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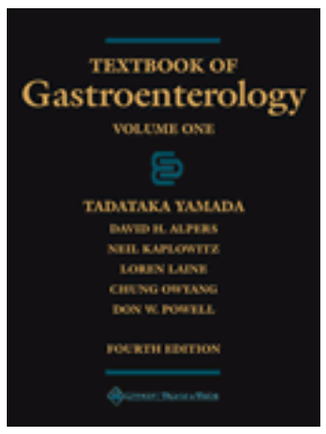

By OkDoKeY

Textbook of Gastroenterology

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PREFACE

In the 16 years or so since we initiated our project to develop a textbook that would serve the needs of students and practitioners of gastroenterology in the 1990s and beyond, the discipline has undergone changes that have exceeded our wildest predictions. The rate of progress in the science of gastroenterology has been so dramatic that the concepts of diagnosis and treatment by which we approached patients with the most common gastrointestinal disorders—such as peptic ulcer disease, cholecystitis, and colon cancer—in the 1980s have been changed fundamentally. *Helicobacter pylori, laparoscopic cholecystectomy,* and *DNA mismatch repair enzyme gene mutations* are terms that were not part of the standard lexicon of even academic gastroenterologists who were already experts in their field. With this challenge before us, we have taken every step to ensure that the fourth edition of the *Textbook of Gastroenterology* is appropriately current. As with the first three editions, every chapter has been updated; in addition, nearly 30 percent of the chapters have new authors to provide a fresh outlook to the material. More importantly, in this edition we have added new chapters to cover the topic of liver diseases, recognizing that most gastroenterologists are also hepatologists. In this regard, we are delighted to introduce our newest associate editor, Neil Kaplowitz, who has assumed responsibility for the hepatology chapters.

These changes have helped retain the major assets of the *Textbook,* namely its modernity, its freshness, and its up-to-date accuracy. Throughout this edition, we have taken great care to maintain the fundamental aims of the *Textbook* in its first and subsequent editions: to present the scientific basis of gastroenterology in such a fashion that it will provide insight into common clinical problems, to detail the basics of a clinician's approach to common clinical problems, to provide at its core an encyclopedic discussion of virtually all of the disease states encountered in practice, and to describe all of the major diagnostic and therapeutic technologies of gastroenterology, both the long standing and the very recent, available to clinicians today.

Above all, as in the first three editions, this edition of the *Textbook of Gastroenterology* is planned to integrate the various demands of science, technology, expanding information, good judgment, and common sense into the diagnosis and management of gastrointestinal patients. Because gastrointestinal complaints are among the most commonly encountered in clinical practice, the *Textbook* is not meant solely for the education of practicing or training gastroenterologists and surgeons but also, and most important in the changing health care environment, primary care physicians and physician extenders. We hope that the collective efforts of the authors of the 162 chapters and the six of us who have edited this edition have succeeded in achieving our goal to provide an outstanding educational work for students and practitioners of the medical science of gastroenterology.

Our efforts were especially facilitated by the expert assistance of Lori Ennis and Barbara Boughen, who collaborated as a team to complement editorial talents with interpersonal skills to maintain the high quality of the text and to deliver the manuscripts in a timely fashion. The authors are indebted to their administrative and secretarial assistants, Carol Arnold, Robbie Loftin, Terri Kirschner, Sue Sparrow, JoAnn Wilson, and Maria Vidrio. In addition, the faculty and fellows of the Gastroenterology Divisions at the University of Michigan, Health System in Ann Arbor, Washington University School of Medicine in St. Louis, and The University of Texas Medical Branch in Galveston provided valuable assistance in reviewing the chapters in the third edition of the *Textbook* as preparations for this, the fourth edition.

The editors wish to express their gratitude to their colleagues at Lippincott Williams & Wilkins who have continued to demonstrate their commitment to quality, integrity, and excellence. Of the many people at Lippincott who have contributed their best efforts to the success of this book, one person deserves special recognition: Beth Barry. This edition would not have been possible without her dedication and extraordinary talent.

Tadataka Yamada, M.D.

PREFACE TO THE FIRST EDITION

The practice of gastroenterology has changed dramatically during the past 20 years. We have witnessed a logarithmic growth in the volume of information concerning the basic biology and biochemistry of the gut. This wealth of new knowledge not only has provided fresh insight into the pathogenesis of gastrointestinal diseases but also has identified the critical role of the gut in the physiology and pathology of other organ systems. There is every reason to expect that the pace of our scientific growth will continue in the years ahead.

In many instances, advances in the science of gastroenterology have been incorporated directly into clinical practice. This has led to the evolution of a large and ever-expanding armamentarium of diagnostic and therapeutic modalities for management of patients with gastrointestinal diseases. The ability to see the organ of pathology and to treat lesions directly without invasive surgical procedures is an advantage almost unique to gastroenterology. As a result, today's clinicians must think in ways not even imagined by their predecessors. Although the textbooks of the past have served us well, modern gastroenterology dictates a more integrated approach to science, technology, and clinical practice. In the *Textbook of Gastroenterology* the Editors address this need.

The *Textbook* begins with a section of chapters describing the basic mechanisms of normal and abnormal gastrointestinal function. This section of the *Textbook* is written so that fundamental scientific concepts can be understood by a reader who is not scientifically oriented. We hope to present the scientific basis of gastroenterology in such a fashion that it will provide insight into common clinical problems. The section is intended to serve both as a guide for clinicians who need to understand the pathophysiology of their patients' disorders and as a resource for serious students of gastroenterology.

A major shortcoming of textbooks that consist solely of descriptions of diseases is the lack of guidance for the reader in applying the information to the management of patients who present with symptoms or signs rather than diagnoses. Therefore, a major section of the *Textbook* consists of detailed chapters on the clinician's approach to patients presenting with common gastrointestinal problems.

As a fully comprehensive textbook, the *Textbook of Gastroenterology* has at its core an encyclopedic discussion of virtually all of the disease states encountered in practice. These chapters, comprising the bulk of the Textbook, have a classical structure that ensures the uniform coverage of all important points.

After the initial evaluation, physicians must choose from a battery of diagnostic and therapeutic modalities as they proceed with patient management. A full section describing all of the major technologies, both the longstanding and the very recent, available to clinicians today comprises the last section of the *Textbook.* This section discusses not only the theory and practical uses of these procedures but also the contraindications and potential complications, the evaluation and assessment of the data obtained, and the future directions of the modality.

The purposes of the *Textbook of Gastroenterology,* then, are multiple: to teach the scientific basis of gastroenterology, to provide practical approaches to common gastrointestinal problems, to serve as an encyclopedic reference for gastrointestinal diseases, and to indicate the current applications and future directions of the technology of gastroenterology. Above all, the *Textbook* is planned to integrate the various demands of science, technology, expanding information, good judgment, and common sense in the diagnosis and management of gastrointestinal patients. The Editors intend the book to be as useful at the bedside as on an office shelf or a student desk. Because gastrointestinal complaints are among the most commonly encountered in clinical practice, we want our readers to include surgeons, primary care physicians, and nurses as well as gastroenterologists and other internists.

To achieve these goals, the finest experts in the field of gastroenterology have written this Textbook. Each chapter is prepared by an authority who is actively engaged in advancing our knowledge of the subject matter of the chapter. We believe that the expertise of this group of physicians and scientists has ensured that the *Textbook* will achieve its aims.

CHAPTER 1

Helen Raybould, Stephen J. Pandol, and Hal Yee

THE INTEGRATED RESPONSES OF THE GASTROINTESTINAL TRACT AND LIVER TO A MEAL

This chapter provides an overview of the mechanisms involved in the regulation of the various responses in the gastrointestinal (GI) tract and the liver to the ingestion of a meal. The coordination of these responses is essential to the overall function of the GI tract, which is to take in nutrients and eliminate wastes, and a key function of the liver, which is to coordinate the distribution and storage of vital nutrients. Each of the processes described in this introductory chapter is explored in greater depth and with extensive references to the literature elsewhere in the textbook.

The GI tract consists of the alimentary canal from the mouth to the anus and the associated glandular organs that empty their contents into the canal. In a general sense, the GI tract adds water, ions, and enzymes to a meal to convert it into an aqueous solution of molecules that can be transported into the blood. Importantly, most of the added substances are absorbed for reuse. The major physiological processes that occur in the GI tract are motility, secretion, digestion, absorption, and elimination. Food is taken into the mouth as large particles containing macromolecules that are not absorbable. The breaking down of food into absorbable material occurs by grinding and mixing the food (motility) with various secretions containing enzymes, ions, and water that enter the GI tract. The enzymes convert the macromolecules into absorbable molecules in a process termed *digestion*. The products of digestion, as well as the secretions from the upper parts of the GI tract, are then transported across the epithelium to enter the blood or lymph by a process termed *absorption*. Secretions and lumenal contents are moved from the mouth to the anus and eliminated by GI motility. The coordination of GI function is regulated in a synchronized way to maximize digestion and absorption by means of multiple control mechanisms.

The liver is anatomically coupled to the GI tract through the biliary tree and portal circulation. It has numerous vital functions, including bile formation and secretion; uptake, storage, and release of nutrients; generation of plasma proteins; detoxification; and immune surveillance. This chapter focuses on the crucial role that the liver plays in the integrated metabolic response to a meal.

The functions of the GI tract and the liver are regulated by three principal types of control mechanisms: paracrine, endocrine, and neural (Fig. 1-1). Paracrine regulation describes the process whereby a chemical released from a sensing cell diffuses through the interstitial space to influence the function of neighboring cells. An example of a paracrine mechanism is the inhibition of gastrin release by somatostatin. Somatostatin is released from cells in the gastric antral mucosa in response to low intragastric pH. Another example is the action of histamine, which is released from enterochromaffin cells in the gastric fundic mucosa, stimulating gastric acid secretion through direct action on the parietal cells.

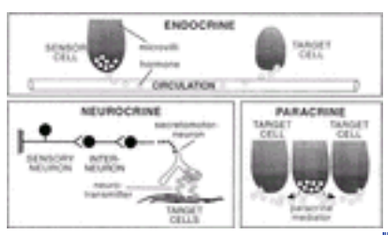

FIGURE 1-1. Three principal types of control mechanisms regulate the function of the gastrointestinal tract—paracrine, endocrine, and neurocrine (neural). Examples of paracrine mechanisms are numerous; in this figure, the target cell is shown as being adjacent to the sensor cell. However, paracrine mechanisms may be mediated over tens or hundreds of microns and may involve many different cell types, including epithelial cells, endocrine cells, nerve terminals, and smooth muscle. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

Endocrine regulation describes the process whereby the sensing cells respond to a stimulus by releasing their contents into the circulation to act on distant target cells. A particular cell responds because it possesses high-affinity receptors specific for the hormone. There are many examples of endocrine regulation in the GI tract and the liver, including the action of gastrin to increase gastric acid secretion, the action of cholecystokinin (CCK) to stimulate pancreatic enzyme secretion, and the action of glucagon to enhance hepatic glycogenolysis and gluconeogenesis.

A neurocrine or neural response is mediated by the release of transmitters from nerves. A neural reflex is the reaction of sensory neurons to stimuli, such as the presence of nutrients or acid in the intestinal lumen, or the stretch and contraction of smooth muscle in the gut wall, leading to the activation of intrinsic and extrinsic neurons that alter the secretory and motor behavior of the gut. An example of a neural reflex is the distention of the esophagus, which leads to the receptive relaxation of the stomach wall. Various sensory neural receptors in the GI tract detect both chemical and mechanical stimuli. The transmission to the target tissue may involve relatively short, neural reflex pathways within the intrinsic nervous system of the gut (i.e., the enteric nervous system) or longer pathways through the spinal cord and brain (Fig. 1-2). Efferent nerves from the brain and spinal cord that innervate the GI tract and the liver are both parasympathetic and sympathetic. In the integrated response of the GI tract to a meal, parasympathetic input is predominant in the regulation of function; these fibers synapse with enteric neurons to effect end-organ responses. Secretomotor enteric neurons are contained within the myenteric and submucosal plexuses—the two nerve plexuses that make up the enteric nervous system. These plexuses contain the neural elements capable of integrating entire reflex circuits without input from the central nervous system (CNS). In contrast to the GI tract, the autonomic nervous system plays a relatively modest role in the hepatic response to a meal, as affirmed by the success of liver transplantation, in which the liver is disassociated from the nervous system.

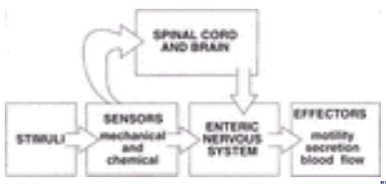

FIGURE 1-2. The control of gastrointestinal (GI) function during the response to a meal is regulated by neural reflexes. The various stimuli present in the meal or generated within the GI tract by digestion stimulate many different sensors. Activation of these sensors stimulates intrinsic and extrinsic afferent neurons, resulting in reflex activity mediated through the enteric nervous system, the prevertebral ganglia, or the central nervous system (spinal cord or brainstem). Activation of these reflexes alters effector function in the GI tract, including motility, secretion, and blood flow. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

The response of the GI tract to a meal is classically divided into phases: cephalic, oral, esophageal, gastric, duodenal, intestinal, and colonic. The liver's response to a meal permits a shift in metabolic conditions from the fasted state to the fed state. In each phase or state, the meal provides certain stimuli that activate control mechanisms resulting in changes in effector function, such as secretion, motility, and metabolism. A particular stimulus generated in an organ of the GI tract can activate effectors not only in the same organ but also in other regions of the GI tract or the liver. These coordinated effects allow for continuous adjustments in motility, secretions, or metabolism to match the unique characteristics of a meal to maximize its digestion, absorption, distribution, and storage.

GASTROINTESTINAL RESPONSE TO A MEAL

Cephalic and Oral Phases

The main feature of the cephalic and oral phases of the response to a meal is the activation of the GI tract. The cephalic phase consists of responses to the auditory, cognitive, visual, and olfactory stimuli induced by the meal. The oral phase includes responses to many of the same stimuli but also includes those initiated in the mouth, which are both mechanical and chemical. The effector responses are mediated through various higher brain centers but ultimately converge in the parasympathetic outflow to the GI tract through the cranial nerves and the vagus nerve (Fig. 1-3). This outflow activates both secretory and motor events, including salivary, gastric, and pancreatic secretions; gallbladder contraction; and relaxation of the sphincter of Oddi. These responses supply the intralumenal environment with water, ions, digestive enzymes, and bile, which are all necessary to initiate digestion and solubilize components of the meal in the aqueous phase so that complete digestion and absorption can occur.

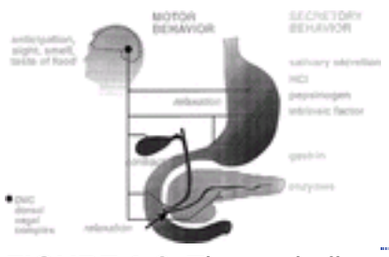

FIGURE 1-3. The cephalic and oral phases of the meal initiate a similar repertoire of responses to activate the digestive processes in the gastrointestinal (GI) tract. The stimuli from either cognitive cues or oropharyngeal stimulation result in activation of parasympathetic outflow, which activates secretory and motor events in both the proximal and distal GI tract. *HCl,* hydrochloric acid. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

During the oral phase, mechanical disruption of the food (i.e., chewing) and the addition of salivary secretions enable swallowing and initiate digestion (Table 1-1). Chewing subdivides the food and mixes it with salivary secretions, including amylase, lingual lipase, water, and mucus. Chewing also results in the formation of a rounded, smooth, and lubricated portion of food (i.e., a bolus) that can be swallowed. Mucus lubricates the food for both chewing and, more importantly, swallowing. The presence of material in the mouth provides both mechanical and chemical stimuli that initiate salivary, gastric, and pancreatic secretion through activation of parasympathetic pathways. Studies in humans indicate that the gastric secretory response to oropharyngeal stimulation is far more robust if the meal is palatable, although the action of chewing bland material will initiate both salivary and gastric secretion. These results indicate that both mechanical and chemical stimuli induced by the meal can mediate the secretory response.

Disruption of food resulting in smaller particles
Formation of bolus for swallowing
Initiation of starch and lipid digestion
Facilitation of taste
Cleaning of mouth and provision of antibacterial action
Clearance and neutralization of refluxed gastric material in the esophagus
Regulation of gastric and duodenal phase responses
Regulation of food intake and eating behavior
Assistance in speech

TABLE 1-1 Functions of Chewing and Salivary Secretion

The components of the cephalic and oral phases of the response to a meal are mediated by purely neural pathways. For example, the salivary secretory response is mediated by parasympathetic effector neurons. In the stomach, responses vary by region. In the proximal stomach, vagal activation stimulates pepsinogen secretion from chief cells, and proton and intrinsic factor secretion from parietal cells. In the distal stomach, vagal activation induces the release of gastrin from endocrine cells in the antrum (i.e., G cells). For this response, vagal efferents synapse with enteric neurons that release gastrin-releasing peptide, which in turn directly activates G-cell gastrin release. Gastrin acts as an endocrine agent on chief and parietal cells to promote their respective secretions. The parietal cell is also activated by histamine that is released from mucosal enterochromaffin-like (ECL) cells. Histamine release is regulated by cholinergic vagal inputs and by gastrin. The acid secreted into the stomach provides the appropriate conditions for the rapid activation of pepsin from its inactive precursor. Thus, protein digestion can be initiated rapidly as food enters the stomach. The overall effect is a complex interaction of neural, paracrine, and endocrine controls initiated by neural activation in the cephalic and oral phases of the responses to a meal.

In a similar manner, the motor effects on the gallbladder and the sphincter of Oddi are mediated by vagal efferent action on the enteric neurons in these organs. Gallbladder contraction is mediated at least in part by the direct action of acetylcholine, released from intrinsic neurons, on smooth muscle. The inhibitory action on the sphincter is mediated by vasoactive intestinal peptide (VIP) and nitric oxide (NO), which are also released from intrinsic neurons.

The responses of the exocrine secretory cells and smooth muscle previously described, as well as the responses in the pancreas and the intestine, result from agonist interaction with specific receptors on the cells, followed by activation of intracellular signaling systems. These signals, in turn, regulate ion transporters, exocytosis, and the contraction and relaxation of smooth muscles. The specifics of these intracellular signals and cellular responses are detailed elsewhere in this textbook.

Esophageal Phase

The pharynx, esophagus, and associated structures have two main functions: to transfer food from the mouth to the stomach, and to protect the gut from the potentially injurious effects of acid and digestive enzymes (Fig. 1-4).

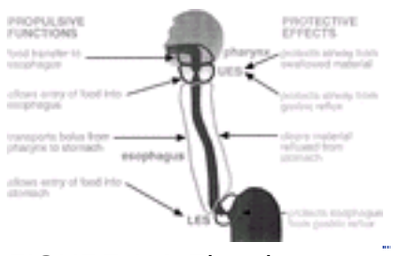

FIGURE 1-4. The pharynx, esophagus, and associated structures (*UES,* upper esophageal sphincter; *LES,* lower esophageal sphincter) subserve both transfer (i.e., propulsive) and protective functions. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

The stimuli provided by the meal in the esophageal phase are predominantly mechanical. Swallowing produces a mechanical stimulation of the pharynx, and the entry of the bolus into the esophagus produces esophageal distention. The sensors involved are mechanosensitive afferents innervating both the pharynx and the esophagus. Activation of these afferents initiates reflex pathways by way of the brainstem or solely by way of intrinsic pathways within the enteric nervous system. These pathways initiate a stereotypic response of muscle relaxation and contraction that is called *peristalsis*. In the pharynx and upper one third of the esophagus (striated muscle), motility is regulated from the nucleus ambiguus in the brainstem. Parasympathetic outflow through the vagus nerve terminating on intrinsic neurons regulates contraction and relaxation of the smooth muscle in the lower two thirds of the esophagus. However, the stimulation of the enteric nervous system of the esophagus is sufficient to produce peristalsis. Pharyngeal and esophageal stimulation and activation of reflex pathways (both intrinsic and extrinsic) produce striated and smooth muscle peristalsis, relaxation of the upper esophageal sphincter (UES) and the lower esophageal sphincter (LES), and relaxation of the proximal stomach (i.e., receptive relaxation).

Another important reflex is a protective mechanism mediated by spinal afferents that terminate in the distal esophageal mucosa. These afferent terminals are sensitive to acid, and their activation leads to reflex closure of the LES and relaxation of the proximal stomach.

The pharynx, UES, esophagus, and LES act in a coordinated manner to propel material from the pharynx to the stomach. The movement of the bolus of food in the mouth, aided by the tongue, initiates a peristaltic wave that carries the bolus through the pharynx. With the coordinated relaxation of the UES, the bolus enters the esophagus, wherein peristaltic waves move it toward the stomach. With each swallow (i.e., pharyngeal stimulation), the LES and the proximal stomach relax, allowing the bolus to pass into the stomach. Distention of the esophagus by the bolus initiates another wave called *secondary peristalsis*. Repetitive secondary peristalsis is often required to clear the esophagus of the meal bolus. Like pharyngeal stimulation, esophageal distention produces relaxation of the proximal stomach and opening of the LES. This allows the stomach to accommodate large volumes of the meal with a minimal increase in intragastric pressure.

Gastric Phase

The gastric phase initiates several responses within the stomach and the other organs of the GI tract, including pancreatic secretion, gallbladder contraction, and relaxation of the sphincter of Oddi (Table 1-2). Stimuli provided by the presence of a meal in the stomach are both mechanical (i.e., distention and stretch of smooth muscle wall) and chemical (e.g., oligopeptides and amino acids produced by protein digestion in the lumen). The pathways mediating the effects of these stimuli are complex, involving neural, endocrine, and paracrine mechanisms. The gastric responses are both secretory (e.g., acid, intrinsic factor, mucus, pepsinogen, gastrin, lipase, bicarbonate) and motor (e.g., inhibition of proximal stomach motility and stimulation of antral peristalsis) (Fig. 1-5). Afferent (i.e., sensory) neurons respond to mechanical and chemical stimuli and activate both intrinsic pathways and parasympathetic outflow via vagovagal reflex pathways. Parasympathetic efferent neurons can be either excitatory or inhibitory, depending on the intrinsic secretomotor neurons on which they terminate. Those that terminate onto neurons that release either acetylcholine or other excitatory neurotransmitters (e.g., substance P) stimulate secretion and motility, and those that terminate on neurons that release VIP or NO generally inhibit function. These neural pathways mediate gastric motor and secretory responses, as well as responses further along the GI tract. The endocrine pathway consists of the release of gastrin, leading to the stimulation of gastric secretion. Paracrine pathways include the release of somatostatin, leading to the inhibition of gastric secretion, and the release of histamine, leading to the stimulation of gastric secretion. These endocrine and paracrine pathways activate responses only within the stomach.

Gastric Responses
Storage of the meal
Secretion of pepsinogen and lipase to initiate digestion
Secretion of H^+ to kill microorganisms and convert pepsinogen to active form
Secretion of intrinsic factor to bind vitamin B_{12} (cobalamin) for absorption
Secretion of mucus and bicarbonate for gastric mucosal barrier and lubrication
Secretion of water for aqueous suspension of nutrients and to make hypoosmotic
Mixing secretions with food and reduction of particle size (grinding) of the meal
Regulation of emptying of contents into duodenum
Distal Gastrointestinal Tract Responses
Stimulation of pancreatic secretion
Contraction of the gallbladder
Increased colonic motor activity
Relaxation of the sphincter of Oddi

TABLE 1-2 Major Functions of the Gastric Phase

FIGURE 1-5. The gastric phase. Once food reaches the stomach, the meal presents many mechanical and chemical stimuli to the gastric wall that initiate new responses or reinforce those that are ongoing. These stimuli initiate both neural and endocrine (humoral) pathways to bring about changes in function. The different regions of the stomach subserve different functions; the proximal stomach (fundus and body) secretes many factors and has a major role as a reservoir. The distal stomach (antrum and pylorus) secretes mucus and bicarbonate; motor coordination of this region is critical in the regulation of gastric emptying. *LES,* lower esophageal sphincter. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

The afferent innervation of the stomach consists of vagal and spinal afferents, the cell bodies of which are in the nodose and dorsal root ganglia, respectively. The central terminals lie in the brainstem and spinal cord, and the peripheral terminals lie in the muscle and mucosa. The sensory terminals in the muscle respond to stretch of the stomach wall, and afferents innervating the mucosa respond to lumenal contents. The terminals in smooth muscle respond to both stretch and active contraction in the terminal fields. They are predominantly vagal afferents; the majority of spinal afferents terminate in the serosa and mesenteric attachments. The mechanism by which mucosal afferents respond to changes in lumenal content is not clear, but it is characteristic of afferents along the length of the GI tract and may be similar to the process that mediates taste. Endocrine or EC cells may release their contents in response to apical stimulation by nutrients, acid, or osmotic stimuli. The release of contents from the basolateral portion of the cells, which are close to the terminals of afferents, stimulates the afferents to initiate neural reflexes.

As the meal enters the stomach, the stretch of the gastric wall activates vagal afferents, which results in vagovagally mediated receptive relaxation of the proximal stomach and an increase in gastric acid secretion, as well as the induction of pancreatic enzyme secretion, gallbladder contraction, and sphincter of Oddi relaxation. Parasympathetic innervation through the vagus nerve is the strongest stimulant of gastric acid secretion. Extrinsic efferent fibers terminate on intrinsic neurons that innervate not only parietal cells, but also ECL cells that release histamine and G cells that release gastrin. Acetylcholine induces acid secretion directly by acting on

parietal cells, and indirectly by stimulating ECL cells and activating neurons that release gastrin-releasing peptide, which, in turn, induces gastrin release from G cells. The complexity of the regulatory systems is further illustrated by the observation that gastrin also can act directly on parietal cells, or indirectly by stimulating ECL cells to release histamine (Fig. 1-6). If the vagus nerve is severed, acid secretion in response to gastric wall stretch is reduced but not abolished. Such results suggest that intrinsic neural reflexes also can mediate the secretory response. Thus, the same stimulus can activate the response through different effector mechanisms. In addition to gastric distention activating this reflex pathway, the products of protein digestion also can activate the same effector pathway, presumably by stimulating the sensory terminals in the mucosa. Furthermore, the products of digestion can activate acid secretion independently of neural pathways by directly stimulating gastrin release from G cells in the antral mucosa.

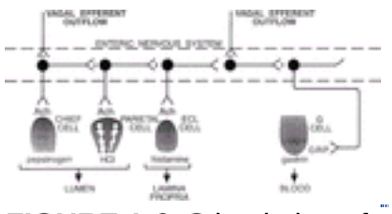

FIGURE 1-6. Stimulation of gastric acid secretion by increased parasympathetic utflow is effectively amplified by the intrinsic neuronal circuitry of the gastric wall. Vagal efferents synapse with intrinsic cholinergic neurons and with neurons containing gastrin-releasing peptide (*GRP*). Activation of these intrinsic neurons stimulates a number of cell types in the gastric mucosa. Neurocrine (GRP and acetylcholine [*Ach*]), paracrine (histamine), and endocrine (gastrin) pathways all contribute to this response. *ECL,* enterochromaffin-like cell; *G cell,* gastrin cell; *HCl,* hydrochloric acid. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

A negative feedback mechanism also exists, whereby a low pH in the gastric antral lumen stimulates the secretion of somatostatin from endocrine cells. Somatostatin exerts a paracrine effect on G cells, decreasing the release of gastrin, which indirectly leads to a decrease in acid secretion.

The motor functions of the stomach include acting as a reservoir for ingested food, fragmenting the food and mixing it with secretions, and emptying of the contents into the duodenum at controlled rates. In a consideration of the motor functions of the stomach, it is easiest to divide the stomach into proximal and distal regions. The proximal stomach produces slow changes in tone compatible with its reservoir function. During the first part of the meal, the proximal stomach relaxes to accommodate the volume of the meal, but later in the response to the meal, tone in the proximal stomach starts to increase, which accelerates gastric emptying by propelling the contents toward the distal stomach. Phasic contractions in the distal stomach are generated by myoelectric activity at a rate of three cycles per minute beginning in the midstomach and moving toward the pylorus. When the nutrient meal is in the stomach, antral contractions serve to mix and grind the gastric contents, resulting in a reduction in the size of solid particles. These contractions also contribute to the emptying phase of the response to a meal. As material is emptied from the stomach, antral contractions are associated with pyloric opening, and gastric chyme is emptied into the duodenum. Only particles less than 2 mm in diameter pass into the duodenum; larger particles are propelled back into the proximal stomach. Gastric motility in the gastric phase of the meal is controlled primarily by neural reflexes mediated by extrinsic circuits but also by the enteric nervous system. These neural reflexes are activated by receptors for chemical and mechanical stimuli in the duodenum, principally nutrients, osmotic load, acid, and stretch of the intestinal wall.

The gastric phase also consists of the activation of the more distal GI tract, including pancreatic secretion, gallbladder contraction, and relaxation of the sphincter of Oddi. These effects are mediated by the same vagal afferent and efferent pathways that mediate the gastric responses, and they prepare the intestine for the gastric emptying of the meal into the proximal small intestine.

Duodenal Phase

The main characteristics of the duodenal phase are the controlled delivery of chyme from the stomach, and the secretion of pancreatic juice and bile into the duodenum to match the digestive and absorptive capacity of the intestine (Table 1-3). The function of this region is highly regulated by feedback mechanisms (Fig. 1-7). The combination of organs regulated during this phase has been termed the *duodenal cluster unit:* stomach, duodenum, liver, biliary tract, gallbladder, and pancreas. Lumenal and wall stimuli activate neural and endocrine pathways to mediate the responses of these organs. The different organ systems in the duodenal cluster unit have a common embryologic origin and are regulated by similar sensory mechanisms and effector pathways. The gastric phase of the response to a meal provides some of the stimuli for the duodenal phase. For example, the products of gastric lipid and protein digestion are potent stimulants of regulation in the duodenum. The important duodenal stimuli are distention, acid, osmotic load, and different nutrients. The sensory pathways consist of spinal and vagal afferents and intrinsic sensory neurons that initiate reflex pathways. Hormonal pathways, especially those mediated by CCK and secretin, are also important. The effector systems regulated include the inhibition of gastric emptying and secretion, the stimulation of pancreatic secretion, gallbladder contraction, relaxation of the sphincter of Oddi, pancreatic and biliary water and bicarbonate secretion, intestinal secretion, and the conversion of small bowel motility from the fasted to fed patterns.

Inhibition of gastric acid secretion
Inhibition of gastric emptying
Stimulation of pancreatic enzyme secretion
Stimulation of pancreatic and biliary ductal ion and water secretion
Stimulation of gallbladder contraction
Relaxation of the sphincter of Oddi
Alteration of intestinal motility from the fasted to the fed state

TABLE 1-3 Major Functions of the Duodenal Phase

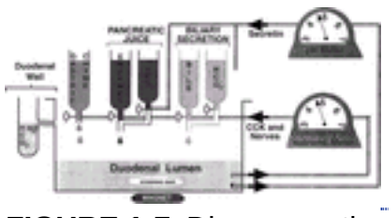

FIGURE 1-7. Diagrammatic representation of the duodenal cluster unit illustrates the control mechanisms and pathways involved in intestinal feedback regulation during the duodenal phase. The nutrient content and osmolality of the meal are sensed (represented as a *meter*) and activate predominantly neural pathways involving cholecystokinin (*CCK*) to open or close various stopcocks to increase pancreatic secretion or delay gastric emptying, for example. The acidity of the meal is sensed and activates secretin release and neural pathways to increase bicarbonate secretion from the pancreas and probably also from the duodenal wall. The contents of the duodenum are mixed with the secretions by the segmental contractions of the smooth muscle wall, represented as a *stirring bar* and *magnetic stirrer*. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

Gastric emptying in the duodenal phase depends on the chemical and physical composition of the gastric contents (i.e., chyme) entering the duodenum. Sensory neurons are both vagal and spinal, responding to nutrients, H^+, distention, and the hyperosmolality of chyme. The effector responses that result in the inhibition or slowing of gastric emptying are not completely understood but involve the activation of vagal efferent outflow, which produces a decrease in antral contractions, contraction of the pylorus, and a decrease in proximal gastric tone. In addition to these extrinsic neural pathways, there also may be local pathways, such as an intrinsic neural reflex, mediating the pyloric contraction induced by acid in the duodenum.

The availability of CCK antagonists has contributed to the understanding that CCK is physiologically important in regulating the function of the duodenal cluster unit in the duodenal phase. The administration of CCK-A receptor antagonists reverses the inhibition of gastric emptying, gallbladder contraction, relaxation of the sphincter

of Oddi, and stimulation of pancreatic enzyme secretion in response to either a meal or the infusion of nutrients into the duodenum. CCK is released from endocrine cells in the proximal intestine in response to lumenal lipid and protein. The action of CCK to inhibit gastric acid secretion and gastric emptying, and to increase pancreatic secretion, is dependent on neural pathways (Fig. 1-8). CCK stimulates specific CCK-A receptors on vagal afferent nerve terminals in the intestinal mucosa and activates vagovagal reflexes to modify gastric and pancreatic function. Thus, CCK, long thought of as a classic hormone, may actually function as a locally acting paracrine effector acting on nerve terminals to initiate reflex neural responses.

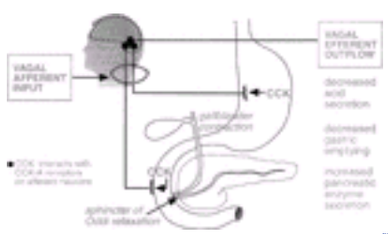

FIGURE 1-8. Cholecystokinin (*CCK*) acts predominantly by way of a vagal afferent pathway to integrate intestinal feedback regulation of the duodenal phase of the meal. (From Undergraduate Teaching Program of the American Gastroenterological Association, 1996.)

The physiological release of CCK in response to a meal is modulated by at least two peptides. One is monitor peptide, which is secreted by pancreatic acinar cells. In addition, there is at least one lumenal CCK-releasing factor. The proposed mechanism of action for both of these releasing factors is that they are constitutively secreted into the intestinal lumen. In the absence of protein or protein products in the intestinal lumen, both factors are efficiently broken down by digestive enzymes and thus are not available to stimulate CCK release. On ingestion of a meal, however, dietary proteins compete with the releasing factors as substrates for the digestive enzymes. The net effect is that the releasing factors escape digestion and become available to stimulate CCK secretion from endocrine cells in the mucosa.

The most important function of the pancreas in the integrated response to a meal is the production, storage, and secretion of digestive enzymes. The major control of pancreatic enzyme secretion is exerted by efferent fibers from the parasympathetic nervous system and by CCK. As indicated in the previous sections, parasympathetic activation of pancreatic enzyme secretion occurs during the cephalic, oral, gastric, and duodenal phases of the response. Secretion during the duodenal phase accounts for about 70% of the total response to a meal. In this phase, secretion is activated by the presence of chyme in the duodenum by way of extrinsic neural pathways, enteropancreatic neural pathways (i.e., through intrinsic neurons that terminate in both the duodenum and pancreas), and endocrine pathways (i.e., through CCK, secretin).

As the meal enters the intestine, it is acidic and hyperosmotic; however, by the time it leaves the duodenum, it has a neutral pH and is isosmotic. This conversion is achieved by the secretion of large volumes of water and bicarbonate ions by the intestinal mucosa, and by the pancreatic and biliary systems. Secretion is regulated mainly by the release from the duodenal mucosa of secretin, which acts as an endocrine agent to increase bicarbonate and water secretion from the pancreatic and biliary ductal systems. The existence of a releasing peptide for proton-induced release of secretin also has been proposed.

The contraction of the gallbladder and the relaxation of the sphincter of Oddi result in the addition of biliary secretion to the meal. During the cephalic, oral, and gastric phases, these responses are mediated by parasympathetic efferent nerves. During the duodenal phase, CCK acting either alone or through a vagal reflex pathway has a pronounced effect on the delivery of bile to the duodenum. Bile is composed of both inorganic and organic constituents. As previously indicated, the inorganic constituents (e.g., water, bicarbonate ions, and other electrolytes) serve to convert gastric chyme into a neutral isosmotic solution. The major organic constituents of bile are conjugated bile acids, phospholipids, cholesterol, and bilirubin; the latter two are excretory products. Both bile acids and phospholipids are essential in maintaining cholesterol in a soluble state and thereby preventing the formation of gallstones. Conjugated bile acids traverse the entire small intestine and are taken up by a receptor-mediated transport system in the terminal ileum. The conjugated bile acids return to the liver by way of the portal circulation and then are secreted back into the biliary system and stored in the gallbladder. This cycling of bile acids is termed the *enterohepatic circulation*. Bile acids have multiple important functions in the process of digestion and absorption of lipids and fat-soluble vitamins. They accomplish these functions in part by possessing polar (hydrophilic) and nonpolar (hydrophobic) regions that allow them to interface between aqueous and lipid environments. Thus, they are involved in the formation of emulsions and micelles and in the binding of lipolytic digestive products to the oil phase of the meal. All these effects enhance digestion and the absorption of fat in the meal.

The highly regulated duodenal phase is responsible for the addition of water, ions, enzymes, bile acids, and other secretions to the meal in proportions that result in an optimal environment for the digestion of nutrients. The process of digestion in the intestinal lumen is facilitated further by the induction of patterns of motility in the duodenum that promote the mixing of intestinal contents with digestive enzymes.

During the intervals between meals, motility of the GI tract is characterized by periods of intense contractions and periods of quiescence. In humans, these periods cycle at about 1.5 hours. The sequential contractions migrate aborally (i.e., toward the anus) and have been termed the *migrating motor complex* (MMC). This complex of migrating contractions starts in the stomach and moves through the intestine and into the colon, sweeping undigested material and contents through the GI tract in the interdigestive period. Initiation of the MMC is dependent on the integrity of the vagal innervation, which serves to release the hormone motilin, which, in turn, activates the MMC. Food in the intestine abolishes the MMC and changes intestinal motility from this fasted pattern to a fed pattern of motility.

Small Intestinal Phase

This phase of the meal is characterized by digestion and absorption. The small intestine is divided into three functional regions: duodenum, jejunum, and ileum. The jejunum is the major organ for intralumenal and surface digestion and absorption of meal nutrients. In addition, a significant portion of the water and ions from both exogenous and endogenous sources is absorbed in the jejunum. The ileum provides another site for absorption. The ileum has a specific role in the absorption of cobalamin (vitamin B_{12}) bound to intrinsic factor secreted by the stomach and bile acids. The ileum also releases hormones that have important functions in the gut. For example, the peptide YY (PYY) inhibits gastric function. The ileocecal valve prevents the large reservoir of colonic bacteria from entering the small intestine. Two characteristics of the small intestine make it especially well adapted for its critical role in nutrient assimilation. First, the small intestine has an enormous surface area as a result of folds in the mucosa, villi on the mucosa, and microvilli on the epithelial cells. Second, the blood flow to the intestine is very high after a meal; it can account for up to 25% of the cardiac output. The enteric nervous system is highly developed in the small intestine, enabling this region of the GI tract to function fairly independently of the extrinsic innervation.

The main responses initiated in the small intestinal phase are alterations in intestinal motility patterns, intestinal secretion, and blood flow, as well as regulation of tight junction permeability. The small intestine exhibits two motor patterns: peristalsis and segmentation. Both of these motility patterns are generated by the enteric nervous system and are independent of the extrinsic nervous system. Peristalsis is characterized by a wave of relaxation followed by a ring of contraction that develops at a point and moves aborally over variable distances. Peristalsis is the primary mechanism by which contents are moved along the intestine in the digestive period. Segmentation is characteristic of the fed state and is the process by which rings of contraction develop at uniform intervals, dividing the lumen into segments. Segmentation is the primary mechanism by which the contents of the intestine are mixed with secretions and moved across the mucosa to enhance absorption.

Other responses initiated in the small intestine are less well understood: for example, regulation of mucosal blood flow, regulation of tight junction permeability, and alteration in mucosal function.

The major changes in the chemical and physical characteristics of food, as well as the absorption of nutrients, trace elements, vitamins, water, and ions, occur in the intestinal phase. Hydrolysis of proteins and carbohydrates and the solubilization and hydrolysis of fats are catalyzed by enzymes in the intestinal lumen (from salivary, gastric, and pancreatic secretions) and in the intestinal brush border. Absorption is the process by which molecules produced by lumenal surface digestion are transported into epithelial cells and then carried into the blood or lymph. In addition to absorbing components of the meal, the small intestine absorbs much of the endogenous secretions. The duodenum and jejunum have the highest absorptive capacities; most absorption occurs in the upper small intestine, although some occurs in the ileum. Molecules are absorbed by two principal mechanisms: transcellular (i.e., passing through enterocytes) and paracellular (i.e., passing between enterocytes). Monosaccharides, amino acids, electrolytes, minerals, and water-soluble vitamins enter the portal circulation and pass through the liver into the systemic circulation. Lipids and fat-soluble vitamins and cholesterol esters (together with lipophilic drugs) enter the lymph, which drains into the systemic circulation.

Colonic Phase

The colonic phase of the response is important in the further reabsorption of water and ions and for the storage and elimination of waste products. The cecum and ascending colon receive about 2 L of ileal effluent daily. Absorptive transport mechanisms reduce the volume to about 200 mL per day. The transverse, descending, and sigmoid portions of the colon store fecal material and transport the material to the rectum. The rectum signals the defecation reflex. In addition, some fermentation of monosaccharides yields free fatty acids that can be taken up by colonic epithelial cells.

The stimuli induced in the colon are both mechanical (e.g., rectal distention) and chemical (e.g., free fatty acids). Extrinsic and intrinsic neural pathways mediate the motility responses of the colon and the defecation response. Hormonal pathways affecting other parts of the GI tract also may be excited in the colon. For example, PYY may be released from the colonic mucosa if the fat content of the meal is high and not all is absorbed by passage through the small intestine. PYY, in turn, slows gastric emptying and transit of the meal through the small intestine, resulting in increased fat digestion and absorption.

The effectors in the colonic phase are both motor and secretory. The colon exhibits both storage motor patterns (i.e., changes in tone) and propulsive motor patterns (i.e., phasic contractions). Rectal distention initiates the defecation reflex. This response consists of both involuntary and voluntary components. Propulsive motility in the descending colon and rectum is increased to move the feces to the anus, where relaxation of internal and external anal sphincters facilitates elimination.

HEPATIC RESPONSE TO A MEAL

An essential function of the liver is to coordinate the distribution of vital metabolic substrates to the tissues of the body. It must perform this task during both the fasted (i.e., postabsorptive) state, when the gut contains no nutrients, and the fed (i.e., absorptive) state, when nutrients ingested during a meal enter the circulation from the gut (Table 1-4). During the fasted state, the energy needs of the tissues must be satisfied by metabolic substrates generated from the body's stores, chiefly the liver, the adipose tissue, and the skeletal muscle. Conversely, during the fed state, the body's energy requirements are met by a portion of the ingested nutrients, and any remaining nutrients must be stored for later use. The integrated response of the liver to a meal directs the crucial metabolic transition from the fasted to the fed state and back.

FASTED STATE	FED STATE
No nutrients in the gut	Nutrient absorption from the gut
Energy needs met by using stored fuel	Energy needs met by using ingested nutrients
Net hepatic glucose release	Glucose is principal fuel source
Glycogenolysis by liver and muscle	Hepatic glucose uptake
Gluconeogenesis by liver	Glycogen synthesis by liver and muscle
Fat catabolism by adipocytes	Fat synthesis and storage by liver and adipocytes
Protein catabolism, especially by muscle	Repletion of depleted proteins
Enhanced fatty acid utilization by most tissues	
Long-term Fast	
Hepatic ketone generation	
Enhanced ketone use by brain and other tissues	
Proportional increase in renal gluconeogenesis	

TABLE 1-4 Major Characteristics of the Fasted and Fed States

Fasted State

During the fasted state, the liver must maintain the concentration of glucose in the blood even though no nutrients are being absorbed from the gut. This is imperative because, under ordinary circumstances, the brain and erythrocytes use only glucose as an energy source. The blood glucose concentration is sustained by hepatic glucose production and by the reduction of glucose utilization by most tissues and organs. An early event in the fasted state is the hydrolysis of glycogen (i.e., glycogenolysis) in the liver into glucose that is released into the blood. The hepatic stores of glycogen are, however, limited, so that during prolonged fasts the liver must synthesize glucose from a variety of precursors (i.e., gluconeogenesis), which include lactate, pyruvate, amino acids, and glycerol. These glucose precursors result from several extrahepatic metabolic processes that are stimulated during a prolonged fast. Glycogen is stored in skeletal muscle as well as in the liver. During a prolonged fast or when a muscle is working, skeletal muscle glycogen is hydrolyzed to form lactate and pyruvate. Lipid hydrolysis (i.e., lipolysis) in adipocytes is stimulated in the fasted state, resulting in the formation of glycerol as well as fatty acids. As the fasted state proceeds, protein mainly located in skeletal muscle is catabolized to amino acids. These precursors are circulated to the liver, where they are converted into glucose to help preserve the concentration of glucose in the blood. Consequently, during the fasted state, the liver is a site of net glucose release.

Gluconeogenesis is capable of supplying less than half of the energy necessary for survival. Therefore, the fasted state is also characterized by a substantial reduction in glucose utilization by most tissues and organs of the body. Glucose generated by the liver is, thus, spared for use by the CNS. Glucose use is reduced by an increase in fat utilization by other tissues. As previously discussed, enhanced lipolysis associated with the fasted state produces fatty acids in addition to the glucose precursor, glycerol. These fatty acids are released to the systemic circulation, where they are used as an energy source by nearly all tissues, except, most notably, the CNS. Fatty acids undergo ß-oxidation, producing hydrogen atoms, which undergo oxidative phosphorylation, and acetyl-CoA, which enters the Krebs cycle, where it is catabolized to generate energy.

During long-term fasts (i.e., longer than 24 to 48 hours), further metabolic changes occur that facilitate survival. The kidneys, which are also capable of performing gluconeogenesis, take on an increasingly more prominent role in glucose production. In addition, the liver converts acetyl-CoA, a product of fatty acid catabolism, as previously discussed, into ketones. These ketones are used as an energy source by many tissues, including the brain. A distinct advantage of shifting the fuel source from glucose to ketones is that muscle is spared as a source of amino acids for gluconeogenesis.

Fed State

The digestion and absorption of a meal drastically alter the metabolic conditions in the body because extrinsic nutrients enter the blood from the gut. During this period, termed the *fed state,* ingested glucose becomes the body's principal energy source. A small amount of this glucose enters the liver, where some of it is used to meet the liver's energy needs; the remainder is converted to glycogen, which will replete the hepatic stores, and to fatty acids and glycerol for synthesis of and storage as triacylglycerols in the adipose tissue. Thus, a key feature of the fed state is net glucose uptake by the liver. Glucose that reaches the skeletal muscle is catabolized for energy and converted to glycogen to replete muscle energy stores. Adipocytes catabolize glucose as well, but they also use it to form a-glycerol phosphate and fatty acids, which are triacylglycerol precursors. Ingested amino acids are taken up by all the body's tissues and organs, including the liver, for use in protein synthesis. Excess amino acids, however, are not stored as protein, but are converted by the liver to a-keto acids, which are catabolized or converted to fatty acids for storage as fat. Thus, the net protein synthesis during the fed state simply replenishes the protein degraded during the fasted state, except in growing children or vigorously exercising adults. The lipid contents of a meal are absorbed by the intestine as triacylglycerol-containing chylomicrons that are carried to the circulation by way of the lymphatics. Lipoprotein lipases in the endothelium hydrolyze the ingested triacylglycerols into fatty acids and glycerol, which the tissues use for energy, and adipocytes store the excess as fat.

Regulation of the Metabolic State

The hepatic response to a meal, which directs the transition from the fasted to the fed state, is regulated by endocrine and neural pathways, as is that of the GI tract, but also by the concentration of metabolic substrates in the sinusoidal blood. Insulin and glucagon, hormones produced in the islet cells of the pancreas, are the principal regulators of the metabolic state, such that the transition from the fasted to the fed state can be explained by an increase in the ratio of insulin to glucagon in the blood. Insulin, the secretion of which is increased during the fed state, affects primarily the metabolism of the liver, adipose tissue, and muscle. Insulin stimulates glucose uptake and use, net glycogen synthesis, net amino acid uptake, and net protein synthesis by muscle. The effect of insulin on adipocytes is to increase glucose uptake and use, and net fat synthesis. The hepatic response to insulin is enhanced glucose uptake, net glycogen synthesis, and net fat synthesis.

Hyperglycemia is the most important stimulus for insulin secretion, although an increase in the blood amino acid concentration also stimulates insulin release. Therefore, the absorption of metabolic substrates during a meal represents a powerful regulator of the insulin concentration in the blood. In addition, insulin secretion is inhibited by sympathetic neural activity and epinephrine, and enhanced by intestinal hormones, including glucose-dependent insulinotropic peptide.

In contrast to insulin secretion, glucagon secretion is increased during the fasted state. Glucagon, which acts primarily on the liver, counters the effects of insulin by stimulating glycogenolysis, gluconeogenesis, and ketone synthesis. Glucagon release is stimulated mainly by hypoglycemia and inhibited by hyperglycemia, although epinephrine and sympathetic neural activity also stimulate its secretion.

Aside from regulating the secretion of insulin and glucagon, epinephrine and the sympathetic nervous system stimulate glycogenolysis by the liver and muscle, gluconeogenesis by the liver, and lipolysis by adipocytes. Hypoglycemia leads to reflex epinephrine secretion and sympathetic neural activity. Although other hormones, such as cortisol and growth hormone, modulate hepatic metabolism, they do not play an important role in the liver's response to a meal.

In addition to the roles that blood glucose and amino acid concentrations play in the hormonal regulation of the metabolic state, the concentration of glucose in the blood of the sinusoids directly modulates hepatic metabolism. As a result of the activity of certain glucose concentration–dependent enzymes in the liver, elevation of the sinusoidal blood glucose concentration stimulates glycogen synthesis and inhibits glycogenolysis. Together, hormones, the autonomic nervous system, and the concentration of metabolic substrates in the blood precisely modulate the hepatic response to a meal.

SUMMARY

Of necessity, this overview of the function of the GI tract and the liver in response to a meal is brief. The details of the function of each organ are provided elsewhere in this textbook. The overview provides the reader with a framework for understanding the regulatory mechanisms and the interdependence of organ functions and control mechanisms required for optimal functioning of the entire process.

CHAPTER 2

John Furness, Nadine Clerc, Fivos Vogalis, and Martin J. Stebbing

THE ENTERIC NERVOUS SYSTEM AND ITS EXTRINSIC CONNECTIONS

The gastrointestinal tract is unique among mammalian organs in that it has extensive intrinsic neural circuits. These circuits form the enteric nervous system, which contains reflex pathways capable of functioning independently of central control, although the central nervous system normally modifies activity within the gut wall. Enteric neurons have essential roles in the control of motility, blood flow, water and electrolyte transport, and acid secretion in the digestive tract. The enteric nervous system is most commonly the medium through which extrinsic neurons control gastrointestinal function.

STRUCTURAL ORGANIZATION OF THE ENTERIC NERVOUS SYSTEM

The enteric nervous system consists of nerve cell bodies and their processes embedded in the wall of the gut. The number of enteric neurons in a human is estimated to be about 100 million, which is about the same number of nerve cells as in the spinal cord. [1], [2] The nerve cell bodies are grouped in small aggregates, the enteric ganglia, which are connected by bundles of nerve cell processes to form two major ganglionated plexuses in the tubular digestive tract: the myenteric plexus, also called the *Auerbach plexus,* and the submucosal plexus, which is often referred to as the *Meissner plexus*.

Locations of the Enteric Ganglia

The myenteric plexus lies between the longitudinal and circular layers of the muscularis externa and forms a continuous network around the circumference of the tubular digestive tract from the upper esophagus to the internal anal sphincter (Fig. 2-1). In the parts of the large intestine where the longitudinal muscle is gathered into teniae, the myenteric plexus is prominent underneath the teniae and is sparser over the rest of the colonic surface.

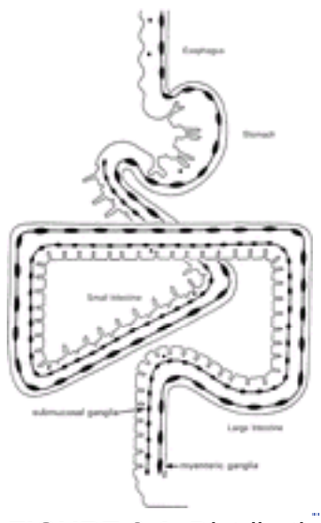

FIGURE 2-1. Distribution of enteric ganglia in the tubular digestive tract. The gastrointestinal tract is depicted in longitudinal section to reveal the myenteric ganglia, which form a continuous plexus from the upper esophagus to the internal anal sphincter, and the submucosal plexus, which is prominent in the small and large intestines. Isolated ganglia occur in the gastric and esophageal submucosa and in the mucosa throughout the digestive tract.

The submucosal plexus of ganglia is significant only in the small and large intestines; extensive networks of linked ganglia are not found in the submucosa of the esophagus and stomach, although isolated ganglia are sometimes encountered in these regions.

Ganglia are occasionally found in the mucosa, specifically in the connective tissue close to the muscularis mucosae. [3] Small ganglia are also found along the extrinsic nerves (i.e., vagus, pelvic, and mesenteric nerves) as they enter the gut. Some of the ganglia associated with the extrinsic nerves are located on the surface of the gut, particularly in the stomach and rectum; these are referred to as *subserosal ganglia*. Enteric ganglia are also present in the gallbladder, biliary ducts, and pancreas.

Nonganglionated Plexuses

A series of nonganglionated plexuses supply effector tissues of the tubular digestive tract: the longitudinal muscle plexus, the circular muscle plexus (and its subdivisions), the plexus of the muscularis mucosae, the mucosal plexus, and the perivascular plexuses (Fig. 2-2). The exception is the striated component of the external musculature of the esophagus, which is found in the upper third of the human esophagus and in a greater proportion of the esophagus in most other species.

The striated muscle cells are innervated at motor endplates, not by a nerve plexus.

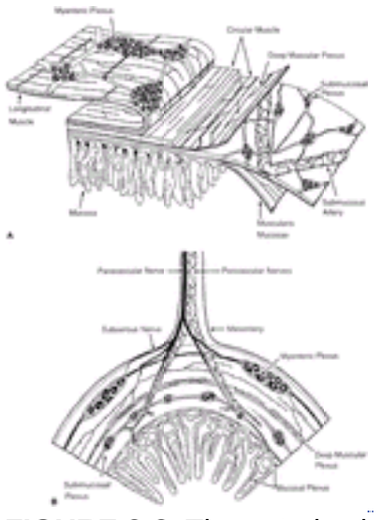

FIGURE 2-2. The enteric plexuses as they are seen (**A**) in whole mounts and (**B**) in transverse section. The drawings depict the small intestine. There are two ganglionated plexuses, the myenteric and the submucosal, in addition to plexuses of nerve fibers in the muscle and the mucosa and around the arterioles. (From ref. [1], with permission.)

The pattern of innervation of the longitudinal muscle differs according to its bulk. In humans, and in other species in which the longitudinal muscle is a thick layer throughout the intestine, the nerve fiber bundles run parallel to the muscle and comprise the longitudinal muscle plexus. In some species, such as the rabbit and the guinea pig, the thickness of the longitudinal muscle of the small intestine is less than about ten muscle cells. In these species, the nerve fiber bundles do not form a plexus within the longitudinal muscle, but rather a tertiary plexus, a component of the myenteric plexus, that lies against the inner surface of the longitudinal muscle (Fig. 2-3). [3], [4] and [5] The processes of individual tertiary plexus neurons ramify extensively on the inner surface of the longitudinal muscle. [6]

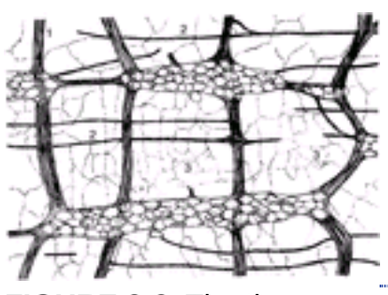

FIGURE 2-3. The three components of the myenteric plexus found in small animals are shown in a drawing of a whole mount from a guinea pig small intestine. Common to all species is the primary component of the plexus (*1*), consisting of the ganglia and internodal strands (interganglionic connectives), and the secondary component (*2*), consisting of nerve strands lying parallel to the circular muscle. The tertiary plexus (*3*) is found only where the longitudinal muscle is thin; in such regions, little or no longitudinal muscle plexus is found within the longitudinal layer (calibration bar = 100 µm). (From ref. [3], with permission.)

The circular muscle plexus is formed by parallel bundles of nerve fibers throughout this muscle layer. In some regions of the gastrointestinal tract, a dense layer of nerve fiber bundles provides additional innervation of the inner part of the circular muscle. The presence of this plexus of nerve fibers and its position in relation to the circular muscle differs among regions. In the small intestine of mammals, including humans, and most other vertebrates, the circular muscle consists of a thick outer layer and a thin inner layer of muscle cells. [7], [8] The dense plexus of nerve fibers located between these two muscle layers in the small intestine is called the *deep muscular plexus*. [9]

The colon of most species lacks an inner specialized layer of circular muscle; a dense layer of nerve fibers, similar to the deep muscular plexus, lies close against the inner surface of the circular muscle, adjacent to connective tissue of the submucosa. A term introduced by Cajal, the *submuscular plexus,* is used to refer to this plexus. [9], [10] and [11] An inner specialized muscle layer has been found in the colon of human and mouse. [12], [13] In the human colon, a deep muscular plexus is found between the two circular muscle layers. The circular layer of the stomach lacks an inner layer of specialized smooth muscle. [10] No dense plexus of nerve fibers near the inner surface of the circular muscle is readily discerned in the stomach of dogs and guinea pigs, although Faussone Pellegrini and colleagues [13] described a submuscular plexus in the human stomach. The circular muscle plexus continues into the circular muscle of the smooth muscle sphincters of the digestive tract without any apparent change in form. The myenteric plexus also continues into the sphincter regions.

The muscularis mucosae throughout the digestive tube consists of outer longitudinal and inner circular layers of smooth muscle innervated by nerve fibers running parallel to the muscle bundles. In the small intestine, muscle bundles extend into the cores of the villi. In the small intestine of small animals, such as mice and rats, the muscularis mucosae is thin and barely discernible in histological sections.

The mucosal plexus is a network of fine nerve fiber bundles that lies beneath the mucosal epithelium. It is sparse in the esophagus but prominent in the stomach, small intestine, colon, and gallbladder. The mucosal plexus in the small intestine is sometimes described as having subglandular, periglandular, and villous components. These components are continuous with one another, although their respective nerve fiber populations differ. [14]

Perivascular plexuses are found around the arteries leading into the gut wall and around the arterioles within the gut. Innervation of veins within the gut wall is sparse or nonexistent, but the mesenteric and hepatic portal veins in most species are innervated. Lymphatic vessels in the gut wall appear to lack innervation.

Myenteric Plexus

The arrangement of an area of the myenteric plexus of the human small intestine is shown in Figure 2-4. The thickness of the ganglia, which are flattened in the plane of the plexus, is usually one to four nerve cells, depending on the state of contraction and the size of the intestine. [15] In any region of the gastrointestinal tract, ganglion size varies widely. In the guinea pig ileum (see Fig. 2-3), ganglia range from a single cell to about 200 nerve cell bodies.

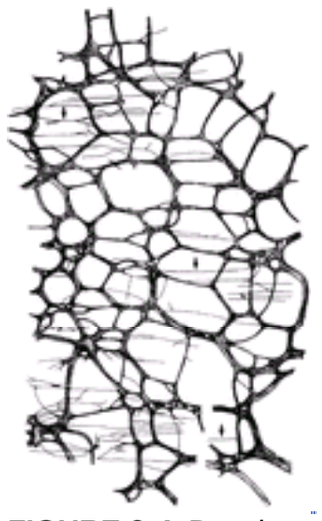

FIGURE 2-4. Drawing of a whole mount of the myenteric plexus of the human small intestine, prepared by Auerbach and published in *Henle's Textbook of Histology* in 1871. Myenteric ganglia, internodal strands, and small nerve trunks of the secondary component of the myenteric plexus (*arrows*) can be seen (calibration bar = 1 mm).

The pattern of ganglia, determined by shape and orientation, differs among regions and species but is often readily identifiable as belonging to a particular part of the intestine. [3], [16] The ganglia are connected to each other by small bands of nerve fibers, known as *internodal strands* or *interganglionic connectives* (see Fig. 2-3 and Fig. 2-4). The meshwork formed by the ganglia and the internodal strands is called the *primary component of the myenteric plexus*. Nerve strands that connect with the primary plexus and run circumferentially constitute the *secondary component of the plexus* (see Fig. 2-3 and Fig. 2-4). [4], [17] Branches from these secondary strands innervate the circular muscle. [18] Nerve fiber bundles (i.e., penetrating fiber bundles) also run through the circular muscle to connect the myenteric plexus with the submucosal and mucosal plexuses. [19]

The major targets for nerve cells in the myenteric plexus are the muscularis externa (which receives most of its innervation from this source), the submucosal ganglia, and other myenteric nerve cells. Myenteric nerve cells also supply the mucosa with nerve fibers, many of which have sensory nerve endings, and they give rise to nerve fibers that project to sympathetic prevertebral ganglia.

Submucosal Plexus

A continuous network of numerous small ganglia is found in the submucosa throughout the small and large intestines. In many species, plexuses of ganglia run in an inner and an outer layer, but some regions in small species have only one layer. [20], [21] The inner and outer plexuses contain different populations of neurons, defined by their morphologic and histochemical characteristics. [22] The electrophysiological properties of the nerve cell populations in the two plexuses also differ. [23] It is assumed that the inner plexus (closer to the mucosa) is mainly concerned with control of fluid movement, and the outer plexus contributes to control of motility of the external muscle. Submucosal ganglia are smaller and less regularly arranged than myenteric ganglia.

The submucosal plexus harbors the cell bodies both of neurons with dual secretomotor/vasodilator function and of neurons that are solely secretomotor. [24], [25] and [26] Some neurons, notably intrinsic primary afferent neurons, project from the submucosa to the myenteric plexus. [27] Other submucosal neurons probably supply the muscularis mucosae of the small and large intestines. [11] Submucosal neurons supply a part of the innervation of the inner circular muscle in some species, including human. [11], [28], [29] and [30] In the stomach, which almost entirely lacks submucosal ganglia, the intrinsic innervation of the mucosa and muscularis mucosae comes from the myenteric plexus.

Ganglia of the Gallbladder, Biliary Ducts, and Pancreas

Because the biliary system and the pancreas develop from diverticula of the small intestine, the ganglia in their walls are part of the enteric nervous system. A plexus of ganglia, similar to the enteric plexuses of the small intestine but differing between species in its relation to tissue layers, is found in the gallbladder, cystic duct, and common bile duct. [3] Numerous nerve fibers are found in the muscle, around blood vessels, and in the mucosa of the biliary tract. Ganglia, connected to each other by small nerve trunks, are scattered through the pancreas, forming a three-dimensional plexus in this solid organ. Nerve fibers are found around the acini and the blood vessels and in the islets. In the gallbladder, the intrinsic neurons control motility and the flux of water and electrolytes. In the pancreas, they appear to be involved in the control of both the endocrine and exocrine components. Nerve fibers connect the plexuses of the biliary system and pancreas with the ganglionated plexuses of the upper small intestine. [31], [32] and [33] These connections are presumed to contribute to coordination between these organs and the gastrointestinal tract.

MICROSCOPIC STRUCTURE OF THE ENTERIC NERVOUS SYSTEM

Shapes of Neurons

The nerve cells of the enteric ganglia can be classified into subgroups according to their shapes. [34], [35], [36] and [37] The first effective classification was by Dogiel, [34] who proposed that the shapes of nerve cells are related to their functions. Dogiel defined three cell shapes: types I, II, and III. The first two are readily recognized in different species and with various staining techniques; the third type is less well defined, but it bears some resemblance to a group later referred to as *filamentous neurons*. Type I neurons are generally flattened in the plane of the ganglia; they have oval cell bodies, prominent flattened (lamellar) dendrites, and a single long axon often characterized by spiny protuberances close to the cell body. Type II neurons have a spheroidal shape and give rise to several axons, usually three to ten, although some type II neurons are pseudounipolar. [35] A few type II neurons have tapering dendrites in addition to several long axonlike processes and are referred to as *dendritic type II cells*. [36], [38]

A variety of other shapes have been described, notably in the pig and guinea pig intestines, which have been studied intensively in recent years. Stach [36] extended Dogiel's classification to include types IV, V, and VI and mini-neurons, based primarily on work in the pig. Type IV neurons may be secretomotor neurons in both the guinea pig and pig. In the guinea pig small intestine, filamentous and small simple neurons (the latter being similar to the mini-neurons in the pig) have been described. [35]

Numbers of Neurons

Counts of nerve cell densities published up to 1985 were tabulated by Furness and Costa. [3] In the myenteric plexus, densities of about 2000 to 20,000 nerve cells per square centimeter were found in a variety of regions and species. In the submucosal plexuses of the small and large intestines, recorded densities range from about 1000 to 5000 nerve cells per square centimeter. Studies emphasize that the accuracy of counts of neurons depends on the methods used and that many published counts are probably underestimates. [2] A thorough study in the guinea pig concluded that there are 6.5 million nerve cells in the myenteric plexus of the small intestine and 7.3 million in the myenteric plexus of the colon. [2]

Ultrastructure

The enteric nervous system consists of a compact arrangement of nerve cell bodies, nerve fiber terminals, bundles of nerve fibers, and neuroglia. Unlike other autonomic ganglia, the enteric ganglia do not contain blood vessels or connective tissue cells, although septa of connective tissue sometimes separate parts of the ganglia in large species, including humans. Nutrients are supplied to the ganglia through the surrounding interstitial fluid. Similarly, hormones or drugs that act on the enteric ganglia are absorbed from the interstitial fluid. It may be significant that the cell bodies of many enteric neurons present large surface areas, devoid of neuroglia, to the extraganglionic space. [39], [40]

Nerve fibers in the enteric plexuses are of two types: fibers of relatively uniform diameter, about 0.2 to 0.5 µm; and fibers that consist of varicosities, usually about 1 to

2 µm in diameter, connected by short intervening segments that measure about 0.2 to 0.5 µm. The nonvaricose axonal segments are nonterminal regions that do not appear to release transmitter, whereas the varicosities are considered to be points of transmitter release. The varicose segments of enteric axons, including those within plexuses and those associated with muscle or mucosal epithelium, are often hundreds of micrometers or several millimeters long. This means that a single axon can contact many nerve cell bodies. The close contacts with nerve cells and their dendrites are of two types: those that have synaptic specializations, consisting of presynaptic accumulations of vesicles and postsynaptic densities; and those in which varicosities containing numerous synaptic vesicles come close to the target neuron, with no intervening glial cell and an absence of postsynaptic density. The latter nonspecialized association is the most common. Most enteric neurons receive both specialized and nonspecialized contacts. [40], [41], [42] and [43]

The nerve fibers that innervate the muscle and epithelium are arranged in bundles in which the individual fibers are partly surrounded by enteric neuroglia. [39] Although there are no obvious postjunctional specializations in which the varicosities come close to muscle cells, such varicosities may make close contacts with muscle cells that are associated with prejunctional specializations, particularly junctional clumping of vesicles. [44]

HISTOCHEMICAL PROFILES AND TRANSMITTER MULTIPLICITY OF ENTERIC NEURONS

The enteric nervous system contains more than 30 substances that affect the activity of neurons, muscle, and epithelial cells—substances that also may be neurotransmitters. [45], [46] and [47] Moreover, individual neurons contain several possible neurotransmitters in addition to other neuron-specific proteins. The combinations of chemical attributes, which are related to the functions of the neurons and their places in nerve circuits, provide a chemical code by which the neurons can be identified. In general, more than one substance contributes to the transmission process, which is therefore described as *plurichemical*. [48]

Co-localized substances do not have equal status as transmitters. [48] One substance (sometimes more) has the major role in transmission; this substance is the primary transmitter. For example, sympathetic vasoconstrictor neurons in many species contain norepinephrine and neuropeptide Y (NPY). The primary transmitter is norepinephrine; NPY has a subsidiary or modifying role. However, the relative contributions of different transmitters can vary between neurons with common functions, depending on the location and the activity of the neurons; the inhibitory muscle motor neurons of the gut are a good example (see section “ Motor Neurons”). Some neurons contain more than one primary transmitter and several subsidiary transmitters or neuromodulators; some classes of enteric neurons have five or more transmitters (Table 2-1).

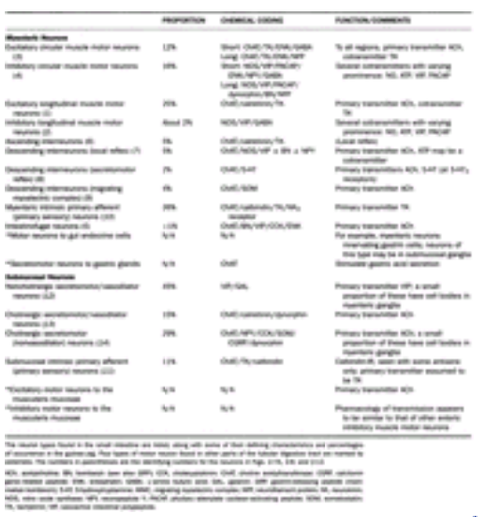

TABLE 2-1 Types of Neurons in the Enteric Nervous System

The chemistries of the subsidiary transmitters of functionally equivalent neurons differ between species. For example, the sympathetic noradrenergic neurons that inhibit intestinal water and electrolyte secretion contain somatostatin in the guinea pig but not in other species, and the noradrenergic neurons that inhibit motility contain NPY in the rat but not in the guinea pig. A broad range of experiments suggest that the primary transmitters are constant between species; many of the observed species differences in neuronal chemistry are probably differences in subsidiary transmitters or neuromodulators. In this chapter, we do not discuss all the substances that may participate in enteric neurotransmission in different species; much of this information is covered in published reviews. [45], [46] and [47]

The immunohistochemical localization of specific combinations of compounds in neurons provides a valuable investigative probe that allows the projections of individual neurons to be determined and their functions to be deduced, even when the roles of the detected chemicals are not known. For example, the inhibitory and excitatory muscle motor neurons in the gastrointestinal tract can be localized by immunoreactivity to vasoactive intestinal polypeptide (VIP) and substance P, respectively.

PHYSIOLOGICAL CHARACTERISTICS OF ENTERIC NEURONS

Electrophysiological Properties of Enteric Neurons

The electrophysiological classification of enteric neurons is confounded because the properties of the neurons differ between regions of the digestive tract, and even in a specific region, properties can vary depending on the circumstances of the experiment. [49], [50] and [51] However, classification is particularly useful when the electrophysiological type can be correlated with the physiological function of the neuron and with its shape and histochemical profile.

In the small intestine of the guinea pig, neurons can be separated into two groups, AH and S, according to their electrophysiological properties (Fig. 2-5). These terms were introduced in 1974, [52] and the classification remains useful, although the reasons for calling them *AH* and *S* are no longer well justified.

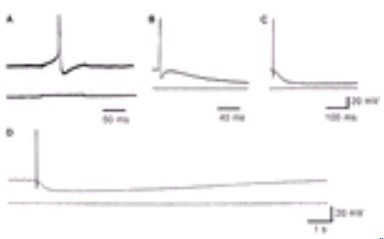

FIGURE 2-5. Action potentials elicited in an S neuron (**A**) and in AH neurons (**B–D**) by intracellular current pulses. The action potential in the S neuron is followed by a brief period of hyperpolarization; the action potentials in the AH neurons are followed by two phases of hyperpolarization (early and late afterhyperpolarizing potentials), one brief (**B**) and the other prolonged (**C,D**). Note the differences in the time scales. (From ref. [52], with permission.)

The S neurons exhibit brief action potentials that are followed by short-duration afterhyperpolarizing potentials (AHPs) lasting 20 to 100 milliseconds. These action potentials are blocked by tetrodotoxin. S neurons have a variety of shapes, which include the Dogiel type I and filamentous morphologies, but never the Dogiel type II morphology. All S neurons have a single axon. They exhibit fast excitatory postsynaptic potentials (EPSPs), and compound fast EPSPs of sufficient amplitude to generate action potentials can be evoked in all S neurons. These EPSPs are referred to as *compound* because they are the summed effects of transmission from

several axons. Slow EPSPs are also recorded from S neurons.

The action potentials of the AH neurons in the guinea pig small intestine are large (i.e., 75 to 110 mV in amplitude) and of longer duration than those of the S neurons, and they have an inflection (hump) on the falling phase. They are normally followed by two separate phases of hyperpolarization (see Fig. 2-5). The early AHP lasts 20 to 100 milliseconds and is similar to the undershoot usually seen in neurons (including S neurons); it is the result of the activation of K^+ channels. The second hyperpolarization, the late AHP, lasts 2 to 25 seconds (see Fig. 2-5); it is the result of the opening of Ca^{2+}-sensitive K^+ channels. [53], [54] and [55] These neurons can generate a soma action potential that is carried by Ca^{2+} and a slow afterhyperpolarization, even when all the voltage-sensitive Na^+ channels are blocked by tetrodotoxin. [52], [56] The Ca^{2+} that enters during the action potential triggers the release of Ca^{2+} from intracellular stores; it is this Ca^{2+} that opens the K^+ channels, which in turn elicit the AHP. [57] In some circumstances, such as during excitation by transmitters or hormones, the prolonged afterhyperpolarization is not apparent. [49] In the guinea pig small intestine, all AH neurons have Dogiel type II morphology. AH neurons in the guinea pig ileum usually do not exhibit fast EPSPs, and when they are recorded, the EPSPs have very small amplitudes. [50], [52], [58] On the other hand, slow EPSPS do occur in AH neurons and can trigger action potentials.

The S and AH nomenclature does not apply, or is less useful, for some other gut regions. For example, in the guinea pig rectum, some neurons that exhibit a prolonged AHP have a single axon and receive fast EPSPs. [59] In the gastric corpus, all myenteric neurons have S characteristics and do not exhibit a prolonged hyperpolarization after the action potential. [60], [61]

Synaptic Transmission in the Enteric Nervous System

Enteric neurons receive fast and slow EPSPs and inhibitory postsynaptic potentials (IPSPs). Transmitter release from terminal axons may be reduced by presynaptic inhibition.

Fast EPSPs are graded in amplitude in relation to the strength of electrical stimulation, and when they are evoked reflexively, individual EPSPs of different amplitudes are observed (see below, Fig. 2-11). Thus, enteric neurons receive multiple fast synaptic inputs. The EPSPs last about 15 to 30 milliseconds. They are associated with an increased membrane conductance, which is attributed primarily to Na^+. Several transmitters contribute to fast EPSPs in S neurons (Fig. 2-6). Acetylcholine (ACh) appears to be one transmitter of fast EPSPs because antagonists of nicotinic cholinergic receptors reduce EPSP amplitude. [52], [62] Some fast EPSPs are in fact completely blocked by nicotinic receptor antagonists, whereas others are reduced only in amplitude, some by as little as 20% (see Fig. 2-6). [63], [64] and [65] In about 60% of neurons with a hexamethonium-resistant fast EPSP, the EPSP after nicotinic block was reduced in amplitude or abolished by PPADS, an antagonist of P_{2X} receptors. [65] This indicates that a purinergic (i.e., adenosine triphosphate [ATP]-mediated) transmission underlies some fast EPSPs. [65], [66] Neurons that use ATP as a transmitter project anally in the myenteric plexus. [65], [66] In about 10% of neurons, a 5-HT_3 receptor antagonist blocks or reduces fast EPSPs that remain in the presence of hexamethonium. [67] Those fast EPSPs that are abolished by hexamethonium are purely cholinergic. However, PPADS and 5-HT_3 receptor antagonists can further reduce responses that have already been reduced by hexamethonium. Thus, at some synapses, ATP and 5-HT could be cotransmitters with ACh. In some S neurons, the three antagonists in combination do not fully block fast EPSPs, which implies the existence of one or more other transmitters of fast EPSPs. Pharmacological and immunohistochemical data suggest glutamate may be a transmitter, acting through AMPA receptors, and that the fast EPSPs seen in a minority of AH neurons may also be mediated by glutamate. [68], [69]

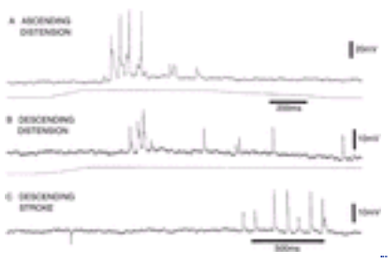

FIGURE 2-11. Intracellular recordings taken from circular muscle motor neurons of the guinea pig small intestine during intestinal reflexes. **A:** The response in an S neuron evoked by a distention of the intestinal wall 25 mm on the anal side of the point of recording. **B:** The response in a different S neuron to distention applied 35 mm on the oral side of the point of recording. **C:** The response of the same neuron illustrated in **B** to mechanical stimulation of the mucosa, without distention, by gentle stroking 35 mm on the oral side of the point of recording. In each case, the stimulus evoked a burst of fast excitatory postsynaptic potentials.

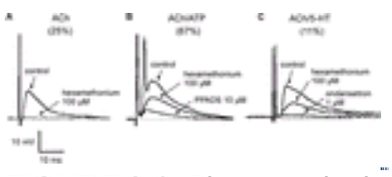

FIGURE 2-6. Pharmacological dissection of fast excitatory postsynaptic potentials (EPSPs) in S neurons of the myenteric plexus of the guinea pig ileum. **A:** A fully cholinergic EPSP is blocked by the nicotinic receptor blocker hexamethonium. **B:** A fast EPSP is partly reduced by hexamethonium. The remaining component is largely blocked by the purinergic P_{2X} receptor antagonist PPADS. **C:** A fast EPSP in which the response remaining after the administration of hexamethonium is blocked by the 5-HT_3 receptor blocker ondansetron. *ACh,* acetylcholine. (From ref. [47], with permission.)

In the guinea pig small intestine, slow EPSPs usually last between 15 and 120 seconds and occur in at least 75% of S-type neurons and in probably all AH neurons. [51], [70] The slow EPSPs evoked by electrical stimulation are likely to be the result of the superimposed actions of several different transmitters, each producing superficially similar synaptic potentials. [70] Electrical stimulation of internodal strands typically evokes cholinergic (via muscarinic receptors) and noncholinergic slow EPSPs in S neurons and in some AH neurons. [71] Other AH neurons exhibit only noncholinergic slow EPSPs. By simultaneously recording from an AH neuron and from an S or another AH neuron in the same ganglion, it has been shown that AH neurons provide slow excitatory synaptic inputs to S neurons and to AH neurons. [72] Other experiments indicate that AH neurons activated by sensory stimuli cause slow EPSPs in AH and S neurons. [51], [73] No evidence suggests that S neurons are a source of slow EPSPs, although adequate research to exclude this possibility has not been carried out.

The nature of the transmitter responsible for the noncholinergic slow EPSPs has been the subject of extensive speculation. [74] Although several substances found within enteric nerve terminals mimic slow EPSPs, including 5-HT, substance P, VIP, and somatostatin, pharmacological and other data indicate that one of the transmitters mediating this response is a tachykinin. [75], [76], [77] and [78] The majority of AH neurons have tachykinin immunoreactivity, but they never contain 5-HT, somatostatin, or VIP. [46]

A combination of membrane changes underlie slow EPSPs: reduction in the K^+ permeability of the neuronal membrane (including inhibition of the gK_{Ca} that is important for the late AHP after an action potential); an increase in Cl^- conductance; and block of a tetraethylammonium-sensitive outward rectifier current. [51], [79], [80] Reducing the permeability of the membrane to K^+ increases the neuronal excitability in two ways. First, it depolarizes the cell membrane, bringing it closer to the threshold potential needed to trigger an action potential. Second, increasing the input resistance of the cell enhances the effect of other currents on the membrane potential, making fast EPSPs more effective.

Inhibition of the outward rectifier current has a profound effect on the excitability state of S neurons; instead of firing briefly in response to depolarization, the neurons can fire tonically and at high rates. [51] Slow EPSPs in AH/Dogiel type II neurons affect neuronal excitability by reducing or abolishing their characteristic slow AHP,

presumably by inhibiting opening of the Ca^{2+}-sensitive K^+ channels. This inhibition facilitates the firing of prolonged, high-frequency trains of action potentials, which is an effect similar to that produced by the inhibition of the outward rectifier current, although through a different mechanism.

An unusual type of slow excitatory transmission, termed *sustained slow postsynaptic excitation* (SSPE), has been described in myenteric AH neurons.[81] Activation of presynaptic axons supplying AH neurons for extended periods (1–30 minutes) at low frequency (1 Hz) gives rise to a slowly developing, sustained increase in the excitability of the neurons associated with depolarization, increased input resistance, and suppression of the late AHP. Successive stimulus trains (duration, 1–4 minutes) elicit successively greater increases in excitability. Increased excitability can last up to 3.5 hours after a stimulus period of 30 minutes. This phenomenon may represent a type of neuronal memory and may be involved in adaptive responses.

IPSPs in myenteric neurons have been reported rarely, and then only in a small proportion of neurons in the guinea pig small intestine.[79,82] It is possible that electrical stimulation of internodal strands with one or more pulses affects both excitatory and inhibitory fibers. The excitatory responses may obscure any IPSPs that are evoked simultaneously. Studies in which enteric reflex pathways are activated by physiological stimuli have also failed to evoke IPSPs in myenteric neurons.[51,73,83,84,85,86] This suggests that any physiological role for this type of synaptic potential may be confined to a small group of myenteric neurons.

Electrophysiological studies of *submucosal* neurons have been performed in preparations from the small intestine, the cecum, and the distal colon of the guinea pig.[87,88,89,90,91] About 90% of all submucosal neurons exhibit fast EPSPs, and a large proportion of these also exhibit slow EPSPs, similar to those observed in myenteric neurons. The fast EPSPs are blocked by hexamethonium and are presumably mediated by ACh acting through nicotinic receptors. The slow EPSPs are probably caused by a reduction in K^+ permeability. Although several substances found within nerve terminals in the submucosal plexus can mimic the slow EPSPs, none has been shown unequivocally to be a transmitter. For example, lesions that cause a complete loss of substance P–containing nerve terminals in the submucosal plexus reduce the number of neurons with slow EPSPs.[92]

Although slow IPSPs are not observed in myenteric neurons, they are commonly seen in submucosal neurons. Stimulation of the internodal strands evokes a substantial IPSP in about 50% of submucosal neurons in the guinea pig small intestine and in as many as 90% of the neurons in the cecum.[87,88,89,90] In contrast, slow IPSPs were not observed in submucosal neurons of the guinea pig distal colon.[91,93] Most submucosal neurons with slow IPSPs also exhibit slow EPSPs, and all have fast EPSPs. In the small intestine, neurons with slow IPSPs contain VIP and are probably noncholinergic secretomotor/vasodilator neurons.[87,92]

The IPSP is caused by a substantial increase in the K^+ permeability of the membrane, which leads to a hyperpolarization of up to 30 mV. Two separate contributions to IPSPs have been identified, adrenergic transmission from the terminals of sympathetic secretomotor inhibitory neurons (see section "Sympathetic Effects on Motility and Secretion") and nonadrenergic transmission from intrinsic neurons.[87,94,95]

Lesion studies reveal that most submucosal neurons receive fast cholinergic input from neurons in the myenteric plexus and from other neurons in the submucosal plexus.[96] The slow EPSPs in many submucosal neurons appear to come from myenteric neurons, but some may come from submucosal neurons.[92,96]

Presynaptic inhibition may also have a role in the enteric nervous system. Sympathetic nerve stimulation reduces the amplitudes of fast EPSPs in both myenteric and submucosal neurons, probably by decreasing the amount of ACh released from the synapses,[97,98] although postsynaptic inhibitory effects may also play a role. It appears that the presynaptic inhibitory effect of the sympathetic neurons is the primary mechanism by which they diminish the contractile activity of the gut.[3] This inhibition is mediated through a-receptors. ACh released from enteric nerve terminals can act presynaptically to regulate the subsequent release of ACh and possibly to modify the release of the transmitters mediating slow EPSPs.[99] Several other putative transmitters contained within enteric nerve terminals (e.g., dynorphin, enkephalin, ?-aminobutyric acid, 5-HT) have been observed in pharmacological experiments to reduce transmitter release in the gastrointestinal tract. No physiological role has yet been established for these latter substances in presynaptic inhibition in the enteric nervous system.

FUNCTIONALLY DEFINED ENTERIC NEURONS

The identification of neurons by function has been derived by combining data from studies of the physiological properties of enteric reflexes, the morphologic identification of those neurons that are present (and could thus fulfill the physiological functions), and the correlation of neurochemical (primarily immunohistochemical) and pharmacological properties of enteric neurons (see Table 2-1). The model species has been the guinea pig. From these data has come a complete definition of the types of neurons in the small intestine,[100] and a thorough, but not yet complete, definition of the neuron types in the colon[101] and the stomach[102,103,104] (Fig. 2-7). Sufficient data are available from other species, notably human, pig, rat, and mouse, to suggest that the data from guinea pig can be extrapolated. The main difference between species is the placement of cell bodies of some neurons; specifically, the cell bodies of motor neurons of the external muscle are present in both the myenteric and submucosal plexuses in human and pig, whereas they are confined to the myenteric plexus in guinea pig, rat, and mouse.

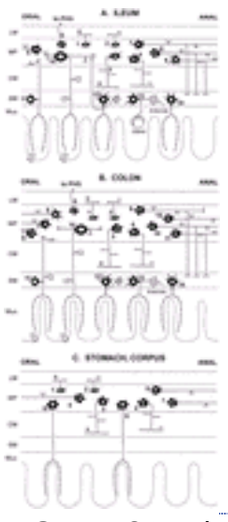

FIGURE 2-7. The types of neurons, as defined by their functions, cell body morphologies, chemistries, and projections, in the ileum (**A**), colon (**B**), and gastric corpus of the guinea pig (**C**). Types 1 through 4 are found in each region; types 5 and 10 through 14 are found in ileum and colon. *1,* Excitatory longitudinal muscle motor neuron; *2,* inhibitory longitudinal muscle motor neuron; *3,* excitatory circular muscle motor neuron; *4,* inhibitory circular muscle motor neuron; *5,* intestinofugal neuron (rare in stomach); *10,* myenteric intrinsic primary afferent neuron (IPAN); *11,* submucosal IPAN; *12,* noncholinergic secretomotor/vasodilator neuron; *13,* cholinergic secretomotor/vasodilator neuron; *14,* cholinergic secretomotor (nonvasodilator) neuron. **In the ileum (A):** *6,* ascending interneuron; *7,* descending interneuron (local reflex); *8,* descending interneuron (secretomotor reflex); *9,* descending interneuron (migrating myoelectric complex). The numbers adjacent to the neurons correspond to the numbers in Table 2-1, which lists all the neuron types according to function and provides data on the percentage of the cell bodies of each type in the myenteric or the submucosal ganglia, in addition to information about their chemistries. **In the colon (B):** *6, 7, 8,* types of ascending interneurons; *9, 15, 16, 17,* types of descending interneurons. **In the corpus of the stomach (C):** *8, 9,* neurons that project from the myenteric plexus to the mucosa; some of these are probably involved in the control of gastric acid secretion. IPANs are not present, and neuron cell bodies in the submucosal layer are rare or absent. *LM,* longitudinal muscle; *MP,* myenteric plexus; *CM,* circular muscle; *SM,* submucosal plexus; *Muc,* mucosa; *PVG,* prevertebral ganglia.

Motor Neurons

Muscle Motor Neurons of the Stomach and Intestines Excitatory neurons innervate the longitudinal and circular smooth muscle and the muscularis mucosae throughout the digestive tract. The primary transmitter of these neurons is ACh, which acts on the muscle through muscarinic receptors.[3] Tachykinins contribute to the excitatory transmission but have a lesser role than ACh, except perhaps in the rat small intestine.[105,106] and[107] The tachykinins in the enteric neurons are substance

P, neurokinin A, neuropeptide K, and neuropeptide-?. Direct experimental evidence has been obtained for the involvement of two of these, substance P and neurokinin A, in neuromuscular transmission. [108] The tachykinin component of excitatory transmission appears to be more prominent at high rates of neuron firing. Immunohistochemical studies and quantitative electron microscopy reveal that ACh and tachykinins are contained in the same excitatory muscle motor neurons. [109], [110] In the guinea pig small intestine, these neurons, and the inhibitory motor neurons, are S neurons by electrophysiological classification. Intrinsic motor neurons that relax the muscle are involved in descending reflexes that facilitate the passage of contents along the bowel and relax regions, such as the stomach, that expand to accommodate and retain their contents. These neurons, called *enteric inhibitory neurons,* release a combination of transmitters that contribute to relaxation in varying degrees, depending on the region and the species. [48], [111], [112], [113] and [114] The neurotransmitters are nitric oxide, ATP, pituitary adenylate cyclase–activating peptide (PACAP), and VIP. VIP and the enzyme nitric oxide synthase are found together in the inhibitory motor neurons. [115] Nitric oxide synthase inhibitors and scavengers of nitric oxide reduce inhibitory transmission in many species, including humans, indicating that nitric oxide is an inhibitory transmitter. [116], [117] However, electrophysiological and pharmacological evidence indicates that at least one other primary transmitter must be involved in the transmission from these neurons. [48] ATP relaxes the muscle, and when the inhibitory neurons are active, increased levels of ATP and its metabolites are detected in perfusates. [118] Suramin, an antagonist of the action of ATP on P_2 purinoceptors in intestinal muscle, can depress transmission from the inhibitory neurons of the guinea pig tenia coli. [119] A contribution of VIP to inhibitory transmission has been demonstrated in some gastrointestinal muscles, but in muscle in other regions, it makes little contribution. [120] Peptide histidine isoleucine (PHI), or its human equivalent, PHM, is derived from the same gene as VIP and has similar effects. Another member of the VIP family, PACAP, also relaxes intestinal muscle and is found in enteric neurons. [121] Immunoneutralization and pharmacological antagonism of the PACAP receptors both indicate that PACAP contributes to enteric inhibitory transmission in some regions. [114], [122] Thus, nitric oxide, VIP or PHI, PACAP, and ATP probably all contribute to the inhibition of transmission to muscle. In most instances, nitric oxide is a primary transmitter, and the contributions of VIP, PACAP, and ATP vary.

Muscle Motor Neurons of the Striated Esophagus The striated muscle of the esophagus is innervated by axons that form motor endplates, but unlike motor endplates elsewhere, individual endplates in the esophagus receive dual innervation, one axon being from a vagal motor neuron with its cell body in the medulla oblongata and the other originating from a cell body in the myenteric plexus. [123], [124] and [125] In the rat, the vagal endings are immunoreactive for calcitonin gene–related peptide (CGRP), and the endings of myenteric origin exhibit nitric oxide synthase immunoreactivity. Double staining with the use of these markers indicates that both fibers make synaptic connections with the muscle, and that the two fiber types are often closely apposed, facilitating presynaptic interaction. [125]

Secretomotor and Secretomotor/Vasodilator Neurons Controlling Fluid Exchange Secretomotor neurons in the small and large intestines and gallbladder enhance water and electrolyte secretion. Some enteric neurons send axons both to the mucosal epithelium, where the effect is to cause water and electrolyte secretion, and to the submucosal arterioles, where they have vasodilator actions (see section “ Enteric Pathways for Secretomotor and Vasodilator Control”). For practical reasons, in most studies of the functions of these neurons, only one outcome of their activity, secretion or vasodilation, has been measured. Nevertheless, if the different experiments are considered together, it can be deduced that some neurons have only a secretomotor role and some a dual secretomotor/vasodilator role (Fig. 2-8). Secretomotor transmission to the mucosa has both cholinergic and noncholinergic components. [126] ACh released from the cholinergic neurons acts on muscarinic receptors on the mucosal epithelium. The primary transmitter of the noncholinergic secretomotor effect is probably VIP. [127] In addition, HCO_3^- secretion in the duodenum and jejunum is augmented by secretomotor neurons. [128] Local vasodilator reflexes can be elicited in the intestines by mechanical or chemical irritation of the mucosa, and substantial evidence indicates that the vasodilator neurons are intrinsic to the intestine and are noncholinergic. [129] Experiments in which single neurons were stimulated, and the resulting changes in the diameter of submucosal blood vessels measured, provide direct evidence for the presence of both submucosal cholinergic and noncholinergic vasodilator neurons. [25], [130] Immunohistochemical studies, combined with denervation, confirm that both intrinsic cholinergic and noncholinergic neurons innervate submucosal arterioles. [131] It is probable that a primary effector of noncholinergic transmission is VIP. [3], [129], [132] In the colon, but not the ileum, the mucosa receives a polarized innervation by descending noncholinergic and ascending cholinergic neurons of the submucosal plexus. [133]

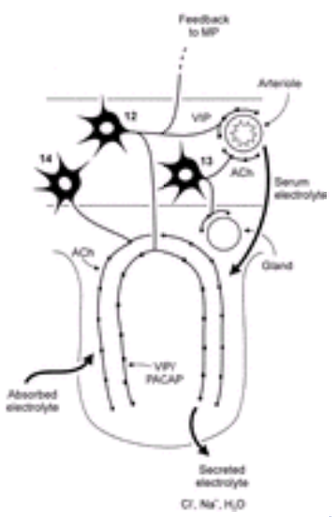

FIGURE 2-8. Neurons with secretomotor and vasodilator actions in the small intestine of the guinea pig. Functional evidence, supported by immunohistochemical and projection data, indicates that secretomotor effects are exerted through motor neurons, some of which are secretomotor/vasodilator and some of which are secretomotor only. The numbers adjacent to the neurons correspond to the numbers in Table 2-1, which lists all the neuron types according to function and provides data on the percentage of the cell bodies of each type in the myenteric or the submucosal ganglia, in addition to information about their chemistries. *ACh,* acetylcholine; *MP,* myenteric plexus, *PACP,* pituitary adenylate cyclase–activating peptide; *VIP,* vasoactive intestinal polypeptide.

Gastric Vasodilator Neurons Gastric acid secretion and blood flow are enhanced when the vagus nerve is stimulated; these effects are reduced by muscarinic antagonists. In most experiments, it is not possible to determine whether vasodilation that is the result of a direct vascular action of cholinergic neurons occurs in addition to a functional hyperemia consequent to the increased secretion. [132] However, Thiefin and colleagues [134] showed that centrally administered thyrotropin-releasing hormone stimulated a vagal pathway in the rat that caused gastric vasodilation after acid secretion was blocked by omeprazole. The increased blood flow in the absence of secretory change was antagonized by atropine. There is also evidence for noncholinergic gastric vasodilator neurons that use VIP as a transmitter. [135]

Gastric Secretomotor Neurons That Stimulate Acid Output Some secretomotor neurons govern gastric acid secretion. These neurons are cholinergic and act on the parietal cells through muscarinic receptors. For a complete description of the roles of these neurons and their relation to the hormonal control of gastric acid secretion, see Chapter 13.

Motor Neurons Innervating Enteric Endocrine Cells A variety of endocrine cells reside in the mucosa of the gastrointestinal tract, and because the mucosa is densely innervated, most of these cells have nerve fibers in close proximity. The best-documented motor neurons innervating the enteric endocrine cells are those controlling the release of gastrin, which is under the influence of vagal and intrinsic gastric pathways. The final neurons in both paths are in the stomach wall. Transmission from the final secretomotor neurons is mediated at least in part by gastrin-releasing peptide. [120] Release from other enteroendocrine cells is also likely to be under neural control. For example, the basal release of motilin is reduced by atropine and by tetrodotoxin and stimulated by muscarinic agonists; these findings suggest that motilin cells receive an excitatory cholinergic input. [136]

Motor Neurons Innervating Lymphoid Tissue (Peyer Patches) Evidence suggests that enteric neurons innervate Peyer patches, [137], [138] and [139] and that receptors for enteric neuron transmitters reside on lymphocytes therein. [140]

Enteric Interneurons

Interneurons have been identified in all gut regions, although their characteristics may vary between regions more than those of other neuron types. For example, the ileum and colon contain the same, or very similar, motor neurons and intrinsic primary afferent neurons, but their complements of interneurons are quite different (see Fig. 2-7).

Within the myenteric plexus, the interneurons form chains of like neurons that run both orad and toward the anus. [140], [141] In the guinea pig small intestine, at least three classes of descending interneurons and one class of ascending interneurons exist. Detailed studies of synaptic connections indicate that the chains formed by two of the types of descending interneurons interconnect. [142] The ascending interneurons appear to be involved in local motility reflexes, as are the descending cholinergic neurons that contain nitric oxide synthase. [26] Another type of descending interneuron, the ACh/somatostatin interneurons, may be linked to the passage of the migrating myoelectric complexes (MMCs). These neurons have a distinctive appearance (filamentous neurons) with numerous branching, tapering, filamentous dendrites [140]; they have very rare connections from intrinsic primary afferent neurons (IPANs). [143], [144] MMCs are waves of excitatory activity that are conducted

aborally along the gut. They are mediated through the intrinsic neural pathways of the small intestine. [145] Interruption of the continuity of the enteric nervous system blocks conduction of the MMC. [146] MMCs begin in the duodenum and travel the full length of the small intestine, but they are not conducted along the colon. It is thus pertinent that the ACh/somatostatin filamentous descending interneurons are found in both the ileum and the duodenum but not in the proximal or distal colon. The third type of descending interneuron, the ACh/5-HT interneuron, is possibly involved in secretomotor reflexes. [26] Interneurons also make connections between the myenteric and submucosal plexuses.

Recordings made during reflexes reveal that some transmission between interneurons is through fast EPSPs. [83] Pharmacological analysis indicates that transmission in the ascending pathway is cholinergic, through nicotinic receptors, whereas descending reflexes are resistant to blockade by the nicotinic antagonist hexamethonium. [75], [111], [147] Pharmacological data reveal that one of the transmitters of descending pathways is ATP, acting at P_{2x} receptors. [148] Nitric oxide, which is probably released from descending interneurons containing nitric oxide synthase, does not appear to be a primary transmitter. It appears to act as a retrograde transmitter, being released from the cell bodies of the interneurons and acting retrogradely on the endings of the terminals of IPANs to reduce transmitter release. [149]

Intrinsic Primary Afferent Neurons

The intrinsic reflex pathways involved in the control of gut movement, blood flow, and secretion are activated through neurons that respond to several stimuli, such as distention, lumenal chemistry, and mechanical stimulation of the mucosa. These intrinsic afferent neurons have now been positively identified as AH neurons with Dogiel type II morphology in the small intestine (see Chapter 3).

About 100 years ago, several investigators showed that enteric motility reflexes could be elicited in segments of intestine that had no neural connections with the central nervous system. [150], [151] and [152] It was therefore assumed that primary afferent neurons were contained in the gut wall. However, it was discovered at about the same time that reflexes, notably cutaneous vasodilator reflexes, could be initiated via axon collaterals even when the axons bearing the collaterals were disconnected from their cell bodies. [153] This led to a controversy about the existence of IPANs—a controversy that has been resolved only in the last decade.

Direct evidence that enteric responses can be initiated by axon reflexes comes from experiments in which reflexes were examined before and after degenerative section of the mesenteric nerve supplying the rat colon. [154] Reflexes initiated by muscle stretch were completely abolished after degenerative section of extrinsic nerves, but reflexes initiated by mechanical stimulation of the mucosa were unaffected. These findings imply that primary afferent fibers for mucosal mechanoreception have cell bodies in the wall of the colon, whereas primary afferent neurons for stretch reflexes in the colon have cell bodies in the dorsal root ganglia. Contrasting results were observed in the guinea pig small intestine. [155] In this case, reflexes initiated by mucosal distortion, and those initiated by radial stretch, were both unaffected by neural isolation of a segment of intestine. Thus, both reflexes depend on IPANs. Similar results have been reported for guinea pig colon. [156]

IPANs have been directly demonstrated by experiments in which activity-dependent changes were used in the labeling, [157], [158] and by intracellular recording. [73], [159] The experiments indicate that the cell bodies of the mucosal mechanoreceptor primary afferent neurons are in the submucosal ganglia, the cell bodies of chemoreceptor afferent neurons are in myenteric ganglia, and that the cell bodies of stretch-responsive primary afferent neurons are also in myenteric ganglia (see Chapter 3). Physiological studies have identified the primary afferent neurons as neurons with AH electrophysiological properties and Dogiel type II morphology in the small intestine of the guinea pig. These findings agree with deductions made from studies of the morphology and projections of Dogiel type II neurons. [19], [160] Neurons with similar morphology, histochemistry, and projections have been found in the guinea pig colon and rat small intestine. [161], [162] and [163] Neurons with Dogiel type II morphology that project to the mucosa, presumed to be IPANs, have been described in the human small intestine. [164] In the pig, Dogiel type II neurons also project to the mucosa and provide terminals in the enteric ganglia [21]; they have some electrophysiological characteristics in common with IPANs in the guinea pig. [165] Unlike their counterparts in the guinea pig and rat, IPANs in the pig are not immunoreactive for calbindin, but they are immunoreactive for CGRP and cholinergic markers; they do not all have such a prominent late AHP as that seen in guinea pig IPANs.

IPANs in the guinea pig are immunoreactive for both tachykinins and choline acetyltransferase. [46], [166] Therefore, they might be expected to transmit using tachykinins and ACh as cotransmitters. They transmit to other IPANs and to S neurons through slow EPSPs [72] that are probably mediated, in part, by tachykinins. [78], [167] Less direct evidence indicates that the AH neurons probably also communicate with other myenteric nerve cells at greater circumferential distances through cholinergic fast EPSPs. [144]

EXTRINSIC CONNECTIONS

Sympathetic Neurons

Norepinephrine is the primary transmitter of the sympathetic postganglionic neurons that supply the gastrointestinal tract. [3] Their cell bodies are in prevertebral and paravertebral ganglia, and their axons run to the gut with the mesenteric nerves. The major roles of these neurons are to regulate blood flow to the gastrointestinal tract, to regulate intestinal fluid and electrolyte secretion to keep it in balance with whole body requirements, and to inhibit gastrointestinal motility (see section "Sympathetic Effects on Motility and Secretion").

Neurons of Vagal and Pelvic Motor Pathways

The vagus nerves contain the axons of neurons whose cell bodies lie within the brainstem. A variety of effects, primarily on the upper gastrointestinal tract, are mediated through these neurons, including control of esophageal peristalsis, relaxation of the proximal stomach, enhancement of gastric peristalsis, stimulation of gastric secretion of acid, promotion of gastrin secretion, stimulation of pancreatic secretion, and modification of gallbladder function. In each case, the vagal neuron does not act directly; it forms synaptic connections with enteric neurons. Vagal efferent neurons also project to the small intestine and proximal colon. It is interesting that Brunner glands in the small intestine appear to be innervated directly by vagal pathways, but not by local intrinsic neurons. [168]

According to the convention used for other cranial autonomic pathways, the vagal neurons of these motor pathways have been called *parasympathetic preganglionic* or *vagal preganglionic neurons*. Reference to vagal input neurons as preganglionic wrongly implies that enteric neurons are relay neurons in parasympathetic pathways. Vagal input neurons are neurons in complex circuits in which enteric reflexes of several types are integrated with signals from the central nervous system and from other parts of the gastrointestinal tract. Most enteric neurons do not receive direct vagal connections. Thus, it is wrong to use the term *parasympathetic* to describe enteric neurons and enteric ganglia.

Transmission from vagal input neurons to enteric neurons is mediated principally by ACh acting on nicotinic receptor; the effects of stimulating vagal motor pathways are blocked or substantially attenuated by drugs that block nicotinic receptors.

The primary gastrointestinal effects of the pelvic nerves are on movement, secretion, and blood flow in the distal colon and rectum. The pathways are analogous to those of the vagus; pelvic efferent neurons with cell bodies in the sacral spinal cord form synapses on enteric neurons at which ACh is an excitatory transmitter acting through nicotinic receptors.

Extrinsic Primary Afferent Neurons

The extrinsic primary afferent neurons are discussed in Chapter 3. These neurons are subdivided into two groups: those with cell bodies in the vagal (nodose) ganglia, and those with cell bodies in the dorsal root ganglia. In general, the vagal afferent pathways carry information about the physiological state of the digestive organs. Impulses conveying pain or discomfort are conducted through the dorsal root ganglion (spinal afferent) pathways.

ENTERIC PATHWAYS FOR MOTILITY CONTROL

The circuits involved in local peristaltic reflexes in the small intestine, and the projections of the component neurons, have been deduced from studies of guinea pig small intestine (Fig. 2-9 and Fig. 2-10; see Fig. 2-7). The general features of these reflex pathways appear to be similar in the guinea pig large intestine and in the

small and large intestines of other species. Less is known about the circuits in the stomach, although propulsive reflexes do occur in this organ. [169] In all species, excitatory and inhibitory motor neurons supply the muscle; the general patterns of small intestine motility are similar (see Chapter 11), and intrinsic reflexes are evoked by the bulk and chemical nature of the lumenal contents.

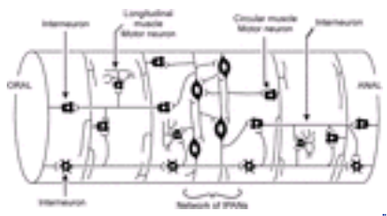

FIGURE 2-9. Pathways for propulsive reflexes in the intestine. A short segment of intestine is represented, on which the first parts of the descending inhibitory and the ascending excitatory reflex pathways are depicted. The patterns in the circuitry for the ascending and descending pathways are similar. Intrinsic primary afferent (sensory) neurons (IPANs) are circumferentially oriented and form self-reinforcing networks. They provide outputs to ascending and descending interneurons and monosynaptic connections to motor neurons. The interneurons form descending and ascending chains and also provide outputs to motor neurons. In the descending pathway, some neurons excite the longitudinal muscle, and some neurons inhibit the circular muscle. Ascending reflex pathways supply inputs to excitatory longitudinal muscle motor neurons and excitatory circular muscle motor neurons. A set of descending interneurons (**bottom part of diagram**) involved in conducting the migrating myoelectric complex receives very little input from IPANs but connects with motor neurons.

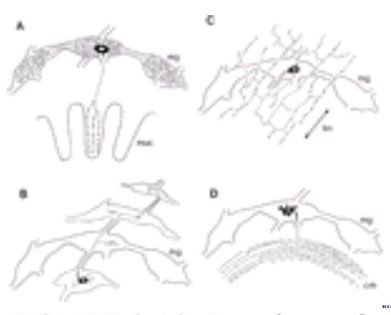

FIGURE 2-10. Drawings of enteric (**A**) intrinsic primary afferent neurons (IPANs), (**B**) interneurons, and motor neurons to (**C**) longitudinal muscle and (lm) (**D**) circular muscle (*cm*). These drawings are derived from studies of the guinea pig small intestine. The morphologies of the IPAN, the interneuron, and the motor neuron innervating longitudinal muscle are determined primarily by filling neurons with intracellular marker dye. The morphology of the motor neuron innervating circular muscle is determined by an evaluation of dye filling, in addition to histochemical and lesion studies. *mg,* myenteric ganglia; *muc,* mucosa.

The movements of the intestine are a result of the contractions and relaxations of the external longitudinal and circular muscles and of the muscularis mucosae. The neural control is superimposed on an underlying rhythm of muscle activity (i.e., slow waves), which occurs at frequencies of about 3 to 12 per minute in humans (see Chapter 6). [170] The major role in forming the pattern of mixing and propulsive movements appears to be taken by the circular layer of the external muscle, areas of which contract (or relax) like an annulus to close or open the lumen partly during both propulsive and mixing movements.

To study enteric muscle motor reflexes, investigators isolate segments of intestine to eliminate the influences of the central nervous system and circulating hormones. This simplifies recording from the muscle and enteric neurons. In these preparations, lumenal distention, or chemical or mechanical stimulation of the mucosa, elicits reflexes whose effects on the circular muscle can be recorded with intracellular microelectrodes. [85], [86], [147], [171], [172] and [173] The major responses of the muscle are depolarizing potentials (i.e., excitation) in the circular layer orad to the site of the stimulus and aborad hyperpolarizing potentials (i.e., inhibition). This polarization of electrical responses is analogous to that of the mechanical events seen when the intestine is distended. Longitudinal muscle motor neurons are also activated by reflex stimuli. [84] The longitudinal muscle contracts at the same time as the circular muscle, although there may be delay between the commencement of contraction in the two layers. [151], [174], [175] Evidence suggests that longitudinal muscle contraction in the colon may in effect pull the intestine over a solid bolus. [175]

Neither the reflex responses to stimulation of the mucosa nor those to distention pass along the gut if the myenteric plexus is cut, but they are unaffected by interruption of the submucosal plexus. [147], [171], [176] Therefore, the reflex pathways excited by mucosal stimulation must pass locally from the sensory receptors in the mucosa to the myenteric plexus. Responses evoked by distention persist if the mucosa is removed, a finding that implies the existence of sensory receptors in the external muscle. [147], [177], [178] These receptors have now been directly demonstrated (see Chapter 3). Mucosal distortion releases 5-HT from enterochromaffin cells in the epithelial lining, [179] which, in turn, activates the endings of the IPANs to initiate peristaltic reflexes. [179], [180] and [181]

In the guinea pig small intestine, IPANs with cell bodies in the myenteric plexus provide extensive varicose networks in the ganglia, primarily close to and surrounding the cell bodies. [19], [38] A minority of the neurons have aborally projecting collateral axons. [182] Ultrastructural studies show that the varicosities of IPAN terminals provide synaptic inputs to the majority of neurons in the myenteric plexus. [183] Electrophysiological studies of the effects of the activation of IPANs through their nerve endings in the mucosa confirm that they communicate with other neurons by means of slow EPSPs, [51], [73] whereas recording at a distance circumferentially implies that some synapses from IPANs cause fast EPSPs. [144] Slow EPSPs are evoked consistently only at rates of electrical stimulation of presynaptic neurons of about 3 Hz or more. In accordance with this finding, the primary afferent neurons fire in response to physiological stimuli in bursts that commonly achieve rates of 10 to 40 Hz. [51], [73]

It is notable that physiological as well as ultrastructural [183] studies indicate that IPANs synapse with other IPANs to form a self-reinforcing network (see Chapter 3). IPANs are numerous, about 500 per millimeter-length of the small intestine. Because the stimuli giving rise to intestinal reflexes (e.g., lumenal chemicals, distention, mucosal distortion) are not spatially confined to submillimeter distances, it can be deduced that reflexes are usually initiated by the more-or-less synchronous activation of a population of several hundred interconnected primary afferent neurons. The summation of the synaptic events caused by the transmissions from the primary afferent neurons results in the nearly simultaneous activation of numerous interneurons and motor neurons. The IPANs in the submucosal plexus project to the myenteric plexus and appear to be directly sensitive to mechanical and chemical stimulation of the mucosa. At least some portion of the reflexes evoked by stimulation of the mucosa is mediated by these neurons.

Enteric reflexes often extend for several centimeters along the intestine from a single point of stimulus, unlike the processes of the majority of IPANs and of motor neurons. This implies that most enteric reflex pathways in the guinea pig include interneurons, which, as previously explained, form interconnecting chains that run along the intestine, and which may individually run for more than a centimeter. [184] The pathway lengths may be greater in other species. Immunohistochemical data indicate that, in the guinea pig small intestine, there is one population of orally directed interneurons, which are cholinergic; and several populations of aborally directed interneurons, many of which are immunoreactive for choline acetyltransferase (see Table 2-1 and Fig. 2-7). Intracellular recordings made from enteric neurons during reflexes evoked by distention and mucosal distortion have generated morphologic data (Fig. 2-11). [83], [84], [85] and [86] Fast EPSPs, which are the output events of interneurons, are seen in neurons at distances of 0.5 to 2.5 cm from the stimuli (see Fig. 2-11). Physiological and structural studies indicate that interneurons make connections with both motor neurons and other interneurons, confirming the pattern of connectivity shown in Figure 2-9. The circuits in Figure 2-9 depict individual neurons, although cohorts of neurons are actually activated. [185]

The projections of motor neurons to the circular muscle have been determined by mapping the responses in the muscle to transmural nerve stimulation in normal pieces of intestine and in pieces in which the nerve paths were lesioned and allowed to degenerate, [186], [187] by microscopic analysis of the effects of nerve lesions, [3] and by the observation of retrograde transport of microcrystals of dye placed on the inner surface of the muscle. [110] Most excitatory and inhibitory motor neurons supplying the circular muscle extend only 1 to 2 mm along the guinea pig intestine, but they run up to half the distance around its circumference, which is about 6 to 8 mm in the guinea pig small intestine (see Fig. 2-10). Thus, the response to reflex activation is spread around the intestine. This finding is consistent with the common

observation that the intestine undergoes annular, not eccentric, contractions or relaxations. The reflex probably involves a summed response to transmission from many motor neurons; Bornstein and colleagues [186], [187] estimated that each smooth muscle cell is influenced by approximately 25 inhibitory motor neurons, and a similar convergence of excitatory influence can be expected. Furthermore, the electrical communication between smooth muscle cells ensures a summation of the effects of the population of motor neurons. Some excitatory motor neurons project in an oral direction for up to 10 mm, and some inhibitory motor neurons project aborad for as much as 30 mm.

In addition to the simple reflex whose neural circuits have now been deduced, the intestine exhibits mixing movements and cyclic changes of activity, collectively called *migrating myoelectric complexes* (MMCs). The MMCs pass along the intestine from the stomach to the terminal ileum. In humans, these cycles last about 90 minutes and are seen between digestive periods. The myoelectric complex is generated by the enteric nervous system. [3] During digestion, the contents of the intestine trigger irregular mixing contractions. The neural pathways depicted in Figure 2-9 presumably provide the building blocks for both the irregular contractile activity and the MMCs, with the irregular contractile activity perhaps representing the superimposition and interaction of simple reflexes. It has been deduced that the descending neuron chains for the MMC are separate from those of local reflexes (see section "Enteric Interneurons" and ref. [144]).

ENTERIC PATHWAYS FOR SECRETOMOTOR AND VASODILATOR CONTROL

Secretomotor and Vasomotor Reflexes Related to Fluid Exchange

Distention, mechanical stimulation of the mucosa, and the application of chemicals to the mucosa cause motility reflexes in the intestine and also evoke secretomotor and vasodilator reflexes. [126], [188], [189], [190] and [191] The secretomotor neurons stimulate the epithelial cells to pump Cl^- into the lumen, which takes with it counter ions, mostly Na^+, and water. It is likely that the same ion channels are responsible for HCO_3^- secretion, the amount secreted being dependent on the lumenal pH. [192] Histochemical studies in the guinea pig small intestine indicate that two of the motor neurons for secretion are also motor neurons for vasodilation (see Fig. 2-8); that is, these secretomotor neurons may cause a physiologically appropriate vasodilation, concomitant with secretion, through collaterals to submucosal arterioles. [131], [193], [194] and [195] Thus, there may not be separate reflexes for secretion and vasodilation, although no evidence suggests a third type of secretomotor neuron projects to the vasculature (see Fig. 2-8). In the small intestine, an important physiological stimulus for secretion appears to be the presence or active uptake of nutrients. Nutrients such as glucose, which are absorbed by a Na^+ cotransporter, draw in Na^+ along with Cl^- and water. At the same time, glucose or its uptake stimulates the enteric secretomotor reflex. [196] Enteric reflexes also cause HCO_3^- secretion in response to duodenal acidification, although other acid-sensitive mechanisms, including a neurally independent stimulation of prostaglandin production, also release HCO_3^-. [128] Secretomotor reflexes can also be initiated pathologically by toxins, such as cholera toxin or enterotoxins, in the lumen.

The enteric secretomotor/vasodilator circuits consist of IPANs with their endings in the mucosa and an integrating circuitry that feeds back to motor neurons with cell bodies in the submucosal ganglia (Fig. 2-12). In some cases, the reflex pathways involve the myenteric ganglia (e.g., cholera toxin–induced secretion [197]), whereas reflexes initiated by mechanical stimulation of the mucosa can be mediated entirely through the submucosal plexus. [126], [189] Pathways from the myenteric plexus also cause vasodilation. [198] There are two types of secretomotor neurons: cholinergic and noncholinergic. The noncholinergic neurons appear to mediate most of the local reflex response, using VIP, or a related peptide, as their primary transmitter. [126], [127], [132] In the guinea pig small intestine, there are two types of cholinergic neurons, those that also contain NPY (and other peptides) and those that contain calretinin. The ACh/calretinin neurons preferentially innervate the glands at the base of the mucosa and have collaterals to submucosal arterioles, whereas the ACh/NPY neurons do not appear to innervate the arterioles. The presence of three classes of secretomotor neurons, two of which also have vasodilator collaterals, may provide a mechanism to balance secretion and vasodilation appropriate to the digestive state (see Fig. 2-8 and Fig. 2-12). The control of secretion in the small intestine contributes to the regulation of whole-body water and electrolyte status. In an equilibrium state, the amount of fluid lost through the kidneys and through respiration and perspiration should be matched by absorption from the alimentary tract. If more fluid is absorbed with nutrients or across the gastric mucosa than is lost, some of the excess can be passed back under the control of secretomotor reflexes. Thus, the source of secreted fluid in the small intestine can be a mixture of serum electrolytes and locally absorbed electrolytes. We postulate that local computation of the need for vasodilation and local absorption to supply electrolytes for secretion determines the relative activation of vasodilator and nonvasodilator secretomotor neurons (see Fig. 2-12).

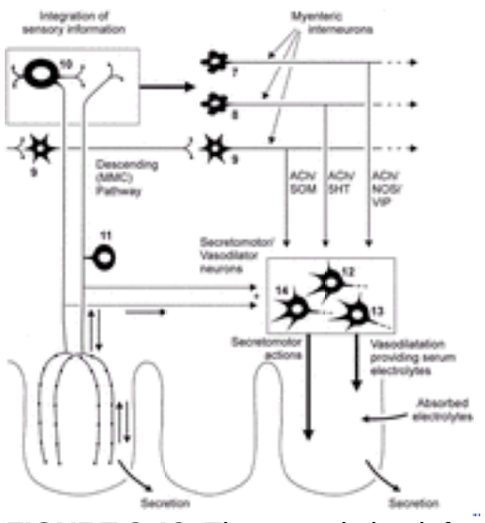

FIGURE 2-12. The neural circuit for secretomotor/vasodilator control in the guinea pig small intestine. Secretomotor responses are mediated both through antidromic action potential invasion of the mucosal endings of intrinsic sensory neurons, and through reflex activation of secretomotor and vasodilator neurons. Fluid absorbed across the mucosa during digestion, and fluid from the vasculature, whose supply is modified by vasodilation, are identified as sources of fluid for secretion. The numbers adjacent to the neurons correspond to the numbers in Table 2-1, which lists all the neuron types according to function and provides data on the percentage of the cell bodies of each type in the myenteric or the submucosal ganglia, in addition to information about their chemistries. *ACh,* acetylcholine; *MMC,* migrating myoelectric complex; *NOS,* nitric oxide synthase; *SOM,* somatostatin; *VIP,* vasoactive intestinal polypeptide.

In addition to reflexes that have secretomotor neurons as their outputs, there is evidence that IPANs may have direct secretomotor effects. The intrinsic sensory neurons are immunoreactive for tachykinins, and their varicose processes are immunoreactive for the vesicular ACh transporter. [166] Thus, their mucosal endings are likely to release ACh and tachykinins, both of which cause secretion. It has been directly shown that action potentials in one process of a sensory neuron traverse the cell body to invade other processes, [199] and the pattern of branching of the neurons indicates that action potentials could be conducted, as an axon reflex, between terminals that branch within the mucosa (see Fig. 2-8). Interestingly, the secretory responses to distention and to mucosal stroking in the guinea pig colon are reduced by tetrodotoxin (which blocks nerve conduction) and by atropine (which blocks the ACh receptors on the epithelium), but not by mecamylamine, an antagonist of cholinergic fast neuroneuronal transmission. [189], [191] The concentration of mecamylamine that was used was shown to block nicotinic receptors in the colon. [191] Moreover, the responses to stroking were not reduced by extrinsic denervation, a finding that indicates they are dependent on the activation of intrinsic neurons. [200] Thus, there is good evidence that ACh released by axon reflex, or by mononeuronal reflexes crossing the IPAN soma, contributes to secretory responses (see Fig. 2-8).

SYMPATHETIC EFFECTS ON MOTILITY AND SECRETION

Sympathetic neurons slow transit along the digestive tract, by constricting the sphincters and by inhibiting the contractile activity of the nonsphincter regions. [201] Inhibition of muscle movement in the nonsphincter parts of the gastrointestinal tract is primarily through actions of the noradrenergic axons in the myenteric ganglia, including the presynaptic inhibition of excitatory transmitter release. The nerve fibers also have direct inhibitory actions on the nonsphincter muscle. In the sphincter regions, the sympathetic neurons contract the muscle. Other sympathetic neurons innervate and inhibit submucosal secretomotor neurons. Another group of noradrenergic sympathetic neurons supply the gastrointestinal vasculature, constricting the arterioles and arteries and the major mesenteric veins. Their physiological

roles, which primarily involve adjustments of the proportion of the cardiac output going to digestive organs, are reviewed elsewhere. [132]

The existence of inhibitory reflexes whose pathways travel to the central nervous system and then back to the intestine through sympathetic ganglia was shown early in the 20th century. [3] Reflexes are conducted from one part of the gastrointestinal tract to another through sympathetic prevertebral ganglia, even when these ganglia are completely isolated from central nervous connections. [202], [203] These enteroenteric reflexes can be studied after the central pathways have been selectively inactivated, or in isolated preparations of intestine with ganglia attached.

Sympathetic enteroenteric inhibitory reflexes that decrease motility are initiated in one part of the gastrointestinal tract; they pass through prevertebral ganglia to other regions and also return to the first region through prevertebral ganglia. The peripheral enteroenteric reflex pathways and their associations with other sympathetic pathways are illustrated in Figure 2-13. Because it is not possible to survey all the evidence for the diagram in this chapter, only major points are discussed. Intestinofugal neurons that synapse in the prevertebral ganglia were demonstrated by intracellular recordings from the ganglia in preparations consisting only of a segment of intestine connected to a ganglion that had been completely removed from the body. [204] Activation of intestinal tension receptors evoked fast EPSPs in many nerve cells in the prevertebral ganglia. [204], [205] and [206] The EPSPs were blocked by the application of nicotinic antagonists to the ganglia, suggesting that they are cholinergic. This conclusion has been consolidated by the observation that all intestinofugal neurons are immunoreactive for choline acetyltransferase. [207] There is also a component of slow excitatory transmission from the intestinofugal neurons, which is a result of the release of a cotransmitter, possibly VIP. [202] Evidence that intestinofugal neurons are second-order neurons that nevertheless may also be directly activated by stretch is summarized in Chapter 3. The cell bodies of the intestinofugal neurons were observed in the myenteric ganglia after the injection of dye transported retrogradely into prevertebral ganglia. [208], [209], [210] and [211]

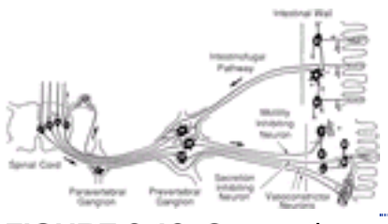

FIGURE 2-13. Connections of sympathetic pathways that affect intestinal motility, fluid exchange, and blood flow. Intestinofugal neurons, which have cell bodies in the wall of the intestine, are probably directly activated by sensory stimuli and indirectly activated by other enteric neurons. The intestinofugal neurons make excitatory synaptic connections with the cell bodies of motility-inhibiting and secretomotor-inhibiting neurons in the prevertebral ganglia. They do not connect with vasoconstrictor neurons.

Sympathetic motility-inhibiting neurons probably have little or no activity in undisturbed humans or animals. [3], [212] The enteroenteric inhibitory reflexes appear to have a protective role for the gastrointestinal tract. [3] For example, in the case of enterogastric reflexes, the slowing of gastric emptying protects the duodenal mucosa from acid and osmotic stress. Enteroenteric inhibitory reflexes affecting other parts of the intestine can be initiated by the distention of any region. Most studies have dealt with reflexes affecting the stomach and intestine, but similar reflex pathways affect the biliary system. [213] Inhibition of the stomach can also be caused by acidity or hypertonicity in the lumen of the upper small intestine. The reflexes that pass through the central nervous system are commonly evoked by noxious stimuli or by pain of visceral origin. For example, biliary colic, abdominal injury, and irritation within the abdominal cavity caused by chemicals or infection activate sympathetic reflexes that inhibit gut motility.

The net movement of water and electrolytes between the gut lumen and body fluid compartments is regulated to maintain overall fluid homeostasis. This is accomplished by sympathetic pathways that can override the enteric secretomotor reflexes (see Fig. 2-13). Axons arising from the sympathetic secretomotor-inhibiting neurons in prevertebral ganglia innervate secretomotor neurons, in which they elicit inhibitory synaptic potentials (see section “Synaptic Transmission in the Enteric Nervous System”). Persuasive evidence indicates that in day-to-day circumstances, the intrinsic secretomotor reflex activity is inhibited by tonic activity of the sympathetic secretomotor inhibitory pathways. Cutting the sympathetic pathways releases the brake on the enteric secretomotor reflex and results in what Bernard, in 1859, called *paralytic secretion*. [214] The level of activity of the sympathetic secretomotor inhibitory neurons responds to measures of whole-body water content and electrolyte status; thus, sympathetic activity is increased and water and electrolyte secretion is reduced in response to hemorrhagic hypotension, unloading of the baroreceptors, or reduction in right atrial pressure. [215], [216], [217] and [218] HCO_3^- secretion is also inhibited by sympathetic nerves. [219]

SUMMARY

Neural control of the gastrointestinal tract is hierarchical and somewhat complex. The neurons influence several aspects of digestive tract function, prominent among these being motility, secretion, and blood flow. At the organ level, control is exerted through local intramural reflexes whose component neurons are within the enteric nervous system. Regions of the gastrointestinal tract are linked through long intramural and extrinsic pathways; these include vagovagal reflexes and sympathetic enteroenteric reflexes. At the next hierarchical level, the gastrointestinal tract is controlled by activity in pathways originating in the central nervous system.

Acknowledgments

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CHAPTER 3

John B. Furness, Martin J. Stebbing, and Nadine Clerc

SENSORY NEURONS OF THE GASTROINTESTINAL TRACT

The sensory system of the gastrointestinal tract is complex. Detection of the state of the gastrointestinal tract involves three systems of sensors: primary afferent neurons, enteroendocrine cells, and immune cells (Fig. 3-1). Each of these detecting systems is more extensive than those of other organs. [1] About 20% of neurons in the enteric nervous system, which contains on the order of 10^8 neurons, are sensory, and more than 50,000 axons reaching the gut through the vagus and splanchnic nerves also are sensory. The gastroenteropancreatic endocrine system contains thousands of enteroendocrine cells, many of which react to their local environment, and from which more than 20 identified hormones are released. The gut immune system senses immunogens and contains 70% to 80% of the body's immune cells.

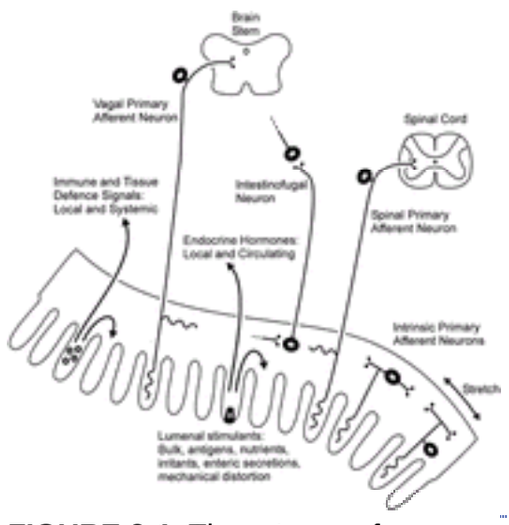

FIGURE 3-1. Three types of sensory signals originate from the gastrointestinal tract. Endocrine messages take the form of hormones released from cells in the mucosal epithelium. The hormones enter the circulation and can act at remote sites, but they also can act locally, on nerve endings, on the epithelium, and possibly on cells of the immune system. Immune messages are conveyed by circulating lymphocytes, which are activated if antigens are presented to them from the lumen or if the mucosal epithelium is breached, from their local tissue environment. Neural messages are conveyed by neurons whose sensitive endings are in the lamina propria, beneath the mucosal epithelium, in the muscle, and in enteric ganglia. Some neurons have cell bodies in the gut wall (IPANs and intestinofugal neurons), and others are in extrinsic ganglia (extrinsic primary afferent neurons; see Fig. 3-2).

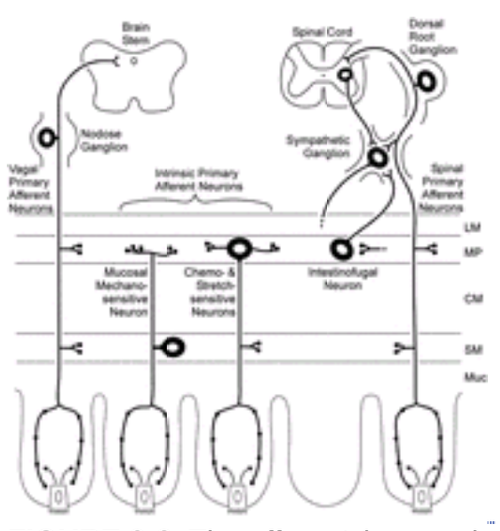

FIGURE 3-2. The afferent (sensory) neurons of the digestive tract. Three classes of *intrinsic* primary afferent neuron (IPAN) have been identified: tension-sensitive neurons with cell bodies in myenteric ganglia, chemosensitive neurons with myenteric cell bodies and processes that detect changes in lumenal chemistry, and mucosal mechanosensitive neurons with submucosal cell bodies and processes that detect mechanical distortion of the mucosa. The enteric afferent neurons are known to be modality selective; each also responds to other stimuli. *Extrinsic* afferent neurons have cell bodies in dorsal root ganglia (spinal afferent neurons) and vagal (nodose and jugular) ganglia. They can be subdivided into several classes (see text). Spinal afferent neurons supply collateral branches in prevertebral ganglia. Intestinofugal neurons are part of the afferent limbs of enteroenteric reflex pathways. Nerve endings in the mucosa can be activated by hormones released from enteroendocrine cells. *LM,* longitudinal muscle; *MP,* myenteric plexus; *CM,* circular muscle; *SM,* submucosa; *Muc,* mucosa.

The neurons that detect the states of tissues are known as *primary afferent neurons, primary* because they are the first neurons in reflex pathways, and *afferent* because they run toward reflex control centers. Three broad classes of afferent neurons are associated with the gut: intrinsic primary afferent neurons (IPANs), which have cell bodies, processes, and synaptic connections in the gut wall; extrinsic primary afferent neurons, which have cell bodies in vagal and dorsal root (spinal)

ganglia; and intestinofugal neurons, which have cell bodies in the gut but send processes to neurons outside the gut wall (Fig. 3-2). The monitoring and control of the digestive system by neurons is hierarchical. The gut contains an extensive collection of neurons—the enteric nervous system—within its walls (see Chapter 2). This intrinsic nervous system is capable of generating appropriate reflex responses to the contents of the intestinal lumen; for example, local reflexes generate mixing movements of the muscle, local changes in blood flow, and secretion of water and electrolytes. The enteric nervous system also participates in reflexes between organs—between the duodenum and stomach, for example, to regulate gastric emptying. Furthermore, the digestive organs send signals to the central nervous system: afferent signals for reflexes that act on the digestive system, signals that mediate coordination with other body systems, and signals that relate to sensations including discomfort, nausea, pain, and satiety.

INTRINSIC PRIMARY AFFERENT NEURONS

IPANs in the small intestine have been studied extensively, and those in the colon, to a limited extent. It is not clear whether neurons with the same function exist in the stomach. If they do, the morphology and cell physiology of most gastric IPANs are different from those of IPANs in the small intestine and colon. IPANs react to at least three types of stimuli: chemical changes in the intestinal lumen, distention of the intestine (even in the absence of the mucosa), and mechanical distortion of the mucosa.

Properties of IPANs

The IPANs identified to date are multiaxonal, with one or more axons that lead to and branch in the lamina propria of the mucosa, just beneath the absorptive epithelium, and axons that enter the myenteric ganglia and supply terminals around several types of nerve cells, including other IPANs, interneurons, and motor neurons. They have a distinctive shape known as *Dogiel type II morphology*. In addition, these neurons have distinct electrophysiological properties. They have broad action potentials that are carried by both Na^{+} and Ca^{2+} currents and are followed by early and late afterhyperpolarizing potentials (AHPs). Moreover, these neurons, unlike all other enteric neurons (at least in the guinea pig small intestine), do not receive prominent fast excitatory postsynaptic potentials (EPSPs). In other regions and species, fast EPSPs are more readily recorded from presumed IPANs. However, they do receive synaptic inputs through which slow excitation is mediated. This is unusual because neither spinal nor vagal afferent neurons receive synapses at their cell bodies. [2] Thus, IPANs differ from extrinsic sensory neurons in that their excitabilities can be modified by synapses at the cell body.

The electrophysiological properties of IPANs are influenced by the recording conditions. If the background synaptic transmission is suppressed, IPANs exhibit the late AHP that identifies them as AH neurons in the terminology of Hirst and colleagues, [3] but when IPANs are acted on by neurotransmitters or hormones, the late AHP can be suppressed or obliterated. [4], [5]

Two inward currents underlie the action potential in the soma of the IPAN, a tetrodotoxin (TTX)-sensitive Na^{+} current (gNa_V) and a TTX-insensitive Ca^{2+} current (gCa_V). [6], [7] In the presence of TTX, the Ca^{2+} current is still sufficient for action potential generation in the soma, but active conduction of action potentials in the processes of the Dogiel type II cells is blocked. The calcium channels are primarily N-type. There is also evidence of a TTX-resistant gNa_V, [8] but its physiological significance is not yet known.

The early AHP is continuous with the falling phase of the action potential and lasts about 50 to 100 milliseconds. The currents of the early AHP have been investigated in some detail. [9] This is a post-spike hyperpolarization similar to that observed in neurons of many types, in the central nervous system and in autonomic ganglia. The major part of the current is formed by the opening of large conductance (BK) channels. [10]

The late AHP, which is characteristic of Dogiel type II neurons when they are in their basal state, has an onset latency of about 70 milliseconds following a single action potential and can last from about 2 up to about 30 seconds. [3], [11] Two conductances increase during the late AHP: an outward Ca^{2+}-dependent K^{+} conductance, gK_{Ca} (an intermediate conductance, IK channel [10]), which exists in all Dogiel type II neurons, and an inwardly rectifying (i.e., depolarizing) cation conductance, g(K,Na), which generates the I_H current and exists in about 80% of these neurons. [12]

Chemosensitive IPANs

Intracellular records taken from nerve cell bodies in the guinea pig small intestine have identified a class of intrinsic neuron that responds to the application of chemicals, such as inorganic acid and short-chain fatty acids at neutral pH, to the lumenal surface of the mucosa of the small intestine. [13], [14] Activation of submucosal and myenteric neurons by the application of glucose to the mucosa also has been reported. [15]

IPANs may indirectly detect changes in the chemical content of the gut lumen via the release of hormones from enteroendocrine cells. Indirect action is inferred because the mucosal epithelium separates the nerve endings from the lumenal environment. Substantial evidence suggests that 5-HT, a potent IPAN stimulant, acts as an intermediate in enteric reflexes. [16], [17] and [18] 5-HT is released when the mucosa is mechanically stimulated to elicit motility reflexes, and the reflex responses are antagonized by drugs that block 5-HT receptors. [16], [17] and [18] Furthermore, mechanical stimulation of the mucosa induces c-Fos expression in IPANs in submucosal ganglia, and this induction is blocked by 5-HT receptor antagonists. [19] Other hormones in gut endocrine cells, such as cholecystokinin (CCK) and motilin, are released by nutrients and act on neurons; however, their roles as reflex intermediaries are untested.

Stretch-sensitive IPANs

IPANs also respond to tension in the muscle and to direct distortion of their processes. [20], [21] The mechanism through which these neurons are activated has been deduced (Fig. -3). The neurons respond phasically at the beginning of a stretch or when their processes are directly deformed. [21] During maintained stretch, the muscle contracts more or less rhythmically, and IPANs continue to be activated. [20] The rate of discharge is proportional to the degree of distention. However, the discharge of action potentials is abolished if the muscle contraction is prevented by muscle relaxants, indicating that active tension in the muscle contributes to the excitation of the tension-sensitive IPANs (see Fig. 3-3). The involvement of the muscle is interesting because it has long been known that intestinal muscle cells are directly sensitive to stretch and respond to it by contracting. [22] This reaction of the smooth muscle may be integral to the response of IPANs during sustained stretch. The neurons themselves possess mechanosensitive ion channels, and when the processes of the neurons are distorted, they discharge action potentials. [21], [23]

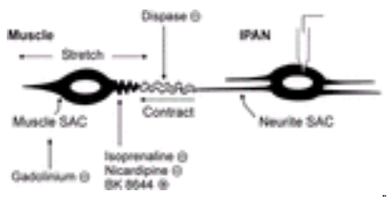

FIGURE 3-3. Mechanism of activation of stretch-sensitive intrinsic primary afferent neurons (IPANs). Stretch opens stretch-activated channels (SACs) in the muscle membrane, which results in muscle contraction. If the muscle SACs are blocked by gadolinium, or if contraction is prevented by isoprenaline or nicardipine, stretch-induced excitation of IPANs ceases. On the other hand, causing the muscle to contract by opening L-type Ca^{2+} channels with BK 8644 activates IPANs. The muscle pulls on IPAN processes through connective tissue, which can be weakened by dispase, preventing IPAN activation in response to stretch. The distortion of IPAN processes opens gadolinium-insensitive SACs, resulting in depolarization and action potential initiation.

Interestingly, the IPANs are inhibited if their cell bodies are distorted, or if a small area of their surface membrane is stretched. [21] This occurs through the opening of BK-type K^{+} channels on the cell soma, which appear to be directly stretch-sensitive. Myenteric nerve cell bodies are distorted by muscle movement, [24] and it is possible that sufficient pressure in the wall of the intestine may inhibit IPANs as part of a protective mechanism that limits the strength of reflex contraction of the

intestine.

Mucosal Mechanoreceptors

Functional evidence for IPANs with cell bodies in submucosal ganglia comes from experiments that have localized the activity-dependent induction of c-Fos and the activity-dependent uptake of styryl dyes. c-Fos immunoreactivity was detected in submucosal nerve cells after mucosal distortion by puffs of nitrogen gas ejected from a pipette. [19] Styryl dyes, which are taken up by active neuron endings and transported back to the cell bodies, also have been used to identify IPANs that are mucosal mechanoreceptors. [15] The results suggest that cell bodies of mucosal mechanoreceptors are in submucosal ganglia and project to the myenteric plexus (see Fig. 3-2). The activation of mucosal mechanoreceptors is likely to be partly indirect, through the release of 5-HT from enterochromaffin cells in the mucosa. [25]

Distention stimuli can activate both mucosal mechanoreceptors and distention-sensitive neurons (myenteric IPANs), explaining why both myenteric and submucosal neurons are revealed after distention. [15] The presence of stretch-sensitive neuron cell bodies in the submucosal ganglia, as well as in myenteric ganglia, has not yet been determined.

Possible Polymodal Nature of IPANs

It appears that intrinsic afferent neurons may be modality preferring, rather than modality specific. For example, primary afferent neurons in myenteric ganglia that respond to chemicals applied to the mucosa can also respond to mechanical stimulation of the mucosa, [14], [26] whereas under some circumstances, mechanical stimulation excites only neurons with cell bodies in submucosal ganglia. [15]

In the guinea pig small intestine, all myenteric neurons with the IPAN morphology project to the mucosa. [27], [28] It is impossible to say whether all these IPANs respond to chemical stimulation from the lumen because to record from them part of the mucosa must be removed. [14] About 50% of the neurons responded to electrical stimulation of the remaining intact mucosa. Thus, it is possible that only 50% of IPANs had intact mucosally projecting axons in the test regions. Of the neurons that responded to focal electrical stimulation of the mucosal surface, 60% responded to chemicals (acid, base, or fatty acid) applied to the mucosa. [14] Thus, at least 60% of myenteric IPANs are chemoreceptive. This may be an underestimate of this population because it is not possible to be certain that the appropriate chemical stimulus reached each nerve ending. In tissue that was stretched to excite stretch-activated IPANs with cell bodies in the myenteric plexus, about 80% of neurons responded to a 40% increase in intestinal circumference. [20] About 70% of these stretch-sensitive neurons responded directly. Thus, the data indicate that some IPANs are activated by more than one sensory stimulus.

Assemblies of IPANs

Physiological studies, [29] as well as ultrastructural studies, [30] indicate that IPANs synapse with other IPANs, thus forming a self-reinforcing network. [14], [31] The neurons are very frequent, about 500 per millimeter length of small intestine. Furthermore, they overlap considerably in their receptive fields; retrograde tracing indicates that each villus is supplied by the axons of 65 IPANs (on average) with cell bodies in the myenteric ganglia. [28] Because the stimuli that give rise to intestinal reflexes (i.e., lumenal chemicals, distention, mucosal distortion) are not spatially confined to submillimeter distances, it can be deduced that reflexes usually are initiated by the more or less synchronous activation of a population of several hundred interconnected primary afferent neurons.

Roles of IPANs

Intrinsic reflexes that affect motility, water and electrolyte secretion, and blood flow all occur in the small intestine. (For details of the reflex circuits, see Chapter 2.) Each of these reflexes is evoked by similar stimuli, although it is not known whether the same, different, or overlapping populations of IPANs contribute to motility, secretomotor, and vasomotor reflexes.

Muscle motor reflexes have been studied as stereotypical responses of the circular muscle; that is, excitation oral and relaxation aboral can be evoked by distention of the muscle (effected without distorting the mucosa), by the application of chemicals to the mucosal surface, and by distortion of the mucosa. [32], [33] and [34] These reflexes occur after the degeneration of extrinsic afferent nerve endings and are therefore the consequences of IPAN activation by sensory stimuli (see Chapter 2).

Secretomotor reflexes are initiated physiologically by chemical or mechanical interaction of lumenal contents with the mucosa, or pathologically by toxins, such as cholera toxin or enterotoxins, in the lumen. [35], [36] Enteric reflexes also cause HCO_3^- secretion in response to duodenal acidification. [37] The enteric secretomotor circuits consist of IPANs with their endings in the mucosa and nerve circuits that pass through the myenteric and submucosal plexuses and feed back to secretomotor neurons with cell bodies in the submucosal ganglia. [35], [36], [38], [39] The secretomotor neurons stimulate the epithelial cells to pump Cl^- ions, which are accompanied by water, into the lumen.

Local vasodilator reflexes in the small intestine are caused by mechanical or chemical irritation of the mucosa, and substantial evidence indicates that the vasodilator neurons are intrinsic to the intestine and transmission from them is predominantly noncholinergic. [40], [41] It is presumed that the first neurons in these reflexes are the IPANs, but this has not been directly shown. In fact, of the reflexes in the intestine, the vasomotor reflexes are the least studied. The same motor neurons have axons that branch to supply both the secretory epithelium and arterioles; thus, some secretomotor and vasodilator reflexes may share the same final neurons (see Chapter 2). This makes physiological sense because the secreted water and electrolyte come indirectly from the vasculature.

The roles of IPANs are to signal changes in the state of the intestine that are the consequences of the presence and nature of its contents. The information is conveyed to other neurons of the enteric nervous system that integrate the information and cause appropriate changes in mixing and propulsive activity, in water and electrolyte transport, and in local blood flow.

EXTRINSIC PRIMARY AFFERENT NEURONS

Extrinsic primary afferent neurons have cell bodies in the nodose and jugular ganglia, from which axons follow vagal pathways to innervate the gut (vagal primary afferent neurons), or in dorsal root ganglia (spinal primary afferent neurons). The fibers of spinal primary afferent neurons in the thoracic and lumbar regions pass through the sympathetic ganglia to reach the gut via the splanchnic and mesenteric nerves. Some fibers originating from thoracic dorsal root ganglia join the vagus nerve, either through the stellate ganglion or directly via small connecting nerves in the thorax. Conversely, many axons of vagal afferent neurons supplying the small intestine are in the celiac branch of the vagus and join with spinal afferent fibers in the mesenteric nerves as they run to the gut. The axons of spinal primary afferent neurons with cell bodies in the sacral ganglia follow the pelvic nerves to reach the colon and rectum.

The types of information carried to the central nervous system by extrinsic afferent neurons are summarized in Table 3-1. Very broadly, the afferent information can be divided into *that which reaches conscious sensation,* such as sensations of gastric or intestinal fullness, warmth, various types of pain (cramp, colicky pain, sharp pain), discomfort, nausea, hunger, and satiety, and *that which is not perceived,* such as the chemical nature of the contents of the intestines and the mechanical activity of the gut during normal digestion, which initiate responses such as gastric acid secretion, pancreatic secretion, and gastric and intestinal motor activity.

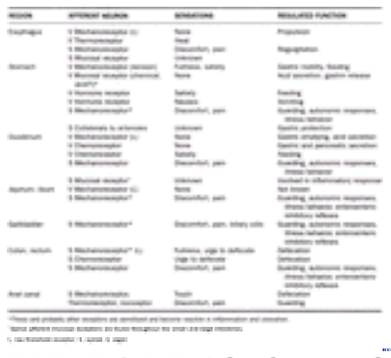

TABLE 3-1 Extrinsic Vagal and Spinal Gastrointestinal Afferent Neurons: Simplified Classification of Neurons, Sensations, and Regulated Functions

The level of specificity and detail of the afferent information that reaches the central nervous system is not known. The central nervous system may be informed of the general state of the digestive organs by the summation of afferent signals. This is suggested by the substantial sizes of the receptive fields of the afferent neurons.[42] It is also suggested by the responses of the organs—for example, acid or enzyme secretion, gallbladder contraction, or gastric relaxation, none of which appear to be finely graded. Moreover, hormones such as CCK that diffuse locally and via the bloodstream act on many afferent nerve endings. Consciously perceived sensations that are conducted by extrinsic primary afferent neurons also are generalized and usually difficult to locate to any particular abdominal organ. Satiety is indirectly perceived; for example, we feel sated after a meal, but the feeling of satiety is not directed to a particular organ.

Conscious perception of the state of the gastrointestinal tract is subject to psychosensory modulation.[43],[44] and[45] Perception is enhanced when attention is paid to the gut and is diminished with inattention. Sensation also is diminished when a painless somatic stimulus is simultaneously applied.[43] A classification of extrinsic primary afferent neurons by the stimuli to which they respond, by the sensations to which they give rise, and by the functions they regulate appears in Table 3-1.

Structural Studies

Large numbers of peripheral axons of extrinsic primary afferent neurons enter the gut. In animals such as cat and rabbit, the abdominal vagus contains about 20,000 to 30,000 primary afferent nerve fibers; the human abdominal vagus contains about 40,000 to 50,000.[46] Spinal afferent fibers also are plentiful; in the greater splanchnic nerve of the cat, there are about 3000 to 4000; in the lumbar splanchnic nerves, about 4000 to 5000; and in the pelvic nerves, about 3500.[46] Almost all the afferent fibers entering the gut are unmyelinated and soon become mixed with the processes of enteric neurons, from which they are indistinguishable by normal light and electron microscopy, with the exception of the intraganglionic laminar endings.

Three types of vagal afferent endings have been identified, mostly by anterograde tracing studies: intramuscular arrays,[47],[48] branching varicose axon endings that innervate the mucosa,[49],[50] and intraganglionic laminar endings (IGLEs).

An intramuscular array consists of an axon that supplies branches, sometimes ten or more, several millimeters long and running parallel to the longitudinal or circular muscle. They are most numerous in the stomach but are found throughout the gastrointestinal tract, from the smooth muscle part of the esophagus to the distal colon, where they are relatively rare.[51] The morphology of intramuscular arrays suggests that they are in-parallel length receptors, even though most physiological evidence suggests that vagal mechanoreceptors in the stomach are tension receptors.[52]

IGLEs were illustrated in 1929 by Lawrentjew,[53] who showed, by their degeneration after vagal nerve section, that they are of vagal origin. Subsequently, their afferent nature was demonstrated when they were found to degenerate after extirpation of nodose ganglion cells, but not after section of the vagus nerve central to the nodose ganglion.[54] The IGLE consists of numerous lamellae, generally about 2 to 5 μm across, that interconnect with each other and form a discontinuous covering of myenteric neurons. In the rat stomach, about six neurons are covered by each IGLE.[47] Experiments have confirmed that IGLEs are mechanoreceptors.[55] IGLEs are very common in the esophageal, gastric, and duodenal myenteric ganglia, and occur at lesser densities in the rest of the digestive tract. In rat, they have been reported to occur at a density of 6.3/mm ^{2} in the gastric corpus, 3.3/mm ^{2} in the duodenum, 0.6/mm ^{2} in the jejunum and ileum, and 0.2/mm ^{2} in the distal colon.[51] The proportion of ganglia innervated also decreases along the digestive tract. After labeling by left vagal ganglion injection, IGLEs were found to supply 88% of ganglia in the esophagus and 31% in the corpus; the proportion decreased to 1% in the distal colon.[56],[57]

The mucosal vagal afferent endings consist of branching varicose fibers that come close up against the mucosal epithelium, and, in the esophagus, may enter the epithelium.[49],[50],[58],[59] Morphologically, these fibers seem indistinguishable from the motor fibers and intrinsic afferent endings that supply the epithelium.

Less attention has been devoted to determining the distributions of spinal afferent neurons with the use of neuronal tracers. In some species, however, a high proportion of spinal afferent neurons contain neuropeptides, particularly tachykinins and calcitonin gene–related peptide (CGRP).[60] Where spinal nerve endings containing these peptides can be distinguished from nerve fibers of intrinsic origin, the peptides have proved to be useful markers. The axons of spinal afferent neurons provide a sparse network of varicose axons in the myenteric ganglia of the esophagus,[61] stomach[60],[62] and small intestine.[63] They are prominent around arterioles in the gut wall,[60],[61],[64],[65] and they also branch within the lamina propria of the mucosa throughout the gastrointestinal tract. Rare fibers are found in the muscle layers.[60],[62],[66] In the cat esophagus, they penetrate deep into the squamous epithelium.[61]

Functional Subtypes Identified by Location of Afferent Endings

Extrinsic primary afferents have been classified as mucosal receptors, muscle receptors, and serosal receptors on the basis of electrophysiological studies of the response patterns to various mechanical and chemical stimuli. Location also discriminates receptors; whether a location is superficial (in the mucosa or the serosa) or deep (in the muscle or myenteric ganglia) can be determined by testing the effect of removing or anesthetizing the mucosa or serosa. Electrophysiological studies, with the use of single-unit recording, imply that most vagal primary afferent fibers innervate the mucosa and muscle layers, whereas the spinal afferent fibers innervate the muscle layers and serosa, but not the mucosa.[67],[68] and[69] However, morphologic data show that the axons of spinal primary afferent neurons innervate the mucosa of the esophagus, stomach, and intestines. These may have nonmechanoreceptor functions.

Muscle Receptors Muscle receptors, found throughout the gastrointestinal tract, are sensitive to variations in muscle tension.[46] Increased discharge when the muscle contracts (i.e., when tension is increased but length is decreased) implies that that they are tension receptors.[70] A carefully controlled study of vagal mechanoreceptor discharge over a range of different gastric volumes and wall tensions confirmed the original designation of these afferents as tension receptors.[71] Muscle receptors exhibit a dynamic response that adapts slowly. Most vagal mechanoreceptors that innervate the gastric muscle fire at low frequency, even when the stomach is empty. Thresholds for their activation vary, but all are in the normal (i.e., nonpathological) range of pressures.[72],[73] Spinal afferent neurons include low- and high-threshold muscle receptors and silent nociceptors.[74],[75] and[76] Silent nociceptors are neurons whose endings are not activated by mechanical forces, except when the tissue they innervate is injured or inflamed. It has been persuasively argued that the term “silent nociceptor” is likely to be a misnomer for most fibers so identified because these fibers, unresponsive to intense mechanical stimuli, may be responsive to other (untested) stimuli; furthermore, not all mechanically insensitive neurons are nociceptors.[77] High-threshold muscle receptors of spinal primary afferent neurons that are sensitive to ischemia are likely to be nociceptors.[74],[75] Spinal mechanoreceptive afferent neurons are generally polymodal. Afferent neurons that respond to colonic distention also respond to bile salts, inflammatory mediators (a mixture of bradykinin, prostaglandin, 5-HT, and histamine), and temperature.[78] These neurons may in fact be mucosal receptors or have collaterals in the mucosa.

Mucosal Receptors Mucosal receptors are found all along the gut. They respond vigorously to mucosal deformation but are likely to be polymodal—that is, able to respond to two or more stimuli, including mechanical deformation (stroking), lumenal chemicals (acid or nutrients), heat or cold, and in some cases increased lumenal osmolarity.[46],[79],[80],[81],[82] and[83] In contrast, feline studies suggest that some mucosal receptors in the esophagus and stomach are selective for temperature,[84] and that others in the small intestine are selective for nutrients, such as amino acids[85] and lipids.[86] This apparent selectivity, however, may have been a consequence of the narrow range of stimuli being tested.[46],[69] A study of sheep demonstrated that duodenal mucosal endings that respond to amino acids are also mechanoreceptors.[81] Nerve fiber recording did not reveal mucosal receptors sensitive to carbohydrates in rat,[87] rabbit,[88] or ferret[83] small intestine. Nevertheless, infusions of glucose, maltose, and mannose into the duodenum evoke reflex responses in rat stomach and pancreas via the activation of vagal afferents,[89],[90]

implying that carbohydrate-sensitive endings are indeed present. Both slow and rapid adaptation to mucosal mechanical stimuli are encountered. [46], [81] Responses to lumenal chemicals generally are slowly adapting. [85], [86] Although present in the stomach and esophagus, thermal receptors appear to be absent from the duodenum. [81]

Serosal Receptors Serosal receptors seem to be activated by high-threshold mechanical stimuli, such as bowel movement, distention, and squeezing, although gentle probing, light squeezing, or puffs of air also elicit responses. [46] Serosal receptors are also sensitive to ischemia. The serosa is continuous with the mesentery and the parietal peritoneum, where receptors with similar properties reside.

Indirect Activation of Mucosal Receptors by Enteroendocrine Cell Products

The stimulatory effects of nutrients on mucosal afferent endings in the small intestine are indirect, which is in accord with the separation of lumenal chemicals from mucosal nerve endings by the intestinal epithelium. The alternative method of activation, directly after passage across the mucosa, probably also occurs (e.g., diffusion of inorganic acid). CCK and 5-HT, which are released in the mucosa, activate the mucosal receptors but not the muscle receptors of vagal primary afferent neurons. [83], [91], [92], [93] and [94] A physiological role for CCK released from enteroendocrine cells and acting on vagal afferent endings has been demonstrated; the activation of vagal primary afferent neuron endings by lipid or protein in the duodenum is blocked by CCK_1 receptor antagonists. [95], [96] 5-HT appears to be an intermediate for the excitation of vagal primary afferent neurons by intralumenal hyperosmotic or hypoosmotic solution in the duodenum and to high concentrations (>275 mM) of the monosaccharides glucose and mannose. [90] Increased discharge of vagal afferent neurons caused by these agents was blocked by the intravenous infusion of the $5\text{-HT}_{3/4}$ antagonist tropisetron, and by the 5-HT_3 antagonist granisetron. In contrast, activation of afferent fibers by duodenal distention was not affected. The intraduodenal chemical stimuli did not elevate plasma 5-HT, implying that the site of action of 5-HT was in the mucosa near its points of release. The activation of afferents mediating vagovagal pancreatic reflexes depends on the release of both CCK and 5-HT from enteroendocrine cells. [97]

Lumenal fat is a stimulus for reflexes from the distal to more proximal ileum, the ileal brake. [98] Release of peptide YY from mucosal enteroendocrine cells of the mid nd distal small intestine and colon leads to slowing of small intestinal transit. Although the neural pathway of this effect is not known, it appears to be a neural reflex. [99]

Efferent Effects of Primary Afferent Neurons

Ultrastructural studies show that afferent fibers contain numerous synaptic vesicles, similar to the endings of unmyelinated motor neurons or interneurons. [58], [100] The axolemma of these fibers is sometimes naked, which suggests that neurotransmitters released by afferent fibers have good access to effector cells. Myenteric neurons are a target because frequent appositions of naked fibers arising from spinal primary afferent neurons occur on the cell bodies. [100] The afferent nerve endings contain bioactive peptides, including CGRP and tachykinins. [60], [63]

Several pharmacological and physiological studies have revealed muscle contraction in response to antidromic stimulation of spinal afferent nerve fibers, [101], [102] and one study has implicated axon reflexes in initiation of the peristaltic reflex (see Chapter 2). Neuropeptides released from primary afferent nerve endings cause vasodilation [103], [104] and [105] and stimulate myenteric neurons [106] and mast cells. [107]

The effects of antidromic release of transmitter from afferent nerve endings on gastric and duodenal motility are most likely of trivial physiological importance. [108] By contrast, vasodilation caused by transmitter released from the peripheral processes of afferent neurons has an important role in mucosal protection (see section "Tissue Protection"). There is also evidence that transmitter released from the mucosal sensory endings of IPANs contributes to the regulation of fluid secretion. [109]

Roles of Extrinsic Primary Afferent Neurons

Extrinsic primary afferent neurons signal to the central nervous system information necessary to regulate organs and behaviors that are beyond the immediate local territories of the afferent endings. The signals that reach the central nervous system convey information about several qualities of the gastrointestinal tract, including the state of distention, the presence of chemicals in the lumen (e.g., nutrients, irritants, and toxins), and the presence of tissue injury and inflammation. The signals can be decoded and interpreted consciously as satiety, pain, or hunger. They can also be used to direct digestive functions automatically; for example, esophageal propulsion, gastric relaxation in response to a meal, and gastric acid secretion may all be controlled through the vagus, defecation via the pelvic nerves, and the control of water and electrolyte transport and blood flow in relation to the relative needs of all organs via sympathetic motor pathways.

Pain and Discomfort For a long period, there was considerable doubt regarding whether the gastrointestinal tract was innervated by pain fibers because clearly injurious insults to the stomach, small intestine, and most of the large intestine are not perceived. [46] On the other hand, there is conscious sensation from the esophagus (e.g., hot food or drink) and from the anal canal. Pain from the abdomen is diffuse, and its origin from a particular organ or site usually is not obvious to the patient. It is no longer uncertain that the gastrointestinal tract is an origin of pain. This pain is evoked by excessive distention (i.e., intralumenal pressure =30 mm Hg) and is exacerbated by inflammation. [110] In humans, excessive distention of the stomach, gallbladder, small intestine, colon, or rectum causes pain that is not felt after bilateral section of the splanchnic (spinal afferent) nerves that carry sensations of pain or discomfort from the abdominal viscera. [111], [112] The quality of the sensation from the digestive tract changes with the intensity of distention, from feelings of fullness to discomfort to pain. [46] Pain signals are believed to be mediated through the activation of high-threshold mechanoreceptors and, in pathological conditions, through afferent fibers that are unresponsive to pressures above the pain threshold in nonpathological conditions (e.g., unresponsive to a lumenal pressure of 70 mm Hg). These unresponsive neurons (silent nociceptors) become mechanosensitive when tissue is inflamed, or when they are exposed to inflammatory mediators. [76] At moderate pressures, low-threshold mechanosensitive afferent neurons are activated to cause reflexes that are unnoticed by a subject without inflammation or other gut pathology. Greater distention activates higher threshold mechanoreceptors and evokes feelings of fullness. Further distention can lead to discomfort and pain. Painful sensations are more likely, and occur at lower thresholds, if the gut is inflamed or if a functional bowel disorder exists, such as irritable bowel syndrome (IBS). This is because the nerve endings in inflamed tissues and in tissue affected by IBS are sensitized. In general, low-threshold primary afferent endings are vagal in origin, and high-threshold nerve endings are of spinal origin. However, some spinal primary afferent neurons are activated by low-threshold mechanical stimuli. [74], [113], [114] Visceral pain is accompanied by somatic and general autonomic reactions, including hunching, abdominal contraction, elevated heart rate, and increased blood pressure. In humans, the intensity of these reactions is directly related to the degree of pain (Fig. 3-4). The somatic and autonomic accompaniments to pain persist when pain itself is prevented by anesthesia. Because they are indications of a stimulus that would normally come to conscious attention, the somatic and autonomic responses observed in a subject under general anesthesia have been called *pseudoaffective responses*. [115] Animal studies confirm that pain is conducted from the intestine through spinal afferent pathways. The neurons are rendered unresponsive by pretreatment with capsaicin, which initially stimulates then desensitizes the endings of primary afferent neurons.

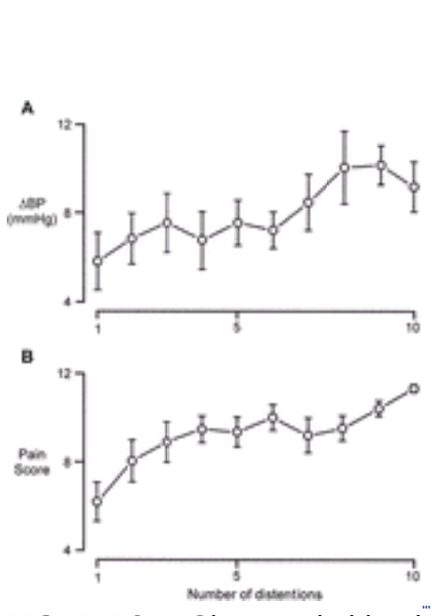

FIGURE 3-4. Changes in blood pressure and psychometric measure of pain perception in human volunteers subjected to inflation of an intrarectal balloon. Successive inflations of the balloon cause more pronounced increases in blood pressure and increased discomfort or pain. (From Ness TJ, Metcalf AM, Gebhart GF. A psychophysiological study in humans using phasic colonic distension as a noxious visceral stimulus. Pain 1990;43:377–386, with permission.)

Chronic changes in the sensitivity of the intestine are observed in IBS. There is a decreased pain threshold to distention of the intestine and exaggerated pseudoaffective responses. [116], [117] and [118] Patients with IBS can feel motility changes that are almost always undetected by healthy individuals. Moreover, contractions within the normal range of amplitude sometimes cause pain in patients with IBS. [119], [120] These and other observations imply a long-lasting increase in the sensitivity of spinal primary afferent mechanisms in IBS.

Nausea The activation of vagal afferent fibers contributes to the nausea and vomiting induced by a number of agents, notably chemotherapy agents, such as cisplatin. In animal models, retching or bursts of synchronous activity in respiratory motor neurons have been used to indicate that nausea or vomiting activity has been elicited. In ferrets, retching and emesis induced by cisplatin or cyclophosphamide were abolished by combined subdiaphragmatic vagotomy and splanchnic nerve section, and were substantially reduced by 5-HT $_3$ receptor antagonists. [121] However, 5-HT $_3$ receptor antagonists did not block retching caused by electrical stimulation of the central end of the severed vagus, leading to the hypothesis that the relevant 5-HT receptors are located peripherally, possibly on the endings of vagal afferent neurons. [122] Subsequent demonstration of 5-HT release by nauseants led to the hypothesis that toxins that induce nausea and vomiting do so by provoking the release of 5-HT from enterochromaffin cells in the proximal small intestine. [123] The 5-HT then stimulates vagal afferent nerve endings, which take signals to the rather loosely defined medullary vomiting centers, including the area postrema and nucleus tractus solitarius. Consistent with the animal studies, 5-HT $_3$ receptor antagonists are effective in reducing chemotherapy-induced and postoperative nausea and vomiting in humans. [124], [125]

Satiety The ingestion of food is dependent on feelings of hunger and satiety, as well as on social and cultural influences. The physiological control center for eating is in the hypothalamus and is influenced by circulating hormones (e.g., leptin), olfactory and taste receptors, and afferent nerve signals conducted from the digestive system to the central nervous system through the vagus nerves. The vagal signals for satiety relate to the volume of food in the stomach, and to the chemical nature of the food products in the duodenum. An animal whose food is diverted by an esophageal cannula and never reaches the stomach provides a model for investigating control through vagal afferents. Studies in such sham-fed animals indicate that gastric distention has a satiating effect, and that the inhibition of eating caused by distention is lost after vagus nerve section. [126], [127] Studies in which the movement of gastric contents is prevented and the composition of the contents is varied indicate that gastric volume, not the nutrients present, is signaled by the vagus. [128], [129] Once the food has entered the duodenum, its nutrient content becomes a factor in influencing satiety. Thus, fat in the duodenum releases CCK from enteroendocrine cells; the CCK, through CCK $_1$ receptors, activates vagal endings and feeding is reduced. [130] CCK acts on mechanoreceptive tension receptors, presumably muscle receptors, as well as on mucosal chemosensitive receptors. [80], [131] Other peptides of gut origin may have roles in determining satiety, possibly through actions on the vagus. These include bombesin, glucagon-like peptide, and ghrelin. [130], [132]

Vagovagal Reflexes Stimuli resulting from the ingestion of food, and the presence of food in the stomach and upper intestine, activate vagovagal reflexes that regulate the activity of the stomach itself and of the pancreas and gallbladder. These reflexes use afferent neurons that end in the nucleus tractus solitarius of the medulla oblongata, integrating circuits within the brainstem, and outputs through vagal efferent neurons that form synapses in enteric ganglia, from which neural connections are made with final effectors (smooth muscle or glands). Vagal afferent endings in the esophagus that are stimulated by swallowing cause a reflex relaxation of the proximal stomach, which is called *receptive relaxation* (i.e., a relaxation in advance of the arrival of the food bolus). [133] The arrival of food in the stomach triggers another reflex through the vagus, the vagovagal accommodation reflex, which involves relaxation of the proximal stomach as well as acid secretion and increased contractile activity in the antrum. [134], [135] A duodenal phase of vagovagal gastric relaxation is triggered by CCK, which is released in response to nutrients, particularly fat, in the lumen of the duodenum. [96], [136] CCK acts on the peripheral ends of primary afferent neurons in the mucosa, relaxing the stomach and thus slowing gastric emptying. The presumed function of this reflex is to allow time for the assimilation of nutrients in the small intestine. Pancreatic function is also influenced by vagovagal reflexes. Nutrients in the upper intestine and stroking of the intestinal mucosa increase pancreatic secretion. This effect is reduced by treatment with capsaicin, or by blocking efferent transmission in the pancreas with atropine. [137], [138] Distention of the body of the stomach, but not the antrum, also increases pancreatic enzyme secretion, again through a vagovagal reflex. [139] Vagovagal reflexes also contribute to the conversion of intestinal motility from the fasted to the fed pattern. [140]

Reflexes Mediated through Spinal Primary Afferent Neurons In addition to conveying pain and discomfort, spinal afferent neurons are involved in the reflex control of the gut, especially the large intestine. The nonnociceptive spinal afferents of the terminal bowel that are concerned with visceral reflexes are analogous to the vagal afferents involved in visceral reflexes. [141] Colonic spinal afferents participate in the control of fecal continence and defecation. Thus, injuries to the pelvic nerves, which carry many of the spinal primary afferent fibers from the descending colon and anal canal, cause derangements of defecation. [142] Defecation reflexes are initiated by rectal distention and chemical or mechanical irritation of the rectoanal mucosa, implying that both muscle tension receptors and mucosal spinal afferent nerve fibers are involved in these reflexes. [143] The section “Intestinofugal Neurons” discusses enteroenteric inhibitory reflexes that pass through prevertebral ganglia without entering the central nervous system. There also are intestinointestinal reflexes in which spinal afferent neurons form the afferent arm and sympathetic pathways are the efferent pathway. [144] These pathways can be activated by distention and also by irritation within the abdominal cavity, such as handling of the intestine, or by infection of the abdominal cavity. It has been suggested that the reflex inhibition of motility through these spinal afferent pathways and through the prevertebral ganglia may facilitate each other because synaptic excitation converges on the noradrenergic neurons in the prevertebral ganglia. [145]

Tissue Protection Neurotransmitters released from the peripheral ends of extrinsic primary afferent neurons may effectively restrict the deleterious consequences of tissue damage. [146] In the stomach, compromising sensory neuron function by pretreating animals with capsaicin aggravates mucosal damage caused by acid, ethanol, nonsteroidal antiinflammatory agents, and other chemicals. [147], [148], [149] and [150] Capsaicin alone is not injurious; in fact, the short-term effect of capsaicin is protective. There is compelling evidence that capsaicin induces the release of the sensory neurotransmitter CGRP from spinal afferent nerve endings, a transmitter that has a major role in protecting the mucosa. [146] Thus, blocking CGRP receptors, or reducing the effectiveness of CGRP by immunoneutralization, compromises gastric protection. [151], [152] The preexperimental application of capsaicin to the spinal afferent pathways, but not to the vagus nerves, blocked the CGRP-dependent hyperemic response to the acute application of capsaicin, indicating that gastric protection is dependent on the integrity of spinal afferent neurons. [153] Because the protection is reduced by the application of the nerve conduction blocker TTX to the stomach, axon reflexes are likely to be involved (i.e., action potential invasion of collateral branches of activated nerve endings). [150] Tachykinins are often co-localized in the gastric spinal afferent neurons, and data indicate that tachykinins, released from the spinal primary afferent endings and acting through NK $_2$ receptors, contribute to gastric protection. [154] The protective mechanism involves vasodilation and increased mucosal blood flow. CGRP and tachykinins cause vasodilation by releasing nitric oxide from vascular endothelial cells, and thus, inhibition of nitric oxide synthase increases gastric damage. [155] Transmitter release from afferent endings in the colon also reduces the severity of damage consequent on inflammation, at least in the acute phase. [156]

INTESTINOFUGAL NEURONS

Intestinofugal neurons are an unusual type of neuron. Their cell bodies are in the gut wall, and their processes run toward the central nervous system and orm synapses in prevertebral sympathetic ganglia. Their presence was deduced by Kuntz and Saccomanno in the 1930s, [157] who found that distention of one region of the gastrointestinal tract inhibits motility in other regions and that these enteroenteric inhibitory reflexes persist after connections with the central nervous system are severed, provided that the integrity of the connections with prevertebral sympathetic ganglia is maintained. Methods to study these reflexes in vitro were developed in the early 1970s, [158] and since that time, the organization of the pathways has been studied in considerable detail. [145] The location of the cell bodies of intestinofugal neurons in the myenteric ganglia of rats and guinea pigs was determined by injecting retrogradely transported dye into prevertebral ganglia. [159], [160], [161] and [162] In pig, some intestinofugal neurons have cell bodies in submucosal ganglia. [163] Intestinofugal neurons are most numerous in the large intestine; in the small intestine, they increase in number distally, and they are rare in the stomach. The axons of intestinofugal neurons form excitatory synapses with the cell bodies of sympathetic neurons in prevertebral ganglia. The axons of the sympathetic neurons project back to the gut. All intestinofugal neurons have the same primary transmitter, acetylcholine. [145], [164], [165]

Roles of Intestinofugal Neurons

The roles of intestinofugal neurons have been analyzed almost exclusively in relation to motility control, although these neurons also innervate sympathetic neurons that inhibit water and electrolyte secretion in the intestine (see Chapter 2). The intestinofugal neurons that affect motility are in the afferent limbs of enteroenteric inhibitory reflexes. These reflexes appear to act primarily on sites in the gastrointestinal tract that are proximal to the sites from which they are initiated. [145] Thus, the reflexes are one of the mechanisms by which more distal parts of the intestine regulate the proximal regions from which they receive products of digestion. The enteroenteric inhibitory reflexes appear to have a protective role in the gastrointestinal tract. For example, enterogastric reflexes slow gastric emptying, thereby protecting the duodenal mucosa from acid and osmotic stress.

OVERVIEW

The gastrointestinal tract can be regarded as a sensory organ. It has three detecting systems: neurons, endocrine cells, and the gut immune system. The neural detection of sensory information is through both intrinsic sensory (primary afferent) neurons and extrinsic sensory neurons. The neural sensory system is responsible for detecting the state of the digestive system. The detected information is used to govern (1) reflexes confined within the gut wall (intrinsic reflexes) that change

motility, blood flow, and transepithelial fluid transfer; (2) extrinsic, but peripheral, reflexes between digestive organs; (3) reflexes through the central nervous system that are not consciously perceived; and (4) conscious sensation, such as satiety, hunger, discomfort, pain, and nausea. Although many of the effects mediated through digestive system afferent neurons have been elegantly dissected, the subtleties of their interactions are still being unraveled. The challenge remains to establish a good linkage between what is known of the responses of individual primary afferent neurons and the integration of the information they provide to yield an output, either a conscious experience or a physiological response.

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CHAPTER 4

Laurence J. Miller

GASTROINTESTINAL HORMONES AND RECEPTORS

The gastrointestinal endocrine system represents the largest and most complex endocrine organ in the body. Whereas traditional endocrine organs are composed of a solid mass of hormone-secreting cells that are homogeneous, or at least organized with a defined spatial orientation relative to one another, the sources of the gastrointestinal regulatory molecules are single endocrine cells and peptidergic neurons that are scattered along the length of the digestive tract. They all share the property of producing and secreting polypeptides. Because these hydrophilic molecules cannot cross a lipid bilayer, they must interact with other molecules that span the plasma membrane of target cells, which can activate the intracellular machinery of those cells. These membrane protein targets of gastrointestinal hormone action are called *receptors*. They have specialized domains for ligand recognition that are accessible to the extracellular space, as well as effector domains for molecular interactions or enzymatic activity that are located intracellularly. The diversity and design of the gastrointestinal endocrine system also necessitate broadening the basic concept of classic endocrinology, which requires that a hormone be delivered to its targets by way of the bloodstream. Although the bloodstream delivers some gastrointestinal hormones, the gastrointestinal endocrine system also uses more direct and focused means of transmitter delivery.

The endocrine cells are scattered among the epithelial cells of the mucosal lining of the gut and the ductular elements that enter the gut. They have an open or closed morphology. Endocrine cells with an open morphology can sense the composition of chyme and secretions through processes that extend into the lumen. A closed morphology facilitates sensing of the interstitial environment. [1,2] Peptidergic neurons also exist and are organized into syncytia, which extend between and along layers of the mucosa and into the solid digestive organs. These neurons are classified as extrinsic or intrinsic, depending on the location of their cell bodies. Extrinsic parasympathetic innervation comes from the vagus and pelvic nerves, whereas sympathetic innervation comes from various sympathetic ganglia. The major source of peptidergic nerves is intrinsic to the gut and is affected little by sympathectomy or vagotomy. There are numerous varieties of both the endocrine cells and the peptidergic neurons of the gastrointestinal tract. Each type of cell has a distinct distribution and is able to sense and respond to a unique complex of stimuli, resulting in the synthesis and secretion of a single predominant peptide.

Each hormone or neurotransmitter affects only those target cells possessing appropriate receptors on their surface. Cells that can be regulated include the vast spectrum of epithelia, smooth muscle cells, and neurons residing at all levels of the digestive tract. Responses span the temporal spectrum, from rapid and transient to delayed and prolonged. The former includes effects on behavior or sensation, secretory or absorptive phenomena, motor events, and metabolic events. The more prolonged effects typically involve cellular growth, differentiation, or morphogenesis. The combination of unique stimuli, limited distribution of distinct types of hormone-secreting cells, and exquisite selectivity of target cells and their responses provides almost unlimited opportunities for regulation. This elegant system integrates digestive functions at all levels of the gastrointestinal tract, thereby optimizing the digestion and absorption of nutrients. For all these reasons, the gastrointestinal endocrine system has been referred to as the *brain of the gut*.

ORGANIZATION OF THE GUT ENDOCRINE SYSTEM

The organization of the gastrointestinal endocrine system is the key to its power and flexibility. Acutely regulated gastrointestinal events are dynamic, complex, and interrelated; the ingestion of a meal initiates changes in secretion, absorption, and transit along the gut and in its associated digestive organs, and the net composition of chyme and its rate of delivery vary markedly over time and space. More delayed and prolonged effects on the differentiation and growth of enteric cells are equally important for the optimal digestive responses for an individual's dietary habits and nutritional requirements. Given the diversity in size and composition of ingested meals, the gastrointestinal endocrine system must be capable of highly varied programs of responses, not only to ensure proper assimilation of the nutrients, but also to make eating the pleasurable experience it represents for the vast majority of the population.

The gastrointestinal endocrine system must be responsive both to intralumenal digestive events and to systemic status. The combination of "open" and "closed" endocrine cells and nerves provides sensitivity toward both worlds, recognizing that the general health, nutritional status, and metabolic needs of the organism can modify the optimal nutritional impact of a meal. With each hormone-secreting cell sensitive to its local environment, and with each such cell strategically located, endocrine cells of the same type in two different locations likely secrete their peptides at different times, reflecting the dynamic changes occurring along the length of the gut after a meal. An additional level of control is made possible by the route of delivery and the mode of action of the secreted regulatory peptide. This can be endocrine, autocrine, neurocrine, or paracrine (Fig. 4-1).

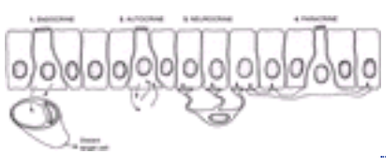

FIGURE 4-1. Modes of delivery of gastrointestinal regulatory peptides from endocrine cells and nerves, illustrating endocrine, autocrine, neurocrine, and paracrine mechanisms.

The term *endocrine* specifically refers to hormonal delivery by way of the bloodstream, providing a mechanism to transport the hormone to distant sites. If this circulation traverses a specialized organ, such as the portal circulation through the liver, the opportunity exists to extract a particular hormone efficiently from the blood, thereby preventing broad systemic distribution. This can differentially affect certain molecular forms of the same hormone. For example, short molecular forms of cholecystokinin (CCK) are extracted in a single pass through the liver, whereas long forms pass through to the systemic circulation. [3] There are also specialized circulatory delivery systems relevant to gastrointestinal peptides, such as those secreted from the pancreatic islets; an islet-acinar portal circulation provides high

concentrations of peptides to the surrounding pancreatic acinar cells. [4]

Autocrine loops are postulated to exist, whereby the peptide synthesized by a cell affects that same cell. This may occur after secretion into the interstitial space, such that back-diffusion allows high local concentrations of peptide to interact with receptors on the plasmalemmal surface of the hormone-secreting cell. Although more difficult to understand from a mechanistic standpoint, it also has been postulated that autocrine effects occur within the cell, independently of hormone secretion. This would require the ability of the peptide, which is within a membrane-bound vesicular compartment, to interact with another molecule playing the role of receptor in an accessible location within the cell. There is no reason why such a molecule must be identical to the receptor molecules we recognize to reside within the plasmalemma with their hormone-binding sites exposed to the extracellular milieu. There has been some indication that molecules, such as guanine nucleotide–binding proteins (G proteins), which are proximal effectors of hormonally stimulated events at the level of the plasmalemma, may also reside on secretory granule membranes. [5], [6]

Neurocrine delivery of peptides usually involves their local release adjacent to a target cell bearing relevant receptors. [7], [8] Neurons are particularly well suited for this mode of delivery because they can have highly specialized long processes that terminate on or adjacent to potential target cells. It is also important to appreciate that this mode of delivery provides a gradient of transmitter concentrations from the point of release. Concentrations may fall off rapidly with distance from this point, limiting action on other receptor-bearing cells in the general area.

Paracrine delivery reflects the identical process originating within an endocrine cell rather than a peptidergic neuron. These cells are also capable of developing specialized processes that allow the secretion of a key transmitter adjacent to its target cell. The first identified example of this was the relationship between somatostatin-secreting cells and gastrin-secreting cells in the gastric antrum. [9]

Once the regulatory peptide is delivered to its target cell, a high level of regulatory diversity is still provided by the specificity of receptors expressed on each cell and by a whole universe of potential signaling and regulatory molecules. A given endocrine cell can further regulate hormone-stimulated events in several different ways: by altering coupling events between receptor and proximal effector, by modifying the complement of molecules intrinsic to each signaling pathway, and by cross talk between the molecules involved in signaling pathways. Each cell also can regulate the receptors it expresses, a phenomenon highly relevant to any understanding of the gastrointestinal endocrine system. This regulation can take the form of modification of the number of receptors on the cell surface, modification of the ability of those receptors to bind ligands or to initiate signaling, movement of receptors into different cellular compartments, and modification of the total cellular complement of receptors by effects on receptor synthesis and degradation. [10], [11] and [12]

A given type of receptor may be present on a wide variety of cells of the body, but the secretion of its hormonal agonist may affect only a small subset of those cells. This may reflect the different limited delivery systems discussed above. It also may reflect a gradient in delivery by diffusion from a point source, with potential target cells also expressing different numbers of receptors on their surfaces. Receptors on distinct cells may be in different states of sensitization or desensitization. A certain type of cell may express a receptor differently at each stage of its life, depending on cell growth, the stage in the cell cycle, and other active metabolic and biochemical processes within the cell.

The gastrointestinal endocrine system represents a system of substantial complexity, which is dynamic and responsive to feedback and cross regulation at the levels of both the hormones and the receptors. It is precisely because of this complexity that the system is so sensitive and has such extraordinary capabilities to keep us well nourished and pleased with our dining experiences.

HISTORY OF GASTROINTESTINAL ENDOCRINOLOGY

Classic endocrinology and gastrointestinal endocrinology were born together in 1902, with the discovery of the first hormone, secretin, by Bayliss and Starling [13] and the recognition that chemical messengers can be carried in the bloodstream. This earliest period in gastrointestinal endocrinology has been termed the *juice-physiology era*. [14] The discovery of gastrin by Edkins followed quickly in 1905. [15] This flourish of activity in the gastrointestinal hormone field was followed by minimal discoveries for many years, while the field of classic endocrinology achieved steady and substantial growth. The solid and homogeneous classic endocrine organs could be studied with relative ease, whereas methods had not yet been developed to gain analogous insights into the scattered diverse gastrointestinal endocrine cells.

Gastrointestinal endocrinology finally achieved its status as a distinct discipline in the chemical era, when a long series of candidate hormones were discovered by means of the techniques of purification and peptide sequencing. The first was gastrin, whose structure was determined in 1964 after it had been extracted from a tumor. [16] Subsequently, the number of recognized potential regulators of digestive function expanded logarithmically. The Gastrointestinal Hormone Laboratory, founded by Mutt and Jorpes at the Karolinska Institutet, [17] was the site of the initial chemical characterization of secretin, CCK, glucose-dependent insulinotropic peptide (GIP), vasoactive intestinal polypeptide (VIP), motilin, gastrin-releasing peptide (GRP), and many other candidate structures.

The next key advance was also methodologic—the development of radioimmunoassay to detect or quantify minute quantities of molecules. Application of this methodology to the quantification of a hormone by Yalow and Berson [18], [19] and its application to CCK [20] initiated the immunologic era of gastrointestinal endocrinology. The usefulness of antibodies was appreciated quickly; these were applied to the localization of hormone-secreting cells by immunohistochemistry and to the quantification of peptide hormones by radioimmunoassay. The usefulness of radiolabeled peptide hormones also was recognized quickly. The radiolabeling of gastrointestinal hormones to highly specific radioactivity made it possible to demonstrate directly the binding of hormones to receptors. The competition-binding technique made it possible to identify receptor subtypes, to define the cellular location of receptors, and to quantify receptor density on a given type of cell.

Since the 1970s, gastrointestinal endocrinology has become increasingly biochemical and molecular. Gastrin led the way into the new era of the molecular biology of peptides. [21] A more biochemical understanding of hormones excited interest in the biochemistry of receptors. During this era of molecular receptorology, [21] receptors increased in status from representing simple sites of hormone binding to being the focus of intense interest. It was critical to understand the structure of receptor molecules before the details of the basis of binding, activation, and regulation became meaningful.

The first insights into the structure of receptors came from affinity labeling approaches, [22], [23] which represented a logical extension of the receptor-binding assay described above. These approaches involved the use of a radiolabeled form of the hormone, as rich a source of receptor as possible, and a chemical method to link the two molecules together covalently. The siting of a photolabile group within the pharmacophoric domain of a hormone has been particularly successful in the covalent labeling of hormone receptors and their domains. [24]

The most exciting and powerful insights in this era have come from cloning the complementary DNA (cDNA) encoding the receptors. If the partial receptor sequence is available, an oligonucleotide probe can be designed. [25], [26] Hybridization screening also can be used to detect a homologous receptor or a known receptor in a new species or tissue. [27] Expression cloning efforts bypass the need for pure receptor and partial receptor sequence. [28] Given the difficulties in purifying what are typically very sparse molecules that are extremely hydrophobic and difficult to work with, this has become a very popular approach to cloning.

Once a receptor cDNA has been cloned and sequenced, a universe of possibilities is revealed. The deduced amino acid sequence provides insights into the receptor class represented. By finding its position in a phylogenetic tree, additional insights can come from analogy with studies of the closest known members of the family. [29] This can take the form of mutagenesis studies that provide insights into structure-function relationships. Chimeric receptors that combine specific regions of two or more receptors can be useful in identifying which domain is important for a given function. [30], [31] Also, truncations and site mutants can help localize such regions. [32] Such receptor constructs can be constructed rapidly and expressed transiently to gain new insights. It is also possible to produce stable cell lines that express such constructs, [33] and to modify the expression of natural or mutagenized hormones and receptors in laboratory animals. A variety of transgenic and null (i.e., knock-out) animals are available to explore the impact of such modifications in an intact animal.

The next era will probably be the era of molecular conformation and molecular interaction. [21] Now that the primary structures of receptors can be catalogued, molecular conformations must be studied, including both resting and active conformations. There will be major interest in the sites and molecular determinants of binding and activation, as well as the use of such insights for the rational design of useful pharmaceuticals. We are already witnessing dramatic advances in drug development resulting from novel strategies of synthesizing and screening vast libraries of potential receptor ligands. [34], [35] These have led to the successful development of numerous nonpeptidyl antagonists, and even nonpeptidyl agonists, for peptide hormone receptors. [36], [37] There is no doubt that for gastrointestinal

endocrinology, the future promises to be even more exciting and successful than the recent past.

HORMONE SECRETORY CELLS

Several cellular sources exist for the synthesis and secretion of gut hormones. As previously noted, these can represent the numerous types of endocrine cells that are isolated and scattered throughout the gut mucosa, as well as the neurons contained within the enteric nervous system. Both cell types share many features: (1) the machinery for peptide biosynthesis and processing, (2) the packaging of products for export into secretory granules and the exocytic secretion of these products, and (3) the specialized processes that may facilitate focused delivery of secretory products. Based on these shared characteristics, similar embryogenesis also has been postulated. This section focuses on each of these features as they relate to the sources of gastrointestinal hormones and neurotransmitters.

In 1968, A.G.E. Pearse [38] introduced the concept of amine precursor uptake and decarboxylation (APUD) and suggested that it is a feature that paraneuronal cells, such as the gastrointestinal endocrine cells, may share with neurons. It can now be concluded that the endocrine cells and enterochromaffin cells of the gastrointestinal tract arise from embryonic endoderm, like the pancreatic islets. [39], [40] All types of intestinal epithelial cells appear to be derived from a single type of pluripotent stem cell precursor. [40], [41], [42] and [43] Gastrointestinal endocrine cells continuously differentiate and follow the highly organized vertical paths of migration along the crypt to the villous axis. These cells have embryological associations and evolutionary relationships similar to those of the endocrine cells of the pancreatic islets. The gastrointestinal hormones gastrin, secretin, and peptide YY are all transiently expressed in fetal islets during development, and the pancreas and duodenum are common sites of gastrin-secreting tumors. [44], [45]

In contrast, the enteric neurons are derived from the neural crest and the neuroectoderm. [8], [46] Some extremely interesting data suggest that the phenotype of cells derived from the neural crest is most dependent on environmental influences in the regions to which they migrate, rather than on the level of the neural axis from which they originate. [46], [47] The complexities of cell types and interactions typical of the enteric nervous system result largely from the mesenchymal substrate that these cells contact. The factors responsible for the development and differentiation of each of the gastrointestinal mucosal endocrine cells are even less well understood.

Despite this apparent difference in their developmental origins, the gastrointestinal endocrine cells and the enteric peptidergic neurons share many biologic and biochemical specializations necessary for polypeptide synthesis, processing, and secretion. Key in this sequence of events is the packaging of the peptide product for export into a secretory granule. The details of this process were clearly defined by Jamieson and Palade, [48], [49], [50], [51] and [52] who studied the pancreatic acinar cell (Fig. 4-2). The identical processes appear to exist in the gastrointestinal endocrine cell.

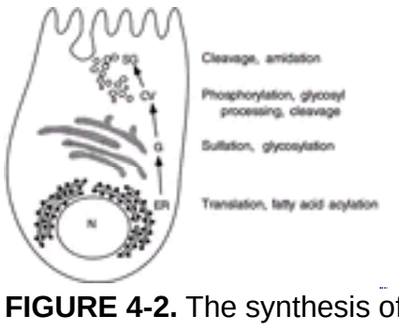

FIGURE 4-2. The synthesis of proteins or peptides for export (i.e., secretion) from polarized epithelial cells follows a highly conserved biosynthetic route. Synthesis begins at the level of the rough endoplasmic reticulum (*ER*) and proceeds through the Golgi apparatus (*G*), condensing vacuoles (*CV*), and secretory granules (*SG*) before exocytosis at the apical pole of the cell.

ENDOCRINE CELL LOCALIZATION AND CHARACTERIZATION

Some gastrointestinal endocrine cells can be recognized by the relatively distinct ultrastructural appearance of their secretory granules [1], [2] (Fig. 4-3). Other gastrointestinal endocrine cells can be tentatively identified not only according to the appearance of their secretory granules but also according to their position along the digestive tract. However, more specific techniques are required to identify most endocrine cells and any of the peptidergic nerves. These techniques are directed toward the polypeptides themselves or to the genes and messenger RNA (mRNA) molecules encoding them (Fig. 4-4). Peptide antisera have been most useful in the immunohistochemical localization of peptides. In situ hybridization has been successfully used to localize relevant hormone mRNA.

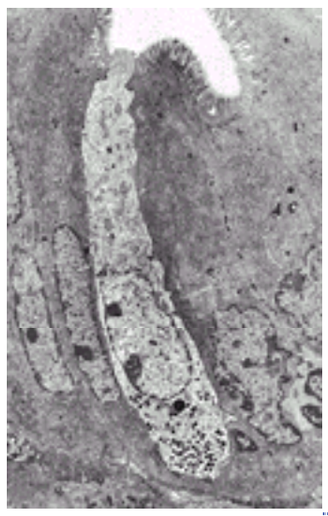

FIGURE 4-3. Electron micrograph of a gastrointestinal endocrine cell in the human jejunum. This cell extends from the lumen to just above the basal membrane and includes the typical electron-dense secretory granules in its basal pole. (From Solcia E, Capella C, Buffa R, et al. Endocrine cells of the digestive system. In: Johnson LR, ed. Physiology of the gastrointestinal tract, 2nd ed. New York: Raven Press, 1987:112, with permission.)

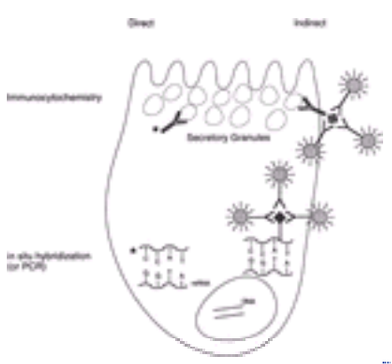

FIGURE 4-4. Methods to identify specific endocrine cells include direct and indirect immunocytochemistry, with antibodies directed toward the hormones themselves, and in situ hybridization (or polymerase chain reaction [PCR]), in which oligonucleotide probes bind to mRNA.

Immunohistochemistry has become a standard technique to localize any antigen in a tissue section to which specific antisera have been raised. With the current understanding of the primary structure of the molecular forms of a given hormone, and of the unique and shared domains within that structure, it is easy to choose a potentially useful antigenic epitope. By conjugating that peptide to a hapten, such as keyhole limpet hemocyanin, a strongly immunogenic antigen can be produced. Most endocrine cells and peptidergic nerves concentrate their peptides in secretory granules in preparation for secretion, providing an ideal concentrated target that has multiple copies for immunolocalization. A potential problem relates to the ability to fix these highly soluble compounds without interfering with the antibody recognition domains. Trials of different fixatives and conditions of fixation may be necessary for an optimal signal.

In situ hybridization is an analogous localization technique based on the specific interaction of an oligonucleotide probe with its complementary sequence within the cell. [53] This most often takes the form of an mRNA molecule. A complementary, single-stranded, antisense riboprobe provides optimal sensitivity and specificity. Here, too, multiple choices exist: fluorescent, colorimetric, or radioactive detection. [54] Limited copy number for a specific mRNA may represent a problem for applying this technique to some hormone genes.

BIOSYNTHESIS AND PROCESSING OF GASTROINTESTINAL HORMONES

The gastrointestinal hormones and neurotransmitters discussed in this chapter are single-chain polypeptides. All are encoded by single-copy genes, containing one or more exons that determine the primary amino acid sequence by a series of nucleotide triplets known as *codons* (Fig. 4-5). For the multiple-exon genes, these coding regions are separated by noncoding introns. While still in the nucleus, the DNA sequences are transcribed by RNA polymerase II to form mRNA. The noncoding regions of the gene, particularly those upstream of the site of the initiation of transcription (i.e., the 5' flanking sequence), often contain *cis*-active regulatory sequences that influence the rate of transcription. Regulation of gene expression typically occurs slowly, over a number of hours, whereas hormone secretion of prepackaged product occurs rapidly, being initiated within seconds of stimulation. During the process of transcription, the sequences complementary to the exons are spliced together while those complementary to the introns are eliminated. Sometimes, there is more than one product of transcription, representing alternatively spliced products in which some potential coding regions are eliminated. This can have substantial functional significance and may occur selectively in a given type of cell or may even be a regulated process occurring only at certain times in that cell. For many hormones, the product of transcription represents a preprohormone, with an amino acid sequence that helps direct the product of translation into the exocytic pathway for export and undergoes additional cleavage and processing before secretion from the hormone-producing cell.

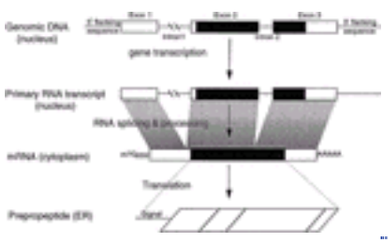

FIGURE 4-5. The gene structure and the molecular biologic events leading to protein synthesis. Posttranscriptional processing events that occur in the nucleus include splicing, capping, and polyadenylation of the RNA to produce mature mRNA. Mature mRNA is transported into the cell cytoplasm, where it directs translation at the surface of the rough endoplasmic reticulum (*ER*).

Several examples exist of alternate processing of a single gene. There may be more than one distinct product within a single gene. Examples of this include the proopiomelanocortin gene, which contains sequences for ß-endorphin, corticotropin, and melanocyte-stimulating hormone, [55] and the proenkephalin gene, which contains five copies of the Met enkephalin sequence and one copy of the Leu enkephalin sequence. [56] VIP, neuromedin A, and peptide histidine isoleucine (PHI) sequences all exist within a single gene. [57] Another phenomenon is the production of distinct gene products by alternate splicing of a single gene, such as that which occurs in the production of calcitonin and calcitonin gene–related peptide. [58] Similarly, alternate splicing of a single gene product may also occur. [59] Several hormone receptors can have a variety of alternately spliced sequences to yield products of different lengths with short deletions or insertions from the predominant species. [60], [61]

The fully spliced mRNA is translocated to the cell cytoplasm, where translation occurs (Fig. 4-6). Translation is the process of producing the polypeptide chain representing the preprohormone that is encoded by the mRNA. Peptide synthesis begins on the ribosome bound to the rough endoplasmic reticulum. Peptides destined for export have a characteristic amino-terminal signal sequence that leads to their segregation within the lumen of the endoplasmic reticulum. [62] As the nascent chain crosses into the lumen of the endoplasmic reticulum, the signal sequence is bound to a signal-recognition particle and cleaved by a signal peptidase to yield the prohormone. Also, posttranslational modifications may begin cotranslationally. For example, sulfation, phosphorylation, glycosylation, and amidation may continue as the peptide moves through the Golgi apparatus and into secretory granules that are at various stages of maturity. The ultrastructural appearance of these secretory granules can be characteristic of a particular type of gastrointestinal endocrine cell. [1] The large secretory granules typify regulated pathways of exocytosis, with constitutive secretory pathways most commonly using much smaller vesicular structures. Glycosylation is a multistep process that occurs through each of these compartments. Tyrosine sulfation seems to occur early in biosynthesis, within the Golgi apparatus. [63]

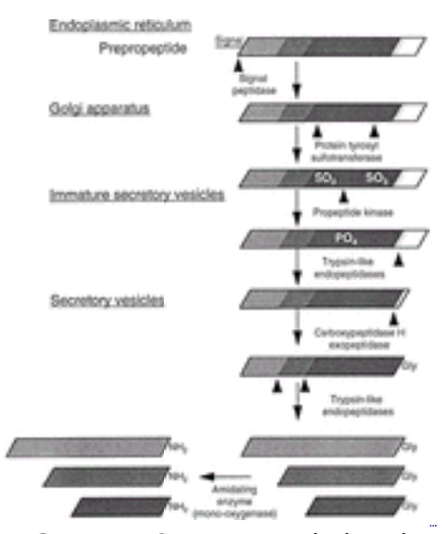

FIGURE 4-6. Posttranslational processing events that occur in distinct cellular compartments in preparation for secretion of a peptide from the cell. These types of reactions are used selectively for different secretory products.

A unique posttranslational processing particularly relevant for peptide hormones is the selective cleavage of the prohormone into fragments of different lengths, with each form possessing biologic activity. [64], [65] Examples of multiple, biologically active forms of a hormone include gastrin and CCK; the multiple forms of these hormones share the carboxyl-terminal sequence critical for activity. Endoproteolytic processing occurs during biosynthesis while the product traverses the late Golgi cisternae and early secretory granule compartments. Dibasic amino acid residues are a common location for such processing. Trypsinlike endopeptidases, such as PC1 and PC2, which are uniquely expressed in endocrine and neuroendocrine cells, have been described to be important in the endoproteolytic processing of

gastrointestinal hormones. [66], [67]

Carboxyl-terminal amidation is a very common posttranslational modification of gastrointestinal hormones. [68], [69], [70] and [71] It occurs in about half of the recognized hormones and peptide neurotransmitters, and was the basis of a discovery strategy used early in the chemical era by Tatemoto and Mutt. [72] A series of enzymatic reactions is necessary to yield an amidated product (1) cleavage at a site of dibasic residues, (2) trimming of the remaining basic residues by carboxypeptidase H, and (3) conversion of the glycine-extended intermediate to the corresponding carboxyl-terminal amide with an a-amidating monooxygenase. These processes occur within the secretory granules themselves.

The processing of gastrin in the antral G cell is an excellent example of these processes. [73] The biologically active region of this peptide that is key for binding to its classic receptor has been carefully isolated to the carboxyl-terminal tetrapeptide-amide of the mature forms of gastrin. [74], [75] The gastrin gene is composed of three exons, with exons 2 and 3 possessing the coding sequences. [76], [77] and [78] The primary mRNA transcript is processed to eliminate approximately 3 kilobases of intron sequence between the first two exons and 130 bases of intron sequence between exons 2 and 3. This is translated to yield preprogastrins of 101 amino acid residues in man and 104 amino acid residues in rat and pig. The signal peptide is then cleaved to yield progastrin, which rapidly undergoes further proteolysis at Arg ^{57,58}, Lys ^{74,75}, and Arg ^{94,95} to yield four fragments. The Arg ^{94,95} cleavage is followed by carboxypeptidase H trimming of carboxyl-terminal basic residues and conversion of the Gly-extended intermediate to the carboxyl-terminal amide. G34 and G17 are the major products of these cleavages. Serine phosphorylation and tyrosine sulfation may also occur. [73], [79] One of the sites of serine phosphorylation is Ser ^{96}, immediately adjacent to the site of cleavage at the dibasic residues, which ultimately results in carboxyl-terminal amidation. Tyrosine sulfation can occur at Tyr ^{87} and Tyr ^{103}. Whether these modifications affect physiological processing remains unclear. [80]

Posttranslational modifications of gastrointestinal hormones are potentially important for their activity. Most of the amidated hormones require modification (i.e., amidation) for recognizable activity. Gastrin exhibits a spectrum of activities, including growth stimulation, which has been attributed to a nonamidated Gly-extended form of the hormone. [81] Presumably, this posttranslational modification is mediated through a receptor that is distinct from the molecule mediating its classic activities. Another key modification is the sulfation of the tyrosine, seven residues from the carboxyl terminus of CCK peptides. [82] This modification is critical, with the desulfated peptide more than 1000-fold less potent. The structurally related gastrin peptides also can exhibit tyrosine sulfation six residues from the carboxyl terminus, but this modification has little or no impact on receptor binding or hormone activity.

HORMONE AND TRANSMITTER MOLECULES

As noted above, all the relevant hormones and neurotransmitters discussed in this chapter represent single-chain polypeptides. They are all hydrophilic and soluble in aqueous media. This property prevents them from traversing the lipid bilayer in the target cell and necessitates the presence of a receptor molecule with which they can interact at the level of the target cell. By first principles, peptides are also flexible. The peptide bonds linking the amino acid residues are stable and strong, broken only by peptidase enzymes under normal biologic conditions. This flexibility enables peptides to sample a large range of potential conformational space, but makes it somewhat difficult to predict the relevant conformation of these molecules as they bind to their targets. Some insights have come from observing the conformationally constrained analogs of some hormones, but such insights are limited. [83]

Dramatic advances in peptide chemistry have permitted the routine synthesis of small peptides. Peptide synthesis can be achieved in solution or by solid phase techniques in which one amino acid residue is added at a time to a growing chain of residues. [84], [85] This technique can be used to produce the natural peptides themselves, or analogs that can provide important primary structure and activity information.

Gastrointestinal hormones can be categorized according to (1) predominant localization in endocrine cells or peptidergic neurons; (2) localization to different levels of the gastrointestinal tract; (3) rapid effects versus more prolonged effects on growth and differentiation; or (4) the presence of structural similarities between the hormones themselves (Table 4-1). A single peptide hormone can be found in endocrine cells and nerves, it can be present at several levels of the gastrointestinal tract, and it can have both acute and chronic growth regulatory effects. This is also true for families of structurally related hormones. A single peptide can interact with and activate more than one type of receptor. Only by appreciating the scope of options for these peptides and their receptors can the complexity and the power of the gastrointestinal endocrine system be understood.

TABLE 4-1 Organization Schemes for Gastrointestinal Hormones, Neuropeptides, and Locally Secreted Growth Factors According to Their Cell of Origin, Anatomical Location, and Structural Similarities

MEASUREMENT OF HORMONES

Hormone concentrations can be quantified by bioassay or by a type of competition-binding assay, such as radioimmunoassay or receptor-binding assay. FONT size=-1>[19], [86] Before the chemical characterization of hormones, concentrations could be determined only in an activity assay. The specificity of an activity assay is unreliable because more than one hormone can produce the same response and because antagonistic agents can influence the net activity. The determination of the chemical identity of many hormones has facilitated the assay of unpurified samples.

Competition-binding assays are based on the competitive inhibition of the specific binding of a radiolabeled form of the hormone to an antibody or receptor. The radiolabeled and natural hormones compete for binding to the antibody or receptor. Radioimmunoassay benefits from the availability of specific antibodies that are easy to handle and store. In contrast, radioreceptor assay depends on the preparation of a receptor that may be difficult to produce and store in an active form.

For all hormone assays, sample collection and handling are critical, and sampling conditions should be reproducible. Samples usually are collected during fasting because hormone concentrations may increase in the postcibal period. Gastrin, CCK, secretin, GIP, and pancreatic polypeptide all increase transiently after a meal, whereas concentrations of most neurotransmitters (e.g., VIP) change minimally. [87] After sample collection, low temperature and protease inhibitors minimize hormone degradation.

Knowledge of hormone concentrations in the circulation can provide important insights. Elevated basal levels may reflect excess secretion or reduced clearance. Excess secretion may develop if hormone-secreting cells proliferate, as occurs in gastrin cell hyperplasia, or if cells have lost their feedback inhibitory mechanisms and secrete constitutively, as occurs in various types of endocrine cell neoplasia. Because polypeptide hormones are cleared by the kidneys, high levels of hormones have been reported to exist in renal failure. [88] In some settings, it may be useful to measure hormone concentrations serially after some type of stimulus, such as the ingestion of a meal or the perfusion of a region of the intestine, to define the timing and characteristics of a normal stimulated response. There are provocative tests

for some endocrine tumors, such as secretin stimulation of gastrin in gastrinoma. [89]

The confirmation of the role of a hormone in a particular physiological process is not simple. Determination of the physiological activity of a particular gut peptide is based on the following criteria: (1) There is a relationship between the appearance of the hormone in the circulation and the physiological event; (2) the physiological effect can be reproduced by infusion of pure or synthetic hormone to achieve the same serum concentration observed under physiological conditions; and (3) the physiological activity can be inhibited by infusion of a specific antibody or receptor antagonist, when available. [90], [91] The circulating concentrations of the hormone that produce the biologic effect should be similar for the natural peptide released physiologically or the synthetic peptide infused into the experimental subject. When multiple molecular forms of a hormone exist, it is important for the infusion to reproduce the forms naturally observed as closely as possible.

Unfortunately, it is more difficult to determine the relevant concentration of gastrointestinal hormones at the level of their receptors. Interstitial concentrations can vary significantly over short distances. There is no good, reproducible method to collect the relevant fluid to measure hormones at their site of action. This is a particular problem for hormones and neurotransmitters released through paracrine or neurocrine mechanisms.

RECEPTORS

Receptor molecules also have a special place in gastrointestinal endocrinology. Hormones and transmitters cannot cross the plasma membrane of target cells to activate intracellular machinery. Therefore, a molecule that spans the lipid bilayer in the target cell plasma membrane, such as a receptor, is required. The receptor molecule has two spatially distinct domains one domain to bind ligands that approach the molecule from the extracellular side of the membrane, and a second to initiate activity cascades within the cell. The cellular response can be triggered by a ligand-induced conformational change that activates an enzymatic activity intrinsic to the receptor. Alternatively, the ligand-induced change in the conformation of the receptor can expose a receptor domain that interacts with an important intracellular molecule, and the interaction initiates the cascade of signaling events typical of that receptor and ligand pair.

With the cDNAs of many receptor proteins now cloned, and their primary structures known, insights into the structural basis of cell activation are being advanced rapidly. Gastrointestinal hormone receptors are grouped into three broad classes according to their membrane topology: (1) the single-transmembrane receptors, (2) receptors with subunits that traverse the membrane multiple times, and (3) the G protein–coupled receptors that traverse the membrane seven times (Fig. 4-7).

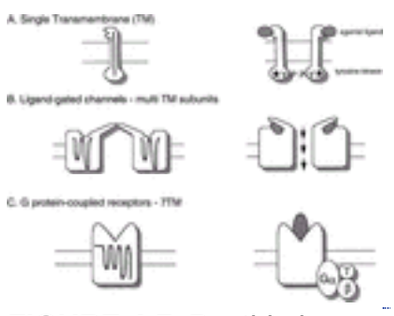

FIGURE 4-7. Peptide hormone receptors can be divided into three groups according to their membrane topology. The basal conformations are shown on the left and activated states on the right. **A:** The single-transmembrane (*TM*) receptors dimerize; some also possess intrinsic tyrosine kinase activity that contributes to the autophosphorylation of the complex. **B:** The ligand-gated complexes undergo a conformational change that opens the channel. **C:** The G protein–coupled receptors become associated with their heterotrimeric G proteins, which then initiate the signaling cascade.

The largest group of single-transmembrane receptors possess intrinsic ligand-triggered enzyme activity. This group includes ligand-triggered protein tyrosine kinases, such as the epithelial growth factor receptor, insulin-like growth factor receptors, platelet-derived growth factor receptor, fibroblast growth factor receptor, and transforming growth factor receptors. [92], [93] and [94] Ligand-triggered membrane guanylate cyclases also exist, such as the receptor for atrial natriuretic peptide. [95] There is also a group of single-transmembrane receptors that do not appear to have any intrinsic enzymatic activity, but rather function by dimerization and initiation of cascades of modular interactions. These include receptors for growth hormone, erythropoietin, prolactin, and several cytokines. [96], [97]

The second group of receptors (ligand-gated channels) have subunits that span the membrane multiple times and associate to form pores or channels. [98] Examples include the nicotinic acetylcholine, GABA $_A$, 5-HT $_3$ (serotonin), glycine, and glutamate receptors. The best characterized is the nicotinic acetylcholine receptor. This complex is composed of five membrane-spanning units (a, a, ß, ?, and d), each having four transmembrane domains, which associate to form a central cation-conducting channel.

The last group is by far the largest group of gastrointestinal hormone receptors and the largest group of receptors in the body—the heptahelical G protein–coupled receptors. [99], [100] and [101] Several thousand of these have been cloned to date, representing 1% to 5% of the genome of different species. They are activated by extraordinarily diverse ligands, ranging from odorants to photons, peptides, and even large glycoproteins and viral particles. There is a somewhat parallel organization of groups of these receptors with structurally similar groups of ligands. [29] These receptors all initiate signaling by coupling with heterotrimeric G proteins. [102], [103] and [104]

The membrane protein tyrosine kinases have extracellular ligand-binding domains and intracellular kinase domains. [92] On activation, these molecules associate into dimers and are autophosphorylated. In addition to the phosphorylation of the receptors themselves, various members of this group phosphorylate and activate phospholipase C, GTPase-activator proteins, and intracellular tyrosine and serine/threonine kinase cascades. These events are closely associated with cell growth, differentiation, and oncogenesis.

The ligand-gated channels are multimers that comprise the same or related subunits and that cross the plasma membrane multiple times. [98] Proteins with four transmembrane domains are the most common subunit structure. The association of these proteins establishes a channel or a water-permeable pore. Ligand binding results in a conformational change, which regulates the size and permeability of the channel.

The heptahelical receptor proteins typically have no intrinsic enzymatic activity. Instead, they signal by associating on their cytosolic face with heterotrimeric G proteins. [102], [103], [104] and [105] These can then regulate a variety of effectors, such as phospholipase C and adenylate cyclase. These receptors have seven segments of approximately 20 to 25 hydrophobic residues representing the putative transmembrane helices (Fig. 4-8). The most highly conserved residues in each family of G protein–coupled receptors are located in these domains. Typically, there are sites of potential N-linked glycosylation in predicted ectodomains. This posttranslational modification likely plays an important role in helping the receptors fold and maintain their solubilities during biosynthesis. There are often serine- and threonine-rich areas in predicted cytosolic domains of these receptors that may be sites for phosphorylation and, thereby, receptor regulation.

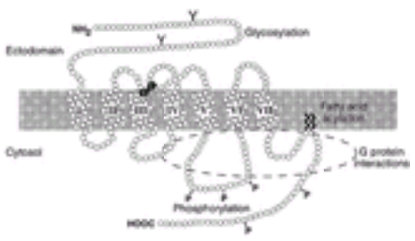

FIGURE 4-8. A prototypical G protein–coupled receptor depicted from the side (**top**) and looking down on the transmembrane helices (**bottom**). Each receptor is predicted to have seven hydrophobic segments traversing the lipid bilayer, with most having sites of N-linked glycosylation in the ectodomain and sites of phosphorylation in cytosolic domains. Also shown is the position of a highly-conserved disulfide bond linking the second and third extracellular loops, and a position of fatty acid acylation that is present in the carboxyl-terminal tail of some of these receptors.

The heterotrimeric G proteins consist of three subunits—a, ß, and ?. [105], [106] The a subunit has intrinsic GTPase activity and is the site of guanine nucleotide binding. The ß and ? subunits remain bound together, binding to the GDP-bound a subunit as a paired unit. There is a cycle of agonist binding to the receptor, association of the GDP-bound G-protein heterotrimer, GTP displacement of GDP, dissociation of the GTP-bound a subunit from the ß? dimer, hydrolysis of the GTP, and reassociation of the GDP-bound a subunit with ß? (Fig. 4-9). Crystal structures have been solved for these G-protein complexes, providing insights into the molecular interactions between receptor and G protein and between G protein and effectors. Early attention was focused on the GTP-bound a subunit as the molecule that interacts with effector proteins to activate them. More recently, it has become clear that the ß? dimer also plays this role for many effector proteins. [107], [108] In mammals, genes for 16 a subunits, 4 ß subunits, and 7 ? subunits have been isolated. [106] The innumerable combination of these various subunits into heterotrimers contributes to the vast diversity of effects and regulatory properties of peptide hormones.

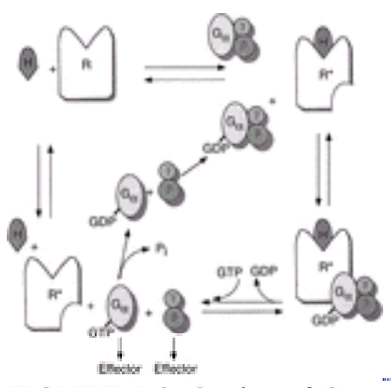

FIGURE 4-9. Cycles of G protein–coupled receptor activation and proximal signaling events at the level of the plasma membrane, which occur in response to agonist occupation. The earliest step is an induced conformational change in the receptor (*asterisk*) that facilitates its association with a G protein. At that point, the G protein is in its GDP-bound trimeric state. The receptor association facilitates GTP exchange for the GDP, which in turn results in the dissociation of the ternary complex and dissociation of the GTP-bound a subunit from the ß? dimer. Both of these can move on to interact with effectors in other compartments within the cell. The a subunit of the G protein has intrinsic GTPase activity that results in the elimination of one of the phosphates and reassociation of the trimeric form of the G protein.

The G protein–coupled receptors relevant to the gastrointestinal hormones fall into two families. Most of the small and carboxyl-terminally amidated peptides, which have focused domains for receptor binding at the carboxyl terminus, are members of the rhodopsin/ß-adrenergic receptor family. The larger peptides with diffuse pharmacophoric domains with amino-terminal determinants of specificity tend to be members of the secretin receptor family [109], [110] (Fig. 4-10). A notable exception is the amino terminal–specific receptor for motilin, which has been cloned and identified as a member of the rhodopsin/ß-adrenergic receptor family. [111] The binding of most agonists acting through this superfamily of receptors is influenced by hydrolysis-resistant analogs of GTP. This property provides an important clue that an action is mediated by this class of receptors.

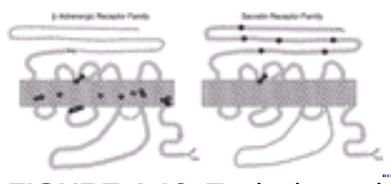

FIGURE 4-10. Typical members of the rhodopsin/ß-adrenergic receptor family and the secretin receptor family of G protein–coupled receptors. The typical signature sequences of the rhodopsin/ß-adrenergic receptor family are not present in the secretin receptor family. Other major differences include a shorter, third intracellular loop and a long amino-terminal tail that includes six highly conserved cysteine residues in the secretin receptor family.

Like the hormones, the gastrointestinal hormone receptors are all the products of single genes. Most do not have signal sequences, and they do not undergo extensive proteolytic processing. Alternate splicing is particularly common in the secretin family of G protein–coupled receptors, in which multiple exons are the rule. [112] The alternatively spliced products can differ substantially in ligand-binding specificity and activation characteristics. [113] The genes encoding the gastrointestinal hormone receptors have been known for less time than those encoding the hormones themselves. As a result, less is known about regulation of the receptor genes. Many receptors undergo substantial posttranslational modifications. Most of the G protein–coupled receptors are glycosylated. This feature helps to establish the appropriate folding and solubilities for biosynthetic transport to the cell surface, and likely helps to protect the receptors from proteolysis. [114], [115] and [116] Palmitoylation of Cys residues within the carboxyl-terminal tail is common in many G protein–coupled receptors. [117], [118], [119] and [120] This modification may bring another loop up to the plasma membrane, and may have a role in regulating access for G-protein coupling. Receptor phosphorylation is another common posttranslational modification, but it occurs predominantly after the mature receptor has been inserted into the plasma membrane. [121], [122] and [123]

Most of the gastrointestinal hormone receptors are present on the cell surface in small to moderate numbers of copies. There are typically fewer than several thousand molecules per cell. In such low numbers and given the very low concentrations of most of the circulating natural ligands, high affinity and specific binding interaction between hormone and receptor are critical. The receptor is an ideal target for regulating the sensitivity of the system. The pharmaceutical industry recognizes this and targets numerous drugs to act at cell surface receptors. Receptors are also a site of substantial endogenous regulation. A shift in the accessibility of the receptor-binding domain to ligand or a reduction in the number of receptors on the cell surface affects the sensitivity of the signaling system.

Receptors are dynamic molecules that have a life cycle in the cell (Fig. 4-11). Most receptors are synthesized at a slow rate, relying on reversible regulatory processes that conserve receptor molecules. The resting cell has a fixed complement of mature receptors on its surface and a small number of molecules in the biosynthetic pathway within the cell. Agonist occupation often initiates many events that can influence the receptor. By definition, agonist occupation of every receptor

initiates signaling processes within the cell. The details of these events vary with the receptor class, family, and even the particular ligand being bound or the cell in which the receptor is expressed. Within many signaling cascades are protein kinases and phosphatases that feed back and act on the receptor itself. For G protein–coupled receptors, this can uncouple the receptor from its proximal G-protein effector. [12] For receptor tyrosine kinases, autophosphorylation is critical. Many receptors undergo internalization processes that remove them from the surface, thus removing the potential for further stimulation. [10], [11] This mechanism protects the cell from damage caused by overstimulation. Some internalized receptors travel to the lysosome and are degraded, whereas others are recycled to the cell surface. For many ligand-occupied receptors, the pair of molecules internalize together and move through the proximal endocytic cascade, in which the endosome becomes acidified and releases the ligand. The ligand can then move independently from its receptor. To be certain of the details of these processes, both ligand and receptor must be studied.

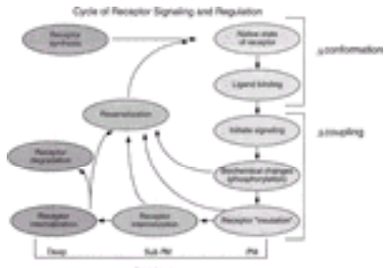

FIGURE 4-11. Events in the life cycle of a receptor in a cell. In the basal state, most of the receptor is usually at the level of the plasma membrane (*PM*). Agonist occupation results in changes in receptor conformation, biochemical modifications, and trafficking through the cell, which impact on accessibility for ligand binding, proximal effector coupling, and signaling events. The cycle is completed by reversal of the biochemical changes and the return of the receptor to the original cellular compartment. Because some receptor is lost to degradation, new synthesis also can replenish the cellular complement of receptor. The details of these cycles differ depending on the ligand, receptor, and cell type.

RECEPTOR CHARACTERIZATION AND QUANTIFICATION

Receptors can be characterized on the basis of their biologic activities or their receptor-binding properties. The availability of analogs of a given hormone that have a spectrum of activities from full agonist to partial agonist to antagonist simplify the characterization of biologic activity. If an activity is mediated by a particular receptor, the receptor antagonist should inhibit it in a competitive manner. Such an activity assay can be used to screen for compounds that are active at a particular receptor. This type of assay is necessary to determine if a new receptor ligand is an agonist or an antagonist.

The binding characteristics of receptors have been the subject of intense investigation. [124] The availability of radiolabeled forms of hormones with highly specific radioactivities that bind normally to their receptors provides a tool to analyze binding. The law of mass action should apply to the binding. Numerous software programs are capable of analyzing the binding data generated from this type of experiment. For such data and analysis to be valid, binding must be performed under conditions in which the concentration of radioligand is below the dissociation constant (K_d) and in which binding is permitted to reach steady state or equilibrium. The method of separating bound and free ligand should be rapid and efficient. The saturability of the binding is a critical feature that focuses on biologically relevant binding of appropriate affinity.

Biochemical characterization of receptors can be performed by affinity labeling. [24], [125], [126] This represents a variant of a receptor-binding study in which the specifically bound radioligand is covalently bound to the receptor molecule and subsequently purified and has provided many important insights into the nature of the receptor molecules. It also represents the only way to study posttranslational modifications of the receptor. By far the most valuable insight into biochemical characterization of receptors has come from cDNA cloning. Several powerful methods provide primary structural information for many gastrointestinal hormone receptors. With the cDNAs of many receptors already cloned, more receptors can be identified by hybridization screens and polymerase chain reaction by using probes designed to recognize domains conserved within families of receptors. Subtypes of receptors and alternatively spliced products can be identified by screening cDNA libraries with probes based on the full-length receptor cDNA. Improved receptor purification techniques facilitate the determination and use of specific sequence information. Expression cloning strategies are powerful and widely applicable to all classes of receptors.

Post-receptor signaling is an independent topic that is discussed in detail in Chapter 15. Such signaling is complicated and may be interdependent. It will undoubtedly become more complex before we understand enough to simplify. The view of signaling as a direct vectorial series of events is overly simplistic. There are numerous points of branching, feedback regulation, and subcellular translocalization at which kinases, phosphatases, lipid metabolites, and many other molecules affect the sensitivity of other molecules and other pathways. The phosphorylation events alone, which are activated by cell stimulation, number in the hundreds or thousands. Themes and rules will develop with increased understanding. It is now clear that many G protein–coupled receptors also stimulate tyrosine phosphorylation events [127] and can influence cell growth and differentiation, as well as the rapidly regulated events normally attributed to such messengers. The tremendous level of cross talk and cross regulation between signaling pathways provides extraordinary opportunity for cell-specific regulation.

HORMONE-SPECIFIC INSIGHTS

Gastrin-Cholecystokinin Family

The gastrin-CCK family of peptides is composed of the various molecular forms of the mammalian polypeptides gastrin and CCK. All share the same carboxyl-terminal pentapeptide-amide (Fig. 4-12). Evolutionary studies of gastrin and CCK suggest a common precursor peptide, with these hormones diverging late in evolution. [128], [129] and [130] Because the biologic information in both is highly localized to relatively small pharmacophoric domains at their carboxyl terminus, various different-length molecular forms of each share these domains. These forms provide an opportunity for differential regulation, clearance, and activity. Both gastrin and CCK have well-established physiological actions.

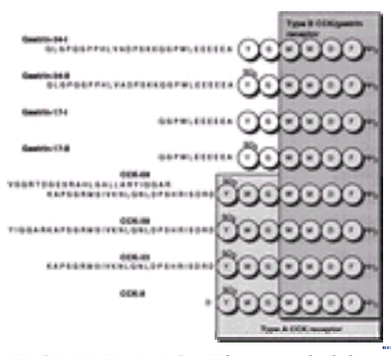

FIGURE 4-12. The variable molecular forms of gastrin and cholecystokinin (*CCK*). All share the same carboxyl-terminal pentapeptide-amide. Shown are the critical domains for recognition by type A CCK receptors and type B CCK/gastrin receptors.

Gastrin The major biologically active forms of gastrin are 17– and 34–amino acid peptides that have a tyrosine residue six residues from the carboxyl terminus, which may (gastrin-17-II, gastrin-34-II) or may not (gastrin-17-I, gastrin-34-I) be sulfated. This posttranslational modification of gastrin appears to have no functional significance, all forms being equally potent and efficacious at traditional gastrin receptors. The critical information in gastrin peptides is in the carboxyl-terminal tetrapeptide-amide, with most extensions and modifications of that domain well tolerated. Gastrin-17 is cleared from the circulation faster (half-life of 7 minutes) than gastrin-34 (half-life of 30 minutes). [78] This results in the situation that most gastrin in the circulation during fasting is gastrin-34, whereas the major form released after

a meal is gastrin-17. The greatest proportion (>95%) of gastrin in the normal circulation is fully processed and contains a carboxyl-terminal phenylalanine-amide. [131] Within some tissues, gastrin can be produced, but not processed, resulting in the presence of nonamidated and carboxyl-extended forms of this peptide. Also, in gastrin-producing neoplasms, processing is often incomplete, resulting in the release of large quantities of nonamidated forms into the peripheral circulation. [131] This has even been proposed as an assay for such neoplasms. [72] The major site of gastrin expression in the adult is open endocrine cells (G cells) in the gastric antrum. Low levels of gastrin are present in the vagus nerve and in pituitary corticotrophs, and transient expression is observed in fetal pancreatic islets and colon. [44], [132] This hormone is found in a number of neoplasms. [133], [134] The highest levels of processed and autonomously secreted gastrin come from islet cell tumors (gastrinomas) in the Zollinger-Ellison syndrome [135] (see Chapter 67). It has also been reported in unprocessed forms in some colon, lung, and ovarian cancers. [133], [134] Excellent radioimmunoassays for gastrin are widely available. Specialized assays for unprocessed forms of this hormone also have been developed. Gastrin secretion from antral G cells is regulated by lumenal, paracrine, endocrine, and neural stimuli. [136] Ingestion of a meal is a strong stimulant of the secretion of this hormone. Small peptides, aromatic amino acids (e.g., phenylalanine and tryptophan), and calcium in a meal are key contributors to its stimulatory effect. Coffee, wine, and beer also have been reported to be stimulants, but the ethanol and caffeine components are not major contributors. Of interest, carbohydrate and fat components of a meal contribute little to the gastrin response. There is an important feedback loop, whereby the major result of gastrin stimulation, acid secretion, results in lowered intragastric pH and reduced G-cell secretion of gastrin (Fig. 4-13). This is mediated by the paracrine effect of somatostatin. The neurocrine mediator GRP stimulates G-cell secretion. Sympathetic innervation and parasympathetic innervation have complex effects. Sham feeding and gastric distention both stimulate gastrin release, and atropine actually enhances this effect.

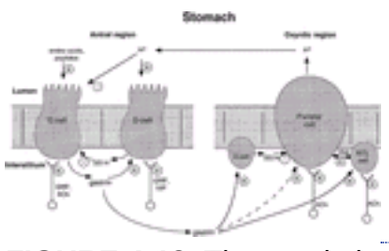

FIGURE 4-13. The regulation of gastric acid secretion by gastrin represents a complex series of interactions and feedback. Some of the key components of this cycle are stimulatory events (+) and inhibitory events (-). Acetylcholine (*ACh*) stimulates the ECL (*enterochromaffin-like*) cell, the parietal cell, and the gastrin-producing G cell. Gastrin-releasing peptide (*GRP*) stimulates both the G cell and the D cell. Gastrin stimulates the D cell, the parietal cell, and also the ECL cell, which then secretes histamine (*Hist*) also to stimulate the parietal cell. Somatostatin (*SS14*) secreted by the D cell inhibits both the G cell and the parietal cell. Acid (H^+) secreted by the parietal cell has regulatory effects as well. *VIP,* vasoactive intestinal polypeptide.

The classic target for gastrin is the gastric parietal cell, which secretes hydrochloric acid. This effect is mediated by a G protein–coupled receptor, which is identical to the most abundant CCK receptor present in the central nervous system (i.e., type B CCK receptor). This receptor is in the rhodopsin/ß-adrenergic receptor family and closely resembles the type A CCK receptor. This is particularly interesting, given the likely shared evolutionary relationship between gastrin and CCK. The receptors also probably evolved from a common precursor, with divergence at some point. It is not yet clear whether the hormones or receptors diverged first, or how the relationship between the molecular basis of binding and activation might compare at these two receptors. Both receptors are coupled with Gq/11. The structural specificity of ligand binding to these two receptors is quite distinct [137], [138] (Table 4-2). The type B CCK/gastrin receptor requires only the carboxyl-terminal tetrapeptide-amide for high-affinity binding and activation. It recognizes all forms of gastrin and CCK almost equally (CCK-8 = gastrin = CCK-8-desulfate = CCK-4). In contrast, the type A CCK receptor has more stringent requirements. At that target, the carboxyl-terminal heptapeptide-amide of CCK is required, with the sulfated tyrosine being critical for high-affinity interaction (CCK-8 >> gastrin = CCK-8-desulfate > CCK-4). Gastrin is recognized poorly, being more than 1000-fold less potent and binding with similarly low affinity.

	APPROXIMATE LIGAND AFFINITIES (nM)	
Ligand	*Type A CCK Receptor*	*Type B CCK/ Gastrin Receptor*
CCK-8	1	2
CCK-8-desulfate	500	10
CCK-4	10,000	10
Gastrin-17	1000	10
L-364,718	1	100
L-365,260	300	2

TABLE 4-2 Specificity of Cholecystokinin (CCK) Receptors

The type B CCK receptor is also expressed on enterochromaffin-like (ECL) cells of the gastric mucosa, on smooth muscle cells at various levels along the gastrointestinal tract, and on diffuse domains of the brain. Experimental antagonists acting at this receptor (L-365,260) have been reported to inhibit gastric acid secretion. [36], [139] However, this result also can be accomplished by a number of other pharmacological routes, such as the use of histamine H_2 receptor antagonists or hydrogen-potassium ATPase inhibitors, which are already in wide clinical use. The most useful application for gastrin receptor antagonists may be for their neuronal effects, which reduce anxiety and panic attacks. [140] As noted, a major physiological effect of gastrin is the stimulation of gastric acid secretion. Of interest, isolated parietal cells respond weakly to gastrin alone. [141], [142] This response is augmented in the presence of histamine or acetylcholine, both of which provide potentiating interactions. Thus, the action of gastrin in vivo to stimulate its receptor on the ECL cell and thereby stimulate histamine release helps provide the optimal background for a brisk acid secretory response. Gastrin also has been shown to stimulate histidine decarboxylase, a key enzyme in histamine synthesis. In addition to its classic acute roles in stimulating acid and histamine secretion, gastrin also appears to function as a growth factor to stimulate mucosal proliferation. [143], [144] and [145] Its role as a growth factor may be unique, with different tissue-specific expression during development and in adult life. During the fetal period, gastrin is expressed in pancreatic islets and in colonic epithelium. Islet expression ends at birth as these cells undergo terminal differentiation. The trophic effect on the gastric oxyntic epithelium has been suggested by the hyperplasia associated with the Zollinger-Ellison syndrome of hypergastrinemia and by the atrophy associated with low levels of gastrin after antrectomy. Chronic high-level gastrin stimulation also has been correlated with enterochromaffin cell hyperplasia. [146] Additionally, in gastrin receptor–deficient mice generated by gene targeting (i.e., receptor knock-out animals), the gastric mucosa was remarkably atrophic, even in the presence of hypergastrinemia. [147] In humans, more than 95% of gastrin peptides normally secreted are fully processed and amidated; the percentage of Gly-extended forms is small. Substantial alterations in gastrin processing have been observed in several pathological states. [148], [149] and [150] These include the increased synthesis that occurs with achlorhydria and with some neoplasms, such as gastrinomas and colonic carcinomas. It has been postulated that Gly-extended gastrins may act as growth factors. [81], [151], [152] and [153] Such forms are several orders of magnitude less potent at stimulating acid secretion, but they may be equipotent or even more potent than processed gastrin in some growth activities. The growth-stimulating actions of Gly-extended gastrin have been reported in the pancreatic tumor cell line AR42J [81], [154] and in Swiss 3T3 cells. [155] Both reports support actions independent of the classic CCK-B receptor. A report of a misspliced form of this receptor as being constitutively active in some colon carcinomas may explain some of these observations. [156] If there is a new and distinct receptor that mediates this activity, it will likely not be structurally related to the type A or type B CCK/gastrin receptors. No reports as of yet demonstrate this action in vivo, and the postulated relationship between gastrin and colon cancer remains unclear.

Cholecystokinin Like gastrin, CCK is found as diverse forms sharing their carboxyl-terminal heptapeptide-amide. Unlike gastrin, which has sulfated and unsulfated forms, all the CCK in the normal circulation is sulfated. This is consistent with the structure-activity relationship described above. Although CCK-8 and possibly CCK-33/39 have previously been thought to be the major forms of this hormone in the circulation, CCK-58 and possibly CCK-83 may be more prominent. [157], [158] It is difficult to measure these very large forms because they are readily cleaved to smaller forms and because little standardized peptide has been available for characterization and validation of assays. A recent report again claims that CCK-33 is the major circulating form. [159] No differential biologic effects of the molecular forms of CCK have been described. Much less is understood about the proteolytic processing of CCK than of gastrin. Although the processing enzymes are likely analogous, the order of cleavages and the mechanisms for tissue-specific processing are not understood. Like gastrin, the various molecular forms are cleared from the circulation at different rates. The small form, CCK-8, has a half-life of only about 1 minute and is cleared almost quantitatively in a single pass through the portal circulation. [3], [160], [161] Larger forms are not cleared by the liver but seem to be metabolized by various capillary beds and by the kidney. CCK is predominantly produced in endocrine I cells scattered throughout the proximal two thirds of the small intestine. [162] It is also found in less abundance in various enteric neurons and in pancreatic islets. [162] CCK-secreting endocrine neoplasms have not been well documented in the literature. The brain contains a large amount of CCK, but the regulation of peptide synthesis and release in the brain is poorly understood. Both protein and fat components of meals stimulate secretion of CCK from I cells; carbohydrates are ineffective. [163] Long-chain triglycerides and fatty acids, both aromatic and aliphatic amino acids (e.g., phenylalanine, tryptophan, valine, and methionine), and small peptides can all stimulate CCK secretion. Measurement of CCK is not as routine as that of gastrin. CCK appears to be present in the circulation in much lower concentrations (basal values < 1 pM CCK versus 100 pM gastrin). Also, because the most common carboxyl-terminal–directed antibodies

can cross-react with both hormones, a selective radioimmunoassay is difficult to establish. A few radioimmunoassays are used in research that incorporate antisera directed against the amino-terminal domain of CCK-8, which are sensitive to the tyrosine-sulfate. [159], [164], [165] The more common assay for CCK uses the ability of the type A CCK receptor to distinguish CCK from gastrin. This is a bioassay in which partially purified peptide is used to stimulate dispersed pancreatic acini to secrete amylase. [166] Feedback regulation also is involved in CCK secretion [167], [168] and [169] (Fig. 4-14). The major targets of this hormone are the rodent pancreatic acinar cells and human pancreatic neurons. When pancreatic exocrine cells are stimulated to release digestive enzymes that make their way into the lumen of the intestine, CCK secretion is reduced. Lumenal administration of a trypsin inhibitor results in increased levels of CCK and increased pancreatic enzyme secretion. Bile acids in the lumen also appear to have a regulatory effect on this feedback phenomenon. [170] Two candidates for representing endogenous CCK-releasing factors have been described. [171], [172] Both have their activities inhibited by trypsin and fulfill the initial expectations of such a mediator, but additional studies will be necessary to define their potential roles in normal physiology.

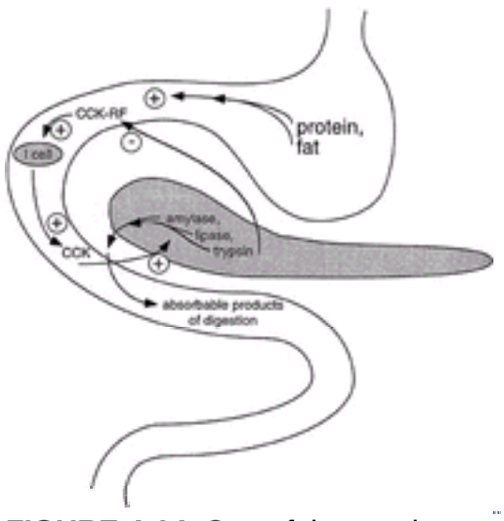

FIGURE 4-14. One of the regulatory cycles for CCK action on the pancreatic acinar cell. Nutrient ingestion results in the release of CCK-releasing factors (*CCK-RF*) into the intestinal lumen. These act on the I cells to secrete CCK, which stimulates pancreatic exocrine secretion. The proteolytic enzymes (particularly trypsin) then degrade the CCK-releasing factors, thereby terminating the cycle.

The type A CCK receptor is structurally similar to the type B CCK/gastrin receptor (50% identical, 66% homologous). [27], [28], [173] It is a member of the rhodopsin/ß-adrenergic receptor family. This receptor has substantial structural specificity, recognizing the carboxyl-terminal heptapeptide-amide of CCK, with a clear requirement for the tyrosine-sulfate, the carboxyl-terminal phenylalanine-amide, the nonoxidized state of the methionine residues, and the residues in almost every position of this peptide. Only the amino terminus is freely available for extension or modification without interference with the binding and biologic activity of CCK. Understanding of the molecular basis of binding the natural agonist peptide to this receptor has improved substantially, with direct evidence of contacts in the extracellular loop and tail domains. [24] Nonpeptidyl antagonists (e.g., L-364,718, lorglumide, PD140548) and even a nonpeptidyl agonist are available for selective action on this receptor. [174], [175] None of these reagents have clinical use. The antagonists induce gallbladder stasis; their use, therefore, is complicated by gallstone formation. The agonist could provide a new mechanism to achieve satiety, but no clinical studies have been reported. CCK is interesting for the diversity of its established physiological and biologic effects. [176] Almost all its actions relate to nutrient homeostasis. Its major digestive effects are to stimulate pancreatic acinar cell secretion of zymogens and to stimulate gallbladder muscularis smooth muscle cell contraction. Both of these events result in the delivery of key ingredients for digestion to the intestinal lumen. It also effects contraction of the pylorus to slow gastric emptying, providing optimal nutrient delivery for digestion. A key neuronal effect of this hormone is to satiate appetite. Less well-established effects include modification of enteric and colonic transit, reduction in acid secretion by its effect as a somatostatin secretagogue, stimulation of secretion of pancreatic polypeptide, and an insulinotropic effect.

Pancreatic Polypeptide Family

The pancreatic polypeptide family includes pancreatic polypeptide (PP), peptide YY (PYY), and neuropeptide Y (NPY). These peptides have extensive homology within their carboxyl-terminal domains, with absolute conservation of an arginine-tyrosine–amide. PYY and NPY have extensive overall homology with each other. These three peptides also have cross recognition of a group of three receptors. Each peptide has a unique distribution and unique functions.

Pancreatic Polypeptide PP was originally identified as a contaminant in preparations of insulin extracted from pancreatic islets. [177] It is a 36–amino acid peptide that is predicted to have a globular shape, resulting from interactions between a polyproline helix and an a helix. [178], [179] Its biologic activity resides in its carboxyl-terminal hexapeptide-amide, with the carboxyl-terminal tyrosine-amide being the critical residue for this activity. It has a short half-life in the circulation (about 7 minutes), with renal clearance apparently most important. PP cells are found in the periphery of pancreatic islets and scattered within the parenchyma of the pancreatic head and uncinate lobe, in contrast to the distribution of glucagon in the pancreatic body and tail. There is a clear PP response to the ingestion of a meal. The response is biphasic; the early increase is neurally mediated and inclusive of cephalic and vagal phases. In many ways, secretion of PP correlates with vagal tone. The nutrients that stimulate gastrin and CCK secretion also stimulate PP secretion. Secretion of this hormone also appears to be stimulated by CCK, gastrin, GRP, neuromedins B and C, and secretin. Although several biologic effects of PP have been proposed, debates continue. [180] This hormone has been thought to inhibit pancreatic exocrine secretion, but antibody neutralization studies have failed to show any effect on pancreatic secretion. A metabolic role as an inhibitor of hepatic glucose production has also been proposed. A series of receptor subtypes that recognize this family of peptides have been confirmed, with the revised nomenclature including the PP1 receptor and NPY receptor subtypes Y1 through Y6. [181], [182] All are structurally related members of the rhodopsin/ß-adrenergic receptor family of the G protein–coupled superfamily of receptors. The PP1 receptor recognizes pancreatic polypeptide better than either of the other family members (PP >> NPY = PYY). Both the Y1 and Y2 receptors recognize PYY and NPY similarly, and much better than PP (PYY = NPY >> PP). There are selective agonists that distinguish between these receptors, with (Leu ^{31}, Pro ^{34})PYY and (Leu ^{31}, Pro ^{34})NPY selective for the Y1 receptor, and carboxyl-terminal fragments (PYY-3-36 and PYY-13-36, NPY-13-36) selective for the Y2 receptor. [183] Much less has been established for the Y4, Y5, and Y6 receptor subtypes. Perhaps the best current use of PP is as a marker for islet cell tumors. Plasma levels have been elevated in a high percentage of islet cell carcinomas and multiple endocrine neoplasia. The percentage of positive values varies with the type of endocrine tumor; VIP-secreting tumors have very high levels, and gastrin-secreting tumors have substantially lower levels.
PYY PYY is also a 36–amino acid peptide. It is found in highest concentrations in enteroendocrine cells in the distal small intestine and colon, [184], [185], [186] and [187] mainly in the basal crypts of L cells. [188], [189] It can act through endocrine mechanisms after release into the bloodstream, or through paracrine mechanisms with specialized basal processes surrounding columnar cells in the vicinity of the endocrine cells. It is released in response to meals. Fat is the major stimulant, but other nutrients (e.g., carbohydrate) and bile acids also stimulate secretion. Neuronal release mechanisms also have been postulated, and in some malabsorptive states, high circulating levels have been observed. GRP also can stimulate its release. Like PP, PYY has a short half-life in the circulation. The actions of PYY are largely inhibitory. [190], [191] It inhibits gastrointestinal motility, pancreatic and gastric secretion, and chloride secretion. It has been given the name *ileal brake* because it helps to establish longer contact times when nutrients reach the distal gut and colon. PYY has also been shown to have trophic effects, but the physiological significance of this action is not yet clear.
NPY NPY is also a 36–amino acid peptide that is structurally homologous with PYY. [192], [193], [194] and [195] It appears to be the neurotransmitter equivalent of PYY, found in the central and peripheral nervous systems. It is one of the most abundant peptides in the brain, along with CCK. [196] It is found in enteric neurons, alone or co-localized with other neuropeptides. [197], [198] and [199] It is especially prominent in sympathetic neurons that innervate blood vessels. Postulated effects of NPY include the stimulation of appetite, alteration of circadian rhythms, and vasoconstriction. [200], [201], [202] and [203] It has many of the same effects as PYY because they both recognize the same receptors, but its physiological role relates mainly to its anatomic distribution.

Tachykinin Family

The tachykinins are a group of biologically active peptides sharing the carboxyl-terminal dipeptide-amide Leu-Met-NH_2. Included in this group are substance P, GRP, the neuromedins, and many mollusk and amphibian peptides (bombesin, physalaemin, eledoisin, kassinin, and phyllomedusin). In this group of transmitters, as in the two groups discussed above, the pharmacophoric domain is predominantly within the carboxyl terminus and well circumscribed. Consistent with this feature, the receptors for this family also are in the rhodopsin/ß-adrenergic receptor family of the G protein–coupled superfamily of receptors.

Substance P (Neurokinin A) Substance P is an 11–amino acid peptide-amide that is widely distributed in the brain, spinal cord, and peripheral and enteric nervous

systems. [204] Its highest concentrations along the digestive tract are in neurons of the esophagus, proximal small intestine, and colon. Substance P stimulates esophageal and intestinal peristalsis and pancreatic secretion, and it inhibits biliary secretion and somatostatin secretion. It likely plays a sensory role along the digestive tract, participating in the afferent limb of various reflexes and possibly mediating pain impulses. In the central nervous system, it probably acts as a neuromodulator.

Gastrin-Releasing Peptide GRP was originally isolated during a search for an analog to the amphibian peptide bombesin. [205], [206] and [207] Like bombesin, GRP stimulates gastrin secretion. It is present in neurons of the stomach, intestine, and colon, as well as in the brain and spinal cord. It also stimulates the release of CCK, motilin, neurotensin, enteroglucagon, insulin, somatostatin, and substance P, and it can stimulate pancreatic acinar cell secretion and gut motility. The release of secretin from S cells is not stimulated by GRP. It also may play a role as an excitatory neurotransmitter, acting on enteric interneurons. Like several of the peptides discussed, GRP appears to have trophic activity. Receptors for this peptide have been described on small cell carcinomas of the lung, where a trophic response may have clinical significance. [208], [209] Several of the neuromedins were isolated during a search for mammalian homologs of the amphibian peptides. Neuromedin C shares its carboxyl-terminal decapeptide-amide with GRP. Neuromedin B is related to ranatensin. Four receptor subtypes are now recognized for this class of ligands. [210] They are currently termed *BB1, BB2, BB3,* and *BB4*. The BB1 receptor recognizes neuromedin B with highest affinity (neuromedin B = bombesin > GRP). The BB2 receptor recognizes GRP best (GRP = bombesin >> neuromedin B). The BB3 receptor has a high degree of homology with the other two but binds both GRP and neuromedin B with low affinities. Its natural ligand has not yet been identified. Its presence has been reported in reproductive organs and lung carcinoma; it may not have a relationship to the gastrointestinal tract. The BB4 receptor was isolated from frog brain and has a unique pharmacological profile—it is highly dependent on the penultimate phenylalanine residue for agonist activity, and it has a unique capability for antagonist recognition. [211]

Somatostatin Family

The two molecular forms of somatostatin are 28 and 14 amino acids in length, with somatostatin-14 representing the carboxyl-terminal domain of somatostatin-28. Both forms of somatostatin contain a critical disulfide bond giving them a cyclic structure [212], [213] and [214] (Fig. 4-15). The amino acid residues 7 through 10 within the short cyclic structure of somatostatin-14 are also critical for activity. Although this hormone was originally identified for its ability to inhibit growth hormone, it is now recognized as having much broader significance.

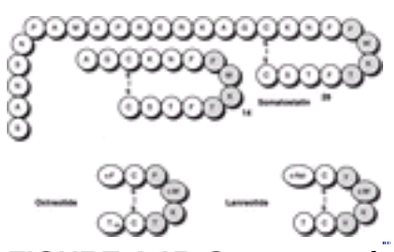

FIGURE 4-15. Structures of somato-statin-14 and somatostatin-28, as well as two prominent synthetic agonist analogs, octreotide and lanreotide, that incorporate unnatural amino acids and terminal blocking groups, prolonging biologic action.

Somatostatin is synthesized and secreted from both neurons and enteroendocrine cells, with the latter (D cells) present as either open or closed. [215], [216] In the stomach, there are highly specialized D cells with characteristic long processes that extend to G cells, parietal cells, and chief cells. [217] These configurations make somatostatin one of the most flexible regulators of the gastrointestinal tract. Its multiple roles as hormone, paracrine transmitter, neurotransmitter, and neuromodulator reflect this flexibility. Along the digestive tract, 90% of somatostatin is present in the mucosa and 10% in the neuromuscular layers. In the stomach, pancreas, and neuromuscular layers of the intestine, somatostatin-14 is the predominant form, whereas both somatostatin-14 and somatostatin-28 are found in the mucosa of the bowel. Somatostatin-14 has a very short half-life of 1 to 3 minutes, whereas somatostatin-28 has a half-life of about 15 minutes. The degradation sites of this hormone are not well defined, but they likely involve capillary beds.

Most of the biologic effects of somatostatin are inhibitory. It inhibits gastric, pancreatic, biliary, and even salivary secretion, in addition to inhibiting the release of a broad variety of gastrointestinal hormones. These include gastrin, CCK, secretin, pancreatic polypeptide, GIP, motilin, glucagon, and insulin. It also inhibits gut motility, but this effect results from the peptide's ability to inhibit cholinergic neurons. Somatostatin decreases splanchnic and portal blood flow.

Somatostatin receptors are in the rhodopsin/ß-adrenergic receptor family of the G protein–coupled superfamily of receptors. There are five recognized receptor subtypes: SSTR1, SSTR2, SSTR3, SSTR4, and SSTR5. [218], [219] and [220] These receptors are structurally homologous, but each couples to distinct G proteins and has a distinct tissue distribution. They are most closely related to the opioid receptors, with which they share about 40% homology. Based on structural similarity, there are two groups of somatostatin receptors: SSTR1 is related to SSTR4, and SSTR2, SSTR3, and SSTR5 are related to each other. SSTR5 is unique among the somatostatin receptors because it has a higher affinity for somatostatin-28 than for somatostatin-14. SSTR1 is found in the stomach and intestine. These receptors most prominently couple to adenylate cyclase; some receptors also activate potassium channels and voltage- and potassium-dependent calcium channels. The somatostatin receptor also may activate a protein tyrosine phosphatase, although the precise molecular mechanism for this is unclear.

An analog of somatostatin that is a long-acting cyclic agonist peptide has become a highly useful clinical reagent. It represents an 8–amino acid cyclic peptide with unnatural residues that make it resistant to proteolysis. It has been used in the treatment of gastrointestinal hormone-secreting tumors, diarrheal disorders, and carcinoid syndrome. [221], [222], [223], [224] and [225]

Motilin Family

Motilin is a 22–amino acid linear peptide that was originally recognized as being secreted into the circulation in a cyclic manner that correlated with increased motor activity along the bowel. It has become one of the most important endogenous prokinetic peptides, and its receptor is recognized as a useful pharmacological target, activated by erythromycin. [226] Despite extensive insight into the structure-activity relationships for the motilin receptor, [227] it has been difficult to isolate and characterize biochemically. This may reflect the substantial interspecific differences in motilin peptides. The most highly conserved sequence resides in the amino-terminal portion of the peptide, and the pharmacophoric domain has been localized to the amino-terminal decapeptide region. [228]

Finally in 1999, with the use of a unique high-throughput screen of compounds that could interact with cloned orphan receptors, the cDNA for the motilin receptor was identified. [111] Sequence analysis showed that it was a member of the rhodopsin/ß-adrenergic receptor family of the G protein–coupled superfamily of receptors, having closest homology to a group of growth hormone secretagogue receptors. One extremely interesting aspect of studying the growth hormone secretagogue receptor was the discovery of ghrelin, an endogenous agonist ligand representing a 28–amino acid peptide that has a high degree of homology with motilin, and that has a unique posttranslational modification of N-octanoylation of the Ser residue in position 3 [229] (Fig. 4-16).

FIGURE 4-16. Alignment of sequences of motilin and ghrelin, members of the motilin family. Ghrelin has a unique posttranslational modification of the serine residue in position 3, representing N-octanoylation.

To date, only a single motilin receptor cDNA has been identified, even though there is pharmacological evidence for more than one pattern of responses to this

hormone. [230] Examination of the molecular basis of these varied responses will likely become an area of active exploration.

Protease-Activated Receptor Family

Proteases, such as trypsin, have traditionally not been thought of as signaling molecules. There is evidence, however, of distinct G protein–coupled receptors in the rhodopsin/ß-adrenergic receptor family of G protein–coupled receptors that are activated by proteolytic cleavage of an amino-terminal peptide sequence. [231] This makes an endogenous peptide ligand at the new amino-terminal end of the receptor accessible for binding to the remainder of the receptor, thus providing a novel molecular mechanism of action. To date, two such receptors have been identified—the thrombin receptor (or protease-activated receptor 1) [232] and the protease-activated receptor 2. [233] A receptor activation mechanism that requires proteolytic cleavage has substantial implications for receptor regulation, particularly for ensuring the inability for resensitization. [234], [235]

These receptors are believed to play roles in the linkage of tissue injury and vascular leakage to cellular responses, such as occurs in hemostasis, inflammation, and angiogenesis. [236] Roles have been postulated for these receptors in the intestine and pancreas as well.

Chemokine Family

The chemokines constitute an extremely large family of ligands and receptors, with biologic activities best described for leukocytes. Although not specific to the gastrointestinal tract, this system has great relevance to it. More than 40 distinct human chemokines have been described and assigned extremely varied original names based on the tissue or cell of origin, the target cell type, the size of the molecule, or the action. A more coherent nomenclature that correlates with the organization of the receptors for these ligands was proposed in 1999. [237] This system classifies chemokines by their structure, based on the number and the spacing of conserved cysteine residues, into four groups: C, CC, CXC, and CX $_3$C. The CC, CXC, and CX $_3$C cytokines have four conserved cysteine residues, with the first two such residues being adjacent to one another, or having one or three other residues between them, respectively. The C cytokines have only two conserved cysteine residues, representing the second and fourth cysteine residues that are present in the other groups of chemokines.

The chemokine receptors are in the G protein–coupled receptor superfamily. They typically recognize more than one chemokine, but these are usually members of a single chemokine family. The receptor nomenclature is based on the ligand specificity, starting with CC or CXC, and followed by R and a number. Of this group, 18 chemokine receptors have been cloned, and these express 25% to 80% sequence identity among themselves.

Another interesting feature of the chemokines and their receptors is the substantial diversity of structure across species, with up to 55% sequence divergence reported. The pressure for the rapidity in evolution of these structures has not been established. Also of note are the nonchemokine ligands for these receptors, which include HIV, in which the chemokine receptor seems to play an important role as a coreceptor for the virus, permitting its entry into the target cell. On tumor cells, these receptors may determine metastatic pattern.

Secretin Family

The secretin family of hormones is characterized by homology that is most evident in the amino-terminal half of moderately large polypeptides (Fig. 4-17). Members of this family with known hormone and receptor sequences include secretin, [25], [238] VIP, [239] pituitary adenylate cyclase–activating peptide (PACAP), GIP, [240] glucagon, glucagon-like peptide-1 (GLP-1), calcitonin, calcitonin gene–related peptide (CGRP), parathyroid hormone, [241] corticotropin-releasing factor (CRF), and growth hormone–releasing factor (GHRF). [242] Some of these are not relevant to the gastrointestinal tract and are not further discussed. It must be recognized, however, that insights from any members of this family may provide important clues relevant to the entire family. Based on the structural homology of hormones, it is expected that the PHI receptor also belongs to this family, although a distinct receptor for this peptide has not yet been cloned.

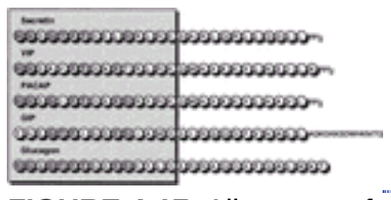

FIGURE 4-17. Alignment of sequences of members of the secretin family. The amino-terminal domain is most critical for agonist action, whereas the pharmacophore extends throughout the entire peptide sequence.

The pharmacophoric domains of these hormones are rather diffuse and spread throughout their primary sequences, usually requiring peptide analogs to span essentially the entire length of the natural hormones for maintenance of biologic activity. In some of these hormones, the amino-terminal residue and a free amino terminus are particularly important. This observation implies the interesting notion that amino-terminally extended precursor molecular forms may not be biologically active. In fact, few of the hormones in the secretin family have more than one molecular form, as is so common for peptide hormones belonging to the rhodopsin/ß-adrenergic receptor family. Some truncated forms of the secretin family hormones have been synthesized and found to continue to bind with high affinity, but these possess markedly reduced biologic activity. Another interesting feature of this family is that its members stimulate dual signaling cascades; low concentrations typically stimulate adenylate cyclase, and higher concentrations stimulate intracellular calcium responses. [243] This also may occur in the rhodopsin/ß-adrenergic receptor family, but it is less common.

Secretin Secretin holds a special place in gastrointestinal endocrinology and in endocrinology generally. Its discovery in 1902 by Bayliss and Starling [13] established the concept of hormones as chemical messengers that travel through the bloodstream to control the function of a target organ. As such, the discovery of secretin gave birth to the whole field of endocrinology. It was therefore the first gastrointestinal hormone, although not the first one to be characterized chemically. Secretin clearly has important physiological functions. Because of its relatively large size and the chemical difficulties encountered in working with this peptide and with its receptor, a detailed molecular understanding of this hormone has been delayed substantially. The recent cloning of cDNAs for members of this receptor family has charged this field with new life and excitement, and it promises to be a fertile area for investigation in the years ahead. Secretin is a 27–amino acid linear peptide with important residues scattered throughout the length of the hormone. [244] It is synthesized in endocrine cells (S cells) that are most abundant in the duodenum and proximal jejunum and present in reduced numbers throughout the distal small intestine. [245] The major stimulus for secretin secretion is unbuffered hydrogen ions as they traverse the duodenum. This stimulus becomes apparent when the duodenal pH decreases to 4 or lower. Secretin secretion also can be stimulated by nutritional acids, such as fatty acids, and by ethanol and components of spicy foods (e.g., 1-phenylpentanol). Bile acids appear to enhance stimulated secretin release. Of interest, the S cell is one of the few enteroendocrine cells that is not responsive to GRP. However, the existence of a secretin-releasing peptide has been postulated. [246] Like most of the peptide hormones, secretin has a short half-life of less than 3 minutes in the circulation. [247] The kidneys appears to be the major site of clearance. The major effect of secretin is the secretion of bicarbonate from biliary and pancreatic ductular epithelium and Brunner glands. [248], [249] and [250] This nicely completes the regulatory cycle, with the acid stimulant of secretin release neutralized by the bicarbonate secreted in response to this hormone. It has been estimated that secretin is responsible for 80% of the bicarbonate response to a meal. In nonhuman species, secretin also has been shown to inhibit gastric secretion and gastric motility, to lower esophageal sphincter tone, and to stimulate insulin release and colonic mucus secretion, but these are likely not physiological actions in humans. [251], [252] Consistent with these observations, the most prominent targets for secretin include ductular epithelial cells in the biliary tree and pancreas, and Brunner gland cells. [248], [249] and [250], [253] Receptors are also believed to be expressed on pancreatic acinar cells of some species, vascular smooth muscle in selected organs, some enteric smooth muscle (particularly in the region of the lower esophageal sphincter), and some enteric mucosal cells. [250], [254], [255], [256] and [257] These targets were initially characterized as having a potent biologic response to secretin, and a lesser response to VIP. Such studies are difficult to interpret because of the cross recognition of many peptides in the family by receptors in the family. Now that the structure of receptors is better understood, the expression of receptors on distinct cells has become easier to identify. The secretin receptor cDNA was initially cloned in 1991 by Ishihara and colleagues. [238] Its structure appeared to have the seven hydrophobic helical transmembrane domains typical of the G protein–coupled receptor superfamily, but other features of the receptor were unique. In fact, the level of homology with previously cloned receptors in the rhodopsin/ß-adrenergic family was only 12%. Subsequent cloning of cDNAs encoding receptors for calcitonin and

parathyroid hormone, [241], [258] which are more similar to secretin receptors, defined a new class of G protein–coupled receptors. The most highly conserved areas are in the transmembrane domains, and a particularly long amino-terminal tail contains six highly conserved cysteine residues that are highly sensitive to reducing and cysteine-reactive reagents. [259] Although the pattern of the disulfide bonds has not been experimentally defined, research supports the existence of three such bonds among these six residues. [260] Another such bond that is likely to be present is the connection between cysteine residues in the first and second extracellular loops, in positions that are conserved throughout the entire superfamily of G protein–coupled receptors. This bond has been characterized for other receptors in the rhodopsin/ß-adrenergic family. [261] Various studies including mutagenesis and photoaffinity labeling have demonstrated the importance of the amino-terminal tail and extracellular loop domains of this receptor for secretin binding and activation. [30], [31], [262], [263] This finding is consistent with the general theme for this superfamily, in which increasingly larger ligands bind to domains moving from the confluence of intramembranous helices to the cellular surface, and ultimately to the extracellular loop and tail domains. Despite having new signature sequences unique to the secretin receptor family, the secretin receptor seems to signal and to be regulated much like the receptors in the rhodopsin/ß-adrenergic family. [264] The secretin receptor is coupled to both Gs and Gq/11. The most potent response to hormonal stimulation is an increase in adenylate cyclase, and less potent is the stimulation of intracellular calcium. [243] The secretin receptor is phosphorylated in response to agonist stimulation, [264] and this phosphorylation interferes with G-protein coupling. Like most G protein–coupled receptors, this receptor is internalized after agonist occupation. [264] The details of the molecular basis of these events are being explored.

VIP VIP is a 28–amino acid linear peptide with close structural homology to secretin. [265] Conformational studies have supported similar structures as well. [266], [267] Unlike the hormone secretin, VIP is exclusively a neurotransmitter. In addition to being present in enteric neurons, VIP is also present in neurons of the brain, spinal cord, lung, urogenital system, and other endocrine organs. VIP has a very short half-life of less than 1 minute in the circulation. Plasma levels accordingly are quite low and unresponsive to the ingestion of a meal. High plasma levels have been reported in the setting of watery diarrhea-hypokalemia-achlorhydria syndrome associated with a VIP-producing endocrine tumor (VIPoma). [268] The VIPoma syndrome provides insight into potential targets and actions of VIP. The normal physiological role of VIP is difficult to determine because of the local actions and absence of reagents with which to dissect its role from those of other related transmitters. Among the potential actions are stimulation of enteric smooth muscle, stimulation of pancreatic exocrine and intestinal secretion, inhibition of gastric acid secretion, and modification of immune function and gastrointestinal blood flow. [269], [270], [271], [272], [273], [274], [275], [276], [277], [278] and [279] The list of tissues and cells containing VIP receptors includes pancreatic acini, pancreatic and biliary ductular epithelial cells, gastric and intestinal epithelial cells, vascular smooth muscle cells, lymphocytes, and a number of epithelial tumors. Direct effects on enteric smooth muscle cells and modulatory effects on interneurons have been demonstrated. VIP is a key candidate for mediating descending relaxation of the peristaltic reflex and relaxation at the lower esophageal sphincter, the internal anal sphincter, and the tenia coli. [280], [281], [282] and [283] Two VIP receptors have been cloned: VIP_1 (or $VPAC_1$) and VIP_2 (or $VPAC_2$) receptors. [239], [284], [285] Both are typical members of the secretin family of G protein–coupled receptors. VIP is also well recognized by the PACAP (or PAC_1) receptor. [113], [286] Both the VIP_1 and VIP_2 receptors have similar structural specificities for VIP, PACAP, and PHI (VIP = PACAP > PHI). Secretin is recognized weakly by the VIP_1 receptor, and not at all by the VIP_2 receptor. The selectivity of the PACAP receptor is quite distinct (PACAP > VIP > PHI). Another interesting feature of the PACAP receptor is prominent alternative splicing, yielding variants that differ in both ligand recognition and intracellular signal transduction pathways. The specific tissue and cellular distribution of these receptors is being characterized. Studies with chimeric constructs incorporating domains of the VIP and secretin receptors have also supported the importance of the amino-terminal domain of these receptors in their selectivity for binding and initiation of active conformations. [30], [31], [287], [288] Such observations indicate the need to understand more about the conformation of the ectodomain of these receptors in an effort to contribute to the rational design of drugs that might act on this important family of receptors.

PACAP PACAP is a 27–amino acid linear peptide that was initially isolated from ovine brain. [289] Unusual for this family of hormones, there is also a second molecular form, PACAP-38, [290] which consists of PACAP with a carboxyl-terminal extension of 11 amino acids. Like many other brain peptides, PACAP is also present in the gastrointestinal tract, [291], [292] in nerve fibers, and in myenteric and submucous ganglia. [292], [293] PACAP-38 is predominant in nonenteric locations (e.g., brain, lung, and testes), whereas PACAP-27 is predominant in the intestine. [294], [295] Like VIP, PACAP is present in the circulation in very low levels, which are unresponsive to the ingestion of a meal. Because of the high degree of homology between PACAP and VIP and the relationship of their receptors, interest has been focused on this neurotransmitter, even though it does not yet have proven physiological functions. Candidate functions include relaxation effects on colonic smooth muscle, stimulation of pancreatic exocrine secretion, and vasodilation. [296], [297] and [298]

GIP GIP was originally called *gastric inhibitory peptide,* until it became clear that inhibition of acid secretion is not a physiological action of this hormone. Instead, its most prominent effect is to stimulate insulin production in a glucose-dependent manner, hence the name *glucose-dependent insulinotropic peptide*. A 42–amino acid linear peptide homologous to secretin and glucagons, [299], [300] GIP is produced in K cells in the proximal intestinal crypts. [301], [302] This peptide hormone is secreted predominantly in response to the ingestion of a meal, with fat the major nutrient stimulant. Both the secretion of GIP and the response of target organs to GIP are affected by cholinergic tone. GIP has a half-life of 18 minutes, which is longer than that of most gastrointestinal peptides. The GIP receptor, like the peptide, is homologous to other members of the secretin receptor family. [240] This is also true for the description of its coupling, signaling, and regulatory events. [303], [304] The main established biologic function for GIP is its role as a stimulant of insulin secretion (incretin). [303] Incretins provided the explanation for the greater insulin responses to nutrients administered orally as opposed to parenterally. [305] There have been many candidates to mediate this action, but GIP and GLP-1 are considered important. For GIP to exert this effect on the cell of the pancreatic islet, the serum glucose level must be above 110 mg/dL, thus providing protection against hypoglycemia. Other metabolic effects of GIP have been described, such as inhibition of hepatic glucose output, inhibition of fat absorption, and inhibition of lipoprotein lipase. The physiological role of GIP in mediating these effects is not clear.

Tyrosine Kinase Receptor Family

The receptors in this family are quite distinct from the G protein–coupled receptors. All are single-transmembrane receptors and all have prominent effects on cell growth (Fig. 4-18). Some also affect cell survival, differentiation, and movement. Many G protein–coupled receptors also appear to mediate effects on cell growth, but they do so through a distinct mechanism. Perturbations of several tyrosine kinase receptors have been associated with tumorigenesis. Activation of G protein–coupled receptors can also stimulate tyrosine phosphorylation events; however, most of the substrates are distinct from those phosphorylated in response to activation of the tyrosine kinase receptor family. [127]

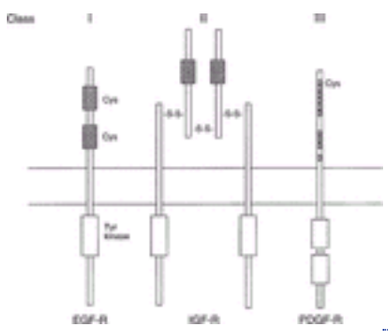

FIGURE 4-18. The three classes of tyrosine kinase receptor molecules. All possess cysteine-rich regions in the ectodomain and tyrosine kinase catalytic domains in the cytoplasm. The classes are dependent on the continuity and spacing of these regions, as well as the subunit structure necessary to achieve a functional unit for the class 2 receptors.

Epithelial Growth Factors Epithelial growth factor (EGF) is a 53–amino acid single-chain polypeptide that contains three intramolecular disulfide bonds. Additional ligands in this family include transforming growth factor-a, amphiregulin, betacellulin, heparin-binding EGF, and the neuregulins. All share the sequence motif CX_7 $CX_{2–3}$ $GXCX_{10–13}$ $CXCX_5$ GXRC. All are produced as single-transmembrane proteins with an amino-terminal proregion, a mature growth factor region ranging in length from 50 to 87 residues, and a relatively short cytosolic domain. Members of the EGF family are active both while anchored to the membrane and after proteolytic processing to release the mature growth factor domain. [92] There are four members of the EGF receptor subfamily, including the EGF receptor encoded by *erb*-b and proteins encoded by *erb*-b2, *erb*-b3, and *erb*-b4. All are single-transmembrane glycoproteins that incorporate two distinct cysteine-rich regions in the ectodomain and a tyrosine kinase catalytic domain and typical sites of potential phosphorylation in cytoplasmic regions. Receptor dimerization or oligomerization is a typical initial step in signaling that is induced by agonist binding. This is followed by cross phosphorylation on tyrosine residues of the receptor and the recruitment of kinase substrates and other signaling proteins. Many of these substrates have Src-homology-2 (SH2) domains. A large number of potential signaling cascades have been described, including the Ras, Raf, and MAP kinase pathways and the JAK/STAT pathways. [306], [307] EGF signaling also includes activation of phosphatidylinositol pathways, with activation of protein kinase C isozymes and increases in intracellular Ca^{2+}. Many gastrointestinal functions can be affected by this family of hormones

and receptors. Included among the most prominent effects are inhibition of gastric acid secretion, protection against injury, and stimulation of intestinal cell growth. There are also putative growth-stimulating and inhibitory effects on a number of gastrointestinal tumors. This provides many interesting possibilities for therapeutic uses of agonists and antagonists acting at these receptors. [308]

Fibroblast Growth Factors Fibroblast growth factors (FGFs) are a group of polypeptides with angiogenic and mitogenic activities that are active in tissue repair and in modulating tissue differentiation. FGFs induce chemotactic activities in fibroblasts and endothelial cells. [94] Nine members of the FGF family have been identified, each with a conserved core sequence of approximately 120 residues and with high affinity for heparin and glycosaminoglycans. FGF1, also known as *acidic fibroblast growth factor,* and FGF2, also known as *basic fibroblast growth factor,* are not efficiently secreted, whereas all other forms possess signal peptides and are secreted. FGF7 is also known as *keratinocyte growth factor (KGF)*. Receptors for the FGFs are monomeric, and like other growth factor receptors, they possess a tyrosine kinase domain. Unlike that of most such receptors, the tyrosine kinase domain of these receptors is discontinuous, with a 14-residue interruption. Also, the FGF receptors have two or three immunoglobulin-like domains within an extracellular domain. At least four distinct genes encode FGFR-1 through FGFR-4. FGFR-1 and FGFR-2 also have multiple alternatively spliced forms. All four types of FGFR can be activated by either the acidic or basic forms of FGF (FGF1 or FGF2). FGF7 (KGF) binds only to the KGFR, which is a distinct splice variant of the FGFR-2. Heparin sulfate proteoglycans on the cell surface also can bind these growth factors, but with low affinity. It has been postulated that this is a site for the collection of the ligand in preparation for binding to its high-affinity receptor. Autophosphorylation of the FGF receptors occurs and is a mechanism for regulation. Tyrosine phosphorylation occurs in response to FGF binding, thereby opening and exposing the kinase domain. Phospholipase C-? can associate with the tyrosine-phosphorylated receptor. Activation also stimulates Shc association with a kinase domain of the receptor, which leads to activation of Grb2/SOS. This in turn leads to the typical series of events activated by other growth factors, such as activation of Raf-1, MAP kinase kinase, and MAP kinase. FGF receptors can form homodimers or heterodimers. The composition of the dimer can determine the details of the signaling initiated. Signaling cascades appear to be quite similar to those described for the EGF receptors. Biologic actions for this group of hormones include prominent effects on angiogenesis, through effects on endothelial cells. Also described are effects on fibroblasts, smooth muscle cells, and various neuronal cells. Like other growth factors, FGFs also have been implicated in tumorigenesis.

Insulin-like Growth Factors Insulin-like growth factors (IGF-1 and IGF-2) are single-chain polypeptides homologous to proinsulin. [93] These hormones were first recognized by the demonstration that the addition of insulin antiserum to neutralize insulin activity failed to inhibit all activities attributed to that hormone. The IGFs are structurally related to insulin, but they bind poorly to traditional insulin receptors. Like insulin, the IGFs have both A and B domains. Unlike insulin, they lack the C domain, which is cleaved in the processing of insulin to achieve its mature two-chain form. IGFs remain a single chain, although they fold to achieve a conformation similar to that of insulin. Like the insulin receptor, the type 1 IGF receptor consists of a heterotetrameric structure with two a subunits and two ß subunits, both of which come from a single gene product. Disulfide bonds link the complex, with the a subunits residing entirely outside the cell and possessing the binding determinants, and the ß subunits spanning the plasma membrane and containing the tyrosine kinase domain intracellularly. As in the insulin receptor, signaling involves tyrosine autophosphorylation, as well as association and phosphorylation of other substrates. In addition to the receptor, there are soluble circulating and interstitial IGF-binding proteins (IGFBPs 1–6). They may affect the volumes of distribution and half-lives of IGF and thereby modify its biologic activities. IGFs affect the growth and differentiation of a variety of tissues.

Toll Receptor Family

All multicellular organisms have innate immunity as a form of host defense against microbes. Infection or infestation leads to early defenses at the levels of phagocytic leukocytes, endothelial cells, and mucosal epithelial cells, with responses mediated by receptors that recognize pathogen-associated molecular patterns, such as the lipopolysaccharide in bacterial cell walls. Activation of such receptors leads to biologic responses that are designed to clear the offending stimulus and to protect the organism. The toll-like receptors are a particularly interesting group of this type of receptor that can recognize and distinguish between various classes of pathogens. [309] They are located on monocytes, macrophages, neutrophils, dendritic cells, intestinal epithelial cells, and endothelial cells. A major signaling target of these receptors is the activation of NF-?B transcription factors, which can affect the expression of various cytokines, adhesion molecules, acute phase proteins, and inducible enzymes.

The toll receptors represent a family of single-transmembrane molecules having multiple copies of leucine-rich repeats in their unique extracellular domain and a cytoplasmic toll/1R (TIR) motif that is related to that found in the interleukin-1 (IL-1) receptor. Because of the latter motif, these receptors signal like the IL-1 receptor. They are thought to dimerize and bind to adapter proteins that initiate signaling. These receptors may represent interesting and possibly productive targets for the treatment of inflammatory and immune-mediated diseases.

HORMONES IN GASTROINTESTINAL DISEASE

As critical as gastrointestinal hormones and their receptors are for the integration and regulation of digestive function in health, their major interest for the clinician is in regard to hormone-secreting tumor syndromes. These syndromes can be dramatic expressions of unregulated overactivity of a particular one of these hormones, without the advantage of the normally active mechanisms for feedback inhibition. Such tumors are typically of the islet cell type, arising in the pancreas, although the duodenum is another common location for gastrinomas. The two most common and dramatic presentations of such tumors, exclusive of the insulinomas and glucagonomas typically seen by the endocrinologist, are gastrin-secreting and VIP-secreting islet cell tumors. [135], [268] Gastrin is produced in the fetal pancreas, but normally it is not produced at all in the adult pancreas. [44] VIP is a normal pancreatic neurotransmitter that is present in very low concentrations under normal circumstances. The syndromes of gastrin overproduction in Zollinger-Ellison syndrome (gastrinoma) and VIP overproduction in the Verner-Morrison syndrome or watery diarrhea-hypokalemia-achlorhydria syndrome (VIPoma) are discussed in detail elsewhere in this book (see Chapter 67 and Chapter 97). Knowledge of the biologic actions of these peptides based on the previous discussion should provide good insight into these clinical syndromes.

The clinical contributions of the gastrointestinal endocrine system to other gastrointestinal diseases is less clear because of the redundancy of the control mechanisms, with more than one hormone and neurotransmitter serving similar functions. Common disturbances of gastrointestinal function, such as irritable bowel syndrome, peptic ulcer disease, gallstone disease, dysmotility states, and even pancreatitis, may be influenced by a diverse menu of hormones and neurotransmitters. Only when the choice of selective pharmacological agonists and antagonists is similarly broad will the roles of these agents be deconvoluted. The reduced clearance of several of these hormones from the circulation in renal insufficiency can be associated with a broad spectrum of results because of the antagonistic effects of elevated peptides. [88], [310] Because nutrient absorption is needed to stimulate the secretion of many gastrointestinal hormones, circulating levels can be influenced markedly by syndromes of maldigestion, such as occurs in pancreatic insufficiency or celiac disease.

Some gastrointestinal peptides and analogs have reached clinical practice. They have numerous diagnostic applications. Pentagastrin is the agent of choice for gastric acid secretory testing. [311] CCK is used for pancreatic function testing, as well as in radiographic studies of gallbladder emptying and pancreatic duct size. [312] Secretin is administered in a provocative test for gastrinoma. [89] Glucagon is used to relax the gastrointestinal smooth muscle activity to facilitate endoscopic, radiologic, and intubation procedures. [313]

In addition to having diagnostic applications, potent agonist analogs of somatostatin are now commonly used as therapeutic agents (see structures of octreotide and lanreotide in Fig. 4-15). A long-acting depot form of lanreotide that can be injected intramuscularly is now available. These ligands have highest affinities for receptor subtypes 2 and 5, with moderate affinity for subtype 3 and very low affinity for subtypes 1 and 4. Somatostatin and its analogs suppress hormone release and hormone action for a number of gastrointestinal peptides. They are being used to treat the diarrhea and flushing of metastatic carcinoid syndrome, the secretory diarrhea of the VIPoma syndrome, the skin lesions of the glucagonoma syndrome, and other refractory diarrheal syndromes and fistulae. [314] Although these agents effectively inhibit acid hypersecretion in gastrinoma, there are better methods for controlling acid secretion, such as hydrogen-potassium ATPase inhibitors and histamine H_2 receptor antagonists. [315], [316] Somatostatin agonists have helped to relieve symptoms, and in rare cases they have induced islet cell tumor regression. They are also being tested for action against other epithelial carcinomas. More selective somatostatin receptor antagonists have been recently developed but are not yet available for clinical use.

Gastrointestinal hormones, such as gastrin-releasing hormone and gastrin, have been implicated as autocrine growth factors in various epithelial malignancies. Antagonists directed toward blocking the action of these peptides may have important clinical applications in the future.

Acknowledgments

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CHAPTER 5

Graham J. Dockray

THE BRAIN-GUT AXIS

The gastrointestinal tract and the central nervous system (CNS) engage each other in two-way communication. The CNS is informed of the state of the gastrointestinal tract by afferent neurons, and is able to control or modulate digestive function though efferent neurons that are part of the autonomic nervous system (Fig. 5-1A). Afferent neurons signal information to the CNS about the chemical environment of the gut lumen, about tension in the gut wall, and about the condition of tissues (e.g., inflammation) (Fig. 5-1B). Efferent neurons determine gastrointestinal responses during the cephalic phase of digestion. They also mediate gastrointestinal responses after reflex stimulation of afferent pathways, activation of somatosensory inputs, and stimulation by higher centers, including, for example, changes in the emotional state.

FIGURE 5-1. Relationships between the brain and the gut. **A:** The gastrointestinal tract down to the midcolon is supplied with efferent and afferent nerve fibers running in the vagal and splanchnic trunks. **B:** Afferent neurons discharge in response to lumenal chemicals, mechanical deformation of their terminal fields, humoral factors, cytokines, and other inflammatory mediators; endocrine cells, mast cells, and cells of the immune system may be influenced by lumenal chemicals. **C:** Efferent neurons in both splanchnic and vagal nerve trunks terminate on myenteric nerve cell bodies from which intrinsic motor neurons influence secretion and motility; sympathetic efferents may also directly supply blood vessels. The response of the enteric motor neurons is also regulated by humoral factors and by other enteric neurons. *CNS,* central nervous system; *gang.,* ganglion.

The major pathways for communication between the brain and gut are the vagal, splanchnic, and sacral nerve trunks; all three contain both afferent and efferent nerve fibers. It has been recognized since the early 20th century that afferent signals associated with the physiological regulation of digestion are typically conveyed by the vagal pathway and painful sensations are conveyed by the splanchnic pathway. [1] Mechanisms that modulate afferent signals have been identified at various levels, including at the peripheral end of afferent fibers and within the CNS, and are a feature in a number of gastrointestinal diseases. [2]

Vagal efferent neurons provide the major parasympathetic route regulating gut function during digestion. Splanchnic efferent neurons constitute the sympathetic outflow to the gut and are activated in stress and adaptation to potentially noxious challenges. The neurons of the enteric ganglia are a major target for both types of efferent pathway (Fig. 5-1C). The motor neurons in these ganglia supply smooth muscle, glandular cells, and blood vessels. The enteric ganglia also function as centers for integration; they communicate with each other, and they receive inputs from intrinsic afferent neurons and from endocrine cells. Ganglion cells act, then, both to modulate and to mediate efferent signals from the CNS. Not surprisingly, the relative importance of information exchange between the alimentary tract and CNS varies between different functions and between different regions of the gut.

DEVELOPMENTAL BIOLOGY OF THE BRAIN-GUT AXIS

The pathway taken by the vagus nerve marks the route by which cells of the neural crest migrate to the gut during development. The molecular mechanisms that determine this migration are now being elucidated. They are of interest because they provide a basis for understanding the developmental biology not just of the brain-gut axis but also of the entire intrinsic nervous system of the gastrointestinal tract. Thus, cells of the vagal neural crest migrate the length of the gut to supply most of the intrinsic innervation, although a small number of cells from the truncal and sacral neural crests also contribute to the colonization of the stomach and distal colon, respectively. Neural crest cells give rise to multiple cell lineages, including neurons, neuroglia, and melanocytes, but not to epithelial enteroendocrine cells as previously thought. [3] The consequences of impaired migration or maturation of neural crest cells are illustrated by Hirschsprung disease, in which a failure of neuronal precursors to colonize the distal part of the colon leads to an aganglionic segment associated with tonic smooth muscle contraction and a dilated proximal segment. Mutations in several genes that regulate neural crest cell survival, migration, and maturation have been identified in people with Hirschsprung disease and in mutant mice with an aganglionic colon.

The development of enteric neural crest cells requires the c- *ret* protooncogene that encodes a receptor tyrosine kinase. The latter forms the transducing component of a signaling pathway activated by glial cell line–derived neurotrophic factor (GDNF). [4], [5] GDNF binds to a second protein, a glycosyl phosphatidylinositol–linked protein, GFRa $_1$, that then activates Ret. [6] Loss-of-function mutations of the c- *ret* gene occur in some cases of Hirschsprung disease. [7], [8] Interestingly, other mutations of this gene are associated with multiple endocrine neoplasia type II. In mice, deletion of either c- *ret* or *GDNF* leads to a failure in the development of the kidney and of neurons throughout the intestine. [9], [10], [11] and [12] The c- *ret* gene is expressed relatively early by neural crest cells, [13] and it is thought that this system is

required for the proliferation and survival of enteric neural precursor cells. A second system known to be required for the maturation of neural crest derivatives involves endothelin-3 (ET-3) and its G protein–linked receptor, the endothelin-B (ET_B) receptor. In some patients with Hirschsprung disease, a missense mutation in the gene encoding the ET_B receptor is associated with impaired Ca^{2+} signaling on stimulation. Similarly, targeted disruption of the ET_B receptor in mice leads to the loss of enteric ganglion cells in the distal colon, but not more proximally. Mice with mutations of the *ET3* gene that prevent the appropriate posttranslational maturation of the ET-3 precursor protein also exhibit impaired development of the enteric nervous system and an aganglionic distal colon. [14] It is thought that the ET-3/ET_B system functions after the c-*ret*/*GDNF* system, [15], [16] and acts to delay neuronal differentiation so that loss of function leads to a failure in the migration of neuronal precursors as a result of premature differentiation. These are not the only control systems determining the maturation of different populations of intrinsic gut neurons. Others include the homeobox gene *Phox2b,* which is required for expression of Ret, and of a second transcription factor, Mash-1, which regulates the expression of a catecholaminergic phenotype in neural crest–derived cells. [17], [18]

METHODS FOR STUDYING THE BRAIN-GUT AXIS

Present knowledge of the organization and function of the brain-gut axis is based on observations based on a wide variety of different experimental techniques. These methods can be loosely grouped into four categories.

1. *Methods directed at the study of organ function.* Methods that measure changes in secretion or motility in the gut after sham feeding, electrical stimulation of nerve trunks, or loss of function after nerve section (e.g., vagotomy) have often provided the first evidence for functional pathways linking the brain and the gut. Similar experiments can often be performed in humans and animals (e.g., barostat measurements of gastric motility). Taken in isolation, however, the results of such studies seldom allow the precise definition of brain-gut interactions because the relative importance of endocrine mediators and nervous reflexes that are extrinsic or intrinsic to the gut is difficult to evaluate in the intact organism. This type of approach can be coupled with the use of drugs to block or activate particular neurotransmitters or types of neurons, but again, interpretation of the data may be impeded by the fact that the same neurochemical mechanisms operate at multiple sites in the gut and the CNS.
2. *Morphologic methods that define the cellular basis of the brain-gut axis.* The nerve pathways linking the brain and the gut can be precisely defined by using tracers that are taken up by nerve cells and transported along axons—for example, fluorescent compounds such as True Blue, and pseudorabies virus, which is transported across synapses and so reveals chains of neurons in a pathway. In addition, the expression of immediate-early gene (e.g., c-*fos*) proteins has been widely used to identify neuronal responses to stimulation, although this technique does not reveal neurons that are inhibited by a particular stimulus. These methods can be coupled with surgical or chemical lesioning of neurons, and with immunocytochemistry for neurochemical characterization of a labeled neuron. Together, such methods provide quantitative information on the organization and neurochemistry of both afferent and efferent pathways, and on CNS pathways. This type of approach is, however, better suited to studies of experimental animals rather than of humans.
3. *Single-unit electrophysiological recording.* The physiological properties of single afferent and efferent nerve fibers, or of CNS neurons, can be studied by electrophysiological recording of their discharge in response to gastrointestinal stimulation. The data obtained allow rigorous analysis of the patterns of signaling, and can provide a formal framework for the description of reflex pathways identified in whole-organism studies or by morphologic methods. Again, however, these methods are more readily applied to animal than human studies.
4. *Methods appropriate for the study of the human brain-gut axis.* A heterogeneous group of methods has been of particular value in human studies. There are disadvantages to each of the techniques listed below, and the combined use of more than one of them therefore offers obvious advantages.
   i. *Evoked potentials.* Before the advent of modern imaging techniques, the activation of human CNS neurons after stimulation (e.g., balloon distention, electrical stimulation) was studied by observing the evoked potentials detected by scalp electrodes. [19], [20] This method is useful for measuring cortical neuronal responses (cortical-evoked potentials) that are relatively superficial in the brain, but it is not useful for studies of the activity of deeper neurons. It can, however, be used for recording responses from spinal neurons, and of the various techniques presently available, it is probably the method of choice for the noninvasive detection of human spinal afferent pathways. The temporal resolution is good (i.e., milliseconds), but the spatial resolution is only 1 to 2 cm.
   ii. *Magnetoencephalography (MEG).* This technique is based on the use of magnetic sensors near the scalp that detect changes in the magnetic field as a consequence of neuronal activity in the underlying part of the brain. The spatial resolution can be 2 to 5 mm, and images can be obtained in milliseconds. Like cortical-evoked potentials, however, MEG can be difficult to use for recording from subcortical regions.
   iii. *Positron emission tomography (PET).* This technique provides a view of neuronal activity as reflected by changes in blood flow. The time lag between changes in neuronal function and blood flow to the relevant region, however, may make it difficult to detect rapid responses. PET allows the identification of brain regions with a resolution of 2 to 8 mm, but the temporal resolution is relatively poor (i.e., 40 seconds). Repeated studies on individuals are difficult because of the risk associated with isotopic exposure and the relatively high cost.
   iv. *Functional magnetic resonance imaging (fMRI).* This imaging technique also detects changes in neuronal activity as reflected by blood flow. The spatial resolution is 2 to 8 mm. Temporal clustering analysis of fMRI data has shown that it is possible to resolve feeding-related changes in hypothalamic function. [21] fMRI is cheaper than PET, and repeated measurements are harmless.
   v. *Transcranial magnetic stimulation (TCMS).* Unlike the other techniques mentioned, this approach does not depend on the detection of neuronal responses; rather, it provides a method to activate specific CNS regions selectively. This is not a new technique, but its introduction to gastroenterology is recent. It can be used repeatedly in humans, and it is useful for the activation of cortical motor regions when coupled with the recording of gut function (e.g., electromyography). [22]
   vi. *Reporting visceral sensations.* Subjects report sensations after stimulation (mechanical, chemical, electrical) of the gut by using visual analog scales or by selecting adjectival descriptors of sensation. This approach is widely used and often valuable when combined with other techniques (e.g., fMRI). [23]

BRAIN-GUT CONNECTIONS

Afferent Neurons

The vagus nerve consists predominantly of afferent nerve fibers. In the rat, between 75% and 90% of vagal fibers are thought to be afferent. [24], [25] Almost all these fibers are unmyelinated or only lightly myelinated, and consequently their conduction velocities are relatively low (0.5 to 5 m/s). The cell bodies of vagal afferent nerve fibers are located in the nodose ganglia, and their central terminals are found in the nucleus of solitary tract (NST). The cell bodies of splanchnic and sacral afferent fibers are found in the dorsal root ganglia, and their central terminals are in the spinal cord. [26]

Different populations of afferent fibers respond to stimuli arising in the mucosa, muscle layers, and serosa. The stimulus in the lumen may be mechanical, thermal, or chemical. In addition, afferent neurons express receptors for various hormones, neurotransmitters, and immunomodulators (see Fig. 5-1B). It is convenient to use the term *afferent* to describe the direction in which information is passed relative to the CNS. However, many visceral afferent neurons (particularly splanchnic ones) synthesize neuropeptides that are transported to terminals both in the CNS and in the periphery, where they are released on nerve stimulation, giving rise to motor (or efferent) functions through an axon reflex. [27], [28]

Some intrinsic gastrointestinal neurons project to the prevertebral ganglia (see Fig. 5-1C). These neurons are therefore distinct from vagal, splanchnic, and sacral afferents, but they function as the afferent arm in some reflexes operating through the sympathetic ganglia. Electrophysiological recording of the discharge of nerve fibers running in the mesenteric nerve bundles detects both extrinsic afferent and intrinsic afferent nerve fibers, and care is required in distinguishing between them.

Organization The vagal afferent supply of the upper gastrointestinal tract is conveyed by the gastric branches of the vagus. More distal regions are innervated by vagal fibers that pass through the celiac–superior mesenteric ganglia and travel with the mesenteric nerve bundles running along blood vessels. There is a loose viscerotopic representation of vagal afferent terminals within the NST. [29], [30] Tracing experiments suggest that within the bowel wall, vagal afferent nerve fibers run in the myenteric plexus, where they form characteristic intraganglionic laminar endings. The innervation of the mucosa is variable. In the intestine, some villi appear to lack an afferent innervation, whereas others exhibit vagal afferent fibers in the lamina propria; afferent fibers do not appear to penetrate the epithelium. [31], [32] Splanchnic afferent nerve fibers enter the spinal cord through the dorsal roots and pass to the gut along blood vessels. They traverse the prevertebral ganglia, where they may supply collateral fibers innervating the ganglion cells, suggesting a direct modulatory role in sympathetic postganglionic discharge (see Fig. 5-1B). In the spinal cord, there are fewer terminals of splanchnic afferent neurons than of somatic afferent neurons. Splanchnic afferents terminate mainly in lamina I of the dorsal horn, although some project to lamina V. [33] The spinal cord neurons that respond to visceral afferent stimulation may also have inputs from somatic afferent nerve fibers. The peripheral terminals of splanchnic afferent nerve fibers can be identified within the gastrointestinal tract with the use of peptide markers such as calcitonin gene–related peptide (CGRP), which is found in fibers in the myenteric plexus, smooth muscle, around submucosal blood vessels, and in the mucosa. [34], [35]
Neurochemistry: Peptide Transmitters Marked neurochemical differences are found among gastrointestinal afferent neurons. About 80% of the spinal afferent neurons serving the rat stomach contain CGRP immunoreactivity, and about 50% contain substance P immunoreactivity. [34], [36] In contrast, fewer than 10% of the vagal

afferent neurons projecting to the gut contain these peptides. The proportion of the spinal afferent neurons associated with substance P or CGRP immunoreactivity is considerably higher in the gastrointestinal tract than in skin or joints. [37] Almost no primary afferent neurons containing somatostatin project to the stomach, however, although dorsal root ganglia neurons containing somatostatin project to the skin and joints. Low doses of the sensory neurotoxin capsaicin selectively stimulate C-fibers, whereas high doses selectively destroy C-fibers, particularly in neonatal animals. [38] Capsaicin-induced lesioning of afferent neurons produces a decrease in the expression of CGRP and substance P and a loss of immunoreactive nerve fibers in the gut. [34], [36] Surgical section of afferent nerve trunks also leads to the decreased expression of CGRP and substance P in primary afferent neurons. Interestingly, this treatment produces an increase in the expression of vasoactive intestinal polypeptide (VIP) and galanin in the afferent neurons. [39] The afferent innervation of the upper gastrointestinal tract does not normally contain VIP, although an exception may be the sacral afferents serving the distal colon of the cat, which reportedly express VIP. [40]

Neurochemistry: Receptors Vagal afferent neurons projecting to subdiaphragmatic structures express cholecystokinin type A receptors (CCK-A). [41] After ligation of the vagal nerve trunk, CCK-A receptors accumulate on the central side of the ligature, indicating transport toward the gut. [41], [42], [43] and [44] Capsaicin inhibits this transport. The data suggest that CCK receptors are expressed by vagal afferent neurons, a conclusion supported by the cloning of CCK-A receptor cDNA from nodose ganglia and the localization by in situ hybridization of CCK-A receptor mRNA to nodose ganglion cell bodies. [44] These neurons also express the putative peptide transmitter cocaine- and amphetamine-regulated transcript (CART). [45] In rabbit nodose ganglion, a common population of cell bodies has been shown to express CCK-A and neuropeptide Y (NPY)–1 and NPY-2 receptors. [46] Vagal afferent neurons also express 5-HT $_3$ receptors; these are probably transported centrally, to the NST, and peripherally, where they are thought to be important in mediating the actions of 5-HT released from intestine. [47]

Efferent Neurons

Organization The cell bodies of parasympathetic preganglionic nerve fibers that run in the vagus nerve are found in the dorsal motor nucleus of the vagus. There is a viscerotopic representation of neurons within the dorsal motor nucleus, and columns of cells corresponding approximately to the branches of the vagal nerve can be identified. [25], [48] The peripheral terminals of vagal efferent fibers have been mapped with the use of tracers such as DiI and PHA-L. [31], [49] Different tracers elicit quantitatively different results, but several conclusions emerge from the available data. Vagal efferent fibers can be shown to supply the gastrointestinal tract as far as the descending colon, but there is a proximal-distal gradient in the relative abundance of these fibers. In the stomach, where the vagal innervation is most dense, virtually all gastric myenteric ganglia are contacted by vagal efferent nerve fibers, but there is little or no supply to the submucosa or mucosa [31], [49] (see Fig. 5-1C). A single vagal efferent fiber may contact many myenteric ganglia. The contacts between preganglionic fibers and ganglion cells exhibit considerable variation; some are clear synaptic-like associations, whereas others are less well defined. The nerve cell bodies of postganglionic sympathetic neurons to the gut are in the prevertebral ganglia, and the preganglionic cell bodies are in the intermediolateral column of the spinal cord. Most of the upper gut receives its innervation from the celiac–superior mesenteric ganglia, and the lower gut from the inferior mesenteric ganglion. The postganglionic fibers run along blood vessels. Enteric ganglion cells are a major target, but so too are submucosal blood vessels (see Fig. 5-1C).

Neurochemistry The predominant phenotype of vagal efferent neurons is cholinergic, although the cell bodies of up to 30% of the neurons of the dorsal motor nucleus projecting to the stomach also appear to express tyrosine hydroxylase and may have a dopaminergic phenotype. [50] Some gastric vagal efferent fibers also contain galanin immunoreactivity, [49] and a population of the motor neurons of the nucleus ambiguus that project to the esophagus contains CGRP immunoreactivity. [51] It has been recognized for several generations that the main synaptic contact between vagal efferent fibers and cells of the myenteric plexus is cholinergic-nicotinic; combined anterograde tracing and immunohistochemistry suggest that gastric myenteric ganglion cells with a vagal input contain 5-HT, VIP, and enkephalin immunoreactivity. [49] The adrenergic phenotype of sympathetic neurons to the gut is well recognized. Many of these neurons also express neuropeptides, however, and the neurons seem to be chemically coded on the basis of their gastrointestinal targets. Thus, neurons of the celiac ganglia that project to submucosal blood vessels in the gut often contain NPY, and those that go to the submucosal plexus contain somatostatin. [52]

CNS Regions and the Digestive Tract

Ascending CNS Pathways Stimulated from the Gut Important early studies of the CNS regions responding to gut stimulation were conducted with cortical-evoked potentials. [19], [20] Subsequently, many groups have made recordings of the activity of identified CNS neurons responding to gastrointestinal stimuli or humoral mediators. [53], [54] and [55] Insight into the organization of CNS regions associated with digestive function has been obtained more recently by means of tracing techniques, the induction of immediate-early gene expression (e.g., c- *fos*), and, in the case of the human brain, the application of PET and fMRI. [19] Major CNS projections from the vagal complex identified by anterograde labeling include those to the paraventricular nucleus, the bed nucleus of the stria terminalis, the central nucleus of the amygdala, as well as those to the parabrachial nuclei and the subthalamus. [31] Many of the same regions are identified by the induction of c-Fos protein in response to nutrients and to the peripheral administration of regulatory peptides. [56], [57], [58] and [59] Noxious stimulation of the spinal afferents (visceral, somatic) increases c-Fos in the spinal cord neurons. [60], [61] Schuligoi and colleagues [62] reported that noxious stimulation of the gastric mucosa with HCl and formalin did not increase the spinal cord expression of c-Fos detected by in situ hybridization, but serosal application of formalin produced a significant increase. Because mucosal stimulation in these experiments increased blood flow, and this response is recognized to be mediated by splanchnic afferent neurons, it seems possible that noxious stimulation of the gastric mucosa may evoke an afferent fiber response of sufficient strength to trigger a local axon reflex but inadequate to recruit spinal nociceptive circuits. In healthy human subjects, PET and fMRI studies have identified responses in the anterior cingulate cortex in response to painful rectal distention. Interestingly, patients with inflammatory bowel disease exhibit differential responses in this region after similar stimulation. [63], [64] The anterior cingulated cortex is probably important more generally in visceral pain. Thus, nonnoxious stimulation of the human esophagus activates the central sulcus, the insular cortex, and the frontal/parietal operculum [65]; noxious stimulation activates the same regions, and also the anterior cingulate cortex and an expanded region of the insular cortex (Fig. 5-2). The CNS regions that respond to visceral stimuli exhibit plasticity; for example, there are prolonged changes in the cortical responses to short-term stimulation of the esophagus. [66] The cellular mechanisms accounting for plasticity and differential responses in disease are still largely unknown.

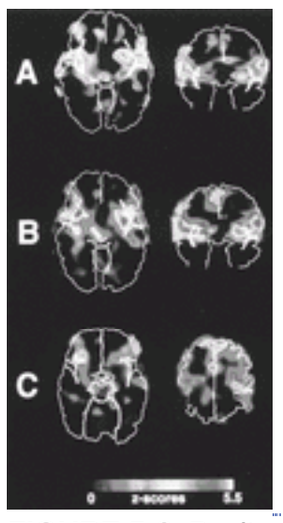

FIGURE 5-2. Positron emission tomography data after esophageal stimulation in humans. Scans of cortical areas activated during definite sensation–no sensation (**A**), pain–no sensation (**B**), and pain–definite sensation (**C**) are shown. **A:** Left and right scans show bilateral activation of the insular and primary somatosensory cortex. **B:** There is greater activation of the insular and primary somatosensory cortex, and some activation of the anterior cingulate gyrus. **C:** The left scan shows activation of the right anterior insular cortex, and the right shows activation of the anterior cingulate cortex. (Adapted from ref. [65], with permission.)

CNS Projections to the Vagal Complex The electrical stimulation of different CNS regions provided convincing early evidence of the role of the CNS in controlling digestive function. [67] Subsequently, retrograde tracing from the vagal complex has revealed the major monosynaptic connections. Thus, the vagal complex receives inputs from the obscure and pallidal raphe nuclei, the paraventricular nucleus, the dorsomedial and lateral areas of the hypothalamus, the central nucleus of the terminal stria, and the insular and medial prefrontal cortexes. [68] In humans, TCMS has been used to examine the cortical control of esophageal function. The relevant area of the cerebral cortex is asymmetric; the greatest esophageal responses were evoked from the right hemisphere in eight of ten subjects and from the left hemisphere in the other two. [69] This asymmetry may explain why the dysphagia that develops after stroke is variable in degree and duration. [22]

SIGNALING FROM GUT TO BRAIN

From the 1950s onward, Paintal, [70], [71] Iggo, [72] and others made electrophysiological recordings of the discharge of single vagal or splanchnic afferent fibers in anesthetized animals. This work has provided the foundation for modern views of visceral afferent nerve function. Individual afferent fibers respond to mechanical deformation of their terminal regions, to the presence of chemical stimulants in the gut lumen, and to humoral agents administered into the circulation. Because visceral afferent nerve fibers may be mechanoreceptors, chemoreceptors, or both, recent classifications of afferent nerve response have been based on the distribution of terminal fields rather than the modality of the receptor. [73], [74]

Mucosal, Muscle, and Serosal Afferents

The responses of vagal afferent nerve fibers to mucosal stimulation, typically in the stomach and proximal small intestine, have been reported by many groups. Less is known about splanchnic afferent responses to mucosal stimulation. [74] All the major macronutrients (i.e., protein, fat, carbohydrate) have been reported to stimulate mucosal afferent nerve fiber discharge. There are also fibers that respond to a variety of nonnutrient chemicals, including acid, alkali, and hypertonic solutions. [74], [75] Many mucosal afferent fibers are multimodal and respond to mechanical as well as chemical stimulation. These fibers, in particular, also respond to light mechanical stimulation, such as brushing or stroking of the mucosa; the responses are rapidly adapting.

The afferent fibers supplying the muscle layers of the gut typically respond to both passive stretch and active contraction, hence the name *in-series tension receptor*. They are characteristically slowly adapting, and if innervating the stomach and proximal small intestine, they have a single receptive field.

The serosal afferent innervation appears to be predominantly spinal in origin. These afferent fibers respond to deformation of the serosa and mesentery, particularly around the bifurcation of blood vessels, thereby conveying information about visceral tension and movement. The receptive fields of these afferent fibers are often punctate.

Mechanoreceptors: Transduction

Sensitivity to mechanical stimulation is a common physiological response and accounts for mechanisms involved in hearing, balance, touch, the regulation of cell volume, and muscle cell responses to stretch. A variety of different cellular mechanisms are now recognized to account for these responses. Many cells have stretch-sensitive ion channels that are gated by changes in membrane tension. The accompanying alterations in membrane potential lead to the propagation of action potentials. Although there is little direct evidence, it is plausible that this type of mechanism is responsible for the sensitivity of muscle and serosal afferent fibers. Balloon distention has been widely used to stimulate mechanoreceptor discharge in visceral afferent fibers.

Afferent fiber populations differ in sensitivity. For example, pelvic afferent fibers responding to distention in the physiological pressure range (<5 mm Hg) are known as *low-threshold fibers*; discharge of these fibers is proportional to distending pressures of up to about 30 mm Hg, which is in the range likely to be perceived as noxious. A separate population of fibers (i.e., high-threshold), however, discharge only in response to high pressures (>30 mm Hg), suggesting a primary function in noxious signaling. [74]

Signals from mucosal afferent fibers that respond to mechanical deformation may be mediated initially by enterochromaffin cells, and many years ago, Bulbring and Lin [76] proposed stimulation of these cells to be the first step in activation of the peristaltic reflex in response to distention of the small intestine. Mechanical deformation at the lumenal surface was postulated to release 5-HT that then stimulated subepithelial nerve fibers. The observation of increased c-Fos immunoreactivity in myenteric neurons after mechanical deformation of guinea pig ileal mucosa, along with evidence of mediation by 5-HT, provides some support for this model. [77]

Chemoreceptors

Transduction The term *chemoreceptor* is used to describe the capacity of afferent nerve fibers to respond to defined chemical agents. Both nonlumenal (neurohumoral) and lumenal chemicals clearly stimulate gastrointestinal afferent nerve fiber discharge. The first humoral stimulants of afferent fibers to be recognized were histamine, 5-HT, and bradykinin. Subsequently, hormones such as CCK were also identified as direct stimulants of visceral afferent nerve fibers. The mechanisms underlying afferent responses to lumenal chemical stimuli are less clear. However, the molecular sensing mechanisms involved in taste perception by the tongue provide a model for understanding what may happen within the gastrointestinal tract. Thus, compounds associated with the taste of salt or acid act by gating of ion channels. [78] Small molecules may therefore penetrate the mucosa to act directly on afferent nerve fibers (e.g., cation channels gated by extracellular H^+). [79] Compounds associated with sweet or bitter tastes are known to act in the tongue through G protein–coupled receptors. [78] a-Gustducin, the GTP-binding subunit of a heterotrimeric G protein, is a putative mediator in these cells. [80] At the cellular and molecular levels, the transduction mechanisms may be similar in the tongue and in the gut, even though taste perception is restricted to the buccal cavity. Consistent with this idea, a-gustducin immunoreactivity has been displayed on the apical membrane of brush cells in the stomach and in the duodenum. These cells are distinct from endocrine cells but are rich in nitric oxide synthase, suggesting that changes in the lumenal environment of the gut trigger the release of nitric oxide, which in turn acts on primary afferent nerve fibers. [81]

Endocrine Cells As Transducers Lumenal stimuli also activate classic enteroendocrine cells, the products of which are putative messengers from the gut to the brain (Fig. 5-3). The mediators include CCK, 5-HT, gastrin, somatostatin, and peptide YY (PYY). CCK is thought to mediate the effects of lumenal protein and fatty acid in the intestine, and 5-HT may mediate the effects of intestinal carbohydrate stimulation. [82] The transduction mechanisms at the apical membrane have, until recently, been uncertain. However, for G cells, it now seems that a G protein–coupled receptor, CaR, responding to both extracellular Ca^{2+} and aromatic amino acids such as L-tryptophan and L-phenylalanine, provides the initial molecular recognition event for multiple lumenal stimuli. [83], [84] After the release of putative endocrine cell mediators, there are two general ways that gut endocrine cell products may signal to the CNS. First, they may penetrate the CNS from the circulation in regions where the blood-brain barrier is absent or leaky; possible examples are pancreatic polypeptide (PP) and ghrelin, which are discussed in subsequent sections. Second, as already noted, primary afferent nerve fibers possess receptors for some of the products of gut endocrine cells (e.g., CCK and 5-HT), which are therefore putative mediators of endocrine-neural signaling pathways. [37] Two schools of thought have emerged regarding how CCK may signal in this way. One line of evidence indicates that CCK stimulates vagal afferent fibers that also function as gastric in-series tension receptors. [85], [86] According to this view, CCK acts by way of the systemic circulation to stimulate vagal afferent fibers serving the gastric musculature. An alternate line of evidence indicates that CCK acts on intestinal mucosal afferent fibers in the subepithelial regions surrounding the CCK-secreting cells. [87], [88] It seems likely that 5-HT released from enterochromaffin cells similarly acts as a paracrine stimulant of local afferent nerve fibers. The pathways have largely been elucidated for rat but probably are similar for humans; for example, lipid-induced depression of gastric motility and sensations of fullness were inhibited by a CCK-A antagonist in healthy human subjects. [89]

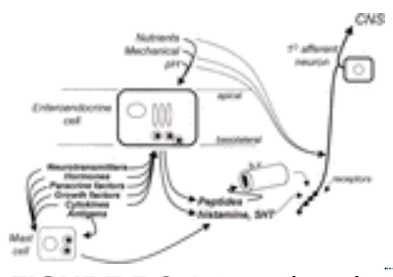

FIGURE 5-3. Interactions between lumenal factors, mast cells, endocrine cells, and primary afferent nerve fibers. The latter may express receptors for endocrine and mast cell secretory products, and may also respond directly to lumenal chemicals that penetrate the mucosa. The discharge of afferent fibers to the smooth muscle may be secondary to passive stretch (distention caused by a meal) or to the contraction of smooth muscle. Peptides released by endocrine cells are thought to act directly on afferent nerve fibers and after transport in the circulation. The receptors expressed by afferent neurons are synthesized in the cell body and may be transported to peripheral terminals. *CNS,* central nervous system; *b.v.,* blood vessel.

Mast Cells and the Extrinsic Innervation of the Gut There is a close relationship between mast cells and substance P–immunoreactive nerve fibers. [90] Mast cell products (i.e., histamine and 5-HT) stimulate the discharge of intestinal afferent fibers, [91] and there is evidence that motor responses to mast cell discharge in vivo are mediated by 5-HT_3 receptors and vagal afferent nerve fibers. [92] In ovalbumin-sensitized rats, antigen challenge increased c-Fos expression in the NST, the lateral parabrachial nucleus, and the paraventricular nucleus. These responses were inhibited by systemic administration of the 5-HT_3 antagonist ondansetron and by

perivagal capsaicin, suggesting a mechanism involving peripheral 5-HT $_3$ receptors and vagal afferent neurons in signaling to the CNS during intestinal anaphylaxis.[93] In addition to the stimulation of afferent fibers by mast cell products, mast cell secretion is stimulated by the activation of vagal efferent fibers (see Fig. 5-3). For example, the administration of a thyrotropin-releasing hormone (TRH) analog that strongly stimulates vagal efferent neurons elicited the release of mast cell markers into the rat intestinal lumen.[94]

Inflammatory Modulation of Afferent Function

Inflammation of the gut wall is a strong stimulant of afferent nerve discharge[95,96] (see Fig. 5-3). Many splanchnic afferent fibers appear normally to be silent; for example, up to 90% of splanchnic afferents to the distal colon are reported to be insensitive to stimulation in the physiological range.[62] Inflammation induced experimentally by the administration of mustard oil, acetic acid, or formaldehyde, however, leads to an increased sensitivity to stimulation.[97] Seemingly silent afferent fibers may play little or no role in the normal physiology of the gastrointestinal tract, but they are important components of the signaling pathway activated by damage or inflammation, and may therefore play a role in gastrointestinal disease states. Activation is likely, in part, to be attributable to the release of cytokines such as interleukin-1ß (IL-1ß), which increase the sensitivity of afferent fibers to stimulation[97] (see Fig. 5-3).

Pain and Visceral Hypersensivity

The noxious stimulation of splanchnic afferent nerve fibers is associated with painful sensations. The sensation of pain may, however, also be evoked by normal or innocuous stimuli, a process sometimes called *hypersensitivity*. It is well recognized that patients with irritable bowel syndrome may report painful sensations in response to distention of the colon at pressures that are not perceived as painful by healthy subjects. The term *allodynia* is used to describe noxious sensations evoked by innocuous stimuli, and the term *hyperalgesia* describes exaggerated responses to noxious stimuli. The mechanisms that account for visceral hypersensitivity have generated considerable interest.[23] These mechanisms are a feature of both inflammatory and noninflammatory conditions (e.g., functional bowel disorders). Changes at any of several different levels in the brain-gut axis may be involved. Thus, in addition to the exaggerated responses of afferent neurons and the recruitment of otherwise “silent” afferent fibers (see previous section), there is also central modulation of afferent stimuli.[2] Perfusion of the lower esophagus with acid increases the sensitivity of the upper esophagus to electrical stimulation, providing direct evidence of central sensitization. Patients with noncardiac chest pain exhibit a marked decrease in their visceral pain threshold.[98] The effects of central sensitization are often relatively long-lasting. For example, irritation of the colon in young rats leads to hypersensitivity in later life despite an absence of apparent pathology in the gut.[99]

The term *wind-up* is sometimes used to describe the increased synaptic transmission at afferent nerve terminals in the spinal cord; this event is thought to involve enhanced transmission by the neurotransmitter glutamate acting at NMDA receptors. In addition, the inhibitory influences on spinal cord cells from nervous pathways that descend in the spinal cord may be altered. Finally, hyperalgesic states clearly are associated with changes in neuronal activation at supraspinal levels, including, for example, the expansion of cortical regions associated with the response to a defined stimulus.[66,96]

EFFERENT FUNCTIONS OF PEPTIDERGIC VISCERAL EFFERENT NEURONS

After the stimulation of visceral afferent nerve fibers, the peptides produced by these neurons are released at both central and peripheral terminals. Peptides released peripherally modulate blood flow, motility, secretion, the discharge of enteric ganglion cells, and a number of functions associated with epithelial protection.[27,28,100] There is now good evidence that noxious agents in the lumen activate splanchnic afferent fibers, which in turn regulate protective or damage-limiting mechanisms within the mucosa, as well as signaling to the CNS (Fig. 5-4). Capsaicin, which is the active ingredient of hot peppers, has been used extensively in studies of primary afferent nerve function.[101] This compound acts on a nonselective cation channel, the vanilloid receptor (VR $_1$), to increase intracellular calcium concentrations.[102] Because the VR $_1$ ion channel is located almost exclusively in primary afferent nerve fibers, capsaicin is a valuable selective stimulant of these neurons. At low doses, capsaicin is excitatory, and at high doses, it produces long-lasting or permanent loss of function.

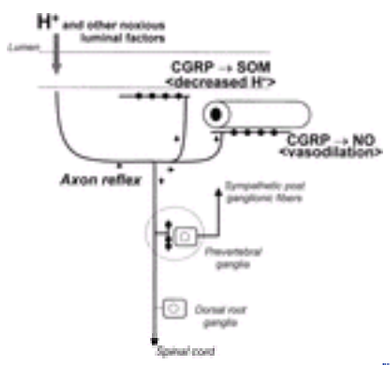

FIGURE 5-4. Axon reflexes in response to noxious chemical stimuli. The latter activate primary afferent neurons that also synthesize neuropeptides (e.g., CGRP) that are transported to their peripheral terminal fields and released by axon reflexes on stimulation. Responses include increased blood flow, probably secondary to stimulation of nitric oxide synthase, and stimulation of somatostatin-producing D cells, which leads to inhibition of acid secretion. *CGRP*, calcitonin gene–related peptide; *SOM,* somatostatin; *NO,* nitric oxide.

Elucidation of the role of peptides released at the peripheral terminals of visceral afferent fibers is complicated for those neuropeptides produced in both afferent and intrinsic neurons (e.g., the tachykinins). However, CGRP-a in the rat stomach has proved to be a valuable model for studies of the relevant mechanisms. CGRP-a is absent from gastric intrinsic neurons and endocrine cells; it is scarce in gastric vagal afferent neurons but abundant in splanchnic afferent nerve fibers. An intact splanchnic afferent innervation mitigates damage of the gastric mucosa inflicted by noxious agents (e.g., ethanol), suggesting that afferent nerve fibers have a protective role.[103] There is good evidence that CGRP-a is a protective mediator in this system. Thus, capsaicin increases the release of CGRP in the perfused rat stomach, and on acute administration, it reduces the lesions produced by ethanol; immunoneutralization of CGRP, or administration of the CGRP antagonist CGRP $_{8–37}$, abolishes capsaicin-induced protection.[100,104,105] The release of CGRP is associated with increased mucosal blood flow mediated by nitric oxide; this hyperemic response is a key part of the protective mechanism (see Fig. 5-4). Multiple other actions of CGRP are likely to be protective, including stimulation of somatostatin secretion and increased somatostatin synthesis at the level of mRNA abundance,[106] which tend to decrease secretagogue-evoked acid secretion.

CNS CONTROL OF THE GUT

The CNS influences all major gastrointestinal functions, including secretion, motility, blood flow, and defense against noxious challenge. A substantial body of literature describes gastrointestinal responses after the administration to defined CNS regions of virtually all the main classic and peptide neurotransmitters, including glutamate, ?-aminobutyric acid, acetylcholine, biogenic amines, corticotropin-releasing factor, opioids, CGRP, CCK, somatostatin, VIP, and neurotensin.[107,108,109] and[110] Both stimulation and inhibition of gastrointestinal motor and secretory responses may be evoked by the same substance in different CNS regions. It is unlikely that all the effects described after the CNS application of neuroactive substances are physiologically relevant. This chapter focuses on the most intensively studied CNS peptide systems regulating gastrointestinal function, particularly those in which the mechanisms and functional significance are clearest. The CNS-peptidergic control of acid secretion has been the subject of several reviews.[109,111]

Thyrotropin-Releasing Hormone

The stimulation of colonic motility and gastric acid secretion in response to the intracisternal administration of TRH was among the first gastrointestinal effects of CNS peptides to be discovered.[112] It is now clear that TRH is a reliable stimulant of a wide range of gastrointestinal responses, including gastric acid and pepsinogen secretion, gastric histamine secretion, gastric mucosal blood flow, gastric motility, and mast cell secretion.[109] In high doses, TRH produces gastric lesions. For the

most part, these effects are mediated by the stimulation of vagal efferent neurons and so are blocked by vagotomy and atropine. They can be mimicked by stable TRH analogs, such as RX77368. There is a rich TRH innervation of the dorsal motor nucleus of the vagus that arises mainly from the obscure and pallidal raphe nuclei. [68] These neurons are also serotonergic. It is likely that the coexisting transmitters, 5-HT and TRH, interact after release. Stimulation of the raphe nuclei, similar to the administration of TRH into the dorsal motor nucleus, increases gastric acid secretion and motility by a vagal cholinergic mechanism. [113], [114] Intracerebroventricular administration of TRH antibodies reduces acid secretion in the pylorus-ligated rat, and immunoneutralization also inhibits the response to raphe nuclei stimulation, providing clear evidence that TRH is an endogenous regulator of the vagal efferent outflow to the upper gastrointestinal tract and may participate in vagovagal reflexes. [113], [115]

Bombesin/Gastrin-Releasing Peptide

Central administration of peptides of the bombesin/gastrin-releasing peptide group can produce effects opposite to those evoked by peripheral administration. In particular, bombesin and gastrin-releasing peptide administered intracisternally inhibit acid secretion, [116] but they generally stimulate it on peripheral administration (reflecting vagal noncholinergic stimulation of the G cell). Retrograde tracing and immunocytochemistry indicate that bombesin-immunoreactive nerve fibers in the dorsal vagal complex originate from cell bodies in the medial parvocellular subdivision of the paraventricular nucleus of the hypothalamus, [117] which provides support for the notion that bombesin-related peptides mediate descending inhibitory effects from the paraventricular nucleus to vagal efferent neurons.

PYY/PP/NPY

The peptides of the PP family (NPY, PYY, and PP itself) influence the brain-gut axis at various levels, almost certainly through a diverse family of receptors (six or more). The different family members vary in their affinity for different, naturally occurring receptors. PP released from endocrine cells of the pancreas is thought to act directly within the vagal complex to stimulate acid secretion and gastric motility. Neurons that are excited by PP have been identified electrophysiologically and characterized as part of a vagovagal circuit activated by antral distention. [118], [119] In contrast, PYY is an inhibitor of acid secretion that is thought to suppress vagal efferent stimuli. Thus, PYY released from the ileum inhibits sham feeding–stimulated acid secretion by a presynaptic effect on the vagal pathway. [120] The complexity of these responses is further illustrated by observations that NPY injected into the paraventricular nucleus inhibited rat gastric acid secretion, but in other hypothalamic regions outside the paraventricular nucleus, it had excitatory effects. [121]

Oxytocin

The intravenous administration of CCK stimulates oxytocin secretion from the posterior pituitary by a mechanism that involves capsaicin-sensitive vagal afferent pathways. [122] Electrophysiological data suggest that oxytocin-secreting neurons of the supraoptic nucleus of the hypothalamus are stimulated by systemic CCK, and by gastric distention, through a vagal mechanism. [123] Intracerebroventricular administration of oxytocin inhibited gastric motility, and an antagonist increased motility, although the antagonist did not influence the response to CCK. [124] Because neurons of the paraventricular nucleus project to the dorsal vagal complex, it seems possible that oxytocin mediates the hypothalamic-vagal pathways regulating digestive function.

VAGOVAGAL REFLEX MECHANISMS

The physiological control of digestion depends on cross talk between the gastrointestinal tract and the brain; the relevant interactions are well illustrated by vagovagal reflexes. Control of pancreatic and gastric secretion, gallbladder contraction, and the motility of stomach and intestine may all include a component that involves a vagovagal reflex. The afferent arm of these reflexes was thought for many years to be triggered by direct mechanical or chemical stimulation of the vagal afferent fibers. It is now clear, however, that local paracrine or hormonal factors also stimulate the afferent arm of vagovagal reflexes; moreover, there may be multiple, interacting inputs to afferent nerve discharge. One of the best-studied examples involves CCK and the reflex regulation of gastric motility and pancreatic secretion. In this context, the primary function of CCK is the regulation of protein and fat digestion in the small intestine. Thus, CCK stimulates the delivery of pancreatic enzymes and bile salts to promote digestion in the intestine; at the same time, it delays further delivery of nutrients to the small intestine by inhibiting gastric emptying and food intake. Taken together, these actions allow matching of the capacity for digestion (enzymes and bile salts) with nutrients already in the small intestine.

Distention of the stomach activates vagovagal reflexes that result in relaxation of the gastric corpus and the stimulation of acid secretion. [125] Gastric relaxation is important early in digestion to accommodate the delivery of food, so that gastric volume increases without an increase in pressure (Fig. 5-5). Electrical stimulation of the vagal nerve trunk generally produces gastric contraction, but relaxation is induced by the same stimulus in the presence of the cholinergic muscarinic antagonist atropine. There are, therefore, a vagal excitatory muscarinic pathway that maintains gastric tone and a vagal inhibitory nonmuscarinic pathway that leads to relaxation of the body of the stomach (and a decrease in intragastric pressure). For some years, VIP was thought to mediate the latter effect, [126] but nitric oxide is also now recognized as a mediator of this response. [127], [128]

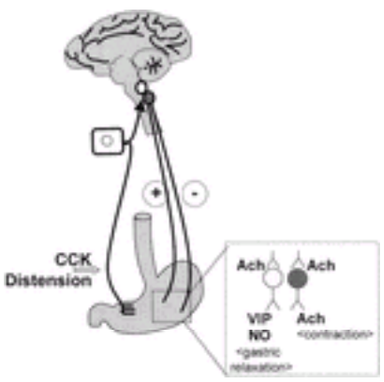

FIGURE 5-5. Vagovagal reflex control of gastric motility. Gastric mechanoreceptors and CCK acting on vagal afferents trigger a vago- vagal reflex that produces inhibition of tone in the gastric corpus. The latter is likely to be secondary both to inhibition of tonically active vagal, cholinergic, excitatory pathways (*shaded*) and to stimulation of vagal inhibitory pathways with VIP and nitric oxide as the final mediators. The reflex is important in receptive relaxation of the stomach early in digestion and in inhibition of gastric emptying (by decreasing the pressure difference between corpus and duodenum) later in digestion. *CCK,* cholecystokinin; *Ach,* acetylcholine; *VIP,* vasoactive intestinal polypeptide.

Neurochemical, electrophysiological, and integrative physiological studies indicate that CCK acts at CCK-A receptors on vagal afferent fibers, which constitute the afferent arm of a vagovagal reflex by which CCK relaxes the gastric corpus [37] (see Fig. 5-5). This response is reduced after capsaicin is topically administered to the vagal nerve trunk, or systemically administered to newborn rats. [129], [130] Lumenal nutrients in the intestine induce CCK release and, like exogenous CCK, relax the gastric corpus by a mechanism sensitive to capsaicin and to CCK-A receptor antagonists. [131], [132] and [133] An important consequence of gastric relaxation in response to CCK is a decrease in the pressure driving the gastric emptying of liquids.

In parallel with the reflex relaxation of the stomach, CCK also appears to activate vagal reflexes leading to the stimulation of pancreatic secretion. It seems that pancreatic acinar cells are controlled both directly by the effects of CCK delivered in the circulation, and indirectly by acetylcholine released from postganglionic parasympathetic neurons as a consequence of CCK-stimulated vagovagal reflexes. [134], [135]

SATIETY MECHANISMS

Food intake is determined by a wide range of influences, including signals from adipose tissue, from the gastrointestinal tract, and arising within the CNS (Fig. 5-6). Together, these mechanisms serve to maintain body weight within narrow limits over long periods in the face of changing metabolic demands. Progress in elucidating the cellular and molecular mechanisms relevant to the control of food intake has been impressive, [136] and it is in part attributable to the study of genetically modified

animals, which has revealed the role of specific transmitters and receptors. [136]

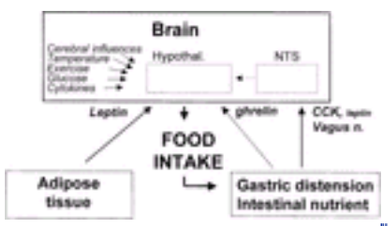

FIGURE 5-6. Schematic representation of the mechanisms controlling food intake. There is inhibition of food intake in the short term (i.e., over the period of a single meal) by gastric distention (which appears not to be nutrient-related) and by the release of CCK from the small intestine (which is related to the fat and protein content of the meal). Both mechanisms result in the activation of vagal pathways terminating in the nucleus of the solitary tract and projecting to the hypothalamus. Feeding inhibits the release of ghrelin from the stomach; ghrelin acts on the hypothalamus to decrease fat utilization. Over the long term, food intake is regulated by leptin released from adipocytes. Among other things, leptin inhibits hypothalamic neuropeptide Y–producing neurons that stimulate food intake. *CCK,* cholecystokinin; *hypothal.*, hypothalamus; *NTS,* nucleus of solitary tract; *vagus n.,* vagus nerve.

Leptin-NPY System

It was thought for many years that fat stores might somehow signal to the CNS to influence food intake (i.e., the lipostatic theory of food intake). The molecular basis for this mechanism has emerged from the characterization of a genetic defect in mice (*ob/ob*)—a homozygous recessive mutation associated with obesity, hyperphagia, decreased locomotion, hypometabolism, diabetes, and infertility. Zhang and colleagues [137] showed by positional cloning that these mice have a mutation in a gene encoding a putative hormone signaling from adipose tissue to the CNS, now known as *leptin*. Subsequently, mutations in the leptin gene were also reported in two obese human subjects. [138] The receptor for leptin is mutated in a different mouse strain (*db/db*) that is also characterized by hyperphagia, hypometabolism, and diabetes. [139] The intracellular signaling pathway activated by the leptin receptor appears to resemble the JAK/STAT pathway. [140] The administration of leptin reduced body weight in *ob/ob* mice and increased energy expenditure. [141], [142] There appear to be important interactions between leptin and several hypothalamic systems. One involves NPY, which is a powerful stimulant of food intake when administered into the ventromedial and paraventricular hypothalamic nuclei. [143]

Leptin secretion reflects the size of the adipose tissue mass, and as leptin reduces NPY synthesis and release, it provides a negative feedback influence on the hypothalamic system that stimulates feeding. [144] Deletion of the NPY gene in *ob/ob* mice partially reverses the phenotype of the latter, providing direct support for the idea that leptin acts through the modulation of NPY release. [145]

The melanocortin-4 (MC-4) receptor is also implicated in the response to leptin. Mice of the agouti strain are hyperphagic and obese, with elevated plasma leptin concentrations; they generate an endogenous peptide antagonist of the MC-4 receptor that, in addition to having CNS effects, blocks the effects of the melanocyte-stimulating hormone on skin melanocytes, giving the animals a yellow color (hence agouti). The intracerebroventricular administration of synthetic MC-4 receptor agonists inhibited feeding in agouti and *ob/ob* mice, and in mice injected with NPY. [146] Moreover, hyperphagia, hyperinsulinemia, and hyperglycemia, similar to that of agouti mice, developed in mice in which the MC-4 receptor had been inactivated. [147] The data suggest multiple signaling systems in the hypothalamus that are activated by leptin. One system is linked to increased feeding and is mediated by NPY; another, which leads to the inhibition of feeding, is linked to melanocyte-stimulating hormone and the MC-4 receptor. However, these are certainly not the only hypothalamic transmitter systems relevant to the control of food intake. Others that are part of this system include catecholamines, 5-HT, glucagon-like peptide I, melanocyte-concentrating hormone, and orexins. [148], [149] and [150]

Gastrointestinal Satiety Signals

The leptin system is thought to provide a mechanism to match energy stores in the body with food intake, and to function as a relatively long-term signaling system. Signals from the gastrointestinal tract that control food intake operate over shorter intervals and limit the size of individual meals, thereby serving to match meal size with the capacity for digestion. Gut hormones are likely to be important regulators of these events, and, as already mentioned, there is considerable support for the idea that CCK may signal to the CNS through vagal afferent fibers. [151], [152] and [153] Thus, exogenous CCK inhibits food intake; this effect is inhibited by abdominal vagotomy, lesioning of the NST, sectioning of vagal afferent rootlets, and the use of capsaicin. [154], [155], [156] and [157] Moreover, the occurrence of missense mutations in the CCK-A receptor in some obese patients provides molecular support for the idea that CCK has a functional role at CCK-A receptors in the control of food intake in humans. [158] It has been suggested that leptin may also act as a gastrointestinal satiety signal. Leptin is produced in gastric chief cells (as well as in adipocytes), and it has been suggested that CCK induces the release of gastric leptin. [159], [160] There appear to be functional leptin receptors on vagal afferent neurons and to be interactions between leptin and CCK in stimulating the discharge of gastric vagal afferent fibers, suggesting a local gastric leptin system that potentiates the satiety effects of CCK. [161]

Physiologically, it is likely that both nutrient and non–nutrient-activated mechanisms contribute to the gastrointestinal control of food intake. One important nonnutritive signal is the volume of the gastric contents. Gastric distention (regardless of nutrient content) inhibits food intake by a vagal mechanoreceptor mechanism. [162], [163] and [164] It seems, however, that there are also interactions between CCK-mediated (nutrient-dependent) mechanisms and gastric volume (non–nutrient-dependent) because there is evidence for potentiation between gastric distention and CCK in the inhibition of food intake. [165] There appears, then, to be synergistic interactions between CCK and leptin, and CCK and nonnutritive stimuli, at the peripheral end of gastric vagal afferent fibers. Further studies are needed to elucidate the CNS projections from the NST to the hypothalamus for nutrient- and non–nutrient-initiated stimuli and to define the ways in which these pathways are integrated with signals from the leptin-adipocyte pathways to control food intake.

In addition to signals from the gut that inhibit food intake, it now appears that there may be other types of gut-brain signals. The peptide ghrelin was discovered as the ligand of orphan receptors that mediate the action of synthetic secretagogues on growth hormone release. [166], [167] Interestingly, ghrelin both releases growth hormone and increases adiposity in rats by decreasing fat utilization. Ghrelin is mainly produced in the stomach; plasma concentrations are increased by fasting and decreased by feeding. [168] It has been suggested that ghrelin signals from the stomach to the hypothalamus to increase metabolic efficiency (see Fig. 5-6).

OVERVIEW

The gastrointestinal tract and brain involve each other in two-way communication. These interactions provide a basis for the gastrointestinal control of CNS responses (e.g., control of food intake) and, conversely, the CNS control of gastrointestinal function (e.g., during the cephalic phase of digestion). More importantly, they provide the capacity for integrating information about the state of the gut contents and inputs from other parts of the body. A direct consequence of these integrative functions is modulation of the signaling between brain and gut during normal digestion. However, the modulation of signaling pathways is also likely to be important clinically, and it may account for the hypersensitivity that is a feature in disease states involving inflammation in the gut, as well as those that do not. The results of a variety of experimental approaches have shown that it is possible to explain in molecular and cellular terms the development of the brain-gut axis and its organization, neurochemistry, and physiology. Impressive progress also is being made in the combined application of CNS imaging techniques and functional studies of the human digestive tract. These approaches should make it possible to define how dysfunction of brain-gut signaling contributes to gastrointestinal disease.

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CHAPTER 6

Gabriel M. Makhlouf

SMOOTH MUSCLE OF THE GUT

The main function of smooth muscle of the gut is to mix and propel intralumenal contents, to enable efficient digestion of food, progressive absorption of nutrients, and eventual evacuation of residues. This function is regulated by the intrinsic electrical and mechanical properties of smooth muscle, such as the ability to maintain tone or undergo phasic contraction, and by alterations in these properties in response to hormonal and neural signals, particularly signals emanating from the enteric nervous system. A distinctive feature of physiological regulation in the gut is that stimuli of hormonal release and neural activation arise within the lumen from the mechanical and chemical properties of food and digestive secretions.

STRUCTURE OF SMOOTH MUSCLE

Muscle Layers

Smooth muscle of the gut consists of a thin, outer, longitudinal layer and a thick, densely innervated, circular layer; the layers derive their names from the orientation of the long axis of muscle cells in them. The layers are separated by laminar septa into bundles about 1 mm long that probably act as contractile units. The muscle cells are embedded in a connective tissue matrix, a product of their synthetic and secretory activity consisting mainly of elastic and collagen fibrils. The layers include glial cells, fibroblasts, and a distinctive population of muscle-like cells, the interstitial cells of Cajal.

Muscle Cells: Membranes and Organelles

Single smooth muscle cells are about 400 µm long and 5 µm wide when fully relaxed. They are spindle-shaped and have a high ratio of surface area to volume (1.5 $\mu m^2/\mu m^3$). Their plasma membranes consist of two specialized structures known as *caveolae* and *dense bands.* [1], [2] and [3]

The caveolae, 70 nm wide and 120 nm deep, are basket-shaped invaginations of the membrane that are arranged in clusters (Fig. 6-1). There are about 150,000 caveolae per cell, and they occupy about one third of the outer surface of the cell but constitute a much larger fraction of the surface of the plasma membrane. The bases of caveolae are surrounded by an abundant endoplasmic reticulum, the site of Ca^{2+} storage and release in smooth muscle. The arrangement suggests that caveolae may be functional equivalents of the transverse tubules in striated muscle. Caveolin-1a, caveolin-1ß, and caveolin-3 are the main structural proteins in smooth muscle caveolae. [4], [5] They bind a variety of signaling molecules and are thought to act as scaffolds to facilitate signaling. In smooth muscle, caveolins also bind transiently to agonist-activated a and ß? subunits of G proteins, prevent their reassociation, and induce desensitization of responses mediated by these G proteins. [6]

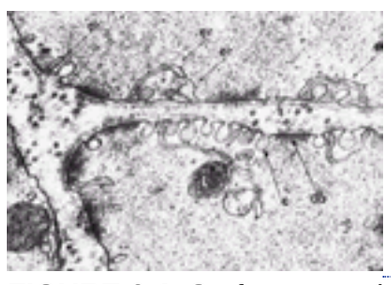

FIGURE 6-1. Surface organization of three adjacent muscle cells from the circular muscle layer of guinea pig ileum. Clusters of basket-shaped caveolae (*c*) surrounded by endoplasmic reticulum (*er*) are separated from each other by dense bands (*db*) (original magnification ×67,000). (From ref. [1].)

Clusters of caveolae are separated from each other by electron-dense structures, 1 to 2 µm long and 0.2 to 0.4 µm wide, called dense bands, that occupy about one half of the surface of the cell (Fig. 6-2). Dense bands are points of attachment of thin actin filaments; like dense bodies, their counterpart in the cytoplasm, they consist mainly of actinin, a protein that is a major component of the Z-line in striated muscle. Intermediate 10-nm-thick filaments, consisting mainly of desmin in visceral smooth muscle, link dense bands in the membrane to dense bodies in the cytoplasm and transmit the force generated by the contractile apparatus within the

cell to the entire surface of the cell. [7]

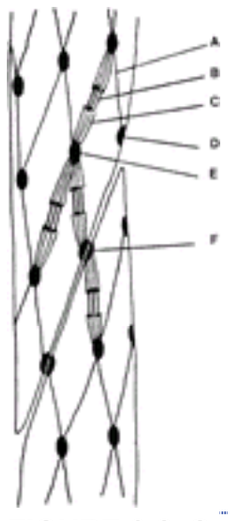

FIGURE 6-2. Organization of the contractile and cytoskeletal apparatus in smooth muscle cells. Thin actin filaments (*C*) emerge from the poles of cytoplasmic dense bodies (*E*) and interdigitate with thick myosin filaments (*B*). Dense bodies (*D*) in the plasma membrane are connected to dense bodies (*E*) in the cytoplasm by intermediate filaments (*A*). When juxtaposed, dense bodies from adjacent cells can form close intermediate junctions (*F*). (Adapted from ref. [7].)

Some dense bands are juxtaposed to dense bands in adjacent cells. At these locations, called *intermediate junctions,* the intercellular space narrows to less than 30 nm and is filled with condensed extracellular matrix (see Fig. 6-2). At other locations, dense bands in one cell are linked by collagen fibrils to dense bands in adjacent cells. Together, intermediate junctions and collagen fibrils anchor adjacent cells to each other, transmit force from one cell to the next, and couple the contractile apparatus of adjacent cells to the rest of the syncytium.

In some regions, patches of the plasma membrane of adjacent cells are closely apposed, and the space between them is reduced to less than 3 nm; the space is bridged by intercellular channels that permit free movement of ions and small molecules. These patches, known as *gap junctions* or *nexuses,* are the most likely sites of electrical coupling between muscle cells. [1], [2] and [3], [7], [8] and [9] Gap junctions permit movement of intracellular regulatory molecules, such as cyclic AMP (cAMP), inositol 1,4,5-triphosphate (IP_3), or Ca^{2+}, and they help to propagate the signal from cell to cell.

Dense bodies and contractile filaments occupy about 80% of the interior of the cell; the remainder is occupied by various organelles, including the nucleus, mitochondria, Golgi apparatus, lysosomes, and rough and smooth endoplasmic reticulum. The last, located immediately beneath and parallel to the plasma membrane, occupies 2% of the cell volume and is the site of Ca^{2+} uptake and release. [10], [11], [12] and [13] It consists of several functional compartments, only one of which is sensitive to IP_3, the membrane-derived messenger responsible for the release of intracellular Ca^{2+} (see Fig. 6-1). [14] The mitochondria, considered a low-affinity, high-capacity storage site, can take up large amounts of Ca^{2+} but only after cell injury, when cytosolic Ca^{2+} increases to more than maximal physiological levels (>1–5 µM). [15]

Contractile Apparatus: Thin and Thick Filaments

Three types of filaments can be differentiated in smooth muscle cells: thin actin filaments (5–7 nm), thick myosin filaments (15 nm), and intermediate desmin filaments (10 nm). Intermediate filaments link dense bodies in the cytoplasm to dense bands on the plasma membrane. Although the arrangement of thin, thick, and intermediate filaments and their attachments to cytoplasmic dense bodies lack the order found in striated muscle, assemblies reminiscent of primitive sarcomeres can be seen.

Thin filaments consist of actin, a ubiquitous, 42-kDa, globular protein (G actin) that polymerizes to form two-stranded helical filaments (F actin) of indeterminate length. [16] Inserted into the grooves of the actin helix is another protein, tropomyosin. Thin filaments have a distinct polarity, and they appear to be inserted into or emerge from the poles of dense bodies and are arranged in bundles that run parallel to the long axis of the cells, with their free ends surrounding and interdigitating with thick myosin filaments (see Fig. 6-2). [1], [2] and [3], [17] The insertion of thin filaments in dense bodies is analogous to that found in Z discs of striated muscle cells; in effect, dense bodies may be viewed as dispersed fragments of Z discs held together and anchored to dense bands of the cell membrane by intermediate filaments.

Thick filaments are aggregates of myosin molecules, a complex 480-kDa protein formed by the association of six different proteins. [16], [17] and [18] These proteins are not covalently linked and can be dissociated from each other into one pair of myosin heavy chains and two pairs of myosin light chains (MLCs) (Fig. 6-3). The heavy chains are coiled around each other to form a rigid, insoluble, helical core or tail. Each strand of the core terminates in a globular head surrounded by two MLCs: a 20-kDa regulatory chain and a 17-kDa essential chain. Each globular head contains a binding site for actin and an actin-activated Mg^{2+}-ATPase.

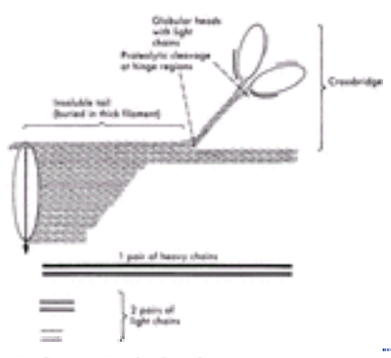

FIGURE 6-3. Component proteins of the myosin molecule. A crossbridge projects laterally from the main segment of the myosin core (i.e., tail). Each crossbridge consists of a pair of myosin heads, each surrounded by a 17-kDa and a 20-kDa myosin light chain and a laterally projecting segment of the myosin core between the two hinge regions. The cores of many myosin molecules form thick filaments with several projecting crossbridges. (Adapted from ref. [7].)

A hinge located at the junction of the globular head and the core enables the head to rotate about the core. Another hinge in the core enables the globular heads to project laterally. The globular heads and the segments of the core between the two hinges are called *crossbridges* because they constitute a link or bridge between thick myosin and thin actin filaments.

Only three to five thick filaments are surrounded by and interdigitate with a much larger number of actin filaments. The ratio of thin to thick filaments is reflected in the relative content of actin and myosin. Visceral smooth muscle contains the same amount of actin as striated muscle (22 versus 28 mg/g of cell) but a lower amount of myosin (20 versus 62 mg/g of cell). [16] Despite the low content of myosin, smooth muscle generates as much force as striated muscle (up to 6 kg/cm^2 of cross-sectional area). [7], [19]

INTERACTION OF CONTRACTILE PROTEINS

The interaction of myosin and actin with hydrolysis of ATP is the fundamental reaction whereby chemical energy is converted into mechanical energy in smooth muscle. The reaction generates force and induces shortening as a result of the sliding of overlapping, interdigitating thin and thick filaments.

Phosphorylation of Myosin Light Chain

An essential step in smooth muscle contraction is phosphorylation of the 20-kd regulatory MLC (MLC $_{20}$) by MLC kinase. [16], [20], [21], [22], [23] and [24] Several steps lead to the activation of this Ca^{2+}-dependent enzyme. An increase in cytosolic free Ca^{2+} occurs when the cell is stimulated. The increase results from the influx of Ca^{2+} into the cytosol through voltage-gated Ca^{2+} channels or from the release of Ca^{2+} into the cytosol from intracellular Ca^{2+} stores. Ca^{2+} sequentially binds to the four binding sites on the regulatory protein calmodulin; the binding of Ca^{2+}-activated calmodulin to MLC kinase forms the active complex Ca^{2+}-calmodulin–MLC kinase. Phosphorylation of MLC $_{20}$ induces a conformational change in the myosin head that greatly enhances the ability of actin to activate myosin–Mg^{2+}-ATPase and to induce the hydrolysis of ATP bound to the myosin head.

Interaction of Myosin and Actin in the Crossbridge Cycle

The interaction of myosin and actin with hydrolysis of ATP occurs in a cycle, the crossbridge cycle, the essential feature of which is a shift in the affinity of myosin for actin. [16] ATP, bound weakly to myosin, is hydrolyzed to ADP and inorganic phosphate (P_i). The products of hydrolysis remain bound to the myosin head, and the energy released is stored in the myosin molecule, which has a high affinity for actin in this state. On the release of ADP and P_i, ATP binds again to myosin, which then reverts to a state of low affinity for actin.

Several models have been proposed to describe how the interaction between phosphorylated myosin heads and actin filaments could generate force, filament movement, and cell shortening. [16], [25] According to one model (Fig. 6-4), the myosin crossbridge (which includes the myosin head, associated MLCs, and a segment of the myosin core) with ATP bound to the myosin head (myosin·ATP) is detached or weakly bound to actin. Hydrolysis of ATP by myosin–Mg^{2+}-ATPase yields a myosin intermediate (myosin·ADP·P_i). Release of P_i from this intermediate is the force-generating step that causes a transition from weak to strong binding of myosin and actin (actin·myosin·ADP) and a change from 90° to 45° in the angle of the myosin head. The change of angle imposes a strain on the crossbridge that is relieved when the actin molecule slides past the crossbridge. In the next step, ADP is released from the myosin head. This step occurs slowly in smooth muscle and limits the velocity of shortening. In smooth muscle, myosin therefore remains bound to actin for a longer period, and it generates more force with less consumption of energy than in striated muscle. After release of ADP, the myosin head is free to rebind ATP (myosin·ATP) and reverts to a 90° angle in which it is weakly bound to the next actin molecule for the start of another cycle.

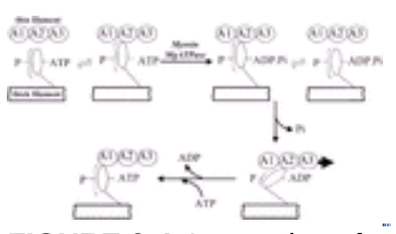

FIGURE 6-4. Interaction of myosin and actin filaments: the crossbridge cycle. At the start of the cycle, the myosin head with ATP bound to it (*myosin·ATP*) is detached or is weakly attached to an actin molecule (*A2*) in the thin filament. Hydrolysis of ATP by the myosin–Mg^{2+}-ATPase yields an intermediate with the products of hydrolysis still bound to it (*myosin·ADP·*P_i); the intermediate is detached or weakly bound to the actin molecule. Release of P_i in the next step causes a transition from weak to strong binding of myosin and actin (*actin·myosin·ADP*) and a change from 90° to 45° in the angle of the myosin head. The strain imposed on the crossbridge by the change of angle is relieved when the actin molecule slides past the crossbridge. ADP is released slowly; the myosin head rebinds ATP (*myosin·ATP*) and reverts to a 90° angle facing the next actin molecule (*A1*) for the start of another cycle. Dephosphorylation of the myosin light chain surrounding a bound myosin head yields a strongly attached, slowly cycling, latch crossbridge. The interaction of only one myosin head in the crossbridge is illustrated. P_i, inorganic phosphate. (Adapted from ref. [16].)

The force generated by crossbridge cycling depends on the number of crossbridges acting in parallel. Because the crossbridges do not cycle in unison, the contraction induced by their concerted action is slow and continuous. Force and shortening velocity depend on the stimulus in smooth muscle; this finding implies that, unlike in striated muscle, the number and cycling rate of activated crossbridges in smooth muscle are regulated.

Crossbridge cycling ceases after the stimulus is withdrawn. Cytosolic Ca^{2+}, MLC kinase activity, and MLC $_{20}$ phosphorylation revert to their resting levels, and the dephosphorylated myosin crossbridges are arrested in a detached state that is characteristic of relaxed muscle. [19], [26]

A different pattern of crossbridge cycling is observed during sustained (i.e., tonic) contraction of smooth muscle. A rapid increase in cytosolic Ca^{2+}, MLC kinase activity, MLC $_{20}$ phosphorylation, and shortening velocity during the initial phase reflects a rapid crossbridge cycling rate; these events precede but are correlated with the increase in muscle tension. [19], [22], [26] Within seconds, despite continued stimulation, cytosolic Ca^{2+} levels and MLC kinase activity decrease to very low levels, whereas MLC $_{20}$ phosphorylation and muscle contraction decrease only slightly (Fig. 6-5). [27] This type of contraction is functionally useful in muscle that is required to maintain the dimensions of a hollow organ against imposed loads with minimal energy consumption. [19]

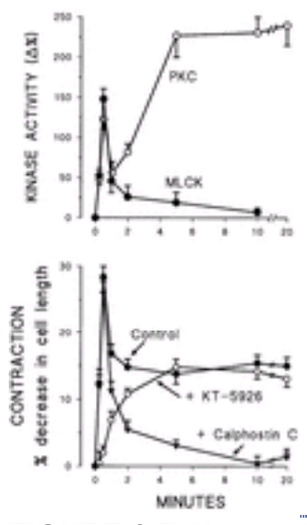

FIGURE 6-5. Upper panel: Time course of activation of protein kinase C (*PKC*) and myosin light-chain kinase (*MLCK*). MLCK activity occurs mainly during the initial phase of contraction. PKC activity is derived from phosphoinositide hydrolysis by PLC-ß during the initial phase and from phosphatidylcholine hydrolysis by phospholipase D during the sustained phase. **Lower panel:** Initial contraction is mediated by Ca^{2+}/calmodulin–dependent activation of MLCK and is inhibited by the MLCK inhibitor, KT-5926; sustained contraction is mediated by PKC and is inhibited by the PKC inhibitor, calphostin C. See details of the pathways in the text and in

Figure 6-6.

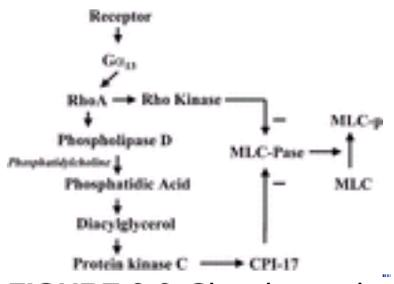

FIGURE 6-6. Signal transduction pathway mediating sustained contraction of smooth muscle. The pathway involves activation of the a subunit of G_{13}, RhoA and its associated kinase, and phospholipase D. Hydrolysis of phosphatidylcholine by phospholipase D generates phosphatidic acid, which is dephosphorylated to diacylglycerol, with resulting activation of protein kinase C. Protein kinase C potently activates CPI-17, an endogenous inhibitor of myosin light chain (*MLC*) phosphatase activity, thereby maintaining MLC phosphorylation (*MLC-p*). A parallel pathway links Rho kinase to the inhibition of MLC phosphatase.

Sustained MLC_{20} phosphorylation and contraction are now known to be Ca^{2+}-independent. MLC_{20} phosphorylation is determined by a signaling pathway that involves sequential activation of the heterotrimeric G protein, G_{13}, and the monomeric G protein, RhoA, followed by activation of Rho kinase, phospholipase D, and one or more isoforms of protein kinase C (PKCe with agonists and PKCa with growth factors), which results in inhibition of MLC phosphatase and maintenance of MLC_{20} phosphorylation and contraction (Fig. 6-6). [28], [29], [30] and [31] An endogenous 17-kd inhibitory protein, CPI-17, that is greatly activated by PKC and inhibits the catalytic subunit of MLC phosphatase, provides the link between PKC activation and MLC phosphatase inhibition. [32], [33]

Unlike tonic contraction, phasic (rhythmic) contraction increases and decreases rapidly in phase with the influx and efflux of Ca^{2+}, driven by rhythmic changes in membrane potential and the opening and closure of voltage-gated ionic channels. A closer correlation prevails during phasic activity among Ca^{2+} levels, MLC kinase activity, MLC_{20} phosphorylation, crossbridge cycling rate, and contraction. Consequently, more energy is required to sustain phasic contraction. Energy is saved, however, because phasic contractile activity usually is maintained in abeyance by a dominant inhibitory neural input.

SIGNAL TRANSDUCTION IN VISCERAL SMOOTH MUSCLE

Source of Activator Calcium

The concentration of Ca^{2+} in the cytosol is the essential determinant of the initial, transient phase of smooth muscle contraction. Two agonist-driven mechanisms lead to an increase in cytosolic Ca^{2+} during contraction. In the first, interaction of a contractile agonist with its receptor on the plasma membrane generates a messenger that causes the release of Ca^{2+} from intracellular stores. In the second, interaction of the contractile agonist with its receptor generates a messenger that induces depolarization of the plasma membrane, which opens voltage-gated Ca^{2+} channels and causes Ca^{2+} influx, followed by Ca^{2+}-induced Ca^{2+} release. The first mechanism occurs in the circular muscle layer, the second in the longitudinal muscle layer. Spontaneous depolarization of the plasma membrane by slow waves or spike potentials can open voltage-gated Ca^{2+} channels in muscle from both layers. By increasing the frequency or amplitude of these depolarizing events, agonists can cause further increase in Ca^{2+} influx through voltage-gated Ca^{2+} channels.

Components of the Transduction Pathway

The transduction of an external signal, such as a neurotransmitter or a hormone, into an internal signal involves sequential activation of three membrane proteins: a receptor and a guanosine triphosphate (GTP)–binding protein (G protein) that couples the receptor to a specific effector enzyme. The effector enzyme acts on membrane-bound or cytoplasmic precursors to generate one or more regulatory signals or second messengers (Fig. 6-7 and Fig. 6-8; see Fig. 6-6).

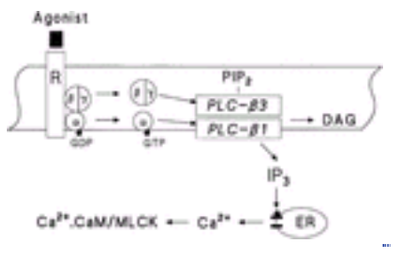

FIGURE 6-7. Signal transduction pathway for Ca^{2+} mobilization in smooth muscle cells of the circular muscle layer during the initial phase of contraction. The sequence involves binding of an agonist to a specific receptor, activation of a transducing G protein and binding of guanosine triphosphate (*GTP*) to its mobile a subunit, and dissociation of the ß? subunits. For receptors that couple to G_q (e.g., CCK-A, muscarinic M_3, 5-HT_2), Ga_q activates phospholipase Cß1 (PLC-ß1); for receptors that couple to G_i, (e.g., muscarinic M_2, adenosine A_1, opioid µ, ?, and d) G_{i}ß? activates PLC-ß3. In circular muscle cells, the immediate substrate hydrolyzed by PLC-ß1 or PLC-ß3 is phosphatidylinositol 4,5-bisphosphate (PIP_2), yielding inositol 1,4,5-triphosphate (IP_3) and diacylglycerol (*DAG*) as second messengers. IP_3 diffuses through the cytosol to interact with IP_3 receptors–Ca^{2+} channels on the membrane of the endoplasmic reticulum (*ER*), causing release of Ca^{2+} into the cytosol. Ca^{2+} binds to calmodulin (*CaM*), and the complex activates MLC kinase (*MLCK*). (See refs. [41] and [55].)

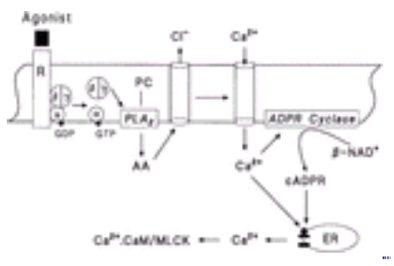

FIGURE 6-8. Signal transduction pathway for Ca^{2+} mobilization in smooth muscle cells of the longitudinal muscle layer during the initial phase of contraction. The sequence involves activation of phospholipase A_2 (PLA_2) by Gß? and hydrolysis of membrane-bound phosphatidylcholine (*PC*) to yield arachidonic acid (*AA*) and lysophosphatidylcholine. Arachidonic acid activates chloride (Cl^-) channels causing Cl^- efflux from the cell, resulting in depolarization of the plasma membrane, opening of voltage-gated Ca^{2+} channels, and Ca^{2+} influx into the cell. The resultant increase in cytosolic Ca^{2+} stimulates Ca^{2+} release (i.e., Ca^{2+}-induced Ca^{2+} release) from endoplasmic Ca^{2+} stores through ryanodine-sensitive receptor–Ca^{2+} channels. In addition, Ca^{2+} activates a membrane-bound ADP–ribosyl cyclase (*ADPR cyclase*) that synthesizes cyclic ADP ribose (*cADPR*) from ß-nicotinamide adenine dinucleotide (ß-NAD^+). cADPR acts as a Ca^{2+}-mobilizing messenger, stimulating Ca^{2+} release by directly activating ryanodine receptor–Ca^{2+} channels and by potentiating Ca^{2+}-induced Ca^{2+} release. (See refs. [57] and [59].)

Receptors consist of external, membrane-spanning, and cytoplasmic domains with features that determine which specific ligand they bind or agonist they recognize and which G protein or membrane enzyme they activate. [34], [35]

G proteins are a large family of closely related proteins that act as signal transducers. [36], [37] Among these are G proteins that stimulate (G_s) or inhibit (G_i and G_o) adenylate cyclase activity and G proteins that stimulate phospholipase C (PLC) activity (G_q). G proteins are heterotrimeric with subunits designated a, ß, and ? in order of decreasing mass. The a subunit (Ga), which serves to differentiate G proteins, contains a single, high-affinity binding site for GTP and possesses GTPase activity; the latter is crucial for terminating the action of G proteins. In the basal state, guanosine diphosphate (GDP) is tightly bound to the a subunit. The binding of a ligand to its receptor enables the ligand-receptor complex to interact with the G protein and stimulate the dissociation of GDP; this opens up a site that is rapidly filled with abundant cytoplasmic GTP. The binding of GTP to the ligand-receptor–G-protein complex causes the following:

1. A decrease in the affinity of the ligand for the receptor and of the receptor for the G protein, thus freeing the receptor for a new cycle of ligand and G-protein binding
2. A decrease in the affinity of the a subunit for the ß? subunit, resulting in the dissociation of a Ga-GTP complex and a ß? dimer that activate different effector enzymes.

The hydrolysis of GTP by the intrinsic GTPase activity of the Ga-GTP complex terminates the activity of the complex; the inactive Ga-GDP complex binds to and inactivates ß? and enables the reassociation of the a, ß, and ? subunits. The slow hydrolysis of GTP is accelerated by two classes of GTPase-activating proteins (GAPs): one class includes effector enzymes such as PLC-ß; the other consists of a group of Ga-GAPs, also known as *regulators of G-protein signaling* (RGS). Certain RGS proteins have been identified that appear to be specific for various a subunits.

G proteins act as transducers that conduct and amplify the external signal and as adapters that allow the same receptor to be coupled to different effector enzymes. The initial focus on a subunits has shifted with the realization that ß? subunits can also activate or inhibit effector enzymes. The involvement of a or ß? subunits depends on the specific receptor activated (see later).

Effector Enzymes and Second Messengers

Several effector enzymes capable of yielding different messengers are involved in the regulation of smooth muscle contraction. They include PLC-ß isoforms, which are activated by the a subunit of G_q or the ß? subunits of G_i and G_o (see Fig. 6-7), phospholipase A_2 (PLA_2), which is activated by G_i (see Fig. 6-8); and phospholipase D, which is activated during the sustained phase of contraction by the a subunit of G_{13} and RhoA (see Fig. 6-6). All can hydrolyze membrane-bound phospholipids. In most cell types, the effector enzyme activated by agonists capable of mobilizing intracellular Ca^{2+} is one of several isoforms of PLC-ß (i.e., PLC-ß1, PLC-ß2, PLC-ß3, and PLC-ß4); the isozymes hydrolyze inositol phospholipids located on the inner leaflet of the plasma membrane. [14], [38], [39], [40], [41] and [42] These phospholipids are products of the sequential phosphorylation of phosphatidylinositol (PI) to phosphatidylinositol monophosphate (PIP) and phosphatidylinositol 4,5-bisphosphate (PIP_2). The last is the immediate substrate hydrolyzed by PLC-ß.

The hydrolysis of PIP_2 generates two messengers: a water-soluble inositol phosphate, IP_3, which diffuses into the cytosol to activate IP_3 receptor–Ca^{2+} channels located in a compartment of the endoplasmic reticulum; and diacylglycerol, which activates various Ca^{2+}-dependent (e.g., a, ß, ?) and Ca^{2+}-independent (e.g., e) isoforms of PKC and initiates their translocation from the cytosol to the plasma membrane (see Fig. 6-7). [40] IP_3 can be converted by sequential phosphorylation to IP_4, IP_5, or IP_6 or by dephosphorylation to IP_2, IP, and inositol. The metabolic products of the two messengers eventually merge to reconstitute PI. Partial depletion of Ca^{2+} stores by IP_3 triggers Ca^{2+} influx into the cell—a process known as capacitative Ca^{2+} influx—and leads to Ca^{2+} release from adjacent Ca^{2+} stores (i.e., Ca^{2+}-induced Ca^{2+} release). The process is manifested by localized changes in Ca^{2+} concentrations (Ca^{2+} oscillations) that sweep through the cell at intervals of 5 to 60 seconds, and it is most effective in the presence of IP_3. [14], [43] IP_3 and Ca^{2+} can flow rapidly (about 10 µm/s) through gap junctions to neighboring cells, propagating the intracellular signal and providing a means for sustained or oscillatory response of the tissue as a whole.

The IP_3 receptor–Ca^{2+} channel is homologous to the ryanodine receptor–Ca^{2+} channel in skeletal and cardiac muscle. [44], [45] and [46] Both receptor types consist of four subunits surrounding a Ca^{2+} channel; each subunit contains a large N-terminal cytoplasmic domain that includes binding sites for IP_3 and Ca^{2+} and the site for regulatory phosphorylation (i.e., inactivation) by cGMP-dependent protein kinases (PKG and PKA). The cytoplasmic domain appears to bridge the space between the endoplasmic and plasma membranes and is postulated to participate in mediating capacitative Ca^{2+} influx. [47]

The involvement of IP_3 in Ca^{2+} mobilization is confined to circular smooth muscle. In longitudinal smooth muscle, Ca^{2+} mobilization involves PLA_2, which hydrolyzes the membrane phospholipid phosphatidylcholine to yield arachidonic acid and lysophosphatidylcholine (see later).

Mobilization of Calcium in Cells of the Circular Muscle Layer

The transduction pathway initiated by the hydrolysis of PIP_2 is fully expressed in cells from the circular muscle layer of the stomach, intestine, gallbladder, and various sphincters. The various steps in this pathway have been examined in detail in dispersed muscle cells devoid of neural elements. The cellular homogeneity of the suspension makes it possible to characterize receptors and intracellular messengers, such as IP_3, diacylglycerol, cytosolic Ca^{2+}, and cyclic nucleotides, and to determine their coupling to mechanical responses, such as contraction and relaxation. [12], [13], [47], [48], [49], [50] and [51]

The exposure of cells derived from the circular muscle layer to a contractile agonist induces rapid contraction (i.e., cell shortening) accompanied by a transient increase in IP_3, cytosolic Ca^{2+}, and Ca^{2+} release (Fig. 6-9 *A*). [12], [13], [51], [52] and [53] These events are closely correlated, and their magnitudes depend on agonist concentration (Fig. 6-9B). The initial increase in IP_3 and Ca^{2+} is followed by the slow reuptake of Ca^{2+} into the cell (capacitative Ca^{2+} influx). The withdrawal of Ca^{2+} from the medium or the addition of Ca^{2+} channel blockers has no effect on the initial increase in Ca^{2+}, but it blocks Ca^{2+} influx. The pattern of response in muscle cells from the circular muscle layer implies that contractile agonists elicit an initial contraction by means of G-protein–dependent activation of PLC-ß, hydrolysis of membrane-bound PIP_2, and generation of IP_3; the latter diffuses through the cytosol to interact with sarcoplasmic IP_3 receptor–Ca^{2+} channels to induce Ca^{2+} release and a transient increase in Ca^{2+}. Depletion of Ca^{2+} stores triggers capacitative Ca^{2+} influx.

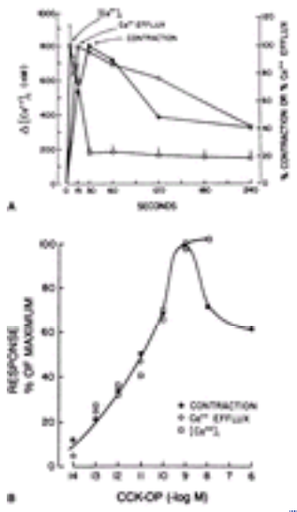

FIGURE 6-9. Time course (**A**) and stoichiometry (**B**) of contraction, cytosolic Ca $^{2+}$ ($[Ca^{2+}]_i$), and Ca $^{2+}$ release from endoplasmic stores, in smooth muscle cells isolated from guinea pig stomach in response to cholecystokinin octapeptide (*CCK-OP*). Similar results are obtained for muscle cells from human stomach and intestine using other agonists. (From ref. [51].)

Studies using a variety of probes confirm this sequence. Direct activation of G proteins by sodium fluoride or GTP?S mimics the effects of receptor-linked agonists, by causing contraction and an increase in IP $_3$ and cytosolic Ca $^{2+}$ levels; the effects of GTP?S, like those of agonists, are abolished by GDPßS. [53], [54] Exogenous IP $_3$ causes contraction (median effective concentration, EC $_{50}$ 0.3 µM), Ca $^{2+}$ release, and an increase in cytosolic Ca $^{2+}$. [13], [53] The responses to IP $_3$, GTP?S, and agonists are inhibited by heparin, which blocks IP $_3$-induced Ca $^{2+}$ release. [53], [54] and [55] In permeabilized muscle cells and in microsomal fractions, IP $_3$, but not other inositol phosphates, binds with high affinity to specific IP $_3$ receptors, and its binding is accompanied by Ca $^{2+}$ release. [55] Although most receptors use G $_q$a and PLC-ß1 to mediate PIP $_2$ hydrolysis, others use G $_{i/o}$ß? and PLC-ß3. Cholecystokinin (CCK) acting through CCK-A receptors, serotonin (5-hydroxytryptamine, 5-HT) through 5-HT $_2$ receptors, and tachykinins through NK $_1$ and NK $_2$ receptors preferentially activate PLC-ß1 through Ga $_q$, whereas somatostatin acting through sst $_3$ receptors, opioid peptides through µ, d, and ? receptors, adenosine through A $_1$receptors, and ATP and uridine triphosphate through P $_{2Y2}$ receptors preferentially activate PLC-ß3 through the ß? subunits of G $_{i1}$, G $_{i2}$, or G $_{i3}$. Acetylcholine acting through muscarinic M $_3$ receptors activates PLC-ß1 through Gaq, and, acting through M $_2$ receptors, it activates PLC-ß3 through the ß? subunits of G $_{i3}$. [53]

Mobilization of Calcium in Cells of the Longitudinal Muscle Layer

The pattern of inositol phospholipid metabolism in longitudinal muscle differs markedly from that in circular muscle. Only a small amount of IP $_3$ is generated in longitudinal muscle, less than 10% of that generated in circular muscle. The preferred substrate is PIP, which yields the inactive IP $_2$ and diacylglycerol. [41], [53], [54], [55], [56] and [57] IP $_3$ does not bind to the endoplasmic and sarcoplasmic membranes of longitudinal muscle cells or cause Ca $^{2+}$ release at maximal concentrations (1 µM); partial Ca $^{2+}$ release is elicited at supramaximal concentrations (10–50 µM).

Contraction and the increase in cytosolic Ca $^{2+}$ induced by agonists in muscle cells from the longitudinal muscle layer are IP $_3$-independent and are abolished in Ca $^{2+}$-free medium or in the presence of Ca $^{2+}$ channel blockers, implying that an initial step involving Ca $^{2+}$ influx is a prerequisite for Ca $^{2+}$ mobilization in these cells. [44], [47], [48] and [49] The initial influx of Ca $^{2+}$ acts as a trigger for Ca $^{2+}$ release from intracellular stores (i.e., Ca $^{2+}$-induced Ca $^{2+}$ release) (see Fig. 6-8). These stores are highly sensitive to Ca $^{2+}$ alone; increments of cytosolic Ca $^{2+}$ in the physiological range (100–500 nM) trigger Ca $^{2+}$ release. In contrast, Ca $^{2+}$ stores in circular muscle develop high sensitivity to Ca $^{2+}$ only in the presence of IP $_3$. Ryanodine, a specific ligand of ryanodine receptor–Ca $^{2+}$ channels in skeletal and cardiac muscle, binds with high affinity to the sarcoplasmic membranes of longitudinal muscle cells and induces Ca $^{2+}$ release (EC $_{50}$ 2 nM) and contraction (EC $_{50}$ 1 nM). [56] The characteristics of ryanodine binding and Ca $^{2+}$ release in longitudinal muscle cells are similar to those in cardiac muscle cells. [44]

The messenger responsible for initiating Ca $^{2+}$ mobilization in longitudinal muscle cells is arachidonic acid, a product of the hydrolysis of phosphatidylcholine by PLA $_2$ (see Fig. 6-8). An initial agonist-induced, G-protein–mediated activation of PLA $_2$ occurs in longitudinal, but not circular, muscle cells. [57] Suppression of PLA $_2$ activity by selective inhibitors abolishes contraction and the increase in cytosolic Ca $^{2+}$ induced by agonists in longitudinal muscle cells. Arachidonic acid in nanomolar concentrations mimics the effect of contractile agonists and causes an increase in cytosolic Ca $^{2+}$ that is abolished by Ca $^{2+}$ channel blockers. The increase in Ca $^{2+}$ reflects arachidonic acid–dependent Ca $^{2+}$ influx followed by Ca $^{2+}$-induced Ca $^{2+}$ release from intracellular stores. Depletion of Ca $^{2+}$ stores attenuates the increase in cytosolic Ca $^{2+}$ by eliminating the component resulting from Ca $^{2+}$-induced Ca $^{2+}$ release; the residual increase in Ca $^{2+}$ reflects the initial step, that is, arachidonic acid–dependent Ca $^{2+}$ influx. Ca $^{2+}$ influx results from activation of Cl $^-$ channels by arachidonic acid, which leads to depolarization of the plasma membrane and opening of voltage-sensitive Ca $^{2+}$ channels (see Fig. 6-8). [58]

The ryanodine receptor–Ca $^{2+}$ channels in longitudinal muscle cells are highly sensitive to cyclic ADP-ribose (cADPR), a product of the hydrolysis of ß-nicotinamide adenine dinucleotide (ß-NAD+) by ADP–ribosyl cyclase, a membrane-bound enzyme present in longitudinal, but not circular, muscle cells (see Fig. 6-8). [59] cADPR formation is stimulated in a concentration-dependent fashion by contractile agonists in longitudinal, but not circular, muscle cells. It binds with high affinity to microsomes (IC $_{50}$ 2 nM), stimulates Ca $^{2+}$ release (EC $_{50}$ 4 nM) by itself, and potentiates Ca $^{2+}$-induced Ca $^{2+}$ release.

In summary, Ca $^{2+}$ mobilization in longitudinal muscle is mediated by an IP $_3$-independent mechanism, initiated by G $_i$-dependent activation of PLA $_2$ and generation of arachidonic acid; the latter activates Cl $^-$ channels and thus causes depolarization of the plasma membrane and opening of voltage-sensitive Ca $^{2+}$ channels. Ca $^{2+}$ influx through these channels induces Ca $^{2+}$ release by activating sarcoplasmic ryanodine receptor–Ca $^{2+}$ channels. Concomitant activation of membrane-bound ADP–ribosyl cyclase generates cADPR, which potentiates Ca $^{2+}$-induced Ca $^{2+}$ release.

Regulation of Cytosolic Calcium at Rest and During Contraction

Resting and agonist-stimulated cytosolic Ca $^{2+}$ concentrations in cells from the longitudinal and circular muscle layers are remarkably similar in various species (e.g., human, guinea pig, rabbit). [13], [53], [54], [55], [56], [57], [58] and [59] Resting levels (70–100 nM) increase two- to threefold during half-maximal contraction and six- to eightfold during maximal contraction. Exposure of permeabilized muscle cells to these concentrations of Ca $^{2+}$ elicits degrees of contraction similar to those elicited by agonists in intact muscle cells.

Smooth muscle cells, like other cells, possess efficient mechanisms to dispose of the Ca $^{2+}$ transients that occur during contraction. In the resting state, the cells maintain low concentrations of Ca $^{2+}$ in the cytosol despite large chemical (e.g., 2 mM Ca $^{2+}$ outside versus 100 nM Ca $^{2+}$ inside the cell) and electrical (e.g., membrane potential of -40 to -80 mV) gradients favoring the movement of Ca $^{2+}$ into the cell. The gradient for Ca $^{2+}$ is maintained because of low permeability of the plasma membrane to Ca $^{2+}$, the presence of efficient Ca $^{2+}$ extrusion mechanisms in the plasma membrane, and a Ca $^{2+}$ uptake mechanism in the endoplasmic reticulum. The Ca $^{2+}$ extrusion mechanisms in the plasma membrane include a calmodulin-dependent Ca $^{2+}$, Mg $^{2+}$-ATPase, which acts as a high-affinity Ca $^{2+}$ pump sustained by ATP hydrolysis that responds to low Ca $^{2+}$ concentrations, similar to those that occur during contraction, and a low-affinity, high-capacity Na $^+$-Ca $^{2+}$ exchanger sustained by the Na $^+$ gradient across the plasma membrane that responds to more drastic changes in cytosolic Ca $^{2+}$ concentrations.

The Ca $^{2+}$ uptake mechanism into the endoplasmic Ca $^{2+}$ stores is also a high-affinity Ca $^{2+}$-ATPase pump, and it participates in dissipating the Ca $^{2+}$ transients

during contraction. However, most of the Ca^{2+} required to replenish endoplasmic Ca^{2+} stores enters the cell from the outside through voltage-gated Ca^{2+} channels in smooth muscle.

Mechanisms of Relaxation by PKA and PKG

Inhibition of initial contraction (i.e., relaxation) results from inhibition of the initial Ca^{2+} transient. Most agents cause relaxation by stimulating the production of cAMP (e.g., ß–adrenergic agonists), cGMP (e.g., nitric oxide [NO]), or both cAMP and cGMP (e.g., vasoactive intestinal polypeptide [VIP] and pituitary adenylate cyclase–activating peptide [PACAP]), leading to the activation of PKA, PKG, or both. [60], [61], [62], [63], [64], [65], [66], [67], [68] and [69] The levels of cAMP and cGMP in gastrointestinal smooth muscle are regulated by the combined activities of cyclases (i.e., membrane-bound adenylate cyclases types V and VI and soluble guanylate cyclase) and phosphodiesterases (PDEs). [70], [71], [72] and [73] Cyclic AMP, which is produced in amounts 10 to 15 times greater than cGMP, is rapidly degraded by cAMP-specific PDE3 and PDE4. [70], [71] Both enzymes are activated by PKA, but only PDE3 is inhibited by cGMP. cGMP is rapidly degraded by cGMP-specific PDE5, which is activated by PKG, and when present, also by PKA. [71], [72] Each protein kinase selectively inhibits the activity of the corresponding synthetic enzyme: PKA inhibits adenylate cyclase, and PKG inhibits soluble guanylate cyclase. [70], [72] Thus, regulatory feedback from the protein kinases inhibits synthesis and accelerates degradation, thereby maintaining levels of cyclic nucleotides within narrow ranges. The interplay of protein kinases, cyclases, and PDEs in the regulation of cyclic nucleotide levels in smooth muscle is depicted in Fig. 6-10.

FIGURE 6-10. Interplay of adenylate and guanylate cyclases with cyclic nucleotide–dependent phosphodiesterases (*PDE*) and protein kinases. Concurrent release of nitric oxide (NO), vasoactive intestinal polypeptide (VIP), and pituitary adenylate cyclase–activating peptide (PACAP) is the physiological norm in gastrointestinal smooth muscle, leading to activation of adenylate cyclase type V/VI (*AC V/VI*) and soluble guanylate cyclase (*sGC*). This leads to concurrent generation of cyclic AMP (cAMP) and cyclic GMP (cGMP) and activation of both cAMP-dependent and cGMP-dependent protein kinases (*PKA* and *PKG*). PKA activates cAMP-specific phosphodiesterase 3 and 4 (*PDE3* and *PDE4*) and inhibits adenylate cyclase activity, thus stimulating degradation and inhibiting further synthesis of cAMP. PKG activates cGMP-specific PDE5 and inhibits soluble guanylate cyclase activity, thus stimulating degradation and inhibiting further synthesis of GMP. PDE5 is further activated by PKA, whereas PDE3 is inhibited by cGMP. Thus, concurrent generation of cAMP and cGMP leads to enhancement of cAMP levels (through inhibition of PDE3 by cGMP) and attenuation of cGMP levels (through activation of PDE5 by PKA). Furthermore, the affinity of cAMP for, and its ability to activate, PKG is greatly enhanced in the presence of cGMP. Because cAMP is present in 10 to 15 times greater abundance than cGMP, it becomes the main activator of PKG. (From ref. [70]. See also ref. [72].)

Although cAMP preferentially activates PKA, it can, at higher concentrations (e.g., with high concentrations of relaxant agonists) also cross-activate PKG (see Fig. 6-10). [68] An increase in both cAMP and cGMP, such as that brought about by corelease of NO, VIP, PACAP, and peptide histidine isoleucine (PHI; *P*eptide with N-terminal *H*istidine and C-terminal *I*soleucine) from the same or adjacent nerve terminals, is the physiological norm during nerve-induced relaxation in the gut. Inhibition of PDE3 by cGMP enhances cAMP levels, whereas activation of PDE5 by PKA and PKG attenuates cGMP levels. Autophosphorylation of PKG by cGMP greatly increases its affinity for the more abundant cAMP. Under these conditions, PKG is activated by both cGMP and cAMP. [73]

Although both PKA and PKG decrease cytosolic Ca^{2+}, they do so by different mechanisms. [53], [66] Common and distinctive cellular targets of PKA and PKG in smooth muscle are depicted in Fig. 6-11. Both protein kinases can inhibit Ca^{2+} mobilization by inhibiting IP_3 formation in circular muscle and arachidonic acid formation in longitudinal muscle. Both also inhibit the activity of membrane Ca^{2+} channels and stimulate the activity of membrane K^+ channels, leading to hyperpolarization of the plasma membrane and interruption of capacitative Ca^{2+} influx into the cell. However, only PKG can phosphorylate the sarcoplasmic/endoplasmic Ca^{2+}/ATPase pump and can accelerate Ca^{2+} reuptake into the stores. [66] Both PKA and PKG can phosphorylate the IP_3 receptor in vitro or in permeabilized smooth muscle cells, but only PKG can phosphorylate the IP_3 receptor in vivo and can thus inhibit IP_3-induced Ca^{2+} release, implying that PKA does not gain access to the IP_3 receptor under physiological conditions. [66], [68], [74] In summary, both PKA and PKG can inhibit the formation of Ca^{2+}-mobilizing messengers, but only PKG can directly inhibit IP_3-induced Ca^{2+} release in circular muscle by phosphorylating the IP_3 receptor–Ca^{2+} channel. PKG likely inhibits Ca^{2+}-induced Ca^{2+} release in longitudinal muscle by phosphorylating the ryanodine receptor–Ca^{2+} channel.

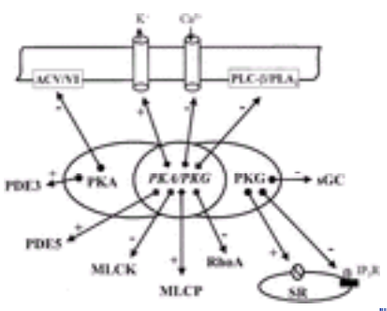

FIGURE 6-11. Distinct and shared cellular targets of cyclic AMP (cAMP)- and cyclic GMP (cGMP)-dependent protein kinases (*PKA* and *PKG*). Only PKA activates cAMP-specific phosphodiesterases 3 and 4 (*PDE3* and *PDE4*) and inhibits adenylate cyclase type V/VI (*ACV/VI*), thus attenuating cAMP levels. Only PKG inhibits soluble guanylate cyclase (*sGC*) activity, stimulates Ca^{2+} uptake into the sarcoplasmic reticulum stores, and inhibits sarcoplasmic Ca^{2+} channels. Other targets are shared by both protein kinases. As shown, either one or both kinases decrease cytosolic Ca^{2+} levels by the following: (*1*) inhibiting the activity of phospholipase C ß (*PLC-ß*) in circular muscle and PLA_2 in longitudinal muscle, thereby diminishing the synthesis of Ca^{2+}-mobilizing messengers; (*2*) inhibiting Ca^{2+} release from sarcoplasmic Ca^{2+} stores (*SR*) by phosphorylating inositol 1,4,5-triphosphate (IP_3) and ryanodine receptors/Ca^{2+} channels; (*3*) stimulating Ca^{2+} uptake into stores by activating the endoplasmic and sarcoplasmic Ca^{2+}-ATPase pump; (*4*) inhibiting the activity of plasmalemmal Ca^{2+} channels; and (*5*) stimulating the activity of plasmalemmal K^+ channels. The resultant hyperpolarization causes further inactivation of plasmalemmal Ca^{2+} channels and suppresses Ca^{2+} influx into the cell. Both PKA and PKG inhibit Ca^{2+}-calmodulin–dependent myosin light chain (MLC) kinase (*MLCK*) activity and stimulate MLC phosphatase (*MLCP*) activity, thereby decreasing MLC phosphorylation during the initial and sustained phases of contraction, and both kinases inhibit RhoA activity, thereby interrupting the pathway that mediates sustained contraction. The net effects of PKA and PKG are to inhibit muscle contraction and to induce relaxation.

In addition to affecting the level of cytosolic Ca^{2+}, PKA and PKG can affect the action of Ca^{2+} by phosphorylating MLC kinase, and thereby decreasing its sensitivity to activation by the Ca^{2+}-calmodulin complex, and by phosphorylating MLC phosphatase, and thus enhancing its activity (see Fig. 6-11). [75], [76] Inhibition of sustained contraction, which as previously noted is Ca^{2+}-independent, can be brought about by phosphorylation of RhoA or phosphorylation of MLC phosphatase through both PKA and PKG. [77], [78]

ELECTRICAL PROPERTIES OF SMOOTH MUSCLE

Resting Membrane Potential

The *resting membrane potential,* defined as the steady-state potential at which the net flow of current (i.e., ions) across the plasma membrane is zero, varies from about -40 to -80 mV in muscle cells of the gut. [79], [80], [81], [82] and [83] Graded differences in resting membrane potential exist between muscle cells in different regions, such as the fundus, corpus, and antrum of the stomach, and between muscle cells located at different depths in the same region, such as cells near the myenteric border and cells near the submucosal border of circular muscle in the antrum or colon.

The membrane potential is largely determined by the activity of the Na^+-K^+ pump (i.e., Na^+, K^+-ATPase), which sets up diffusion gradients for K^+ (162 mM inside versus 5 mM outside) and Na^+ (136 mM outside versus 14 mM inside) across the membrane. The permeability of the membrane to K^+ is much greater than to Na^+, and the flow of K^+ ions down their electrochemical gradient creates a diffusion potential that is the major contributor to the resting membrane potential. K^+ ions flow through passive K^+-selective channels that remain open at rest. [84]

In addition to setting up ionic gradients, the Na^+-K^+ pump is electrogenic, moving three Na^+ ions out of the cell for every two K^+ ions into the cell; the net outward flow of positive charge can contribute up to 30 mV to the resting membrane potential. Variability in the direct contribution of this pump may account for regional differences in resting membrane potential. A Cl^- pump, which maintains low Cl^- concentrations (55 mM) in the cell, can contribute up to 10 mV to the resting membrane potential; its effect is partly offset by the tendency of Cl^- ions to diffuse out of the cell.

Gated Ion-Selective Channels

In addition to passive ion-selective channels, the plasma membrane contains ion-selective channels that can be regulated by membrane potential (i.e., voltage-gated channels) and by various humoral, hormonal, or neural agents (i.e., agonist- or ligand-gated channels). Ligands can activate channels directly and through G proteins in the membrane. Ligands can also activate, inhibit, or modulate voltage-gated channels through second messengers.

Channels in the plasma membrane usually are selective for one ion, such as K^+ or Ca^{2+}, although some allow the passage of more than one ion. The two main types of ion channels involved in the regulation of rhythmic activity of smooth muscle of the gut are selective for K^+ or Ca^{2+}. The properties of Ca^{2+} and K^+ channels were first characterized in amphibian gastric muscle cells [85], [86] and [87] and subsequently in muscle cells from various regions of the mammalian gut. [89], [90], [91] and [92] The flow of ions in single channels can be measured in small patches of plasma membrane. [80], [81], [88], [90] The patches can be electrically isolated by suction into the tip of a micropipette, where they form a tight seal that makes it possible to record current flow in one or only a few channels in the patch. The patches can remain attached to the rest of the plasma membrane or can become fully detached such that the inner (i.e., inside-out patch) or outer (i.e., outside-out patch) surface of the membrane faces the external medium. Each configuration has its advantages. Inside-out patches are useful for examining the role of intracellular messengers; outside-out patches are useful for examining the influence of extracellular ions; and patches in the whole cell configuration are useful for examining the effect of ligands and second messengers.

The use of patches has made it possible to characterize ion channels in terms of their ion selectivity, membrane density, activation and inactivation kinetics, voltage or ligand dependence, and dependence on changes in intracellular Ca^{2+}. The channels are electrically defined by their conductance (i.e., reciprocal of resistance), which is expressed in picosiemens (pS) as the amount of current flowing through the channel in response to an electrical gradient (i.e., current/voltage).

Voltage-Gated Calcium Ion Channels

Voltage-gated Ca^{2+} channels have been identified in muscle cells from the stomach and intestine of several mammals. [92], [93] and [94] The channels carry the inward Ca^{2+} current responsible for the upstroke of the fast action potential. The channels are activated rapidly by depolarization of the plasma membrane to about -40 mV but are inactivated more slowly. Inactivation occurs as a result of Ca^{2+} influx and membrane depolarization. The voltage range of activation (-40 to -10 mV) overlaps with that of inactivation (-60 to 0 mV); at - 40 to -50 mV, some channels remain open and can carry a steady inward Ca^{2+} current. [93], [94] The potential at which such a current may flow is close to resting membrane potential in some muscle cells and usually is attained during the plateau phase of a slow wave.

Voltage-Gated Potassium Ion Channels

Several types of K^+ channels have been identified in gastric and intestinal smooth muscle. [89], [90], [91], [92], [93], [94], [95], [96], [97], [98], [99], [100] and [101] The channels differ in their conductance, ranges of voltage activation, and Ca^{2+} sensitivities. The most widely distributed is a high-conductance (100 pS), Ca^{2+}-activated, voltage-sensitive K^+ channel. [89], [91], [95], [96] and [97] Current through this channel flows outward and can be blocked by barium (Ba^{2+}) and tetraethylammonium. During resting conditions, when cytosolic Ca^{2+} concentrations are low ($<10^{-7}$ M), relatively few channels are open. On stimulation, the increase in cytosolic Ca^{2+} induces activation of large numbers of K^+ channels, which carry an outward current that drives the membrane potential to its resting state (i.e., the K^+ equilibrium potential). A stimulus that acts by inducing membrane depolarization and Ca^{2+} influx is terminated. A second voltage-sensitive K^+ channel with lower conductance (50 pS) has been identified; it opens up on prolonged depolarization, as occurs during the plateau phase of slow waves. [90] Other K^+ channels detected in gastrointestinal smooth muscle include delayed-rectifier, apamin-sensitive, and ATP-sensitive K^+ channels. An apamin-sensitive K^+ channel expressed in some human and guinea pig smooth muscles (chiefly teniae coli) mediates the relaxant effect of PACAP in these tissues. [98]

RHYTHMIC ELECTRICAL ACTIVITY OF SMOOTH MUSCLE

Control of Rhythmic Electrical Activity by Calcium and Potassium Ion Channels

Ca^{2+} channels and Ca^{2+}-activated K^+ channels constitute the electrical apparatus that sustains rhythmicity in smooth muscle. The Ca^{2+} sensitivity of K^+ channels links their activity to that of Ca^{2+} channels and creates the dynamic framework for rhythmic electrical activity. Activation of Ca^{2+} channels induces an inward flow of Ca^{2+} ions that depolarizes the membrane and increases cytosolic Ca^{2+}. Depolarization and an increase in cytosolic Ca^{2+} inactivate the Ca^{2+} channels and activate the K^+ channels, by inducing an outward flow of K^+ ions. Suppression of the inward flow of Ca^{2+} ions and enhancement of the outward flow of K^+ ions restore the resting membrane potential. The speed, amplitude, and duration of these cycles of depolarization and repolarization depend on the relative proportions of Ca^{2+} and K^+ channels, modulation by neural and humoral agents, participation of other voltage-gated channels, and coupling of muscle cells to each other and to pacemaker cells.

Fast Action Potentials

Only *fast action potentials* (i.e., spike potentials) occur in isolated muscle cells, spontaneously or after application of small depolarizing currents; slow waves are not seen. [80], [85], [90], [93] Cell dispersion uncouples muscle cells from pacemaker regions responsible for slow wave activity and from other muscle cells in the syncytium. Small inward currents can cause rapid and complete depolarization of isolated muscle cells, with resulting maximal activation of Ca^{2+} channels and rapid influx of Ca^{2+}. The depolarization and substantial increase in cytosolic Ca^{2+} inactivate the Ca^{2+} channels and induce massive activation of the K^+ channels. The rapid depolarization and repolarization are characteristic of fast action potentials.

Fast action potentials lasting 0.1 to 0.2 seconds can occur in intact muscle. [80], [81], [82] and [83] They occur spontaneously in regions of the gut where the resting membrane potential is more positive than a threshold of -30 mV or can be raised above that threshold by neural stimulation. In other regions, spike potentials occur only after the membrane has been depolarized by slow waves. Whether they occur spontaneously or are superimposed on slow waves, spike potentials are accompanied by muscle contraction. However, they are not essential for contraction, which can also be generated by changes in the amplitude and duration of the plateau potential of slow waves.

Slow Waves

Profile A typical *slow wave* consists of the following sequence: rapid depolarization (i.e., upstroke), partial repolarization, a sustained plateau lasting several seconds, and complete repolarization to the resting membrane potential (Fig. 6-12). [82], [83] In some regions, usually pacemaker regions, the upstroke is preceded by a slow, low-amplitude depolarization (i.e., pacemaker potential) that may trigger the subsequent slow wave.

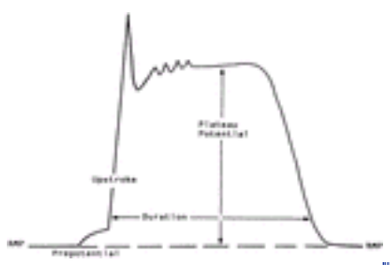

FIGURE 6-12. Profile of a typical slow wave. A slight depolarization (i.e., prepotential) of the resting membrane potential (*RMP*) precedes and may trigger slow waves in some regions. A rapid upstroke is followed by partial repolarization, a plateau potential of variable duration on which may be superimposed small oscillations or spike potentials, and complete repolarization.

The amplitudes, durations, and frequencies of slow waves vary with the location of the muscle. [81] Frequency decreases aborally in human and canine stomach (i.e., 3–5 cycles/min in the corpus to 1.5 cycles/min in the antrum) and intestine (i.e., 12 cycles/min in the duodenum, 7 cycles/min in the ileum, and 5–6 cycles/min in the proximal colon). The frequency gradient is continuous and intrinsic to each region; segments of intestine obtained sequentially along the main axis oscillate at progressively decreasing frequencies. In the canine stomach, the decreasing gradient in frequency is accompanied by an increasing gradient both in the resting membrane potential (i.e., -51 mV in the corpus to -71 mV in the antrum) and in the duration of the plateau potential (i.e., 5 seconds in the corpus to 20 seconds in the antrum). Gradients in resting membrane potential and in the amplitude of plateau potential also occur in the transverse direction, within the thickness of circular muscle. [82] The decrease in the amplitude of the plateau potential in the transverse direction reflects the decay of slow waves with increasing distance from the pacemaker regions at the borders of circular muscle.

Site of Origin Slow waves originate in pacemaker regions located at the myenteric and submucosal borders of circular muscle. [81], [82] and [83] These regions contain a network of cells known as the interstitial cells of Cajal, which act as pacemakers cells capable of initiating rhythmic electrical activity. [102], [103], [104], [105], [106], [107], [108], [109], [110] and [111] The interstitial cells of Cajal are a distinctive population of muscle-like, stellate cells with large nuclei and an abundance of surface caveolae, mitochondria, and rough endoplasmic reticulum. The cells make contact with each other and with muscle cells and nerve terminals. [110] Single interstitial cells isolated from the canine colon appear to be spontaneously active, generating wavelike depolarizations analogous to slow waves. [108], [111] The activity of interstitial cells may not be uniform in all regions, as is evident from the decreasing frequency of slow waves aborally. In some regions (e.g., canine colon), slow waves initiated at the myenteric border differ in configuration and frequency from those initiated at the submucosal border. [102], [105]

Propagation Slow waves originating in the myenteric or submucosal pacemaker regions propagate rapidly around circular muscle and throughout its thickness in the transverse and long axis of the gut. Circumferential propagation is rapid and is facilitated by the peripheral networks of interstitial cells; propagation within the syncytium is facilitated by intramuscular interstitial cells and by the abundance of gap junctions in circular muscle. Slow waves originating in the myenteric pacemaker region of circular muscle also spread to the longitudinal muscle. [104], [105], [112], [113], [114], [115] and [116] After they spread circumferentially, slow waves propagate in oral and aboral directions as discrete rings of excitation capable of eliciting segmental contractions. Propagation in only one direction (i.e., oral or aboral) can occur if the frequency gradient is steep and if the conduction velocity is fast enough to allow slow waves originating in one segment to set the pace of slow waves in another segment [83] or if inhibitory neural input to one segment limits propagation to that segment. [114] Inhibitory neural input to muscle (or pacemaker cells) hyperpolarizes the resting membrane potential and prevents the occurrence of a slow wave, or it decreases the plateau potential and prevents the development of a contraction. Inhibitory neural input reflecting the corelease of NO, VIP, and related neuropeptides appears to predominate normally, masking rhythmic electrical and contractile activity. [114]

Ionic Mechanisms The ionic mechanisms underlying slow wave activity may differ slightly from one region to another. [80], [81], [82] and [83] The rapid upstroke of the slow wave could reflect the inward flow of current through channels of mixed cationic selectivity that are activated at high resting membrane potentials (-60 to -70 mV). Ca^{2+} currents appear to be involved, because the upstroke potential is reduced in the absence of Ca^{2+}. The plateau potential corresponds to the level at which Ca^{2+} channels are activated, and its amplitude reflects the combined activity of voltage-gated Ca^{2+} and K^{+} channels. Ca^{2+}-activated K^{+} channels appear to be the main type of K^{+} channel involved in repolarization of the membrane.

Role in Phasic Contraction The amplitude and duration of the plateau potential determine the magnitude of Ca^{2+} influx through voltage-gated Ca^{2+} channels and can be modulated by excitatory and inhibitory neurotransmitters (Fig. 6-13). Excitatory neurotransmitters, such as acetylcholine or tachykinins, increase the amplitude and duration of the plateau potential and cause a concentration-dependent increase in cytosolic Ca^{2+} and contraction. [104], [117], [118] The increase in Ca^{2+} is accentuated by Ca^{2+} release from Ca^{2+} stores. Spike potentials can be superimposed on plateau potentials and can further augment cytosolic Ca^{2+} concentration and contraction. In some regions, such as the distal antrum and inner lamella of circular muscle in the small intestine, spike potentials appear to be necessary for contraction. [81], [83], [113], [119] Inhibitory neurotransmitters (e.g., NO, VIP, PACAP) decrease the amplitude and duration of the plateau potential or reduce the frequency of spike potentials and prevent the development of contraction associated with a slow wave. These neurotransmitters act predominantly by activating PKA and PKG, which inhibit Ca^{2+} channel activity and stimulate K^{+} channel activity (see Fig. 6-11).

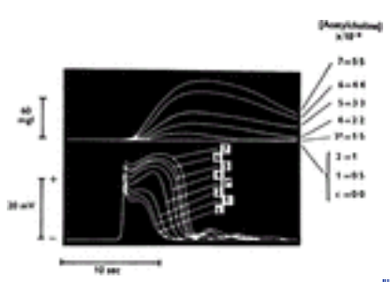

FIGURE 6-13. Relation between the amplitude and duration of the plateau potential (**lower panel**) and the corresponding contraction (**upper panel**) of longitudinal muscle of canine antrum in response to acetylcholine. (From ref. [104].)

Regional Patterns of Contractile Activity

The magnitude of the resting membrane potential, the form, frequency, and site of origin of slow waves, the occurrence and frequency of spike potentials, and the extent of excitatory or inhibitory neural input can be correlated with the neuromuscular function of various regions in the gut. The pattern in humans parallels the pattern observed in dogs.

Stomach The proximal-to-distal gradient in the resting membrane potential of circular smooth muscle in the stomach is depicted in Figure 6-14. The membrane potential in the orad segment or fundus is low, about -50 mV, and lies near or above the threshold for the opening of Ca^{2+} channels and contraction. [81], [120] The segment normally is tonically contracted and is devoid of rhythmic electrical activity. Small changes in stimulatory (i.e., depolarizing) or inhibitory (i.e., hyperpolarizing) neural input can cause further contraction or can induce relaxation of the fundus, features that render this tonic segment suitable for receiving (i.e., receptive relaxation) and discharging (i.e., tonic contraction) a meal into the middle segment or corpus of the stomach. [120]

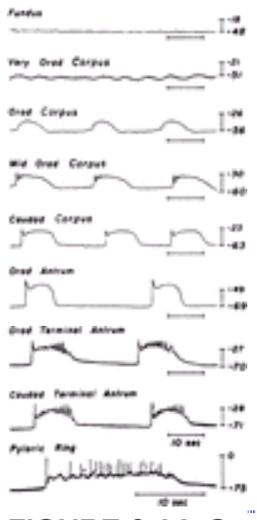

FIGURE 6-14. Gradients in resting membrane potential and profile of slow waves in various regions of the canine stomach recorded with intracellular electrodes. From the proximal to the distal part of the stomach, the resting membrane potential becomes increasingly negative, and the duration of slow waves becomes increasingly longer; potential oscillations and spike potentials are evident in the distal stomach and pylorus. (From ref. [81].)

The corpus is the site of spontaneous pacemaker activity in the stomach and has a higher resting membrane potential. The corpus can undergo both tonic and phasic contraction. Tonic contraction induced by excitatory neurotransmitters is mediated by the release of intracellular Ca^{2+}; the concomitant decrease in the resting membrane potential is not sufficient to activate Ca^{2+} channels and to cause Ca^{2+} influx (Fig. 6-15). [111] Phasic contraction of the corpus is determined by the amplitude and duration of the plateau potential. Small changes in amplitude or duration induced by excitatory neurotransmitters result in substantial changes in the magnitude of phasic contraction.

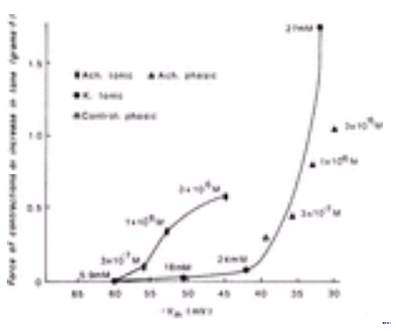

FIGURE 6-15. Comparative effects of acetylcholine (*Ach*) and extracellular K^{+} on membrane potential and contraction in circular muscle from the corpus of the canine stomach. Acetylcholine caused an increase in tonic contraction; the depolarization induced by acetylcholine does not exceed the threshold for opening of Ca^{2+} channels, implying that the source of Ca^{2+} for agonist-induced tonic contraction is intracellular. During slow wave activity, the magnitude of phasic contraction in response to acetylcholine correlates with the amplitude of the plateau potential. Contraction induced by extracellular K^{+} results from depolarization and an influx of Ca^{2+} and coincides with the pattern of phasic contraction induced by acetylcholine. (From ref. [81].)

Intrinsic pacemaker rhythm is highest in the orad corpus (5 cycles/min) and decreases progressively throughout the rest of the corpus, antrum, and pylorus. Slow waves originating in the corpus propagate to and pace antral muscle. Slow waves originating in the distal antrum and pyloric sphincter have prolonged plateau potentials on which spike potentials are usually superimposed. As a whole, the antrum has little tone and is best suited for propagation of slow waves and contractions originating in the corpus. [81], [118], [119] Antral slow waves propagate aborally to the pyloric sphincter, where they pace longitudinal muscle and the outer layer of circular muscle. The slow waves decay before reaching muscle cells in the inner layer of circular muscle; muscle cells in this region are electrically quiescent and may be responsible for the intrinsic tone of the sphincter. Opening of the sphincter is mediated by an inhibitory (i.e., relaxant) neural reflex triggered by distention of the distal antrum; distention occurs when the gastric contents are propelled aborally by phasic or peristaltic contractions originating in the pacemaker region in the corpus of the stomach.

Small Intestine In most mammals, including humans, there is a decreasing gradient in slow wave frequency from the duodenum to the ileum. Slow waves originate in a pacemaker region located at the myenteric border of circular muscle from where they propagate to the bulk of circular muscle and to longitudinal muscle. [111], [112] Removal of a thin layer at the myenteric border of circular muscle abolishes slow waves in longitudinal muscle and in the bulk of circular muscle. Propagation around and in the long axis of the intestine occurs preferentially through circular muscle. The configuration of slow waves in the small intestine is similar to that in the corpus of the stomach and consists of a rapid upstroke followed by a sustained plateau potential. Excitatory and inhibitory neural input influences the amplitude of the plateau potential and the frequency of spike potentials, and determines the occurrence and magnitude of phasic contraction in the intestine. [111], [112]

Colon There is a decreasing gradient in resting membrane potential across circular muscle of the canine colon from -80 mV in muscle cells at the submucosal border to -45 mV in muscle cells at the myenteric border (Fig. 6-16). Rhythmic electrical activity is mediated by two pacemaker regions, one at the myenteric border and the other at the submucosal border. Each pacemaker region generates slow waves with distinctive forms and frequencies that spread passively and summate in the bulk of circular muscle, to yield waves of mixed form. [82], [104], [105], [121]

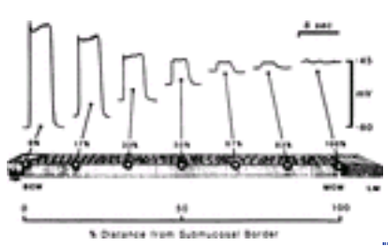

FIGURE 6-16. Records of slow waves obtained at various depths from the submucosal border of circular muscle in canine colon. Two gradients are evident: a decline in the amplitude of the plateau potential from the submucosal (*SCM*) to myenteric (*MCM*) border is matched and offset by a decrease in the resting membrane potential. The plateau potential during slow wave activity attains or exceeds the threshold potential throughout the thickness of circular muscle. Slow waves of a different configuration originate at the myenteric border; as they propagate in circular muscle, they summate with slow waves originating in the submucosal border. (From ref. [106].)

Slow waves originating at the submucosal border have a frequency of five to six cycles per minute, a configuration similar to that found in the small intestine and corpus of the stomach, and a plateau potential lasting 3 to 15 seconds (see Fig. 6-16). [82] The amplitude of the plateau potential decreases as the waves spread toward the myenteric border; the decrease is matched and offset by the decrease in resting membrane potential such that the plateau potential is maintained at about -45 mV, which is close to the threshold for mechanical activity. Slow waves originating at the myenteric border of circular muscle have a frequency of 17 cycles per minute and a sinusoidal configuration. [82], [104], [105], [121] The waves, called *myenteric potential oscillations,* spread to and pace muscle cells in the longitudinal muscle layer. They also spread in circular muscle toward the submucosal border, their amplitude decreasing as a function of distance from the myenteric border. The waves summate with waves originating at the submucosal border; the encounter boosts the plateau potential of waves that originate at the submucosal border and elicits contractions at the rate of six per minute. In longitudinal muscle, myenteric potential oscillations generate fast action potentials. The frequency of these fast transients and of longitudinal muscle contractions is regulated by neural input from the enteric nervous system. Fast action potentials are not generated in circular muscle and only rarely propagate from longitudinal to adjacent circular muscle cells. Excitatory and inhibitory input from neurons of the enteric nervous system is directed at both pacemaker regions, where the density of innervation is highest. [113] Input from these neurons affects slow wave and phasic contractile activity in the same way it does in the small intestine.

Stimulus-Contraction Coupling in Syncytia: Tonic and Phasic Contraction

The signal transduction pathways described in isolated circular and longitudinal muscle cells regulate tonic contraction and relaxation of intact, syncytial muscle.

Small depolarizations caused by hormones or excitatory neurotransmitters are not sufficient to cause tonic contraction, except in some regions of the gut, such as the fundus of the stomach; there the resting membrane potential is close to the mechanical threshold, which is the membrane potential at which Ca^{2+} channels are presumed to open. In circular muscle of the intestine and corpus of the stomach, tonic contraction induced by excitatory neurotransmitters occurs at membrane potentials more negative than the mechanical threshold, implying that the source of Ca^{2+} responsible for tonic contraction in these regions is intracellular (see Fig. 6-15). [111], [112] However, when circular or longitudinal muscle is depolarized by a slow wave, relatively small changes in the plateau potential imposed by the effect of an excitatory neurotransmitter are sufficient to induce Ca^{2+} influx and contraction. The contraction is phasic, coinciding with and determined by the amplitude of the plateau potential; mobilization of intracellular Ca^{2+} by the excitatory neurotransmitter amplifies the phasic contraction.

In some regions, spike potentials superimposed on the plateau potentials produce the requisite depolarization for contraction. Spike potentials are not seen in circular muscle of the colon, [82] the small intestine except for a thin inner lamella, [111] and the corpus of the stomach. [81] In the proximal colon, slow waves emanating from the pacemaker region at the myenteric border of circular muscle elicit spike potentials when they spread to the longitudinal muscle only. Spike generation appears to be more prevalent in longitudinal muscle and may be related to signal transduction pathways and channel characteristics in this muscle.

As previously noted, hormones and inhibitory (i.e., relaxant) neurotransmitters act through PKA or PKG to decrease cytosolic Ca^{2+} and the sensitivity of contractile proteins to Ca^{2+} (see Fig. 6-11). They do so by inhibiting Ca^{2+} release and enhancing sequestration of Ca^{2+} in intracellular stores, [66], [68] a mechanism well suited to the relaxation of tonic contraction, or by hyperpolarizing the plasma membrane and reducing Ca^{2+} influx, a mechanism well suited to the relaxation of phasic contraction (see Fig. 6-11). The hyperpolarization is mediated by inhibitory phosphorylation of voltage-gated Ca^{2+} channels and stimulatory phosphorylation of voltage-gated, Ca^{2+}-dependent K^{+} channels. The hyperpolarization determines the profile or occurrence of slow waves and thus determines the occurrence of phasic contractions. It has traditionally been labeled the *inhibitory junction potential* because it leads to inhibition of phasic electrical and contractile activity.

NEURAL REGULATION OF SMOOTH MUSCLE BY THE MYENTERIC PLEXUS

The intrinsic electrical and mechanical properties of smooth muscle are modulated by neurotransmitters released from neurons of the enteric nervous system, especially neurons of the myenteric plexus. These neurons constitute the final neural pathway regulating smooth muscle activity. Neurons of the submucosal plexus innervate the innermost layers of circular muscle in large species, such as humans and dogs.

Two populations of sensory neurons have been identified. The first, activated by mucosal stimuli, is wholly intrinsic, and the second, activated by muscle stretch and painful stimuli, has neuronal cell bodies in the dorsal root ganglia. Extrinsic neurons of the sympathetic and parasympathetic systems influence smooth muscle indirectly by acting on neurons of the myenteric plexus. Adrenergic and peptidergic neurons in prevertebral and paravertebral ganglia synapse with and inhibit the activity of cholinergic and noncholinergic neurons of the myenteric and submucosal plexuses.

Neuronal Topography

The neural organization of the enteric nervous system is well conserved in mammals and has been extensively studied in guinea pigs, rats, dogs, and humans (see Chapter 2). [122], [123], [124], [125] and [126] Neurons in the myenteric plexus synapse with neurons in the myenteric and submucosal plexuses and paravertebral ganglia and innervate cells in the circular and longitudinal muscle layers. The fibers that make up the deep plexus close to the submucosal border of circular muscle are derived from neurons of the myenteric and submucosal plexuses in large species.

Advances in immunocytochemical and imaging techniques have made it possible to map these neurons and to correlate their morphologic, electrophysiological, and neurochemical properties, specifically their content of neurotransmitters. Neurons of the myenteric plexus fall into two broad categories: about 25% contain VIP or PACAP together with NO synthase (NOS), the enzyme responsible for synthesis of NO in nerve terminals; and about 60% contain acetylcholine, usually together with the tachykinins, substance P, and neurokinin A (NKA). There is virtually no overlap between these categories of neurons. [122], [123], [124], [125], [126], [127] and [128] VIP neurons also contain a homologous peptide, designated PHM (i.e., *P*eptide with N-terminal *H*istidine and C-terminal *M*ethionine) in humans and PHI in animals, which is derived from the same precursor, pro-VIP. Substance P neurons contain NKA, also known as substance K, which is derived from the same precursor, ß-protachykinin. The two main categories of neurons correspond to major roles for acetylcholine and the tachykinins as excitatory neurotransmitters and for VIP, its homologs, and NO as inhibitory neurotransmitters. Excitatory neurotransmitters stimulate Ca^{2+} release, depolarize the plasma membrane (i.e., trigger *excitatory junction potentials*) and induce Ca^{2+} influx, increase muscle tone, and induce phasic contraction. Inhibitory neurotransmitters inhibit Ca^{2+} release, hyperpolarize the plasma membrane (i.e., trigger inhibitory junction potentials) and inhibit Ca^{2+} influx, decrease (i.e., relax) muscle tone, and inhibit electrical slow wave and phasic contractile activity.

Subpopulations of neurons in the two main categories contain one or more of the following: bombesin, also known as gastrin-releasing peptide; neuropeptide Y; the opioid peptides, dynorphin, methionine-enkephalin and their derivatives; and galanin. A few neurons contain ?-aminobutyric acid (GABA), serotonin (<3%), or somatostatin (<5%). Neurons that contain serotonin or somatostatin act mainly as interneurons and influence smooth muscle cells indirectly by means of other neurons.

Peptide and Other Neurotransmitters

Pharmacological Profile Peptide and nonpeptide neurotransmitters are released from axonal varicosities near muscle cells. They diffuse across distances ranging from 20 to 100 nm to interact with receptors on muscle cells or on the same (autoreceptors) or adjacent nerve terminals. The presence of receptors on muscle cells and the possibility of direct action can be determined in isolated muscle cells by measuring the binding of specific radioligands, the release of intracellular messengers (e.g., increase in cytosolic Ca^{2+}, IP_3, or cyclic nucleotides), or the mechanical response of the cell (i.e., contraction or relaxation). [129], [130] Determining the presence of receptors on neurons or their terminals and the possibility of indirect, neurally mediated action of neurotransmitters on muscle cells requires the use of innervated muscle strips; the neurally mediated component of a mechanical response is identified by its sensitivity to blockade by neurotoxins. [129], [130] and [131]
Direct and neurally mediated response. When given exogenously, peptide and nonpeptide neurotransmitters can cause contraction or relaxation of smooth muscle cells directly or indirectly by stimulating or inhibiting the release of other excitatory or inhibitory neurotransmitters. Bombesin and CCK, for example, cause contraction in isolated muscle cells and innervated muscle strips; in the latter, they also elicit release of acetylcholine and substance P. [132], [133], [134] and [135] Their effect in strips is in part neurally mediated and can be partially blocked by the axonal blocker, tetrodotoxin, or by a combination of muscarinic and tachykinin antagonists.
Linkage between VIP/PACAP and NO. VIP, PACAP, PHI, and PHM cause relaxation of isolated muscle cells and muscle strips. [60], [61], [62], [63] and [64], [68], [136], [137] The effects of PHI and its human counterpart, PHM, are mediated by cAMP, whereas the effects of VIP and PACAP are mediated by both cAMP and cGMP. [54], [55], [59] The increase in cGMP is the result of stimulation of NO production in smooth muscle cells by VIP and PACAP (see later). [63], [67], [68] A unique interplay exists between VIP or PACAP and NO in smooth muscle of the gut. NO produced in nerve terminals regulates VIP and PACAP release from the same or adjacent nerve terminals (Fig. 6-17). The release of VIP and PACAP from these terminals regenerates NO in target muscle cells (Fig. 6-18; see Fig. 6-17). [64], [67], [68], [138], [139], [140], [141], [142], [143], [144] and [145]

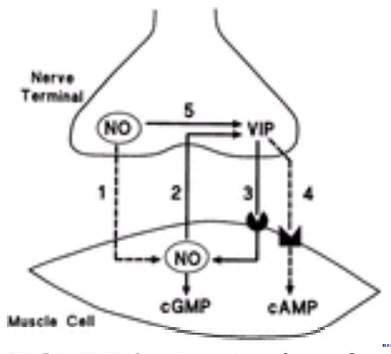

FIGURE 6-17. Interplay of vasoactive intestinal polypeptide (*VIP*) and nitric oxide (*NO*) in the regulation of smooth muscle relaxation. VIP and neuronal NO synthase are often located in the same enteric neurons. NO produced in nerve terminals regulates VIP release (*5*) and diffuses to muscle cells (*1*). VIP released from nerve terminals interacts with one class of natruretic peptide receptors (*NPR-C*) coupled to an endothelial-type NO synthase in muscle cells, leading to the production of

NO and cyclic GMP (*cGMP*) (*3*). NO produced in muscle cells diffuses to nerve terminals, where it facilitates VIP release (*2*). VIP interacts with $VPAC_2$ receptors (previously known as VIP_2/or $PACAP_3$ receptors) coupled to adenylate cyclase to generate cyclic AMP (*cAMP*) (*4*). The amount of NO produced in muscle cells (60% to 80%) is much larger than that produced in nerve terminals (20% to 40%). The linkage between NO production and VIP release and the dual sources of NO explain why NO synthase inhibitors are potent inhibitors of NO production, VIP release, and relaxation and why the VIP antagonist $VIP_{10–28}$ is a potent inhibitor of NO production and relaxation. Oxyhemoglobin neutralizes extracellular NO and blocks only pathways 1 and 2. Identical pathways link the release of pituitary adenylate cyclase-activating peptide (*PACAP*) and NO; PACAP interacts with the same two classes of receptors as VIP.

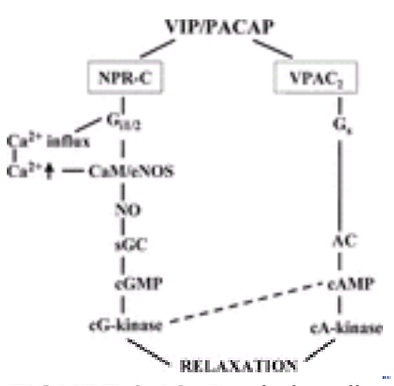

FIGURE 6-18. Dual signaling cascades initiated by the interaction of vasoactive intestinal peptide (*VIP*) and pituitary adenylate cyclase–activating peptide (*PACAP*) with two classes of receptors. VIP and PACAP interact with $VPAC_2$ receptors (previously known as VIP_2 receptors or $PACAP_3$ receptors) coupled by G protein G_s to activation of adenylate cyclase (*AC*), formation of cyclic AMP (*cAMP*), and activation of cAMP-dependent protein kinase (*PKA)*; high agonist concentrations generate higher levels of cAMP that can also cross-activate cyclic GMP (*cGMP*)-dependent protein kinase (*PKG; dotted line*). VIP and PACAP interact with one class of natriuretic peptide receptors (*NPR-C*) coupled by G proteins G_{i1} and G_{i2} to stimulation of Ca^{2+} influx into the cell and activation of a constitutive Ca^{2+}-calmodulin (*CaM*)–dependent isoform of nitric oxide synthase (endothelial type NOS [*eNOS*]). The resultant increase in nitric oxide (*NO*) activates soluble guanylate cyclase (*sGC*), stimulates cGMP formation, and leads to activation of PKG. Both kinases act concurrently to cause relaxation and hyperpolarization of smooth muscle cells. CG-kinase, PKG; cA-kinase, PKA. (Adapted from refs. [67] and [147].)

There is much evidence to support the dual interplay between VIP or PACAP and NO in various regions of the gut in mammalian species (human, dog, opossum, rabbit, guinea pig, rat, and mouse). In isolated myenteric ganglia devoid of muscle cells, the nicotinic agonist, dimethylphenylpiperazinium, stimulates NO production and VIP release; NOS inhibitors abolish both NO production and VIP release, implying a dependence of VIP release on NO production. [138], [139] Exogenous NO causes VIP release from these ganglia, but exogenous VIP has no effect on NO production. [138] Studies in synaptosomal membranes have confirmed that NO donors stimulate VIP release, whereas VIP does not stimulate NO production. [146] In isolated muscle cells devoid of neural elements, VIP and PACAP, but not the homologous peptides, PHI, PHM, or secretin, stimulate NO production. [63], [67], [139], [140] The processes that occur in neurons and muscle cells are reflected in VIP/PACAP release and NO production occurring during nerve stimulation of muscle strips. [63], [67], [139], [141] NOS inhibitors abolish NO production induced by all neural stimuli, and they also abolish VIP release induced by low stimuli and partially inhibit VIP release induced by intense stimuli, indicating that VIP and PACAP release is mediated by NO but can occur independently of it during intense nerve stimulation. [139] The VIP antagonist, $VIP_{10–28}$, which blocks the postjunctional effects of VIP on muscle cells, inhibits NO production by 60% to 80%, indicating that VIP- and PACAP-induced NO production in muscle cells is the major source of NO. [139], [142] The effectiveness of NOS inhibitors in blocking muscle relaxation induced by nerve stimulation led investigators to conclude incorrectly that relaxation was exclusively mediated by NO released from nerve terminals. NOS inhibitors elicit their potent effects by acting at several locations: by suppressing NOS production in nerve terminals, the inhibitors also suppress VIP and PACAP release and thus prevent VIP/PACAP-mediated regeneration of NO in muscle cells. The cellular mechanisms in VIP- and PACAP-induced relaxation involve the interaction of VIP with two distinct receptor types (see Fig. 6-18): $VPAC_2$ receptors (previously known as VIP_2 or $PACAP_3$ receptors), which are coupled through G_s to the activation of adenylate cyclase; and natriuretic peptide receptors type C (NPR-C), which are coupled through G_{i1} and G_{i2} to the activation of NOS in smooth muscle cells. $VPAC_2$ receptors are also recognized by PHI and PHM but with lesser affinity, whereas NPR-C receptors are recognized exclusively by VIP and PACAP, as well as by natriuretic peptides. [67], [143] The distinctive feature of NPR-C is the presence of a truncated 37-amino acid intracellular domain devoid of kinase or guanylate cyclase activity. Site-directed mutagenesis corroborated by studies using peptide fragments derived from the intracellular sequence of NPR-C have identified a 17-amino acid intracellular segment that determines the ability of NPR-C to activate G_{i1} and G_{i2}. [147], [148] and [149] The interaction of VIP and PACAP with NPR-C initiates a signaling cascade involving Ca^{2+} influx that leads to Ca^{2+}-calmodulin–dependent activation of a membrane-bound constitutive NOS, identified by in situ polymerase chain reaction on single muscle cells as endothelial NOS (eNOS) (see Fig. 6-18). [144], [145] The resultant increase in intracellular NO activates soluble guanylate cyclase, and this leads to the generation of cGMP and the activation of PKG. Together, PKA and PKG are responsible for the relaxation of muscle tone. [63], [67], [68] As previously discussed, the two kinases also cause hyperpolarization of the muscle cells, which can prevent the development of phasic contractions in innervated muscle strips. Although the amount of eNOS in muscle cells is small and immunocytochemical detection is difficult, eNOS coupling to G proteins greatly amplifies its effect. In smooth muscle cells of the gut, eNOS is susceptible to inactivation by PKC. [150] A common experimental flaw, whereby the tone of muscle strips is increased using contractile agonists that activate PKC, leads to the suppression of NOS activity in muscle cells and precludes detection of the VIP-NOS linkage in these cells. The mechanisms of action of VIP and PACAP are different in teniae coli, and the receptors for both peptides are distinct from their counterparts in other regions of the gut. Smooth muscle cells in this region are devoid of eNOS. VIP interacts with a specific receptor that does not recognize PACAP. PACAP interacts with a receptor that does not recognize VIP or couple to adenylate cyclase but that couples to the activation of apamin-sensitive K^+ channels. [98], [151]

GABA, somatostatin, and opioid peptides. GABA, acting through $GABA_A$ receptors, causes relaxation in muscle strips but not in isolated muscle cells. The relaxation induced by GABA in muscle strips is accompanied by the release of VIP and NO and can be blocked by VIP antagonists and NOS inhibitors (see Fig. 6-17 and Fig. 6-18). Opioid peptides have an opposite effect. They cause a transient tonic contraction, followed by phasic contractions in previously quiescent muscle strips. The transient tonic contraction is a direct effect of opioid peptides on smooth muscle and is demonstrable in isolated muscle cells. [152] The phasic contractions result from inhibition of the background release of inhibitory neurotransmitters (e.g., VIP, PACAP, and NO) that normally mask phasic contractile activity. [137] Elimination of this background with VIP antibody, VIP antagonists, or NOS inhibitors induces phasic contractions in quiescent muscle strips. Somatostatin stimulates VIP (and probably PACAP) release and NO production indirectly through pathways that inhibit opioid peptide release. The inhibition of opioid peptide release eliminates the restraint exerted by opioid neurons on VIP, PACAP, and GABA neurons, resulting in stimulation of VIP and PACAP release and NO production. The interplay of somatostatin, opioid, and GABA neurons is expressed in the regulation of VIP and PACAP release and NO production during the descending phase of peristalsis (Fig. 6-19).

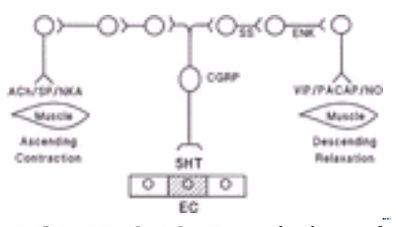

FIGURE 6-19. Regulation of the peristaltic reflex by neurons of the myenteric plexus. The reflex has ascending and descending phases: the descending relaxation and the ascending contraction in circular muscle shown here are accompanied by reciprocal descending contraction and ascending relaxation of longitudinal muscle (*not shown*). The reflex can be triggered by mucosal stimuli and circular muscle stretch. Mucosal stimuli (but not muscle stretch) release serotonin (5-hydroxytryptamine; *5-HT*) from intestinal enterochromaffin (*EC*) cells that acts on 5-HT_4 receptors located on the nerve terminals of intrinsic sensory calcitonin gene–related peptide (*CGRP*)–containing neurons with cell bodies in the wall of the intestine. CGRP acts on modulatory interneurons coupled to (*a*) inhibitory motor neurons that release the relaxant neurotransmitters vasoactive intestinal polypeptide (*VIP*), pituitary adenylate cyclase–activating peptide (*PACAP*), and nitric oxide (*NO*), and (*b*) excitatory motor neurons that release the contractile neurotransmitters acetylcholine (*ACh*), substance P (*SP*), and neurokinin A (*NKA*). The interneurons consist of somatostatin (*ss*) and opioid neurons (enkephalin; *ENK*) connected in series. The increase in somatostatin inhibits the release of opioid peptides, thereby suppressing the inhibitory restraint of opioid neurotransmitters on release of VIP, PACAP, and NO. The reflex triggered by muscle stretch does not involve release of 5-HT; muscle stretch activates extrinsic CGRP-containing sensory neurons with cell bodies in the dorsal root ganglion. These sensory neurons

couple to the same modulatory interneurons and motor neurons. The pathways mediating the reflex are identical in rats and humans.

Physiological Profile The pharmacological profile of actions described in the previous section provides a framework for understanding the regulatory role of neurotransmitters released from neurons of the myenteric plexus. Excitatory motor neurons release one or both types of contractile neurotransmitters: acetylcholine and the tachykinins, substance P and NKA. [153] Inhibitory motor neurons release the relaxant neurotransmitters, VIP with PHI or PHM, PACAP, and NO. Other neurotransmitters modulate the release of excitatory and inhibitory neurotransmitters from motor neurons, as previously noted. Peptides expressed in neurons of the myenteric plexus may be identified as neurotransmitters if they meet the following criteria: [130]

1. Demonstration of peptide synthesis in neurons and presence of the peptide in nerve terminals close to target cells, such as muscle cells, or interstitial cells of Cajal
2. Release of the peptide by physiological stimuli, such as chemically or mechanically induced reflexes
3. Coupling of peptide release to a putative function, as evident from blockade of a physiological response by a specific antiserum or antagonist
4. Mimicry of the response by exogenous application of the peptide, provided the substance does not evoke unrelated pharmacological effects that mask the physiological response.

Several excitatory and inhibitory peptide motor neurotransmitters, such as VIP, PACAP, substance P, and NKA, fulfill these criteria. The example of VIP demonstrates its role as a relaxant motor neurotransmitter (see Fig. 6-18 and Fig. 6-19). VIP and neuronal NOS (nNOS) are colocalized in neurons of the myenteric plexus that innervate mainly circular muscle throughout the gut. [122], [123], [124], [125], [126], [127] and [128] VIP and NO are released concurrently in response to neural stimulation, and their release is accompanied by a proportional increase in relaxation. [136], [137], [139] VIP and its homologs cause relaxation in smooth muscle from all regions of the gut, including the stomach, intestine, gallbladder, and sphincters. [63], [67], [139], [140], [154], [155] The effect of VIP is partially the result of its ability to stimulate NO production in muscle cells (see Fig. 6-18). NO is released from nerve terminals by activation of nNOS and from smooth muscle cells by activation of a VIP/PACAP-dependent eNOS. [139] Neutralization of VIP with specific VIP antiserum, blockade of its effect with selective VIP antagonists, inhibition of VIP release from nerve terminals, and inhibition of its relaxant effect in muscle cells by NOS inhibitors inhibit neurally induced relaxation in all regions of the gut. [64], [139], [142], [154], [155] and [156] Reflex activation of myenteric neurons by physiological stimuli (e.g., stretch or mucosal stimulation) causes VIP and PACAP release, NO production, and muscle relaxation. VIP or PACAP antiserum and VIP/PACAP antagonists inhibit relaxation and NO production by suppressing the effect of VIP and PACAP on muscle relaxation and NO production. NOS inhibitors block NO production in nerve terminals and muscle cells and inhibit NO-dependent VIP and PACAP release, effectively blocking nerve-induced relaxation (see Fig. 6-18 and Fig. 6-19). [139], [141]

Peristaltic Reflex

The peristaltic reflex exemplifies the role of enteric neurons in regulating a physiological motor function (see Fig. 6-19). [142], [156], [157], [158], [159], [160], [161], [162], [163], [164], [165], [166], [167], [168], [169] and [170] The reflex can be evoked by stroking, which stimulates sensory nerve terminals in the mucosa, or by radial stretch, which stimulates sensory nerve terminals in circular muscle. The reflex consists of an orad (or ascending) and a caudad (or descending) component or phase. During the caudad phase, circular muscle relaxes while longitudinal muscle contracts; during the orad phase, circular muscle contracts while longitudinal muscle relaxes. Reciprocal contraction and relaxation of the two muscle layers maintain the dimensions of the segment (see Fig. 6-19). [163] In vitro stimulation of hollow or flat segments of intestine at the orad end to elicit only the caudad (i.e., descending) phase, or at the caudad end to elicit the orad (i.e., ascending) phase, makes it possible to identify the types of neurotransmitter released and their functional coupling to each component of the peristaltic reflex. Compartmented flat-sheet colonic preparations are particularly useful in examining the reflex in humans, which is identical to that observed in rat and guinea pig. Sensory neurotransmitters are released into the central compartment, where stimuli are applied, but not into the peripheral compartments, where orad and caudad mechanical responses are measured. [164], [165] and [166]

Excitatory and Inhibitory Motor Neurons VIP, PACAP, and NO are released during and are responsible for the descending relaxation of circular muscle; VIP or PACAP antiserum, VIP or PACAP antagonists, and NOS inhibitors inhibit the descending relaxation (see Fig. 6-19). [142], [156], [157] Acetylcholine, substance P, and NKA are released during and are responsible for the ascending contraction of circular muscle; contraction is partially inhibited by muscarinic antagonists and is abolished by a combination of muscarinic antagonists and tachykinin antagonists or antibodies (see Fig. 6-19). [157], [158] The pattern illustrates the involvement of the main excitatory and inhibitory motor neurotransmitters of the myenteric plexus in regulating the motor limb of the peristaltic reflex.
Modulatory Interneurons Interneurons of the myenteric plexus participate in the peristaltic reflex by modulating the release of excitatory and inhibitory neurotransmitters from motor neurons. [159], [160], [161] and [162] The release of somatostatin increases during the descending phase, whereas the release of opioid peptides decreases. [160], [161] In the resting state, opioid interneurons exert a continuous restraint on VIP/PACAP/NOS motor neurons. This restraint is eliminated during the descending phase when opioid peptide release is decreased, leading to an increase in VIP, PACAP, and NO release. [159], [160], [161] and [162] Opioid inhibitors applied during the descending phase enhance the release of VIP, PACAP, and NO and increase muscle relaxation; the application of opioid peptides has the reverse effect. The decrease in opioid peptide release, which eliminates opioid restraint on VIP/PACAP/NOS motor neurons, is mediated by the increase in somatostatin during the descending phase. Neutralization of somatostatin with somatostatin antibody increases opioid peptide release and decreases VIP, PACAP, and NO release and muscle relaxation. Exogenous somatostatin has the opposite effect, enhancing VIP, PACAP, and NO release, as well as circular muscle relaxation. [159], [161] The interplay of somatostatin and opioid interneurons that regulates the activity of VIP/PACAP/NOS motor neurons also regulates the activity of VIP/PACAP/NOS interneurons. These interneurons synapse with cholinergic/tachykinin motor neurons that innervate longitudinal muscle. [163] The activation of these interneurons results in contraction of longitudinal muscle during the descending phase concurrently with relaxation of circular muscle. A switch in the activity of somatostatin interneurons during the ascending phase presumably leads to a reverse pattern of circular muscle contraction and longitudinal muscle relaxation, which is characteristic of the ascending phase of the peristaltic reflex. P> **Sensory Neurons Mediating Peristalsis** Two distinct populations of sensory neurons that can be differentially activated and desensitized mediate the peristaltic reflex, which is elicited by muscle stretch and mucosal stimulation. [164], [165] and [166] Muscle stretch activates the intramuscular nerve terminals of extrinsic sensory neurons, which have cell bodies in the dorsal root ganglion and axonal projections to myenteric neurons. Mucosal stimulation activates intrinsic (i.e., enteric) sensory neurons with nerve endings in the mucosa. Activation is initiated by the release of 5-HT from mucosal enterochromaffin cells that acts on 5-HT $_4$ receptors located on sensory nerve terminals and causes the release of the sensory neurotransmitter, calcitonin gene–related peptide (CGRP). [165], [166], [167], [168], [169] and [170] The addition of lumenal 5-HT or selective 5-HT $_4$ agonists induces CGRP release and triggers the peristaltic reflex. Selective 5-HT $_4$ antagonists block CGRP release and the mechanical components of the reflex induced by mucosal stimulation. CGRP antagonists block the ascending and descending phases of the reflex. The release of 5-HT and CGRP and the effects of 5-HT $_4$ agonists, 5-HT $_4$ antagonists, and CGRP antagonists are elicited only in the compartment where stimuli are applied, clearly identifying 5-HT and CGRP as components of the sensory limb of the reflex. Exposure of the intestinal lumen to 5-HT $_4$ agonists initiates peristaltic activity and stimulates propulsion of intralumenal contents. [168], [169] and [170] Addition of opioid d-receptor antagonists suppresses the inhibitory restraint exerted by opioid interneurons on excitatory and inhibitory motor neurons and greatly enhances the propulsive effect of 5-HT $_4$ agonists. [169] The potent synergism between opioid antagonists and 5-HT $_4$ agonists at near-threshold concentrations endows the combination with therapeutic potential. Events corresponding to the two phases of the peristaltic reflex underlie propulsion and the opening and closure of various sphincters, including the lower esophageal, pyloric, choledochal, ileocecal, and internal anal sphincters. [154], [155], [171] In these regions, VIP (and likely PACAP) acting directly and by means of NO appears to be the main neurotransmitter responsible for sphincter relaxation; acetylcholine and probably tachykinins participate in sphincter contraction.

HORMONAL REGULATION OF SMOOTH MUSCLE FUNCTION

Hormonal influences on smooth muscle activity are evident during and in between meals. The example of CCK illustrates the interplay of hormonal and neural influences. After ingestion of a meal, CCK is released into the circulation from the upper small intestine; it causes both direct and cholinergically mediated contraction of muscle cells in the gallbladder and neurally mediated relaxation of muscle cells in the sphincter of Oddi. Relaxation of the sphincter is accompanied by the release of VIP and is blocked by VIP antiserum, VIP antagonists, and NOS inhibitors, implying that CCK causes relaxation by stimulating the release of VIP and NO from intramural neurons. [155]

Motilin illustrates the participation of a hormone in the regulation of smooth muscle activity between meals. In humans and other mammals, cycles of electrical and contractile activity, described in greater detail in Chapter 11, recur at 1.5- to 2-hour intervals between meals. The cycles consist of four distinct phases, collectively known as the *interdigestive myoelectric complex* or *migrating motor complex.* The cycles culminate in phase III, a 5- to 10-minute period of intense phasic contractile activity. Cycles typically begin in the stomach and migrate aborally throughout the small intestine; some cycles appear to begin in the small intestine. Motilin, a peptide released from endocrine cells of the upper small intestine, appears to be responsible for initiating cycles that begin in the stomach. Peaks of motilin coincide with the onset of phase III in the stomach. Neutralization of circulating motilin with motilin antiserum disrupts phase III activity, and infusion of motilin in concentrations that mimic circulating levels induces premature phase III activity. [172], [173] Cycles that begin in the intestine are not controlled by motilin and may be regulated by input

from enteric and extrinsic neurons. The neural and hormonal mechanisms that cause suppression of the cycles on ingestion of a meal are unknown.

HUMORAL REGULATION OF SMOOTH MUSCLE FUNCTION

In addition to neurotransmitters and circulating hormones, humoral agents produced by nonneural cells within the smooth muscle layer influence smooth muscle activity. These include ATP, histamine, serotonin, adenosine, and eicosanoids, such as prostaglandins, thromboxanes, and leukotrienes. Receptors for most of these agents have been identified on smooth muscle cells of the gut.

ATP, whether released as a neurotransmitter or as a metabolic product, interacts with two P_2 receptors on muscle cells. [174] The term P_2 is preferable to purinergic because it recognizes that purine and pyrimidine nucleotides can act as preferential ligands of various receptor subtypes. At low concentrations, ATP interacts with P_{2Y2} receptors coupled by G_q to PLC-ß1 and by G_{i3} to PLC-ß3 and elicits IP_3-dependent Ca^{2+} release and muscle contraction. At higher concentrations, ATP also interacts with ligand-gated P_{2X1} receptors that act as cationic channels and cause depolarization of the plasma membrane and Ca^{2+} influx through voltage-gated Ca^{2+} channels. At high concentrations, ATP also activates K^+ channels; the notion that activation of these channels on release of ATP from inhibitory nerves is responsible for initial hyperpolarization has some proponents.

ATP is rapidly degraded to adenosine, which also interacts with two types of receptors, A_1 and A_2, on smooth muscle cells of the intestine. [175] These receptors are coupled to three signaling pathways. A_2 receptors are coupled by G_sa to the activation of adenylate cyclase, whereas A_1 receptors are coupled by G_{i3}a to the inhibition of adenylate cyclase and by G_{i3}ß? to the activation of PLC-ß3 and IP_3-dependent Ca^{2+} release in circular muscle and to the activation of PLA_2 and Ca^{2+}-induced Ca^{2+} release in longitudinal muscle. [175] The net effect of adenosine is contraction, which is augmented after blockade of A_2 receptors; blockade of A_1 receptors unmasks relaxation.

Two types of receptors for serotonin, 5-HT_2 and 5-HT_4, coexist on smooth muscle cells of the stomach and intestine. The 5-HT_2 receptors mediate contraction through the increase of cytosolic Ca^{2+}, and the 5-HT_4 receptors mediate relaxation through an increase in cAMP. [176] Similarly, two types of receptors for histamine, H_1 and H_2, coexist on smooth muscle cells of the stomach and intestine. H_1 receptors mediate contraction through an increase in Ca^{2+}, and H_2 receptors mediate relaxation through an increase in cAMP. [177] For both 5-HT_2 and histamine H_1 receptors, Ca^{2+} mobilization in circular muscle is mediated by means of PLC-ß1 and IP_3-dependent Ca^{2+} release, while in longitudinal muscle it is mediated by means of PLA_2 and Ca^{2+}-induced Ca^{2+} release. The net effect of 5-HT and histamine is contraction, reflecting the dominant influence of 5-HT_2 and H_1 receptors, respectively. The blockade of 5-HT_4 or H_2 receptors augments the contraction, whereas blockade of 5-HT_2 or H_1 receptors unmasks relaxation.

The influence of histamine, serotonin, adenosine, and various eicosanoids probably is more pronounced when smooth muscle is inflamed. Humoral agents are released from cells that lie in proximity to muscle cells and to myenteric neurons and their terminals. These agents can act directly on muscle cells, or they can act indirectly by stimulating or inhibiting the release of neurotransmitters and thereby influencing physiological responses.

SUMMARY

Smooth muscle of the gut exhibits variable tone on which are superimposed rhythmic contractions, driven by cycles of depolarization and repolarization (slow waves) that originate in pacemaker cells (interstitial cells of Cajal) located mainly at the periphery of circular muscle. Ca^{2+} influx occurs when the depolarization attains a level at which voltage-gated Ca^{2+} channels are activated, triggering a transient contraction superimposed on muscle tone. Release of excitatory neurotransmitters (mainly acetylcholine and the tachykinins, substance P, and NKA) from enteric motor neurons accentuates depolarization and Ca^{2+} influx and initiates Ca^{2+} release, causing an increase in both tone and rhythmic contraction. Release of inhibitory/relaxant neurotransmitters (mainly VIP, PACAP, and NO), enhanced by VIP/PACAP-dependent generation of NO from smooth muscle cells, induces hyperpolarization of the plasma membrane and suppresses rhythmic contractile activity (hence the term *inhibitory*) and attenuates muscle tone (i.e., induces relaxation). In some regions, interstitial cells within the muscle mass may participate in relaying neural input to smooth muscle.

The effects of excitatory and inhibitory neurotransmitters on tone (and, to a lesser extent, on rhythmic contractions) are mediated by G-protein–coupled receptors. Muscle contraction occurs in two phases. The initial phase consists of a transient contraction mediated by Ca^{2+}-calmodulin–dependent activation of MLC kinase and phosphorylation of MLC_{20}. Ca^{2+} mobilization during the initial phase involves G-protein–dependent activation of PLC-ß and stimulation of IP_3-dependent Ca^{2+} release in circular muscle and G-protein–dependent activation of PLA_2 and arachidonic acid–stimulated Ca^{2+} influx, followed by Ca^{2+}- and ADP ribose–induced Ca^{2+} release in longitudinal muscle. The second phase consists of a sustained Ca^{2+}-independent contraction during which MLC_{20} phosphorylation is maintained by PKC- and Rho kinase-dependent inhibition of MLC phosphatase. The pathway that leads to inhibition of MLC phosphatase involves activation of the heterotrimeric protein, G_{13}, the monomeric G protein, RhoA and its associated kinase, phospholipase D, and PKC.

Relaxation is mediated by NO- and VIP/PACAP-dependent stimulation of cAMP and cGMP and activation of both PKA and PKG. cAMP and cGMP levels are maintained within narrow limits by PKA/PKG-dependent feedback mechanisms that activate PDEs and inhibit cyclases. PKA inhibits adenylate cyclase and activates cAMP-specific PDE3 and PDE4 and cGMP-specific PDE5, whereas PKG inhibits soluble guanylate cyclase and activates PDE5. The concurrent stimulation of cGMP enhances cAMP levels by inhibiting PDE3 activity and greatly increases the affinity of cAMP for PKG. Thus, during physiological neural activity when both cyclic nucleotides are present, the more abundant cAMP preferentially activates PKG. Both protein kinases inhibit PLC-ß and PLA_2 activities, but only PKG directly inhibits IP_3-induced Ca^{2+} release. These properties are mainly responsible for the ability of PKA and PKG to inhibit initial contraction. The inhibition of sustained contraction is mediated by the ability of both protein kinases to inhibit RhoA activity and to stimulate MLC phosphatase activity.

The interplay of smooth muscle cells, interstitial cells, and enteric neurons (interneurons, sensory neurons, and excitatory and inhibitory motor neurons) is seen to best advantage in the regulation of the peristaltic reflex. The reflex is triggered by mucosal stimuli produced by the passage of digesta through the intestine, and only exceptionally by distention, which uses an extrinsic sensory pathway. Mucosal stimulation releases 5-HT from enterochromaffin cells; 5-HT acts on 5-HT_4 receptors located on the nerve terminals of intrinsic, sensory CGRP-containing neurons; the information is relayed sequentially through somatostatin and opioid interneurons to the ascending and descending excitatory and inhibitory motor neurons. The ascending limb of the reflex involves the release of acetylcholine and tachykinins from excitatory motor neurons and mediates orad contraction of circular muscle; the descending limb, which mediates caudad relaxation of circular muscle, involves the combined release of VIP, PACAP, and NO from inhibitory motor neurons, accentuated by VIP/PACAP-mediated NO production in muscle cells. A distinct set of VIP/PACAP/NOS interneurons that synapse with tonic cholinergic/tachykinin motor neurons innervating longitudinal muscle mediates reciprocal caudad contraction and orad relaxation of longitudinal muscle. The ability of 5-HT_4 agonists to initiate peristalsis, particularly when combined with opioid antagonists, underlies their therapeutic potential.

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CHAPTER 7

Richard S. Blumberg and William F. Stenson

THE IMMUNE SYSTEM AND GASTROINTESTINAL INFLAMMATION

The intestinal epithelial cell surface represents a vast frontier of body surfaces that must be defended by the immune system. The intestinal immune system must defend against the many infectious and toxic assaults that may breach the epithelium and cause intestinal injury. The immune system also must recognize epithelial cell transformation, to which the intestine may be uniquely prone, because toxin exposure and the high proliferation rate of the epithelial cells increase the risk for cytogenetic error and malignant transformation. [1] The intestinal immune system must simultaneously ignore the multitude of commensal organisms and dietary antigens that are not threats to the host.

Because specific immunity is driven by antigen recognition, the components of the immune system that protect the gut are presented with a significant challenge in differentiating foreign or nonself material from self-antigens by responding to the former and ignoring the latter. Except for a few other epithelial surfaces, no other organ system is presented with this combination of problems in such a dramatic fashion. The skin is covered by a horny layer of epithelial cells that effectively excludes most antigens. As a result of constant antigenic exposure, the gut possesses abundant lymphoid cells (i.e., B and T lymphocytes) and myeloid cells (i.e., macrophages, neutrophils, eosinophils, mast cells). The mucosal immune system of the gastrointestinal tract represents one of the largest immunologic compartments in the body. [2]

To deal with this challenge, the gut-associated lymphoid tissue (GALT) has evolved several important modifications of antigen processing, humoral immunity, and cellular immunity to cope with its organ-specific responsibilities. These include flattened epithelial microfold (M) cells that transport antigens, the specialized epithelial cells of the domes overlying lymphoid aggregates that sample lumenal antigens selectively, the immunoglobulin A (IgA) system that helps to exclude and remove foreign antigens, and unique mechanisms of generating local specific secretory immunity in the context of systemic tolerance. [3], [4], [5], [6] and [7] The functional segregation of intestinal from systemic compartments reiterates the separate role of the intestine as a unique lymphoid organ that is linked to other mucosal surfaces, such as those of the lung, breast, and genitourinary tract, to create a common mucosa-associated lymphoid tissue (MALT). [8] Functional linkage of the MALT is accomplished by the regulated traffic of lymphocytes between the affiliated tissues. [9]

Advances in molecular biology have provided important insights into the general operation of the immune system, including the manner in which the immune system processes, presents, and recognizes antigens through the major histocompatibility complex, T-cell receptors, and immunoglobulins; the mechanisms by which the responsive cellular elements are stimulated after antigen contact through accessory molecules, adhesion molecules, and signal transduction; and the manner in which the activated immune cells carry out their functional destiny through the production of cytokines and other humoral factors and through cytolysis. The immune response generated by these events usually is self-limited after the foreign antigen is cleared. Without self-regulation or with persistent uncleared antigen, chronic inflammation results, as in inflammatory bowel disease. [10]

INNATE AND ADAPTIVE IMMUNITY

Two types of immunity normally function along the mucosal surfaces: natural (innate) and acquired (adaptive). Although natural immunity is nonspecific and rapidly mobilized, acquired immunity is specific for antigens on foreign substances and is mobilized slowly over several days or weeks after an initial or primary exposure. In general, specific immunity amplifies the initial protection provided by natural immunity in a focused, antigen-specific manner, in part through the further mobilization of the natural immune components. In the adaptive immune system, lymphocytes interact with antigens through receptors (the T-cell receptor and the B-cell receptor) that are specific for that antigen. When a specific antigen is presented to one of the few lymphocytes with a receptor capable of recognizing it, that lymphocyte undergoes clonal expression. Unlike natural immunity, adaptive immunity is characterized by the acquisition of memory during the course of a specific immune response, which produces a more rapid mobilization of directed, specific immunity in future or secondary antigen encounters.

Natural intestinal immunity has two components: nonimmunologic and immunologic. The mucosal surface of the intestine expresses a variety of nonimmunologic physiochemical barriers to exclude, inactivate, or clear pathogenic substances and organisms. These barriers include gastric acid, digestive enzymes, and other potentially antipathogenic factors, such as bile acids, lysozymes, intestinal mucus, normal peristalsis, indigenous microbial flora, and the epithelial cell itself, whose tight junctions are usually impenetrable when intact. In diseases such as the acquired immune deficiency syndrome (AIDS), defects in one or more of these factors may increase susceptibility to invasive pathogens.

A second class of natural immunity is composed of immunologic factors. [11] These include cellular and soluble elements. Virtually all classes of cells participate in innate immunity, including natural killer (NK) cells, a subclass of T cells called *NK-T cells,* phagocytes, mast cells, and epithelial cells, among others. In response to specific cell surface signals delivered by a variety of receptors, these cells either engulf microorganisms (e.g., phagocytes) or secrete a wide variety of soluble substances. The latter function to remove pathogens (e.g., cryptdins from intestinal epithelial cells), recruit other cell types (e.g., interleukin-8 [IL-8] from intestinal epithelial cells), arm other cell types against microbial invasion (e.g., interferon-? [IFN-?] from NK-T cells), remove cells altered by infection or malignancy (e.g., granzymes and perforins from natural killer cells), or modify specific immune responses (e.g., IL-4 from NK-T cells). Many of these responses are initiated by interactions between specific components of microbes and either soluble or cell surface receptors on host cells (pattern recognition receptors) that recognize characteristic microbial structures, such as the recognition of bacterial lipopolysaccharide by toll receptors (Table 7-1). In contrast to the adaptive immune system, in which there are many thousands of distinct T-cell and B-cell receptors, there are relatively few (several hundred) receptors in the innate immune system. The innate immune system receptors recognize a few highly conserved molecules that exist in large groups of microorganisms and are termed *pathogen-associated molecular patterns*. These microbial molecules, which include lipopolysaccharides, peptidoglycans, and lipoteichoic acids, share certain attributes: (1) None of them are

expressed by the host, (2) they are required for microbe survival, and (3) they are shared by a broad group of pathogens. The receptors of the innate immune system are expressed on antigen-presenting cells, which include monocytes, macrophages, dendritic cells and B cells; some are also expressed on intestinal epithelial cells. The receptors of the innate immune system—pattern recognition receptors—include toll-like receptors, which recognize lipopolysaccharides, and macrophage mannose receptors, which recognizes carbohydrates with large numbers of mannose units, characteristic of microorganisms. Pattern recognition receptors are expressed uniformly on large numbers of cells of a given class. For example, all macrophages express the same mannose receptor, in contrast to the acquired immune system, in which different T cells express thousands of different T-cell receptors. Recognition of bacterial products by pattern recognition receptors results in prompt intracellular signaling events and a biologic response. For example, recognition of lipopolysaccharide by toll-like receptors results in the activation of nuclear factor-?B (NF-?B) and, as a consequence, the induction of a wide variety of inflammatory- and immune-response genes. This prompt biologic response in the innate immune system occurs over a few hours, in contrast to the much slower response in the acquired immune system, which involves clonal expression of relevant lymphocytes over several days. The innate immune system, therefore, is much better situated than the acquired immune system to deal with acute infections caused by pathogens new to the host.

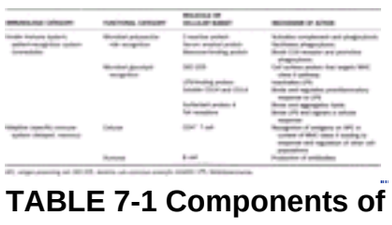

TABLE 7-1 Components of Innate and Adaptive Immune Responses

Specific immunity also has two components: humoral and cellular. Humoral immunity is represented by antibodies derived from B lymphocytes and defends against extracellular events. Cellular immunity is derived from T lymphocytes and provides protection from deleterious intracellular events that are not amenable to the effects of antibodies.

Both types of specific immunity possess several properties that differentiate them from nonspecific immunity. First, to respond to the wide variety of potential antigens to which humans may be exposed during a lifetime, a large number of clonally distinct lymphocytes specific for particular determinants or epitopes on a molecule are maintained. The molecular basis for this clonotypic specificity is determined by a cell surface receptor (i.e., immunoglobulin for B cells and T-cell receptor for T cells) that is unique to each cell or clone. Second, each cell or clone expressing its unique or clonotypic receptor is selected to differentiate a self-antigen from a nonself or foreign antigen. This process, which is primarily the result of positive selection (i.e., selection of good receptors) and negative selection (i.e., deletion of bad receptors) of B cells in the bone marrow and T cells in thymus during lymphocyte development, results in the production of an army of lymphocytes that are tolerant or nonresponsive to self. [12], [13] and [14]

In the absence of tolerance, inappropriate recognition of self may occur, resulting in autoimmunity. The intestine, liver, and peritoneum participate in the extrathymic pathways of T- and B-cell development, and self-reactive lymphocytes that escape from the normal selection processes may reside within these organs. [14], [15] The fact that intestinal autoimmunity is not commonly observed suggests that peripheral mechanisms in the intestine prevent the activation of potentially pathogenic clones (see section “ Autoimmunity and Oral Unresponsiveness”).

The specific immune system generates memory during the course of the immune response that results in the expansion of long-lived cells, representing the progeny of the original responsive antigen-specific B-cell or T-cell clones. After reexposure to the original offending antigen, the adaptive immune system responds more rapidly, is quantifiably larger, and becomes more specific. In the case of B cells, for example, immunoglobulins are generated that bind more avidly to their antigen.

COMPONENTS OF THE IMMUNE SYSTEM

T Lymphocytes

T lymphocytes play a crucial role in immune effector function and regulation by recognizing and eliminating infectious agents and tumors and by orchestrating a coordinated immune defense through their regulatory effects on other cell populations. Lymphoid progenitors destined to be T cells arise from pluripotent stem cells in the bone marrow and migrate to the thymus. Within the thymus, maturation and education about self-antigens occur, leading to the production of self-tolerant T cells that migrate to and populate the secondary lymphoid organs, including the spleen, lymph nodes, and tonsils. The intestine contains secondary lymphoid aggregates in the form of Peyer patches (see section “ Peyer Patches and Lymphoblast Homing”).

Unlike B cells, which recognize determinants on whole soluble antigens, most T cells recognize processed nominal antigens or peptide fragments from the whole antigen in association with components of the major histocompatibility complex (MHC) on the surface of an antigen-presenting cell. [16] The antigen-specific or clonotypic receptor on the cell surface of T cells that is responsible for the recognition of antigen in the context of an MHC molecule is the T-cell receptor. The T-cell receptor consists of immunoglobulin-like, heterodimeric glycoproteins, called *aß* and *?d heterodimers*. [16], [17] Rarely, the T-cell receptor exists on the cell surface as a ßß homodimer. Most mature T lymphocytes in the peripheral blood and intestine express the aß heterodimer, and the others express the ?d receptor. Transfection experiments and, more recently, x-ray crystallography have proved that the aß heterodimer achieves recognition of a peptide in association with an MHC molecule. [18] The mechanism of recognition of the ?d T-cell receptor is not as clearly defined. [19] However, T cells bearing a ?d T-cell receptor have been shown to recognize a variety of target cell structures, including classic and nonclassic MHC class I molecules (e.g., recognition of MHC class I chain-related gene A [*MICA*] by ?d T cells in the human intestinal epithelium serves to remove impaired epithelial cells), extracts of mycobacteria, heat shock proteins, viral glycoproteins, and small nonpeptide phosphorylated molecules.

The protein structures of the T-cell receptor a, ß, ? and d chains, as deduced from their complementary DNAs, consist of a leader peptide, a long amino-terminal extracellular domain, a transmembrane region, and a short cytoplasmic tail. [20] These protein segments are organized into variable (V), joining (J), constant (C), and, for the ß and d chains, diversity (D) regions. Like immunoglobulins, these structural regions are each encoded by distant gene segments on chromosome 14 (e.g., T-cell receptor a and d) and chromosome 7 (e.g., T-cell receptor ß and ?) that recombine during T-cell development in the thymus (Fig. 7-1). In the case of the T-cell receptor ß chain, for example, there are approximately 60 different V gene segments, organized into 24 different V-region families, 2 D-region segments, 13 J-region segments, and 2 C-region segments. The sequential recombination of a D-region gene to a J-region gene and a V-region gene to a D-region gene, with looping out and deletion of the intervening genomic sequences, results in a functional transcriptional unit that is transcribed, spliced appropriately, and translated. These genetic recombination events are orchestrated by protein products of the recombination-activating genes (RAGs). In the absence of functional RAG proteins, T- and B-cell development does not occur.

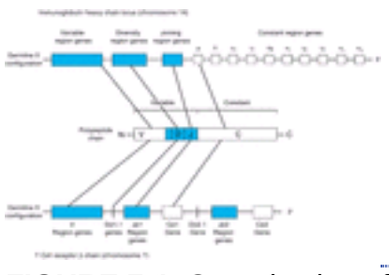

FIGURE 7-1. Organization of the immunoglobulin heavy chain locus on chromosome 14 and the T-cell receptor ß-chain locus on chromosome 7. In both, distant clusters of gene segments encoding variable (*V*), diversity (*D*), joining (*J*), and constant (*C*) regions recombine during T-cell development in the thymus and B-cell development in the bone marrow to create a functional transcriptional unit that is translated into a polypeptide chain. The polypeptide chain contains a variable region

that is involved in antigen binding (i.e., soluble antigens for B cells and peptide fragments in the context of an MHC component for T cells), and a constant region that is involved in anchoring the immunoglobulin or T-cell receptor protein to the plasma membrane as an integral membrane protein. Alternative immunoglobulin transcripts are expressed that encode a secreted form of the molecule in which the constant region domain (i.e., Fc portion) of the protein possesses an effector function (e.g., complement fixation, opsonization, antibody-dependent cell-mediated cytotoxicity, immediate hypersensitivity). Most of the variable region is concentrated at the junction of VDJ regions (*shaded areas*), which are involved in antigen (e.g., immunoglobulin) and peptide/MHC (e.g., T-cell receptor) binding. In the case of the T-cell receptor, more amino-terminal sequences in the variable region are primarily involved in interactions with the MHC protein outside the peptide-binding pocket.

To allow for the multitude of potential antigenic interactions, diversity of the T-cell receptor is accomplished by several mechanisms: the existence of multiple V-, D-, and J-region genes; recombination of the D region in multiple reading frames; addition of nucleotides not included in the germ-line configuration at the V-D and D-J junctions by activity of terminal deoxynucleotidyl transferase (non–germ-line or N-region additions); and different combinations of either the T-cell receptor a and ß or ? and d chains. [21] As a result, most of the variability of the T-cell receptor is concentrated at the V-D-J junction or complementarity-determining region 3 (CDR3) part of the receptor. This hypervariable region of the T-cell receptor, or NDN region, represents the clonotypic determinant of the receptor, with sequences in the more amino-terminal region of the T-cell receptor (CDR1 and CDR2 regions) more involved in MHC interactions. The average length of the CDR3 region of the T-cell receptor a and ß chains is about ten amino acids. The longer length of the T-cell receptor ?- and d-chain CDR3 regions (20+ amino acids) is consistent with the hypothesis that counterligands for these receptors may be recognized in a manner that is more similar to that of immunoglobulins. Simultaneous crystallization of the T-cell receptor aß heterodimer and MHC class I molecule has shown that the CDR3 region of the T-cell receptor is the major contact site for antigenic peptide. [18]

Intimately associated with the aß or ?d T-cell receptor in a noncovalent fashion on the cell surface are at least four proteins of the cluster of differentiation 3 (CD3) complex: ?, d, e, and ?. The CD3 complex likely has a stoichiometry that consists of two e and ? chains and one ? and d chain. Although invariant and not contributing to the antigen specificity of the T-cell receptor, the CD3 proteins are members of the immunoglobulin supergene family. The CD3 proteins are involved in the assembly and transport of the T-cell receptor to the cell surface and in signal transduction after the T-cell receptor binds antigen in the context of MHC. The T-cell receptor and CD3 complex together play the central role in cognate or antigen-specific interactions with an antigen-presenting cell.

Several molecules play important roles in noncognate or antigen-nonspecific interactions between a T cell and an antigen-presenting cell. T cells are segregated into distinct functional subtypes by the expression of combinations of these molecules as defined by the CD antigens. T cells can be broadly divided into two categories: cells that express CD4 and cells that express CD8. Cells that express the CD4 molecule, a 60-kd glycoprotein, help coordinate immune responses and recognize antigenic peptides in the context of class II MHC molecules. Most cells that express CD8, a 32-kd heterodimeric glycoprotein, have the aß heterodimer, and a minority have an aa homodimer. The CD8-positive ($CD8^+$) T cells recognize antigenic peptides in the context of class I MHC molecules, whereupon they elicit cytolysis or secrete cytokines that influence immune response networks. [22] The antigen-independent conjugate formation between CD8 and class I MHC and between CD4 and class II MHC determinants provides a stabilizing environment for the ternary interactions between the T-cell receptor and its target antigenic peptide and MHC complex. Through interactions with other intracellular ligands, such as the nonreceptor lck protein tyrosine kinase, the CD4 and CD8 molecules play an essential role in signal transduction in addition to their noncognate cell surface binding activities. [23]

In addition to CD4–MHC class II and CD8–MHC class I determinants, other important T-cell and antigen-presenting cell interactions play a role in modulating adhesion and signal transduction. T-cell activation requires two events. First, the T cell must adhere to the local connective tissue matrix and antigen-presenting cell, which stabilizes subsequent intercellular interactions and may provide intracellular signals for subsequent events. Second, the T cell must appropriately receive two distinct intracellular signals from cell surface molecules: an antigen-specific signal (signal 1) through the T-cell receptor and CD3 complex (i.e., cognate signal) and an antigen-independent signal (signal 2) through an accessory molecule on the T cell (i.e., noncognate or costimulatory signal) that is delivered by its ligand counterpart on an antigen-presenting cell. [24] Delivery of these two signals provides the appropriate impulse for activation.

In response to the composite of cognate and noncognate cell surface interactions, a signal is transmitted intracellularly, resulting in a cascade of early biochemical and later nuclear events that lead to the expression of new cell surface antigens important to further intercellular interactions, cell growth, and the functional expression of the cell. The signal transduction pathways initiated by T-cell receptor–CD3 complex ligation and binding of the CD4 and CD8 coreceptor are highly complex and include activation of tyrosine kinases (lck, fyn, ZAP70), leading to activation of phospholipase C?1, ras, and phosphatidylinositol 3-kinase, leading in turn to mitogen-activated protein kinase (MAPK) and protein kinase C activation and calcium mobilization. Subsequent activation of cytosolic and translocated nuclear factors, such as nuclear factor of activated T cells (NFAT), initiate new gene transcription, such as IL-2 production. [23]

In the absence of the costimulation provided by a second signal, clonal anergy (i.e., tolerance) to the antigen or apoptosis (i.e., cell death) may occur. [25] The requirement for two signals is probably fundamental to antigen-specific events and presumably plays an important role in extrathymic mechanisms of avoiding autoimmunity. Costimulation is most important to activating naive (virgin) T cells and less so to previously activated T cells, which may depend more on cytokine signals (e.g., from IL-15). [26] Some of the accessory molecular interactions between a T cell and an antigen-presenting cell are summarized in Table 7-2. In particular, the signal delivered to several CD28-related molecules on the T cells by several CD80/CD86-related molecules on the antigen-presenting cell may be critical for T-cell activation. [27], [28] A molecule homologous to CD28, CTLA-4, is expressed on activated T cells and uses CD80 and CD86 as its ligands. CTLA-4 may function to provide a corollary down-regulatory signal to T cells on binding CD86. Blockade of the CD28-CD80/CD86 pathway with recombinant decoy molecules resembling CD28 suppresses cellular activation. [28]

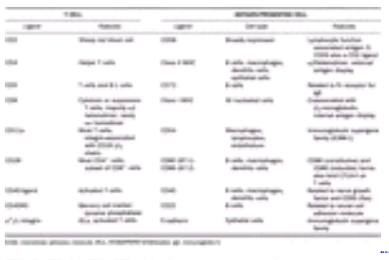

TABLE 7-2 Ligand Interactions Between T Cells and Antigen-Presenting Cells

Major Histocompatibility Complex and Antigen Presentation

The strict dichotomy between CD4- and CD8-bearing T cells in terms of their recognition of antigens in the context of class II and class I MHC, respectively, has important functional implications. An understanding of the molecular basis of antigen processing is essential to appreciate these effects.

The human MHC is also known as the *human leukocyte antigen (HLA) complex*. The MHC or HLA products and the genes that encode them are subdivided into three classes. The MHC class I and class II glycoproteins are involved in antigen presentation to T cells. The three main MHC class I genes are designated *HLA-A, HLA-B,* and *HLA-C;* the three main MHC class II genes are designated *HLA-DP, HLA-DQ,* and *HLA-DR*. The class III MHC genes encode for a variety of functionally distinct proteins (e.g., complement components, heat shock proteins, and tumor necrosis factors).

The genes for the MHC class I and II molecules and several genes that are involved in antigen processing, such as the proteosome-like genes and transporter genes, are encoded in the MHC locus on the short arm of human chromosome 6. Each MHC class I protein is expressed on the cell surface as a dimer composed of two noncovalently linked polypeptide chains: a 43-kd integral membrane glycoprotein called the *heavy* or *a chain,* encoded by the *HLA-A, HLA-B,* and *HLA-C* genes, and an associated non–MHC-encoded, nonglycosylated 12-kd protein called *ß2-microglobulin*.

The a or heavy chain of the MHC class I molecule consists of three extracellular folded domains, a_1, a_2, and a_3, that are each encoded by separate exons and share structural homology with immunoglobulins. The a_1 and a_2 domains of the *HLA-A, HLA-B,* and *HLA-C* genes are highly polymorphic, adjacent, and roughly parallel to one another. Based on the defined crystal structure of an HLA-A2 molecule, the two domains form a groove consisting of a series of ß-pleated sheets

bounded by two a helices that bind a peptide of relatively strict length (nine amino acids). [29], [30] The degree of observed variability within the a_1 and a_2 domains suggests that antigenic peptides reside in the groove, and that exposed surfaces of the a helices interact with the V region of the T-cell receptor. Because each person carries six different HLA-A, HLA-B, and HLA-C alleles and each allelic gene product is capable of binding a variety of different peptides, the peptide-carrying capacity of the endowed class I MHC is theoretically enormous; the estimate is about 50 HLA-A, 97 HLA-B, and 34 HLA-C alleles in the human population. [31]

Certain molecules encoded within the HLA class I region, such as HLA-E, HLA-F, HLA-G, and Hfe, and the MHC class I chain-related gene A (*MICA*), are unique in the absence of extensive polymorphism within their a_1 and a_2 domains. These nonpolymorphic molecules may be involved in immunologic recognition or other unique functions. [32] For example, leader peptides from HLA-G are presented by HLA-E to inhibitory receptors on natural killer cells. [33] Distinct alleles of Hfe have been linked to the pathogenesis of hemochromatosis. [34] These molecules are considered to be nonclassic MHC class I or MHC class Ib molecules. Some MHC class Ib molecules (e.g., CD1 and the neonatal Fc receptor for IgG) are encoded by genes outside the MHC locus on chromosome 6. The CD1 molecules appear to present lipid antigens to T cells. [35]

Class II molecules are nondisulfide-linked heterodimeric glycoproteins consisting of a 32- to 34-kd a chain and a 29- to 32-kd ß chain that are encoded by different genes in the MHC class II genetic locus. Significant allelic variation exists for the *DRß* (126 alleles), *DPß* (60 alleles), and *DQß* (26 alleles) genes, in contrast to the *DRa* (2 alleles), *DPa* (8 alleles), and *DQa* (15 alleles) genes. [31] The extracellular portion of a class II molecule consists of two domains, approximately 90 amino acid residues long, called a_1-a_2 and $ß_1$-$ß_2$, respectively. Similar to the class I MHC, x-ray crystallographic studies of HLA-DR1 show that the a_1 and $ß_1$ domains of an MHC class II molecule form a peptide-binding pocket with somewhat less stringency in peptide length (15–18 amino acids) than MHC class I–associated peptides. [36] The class II MHC is highly polymorphic, with the polymorphic residues concentrated in the membrane-distal a_1 and $ß_1$ domains, which are involved in peptide binding. The three-dimensional structure revealed by x-ray crystallography shows that each MHC class II molecule naturally exists as a dimer similar to immunoglobulin. Multimeric receptor forms are also possible for other molecules involved in cognate recognition (e.g., MHC class I, T-cell receptor) and probably increase the avidity of the reaction with the apposing cell.

Although extensive structural similarities exist between class I and class II MHC molecules, the origin of the derived peptide differs substantially. MHC class I peptides usually are derived from intracellular proteins, and MHC class II peptides usually are derived from the extracellular milieu. Distinct cellular compartments and trafficking pathways determine the source of antigenic peptides for the two classes of MHC molecules. MHC class I molecules primarily bind cytoplasmic proteins (e.g., viral proteins in infected cells) that are degraded into peptides by a large cytoplasmic proteolytic complex derived from at least two genes in the HLA class II region that code for low-molecular-mass polypeptides 2 and 7. These peptide fragments are transported into the endoplasmic reticulum by an endoplasmic reticulum membrane–associated heterodimer that is derived from the gene products of the MHC class II–associated genes, transporter-associated protein 1 (TAP1) and TAP2. In the endoplasmic reticulum, the transported peptides bind to the MHC class I groove or binding pocket. On loading with peptide, $ß_2$-microglobulin associates with the MHC class I heavy chain, and a stable conformation is assumed that allows cell surface transport of the MHC class I peptide complex. [37]

Class II MHC molecules bind peptides derived from exogenous proteins that have been internalized by means of antigen-presenting cells in a coated pit into the endocytic pathway, where antigen degradation generates functional peptides. Intersection of the endocytic pathway with class II MHC molecules derived from the trans–Golgi network results in binding of the exogenous peptide to the peptide-binding groove of the class II proteins and the release of the invariant or ? chain, which stabilizes class II molecules before peptide binding. Before release of the MHC class II–associated invariant chain, the ? chain is proteolytically degraded to a smaller fragment, CLIP, which remains associated with MHC class II until exchange with exogenous peptide occurs. The exchange is facilitated by a nonpolymorphic product of the MHC class II locus, HLA-DM, which is restricted to an intracellular location within a specialized MHC class II compartment related to lysosomes. The resulting peptide-protein complex is then translocated to and expressed on the cell surface. [37] The antigen-processing machinery associated with the CD1-related molecules (CD1a-d) for presentation of glycolipids overlaps with and is highly homologous to the MHC class II pathway and is considered to be a third pathway of antigen presentation. [35]

At any given time, cells display a summary of their internal antigenic exposure through class I molecules and their external antigenic exposure through class II MHC molecules. Under normal circumstances, self-proteins are displayed, leading to a state of tolerant inactivity. Under abnormal circumstances, deleterious intracellular events lead to the display of MHC class I molecules bound with foreign peptides, the activation of $CD8^+$ T cells, and the generation of cytolytic T cells. Similarly, extracellular exposures to foreign substances result in the cell surface display of class II MHC molecules bound with foreign peptides, leading to the activation of $CD4^+$ T cells, the induction of B cells through helper T-cell activity, and immunoglobulin production.

The CD8–MHC class I pathway is propelled by internal cellular events. Because virtually all nucleated cells express MHC class I molecules, essentially all cells can act as antigen-presenting cells for $CD8^+$ T cells, providing important protection against harmful intracellular processes such as viral infection and neoplasia. The CD4–MHC class II pathway is responsive to external cellular events, but the relative restriction of constitutive MHC class II expression and antigen-presenting capacity to B cells, macrophages, dendritic cells, venular endothelial cells, and probably intestinal epithelial cells of the small intestine and colon during inflammation limits the possibilities and consequences of activating $CD4^+$ T cells. Moreover, the restricted expression of accessory molecules essential to T-cell activation, such as CD80 and CD86, also imposes strict limitations on antigen-initiated T-cell activation.

Immunoglobulins

When expressed on the surfaces of B lymphocytes, immunoglobulin molecules serve as clonotypic receptors, comparable to the T-cell receptor on T cells. [38] After binding their cognate antigens, the surface immunoglobulins initiate a cascade of events that may activate the B cell, prompt clonal proliferation, and generate plasma cells. The immunoglobulins that are secreted as a result of this activation function as antibodies, traveling throughout tissue fluids to detect and bind to the antigenic molecules that first triggered their production.

Each B cell expresses a unique immunoglobulin molecule that contains hypervariable regions conferring its antigenic specificity or idiotype. The secreted antibodies consist of two identical light chains of about 24 to 28 kd, each linked by disulfide bridges to two identical heavy chains of about 55 to 70 kd that are linked by disulfide bridges to each other. Each chain consists of repeating homologous globular domains, called *immunoglobulin domains,* in tandem. [17]

There are two immunoglobulin light chain types (? and ?) and five immunoglobulin heavy chain isotypes (?, µ, a, e, and d). There are two isotypes of IgA (IgA1 and IgA2) and four isotypes of IgG (IgG1, IgG2, IgG3, and IgG4), which are determined by the C-region gene segment (see Fig. 7-1). Alleles, or allotypes, of these isotypes also exist. Although the biologic function of the antibody (e.g., complement fixation) is determined by the Fc portion of the isotype, the antigen specificity of the antibody is determined by the amino terminus of the heavy and light chains, which are encoded by the V-, D-, and J-region gene segments—the idiotypic region of immunoglobulins. The polymorphism of the V region is further concentrated into three hypervariable stretches, called *complementarity-determining regions (CDR1–3),* the tertiary structure of which complements that of the bound antigen and plays an important role in antigen binding. Four protein segments intervene between the three CDR regions. These four segments are relatively nonpolymorphic and are called *framework regions (Fr1–4)*.

The diversity of immunoglobulins for interaction with the wide variety of environmental antigens is accomplished by mechanisms similar to those described for the T-cell receptor. Unlike T-cell receptors, immunoglobulins also increase diversity by somatic mutation of the variable gene sequence after recombination of immunoglobulin gene segments has taken place, which accounts for the avidity maturation of immunoglobulins during secondary antigen encounters. Most avidity maturation generated by somatic mechanisms involves the CDR1–3 regions and, less commonly, the Fr1–4 regions.

B Lymphocytes

B lymphocytes are the progeny of hematopoietic stem cells in bone marrow and fetal liver. Pre–B cells express cytoplasmic IgM heavy chain and, in the absence of cell surface IgM, are antigen-independent. Further antigen-independent maturation within the bone marrow leads to the development of antigen-responsive, membrane IgM- and IgD-positive B cells expressing the same V region or idiotype. The membrane IgM on mature B cells is noncovalently associated with a disulfide-linked heterodimer, consisting of Ig-a and Ig-ß transmembrane glycoproteins, that participates in IgM cell surface transport and signal transduction after antigen binding. [39] Similar to the CD3 complex association with the T-cell receptor, Ig-a and Ig-ß probably link the surface immunoglobulin with cytoplasmic protein kinases, which become functionally activated after antigen binding.

Like T cells, B-cell subsets can be identified on the basis of phenotypic markers other than membrane immunoglobulin. CD5, a T-cell marker, is expressed by 5% to

10% of B-lineage cells. These CD5 $^{+}$ B cells, or B-1 cells, are capable of autoantibody production and may mature outside the bone marrow within the peritoneal cavity, based on studies in mouse models. [14] Within the privileged space of the peritoneum, these cells are not likely to encounter their antigen and be activated to produce antibody. Their expansion in autoimmune disease suggests that under certain circumstances they may be activated to express autoantibody. Most B cells do not express CD5; they are known as *B-2* or *common B cells*. Other B-cell molecules of potential significance include CD19, CD20, and CD40. These molecules are expressed by all human B cells and are involved in B-cell activation, proliferation, and, for CD40, isotype switching. [40] CD40 binds to CD40-ligand on an activated T cell and functions as an important costimulatory molecule. [41]

Outside the bone marrow in the periphery, the stimulation of mature B cells by an appropriate soluble antigen results in the clonal expansion of the responding B cell and the generation of a secreted form of the membrane-bound immunoglobulin using the same idiotype and, therefore, the same antigen specificity. Some of the clonally expanded B cells differentiate into memory cells; antigen-unstimulated B cells have a relatively short half-life. After further modulation by antigen-specific T cells, B cells switch the isotype form associated with the antigen-specific V region (usually to isotypes of IgG in the periphery and IgA in the GALT) and differentiate into plasma cells, which secrete extremely high levels of immunoglobulin. These stages of B-cell development depend on specific cytokines released by antigen-specific T cells, the most important of which are likely to be transforming growth factor-ß (TGF-ß), IL-4, IL-5, and IL-6.

Natural Killer Cells

Natural killer cells, which comprise about 10% of the peripheral blood lymphocytes, express neither membrane immunoglobulin nor the T-cell receptor–CD3 complex. [42] They were originally considered to be neither B cells nor T cells and were referred to as *null cells*. They may express some T-cell differentiation antigens, such as CD2 and CD8 (as an aa homodimer), and myelomonocyte differentiation antigens, such as CD11b, CD11c, CD14, and CD15. These large, granular lymphocytes characteristically express CD56, which is homologous to neural cell adhesion molecule, and sometimes express CD16, a low-affinity receptor for the Fc portion of IgG. CD16 is often associated with the CD3-? homodimer found in the T-cell receptor–CD3 complex.

Natural killer cells are a major component of the natural immune system. They elicit cell-mediated cytotoxicity, which plays an important role in resistance to intracellular pathogens and in the elimination of tumor cells. Although natural killer activity is a component of natural immunity, it can be modulated by T-cell–derived cytokines, such as IL-2. In turn, natural killer cells represent an important source of cytokines, such as IFN-?.

Natural killer cells are stimulated to elicit cytotoxic activity by one of two mechanisms: through binding of a target cell opsonized by IgG by means of CD16 on the natural killer cell (i.e., antibody-dependent cell-mediated cytotoxicity) or through the recognition of target cells that lack HLA-A, -B, or -C expression. Because many viruses and tumors, including intestinal cancers, down-regulate HLA-A, -B, or -C expression to evade adaptive immune responses involving classic MHC class I molecules, this mechanism of recognition is extremely important. Natural killer cells express cell surface receptors for specific HLA alleles, which, when engaged by HLA proteins, inhibit killer cell function. These killer-inhibitory receptors (KIRs) include molecules such as NKB1 (p70 KIR), which binds HLA-Bw4, and p58.1 KIR, which binds HLA-Cw4. In the absence of these inhibitory signals by MHC class I molecules, killer-activating receptors (KARs) such as p50 KAR stimulate natural killer cell activity by inducing the release of proteins such as granzyme and perforin, which kill the target cell.

Myelomonocytic Cells

Cells of the myelomonocytic lineage include monocytes and their progeny (macrophages and dendritic cells) and polymorphonuclear leukocytes (neutrophils, eosinophils, and basophils). They evolve from a common myeloid progenitor cell in the bone marrow in response to cytokines, including several interleukins (e.g., IL-1, IL-3, and IL-6) and the macrophage and granulocyte colony-stimulating factors (e.g., G-CSF, GM-CSF, M-CSF).

Monocytes, mononuclear phagocytes that mature into macrophages when residing within tissues, and dendritic cells play central roles in natural immunity through their phagocytic function and inflammatory cytokine secretion, and in specific immunity through their ability to endocytose, process, and present foreign antigens to T cells. [43] Monocytes within the peripheral blood almost uniformly express the CD14 marker (see Table 7-1) and shed this molecule within tissues such as the lamina propria of the intestine, which they enter during an inflammatory response, such as occurs in inflammatory bowel disease. These recruited monocytes evolve into tissue macrophages that have active phagocytic functions. The phagocytic function of macrophages is aided by their expression of receptors for the Fc portion of immunoglobulins and complement components (C3b) that coat foreign antigens. These phagocytic functions are important not only for eradicating invading microorganisms but also as the essential first step in initiating antigen processing and presentation for MHC class II pathways, which is a prerequisite for activating CD4 $^{+}$ T cells that orchestrate further immune responses. Macrophages promote inflammation by secreting IL-12 and IL-18, which drive CD4 $^{+}$ T cells to secrete IFN-? that in turn activates the phagocytic and antigen presentation functions of the macrophage.

Dendritic cells are the most potent antigen-presenting cells. The GALT, both organized (Peyer patches) and nonorganized (lamina propria and epithelium), contains dendritic cells. [44] Within tertiary lymphoid tissues such as the lamina propria, dendritic cells are considered to be immature; they exhibit active phagocytic functions but poor antigen presentation functions. On migration to regional lymph nodes, such as the Peyer patches and mesenteric lymph nodes, dendritic cells mature and up-regulate their antigen presentation functions through the expression of important costimulatory molecules, such as CD80 and CD86. A characteristic feature of dendritic cells is their heterogeneity, which likely correlates with specific immunologic functions. In the mouse, three subsets of dendritic cells, defined by CD11c expression, have been identified that express either CD11b (myeloid), CD8 (lymphoid), or neither marker (double-negative). Although the function of the double-negative subset remains unknown, the myeloid and lymphoid subsets appear to be responsible for inducing differentiation of naive T cells into Th2/Th3 and Th1 subsets, respectively.

Nonimmune Parenchymal Cells

A variety of parenchymal cells, which are not considered immune cells per se, exhibit important immunologic functions, thus creating an immunophysiological network. These cell types do not likely function to initiate specific immune responses through the education of naive lymphocytes but function to integrate and enhance ongoing immune responses and contribute to immunopathology in disease states. [45] These cell types include intestinal epithelial cells (see section “ Epithelial Cells” under “Gastrointestinal Inflammation”), mesenchymal cells (e.g., fibroblasts), smooth muscle cells, and endothelial cells. Fibroblasts, through the expression of cell surface molecules, connective tissue components, and cytokines, regulate local lymphocyte survival and function and contribute to TGF-ß–mediated fibrosis associated with chronic inflammation. Smooth muscle cells can present antigens in an MHC class II–restricted fashion in the context of inflammation and respond to inflammatory cytokines such as IL-4 with alterations in intestinal motility, which may play a role in increasing peristalsis, an attribute important in eradicating lumenal pathogens. Endothelial cells play a key role in regulating tissue inflammation by both directing the recruitment of leukocytes and myeloid cells (see section “ Adhesion Molecules and Cell Trafficking”) and secreting a variety of soluble mediators that enhance inflammation.

Cytokines

Cytokines are an ever-expanding group of protein hormones secreted by a variety of cell types—including immune cells (e.g., lymphocytes, macrophages) and nonimmune cells (e.g., endothelium, epithelium, fibroblasts, smooth muscle)— that have autocrine, paracrine, and endocrine functions (Table 7-3). The effect of each cytokine depends on the cytokine receptor expression of target cells, which depends on the cell’s differentiation state and previous cytokine exposure, reflecting the networks that exist between cytokine action. The pleiotropism of cytokine action creates significant repetition within these networks.

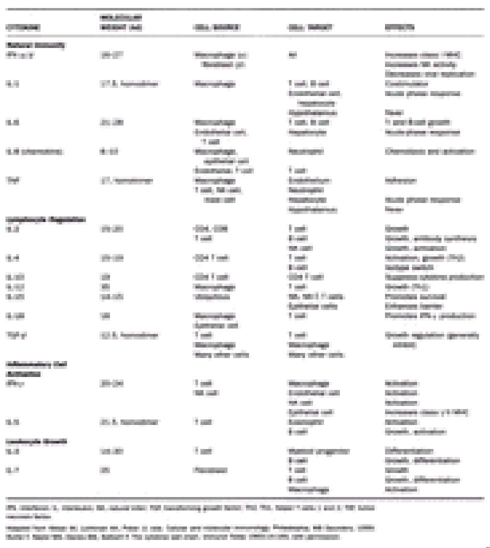

TABLE 7-3 Cytokines

Although a given cell type can secrete a variety of different cytokines and significant redundancy exists in their actions, some patterns allow functional classification of the cytokines and the cells that produce them (Fig. 7-2). Helper T (Th) cells that have been persistently stimulated can be divided into Th1 cells, which predominantly secrete IL-2 and IFN-?, a profile that supports the early events of T-cell and B-cell development (IgG1 production) and cell-mediated immunity (delayed-type hypersensitivity); Th2 cells, which predominantly secrete IL-4, IL-5, IL-6, IL-10, and IL-13, a profile associated with humoral immunity (IgG4 and IgE production); Th3 cells, which predominantly secrete TGF-ß, a profile associated with suppression; and T-regulatory (Tr1) cells, which predominantly secrete IL-10, a profile also associated with suppression. [46] Consistent with these phenotypes, T cells often are considered either effector (Th1, Th2) or regulatory (Th3, Tr1) cells. $CD4^{+}$ cells, which express CD25, the IL-2 receptor a chain ($CD4^{+}CD25^{+}$ cells), may be a particularly important T-regulatory cell capable of secreting IL-10 and TGF-ß, responsible for preventing autoimmune gastritis and inflammatory bowel disease. [47] Th0 cells exhibit a secretory pattern of cytokines that includes those associated with Th1 and Th2 patterns and also represents newly stimulated T cells. [48], [49] Th0 cells are driven to become either Th1 cells by IL-12, which stimulates the transcription factor T-bet, or Th2 cells by IL-4, which stimulates the transcription factor GATA-3. [50] Although there has been considerable interest in defining T-cell clones in this manner, most T-cell clones are not so easily defined. In addition, because many cell types secrete several cytokines, it is probably best to ascribe combinations of cytokines as type 1 and type 2 when identifying patterns of cytokine production so as not to overemphasize the source of the cytokine. These issues are important owing to the increased evidence that certain idiopathic diseases, such as inflammatory bowel disease, and inappropriate immune responses to certain infectious pathogens may be associated with distinct cytokine profiles. CD8-bearing T cells may also preferentially secrete combinations of cytokines that fall into either a type 1 or type 2 pattern. One possible classification of cytokine activity is provided by their association with natural immunity, regulation of lymphocyte function, activation of inflammatory cells, and regulation of leukocyte growth (see Table 7-3).

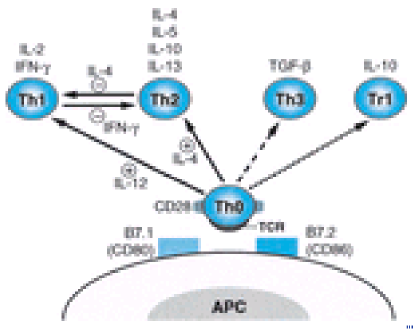

FIGURE 7-2. Helper T-cell differentiation. After the uptake and processing of the antigen, a professional antigen-presenting cell (*APC*), such as a macrophage or dendritic cell, provides two signals to a naive helper T cell (*Th0*); signal 1 is provided by presentation of the nominal peptide of the antigen in the context of an MHC class II molecule to the T-cell receptor on the T cell (*TCR*), and signal 2, the costimulatory signal, is provided by either CD80 or CD86 to CD28 on the T cell. In the presence of IL-12, the Th0 differentiates to a Th1 phenotype, and in the presence of IL-4, the Th0 differentiates to a Th2 phenotype. The Th0 cell can also differentiate to a Th3 or T-regulatory 1 (Tr1) phenotype, although the differentiating signals are unknown. *IL*, interleukin; *TGF-ß*, transforming growth factor-ß; *IFN-?*, interferon-?.

The redundancy in cytokine effects probably is related to similarities between cytokine receptors and their associated signal transduction pathways, which can be grouped into four varieties. [51] The hematopoietic cytokine receptor (R) family includes IL-2Rß, IL-2R?, IL-3R, IL-5R, IL-6R, IL-7R, IL-9R, GM-CSFR, G-CSFR, LIF-R, and gp130R. A common feature of these receptors is that they function as multimers with each other and other nonhematopoietic cytokine receptor family members, such as IL-2aR or IL-15aR. The interferon receptor family includes the receptors for type I interferons (IFN-a and IFN-ß) and type II interferon (IFN-?), which share extracellular fibronectin-like domains with the hematopoietic cytokine receptors. The tumor necrosis factor receptors (TNFR) consist of two types: p55 (TNFR1) and p75 (TNFR2). They are homologous to CD95 (Fas antigen), CD40, CD27, and nerve growth factor receptor and are involved in either proliferation or programmed cell death (apoptosis). [25] The receptors for chemokines (either C-C or C-X-C) are seven-membrane–spanning G-protein–linked receptors that are coupled to cell activation through calcium mobilization. The signal transduction pathways that transfer cell surface information to the intracellular milieu for the other cytokine receptors include the JAK/STAT and sphingomyelinase-ceramide pathways. The interferon receptor family and some members of the hematopoietic receptor family (including IL-3, IL-4, IL-6, and GM-CSF) bind and activate the Janus kinases (JAK), which are protein tyrosine kinases that phosphorylate various signal transducers and activators of transcription (STAT) factors that bind and transactivate DNA elements such as IFN-stimulated response elements (ISRE). The TNF receptor family and IL-1R activate sphingomyelinases that induce the release of ceramide as a second messenger from sphingomyelin, thus coupling cell surface receptor events to cellular responses.

CELLULAR INTERACTIONS IN THE IMMUNE SYSTEM

Although lipid and polysaccharide antigens may activate mature B cells to produce immunoglobulins, primarily of the IgM class, B-cell responses to protein antigens require the help of T cells. In the latter process, two requisite events occur. First, the binding of the protein antigen to B cells through membrane immunoglobulin activates the B cell, making it receptive to other signals delivered by T cells. Second, the protein antigen that is bound to membrane immunoglobulin is internalized through coated pits into the endocytic pathway, leading to peptide loading of the class II MHC molecule that the B cell constitutively expresses. Peptide fragments derived from the antigen complementary to the idiotype of membrane immunoglobulin of the B-cell clone are presented to antigen-specific $CD4^{+}$ T cells (Th1- and Th2-type cells). The close intercellular contact required for this B-cell and T-cell interaction makes available the cytokines expressed by helper T cells. The cytokines function as the necessary second signals to B cells for prompting immunoglobulin isotype switching and secretion. The T cell similarly requires and receives complementary signals from the B cell that are a prerequisite for its activation.

The generation of effective cell-mediated immune responses requires the cooperation of cell populations. Delayed hypersensitivity is a T cell–mediated reaction at the site of a perceived foreign substance, as is observed during the course of skin testing with antigens such as purified protein derivative and *Candida* antigen. It is a clinically important immune response to invasive intracellular organisms and to tumors. It is characterized pathologically by granulomas formed by the aggregation of monocytes or macrophages. Foreign antigen, taken up by resident class II molecule–bearing antigen-presenting cells, such as dendritic cells and macrophages, is presented as peptide fragments to antigen-specific $CD4^{+}$ T cells (predominantly Th1 cells). Antigen binding results in T-cell activation, causing the clonal expansion of the antigen-responsive T cells, cytokine secretion by the T cells, and monocyte recruitment. Primarily through the activity of IFN-?, recruited monocytes differentiate into activated macrophages that express higher levels of MHC class II molecules on the cell surface, secrete inflammatory mediators, kill microorganisms and tumor cells, and secrete additional proinflammatory cytokines.

The effective generation of $CD8^{+}$ T cells directed at infected or transformed target cells expressing foreign peptides in the context of class I MHC molecules requires the cooperation of $CD4^{+}$ T cells. The $CD4^{+}$ T cells, which are responsive to peptide fragments derived from internalized antigens, secrete cytokines (e.g., IL-2, IL-4,

IFN-?) that provide the requisite costimulation for CD8 $^{+}$ T-cell differentiation into cytotoxic T cells from precursors normally present at low concentrations in the peripheral blood. Cytolysis of the foreign antigen-bearing target cells is effected by perforin, a membrane pore–forming protein, and granzyme, a cytolysin, stored in preformed granules. CD4 $^{+}$ T cells can also function as cytolytic effectors, primarily through the expression of Fas ligand (CD95 ligand, or CD95L). CD95L is induced on T cells early after T-cell receptor/CD3 complex–mediated activation of T cells. CD95L-bearing cells induce CD95-bearing (or Fas-bearing) cells. Because CD95 is widely expressed, including expression in cells of the lymphoid lineage, the CD95L-CD95 pathway also represents a major mechanism of down-regulating immune responses.

This overview of the immune system is necessarily brief. For more in-depth discussions of these and other topics, the reader is referred to several excellent texts. [52], [53] and [54]

GUT-ASSOCIATED LYMPHOID TISSUE

The cellular elements of the GALT are organized into functional compartments that are anatomically contained within the lamina propria and epithelium. The lamina propria is composed of an organized compartment, including Peyer patches and follicle-associated epithelium, and a nonorganized compartment that is loosely distributed throughout the lamina propria. Peyer patches, which are unencapsulated lymphoid nodules, constitute an afferent limb of the GALT that recognizes antigens through the specialized sampling mechanism of the M cell contained within the follicle-associated epithelium. Detection of substances that move across the small bowel epithelium results in the education and dissemination of B and T lymphoblasts to other tissues linked to the MALT, such as the lungs, breast, and genitourinary tract, and the loosely affiliated compartment of the lamina propria. The lamina propria represents an efferent or effector limb of the GALT, which is populated by lymphoid cellular effectors, such as B cells, plasma cells, T cells, and natural killer cells, and by mononuclear and polymorphonuclear phagocytes and mast cells. The immune compartment within the epithelium consists of a resident population of T cells, the intraepithelial lymphocytes. Evidence from rodent models indicates that dendritic cells found adjacent to the intestinal epithelium may also obtain local antigens and migrate to regional lymph nodes (mesenteric lymph nodes), where immune responses are initiated. [55]

Peyer Patches and Lymphoblast Homing

The follicle-associated epithelium contains M cells, which are derived directly from undifferentiated, immature epithelial stem cells in the crypts that surround the organized lymphoid follicles called *Peyer patches* (Fig. 7-3). The evolution of M cells may be influenced by subjacent B cells within the lamina propria. The morphologic feature that differentiates M cells from absorptive epithelial cells is fewer, shorter, and wider microvilli. M cells cover the lymphoid follicles in the gastrointestinal tract and provide a site for the selective sampling of intralumenal antigens. [3], [56] Clusters of M cells transport particulate molecules such as horseradish peroxidase and ferritin through a transcellular vesicular pathway into the underlying lymphoid tissues of the Peyer patches. Various microorganisms initiate an immune response through this pathway and, in certain circumstances, use the pathway to invade tissues. The mechanism is unknown, but it may involve specific carbohydrate interactions because M cells absorb certain lectins. Among the many infectious agents known to undergo endocytosis and transport by M cells are the human immunodeficiency virus, reoviruses, *Vibrio cholerae,* and species of mycobacteria.

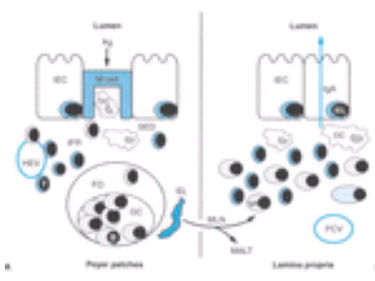

FIGURE 7-3. Antigen processing and lymphocyte trafficking in the gut-associated lymphoid tissue (GALT). **A:** Peyer patches (afferent or inductive limb of GALT). Antigen (Ag) is transported across M cells into a subepithelial pocket containing naive B and T cells in close proximity to professional antigen-presenting cells, such as macrophages and dendritic cells. The antigen-presenting cells engulf, process, and present the antigens, leading lymphocytes to recognize the lumenal antigen. The activated T- and B-cell blasts emigrate from the Peyer patches via regional lymphatics into the mesenteric lymph nodes (MLNs). **B:** Lamina propria and epithelium (efferent or effector limb of GALT). The T- and B-cell blasts migrate through the MLNs and from there enter the thoracic duct and subsequently home to other tissues associated with the mucosa-associated lymphoid tissue (MALT) and back to the loosely affiliated lamina propria and epithelium as intraepithelial lymphocytes (IELs) after emigration through the postcapillary venules (PCVs) of the lamina propria. The B cells differentiate into plasma cells, where they secrete predominantly immunoglobulin A (IgA) and, to a lesser extent, IgG. See text for details. *IEC*, intestinal epithelial cell; *HEV*, high endothelial venule; *IFR*, interfollicular region; *SED*, subepithelial dome; *FO*, lymphoid follicle; *GC*, germinal center; *EL*, efferent lymph.

The antigens and microorganisms transported ablumenally by the M cell come into contact with lymphocytes, macrophages, and dendritic cells that have migrated into the lymphoid aggregates or Peyer patches below the M cells (see Fig. 7-3). Some of these mononuclear cells enter an intercellular space or central hollow that indents the M cell. [56] Naive B and T lymphocytes that have never encountered their cognate antigen express a combination of cell surface receptors that directs them to emigrate into lymphoid aggregates such as Peyer patches associated with follicle-associated epithelium. [57] This emigration occurs through interactions between cell surface receptors on the naive lymphocytes and their counterligands on specialized endothelium, the high endothelial venules present within the organized lymphoid structures. M cells transport selected antigens from the intestinal lumen to facilitate macrophage (and dendritic cell) processing and antigen presentation to the naive lymphocytes in the intestinal lymphoid follicles. A specific mucosal immune response is initiated by these interactions. This interaction may preferentially direct naive T cells to either a Th2, Th3, or Tr1 phenotype over a Th1 phenotype. Such an outcome is likely a consequence of the properties of the dendritic cells within the Peyer patches and plays an important role in generating mucosal tolerance. During infection or pathological conditions, such as inflammatory bowel disease, a Th1 bias may occur relative to Th3 or Tr1. The observation of lysosomal organelles containing MHC class II components in M cells confirms that, under certain circumstances, M cells also may serve as primary antigen-presenting cells. [58]

Activated lymphocytes from intestinal lymphoid follicles begin a maturational journey in which they leave the intestinal tract and migrate into afferent lymphatics that drain into mesenteric lymph nodes. [59], [60] During this process, the lymphocytes mature into T and B lymphoblasts enriched in IgA-bearing B cells. The B lymphocytes become surface IgA-bearing lymphoblasts after being promoted to switch their immunoglobulin isotype by regulatory (i.e., “switch”) T cells within Peyer patches. [6], [7], [61] Switch T cells control isotype switching and B-cell clonal expansion by secreting cytokines (e.g., IL-2, IL-4, IL-5, IL-6, IL-10, and TGF-ß). Lymphocytes then enter the efferent lymphatics of the mesenteric lymph nodes and pass through the thoracic duct into the peripheral blood. These lymphocytes subsequently reenter the loosely affiliated lamina propria through interactions with flat endothelial cells of the postcapillary venules. After homing to mucosal sites, B lymphoblasts mature into IgA-secreting plasma cells under the control of antigen-activated T lymphocytes that have completed a similar maturational journey. Lymphoblasts that have homed to the gastrointestinal mucosa and have matured into effector cells provide protective immunity within the lamina propria.

Lymphoblasts recirculate or home to the sites of the original antigenic stimulation and to other mucosal secretory sites. After antigenic stimulation in the gastrointestinal tract, IgA lymphoblasts circulate to the mucosal secretory sites of the breast, lung, and eye, where antigen-specific antibodies are secreted. [59], [62] A breast-feeding mother can passively transfer secretory IgA in the breast milk to her nursing child. The transferred breast milk secretory IgA protects the infant against bacteria or viruses found within the mother’s gastrointestinal tract, supporting the importance of the common mucosal immune system. [6] Homing of stimulated lymphoblasts to mucosal secretory sites allows the secretion into lung, breast, and eye fluids of protective antibodies directed against antigens recognized within the gastrointestinal lumen. The intestinal immune system thus has components that allow selective antigen sampling and subsequent induction of immune responses that provide protection for the gastrointestinal tract and other mucosal surfaces.

The selective interaction between lymphocyte-specific proteins and counterligands on endothelial cells in specific organs regulates the distribution of lymphoid effector cells to the intestine and other mucosal secretory sites. Antigenic stimulation and chronic inflammation result in a rapid increase in the number of endothelial venules. The number and adhesiveness of endothelial venules increases because of enhanced differentiation and stimulated proliferation. [63], [64] and [65] Cytokines, including IL-1, IFN-?, and TNF-a, increase lymphoblast adherence to endothelial cells, trigger the development of endothelial cell differentiation markers, and

enhance the expression of endothelial adhesion molecules. The increased expression of adhesion molecules on endothelial cells stimulates an increase in the influx of antigen-specific, sensitized lymphocytes into areas of chronic inflammation or areas where cell-mediated host defense processes are needed. Endothelial venules are closely associated with dense, lymphocytic infiltrates, particularly if the mononuclear cell–mediated processes are persistent. [63], [64]

Nonorganized Lamina Propria

The efferent, or effector, compartment of the lamina propria consists of T cells, B cells, plasma cells, natural killer cells, phagocytic cells, and mast cells. About 60% of the lymphoid cells are T cells. $CD4^+$ and $CD8^+$ T cells occur in a ratio of about 2:1, which approximates their concentrations in peripheral blood. [65], [66] Virtually all T cells within the lamina propria express the aß T-cell receptor. Closer examination of aß T-cell receptor usage shows some skewing, with an overabundance of certain receptors, suggesting the expansion of some T-cell clones. [65] The CD45 isoform expression confirms that these cells are memory cells, indicating previous encounters with antigens, presumably in the Peyer patches (see Table 7-2). [66] The $CD4^+$ cells in the lamina propria exert a helper-inducer function for immunoglobulin production, a major function of lamina propria T cells. [67], [68] Consistent with this, in response to antigen, lamina propria lymphocytes (LPLs) respond primarily with cytokine production rather than proliferation. [67] Most $CD8^+$ LPLs as well as a significant proportion of $CD4^+$ LPLs express a unique $ß_7$-integrin associated with a novel a chain, a ^{E}, which plays a role in intestinal epithelial cell binding. [69], [70] The population of $CD8^+$ LPLs probably contains precursors, such as TCR-aß $^+$, $CD8^+$, a $^E ß_7^+$ ($HML\text{-}1^+$), and $CD45RO^+$ T cells, in transit to the epithelium.

A set of lymphocytes in the intestine are cytotoxic effector precursor cells that can participate in host mucosal defense mechanisms when needed, without continuously causing damage to the surrounding tissue when not needed. Functional precursor natural killer cells and cytotoxic T lymphocytes can be enriched from isolated lamina propria mononuclear cells. LPLs can be induced to mediate cell-mediated cytotoxicity by incubation with IL-2, IFN, lectins, and monoclonal antibodies directed against the T-cell receptor, typical of antigen-primed effectors and consistent with their CD45RO phenotype. [71], [72], [73], [74], [75], [76] and [77] Although cytotoxic effectors are present, the intestinal LPLs generally are poor mediators of cell-mediated cytotoxicity in a variety of systems, including spontaneous cell-mediated cytotoxicity, antibody-dependent cellular cytotoxicity, and cell-mediated cytolysis. [71], [72]

Most mucosal T lymphocytes are also $CD95L^+$ and $CD69^+$ and exhibit elevated levels of cytoplasmic Ca^{2+}, consistent with an activated phenotype. [78] Controlled activation of the intestinal immune system may be important in regulating effector cell function. This includes cytotoxic function that may be directed at the lymphocytes themselves for the purposes of down-regulating immune responses. [79] In this way, the gut can remain in a state of physiological inflammation, poised for intervention when necessary but maintaining a general tone of restraint. Humoral extracts from the healthy lamina propria, containing humoral substances of unknown origin, are capable of suppressing the activation of peripheral nonintestinal lymphoid populations, which may account for the low level of responses to proliferative signals delivered by the T-cell receptor–CD3 complex. [80]

About 40% of the lymphoid cells in the lamina propria are B cells derived primarily from precursors in Peyer patches. [81], [82] These B cells and their progeny plasma cells are predominantly focused on IgA synthesis and focused to a lesser extent on IgM, IgG, and IgE synthesis. Lamina propria B cells are induced to differentiate terminally into IgA-secreting cells by IFN-?, IL-4, IL-5, IL-6, and IL-10. [4], [7], [62], [83], [84], [85], [86], [87], [88] and [89] IL-4 and IL-5 activate resting B cells and induce the division and growth of activated B cells. IL-6 is critical for the terminal differentiation of IgA plasma cells, resulting in the secretion of large amounts of IgA. The sequence of cytokine-mediated events that regulates B-cell growth, differentiation, and development toward IgA production involves both Th1 (IFN-?) and Th2 (IL-4, IL-5, IL-6, IL-10), indicating the importance of this antibody to mucosal protection. In diseases such as inflammatory bowel disease, the numbers of lamina propria B cells and plasma cells, which produce IgG, are markedly increased. [10]

Immunoglobulin Secretion

A major protective immune mechanism for the intestinal tract is the synthesis and secretion of dimeric IgA. The intestine contains more than 70% of the immunoglobulin-producing cells in the body. [2], [90] Although IgG and IgM antibodies are also produced by lamina propria B cells within the normal intestine, the predominant antibody synthesized and secreted is IgA. Evidence suggests that IgE and even IgG also may be important secretory immunoglobulins (Fig. 7-4). [90], [91] and [92]

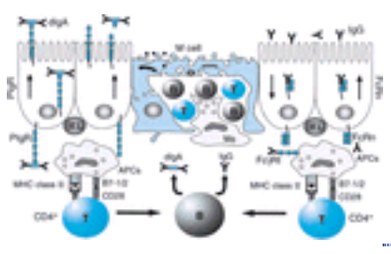

FIGURE 7-4. Immunoglobulin transport across intestinal epithelial cells. The interaction between professional antigen-presenting cells (APCs) and $CD4^+$ T cells in the lamina propria induces local B cells to secrete predominantly immunoglobulin A (IgA) and IgM and, to a lesser extent, IgG, except in inflammatory conditions such as inflammatory bowel disease, in which significant amounts of IgG are produced. IgA is secreted from the B cell as a dimer in association with J chain (dIgA). This polymeric IgA and IgM (which is pentameric) is taken up by the polymeric immunoglobulin receptor (pIgR) along the basal surface of the intestinal epithelial cell. The pIgR then transports the dimeric IgA and IgM across the cell in a transcytotic pathway to the apical cell surface. At the apical cell surface, cell surface proteases cleave the pIgR, releasing the polymeric IgA and IgM in association with secretory component, a proteolytic fragment of the pIgR. In a similar manner, the neonatal receptor for IgG (*FcRn*) transports local IgG transcellularly in a transcytotic pathway. However, in contrast to the pathway associated with the pIgR, the FcRn pathway is bidirectional and not associated with cleavage of the FcRn.

There are two IgA subclasses: IgA1 and IgA2. In serum, less than 15% of the IgA is IgA2, but in external secretions, as much as 50% of IgA is IgA2. [93] One possible explanation for the preferential production of IgA2 in intestinal secretions is the observation that IgA1 is easily cleaved by proteases produced by bacteria, but IgA2 is more resistant to cleavage and may survive longer in the intestinal lumen. [94] Plasma cells produce IgA in monomeric form or as a dimeric structure in which two IgA monomers are joined by a polypeptide called *J chain*. J chain is also produced within the plasma cell. [95] J chain produces polymerization of IgA and IgM. By subsequent interaction with another polypeptide on the basal surface of the epithelial cell, called *polymeric immunoglobulin receptor* (pIgR), J chain participates in the transport of polymeric IgA and IgM molecules across the intestinal epithelial cell into the lumen. [90], [96], [97]

pIgR, produced by epithelial cells, is a membrane receptor that exhibits selective binding to polymeric immunoglobulins such as IgA and IgM, which contain J chain. [95], [98], [99], [100], [101], [102], [103] and [104] pIgR is synthesized in the rough endoplasmic reticulum of the epithelial cell, glycosylated in the Golgi complex, and directed to the basolateral cell surface by a sorting signal in the cytoplasmic tail. [101], [102] After the initial interaction between polymeric IgA and the pIgR, the entire IgA-pIgR complex undergoes endocytosis into the basal side of the epithelial cell. The IgA-pIgR complex is then transported to the apical surface of the epithelial cell in endocytic vesicles that fuse with the apical membrane and subsequently release a portion of the pIgR, secretory component, as an intact secretory IgA molecule into the intestinal lumen.

Secretory component remains bound to polymeric IgA after its release into the external secretions, although it can also be found by itself in mucosal secretions as a secreted glycoprotein not bound to polymeric immunoglobulins. [95], [98], [99], [100], [101], [102], [103] and [104] Secretory component may prevent proteolytic degradation of the secretory IgA molecule and may stabilize the structure of the polymeric IgA complex, protecting the secretory IgA that is secreted into a hostile environment containing numerous proteolytic enzymes, bacteria, and other substances that could otherwise rapidly degrade it.

IgA is also translocated across the hepatocyte or bile duct epithelium into the bile. [105], [106] IgA in the bile is carried into the duodenum. Important interspecies differences have been observed; IgA and secretory component are major components of the bile of the rat and rabbit, whereas the bile of sheep, dogs, and humans

contains much smaller amounts of IgA. These differences in IgA translocation into bile appear to be related to the presence of pIgR on hepatocytes in rats and rabbits, resulting in highly efficient movement of IgA into the bile, but pIgR is expressed only on biliary epithelium in humans, resulting in less efficient IgA translocation. The presence of secretory IgA in bile provides passive immunity and protection for the biliary tract and the proximal parts of the small bowel. A second implication of the hepatobiliary secretion of IgA is that complexes of IgA and antigen can be transported into the bile from the circulation. Hepatic removal of IgA-antigen complexes may protect against harmful absorbed substances, including dietary antigens and bacterial products.

The major function of secretory IgA in host defense is protection against bacteria, viruses, and lumenal antigens. [2], [4], [59] Secretory IgA inhibits the adherence of bacteria to epithelial cells and prevents their effective colonization and multiplication. Secretory IgA neutralizes bacterial toxins and prevents their action on intestinal epithelial cells. The major antiviral properties of IgA are neutralization of viral activity and passive protective immunity. Secretory IgA also blocks the absorption of antigens from the gut and may be particularly important in disease states in which the mucosal barrier is broken. IgA also is able to neutralize intracellular pathogens within a cell that possesses a transcytotic pathway for IgA. [107] The intersection of endocytic compartments containing pathogens and secretory IgA leads to neutralization of the foreign substances and their excretion after transport to the apical surface.

IgA does not activate complement and does not enhance cell-mediated opsonization or destruction of infectious organisms or antigens. In this lack of complement activation, IgA sharply contrasts with other immunoglobulins, such as IgG, which can also be secreted by B cells in the intestine and initiate important complement-mediated and cell-mediated protective events within the intestine.

Transcellular pathways for IgE and IgG transport also exist in epithelial cells. The activities of CD23 (FceR1, or IgE receptor), whose expression in epithelial cells is regulated by IL-4, appear to direct IgE vectorially from the lumen into the tissues. [108] Such a pathway may be very important to intestinal allergic responses. High concentrations of IgE in the lumen during parasitic infestations suggest an alternate pathway of IgE transport from the tissues into the lumen. However, the molecular basis for this pathway is unknown.

Transcytosis of IgG also occurs across intact epithelial barriers during adult life in rodents and humans. [109], [110] This process is mediated by the MHC class I–related molecule FcRn (neonatal Fc receptor for IgG). In contrast to the pathway related to the pIgR, the FcRn-mediated pathway is bidirectional (apical to basal and basal to apical) and is not associated with proteolytic cleavage of the transporting receptor such that multiple rounds of transport are possible. The FcRn binds to the Fc portion of IgG at acidic pH (pH 6.0), the pH of endosomes, and releases IgG at neutral pH (pH 7.4), the pH of the interstitium. Such a pathway may endow the epithelium with a means to monitor the antigens within the lumen without disrupting the epithelial barrier.

Intraepithelial Lymphocytes

The epithelium of the human intestine contains a unique population of lymphoid cells, the intraepithelial lymphocytes (IELs), that reside between intestinal epithelial cells along their basolateral surface. [111] A few IELs are $CD4^+$ T cells, but most IELs express the CD8 aß heterodimer, the CD45RO isoform, and the aß T-cell receptor, indicating that they are memory cells driven by MHC class I or class I–like molecules. [112], [113] Between 5% and 30% of these T cells, especially in the colon, express the ?d T-cell receptor. [114] Virtually all the ?d T cells in the human intestine reside within the epithelium, suggesting that they are responsive to specific chemotactic factors or antigens within this location. [115] More than 95% of IELs express the unique $a^E ß_7$-integrin, HML-1, which, in view of its role in epithelial cell binding, probably plays an important role in the epithelial cell tropism of these cells. [69], [116] The $a^E ß_7$-integrin binds E-cadherin, an immunoglobulin supergene family member, on the basolateral surface of the intestinal epithelial cell. [73] In the presence of TGF-ß, presumably secreted by local intestinal epithelial cells, high levels of this integrin are expressed somewhat reciprocally to the $a^L ß_2$-integrin lymphocyte-function associated antigen-1 (LFA-1; i.e., CD11a/CD18), a $ß_2$-integrin commonly expressed by peripheral blood lymphocytes. [69], [116]

Despite their contiguity to the gut lumen, potential exposure to a variety of antigens, and the expectation that these cells express a diverse, polyclonal array of aß and ?d T-cell receptors, IELs within the small and large intestine are oligoclonal and express a small number of aß and ?d T-cell receptors based on an analysis of CDR3 regions. [65], [117], [118] A limited variety of T-cell clones are, in fact, widely disseminated throughout the intestinal epithelium. [119] With a CD45RO phenotype, [120] these cells are memory cells that recognize an extremely limited number of antigens, not the multitude of lumenal antigens. The abundant expression of CD8 further indicates that these cells recognize these putative antigens in the context of an MHC class I or class I–like molecule such as CD1 or the MHC class I chain–related gene A gene product. [34], [121], [122] The CD8 expression of IELs suggests that they function biologically as cytolytic effectors as a consequence of antigenic recognition. IELs exhibit a high level of cytolytic activity in a variety of in vitro systems, especially after activation, [123] and likely in disease states in vivo. IELs in situ, however, do not express granzyme, perforin, or CD94L. [124] This suggests that their major biologic function in health is the secretion of cytokines (e.g., IFN-? and keratinocyte growth factor), which regulate epithelial cell function and possibly responses to lumenal antigens. [125] ?d IELs, for example, may play a role in regulating responses to orally delivered antigens. [126] On activation, IELs may acquire cytolytic machinery that can contribute to epithelial cell death through apoptosis. Their cytolytic capabilities, large number, and extremely limited T-cell receptor repertoire together indicate that IELs are a regionally specific population of cells involved in immunosurveillance against abnormal epithelial cells based on the recognition of a limited number of proteins not normally expressed on the cell surface of intestinal epithelial cells. IELs may be the first line of defense against deleterious epithelial events. Their numbers are markedly increased in intestinal graft versus host disease, gluten-sensitive enteropathy, and protozoal infections of the epithelium, such as those caused by *Cryptosporidium* and *Isospora* species.

Human IELs and the $CD8^+$ T cells within the lamina propria share many phenotypic characteristics, but their origin is unclear. Studies of murine systems suggest the existence of two IEL populations: thymus-dependent (i.e., selected in the thymus) and thymus-independent (i.e., selected in the intestine) lymphocytes. [127] A feature of thymus-independent mouse IELs is the expression of CD8 as an aa homodimer rather than the more common aß heterodimer and the expression RAG transcripts. Considering the rarity of this form of CD8 in normal human adult intestine, the extrapolation from the mouse models to humans remains uncertain. Nonetheless, mouse models of rotavirus and reovirus infections indicate that TCR-$aß^+$, $CD8^+$, MHC class I–restricted, virally specific cytotoxic effectors can be found within the epithelium that are probably derived from cytotoxic T-cell precursors in the Peyer patches. [128], [129] These results, taken together with those of other studies showing that most IELs are restricted by classic MHC class I molecules, [130] suggest that IELs are likely to be the clonally expanded progeny of MHC class I–restricted cytolytic effector cells.

AUTOIMMUNITY AND ORAL UNRESPONSIVENESS

Autoimmunity

Despite the processes of negative and positive selection that occur within the thymus for T cells and within the bone marrow for B cells, healthy persons maintain low numbers of autoreactive T-cell and B-cell clones. Rather than a failure of immunologic selection within the central immunologic compartments, the existence of these autoreactive clones may represent the need for B-cell and T-cell clones that recognize dominant microbe-derived peptides to which a person may be exposed—peptides that resemble or mimic peptides derived from normal self-antigens. [131]

The intestine also may be a site of thymus-independent T-cell maturation. [128] The evidence remains unclear for humans, but mice have a pathway of T-cell development in which the intestinal epithelial cell may play a central role in T-cell selection. Studies of mouse models suggest that the selection pathways operating in the intestine may not involve classic MHC class I or II molecules. [15]

The normal host must therefore maintain active peripheral mechanisms within tissues, including the intestine, to prevent potential autoreactive clones from becoming autoimmune clones that participate in immune-mediated diseases. This presumably is accomplished by suppressor T-cell networks, the effects of immunosuppressive cytokines (e.g., TGF-ß, IL-10), the negative feedback effects of antibodies on B cells, the presentation of antigenic peptides to the T-cell receptor in the absence of an appropriate costimulatory signal, and the existence of anti-idiotype networks (i.e., antibodies that bind to the antigen-binding site of an antibody and block its function). [132] The end result of these potential mechanisms is active suppression, clonal anergy, or clonal deletion through apoptosis.

Because the gastrointestinal tract must continuously mount immune responses against bacteria, viruses, and other antigens, molecular mimicry or immune dysregulation leading to the activation of self-reactive T cells and the development of autoimmune diseases in genetically predisposed hosts is possible. These mechanisms are indirectly supported by several mouse gene knock-out models. In mice made deficient in the genes for IL-10, TGF-ß, IL-2, IL-2R, MHC class II molecules, and TCR-a, an inflammatory bowel disease–like process develops that includes the production of autoantibodies. [133], [134], [135], [136] and [137] This suggests that

the loss of crucial control mechanisms may allow the release of autoreactive clones that are triggered by microbial antigens because bacterial colonization may be essential for the development of the disease process in these animals. Autoimmunity appears to be relevant in the development of chronic inflammatory disorders of the gastrointestinal tract. Autoimmune responses to tissue transglutaminase stimulated by gliadin in genetically susceptible individuals who express particular MHC class I molecules may be an essential feature of celiac sprue. [138] In autoimmune gastritis, inappropriate cellular responses to the H^+,K^+-ATPase in association with excess Th1 cytokine production are observed. [139]

After the autoreactive T-cell clones have been activated, a variety of effector mechanisms can be brought into play. Cytokines can destroy host cells directly and activate other cell types to mediate tissue damage. Cytokines such as IFN-? enhance MHC class II expression on cells such as epithelial cells, leading to increased presentation of autoantigens or lumenal antigens, which may result in other potential molecular mimicry events. T-cell regulation of B cells through cytokines may lead to the generation of IgG autoantibodies, which, through activation of complement, may lead to the production of chemotactic factors and an increasing influx of various cell types, including macrophages, granulocytes, and eosinophils. In models of chronic idiopathic intestinal inflammation, an imbalance of cytokines is common; for example, in a situation of excessive production of type 1 cytokines relative to type 2 cytokines, inhibition of the type 1 cytokines mitigates the inflammation.

Oral Tolerance

Because the gastrointestinal tract contains many antigens, including dietary proteins, that can lead to cross-reactivity and activation of self-reactive T cells, an important function of the gastrointestinal immune system is the generation of a state of tolerance to mucosal antigens. The gastrointestinal tract exhibits a fascinating example of specific tolerance to orally ingested antigens, called *oral tolerance*. [140], [141], [142] and [143]

The oral administration of antigens can lead to systemic antigen-specific unresponsiveness, which results in the lack of specific T-cell and B-cell responsiveness to those antigens. Concurrently, local specific secretory immunity can develop, resulting in lymphoblasts capable of IgA production. [143] This dichotomy between mucosal and systemic compartments appears to reflect a solution to the need for excluding the specific antigen during future encounters and avoiding inappropriate systemic responsiveness. The ability of antigens in the gastrointestinal tract to induce a state of functional anergy is precise. The state of unresponsiveness or anergy holds only for the specific antigen that was presented orally to the gastrointestinal tract and depends on the nature of the antigen. Not surprisingly, the gastrointestinal tract possesses mechanisms to protect against mounting adverse immunologic reactions. If this were not the case, numerous bacterial and viral antigens and food components could lead to frequent cross-reactive immunologic stimulatory events and result in intestinal autoimmune disorders, and many food substances could give rise to diverse and uncontrollable food-induced allergic reactions.

In model systems, the induction of oral tolerance depends on the type of antigen, the amount of antigen, the frequency of antigen sensitization, the type and genetic background of the animal being studied, the age of the animal, and the particular immune response being evaluated. [140], [141] and [142] Two mechanisms of tolerance have been hypothesized: T-cell suppression and clonal anergy. The application of low doses of antigen to the intestine appears to induce the development, within Peyer patches and mesenteric lymph nodes, of $CD4^+$ T cells capable of secreting TGF-ß (Th3) and $CD4^+$ T cells with a Th2 cytokine profile, capable of secreting IL-4 and IL-10. [141] These cell types migrate to a variety of lymphoid and nonlymphoid tissues, where they inhibit the generation of antigen-specific effector cells. High doses of a tolerogen appear to induce the clonal anergy or apoptosis of antigen-specific Th1 cells. [142] The administration of high doses of antigen may lead to systemic leakage of the antigen, causing inappropriate antigen presentation in the periphery by antigen-presenting cells that lack the necessary costimulatory second signal.

The successful induction of oral tolerance may help prevent the initiation of autoimmune diseases. Mice prone to the development of autoimmune disease exhibit defects in the induction of oral tolerance. The complex regulatory processes operating in the mucosal immune system facilitate the development of a local protective mucosal immune response against pathogenic organisms and prevent the development of adverse systemic autoimmune reactions to the same antigens. Although oral tolerance creates an impediment to vaccination, certain mucosal adjuvants, such as cholera toxin, can reverse oral tolerance. [144]

Oral tolerance is used to advantage in treating several autoimmune diseases. In murine models, feeding myelin basic protein, collagen type II, S antigen (i.e., retinal autoantigen), porcine insulin, and class II MHC peptides suppresses experimental allergic encephalitis, collagen- or adjuvant-induced arthritis, experimental allergic uveitis, diabetes mellitus, and organ transplant rejection, respectively. [145], [146], [147] and [148] This approach is being tested in the treatment of systemic human diseases and inflammatory conditions of the intestine itself. [148]

GASTROINTESTINAL INFLAMMATION

The process of inflammation consists of the activation of neutrophils and monocytes and the effects of that activation on endothelial cells, epithelial cells, and other cell types. Inflammation is a prominent component of a number of important gastrointestinal diseases, including gastroesophageal reflux disease, pancreatitis, peptic ulcer disease, *Helicobacter pylori* gastritis, inflammatory bowel disease, and various enteric infections.

The immune response and the inflammatory response are entwined. From one point of view, the inflammatory response is an effector mechanism for the immune response. For example, when a bacterial infection activates the immune system, antibodies that are specific to the invading bacteria are produced and bind to the bacteria, and neutrophils that express antibody receptors on their surface efficiently phagocytose the antibody-coated bacteria. In this example, a portion of the inflammatory response—that is, phagocytosis by neutrophils—acts as an effector mechanism for the humoral component of the immune response. From another point of view, the inflammatory response is a damage control mechanism that is invoked only if the immune response breaks down or is overwhelmed. In most instances, immune processes eliminate foreign antigens without the development of clinically apparent inflammation. The histology of the normal small intestine and colon demonstrates immune activation and a low-grade inflammatory response, which result from the stimulation of lamina propria immune cells by lumenal antigens. In health, this limited immune activation and low-grade inflammation are clinically silent. The development of clinically apparent inflammation suggests that the immune system has been overwhelmed by a large antigen load, by an antigen in a location inaccessible to the immune response (e.g., osteomyelitis), or by an antigen that is resistant to the immune response (e.g., tuberculosis). Some chronic inflammatory diseases, such as inflammatory bowel disease, may result from dysregulation of the immune response, resulting in an inappropriately prolonged and inappropriately amplified immune activation. [149]

Inflammation also can be viewed as a component of the larger process of wound healing [150] (Fig. 7-5). After injury, blood clotting and fibrinolysis are followed by the infiltration of inflammatory cells. Surviving epithelial cells migrate to cover the epithelial defect induced by injury. This process overlaps with the influx of inflammatory cells. The migration of inflammatory cells and the repair of the epithelium are regulated by the same cytokines and mediators. Events that affect inflammation also affect epithelial healing, and events that affect epithelial healing affect inflammation.

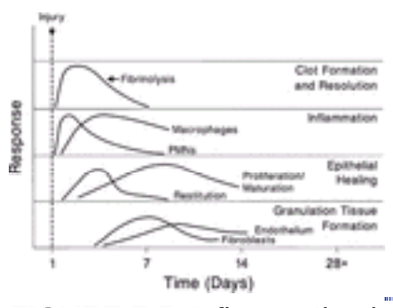

FIGURE 7-5. Inflammation is a component of the larger process of wound healing. Each curve represents the relative intensity of the biologic activity or the concentration of the biochemical or structural component with a time course beginning with injury on day 1. The earliest events are clot formation and resolution. At about the same time that clot forms, neutrophils begin to enter the injured area. Soon after neutrophil migration begins, monocytes enter the area of injury and differentiate into macrophages. Epithelial cells from the edge of the wound rapidly migrate to restore the integrity of the epithelial layer. Full epithelial healing requires the proliferation of additional epithelial cells. Later stages of wound healing include the proliferation of fibroblasts and neovascularization.

Various initiating agents, including aspirin, ethanol, and radiation, result in gastrointestinal inflammation, but the evolutionary pressures that formed the inflammatory

response were not the pressures of aspirin, ethanol, and radiation, but rather those of infectious agents. Our ancestors were exposed to contaminated food and water and poor sanitation, which resulted in viral, bacterial, and parasitic gastrointestinal tract infections. The inflammatory response evolved to deal with these infections. Many of the inflammatory diseases of the gastrointestinal tract today reflect the activation of defense mechanisms that evolved in response to infectious agents but have been adapted (or maladapted) to defense against noninfectious insults. A corollary to this observation is that if the inflammatory response defends against infectious agents, then therapeutic attempts to diminish the inflammatory response may impair the host's ability to respond to infection.

The immune response in the GALT differs in many respects from the systemic immune response. In contrast, gastrointestinal inflammation is similar to inflammation in other organ systems. The inflammatory cells, the cytokines, and the mediators that respond to shigellosis are similar to those that respond to pneumococcal pneumonia. One distinguishing feature of gastrointestinal inflammation is the enormous antigen load in the intestinal lumen. No other organ is faced with foreign antigens in such abundance. This antigen load, particularly the bacterial component, is the stimulus for the chronic low-grade inflammatory response in the healthy gastrointestinal tract. The differences between the histology of the gastrointestinal tract of germ-free rodents and that of conventional rodents are striking. [151] The expected difference is fewer lymphocytes and macrophages in the germ-free animals; the Peyer patches are smaller with fewer germinal centers, and plasma cells are absent. The nonimmune cells also exhibit differences. In germ-free animals, the villi are thinner and the crypts shallower. Epithelial proliferation is diminished, and the time required for epithelial cells to migrate from the crypt to the villus tip is doubled. It is not known what portion of these differences represents the direct effects of bacteria on epithelial cells and what portion represents indirect effects mediated through immune cells.

The epithelium is essential to the control and regulation of the inflammatory response in the gastrointestinal tract. The intact epithelium acts as a barrier, preventing the immune cells in the lamina propria from being overwhelmed by the antigens in the lumen. Breaks in the epithelial monolayer result in a clinically apparent inflammatory response. Epithelial breaks expose lamina propria immune cells to the several hundred species of bacteria that flourish in the human colon. The diversity of antigenic stimuli triggers nonspecific inflammatory responses. In diseases marked by breaks in the epithelial barrier, the immune or inflammatory response to the initiating agent may be difficult to distinguish in the midst of the response to the normal intestinal flora that invade through the epithelial breaks.

Gastrointestinal inflammation is remarkable in that diverse initiating events, such as infections, ischemia, radiation, and chemical toxins, all induce inflammatory responses that are clinically, endoscopically, and histologically similar. The colonoscopic and histological appearances of ulcerative colitis, radiation proctitis, shigellosis, and ischemic colitis are similar. The commonality of the inflammatory responses is explained partly by the similarity between the proinflammatory cytokines, the patterns of leukocyte migration, and the inflammatory mediators induced by these initiating events (Fig. 7-6). Moreover, in each of these diseases, the inflammatory response is largely directed against normal colonic flora that has activated the immune cells of the lamina propria as a result of the loss of epithelial integrity.

FIGURE 7-6. The intestine has a stereotyped inflammatory response to injury. One of the remarkable features of intestinal inflammation is that a wide variety of initiating events—including infection, ischemia, radiation, and chemical toxins—can induce intestinal inflammatory responses that are remarkably similar histologically, endoscopically, and clinically. The reason for the commonality of the inflammatory responses induced by diverse initiating events is that the proinflammatory cytokines, patterns of leukocyte migration, and mediators of inflammation induced are all largely similar.

Gastrointestinal inflammation is distinguished also by the organ-level physiological response, primarily increased motility and secretion. [152], [153] and [154] The gastrointestinal tract responds to enteric infections by attempting to wash out the offending microbes with increased electrolyte and water secretion and increased motility. Increased secretion and motility result in cramps and diarrhea, which are prominent clinical features of enteric infections. The increases in secretion and motility are mediated by the inflammatory response. Because the inflammatory response in the gastrointestinal tract is stereotyped irrespective of the initiating event, increased secretion and motility (and the resultant cramps and diarrhea) are also part of the clinical picture in those inflammatory diseases (e.g., radiation enteritis and inflammatory bowel disease) in which there are no pathogenic organisms to wash out.

ADHESION MOLECULES AND CELL TRAFFICKING

A healthy gastrointestinal tract contains many neutrophils, macrophages, and lymphocytes. Most of these cells are found in the lamina propria, the space between the epithelium and the muscularis mucosae. These three cell types arise from the bone marrow, but their life histories are different.

Neutrophils differentiate in the bone marrow before entering the peripheral blood. Leaving the peripheral circulation, they enter the gastrointestinal tissues by binding to adhesion molecules expressed on the endothelium of postcapillary venules. After only 1 or 2 days in the gastrointestinal tract, neutrophils pass between epithelial cells into the intestinal lumen, where they die and are expelled in the stool. Although neutrophils differentiate in the bone marrow, they are primed and activated in the lamina propria. [155] Priming enhances the ability of neutrophils to produce reactive oxygen species, and activation induces the production of reactive oxygen species. Neutrophil priming and activation are mediated by interaction with particulate stimuli (e.g., bacteria) or by stimulation with soluble factors, such as cytokines, inflammatory mediators, and bacterial products (e.g., endotoxin). Neutrophils are incapable of proliferation; the increase in the number of neutrophils in the lamina propria in inflammatory states reflects increased trafficking out of the bloodstream and into the gastrointestinal tissues.

Gastrointestinal macrophages are derived from circulating monocytes produced in the bone marrow. [156], [157] The large increase in the macrophage numbers in clinically apparent inflammation reflects the increased migration of monocytes out of the bloodstream and into the lamina propria rather than proliferation of the resident macrophages. Monocytes enter the circulation and, like neutrophils, bind to adhesion molecules expressed on endothelial cells in the postcapillary venules of the intestine. After binding to these adhesion molecules, monocytes pass between endothelial cells and enter the gastrointestinal tissue.

After entering the lamina propria, the monocyte begins to differentiate into a mature macrophage. As the monocyte differentiates, it can acquire capacities for phagocytosis, proliferation, and bacterial killing. Macrophage differentiation is controlled by cytokines and other soluble factors present in the lamina propria; different combinations of cytokines and mediators result in macrophages with different phenotypes. Macrophage phenotypes are characterized by their surface receptors; these surface receptors determine the stimuli to which the macrophage can respond. No detailed surveys exist of the life span of macrophages in the gastrointestinal tract, but as a group their stay is far longer than that of neutrophils.

For the most part, neutrophil and monocyte trafficking in the gastrointestinal tract is similar to that in other organ systems. The mechanisms of leukocyte trafficking in various organs are qualitatively similar, but the number of leukocytes passing through the gastrointestinal tract greatly exceeds that in other organs. The gastrointestinal tract has a large surface area; as a result, even the modest degree of inflammation seen in the healthy small intestine and colon represents the trafficking of a substantial number of monocytes and neutrophils. In diffuse inflammatory diseases of the gastrointestinal tract, such as ulcerative colitis, the trafficking of leukocytes through the inflamed mucosa expands to the point that most leukocytes produced in the bone marrow travel through the gastrointestinal mucosa into the lumen. [158]

The migration of neutrophils and monocytes from the peripheral circulation into the lamina propria of the gastrointestinal tract is mediated by the expression of adhesion molecules on vascular endothelial cells and on the leukocytes themselves [159], [160] (Fig. 7-7). Adhesion molecules play an important role in many biologic processes—not only in interactions between inflammatory cells and vascular endothelium but also in interactions between different inflammatory cell types, between inflammatory and noninflammatory cells, and in the binding of cells to the extracellular matrix. [161] The adhesion molecules that participate in the binding of inflammatory cells to vascular endothelium fall into three groups: selectins, ß $_2$-integrins, and the immunoglobulin superfamily of adhesion molecules (Table 7-4).

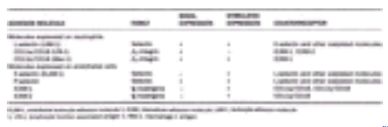

TABLE 7-4 Adhesion Molecules Involved in Neutrophil Binding to Endothelial Cells

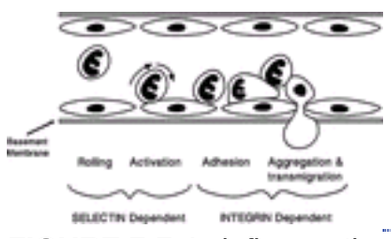

FIGURE 7-7. In inflammation, neutrophils bind to the vascular endothelium through cellular adhesion molecules. Cytokines and inflammatory mediators induce the expression of adhesion molecules on neutrophils and endothelial cells. The initial interactions are mediated by selectins. The weak bonds formed by selectins cause the neutrophils to slow down and roll along the endothelial surface. Later, stronger bonds are formed between the integrins on the surface of the neutrophils and adhesion molecules on the endothelial cells. These stronger bonds firmly attach the neutrophil to the endothelium; after attaching to the endothelial cells, the neutrophils migrate between them.

As inflammation is initiated, leukocytes and endothelial cells express selectins. The three members of the selectin family are L-selectin (leukocyte adhesion molecule-1 [LAM-1]), E-selectin (endothelial leukocyte adhesion molecule-1 [ELAM-1]), and P-selectin. [162], [163] The natural ligands for all three selectins are sialylated Lewis X oligosaccharides, which are found on almost all cell types. Among the molecules with sialylated Lewis X moieties are the selectins themselves, so that L-selectin on neutrophils can bind to E-selectin or P-selectin on endothelial cells.

L-selectin is expressed on lymphocytes, monocytes, and neutrophils; it mediates their adherence to endothelial cells. L-selectin expression is stimulated by inflammatory cytokines (e.g., IL-1 and TNF-a), mediators of inflammation (i.e., leukotriene B $_4$ [LTB $_4$]), and lipopolysaccharide. E-selectin is found only on stimulated endothelial cells, where it promotes leukocyte adherence. It is expressed in response to cytokine stimulation (e.g., IL-1, TNF-a, and IFN-?). P-selectin is expressed on platelets and is involved in thrombosis. It is also expressed on endothelial cells and is involved in leukocyte adhesion. In contrast to E-selectin, which is expressed only in response to stimulation, P-selectin is constitutively expressed on endothelial cells, at least in some organs. Proinflammatory cytokines, histamine, and lipopolysaccharide induce P-selectin expression, but different organs have different levels of sensitivity. Lipopolysaccharide induces endothelial expression of P-selectin and E-selectin, whereas histamine induces P-selectin but not E-selectin. [164]

Selectin bonds are responsible for leukocyte rolling. The selectin bonds that form between leukocytes and endothelial cells are weak; the weakness of these bonds allows leukocytes to roll along the surface of the endothelium by making and breaking selectin-mediated bonds (see Fig. 7-7). Rolling reduces leukocyte velocity before the formation of stronger adhesion bonds that fully immobilize leukocytes on the surface of the endothelium. These stronger bonds are formed between ß $_2$-integrins expressed on the surface of the leukocytes and intercellular adhesion molecules (ICAMs)-1 and -2, members of the immunoglobulin superfamily of adhesion molecules, which are expressed on endothelial cells.

The integrins form a large group of adhesion molecules. [165] Each integrin is a heterodimer that consists of noncovalently associated a and ß subunits. Integrins are divided into subfamilies based on common ß subunits; the ß $_1$ and ß $_2$ subfamilies are the most important in inflammation. ß $_1$-Integins are involved in lymphocyte trafficking. ß $_2$-Integrins are involved in the adhesion of monocytes and neutrophils to endothelial cells. One ß $_2$-integrin, CD11a/CD18 (LFA-1), binds to both ICAM-1 and ICAM-2 on endothelial cells. Another ß $_2$-integrin, CD11b/CD18 (macrophage-1 antigen [Mac-1]), binds only to ICAM-1. CD11a/CD18 is expressed on neutrophils in the basal state, and its expression is not enhanced by cytokines or inflammatory mediators. In contrast, the expression of CD11b/CD18 on leukocytes is induced by products of bacteria (e.g., *N*-formyl-methionyl leucyl phenylalanine [fMLP]) and inflammatory cells (e.g., TNF-a, IL-1, IL-8, LTB $_4$, and platelet-activating factor [PAF]) (Fig. 7-8).

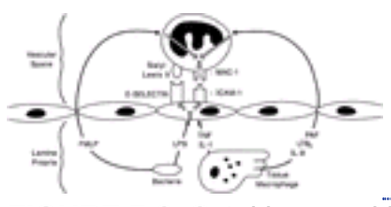

FIGURE 7-8. Cytokines and inflammatory mediators induce leukocytes and endothelial cells to express adhesion molecules. The expression of adhesion molecules on leukocytes (e.g., MAC-1) and endothelial cells (e.g., E-SELECTIN, ICAM-1) is induced by various agents found in inflammatory processes. Among these agents are bacterial products, including fMLP and lipopolysaccharide. Also important in the expression of adhesion molecules are cytokines (TNF, IL-1, IL-8) and lipid mediators of inflammation (LTB $_4$, PAF) produced by tissue macrophages and other inflammatory cells. Other cytokines (e.g., TGF-ß, IFN-?) also are involved in the expression of adhesion molecules. *PAF*, platelet-activating factor; *LTB $_4$* , leukotriene B $_4$; *FMLP*, *N*-formyl-methionyl leucyl phenylalanine.

The third group of molecules important in the binding of leukocytes to endothelial cells is the immunoglobulin superfamily of adhesion molecules. The most prominent members of this family, ICAM-1 and ICAM-2, [159] are expressed on endothelial cells in the basal state; ICAM-2 is expressed at higher levels than ICAM-1. The expression of ICAM-2 is not increased by cytokine stimulation, whereas ICAM-1 expression is enhanced by IL-1, TNF-a, and IFN-?. Thus, the relative importance of ICAM-1 increases in inflammation. ICAM-1 binds both CD11a/CD18 and CD11b/CD18, whereas ICAM-2 binds only CD11a/CD18.

Enhanced expression of adhesion molecules occurs in human diseases marked by gastrointestinal inflammation and in animal models of inflammation. In inflamed colonic mucosa, the expression of ICAM-1, E-selectin, and P-selectin is up-regulated in the vascular endothelium. [166], [167] and [168] Quantitative immunohistochemistry reveals increased numbers of E-selectin–positive vessels in inflamed areas of a colon affected by ulcerative colitis in comparison with the healthy colon. [169]

The role of adhesion molecules in the pathogenesis of intestinal inflammation was carefully addressed in a study of indomethacin-induced enteritis in the rat. [170] Indomethacin treatment results in mucosal ulceration and granulocyte infiltration in the rat intestine and a corresponding inflammatory response in the mesentery characterized by an increase in the number of adherent and extravascular leukocytes and a reduction in leukocyte rolling velocity. The role of adhesion molecules was assessed by the coadministration of indomethacin and monoclonal antibodies directed against P-selectin, E-selectin, or CD11b/CD18. The indomethacin-induced leukocyte–epithelial cell adhesion in mesenteric venules was reduced with coadministration of monoclonal antibodies against CD11b/CD18 or E-selectin but not by the monoclonal antibody against P-selectin. This study suggests that both CD11b/CD18 (expressed on neutrophils) and E-selectin (expressed on endothelial cells) contribute to neutrophil accumulation.

After neutrophils attach firmly to the endothelium, they migrate between the endothelial cells into the extravascular space. On neutrophils, L-selectin (LAM-1) and CD11b/CD18 are expressed consecutively—that is, the expression of CD11b/CD18 increases as LAM-1 is shed. Immunohistochemistry of inflamed tissue demonstrates that some intravascular neutrophils are LAM-1 positive and others are CD11b/CD18 positive, but extravascular neutrophils are all CD11b/CD18 positive and LAM-1 negative, [171] suggesting that both LAM-1 and CD11b/CD18 are important in the adhesion of neutrophils to endothelial cells but that only CD11b/CD18 is important in the subsequent neutrophil migration into the extravascular space.

The final step in leukocyte trafficking in intestinal inflammation is the passage of leukocytes between epithelial cells and out into the lumen. [172], [173] Leukocytes that have passed into the lumen can be found in the stool; indeed, the presence of fecal leukocytes signifies inflammation of the gastrointestinal tract. Adhesion molecules, including ICAM-1, are expressed on epithelial cells in the presence of inflammation, and neutrophil chemotactic factors have been identified in the colonic lumen.

Adhesion molecules are possible therapeutic targets in gastrointestinal inflammation. Antibodies to adhesion molecules, both neutrophil adhesion molecules and endothelial adhesion molecules, have been used to block leukocyte trafficking. Monoclonal antibodies directed against ICAM-1 prevent the passage of neutrophils between endothelial cells. [174] A set of experiments in which antibodies against different components of the ß $_2$-integrins were used defined their role in the migration of neutrophils between endothelial cells. [175], [176] Antibodies against CD18 inhibited neutrophil migration, and antibodies against CD11a or CD11b also inhibited migration. Therefore, the binding of both CD11a/CD18 and CD11b/CD18 to ICAM-1 appears to be important in the migration of neutrophils between endothelial cells. In contrast, antibodies to L-selectin did not inhibit neutrophil migration, which is consistent with the finding that extravascular neutrophils are CD11b/CD18 positive but L-selectin negative. Protein synthesis can be blocked by administering the appropriate antisense mRNA, which will bind to the sense mRNA and prevent translation. Antisense mRNA for ICAM-1 was given intravenously to a small group of patients with steroid-treated Crohn disease in an attempt to improve their clinical status by diminishing leukocyte trafficking. A better clinical outcome was observed in the patients receiving ICAM-1 antisense mRNA than in those receiving placebo. [177]

Lymphocyte trafficking shares some similarity with neutrophil and monocyte trafficking, but the differences are substantial. Lymphocyte trafficking is mediated by the expression of receptors and counterreceptors on lymphocytes and endothelial cells. The interaction of lymphocytes with endothelial cells (like the interaction of leukocytes with endothelial cells) involves three families of cell surface proteins: integrins, selectins, and immunoglobulin-like adhesion receptors. Lymphocytes destined for the intestine display some integrins (e.g., CD11a/CD18) that are also expressed on leukocytes, but they also express lymphocyte-specific integrins, including a ^{4}ß $_7$, an integrin expressed in CD4 and CD8 gut trophic lymphocytes, and a ^{E}ß $_7$, which is expressed on almost all intraepithelial lymphocytes and 40% of lamina propria lymphocytes. [178], [179] and [180]

The a ^{4}ß $_7$-integrin binds to MAdCAM-1, a member of the immunoglobulin superfamily that is selectively expressed on the high endothelial venules of mucosal lymphoid organs. [70] Thus, the migration of selected lymphocyte populations to the intestine is directed by the selective expression of a ^{4}ß $_7$ on these lymphocytes and the selective expression of MAdCAM-1 on certain endothelial populations.

The a ^{4}ß $_7$-integrin binds to E-cadherin on the basolateral surface of endothelial cells. This interaction directs the migration of intraepithelial lymphocytes to the epithelium after they leave the vascular space. [69], [116]

Selectins play a role in the interaction of lymphocytes with the endothelium, just as they do with leukocytes and the endothelium. By nature of the lectin-binding domain, all selectins bind fucosylated lactosamine structures, related to the sialylated Lewis X (sLe ^{X}) blood group antigens. The sLe ^{X} blood group antigens decorate molecules such as MadCAM-1, making them counterligands for L-selectin–bearing lymphocytes. E-selectin plays a role in the adhesion of a subpopulation of memory lymphocytes, neutrophils, and monocytes, and the concentration of E-selectin is increased in inflammatory lesions of the intestine. [181]

Regulated expression of these molecules controls the migration of naive lymphocytes into Peyer patches through high endothelial venules and the subsequent homing of activated memory lymphocytes into the effector compartments of the loosely affiliated lamina propria. Naive T lymphocytes that migrate into Peyer patches in search of an antigen encounter are CD3 $^+$, CD45RA $^+$, L-selectin $^+$, and a ^{4}ß $_7$ ^{Lo}; however, antigen-activated T lymphocytes that migrate into lamina propria are CD3 $^+$, CD45RO $^+$, L-selectin $^-$, and a ^{4}ß $_7$ ^{Hi}. Similarly, naive B cells with a phenotype of surface IgD $^+$, CD20 $^+$, L-selectin $^+$, and a ^{4}ß $_7$ ^{Lo}, and memory B cells with a phenotype of surface IgD $^-$, CD20 $^+$, L-selectin $^-$, and a ^{4}ß $_7$ ^{Hi} are preferentially observed in Peyer patches and lamina propria, respectively. [182]

LEUKOCYTE CHEMOTAXIS AND ACTIVATION

After the neutrophil has adhered to the endothelium and passed between the endothelial cells, it must migrate to the site of bacterial invasion or other injury, a process called *chemotaxis*. After entering the affected tissue, the neutrophil must acquire the ability to produce reactive oxygen species in maximal amounts. [183] The process of enhancing the capacity of the neutrophil for producing reactive oxygen species is called *priming,* and the stimulation of their production is called *activation*.

A chemoattractant is a molecule that stimulates the migration of neutrophils, monocytes, eosinophils, and other cells. Chemotaxis is the migration of a cell in the direction of the higher concentration of a chemoattractant. [184] Chemotactic agents act by binding to specific receptors on the leukocyte plasma membrane; binding of a ligand to its receptor results in intracellular signaling events, reorganization of the cytoskeleton, and movement of the leukocyte. Among the agents that induce neutrophil chemotaxis are fMLP, IL-8, the complement component C5a, PAF, and LTB $_4$; each chemotactic agent has a separate receptor on the neutrophil plasma membrane. The direction of neutrophil migration after the binding of a chemoattractant to its receptor is determined by the location of the receptor on the neutrophil plasma membrane. Receptors are distributed over the surface of the cells, and migration occurs in the direction of the receptor where ligand binding has occurred. Monocytes and eosinophils also are capable of chemotaxis, but they respond to a different range of agents.

Study of the chemotactic agents that affect neutrophils (e.g., fMLP, IL-8, C5a, PAF, and LTB $_4$) reveals ties between neutrophil chemotaxis and bacterial infection. The agents that induce neutrophil chemotaxis suggest that the inflammatory response developed to deal with bacterial infections. fMLP is a tripeptide derived from *Escherichia coli* and other bacteria [185]; binding of fMLP to its receptor results in neutrophil migration toward the bacteria. IL-8 is produced by a variety of cell types, including intestinal epithelial cells and activated monocytes. Intestinal epithelial cells produce IL-8 when infected with invasive bacterial species such as *Salmonella.* [186] IL-8 serves as an early warning system for host events by triggering the influx of neutrophils to sites of bacterial invasion in the intestine. C5a production is part of the immune response to bacterial invasion. [187] The binding of immunoglobulins to invading bacteria results in complement activation and the production of C5a, which attracts neutrophils to the site of the antibody-coated bacteria. PAF and LTB $_4$ are secondary amplification agents for the inflammatory response. Phagocytosis of invading bacteria activates neutrophils and macrophages, resulting in the production of PAF and LTB $_4$, which in turn induce the migration of other neutrophils and macrophages to the site of bacterial invasion. [188] Thus, each of the neutrophil chemoattractant receptors can be related, directly or indirectly, to neutrophil migration to sites of bacterial invasion. The multiplicity of mechanisms stimulating neutrophil migration toward sites of bacterial infection suggests the evolutionary importance of this response.

The adhesion of neutrophils and monocytes to endothelial cells and their migration to the site of inflammation are two highly integrated components of the inflammatory response. The regulation of these events is coordinated by cytokines and inflammatory mediators. A single biologic event, such as the phagocytosis of bacteria by a macrophage, results in the production of agents (e.g., IL-1) that induce the expression of adhesion molecules but not neutrophil chemotaxis, and other agents (e.g., IL-8 and LTB $_4$) that induce the expression of adhesion molecules and neutrophil chemotaxis. Some soluble mediators, such as eosinophil chemotactic factor, are chemotactic for only one cell type, whereas others act on multiple cell types; for example, LTB $_4$ and PAF are chemotactic for both monocytes and neutrophils. The mediators that induce the expression of adhesion molecules and leukocyte chemotaxis constitute a complex and highly redundant network. In chronic inflammatory diseases, multiple agents that induce leukocyte adhesion and chemotaxis act in parallel (Fig. 7-9). The redundancy in the network suggests that therapeutic agents aimed at one specific element in this complex network may not be successful in blocking the inflammatory response.

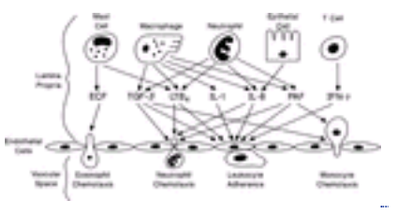

FIGURE 7-9. Chemotactic factors are part of a complex and redundant network. In the intestine, multiple cell types in the epithelium and lamina propria are capable of producing factors that promote the expression of adhesion molecules on leukocytes and endothelial cells or the chemotaxis of leukocytes. These factors include eosinophil chemotactic factor (ECF), transforming growth factor-ß (TGF-ß), leukotriene B_4 (LTB_4), IL-1, IL-8, platelet-activating factor (PAF), and interferon-? (IFN-?). Some inflammatory cells, especially macrophages and neutrophils, produce several of these chemotactic and adherence factors. Some factors act specifically on one cell type, whereas others act on multiple cell types. Some factors affect the expression of adhesion molecules and chemotaxis, whereas others affect only the expression of adhesion molecules.

The complexity of the network of chemotactic agents raises the question of whether all these factors operate in every gastrointestinal inflammatory event or whether some factors are prominent in certain inflammatory events but not in others. The question remains to be answered. The composition of the inflammatory infiltrate may yield some insight. For example, a large number of eosinophils suggests the presence of eosinophil chemotactic factors. In most inflammatory states, it is likely that more than one chemotactic factor is operative and that different factors are more or less prominent as the inflammatory response develops. In response to infectious agents, neutrophil infiltration typically occurs within a few hours and monocyte infiltration occurs later, suggesting that factors promoting neutrophil chemotaxis appear earlier in the development of inflammation than those promoting monocyte chemotaxis.

Circulating neutrophils are not capable of producing reactive oxygen species in maximal amounts. After entering the tissue, neutrophils are primed by IL-1, TNF-a, granulocyte-macrophage colony-stimulating factor (GM-CSF), or lipopolysaccharide, which enhance their ability to make reactive oxygen species. Neutrophils also can be primed by exposure to the extracellular matrix. Exposure to C5a, LTB_4, or fMLP can activate the neutrophil respiratory burst with the production of superoxide, singlet oxygen, and hydroxyl radical. More typically, activation of the respiratory burst in neutrophils is the result of the phagocytosis of bacteria. Neutrophils kill phagocytosed bacteria with reactive oxygen species generated intracellularly within the phagosome. [189] Although the generation of reactive oxygen species serves the useful purpose of killing bacteria, considerable evidence indicates that reactive oxygen species produced by neutrophils contribute to the tissue destruction seen in acute and chronic inflammatory diseases of the gastrointestinal tract, including inflammatory bowel disease. [190]

fMLP is a molecule of particular interest in considering neutrophil chemotaxis and activation in the gut. [185] *E coli* and other bacteria found in normal colons produce a series of formulated oligopeptides, of which fMLP is the prototype. These formulated oligopeptides bind to fMLP receptors on neutrophils and macrophages. The intact epithelial monolayer acts as a barrier to the passage of fMLP, preventing the activation of lamina propria neutrophils by fMLP. Diseases marked by defects in the epithelial monolayer result in the exposure of lamina propria neutrophils to fMLP and the recruitment of additional neutrophils to the area of epithelial damage. fMLP can cross the epithelial monolayer by paracellular pathways, [191] but in health, the epithelial monolayer usually blocks its passage. Increased paracellular permeability caused by drugs, disease states, or genetic disposition can enhance the passage of fMLP across the monolayer and thus enhance neutrophil recruitment. fMLP suppositories have been used to induce neutrophil migration into the colon in animal models of colitis, demonstrating that even in health, the colonic epithelial monolayer is not totally impermeable to fMLP.

Study of the neutrophil fMLP receptors has demonstrated how receptor-ligand interactions also regulate other neutrophil functional activities. Neutrophils have two classes of fMLP receptors: high-affinity receptors and low-affinity receptors. Binding of fMLP to the high-affinity receptor results in chemotaxis; binding to the low-affinity receptor activates the respiratory burst and other neutrophil functions involved in bacterial killing, suggesting a mechanism, mediated by high-affinity fMLP receptors, by which neutrophil migration is induced by low concentrations of a bacterial product at a considerable distance from the bacteria. As the neutrophils migrate toward the bacteria, the concentration of fMLP increases and low-affinity receptors are activated. Binding of fMLP to low-affinity receptors initiates a series of events, particularly activation of the enzymes required for producing reactive oxygen species that prepare the neutrophils for bacterial killing. The interaction of the chemotactic and neutrophil-activating components of fMLP activity was demonstrated by a study in which an fMLP solution was infused into the rat colon. This infusion induced neutrophil infiltration, increased blood flow, and increased endothelial and epithelial permeability. [185] The increase in epithelial permeability was reduced by administering antioxidants and by depleting neutrophils. The ability of antioxidants to block the effects of fMLP on permeability suggests that these effects are mediated by activation of the respiratory burst in neutrophils.

There are disease-specific chemotactic agents. Gastritis caused by *H pylori* is associated with the infiltration of neutrophils into the gastric mucosa. A neutrophil chemotactic agent secreted by *H pylori* has been cloned. [192] Whether this chemotactic factor binds to a previously described receptor for another chemotactic factor or to a novel receptor has not been established. The contribution of this *H pylori*–associated chemotactic factor to the total infiltration of neutrophils in *H pylori*–infected gastritis is not known.

Neutrophil migration has been studied more extensively in ulcerative colitis than in other gastrointestinal diseases. [193] Mucosa affected by ulcerative colitis has high levels of two major chemotactic factors, IL-8 and LTB_4. [194] IL-8 production by both macrophages and epithelial cells is induced by IL-1 and TNF-a, proinflammatory cytokines measured at increased levels in ulcerative colitis. LTB_4 is produced primarily by activated neutrophils, and the mucosa affected by ulcerative colitis is heavily infiltrated with activated neutrophils. The identity of the major neutrophil chemotactic factor in ulcerative colitis was pursued by placing homogenized colonic mucosa from human surgical resections in Boyden chambers and measuring neutrophil chemotaxis. [193] Homogenized mucosa from resections of tissue affected by ulcerative colitis induced a level of neutrophil chemotaxis that was several times that seen with normal mucosa. Most neutrophil chemotactic activity was lipid extractable. Administration of a specific antibody against LTB_4 neutralized about 60% of the total chemotactic activity and nearly all the lipid-extractable chemotactic activity. The data were interpreted to suggest that LTB_4 is the dominant, but not sole, neutrophil chemotactic factor in the mucosa affected by ulcerative colitis. Inhibitors of 5-lipoxygenase, the key enzyme in the synthesis of LTB_4, are effective in the treatment of animal models of colitis, such as acetic acid–or trinitrobenzenesulfonic acid–induced colitis. These animal studies support the hypothesis that LTB_4 is a major chemoattractant in ulcerative colitis and that the pharmacological modulation of LTB_4 levels may diminish neutrophil recruitment and inflammation; however, trials of an inhibitor of 5-lipoxygenase and an inhibitor of 5-lipoxygenase–activating protein (FLAP) in patients with acute ulcerative colitis did not demonstrate a significant difference in the clinical response between the treated and placebo groups. [195] LTB_4 levels were measured in rectal dialysates before and after treatment with the FLAP inhibitor. Pretreatment levels of LTB_4 in dialysates of patients with ulcerative colitis were several times the levels measured in healthy persons. A single dose of the FLAP inhibitor reduced levels of LTB_4 in dialysates by more than 95% for longer than 24 hours, achieving the biochemical goal of reducing LTB_4 levels without inducing a clinical response. Despite success in animal models, leukotriene synthesis inhibitors probably failed to treat ulcerative colitis in humans because inflammation had been present for weeks to months, whereas in the animal model trials, inflammation had been present for hours to days. The prolonged course of inflammation in ulcerative colitis may have allowed time for the development of a more complex and redundant network of chemotactic agents.

LIPID MEDIATORS OF INFLAMMATION

Prostaglandins, leukotrienes, and PAF are referred to collectively as *lipid mediators of inflammation*. In many circumstances, they are produced by the same cell types in response to similar stimuli. Phagocytosis in macrophages results in the production of prostaglandin E_2 (PGE_2), LTB_4, and PAF. Although the lipid mediators are structurally distinct, they have a number of overlapping biologic effects. PGE_2, LTB_4, and PAF enhance vascular permeability; LTB_4 and PAF activate neutrophils.

Prostaglandins and leukotrienes are the products of arachidonic acid metabolism and are referred to as *eicosanoids*. [196], [197] and [198] Prostaglandins are produced through the cyclooxygenase pathway, whereas leukotrienes are produced through the 5-lipoxygenase pathway (Fig. 7-10). Two distinct cyclooxygenases are involved in prostaglandin synthesis. COX-1 is a constitutive enzyme found in most mammalian cell types. In the gastrointestinal tract, COX-1 is expressed in lamina propria mononuclear cells, fibroblasts, muscle cells, epithelial cells, and vascular endothelial cells. The expression of COX-2 is induced by IL-1, TNF-a, and other proinflammatory cytokines. COX-2 expression can be induced in macrophages, fibroblasts, epithelial cells, and other cell types.

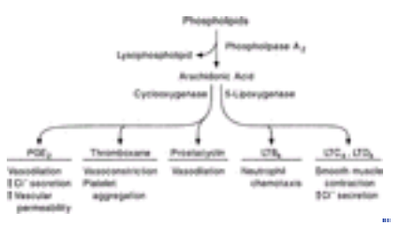

FIGURE 7-10. Arachidonic acid is metabolized to many proinflammatory products. Intracellular levels of arachidonic acid are quite low. The major rate-limiting step in the production of arachidonic acid metabolites is the release of arachidonic acid from phospholipids by the activation of phospholipase A_2. Most agents that induce the synthesis of arachidonic acid metabolites do so by activating phospholipase A_2. There are two major pathways for arachidonic acid metabolism: the cyclooxygenase pathway and a series of lipoxygenase pathways (5-, 12-, and 15-lipoxygenase); 5-lipoxygenase is the most prominent. Essentially all mammalian cells contain cyclooxygenase. The particular mixture of cyclooxygenase metabolites produced by a given cell is a function of the downstream enzymes (e.g., thromboxane synthase and prostacyclin synthase).

Under most circumstances, the rate-limiting step in the synthesis of prostaglandins is the availability of arachidonic acid. Cells have almost no free arachidonic acid; the activation of phospholipases (particularly phospholipase A_2) makes arachidonate available by releasing it from membrane phospholipids. A wide variety of stimuli can activate phospholipase A_2, resulting in the release of arachidonate. Phagocytosis in macrophages activates phospholipase A_2, as do receptor-mediated events, such as the binding of bradykinin to its receptor. Phospholipase activation typically results in the rapid release of arachidonate and the production of prostaglandins within a few seconds to 1 or 2 minutes. Prostaglandin synthesis also can be regulated by the infiltration of prostaglandin-producing cells into the gastrointestinal tract. The large increase in prostaglandin levels in inflammatory bowel disease may reflect the trafficking of prostaglandin-producing monocytes and macrophages into the lamina propria. A third mechanism for the regulation of prostaglandin production is the promotion of COX-1 and COX-2 synthesis. COX-2 synthesis can be induced by proinflammatory cytokines (e.g., IL-1, TNF-a) and phorbol esters. The induction of COX-2 expression occurs over a few hours; thus, the regulation of prostaglandin production by the induction of COX-2 occurs over a longer time frame than the regulation of prostaglandin production by phospholipase activation. Among the products of arachidonic acid metabolism through the COX pathway are PGE_2, thromboxane A_2, and prostacyclin. PGE_2 is made by macrophages, epithelial cells, and fibroblasts. Its biologic effects include vasodilation, increased epithelial Cl^- secretion, increased vascular permeability, and increased intestinal motility. PGE_2 also plays an important role in wound repair; in the face of injury, PGE_2 promotes epithelial cell proliferation. Thromboxane is produced primarily by platelets and results in vasoconstriction and platelet aggregation. Prostacyclin is produced by a number of cell types, including vascular endothelial cells. It is a potent vasodilator.

In the healthy gastrointestinal tract, COX-1 is present in abundance, but COX-2 is not expressed. [199] COX-2 expression has been identified in two animal models of gastrointestinal inflammation: acetic acid–induced gastric ulcers in the mouse and trinitrobenzenesulfonic acid–induced colitis in the rat. [200] The administration of selective COX-2 inhibitors in each of these models resulted in inhibition of spontaneous healing. These studies demonstrate that prostaglandins are involved in wound healing and suggest that the induction of COX-2 synthesis in gastrointestinal injury plays a role in enhancing wound healing. Prostaglandins play a role in down-regulating the immune response in the intestine. [201] A population of intestinal stromal cells constitutively express COX-2; these cells produce PGE_2, which is responsible for shutting down the immune response. [202]

Leukotrienes are made through the 5-lipoxygenase pathway. The cellular distribution of 5-lipoxygenase is much more limited than that of COX-1 and COX-2. LTB_4, a potent neutrophil chemotactic agent, is made in neutrophils, macrophages, and mast cells. The peptidyl leukotrienes (LTC_4, LTD_4, and LTE_4) are made in macrophages, mast cells, and eosinophils. They increase vascular permeability and induce vasoconstriction and smooth muscle contraction, and they may induce epithelial Cl^- secretion.

Inflamed mucosa from patients with ulcerative colitis and Crohn disease contains markedly elevated levels of PGE_2 and LTB_4. [203] Analysis of neutrophil chemotaxis in ulcerative colitis demonstrated LTB_4 to be the major neutrophil chemotactic factor; however, 5-lipoxygenase inhibitors, used as single therapeutic agents, have not been found to be effective in placebo-controlled trials in ulcerative colitis. Nonsteroidal antiinflammatory drugs (NSAIDs) inhibit prostaglandin production through COX-1 and COX-2. In many inflammatory diseases, NSAIDs diminish inflammation and relieve clinical symptoms, but in ulcerative colitis, NSAIDs exacerbate clinical activity. Elevated PGE_2 levels in ulcerative colitis are associated with enhanced COX-2 expression. Immunohistochemical studies demonstrated COX-2 expression in epithelial cells from inflamed tissue affected by ulcerative colitis. [204]

PAF is a phosphatidylcholine with an ether-linked alcohol fatty acid at the first carbon and an acyl-linked acetyl group at the second carbon. [205] There are no intracellular stores of PAF, and its synthesis is initiated by the remodeling of phosphatidylcholine through the activation of phospholipase A_2. PAF is made by neutrophils, macrophages, mast cells, and eosinophils, and it is often produced in parallel with prostaglandins and leukotrienes. The biologic effects of PAF include enhanced vascular permeability, vasoconstriction, platelet aggregation, neutrophil chemotaxis, smooth muscle contraction, and epithelial Cl^- secretion. Many of these biologic effects overlap with those of prostaglandins and leukotrienes. PAF is rapidly degraded to the inactive metabolite lyso-PAF by the enzyme acetylhydrolase. Acetylhydrolase has both intracellular and secreted forms. Intestinal epithelial cells secrete acetylhydrolase, which may be an important mechanism for the defense of the gastrointestinal tract against PAF. [206] Necrotizing enterocolitis is an often fatal disorder seen in premature infants; PAF may be an important mediator in its pathogenesis. An animal model of necrotizing enterocolitis was created by injecting PAF into the superior mesenteric artery of a rat. [207] Ischemia and necrosis in the small intestine resulted, a clinical picture similar to necrotizing enterocolitis in humans.

NITRIC OXIDE

Nitric oxide (NO) is a small, biologically active compound formed when nitric oxide synthase (NOS) oxidizes the guanidino nitrogen of arginine. The biologic effects of NO include actions as a vasodilator, a neurotransmitter, and an important component of the inflammatory response. [208], [209]

The three distinct isoforms of NOS differ in their cofactor requirements, tissue distribution, transcriptional regulation, and posttranslational modification. Type 1 NOS (NOS-1) is found in neural tissue. The nitric oxide produced by NOS-1 is the principal nonadrenergic noncholinergic (NANC) neurotransmitter in the gastrointestinal tract. NOS-1 is found in enteric nerves in the myenteric plexus [210] and circular muscular and is present in neurons containing vasoactive intestinal polypeptide (VIP). NO produced through NOS-1 participates in the control of peristalsis and sphincter function in the gut. Excess production of NO through NOS-1 may contribute to disorders of bowel motility, including ileus.

Type 3 NOS (NOS-3) is found in vascular endothelial cells. NO produced by endothelial cells relaxes vascular smooth muscle cells and dilates the vasculature. Inhibition of NOS-3 in vivo causes an increase in blood pressure; hypertension is the major side effect of nonspecific NOS inhibitors. The effects of NOS-3 inhibition on blood flow are organ specific. In rats, administration of L-NMMA, an NOS inhibitor, increases vascular resistance in the stomach, pancreas, and mesenteric bed but not in the colon or small intestine. [211] NOS-1 and NOS-3 synthesize nitric oxide in relatively small quantities; the physiological effects of nitric oxide as a vasodilator and a neurotransmitter occur at quite low concentrations. NOS-1 and NOS-3 are calcium/calmodulin-dependent enzymes.

In contrast to NOS-1 and NOS-3, which are constitutive, type 2 NOS (iNOS) is inducible by proinflammatory cytokines (e.g., IL-1, TNF-a, IFN-?) and by bacterial lipopolysaccharide. NOS-2 is also called *inducible NOS (iNOS)*. NOS-1 and NOS-3 form NO in picomole concentrations, whereas iNOS forms NO in nanomole concentrations. iNOS produces nitric oxide in quantities that greatly exceed those required for its physiological functions as a vasodilator or neurotransmitter. In macrophages, the large quantity of NO produced by iNOS is used in killing bacteria and tumor cells. [212] NO works in conjunction with reactive oxygen species (e.g., hydroxyl radical, superoxide, and hydrogen peroxide) generated within phagosomes to kill phagocytosed bacteria.

iNOS is induced in intestinal epithelial cells in inflammatory states. It is induced in epithelial cells in vitro by IL-1, TNF-a, IFN-?, and lipopolysaccharide [213]; in vivo administration of endotoxin to rats induces the expression of iNOS in enterocytes. [214], [215] Infection of IFN-?–primed intestinal epithelial cells with *Salmonella* induces iNOS expression. [216] Immunohistochemical studies have revealed induction of iNOS in colonic epithelial cells from areas of inflammation in surgical resections from patients with Crohn disease, with ulcerative colitis, and with diverticulitis. [217] iNOS was not seen in epithelial cells from healthy colon or in epithelial cells from uninvolved areas of surgical resections. Elevated levels of the proinflammatory cytokines IL-1 and TNF-a in the mucosa of patients with Crohn disease and ulcerative colitis may explain the expression of iNOS by epithelial cells in these diseases.

The functional role of epithelial cell–produced NO in mucosal inflammation is not clear. NO produced by epithelial cells dilates mucosal blood vessels; increased circulation acts to wash away noxious products of inflammation. Some evidence indicates that NO may help to maintain the integrity of the epithelial barrier and the normally low permeability of the enterocyte monolayer to macromolecule transit. In studies of ischemia-reperfusion in the cat intestine, inhibition of NO synthesis resulted in marked increases in microvascular permeability and intestinal epithelial permeability. [208] Some question exists as to whether the net effect of NO generated by iNOS is to enhance or diminish gastrointestinal damage in inflammatory states. Most evidence suggests that NO, especially at low to moderate levels of production, is protective; there is some evidence that NO at high levels may promote gastrointestinal damage. Administration of endotoxin to rats induces the

expression of iNOS in the intestine and promotes gastrointestinal injury. Pretreatment of rats with an NOS inhibitor before the administration of endotoxin preserves the viability of enterocytes and diminishes the inflammatory response. [208] Moreover, the gastrointestinal injury associated with the lumenal administration of trinitrobenzenesulfonic acid is prevented by the administration of NOS inhibitors (e.g., L-NAME and aminoguanidine). In contrast to the detrimental effects of high levels of NO on mucosal integrity, low levels may enhance mucosal integrity. In the rat stomach, coadministration of indomethacin and the NOS inhibitor L-NMMA induces gastric mucosal injury. [218]

EPITHELIAL CELLS

Epithelial cells interact with the inflammatory response at several levels. The secretion of electrolytes and water by epithelial cells is an important part of the gastrointestinal response to inflammation. The interaction of inflammation and epithelial cell electrolyte and water secretion has been studied by assessing the effects of individual inflammatory mediators on electrolyte and water secretion, or by identifying the mediators that regulate electrolyte and water secretion in specific inflammatory states. Histamine, PGE_2, serotonin (5-hydroxytryptamine, 5-HT), and LTB_4 are just a few of the inflammatory mediators that induce epithelial cell Cl^- secretion (Fig. 7-11). Neural mechanisms also affect epithelial cell Cl^- secretion directly or indirectly through inflammatory cells. The neurotransmitter acetylcholine induces Cl^- secretion in epithelial cells directly. [219] Neuropeptides such as substance P, VIP, and NPY induce mast cell activation, resulting in the release of histamine and 5-HT, and thus activate epithelial cell Cl^- secretion. [220] Cl^- secretion is accompanied by Na^+ secretion and, consequently, by the passage of water across the epithelium into the intestinal lumen. Diarrhea is the clinical manifestation of the enhanced enterocyte Cl^- secretion induced by these mediators. These same inflammatory mediators induce the secretion of mucus by goblet cells in the gastrointestinal tract. Diarrhea protects the host from infectious agents and their toxins by speeding their passage through the gastrointestinal tract and out of the organism. Mucus secretion protects the host from infectious agents in the gastrointestinal tract by preventing the binding of the infectious agents and their toxins to epithelial cells.

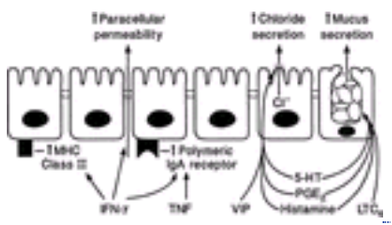

FIGURE 7-11. Intestinal epithelial cells respond to cytokines and inflammatory mediators. *5-HT,* 5-hydroxytryptamine, or serotonin; *IFN-?*, interferon-?; *LTC_4,* leukotriene C_4; *PGE_2,* prostaglandin E_2; *VIP,* vasoactive intestinal polypeptide.

The interaction of inflammation and epithelial electrolyte and water secretion has also been studied by identifying the inflammatory mediators that regulate electrolyte and water secretion in specific inflammatory conditions. One relatively simple animal model of intestinal inflammation is the sensitization of rats to egg albumin followed by antigenic challenge. In this model, antigenic challenge results in increased Cl^- secretion and increased paracellular permeability. [221] The increase in paracellular permeability was demonstrated by the increase in the uptake of ^{51}Cr-EDTA from the intestinal lumen after antigen challenge of previously sensitized rats. The relative contribution of various inflammatory mediators to the increase in Cl^- secretion seen in this model was tested with a series of blocking agents. Ketanserin (an antagonist of 5-HT), diphenhydramine (a histamine H_1 antagonist), and piroxicam (a COX inhibitor) inhibited egg albumin–induced Cl^- secretion by 30%, 42%, and 52%, respectively. The combination of piroxicam and diphenhydramine inhibited the secretory response by 82%. These data suggest that even in this simple model, more than one inflammatory mediator contributes to the increase in Cl^- secretion, and the mediators involved indicate that more than one cell type produces mediators. Histamine and 5-HT are products of mast cell activation. PGE_2, the prostaglandin most likely involved in Cl^- secretion, is produced by fibroblasts or by the epithelial cells themselves. In a similar study, challenge of *Trichinella*-immunized rats with *Trichinella* larvae resulted in increased intestinal fluid secretion. [222] Treatment of the rats with the combination of indomethacin, a COX inhibitor, and diphenhydramine ablated the increase in the fluid secretion induced by exposure to *Trichinella.* In the *Trichinella* model, increased fluid secretion is caused by a combination of prostaglandins and histamine, whereas in the egg albumin model, increased secretion results from a combination of prostaglandins, histamine, and 5-HT.

The epithelium also interacts with the inflammatory response in its function as a barrier. The epithelium separates lumenal antigens from lamina propria inflammatory cells, thus preventing activation of the inflammatory cells by lumenal agents. The effectiveness of the epithelium as a barrier is influenced by the junctions between the epithelial cells. [223], [224] The tighter these junctions, the more effective the epithelium is as a barrier to the passage of noxious agents. The junctions between epithelial cells are fairly loose in the proximal intestine and become progressively tighter in the distal small intestine and colon. Epithelial permeability usually is assessed by measuring the flux of tracer molecules either from the bloodstream into the intestinal lumen or from the intestinal lumen into the bloodstream. One commonly used probe is ^{51}Cr-EDTA. This compound is placed in the intestinal lumen, the animal is challenged, and then the recovery of ^{51}Cr-EDTA in the blood is assessed. Epithelial permeability is affected by inflammatory events. IFN-?, a cytokine produced during intestinal inflammation, increases paracellular permeability by opening the tight junctions between epithelial cells. [225] Increased paracellular permeability has been described in Crohn disease and in several animal models of gastrointestinal inflammation. [226]

Absorptive intestinal epithelial cells can also function as antigen-presenting cells. [227], [228], [229] and [230] Epithelial cells of the small intestine constitutively express MHC class II molecules, possibly as a consequence of the intraepithelial lymphocyte secretion of IFN-?. [231] Colonic epithelial cells do not normally express measurable levels of MHC class II molecules, except in the setting of inflammation, presumably in response to local cytokine production. [232] In vitro studies of intestinal epithelial cell function show that they take up, process, and present soluble antigens to $CD4^+$ T cells in the context of MHC class II molecules. [233] Although soluble antigens can be taken up apically and basolaterally, MHC class II molecules primarily segregate in the basolateral region, where antigen presentation to antigen-specific, MHC class II–restricted T cells occurs. There is evidence that under normal conditions, intestinal epithelial cells take up, process, and present soluble antigens from the lumen in vivo. The implication of these studies is that intestinal epithelial cells may augment or modify afferent pathways that normally result from antigenic events within Peyer patches. Despite in vitro evidence for functional class II MHC expression, intestinal epithelial cells seem to engage and stimulate preferentially $CD8^+$ cells that exhibit suppressor activity—an activity that may contribute to the suppressor tone of the intestine. [228], [229]

One consequence of many forms of intestinal injury is the development of breaks in the epithelial barrier. These breaks are seen in infectious disease, in inflammatory bowel disease, in celiac disease, and in injury caused by radiation and chemotherapeutic agents. There is an orchestrated response to these breaks, beginning with the rapid migration of epithelial cells shouldering the wound to cover the defect. Epithelial cells elongate and thus cover broad areas of denuded mucosal surface.

INFLAMMATION AND EPITHELIAL CELL GENE EXPRESSION

In the presence of inflammation, epithelial cells express a series of genes that are not expressed in the absence of inflammation. Among these are genes for COX-2, [204], [234] iNOS, [216], [217] and IL-8. [186] As previously discussed, an immunohistochemical study of human colon revealed expression of iNOS in epithelial cells from areas of inflammation in ulcerative colitis, Crohn disease, and diverticulitis, but not in epithelial cells from uninflamed areas of the same surgical resections. [217] In parallel studies of the cellular distribution of iNOS and COX-2 in ulcerative colitis resections, these proteins were expressed in exactly the same populations of epithelial cells in areas of inflammation. [204] This colocalization suggests a common regulatory mechanism for the expression of these genes.

Intestinal epithelial cell lines infected with *Salmonella* species and other invasive bacteria express IL-8 and COX-2, whereas uninfected cells or cells infected with noninvasive bacteria do not. [186], [234] Intestinal epithelial cell lines in culture were infected with *Salmonella dublin* and then stained for COX-2 expression and for intracellular *Salmonella*. The *Salmonella*-infected cells expressed COX-2, as did a few contiguous epithelial cells. In contrast, cells that were not infected themselves or were not adjacent to infected cells did not express COX-2. The expression of COX-2 in cells adjacent to an infected cell raises the possibility of a paracrine effect mediated by a soluble factor. *Salmonella* infection also induced epithelial cell COX-2 expression in human intestinal xenografts transplanted into mice with severe combined immunodeficiency disease (SCID), which demonstrates that infection from *Salmonella* induces COX-2 expression in normal intestinal epithelial cells as well

as in transformed epithelial cell lines. Infection of the colonic epithelial cell line HT-29 with *Salmonella* resulted in the expression of COX-2 and, as a consequence, a 50-fold increase in the production of PGE_2, a potent stimulus for Cl^- secretion in epithelial cells. Supernatants from *Salmonella*-infected epithelial cell lines increased Cl^- secretion by polarized intestinal epithelial cells mounted in an Ussing chamber. The demonstration that the PGE_2 in conditioned media from *Salmonella*-infected epithelial cells in culture can induce Cl^- secretion in vitro suggests that the diarrhea affecting humans infected with *Salmonella* may be the result of PGE_2 produced by COX-2 in *Salmonella*-infected epithelial cells. One of the protective responses to infection with enteric pathogens is an increase in electrolyte and water secretion to wash the pathogens out of the gastrointestinal tract. This study proposes that one mechanism for this protective response is the induction of epithelial cell COX-2 expression, which results in increased prostaglandin production and, thus, increased Cl^- secretion.

The genes for IL-8, iNOS, and COX-2 each have an NF-?B site in their promoters. NF-?B is an important transcription factor in the regulation of the synthesis of numerous inflammation-related proteins (e.g., TNF-a, IL-1, ICAM-1, E-selectin, IL-8, iNOS, and COX-2.) [235] NF-?B is a heterodimer that consists of p50 and p65 subunits. In unstimulated cells, NF-?B is bound to I?B-a, which is found in the cytoplasm. When cells are stimulated, I?B-a is phosphorylated and degraded, and NF-?B is released. The release of NF-?B from I?B-a allows NF-?B to enter the nucleus, where it binds to the promoter regions of target genes. NF-?B is activated by proinflammatory cytokines (e.g., TNF-a and IL-1), oxidants, phorbol esters, PAF, and lipopolysaccharide. Some of these agents (e.g., IL-1, TNF-a) are likely to be present in inflammatory states. The importance of NF-?B in the response to infection is demonstrated by the finding that mice lacking the p50 subunit of NF-?B are unable to clear *Listeria* species and other organisms effectively. [236]

Activation of NF-?B results in the parallel stimulation of a number of important genes involved in the inflammatory response. It is this parallel stimulation that probably accounts for the coexpression of iNOS and COX-2 in the same population of epithelial cells in ulcerative colitis. The presence of NF-?B response elements in the genes for E-selectin, ICAM, IL-8, and TNF-a allows for the coordinated expression of a series of proteins involved in the adhesion of neutrophils to epithelial cells, the migration of neutrophils from the vascular space into gastrointestinal tissue, and the activation of those neutrophils. IL-1, TNF-a, and NF-?B are involved in a cycle of activation that results in amplification of the inflammatory response. IL-1 and TNF-a both activate NF-?B; in turn, the synthesis of IL-1 and TNF-a is promoted by the binding of NF-?B to response elements in their promoters. This positive regulatory cycle amplifies and perpetuates the inflammatory response. Although NF-?B has received the most attention, other transcription factors are involved in the regulation of genes associated with inflammation. NF-IL6 sites are presented in the promoters of several of these genes. Preliminary studies suggest that NF-IL6 may be as important as NF-?B in the regulation of these genes. [237]

MAST CELLS

Mast cells are inflammatory cells with large granules containing preformed mediators of inflammation (e.g., histamine and 5-HT). [238] In response to stimulation, mast cells release these granules and produce newly formed non-granule–associated mediators (e.g., NO, PGD_2, PAF, and leukotrienes). In the healthy gastrointestinal tract, mast cells are found in the lamina propria, submucosa, and muscle layers and on the serosal surface. [238] More numerous and activated mast cells have been observed in the gastrointestinal mucosa of patients with helminthic infections, ulcerative colitis, Crohn disease, gastritis, and celiac sprue.

In rodents, the two distinct subpopulations of mast cells are connective tissue mast cells and mucosal mast cells. The connective tissue mast cells, such as those found in the rat peritoneum, have granules that contain 5-HT, heparin, and large amounts of histamine. They are capable of making PAF, NO, PGE_2, TNF-a, IL-1, IL-3, IL-4, IL-6, IL-10, and IFN-?. In contrast, rat mucosal mast cells have granules that contain 5-HT and small amounts of histamine. They are capable of making PAF, NO, PGE_2, leukotrienes, and TNF-a. Connective tissue mast cells and mucosal mast cells arise from a common progenitor cell. There appear to be two subpopulations of mast cells in humans, but the distinctions are not as clear as they are in rodents.

Mast cells can be activated by a variety of factors, but IgE-dependent antigen activation is the most common. Antigen-specific IgE binds to receptors on the surface of the mast cell through its Fc component. Exposure of the mast cell to an appropriate antigen results in cross-linking of the IgE molecules, which in turn results in activation of the mast cell. IgE-mediated mast cell activation is an effective defense mechanism against intestinal worms and other parasites. Parasite antigens cross-link IgE molecules on intestinal mast cells. Mast cell activation releases substances that promote intestinal motility and increase electrolyte and water secretion. Histamine, PGD_2, and peptidyl leukotrienes are released by activated mast cells; all enhance epithelial cell Cl^- secretion and promote intestinal motility. These physiological responses allow the infected host to wash the parasites out of the digestive tract. IgE-mediated mast cell activation is also important in allergic disorders. Pollen cross-links IgE molecules on mast cells in the nasal mucosa, causing allergic rhinitis. Food antigens activate mast cells in the gastrointestinal tract by similar mechanisms. There would appear to be no evolutionary advantage to being able to mount an allergic response to food antigens, but there would be an evolutionary advantage to being able to mount a response to gut parasites that would clear them from the gastrointestinal tract. It may be that food allergies represent the maladaptation of an inflammatory response designed to deal with intestinal parasites.

In addition to IgE-dependent antigen activation, mast cells also can be activated by the calcium ionophore A23187 and by the complement components C3a and C5a. Some subpopulations can be activated by substance P; substance P activation is of particular interest in that substance P is a neurotransmitter that can be released by neural activation in the gastrointestinal tract. Mast cell activation by substance P would provide a mechanism for the induction of intestinal inflammation by neural activation. There are both anatomic and functional interactions between mast cells and the enteric nervous system. In the rat, infection with the intestinal nematode *Nippostrongylus brasiliensis* results in mast cell hyperplasia. Immunohistochemical studies reveal that most of these mast cells are juxtaposed to enteric nerves. There are bidirectional interactions between nerves and mast cells. Neurotransmitters, particularly substance P, cause mast cell degranulation, and mast cells in turn release VIP, which can act as a neurotransmitter.

Mast cell activation plays an important role in various allergic reactions. Exposure to an antigen results in B-cell activation and the production of antigen-specific IgE, which binds to specific receptors on mast cells. Reexposure to the sensitizing antigen results in IgE cross-linking and mast cell activation. Earlier in this chapter, we described the results of studies in which sensitized rats were challenged with albumin; Cl^- secretion increased, as did paracellular permeability. A role for mast cell activation in this process was demonstrated by the finding that Cl^- secretion in response to egg albumin can be significantly diminished by the coadministration of diphenhydramine, an antagonist of the histamine H_1 receptor. [221] Mast cells are the dominant source of histamine in the gastrointestinal tract.

Additional evidence that mast cells are involved in the mediation of enhanced Cl^- secretion after antigen exposure comes from studies with the mast cell–deficient mouse (W-W^V). Antigen challenge in sensitized W-W^V mice results in a 70% decrease in Cl^- secretion in comparison with antigen challenge in wild-type littermates. [239] These data suggest that at least in this model, mast cell–derived histamine is an important mediator of epithelial cell Cl^- secretion. The Cl^- secretion measured in the mast cell–deficient mice (30% of that in the wild-type mice) could be inhibited with NSAIDs, suggesting that the prostaglandins that induce Cl^- secretion are not of mast cell origin.

MOTILITY

The musculature of the gastrointestinal tract is at one level an effector mechanism for the inflammatory response. One mechanism for eliminating pathogenic organisms is to enhance motility and push the offending organisms out of the gut. The cramps and diarrhea that accompany intestinal inflammation are in part the products of inflammation-induced alterations in gastrointestinal motility.

Altered motility has been documented both in animal models of inflammation and in chronic inflammatory diseases in humans. The intrarectal administration of acetic acid in the cat results in a biphasic change in motility in the sigmoid colon. [240] Three days after the administration of acetic acid, spontaneous motility in the colon decreases. This period corresponds to the peak of acute inflammation. Motility returns to normal within a week; later, as the inflammation continues to resolve, motility is increased over normal. In conditions marked by chronic inflammation, changes in motility are more complex. Active ulcerative colitis in humans is associated with diminished sigmoid activity, which returns to normal as the colitis goes into remission. In healthy subjects, there is a reflex increase in colonic motility after meals; in patients with ulcerative colitis, this reflex increase in motility is decreased. [241]

The changes in motility associated with inflammation do not require inflammation in the muscle itself. Superficial mucosal inflammation can result in dysmotility in the underlying muscle, even though the muscle is histologically normal. [242] Moreover, inflammation in one part of the gastrointestinal tract can result in dysmotility in remote parts; for example, colitis can be associated with gastric dysmotility.

Motility of the gastrointestinal tract is affected by inflammation at several levels; mediators of inflammation affect smooth muscle contraction by direct interaction with

the muscle cell, inflammatory cytokines affect the growth of smooth muscle, and inflammation affects muscle contraction indirectly through effects on the neural and hormonal elements that regulate smooth muscle contraction.

Some mediators of inflammation, including prostaglandins, PAF, leukotrienes, and serotonin, act directly on smooth muscle cells. The effects of mediators on muscle contraction can be specific to a species, a region of the gastrointestinal tract, or a muscle type. For example, PGE_2 contracts colonic longitudinal muscle but relaxes colonic circular muscle. A few studies have attempted to define which inflammatory mediators contribute to the altered motility seen in specific inflammatory conditions. Oral challenge of ovalbumin-sensitized rats results in a disruption of the normal migrating motor complex. [243] This effect can be reduced by treating the rats with an antagonist of the 5-HT_1 and 5-HT_2 receptors. These findings suggest a role for 5-HT in the dysmotility associated with anaphylaxis.

Inflammatory cytokines affect smooth muscle growth and regulate enzymes involved in stricture formation. [244] Chronic inflammation is associated with stricture formation. In Crohn disease, smooth muscle hypertrophy is found in the muscularis mucosae. [245] Smooth muscle hypertrophy also is seen in the intestines of rodents infected with *Trichinella spiralis* and *N brasiliensis*. The mechanism of this inflammation-associated smooth muscle hypertrophy is not clearly understood; however, the proinflammatory cytokine IL-1, which is present in abundance in acute and chronic inflammation, is mitogenic for smooth muscle cells. IL-1 activates collagenase and inhibits collagen synthesis in human intestinal smooth muscle cells. [246]

Inflammation influences smooth muscle function through its effects on the enteric nervous system and hormones. Crohn disease is associated with structural abnormalities of enteric nerves and smooth muscle even in unaffected areas of the gastrointestinal tract, [247] which may explain the findings of dysmotility in parts of the gastrointestinal tract remote from the site of inflammation. Intestinal inflammation also has been associated with changes in the levels of neuropeptides. Substance P levels are increased in ulcerative colitis. [248] Substance P may play a role in the increased sensibility of the rectum to distention in patients who have ulcerative colitis. [249] In an animal model of ileitis, inflammation increased the noncholinergic excitation induced by substance P. [250]

NO levels increase in inflammatory bowel disease and in animal models of inflammation. NO is thought to be the major inhibitory neurotransmitter in the intestine. Immunohistochemical studies suggest that in human inflammatory bowel disease, most of the inducible NO synthesis is expressed in epithelial cells. Whether NO produced in epithelial cells can influence contractions in the muscularis propria is not clear.

The interaction between the immune system and smooth muscle cells is bidirectional. Cytokines affect smooth muscle function, and smooth muscle cells can produce cytokines. In vitro incubation of rat intestinal smooth muscle cells with IL-1 results in the production of IL-6. The functional importance of in vivo cytokine production of smooth muscle cells is not clear.

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CHAPTER 8

James Madara and James M. Anderson

EPITHELIA: BIOLOGIC PRINCIPLES OF ORGANIZATION

All cavities within the alimentary tract, from the small ducts and acini of the pancreas to the gastric lumen, are lined by sheets of polarized epithelial cells. A common feature of all epithelia is their ability to create selective barriers and to accomplish vectorial transport. These properties are based on the structural polarity of individual cells, their mechanisms of cell-cell and cell-substrate interaction, and their higher-order organization with other cell types. This chapter reviews both the origins of cell polarity and cell interactions and examines how these interactions underlie the structural organization of the gut wall and the biology of gut epithelial barriers. Some aspects of epithelial transport are discussed to emphasize the associations among epithelial transport, barrier function, and structure, but more detailed reviews of active transport processes across specific epithelia are found in Chapter 13, Chapter 14, Chapter 15 and Chapter 16.

ORGANIZATION OF THE GUT WALL

Figure 8-1 depicts the relation of the epithelial layer to other components of the gut wall. Four principal layers exist: mucosa, submucosa, muscularis propria, and serosa or adventitia. The mucosa consists of the epithelium, an underlying layer of loose connective tissue carrying nerves and vessels (i.e., lamina propria), and a thin layer of smooth muscle (i.e., muscularis mucosae). The mucosa also contains an array of lymphocytes, mast cells, macrophages, and, in disease states, polymorphonuclear leukocytes, all of which may be capable of modulating epithelial function.

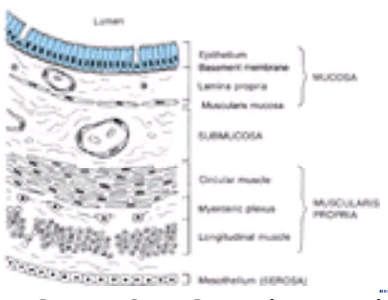

FIGURE 8-1. Generic organization of the gut wall.

An underlying layer of fibroconnective tissue called the *submucosa,* which contains nerves, vessels, and lymphatics, supports the mucosa. The submucosa rests on the muscularis propria, which is composed of two or three layers and is home to the myenteric plexus (see Chap. 2). In most instances, organs are encased by a delicate layer of fibrofatty tissue that supports a continuous layer of mesothelial cells called the *serosa.* Where no serosa exists, as in the esophagus, the fibrofatty tissues of surrounding structures interlace intimately with the external portion of the muscularis propria. These organs are said to have an adventitial, rather than a serosal, encasement.

Grasp biopsies retrieved endoscopically usually go no deeper than the muscularis mucosae, although thin wisps of submucosal tissues occasionally may be seen. Suction biopsies more consistently penetrate the submucosa, although only the most superficial portion of the submucosa is obtained. Deeper portions of the wall appear in endoscopic samples by accident, such as in an aggressive snare of a sessile mucosal lesion.

Organ-specific specializations in epithelial structure exist. Most of the gastrointestinal tract is lined by a simple columnar epithelium (Fig. 8-2; see Fig. 8-1). In contrast, the mouth and esophagus are lined by a nonkeratinizing squamous epithelium that is capable of withstanding the mechanical stress of swallowing but plays no role in transepithelial transport. Hepatocytes are unique, because the apical (canalicular) surface is organized not at one end but as a 3-μm band circumscribing the cell. The liver also lacks extensive subepithelial layers, and capillaries are separated from hepatocytes by only a rudimentary basement membrane in the space of Disse. The folding of epithelial layers also exhibits significant interorgan variation, such as the cuplike acinar organization of the exocrine pancreas and the deep extensions of the epithelium into the lamina propria of Brunner glands in the duodenum. Certain central features, however, are maintained in all epithelial tissues.

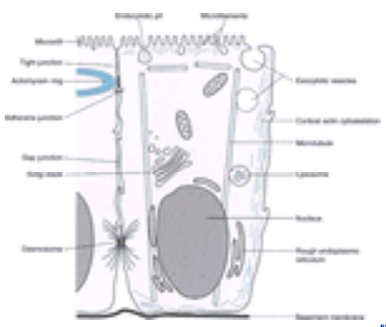

FIGURE 8-2. Generic organization of a gut epithelial cell.

ORGANIZATION OF EPITHELIAL CELLS AND SHEETS

To function properly as a barrier, epithelial cells must assemble into a multicellular sheet (see Fig. 8-1 and Fig. 8-2). Requirements for this higher organization include the following: mechanisms to generate individual cell polarity, cell-cell adhesion, and formation of cell-cell junctions; stabilization of cell shape by the cytoskeleton; and stabilizing interactions with the basement membrane.

Generation of Cell Polarity

A hallmark of epithelial cells is structural and functional polarity. Within each cell, the organelles and components of the secretory and endocytic pathways have specific spatial relationships (Fig. 8-3; see Fig. 8-2). These are largely maintained by attachment to cytoskeletal elements formed by filaments of actin and tubulin. Both microfilaments and microtubules are structurally polarized, with so-called plus and minus ends, and the cell uses this polarity to establish and maintain subcellular asymmetry. The plasma membrane is separated into apical and basolateral domains, and within these there exist microdomains that organize protein complexes of receptors, transporters, signal transduction proteins, and cell junctions. [1] Two examples make obvious the functional requirement for polarity: the sodium (Na^+)-dependent solute transporters must be positioned on the apical cell membrane to accomplish nutrient uptake from the gut lumen; and zymogen granules must be secreted onto the apical pole of the pancreatic acinar cell to reach the duodenum (see Fig. 8-3). Failure of zymogen granule targeting results in intracellular activation of proteases and pancreatitis. [2] The polarized distribution of surface components is maintained by protein sorting by the intracellular secretory machinery, by selective retention at the plasma membrane, and by the intramembrane fence function of the tight junction (see Fig. 8-3).

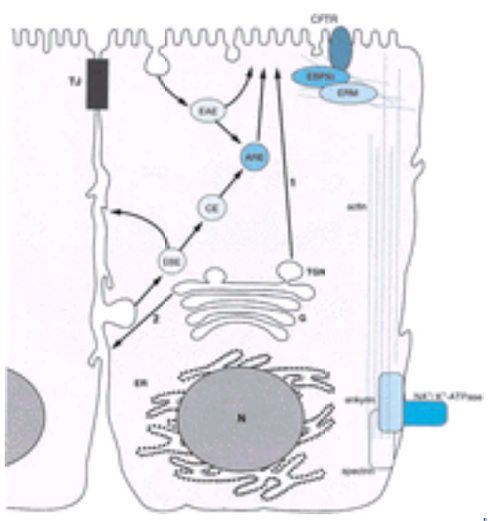

FIGURE 8-3. Three mechanisms for generating epithelial cell polarity. **First:** Membrane and secretory proteins synthesized in the endoplasmic reticulum (ER) pass through the Golgi apparatus (G) and are initially sorted in the trans-Golgi network (TGN). Some proteins are delivered directly to the apical (1) or basolateral (2) surface. Others (e.g., poly-IgA receptor) are delivered to the basal surface and are transcytosed to the apical surface through early basolateral endosome vesicles (EBE). Many receptors are recycled on their respective membranes through early apical endosomes (EAE) or EBE. Crossover between the apical and basal compartments and maintaining fidelity of each can occur through apical recyling endosomes (ARE) and common endosomes (CE). **Second:** Once inserted, many proteins are anchored to the cortical actin cytoskeleton;for example, the Na^+,K^+-ATPase is bound through spectrin and ankyrin; cystic fibrosis transmembrane conductance regulator (CFTR) is bound through EBP50 (ezrin-radixin-moesin family of proteins [ERM]–binding phosphoprotein of 50 kd) and ezrin. **Third:** Tight junctions (TJ) create a barrier to diffusion of components between the membrane domains.

Extracellular Cues Induce Cell Polarity

The spatial cues that induce initial cell asymmetry, and maintain it, arise from contact with the extracellular matrix and with other cells. Important receptors include the large protein families of integrins, heparan sulfate proteoglycans (e.g., syndecan), and others. [3], [4] These respond to ligation of matrix components such as collagens, fibronectin, and laminin by inducing initial polarity of the cytoskeleton, organelles, and intracellular vesicular compartments. These cues also alter or maintain differentiation by controlling gene transcription.

A key transducer of extracellular cues in gastrointestinal epithelia is the cell-cell adhesion molecule, cadherin. The epithelial form, E-cadherin, is a 120-kd calcium (Ca^{2+})-dependent homotypic adhesion molecule found in all epithelial cell types of the gastrointestinal tract (Table 8-1; see Fig. 8-2). [5] In many cell types, E-cadherin is focused at the adherens junction where, through the cytoplasmic linker proteins, a-actinin, and a- and ß-catenin, it is coupled to a perijunctional ring of actin filaments (see Fig. 8-2 and Table 8-1). E-cadherin is also capable of heterotypic adhesion to a $E ß_7$ integrin on intraepithelial lymphocytes. [6] Many experiments support the idea that cell-cell adhesion mediated by cadherin triggers a cascade of cellular events that induces polarity. In addition, cadherin influences gene transcription and inhibits cell motility. [7] This explains why loss of cadherin is strongly correlated with acquisition of an invasive phenotype for many human gastrointestinal neoplasms. Another link to cell proliferation occurs through ß-catenin, which, when not bound to cadherin, functions as pro-growth transcription activator. The activity of cytosolic ß-catenin normally is down-regulated by the adenomatous polyposis coli (APC) gene, thus explaining why mutations in *APC* lead to enhanced ß-catenin activity and growth of adenomas in the colon and demonstrating the functional connections among cell adhesion, polarity, growth, and tumor invasion. [8], [9] Likewise, altered expressions of other cell-cell and cell-matrix molecules are either primary or secondary causes of neoplastic transformation in the gastrointestinal tract, again because of their inductive role in maintaining polarity and the differentiated phenotype.

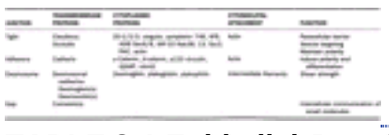

TABLE 8-1 Epithelial Junctions: Molecular Components and Function

Polarized Delivery of Proteins to the Cell Surface

Plasma membrane proteins pass through a series of distinct vesicular compartments as they are sorted to either the apical or basolateral surface. They share a common site of synthesis on ribosomes bound to the rough endoplasmic reticulum, undergo posttranslational modification in the Golgi apparatus (e.g., glycosylation), and are first sorted into distinct vesicles in the trans-Golgi network (see Fig. 8-3). From this point, the patterns of vesicle sorting are specific to different cell types. In hepatocytes, all membrane proteins are first delivered to the basolateral surface; subsequently, the apically destined proteins are “transcytosed” to the canalicular surface. [10] An easily rationalized example of this type of sorting is offered by the poly-immunoglobulin A (IgA) receptor, which picks up IgA from the serum on the basal surface and is then transcytosed and released as secretory component into the bile. [10] Why the apical transporters and channels first traffic through the basal membrane in hepatocytes remains unclear.

Intestinal epithelial cells appear to use a combination of both indirect (transcytosis) and direct sorting for delivery of polarized membrane proteins. [11] In addition, all cell types have active pools of vesicle compartments that recycle proteins on either the apical or basolateral surfaces, called, respectively, *early apical or basolateral*

recycling endosomes (see Fig. 8-3). These are used by receptors cyclically to internalize compounds such as low-density lipoprotein (LDL) and transferrin after being reinserted back onto the correct surface. Proteins in these early endosome compartments can also cross among trafficking pathways in at last two compartments located deeper in the cell, called the *common endosome* and the *apical recycling endosome.* For example, the poly-IgA receptor is transcytosed from the basal to the apical surface through early basal endosomes and the common and apical recycling endosomes. Degradation of proteins from both surfaces is accompanied by sorting early endosomes to a common late endosome and into lysosomes. The polarized sorting of lipid is discussed in Chapter 18.

Signals must exist for sorting proteins among different compartments and for keeping the various compartments biochemically distinct. Emerging themes for maintaining specificity are the presence of distinct members of the Rab family of small guanosine triphosphate (GTP)-binding G proteins on each vesicle type and the presence of different adaptor proteins coating the surface of each compartment. The specificity for recognizing and fusing with the proper surface domain is based on distinct members of the syntaxin, SNAP, and Rab protein families in the vesicles and plasma membrane; these mechanisms are similar to those responsible for synaptic vesicle cycling in neurons, [12] where they are better understood. Although in general these mechanisms remain poorly understood in gastrointestinal epithelia, two excellent examples are the machinery used by zymogen granules for recognition and fusion with the acinar cell membrane [13] and that used for the fusion of membranes containing hydrogen, potassium-adenosine triphosphatase (H^+,K^+-ATPase) with the apical surface of the parietal cell in the stomach. [14]

Initial sorting of proteins to either apical or basolateral surfaces is based on signals within the proteins themselves. Apical sorting signals usually are located in the extracellular or transmembrane domains. These signals can be specific carbohydrate moieties added, like a Zip code, while the protein passes through the Golgi apparatus. [15] Addition of the lipid glycosylphosatidylinositol (GPI), such as found on alkaline phosphatase, also creates an apical targeting signal. The lipid portion of the GPI anchor partitions into cholesterol-rich lipid microdomains, called *rafts,* that somehow are sorted to the apical surface. [16] At present, three types of basolateral sorting signals are recognized:

1. The so-called tyrosine-based motif exemplified by the LDL receptor (tyrosine-X-X-[hydrophobic residue])
2. The di-leucine sequence found on the immunoglobulin Fc receptor
3. A variety of unique motifs.

The sorting machinery that reads these signals is based on a large and diverse family of heterotetrameric adapter proteins (AP1, 2, 3, and 4). For example, the µ1B subunit of AP1 binds the LDL tyrosine motif, and this interaction is required for basolateral delivery of the receptor. [17] How the AP complexes themselves attain specific subcellular locations remains unknown.

Stabilization at the Membrane Surface

Once proteins are delivered to the correct plasma membrane domain, they can be retained through interaction with a cortical network of actin-based cytoskeletal proteins (see Fig. 8-3). For example, the Na^+,K^+-ATPase is stabilized on the basolateral membrane domain by attachment to the cytoskeleton through ankyrin and spectrin. [18] More recent is the recognition that many transmembrane proteins are linked through their extreme C-terminals to so-called PDZ domains within cortical cytoskeletal proteins. PDZ domains bind in a sequence-specific manner to the C-terminal three to seven amino acids of transmembrane proteins. Proteins with PDZ domains typically contain many additional protein-binding motifs that serve to cluster other components involved in regulating a specific channel or receptor signaling pathway. A good example is provided by cystic fibrosis transmembrane regulator (CFTR), which apart from its chloride (Cl^-)-secretory function also regulates electrolyte absorption by influencing the activity of the epithelial Na^+ channel in absorptive epithelial cells such as in the colon. The tail of CFTR is bound to a PDZ domain in a protein called EBP50 (ERM-binding phosphoprotein of 50 kd), which links it to the cortical actin cytoskeleton through an ERM (ezrin-radixin-moesin) protein (see Fig 8-3) and several signaling proteins including protein kinase A, which directly regulates the epithelial Na^+ channel. [19] There is growing interest in the study of cortical cytoskeletal coordination and scaffolding of epithelial cell signaling in the gastrointestinal tract. [1]

Stabilization of Cell Shape by the Cytoskeleton

The cytoskeleton is considered here in the context of the villus absorptive cell of the small intestine, a cell type that has become an important model for studies of cytoskeletal structure and function in nonmuscle cells. Interorgan differences in cytoskeletal organization largely represent variations on a single theme.

Stabilization of epithelial cell structure requires support for the apex of the cell, which confronts the turbulent environment of the gut lumen. Alimentary tract epithelia, except for esophageal squamous epithelia, display microvillus projections on the apical surface. Microvilli are particularly well formed in absorptive cells. A bundle of 20 to 30 microfilaments composed of F-actin resides in the core of these microvilli. [20]

Individual microfilaments are cross-linked to each other by filamentous structures consisting of filamin, a 68-kd actin-bundling protein (with two actin-binding domains that permit tight actin filament bundling), and villin, a 95-kd protein that can bundle or sever F-actin microfilaments, depending on intracellular Ca^{2+} concentrations. Villin knock-out mice demonstrate impaired intestinal epithelial wound healing that appears to result from the inability of microvilli to break down, which is a requirement for the transition from the epithelial to mesenchymal phenotype during wound closure. [21] The microfilament core is linked laterally to the microvillus plasma membrane by a member of the myosin family (myosin 1A). The membrane-myosin-actin link determines the length of the microvillus. The microvillus actin-bundle rootlets jut into the apical pole of the cell and associate with a terminal web of structural proteins. The density of this terminal web is most marked in cell types that do not have substantial exocytosis of secretory vesicles. Fine filaments, composed of type II myosin, link adjacent rootlets. A heteromeric spectrin-like molecule (i.e., TW260/240) also resides at this location and may be important in linking actin filaments and plasma membranes and in assisting in the stabilization of the cell surface.

Columnar epithelial cells display circumferential belts of actin and myosin II around their apices (see Fig. 8-2) that can contract in the presence of micromolar concentrations of Ca^{2+} and millimolar concentrations of ATP. [20] This actomyosin belt has been referred to as the *contractile ring.* The tension exerted by this ring on cell-cell contacts may modulate the barrier function of epithelia (discussed later in this chapter). A cortex of cytoskeletal elements, including actin and numerous cross-linking molecules, also wraps the basolateral pole on the epithelial cell. This cortex plays a crucial role in establishing apical-basolateral polarity, cell junctions, and transmembrane signaling [18] (see Fig. 8-2).

Cables composed of aggregates of 10-nm intermediate filaments course through the cells and function as support cables for structural buttressing. Such tonofilament cables associate with plasma membranes at sites where specialized cell-cell junctions, called *desmosomes,* reside (see Fig. 8-2). The role of desmosome and intermediate filaments in the gastrointestinal tract is highlighted by hepatocyte fragility in mice genetically altered to express a mutant form of keratin-18. These mice also have an increased susceptibility to the hepatotoxins acetaminophen and griseofulvin. [22]

The 25-nm-diameter microtubules, composed of tubulin, create a polarized array throughout the cell. Two large families of microtubule-dependent motor proteins exist, the kinesins and dyneins, which move in the opposite direction on polarized microtubule arrays. Most organelles and all the intracellular sorting compartments are attached to the microtubule network through adaptor proteins and use the motor proteins for trafficking and to maintain their proper subcellular localization. For example, if microtubules are disrupted, intestinal epithelial cells misdirect the apical membrane protein aminopeptidase to the basolateral domain

Intercellular Junctions

Other than the squamous epithelium of the esophagus and perhaps a few enteroendocrine cells, all other alimentary epithelia share a set of distinct intercellular junctions (see Fig. 8-2 and Table 8-1). These include, from the apical direction, the tight and adherens junctions, which form continuous circumferential contacts, and below these, desmosomes and gap junctions, which form macular or spot contacts. Together they seal the paracellular space, provide communication among cells, and stabilize the epithelial monolayer.

In thin sections viewed under the electron microscope, tight junctions appear as 100- to 300-nm-deep zones where adjacent cells closely abut (Fig. 8-4A). Series of punctate fusions or “kisses” between these plasma membranes form a sealing barrier. These fusion sites are arrayed in a linear anastomosing fashion around the cell and correspond to the netlike series of grooves or strands seen in replicated fracture faces of epithelial cells (Fig. 8-4B). The strands are formed by members of the claudin family of tetraspan adhesion molecules, which make contact in the intercellular space to create a selective diffusion barrier. Occludin, a 60-kd transmembrane protein whose function remains unknown, is also found in the strands of some tight junctions. In the intestine, occludin also is expressed diffusely along the entire

lateral cell membrane. Research has shown that occludin knock-out mice are viable. [23]

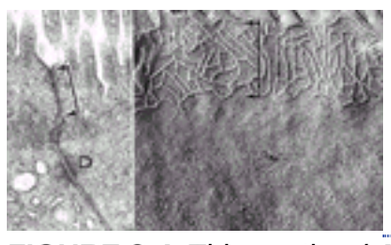

FIGURE 8-4. Thin section (**A**) and freeze fracture electron micrographic appearance (**B**) of the tight junction of an intestinal absorptive cell. In thin sections, the tight junction is a zone of close apposition between the lateral membranes of adjacent cells. By freeze fracture, the strands represent membrane fusion sites within the tight junction zone. Members of the claudin family of transmembrane proteins form the strands.

The cytoplasmic face of tight junctions contains a large number of proteins that fall into different functional categories. Some link the barrier-forming proteins to the actin cytoskeleton, others play a role in creating cell polarity, some form a specialized zone for vesicle targeting, and still others influence gene transcription (see Table 8-1). Many reviews of this complex structure are available. [24], [25] Of special interest is the direct physical link of the claudins to the perijunctional ring of actin and myosin through a PDZ protein called ZO-1, [26] which may play a role in cytoplasmic control of the paracellular barrier. The tight junction barrier is dynamic, and there are many other intracellular and extracellular signals that regulate barrier function under physiological and pathological conditions. [24], [27]

The squamous epithelium of the esophagus is somewhat peculiar. Unlike other alimentary epithelia, the epithelium lining the esophagus is multilayered (i.e., stratified). Data suggest that most layers are relatively devoid of tight junctions, and, where tight junctions do exist, they are discontinuous rather than circumferential. The intercellular barrier appears to be created by secreting sheets of a lipidlike material. [28]

Directly below the tight junction lies a zone, the adherens (or intermediate) junction, in which the lateral membranes of adjacent cells lie parallel to each other and adhere by means of E-cadherin–based interactions (see Fig. 8-2). At this site, the previously mentioned perijunctional contractile ring of actin and myosin resides and attaches to E-cadherin through a-actinin, vinculin, and a- and ß-catenin. [9] Directly below the adherens junctions are desmosomes (see Fig. 8-2 and Table 8-1). Distant relatives of the cadherin family, called *desmogleins* and *desmocollins,* form cell contacts at desmosomes. [29] In contrast to E-cadherin, these transmembrane glycoproteins have evolved different cytoplasmic sequences allowing them to associate with intermediate filaments rather than with actin microfilaments. Consequently, they anchor the cytokeratin-based cytoskeleton between neighboring cells and provide resistance to mechanical shear.

To varying degrees, all epithelial cells of the gastrointestinal tract express gap junctions. Here, the cytoplasm of adjacent cells is in physical continuity through transmembrane channels formed by members of the connexin protein family. Six connexins assemble on each membrane to form a channel and, by adhering across the paracellular space, create a lumen isolated from the extracellular space. Signaling molecules up to about 1500 d (e.g., Ca^{++}, inositol triphosphate) and small nucleotides can diffuse freely between cells and coordinate physiological responses. [30] There are numerous connexin genes in humans, and each shows organ-specific diversity and allows for organ-specific regulation of communication. Gap junctions coordinate epithelial function by allowing sheets of cells to behave as syncytia, for example, coordinating exocytosis of zymogen granules from the pancreas. [13] Gap junction communication in the liver, working through intracellular Ca^{++} waves, modifies bile secretion in response to glucagon and vasopressin. [31] Gap junctions are down-regulated in gastrointestinal cancer, probably reflecting the indifference to extracellular control that is characteristic of the undifferentiated phenotype. [32]

Basement Membranes

The basement membrane was introduced previously as a source of signals for inducing epithelial cell polarity. It also serves an important structural and supportive role. All alimentary epithelia reside on a basement membrane that measures 20 to 40 nm deep; consists of a faintly fibrillar network; and rests on an underlying, complex extracellular matrix. Although the basement membrane in the alimentary tract is incompletely characterized, it appears to be similar to basement membranes found elsewhere. [33] The major elements of the basement membrane include laminin, heparan sulfate proteoglycan, and type IV collagen; minor but possibly important constituents such as thrombospondin and entactin also exist.

Laminin, a 850-kd glycoprotein, consists of an assembly of three chains: a straight A chain and two B chains, which helically wrap a portion of the A chain and flare off at an acute angle. This arrangement imparts a cross shape on rotary-shadowed images of this molecule. Laminin exhibits specific binding sites for type IV collagen, heparan sulfate proteoglycan, cell surface laminin receptors, and entactin. [33] In similar fashion, many matrix components display binding sites for several additional components, thus adding to the complexity of interactions between the epithelial cell and its surrounding environment.

The major proteoglycan of the basement membane is heparan sulfate proteoglycan. Proteoglycans consist of long chains of glycosaminoglycans (e.g., heparan sulfate glycosaminoglycans) linked to a protein core. The structure of these massive molecules (i.e., molecular weight in excess of 10^{6}) is often likened to a test-tube brush, with the bristles representing the glycosaminoglycan extension. Proteoglycans probably organize water within the basement membrane. They hydrate this environment through their capacity to bind water, and they may be able to impart solute-sieving characteristics under conditions of bulk water flow.

Type IV collagen originates as a triple-stranded helical molecule, which, unlike other collagens, does not have its propeptides sheared from it after deposition in the extracellular space. Partially as a result of this, collagen IV does not cross-link into dense fibrils, as many other collagen species do; instead, it assumes a loose, netlike structure by associating with other collagen IV molecules. This meshlike structure of collagen IV may provide the basic structure to the basement membrane.

In addition to the specific components of the basement membrane mentioned previously, other key elements are present in the remainder of the extracellular matrix. For example, the adhesive glycoprotein fibronectin exists in soluble plasma and cell surface forms, and it is found in a highly insoluble fibrillar form in the extracellular matrix. Fibronectin has binding sites for heparan sulfate proteoglycan, collagens, and epithelial cell surface receptors and serves as one link between the cell surface and the matrix underlying the basement membrane.

Basement membrane components can exert significant effects on epithelia, including effects on proliferation, adhesion, migration, differentiation, and perhaps even barrier function. [33], [34] In the intestine, type IV collagen is produced primarily by mesenchymal components, heparan sulfate proteoglycan by epithelial cells, and laminin by mesenchymal and epithelial cells. Many components of basement membranes have multiple binding sites. For example, laminin can bind type IV collagen and heparan sulfate proteoglycan, but it also has a cell binding site.

Basement membrane components bind to a family of epithelial cell surface molecules called *integrins,* which bind to the actin cytoskeleton within the cell by way of linking proteins. Through such associations, structural elements within the cell are able to connect with, and potentially be influenced by, events occurring within the basement membrane and even deeper in the extracellular matrix.

EPITHELIAL BARRIERS

This section addresses the question of how intralumenal threats are restricted to the lumen by the epithelial barrier (Fig. 8-5). At various sites in the alimentary tract reside such threats as H^{+}, chemotactic peptides, undigested potentially antigenic proteins, and bacteria. It is not surprising that the epithelial barrier consists of numerous components; some are site specific and some are universal. To discuss these barriers, we arbitrarily divide them into two major categories: those that are extrinsic to the epithelium (although in some instances produced by the epithelium) and those provided by the physical presence of the epithelium, which we arbitrarily categorize as intrinsic barriers.

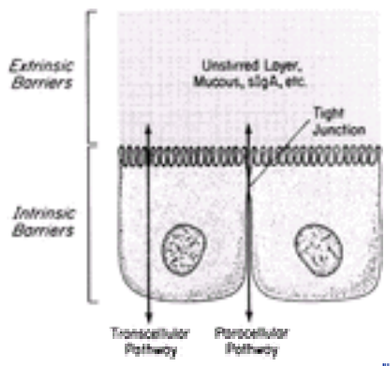

FIGURE 8-5. Gut epithelial barriers.

Extrinsic Barriers

Extrinsic barriers confront the microenvironment overlying the epithelia.

Mucus All alimentary epithelia are coated with a layer of mucus, which protects against bacteria and surface shear. Most surfaces, including those of the stomach, the intestine, the biliary and pancreatic ducts, and the gallbladder, contain specialized cell types that synthesize, package, and secrete mucin. In the esophagus, however, mucin is derived primarily from small glands that lie under the epithelium and connect to the lumen by way of delicate ducts. Although the precise chemical nature of mucus varies throughout the alimentary tract, the various mucin molecules share common features. They are viscous, polydispersed glycoproteins (250–20,000 kd) composed of about 80% carbohydrate by mass. At least eight human mucin-producing genes (*MUC* genes) have been identified. *MUC2* is the predominant form in intestinal and colonic surfaces; [35] its level is altered in inflammatory bowel disease and cancer. Mucin can act as a barrier by behaving as a viscous hydrated gel that undoubtedly attenuates shear forces that the epithelium otherwise would experience from particulates; these are driven down the alimentary tract by propulsive force. In addition, carbohydrate groups on the mucin molecules have the potential for binding to bacterial surfaces and inhibiting surface colonization. In some instances, mucin carbohydrates specifically replicate epithelial carbohydrate binding sites to which bacteria can attach, presumably preventing colonization in a more specific fashion. Given their extensive glycosylation, mucins can cross-link several bacteria and therefore serve to aggregate bacteria. Such aggregation presumably aids in bacterial clearance by intestinal propulsive activity. The protective effects of mucin are highlighted further by the finding that exposure of epithelial surfaces to threats such as bacterial toxins and noxious chemicals often results in a reflexive secretory release of mucins. The expression of mucin genes in goblet cells and mucin secretion respond to intestinal microbes and host-derived inflammatory mediators [36] and are altered by infections, such as *Helicobacter pylori* in the stomach. [37] It has been observed that the diffusion coefficients of hydrophilic molecules are substantially lower in mucin than in free solution. Some researchers have suggested that this alteration would diminish contact between the epithelial surface and lumenal threats such as H^+. Given the depth of the mucin layer and the duration that lumenal contents are in contact with the epithelium, however, small lumenal molecules probably have sufficient time to equilibrate within the mucous gel.

Unstirred Layer Because of the propulsive movements of the alimentary tract, the lumen can be rather turbulent. A body of evidence suggests that this turbulence or convective force does not extend to the epithelial surface. [38] The view is that the apical microenvironment of epithelial cells is still (i.e., unstirred); moving toward the lumen, however, greater degrees of convection are achieved. Estimates of the depth of the unstirred layer range from 300 to 800 µm. The physiological significance of this layer has not been fully determined. [39] Consideration of the unstirred layer is important to investigators bent on achieving accurate kinetic data for epithelial transport systems; if surface convection is absent, transport must depend on the diffusive movement of molecules toward the epithelial surface. If uptake of the molecule by the epithelial transport system is faster than the diffusion to the epithelial surface, however, the concentration of the molecule at the epithelial surface will not be equivalent to the concentration of the molecule in the center of the lumen. To measure the kinetics of uptake of various molecules by epithelial cells, this confounding issue of unstirred layers must be confronted. Such considerations are particularly important when evaluating uptake of molecules such as lipids; these have permeability coefficients that are several orders of magnitude less in water than across lipid membranes. These considerations led to the unproven and perhaps erroneous conclusion that the unstirred layer plays a substantial role in physiological barrier function. First, as with mucus, the exposure of lumenal content to the epithelial surface often is long enough to permit substantial diffusive equilibration. Second, in absorptive epithelia, net inward water flow normally occurs after initiation of absorption, and this would establish inward convective movement at the epithelial surface. Third, experimental measurements of unstirred layers may be artifactual overestimates when they are determined in anesthetized animals. [40] For example, in the conscious rat, the unstirred layer of the small intestine is 100 µm or less, but this value increases dramatically with anesthesia, either alone or with laparotomy, to 200 and 600 µm, respectively. Although consideration of unstirred layers continues to be important to the investigator studying transport kinetics, it is less certain that this phenomenon contributes substantially to epithelial barrier function.

Secretory IgA The epithelial surfaces in the alimentary tract for the most part are bathed by secretory IgA. Secretory IgA is produced as a dimer in the subepithelial space by plasma cells, transported by transcytosis across epithelial cells by he poly-Ig receptor, and released into the lumen as a consequence of proteolytic clipping of the receptor as it reaches the apical membrane. Secretory IgA, by binding to lumenal threats such as pathogenic bacteria or important antigens such as cholera toxin, acts as a barrier to antigens. Although of extreme importance in host defense, this barrier is highly specific and dependent on antigenic sensitization. Secretory IgA binding to the surfaces of pathogens may not only impede pathogen-epithelial interactions over most of the epithelial surface, but may also actually enhance pathogen-epithelial interactions at selected sites such as the M cells, a cell type responsible for the afferent limb of intestinal immunity. [41]

Secreted HCO_3^- All the extrinsic barriers previously discussed exist throughout the alimentary tract; however, other extrinsic barriers have regional variation. A case in point is the net bicarbonate (HCO_3^-) secretion by epithelial surfaces that interface with acidic lumenal compartments: the stomach and the duodenum. Several microelectrode studies have shown that, whereas the pH of the gastric lumen may be 2 to 3, the pH at the surface of epithelial cells is at or near 7. [39] Initially, retarded diffusion of H^+ because of mucus and unstirred layers was thought to be the basis of this observation. As discussed, however, such explanations were inadequate given the time over which diffusive equilibration of H^+ within such barriers could occur. It has since been recognized that gastric surface foveolar cells and duodenal villus absorptive cells secrete HCO_3^- across the apical membrane and potentially assist in creating a microenvironment of neutral pH. This secretory process is an example of a highly specific and regionally localized extrinsic epithelial barrier.

Hydrophobic Layer If a researcher takes an epithelial surface such as that of the stomach or small intestine, blots the surface with absorbent material, places a drop of water on it, and carefully measures the angle at which the drop of water intersects the surface, a reasonable estimate of surface hydrophobicity can be obtained. [42] This highly imaginative yet beautifully simple approach has been used by investigators to demonstrate that at least some alimentary epithelial surfaces are coated with a hydrophobic barrier extrinsic to the epithelial cells. For example, a perfectly wetted surface would have a contact angle for water of 0°, and an extremely nonwetable surface would have a contact angle of 108°. Because the contact angle for aqueous solution on mammalian gastric mucosa is about 80°, it has been concluded that this is a nonwetable, hydrophobic surface. A hydrophobic layer provides a physical barrier to ions in aqueous solution. Questions remain about the chemical nature of hydrophobic barriers and the extent to which they are induced by the methods used to demonstrate them. Acceptance of this potentially important barrier and its distribution throughout the alimentary tract must await further developments in the field.

Antimicrobial Peptides Gut epithelial cells produce and secrete peptides with antimicrobial functions. Several classes of peptides have been isolated from humans, including members of the defensin, cathelicidin, and histatin families. Paneth cells at the base of the crypts in the small intestine and ascending colon produce several defensin peptides, termed cryptdins. [43] There are more than 20 defensins, and they are released specifically in response to bacteria, and not fungi or parasites. Some defensins not only exert direct antibacterial activity but also orchestrate a protective host response by signaling to immune cells and by stimulating apical Cl^- and water secretion to flush the lumen. [44] Paneth cells also release certain enzymes with antimicrobial activity, including lysozyme and type II phospholipase A_2.

Intrinsic Barriers

Extrinsic barriers differ in their relative contribution to different regions of the gastrointestinal tract. In contrast, the universal intrinsic barrier in all tissues is formed by the continuous sheet of epithelial cells that creates a separation of lumenal compounds from the subepithelial fluids.

Classically, discussions of the barrier function of epithelia consider the two pathways available to passive permeation: the transcellular pathway and the paracellular pathway (see Fig. 8-5). The exact physical site where solutes cross the epithelial cell layer was a topic of considerable controversy beginning in the mid 1800s. It seemed implausible that charged ions could traverse a lipid surface, until it was recognized in the 1960s that proteins insert into and form channels across the bilayer. Likewise, the paracellular space was thought to be impermeable and unregulated; a misconception perpetuated well into the 20th century owing to the static

appearance of intercellular contacts seen in early electron micrographs (note the use of the inappropriate term “tight” junction). Eventually, our present understanding of the transcellular and paracellular pathways emerged, along with appreciation that both are physiologically regulated and vary widely in different tissues. [45] We follow this division in our discussion of passive transepithelial movement of hydrophilic solutes. The principles guiding the movement of water and hydrophobic molecules across the epithelium are distinct from those governing the movement of hydrophilic molecules, however. Hydrophobic compounds can cross epithelial cells directly by virtue of their solubility in the lipid bilayer of the plasma membrane. Physiologically, the most important transepithelial movement of hydrophobic compounds occurs during fat absorption (see Chap. 18). Water permeation is complex and is reviewed separately.

Transcellular Pathway The transcellular pathway is highly restrictive to passive flow of hydrophilic solutes. To diffuse passively across a cell, an ion or other hydrophilic solute must interact with three barriers in series: the apical membrane, the cytosol, and the basolateral membrane (see Fig. 8-5). The lipid bilayers of the apical and basolateral membranes severely restrict permeation of hydrophilic solutes and tend to preserve transmembrane electrochemical gradients. For example, the resistance to passive ion flow across model lipid bilayers ranges from 10^{6} to 10^{9} Ohm·cm^{2}. [46] Similarly, as determined in osmotic experiments using such model membranes, the reflection coefficients for NaCl, glucose, and sucrose approximate 1 (i.e., low permeability). [46] Biologic membranes (i.e., lipid bilayers plus integral membrane proteins) also have resistances that restrict passive ion flow, usually in the range of 10^{3} to 10^{4} Ohm·cm^{2}. Such resistance is substantial considering that intact alimentary epithelia range in resistance from about 5×10^{1} to 5×10^{3} Ohm·cm^{2} (Table 8-2).

SITE	Ohm·cm^2
Esophagus (rabbit)	1659
Gallbladder (rabbit)	21
Small intestine	
Duodenum (rat)	67
Ileum (rabbit)	115
Colon (rabbit)	286–500

TABLE 8-2 Examples of Transepithelial Electrical Resistance Along the Alimentary Tract of Mammals

The ability of a lipid membrane to retard passive flow of hydrophilic solutes allows epithelial cells to maintain cytosolic ion concentrations far different from those in the interstitial space (e.g., Na^{+} < 40 mmol/L, K^{+} > 100 mmol/L, Ca^{2+} in the nanomolar range). As indicated by these data and as would seem ideal, the major component of ion movement across biologic membranes appears to be mediated by integral membrane proteins such as transporters, pumps, and channels (see Chap. 14). In the absence of specific transport pathways, intact biologic membranes can be considered virtually impermeable to hydrophilic solutes 0.5 nm or larger. In contrast to hydrophilic solutes, hydrophobic molecules readily permeate the lipid bilayer. For example, saturated fatty acids are calculated to exhibit astounding permeability coefficients across jejunal epithelial cell microvillus membranes: 10^{6} to 10^{7} cm/s compared with 10^{-5} to 10^{-6}cm/s in aqueous solution. The remaining barrier, other than the two plasma membranes, to restrict transcellular passive permeation is the cytosol. Because the cytosol has the characteristics of a hydrated gel, the resistance afforded by this cellular compartment is likely to be only marginally greater than that of free solution. It is possible to conclude that plasma membranes are the key barriers that restrict the passive movement of hydrophilic solutes across cells. In the 1960s, the observation that various small hydrophilic solutes could passively leak across gut epithelia led to the conclusion that small transepithelial pores were scattered through this epithelium. It was mistakenly assumed, however, that molecules could not permeate intercellular junctions, and the language used to describe the location of these pores in the early literature incorrectly implies that the pores are transcellular.

Paracellular Pathway The paracellular pathway is a major pathway for passive solute permeation. Although plasma membranes tend toward high resistance, alimentary epithelia, with the exception of the esophageal epithelium, have low net resistance (50–100 Ohm·cm^{2}), meaning they are relatively leaky (see Table 8-2). It follows that the remaining pathway, the paracellular pathway, must be largely responsible for such leaks in these epithelia. [47] The paracellular pathway consists of the apical intercellular tight junction and the underlying paracellular space. Under most conditions, tight junctions are the rate-limiting barrier restricting passive movement of hydrophilic solutes through the paracellular space. However, collapse of the subjunctional paracellular space may contribute to paracellular resistance in leaky epithelia, such as occurs in the gallbladder and small intestine. In the resting physiological state, tight junctions may leak small quantities of molecules the size of monosaccharides and disaccharides (i.e., fluxes on the order of 10^{-2}–10^{-4} mol/L · cm^{-2} · h^{-1}), or up to a physical cutoff of about 15Å in hydrodynamic radius. Such leakiness is physiologically regulated and, under some conditions, greatly increased. [27] As noted, the electrical resistance and even ion selectivity of tight junctions vary throughout the gastrointestinal tract. After the highly selective properties of transcellular transport have generated specific ion and solute gradients, the net composition on either side can be modified further by selective back-diffusion through the tight junction. Transcellular gradients in the leaky epithelia of the gallbladder, liver, and small intestine are quickly dissipated by paracellular movement of water and counter ions, and this explains the isosmotic nature of secretions in these tissues. In contrast, in tighter epithelia such as in the colon, tight junctions can maintain the ion gradients required for net movement of water out of the lumen. The selective and variable properties of tight junctions appear to be based on the differential expression of a family of transmembrane proteins called *claudins.* There are at least 20 members of this barrier-forming protein family in humans. Of the few that have been studied, each has a unique distribution within gastrointestinal epithelia, [48] for example, showing gradients along the crypt-villus axis or along the hepatic lobule. When expressed in cultured epithelial models, some claudins induce an increase and others a decrease in paracellular electrical resistance, and one claudin (claudin-4) has been shown to decrease the permeability of Na^{+} relative to Cl^{-}. [49] The expression of claudin-1 has been shown in cultured intestinal epithelial cells to be regulated by proinflammatory cytokines. [50] Human mutations in two nonintestinal claudins lead to epithelial diseases. Together, these early studies suggest there will soon be major insights into the molecular basis for defining and regulating paracellular transport in normal tissues and in disease.

Water Movement Across the Epithelial Barrier

Despite the obvious importance of fluid transport across gastrointestinal epithelia, controversy remains about the relative importance of the transcellular versus paracellular routes. One route for transcellular water movement is through transmembrane channels created by members of the aquaporin (AQP) protein family. These small (30-kd) integral membrane proteins are well studied in tissues specialized for regulated water transport, such as the collecting duct of the kidney. However, their relative contribution to the large movements of water in the gastrointestinal tract remains unresolved. At present, seven AQP water channel proteins have been described in gastrointestinal epithelia, including AQP1 in intrahepatic cholangiocytes, AQP3 and AQP4 in colonic surface epithelium, AQP4 in gastric parietal cells, AQP5 in salivary gland, AQP7 in small intestine, AQP8 in liver, pancreas, and colon, and AQP9 in liver. Indirect evidence for a role in water transport comes from the phenotype of mice with genetic deletions of AQP1 and AQP4 that show defects in dietary fat processing and colonic fluid absorption, respectively. [51]

A second route for transcellular water movement is across the lipid membranes. Movement of water is much less restricted than movement of hydrophilic solutes. [46] For example, the water diffusional permeability coefficient of model lipid bilayers is in the range of 2×10^{-5} to 2×10^{-2} per second, depending on the details of membrane composition. This range is the approximate range of water permeabilities in biologic membranes. How water permeates a biologic membrane in the absence of a specific channel is uncertain; however, given the equality of the diffusional and osmotic permeability coefficients for water moving across lipid bilayers, it appears that water movement occurs as independent diffusional movement of individual molecules rather than as cooperative laminar flow. One picture that emerges from such studies and that satisfies many of the observed features of water movement across membranes is that water diffuses across the nonpolar portion of the outer membrane leaflet, dissolves in the central polar portion of the membrane in proportion to its molar fraction in the outer aqueous phase, and moves by diffusion across the inner leaflet, assuming an inwardly directed gradient favoring water movement.

Because hydrophilic solutes the size of water can permeate tight junctions, water movement can occur across epithelia by the paracellular route. Although there is agreement that water crosses by the transcellular and paracellular routes, the relative partitioning of water flow between these two pathways is controversial. It has been estimated that about 50% of the water absorption that occurs during stimulated intestinal absorption is paracellular. [52] Data from other epithelia show that interepithelial differences in hydraulic conductivity, a measure of force-induced water flow, correlate reasonably well with paracellular resistance. Similarly, correlations between hydraulic water permeability (determined at hydrostatic pressures that do not move water transcellularly: 24.4 cm H_2O) and transepithelial voltage gradients, an indirect measure of junctional permeability, correlate inversely and well over a wide variety of epithelia. [53]

In aggregate, these observations suggest that the paracellular pathway is a major route for water flow across epithelia when such flow is driven by hydrostatic or osmotic pressures. In other specialized epithelia, such as that of the urinary bladder, most water uptake appears to be transcellular, through aquaporins, not paracellular. A basic consideration in such experiments is whether diffusional or convective movement is being measured. This issue is crucial because diffusional water movement varies with the square of the pore radius (r^{2}), and convective or force-associated water movement varies with the fourth power of the pore radius (r^{4}) as predicted by the Kedem-Katchalsky equation. [54] Although paracellular pathways may contribute little to diffusional water movement, they are important in convective water flow. Moreover, convective water flow secondary to osmotic pressures generated by nutrient or ion transport or by countercurrent vascular flow in the lamina propria is probably the more physiologically significant type of transepithelial water movement in the alimentary tract. Despite the characterization of

aquaporins and tight junction proteins, the basic issue of the route of water flow across gut epithelia remains surprisingly open.

EPITHELIAL HOMEOSTASIS AND RESPONSES TO DISEASE AND INJURY

Inductive Effects of Commensal Bacteria

Bacteria normally colonize the entire human gastrointestinal tract, with the highest concentration and number of species (>400) in the colon. [55], [56] Destructive interactions occur between the epithelium and numerous well characterized pathogenic species such as *Salmonella, Shigella,* and *Clostridium difficile* (see Chap. 88). In contrast, normal function of the gut actually is highly dependent on the resident commensal bacteria species. Such probiotic effects take several forms, including competing with pathogens for attachment to the epithelial surface, release of antimicrobial compounds, or synthesis of fatty acid metabolites that are toxic to the pathogenic species. Studies in germ-free mice demonstrate that normal development of immune cell lineages in the bone marrow and the lamina propria and local humoral defense depend on the presence of the commensal bacteria in the gut. [57], [58] Inductive effects on epithelial cell gene transcription have also been observed. For example, introducing a commensal *Bacteroides* species in germ-free mice extensively alters the transcription profile toward gene products that enhance nutrient uptake and metabolism. [59]

Although the molecular mechanisms remain poorly understood, reports of the probiotic effects of commensal bacteria appear in the clinical literature. For example, administration of *Lactobacillus acidophilus* reduces diarrhea in patients undergoing pelvic radiotherapy, possibly by reducing radiation damage or accelerating healing of the intestinal epithelium. [60] Similarly, ingestion of a mixture of commensal anaerobes prevents recurrent epithelial infections typically observed in patients with ileal pouch–anal anastomosis for ulcerative colitis. [61] The probiotic and pathogenic effects of bacteria on the intestinal epithelium are reviewed in detail in Chapter 25, Chapter 74, and Chapter 88.

Forms of Epithelial Injury Without Enhanced Permeation of Epithelial Monolayers

Epithelial injury is most readily apparent when gaps within the epithelium such as erosions or ulcerations are present; however, because the gut has remarkable ability for repair, there exist numerous forms of injury that do not result in a functionally significant defect. Futhermore, the life span of individual epithelial cells is influenced by physiological events, such as exposure to pancreatic proteases. Thus, one should view epithelial injury as spanning a spectrum that includes normal physiological challenges.

Gut epithelial turnover occurs, on average, once each week by a process of proliferation, migration, apoptosis, and sloughing. For example, gastric surface cells arise from the proliferative zone called the *gastric pit,* migrate to the surface as they become differentiated foveolar cells, and ultimately slough into the lumen. By removing physiological agents from the lumen, the half-life of epithelial cells often can be lengthened. Integral membrane proteins of the cell surface also turn over, displaying half-lives shorter than those of epithelial cells; however, a protein-depleted cell surface does not ultimately result, because these lost or damaged components are replaced constantly by individual epithelial cells. Undoubtedly, turnover of lipids in the membrane also occurs, but documentation is difficult because of the proclivity of membrane lipids to exchange side chains under the influence of membrane phospholipases. Components of individual epithelial cells turn over at rates that can be altered by normal physiological events, and these events also can modulate the life span of the cell. Thus, in its most subtle form, epithelial injury could be viewed as alteration in surface (or cytoplasmic) constituents without change in actual cell number.

A dramatic type of physiological gastrointestinal epithelial injury, limited to the membrane, has been documented. [62], [63] Because of the physical trauma accompanying events such as gut motility, the apical plasma membrane of epithelial cells can transiently break down. In such instances, macromolecular tracers present in the lumen leak into and label the cytoplasm of these cells. These tracers do not appear to leak across the basolateral membrane, and no evidence has been found to suggest that such sites may be foci of transepithelial macromolecular leaks. Because these labeled cells subsequently remain in the epithelium and retain normal appearances for 48 hours, it appears that such transient defects in apical membrane permeability are rapidly repaired. Experiments using cultured epithelial cells have similarly shown that, when scraped or trypsinized, plasma membranes can undergo a cycle of transient breakdown and rapid repair. Transient plasma membrane breakdown and subsequent rapid repair may be considered a second variety of physiological injury.

Subtle forms of epithelial injury also can lead to a diminished life span of individual absorptive cells. Because epithelial monolayers may extrude single damaged cells without loss of the epithelial barrier (even transiently), such events can occur while barrier function remains intact. [64] Dramatic remodeling of the intercellular junctions and extrusion of the apoptotic cell accomplish this without creating a break in the barrier. Such a mechanism is required, considering that about 10% of the proliferating cells in the intestinal crypt undergo apoptosis daily. [65]

Confluent Epithelium and Tight Junction Permeation

Diseases exist in which the epithelium remains confluent, but tight junction permeation is enhanced to inert solutes that are the size of disaccharides and larger. One example is celiac sprue. [66] At least some of the protein-losing gastropathies may fall into this category as well, and it has been suggested that patients with Crohn disease may exhibit a primary defect in tight junction permeability, [67] and that permeability increases before clinical manifestation of a flare in disease severity.

The suggestion that disease states could affect tight junction barrier function arose from various studies indicating that this crucial barrier is physiologically regulated. As described in Chapter 14, active transepithelial transport of nutrients such as glucose occurs across the villus absorptive cells. It appears that the presence of glucose within the lumen results in structural deformation of villus absorptive cell tight junctions and enhanced permeability of junctions to molecules the size of amino acids and glucose and perhaps to substantially larger molecules. The initial triggering event is activation of the Na^+-glucose cotransporter. Mild cytoplasmic alkalinization and subsequent activation of the Na^+/H^+-exchanger occur next. Activation of myosin light-chain kinase, phosphorylation of myosin light chain, and contraction of the perijunctional ring are downstream events. [68] The steps between apical and distal events in this pathway remain unknown. As active transcellular absorption of Na^+ and water proceeds, a transepithelial osmotic gradient of Na^+ and glucose is thought to develop, which drives water movement inward across the tight junction. Because such altered tight junctions have abnormally low reflection coefficients for nutrient-sized molecules, substantial paracellular nutrient uptake by way of solvent drag follows. Demonstration of glucose-induced absorption of undegradable octapeptides (e.g., D-amino acid–substituted) also suggests that this pathway can be used to deliver bulk quantities of stabilized bioactive peptides as oral pharmaceuticals.

If correct, this theory would explain why the intestine continues to absorb increasing amounts of nutrients from the lumen as lumenal nutrient concentrations rise far above the concentration at which the Na^+-glucose cotransporter is saturated. [69] Most important, this phenomenon highlights how transcellular active transport events can be intertwined intimately with paracellular barrier function and shows the plasticity that tight junction barrier function exhibits even in physiological states. In a variety of nonalimentary epithelia, classic intracellular mediators such as Ca^{2+}, cyclic adenosine monophosphate (cAMP), and protein kinase C and inflammatory mediators also can affect tight junction structure or permeability. [27] Such modulation of tight junction permeability may underlie the abnormal epithelial permeability encountered in diseased, confluent epithelia.

Modulation of Epithelia by Subepithelial Elements

Gut epithelial function may be modulated by a host of local factors derived from nonepithelial sources. Specific examples of such influences are provided in detail in chapters dealing with specific epithelia. In this chapter, the point is made that such events may occur throughout the alimentary tract in the physiological state and that subepithelial-epithelial interactions may be altered in states of enhanced epithelial permeation.

Before dealing with the effects of enhanced permeation on these subepithelial-epithelial interactions, the following are examples of such interactions in health. Alimentary epithelia rest on a structurally complex lamina propria, which, as shown in Figure 8-6, contains many candidate elements that could influence epithelial transport events or barrier function. Nerves, microvascular myofibroblasts, and a host of immune cells—including mast cells, macrophages, and lymphocytes—are within micrometers of epithelial cells. The factors derived from such elements may affect epithelial function profoundly. [70] Studies of these factors are performed largely by modulating one subepithelial component or by adding one agent known to be produced by subepithelial components to an in vitro epithelial sheet. Although data derived from these studies provide a framework for understanding how subepithelial elements influence epithelia, the net effects of orchestrated subepithelial events on epithelial function may not be understood clearly for some time.

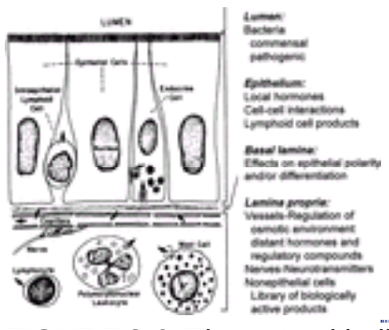

FIGURE 8-6. The gut epithelium is in proximity to lumenal, vascular, neural, and immune factors that substantially influence epithelial function.

Various examples of the dramatic influence of subepithelial signals on epithelial function now exist. For example, the lymphocyte-derived chemokine, interferon-?, induces cultured epithelial cells to down-regulate surface expression of various transporters and elicits surface expression of molecules with immune accessory function. [34] In nature, such cytokine-elicited *phenotype switch* of epithelia is undoubtedly complex, given the additive, negative, or synergistic interactions that can result from the concurrent presence of more than one cytokine.

Neurons of the enteric nervous system also produce a host of bioactive compounds, including peptides (e.g., vasoactive intestinal polypeptide, somatostatin, and cholecystokinin), catecholamines, and acetylcholine. [71] Moreover, termini from these nerves project to subepithelial positions, often within 1 µm of epithelial cells. The compounds produced by neurons influence epithelial functions ranging from acid secretion by gastric parietal cells to ion absorption by intestinal absorptive cells. Other examples of subepithelial influences on epithelial function include histamine, a product of mucosal mast cells, which produces a range of effects (e.g., induction of gastric acid secretion, intestinal Cl ¯ secretion) on alimentary epithelia. Mucosal mast cell activation in disease states in response to enhanced epithelial permeability must be considered another important modulator of epithelial function that may arise locally.

It is likely that enhanced epithelial permeation in disease states substantially modifies epithelial interactions with subepithelial elements and further modifies epithelial function. For example, polymorphonuclear leukocytes (PMNs) migrate across the gut epithelium and collect in gastric pits or intestinal crypts. Indeed, this histological event defines active inflammation in many alimentary inflammatory states. When crossing the epithelium, PMNs squeeze through intercellular tight junctions (Fig. 8-7), and they induce a reversible increase in per- meability. [72] Polarized interactions involving PMNs also can influence epithelial function differentially. For example, PMNs interacting with the apical membranes of cryptlike epithelial cells can elicit electrogenic Cl ¯ secretion as a result of paracrine release of 5'-AMP by PMNs, surface conversion of this signal by CD73 on the apical membrane of epithelial cells, and binding of the resulting adenosine signal to the apical A2b adenosine receptor.

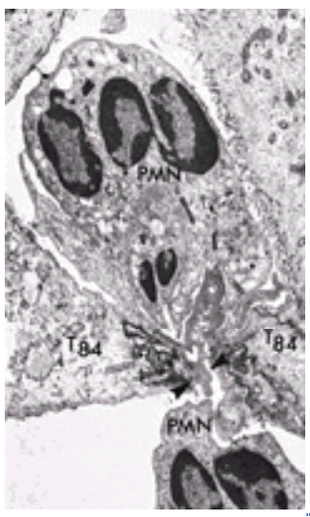

FIGURE 8-7. In an in vitro model of gut epithelial acute inflammation, neutrophils squeeze through an intercellular tight junction and induce only a small increase in junctional permeability. Mediators released from cells of the immune system or other subepithelial cells, such as fibroblasts, also are likely to influence epithelial transport and barrier function, even without direct epithelial cell–immune cell contact, such as that seen here. (From Nash S, Stafford J, Madara JL. Effects of polymorphonuclear leukocyte transmigration on the barrier function of cultured intestinal epithelial monolayers. J Clin Invest 1987;80:1104, with permission.)

It is now clear that not only do subepithelial signals influence epithelial characteristics, but also epithelia can regulate subepithelial events. For example, in response to the detection of apical pathogens such as *Salmonella typhimurium,* epithelial cells release many cytokines that orchestrate transepithelial migration of PMNs. [73]

Healing of Epithelial Wounds

When epithelial proliferation no longer keeps up with accelerated epithelial loss resulting from injury, a defect in the epithelial barrier ensues. Erosions or ulcers develop that also destroy the underlying lamina propria and muscularis mucosae. To evaluate this situation in terms of the concepts reviewed in this chapter, such an injury may be viewed as a dramatic focal expansion of the paracellular pathway associated with loss of the rate-limiting barrier to paracellular flow: the intercellular tight junction. Thus, in open epithelial wounds, the surface area exposed to unrestricted permeation by noxious lumenal solutes in orders of magnitude is greater than if two viable cells are separated at the tight junction by 1 or 2 µm. Governed by Fick's first law of diffusional permeation (P_D), the rate of P_D directly relates to the surface area of the pore (wound), and this finding provides qualitative grounds for the intuitive realization that small breaks encompassing several cell widths expose subepithelial tissues to huge pulses of lumenal solutes. As a result, transepithelial leaks of paracellular tracer molecules, such as polyethylene glycol-400, can increase by more than 200% in patients with erosive intestinal disease, even though the eroded surface may represent a minute percentage of the net intestinal surface area. [67]

Once wounded, epithelia often respond by enhanced proliferation in an effort to replenish cells in the eroded area. An increased fraction of progenitor cells is stimulated to move through the S, G_2, and M phases of the cell cycle. Because the S phase of the cell cycle alone is 6 to 12 hours long, however, the benefit of this proliferative response is not recognized for many hours. Although the stimuli for enhanced proliferation in gut epithelia are not precisely known, various classic growth factors (e.g., epidermal growth factor) may participate in such responses (see Chap. 4 for a detailed description of peptide-elicited mucosal growth). Stimulated production of new epithelial cells requires a minimum of several hours to increase the rate of epithelial cell renewal. In the short term, reepithelialization of small wounds occurs by the remarkable process known as *restitution,* [34] characterized by the rapid spreading of epithelial cells shouldering the wound. By converting from a tall columnar to a spread and somewhat flattened phenotype, these cells produce a maximal footprint on the basement membrane and in so doing attenuate the diameter of the wound. Because an entire collar of cells spreads and surrounds the wound, the wound closes in the time frame of cytoskeletal motility events (i.e., minutes) rather than of cell cycle events (i.e., hours).

Signals involved in the stimulation of restitution are beginning to be understood. Several growth factors (e.g., epidermal growth factor, fibroblast growth factor, and hepatocyte growth factor) enhance restitution by increasing the number of motile, spreading epithelial cells. [34] Several such factors signal through a common transforming growth factor-ß1 (TGF-ß1)–dependent pathway. In fact, TGF-ß1 itself may be released from the epithelial cytoplasm in the process of injury. Other factors that promote healing by stimulating restitution (and perhaps replication) include peptides secreted by epithelia known as trefoil factors. Mice in which the gene encoding intestinal trefoil factor was disrupted experienced difficulty in healing injured mucosa. [74] These various factors use different mechanisms to promote

restitution. Some peptides may stimulate epithelial spreading and motility, and others may do so through a shared TGF-ß1–dependent pathway; still others (e.g., trefoil peptides) may promote the viscosity of the overlying mucous gel, thus potentially protecting the underlying restituting epithelia. [75]

The precise signal transduction pathways and targeted molecular events influenced by restitution-enhancing factors are not well understood currently but undoubtedly include cytoskeletal events and cell-matrix interactions. Roles for laminin isoform a6 and integrin laminin receptors a3ß1 and a6ß4 in the restitution of model intestinal epithelia have been defined. [76]

Nonmatrix elements in the lamina propria also may influence epithelial resealing after injury. In a model of focal intestinal epithelial denudation, restitution is aided by myofibroblast-mediated contraction of the lamina propria. This contractile event effectively diminishes the size of the defect to be reepithelialized.

During the early 1980s, a phenomenon called *cytoprotection* was popularized. Certain compounds (e.g., prostaglandins) that were in concentration too low to influence epithelial proliferation appeared to promote epithelial resealing of open wounds; however, cell injury in these early studies was judged largely by gross inspection of epithelial surfaces. Subsequent microscopic studies revealed that surface epithelial cells often were injured equally in control and cytoprotected mucosae. [77] A major difference between control and cytoprotected groups appeared to be vascular. Cytoprotection prevented extravasation of red blood cells from subepithelial vessels. It is also clear that cytoprotection reduced the depth of mucosal necrosis in some models, which ultimately may have clinical importance.

Integration of Barrier Function, Repair, and Epithelial-Immunological Cell Interactions

The defense of the mucosa against its harsh lumenal environment consists of the integration of the concepts and principles defined in this chapter. The epithelial barrier retards passive movement of molecules across it and is itself continuously repaired by rapid epithelial renewal, restitution, and other mechanisms. Moreover, resident intraepithelial lymphocytes and lamina propria lymphocytes and macrophages, along with the M cell and IgA secretory immune system, interface with this epithelial barrier.

The gut epithelial barrier, which restricts passive permeation, is complex and dynamic. Maintenance of this barrier in health depends on the integrity of cellular plasma membranes and tight junctions and the elaboration of epithelial secretory products such as HCO_3^-. Focal denudation of this barrier results in permeation by a host of threatening lumenal compounds, including antigens, proteases, H^+, and factors chemotactic for inflammatory cells. By initiating inflammation and acting on subepithelial tissues, such factors can further influence epithelial transport and barrier function. Repair of epithelial injury is also complex, and restitution and enhanced epithelial cell proliferation are likely to play major roles. Future studies should be directed toward examining the molecular mechanisms by which such barriers are maintained and repaired.

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CHAPTER 9

Piero Biancani, Karen M. Harnett, and Jose Behar

ESOPHAGEAL MOTOR FUNCTION

The esophagus is responsible for transporting food from the mouth to the stomach and for preventing retrograde movement of esophageal or gastric contents, movement that may result in reflux of gastric secretions or regurgitation of food. It is a hollow tube closed at both ends by the upper esophageal sphincter (UES) and the lower esophageal sphincter (LES). Its lumen is lined with squamous mucosa, containing longitudinally oriented muscle fibers and connected to the muscularis propria by a loose network of connective tissue fibers of the submucosa. The muscularis propria consists of an inner circular muscle layer with fibers oriented along the circumference of the tube and an outer longitudinal layer with fibers oriented along its axis.

The pharynx and proximal esophagus contain striated muscle controlled entirely by the swallowing center, located in the brainstem, through the vagus nerves. The control mechanisms for the lower two thirds of the esophagus, which contains smooth muscle, are quite different from the striated muscle portion. In the smooth muscle portion of the esophagus, peristalsis is controlled primarily by intrinsic neural networks located between the longitudinal and circular muscle layers (i.e., the Auerbach plexus) and in the submucosa (i.e., the Meissner plexus) and is only modulated by central mechanisms in the swallowing center. Myogenic factors, however, may also play a role in peristalsis.

Deglutition, which is the normal stimulus that initiates pharyngoesophageal motor activity and results in propulsion of the bolus from the pharynx to the stomach, is divided into buccopharyngeal and esophageal phases. Because of the high speed of contraction in the pharynx, deglutition has been studied by cinefluoroscopy, manometry, and combinations of both methods. The slower esophageal contractions have been studied mainly by manometry.

In humans and most mammals, the esophagus propels its contents in a caudad direction, but it behaves only as a passive conduit when orad transport occurs. Esophageal function is complicated by the fact that the airways and digestive tract cross in the pharynx, and their respective contents must be kept separate; this requires precise control and coordination of swallowing and respiration, controlled by the rich innervation of pharyngeal muscles.

Initiation of swallowing is voluntary, but continuation is mediated reflexly by sensory neurons with input to the swallowing center. In the buccopharyngeal phase, the bolus is gathered on the tongue in preparation for swallowing and is confined to the oral cavity by apposition of the soft palate to the posterior portion of the tongue. The voluntary portion of swallowing consists of closing the mouth and pressing the tip of the tongue against the palate while a progressive wave of contraction travels through the body of the tongue, which squeezes the bolus toward the pharynx. Then follows a series of involuntary events, consisting of transient suppression of respiration, during which the bolus slides to the base of the tongue and is pushed by the tongue toward the oropharynx, which is elevated and forced open. The posterior thrust of the tongue propels the bolus into the pharynx. The descending wave of pharyngeal peristalsis begins and continues through the open UES. A rapid series of events occurs in the pharynx: closure of the velopharyngeus to prevent reflux into the nose, closure of the larynx to prevent aspiration, pharyngeal peristalsis to clear the bolus out of the pharynx, and displacement of the larynx upward and forward to move out of the path of the bolus and to force open the cricopharyngeal region and the UES.

Relaxation of the UES occurs immediately, almost simultaneously with the initiation of swallowing, and has a brief duration. In the esophageal phase, the bolus is forcefully injected into the esophagus through the opened UES. A primary peristaltic wave is initiated in the pharynx and continues into the upper portion of the cervical esophagus. As the bolus enters into the body of the esophagus in upright subjects, it descends by gravity and is propelled by peristalsis. The relative contribution of gravity and peristalsis depends on bolus consistency. In the upright position, liquids travel faster by gravity than the peristaltic contraction that follows the swallow. Liquids travel fast enough to impact on the closed LES.

After the peristaltic wave reaches the liquid bolus, the LES is opened, and the bolus is pushed through the sphincter. Occasionally, small amounts of fluid may remain in the distal esophagus, behind the peristaltic wave, necessitating a second contraction unrelated to swallowing and triggered by local esophageal distention (i.e., secondary peristalsis), to clear the esophagus completely. Solid boluses are propelled by peristalsis even in the upright position, but their movement is helped by gravity. Progression of the bolus does not outstrip peristalsis; the bolus and the peristaltic wave move through the esophagus at the same speed. The bolus, propelled by the peristaltic contraction, forces the opening of the relaxed LES.

Because gastric contents are damaging to the esophageal squamous epithelium, esophageal function must be designed so one-way flow of nutrients occurs, preventing reflux, except for occasional events such as vomiting or belching. The presence of tonic contraction in the UES and LES prevents or minimizes gastroesophageal reflux or esophagopharyngeal regurgitation. If gastroesophageal reflux occurs, however, the refluxed material is cleared by secondary peristalsis triggered by localized distention of the esophagus.

INNERVATION

Esophageal peristalsis and sphincter function are controlled by the autonomic nervous system, with contributions from the parasympathetic, sympathetic, and enteric divisions.

Parasympathetic Nervous System

The central control of esophageal and LES function arises from preganglionic parasympathetic neurons originating in the dorsal vagal complex located in the dorsomedial hindbrain medulla [1], [2], [3] and [4] (Fig. 9-1). The dorsal vagal complex comprises two nuclei, the *nucleus tractus solitarius* (NTS), which receives the sensory input from the viscera, and the *dorsal motor nucleus of the vagus* (DMV), which contains the preganglionic motor output to the viscera. [5] The NTS comprises subdivisions that include the central (NTScen), intermediate (NTSint), and interstitial (NTSis) subnuclei.

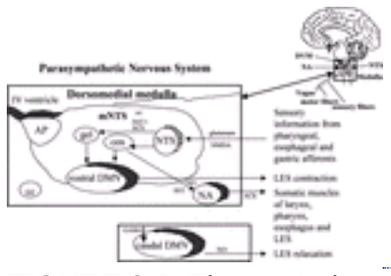

FIGURE 9-1. The central control of esophageal and lower esophageal sphincter (*LES*) function arises from preganglionic parasympathetic neurons originating in the dorsal vagal complex located in the dorsomedial hindbrain medulla (*upper right corner*). A summary diagram of the dorsomedial medulla oblongata is shown on the *left.* The dorsal vagal complex comprises two nuclei: the nucleus tractus solitarius (*NTS*), which receives the sensory input from the viscera, and the dorsal motor nucleus of the vagus (*DMV*), which contains the preganglionic motor output to the viscera. The neurocircuitry controlling swallowing begins with sensory afferents from the esophagus projecting to the NTS and terminating on premotor neurons in the subnucleus centralis (*cen*). Cell bodies from the subnucleus centralis project directly to somatic motor neurons within the nucleus ambiguus (*NA*) that control the buccopharyngeal pattern generator for swallowing. This provides a direct transfer of information coming into the NTS from peripheral sensory endings in the esophagus to the motor neurons in the NA that innervate the muscles involved in deglutition. Relaxation of the LES is controlled by long-loop neural feedback involving the vagus nerve and the DMV. Excitatory and inhibitory control of LES pressure is mediated by vagal efferent neurons located in two distinct sites in the DMV. Excitatory neurons located in the rostral DMV produce LES contraction, and inhibitory neurons in the caudal DMV produce LES relaxation. The neurotransmitters involved in the vagal control of the LES include the neurotransmitters in the subnucleus centralis and medial subnuclei of the NTS (*mNTS*) that mediate afferent integration of swallowing and LES function and the neurotransmitters that control vagal efferent outflow from the DMV to the LES. L-Glutamate is a primary afferent neurotransmitter released in the NTS from esophageal afferents. Control of vagal preganglionic neurons may involve the inhibitory neurotransmitter ?-aminobutyric acid (*GABA*). Most preganglionic neurons in the DMV are cholinergic. Nitrergic neurons form a separate population from the cholinergic neurons in the DMV and peripherally released NO mediates LES and gastric relaxation. *ACh,* acetylcholine; *a1,* a1 adrenergic receptors; *AP,* area postrema; *cc,* central canal; gel, subnucleus gelatinosus; *NMDA, N*-methyl-D-aspartate; *NO,* nitric oxide; *5HT2,* serotonin; *SST,* somatostatin. (Adapted from ref. [59].)

Sensory neurons from the entire esophagus run in the vagus nerve. In the lower esophagus, they are also found in the splanchnic and thoracic sympathetic nerves. The cell bodies of the vagal fibers reside in the nodose ganglion, and those of the sympathetic fibers are found in the vertebral ganglion of the thoracolumbar region. Sensory neurons within the nodose ganglion contain the neurotransmitters glutamate, d-aminobutyric acid (GABA), catecholamines, serotonin, acetylcholine (ACh); and the neuropeptides substance P, vasoactive intestinal polypeptide (VIP), neurokinin A, calcitonin gene–related peptide (CGRP), galanin, enkephalin, somatostatin, cholecystokinin (CCK), neuropeptide Y, and nitric oxide (NO). [6], [7], [8], [9] and [10] These afferent fibers innervate the serosa (adventitia), the longitudinal and circular muscle, and the mucosa of the esophagus. Afferents innervating the muscle are sensitive to distention and chemicals, and afferents innervating the mucosa are sensitive to light touch, pH, and chemicals such as hypertonic saline, sodium hydroxide, serotonin, bradykinin, and prostaglandins. [11]

The DMV innervates the smooth muscle of the LES [12], [13] and stomach. [14], [15] The motor innervation of the striated muscle of the pharynx, esophagus, and LES includes somatic nerves with cell bodies located in the nucleus retrofacialis and the rostral portion of the nucleus ambiguus. These nerves end at the motor endplate of the striated muscle fibers. The transmitter released by these neurons is ACh, acting through nicotinic cholinergic receptors in the endplate of the striated muscle fibers. [16] The somatic motor nerves supplying the pharyngeal muscles, including the UES, accompany the vagus and branch out into the pharyngeal nerve at the level of the nodose ganglion. Most nerves supplying esophageal striated muscle originate from the vagus in the upper cervical region, but some also arise from the recurrent laryngeal nerve. All these somatic nerve fibers are unmyelinated in their terminal portions. [17], [18]

The neurocircuitry controlling swallowing begins with sensory afferents from the esophagus projecting to the NTS and terminating on premotor neurons in the NTScen [5], [19] (see Fig. 9-1). Cell bodies from the NTScen project directly to somatic motor neurons within the nucleus ambiguus [19] that control the buccopharyngeal pattern generator for swallowing, [20], [21] thereby providing a direct transfer of information coming into the NTS from peripheral sensory endings in the esophagus to the motor neurons in the nucleus ambiguus that innervate the muscles involved in deglutition. Sequential, peristaltic contraction of the esophagus is coordinated by the brainstem–pattern generator circuit involving the NTS and is modulated by vagal afferents. The swallowing central pattern generator is an interneuronal network of premotor neurons that organizes the sequential activity of the motor neurons active during swallowing. Two neuroanatomically distinct subnetworks of premotor neurons are involved in the buccopharyngeal and esophageal phases of swallowing. [19], [22], [23] and [24] Palatal, pharyngeal, and laryngeal afferents overlap and terminate in the NTSint and NTSis. [25] In contrast, esophageal afferents terminate in the NTScen and do not overlap in their distribution with palatal, pharyngeal, and laryngeal afferents. [25] Motor neurons projecting to the pharynx and larynx are located in the semicompact and loose formation of the nucleus ambiguus, and esophageal motor neurons reside in the compact formation of the nucleus ambiguus. Although distinct neuroanatomic circuits control the buccopharyngeal and esophageal phases of swallowing, neuropharmacological data suggest that a central mechanism coordinates the phases without peripheral feedback. [26], [27], [28] and [29] Using Barth PRV, an attenuated vaccine strain of swine a-herpesvirus, a subpopulation of esophageal premotor neurons were found to project to pharyngeal and buccopharyngeal premotor neurons in the NTSint and NTSis, providing a synaptic link within the NTS for the central coordination of the two deglutitive phases. [30], [31]

Esophageal peristaltic contraction depends on cholinergic (muscarinic) excitation and NO-mediated inhibition. [32] NO may be involved in the central reflex control of esophageal peristalsis. [33] NO synthase, the NO-producing enzyme, is present in premotor neurons of the NTScen that innervate esophageal motor neurons in the nucleus ambiguus. Nicotinamide adenine dinucleotide phosphate (NADPH)-diaphorase staining of both the somata and terminals of esophageal premotor neurons suggests that NO is involved in neurotransmission in the NTScen and at the site of synaptic contact between esophageal premotor neurons and motor neurons in the nucleus ambiguus. [34] In addition to NO, esophageal premotor neurons of the NTS are tonically inhibited by GABAergic neurons through GABA receptors. The expression of $GABA_{Aa1}$ mRNA subunit within esophageal premotor neurons of the NTS supports a $GABA_{Aa1}$ role for GABA in the brainstem circuit that controls esophageal peristalsis. [35]

Relaxation of the LES is controlled by *long-loop neural feedback* involving the vagus nerve. [5] The integration center of vagal control of LES relaxation is the DMV of the dorsal vagal complex. The vagal motor neurons innervating the LES are clustered within two distinct populations in the DMV, one rostral and one caudal to the obex. The *obex* is an anatomic landmark that marks the opening of the central canal into the fourth ventricle. Studies indicate that excitatory and inhibitory control of LES pressure may be mediated by vagal efferent neurons located in these two distinct sites in the DMV. Excitatory neurons arise from the area rostral to the obex, and inhibitory neurons are located caudal to the obex. [4] Stimulation of rostral neurons by microinjection of the excitatory neurotransmitter L-glutamic acid, increases LES pressure, whereas stimulation of caudal neurons decreases LES pressure. The axons of these efferent neurons are carried in the vagus nerves. The axons branch into the esophageal plexus surrounding the body of the esophagus and enter the esophagus at various levels. They travel within the esophageal body for some distance before synapsing on postganglionic neurons in the enteric esophageal plexuses. These data suggest that both peripheral ascending and central descending inputs related to esophageal function are integrated in the NTS, leading to activation of separate opulations of excitatory and inhibitory preganglionic neurons in the DMV. The contribution of vagal inhibitory and excitatory neurons modulates basal LES pressure, as well as the frequency, amplitude, and duration of spontaneous and swallow-induced LES relaxations. The demonstration of reduced gastric motility and tone after balloon distention of the rat thoracic esophagus supported this view. [36] Esophageal distention resulted in activation of neurons in the solitary tract, pars centralis, the recipient of esophageal afferent projections from the vagus nerve. Activation of the NTS by balloon distention resulted in excitation of lateral and caudal neurons in the DMV and inhibition of medial and rostral DMV neurons.

The neurotransmitters involved in the vagal control of the LES include the neurotransmitters in the NTScen and medial subnuclei of the NTS that mediate afferent integration of swallowing and LES function and the neurotransmitters that control vagal efferent outflow from the DMV to the LES. L-Glutamate (or a closely related substance) is a primary afferent neurotransmitter released in the NTS from esophageal afferents. [5], [37] Microinjection of L-glutamate or NMDA agonists into the NTScen induces swallowing, whereas NMDA antagonist injection inhibits the esophageal component of swallowing. [38], [39] Expression of NMDA receptors within the brainstem circuit that controls esophageal swallowing has been reported. [40] These data suggest that excitatory amino acid release in the NTS in response to gastric distention or pharyngeal stimulation may induce swallowing. Pharyngeal and esophageal components of peristalsis can be stimulated by the microinjection of ACh, [20] serotonin agonists, [41] and a-adrenergic agonists [42] into the NTS. Binding sites for CCK (A and B subtypes) and dopamine (D_2 subtype) have been identified in the NTS. [43] Somatostatin and NO synthase are present in neurons projecting to the laryngeal and pharyngeal motor neurons of the nucleus ambiguus, suggesting that a relay between sensory information and the activation of swallowing is nitrergic [44] and somatostatinergic. [33]

Direct neurotransmitter control of vagal preganglionic neurons may involve the inhibitory neurotransmitter GABA (see Fig. 9-1). [5] Most preganglionic neurons in the

DMV are cholinergic (Fig. 9-2). In addition, catecholaminergic and nitrergic abdominal vagal preganglionic neurons exist in the DMV. The catecholamines are colocalized with ACh in the DMV neurons; however, it is unclear whether the catecholamines contribute to the central control of LES pressure. In contrast, nitrergic neurons form a separate population from the cholinergic neurons in the DMV, and peripherally released NO mediates LES [45], [46] and gastric relaxation. [47], [48], [49], [50], [51], [52] and [53] The nonadrenergic noncholinergic (NANC) inhibitory control of relaxation involves cholinergic preganglionic nerves that innervate NANC myenteric postganglionic neurons (see Fig. 9-2). In addition, NO synthase is present in preganglionic neurons in the DMV that innervate the gastrointestinal tract, [54], [55] and [56] providing a novel pathway in parallel to the more traditional view of vagal NANC inhibition. [5] NO synthase–containing preganglionic neurons are concentrated in two populations in the DMV, one caudal and the other rostral to the obex. Because L-glutamate stimulation of caudal DMV neurons causes LES relaxation, [4] it is possible that the nitrergic preganglionic neurons in the caudal DMV mediate LES relaxation. Hornby and Abrahams [5] reported that stimulation of nitrergic preganglionic vagal motor neurons produces gastric relaxation. Gastric relaxation was stimulated by microinjection of substance P into the nucleus raphes obscurus, a hindbrain nucleus with functional connections to the dorsal vagal complex. [57], [58] With this model, they showed that centrally evoked relaxation depends on simultaneous inhibition of cholinergic tone and activation of NO and VIP pathways. [59] These studies demonstrate a role of preganglionic vagal cholinergic tone and NO release in gastric relaxation. The mechanism for centrally mediated vagal cholinergic and NANC control of LES relaxation is unknown; however, the neurocircuitry for the control of intragastric and LES pressure may be similar because the LES relaxation associated with deglutition is combined with gastric adaptive relaxation to accommodate the bolus of food.

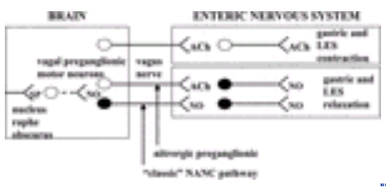

FIGURE 9-2. The “classic” nonadrenergic noncholinergic (*NANC*) inhibitory control of lower esophageal sphincter (*LES*) and gastric relaxation consists of cholinergic preganglionic nerves that innervate NANC myenteric postganglionic neurons. Nitric oxide (*NO*) synthase, however, also is present in preganglionic neurons in the DMV that innervate the gastrointestinal tract and provide a novel pathway in parallel to the more traditional view of vagal NANC inhibition. Microinjection of substance P (*SP*) into the nucleus raphes obscurus stimulates the dorsal vagal complex and produces vagally mediated gastric relaxation. *Solid lines* represent monosynaptic pathways; the *dashed line* represents polysynaptic pathways. (Adapted from ref. [5].)

Sympathetic Nervous System

The cells that give rise to the preganglionic sympathetic fibers innervating the pharynx, esophagus, and LES originate in the intermediolateral columns of the spinal cord (segments T1–T10) (Fig. 9-3), and their preganglionic fibers enter the cervical, thoracic, and possibly celiac ganglia. [60] Most preganglionic fibers pass through the greater splanchnic nerves, terminate in the celiac ganglia, and then synapse with postganglionic neurons. [3], [61] Most postganglionic axons originate in the celiac ganglia or reach the esophagus by way of perivascular fibers. Most postganglionic axons synapse in the myenteric or in the submucosal plexus, and few innervate the smooth muscle directly. [16], [61]

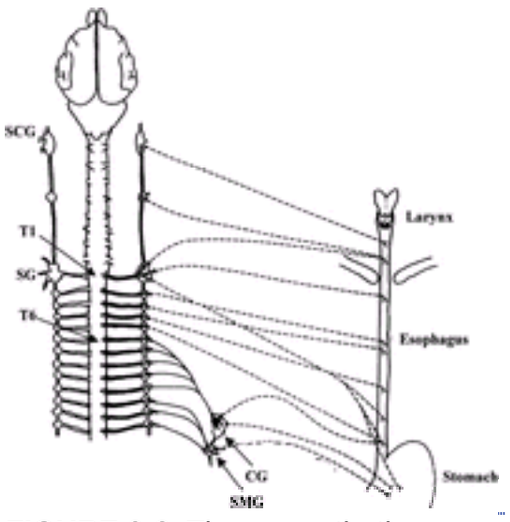

FIGURE 9-3. The sympathetic nervous system. The cells that give rise to the preganglionic sympathetic fibers innervating the pharynx, esophagus, and lower esophageal sphincter originate in the intermediolateral columns of the spinal cord (segments T1–T10). Their preganglionic fibers enter the cervical, thoracic, and celiac ganglia. Most preganglionic fibers pass through the greater splanchnic nerves and terminate in the celiac ganglia, where they synapse with postganglionic neurons. Most postganglionic axons originate in the celiac ganglia or reach the esophagus by way of perivascular fibers, and most postganglionic axons synapse in the myenteric or in the submucosal plexus. A few innervate the smooth muscle directly. *CG,* celiac ganglion; *SCG,* superior cervical ganglion; *SG,* stellate ganglion; *SMG,* superior mesenteric ganglion; *T1,* first thoracic spinal cord segment; *T6,* sixth thoracic spinal cord segment. (Adapted from ref. [60].)

Enteric Nervous System

The esophagus has a rich network of intrinsic neurons in the submucosa (i.e., the Meissner plexus) and between the circular and longitudinal muscle layers (i.e., the Auerbach plexus). The enteric neural network receives instruction from and sends signals to the central nervous system by way of the vagi, the adrenergic ganglia in the thoracic sympathetic chain, and the celiac ganglia. This network of enteric neurons is capable of producing secondary peristalsis in the smooth muscle portion even when the esophagus is separated from the central nervous system and is isolated in an organ bath. [62]

At the turn of the 20th century, Bayliss and Starling [63], [64] and [65] described descending relaxation in the intestine that persisted when the intestine was extrinsically denervated by resecting vagal and mesenteric fibers. Langley and Magnus [66] reported that this reflex was maintained in vitro in the absence of any extrinsic innervation. Based on this evidence, the intrinsic neurons were thought to constitute a separate enteric division of the autonomic nervous system. It has become apparent that this enteric network is composed of a variety of neural cells containing numerous neuropeptides that also are found in the brain. Peptides present in esophageal and LES neurons include the following: VIP; neuropeptide Y, which is often present in neurons containing VIP or catecholamines; CGRP; substance P, which is often present with enkephalins and galanin; and pituitary adenylate cyclase–activating peptide (PACAP), which is often present with VIP. [6], [7], [34], [67], [68], [69], [70], [71], [72], [73] and [74]

In addition to the neuropeptides, NO-releasing nerves regulate esophageal and LES smooth muscle function. [75] Stimulation of intrinsic esophageal nerves releases NO from the LES. [76] NO synthase colocalizes with such neuropeptides as VIP, CGRP, and galanin and rarely with neuropeptide Y and substance P, [77] a finding suggesting that the nerves responsible for peristalsis in the esophagus may act by releasing NO along with other inhibitory neuropeptides, such as CGRP, galanin, and VIP, but not excitatory substances, such as neuropeptide Y and substance P. [75], [77]

PHARYNX AND OROPHARYNX

Initiation of swallowing is voluntary, but its continuation is mediated reflexly by sensory neurons with input to the swallowing center located in the brainstem. The

tongue is the major participant in the oral phase. It contributes to the formation, containment, and propulsion of the bolus; it adapts to bolus size and creates a larger propulsive chamber with increasing bolus sizes. During deglutition, the tongue exhibits a pattern of surface motion suggesting a lingual propulsive activity that appears as a wave along the central groove of the tongue. Propulsion is accomplished with the tip of the tongue pressing against the palate, with its lateral aspects sealing against the alveolar ridges and pharyngeal walls, and with the central groove of the tongue exhibiting appropriate motions to push the bolus toward the pharynx. This process is followed by a transient suppression of respiration while the bolus slides over the base of the tongue into the pharynx. [78]

Pharyngeal Anatomy

The pharynx, connecting the nose and mouth on the proximal end to the esophagus and trachea on the distal end, is responsible for separating food and air as they pass through this area (Fig. 9-4). The necessary fine motor control is reflected by the complexity of this structure. An excellent representation of pharyngeal muscles and deglutition sequence is shown in the article by F. H. Netter, [79] found in the Ciba Collection of Medical Illustrations.

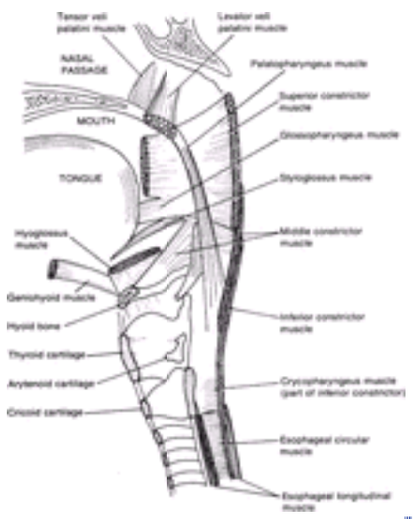

FIGURE 9-4. The pharynx, connecting the nose and mouth on the proximal end to the esophagus and trachea on the distal end, is responsible for separating food and air as they pass through this area. This process requires fine motor control and is reflected by the complexity of this structure. The pharynx consists of several distinct muscle groups, and in its lower portion, it is supported anteriorly by arytenoid, cuneiform, corniculate, and cricoid cartilages. Traditionally, the pharynx has been divided into the nasopharynx, the oropharynx, and the hypopharynx. The nasopharynx, extending from the base of the skull behind the soft palate to the distal edge of the soft palate, is not part of the alimentary tract. Muscles in the nasopharynx, such as the tensor veli palatini, levator veli palatini, and others, contribute to elevating the soft palate and closing the nasopharyngeal passage during swallowing, to prevent bolus entry into the nasal passage. The oropharynx extends from the soft palate above to the base of the tongue and the level of the hyoid bone below and contains the upper border of the epiglottis, called the valleculae. In this area, the respiratory and gastrointestinal tracts cross. Muscles in the oropharynx are responsible for bolus propulsion (e.g., middle constrictor) and for elevation (e.g., palatopharyngeus) and forward displacement (e.g., geniohyoid) of the pharynx. The hypopharynx extends from the valleculae at the base of the tongue to the lower border of the cricoid cartilage and contains the inferior constrictor muscle and the upper esophageal sphincter. (Adapted from ref. [79].)

The pharynx is a hollow cylinder, 12 to 14 cm long, extending from the base of the skull to the lower border of the cricoid cartilage and bordering posteriorly on the cervical spine. It consists of several muscle groups and in its lower portion is supported anteriorly by the arytenoid, cuneiform, corniculate, and cricoid cartilages.

Traditionally, the pharynx has been divided into three segments: nasopharynx, oropharynx, and hypopharynx. The nasopharynx extends from the base of the skull, behind the soft palate, to the distal edge of the soft palate and is not part of the alimentary tract; however, muscles located in the nasopharynx contribute to elevating the soft palate and closing the nasopharyngeal passage during swallowing, thus preventing bolus entry into the nasal passage. The mechanism of nasopharyngeal closure during belching and swallowing is different. During swallowing, nasopharyngeal closure consists of palatal elevation and adduction of the superior pharyngeal muscle, whereas during belching, only palatal elevation occurs. [80] The oropharynx extends from the soft palate above to the base of the tongue and the level of the hyoid bone below, and it contains the upper border of the epiglottis, called the *valleculae.* In this area, the respiratory and gastrointestinal tracts cross. The hypopharynx extends from the valleculae at the base of the tongue to the lower border of the cricoid cartilage and contains the UES.

The muscle groups participating in deglutition are those of the soft palate, pharyngeal isthmus, tongue, and hyoid bone—pharyngeal constrictors and muscles that elevate and displace the pharynx in a forward direction (Fig. 9-5). They may be classified as extrinsic muscles, which are responsible for altering the shape of the pharynx and closing the airways, and intrinsic muscles, which are responsible for collapsing the lumen of the pharynx and propelling the bolus.

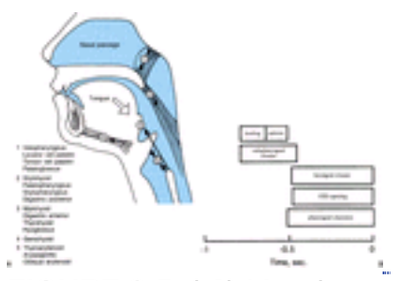

FIGURE 9-5. (**A**) Muscle groups participating in deglutition may be classified as extrinsic muscles, which are responsible for altering the shape of the pharynx and closing the airways, and intrinsic muscles, which are responsible for collapsing the lumen of the pharynx and propelling the bolus. The extrinsic muscles, including the levator veli palatini, tensor veli palatini, and palatoglossus, are located in the nasopharynx; they raise and tense the soft palate and uvula and close the nasal passage, to prevent pressure generated in the mouth from being dissipated through the nose. The stylohyoid, palatopharyngeus, stylopharyngeus, digastric posterior, and other muscles located posteriorly cause elevation, and the geniohyoid, mylohyoid, digastric anterior, thyrohyoid, and other muscles located anteriorly cause forward displacement of the larynx and pharynx and contribute to opening the upper esophageal sphincter (*UES*). The thyroarytenoid, aryepiglottic, and oblique arytenoid muscles and others close the larynx to prevent food from entering the trachea. (**B**) During swallowing, a rapid series of events occurs: closure of the velopharyngeus to prevent reflux into the nose; closure of the larynx to prevent aspiration; pharyngeal peristalsis to clear the bolus out of the pharynx; laryngeal upward and forward displacement to move the larynx out of the path of the bolus and to force open the cricopharyngeal region; and opening of the UES. The *graph* depicts the temporal sequence of some of the events occurring during swallowing of a 1-mL liquid bolus. Time zero represents the end of the swallow, determined by the occurrence of UES closure. Velopharyngeal closure occurs as the bolus is gathered on the tongue (i.e., loading) and propelled forward (i.e., pulsion). Laryngeal closure and UES opening occur later, during the phase of pharyngeal clearance. (Adapted from ref. [78].)

The extrinsic muscles, including the levator veli palatini, tensor veli palatini, palatoglossus, and others, are located in the nasopharynx; with the palatopharyngeus, they raise and tense the soft palate and uvula and close the nasal passage, thus preventing pressure generated in the mouth from being dissipated through the nose. When selective paralysis of these muscles occurs, as in poliomyelitis, the bolus sometimes is pushed into the nasopharynx. [81] The stylohyoid, styloglossus, palatopharyngeus, stylopharyngeus, digastric posterior, and other muscles located posteriorly cause elevation, and the geniohyoid, mylohyoid, digastric anterior, thyrohyoid, and other muscles located anteriorly cause forward displacement of the larynx and pharynx and contribute to opening the UES. Activation of these groups of muscles causes negative pressure to develop in the hypopharynx.

During the pharyngeal phase of swallowing, the area located ahead of the bolus exhibits an incrementally decreasing pressure caudally, which may facilitate bolus

transport and may contribute to airway protection. [82] The combination of the negative pressure in front of the bolus and the positive pressure caused by the tongue, palate, and proximal pharynx behind the bolus imparts a powerful propulsive movement, resulting in high-speed injection of the bolus into the esophagus. [83], [84] When swallowing occurs, the thyroarytenoid, aryepiglottic, oblique arytenoid, and other muscles close the larynx, thus preventing food from entering the trachea.

The intrinsic pharyngeal muscles are the superior, middle, and inferior pharyngeal constrictors (see Fig. 9-4). These muscles overlap like tiles on a roof and insert into a collagenous sheet, called the *buccopharyngeal aponeurosis.* In the upper pharynx, this structure is attached to the prevertebral fascia by a median raphe; distally, the constrictors are vertically mobile in relation to the prevertebral fascia and allow considerable axial movement during swallowing. [85]

The inferior pharyngeal constrictor has two anatomic components: the thyropharyngeus, which extends posteriorly from the thyroid cartilages and overlaps the middle constrictor, and the cricopharyngeus. The cricopharyngeus consists of a horizontal muscle loop surrounding the esophageal inlet, constituting part of the UES, and an oblique component on its posterior and proximal portion that attaches to the median raphe. This arrangement creates a thin, triangular area in the posterior aspect of the hypopharynx, called the *Killian triangle.* This area is structurally weak and susceptible to failure, producing outpouching of the mucosa through the muscular layer, called a *Zenker diverticulum.* [86] The term *cricopharyngeus muscle* commonly refers to the horizontal portion of the cricopharyngeus. The UES consists of the horizontal cricopharyngeus muscle and portions of the inferior constrictor. The sphincter is part of the pharynx. Some evidence suggests that muscles of the proximal esophagus do not contribute to UES function. [84], [85], [86], [87] and [88]

Pharyngeal Function and Control

Mastication of food, mixing it with saliva, and forming a bolus of a size and consistency appropriate for swallowing occur in the mouth under voluntary control. The deglutitive functions of the tongue include bolus containment, volume accommodation, and bolus propulsion. The tongue's graded actions are capable of accommodating boluses of different volumes and are responsible for increasing intrabolus pressure for vigorous bolus expulsion. [78] Tongue propulsive force and the clearing pressure during the swallow modulate bolus viscosity. The anterior two thirds of the tongue shows both greater force development and greater modulation than the tongue base. [89] Based on evidence obtained from simultaneous pressure measurements and cinefluorography, it has been suggested that the driving pressure produced by the tongue and the negative pressure generated in the pharyngoesophageal segment contribute significantly to the driving force of the bolus. When these components are absent or reduced in patients with tongue impairment or who have undergone laryngectomy, the transit of a food bolus is impaired. [83]

The presence of a bolus in the oropharynx activates a variety of receptors located at the base of the tongue, tonsils, soft palate, uvula, posterior pharyngeal wall, and larynx. [16], [90], [91] The afferents carrying the input to the swallowing center are thought to run in the maxillary branch of the trigeminal nerve, the glossopharyngeal nerve, and the superior laryngeal branch of the vagus nerve. [92], [93] The pharyngo-UES contractile reflex is activated by pharyngeal mucosal mechanoreceptors whose afferent limb is the glossopharyngeal nerve and whose efferent limb is the pharyngoesophageal branch of the vagus nerve. [94] The initial response to injection of water into the pharynx is inhibitory, and it can block ongoing primary and secondary peristalsis in the esophageal body. [95] Continuation of deglutition in the pharynx and striated esophagus is under involuntary reflex control by motor nuclei in the swallowing center. The swallowing center that controls the deglutition reflex is located in or near the NTS in the brainstem. [16], [96], [97]

Afferent mucosal receptors, sensitive to pressure and taste, are present through the pharynx and larynx. The epiglottal edge appears to be the most sensitive trigger zone for swallowing. [98] Afferent receptors activate glossopharyngeal and vagal fibers that innervate the swallowing center. Central recognition of the incoming sensory stimulus is thought to be accomplished by a pattern recognition system in the brainstem that identifies the stimulus as appropriate for swallowing and generates the required neuromuscular response. [97], [99] The swallowing center then initiates a pattern of excitation and inhibition of medullary motor neurons; it interacts with other brainstem centers and coordinates cessation of breathing, initiation of swallowing, opening of the UES, and peristalsis in the striated portion of the esophagus. [90], [100], [101]

Sensory loss in the oropharyngeal isthmus, pharynx, or larynx that may be caused by local anesthesia often is associated with impairment of swallow or aspiration, suggesting that proper coordination of swallowing and respiration may depend in part on the appropriate input from receptors located in the pharynx or larynx. [102], [103] The sensory afferents are connected to motor nuclei in the brainstem by interneurons. Discrete motor input is distributed to all muscle groups in the pharynx, larynx, and soft palate through motor fibers in the glossopharyngeal and vagus nerves, except for the tensor veli palatini, which is innervated by the trigeminal nerve. The axons of the motor neurons to the pharyngeal muscles run through the vagi into the superior pharyngeal nerves, arising from the vagi at the level of the nodose ganglion. These nerve fibers form the pharyngeal plexus and control the pharyngeal muscles, including the cricopharyngeus and the upper portion of the esophagus.

Videofluoroscopic analysis of the pharyngeal phase of swallowing reveals rapid succession or almost simultaneous occurrence of multiple component actions: laryngeal closure that results from elevation of the larynx; nasopharyngeal closure that results from the elevation of the soft palate; UES opening associated with sphincter relaxation and traction of the anterior wall resulting from laryngeal elevation and contraction of the suprahyoid and infrahyoid muscles; bolus propulsion accomplished by the actions of the tongue; pharyngeal clearance that results from the longitudinal shortening and elevation of the larynx; and transverse contractions, which are aborally propagated and responsible for the emptying of any residue. Compromising any of these components can cause dysphagia. [104]

Longitudinal contractions, observed early in the swallow, are caused by contraction of the stylopharyngeus muscle, which changes the conformation of the pharynx and transforms it from a respiratory to a deglutitive organ. They approximate the UES to the base of the tongue. Transverse contractions occur late in the swallow and are aborally propagated as the bolus tail travels the length of the pharynx. This peristaltic action is performed entirely by the posterior wall of the pharynx because the anterior wall is composed of cartilaginous structures that are devoid of any electromyographic activity. The speed of this highly complex function varies with the volume of the bolus. It is progressively slower with increasing bolus volumes. Only the duration and amplitude of contraction of the pharyngeal constrictors associated with pharyngeal clearance remain constant regardless of bolus size. [105]

The oropharyngeal swallow accommodates a range of bolus volumes. Volume accommodation is accomplished by augmenting and prolonging reconfiguration from a respiratory to deglutitive pathway. [106] When bolus volume is increased, more time is allocated for reconfiguration, that is, sustained laryngeal elevation and hyoid excursion. In terms of bolus expulsion volume, accommodation is accomplished within the same period by using increased vigor of expulsion. [105]

The descending wave of pharyngeal peristalsis begins and continues through the open UES. Within 1 to 2 seconds, the velopharyngeus closes to prevent reflux into the nose, the larynx closes to prevent aspiration, pharyngeal peristalsis clears the bolus out of the pharynx, and the larynx is displaced upward and forward to move it out of the path of the bolus and to force open both the cricopharyngeal region and the UES. [107]

UPPER ESOPHAGEAL SPHINCTER

Functionally, the UES is defined as a high-pressure zone, 2 to 4 cm wide, separating the pharynx from the body of the esophagus. It is composed of both opening and closing muscles. The opening muscles include the thyrohyoid and the geniohyoid, and the closing muscles include the cricopharyngeus, thyropharyngeus, and those of the cervical esophagus. The relative contribution of each muscle to the opening and closing of the UES varies with the physiological state. Although the cricopharyngeus may be the primary muscle of the UES, it cannot account for all the observed functions of the UES. [108] There is some uncertainty about the muscles responsible for maintaining the high pressure, because the width of the horizontal cricopharyngeus accounts for only 1 to 1.5 cm of the UES. It appears, however, that the UES high-pressure zone encompasses the cartilaginous hypopharynx, including the cricoid cartilage anteriorly and the cricopharyngeus and other components of the inferior pharyngeal constrictor. [16], [79], [109], [110] Data obtained from animal studies show good correlation between intralumenal pressure and electromyographic activity in the cricopharyngeus and inferior pharyngeal constrictor. [87]

The UES or pharyngoesophageal segment maintains closure of the proximal end of the esophagus, [111] and it constitutes an additional barrier to refluxed materials entering the pharynx (Fig. 9-6). It also prevents air from entering the esophagus, because intraesophageal pressure during inspiration is lower than pharyngeal pressure. In humans, the UES exhibits an asymmetric pressure profile longitudinally and radially. [112], [113], [114], [115] and [116] Peak pressure is highest in the pharyngoesophageal segment, lower in the inferior constrictor area, and lowest at the level of the tongue base. [116] Radial asymmetry is considerable in the UES, with the highest pressures occurring in an anteroposterior direction and peak pressures occurring at slightly different locations in the anterior and posterior directions. In the anterior direction, the peak pressure occurs 1 cm distal to the upper border of the high-pressure zone, and in the posterior direction, it occurs 2 cm distal to the upper border of the high-pressure zone. [115] The asymmetry disappears in patients who have undergone laryngectomy, suggesting that this feature may be caused by squeezing of the esophagus against the laryngeal cartilages forming the anterior wall of the sphincter. [115] The asymmetry also may result from different mechanical

constraints at different levels of the pharynx, or it may be a reflection of the neural control of swallowing in the brainstem. [116]

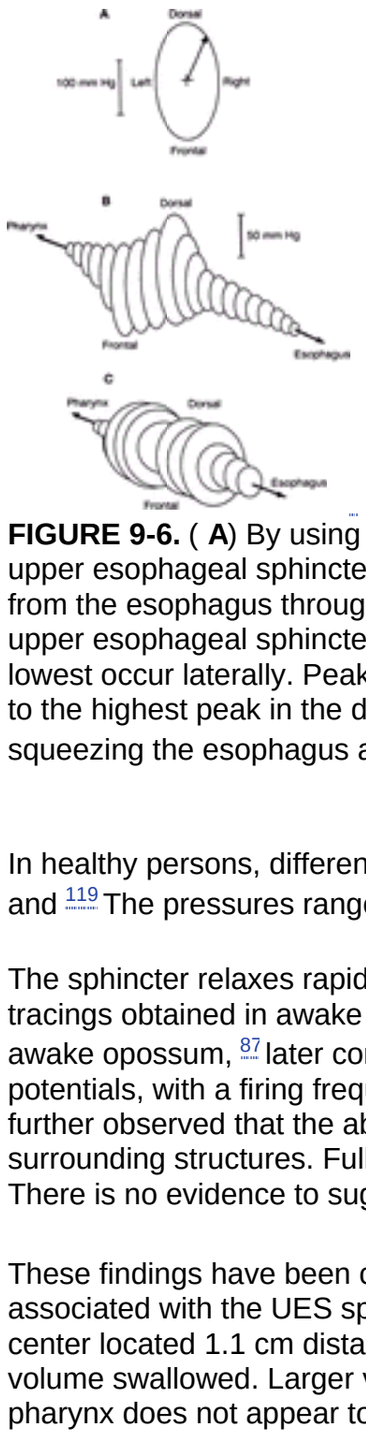

FIGURE 9-6. (**A**) By using multiple pressure sensors oriented in different directions, it is possible to measure pressure as a function of the radial orientation within the upper esophageal sphincter. In this sphincter, pressure is highest in the frontal and dorsal directions and is lowest in the lateral directions. (**B**) As the probe is moved from the esophagus through the sphincter and into the pharynx, a three-dimensional representation of the sphincter's pressure profile is obtained. In humans, the upper esophageal sphincter exhibits an asymmetric pressure profile longitudinally and radially. The highest pressures occur in an anteroposterior direction, and the lowest occur laterally. Peak pressures occur at slightly different locations in the anterior and posterior directions. The highest peak in the frontal direction occurs orad to the highest peak in the dorsal direction. (**C**) The asymmetry disappears in patients who have undergone laryngectomy, suggesting that it may be caused by squeezing the esophagus against the laryngeal cartilages, thus forming the anterior wall of the sphincter. (Adapted from ref. [115].)

In healthy persons, different resting pressures have been recorded by different investigators and by the same investigators using different techniques. [111], [115], [117], [118] and [119] The pressures range between 40 and 100 mm Hg, with pressures in the lateral orientations as low as 30% of the pressures in anterior or posterior orientations.

The sphincter relaxes rapidly and exhibits significant longitudinal excursions during swallowing that cause considerable confusion in the interpretation of pressure tracings obtained in awake persons. In experimental animals, anesthesia affects the spike potentials associated with contraction of striated muscles. Studies of the awake opossum, [87] later confirmed in studies of the cat, [120] showed that the cricopharyngeus and the inferior pharyngeal constrictor exhibit continuous electrical spike potentials, with a firing frequency proportional to the resting tone present in the sphincter and relaxation occurring after abolition of the spike potential. The studies further observed that the abolition of spike potentials reduced resting tone only by half, because some passive pressure was present as a result of the elasticity of the surrounding structures. Full relaxation occurred on contraction of the geniohyoid muscle, which caused forward displacement of the larynx and forced open the UES. There is no evidence to suggest that esophageal fibers distal to the cricopharyngeus are involved in maintenance of sphincter closure.

These findings have been confirmed in humans using a combination of videofluoroscopy and appropriate intralumenal manometry. [121] The high-pressure zone associated with the UES spanned about 3 cm, but the width of the high-pressure zone with pressure greater than 25% of peak pressure averaged 1.6 cm, with its center located 1.1 cm distal to the vocal cords. On swallowing, the UES moved orad 2 to 3 cm, with the extent of movement and opening size dependent on the volume swallowed. Larger volumes are accommodated by greater axial excursions, wider openings, and prolonged opening intervals. Peristaltic velocity in the pharynx does not appear to be affected by bolus volume.

Deglutition evokes an almost immediate UES relaxation, which briefly precedes pharyngeal contractions and, within 2 seconds, relaxes the LES. LES relaxation is observed as the peristaltic contraction appears within the middle third of the esophagus. [122] The UES relaxes briefly (<1 second) and fully to the level of pharyngeal or atmospheric pressures.

Neural input to the UES is required for maintenance of high resting pressure and for precise coordination of relaxation with swallowing. In the opossum, sphincter tone is mediated through neural fibers in the vagal trunks that originate in the nucleus ambiguus of the medulla. [123] Vagal transection abolishes activity in the cricopharyngeus and inferior pharyngeal constrictor muscles. [87] These muscles normally exhibit continuous spike activity, which is abolished by curare. Tone alters with changing firing frequency, indicating that motor neuron activity is responsible for maintenance of tonic muscle activity. Cessation of firing causes relaxation. Swallowing is further facilitated by forward displacement of the pharynx, which forces the sphincter to open. [87]

Resting tension in the UES is influenced by breathing, because it increases during inspiration when thoracic and esophageal resting pressures decrease. [124], [125] The change in pressure cannot be the result of transmission of pressure from the body of the esophagus but probably results from reflex neural activity. In the opossum, this increase in resting pressure is associated with increased spike activity in the cricopharyngeus and persists until stage 3 anesthesia, at which point, activity of UES muscles is reduced or abolished. [87] UES pressure is reduced during sleep and increases in persons under acute emotional stress. [124], [126]

Intrabolus pressure is an important determinant of UES opening in the healthy oropharynx. The healthy UES has residual opening capacity that can be demonstrated by altering body posture. These findings were confirmed in humans by using a combination of videoradiography and intralumenal manometry. Hypopharyngeal intrabolus pressure increases significantly in the horizontal compared with the upright position, resulting in increased maximal sphincter diameters during bolus flow, shorter duration of sphincter opening, and increased transsphincteric flow. [127]

Distention with small air-filled balloons or injection of water or acid into the body of the esophagus causes an increase in UES pressure. [128] The increase is greater when the bolus is closer to the UES and is greater for acid than for water or saline in healthy persons. [129] In infants with gastroesophageal reflux, the response to acid perfusion is not different from that of healthy age-matched controls, but in patients with esophagopharyngeal regurgitation, neither saline nor acid causes a significant increase in UES pressure. [130], [131]

Increased UES pressure, in response to reflux, [132] has been observed in patients with esophagitis and in healthy volunteers. Mechanisms responsible for this reflex were examined in the dog. [133] In this species, UES pressure increases in response to acid perfusion and to balloon distention of the proximal esophagus. The response to acid increases with decreasing pH and with proximity of the distending balloon to the UES. The response to intraesophageal acid perfusion at all levels within the esophagus is abolished by bilateral blockade of the vagosympathetic trunks. The response to balloon distention of the distal esophagus is abolished, but the response to distention of the proximal esophagus is only reduced. These data suggest that the afferent pathways for the response to acid are found exclusively in the vagosympathetic trunks, presumably arising from branches of the recurrent laryngeal nerves. Responses induced by distention, at least in the proximal esophagus, may partially depend on another pathway.

Injection of air into the body of the esophagus or distention by large balloons relaxes the UES. [134] The relaxation facilitates expulsion of the air bolus by belching. The afferent mechanisms mediating discrimination between distention by fluid, which causes contraction, or by air, which causes relaxation, are unknown. It is possible that the spatial pattern and rapidity of esophageal distention may provide discrimination in the response to fluid or air. [134] When refluxed contents reach the upper esophagus, the UES prevents further retrograde movement. [135] During deep sleep, however, the UES pressure decreases, possibly resulting in a reduced ability to prevent reflux at a time when the risk of reflux is higher. [124]

UES relaxation occurs during deglutition, belching, or vomiting. Relaxation during swallowing has two components. The first is inhibition of neural input arising from motor neurons in the brainstem and resulting in cessation of spike activity in the muscles of the UES. [87], [136], [137] The second is forceful opening of the UES, resulting from elevation and forward displacement of the larynx by muscles of the oropharynx. The styloglossus, stylohyoid, palatopharyngeus, stylopharyngeus, digastric

posterior, and other muscles located on the posterior aspect of the pharynx presumably contribute to elevation, and the geniohyoid, mylohyoid, digastric anterior, thyrohyoid, and other muscles located anteriorly contribute to forward displacement. Inhibition of tonic muscle activity by itself is not sufficient to cause relaxation, and forceful opening of the sphincter is required to abolish resting pressure.

Pharyngeal peristalsis may force the bolus through the sphincter even in the absence of forceful opening; conversely, displacement of the larynx may force the UES open, even in the absence of cricopharyngeal inhibition. Impairment of relaxation or opening of the sphincter may affect the smooth coordination of swallowing. Impairment of cricopharyngeal relaxation results in a prominent bar across the cricopharyngeal region (i.e., cricopharyngeal achalasia), and paralysis of the suprahyoid pharyngeal muscles may cause paralytic achalasia of the UES. [16]

ESOPHAGUS

Esophageal Anatomy

The body of the esophagus is about 18 to 25 cm long, extending from the inferior border of the UES to the upper border of the LES. The length of the esophagus correlates with body height and usually is longer in men than in women. It has a longitudinally oriented muscle layer in the muscularis mucosae and two muscle layers in the muscularis propria, with the inner one oriented along the circumference (i.e., circular layer) and the outer one along the axis (i.e., longitudinal layer).

Although the muscle in the muscularis mucosae is longitudinally oriented and smooth throughout the whole length of the esophagus, the muscularis propria is composed of striated muscle in the most proximal portion. Esophageal striated muscle has histological characteristics different from those of other striated muscles. These differences include the type of motor innervation and endplates, arrangement of the muscle fibers, fiber diameter, and type of myosin adenosine triphosphatase. [17], [138], [139] and [140]

It is generally accepted that the junctional area between the striated and smooth muscle occurs in the middle third of the esophagus. Data suggest that, on average, the proximal 4.1 cm of the circular and 5.1 cm of the longitudinal layers are entirely striated muscle. The point at which the circular layer is about half smooth and half striated muscle occurs 4.7 cm distal to the cricopharyngeus. [141] A 4- to 8-cm transition area (i.e., middle third of the esophagus) has various proportions of striated and smooth muscle; the distal 10 to 14 cm consist exclusively of smooth muscle whose fibers are oriented in a circular or an elliptical direction. [142] This distribution is typical of the human esophagus, but there are considerable species differences in the relative proportions of striated and smooth muscle. A distribution similar to that in humans occurs in other primates, cats, and opossums. The pig esophagus is predominantly striated muscle, with a short smooth muscle portion. In dogs and sheep, the esophagus is entirely striated. In some species, such as the rat, the striated muscle portion includes the LES. [143] Knowledge of the muscle composition of the esophagus is crucial for the interpretation of physiological and pathophysiological data.

Esophageal Function

Cinefluoroscopic studies of barium swallows have demonstrated that as an ingested bolus enters the body of the esophagus (Fig. 9-7), it travels by gravity or peristaltic propulsion, depending on its consistency. Liquids travel by gravity if the subject is upright. Liquids fall fast enough (within 2 seconds) to find the LES closed, causing distention of the distal esophagus and creating the radiological appearance of the phrenic ampulla. After the peristaltic contraction reaches the liquid bolus, it pushes it through the relaxed LES. When the subject is supine, the column of barium travels just ahead of the peristaltic wave. Small amounts of fluid may remain in the distal esophagus after the peristaltic wave.

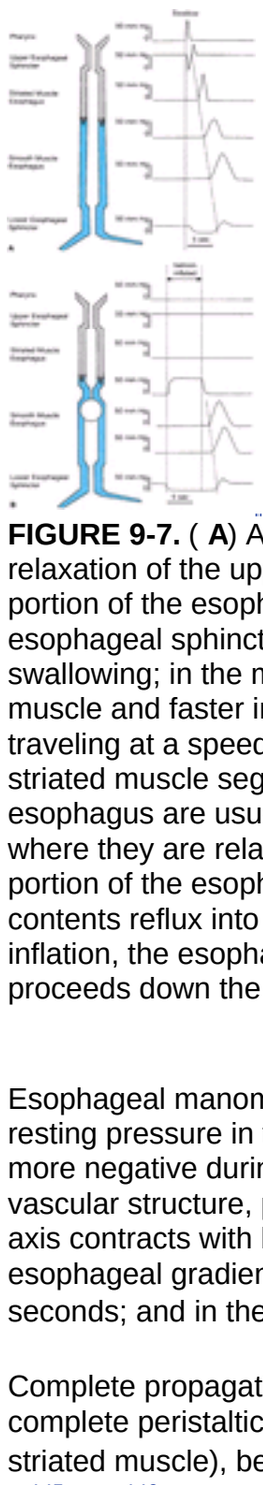

FIGURE 9-7. (**A**) At rest, the normal esophagus is quiescent, without any spontaneous contractions, and the sphincters are tonically contracted. Swallows trigger relaxation of the upper and lower esophageal sphincters and give rise to a peristaltic contraction traveling smoothly through the striated and then the smooth muscle portion of the esophagus. Each location along the esophageal axis contracts with a latency that increases gradually from the upper esophagus to the lower esophageal sphincter. The latencies are site dependent and reproducible. In the upper third of the esophagus, contraction occurs within 1 to 2 seconds after swallowing; in the middle third, within 3 to 5 seconds; and in the lower third, between 5 and 8 seconds. The velocity of the peristaltic wave is slower in the striated muscle and faster in the smooth muscle segments of the esophagus. Contractions reach the smooth muscle segment within 2 seconds after the onset of the swallow, traveling at a speed of about 3 cm per second; in the smooth muscle segment, the velocity of propagation may be as fast as 5 cm per second. The contractions in the striated muscle segment are shorter (1–2 seconds), and in the smooth muscle segment, they are longer (4–7 seconds). Contractions in the distal one third of the esophagus are usually stronger (50–150 mm Hg) than those in the upper third (40–120 mm Hg), and both are stronger than those in the middle third (20–80 mm Hg), where they are relatively weak, probably occurring at the transition between the striated and the smooth muscle. (**B**) A similar sequence occurs in the smooth muscle portion of the esophagus during secondary peristalsis, which happens when a bolus remains in the esophagus after ineffective primary peristalsis or when gastric contents reflux into the esophagus. Secondary peristalsis is thought to be caused by distention and can be demonstrated by inflating a balloon in the esophagus. On inflation, the esophagus contracts proximal to and relaxes distal to the balloon, including the lower esophageal sphincter. When the balloon is deflated, peristalsis proceeds down the esophagus.

Esophageal manometry shows that the normal esophagus at rest is quiescent without any spontaneous contractions, but the sphincters are tonically contracted. The resting pressure in the esophagus is an approximate reflection of thoracic negative pleural pressures. These pressures vary with the respiratory cycle and become more negative during inspiration. Occasionally, a 1- to 3-cm-long high-pressure zone is present in the middle esophagus, resulting from extrinsic compression by a vascular structure, possibly the aorta. Swallows trigger peristaltic contractions traveling along the entire length of the esophagus. Each location along the esophageal axis contracts with latency that increases gradually from the upper esophagus to the LES. The latencies are site dependent and reproducible, constituting the esophageal gradient. In the upper third of the esophagus, contraction occurs within 1 to 2 seconds after swallowing; in the middle third, it occurs within 3 to 5 seconds; and in the lower third, it occurs between 5 and 8 seconds. [144]

Complete propagation of the peristaltic contraction takes place after a swallow if a second swallow does not occur within at least 5 seconds after the first one. The complete peristaltic sequence is inhibited if a second swallow occurs within 5 seconds or less; a contraction may appear in the upper third of the esophagus (i.e., striated muscle), because it may occur before the second swallow is taken, but the peristaltic sequence is always inhibited in the lower two thirds of the esophagus. [144], [145] and [146] After repeated swallows at short intervals, no contractions are observed in the entire esophagus until after the last swallow, which is followed by a high-amplitude contraction. The inhibition of peristalsis by repeated swallows has been called *deglutitive inhibition.* Deglutitive inhibition maintains esophageal atony during guzzling, (i.e., taking repeated swallows at very short intervals). The second swallow affects the motor response to the first; the first swallow may also impair the response to the second if the two swallows are taken at short intervals. [147] Weak swallows may not evoke a full esophageal response; they may induce LES relaxation without esophageal contractions. [148] Occasionally, even in healthy persons, deglutition may induce simultaneous contractions within a 5- to 10-cm segment.

The velocity of the peristaltic wave is slower in the striated muscle and faster in the smooth muscle segments of the esophagus. Contractions reach the smooth muscle segment within 2 seconds after the onset of the swallow, traveling at a speed of about 3 cm per second. In the smooth muscle segment, the velocity of propagation may be as fast as 5 cm per second.

Motility studies indicate that the size and viscosity of an ingested bolus affect the speed and force of the esophageal contractions. Increasing the size of the bolus slows its velocity and increases the force of the contractions. Increased viscosity also increases the force of contraction and decreases the speed of peristalsis. [149], [150] Peristalsis also is affected by the temperature of the bolus; warm boluses increase its velocity, and cold boluses reduce it. [151]

Each contraction is preceded by a slight fall in pressure, probably related to alterations in the respiratory pattern during swallowing. [152] Pressures then rise rapidly, usually peaking within 1 to 2 seconds from the onset of the contraction. Normal esophageal contractions have spikelike configurations, particularly in the striated muscle segment of the esophagus (i.e., upper third), but occasionally they may be biphasic. The configuration of the pressure wave may depend on whether the ingested bolus is liquid or solid, but it usually exhibits an initial hump or plateau before rising rapidly. The initial pressure change may reflect the pressure created in the esophagus by the presence of the bolus, preceding the peristaltic wave. Dry swallows reveal that the contractions in the striated muscle segment are shorter (1–2 seconds) than in the smooth muscle segment (4–7 seconds). Contractions in the distal one third of the esophagus are usually stronger (50–150 mm Hg) than those in the upper third (40–123 mm Hg), and both are stronger than those in the middle third (20–80 mm Hg), where they are relatively weak, perhaps because of the transition between striated and smooth muscle. The force of the contractions can vary from segment to segment and even from swallow to swallow. [153]

Segmental peristaltic contractions may occur to prevent the injurious effect of reflux by clearing the esophagus of any refluxed contents. This response is similar to secondary peristalsis, which occurs as result of distention by food, gas, and acid. It is not clear whether local esophageal distention is always necessary to trigger esophageal clearing, because acid alone may evoke peristaltic contractions. [154] In addition, acid increases the perception of esophageal distention. [155]

Control of Esophageal Striated Muscle

Esophageal electromyographic responses can be evoked by stimulation of either the right or left hemisphere, with the largest amplitude responses obtained anterior to those of the thenar eminence, indicating that the esophagus may be represented on either the anterior aspect of the motor cortex or on the premotor cortex. These studies suggest that esophageal motor function shows bilateral cortical representation with consistent interhemispheric asymmetry. [156]

Contraction of esophageal striated muscle depends entirely on neural input arising from neurons located in the nucleus ambiguus and is centrally organized by sequential discharge of motor neurons controlling progressively distal segments in the body of the esophagus (Fig. 9-8). [1], [2], [157] Motor fibers course along the vagus and separate in the upper portion of the neck. These cholinergic fibers release ACh and stimulate nicotinic cholinergic receptors on the motor endplates of the striated muscle fibers. Bilateral vagotomy high in the neck, above the origin of the pharyngoesophageal fibers, completely abolishes peristalsis in this portion of the esophagus, but peristalsis in the smooth muscle portion can still be initiated by local distention. [16] Unilateral vagotomy has no effect, and transection of the striated muscle segment does not affect the normal progression of the peristaltic contractions evoked by swallowing. Peristalsis in the striated muscle portion of the esophagus does not depend on the continuity of esophageal plexuses but is under central nervous control. [158], [159]

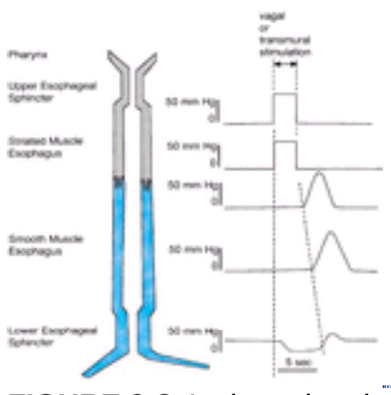

FIGURE 9-8. In the striated muscle portion of the esophagus, peristalsis is controlled directly by the central nervous system, and sequencing of peristalsis occurs in the swallowing center. Motor neurons are activated sequentially, first those innervating the proximal striated esophagus and then those activating the more distal portion of the striated muscle. In the opossum, after division of the vagus at the cervical level, stimulation of the distal end, which activates simultaneously all vagal fibers, produces simultaneous contraction of the striated esophagus and relaxation of the lower esophageal sphincter. At the end of the stimulus, peristalsis occurs in the smooth muscle esophagus. This suggests that, although peristalsis in the striated esophagus is under central nervous system control, local mechanisms are responsible for peristalsis in the smooth muscle portion. Even in the smooth muscle portion, central input may regulate peristalsis. Two types of vagal fibers may participate in smooth muscle peristalsis. Short-latency fibers, firing within 1 second of swallowing, coincide with the onset of inhibition in the esophagus and lower esophageal sphincter and may stimulate neurons that release inhibitory neurotransmitters. Long-latency fibers, discharging their impulses within 1 to 5 seconds, may coincide with the onset of contraction at various locations in the esophagus. These fibers may activate motor neurons to release excitatory neurotransmitters. It is possible that, on swallowing, the central nervous system mediates initial inhibition throughout the esophagus by way of a nonsequential mechanism activating short-latency fibers that results in hyperpolarization in the esophagus and relaxation of the lower esophageal sphincter. As hyperpolarization ends, sequential excitatory input through the long-latency fibers causes sequential contraction and peristalsis in the smooth muscle esophagus.

Other types of esophageal responses in the striated muscle portion also appear to depend on central control, including secondary peristalsis, the inhibition evoked by frequent swallows, and reversed peristalsis in ruminants. [160] Extrinsic denervation or bilateral cervical vagotomy eliminates the esophageal contractions evoked by esophageal distention in this segment. Frequent swallows taken at short intervals abolish the response of the striated muscle of the esophagus until the last swallow occurs. [146] Because the striated muscle is under direct central control, swallow-induced inhibition probably originates in the swallowing center.

The strength of contraction in the striated muscle may be modulated by a variety of sensory inputs, including bolus volume and temperature. [151], [161], [162] An increase in volume augments the force and duration of the contractions while slowing the velocity of propagation. Warm boluses increase the force, and cold boluses decrease the force of the contractions. These effects may depend on afferent input to the central nervous system with subsequent modulation of vagal control of the striated esophageal fibers. [163]

Control of Esophageal Smooth Muscle

Control of peristalsis in the smooth muscle segment of the esophagus (see Fig. 9-8) is influenced by sequencing of motor neurons in the swallowing center, resulting in progressive stimulation of intramural cholinergic neurons. [164] Central sequencing alone, however, cannot account for the occurrence of sequential peristaltic contractions in response to stimulation of the decentralized efferent vagus at the cervical level. [165] Long latencies in response to field stimulation have been observed, even in vitro in muscle strips taken from the distal esophagus, when compared with strips from the proximal esophagus, which exhibit shorter latencies. [166]

Control of peristalsis also may arise from peripheral mechanisms located in the intramural plexuses, because stimulation of decentralized vagal efferent nerves evokes peristalsis that is similar to the peristaltic wave produced by swallowing. [165], [167] Nerve conduction studies indicate that neural stimuli initiated by swallowing propagate with a speed of 5 to 6 cm per second and therefore reach the intramural neurons of the entire length of the esophagus at about the same time, but the timing of the contractions reveals an aboral increment in latency of contraction during peristalsis. [168] In vitro distal esophageal strips exhibit longer latencies than proximal strips. [166], [169] Esophageal transection at the level of the smooth muscle segment markedly affects the propagation of the peristaltic wave below the level of the transection, but transection of the striated esophagus does not affect the orderly transmission of peristalsis across the cut, suggesting that the integrity of the

intramural mechanisms is essential for the normal progression of peristalsis in the smooth but not in the striated esophagus. [159]

Peripheral control is illustrated also by distention of the esophageal smooth muscle segment after extrinsic denervation. [62], [170] Peristalsis induced by balloon distention always is propagated in the caudad direction after extrinsic denervation with cervical vagotomy and even in the isolated esophagus mounted in an organ bath. Esophageal peristalsis evoked by balloon distention is similar to that caused by swallowing, with a similar sequence of inhibition or hyperpolarization during distention followed by a latency period and contraction or depolarization after balloon deflation. Repeated stimulation of the decentralized efferent vagal fibers at short intervals inhibits the esophageal responses, with contraction occurring only after the last stimulus. [171] This experimental model mimics deglutitive inhibition and suggests that the mechanism responsible resides in the intramural neurons.

Primary peristaltic waves in this segment are affected by the presence of a bolus and by central mechanisms. The bolus modifies the speed, duration, and force of the contractions. [172] Central mechanisms are also capable of modulating the occurrence and features of esophageal contractions, including the polarity of peristalsis, as occurs under some pathophysiological conditions. Changing the parameters of vagal stimulation can induce peristaltic or antiperistaltic sequences at different speeds of propagation. [165]

The preganglionic fibers that mediate this control branch out from the thoracic vagus nerve, to form a plexus around the esophagus, and then enter into the esophageal wall, where they travel a few centimeters before synapsing with the myenteric neurons. [173], [174] They arise from the dorsal motor nucleus and may synapse to the intramural neurons. [2], [3] During swallowing, central regulation may be mediated by two types of vagal fibers. Short-latency fibers firing within 1 second may stimulate neurons that release inhibitory neurotransmitters, and long-latency fibers discharging their impulses within 1 to 5 seconds may activate motor neurons to release excitatory neurotransmitters. [175], [176]

Central mechanisms also control the contractions of the longitudinal muscle layer. Swallowing induces peristaltic sequences with gradual activation from orad to caudad segments and a correspondent progressive increase in latencies. [177] Unlike the responses observed in the circular muscle layer, stimulation of decentralized vagal efferent fibers causes simultaneous contractions in the longitudinal muscle layer. [165] In the human esophagus, both circular and longitudinal muscle contractions occur as propagating segments during peristalsis, with the longitudinal muscle contraction leading the circular muscle contraction. Propulsive force occurs during proximal circular and distal longitudinal muscle contraction. [89]

Esophageal peristalsis is characterized by a gradient, with increasing latency of contractions along the length of the esophagus. The latent period is the interval between the swallow or electrical stimulus and the onset of contraction at a given site in the esophagus. In humans, it is 2 seconds in the proximal smooth muscle esophagus and 5 to 7 seconds just above the LES. [144] The nature of the latency gradient is controversial, but it can be changed by varying the parameters of vagal stimulation or by pharmacological manipulation of the esophageal contractions, suggesting that latency may result from the interaction between the initial inhibition and subsequent excitation of esophageal smooth muscle. [165] The muscarinic cholinergic antagonist atropine increases the latency of contraction, and the acetylcholinesterase inhibitor physostigmine decreases it. [176], [178]

The data support the view that peristalsis is largely determined by neurally dependent esophageal gradients, because the esophageal response to low-frequency electrical stimuli is tetrodotoxin sensitive and is aborally transmitted no matter where in the esophagus the stimulus is applied. [165]

Preganglionic vagal fibers synapse with intramural excitatory neurons through activation of nicotinic receptors. The nicotinic ganglionic inhibitor hexamethonium blocks cervical vagal stimulation. [167] The nature of the postganglionic intramural neurons varies with the animal species studied. It has been suggested that these neurons are exclusively inhibitory, using NANC transmitters. Stimulation of these neurons results in an initial inhibition, followed by a rebound contraction after cessation of the stimulus. [179] Studies in the opossum and cat suggest that the inhibition occurring during the stimulus and the “off” contraction occurring at the end of the stimulus are mediated by separate transmitters, if not by different neurons, although considerable species-related differences occur.

Swallowing causes a single contraction at each level of the esophagus after a given latency. Brief stimulations (1 second) of the efferent vagal fibers or of circular muscle strips also evoke a single contraction with a given latency [178]; however, the esophageal response induced by prolonged stimulation (5–20 seconds) is dissociated into an “on” contraction that occurs during the stimulus and an “off” contraction after the end of the stimulus. Because the act of swallowing is brief, it is conceivable that the on and off responses represent dissociations of one or more excitatory neurons during artificially prolonged stimulation. The on responses are not seen at lower stimulus frequencies, which are thought to be more representative of physiological events. [178] It is unclear how the results of these experimental studies may relate to the esophageal response evoked by deglutition.

In the opossum, cholinergic and NANC excitatory neurons are responsible for the off response; proximal segments are controlled primarily by cholinergic fibers, and distal segments are controlled by noncholinergic motor neurons. [178] In the cat esophagus, the off response seems to be entirely under cholinergic control. The contractions induced by field stimulation in vitro or by vagal stimulation are blocked by atropine. [144], [179], [180] Because atropine decreases the amplitude of pressure generated by swallowing and the force developed in vitro by esophageal muscle in response to electrical (i.e., neural) stimulation, [181], [182] and [183] cholinergic fibers also contribute to the human esophageal contractions.

The intramural neural pathway that mediates the descending peristaltic reflex induced by balloon distention in the opossum involves long descending neurons that depend on NO as a final mediator. [184] NO is produced from L-arginine by the enzyme NO synthase, which is present in myenteric neurons. [185], [186] NO synthase is reversibly inhibited by some analogs of L-arginine, including *N*-nitro-L-arginine methyl ester and *N*-nitro-L-arginine. NO synthase inhibitors decrease the latency and amplitude of off contractions in the in vitro esophageal circular muscle [187] and in the in vivo esophagus in response to swallowing. [45], [188] *N*-nitro-L-arginine dose dependently reduces the propagation time of contraction in the distal esophagus. [189] In vivo, this effect is more pronounced in the distal esophagus, where latency is longer. NO synthase inhibitors reduce the latency gradient so contraction in response to swallowing occurs almost simultaneously in the smooth muscle portion of the esophagus. In vivo infusion of recombinant human hemoglobin (rHb1.1), which inactivates NO by binding with it, interferes with esophageal peristalsis, LES relaxation, and precipitates esophageal spasm. [190], [191] and [192] These studies suggest that endogenous NO may mediate the coordinated latency between swallowing and progressive contraction along the esophageal body that is responsible for peristalsis. [45], [190], [191], [193], [194]

The hypothesis that esophageal smooth muscle response may result from more than one neurotransmitter is supported by the finding that it is possible to dissociate esophageal inhibition and esophageal excitation by using selective antagonists. The initial inhibition may be partially mediated by VIP-containing neurons, which are present in the body of the esophagus. [195] Esophageal muscle strips tonically contracted with bethanecol relax during field stimulation; this neurally mediated (i.e., tetrodotoxin-sensitive) relaxation is partially blocked by VIP antiserum without affecting the force and duration of the off contractions, which are blocked by atropine. [144] In addition, NO colocalizes with such neuropeptides as VIP, CGRP, and galanin, but it shows little colocalization with neuropeptide Y and substance P, suggesting that the nerves responsible for peristalsis in the esophagus may act by releasing NO along with other inhibitory neuropeptides, but not excitatory substances, such as neuropeptide Y and substance P. [75], [77]

Esophageal muscle is relaxed at rest and contracts in response to vagal or field stimulation with a brief and forceful contraction. In the cat, this contraction is mediated by M_2 muscarinic receptors [196] linked to a pertussis toxin–sensitive guanosine triphosphate (GTP)-binding protein of the G_{i3} type and results in the activation of at least three phospholipases acting on membrane phospholipids. Phosphatidylcholine-specific phospholipase C and phospholipase D hydrolyze phosphatidylcholine to produce diacylglycerol without the production of inositol phosphates. [197], [198] A cytosolic phospholipase A_2 (PLA_2) [199], [200], [201] and [202] is also activated, resulting in the production of arachidonic acid. [203] Activation of these phospholipases requires the presence of Ca^{2+}, which is provided by the influx of extracellular Ca^{2+} through voltage-dependent channels, [204] possibly augmented by the release of Ca^{2+} from intracellular stores. [205], [206] Once diacylglycerol is produced, the Ca^{2+}-independent protein kinase C (PKC_e) is activated and contraction proceeds through a calmodulin-independent pathway that may not be directly regulated by activation of myosin light chain kinase. [207], [208] and [209] Arachidonic acid, produced by PLA_2, which is also Ca^{2+}-dependent, potentiates the diacylglycerol-induced activation of PKC and contributes to the initiation of PKC activity and ultimately to the strength of contraction (Fig. 9-9). [203]

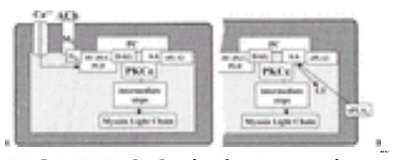

FIGURE 9-9. (**A**) Normal esophagus. Contraction of esophageal circular muscle in response to maximally effective doses of the cholinergic neurotransmitter acetylcholine (*ACh*) is mediated through muscarinic M_2 receptors linked to pertussis toxin–sensitive G proteins of the G_{i3} type. It results in activation of at least three phospholipases acting on the membrane phospholipid phosphatidylcholine (*PC*). Phosphatidylcholine-specific phospholipase C (*PC-PLC*) and phospholipase D (*PLD*) hydrolyze PC to produce diacylglycerol (*DAG*) without production of inositol phosphates. A 100-kd cytosolic phospholipase A_2 (*cPLA* $_2$) is also activated, resulting in the production of arachidonic acid (*AA*). Activation of these phospholipases requires the presence of Ca^{2+}, which may be provided by the influx of extracellular Ca^{2+} through voltage-dependent channels or by release from Ca^{2+} stores. AA produced by PLA_2 potentiates the DAG-induced activation of protein kinase C (*PKCe*), contributing equally to the initiation of PKC activity and to the strength of contraction. (**B**) Esophagitis. The roles of PLA_2 and AA and its metabolites are altered by the inflammatory responses associated with the induction of experimental esophagitis by repeated esophageal acid perfusion. In normal esophageal circular muscle, AA, produced by a $cPLA_2$, directly contributes to ACh-induced contraction by potentiating DAG-induced activation of PKC, and AA metabolites such as prostaglandins and leukotrienes (*LTs*) do not play a major role in contraction. After the induction of acute experimental esophagitis, the amplitude of response to ACh in cells or muscle strips of esophageal muscle is not altered greatly, but contraction is mediated in part by a low-molecular-weight (14-kd) secreted PLA_2 ($sPLA_2$). $sPLA_2$ and $cPLA_2$ differ in several respects, including molecular weight, substrate specificity, and localization within an organism. In addition, after esophagitis, AA is metabolized to LTs; because resting levels of LTs are greater than in normal circular esophageal muscle, LT levels increase in response to ACh, and ACh-induced contraction is reduced by LT antagonists. In animals with esophagitis, LTs contribute to contraction by activating LT-selective membrane receptors and by potentiating the DAG-induced activation of PKC.

K^+ channels play a role in maintaining the membrane resting potential. Some K^+ channels are active at or near the resting membrane potential and may participate in maintaining membrane polarization. [210], [211] Others are activated at more positive potentials as the membrane is depolarized and may contribute to ending depolarization and restoring the resting membrane potential. K^+ channels in many tissues are targets for modulation by inhibitory as well as excitatory factors. [212], [213] Agents that cause relaxation of esophageal muscle activate K^+ currents and contribute to hyperpolarization. For example, NO activates a K^+ current in the opossum esophagus, contributing to inhibitory junction potentials, [214], [215] and K^+ channels are activated by cyclic AMP (cAMP)-dependent pathways. [216] In addition, K^+ currents are targets for suppression by excitatory pathways. Cholinergic excitation suppresses spontaneous outward K^+ currents in esophageal muscle, [217] and it inhibits some cloned smooth muscle K^+ currents. [218]

Chloride ($C1^-$) channels may play a role in inducing depolarization. [219], [220] It has been suggested that agonist receptor binding and G-protein activation induces arachidonic acid formation by activation of a cytosolic PLA_2. Arachidonic acid in turn causes opening of $C1^-$ channels and efflux of $C1^-$ from the cell, resulting in depolarization, opening of voltage-sensitive Ca^{2+} channels and Ca^{2+} influx into the cell. Ca^{2+}-influx stimulates the production of cyclic adenosine diphosphate–ribose, which acts directly on ryanodine-sensitive Ca^{2+} channels in the endoplasmic reticulum and causes Ca^{2+} release. [221] $C1^-$ channels are another site of action for NO, which inhibits $C1^-$ efflux. [220]

The roles of PLA_2 and arachidonic acid and its metabolites are altered by the inflammatory responses associated with experimental esophagitis [222] induced by repeated esophageal acid perfusion. [223] In normal esophageal circular muscle, arachidonic acid produced by a 100-kd cytosolic PLA_2 contributes to ACh-induced contraction by potentiating diacylglycerol-induced activation of PKC; arachidonic acid metabolites such as prostaglandins and leukotrienes do not play a major role in contraction. [203] After induction of acute experimental esophagitis, the amplitude of response to ACh in cells or muscle strips of esophageal muscle [224] is not greatly altered, but contraction is mediated in part by a low-molecular-weight (14-kd) secreted PLA_2 ($sPLA_2$). [222] $sPLA_2$ and cytosolic PLA_2 are different in several respects, including molecular weight, substrate specificity, and localization within an organism. [225], [226] After induction of esophagitis, arachidonic acid is metabolized to leukotrienes, as demonstrated by the following observations: resting levels of leukotrienes are greater than in normal circular esophageal muscle, leukotriene levels increase in response to ACh, and ACh-induced contraction is reduced by leukotriene antagonists. In animals with esophagitis, leukotrienes contribute to contraction by activating leukotriene-selective membrane receptors [227] and by potentiating the diacylglycerol-induced activation of PKC (see Fig. 9-9).

LOWER ESOPHAGEAL SPHINCTER

Anatomy

An anatomic sphincter distinct from the body of the esophagus is found in human autopsies and consists of an asymmetric, thickened, ringlike structure that angles obliquely upward from the lesser to the greater gastric curvature. The length of the thickened area along the lesser curvature is about 2.5 cm and about 3 cm along the greater curvature. The LES proper is composed of complete muscle rings. Distally, the rings are split into two segments: one straddles the greater curvature and is parallel to the sling fibers of the stomach, and the other consists of short clasps straddling the lesser curvature and connecting at an angle to the sling fibers. [228] The squamocolumnar junction is present within this structure. In similar studies on the esophagogastric junction of cats, a clear correlation was found between the manometrically defined high-pressure zone and the thicker segment of circular muscle, with the squamocolumnar junction slightly distal to the high-pressure point. [229] The thickness of the circular muscle increases from the esophagus toward the LES, peaks at the in vivo high-pressure zone, and then decreases toward the stomach.

Electron microscopic studies reveal that the sarcolemma of the LES muscle has more evaginations than that of the esophageal muscle. [230], [231] It is unclear whether this morphologic feature has functional significance or whether it results from the tonically contracted state of the muscle. More abundant mitochondria and a more developed endoplasmic reticulum also differentiate the LES muscle from the circular muscle of the esophageal body. [231]

Basal Pressure

At rest, the LES is tonically contracted. The contraction is maintained by muscular mechanisms, even in the absence of neural input, [232], [233] and [234] and it creates an effective barrier consisting of a 3- to 5-cm-long high-pressure zone that effectively separates the esophagus from the stomach. The skeletal muscle crural diaphragm appears to contribute to the pressure barrier. [235] The pressure in the high-pressure zone ranges from 12 to 30 mm Hg above gastric pressures and exhibits radial and axial asymmetry, which may be attributed in part to extrinsic compression by the crural diaphragm. [236] The highest pressures are usually at the point of respiratory reversal, where the esophagus crosses the diaphragm. [237] This point separates the thoracic esophagus, where pressures decrease with inspiration, from the abdominal esophagus, where pressures increase with inspiration because the descending diaphragm compresses the abdominal contents.

LES pressures are affected by respiratory excursions and may be augmented by diaphragmatic contraction. [238], [239] Mid- or end-expiratory pressures are an indication of LES strength. The end-expiratory pressure is a result of the tonic activity of the smooth muscles of the LES. The pressure at the esophagogastric junction increases during inspiration, owing to the effect of the crural diaphragm. There is a reflex contraction of the LES during periods of increased intra-abdominal pressure, and the crural diaphragm contributes to this reflex. [235], [240] The end-expiratory pressure in the resting state is caused by the contraction of the LES, whereas the additional increase in pressure during inspiration is caused by the crural diaphragm. [238], [241] These two sphincters seem to relax simultaneously during swallowing, belching, and vomiting. [242] Gastroesophageal reflux secondary to transient LES relaxation is also accompanied by the simultaneous relaxation of the LES and the crural diaphragm. [240], [243], [244]

Extrinsic support of the LES by the crural diaphragm may be important, and loss of this support through development of hiatus hernia may contribute to LES dysfunction. The diaphragmatic and LES smooth muscle components normally supplement each other to maintain competence in static conditions and during dynamic stresses associated with increased intra-abdominal pressure or swallowing. These sphincteric components also interact with each other pathophysiologically. Hiatus hernia reduces LES pressure and alters the dynamic responsiveness of the LES by spatially separating pressure components derived from the intrinsic LES and the extrinsic compression of the esophagus within the hiatal canal. In patients with hiatus hernia, the esophagogastric junction high-pressure zone has two discrete segments, one proximal to the squamocolumnar junction and one distal, attributable to the extrinsic compression within the hiatal canal. Inspiration and abdominal

compression augments pressure mainly in the distal segment. [236], [245]

LES smooth muscle exhibits spontaneous tone and myoelectric activity that is characterized by continuous spike-burst activity with or without short phasic contractions. [112], [246] Spikes occur at a frequency of 15 to 40 per minute and may be associated with minicontractions, 5 to 15 mm Hg in amplitude. Partial relaxation of the LES is accompanied by complete cessation of electrical activity, and there is substantial residual LES tension that is not accompanied by any detectable electrical activity. Phasic contractions and electric spike activity become prominent after the administration of bethanecol or during phases II and III of the migrating myoelectric complex. [247] Phase III of the migrating myoelectric complex begins in the LES, lasts for about 15 minutes, and is characterized mostly by an increase in short phasic contractions. [247] The control mechanisms have not been established completely, but they appear to be under vagal control and are associated with a rise in circulating levels of the peptide motilin. Feeding, barbiturates, and cholinergic antagonists abolish electrical activity and associated contractions.

The composition of a meal also affects LES pressure; it is lower after a fatty meal and higher after a protein meal. [248] In the anesthetized cat, distal esophageal acidification causes an increase in LES pressure. [249] The physiological role of this response is unclear; it may be designed to tighten the sphincter to prevent further gastroesophageal reflux.

Gastrointestinal hormones or regulatory peptides may affect in vivo LES pressure, but it is difficult clearly to demonstrate a physiological role for many of them because of the lack of selective antagonists and because, when infused at concentrations similar to the circulating levels, many have little effect on the LES. The pharmacological effects of these hormones and endogenous compounds are well established, including some of their mechanisms of action. Gastrin causes LES contraction by direct muscle action. [167], [250] Other hormones, such as motilin, bombesin, pancreatic polypeptide, galanin, and substance P, cause LES contraction when used in pharmacological doses. [251], [252], [253], [254] and [255] In contrast, secretin, CCK, glucagon, gastric inhibitory peptide, VIP, peptide histidine isoleucine (PHI), CGRP, and neurotensin decrease LES pressures, although significant species differences occur. [256], [257], [258], [259], [260], [261], [262], [263] and [264] For instance, CCK causes LES relaxation in humans and in cats and causes LES contraction in the opossum. [257], [263], [264] It causes relaxation by stimulating the intramural NANC inhibitory neurons and causes contraction by direct muscle action. [263], [264] Glucagon relaxes the human LES, probably by direct action, but it contracts the cat LES by releasing catecholamines from the adrenal medulla. [256]

Some experiments suggest a physiological role for selected hormones. Intravenous infusion of neurotensin in humans at or below the serum levels obtained postprandially causes a dose-dependent decrease in pressures and may involve a cholinergic pathway. [265] Neurotensin levels increase after intragastric infusion of fat, and neurotensin antibodies reduce the decrease in LES pressure induced by fat infusion, suggesting that neurotensin may participate in this pathway. [266] Increases in the circulating levels of pancreatic polypeptide correlate with a rise in LES pressures after a protein meal and are abolished by duodenal exclusion from the meal. [253] Somatostatin has no effect on basal LES pressures but blocks the postprandial rise in LES pressures. [267] These observations suggest that a meal-induced rise in LES pressures is mediated by hormonal mechanisms, and it is known that somatostatin inhibits hormonal release. [268], [269] Gastroduodenal hormones may play a physiological role in regulating LES pressures during the digestion of certain foodstuffs (e.g., fatty meals, chocolate, alcohol, coffee) and during changes in gastroduodenal pH (e.g., acid or alkaline solutions), which may decrease or increase LES pressures in humans. [270], [271]

Basal LES pressures in vivo or basal circular muscle tension in vitro are maintained even in the absence of neural input. [232], [234], [272], [273] Bilateral cervical vagotomy and denervation with the neural poison tetrodotoxin do not eliminate in vivo LES tension in most animal species; in the opossum, LES pressures do not change at all. [234] In vivo experiments, however, usually are performed in anesthetized animals. General anesthesia depresses LES pressure in the baboon, suggesting that the neural input may be reduced or abolished in some in vivo experiments. [274] Some evidence indicates the presence of a neural contribution to the genesis or modulation of LES pressures in several animal species. Atropine reduces LES pressures in conscious humans and dogs, but similar doses of atropine do not affect LES pressures in conscious opossums and monkeys or in anesthetized opossums and cats. [178], [234], [272], [274], [275] and [276] Atropine has no effect on the tone of human LES smooth muscle strips, [277] a finding suggesting that cholinergic modulation of LES pressure may be of central origin. The precise contribution of cholinergic input to LES pressure is difficult to estimate because resting pressures may be modulated by excitatory and inhibitory influences. A decrease in tonic excitatory cholinergic input may enhance the tonic inhibitory effect. In the dog, however, LES pressures clearly are dependent on vagal tonic activity. [278], [279] In the cat, some adrenergic input contributes to LES pressure, and sympathetic nerves also mediate a reflex contraction. [272] The a-adrenergic blocking agent phentolamine decreases LES pressures by 25%, and a similar reduction is seen after tetrodotoxin administration. [272] Some of the controversies surrounding the significance of neural input in the genesis or modulation of LES pressures may reflect species differences.

The LES circular muscle is a major determinant of LES tone, although the relative neurogenic contribution may vary with the animal species. Functionally, this muscle is distinguished by the ability to maintain sustained tonic contraction in vivo and in vitro, even after the removal of neurogenic contributions by the administration of high doses of tetrodotoxin. [232], [233] and [234] Functional differences in esophageal and LES muscle may be related in part to the presence of different contractile proteins in the two types of muscle. [280] Maintenance of tone also may be related to the sustained activity of a pancreatic-like PLA_2 (Fig. 9-10). [281], [282]

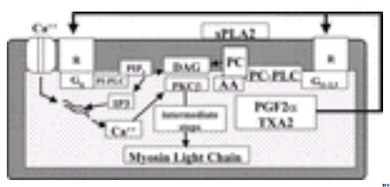

FIGURE 9-10. Resting lower esophageal sphincter (LES) tone is associated with the spontaneous activity of phosphatidylinositol-specific phospholipase C (*PI-PLC*) and phosphatidylcholine-specific phospholipase C (*PC-PLC*) resulting in the production of inositol 1,4,5-trisphosphate (IP_3) and diacylglycerol (*DAG*) and the activation of a Ca^{2+}-sensitive protein kinase Cß (*PKCß*). A low-molecular-weight (14-kd) secreted, pancreatic-like (group I) phospholipase A_2 (*sPLA* $_2$) may play a role in resting LES tone by producing arachidonic acid (*AA*), which is then metabolized to prostaglandin F_2a (*PGF* $_2a$) and thromboxane A_2 (*TXA* $_2$). PGF_2a and TXA_2 are membrane permeable and bind to specific G-protein–linked receptors, to cause activation of PI-PLC and PC-PLC, producing second messengers and contraction. G_q, $G_{i1-2,3}$, G proteins; PIP_2, phosphatidylinositol biphosphate; *R,* receptor.

The PLA_2 enzymes comprise a growing family of enzymes that catalyze the hydrolysis of glycerolphospholipids at the *sn*-2 position, producing free fatty acids and lysophos- pholipids. [283], [284] and [285] These enzymes regulate phospholipid-acyl turnover for membrane repair or for the production of arachidonic acid. Specific cellular processes, including the regulation of PKC and phospholipase C-? and the modulation of Ca^{2+} transients, are controlled by arachidonic acid. This fatty acid is also the precursor to biologically active lipids including prostaglandins, leukotrienes, thromboxanes, and platelet activation factor.

The presence of an immunoreactive group I–like $sPLA_2$ in the human LES circular muscle layer has been demonstrated by Western blot analysis, [281] but the precise amino acid sequence of this enzyme remains to be determined. $sPLA_2$ may mediate LES tone through the production of arachidonic acid and its metabolites, such as prostaglandins F_2a and thromboxanes A_2/B_2, which are present in LES circular muscle. Selective group I $sPLA_2$ inhibitors and cyclooxygenase inhibitors reduce the basal tone of in vitro LES muscle strips and $sPLA_2$-induced contraction of isolated LES cells from cats and humans. [281], [282] These arachidonic acid metabolites maintain activation of G proteins such as G_{i3}, $G_{i1/2}$, and G_q. G_{i3} and G_q are coupled to phosphatidylinositol-specific phospholipase C and phosphatidylcholine-dependent phospholipase C. [196], [286] Low-level activities of these phospholipases produce threshold levels of the second messengers diacylglycerol and inositol 1,4,5-trisphosphate (IP_3). IP_3 and diacylglycerol, produced at submaximal levels, act synergistically; their interaction is dependent on Ca^{2+} release and is mediated through the sensitive PKCß, to maintain a PKCß-dependent basal tone. [208], [287], [288]

In contrast to spontaneous tone, contraction induced by maximally effective doses of the cholinergic neurotransmitter ACh is mediated through muscarinic M_3 receptors, linked to pertussis–toxin insensitive GTP-binding proteins of the G_q-G_{11} type. They activate phospholipase C, producing IP_3 and diacylglycerol. IP_3

causes the release of intracellular Ca $^{2+}$ and the formation of a Ca $^{2+}$-calmodulin complex, resulting in activation of myosin light-chain kinase and contraction through a calmodulin-dependent pathway (Fig. 9-11A). [287]

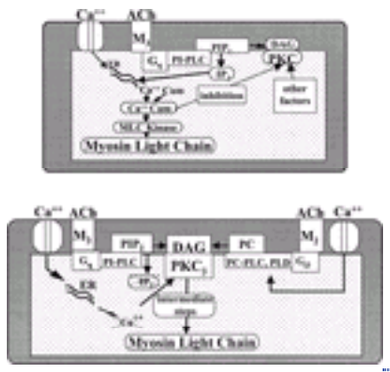

FIGURE 9-11. (**A**) Contraction of lower esophageal sphincter (LES) circular muscle in response to maximally effective doses of the cholinergic neurotransmitter acetylcholine (*ACh*) is mediated through muscarinic M $_3$ receptors, linked to pertussis toxin–insensitive G proteins of the G $_q$–G $_{11}$ type. They activate phospholipase C (*PLC*), which hydrolyzes phosphatidylinositol bisphosphate (*PIP* $_2$), producing inositol 1,4,5-triphosphate (*IP* $_3$) and diacylglycerol (*DAG*). *IP* $_3$ causes the release of intracellular Ca $^{2+}$ in levels sufficient to form a Ca $^{2+}$-calmodulin (*Ca* $^{++}$*Cam*) complex. The Ca $^{2+}$-calmodulin complex activates myosin light-chain (*MLC*) kinase, resulting in contraction by means of a calmodulin-dependent pathway. In addition, activated calmodulin may inhibit protein kinase C (*PKC*), eliminating any PKC-dependent contributions. (**B**) In a model of acute experimental esophagitis (*AE*) in the cat, resting in vivo LES pressure, spontaneous in vitro tone, levels of IP $_3$, and releasable intracellular Ca $^{2+}$ stores were reduced. Because the release of Ca $^{2+}$ from intracellular stores is reduced in AE, the available Ca $^{2+}$ may be insufficient to activate calmodulin; and thus, a PKC-dependent pathway may be unmasked that otherwise would be suppressed by calmodulin activation. After AE, the contribution of M $_3$ receptors, G $_{q/11}$, and phosphatidylinositol-specific PLC (*PI-PLC*) is reduced. M $_2$ receptors, G $_{i3}$-type G proteins, and phosphatidylcholine-dependent PLC and phospholipase D (*PC-PLC, PLD*) are activated and produce diacylglycerol, which, in turn, causes contraction through a PKC-dependent pathway. These changes in the functional signal transduction pathway are mimicked in normal LES muscle by acute depletion of intracellular Ca $^{2+}$ stores by thapsigargin and may therefore be related to the impaired release of Ca $^{2+}$ from intracellular stores, which arises from both impaired production of IP $_3$ and depletion of releasable Ca $^{2+}$ stores subsequent to induction of AE. *ER,* endoplasmic reticulum.

Therefore, unlike LES tone, which is associated with submaximal phospholipase C activity and activation of a PKCß-dependent pathway, maximal cholinergic stimulation activates a calmodulin-dependent pathway. The mechanisms responsible for a switch from a PKC-dependent to a calmodulin-dependent pathway are not entirely clear. They may result from the different Ca $^{2+}$ requirements of calmodulin and PKC. Lower Ca $^{2+}$ levels are required for PKC activation than for calmodulin activation. [286], [287], [289] For instance, relatively low (180 nmol/L) cytosolic Ca $^{2+}$ levels can support contraction induced by the PKC agonist diacylglycerol but not by calmodulin, which requires Ca $^{2+}$ levels approaching 1 µmol/L. [287] In addition, when Ca $^{2+}$ levels are elevated sufficiently to activate calmodulin, calmodulin may inhibit PKC. [290], [291] and [292] Thus, it is possible that at Ca $^{2+}$ levels insufficient to activate calmodulin, contraction will be PKC dependent. In contrast, at Ca $^{2+}$ levels sufficient to fully activate calmodulin, the contraction will be calmodulin dependent, and PKC activity will be inhibited.

These observations may be of some help in understanding the following changes reported in a model of acute experimental esophagitis in the cat. Repeated perfusion of the esophageal lumen with 0.1 N hydrochloric acid for 3 to 4 days cause a reduction in resting in vivo LES pressure, [293] spontaneous in vitro tone, levels of 1,4,5-IP $_3$, and releasable intracellular Ca $^{2+}$ stores. [223], [224], [289] Acute experimental esophagitis also causes a shift in the intracellular pathway mediating the response to a maximally effective dose of ACh from a calmodulin-dependent to a PKC-dependent pathway. [286] After acute experimental esophagitis, contraction induced by a maximally effective dose of ACh is mediated through M $_2$ muscarinic receptors, linked to G $_{i3}$-type G proteins, which activate phosphatidylcholine-dependent phospholipase C and phospholipase D to produce diacylglycerol. Diacylglycerol causes contraction through a PKC-dependent pathway (Fig. 9-11B). These changes in the functional signal transduction pathway are mimicked in normal LES muscle by acute depletion of intracellular Ca $^{2+}$ stores by thapsigargin. [286] Therefore, they are related to the impaired release of Ca $^{2+}$ from intracellular stores. This impairment arises from both impaired production of IP $_3$ [224] and depletion of releasable Ca $^{2+}$ stores [289] subsequent to the induction of acute experimental esophagitis. Because the release of Ca $^{2+}$ from intracellular stores is reduced in acute experimental esophagitis, the available Ca $^{2+}$ may be insufficient to activate calmodulin; thus, a PKC-dependent pathway is unmasked that would otherwise be suppressed by calmodulin activation. Depletion of releasable Ca $^{2+}$ by inflammation secondary to acute experimental esophagitis may be the central event from which all other observed changes follow.

The significance of these subchronic changes in the cat to the understanding of esophagitis in humans, in whom gastroesophageal reflux disease is likely to develop over a longer period, remains to be established. In a chronic model of esophagitis obtained by performing a myotomy of LES circular muscle, we find similar, if less accentuated, endoscopic, histological, and functional changes 6 months after surgery. [294] Metaplasia was present in some specimens 10 months after myotomy. [295] In addition, in the chronic model, the suppression of hydrochloric acid secretion, either after the onset of mild chronic esophagitis or at the time of myotomy, reverses or prevents the changes in smooth muscle signal transduction, presumably by inhibiting or preventing the injury caused by reflux. [294]

Relaxation

Transient Relaxation At times, particularly after meals, reflux of gastric contents may occur as a result of transient relaxations of the LES (TLESRs) [243] unrelated to swallowing or secondary peristalsis. [154], [296], [297] and [298] TLESRs occur in healthy persons, but they are more frequent in patients with gastroesophageal reflux disease. [154], [298] In healthy persons, TLESRs are most likely associated with expulsion of air from the stomach, [299], [300] whereas in patients, higher percentages of TLSERs are associated with reflux. [301], [302] In patients, TLESRs are the most common mechanism of reflux, accounting for the largest proportion of reflux episodes. [297], [298] The proportion of reflux episodes related to TLESRs, however, is greater in patients without endoscopic evidence of reflux esophagitis, in whom reflux occurs almost exclusively because of TLESRs, and it decreases with increasing severity of the disease. In patients who have severe esophagitis, the prevalence of reduced LES basal pressures probably plays a greater role in facilitating TLESR-unrelated reflux. [297], [303] Vagal pathways and the hindbrain control the occurrence and pattern of TLESRs, [304] although many aspects of these mechanisms are not well understood. [243], [305] TLESRs occur more frequently in the postprandial state when the stomach accommodates the meal by a receptive relaxation of the fundus; the resultant distention relaxes the LES, perhaps to facilitate belching. TLESRs may represent a silent belch. [299], [300] As the pH of gastric contents decreases, the probability of TLESRs increases. [306] In addition, pharyngeal stimulation, (e.g., by intubation), [307] which by itself may induce LES relaxation, increases TLESR frequency. [308], [309], [310], [311] and [312] Weak swallows also may evoke LES relaxations, without triggering an esophageal response in humans, [148], [311] or in the opossum, in which isolated and transient LES relaxations are also triggered by weak electrical stimulation of the vagus. [313] The latter non–swallow-associated relaxations, however, do not involve inhibition of the crural diaphragm, as occurs with swallow-induced relaxation [240] or TLESRs, nor do they involve the esophageal common cavity and acid reflux, and therefore, they may not be representative of proper TLESRs. [311] TLESRs triggered by gastric distention may use NO and CCK as neurotransmitters, because they are increased by intravenous CCK and are blocked by NO synthase inhibitors [314] and by CCK-A antagonists. [315], [316], [317] and [318] The frequency of TLESRs increases during intravenous infusions of CCK and after fatty meals. [319], [320] CCK infusion and fatty meals cause a rise in circulating CCK levels; the associated increase in TLESRs is mediated through CCK-A receptors, because CCK-A antagonists inhibit the increase in TLESRs. Muscarinic receptors may be involved in TLESRs, because TLESRs are also antagonized by atropine. [312], [321], [322], [323] and [324] GABA agonists, such as baclofen, inhibit TLESRs, [304], [325], [326] and [327] possibly by acting on receptors located in the DMV. [5], [328] It is thought that afferents located in the subdiaphragmatic vagus, particularly in its ventral branch, [14], [25], [243] initiate the non–swallow-induced relaxations that occur during belching, TLESRs, gastroesophageal reflux, and vomiting. [329] The afferent and efferent neural pathways responsible for swallow- and non–swallow-induced relaxations have been compared in the mouse. [329] The efferent limb of both swallow-associated and non–swallow-associated relaxations lies in the preganglionic vagal inhibitory pathway to the postganglionic nitrergic neurons in the LES. [315] Both relaxations can be blocked by bilateral cervical vagotomy, cervical vagal cooling, or chemical blockers of

neuronal NO synthase. [243], [330], [331] The afferent arms of these two types of relaxation, however, differ from one another. The afferent arm of the swallow-induced relaxation lies in the pharyngeal and superior laryngeal nerves, and the central neural circuit for the swallowing reflex is in the medullary subnuclei. [23], [26], [27], [101] Electrophysiological and tracer studies in the rat and larger animal species and c-*fos* expression in the mouse have further localized the brainstem circuit of the swallowing reflex to include interneurons in interstitial, intermediate, and central subnuclei, and motor neurons in the nucleus ambiguus and in the DMV. [332], [333] and [334] Non–swallow-induced relaxations, in contrast, are initiated through gastric afferents in the subdiaphragmatic vagus and activate neurons in the medial, dorsomedial, and commissural solitary subnuclei and motor neurons in the caudal part of the DMV. [329]

Swallow-Induced Relaxation The passage of an ingested bolus is expedited by LES relaxation and by the force of peristaltic contractions. Within the first 2 seconds after swallowing, LES resistance to bolus transit is caused by resting LES pressures. As the LES relaxes, it is passively forced open by the bolus that has been propelled by the peristaltic wave. Although accurate measurements are difficult because of motion artifacts resulting from longitudinal esophageal movement during swallowing, it is estimated that the LES relaxes fully. LES relaxation occurs within 2 seconds of deglutition, at a time when the peristaltic wave appears in the middle esophagus, at the beginning of the smooth muscle segment. Within 1 second, LES pressures fall to equal those of the stomach or esophagus, depending on the location of the pressure sensor within the LES. When ingested boluses reach the LES, it is relaxed but closed. The bolus forces the LES open by its own weight or with the aid of the peristaltic contraction. Deglutition induces an initial inhibition of the entire smooth muscle of the esophagus, and LES relaxation is part of this inhibitory response. After 5 to 7 seconds, the LES recovers its initial pressure and then undergoes an aftercontraction, which probably represents the end of the peristaltic pressure wave as it reaches the distal end of the esophagus. [149] Circular muscle strips obtained from the LES proper exhibit relaxation only, but more proximal strips reveal relaxation followed by an after-contraction, perhaps because of the presence of mixed LES and esophageal muscle fibers. [229] The inhibitory innervation that mediates LES relaxation on swallowing or as a result of gastroesophageal distention has been studied extensively. [179], [335], [336] LES relaxation induced by swallowing is mediated by the vagus nerve, which synapses with intramural inhibitory neurons. Electrical stimulation of decentralized efferent vagal fibers in vivo and field stimulation of LES muscle strips in vitro cause full relaxation, with concomitant circular muscle hyperpolarization. [112], [337] The vagus nerve uses ACh as a ganglionic transmitter acting through nicotinic and muscarinic receptors, because transmission is blocked by a combination of hexamethonium (i.e., nicotinic blocker) and atropine (i.e., muscarinic blocker). LES relaxation can be triggered by distention from both sides of the esophagogastric junction. [338] This relaxation induced by esophageal distention is volume dependent; it is brief during inflation with small volumes but persists during inflation with large volumes. [339], [340] The relaxation is unaffected by cervical vagotomy and therefore is mediated by intramural neurons. The intramural neurons of these reflexes use NANC inhibitory neurotransmitters, and relaxation is not affected by adrenergic or cholinergic blockers. It is only antagonized by tetrodotoxin, which denervates smooth muscle without affecting its contractile function. [341] Convincing evidence supports a role for NO as a neurotransmitter responsible for LES relaxation. NO is produced by NO synthase from the amino acid L-arginine. Three isoforms of NO synthase derived from three different genes have been identified: neuronal (type I), inducible (type II), and endothelial (type III). Inducible NO synthase is not present under physiological conditions; it is induced during inflammation and tissue injury. Neuronal and endothelial forms of NO synthase are constitutively present and play a role under physiological conditions. Neuronal NO synthase is a soluble cytosolic enzyme present primarily in nerves and is the source of NO that is involved in neurotransmission, whereas endothelial NO synthase is particulate and membrane bound and is present mainly in endothelial cells and smooth muscle cells. [342] Endothelial NO synthase produces NO that is involved in producing endothelial-dependent relaxing factor, [343] and it has been proposed to play a role in neuromuscular transmission. [344] The enzyme NO synthase has been identified in neurons of the myenteric plexus and has been found to be colocalized with VIP, which may be a second inhibitory neurotransmitter in the LES as well as in the esophageal body. NO is released on neural stimulation in the esophagus, LES, and stomach. [76], [185], [345], [346] and [347] In the LES, NO has a marked inhibitory effect. Many studies performed in vitro and in vivo have shown that inhibition of NO synthesis by NO synthase inhibitors results in blockade of neurally mediated LES relaxation, and NO-induced LES relaxation is associated with an increase in cyclic guanosine monophosphate (cGMP), as occurs during neural stimulation. [45], [187], [310], [348], [349], [350] and [351] Conclusive evidence that neuronal NO synthase plays a role in LES relaxation has been obtained by comparing wild-type and genetically engineered, neuronal NO synthase–deficient mice. LES ring preparations developed spontaneous tone in all animals. In wild-type mice, electrical field stimulation produced frequency-dependent relaxation, which was abolished by the NO synthase inhibitor *N*(omega)-nitro-L-arginine methyl ester, whereas in neuronal NO synthase–deficient mice, the relaxation was absent. [352] Evidence has been presented in support of VIP as one of the neurotransmitters of these neurons. VIP-containing neurons have been demonstrated in the submucosal plexus by immunofluorescent methods. [68], [71], [353], [354] VIP relaxes the LES in vivo and in vitro by direct muscle action. [256], [355], [356] Electrical stimulation of in vitro LES muscle strips relaxes the LES and releases VIP in the muscle bath, and VIP antiserum partially reduces the LES relaxation induced by VIP and evoked by vagal or field stimulation. [356], [357] There is some evidence that PHI in the cat and, to a lesser extent, CGRP in the opossum also may participate as inhibitory neurotransmitters. [358], [359] Like VIP, PHI and CGRP relax the LES fully or in part by a direct action on the muscle. [358], [359] and [360] PHI is of some interest because it is derived from the same precursor as VIP and coexists with VIP in the same neurons. [361] PACAP-27– and PACAP-38–immunoreactive nerve structures have also been found in the cat and the human LES, with an abundance in the circular smooth muscle layer. PACAP-27 immunoreactivity often was colocalized with VIP immunoreactivity. [74] PACAP, however, has a less potent relaxant action than VIP, and its functional importance remains to be established. It is thought that VIP released from nerve terminals acts on neural terminals to release NO and on gastric muscle cells to stimulate production of NO by the muscle. [344], [362], [363], [364] and [365] These observations have been confirmed in part by studies of mutant, NO synthase–deficient mice, which suggest that VIP is a prejunctional neurotransmitter that causes NO release by activating a "neuronal" NO synthase. [366] The finding of a small amount of NO synthase in intestinal muscle has provided further confirmation. [367] The notion that multiple neurotransmitters may interact to produce LES relaxation may resolve inconsistencies and discrepancies that follow from the assumption that any one neurotransmitter is the only one responsible for LES relaxation. Reports of the colocalization of NO synthase, VIP, PACAP, CGRP, and galanin in the myenteric neurons of the distal esophagus support this concept. [46], [368], [369] Several investigations suggest that carbon monoxide also may contribute to LES relaxation. Carbon monoxide is generated by heme oxygenase isoenzymes in the degradation of heme- containing molecules. In the LES and other sphincters, heme oxygenase is present in neuronal and nonneuronal cells, and it is colocalized with VIP and NO synthase in myenteric neurons. [370] Similar to NO, carbon monoxide reduces spontaneous LES tone by increasing cGMP but not cAMP levels. Carbon monoxide may contribute to VIP-, PHI-, and PACAP-induced relaxations, because inhibition of the carbon monoxide–producing enzyme heme oxygenase causes a rightward shift for the relaxation responses to these peptides. [371] Anatomic evidence for the presence of a carbon monoxide–producing enzyme is strong, [371], [372], [373] and [374] whereas functional evidence is considerably weaker. Certain metalloporphyrins, which are inhibitors of heme oxygenase and have been widely used as pharmacological tools to establish a messenger role for carbon monoxide, also are associated with a large range of nonspecific effects. [375] For example, zinc protoporphyrin attenuates LES relaxation induced by VIP and other adenylate cyclase–mediated smooth muscle relaxants (e.g., isoproterenol). [376], [377] In summary, the esophagus functions as an active neuromuscular conduit that produces primary peristaltic contractions to transport a bolus from the pharynx to the stomach. Prevention of retrograde reflux of gastric contents is accomplished by maintaining tonically contracted LES and UES and by the clearance function of secondary peristalsis. Primary peristalsis is initiated by swallowing, which is voluntary. Its continuation is mediated reflexly by sensory neurons with input to the swallowing center located in the brainstem. The swallowing center initiates a pattern of excitation and inhibition of medullary motor neurons; interacts with other brainstem centers; and coordinates cessation of breathing, initiation of swallowing, opening of the UES, and peristalsis in the striated portion of the esophagus. Peristalsis in the smooth muscle segment of the esophagus depends on peripheral mechanisms in the intramural plexuses, which evoke peristalsis by activating inhibitory and cholinergic excitatory intramural neurons, but it also can be influenced by the sequencing of motor neurons in the swallowing center, resulting in progressive stimulation of intramural cholinergic neurons. The interplay between these inhibitory and excitatory neurons produces propulsive contractions that travel the entire length of the esophagus and LES relaxation. Whereas UES tone is dependent primarily on continued vagal input, specialized myogenic mechanisms contribute to the maintenance of LES tone.

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CHAPTER 10

William L. Hasler

PHYSIOLOGY OF GASTRIC MOTILITY AND GASTRIC EMPTYING

Motor activity of the stomach serves distinct roles under fasting and fed conditions. Interdigestive patterns clear the stomach of undigested debris and sloughed epithelial cells. After eating, the stomach accommodates the ingested bolus, which is then ground and dispersed into fine particles that are delivered to the duodenum at a controlled rate. The stomach can be divided into three functional regions: the proximal stomach (cardia, fundus, and proximal body), the distal stomach (distal body and antrum), and the pylorus. Coordinated actions of these regions with feedback control from the small intestine regulate the emptying of gastric contents. Gastric motor activity is controlled by myogenic properties, extrinsic and intrinsic innervation, circulating hormones, and external influences from the central nervous system.

SMOOTH MUSCLE CHARACTERISTICS OF THE STOMACH

The complex anatomic orientation of the gastric smooth muscle layers serves the needs of regulating nutrient dispersion and emptying. Located below the serosa, the longitudinal muscle layer is prominent in the distal stomach and is mostly continuous with the duodenal longitudinal layer, although some fibers terminate in the pylorus. The circular muscle layer, located below the longitudinal layer, is prominent in all gastric regions. The gastric circular layer is electrically isolated from its duodenal counterpart by a connective tissue septum at the pylorus. The innermost and least complete muscle layer, the oblique layer, is present on the lesser curvature near the cardia and is continuous with the gastroesophageal junction. Specialized pyloric smooth muscle is composed of two circumferential loops that coalesce over the lesser curvature. The pylorus is mainly circular smooth muscle, but it is reinforced by antral longitudinal muscle fibers and by connective tissue from the mucosa and smooth muscle.

INNERVATION OF THE STOMACH

Gastric smooth muscle is innervated by extrinsic nerves that relay information to and from the extragastrointestinal ganglia, the spinal cord, and the central nervous system, and by intrinsic nerves in the gastric wall. The extrinsic supply is provided by the vagus and splanchnic nerves, which contain both efferent and afferent fibers. The major intrinsic innervation projects from the myenteric plexus at the interface between the longitudinal and circular layers. The submucous plexus along the lumenal aspect of the circular layer may play a minor role in some reflex activities.

Efferent Extrinsic Innervation

Only a few vagal fibers provide efferent modulatory input to the gastric myenteric plexus. [1] However, immunohistochemical measurement of myenteric c-*fos* expression in rats indicates a widespread functional efferent vagal innervation of the enteric nervous system. [2] The vagus contains three groups of efferent fibers: preganglionic parasympathetic cholinergic nerves, which supply excitatory enteric neurons; preganglionic cholinergic nerves, which supply inhibitory enteric neurons; and sympathetic fibers from the superior cervical and stellate ganglia. [3] Cell bodies of most efferent vagal fibers reside in the dorsal motor nucleus of the vagus. [4] Stimulation of efferent vagal cholinergic neurons activates nicotinic receptors within enteric ganglia and increases gastric motor activity. Most efferent fibers exhibit a low threshold to electrical stimulation. Some efferent fibers that exhibit a high threshold to electrical stimulation inhibit motor activity through the release of nitric oxide (NO) and vasoactive intestinal polypeptide (VIP). [5], [6]

Sympathetic innervation from splanchnic nerves originates from neuronal cell bodies in the prevertebral celiac ganglia. Preganglionic cholinergic neurons project from the inferomedial spinal cord to the prevertebral ganglia, where they synapse through nicotinic receptors. Postganglionic noradrenergic neurons project to the enteric ganglia through the splanchnic nerves and generally inhibit excitatory myenteric transmission. [7] Smaller numbers of sympathetic fibers project to the smooth muscle, where they also have inhibitory motor effects.

Afferent Extrinsic Innervation

Afferent fibers outnumber efferent fibers by tenfold in the vagus and by threefold in the splanchnic nerves. Vagal afferents terminate in the nucleus tractus solitarius and area postrema. [8] Nucleus tractus solitarius neurons project to the dorsal motor nucleus of the vagus and nucleus ambiguus and to higher cerebral centers. [9] Sensory information from the stomach is also transmitted to the dorsal horn of the spinal cord through the splanchnic nerves, from where second-order neurons project centrally.

The stomach is richly supplied with sensory fibers (Fig. 10-1). Free mucosal nerve endings respond to stroking or to chemical stimuli such as hydrochloric acid. [10] Smooth muscle stretch receptors or mechanoreceptors are activated during passive distention or active contractions or if they are exposed to extreme temperatures. [11], [12] Two types of mechanoreceptors are evident. Intramuscular arrays serve as stretch receptors to mediate tonic activities, whereas intraganglionic laminar endings integrate tension and neuronal activity into propagative motor programs such as peristalsis and emptying. [13] Neurons in the nucleus tractus solitarius and the dorsal motor nucleus of the vagus are activated by gastric distention, indicating vagal transmission of sensory information from gastric mechanoreceptor activation. [14] Mesenteric and serosal receptors respond to tension on the viscera and forceful contractions and may mediate perception of visceral pain. In many gut regions, nociceptive sensory input is carried by the splanchnic nerves. Afferent pathways that mediate extended neural reflexes may be vagovagal, vagosplanchnic, or splanchnosplanchnic, and they are further modified by central nervous system input.

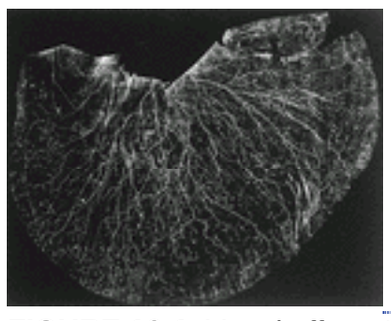

FIGURE 10-1. Vagal afferent innervation of the stomach. The fiber bundles enter from the lower esophageal sphincter (**top**) and radiate to the greater curvature. In the antrum (**upper right**), the gastric branch of the vagus courses across the pylorus to innervate the proximal duodenum. (From ref. [13].)

Intrinsic Innervation of the Stomach

The enteric nervous system possesses the components needed for complete reflex activities, including afferent neurons, interneurons, and motor neurons, and it can initiate many physiological motor patterns in the absence of extrinsic input. Myenteric neurons contain numerous neurotransmitters. Excitatory neurons containing acetylcholine and tachykinins (i.e., substance P and neurokinin A) project to circular muscle. Inhibitory motor neurons containing NO and VIP project aborally and mediate the relaxation phase of peristalsis. [15], [16] The actions of NO are mediated by stimulation of cyclic guanosine monophosphate–dependent kinase, whereas VIP acts through cyclic adenosine monophosphate generation. [17] Serotonin serves a modulatory role in enteric neurotransmission. In some species, excitatory ascending pathways are activated by serotonin (5-hydroxytryptomine, 5-HT) acting on 5-HT $_3$ receptors, whereas by occupying 5-HT $_{1P}$ receptors, serotonin can produce descending inhibition. [18] Other relevant modulators include norepinephrine, endogenous opioids, adenosine triphosphate, peptide histidine isoleucine, ?-aminobutyric acid, neuropeptide Y, gastrin-releasing peptide (GRP), and histamine. [16], [19], [20], [21] and [22]

Both stimulatory and inhibitory neurons in the myenteric plexus interact with the interstitial cells of Cajal in the circular muscle of the proximal gut. [23] Interstitial cells of Cajal are histologically distinct from neurons and smooth muscle cells. In mutant animals deficient in intramuscular interstitial cells, cholinergic excitatory and nitrergic inhibitory functions are impaired, indicating an important physiological role for interstitial cells of Cajal in gastric neurotransmission. [23]

REGIONAL MOTOR PATTERNS IN THE STOMACH AND DUODENUM

Proximal Stomach

The proximal stomach serves to accommodate and store ingested food, to regulate intragastric pressure, and to propel chyme into the distal stomach in a tonic fashion. To accomplish these tasks, the proximal stomach possesses distinct myogenic and neural characteristics.

Electrical Activity The resting membrane potential decreases from -48 mV in the fundus to -71 mV in the antrum. The electrical threshold for gastric contraction is -50 mV. Thus, under basal conditions, the fundus is in a state of continual partial contraction or tone, which is modulated by neural or hormonal input. Minor depolarizations or hyperpolarizations in membrane potential produce significant increases or decreases in tone, respectively. In contrast to the distal stomach, the proximal stomach does not exhibit rhythmic fluctuations in membrane potential.
Contractile Activity The proximal stomach exhibits two distinct contractile patterns. Slow, sustained contractions up to 6 minutes in duration constitute 80% of proximal gastric motor activity and determine the basal intragastric pressure. [24] Superimposed on these slow tonic changes are more rapid phasic contractions up to 30 seconds in duration. Regulation of proximal gastric motility allows the maintenance of a stable intragastric pressure in response to ingested volumes in excess of 1 liter. [25] In dogs, intragastric pressure initially drops after eating and then increases above basal levels 30 to 90 minutes postprandially. [26] In humans, intragastric pressure decreases from 12.9 to 9.8 mm Hg in the first 30 minutes after eating; tone does not return to basal levels until all solids have emptied from the stomach. [27], [28] Maintenance of gastric pressure is controlled by two proximal gastric reflexes: receptive relaxation and gastric accommodation. Receptive relaxation is the reduction in proximal gastric tone that occurs with the act of swallowing. Transfer of the swallowed bolus into the stomach is not required to activate receptive relaxation; the reflex occurs with a dry swallow or mechanical stimulation of the pharynx or esophagus. [29] Gastric accommodation, the relaxation of the proximal stomach in response to gastric distention, is mediated by stimulation of gastric mechanoreceptors and does not require pharyngeal or esophageal stimulation. [29], [30] Because of the accommodation reflex, 80% of fluid placed in the stomach is retained in the fundus and proximal body. Receptive relaxation and accommodation are mediated by vagovagal reflex arcs through the nucleus tractus solitarius. [31]
Neurohumoral Control of Motility Extrinsic innervation exerts an important regulatory influence on proximal gastric tone. Vagal cooling reduces proximal gastric motor activity and demonstrates basal vagally mediated tone. [32] Truncal or proximal gastric vagotomy produces decreased gastric distensibility and increased intragastric pressure after bolus ingestion. [33], [34] Electrical activation of low-threshold vagal fibers increases fundus tone, whereas stimulation of high-threshold fibers relaxes the proximal stomach. [6], [35] Sympathetic nerves also modulate proximal gastric activity. Splanchnicectomy augments the increase in intragastric pressure evoked by vagal activation. [36] Several mediators modify motor activity in the fundus. NO is a major physiological mediator of proximal gastric relaxation. The nitrergic agent nitroglycerin relaxes the proximal stomach, whereas administration of NO synthase inhibitors increases fundic tone, an effect blocked by vagal cooling or atropine; this finding indicates that NO acts presynaptically on vagal cholinergic efferent nerves. [37] Receptive relaxation is postulated to result from VIP release by fundic nerves. [6], [29] Agents that relax the proximal stomach or enhance fundic compliance include cholecystokinin (CCK), secretin, VIP, gastrin, somatostatin, dopamine, GRP, glucagon, bombesin, and the a $_2$-adrenergic receptor agonist clonidine, whereas motilin and thyrotropin-releasing hormone (TRH) increase fundic pressure. [38], [39] and [40] Secretin relaxes the stomach by activating vagal afferent pathways originating in the gastroduodenal mucosa. [41] Pentagastrin-evoked relaxation is mimicked by histamine and is abolished by acid-suppressing agents, suggesting mediation by a mechanism involving gastric secretion.
Reflex Modulation of Motility Neurohumoral mechanisms participate in the reflex control of proximal gastric motility by other regions of the gut. Duodenal balloon inflation reduces fundic tone through vagal and splanchnic nonadrenergic, noncholinergic pathways. [42] Vagal application of the sensory neurotoxin capsaicin attenuates the decrease in fundic pressure after duodenal distention at low inflation volumes. Capsaicin treatment of the celiac and superior mesenteric ganglia reduces the response to all volumes of balloon inflation, indicating activation of vagal sensory pathways by low levels of distention and activation of splanchnic nerve pathways by more intense stimuli. [43] Other enterogastric reflexes, such as the inhibition of proximal gastric activity by duodenal protein or lipid perfusion, are variably mediated by vagal nonadrenergic, noncholinergic pathways possibly involving NO. [42], [44] Enterogastric reflexes are not strictly localized to the duodenum. Colonic distention also inhibits proximal gastric motor function. As part of the ileal brake, ileal perfusion of glucose reduces fundic tone. [26] CCK is postulated to inhibit proximal gastric tone during duodenal perfusion of lipids or certain amino acids such as L-tryptophan. Capsaicin inhibits the reduction in intragastric pressure evoked by CCK, indicating participation by afferent pathways. [45] Fundic relaxations in response to duodenal acid exposure are mediated by multiple pathways. At low hydrochloric acid perfusion rates, relaxation is mediated by endogenous secretin, which acts on vagal afferent pathways. [46] Higher acid loads induce CCK release and have direct action on afferent nerve pathways.

Distal Stomach

The distal stomach exhibits electrical and contractile properties distinct from those of the proximal stomach that serve to grind and triturate solid food and to regulate gastric emptying of solid and, to a lesser extent, liquid meals.

Rhythmic Electrical Activity The distal stomach exhibits a more negative membrane potential than the proximal stomach. Superimposed on the resting membrane potential is a rhythmic depolarization, known as the *slow wave,* consisting of an initial rapid depolarization followed by a more prolonged plateau potential (Fig. 10-2). A site along the greater curvature in the proximal gastric body exhibits the highest oscillatory frequency (3 cycles/min [cpm] in humans) and acts as the dominant pacemaker to entrain the rest of the stomach. [47] The cells of origin of pacemaker activity are believed to reside in the interface between the longitudinal and circular muscle layers. Interstitial cells of Cajal exhibit rhythmic depolarizations in the small intestine and colon and are thought to generate slow wave activity in these organs. Interstitial cells of Cajal are prominent in the myenteric region of the gastric antrum and corpus and may serve a similar pacemaker role in the stomach. [48] Although the slow wave is a myogenic characteristic, altered neural input can destabilize slow wave rhythmicity. Acute vagotomy induces slow wave disorganization with generation of ectopic antral pacemakers. [49]

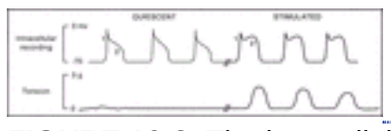

FIGURE 10-2. The intracellular electrical activity of distal gastric smooth muscle and the resultant contractile response are displayed under quiescent conditions and after stimulation with a contractile agonist. In the unstimulated state, distal gastric smooth muscle exhibits rhythmic electrical activity with an initial upstroke (*1*), followed by a plateau potential (*2*). Because the amplitude of this electrical depolarization does not reach a critical threshold, no contraction occurs. With stimulation, the plateau is prolonged and enhanced, so a threshold depolarization is achieved, resulting in phasic contractions in the distal stomach. (Adapted from Kim CH. Electrical activity of the stomach: clinical applications. Mayo Clin Proc 1986;61:205.)

Slow waves are propagated distally through the smooth muscle. Unlike the heart, the stomach has no specialized electrical conduction pathways. Conduction is faster circumferentially than along the longitudinal axis; therefore, the slow wave propagates in well-defined rings of depolarization. [50] Slow wave propagation is slightly faster along the greater curvature, such that myoelectric activity from the greater and lesser curves reach the pylorus simultaneously. The propagation velocity increases from 0.5 cm per second in the gastric body to 4 cm per second in the distal antrum. Slow waves do not propagate proximally into the fundus because of its less negative resting membrane potential and other properties that limit its excitability. Gastric myoelectric rhythmicity matures late in fetal and postnatal development. The percentage of time that normal 3 cpm slow wave activity is demonstrable increases from 37% in preterm infants to 66% 6 months after birth. [51] Anatomic correlates of these observations indicate that neuronal networks within the enteric nervous system are immature at birth, and connections between the interstitial cells of Cajal and smooth muscle cells are poorly coupled. [52]

Contractile Activity Under quiescent conditions, the gastric slow wave is of insufficient amplitude to evoke significant contractions. Although the initial rapid depolarization of the slow wave may induce trivial pressure waves, the plateau potential remains more negative than the contractile threshold. Contractile agonists increase the duration and amplitude of the plateau potential (see Fig. 10-2) and, in some cases, induce action potentials (+100 ms) that are intense depolarizations superimposed on the plateau potential. [53] These events provide the depolarization needed to exceed the contractile threshold. In contrast, relaxing agents reduce the plateau potential amplitude or duration or prevent the stimulatory effects of contractile agonists, although VIP inhibits motor activity without altering slow wave morphology, suggesting the involvement of other mechanisms. [54] Because the contractile threshold is exceeded only during each plateau potential, the slow wave determines the maximal distal gastric contractile frequency (3 cpm in humans). Phasic antral contractions may exceed 100 mm Hg, migrating with the propagating slow wave as a ring and increasing in amplitude and velocity as the slow wave approaches the distal antrum. Because of neurohormonal input, not all ring contractions traverse the entire distal stomach. Some contractions subside before reaching the pylorus, whereas others initiate in the distal antrum. The distal stomach exhibits distinct phasic motor patterns under fasting and postprandial conditions. The fasting *migrating motor complex* (MMC) is a stereotypical pattern that clears the stomach of undigested debris. Loss of the MMC in certain disease states may promote gastric bezoar formation. After eating, the MMC is replaced by a *fed contractile pattern* of variable intensity and duration that grinds and triturates the ingested nutrients.

Migrating motor complex. The MMC clears the stomach and intestine of undigested food particles, mucus, and sloughed epithelial cells during fasting and has been termed the gastrointestinal housekeeper. The MMC consists of three phases with a combined duration of 84 to 112 minutes (Fig. 10-3). [55] Phase I is a period of relative motor quiescence, representing 40% to 60% of the cycle. High-amplitude contractions are not observed during phase I, but diminutive pressure waves of +9 mm Hg in amplitude can occur in phase with the slow wave. [56] Phase II, comprising 20% to 30% of the cycle, exhibits increasing but irregular contractions. Phase III is a 5- to 10-minute period of intense, rhythmic, lumenally occlusive contractions that begin in the gastric body and propagate unimpeded to the pylorus. The frequency of contractions during phase III approaches that of the slow wave. [55] Of the phase III complexes, 71% begin in the stomach, whereas 18% originate in the proximal duodenum, and 10% and 1% start in the distal duodenum and proximal jejunum, respectively. [57] Phase IV, a brief period of transitional motor activity from phase III to phase I, has been described. MMC cycling exhibits diurnal variation, with fewer complexes occurring during sleep (0.25/h) than wakefulness (0.64/h). [58] Phase I is prolonged during sleep, whereas phase II is shortened. The MMC is established early in life, although some infants exhibit an immature pattern of isolated pressure waves alternating with clustered phasic contractions. [59] MMC activity is not impaired in the elderly. [60] MMC cycles are more frequent and phase III complexes are shorter in women than in men. [58]

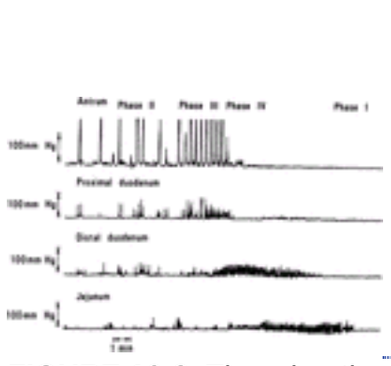

FIGURE 10-3. The migrating motor complex from a healthy human consists of four phases: phase I, a period of motor quiescence; phase II, a period of irregular contractile activity; phase III, a brief complex of intense rhythmic contractions that propagate from the distal stomach into the small intestine; and phase IV, a transitional phase back to the quiescence of phase I. (From Rees WDW, Malagelada JR, Miller LJ, Go VLW. Human interdigestive and postprandial gastrointestinal hormone patterns. Dig Dis Sci 1982;27:321.)

The propulsive characteristics of interdigestive gastric motor activity vary depending on the MMC phase. Phase III contractions are highly propagative. [55] In contrast, phase II motor activity exhibits little coordination from one gastric recording site to the next and is believed to represent a period in which fasting gastric contents are mixed. Emptying of inert liquids and endogenous secretions is more rapid during phase III than in phases I and II. [61]

Fed motor pattern. The fed motor pattern is induced 5 to 10 minutes after eating and persists as long as food remains in the stomach. Intermittent phasic contractions of irregular amplitude similar to those of phase II of the MMC comprise the fed pattern (Fig. 10-4). As with phase II, half of the slow wave cycles are associated with contractions. Fluoroscopic studies of postprandial motor activity demonstrate antral contractions that propel the ingested material distally, only to be retropelled back into the proximal stomach, thus serving to mix and grind the food. [62] Ultrasonographic investigations indicate that this distal gastric motor function shortens the gastric length and alters the configuration of the stomach. [63] Furthermore, interaction of fed gastric contractions with the incisura facilitates the breaking up of solid gastric contents.

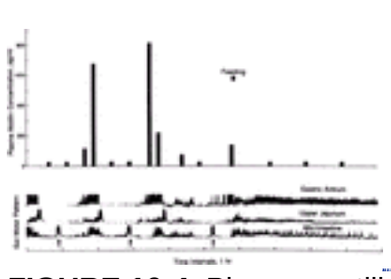

FIGURE 10-4. Plasma motilin levels during fasting and the fed state from a representative canine experiment. Fasting motor activity shows regular cycling of the migrating motor complex. Each antral phase III complex is accompanied by a significant increase in motilin concentration. After a meal, the motor pattern changes to one of irregular but continuous phasic contractions for the duration of time that food remains in the stomach. The fed motor pattern is associated with suppression of plasma motilin levels. (Adapted from ref. [82].)

Postprandial contractile amplitudes depend on the consistency and composition of the ingested material. Antral contractions evoked by eating particulate nutrients are more intense than those induced by an equivalent homogenized meal. [64] The duration of the fed period is proportional to the number of calories consumed, with fats inducing a more prolonged response than proteins or carbohydrates. [65] As little as 17 g of fat may induce a prolonged fed pattern. Nutrients are not strictly necessary to evoke the fed pattern. Sham feeding evokes transient increases in antral contractility and delays the onset of the next phase III. This phenomenon is blocked by the CCK antagonist loxiglumide, suggesting mediation by endogenous CCK. [66] Physical factors modify the characteristics of the fed pattern. The presence of plastic spheres in saline induces stronger antral contractions than a similar amount of saline alone. Increasing the viscosity with polycarbophil, a gel-forming compound, reduces antral contractile amplitudes but prolongs the fed pattern for several hours. [67] More than 50% of individuals fed continuously through an intragastric catheter revert to a fasting pattern, indicating that nutrient inhibition of interdigestive motor activity is self-limited. [68] Intravenous nutrient administration disrupts normal fasting gastric motor activity; this indicates that the systemic effects of a meal may contribute to selected components of the fed motor response. Intravenous amino acid

solutions reduce phase II duration and suppress antral phase III activity. [69]

Neurohumoral Control of Motility Neural pathways are important physiological regulators of distal gastric motor activity. Efferent vagal stimulation of low-threshold fibers evokes antral contractions that are sensitive to atropine, indicating cholinergic mediation. In contrast, stimulation of high-threshold fibers decreases antral motor activity, most likely through release of VIP, and this reduces the sensitivity of gastric smooth muscle to calcium. [6], [70] In dogs, NO synthase inhibitors increase antral contractions, whereas NO precursors decrease motor activity, indicating a physiological relaxant role for NO. [71] Administration of the phosphodiesterase inhibitor sildenafil inhibits antral motor activity by enhancing the action of endogenous NO. [72] Infusion of NO synthase inhibitors evokes MMC-like contractions in fed rats, whereas the NO donor sodium nitroprusside induces complexes similar to the fed pattern, suggesting that NO may be the final mediator controlling fasting and postprandial motility in some species. [73] Hormones and neurotransmitters can modify distal gastric electromechanical function. CCK, bombesin, substance P, and some opiates increase antral contractions, but the physiological relevance of these effects is unknown. [74] Conversely, secretin, somatostatin, glucagon, GRP, TRH, neurotensin, calcitonin, peptide YY, prostaglandin E_2, the a_1-adrenergic receptor agonist phenylephrine, and the a_2-adrenergic receptor agonist clonidine inhibit antral motor activity. [75] Some compounds have dual effects; for example, the 5-HT_3 receptor agonist m-chlorophenylbiguanide increases antral motility under fasting conditions but reduces the amplitude of postprandial antral contractions. [76]

Neurohumoral regulation of the MMC and fed motor pattern. Neurohumoral control of the gastric MMC is complex and incompletely understood. Phase III persists after sectioning of the vagus and splanchnic nerves. Autotransplantation of the stomach or the entire gut with denervation of all extrinsic input does not interrupt spontaneous phase III events in the excluded gastric segment, suggesting that intact extrinsic innervation to the stomach is not needed for phase III initiation. [77], [78] Nevertheless, vagal recordings show increased efferent firing during antral phase III, suggesting participation of vagal pathways. Additionally, vagal cooling abolishes antral phase III patterns; this finding indicates that fasting gastric motor function can be modulated by the vagus. [79] The inhibition of phase III by vagal cooling is not reversed by the a- and ß-adrenergic receptor antagonists phentolamine and propranolol, showing that unopposed adrenergic activity is not responsible for the phenomenon. [80] In contrast to its effects on phase III activity, bilateral vagotomy decreases or abolishes gastric phase II, suggesting that the different MMC phases are regulated by distinct pathways. [81] Many lines of evidence suggest the presence of a hormonal mediator of gastric phase III activity. Initiation of antral phase III correlates temporally with elevations in motilin, a hormone localized to duodenal mucosal cells (see Fig. 10-4). [82], [83] Premature antral phase III is inducible by motilin infusion in dogs and humans. Infusion of motilin antibodies, which suppress circulating motilin levels, abolishes gastric phase III in dogs for several hours. [84] Resection of the duodenum (and presumably most motilin-secreting tissue) eliminates antral phase III in dogs. [85], [86] Motilin stimulates antral phase III in part through cholinergic pathways, because atropine reduces its contractile effects. [87] Duodenal phase III activity, however, can persist in the presence of atropine, indicating control by noncholinergic mechanisms. Depletion of endogenous serotonin by *p*-chlorophenylalanine suppresses spontaneous and motilin-induced gastric phase III, suggesting involvement of serotonergic pathways. [88] Motilin also releases NO. [89] NO synthase inhibitors increase MMC frequency and shorten the duration of phase I in humans; this indicates modulation of MMC activity by endogenous NO pathways. [90] Similarly, in dogs, inhibition of NO synthesis induces premature antral phase III. [91] These effects are associated with increased motilin levels during inhibition of NO synthesis and indicate regulation of motilin release by endogenous NO pathways (Fig. 10-5). [91] The physiological stimulus for motilin release is unknown, although motilin levels increase after vagal stimulation. Extrinsic denervation of the upper gut, however, does not interrupt motilin cycling. [78] Evidence suggests that there is plasticity in the neural regulation of the gastric MMC. In dogs who underwent resection of the pylorus, duodenum, and upper jejunum, MMC cycling resumed 1 to 4 months postoperatively. Reemergence of the MMC was independent of motilin cycling, but it relied on vagal cholinergic and adrenergic efferent pathways. [92]

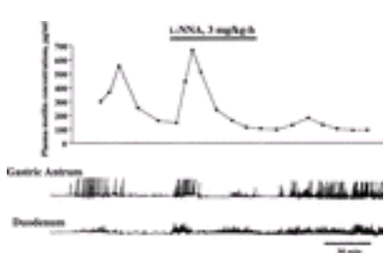

FIGURE 10-5. Effects of nitric oxide (*NO*) synthase inhibition with $N^?$-nitro-L-arginine (*L-NNA*) on gastroduodenal motility and plasma motilin release in a fasted dog. L-NNA induced premature phase III contractions in the antrum and duodenum with an associated increase in plasma motilin levels. (From ref. [91].)

Motilin receptors are present on antral smooth muscle and on nerves within the stomach wall. Receptor subtypes in these regions may be distinct, suggesting possible differential roles in regulating upper gut motor function. [93] The existence of two or more motilin receptor subtypes is supported by neurotransmitter binding studies that show different affinities for two motilin receptor agonists in membranes from neural and muscular tissues of the stomach. [93] Other factors modulate gastric MMC activity. The opioid receptor antagonist naloxone prolongs MMC cycling from 103 to 219 minutes and delays plasma motilin peaking, suggesting that endogenous opiates may regulate motilin release. [94] Naloxone does not prevent phase III evoked by exogenous motilin; this finding confirms that motilin does not act through opiate-dependent pathways. The histamine H_2 receptor antagonist famotidine shortens the MMC cycle length, and this also suggests a modulatory role for endogenous histamine. [95] Pancreatic polypeptide cycles in phase with gastric phase III; however, exogenous pancreatic polypeptide does not induce premature phase III activity. Furthermore, pancreatic resection, which removes pancreatic polypeptide–secreting cells, does not alter the antral MMC. [96] Motilin releases pancreatic polypeptide through vagal cholinergic pathways, indicating that motilin may regulate physiological functioning of this hormone. [97] Secretory activities of the upper gut may influence gastric MMC activity. Fasting pancreaticobiliary and secretory immunoglobulin A output cycle in phase with the MMC, and increases in biliary secretion correlate with motilin release. [98], [99] Additional peaks of pancreatic secretion occur during fasting that are independent of antral phase III and motilin cycling. [100] Duodenal bile acid perfusion suppresses antral contractions, suggesting that endogenous bile release does not mediate the gastric phase III. Antral phase III complexes do not develop when the duodenal pH is less than 7.0 regardless of the plasma motilin level. [101] Furthermore, duodenal hydrochloric acid perfusion abolishes the gastric phase III; this indicates that duodenal alkalinization is a prerequisite for MMC cycling. Gastric acidification to pH 1.0 inhibits spontaneous and motilin-induced gastric phase III through a vagovagal reflex. [102] Patients with chronic pancreatic insufficiency exhibit MMC cycles of shorter duration, indicating an interrelation between antroduodenal and pancreatic function. [103] Neurohumoral factors control postprandial gastric motor function, although the specific mediators of the fed pattern are unknown. Distention of the proximal stomach produces irregular phasic contractions mimicking the fed pattern in the proximal intestine, indicating a possible role for gastric mechanoreceptor-activated pathways. [104] Infusion of a meal into an extrinsically denervated, autotransplanted loop of small intestine interrupts antral MMC activity; this indicates that external innervation is not needed for induction of the fed pattern. [105] However, as with the MMC, extrinsic neural activity can modulate postprandial motor function. Bilateral vagotomy increases the threshold for inducing the fed pattern and shortens its duration. [81] Autotransplantation of the entire gut increases the number of calories needed to abolish MMC activity and induce the fed pattern. [106] Finally, vagal cooling converts the fed pattern to intermittent phase III activity. [107] When blood from a fed animal is perfused into an isolated stomach preparation, motor complexes similar to the fed pattern are generated, suggesting the presence of an unknown circulating mediator. [108] Many candidates for the humoral inducer of the fed pattern have been proposed, but none are proven. Plasma gastrin rises after eating. Physiological infusions of gastrin increase the amplitude and frequency of antral contractions in association with increases in the percentage of slow waves that reach a contractile threshold. [109] These effects, however, are too greatly delayed and are too short in duration for gastrin to represent the major mediator of the fed pattern.

Reflex Modulation of Motility As with the proximal stomach, distal gastric motor activity is subject to reflex modulation from other gut regions. In ferrets, distention of an excluded fundus pouch evokes antral contractions through a vagal cholinergic pathway. [110] A vagal fundoantral reflex is also observed on distention of the human and the canine proximal stomach. [33], [35], [49] During fasting, proximal gastric distention disrupts MMC cycling, suggesting that the fundoantral reflex may regulate physiological induction of the fed pattern to stimulate mixing when the fundus is distended by an ingested bolus. [111] Circumferential myotomy of the midstomach promotes increased antral motor activity after meal ingestion, suggesting that intrinsic connections within the gastric wall mediate a tonic inhibition on the antrum by the fundus. [112] The distal stomach also responds to feedback from extragastric sites. Administration of saline or water to the larynx and epiglottis inhibits antral motor activity in anesthetized rats. [113] Rectal distention reduces antral motor activity and induces slow wave instability. [114] Duodenal distention inhibits antral motor activity, a reflex partly blocked by vagotomy or splanchnicectomy and abolished by both. [42] In contrast, duodenal distention–evoked inhibition of antral motor activity is unaffected by duodenal transection, excluding a prominent role for intrinsic neural pathways in this reflex. Intraduodenal perfusion of lipids, proteins, or hydrochloric acid reduces spontaneous antral contractions—effects that are reduced but not abolished by vagotomy. [115] In rats, duodenal perfusion of glucose and amino acids increase vagal afferent firing. [116] A role for afferent neurons is suggested by studies of capsaicin, which, when applied to the vagus nerves, partially reverses the inhibitory effects of protein, glucose, and trypsin inhibitor on distal gastric motor activity. [117] Intraduodenal lipids reduce motor activity in the denervated stomach and autotransplanted gastric pouches, indicating regulation by hormonal pathways. [118] The ability of the CCK antagonist L364,718 to reverse the inhibitory effects of intraduodenal protein on gastric motility in rats suggests that CCK may mediate enteroantral reflex activity in that species through action on CCK-A receptors. [117] The inhibitory effects of the different nutrient classes are dependent on the chemical nature of the nutrient that is perfused. Protein or amino acid perfusates containing

L-tryptophan potently inhibit antral motility. [115] Long-chain triglycerides are more effective inhibitors of distal gastric contractions than short- or medium-chain lipids. [119]

Pylorus

The pylorus serves as a sieve to regulate outflow of intralumenal contents from the stomach. To fulfill this role, the pylorus possesses unique smooth muscle and neural properties that distinguish it from the surrounding structures. Because of its thickness and the density of connective tissue it possesses, the pylorus acts as a mechanical stricture to the passage of large particles. The sphincteric properties of the pylorus are aided by a redundant, highly folded mucosa that narrows the lumenal diameter. The nerve density in pyloric circular muscle is three to five times greater than that in the adjacent antral circular layer, a feature indicating that regulation of pyloric motor activity is different from the surrounding regions. [120] The sensory nerve supply projecting from the pylorus to brainstem vagal nuclei is greater than from the duodenum, suggesting regional differences in extrinsic afferent innervation. [121] Increased numbers of neurons containing VIP, substance P, enkephalins, neuropeptide Y, and galanin are present in the pylorus as compared with the antrum and duodenum. [120], [122]

Electrical Activity The pylorus exhibits histological characteristics suggesting that its electrical characteristics are different from those of the distal stomach. The density of gap junctions interconnecting pyloric smooth muscle cells is less than in the antrum. [120] Prominent interstitial cells of Cajal interact with pyloric myocytes and may act as pacemaker cells in the pylorus. [123] The pyloric slow wave is entrained to the same frequency as in the distal stomach (3 cpm), but most antropyloric slow waves are not propagated into the duodenum, because of the thick, fibrous septum. In contrast, some spike potentials can cross the pyloric region in patches and may form the basis for gastroduodenal coordination. [124]

Contractile Activity The pylorus is a barrier to gastric emptying that is modulated by internal and external influences. In dogs, the pylorus has a resting pressure of 10 mm Hg and exhibits spontaneous contractions. [123] In humans, resistance to flow also is provided by tonic and phasic pyloric motor activity, although the presence of basal pressure is not reliably demonstrable. In humans, pyloric smooth muscle relaxes during electrical stimulation, indicating a predominance of inhibitory neural input, whereas in the dog, electrical stimulation produces pyloric contraction. [125] The ability of the neural toxin tetrodotoxin to enhance phasic pyloric contractions indicates that this motor activity is an inherent property of the smooth muscle. [125] The pylorus exhibits characteristic motor patterns under fasting and fed conditions. During phase III of the MMC, the pylorus is open, and fasting gastric contents exit into the duodenum. After eating, the pylorus exhibits a complex motor response of prolonged periods of closure interrupted by brief intervals during which antral contents pass into the intestine. [126] In general, pyloric contractions are synchronized with fed antral activity. However, some foods, such as milk, evoke isolated pyloric contractions with antral inhibition before induction of a fed antral pattern. Large particles do not traverse the pylorus during the fed state, indicative of enhanced sieving after meal ingestion. [127] The role of the pylorus as a sieve is illustrated by imaging of antropyloroduodenal motility after a meal. On cinefluoroscopy, a minor ring contraction in the gastric body is followed 2 to 3 seconds later by an intense contractile ring that obliterates the lumen. The minor ring contraction induces pyloric closure as the larger contraction approaches the midantrum. The intense contractile ring further propagates into the distal antrum, propelling trapped food against the occluded pylorus and producing grinding and mixing. This is followed by retropulsion of the mixed bolus into the proximal stomach as the major contractile wave reaches the pylorus. Because the pylorus is closed by the initial minor ring contraction, all gastric emptying occurs early in the cycle before the major contraction reaches the midantrum. The relative timing of the two waves is dependent on the latent period between the initial rapid depolarization of the slow wave, which induces the minor contraction, and the peak of the plateau potential, which is responsible for the major contraction. Because this is a myogenic phenomenon, the timing of the two contractile rings is not dependent on neural, hormonal, or meal-related factors. [128] Ultrasonographic studies show that most transpyloric flow occurs during periods of prolonged pyloric opening not associated with occlusive antral or duodenal contractions with the formation of a common antropyloroduodenal chamber. [129] Furthermore, bursts of duodenogastric reflux are demonstrable before pyloric closure, suggesting that duodenal mixing and retropulsion occur. [129], [130] It is hypothesized that shear forces induced by the sudden change in velocity of the intragastric contents and the squeezing action by the antral walls disperse large particles into smaller ones that subsequently are delivered to the duodenum. Resection of the pylorus does not prevent postprandial sieving and trituration, suggesting that other gastroduodenal regions also possess these capabilities. [131] Resection of both the antrum and pylorus results in loss of this function, with passage of larger, undispersed particles.

Neurohumoral Control of Motility Neurohumoral regulation of pylorus motor function is distinct from that of the duodenum and distal stomach. Electrical vagal stimulation produces pyloric contraction at low frequencies, and relaxation with increases in transpyloric flow at high frequencies, suggesting the presence of both excitatory and inhibitory vagal pathways. [123], [132] That vagally mediated pyloric contractions are blocked by naloxone indicates mediation by opioid pathways. This finding correlates with the documentation of large numbers of enkephalin-containing fibers in pyloric smooth muscle and pyloric branches of the vagus. [133] Electrical stimulation of the splanchnic nerves evokes pyloric contraction. Pyloric motor activity also is influenced by ascending and descending intrinsic neural pathways. Electrical stimulation of duodenal muscle induces pyloric contractions, whereas antral muscle depolarization relaxes the pylorus. Administration of NO donors blunts basal pyloric pressure waves, reduces the pyloric motor response to intraduodenal lipids, and enhances transpyloric flow after eating, confirming the ability of nitrergic mediators to reduce pyloric motor activity. [134], [135] NO synthase inhibitors block the inhibition of pyloric motility evoked by vagal stimulation, antral depolarization, or electric field stimulation of the pylorus and reduce transpyloric flow; these findings suggest that NO is a common mediator of pyloric relaxation (Fig. 10-6). [136], [137] NO may act in concert with purinergic nerves to mediate pyloric relaxation, as suggested by the observation that combined administration of NO synthase inhibitors and P_{2X} purinoceptor antagonists more potently inhibit electrically induced pyloric relaxations than NO synthase inhibitors alone. [138] Several agents have potent effects on pyloric motor activity. VIP, peptide histidine-isoleucine, galanin, prostaglandin E_1, and serotonin relax the pylorus, whereas CCK, secretin, and histamine produce pyloric contraction. [133], [139], [140] Although CCK has a limited role in enteropyloric reflex activity, the finding that the CCK antagonist L364,718 reduces pyloric motility after a meal suggests that CCK plays some role in the postprandial state. [141]

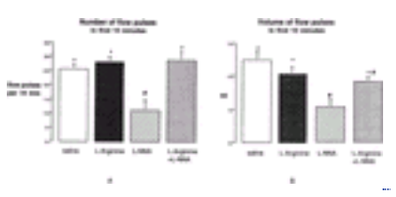

FIGURE 10-6. Effects of nitric oxide (*NO*) synthase inhibition on transpyloric flow. The NO synthase inhibitor $N^{?}$-nitro-L-arginine (*L-NNA*) decreases both the number and volume of flow pulses across the pylorus; this indicates that NO is a physiological relaxant in that region. The NO precursor L-arginine reverses the effects of L-NNA. *, $P < 0.05$ cf L-NNA; #, $P < 0.05$ cf saline. (From ref. [137].)

Reflex Modulation of Motility Pyloric motor activity is modified stimulation of the small intestine. Duodenal perfusion of lipids, amino acids, glucose, hypertonic saline, or hydrochloric acid produces pyloric closure and decreases transpyloric flow. [25] Ileal triglyceride perfusion evokes isolated pyloric contractions. [142] Ileal delivery of short-chain fatty acids increases pyloric tone and reduces the stroke volume of transpyloric fluid pulses. [143] Combined cinefluoroscopy and manometry recordings demonstrate that the pylorus is closed 98.5% of the time that pyloric tone exceeds 4 mm Hg. [144] Induction of isolated pyloric contractions is associated with inhibition of liquid gastric emptying. [142], [145] Pathways that mediate reflex stimulation of pyloric motor activity are dependent on the stimulus. Isolated pyloric contractions evoked by duodenal hydrochloric acid are antagonized by atropine and the ganglionic blocker hexamethonium but are not blocked by vagotomy, indicating mediation by nonvagal cholinergic pathways. [146] Similarly, the response to intraduodenal glucose is blocked by atropine. The ability of the topical anesthetic lidocaine (Xylocaine) to prevent acid-induced pyloric contraction is evidence of a role of mucosal receptors in initiating this enteropyloric reflex. [146] The 5-HT_3 receptor antagonist zacopride reduces pyloric motor responses to intraduodenal hydrochloric acid, indicating that serotonergic nerves also play a role. Although atropine reduces the pyloric response to intraduodenal lipids, it is postulated that hormones such as CCK also modulate the pyloric motor response. However, other studies suggest that CCK plays a limited role in enteropyloric reflex activity under physiological conditions. Mediation by opioid pathways is suggested by the ability of naloxone to block pyloric contractions induced by intraduodenal amino acids. [147] The inability of naloxone to block lipid-evoked pyloric contractility is evidence of differential regulation of pyloric motility by different nutrient classes. [148]

Duodenum and Small Intestine

The small intestine modulates gastric emptying and plays a pivotal role in regulating gastric and pyloric motor activity. The duodenum possesses an electrical pacemaker distinct from that of the stomach, with a frequency of 11 to 12 cpm. Some electrical activity from the stomach propagates across the pylorus, despite the presence of the fibrous septum, and is responsible for intermittent antroduodenal coordination with one gastric slow wave cycling in phase with every three to four duodenal slow waves. This promotes propagation of some antral pressure waves into the duodenum, most commonly during phase II. [149], [150] Because of the higher duodenal slow wave frequency, additional duodenal contractions interspersed between these coordinated antroduodenal waves are generated. The inability of the neural toxin tetrodotoxin to prevent antroduodenal coordination indicates that electrical propagation is carried by pyloric longitudinal muscle fibers and not by neural

pathways. [151]

The duodenum exhibits other contractile patterns as well. After eating, isolated duodenal contractions segment and mix food particles with pancreaticobiliary secretions. Intraduodenal lipid or bile perfusion decreases intestinal flow, an effect associated with increases in duodenal diameter with fewer contractions and reductions in propagation distances. [152], [153] In contrast, hydrochloric acid and hyperosmolar solutions retard duodenal propagation by inducing tonic duodenal lumenal occlusions. [152] Duodenal myotomy reduces the evoked duodenal contractions with enhancement of lumenal propulsion. [154] Under some conditions, strong retroperistaltic contractions in the duodenum propel intestinal contents orally and induce duodenogastric reflux. Studies demonstrating retrograde duodenal motor activity during late phase III of the MMC imply the presence of a retroperistaltic pump that may regulate fasting duodenal pH. [155] Duodenogastric retropropulsion of bile in the refluxate is minimized by phase III–associated closure of the sphincter of Oddi. [156] After eating, 40% to 50% of the pressure waves in the initial 5 to 6 cm of the duodenum are retrograde and may contribute to controlling gastric emptying. [157]

Just as stimulation of the small intestine modulates gastric contractile function, gastric perturbations can alter intestinal motor patterns. Gastric distention delays intestinal transit, a phenomenon known as the *gastroenteric reflex.* [25] This reflex is unaffected by vagotomy but is reduced by small intestinal denervation and by severing connections from the celiac plexus. [25], [158] The abilities of L-arginine applied to celiac ganglion neurons to enhance the reflex and NO synthase inhibitors to block the reflex indicate mediation by celiac plexus NO release. [159] The gastroileal reflex, the increase in ileal propulsion after eating, is abolished by intestinal transection, indicating mediation by intrinsic nerves. [160] Decreases in pressure at the ileocecal junction are reported in response to nutrient ingestion. [161]

GASTRIC EMPTYING

The coordinated actions of the distinct regions of the stomach with feedback from the small intestine regulate emptying of gastric contents into the duodenum. Intragastric contents are categorized as liquids, digestible solids, fats, and indigestible solids because of the differential handling of each by the stomach.

Gastric Emptying of Liquids

Kinetics of Emptying of Inert Liquids Gastric emptying of inert liquids such as water or isotonic saline follows a single exponential curve, termed *first-order kinetics,* with a time to 50% emptying of 8 to 18 minutes. The volume of liquid emptied into the duodenum in a given time is a constant fraction of the volume that remains in the stomach. [162] Thus, a 300-mL saline bolus will empty at a rate twice as fast as a 150-mL load. Studies in preterm infants indicate that mature patterns of liquid emptying develop by the 32nd gestational week; before then, liquid emptying is variably delayed. [163], [164] Some investigations suggest that liquid emptying is slower in women than in men. [165] Nutrient-containing liquids are emptied more slowly in elderly persons (mean age, 73 years) than in young controls. [166]

Factors that Modify Liquid Emptying The nutritional properties of an ingested liquid modify the speed at which it exits the stomach. Consequently, carbohydrate-, protein-, or fat-containing liquids can be digested and absorbed completely before they reach the distal small intestine. [167] Feedback from the small intestine delays gastric emptying of liquid nutrients because of alteration of the first-order kinetic pattern. Gastric emptying of nutrient liquids exhibits an initial rapid phase lasting 5 to 30 minutes, followed by a slower phase in which the nutrient is emptied at a constant rate for up to 120 minutes. [168] Because of the linearity of liquid nutrient emptying, a 300-mL liquid meal of 11% glucose empties at the same rate as a 150-mL liquid 11% glucose meal. [169] Liquids with high caloric density empty more slowly than foods with fewer calories per unit volume (Fig. 10-7). [169], [170] and [171] Liquid nutrient emptying is controlled to a rate that delivers about 200 kcal per hour into the duodenum by modulating the duration of the rapid emptying phase, regardless of whether calories are in the form of fats, proteins, or carbohydrates. [172] Thus, 0.25 M glucose empties four times as fast as 1 M glucose. The concentrated glucose solution shortens the rapid emptying phase so that the slower linear phase delays its exit from the stomach, while the dilute solution prolongs the rapid emptying phase to enhance duodenal delivery. Caloric control of liquid emptying is operative within a limited range of concentrations. A 1 M glucose solution maximally inhibits liquid emptying. Concentrations greater than 1 M empty from the stomach faster than 200 kcal per hour. [170] Similarly, duodenal caloric delivery of milk protein solutions of high caloric density is greater during the early phase of liquid emptying than for less concentrated protein hydrolysates. [171]

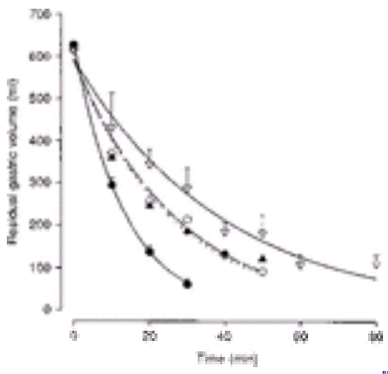

FIGURE 10-7. Gastric emptying profiles of different nutrient liquids. Glucose (*filled circles*) emptied the fastest, whereas liquids with higher caloric density such as milk protein (*open triangles*), pea peptide hydrolysate (*open circles*), and whey peptide hydrolysate (*filled triangles*) emptied much more slowly. (From ref. [171] .)

Other nutrient characteristics regulate liquid emptying as well. Osmolarity is an independent factor in the regulation of liquid emptying. Carbohydrates and most amino acids modulate duodenal delivery, in part, by action on intestinal osmoreceptors. Emptying of glucose solutions is slower than emptying of isocaloric, hypotonic glucose polymer solutions. [173] Vagal osmoreceptor-activated pathways mediate this response, although a dependence on intact celiac ganglia also has been reported. [174] Fructose is a less potent inhibitor of gastric emptying than glucose or xylose, suggesting the participation of other factors as well. [175] L-Tryptophan is considered separately from other amino acids in that it delays liquid emptying in isotonic solutions; this suggests the presence of specific intestinal L-tryptophan receptors. [176] Isocaloric starch, disaccharide, and monosaccharide solutions are equipotent at delaying liquid emptying, as are isocaloric protein and amino acid solutions, suggesting that the digestive products of carbohydrate and protein hydrolysis are the major regulators of duodenal delivery of nutrients. [177] This theory is supported by the report that liquid emptying is not inhibited by a nonhydrolyzable sucrose polyester compound. [178] Fatty acids, rather than triglycerides, mediate emptying of liquid lipid solutions, because triglycerides do not inhibit emptying in exocrine pancreatic insufficiency. [179] Medium-chain fatty acids of 12 to 14 carbons are more potent than longer- or shorter-chain fatty acids. [180] A requirement for endogenous bile is suggested by the observation that biliary diversion prevents the inhibitory effect of fat on gastric emptying. [179] The mechanism by which nutrients slow liquid emptying is unknown, although the effects of protein meals are blocked by antibodies to VIP. [174]

Finally, some studies suggest that the composition of meals ingested previously can modify liquid nutrient emptying. In rats fed a high-protein diet for 3 weeks, gastric emptying of liquid peptones is faster than in animals fed diets lower in protein content (Fig. 10-8). [181]

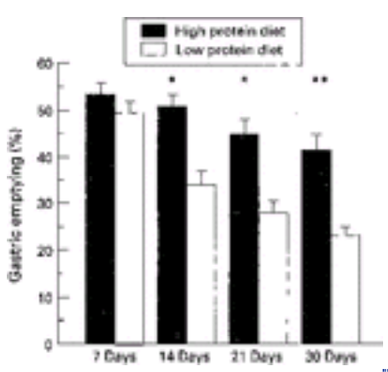

FIGURE 10-8. Gastric emptying of a peptone meal in rats fed a high- or low-protein diet for 7, 14, 21, and 30 days. Long-term ingestion of high-protein meals leads to accelerated emptying of peptone meals. (From ref. [181] .)

Noncaloric physicochemical factors modulate rates of liquid gastric emptying. Emptying of glucose solutions is faster during filling of the stomach than when measured

after meal completion and is dependent on the volume delivered rather than on the rate of gastric filling. [182] Titratable acid delivery to the duodenum is constant regardless of the pH or lipid solubility of acids in the stomach. [183] Large quantities of a weak acid are more potent inhibitors of liquid emptying than trivial amounts of a strong acid such as hydrochloric acid. The mechanism of the inhibitory effects of acid on liquid emptying is not completely understood. Secretin is released on duodenal acid exposure; however, the rapidity of the reflex response to duodenal acidification is more consistent with neural mediation. [184] The actions of intraduodenal hydrochloric acid on liquid emptying require an intact pylorus, suggesting that acid-evoked tonic and phasic pyloric activities are important. [174] Increases in viscosity slow the rate of liquid emptying. [67], [185] Gravity has minimal effects on liquid emptying of carbohydrate or acid solutions, although saline empties slightly faster in a person who is upright than in one who is supine. [186] When a nutrient liquid such as soup is ingested as part of a mixed meal, assuming a supine position may prolong emptying by 50%. [187] Maintaining an upright posture after drinking a nonnutritive liquid produces more long antroduodenal pressure waves and isolated pyloric contractions than does maintaining a left lateral decubitus position. [188] Investigations suggest that liquid emptying is modified by postabsorptive factors. In healthy humans, liquid emptying is slowed by 38% during intravenous hyperalimentation. [189] When half of the amino acids are replaced with branched-chain amino acids, this delay in emptying is attenuated, suggesting that different parenteral nutrients may have distinct effects. Similarly, patients receiving intravenous fat emulsions exhibit delays in gastric emptying.

Roles of Different Physiological Regions in the Control of Liquid Emptying Liquid gastric emptying traditionally has been considered to be controlled by tonic motor activity in the proximal stomach, but investigations have delineated important roles for other regions. If the proximal stomach is extrinsically denervated or resected or if fundoplication is performed, intragastric pressure rises after eating and liquid emptying is accelerated as a consequence of impaired receptive relaxation and accommodation. [64], [190] Receptive relaxation is an important modulator of liquid emptying. Liquid meals delivered to the stomach through orogastric tubes empty faster than swallowed boluses of the same liquid. [191] It has been postulated that the fundus generates a pressure gradient from the stomach to the duodenum that serves to expel liquids from the stomach. However, questions about this model have been raised. Diabetic patients with normal fundus tone commonly exhibit delayed liquid emptying. [192] Furthermore, a pressure gradient between the fundus and duodenum has never been demonstrated. When the pressure in the proximal stomach is maintained 2 cm H_2O lower than that of the duodenum, liquid emptying is unimpaired. [25] These findings suggest that regions other than the proximal stomach play important roles in the emptying of liquids. It is hypothesized that the distal stomach acts as a pump to facilitate liquid emptying. [44], [193] Although antral contractions are not required for liquid emptying, the degree of antral pressure wave activity correlates with expulsion of liquids into the duodenum. [67], [194], [195] Furthermore, when antral contractions are induced by vagal electrical stimulation, liquid emptying is enhanced. In pigs, emptying of nonnutrient liquids occurs in association with non–lumen-occluding gastric corpus contractions that become occlusive in the distal antrum and pylorus, thereby producing discrete flow pulses into the duodenum. [196] Rapid magnetic resonance imaging demonstrates antral contractions of greater amplitude during emptying of nutrients of low caloric density (e.g., 10% dextrose) compared with emptying of more concentrated solutions (e.g., 25% dextrose), indicative of differential regulation of liquid nutrient emptying by action on the distal stomach. [193] If the proximal stomach is surgically excluded, coordinated antropyloric pressure waves generate fluid flow pulses that preserve normal liquid emptying. [197] Antral motor activity also can delay liquid emptying. If the antrum is resected, the initial phase of liquid emptying is accelerated, suggesting that the distal stomach has an inhibitory role during the first several minutes after liquid intake. [198] Furthermore, the antral lumenal diameter is less after ingestion of a liquid nutrient than after ingestion of an inert liquid; this finding indicates that a calorie-containing fluid may be retained by motor properties of the distal stomach. [199] The pylorus plays a crucial role in the emptying of certain liquids. Early studies failed to document an effect of stent placement across the pylorus on the rate of liquid emptying. [200] More recent studies show acceleration of the initial rapid phase of liquid emptying with pyloric stenting without any affect on the subsequent slower linear phase. [201] This finding correlates with observations that pyloroplasty or pyloric myotomy enhance early liquid emptying and with the demonstration that pyloroplasty enhances emptying after highly selective vagotomy. [25], [131], [198], [202] Pyloroplasty also abolishes the resistance to transpyloric flow evoked by hydrochloric acid or lipids. [25] If the duodenum is transected, liquid nutrients do not induce isolated pyloric contractions and are emptied at an accelerated pace, suggesting an important role for stimulated pyloric motility in the control of liquid delivery into the duodenum. [203] Duodenal motor activity also regulates liquid emptying. The absence of duodenal motor activity is associated with accelerated emptying, whereas the presence of continuous duodenal contractile activity correlates with delayed liquid gastric emptying. [128] Enhanced liquid emptying is also noted after circular myotomy of the duodenal wall. [154]

Gastric Emptying of Digestible Solids

Kinetics and Flow Patterns in Solid Emptying The delivery of digestible solid residue to the duodenum is slower than for liquids, with a time to 50% emptying of about 2 hours. [191] The emptying of solids exhibits an initial lag phase, which persists up to 1 hour, during which time little or none of the ingested food leaves the stomach. [204], [205] Cinefluoroscopy demonstrates extensive mixing and retropulsion during the lag phase, during which the food is dispersed into fine particles. The lag phase is followed by a linear emptying phase, during which time the dispersed particles are slowly delivered to the duodenum. [204], [205] This linear phase, which lasts 1 or more hours until the stomach is nearly empty, exhibits *zero-order kinetics;* in other words, lumenal contents leave the stomach at a constant rate that is independent of the volume remaining in the stomach. [206] Solid phase emptying terminates in a third phase of very slow transit of the last remaining particulate gastric contents. [204], [205] In general, solid emptying is slower in premenopausal women than in age-matched men regardless of the phase of the menstrual cycle (Fig. 10-9). [165], [207] This delay in women stems from a slower linear emptying rate, without differences in the lag period. [207], [208] and [209] In rats, oophorectomy accelerates emptying, indicating a physiological inhibitory role for hormonal factors. [210] Finally, studies show that the rate of solid emptying correlates positively with body mass index. [211]

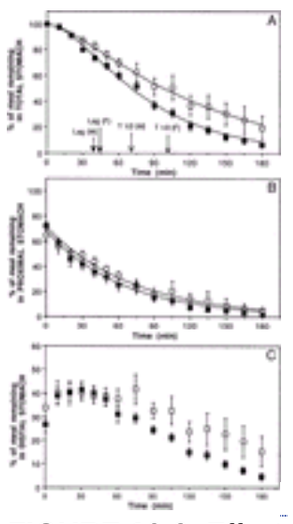

FIGURE 10-9. Effects of gender on solid phase gastric emptying in humans. Women (*open squares*) have delayed emptying compared with men (*filled squares*) as a consequence of a slower linear emptying phase (**A**). In contrast, there is little effect on the initial lag phase of solid emptying. In this study, there were no differences in proximal gastric retention of a solid meal (**B**); however, women exhibited prolonged distal gastric retention compared with men (**C**). (From ref. [207].)

Several theories have been proposed to explain the differential handling of solids and liquids by the stomach. The J shape of the stomach may promote selective retention of solids. [149] In this model, the liquid fractions of a meal are expelled through the pylorus by coordinated gastric motor activity, whereas the particulate components settle because of the effects of gravity. Other investigators suggest that solid particles empty more slowly because of basic fluid mechanics. In these theories, smaller particles are carried in the center of the fluid stream and are expelled with the liquid phase. In contrast, larger particles are propelled in the periphery of the fluid column, which moves more slowly than the center and does not reach the distal antrum until after pyloric closure. This produces retropulsion of larger particles until they can be reduced in size. In this model, contact of the particles with the antral walls is not required; rather, the shearing forces from sudden changes in fluid direction are sufficient to disperse the solids.

Factors that Modify Emptying of Digestible Solids Solid meals exhibit nutritional and physical properties that modify the rate at which they are delivered to the intestine. Radiolabeled eggs or noodles are passed into the duodenum faster than an equicaloric meal of 10-mm cubes of radiolabeled liver. [204], [205] Similarly, when eggs are fed as homogenized preparations, or as 2.5- or 5-mm cubes, the homogenized eggs empty most rapidly, whereas the 5 mm cubes are emptied most slowly. [212] These differences are caused by modulation of the initial lag phase. Larger particles prolong the lag phase, whereas evenly dispersed suspensions have relatively short lag phases. After completion of the initial lag period, a homogenized meal empties at the same linear rate as an unhomogenized one. In humans and dogs, solids are delivered to the duodenum as finely dispersed suspensions of particles less than 1 mm in diameter. [213] After the ingestion of solid chicken pieces, the mean diameter of recovered solid particles is 0.05 mm, and 95% are less than 0.5 mm in diameter. If homogenized food with a mean particle size less than 0.25 mm is consumed, the initial lag phase is nearly abolished. [204] Increasing the viscosity of a solid meal impairs the ability of the stomach to discriminate between large and small particles and leads to duodenal delivery of larger particles. The effects of posture on solid emptying are controversial—some studies show no effect and others

show delay, primarily by inhibiting the linear emptying phase. [187], [214] The caloric content and character of the solid food regulate the rate of solid emptying. Addition of fats, triglycerides, or carbohydrates such as glucose, fructose, or xylose to a solid meal delays its emptying, presumably by prolonging the lag phase. [215], [216] In contrast, if a low-calorie substance such as lettuce is added to a solid meal to enhance its volume but not its nutritive value, solid emptying is accelerated. [217] The quantity of liquid consumed with the solid meal alters the rate of duodenal delivery of solid nutrients. In a mixed solid-liquid meal, the liquids are emptied more rapidly than the solids, suggesting that the stomach is capable of distinguishing the two phases when solids and liquids are presented simultaneously. [218] Furthermore, the presence of liquid in the stomach prolongs the lag phase of solid emptying. [219] In fact, liquid consumption 90 to 180 minutes after a solid meal can blunt antral contractions and can induce a second lag phase of solid phase gastric emptying. [219], [220]

Roles of Different Physiological Regions in the Control of Solid Emptying Solid emptying results from the combined action of the pylorus and distal stomach. In humans who have undergone vagotomy and pyloroplasty, there is no increase in particle sizes delivered to the duodenum. [221] /SUP>Similarly, pyloric resection in dogs does not change particle sizes, indicating that the pylorus is not solely responsible for the controlled delivery of fine suspensions to the intestine. [131], [213] The rate of solid emptying is dependent on the magnitude of antral motor activity. [64] Humans who have undergone vagotomy and antrectomy deliver 30% of the solid meal residue to the duodenum as particles larger than 1 mm in diameter. [221] In these persons, solid phase emptying exhibits an early acceleration with loss of the lag phase. Similarly, in dogs, when the distal 3 cm of antrum and the pylorus are resected, 30% of the particles emptied by the stomach are larger than 1 mm in diameter. [131], [213] Control of particle size is regulated by coordinated antropyloric motor activity. These findings have important nutritional consequences. In dogs ingesting carbon 14 (^{14}C)-labeled fat within a chicken liver meal, 85% of the radiolabel is absorbed before reaching the midintestine. [222] If vagotomy and antrectomy are performed, only 43% of the radiolabel is absorbed after reaching the same site, suggesting that excision of the distal stomach can induce nutrient malabsorption. The roles of the proximal stomach and small intestine in controlling solid emptying are less thoroughly explored. As stated previously, 30% of particles delivered to the duodenum after a solid meal are larger than 1 mm in diameter after antrectomy with or without pylorectomy. Thus, 70% of food material is expelled as fine particles, suggesting that the proximal stomach has some capacity to disperse solid nutrients. [131], [213], [221] The ability of fats to slow solid emptying after antrectomy indicates that intestinal feedback persists in the absence of the distal stomach. [216]

Gastric Emptying of Fats

High-fat foods are handled differently by the stomach, and their emptying patterns are distinct from those of other liquids and solids. Many fats are solid or semisolid before ingestion. After consumption, they are warmed to body temperature and are converted to a liquid phase, which is emptied more slowly than nonlipid liquids. Emptying of fats exhibits an initial lag period, although solid and liquid lipids are handled separately. [223] Solid fats empty at a rate similar to that of liquid lipids in the first hour but are evacuated more slowly thereafter. [224] During the initial lag period, magnetic resonance imaging demonstrates marked increases in to-and-fro movements in the antrum that most likely enhance fat emulsification. [225]

The reasons for delayed emptying of fats are multifactorial. Fats have specific gravities less than 1 g/cm ^{3} and float on top of aqueous liquids in the stomach. [226] In contrast, nonlipid solids have specific gravities greater than 1.2 g/cm ^{3} and settle in dependent fashion. Fats are delivered to the duodenum at the same rate as low-density spheres; this reinforces the importance of specific gravity as a determinant of gastric emptying. Because of poor aqueous solubility, fats coalesce into large globules and fail to disperse into fine particles. Also because of their poor solubility, lipids adhere to solid food particles and thus are retained in the stomach until solid emptying proceeds. [223] Fats potently activate inhibitory enterogastric reflex pathways that further delay emptying. The products of lipid digestion appear to be more important for regulating emptying of fats, because patients with pancreatic exocrine insufficiency exhibit faster emptying of lipids than do healthy persons. [227] Studies in dogs and humans show that inhibition of lipolysis by orlistat accelerates gastric emptying of lipids, supporting this hypothesis. [228], [229]

Gastric Emptying of Indigestible Solids

Indigestible solids, the nonnutritive fibrous residue from a meal, are evacuated from the stomach in a fashion most dependent on particle size. Indigestible spheres smaller than 1 mm in diameter pass into the intestine during the fed period, often faster than solid nutrients. [230] Larger spheres pass more slowly, usually after an initial lag period, with spheres up to 3 mm in diameter passing with the calorie-containing solid meal components. [231], [232] Particles as large as 7 mm do not empty with solid food at all and are retained until the fed motor pattern terminates and gastric phase III activity resumes. The main characteristic that distinguishes phase III from the fed pattern is the presence of an open pylorus during fasting that permits intestinal delivery of large particles. In some reports, undigested materials as large as 2 cm in diameter can pass into the intestine during fasting. From a clinical perspective, particle size is relevant to the design of encapsulated medications. Drug absorption from 0.7-mm pellets is faster than absorption from 3.6-mm pellets, which are delivered to the intestine only with reinitiation of fasting motor activity. [233]

Other physical factors participate in the emptying of indigestible solids. Specific gravity is an independent determinant, with spheres denser than 2.0 g/cm ^{3} or less dense 0.5 g/cm ^{3} emptying more slowly than spheres with a density of 1.0 g/cm ^{3}. [230], [234] Viscosity of gastric contents plays a role in emptying of indigestible spheres. The addition of guar gum to increase viscosity accelerates emptying of 3.2-mm spheres. [235] Compressibility of the gastric compound is important, with soft indigestibles emptying faster than hard ones. [236]

Mediation of emptying of indigestible solids by the distal stomach has been demonstrated in canine models. Distal antrectomy accelerates emptying of indigestible spheres, although excision of the fundus has no effect. [237] Interruption of vagal innervation of the proximal stomach does not modify emptying of indigestible spheres, whereas distal gastric vagotomy delays their passage into the duodenum.

Differential Intragastric Distribution of Solids and Liquids

The differential emptying patterns for solids and liquids provide evidence that the stomach sequesters certain meal components, whereas others are processed for duodenal delivery. Fluoroscopy demonstrates retention of large pieces of ingested food in the fundus, although fine particles are mixed in the antrum. Quantitative scintigraphy shows constant levels of radioactivity in the antrum after a solid meal with progressive decreases in fundic activity, indicating that solids are stored proximally and are delivered to the distal stomach at constant rates for trituration. Studies of distribution of radiolabeled solids such as liver, chopped beef, or egg cubes indicate that the initial lag phase of solid emptying is characterized by antral filling to a maximal level of radioactivity that remains constant during the linear emptying phase while more proximal activity decreases. [212], [238]

Ingested liquids also are retained in the proximal stomach to maintain a constant volume in the distal stomach. [218], [239] Addition of lipid substances such as margarine to ingested liquids promotes redistribution into the fundus with associated delays in emptying. [240] The caloric content of a consumed liquid regulates proximal gastric storage. The antral diameter is greater after a nonnutritive meal than after ingestion of caloric liquids, and fundic retention is less. When liquids and solids are consumed together, the solid fraction is retained in the proximal stomach, whereas the liquid portion is delivered to the antrum. [241] If liquids are replaced by carbonated water, the proximal stomach initially retains a greater proportion of the solid meal, suggesting that gaseous distention of the stomach also modifies intragastric distribution. [242] The size of the solid meal influences the rate of liquid emptying. A large solid meal retards the transfer of liquid from the proximal stomach to the antrum and delays the delivery of liquid to the duodenum. [243]

Indigestible solids are distributed separately from digestible solids. When liver is ingested along with 2.4-mm-diameter plastic spheres, the liver is retained within the fundus, whereas the spheres are transported to the distal stomach, where they remain until the late postprandial period.

External Regulation of Gastric Emptying

There is extensive modulation of gastric emptying by external influences. Feedback inhibition of emptying by intestinal factors is crucial to regulating duodenal caloric delivery. Additional control is provided by extrinsic and intrinsic nerves and from hormones and neurotransmitters. Finally, gastric motor function is subject to modulation by central nervous system factors.

Feedback Inhibition from Distant Sites Feedback from the small intestine is a major regulator of gastric emptying. Duodenal pH receptors are postulated to mediate inhibition of gastric emptying evoked by duodenal acidification. [244] Duodenal perfusion of nutrients such as amino acids nearly abolishes solid emptying and also reduces liquid emptying. [245] If a duodenal fistula is placed to divert nutrients away from intestinal mucosal receptors, liquid emptying exhibits first-order kinetics, in contrast to the curvilinear pattern seen with an intact enterogastric reflex. Intestinal nutrient perfusion modifies solid emptying by altering the duration of the lag phase. Duodenal amino acid perfusion enhances the sieving properties of the stomach and pylorus and increases the percentage of particles less than 1 mm in diameter delivered to the duodenum from 66% to 82%. [245] Many agents act on intestinal osmoreceptors to increase duodenal outflow resistance and to delay emptying. [246] Inhibition of gastric emptying by lipids appears to require the intralumenal presence of component fatty acids, because intestinal perfusion of nonhydrolyzable fats does not inhibit gastric emptying. [178] Duodenal lipid-evoked inhibition of gastric emptying may also require the generation of chylomicrons, because administration of surfactants that prevent chylomicron formation abolishes this enterogastric reflex (Fig. 10-10). [247]

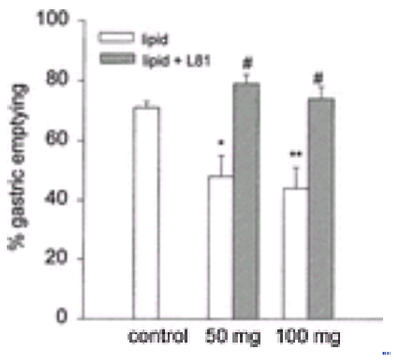

FIGURE 10-10. Effects of Pluronic L-81, a detergent that inhibits chylomicron formation, on the retarding of gastric emptying by duodenal lipid perfusion. Lipid perfusion significantly delays gastric emptying in rats. This inhibition is prevented by treatment with two doses of L-81, indicating that the effects of lipid are mediated by generation of chylomicrons. (From ref. 247.)

The degree of inhibition of gastric emptying by intestinal feedback depends on the length of intestine exposed to the stimulus. Osmoreceptors mediating hypertonic saline–evoked inhibition of emptying are located only in the duodenum, whereas acids, lipids, and glucose have inhibitory effects on longer segments of small intestine. 184, 248 Maximal inhibition of liquid emptying is seen with exposure of the proximal 150 cm of small intestine to acid, glucose, or oleic acid. 169, 249 However, perfusion of only the proximal 15 cm of duodenum with hydrochloric acid or oleic acid, but not glucose or lactic acid, delays delivery of liquids into the intestine. 250 Capsaicin-sensitive afferent nerves mediate enterogastric feedback regulation of gastric emptying in response to some stimuli. Intestinal capsaicin perfusion abolishes the inhibition of emptying induced by intestinal hydrochloric acid, but it reduces the inhibition evoked by duodenal glucose or lipid by only 59% and 42%, respectively. 251 Similarly, vagal capsaicin attenuates lipid-induced inhibition of gastric emptying by 57%. 252 Capsaicin also prevents the inhibitory effects of mechanical stimuli such as duodenal balloon inflation on gastric emptying of saline. 253 Regions other than the proximal small intestine regulate gastric emptying. Nonpainful rectal balloon inflation slows emptying of solids. High-fat soups delay gastric emptying more when ingested orally than when given by intragastric infusion, indicating modulation by orosensory stimulation. 254 Gastric emptying of solids is potently inhibited by glucose or oleic acid perfusion of the ileum, in a phenomenon known as the *ileal brake*. 255 The involvement of opiate pathways is suggested by studies showing blockade of the ileal brake by naloxone. 256 Certain lipids are capable of activating the ileal brake (i.e., petroselinic, oleic, myristoleic, erucic, linoleic, and linolenic acids, corn oil, lecithin, and deoxycholic acid), whereas others cannot (e.g., taurocholic acid). 257

Neurohumoral Modulation

Role of extrinsic and intrinsic innervation in control of gastric emptying. Intact extrinsic innervation is essential for normal gastric emptying of solids and liquids. Thoracic spinal cord transection acutely delays emptying of nutrient liquids. 258 Truncal or proximal vagotomy produces rapid duodenal delivery of liquids. 237, 259 The addition of pyloroplasty enhances the rapid liquid emptying after vagotomy. 202 This rapid emptying is more pronounced when the person is erect than when the person is supine, indicating that the vagus controls postural effects on gastric motor function under normal conditions. 260 In contrast, truncal vagotomy retards overall emptying of digestible and indigestible solids and can lead to bezoar formation in some patients, although the earliest phase of solid emptying may be accelerated. 260, 261 Proximal vagotomy has little effect on solid phase emptying. 259 The intrinsic innervation of the distal stomach also regulates liquid emptying. If the antrum is transected 2 cm proximal to the pylorus and is then reanastomosed, transpyloric flow pulse volumes are reduced and gastric emptying is retarded. 262 This operation does not modify the inhibition of gastric emptying, the inhibition of antral contractions, or the stimulation of pyloric motor activity evoked by duodenal glucose, indicating that intrinsic gastric pathways are not involved in enterogastric reflex activities. However, the demonstration that duodenal transection in pigs impairs nutrient-evoked delay of gastric emptying indicates that intrinsic duodenal neurons participate in enterogastric reflex inhibition. 203

Roles of specific neurohumoral transmitters in control of gastric emptying. Many hormones and neurotransmitters modify gastric emptying, but their physiological relevance is largely unknown. The role of NO in modulating emptying appears to be species dependent. In humans, NO donors delay gastric emptying, whereas NO synthase inhibitors have accelerating properties. 134, 263 In rodent models, NO synthase inhibitors delay gastric emptying by modulating intestinal feedback control. 264 The delay in emptying that occurs with NO synthase inhibition in rodents is associated with increased pyloric and duodenal contractions; therefore, NO may facilitate gastric emptying by inhibiting obstructing pyloroduodenal motor activity. 265 Opiates of different receptor subclasses have inhibitory and excitatory effects on gastric emptying through naloxone-sensitive and naloxone-insensitive pathways. 266 The ability of naloxone to accelerate gastric emptying indicates an inhibitory role for endogenous opioid pathways in controlling gastric emptying. Emptying delays induced by meperidine are reversed by the a-adrenergic receptor antagonist phentolamine, suggesting mediation by adrenergic pathways. 267 Modulation of emptying by serotonergic pathways likely depends on the receptor subtype that is activated. 5-HT $_3$ receptor antagonists and 5-HT $_1$ agonists, such as sumatriptan, delay gastric emptying; this action indicates inhibitory effects of 5-HT $_3$ pathways and excitatory actions of 5-HT receptors. 268, 269 The enhancement of emptying by the histamine (H $_2$) antagonist nizatidine suggests that endogenous histamine may serve as a physiological inhibitor of gastric emptying. 270 Studies demonstrating acceleration of emptying after administering antibodies to secretin suggest that secretin may modulate intestinal nutrient delivery. 271 Blockade of GRP receptors delays emptying of solids, and this also indicates a physiological role for GRP. 272 Antibodies to calcitonin gene–related peptide (CGRP) prevent the inhibition of gastric emptying by intestinal hydrochloric acid or hypertonic saline, suggesting participation of CGRP pathways in enterogastric reflex inhibition. 273 Agents that delay gastric emptying include gastrin, secretin, somatostatin, neurotensin, peptide YY, enteroglucagon, oxyntomodulin, prostaglandin E $_1$ and E $_2$, neuromedin B, amylin, glucagon-like peptide I, epidermal growth factor, transforming growth factor a, and interleukin-1 ß. 38, 109, 274, 275, 276, 277 and 278

Role of cholecystokinin in emptying of nutrient meals. CCK, released by duodenal lipids or proteins, has been extensively investigated as a physiological regulator of gastric emptying; however, its specific role remains incompletely defined. Physiological CCK infusions inhibit liquid emptying and stimulate antral motor activity, whereas higher doses increase pyloric contractions. 74, 279 CCK infusions that mimic the postprandial state produce fundic relaxation and increase proximal gastric compliance. 280 The inhibitory response of the stomach to CCK exhibits plasticity depending on prior dietary exposure. Oleate-induced inhibition of gastric emptying is attenuated in rats fed a high-fat diet. 281 In the same model, exogenous CCK administration less potently inhibits gastric emptying, indicating a loss of sensitivity to the peptide (Fig. 10-11). The physiological significance of these different effects is uncertain, and their mediation is poorly characterized. Antrectomy, in combination with pyloroplasty, inhibits the effects of physiological CCK infusions on gastric emptying. Surgical resection of the fundus prevents CCK inhibition of emptying in some studies but has no effect in others. 282 However, serosal application of the myenteric denervating agent benzalkonium chloride does not prevent the inhibition of gastric emptying by CCK, suggesting that antral neurons are not involved. 283

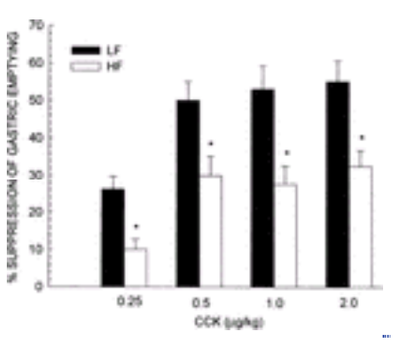

FIGURE 10-11. Gastric emptying of saline is inhibited by cholecystokinin (*CCK*) in rats fed a low-fat (*LF, dark bar*) and high-fat (*HF, white bar*) diet. For rats fed a low-fat diet, CCK inhibits gastric emptying in dose-related fashion. This inhibitory effect on emptying is blunted by the long-term ingestion of a high-fat diet. (From ref. 281.)

Although not universally accepted, the weight of evidence is leaning toward an intrinsic role for CCK in the inhibition of gastric emptying, based on investigations using selective CCK receptor antagonists. The CCK antagonist loxiglumide usually accelerates emptying of mixed meals, glucose, lipids, and radiopaque markers, although some studies have reported no enhancement. 284, 285, 286 and 287 Enhancement of distal gastric contractions by loxiglumide has been observed, indicating that CCK may regulate gastric emptying by inhibiting antral motor function. 288 Similarly, the potent CCK antagonists L364,718, devazepide, and linitript accelerate gastric emptying in some investigations but not others (Fig. 10-12). 141, 289, 290, 291, 292 and 293 One study using L364,718 demonstrated blockade of the enterogastric inhibition of gastric emptying by duodenal maltose perfusion. 294 The ability of devazepide to blunt the retardation of emptying evoked by a peptone meal decreased as the caloric density increased, indicating the involvement of CCK-independent factors with greater nutrient loads. 295

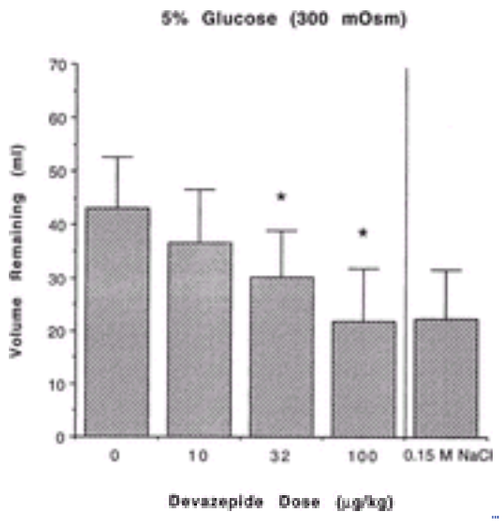

FIGURE 10-12. Volume of a glucose test meal remaining in the stomach 10 minutes after several doses of the cholecystokinin receptor antagonist devazepide. Devazepide produces a dose-dependent acceleration of gastric emptying, with the highest dose producing emptying rates comparable to those of a nonnutritive saline load. (From ref. 289.)

CCK effects on gastric emptying likely are mediated by action on vagal afferents, because perivagal capsaicin abolishes the delay in emptying evoked by CCK. 45 In rats, intraarterial or intraperitoneal CCK increases firing in gastric vagal mechanoreceptor afferents. 296 The ability of devazepide to block the increase in firing rates of gastric vagal afferents evoked by CCK indicates mediation by CCK-A receptors. 297 In another study, CCK amplified vagal afferent discharges induced by duodenal nutrients, indicating an integrative response. 298 Some investigations suggest the additional involvement of splanchnic pathways. 279 Devazepide attenuates the inhibition of gastric emptying evoked by duodenal lipids. 252 However, that there is no additive effect of devazepide and vagal capsaicin on preventing the inhibitory effects of lipids suggests that CCK acts solely on vagal afferent CCK-A receptors in this model. The actions of CCK on gastric emptying are mediated by the release of secondary transmitters that then act on selected gastric sites. Studies in cats show that CCK-evoked catecholamine release from splanchnic efferent nerves produces excitatory and inhibitory effects on gastric muscle through action on a_2- and ß-adrenergic receptors, respectively. 299 In rats, serotonin (i.e., 5-$HT_{2A/2C}$) receptors are postulated to mediate the inhibitory effects on gastric emptying of endogenous CCK released by intestinal lipid perfusion. 300

Central Nervous System Modulation Central nervous system input is a crucial regulator of gastric motor function. Noxious noise modifies fasting gastric contractions in dogs, and mental stress prolongs MMC cycling in humans. 301 Physical restraint of rats replaces the MMC with irregular, continuous phasic contractions. The fed motor pattern is affected by acoustic stress and by immersion of the hand in cold water. 302 Cold stress induces isolated pyloric contractions, inhibits antral contractions, stimulates duodenal phase III, and prolongs liquid emptying. 303, 304 Experimentally induced vertigo, noise stress, restraint stress, cold pain, and ischemic pain all delay gastric emptying. 302, 305 In humans, gastric motor activity is increased during anger and is decreased during fear and depression. Multiple neurohumoral pathways participate in these stress responses, as demonstrated by the associated release of norepinephrine and endorphins and by their blockade after truncal vagotomy or with antagonism of adrenergic or opioid receptors. 306, 307 Corticotropin-releasing factor (CRF) has been extensively investigated as a prominent mediator of stress effects on gastric motor function. Infusion of CRF into the brain or spinal cord delays gastric emptying through a vagal cholinergic pathway. 308, 309 Some stressors evoke intracerebral CRF release. The peripheral effects of many stressors are inhibited by prior administration of CRF receptor antagonists, suggestive of a physiological role for this peptide. 309, 310 and 311 The ability of the selective CRF_2 ligand urocortin to disrupt gastric motility when given intracerebroventricularly and the failure of CRF_1 receptor antagonists to block the effects of CRF or urocortin suggest that the CRF_2 receptor mediates the central responses to CRF. 312, 313 Centrally administered CRF antagonists reverse intracisternal interleukin-$1_ß$–evoked delays in gastric emptying, indicating a possible physiological interaction of inflammatory mediators with central CRF pathways. 314 The ability of a somatostatin analog infused into the fourth ventricle to block the effects of CRF on gastric emptying is suggestive of mediation by endogenous somatostatin pathways. 315 Central neural NO pathways can modulate physiological fasting gastric motor activity. Inhibition of central NO synthase suppresses gastric and duodenal phase III but has no effect on jejunal cycling or on solid emptying; these findings indicate differential central regulation of proximal and distal motor function and on fasting and fed contractile patterns (Fig. 10-13). 316 Central administration of NO synthase inhibitors increases tonic and phasic pyloric motor activity. 317 The ability of truncal vagotomy to block this effect indicates mediation by vagal pathways. The inability of the ganglionic blocker hexamethonium to prevent the gastric relaxation evoked by NO synthase inhibition implies that nonnicotinic pathways participate in this response. 318 The presence of NO synthase activity in the dorsal vagal complex of the brainstem implies a role for NO in the afferent regulation of gastric motor function. 319

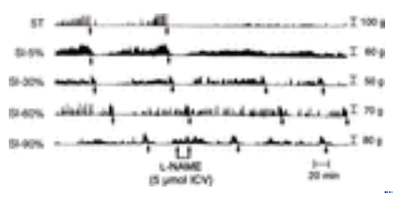

FIGURE 10-13. Effects of intracerebroventricular (*ICV*) injection of the nitric oxide (*NO*) synthase inhibitor *N* G-nitro-L-arginine methyl ester (*L-NAME*) on gastroduodenal motor activity are shown. Central L-NAME inhibited phase III activity in the stomach (*ST*) and duodenum but not in the distal small intestine (*SI*). (From ref. 316.)

Other central nervous system neurotransmitters modulate gastric motor function as well. Central TRH administration increases gastric motor activity by action on vagal pathways. 320, 321 The excitatory effects of TRH are abolished by intracisternal injection of TRH mRNA antisense oligonucleotides, which block the function of TRH. 322 TRH antibodies also block stimulation of gastric contractions evoked by central injection of glutamate, and TRH receptor 1 antisense oligonucleotides block the delay in gastric emptying evoked by cold stress, suggesting that TRH may represent a physiological activator of gastric motility in response to varied central nervous system stimuli. 323, 324 Oxytocin infusion into the dorsal motor nucleus of the vagus reduces gastric motor activity, whereas central administration of an oxytocin antagonist increases fasting gastric activity. 325 Oxytocin receptor antagonists also prevent the inhibition of gastric motor activity evoked by electrical stimulation of the paraventricular nucleus of the hypothalamus, suggesting a possible physiological role for oxytocin as a central inhibitor of gastric motor activity. Central adrenomedullin infusion inhibits gastric emptying through action on adrenal-dependent, CRF- independent, ß-adrenergic pathways. 326 Central administration of CCK, opiates, bombesin, tachykinins, atrial natriuretic factor, ?-aminobutyric acid, calcitonin, CGRP, substance P, or peptide YY inhibits gastric emptying or motility, in many instances through vagal pathways. 308, 320, 327, 328 Conversely, pituitary adenylate cyclase–activating polypeptide-38, pancreatic polypeptide, serotonin, and glutamate increase gastric motor activity when they are administered into selected brainstem nuclei. 329, 330 An important role for the brain-stomach axis is the regulation of food intake. The effects of centrally acting agents on gastric motor function may play pivotal roles in controlling appetite. CCK acting on CCK-A receptors in the gut suppresses food intake through activation of vagal pathways projecting to the nucleus tractus solitarius, lateral parabrachial nucleus, amygdala, and higher sites. 331 Rats lacking CCK-A receptors develop obesity, indicating the importance of CCK as a satiety hormone. 331 When given peripherally, urocortin decreases food intake in lean and obese mice in association with delays in gastric emptying. 332 Leptin is a hormone secreted by adipose tissue and gastric mucosa that plays a prominent role in the control of food intake. 333 When injected into the cerebral ventricles, leptin inhibits gastric emptying through leptin receptors on cholinergic neurons in the dorsal motor nucleus of the vagus. 334, 335 Leptin applied to the stomach activates a subpopulation of vagal afferent neurons that also respond to CCK. 333 When given together, leptin and CCK have an additive effect on vagal discharges (Fig. 10-14). These findings suggest that leptin may modulate the effects of CCK to regulate food intake. Conversely, ghrelin, a peptide secreted by the stomach, was shown to decrease gastric vagal afferent activity and to increase gastric motor activity in rats. 336 Furthermore, ghrelin may act through Y-1 receptors for neuropeptide Y expressed in the arcuate nucleus to increase food intake and to decrease energy use. 336 Ghrelin levels are suppressed by leptin, suggesting that this gastric peptide may act downstream from leptin to control appetite.

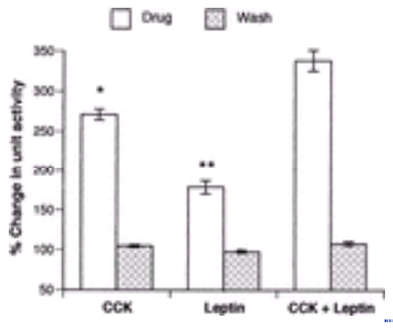

FIGURE 10-14. Effects of cholecystokinin (*CCK*) and leptin on firing of nucleus tractus solitarius neurons receiving gastric vagal afferent input. Both CCK and leptin produce increases in afferent vagal firing; when given together, their effects are subadditive. (From ref. 333.)

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CHAPTER 11

William L. Hasler

MOTILITY OF THE SMALL INTESTINE AND COLON

The motor activities of the small intestine and colon are characterized by contractile patterns that serve the requirements of each organ. The small intestine processes and absorbs nutrients for distribution to the rest of the body, whereas the colon extracts water, digests some meal residue, and processes feces for expulsion. Two basic motor patterns, mixing and propulsion, subserve these functions. In the small intestine, triturated food from the stomach is mixed with bile and pancreatic enzymes for digestion and is propelled aborally. During fasting, mixing and propulsion clear the small intestine of undigested solids and sloughed enterocytes. In the colon, prominent mixing patterns afford time for the slow process of fecal desiccation, and less organized propulsion patterns result in slow transit.

Two sphincters in the lower gut regulate transit within and expulsion from the gut lumen. The ileocecal junction prevents reflux of feces from the cecum to the ileum, but its role in modulating delivery of ileal contents to the colon is less clear. The internal and external anal sphincters in concert with pelvic floor muscles control evacuation of solid waste from the body until it is socially convenient. In addition, sphincteric functionality has been attributed to the rectosigmoid junction. [1] Anatomic analyses of this region show a mucosal “rosette” nearly 3 cm in length associated with a narrowed contractile segment that halts fecal transit from the sigmoid colon to the rectum. [2]

Motility of the small intestine, colon, and sphincteric regions is controlled by myogenic characteristics, extrinsic and intrinsic nerves, and circulating hormones. External influences from meals, during sleep, with central nervous system activation, or from immune factors modulate this contractile function, as do effects of intralumenal stimuli.

ANATOMIC CONSIDERATIONS

The small intestine is about 600 cm long, including the duodenum, and the jejunum (40% of the length), and the ileum (60%). Its wall has two regions of muscle tissue, the muscularis externa and the muscularis mucosa. The muscularis externa consists of an outer longitudinal layer and an inner circular layer, oriented at 90° angles to each other. The circular layer is subdivided into inner and outer layers. The muscularis externa is the major effector of mixing and propulsion, whereas the role of the muscularis mucosa is poorly understood. Circular muscle thickening is observed at the ileocecal junction.

The structure of the colon is very different from that of the small intestine. The ileocecal junction attaches to the cecum, a sac-like structure that likely serves a storage function. The ascending, transverse, and descending regions possess a circumferential circular muscle layer with three overlying longitudinal muscle strips, the teniae coli, 120° apart. Haustra are produced by circular and longitudinal muscle contractions that narrow the lumen and shorten the length of the colon. The longitudinal muscle envelops the circumference of the rectosigmoid colon. The rectum exhibits transverse folds (usually two to three) extending beyond the midline of the rectal lumen that contain mucosa as well and circular and longitudinal muscle. [3] These structures provide a shelving function to retard fecal passage.

The anus is a specialized region with both smooth and striated muscle. The internal anal sphincter is a thickened projection of the rectal circular smooth muscle. Rectal longitudinal fibers fan out at the anal verge, pass through the sphincter muscles, and insert in the perianal subcutaneous tissue. The external anal sphincter, a striated structure, is composed of three muscles: the deep external sphincter, the superficial external sphincter, and the subcutaneous external sphincter. The levator ani muscles (puborectalis, pubococcygeus, and iliococcygeus) form a sling to surround the distal rectum and maintain continence when they are contracted.

SMALL INTESTINAL AND COLONIC SMOOTH MUSCLE

Cellular and Electrical Characteristics

Small intestinal and colonic muscle cells are spindle shaped and uninucleate. In contrast to skeletal muscle, there are no specialized regions for neuronal interaction. These cells are electrically active with resting membrane potentials of -40 to -80 mV that are maintained by Na^+,K^+-ATPase activity, and they exhibit spontaneous contractions activated by stretch. Small intestinal myoelectric activity exhibits ubiquitous membrane potential fluctuations of 3 to 15 mV oscillating at 11 to 12 cycles per minute (cpm) in the duodenum and at lower frequencies in distal regions. Extracellular recordings of this slow wave activity show sinusoidal patterns or rapid biphasic deflections from zero potential. Intracellular recordings demonstrate rapid depolarizations followed by partial repolarizations to prolonged plateau phases of depolarization. The slow wave cycle terminates with full repolarization to the resting membrane potential. In vivo slow wave activity also is present in the colon and cycles in two frequency ranges, from 2 to 6 cpm and from 9 to 13 cpm. [4] Colonic slow waves mediate short duration contractions (SDCs). In addition, rapid electrical oscillations at frequencies of 25 to 40 cpm, termed *myenteric potential oscillations,* regulate long-duration colonic contractions. [5], [6] Excised human colon circular muscle strips exhibit spontaneous slow waves of 12 mV in amplitude, 9.4 seconds in duration, and at frequencies of 2 to 4 cpm. [7] Near the myenteric edge of these strips, rapid membrane potential fluctuations at a frequency of 18 cpm are observed.

Smooth Muscle Contraction

In the small intestine, phasic contractions are controlled by the slow wave, although tonic contractions lasting 10 seconds to 8 minutes have been recorded from the circular muscle. [8] In most instances, the slow wave produces insufficient membrane depolarization to initiate contraction. Neurohumoral stimuli or exogenous administration of excitatory agonists evoke motor activity by increasing the duration and amplitude of the slow wave plateau potential or by inducing spike potentials of brief duration (10–100 ms) but high amplitude (50 mV) in phase with the slow wave. Spike potentials generated in isolated longitudinal and circular muscle propagate only a few millimeters. Intestinal relaxation results from removal of a contractile stimulus or application of an active relaxant agent that acts to inhibit spike potentials,

to reduce plateau potential amplitude or duration, or to induce membrane hyperpolarization.

Myoelectric regulation of colonic motor function is more complicated. In colonic circular muscle, contractile activity is generated from the summation of slow wave activity generated along the submucosal border and myenteric potential oscillations generated along the myenteric border. [6] This summed activity is responsible for the complex waveforms observed in colonic recordings. Contractile agonists evoke colonic contractions by increasing myenteric potential oscillation amplitude, enhancing slow wave plateau potential duration and amplitude, or generating spike potentials. [9] Relaxant agents inhibit myenteric potential oscillations, reduce slow wave activity, or evoke membrane hyperpolarization.

Interstitial Cells of Cajal as Generators of the Slow Wave

The interstitial cells of Cajal (ICCs) are believed to be responsible for generation of the slow wave. ICCs with pacemaker properties are localized to the interface of the circular and longitudinal muscle in the small intestine and along the submucosal surface of the circular layer of the colon. A second population of ICCs in the colon is present at the interface of the circular and longitudinal layers and may underlie the generation of myenteric potential oscillations. [6] ICCs are uninucleate with thin cytoplasms and numerous mitochondria indicative of high metabolic activity, as well as abundant surface caveolae with prominent endoplasmic reticula inferring active ion transport. ICCs are distinct from neurons, glia, fibroblasts, and myocytes. [10] In humans, their greatest density is in the transverse colon, which is postulated to represent the physiological pacemaker site. [11]

Substantial evidence points to ICCs as generators of spontaneous electrical rhythmicity. In cat intestine, rhythmic oscillations are generated only by tissue containing ICCs. [12] In colonic circular muscle, slow waves are maximal in amplitude at the submucosal border and become weaker in the myenteric region, indicating the importance of submucosal ICCs. Intracellular recordings from ICCs in canine colon show cyclic oscillations 37 mV higher than the resting membrane potential at 4.6 cpm. [13] Similarly, ICCs from mouse small intestine exhibit a frequency of 14 cpm with a mean amplitude of 13 mV. [14] Cyclic fluctuations in intracellular calcium underlie this electrical rhythmicity. [15] ICC-generated slow waves are reduced by the removal of extracellular calcium but are insensitive to L-type calcium channel blockers. [14] Rhythmicity in cultured ICCs is dependent on calcium release from intracellular stores. [16], [17] Selective injury to ICCs by administration of methylene blue plus intense illumination disrupts the slow wave and provides evidence of the pacemaker capabilities of these cells. [18]

Intrauterine ICC maturation requires signaling by the c-Kit tyrosine kinase receptor. [19] In human fetal small intestine, c-Kit immunoreactivity is demonstrable at a gestational age of 13 weeks. [20] A continuous layer of ICCs forms from week 17 to birth. Staining for c-Kit and smooth muscle myosin heavy chain, investigators have suggested that ICCs and longitudinal muscle cells may share a common mesenchymal progenitor cell. [21] Down-regulated in cells destined to become muscle cells, c-Kit is up-regulated in cells that become ICCs. [22] ICCs mature normally in glial cell line–derived neurotrophic factor knock-out mice that exhibit no enteric neurons, indicating that the enteric plexuses are not required for ICC development. [23] Studies measuring c-Kit expression provide the most convincing support for ICCs as physiological regulators of the slow wave. Mice with mutations in the kit gene lack ICCs and exhibit impaired peristalsis. [24], [25] In animals with mutations in the steel factor, the ligand for c-Kit, ICCs are absent in the small intestinal myenteric plexus, and slow wave cycling is not detected (Fig. 11-1). [19] Finally, in neonatal mice, administration of neutralizing monoclonal antibodies to c-Kit abolishes small intestinal rhythmicity and reduces the number of ICCs, which then assume the appearance of smooth muscle cells. [26], [27]

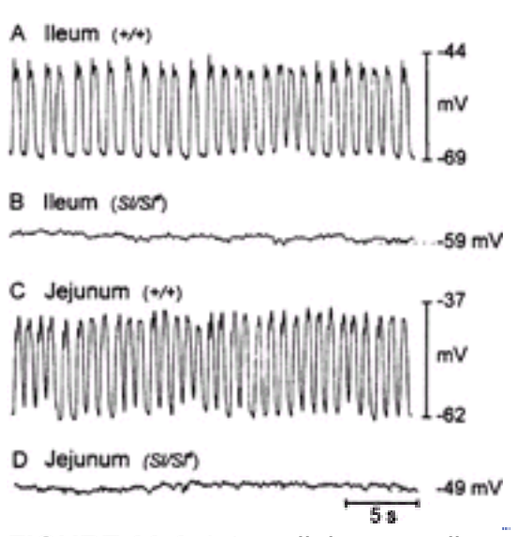

FIGURE 11-1. Intracellular recordings from circular smooth muscle cells from mouse small intestine. In tissue from wild-type mice (+/+), typical slow wave activity is prominent both in the ileum (**A**) and jejunum (**C**). In contrast, steel mutants (*Sl/Sl* ^{d}) which lack the ligand for c-Kit, exhibit no evidence of spontaneous slow waves in either the ileum (**B**) or jejunum (**D**). (From ref. [19].)

Coupling of Small Intestinal and Colonic Contractions

Migration of small intestinal myoelectric and motor activity is regulated by intrinsic electrical coupling properties. Intestinal slow wave propagation requires continuity of both muscle layers. After excision of a 1-mm band of circular muscle, longitudinal slow wave propagation is limited to distances of 3 to 6 mm. [28] Removal of more than 5 mm of longitudinal muscle abolishes slow wave propagation, whereas excision of shorter bands permits inefficient propagation. Significant coupling exists between longitudinal and circular muscle cells in the small intestine. In the circular layer, gap junctions or nexuses provide low-resistance pathways for electrical conduction and for passage of low-molecular-weight second messenger compounds. Close coupling also is observed in the longitudinal layer, which possesses few or no nexuses. In this layer, other structures such as peg-and-socket junctions may mediate intercellular communication. [29] Interconnecting bridges between the circular and longitudinal layers may provide low-resistance coupling across the myenteric region. Furthermore, ICCs are extensively innervated and form close electrical contacts with smooth muscle cells from both layers. Research suggests that gap junctions may not be needed for the pacemaking function of ICCs; rather, ICC coupling may use a syncytium of circular muscle cells. [30] In the deep muscular plexus of the circular layer, specialized ICCs form close junctions with smooth muscle cells as well as nitric oxide (NO) synthase–like and substance P–like immunoreactive axonal varicosities; these findings suggest that ICCs may provide a pathway for nerve-muscle communication. [31], [32] Furthermore, ICCs express M_2 and M_3 muscarinic receptors, NK_1 and NK_3 neurokinin receptors, VIP_1 vasoactive intestinal polypeptide receptors, and stem cell factor, indications of modulation by neuronal pathways. [33]

Because small intestinal smooth muscle coupling is close, cells with the highest slow wave frequency entrain adjacent cells in a phenomenon termed *phase lock.* In humans, the dominant pacemaker extends from the pylorus to the ligament of Treitz. Slow wave propagation is faster in the longitudinal axis (7–10 cm/s) than in the transverse axis (1 cm/s). [34] Uncoupling of slow wave synchrony develops over the long distances encountered in the small intestine and is more pronounced in the ileum than in the duodenum. [35] The human intestinal slow wave frequency decreases from 11 to 12 cpm in the duodenum to 7 to 8 cpm in the distal ileum. In most species, the decrease in frequency from the duodenum to the ileum occurs in stepwise fashion, producing alternating frequency plateaus and regions of variable frequency. Frequency plateaus are prominent more proximally but are often undetectable in the ileum. [35] Consequently, there are expansive regions of myoelectric coupling in the upper gut that promote long distance contractile propagation, whereas uncoordinated ileal slow waves limit the migration distance. In addition to regional variations in frequency, slow wave propagation velocities decrease by up to 50% from the duodenum to the distal ileum. These phenomena are advantageous for efficient digestion. Proximally, nutrients are propelled over a large mucosal surface area for rapid digestion and absorption, whereas distally, retarded propulsion permits absorption of slowly digested and absorbed substances such as fats and bile.

In the colon, slow wave migration requires continuity of the submucosal aspect of the circular muscle. [36] The slow wave is considerably less well entrained across different colonic regions compared with small intestinal myoelectric complexes because of a lesser degree of intercellular electrical coupling. Colonic slow wave frequencies and propagation velocities are highly variable, in part because of a marked reduction in circular muscle nexus density as compared with the small

intestine. [37] Unlike in the small intestine, there is no decrement in slow wave frequency from the ascending colon to the rectum. Colonic slow wave propagation is faster circumferentially (1.6 cm/s) than longitudinally (1–5 mm/s), providing myoelectric coordination of slow propagating segmenting ring contractions. [38], [39]

In addition to regulating peristalsis, slow wave activity generated by the ICCs controls the nonpropagative mixing activities of the gut. Mixing movements within the longitudinal layer are the consequence of spontaneous calcium waves generated by pacemaker sites that spread in all directions and that terminate by collision with calcium waves from other pacing sites (Fig. 11-2). [40]

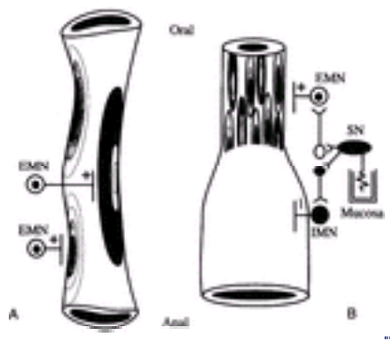

FIGURE 11-2. Neural mechanisms underlying mixing (**A**) and peristalsis (**B**). Mixing is regulated by pacing sites that propagate over large regions evoking localized shortening. Peristalsis results from activation of polarized nerve pathways that induce propagation orad to and suppress propagation caudad to the stimulus. *EMN,* excitatory motor neuron; *IMN,* inhibitory motor neuron; *SN,* sensory neuron located in the mucosa. (From ref. [40].)

Relative Functions of Longitudinal and Circular Muscle

The distinct functions of longitudinal and circular muscle in the small intestine are poorly understood. The circular layer is believed to mediate both mixing and propulsion, because lumenal occlusion and displacement of gut contents require contraction of this layer. The most basic small intestinal contractile pattern, segmentation, results from reciprocal inhibition and disinhibition of adjacent circular muscle. Although longitudinal muscle likely does not have potent propulsive capabilities, contraction of this layer theoretically could shorten the gut and accelerate transit. Longitudinal contraction also can increase lumenal diameter to facilitate movement of large boluses. The timing of contractions in the two layers is controversial; some investigations have observed concurrent motor activity, and others have reported 90° to 270° phase lags between circular and longitudinal contractions. [41], [42] In guinea pig ileum, independent contractions of the two layers are noted at low intralumenal pressures, whereas simultaneous contractions occur at higher pressures. [43] Spatiotemporal mapping has shown the contraction of the longitudinal layer immediately preceding and during the propulsive period of circular muscle contraction. [44] However, when the circular contraction occluded the lumen, longitudinal lengthening occurred.

Less is known about the functions and timing of circular and longitudinal muscle in the colon. Circular muscle contractions are lumenally occlusive, whereas those of the longitudinal layer may shorten the colon. Longitudinal contraction was recorded preceding circular contraction in one study, although other investigators observed similar electrical responses of the two layers during contraction. [45] A model of peristalsis has demonstrated synchronous contraction orally and relaxation aborally of the longitudinal and circular layers in response to a lumenal stimulus. [46]

INNERVATION OF THE SMALL INTESTINE AND COLON

Gut smooth muscle is innervated by extrinsic nerves relaying information to and from the extraintestinal ganglia, the spinal cord, and the central nervous system and by intrinsic nerves within the intestinal wall. The vagus and splanchnic nerves supply the small intestine, ileocecal junction, and proximal colon, whereas the pelvic nerves provide input to the colon and internal anal sphincter. Transcranial magnetic stimulation reveals bilateral cerebral motor cortex representation in the innervation of the anus and rectum. [47] The external anal sphincter and pelvic floor muscles receive sacral spinal input from the pudendal nerves. [48] The myenteric plexus provides the major intrinsic innervation to the small intestine and colon, although the submucous plexus may play a minor role in some reflexes. The number of intrinsic neurons greatly exceeds the number of vagal, pelvic, or splanchnic fibers. The human enteric nervous system contains 10 to 100 million neurons versus 2,000 efferent fibers in the vagus; thus, most motor activities are directed by intrinsic nerves, whereas extrinsic innervation provides a modulatory function.

Extrinsic Innervation

Efferent Neural Pathways Efferent extrinsic fibers are carried in parasympathetic and sympathetic tracts. Most efferent fibers terminate in the myenteric plexus and form connections with enteric ganglia, although some sympathetic axons terminate directly on sphincteric smooth muscle. The vagus nerves contain three groups of efferent fibers: preganglionic parasympathetic excitatory cholinergic nerves to the enteric plexuses, preganglionic inhibitory cholinergic nerves to the myenteric plexus, and sympathetic fibers from the cervical ganglia. Efferent vagal cholinergic neurons activate nicotinic receptors within enteric ganglia with excitation of motor activity. In general, these fibers exhibit a low threshold to electrical stimulation. The cell bodies of these efferent nerves reside in the brainstem vagal dorsal motor nucleus. Those fibers exhibiting high thresholds to electrical stimulation are inhibitory to motor activity via release of VIP and NO. The efferent vagal supply is maximal in the upper gut, although anterograde tracing studies from the rat dorsal motor nucleus confirm vagal innervation to much of the colon. [49] In ferrets, cooling the vagus nerves, thereby inducing temporary vagotomy, abolishes phasic motility in the proximal colon, whereas vagal stimulation evokes proximal colonic contractions. [50] Pelvic nerve fibers arise from cell bodies in the sacral spinal cord and enter the colon at the rectosigmoid junction. From there, they travel in shunt fascicles to innervate the myenteric plexus. Unlike enteric nerves, shunt fascicles have a perineurium, myelinated fibers, and their own blood supply. Pelvic nerve stimulation evokes generalized colonic contractions of the circular and longitudinal muscle layers and accelerates colonic transit. [45], [51] Sympathetic splanchnic innervation is different from vagal parasympathetic innervation in that neuronal cell bodies reside outside the gut in the prevertebral ganglia (i.e., the celiac, and superior and inferior mesenteric ganglia). Preganglionic cholinergic neurons project from the spinal cord to the prevertebral ganglia. Noradrenergic postganglionic neurons then project to the enteric ganglia. Such sympathetic innervation generally inhibits excitatory cholinergic transmission in the myenteric plexus of both the small intestine and colon. [45], [51] The functional significance of these pathways is exemplified by the long inhibitory intestinal reflexes that decrease motility through neural arcs involving the prevertebral ganglia. Conversely, ablation of the prevertebral ganglia has little effect on colonic transit. Numerous neurotransmitters participate in extrinsic neural control of small intestinal and colonic motor function. Peptidergic fibers containing somatostatin, substance P, cholecystokinin (CCK), neuropeptide Y (NPY), enkephalins, and other transmitters are present in the vagus and splanchnic nerves.

Afferent Neural Pathways Afferent fibers outnumber efferent fibers by tenfold in the vagus and by almost threefold in the splanchnic nerves. Vagal afferent fibers terminate in the brainstem nucleus solitarius. Neurons in the nucleus solitarius project to the vagal dorsal motor nucleus and the nucleus ambiguus. Sensory information from the small intestine and colon is transmitted to the dorsal horn of the spinal cord by the splanchnic nerves. From there, second-order neurons project to the brainstem and cerebral cortex. The anus is innervated by sensory fibers that project through the pudendal nerve. The small intestine and colon are richly supplied with sensory fibers. Free mucosal nerve endings respond to stroking or to chemical stimuli such as hydrochloric acid. Mucosal receptors in the anal canal are sensitive to mechanical stimuli, temperature, and electrical current. [52] Thermoreceptor exposure in the small intestine activates perceptual responses and reflex motor responses in the stomach. [53] Mechanoreceptors are activated by passive distention or during active contractions. Pelvic nerve afferent fibers from the colon respond to distention, heat, and bile salts, indicating the polymodal nature of the receptors. [54] That excision of the mucosa, submucosa, and the inner circular muscle does not abolish responses to stretch localizes mechanoreceptors to the outer muscle layers or the myenteric plexus. [55] Mesenteric and serosal receptors respond to tension or to forceful contraction and may mediate visceral pain perception. Intrinsic afferent neurons that mediate local neural reflexes project within the enteric plexuses. Afferent pathways that mediate extended reflexes involve vagal, pelvic, and splanchnic nerves and may be modulated by cerebral input. Afferent information involving perception of nonnoxious stimulation may be transmitted via vagal, pelvic, or splanchnic pathways, whereas nociceptive input is believed to be carried mainly by the splanchnic nerves. Significant interaction between motor and sensory function is evidenced by studies using dual magnetic stimulation of the cerebral motor cortex, in which cortical transmission to the anus was facilitated by prior electrical stimulation of the lumbosacral root or pudendal nerve. [56] Plasticity of sensory nerve activity is shown by experiments that demonstrate reductions in ileal vagal afferent sensitivity with prior exposure of the distal ileum to short-chain fatty acids. [57]

Intrinsic Innervation

The density of myenteric neurons ranges from 3,700 to 12,170 cells/cm [2] in the cat small intestine, approximating that of the spinal cord. [58] In humans, an additional deep muscular plexus innervates the interface of the inner and outer circular muscle layers of the small intestine and colon and receives input from the myenteric plexus. The submucous plexus provides less than 1% of the axons to smooth muscle and projects instead to the mucosa, where it likely mediates secretory and absorptive activities. [59] In the colons of some species, the outer layer of the submucous plexus provides important input to the circular muscles and ICCs. [60], [61]

The enteric nervous system possesses the elements for complete reflex activities, including afferent neurons, interneurons, and motor neurons, and thus can mediate physiological motor patterns in the absence of extrinsic input. Myenteric ganglia resemble the central nervous system in that only neurons and glial cells are present. In the absence of blood vessels and connective tissue cells, neuronal nourishment diffuses from the interstitial fluid. Neurotransmitters released from axonal varicosities diffuse 20 to 100 nm to specific receptors on muscle cells or neurons. In the small intestine, motor neurons typically project 1 to 2 mm longitudinally, although some fibers extend for 30 mm. Excitatory fibers run cephalad, whereas inhibitory fibers project in a caudad direction. Reflex projection of 100 cm or more implies extensive interneuronal connections. [62] Most myenteric neurons project to other myenteric neurons and to the circular muscle; fewer supply the submucous ganglia. The less well supplied longitudinal muscle receives fibers primarily from excitatory motor neurons with little inhibitory input.

Eighty percent to 90% of myenteric neurons contain either tachykinins (40%–45%) or VIP (40%–45%), with no overlap between the two groups. Tachykinin neurons containing substance P, neurokinin A, and acetylcholine mediate most of the excitatory functions of intestinal smooth muscle. [63] In rat intestinal longitudinal muscle, neural stimulation evokes contractions that are incompletely blocked by atropine. [64] The atropine-resistant fraction is abolished by tachykinin receptor antagonists. The inhibitory supply to the small intestine and colon is provided by NO- and VIP-containing myenteric neurons. [65] Inhibitors of NO synthesis block ileal relaxations evoked by electrical depolarization, serotonin (5-HT), ATP, and ?-aminobutyric acid (GABA). [66] Furthermore, inhibition of NO synthase evokes duodenal contractions in rats, indicating tonic inhibition by NO pathways. [67] Similarly, antagonists or antisera to VIP prevent neurally mediated relaxation. [68], [69] A related transmitter, pituitary adenylate cyclase–activating peptide (PACAP), also may act as a physiological relaxant in some regions through activation of PACAP $_{1}$/VIP receptors. [70] NO synthase, the enzyme responsible for NO generation, colocalizes with VIP in colonic neurons, suggesting that the two transmitters perform interrelated functions. [71] In some models, VIP evokes NO-dependent relaxations through an intermediate transmitter, such as GABA acting on GABA $_{A}$ receptors. [72] Some VIP neurons contain NPY, calcitonin gene–related peptide (CGRP), gastrin-releasing peptide (GRP), and galanin. ATP is an important inhibitory neurotransmitter for some gut activities. In rat colon, nearly all ATP-containing myenteric neurons exhibit NO synthetic capabilities. [73] In rat anus, the relaxant response to ATP is reduced by a combination of an NO synthase inhibitor, tubocurarine and apamin, indicating partial NO pathway participation in purinergic innervation. [74] Finally, tachykinin pathways may interact with nitrergic function in some stimulated states, such as lumenal distention. In guinea pig small intestine, NK $_{1}$ receptor activation, mediated by NO release, evokes both stimulatory and inhibitory effects on distention-evoked contractions. [75]

Intrinsic neurons distinct from tachykinin-, NO-, and VIP-containing cells likely function as interneurons to modulate gut motor activity. Opioid neurons regulate both excitatory and inhibitory transmission. Enkephalinergic neurons project orally to both muscle layers, whereas neurons containing GRP and NPY project in an aboral direction. [76] Orphanin FQ immunoreactivity is expressed in excitatory neurons projecting to both muscle layers and to a few interneurons. [77] Neurons containing substance P and CGRP may mediate sensory function in the colon. [78] Colon smooth muscle from mice with a knock-out of the dopamine transporter exhibits impaired contractions to electrical stimulation, indicating a physiological role for endogenous dopamine. [79] Endogenous cannabinoid pathways may inhibit small intestinal motility by acting on CB $_{1}$ receptors. [80]

PHYSIOLOGICAL AND PATHOPHYSIOLOGICAL MOTOR PATTERNS UNDER BASAL FASTING CONDITIONS

Small Intestine

During fasting, the small intestine exhibits a prominent cyclic motor pattern, the migrating motor complex (MMC). Other patterns that occur less frequently and with less regularity also play important roles in small intestinal transit, in both health and illness.

Migrating Motor Complex

Physiological characteristics. The MMC is an organized fasting pattern termed the *intestinal housekeeper* that propels undigested food residue and sloughed enterocytes from the proximal gut. [81] The rapid development of small intestinal bacterial overgrowth in rats given morphine to disrupt MMC cycling shows the importance of this motor pattern (Fig. 11-3). [82] The electrical correlate of the MMC is the migrating myoelectric complex. [83] The MMC consists of four phases lasting 84 to 112 minutes (Fig. 11-4). Phase I is a period of motor quiescence lasting 40% to 60% of the cycle length. Phase II, occupying 20% to 30% of the cycle, exhibits irregular phasic contractions involving about half of the slow wave cycles. The duodenal cross-sectional area is greater during phase II than phase I, possibly to accommodate pancreaticobiliary secretions. [84] Phase III is a 5- to 10-minute period of lumenally occlusive, rhythmic contractions, most of which propagate aborally. Some duodenal phase III contractions propagate in an orad direction, however, indicative of a physiological retroperistaltic pump. [85] This retroperistaltic activity is associated with duodenogastric reflux of bicarbonate and immunoglobulin A, which have been proposed to reconstitute the antral mucosal barrier during fasting. [86] The maximal phase III contractile frequency is determined by the slow wave frequency (11–12 cpm in the duodenum, 7–8 cpm in the ileum). Phase III complexes propagate more slowly (4–6 cm/min) than the slow wave because of a loss of slow wave phase locking and shortening of frequency plateaus. [87] Nevertheless, individual phase III contractions propagate over longer distances than phase II contractions. [88] The length of intestine in a given phase III complex decreases from 40 to 60 cm in the duodenum to 5 to 10 cm in the ileum. Phase IV is a transition period of irregular contractions between phase III and phase I.

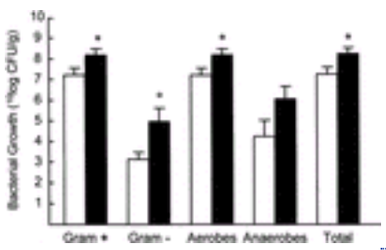

FIGURE 11-3. Growth of different bacteria in the duodenum of rats is shown after administration of placebo (*open bars*) or morphine (*black bars*) to disrupt migrating motor complex (*MMC*) cycling. Morphine led to increases in total bacteria as well as both gram-positive and gram-negative aerobes. (From ref. [82].)

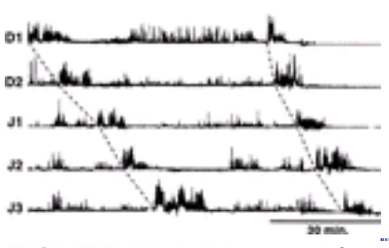

FIGURE 11-4. Manometric recordings of the different phases of the human migrating motor complex (MMC) cycle from the duodenum (D1, 2. to the jejunum (J1, 2, 3). The *dashed lines* show propagation of the leading edge of two phase III complexes. (From ref. [90].)

Although typically present during fasting in healthy persons, MMCs exhibit variability in several parameters. However, sequential manometric recordings in the same patients demonstrate that intraindividual MMC characteristics are very reproducible. [89] In nearly all healthy subjects, at least one phase III develops during 6 hours of fasting. [90] Seventy-one percent of phase III complexes originate in the stomach, with 28% beginning in the duodenum and 1% in the proximal jejunum. [91] The fraction

of MMC complexes that originate in the stomach or duodenum linearly correlates with the duration of fasting. [92] Furthermore, MMC cycle duration is nearly twice as long when the previous phase III complex originated in the stomach versus the duodenum. [93] One half of MMCs propagate beyond the midjejunum, and only 10% reach the distal ileum. Infants born at term exhibit physiological phase III activity, albeit with an increased number of nonpropagated clusters; preterm babies, however, often exhibit low-amplitude, disorganized clustered contractions of short duration without recognizable phase III activity. [94] There are no qualitative differences in MMC characteristics between young (18–39 y) and old (40–69 y) persons, although phase III propagation velocity may decrease in persons older than 80 years. [95]

Neural regulation. Neural regulation of the intestinal MMC involves extrinsic and intrinsic input. Extrinsic innervation from the vagus and splanchnic nerves serves a modulatory function. Bilateral truncal vagotomy, removal of the superior and inferior mesenteric ganglia, total sympathectomy, and complete extrinsic denervation of the small intestine do not prevent MMC cycling, although cycle duration and regularity may be altered. [96], [97] and [98] Bilateral vagotomy increases cycle duration, whereas section of the extrinsic nerves to the jejunum and ileum decreases the MMC cycle length, reduces the percentage of phase III complexes that propagate to the ileum, and disrupts coordination of duodenal and jejunal phase III activity. [96], [99] Vagal cooling in dogs has no effect on intestinal phase III, but it shortens intestinal phase II activity, an effect suggesting a vagally mediated pathway for maintenance of phase II activity and a vagally independent pathway for phase III. [100] Enteric ganglia coordinate small intestinal MMC propagation. Isolated denervated intestinal segments exhibit spontaneous phase III activity that propagates aborally; however, cycling in the excluded segment is out of phase with the main segment, suggesting that continuity of the enteric nervous system synchronizes cycling from one intestinal region to the next. [101] Likewise, intestinal transection into many segments with subsequent end-to-end reanastomosis promotes independent phase III activity in each segment. [102] In this model, neuronal growth across the anastomosis is noted at 28 days, and coordinated cycling resumes at 45 days, features indicative of regeneration of enteric nerve connections. [102], [103] If a segment of colon is interposed between the small intestinal segments or if end-to-side or side-to-side intestinal anastomoses are performed, coordination does not resume; this shows a requirement for continuity of small intestinal–type enteric nerves. [104] In guinea pigs, longitudinal myomectomy with circumferential interruption of myenteric ganglia blocks propagation of 50% to 60% of phase III complexes, whereas complete transection with reanastomosis blocks more than 80% of complexes, indicating an important role for the myenteric plexus in MMC continuity. [103] Other investigators have demonstrated disruption but not abolition of MMC cycling with serosal application of benzalkonium hydrochloride, which selectively destroys myenteric ganglia, suggesting that the deep muscular plexus and submucous plexus have some role in maintaining the MMC. [105] Cholinergic and noncholinergic pathways participate in MMC contractions. In dogs, intravenous administration of atropine, the ganglionic blocker hexamethonium and the neural toxin tetrodotoxin eliminate MMC cycling. [106] Close intraarterial injection of these agents prevents MMC propagation distal to the site of infusion. [107] Adrenergic receptor antagonists disrupt but do not abolish MMC cycling. NO synthase inhibitors evoke premature MMC activity, then more rapid cycling, indicating that endogenous NO may be a physiological inhibitor of fasting motor function. [108], [109] Conversely, the NO donor sodium nitroprusside disrupts MMC activity in rats and induces a postprandial-like pattern. [110] Similarly, the phosphodiesterase inhibitor sildenafil, which prevents cyclic GMP degradation after NO stimulation, disrupts MMC cycling. [111] Selective VIP receptor antagonists block the disruptive effects of NO donors, indicating that NO may mediate its actions through VIP release. [112] After small intestinal transection and reanastomosis in guinea pigs, immunoreactivities for VIP, GRP, and somatostatin are decreased distal to the anastomosis. [103] Recovery of MMC cycling is associated with recovery of these peptides and nerve regrowth, suggesting that peptidergic nerves within the myenteric plexus may contribute to coordinated MMC activity. Endogenous 5-HT has been promoted as a physiological modulator of the intestinal MMC. 5-HT increases the frequency of intestinal contractions, shortens the MMC cycle length, accelerates propagation patterns, and converts mixing patterns to propulsive ones. [113], [114] In rats, ablation of myenteric 5-HT neurons with 5,6- and 5,7-dihydroxytryptamine prolongs MMC periodicity and decreases propagation velocities. [115] Finally, 5-HT $_3$ receptor antagonists inhibit phase III activity or prolong cycle length. [116], [117] Other neural factors may modulate intestinal MMC activity. Morphine induces premature intestinal phase III activity, whereas the μ-opioid receptor antagonist naloxone prolongs MMC periodicity from 103 to 219 minutes. [102], [118] Consequently, opioid peptides may be intermediaries in generating intestinal phase III activity and may function in recovery of phase III cycling after a meal. Neurokinin A and substance P increase phase II activity in the human small intestine, suggesting that tachykinins may selectively regulate that phase of the MMC. [119]

Hormonal mediation. Induction of phase III in the stomach and proximal small intestine results from release of the hormone motilin from the duodenal mucosa. [120] Motilin receptors are expressed in human duodenal and colonic enteric neurons. [121] The motilin receptor is G-protein coupled and is 52% identical to the receptor for growth hormone secretagogues. Motilin also is localized to the hippocampus, thalamus, hypothalamus, amygdala, cerebellum, and vagus in animal models; however, its physiological function in these sites is uncertain. [122], [123], [124] and [125] Antral phase III complexes temporally correlate with plasma motilin elevations in healthy humans, and premature antral phase III activity is inducible by motilin infusion. [126] Motilin-evoked phase III is identical in duration, amplitude, and propagation velocity to spontaneous complexes. [127] In dogs, gastroduodenal phase III, abolished for several hours after infusion of motilin antisera, is replaced by irregular phasic contractions. [128] Likewise, duodenal excision with removal of motilin-secreting tissue alters fasting antroduodenal motility in dogs. [129] The motilin-dependence of MMC cycling exhibits neural plasticity. After duodenectomy, dogs exhibit a recovery of gastroduodenal MMC-like activity 1 to 4 months postoperatively that is vagally mediated and dependent on cholinergic and adrenergic efferent pathways. [130] The physiological stimulus for motilin cycling is not known. Cyclic motilin fluctuations are blocked by atropine and hexamethonium, suggesting regulation by cholinergic pathways. [131] Motilin release is evoked by vagal stimulation, cholinergic agonists, opioid agents, and duodenal pH changes, but the roles of these stimuli in the regulation of motilin release are unclear. [131], [132] and [133] Similarly, NO synthase inhibitors evoke phase III–like antroduodenal complexes with associated increases in plasma motilin. [134] There are differences in the motilin-dependence of phase III complexes in the proximal and distal small intestine. Ectopic complexes originating distal to the ligament of Treitz often are unassociated with plasma motilin elevations. [135] Furthermore, the effects of motilin antisera on distal jejunal and ileal phase III complexes are minimal. [102], [128] Similarly, ectopic phase III complexes form in the jejunum and ileum after resection of duodenal motilin-producing tissues. [129] These findings suggest that “programming” of phase III activity in the medial and distal intestine is a motilin-independent phenomenon that is entrained by the actions of motilin on the antrum and duodenum. The only peptides other than motilin known to cycle in phase with the MMC are somatostatin and pancreatic polypeptide; however, it is unlikely that either is a mediator of the complex. [136], [137] Somatostatin evokes intestinal phase III complexes every 20 to 30 inutes, but the complexes are not physiological because gastric motor activity is suppressed. [136] Furthermore, somatostatin-evoked complexes do not exhibit a phase II–like pattern of irregular contractions, and they delay rather than accelerate transit. It is conceivable that somatostatin may play a role in motilin-independent cycling in the distal intestine. Plasma levels of pancreatic polypeptide, a peptide produced by the pancreas, also peak just before phase III. [138] Furthermore, pancreatic polypeptide is released by exogenous motilin administration through vagal, cholinergic pathways involving 5-HT $_3$ receptors. [139] However, pancreatic autotransplantation has no effect on intestinal phase III cycling despite its disruption of pancreatic polypeptide fluctuations. [140] In addition, pancreatic polypeptide infusion to levels mimicking those during phase III has no effect on fasting intestinal motility. [141] The importance of other hormones in regulating MMC activity is speculative. Glucagon-like peptide 1 (GLP-1) prolongs MMC cycle duration with slowing of transit through activation of NO-dependent pathways. [142] Ileal resections reduce phase I duration, shorten the MMC cycle, and induce intestinal clusters, suggesting modulatory roles for peptides specifically released by the distal small intestine. [143]

Other associated cyclic phenomena. The intestinal MMC cycles in phase with motor activity of the gallbladder and sphincter of Oddi as well as with several secretory functions. Gastric acid and pepsin production and intestinal fluid secretion increase before duodenal phase III. [144] Similarly, the release of bicarbonate, bile acids, bilirubin, pancreatic enzymes, and lumenal secretory immunoglobulin A peaks just before or during phase III. [138], [145] Qualitatively, this relationship persists at night; however, there is a relative reduction in motor activity with preservation of exocrine output providing evidence for differential circadian rhythms for motility and enzyme release (Fig. 11-5). [146] Furthermore, duodenectomy and diseases such as chronic pancreatitis disrupt the synchrony of MMC activity and pancreatic exocrine cycling, suggesting differential regulation of the two phenomena. [129], [147]

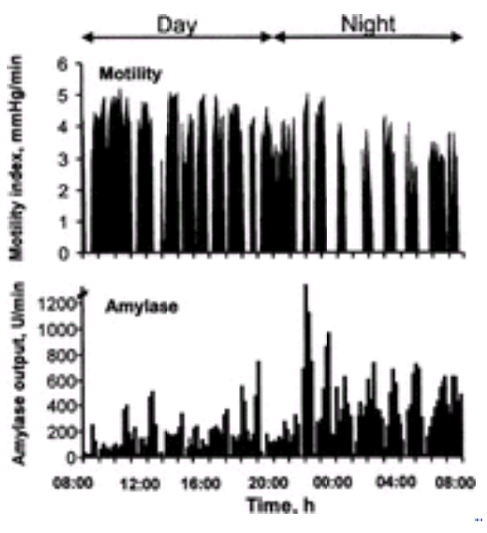

FIGURE 11-5. Circadian cycling of jejunal motility (**top**) and amylase output (**bottom**). Pancreatic secretion exhibits a close correlation with motor cycling, however differential regulation is shown by the observation that enzyme output increases at night while contractile activity is unchanged. (From ref. [146].)

These secretory activities may modulate the induction of MMC cycling. Motilin is released into the circulation after exposure of duodenal mucosa to bile. [148] Furthermore, experimental bile duct occlusion or diversion of bile away from the duodenum disrupts motilin cycling with the loss of duodenal phase III and induction of ectopic jejunoileal complexes. [149] Bile flow restoration and exogenous bile acid perfusion reverse these effects. [150] Despite this information, other studies dispute the role of bile in MMC generation. [151] Peak pancreatic secretion has been observed both in phase and out of phase with phase III cycling. [152] Phase III–independent secretory peaks occurred in subjects with longer MMC cycle lengths.

Giant Migrating Complexes Intense contractile waves that propagate aborally for long distances in the intestine are observed in experimental animals during hypoxia, anemia, and gangrene, after laparotomy, and after death. This phenomenon has been given many names including peristaltic rush, prolonged propagated contractions, migrating action potential complexes, power contractions, and giant migrating contractions (GMCs). Small intestinal GMCs are two to three times greater in amplitude and four to five times longer in duration than individual phasic contractions, and they propagate at 1 cm per second. [153], [154] The duration of an intestinal GMC generally is longer than that of a single slow wave, indicating dissociation of the slow wave regulation of phasic contractile activity. [153] During a GMC, myoelectric measurements record intense bursts of spike potentials lasting 4 to 16 seconds that obscure the slow wave. [154] GMCs typically begin in the jejunum or ileum during fasting and migrate to the ileocecal junction. They are intensely propulsive of ileal contents, in contrast to phase III. [153] Nearly half of the GMCs are associated with propagating sequences in the cecum, indicating a coordinated evacuation mechanism in the distal gut (Fig. 11-6). [155] Consequently, intestinal GMCs are postulated to clear debris from the ileum and to prevent coloileal reflux. Intestinal GMCs, rare in health (0.03/h), are induced by noxious stimuli such as intravenous morphine, intragastric vinegar, ileal perfusion of feces or short-chain fatty acids, ionizing radiation, and infection with *Vibrio cholerae, Clostridium perfringens, Clostridium difficile,* noninvasive *Escherichia coli, Shigella* organisms, and *Trichinella spiralis*. [154] Induction of GMCs is associated with decreases in ileal pH, suggesting that coloileal reflux may be a physiological stimulant of this motor pattern. [156] Diarrhea, fecal urgency, and abdominal discomfort temporally correlate with GMCs during ileal inflammation, a correlation suggesting a role in symptom induction. In enteropathogenic *E coli* infection, GMCs present before the onset of diarrhea, confirming that they are not merely a response to the diarrheal state. [157] Propagation of small intestinal GMC activity is controlled by enteric neural pathways, whereas extrinsic nerves regulate inhibition of motor activity orad to the complex. [158] Research suggests that neurokinin (NK_3) receptors on presynaptic neurons may mediate GMCs in the small intestine. [159]

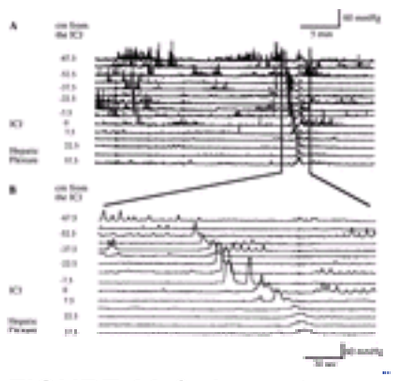

FIGURE 11-6. A spontaneous giant migrating contraction (GMC) originating in the ileum is shown propagating across the ileocecal junction (ICJ) into the proximal colon. (From ref. [155].)

Other Aborally Migrating Patterns Intensely propulsive contractions also occur in clusters termed migrating clustered contractions, discrete clustered contractions (DCCs), or the minute rhythm. [153], [160] DCCs occur while fasting and after eating and consist of 3 to 10 contractions preceded and followed by 1 minute of motor quiescence (Fig. 11-7). DCCs migrate at 5 to 10 cm per minute over distances of 2 to 40 cm and are proposed, along with GMCs, to be a physiological means of emptying the ileum. [153], [160] Unlike GMCs, DCCs rarely extend into the proximal colon. [155] In some studies, increased DCCs are observed in irritable bowel syndrome, intestinal pseudoobstruction, and partial small bowel obstruction. [161]

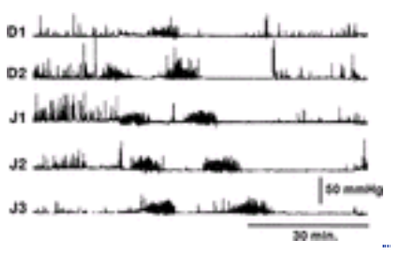

FIGURE 11-7. Discrete clustered contractions (DCCs) occurring in human duodenum (D1, 2) and jejunum (J1, 2, 3). (From ref. [90].)

An extremely propagative pattern, called the *rapidly migrating contraction,* migrates 200 cm at more than 30 cm per second. [162] In contrast to intestinal GMCs, rapidly migrating contractions occur predominantly in the proximal small intestine and are associated with disruption or loss of slow wave activity.

Retrograde Peristaltic Contractions After administration of emetic agents, complexes migrate orally from the medial small intestine to the duodenum before vomiting. These retrograde peristaltic contractions (RPCs) exhibit periods of motor inhibition immediately before and after the complex that are followed by several phasic contractions and then a second inhibitory period. [163] Vagotomy abolishes the entire phenomenon, whereas atropine prevents only the RPC itself. Although most often associated with retching or vomiting, RPCs do occur in their absence. Conversely, retching and vomiting can occur in the absence of RPCs. The purported function of RPCs is to evacuate intestinal contents into the stomach so that they may be expelled during emesis. RPCs exhibit contractile amplitudes 1.3 to 1.8 times greater than normal intestinal contractions with durations 2 to 4 times longer and rapid migrations (8–10 cm/s) over distances greater than 100 cm. [164] RPCs are preceded by slow wave obliteration before generation of electrical bursts that migrate orally. [165] Investigations in dogs involving autotransplantation of small intestinal segments in dogs indicate that retrograde motor activity is controlled by extrinsic neural pathways. [166]

Colon

The movement of colonic contents in humans is regulated by the concerted actions of several distinct motor patterns and their corresponding myoelectric complexes. Additional organized motor complexes are described in some species.

Short and Long Duration Contractions SDCs are stationary pressure waves that mix fecal material to effect water extraction. The myoelectric phenomena underlying SDCs are short spike bursts that occur in phase with colonic slow wave activity. SDCs persist for a mean of 8 seconds, occur at frequencies of 4 to 6 cpm in dogs and 2 to 13 cpm in humans, and are recorded from in vitro circular muscle preparations, suggesting that this is their site of generation. [167], [168] Long duration contractions (LDCs) may be stationary, or they may propagate for short distances in either direction, thus promoting both mixing and local propulsion of feces. In the ascending and transverse colon, many LDCs migrate orally, whereas in the more distal colon, LDCs typically migrate in an aboral direction. [169] The myoelectric correlates of LDCs, long spike bursts, are action potentials generated by myenteric potential oscillations. In contrast to short spike bursts, long spike bursts do not occur in synchrony with colonic slow waves. Although long spike bursts oscillate at 25 to 40 cpm, a single, tetanic LDC usually persists for the length of the electrical complex because of limitations on the rates of relaxation of colonic smooth muscle. LDCs persist for 20 to 60 seconds and are recorded from in vitro longitudinal muscle preparations, suggesting the presence of myoelectric control from a different site than for SDCs. [167], [168], [170] Compared with SDCs, LDCs are of greater amplitude. [171] In general, most LDCs occur during the day, with increased frequency after eating and after awakening. [172]

Giant Migrating Contractions Colonic GMCs, also termed *high-amplitude propagated contractions,* propagate aborally over extended distances and evoke mass movements of feces. [173] The myoelectric correlates of colonic GMCs are migrating, long spike bursts, which are believed to be generated from myenteric potential oscillations. The importance of this motor pattern in fecal expulsion is shown by studies in dogs in which prominent colonic GMCs precede the need to defecate. [173] In ambulatory manometric studies of unprepared colons in healthy humans, propagating pressure waves of high amplitude (>105 mm Hg) and prolonged duration (>14 s) were observed to occur about 10 times daily, most prominently after awakening, after eating, or in association with defecation (Fig. 11-8). [174] Such patterns may

originate as early as 1 hour before stool expulsion. [175] In general, the time interval to defecation correlates negatively with the amplitude of the contractile complex. Most sequences in humans originate in the cecum. [176] Ninety percent of GMCs associated with defecation in dogs are accompanied by propagation to the rectum and relaxation of the internal anal sphincter. [177] In human scintigraphic studies, oleic acid infusion into the cecum produces rapid tracer shifts from the ascending colon to the splenic flexure and descending colon that temporally correlate with the induction of colonic GMCs. [178] Colonic GMCs are evoked by distention and are increased during experimental diarrhea caused by laxatives, abdominal radiation, intracolonic acetic acid, and intravenous cisapride and may thus be partially responsible for the increased defecation frequency and impaired fluid absorption that occur with diarrhea. [179] Orphanin FQ accelerates colonic transit in rats by inducing GMCs. [180] Experimental colonic inflammation enhances the stimulation of colonic GMCs by substance P, suggesting the involvement of neuropeptide mechanisms in producing inflammatory diarrhea. [181] Colonic GMCs evoked by intracolonic glycerol are reduced by lidocaine, hexamethonium, and atropine, indicating mediation by mucosally activated cholinergic pathways. [182] Investigations indicate that colonic GMCs may be mediated by activation of NK_1 receptors on smooth muscle cells. [159]

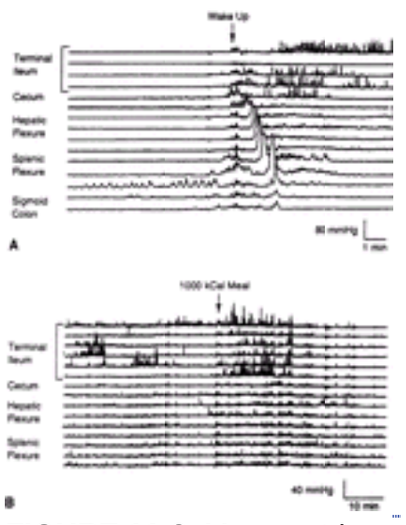

FIGURE 11-8. Manometric recordings of human colonic contractions after awakening and meal ingestion. During sleep, colonic motor activity is minimal. After awakening, colonic contractions increase, and a colonic giant migrating contraction (*GMC*) is observed to propagate from the cecum to the sigmoid colon (**A**). Ingestion of a 1,000-kcal meal evokes increased contractile activity at all levels of the colon (**B**). (From ref. [176] .)

Rectal Motor Complexes Specialized motor patterns may facilitate the storage function of the rectum. Ambulatory manometric studies demonstrate rectal motor complexes that occur about 16 times per day with average durations of 5 to 15 minutes (Fig. 11-9). [183] Motor complexes occur more commonly in the distal rectum compared with the proximal rectum. Contractile complexes consist of two to three contractions per minute, with a mean amplitude of 58 mm Hg. Such rectal motor activity occurs during the day and nocturnally and is not related to meal ingestion; however, most rectal events temporally occur in relation to contractions more proximally in the colon. [184] For the most part, rectal motor complexes do not propagate either in an orad or aboral direction, although some studies support their association with motor events more proximally in the colon. [176] Anal relaxations are not observed during rectal motor complexes, consistent with a role in maintenance of fecal continence. [185]

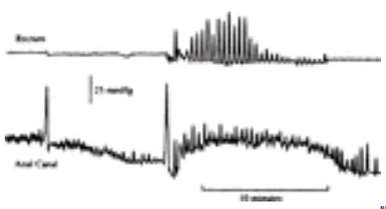

FIGURE 11-9. A rectal motor complex and the associated anal manometric recording. This rectal complex exhibits a duration of about 10 minutes and is associated with an increase in anal tone. (From ref. [185] .)

Migrating Motor Complexes Organized patterns known as MMCs are observed in some species but not in humans. The myoelectric correlates, migrating myoelectric complexes, cycle every 40 to 50 minutes in dogs. Colonic MMCs propagate over more than half the length of the colon with a duration of 30 to 120 seconds. [186] There is no coordination of colonic MMC activity with the small intestinal MMC.

PHYSIOLOGICAL AND PATHOPHYSIOLOGICAL MODULATORS OF SMALL INTESTINAL AND COLONIC MOTILITY

Motor patterns of the small intestine and colon under fasting conditions can be modified by external stimulation. Additional intralumenal stimuli also may modulate lower gut function.

External Stimuli

Responses to Meal Ingestion

Small intestinal fed motor pattern

Physiological characteristics. After a meal, the MMC is replaced by a fed pattern of intermittent phasic contractions of varying amplitude, occurring throughout the small intestine and peaking 10 to 20 minutes after eating. [187] The number of isolated pressure waves during the fed period is greater than during a comparable phase II period. [188] The myoelectric correlate consists of random bursts of spike potentials in phase with the slow wave. [189] The fed pattern serves to mix and propel intestinal contents. Forty-four percent of fed contractions do not propagate. Of contractions that do, 90% migrate less than 30 cm and 66% migrate less than 9 cm. In the proximal duodenum, 40% to 50% of fed contractions are retrograde. [190] This duodenogastric propulsion may modulate gastric emptying. However, most pressure waves propagate in an antegrade direction for distances of 1.5 to 4.5 cm in the distal duodenum during duodenal lipid perfusion. [191] The small intestinal fed pattern, which persists throughout life, has been recorded in preterm infants. [192] Other motor patterns are seen after meal ingestion. A transitional motor pattern observed immediately after meal ingestion is characterized by highly propagative contractile clusters that expose extended regions of mucosa to nutrients. [193] Intense individual migrating contractions occurring postprandially have amplitudes twice those of normal fed phasic contractions and durations equal to two slow wave cycles. [88] Postprandial ileal contractions, once thought to be weaker than those in the duodenum, have been shown in dogs to have increased contractile amplitudes. [194] Intense pressure waves at frequencies of 19 to 24 cpm have been observed in the terminal ileum and appear to develop 1 to 4 hours after eating when chyme reaches the colon. [195] Myoelectric recordings from dogs show that this activity is accompanied by spike bursts similar to myenteric potential oscillations in the colon. Finally, a delayed tonic relaxation of the ileum is induced by local exposure to bile acids and triglycerides not absorbed in the proximal intestine. [196] The duration of the fed motor pattern is dependent on the caloric content and qualitative aspects of the meal. [197] In dogs, a mixed 450-kcal meal induces a fed pattern lasting longer than 3 hours. The threshold for inducing the fed state in humans is not known, although a 345-kcal meal disrupts MMC cycling for more than 90 minutes. [198] Peanut oil, consisting mostly of 18-carbon triglycerides, induces fed contractions for longer periods (>8 hours) than equicaloric sucrose or milk protein meals. [199] In humans, a 400-kcal meal with 9% fat disrupts MMC activity for 294±21 minutes, whereas a meal with 50% fat prolongs the fed period to 410±42 minutes. [200] Long-chain triglycerides convert the MMC to a postprandial pattern, whereas medium-chain triglycerides in equicaloric amounts have no effect. [201] The addition of guar to a glucose drink prolongs the duration of the fed pattern. [202] Intravenous amino acids shorten the cycle length of the MMC but do not induce a fed motor pattern, indicating that participation of the gut lumen is requisite. [203]

Neural regulation. Extrinsic innervation of the small intestine plays an important role in inducing the fed pattern. In dogs, the sight or smell of food disrupts MMC cycling, suggesting that cephalic phase initiates the fed pattern. In humans, sham feeding disrupts duodenal phase III but does not evoke fed contractions. [204] Bilateral vagotomy, splanchnicectomy, mesenteric ganglionectomy, and total extrinsic denervation do not prevent induction of the fed state, but bilateral vagotomy shortens its duration and increases the latency from the time of eating to the onset of fed contractions. [96], [101] Complete denervation reduces the number of propagated fed contractions, the mean distance of propagation, and the rate of intestinal transit. Jejunoileal autotransplantation, which produces extrinsic denervation, increases the number of calories needed to abolish MMC cycling and to initiate the fed pattern. [205] Finally, vagal cooling during the early postprandial period converts the fed pattern

to intermittent phase III activity. [100] These findings indicate modulation of the fed state by extrinsic neural pathways. Intrinsic innervation also modulates propulsion during the fed period. Intestinal transection and reanastomosis in dogs decrease the frequency and amplitude of fed contractions with decreased propagation. [206] In the absence of continuity of the enteric nervous system, the extrinsic nerves alone are sufficient to suppress fasting motility in excluded intestinal regions. If nutrients are perfused into an isolated, but extrinsically innervated intestinal loop, the MMC is disrupted in the unconnected main portion of the intestine. [207] As with the MMC, atropine and hexamethonium abolish fed motor and spike potential activity, emphasizing the importance of cholinergic innervation. [208] Infusion of an NO synthase inhibitor shortens the duration of the fed pattern, suggesting that endogenous NO pathways are physiological regulators of postprandial function. [108]

Hormonal mediation. Despite much investigation, the role of CCK as a mediator of the fed state remains unproven. CCK levels increase five- to tenfold after eating but do not persist for the duration of the fed state. CCK and its analogs increase intestinal motility, although the motor pattern exhibits nonphysiological characteristics such as prominent retrograde contractions and a preferential stimulation of proximal intestine contractions. [209] CCK inhibits MMC activity but does not prevent motilin cycling. [209] In rats, duodenal perfusion of trypsin inhibitor releases endogenous CCK, which acts on vagal afferent CCK-B receptors to stimulate a central CCK-A receptor pathway to disrupt MMC cycling. [210] Intravenous CCK-B receptor blockade and central administration of a CCK-A receptor antagonist prevent the disruption of MMC activity in response to nutrients in rats. [211] In dogs, the CCK receptor antagonists loxiglumide and MK-329 reduce but do not prevent the fed response or the interruption of MMC cycling after eating. [212] Thus, the role of CCK in mediating the fed response may be species dependent. Other compounds inhibit the MMC and induce complexes similar to the fed pattern, including gastrin, insulin, glucagon, neurotensin, neuromedin-N, enkephalins, and prostaglandin E_2. [96] As with CCK, gastrin does not reproduce the fed pattern, but it does stimulate proximal intestinal motility. Gastrin release also does not persist for the duration of the fed period. [209] Peanut oil, which induces a fed pattern, does not evoke gastrin or insulin release. [213] Conversely, glucose increases insulin levels but does not induce a fed pattern. Indomethacin infusion induces a fed pattern in dogs, but prostaglandin E_2 also disrupts MMC cycling. [214] Neurotensin has been proposed as a mediator of fed motility based on its ability to convert the fasting to a fed pattern along the entire small intestine in rats and humans. [215] In rats, neurotensin antagonists also reduce the duration of the fed pattern. [216] Although motilin is important in initiating fasting activity, it plays no role in the fed state. After eating, motilin levels decrease drastically. If exogenous motilin is given during the fed state, no phase III activity develops. [127] In addition, motilin antisera generate motor patterns similar to those seen in the fed state, suggesting that suppression of motilin release may be necessary for induction of the fed state. [128]

Reversion to the fasting motor pattern. The mechanism for reversion to MMC cycling after completion of the fed pattern is poorly understood. The first MMC after eating begins distal to the duodenum, implying that factors required to initiate normal complexes are not yet operational. [199], [217] There is a strong correlation between initiation of duodenal phase III and completion of gastric emptying. [218] However, in dogs, continuous intraduodenal perfusion of nutrients induces the fed motor pattern for only a finite time, after which the fasting pattern returns, indicating that the presence of intestinal nutrients does not prevent resumption of fasting motor activity. In humans, continuous duodenal feedings produce persistence of the fed pattern for at least 6 hours, whereas phase III activity resumes 4 hours after initiation of intragastric feedings, indicating differential inhibitory effects of the two regions on MMC cycling. [219]

Gastrocolonic response. As in the small intestine, eating produces simultaneous increases in motor activity throughout the colon (see Fig. 11-8). [220] The sigmoid colon exhibits a greater phasic response than the transverse colon. [221] The effect of eating on regional tone is less certain; some studies report greater responses in the transverse colon, and others report larger increases more distally. [221], [222] Myoelectric recordings in humans show increases in propagative and stationary long spike bursts that peak 15 minutes after eating. In some instances, mass fecal movements with defecation may occur during the gastrocolonic response. Prolonged manometric studies in dogs show disruption of the colonic MMC for 582 minutes after eating. [223] Although the fat content of a meal was thought to relate to the magnitude of the gastrocolonic response, studies show no difference in the response to low- and high-fat meals. [224] Mediators of the gastrocolonic response are incompletely defined. The response is abolished by the anticholinergic agent clidinium, indicating mediation by cholinergic pathways. [225] In humans, the 5-HT_3 receptor antagonist ondansetron blunts the tonic response to eating. [226] The human gastrocolonic response consists of a mechanoreceptor component activated by gastric distention and a chemoreceptor component stimulated by intestinal nutrients. [227] Both the mechanoreceptor and chemoreceptor components are partially mediated by 5-HT_3 pathways (Fig. 11-10). [228] Neurotensin receptor antagonists suppress the early fed response in the rat distal colon and block the late response in the proximal colon. [216] In one investigation, spinal cord section did not abolish the gastrocolonic response, but a second study in dogs showed that paraaortic and presacral denervation reduced the colonic response to intragastric lipids, suggesting modulation by the pelvic plexus. [229] Postprandially released transmitters such as CCK and gastrin evoke increases in colonic motor and myoelectric activity in some models; however, a human study showed no increase in colonic motor activity or transit during CCK administration at doses that stimulate pancreatic enzyme secretion and gallbladder contraction. [230] Furthermore, a study in dogs reported that CCK increased phasic colon contractions but decreased tone, whereas meals provoked increases in both. [231] However, also in dogs, the CCK receptor antagonist loxiglumide was shown to blunt the increase in colonic motor activity in response to eating, suggesting possible species-dependent effects. [232] Similar studies in rats using selective antagonists have shown potential roles for both CCK-A and CCK-B receptor mediation of the gastrocolonic response.

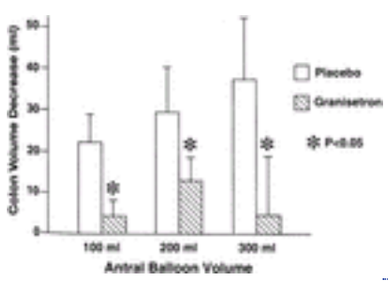

FIGURE 11-10. As a model of the mechanoreceptor component of the gastrocolonic response, inflation of an antral balloon leads to decreases in volume of a colonic recording balloon indicative of increased tone. This tonic contraction is blunted by granisetron, showing mediation by 5-HT_3 pathways. (From ref. [228].)

Motility during Sleep Small intestinal motor patterns exhibit subtle differences during wakefulness and sleep. MMC complexes migrate 2.5 times faster during the day but exhibit higher contractile amplitudes at night. [198] MMC periodicity and phase II duration are shorter during sleep, whereas phase I duration is prolonged. [233] The summed motility index during wakefulness exceeds that during sleep. Prolonged manometric recordings show no correlation of the sleep stage with MMC cycling and no synchrony of rapid eye movement sleep with intestinal phase III. [234] In contrast to fasting activity, fed patterns in humans who fall asleep soon after eating are not different from complexes recorded while people are awake. [235] Colonic motility also exhibits qualitative changes during sleep. Propagating contractions are infrequent during sleep but increase after awakening (see Fig. 11-8). [220] Awakening evokes a threefold increase in motility, including high-amplitude events in isolated segments as well as propagated complexes across long stretches of colon. [174], [236] Nocturnal suppression of colonic motility correlates with the depth of sleep and with the elimination of all propagated contractions during slow wave sleep. Conversely, during rapid eye movement sleep, colonic contraction frequencies increase to levels observed during stage 2 sleep. [237]

Central Nervous System Modulation The central nervous system has pronounced modulatory effects on small intestinal and colonic function. Intestinal phase III is reduced by depression and by acoustic and mental stress (mental arithmetic, video games, driving in rush hour traffic). [238] Phase II and DCCs also are suppressed by stress, whereas acoustic stress prolongs the fed pattern in dogs. [239] MMC disruption depends on the nature of the stressor and the time of day it is administered. Abrupt nighttime awakening does not modify the MMC, despite the subjects' perception that the nocturnal stressor is more noxious than daytime stressors. [238] Stress-induced effects on intestinal motility usually are associated with delayed transit. The colon also is susceptible to stress. Studies using stressful interviews note increases in rectal motor activity and erythema when unpleasant topics are broached. [240] In rats, conditioned fear increases colonic spike bursts. Different forms of stress elicit distinct effects on colonic motility; psychological stress induces propagated contractions, and physical stress evokes simultaneous contractions (Fig. 11-11). [241] Different regions may respond differently to stress. Tail shock in rats suppresses proximal colonic motility by reducing LDCs but increases fecal output from the distal colon. [242] A complex neurohumoral response is associated with stress. Prolongation of the intestinal fed pattern with acoustic stress is associated with gastrin, pancreatic polypeptide, and somatostatin release. [239] The delay in intestinal transit evoked by cold water immersion of the forearm is attenuated by ß-adrenergic receptor blockade. [243]

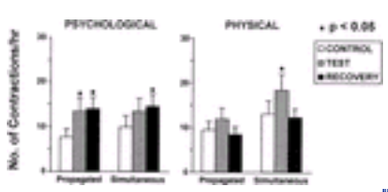

FIGURE 11-11. Effects of different stresses on colonic motor function. Psychological stress evokes predominantly propagated contractions during the stressor, whereas physical stress stimulates simultaneous contractions. (From ref. [241].)

Several compounds administered centrally can modify intestinal and colonic motor patterns. Inhibition of NO synthase in the brain suppresses duodenal phase III in dogs through vagal pathways. [244] Intracerebroventricular (ICV) infusion of NO synthase inhibitors suppresses colonic activity, whereas peripheral administration increases colonic contractions. [245] Destruction of hypothalamic and locus ceruleus adrenergic pathways with 6-hydroxydopamine lengthens MMC periodicity. [246] ICV CCK in-creases and ICV somatostatin decreases MMC cycle duration. [238] ICV calcitonin, CGRP, neurotensin, and opioids evoke intestinal phase III activity; NPY delays intestinal transit. [247] ICV neurotensin delays MMC disruption by a meal, whereas ICV atropine or substance P shortens the fed state. [248] ICV galanin restores MMC cycling during the fed period through opioid pathways. [249] Injection of a GABA antagonist into the dorsomedial hypothalamic nucleus increases jejunal contractions in rats, suggesting physiological regulation by central GABA pathways. [250] ICV GLP-1 increases fecal output, whereas orphanin FQ retards colonic transit. [251], [252] Infusion of CCK into the paraventricular hypothalamic nucleus slows colonic transit by action on CCK-B receptors. [253] ICV thyrotropin-releasing hormone accelerates intestinal transit in rats through cholinergic and serotonergic pathways and evokes colonic GMCs. [254] ICV injections of a $_2$-adrenergic receptor agonists, morphine, and leu-enkephalin blunt the increase in external anal sphincter activity elicited by pudendal nerve stimulation. [255] A large body of evidence indicates that corticotropin-releasing factor (CRF) is a physiological mediator of stress effects on the lower gut. Many stressors are associated with intracerebral CRF release. In mice, ICV CRF mimics the stressful effects of cold on intestinal transit. [256] In rats, vagotomy and naloxone prevent ICV CRF inhibition of intestinal transit, suggesting the importance of vagal and opioid pathways. [257] MMC inhibition by ICV CRF in dogs is associated with suppression of motilin cycling. [258] ICV CRF and stress increase colonic activity in rats by action on the locus ceruleus and the hypothalamic paraventricular nucleus. [259], [260] CRF-related peptides, including urocortin and sauvagine, also stimulate colonic transit. [261] The effects of CRF and stress on intestinal function are reversed by CRF antagonists, supporting a physiological role for this peptide. [262] CRF acts through activation of CRF $_1$ receptors, whereas urocortin acts on receptors of the CRF $_2$ subtype. [261], [263] The ability of CRF antagonists to block the increase in colonic contractions evoked by interleukin-1 $_\beta$ indicates a role for central CRF in pathophysiological inflammatory stress. [264] The effect of a CRF receptor antagonist to block the increase in colonic transit elicited by NPY infusion into the paraventricular nucleus indicates that hypothalamic pathways projecting to the brainstem regions direct gut stress responses. [265] Vasopressin may be an intermediate transmitter, because vasopressin antagonists block the effects of emotional stress and ICV CRF on colonic motility in rats. [266] CRF-stimulated defecation in rats may be mediated by serotonin release with action on 5-HT $_3$ receptors. [267]

Immune Modulation Inflammatory conditions may be associated with altered intestinal and colonic motility. Oral albumin evokes diarrhea in rats sensitized by intraperitoneal egg albumin injection, (i.e., antigen sensitized). [268] This intestinal anaphylaxis is associated with MMC disruption and induction of propagating, high-amplitude, clustered contractions, which are prevented by the 5-HT antagonists methysergide and cinanserin and the prostaglandin synthesis inhibitor indomethacin. [269] Similarly, colonic antigen challenge to egg albumin–sensitized rats increases colonic myoelectric activity. [270] The importance of afferent pathways in mediating these responses is shown by the ability of capsaicin to prevent antigen effects in the small intestine. [271] The role of mucosal mast cells is demonstrated by the abilities of the mast cell stabilizers and degranulation inhibitors (doxantrazole, cromoglycate, eucalyptus bioflavonoids [Quercetin]) to blunt antigen-induced intestinal and colonic motor responses and diarrhea. [270] Furthermore, mast cell degranulation evoked by BrX-537A inhibits antigen-evoked colonic myoelectric activity in rats through vagal afferent fibers and 5-HT $_3$– and substance P–dependent pathways. [272] However, other studies show participation of nonvagal pathways as well. Histologically, granulated mast cells are reduced at sites of antigen challenge. [273] In rat colon longitudinal muscle, antigen-induced contraction results from IgE-mediated mast cell activation, which then evokes release of contractile stimulants. [274] The effects of intestinal anaphylaxis on intestinal function in antigen-sensitized animals may involve central nervous system activation. In sensitized rats, egg albumin MMC disruption is associated with increased c-*fos* expression in the nucleus tractus solitarius and lateral parabrachial and paraventricular hypothalamic nuclei through vagal afferent pathways. [275] Intestinal infections also disrupt gut motor function. In rats, *Yersinia enterocolitica* and *E coli* alter MMC cycling through free radical generation. [276] Similarly, fasting intestinal myoelectric activity is increased with induction of spike potential bursts in rats infested by *Hymenolepsis diminuta*. [277] Gastrointestinal transit is slowed and ileal smooth muscle is thickened in mice infected with *Schistosoma mansoni*. [278] Infection with *Nippostrongylus brasiliensis* enhances jejunal motor responsiveness to carbachol and neurokinin A, an effect prevented by mast cell degranulation with BrX-537A. [279] Increased responses to CCK after infection with this organism are blocked by vagotomy. [280] Information on the role of the immune system in gut function has been provided by study of the nematode *T spiralis,* which produces trichinosis. Infection with *T spiralis* reduces jejunal myenteric neuron membrane potential and increases neuronal metabolic activity. [281] Structural damage to the ICC network with slow wave destabilization also is observed. [282] In rats, *T spiralis* infection disrupts intestinal motility and induces GMCs. [283] In previously infected rats, subsequent exposure to *T spiralis* larvae evokes muscle contraction, which can be blocked by a 5-HT antagonist and the mast cell stabilizer doxantrazole. [284] Jejunal muscle from *T spiralis*–infected rats exhibits increased responses to the muscarinic agonist carbachol, CCK, and 5-HT, in addition to enhanced peristalsis. [285], [286] In contrast to its excitatory effects on smooth muscle, *T spiralis* inhibits myenteric nerve activity and reduces acetylcholine and norepinephrine release, with peak effects 6 days after infection. [287] *T spiralis* reduces the intestinal content of substance P by 73% and VIP by 59%, and markedly inhibits NO synthase type 2 gene transcription, protein expression, and enzyme activity. [288], [289] The effects of *T spiralis* infection on smooth muscle and myenteric nerves may persist for 1 to 2 months after acute infection resolves (Fig. 11-12). [290], [291]

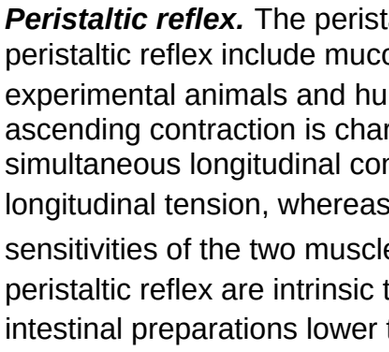

FIGURE 11-12. Contractile responses of intestinal muscle tissues to carbachol (*open bars*) and potassium chloride (*black bars*) at various time points after infection with *Trichinella spiralis*. Both stimuli evoke exaggerated contractions for 42 days after acute infection. (From ref. [291].)

Several immune mechanisms may underlie the effects of infection with *T spiralis*. Macrophages and lymphocytes penetrate intestinal longitudinal muscle, and their cellular protrusions contact myocytes. [292] The inhibition of myenteric nerve activity accompanying *T spiralis* infection is preserved in athymic rats, suggesting participation of T-cell–independent pathways. Conversely, the induction of increased contractions by carbachol and 5-HT is not observed, indicating T-cell–dependent responses as well. [285], [287] Reconstitution of CD $_4$ T cells to mice that are athymic, CD $_4$ cell deficient, or major histocompatibility complex II deficient restores the enhanced muscle contractility, confirming the role of T cells. [293] The ability of an interleukin-1 (IL-1) antagonist to prevent the neural effects of *T spiralis* infection suggests a role for IL-1 release, likely from macrophages. Increased muscle contraction with during infection with *T spiralis* is blunted in IL-5–deficient mice, indicating a role for this interleukin as well. [294] Sustained increases in smooth muscle cyclooxygenase-2 have been observed in mice infected with *T spiralis*. [295] Selective in vitro cyclooxygenase-2 inhibition reduces smooth muscle responses to agonist stimulation.

Internal Stimuli

Localized Reflexes

Peristaltic reflex. The peristaltic reflex is demonstrable in the small intestine and colon and produces aboral propulsion of lumenal contents. Stimuli that evoke the peristaltic reflex include mucosal pinching, hypertonic saline infusion, and insertion of a solid bolus. The peristaltic reflex consists of two phases that are observable in experimental animals and humans: an excitatory response proximal to stimulation (ascending contraction) and a distal inhibition (descending relaxation). [88], [296] The ascending contraction is characterized by simultaneous circular muscle shortening and longitudinal muscle relaxation, whereas the descending relaxation involves simultaneous longitudinal contraction and circular relaxation. Distention of guinea pig ileum to pressures of 0.5 to 1.5 cm H $_2$O, induces isolated increases in longitudinal tension, whereas at 1.5 to 3.0 cm H $_2$O, the longitudinal tension increase is followed by a propagative circular contraction, indicating differential sensitivities of the two muscle layers. [297] Afferent neuron activation is needed to elicit the peristaltic reflex. Cell bodies of the primary sensory neurons initiating the peristaltic reflex are intrinsic to the intestinal wall. [298] Radial stretch is the most potent stimulus for inducing the reflex. [51] Faster stretching and the use of longer intestinal preparations lower the threshold for peristalsis. [299] Receptors that sense radial stretch are mucosal in location, because stripping the mucosa or applying

lumenal topical anesthetics abolishes the reflex. Nonmucosal receptors also are suggested by experiments in which blunted peristaltic reflexes persist despite chemical destruction of the mucosa by silver nitrate or tannic acid. [300] Transmitters that mediate the peristaltic reflex have been extensively studied. Ascending contractions are partially inhibited by atropine at low levels of stimulation, whereas tachykinin receptor antagonists or antisera block contractions induced by intense radial stretching, indicating dual cholinergic and tachykinin mediation. [297], [301] Acetylcholine, substance P, and neurokinin A are released by radial stretch. [301] Muscarinic M_1 receptors participate in interneuronal transmission, whereas M_3 receptors mediate the smooth muscle contraction. [302] In guinea pig preparations, NK_1, NK_2, and NK_3 receptors all participate in peristalsis in concert with cholinergic pathways. [303], [304] The mediators of the descending relaxation likely are VIP and NO because both are released during peristalsis. [305] NO synthase inhibitors such as *N*-?-nitro-L-arginine and *N*-nitro-L-arginine methyl ester inhibit descending relaxations elicited by colonic distention. [46], [306] Capsaicin-sensitive afferent neurons mediate both ascending and descending components of the peristaltic reflex. [307] Other peptidergic neurons participate in the reflex. It is hypothesized that stretch activates aborally projecting somatostatin neurons that inhibit the activity of opioid neurons causing a reduction in met-enkephalin release. [308] The decrease in met-enkephalin produces relaxation by VIP and NO release. d-Opioid receptor antagonists act synergistically with 5-HT_4 receptor agonists to facilitate colonic peristalsis, also indicating a physiological modulatory role for opioids in the peristaltic reflex. [309] Serotonin is released with gut distention and lowers the threshold for evoking the peristaltic reflex, indicating an important role for this transmitter. [310] Mucosal stimulation initiates the reflex by activating 5-HT_4/5-HT_{1P} receptors on sensory neurons containing CGRP in human intestine and 5-HT_4/5-HT_{1P} and 5-HT_3 receptors in guinea pig colon. [311] In human colon, the 5-HT_3 antagonist granisetron has no effect on the ascending contraction or descending relaxation. [228] In isolated human, rat, and guinea pig colon tissue, 5-HT_4 agonists activate CGRP pathways that evoke ascending substance P release and descending VIP release (Fig. 11-13). [312] In guinea pig colon, 5-HT_3 and 5-HT_4 receptors may be arranged in parallel in the mediation of the peristaltic reflex. [313] In this species, selective 5-HT_3 and 5-HT_4 receptor antagonists are additive in their inhibition of colonic peristalsis. [314] In marmoset small intestine, it is hypothesized that 5-HT_4 receptors are activated by low concentrations of 5-HT, whereas higher concentrations produce 5-HT_3 receptor activation. [315]

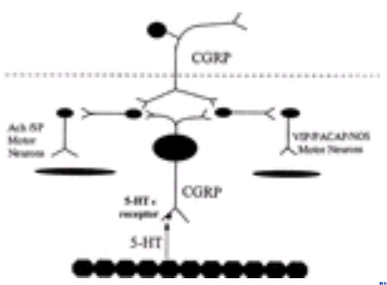

FIGURE 11-13. Neural pathways underlying the peristaltic reflex. Mucosal stimulation evokes 5-HT release, which activates release of calcitonin gene–related peptide (*CGRP*), which then stimulates ascending motor pathways containing the contractile agents acetylcholine (*Ach*) and substance P (*SP*) and descending motor pathways containing the relaxant agents vasoactive intestinal polypeptide (*VIP*), nitric oxide (*NO*), and pituitary adenylate cyclase–activating peptide (*PACAP*). (From ref. [312].)

Other peptides can modulate the peristaltic reflex, including GABA and PACAP. Endogenous purinergic pathways suppress peristalsis by activation of P_2 receptors. [316] Stimulation of cannabinoid CB_1 receptor pathways inhibits peristalsis in guinea pig ileum. [317] The demonstration that a CB_1 antagonist increases activity in mouse colon suggests that endogenous cannabinoids may physiologically modulate peristalsis. [318] In rat duodenum, ascending contractions are blocked by CCK by simultaneous activation of CCK-A and CCK-B receptors. [319] Activation of endothelin ET_A receptors stimulates peristalsis in guinea pig small intestine, whereas ET_B receptor occupation inhibits peristalsis. [320] The ability of an ET_B antagonist to enhance peristalsis suggests a physiological modulatory role for ET_B receptors. Although traditionally considered to be mediated by intrinsic nerves, some evidence suggests that extrinsic pathways may modulate the peristaltic reflex. In guinea pig colon, sympathetic nerve stimulation inhibits cholinergic components of the reflex. [321] In isolated tissues, extrinsic denervation decreases basal CGRP levels and abolishes peristaltic reflexes activated by muscle stretch but not mucosal stimulation. [322] In these models, extrinsic sensory pathways are postulated to mediate responses to stretch, whereas intrinsic pathways activate peristalsis with mucosal stimulation. That CGRP antagonists abolish peristalsis evoked by both stimuli indicates CGRP involvement in both extrinsic and intrinsic pathways.

Rectoanal inhibitory reflex. Rectal distention produces an abrupt volume-dependent decrease in internal anal sphincter pressure, known as the *rectoanal inhibitory reflex.* The reflex also is activated by rectal electrical stimulation. In contrast, external anal sphincter pressure increases on rectal distention. The rectoanal inhibitory reflex permits efficient defecation on rectal filling, whereas reflex external sphincter contraction prevents accidental leakage of rectal contents until they may be volitionally evacuated. The reflex also may be important for anal canal sampling to discriminate among solids, liquids, and gas. The rectoanal inhibitory reflex is mediated by intrinsic neural pathways between the rectum and the internal sphincter, although some investigators suggest a role for spinal pathways. [323], [324] Rectal sensory fibers are responsible for activating the reflex, although fibers in the pelvic floor muscles may play some role. The reflex is abolished by rectal transection at the anal verge, but it can regenerate within 2 years of surgery in humans. [325] Internal anal sphincter relaxation in response to rectal stimulation is a consequence of VIP and NO release. [326]

Extended Reflexes

Inhibitory intestinointestinal reflex. The inhibitory intestinointestinal reflex represents a possible model for ileus. It is characterized by profound motor inhibition of up to several hundred centimeters of intestine as a consequence of abrupt stretching or dilation of a localized intestinal segment. This is associated with slow wave disruption generating dysrhythmic myoelectric activity. [327] In humans, the inhibitory effects of distention are more pronounced in the proximal small intestine than in the ileum. [328] The relevance of the reflex is that when distention results from mechanical obstruction or another cause, the bowel responds with decreased motility and tone, thus serving a protective function. In contrast to peristalsis, the intestinointestinal reflex is not mediated by mucosal receptors, because excision of the mucosa and submucosa has no effect. Instead, removal of the longitudinal muscle prevents the reflex. In dogs, abrupt distention of an excluded but not extrinsically denervated intestinal segment abolishes spike activity both in the excluded loop and the intact intestine through splanchnic pathways. [329] The reflex is abolished by spinal cord transection below T7, indicating a need for intact thoracolumbar spinal pathways.

Intestinal reflexes involving the stomach. *Enterogastric and enteropyloric reflexes* are defined as the respective inhibition of gastric emptying and stimulation of pyloric motility by small intestinal stimulation. Duodenal distention reduces intragastric pressure, inhibits antral motility, and induces pyloric contractions. [330] Capsaicin-sensitive vagal afferents mediate the inhibitory response to low volumes, whereas spinal afferents mediate the response to high volumes of duodenal inflation. [331] Similarly, delays in gastric emptying evoked by duodenal carbohydrate and protein perfusion are associated with increased vagal afferent activity and are inhibited by capsaicin treatment of vagal and spinal afferents. [332], [333] Intrinsic pathways are involved in pyloric responses, because duodenal transection prevents induction of pyloric contractions by duodenal distention. [331] Conversely, duodenal transection does not block inhibition of antral contractions or the delay in gastric emptying induced by duodenal distention, indicating mediation by pathways other than ascending intraduodenal nerves. [331] In some models, enterogastric reflexes are blocked by the CCK antagonist MK-329. [332], [334] The magnitude of the enterogastric response depends on the stimulus intensity, its nature, and the length and region of intestine that is stimulated. Exposure of 15 cm of duodenum to hydrochloric acid inhibits gastric emptying, whereas lactic acid has no effect. [335] Perfusion of 150 cm of intestine with glucose induces maximal inhibition, whereas exposure of 15 cm has no effect on gastric emptying. [336] Duodenal peptone perfusion is more effective than equicaloric glucose perfusion. [333] Complementary to enterogastric reflexes, gastric distention abolishes fasting duodenojejunal motor activity and delays intestinal transit. [337] This gastroduodenal inhibitory reflex is unaffected by vagotomy but is reduced by celiac plexus sectioning or by small intestinal denervation, showing mediation by nonvagal pathways. [338] Mediation of this reflex by NO release in the celiac plexus also has been reported. [339]

Ileal brake. Gastric motility is inhibited more by ileal glucose perfusion than by duodenal administration. [340] Similarly, ileal short-chain fatty acid perfusion potently reduces antral contractions and reduces transpyloric flow. [341], [342] Ileal lipid or carbohydrate perfusion also reduces duodenal and jejunal motility and proximal intestinal transit more potently than proximal perfusions. [343] These inhibitory nutrient effects define the *ileal brake,* which likely serves a protective function to prevent the distal intestine from being overwhelmed by massive nutrient loads. Lipids are the most potent stimulators of the ileal brake, with oleate being 20-fold more potent than glucose at inhibiting gastric emptying. [344] Mediators of the ileal brake are incompletely characterized. a $_1$-Adrenergic receptors and ß $_1$-adrenergic receptors, opioid, and 5-HT_3 antagonists blunt ileal lipid-induced motor inhibition. [345], [346] Naloxone antagonizes the ileal brake only when administered to the proximal intestine, indicating a dependence on opioid pathways located on the efferent end of the reflex. [347] Ileal lipids and short-chain fatty acids release GLP-1, neurotensin, and peptide YY (PYY). [348], [349] and [350] Intravenous PYY prolongs the duodenal MMC cycle length and delays intestinal transit, mimicking the effects of ileal nutrients. [349] PYY immunoneutralization accelerates intestinal transit inhibited by ileal lipids, confirming dependence of the ileal brake on endogenous PYY. [351] Inhibition of duodenal motility by ileal protein is reversed by a GLP-1 receptor antagonist, indicating a role for this peptide as well. [352]

Extended colonic reflexes. Ileal distention evokes colonic relaxation, whereas colonic distention retards intestinal transit, reduces ileal motility, and disrupts

intestinal slow wave cycling. [353], [354] Sectioning the splanchnic nerves and ablating the prevertebral ganglia abolish these colonic reflexes, indicating mediation by extrinsic sympathetic pathways. [355] Topical anesthesia of the rectum prevents inhibition of intestinal contractions by rectal distention, indicating participation of mucosal receptors. [353] Colonic perfusion of mixed nutrients inhibits duodenal fasting activity. [356] Colonic distention delays the onset of MMC activity by activation of nicotinic ganglionic receptors. In dogs, the effects of ileal distention on colonic tone are not solely mediated by adrenergic, nicotinic, or NO pathways. Colonic perfusion of lactose and short-chain fatty acids reduces gastric tone, indicative of a colonic brake. [357] The role of PYY in the mediation of the colonic brake is uncertain, although colonic perfusion of short-chain fatty acids evokes PYY release. [358] Colonic stimulation also evokes reflex responses within the colon. Inflation of a distal colon balloon inhibits motility of a proximal colonic loop through a $_2$-adrenergic receptor–mediated pathways. [359] Distension of the rectosigmoid junction distention elicits increased rectal pressure and anal relaxation, with resultant expulsion of a rectal balloon. [360] This rectosigmoid-rectal reflex is postulated to facilitate defecation. Conversely, sigmoid colon distention increases rectosigmoid junction pressure by activation of mucosal receptors, possibly promoting fecal retention. [361], [362]

CORRELATION OF MOTOR PATTERNS WITH SMALL INTESTINAL AND COLONIC TRANSIT

Small Intestinal Transit

Propulsion of small intestinal contents depends on the lumenal caliber, wall tone, and amplitude of phasic contractions. Scintigraphic studies report a mean intestinal transit time of 221±49 minutes in healthy humans, with a range of 131 to 322 minutes. [363] Many studies report no effect of age or gender, although some have observed slower intestinal transit in women. [364], [365] As expected from manometry studies, transit in the upper intestine is more rapid than distally. In rats, transit across the proximal half of the intestine is 30 minutes versus 2.5 hours in the distal half. [366] In contrast to the stomach, where solids and liquids are handled differently, [131] I-labeled solids and ^{99m}Tc-DTPA–labeled water are propelled through the intestine at similar speeds. [367], [368] However, evidence suggests that the ileum may selectively retain bran while allowing liquids to pass into the colon. [369] Small intestinal nutrient transit depends on the caloric density and the nutrient class. Inert substances such as guar have little effect on total intestinal transit time. [370] In studies of different protein solutions, intestinal transit slowed in proportion to the calories delivered, with a resultant increase in the amount of protein absorbed in the proximal intestine, thus maintaining a constant level of absorption across a broad range of nutrient loads. Similarly, intralumenal lipids delay intestinal transit in a manner dependent on their caloric density. [371] Proteins tend to delay small intestinal transit to greater degrees than lipids. [368]

Combining manometry with cinefluoroscopy has permitted the correlation of fasting and fed patterns with the movement of intestinal contents. Intestinal transit of inert substances is four times faster in phase III than in phase I of the MMC. Fifty percent of total flow occurs during phase III, which efficiently clears the small intestine of retained material. [164] Transit also occurs in phase II, with rapid propulsion occurring during the transition from phases II to III. On cineradiography, transit during phase III is characterized by intermittent boluses of 4 to 5 cm in length separated by 1- to 2-cm ring contractions. [372] After eating, the distribution of intestinal contents becomes more uniform. In dogs and humans, postprandial transit is faster and exhibits lesser fluctuations than during fasting. [164]

Colonic Transit

Colonic transit in healthy persons is characterized by a slow movement of lumenal contents from the cecum to the rectum over 1 to 2 days. Mean transit times are slightly faster in men, with associated increases in fecal weight. [373], [374] Middle-aged women tend to exhibit slower transit than younger women. [365] /SUP>Propulsion through the different colonic regions is not uniform. Ingested radiopaque markers dwell in the rectosigmoid colon the longest both in men and women, indicative of a storage function for this region. [375] Prolonged residence of markers in the ascending colon also has been observed. Early radiographic observations noted retropulsion from the pacemaker area in the transverse colon to the ascending colon, with proximal retention of feces. However, as fecal volume in the ascending colon increases, retroperistaltic patterns give way to lumenally occlusive, aborally propagating contractions. Solids and liquids are handled differently in discrete regions of the colon. Solid residue is initially retained in the ascending colon, whereas liquids are propelled aborally. [376] In the transverse colon, solids and liquids are propelled similarly. Studies using nonabsorbable isotonic electrolytes show that the distal colon exhibits significant fluid retentive capabilities, suggesting that this region regulates stool output during diarrheal conditions. [377] With defecation, different regions exhibit different emptying efficiencies, with evacuation of 20% of marker from the right colon, 32% from the left colon, and 66% from the rectum. [378]

SPHINCTERIC FUNCTION OF THE LOWER GASTROINTESTINAL TRACT

Ileocecal Junction

The ileocecal junction exhibits characteristics distinct from the ileum and colon; it has a localized high-pressure zone in animal models that is not abolished by neural toxins. Manometric recordings in dogs show a basal pressure of 30 to 40 cm H_2O, with phasic contractions greater than 100 cm H_2O. [379] In humans, a high-pressure zone is less reliably observed, although phasic contractions are often noted. [380] Studies in patients with temporary ileostomies demonstrate high-pressure regions 5 cm in length, with mean pressures of 10 mm Hg (Fig. 11-14). [381] Slow waves and spike potentials migrate across the canine and feline ileocecal junction into the colon. [382] In dogs, more than 50% of MMCs traverse the ileocecal junction in dogs, but, in humans, the structure participates in very few MMC cycles. [383] In dogs, more than 80% of phase III complexes traversing the canine ileocecal junction evoke colonic contractions. [355] In scintigraphic studies, fasting flow across the canine ileocecal junction is maximal before phase III, whereas in humans, changes in flow do not correlate with MMC phases. [383] Eating increases motor activity and flow across the ileocecal junction that, in dogs, are maximal 4 hours after a meal. [384] In humans, meals increase ileocecal junction tone and the proportion of time occupied by phasic activity. [381] GMCs and DCCs readily traverse the ileocecal junction and decrease ileocecal junction phasic and tonic activity. [381], [383]

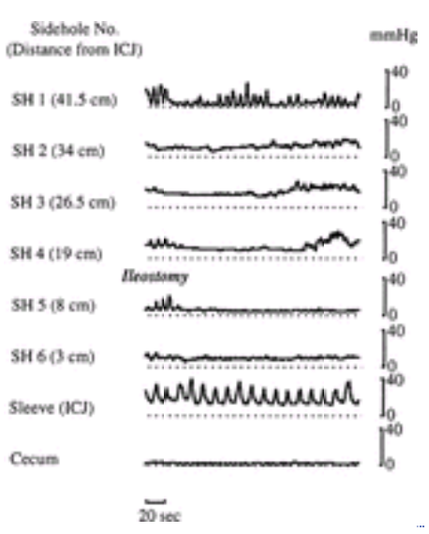

FIGURE 11-14. Pressure recordings from sideholes (*SH1 to 6*) in the ileum, a sleeve sensor in the ileocecal junction (*ICJ*), and the cecum from patients with ileostomies. The ileocecal junction exhibits increases in tone and phasic motor activity unrelated to basal ileal or cecal motility. (From ref. [381].)

The ileocecal junction safeguards against fecal reflux from the colon into the ileum. Attempts to propel material in retrograde fashion across the structure meet with resistance. In humans and animals, surgical excision of the ileocecal junction increases ileal bacterial counts, suggesting that the structure maintains relative sterility of the ileum. [385] In canine and human tissues, colonic distention evokes reflex ileocecal junction contraction, which is unaffected by transection of vagal or pelvic nerves. [380], [381], [386] Conversely, splanchnic nerve transection blocks the reflex, indicating the importance of extrinsic sympathetic pathways. The anatomic conformation of the ileocecal junction likely contributes to this response in that at its point of insertion into the cecum, it is maintained at a

constant acute angulation by fibrous connections. Severance of this fibrous tissue such that the acute angulation is lost renders the ileocecal junction incompetent, and coloileal reflux results. [387]

The role of the ileocecal junction as a barrier to aboral flow from the ileum into the colon is less clear. Early studies showed that the structure does not impede ileal liquid transit. Some manometric studies have reported reflex ileocecal junction relaxations with ileal distention, whereas others have noted reflex contractions. [388] Colonic filling from the ileum is characterized by bolus movements separated by intervening periods of stasis, suggesting that the ileum regulates cecal delivery. [389] Studies with Heidelberg capsules show lag times of 0.8 to 2.5 hours for this passage of solids across the junction, but other investigations show no discrimination of solid versus liquid passage into the cecum. [390] Furthermore, patients with surgical resection of the ileocecal junction exhibit normal transit of radiolabeled beads, suggesting little importance as a regulator of forward flow. [391] However, excision of a long segment of small intestine and removal of the ileocecal junction accelerate transit, showing that the structure can be a barrier to aboral propagation under conditions of high flow.

Anus and Pelvic Floor

The mean length of the anal canal is 2.8 cm, shorter around its circumference in women than in men. [392] There is a symmetric, functional region of elevated pressure extending above this level that helps to maintain fecal continence. Maximal pressures occur in the distal 1 to 2 cm of the anus, but pressures greater than intrarectal levels are measured 6 cm above the most proximal aspect of the anus. The internal anal sphincter provides 85% of the resting anal tone, which in humans is 40 to 80 mm Hg (Fig. 11-15). With voluntary contraction of the external anal sphincter, pressures exceed 150 mm Hg. [393] Maximal squeeze pressures are higher and squeeze durations are longer in men, but resting tone is similar in men and women. [394] Both resting tone and maximal squeeze pressure decrease with increasing age. [395] Age-related changes include decreased external sphincter thickness but increased internal sphincter thickness. [392] With sudden rectal distention, the contribution of the internal sphincter drops to 40% of the resting pressure, whereas the structure contributes 60% of the tone with constant rectal stimulation. [48] Ambulatory manometry records 14 to 19 spontaneous anal relaxations occurring per hour while awake and 4 per hour during sleep, each lasting 16 to 19 seconds. [183] Some of these relaxations are correlated with flatus passage.

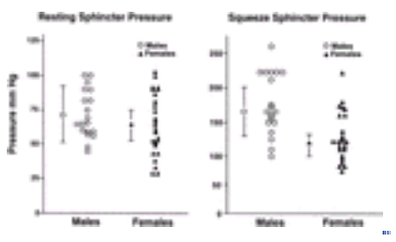

FIGURE 11-15. Resting anal sphincter pressure and maximal anal squeeze pressure of healthy men and women. (From ref. [394].)

The internal anal sphincter exhibits smooth muscle cell characteristics that facilitate its sphincteric function. Spontaneous electrical activity occurs in three regions: an upper region 15 to 20 mm from the anal verge with a frequency of 6.8 cpm, a transitional region with a frequency of 15.9 cpm, and a lower region within 5 mm of the anal verge with a frequency of 24.1 cpm. [396] Tone is generated in the two lower regions, whereas relaxant responses are observed in all regions with electrical depolarization.

Coordination of Defecation Passage through the colon promotes fecal dehydration, such that on reaching the rectum the stool is semisolid to solid. The rectum exhibits a compliant wall that allows it to serve as a reservoir for fecal material until it can be expelled. The internal anal sphincter provides tone to prevent accidental loss of stool, whereas the external sphincter voluntarily contracts if unwanted loss of feces is impending. In contrast to most striated muscle structures, the external anal sphincter and puborectalis muscle exhibit spontaneous tone, which promotes fecal continence. Indeed, maximal anal canal squeeze pressures are observed where the puborectalis overlaps the external sphincter. [397] The uppermost loop of the external sphincter, formed from the deep external sphincter muscle, attaches anteriorly on the pubis. The intermediate loop, formed from the superficial external sphincter, attaches posteriorly to the coccyx. The base loop, formed from the subcutaneous external sphincter, attaches to the perianal skin. Tight contraction of the three loops produces anal occlusion. Electromyographic studies indicate that the pelvic floor musculature behaves as a single muscle, with all muscle groups activating in response to a localized stimulus. [398] The anorectal angle created by the puborectalis muscle provides a functional obstruction to accidental loss of stool at rest. [399] Epithelial nerve endings in the rectum and anus allow the differentiation of solids, liquids, and gases before volitional expulsion. The rectum is innervated with ganglion cells and nerve fibers that mediate activities of the rectoanal region, including the rectoanal inhibitory reflex. The rectum has few free or organized nerve endings and is insensitive to most stimuli. In contrast, the transitional zone above the pectinate line, the anal crypt region, and the anal canal possess many free nerve endings and organized nerve endings that sense anal canal contents. Defecation involves the interaction of several different structures in and surrounding the rectum and anus. Tonic puborectalis contraction maintains an anorectal angle of 90° to preserve continence. Sitting erect contributes to continence by tightening the anorectal angle. [400] With defecation, puborectalis relaxation produces a more acute rectoanal angle (110°), and the internal anal sphincter relaxes. Fecal evacuation is aided by assuming a position in which the hips are flexed, further opening the rectoanal angle. Rectal contraction increases lumenal pressure to provide a propulsive force for defecation. Contraction of the rectus abdominus muscles, diaphragm, and terminal aspects of the other levator ani muscles increases intra-abdominal pressure and provides additional force. With straining, there is a physiological descent of the pelvic musculature of about 1 cm; this value is increased in incontinent patients. [401] After fecal passage, a rebound contraction of the external sphincter and pelvic floor muscles occurs. In contrast to defecation, the muscular events associated with flatus passage do not open the rectoanal angle. Rapid abdominal pressure increases, and pelvic floor muscle contractions, coupled with colonic contractions and increased rectal pressure, force gas past the acutely angled anorectum with retention of feces. [402]

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CHAPTER 12

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MOTILITY OF THE BILIARY TRACT

Understanding biliary tract function is a clinically significant goal. In the United States alone, more than 20 million people have gallstones or have had a cholecystectomy. Furthermore, the total direct costs associated with biliary tract disease, which amount to billions of dollars annually, are greater than the costs for any other gastrointestinal ailment, including colorectal cancer and peptic ulcer disease. [1] Abnormal motility is a hallmark of biliary tract disorders, including those of both calculous and acalculous origins; therefore, it is important to understand how motility of the gallbladder and sphincter of Oddi (SO) are normally regulated and what changes in these structures lead to dysmotility under pathophysiological conditions.

This chapter provides an overview of the current knowledge of biliary tract motor activity. Because motility primarily involves the activities of two cell types, neurons and smooth muscle, the chapter is organized to provide a summary of the basic physiology of the nerves and smooth muscle in the gallbladder and SO, with clinical correlations provided whenever possible. These sections also describe how the nerves and smooth muscle of these organs function during the bile retention and bile flow phases of the feeding cycle. Finally, a section at the end of the chapter describes what is currently known about the roles of nerves and smooth muscle in the biliary tract under pathophysiological conditions.

GALLBLADDER

The incidence of abnormal gallbladder motility in patients with biliary disease is high, and therefore evaluation of gallbladder motility is a common diagnostic approach for patients with clinical evidence of biliary dysfunction. Functional radiographic imaging (biliary scintigraphy) is the most common means of evaluating biliary motility. Functional biliary scintigraphy is performed after administration of a chemical analog of cholecystokinin (CCK) to a patient who has previously received iminodiacetic acid radiotracer. Radiographic images are captured over a 20- to 30-minute period to determine the gallbladder ejection fraction, which is calculated using a standard time-activity curve. Functional scintigraphy has an estimated sensitivity of greater than 82%, a specificity of greater than 87%, and a positive predictive value for detecting abnormal gallbladder function of greater than 90%. [2], [3] and [4] It is therefore a valuable tool for predicting whether a given patient's symptoms will be resolved by cholecystectomy. Numerous reports suggesting that patients with diminished gallbladder emptying, as determined by functional scintigraphy, benefit from laparoscopic cholecystectomy further support this approach. [5]

Given that biliary tract dysmotility is so prevalent in gallbladder disease, a comprehensive understanding of the neurons and smooth muscle that are responsible for gallbladder contractile activity is critical. Determination of the cellular mechanisms that are responsible for normal and pathological gallbladder motility is difficult in humans; therefore, much of what is known about the structure and function of gallbladder neurons and smooth muscle is derived from animal studies.

Gallbladder Neurobiology

The wall of the gallbladder consists of a mucosal layer with an underlying lamina propria, a muscularis layer, and a serosal layer. Neural networks can be found in each of these layers, and these neural networks are interconnected by nerve bundles. The morphology of nerves of the gallbladder have been investigated in many species including human, rhesus monkey, pig, dog, cat, marmoset, guinea pig, North American opossum, Australian brush-tailed possum, and mouse. [6], [7], [8], [9], [10], [11] and [12] The serosal plexus is the most prominent network of nerves in the gallbladder. It contains small, irregularly shaped ganglia that are connected by bundles of unmyelinated axons. The serosal plexus is connected to nerve bundles that follow the extensive vascular distribution in this layer. The neural plexus of the muscularis, which does not contain ganglia, is prominent in humans and other larger species. The mucosal plexus consists of nerve bundles that travel in the lamina propria, with branches that pass through the mucosa and often terminate near the epithelial cells. In some species, including humans, the mucosal plexus contains occasional small ganglia consisting of one to three neurons.

The structure of gallbladder neurons has been studied in animal models by injecting individual neurons with intracellular markers. [10], [13] Gallbladder neurons of these species are simple in structure when compared with intestinal neurons, because they consist only of a cell body and one or two long processes with no appreciable dendritic arborization. The axons typically pass from the ganglion of origin into interganglionic fiber bundles, where they travel for some distance before terminating in the muscular plexus. Unlike ganglion cells in the submucosal plexus of the bowel, gallbladder neurons do not project to the vascular nerve bundles, indicating that gallbladder ganglion neurons do not contribute to the modulation of vascular resistance in the gallbladder.

Neuroactive Compounds in Gallbladder Nerves

Gallbladder neurons. Evaluating the influence of gallbladder neurons on gallbladder function requires the identification of the neurochemical phenotypes of these cells. The complexity of the enteric nervous system is reflected by the large number of neuroactive compounds that are found in the neural plexuses of the gut. Studies of the neurotransmitter content of gallbladder neurons have revealed that many putative neurotransmitters also exist in this system. However, unlike in the gut, where distinct populations of cholinergic and noncholinergic neurons exist, all gallbladder neurons are apparently cholinergic because all are immunoreactive for the essential biosynthetic enzyme for acetylcholine choline acetyltransferase (ChAT). [9], [14] In the guinea pig, the most extensively studied species, the overall population of cholinergic neurons can be divided into two distinct subpopulations based on chemical coding patterns. [15], [16] The larger population, representing more than 80% of the neurons, is immunoreactive for substance P, neuropeptide Y (NPY), somatostatin, and orphanin FQ (OFQ, also known as nociceptin), as well as ChAT. The remaining neurons are immunoreactive for ChAT, in addition to vasoactive intestinal polypeptide (VIP), pituitary adenylate cyclase–activating polypeptide (PACAP), and nitric oxide synthase (NOS). Chemical coding of gallbladder neurons has also been described in other species, including human. [9], [11], [12], [17], [18] Most human gallbladder neurons express VIP, NPY, somatostatin, and PACAP, and most of these neurons are also tachykinin immunoreactive. [9], [11], [12], [17] NOS immunoreactivity and nicotinamide adenine dinucleotide phosphate (NADPH) dehydrogenase staining has also been reported in a small subset of human gallbladder neurons, but unlike the pattern seen in the guinea pig, the NOS-positive neurons are VIP negative. [17]

Sympathetic nerve fibers. Catecholamine histofluorescence and antibodies directed against the biosynthetic enzymes tyrosine hydroxylase (TH) and dopamine ß-hydroxylase have been used to identify the sympathetic postganglionic nerves in the wall of the gallbladder. [6], [8] Injection of axon tracers into the gallbladder wall demonstrates that sympathetic postganglionic projections to the gallbladder arise in the celiac ganglia. [8] Sympathetic postganglionic nerves pass along serosal blood vessels, and they are also abundant in the ganglionated plexus of the gallbladder, with numerous axonal varicosities surrounding gallbladder neurons. [8] In the perivascular plexus of the guinea pig and the human gallbladder, the sympathetic nerve fibers also are immunoreactive for NPY, and our research in the guinea pig has confirmed OFQ immunoreactivity. [16]

Sensory nerve fibers. It is not yet possible to distinguish sensory (afferent) fibers that arise from spinal ganglia versus those from nodose ganglia; however, the ganglionated plexus of the gallbladder clearly is rich in afferent nerve fibers that are immunoreactive for both substance P and calcitonin gene–related peptide (CGRP).[8], [9], [14] These nerves are likely to be extrinsic primary afferent fibers because gallbladder neurons are not immunoreactive for CGRP.[9], [12] The substance P/CGRP–immunoreactive nerve fibers are abundant in ganglia, in interganglionic fiber bundles, and in the perivascular plexus. Within the ganglia, the substance P/CGRP fibers appear to ramify and give rise to processes that terminate on gallbladder neurons. Substance P/CGRP–positive nerve fibers also are immunoreactive for PACAP,[16] suggesting that activated afferent nerves can release tachykinins, CGRP, and PACAP.

Electrical and Synaptic Properties of Gallbladder Neurons

Electrical properties. The electrical properties of gallbladder neurons have been studied in guinea pig, opossum, and human.[10], [13], [19] Unlike enteric neurons, gallbladder neurons can be classified into a single group on the basis of their electrical properties. They are relatively inexcitable, because they rarely exhibit spontaneous action potentials, and they fire stimulus-induced action potentials only at the onset of a depolarizing current pulse. These features indicate that gallbladder neurons are normally quiescent and must be driven by excitatory inputs to release their neurotransmitters onto their target tissues, such as smooth muscle. The action potential of the gallbladder neuron consists of a rapid upstroke that involves the influx of Na^+ and Ca^{2+} ions. An afterhyperpolarization of moderate duration that can be divided into an early and a late phase follows the action potential. These early and late phases involve two sequential Ca^{2+}-activated K^+ conductances.[13] The early phase is mediated by large-conductance, Ca^{2+}-activated K^+ (BK) channels, as well as by delayed rectifier K^+ channels. The late phase is sensitive to apamin, a selective blocker of small conductance, Ca^{2+}-activated K^+ (SK) channels. The late phase of the afterhyperpolarization apparently contributes to the inexcitability of gallbladder neurons because inhibition of this phase dramatically increases the excitability of these cells.[13]

Synaptic inputs. Gallbladder neurons are not inherently active and therefore must be driven by synaptic inputs to send signals to the smooth muscle. Fast excitatory postsynaptic potentials (EPSPs) and slow EPSPs, but not inhibitory synaptic potentials, can be elicited by focal stimulation of interganglionic axon bundles.[10], [13], [19] Fast excitatory synaptic input to gallbladder neurons is the principal driving force in the neuromuscular axis of the gallbladder. Stimulation of interganglionic nerve bundles, or cystic nerves, elicits fast EPSPs in essentially all gallbladder neurons.[10], [13], [19], [20] Fast EPSPs in the gallbladder are mediated exclusively by acetylcholine, as evidenced by the finding that they are completely abolished by the nicotinic receptor antagonist hexamethonium. Gallbladder neurons receive a significant input from vagal preganglionic nerves. Stimulation of vagus nerves in vivo elicits gallbladder contraction.[21], [22] and [23] Furthermore, neurons in the dorsal motor nucleus of the vagus are labeled after retrograde axonal tracers are injected into the gallbladder wall.[8] These vagal postganglionic projections represent the major source of nicotinic synaptic input to gallbladder neurons.[20] In animals that have been vagotomized, cystic nerve stimulation does not elicit a synaptic response. Therefore, unlike the ganglia of the intestines, in which most fast synaptic inputs arise from other ganglion cells, fast synaptic input to gallbladder neurons is mainly derived from vagal preganglionic nerve terminals. However, in about 20% of the neurons tested in vagotomized preparations, stimulation of interganglionic fiber bundles elicits a fast EPSP, indicating that interganglionic communication among gallbladder neurons does exist. Stimulation of interganglionic connectives at high frequency (10–20 Hz) elicits a slow EPSP in 20% to 30% of gallbladder neurons.[10], [13], [24] The slow EPSP consists of a depolarization of up to 8 mV, lasting about 1 minute. Unlike slow excitatory synaptic events in the gut, which involve a decrease in K^+ conductance, the slow EPSP in gallbladder ganglia is associated with a decrease in input resistance and is mediated by an opening of nonselective cation channels. The slow EPSP in gallbladder ganglia is mediated by the release of peptides from extrinsic afferent fibers that may be associated with the subjective experience of biliary pain. A subset of the sensory fibers in the wall of the gallbladder is activated only by noxious stimulation of the biliary system, indicating that these are pure visceral nociceptors.[25] Within the gallbladder, the pain afferents are likely to be immunoreactive for both substance P and CGRP.[8], [9], [26] In the gut, substance P is released from the peripheral endings of sensory nerve fibers as part of the process of inflammation.[27] Capsaicin, which causes peptide release from small-diameter afferent fibers, induces the release of substance P and CGRP when it is applied to the gallbladder in vitro.[26] When applied to gallbladder neurons, tachykinins and CGRP, as well as capsaicin, cause a membrane depolarization that is mediated by neurokinin 3 receptors and resembles the slow EPSP.[24], [28] Furthermore, neurokinin 3 receptor blockade inhibits the stimulus-induced slow EPSP and the depolarization caused by capsaicin.[24] Therefore, sensory fibers that pass through gallbladder ganglia can act locally to modulate ganglionic output by synapsing on gallbladder neurons and increasing their excitability. This local axon reflex circuit may contribute to a cycle that induces the subjective experience of biliary pain in gallbladder disease. Increased ganglionic output caused by slow EPSPs may lead to increased gallbladder contractility. If bile outflow from the gallbladder is inhibited or obstructed, pressure may then increase to higher levels and may lead to further activation of sensory fibers. The activated sensory fibers would then elicit additional stimulation of gallbladder neurons, leading to further increases in intralumenal pressure and enhanced perception of pain.

Gallbladder Smooth Muscle Cells

Gallbladder smooth muscle cells are arranged in interposed bundles that are oriented in various directions within the muscularis of the gallbladder.[29] Within each bundle, these smooth muscle cells appear to be coupled because intact cells have a very low input resistance;[30] although, on ultrastructural examination, gap junctions are relatively sparse.[29]

The Action Potential: Ca^{2+} and Voltage-Activated K^+ Channels The electrical properties of gallbladder smooth muscle cells have been investigated in intact preparations using intracellular microelectrodes and in isolated myocytes using patch clamp recording techniques.[30], [31] Intact gallbladder smooth muscle cells generate spontaneous action potentials that consist of four phases:[30]

1. A rapid depolarization upstroke
2. A transient repolarization downstroke almost to the resting membrane potential
3. A plateau phase
4. A repolarization back to the resting membrane potential.

Spontaneous action potentials typically occur at a frequency of 0.3 to 0.4 Hz, which is significantly higher than the frequency of slow wave action potentials in smooth muscle of the gut. Two of the principal channels that are involved in the gallbladder smooth muscle action potential are the L-type Ca^{2+} channel and the delayed rectifier K^+ channel. The L-type Ca^{2+} channel blocker, nifedipine, abolishes the gallbladder smooth muscle action potential in intact tissue, as well as the voltage-activated inward current in isolated cells.[30], [31] The amplitude and duration of the spike and plateau phases of the action potential are augmented by the voltage-activated K^+ (K_V) channel blocker, 4-aminopyridine, and most of the voltage-activated outward current is mediated by K_V channels.[30], [32] Whereas dihydropyridine-sensitive Ca^{2+} channels and K_V channels are clearly involved in the action potential of gallbladder smooth muscle, the mechanisms responsible for pacing of the action potential and the generation of the plateau phase have not been resolved.

BK Channels Inhibition of BK channels with tetraethylammonium or charybdotoxin does not affect the action potential shape or the membrane potential of gallbladder smooth muscle. However, BK channels have been identified in single-channel recordings from gallbladder smooth muscle.[30] Charybdotoxin and tetraethylammonium cause a slight decrease in the voltage-activated outward current. We have demonstrated that BK channels are responsible for spontaneous transient outward currents in gallbladder smooth muscle.[33] These currents are activated by local Ca^{2+} release events (i.e., sparks) from the endoplasmic reticulum, and they are potentiated by caffeine and inhibited by ryanodine. Although the functional relevance of sparks and transient outward currents in gallbladder smooth muscle is not completely understood, they are likely to play a role in the regulation of gallbladder smooth muscle excitability because they are inhibited by CCK.

ATP-Sensitive K^+ Channels Another type of K^+ channel that has been identified in gallbladder smooth muscle is the ATP-sensitive K (K^+-ATP) channel.[34], [35] and [36] Direct activation of the K^+-ATP channel with lemakalim or pinacidil causes a prolonged hyperpolarization that is associated with an elimination of spontaneous action potentials. Activation of K^+-ATP channels is responsible for the inhibitory effects of CGRP and agonists of H_2 receptors for histamine.[35], [37] Receptor-mediated opening of the K^+-ATP channel in gallbladder smooth muscle involves the activation of the cyclic AMP (cAMP)–adenylate cyclase–protein kinase A signal transduction cascade.[34], [36] Activation of protein kinase C inhibits K^+-ATP channel activity in gallbladder smooth muscle,[36] but gallbladder smooth muscle K^+-ATP channels are not as prone to inhibition by protein kinase C as other smooth muscle cell types. Because activation of protein kinase A is a major mechanism for inhibition of smooth muscle, and because the effects of forskolin are completely blocked by glibenclamide,[34] it is likely that K^+-ATP channels are actively involved in receptor-mediated relaxation of gallbladder smooth muscle.

Gallbladder Emptying

Neuroendocrine Control of Gallbladder Contraction Numerous neurotransmitters and hormones are capable of causing gallbladder contraction, as demonstrated in functional studies involving measurement of gallbladder pressure or muscle strip tension (Fig. 12-1). The three compounds that clearly participate in gallbladder contractile events are CCK, acetylcholine, and tachykinins.

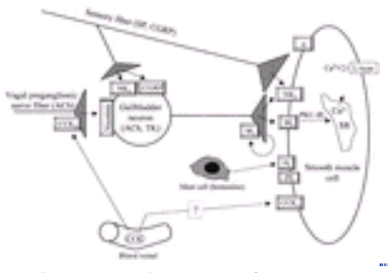

FIGURE 12-1. Excitatory transmitters, hormones, inflammatory mediators, receptors, and ion channels that have been identified in the neuromuscular axis of the gallbladder. Nerve terminals are represented by *triangles,* receptors are represented by *framed boxes,* and ion channels are represented by *boxes with arrows* indicating the direction of ion flow. a is an adrenergic receptor. *ACh,* acetylcholine; *CCK,* cholecystokinin; *CGRP,* calcitonin gene–related peptide; *EP,* prostaglandin E; *H,* histamine; *IP* $_3$*,* inositol 1,4,5-trisphosphate; *M,* muscarinic; *NK,* neurokinin; *PKC,* protein kinase C; *SP,* substance P; *SR,* sarcoplasmic reticulum; *TK,* tachykinins.

Cholecystokinin. One of the first and most significant hormones to be identified, CCK, was named in 1928 by Ivy and Oldberg [38] for its ability to contract the gallbladder. The mechanisms of CCK action in the gallbladder are described in greater detail later, in the section devoted to postprandial gallbladder contraction.
Acetylcholine. As described previously, all gallbladder neurons are cholinergic, and stimulus-induced release of acetylcholine from these neurons results in a contraction of gallbladder smooth muscle. Although M $_1$ to M $_4$ muscarinic receptors have been reported in the gallbladder, [39], [40], [41] and [42] the main muscarinic receptor subtype in the smooth muscle cells is M $_3$, whose activation induces smooth muscle contraction. [39], [40] Parkman and colleagues [42] demonstrated that the M $_1$ and M $_2$ receptors may provide an automatic feedback mechanism in the gallbladder because they are located on nerve terminals. Activation of M $_1$ receptors promotes transmitter release and M $_2$ receptor activation inhibits transmitter release. M $_2$ activation is linked to the phospholipase D–protein kinase C pathway, [43] whereas activation of M $_3$ receptors leads to phosphatidylinositol hydrolysis by phospholipase C as well as an inhibition of cAMP formation. [43], [44]
Tachykinins. Tachykinins are prevalent in the nerves of the gallbladder, including extrinsic afferent nerves as well as gallbladder neurons and their axons. Tachykinins produce a direct, concentration-dependent contraction of the isolated gallbladder, with a rank order potency of neurokinin A ? neurokinin B ? substance P, [23], [26], [45] which is a characteristic of neurokinin 2 receptors. In the gallbladder, the binding of tachykinins to neurokinin receptors is linked to protein kinase activation. [45] The finding that tachykinins are coexpressed with acetylcholine in gallbladder neurons indicates that these compounds may act together to promote gallbladder emptying on vagal stimulation.
Postprandial Gallbladder Contractions Postprandial gallbladder contraction is triggered by gastric emptying leading to the release of CCK from enterochromaffin cells in the epithelial lining of the duodenum. Although the concept that CCK acts as a hormone to cause gallbladder emptying is well established, it is also likely that CCK acts at several sites to promote functional gallbladder motility (Fig. 12-2). The most direct means of gallbladder contraction is for hormonal CCK to act on receptors located on gallbladder smooth muscle, which expresses the CCK-A but not the CCK-B receptor. [46] It is likely that low-affinity CCK-A receptors are present on gallbladder smooth muscle, because the EC $_{50}$ for the CCK-induced gallbladder contraction is in the 10- to 50-nM range. [47], [48] and [49] However, it is unclear whether the CCK receptors on gallbladder smooth muscle are a normal physiological site of action for CCK, because postprandial concentrations serum concentrations of CCK are in the 10- to 20-pM range, [50], [51] which is far lower than the threshold necessary for a direct action of CCK on gallbladder muscle strips. Therefore, a neural mechanism must be involved in the prokinetic effects of postprandial CCK release.

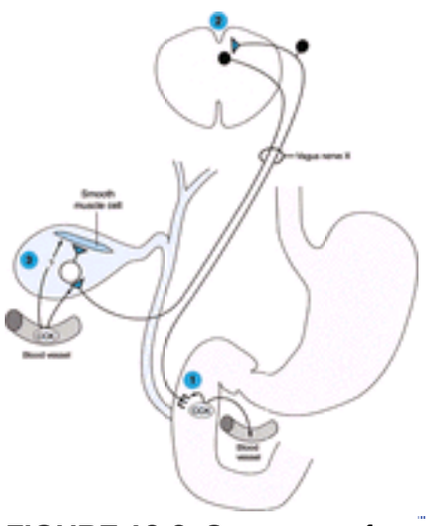

FIGURE 12-2. Sequence of events leading to postprandial gallbladder contraction: *(1)* cholecystokinin (CCK) released from the duodenal mucosa activates vagal afferent fibers and enters the bloodstream; *(2)* increased vagal afferent activity leads to increased vagal preganglionic output; *(3)* within the gallbladder, hormonal CCK acts at physiological concentrations on receptors located on vagal preganglionic nerve terminals in gallbladder ganglia. Activation of these presynaptic receptors leads to an increase in acetylcholine release and enhanced activation of gallbladder motor neurons. CCK may also act directly on gallbladder smooth muscle, but the receptors at this location appear to be low-affinity CCK-A receptors, which normally would not be activated by physiological concentrations of CCK.

Meal-induced gallbladder contractions and contractions induced by *physiological* concentrations of CCK in vivo are significantly attenuated by neural blockade in several species, including human. [51], [52], [53], [54], [55], [56], [57], [58], [59] and [60] Furthermore, hexamethonium, which blocks the vagal preganglionic input to gallbladder neurons, inhibits CCK- and meal-induced gallbladder contractions. [51], [57], [58] These data indicate that CCK can act on nerves to promote gallbladder motility. Results of electrophysiological studies suggest that CCK does not have a direct effect on gallbladder neurons, but it does have a potent presynaptic excitatory effect on nerve terminals in gallbladder ganglia, thereby increasing the release of acetylcholine onto gallbladder neurons. [10], [20], [61] CCK increases the amplitude of the cholinergic fast EPSPs in gallbladder ganglia, and typically converts subthreshold synaptically mediated depolarizations to responses that are accompanied by one or more action potentials. [10], [61] The action of CCK in gallbladder ganglia peaks at 1.0 nM and has a median effective concentration (EC $_{50}$) of 33 pM. Furthermore, in the presence of 10 pM CCK, synaptic currents increase about 20%. Thus, the neural action of CCK in the gallbladder occurs within the physiological concentration range. The nerve terminals that are sensitive to CCK are vagal preganglionic nerve terminals because these synaptic inputs are eliminated by vagotomy, as previously described. [20] Vagal afferent nerve fibers in the duodenum are another likely site of action for CCK released from enterochromaffin cells. Subdiaphragmatic vagal afferent fibers are sensitive to CCK, and postprandial physiological responses, such as increased gastric motility and pancreatic secretion, have been attributed to CCK-mediated increases in vagal afferent activity. [62], [63] After a meal, CCK stimulates vagal afferent nerve fibers, which act in the vagal motor complex to increase the rate of firing of vagal preganglionic neurons. Furthermore, CCK acts in gallbladder ganglia, as previously discussed, to increase the amount of acetylcholine released from the vagal motor terminals each time they are activated. The question remains: If CCK is normally acting through the neural mechanisms described, how does CCK-induced gallbladder emptying still occur (albeit diminished) after vagotomy or after liver transplantation with the donor gallbladder intact? The answer to this question probably lies in the fact that gallbladder emptying involves increased gallbladder tone in concert with a decrease in resistance at the SO. As described later, it is likely that the circuitry responsible for the CCK-induced decrease in SO resistance is left intact after vagotomy or liver transplantation. Therefore, changes in SO tone, in combination with the myogenic tone of the gallbladder, may lead to the flow of bile. Another potential mechanism for CCK-induced gallbladder emptying after vagotomy is that gallbladder denervation could lead to a sensitization of gallbladder smooth muscle, resulting in a leftward shift of the concentration-effect curve for CCK. This phenomenon is observed in patients who have had a truncal vagotomy—gallbladder contraction in response to CCK is significantly enhanced. [64]
Gallbladder emptying after vagotomy may be mediated through an enterobiliary neural reflex. Studies in the guinea pig and the Australian possum have demonstrated that the gallbladder receives projections from neurons located in the myenteric plexus of the duodenum. [8], [65] It is plausible that lumenal stimuli or mucosal CCK release may result in the activation of these neurons, which directly signal the gallbladder. Whereas the latter scheme could not contribute to gallbladder emptying after liver transplantation, the enterobiliary circuitry may contribute to gallbladder emptying after truncal vagotomy.
Interprandial Gallbladder Contractions During phase II of the migrating myoelectric complex (MMC), there is an increase in gallbladder pressure, accompanied by a transient flow of bile from the gallbladder to the duodenal lumen. This interprandial gallbladder motor response, which occurs in association with elevated antral and duodenal motor activity, has been demonstrated in several species including human, and it is thought to help maintain the enterohepatic circulation of bile salts. [66]
The mechanisms responsible for the phase II gallbladder contraction have not been clearly resolved, but they appear to involve a neural component because spontaneous and motilin-induced gallbladder contractions are reduced or abolished with atropine or hexamethonium. [67] However, it is not clear whether the neural

component involves the activation of a vagal reflex or whether it is limited to actions on intrinsic reflex circuits of the bowel. In one study, performed in dogs, inhibition of vagal reflex activity by transient vagal chilling or by acute vagotomy led to a decrease in MMC-related gallbladder emptying. [68] In contrast, MMC-related contractile activity is relatively normal after chronic vagotomy. [69], [70] and [71] If the biliary motor response that accompanies the MMC does not involve an extrinsic neural circuit, it may involve direct neural interactions between the gut and the biliary tree, or humoral factors may contribute. The gastrointestinal hormone motilin is thought to initiate the MMC because plasma motilin levels increase during or just before the onset of MMC activity, and exogenous motilin administration initiates an early burst phase of the MMC in the upper gastrointestinal tract, [72], [73] including the gallbladder. [52] Exogenous administration of motilin, or motilin analogs such as erythromycin, increases gallbladder motor activity, [74] and motilin induces the contraction of isolated gallbladder myocytes. [75] However, it is not yet clear how endogenous motilin would directly activate MMC-related gallbladder activity, which is primarily limited to phase II, because peak levels of motilin in the serum correspond to phase III of the MMC, when the gallbladder is quiescent.

Remaining Questions Related to Biliary Tract Contractility Although much has been learned in the last 2 decades regarding postprandial and interprandial gallbladder contractions, the mechanisms of excitation-contraction coupling in gallbladder smooth muscle cells remain less clear. A few studies, involving measurement of the percentage of shortening of gallbladder myocytes, have elucidated the intracellular pathways that mediate agonist-induced responses, whereas precisely how the activation of M_3 or CCK-A receptors leads to Ca^{2+} mobilization and contractile protein activation requires further clarification. Intracellular electrophysiological studies have shown that gallbladder smooth muscle cells generate spontaneous action potentials that are likely to be associated with increases in global Ca^{2+} and probably contribute to the basal tone of the gallbladder. Unfortunately, the generation of these events is not understood, nor is it known whether they are modulated in response to neurohormonal signals. Because gallbladder dysmotility is clearly a predisposing factor to a variety of clinical disorders, a thorough understanding of how basal tone in the gallbladder is maintained and how agonists contract gallbladder smooth muscle will advance the understanding of clinical biliary disease and may lead to therapeutic alternatives.

Gallbladder Filling

During fasting, gallbladder filling is related to the rate of hepatic secretion from the liver and the resistance to flow through the SO into the duodenum. During this process, hepatic secretory pressure is relatively high (=25–30 mm Hg), as is SO pressure (~11–30 mm Hg), whereas pressure within the biliary tree is maintained at a relatively low level (~5–15 mm Hg) because of the gallbladder's ability to accommodate newly synthesized bile. [66] The accommodation of the gallbladder during fasting relies on the ability of the epithelium to concentrate bile and on the receptive expansion of the gallbladder. The gallbladder therefore contributes to the modulation of pressure within the biliary system, and its contribution is likely to involve both passive and active mechanisms.

Passive Filling The fibroelastic properties of the gallbladder wall likely facilitate filling, allowing the organ to expand as bile is routed from the biliary tree to the gallbladder. The sheer size of this organ, relative to other structures in the biliary tree, favors the passage of bile into the gallbladder. According to the law of Laplace, tension in the wall of a hollow elastic structure is proportional to the product of pressure times diameter. Because the diameter of the gallbladder is significantly greater than that of the bile ducts, the path of least resistance would be gallbladder filling and expansion while the SO is resisting bile flow. As the gallbladder fills and its diameter increases further, the propensity for bile to flow into the gallbladder increases. Although this principle has never been applied to the gallbladder, many visceral structures that undergo expansion, including the urinary bladder, the alveoli, the heart, and the eyeball, have been shown to behave according to the law of Laplace. Since the early 1990s, several stretch-activated channels have been identified. Although this has not yet been investigated in the gallbladder, it is highly likely that gallbladder smooth muscle cells express stretch-activated channels that are activated as gallbladder tone increases slightly as a result of filling. These could be channels that induce a depolarization of the smooth muscle resulting in increased gallbladder basal tension or channels that elicit hyperpolarization and decreased tension. In cardiac myocytes, a stretch-sensitive channel with the properties of the BK channel has been identified. [76] As described previously, BK channels in gallbladder smooth muscle are responsible for spontaneous transient outward currents that are activated by sparks—local, ryanodine-sensitive, Ca^{2+}-release events. [30], [33] It remains to be determined whether the open-state probability of these channels and associated spontaneous transient outward current activity are altered by changes in smooth muscle tension or whether other, yet to be identified channels exist in gallbladder smooth muscle and contribute to the maintenance of myogenic tone during gallbladder filling.

Active Filling Several candidates exist as potential mediators of active gallbladder relaxation and resultant gallbladder filling (Fig. 12-3). Neurotransmitters that have an inhibitory effect on gallbladder smooth muscle include CGRP, norepinephrine, VIP, PACAP, and NO. Humoral factors that relax the gallbladder include pancreatic peptide and somatostatin. However, it is difficult to determine which of these compounds actually contribute to normal physiological gallbladder expansion, because many of their effects on the gallbladder have been observed at supraphysiological concentrations. Several potential schemes could contribute to this process, as described in the following discussion.

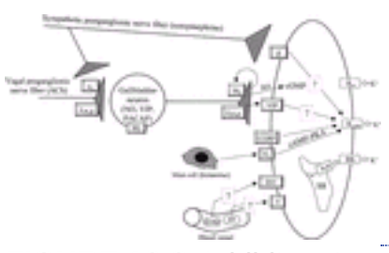

FIGURE 12-3. Inhibitory transmitters, hormones, inflammatory mediators, receptors, and ion channels that have been identified in the neuromuscular axis of the gallbladder. Nerve terminals are represented by *triangles,* receptors are represented by *framed boxes,* and ion channels are represented by *boxes with arrows* indicating the direction of ion flow. a_2 and ß are adrenergic receptors, and *d, ?*, and *μ* are opioid receptors. *ACh,* acetylcholine; *BK,* large-conductance Ca^{2+}-activated K^+ channel; *CGRP,* calcitonin gene–related peptide; *PE,* prostaglandin E; *H,* histamine; K_{ATP}, ATP-sensitive K^+ channel; K_V, voltage-activated K^+ channel; *M,* muscarinic; *NO,* nitric oxide; *PACAP,* pituitary adenylate cyclase–activating peptide; *PKA,* protein kinase A; *PP,* pancreatic polypeptide; *RyR,* ryanodine receptor; *SOM,* somatostatin; *SR,* sarcoplasmic reticulum; *SST,* somatostatin receptor; *VIP,* vasoactive intestinal polypeptide; *Y,* neuropeptide Y/PP receptor.

Release of inhibitory neuroactive compounds from gallbladder neurons. In the bowel, distinct subpopulations of enteric neurons (i.e., inhibitory motor neurons) act to decrease muscle tone. Compounds that have been proposed to contribute to relaxation of intestinal smooth muscle include NO, VIP, PACAP, and ATP. As mentioned previously, subsets of gallbladder neurons are immunoreactive for VIP or PACAP, [12], [15], [16] and [17] and they are immunoreactive for NOS. [12], [14], [16], [17] The neuroactive peptides VIP and PACAP cause relaxation of resting or precontracted gallbladder muscle strips from several species including human, [77], [78], [79] and [80] probably through the activation of adenylate cyclase. VIP is released by nerves in the gallbladder in response to electrical stimulation of the vagus nerves, further evidence of its neurotransmitter capability. [81], [82] In addition, relaxant responses to exogenous VIP vagal stimulation can be antagonized by VIP antisera. [79] Evidence indicating that NO relaxes gallbladder smooth muscle includes the finding that inhibition of NOS results in increased gallbladder tone and an enhancement of agonist-induced contractions. [83], [84] NOS inhibition also reduces stimulation-induced neurogenic relaxations, and NO donors have an inhibitory effect on gallbladder tone. [83], [84], [85] and [86] CGRP, which is present in extrinsic sensory fibers, can decrease tension in gallbladder muscle strips and cause a hyperpolarization of gallbladder smooth muscle cells. It is unlikely, however, that CGRP released from gallbladder afferent fibers contributes to relaxation because tachykinins are co-stored with CGRP in these nerve fibers, [8], [9], [14] and tachykinins have an excitatory effect on gallbladder smooth muscle. [23], [26], [45], [87] Carbon monoxide may also play a role in gallbladder function. The enzyme that synthesizes carbon monoxide, heme oxygenase 2, is present in canine gallbladder nerves, and copper protoporphyrin IX, an inhibitor of heme oxygenase, inhibits nonadrenergic noncholinergic stimulus-induced relaxations. [88] In the bowel, ATP is a mediator of nerve-induced relaxations. However, it is unlikely that ATP contributes to gallbladder relaxation because the application of exogenous ATP leads to a gallbladder contraction. [89] Despite the results described, it is difficult to conceive how gallbladder neurons could provide an unambiguous inhibitory signal to gallbladder smooth muscle because all gallbladder neurons express ChAT and therefore are likely to be cholinergic and excitatory. [9] Activation of these neurons would lead to the release of acetylcholine and other compounds such as NO or VIP, thus providing the gallbladder smooth muscle with a mixed signal.

Increased sympathetic neural activity in the gallbladder. Another possibility is that gallbladder relaxation involves the release of norepinephrine from sympathetic nerves. Stimulation of the splanchnic nerves leads to a decrease in gallbladder tone, [90], [91] and [92] indicating that sympathetic activity contributes to interprandial gallbladder filling. It is likely that activation of sympathetic neural inputs can lead to decreased gallbladder tone through the direct effects on gallbladder smooth muscle and by inhibiting vagal input to gallbladder neurons. Behar and colleagues demonstrated that electric field stimulation–induced relaxation of cat gallbladder muscle strips can be completely blocked by the ß-adrenergic receptor antagonist, propranolol, whereas receptor antagonists or NOS inhibition have no effect. [80] These data are consistent with reports demonstrating that sympathetic nerve stimulation can result in gallbladder relaxation and indicate that, at least in larger species, sympathetic nerves may promote gallbladder filling through direct actions on gallbladder smooth muscle. In addition to relaxing gallbladder smooth muscle directly, sympathetic nerves may facilitate gallbladder filling by decreasing vagal tone in the organ. Yamasato and Nakayama [92] reported that stimulation of sympathetic nerves, at intensities lower than necessary to relax the gallbladder directly, results in a decrease in the excitatory response to vagal stimulation.

Furthermore, application of exogenous norepinephrine or release of norepinephrine from sympathetic nerves causes activation of a $_2$-adrenergic receptors located on vagal terminals in gallbladder ganglia and suppression of vagal synaptic input to gallbladder neurons. [20], [93] In smaller species, relaxations are not elicited in muscle strips in response to electric field stimulation, whereas in larger species, including human, relaxations can be activated even without precontraction of the tissue. The interspecies variation may be associated with differences in the distributions of sympathetic nerve fibers. In the guinea pig, TH-immunoreactive nerve fibers are restricted to the vascular and ganglionated plexuses. In larger species, TH-immunoreactive nerves are distributed throughout the muscularis as well as the ganglionated and vascular plexuses. Taken together, these data support the suggestion that sympathetic activity can cause a decrease in gallbladder tone by inhibiting the vagal input to gallbladder neurons and through direct effects on gallbladder smooth muscle. However, it is still unclear whether such a system is active primarily at times of physiological stress or whether it contributes to the normal accommodation of the gallbladder wall during interprandial filling. Indeed, in models of hemorrhagic shock, gallbladder contractility is significantly decreased. [94]

Humoral factors that relax gallbladder smooth muscle. Because pancreatic polypeptide relaxes gallbladder muscle strips, [95] this peptide has been implicated as a potential contributor to gallbladder filling. Furthermore, elevated pancreatic polypeptide levels have been implicated in the origin of clinically significant biliary stasis, as is seen in patients with diabetes mellitus. [96] Increases in pancreatic polypeptide levels have been measured in response to liquid test meals, intraduodenal feeding, and CCK infusion. [97], [98] However, there is no correlation between pancreatic polypeptide levels and gallbladder motor activity in healthy individuals, [99] indicating that pancreatic polypeptide probably does not contribute to interprandial gallbladder filling. Pancreatic polypeptide may play an indirect role in the control of gallbladder motility by modifying pancreatic secretion or intestinal motility, but this has not been resolved. Somatostatin is another compound that may contribute to gallbladder filling, because several lines of evidence indicate that somatostatin decreases gallbladder contractility. Long-term therapy with the somatostatin analog octreotide for diseases such as acromegaly [100], [101] and the existence of somatostatin-secreting tumors [102] both interfere with gallbladder contractility. Gallbladder hypomotility in celiac disease has been associated with elevated somatostatin levels, increased gallbladder fasting volume, and decreased CCK release. [103] In addition to its well-known inhibitory effect on the release of mucosal CCK, somatostatin can act locally to decrease gallbladder motility. Somatostatin decreases CCK- and carbachol-induced gallbladder contractions, gallbladder pressure, and the release of acetylcholine from gallbladder muscle strips. [104], [105] Somatostatin also inhibits CCK-induced gallbladder contractions in isolated cells. [106] In healthy subjects, somatostatin enhances gallbladder relaxation and reduces CCK secretion in the late postprandial phase. [107] Although these results infer an association between somatostatin and gallbladder volume under pharmacological or pathological conditions, there is no direct evidence to indicate that somatostatin plays a physiological role in gallbladder filling.

SPHINCTER OF ODDI

The SO oversees the flow of about 3 L of bile and pancreatic juices per day, and it is the busiest nonvascular intersection in the body. Because of its vast interspecies variation in morphology and function, it has been the subject of more controversy than any other structure in the gastrointestinal tract. The tasks of the SO are to regulate the flow of bile and pancreatic juices into the duodenum, to facilitate gallbladder filling, and to prevent the reflux of lumenal contents from the duodenum into the biliary tree. Although the precise mechanisms of SO function are not yet fully understood, research has demonstrated that changes in SO motility involve neurohormonal regulation of nontraditional neural circuits that orchestrate coordinated signals from SO ganglia to the surrounding circular smooth muscle.

Sphincter of Oddi Neurobiology

Most studies of SO ganglia have involved the guinea pig and the Australian possum. Therefore, most of the following information relates to ganglia in the SO region of these two species; of note, considering the aforementioned interspecies variation. [108]

Morphology of the Sphincter of Oddi Ganglia Ganglia in the guinea pig SO are similar in shape and density to the ganglia of the myenteric plexus of the duodenum. [109], [110] In fact, interganglionic nerve bundles can be traced between the myenteric plexus of the duodenum and that of the SO. [109] Most of these neurons have Dogiel type I morphology, consisting of a single long process and several short processes that measure less than the diameter of the cell body. The processes of SO neurons typically leave the ganglionated plexus to innervate the smooth muscle of the SO, and therefore most SO neurons are thought to be motor neurons.
Neurotransmitters of the Sphincter of Oddi Immunohistochemical studies have revealed the presence of numerous neuroactive compounds in SO nerves. [113], [114], [115], [116], [117], [118], [119], [120], [121], [122], [123], [124], [125], [126], [127], [128], [129] and [130] For example, among the species investigated, there is evidence of acetylcholine, NO, VIP, tachykinins, CGRP, NPY, OFQ, somatostatin, enkephalin (ENK), galanin (GAL), serotonin (5-hydroxytryptamine; 5-HT), peptide histidine isoleucine (PHI), gastrin-releasing peptide, and bombesin. The most thorough investigation of neurochemical content and distribution in the SO has been conducted in the guinea pig. Guinea pig SO ganglia consist of two major subpopulations of neurons: those that are immunoreactive for ChAT and those that are immunoreactive for NOS. [110], [111] The cholinergic (ChAT-positive) neurons, which are thought to be excitatory motor neurons, represent about two thirds of the population and are also immunoreactive for tachykinins and/or ENK. The nitrergic neurons, which are thought to be inhibitory motor neurons, are also immunoreactive for either VIP or NPY, but not for both. SO neurons have also been found to be immunoreactive for bombesin, [112] OFQ, [113] and 5-HT [114]; whether these compounds are expressed by cholinergic or nitrergic neurons is not yet known. Nerve fibers, thought to be extrinsic afferent axons, are immunoreactive for tachykinins and CGRP and are abundant in the ganglionated plexus of the guinea pig SO. [110] TH-immunoreactive, sympathetic postganglionic nerves also are abundant in guinea pig SO ganglia and nerve bundles. [115] Less is known about the colocalization of various compounds in other species. Considerable neurotransmitter diversity exists, and a nitrergic innervation of the SO has been reported in all species where this has been investigated. In the human SO, neural immunostaining has demonstrated NOS, VIP, PHI, NPY, CGRP, GAL, somatostatin, tachykinin, and ENK, and histochemical staining was positive for NADPH dehydrogenase. [11], [116], [117] and [118] Nerve fibers of the rhesus monkey SO are immunoreactive for ENK, NPY, and TH, and NADPH dehydrogenase staining also has been reported. [11], [119] In the pig, immunoreactivities for NOS, VIP, NPY, GAL, PHI, CGRP, tachykinin, and bombesin have been reported. [18], [117], [120] Feline SO nerves have been shown to be VIP and tachykinin immunoreactive. [23], [79], [121], [122] In studies of the Australian possum SO, ChAT, TK, GAL, gastrin-releasing peptide, and somatostatin immunoreactivities have been demonstrated, [123], [124], [125] and [126] and NADPH dehydrogenase staining suggests a nitrergic innervation. [127]
Electrophysiology Thus far, the electrophysiological properties of SO neurons have been investigated exclusively in the guinea pig. SO neurons can be classified as tonic or phasic cells on the basis of their active electrical properties. [109] Tonic cells are quite excitable, often exhibiting spontaneous activity and generating action potentials throughout a depolarizing current pulse. Phasic cells, in contrast, are relatively inexcitable, without spontaneous activity, and are capable of generating action potentials only at the onset of a depolarizing current pulse. An extensive study combining intracellular recording and dye injection, along with chemical coding, has revealed that there is no relationship between the electrical properties of SO neurons (tonic versus phasic) and their chemical coding patterns (cholinergic versus nitrergic). [128] Therefore, it is conceivable that the excitability of SO neurons shifts between excitable and inexcitable states, possibly in response to some humoral or neural signal. This type of neuroplasticity could be associated with changes in SO activity that occur during the feeding cycle. Stimulation of interganglionic fiber bundles in the SO elicits fast EPSPs in most neurons and slow EPSPs in about 25% of the cells, whereas inhibitory postsynaptic potentials are detected in about 10% of the neurons. [109], [115], [129] Fast EPSPs are mediated by nicotinic receptors, and fast excitatory synaptic input to SO neurons arises in part from the myenteric plexus of the duodenum. Neural connections between the duodenum and the SO are discussed in greater detail in the later discussion of SO postprandial activity. Slow EPSPs are mediated, at least in part, by the release of tachykinins from extrinsic sensory fibers, and they involve the activation of neurokinin 3 receptors. [129] Sympathetic postganglionic nerves, by releasing norepinephrine, initiate inhibitory postsynaptic potentials in SO neurons and inhibit fast EPSPs through actions on presynaptic nerve terminals. [115]

Sphincter of Oddi Smooth Muscle

Human and animal studies of the SO have established that the sphincter is not a simple and passive smooth muscle portion of the biliary system. Rather, it exhibits basal tonicity, spontaneous phasic contractions, and peristaltic activity that modulates bile flow into the duodenum. [130], [131] and [132] The human SO normally is characterized by prominent phasic contractions that are superimposed on a modest basal pressure. [133] In a landmark study, pressures were recorded from the human SO to determine the effect of neurohumoral mediators on SO function. [134] The basal SO pressure was 4 mm Hg higher than the common bile duct or pancreatic duct pressures. Pronounced phasic contractions superimposed on the basal SO pressure occurred at a frequency of 4.1 per minute. The phasic SO contractions were about 100 mm Hg in amplitude and 4.3 seconds in duration. These contractions were unique to the SO and did not exist in the duodenum, the common bile duct, or the pancreatic duct. The phasic contractions were altered by intravenous infusions of CCK, glucagon, and secretin, suggesting a neurohumoral component to the modulation of SO motility in the human.

In general, the interpretation of the actions of various compounds in the SO has been difficult because most compounds mediate their effects both through direct actions on the smooth muscle and indirect neural responses. For example, exogenously administered CCK has a direct contractile effect on SO smooth muscle as well as contractile and relaxant effects that are mediated by neurotransmitters release. The neurally mediated effects of CCK are species specific; both contraction and relaxation are detected in species whose sphincter acts as a pump, [135] and simple relaxation is detected in species such as human and cats where the sphincter

acts primarily as a resistor. [136], [137] and [138]

Electrical field stimulation of the SO evokes a twitchlike contraction followed by relaxation. [127], [139], [140] and [141] Muscarinic blockade in the presence of guanethidine alters this pattern to simple relaxation, [139], [140], [142] consistent with the existence of inhibitory nonadrenergic noncholinergic innervation. NO is the primary inhibitory neurotransmitter in the SO. [127], [140], [143], [144] Neurally released NO increases cAMP and cGMP levels in SO smooth muscle, [143], [145] and it induces the activation of K^+-ATP channels. [146] This pathway may be important in the physiological regulation of the bile duct pressure, because NOS blockers increase SO resistance to flow, [142] and NO donors inhibit SO contraction frequency and decrease basal pressure. [116]

Postprandial Decreases in Sphincter of Oddi Resistance

Release of CCK from enterochromaffin cells in the duodenum results in changes in SO function that promote the flow of bile in concert with increased gallbladder motility. In some species, including human, cat, dog, and Australian brush-tailed possum, this mainly involves a decrease in SO tone. [56], [137], [147], [148] and [149] In other species, such as guinea pig, rabbit, and North American opossum, CCK release leads to a pumping response that involves rhythmic fluctuations in SO tension. [47], [150], [151] and [152] In the opossum, bile flow appears to involve peristalsis through the particularly long SO region characteristic of this species. Regardless of the species or the type of response, it appears that the physiological effect of CCK is to decrease resistance and therefore to promote bile flow.

The effect of CCK on SO tone involves a neural mechanism, because the CCK-induced increase in bile flow through the SO is inhibited by muscarinic blockade with atropine or by complete neural blockade with tetrodotoxin. [56], [57], [150], [151] and [152] Furthermore, inhibition of VIPergic signals with VIP antiserum, or NO release by NOS inhibitors, suppresses CCK actions. [153], [154] These observations led to the theory that CCK alters SO tone through a hormonal effect on SO neurons. Indeed, the application of CCK to guinea pig SO neurons causes a prolonged depolarization and bursts of action potentials. [155] However, as for gallbladder smooth muscle, the concentration-dependent nature of this response is out of the physiological range. [50], [51] These data indicate that the neural mechanism by which CCK decreases SO resistance probably does not involve a direct action of hormonal CCK on SO neurons. One possibility is that CCK, released locally at high concentrations from enterochromaffin cells, has a paracrine effect on SO neurons, and another possibility is that CCK acts on duodenal neurons that provide regulatory input to SO ganglia.

Evidence is mounting in support of the concept that CCK released from duodenal enterochromaffin cells activates duodenal enteric neurons, and these neurons signal SO ganglia, initiating postprandial changes in SO function (Fig. 12-4). [156] In the guinea pig [157] and the Australian possum, [65] retrograde axonal tracing studies have confirmed the existence of a direct neural projection from the myenteric plexus of the duodenum to the SO. In the guinea pig, the neurons of this duodenum-SO projection are cholinergic, [157] and neurons in SO ganglia receive nicotinic excitatory synaptic input from the duodenal myenteric plexus. [158] Experiments involving electrical recordings from duodenal neurons that were retrogradely labeled from the SO have demonstrated that all of these neurons express CCK receptors, and many of these neurons are intrinsic primary afferent neurons. [157] Intrinsic primary afferent neurons in the intestines are known to send projections to the lamina propria, where they may be activated by local release of CCK from enterochromaffin cells. [159], [160] and [161] These data support the view that CCK released from duodenal enterochromaffin cells could initiate changes in SO resistance by way of a local neural circuit. However, the involvement of this neural circuit in postprandial changes in SO tone is not certain. Other potential functions for this circuit include the mediation of changes in SO tone that accompany interprandial bile flow during phase II of the MMC, as discussed in the next section. Alternatively, it may provide a means of increasing SO tone when lumenal pressure in the duodenum is elevated, thus inhibiting the flow of duodenal contents into the biliary tract.

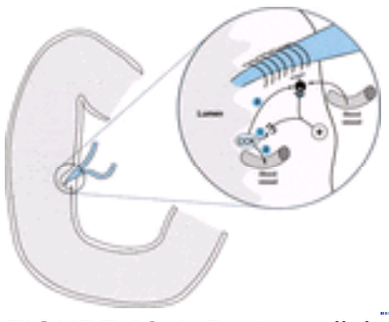

FIGURE 12-4. Postprandial relaxation of the sphincter of Oddi. Cholecystokinin (*CCK*) released from the duodenal mucosa activates inhibitory motor neurons in sphincter of Oddi (SO) ganglia, which, in turn, decrease the basal tone of the SO. CCK probably causes a relaxation of the SO by activating duodenal neurons that provide synaptic input to SO ganglia (1). Hormonal CCK may activate SO neurons, but these neurons are not sensitive to the concentrations of CCK that are found in the serum (2). Another possibility is that locally released CCK acts directly on SO neurons through a paracrine mechanism (3).

Interprandial Sphincter of Oddi Activity

Between meals, SO resistance is maintained by phasic contractions of the smooth muscle that are superimposed on a basal level of tonic pressure that is adequate to route hepatic bile to the gallbladder. During the fasting state, bile flowing toward the small bowel is diverted into the gallbladder by the resistance encountered at the SO—resistance maintained by basal and phasic contractions. Thus, intraductal pressures are increased, and bile flow is diverted to the region of lowest resistance, the cystic duct and gallbladder. High pressures in the biliary system also trigger relaxation and compliance of the gallbladder. Resistance at the SO and increased compliance of the gallbladder allow for the diversion of most of the bile away from the gut and into the gallbladder. There are, however, interprandial changes in SO activity that correspond to phase II of the MMC.

The interprandial SO activity associated with the MMC involves a decrease in SO resistance that accompanies increased gallbladder motor activity and bile flow. However, the MMC-related flow of bile does not depend on increased gallbladder motility, because dogs that have undergone cholecystectomy also exhibit cyclic bile flow that corresponds to the MMC. [162] Changes in SO activity that accompany the MMC have been documented in many species, including human. [67], [162], [163] As for the gallbladder, the mechanisms underling the MMC-related changes in SO activity are not entirely clear; however, it appears likely that the duodenum-SO neural circuitry is involved. After truncal vagotomy, MMC-related SO activity is normal in the opossum. Furthermore, relocation of the duodenal papilla in the opossum, which leaves the blood supply and extrinsic nerves intact but eliminates myoneural continuity between the duodenum and the SO, leads to a marked reduction in electrical activity of the SO during phases II and III of the MMC. [164] In addition, surgical relocation of the duodenal papilla reduces the responsiveness of the SO to motilin. [165] Together, these data support the concept that an intrinsic neural circuit mediates the interprandial SO motor response, but because some cyclic activity remains after translocation of the SO, it is likely that vagal reflex activity or humoral factors also are involved.

PATHOPHYSIOLOGY OF THE BILIARY TRACT

Mechanisms of Biliary Stasis in Cholesterol Disease

The gallbladders of patients with cholesterol gallstones often demonstrate abnormal motility. These patients have increased residual gallbladder volume interprandially, as well as decreased gallbladder emptying after a meal [166], [167] or in response to exogenous CCK administration. [166], [168] Furthermore, gallbladder muscle strips from patients with cholesterol gallstones demonstrate impaired contractile response to a variety of stimuli, as compared with gallbladder muscle strips from patients with pigment stones. [169], [170]

Decreased gallbladder smooth muscle contractility appears to be a critical link in the chain of events that leads to cholesterol gallstone formation and associated disease. Ground squirrels and prairie dogs that are fed a high-cholesterol diet undergo a progressive increase in cholesterol saturation in the bile, a decrease in gallbladder contractility, and, ultimately, cholesterol stone formation. [171], [172] If the decrease in gallbladder motility is offset by the administration of the prokinetic

agents cisapride or erythromycin, the formation of cholesterol crystals is prevented in cholesterol-fed ground squirrels. [173], [174] Conversely, if gallbladder stasis is pharmacologically induced by the administration of a CCK-A antagonist, gallstone formation occurs in animals that are fed either a normal diet or a cholesterol-enriched diet. [175]

These clinical and basic scientific data indicate that gallstone formation results from an elevation in cholesterol concentrations within the bile and an associated decrease in gallbladder contractility. The decrease in contractility is directly linked to cholesterol enrichment. Gallbladder smooth muscle that is exposed to cholesterol-rich liposomes exhibits diminished contractility similar to that observed in human gallbladders with cholesterol stones and in animals fed a high-cholesterol diet. [176] These results support the hypothesis that cholesterol itself can have a direct effect on gallbladder smooth muscle.

Two possible sites for the contractile defects caused by cholesterol are the cell membrane and the contractile machinery of gallbladder smooth muscle cells. However, the possibility that cholesterol disrupts the contractile machinery of these cells has been largely excluded. No difference is detected between the contractile responses of gallbladder muscle strips from control and cholesterol-fed ground squirrels that are exposed to the Ca^{2+} ionophore A-23187, which facilitates access of Ca^{2+} to the contractile proteins and causes contractions. [177] Therefore, it is likely that the primary smooth muscle defect in cholesterol gallstone disease does not reside in the contractile apparatus, but rather involves a disruption in the agonist-induced mobilization of Ca^{2+}. This may result from impaired ligand binding or altered ion channel activity.

To test whether cholesterol enrichment alters gallbladder smooth muscle ion channel activity, we exposed whole-mount preparations and isolated myocytes to cholesterol-loaded cyclodextrins. [178] Elevated membrane cholesterol results in a disruption of spontaneous action potentials, a decrease in the depolarization caused by the L-type Ca^{2+} channel opener Bay K 8644, and a suppression of voltage-activated Ca^{2+} currents. Conversely, K_V currents and K^+-ATP channels are not affected by cholesterol enrichment. In addition to altering Ca^{2+} channel activity, cholesterol enrichment of the membrane alters the activation of cytosolic membrane receptors. For example, the hyperpolarization to CGRP is suppressed after cholesterol enrichment, even though the channel that mediates this response, the K^+-ATP channel, is not altered by cholesterol. Furthermore, gallbladders from patients with cholesterol stones have impaired CCK receptor binding as compared with those from patients with pigment stones. [179] These data indicate that cholesterol enrichment results in selective deficits, rather than a nondiscriminatory disruption of membrane protein function, and that the biliary stasis associated with cholesterol stone formation is likely to involve altered Ca^{2+} channel and receptor function.

Acalculous Cholecystitis and Gallbladder Dysmotility

Conditions other than elevated biliary cholesterol also lead to decreased gallbladder contractility and cholecystitis, with or without stones. For example, biliary colic in the absence of gallstones is an increasingly prevalent complication among patients in the intensive care unit and in patients without predisposing illness. [180], [181] and [182] This condition, commonly referred to as *acute acalculous cholecystitis,* is often associated with trauma or extensive surgery. In a retrospective chart review over 53 months in a tertiary care center, 27 cases of acalculous cholecystitis were observed. Of these cases, 52% occurred in the intensive care unit and 63% occurred in patients recovering from unrelated surgical procedures, and acalculous cholecystitis accounted for 14% of all cases of acute cholecystitis. [183] The mortality rate for all patients with acalculous cholecystitis was 41%, illustrating the potential lethality of this clinical entity.

The development of acalculous biliary colic, like cholesterol-related gallbladder disease, appears to involve decreased gallbladder motility leading to an increase in the resting volume of the gallbladder and cholecystitis. [184], [185] and [186] The clinical complexity of a critically ill patient makes it difficult to sort among interrelated factors and to determine the origin of a complication such as acute acalculous cholecystitis. Factors implicated in the pathophysiology of acalculous cholecystitis include parenteral feeding, altered bile composition, ischemia, infection, and functional obstruction of gallbladder outflow. [184], [185] and [186] Decreased gallbladder motility may also result as a consequence of therapeutic agents that surgical patients and trauma victims receive to alleviate pain. A summary of the major findings in these areas follows.

Parenteral Feeding During total parenteral nutrition (TPN), biliary stasis and hypomotility have been well documented, contributing to the development of biliary dilation, sludge accumulation, and acute cholecystitis. In most patients, TPN induces gallbladder stasis but does not increase the biliary lithogenic index. [187] Interruption of the enterohepatic circulation and gallbladder stasis are part of the pathogenesis as a consequence of the lack of a natural stimulus for CCK release. However, during prolonged enteral nutrition, plasma CCK levels are significantly increased and gallbladder contractility is preserved, thus minimizing the risk of acute acalculous cholecystitis. [98] Furthermore, intravascular infusion of CCK in human or animal models ameliorates the hepatobiliary dysfunction caused by TPN. [187], [188] Increased circulating levels of CCK also can be induced by intermittent, rapid intravenous infusion of amino acids. [189], [190] However, continuous administration of amino acids for longer than 2 hours (as occurs in TPN) is accompanied by a decline in CCK release. [191] Taken together, these findings indicate that intravenous administration of amino acids or CCK may circumvent the biliary dysmotility associated with TPN, but therapy of this kind would probably be most beneficial if it is administered periodically, rather than continuously.

Opiate Analgesic Administration Another important factor leading to gallbladder stasis in the critically ill patient is opiate analgesic administration. In addition to acting as potent analgesics, opiate compounds are well known for their ability to alter gastrointestinal motility. Therefore, we have investigated the actions of opioid agonists at various sites along the neuromuscular axis of the gallbladder. [192] Although opioid agonists had no detectable effect on gallbladder neurons or gallbladder smooth muscle cells, selective agonists of the d, ?, and µ receptor subtypes caused a potent decrease in the amplitudes of fast EPSPs in gallbladder ganglia and neurogenic contractions of gallbladder smooth muscle. Furthermore, immunoreactivities for all three receptor subtypes were observed in the ganglionated plexus of the gallbladder. These data indicate that opioid agonists have a potent inhibitory effect on gallbladder motility by decreasing the release of acetylcholine and other excitatory neurotransmitters. The sites of action include both the vagal terminals in gallbladder ganglia and the neuromuscular terminals of gallbladder neurons. Because opioid agonists also increase SO resistance, bile flow is likely to be severely inhibited by opioid compounds, and continued administration of these substances may contribute to the onset of acalculous cholecystitis.

Inflammatory Mediators The cause-and-effect relationship between biliary stasis and acalculous cholecystitis is not well understood. However, the actions of several proinflammatory agents that are likely to be involved in acute cholecystitis have been investigated. These include histamine, prostaglandins (PGs), and reactive oxygen species. Histamine is a well-recognized inflammatory mediator that can be released from mast cells to cause vasodilation, increased vascular permeability, gastric secretion, and contraction of bronchiolar and gastrointestinal smooth muscle. The gallbladder wall is rich in histamine-containing mast cells, which are distributed in the mucosa and muscularis and serosa layers. [193] In gallbladder smooth muscle, both H_1 and H_2 histamine receptors are present, with H_1 receptors causing contraction and H_2 receptors mediating relaxation. [193] However, the net effect of histamine in gallbladder muscle strips is contraction. [193], [194] Furthermore, activation of H_1 receptors is associated with a depolarization of intact gallbladder smooth muscle, and activation of H_2 receptors is associated with a hyperpolarization that is mediated by the opening of K^+-ATP channels. [37] In contrast to the ganglia of the gut, where histamine has dramatic effects, no actions of histamine are detected in gallbladder ganglia. [37] Although there is no direct evidence for a role of histamine in gallbladder inflammation, it is possible that acute cholecystitis is associated with mast cell infiltration and degranulation. Thus, endogenously released histamine may exert an excitatory effect on gallbladder smooth muscle, resulting in a prokinetic protective effect. PGs, particularly PGE_2, have been shown to be intimately associated with cholecystitis. [195] Early studies of diseased human gallbladders have demonstrated that both the mucosa and the muscularis of the organ produce high levels of PGE_2. [196] Furthermore, a correlation between severity of inflammation and PGE_2 concentrations has been described. [197] PG release may be prolonged or augmented by bradykinin, because gallbladder distention and progressive inflammation stimulate local bradykinin formation, thereby stimulating PG release from the inflamed gallbladder. [198] The concept that PGs play a role in cholecystitis is supported by human and animal model studies that have shown that symptoms of acute cholecystitis are significantly reduced by the cyclooxygenase inhibitor indomethacin. [199] PGE_2 has been shown to cause a concentration-dependent contraction of the gallbladder, [200] as well as a significant reversal in net fluid movement from absorption to secretion, including an increase in mucin secretion. [201], [202] These actions are consistent with a cytoprotective role for PGE_2 in the gallbladder, at least in acute inflammation, acting by expelling gallbladder contents and preserving mucosal integrity. However, PGE_2 also has been shown to decrease gallbladder ganglionic output by hyperpolarizing gallbladder neurons, [203] thereby inhibiting neurogenic contractions of the gallbladder. Thus, the direct contractile effects of PG may contribute to the pain reported by patients suffering acute cholecystitis, because these contractions would not be accompanied by SO relaxation. Furthermore, the neural effects of PG in the gallbladder would lead to a decrease in postprandial contractions because, as previously described, excitatory neural activity is involved in normal postprandial gallbladder emptying. Therefore, in acute cholecystitis, PGs may contribute to gallbladder dysmotility by causing the gallbladder to contract when it normally would not and by preventing the gallbladder from contracting when it normally would.

Reactive Oxygen Species Another potential contributor to changes in gallbladder motility during the inflammatory state is the generation of reactive oxygen species, such as H_2O_2, $O_2^{-}\cdot$, $OH^{-}\cdot$, monochloramine, and peroxynitrite. As observed with histamine and PGs, contradictory effects of reactive oxygen species on gallbladder motility have been reported. For example, reactive oxygen species induce a reduction in agonist-induced gallbladder contraction, whereas others have a direct contractile effect. [86], [204] The exact importance of these inflammatory mediators in acute acalculous cholecystitis remains to be determined.

Sphincter of Oddi and Postcholecystectomy Syndrome

During the fasting state, bile flow into the small bowel is diverted into the gallbladder by the resistance at the SO because of the basal and phasic contractions previously described. After cholecystectomy, the pressure reservoir represented by the gallbladder is lost, and the ability of the biliary tree to accommodate hepatic bile is compromised. In most persons, bile manages to flow through the SO interprandially, without dramatic increases in pressure within the biliary tree. In a subset of the population, however, bile does not flow as readily through the SO, leading to increased intrabiliary pressure and postoperative pain. [205], [206] In some patients with postoperative biliary pain, biliary manometry has demonstrated abnormal pressure profiles in the SO with changes in amplitude, frequency, and direction of contractions. [206], [207] and [208] In an attempt to alleviate the pain experienced by these patients, endoscopic sphincterotomy, stent placement, and surgical sphincteroplasty have all been used, with variable results. [209]

OVERVIEW

Although the neuromuscular reflexes of the gallbladder and SO appear to be less complicated than those of the bowel, bile retention and bile flow require coordinated neuromuscular reflex interactions. Normal gallbladder filling requires relatively high basal pressure in the SO, in concert with an expansion of the gallbladder. This increase in gallbladder diameter requires the passive fibroelastic features of the gallbladder wall as well as myogenic and neurohormonal relaxation of gallbladder smooth muscle. Postprandial bile flow occurs in response to the release of CCK from the duodenal mucosa. To contract the gallbladder, CCK acts locally to activate vagal afferent nerves, and it also acts within gallbladder ganglia to increase acetylcholine release from vagal preganglionic nerve terminals. CCK release also leads to a decrease in the resistance of the SO by activating inhibitory motor neurons in SO ganglia, by way of a duodenal-SO neural circuit.

A common feature of biliary tract disease is abnormal motor activity. In cholesterol gallstone disease, decreased motility involves a disruption of Ca^{2+} channels in gallbladder smooth muscle and a decrement in the ability of agonists to bind to membrane receptors. Acalculous cholecystitis is also associated with biliary stasis, but the contributing factors have not been clearly resolved. Likely causes include decreased CCK release resulting from parenteral feeding, opioid inhibition of excitatory neurotransmission in the gallbladder when opiate analgesics are administered, and the effects of inflammatory mediators. As detailed in this chapter, much has been learned about how gallbladder nerves and smooth muscle operate. However, a clearer understanding of the precise subcellular mechanisms of gallbladder smooth muscle excitability and contractility will aid in the development of clinical strategies for preventing biliary stasis and subsequent biliary disease.

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CHAPTER 13

John Del Valle and Andrea Todisco

GASTRIC SECRETION

The stomach is a complex organ capable of secreting a great variety of products into the gastric lumen, vasculature, and interstitium. Although hydrochloric acid (HCl) is the primary gastric secretion, the stomach also secretes pepsinogen, mucus, bicarbonate (HCO_3^-), intrinsic factor (IF), prostaglandins, regulatory peptides, and other chemical messengers. This chapter discusses each of these, with particular emphasis on gastric acid secretion. Classic human and whole animal studies are integrated with more recent information gained at the cellular and molecular level, and this integrated model of acid secretion is placed within the fabric of current clinical practice.

Although it may appear to be axiomatic that the stomach secretes HCl, general acceptance of this fact occurred only after centuries of contention and controversy. Baron [1] attributed the 17th century Flemish physician Jean-Baptiste van Helmont with the first recognition that acid in gastric juice caused digestion. He thought it was derived from the spleen. About the same time, Sylvius, a professor of medicine in Leyden, proposed that gastric acid was a chemical digestive factor but thought it was secreted by the pancreas. For more than a century, there were numerous reports that acid was not present in gastric juices or, if acid was found, that it was a product of fermentation. Even the possibility of secretions arising from the stomach was in doubt, because many investigators asserted that gastric fluid was retained saliva.

The issue was settled by William Beaumont, [2] who demonstrated conclusively in 1825 and 1826 that Alexis St. Martin, a patient with a posttraumatic gastrocutaneous fistula, secreted gastric juice that could dissolve food inside and outside the stomach. Beaumont sent bottles of this gastric juice to three chemists, all of whom demonstrated it to be acidic.

Baron [1] gives credit to William Prout for the first qualitative and quantitative assessment of the composition of gastric juice in his article of 1823 entitled, “On the Nature of the Acid and Saline Matters Usually Existing in the Stomachs of Animals.” Prout identified free HCl in the gastric juice of rabbits, hares, horses, calves, dogs, and humans. In February 1824, two German workers, Tiedemann and Gmelin, unaware of Prout’s work, reported to the French Academy their proof that gastric acid was HCl. However, rival French workers reported that gastric acid was lactic acid, and this view held sway with many authorities throughout the 19th century despite the work of Prout, Tiedemann, and Gmelin.

Other workers who should be recognized are Schmidt, who showed in a series of experiments in 1852 that HCl was present in resting juice, and Dodds and Robertson, [3], [4] who, in 1930, demonstrated that the lactic acid in gastric juice was a product of fermentation.

The study of the regulation of gastric acid secretion also has a long history. William Beaumont reported that “fear and anger suppressed secretion by the stomach.” [2] Pavlov [5] showed that the sight and smell of food were powerful stimuli of gastric acid, and by introducing innervated and denervated gastric pouches, he established methods to study regulation of acid secretion that have prevailed for almost a century. One of his colleagues, Bechterew, [6] reported in his 1911 investigations into electrical stimulation of the frontal cortex as a stimulus of acid secretion that the effect was inhibited by vagotomy. In 1905, Edkins [7] demonstrated that extracts of canine antral mucosa stimulated acid secretion when they were injected intravenously into dogs. He thought this was caused by the presence in the mucosa of a chemical stimulator of acid secretion, which he called *gastrin.* Although similar observations were made by Lim in 1922, [8] studies of this mucosal factor were confounded by the presence in tissues under study of histamine, another powerful stimulus of acid secretion. [9] Gastrin remained a hypothetical substance until 1964 and 1965, when Gregory and colleagues, [10] having purified gastrin from hog antral mucosa, determined its structure and synthesized it in its biologically active form.

Inhibitory influences on acid secretion have been studied for many years. In 1929, Feng and colleagues [11] described a hormonally mediated inhibitor of acid secretion that was released after fat was placed in the duodenum. They called this putative hormone *enterogastrone.* Since then, there have been many attempts to identify this agent. Although it is beyond the scope of this chapter to highlight the discovery of each of the regulatory peptides implicated in acid secretion, special mention should be made of the isolation from ovine hypothalami of somatostatin, a powerful endocrine and exocrine inhibitor, by Brazeau in Guillemin’s laboratory in 1973. [12]

Until the late 1970s, considerable controversy surrounded the relative roles of gastrin, acetylcholine, and histamine in mediating acid secretion. Some researchers proposed that histamine was the final common pathway for acid stimulation. In a series of studies, Gillespie and Grossman [13] showed that these secretagogues potentiated the effects of each other, and these investigators proposed that there were individual receptors for these ligands on the parietal cell membrane. Soll and colleagues [14], [15] confirmed this hypothesis by demonstrating receptor characteristics for gastrin, acetylcholine, and histamine on isolated canine parietal cells. Somatostatin receptors have also been identified on these cells. [16]

ANATOMY OF GASTRIC MUCOSA

A detailed understanding of gastric mucosal structure provides insight into the functional events occurring during gastric secretion. The epithelial lining of the stomach lumen consists of thick, vascular folds, called rugae, invaginated with microscopic gastric pits. Each pit opens into four to five gastric glands. The epithelial cells lining

the gastric glands are highly specialized and different from the surface epithelial cells. Glands from the cardiac region of the stomach provide the transition from esophageal squamous epithelium to gastric columnar epithelium. They contain mucous and endocrine cells and comprise less than 5% of the gastric gland area. Most gastric glands (75%) occur in the oxyntic mucosa and are responsible for acid secretion (Fig. 13-1). They include parietal, chief, mucous neck, endocrine, and enterochromaffin cells. [17] The pyloric glands cover the gastric antrum and pylorus and contain gastrin cells (G cells), mucous cells, and other endocrine cells. Each of these cell types has evolved into a highly specialized secretory cell that contributes to gastric secretion.

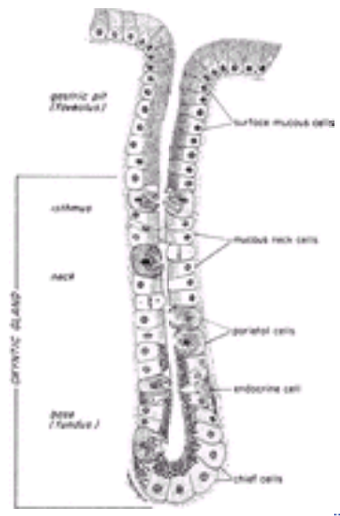

FIGURE 13-1. Oxyntic gastric gland. (Adapted from Ito S, Winchester RJ. The final structure of the gastric mucosa in the bat. J Cell Biol 1963;16:541.)

Cells

The parietal or oxyntic cell is the most distinctive cell of the gastric mucosa. It is usually found in the neck or isthmus of oxyntic glands bulging into the glandular lumen. The unstimulated parietal cell has prominent cytoplasmic tubulovesicles and an apical intracellular canaliculus lined with stubby microvilli. On stimulation, a dense meshwork of intracellular canaliculi rapidly forms while tubulovesicles disappear. [18], [19] The canaliculi contain a large number of elongated microvilli formed by extensive microfilaments that have a central cytoskeletal core of actin filaments stabilized by other proteins. It is across this apical canalicular surface that HCl is secreted. Acid secretion is an active transport process and requires significant amounts of energy. To provide this energy, parietal cells have numerous mitochondria, which account for 30% to 40% of total cellular volume. [20]

One prominent feature of parietal cells is their lack of the microvillous glycocalyx that is present on other cells in the gastric glands. Parietal cells are characterized by basolateral membrane folds that increase surface area for HCO_3^- exchange. The functional importance of these features is discussed in later sections of this chapter.

Chief cells are pepsinogen-secreting exocrine cells found in the base or fundus of oxyntic glands. Zymogen granules containing proenzymes are located in the apical cytoplasm and release their contents by exocytosis. The apical membrane has a few short microvilli covered by a thin coating of glycoprotein or glycocalyx. An abundant rough endoplasmic reticulum extends upward from the basal cytoplasm toward the apical granules. Functional characterization of chief cells has been aided by the ability to obtain relatively pure populations of chief cells.

Mucous neck cells are located in the isthmus or neck region of oxyntic glands (see Fig. 13-1). A transition zone from mucous neck cells to surface mucous cells appears near the junction of gastric glands and gastric pits. The mucous neck cells are the stem cell precursors for all of the gastric epithelial cells, including the surface mucous, parietal, chief, and endocrine cells. [21], [22]

Mucous neck cells differ in appearance from surface mucous cells. All mucous cells synthesize large amounts of mucin in prominent Golgi stacks, and these glycoproteins are transported by vesicles to large apical mucous granules. Mucous neck cells contain acidic glycoproteins, indicating sulfated forms, and surface mucous cells contain a neutral mucosubstance. [23], [24] Mucous granules are larger and often paranuclear in mucous neck cells compared with surface mucous cells. Mucous neck cells have abundant ribosomes and moderate amounts of rough endoplasmic reticulum. Their function as secretory cells and mucosal stem cells is different from the presumed function of surface mucous cells in mucosal defense. Surface mucous cells line the gastric pits and cover the entire lumenal surface of the stomach. They migrate up from the gastric pits and are replaced every 1 to 3 days. [25] They are thought to protect the stomach from injury by acid, pepsin, ingested materials, and pathogens by secreting mucus and HCO_3^- to form a protective gel. The apical portion of the surface mucous cell is packed with secretory granules. Short microvilli extend from the apical membrane and are covered by a glycocalyx. Secretion of granular mucus appears to occur by exocytosis, apical expulsion, and cell exfoliation. [26]

There are many different types of endocrine cells scattered throughout the gastric mucosa. Their secretory products have important endocrine and paracrine effects on acid secretion. Immunohistochemical techniques have enabled characterization of these cells based on their secretory granule contents. [20], [27] Gastric endocrine cells secrete gastrin, somatostatin, and enteroglucagon. Other morphologically distinct gastric endocrine cells may contain additional candidate hormones, but they await further characterization.

Gastric endocrine cells can be classified as open cells, which have apical membranes in contact with the glandular lumen, or closed cells, which are located near the epithelial basement membrane and do not border on the lumen of the gland. The prototypical open endocrine cell is the G cell. The basilar portion of the cell is packed with secretory granules, [28] from which gastrin is released by basilar exocytosis or emiocytosis, [29] consistent with the rapid postprandial appearance of the hormone in the bloodstream. The apical portion of the cell narrows until only a small microvillous border opens on the glandular lumen. The apical membrane may contain lumenal receptors that can detect amino acids or their amine derivatives, which are thought to stimulate G cells during feeding. [30], [31] The model of a closed gastric endocrine cell is the fundic somatostatin cell (D cell). By immunohistochemical staining, these cells are revealed to have long, slender processes that terminate on or near parietal and chief cells. [32] These processes presumably mediate the paracrine effect of somatostatin.

The primary cell containing mucosal histamine in the dog stomach is the mast cell. [33] Histamine-containing mast cells also occur in the human stomach. Mast cells from canine fundic mucosa have been enriched by elutriation and shown to contain characteristic dense granules that stain metachromatically. [34] Some species, including rats and humans, have histamine in endocrine-like cells that contain large granules and have the characteristic appearance of enterochromaffin-like (ECL) cells. [35] The relative proportion of these two histamine cell types in humans is unknown. Although in situ morphologic studies have not been definitive, it appears that these cells exist in the lamina propria in proximity to the glandular cells. The functional characteristics of these cells are discussed in later sections of this chapter.

The origin of gastric epithelial cells and the molecular events important in their differentiation are becoming increasingly clear. Electron microscopic examination and the measurement of DNA synthesis in gastric cells indicate that the isthmus of the gastric gland (see Fig. 13-1) constitutes the proliferative zone for the stomach. [36], [37] and [38] Within this region are three populations of cells that do not contain granules and are characterized on the basis of their Golgi apparatus. Parietal cells originate from precursor cells (preparietal cells), which mature as they migrate toward the neck and base of the gland. Karam and colleagues [39] demonstrated that there is functional heterogeneity of parietal cells along the pit-gland axis. Using quantitative morphometry, these investigators observed that parietal cells in the isthmus and neck of the rabbit gastric gland responded to secretagogues in an appropriate manner, whereas those in the base showed minimal morphologic change after exposure to different stimuli. The factors responsible for decreased function of senescent parietal cells are unclear.

Molecular approaches with various transgenic mouse models have been used to unravel the molecular signals directing differentiation of the distinct gastric cell lineages. Li and colleagues [40] capitalized on the unique expression of the ß subunit of H^+,K^+-ATPase in parietal cells to target expression of the simian virus 40 T

antigen (SV40TAg) to gastric glands. These investigators successfully amplified the population of preparietal cells in this animal model and noted a block in cell differentiation accompanied by hypochlorhydria and iron deficiency. In addition, SV40TAg-induced preparietal cell proliferation was accompanied by apoptosis, which appeared to occur in a p53-independent manner. The block in the development of mature parietal cells in this model was accompanied by the absence of zymogenic cells. This key observation supported the novel concept that potential signals are generated by mature parietal cells, which, in turn, are important in regulating the differentiation of other cell lineages within the stomach. This hypothesis was further confirmed by analyzing gastric glands of transgenic mice in which parietal cells were ablated by diphtheria toxin. [41] In addition to inhibited development of mature parietal cells, these investigators observed that differentiation of zymogenic cells was blocked, with accumulation of pre-neck cells and amplification of pit cells. Together, these studies are consistent with the hypothesis that interactions among different cell lineages are important in regulating cell differentiation in the stomach.

Studies of genetically engineered mice in which the cholecystokinin B (CCK-B) receptor or the peptide gastrin have been ablated demonstrate the development of gastric mucosal atrophy accompanied by a reduced number of parietal and ECL cells. These findings confirm the importance of gastrin in gastric mucosal cell development and differentiation. [42], [43] Similarly, ablation of the genes encoding the histamine H_2 receptor (H2R) has revealed the presence of a series of morphologic abnormalities of the gastric mucosa. [44] In particular, the stomach of H2R knock-out mice exhibits a markedly hypertrophic gastric mucosa with enlarged folds and elevated gastrin levels. Immunohistochemical analysis of the gastric mucosa of these mice demonstrates increased numbers of parietal and ECL cells. The parietal cells of these animals, however, appear to be smaller than in the wild-type mice, exhibiting enlarged secretory canaliculi with a lower density of microvilli and few typical tubulovesicles in the narrow cytoplasm. As a consequence of these morphologic changes, the H2R-deficient mice demonstrate abnormalities in secretagogue-stimulated gastric acid secretion. In particular, both gastrin and histamine fail to stimulate gastric acid secretion, whereas the cholinergic agonist carbachol retains the ability to induce gastric acid secretion to a level identical to that observed in the wild-type mice.

Interestingly, the enlargement of the secretory canaliculi and reduced number of microvilli and tubulovesicles also occur after the selective deletion of either the H^+,K^+-ATPase a [45] or ß [46] subunits in mice. Moreover, targeted deletion of the Na^+/H^+ exchanger isoform 2 (NHE2) gene appears to lead to a significant decrease in the number of parietal and zymogenic cells and to a marked increase in parietal cell degeneration, suggesting that NHE2 plays an important role in parietal cell viability. [47] The exact mechanisms responsible for these observations have not yet been characterized.

Significant alterations in gastric mucosal morphology and gastric acid secretion were also detected in mice deficient in the trefoil factor 2 (TFF2)/spasmolytic polypeptide (SP) gene. [48] TFF2/SP is a small polypeptide secreted by the gastric mucous neck cells. [48] Expression of TFF2/SP appears to be increased at sites of gastrointestinal ulceration and inflammation, suggesting that it may play an important role in the stimulation of reparative and cytoprotective mechanisms. [48] TFF2/SP-deficient mice appear to have an increased number of activated parietal cells, gastric acid secretion is increased, and gastrin is undetectable. In addition, these animals exhibit decreased gastric mucosal thickness and proliferation rates, and increased susceptibility to indomethacin-induced gastric ulcerations. [48] Taken together, these observations suggest that TFF2/SP promotes mucosal healing through the stimulation of cell proliferation and the inhibition of gastric acid secretion. The precise mechanisms responsible for TFF2/SP-mediated inhibition of gastric acid secretion remain to be elucidated.

Innervation

As is the case for the entire gastrointestinal tract, the stomach is innervated by the central and enteric nervous systems. This innervation mediates secretion and motor activity through efferent fibers and detects chemical or mechanical stimuli by means of afferent fibers. A full description of the enteric nervous system's structure and function is found in Chapter 2.

Central efferents are carried by the parasympathetic vagal branches and the sympathetic greater splanchnic nerve. [49] Vagal preganglionic fibers arise from the dorsal motor nucleus of the brain stem and descend to the thorax, where they branch to form an esophageal plexus. These fibers merge into anterior and posterior vagal trunks just before they cross the diaphragmatic esophageal hiatus. The anterior trunk gives off a gastric branch that runs along the lesser curvature and supplies the anterior surface of the stomach down to the pylorus. Pyloric branches innervate the antrum, pylorus, and proximal duodenum. The posterior vagal trunk provides a similar gastric branch that supplies the posterior surface of the stomach. The preganglionic fibers synapse in intramural ganglionated plexuses. Postganglionic fibers may be the final motor neurons or interneurons communicating with the enteric nervous system. Parasympathetic stimulation usually causes an increase in secretory and motor activity. Gastric sympathetic efferents emerge from spinal cord segments of T5 through T9. These preganglionic fibers run along the greater splanchnic nerve and synapse in the celiac ganglion. Postganglionic axons extend to the stomach and enter it in association with blood vessels. These sympathetic fibers provide rich innervation to the gastric blood vessels and the myenteric and submucous plexuses. In general, sympathetic stimulation causes a counterbalancing inhibition of gastric secretory and motor activity and constriction of blood vessels.

Afferent sensory neurons are exemplified by mucosal chemoreceptors and myenteric mechanoreceptors. These receptors communicate with the central and the enteric nervous systems. Afferent nerve processes account for about 80% of vagal fibers and 20% of the greater splanchnic nerve's fibers. [50], [51] The incoming information carried by these nerves is processed in central sensory nuclei and initiates neurally mediated gastric reflexes. Sensory neurons in enteric ganglia also initiate local reflexes such as gastric peristalsis. Cross talk between central and enteric fibers occurs continually in the enteric plexus as it modulates the effects of motor neurons.

Vasculature

Gastric mucosal blood flow maintains epithelial integrity and is an essential component of mucosal defense. The stomach receives its blood supply from the celiac axis through six major arteries. The right and left gastric arteries supply the lesser curvature and extend over the anterior and posterior surfaces of the stomach. In a similar fashion, the left and right gastroepiploic arteries supply the greater curvature. Short gastric arteries from the splenic artery perfuse the upper stomach, and the gastroduodenal artery serves the pyloroantral region. These arteries provide large arterioles that pierce the gastric muscle wall.

The microvasculature of the stomach is illustrated in Figure 13-2. The entering arterioles provide smaller arterioles that extend to the submucosal plexus and to the muscle layers. [52] A capillary layer in the muscle drains into the venous collecting system. The afferent arterioles supplying the submucosal plexus are innervated by sympathetic fibers coursing with the entering blood vessels. The submucosal arteriolar plexus does not communicate with the venous plexus directly but rather provides mucosal arterioles to the base of the gastric glands. The mucosal arterioles branch into capillaries that ascend perpendicularly between the glands to the epithelial surface. These ascending capillaries interconnect horizontally, forming a lattice around the gastric glands. Ascending capillaries receive HCO_3^- secreted from the basolateral surface of stimulated parietal cells and carry it to the surface epithelium. This phenomenon is called the *alkaline tide.* At the surface epithelium, the HCO_3^- can assist in buffering any H^+ ion back-diffusion. At the mucosal surface, the fenestrated capillaries empty into venules that drain into the submucosal venous plexus and efferent veins that follow the course of the primary arterial branches. [53] This anatomic arrangement allows muscle layer blood flow to be in parallel with mucosal flow while submucosal flow is in series with the mucosa. This type of microvascular system allows selective decreases in mucosal blood flow while muscular blood flow is maintained.

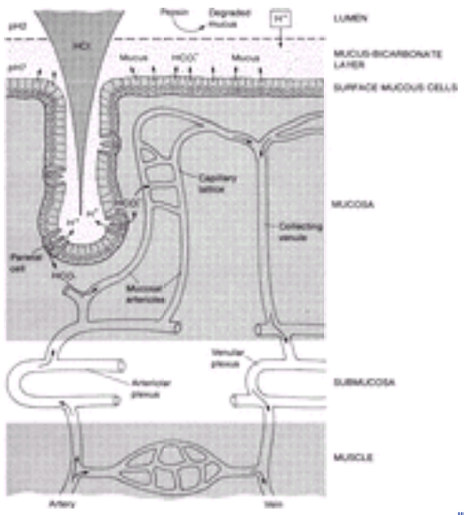

FIGURE 13-2. Gastric microvasculature. (Adapted from ref. [52] and Koelz HR, Fimmel CJ. Garner A, et al. The stomach and duodenum. In: Kern F, Blum AL, eds. The gastroenterology annual, 3rd ed. New York: Elsevier, 1986:28.)

Mucosal blood flow accounts for 70% to 80% of total gastric blood flow in basal and stimulated states. [54], [55] Previous studies using aminopyrine clearance techniques suggest that stimulation of acid secretion increases mucosal blood flow, but interpretation of these results must be tempered by the knowledge that acid secretion affects aminopyrine clearance and may account for the apparent increase in mucosal blood flow. [56] Acid secretagogues appear to have no effect on mucosal blood flow using more recently developed indicator-dilution techniques. When gastric perfusion pressures are adequate, gastric acid secretion varies independently of blood flow. [57] These conclusions have challenged the long-held hypothesis that secretagogue stimulation induces a parallel increase in acid secretion and mucosal blood flow. [58]

The innervation of the gastric microvasculature partially reveals the mechanisms regulating mucosal blood flow. The submucosal arterioles appear to be innervated by sympathetic nerve fibers. [59] When stimulated, these fibers constrict the arterioles and decrease mucosal blood flow temporarily. [60], [61] After 3 to 4 minutes of prolonged sympathetic stimulation, flow increases. [62] This response has been described as autoregulatory escape from adrenergic vasoconstrictor influence. [63] Anatomic details support these physiological findings and suggest a regulatory function at the level of the submucosal arterioles. [59]

REGULATION OF ACID SECRETION

The regulation of acid secretion can be subdivided into the supracellular influences that have been the focus of classic physiology since Beaumont and the cellular mechanisms that have been elucidated more recently. This section concentrates on supracellular influences and reviews the in vivo studies of acid secretion.

Integrated Control

The integrated mechanisms that control acid secretion can be viewed as an arrangement of regulatory strata. These include neural control in two forms: long reflex or cephalovagal arcs and local intragastric reflex arcs. A second tier of control is exerted by humoral substances acting in an endocrine fashion, such as gastrin, or in a paracrine manner, such as histamine. Somatostatin may function as an endocrine and paracrine factor. The role of many such peptides remains uncertain. The direct influence of chemical factors is an additional tier of regulation, typified by the stimulatory action of amino acids and amines on gastrin release and the inhibitory effect of gastric acid on gastrin release.

These strata are not distinct but rather are closely intertwined; when discussing the phases of postprandial acid secretion, Grossman [64] asserted that the individual contributions made by the various phases could not be differentiated. Nevertheless, classic physiology has attempted to isolate and study factors that influence acid secretion. The results indicate that there are three principal stimuli of parietal cell acid secretion: histamine, acetylcholine, and gastrin. The principal inhibitory secretagogue appears to be somatostatin, although other chemical modulators, including peptides, growth factors, and prostaglandins, may also have roles.

When reviewing these in vivo studies, several confounding facts should not be forgotten. First, acute responses should be differentiated from chronic adaptive changes. For example, the effect on gastrin and somatostatin cell number during acute achlorhydria differs from the changes seen in chronic achlorhydria. [65] By analogy, data in studies using vagotomy may signify the chronic adaptive response to achlorhydria rather than the effect of withdrawal of vagal innervation alone. Second, acid secretagogues potentiate the response to one another. This central point, which is crucial to the understanding of studies of supracellular and cellular regulation, was first appreciated by Grossman. [64] He defined a potentiated response as one in which the effect of two agents in combination is greater than the sum of their separate effects when administered alone. Gillespie and Grossman [13] demonstrated in dogs with Heidenhain pouches that the acid secretory response to a combination of Urecholine (bethanechol chloride) and gastrin or to Urecholine and histamine greatly exceeded the maximal response to gastrin, histamine, or Urecholine alone (Fig. 13-3). These investigators subsequently demonstrated the corollary effect using antagonists of acid secretion. Metiamide, an H_2 receptor antagonist, inhibited acid secretion stimulated by histamine, pentagastrin, 2-deoxyglucose (which activates central vagal stimulation), and food. Atropine sulfate also inhibited the response to all of these stimulants except histamine. [66] With great perception, Grossman deduced that there were receptors for histamine, gastrin, and acetylcholine on the parietal cell that interacted with one another. Because of these interactions, it is impossible to predict the relative importance of each secretagogue on the basis of individual in vivo stimulation or inhibition studies. The phenomenon of potentiation is the basis for the efficacy of many of the acid-reducing therapies for peptic ulceration, especially the use of H_2 antagonists. A third problem is that stimulatory and inhibitory influences are active simultaneously. This is true in the basal interprandial and the postprandial state.

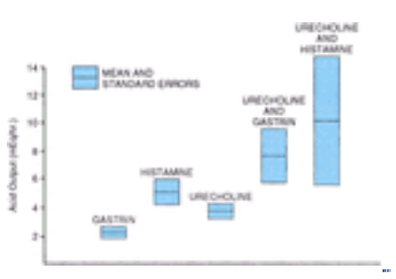

FIGURE 13-3. Maximal acid responses to gastrin extract, histamine, and Urecholine, alone and in combination in dogs with Heidenhain pouches. (From ref. [13].)

Methods for Measuring Acid Secretion

Animal studies have used dogs, rats, rabbits, and mice. There are great species differences in basal secretion and responsiveness to secretagogues. [67] Classic studies of dogs have used vascularly perfused isolated pouches of gastric mucosa. The fundic pouches with intact vagal innervation are called *Pavlov pouches,* and vagotomized fundic pouches are called *Heidenhain pouches.* It is assumed that secretion from a vagally innervated fundic pouch is an accurate index of secretion from the main stomach. [64] These gastric pouches have been used to define many aspects of regulation of gastric acid secretion, including cephalic influences, the role of gastrin, long cephalovagal and local reflex neural arcs, and the inhibitory feedback control of acid secretion by intralumenal acid.

Aspiration of gastric juice is the simplest and most widely used method of estimating acid secretion in humans. To perform these tests, a fine-bore nasogastric tube is inserted under fluoroscopic control into the most dependent part of the stomach under fluoroscopic control of a fasted volunteer, and several measurements are made: basal acid output (BAO), which estimates resting secretion, and maximal acid output (MAO) or peak acid output (PAO), which estimates the acid secretory

response to an exogenous secretagogue. BAO is measured by aspirating the gastric contents for four consecutive 15-minute periods. The H^+ ion concentration of the aspirate is estimated by titration with a basic solution of known concentration. The BAO is expressed as mEq H^+ per hour and is the sum of the measured acid output in four unstimulated test periods. The expected range for BAO in healthy adults is 0 to 11 mEq H^+ per hour (Fig. 13-4). [68] The measurement of BAO is usually combined with the measurement of MAO or PAO. In this test, acid output is stimulated by a supraphysiological dose of an exogenous secretagogue. This is usually pentagastrin, which may be administered by a subcutaneous or intramuscular injection (6 µg/kg) or by continuous intravenous infusion (6 µg/kg/h). Other secretagogues that have been used are histamine or a histamine analog, betazole. [17] The PAO is calculated by multiplying by 2 the sum of the two highest outputs recorded in the four test periods. The MAO is the sum of acid output of four consecutive 15-minute collection periods. The expected range for PAO in healthy adults is 10 to 63 mEq per hour (see Fig. 13-4). [68]

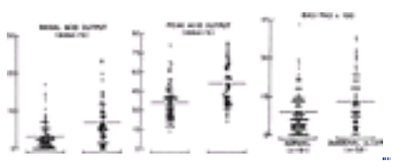

FIGURE 13-4. Basal acid output (*BAO*), peak acid output (*PAO*), and BAO/PAO ratio in 91 healthy adults and in 58 patients with duodenal ulcers. (From ref. [68] .)

MAO and PAO are a reflection of the total number of parietal cells, also called the *parietal cell mass,* [69] which is influenced by gender, body weight, lean body mass, and age. [70] MAO and PAO are lower in women than men because of a smaller parietal cell mass and a lower sensitivity of that parietal cell mass to exogenous secretagogues. [71] Stimulated acid output in children is comparable to that in adults when expressed as a function of body weight, but acid secretion declines in the elderly. [72] , [73]

BAO, MAO, and PAO underrepresent actual basal and stimulated acid secretion, because these methods do not account for acid lost through the pylorus, acid neutralization by gastric HCO_3^- and refluxed duodenal juice, and acid losses resulting from back-diffusion of H^+ through the gastric mucosa. Nonetheless, these measurements have proved useful in defining the pathophysiology of peptic ulcer disease [68] and in diagnosing Zollinger-Ellison syndrome.

Another method of estimating acid secretory ability was introduced by Fordtran and Walsh using continuous in vivo intragastric titration. [74] By this means, the acid secretory response to the physiological stimulation of ingested food is estimated. Continuous intragastric titration requires placement of a double-lumen tube into the most dependent part of the stomach, usually under fluoroscopic visualization. One lumen allows frequent sampling of small volumes (2–3 mL) of gastric contents. The pH of the sample is immediately measured, and the gastric juice is returned to the stomach. The port of the second tube is positioned 10 cm proximal to the sampling port and is used to infuse $NaHCO_3$. Throughout the study, the gastric pH is maintained at an arbitrary value, usually pH 5.5, by infusing $NaHCO_3$. A homogenized meal buffered to pH 5.5 is eaten. The amount of $NaHCO_3$ necessary to maintain the pH of the gastric juice at pH 5.5 is a measure of the postprandial acid secretory response. Because this is a cumbersome procedure, it has not gained widespread use outside specific research investigations. However, continuous intragastric titration has proven to be a useful method of measuring postprandial acid secretory responses. [75] , [76]

A different approach to studying continuous acid responses is to measure intragastric pH by using an indwelling probe or radiotelemetric capsule. [77] , [78] This does not estimate total H^+ concentration because it does not account for the volume of gastric juice and buffering capacity. Further studies with these techniques are required before their specific utility in humans can be determined.

Basal or Interprandial Acid Secretion

Basal unstimulated acid secretion exhibits a circadian variation, with the highest secretion at night and the lowest in early morning. [79] This variation is not matched by changes in circulating serum gastrin levels. In humans, vagotomy greatly reduces or abolishes interprandial acid secretion in patients with duodenal ulcer. [69] , [76] , [80] The caveats expressed about the influence of potentiation and chronic adaptive changes on studies of the effect of vagotomy on gastric endocrine and exocrine responses are pertinent when interpreting these data, although vagotomy does not affect parietal cell number. [81] Twice-daily administration of an H_2-antagonist to patients with a duodenal ulcer reduces interprandial acid secretion, although it does not abolish it. [76] This suggests that H_2 receptor activation is also capable of influencing BAO. It appears likely that basal interprandial acid secretion is the result of combined cholinergic and histaminergic stimulation.

The cholinergic regulation of basal gastric acid secretion is complex. In dogs with gastric pouches, denervation of the antral pouch markedly reduces resting output of acid from the main innervated stomach and from the denervated fundic pouch without changing plasma gastrin levels. [82] This suggests there is a local interneuronal reflex arc innervated by the vagus and independent of plasma gastrin that carries acid stimulatory signals from the antrum to the fundus. These data are mirrored in humans; the small amount of basal acid secretion found in some patients who have undergone vagotomy can be abolished by antrectomy plus vagotomy. [80]

Stimulation of Acid Secretion

The physiological stimulus for acid secretion is food. Traditionally, food-stimulated acid response has been described in three phases: cephalic, gastric, and intestinal. These phases refer to the sites of origin of the stimuli and do not imply mechanisms by which acid secretion is stimulated or inhibited. These phases occur concurrently, not consecutively. The acid secretory response at any instant represents the sum of all the stimulatory and inhibitory influences.

Cephalic Phase The nervous system's influence on gastric acid secretion was first recognized by William Beaumont and Pavlov. In 1928, Farrell [83] showed that the vagus was the sole gastrocephalic neural link involved in gastric secretion, and this observation remains valid. The cephalic phase contributes in some degree to total postprandial acid but was estimated by Richardson and colleagues [84] to be as much as 50%. Feldman and Richardson [85] studied the relative contribution of thought, sight, smell, and taste of food to the cephalic phase of gastric acid secretion in humans (Fig. 13-5). Merely discussing appetizing food for 30 minutes without sight, smell, or taste produced an average of 66% of the total cephalic response estimated by the time-honored method of modified sham feeding. Serum gastrin levels also significantly increased. The sight of food or smell of food alone, although still producing significant acid secretion and gastrin release, was a considerably less potent stimulus than conversation. These data show by subtraction that taste is also an important component of the cephalic phase.

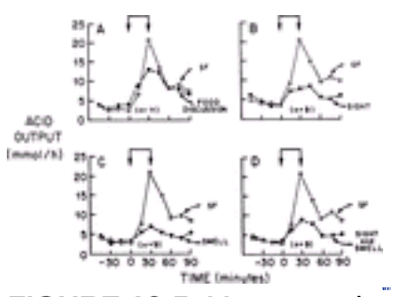

FIGURE 13-5. Mean gastric output in the same persons in response to sham feeding (*SF*), compared with (**A**) discussion of favorite foods, (**B**) sight of appetizing food, (**C**) smell of appetizing food, and (**D**) a combination of sight and smell. (From ref. [85] .)

The mechanisms by which the senses stimulate acid secretion are less certain. Bechterew [6] induced acid secretion in dogs by electrically stimulating the frontal cortex. This effect was lost in vagotomized animals. Two other forms of study have been used: electrical stimulation of various sites in the brain and injection of putative neuromodulatory substances into the brain. The putative ligands include peptides; classic neurotransmitters such as acetylcholine, ?-aminobutyric acid agonists, and catecholamines, and prostaglandins. There are many interrelated mechanisms by which cephalovagal input mediates the cephalic phase of stimulated

acid secretion. Vagal innervation acts directly on parietal cells. This was deduced from studies of dogs with antral and fundic gastric pouches. Denervation of the antral pouch reduced or abolished the serum gastrin response to sham feeding but did not abolish acid secretion by the vagally innervated fundic pouch. [82] Corroborative evidence for this mechanism is afforded by the demonstration of acetylcholine receptors on isolated canine parietal cells. A second pathway in the cephalic phase of gastric acid secretion is the release of circulating gastrin from the gastric antrum. The thought, sight, or smell of food and modified sham feeding increase serum gastrin levels. [80] Isolated canine gastrin cells possess acetylcholine receptors. The plasma gastrin response to sham feeding in humans is regulated by intragastric pH and is not observable when gastric pH is maintained at pH 2.5. [86] This effect also is atropine sensitive. However, vagal control of postprandial gastrin release is complex. Low doses of atropine, a muscarinic antagonist, enhance rather than reduce the gastrin response to sham feeding, insulin-induced hypoglycemia, or feeding. [87], [88], [89], [90] and [91] Similarly, parietal cell vagotomy, although markedly reducing acid secretion, enhances gastrin release in response to sham feeding, [87], [92] insulin-induced hypoglycemia, [93] or intragastric nutrient infusion. These effects are independent of changes in intragastric pH. [89] This result suggests that the vagus carries cholinergic fibers that, in the absence of atropine, mediate directly or indirectly an inhibitory control of gastrin release. Studies of dogs with Heidenhain pouches and gastric fistulae demonstrate that truncal vagal denervation reduces gastrin release but elevates acid secretion by the pouch. [94] These data can be interpreted to suggest cholinergic release of a substance that can inhibit acid secretion independently of its effects on gastrin release. Although the nature of this substance has yet to be elucidated, somatostatin is one candidate. [91] However, Feldman and colleagues [88] were unable to demonstrate a rise in the level of plasma somatostatin in humans during sham feeding, and the role of somatostatin in regulating the cephalovagal control of gastric acid secretion whether directly on the parietal cell or by inhibition of gastrin release remains uncertain. To define more precisely the role of gastrin in sham feeding–mediated gastric acid secretion in dogs, Kovacs and colleagues [95] developed a monoclonal antigastrin antibody capable of immunoneutralizing circulating gastrin in vivo. By means of this antibody, these investigators demonstrated that immunoneutralization of gastrin leads to a significant inhibition of sham feeding–stimulated gastric acid output, suggesting that gastrin released by central vagal stimulation is an important mediator of sham feeding–stimulated gastric acid secretion in dogs. The components of the central nervous system involved in modulating gastric secretion include the dorsal motor nucleus of the vagus (DMNV), the nucleus tractus solitarius (NTS), and the hypothalamus. The DMNV supplies stimulatory efferent fibers to the stomach through the vagus nerve. [96], [97] Stimulation of the DMNV results in activation of gastric secretion, [98], [99] whereas ablation of this nucleus abolishes the secretory process. [100] It appears that the role of the DMNV is not to initiate secretion, [101] but instead to integrate central input from the hypothalamus and visceral input from the NTS. Regions of the hypothalamus important for regulating gastric secretion include the ventromedial hypothalamus, which appears to provide tonic inhibition, [102], [103] the lateral hypothalamus, and the adjacent medial forebrain bundle, which together mediate secretion in response to hypoglycemia. [104], [105] and [106] Visceral afferents and taste fibers deliver input into the NTS, demonstrating its potential involvement in stimulating gastric secretion in response to taste. The NTS is also thought to mediate stimulation of acid secretion in response to hypoglycemia. [107] Sympathetic and vagal afferents relay sensory information (mechanical, chemical and thermal) from the stomach to the central nervous system. [108] Peptidergic factors also play an important role in modulating central nervous system–regulated gastric secretion. Direct injection of peptides into the brain has led to the discovery of factors that either stimulate or inhibit gastric secretion. [109] The peptides observed to either activate or inhibit gastric acid secretion by central injection are summarized in Table 13-1. This represents a partial summary of these peptides, the result of the numerous elegant studies examining this important subject. [109], [110], [111], [112], [113], [114], [115], [116], [117], [118], [119] and [120]

INHIBITORS	STIMULANTS
CRF	TRH
β-endorphin	Somatostatin
Bombesin	NPY
Neurotensin	Galanin
Calcitonin	PYY
CGRP	Ghrelin
Interleukin 1	Orexin A

Note: CRF, corticotropin-releasing factor; CGRP, calcitonin gene–related peptide; TRH, thyrotropin-releasing hormone; NPY, neuropeptide Y; PYY, peptide YY.

TABLE 13-1 Peptides that Affect Gastric Acid Secretion After Injection Into the Central Nervous System

Gastric Phase When food enters the stomach, it initiates the gastric phase of acid secretion. This is usually divided into two components: a physical component caused by distention of the stomach and a chemical component in which chemical effectors interact with gastric cells. The acid secretory response to distention results from presumed stretch receptors in the gastric tissue. This response is the stereotypical long vasovagal reflex arc. Distention of the gastric fundus and corpus in humans stimulates acid secretion. The effect is almost abolished by proximal gastric vagotomy and at least in part is independent of changes in serum gastrin levels. [121], [122] and [123] Distention-induced acid secretion is a complex phenomenon in which antral and fundic responses can be viewed separately. Antral distention produces gastrin release in dogs [124] and humans. [125] Debas and associates [124] called this the *pylorooxyntic reflex.* In dogs with antral and fundic pouches, this response was inhibited by antral acidification [124]; however, in humans, albeit those with intact stomachs, it appeared to be independent of lumenal pH. [125] In dogs, the pH sensitivity of the antral distention response required an intact cerebrovagal link to the antrum. In humans, low-dose atropine, which inhibited the acid secretory response to antral distention, nonetheless enhanced the elevation of serum gastrin levels. This suggests that there is an atropine-sensitive inhibitory pathway restraining the gastrin response to antral distention. Fundic distention in dogs with vagally innervated fundic pouches and vagally innervated antral pouches produces gastrin release and acid secretion. [126] Both responses are lost when the antral pouch is maintained at pH 2.5. This vasovagal reflex has been called the *oxyntopyloric reflex.* There is a similar vagally dependent acid stimulatory response to fundic distention in humans, but it appears not to be mediated through elevated serum gastrin. [121] The roles of histamine and acetylcholine in mediating distention-induced acid secretion are unclear. As may be predicted from our knowledge of the potentiation interrelations of the three principal secretagogues—gastrin, acetylcholine, and histamine, the acid response to gastric distention in humans is abolished by vagotomy or cimetidine. [127], [128] However, this may indicate the facilitory effect of background secretion of these secretagogues only. Gastric release of acetylcholine or histamine in response to distention has not been shown. Food interacts with the gastric mucosa to cause acid secretion in a manner that is independent of stretch by the food bolus. At least four constituents of food produce this stimulatory effect: peptic digests of proteins, ethanol, coffee, and Ca^{2+}. Whole proteins are poor stimuli of gastric acid secretion, but peptic digests of the same proteins are effective. [129] The breakdown products of protein, amino acids, and amines produce acid secretion principally through the release of gastrin. [31], [130], [131] The aromatic amino acids phenylalanine and tryptophan are the most potent stimuli of gastric acid secretion and gastrin release in vivo. [132] Their amine derivatives may contribute significantly to this response. [31], [130] Circulating gastrin is the principal mediator of postprandial gastric acid secretion. This has been demonstrated in human studies in which the increment in plasma gastrin that occurred after an intragastric infusion of amino acids or ingestion of a protein-rich meal was reproduced with an intravenous infusion of gastrin, and a similar acid secretory response to endogenous and exogenous gastrin was observed. [75], [131] These studies suggest that, despite the cephalovagal component to gastrin release, it is the chemical response to protein breakdown products that is the principal stimulus for postprandial gastrin. [131] Although amines and amino acids can cause gastrin release by direct action on the G cell, [31], [132] this phenomenon must be viewed within the larger orchestrated physiological response to ingested nutrients. Nutrients stimulate the release of many peptides into the circulation, including somatostatin, CCK, secretin, gastric inhibitory peptides, enteroglucagon, and peptide YY, which may influence acid secretion directly or by affecting gastrin release. Integrating these data with data derived from whole-animal physiology has proven difficult. Atropine enhances gastrin release in many circumstances, although according to the in vitro model previously described, inhibition or no change would be expected. Agents that block the gastrin response to bombesin do not affect the gastrin response to food. Studies in humans suggest also that ß-adrenergic innervation may play a role in gastric acid secretion. Administration of terbutaline, a $ß_2$-adrenergic receptor agonist, enhances serum gastrin but inhibits acid secretion in response to intragastric infusion of a homogenized meal. [133] Although it is likely that numerous neurohumoral mediators such as somatostatin and gastrin-releasing peptide and cholinergic and adrenergic innervation are significant factors in postprandial gastrin release, their relative roles in the integrated physiology of gastrin release remain as yet undefined. Studies of humans have demonstrated a modest acid stimulatory response or no response to direct intragastric infusions of pure alcohol. [134], [135] and [136] Red wine and beer, however, are potent stimuli of acid secretion and serum gastrin. [134], [135] and [136] These effects are probably caused by amines or amino acids in the beverage that stimulate gastrin release and not by a direct effect of its alcohol content. Caffeine stimulates acid release in humans. [137] McArthur and colleagues [138] showed that many household beverages were potent acid stimuli, including Tab, coffee, beer, and milk, each of which caused a pentagastrin-stimulated MAO of greater than 70%. Decaffeinated coffee was a potent stimulus, showing that it is not only the caffeine in coffee that is an acid stimulant. These studies did not attempt to control for the cephalic phase of acid secretion, nor was gastrin measured; it is therefore impossible to draw conclusions about the mechanisms that underlie these observations. Oral ingestion of calcium carbonate stimulates gastrin release and acid secretion in humans. [139] This action is independent of acid buffering capacity and is presumed to be an effect of dissociated Ca^{2+}.

Intestinal Phase The entry of chyme into the small intestine initiates the intestinal phase of the acid secretory process. The primary stimulatory factors are distention, proteins, and the products of protein digestion. [140], [141] Quantitating the significance of the intestinal phase to the stimulatory limb of acid secretion has proven controversial, perhaps because there are definite potentiation phenomena between the acid secretory response to intestinal nutrients and gastrin or histamine. [142] Serum gastrin levels do not appear to mediate the intestinal phase of acid secretion in dogs or humans. [141], [143] The acid stimulatory response to intestinal nutrients is preserved in vagotomized animals, indicating that circulating stimuli are involved to some degree. [144] This role may be filled in part by circulating amino acids, which have been shown to stimulate acid secretion without elevating serum gastrin. [145] There have been numerous attempts to isolate a distinct acid stimulatory peptide hormone (i.e., enterooxyntin) from small bowel mucosa, albeit without convincing success. [146], [147], [148] and [149]

Inhibition of Acid Secretion

Cephalic Influence Evidence suggesting that there may be cephalic inhibitory influence acting on gastric acid secretion comes from intracerebral microinjection studies. [150] Several of the peptides that can lead to this inhibitory event after intracerebroventricular injection are outlined in Table 13-1. It appears that vagal fibers carry inhibitory and stimulatory messages to the parietal cells. As with the cerebral microinjection studies in which gastric acid is stimulated, however, there are no data to indicate which, if any, of these observations are relevant to the cephalic phase of gastric acid secretion. In addition, the complex response of stimulated serum gastrin levels to low-dose atropine described previously suggests that there are vagal inhibitory influences on gastrin release.

Gastric Mediation Just as vagal inhibitory fibers have been implicated in the cephalic acid secretory response, there appear to be vagally mediated inhibitory neural arcs involved in the distention-induced acid secretory response. Studies of dogs and humans led Debas and colleagues [124], [151], [152] to conclude that antral distention in addition to stimulating serum gastrin release results in the release of an inhibitor of acid secretion. Whether this effect is humoral or neurocrine is not clear, but it does depend on an intact vagus. Gastrin release in response to nutrients, sham feeding, and antral distention is inhibited by the presence of acid in the gastric antrum. [86], [124], [153] An intralumenal pH of 3 appears to be the threshold for initiating this response. [154] The mechanisms of this negative-feedback loop probably include somatostatin release as a paracrine or endocrine gastrin inhibitor. [154] Evidence from in vitro human studies suggest that gastric acid regulates somatostatin release locally in the stomach and postprandial circulating somatostatin. [155], [156] An alternative mechanism for the inhibition of gastrin release at low intralumenal pH is that an acidic milieu causes amines to be protonated, and as charged particles, they are not taken up by G cells. [157] This cannot be the complete answer because, at pH 2.5, cephalovagal stimulation of gastrin is inhibited. [86] It is probable that somatostatin acts directly on the parietal cell to inhibit acid secretion by means of paracrine or endocrine pathways. [16], [158]

Intestinal Inhibition The observation that nutrient infusion into the small intestine inhibits acid secretion is an old one. Feng and colleagues [11] proposed that fat infusion into the small bowel inhibited acid secretion by the release from the small bowel mucosa of a circulating inhibitory hormone they called enterogastrone. There have been at least seven candidate peptides for this role: somatostatin, neurotensin, gastric inhibitory peptides, peptide YY, secretin, CCK, and galanin. [159], [160], [161], [162], [163], [164], [165], [166] and [167] It is probable that enterogastrone is not a single entity but rather a physiological response to more than one circulating acid inhibitor.

CELLULAR BASIS OF ACID SECRETION

Analysis of the regulatory effects of gastric secretagogues in vivo has provided important insight into the mechanisms of acid secretion in animals and umans. Interactions among neurocrine, paracrine, and endocrine signals and their effects on the parietal cell ultimately must be studied in the intact organism. However, the complexity of the regulatory effects converging on the parietal cell makes integrated in vitro analysis a necessity. In vitro models of acid secretion include gastric mucosa preparations, gastric glands, and isolated enriched parietal cells. Gastric endocrine cells effecting acid secretion have also been enriched and analyzed. These models have provided an understanding of the cellular events leading to acid secretion.

In Vitro Models

Isolated preparations of intact gastric mucosa allow the study of secretory events while maintaining all the mucosal cell types in their usual cellular environment. Cell polarity, tight junctions, gap junctions, desmosomes, and certain paracrine effects are maintained in such models. Physiological neurocrine and endocrine effects are absent but can be mimicked by bathing one or both surfaces with the desired neurotransmitter or hormone. One such model, the isolated bullfrog mucosa, has been used to study histamine release and acid secretion. [168] Mucosal strips are bluntly dissected and are mounted as a sheet between two sides of a Lucite chamber. A flow-through system can be used to ensure rapid changes of the serosal solution. Acid secretion into the mucosal solution is measured with a pH stat by maintaining the pH at 7.0 with isotonic 15 mM NaOH. Other types of mucosal studies use explants maintained in organ culture or fragments of mucosa suspended in tissue culture solutions. [169], [170] and [171] Studies using these types of models have been supplanted by gastric gland and isolated cell studies.

The initial steps in the preparation of glands and cells are similar. [172], [173] The gastric mucosa is bluntly dissected away from the submucosa, finely minced, and then dispersed with pronase or crude collagenase. The viability rate of parietal cells obtained with this technique generally exceeds 95% as judged by trypan blue exclusion.

After digestion of the gastric mucosa, the glands can be separated from the cells and debris by several sedimentation washes at unit gravity. The large size of the glands allows them to sediment rapidly, essentially free of nonglandular material. Separation of isolated cells can be achieved by velocity and density separation techniques. Counterflow elutriation is a velocity separation technique that separates cells predominantly on the basis of size. [173] Enriched fractions routinely contain 50% to 70% parietal cells that maintain their viability and biologic activity. Antral G cells and fundic D cells have also been enriched by elutriation. [174] A second purification step using a density gradient can be added to enrich parietal or endocrine cells further.

There is a potential limitation in using isolated parietal cells for physiological studies. After the cells are separated from the microenvironment found within the gastric mucosa, their ability to respond to secretogogues in a tissue-specific fashion diminishes within 8 to 10 hours after isolation (Del Valle and Yamada, unpublished observations). To circumvent this problem, Chew and Ljunstrom [175] developed a system for culturing isolated parietal cells so they retain differentiated structure and function for as long as 72 hours after isolation. These cultured cells express receptors for the appropriate secretagogues and maintain expression of parietal cell–specific genes such as H^+,K^+-ATPase. This model provides an important tool for characterizing the multiple factors involved in regulating parietal cell physiology.

Gastric secretion has been analyzed in different species with each of these models. Species variety may account for certain discrepant results occasionally observed with different models. Many analyses of the cellular events controlling acid secretion have used canine gastric mucosal cells enriched by counterflow elutriation. The data obtained using canine cells can be correlated with the large body of data obtained with in vivo acid secretory studies in dogs. Moreover, canine parietal cells have several features in common with human parietal cells.

Indirect measures of parietal cell function have been developed to quantitate the biologic effect of various secretagogues and inhibitors. The development of these assays has made it possible to correlate the binding of ligands with their functional effects. Oxygen consumption has been shown to correlate with HCl secretion in both in vivo and ex vivo studies. [176], [177] Oxygen consumption in isolated enriched parietal cells increases in response to gastrin, carbachol, and histamine, probably reflecting the activity of the proton pump. [173] Glucose oxidation can also be used as a measure of parietal cell metabolic activity. [178] Morphologic transformation of parietal cells in response to secretagogues produces dramatic changes in cell appearance. Resting cells are filled with tubulovesicles, and stimulated cells rapidly develop a dense intracellular canalicular network communicating with the cell's apical surface. Nomarski optics can visualize this transformation in living cells, and fluorescent microscopy with acridine orange can show the accumulation of fluorescent dye in the newly generated acid spaces. [179]

The accumulation of weak bases in membrane-bound acid spaces is the basis of the [^{14}C]aminopyrine uptake assay. [15], [180] Uncharged aminopyrine is lipid soluble and easily crosses cellular membranes. On entering an acidic compartment, aminopyrine is protonated and loses its lipid solubility. This sequestration of [^{14}C]aminopyrine correlates with intracellular acid formation and is the most commonly used assay for parietal cell stimulation. Other methods involving measurement of the proton pump association with canalicular membranes and correlation of morphologic changes in parietal cells with functional measurement of acid production using fluorescence microscopy techniques hold promise for the dynamic study of parietal cell physiology. [181], [182] and [183]

Receptors

Separation of gastric cells into highly enriched cell populations has made it possible to identify and characterize specific receptors on each cell type. [184] Purified cells maintain intact receptors through the separation procedure and bind radiolabeled ligands specifically. The binding of a ligand should correlate with a functional assay over the same dosage range to confirm that a specific cell type has biologically active receptors. An example is the correlation of carbachol binding to parietal cells and the induction of aminopyrine uptake by carbachol.

After functional receptors have been demonstrated, they can be characterized in binding studies with available receptor agonists and antagonists. Further characterization of receptors has been achieved by solubilizing them from cell membranes and cross-linking them to specific radiolabeled ligands. The solubilized

form of the receptor can be examined for size, subunit structure, or ligand-induced autophosphorylation.

The receptors can be purified, their amino acid sequence analyzed, and their genes eventually cloned. The application of molecular biologic techniques to the study of cell surface receptors has greatly enhanced our understanding of their structure and function. Several receptors that are critical for gastric secretory function have been cloned. These include the histamine H_2, somatostatin, M_3 muscarinic, and gastrin or CCK-B receptors. [185], [186], [187] and [188] Analysis of the amino acid sequence of these cloned receptors indicates that they belong to the family of heptahelical or G protein–linked receptors. Structurally, these receptors consist of a single amino acid chain, containing seven hydrophobic domains, that presumably traverses the cell surface membrane (i.e., transmembrane regions). Through studies using recombinant molecular technology (e.g., site-directed mutagenesis, chimeric receptor studies), the structural components of G protein–linked receptors important for ligand binding and biologic action have been elucidated, and several general characteristics of this receptor family have emerged. For example, the specificity of ligand binding can be determined by the hydrophobic transmembrane regions of the receptor and the extracellular domains. Furthermore, coupling to the G proteins that are responsible for secretagogue-mediated signal transduction involves interaction principally with the third intracellular loop of the receptor. Receptor regulation (e.g., uncoupling, sequestration, down-regulation) appears to involve kinase-mediated phosphorylation events targeted to serine and threonine residues found within the carboxyl terminal portion of these receptors. Detailed structure-function analyses of the receptors important in gastric secretory function are in progress.

The question of whether acid secretagogues act directly or indirectly on parietal cells could be answered after isolated parietal cell preparations were developed. Initial studies showed that histamine, carbachol, and gastrin increased canine parietal cell oxygen uptake. [173], [189] Later studies revealed that [^{14}C]aminopyrine accumulation was also increased by each of these secretagogues. [14], [15] By using isolated rabbit gastric glands, somewhat different results were obtained; aminopyrine accumulation could be demonstrated in response to histamine and carbachol but not to gastrin. [180] These differences may reflect species variability. For example, rabbits, which are known to feed continuously, may not have developed the ability to marshal an acid secretory response to intermittent bolus feeding as mediated by gastrin. Alternatively, these differences may reflect differences in the methods of study. The experiments of Del Valle and colleagues [190] have demonstrated marked increases in intracellular Ca^{2+} concentration ($[Ca^{2+}]_i$) in response to administration of gastrin 17 in single isolated rabbit parietal cells, suggesting that gastrin receptors are present on rabbit cells as well.

The concept that secretagogues such as acetylcholine and gastrin directly activate parietal cells was placed in doubt by a study that demonstrated the expression of histamine H_2, gastrin, and muscarinic receptors on immunocytes found in gastric lamina propria instead of on parietal cells. [191] This controversial issue was revisited by Diaz and colleagues. [192] Using in situ hybridization histochemistry and autoradiography with a highly selective H_2 receptor radioligand (^{125}I-aminopotentidine), a specific signal for H_2-specific gene transcripts was detected only within parietal cells of the gastric epithelium. The discrepant results obtained by these two investigative teams may be attributed to the less sensitive oligonucleotide probes used by the former group as compared with the highly specific H_2 receptor antisense riboprobe used by the latter. The classic secretagogues described earlier can clearly act directly on gastric parietal cells as initially postulated by multiple investigators, and the possibility that receptors for these ligands are also located on immunocytes in the lamina propria should not be excluded.

Although the critical role of the H_2 receptor in regulating gastric acid secretion had been well established, the structural components of this receptor that determine H_2 selectivity were unknown. The success of Gantz and associates [185] in cloning the gene encoding the H_2-histamine receptor provided the essential tools required for studying this question further. Key areas of homology in the structures of the H_2-histamine and ß$_2$-adrenergic receptors suggested specific transmembrane amino acids that could be important for histamine binding. On the basis of studies involving the expression of H_2-histamine receptors with site-directed mutations, a model for histamine binding and action on the H_2 receptor has been proposed. [193] An aspartic acid residue (Asp98) in the third transmembrane domain is essential for histamine binding and action, and an aspartic acid (Asp186) in the fifth transmembrane defines H_2 selectivity. A threonine (Thr190) in the fifth transmembrane domain is important in establishing the kinetics of histamine binding but is not essential for H_2 selectivity.

The presence of a specific muscarinic receptor on canine parietal cells is supported by specific blockade of carbachol's biologic effects with atropine. The dissociation constant for atropine inhibition of carbachol-induced aminopyrine uptake, 1.3 nM, is consistent with dissociation constants observed with muscarinic receptors in other tissues. [15] In addition to inducing oxygen uptake and aminopyrine accumulation in a dose-dependent manner, carbachol produces a parallel increase in the turnover of membrane inositol phospholipids. [194] Pharmacological studies indicate that the parietal cell muscarinic receptor is of the M_3 subtype, which has been confirmed by the molecular cloning of this receptor. [195], [196] Rat parietal cells have also been shown to have M_3 muscarinic receptors that stimulate aminopyrine uptake, increase inositol phospholipid turnover, and bind [*N*-methyl-^{3}H]scopolamine. [197]

Gastrin receptors have been localized to isolated rat [198] and canine [14] parietal cells. Binding studies with purified parietal cells reveal specific binding that is rapid and saturable. Proglumide, a CCK or gastrin receptor blocker, inhibits gastrin binding and stimulation of parietal cell function. CCK-8 is equipotent with gastrin 17 in displacing radioligand and in stimulating aminopyrine uptake. [14] This suggests that the parietal cell gastrin receptor binds and responds to either ligand in a similar fashion. The gastrin receptor has been characterized further by cross-linking studies in two species. Using canine parietal cell membranes, cross-linking studies with ^{125}I-gastrin 2–7 revealed a single gastrin receptor with a molecular weight of 74 kd. [199] Half-maximal inhibition of radiolabeled gastrin binding in these canine parietal cell preparations was 3×10^{-10} M, in agreement with the potency of gastrin in stimulating aminopyrine uptake. [14] Similar studies with detergent extracts of porcine gastric mucosal membranes using ^{125}I-[Nleu^{15}]gastrin 2–17 as a ligand resulted in the cross-linking of a 78-kd binding protein. [200] In contrast to the studies using canine parietal cells, however, 50% inhibition of binding required 2×10^{-6} M [Nleu^{15}]gastrin 17. The apparent reduction in affinity may be an artifact of the extraction technique used. In both species, the receptor appears to be a single protein with no disulfide-linked subunits. These studies confirm the presence of a specific gastrin receptor on parietal cells. The gastrin receptor appears to be coupled to membrane inositol phospholipid turnover and protein kinase C (PKC) activation. [194], [201]

Kopin and colleagues [188] isolated a cDNA clone encoding the gastrin receptor found on parietal cells. They determined that it is a member of the G protein–linked receptor family and, when expressed, it has a molecular weight similar to that reported in the receptor cross-linking studies. Expression of the cloned receptor confirmed that it is coupled to membrane inositol phospholipid turnover and mobilization of $[Ca^{2+}]_i$. Song and colleagues [202] cloned the gene encoding the human gastrin or CCK-B receptor and localized it to a region of chromosome 11 (11p15.4). It appears that the gene produces two different receptor proteins as a result of alternative RNA splicing. The functional difference between the two receptor isoforms remains to be established.

As with many peptides, gastrin is synthesized as a precursor molecule that undergoes posttranslational processing to become a C-terminus amidated product, which is presumed to be the sole biologically active form of this hormone. [203] Indeed, posttranslational processing intermediates of gastrin, specifically the C-terminus glycine-extended form (G-Gly), serve as substrate for amidation, are stored in gastric tissues, and are secreted into the circulation with amidated peptide reaching plasma concentrations roughly equivalent to those of processed gastrin.

The question whether posttranslational processing intermediates of gastrin exert a biologic effect was addressed by Seva and colleagues. [204] These investigators demonstrated that G-Gly exerts growth promoting effects by activation of non–CCK-B receptors. [204] This finding demonstrated that glycine-extended intermediates of prohormone processing have independent and hitherto unrecognized important biologic actions. Subsequently, Kaise and associates [205] showed specific binding of [^{125}I-Leu^{15}]-G2-17-Gly to gastric canine parietal cells, which was dose-dependently displaced by G2-17-Gly but not by gastrin or by the specific gastrin/CCK-B receptor antagonist L365,260. Thus, gastric parietal cells appear to express specific and distinct receptors for both gastrin and G-Gly.

The role of G-Gly in gastric acid secretion has been the focus of ongoing investigative efforts. An early study conducted in isolated canine gastric parietal cells demonstrated that G-Gly was at least four orders of magnitude less potent than gastrin in stimulating gastric acid secretion. [206] Interestingly, inhibition of gastrin amidation by copper chelation with diethyldithiocarbamate led to an unexpected increase in both basal and gastrin-stimulated gastric acid outputs, suggesting that chronically elevated levels of G-Gly could have a stimulatory effect on the gastric secretory process. [207] Higashide and colleagues [208] demonstrated that infusion of G-Gly alone did not stimulate gastric acid secretion in rats. However, administration of G-Gly in combination with gastrin significantly potentiated the response observed in the presence of gastrin alone, further supporting a physiological stimulatory role for G-Gly on gastric acid secretion. [208] Chen and associates [209] observed similar potentiating effects on gastric acid secretion after administering G-Gly in combination with gastrin to gastrin-deficient mice. Interestingly, in these animals, G-Gly infusion did not affect the number of either ECL or parietal cells, the proliferation rate of the fundic epithelium or the level of expression of H^+,K^+-ATPase. Administration of G-Gly, however, appeared to prevent the formation of vacuolar canaliculi and lipofuscin bodies, suggesting that G-Gly could synergize with gastrin

to stimulate gastric acid secretion by preventing parietal cell degradation. [209]

Another possible mechanism for these observations was presented in a study performed in cultured canine gastric parietal cells. [205] Preincubation of parietal cells with G-Gly enhanced the acid secretory response of the cells to histamine, whereas acutely administered G-Gly had no effect. [205] G-Gly dose-dependently increased both the expression and transcription of the gene encoding the a subunit of H^+,K^+-ATPase, suggesting that G-Gly may have a functional role in potentiating gastric acid secretion by enhancing the expression of the gene encoding the protein responsible for H^+ generation. [205] Although these observations are in apparent discordance with those reported in the study of the gastrin-deficient mice, it is possible that these genetically engineered animals exhibit developmental abnormalities in the mechanisms regulating the expression of H^+,K^+-ATPase. Alternatively, these findings may reflect species-specific differences regarding the ability of G-Gly to induce H^+,K^+-ATPase gene expression.

The effects of somatostatin on isolated enriched canine parietal cells have been evaluated. Somatostatin dose-dependently inhibits histamine-induced aminopyrine uptake and cAMP production and pentagastrin-stimulated aminopyrine accumulation. [16] Somatostatin binding sites have been identified using ^{125}I-[Leu^{8}-D-Trp^{22}-Tyr^{25}]somatostatin 28 as radioligand. Somatostatin 14 and somatostatin 28 are equally potent at displacing bound ligand and at inhibiting aminopyrine accumulation. Scatchard analysis of the binding data revealed two binding sites with dissociation constants of 3.2×10^{-9} M and 2.1×10^{-7} M, respectively. Crude membranes prepared from 95% to 100% pure parietal cells were incubated with ^{125}I-[Leu^{8}-D-Trp^{22}-Tyr^{25}]somatostatin 28 and cross-linked with disuccinimidyl suberate. [210] After solubilization, a single sharp band with no disulfide linkages was identified corresponding to a membrane receptor with a molecular weight of 99 kd. These parietal cell somatostatin receptors may be activated by means of a direct paracrine effect mediated by long cytoplasmic processes that extend from fundic mucosal D cells and appear to terminate on parietal cells. [32] Endocrine inhibition of acid secretion by postprandial serum somatostatin may also be mediated through these receptors.

Five members of the somatostatin receptor family (SSTR1–SSTR5) have been cloned. [180] and characterized with distinct pharmacological properties and tissue distribution. All five SSTR subtypes are expressed throughout the rat gastrointestinal tract and, specifically, in all layers of the gastric mucosa. [211] The development of highly specific and selective agonists [212] has led to more precise functional characterization of these different somatostatin receptors. In particular, SSTR2 appears to mediate the inhibition of gastric acid secretion in rats, dogs, and humans, [212], [213] and [214] a finding confirmed by selective ablation of the *SSTR2* gene. [215] Moreover, Prinz and colleagues [216] demonstrated that SSTR2 is the predominant somatostatin receptor subtype expressed on rat ECL cells, where it mediates the inhibition of histamine release. Therefore, although all five SSTRs are expressed in the stomach, SSTR2 appears to be the receptor subtype most involved in regulating the gastric secretory process.

The presence of other receptors on parietal cells has been suggested but awaits confirmation. Prostaglandins E_2 (PGE_2) and I_2 (PGI_2) inhibit histamine-stimulated aminopyrine accumulation and cAMP generation in enriched canine parietal cells. [217] These agents have no effect on aminopyrine uptake induced by gastrin, carbachol, or dibutyryl cAMP. Specific binding sites for PGE_2 exist in porcine fundic mucosa with subcellular membrane fractionation, suggesting localization to the plasma membrane. [218], [219] and [220] Prostaglandins have displaced bound [^{3}H]PGE_2 and inhibited histamine-stimulated aminopyrine uptake in isolated rabbit parietal cells. [221] Together, these findings strongly suggest the presence of prostaglandin receptors on parietal cells. Ding and colleagues [222] confirmed this observation by documenting gene expression of the prostaglandin receptor subtypes EP_3 and EP_4 on rat gastric parietal cells.

Although it is fairly clear that H_3 receptors are expressed on ECL cells, serving a role in autocrine regulation of histamine release, their presence on parietal cells remains controversial. The H_3 receptor agonist methylhistamine is a potent stimulant of aminopyrine uptake in isolated cultured rabbit parietal cells. [223] This observed stimulatory effect can be blocked by the H_2 receptor antagonist ranitidine, placing in question the selectivity of the H_3 receptor agonist and the significance of the H_3 receptor in parietal cell physiology.

The sympathetic arm of the autonomic nervous system may be expected to counterbalance the stimulatory effect of muscarinic receptors on acid secretion. Although it is possible that ß-adrenergic agonists may inhibit acid secretion through stimulation of somatostatin release [224] and inhibition of histamine release by fundic mucosal mast cells, [184], [225] direct studies with parietal cells support the presence of stimulatory ß-adrenergic receptors. [226]

Epidermal growth factor (EGF) and transforming growth factor-a (TGF-a) have been implicated in the inhibitory modulation of gastric acid secretion. [227], [228] and [229] TGF-a shares structural homology with EGF and is expressed within parietal cells. Several studies have demonstrated that parietal cells express the receptor for EGF, which recognizes EGF and TGF-a. [230], [231] Both these growth factors inhibit parietal cell function in a similar fashion. [232], [233], [234] and [235] TGF-a is secreted by parietal cells, and it may function as an autocrine regulatory factor. The presence of secretin, glucagon, or opioid receptors on parietal cells has been suggested but requires confirmation.

Beales and Calam [236] have suggested that parietal cells may express receptors for cytokines. In a series of studies conducted in isolated rabbit gastric parietal cells, tumor necrosis factor-a and interleukin-1ß were shown to inhibit gastric acid secretion, as assessed by the accumulation of the weak base aminopyrine, both in the basal state and after stimulation with histamine, gastrin, and carbachol. [236] In contrast to these observations, Yakabi and colleagues [237] reported that interleukin-8 enhanced in vivo gastrin-stimulated gastric acid secretion in rats. Thus, cytokines appear to have complex regulatory effects on gastric acid secretion, which might be mediated, at least in part, by a direct action of these agents on the gastric parietal cells.

An overview of the interactions of ligands and receptors involved in acid secretion is provided in Figure 13-6. The parietal cell has stimulatory receptors for gastrin, acetylcholine, and histamine. Several lines of evidence suggest that parietal cells in humans may be exposed continuously to basal levels of acetylcholine and histamine. Gastrin can account for most of the postprandial increase in gastric acid secretion. [131] Although a portion of the postprandial response is the result of a direct effect of gastrin on parietal cells, a significant component may be caused by the ability of gastrin to stimulate histamine release from ECL cells (see the section on histamine). The parietal cell also has inhibitory somatostatin receptors that counteract the secretagogue effects. Vagal nerve fibers may enhance their acid stimulatory effect through stimulatory muscarinic receptors on G cells and inhibitory muscarinic receptors on D cells. [224] Adrenergic fibers appear to stimulate D-cell secretion and inhibit release by histamine-containing cells, counterbalancing the vagal effects. [225] Gastrin and CCK stimulate parietal cells and D cells. [238] Gastrin-releasing peptide acts as a neurotransmitter that stimulates G cells, and somatostatin may function as a paracrine G-cell inhibitor. [239], [240] and [241] Further structural and functional analyses of each of these receptors will greatly enhance the understanding of gastric secretion.

FIGURE 13-6. Regulation of gastric acid secretion. Major gastric mucosal ligand-receptor interactions regulate parietal cell HCl secretion. *D cell,* somatostatin cell; *G cell,* gastrin cell. (Adapted from Feldman M. Acid and gastrin secretion in duodenal ulcer disease. Regul Pept Lett 1989;1:1.)

Intracellular Signal Transduction

As previously described, the gastric parietal cell is a complex biologic structure that is controlled by a broad variety of growth factors, hormones, and

neurotransmitters. These agents interact with specific receptors on the cell surface, initiating a flow of information that moves to the cell nucleus along highly organized and complex signal transduction pathways. Once in the nucleus, these signals are known to activate specific programs of transcriptional events that lead to the expression of specialized cellular functions. Numerous signal transduction pathways have been studied and characterized in the gastric parietal cells.

Receptors linked to adenylate cyclase influence intracellular levels of cAMP. Such receptors are coupled to inhibitory (G_i) or stimulatory (G_s) GTP-binding proteins (Fig. 13-7). [242] G_i attenuates adenylate cyclase activity, decreasing cAMP levels. The ability of pertussis toxin selectively to block the actions of G_i can be used experimentally to dissect second messenger pathways. G_s, which increases adenylate cyclase activity and cAMP levels, can be stimulated selectively by cholera toxin. Increases in cytoplasmic cAMP levels result in activation of cAMP-dependent protein kinases and consequent phosphorylation of various intracellular proteins that appear to mediate the effects of the ligand-receptor interaction.

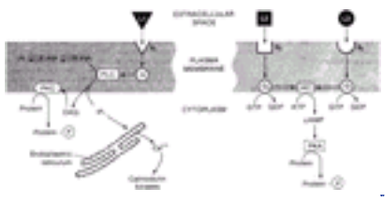

FIGURE 13-7. Signal transduction pathways in parietal cells. Ligands (*L*) interact with receptors (*R*) to initiate the target cell response. The **left panel** shows the membrane inositol phospholipid pathway activated by gastrin and acetylcholine (*L1*). A guanine nucleotide–binding protein (*G*) activates phospholipase C (*PLC*), which hydrolyzes phosphotidylinositol 4,5-bisphosphate (PIP_2) to diacylglycerol (*DAG*) and inositol 1,4,5-trisphosphate (IP_3). DAG activates protein kinase C (*PKC*), which phosphorylates (*P*) target proteins, whereas IP_3 induces intracellular calcium mobilization, which activates kinases. The **right panel** depicts the cAMP pathway used by histamine (*L2*) and somatostatin (*L3*) in parietal cells. The histamine receptor (R_2) acts through a stimulatory G protein (G_s), which activates adenylate cyclase (*AC*) and generates cAMP. An increased level of cytoplasmic cAMP activates protein kinase A (*PKA*) and results in phosphorylation of parietal-cell effector proteins. The somatostatin receptor (R_3) activates an inhibitory G protein (G_i), which inhibits adenylate cyclase and decreases cAMP generation.

A second major intracellular signal transduction cascade involves the turnover of membrane phospholipids, specifically the inositol phospholipids. Receptors are linked to this pathway through a G protein, and the initial events that are activated by occupancy of their receptor lead to phospholipase C–induced hydrolysis of phosphatidylinositol bisphosphate (PIP_2) to diacylglycerol and inositol triphosphate (IP_3). [243] IP_3 causes the release of Ca^{2+} from intracellular stores, and diacylglycerol promotes the translocation of a Ca^{2+}-phospholipid–dependent protein kinase (i.e., PKC) from the cytoplasm to its active site on the cell membrane. [244], [245] Increases in $[Ca^{2+}]_i$ activate various Ca^{2+}-dependent enzyme systems, such as the calmodulin kinases, and promote the translocation and activation of PKC.

Although the classic teaching had been that receptor activation is linked to a single signal transduction cascade, it is evident that interaction between postreceptor signaling pathways can occur at multiple levels. [187] One example of this in gastric secretion is the ability of the H_2 histamine receptor to stimulate through separate pathways an increase in cAMP and $[Ca^{2+}]_i$. [246] Del Valle and colleagues [246] have confirmed that stimulation of the cloned canine H_2 receptor can lead to an increase in adenylate cyclase and IP_3 or $[Ca^{2+}]_i$ activity through independent pathways.

Histamine's acid stimulatory action appears to be mediated primarily through its ability to increase cAMP production in parietal cells. [247] This increase parallels histamine's ability to stimulate aminopyrine uptake and oxygen consumption. Histamine increases cAMP-dependent protein kinase activity in enriched rabbit parietal cells. [248] The substrates for this enzyme activity have not been fully characterized, but they are thought to mediate the effects of histamine stimulation. [249]

The acid stimulatory actions of gastrin and carbachol appear to be mediated by Ca^{2+}-dependent pathways. Gastrin and carbachol exert similar effects on membrane inositol phospholipid turnover in parietal cells. Both agents induce a time-dependent decrease in PIP_2 and increase in the formation of IP_3. [194] These effects parallel increases in [^{14}C]aminopyrine accumulation. The increase in IP_3 may mediate the mobilization of $[Ca^{2+}]_i$ observed in parietal cells stimulated with gastrin or carbachol. In the case of carbachol, increased $[Ca^{2+}]_i$ may occur as a result of enhanced mobilization of Ca^{2+} from intracellular stores or through influx of extracellular Ca^{2+} across the cell membrane. [250], [251] and [252] It is presumed that the elevated levels of cytoplasmic Ca^{2+} then activate several enzyme cascades, including the calmodulin kinase family and, in concert with diacylglycerol, the second product of PIP_2 breakdown, PKC. Gastrin and carbachol dose-dependently increase membrane-associated PKC activity. [201] Although gastrin and carbachol increase membrane inositol phospholipid turnover and PKC activity in canine gastric parietal cells to a similar degree, carbachol is a more potent stimulant of acid production. [201] Takeda and associates [253] observed that carbachol is more efficacious than gastrin in stimulating the entry of extracellular Ca^{2+} into parietal cells. It appears that these two secretagogues regulate the entry of Ca^{2+} into parietal cells by distinct mechanisms. Gastrin stimulated Ca^{2+} entry from extracellular sites by opening Ca^{2+} channels that are linked to the ability of this stimulant to deplete $[Ca^{2+}]_i$ stores. Carbachol opens similar Ca^{2+} channels and opens channels that are not regulated by the depletion of $[Ca^{2+}]_i$ pools. These differences in Ca^{2+} regulation may account for the more potent stimulatory action of carbachol on canine gastric parietal cells.

Furthermore, research suggests that the induction of $[Ca^{2+}]_i$ release in response to gastrin but not carbachol is a function of activation of the cAMP pathway. [254] In fact, incubation of rabbit parietal cells with gastrin in the presence of the histamine H_2 receptor blocker cimetidine inhibits gastrin-mediated increases in $[Ca^{2+}]_i$. The addition of dibutyryl adenosine 3',5' phosphate in addition to cimetidine completely restored this response, suggesting that in the rabbit, H_2 receptor activity potentiates the effect of gastrin on parietal cell function. Athmann and colleagues [255] confirmed these observations, demonstrating that in isolated rabbit gastric glands, ranitidine blocks $Ca^{2+}]_i$ release induced by gastrin (1 nM). Interestingly, these investigators also reported that a supraphysiological, tenfold higher dose of gastrin (10 nM) induced a Ca^{2+} signal that was not inhibited by ranitidine. [255] The specific intracellular targets and the biologic significance of this ranitidine-insensitive Ca^{2+}signal remain to be elucidated.

Although it appears that the plateau or steady phase of carbachol-mediated increases in Ca^{2+} is essential for activation of the secretory process, [256] this may not be the only signaling event important in muscarinic receptor–mediated acid secretion. Similar Ca^{2+} levels can be achieved with the calcium ionophore, ionomycin, and the Ca^{2+}-ATPase inhibitor thapsigargin, [257] but a corresponding increase in acid production similar to that observed with carbachol is not observed. The additional signaling events linking M_3 receptor activation and acid secretion require further investigation, but they may include differential activation of downstream kinase pathways.

Ca^{2+}/calmodulin-dependent protein kinase II (CaMKII) is one of the best-characterized targets of intracellular Ca^{2+}. Tsunoda and colleagues [258] demonstrated that carbachol can lead to the activation of CaMKII in parietal cells. The specific components of the carbachol-activated signaling cascade are for the most part unknown. Parente and colleagues [259] have reported the purification and cloning of a novel phosphoprotein, designated Ca^{2+}-sensitive phosphoprotein of 28 kd (CSPP28), from rabbit parietal cells. This protein was phosphorylated by CaMKII in response to cholinergic stimulation, but not when cells were treated with phorbol esters. CSPP28 may represent an important element in Ca^{2+} signal transduction, but its specific role in parietal cell biology remains unknown. Because CaMKII regulates cytoskeletal function, this protein kinase may play a role in the dramatic morphologic transformation of parietal cells during stimulation.

Carbachol-mediated stimulation of various cell types leads to alterations in cell volume that are linked to the regulation of ion channel conductance. Negulescu and colleagues [260] examined the role of parietal cell volume in regulating carbachol-mediated changes in cytosolic Ca^{2+}. These investigators observed that carbachol-mediated opening of Ca^{2+} channels leads to the loss of ions and water from the parietal cell and subsequent cell shrinkage. The decrease in cell volume

may inhibit Ca $^{2+}$ channels, providing a feedback mechanism to reduce further cell shrinkage resulting from ion fluxes.

The action of PKC in parietal cells is complex. Direct activation of PKC with phorbol esters results in enhanced acid secretory activity, [261] and inhibition of PKC by the specific inhibitor Ro 31-8220 blocks carbachol-stimulated aminopyrine uptake in isolated rat parietal cells. [262] However, phorbol ester pretreatment of canine parietal cells with phorbol esters decreases the stimulatory effects of subsequent treatment with carbachol and gastrin, [261] whereas Ro 31-8220 potentiates [^{14}C]aminopyrine uptake from isolated rabbit gastric glands stimulated by both carbachol and histamine. [263] Numerous factors may account for these contradictory observations. PKC may induce down-regulation of muscarinic and gastrin receptors on parietal cells. [261] The mechanism for this effect is unclear but may involve receptor phosphorylation. In addition, because PKC comprises a large family of proteins with different biochemical and functional properties, [264] species-specific differences in both the function and the cellular localization of PKC isoforms may exist. [263] Chew and colleagues [263] analyzed the different PKC subtypes in rabbit parietal cells and reported abundant levels of both the novel isoforms PKC-e and PKC-µ and of the atypical isoforms PKC-?, PKC-?, and PKC-?. In contrast to a previous study performed in canine parietal cells, [265] low levels of the classic isoforms PKC-a and ß were measured in the rabbit parietal cells. These investigators observed, with a confocal microscope, that PKC-e was localized to a parietal cell compartment that bore resemblance to that containing filamentous actin, suggesting that PKC could negatively regulate gastric acid secretion through phosphorylation and modification of cytoskeletal proteins. [263] A newer protein kinase, protein kinase D (PKD), has been characterized and found to be induced by phorbol esters. [266], [267] Although it is currently unknown whether PKD or other related protein kinases are present in gastric cells, this intriguing finding supports the hypothesis that some of the effects of phorbol esters on acid secretion could be mediated by protein kinases other than PKC. Further studies will clarify the function of PKC in the stomach.

In addition to these well-established signal transduction pathways, mammalian parietal cells express multiple members of a family of protein kinases known as mitogen-activated protein kinases (MAPKs). [268], [269] and [270] In particular, parietal cells appear to express the extracellular signal–regulated protein kinases (ERKs), [268], [270] the Jun N-terminal kinases (JNKs), [271] and the p38 kinases, [272] molecules known to regulate multiple important cellular functions [273], [274] (Fig. 13-8). The ERKs are important elements in a cascade of biochemical reactions that involves the small GTP-binding protein Ras, as well as upstream protein kinases such as Raf and MAPK/ERK. [275], [276] ERK activation is known to target numerous cellular proteins, including downstream protein kinases such as 90-kd S6 kinase (RSK) [275], [276] and transcription factors, such as Elk-1, that regulate the activity of the promoter of the early response gene c- *fos* through the serum response element (SRE). [277], [278] These kinases appear to play a crucial role in the process of amplification, integration, and transmission of extracellular signals from the cell surface to the nucleus, leading to the induction of cellular growth and proliferation and, in some systems, cellular differentiation. [274], [275] and [276]

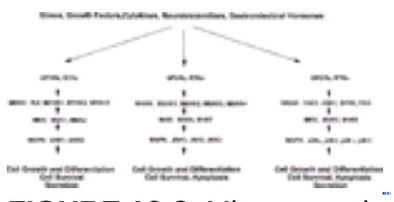

FIGURE 13-8. Mitogen-activated protein kinase (*MAPK*) pathways in the gastric parietal cells. Many different extracellular signals activate kinase cascades that lead to the induction of members of the MAPK family of protein kinases. These include the extracellular signal–regulated protein kinases (*ERK*), p38 kinases, and the Jun N-terminal kinases (*JNKs*). These protein kinases appear to regulate several important cellular functions, including growth, apoptosis, survival, differentiation, and secretion. *GPCRs,* G protein–coupled receptors; *RTKs,* receptor tyrosine kinases.

Several studies have examined the regulation of ERKs by gastric acid secretagogues. Takeuchi and colleagues [270] reported that carbachol was the most potent inducer of ERK2 activity in isolated gastric canine parietal cells. Gastrin and EGF had weaker stimulatory effects, whereas histamine induced no response. The effect of carbachol appeared to be independent of Ca $^{2+}$ signaling. PD98059, a selective inhibitor of the upstream ERK activator, MEK dose-dependently inhibited both carbachol- and EGF-stimulated ERK2 activity. [270], [279] Similarly, Nakamura and associates [268] observed that carbachol and EGF induced the ERKs in isolated rabbit parietal cells.

The functional relevance of ERK activation in the stomach has been the focus of continued investigation. Initial studies reported the divergent effect of EGF on gastric acid secretion. Under acute conditions, EGF had an inhibitory effect on acid secretion, whereas prolonged administration of EGF increased both basal and maximal acid secretion in vivo and acid production in isolated parietal cells in vitro. [269], [280], [281] Some studies suggested that these effects of EGF could be mediated by the activation of protein tyrosine kinases because they were fully reversed by the addition of protein tyrosine kinase inhibitors. [269], [280] In addition, Chew and associates [269] observed that inhibition of the chronic stimulatory effect of EGF by these agents was associated with a decrease in phosphorylation of a 44-kd protein identified as an ERK isoform. Taken together, these results suggested that both the ERKs and an unidentified protein tyrosine kinase were likely to be important in the regulation of the gastric parietal cells.

Our understanding of the function and physiological role of the ERK pathway has been significantly enhanced by the use of PD98059. To examine the importance of ERK activation in gastric acid secretion further, Takeuchi and colleagues [270] tested the effect of PD98059 on carbachol-stimulated uptake of [^{14}C]aminopyrine. Acute inhibition of the ERKs by PD98059 led to a small increase in [^{14}C]aminopyrine uptake and to complete reversal of the inhibitory effect of EGF on parietal cell activation induced by either carbachol or histamine. [270] In contrast, exposure of the cells to PD98059 for 16 hours reversed the chronic stimulatory effect of EGF on [^{14}C]aminopyrine uptake induced by carbachol, leading the investigators to conclude that whereas the acute effect of the ERKs on gastric acid secretion appears to be inhibitory, the activation of transcription factors and of early gene expression could produce chronic stimulatory effects (Fig. 13-9). [270] Further research has shown that the acute inhibitory effect of EGF on gastric acid secretion could be mediated by a PKC-dependent pathway. [265] Thus, because PKC has been shown to activate c-Raf, a kinase involved in the activation of the ERKs, [282] it is possible that the acute inhibitory effect of EGF on aminopyrine uptake could be mediated by a signaling cascade involving activation of PKC, c-Raf, and MEK, leading finally to induction of the ERKs. Furthermore, Kaise and associates [281] demonstrated that EGF is able to induce transcription of the a subunit of the H $^{+}$,K $^{+}$-ATPase gene through a novel EGF-response element (ERE) located between bases–162 and–156 (5'-GACATGG-3') relative to the cap site. This ERE is homologous to the 3' half-site of the c- *fos* serum response element. Accordingly, the authors suggested that the stimulatory effect of EGF on gastric acid secretion could be mediated by induction of the H $^{+}$,K $^{+}$-ATPase gene. [281] Although a direct link between induction of the ERKs and c- *fos* and stimulation of the H $^{+}$,K $^{+}$-ATPase gene has not been yet demonstrated, it is possible that this gene may contain specific DNA regulatory elements that receive input from signaling pathways involving the ERKs and c- *fos*.

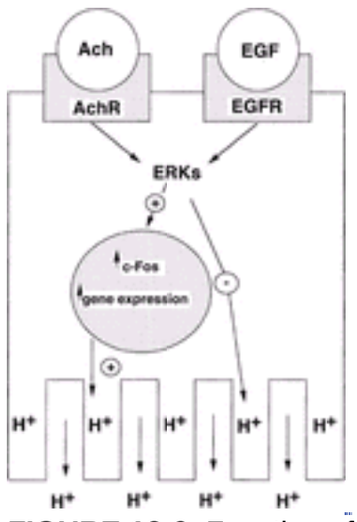

FIGURE 13-9. Function of the extracellular signal–regulated protein kinases (*ERKs*) in the gastric parietal cells. Both acetylcholine (*Ach*) and epidermal growth factor (*EGF*) induce a cascade of events in parietal cells that results in ERKs activation. Whereas the acute effect of the ERKs on gastric acid secretion appears to be inhibitory, the activation of transcription factors and of early gene expression may be responsible for their chronic stimulatory effects.

p38 kinase is another member of the MAPK family of protein kinases. [273] Like the MAPKs, p38 kinase is regulated by a multistep cascade of biochemical reactions. One of the first steps in the complex chain of events that leads to the induction of p38 kinase is the activation of Rac and Cdc42, small GTP-binding proteins belonging to the Rho family of GTPases. [273] These molecules are responsible for the induction of downstream protein kinases such as the dual-specificity kinases MKK3 and MKK6, which, in turn, phosphorylate p38 kinase on both tyrosine and threonine residues, leading to its activation. [273] Research indicates that p38 kinase is activated in response to both physical and chemical stress, cytokines, hematopoietic growth factors, neurotransmitters such as carbachol and isoproterenol, and CCK. [272], [273], [283], [284] Activation of p38 kinase has been linked to regulation of programmed cell death, skeletal muscle differentiation, organization of the cellular cytoskeleton, and phosphorylation of transcription factors. [273], [284], [285], [286] and [287] Pausawasdi and colleagues [272] used SB-203580, a specific p38 kinase inhibitor, [286] to investigate the regulation and the functional relevance of p38 kinase in carbachol-stimulated gastric acid secretion. These investigators have observed that SB-203580 dose-dependently potentiates carbachol induction of aminopyrine uptake in isolated and cultured canine gastric parietal cells, suggesting that p38 kinase has a negative regulatory effect on gastric acid secretion. [272]

Because p38 kinase is known to play an important role in the organization of the actin cytoskeleton, [273], [284] a possible mechanism of p38 kinase–induced inhibition of gastric acid secretion could involve changes in the actin cytoskeleton of the gastric parietal cells. The cytoskeleton is an important element in secretagogue-stimulated gastric acid secretion because, during this process, the parietal cell undergoes dramatic morphologic modifications that lead to the translocation of H^+,K^+-ATPase from cytoplasmic tubulovesicular structures to the apical plasma membrane. [18], [19] and [20] Accordingly, p38 induction could result in phosphorylation of cytoskeletal proteins and in significant changes in the organization of the actin cytoskeleton. These events could be responsible for alterations in the process of H^+,K^+-ATPase insertion into the parietal cell apical membrane and for the inhibition of gastric acid production.

Considering the lack of specific JNK inhibitors, it is unclear whether these kinases are involved in the process of gastric acid secretion. However, studies conducted in canine parietal cells have suggested that JNK activation may be an important step in the parietal cell response to stress and inflammation. [271]

Another signal transduction pathway that has been shown to play an important role in the mediation of some of the physiological actions of EGF is that involving phosphoinositide 3-kinase (PI_3-K) and protein kinase B/Akt. [287], [288], [289] and [290] Three major isoforms of Akt have been identified and described thus far. [288] Whereas Akt 1 and Akt 2 are ubiquitously expressed, Akt 3 appears to be expressed predominantly in the brain, heart, and kidney. [290] Activation of Akt is known to induce cellular growth and survival and to promote the expression and maintenance of highly differentiated cellular phenotypes. [287], [288], [291], [292] Furthermore, Akt has been shown to contribute to the regulation of vesicular trafficking and endocytosis. [292] Akt is homologous to the PKA and PKC families of protein kinases, [288], [293] and its activity is regulated by growth factors through the induction of PI_3-K. [288], [293] Phosphorylation of Akt appears to be critical for its activation. [288], [293], [294] The major phosphorylation sites required for activation of Akt 1 have been identified as threonine 308 and serine 473, which are the target of the phosphoinositide-dependent kinases 1 (PDK1) and 2 (PDK2), respectively. [288], [293] Similarly, Akt 2 and Akt 3 are also phosphorylated on serine and threonine residues (Thr309 and Ser474 in Akt 2 and Thr305 and Ser472 in Akt 3) by PDK1 and PDK2 (Fig. 13-10). [288], [290] Todisco and associates [295] reported that EGF regulates the expression of the H^+/K^+-ATP-ase a subunit gene through a signal transduction pathway that involves the activation of Akt. These findings suggest a novel role for Akt in the regulation of the secretory function of the gastric parietal cells.

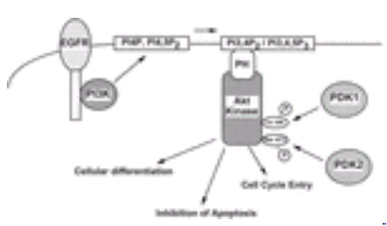

FIGURE 13-10. Activation and function of the Akt signal transduction pathway. Akt is a serine-threonine protein kinase involved in the regulation of numerous important cellular functions such as inhibition of apoptosis and stimulation of cellular growth and differentiation. Induction of Akt requires the recruitment of the kinase to the plasma membrane by phosphoionositide 3-kinase–mediated phosphorylation of membrane phospholipids and phosphorylation of Ser 473 and Thr 308 of Akt by the phosphoinositide-dependent kinases PDK1 and PDK2. PI4P, phosphatidylinositol-4-phosphate; $PI4,5P_2$, phosphatidylinositol-4,5-bisphosphate; $PI3,4P_2$, phosphatidylinositol-3,4-bisphosphate; $PI3,4,5P_3$, phosphatidylinositol-3,4,5-triphosphate; PH, pleckstrin homology domain.

The mechanism by which somatostatin inhibits parietal cells has been explored. [295] In the case of histamine-stimulated acid secretion, somatostatin appears to inhibit the generation of cAMP through an inhibitory guanine nucleotide–binding protein that regulates adenylate cyclase activity. However, somatostatin is also able to inhibit acid secretion induced by dibutyryl cAMP. The stimulatory effects of gastrin and carbachol are also inhibited without altering the turnover of membrane inositol phospholipids or the activation of PKC induced by these agents. Somatostatin appears to act on parietal cells at a site distal to the activation of the intracellular signal transduction cascades. This action may be mediated through the induction of protein dephosphorylation, the inhibition of cellular secretion, or some other yet undetermined mechanism. [296], [297] and [298] For example, somatostatin inhibits the induction of the early response gene c-*fos* in canine parietal cells stimulated by both carbachol and histamine. [299] As in many other physiological systems, c-*fos* plays an important role in cellular activation. Thus, the ability of somatostatin to inhibit c-*fos* expression may represent an important regulatory mechanism in gastric parietal cell physiology.

Regulation of Parietal Cell Genes

The stimulation of parietal cells induces several cellular events, including morphologic transformation, rapid changes in enzyme location and activity, and opening of ion channels. The resting parietal cell contains a collapsed canalicular system and cytoplasmic tubulovesicles containing the gastric proton pump, H^+,K^+-ATPase. The stimulated cell rapidly develops a richly interdigitating intracellular canalicular system bulging with microvilli with concomitant loss of cytoplasmic tubulovesicles (Fig. 13-11). [19], [300] The microvilli have a central cytoskeletal core of actin filaments stabilized by other proteins. [301] These filaments appear to mediate the fusion of tubulovesicles with the canalicular system. This fusion translocates H^+,K^+-ATPase from vesicular membranes to the canalicular membrane, where it actively pumps H^+ ions in exchange for K^+ (Fig. 13-12). [302], [303] For each proton that is secreted, an intracellular OH^- ion is generated. This alkaline challenge is handled by

carbonic anhydrase II (CAII)–mediated conversion of OH^- to HCO_3^-, which is exchanged for Cl^- at the basolateral membrane.

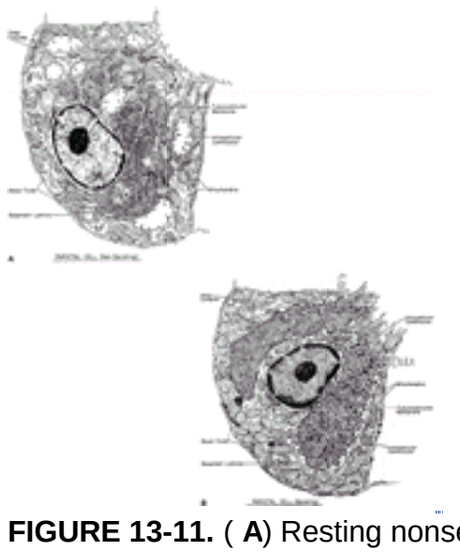

FIGURE 13-11. (**A**) Resting nonsecretory parietal cell. (**B**) Stimulated acid secretory parietal cell. The stimulated cell develops a richly interdigitating intracellular canalicular system that bulges with microvilli. There is a concomitant loss of cytoplasmic tubulovesicles. (From ref. [20].)

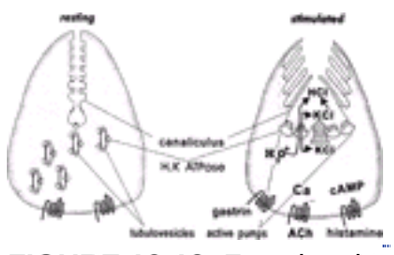

FIGURE 13-12. Functional transformation of a secreting parietal cell. This model illustrates a resting parietal cell (**left**) with collapsed secretory canaliculi and cytoplasmic tubulovesicles expressing H^+,K^+ATPase and a stimulated parietal cell (**right**) with formed secretory canaliculi expressing active H^+,K^+-ATPase pumps. (Adapted from ref. [343].)

There are many proteins involved in generating and secreting gastric acid. Stimulation of parietal cells presumably involves the activation of nascent proteins and the induction of the genes responsible for these effector molecules. The effect of acid secretagogues on the genes coding these enzymes and structural proteins has been analyzed.

The induction of specific gene transcription has been studied in isolated canine parietal cells. [304] Significant increases in CAII RNA levels were induced by carbachol, gastrin, and histamine. Maximal stimulation was reached within 20 minutes for histamine and carbachol and within 60 minutes for gastrin. To determine whether the observed increases in mRNA levels were the result of increased transcription or decreased degradation, nuclear run-off experiments were performed. They revealed increased transcription within 15 minutes for each agent. H^+,K^+-ATPase mRNA levels were also induced in the same fashion as for CAII. [305] This coordinated induction of both enzymes could be blocked with the competitive inhibitors of each secretagogue.

Because CAII appears to be the catalyst of the reaction responsible for the elimination of OH^- produced in the generation of H^+, increased transcription of CAII RNA in stimulated parietal cells could result as a secondary effect of the induction of H^+,K^+-ATPase. To explore this possibility, parietal cells were pretreated with omeprazole, an agent known to inactivate H^+,K^+-ATPase irreversibly. [306] Under these circumstances, carbachol still induced CAII RNA, with the same kinetics as in untreated cells. Carbachol appears to stimulate CAII gene expression without dependence on OH^- ion generation by H^+,K^+-ATPase.

Secretagogues stimulate actin expression. Although actin often is used as a control or housekeeper gene for such studies because its expression tends to remain constant, in the case of the parietal cell, actin plays a crucial role in the acid secretory process. The induction of actin gene expression may serve as a particularly useful marker for acid secretion instead of using H^+ generation. This type of analysis can elucidate the effect of acid secretagogues on parietal cell function at the molecular level.

Muraoka and associates [307] undertook an extensive evaluation of the regulatory elements controlling H^+,K^+-ATPase gene expression. They transfected cultured canine gastric parietal cells with reporter gene constructs composed of the first exon and various lengths of the 5'-upstream regulatory region of the canine H^+,K^+-ATPase a subunit ligated to a luciferase reporter plasmid. Their data indicate that basal transcriptional activity of the H^+,K^+-ATPase gene is mediated through binding of the nuclear transcriptional factor SP1 to the 5'-GCTCCGCCTC-3' nucleotide sequence residing between bases –47 to–38 relative to the putative cap site. [307]

Shigehiko and Maeda and their colleagues [308], [309] identified regions within the promoters of both the a and ß subunits of the H^+,K^+-ATPase gene that were recognized by gastric-specific nuclear proteins. These investigators subsequently cloned two novel nuclear proteins, named GATA-GT1 and GATA-GT2, from pig gastric mucosa. These bound specifically to the sequences (G or C)RR(G or C)NGAT(A or T)RY, in which R and Y are unspecified purine and pyrimidine bases, respectively. GATA-GT1 and GATA-GT2 belong to the family of the GATA transcription factors that are known to play an important role in tissue-specific gene expression. [309] Northern blot analysis of several different tissues revealed that these nuclear proteins were expressed predominantly in the gastric mucosa and to a lesser degree in the intestine, but not in the brain, heart, liver, kidney, spleen, or lung. [308] Maeda's group speculated that these transcription factors were responsible for the specific expression of the H^+,K^+-ATPase gene in the gastric mucosa.

Acid Secretory Processes

The gastric epithelium secretes a fluid of almost isotonic HCl through an active transport process. Acid is secreted at a pH of 0.8, but the parietal cell cytosolic pH is about 7.2. The parietal cell alone is responsible for this remarkable H^+ ion concentration gradient of 2.5 million–fold. Significant amounts of mitochondrial energy are required when the parietal cell is signaled to transform from a relatively quiet resting state to an actively secreting state. Dramatic changes in cellular membranes, cytoskeletal architecture, membrane ion conductances, and ATPase activities are only a portion of the events that accompany cell activation. Equally impressive is the compressed time frame over which these changes occur. It has been suggested that the family of isoenzymes, kinase/phosphocreatine (CK/DCr), which catalyze the exchange of high-energy phosphate groups between phosphocreatine and ADP, are involved in parietal cell transformation. The CK/DCr system couples to the K^+-ATPase [310] and the Ca^{2+}-ATPase found in sarcoplasmic reticulum. [311] The isoform BB-CK (B, brain) has been identified in the parietal cells of different species, [312] and this enzyme appears to colocalize with H^+,K^+-ATPase in purified vesicles prepared from porcine parietal cells. These data suggest that H^+,K^+-ATPase may obtain the required ATP generated by the BB-CK colocalized in the same cellular compartment. [313]

The stimulation of parietal cells induces the formation of a dense apical meshwork of intracellular canaliculi packed with long microvilli. [314] The apical cell membrane surface area increases five- to tenfold after stimulation. This increase coincides with the disappearance of most of the cytoplasmic tubulovesicles seen in the resting

parietal cell. The mechanism of this increase in surface area appears to be the fusion of tubulovesicles and apical membrane. Immunocytochemical studies using a monoclonal antibody to H^+,K^+-ATPase demonstrate the translocation of the enzymes from tubulovesicles in the resting cell to the apical membranes in the stimulated cells, supporting the concept of secretagogue-induced membrane fusion and enzyme redistribution. [302] The fusion of tubulovesicles with the apical membrane is directed by cytoskeletal microfilaments composed of actin and other regulatory proteins. Membranes are recycled back to the tubulovesicles as cells return to the resting state, a process that also appears to be mediated by actin-containing microfilaments. [301] These microfilaments anchor at regularly spaced intervals in the apical membrane and do not appear to associate with resting tubulovesicles. Cytochalasins, agents that inhibit actin polymerization, inhibit acid secretion. [315] Colchicine and vinblastine also are capable of inhibiting acid secretion. [316] These studies indicate the essential importance of actin-mediated H^+,K^+-ATPase translocation in the initiation of acid secretion.

Forte and colleagues [317] have analyzed in detail the biochemical and functional properties of actin in the gastric parietal cells. In a series of elegant studies, these authors have demonstrated that most parietal cell actin exist in the filamentous form (F-actin), which is an important component of the apical cell microvilli, whereas only a small fraction appears to be in the monomeric state (G-actin).

In some systems of regulated vesicular transport, F-actin is known to form a meshwork of filaments just beneath the cell surface, creating a barrier for the recruitment of vesicles to the apical plasma membrane. According to this model, F-actin depolymerization to the monomeric G form facilitates vesicle translocation. In contrast to this theory, Forte's group [317] did not detect any significant changes in the steady-state ratio of F-actin and G-actin during the process of parietal cell activation by the gastric acid secretagogues. Thus, although these observations do not exclude the possibility that rapid exchange between F-actin and G-actin may occur, it appears that during induction of gastric acid secretion, the parietal cell maintains actin in a highly polymerized state. [317]

These investigators [318] also demonstrated that the parietal cells contain pools of actin that display considerable functional differences. Using latrunculin B, a compound known to bind and inhibit the function of G-actin, they demonstrated that whereas G-actin is important for lammellipodia formation, F-actin, which is less sensitive to latrunculin B, appears to play a crucial role in the process of secretagogue-stimulated gastric acid secretion. In fact, alteration of F-actin function by either a high dose of latrunculin B or by cytochalasin D, an agent known to fragment F-actin filaments, resulted in inhibition of gastric acid secretion. [318]

Hanzel and colleagues [319] have identified one of the membrane proteins involved in parietal cell cytoskeletal transformation as ezrin, an 80-kd membrane protein found in several cell types. Ezrin is associated with the actin filaments in the microvilli of stimulated parietal cells and is phosphorylated during cAMP-mediated stimulation. Actin, an important structural protein for cellular scaffolding, is expressed as several isoforms, including the ß and ? subtypes found in parietal cells. [320] In a series of elegant experiments using ultrastructural immunocytochemistry and biochemical protein purification, Yao and associates [321] confirmed that ß-actin is localized in canalicular microvilli and in the apical portion of parietal cells. [321] The authors also reported the selective association of ezrin with ß-actin. These important observations have begun to shed light on the potential role of ezrin in parietal cell secretion. Moreover, they support the novel concept that actin isoforms may be distributed in different cellular domains, exerting differential functions by association with isoform-specific actin-binding proteins.

The molecular events regulating the dramatic recycling of H^+,K^+-ATPase–containing membranes have been the focus of intense investigative efforts. Study of other cell systems has begun to elucidate the important factors in the secretion process. One such family of factors has been the low-molecular-weight GTP-binding proteins. [322] The members of this superfamily of proteins have certain characteristics: molecular weights of 20 to 30 kd, significant homology with the oncogenic peptide Ras, ubiquitous expression, and involvement in a host of cellular functions, including vesicle trafficking. Several members of the Rab category of Ras proteins are expressed in gastric parietal cells. [323], [324], [325], [326] and [327] Rab 25, Rab 324, and Rab 11 (previously thought to be Rab 2) [325], [326] have been identified in rabbit parietal cells. More importantly, these specific proteins cosegregate with a-H^+,K^+-ATPase during parietal cell activation, suggesting a role for these small GTP-binding proteins in membrane recycling. [325], [327] The importance of Rab 11a in the process of parietal cell activation has been demonstrated by Duman and colleagues. [328] Transduction of isolated and cultured rabbit parietal cells with an adenoviral vector expressing a dominant negative Rab 11a gene led to a block in the translocation of the H^+,K^+-ATPase to the apical plasma membrane and to the inhibition of gastric acid secretion. [328]

Developments in the field of cell biology have shed light on the molecular pathways important in cellular secretory processes. Work in neuronal systems and yeast have demonstrated the potential involvement of highly conserved protein complexes in vesicle fusion and docking, such as the soluble *N*-ethylmaleimide–sensitive factor attachment protein receptors (SNAREs). [329], [330] These can be vesicle-related (v-SNARE) or target membrane–related (t-SNARE), binding to each other to provide a molecular framework for the attachment of soluble factors. [331] Members of these protein families include VAMP-2 (vesicle-associated membrane protein-2, identified as a v-SNARE in brain), and the syntaxin 1A/1B and SNAP-25 (synaptosome-associated protein), which are categorized as t-SNAREs and are thought to play a role in the recycling of synaptic vesicles. [332] The secretory carrier membrane proteins or SCAMPs found in pancreatic zymogen granules and synaptic vesicles may also play a role in vesicle recycling. Calhoun and Goldenring [327] have explored the potential role of these novel trafficking-related proteins in the parietal cell secretory process. The involvement of VAMP-2, SCAMP, Rab 11, and Rab 25 in membrane trafficking was determined by immunoisolation of highly purified parietal cell tubulovesicles. These investigators reported that VAMP-2, SCAMPs, and the two Rab proteins were present on H^+,K^+-ATPase–containing tubulovesicles and suggested that Rab proteins take part in the assembly of the SNARE complex. These observations support a theory originally proposed by Forte that parietal cell tubulovesicles may be derived from a cell surface recycling system.

Other studies have demonstrated that the cytoskeletal proteins actin, spectrin, and ankyrin co-purify with the H^+,K^+-ATPase from parietal cell microsomal membranes and cosegregate with H^+,K^+-ATPase in resting and secreting parietal cells. Ankyrin may mediate the interaction of H^+,K^+-ATPase with the structural membrane proteins actin and spectrin and may thereby function to maintain the polarized distribution of the enzyme to the apical portion of the parietal cell. Festy and colleagues [333] screened a cDNA bank of rabbit gastric fundic mucosa by two-hybrid assays and demonstrated that the rabbit H^+,K^+-ATPase binds through its amino-terminal end to the spectrin binding domain of ankyrin III. The two proteins bind directly to one another, constituting an important anchoring system for the H^+,K^+-ATPase to the plasma membrane. [333]

Studies of H^+,K^+-ATPase activity have been performed using membrane vesicles obtained from resting and stimulated parietal cells. [314] Vesicles from light microsomal membrane fractions, presumably containing the tubulovesicles, exhibit most of the H^+,K^+-ATPase activity in resting parietal cells. [334] Heavier membrane vesicles associated with microfilaments contain the majority of H^+,K^+-ATPase activity in stimulated parietal cells. The membranes of these acid-secreting vesicles have a K^+-Cl^- cotransport system that is lacking in membrane preparations from resting cells. [335]

Studies have confirmed that H^+,K^+-ATPase requires extracellular K^+ for electroneutral exchange with H^+. [336], [337] It appears that concomitant with membrane or enzyme translocation, associated K^+ and Cl^- conductances are activated. [335] Opening of this conductance as it moves from tubulovesicles to the canalicular membrane allows H^+,K^+-ATPase to generate H^+. The K^+-Cl^- conductance is intimately associated with H^+,K^+-ATPase, and its function is required for acid secretion. Several other ion channels are involved in maintenance of cellular homeostasis during acid secretion. A schematic representation of parietal cell conductances and pumps is shown in Figure 13-13. Each H^+ ion that is generated results in formation of an intracellular OH^- ion, which reacts with CO_2 in a reaction catalyzed by CAII to form HCO_3^-. A mechanism located on the basolateral membrane permits the exchange of intracellular HCO_3^- for extracellular Cl^-. This Cl^- provides part of the intracellular pool for transport by the apical K^+-Cl^- conductance system. Several types of Cl^-/HCO_3^- exchangers have been described. The one expressed in parietal cell basolateral membranes has been identified as the anion exchanger 2. [338] Parietal cells also express the Na^+-H^+ exchanger NHE1 on basolateral membranes, but to a much lesser degree than in mucous neck and chief cells. Moreover, the level of CAII expression appears to correlate with the level of NHE1 expression in these same cells. The low level of NHE1 in parietal cells may suggest that another exchanger such as NHE4 [339] may play a more important role in the acid secretory process. Although not shown in Figure 13-13, a Na^+-HCO_3^- cotransporter has also been located on parietal cells. [340] The role of this cotransporter in maintaining parietal cell homeostasis has not been determined.

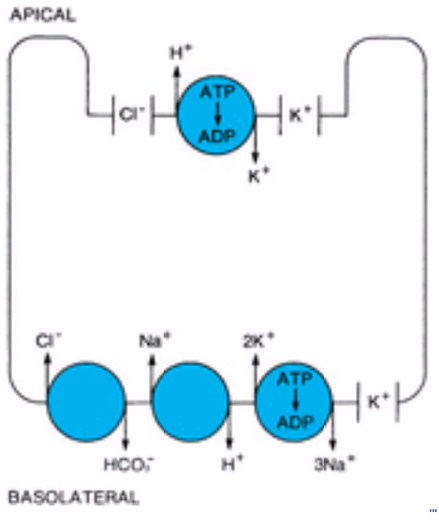

FIGURE 13-13. Ion transport pathways in parietal cells. The apical membrane contains the H $^{+}$,K $^{+}$-ATPase pump and K $^{+}$ and Cl $^{-}$ conductances. The basolateral membrane also has a K $^{+}$ conductance, Cl $^{-}$/HCO $_{3}$ $^{-}$ exchange, Na $^{+}$/H $^{+}$ exchanges, and Na $^{+}$,K SUP>+-ATPases to maintain cellular homeostasis during secretory and resting states.

A second mechanism located on the basolateral cell membrane provides for exchangers of extracellular Na $^{+}$ for intracellular H $^{+}$. The Na $^{+}$ or H $^{+}$ and Cl $^{-}$/HCO $_{3}$ $^{-}$ exchangers are coupled functionally as cellular pH monitors handling excess acid or base loads, respectively. Although physically separate, these exchange mechanisms act as a functional basolateral Na $^{+}$ and Cl $^{-}$ cotransport mechanism. The basolateral membrane also has a K $^{+}$ conductance, which allows intracellular K $^{+}$ to follow the concentration gradient. K $^{+}$ uptake by the parietal cell can occur by means of a basolateral Na $^{+}$,K $^{+}$-ATPase or Na $^{+}$ pump, which appears to translocate three intracellular Na $^{+}$ ions for two extracellular K $^{+}$ ions. The net effect of these ion channels is to maintain cellular homeostasis in the face of HCl secretion. The osmotically active HCl generated in the canalicular lumen results in the flow of H $_{2}$O across the cell. This flow leads to the formation of gastric fluid associated with acid secretion. The gastric H $^{+}$,K $^{+}$-ATPase or H $^{+}$ ion pump has been studied in detail. [341] This membrane protein is a member of the P-ATPase family that includes Na $^{+}$- or K $^{+}$- and Ca $^{+}$-ATPases. Structural homology between these family members suggests that they probably diverged from a common ancestor molecule. [342]

Significant progress has been made toward understanding the structural characteristics of the gastric H $^{+}$,K $^{+}$-ATPase. [343] This critical enzyme in parietal cell biology consists of two subunits, a and ß. The a subunit is larger. The amino acid sequences of the rat, porcine, and rabbit enzymes have been deduced from the corresponding cloned cDNA structures. [344], [345], [346] and [347] Interspecific conservation of the a subunit is greater than 97%; the protein length ranges from 1033 amino acids in the rat to 1035 residues in the rabbit. The gene corresponding to the human a subunit has been cloned and sequenced; [326] it contains 22 exons and has a corresponding deduced protein length of 1035 amino acids.

Several experimental procedures have been used to establish the secondary structure of the a subunit (Fig. 13-14). Initial hydropathy plots of the deduced amino acid sequence suggested that this protein was anchored by seven to nine potential transmembrane domains with a large cytoplasmic site containing an ATP-binding region. [344] Although still controversial, more recent studies capitalizing on a combination of proteolytic cleavage of the enzyme, in vitro translation experiments, and inhibitor-binding studies suggest that the a subunit may contain ten transmembrane segments with a large cytoplasmic domain. [343] The cytoplasmic domain is presumed to include ATP-binding, phosphorylation, and energy transduction sites. Labeling studies suggest that the proton pump inhibitors omeprazole, lansoprazole, and pantoprazole bind to the cysteine residues located at positions 813 and 822. These residues lie near a predicted extracytoplasmic loop between transmembrane domains 5 and 6, and they are thought to be essential for proton pump inhibition of H $^{+}$,K $^{+}$-ATPase. [348], [349] and [350] Elucidation of the tertiary structure of this enzyme requires high-resolution crystallography studies.

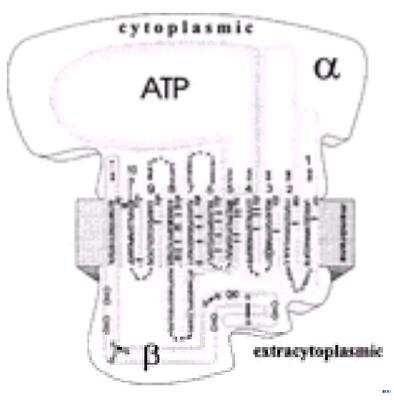

FIGURE 13-14. Model of the two-dimensional structure of the H $^{+}$,K $^{+}$-ATPase illustrating the a and ß subunits. The a subunit contains ten putative transmembrane domains. Hydrophilic amino acids within the postulated membrane anchored domains are shown. (Adapted from ref. [343].)

The cDNA of the H $^{+}$-K $^{+}$-ATPase ß subunit has been isolated and sequenced from several species, including rabbit, [351] rat, [352], [353] and [354] hog, [355] mouse, [356] and human. [357] The average length of this subunit ranges from 290 to 294 amino acids, and hydropathy plots suggest a single transmembrane domain. Although the role of the ß subunit has not been established, several lines of investigation suggest that it may be important for assembly and stabilization of the functional a subunit. Insight into the functional role of the H $^{+}$,K $^{+}$-ATPase ß subunit has been gained by studies conducted in transgenic animals. [358] Courtois-Coutry and colleagues [358] noted that the cytoplasmic tail of the H $^{+}$,K $^{+}$-ATPase ß subunit displays a four-residue sequence homologous to tyrosine-based endocytosis signals. Mutation of this motif's tyrosine and expression of the mutated H $^{+}$,K $^{+}$-ATPase ß subunit in transgenic animals led to constitutive secretion of acid and to the continuous expression of the mutated H $^{+}$,K $^{+}$-ATPase at the cell surface. Thus, the H $^{+}$,K $^{+}$-ATPase ß subunit appears to play an important role in the process of H $^{+}$,K $^{+}$-ATPase internalization and in the termination of gastric acid secretion. [358]

The timing of the association between a and ß subunits of the H $^{+}$,K $^{+}$-ATPase and the initial steps in cellular trafficking for this enzyme have been determined. Crothers and associated [359] used the method of in vivo protein metabolic labeling in conjunction with subcellular fractionation to examine these issues in rabbit parietal cells. These investigators observed that the a and ß subunits associated early in the endoplasmic reticulum. Moreover, the newly synthesized H $^{+}$,K $^{+}$-ATPase migrated sequentially through at least three distinct membrane pools. Future studies should reveal the impact of pharmacological manipulation on synthesis and trafficking of this important enzyme in parietal cells.

H $^{+}$,K $^{+}$-ATPase can be inactivated completely by omeprazole, lansoprazole, rabeprazole, and pantoprazole. These compounds become cyclic in the presence of acid and react with available sulfhydryl groups to form a covalent interaction that irreversibly inactivates the enzyme. The blockade of proton transport is virtually complete and has been used clinically as described in Chapter 66.

OTHER GASTRIC SECRETORY PRODUCTS

Histamine

Histamine plays a critical role in regulating gastric acid secretion through the activation of the parietal cell H_2 receptor. There is evidence that histamine mediates in part the stimulatory effect of other secretagogues (e.g., gastrin, carbachol) on parietal cells. The relative importance of the two histamine-containing cell types found in the stomach (i.e., mast cells, ECL cells) in regulating gastric acid secretion has been the source of some debate. The ratio of the two cell types in gastric mucosa depends on the species examined. Early studies using isolated mast cells obtained from canine gastric mucosa indicated that gastrin and cholinergic agents did not stimulate histamine release, and this observation was used to support the contention that these secretagogues induced acid secretion by direct action on parietal cells rather than by stimulating histamine release. However, these data were at odds with studies demonstrating that pentagastrin stimulated the release of histamine from gastric mucosal preparations. Chuang and colleagues [360] shed light on this controversy by successfully isolating and characterizing a population of non–mast cells from canine oxyntic mucosa that contain histamine and have the morphologic characteristics of ECL cells. Gastrin nd carbachol stimulate histamine release from ECL cells when they are isolated and cultured. It is possible that the secretagogue actions of gastrin and carbachol can be mediated, at least in part, through the action of histamine. Such studies reinforce the concept that regulation of gastric acid secretion involves a complex interplay between multiple pathways and receptors.

Development of the isolated ECL cell model has facilitated the study of this important source of gastric histamine. Gastrin-stimulated histamine release from ECL cells occurs through the CCKB/gastrin receptor, which is coupled to a Ca^{2+} signaling pathway. [361], [362] Histamine release is also stimulated by factors that increase cAMP, such as agonists of the ß-adrenergic receptor, forskolin, and pituitary adenylate cyclase–activating peptide (PACAP). [361], [363] Research indicates that the PACAP type I receptor subtype regulates histamine release and $[Ca^{2+}]_i$ mobilization. [364] As one would anticipate, ECL cells are also under inhibitory restraints. Somatostatin binds to an SSTR2 receptor subtype, inhibiting histamine release and Ca^{2+} signaling. [365] Histamine appears to regulate its own release through an H_3 receptor subtype located on ECL cells, thus creating an autocrine feedback loop. [361] Peptide YY has also been observed to inhibit histamine release through a Y_1 receptor subtype. [363]

Histamine is generated in ECL cells by decarboxylation of histidine by the enzyme histidine decarboxylase (HDC). [366], [367] HDC is transcriptionally activated by both gastrin and acetylcholine. Studies by Hocker and colleagues [368], [369] have demonstrated that gastrin stimulates HDC transcription through a PKC- and MAP kinase–dependent pathway. In view of reports demonstrating the development of ECL hyperplasia and gastric carcinoid tumors in animals receiving large doses of proton pump inhibitors, there has been increased interest in understanding the regulation of ECL cell proliferation. [370] Both in vivo and in vitro studies have demonstrated that gastrin has a direct proliferative effect on these cells, thus supporting the concept that hypergastrinemia resulting from potent acid suppression is responsible for increased ECL proliferation in these animal models. [371], [372], [373], [374], [375], [376] and [377] The relevance of these observations to human physiology is unclear, considering the studies demonstrating that the human ECL cell is terminally differentiated and is not capable of undergoing proliferation. [378]

An attempt to determine the relative importance of histamine-containing mast cells and ECL cells in the regulation of gastric acid secretion was made by Stechschulte and colleagues. [379] These investigators observed that mast cell–deficient mice had about 50% of the normal quantity of gastric histamine in addition to diminished basal and stimulated acid secretory activity. They concluded that non–mast cell histamine may account for only part of the basal and stimulated gastric secretory response.

Pepsinogen

The secretion of pepsinogen by the gastric mucosa occurs in response to food ingestion, as described by Langley in 1886. [380] Pepsinogens are inactive proenzymes that are autocatalytically cleaved under acidic conditions to generate their active form, pepsin. The proenzyme is synthesized in exocrine chief cells found at the base of oxyntic glands and in mucous neck and mucous cells in cardiac, oxyntic, and pyloric glands. [381] Chief cells package pepsinogen in apical granules, where it is stored until the cells are stimulated. Stimulation of chief cells by various secretagogues induces exocytosis of granule contents into the glandular lumen. Concomitant stimulation of parietal cells provides the acidic lumenal conditions needed for rapid production of pepsin, the major acid protease activity in the stomach. Pepsin is a member of the group of proteolytic enzymes classified as aspartic proteases. [382] Members of this enzyme family are found in an array of organisms extending from plants and retroviruses to humans. The stomach is the best source of aspartic proteases in mammals, and pepsin is the representative member.

This protease initiates protein digestion and is particularly active in proteolysis of collagen, a major protein component of meat. Peptides generated by the proteolytic activity of pepsin act as signals for secretion of digestive hormones such as gastrin and CCK. These peptide signals initiate the coordinated digestive response necessary for the absorption of nutrients.

Pepsinogens have been electrophoretically separated into seven isozymogens. [383] Five fractions (i.e., fractions 1 through 5) that migrate toward the anode rapidly are immunologically similar and have been named group I pepsinogens (PGI or PGA). Group II pepsinogens (i.e., fractions 6 and 7) migrate slightly slower in gels and are antigenically similar (i.e., PGII, PGC, or progastricsin). Both groups are active in acidic conditions (pH 2–3.5) and are inactivated at a pH higher than 5. Despite these similarities, many biochemical and immunochemical differences between PGI and PGII are the subject of intense investigation.

The distribution of the two pepsinogen groups in gastrointestinal tissues varies. Although both groups are found in the gastric body, group I pepsinogen being the most abundant, only group II pepsinogens are found in the gastric antrum, proximal duodenum, and the Brunner glands. The presence of different isozymogens could be the result of multiple gene loci, multiple alleles at a single gene locus, alternate posttranscriptional processing of RNA, or posttranslational modifications of primary gene products. Genetic analysis has revealed that the two groups have different gene loci. A human pepsinogen I gene has been isolated, sequenced, and localized to chromosome 11q12-13. [384], [385] and [386] Analysis of its locus indicates the presence of multiple pepsinogen I genes. [387] This type of multigene protein polymorphism is rare and suggests that the pepsinogen gene complex is undergoing evolutionary gene duplication and selection. Other mechanisms of isozymogen diversity, such as posttranslational modification, may also be active.

Taggart and colleagues [388] isolated and sequenced the cDNA and gene coding human pepsinogen II (PGC, progastricsin). Analysis of the genomic clones suggests there is a single *PGC* gene located on chromosome 6. Structural comparisons between PGI (i.e., PGA) and PGII (i.e., PGC) demonstrate that they share 60% nucleotide and 50% amino acid identity. No immunologic cross-reactivity was detected between the two groups of human pepsinogens A and C. This lack of immunologic cross-reactivity may result from divergent evolution of sequences located on the surface of the enzymes, in contrast to the highly conserved sequences at the biologically active sites of the zymogen located within the binding cleft of the enzyme.

The stimulation of pepsinogen secretion has been analyzed in intact animals, gastric mucosal preparations, gastric glands, and isolated chief cells. Despite the use of different species in these models, certain generalizations about the mechanisms of chief cell activation are supported by the data. Acetylcholine and its analogs appear to stimulate chief cells directly through muscarinic receptors. [389], [390] Other neuronal mediators, such as gastrin-releasing peptide, may also mediate vagal stimulation of pepsinogen secretion. [391] The adrenergic agonist isoproterenol has a stimulatory effect in vitro, [392] although the physiological significance of this effect has not been demonstrated in intact animals. The ability of histamine to stimulate pepsinogen secretion remains controversial, and conflicting data preclude any definitive conclusion. [381]

Although gastrin stimulates chief cells only weakly, CCK-8 is a potent stimulus of pepsinogen secretion in vitro. [393] The finding that peripherally administered CCK in vivo only weakly stimulates pepsinogen secretion may reflect CCK's function as a neurocrine, rather than endocrine, mediator of chief cell stimulation. Alternatively, it may reflect the mixed effects of a direct stimulatory effect on chief cells and an indirect inhibitory effect such as could be mediated by CCK-induced somatostatin release.

Secretin has been reported to stimulate pepsinogen secretion in vivo and in vitro. [393], [394] The intracellular signal transduction mechanisms activated by the various chief cell secretagogues have been analyzed. Secretin, isoproterenol, and vasoactive intestinal polypeptide stimulate pepsinogen release through cell surface receptors linked to the activation of adenylate cyclase and cAMP formation. [381] Several peptide hormones including somatostatin, neuropeptide Y, and peptide YY have been found to inhibit pepsinogen release from isolated chief cells through a pertussis toxin–sensitive mechanism. Other secretagogues, such as cholinergic agonists and CCK, stimulate pepsinogen release in chief cells by activating the IP_3 and $[Ca^{2+}]_i$ signaling pathway. [395] and [396] Activation of the chief cell muscarinic receptor leads to a modest increase (15%) in PKC enzyme activity while stimulating a threefold increase in pepsinogen secretion. [397] Of interest, doses of carbachol capable of inducing pepsinogen release did not alter PKC activity, suggesting that other pathways independent of PKC, such as changes in cytosolic Ca^{2+}, may be sufficient for enzyme release. However, a maximal stimulatory effect of carbachol on pepsinogen release was achieved only at doses that also led to PKC activation. From these data, it appears that PKC, although not essential for enzyme release, may be involved in modulating the maximal response achieved by activation of the

muscarinic receptor.

Further insight into the postreceptor events involved in secretagogue-mediated pepsinogen secretion have been gained. The involvement of the Ca^{2+} signaling pathway as an important mechanism for chief cell activation has been reaffirmed. [398] Moreover, it appears that the increase in cytosolic Ca^{2+} is important for the ability of ligands such as CCK-8 to potentiate the stimulatory effect of agents such as secretin, which activates cAMP production in chief cells. This potentiation in pepsinogen release occurs independent of the Ca^{2+} source (intracellular or extracellular). As is the case in other cell systems, the Ca^{2+}/CaMKII is also involved in Ca^{2+}-mediated pepsinogen secretion. [399] Protein phosphatase-2B (calcineurin) is a Ca^{2+}/calmodulin-dependent serine/threonine protein phosphatase involved in signaling of cells from a variety of tissues. [400], [401] Raufman and colleagues [402] demonstrated that gastric chief cells express this phosphatase and that it modulates Ca^{2+}-induced potentiation of the cAMP signaling pathways at the level of adenylate cyclase. [403] Moreover, this specific phosphatase appears to be involved in Ca^{2+}-induced pepsinogen secretion by facilitating phosphorylation of a specific 55-kd cytoskeletal protein, which, in turn, regulates exocytosis.

As in gastric parietal cells, Rab proteins appear to be involved in chief cell intracellular trafficking. [404], [405] Specifically, Rab3 proteins appear to regulate pepsinogen secretion at one of the final steps of exocytosis. [404] Chief cells also express two isoforms of Rab-GDP-dissociation inhibitor (Rab-GDI), [405] which are proteins that, in addition to inhibiting the dissociation of GDP from Ras, inhibit the binding of GTP to GDP-bound Rab, thus removing Rab proteins from membranes. [406], [407] and [408]

The mechanism by which PKC modulates (i.e., augments) Ca^{2+}-induced pepsinogen secretion is unclear. A member of the family of proteins known as MARCKS (myristoylated alanine-rich C kinase substrate) has been suggested to play an important role in mediating the cross talk between PKC and Ca^{2+} calmodulin-stimulated pepsinogen secretion. [409] Specifically, a MARCKS protein may serve as a Ca^{2+}/calmodulin buffer, which, on phosphorylation, releases calmodulin from membranes and makes it available to bind and activate other signaling proteins such as Ca^{2+}/calmodulin kinase II and calcineurin. [410] In a series of immunologic and biochemical studies, Raufman's group presented evidence to suggest that an acidic 72-kd-protein (pp72) serves as a MARCKS protein in chief cells. It appears that PKC-induced phosphorylation and Ca^{2+}/calmodulin binding compete for similar regions on pp72. Therefore, PKC may enhance Ca^{2+}-induced pepsinogen secretion by phosphorylating MARCKS, decreasing its affinity for calmodulin, and facilitating activation of downstream targets important for pepsinogen secretion such as Ca^{2+}/calmodulin kinase II and calcineurin. [410]

An additional signaling pathway involved in pepsinogen secretion is the nitric oxide (NO) system. [411], [412] Gastric chief cells express a Ca^{2+}/calmodulin–dependent NO synthase that initially was observed to mediate pepsinogen secretion stimulated by leukotrienes (LTB_4, LTC_4, LTD_4, and LTE_4). [411] This pathway also appears to be involved in the action of Ca^{2+}-dependent agonists such as carbachol, gastrin, CCK, thapsigargin, and Ca^{2+} ionophore.

Pepsinogens are found in the gastric secretions, serum, urine, and seminal fluid. [413], [414] Their physiological role in nongastric fluids is unknown. Measurement of pepsinogens in these fluids is performed with specific radioimmunoassays. [413] Attempts to correlate serum pepsinogen groups with risk of peptic ulcer disease have proven to be of modest clinical value (see Chapter 66). Azuma and colleagues, [415] studying Japanese patients with gastric ulcers, found no genetic polymorphism for the pepsinogen A gene, but observed a 100-base pair insertion-deletion restriction fragment–length polymorphisms in the pepsinogen C gene. Genotypes that contained the small fragment were significantly more common in patients with gastric body ulcers than in patients with angular or antral ulcers or in controls. The clinical implication of this interesting observation has not been determined. Advances in our understanding of pepsinogen physiology and pathophysiology may lend these measurements new clinical significance.

Mucus

Gastric epithelium is partially protected from acidic autodigestion by a mucous gel that covers the entire surface of the stomach (for further discussion of gastric mucosal defense mechanisms, see Chapter 66). This gel acts as a barrier, protecting the gastric mucosa from acid, pepsin, bile salts, alcohol, and other injurious agents. The barrier consists of an unstirred layer of mucus, HCO_3^-, surface phospholipids, and water. A prominent pH gradient extending from the lumen (pH 2) to the epithelial cell surface (pH 7) is maintained by this gel. [416] Bhaskar and colleagues [417] shed light on the physical-chemical basis for the barrier function of gastric mucus. In a series of elegant in vitro experiments, these researchers demonstrated that injection of HCl through solutions of porcine gastric mucin produces viscous fingering patterns that depend on pH, mucin concentration, and acid flow rate. If the pH is greater than 4, discrete fingers are observed; at a pH of less than 4, these fingers are not seen, and HCl does not penetrate the mucin solution. Acid secreted by the gastric gland that exists in a pH 5 to 7 environment can penetrate the mucus gel layer through narrow fingers. In contrast, HCl in the gastric lumen (pH 2) is prevented from diffusing back to the epithelium by the high viscosity of the gastric mucus gel on the lumenal side.

Mucin, a high-molecular-weight glycoprotein, is secreted by surface mucous cells, mucous neck cells, and glandular mucous cells. Polymerization of mucin subunits by means of disulfide bonds is essential for the formation of the hydrated gel. [418] The precise structural organization and approximate molecular weight of mucin polymers are unknown. Two significantly different models of gastric mucin polymerization have been proposed and are described in other reviews. [419], [420] The peptide backbone of mucin contains many serine, threonine, and proline residues (>40% molecular weight) that serve as the amino acid anchors for the glycosyl residues that branch from the protein. Partial and complete cDNA clones for mucin have been isolated, and deduced peptide sequences confirm this amino acid preponderance. [421], [422], [423] and [424] The structural analysis indicates that the mucin backbone contains extended arrays of tandemly repetitive peptides rich in threonine or serines that are potential sites for *O*-glycosylation.

Multiple mucin gene products are expressed at various sites. Specific cDNA clones coding mucins found in submaxillary glands, tracheobronchial tree, and intestinal mucosa have been isolated and characterized. [425], [426], [427] and [428] Toribara and colleagues [424] isolated a cDNA clone coding human gastric mucin, which is characterized by tandem repeat sequences rich in threonine, serine, and proline. [424] This gene is expressed primarily in the stomach and the gallbladder and has been localized on chromosome 11 (11p15.4-11p15.5). The unique gastric mucin has been called MUC6. The mucin that is synthesized in the rough endoplasmic reticulum undergoes vesicular transport to the Golgi apparatus for glycosylation. The major portion of mucin is heavily glycosylated, and nonglycosylated or "naked" regions of the peptide are joined to other mucins by disulfide bridges. The initial peptide-carbohydrate linkage involves glycosidic bond formation between *N*-acetylgalactosamine and the hydroxyl groups of serine or threonine residues. This type of *O*-linked glycosylation involves the addition of individual carbohydrate moieties, rather than the transfer of preassembled oligosaccharides, to gradually lengthening chains. [429] Fucose, galactose, *N*-acetylglucosamine, and *N*-acetylgalactosamine make up more than 95% of the sugar moieties in each chain. [430] Each mucin peptide contains up to several hundred linear or branched-chain oligosaccharides. This high carbohydrate content (>50% by weight) results in a highly viscoelastic substance that expands when hydrated. These properties may be essential to the protective function served by gastric mucus.

The intracellular transport of mucin proceeds through the cis-, medial-, and trans-Golgi cisternae before transfer to the apical mucous granule. Cytoplasmic transport vesicles presumably bud off Golgi membranes and transfer mucus to its target organelles. Fusion of vesicle and granule membranes results in gradually enlarging mucous granules that ultimately pack the apical cytoplasm. Intact microtubules appear to be needed for transport of secretory granules from the Golgi apparatus to the cell surface. Basal secretion of mucus occurs continually throughout the life span of mucous cells. [431] In vitro pulse labeling of human mucous granules reveals that intracellular transit and release occurs in 20 to 24 hours. In contrast to basal secretion, stimulation of mucous cells results in fairly rapid fusion of granular and apical cell membranes or extrusion of mucous granule contents. Fusion of subjacent mucous granules enhances the secretory response. This process is called *compound exocytosis* and results in a dramatically cavitated apical cell surface during stimulated secretion.

Study of the regulation of mucous cell secretion has been impeded by the inherent difficulties in quantitatively retrieving and measuring secreted mucus. Hydration of secreted mucus forms a viscous gel that adheres to mucosal cells. Attempts to quantify mucus secretion by morphologic methods, [432] release of radiolabeled glycoproteins, [433] and radioimmunoassay [434] have met with only limited success. Despite these limitations, several factors that affect mucin secretion have been identified and can be summarized.

Cholinergic agonists stimulate secretion in a portion of gastric mucous cells, but adrenergic neurotransmitters do not appear to influence release of secretory granules. Gastrin and CCK may increase feline gastric lumenal carbohydrate content, but the source of this presumed mucus secretion has not been determined. [435] Similarly, secretin has also been shown to increase the carbohydrate content of secreted gastric mucus in humans, but the cellular origin of this effect also is unknown. [436] PGE and PGF have been reported to stimulate the release of soluble and insoluble mucin from gastric mucosa, with a resultant increase in mucous gel

thickness. [437], [438] This increase may be produced by prostaglandin-induced HCO_3^- secretion, creating an outward alkaline flow carrying previously secreted mucins from gastric crypts and glands. [439] After gastric mucosal injury has occurred, multiple inflammatory and immune cytokines may be present in the mucosa. It seems plausible that these factors directly or indirectly stimulate mucous cell secretion as part of the reparative process. [418] Candidate secretagogues include leukotrienes, immune complexes, and mast cell histamine.

Boland and colleagues [440] established a system for isolating and culturing canine gastric mucous cells and demonstrated that the cells synthesize and secrete mucin and phospholipids. [440] Mucin glycoprotein and surface-active phospholipids may form a biochemical complex that is analogous to pulmonary surfactant, and it is this complex that may be critical for providing the gastric epithelium its protective barrier. PGE_2 may enhance mucosal defense through the release of surface-active phospholipids by gastric mucous cells. Other studies have demonstrated that the same secretagogues that stimulate gastric acid secretion (e.g., histamine, gastrin, carbachol) can stimulate mucin synthesis and release of surface-active phospholipids. [441], [442] Gastrin, in particular has been shown to stimulate the biosynthesis of mucin from surface mucus cells of the rat through NO-dependent mechanism, because NO synthase inhibitors were able to block the stimulatory action of gastrin. [443] These observations suggest that gastric acid secretion (i.e., aggressive factor) and gastric mucosal defense are regulated in a coordinated or parallel fashion.

Mucous cells presumably have various receptors mediating stimulatory effects on their basolateral membrane, but no specific receptors have been demonstrated on these cells. Part of the difficulty in performing radioreceptor binding studies in this cell type is the high level of nonspecific ligand binding to the mucous gel. Functional studies have suggested that carbachol, histamine, and gastrin act directly on gastric mucous cells. Seidler and Pfeiffer [444] demonstrated that acetylcholine stimulates IP_3 formation, $[Ca^{2+}]_i$ mobilization, and mucin release from isolated gastric mucous cells. However, neither dibutyryl AMP nor phosphodiesterase inhibitors have been shown to stimulate mucin release from intestinal mucous cells. [445] Conversely, a Ca^{2+} ionophore has been shown to induce visible loss of mucous granules from guinea pig gastric mucosa and to increase the thickness of the mucous gel. [446] Together, these results suggest that gastric mucous cells may be stimulated by secretagogues that use the Ca^{2+}-mediated intracellular pathways.

Research has indicated that activation of protease-activated receptor 2 (PAR-2) leads to the secretion of gastric mucus through the release of calcitonin gene–related peptide and tachykinins from sensory neurons, suggesting a novel mechanism for the regulation of mucus secretion in the stomach. [447] Much remains to be learned about the regulation of gastric mucous cell secretion in the basal and stimulated states.

Bicarbonate

Gastric mucosal defense depends on a mucus-HCO_3^- barrier coating the entire lumen of the stomach. This barrier exists as a gel with a pH gradient that provides a neutral microenvironment at the epithelial surface. The gradient is generated by the secretion of HCO_3^- by gastric surface mucous cells. HCO_3^- uptake at the basolateral membrane and secretion at the apical membrane are metabolically dependent processes. [448] CA, the enzyme responsible for HCO_3^- generation, has also been localized to the apical matrix and microvillous cores of surface epithelial cells. [449] HCO_3^- secretion appears to be mediated by a Cl^- and HCO_3^- exchange mechanism on the lumenal surface of gastric epithelial cells. [450] In addition to active secretion, passive efflux of HCO_3^- presumably occurs through a paracellular route. Mucosal delivery of HCO_3^- ions is enhanced by fenestrations of the capillaries supplying the epithelial cells. [53] The relative proportion of passive and active HCO_3^- secretion in basal and stimulated states has been difficult to determine and depends on the type of model studied.

Several models have been used to measure gastric HCO_3^- secretion. Mucosal membranes can be mounted in chambers and bathed with appropriate solutions. [451], [452] and [453] Titration of lumenal secretions is used to calculate HCO_3^- release. In vivo measurements have been performed in fundic and antral pouches and are described elsewhere. [454], [455] In humans, gastric HCO_3^- secretion can be measured in vivo by back-titration after complete pharmacological inhibition of acid secretion. [456] Another technique involves rapid perfusion of the stomach (30 mL/min), with continuous measurement of PCO_2 and pH in the gastric aspirate. [457] HCO_3^- and H^+ secretion can then be calculated by the Henderson-Hasselbalch equation. This technique relies on the assumption that all lumenal CO_2 results from neutralization of H^+ ions by HCO_3^-. A third approach uses a two-component model that calculates gastric HCO_3^- secretion from measurements of gastric juice volume, H^+ concentration, and osmolality. [458] This model assumes a fixed relation between the osmolality of plasma and that of gastric secretions such that a decrease in osmolality occurs only as a result of H^+ and HCO_3^- neutralization. This method results in a higher calculation of gastric HCO_3^- secretory rate (2300 µEq/h) than obtained with other methods (400 µEq/h).

Because of the quantitative differences observed with the various methods used, Odes and colleagues [459] performed detailed in vivo and in vitro studies using the different techniques outlined. They observed that in the acid-suppressed stomach, gastric HCO_3^- is accurately determined by back-titration. However, measurement of HCO_3^- in the acid-secreting stomach is not accurate with any of the methods tested. Despite these quantitative differences, responses to various stimuli are qualitatively similar with all the techniques.

Regulation of gastric HCO_3^- secretion is an important component of gastric mucosal defense. Sham feeding and electrical stimulation of the vagus induce gastric HCO_3^- secretion. [460], [461] and [462] This neural control mechanism is effectively blocked by atropine and benzilonium bromide. Intravenous infusion of cholinergic agents such as bethanechol also stimulate HCO_3^- and H^+ secretion. [456], [458] The coordinated stimulation of acid and HCO_3^- secretion by vagal transmitters is not reproduced by gastrin or histamine. Local regulation of HCO_3^- secretion is initiated by the presence of lumenal acid. Studies using canine denervated pouches [463] and isolated frog mucosal strips [464] suggest that a humoral factor that stimulates HCO_3^- secretion is released in response to lumenal acid. Although exogenous prostaglandins have been shown to stimulate gastric and duodenal HCO_3^- secretion, the physiological role of endogenous prostaglandins in HCO_3^- control is less clear. [448], [465] The inhibition of gastric HCO_3^- secretion is not well characterized. Vagal stimuli and lumenal acid appear to stimulate surface mucous cells to secrete HCO_3^-. Acid stimulation also results in delivery of HCO_3^- from the parietal cells to the surface epithelium by ascending mucosal capillaries (i.e., the alkaline tide). These regulatory controls ensure simultaneous secretion of gastric acid and HCO_3^-.

Intrinsic Factor

IF, a 45-kd glycoprotein present in gastric secretions, is essential for the absorption of cobalamin (vitamin B_{12}) in the terminal ileum by receptor-mediated endocytosis. Its existence was first postulated by Castle in 1929. [466] Human IF is synthesized and secreted by parietal cells. [467] Parietal cells also are the source of IF in cats, rabbits, monkeys, guinea pigs, and oxen. [444] In rodents such as the rat and mouse, chief cells produce IF, and its source in pig is gastric mucous cells. [469], [470] The structure of rat IF, as deduced from a cDNA clone, indicates a primary amino acid sequence of 421 amino acids, with a putative signal sequence of 22 amino acids. [470] The cobalamin-binding domain is thought to reside in the NH_2-terminal half of the protein. There is 80% identity between the predicted amino acid sequences corresponding to rat and human IFs. [471] The single human IF factor gene, located on chromosome 11, and the human IF appears to have a high degree of conservation with monkey, rat, mouse, and cow IFs.

IF is secreted in amounts far exceeding that necessary for cobalamin absorption. [472] Secretion of IF is stimulated by the same pharmacological agents as acid secretion—pentagastrin, histamine, and cholinergic agonists—but this secretory response is not linked to acid secretion. [473] For example, omeprazole does not alter basal or stimulated IF secretion in humans, nor does it alter the absorption of labeled cobalamin. [474], [475] The intracellular responses mediating IF secretion involve the cAMP pathway when stimulated by histamine. [473] Whether pentagastrin and acetylcholine, which appear to act through Ca^{2+}-phospholipid–dependent pathways when stimulating acid secretion, act in a similar fashion when stimulating IF secretion is unknown.

Inhibitory regulation of IF secretion is poorly understood. Somatostatin inhibits histamine- or pentagastrin-stimulated IF secretion in isolated guinea pig gastric glands. [476] EGF is reported to inhibit histamine-stimulated IF secretion in rabbit isolated gastric glands and in humans. [477], [478] The physiological significance of these

observations is unknown.

IF resists digestion by gastric acid and proteolytic enzymes under normal circumstances. A rare kindred with cobalamin malabsorption caused by an abnormal IF that was susceptible to acid and proteolysis has been reported. [479] An absence of IF secretion occurs rarely in persons, usually children, with normal acid secretion. [480], [481] The discussion of the significance of IF in health and disease is continued in Chapter 20.

Human tissue studies have demonstrated that IF is expressed not only in parietal cells, but also in gastric chief cells and enteroendocrine cells. [482] The expression of IF in these latter sites appears to be lower than the level of expression in parietal cells. Some investigators have suggested that the principal reason for the differential expression of IF in different cell types lies in the variation between untranslated region of the human and rat's gene, [483] however, the observation that IF can also be expressed in human chief cells goes somewhat against this theory. [482] Questions still remain regarding the interspecific variation in IF expression and the cellular pathways that regulate IF synthesis and secretion.

Prostaglandins

The role of prostaglandins in gastric acid secretion and mucosal defense has been an area of active research. Prostaglandins are 20-carbon fatty acid derivatives that have diverse biologic activities and are produced in many different tissues. They are synthesized from arachidonic acid, a product of enzymatic cleavage of cell membrane phospholipids by phospholipases, particularly phospholipase A_2. [484] Cyclooxygenase rapidly metabolizes free arachidonic acid to cyclic endoperoxides that are transformed to various prostaglandin subtypes by tissue-specific processing enzymes (Fig. 13-15). PGE_2 and PGI_2 (or prostacyclin) are synthesized and secreted by the gastric mucosa of various species. The presence of these two predominant forms in homogenates of gastric mucosa has been confirmed by bioassay, radioimmunoassay, and gas chromatography with mass spectrometry. Biosynthetic studies using radiolabeled arachidonic acid have confirmed that gastric mucosa can synthesize both these prostaglandins. The specific mucosal cell types synthesizing the various prostaglandins have not been identified. Although canine and rat parietal cell fractions appear to form several prostaglandin subtypes, other cells, such as mucous and chief cells, may also synthesize prostaglandins.

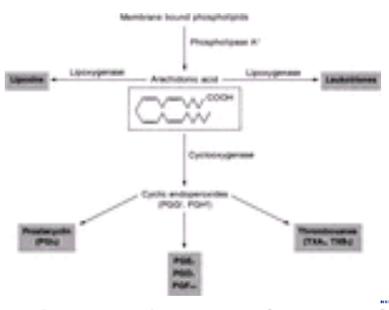

FIGURE 13-15. Pathways of arachidonic acid metabolism.

Gastric prostaglandins are likely to have paracrine effects on other gastric mucosal cells. One example is direct inhibition of acid secretion by prostaglandins. Studies of canine parietal cells show that PGE_2 decreases [^{14}C]aminopyrine accumulation and cAMP production. [217] This effect appears to be mediated through prostaglandin receptors activating inhibitory guanine nucleotide–binding proteins. [485] Inhibition of parietal cells could be one effect of a prostaglandin-mediated negative-feedback loop regulating gastric acid secretion.

Prostaglandins may also function as lumenal hormones. PGE_2 has been demonstrated in the gastric juice of several species, including humans, cats, dogs, and rats. [484] Secretion of PGE_2 into lumenal fluid is maximal during pentagastrin-stimulated acid secretion. The presence of prostaglandins in gastric juice and gastric mucosa may facilitate the diverse cytoprotective functions ascribed to these agents, in addition to their antisecretory effects. These functions include the following: stimulation of mucus, phospholipid, and HCO_3^- secretion; enhancement of mucosal blood flow; reduction of mucosal H^+ ion back-diffusion; and stimulation of mucosal cell turnover. Prostaglandins may function in this way to mediate mucosal defense.

Prostaglandin deficiency may predispose to gastric mucosal injury. Cyclooxygenase inhibitors such as aspirin and non-steroidal antiinflammatory agents produce a spectrum of mucosal injuries in the stomach. Immunization against prostaglandins has caused ulcers in rabbits. [486], [487] Part of the cause of these ulcerations is a decrease in prostaglandin-mediated mucosal defense. These observations have served as the basis for the development of prostaglandins for use in the treatment of acid or peptic diseases of the stomach (see Chapter 66).

CLINICAL IMPLICATIONS OF GASTRIC SECRETION

Gastric acid secretion is a well-studied process with a history involving many of the legendary names in gastrointestinal research, such as Beaumont, Pavlov, and Grossman. There are few bodily functions that have been examined so completely in vivo and in vitro as the stomach in processing food and initiating the digestive process. It should be remembered, however, that human patients with no stomachs can live normal lives if they are provided with vitamin B_{12}.

Disorders Associated with Increased Acid Secretion

Despite the landmark discovery of *Helicobacter pylori* and its central role in the pathogenesis of peptic ulcer disease, gastric acid continues to be an important factor in the equation leading to disruption of gastric and duodenal mucosal integrity. The mechanisms by which this bacterium, which resides in the gastric mucosa, produces duodenal ulcerations remain unknown. Abnormalities in the gastric secretory process have been described in patients with *H pylori* and duodenal ulcers. This subject is discussed in greater detail in Chapter 66, but a brief overview is included here. Patients with duodenal ulcers who are infected with *H pylori* have a threefold elevated BAO and an increased maximal acid response to gastrin as compared with *H pylori*–negative volunteers. [488] A more dramatic difference is noted between these groups of patients when the gastric acid response to gastrin releasing peptide is examined. A sixfold increase in BAO occurs in those infected with *H pylori*. These abnormalities are reversible with eradication of the organism. Additional abnormalities observed in *H pylori*–infected individuals include increased basal and meal-stimulated gastrin levels [489], [490] and decreased gastric somatostatin levels. [491] More recent work has demonstrated that infection with *H pylori* may result in alteration of the pathways important in inhibiting gastric secretion. Specifically, *H pylori* infection in patients with duodenal ulcers decreases the inhibitory action of CCK. This defect in CCK-mediated inhibition is corrected after eradication of *H pylori.* [492], [493] Olbe and colleagues [494] observed that infection with this bacterium blocked antral distention–mediated inhibition of gastrin release and acid secretion in a manner that was reversible with *H pylori* eradication. Of note, the abnormality occurred independent of duodenal ulcer status. From these data, it is theorized that dysregulation of acid secretion in patients infected with *H pylori* leads to the delivery of greater amounts of acid to the duodenum that, in turn, promotes ulcer development. The observation that a block in antral distention–mediated inhibition of acid secretion occurs independent of ulcer formation suggests that other factors must be involved in the pathogenesis of this common disorder. Although these are interesting theories, the role of gastric secretory abnormalities in *H pylori*–mediated peptic ulcer disease is not established.

Zollinger-Ellison syndrome is a disorder in which there is autonomous hypersecretion of gastrin by an islet cell tumor, usually situated in the pancreas. There is gross hypersecretion of acid in the basal state and a poor response to exogenous secretagogues. The BAO usually is greater than 15 mmol per hour, and the ratio of BAO to MAO usually is greater than or equal to 0.6. These tests are useful for differentiating hypergastrinemia caused by Zollinger-Ellison syndrome from the more common situation in which serum gastrin levels are elevated as a consequence of achlorhydria.

Retained gastric antrum syndrome is a rare consequence of antrectomy and Billroth II gastrojejunostomy, in which the gastric secretions are directed away from a distal segment of unresected antrum. Because the antral segment is not in contact with the acidic gastric contents, the normal negative-feedback loop in which gastrin release is inhibited at low intralumenal pH concentrations is not activated. This results in unrestrained secretion of gastrin and continuous stimulation of acid secretion. Some patients with retained antrum syndrome have recurrent peptic ulceration.

Excess production of histamine is an unusual cause of gastric acid hypersecretion. It is found with systemic mastocytosis, foregut carcinoid tumors, and basophilic leukemia. Even rarer causes of significant hypersecretion are extensive resections of the small bowel and raised intracranial pressure. [495], [496]

Disorders of Gastric Acid Hyposecretion

The most common hyposecretory disorder is chronic atrophic gastritis. This condition is accompanied by hypochlorhydria or achlorhydria. Because of the absence of gastric acid, the negative-feedback control of gastrin release is interrupted, and these patients have elevated serum gastrin levels, frequently as high as 1000 pg/mL or more. [497] These patients may be misdiagnosed, on the basis of hypergastrinemia, as having Zollinger-Ellison syndrome unless acid secretory studies are performed.

Chronic atrophic gastritis often is accompanied by an absence of IF secretion. This deficiency leads to pernicious anemia, a disease characterized by the failure of cobalamin absorption in the terminal ileum and megaloblastic anemia. These patients usually display circulating antibodies to IF and parietal cells, which may have an etiopathogenetic role in inducing or maintaining IF or acid hyposecretion. [498], [499] There are rare cases, usually among children, of IF hyposecretion in the presence of normal acid secretion. [480], [481]

Reduced acid and pepsin secretion often occurs in patients with gastric ulcers, gastric polyps, and gastric carcinoma. Hyposecretion of gastric acid is common after partial gastrectomy or vagotomy procedures.

Several reports have documented decreased gastric acid and IF secretion in patients with advanced acquired immunodeficiency syndrome (AIDS). [500], [501], [502] and [503] In contrast, similar abnormalities have not been observed in the early stages of human immunodeficiency virus (HIV) infection. [503], [504] Although the pathogenesis of these secretory abnormalities is unknown, preliminary research has demonstrated several secretory and parietal cell morphologic abnormalities in HIV-infected patients. [505] Despite the limited number of patients examined, several interesting observations were made. Fasting gastric pH and IF concentrations were significantly different in patients with AIDS and in the early stages of HIV infection as compared with a group of healthy volunteers. As previously noted, MAO output and Schilling test (1 and 2) results were decreased only in AIDS. At the histological level, vacuolar degeneration (light microscopy), reduced tubulovesicles, and dilated intracellular canaliculi with loss of microvilli (electron microscopy) were observed in gastric biopsy specimens obtained from HIV-infected patients. Moreover, there was evidence of HIV antigens (gp120, gp41, p24, and p17) in parietal cells obtained from patients in both the early and late stages of HIV infection. Although a preliminary report, the histological findings are quite dramatic and suggest that HIV may be directly responsible for infecting and altering parietal cell structure and function.

Pharmacology of Acid Secretion

The advances in the pharmacology of acid secretion suppression mirror the advances in the physiological understanding discussed in the preceding sections of this chapter. Pharmaceutical products that interact at almost every stage of acid secretory regulation are available. H_2 receptor antagonists are the prototypical acid secretory antagonists. These agents reduce but do not abolish stimulated acid secretion. Because of the influence of potentiation, H_2 antagonists are able to reduce acid secretion in response to histamine or to pentagastrin and acetylcholine. They are effective in promoting ulcer healing.

PGE and PGI selectively inhibit histamine-stimulated parietal cell function in vitro. One mechanism for this effect of PGE analogs is their interaction with the pertussis toxin–sensitive guanine nucleotide–binding regulatory protein of adenylate cyclase in the parietal cell basolateral membrane. PGEs are effective inhibitors of acid secretion in vivo. [505] Part of this action may reflect prostaglandin-mediated release of somatostatin. [506] Enprostil, a PGE agonist, inhibits serum gastrin levels. [505]

Direct interference with the parietal cell proton pump is possible using substituted benzimidazoles (e.g., omeprazole), which inhibit acid secretion by blocking parietal cell H^+,K^+-ATPase. [507] These agents inhibit the acid secretory effects of all known stimuli. [507] They are the most powerful antisecretory agents available for use in humans and can produce absolute achlorhydria. [508] Compared with cimetidine, benzimidazoles are more effective in relieving symptoms and in healing ulcers in patients with duodenal ulcers. [509], [510] Just as achlorhydria resulting from chronic atrophic gastritis and pernicious anemia causes marked hypergastrinemia, omeprazole-induced achlorhydria also is accompanied by sustained hypergastrinemia. The knowledge that gastrin is a growth-promoting hormone has raised concerns that long-term administration of omeprazole may promote gastric tumor formation, as can occur in patients with pernicious anemia or after gastric surgery for chronic duodenal ulcer. [511], [512] and [513]

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CHAPTER 14

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ELECTROLYTE SECRETION AND ABSORPTION: SMALL INTESTINE AND COLON

The goal of this chapter is to review the cellular, molecular, and regulatory aspects of fluid and electrolyte transport in the mammalian intestinal tract. The intestine can absorb vast quantities of fluid from the intestinal lumen. This is a vital function, because of the presentation of about 9 L of fluid are presented to the intestine each day (Fig. 14-1). Greater than 98% of this fluid load is absorbed to preserve health. In addition to its ability to absorb fluid, the intestine also is capable of secretion. In fact, most of the daily fluid load originates from secreted rather than ingested fluids. Fluid secretion is necessary to aid in the digestion and absorption of nutrients, by maintaining a fluid environment for the mixing of food with digestive enzymes. The balance between absorptive and secretory processes is closely regulated. There is normally a net absorption of fluid, but certain conditions may result in excessive stimulation of secretion, which can overwhelm the combined absorptive capacity of the small and large intestines and can result in the pathological condition of secretory diarrhea. The most dramatic example is cholera; patients can experience drastic fluid loss in the stool of up to 20 L per day. [1]

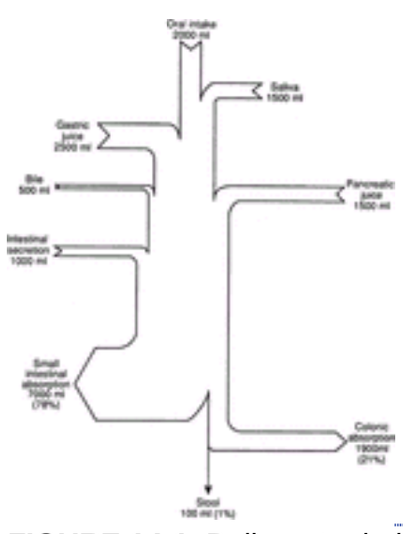

FIGURE 14-1. Daily water balance in the healthy human gastrointestinal tract. The amount of oral intake varies among individuals depending on the types of meals taken. Note that even in health there is a significant secretory flux of fluid from the intestine (1000 mL). The small intestine is responsible for absorbing almost 80% of the daily fluid load, largely in association with nutrient uptake. The colon absorbs most of the remaining fluid, with an efficiency of about 95%, leaving only about 1% of the daily fluid load to be lost to the stool. (From Barrett KE, Dharmsathaphorn K. Transport of water and electrolytes in the gastrointestinal tract: physiological mechanisms, regulation and methods for study. In: Narins RG, ed. Maxwell and Kleeman's clinical disorders of fluid and electrolyte metabolism, 5th ed. New York: McGraw-Hill, 1994:493.)

Intestinal fluid movement is predominantly controlled by the active transport of Na^+, K^+, bicarbonate (HCO_3^-), and Cl^- ions across the intestinal epithelium. Fluid follows the direction of electrolyte movement to maintain isotonicity between the intestinal lumen and tissue compartments. In addition to this vital role in fluid homeostasis, electrolyte transport has important roles in other physiological processes. For example, mucosal protection in the duodenum is dependent on duodenal secretion of HCO_3^-. [2], [3] Plasma electrolyte homeostasis is affected by electrolyte transport in both the colon and the kidney. [4] The uptake of peptides, amino acids, sugars, and bile acids requires various specialized ion-dependent transport mechanisms in the small intestine. [5], [6] and [7]

Many dramatic advances have been made in understanding intestinal electrolyte transport from the molecular to the whole-tissue level. Molecular cloning of transport, regulatory, and structural proteins has provided new insight into the regulation of electrolyte transport processes at the molecular level and has defined the underlying basis for genetic disorders of intestinal transport. Advances in cellular biochemistry have shown how different second messenger pathways interact with one another, with transport proteins, and with the cytoskeleton to allow the intestinal epithelium to respond appropriately to changes in the extracellular environment. Enhanced knowledge of epithelial development and tight junctional structure has increased the understanding of cellular diversity and of how the epithelium acts as a barrier to lumenal toxins. At the whole-tissue level, our knowledge of intercellular communication among the mucosal immune system, the enteric nervous system, and the intestinal epithelium is growing constantly. This chapter therefore emphasizes molecular mechanisms of intestinal epithelial ion transport processes as well as the intracellular and extracellular factors involved in their regulation.

INTESTINAL EPITHELIUM

Structural Properties of Epithelial Cells

The intestinal epithelium is a continuous monolayer of cells (i.e., enterocytes) lining the entire lumen of the gut. It simultaneously forms a barrier to the nonselective movement of substances between the body and gut lumen and expresses proteins that permit the selective transport of desired substances. The capacity of the intestine to absorb and to secrete is enhanced by the massive surface area of the epithelium, which is greatly amplified (by about 600-fold in the small intestine) by folds, villi, and microvilli. In addition, regulated contractions of the intestinal smooth muscle layer can increase or decrease the flow of lumenal contents, allowing

optimal contact time of nutrients and fluids with the epithelium.

Two essential features of the intestinal epithelium (and all other epithelia) that enable it to conduct its barrier and electrolyte transport functions are the ability of the component cells to form homotypic intercellular tight junctions and the ability to develop functional polarity. The basis for these features is discussed below.

Tight Junctions The intestinal epithelium forms a continuous barrier with each cell joined to its neighbors by apical junction complexes. These structures, which are composed of tight junctions and adjacent adherens junctions, link neighboring cells close to the apical pole. The junctional complexes form a regulated barrier to the movement of substances, not only between the cells but also within the plane of the epithelial cell plasma membrane. In response to the movement of ions and other solutes through epithelial cells, counterions and fluid can be driven paracellularly by chemical and osmotic gradients through the tight junctions, which behave as selective pores. At the ultrastructural level, freeze-fracture techniques reveal that epithelial tight junctions consist of a dense network of protein strands. [8] In general, the number of strands constituting the tight junction is directly related to the permeability of the pore. [9] Tight junction permeability is not constant along the length of the intestinal tract and generally decreases aborally. Recent years have seen an explosion in our knowledge of the molecular architecture of tight junctions (Fig. 14-2). [10] Many of the transmembrane components of tight junctions have been identified. [11] The first of these was occludin, a 65-kd transmembrane protein inserted into the plasma membrane in such a way as to expose two hydrophobic extracellular loops rich in glycine and tyrosine residues. [12], [13] These loops are believed to interact with partners on adjacent cells to form homotypic intercellular bonds (see Fig. 14-2). However, although occludin undoubtedly is involved in the formation of tight junctions, studies of occludin knock-out mice, in which intestinal tight junction structure and function appear to be unaltered, [14] indicate that other proteins are likely to be involved. This led to the discovery of the claudins, a family of proteins that, to date, consists of 20 members ranging in sequence identity from about 10% to 70%. With a molecular weight of about 22 kd, claudins are considerably smaller than occludin, but they are structurally similar in that they possess intracellular amino and carboxy termini and two extracellular loops. Claudins exhibit stronger adhesion characteristics than occludin, and several lines of evidence suggest they may be the primary sealing and pore-forming components of the tight junction. [10], [15] Claudins also display a varied tissue distribution, which may explain the high degree of heterogeneity in tight junction permeability between different tissues. [16] Evidence suggests that claudins also may be involved in regulating the ionic selectivity of tight junctions. Paracellin, a member of the claudin family, has been identified in renal epithelial cells, where it appears to confer cation, particularly Mg^{2+} and Ca^{2+}, selectivity on the tight junctions of the thick ascending limb of Henle. [17] Another transmembrane component of tight junctions is junctional adhesion molecule (JAM). [18], [19] and [20] A member of the immunoglobulin superfamily, JAM is structurally unrelated to claudins and occludin. In addition to regulating the sealing and permeability of tight junctions through the formation of intercellular homotypic bonds, JAM may play a role in regulating leukocyte migration through the tight junction. [18]

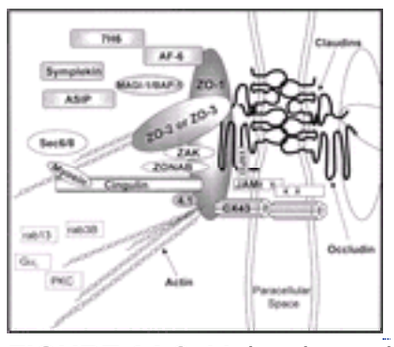

FIGURE 14-2. Molecular architecture of epithelial tight junctions. The transmembrane proteins occludin, the claudins, and junctional adhesion molecule (*JAM*) are the pore-forming constituents of the tight junction. Each of these proteins forms extracellular homotypic contacts with partners on neighboring cells. It is thought that the claudins, of which there are multiple isoforms, confer ionic selectivity on the tight junction. On the intracellular face of the tight junction, claudins and occludin associate directly with the ZO proteins, ZO-1, ZO-2, and ZO-3. Through several motifs that allow protein-protein interactions, the ZO proteins are, in turn, associated with the cellular actin cytoskeleton and a variety of intracellular signaling proteins that are involved in modification of tight junction permeability. *ASIP,* atypical PKC–isotype-specific interacting protein; *BAP-1,* BAI-associated protein; *PKC,* protein kinase C; *VAP,* vesicle-associated membrane protein (VAMP)-associated protein; *ZONAB,* ZO-1–associated nucleic acid–binding protein. (From ref. [10]. Copyright 2000 by the American Physiological Society.)

On the cytoplasmic face of tight junctions, occludin, claudins, and JAM are associated with a myriad of intracellular regulatory proteins. The most thoroughly studied of these are the zonula occludens (ZO) proteins: ZO-1, ZO-2, and ZO-3. [21], [22] and [23] These are members of a class of proteins called the *membrane-associated guanylate kinase homologs* (MAGUKs), a family of proteins characterized by a complex multidomain structure. [23] In addition to a guanylate kinase domain, these proteins contain several motifs involved in protein-protein interactions, including SH3 domains, [24] PDZ domains that bind integral membrane proteins, [25], [26] and docking sites (YEXV motif) for the SH2 domains of nonreceptor tyrosine kinases. [27] A novel ZO-1 interacting protein has been identified in kidney epithelial cells, which adds an exciting new dimension to the way in which ZO-1 may regulate cell function. [28] This protein, termed *ZONAB* (for ZO-1–associated nucleic acid–binding protein), is a transcription factor that preferentially activates the ErbB2 promoter. ErbB2 is a receptor tyrosine kinase involved in cell growth and differentiation. Thus, ZO-1 may play a fundamental role in regulating epithelial dynamics. Many other proteins also have been localized to tight junctions, including cingulin, the 7H6 antigen, low-molecular-weight G proteins, heterotrimeric G proteins, and tyrosine kinases. [29], [30] and [31] Although the functions of several of these proteins have yet to be elucidated, the aggregation of diverse signaling proteins at tight junctions clearly suggests that these structures are closely regulated. Apical junction complexes appear to be linked to the cytoskeleton by actin microfilaments, which form a dense perijunctional ring underlying the tight junctions. Actin microfilaments also appear to make direct contact with tight junction proteins including the ZO proteins and occludin. [32] Agents causing elevations in intracellular second messengers, such as Ca^{2+} and cyclic adenosine 3',5' monophosphate (cAMP) alter junctional permeability, probably through mechanisms involving contraction of the cytoskeleton and the perijunctional ring. [33] Other signaling molecules believed to be involved in the regulation of tight junction permeability include protein kinase C, myosin light-chain kinase, mitogen-activated protein kinases, and phosphatidylinositol 3-kinase (PI3-K). [34], [35], [36] and [37] Extrinsic factors, such as migrating neutrophils, [38] mucosal pH, [39] and dietary and bacterial toxins [40] also regulate tight junction permeability. Epithelial tight junction permeability also may be regulated by the absorption of nutrient-derived solutes, such as glucose. [35] Increases in lumenal glucose concentrations activate SGLT1, a cotransporter that couples glucose uptake to a favorable electrochemical gradient for Na^{+} absorption. Increased SGLT1 activity is associated with increases in tight junction permeability through an indirect mechanism involving the Na^{+}/H^{+} exchanger, NHE3. [41] This, in turn, reduces the barrier properties of the epithelium, leading to solvent drag of fluid and subsequent increased uptake of such solutes through the paracellular route. The validity of this solvent drag hypothesis has been questioned because increased paracellular flux in the presence of lumenal glucose has not been observed in all experimental models. For example, at SGLT1 substrate concentrations likely to exist in the intestinal lumen, the paracellular pathway contributes little (i.e., less than 5%) to the observed uptake of sugar from the intestinal lumen. [42] However, more recent in vivo studies in humans have shown that Na^{+}/glucose transport may dramatically increase paracellular absorption up to 45%. [43] Thus, despite the uncertainty of the solvent drag hypothesis and of its contribution to fluid absorption under normal conditions, both in vivo and in vitro studies provide convincing evidence that coupling between transcellular and paracellular pathways for solute transport can exist within the same cell. In summary, the opening of tight junctions can have significant consequences for intestinal physiology because as tight junctions become more permeable, the barrier to toxigenic substances, nutrients, electrolytes, and water is diminished.

Functional Polarity For fluid movement to occur, intestinal epithelia must transport solutes in a vectorial fashion Vectorial transport requires functional polarity of the epithelial cells, in which the proteins involved in electrolyte transport are asymmetrically distributed between the apical (lumen-facing) and basolateral (serosa-facing) domains of the cellular plasma membrane. Epithelial polarity is important not only for the differential localization of ion transport proteins but also for the localization of receptors for various neurotransmitters and other regulatory mediators. Many intestinal epithelial receptors, such as those for acetylcholine, histamine, and neuropeptides, are expressed only on the basolateral side, which is not surprising considering that the basolateral domain of the epithelium is in intimate contact with enteric nerves and cells of the mucosal immune system. Conversely, receptors for endogenous or exogenous substances delivered through a lumenal route (e.g., guanylin or bacterial enterotoxins, respectively) are found on the apical pole of epithelial cells. Some receptors are expressed bilaterally, such as those for adenosine and kinins. [44], [45] The cellular mechanisms responsible for differential localization of membrane proteins to the apical and basolateral domains of epithelial cells are only beginning to be understood. Extracellular contact, either between adjacent cells or between cells and the extracellular matrix, appears to be an important cue for the development of this epithelial polarity. [46], [47] Important components for sorting proteins to the apical or basolateral compartments are the trans-Golgi network, the cytoskeleton, and various biochemical signals that regulate membrane trafficking events. In the trans-Golgi network, proteins destined for the apical and basolateral membranes are sorted into distinct vesicles. The role of the cytoskeleton is demonstrated by the observation that depolymerization of the cytoskeleton results in the delivery of normally apical proteins to the basolateral membrane. [48], [49] Specific biochemical signals on the targeted proteins preferentially direct membrane proteins to either the apical or the basolateral domain. Basolateral sorting signals usually are short cytoplasmic peptides that contain a conserved tyrosine-based motif. In contrast, apical sorting signals typically are found in the extracellular domain and often involve *N*-glycosylation of the protein. It has been proposed that proteins

bound for the apical membrane are shipped from the Golgi network in glycosphingolipid rafts, and clustering of proteins into these rafts occurs by means of their glycophosphoinositol anchors. [46] It is also thought that specific peptide sequences, or single amino acid residues, within a particular protein may play an important role in directing membrane trafficking. [50] Within the membrane, mechanisms also exist that help to direct and maintain polarity. For example, it is believed that the plasma membrane contains targeting patches in which specific docking molecules, such as target membrane–related soluble *N*-ethylmaleimide–sensitive factor attachment protein receptors (t-SNAREs), are found. Such docking molecules have the ability to recognize specific proteins on the surface of transport vesicles, such as vesicle-related SNAREs (v-SNAREs). [46] The cytoskeleton also helps to anchor some proteins at specific sites within the membrane, and tight junctions act as a "fence" to prevent free movement of proteins between the apical and basolateral domains within the plane of the membrane.

Epithelial Organization and Diversity

In addition to the common epithelial features described, there is a wide diversity in cell type and function within the intestinal epithelial layer. This diversity is apparent along the length of the intestine (the jejunocolonic axis) and among the cells that constitute an individual crypt and villus unit (the crypt-villus axis).

Crypt-Villus Axis The predominant cells within the intestinal epithelium are the enterocytes, the transporting epithelial cells that perform the absorptive and secretory functions of the intestine. In general, the proteins specifically involved in digestion and absorption are expressed at highest levels in the enterocytes near the villus tip, whereas the proteins involved in secretion are most abundant in crypt enterocytes. Thus, there is a functional heterogeneity in electrolyte, fluid, and nutrient transport along the crypt-villus axis. Indeed, for many years it was believed that absorption and secretion were spatially distinct processes, with absorption occurring only across villus cells and secretion restricted to the crypts. [51] However, it has become apparent that this scheme was an oversimplification. Studies of the rat small intestine demonstrate that secretory mechanisms may be present in some villus cells. [52] Further, although colonic crypt cells secrete Cl^- in response to neurohumoral agonists, they constitutively absorb Na^+ and water in the absence of secretory stimuli. [53] These physiological observations are complemented by new information about the cellular expression of individual transport proteins, discussed in the section "Electrolyte Transport Proteins." Other epithelial cells also contribute to and regulate the functions of the transporting cells. Goblet cells located in the crypts secrete mucus. [54] Paneth cells also located in the crypts contribute to mucosal defense by producing antibacterial substances (e.g., lysozyme, defensins). [55], [56] They also may have a role in regulating epithelial development or repair by producing growth factors. [57] Enteroendocrine cells, which are interspersed between the enterocytes, contain a variety of hormones, neuropeptides, and serotonin. [58] These mediators can be released in response to various lumenal stimuli, including bacterial toxins and distention, and may act in a paracrine fashion to alter epithelial ion and fluid transport. [59] All four epithelial cell types are replenished constantly from stem cells found near the base of each crypt. As cells divide, most are pushed upward along the crypt toward the villus tip, from which they are shed after a period of 4 to 7 days. [60], [61] The 4- to 7-day cycle is completed when the migrating cells undergo apoptosis at the villus tip (or surface epithelium in the colon) and are shed into the lumen or reabsorbed.

Jejunocolonic Axis The intestinal tract also is heterogeneous along its longitudinal axis, reflecting the different functions of each intestinal segment. The proximal segments of the small intestine absorb large volumes of fluid against relatively small electrochemical gradients. Resistance across the epithelium of the small intestine is low. The duodenum, jejunum, and ileum exhibit regional differences in terms of electrolyte, nutrient, vitamin, mineral, and bile acid transport. The colon has two distinct areas characterized by functional differences in electrolyte transport. The epithelium of the proximal colon exhibits intermediate resistance between leaky and tight, whereas the distal colon has a semitight epithelium that absorbs Na^+ against a steep gradient. Perfusion studies have shown that electrolyte concentrations in the intestinal lumen mirror those of plasma in all intestinal segments except the distal colon, where the Na^+ concentration is reduced and that of K^+ is increased. [62] Differential sensitivities to various secretagogues, aldosterone, amiloride, and amiloride analogs have been found between the proximal and distal colon. [63], [64] A clinical correlate to these physiological observations is the finding that surgical removal of the right colon is associated with diarrhea and salt wasting, supporting the theory of a specialized function for this segment of the intestine. [65]

Regulatory Cells

Epithelial ion transport is regulated by a wide array of extracellular stimuli, which include lumenal factors and blood-borne, neuronal, and immunologic mediators (Fig. 14-3). Histological examination of the lamina propria reveals various immune cell types (the mucosal immune system), an extensive neuronal network (the enteric nervous system), and enteroendocrine cells that are interspersed among the cells of the epithelial layer. The epithelium also has an underlying layer of specialized mesenchymal cells called *myofibroblasts.* Mediators released from all these cell types contribute to the regulation of epithelial transport function; both the mucosal immune system and the enteric nervous system integrate regulation of epithelial transport.

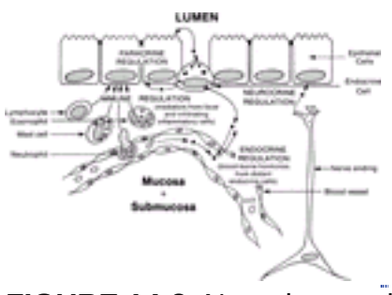

FIGURE 14-3. Neurohumoral and immune regulation of the intestinal epithelium. Endocrine cells in the crypt region release peptides and other substances across either their basolateral or apical membranes. In many cases, local diffusion of these bioactive substances can regulate nearby ion transporting cells (paracrine regulation). These substances, or others from more distant endocrine organs, also can enter the bloodstream to regulate the function of distant portions of the epithelium (endocrine regulation). Enteric nerve endings release peptides and other neurotransmitters that regulate both the epithelium and muscle layers (neurocrine regulation). Immunologic effector cells (e.g., mast cells, eosinophils, neutrophils, and lymphocytes) in the lamina propria can also be simulated to release substances that regulate the epithelium (immune regulation).

Neuroendocrine Cells Enteroendocrine cells and enteric nerves effect paracrine regulation of epithelial function. Enteroendocrine cells are specialized cells located in the crypt region that contain various messengers released in response to lumenal stimuli. [56] These messengers relay signals to coordinate the reaction of both local and distant epithelial cells. Nerve endings of neurons of the submucosal plexus are located close to the intestinal epithelium. Neural connections also run between the submucosal and myenteric plexuses; this may be important in coordinating epithelial transport with gut motility. The central nervous system also plays a role in the regulation of fluid and electrolyte transport by way of the vagus nerve. Endocrine regulation provides distant coordination among different portions of the gastrointestinal tract and between the gut and other organs. This coordination results from the release of peptides or neurotransmitters into the bloodstream.

Immune Cells The gastrointestinal tract is extremely rich in immune effector cells, [66], [67] and this is not surprising considering its continuous exposure to bacterial, viral, and dietary antigens. Immune cells of the gut are contained primarily within the lamina propria and consist of lymphocytes, plasma cells, mast cells, and phagocytes. Classically, the mucosal immune system is broadly divided on a functional basis into afferent and efferent components, the afferent component consisting of lymphocyte-mediated antigen recognition and presentation, and cytokine production, which, in turn, recruit the efferent component consisting of mast cells, phagocytes, and their mediators. In recent years, however, the division between the afferent and efferent arms of the mucosal immune system has become blurred as it becomes more apparent that practically all cells within the intestinal mucosa are involved in both the initiation and propagation of mucosal immune responses. It is also clear that, in addition to their classical immunomodulatory functions, many cytokines produced by lymphocytes also affect both epithelial barrier function and ion transport. [68], [69] and [70] Regulation of intestinal ion transport by mast cells, phagocytes, and their mediators has also been demonstrated. [66], [71], [72]

Myofibroblasts Myofibroblasts form a continuous sheath underlying the basement membrane of the intestinal epithelial layer. Through the production of growth factors, cytokines, and extracellular matrix proteins, these cells are believed to play an important role in regulating epithelial cell growth and differentiation. [73] Studies by Berschneider and Powell and their associates [74], [75] have demonstrated that this fibroblastic sheath may also contribute to the regulation of epithelial transport. On stimulation by an array of neuroimmune mediators, myofibroblasts produce prostaglandins, which act in a paracrine fashion to stimulate epithelial Cl^- secretion. Exposure of myofibroblasts to cytokines, such as tumor necrosis factor a (TNF-a) and interleukin-1, further increases the potentiating effect of fibroblasts on agonist-stimulated Cl^- secretion, as a result of up-regulated cyclooxygenase expression. [76] Endothelial cells, which line the blood vessels within the mucosal layer, may act like myofibroblasts in that they produce prostaglandins in response to inflammatory stimuli, which could stimulate epithelial secretion in a paracrine fashion. [77]

PRINCIPLES OF EPITHELIAL TRANSPORT

Transepithelial and Housekeeping Transport

Substances may traverse the intestinal epithelium by multiple routes. Transcellular processes require entry of electrolytes across one membrane barrier, transit through the cell cytosol, and exit across the opposite membrane. The functional polarity of epithelial cells is the basis for this net (vectorial) movement of electrolytes through the asymmetric distribution of different membrane transport proteins in the opposing apical and basolateral membranes of the cells. Paracellular transport is the passive flux of electrolytes and water across tight junctions. Paracellular flux occurs entirely extracellular to the epithelial cells and can only produce vectorial transport as a dissipation of the transepithelial chemical and electrical gradients established by active transcellular transport.

Types of Transmembrane Transport

Charged species such as ions cannot traverse the lipid core of the plasma membrane. This is vital to life, because the regulation of cellular biochemistry is dependent on the tight control of intracellular ionic composition. Therefore, specialized proteins are inserted into the plasma membrane to mediate and regulate ion movement. Ions can cross the membrane through these proteins with either the consumption of cellular energy (active transport) or by flowing down existing electrical and/or chemical gradients (passive transport). Overall, transepithelial transport involves a combination of both active and passive transport mechanisms. Ultimately, all of the net transepithelial transport mechanisms discussed in this chapter require at least one active transport step to drive vectorial transport. In these cases, net passive transport of a substance through the paracellular or transcellular route occurs only in response to transepithelial electrochemical gradients established by the active transport processes.

In addition to the distinction of active versus passive transport across a membrane, all transport reactions also can be grouped into one of two classes: electrogenic or electroneutral. *Electrogenic* transport moves net charge across the membrane either by the flux of single ions (e.g., ion channels) or by combinations of transport substrates whose charge is unequal (e.g., Na^+/glucose cotransporter). Because no aqueous solution can have unequal amounts of cations and anions, electrogenic transport requires compensatory flux by another electrogenic transport process to maintain electrical neutrality. Very often, electrical neutrality is maintained by compensatory paracellular transport of an oppositely charged ion through the tight junctions. *Electroneutral* transporters also mediate a net flux of ions, but they do so without moving net charge across the membrane during a transport cycle. Electroneutral transporters may mediate an exchange of equal charges across the membrane (e.g., Na^+/H^+ exchanger), or a cotransport of equal and opposite charges (e.g., $Na^+/K^+/2Cl^-$ cotransporter). Importantly, the electrical gradient across the membrane is not a driving force affecting electroneutral carriers, and so the rate and direction of transport by electroneutral carriers are determined solely by the transmembrane chemical gradients of the transported ions. As discussed later (see the section “ Transepithelial Electrolyte Transport ”), combinations of different transport proteins can also result in overall transepithelial transport that is either electrogenic or electroneutral.

Fluid Transport

Physiology of Water Flux Water is crucial to physiological processes, and the amount of water in the intestinal lumen is closely regulated. A large amount of water is secreted by various gastrointestinal organs to facilitate digestive and other intestinal processes (see Fig. 14-1). The daily fluid load varies according to the amount and composition of meals, but it approximates 9 L. In healthy humans, 65% to 80% of the secreted or ingested water is absorbed in association with nutrient and electrolyte absorption in the small intestine, so the colon receives only 1500 to 2000 mL. [78] The colon absorbs most of this remaining fluid with high efficiency; thus, normally, fluid excretion in the stool is only about 100 mL. [79]

Molecular Mechanisms of Water Transport Researchers are still developing an understanding of how water crosses the intestinal epithelium. One general principle, however, is that water always flows across membranes in response to electrolyte and/or osmolyte fluxes. The net movement of electrolytes causes local accumulation of osmolytes, which drives compensatory water diffusion to balance osmolarity. In most cases, water molecules are moved into and out of the lumen by passive processes requiring no further input of cellular energy beyond that required to cause electrolyte transport. There are multiple routes for passive water diffusion across epithelia. If the actively transported electrolytes accumulate in the small extracellular spaces between adjacent epithelial cells (during absorption) or in the gut lumen (during secretion), there is an osmotic driving force for water to equilibrate through the tight junctions. This is believed to be the mechanism for fluid absorption in the proximal small intestine. Conversely, if the electrolyte accumulates intracellularly, there is a driving force for water to equilibrate across the cell membrane, which provides a transcellular route for water transport. The transcellular route is predicted to be more important in tight epithelia (e.g., the distal colon), because ion flow through the tight junctions is restricted in this setting. Like ions, water has difficulty crossing lipid membranes; therefore, membrane proteins must enhance water transport across these barriers. Attention has focused on two classes of proteins that increase water permeability across intestinal membranes: aquaporins and Na^+-dependent solute transporters. Aquaporins are a gene family that function as water channels in the plasma membrane. Since their initial discovery, [80] three aquaporin isoforms have been suggested to contribute to intestinal water transport. AQP3 has been shown to be abundant in the surface/villus cells of the colon, jejunum, and ileum, although whether AQP3 is apical [81] or basolateral [82], [83] remains controversial. In contrast, AQP4 has been shown reproducibly to be a basolateral protein in the small intestine and colon, but its predominant location—crypt cells or surface/villus cells—is uncertain. [82], [84] The AQP8 isoform also has been found in surface/villus cells of the jejunum and colon. [82] At present, the only functional evidence that links any specific isoform to intestinal water transport comes from AQP4 knock-out mice, which showed a modest (30% to 40%) decrease in water absorption by the proximal colon. [84] This highlights redundant pathways for intestinal water flux under normal circumstances. Another important route for transcellular water flux is through Na^+-dependent solute transporters. SGLT1 moves about 210 to 260 water molecules across the membrane each time it transports two Na^+ ions and a single glucose molecule into the cell, possibly in the form of water molecules that hydrate the cotransporter glucose-binding site. [85] Results suggest that other cotransporters also enhance water fluxes. [86] Water may be a transport substrate of SGLT1 even under isotonic conditions, [87], [88] although evidence suggests that part of SGLT1-stimulated water flow is the result of microscopic osmotic gradients driving conventional passive water diffusion. [87] Independent of the precise model, it has been estimated that the absorption of up to 5 L of water per day could be mediated by SGLT1 alone. [85] In the intestine, water transport is regulated indirectly by the regulation of electrolyte transport. When electrolytes and osmolytes remain in the intestinal lumen (e.g., lumenal lactose in lactase-deficient subjects, or during active electrolyte secretion), lumenal hyperosmolarity decreases water absorption. The opposite is also true: when molecules are absorbed and leave the lumen, they increase water absorption. Use of this osmotic driving force constitutes the basis for oral rehydration solutions, in which a low-osmolarity substrate (e.g., starch) in the lumen is digested (e.g., to glucose) and the products are avidly absorbed (e.g., by SGLT1), thus stimulating water absorption. [89]

ELECTROLYTE TRANSPORT PROTEINS

This section describes the building blocks of epithelial transport—individual membrane transport proteins. Different combinations of these transport molecules are expressed in individual cells to perform specific electrolyte transport events. Many electrolyte transport proteins have been cloned, providing unprecedented information about their structure and function. In many cases, there have also been striking advances because mutations in these proteins underlie inherited diseases of intestinal transport. The electrolyte transport proteins known to exist in gastrointestinal epithelia and their participation in transepithelial electrolyte transport are summarized in Table 14-1. The following sections emphasize cloned transport proteins that are expressed in the intestine. Because each transport protein is a member of a gene family of related protein isoforms, it is sometimes unknown which isoforms participate in particular transepithelial intestinal electrolyte transport mechanisms. Moreover, some important transport proteins remain to be cloned, and some proposed mechanisms of transepithelial transport remain more speculative than confirmed.

TRANSPORT PROTEIN	POLARITY	GENES	TRANSEPITHELIAL FUNCTION	NOTES
ATPase pumps				
Na^+, K^+-ATPase	Basolateral	α1, subunit: ATP1A1; β1, subunit: ATP1B1	Ion active transport	α1/β1 dimer is the only intestinal epithelial isoform
H^+, K^+-ATPase	Apical	α subunit: ATP4A; ATP4B?/ATP4A2	Colonic K^+ and SCFA absorption	β subunit needed for function
Exchangers and cotransporters				
Na^+/glucose cotransporter	Apical	SGLT1	Solute-coupled Na^+ uptake	Example of Na^+-coupled cotransporter
H^+/peptide cotransporter	Apical	PEPT1	Proton-coupled osmotic absorption	Example of proton-coupled cotransporter
H^+/monocarboxylate cotransporter	Apical and basolateral	MCT1	SCFA absorption?	SCFA transporter
Na^+, H^+ / $2CO_3^-$ cotransporter	Basolateral	NBC3	CO_2 and H^+ secretion	
Na^+/H^+ exchangers	Apical	NHE3	Small intestinal electroneutral Na^+ absorption	
	Apical	NHE2	(Electroneutral Na^+ absorption?); HCO_3^- secretion?	Potential role in colonic Na^+ absorption
	Basolateral	NHE1	HCO_3^- secretion	Ubiquitously expressed, cellular pH regulation
Cl^-/HCO_3^- exchangers	Apical	CLD, (AE1?)	Electroneutral Cl^- and K^+ absorption, HCO_3^- secretion	AE1 controversial
	Basolateral	AE2, (AE1?)	Electroneutral Cl^- absorption?	AE2 controversial
Na^+/Cl^- cotransporter	Unknown	NCCT	(Electroneutral NaCl absorption?)	Putative apical NaCl uptake mechanism
K^+/Cl^- cotransporter	Unknown	KCC1	Electroneutral K^+ or Cl^- absorption	Basolateral route for KCl exit
Channels				
Na^+ channel	Apical	α subunit: SCNN1A (ENaCα); SCNN1B (ENaCβ); SCNN1G (ENaCγ)	Electrogenic Na^+ absorption	Amiloride-sensitive channel; others may exist
Cl^- channels	Apical	CFTR	Cl^- secretion	cAMP activation, also conducts bicarbonate
	Apical	CLCN2	Cl^- secretion?	Ca^{2+} activated
	Apical	CIC-2	Cl^- secretion?	Outwardly rectifying; volume sensitive; Outwardly rectifying
K^+ channels	Endosomal	CIC-5	Cl^- secretion?	
	Apical	Unknown	K^+ secretion	
	Basolateral	KVLQT1, ISK1	K^+ secretion, electrogenic and solute-coupled Na^+ absorption	
HCO_3^- channel	Apical	Unknown	HCO_3^- secretion	May be CFTR

TABLE 14-1 Key Epithelial Transport Proteins and Their Participation in Transepithelial Electrolyte Transport

ATPase Pumps

Ultimately, a transport ATPase is required to provide the necessary driving force for net transepithelial ion transport to occur in mammalian cells. Among the known transport ATPases, the Na^+,K^+-ATPase and H^+,K^+-ATPase are the best understood and have well-established roles in intestinal electrolyte transport. These proteins have related sequences (60% to 70% homology), and both are members of the P-type ATPase multigene family, so named because all members of this family become phosphorylated during part of the transport cycle. Other ATPases exist, but they have less certain roles in intestinal transport physiology. Ca^{2+}-ATPase in the basolateral membrane may contribute to transcellular Ca^{2+} absorption stimulated by vitamin D. [90], [91] Deeper uncertainties exist about ion-dependent ATPases that have been identified by biochemical assays, but that have not yet been shown to mediate ion transport (e.g., HCO_3^--ATPase, Cl^--ATPase, and Mg^{2+}-ATPase).

Na^+,K^+-ATPase In the presence of Mg^{2+}, the Na^+, K^+-ATPase catalyzes the efflux of three Na^+ ions from the cell and the uptake of two K^+ ions at the expense of hydrolyzing one ATP molecule per cycle. [92] Because more cation is pumped out than is replaced, the Na^+ pump is electrogenic and generates a negative intracellular electrical potential. Na^+, K^+-ATPase is localized basolaterally and is expressed in all intestinal epithelial cells. [93] It acts to maintain a low intracellular Na^+ concentration and a high intracellular K^+ concentration. The Na^+ electrochemical gradient then is used as a driving force for Na^+ influx through Na^+ channels or through the many secondary active cotransporters and exchangers for which Na^+ is required (Fig. 14-4, Fig. 14-5 and Fig. 14-6). Inactivation of the Na^+, K^+-ATPase with cardiac glycosides such as ouabain will inhibit all these active transport mechanisms in the intestine.

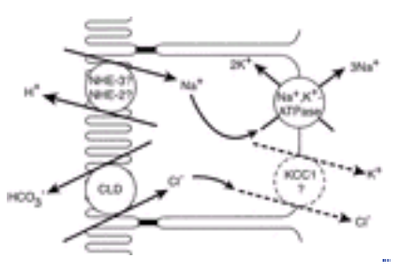

FIGURE 14-4. Electroneutral NaCl absorption in the small intestine and colon. As described in the text, apical NaCl entry through Na^+/H^+ and Cl^-/HCO_3^- exchange is the most strongly supported model in mammalian intestine, although directly coupled NaCl cotransport remains a possibility. In both models, Na^+ exits the cell through the basolateral Na^+,K^+-ATPase, but the route of Cl^-efflux is more speculative.

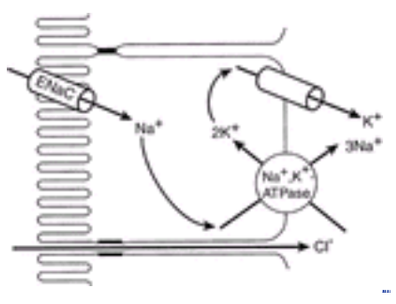

FIGURE 14-5. Electrogenic Na^+ absorption in the distal colon. Na^+ enters cells at the apical membrane through Na^+ channels and leaves cells at the basolateral membrane through the Na^+,K^+-ATPase. Details of the model are described in the text.

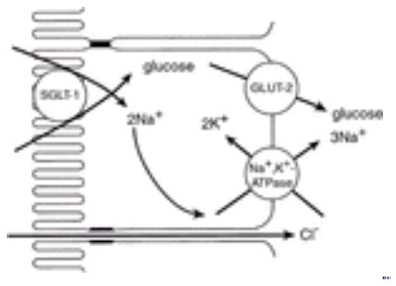

FIGURE 14-6. Solute-coupled Na^+ absorption in the small intestine. The model presents the mechanism for glucose as a generic solute and is representative of Na^+-coupled absorption of glucose, many amino acids, certain B vitamins, and bile salts. Details of the model vary among solutes because of differences in the charge of different solutes and the stoichiometry with transported Na^+ ions. Details of the model are discussed in the text.

The Na^+ pump exists in the membrane as a heterodimer of a and ß subunits, and the a1/ß1 isoforms of these subunits predominate in the intestine. [94] The a subunit is an integral membrane protein with at least eight transmembrane segments, [95] and it contains the ouabain binding site and all ion binding and catalytic sites. [96] The ß subunit has no known role in mediating ion transport, but it is an integral membrane protein essential for chaperoning the heterodimer through correct assembly and membrane targeting. [97], [98]

H^+,K^+-ATPase In several species, the apical membranes of cells in the distal colon contain H^+, K^+-ATPase activity. Similar to the gastric H^+, K^+-ATPase to which it is related, the colonic form mediates cellular efflux of H^+ into the gut lumen in exchange for K^+ uptake, with hydrolysis of ATP providing energy. [99] Activity of this pump is variably inhibited by the Na^+, K^+-ATPase inhibitor ouabain and the gastric H^+, K^+-ATPase inhibitor SCH28080, suggesting that the colonic enzyme shares common structural features with both of these pumps. [100], [101], [102], [103], [104] and [105] One reason for the variability in inhibitor responses is that a second, ouabain-insensitive K^+-ATPase probably exists. [100] Moreover, because the only cloned isoform of the colonic H^+, K^+-ATPase is not inhibited by SCH28080, there is

some controversy about whether it is the predominant apical isoform. [102] Antibodies detect H^+,K^+-ATPase protein in both the surface cells and crypt cells, although the cloned isoform may be predominantly expressed in the surface cells. [102], [105] Unlike the Na^+,K^+-ATPase, the colonic H^+,K^+-ATPase does not play a dominant role in regulating intracellular levels of its transported ions. Although cytosolic pH can be measurably affected by the activity of H^+,K^+-ATPase, other transporters (e.g., Na^+/H^+ exchangers) are likely to have greater impact on control of the resting cytosolic pH. [106], [107] In the distal colon of the guinea pig, protons secreted by the colonic H^+, K^+-ATPase are important for stimulating uptake of lumenal short-chain fatty acids (SCFAs), facilitating absorption of SCFAs in their nonionized (i.e., protonated) form. [108] The colonic H^+, K^+-ATPase also is important for K^+ absorption, especially in infant animals who need to retain more K^+ than adults to avoid growth retardation. [109] Both an a and a ß subunit of the colonic H^+,K^+-ATPase have been cloned. [110], [111] Coexpression of the a subunit with either the cloned HKß or a heterologous ß subunit (either from gastric H^+,K^+-ATPase or Na^+,K^+-ATPase) is sufficient to reconstitute observable H^+,K^+-ATPase activity in the plasma membrane. [112], [113] Evidence suggests that the ß subunit of the Na^+,K^+-ATPase may serve this role physiologically, as it associates with the HKa subunit in the apical membrane of colonocytes. [114] The a subunit is predicted to have multiple transmembrane spanning segments, [110] but no detailed mapping of functional sites has been reported.

Exchangers and Cotransporters

Using a limited number of ATPase pumps, abundant assortments of electrolytes and solutes are actively transported across the intestinal epithelium. This is possible because many different carrier proteins convert the energy of Na^+ or proton gradients into net transport of other electrolytes and solutes. In addition to these secondary active exchangers and cotransporters, the passive transporters play a key role in facilitating movement of ions across the plasma membrane at faster rates than could be expected by simple diffusion.

Na^+-Solute Cotransporters Many food-derived products including glucose, amino acids, and vitamins, as well as endogenous lumenal solutes such as bile acids, are taken up by specific Na^+-coupled cotransport proteins in the apical membrane of enterocytes. These cotransporters are the predominant route by which such diverse substances enter the body and are also responsible for driving the majority of Na^+ and water absorption after a meal. Most of these cotransporters are found in the small intestine, where organic nutrients are predominantly absorbed. Among the mammalian Na^+-coupled nutrient transporters, the intestinal Na^+/glucose cotransporter (SGLT1) was the first to be cloned and has been studied the most extensively. [115] Each transport cycle of the protein results in the uptake of two Na^+ ions and one glucose molecule. Its electrogenic nature renders the cotransporter sensitive to changes in membrane potential. [116] SGLT1 functions as a single polypeptide chain, but its topology is controversial. Between 11 and 14 transmembrane segments have been proposed, and results have conflicted in assigning the C-terminal to either the intracellular or extracellular domain. [117], [118] Despite these uncertainties, studies of chimeras between SGLT1 and a related cotransporter (SGLT2) have ascertained that the C-terminal portion of SGLT1 contains the sugar-binding site. [119] Further, a naturally occurring missense mutation near the C-terminal (R499H) was found in a patient with glucose-galactose malabsorption, and the mutant protein had a tenfold lower sugar affinity than the wild-type protein but no alteration in Na^+ affinity. [120] Similar approaches are being used to evaluate the mechanisms certain cloned intestinal Na^+/amino acid, Na^+/bile acid, and Na^+/vitamin cotransporters. [116] These Na^+-coupled nutrient cotransporters are predicted to contribute to water and Na^+ absorption in the intestine, but they are discussed in more detail in Chapter 16, Chapter 19, and Chapter 20.

H^+-Solute Cotransporters The intestinal proton-coupled peptide cotransporter (PEPT1) is expressed in the apical membrane of small intestinal cells and can transport a wide variety of dipeptides and tripeptides. The cloned transporter is electrogenic, and the proton activation kinetics suggest a 1:1 stoichiometry of proton:dipeptide transport for uncharged peptide substrates. [121], [122] From the standpoint of fluid and electrolyte absorption, the major contribution of PEPT1 is to stimulate water transport. PEPT1 remains an enigma for transport physiologists because of its capacity to accommodate an extremely broad range of peptide substrates. The molecular basis of this promiscuity is not fully understood. A cloned proton-coupled monocarboxylate cotransporter (MCT1) has been identified in membranes of both small and large intestine epithelial cells. [123] MCT1 functions to reproduce nonionic diffusion through coupling flux of monocarboxylate anions and protons. [124] The transporter is inhibited by a-cyano-hydroxycinnamates, and lactate is a favored substrate. [124] Evidence suggests a physiological role for colonic MCT1 as a route for basolateral SCFA flux from cells as part of transcellular SCFA absorption, but there are conflicting reports about the ability of a-cyanohydroxycinnamates to affect SCFA fluxes in colonocytes [125], [126] and the localization of MCT1 to either the apical or basolateral membrane. MCT1 may be most important physiologically in the transport of monocarboxylates that are too hydrophilic for efficient flux by nonionic diffusion through the lipid bilayer.

$Na^+/K^+/2Cl^-$ Cotransporter $Na^+/K^+/2Cl^-$ cotransport is present in the basolateral membrane of certain intestinal epithelial cells, and it participates in electrolyte secretion. This cotransport is electroneutral because each transport cycle moves equal numbers of cations (1 Na^+ plus 1 K^+) and anions (2 Cl^-) into the cell. The $Na^+/K^+/2Cl^-$ cotransporter plays a key role in Cl^- secretion as the predominant route for basolateral Cl^- uptake, [127] and it also participates in K^+ secretion by providing a second route for basolateral K^+ uptake in addition to the activity of Na^+,K^+-ATPase. [128], [129] The $Na^+/K^+/2Cl^-$ cotransporter isoform NKCC1 is expressed in predominantly the basolateral membrane of human colonic intestinal epithelial cells. [130], [131] It functions as a single polypeptide chain, and it has 12 putative transmembrane segments. [130] NKCC1 is inhibited by bumetanide and furosemide, which affect anion binding to the transporter. [132] A distinct Na^+/Cl^- (or $Na^+/K^+/Cl^-$) cotransporter has been proposed to exist in the apical membrane of enterocytes, as part of the electroneutral Na^+ absorptive mechanism. Similar apical cotransporters have been cloned from the kidney medulla, where they participate in Na^+ reabsorption, [133] and one highly homologous isoform (with as yet untested function) is expressed in the human small intestine and colon. [134] However, although strong evidence supports the physiological importance of apical Na^+/Cl^- cotransport for Na^+ absorption in winter flounder intestine, [135], [136] mammalian intestinal epithelial cells likely use other mechanisms to mediate electroneutral Na^+ uptake (see the following sections).

Na^+/H^+ Exchangers Na^+/H^+ exchange activity is present in all segments of the small intestine and colon. It is defined as the tightly coupled uptake of one Na^+ ion in exchange for the efflux of a proton. [137], [138] This electroneutral exchange reaction is important for intracellular pH regulation, as well as transepithelial Na^+ absorption. Na^+/H^+ exchange is observed in both the apical and basolateral membranes of epithelial cells from small intestine [139] and colon. [140], [141], [142] and [143] Four cloned isoforms of the Na^+/H^+ exchanger (NHE) gene family have been detected in gastrointestinal epithelia. The NHE1 isoform is present in virtually all cells of the body and is believed to be responsible for cellular pH regulation. [144] NHE1 is found in the basolateral membrane of intestinal epithelial cells. [145] NHE2 and NHE3 are predominantly epithelial isoforms of the exchanger that are expressed in the apical membranes of intestinal cells. [146] NHE4 is also expressed in gastrointestinal tissues (predominantly stomach) but there is no evidence that NHE4 contributes to transport responses in the small or large intestines. Each epithelial NHE isoform has a distinct distribution of mRNA abundance along the small and large intestines, which varies among species. [147], [148] There are also gradients of NHE isoforms and Na^+/H^+ exchange function along the crypt-to-villus and crypt-to-surface axes. Based on immunoreactivity, NHE1 is expressed at all sites along these axes, [145] but NHE3 is expressed preferentially on the villus in the small intestine and on the surface and in the upper third of the colonic crypts. [149] In contrast, functional apical Na^+/H^+ exchange activity has been observed along most of the ileal villus-crypt axis using isolated membranes [139] and in the colonic crypts using optical approaches to study either crypts in situ or in isolation. [150], [151] All NHE isoforms have 10 to 12 predicted transmembrane segments with cytoplasmic C- and N-termini. [146] Overall, NHE isoforms are 50% to 60% homologous within a species, with the C-terminal cytoplasmic tail having the most sequence divergence. The C-terminal portion is important for second messenger regulation of NHE isoforms, and several portions of this domain have been identified as crucial for mediating the effects of Ca^{2+}, calmodulin, cAMP, activation by intracellular protons, and growth factors. [146], [152], [153], [154] and [155] Evidence also suggests that, in some cases, NHE regulation requires accessory phosphoproteins, because signaling pathways do not always alter phosphorylation of NHE proteins. [156] As demonstrated by kinetic studies, all isoforms have different affinities for Na^+ and different inhibition constants for amiloride and other inhibitors, reflecting their different primary structures. [146] NHE2 and NHE3 are apical membrane proteins in intestinal cells, [146] and therefore they may contribute to Na^+ absorption. Several results suggest that NHE3 probably is responsible for electroneutral Na^+ absorption in the small intestine. First, mineralocorticoids increase the rate of ileal brush border Na^+/H^+ exchange; this correlates with an increase in the abundance of NHE3 mRNA without an effect on mRNA for NHE1 or NHE2. [157] Second, the dose-dependent inhibition of Na^+ and water absorption in dogs by dimethylamiloride is most consistent with NHE3 inhibition. [158] Finally, the patterns of second messenger regulation of Na^+ absorption and NHE3 expression are closely matched (i.e., inhibition by Ca^{2+} and stimulation by phorbol esters), but differ from the responses of NHE2. [146] Conversely, in rabbit ileum, evidence suggests that NHE2 and NHE3 each contribute about equally to basal Na^+ uptake in brush border vesicles. [159] Deeper controversy is emerging about the contribution of each NHE isoform to colonic Na^+ absorption. Evidence suggests that colonic NHE2 may play a role in regulating this process. [160], [161] In rat proximal colon, evidence shows that NHE2 is the predominant contributor to basal Na^+ absorption, [162] although the role of NHE3 seems more dominant in isolated membrane vesicles from the same tissue. [160], [163] Overall, it appears that the role of NHE3 predominates, at least in mice, because NHE3 knock-out mice have obvious diarrheal disease, whereas NHE2 knock-out mice have no identified intestinal disorder. [164], [165]

Cl^-/HCO_3^- Exchangers Cl^-/HCO_3^- exchange in the apical membrane acts in concert with Na^+/H^+ exchange to mediate electroneutral NaCl uptake in colonocytes

and small intestine cells. [152], [166] Unlike the apical membrane Na^+/H^+ exchange, which is expressed predominantly in villus cells of the small intestine, Cl^-/HCO_3^- exchange appears to be equally prevalent in the apical membranes of both crypt and villus cells, suggesting that it contributes to other transport mechanisms in addition to NaCl absorption. [139] In the colon, apical Cl^-/HCO_3^- exchange is observed predominantly in the surface cells, although Cl^-/OH^- exchange remains relatively active in crypt membranes. [167] This and other kinetic evidence suggests that apical Cl^-/HCO_3^- and Cl^-/OH^- exchange may not be mediated by the same protein in either the small intestine or colon. [166], [167] and [168] Molecular biologic approaches have identified many candidate proteins that may perform the apical Cl^-/HCO_3^- exchange reaction. Multiple isoforms of the anion exchanger (AE) gene family are expressed in the small and large intestines. Among the three known AE2 transcripts (AE2a, AE2b, AE2c1), AE2a and AE2b are abundant in the intestine, and, like most AE2 epitopes, they are located basolaterally. [84], [169], [170] Results have also suggested the presence of an apical AE1 isoform and a basolateral AE3 isoform in the intestines, but their existence remains highly controversial. [170], [171] The function and localization of these and other unidentified AE isoforms remain to be confirmed. A more definite role in intestinal Cl^- absorption has been assigned to the CLD gene product (formerly called DRA). Mutations in CLD are responsible for congenital chloride diarrhea, [172] a disease characterized by a chloride-rich, acidic stool. The protein is known to be expressed in the apical membrane of colonocytes. [173] CLD is the strongest candidate for an apical Cl^-/HCO_3^- exchanger in the small intestine and colon, with ion transport measurements demonstrating that CLD is a Cl^-, HCO_3, and divalent anion transporter. [174], [175] and [176] CLD mutations identified in patients with congenital chloride diarrhea do, in fact, lead to defective anion transport. [176]

Other Exchangers and Cotransporters In addition to those described earlier, other intestinal electrolyte cotransporters have been cloned, including Na^+/PO_4^- and Na^+/SO_4^- cotransporters. [177], [178] However, it remains to be established whether these proteins contribute to the observed active transport of SO_4^- and PO_4^- across the intestinal epithelium. [179] An intestinal K^+/Cl^- cotransporter has also been cloned. [180] This cotransporter is a logical mechanism for electroneutral basolateral exit of K^+ and Cl^- from intestinal epithelial cells that absorb K^+ and/or Cl^- in an electroneutral fashion, and its expression increases during dietary potassium depletion. [181], [182] In addition, numerous transport proteins can be defined functionally in isolated membrane vesicles, but are not yet cloned. These include $SCFA/HCO_3^-$ exchange and an electrogenic Na^+-HCO_3^-(OH^-) cotransporter. The roles of these transport reactions in epithelial transport remain to be established.

Ion Channels

An ion channel acts as a gated pore that is selective for certain ions. [183], [184] Open channels are passive pathways for electrolyte movement. *Gating* controls the amount of time that the pore spends in an open versus closed configuration. A single open channel conducts a rapid flux of ions across the membrane with a characteristic *unitary conductance* (one pS ~500 univalent ions per millisecond). The conductance of a given channel is an intrinsic biophysical signature, whereas gating of epithelial channels is most often regulated by second messengers. Compared with ion channels in excitable cells, membrane voltage has only modest effects on gating of epithelial channels, although this should be distinguished from the large effect of membrane voltage on the magnitude of ionic currents across the open channel, a completely separate feature.

Na^+ Channels Apical Na^+ channels contribute to electrogenic Na^+ absorption in many distal colonic epithelia. Epithelial Na^+ channels are selective for Na^+ (>10:1 versus K^+), have 5 pS unitary conductances, and are *inwardly rectifying* (i.e., they mediate cation efflux less efficiently than cation influx). [185], [186] Epithelial Na^+ channels are inhibited by nanomolar concentrations of phenamil and amiloride analogs, but the profile of inhibitor sensitivity is distinct from that of Na^+/H^+ exchangers. [186] Studies show that not all electrogenic Na^+ flux is equally sensitive to the same channel inhibitors. [187], [188] and [189] It is currently unclear whether this is the result of different regulatory or structural states of the known epithelial Na^+ channel, or whether a second biochemically distinct and amiloride-insensitive Na^+ channel exists in some portions of the colon. [190] The amiloride-sensitive epithelial Na^+ channel (ENaC) is composed of three subunits: a, ß, and ?. The subunits share 35% homology, and each is an integral membrane protein predicted to span the membrane twice. [191], [192] Expression of the ß and ? subunits is required to target the channel to the plasma membrane efficiently. [192] This is consistent with the observation that certain activating mutations in the ß or ? subunits (see the later discussion of Liddle syndrome) cause increased expression of Na^+ channels in the membrane as well as increased activity of the expressed channels. [193], [194] In contrast, a glycine near the N-terminal of the a subunit is involved in channel gating. [195] Although the predicted minimum configuration of the epithelial Na^+ channel is a heterotrimeric protein, it is likely that the native channel is more complex. Biochemical purification suggests that five or six polypeptides of widely different molecular weight are components of the channel. Some of these components may be differentially glycoslyated forms of the known subunits, but some of the peptides have been identified as regulatory proteins and cytoskeletal elements that modify channel function. [190]

Cl^- Channels Apical Cl^- channels are essential components of Cl^- secretion. Cl^- secretion in all parts of the intestine and colon is controlled predominantly by the ability of specific second messengers to regulate channel gating and localization. However, multiple Cl^- channel types are observed in intestinal epithelial cells, sometimes making it difficult to assign a functional role to individual proteins. A cAMP-activated channel in the apical membrane is known to participate in hormone-stimulated Cl^- secretion, and it is encoded by the cystic fibrosis transmembrane conductance regulator gene (*CFTR*) that is responsible for cystic fibrosis. [196] The normal CFTR channel is selective for Cl > Br > I, has a unitary conductance of 8 to 10 pS, and is *nonrectifying* or *linear* (i.e., it conducts ions with equal efficiency in either an inward or outward direction). [197] The CFTR protein also is able to mediate the conductance of other ions (notably HCO_3^-) [198], it participates in a subset of cAMP-regulated apical membrane endocytosis and exocytosis events, [199] and it regulates the activity of other Cl^- and Na^+ channels. [200], [201], [202] and [203] The CFTR channel is a single polypeptide with 12 membrane-spanning domains, two cytoplasmic nucleotide-binding domains, and a “regulatory” (R) domain that contains most of the phosphorylation sites. [196], [204] It has been shown that CFTR Cl^- channel gating is complex, involving coordinated phosphorylation of multiple R-domain serine residues and ATP hydrolysis at, and interaction between, the two nucleotide-binding domains. [204] In healthy intestine, agonists acting through cAMP or Ca^{2+} stimulate Cl^- secretion. However, in patients with cystic fibrosis, neither cAMP nor Ca^{2+} is able to elicit Cl^- secretion. [205] This is in contrast to airway epithelia of patients with cystic fibrosis, where a Ca^{2+}-activated Cl^- channel functions in the absence of normal CFTR. [206] The reasons underlying the failure to detect such a Ca^{2+}-activated channel in the intestine of patients with cystic fibrosis is not yet understood at a molecular level, but it could involve the lack of an appropriate channel protein, a defect in the ability of CFTR to regulate another anion channel, and/or the existence of negative regulatory mechanisms in the intestinal epithelium (see the section “ Regulation of Electrolyte Transport ”). Molecular cloning has revealed other candidate Cl^- channels. ClC-2 and ClC-5 are cloned Cl^- channels that are expressed in intestine. [207], [208] and [209] ClC-5 is particularly intriguing because it is expressed only in kidney and colon and it has a permselectivity of I>Cl, is outwardly rectifying, but is not activated by cAMP. [207] Because ClC-5 is located in endosomes of intestinal epithelial cells, [209] it could be inserted into the plasma membrane upon appropriate regulatory signals. ClC-2 is expressed in the apical membrane of intestinal epithelial cells, and it may contribute to Cl^- secretion. [210], [211] Because ClC2 was apparently operational in CFTR knock-out mice, it is unlikely to be one of the proteins that requires CFTR to function. [211] /SUP>ClC channels are activated by cell swelling and, at a minimum, probably participate in the ion efflux necessary for cell volume regulation. [212], [213]

K^+ Channels K^+ channels in the apical membrane of epithelial cells are involved in K^+ secretion, and K^+ channels in the basolateral membrane are involved in Cl^- secretion. K^+ channels also are involved in electrogenic Na^+ absorption and Na^+-coupled solute absorption, but the polarity of involved K^+ channels involved in these processes is less certain. The characterization of apical K^+ channels has come predominantly from biophysical and physiological studies. The presence of apical K^+ channels has been inferred indirectly by adding lumenal K^+ channel blockers (e.g., barium and tetraethylammonium [TEA]) and monitoring the effects on global electrical properties of the rat colonic mucosa. [214], [215] Treatment with aldosterone increases the apical K^+ conductance, suggesting an increase in K^+ channel activity or number. [216] A preliminary report identified an apical 210-pS K^+ channel in rat distal colon as a candidate for the predominant apical K^+ conductance. [217] Basolateral K^+ channels have been studied more extensively. Patch clamping has identified a basolateral channel in human and rat colonic crypts that was selective for K^+ (49:1 versus Na^+), and it had a unitary conductance of 23 pS. [218], [219] Open probability of this channel was increased four- to tenfold by cAMP or micromolar Ca^{2+}, and the channel was blocked by quinidine and diphenylamine carboxylate (DPC), but not by TEA. [218], [220] The second messenger sensitivity of the 23-pS channel suggests that it contributes to both Ca^{2+}- and cAMP-stimulated Cl^- secretion. A second K^+ channel with unitary conductance of 138 pS has also been observed in human colonic crypts. The 138-pS channel has a distinct inhibitor profile; it is inhibited by TEA and quinidine, but not DPC, and it is highly K^+ selective (190:1 K:Na). [220] rSK1 and KVLQT1 channels have emerged as basolateral K^+ channels in the colon that respond to Ca^{2+} and cAMP, respectively. [221], [222] /SUP>

TRANSEPITHELIAL ELECTROLYTE TRANSPORT

The preceding section reviewed th*transmembran*e transport proteins that serve as basic building blocks for intestinal electrolyte transport of +, K +, Cl -, and HCO_3^-. This section discusses how these proteins are combined to produce the major *transepithelia* electrolyte transport events observed in the intact intestine.

In general, net transepithelial transport requires the participation of at least two transport proteins to mediate ion uptake across one plasma membrane domain and exit of ions across the plasma membrane domain at the opposite pole of the epithelial cell. By definition, active transepithelial transport also requires involvement of a primary active transporter to provide the energy for net absorption or secretion of the ion. When the pump also serves as an uptake or exit step for the ion in question, only two transport

proteins are required. Net transepithelial electrolyte transport across the epithelium can be either electrogenic or electroneutral. However, transepithelial transport mechanisms can be electrogenic only when both the apical and basolateral membranes mediate electrogenic ion fluxes as part of the mechanism, or when transcellular fluxes drive electrogenic paracellular fluxes. For instance, the electrogen+,K +-ATPase is involved in both electroneutral and electrogenic N+ absorption, depending on the accessory Na + transport events.

The voltage clamp and Ussing chamber are used in combination to define the presence of active electrolyte transport between the lumenal and serosal surfaces of a tissu*Ussing chambe*r permits mounting of intestinal tissue (or a cultured cell monolayer) between two compartments to measure transport between the lumenal and serosal reservoirs. If tissue in an Ussing chamb*voltage clamped* eliminating the driving force for transepithelial ion movement (i.e., transepithelial voltage is clamped to zero), the magnitude of the current required to offset the potential difference (term*short circuit current* or I_{sc}) will reflect active, net electrogenic ion transport. In the same configuration, measurement of net transepithelial ion fluxes with isotopes can be used to detect transport of specific ions that is either electrogenic or electroneutral.

Classically, absorptive and secretory functions of the intestine are considered independent, and absorptive function may remain intact during periods when the secretory function is excessively stimulated. This is a clinically important concept because some antidiarrheal strategies (e.g., oral rehydration solutions) rely on stimulating absorption in the presence of abundant secretion. However, more recent evidence suggests that there are functional, and potentially molecular, linkages between the absorptive and secretory mechanisms. As discussed previously, more recent data have challenged the long-held dogma that villus cells are purely absorptive and crypt cells are purely secretory, and they have confirmed the presence of at least a subset of cells that mediate both absorption and secretion (at least in the colon).[223] Further, evidence that expression of the CFTR C- channel affects activity of the epithelial N+ channel provides an explanation for the observation that patients with cystic fibrosis have increased electrogenic Na+ absorption.[203] Thus, regulation of Cl- secretion and Na+ absorption can be intertwined, and cells that express machinery for both secretory and absorptive functions may have a significant role + absorption.

Electrolyte Absorptive Mechanisms

The small intestine performs electroneutral NaCl absorption and N+-coupled nutrient absorption, and it is responsible for most of the absorption of nutrients and water by the intestine as a whole+ absorptive mechanisms in the colon vary considerably as a function of both species and segment; however, in most species including human, the large intestine absorbs+ avidly through both an electrogenic mechanism involving apical N+ channels and an electroneutral NaCl absorptive mechanism similar to that in the small intestine.[4, 63, 224, 225] The colon also is responsible for SCFA absorption and some + absorption, and it is essential for conservation of fluid and electrolyte[4, 226]

Electroneutral NaCl Absorption A significant fraction of the Na+ and Cl- absorbed by the intestinal tract is electroneutral and is mutually dependent on the alternate ion (i.e., electroneutr+ absorption requires the presence of C- and vice versa).[227] Postprandially, after the absorption of nutrients has been completed, or in the fasting state, electroneutral NaCl absorption is the major route f+ absorption in the small intestine[158] This NaCl absorptive mechanism is also the principal route for N+ absorption in the proximal colon, with less prominence in the distal colc[4] The relative importance of electroneutral versus electrogenic N+ absorption in the colon also varies as a function of mineralocorticoid statu[4] To explain these coupled fluxes, two models of NaCl uptake in the apical membrane have been proposed: an electroneutral+/Cl - cotransporter[228] and the combined action of Na+/H + and Cl-/HCO_3 - exchangers. In either model, it is generally agreed that the I+,K +-ATPase in the basolateral membrane serves as the exit step for N+. Figure 14-4 outlines the dual exchanger model, which is the most widely accepted model for human NaCl absorption at this time. Abundant information supports the existence of electroneutral NaCl absorption composed of a Na+/H + exchange working in parallel with a C-/HCO_3 - exchange pathway[229, 230] Both Na +/H + exchange and C-/HCO_3 - exchange are present in vesicles prepared from the apical membranes of rabbit ileal villus c[139] In this model (see Fig. 14-4), coupling of the paired exchangers occurs through changes in intracellular pH. For example, the + gradient drives Na+ uptake and H+ efflux by Na+/H + exchange, which alkalinizes the cytoplasm and increases the activity of the Cl -/HCO_3 - exchanger. The action of carbonic anhydrase produces HCO_3^-, which then leaves the cell in exchange for uptake of lumenal -. The net reaction is Na+ and Cl- uptake in exchange for H+ and HCO_3 - efflux. Once in the cell, Na+ is pumped out by Na +,K +-ATPase, and Cl- follows by way of an electroneutral transport protein that has yet to be identified, but it is likely to be+/Cl - cotransporter.[180]

Electrogenic Na+ Absorption In humans, electrogenic, amiloride-sensitive N+ absorption accounts for about 50% of the N+ reabsorbed in the distal colon. There is a decreasing gradient of activity through the transverse colon and only minimal levels in the proximal colon.[224, 225] Aldosterone, a mineralocorticoid well known for its N+-retaining and K+-wasting effects on the distal nephron, also exerts profound effects on the distal colon. Induction of secondary hyperaldosteronism by the feeding of a low-sodium diet or administration of exogenous aldosterone converts the rat distal colon from electroneutral, NaCl absorption to amiloride-sensitive, electroge+ absorption.[231, 232 and 233] A similar induction of electrogenic Na+ absorption occurs in certain other species. Figure 14-5 shows the proposed mechanism of electrogenic N+ absorption by the surface epithelium of the distal colon. Lumenal N+ enters colonocytes by a Na+ channel in the apical membrane and exits by the basolateral I+,K +-ATPase. In the process of driving transepithelial Na+ absorption, Na+,K +-ATPase also catalyzes K+ uptake and creates a charge imbalance, and both effects must be compensated to sustain + absorption. The compensatory ion flux is probably achieved by the electrogenic efflux of K+ through K + channels. Whether apical or basolateral I+ channels perform this function remains controversial. It is likely that apical+ conductance is predominantly involved in + secretion, a crypt cell function that is not linked to N+ absorption[128, 234, 235] The simplest model suggests that basolateral I+ channels provide the compensatory flux and recycle + across the basolateral membrane. Finally, paracellular flux of - is driven by Na+ movement to restore electroneutrality between lumenal and serosal compartments.

Solute-Coupled Na+ Absorption The absorption of many nutrients, and also of bile acids (in the terminal ileum), is mediated by+-coupled cotransporters in the small intestine. The combined action of these transporters contributes significantly to+ and water absorption.[236] A common cellular mechanism (Fig. 14-6) is believed to mediate all solute-coupled N+ absorption that occurs in villus cells. Lumenal N+ enters through apical Na+-coupled cotransport and exits through the basolateral N+,K +-ATPase. The Na+-solute cotransporter uses the energy of the electrochemical gradient for I+ to drive intracellular accumulation of the cotransported solute above its equilibrium value. When two+ ions are coupled to the uptake of a single solute molecule (e.g., Na +/glucose cotransporter), the intracellular accumulation of solute can be nearly 100-fo[237, 238] Efflux of the solute down its steep concentration gradient across the basolateral membrane is then efficiently mediated by facilitated diffusive carriers, which are passive. For example, the GLUT2 passive glucose transporter performs this function for glucose absorpt[239] Electrogenic Na+ absorption stimulates absorption of a compensatory - anion by the paracellular route, and recycling of + through basolateral K+ channels ensures cellular electroneutrality.

SCFA Absorption and SCFA-Stimulated Na+ Absorption SCFAs are produced in the colon by bacterial catabolism of unabsorbed carbohydrate and protein. The combined concentration of the predominant lumenal SCFAs (acetate, propionate, and butyrate) is 100 to 150 mM, so they are the major anions and osmolytes in the colonic lum[226] Absorbed SCFAs account for 7% to 10% of ingested calories, and they serve as an important energy source for colonic epithelial c[240] SCFAs also stimulate electroneutral Na+ absorption, up to fivefold in humans[241] SCFAs stimulate Na+ absorption by activation of apical N+/H + exchange in colonocytes that leads to the subsequent exit of I+ through the basolateral Na+,K +-ATPase. Evidence suggests that lumenal SCFA uptake causes intracellular acidification and lumenal alkalinization, which are both known activators c+/H + exchangers.[242, 243] The mechanisms of SCFA uptake that lead to the change in pH are controversial, but it appears that both nonionic diffusion (uptake of the protonated form of these weak acids without intervention of a transport protein) and carrier-mediated transport by anion transporters (e.g., $SCFA_3$ - exchange, MCT1) both play a role[244, 245, 246 and 247] The ability of SCFA-induced acidification to stimulate apical, but not basolateral, N+/H + exchangers of colonocytes has been perplexing. The regulation of extracellular pH in microenvironments directly adjacent to colonocyte membranes may partially expl[248, 249] Physiological SCFA gradients cause alkalinization of the lumenal surface and acidification of the basolateral surf[242] which preferentially activates apical but not basolateral N+/H + exchange.[243] There are several candidate mechanisms for SCFA efflux across the basolateral membrane including nonionic diffusion, SCFA anion transport by SCFA/HCO_3 - exchange, and SCFA/H+ cotransport by MCT1.[124, 250]

K + Absorption In human metabolic balance studies, about 85% of ingested + is absorbed in the small intestine, with passive absorption driven by prevailing electrochemical gradients being sufficient to explain this u[62] In contrast, active electroneutral K + absorption occurs in the distal colon[99, 251] and it is believed to mediate absorption of 5% to 7% of + ingested daily,[62] leaving 3% to 5% to be lost in fecal water. The absolute magnitude of colonic+ fluxes are underestimated by these values because colonic K + secretion also occurs. No single model of colonic I+ absorption predominates. An apical I+,K +-ATPase promotes uptake of I+ from the lumen.[100, 101] Because evidence suggests that not all apical I+ uptake is coupled to proton efflux, there may be other modes of I+ entry as well.[252] K + likely leaves the cell by the I+/Cl - cotransporter KCC1, shown to be expressed in the intestin[253] Because not all K+ absorption is Cl- dependent, there is likely to be a second (or alternative) route for basolateral K+ efflux that is not as tightly coupled to C- anions.[4] The Cl --dependent portion of K+ absorption probably uses apical C-/HCO_3 - exchange to neutralize protons extruded by the +,K +-ATPase, providing a source of intracellular C- to drive the basolateral K+/Cl - cotransporter. Evidence also has shown that SCFA uptake by the guinea pig distal colon requires activity of tI+,K +-ATPase; therefore, an alternative model may exist in which+ absorption regulates SCFA fluxes and vice vers[108]

Genetic Defects in Electrolyte Absorptive Processes

Defects in NaCl absorption. Congenital chloride diarrhea produces profound C--rich, acidic diarrhea commencing at birth, resulting in systemic hypochloremic, hypokalemic acidosis with volume depletion. First described in 1[254, 255] the disease has been described in more than 60 children. The histology of the intestinal mucosa is found to be normal on routine examination. Intubation studies have revealed that the transport defect is limited to the ileum ar[256] Na + absorption against a chemical gradient remains intact and is accompanied by I+ secretion.[257, 258] The gene that causes congenital chloride diarrhea*CLD*) has been cloned.[172] Although the disease can be accounted for by a defective -/HCO_3 - exchanger, the defect may be more subtle than a total lack of anion exchange. Indeed, increases in lumenal HCO_3 - concentrations increase C- secretion in these patients, suggesting that residual C-/HCO_3 - exchange may still occur[256, 257] It is unclear whether this is the result of an alternative anion exchange protein in the apical membrane, or whether it is residual CLD function. Congenital sodium diarrhea is an even rarer disorder. This disease produces a sodium-rich, alkaline stool and results in systemic acidosis. Intubation studies have revealed that the usual inverse relation between N+ and H + fluxes does not hold, suggesting a defect in N+/H + exchange.[259] This defect has been confirmed in studies demonstrating a clear lack of proton-driven + uptake in jejunal vesicles[260] Field and Semrad[1] reported that the distribution of NHE3 mRNA does not correspond to the site of the transport defect in congenital sodium diarrhea, tentatively suggesting that anoth+/H + exchanger isoform may be affected. However, a genetic study failed to find an association with mutations of NHE1, NHE2, NHE3, or NHE5[261] demonstrating that the defect is associated with a yet uncloned NHE isoform or with an essential regulator of a known apical NHE.

Defects in electrogenic Na+ absorption. Liddle syndrome is an autosomal dominant disorder leading to salt-sensitive hypertension. It has been shown to be caused by mutations in the amiloride-sensit+ channel that is present in both distal colonic and renal epithelia.[262] The disorder leads to increased expression and activity of N+ channels because of truncation or frameshift mutations in the cytoplasmic C-terminal tail of a or ? subunits of the chan[193, 194] Curiously, no disorders of intestinal electrolyte absorption have been reported in Liddle syndrome despite severe defects in renal function, suggesting that down-regulation of alternati+ absorptive processes in the colon may compensate for any increased electrogenic absorption; however, this hypothesis remains untested. At least a subset of type 1 pseudohypoaldosteronism (the autosomal recessive version) is also caused by mutations in the epithe+ channel. In contrast to the gain-of-function mutants seen in Liddle syndrome, the mutations in type 1 pseudohypoaldosteronism diminish channel function and cause salt wasting. Intestinal malfunctions have been noted in this disease, including elevat+ levels in the stoo[263] and a lack of colonic responsiveness to mineralocorticoid[264] The disorder can be caused by frameshift, premature termination, and missense mutations in the a, ß, or ? subunits of the chan[265, 266] Cystic fibrosis leads to an increase in electrogenic N+ absorption, although cystic fibrosis is primarily a defect in C- secretory function caused by malfunction of a C- channel (CFTR). However, the expression of normal CFTR protein is thought to limit I+ channel activity, and the lack of CFTR (or presence of mutant CFTR) therefore removes an inhibitory influence on the amiloride-sensitive Na+ channel.[203] An activated CFTR Cl- channel is required to mediate the inhibitory regulatio[202, 203, 267]

Defects in nutrient-coupled Na+ uptake. Glucose-galactose malabsorption is a rare, autosomal recessive disorder in which +-coupled uptake of glucose and galactose is defective. Food ingestion leads to osmotic diarrhea, which can be treated by eliminating glucose and galactose from the diet. Children who have this disorder can maintain normal+ balance without glucose or galactose in their diets, illustrating the reserve capacity of the distal small intestine and colon for water ar+ absorption. Screening of 33 patients identified multiple mutations in SGLT1. These mutations caused transport defects resulting from impaired trafficking of SGLT1 to the plasma membrane and/or defects in glucose transport ki[120, 268]

Electrolyte Secretory Mechanisms

Secretory mechanisms throughout the gastrointestinal tract center around the - anion. HCl is the major secretory product of the stomach. In other intestinal segments, the predominant secreted ion is eith- or HCO_3 -.[269, 270] However, HCO_3 - secretion may be related to, or require, active C- secretion.[271, 272]

Electrogenic Cl- Secretion Electrogenic Cl- secretion is found in all segments of the gastrointestinal tract from the duodenum to the distal colon, presumably reflecting the common need for a mechanism to maintain hydration of the lumenal c Figure 14-7 introduces the model of C- secretion. Uptake of C- across the basolateral membrane is through the electroneutral N+/K +/2Cl - cotransporter.[127] A key function of the Na+/K +/2Cl - cotransporter is to use the energy of the N+ gradient to accumulate Cl - intracellularly above its electrochemical equilibrium. Under these conditions, - will exit the cell across the apical membrane when - channels are opened. Na+,K +-ATPase provides energy for this overall mechanism and recycles N+ across the basolateral membrane. K+ channels in the basolateral membrane allow for + recycling, maintain cellular electroneutrality by compensating for - efflux, and keep the cell hyperpolarized (i.e., negative intracellular voltage compared to outside the cell) s- Cl efflux across the apical membrane can be sustaine[273] It is believed that Na+ follows passively by paracellular flux through tight junctions to maintain electroneutrality between lumenal and serosal compartments. The product of the cystic fibrosis gene, CFTR,

encodes one Cl^- channel in the apical membrane, but other channels also may be important.

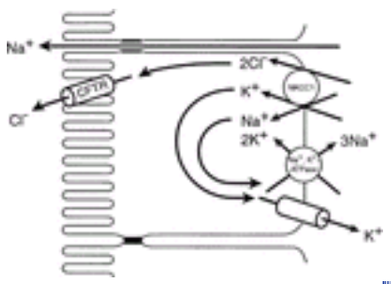

FIGURE 14-7. Cl^- secretion in the small intestine and colon. Cl^- enters cells at the basolateral membrane through $Na^+/K^+/2Cl^-$ cotransport and leaves cells at the apical membrane through Cl^- channels. Details of the model are described in the text.

HCO_3^- Secretion HCO_3^- secreted by the duodenal epithelium contributes to the mucus-HCO_3^- layer that overlies duodenal epithelial cells and thus may be an important protective factor that defends against duodenal ulcer.[270] Although the duodenum secretes large amounts of HCO_3^-, luminal HCO_3^- concentrations in the upper small intestine are relatively low because of neutralization by gastric acid. HCO_3^- secretion also takes place in ileum and colon, but its physiological role in these sites is less clear.[269] One suggestion is that apical Cl^-/HCO_3^- exchange in the colon acts to conserve Cl^- at the expense of HCO_3^- secretion. Conversely, because congenital chloride diarrhea causes systemic alkalosis, the lower gastrointestinal tract may play a significant role in acid-base homeostasis. More than one mechanism likely exists for duodenal HCO_3^- secretion. In the two models depicted in Figure 14-8, a common source of secreted HCO_3^- anion is carbonic anhydrase, which hydrates CO_2 to produce intracellular HCO_3^- and a proton. The proton is eliminated by basolateral Na^+/H^+ exchange, and Na^+ is recycled across the basolateral membrane by Na^+,K^+-ATPase. The models diverge in describing how HCO_3^- exits across the apical membrane. In an electroneutral mechanism, HCO_3^- is exchanged for intraluminal Cl^-.[271] An electrogenic mechanism has also been proposed, involving HCO_3^- secretion through apical channels.[274] It has been shown that the CFTR Cl^- channel can conduct HCO_3^-, making it a logical candidate for such a HCO_3^- secretory channel.[198] Convincing evidence from both patients with cystic fibrosis and CFTR knock-out mice suggests that CFTR plays an essential role in HCO_3^- secretion, both by conducting HCO_3^- and by regulating Cl^-/HCO_3^- exchange.[275], [276] and [277]

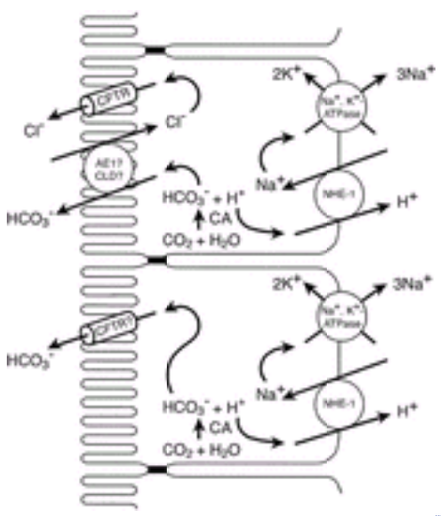

FIGURE 14-8. HCO_3^- secretion in the duodenum. The two models depicted differ in the mechanism for HCO_3^- exit at the apical membrane. As described in the text, both mechanisms may be important in mammalian duodenum. In both models, H^+ is produced by carbonic anhydrase (*CA*), which requires compensatory efflux of a proton by Na^+/H^+ exchange.

HCO_3^- secretion in colon is stimulated by SCFA,[226], [278] and it may occur by a mechanism that differs from those previously described. SCFA uptake is known to cause alkalinization of the colonic lumen, most likely because of nonionic uptake of protonated SCFA by tissues, which removes acid equivalents from the lumen.[242], [244] The resulting transepithelial pH gradient causes a vectorial titration of other weak acids (e.g., CO_2) across the epithelium and results in lumenal accumulation of the basic form (e.g., HCO_3^-). In support of this model, transepithelial CO_2/HCO_3^- gradients have been shown to drive lumenal pH changes consistent with transepithelial nonionic diffusion of CO_2.[244] The relative importance of this nonionic diffusion model versus one using apical Cl^-/HCO_3^- exchange has not been assessed.

K^+ Secretion All portions of the mammalian colon can perform active K^+ secretion.[4] K^+ secretion is enhanced by a low-sodium diet, which leads to increased plasma levels of aldosterone, and by acute K^+ loading.[128], [279], [280] K^+ secretion also can be stimulated by cAMP-dependent secretagogues and seems to occur concurrently with Cl^- secretion.[235] However, despite this evidence for acute regulatory mechanisms, the physiological role of active K^+ secretion remains unknown. The model of active K^+ secretion (Fig. 14-9) closely parallels that of Cl^- secretion. Because inhibitors of the $Na^+/K^+/2Cl^-$ cotransporter block K^+ secretion, at least in some tissues (e.g., rat and guinea pig colon), the $Na^+/K^+/2Cl^-$ cotransporter may be an important route for basolateral K^+ uptake.[129] K^+ secretion ultimately is dependent on the Na^+,K^+-ATPase pump to establish driving gradients, and in some tissues (e.g., rabbit proximal colon), the ATPase may supply the secreted K^+ instead of the $Na^+/K^+/2Cl^-$ cotransporter.[281] In contrast to Cl^- secretion, active K^+ secretion is sensitive to inhibition by K^+ channel blockers applied lumenally (e.g., barium and TEA).[282] The entire process is electrogenic, producing a current opposite to that produced by Cl^- secretion or Na^+ absorption. Although K^+ secretion is believed to be a function of colonic crypts, the interrelationship with Cl^- secretory cells at the same site is largely unexplored.[235]

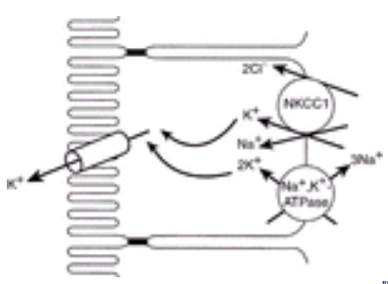

FIGURE 14-9. K^+ secretion in the colon. K^+ uptake at the basolateral membrane is mediated by either K^+,K^+-ATPase or $Na^+/K^+/2Cl^-$ cotransport, depending on the species and the intestinal segment. K^+ efflux at the apical membrane is through K^+ channels. Details of the model are described in the text.

Genetic Defects in Electrolyte Secretory Mechanisms

Defects in Cl^- secretion. Cystic fibrosis is an autosomal recessive disease that causes defective epithelial transport in numerous organs.[283] It is the most common genetic disease of ion transport, with one of every 20 whites being a heterozygote carrier and harboring a mutant *CFTR* allele. There are more than 800 naturally occurring missense, frameshift, and truncation mutations in *CFTR* that cause defective Cl^- secretion by either diminishing the amount of CFTR protein in the membrane (i.e., defective trafficking or synthesis) or decreasing appropriate Cl^- channel opening (i.e., defective ATP gating, cAMP activation, or ion conductance).[284] Mutations occur in all portions of the CFTR molecule, but with the highest frequency occurs in the first nucleotide-binding domain, which controls channel gating. The most common mutation, found in 70% of all mutant *CFTR* alleles, is a triplet deletion that removes a phenylalanine (?F508) and results primarily in defective trafficking of CFTR to the plasma membrane. Intestinal obstruction and meconium ileus associated with cystic fibrosis in newborns appear to result from the inability to maintain appropriate viscosity of lumenal contents. Intestine from patients with cystic fibrosis fails to exhibit normal secretory responses.[205], [285] It has been suggested that one reason for the prevalence of the disease is a heterozygote advantage of being partially protected from the severe, life-threatening consequences of infectious secretory diarrhea, although others have disputed this hypothesis.[286], [287] Another finding in this regard is that the ability of *Salmonella* to invade intestinal epithelial cells is significantly impaired in cells carrying one mutant allele of *CFTR*, which might confer some resistance to typhoid fever.[288]

Other Disorders of Water and Electrolyte Transport

Although genetic diseases associated with specific alterations in transport proteins have provided unique insights into the underlying molecular physiology of electrolyte transport, it should be recognized that the most common diseases of fluid and electrolyte absorption are not related to transporter mutations. This section briefly describes other disorders of intestinal fluid and electrolyte transport, some of which are far more common than the diseases discussed earlier.

Infectious Diarrhea Diarrhea caused by infectious agents is sometimes only an inconvenience in the developed world, but it is a major killer in nonindustrialized nations, responsible for about 5 million deaths per year worldwide. Many infectious diarrheas result from infections with microorganisms capable of producing classical enterotoxins. Cholera is the prototype; a multimer toxin binds to receptors on the apical surface of intestinal epithelial cells and thereby simulates a sustained increase in intracellular cAMP, which, in turn, inhibits electroneutral NaCl absorption and evokes profound Cl^- secretion. Other bacterial toxins may subvert other cellular signaling mechanisms or activate subepithelial cell types to evoke secretion (see the section "Regulation of Electrolyte Transport"). Information is also emerging regarding the mechanisms by which nontoxigenic pathogens, such as invasive bacteria, evoke diarrhea. Such organisms may evoke a program of gene expression in intestinal epithelial cells that predisposes them to display increased secretory responses to endogenous agonists, as well as diminished barrier function.[289]

Microvillus Inclusion Disease This is a rare hereditary disease that commences in the first few days of life with the appearance of severe diarrhea. The histological appearance of the intestinal epithelium is abnormal, characterized by inclusions of brush border membranes as vesicular structures within the cytoplasm of villus cells and a corresponding absence of apical microvilli. It has been suggested that cytoplasmic transport of Golgi-derived vesicles destined to fuse with the apical membrane may be defective in microvillus inclusion disease.[290], [291] Endoscopic biopsies from patients with this disorder showed markedly reduced levels of several apical transport proteins, with those transporters instead residing diffusely in the apical cytoplasm. Levels of basolateral transporters, in contrast, were normal. This led to the hypothesis that the disease results from a genetic defect specifically affecting a late stage of apical membrane trafficking.[292]

Disaccharide Intolerance This term encompasses genetic variability in the ability to conduct brush border digestion of carbohydrates, such as reduced levels of lactase in adulthood, or sucrase-isomaltase deficiency. Such conditions cause osmotic diarrhea

resulting from a reduced rate of brush border hydrolysis of specific disaccharides; only monosaccharides, not disaccharides, can be absorbed by villus enterocytes. The undigested disaccharides remain in the lumen, in contrast to specific peptidase deficiencies, which do not usually result in fluid and electrolyte transport abnormalities because the intestinal epithelium can absorb both free amino acids and small oligopeptides.

Celiac Disease This genetic disorder is characterized by a sensitivity to gluten: a water-insoluble protein found in certain cereal grains, notably w[293] The disease results in diarrhea and nonspecific nutrient malabsorption, which can be rapidly reversed by elimination of offending substances from the diet. Celiac disease causes characteristic and striking morphologic changes in the small intestine, including loss of villi, damage to remaining epithelial cells, and crypt hyperplasia. The underlying cause of the disease is unknown but may include defective digestion of gluten or an inappropriate immune response to normal or improperly processed gluten molecules. The accompanying diarrhea may have both osmotic and secretory components, with the latter mediated, in part, by inflammatory mediators released by activated immune cells in the lamina propria.

Inflammatory Bowel Diseases Patients with inflammatory bowel diseases often present with diarrheal disease, although constipation also is common. It is widely believed that the disorders of fluid and electrolyte absorption in patients with inflammatory bowel disease result from elevated levels of cytokines and other mediators in the inflamed bowel, although disruption of genes involved in maintaining an epithelial barrier also may play a role. Evidence for altered barrier function as a predisposing factor in inflammatory bowel disease has been derived from studies of mice lacking the multidrug resistance protein [294] This protein, expressed in the epithelium, may be responsible for effluxing toxic substances inappropriately absorbed from the gut lumen, and it may also regulate the expression and function of other transport proteins, such as CFT[295] Likewise, disruption of electrolyte absorption, including Na^+ nutrient cotransport, electrogenic Na^+ absorption, and electroneutral NaCl absorption, have also been reported in animal models of intestinal inflammatio[296], [297] and [298]

REGULATION OF ELECTROLYTE TRANSPORT

With molecular information regarding the transport proteins that constitute specific transport mechanisms in the small and large intestines as a foundation, one can study the pathways that regulate these transport mechanisms. Ultimately, all regulatory mechanisms are targeted to the specific protein components of the transport mechanisms, by modulating their activity, abundance, or localization within the epithelial cell.

Appreciation of the more proximal portions of the regulatory cascades has increased. In general, proximal mechanisms responsible for regulating electrolyte transport can be divided into two classes. The first consists of mechanisms operative within the epithelial cells themselves. The second class of regulatory mechanisms is intercellular and includes the release of hormones and neurotransmitters from endocrine cells and enteric nerves, the influence of mediators released by cells of the intestinal immune system, and the role of mesenchymal elements such as fibroblasts.

The kinetics of regulatory processes also should be considered. Intestinal water and electrolyte transport is subject to both acute and chronic regulation. Acute regulation accommodates rapid changes in the lumenal or neurohumoral environment of the epithelium, such as that required in response to the ingestion of a meal. This acute level of regulation usually is provided for by signal transduction events that alter the chemical nature or location of transporter proteins. In general, transport rate is increased by making individual transport molecules work more rapidly or by inserting more transport molecules into the plasma membr[199] The former type of regulation usually occurs on a rapid time scale, in a matter of seconds to minutes. Membrane insertion of transport molecules also can occur on the same time scale if it is mediated by trafficking of transporter-laden vesicles to fuse with the plasma membrane; it may take several hours if translation of new proteins is required. Rapid vesicle trafficking (and interaction with cytoskeletal proteins) is now known to be important for the regulation of certain electrolyte transport proteins. Second messenger–induced phosphorylation of the transport molecule or associated regulatory molecules, with or without transporter insertion, often mediates the rapid regulatory effects[299] Conversely, more protracted changes in whole-body electrolyte status, food intake, or development are balanced by long-term effects on transport regulation that are mediated at a genomic level and are often, although not invariably, the result of alterations in the expression levels of specific transport proteins. A schematic depiction of the various levels at which transport can be regulated is pro Figure 14-10.

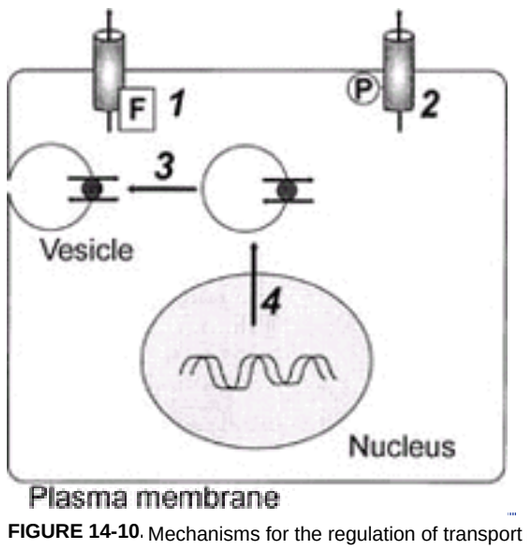

FIGURE 14-10 Mechanisms for the regulation of transport protein function at the level of the epithelium. Four levels of regulation are currently recogni*arabic numerals*). *Mechanism 1* involves direct interactions of regulatory factors*F*) with transport proteins. An example of this mechanism is the interaction of Ca^{2+} with the Ca^{2+}-activated K^+ channel. *Mechanism 2* involves covalent modification of transport proteins, most often mediated by kinase-dependent phosphorylation events. An example of this mechanism is the opening of the cystic fibrosis transmembrane conductance regulatc*CFTR*) Cl^- channel in response to phosphorylation by protein kinase *Mechanism 3* involves changes in the insertion of preformed transport proteins from vesicular pools into the plasma membrane or a change in their rate of retrieval from the plasma membrane. This mechanism appears to involve cytoskeletal cooperation. An example is the increased numbe$Na^+/K^+/2Cl^-$ (*NKCC1*) cotransporters in the basolateral membrane of secretory epithelial cells after an increase in cAM*Mechanism 4* involves changes in the synthesis of specific transport proteins at the level of gene transcription or translation. An example is the increase in epithelNa^+ channel (*ENaC*) expression in response to mineralocorticoids. For further details, see text.

Intracellular Regulatory Mechanisms

This section focuses on a stepwise analysis of the mechanisms intrinsic to the epithelium that ultimately mediate changes in transport function. The intestinal epithelium has evolved a complex network of pathways that regulate its function in a largely autonomous fashion, once an initial signal is received. In addition, signaling cascades intrinsic to the epithelium may modulate its responsiveness to subsequent stimulation.

Receptor Binding The initial step in the activation of the intracellular regulatory mechanisms that control ion transport is the binding of various hormones and neurotransmitters to membrane receptors on epithelial cells. Generally, endogenous regulatory peptides and neurotransmitters bind to and activate receptors on the basolateral aspect of epithelial c[300] whereas bacterial toxins act on the apical membran[301] There are, however, well-recognized exceptions to this polarization. For example, receptors for the endogenous agonists adenosine and bradykinin are found on both sides of the epitheli[44], [302] and this may reflect the finding that these agonists (or their precursors) are the products of inflammatory cells capable of traversing the epithelium and releasing mediators into the lumenal compartmen[303] Similarly, receptors for guanylin, the endogenous ligand for the receptor that binds the heat-stable enterotox*Escherichia co* (ST_a), are located in the apical membrane, implying that this endogenous peptide is secreted specifically into the lumen to produce its biologic effects. Conversely, epidermal growth factor (EGF), normally present in the lumen, has basolaterally localized receptors. It has been suggested that EGF functions as a “lumenal surveillance peptide” in that it regulates epithelial function only in the setting of mucosal damage, where it can gain access to the basolateral aspect of the epithe[304] Some hormones influence epithelial cell function by interacting with intracellular receptors, initiating nuclear signaling events that lead to changes in gene expression. Epithelial cell receptors thus receive signals from either sensor cells (i.e., endocrine cells, neurons, or immune cells) or microorganisms and initiate an appropriate biologic response. For microbial toxins, the secretory response that is initiated may be a primitive defense mechanism designed to wash away the offending microorgan[66] Many substances have been identified that directly affect the Cl^- secretory function of intestinal epithelial cells and, accordingly, are thought to bind to epithelial receptors. The effects of such agents, and the second messenger mechanisms that they initiate, are summariz Table 14-2. The agonists listed in Table 14-2 are limited to those thought to be of physiological relevance. It is of interest that many of the agonists listed are capable of simultaneously inhibiting electroneutral NaCl absorption (although not, importanNa^+/nutrient absorption). Conversely, other substances, such as peptide growth factors, inhibitCl^- secretion yet up-regulate NaCl absorption. This reciprocity in the regulation of Cl^- secretion versus NaCl absorption ensures that the transport functions of different epithelial cells normally do not work at cross-purposes. Further, the independence of the nutrient-coupled processes allows ongoing secretion even during the digestion of a meal and provides for the efficacy of oral rehydration.

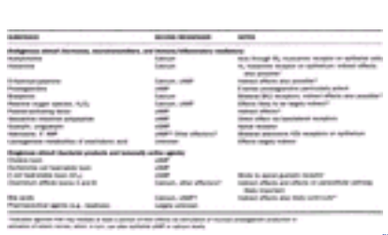

TABLE 14-2 Endogenous and Exogenous Regulators of Intestinal Chloride Secretion

Signal transduction pathways, previously thought to couple receptor occupancy to downstream effectors, can no longer be thought of as linear cascades. A more complete understanding of these pathways includes the concepts of receptor cross talk and branching of signaling mechanisms. Some of the effects of various agonists on intestinal transport may actually be the result of promiscuous coupling to alternate signal transduction pathways. Similarly, the spatial segregation of receptors and effectors in epithelial cells influences the precise mechanisms underlying transport regulation, which can result in signaling asymmetry, whereby the binding of an agonist to the basolateral membrane results in the assembly of signaling components at the apical membrane. [305] Certain protein kinases, including one isoform of the cAMP-dependent protein kinase, protein kinase A, may be selectively anchored in specific subcellular domains. The anchoring of protein kinase A is mediated by a family of proteins known as AKAPs (A-kinase anchoring proteins)[306]

Epithelial Signal Transduction Pathways Once hormones or toxins bind to epithelial cells, they initiate a cascade of events that ultimately results in the regulation of ion transport proteins. Important constituents of this cascade are the small molecules that act within the cell as second messengers. Altered levels of these second messengers control the level of activity of various protein kinases and other effectors and thereby regulate, directly or indirectly, the activity and/or abundance in the membrane of the various transport proteins that constitute the transport mechanisms. The classic second messengers recognized for their involvement in transport regulation are cAMP, cGMP, and free cytosCa^{2+}. More recent studies have identified many novel additional messengers that amplify, antagonize, or modify the effects of these classic messengers within the epithelium.

Cyclic nucleotides. cAMP and cGMP have long been recognized to play important roles in the control of intestinal epithelial ion trans[307], [308] An increase in the level of cAMP or cGMP within the epithelium stimulates Cl^- secretion and inhibits the neutral NaCl absorptive mechanism. cAMP, at least, may also play a role in stimulating the Na^+-glucose absorptive pathway[309] and duodenal HCO_3^- secretion.[274] Several key endogenous hormonal regulators of ion transport, including prostaglandins and vasoactive intestinal polypeptide (VIP), act through an increase in cAMP productio[310], [311] and [312] Exogenous substances may also activate the cAMP pathway. A well-known example is cholera toxin, which causes irreversible activation of G_s^{α} protein by

ADP-ribosylation, leading to prolonged adenylate cyclase activation and elevation of cAMP.[313] Similarly, the apical receptor for the *E coli* heat-stable enterotoxin (ST_a) is a membrane-bound guanylyl cyclase.[314, 315] The binding of toxin to this receptor thereby increases intracellular cGMP concentrations, stimulating Cl^- secretion and inhibiting NaCl absorption.[316, 317] The existence of a receptor for ST_a implied the existence of an endogenous ligand for this system.[318] Such a ligand was subsequently identified and named *guanylin*.[318] Guanylin is produced by intestinal epithelial cells and goblet cells.[319] It may play an important role in normal intestinal water and electrolyte homeostasis, although the details of this role have yet to be fully elucidated.[320] The guanylin family of peptides may also link the salt homeostatic mechanisms of the intestine and kidney.

Intracellular Ca^{2+}. The other primary second messenger system known to regulate intestinal ion transport involves increased levels of free cytosolic Ca^{2+}.[307] Increases in this messenger stimulate both Cl^- and HCO_3^- secretion while inhibiting electroneutral NaCl absorption. The starting point for this mechanism is the receptor-mediated activation of phospholipase C, which cleaves membrane phosphatidylinositols to yield a Ca^{2+}-mobilizing messenger, inositol 1,4,5-trisphosphate (IP_3), and a messenger capable of activating certain isoforms of protein kinase C, diacylglycerol.[321] IP_3 is the messenger responsible for stimulating the release of Ca^{2+} from intracellular stores.[322] Release of Ca^{2+} from stores also triggers the influx of Ca^{2+} from the extracellular milieu.[322] The actions of hormones and neurotransmitters that mediate their effects through cytoplasmic Ca^{2+} are more complicated than the direct pathways described for the cyclic nucleotides. There is a relatively poor correlation between the magnitude and duration of increases in Ca^{2+} and the resulting ion transport responses that result.[323] Whereas Ca^{2+} alone appears to be sufficient to activate epithelial Cl^- secretion, the end effects of this messenger also can be modified by other substances within the cell.[324] For example, other products of phospholipid turnover, including diacylglycerol, inositol 3,4,5,6-tetrakisphosphate (IP_4), or 3-phosphorylated inositol phospholipids produced by the enzyme PI3-K, may be involved in modulating Ca^{2+}-dependent ion transport responses.[325, 326] These auxiliary messengers have been shown to have reciprocal effects on secretory and absorptive processes, for example, inhibiting Ca^{2+}-dependent Cl^- secretion while stimulating electroneutral NaCl absorption.[325, 327]

Downstream targets of epithelial signaling pathways. The downstream effects of cyclic nucleotides and Ca^{2+} within epithelial cells are mediated largely through the actions of specific protein kinases and phosphatases. These downstream effectors alter the conformation of transport proteins or other intermediaries and thereby alter transporter activity. The mechanisms whereby these enzymes alter ion transport have been best defined for protein kinase A. This enzyme phosphorylates CFTR, resulting in channel opening.[328, 329] The efficiency of this process likely is enhanced by clustering of protein kinase A and CFTR in a signaling complex, with the participation of scaffolding proteins including AKAP.[330] Likewise, various kinases, including protein kinase A, alter the activity of Na^+/H^+ exchanger isoforms.[331, 332] For NHE3, the apparent epithelial isoform involved in small intestine NaCl absorption, the reported effect of protein kinase A is to inhibit transporter activity. This phosphorylation requires a cofactor, one of two closely related accessory proteins termed E3KARP (NHE3 kinase regulatory protein) or NHERF (NHE regulatory factor).[333, 334] Further, as discussed for CFTR, scaffolding interactions are critical in ensuring proper NHE3 regulation by protein kinase A.[335] Moreover, as alluded to earlier, an intracellular signaling pathway that enhances Cl^- secretion also causes a concomitant inhibition of NaCl absorption. Similarly, guanylin and ST_a appear to activate Cl^- secretion and to inhibit NaCl absorption through the type II cGMP-dependent protein kinase that is expressed in the intestinal epithelium.[336] The downstream basis of these cGMP-dependent effects is not entirely clear, however, because this kinase neither phosphorylates nor activates CFTR.[329] Moreover, in some segments of the intestine, the effects of cGMP may be mediated by cross-reactivity with protein kinase A.[336] The protein kinases or other effectors responsible for mediating the secretory effects of Ca^{2+} are less well understood. Increases in Ca^{2+} within the epithelial cytosol can activate the Ca^{2+}-calmodulin–dependent (CaM) kinase directly and also act in concert with diacylglycerol to translocate protein kinase C to the plasma membrane and induce its action.[321, 337] A family of Cl^- channels activated by Ca^{2+} and CaM kinase II has also been cloned.[338] It seems possible that such channels may also contribute to Cl^- secretion in response to Ca^{2+}-mobilizing agonists in the intestine, because they are expressed in native intestine and intestinal epithelial cell lines, and a related channel has been cloned from porcine intestine.[339] The relative importance of these channels, as opposed to CFTR, in setting the overall level of secretion has yet to be elucidated, although a channel with similar properties may account for residual Cl^- secretion in the intestine of some patients with cystic fibrosis or in murine models of the disease.[340, 341] Protein kinase C may mediate both decreased epithelial absorption and increased secretion, although its effect on the latter process is controversial.[342] In fact, although protein kinase C activation is capable of inducing active Cl^- secretion in some systems, a more prominent effect of this enzyme appears to be its ability to reduce secretory responses induced by both cAMP and Ca^{2+}-mobilizing agonists.[343, 344] This may result, in part, from the involvement of at least some protein kinase C isoforms in regulating vesicular trafficking of basolateral transport proteins.[345] Some effects of Ca^{2+} within the epithelium are undoubtedly mediated by a direct effect of the cation itself on basolateral, Ca^{2+}-activated K^+ channels.[346] For the Cl^- secretory process, K^+ channel opening increases the driving force for Cl^- exit across the apical membrane.[346] In fact, molecular characterization of a candidate K^+ channel involved in intestinal Cl^- secretion, hIK1, has revealed that the channel is activated by either calcium or protein kinase A in an ATP-dependent fashion, but not by CaM kinase or protein kinase C.[347] Other transport proteins also may be modified as a result of kinase cascades. For example, the basolateral NKCC1 cotransporter involved in Cl^- secretion is phosphorylated by protein kinase A and is thereby activated; this activation is balanced, in turn, by phosphatase action.[348] Furthermore, the intracellular Cl^- concentration also seems to regulate cotransporter activity by impeding its phosphorylation when Cl^- concentrations are high.[348] Thus, intracellular Cl^- self-regulates the rate of anion entry into the cell. Overall, these mechanisms balance Cl^- exit and entry in the actively secreting epithelial cell. Transport regulation also can be mediated by second messenger cascades that alter transport protein localization rather than activity (Fig. 14-10). Several transport mechanisms are regulated in an agonist-sensitive fashion by changing the rate of transport protein insertion into, or retrieval from, the plasma membrane. This is a common mechanism whereby enhanced transport can be accomplished rapidly, having been described for NKCC1,[349] NHE3,[131, 350] SGLT1,[332] Na^+ channels,[351] and Na^+,K^+-ATPase.[352] Transport protein insertion may be stimulated by known signaling pathways, such as by cAMP for NKCC cotransport[353] or PI3-K for NHE3 and Na^+,K^+-ATPase.[325, 354] Similarly, acute effects of the mineralocorticoid aldosterone may partly reflect the insertion of additional Na^+ channels and/or active Na^+,K^+-ATPase pumps into the appropriate membrane of the epithelial cell.[351] As noted previously, transporter abundance also can be reduced by retrieval from the plasma membrane, downstream of protein kinase C activation.[345] The resulting alterations in the abundance of the various transport proteins in the membrane can be considered a complementary mechanism for regulation to those previously described that alter determinants of transport protein activity (e.g., open probability, substrate affinity, or turnover number). A final mechanism whereby epithelial signaling cascades can alter transport activity is brought into play for chronic regulation. Certain hormones, particularly steroids, alter transport activity by transcriptional regulation; the entire pool of transporters within the cell is increased with an accompanying increase in transport capacity. Chronic regulation is an adaptive response to long-term changes in whole-body status, such as occur during fasting and malnutrition, or if salt intake is modified substantially.[355] This mechanism has been best established for the regulation of colonic Na^+ transport by aldosterone.[356] This hormone stimulates the expression of both epithelial Na^+ channels (ENaC) and the Na^+,K^+-ATPase, thereby increasing electrogenic Na^+ absorption.[356, 357] Similar mechanisms almost certainly alter the expression of other transport proteins in the intestine, but they remain relatively underexplored. Note, however, that not all effects of steroid or other hormones on transport regulation are mediated at the genomic level. For example, estradiol rapidly down-regulates Cl^- secretion in the female rat colon without altering Na^+ absorption.[358] One setting in which transcriptional regulation of transporters has a clearly established role, however, is during development.[359] The fluid and electrolyte transport capabilities of the gut are not fully developed at birth in most mammalian species. Development of the intestine is characterized by the appearance and modification of specific transport proteins and changes in the regulation of these pathways. The newborn intestine has elevated levels of ion absorption, in particular Na^+-dependent glucose absorption in the small intestine and amiloride-sensitive Na^+ absorption in the colon.[360] In preterm human infants, the appearance of colonic Na^+ absorption precedes that in the kidney, suggesting that the colon may be a major site for Na^+ conservation in young infants.[361] A subsequent decline in amiloride-sensitive Na^+ absorption correlates with a sharp decline in circulating levels of mineralocorticoids.[362] In contrast, the Cl^-/HCO_3^- exchanger is absent in the colon of preterm infants and only slowly develops during the first year of life.[363] Low levels of Cl^-/HCO_3^- exchange may account for the susceptibility of infants to Cl^- depletion. In the rat colon, Na^+,K^+-ATPase activity is elevated at birth and declines to adult levels during the first few days of life.[364] Conversely, the amount of Na^+,K^+-ATPase mRNA is low at birth and increases with age.[94] Cyclic nucleotide-dependent Cl^- secretion also is fully functional in newborn and infant intestine. Interestingly, the number of guanylin/ST_a receptors is high in infants and declines rapidly with increasing age.[365] Some scientists have speculated that the ability of guanylin to stimulate Cl^- secretion through CFTR may be involved in the clearance of meconium from the newborn intestine,[366] which could account for the finding of meconium ileus in babies with cystic fibrosis.[367] Conversely, bile acids, which normally elicit a Cl^- secretory response in adult colon, do not stimulate Cl^- secretion in newborns.[368, 369] There also is a developmental delay in the appearance of active ileal reabsorptive mechanisms for bile acids.[370] Thus, the concomitant absence of a Cl^- secretory response to these agonists in the colon has obvious beneficial consequences. In fact, newborn intestine may have a more general maturational delay in all receptor-mediated Ca^{2+}-dependent secretion,[369] secondary to reduced phospholipase C activity.[371]

Interactions among second messengers. Hormones that affect intestinal ion transport through different second messengers may have synergistic effects when they are supplied in combination.[372] The physiological implication of this observation is that greater effects on ion transport can be achieved for a given hormone level. The pathophysiological implication is that the combinations of mediators that are commonly encountered in the setting of intestinal inflammation can cause profound secretory diarrhea. The underlying basis of these synergistic interactions among messengers has been studied extensively. For Cl^- secretion, the synergism appears, in part, to depend on the transport proteins activated by a given messenger. Thus, Cl^- secretion induced by cAMP is regulated primarily by the opening of apical CFTR Cl^- channels, whereas Ca^{2+} appears to act predominantly by opening basolateral K^+ channels. When both second messengers are elevated simultaneously, the rate-limiting step for each type of secretion is effectively removed. This may be an oversimplification, however, given emerging evidence for Ca^{2+}-activated Cl^- channels in the intestine.[373] Moreover, synergism also may result from interactions at the level of second messenger generation. cAMP has been shown to modify Ca^{2+} mobilization responses within cultured epithelial cells.[374] Some agonists of secretory processes may also stimulate the secondary production of additional messengers that inhibit or antagonize the effects of the initial messenger.[327] Some of these agents may serve only to induce negative signals within the epithelial cell, without being agonists of secretion themselves. An example is the peptide growth factor, EGF, which inhibits epithelial Cl^- secretion evoked by Ca^{2+}-dependent agonists without acting as a secretagogue itself.[375] Conversely, EGF acts to stimulate both NaCl and glucose-coupled Na^+ absorption.[376, 377] All these processes involve common signaling pathways in which PI3-K plays a central role.[304] Other neurohumoral agents may interfere with the initial production of second messengers. Somatostatin, acting through an SS_1 receptor, inhibits agonist-stimulated accumulation of cAMP in colonic epithelial cells, presumably through the activation of a linked inhibitory G protein (G_i).[378] This effect may underlie, at least in part, the clinical efficacy of somatostatin analogs in cases of severe secretory diarrhea.[379]

Intercellular Regulatory Mechanisms

In addition to the regulatory mechanisms intrinsic to the epithelium itself, transport function is regulated by a plethora of other influences that arise as a consequence of intercellular interactions. The mediators of such effects include those released by endocrine cells, nerve endings, and resident and infiltrating immune and inflammatory cell types (see Fig. 14-3).

Endocrine and Paracrine Regulation Both endocrine (i.e., blood-borne hormones from distant sites) and paracrine (i.e., local) factors are of critical importance in the regulation of epithelial transport. Endocrine cells located within the epithelium sense the nature and composition of lumenal contents and can also mediate responses to extraintestinal signals, such as those from neural inputs. The hormones released by these cells act on epithelial receptors to alter ion transport. Table 14-2 presents a partial listing of some of the neurohumoral agents that have been identified as potential physiological regulators of intestinal ion transport and their presumed mechanism of action. The topic of gastrointestinal hormones is covered in Chapter 4.

Neural Regulation Another key regulatory mechanism for intestinal ion transport is that provided by the enteric nervous system.[380] Various ion transport responses are modified in the absence of neuronal transmission, and electrolyte transport likewise can be stimulated by directly activating resident nerves using electrical stimulation. Neurotransmitters released by enteric nerve endings can bind directly to enterocytes, as described for acetylcholine,[381] and can affect their function in much the same way that has been described for endocrine and paracrine hormones. Enteric neurotransmitters identified as being able to alter intestinal ion transport are included in Table 14-2. Neurotransmitters also may indirectly affect ion transport through their ability to release secondary hormones and mediators. In addition, the enteric nervous system responds to mediators released from endocrine or immune cells,[382] or to lumenal secretagogues, such as bacterial toxins and bile acids.[380] Neural regulation, particularly through cholinergic pathways, appears to be important in maintaining the basal ion transport tone of the intestine. The enteric nervous system probably is also critical for the integration of intestinal electrolyte transport with motility.[383] Activation of the enteric nervous system has various consequences, depending on the intestinal segment examined. In the duodenum, nerve stimulation evokes HCO_3^- secretory responses.[384] In the jejunum, ileum, and distal colon, the most prominent effect is a stimulation of electrogenic Cl^- secretion.[380] In contrast, electrical field stimulation of guinea pig proximal colon inhibits electroneutral Na^+ and Cl^- absorption.[385] Neural regulation of transport function also is likely to be important in coordinating net fluid transport with the passage of contents through the intestine. Thus, stroking of the mucosa, which may model the physical passage of a food bolus during peristalsis, evokes the secretion of both fluid and mucus.[386] This reflex may be a local one, likely involving acetylcholine and 5-hydroxytryptamine,[56, 387] that serves to lubricate the mucosa and protect it from mechanical damage. Neural regulation of epithelial function, in part mediated by central input, also has been implicated in pathophysiological settings. Rats subjected to acute systemic stress show significant increases in baseline secretion and permeability in their intestinal tissues when they are examined in vivo and in vitro.[388] Likewise, the enteric nervous system clearly plays a role in mediating at least a portion of the Cl^- secretory response to specific enteric pathogens such as cholera, *Clostridium difficile*, and rotavirus.[389, 390] and [391]

Immune Regulation Knowledge about the ways in which cells and mediators of the intestinal immune system can affect ion transport has expanded rapidly.[66] This knowledge enhances understanding of the dysfunction of intestinal ion transport that accompanies intestinal inflammatory disorders. Immune cell types also may play a broader role in the control of intestinal fluid and electrolyte homeostasis under noninflammatory conditions than previously thought. For example, mice that are genetically deficient in tissue mast cells not only display defective ion transport responses to antigen challenge, but also have reduced responsiveness to the Cl^- secretory effects of electrical field stimulation and bile acids.[392, 393] In cooperation with the enteric nervous system, mast cells also may mediate Cl^- secretory responses to certain bacterial enterotoxins, most notably the toxins produced by *C difficile*.[390, 394]

Mast cells. Mast cells are the primary effector cells involved in allergic reactions, including those to foods, and they also have been implicated in the pathogenesis of inflammatory bowel disease.[395] In a variety of animal models, immunologic (IgE-dependent) activation of resident mast cells leads to a stimulation of Cl^- secretion.[66] These responses reflect the combined effects of the various mediators that are released from mast cells on activation; the relative contribution of various substances is species dependent. Treatment of human small and large intestine tissue segments with anti-IgE has been reported to produce Cl^- secretory responses that involve histamine, 5-hydroxytryptamine, and functional enteric neurons.[396, 397] The functional association of mast cells and nerves in controlling ion transport is mirrored by a morphologic association; mast cells and nerves are in close proximity in the intestinal mucosa.[398] Mast cell mediators capable of inducing Cl^- secretion include histamine, adenosine, platelet-activating factor,

cysteinyl leukotrienes, and prostaglandin D_2.[399] Mast cells also may be important indirect regulators of ion transport by virtue of their ability to influence other inflammatory or mesenchymal cell types. The mast cell produces various cytokines and other chemotactic factors that attract inflammatory cells.[400] These latter cells, in turn, can alter ion transport. Substances capable of recruiting neutrophils and other inflammatory cells also can be released by the epithelium itself in response to challenge with inflammatory cytokines or in response to bacterial invasion.[401,402]

Neutrophils. Neutrophils accumulate at inflammatory foci in response to peptide and lipid chemotactic factors. The bacterial chemotactic peptide f-met-leu-phe has been used experimentally to activate resident neutrophils. In colonic and ileal tissues from rats, rabbits, and humans, f-met-leu-phe induces a Cl^- secretory response that is attenuated by cyclooxygenase inhibitors and is at least partially dependent on enteric ne[397,403,404] Several mediators released from activated neutrophils may contribute to secretory responses, including neutrophil-derived oxidant[405] as well as 5'AMP. This latter mediator is interesting in that it exhibits only apical activ[303] reflecting a requirement for an apically localized 5' ectonucleotidase that cleaves the 5'AMP to its final active mediator, adenosine. 5'AMP may be an important stimulus of intestinal Cl^- secretion after neutrophil migration into the intestinal lumen, as occurs in a crypt absce[406]

Other immune effector cells. Other inflammatory cells have not been examined as extensively as the mast cell or neutrophil for their ability to regulate epithelial ion transport functions. It seems likely, however, that such cells, including eosinophils, lymphocytes, and monocyte/macrophages, also participate in the control of electrolyte transport because they all can synthesize potent mediators that have either direct or indirect effects on the epi[66] Such cells also may cooperate to influence the transport and barrier properties of the epithelium. For example, activation of T cells with consequent synthesis of interferon-? in the presence of monocytes induces the latter cell type to synthesize TNF-a. This cytokine and other soluble mediators produced by the monocyte, in turn, can selectively down-regulate secretory responses of intestinal epithelial cells to a variety of agonists, as well as diminish their barrier prop[407] In fact, many studies reveal that the overall effect of ongoing inflammation is to down-regulate overall transport function of the intestine, including Na^+ or NaCl absorption[408] and to render the epithelium more permeable. These effects, in sum, predispose to diarrhea. Paneth cells form part of the innate immune system. These cells, localized at the base of intestinal crypts, synthesize a variety of antimicrobial products, including a family of peptides known *cryptdins*. Research suggests that certain of these latter molecules, which are cationic, may be capable of forming anion channels in the apical membrane of colonocytes.[409] Because these peptides are secreted into the crypt lumen by activated Paneth ce[53] their electrophysiological effects also may represent another mode of immune-epithelial interactions that modulate transport.

Role of mesenchymal cells. Immune cells also may interact with the structural elements of the mucosa to control epithelial function. Thus, the myofibroblast sheath underlying the epithelium is thought to act as a regulatory site, whereby signals from elements in the lamina propria, including immune cells, can be amplified, suppressed, translated, or spatially restrict[410] An example of such indirect regulation through myofibroblasts is demonstrated by the coculture of epithelial cells with myofibroblasts that up-regulates the secretory responses of the epithelial cells to both mast cell and neutrophil produ[410] This effect may be attributable, at least in part, to the ability of myofibroblasts to synthesize prostaglandin E_2. In turn, the capacity of the myofibroblast layer to synthesize this mediator may be selectively enhanced in the setting of local inflammation, through the induction of cyclooxygena[76]-2.

Interactions between Intercellular Regulatory Mechanisms The epithelium can respond to signals from a variety of regulatory systems, including those supplied by the endocrine, neurocrine, and immune systems, and there is substantial interplay among the mediators produced. As noted previously, mediators that exert their effects on ion transport through different intracellular signaling mechanisms can display synergistic interactions when supplied to the epithelium in combination. Further, because neural input provides a basal tone to the epithelial cells, they can respond more readily to increased levels of other hormones or inflammatory mediators. Evidence of the importance of this neurally defined tone is provided by the observation that the secretory responses of intestinal tissues to a wide variety of substances are reduced by neurotransmission blocking agents such as tetrodotoxin or atro[411] Thus, even for hormones and immune mediators known to affect the epithelium directly, the degree of neural input appears to be of paramount importance in setting the cellular sensitivity of the syste[412] The topic of intestinal neuroimmunophysiologc[66] has aroused interest because of the intimate spatial relations between the enteric nervous system and the immune elements of the intestine. These spatial relationships appear to have functional correlates. Ion transport responses stimulated by electrical field stimulation can be reduced by antagonists of inflammatory mediators; conversely, the stimulation of immune cells such as mast cells has measurable effects on enteric nerves and consequent indirect effects on ion transpc[66] Another indirect means whereby immune and inflammatory mediators have been shown to influence ion transport function is by the chronic regulation of epithelial responsiveness. Tissues from sensitized animals are more responsive to the Cl^- secretory effects of histamine and substance P[413] Long-term exposure to mast cell products similarly has been shown to enhance the responsiveness of cultured colonic epithelial cells to the Cl^- secretagogues carbachol, VIP, and ST_a without altering their basal electrical properties or their responsiveness to prostaglandins or adenos[414] Although the underlying mechanisms of these effects are not known, their existence suggests that the epithelium may be primed to display secretory responses in the setting of intestinal inflammation or infection. Similarly, sensitization primes the epithelium for rapid antigen transfer from the lumen by the expression of the low-affinity IgE receptor CD23, [415] further amplifying abnormal transport responses in the setting of gastrointestinal allergi[415] Certain other factors indirectly influence fluid and electrolyte transport by intestinal epithelial cells. These include acid-base homeosi[416] gastric and intestinal motility,[383] lumenal flow rates,[417] intestinal permeability[416] oncotic pressure of the blood, arterial pressure, venous pressure, plasma volume, lumenal pressi[418] and physical and psychological stress[419] Some of these factors may have important clinical implications. Retardation of the flow of intestinal contents achieved by the coordinated contraction of gastrointestinal smooth muscle allows more contact time for absorption and thereby may decrease stool[379] Antidiarrheal drugs, particularly the synthetic opiates, act mainly through their effects on gut motilit[379] although neuronal, hormonal, and immunologic influences also are involved. Intestinal permeability increases in diseases that cause mucosal injury, such as inflammatory bowel disease and celiac disease.[420] In theory, an increase in intestinal permeability may allow oncotic and hydrostatic pressure gradients to exert greater effects on electrolyte transport. Increased permeability also may allow macromolecules, normally excluded by the epithelium, to diffuse across the intestinal mucosa more readily. This nonspecific increase in macromolecular permeability should be contrasted with the specific CD23-mediated increase in transcytotic antigen transfer alluded to in the previous pa[415] Nevertheless, both mechanisms may amplify immune responses to transferred macromolecules and may thereby further enhance immune regulation of epithelial fun[66] on.

CONCLUSIONS

The ability of the intestine to control the fluidity of intestinal contents is clearly of key importance for many digestive functions. Both absorption and secretion of electrolytes, as well as other solutes, regulate this fluidity. Over the last few years, an explosion of molecular information regarding the precise structure and function of the transport proteins involved in these transport mechanisms has provided unprecedented insights into the basis of transport and its regulation. Moreover, the identification of various genetic disorders in which epithelial transport function is compromised has provided a deeper understanding of molecular physiology and pathophysiology. The classic disease of intestinal transport is secretory diarrhea; however, it also is apparent that many other disease states can result when transport function is either compromised or overexpressed. In the long-term, the molecular insights described in this chapter will form the basis for advances in the treatment of patients with such conditions.

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CHAPTER 15

Chung Owyang and John A. Williams

PANCREATIC SECRETION

The pancreas is an organ with exocrine and endocrine functions. The secretions of the exocrine pancreas, including digestive enzymes and bicarbonate, affect the digestion and absorption of nutrients. The endocrine pancreas releases hormones that regulate metabolism and the disposition of the breakdown products of food within the body. The combined exocrine and endocrine functions make the pancreas one of the most important and complex organs involved in the assimilation of food.

In humans and other mammals, the exocrine pancreas consists of clusters of acini that form lobules separated by loose connective tissue. Eighty percent or more of the pancreas consists of acini. Each acinus is a sphere composed of 20 to 50 pyramidal cells arranged with their broad bases around the circumference and their apices pointed toward a central lumen. Each acinus is drained by a ductule; the most proximal cells of the ductules, which extend into the lumen of the acinus, are called *centroacinar cells*. The ductules drain through a series of ducts of increasing caliber until the main ducts are reached.

Distributed within the pancreas are the islets of Langerhans, containing the cells of the endocrine pancreas. Morphologic studies have revealed cell-to-cell contact between the exocrine and endocrine tissue and direct connections between the capillaries of the islets and the acini. [1], [2] These morphologic arrangements may reflect the regulatory influences of the islet hormones on the function of the exocrine pancreas and vice versa. Of the pancreatic hormones, glucagon, somatostatin, and pancreatic polypeptide (PP) inhibit pancreatic exocrine secretion. [3], [4] and [5] Insulin potentiates the stimulatory effect of cholecystokinin (CCK) on pancreatic exocrine secretion. [6] In addition, exocrine pancreatic secretion can influence pancreatic hormone release.

Over the last decade, basic science research has significantly advanced the understanding of the control of pancreatic secretion. Several new observations have caused the paradigm shift from hormonal-neural control of pancreatic secretion to neurohormonal control with significant control system plasticity. The most significant findings include the following: (1) the cloning of the CCK receptor with the subsequent recognition that the CCK-A receptor is virtually absent from the human pancreas [7], [8]; (2) the discovery that CCK acts on CCK-A receptors on vagal afferent fibers to mediate pancreatic secretion [9], [10]; (3) the observations that most inhibitory mediators of pancreatic exocrine secretion target the dorsal vagal complex, [11], [12] and [13] suggesting a central site of coordination of pancreatic secretion; and (4) the discovery of several protease-sensitive CCK-releasing factors [14], [15] in the proximal intestine that participate in the feedback regulation of pancreatic enzyme secretion. This chapter highlights the details of these discoveries.

FORMATION AND COMPOSITION OF PANCREATIC JUICE

The human pancreas daily secretes about 1 L of juice, consisting mostly of water, electrolytes, and digestive enzymes. The morphologic appearance of the different cells of the exocrine pancreas and the results of micropuncture studies suggest that the acinar cells secrete digestive enzymes and that the ductal cells are mainly responsible for an electrolyte secretion that is rich in bicarbonate. [16] This thesis is further supported by animal experiments in which administration of a toxin or a special diet selectively destroyed acinar or duct cells. Administration of alloxan, which destroys ductal but not acinar cells, results in diminished secretion of fluid and bicarbonate. [17] Ethionine administration damages acinar cells with a concomitant reduction in enzyme secretion, but the secretion of fluid and electrolytes is relatively unaffected. [17]

Water and Electrolytes

Pancreatic electrolytes are secreted in a clear, alkaline fluid that is isosmotic with extracellular fluid. Evidence suggests that water enters the juice passively along osmotic gradients established by the active secretion of electrolytes or other solutes. The major cations in the pancreatic juice are Na^{+} and K^{+}; both are secreted at concentrations similar to their plasma concentrations. The concentrations of both cations are constant and independent of secretory rates (Fig. 15-1). The major anions in the pancreatic juice are HCO_3^{-} and Cl^{-}, the concentrations of which depend on flow rates. As the flow rate increases, the HCO_3^{-} concentration rises asymptotically, approaching a plateau value in humans of about 150 mEq/L at about 30% to 50% of the maximal secretory rate. Because the Cl^{-} concentration falls reciprocally with the increasing secretory rate, the sum of the two anions remains constant and approximately equal to the sum of Na^{+} and K^{+} at all secretory rates. In humans and other animal species, the pancreatic juices also contain Ca^{2+} (1–2 mEq/L) and traces of Mg^{2+}, Zn^{2+}, HPO_4^{2-}, and SO_4^{2-}.

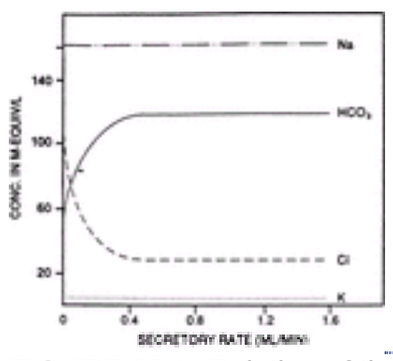

FIGURE 15-1. Relation of the secretory rate to the electrolyte composition of pancreatic juice.

The application of micropuncture and electrophysiological measurements and the use of fluorescent probes and ultrastructural analysis in the study of pancreatic duct cell function have provided important information on the mechanism of ductal electrolyte secretion. Several comprehensive reviews on this topic have been published. [18], [19] Micropuncture studies in rats, cats, and rabbits show that secreted HCO_3^- originates from the intralobular ducts and small interlobular ducts. In the large ducts, some of the HCO_3^- may be exchanged with Cl^- (Fig. 15-2). Evidence supporting a ductal origin of HCO_3^- in humans comes from studies demonstrating that the biochemical lesion of cystic fibrosis is a defect in cyclic AMP (cAMP)–regulated membrane Cl^- conductance in epithelial cells. [20], [21] In the pancreas, this is associated with defective secretion of HCO_3^-. [22] Immunofluorescence studies show that the cystic fibrosis transmembrane conductance regulator (CFTR) is localized to the apical domain of centroacinar cells and intralobular duct cells. [23] CFTR is identical to the small conductance (4 pS) Cl^- channels in the apical plasma membrane of rat pancreatic duct cells, which are regulated by cAMP. [24]

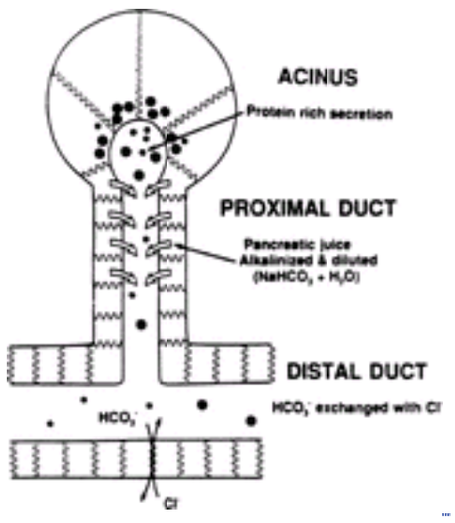

FIGURE 15-2. Ductal origin of secretin-dependent pancreatic HCO_3^- secretion. *Open arrows* indicate secretion of $NaHCO_3$ and water from the proximal duct cells. Some of the secreted HCO_3^- is exchanged with Cl^- in the distal ducts (*curved, solid arrows*). (From ref. [18].)

Electrophysiological studies have shown that by increasing Cl^- exit into the lumen, the CFTR Cl^- channels facilitate the function of a Cl^-/HCO_3^- exchange protein present in the apical membrane. The net result is a recycling of Cl^- and secretion of HCO_3^-. [19], [25] The CFTR channel has some permeability to HCO_3^-, but it is not believed to be a major pathway for HCO_3^- exit into the lumen. An additional function of CFTR is to depolarize the duct cell, with depolarization resulting in basolateral membrane transport (see later discussion).

The mechanism at the basolateral membrane of the duct cell is less well understood. Originally, HCO_3^- was thought to be derived in duct cells from CO_2 under the influence of carbonic anhydrase, which is present in the duct epithelium. [26] Although H^+ extrusion often is shown to be mediated by a Na^+/H^+ exchanger in the basolateral membrane, this protein is now known to play a role in basal cell pH regulation and to account for no more than 1% to 2% of stimulated H^+ extrusion (i.e., equivalent to HCO_3^- uptake). [27] Moreover, both experimental evidence and mathematical models have indicated that some form of energy input is necessary to obtain secretion of 150 mM HCO_3^-. The rat, unfortunately, is a poor model because it secretes HCO_3^- at only 70 to 90 mM. Two competing theories have emerged to account for higher HCO_3^- secretion rates in other species. Based on study primarily of pig pancreas, Veel and colleagues [27] have presented evidence for a primary active proton efflux mediated by a vacuolar H^+-ATPase. The ATPase at rest is located in a tubulovesicular compartment and inserted into the basolateral plasma membrane by exocytosis in response to stimulation by secretion. [28] The action of the ATPase to extrude H^+ allows HCO_3^- to accumulate in the cell and to exit by the apical mechanism previously described. Colchicine, a microtubule-depolymerizing drug, was shown to block secretion-stimulated insertion of tubulovesicles into the basolateral plasma membranes and to inhibit secretin-invoked HCO_3^- secretion by 60%. [29]

The other mechanism has emerged from work on isolated pancreatic ducts from guinea pig, which are capable of secreting 150 mM HCO_3^-. Secretion was blocked by omitting Na^+ from the bath, but not by inhibiting Na^+/H^+ exchange or vacuolar H^+-ATPase activity. [19] Based on an analysis of intracellular pH and Na^+ influx, Ishiguro and colleagues [30], [31] concluded that Na^+-HCO_3^- cotransport accounted for 75% of secretion-stimulated bicarbonate secretion. In their model, the energy of the Na^+ gradient maintained by Na^+, K^+-ATPase present on the basolateral plasma membrane [32] serves to drive HCO_3^- into the cell. This model also agrees with earlier data from the study of perfused cat pancreas showing that fluid secretion increases as a function of perfusate HCO_3^-. [26]

Some details of the nature of the protein mediating Na^+-HCO_3^- transport have emerged. A Na^+-HCO_3^- cotransporter (NBC) was originally cloned from amphibian kidney, and subsequently, a family of at least four NBCs has been molecularly characterized. The NBC1 form has alternatively spliced variants, termed *p* and *k* for *pancreatic* and *kidney*, that differ at the amino terminal as a result of alternate mRNA splicing. [33] pNBC1 has been localized to the basolateral membrane of pancreatic duct cells by immunohistochemistry. [34] The human pNBC1 analog codes for a 1079–amino acid protein, and when expressed in *Xenopus* oocytes, it induces $NaHCO_3$ uptake. [33] Although the stoichiometry of Na^+ and HCO_3^- varies in different cells, it appears to be 1 Na^+ to 2 HCO_3^- in pancreatic duct cells. [35] Because the transporter is electrogenic, it is influenced by the Na^+ gradient, the HCO_3^- gradient, and the membrane potential. In healthy duct cells, transporter-mediated HCO_3^- uptake is enhanced by depolarization induced by Cl^- exit through CFTR. [36] Thus, in this model, secretin, by activating CFTR, increases HCO_3^- uptake via the pNBC exchanger. The transcellular secretion of anions sets up a lumenal electronegative potential, enabling cations such as Na^+ and K^+ to reach the pancreatic juice by moving passively through the paracellular pathway driven by the electrical gradient that increases in response to a stimulation. [37] A cellular model depicting these events in a pancreatic duct cell is shown in Figure 15-3.

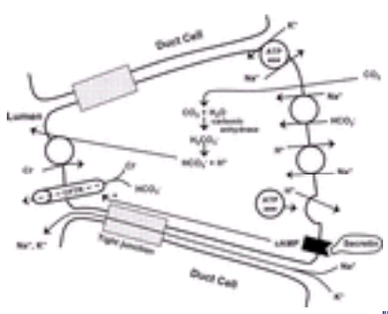

FIGURE 15-3. Cellular model of pancreatic ductal ion transport events that bring about the primary secretion of pancreatic juice. HCO_3^- enters the cell by means of the Na^+-HCO_3^+ cotransporter on the basolateral membrane or by diffusion of CO_2 that becomes hydrated to carbonic acid under the influence of carbonic anhydrase. Transport of H^+ out of the cell by the Na^+/H^+ exchanger or the vascular H^+-ATPase contributes to the provision of intracellular HCO_3^-. Other important

components at the basolateral membrane are the secretin receptor, which activates the production of cAMP, a Na^+, K^+-ATPase that maintains the low level of intracellular Na^+, and a K^+ channel (not shown) that allows K^+ to exit from the cell. The apical membrane contains the cystic fibrosis transmembrane conductance regulator (CFTR) Cl^- selective anion channel and a Cl^-/HCO_3^- exchanger, which together bring about secretion of HCO_3^- and a net lumenal negative potential. The transcellular electrical gradient between blood and lumenal fluid drives the secretion of Na^+ and K^+. The activation of the CFTR Cl^- channel also depolarizes the ductal cell and thereby links events at the apical membrane to the activation of Na^+-HCO_3^- cotransport at the basolateral membrane.

Enzymes

Depending on the species, the enzyme component of pancreatic juice is mixed in various proportions with the aqueous component. Human pancreatic juice contains a concentration of 0.7% to 10% protein. Most proteins are enzymes and proenzymes; the remainder are plasma proteins, trypsin inhibitors, and mucoproteins. The four major enzyme groups are amylolytic, lipolytic, proteolytic, and nucleolytic. The proteolytic enzymes, which include trypsinogen, chymotrypsinogen, procarboxypeptidase, and proelastase, account for most enzymes in the juice and are secreted as inactive proenzymes. After entering the intestinal lumen, trypsinogen is converted by enterokinase, an enzyme secreted by the duodenal mucosa, to the biologically active trypsin. Trypsin autocatalytically activates trypsinogen and converts chymotrypsinogen and other proteolytic enzymes into their active forms.

Pancreatic juice also contains a low concentration of trypsin inhibitor, a polypeptide that, at pH of 3 to 7, combines with and inactivates trypsin in a 1:1 ratio. Trypsin inhibitor also partially inhibits chymotrypsin. Trypsin inhibitor in the pancreas is thought to protect the organ against autodigestion by small amounts of active trypsin within the pancreas. Because it is present in minute quantities, the proteolytic activity of fully activated pancreatic juice in the intestinal lumen is not inhibited. Unlike the proteolytic enzymes, amylase, lipase, and ribonuclease are secreted by the acinar cells in their active forms. Pancreatic juice also contains a 10-kd peptide, called *colipase*, that is essential for optimal lipolysis. [38] It facilitates lipase action by binding with bile salt–lipid surfaces to increase the interaction of lipase with triglyceride. [39] In the presence of bile salts, colipase lowers the optimal pH of lipase from 8.5 to 6.5, the usual pH in the proximal intestine.

Pancreatic enzymes are synthesized within acinar cells and packaged into zymogen granules. [40], [41] The entire process, from synthesis to the point at which the enzymes are ready to be secreted into the lumen, requires about 50 minutes. Total enzyme synthesis is estimated at 20 mg/g of dry tissue per hour or 10 million enzyme molecules per acinar cell per minute. [16] Studies in rats demonstrated that CCK plays an important role in the regulation of gene expression of pancreatic enzymes. [42] Intraduodenal infusion of soybean trypsin inhibitor raises plasma CCK and increases trypsinogen I and chymotrypsinogen ß mRNA levels fivefold after 48 hours. In contrast, soybean trypsin inhibitor infusion has no effect on amylase mRNA levels. Similar effects on pancreatic enzyme mRNA levels are observed after intravenous infusion of CCK to plasma levels comparable to those obtained with soybean trypsin inhibitor. In addition to their effects on gene regulation, CCK and other hormones may exert posttranscriptional control to regulate the synthesis of specific digestive enzymes as mRNA is translated by ribosomes. [43]

Research has begun to reveal the mechanisms by which hormones, especially CCK and insulin, regulate protein synthesis at a translational level and account for the stimulation of pancreatic digestive enzyme synthesis with each meal. CCK and insulin can stimulate acinar protein synthesis in vitro without changes in mRNA. [44] These actions appear primarily at the initiation step in translation and involve the rate-limiting initiation factor eIF4E. Acinar secretagogues and insulin, through a series of steps most likely involving phosphatidylinositol 3-kinase (PI_3-K) and the mammalian target of rapamycin (mTOR), lead to the phosphorylation of the binding protein for eIF4E, and thereby the release of eIF4E, which becomes incorporated into a complex that binds the mRNA 5' cap. [45], [46] Through a separate pathway, CCK also activates the phosphorylation of eIF4E, which increases its affinity for the mRNA cap. In addition, mTOR activates ribosomal S6 kinase (p70 S6K), thereby phosphorylating S6, which increases the translation of messages with terminal polypyrimidine tracts. [47] The importance of the mTOR pathway is shown by the fact that rapamycin can block the stimulation of acinar protein synthesis in vitro. [45]

According to the classical model of Palade, amino acids are actively transported into the acinar cells, and protein synthesis occurs in the ribosomes. The newly synthesized proteins eventually find their way to the rough endoplasmic reticulum. [48] Translation of RNAs for all classes of proteins begins on polysomes, which are free in the cytosol. Pancreatic enzymes and a variety of other exportable proteins are synthesized with an amino acid terminal peptide extension called the *signal peptide*, which recognizes the endoplasmic membrane and allows the attachment of the polysome to the membrane. [48], [49] Translation is temporarily halted when the signal peptide emerges from the ribosomal subunit and interacts with the signal recognition particle, which is associated with the large ribosomal subunit. The protein-RNA complexes find their way to the endoplasmic reticulum membrane, where they interact with a membrane protein known as the *docking protein*. [48] Interaction between the signal recognition particle and the docking protein permits the completion of translocation, after which the ribosomal subunits, signal recognition particle, and RNA dissociate from the endoplasmic reticulum while the protein crosses the endoplasmic reticulum membrane into the cisternae.

The need for the signal recognition particle and docking protein complex to form on the endoplasmic reticulum before the translation of RNAs containing a signal peptide codon can be completed has important physiological implications. This process ensures that potentially noxious proteins, such as proteinases, cannot gain access to the cytosolic compartment. It also provides an initial mechanism for sorting proteins not destined for export from those that must be processed by the endoplasmic reticulum–Golgi pathway and packaged for later secretion. [50] Inside the cisternae of the endoplasmic reticulum, pancreatic secretory proteins undergo conformational changes, assuming tertiary and, in some cases, quaternary structure. These structural changes may account for the irreversible segregation of proteins within the rough endoplasmic reticulum.

The transfer of pancreatic enzyme proteins from the endoplasmic reticulum to the Golgi complex occurs within 20 to 30 minutes of synthesis. [51] Their transfer is mediated by vesicles arising from pinched-off transitional elements of the rough endoplasmic reticulum, which act as transport containers for the secretory proteins. [51] Further modification and concentration occur in the Golgi complex and may result partially from the interaction of the predominantly basic secretory proteins with polyanionic substances formed in the Golgi complex. [52] It is also facilitated by the relatively acidic pH within the Golgi complex. After their formation in the Golgi complex, secretory granules move to the apical portions of the acinar cell by a mechanism involving microtubules, where they remain until an appropriate neurohormonal stimulus triggers exocytosis.

Pancreatic enzymes from a single cell probably are secreted in a fixed ratio that is independent of the nature of the stimulus and of the rate of secretion but determined at the time of synthesis. This phenomenon may be explained by the model proposed by Scheele and Palade, [53] which states that secretory proteins are mixed together in the zymogen granule and discharged in parallel. Under certain experimental conditions, there may be nonparallel secretion of pancreatic enzymes. In humans, increasing doses of CCK infusion result in pancreatic secretion characterized by a greater response of lipase than chymotrypsin concentrations, both of which are greater than the amylase concentration. [54] Similar nonparallel secretion has been reported by Dagorn and colleagues in rats and humans. [55], [56]

Several possible explanations for nonparallel secretion have been proposed: different rates of enzyme synthesis in response to different degrees of stimulation; the existence of a soluble cytoplasmic pool of enzyme proteins that are in equilibrium with those contained in zymogen granules; and different enzymic contents within populations of acinar cells. Morphologic studies show considerable differences in cell sizes in periinsular or teleinsular acinar cells. [57] This, coupled with the observation that the enzyme content and the ratio of amylase to chymotrypsin varies widely among granules taken from the same animal, supports the latter hypothesis and suggests that nonparallel secretion is the result of exocytosis from heterogeneous cells within the pancreas. [58], [59]

Although much controversy exists regarding short-term deviations from parallel secretion, there is little doubt that long-term adaptation of enzymes to diet occurs in animals. Adaptation occurs in rats fed diets containing a preponderance of carbohydrate, protein, or fat, as indicated by increased pancreatic content, mRNA, and rates of synthesis and secretion of the appropriate class of hydrolytic enzymes by the pancreas. [60] Moreover, the dietary effects are thought to be mediated by specific hormones. Insulin mediates the increased amylase synthesis, and CCK released by protein increases the synthesis of proteases. Although such adaptive changes are unreported in humans, preferential secretion of lipase occurs in the chronic renal failure associated with hypercholecystokininemia, consistent with the adaptive change observed in rats after chronic CCK administration. [61]

STIMULATION OF PANCREATIC SECRETION

Mediation of postprandial pancreatic secretion has been ascribed mainly to the hormones secretin and CCK and to vagovagal reflexes that activate cholinergic postganglionic neurons in the pancreas. Considerable knowledge has been gained about these classical regulatory mechanisms, but the picture has become

increasingly complicated by evidence suggesting that other regulatory peptide hormones and neurotransmitters are also involved.

Hormonal Mechanisms

Secretin Secretin is synthesized by S-type enteroendocrine cells of the small intestine and released during a meal. Mutoh and colleagues [62] identified BETA2/neuroD, a transcription factor expressed in the intestinal mucosa cells of mice that also express secretin. This may be the first transcription factor identified that specifically activates cell type–specific expression of an intestinal hormone gene. Secretin is the most potent and efficacious stimulant of pancreatic fluid and HCO_3^- secretion in humans and all other species tested. Duodenal pH is the major regulator of secretin release. The threshold value for secretin release and stimulation of pancreatic HCO_3^- secretion is pH 4.5. [63], [64] Below this pH, pancreatic HCO_3^- output is related to the total amount of titratable acid presented to the duodenum. The increase in postprandial secretin levels in humans amounts to only a few picomolar increments because of the buffering of an appreciable amount of acid produced in the stomach by food and the neutralization of the remaining acid entering the duodenum by pancreaticobiliary secretion. [65] The pH of gastric chyme in the first portion of the duodenum is in the range of 4 to 5.0. [66] However, dilute HCl infused into duodenum at a rate of 2 to 4 mmol/hour can increase plasma secretin significantly in humans and dogs. [65], [67], [68] H^+ bound to solid food particles may be a potent stimulus of pancreatic HCO_3^- secretion. [69] The H^+ slowly diffusing from the particles stimulates pancreatic HCO_3^- secretion by triggering H^+ receptors located in the more distal small intestine. The mechanism by which HCl stimulates the release of secretin is unclear. Studies of rats indicate that H^+ releases a secretin-releasing factor into the upper intestinal lumen to stimulate the release of secretin. [70] This factor has a molecular weight of more than 5000 and is heat stable and trypsin sensitive. The secretin-releasing peptide(s) has not yet been completely characterized, but investigators have reported that purified porcine pancreatic phospholipase A_2 (PLA_2) stimulates secretin release from rat intestinal S cells. [71] Furthermore, PLA_2-like immunoreactivity has been demonstrated in the intestinal mucosa and released into the lumen during duodenal acidification. [72] Pretreatment with a specific anti-PLA_2 antiserum abolished the secretin-releasing bioactivity of the intestinal acid perfusate, suggesting that intestinal PLA_2 is a secretin-releasing peptide. Nonacid factors may play a role in the postprandial release of secretin. Among the major components of a mixed meal, fatty acids such as oleic acid and other digestive products of fat can increase plasma secretin levels and pancreatic HCO_3^- secretion. [73], [74] Bile in the upper small intestine can also stimulate secretin release. [75] However, the physiological importance of these nonacid factors in the release of secretin is questionable, because postprandial plasma secretin does not increase in subjects who are achlorhydric or in healthy subjects in whom meal-induced acid secretion is neutralized with $NaHCO_3$. The pancreas appears to be sensitive to the small amounts of secretin released into the circulation after a meal. Secretin given in a dose that mimics postprandial plasma secretin levels can stimulate pancreatic secretion of water and HCO_3^-. [76], [77] Administration of secretin antiserum to conscious dogs greatly reduces the pancreatic HCO_3^- response to a meal. [78] The site(s) by which secretin acts to stimulate pancreatic secretion is not fully understood. In vitro animal models clearly demonstrate that secretin stimulates HCO_3^- secretion in isolated ducts or duct fragments. [30], [31] With the use of a radiolabeled ligand ^{125}I-secretin and autoradiography, a secretin-binding site was demonstrated on pancreatic acini and duct cells. [79] No binding was evident on pancreatic islets or vascular structures. This supports but does not necessarily prove that secretin acts directly on the pancreas to stimulate pancreatic secretion. On the other hand, in vivo studies indicate that secretin may act on vagal afferent fibers to stimulate pancreatic secretion. [80] When secretin is given with CCK or with simultaneous vagal stimulation, the full postprandial HCO_3^- response is observed, suggesting that the synergistic effects of secretin with CCK or acetylcholine probably account for most of the postprandial HCO_3^- secretion. [77], [81], [82] In the isolated perfused rat pancreas, with the use of electrical field stimulation, it was shown that intrapancreatic cholinergic neurons potentiated the pancreatic exocrine secretion stimulated by intraarterial infusion of secretin, suggesting that cholinergic innervation and secretin may act synergistically to control pancreatic secretion. [83]

Cholecystokinin CCK is the other gut hormone that plays an important role in pancreatic secretion. It is released by hydrolytic products of digestion such as amino acids and fatty acids. In dogs, proteins do not stimulate pancreatic secretion, but crude enzyme digests of protein that contain peptides and amino acids are effective stimulants of pancreatic enzyme secretion, presumably by means of CCK release. [84], [85] Undigested fat is ineffective, but products of lipolysis such as fatty acids are the most potent stimulants of CCK release. [86] Factors that influence CCK response to fatty acids include their chain length, degree of saturation, concentration, and total load. [87] The mechanism by which nutrients stimulate the release of CCK is not clear. In species such as the rat, in which feedback inhibition of pancreatic enzyme secretion occurs, CCK release may be mediated by a trypsin-sensitive CCK-releasing peptide. [88] Peptone in the duodenum stimulates serotonin release from the intestinal enterochromaffin cells. The serotonin released into the submucosa activates the sensory substance P neurons. Signals would then be transmitted to cholinergic interneurons and to epithelial CCK-releasing peptide–containing cells via cholinergic secretomotor neurons. [88] In this manner, CCK release may be controlled by the level of active intralumenal proteases. [89], [90] and [91] Proteins, the major food stimulants of CCK secretion in the rat, may bind or inhibit intralumenal endopeptidases, which would otherwise inactivate the CCK-releasing peptide. [92] Research indicates that ethanol-stimulated CCK release is also mediated by a CCK-releasing peptide. [93] In dogs, the mechanism of CCK release may be nutrient dependent. [94] CCK release in response to sodium oleate but not tryptophan or HCl is atropine sensitive. [94] It is conceivable that the release of CCK by sodium oleate may be mediated by a CCK-releasing peptide, the release of which is mediated by cholinergic input, and tryptophan or HCl may stimulate CCK cells directly. [92] Under fasting conditions, the plasma CCK levels are low, averaging about 1 pmol/L in humans. [95], [96] and [97] After the ingestion of a protein- and fat-rich meal, the concentration increases to 6 to 8 pmol/L within 10 to 30 minutes, followed by a gradual decline to basal levels during the ensuing 3 hours. [96], [97] Several molecular forms of CCK appear to be released into the circulation postprandially, including CCK-58, CCK-33, CCK-22, CCK-12, and CCK-8. [98] Their relative contribution to the CCK activity of plasma in basal and stimulated states remains to be determined. CCK plays an important role in the stimulation of pancreatic enzyme secretion during the postprandial state. Infusion of physiological doses of CCK produces the same levels of pancreatic enzyme secretion as during the postprandial state. [99] Furthermore, administration of lorglumide or MK-329, both of which are potent CCK antagonists, produces a 50% to 60% inhibition of meal-stimulated pancreatic secretion in dogs. [100] These antagonists have similar effects on meal-stimulated pancreatic secretion in humans. [101] CCK can stimulate fluid and HCO_3^- secretion to some extent. [102] The effect on HCO_3^- secretion is weak but physiologically relevant, because CCK potentiates the action of secretin on the pancreas. [103] In intact dogs and humans, CCK-stimulated pancreatic enzyme secretion is not potentiated by secretin. [81], [99], [104] The mechanisms by which CCK acts to stimulate pancreatic enzyme secretion remain controversial. In vitro studies using dispersed pancreatic acini demonstrate that CCK-stimulated amylase release is insensitive to atropine or tetrodotoxin, indicating a direct action on pancreatic acini. [105] The effects of cholinergic agonists and CCK in isolated preparations of pancreatic acini are additive, suggesting that CCK may stimulate enzyme secretion by a cholinergically independent mechanism. [106] However, in vivo studies of humans and dogs have demonstrated that pancreatic secretion stimulated by CCK can be blocked by atropine, implying the involvement of cholinergic pathways. [107], [108] and [109] Furthermore, enzyme output in response to low doses of CCK is reduced in patients after vagotomy compared with healthy controls. [110] It appears that CCK can act through atropine-sensitive and -insensitive pathways to stimulate pancreatic exocrine secretion. Human studies have demonstrated that infusions of CCK-8 to produce plasma CCK levels similar to those observed after ingestion of a standard mixed meal stimulate pancreatic enzyme output predominantly in an atropine-sensitive manner. [107] Furthermore, studies in rats indicate that physiological doses of CCK act through stimulation of vagal afferent pathways originating from the duodenal mucosa (Fig. 15-4). [111] CCK receptors in the rat vagus nerve have been detected with the use of in vitro receptor autoradiography. [112] Like those in the pancreatic acini, vagal CCK receptors exist in both high- and low-affinity states. [9], [10], [113] Under physiological conditions, CCK appears to act through high-affinity vagal CCK-A receptors to mediate pancreatic enzyme secretion. [10] In contrast, the effect of CCK on satiety is mediated by low-affinity vagal CCK-A receptors. These findings suggest that different affinity states of the vagal CCK receptors mediate different digestive functions. Therefore, under physiological conditions, CCK stimulates postprandial pancreatic enzyme secretion through cholinergic pathways rather than through direct action on the pancreatic acinar cell. The muscarinic receptor on the pancreatic acini that mediates these responses appears to be an M_3 muscarinic receptor sensitive to 4-diphenylacetoxy- *N*-methylpiperadine-methiodide. [114]

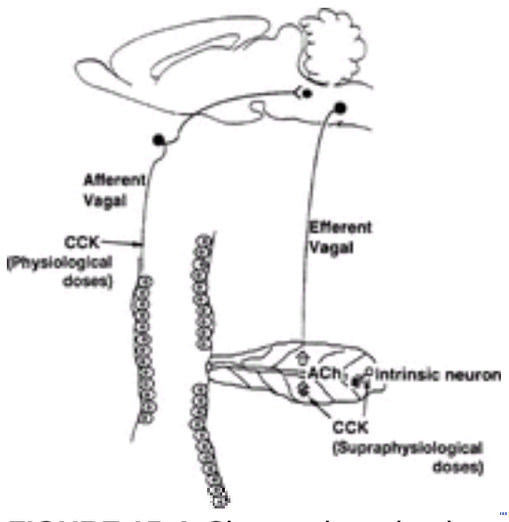

FIGURE 15-4. Sites and mechanisms of action of cholecystokinin (CCK) to stimulate pancreatic enzyme secretion. Doses of CCK-8 that produce physiological plasma CCK levels act through stimulation of the vagal afferent pathway originating from the gastroduodenal mucosa. In contrast, doses that produce supraphysiological plasma CCK levels act on intrapancreatic neurons and, to a lesser extent, on pancreatic acini. (From ref. [111].)

The molecular cloning of the CCK receptor gene and the subsequent recognition that its expression is virtually absent in the human pancreas [7], [8] further supports the possibility that CCK may be acting at an extrapancreatic site. A study indicated that human acini did not respond to CCK agonists, although they responded to a muscarinic agonist. [8] In contrast, the cells responded to CCK agonists after adenovirus-mediated gene transfer of CCK-A receptors. [8] Quantitative reverse transcriptase–polymerase chain reaction (PCR) demonstrated that the message levels of CCK-A receptors were about 30-fold lower than those of CCK-B receptors, which were about 10-fold lower than those of M_3 muscarinic receptors. In situ hybridization did not detect CCK-A receptor mRNAs in adult human pancreas. These observations indicate that human pancreatic acinar cells do not respond to CCK receptor agonists and show that this is caused by an insufficient level of receptor expression.

Serotonin Intestinal serotonin (5-hydroxytryptamine [5-HT]) appears to play an important role in the mediation of postprandial pancreatic secretion. [115], [116] 5-HT is found in abundance in intestinal enterochromaffin (EC) cells, where it is stored in subcellular granules and released by exocytosis when the EC cells are stimulated. EC cells have a morphology consistent with a “sensory” paracrine role. [117] 5-HT is released in response to a wide variety of stimuli, [118] including acidification of the duodenal lumen, [119] instillation of hypertonic glucose, sucrose, and maltose solutions, [115], [120] vagal stimulation, [121] and mechanical stimulation. [122] Studies have shown that 5-HT may increase the discharge of vagal afferent fibers from the stomach and proximal intestine in ferrets [123], [124] and stimulate vagal nodose ganglia activity in rats. This sequence in turn stimulates pancreatic secretion via the vagovagal reflex through a cholinergic pathway. [115] Studies in rats have indicated that administration of the CCK-A receptor antagonist inhibited 54% of postprandial pancreatic protein secretion. The combination of the CCK-A receptor antagonist and a 5-HT_3 antagonist almost completely abolished exocrine pancreatic secretion, [114] suggesting that non–CCK-dependent pancreatic stimulants account for about 50% of postprandial pancreatic secretion and that 5-HT plays a critical role in mediating non–CCK-dependent pancreatic secretion. Similarly, pancreatic HCO_3^- secretion and secretin release in response to duodenal acidification in rats were inhibited by the 5-HT_3 antagonist ondansetron and the 5-HT_2 antagonist ketanserin. [125] In addition, pancreatic fluid and HCO_3^- secretion stimulated by exogenous secretin was also inhibited by these 5-HT antagonists. Thus, 5-HT appears to regulate acid-stimulated exocrine pancreatic secretion through modulation of both the release and action of secretin via two 5-HT receptor subtypes. Under certain experimental conditions, activation of the 5-HT pathway may inhibit pancreatic exocrine secretion. It has been reported that in isolated guinea pig pancreatic ducts, 5-HT inhibited basal and secretin- or acetylcholine-stimulated fluid secretion. This effect was antagonized by a 5-HT_3 agonist, indicating the involvement of a direct 5-HT_3 inhibitory pathway. [126] On the other hand, administration of a 5-HT_3 antagonist, azasetron, in rats stimulated pancreatic secretion of fluid and protein. [127] This effect was abolished by atropine but not by vagotomy, suggesting the involvement of a vagus-independent cholinergic pathway. Researchers recently identified the presence of 5-HT–containing cells in guinea pig pancreatic ducts and 5-HT_3 receptors on their basolateral cell membranes. [126] The elevation of intraductal pressure reduced secretin-stimulated fluid secretion, an effect that was attenuated by a 5-HT antagonist. These authors proposed a distention-stimulated negative feedback on fluid secretion by pancreatic duct cells.

Other Hormones and Stimulatory Factors

Insulin plays a significant role in modulating exocrine pancreatic secretion. [6] Studies have demonstrated that insulin potentiates the secretory response to secretin plus CCK [128] and that ouabain, an inhibitor of Na^+, K^+-ATPase activity, abolishes this stimulatory action of insulin. Insulin potentiates the stimulatory action of secretin on Na^+, K^+-ATPase activity, without affecting the binding of secretin to pancreatic acini. Physiologically, this action of insulin is important because immunoneutralization experiments in conscious rats show that pancreatic secretion of water, HCO_3^-, and protein stimulated by a meal or by a combined intravenous administration of physiological doses of secretin and CCK-8 is markedly reduced when the circulating insulin is neutralized with a rabbit anti-insulin antibody. [129] Similar observations have been made in studies of an isolated perfused rat pancreas preparation. These studies indicate that insulin is needed locally for the action of secretin and CCK on the exocrine pancreas. A clinical confirmation of this hypothesis is that pancreatic enzyme secretion is frequently reduced in human diabetics who have no overt pancreatic disease. [130]

In view of the structural similarity between CCK and gastrin (see Chapter 4), it is not surprising to find that gastrin stimulates pancreatic enzyme secretion. In the dog, gastrin is about one-third as effective as CCK on a molar basis in stimulating the pancreas. Irrigation of a canine antral pouch with peptone and liver extracts at neutral pH stimulates pancreatic protein secretion, but irrigation with acidified solutions does not. [131] The stimulatory effects of the protein solution may be caused by the release of gastrin, and the inhibitory effects of acidification may be the result of the suppression of gastrin release. The evidence, however, is indirect; it is unlikely that the postprandial increase in plasma gastrin is sufficient to significantly stimulate pancreatic secretion.

Bombesin (i.e., gastrin-releasing peptide in mammals), a polypeptide isolated from the skin of frogs and the human alimentary tract, stimulates pancreatic secretions that contain small amounts of HCO_3^- and high concentrations of enzymes in humans. [132], [133] Bombesin can act directly on the pancreas by means of specific receptors identified on pancreatic acinar cells. [134] It has been suggested that bombesin exerts its stimulating effect on the exocrine pancreas indirectly by promoting the release of CCK from the small intestinal mucosa. [135] In other systems, bombesin has been reported to exert its effect by way of a cholinergic pathway. [136] Bombesin stimulates amylase release from rat pancreatic lobules through the release of acetylcholine from intralobular nerves. [137] In contrast, in vivo studies indicate that the action of bombesin in rats is probably direct, however, because the combined administration of atropine and the CCK antagonist proglumide does not affect pancreatic protein output stimulated by bombesin. [138] Similarly, the stimulatory action of bombesin on pancreatic secretion in humans is not affected by the CCK receptor antagonist loxiglumide. [139] The physiological importance of bombesin in pancreatic secretion is brought into question by studies demonstrating that bombesin receptor antagonists do not influence postprandial enzyme secretion in intact rats. [140]

The tridecapeptide neurotensin has been shown to stimulate pancreatic secretion in humans and dogs. [141], [142] In rats, the mechanism of stimulation appears to be neurally mediated, involving capsaicin-sensitive sensory fibers and cholinergic vagal efferent pathways. [143] Neurotensin is released by intestinal fatty acids, raising the interesting possibility that neurotensin may play a significant role in mediating pancreatic secretion stimulated by fat. [142] However, exogenous infusion of neurotensin in doses that stimulate pancreatic secretion results in plasma levels much higher than those that occur after a normal meal. [141], [142] Neurotensin stimulates HCO_3^- secretion but decreases enzyme secretion stimulated by secretin and cerulein, a CCK analog, in humans. [141] These observations do not support a role for neurotensin as a regulator of meal-stimulated pancreatic secretion.

Nitric oxide (NO), a ubiquitous substance that is present in the neurons and vascular endothelium of the pancreas, [144] also appears to play a significant role in regulating pancreatic secretion. In rats, inhibition of NO production with N^G-nitro-L-arginine-methyl ester (L-NAME) reduced basal amylase secretion by 60%. [145] The NO synthase inhibitor also inhibited pancreatic enzyme secretion in response to a meal, duodenal infusion of acid, or intravenous infusion of physiological doses of secretin or CCK in anesthetized and conscious rats. [146] In humans, graded doses of the NO synthase inhibitor N^G-monomethyl-L-arginine dose-dependently reduced pancreatic enzyme secretion stimulated by secretin and cerulein. [147] Because the NO synthase inhibitor did not inhibit amylase release or changes in intracellular Ca

$^{2+}$ concentration in rat pancreatic acini stimulated by carbachol and CCK-8, [148] it is likely that the effect of NO on exocrine pancreatic secretion is indirect. NO synthase is abundant in intrapancreatic nerves [149] and ducts, the enteric nervous system, and the vagus nerve. [150] The source of NO that mediates the action of CCK and secretin on pancreatic secretion is not yet clear.

Administration of L-NAME reduces CCK-stimulated pancreatic microvascular blood flow and at the same time decreases pancreatic fluid and protein output in cats. [151] This observation may have clinical importance, because inadequate blood flow has been associated with clinical pancreatitis. It has been reported that administration of a NO synthase inhibitor in rats with edematous pancreatitis causes a decrease in pancreatic blood flow and exacerbates cerulein-induced pancreatitis. [152] Conversely, treatment with the NO donor L-arginine before and after cerulein injection increases pancreatic blood flow and reduces the severity of cerulein/water immersion–induced hemorrhagic pancreatitis. These observations suggest that NO may have a protective role against the development of pancreatitis, possibly because it increases pancreatic blood perfusion.

Neural Mechanisms

Parasympathetic Nervous System The pancreas is innervated by parasympathetic and sympathetic nerve fibers. The parasympathetic fibers pass to the pancreas directly through the vagus nerves and indirectly through the celiac ganglion, the splanchnic nerves, and perhaps the intramural plexus of the duodenum. The functional effect of vagal stimulation of the pancreas varies greatly with the species and with the experimental conditions. [153] In the rat, both CCK and non–CCK-mediated lumenal stimuli evoke pancreatic enzyme secretion via stimulation of a vagal afferent pathway originating from the duodenal mucosa. [116], [154] Acute vagotomy or administration of atropine completely abolishes pancreatic secretion stimulated by these agents. [116], [154] In the dog and rabbit, vagal stimulation has a small stimulatory effect on enzyme output, but the increase is smaller in magnitude than with CCK stimulation. This response persists after removal of the stomach and intestine, indicating a direct stimulatory effect on the pancreas. In cats, stimulation of the vagus results in some increase in the secretion rate that does not depend on hormones. The secretion of enzymes is blocked by atropine, but atropine has no effect on HCO_3^- secretion. In pigs, vagal stimulation results in copious secretion of pancreatic juice rich in enzymes and HCO_3^-, even after extirpation of the stomach and intestine. In humans, the vagus appears to play an important role in the mediation of pancreatic secretion. Insulin-induced hypoglycemia, which is presumed to stimulate the vagus, augments secretin-stimulated pancreatic protein output. [155] Vagotomy reduces the HCO_3^- secretory response to exogenous hormones. Maximal enzyme secretion is not significantly affected, but the sensitivity of the pancreas to submaximal doses of CCK is decreased. [110] Vagotomy reduces pancreatic enzyme responses to intestinal stimulants and food. [110], [156] It seems that the cholinergic stimulation primarily modulates the action of gut peptides on pancreatic secretion but has no physiologically relevant effect on the release of CCK or secretin. [157] There are volume receptors and osmoreceptors in the human duodenum. Stimulation of these receptors by distention or administration of a hyperosmolar solution elicits a pancreatic enzyme response mediated by cholinergic neurons. [158], [159] Increased firing rates in peripheral afferent vagal neurons and in central sites have been recorded after gastric distention and intestinal perfusion with amino acids and HCl. [160], [161] and [162] The neurotransmitters involved in central vagal transmission and regulation of pancreatic secretion remain to be identified. Microinjection of the thyrotropin-releasing hormone analog in the dorsal vagal complex of rats stimulates pancreatic juice flow and enzyme output in a dose-dependent manner. [163] Vagotomy and atropine eliminate this stimulatory effect, indicating that thyrotropin-releasing hormone modulates pancreatic exocrine secretion through the dorsal vagal complex. Intrapancreatic postganglionic cholinergic neurons regulate enzyme and HCO_3^- secretion. These neurons are activated by central input during the cephalic phase and by vagovagal reflexes initiated by gastric- and intestinal-phase stimulation. Acetylcholine released by the intrapancreatic neurons may act directly on acinar cells or potentiate the action of secretin on HCO_3^- secretion from duct cells. The interaction of acetylcholine and CCK is additive. The enteropancreatic reflex also may play a role in mediating postprandial enzyme secretion. [159] This is especially important after chronic vagotomy. [164]

Sympathetic Nervous System Adrenergic innervation of the pancreas occurs mainly through the splanchnic nerves. In the pancreas, most of the fibers are distributed to the blood vessels, and a few pass to the acini or ducts. [103] Signals of the splanchnic nerves usually inhibit exocrine and endocrine pancreatic secretion; stimulation of the nerves usually decreases the response to pancreatic stimulants, but splanchnicectomy increases it. [103], [165] The pancreatic inhibitory effect of splanchnic nerve stimulation appears to be synchronous with and dependent on intense vasoconstriction that is caused by stimulation of the ß-adrenergic receptors on blood vessels. In isolated guinea pig pancreatic acini, norepinephrine alone has no effect on the response to submaximal concentrations of CCK-8. [166] Epinephrine produces a modest stimulation of enzyme output in mouse and rat pancreas in vitro, and the stimulatory effect is inhibited by ß-adrenergic receptor antagonists. [167] No clear pattern emerges from the many studies of the regulation of exocrine pancreatic secretion by the sympathetic nervous system. The major role of the adrenergic mechanism appears to be the inhibition of fluid and HCO_3^- secretion, which is mediated partially by vasoconstriction.

Enteropancreatic Neural Reflex Functional and anatomic enteropancreatic neural connections have been shown by antegrade and retrograde tracer studies; neurons in ganglia of the myenteric plexuses of the stomach and duodenum project to the pancreas. [168] Activation of the myenteric neurons in the duodenum can influence the exocrine and endocrine pancreatic function in the rat. These enteropancreatic neural pathways have cholinergic and serotoninergic components. [168], [169] The cholinergic nerves from the duodenum stimulate intrapancreatic neurons by way of nicotinic synapses. Abundant enteropancreatic serotonergic axons may inhibit pancreatic secretion through presynaptic receptors 5-HT_{1P} on cholinergic nerves. [169] Further studies are needed to define the physiological role of the serotonergic enteropancreatic neural pathways.

Peptidergic Nervous System Immunocytochemical studies have revealed several peptides in nerve cell bodies or fibers in the pancreas. Among these, nerve fibers and cell bodies containing vasoactive intestinal polypeptide (VIP) are the most abundant. [170] The VIP fibers appear to surround the cell bodies of intrapancreatic ganglia and innervate duct cells. In pigs, VIP is the neurotransmitter that mediates much of the HCO_3^- secretory response to electrical stimulation of the vagus nerve. [171] Vagal stimulation after administration of atropine increases the pancreatic venous outflow of VIP and pancreatic HCO_3^- secretion; somatostatin blocks both effects. [171] The time courses for increased venous efflux of VIP and for increased HCO_3^- secretion after vagal stimulation are similar, and a specific VIP antiserum reduces the HCO_3^- response to vagal stimulation. [170] The importance of intrapancreatic neuronal VIP as a regulator of pancreatic secretion may be species specific. VIP is a weak partial agonist in humans. [172] In some species, VIP may also induce pancreatic vasodilation and increase blood flow in response to the activation of the exocrine pancreas. The neuropeptide galanin, which has been found in intrapancreatic nerve endings surrounding the endocrine and within the exocrine pancreas, appears to have a modulatory effect on exocrine pancreatic secretion. Depending on the experimental models used, galanin may stimulate or inhibit pancreatic secretion. With the use of an isolated perfused rat pancreas, it has been shown that porcine galanin in concentrations of 1 and 10 nmol/L significantly enhances CCK-stimulated amylase secretion and stimulates insulin release. [173] However, at higher concentrations (1–100 nmol/L), galanin inhibits insulin secretion and has no effect on CCK-stimulated amylase secretion. Because insulin potentiates CCK-stimulated enzyme secretion, low concentrations of galanin probably act through the release of insulin to stimulate pancreatic secretion. In contrast, galanin appears to be a potent inhibitor of the pancreatic exocrine secretion stimulated by either bombesin, secretin, or CCK in intact animals. [174] Galanin completely inhibits 2-deoxy-D-glucose–stimulated amylase secretion in anesthetized rats and significantly inhibits veratridine-stimulated release of acetylcholine in rat pancreatic lobules. [175] Thus, in intact animals, galanin appears to inhibit pancreatic secretion by inhibiting cholinergic transmission. Other peptidergic neurotransmitters identified in the pancreas include the carboxyl-terminal tetrapeptide of gastrin or CCK, [165], [176] gastrin-releasing peptide, [177] substance P, [165], [176] peptide histidine isoleucine, [178] neurotensin, [179] neuropeptide Y, [178], [180] enkephalin, [165], [176] and calcitonin gene–related peptide (CGRP). [181] Pharmacological studies have found that CCK, gastrin, substance P, gastrin-releasing peptide, peptide histidine isoleucine, neurotensin, and CGRP stimulate and that enkephalin and neuropeptide Y inhibit exocrine pancreatic secretion. The physiological relevance of their mediation of pancreatic secretion is unknown.

Intrapancreatic Nerves

Pancreatic ganglia share many of the characteristics of the enteric neurons and receive input from the parasympathetic, sympathetic, and enteric nervous systems. The vast majority of pancreatic neurons (86%) are choline acetyltransferase (ChAT) positive. [182] All ChAT-positive pancreatic neurons possess neuropeptide Y immunoreactivity, and most also express NO synthase. [182] Most pancreatic neurons innervate the acini. Intrapancreatic neurons receive fast and slow excitatory postsynaptic potentials (EPSPs); 5-HT appears to inhibit the slow EPSPs through a 5-HT_{1P} receptor in some neurons. 5-HT and 5-HT_{1P} agonists are known to inhibit amylase secretion. Furthermore, intravenous injections of 5-HT_3 receptor antagonists, such as azasetron and granisetron, increase pancreatic protein output in conscious rats. [127] Blockade of this effect by atropine, but not truncal vagotomy, [127] is consistent with a site of action on intrapancreatic ganglia and nerves.

INTRACELLULAR CONTROL OF PANCREATIC SECRETION

Receptors

Most of the hormones and neurotransmitters that stimulate pancreatic secretion do so by directly regulating acinar and duct cells, but some may regulate indirectly by their actions on nerves or blood vessels. To determine the regulatory pathway, it is important to identify the physiological effects on isolated acinar and duct cells and to localize the high-affinity receptors for each regulator to its target cell. Because of the great preponderance of acinar cells in the pancreas, several preparations of isolated cells or pancreatic acini have been established. By using amylase secretion as the criterion for functional response, studies of the effects of agonists and antagonists on secretion have identified the presence of specific receptors on acinar cells. The receptors have been confirmed by binding studies with radiolabeled analogs and antagonists. [183], [184] Electron microscopic autoradiography and confocal fluorescence microscopy have localized binding to the basolateral membrane domain, although bound ligand may be internalized subsequently by an energy-dependent process. [185], [186] Preparations of isolated duct segments or cultured monolayers of duct cells also have been developed. [30], [187]

Through such studies, acinar cells from a variety of species, including humans, have been shown to bear receptors for CCK, bombesin, acetylcholine, VIP, and secretin. CCK interacts with both CCK-A receptors, which are highly specific for CCK and CCK-B, or gastrin receptors, which respond to both CCK and gastrin. Much is known about CCK-A and M_3 muscarinic receptors and their signaling, because of their presence on rodent acinar cells; however, the human pancreas contains few CCK-A or CCK-B receptors, and little is known about their function on acinar cells. CCK receptors on afferent nerves appear to have properties similar to those on acinar cells. Less is known about duct cell receptors because of the relatively small number of these cells in the pancreas and the greater difficulty in studying their physiological functions (e.g., ion transport). Duct cells bear receptors for secretin, ATP, CCK, and acetylcholine and may have receptors for VIP.

Receptors for major pancreatic secretagogues belong to the receptor family characterized structurally by seven hydrophobic transmembrane domains and functionally by their interaction with guanine nucleotide–binding proteins (i.e., G proteins). [188] The M_3 muscarinic receptor, the bombesin receptor, and the CCK-A and CCK-B receptors have been cloned and found to possess similar structures. The cloned secretin receptor, although possessing seven transmembrane regions, is somewhat different in its amino acid sequence. [189]

Structure-function studies with site-directed mutagenesis and chimeric receptors have established some general principles of function for the G protein–coupled receptor family. [188], [190] The transmembrane segments may form a pocket for the binding of small molecules, such as acetylcholine, and the extracellular amino-terminal end and loops may be important in the interaction with peptide molecules. The third cytoplasmic loop, projecting between the fifth and sixth transmembrane domains, is thought to interact with the appropriate G protein, and the serine and threonine residues in the cytoplasmic carboxyl-terminal tail may be involved with regulatory mechanisms such as desensitization and down-regulation by way of phosphorylation. The function of glycosylation on externally directed sites at the amino terminus is not yet well established, but these sites may play a role in ligand binding or in the intracellular processing of new receptors and their insertion into the plasma membrane. A number of techniques are now being applied to elucidate the details of ligand-receptor interaction.

Transmembrane Signaling

Although all membrane receptors are integral proteins spanning the lipid bilayer, the pancreatic secretagogue receptors convey information by interaction with G proteins. G proteins are heterotrimeric proteins with unique a subunits and a smaller number of shared ß? subunits. [191] Acinar cells possess a_s and a_i subunits, which stimulate and inhibit adenylate cyclase, respectively. These subunits can be ADP-ribosylated by cholera toxin and pertussis toxin, which permanently activate or inhibit adenylate cyclase, respectively. Acinar cells also possess a_q and a_{11} subunits that activate phospholipase C. [192] The full complement of a and ß? subunits expressed in acinar and duct cells and their functions still need to be fully explored.

The a subunit possesses the guanine nucleotide–binding site, which, in the resting state, is occupied by GDP. After the receptor binds its ligand, it interacts with the G protein to catalyze the exchange of GTP for GDP. The GTP-a subunit dissociates from the ß? complex and activates its effector (i.e., phospholipase C, adenylate cyclase). The system is amplified because the lifetime of the GTP–a subunit complex is much longer than that of the hormone-receptor complex. Eventually, GTP is cleaved to GDP by an intrinsic GTPase activity, and the a subunit reassociates with the ß? subunit. It is because of this cycle that secretagogues stimulate GTPase activity in pancreatic membranes and that nonhydrolyzable analogs of GTP, such as GTP?S, activate adenylate cyclase or phospholipase C in pancreatic membranes or permeabilized acinar cells. [193]

The final component in transmembrane signaling is the membrane effector that generates the intracellular messenger. The two major effector enzymes in acinar cell membranes are the polyphosphoinositide-specific phospholipase C, which cleaves phosphatidylinositol 4,5-bisphosphate, producing inositol 1,4,5-trisphosphate (IP_3) and 1,2-diacylglycerol (DAG), and adenylate cyclase, which converts ATP to cAMP. [194] Multiple forms of phospholipase C have been purified, cloned, and shown to be expressed in a variety of tissues. [195] The $ß_1$ and $ß_3$ forms are differentially activated in rat acini by CCK, carbachol, and bombesin. [196] Adenylate cyclase has also been cloned, and its primary structure is consistent with its role as an integral membrane protein with multiple membrane-spanning domains, although its catalytic site is clearly intracellular. [197] Multiple isoforms of adenylate cyclase exist, some of which are also regulated by G-protein ß? subunits, Ca^{2+}, and protein kinases. Adenylate cyclase is clearly the major effector enzyme in duct cells, but there is some evidence for the participation of a phospholipase C. Other membrane effectors in the pancreas may include phosphatidylcholine-specific phospholipases C [198] and D, [199] PLA_2, Na^+/H^+ ion exchanger, and various ion channels. However, these may be regulated by intracellular messengers rather than directly by G proteins.

Intracellular Messengers

The major intracellular messengers involved in the regulation of pancreatic secretion are IP_3, Ca^{2+}, DAG, and cAMP. [194], [200] The first three are predominant in the acinar cell and increase after the activation of phosphoinositide-specific phospholipase C by CCK and acetylcholine, whereas cAMP is the predominant messenger in duct cells, where it is produced in response to secretin.

Phosphatidylinositol and its polyphosphate derivatives—phosphatidylinositol 4-phosphate (PIP) and phosphatidylinositol 4,5-bisphosphate (PIP_2)—constitute about 10% of membrane phospholipids. PIP_2 serves as a precursor for IP_3. When acinar cells are stimulated with acetylcholine or CCK, there is a rapid fall in prelabeled PIP_2 and PIP, an increase in DAG, and a delayed rise in phosphatidic acid. This indicates the primary breakdown of polyphosphoinositides, with production of DAG, which is subsequently converted to phosphatidic acid by DAG kinase. When the production of the water-soluble inositol phosphatases is assessed after labeling with ^{3}H-inositol, the IP_3, IP_2, IP_1, and inositol products are all increased. Because IP_3 can be produced only by hydrolysis of PIP_2, this is presumed to be the primary event, with further synthesis of PIP and PIP_2 from phosphatidylinositol by the action of a phosphatidylinositol kinase.

Although production of IP_3 was initially measured by prelabeling with ^{3}H-inositol, it is possible to measure the actual mass of cellular IP_3 by a competitive binding assay. Such measurements show a rapid increase within 5 seconds in rat acini stimulated with CCK, carbachol, or bombesin (Fig. 15-5). This 10- to 30-fold increase rapidly declines after 30 to 60 seconds to a smaller, sustained plateau. Coincident with this rapid peak of IP_3 is a rapid increase in DAG of similar magnitude (Fig. 15-6), which is thought to arise simultaneously from the hydrolysis of PIP_2. The later, larger increase in DAG may result from the hydrolysis of phosphatidylinositol or phosphatidylcholine by distinct phospholipases. [198], [199] These results mean that IP_3 and DAG can act as separate signals and are not necessarily in lockstep. Phosphorylation of 1,4,5-IP_3 produces 1,3,4,5-IP_4, which can be dephosphorylated to yield 1,3,4-IP_3. Whether a biologic role exists for these other inositol phosphates and for additional degradation products, such as IP_2 and IP_1, has not been established.

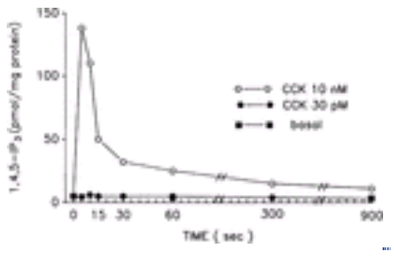

FIGURE 15-5. Time course of inositol 1,4,5-trisphosphate (*1,4,5-IP* $_3$) increase induced by cholecystokinin (*CCK*) in rat pancreatic acini. (From ref. [198].)

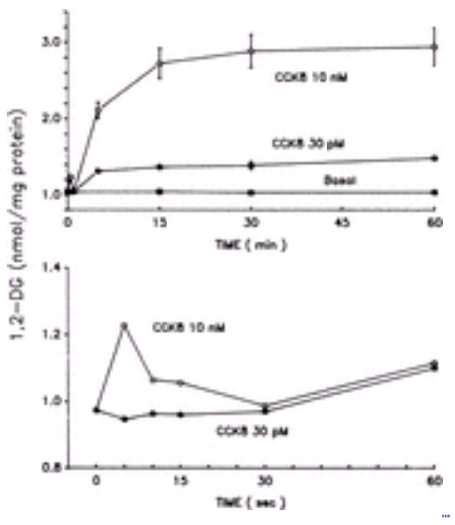

FIGURE 15-6. Time course of 1,2-diacylglycerol (*DG*) increase induced by cholecystokinin (*CCK*) in rat pancreatic acini. (From ref. [198].)

The established function of IP $_3$ is to bind to a receptor on an intracellular Ca $^{2+}$ storage site and release Ca $^{2+}$ into the cytoplasm. The IP $_3$ receptors are large, 260-kd molecules that form tetramers, which function as ligand-gated ion channels. Three forms of IP $_3$ receptors (types I, II, and III) are known, and all exist in acinar cells. [201] The sensitivity of these receptors to IP $_3$ can also be regulated by phosphorylation of the IP $_3$ receptor channel, by G-protein ß? subunits, Ca $^{2+}$, and thiol-reacting reagents. Modulation of IP $_3$ sensitivity may explain how certain agonists can release Ca $^{2+}$ without a measurable increase in IP $_3$ levels. IP $_3$ receptors undergo down-regulation in response to prolonged stimulation. Another homologous gated intracellular Ca $^{2+}$ channel, the ryanodine receptor, exists in muscle and other cells and mediates Ca $^{2+}$-induced release. The existence of ryanodine receptors in acinar cells has recently been verified.

The morphologic identity of the Ca $^{2+}$-sequestering and -releasing organelle in the pancreas is not clear. Probably multiple membrane-bound compartments all related to the endoplasmic reticulum have the ability to take up and release Ca $^{2+}$. Some investigators have suggested the existence of multiple populations of vesicles with different Ca $^{2+}$ transport or release characteristics. [202] Immunocytochemistry is being used to localize IP $_3$ receptors in intact cells. [203], [204] All three IP $_3$ types are present primarily in the apical pole of the cell just under the lumenal membrane and partially overlapping the submembranous network of actin filaments. Several types exist in the nuclear membrane but in fewer numbers.

Much of the knowledge about secretagogue-induced changes in the intracellular Ca $^{2+}$ concentration ([Ca $^{2+}$] $_i$) in acinar cells has been obtained by using fluorescent Ca $^{2+}$ probes such as fura-2. These studies have been carried out on suspensions of cells or acini and on individual cells by microspectrofluorometry [205] and digital imaging. [198], [206] Such studies have shown that high concentrations of CCK, bombesin, and cholinergic analogs cause a rapid five- to tenfold increase of [Ca $^{2+}$] $_i$, which declines over 2 to 5 minutes to a level slightly above the basal level (Fig. 15-7). This initial large increase is essentially independent of extracellular Ca $^{2+}$, but the small, sustained plateau increase in [Ca $^{2+}$] $_i$ absolutely depends on extracellular Ca $^{2+}$. The initial increase shows a similar time course and dependence on secretagogue concentrations, as does the rapid increase in IP $_3$, and is presumed to depend on IP $_3$-stimulated release of intracellular Ca $^{2+}$. Much of this released Ca $^{2+}$ is extruded from the cell, as shown by earlier studies documenting an increased efflux of prelabeled ^{45}Ca $^{2+}$ and a fall in total acinar Ca $^{2+}$. [200], [207] All three of the major phospholipase C–activating secretagogues (i.e., CCK, acetylcholine, bombesin) access the same intracellular pool of Ca $^{2+}$, and after maximal stimulation by one agonist, the addition of another has no additional effect. After removal of the agonist, the IP $_3$-releasable intracellular pool refills over 2 to 10 minutes as Ca $^{2+}$ is taken up from the medium and resequestered.

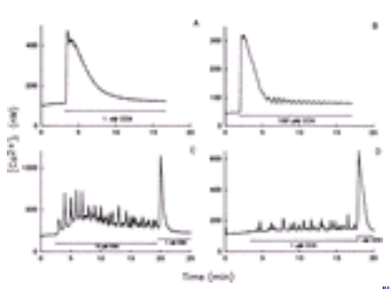

FIGURE 15-7. Increase in intracellular calcium ion concentration (*[Ca* $^{2+}$*]* $_i$) in individual rat acinar cells in response to various concentrations of cholecystokinin (*CCK*). (From ref. [205].)

The sustained increase in [Ca $^{2+}$] $_i$ and the refilling of the intracellular Ca $^{2+}$ pool involve activation of a poorly understood Ca $^{2+}$ entry mechanism. Studies measuring ^{45}Ca $^{2+}$ uptake and the influx of Mn $^{2+}$, which quenches the intracellular fluorescence of fura-2, indicate that the phospholipase C–coupled secretagogues increase Ca $^{2+}$ influx two- to fourfold. [207] The influx mechanism is blocked by La $^{3+}$ but is not voltage-dependent or sensitive to organic Ca $^{2+}$ channel blockers. Patch clamp studies have not yet clearly defined a constituent channel. It is better defined as an entry mechanism. The mechanisms controlling Ca $^{2+}$ influx are sensitive to the state of the intracellular Ca $^{2+}$ stores, as most directly shown by using inhibitors of the microsomal Ca $^{2+}$-ATPase such as thapsigargin, which releases intracellular Ca $^{2+}$ and activates the Ca $^{2+}$ influx mechanism, bypassing the receptor and the generation of inositol phosphates. Evidence also exists for tyrosine phosphorylation in gating Ca $^{2+}$ influx. That a sustained increase in [Ca $^{2+}$] $_i$ is important for sustained amylase release is demonstrated by the fact that La $^{3+}$ inhibits the sustained component of amylase release, similar to removal of Ca $^{2+}$ from the medium. [208]

Recordings of the [Ca $^{2+}$] $_i$ from individual cells of rat and mouse acini have revealed that low physiological concentration of CCK, acetylcholine, and bombesin induce a different pattern of Ca $^{2+}$ increase characterized by oscillations in [Ca $^{2+}$] $_i$ increases. [205], [206], [209] Superimposed on any steady increase are phasic increases in [Ca $^{2+}$] of up to 500 nmol/L (see Fig. 15-7). These oscillations, which occur one to four times per minute, are relatively independent of extracellular Ca $^{2+}$ and involve the release and reuptake from the intracellular Ca $^{2+}$ stores. That [Ca $^{2+}$] oscillations can drive secretion is shown by the fact that the CCK analog JMV-180 induces Ca

$^{2+}$ oscillations and can stimulate maximal amylase release. [210]

Confocal digital imaging of Ca^{2+} in acinar cells has shown that the $[Ca^{2+}]_i$ increase is initiated in the apical pole of the cell and then spreads basally. [211] The response to lower agonist concentrations is a series of local increases in Ca^{2+} in the apical pole of the cell. [212] Improved imaging techniques have shown that local Ca^{2+} spikes are the result of pacemaker hot spots of Ca^{2+} release that entrain the surrounding region. [213] Interestingly, in the same cell, different agonists (i.e., carbachol, CCK, and bombesin) initiate $[Ca^{2+}]$ increases in distinct apical areas, indicating compartmentalization of signaling. [214], [215] Further complicating the process, protein kinase A phosphorylation of IP_3 receptors can modulate the pattern of Ca^{2+} increase. [216] Besides the apical-to-basal spread of Ca^{2+} in intact acini, Ca^{2+} waves appear to spread from cell to cell around an acinus. Gap junction coupling remains open as these Ca^{2+} waves spread but closes in response to supermaximal stimulation. [217] This cell-to-cell spread [217] increases the cellular sensitivity, allowing acinar activation to be triggered by the most sensitive cell.

Intracellular Messenger–Induced Secretion The evidence to support the importance of Ca^{2+}, DAG, and cAMP as intracellular mediators regulating pancreatic secretion is based on the ability of artificial changes in the level of each messenger to influence secretion. [183], [200] In the case of Ca^{2+}, the discovery that certain antibiotics, such as A23187 and ionomycin, functioned as Ca^{2+} ionophores and could be used to increase Ca^{2+} influx and trigger secretion provided one of the cornerstones of evidence for the importance of Ca^{2+} in this process. The Ca^{2+}-ATPase inhibitor thapsigargin also increases $[Ca^{2+}]_i$ and can be used to define the contribution of Ca^{2+} in stimulating secretion. [218] The discovery that certain phorbol esters could activate protein kinase C in a manner similar to DAG led to extensive studies of the activation of this pathway. [219] Ca^{2+} ionophores and phorbol esters stimulate acinar cell secretion, and their effects are additive or potentiating. In the case of cAMP, derivatives such as dibutyryl-cAMP or Br-cAMP that are lipophilic or phosphodiesterase resistant have been used to activate the pathway normally initiated by secretin or VIP. In most species, cAMP derivatives used alone have minimal effects on acinar cell secretion but potentiate the effects of agents working by means of Ca^{2+} and DAG.

Mechanism of Action of Intracellular Messengers The intracellular messengers active in pancreatic acinar cells have been identified and characterized, but much less is known about the mechanisms by which they act to induce granule exocytosis, fluid secretion, protein synthesis, and gene expression. Although other mechanisms may exist, all the intracellular messengers activate protein kinases and phosphatases and thereby regulate the state of protein phosphorylation. Considerable data suggest that changes in the phosphorylation of regulatory proteins mediate the acini or hormones and neurotransmitters in a variety of tissues. In support of this postulate, more than 25 phosphoproteins (not all identified) that are regulated by pancreatic secretagogues have been visualized by two-dimensional gel electrophoresis in pancreatic acini. [220], [221] Some are uniquely regulated by Ca^{2+}, phorbol esters, or cAMP, and others are regulated by multiple second messengers. When a Ca^{2+} ionophore and a phorbol ester are combined, they reproduce all the phosphorylation changes induced by secretagogues. Several Ca^{2+}-activated kinases have been identified in pancreatic acinar cells, including Ca^{2+}/calmodulin-activated type II and type III kinase and myosin light chain kinase. [194], [200] Although some kinases are highly substrate specific, calmodulin-activated type II kinase is a multifunctional kinase that acts on several proteins. Its activation by CCK is supported by its temporary conversion to a Ca^{2+}-independent form. [222] Protein kinase C, originally described as a Ca^{2+}-, phospholipid-, and DAG-dependent kinase, [219] is also present in acinar cells. Multiple isoforms are present, including a, d, e, and ?, which include both classical and atypical forms. [223] In addition to Ca^{2+}- and cAMP-activated kinases, acinar cells contain the major classes of serine/threonine phosphatases (i.e., PP1, PP2A, and PP2B). [224], [225] Although some of these are constitutively active and are involved in reversing phosphorylation induced by kinases, PP2B, or calcineurin, is specifically activated by Ca^{2+} through calmodulin. This phosphatase, which is activated by pancreatic secretagogues, is blocked by the immunosuppressant cyclosporine, which also partially inhibits secretagogue-activated amylase secretion. [225] A specific substrate protein of 24 kd, CRHSP-24, also exists in acini, but its function is unclear. [226] The role of intracellular messengers and effectors in pancreatic enzyme secretion is summarized in Figure 15-8. Stimulation of secretion normally involves synergistic interactions among intracellular messengers. In the case of acetylcholine and CCK, this includes interactions between Ca^{2+}- and DAG-activated pathways. Agents such as VIP and secretin, which increase cAMP, add a further interaction at the post–intracellular messenger level. Proteins localized on the granule and lumenal plasma membrane and several soluble and cytoskeletal proteins may be involved in exocytosis. In pancreatic duct cells, the same intracellular messengers and kinases may regulate ion pumps, carriers, and channels involved in fluid and electrolyte secretion.

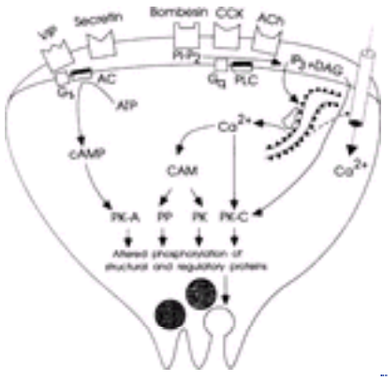

FIGURE 15-8. Stimulus-secretion coupling of pancreatic acinar cell protein secretion. In one intracellular pathway, receptors for vasoactive intestinal polypeptide (*VIP*) and secretin couple through a G protein (G_S) to activate adenylate cyclase (*AC*), and the cAMP produced activates protein kinase A (*PK-A*). In the other and quantitatively more important pathway, receptors for bombesin, cholecystokinin (*CCK*), and acetylcholine (*ACh*) couple through G proteins of the Gq family (*Gq*) to activate phospholipase C (*PLC*). This enzyme hydrolyzes phosphatidylinositol 4,5-bisphosphate (*PI-P2*) to produce inositol 1,4,5-trisphosphate (IP_3) and diacylglycerol (*DAG*). IP_3 releases Ca^{2+} from intracellular stores in the endoplasmic reticulum, activating Ca^{2+} influx across the plasma membrane. Ca^{2+}, by binding to calmodulin (*CAM*), activates several protein kinases (*PK*) and one protein phosphatase (*PP*). Diacylglycerol with Ca^{2+} activates several species of protein kinase C (*PK-C*). This battery of protein kinases and phosphatases, by altering phosphorylation of diverse proteins, brings about the secretion of digestive enzymes and other cellular effects on growth and metabolism.

Zymogen Granules and Exocytosis The terminal steps in secretion involve fusion of the zymogen granules with the lumenal membrane of the acinar cell. This fusion event shares basic mechanisms with other membrane fusion events from yeast to neurons. Two types of proteins, SNARE proteins and small G proteins of the Rab family, appear to play prominent roles. In the SNARE paradigm, transport vesicles destined to fuse with another membrane possess a set of proteins, termed *v-SNAREs*, that interact with t-SNAREs—that is, proteins on the target membrane—and with soluble attachment factors. [227] Synaptobrevins/VAMPs, which act as v-SNAREs on synaptic vesicles, are present on zymogen granules, with both VAMP2 and cellulobrevin present. [228] t-SNAREs on the plasma membrane of neurons include SNAP-25 and syntaxin. Multiple isoforms of syntaxin in acini have been identified. Syntaxin 2 is present on the apical plasma membrane, [229] but other syntaxins are also present on zymogen granules. The interaction of VAMPs and syntaxins in acini remains poorly understood. The additional importance of cytosolic proteins was shown by the observation that permeabilizing acini with a-toxin, which produces small pores that prevent protein leakage, prevented the rundown of secretion seen with streptolysin-O permeabilization that induces large pores. Moreover, cytosol from brain or lacrimal glands restores secretion in streptolysin-O–permeabilized acini. One of the responsible proteins is CRHSP-28, a Ca^{2+}-regulated phosphoprotein that associates with zymogen granules through a 70-kd protein. [230] Whether Ca^{2+} in acini directly regulates a SNARE or cytosolic protein or indirectly through activating a kinase or phosphatase is unknown. An experimental approach that should prove useful in this work is monitoring secretion by following the capacitance of patch-clamped acinar cells. Low concentrations of acetylcholine, which are known to induce $[Ca^{2+}]$ oscillations in the apical pole of the cell, also induced an increase in membrane capacitance without an increase in membrane conductance. [231] Work on Rab species in the pancreas has focused on Rab3, because Rab3A is present on synaptic vesicles and an immunoreactive Rab3 species has been identified in pancreas. Studies using PCR amplification and immunocytochemistry with specific antibody have identified conclusively the presence of Rab3D on zymogen granules [232], [233] as well as on granules in salivary and lacrimal glands, and on chief cells of the stomach. Overexpression of Rab3D in acinar cells of transgenic mice enhances a component of amylase secretion, [234] indicating that Rab3D may be rate-limiting for secretion. Another cellular component believed to be involved in exocytosis is the cytoskeleton. It may serve as a barrier, preventing premature secretion, and it may be involved in the movement of zymogen granules to the cell surface. The former role may be mediated by an actin network under the plasma membrane. Evidence of this role for actin is the observation that introducing the monomeric actin-binding protein ß-thymosin into permeabilized acini to shift the monomer-polymer equilibrium induces secretion in a Ca^{2+}-independent manner. [235] Moreover, high concentrations of CCK and ß-thymosin, the actin monomer–binding protein, decreased polymerized actin at the apical membrane. In contrast, phalloidin, which binds and stabilizes polymerized actin, inhibited secretion by the permeabilized cells.

Intracellular Pathways Leading to Growth and Gene Regulation Pancreatic secretagogues, particularly CCK, have long been known to induce pancreatic hypertrophy, hyperplasia, and gene expression. Initially, it was assumed that the same intracellular mediators that regulate secretion might mediate these effects. It is

now clear, however, that nonsecretagogues, such as insulin and epidermal growth factor, can also regulate nonsecretory pancreatic functions and that classical secretagogues, such as CCK, activate novel signaling pathways, many of which involve tyrosine phosphorylation. There are also cases in which a nonsecretagogue input, such as the extracellular ligation of integrins, leads to both tyrosyl phosphorylation and an increase in intracellular Ca^{2+}. [236] Because of the increasing evidence that cytokines can affect the pancreas, at least in pancreatitis, it seems likely that cytokine receptors also activate intracellular signaling. G protein–coupled receptors can activate tyrosine kinase signaling pathways in a variety of cells. [237] Most growth factor receptors include an intracellular tyrosine kinase domain, and autophosphorylation of the receptor on tyrosines leads to docking of signaling molecules such as phosphoinositol 3-kinase, phospholipase C-?, and adapter molecules such as Grb-2 and Shc. G protein–coupled receptors do not possess tyrosine kinase activity, nor are they tyrosyl phosphorylated. However, CCK is known to increase tyrosyl phosphorylation of multiple acinar proteins [238] and to activate the mitogen-activated protein kinase (MAPK) cascade involving Ras-Raf-MEK-ERKs. [239], [240] The kinase $p90^{rsk}$, which is downstream of ERKs and acts to phosphorylate nuclear transcription factors and cytoplasmic targets, also is activated by CCK. [241] The early events by which secretagogues may activate Ras and the MAPK cascade leading to ERKs have been studied. In rat acini, CCK, carbachol, and bombesin, as well as epidermal growth factor, increased the tyrosyl phosphorylation of Shc, [242] leading to the binding of Grb2 and SOS. Because SOS is a guanine nucleotide exchange factor for Ras, this could mediate an activation of Ras in response to CCK. Shc phosphorylation induced by CCK was shown to be largely protein kinase C–dependent, whereas that due to epidermal growth factor was fully protein kinase C–independent. [242] In pancreas-derived AR42J cells, which possess primarily CCK-B receptors, gastrin has also been shown to increase tyrosyl phosphorylation of Shc. [243] In AR42J cells, gastrin also induced the tyrosyl phosphorylation of the docking protein, insulin receptor substrate 1 (IRS-1). [244] Because Grb-2 also binds to IRS-1, this is a potential alternative means of activating the MAPK cascade. IRS-1 has not been identified in normal acinar cells, although it is overexpressed in some pancreatic cancer cells. The major unanswered question, however, is the identity of the tyrosine kinase that phosphorylates Shc and IRS-1 and how it is activated. Interest has centered on cytoplasmic tyrosine kinases of the Src family, and published data would suggest that protein kinase C activates Src. Two other MAPK cascades, called *stress-activated protein kinase (SAPK)* and *p38 MAPK,* lead to activation of Jun kinase. Both of these pathways exist in acinar cells and can be activated by CCK as well as other extracellular signals. [245], [246] Activation of SAPK is an early event in secretagogue-induced pancreatitis, although it is not clear whether it is a cause or effect. [247] A known effect of p38 MAPK is to activate MAPKAP kinase-2, which phosphorylates Hsp27 and thereby regulates actin polymerization. [246], [248] Both SAPK and p38 are activated by dual phosphorylation on tyrosine and threonine residues by upstream dual function kinases related to MEKs. [249] These are activated by a serine/threonine kinase analogous to Raf that is farther upstream. However, the three MAPK cascades can be activated independently. Evidence exists for two other signaling pathways in pancreatic acini. [194] The first is the activation of focal adhesion kinase, or $p125^{FAK}$, and its downstream target, paxillin. Both of these molecules are associated with the cytoskeleton and, in other cells, are activated by growth factors and integrins. CCK has been shown to increase the tyrosine phosphorylation of $p125^{FAK}$ and paxillin in rat acini. [250] This effect is probably mediated by the small G protein Rho. The second is the pathway through phosphatidylinositol 3-kinase (PI3-K) to $p70^{S6K}$, the kinase that phosphorylates ribosomal protein S6. CCK, carbachol, and bombesin activate $p70^{S6K}$ in rat acini by a pathway that can be blocked by PI3-K inhibitors such as wortmannin, and by rapamycin, which binds intracellularly to the immunophillin FKBP-12. [47] Because $p70^{S6K}$ also mediates the initiation of protein translation, this pathway probably underlies, at least in part, the stimulation of protein synthesis in acinar cells by pancreatic secretagogues discussed earlier. Identification of intracellular regulation by both conventional and novel pathways is a rapidly developing field, and further progress is expected.

INHIBITION OF PANCREATIC SECRETION

The regulation of pancreatic secretion depends on a balance between inhibitory and stimulatory influences on the gland, which are exerted through hormones and the autonomic nervous system. Although much has been written about pancreatic stimulation, less is known about the inhibitory influences on the pancreas.

Inhibitory Phase of Pancreatic Secretion

In humans, hyperglycemia induced by intravenous infusion of glucose inhibits the pancreatic secretory response to a test meal. [251] Similarly, intravenous infusion of amino acids inhibits the human pancreatic enzyme response to intestinal amino acid perfusion. [252] Although the mechanisms responsible for these observations are unknown, the secondary release of inhibitory hormones is postulated. Pancreatic glucagon exhibits characteristics consistent with such an inhibitory hormone. In most of the studies, glucagon inhibits pancreatic secretion stimulated by secretin and CCK, alone or in combination, or by ingestion of a test meal in dogs, cats, rats, and humans. [3], [253], [254] and [255] The inhibitory effect is characterized by reduction of the volume of flow and of HCO_3^- and enzyme secretion.

Pancreatic glucagon is secreted concomitantly with the hyperaminoacidemia observed after the intestinal perfusion of amino acids or a high-protein meal. [208] This postprandial level of glucagon may be sufficient to inhibit secretin- or CCK-stimulated pancreatic secretion.

Another pancreatic hormone, somatostatin, may also play a role in the inhibition of pancreatic secretion. Somatostatin-containing cells are also present in the upper gastrointestinal tract and the central nervous system. Somatostatin is processed to multiple molecular sizes and interacts with a variety of receptors on acinar cells, dorsal root ganglia, and islets of Langerhans, and on nerves of the peripheral and central nervous systems. One mechanism to provide a degree of specificity could be the release of different forms of somatostatin. For example, glucagon-like peptide 1, oleic acid, and gastrin-releasing peptide stimulate the secretion of both somatostatin-28 and somatostatin-14, but secretin induces a preferential release of somatostatin-14. [256] In humans, pharmacological doses of somatostatin cause marked inhibition of CCK-stimulated pancreatic enzyme secretion and modest inhibition of secretin-stimulated HCO_3^- secretion. [4], [257] Studies in rats demonstrate that somatostatin inhibits 2-deoxy-D-glucose– and CCK-evoked pancreatic enzyme secretion through a vagal pathway. [11] Somatostatin does not act on peripheral vagal afferent or efferent pathways or directly on pancreatic acini; it exerts its inhibitory action at a central vagal site. [11] Studies of the perfused canine pancreas have demonstrated that somatostatin is released from the pancreas during perfusion with high concentrations of amino acids or glucose. [258] This peptide may exert a paracrine inhibitory effect on the exocrine pancreas.

Six somatostatin receptor (SSTR) subtypes have been cloned from five genes; SSTR2 is expressed in an A and a B form by variant mRNA splicing. Inhibition of pancreatic secretion is mediated by SSTR2 receptor agonists (e.g., octreotide), whereas SSTR5 inhibits insulin release. In addition, SSTR2A has been localized to acinar cells, and to glucagon and PP immunoreactive islet cells. [259] Although somatostatin is one of the few peptides to inhibit pancreatic secretion in an isolated pancreas, the role of the acinar cell SSTR may be to modulate the nonparallel secretion of digestive enzymes [260] rather than to inhibit secretion. It is believed that somatostatin inhibits pancreatic exocrine secretion mainly through a central cholinergic mechanism. [11]

Intrajejunal perfusion of hypertonic glucose (50%) produces dose-related inhibition of secretin-stimulated pancreatic fluid and HCO_3^- secretion in humans. [261], [262] Similar inhibition occurs with intraintestinal hypertonic (9%) NaCl infusion in dogs. At least part of this inhibitory effect has been attributed to the release of enteric glucagon. [252] Infusion of oxyntomodulin, a 37–amino acid, glucagon-containing peptide isolated from porcine lower intestine, inhibits basal and cerulein-stimulated pancreatic secretion of HCO_3^- and enzymes. [263] This intestinal glucagon is ten times more potent than pancreatic glucagon.

Factors in the ileum and colon can inhibit pancreatic secretion. Hage and colleagues [264] demonstrated that infusion of oleic acid into the proximal colon of conscious dogs inhibits secretin-stimulated pancreatic secretion. Similar inhibitory effects have been observed in rats. [265] In the anesthetized cat, infusion of oleic acid or hypertonic glucose or saline into the colon or terminal ileum inhibits secretin- or CCK-stimulated secretory volume and HCO_3^- and enzyme output from the pancreas. [266]

In humans, nutrients (e.g., lipid) in the colon inhibit CCK-stimulated pancreatic enzyme and HCO_3^- output. [267] These late postprandial events may serve as physiological signals to reduce exocrine pancreatic secretion after digestion and the absorption of nutrients are complete. The inhibitory effect of nutrients in the distal gut on pancreatic secretion appears to be independent of the vagus and splanchnic nerves. [266] Cross-circulation studies in the rat have shown that a humoral factor mediates the inhibition of pancreatic enzyme secretion induced by colonic perfusion of oleic acid.

Harper and colleagues [268] used the term *pancreotone* to describe an inhibitory substance extracted from the colonic mucosa. The function of pancreotone is abolished when the extract is preincubated with trypsin, demonstrating it is a peptide. Peptide YY, a 36–amino acid peptide named for its amino- and carboxyl-terminal tyrosines, is abundantly present in the distal small intestine, colon, and rectum. [269] This peptide is released by fat and, to a lesser degree, protein in the distal gut or colon. The infusion of this peptide in dogs significantly inhibits basal and meal-stimulated pancreatic HCO_3^- and enzyme secretion. [270] Physiological experiments demonstrate that intraileal, but not intracolonic, carbohydrate increases plasma peptide YY levels and decreases amylase secretion in dogs. [271] These observations support the

hypothesis that peptide YY is at least a component of pancreotone in dogs. In humans, ileal perfusion of carbohydrate inhibits exocrine pancreatic secretion. Glucagon-like peptide 1 (GLP-1), another ileal hormone, but not peptide YY, is elevated in the circulation during ileal infusion of carbohydrate. GLP-1 does not appear to act directly on the pancreas to inhibit exocrine secretion. In anesthetized pigs [272] with cut splanchnic nerves, intravenous infusion of GLP-1 inhibited hypoglycemia-induced pancreatic HCO_3^- and protein secretion. These effects were not observed in vagally stimulated, isolated, and perfused porcine pancreas, [272] suggesting that GLP-1 acts via a central mechanism. Subsequent studies in rats indicated that the inhibitory action of GLP-1 depends on intact vagus nerves. [12] GLP-1 acted on the dorsal vagal complex to inhibit pancreatic enzyme secretion. [12] Similarly, oxyntomodulin, which is released after ileal administration of nutrients, also inhibits pancreatic secretion by a vagus-dependent central mechanism. [273] Because oxyntomodulin does not interact with the receptor for glucagon or truncated GLP-1, its effects appear to involve an oxyntomodulin-specific receptor. Although the mediator(s) of ileal carbohydrate-induced inhibition of exocrine pancreatic secretion has not been firmly identified, it appears that the action of these potential mediators is dependent on central neural pathways.

PP, a peptide closely related to peptide YY, is another hormone that may play an important role in regulating pancreatic exocrine secretion. PP is localized in the islets of Langerhans and between the acinar cells of the exocrine pancreas. [274] Its only apparent physiological actions are to inhibit pancreatic and biliary secretion. The secretion of PP is governed mainly by a cholinergic mechanism. [275] Postprandial release of PP is mediated by a long vagovagal reflex and short local cholinergic pathways. [275] Vagal cholinergic activity is the most powerful stimulant of PP release, and it is also key to most other stimulation of the PP cell. [275]

In humans and dogs, infusion of physiological concentrations of PP inhibits basal and stimulated pancreatic secretion. [5], [276] In vivo, PP appears to act preferentially by inhibiting vagal stimulation. [277] In vitro, PP inhibits pancreatic enzyme secretion by way of the presynaptic modulation of acetylcholine release. [278] Because its secretion is under cholinergic control and it acts by interfering with cholinergic transmission, PP is an ideal candidate to modulate pancreatic secretion stimulated by the cholinergic enteropancreatic reflex. After ingestion of a meal, the enteropancreatic reflex is activated to stimulate pancreatic enzyme secretion and PP release. PP inhibits cholinergic transmission and reduces pancreatic enzyme secretion. PP may play an important role in the feedback regulation of pancreatic enzyme secretion activated by the enteropancreatic reflex. The primary target of PP appears to be the central nervous system. [13] PP receptors have been identified in discrete locations in the hypothalamus, limbic system, brainstem, and other central locations with the use of receptor autoradiography. [279] In contrast, mRNA is almost undetectable, [280] suggesting that peripheral PP is modulating central neural function through sites with an incomplete blood-brain barrier. Microinjection of PP into the dorsal motor nucleus inhibits pancreatic secretion stimulated by CCK, suggesting that the dorsal motor nucleus of the vagus is a site for neural feedback inhibition of pancreatic exocrine secretion. [281]

Although the list of peptides known to inhibit exocrine pancreatic secretion has expanded, little is known about the mechanisms through which hormones or neurotransmitters inhibit pancreatic enzyme secretion. An important feature shared by these agents is the lack of direct inhibition of the pancreatic acinar cells. Many substances suppress pancreatic enzyme secretion in vivo but do not act directly on the acinar cell to suppress enzyme release. Based on animal studies, it appears that peptides such as PP, somatostatin, CGRP, enkephalin, and pancreastatin inhibit pancreatic enzyme secretion by modulating cholinergic transmission, and most, if not all, act through a central vagal site. [277], [278], [282], [283], [284] and [285] Intracerebroventricular administration of CGRP inhibitors stimulates (basal) pancreatic secretion in conscious rats, and this appears to be mediated by sympathetic nonadrenergic efferents through the a-adrenergic receptor. [286] In contrast, CGRP in the central nervous system inhibits pancreatic enzyme secretion stimulated by 2-deoxy-D-glucose and CCK by modulating vagal parasympathetic outflow. [287]

Feedback Regulation of Pancreatic Secretion

A series of observations suggests that the intralumenal action of pancreatic proteases plays an important role in regulating pancreatic enzyme secretion. [89], [288] The underlying concept of feedback regulation of the pancreas is based primarily on studies of rats that show that diversion of pancreatic juice from the duodenum stimulates CCK release and pancreatic enzyme secretion. [90] However, intraduodenal administration of trypsin or chymotrypsin inhibits the release of CCK and pancreatic enzymes. [90] This phenomenon is specific for activated proteases and is not observed with inactivated trypsin, amylase, lipase, or $NaHCO_3$. Subsequent studies demonstrated that intravenous infusion of proglumide, or L364,718, a specific CCK receptor antagonist, abolishes the increase in pancreatic exocrine secretion evoked by the diversion of bile-pancreatic juice. [90], [289] These observations indicate that feedback inhibition of pancreatic secretion by trypsin is mediated by inhibiting the release or action of CCK.

The increased plasma CCK levels and pancreatic secretion after diversion of pancreatic juice appear to be mediated by a trypsin-sensitive substance secreted by the proximal small intestine, which has been designated *CCK-releasing factor (CCK-RF)*. [92] When trypsin is present, this peptide is cleaved and inactivated. CCK-RF may act as a mediator of pancreatic enzyme secretion in response to dietary protein intake in rats. Dietary protein in the intestine competes for the trypsin that would otherwise inactivate CCK-RF. [91] The resulting increase of CCK-RF in the intestinal lumen enhances CCK release, stimulating pancreatic enzyme secretion. Although this appears to be the principal mechanism regulating CCK release in rats, it is not known whether the same mechanism operates in other species.

A number of CCK-releasing peptides have been identified, including one from porcine mucosa [14] and another from rat upper intestinal fluid. [15] Herzig and colleagues [14] isolated and purified a CCK-releasing peptide from porcine small bowel mucosal extracts. This trypsin-sensitive peptide, which is secreted intraduodenally, releases CCK and stimulates pancreatic secretion in rats (Fig. 15-9). Peptide sequencing and mass spectrometry have shown that this peptide is identical to the porcine diazepam-binding inhibitor. Diazepam-binding inhibitor–like immunoreactivity has been found in intestinal mucosal cells, [290] and this peptide is secreted into the lumen under neural regulation. [291] Peptone in the duodenum stimulates serotonin release, which activates the sensory substance P neurons in the submucous plexus. Signals are then transmitted to cholinergic interneurons and to epithelial CCK-releasing peptide–containing cells by way of cholinergic secretomotor neurons. [291] This enteric neural circuitry, which is responsible for the secretion of CCK-releasing peptide, may in turn play an important role in the postprandial release of CCK. Spannagel and colleagues [15] successfully purified another lumenal CCK-releasing peptide from the duodenal secretion of rats. Partially purified fractions increase the release of CCK and stimulate pancreatic secretion of fluid and protein. Amino acid sequencing and mass spectral analysis of this peptide reveal that it is composed of 70 to 75 amino acid residues and has a mass of 8136 kd. The monitor peptide [292] in rat pancreatic juice, which is secreted into the duodenal lumen, is another CCK-releasing peptide. The physiological significance of these releasing peptides is unknown.

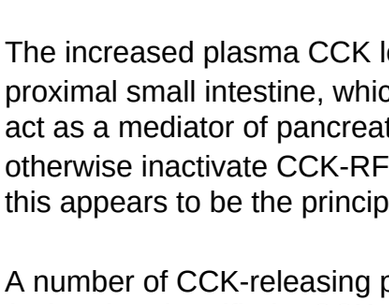

FIGURE 15-9. A schematic representation of the postulated mechanism by which cholecystokinin-RP (*CCK-RP*) stimulates the secretion of CCK postprandially. CCK-RP is secreted into the proximal small intestine and inactivated by trypsin. When food enters the duodenum postprandially, protein binds to trypsin and prevents the inactivation of CCK-RP. CCK-RP stimulates CCK cells in the duodenum to release CCK into the bloodstream. CCK, in turn, stimulates pancreatic enzyme secretion. (From ref. [14].)

Despite several attempts to demonstrate a protease-sensitive feedback mechanism in humans, the issue remained controversial because the technical limitations in removing or blocking intralumenal protease activity made studies of humans difficult. Using a different approach, researchers have reported that intestinal administration of trypsin or chymotrypsin in humans suppresses CCK release and partially blocks the pancreatic response to intestinal administration of amino acids or oral ingestion of a test meal. [97], [293] These results support the existence of feedback regulation of pancreatic enzyme secretion in humans.

Using an alternative approach, Dlugosz [294] and Hotz [295] and their colleagues observed that duodenal infusion of aprotinin, a trypsin inhibitor, had no effect on basal pancreatic enzyme secretion. Similar findings were reported with use of the trypsin inhibitor FOY-305. [296] Neither compound, however, strongly inhibits human

chymotrypsin. Liener and colleagues [297] demonstrated that Bowman-Birk soybean trypsin inhibitor, an inhibitor of chymotrypsin and elastase, markedly stimulates pancreatic enzyme secretion in humans. These observations suggest that the removal of trypsin and other proteases such as chymotrypsin and elastase would evoke pancreatic enzyme secretion in humans.

Studies of rats show that biliary secretion may also participate in the feedback regulation of CCK release. Secretion of bile into the proximal intestine inhibits CCK release and pancreatic secretion by protecting pancreatic proteases from autodigestion in the lumen. [298]

Postprandial pancreatic enzyme secretion is under hormonal and neural control. Distention of the duodenum or administration of hyperosmolar solutions into the duodenum elicits pancreatic enzyme secretion without raising plasma CCK levels. [159] This stimulatory effect is inhibited by atropine, suggesting that it is cholinergically mediated. In contrast to amino acid–stimulated pancreatic enzyme secretion, pancreatic responses to stimulation by volume or osmolality in the duodenum are not suppressed by trypsin. [159] This indicates that feedback regulation of pancreatic secretion by trypsin is stimulus specific and is mediated by inhibiting CCK release. The enteropancreatic reflex is unaffected by intralumenal proteases.

The existence of a feedback regulation of pancreatic enzyme secretion in humans may have important clinical implications. It is conceivable that in patients with chronic pancreatitis, decreased pancreatic enzyme secretion may result in elevated plasma CCK levels, reflecting a failure in the feedback modulation of CCK release. This may cause hyperstimulation of the pancreas and produce pain. Effective enzyme replacement therapy may reduce pancreatic stimulation, decrease intraductal pressure, and diminish pain. Large doses of pancreatic extract have reduced pain in some patients with chronic pancreatitis (see Chapter 95). [299], [300] This exciting observation awaits confirmation.

PATTERNS OF SECRETION

Basal Secretion

Under basal conditions, pancreatic secretion occurs at very low rates, although a small amount of enzymes is always present in the pancreatic juice. Basal secretion rates of enzymes and HCO_3^- are about 10% and 2% of maximal levels, respectively. A pattern of cyclic change in basal pancreatic secretion has been demonstrated in dogs [301] and humans, [302] and this is characterized by brief increases in HCO_3^- and enzyme secretion, which recur every 60 to 120 minutes during the interdigestive period. These bursts of pancreatic secretory activity are temporarily associated with periods of increased motor activity in the stomach and proximal intestine known as *interdigestive migrating motor complexes (IMMCs)*. [301], [302]

Associated with the bursts of pancreatic secretion are brief increases in gastric acid and biliary secretion. [302] Plasma motilin and PP levels also fluctuate in phase with the IMMCs. [302], [303] The concentrations of pancreatic enzymes and bile acids during the transient surge of pancreaticobiliary secretion are similar to maximal postprandial outputs, although the concentrations diminish rapidly with the onset of type III duodenal motor activity. [302], [303] It has been postulated that the cyclic secretion of pancreatic and biliary juice may be important in the digestion of residual food particles or cellular debris in the gastrointestinal tract during the interdigestive period.

The mechanism of control of the cyclic patterns of pancreatic secretion is unclear. Bursts of increased acid secretion are unlikely to be the principal mediators for the cyclic changes in pancreatic secretion, because removal of gastric acid by aspiration or through a fistula does not affect the pattern of interdigestive pancreatic secretion. [302], [303] Infusion of motilin prematurely initiates cyclic pancreatic secretion and shortens the periodicity between peaks. [304] Administration of motilin antiserum abolishes the cyclic pattern of pancreatic secretion. [305] Cholinergic blockade with atropine also markedly decreases trypsin output and abolishes interdigestive motor activity. However, administration of phentolamine, an a-adrenergic blocker, increases basal trypsin output fourfold without disrupting the periodicity of pancreatic secretion. [306] These observations suggest that motilin and the autonomic nervous system are important in the initiation of the cyclic pancreatic secretion that occurs during fasting.

Prandial and Postprandial Secretion

After ingestion of a meal, the exocrine pancreas is stimulated to secrete enzymes and HCO_3^-. Total postprandial pancreatic output is about 60% to 70% of the output attained in response to maximal stimulation with intravenous infusion of CCK. [99] The stimulatory effect of a meal can be described by separating its components into cephalic, gastric, and intestinal phases (Table 15-1).

PHASE	PANCREATIC RESPONSE (%)	STIMULANTS	MEDIATORS
Cephalic	25	Sight, smell, taste, eating	Vagal innervation
Gastric	10	Distention	Vagal-cholinergic pathways
Intestinal	50–70	Amino acids	Cholecystokinin, secretin
		Fatty acids	Enteropancreatic reflexes
		Ca^{2+}, H^+	Other hormones (?)
		Distention	

TABLE 15-1 Three Phases of Postprandial Pancreatic Secretion

Cephalic Phase In humans and experimental animals, pancreatic secretion rich in enzymes is stimulated by the sight, smell, and taste of appetizing food. [307] This cephalic effect in dogs amounts to 25% of the enzyme response to an ordinary meal. In humans, the contribution of the cephalic phase to the postprandial pancreatic enzyme secretion appears to be larger and amounts to 50% of the maximal responses induced by exogenous secretin and CCK. [308] The pancreatic response to sham feeding lasts only for the duration of feeding. [308] The vagus nerve appears to be important in mediating the cephalic phase, as this phase can be completely abolished by vagotomy in rats. [309] Administration of an anticholinergic drug decreases or abolishes the pancreatic response to sham feeding in humans. [308] The efferent cholinergic fibers probably act directly on the pancreas because vagal stimulation causes pancreatic secretion in dogs even when the pancreas is perfused extracorporeally to eliminate any humoral effects of nerve stimulation. [310] Sham feeding increases gastric acid secretion, but because sham feeding induces a pancreatic response in patients who are achlorhydric, it is unlikely that gastric acid secretion contributes significantly to the cephalic phase of pancreatic enzyme secretion.

Gastric Phase In dogs and humans, gastric distention increases the rate of pancreatic enzyme secretion. [311], [312] Gastric distention to a volume of 250 to 400 mL with a balloon doubles the basal pancreatic protein output. [313] Although the actual contribution of the gastric phase to the total postprandial pancreatic secretion has not been determined in humans, the magnitude of the distention-induced pancreatic response in dogs is about 20% of the maximal CCK response over a range of distention from 300 to 1500 mL. [311] Vagotomy and atropine reduce or abolish the pancreatic response to gastric distention, suggesting that it is mediated mainly by vagal cholinergic pathways. [312], [313] The mechanoreceptors appear to be located in the body of the stomach because distention of antral pouches does not stimulate pancreatic secretion, whereas distention of the remaining stomach does. [313] The presence of food in the stomach also releases antral hormones such as gastrin or gastrin-releasing peptide, which can stimulate pancreatic secretion directly or indirectly. However, this possibility is considered unlikely because a transplanted portion of pancreas does not respond to gastric distention in dogs. [313] Moreover, distention of the intact stomach has only a slight effect on gastrin release in humans and dogs. [314], [315] The stomach facilitates digestion by fractionating solid food into small particles and by initiating digestion of dietary proteins and lipids by pepsin and gastric lipase, respectively. [316], [317] Gastric emptying is important in determining the rate of delivery of acid and nutrients into the duodenum, thereby determining the pattern and magnitude of the intestinal phase of pancreatic secretion. Abnormal postprandial pancreatic enzyme secretion is common after gastric surgery. [156]

Intestinal Phase The intestinal phase is the most important phase of postprandial pancreatic secretion. In humans and animals, the delivery of food into the small intestine stimulates pancreatic enzyme secretion to about 70% of the maximal level. [156] The major hormonal mediators of the intestinal phase of pancreatic secretion are secretin and CCK. Intestinal serotonin also appears to play an important role in the mediation of postprandial pancreatic secretion through the vagal cholinergic pathway. [115], [116] The intestinal mucosa has receptors for important vagal cholinergic reflexes that regulate pancreatic HCO_3^- and enzyme secretion. The proximal intestine plays an important role in the stimulation of pancreatic HCO_3^- secretion, primarily by the release of secretin. Although duodenal pH is the major regulator for the release of secretin, nonacid factors such as fatty acids and bile may also participate. The physiology of secretin was discussed in a previous section. Among the hydrolytic products of digestion, amino acids and fatty acids are potent stimulants of enzyme secretion but have only a weak effect on water and HCO_3^- secretion. Amino acid mixtures are more potent than fatty acids, and among the amino acids, only phenylalanine, valine, and methionine stimulate enzyme secretion in humans,

but in dogs, phenylalanine, leucine, tryptophan, oligopeptides, and casein are effective. [318] The pancreatic response to intestinal perfusion with amino acids above a concentration of 8 mmol/L depends on the total load administered. [319] This dependence on load rather than concentration is the result of exposure of longer segments of small intestine to amino acids at a concentration above a threshold value. In humans, the mechanisms responsible for the pancreatic response to amino acids are confined to the duodenum and jejunum; amino acid perfusion into the ileum elicits no response. Undigested fats are ineffective in stimulating pancreatic secretion, but fatty acids in micellar form are potent pancreatic stimulants when present in micellar form. [74] Monoglycerides, the other product of lipolysis, also stimulate pancreatic secretion. [74], [318] The chain length of fatty acids influences their potency in stimulating pancreatic secretion. In humans, the order of potency is C18 > C12 > C8. [320] Other factors that influence pancreatic response to fatty acids include the degree of saturation, the concentration and total load, and the concentration of bile salts relative to fatty acids. [317] In humans, intestinal perfusion of 10 mmol of monoolein per liter produces a pancreatic enzyme output greater than that stimulated by intestinal amino acids and almost equal to the maximal response to exogenous CCK. [321] The release of CCK and intestinal serotonin by nutrients and mechanical factors appears to play a major role in mediating the intestinal phase of pancreatic enzyme secretion. Plasma CCK levels increase after oral or intraduodenal administration of fat and protein or amino acids. [95], [96] and [97], [322] Administration of proglumide, a CCK receptor antagonist, partially inhibits pancreatic secretory responses to intestinally perfused amino acids and fat emulsions. In contrast, a wide variety of non–CCK-dependent stimuli, such as acid, carbohydrates, and mechanical factors, stimulate pancreatic secretion through intestinal serotonin. [115], [116] Serotonin, in turn, stimulates submucosal vagal afferent fibers to evoke pancreatic exocrine secretion through a vagal cholinergic pathway. Increased firing rates in peripheral afferent neurons and in central sites have been recorded during intestinal perfusion with amino acids. [161], [323] This finding, coupled with the observation that truncal vagotomy or administration of atropine markedly increases the latency of the pancreatic secretory response to intestinal nutrients but not to CCK, indicates the participation of vagovagal cholinergic reflexes. [324] Ca^{2+}, which is intimately involved in the action of CCK on pancreatic acinar cells, stimulates the pancreas. In humans, intraduodenal perfusion of Ca^{2+} solutions in concentrations similar to those found in the duodenum after ingestion of a meal stimulates pancreatic enzyme secretion and gallbladder contraction. Intraduodenal Ca^{2+} concentrations of 12 to 25 mmol/L induce pancreatic enzyme responses similar to the maximal enzyme output evoked by intravenous infusion of CCK. [325] Intestinal perfusion with $MgSO_4$, $MgCl_2$, and $ZnSO_4$ also stimulates pancreatic enzyme secretion by undetermined mechanisms. [326] The human duodenum contains receptors for volume and osmolality that mediate pancreatic enzyme secretion. Volume distention or hyperosmolar solutions in the duodenum elicit pancreatic enzyme secretion via intestinal serotonin without raising plasma CCK levels. [115], [159] This enzyme secretion is inhibited by atropine, suggesting mediation by cholinergic pathways. [159] The volumes of saline required to induce pancreatic secretion are as low as 1 to 5 mL/min, within the range observed in the duodenum postprandially. The degree of stimulation by volume receptor or osmoreceptor activation is 15% to 20% of the maximal enzyme response to CCK. [158] Therefore, both CCK- and non–CCK-dependent stimuli act in concert to mediate the intestinal phase of pancreatic secretion.

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CHAPTER 16

Steven A. Weinman and Nyingi Kemmer

BILE SECRETION AND CHOLESTASIS

The formation and secretion of bile are among the most central and earliest recognized functions of the liver. [1], [2], [3], [4], [5], [6] and [7] In addition to its digestive function as the source of intraintestinal bile acids, [8] bile formation is necessary for numerous other functions of the liver. These include the obvious functions, such as elimination of toxins, heavy metals, and xenobiotics, [9], [10] and [11] and the less obvious, such as regulation of whole-body lipid homeostasis [12], [13] and prevention of liver toxicity from endogenously produced bile acids. [14] *Cholestasis* is a term that was initially defined in terms of morphologic criteria, [15] but it also refers to a syndrome of serum biochemical abnormalities in conjunction with distinct functional abnormalities. The unifying process of cholestasis is an abnormality of the composition and rate of bile formation. [16], [17]

This chapter describes the cellular and molecular processes responsible for bile formation and the mechanisms that result in cholestasis. Knowledge in this field has increased dramatically, and many excellent detailed review articles on this subject are available. [18], [19], [20], [21], [22], [23], [24], [25] and [26] Specific cholestatic states are discussed as examples of the mechanisms involved, but the focus of this chapter is on cellular processes, and the reader is referred to other chapters for specific discussions of diagnosis and management of cholestatic liver diseases such as drug-induced cholestasis, primary sclerosing cholangitis, primary biliary cirrhosis, and the approach to the jaundiced patient.

GENERAL ASPECTS OF BILE FORMATION

Bile is the secretory product of the liver and bile ducts. It flows through the biliary tract and empties into the duodenum. Bile secretion results from the transport activities of several different epithelia working in concert. It is initially produced by hepatocytes, and its volume and composition are subsequently altered as it passes through the bile ducts. [6], [27] Bile is a complex suspension that has an electrolyte composition similar to that of plasma, except for higher HCO_3^- concentration. In addition, bile contains significant concentrations of bile acids, phospholipids, cholesterol, proteins, amino acids, and peptides. [16] Heavy metals, particularly copper, are also excreted in bile, and biliary metal excretion is the major mechanism for their elimination.

Anatomy and Physiology of Bile Secretion and Enterohepatic Circulation

Hepatocytes are specialized epithelial cells with distinct basolateral and apical membrane domains. They are depicted schematically in Figure 16-1. The sinusoidal or basolateral membrane surrounds the majority of the cell, and the canalicular or apical membrane occupies a small area of the cell surface near the points of contact between adjacent cells. The canalicular membranes, from which numerous microvilli protrude, make up only about 15% of the total surface area. [28] Opposing canalicular membrane domains border an enclosed space called the *bile canaliculus* measuring about 1 to 2 µm. [6] At the point of contact of the two cell membranes is the tight junction, also known as the *zonula occludens.* Tight junctions constitute a permeability barrier that consists of several transmembrane proteins including members of the claudin family and occludin. These proteins form a seal between the membranes of adjacent cells. [29] Intracellular proteins such as ZO-1, ZO-2, and numerous others serve as anchors for the claudin molecules. [30], [31] and [32] These tight junctions are cation selective, and they form a barrier between the bile canaliculus and the sinusoidal space that prevents back-diffusion of the secreted organic anions. [33], [34]

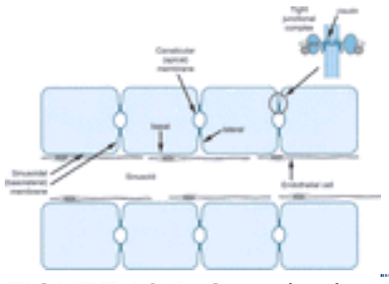

FIGURE 16-1. Organization of hepatocyte membrane domains. The relationship of canalicular and basolateral membrane domains is represented for the heptaocyte plates and sinusoidal space. The sinusoidal space and canalicular space are separated from each other by the tight junctional permeability barrier, which is composed of the extracellular loops of claudin molecules from adjacent cells.

Bile is produced as a result of the osmotic consequences of solute secretion. [35] The hepatocytes directly secrete solutes, primarily bile acids, into the canalicular space, and water follows passively driven by osmotic forces across the tight junctions and the cell membranes themselves. [16] The bile canaliculi encircle the hepatocytes in a beltlike fashion and form an interconnecting three-dimensional network. This network communicates with the canals of Hering, which form the transitions to the smallest bile ductules. The flow into the canalicular network thus results in net fluid flux into the biliary tree. Small bile ducts can secrete or absorb fluid and consequently modify the composition and amount of bile. [36], [37] and [38] They serve as tributaries of the larger ducts. During fasting, the sphincter of Oddi contracts and the gallbladder relaxes, causing bile to enter the gallbladder for storage, but postprandially, the gallbladder remains contracted and the sphincter of

Oddi relaxes, resulting in bile's bypassing the gallbladder and emptying directly into the duodenum.

In the duodenum and proximal ileum, some bile acids, particularly unconjugated bile acids, are passively absorbed, but conjugated bile acids are primarily absorbed in the terminal ileum. This absorption results from the apical Na^+-dependent bile acid transporter (ASBT), a Na^+-dependent transport protein located in the apical membrane of the ileal enterocytes. [39] Within the enterocytes, bile acids interact with a cytosolic bile acid binder, the ileal lipid-binding protein, [40], [41] and are transported to the basolateral membrane. From here they are secreted into the superior mesenteric vein by an Na^+-independent organic anion exchange system. [42] The identity of this ileal basolateral transporter appears to be a protein that results from alternative splicing of the same gene that produces the ASBT protein. [43] In this case, a truncated protein molecule localizes to the basolateral instead of the apical membrane and mediates Na^+-independent transport of bile acids from the cells into the portal circulation. [43] The bile salt bound to albumin is then transported into the portal venous blood, through the fenestrae between the sinusoidal endothelial cells, and passes into the space of Disse. It is efficiently extracted by specific transporters present in the basolateral membrane of the hepatocytes. [44] The uptake of bile salts occurs predominantly in periportal zone 1 hepatocytes. [45] Although 98% of bile salts are typically extracted from the portal blood during a single pass, the fractional extraction varies considerably according to the particular bile acid molecule. The general scheme for this enterohepatic circulation of bile acids is depicted in Figure 16-2.

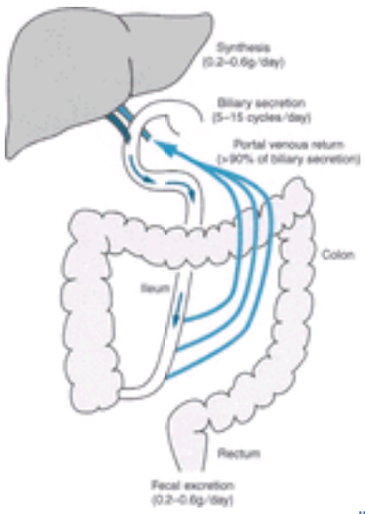

FIGURE 16-2. Schematic representation of the enterohepatic circulation. The routes of secretion and absorption of bile acids are indicated.

Multiple solutes are responsible for bile flow, but the primary ones are organic anions, and the most important of these are bile acids. Bile acids are amphipathic products of cholesterol metabolism that have several important functions. [46] They are natural detergent molecules that assist in solubilizing the lipid components of bile and are critical for solubilization, digestion, and absorption of lipids in the intestine. In addition, they serve as the primary vehicle for cholesterol elimination, [13] they can specifically trigger apoptosis, [47], [48] and they bind to transcription factors altering gene expression. [49], [50] and [51] Bile acids are synthesized de novo from cholesterol within the hepatocytes. They are also efficiently recycled through the enterohepatic circulation. Each day, up to 30 g of bile acid is secreted into bile, but only approximately 3 g is newly synthesized.

During the transit of bile through the bile ducts, its composition and volume are modified. [52] Some bile acids in the biliary tract are absorbed by the biliary epithelial cells and are returned into the portal circulation without entering the intestine. This process is known as *cholehepatic shunting* and serves to amplify the amount of fluid flow produced by a given net bile acid secretion. [53] Cholangiocytes take up unconjugated bile acids passively after protonation by H^+ derived from the activity of carbonic anhydrase present in the cholangiocytes. [53] In addition, conjugated bile acids can be taken up by the activity of a specific Na^+/bile acid cotransport protein, ASBT, which is present in the apical membrane of both cholangiocytes and ileal absorptive epithelial cells. [39], [54] Intracellular bile acids are then transferred to the basolateral membrane and are transported through the periductal capillary plexus into the sinusoids for uptake into the hepatocytes. Cholehepatic shunting plays an important role in bile formation, but it also is a mechanism of removal of potentially toxic bile acids during pathological states, such as bile duct obstruction. It has also been postulated to play a signaling role in which hepatic bile acid secretion can alter the function of the cholangiocytes themselves. [55]

Role of Bile Acids in Bile Flow

The concept that bile acids increase bile flow because they provide an osmotic driving force for filtration of water and electrolytes was first proposed by Sperber, [35] and it was subsequently confirmed by Brauer and colleagues, [56] who demonstrated that the administration of increasing loads of bile acids to rats led to an increase in bile flow. It was further demonstrated that the amount of bile acid in bile exhibited a linear correlation with the amount of bile flow. [1], [57] Although there were no circumstances in which bile acids were absent from bile, the linear relationship allowed an extrapolation back to a theoretical quantity of bile flow that would be predicted to occur in the complete absence of bile acids. This type of analysis has led to the concept that bile flow has two discrete components: bile acid–independent flow, which would occur entirely in the absence of bile acids, [58] and bile acid–dependent flow, which is a result of the osmotic coupling of water to bile acid transport. [59]

The concepts of bile acid–dependent flow and bile acid–independent flow are useful descriptions of the type of data obtained from perfused livers, assuming that the liver is a scientific "black box" with analysis restricted to measuring input and output only. Unfortunately, this concept suggested that there are two fundamentally different mechanisms at work in the liver, such as organic ion transport for bile acid–dependent flow and inorganic salt transport for bile acid–independent flow. [16] We now know that essentially all canalicular bile formation is the consequence of organic ion transport, and the fraction referred to as bile acid–independent flow results largely from the transport of other organic compounds such as glutathione and its conjugates, [60], [61] by mechanisms fundamentally the same as for bile acids.

Fortunately, since the early 1990s, dramatic advances have been made in the identification and characterization of the transport proteins responsible for hepatic transport and bile formation. We are now able to understand the mechanisms of bile formation on a molecular rather than a "black box" level. The concepts of bile acid–independent and bile acid–dependent flow have thus given way to a specific understanding of what is being transported and how. This new information has allowed an explanation of the transport specificity of the liver and the mechanisms of decreased bile flow in cholestasis. It has also successfully predicted the phenotype of genetic and acquired liver diseases. To understand the mechanisms of bile formation and cholestasis, therefore, it is critical to consider the identities and regulation of specific transport proteins at each of the critical steps of the process.

HEPATIC TRANSPORT MECHANISMS

Inorganic Ion Transport

Hepatocytes possess multiple transport proteins for inorganic ions. These primarily function to generate transmembrane ion gradients, to regulate intracellular pH and cell volume, and to create the electrical potential difference between the cell interior and the extracellular space. Although inorganic ion transport in hepatocytes generates the electrochemical gradients necessary for bile acid transport, it is not directly responsible for bile secretion. This differs from the situation in most fluid-transporting epithelia such as bile duct epithelial cells, in which inorganic ion transport is directly responsible for fluid secretion. The inorganic ion transport proteins in hepatocyte are indicated in Figure 16-3.

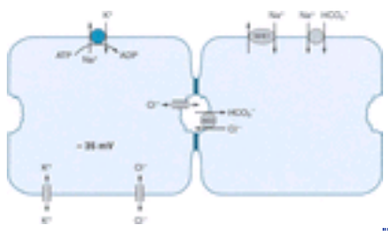

FIGURE 16-3. Inorganic ion transport pathways of the hepatocytes. Ionic gradients are maintained by Na^+,K^+-ATPase, which, in conjunction with K^+ and Cl^- channels, maintains the cell interior at an electrical potential of approximately -35 mV with respect to the sinusoid. Cl^- is distributed across the membrane passively in accordance with electrochemical equilibrium, and there is normally no net driving force for Cl^- secretion. The Na^+/H^+ exchange protein, NHE-1, the Na^+/HCO_3^- transporter, and the anion exchange protein, AE2, play important roles in pH regulation.

Na^+,K^+-ATPase is present in the basolateral membrane, [62] and it uses the energy of ATP hydrolysis to exchange Na^+ and K^+ and thus generates the low-Na^+, high-K^+ intracellular environment present in all mammalian cells. A cell negative membrane potential is generated by the presence of K^+ channels in the plasma membranes. [63], [64] The efflux of K^+ through these channels produces a cell negative diffusion potential. Hepatocytes also have multiple Cl^- channels in both the sinusoidal and canalicular membranes. [65] This allows Cl^- to distribute at its electrochemical equilibrium, and this process reduces the membrane potential to approximately -30 to -40 mV, cell interior negative. [66]

Several other transporters are primarily involved in pH regulation. [67] These are the Na^+/H^+ exchanger, NHE-1, [68], [69] and a Na^+-HCO_3^- cotransporter. [70] Both of these regulate intracellular pH in response to an acid load. The Na^+-HCO_3^- cotransporter is quantitatively more important in this response. [71] At the canalicular membrane, a Cl^- channel [72] and the Cl^-/HCO_3^- exchanger AE2 [73], [74] and [75] are present. In bile duct epithelial cells, this combination of a Cl^- channel and Cl^-/HCO_3^- exchanger results in electrogenic Cl^- and HCO_3^- secretion (see later). However, Cl^- is at electrochemical equilibrium in the hepatocyte, and there is thus no driving force for net Cl^- efflux. In hepatocytes, therefore, the AE2 transporter is primarily involved in pH regulation and not in fluid secretion.

Cell volume regulation is also extremely important for hepatocytes. Unlike most other cells, hepatocyte function requires cycles of swelling and shrinkage. After a meal, the concentrations of amino acids and sugars increase in the portal circulation. Hepatocytes take up large quantities of amino acids and other nutrients and consequently swell. [76] They respond to cell swelling by activation of K^+ and Cl^- channels. [77], [78] and [79] This produces a net flux of KCl out of the cell and reduces cell volume. [67] In addition, the Cl^- channels activated by cell swelling are also permeable to organic substrates such as taurine, and efflux of taurine contributes to volume regulation. [80] The mechanisms by which cell swelling activates ion channels are not well understood. Hepatocytes release ATP in response to cell swelling, and this ATP can bind to purinergic receptors on the cell membrane that activate Cl^- channels. [81] In addition, cell swelling directly activates protein kinases, and these play a role in channel activation as well. [82]

Uptake Transporters

Specific transport proteins at the sinusoidal membrane of hepatocytes account for the ability of the hepatocyte to take up substances from sinusoidal blood and to move them into the cells. With few exceptions, the transported molecules do not cross lipid bilayers, and their uptake is entirely determined by the properties of the transport proteins present in the sinusoidal membrane. Differences in the identities and quantities of these account for the different uptake properties of hepatocytes in different species, in different individuals, and at different times in the same individual. The most important uptake transporters are Na^+/bile acid cotransport protein (NTCP), the Na^+-dependent bile acid transporter, [83], [84] and a family of organic anion conjugate transporters known as the OATP family. [85] Members of the OATP family are responsible for the Na^+-independent uptake of bile acids, [44] unconjugated bilirubin, many other organic anions, and even some cationic and amphipathic substances. [10], [86], [87] Other important groups of transporters include the OAT family of anion transporters [88] and the OCT family of cation transporters. [89] These transporters are summarized in Table 16-1, and a schematic representation of their localization is demonstrated in Figure 16-4.

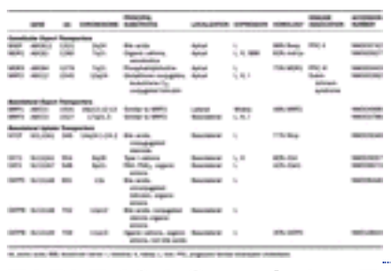

TABLE 16-1 Organic Ion Transport Proteins of the Human Hepatocyte

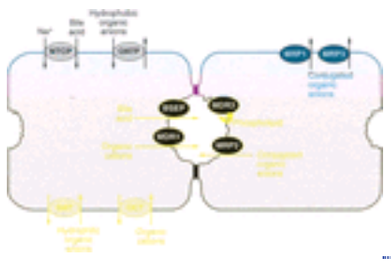

FIGURE 16-4. Organic ion transporters of the hepatocytes. Uptake at the basolateral membrane is mediated by an Na^+/bile acid cotransport protein, NTCP, and three families of organic ion exchanger transporters. The symbol *OATP* represents the organic anion transporter protein family, *OCT* represents the organic cation transporter family, and *OAT* represents the organic anion transporter family. Export from the hepataocyte into the canalicular space results from the ATP-dependent transport proteins BSEP, MRP2, MDR1, and MDR3. The ATP-dependent transport proteins MRP1 and MRP3 transport substrates out of the cells into the sinusoidal blood. See text for details.

Sodium-Bile Acid Cotransport Protein The first liver-specific transport protein to be identified was NTCP. [44], [90] The existence of a coupled Na^+/bile acid transporter was inferred from extensive studies demonstrating that the major fraction of bile acid transport was Na^+ dependent. [91], [92] The molecule responsible for this coupling, Na^+-taurocholate transport protein (Ntcp), was identified in 1989 by expression cloning with rat liver mRNA. [93], [94] Subsequently, a strongly homologous molecule (NTCP) was shown to be present at the basolateral membrane of human liver. [95] Its expression is highly liver specific, it localizes exclusively to the basolateral membrane of hepatocytes, and it is expressed similarly in periportal and pericentral hepatocytes. [96] Human NTCP is a 349-amino acid, 50-kd protein exhibiting a 77% identity with rat Ntcp. [96], [97] The human protein, NTCP, has a higher affinity for taurocholate than does rat Ntcp (K_m of 6 µM versus 25 µM) and also mediates uptake of a variety of bile salts in addition to taurocholate including glycocholate, chenodeoxycholate, glycochenodeoxycholate, taurochenodeoxycholate, and tauroursodeoxycholate. [98] Transport is strongly coupled to Na^+ with a stoichiometry of 2 Na^+ transported for each bile acid molecule. [99], [100] This coupling strategy results in an electrogenic transport process, [101], [102] and [103] which greatly increases the driving force for uptake and increases the maximum intracellular bile acid concentrations that can be achieved. It has been estimated that it can elevate intracellular bile acid concentrations at least tenfold higher than in the sinusoidal blood. [104] NTCP is a member of a small family of Na^+/bile salt cotransporters within the superfamily of Na^+/solute symporters. [105], [106] Human NTCP (Na^+/taurocholate cotransporting polypeptide) was mapped to chromosome 14q 24.1-24.2. [98] In addition, a structurally related protein, ASBT, [107] mediates uptake of bile acids in the

ileum as well as the bile duct and has been identified in rat and human ileum. [39], [54], [108], [109] These Na $^{+}$-dependent bile acid transporters are unique within the superfamily of Na $^{+}$/solute symporters in that they possess only seven membrane-spanning domains. [106], [107] The function of NTCP is critical for bile acid transport, and it is responsible for Na $^{+}$-dependent bile acid transport. Under most conditions, this accounts for one half to two thirds of the total bile acid uptake, and it is critical for achieving the rapid clearance of bile acids from portal blood. NTCP is strongly conserved among all mammalian species examined, and its critical role in uptake is reflected by its being an important target of regulation of expression during cholestasis. [20], [110], [111]

Microsomal Epoxide Hydrolase A second protein, microsomal epoxide hydrolase (mEH), has also been proposed to contribute to Na $^{+}$-dependent bile acid uptake. [112] This protein is present in both the endoplasmic reticulum and the hepatocyte sinusoidal membrane, [113] and it can mediate Na $^{+}$-dependent bile acid transport in reconstituted membrane preparations, [114] as well as in stably transfected MDCK cells. [115] However, unlike NTCP, its role in physiological bile acid uptake is uncertain. This is because its expression in other cell lines did not alter bile acid transport, [116] and it is also because the substrate and inhibitor specificity proposed for mEH does not correspond to that of the native hepatocyte. It is thus possible that mEH may serve as an intracellular bile acid transporter, but its role in sinusoidal bile acid uptake is uncertain.

OATP Family of Transporters The OATP family consists of a large number of related molecules with a broad tissue distribution and substrate specificity. [85], [110] To date, 15 members have been identified, 8 in humans (OATP-A to OATP-F, OATP8, and hPGT), [86], [117], [118], [119] and [120] and 7 in rodents (Oatp1 to Oatp5, OAT-K1/2, and rPGT). [90], [121], [122], [123] and [124] These transporters are classified within the gene superfamily of solute carriers (SLC) as gene family SLC21A. The initial member of this family to be discovered is now called Oatp1 and was identified by expression cloning from rat liver. [125], [126] It accomplishes Na $^{+}$-independent transport of bile acids, conjugated estrogens, conjugated bilirubin, cysteinyl leukotrienes, thyroid hormones, and many other substrates, [125], [127] by a process of anion exchange. [128] In addition to uptake into the cell, Oatp1 mediates the efflux of reduced glutathione (GSH) from hepatocytes into the sinusoid. [129] Because GSH is synthesized in hepatocytes and its intracellular concentrations are much higher than those in sinusoidal blood, efflux of GSH is likely to provide the driving force for electroneutral organic ion uptake. [130] Oatp1 localization is not restricted to liver, and it is present in other tissues as well. [90], [131] It is a 670-amino acid, 80-kd protein expressed at the basolateral membrane of the hepatocytes, the S_3 segment of the proximal tubule, and the apical surface of the epithelial cells of the choroid plexus. Oatp2 is a 661-amino acid, 92-kd protein, expressed at the basolateral membrane of rat hepatocytes, [132] the choroid plexus, [133] the retina, and the endothelial cells of the blood-brain barrier. [124] It also functions as an anion exchanger and mediates the uptake of bile salts, cardiac glycosides, and cyclic proteins. [132], [134] Oatp4 is another rat protein that shares 43% and 44% amino acid identities with Oatp1 and Oatp2, respectively, and is expressed exclusively in the liver, where it mediates the uptake of bile salts, conjugated steroids, and thyroid hormones. [122] Additional family members such as Oatp3, rPGT and OAT-K1/2 are not expressed in the liver. [135], [136] and [137] Unlike the situation for NTCP, in which the protein is directly homologous among species, the human OATP molecules are not the direct homologs of the earlier-identified rodent proteins. [86], [118] This has caused some confusion in the nomenclature of the human OATP proteins. Table 16-1 presents the characteristics of the human OATP proteins and the systematic gene names, as well as the names that are commonly used in the literature. OATP-C, which has also been called OATP2, or liver–specific transporter (LST-1), appears to be the primary organic ion uptake transporter in human liver. [117], [119], [138] It is a 691-amino acid protein that is primarily expressed in the basolateral membrane of hepatocytes and transports bile acids, conjugated bilirubin, conjugated sterols, eicosanoids, and thyroid hormones. Its significance is demonstrated by its ability to transport albumin-bound unconjugated bilirubin, unlike most of the other members of the OATP family. [139] It is most homologous to the rodent transporter Oatp4. OATP8 is another important multispecific transport molecule at the basolateral membrane of human hepatocytes. It has an 80% identity to OATP-C, [140] and it also has a broad substrate specificity mediating transport of bile acids, conjugated sterols, bromosulphophthalein, thryroid hormones, and organic cations. [86] OATP-B, a 643-amino acid protein that is expressed predominantly in the basolateral membrane of the hepatocytes, [86] is the third human liver OATP. It has a more limited substrate specificity than OATP-C or OATP8 and does not transport bile acids. [86] The first human OATP to be identified, OATP-A, is not thought to be an important hepatic transporter. Although it is expressed in the hepatocytes, it is found predominantly in the endothelial cells of the blood-brain barrier, where it transports bile salts, organic anions, conjugated steroids, and numerous drugs. [141], [142] and [143]

Other Uptake Transporters Several other groups of transporters are present in the basolateral membrane of hepatocytes and play a role in uptake. The OAT family comprises a group of transporters that play a role in anion transport in both liver and kidney. [88] The substrate specificity of the OAT family members differs from that of the OATP family in that substrates tend to be more hydrophilic. Nonetheless, there is considerable overlap. OAT1, the initially identified member of the group, is present only in kidney, where it functions as the high-affinity paraaminohypuritic acid transporter. This is the probenecid-inhibitable transporter that is largely responsible for efficient tubular anion secretion. [144], [145] A similar transporter, OAT2, is present exclusively in the liver, [146] a third family member, OAT3, is present in both kidney and liver, [147] and OAT4 is present in kidney and placenta. [148] The relative distribution of OATP and OAT family members between kidney and liver plays an important role in the well-known transport differences of these two organs. More bulky hydrophobic anions (OATP substrates) are preferentially taken up by hepatocytes and are excreted largely in bile rather than in urine. More hydrophilic organic anions (OAT substrates) are preferentially secreted by the renal tubules and are excreted largely in urine rather than in bile. The members of the OAT family have a 12-transmembrane domain structure and function largely as electroneutral anion exchangers. [88] In spite of these similarities, they do not share sequence homology with the OATP family. They do, however, share homology with the organic cation transporters of the OCT family. [149] The organic cation transporters mediate the uptake of small type I cations such as endogenous amines and a wide range of drugs. [89] The rat OCT1 is a 556-amino acid protein that is expressed in the basolateral membrane of the hepatocyte, kidney, colon, and small intestine. [149], [150], [151], [152] and [153] Two homologous human transporters have been cloned. The hOCT1 has a 78% identity with rat OCT1 and is expressed predominantly in the liver, whereas hOCT2 is expressed predominantly in the kidney. [154], [155] These molecules function as electrogenic cation transporters. [151], [156] Although the organic cation transporters appear to play little role in bile formation, their substrates include many important therapeutic drugs and xenobiotics.

Intracellular Movement

After uptake of organic solutes across the basolateral membrane of the hepatocytes, the solutes are transferred to the apical domain within seconds to minutes. [157] Many of the solutes transported by the hepatocytes are hydrophobic organic ions, and as a consequence they tend to bind to proteins, partition into various cellular membrane fractions, and become associated with intracellular organelles. It has been estimated that these interactions reduce the effective diffusion constants for intracellular molecules by a factor of 1000 compared with what would be expected for diffusion in free solution. [158] This intracellular binding reduces the tendency of substrates to efflux back out the sinusoidal membrane, but it also has the potential to reduce the delivery of substrates to the canalicular membrane. Transcellular transport thus requires mechanisms for these molecules to traverse the hepatocyte cytoplasm and to become available as substrate for the canalicular transporters. [159], [160] and [161]

High-affinity intracellular bile acid binding proteins are present in hepatocytes. The bile acid binding proteins are primarily 3a-hydroxysterol dehydrogenases. [162], [163] and [164] These molecules bind bile acids in the low micromolar range and have been cloned from rat and human liver. [165], [166] Other substrates also bind to intracellular binding proteins in high affinity. These include fatty acid binding protein for free fatty acids [167] and glutathione *S*-transferases, previously referred to as ligandin, [168], [169] for bilirubin.

Several functions for intracellular binding of bile acids and other substrates have been proposed. [158], [160] Because the proteins remain in the aqueous phase of the cytoplasm, they may serve to prevent bile acids from partitioning into fixed lipid membranes and organelles. In this case, protein binding serves to increase the diffusion coefficient for these molecules. [170] They may also serve as an intracellular sink preventing reflux of bile acids through the sinusoidal membrane. Lateral diffusion within lipid bilayers, as well as vesicle-vesicle transfer, is another mechanism that plays a role in intracellular bilirubin diffusion. [171] For molecules in which vesicle-vesicle transfer is important, binding proteins may interfere with this process and may actually slow the process of intracellular movement. [160]

Transcytotic vesicular transport also plays a role in hepatic transport in some situations. Certain proteins, particularly monomeric IgA, bind to specific receptors at the sinusoidal membrane and undergo receptor mediated endocytosis, transcellular trafficking, and exocytosis at the canalicular membrane. [172] Other molecules, including bulky organic solutes such as class II cations and proteins such as transferrin, translocate by this mechanism. [173], [174] and [175]

Whether small molecules, such as bile acids, are also packaged in vesicles and are exocytosed is doubtful. Several investigators have shown that a microtubule-dependent pathway is involved in bile acid secretion, [176], [177] and [178] but these results are likely because bile acids induce a vesicle mediated insertion of transporters into the canalicular membrane, [179] and not because the vesicles themselves contain significant amounts of bile acids. [175] This mechanism is discussed in detail later.

Hepatic Copper Transport The hepatic transport of copper also involves a vesicular transport process. Biliary excretion of copper is the exclusive mechanism by which copper is eliminated from the body, and a defect in this process, as occurs in Wilson disease, results in massive accumulation of copper in liver, brain, and other organs. [180] The major copper transport protein is a metal transporting (P-type) ATPase known as ATP7B. This protein is exclusively found in the liver and is

localized primarily in an intracellular trans-Golgi site. [181] Copper transport into this vesicular space is necessary for copper incorporation into ceruloplasmin and other proteins, but how this subcellular localization results in biliary copper excretion is unclear. One possibility is that ATP7B transports copper into a population of intracellular vesicles that then traffic to the canalicular membrane and discharge their contents into the bile. [182], [183] An alternative hypothesis is based on the observation that ATP7B moves away from the trans-Golgi and associates with the canalicular membrane when cellular copper content is elevated. If ATP7B directly inserted in the canalicular membrane under these conditions, it could directly pump copper into the canalicular space. [184] At the present time, this issue remains unresolved.

Canalicular Transport

The secretion of bile salts and other molecules from the cytoplasm of the hepatocytes across the canalicular membrane into the bile canaliculi is the rate-limiting step in bile formation and requires direct energy input. [6], [16] It is achieved by the action of several different ATP-dependent transport proteins. [185] These transport proteins are part of the superfamily of ATP-binding cassette (ABC) proteins and share similar structural and functional characteristics with each other. [186] In addition to canalicular membrane organic ion transporters, this family includes the cystic fibrosis transmembrane transporter (CFTR), [187] the sulfonylurea receptor, [188] and certain bacterial transport proteins. [189] Four different ABC transporters have been shown to play a significant role in canalicular secretion of biliary components. [190]

Bile Salt Exporting Protein Although NTCP can accomplish at most a tenfold increase in bile salt concentration from sinusoidal blood into the cell, biliary bile salt concentrations can be more than one hundredfold greater than that in the cell. In humans this concentrative transport step is accomplished almost exclusively by bile salt exporting protein (BSEP). [191] This molecule was initially identified as a protein of unknown function present exclusively in the liver. It shares significant homology with the multidrug resistance P-glycoptein. It was thus initially designated *sister of P-glycoprotein* or spgp. [192] This protein was subsequently shown to transport bile acids with a substrate specificity and concentration dependence identical to that seen for the bile acid transport process in isolated liver canalicular membranes. [193] Definitive proof that BSEP is the only significant bile salt export protein in human liver was provided by the observation that the genetic cholestatic disorder PFIC2 mapped to the BSEP gene locus on chromosome 2q24. [191], [194] These patients have mutations of BSEP that result in failure of the protein to function and a near complete absence of bile acid export into bile (see later). The rat, mouse, and human BSEP genes have been cloned, and the gene product is exclusively expressed in the liver. [195] Immunofluorescence microscopy demonstrates that the protein is localized in the canalicular membrane and in subcanalicular vesicles. [196] The rat protein is approximately 160 kd and shares almost 50% amino acid homology with the canalicular cation transporter, Mdr1b. [193] Although loss of human BSEP results in a near total elimination of biliary bile acid excretion, [191] a knock-out mouse missing bsep only displayed a 50% reduction in bile acid secretion. [197] This demonstrates that in mouse, unlike in humans, other proteins at the canalicular membrane can transport bile salts. This further demonstrates that the precise molecules that carry out hepatic transport and the relative proportions of each are species specific.

Multidrug Resistance Associated Protein 2 Multidrug resistance associated protein 2 (MRP2) is an ABC transport protein that is primarily expressed in the canalicular membrane of hepatocytes, [198], [199] and it serves as a primary, high-affinity transporter for conjugated organic anions including conjugated bilirubin, [200] glutathione conjugates, and cysteinyl leukotrienes. [201] In addition, it transports other substrates including sulfated bile salts, conjugated estrogens, antibiotics, and other exogenous compounds. [202] MRP2 also transports unconjugated GSH from the hepatocyte into the bile, [130] albeit with a relatively low affinity. [203] However, because intracellular GSH concentrations are high, it is responsible for a significant fraction of canalicular GSH secretion. MRP2 is a 1545-amino acid protein coded by a gene located on chromosome 10q 23-q24. [204] It is a member of a family of closely related transport proteins that differ more in their tissue and membrane localization than in their transport function. [198], [199], [205] In humans, the MRP family consists of six members. MRP2 is the only MRP detected in the apical membrane, but MRP1 and MRP3 are present in the basolateral membranes, where they function as sinusoidal efflux pumps. [202] These become important during cholestasis (see later). MRP2 is specifically absent from the canalicular membrane in the human disease Dubin-Johnson syndrome, [206] as well as in two mutant rat strains, TR-/GY and EHBR. [207], [208] and [209] Although MRP2 is important in organic anion and bilirubin transport, it plays little role in bile formation. Patients with Dubin-Johnson syndrome have an almost complete absence of bilirubin conjugates and glutathione conjugates in bile, they develop a black pigment in their liver, but they are otherwise normal. They have an up-regulation of other MRP family members in the sinusoidal membrane, and as a consequence they are able to eliminate MRP substrates into the blood for eventual renal rather than biliary excretion. [210]

Multidrug Resistance Protein 3 MDR3 is an ABC transport protein that is expressed in high levels in the canalicular membrane of the hepatocyte, where it functions as a phospholipid transporter. [211] Several studies have demonstrated that MDR3 and the homologous protein in mice, mdr2, serve to translocate phosphatidylcholine (PC) from the inner to the outer leaflet of the lipid bilayer of the canalicular membrane. [212], [213] This process is a critical step in the mechanism by which phospholipid is secreted into bile (see later). Human MDR3 and its mouse homolog, mdr2, are 170-kd proteins and are the major ABC proteins of the canalicular membrane. [214], [215] and [216] They were initially identified by homology with the multidrug resistance P-glycoprotein, MDR1. This latter protein transports many cationic drugs, and its overexpression renders malignant cells resistant to many chemotherapy agents. [217] MDR3, a closely related molecule, fails to produce multidrug resistance as a consequence of overexpression. However, knock-out mice with disruption of the *mdr2* gene develop progressive hepatic fibrosis and bile duct destruction and have a complete absence of phospholipid secretion into bile. [218] A similar syndrome, progressive familial intrahepatic cholestasis type III (PFIC III; see later), also occurs in humans with mutations of the MDR3 protein. [219] Defects in this gene also contribute to gallstone susceptibility in association with cholestasis. [220], [221]

Multidrug Resistance Protein 1 MDR1 is a 1280-amino acid, 170-kd protein that is responsible for the secretion of a variety of large hydrophobic compounds, especially organic cations. [217] This transporter was first identified as a protein that becomes overexpressed in some cancer cells and consequently renders these cells resistant to a diverse array of chemotherapy agents. It was subsequently determined that the mechanism of this multidrug resistance is that MDR1 functions as a drug efflux pump with broad specificity for cationic chemotherapy agents. [222] The protein is normally expressed at the apical membranes of several epithelia but most notably in the canalicular membrane of hepatocytes, [214], [223] where it functions as an important mechanism for the secretion of drugs and organic cations into bile. [10], [224] To date, no specific endogenous substrate has been identified for either MDR1 or its mouse homologs mdr1a and mdr1b.

Biliary Lipid Secretion

Lipids make up an important component of bile. Biliary lipid secretion is necessary for maintenance of whole-body lipid homeostasis and prevention of intrahepatic bile acid toxicity. [13] The major lipid components of bile are bile salts, phospholipid primarily as PC, and cholesterol. The monomeric forms of phospholipid and cholesterol have limited aqueous solubility and exist in bile in the form of micelles and vesicles. The mechanism of biliary lipid secretion differs from that of soluble small molecules. The general process is one in which PC and cholesterol are inserted into the canalicular membrane and then are subsequently solubilized from the outer leaflet of the canalicular membrane into bile by a process dependent on bile acid. [12], [13], [225], [226]

Phospholipid secretion depends on several steps. First, cellular PC is inserted into the canalicular membrane. The primary process responsible for this is a membrane-membrane exchange process mediated by the PC transfer protein (PC-TP). [227], [228] and [229] This protein results in the selective addition of PC to the inner leaflet of the canalicular membrane. Flipping of PC from the inner leaflet to the outer leaflet of the membrane is accomplished by the MDR3 protein. [211], [230], [231] and [232] This step is critical, and the absence of a functional “flippase” in the canalicular membrane results in the complete absence of biliary phospholipids in both humans and mice. [218] [219] Phospholipid is ultimately removed from the outer leaflet of the canalicular membrane and appears in bile as small vesicles. This process is dependent on bile salts. Electron microscopic evidence suggests that it may involve direct budding of vesicles off the canalicular membrane, [225], [233] in addition to possible transfer from the canalicular membrane to biliary micelles. [234] Although bile acids are essential for biliary phospholipid secretion, the exact mechanism of this effect has been called into question by the observation that knock-out mice that fail to express BSEP have elevated levels of biliary phospholipid excretion. [197] This result suggests that intracellular bile acid, rather than secreted bile acid, is necessary for PC secretion.

Biliary cholesterol secretion also results from extraction from the canalicular membrane that depends on bile acid. In this case, secreted cholesterol appears to be derived from high-density lipoprotein and not from newly synthesized cholesterol. A hepatic high-density lipoprotein receptor, SR-BI, results in the uptake of cholesterol destined for biliary secretion, [235], [236] but the mechanisms by which cholesterol is inserted into the canalicular membrane and is ultimately solubilized into biliary micelles have not been determined. [226]

Mechanisms of Bile Ductular Secretion

After the process of fluid secretion into the canalicular space, the bile flows through the bile ductular system before its emptying into the duodenum. Far from a passive conduit, the biliary epithelium serves many functions, including secretion and absorption of fluid and electrolytes. [237] In addition, bile ducts are able specifically [52], [238] to absorb bile acids. [239] This serves both to detoxify bile and to amplify bile flow through cholehepatic shunting. [240] The biliary ductules comprise only

3% to 5% of the total liver cell population, but they can secrete up to 40% of total bile flow in humans. [241] Unlike the situation in hepatocytes, fluid secretion by the biliary epithelium is not the product of organic ion secretion. Rather, it is a process of Cl^--dependent HCO_3^- secretion similar to that which occurs in other salt transporting epithelia such as the airway epithelium. [242] Biliary secretion is under intense hormonal control, particularly by the secretagogue secretin, which increases intracellular cyclic AMP (cAMP) stimulating secretion of HCO_3^--rich fluid. Other hormones and neuropeptides such as acetylcholine, somatostatin, gastrin, bombesin, substance P, vasoactive intestinal polypeptide, and endothelin also modulate bile flow. [237], [243] Figure 16-5 diagrams the transport proteins responsible for bile ductular secretion.

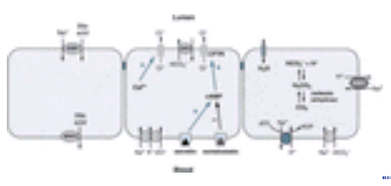

FIGURE 16-5. Bile duct epithelial cell transport. Transport of electrogenic fluid and HCO_3^- results from the involvement of multiple transporters. The $Na^+/K^+/2\ Cl^-$ cotransporter uses the energy of the Na^+ gradient to bring Cl^- into the cell at concentrations above its electrochemical equilibrium. Cl^- is secreted into the lumen primarily through the cAMP-dependent Cl^- channel, CFTR (cystic fibrosis transmembrane conductance regulator). It is exchanged for HCO_3^- by AE2 at the apical membrane. The process is primarily regulated by secretin, which increases cAMP levels in the cell. Bile acids can be absorbed from the lumen by the ASBT transport protein. A water channel, aquaporin-1, is present at the apical membrane. These processes are represented in different cells only for clarity of the figure. See text for further details.

The cholangiocytes are the epithelial cells that line the biliary ductules. They are polarized cells with transcellular transport resulting from the asymmetric localization of transport proteins. The basolateral membrane contains a Na^+,K^+-ATPase that establishes the ionic gradients and a K^+ channel that serves to maintain the intracellular negative potential. [244] An electroneutral $Na^+/K^+/2\ Cl^-$ cotransporter at the basolateral membrane is driven by the Na^+ gradient and results in the accumulation of intracellular Cl^- concentrations greater than that which would be expected by electrochemical equilibrium. [245] This is important because of the presence of the CFTR Cl^- channel at the apical membrane facing the ductular lumen. [246] Because Cl^- is above electrochemical equilibrium, a stimulus, such as secretin, that opens the apical membrane Cl^- channels will result in a process of Cl^- secretion into the lumen. [247], [248] A water channel, aquaporin-1, is present in the apical membrane and allows a water flux to be driven by osmotic forces. [249]

At the same time, the Na^+ gradient across the basolateral membrane drives two processes that tend to alkalinize the cytosol. These are electrogenic Na^+-HCO_3^- cotransport [250], [251] and Na^+/H^+ exchange. [252] The cycle is completed by an apical membrane Cl^-/HCO_3^- exchanger, AE2, which exchanges the secreted Cl^- in the lumen for intracellular HCO_3^-. [74] The net process is one of electrogenic HCO_3^- secretion. The entire system is maintained by the Na^+,K^+-ATPase, and the energy supplied by this transporter is transduced into net electrogenic HCO_3^- secretion from blood to bile, Cl^- recycling into and out of the cell across the apical membrane, Na^+ and K^+ recycling across the basolateral membrane, and passive Na^+ entry into bile through the tight junctions. This process of secretin-stimulated fluid secretion occurs primarily in the larger branches of the intrahepatic ducts, those with diameters greater than 15 µm, and not primarily in the smallest ducts with diameters less than 8 µM. [253] The CTFR is a critical component of this process. It is a cyclic AMP-dependent apical membrane Cl^- channel on which this process relies. Similar to the situation in respiratory epithelia, a defect in this Cl^-channel, as occurs in cystic fibrosis, reduces fluid secretion. [254] Under these circumstances, a Ca^{2+}-dependent Cl^- channel can substitute for CFTR and can allow some biliary secretion to occur. [255]

MECHANISMS OF REGULATION OF BILE FORMATION

A fundamental characteristic of the liver is that it serves multiple different functions and is able to adapt its functional activity to suit physiological and pathophysiological demands. The ability of the liver to function in the production of bile is thus quite variable and under some circumstances can be dramatically reduced. This reduction in the transport activity of hepatocytes and bile ducts manifests itself as cholestasis. Because bile formation is largely a consequence of the activity of organic ion transport proteins, the major mechanisms involved in regulation of bile flow are changes in the function, localization, and expression of these transport proteins. Other changes such as alterations in structure and function of tight junctions [256] and bile duct secretory processes [243] also play a role. This allows the liver to adapt to conditions that require changes in bile flow. The liver is able to modify its transport function by several mechanisms. In the short term (minutes to hours), changes in intracellular signaling result in modifications of transport proteins themselves, [257] as well as in redistribution of transport proteins between intracellular and plasma membrane pools. [179] In the longer term (hours to days), changes in expression of specific transport proteins play the more significant role. [258]

Alterations in Function of Hepatocellular Transporters

Signaling events within hepatocytes and biliary epithelial cells produce rapid changes in transport. [259] NTCP is subject to rapid regulation of its function on several levels. First, because it transports 2 Na^+ ions with each negatively charged bile acid, [99] the net transport is electrogenic and its rate is strongly influenced by changes in the intracellular negative membrane potential. [260] Hepatocytes normally have a membrane voltage of -30 to -40 mV, but this changes in pathological states, [66], [261], [262], [263], [264], [265] and [266] with a consequent increase or decrease in bile acid entry through NTCP.

In addition, NTCP itself is phosphorylated, and its phosphorylation state controls its distribution between intracellular and membrane sites. [267], [268] and [269] The phosphorylation state of NTCP is controlled by a balance of kinase and phosphatase activity and a cAMP-mediated increase in phosphatase activity reduces the phosphorylation state of NTCP. The dephosphorylated molecule is preferentially translocated to the sinusoidal membranes from an intracellular pool. [267]

Other important transport proteins can also be directly phosphorylated, but the importance of phosphorylation to regulation of transport is not clear. Protein kinase C (PKC) phosphorylation sites are present on MRP2, and activity of canalicular multispecific organic anion transport is modulated by PKC activators. [270] However, it is not known whether this effect results from direct effects on the protein or other changes such as membrane trafficking (see later). Oatp1 in rats has also been shown to be phosphorylated, [271] but the consequences of this on transport are not clear. There is additional evidence that phosphatidylinositol 3-kinase plays a role in transporter regulation. It regulates insertion of transporters into the canalicular membrane (see later), and its activation also increases the transport capacity of canalicular membrane vesicles. [272] The mechanism of this stimulation is unknown.

Alterations in Transporter Targeting

A major mechanism that produces rapid changes in the transport capacity of hepatocytes is redistribution of transport molecules between cellular sites. [179], [273], [274] Although this can occur for both the uptake transporters as well as the canalicular membrane efflux pumps, it is a more critical mechanism at the canalicular membrane, where transport capacity is rate limiting for overall bile flow. Redistribution of preformed canalicular transporters occurs rapidly in response to increased intracellular cAMP, [274] elevated intracellular bile acid concentrations, [275], [276] and osmotically induced changes in cell volume. [196], [277], [278]

Intracellular bile acid themselves increase the cellular capacity for maximal bile acid secretion. This process depends on the function of microtubules. [157] Electron microscopic studies of hepatocytes have shown that exposure of isolated hepatocytes to taurocholate induces an increase in the number of subcanalicular vesicles and a transformation of the subcanalicular network from primarily small vesicles to one of elongated tubules. [279] This process results in the insertion of BSEP-containing vesicles into the canalicular membrane [272] and an increase in the capacity of the canalicular membrane to transport bile acids. [280] The four major canalicular membrane transport proteins all actively shuttle between the canalicular membrane and an intracellular “subapical” vesicular pool. [179] Although the canalicular transport proteins undergo dynamic redistribution, the process is quite specific, and other canalicular membrane proteins remain in the canalicular

membrane while transporters are internalized. [179], [275]

Transporter cycling is dependent on the generation of lipid signalizing molecules though the action of phosphatidylinositol-3-kinase. [281] Transporter recruitment to the canalicular membrane is entirely blocked by inhibitors of phosphatidylinositol 3-kinase and the lipid products of phosphatidylinositol 3-kinase themselves directly stimulate transporter insertion. [179] As discussed earlier, phosphatidylinositol 3-kinase therefore plays a dual role in regulation of transport, stimulating transporter insertion and directly modulating transporter activity. The response to cell volume changes also appears to involve phosphatidylinositol 3-kinase. In addition, hepatocyte swelling activates several different members of the MAP kinase family including kinases of the Erk and p38MAPK type. [259]

Dynamic redistribution of canalicular membrane transporters between intracellular and canalicular membrane sites is important for liver function during physiological conditions. Hepatocytes normally undergo cycles of swelling and shrinkage in response to the solute load imposed by meals and subsequent uptake of amino acids and sugars. [282], [283] Because cell swelling causes insertion of MRP2 and BSEP into the canalicular membrane and cell shrinkage causes their internalization from the canalicular membrane, [259] this mechanism is likely to coordinate bile secretion rates with meals. Rapid redistribution of preformed transporters appears to be less important for the uptake transporters at the sinusoidal membrane. Although cAMP-dependent translocation occurs for NTCP, the uptake step is not rate limiting, and this may not play a major role in regulation of bile formation. Transporter translocation has not been shown to be important for regulation of the OATP family of transport proteins.

Regulation of Transporter Gene Expression

The primary mechanism by which the transport function of the liver is regulated is through changes in the expression levels of individual transport proteins. [257], [258] This mechanism plays an important role in the adaptation of the liver to inflammation [284], [285] and bile duct obstruction, [18] and it plays a dominant role in the development of liver transport function in ontogenesis. [26] The mechanisms that control expression of hepatic transport proteins are currently under active investigation, and although much information has been obtained, a complete understanding of how expression is coordinated under varying conditions has not yet emerged.

Transcription of the hepatic transporter genes, like all other cellular genes, is under the control of 5' upstream regulatory element sequences in the genes and the presence of activator proteins in the nucleus that bind to those elements. [286] Transporter mRNA transcription rates are rapidly altered by changes in the amount or function of these nuclear transcription factors. The nuclear localization and DNA binding ability of the transactivator proteins themselves are strongly regulated. This regulation can occur after transcriptional activation of the transcription factors themselves, after proteolysis steps that activate or inactivate factors, or after ligand binding, which creates an active factor complex. [286], [287] The transcription factors controlling transporter gene expression are multiple and interacting, and as a consequence environmental changes result in coordinated changes in expression. [258]

Investigators have demonstrated that bile acids themselves are the endogenous ligands for a particular nuclear receptor, the farnesoid X receptor (FXR). [50], [51], [288] FXR is a cytosolic transcription factor that binds bile acids with low micromolar affinity. [288], [289] On bile acid binding, FXR can complex with the retinoid X receptor (RXR), [290] to form an active transcription factor complex. [49] For genes with an FXR/RXR response element, such as the intestinal bile acid binding protein, [291] bile acids induce expression. Intracellular bile acids can also suppress transcription as for a key enzyme involved in bile acid synthesis, cholesterol 7a-hydroxylase. [292] In this case, bile acid binding to FXR results in formation of an FXR/RXR complex, which then competes for RE binding of another transcription factor (LXR/RXR) and decreases expression. [293] The extent to which this system controls hepatic transporter expression is not yet clear, but retinoid binding protein response elements are present in the NTCP promoter, [294] and they could be down-regulated by bile acids, similar to the situation with cholesterol 7a-hydroxylase.

Regulation of NTCP expression is critical because it controls bile acid entry into the hepatocytes. Its expression is sharply down-regulated in inflammation and other cholestatic situations, and this prevents bile acid–induced cytotoxicity (see later). Promoter elements that have been identified to control NTCP expression are the liver-specific transcription factors Hex [295] and HNF-1 and an uncharacterized footprint B binding protein (FpB BP). [296], [297] Expression is also under the control of signal transducer and activator of transcription 5 (STAT5), [298] and it involves the RXRa/RARa heterodimer. [294] Down-regulation of NTCP expression during inflammation can be explained in rats by an endotoxin-induced down-regulation of HNF-1 and FpB BP. [297] These important effects of endotoxin are likely mediated by cytokines. This is supported by the observation that interleukin-1ß (IL-1ß) down-regulates RXRa/RARa heterodimer activity and NCTP promoter activity. [294] In contrast, the hormone prolactin increases NTCP expression, and this results from a prolactin-induced phosphorylation and activation of STAT5. [298]

Details are less clear about the elements of promoter regulation of the other transporter proteins. Unlike NTCP, BSEP expression is only slightly decreased by whole-animal endotoxin treatment, bile duct ligation, or ethinyl estradiol treatment. [258] Transcription factors that bind to the promoter elements of the *BSEP* gene include C/EBP-ß, the hepatocyte-specific factor HNF-3ß, and the FXR/RXR heterodimer. Because this last transcription complex is directly activated by bile acids, it could potentially modulate coordination of BSEP expression with intracellular bile acid concentration. [258]

Regulation of OATP family expression is complex because of the multiple members of the family. In humans, promoter elements have only been identified for OATP-A, [299] although this family member is now known to be expressed primarily in brain. OATP-C and OATP8 are more significant transporters in liver. [86] OATP-A expression is stimulated by taurocholate in promoter constructs in HepG2 cells, and because OATP-mediated transport is bidirectional, this could enhance sinusoidal efflux of bile acids in cholestasis. Promoter element that have been identified include AP-1, AP-2, AP-3, HNF-1, HNF-3, C/EBP, and the glucocorticoid response element, GRE. [299]

Expression of MRP2, the canalicular organic anion transporter, is rapidly down-regulated in experimental models of cholestasis. [24], [300] This can in part be mediated by IL-1ß, which results in a decrease in the expression of RXRa/RARa heterodimers. [294] Promoter response elements identified in the human MRP2 promoter include AP-1, SP-1, HNF-1, HNF-3ß, and C/EBP. [301], [302] and [303]

Regulation of the canalicular phospholipid transporter, MDR3, has been extensively studied because of its role in disorders of lipid metabolism. [258] Its expression does not appear to be altered by endotoxin or during inflammation, but it can be strongly induced by feeding of hydrophobic bile acids, pharmacological inhibition of cholesterol synthesis, and induction of the transcription factor PPARa. [258], [304] Responsive elements in the 5' UTR of the gene include SP-1, AP-2, and C/EBP. [305]

Cytoskeleton and Tight Junction Changes

Alterations in the structure of the cytoskeleton and tight junctions occur in several conditions in which bile flow is reduced. Actin filaments serve as an intracellular scaffolding mechanism and are necessary for the targeted delivery of vesicles and the maintenance of cell polarity. A loss of the normal uniform actin filament distribution and a clumping of actin filaments around the bile canalicular membrane have been observed in multiple different models of cholestasis including bile duct obstruction and cholestatic bile acids and drugs. [306], [307] Vesicular transport along microtubules mediated by the molecular motor protein kinesin is also inhibited in cholestasis, [308] and these processes together alter transport function by changing the pattern of transport protein insertion and retrieval from the cell membranes.

The paracellular pathway, controlled by the proteins that make up the tight junctional complex, is also altered in cholestasis. [256] Both bile duct obstruction and estrogen-induced cholestasis produce changes in the distribution of the intracellular tight junction protein, ZO-1, [309] and a reduction in the number of claudin strands that compose the tight junctional seal. [310] These effects produce an increase in the permeability of the tight junctions to large molecules. [310] Alterations in the organization of tight junction proteins occur in human cholestatic diseases, [311] and they may contribute to the decrease in bile flow under these conditions.

Regulation of Bile Duct Secretion

Short-term regulation as a consequence of transport protein phosphorylation is the major mechanism of regulation of bile duct secretion. [237], [243] The rate-limiting step for biliary HCO_3^- secretion is the activity of the CFTR Cl^- channel at the apical membrane (see Fig. 16-5). Channel activity of CFTR is controlled by protein kinase A–mediated phosphorylation of its regulatory domain. [187], [312] In the biliary epithelium, a secretin-induced increase in intracellular cAMP is the primary mechanism that activates Cl^- channels and net secretion. [247] In addition, secretin also causes a microtuble-dependent insertion of aquaporin-1 water channels into the apical

membrane further enhancing fluid secretion. [249]

Other hormones and neuropeptides also play a role in regulation of ductular secretion, particularly in modulating the effects of secretin. Somatostatin interacts with somatostatin receptor subtype 2 on the bile duct epithelial cells and decreases cAMP and biliary secretion. [313] Gastrin has no effect on basal secretion, but it inhibits the secretin-induced choleresis. [314] Ductular fluid secretion is stimulated by bombesin, [315], [316] vasoactive intestinal polypeptide, [317], [318] and acetylcholine. [317], [318] It is inhibited by endothelin. [319]

An additional mechanism that regulates bile ductular secretion is the presence of micromolar concentrations of ATP in the ductular lumen. [320] In response to cell swelling and other stimuli, hepatocytes are able to release ATP into the canalicular space. [321], [322] This ATP interacts with purinergic P_{2u} receptors on the apical membrane of cholangiocytes that increase intracellular calcium, activate Cl^- channels, and stimulate secretion. [323], [324] In addition, ATP may be released by the cholangiocytes themselves resulting in a mechanism for both autocrine and paracrine regulation of bile duct function.

MECHANISMS OF CHOLESTASIS

Cholestasis is defined as the cessation or reduction of bile flow in a pathological situation. In many circumstances, this results from a coordinated response and is not merely a manifestation of nonspecific injury. The general principle of the response is that transcellular transport is diminished, but at the same time bile acid accumulation in the cell is minimized. This coordinated response provides for an alternative sinusoidal efflux pathway for toxins, conjugated bile acids, conjugated bilirubin, leukotrienes, and GSH. At the same time, biliary excretion of drugs is preserved and hepatocellular uptake of amino acids is increased, perhaps as a mechanism to support the massive protein synthesis required for the acute phase reaction. Other changes in liver function and structure also occur in many cholestatic settings. These include changes in cytoskeletal architecture, [307] alterations in tight junctional permeability, [310] and a decrease in the fluidity of the canalicular membrane. [325] In this section, we consider the specific changes that occur in several cholestatic situations. Numerous detailed reviews have appeared on this subject. [18], [21], [23], [24], [25] and [26], [284], [326], [327] and [328] Table 16-2 summarizes some of the changes in transporter activities that occur in acquired cholestasis syndromes.

TABLE 16-2 Changes in Transporter Gene Expression Associated with Cholestasis

Inflammatory Cholestasis

Cholestasis is frequently associated with inflammation, in particular that caused by gram-negative bacteremia. Inflammatory cholestasis is mediated primarily by cytokines, such as tumor necrosis factor-a, IL-1, and IL-6, that are released in response to bacterial lipopolysaccharide (LPS) stimulation of activated macrophages and Kupffer cells. The effect of these cytokines has been extensively studied in several experimental models. [329], [330] and [331] The best studied model is intraperitoneal LPS administration to rats. This treatment results in a dramatic decrease in bile flow and bile acid output that lasts several days. Within 3 hours of administration of LPS, Mrp2 redistributes from the canalicular membrane to a population of subapical vesicles. [332] This relocation results in reduced excretion of glutathione conjugates and bilirubin by Mrp2.

In addition to the rapid decrease in the Mrp2-mediated secretion, there is a slower onset of a decrease in bile acid transport. The amount of Ntcp present at the basolateral membrane is decreased, and this occurs primarily from reduced trans- cription. [330], [333] This is mediated by a reduction in transcription factors that bind to response elements in the Ntcp promoter region, specifically hepatocyte nuclear factor 1 (HNF-1 [333]), FpB BP, and the RXRa/RARa heterodimer. [294] These changes result in the impairment of Na^+-dependent bile acid uptake and thus a reduction in the concentration of bile acids within the hepatocyte. Expression of the canalicular bile acid transporter Bsep is relatively preserved compared with that of Ntcp, [334] but overall canalicular membrane excretion of bile acids from the hepatocytes is decreased. At the same time as biliary bile acid excretion is decreased, there is an up-regulation of expression of Mrp1 and Mrp3 at the basolateral membrane. [335] This provides an alternative sinusoidal efflux pathway for organic anions and is partially responsible for the appearance of conjugated bilirubin and bile acids in the plasma of patients with inflammatory cholestasis. Uptake of non–bile acid organic anions is not decreased to the same degree as bile acid uptake. Expression of Oatp1 in rat models is somewhat reduced, [285] but the functional consequences of this are unclear because a decrease in Na^+-independent bile salt uptake does not occur. [330], [333]

In humans, studies have been more limited. In a group of patients with inflammatory cholestasis, mostly patients with cholestatic alcoholic hepatitis, there is also a reduced expression of NTCP, OATP-C, and BSEP. In contrast to the situation in rodents, however, mRNA levels of MRP2 are preserved. [336] Despite these differences between the experimental rodent models and human disease, both situations are characterized by a decrease in transcellular bile acid transport. Bile acid uptake is decreased to a greater degree than canalicular efflux, and a sinusoidal efflux pathway for organic ions and bile salts is increased. This reduces the accumulation of bile acids within the hepatocytes and allows for entry of these compounds into the blood for eventual urinary excretion. The changes in gene expression are summarized in Table 16-2.

Bile Duct Obstruction

Obstructive cholestasis is a pathological condition resulting from obstruction in the extrahepatic bile duct as commonly results from choledocholithiasis or neoplasm. It has been extensively studied in the model of common bile duct ligation in rats. The inability of bile to enter the intestine results in an increase in bile acid concentrations in the bile duct lumen and within the hepatocytes, [337] as well as an increase in the pressure within the biliary tree. In response, expression of Ntcp and Oatp1 at the basolateral membrane is reduced, [338] limiting hepatocellular uptake of toxic bile acids. Short-term common bile duct ligation also results in relocation of Mrp2 from the apical membrane to a subapical vesicular compartment similar to that seen in inflammatory cholestasis. [339] There are also specific mechanisms that decrease canalicular efflux. Mrp2 expression [334] is dramatically decreased, [300] and this impairs secretion of sulfated bile acids as well as conjugated bilirubin. However, Bsep is reduced by only 20%, and therefore some efflux of bile acids into the biliary space continues. [334] This serves to protect hepatocytes from accumulating toxic levels of bile acids.

Several mechanisms further serve to prevent accumulation of toxic compounds intracellularly and in the bile duct lumen. Tight junctional structure is altered with a decrease in junctional strands and a marked increase in paracellular permeability that allows reflux of biliary components into blood. [310] As in inflammatory cholestasis, there is also up-regulation of Mrp1 and Mrp3 at the basolateral membrane of the hepatocytes. [335], [339], [340] and [341] This allows the efflux of bile acids into plasma, thus preventing further hepatocellular toxicity. In the bile duct, there is proliferation of cholangiocytes, [36], [342] as well as an increase in the expression of the ASBT at the apical membrane of the cholangiocyte. [343] This provides a mechanism for the removal of conjugated bile acids from the lumen of the bile duct. Furthermore, up-regulation of Mrp3 expression at the basolateral membrane of the cholangiocytes results in efflux of bile acids. [344] Finally, there is a marked increase in ductal HCO_3^- secretion resulting from up-regulation of secretin receptors. Therefore, although ductal obstruction prevents net bile flow, the compensatory mechanisms result in limitation of bile acid accumulation within hepatocytes and the duct lumen and profound dilation of the obstructed duct.

Estrogen-Induced Cholestasis

Estrogen-induced cholestasis is complex process resulting from changes in gene expression, direct inhibition of transport proteins, and other factors. It can occur with oral contraceptive use and can manifest as intrahepatic cholestasis of pregnancy. This syndrome is characterized by pruritus and cholestasis during the third trimester and accounts for about 20% of pregnancy-associated liver disorders. [345], [346] The administration of the synthetic estrogen ethinyl estradiol to male rats serves as an experimental model to study the pathogenesis of estrogen-induced cholestasis. In this model, there is a dramatic decrease in expression of Ntcp [334] associated with a

reduction in bile flow. Bsep expression is also reduced, but as in the other models of cholestasis, this effect is much smaller than that on Ntcp. [18] The result is impaired bile acid secretion. In addition, decreased expression of Oatp1 has also been observed, [347] which reduces the Na^+-independent uptake of bile acids and organic anions as well.

Conjugated estrogens themselves also are direct inhibitors of bile acid transporters. Ethinyl estradiol 17ß-glucuronide is a direct inhibitor of BSEP. However, it inhibits BSEP only when it is present in the canalicular space. It must therefore first be secreted before it produces its cholestatic effect. [348], [349], [350] and [351] Because it is transported into the canalicular space primarily by Mrp2, it is cholestatic in normal livers, but not in liver lacking expression of Mrp2 such as mutant rats or patients with Dubin-Johnson syndrome.

Lithocholic Acid–Induced Cholestasis

Lithocholic acid (LCA) is a monohydroxyl bile acid derived from bacterial 7a-dehydroxylation of chenodeoxycholic acid. LCA, together with its taurine and glycine conjugates, induces a cholestatic decrease in bile flow in animals and in perfused liver models. Several mechanisms have been postulated, but unlike the situation for other experimental models of cholestasis, there is not yet a detailed understanding of the effects of LCA on transporter expression and function.

LCA infusion produces a rapid cholestatic effect on the liver. It decreases canalicular excretion of both bile acids and organic anions, [352] it alters the distribution of cytoskeletal proteins, [325] it increases tight junction permeability and reflux from the liver into plasma, [352] and it reduces canalicular membrane fluidity by mobilizing cholesterol to the apical membrane. [353] This decreased membrane fluidity may contribute to the inhibition of canalicular transport. Like other highly hydrophobic bile acids, LCA increases intracellular Ca^{2+} and has specifically been shown to activate PKC. [354] These effects likely play a role in the cellular functional changes.

Although LCA is a known cause of cholestasis in experimental models, its significance in humans is unclear. LCA is found in minute amounts in human bile and serum. In human liver, in addition to its conjugation with taurine and glycine, LCA undergoes sulfation resulting in a decrease in hepatotoxicity and cholestasis. It has been shown that LCA, unlike other bile acids, is a high-affinity ligand for the nuclear receptor PXR. [355] LCA binding to PXR up-regulates the expression of the enzyme CYP3A11, which hydroxylates LCA and converts it to a less toxic derivative. In addition, LCA binding to PXR up-regulates expression of Oatp2. In rats, this would increase uptake of LCA, possibly allowing it to be more efficiently removed from blood and detoxified. [355]

Hereditary Cholestasis Syndromes

Several inherited defects in hepatic transport proteins result in clinically significant cholestatic liver diseases (Table 16-3). In addition, they provide enormous fundamental insights into the function of the transporters and the consequences of their failure.

TABLE 16-3 Summary of Hereditary Cholestasis Syndromes

Progressive familial intrahepatic cholestasis (PFIC), also known as Byler disease, is an autosomal recessive disorder that was first described in an Amish kindred. [356] It is now known that there are several distinct subtypes of this disorder, and molecular and genetic studies have identified the genes responsible for three of these. PFIC I is caused by a defect in a protein produced by the familial intrahepatic cholestasis 1 *(FIC1)* gene. This gene maps to chromosome 18q 21-22, and at least five mutations have been identified. [357] The *FIC1* gene codes a P-type ATPase that may be involved in the transport of aminophospholipids. It is expressed in the liver, small intestine, and pancreas, and a defect can cause cholestasis, diarrhea, or pancreatitis.

Benign recurrent intrahepatic cholestasis (BRIC) is a clinically milder variant of PFIC I. It is a recurrent cholestatic disorder of adults that was first described by Summerskill and Walshe in 1959. [358] Houwen and associates [359] mapped the genetic defect in BRIC to the *FIC1* gene. The process by which these mutations lead to cholestasis is not understood. Whereas PFIC I leads to progressive liver disease, BRIC is a milder, nonprogressive disease presenting either in childhood or adulthood. The reason for this variability in phenotypic expression is unknown.

PFIC II is a subtype of Byler disease that is clinically indistinguishable from PFIC I but is caused by a different genetic abnormality. The PFIC II locus has been mapped to the *BSEP* gene on chromosome 2q24 locus. [194] Immunostaining of the liver of these patients shows an absence of BSEP at the canalicular membrane. [191] These patients have a near-total absence of bile acids from the bile and consequently develop bile acid hepatotoxicity as a result of accumulation within the hepatocytes. The relatively severe phenotype and progressive liver failure resulting from absence of BSEP in PFIC II demonstrate the importance of this molecule in preventing the toxic effects of bile acid accumulation in hepatocytes. It is noteworthy, therefore, that BSEP expression is relatively preserved in most acquired cholestatic syndromes. Total loss of BSEP, as in PFIC II, results in severe liver toxicity.

PFIC III is a subtype that presents as a somewhat more aggressive disease, and unlike PFIC I and PFIC II, the affected patients have a high ?-glutamyltranspeptidase level. It is caused by a mutation of the *MDR3* gene on chromosome 7q21, [360] and it leads to a near absence of PC secretion into the bile. Because phospholipids are crucial for forming micelles with bile acids, the absence of biliary phospholipid increases free bile acid concentrations in the biliary tract. The consequences of this are injury and proliferation of bile ductules resulting in the release of ?-glutamyltranspeptidase. *MDR3* mutations are also associated with milder diseases that manifest in adulthood. Studies suggest that defects in MDR3 may be involved in hereditary susceptibility to cholelithiasis in association with cholestasis. [221]

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CHAPTER 17

Peter G. Traber

CARBOHYDRATE ASSIMILATION

Carbohydrates are the most abundant of the four major biomolecules, which also include proteins, nucleic acids, and lipids. Carbohydrates include sugars (mono- and disaccharides), starch, and fiber, and they constitute between 40% and 50% of the total daily calories as a source of fuel for humans. The ingestion and assimilation of carbohydrates should be viewed as the first step in carbohydrate homeostasis, which is crucial for the function of all organs. Carbohydrate absorption occurs along the entire length of the alimentary tract. After absorption, the liver, endocrine pancreas, muscle, and fat become the principal sites of carbohydrate homeostasis.

CHEMISTRY AND STRUCTURE OF CARBOHYDRATES

Carbohydrates, or saccharides, are composed of a series of aldehyde and ketone compounds that contain multiple hydroxyl groups. [1] Monosaccharides consist of a single aldehyde or ketone unit, and oligosaccharides contain two to ten monosaccharide units. The most common oligosaccharides available as human fuel sources are the disaccharides lactose and sucrose. Polysaccharides contain many monosaccharide units joined in long linear or branched chains. In biologic systems, carbohydrates serve as sources of fuel, energy stores, intermediates in metabolic pathways, and structural components of cells.

Monosaccharides have the molecular formula $(CH_2O)_n$. The simplest sugar, D-glyceraldehyde, has an n of 3 and one asymmetric carbon. Because of the stereoisomerism of asymmetric carbon atoms, the diversity of chemical structures increases with the addition of each successive carbon atom. The most important monosaccharides used as fuel sources are the hexoses D-glucose and D-galactose (Fig. 17-1A,B). Dihydroxyacetone is the three-carbon ketose that is the precursor of the most important six-carbon ketone sugar, D-fructose (Fig. 17-1C). The stable structure of hexose is as a six-member ring (i.e., pyranoses) that closes in either an a or ß form based on the position of the hydroxyl group on carbon 1 (see Fig. 17-1A,B). Fructose forms a five-membered ring called a *furanose* (see Fig. 17-1C). These ring structures form the building blocks of oligosaccharides and polysaccharides.

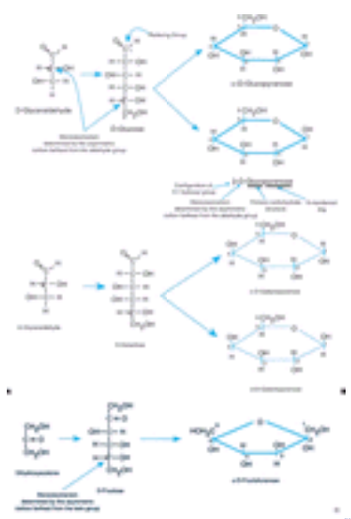

FIGURE 17-1. Carbohydrate structure. Linear and closed-ring forms of the chief nutritional carbohydrates. **A:** D-Glucose. **B:** D-Galactose. **C:** D-Fructose.

The chemical bonds that link monosaccharides to form oligosaccharides and polysaccharides are crucial to the biologic properties of the resultant molecules. Two D-glucose molecules linked by an a(1,4)-glycosidic bond form maltose, a readily digestible oligosaccharide (Fig. 17-2). If the D-glucose molecules are linked by a ß(1,4) linkage, the resultant molecule is cellobiose, the structural building block of cellulose, a compound that is indigestible by human enzymes. The structures of the two major dietary disaccharides, lactose and sucrose, also are shown in Figure 17-2.

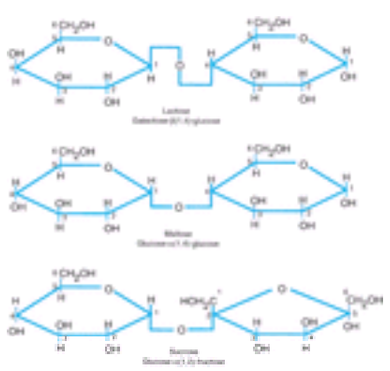

FIGURE 17-2. Structure of the three major nutritional disaccharides: lactose, maltose, and sucrose.

DIETARY CARBOHYDRATES

In Western cultures, 200 to 300 g of carbohydrates account for 40% to 50% of the total daily food energy ingested by humans. [2], [3] and [4] A survey conducted in 1985 showed that in the United States, carbohydrates, fats, and proteins accounted for 47%, 37%, and 16%, respectively, of the total daily ingested calories. [4] More recently, carbohydrate intakes in the United States have varied widely because both high- and low-carbohydrate dietary regimens are advocated. The sources of carbohydrates include complex forms, such as starch, and simple sugars, including disaccharides such as sucrose and lactose and monosaccharides such as glucose and fructose.

Except for lactose, which is found in milk, and small amounts of glycogen contained in meats, all carbohydrates are derived from plants. In fact, most of the glycogen in meat is depleted during the slaughter of the animal and therefore is not available as an energy source when ingested. [5] A number of ingested plant carbohydrates are not digestible by humans, including fiber and a variety of oligosaccharides. [6] These oligosaccharides contain three to eight linked monosaccharides and are ingested mainly as a-galactosides in legumes (e.g., raffinose and stachyose) and fructans (e.g., fructooligosaccharides), which are constituents of some vegetables, such as onions and Jerusalem artichokes.

Starch, a soluble polymeric molecule found in the cell walls of many plants, accounts for 40% to 60% of total ingested carbohydrates, [2] or about 22% of total calories. [4] The two forms of starch, a-amylose and amylopectin (Fig. 17-3), are present in many common foods, including grains (e.g., wheat, rice, barley) and vegetables. a-Amylose is a linear polymer of glucose joined by a series of a(1,4)-glycosidic bonds. Amylopectin is a branched form of a-amylose with a(1,6)-glycosidic linkages at the branch points, which occur every 15 to 25 molecules. [7] The ratio of a-amylose to amylopectin is variable, depending on the source of the starch. The physical form of starch in food is important in the process of digestion. [6] Most starch can be hydrated to form a gel, which gives particular foods their typical consistency. In raw plant foods, starches are enclosed in granular structures that are variably disrupted in food processing or cooking. Starch granules may be more or less disrupted with variable hydration of the starch. These physical characteristics of starch-containing foods have important implications for digestion and absorption.

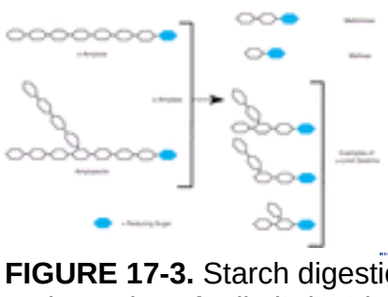

FIGURE 17-3. Starch digestion products. Amylopectin and a-amylose, the two molecular forms of starch, are digested by lumenal a-amylase into maltotriose, maltose, and a series of a-limit dextrins, a few examples of which are shown.

The percentage of simple sugars in Western diets has increased during the past several decades. [2], [4], [5] Sucrose is a natural ingredient of most fruits and vegetables. In addition, purified sucrose, used as table sugar and as a sweetener for many foods, is derived from certain plants that contain a high sucrose content, such as sugarcane, beets, and maple sap. The mean daily intake of sucrose per person in the United States is 41 g per day, or about 9% of the caloric intake and 30% of the carbohydrate intake. [2], [8] Of this ingested sucrose, 67% is added to foods as a sweetener. Lactose, the other main disaccharide, is the major carbohydrate in milk and serves as a food additive in many processed foods. The intake of lactose per person is highly variable depending on race and age, but it can account for up to 10% of the total carbohydrate calories consumed by the population in the United States.

The primary monosaccharides in foods are fructose and glucose. Fruits and vegetables contain various amounts of these two sugars as well as the disaccharide sucrose (Table 17-1). The average daily intake of free glucose per person has been estimated to be 20 g. [2] The main source of fructose in the diet is high-fructose corn syrup, which is added as a sweetener to foods because it is the sweetest of the sugars. [2] For this reason, the mean daily intake of fructose per person in the United States is 16 g; for some persons who ingest large amounts of foods sweetened with high-fructose corn syrup, fructose intake can be up to 150 g per day. [2], [8] The sugar alcohol sorbitol, or D-glucitol, is commonly found in many fruits; however, increasing amounts of sorbitol are being added to foods as a nonnutritive sweetener [2] and may cause gastrointestinal complaints in some persons because of its poor absorption by the small intestine. [9] Other polyols also are being used as food additives, including xylitol, mannitol, lactitol, and maltitol.

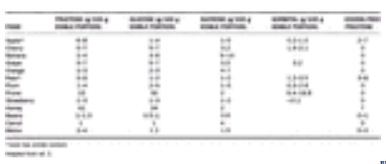

TABLE 17-1 Food Content of Fructose, Glucose, Sorbitol, and Sucrose

Based on the health risks of high-fat diets, it has been recommended that the percentage of carbohydrates in the U.S. diet be increased. [4] There is concern, however, about the type of carbohydrate that will account for the increment in intake. For example, an increase in the fructose intake may have deleterious health consequences, increasing hyperlipidemia and inducing other metabolic changes. [4] Similarly, the ratio of amylopectin to a-amylose may be important because the insulin response and postprandial serum glucose levels after a meal of amylopectin are significantly higher than those after a meal primarily composed of a-amylose. [4] Therefore, the carbohydrate composition of the diet may be of importance for the overall health of the population.

DIETARY FIBER

The typical human diet comprises multiple nondigestible carbohydrates called *fiber*. The amount of fiber in the diet varies widely with the region of the world and the regional dietary customs. Fiber is composed of nonstarch polysaccharides, which are integral structural components of plant cell walls. The wall that surrounds plant cells is a complex structure that differs in chemical composition among plants, in different portions of the same plant, and during developmental processes in the same plant. The highly variable cell wall properties of different plants lead to many varieties of fibers in nature. [10] The structure of many of the fibers in the human diet have not been studied extensively. Fiber can be categorized into soluble and nonsoluble forms, of which representative types are listed in Table 17-2.

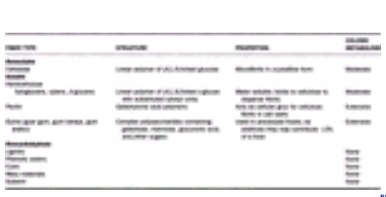

TABLE 17-2 Dietary Fiber

Dietary fiber has numerous effects on metabolism and digestive processes. [10], [11], [12] and [13] Unabsorbed fiber reaching the colon serves as a fuel for colonic bacteria. [13] The degree to which fiber is metabolized by bacteria varies with the type of fiber (see Table 17-2). The bacterial products of fiber metabolism include short-chain fatty acids, hydrogen, and methane. [13], [14] and [15] Short-chain fatty acids serve as a fuel for colonocytes. [16], [17] The fiber content of food affects the rate of intestinal absorption of carbohydrates by several mechanisms. One mechanism is to slow gastric emptying based on the viscosity of the gastric contents. Another is to impede digestion in the intestinal lumen by decreasing the accessibility of starch to enzymes. [10] It is known that the ability of fiber to complex with organic compounds may affect the absorption of certain drugs. [10] Other effects include changes in sterol metabolism [10] and an increase in stool weight. [10], [18]

Evidence suggests that dietary fiber may have a beneficial effect on a number of disease processes, [12], [19] including lipid disorders and colorectal cancer. Studies have shown that water-soluble fiber in the diet reduces low-density lipoprotein levels by about 10%, but it has no effect on the levels of high-density lipoproteins and triglycerides. [20] Based on the results of epidemiologic studies, it has been concluded that an inverse relationship exists between the amount of ingested fiber and the incidence of colorectal cancer. [21], [22], [23], [24], [25] and [26] Cautionary notes have been sounded on the interpretation of the results of these studies because of the possible variance in dietary components and socioeconomic parameters among populations. [23], [27] Although the mechanism of the effect of fiber on the incidence of colorectal cancer is unknown, it is possible that the colonic metabolites of fiber, particularly short-chain fatty acids (e.g., butyrate), are involved in growth regulation. [24], [28]

CARBOHYDRATE ASSIMILATION

Organs throughout the alimentary tract have important roles in the digestion and assimilation of ingested carbohydrates. The most important processes include the enzymatic hydrolysis of starch in the lumen of the intestine, the hydrolysis of oligosaccharides and disaccharides by enzymes located in the apical membrane of enterocytes, and the transport of monosaccharides across the apical membrane of enterocytes. [6], [29] These processes are described in detail in the following sections, and the overall efficiency of the process and the rate-limiting steps for the assimilation of different types of carbohydrates are discussed.

Several ancillary processes have a small overall effect on carbohydrate assimilation. Effective mastication of certain vegetable products is important for lumenal hydrolysis of the starch, which may be located within a layer of waxy cellulose. There is some evidence that the motility of the stomach and small intestine may play a small role in absorption. [30], [31] Overall starch absorption increases if the transit time through the small intestine is slowed by pharmacological means, [30] whereas absorption is diminished if transit is hastened. [30], [31]

LUMENAL PHASE OF DIGESTION STARCH HYDROLYSIS

Initial digestion of starch occurs in the lumen of the alimentary tract and is catalyzed by a-amylase. [31], [32], [33] and [34] This enzyme has a high level of activity for the cleavage of internal a(1,4)-glycosidic linkages. [32] It has no activity for the hydrolysis of a(1,6) linkages, terminal glucose residues, or a(1,4) bonds adjacent to a(1,6) linkages. [32] Lumenal hydrolysis of a-amylose produces maltose and maltotriose, and hydrolysis of amylopectin produces maltose, maltotriose, and a-limit dextrins, which are a series of oligosaccharides containing four or more glucose molecules having an a(1,6) linkage (see Fig. 17-3). [7], [32], [33] In the aqueous environment of the gastrointestinal lumen, the process of starch hydrolysis is efficient and is completed in the proximal jejunum [33], [35]; however, some important characteristics of solid food particles limit starch hydrolysis and allow a percentage of starch to enter the colon undigested.

The a-amylase protein is synthesized and secreted into the alimentary tract by the parotid gland and the pancreas. Salivary a-amylase, which exhibits optimal activity within a narrow pH range of 6.6 to 6.8, initiates starch digestion but is of only minor significance because it is rapidly inactivated in the acid pH environment of the stomach. Short-chain glucose polymers in the diet may stabilize the enzyme and allow maintenance of activity at acid pH. [36] This stabilization phenomenon may help to ensure amylase activity for carbohydrate assimilation in neonates and in patients who have chronic pancreatic insufficiency. [3] In healthy persons, the survival of amylase in the proximal intestine is increased by the presence of nutrients in the lumen. [37] Pancreatic amylase is secreted into the lumen in quantities far greater than are required for the hydrolysis of starch. The completeness of starch hydrolysis is more dependent on the state of the ingested starch than on the amount of pancreatic amylase secreted. For example, the physicochemical properties of different varieties of rice [38] and the preparation of other cereal foods [39] affect the ability of amylase to hydrolyze the starch component of the foods. Amylase concentration becomes limiting for starch hydrolysis only in severe cases of pancreatic insufficiency, in which lumenal amylase activity levels are reduced to below 10% of normal (see Chapter 95). [40] Human milk contains amylase activity, which may be important for carbohydrate digestion in infants. [41]

Amylase proteins are encoded by a clustered gene family located on human chromosome 1 and mouse chromosome 3. [42] In humans, the AMY1 gene is expressed in the parotid gland, and the AMY2 gene is expressed in the pancreas. [43] Purified preparations of the two isoenzymes exhibit identical activities at neutral pH, but their molecular weights differ, as do other biochemical properties. [44] The sequences of the pancreatic and salivary cDNAs are 94% similar, encoding for polypeptides with the same number of amino acids. [45] For additional information on the structure, synthesis, and secretion of amylase, see Chapter 15.

BRUSH BORDER CARBOHYDRASES

Brush border saccharidases are ectoenzymes that are anchored to the apical membrane of enterocytes. These enzymes are responsible for the further breakdown of lumenal hydrolysis products (i.e., maltose, trimaltose, and a-limit dextrins) and ingested disaccharidases. The result of metabolism by these enzymes is the production of monosaccharides that can be transported across the apical membrane. Table 17-3 outlines the characteristics of each enzyme.

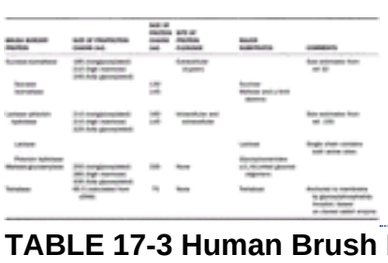

TABLE 17-3 Human Brush Border Proteins with Carbohydrase Activity

Sucrase-Isomaltase

Sucrase-isomaltase (SI) has been one of the most intensively studied membrane proteins; consequently, an enormous amount of literature has accumulated concerning its function, synthesis, processing, sorting, and regulation.

Enzyme Activity SI, a bifunctional disaccharidase with two active sites, constitutes about 10% of the total brush border protein mass. [46] The isomaltase active site hydrolyzes maltose at the a(1,4)-glycosidic linkage and a-limit dextrins at the a(1,6) linkage. [47] Although SI does hydrolyze isomaltose, this natural substrate is not abundant; therefore, the name *a-limit dextrinase* has been suggested in place of *isomaltase*. [48] The sucrase active site cleaves sucrose to glucose and fructose and appears to be the only enzyme in the intestinal tract that can perform this function. The enzyme accounts for 100%, 90%, and 80% of the sucrase, isomaltase, and maltase activity, respectively, in the intestine. [46] One group of investigators has reported that SI is a cAMP-dependent chloride channel on the basis of current inhibition in the presence of antibodies. This intriguing functional characteristic of SI must be confirmed with further research. [49]
Protein Structure The primary structure of the SI molecule was determined by cloning of the complete rabbit, [50] human, [51] and rat [52] cDNAs. The coding sequence of the human SI mRNA is 5481 bases long and encodes a 1827-amino acid polypeptide. [51] The deduced molecular weight of this polypeptide correlates well with the measured molecular weight of the nonglycosylated human SI protein of 185 kd. [53] The nucleotide sequence of rabbit cDNA has 83% overall identity to the human sequence [50], [51] and a similar level of identity in the cloned regions of rat [54] and mouse [55] cDNAs. In both rabbit and human cDNAs, the isomaltase and sucrase

subunits have a high degree of sequence identity. In fact, 13 of 15 amino acids surrounding the aspartate residues at both active sites are conserved, [50], [53] which has led to the hypothesis that the two subunits were formed by a gene duplication event. [47], [50], [56] In addition, SI may be a member of a larger gene family [57], [58] with homology to lysosomal glucosidase. [58] The primary structure of SI predicts a molecular model that can be represented by five different domains (Fig. 17-4). [47], [50], [56] The amino terminus of the molecule forms a short intracytoplasmic domain containing about 12 amino acids. A transmembrane domain of highly hydrophobic amino acids that predicts an a-helical structure serves as the membrane anchor for the molecule; the length of this segment indicates that it can cross the membrane only once. The first portion of the extracellular domain forms a highly hydrophilic segment, or stalk, of about 180 amino acids, including multiple serine and threonine residues, which suggests a high degree of glycosylation. [46], [47], [50], [56] The length of this stalk differs somewhat among species, with that of the rat being longer than that of the rabbit and human. [59] The significance of this finding is unknown. The isomaltase subunit is next, followed by the sucrase subunit, which in the completely processed enzyme is linked to the isomaltase subunit by noncovalent ionic bonding.

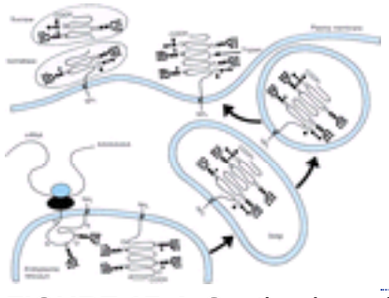

FIGURE 17-4. Synthesis and processing of sucrase-isomaltase. The processing of sucrase-isomaltase, from synthesis of the polypeptide chain on ribosomes to insertion of the protein in the apical membrane.

Synthesis and Processing SI is encoded by a single gene in the human, [59], [60] rat, [59] and mouse. [55] The human gene, which is located on chromosome 3 at locus 3q25-26, [61] is estimated to be about 55 kb in size. [51] As for many eukaryotic genes, the 5'-flanking region of the SI gene has a number of DNA regulatory regions that control initiation of gene transcription. [62], [63] Transgenic mouse experiments show that SI gene expression in the mouse intestine is regulated by many functional *cis*-acting DNA elements. [64], [65] and [66] The elements necessary to direct intestinal epithelial cell–specific expression are embodied in a 201-nucleotide, evolutionarily conserved, 5'-flanking region of the gene. [66] This mouse SI gene promoter directs expression of a reporter transgene to enterocytes in the proper developmental- and differentiation-dependent patterns. Studies with transgenes containing longer segments of the 5'-flanking region of the gene showed that other elements are important for modulating the expression of the SI gene. [65] DNA regulatory elements and associated DNA binding proteins within the evolutionarily conserved SI promoter region have been assessed. [55], [67], [70] At least two types of transcriptional proteins are involved in SI promoter transcription, including hepatocyte nuclear factor 1 (HNF-1), [67], [68] and caudal-related homeodomain proteins (Cdx). [70] The requirement for the HNF-1 binding site has been confirmed in transgenic mice, and the activation of transcription is modulated by the ratio of HNF-1a to HNF-1ß. [68] There is some evidence that GATA factors facilitate maximal transcriptional activation of the SI promoter, [69] although the mechanism remains poorly defined. The interaction of tissue-specific, tissue-restricted, and ubiquitous transcription factors facilitates the transcription of genes in a single cell type. It is not understood how these transcription factors interact with each other or with coactivator proteins to direct the complex patterns of SI gene transcription. Following synthesis and processing, the mature mRNA for SI is translated in the cytoplasm to yield a single protein of the predicted size (see Fig. 17-4). [47], [53] The amino terminus forms a short-membrane anchor that remains embedded in the endoplasmic reticulum (ER) membrane. [47], [71] The amino-terminal intracellular tail of the SI protein is phosphorylated by cAMP-dependent protein kinase, which suggests that phosphorylation affects the physiological function of the protein. [72] The SI protein is used as a model for examining the posttranslational processing of membrane proteins. Glycosylation of proteins is performed by multiple glycosylases present in the ER and Golgi apparatus. [73] Protein *N*-glycosylation and the addition of a branched oligosaccharide to an asparagine residue occur during translation of the protein in the ER. Because this oligosaccharide contains nine mannose molecules, proteins with this modification have been called *high-mannose forms*. This common precursor oligosaccharide may be modified in the Golgi apparatus by the removal of mannose residues and the addition of other sugars. The secondary structure of the protein determines which asparagine-linked oligosaccharides are further modified. *O*-glycosylation occurs at the hydroxyl-group oxygen of serine and threonine residues in the Golgi apparatus. The fully glycosylated SI molecule has a molecular weight of 245 kd, and both *N*- and *O*-glycosylation occur equally on the isomaltase and sucrase subunits. [53] Although these data apply to the human protein, other species show essentially the same pattern of synthesis, with slight differences in the molecular weights of the forms. [71], [74], [75] and [76] The fully glycosylated SI molecule is transported uncleaved to the apical membrane of the enterocyte. Electron microscopic studies suggest that the transport vesicles move from the Golgi apparatus directly to the apical membrane. [77] Cleavage of the molecules into the two subunits (145 kd for isomaltase and 130 kd for sucrase) occurs by tryptic digestion in the lumen of the human intestine. [46], [47], [53] The two subunits remain associated by hydrostatic bonds. In studies using electron microscopy of reconstituted membranes, it was shown that SI exists as a dimer in the membrane. [78] The degradation of SI in the lumen of the intestine is mediated by pancreatic proteases. [79], [80] Understanding of the processing and transport of the SI molecule has been instrumental in elucidating the site of the defect in inherited SI deficiency in humans. [81] The enzymatic deficiency in these persons appears to be related to a number of different defects in glycosylation and intracellular transport, and it may also involve catalytic activity. [81], [82]

Regulation

Spatial and regional regulation. The expression of genes in the intestine exhibits a complex spatial pattern along the vertical (crypt-to-villus) and horizontal (proximal-to-distal) axes. [83] By using sections of frozen tissue to sample villus and crypt cells sequentially, it was demonstrated that there is little sucrase enzyme activity in the crypt, maximal activity in the lower and mid villus, and decreased levels in the villus tip cells. [84] This pattern of enzyme activity is consistent with the localization of SI protein expression that was determined when antibodies became available to study the pattern of SI protein expression by immunohistochemistry. [77] In the mouse, SI protein in the intestinal mucosa is not detectable in crypt cells, but it is expressed from the crypt-villus junction to the villus tip, with a minimal decrease in intensity at the tips. [64] This pattern of expression is generally accepted, although some data derived from monoclonal antibody studies show that SI protein is expressed to some degree in the crypts of the human small intestine. [85] SI mRNA is also first expressed in the upper region of the crypt near the crypt-villus junction. [59], [64], [86] Within only a few cell positions, the levels of SI mRNA are maximal and remain at a similar level about two thirds of the way up the villus; levels decrease toward the villus tip. These data suggest that the major mechanism for regulating the expression of the SI protein along the crypt-villus axis is the steady-state level of SI mRNA; however, posttranscriptional and posttranslational regulation likely plays a role in the expression of functional SI protein along the intestinal crypt-villus axis, [85], [87] and in colon carcinoma cell lines that undergo partial enterocyte differentiation. [88] Experiments in transgenic mice suggest that the complex patterns of SI mRNA expression along the crypt-villus axis most likely result from events associated with transcriptional regulation of the gene. [64], [65] and [66] Many functional differences exist between the jejunum (proximal segment) and the ileum (distal segment) that reflect differences in the expression of different genes, or gradients of gene expression, along the proximal-distal axis of the intestine. For example, SI activity is four- to fivefold greater in the jejunum than in the ileum [87]; however, sucrase mRNA appears to be similar in the two regions. [64], [89] The pattern of glycosylation in the Golgi apparatus differs slightly between the two regions, but the resultant protein levels seem to be unaffected. [87], [89] The major difference in regulation of SI expression between the jejunum and ileum appears to be at the level of mRNA translation. [89]

Developmental regulation. SI has served as a model to examine developmental processes in the intestine because its expression mirrors many of the developmental transitions that occur in the establishment of the adult intestine. [66], [90], [91], [92] and [93] In the human small intestine, SI is expressed between weeks 8 and 14 of gestation, and high-level expression persists through birth and into adulthood. [90] The mechanism for the induction of SI in human small intestine appears to be at the level of mRNA. [90], [94] In rats, mice, and rabbits, the maturation of the small intestine is completed in the neonatal period. [91] At birth, SI enzymatic activity is not detectable, and dramatic induction occurs at the time of weaning. [95], [96] Evidence suggests that activation of the SI gene occurs in two stages. [66] SI mRNA is first detectable at low levels in the mouse fetus after the transition of the endoderm into a simple polarized epithelium. [66] Low levels of SI mRNA expression are maintained until the suckling-weaning transition that occurs in the third week of life, when a dramatic induction of SI mRNA and protein takes place. [66] Transgenic mouse experiments have shown that a short, evolutionarily conserved SI promoter [66] is sufficient to direct the induction of expression at the suckling-weaning transition. These data indicate that transcriptional regulation is responsible for the patterns of expression during development. Moreover, the two stages of SI mRNA expression suggest that epithelial cells can transcribe the SI gene late in fetal development, but missing factors come into play at weaning to allow full expression of the gene. The dramatic induction of SI in the rodent intestine appears to be a genetically programmed event, largely unaffected by the type of ingested food. [91], [92] and [93], [97] Stress in suckling pups or the administration of corticosteroids or thyroid hormone results in premature induction of SI before the normal time of weaning. [91], [92] and [93], [97] It appears that the normal induction of SI at the suckling-weaning transition and the precocious induction of SI by corticosteroids occur through separate molecular pathways. [66], [98] Studies in rodents have shown that differences in corticosteroid regimens significantly affect the response of SI in the developing intestine, [99] which may explain conflicting results obtained in human studies. SI expression in the colon of humans and rodents is transient. [100], [101] and [102] In humans, SI mRNA, protein, and enzymatic activity are found in the colon between weeks 12 and 28 of gestation, concurrent with the histological appearance of villus and crypt structures

resembling those of the small intestine. Expression of the SI gene and these histological features are subsequently lost after gestational week 28 and are absent in the newborn colon. [101] Although this matter remains controversial, [103], [104], [105], [106] and [107] results of studies using the reverse transcriptase-polymerase chain reaction suggest that SI mRNA is not expressed in adult colon. [105] Importantly, SI is expressed in neoplastic tissue from the colon, including adenomas and adenocarcinomas, [103], [104], [105] and [106], [108] which may represent a reversion to a fetal type of regulation of the SI gene. A similar pattern of developmental expression is found in the rodent colon, although the process occurs in the postnatal animal, as it does in small intestine maturation. [109]

Nutritional regulation. Changes in diet have a marked effect on the expression of SI. [87] Starvation leads to a rapid decline in brush border proteins and SI activity. [110], [111], [112], [113] and [114] This decline in SI activity is restored rapidly after refeeding. The type of ingested carbohydrate is important for the regulation of SI expression. Although both starch and sucrose induce SI activity, sucrose is a more potent inducer. [87], [115], [116], [117] and [118] The mechanism of induction of SI in rats appears to be at the level of mRNA. [115] There is evidence that feeding rats fructose, but not glucose, increases transcription of the SI gene and alters the binding of proteins to the promoter. [119] Study of the intestinal cell line Caco-2 has shown that a short promoter region of the human SI gene (nucleotides -370 to +30) can down-regulate SI transcription in the presence of glucose. [120] Although this effect may be related to the neoplastic origin of the cell line, it underscores the complex physiological regulation of SI gene transcription in response to glucose. There is also a circadian pattern to SI activity that is related to meal ingestion; the pattern can be shifted by altering the time of feeding. [87] The short-term regulation of the meal-related diurnal differences is not controlled by food intake, but the long-term regulation of this phenomenon is clearly related to feeding. [87] Research has also shown that glucagon-like peptide 2 can increase the expression of SI in the intact intestine. [121]

Pathophysiological regulation. The levels of SI and other saccharidases may decrease with infection and inflammation. [122] In some cases, a decline in enzyme activity leads to malabsorption of carbohydrates and symptoms of diarrhea, flatulence, and weight loss. In most disease processes, however, the diminished levels of SI are associated with global dysfunction of the small intestine mucosa with abnormal absorption of other nutrients, abnormal absorption of water, and abnormal electrolyte absorption and secretion. Cytokines may mediate the inflammation-related inhibition of SI expression in intestinal epithelial cells in vivo and in intestinal cell lines. [123] Experiments in cell lines suggest that interleukin-6 and interferon-? specifically inhibit SI gene expression. [123] Although most diseases produce a decline in SI activity, diabetes mellitus results in a striking up-regulation of SI activity. [87] This increased function of the small intestine mucosa is not limited to SI activity; monosaccharide transport, amino acid transport, and mucosal hypertrophy also are increased. [87], [124], [125], [126], [127], [128] and [129] The mechanism of SI induction in experimental diabetes in rats appears to be multifactorial. There may be changes in protein degradation, [126] an increase in SI mRNA, [87] and a greater number of SI-expressing cells along the crypt-villus axis. [130] The increase in the absorptive capacity of the small intestine for sugars may be of importance in the pathogenesis of complications associated with diabetes. Administration of acarbose, a drug that inhibits SI, decreases postprandial glucose levels and hemoglobin A_{1c} levels in experimental models of diabetes and in humans with diabetes. [131] Furthermore, diabetic rats given acarbose had less severe diabetic neuropathy than did control animals. [132]

Lactase–Phlorizin Hydrolase

One of the defining characteristics of mammals is that neonates are fed milk produced by their mothers. The major carbohydrate in milk is the disaccharide lactose, which is produced exclusively in the mammary glands by lactose synthase. Brush border lactase is the only enzyme that cleaves lactose into glucose and galactose and is therefore essential for mammalian survival early in life. Lactase has other enzymatic activities, including phlorizin hydrolase, glycosylceramidase, and ß-galactosidase activity. [133] It has been suggested that glycosylceramidase activity may play a role in the digestion of glycolipids. [133], [134]

Structure and Synthesis Lactase–phlorizin hydrolase (LPH) cDNAs and genes from a number of species have been cloned. The human LPH structural gene is about 55 kb long with 17 exons [135] and is located on the long arm of chromosome 2. [136] The human LPH cDNA encodes a single polypeptide chain containing 1972 amino acids. [137] Various lengths of the 5'-flanking regions of the human, [135] pig, [138] and rat [139], [140] LPH genes have been characterized. The LPH promoter is well conserved across species within the first 150 nucleotides from the start of transcription. [138] Nucleotides -1000 to -24 of the pig LPH gene [141] and nucleotides -2038 to -15 of the rat LPH gene [140] are able to direct transcription to the intestinal epithelium in transgenic mice. Studies in intestinal cell lines have identified functional DNA elements in the LPH promoter that interact with nuclear transcription factors. [138], [142], [143], [144], [145], [146] and [147] Both Cdx and HNF-1 proteins bind to promoter elements similar to the SI promoter element, [68], [142], [143], [145], [146] and it appears that these two proteins interact. [143] A single Cdx2 molecule binds to the LPH promoter, whereas the SI promoter binds two Cdx molecules at two closely linked sites. [142] Single Cdx-binding elements are also active regulatory units in the transcription of other intestinal gene promoters. [148] Members of the GATA-type zinc-finger transcription factor family have emerged as important activators of the LPH promoter [68], [145], [146] and appear to interact with HNF-1 in the transcriptional activation of LPH. [68] It is of great interest that transcription factors similar to those that regulate the SI gene promoter may also regulate the LPH gene despite very different patterns of regulation. Analysis of the primary amino acid sequence and multiple biochemical experiments using native LPH protein and cDNA-derived protein from transfected cells have provided a clear picture of the processing and structure of the mature enzyme. Although both LPH and SI are anchored to the apical membrane and have two active sites on the lumenal side of the membrane (Fig. 17-5), their structures differ markedly. The carboxyl-terminal end of the LPH polypeptide is embedded in the plasma membrane, leaving the amino-terminal end in the lumen. The 19 amino-terminal amino acids of the propeptide are cleaved in the ER, leaving the amino-terminal end of the molecule free in the interior of the ER. After synthesis of the polypeptide, the carboxyl-terminal end remains embedded in the membrane and serves as an anchor. This anchor consists of a cytoplasmic domain containing 26 amino acids and a membrane-spanning region containing 19 hydrophobic amino acids. [137] Correct folding of pro-LPH in the ER is dependent on intramolecular disulfide bonds. [149] The prepolypeptide has a fourfold symmetry, and the mature lactase has a twofold symmetry, suggesting that the two domains may harbor the active sites of the enzyme. [137] This is similar to the symmetry of the SI molecule, although the repeated units in LPH are not cleaved. The active sites in the mature protein have been identified: the lactase site at Glu1271 and the phlorizin hydrolyase site at Glu1747. [150]

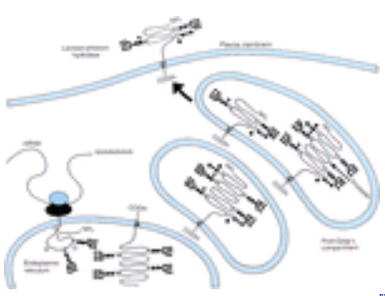

FIGURE 17-5. Synthesis and processing of human lactase–phlorizin hydrolase.

In humans, a single polypeptide precursor of about 210 kd is synthesized from a single mRNA (see Fig. 18-5). [151] The precursor is cotranslationally *N*-glycosylated in the ER and then transported to the Golgi apparatus, where some of the *N*-glycosylated groups are modified and *O*-glycosylation occurs, [151], [152] resulting in a 215-kd high-mannose form and a 225-kd complex glycosylated form of LPH. The glycosylation process is apparently not invariant because the final plasma membrane enzyme exists in two forms, one with *N*-glycosylation only and another with both *N*- and *O*-glycosylation. [152] The latter form is more active enzymatically. [152] The mature LPH enzyme in the brush border represents a cleavage product of the glycosylated precursor molecule. [47], [133], [152], [153] The first cleavage occurs intracellularly at a dibasic residue (Arg734-Leu735), resulting in a 160-kd intermediate form of LPH. [154], [155] The cleaved polypeptide is structurally similar to the mature enzyme, but it is devoid of enzymatic activity. [156] The human enzyme is cleaved before the appearance of the protein on the plasma membrane [151] in a post-Golgi compartment [157] after complex glycosylation. [152], [158] Proteolytic cleavage is not required for enzymatic activity in either polarized [159] or nonpolarized [160] cells, nor for proper sorting to the apical membrane. [159] The human LPH enzyme is cleaved at a second site by extracellular trypsin (Arg868-Ala869) after it is in the brush border membrane, to yield the mature 145-kd form. [154] In contrast to the cleavage of human LPH, that of rat LPH appears to occur on the plasma membrane in several steps. [133], [161], [162]

Regulation Regional differences in the expression of LPH occur along the vertical and horizontal axes of the intestine. In situ hybridization and immunohistochemistry of small intestine tissue from neonatal rats show a complex pattern of expression of LPH mRNA along the crypt-villus axis. [163] Prenatally, LPH mRNA and protein are expressed in all villus-associated enterocytes. [163] Immediately after birth, the LPH mRNA is restricted to enterocytes located at the base of villi, in a fashion similar to the pattern of expression of SI mRNA in the adult intestine. The protein, however, persists throughout all villus enterocytes, suggesting some posttranscriptional regulation of the maturation process. [163] The regulation along the horizontal intestinal axis is complex, involving pretranslational and translational regulation in the proximal and distal intestine. [164], [165] and [166] The developmental regulation of LPH has been studied extensively. [47], [93], [97], [133] In humans, LPH is expressed in fetal small intestine just after the onset of expression of SI. [90], [100] This expression is maintained throughout development and during childhood. LPH is expressed in the neonatal rat colon [101], [167] in the same manner as SI. At some time during childhood or adolescence, LPH activity declines to 5% to 10% of childhood levels in most populations worldwide. [47], [168] The age at which the enzyme disappears varies, depending on the heritage of the individual. Ingestion of milk or milk products by

persons with diminished LPH activity leads to flatulence, abdominal cramps, and diarrhea. [168] In selected human populations, such as those in northern Europe, where dairy cattle have been developed as a continuing source of milk products, intestinal LPH activity persists throughout adulthood. [47], [93], [97], [133] This phenotype is inherited as an autosomal recessive trait, and evidence of intermediate levels of LPH activity has been found in heterozygous persons. [47] Therefore, the aberrant allele in the human population is considered to be the one that leads to persistence of the enzyme, not the deficiency. Support is given to this concept by the fact that in nearly all other species of mammals, LPH activity is lost after weaning. [47], [93], [97], [133] Thus, LPH activity declines at the same time that SI activity increases in the small intestine. [47], [91], [92] The decline in LPH activity is hastened by pharmacological treatment with corticosteroids in parallel with the induction of SI. Hypolactasia in humans appears to involve transcriptional and posttranscriptional mechanisms. [166], [169], [170] and [171] The mechanisms involved in the postweaning decline of LPH activity in many animal species remain uncertain. The clinical syndrome and the mechanisms for hypolactasia are discussed in Chapter 77. LPH activity is sensitive to infectious and inflammatory diseases that affect the intestine. Therefore, after a bacterial, viral, or parasitic infection, residual symptoms resulting from the loss, and slow recovery, of LPH activity may occur. Infection with human immunodeficiency virus type 1 leads to decreased LPH activity and malabsorption of lactose. [172] In adults, LPH activity is resistant to the effects of starvation, [97] whereas in children, malnutrition causes a reduction in LPH mRNA. [173]

Maltase-Glucoamylase

Maltase-glucoamylase is a bifunctional enzyme that is responsible for the removal of glucose from the nonreducing end of short glucose polymers at a(1,4) bonds. [174] Both subunits of the enzyme appear to have the same substrate specificity and activity. The enzyme has low but measurable activity for hydrolysis of a(1,6) bonds. [175] Maltase-glucoamylase accounts for about 2% of the brush border protein and 20% of the total maltase activity in the small intestine. [93] Relatively little is known about the structure of the enzyme, although the human cDNA has been cloned. [176], [177] Much can be learned from a number of careful biochemical studies of enzymes isolated from the small intestine of rats, [76], [178] rabbits, [179] pigs, [175], [180], [181] and humans. [174], [182], [183]

Maltase-glucoamylase purified from human small intestine brush border membranes is a monomeric membrane protein of about 335 kd (others report 355 kd). [182], [183] The protein is synthesized as a polypeptide with a molecular weight of 255 kd, which is rapidly *N*-glycosylated to a 285-kd high-mannose form. [182] Further processing results in *N*-linked complex glycosylation and *O*-glycosylation, producing the mature enzyme. [182] The final sugar content of the molecule is about 30% of the total weight. [174], [182], [183] There is no evidence that proteolytic cleavage occurs intracellularly or extracellularly by pancreatic proteases. [182]

The characteristics of maltase-glucoamylase in other species differ markedly from those of the human enzyme. [175], [180], [181] The pig enzyme has a molecular weight of 240 kd. After insertion into the apical membrane, pancreatic proteases cleave the enzyme into smaller forms of 135 kd and 125 kd. [175], [181] The molecule is anchored to the membrane by an amino-terminal sequence and exists in the membrane as a homodimer. [180] The rat maltase-glucoamylase is synthesized as a polypeptide of 145 kd, which undergoes partial cleavage to yield two subunits with molecular weights of 130 and 145 kd. [72] The functional significance of these structural differences is unknown.

The developmental regulation of maltase-glucoamylase differs somewhat from that of SI and LPH. The enzyme is expressed at significant levels in the small intestine of suckling rats and mice, accounting for all the maltase activity at this developmental time because SI activity is not detectable. [93] At weaning, maltase-glucoamylase activity is induced to adult levels, but the increase in activity is less than that occurring with the induction of SI. [93], [184] Furthermore, pharmacological doses of corticosteroids before weaning lead to precocious induction of the enzyme, but the fold induction is less than that for SI. [93], [185], [186] Expression of maltase-glucoamylase in the human small intestine has not been investigated carefully during development, but the available data show that it is expressed early in development and that expression continues at similar levels after birth and into adulthood.

Trehalase

Trehalose is a disaccharide of glucose that is found in insects, yeast, and mushrooms. The mammalian intestine brush border contains small quantities of an enzyme that is able to hydrolyze this disaccharide. [187], [188] Although the total trehalase protein in the brush border amounts to only 0.1% of the total brush border proteins, it is important for the assimilation of foods that contain trehalose; for example, in persons who are trehalase deficient (as are 8% of Greenlanders), severe diarrhea develops after the ingestion of mushrooms. [187], [188], [189] and [190]

The cDNA for rabbit, human, rat, and mouse intestinal trehalase encodes a protein of 576 to 578 amino acids with a molecular weight of about 65.5 kd. [191], [192] and [193] There are four potential *N*-glycosylation sites in the rabbit cDNA, which may account for the higher molecular weight of the purified protein. [191] The rabbit cDNA encodes authentic trehalase by demonstrating enzymatic activity in frog oocytes injected with synthesized mRNA. [191] The protein is markedly different from the other brush border carbohydrases, because there is a cleaved signal peptide and the protein is fixed to the membrane by a glycosylphosphatidylinositol anchor. [191] It has no homology with other known proteins except for *Escherichia coli* trehalase, which shares 35% amino acid identity. [191] The mRNA for rabbit trehalase is distributed in a pattern commensurate with the expectations from previous studies of enzymatic activity. High levels of a 1.9-kb rabbit mRNA were measured in the small intestine and kidney, with low levels detected in the liver. [191] A 2.1-kb mouse mRNA was found only in the intestine and kidney. [192]

The developmental pattern of trehalase expression in the small intestine mirrors that of SI expression; levels in the human fetus and adult are similar, [93] whereas levels in rodents are very low during the suckling period, with marked induction at weaning. [93], [99], [184] Premature trehalase expression in rodent intestine can be induced with corticosteroids. [99], [184]

Integrated Function of Brush Border Enzymes

The membrane carbohydrases have individual activities for specific disaccharides. They also work to further hydrolyze the products of amylase hydrolysis, which include maltose, maltotriose, and a-limit dextrins. The generally accepted scheme for this process is shown in Figure 17-6. Maltase-glucoamylase has maximal activity for the removal of nonreducing sugars from oligosaccharides (i.e., linear and a-limit dextrins) that have between five and nine glucose molecules. [174] The sucrase and isomaltase subunits are also capable of metabolizing these oligosaccharides. [194] The a(1,6) bonds are resistant to hydrolysis until the adjacent nonreducing sugar is removed. Once the a(1,6) linkage is exposed, isomaltase is able to cleave this bond; glucoamylase also has minimal activity, but its role in completing this reaction is unknown. Hydrolysis of maltose can be accomplished by any of the carbohydrases, although the primary ones are sucrase and glucoamylase. This process yields monosaccharides that then can be absorbed across the epithelium.

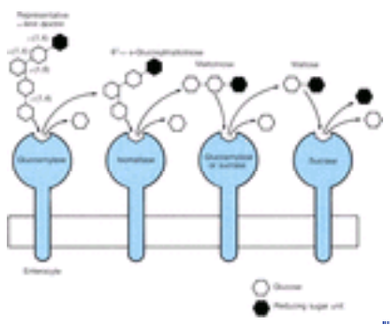

FIGURE 17-6. Overall process of digestion to monosaccharides. The hydrolysis of a-limit dextrins is a collaborative effort between multiple brush border enzymes.

ABSORPTION OF MONOSACCHARIDES

The only carbohydrates that can be absorbed across the epithelial cells of the small intestine mucosa are monosaccharides, or simple sugars. Digestion by

intralumenal and brush border processes is required to present simple sugars to the transport systems in the apical membrane of enterocytes. The transport processes involved in the absorption of monosaccharides across the enterocyte are passive or facilitated diffusion and Na^+-coupled active transport. The sugars that are normally presented to the enterocyte for absorption are glucose, galactose, and fructose. The mechanisms for the transport of each of these sugars has been elucidated, and this knowledge has led to a working molecular model for the absorption of each of these sugars (Fig. 17-7). As discussed in the following sections, these mechanisms and their relative contributions to absorption remain the subject of debate. [195]

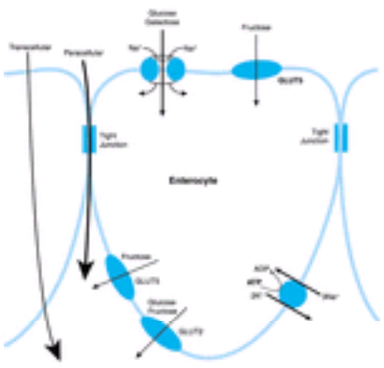

FIGURE 17-7. Transport processes responsible for movement of monosaccharides across intestinal epithelium in the enterocyte. Note that evidence suggests that GLUT2 may exist in the apical membrane when lumenal glucose levels are high (see refs. [195] and [228]).

Na^+-Coupled Glucose and Galactose Transport

The primary process for the active absorption of glucose and galactose is the active cotransport of Na^+ and sugar molecules across the apical membrane (see Fig. 7-7). [52], [196] Many studies of whole intestine mucosa, isolated enterocytes, and membrane vesicles isolated from the apical membrane fragments of the intestinal epithelium have shown that glucose and galactose are transported across the apical membrane in a Na^+-coupled electrogenic process. [52], [196], [197], [198] and [199] The active component of this system is dependent on a Na^+ gradient that is created and maintained by Na_+, K_+-ATPase (see section " Structure and Function of Na+,K+-ATPase in Enterocytes"). The concentration of solute in vitro at which the transport process is at half-maximum, or the enzyme's K_m for the transport of D-glucose, is between 0.2 and 0.5 mmol/L, and the K_m for the transport of extracellular Na^+ is between 2 and 7 mmol/L. [200] Transport studies suggest that there is a stoichiometry of 2 Na^+ for every hexose molecule. [198], [199] The transporter has been estimated to be about 75 kd in size and exists in the membrane as a homotetramer. [199], [201] These data led to a working molecular model of how the transporter may function (Fig. 17-8).

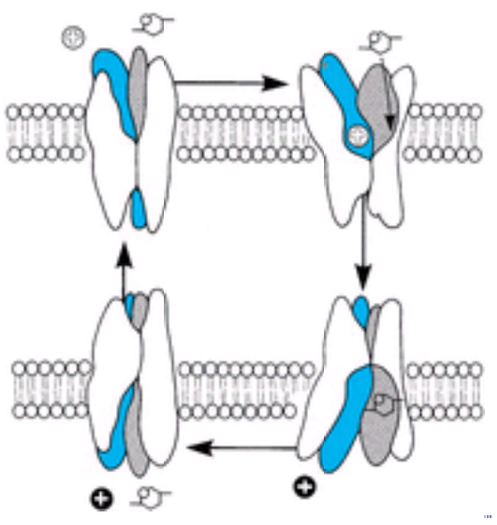

FIGURE 17-8. In the proposed mechanism for Na^+/glucose cotransport across the apical membrane, the transporter complex is likely to be a tetramer. (From Wright EM, Turk E, Zabel B, et al. Molecular genetics of intestinal glucose in transport. J Clin Invest 1991;88:1435.)

Knowledge of the molecular mechanism of this transporter advanced rapidly after cloning of the cDNA encoding the rabbit intestine Na^+/glucose cotransporter SGLT1. [202] The cloned cDNA encodes transport activity with the same relative specificity as the previously characterized native transport system D-glucose > a-methyl-D-glucose > D-galactose > 3- *O*-methyl-D-glucopyranose > > > L-glucose. [202], [203] Furthermore, the transport process in oocytes expressing the protein was found to be electrogenic, [204], [205] like transport of the native protein in the small intestine. The cDNA encodes a protein of 662 amino acids with a predicted molecular weight that correlates well with the biochemically defined size. Computer-aided analysis and biochemical studies have provided a model of the membrane topology of SGLT1 and other cotransporters in this gene family. SGLT1 is predicted to have 14 membrane-spanning domains, with one asparagine-linked carbohydrate group on the third extracytoplasmic loop. [206], [207] and [208]

The importance of the cloned transporter and proof that it is the major, if not the only, Na^+/glucose cotransporter in the intestine was shown in studies of two siblings with hereditary glucose galactose malabsorption, a rare autosomal recessive disease. [209] These two sisters had severe diarrhea in the first days of life that resolved with the elimination of glucose and galactose from the diet. A point mutation was identified in the genomic DNA of both sisters that resulted in a change of amino acid residue 28 from an aspartate to an asparagine. [209] Analysis of DNA from the parents proved that they were heterozygous for the mutation. The mutant mRNA was tested in *Xenopus* oocytes and was found to be nonfunctional for D-glucose transport. The mutant protein was synthesized and processed normally but was nonfunctional. [209] Since the description of the mutation in these two sisters, a variety of mutations in the coding region of the SGLT1 gene have been identified, many of which lead to the expression of proteins that are dysfunctional because of cellular trafficking abnormalities. [52], [210], [211], [212], [213] and [214]

The identification of the cDNA encoding SGLT1 led to additional biochemical analysis of the synthesis and transport properties of this Na^+/glucose cotransporter protein. [215] Inhibition of *N*-glycosylation in mammalian cells transfected with the cDNA decreased glucose transport activity by 80%, suggesting that this posttranslational modification is important for expression of the protein or for protein activity. [215] In vitro analysis of glycosylation showed that of the two potential *N*-glycosylation sites, only aspartate, [248] which resides on the external side of the membrane, is glycosylated. [216] Mutation of this aspartate to a glycine eliminated glycosylation of the protein; however, only a minimal decrease in activity occurred in *Xenopus* oocytes. [216] Glycosylation studies of the native protein suggested that a single *N*-linked residue was glycosylated and that partial deglycosylation did not affect the function of the transporter. [217] These data suggest that a single residue is *N*-glycosylated in the ER followed by modification in the Golgi apparatus. The protein does not require glycosylation for function, but glycosylation may be required for proper processing of the protein in mammalian cells. [215]

The expression and activity of glucose transport in the intestinal brush border are regulated by both short-term and longer-term processes. In the short term, activity of glucose transport is increased by both protein kinase A– and protein kinase C–dependent processes. [200] The mechanism of this enhanced activity is an increase in the number of membrane transporters, mediated by changes in exocytosis and endocytosis of membrane vesicles that contain the transport protein. [200] Study of the

transcriptional regulation of the SGLT1 gene in cell lines has shown involvement of HNF-1 and Sp1 proteins. [218] Longer-term regulation of glucose transport is mediated by changes in the expression of SGLT, which is controlled by changes in nutrient intake. [219], [220] and [221] Nutrient transport in the healthy intestine and in various disease states has been reviewed in the literature. [222], [223]

Facilitative Monosaccharide Transport

D-Glucose that is absorbed across the apical membrane crosses the basolateral membrane as the last step in intestinal absorption by a process of facilitative diffusion (not requiring energy) (see Fig. 17-7). [224] Because glucose is required by every cell in the body, facilitative glucose diffusion is a ubiquitous property of mammalian cells; however, different transport properties and regulatory characteristics are required by different cells. To meet these different requirements, there exists a family of genes (GLUT) that encode facilitative sugar transport proteins. [225], [226], [227], [228] and [229] The GLUT family of genes, their transport properties, and their tissue distributions are outlined in Table 17-4. The two genes that are expressed in the small intestine are GLUT2, the basolateral membrane–associated glucose transporter, and GLUT5, an apical membrane fructose transporter.

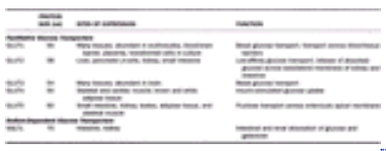

TABLE 17-4 Glucose Transporters

GLUT2 has molecular structural characteristics similar to those of the other members of this family of transporters. [225], [226] and [227] The protein has 500 amino acids with many hydrophobic residues that predict a total of 12 membrane-spanning domains (Fig. 17-9). There is a one long extracellular loop between membrane-spanning domains 1 and 2 that contains an asparagine that is *N*-glycosylated and one long cytoplasmic loop between membrane-spanning domains 6 and 7; the remainder of the connecting sequences for the membrane-spanning domains are short. The association of GLUT molecules has been investigated for several of the genes. GLUT1 appears to be a dimer, whereas GLUT4 appears to exist as a monomer. [225], [226] and [227] It has not been determined whether GLUT2 exists in the membrane as a monomer or as a multimer.

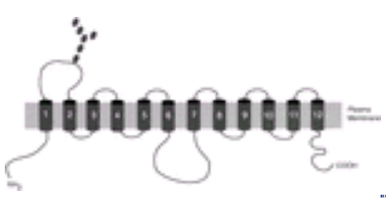

FIGURE 17-9. GLUT2 basolateral glucose transporter. This model of the monomer is suggested by the primary amino acid sequence. (From ref. [225].)

The transport properties of the GLUT2 protein have been studied by expressing it in *Xenopus* oocytes. [225], [226] and [227] The transporter has a high K_m for glucose (17 mmol/L) and is responsible for transport in the liver, kidney, and pancreatic ß-cell, as well as in the intestine. [225], [226] and [227] Therefore, the transporter has a high-level capacity for glucose transport. Evidence suggests that this protein also can transport fructose. The GLUT2 transporter protein is expressed on the basolateral membrane of enterocytes and renal tubular cells. [227] There is evidence that GLUT2 may also reside in the apical membrane and contribute to the facilitated diffusion of glucose, and that this process is regulated by a protein kinase C–dependent pathway. [195], [228]

Fructose Transport and Absorption

Studies in both humans and other animals have shown that there is a saturable, facilitative transport system for fructose in the intestinal epithelium that is less active than the transport system for glucose and galactose. [2], [230] Despite this lower level of activity, the fructose transport capacity of the entire small intestine mucosa is estimated to be large. [2] In vivo studies in humans, however, have shown that most individuals have a capacity for the absorption of fructose that is much more limited than the estimated value. [2] In these studies, hydrogen breath tests have been used to determine the threshold for malabsorption of fructose in test subjects.

The protein responsible for most apical membrane fructose transport is a member of the facilitative monosaccharide transporter family called *GLUT5*. Human [231] and rat [232] cDNAs have a similar predicted amino acid primary sequence: a protein of 502 amino acids with a molecular weight of 55.51 kd in the rat [232] and a protein of 501 amino acids in humans. [231] Analysis of this amino acid sequence suggests that there are 12 membrane-spanning domains, as for the other GLUT molecules. There is also one potential *N*-linked glycosylation site that is predicted to be located on an extracellular loop of protein.

Despite the similar structural characteristics of human and rat GLUT5 molecules, evidence of functional differences was revealed after assessment of GLUT5 in a *Xenopus* oocyte expression system. Human GLUT5 transports fructose and is specifically inhibited by excess fructose but not by glucose. [231] In contrast, the transport of fructose by rat GLUT5 is inhibited by both fructose and glucose, and furthermore, rat GLUT5 is able to transport 2-deoxyglucose. [232] These data suggest that GLUT5 in the rat facilitates the transport of both glucose and fructose, whereas human GLUT5 transports fructose exclusively. The functional significance of these findings requires assessment in the whole animal.

The tissue distribution of GLUT5 also differs between rats and humans. Human GLUT5 mRNA and protein are expressed in the small intestine, kidney, testes, adipose tissue, and skeletal muscle. [233] Rat GLUT5 mRNA is expressed in the small intestine, kidney, and, at lower levels, in the brain, but it is not expressed in adipose tissue, testes, or skeletal muscle. [232]

Little fructose is metabolized in the enterocyte. [2] Fructose is transported across the basolateral membrane and is taken up and metabolized rapidly by the liver, resulting in low postabsorptive blood levels of fructose. [2] Some in vivo experiments in human subjects have raised the question of whether more than one type of fructose transport process exists. [2] These experiments show that malabsorption of fructose in humans can be prevented by the simultaneous administration of glucose. [2] Although there are possible mechanisms by which a single fructose transport process can yield these results, it has been argued that another glucose-responsive system may be present in enterocytes. Fructose likely is transported across the basolateral membrane by GLUT2, [229], [234] and evidence of GLUT5 expression on the basolateral membrane suggests that it too may be a fructose transporter. [235] It is also possible that GLUT2 on the apical membrane transports fructose. [195], [228]

Expression of the GLUT5 protein is regulated by the presence of its substrate. [221] If experimental animals are fed fructose, the expression of GLUT5 mRNA and protein increases an induction process that requires fructose to be in contact with the brush border membrane. [221] Precocious expression of GLUT5 in suckling animals is also induced by lumenal fructose. [236]

Structure and Function of Na^+,K^+-ATPase in Enterocytes

Na^+,K^+-ATPase is found in the membrane of nearly every cell in the animal kingdom and is essential for cell viability. [237] This integral membrane protein uses ATP

energy to transport 3 Na^+ out of the cell and 2 K^+ into the cell, thus maintaining an ionic and electrochemical gradient between the intracellular and extracellular space. Na^+, K^+-ATPase, localized on the basolateral surface of the enterocyte, functions in the absorption of monosaccharides by maintaining low intracellular Na^+ concentrations, thereby providing a large gradient for transport from the intestinal lumen across the apical membrane.

Na^+,K^+-ATPase comprises two proteins: the a and ßsubunits. [237], [238] The a subunit, also called the *catalytic subunit,* is 112 kd in size and contains the binding sites for ATP and cardiac glycosides. The 35-kd ß subunit is required for activity, but its specific function has not been established. Isozymes of the a and ß subunits have a complex pattern of tissue distribution. [238], [239] and [240] The significance of the differential expression of the subunits in various tissues has been evaluated, [240] but the implications for intestinal epithelial function are unknown.

Passive Movement across the Epithelium

Intestinal absorption of glucose continues in a linear fashion when glucose levels exceed those that saturate the Na^+/glucose cotransport system. [195] Therefore, other mechanisms for glucose transport across the enterocyte are thought to exist. Two potential mechanisms for the passive movement of monosaccharides across the intestinal epithelium are paracellular and transcellular pathways (see Chapter 8).

The paracellular pathway allows water and solutes to pass between epithelial cells and has been used to explain the absorption of glucose at high lumenal levels that oversaturate the SGLT1 system. The zona occludens forms a high-resistance seal between epithelial cells, the tightness of which can be altered by physiological processes. [241] One cellular mechanism for altering this intercellular seal may be the contraction of the cellular cytoskeleton that is connected to the zona occludens apparatus, which results in separation of the cells at the tight junction. [241] A trigger for this altered junction permeability is apical membrane Na^+-coupled glucose transport. [241] Therefore, it has been hypothesized that the absorption of glucose may lead to an increase in paracellular permeability and the movement of water and solutes by the paracellular route. [241] At times of high intralumenal glucose concentration, this transport mechanism may be significant.

Passive diffusion of monosaccharides across the apical membrane of the enterocyte (i.e., a transcellular pathway) is very inefficient because sugars are water-soluble molecules that are efficiently excluded by lipid bilayers. Diffusion would be enhanced if there were a facilitated transporter, as there is for the basolateral membrane. As mentioned previously, GLUT2 may be inserted into the apical membrane via a regulated trafficking pathway. [228] This would lead to facilitated diffusion of glucose across the apical membrane, and because GLUT2 is a high-capacity transporter, this could account for linear increases in glucose transport exceeding the capacity of the SGLT1 system.

The contribution of the paracellular pathway versus facilitated diffusion remains controversial. However, it is likely that one or the other predominates because the presence of a facilitative transport protein in the apical membrane would dissipate the glucose gradient that is hypothesized to drive the paracellular movement.

Water Movement Accompanying the Transport of Sugars

Water crosses the intestinal epithelial cell layer as monosaccharides are transported across the cell; this is the rationale for adding glucose to oral rehydration solutions. The mechanism by which water crosses the epithelial cell layer is poorly defined. One hypothesis is that the driving force for water movement is a localized, intracellular osmotic gradient created by the active transport of monosaccharide molecules across the membrane. This mechanism probably is not the major mechanism, however, because the basolateral and brush border membranes of enterocytes are relatively impermeable to water. In some epithelial tissues, such as the renal collecting ducts, specialized transmembrane proteins called *aquaporins* transport water. [242], [243] Aquaporins have not been identified definitively in the intestine, however. Another widely held hypothesis is that most water movement in the intestine occurs by the paracellular route. Because the intestinal epithelium is semipermeable, the tight junctions can act as size-selective pores, allowing the passage of small molecules for which there are no specific transporters, including water. As discussed previously, the paracellular pathway of water movement may be regulated by changes in the epithelial cells. The driving force for this process is also assumed to be osmotic gradients across the epithelial junctions as well as bulk flow.

Wright and colleagues [244], [245], [246] and [247] introduced a revolutionary concept for intestinal water transport. Experiments in *Xenopus* oocytes have shown that SGLT acts as a low-capacity water channel in the absence of glucose. When glucose and Na^+ are present in the extracellular medium, SGLT can cotransport 264 water molecules for every 2 Na^+ and every 1 molecule of glucose. The capacity of this water-transporting system in a human who ingests an average carbohydrate diet is estimated to be 5 L of water per day. Transporters of other molecules, including amino acids, may also use this mechanism of water transport. Interestingly, GLUT2 may also transport water, which would allow apically located GLUT2 to facilitate the movement of water molecules across the apical membrane. [195] Quantitative in vivo experiments will be required to assess the relative contribution of this type of water transport associated with monosaccharide transport and that of paracellular water movement.

SPATIAL LOCALIZATION OF HYDROLYSIS AND TRANSPORT ALONG THE CRYPT-VILLUS AXIS

In a consideration of physiological processes, it is important to recognize that the intestine is lined with a complex epithelium in which each enterocyte does not have the same phenotypic characteristics. The epithelial cells of the small intestine are generated from a fixed stem cell population that has been localized to the lower portion of intestinal crypts. [83], [248], [249] and [250] These stem cells give rise to four primary epithelial cell types that reside in the intestinal mucosa: absorptive enterocytes, goblet cells, enteroendocrine cells, and Paneth cells. [249], [250], [251] and [252] Precursors of absorptive enterocytes in the crypts constitute about 90% of the cells in the crypt compartment, and mature absorptive enterocytes constitute more than 95% of cells located on the intestinal villus. [252] The other three primary phenotypes make up a small, but important, percentage of the total number of cells. Absorptive enterocytes migrate from the crypt compartment to the intestinal villus and are extruded into the intestinal lumen from the villus tip. The migration of cells occurs in a linear fashion, with cells moving vertically from their site of origin in the crypts. [253], [254] and [255] This process occurs over about 3 days in mice and over 4 to 5 days in humans. [248] Therefore, the epithelial lining of the mucosa is renewed continually at a relatively rapid rate.

The phenotype of enterocytes changes as cells move from the undifferentiated crypt compartment to the villus. Changes in phenotype are mediated by the expression or repression of sets of genes. The pattern of expression along the crypt-villus axis for multiple genes involved in carbohydrate assimilation has been determined. These data provide information that indicates where sugar absorption occurs. As discussed previously, a number of investigators have examined the pattern of expression of SI mRNA, protein, and activity along the crypt-villus axis. These studies suggest that there is little SI in crypts and that SI levels are maximal in the mid villus region. LPH gene expression in rodents is remarkably similar to that of SI. [163] In the small intestine of adults who have hypolactasia, LPH protein expression is patchy. [256] GLUT5 mRNA expression along the crypt-villus axis is nearly identical to that of SI mRNA, an intriguing finding inasmuch as GLUT5 transports fructose, one of the major products of SI enzymatic action. [232], [257] The pattern of SGLT mRNA expression in the crypts and lower villus region is the same as that of SI and GLUT5, but in the villus tip cells, expression is maintained at high levels. [257] GLUT2 expression is limited to the basolateral membrane of mature villus enterocytes. [258]

The components of the brush border carbohydrate assimilation system appear to be expressed in a coordinate pattern, suggesting that similar regulatory mechanisms may be involved in directing their expression. The enterocytes located in crypts probably do not normally function in carbohydrate assimilation; however, the expression of proteins involved in sugar assimilation may be expressed earlier along the crypt-villus axis in certain pathological conditions, such as diabetes mellitus. [130] This is an important level of regulation in addition to the level of expression in individual enterocytes.

EFFICIENCY AND RATE-LIMITING STEPS OF CARBOHYDRATE ASSIMILATION

The molecular mechanisms for the biochemical processes involved in carbohydrate absorption are being established, as indicated in the preceding sections; however, the integration of this information for an understanding of the absorption of real food in humans has lagged behind the basic scientific discoveries. Therefore, some controversy, as well as misleading statements, can be found in the literature. In simplistic terms, the efficiency of starch absorption can be related to two factors: the availability of food starch for intralumenal hydrolysis and the activity of the brush border enzymes and transport processes for absorption. A meal of soluble starch is assumed to be completely available for hydrolysis; therefore, the efficiency of absorption is dependent only on the brush border hydrolases and monosaccharide transport processes. Under these conditions, the traditional view has been that D-glucose transport at the apical membrane is the rate-limiting step. In reality, however, all ingested starch is not completely available for lumenal hydrolysis. Therefore, the rate-limiting step may be viewed as being dependent on properties of

the food itself.

A certain portion of starch is not digested or absorbed in the small intestine. [259], [260], [261], [262] and [263] In fact, a small degree of starch malabsorption should be considered a normal physiological phenomenon. [262] The quantity of starch malabsorption after ingestion of a variety of foods has been tested by ileal intubation, [260], [261] in patients with an ileostomy, [263] and by hydrogen breath tests. [259], [261] These experiments indicate that the passage of carbohydrate into the colon is common for many food types. The degree of malabsorption of ingested starch in healthy volunteers ranges from 2% to 20%. [260] The types of compounds that are complexed with starch, as well as the degree of food processing, appear to affect hydrolysis. [259], [264], [265] Sucrose also is partially malabsorbed in otherwise healthy persons, [266] as is lactose in subjects with normal LPH levels. [262] Therefore, the accessibility or contact time of disaccharides to brush border enzymes also may be a limiting factor.

Starch and oligosaccharides that reach the colon cannot be absorbed in the absence of bacteria. [266] Carbohydrates in the colon are metabolized rapidly by the colonic bacterial flora to short-chain fatty acids, including acetic, butyric, and propionic acids, which are absorbed rapidly by the colonic mucosa. [266], [267] and [268] These short-chain fatty acids serve as a primary fuel for colonocytes. [17] The energy derived from the metabolism of carbohydrates in the human colon is as high as 5% to 10% of total energy requirements. [267] Research has been directed at determining whether short-chain fatty acids provide a beneficial fuel during intestinal inflammation. [16], [269], [270]

Another gauge of the availability of starch from different sources is the glycemic index. [271], [272] This index is calculated by measuring the area under the serum glucose curve for 2 hours after a meal and comparing it with the area obtained for a reference food (e.g., glucose, white bread) set at 100%. This index does not assess the completeness of absorption of the available glucose; rather, it measures how rapidly the glucose is absorbed. When this method is used, different foods containing the same amount of total glucose demonstrate a wide range of glycemic indices (Table 17-5). It is difficult to predict what the glycemic index will be for a particular food. Furthermore, the difference between simple and complex carbohydrates is not straightforward because some complex forms (e.g., potato) have higher indices than simple sugars (e.g., sucrose; see Table 17-5). [271] This measure has been found to be useful in the treatment of diabetes mellitus.

FOOD	GLYCEMIC INDEX* (%)
Percentage of response to glucose	
Glucose	100
Corn flakes	80
Rice	72
White bread	69
Corn	59
Oatmeal	49
Kidney beans	29
Percentage of response to white bread	
White bread	100
Whole grain rye	58
Whole meal wheat	99
Rice	83
Spaghetti	66
Potato (russett, baked)	135
Potato (sweet)	70
Apple	53
Banana	79
Pear	47
Raisins	93
Fructose	30
Glucose	138
Honey	126
Maltose	152
Sucrose	86
Whole milk	49
Ice cream	52
Yogurt	52

*Area under 2-hour glucose curve.
Data from refs. 271 and 272.

TABLE 17-5 Glycemic Index of Selected Foods

According to traditional theories, the absorption of oligosaccharides, including both starch hydrolysate and disaccharides, is dependent on brush border hydrolytic enzyme systems and monosaccharide transport. Sucrose, maltose, maltotriose, and a-limit dextrins are hydrolyzed completely to their component monosaccharides, and the rate-limiting step for absorption is transport of the monosaccharides across the apical membrane. In contrast, the rate-limiting step in the absorption of lactose is hydrolysis by LPH. In vitro studies have questioned these time-honored concepts. The transport of glucose polymer–derived glucose was investigated in rabbit jejunal mucosa by measuring unidirectional glucose flux in the presence and absence of various enzyme inhibitors. [273] The findings suggest that brush border hydrolysis may be the limiting factor in the absorption of glucose polymer–derived glucose. [273] Furthermore, studies in humans have shown that 2% to 4% of ingested sucrose may enter the colon [266]; the level at which sucrose malabsorption occurs is unclear.

One important physical aspect of the lumenal content–enterocyte interface is the unstirred water layer. [274], [275] and [276] This layer is a stationary region of the lumenal fluid that is in contact with the mucosal surface of the intestine. Transport of solute molecules across this region of lumenal fluid occurs by simple diffusion based on concentration gradients. The thickness of this layer has been estimated by using a number of experimental methods. [274], [275] A practical assessment of these data suggests that the thickness of this layer in vivo has been grossly overestimated, and that the contribution of the unstirred water layer to the resistance to oligosaccharide and monosaccharide absorption is much less than previously reported. [276] The thickness of the unstirred water layer may increase in disease states (e.g., celiac sprue) and thereby contribute to malabsorption. [277]

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CHAPTER 18

Nicholas O. Davidson

INTESTINAL LIPID ABSORPTION

Dietary lipid represents a major caloric source in most Western cultures. In addition to the 120 to 150 g of lipid consumed each day, the small intestine processes 40 to 50 g of biliary lipid, together with small amounts of lipid contributed by sloughed mucosal cells and bacteria. Tremendous advances have been made in understanding how this process is efficiently coordinated, and the widespread attention given the issues of diet and cardiovascular health has renewed investigation of the role of the small intestine as an active participant in systemic lipoprotein metabolism.

INTESTINAL LIPID BALANCE

The major form of dietary lipid consumed each day is long-chain (>14 carbon atoms) triglyceride, combined with smaller amounts (=10 g/d) of the phospholipid lecithin. The predominant sources of intralumenal lecithin arise from biliary secretion (10–20 g/d) and membrane phospholipid from desquamated intestinal cells. Most Americans consume 200 to 500 mg of cholesterol daily, which, in conjunction with large fluxes of biliary cholesterol (1–2 g/d), constitutes a major source of the body's daily metabolic requirements for this sterol. Plant sterols, principally ß-sitosterol, and shellfish sterols may be substantial dietary lipid components. Each person daily ingests other complex lipids, such as waxes, lipovitamins (i.e., A, D, E, K), lipid-soluble by-products of commercial food processing, and hydrophobic xenobiotics, including potentially toxic insecticides and preservatives. Absorption of intestinal lipids is highly efficient (>95% coefficient of absorption for triglyceride, <5% for ß-sitosterol), and the underlying physiological parameters regulating uptake, intracellular processing, and secretion are distinctive.

INTRALUMENAL LIPID DIGESTION

Intestinal lipid digestion is a complex, multistep process, depending first on the effective dispersion of fat into a stable form with a large surface area. This process is called *emulsification*. Second, several lipolytic enzymes become sequentially adsorbed to this emulsion and, under appropriate conditions, mediate the digestion of long-chain triglyceride, phospholipid, and sterol ester bonds, a process called *lipolysis*. Lipolysis also involves important phase transitions; the controlled, sequential evolution of lipolysis is integral to the successful completion of digestion and solubilization of lipid. These lipolytic products subsequently undergo incorporation into aggregated mixtures of bile salts and biliary lecithin for delivery to the microvillus membrane of the villus cell. This step is called *micellar uptake*, a process that necessitates traversing the aqueous milieu of the small bowel lumen. The physical and apparent barriers limiting this process are considered in subsequent sections.

Intragastric Events

Lipid digestion begins in the stomach with the initiation of emulsification. The interaction of triglyceride droplets with dietary and other intralumenal sources of lecithin is a key step in providing stability to the crude lipid emulsion resulting from the shearing action of gastric peristalsis. Proteolytic fragments resulting from peptic digestion also stabilize the emulsion.

The properties of such emulsions have been characterized in vivo and in vitro by using phase equilibria and magnetic resonance spectroscopy with carbon 13. [1,2] Studies of the initial properties of a model emulsion consisting of triglyceride, phospholipid, cholesterol, and water predict a surface layer of lecithin consisting of about 3% triglyceride by weight and small amounts of cholesterol. [1] Most of the triglyceride is sequestered within the core of this emulsion and exists as a homogeneous oil phase. In this conformation, limited quantities of surface triglyceride are accessible for lipase digestion. The rapid exchange that occurs between core and surface triglyceride molecules continuously exposes new substrate to lipase, facilitating the efficient initiation of lipid digestion.

Intragastric lipolysis is of substantial quantitative importance in humans and experimental animals. Lipase activity has been demonstrated in the gastric contents of neonates born prematurely as early as 26 weeks of gestation and in mucosal biopsy specimens from adults up to 80 years of age, although enzyme activity decreases in persons older than 60 years. [3,4] Estimates based on studies in ruminants and extrapolated to data for humans suggest that normal intragastric lipolysis may account for 20% to 30% of total intralumenal lipid digestion. [4,5] A relative or absolute decrease in pancreatic lipase secretion may increase the percentage contribution of intragastric lipid digestion to more than 90%.

Earlier evidence suggested that two distinct lipases were detectable in human gastric contents: lingual lipase and a gastric lipase principally of fundic origin. [4,5] and [6] Although rodents may have more than one source of gastric lipase activity, this is unlikely to be the case in humans. In one study, the lipase activity was measured in tissue supernatants from numerous regions of the human tongue (including Ebner glands), the pharynx, and the upper gastrointestinal tract. Virtually all the lipase activity was found in the gastric fundus, with less than 0.015% of the total activity attributable to a lingual source. [4] More recent studies [7] have evaluated the specific activity of lipases from gastric content in comparison to the recombinant proteins, finding that although the absolute activity is lower than predicted from in vitro studies, total lipase activity is accounted for by secretion of gastric lipase without the need to invoke an alternative source.

Intragastric lipolysis differs in several important respects from lipolysis in the small intestine. Based on assay of human gastric contents and biopsy of gastric fundic cells, preduodenal lipase activity displays a broad pH range with an optimum of about 4.0 to 5.5 and preferential activity against the 3-position long-chain fatty acid ester bond of triglyceride. [3,4] and [5,8] The major gastric lipolytic products are diglyceride and fatty acid. Activity of the purified enzyme in vitro is inhibited in the presence of bile salts, and a similar effect is observed after bile salts are added to human gastric aspirates, particularly in concentrations greater than 5 mmol/L. [3,8] This suggests that intragastric lipolysis is sensitive to interfacial denaturation, although at the bile salt concentrations typically encountered in neonates (i.e., 1–2.0 mmol/L), in whom intragastric lipolysis may be particularly important, little inhibitory effect is predicted. [3] In addition to its low pH optimum and inactivation by bile salts,

preduodenal lipase is resistant to pepsin and requires no cofactors. [9] At neutral to alkaline pH and in the presence of bile salts, preduodenal lipase is rapidly and extensively degraded by pancreatic proteases, so that it unlikely that it would function effectively in the upper small intestine under normal circumstances. [10] Crystal structure analysis of recombinant human gastric lipase has confirmed these predictions. [11]

In the stomach, crude dietary lipid undergoes emulsification and initiation of lipolysis. This contribution to lipid digestion may assume particular importance in neonates, in whom preduodenal lipase secretion can compensate for a developmental deficiency in pancreatic lipase secretion, resulting in intragastric lipolysis of 60% to 70% of dietary triglyceride. [12] Similarly, triglyceride absorption is maintained at about 50% in children with a congenital absence of pancreatic lipase. [13], [14] Patients with cystic fibrosis and exocrine pancreatic insufficiency typically exhibit various degrees of steatorrhea. [15] In one study, preduodenal lipase activity in five such patients accounted for more than 90% of the total lipase activity measurable at the ligament of Treitz. [15] A notable contributor to the apparent preservation of preduodenal lipase activity in these patients was the low postprandial intralumenal pH of about 4.0 to 5.0, presumably reflecting decreased bicarbonate secretion, which would permit continued lipolysis by gastric, but not pancreatic, lipase. [15] Gastric lipase activity also may play a role after total gastrectomy, when fat malabsorption is not uncommon. [16] At a pH of 5.0 to 6.0, essentially all the liberated fatty acids remain protonated and, with diglyceride, are dissolved in the oil phase of the triglyceride emulsion. This emulsion, containing partially digested triglyceride and its lipolytic products, enters the small intestine, where most intralumenal fat digestion takes place.

Small Intestine Events

Several key events governed by the physicochemical environment of the upper small intestine act in concert to augment the rate and extent of lumenal lipolysis. First, at the duodenal pH of 6.0 to 7.5, the fatty acids released by intragastric lipolysis become ionized. They migrate to the surface of the emulsion, where, as charged particles, they assist in emulsification and help anchor colipase to the triglyceride emulsion. [1] This is an essential step in initiating pancreatic lipase digestion. Second, various hormonal stimuli induce gallbladder contraction, resulting in the entrance of bile into the duodenum. The combination of alkaline pH, adequate Ca^{2+}, bile salts, and lecithin in the presence of lipolytic enzymes initiates a dynamic and dramatic cascade of events.

Lipolytic enzymes encounter the triglyceride emulsion resulting from the gastric processing of ingested fats, and in the presence of appropriate cofactors, ionic conditions, and pH, they accelerate intralumenal lipolysis. [13], [14] and [15], [17], [18], [19], [20], [21], [22], [23], [24], [25], [26], [27], [28], [29], [30], [31], [32], [33], [34], [35], [36], [37], [38], [39], [40] and [41] Pancreatic lipase and colipase are secreted from acini in equimolar amounts. Effective activation of pancreatic lipase requires several interdependent steps. After bile salts clear the lipid emulsion from adsorbed protein, the colipase-lipase enzyme complex attaches to the triglyceride emulsion—colipase physically anchors lipase in an appropriate configuration to expose its active site. To achieve this anchor, colipase binds directly to the triglyceride emulsion and indirectly to the bile salts through micelle formation. The bile salts then bind to the emulsion surface. These interactions are augmented by the presence of ionized fatty acids. [18], [21], [22], [26], [39] Pancreatic lipase activity in vitro is inhibited by bile salts and phospholipid through desorption of surface protein (including lipase) from the lipid emulsion. This inhibitory process is specifically reversed by colipase.

Colipase-dependent anchoring of lipase to the lipid emulsion is further enhanced by phospholipase A_2 digestion of the phospholipid on the surface of the lipid emulsion, allowing exposure of the triglyceride core to the colipase-lipase complex. Phospholipase A_2 digestion requires bile salts and Ca^{2+} for activation, which may further assist colipase-lipase–mediated triglyceride lipolysis by providing a mechanism for removal of the lipolytic products. As previously described, dietary and biliary phospholipids play an important role in stabilizing the crude triglyceride emulsion. The major intralumenal phospholipid is phosphatidylcholine (i.e., lecithin), which is unabsorbable intact and requires hydrolysis by phospholipase A_2 to yield a mole of fatty acid and a mole of lysolecithin per mole of substrate. Lysolecithin has a high aqueous solubility and undergoes rapid diffusion and uptake. The newly generated long-chain fatty acids undergo micellar solubilization and participate in lipolytic product phase formation.

Sterol and lipovitamin esters are largely sequestered within the oily core of the triglyceride emulsion and undergo hydrolysis of their fatty acid ester bonds through the actions of cholesterol esterase. [36], [37] Bile salts are an obligatory component of this reaction, serving as an essential enzyme cofactor and as a vehicle for effective solubilization of the extremely hydrophobic sterol and lipovitamin alcohols.

The dynamic aspects of intralumenal fat digestion have been elucidated in a series of elegant in vitro and in vivo studies. With the use of mixtures of triglyceride, pancreatic lipase, colipase, and bile salts in a buffered solution at duodenal pH of about 6.5, researchers have demonstrated the existence of at least two sequential product phases of triglyceride lipolysis, as visualized by light microscopy. [42] The initial products were contained within a lamellar liquid crystalline phase composed of a shell of birefringent calcium soaps (i.e., a 1:2 mixture of Ca^{2+} and fatty acid) encapsulating an oily core of unhydrolyzed triglyceride. When a similar analysis was conducted at pH 5.0, no product phases were visible, suggesting that because the liberated fatty acids are minimally ionized at gastric pH, calcium soap formation does not occur. [2] Development of this calcium soap phase can be preserved in vitro by performing incubations in the absence of bile salts, suggesting that this may be a transient, intermediate phase of intralumenal lipolysis. [2]

In the presence of bile salts, a second phase is formed, the composition of which varies continuously as a function of the molar ratio of lipolytic products to bile salts. This phase continuum is referred to as the *viscous isotropic phase* and is composed largely of monoglyceride and protonated fatty acid in a 1:1 molar ratio. [42] The initial products of lipolysis, visible 5 minutes after the addition of lipase in vitro, are spherical vesicles 11 to 30 nm in diameter. As lipolysis proceeds to completion, lamellar products accumulate at the surface of unhydrolyzed triglyceride emulsions. In the presence of physiological micellar concentrations of bile salts, multilamellar product phases and lamellar product vesicles are also detected in vitro, with product vesicles averaging 50 to 250 nm in diameter. A similar situation is encountered in vivo; after intralumenal lipid instillation, a rough multilamellar product appears initially (Fig. 18-1), giving way to an abundance of product vesicles as bile salts enter the intestinal lumen, inducing micellar solubilization of lipolytic products for delivery to the microvillus membrane.

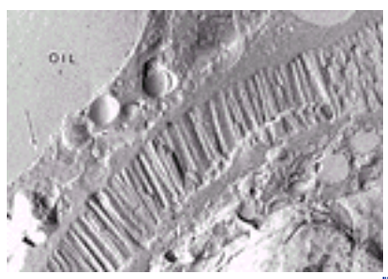

FIGURE 18-1. Morphology of intestinal lipid digestion in vivo. Lamellar product vesicles (*arrows*) are adjacent to a large triglyceride droplet. The product phases are in proximity to the enterocyte brush border. (*Bar* = 500 nM; from Rigler MW, HonKanen RE, Patton JS. Visualization by freeze fracture, in vitro and in vivo, of the products of fat digestion. J Lipid Res 1986;27:836.)

The multilamellar phase of in vitro and in vivo systems appears to correspond to the viscous isotropic phase previously demonstrated by light microscopy. [42] It is proposed that as lipolysis proceeds in the upper small intestine, the lipolytic products (i.e., monoglyceride and fatty acid) initially accumulate to form rough multilamellar phases (e.g., viscous isotropic phase). The density and size of these phases vary according to divalent cation and bile salt concentrations. In the presence of bile salts above a critical micellar concentration, lipolytic products are partitioned into mixed micelles, the size of which reflects the ratio of bile salt to lipolytic product. This process is represented schematically in Figure 18-2. The concordance of the results of in vitro analyses with in vivo experiments indicates the likelihood that this phase transition complex occurs physiologically in humans.

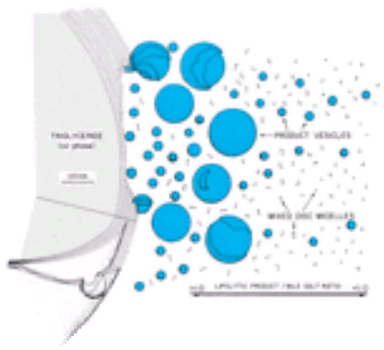

FIGURE 18-2. Accumulation of lipolytic product (LP) lamellae and their subsequent dispersion into vesicles, illustrating the formation of product vesicles whose density and size vary as a function of the LP–bile salt ratios. At high LP–bile salt ratios, large LPs are obtained, and discoidal structures of high flotation density are obtained at higher bile salt–LP ratios. (Adapted from Rigler MW, HonKanen RE, Patton JS. Visualization by freeze fracture, in vitro and in vivo, of the products of fat digestion. J Lipid Res 1986;27:836.)

Micellar Uptake and Delivery of Lipolytic Products to the Brush Border

Adequate lumenal concentrations of conjugated bile salts are a key factor governing the effective solubilization of lipolytic products. Lipolytic products and phospholipid (i.e., biliary lecithin) become incorporated into these aggregates, producing mixed micelles. The more hydrophobic products of lipolysis partition preferentially into the core of the micelles and traverse the aqueous medium of the intestinal lumen by virtue of micellar solubilization. Examples of lipids that depend critically on this mechanism include cholesterol, fat-soluble vitamins, and plant sterols.

In the absence of bile salts at effective micellar concentrations, which can be caused by biliary obstruction, bacterial deconjugation (e.g., bacterial overgrowth), or abnormal small intestine acidity (e.g., acid hypersecretory states), absorption of hydrophobic compounds is reduced virtually to zero. In contrast, absorption of fatty acids and monoglycerides is facilitated by, but is not exclusively dependent on, micellar solubilization. As a result, triglyceride malabsorption is relatively mild (<30%) in patients with total biliary obstruction, in whom lipolysis would be expected to proceed normally but the formation of micelles from lipolytic products would be absent.[43] The explanation presumably reflects the phase equilibria produced after triglyceride digestion. In the absence of bile salts, fatty acids and monoglycerides exist as a lamellar, viscous isotropic phase from which monomeric fatty acids can be delivered to the brush border membrane for uptake.

Effective solubilization of lipolytic products within the bulk phase of lumenal contents is one mechanism by which diffusional resistance to mucosal uptake may be overcome. The diffusion of hydrophobic molecules through this aqueous medium, composed of a dense mucous layer overlying the brush border glycocalyx, and their delivery to the brush border membrane are components of a major rate-limiting process in lipid absorption. One explanation of the behavior of solutes within the bulk phase of lumenal contents is the unstirred layer hypothesis, which predicts that contents in the center of the lumen are ideally mixed while contents at the outermost region of the loop (i.e., closest to the brush border membrane) remain functionally unstirred. Quantitation of this unstirred layer has produced figures for the rat in the range of 100 to 1100 µm.[44], [45] and [46] In contrast, the laminar flow model predicts that solutes would be mixed by a physical process that depends on their relative positions with respect to an ideal central axis of vectorial flow.[47] The essential difference between the two models resides in the assumption that the laminar flow model has fixed physical dimensions derived from the direct determination of solute movement relative to fluid movement in longitudinal and horizontal axes and from the determination of epithelial compared with lumenal resistance to solute movement. The laminar flow hypothesis is supported by the observation that absorption of a variety of probes is independent of lumenal volume.[47] The results obtained in these experimental systems provide a useful basis for understanding lumenal resistance to solute movement in mammalian intestine.

Intestinal brush border membranes contain a heparin-like binding site that mediates specific, saturable, and heparin-displaceable binding of pure pancreatic cholesterol esterase (100-kd peptide) and pancreatic lipase, but not amylase or ribonuclease.[48], [49] These findings imply that binding of lipolytic enzymes may occur at the brush border membrane, obviating the requirement for lipolytic products to traverse an aqueous diffusion barrier. These investigators postulated that in situ lipolysis of cholesteryl ester or triglyceride releases fatty acids, monoglyceride, and free cholesterol in proximity to the brush border membrane, where they undergo rapid uptake.[48], [49]

Lipolysis and Uptake of Phospholipid and Medium-Chain Triglyceride

A triglyceride with fatty acyl groups that have a 6- to 12-carbon chain length is referred to as a *medium-chain triglyceride*. Its lumenal and intracellular metabolism differs in several respects from that of a long-chain triglyceride. First, medium-chain triglycerides form more expanded surface films as emulsions than do long-chain triglycerides, facilitating intragastric lipolysis.[1] Second, as a result of their intrinsically higher aqueous solubility, medium-chain fatty acids released by intragastric and small intestine lipolysis are effectively absorbed by gastric and small intestine epithelium and undergo rapid transepithelial delivery. Third, medium-chain fatty acids do not become activated to their coenzyme A (CoA) derivative for reassembly into complex lipid but rather undergo direct secretion into the portal vein, where they become bound to albumin for transport to the liver.[50], [51] Fourth, although intrinsically more susceptible to lipase digestion than long-chain triglycerides, a proportion of medium-chain triglycerides may be absorbed directly, without prior lipolysis. Direct proof of this last metabolic event is lacking in humans.

The role of phospholipid hydrolysis in the regulation of lipid absorption is unresolved. Rats treated with a competitive phospholipase A_2 inhibitor demonstrated reduced cholesterol and triglyceride transport into lymph,[52] supporting a role for lumenal phospholipid hydrolysis in lipid absorption. Interestingly, although human studies have shown that lumenal phospholipid may interfere with cholesterol absorption, the absorption of triglyceride, phospholipid, or cholesterol was unchanged in phospholipase A_2 -/- mice,[52] suggesting that other lipases may accomplish the task of phospholipid hydrolysis.

Bile Salt Absorption

Micellar lipid delivery involves the disgorgement of micellar contents at the brush border membrane for uptake. Conjugated bile salts are passively absorbed as monomers throughout the upper small intestine, and because of the efficient processes of hepatic uptake after portal vein delivery and reexcretion into bile, they become rapidly available again to participate in micellar lipid solubilization. In addition to passive uptake throughout the small intestine, an active Na^+-coupled uptake process in the mature ileum retrieves more than 95% of the intralumenal bile salts. Ileal absorption of bile salts is mediated through the apical Na^+-dependent bile transporter (ASBT), originally referred to as the *ileal bile acid transporter (IBAT)*.[53] ASBT demonstrates 35% amino acid identity and 63% similarity to the rat liver Na^+-coupled bile acid transporter.[53] Human IBAT cDNA cloning and population screening identified a mutation in a patient with Crohn's disease that abolishes taurocholate transport.[54] Cellular transport of bile acids in the ileal enterocyte is mediated by the ileal bile acid–binding protein (I-BABP), whose gene transcription is tightly regulated by altered bile acid flux through mechanisms involving the nuclear orphan receptor FXR.[55] Bile acid–dependent regulation of gene expression is discussed in detail elsewhere (see Chapter 16). Once delivered across the cytosolic compartment in association with I-BABP, the efflux of bile salts appears to involve transport through a truncated version of ASBT (t-ASBT) that arises through alternative RNA splicing.[56] Alternative RNA splicing results in skipping of exon 2 of the ASBT gene product and produces a frameshift in the alternatively spliced mRNA. This generates t-ASBT, a novel peptide of 154 residues that is expressed in the basolateral membrane of ileal enterocytes and cholangiocytes and functions as an efflux protein.[56]

Colonic Events

Unabsorbed long-chain fatty acids that enter the colon undergo a series of bacterial modifications, principally hydroxylation; they are not absorbed by this organ. In healthy persons, no undigested triglyceride is found in the stool, and the standard fecal fat estimate of approximately 7 g/d reflects the cumulative total excretion of saponification products (i.e., fatty acids) that arises principally from membrane phospholipid and bacteria. Estimates of normal triglyceride fractional absorption (>95%) likely underestimate the astonishing efficiency of this process.

Short-chain fatty acids are the principal aqueous solute in colonic contents, with a total concentration of 100 to 240 mmol/L; of these, over 90% are acetic, propionic, and *n*-butyric acids.[57] Short-chain fatty acids arise from bacterial fermentation of unabsorbed carbohydrate and may be an important energy source for colonocytes in health and disease.[58] Human fecal matter contains about 500 mg of bile salts and 200 to 500 mg of cholesterol and its bacterial degradation products, cholestanol

and cholestanone. [59] Bile salts are principally deconjugated and dehydroxylated in the colon, and in health, a balance is maintained between these daily fecal losses and de novo bile salt synthesis by the liver. [59]

Brush Border Membrane Events

The mechanism of long-chain fatty acid uptake across the intestinal microvillus membrane is incompletely understood. This subject is well covered in two reviews. [60], [61] Although a favorable concentration gradient likely exists for passive diffusion of these lipophils, biochemical evidence suggests the presence of a specific carrier. The cloning and characterization of a family of fatty acid transport proteins (FATPs) [62] led to the identification of FATP4, a protein of about 64 kd that is expressed at high levels in the apical membrane of mammalian enterocytes. [63] The cloning of this family of transporters has fulfilled earlier predictions of the biochemical and kinetic parameters of fatty acid uptake. FATP4 transfection of heterologous cells induces saturable, specific, and temperature- and substrate-dependent uptake of long-chain fatty acids, but not cholesterol or other hydrophobic sterols, suggesting that FATP4 likely is the elusive intestinal fatty acid transporter. [63] The role of other proteins previously thought to be involved in fatty acid transport across the brush border membrane, including the microvillus membrane fatty acid–binding protein (MVM-Fabp), which was subsequently identified as a mitochondrial isoform of aspartate aminotransferase, [64] must now be reexamined. Further information concerning the role of these FATP genes in fatty acid transport by the enterocyte will undoubtedly emerge from gene-targeting approaches.

Cholesterol uptake across the microvillus membrane of small intestinal enterocytes has been postulated to be protein mediated, although the identification of such transporters has been long sought. A breakthrough was the identification of an ATP-binding cassette (ABC)–type cholesterol transporter, ABCA1, whose function is defective in Tangier disease. [65], [66] and [67] Tissues of patients with Tangier disease (including the small intestine) accumulate cholesterol, and their serum contains very low levels of high-density lipoprotein (HDL) as a result of the failure of cholesterol to be transferred to an acceptor HDL particle in the extracellular fluid. ABCA1 is postulated to function as a cholesterol export pump, although whether it is uni- or bidirectional is unclear. In addition, the tissue- and cell-specific distribution of ABCA1 is somewhat controversial. Studies in mice suggest that ABCA1 mRNA is expressed and regulated in the intestine, [68] whereas in situ hybridization studies of baboon intestine suggest that ABCA1 mRNA is located in macrophages in the lamina propria. [69] Accordingly, intestinal expression of ABCA1 protein is yet to be demonstrated, and its membrane topology (i.e., brush border or basolateral) will have to be reconciled with any putative cholesterol transport function. Pending the resolution of these questions, other studies have demonstrated metabolic regulation of distinct components of cholesterol transport. Specifically, nuclear hormone receptors involved in cellular lipid metabolism, including oxysterol receptors (LXRs) that function as obligate partners of retinoid X receptors (RXRs), were demonstrated to regulate cholesterol absorption through the transcriptional induction of ABCA1 expression in the intestine. [68] Mice treated with a synthetic rexinoid, a ligand agonist of RXR, demonstrated up-regulation of ABCA1 expression in the intestine, with decreased cholesterol absorption. [68] Consistent with these observations, ABCA1 -/- mice show mild increases in cholesterol absorption. [70], [71] In contrast, another report suggests that ABCA1 -/- mice generated in a C57 genetic background manifest an increase in cholesterol absorption. [72] These findings considered together suggest as yet unresolved issues concerning an important new mechanism of cholesterol uptake by the human intestine. A further breakthrough in the understanding of brush border events regulating lipid uptake came with the cloning of yet another set of ABC-type transporters whose function is defective in humans with sitosterolemia. Affected individuals absorb large amounts of plant sterols, structurally related to cholesterol, whereas healthy subjects absorb less than 5% of these sterols. This presumably is a consequence of the ability of the enterocyte transporter to discriminate subtle differences in sterol structure. By using a combination of genetic linkage and fine mapping, the locus was narrowed, and candidate genes identified as half-transporters, referred to as *ABCG5* and *ABCG8*, were located on chromosome 2p21. [73], [74] The two genes are arranged in tandem and transcribed in opposite directions, each encoding a half-transporter. Accordingly, these ABC-type transporters are postulated to function as heterodimeric sterol exporters with the ability to discriminate cholesterol from other, related sterols. [73], [74] Investigators postulate that these transporters function in concert, but the relationship of ABCA1 to the function of the ABCG5 and ABCG8 transporters is unknown. Conceivably, these transporters participate in a coordinated manner whereby cholesterol and other sterols are discriminated and selectively transported across the membrane.

INTRACELLULAR EVENTS IN LIPID REASSEMBLY

Intracellular Transport of Long-Chain Fatty Acids

After brush border membrane uptake, long-chain fatty acids and monoglyceride must be translocated across the aqueous cytosolic compartments of the enterocyte and delivered to the smooth endoplasmic reticulum (ER) for incorporation into complex lipid. Directed intracellular trafficking is facilitated by fatty acid–binding proteins (Fabps), members of a larger supergene family that includes several vitamin A–binding proteins and other proteins whose functions are unknown. Mammalian enterocytes express mRNAs for two cytosolic Fabps, intestinal Fabp (Fabpi) and liver Fabp (Fabpl), [75] based on the organ from which each was initially isolated. The mRNAs encoding these proteins are highly abundant, accounting for 2% to 3% of the total intestinal translation products and representing 1% to 2% of the soluble cytosolic protein mass. [76], [77], [78] and [79] Predictions from the cDNA sequence [80], [81] suggest Fabpl is a 127-residue peptide with a molecular weight of 14,273 d, whereas Fabpi mRNA encodes a slightly larger peptide of 132 residues and a molecular weight of 15,124 d, with more than 80% nucleotide and amino acid homology between rat and human genes. [75], [80], [82] Human *FABPL* is located on chromosome 2, and *FABPI* on chromosome 4. [82] Both Fabps are synthesized as obligate intracellular peptides, neither being destined for export. The tissue distribution of the two cytosolic Fabps differs. Fabpi mRNA is essentially confined to the small intestinal enterocyte. When transcript abundance is normalized to that in the small intestine (100%), less than 8% of the small intestine signal is present in colon, and the stomach and the liver express 2% to 4%, respectively. [83] By contrast, Fabpl mRNA is expressed in the liver at 50% to 70% of the level found in the small intestine, and both the stomach and colon express 2% to 6% of the small intestine Fabpl mRNA. [83]

Regulation of the biosynthesis of intestinal and liver Fabps has been studied by means of various techniques. Earlier studies demonstrated that the cytosolic concentration of immunoassayable Fabp increases in a gradient from the villus to the crypt and from the proximal to the distal small intestine. [78] The developmental expression of the two cytosolic Fabps reveals a coordinated increase in Fabpl and Fabpi mRNA abundance in intestinal mucosa after birth, detectable at day 19 of gestation and peaking in adulthood. [83] Complementary approaches of cDNA hybridization and classical protein turnover kinetics have established that Fabpl mRNA abundance and translatable activity are higher in the jejunum of female than of male rats. [76], [77] However, the turnover of newly synthesized Fabpl in female rats is also faster than in male rats, producing no net effect on steady-state cytosolic Fabpl concentrations. There are no sex-related differences in the cytosolic concentration or translatable activity of Fabpi. Clofibrate, a hypolipidemic agent that affects hepatic fatty acid metabolism through peroxisome proliferator activator receptor-a, doubles the intestinal Fabpl mRNA level and protein concentration in the intestine but has no effect on Fabpi.

A consensus of data supports the hypothesis that the two cytosolic Fabps are independently regulated and probably play distinct roles in intracellular fatty acid metabolism. Both Fabps are expressed in *Escherichia coli,* and their structures and ligand-binding affinities have been determined. [84], [85] and [86] Fabpl has a binding capacity of 2 mol fatty acid per 1 mol protein, and Fabpi binds in 1:1 stoichiometry. Further studies have demonstrated that Fabpl exhibits a higher affinity for polyunsaturated fatty acids than for saturated species, and Fabpi exhibits a broadly similar affinity for saturated and polyunsaturated fatty acids. These data support the concept that intracellular Fabps may target substrate fatty acid to selective compartments for incorporation into triglyceride, phospholipid, or cholesteryl or retinyl esters. Fatty acids entering the enterocyte from the basolateral membrane (i.e., plasma derived) are thought to be metabolically distinct from fatty acids absorbed across the brush border. [87] The differences in ligand-binding stoichiometry, underlying mechanisms of conformational interaction, and pH sensitivity may be correlated with their distinctive physiological roles in enterocyte fatty acid trafficking. [84]

The *FABPI* locus was examined in Pima Indians as a possible basis for the linkage observed between markers on chromosome 4q and certain measures of insulin action. [88] A common genetic polymorphism in the coding region of *FABPI* was identified—that is, a single G-to-A transition generating an alanine-to-threonine substitution in the Fabpi protein. [89] The mutant protein demonstrated increased binding of long-chain fatty acids in vitro. [89] Furthermore, expression of the mutant protein in Caco-2 cells resulted in increased transport of both long-chain fatty acids and triglyceride. [90]

Analysis of the role of Fabpi through gene targeting in mice has yielded some surprising findings. Homozygous Fabpi -/- mice absorb fat normally, and male but not female animals gain weight at a faster rate than do wild-type animals when fed either a chow diet or a high-fat diet. [91] Plasma insulin levels were higher in Fabpi -/- mice of both sexes but were not associated with alterations in plasma glucose levels. Thus, the male Fabpi -/- phenotype shows some features of the Pima Indian phenotype previously described. Further study of the metabolic targets of Fabps will undoubtedly reveal new functions for these abundant genes. The fatty acid transport function of these genes has been reviewed recently elsewhere. [92]

Intracellular Transport of Sterols

Intestinal absorption and metabolic processing of dietary and biliary sterols—principally cholesterol—is a major component of the body's homeostatic control mechanisms for regulating sterol uptake. The intestine is the portal of entry of the fat-soluble A, D, E, and K vitamins, and elaborate conservation mechanisms have evolved to ensure constant availability of these essential nutrients, particularly vitamin A.

From the perspective of regulating intracellular cholesterol traffic, the enterocyte receives most of its daily flux of cholesterol from lumenal sources (i.e., dietary and biliary), but it is also capable of de novo cholesterol synthesis from 2-carbon units. The intestinal cell also expresses low-density lipoprotein (LDL) receptors on its basolateral membrane, which facilitate the endocytotic uptake of circulating plasma LDL. [93] The interactions of these sources of intestinal cholesterol and their role in regulating cholesterol metabolism in the enterocyte have been studied extensively.

Cholesterol delivery to various intracellular locations is facilitated by means of its 1:1 stoichiometric association with a specific carrier protein, sterol carrier protein 2 (SCP2). [94] SCP2 was first isolated from rat liver as a 13-kd protein that participated in the intracellular movement of cholesterol within hepatocytes and steroidogenic tissues. [94], [95] Cloning the rat and human SCP2 genes revealed substantially more information. [96], [97], [98] and [99] The rat gene spans 20 to 30 kilobases (kb) and is transcribed into four distinct mRNA species that encode a 58-kd protein, SCPx, and a smaller, about 14-kd protein, SCP2. [98] SCP2 is expressed in rat small intestine enterocytes [100] and colocalized to the subapical region with the peroxisomal marker PMP70, suggesting that SCP2 is indeed a peroxisomal protein, not a mitochondrial protein, as previously thought. [101] SCP2 gene expression in the rat is subject to tissue-specific and developmental regulation with a temporally distinct pattern of increase in SCP2 mRNA abundance in the liver and small intestine in the neonatal period. [102] The function of SCP2 may involve peroxisomal catabolism of methyl-branched fatty acyl-CoA, as inferred from the study of gene-targeted mice. [103] These mice demonstrated alterations in peroxisomal morphology associated with hypolipidemia and neuropathy with accumulation of phytanic acid. Other studies have suggested roles for SCP2 in cholesterol efflux [104] and biliary cholesterol secretion. [105]

Intracellular Transport and Metabolism of Vitamin A

All mammals require vitamin A for effective cell growth and differentiation. The two major sources of this nutrient are carotenoids, principally ß-carotene from plants, and retinyl esters derived from animal tissues. Carotenoids must be converted, after uptake into the enterocyte, into retinol. This conversion is a two-step procedure involving cleavage by 15,15'-dioxygenase to yield two molecules of retinaldehyde, which is subsequently reduced to retinol. [106] Dietary retinyl esters must first undergo hydrolysis to retinol before absorption, and although details are lacking, the evidence suggests that ß-carotene and retinol are absorbed into the enterocyte by passive diffusion.

The intestine synthesizes specific binding proteins that provide a mechanism by which cellular vitamin A is stored and delivered for esterification before secretion in the form of retinyl ester. There are at least four intracellular cellular vitamin A–binding proteins in mammals, including cellular retinol-binding protein I (CRBP I), cellular retinol-binding protein II (CRBP II), and cellular retinoic acid–binding protein types I and II (CRABP I and CRABP II). [107] These intracellular vitamin A–binding proteins are distinct from the serum retinol-binding protein RBP. [108]

The genes that encode these proteins form part of a supergene family that has at least 11 known member products, including the Fabps, adipocyte 422 protein, and myelin P2 protein. The two principal intracellular vitamin A–binding proteins of relevance to the intestine are CRBP I and CRBP II. These two peptides exhibit extensive amino acid and nucleotide homology, and the genes for both are located on human chromosome 3. [109] The tissue distribution of the two mRNAs is distinct, with CRBP II gene expression virtually confined to the enterocyte and CRBP widely distributed among vitamin A–responsive cells; CRBP immunoreactivity in the gut is confined to the submucosa and lamina propria. [110], [111]

CRBP II is abundant, representing about 1% of the soluble protein in rat jejunum and 0.4% of the soluble protein in human proximal small bowel. [112] The highest levels are detected in villi of the proximal small bowel, illustrating a striking gradient vertically (i.e., villus to crypt) and horizontally (i.e., jejunum to ileum). Studies further suggest that CRBP II undergoes developmental regulation, with peak mRNA abundance occurring just before birth, at 21 days' gestation in the rat. [111] This finding is compatible with the observation that cellular retinol flux undergoes changes along the villus-crypt axis. Cloning and sequencing of the retinoic acid receptor gene suggest that this new member of the steroid-thyroid hormone receptor gene family may function as a vertebrate morphogen. [113] Further elucidation of the structure of its intestinal homolog may provide important insights into the factors controlling cellular maturation and differentiation. Gene-targeting studies should provide conclusive information concerning the role of CRBP II.

Retinol is secreted by the enterocyte in the form of retinyl ester, predominantly in the hydrophobic core of triglyceride-rich lipoproteins. The mechanism by which intestinal retinol undergoes esterification has been the subject of considerable debate. Potential mechanisms include the reversible action of pancreatic cholesterol esterase, microsomal conversion using acyl-CoA:cholesterol acyltransferase 1 or 2 (ACAT1 or ACAT2), or acyl-CoA:retinol acyltransferase (ARAT). [114] The latter two enzymes have been favored because of the observed fatty acid specificity of the esterification reactions as evidenced by gas-liquid chromatographic analysis of human and rat lymph retinyl esters. [106] The role of ACAT enzymes is discussed further in the next section.

When retinol is presented as a complex with CRBP II, microsomes are able to use endogenous acyl donors and synthesize retinyl esters with a fatty acid composition strikingly similar to that described for rat lymph. [51], [115] Moreover, retinol–CRBP II complexes are unavailable for acyl-CoA–directed esterification, but retinol complexed to albumin is effectively esterified in an acyl-CoA–driven reaction. The rates of retinyl ester production by rat intestinal microsomes using this acyl-CoA–independent mechanism are calculated to be more than 1 µmol/d, which is more than sufficient to meet physiological requirements. These observations suggest that a major function of CRBP II may be to direct intestinal retinol to an appropriate milieu for esterification. [51], [115] Retinol targeted to a non–acyl-CoA–driven reesterification process would permit distinct metabolic compartmentalization. Although direct evidence is lacking, this mechanism could obviate competition between retinol and other intracellular lipid components, such as cholesterol for acyl-CoA donors.

Intestinal Cholesterol Metabolism

Dietary and biliary sources provide the 1 to 2 g of cholesterol that enters the intestinal lumen each day. Biliary cholesterol is essentially all free sterol, but 10% to 20% of dietary cholesterol is cholesteryl ester that must undergo hydrolysis before absorption. As previously discussed, intestinal cholesterol uptake is likely to involve one or more ABC-type transporters (ABCA1, ABCG5, and ABCG8) whose interactions may regulate the net influx of sterols. Membrane cholesterol transport may require interaction with protein components capable of mediating specific interactions with sterol acceptors, such as SCP2. From the brush border, cholesterol is transported, presumably by SCP2 and other acceptor proteins, to the smooth ER for reesterification. Although up to 25% of cholesterol may be secreted from the enterocyte and transported in mesenteric lymph as free cholesterol, most intracellular cholesterol destined for export undergoes reesterification.

Cholesterol esterification in the intestine is mediated by one of two ACAT enzymes. ACAT1 is expressed in numerous sites, with high levels in the adrenal cortex, macrophages, and sebaceous glands. [116] Intestinal cholesterol absorption, as well as hepatic and intestinal ACAT activity, were normal in the ACAT1 -/- mice, suggesting that another enzyme may function in the intestine and liver. [117] As suspected, a second ACAT gene (ACAT2) was subsequently identified whose expression is limited to the intestine and liver and whose function has recently been suggested through gene targeting in mice. [118] These mice demonstrated reduced cholesterol absorption and were protected against dietary cholesterol-induced hypercholesterolemia and cholesterol cholelithiasis. The intestine and liver of these animals also failed to accumulate cholesteryl ester when challenged with a dietary cholesterol load. [118] The results of experiments with ACAT2 -/- mice suggest that hepatic and intestinal cholesterol metabolism is regulated by distinct cholesterol-esterifying enzymes compared to other tissues.

The available evidence suggests that several discrete metabolic compartments exist for intestinal cholesterol. Absorbed lumenal (i.e., dietary and biliary) cholesterol appears to regulate ACAT2 activity and may be the preferred substrate. Newly synthesized cholesterol plays an important role in delivering cholesterol for cell membrane biosynthesis, perhaps in part related to growth needs but distinct from the diet-related export demands of the enterocyte. The factors that regulate LDL internalization and the role that this source plays in the traffic of intracellular cholesterol are unresolved and together form a focus of active investigation.

Intestinal Triglyceride Synthesis and Metabolism

Triglyceride is the most abundant lipid processed by the intestine and is almost completely absorbed by the enterocyte after intralumenal lipolysis. Approximately 75%

of absorbed lumenal fatty acids are resynthesized into triglyceride and secreted by the enterocyte as lipoprotein. [119], [120] The remaining 25% serve as substrate for phospholipid and other intracellular lipid resynthesis events or undergo transport to the liver bound to albumin through the portal vein. [50], [121], [122] The intestine also synthesizes and secretes triglyceride during fasting, with estimates of 10% to 40% of circulating plasma triglyceride being derived from the intestine after an overnight fast. [123]

There are two major pathways for intestinal triglyceride synthesis. The glycerol-3-phosphate pathway is the major route during periods of limited availability of lumenal monoglyceride and fatty acid. [123], [124] The substrates for triglyceride synthesis through this pathway, glycerol-3-phosphate and lysolecithin, are derived from glucose metabolism (i.e., dihydroxyacetone phosphate) and biliary phospholipid, respectively. [123], [125] The importance of biliary phospholipid as a substrate for triglyceride synthesis is highlighted by the fact that 75% of the fatty acids in infused lecithin are incorporated into secreted triglyceride during fasting. [126]

Enzymes catalyzing triglyceride synthesis through the monoacylglycerol pathway are located in the smooth ER, in contrast to the enzymes of the glycerol-3-phosphate pathway, which are located in the rough ER. [127], [128] Although the liver also contains monoacylglycerol acyltransferase, the intestinal form of the enzyme appears to be distinct based on substrate specificity, thermolability, and susceptibility to various detergents. [129] The intestinal enzyme is stereospecific in that almost 90% of 2-monoacylglycerol is converted into 1,2-diacylglycerol. [130] The molecular cloning of acyl-CoA:diacylglycerol acyltransferase (DGAT) has provided a new avenue for the investigation of triglyceride synthesis. [131] Structural and biochemical studies of DGAT were hitherto impossible because DGAT is an integral membrane protein. With the availability of reagents, these and other questions, including the tissue- and cell-specific distribution (ubiquitous with high levels in liver and intestine), were addressed. [131] After DGAT was cloned, its function was established by gene targeting in mice. The DGAT -/- mice demonstrated both resistance to obesity and defective lactation. However, intestinal triglyceride absorption was normal, although the livers of mice fed a high-fat diet accumulated less triglyceride. [132] These findings imply the presence of another DGAT enzyme involved in triglyceride synthesis in certain tissues, including the small intestine, perhaps analogous to the multiple ACAT enzymes that mediate cholesterol esterification.

Intestinal Phospholipid Synthesis and Metabolism

Phospholipids, particularly phosphatidylcholine (i.e., lecithin), are essential components of cell membranes. Lecithin is also a key component of lipoproteins, providing a surface coat for the inner core of triglyceride and cholesteryl ester. There are two major synthetic pathways for phospholipid in the enterocyte: the phosphatidic acid–phosphorylcholine pathway and the reacylation of absorbed lysolecithin. [126], [133], [134] and [135] Phosphatidylethanolamine is methylated in a third pathway in the liver, but this mechanism is not important in the intestine. [136]

The quantitative importance of either pathway for intestinal phospholipid synthesis is unknown. [137], [138] Lysophosphatidylcholine acyltransferase is more responsive to lipid feeding than choline phosphotransferase, suggesting that the lysolecithin reacylation pathway may be more important postprandially. [139] During periods of fasting and low levels of lumenal lecithin, the phosphatidic acid–phosphorylcholine pathway is the main synthetic route. [126], [138] Postprandially, with higher levels of biliary and dietary lysolecithin available, there is increased synthesis of lecithin through the reacylation pathway. During very high rates of experimental lecithin infusion into the lumen of the rat intestine, lymphatic chylomicron-phospholipid is exclusively derived from the infused lecithin through the reacylation pathway. [138]

INTESTINAL LIPOPROTEIN ASSEMBLY AND SECRETION

Enterocyte Morphology and Pathways of Lipoprotein Assembly

The morphology of the intracellular events occurring during lipid absorption has been studied extensively in human and rat intestine (Fig. 18-3). [140], [141] and [142] Serial electron micrographs reveal that large droplets of lipid form near the microvillus membrane 5 to 10 minutes after lipid instillation into the intestinal lumen. These droplets represent resynthesized triglyceride in and adjacent to the smooth ER. The overall process of chylomicron assembly and secretion is coordinated through the sequential action of several co-dependent steps (Fig. 18-4). The enzymes involved in triglyceride and phospholipid synthesis are located in the smooth and rough ER. [137] There may be metabolically distinct pools of intracellular triglyceride, one pool destined for chylomicron secretion derived from the 2-monoglyceride pathway, and another, not destined for immediate export, derived from the phosphatidic acid pathway. [122] In addition, there is evidence for the sequential assembly of chylomicrons and their compartmentalization based on different transport rates through the secretory apparatus. [143], [144]

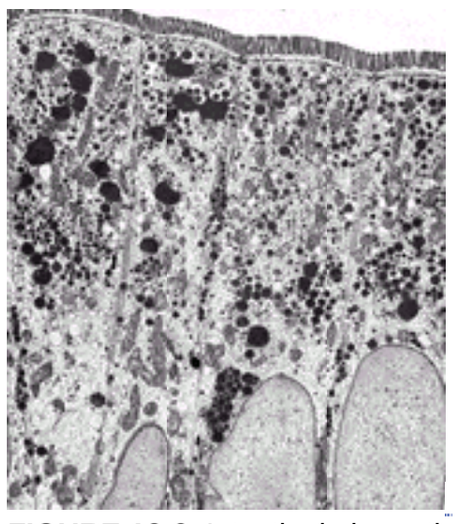

FIGURE 18-3. Intestinal absorptive cells after lipid feeding. This electron micrograph, taken 2 hours after lipid feeding, shows the apical cytoplasm to be engorged with osmiophilic droplets bounded by smooth endoplasmic reticulum. Golgi zones and intracellular spaces contain chylomicron-size particles. (Original magnification ×8000; adapted from ref. [141].)

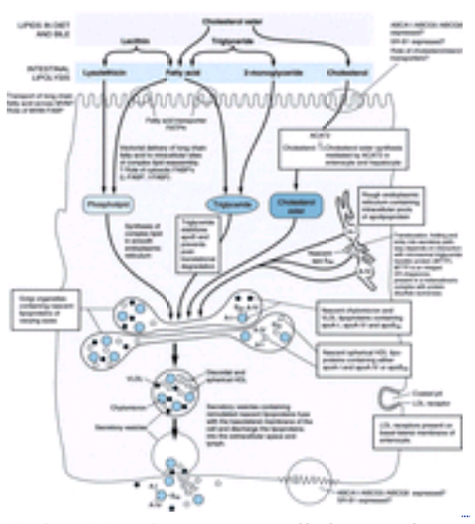

FIGURE 18-4. Intracellular pathways of intestinal lipoprotein assembly. Lumenal lipids are absorbed across the microvillus membrane and transported to the smooth endoplasmic reticulum, where they serve as substrates for the synthesis of triglyceride, lecithin, and cholesteryl ester—the lipids of nascent lipoproteins.

Apolipoproteins from the rough endoplasmic reticulum are mobilized during lipoprotein formation onto lipid to form nascent lipoproteins in Golgi organelles. Lipoproteins are remodeled in the Golgi organelles and the secretory vesicles. These secretory vesicles fuse with the basolateral membrane of the enterocyte and release the nascent lipoproteins into the lymph.

Apolipoproteins synthesized in the rough ER are transferred onto newly synthesized lipid, as evidenced by the decrease in apo B immunostaining in the rough ER and a corresponding increase in apo B immunostaining in the smooth ER after lipid feeding. [145] The distribution of apolipoproteins within the enterocyte is such that during fasting, about 90% of apolipoprotein is not bound to lipoprotein, but remains associated with the ER. [146] Lipid feeding mobilizes 5% to 10% of this apolipoprotein from the non–lipoprotein-bound pool onto newly assembled lipoproteins. [147] The mechanism by which apolipoproteins associate with lipids to form nascent intracellular lipoproteins is likely to involve the microsomal triglyceride transfer protein (MTTP). Other studies have suggested that microtubules may be involved in intracellular lipid transport. [148]

The Golgi apparatus membranes, which are located close to the nucleus of the cell, become filled with lipid droplets corresponding in size to chylomicrons and very-low-density lipoprotein (VLDL)–like particles after lipid feeding (see Fig. 18-3 and Fig. 18-4). In the liver, lipid and apolipoproteins are added to nascent lipoproteins in both the ER and the Golgi organelles (Fig. 18-5), and similar remodeling probably occurs in the intestinal enterocyte. [149] Golgi organelles are thought to be precursors to secretory vesicles, which are larger vesicles filled with lipid droplets of various sizes that represent nascent forms of intracellular lipoproteins before secretion. [142] These secretory vesicles fuse with the basolateral membranes and release nascent lipoproteins into the extracellular space and mesenteric lymph. [141], [142] The limiting rate during chylomicron formation and secretion appears to be the rate at which lumenal triglyceride is absorbed, not the rate of apolipoprotein synthesis. [146], [150] Secretion rates of apolipoprotein in the proximal and distal rat small intestine appear to be similar, although distal intestine has been shown to transport lipid more slowly than proximal intestine. [150] After lipid infusion directly into human jejunum and ileum, no morphologic differences between enterocytes from the two sites are seen, suggesting similar fat absorption and lipid secretion in the proximal and distal bowel. [151] Functional differences, however, may exist in the capacity of the ileum to transport large quantities of lipid, as inferred from the reduced expression of most genes involved in lipid transport and lipoprotein assembly.

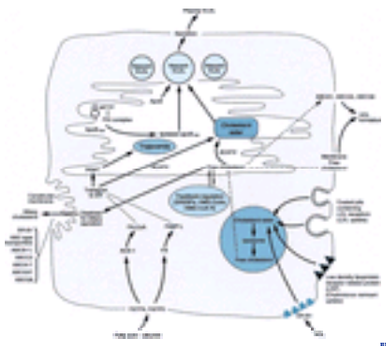

FIGURE 18-5. Intracellular pathways of hepatocyte lipoprotein assembly and secretion. Several distinct pathways are involved in the complex regulation of hepatic lipid flux. Fatty acid uptake by the hepatocyte is carrier mediated and results in fatty acid delivery to acyl-CoA synthase (ACS-1) and also to liver fatty acid–binding protein (Fabpl). Fatty acids are delivered to the endoplasmic reticulum (ER) for assembly into complex lipid (cholesteryl ester and triglyceride). Key enzymes involved in these reactions include acyl-CoA:cholesterol acyltransferase 2 (ACAT2) and acyl-CoA:diacylglycerol acyltransferase (DGAT), respectively. The newly synthesized triglyceride is transferred to the nascent apo B protein by the microsomal triglyceride transfer protein (MTTP), which is an ER chaperone. MTTP exists bound to protein disulfide isomerase and is required for lipidation of apo B. These particular features of hepatic lipoprotein assembly are indistinguishable from those in the small intestine. Other apolipoproteins are added to the nascent very-low-density lipoprotein (VLDL) particle, including apo E, which is not expressed in the enterocyte. The VLDL particle is directed through the Golgi apparatus for secretion into plasma. Cholesterol entry into the hepatocyte involves lipoprotein uptake by several different receptors. In humans, most lipoprotein cholesterol is carried by low-density lipoprotein (LDL), and the LDL receptor accounts for most receptor-dependent cholesterol uptake. Other receptors are shown to indicate their role in high-density lipoprotein (HDL) uptake: class B scavenger receptor (SR-B1) and LDL receptor–related protein (LRP), chylomicron remnants. In addition, several members of the ABC transporter family likely play a role in cholesterol secretion, both into bile and into the systemic circulation. However, it is as yet unclear whether the expression of these HDL transporters (ABCA1, ABCG5, and ABCG8) is polarized in the hepatocyte and what if any role they play in the delivery of cholesterol into bile.

As alluded to previously, there are many similarities in intestinal and hepatic lipoprotein assembly and secretion, but also important points of divergence. These are summarized in Figure 18-5.

Regulation of Intestinal Apolipoprotein Gene Expression

Apolipoproteins are the protein components of lipoproteins, the vehicles by which water-insoluble lipid is transported through the aqueous medium of plasma. Lipid secretion from the enterocyte is largely in the form of chylomicrons and VLDLs. The liver secretes VLDLs but not chylomicrons (see Fig. 18-5), for reasons that are incompletely understood. Both chylomicrons (>100 nM) and VLDLs (30–90 nM) are large, lipid-rich particles that transport neutral lipid from its site of synthesis or reassembly to other tissues for use as an energy substrate or for the delivery of cholesterol. VLDLs and chylomicrons have a distinctive pattern of apolipoproteins associated with their surface, apolipoproteins that play a large role in directing the particles to the appropriate receptors and enzymes involved in their metabolism.

There are at least six apolipoprotein genes, whose presence in the small intestine has been demonstrated by immunologic detection of the final gene product or by hybridization to an appropriately sized transcript (Table 18-1). Of these apolipoprotein genes, three are expressed abundantly (i.e., apo A-I, apo A-IV, apo B), and these are discussed in this chapter. For further information, the reader is referred to excellent reviews of the molecular genetics of apolipoproteins and the structure-function and evolutionary aspects of apolipoprotein genes. [152], [153], [154], [155] and [156]

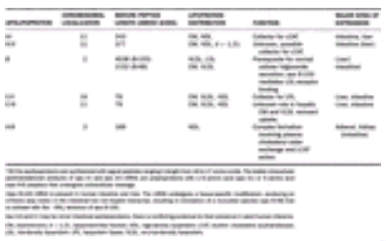

TABLE 18-1 Human Intestinal Apolipoproteins*

Apolipoprotein gene structure in mammals shows a high degree of conservation, with four of the six genes expressed in human small intestine showing distinct similarities; the structure of the apo B gene and predicted structure of the apo A-IV gene are quite different. The major apolipoproteins demonstrate such structural conservation that they are predicted to have arisen from a common ancestral gene. [157] The basic organization is a four-exon, three-intron gene structure; the exception is apo A-IV, which lacks an intron in the 5' noncoding region of the corresponding mRNA. [158] The observed structural similarity in the genes reflects structural conservation in the gene products, in which multiple repeats of 22 amino acids confer a marked a-helical amphipathic secondary structure that contributes to their essential function as lipid-binding proteins. [159], [160] Like most proteins destined for export, all the intestinal apolipoproteins are synthesized with signal peptides that are cotranslationally cleaved, and the stable posttranslational forms of apo A-I and apo A-II mRNA contain an additional prosegment that undergoes extracellular

cleavage. [157], [161], [162]

Apo A-I is an abundant intestinal mRNA that codes for about 1% to 2% of newly synthesized protein in the rat. [161] In humans, apo A-I is the major protein component of HDL and is the principal cofactor for the enzyme lecithin:cholesterol acyltransferase (LCAT), which is responsible for plasma cholesterol esterification. [163], [164] Several studies suggest that the intestine synthesizes about half of the body's daily input of this apolipoprotein. [165] Attention given to the importance of HDL as a protective factor against atherosclerosis and evidence demonstrating that ambient serum levels of apo A-I may be an even more potent predictive factor have focused attention on factors that influence the biosynthesis of apo A-I. [166]

Studies of the rat suggest that apo A-I is synthesized throughout the small intestine, with the highest levels produced in the proximal jejunum and a nadir reached in the terminal ileum. [167] Additional rat studies have demonstrated that intestinal apo A-I synthesis and mRNA content are unaltered after acute triglyceride feeding or after 3 to 6 weeks of sustained exposure to diets containing widely discrepant quantities, 0% to 30% by weight, of triglyceride. [168] Intestinal apo A-I synthesis does not respond to diets composed of butter fat or corn oil, and dietary cholesterol augmentation has no effect, suggesting that, at least in this species, intestinal apo A-I gene expression appears to be largely constitutive. Apo A-I synthesis demonstrates regional sensitivity to the removal of biliary lipid components, with no effect observed in the jejunum but marked suppression in the ileum. [167] The molecular basis and physiological consequences of this regional heterogeneity are unknown.

Intestinal apo A-I gene expression undergoes marked induction during development, with an increase in mRNA abundance occurring at birth, presumably with the onset of suckling; subsequent mRNA levels appear to remain relatively constant until adulthood. [161] Intestinal apo A-I gene expression may be sensitive to alterations in thyroid hormone status, with such changes mediated at a translational or posttranslational level. [169]

Apo A-IV expression constitutes another abundant intestinal mRNA species, coding for as much as 3% of newly synthesized protein. [170] Its distribution in plasma differs from that of apo A-I and all the other members of this gene family in that it circulates largely unbound to lipoproteins, although about 25% of the plasma apo A-IV in humans is associated with HDL. [163] Its regulation within enterocytes is also distinct from that of apo A-I, as it is responsive to cellular triglyceride flux. [167], [171] The synthesis rates and mRNA abundance of jejunal and ileal apo A-IV double within 4 to 6 hours of ingestion of a fat bolus, and under fasting and postprandial conditions, the proximal to distal gradient of synthesis rates and mRNA abundance is maintained. [171] Further studies demonstrated striking developmental induction of intestinal apo A-IV mRNA abundance, with a greater than 20-fold increase occurring at birth, followed by a gradual decline over the ensuing 14 days. [172] The intestine is the major synthetic and secretory source of apo A-IV, and although no specific function has yet been attributed to this protein, studies suggest that it may act as a potential cofactor for LCAT, although less effectively than apo A-I. [173] Studies in rats have suggested that apo A-IV may be a factor in regulating food intake, and in this regard, its effects appear to be centrally mediated. [174] Transgenic mice expressing high levels of human apo A-IV in the intestine demonstrated no alterations in food intake. [175] Another promising area for investigation was suggested by the demonstration that human apo A-IV transgenic mice bred into the atherosclerosis-susceptible apo E–deficient strain were protected against atherosclerosis. [176]

Apo C-III is a minor intestinal apolipoprotein whose mRNA codes for less than of 0.5% total protein synthesis. [177] Its gene is localized on chromosome 11 between and in a reverse orientation to the apo A-I and apo A-IV genes. [178] Apo C-III probably functions in regulating the hepatic uptake of remnant lipoprotein particles. Studies have demonstrated that, like apo A-IV, apo C-III is regulated after increases in intestinal triglyceride, with accumulation of mRNA and its translational product. [177], [178]

Apo A-II, apo C-I, and apo C-II are minor intestinal apolipoproteins; their regulation has not been studied extensively. [170], [179], [180] Apo D mRNA codes for a protein found in human plasma in association with HDL, whose function is partially understood. This mRNA has been demonstrated in human intestine, [181] but its regulation mechanism is unknown in the gut.

Apo B is synthesized in mammalian enterocytes and hepatocytes as an obligate component of triglyceride-rich lipoproteins (e.g., chylomicrons, VLDL). Based on a centile score in human plasma, two major molecular forms of apo B are found: apo B-100 (~512 kd) and an unglycosylated form, apo B-48 (~241 kd). [182] These isoforms are synthesized in an organ-specific fashion as a result of posttranscriptional RNA editing (described in the following paragraph). Apo B plays an essential role in the assembly of lipoproteins within the liver and small intestine. In addition, apo B-100, the major protein component of circulating plasma LDLs, expresses a domain in its carboxyl terminus that mediates the high-affinity interaction with the LDL receptor and plays a central role in cholesterol catabolism. [183], [184] Because this domain is absent from apo B-48, intestinal lipoproteins such as chylomicrons, which contain apo B-48, are catabolized through a different receptor, referred to as the *chylomicron remnant receptor*. [152]

Mammalian apo B is the product of a single gene that has been mapped in humans to chromosome 2. [152] Its genomic organization is distinct from that of all other apolipoprotein genes. Apo B has 29 exons; exon 26 spans 7572 bases, so that it is one of the largest mammalian exons studied. [152], [183] The underlying mechanism for the organ-specific production of apo B-100 and apo B-48 has been the focus of considerable study. A single apo B gene is transcribed into a 14-kb mRNA in the liver and small intestine. In the liver, this apo B mRNA contains a genomically templated CAA codon that encodes a glutamine residue in apo B-100. In the small intestine, apo B is transcribed into an mRNA in which a site-specific deamination of the cytidine residue in this codon produces a UAA or translational stop. [157], [185] As a result of posttranscriptional RNA editing, intestinal apo B mRNA is translated as a truncated protein that is colinear with the amino-terminal 48% of apo B-100. [157], [185] Apo B mRNA editing occurs in the human small intestine but not in the liver. [157], [185], [186] In contrast, rat and mouse livers contain edited and unedited species of apo B mRNA and secrete apo B-100 and B-48. [187] Apo B mRNA editing is developmentally regulated in the human small intestine and is subject to hormonal and nutritional modulation in the rat liver. [187], [188] and [189] The functional consequence of intestinal apo B mRNA editing is that intestinal chylomicrons (apo B-48) are cleared from the circulation within 30 minutes, and LDL particles (e.g., apo B-100) have a half-life longer than 2 days. [190] Apo B mRNA editing can thus be viewed as an adaptive mechanism for the rapid delivery of intestinal triglyceride to the liver.

The underlying molecular mechanisms have been elucidated and involve protein factors that mediate site-specific deamination in the context of a distinct AU-rich RNA motif. [188] The catalytic subunit of the apo B mRNA editing enzyme, apobec-1, has been cloned and is expressed exclusively in the human small intestine. [191], [192] Apobec-1 knock-out mice phenotypically express exclusively apo B-100. [193] There was no apparent defect in intestinal lipid absorption, suggesting that apo B-100 and apo B-48 can function interchangeably as regards intestinal lipoprotein assembly and secretion. [193], [194]

Despite its evident requirement in the assembly and secretion of triglyceride-rich lipoproteins, intestinal apo B synthesis is not regulated in response to dietary triglyceride augmentation but may be regulated by various components of bile. [195], [196] Intestinal apo B synthesis is decreased in the rat after removal of biliary lipid and reexpressed after introduction of components such as fatty acids and bile salts. The postulated mechanism appears to be the provision of substrate (i.e., fatty acid) for microsomal triglyceride assembly that presumably reaches a threshold level at which intestinal apo B synthesis is reexpressed. Augmentation of the triglyceride flux above this putative threshold produces no further change in apo B synthesis. Regulation of apo B synthesis and posttranslational stability in the context of triglyceride availability has been reviewed. [197]

The developmental regulation of apo B gene expression in the rat is different from that of apo A-I or apo A-IV. [198] After an increase at birth, apo B levels decline until day 8, when a second increase occurs, followed by a second decline to a nadir at age 24 to 35 days. After the second decline, expression increases to adult levels. Apo B RNA editing also is developmentally regulated in mammalian small intestine. [186], [187], [189], [199]

The major evidence that apo B is essential to the process of triglyceride-rich lipoprotein secretion has been derived from studies of humans with syndromes of defective chylomicron assembly and secretion and abnormal apo B gene expression (Table 18-2). The prototype of such syndromes is abetalipoproteinemia, an autosomal recessive condition characterized by mild fat malabsorption and several systemic abnormalities, including ataxia, acanthocytosis, and retinitis pigmentosa. [200]

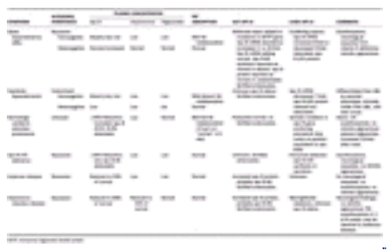

TABLE 18-2 Syndromes of Defective Chylomicron and Very-Low-Density Lipoprotein Secretion

Parents of the affected persons are obligate heterozygotes but manifest normal levels of plasma apo B and no evidence of fat malabsorption. Conclusive studies demonstrated that the molecular basis for abetalipoproteinemia is a defective MTTP gene. [201] Earlier studies of the intestine of two homozygous patients with abetalipoproteinemia revealed an absence of immunoreactive apo B, a finding in contrast to the report of quantitatively normal amounts of immunoreactive apo B-48 (and apo B-100) epitopes in another patient with this disorder. [202], [203] The molecular heterogeneity of abetalipoproteinemia is exemplified by divergent reports of absent or normal apo B-48 synthesis by intestinal explant cultures. [204], [205] Other studies of the livers of two patients with abetalipoproteinemia revealed increased amounts of apo B mRNA, findings that should be contrasted with other work reporting fivefold decreased levels of apo B mRNA. [206], [207] Studies of intestinal apo B mRNA in abetalipoproteinemia have demonstrated an increased abundance of a normally edited transcript. [206], [208] The most reasonable conclusion is that abetalipoproteinemia is a heterogeneous condition in which triglyceride accumulation within hepatocytes and small intestinal cells is accompanied by inconsistent alterations in apo B gene expression. It bears emphasis that apo B mRNA abundance was unaffected in a murine model of abetalipoproteinemia generated through conditional gene targeting of the *Mttp* allele in embryonic stem cells. [209] The molecular basis for abetalipoproteinemia is different from a genetically distinct entity referred to as *hypobetalipoproteinemia*. [210], [211] The distinguishing features in hypobetalipoproteinemia include autosomal codominant inheritance and low plasma apo B levels in obligate heterozygotes. Studies have shown a sevenfold decrease in hepatic apo B-100 mRNA levels, compared with a sixfold increase in abetalipoproteinemia and a reduced intracellular apo B-100 mass. [211] Hypobetalipoproteinemia may represent a translational abnormality with an unstable or an abnormal apo B mRNA.

Patients with several other syndromes have various degrees of fat malabsorption and are unable to assemble and secrete triglyceride-rich lipoproteins from enterocytes or hepatocytes. These syndromes are summarized in Table 18-2 and collectively form the basis for investigating the molecular details of apo B assembly and processing and its requisite involvement in chylomicron secretion. [200], [201], [202], [203], [204], [205], [206], [207] and [208], [210], [211], [212], [213], [214], [215], [216], [217] and [218]

Characterization of Intracellular Intestinal Lipoproteins

Lipoproteins are multimolecular aggregates of lipid and protein, with a configuration that allows the transport of intensely hydrophobic lipid through the aqueous milieu of cellular cytosol and plasma compartments. The apolar lipid core of the particle contains triglyceride and cholesterol ester, and the surface coat is composed of cholesterol, lecithin, and apolipoproteins. The intestine assembles and secretes chylomicrons, VLDL, HDL, and perhaps LDL (see Fig. 18-4). Chylomicrons (80–500 nm) are assembled during periods of lipid absorption and have been isolated from within intestinal Golgi organelles. Intracellular chylomicrons are composed of 13% to 25% protein and 75% to 85% lipid, two thirds of which is triglyceride. [146], [168], [219] In contrast, extracellular chylomicron composition in lymph and plasma is greater than 95% lipid, 90% of which is triglyceride. [220], [221] The major apolipoproteins on the surface of intracellular chylomicrons are apo B-48, apo A-I, and apo A-IV. These proteins are synthesized within the enterocyte.

VLDL-size particles (25–80 nm) are assembled in the enterocyte during fasting and lipid feeding. Apolipoproteins comprise about 15% of intracellular VLDL—apo B-48, apo A-I, and apo A-IV. As with chylomicrons, the lipid moiety of VLDL is largely triglyceride and phospholipid. [219] The compositional resemblance of chylomicrons and VLDL suggests that these particles represent a size continuum of the same type of lipoprotein whose volume depends on the availability of intracellular triglyceride. [222] There appear to be separate pathways for chylomicron and VLDL assembly, based on the demonstration of chylomicrons and VLDLs in different Golgi organelles, together with studies showing selective inhibition of chylomicron secretion despite persistent VLDL secretion. [223], [224]

Lipoprotein particles of the size of LDL have been isolated from within enterocytes and partially characterized. [146] The particles contain newly synthesized apo A-IV and apo B and trace amounts of apo A-I. The lipid composition is 40% phospholipid and 31% triglyceride. In rats given galactosamine, which profoundly reduces the hepatic synthesis of lipoproteins, there appear to be intestinally derived particles of the size of LDL in mesenteric lymph. [225] However, these data should not be construed to indicate that the intestine is normally a source of plasma LDL.

HDL particles, although too small to be seen in electron micrographs of unfractionated enterocytes, have been isolated within Golgi vesicles of rat enterocytes. [226] Intracellular HDL particles have been visualized as spherical 6- to 13-nm particles. Compositional analysis has revealed two distinct populations, one containing apo A-I and A-IV and the other containing apo B-48 as the surface apolipoprotein. The particles are composed of 70% protein and 30% lipid, one half of which is phospholipid. Whether the HDL particles that contain apo B-48 are metabolic precursors to chylomicrons is unknown, as is their fate.

Mesenteric lymph contains a population of discoidal and spherical HDL particles that are secreted from the intestine. [227], [228] Discoidal particles make up about 50% of fasting levels of lymph HDL; these particles are enriched in apo A-I and phospholipid relative to plasma HDL. Apo A-I on these HDL particles is metabolically derived by de novo intestinal synthesis. Metabolic labeling experiments in rats provide good evidence that discoidal HDL particles are synthesized within enterocytes and do not simply reflect lipolytic products of triglyceride-rich lipoproteins or ultracentrifugation artifacts. [227] Spherical lymph HDL particles are also relatively protein- and phospholipid-enriched compared with plasma HDL particles, and they contain core cholesteryl ester that appears to have an intestinal source (i.e., ACAT derived) rather than a plasma (i.e., LCAT derived) source, as evidenced by the high ratio of saturated to unsaturated fatty acid esters. [227] These spherical particles resemble intracellular HDL particles.

Further evidence that newly synthesized HDL particles are secreted by the intestine comes from examination of the specific activity of lipoprotein phospholipid after administration of choline-labeled bile lecithin. [229] The phospholipid of lymph HDL has a lower specific activity than VLDL phospholipid, indicating that the two particles are derived from distinct synthetic pathways. [229]

Mesenteric Lymph Lipoproteins

Studies of rats have characterized the principal species of intracellular apolipoproteins. After secretion into the lymph compartment, several alterations occur in apolipoprotein composition, largely as a result of the passive exchange of surface protein components with lipoproteins in lymph. [220] It is likely, although unproved, that a similar situation exists in humans. Rat mesenteric lymph chylomicrons and human chyluric chylomicrons demonstrate a striking degree of homology in their apolipoprotein compositions. [220] In addition to apo B-48, apo A-IV, and A-I, human and rat lymph-derived chylomicrons contain substantial quantities of albumin, apo E, and apo C. Human chyluric and pig intestinal lymph chylomicrons also contain a peptide of a size compatible with apo B-100, suggesting that apo B-100 transfers onto lymph chylomicrons or, more likely, that hepatic VLDL (containing apo B-100), secreted into the cisterna chyli, contaminates the chylomicron fraction. [228], [230]

The presence of discoidal HDL in mesenteric lymph and in perfusates from rat liver, but not in plasma, suggests that this form of lipoprotein is secreted directly into lymph. It is postulated that discoidal HDL becomes rapidly converted to a spherical morphology after the action of LCAT, which converts free cholesterol to cholesteryl ester and enriches the core of these particles with hydrophobic lipid. [227] During fasting, 75% to 85% of lymph apo A-I is in the HDL fraction. [147], [231], [232] Essentially all apo B in fasting lymph is in the chylomicron-LDL fraction. [147] During lipid feeding, 25% to 50% of apo A-I and 67% to 100% of apo B in lymph is in chylomicrons and VLDL. [147], [233]

Lipid Effects on Intestinal Lipoprotein Assembly and Secretion

Triglyceride Effects on Apolipoprotein Synthesis and Secretion There is no consensus regarding the regulatory effect of triglyceride feeding on human intestinal apolipoprotein synthesis. Some studies suggest that acute triglyceride feeding increases apo A-IV synthesis, but not apo A-I or apo B-48 synthesis. [167], [171], [195], [234] Similarly, chronic triglyceride feeding has no affect on the synthesis of apo A-I or apo B-48. [168] Older studies based on the immunofluorescence or immunoperoxidase

staining of apo A-I, apo A-IV, or apo B demonstrated an increase in intracellular apolipoprotein content in response to triglyceride feeding, which can be interpreted as being consistent with increased synthesis. [232], [235], [236] It may be that alterations in the intracellular location of apolipoproteins, as occurs during lipid absorption, account for differences in staining characteristics. Reports of enterocyte apolipoprotein content after lipid absorption have shown increased, decreased, and unchanged apo A-I and apo B content. [167], [236], [237] and [238] Considered together, the available evidence suggests that only apo A-IV gene expression is responsive to alterations in mucosal triglyceride flux. In this regard, cell culture studies have suggested that apo A-IV synthesis is regulated transcriptionally through the secretion of leptin. [239] These findings, however, have not been confirmed in rats, the results showing no adaptation to chronic high-fat intake and no relation to serum leptin levels. [240] The secretion of apolipoproteins A-I, A-IV, and B into the lymph, unlike their intracellular content or synthesis, increases during lipid feeding. [232], [233], [234], [235], [236], [237] and [238], [241], [242] During lipid absorption, however, lymph flow rates and the transfer of proteins from plasma to lymph increases. Thus, conclusions about enterocyte synthesis based on lymph apolipoprotein measurements are difficult to interpret. [231], [233], [242], [243] The effect of lipid on intestinal apolipoprotein secretion has been studied extensively. In patients with chyluria, in whom intestinal lymph is directly secreted into urine, apo A-I and apo A-IV increase after lipid feeding. [244] Examination of human thoracic lymph after lipid feeding has shown an increase in secretion of pro–apo A-I. [245] However, when plasma apolipoprotein levels are experimentally reduced in rats treated with ethinyl estradiol, only apo A-IV increases in lymph during triglyceride absorption, suggesting that the observed postprandial increases in apo A-I and apo B-48 may be the result of plasma filtration. [246] The issue of increased intestinal secretion or plasma filtration of apo B-48 and apo A-I remains unsettled. A unifying hypothesis is needed to account for the apparently conflicting observations of increased lymph apolipoprotein secretion without accompanying changes in intestinal apolipoprotein synthesis rates after a fatty meal. [146] Such a hypothesis may be derived from the observation that a large intracellular pool of apo A-I and apo B is principally microsomal and not associated with lipoprotein. After a fatty meal, a significant shift occurs, increasing by twofold to threefold the lipoprotein-associated fractions of both apolipoproteins, which are then destined for export, while producing minimal changes (<10%) in the total intracellular pool size of apolipoproteins. [146]

Triglyceride Effects on Lipoprotein Composition The fatty acid composition of chylomicron triglyceride mirrors the composition of triglyceride in the intestinal lumen. [222], [247], [248] The size of secreted chylomicrons is affected by the quantity and type of lipid absorbed. The more triglyceride that is absorbed, the larger the chylomicrons. [247] The composition of the triglyceride also affects the size of chylomicrons. In general, chylomicrons increase in size as the lipids contained within them become less saturated. [247] When saturated fats such as palmitate are fed, smaller chylomicrons are secreted into lymph than when unsaturated fatty acids are fed, although this observation has not been consistent. [222], [233] The presence of unsaturated fatty acids also affects the composition of chylomicrons in that there is less apo B-48 on chylomicrons containing unsaturated triglyceride. [233] During active lipid absorption, the uptake of free fatty acids from plasma into the enterocytes and the incorporation of these fatty acids into newly synthesized triglyceride are increased. [85] Triglyceride feeding affects the triglyceride component of lipoproteins and the phospholipids. The fatty acid composition of lymph lipoprotein phospholipids at least partially reflects the fatty acids present in the intestinal lumen. This observation is consistent with the concept that phospholipids are in part derived from lumenal triglyceride during triglyceride feeding. [229] Phospholipids are synthesized de novo during triglyceride feeding, suggesting that reacylation of absorbed lysolecithin and the phosphatidic acid–cytidine diphosphate choline pathways contribute to lipoprotein phospholipid. Triglyceride feeding also increases the phospholipid content of lymph HDL. [229], [249]

Effect of Bile Diversion and Fasting Reduced intestinal lipid flux has been produced experimentally by using a combination of bile diversion and fasting. The lack of biliary lipid, bile salts, and exogenous lumenal lipid reduces triglyceride output in lymph by 85% and results in the almost total disappearance of VLDL from Golgi organelles. [231] Apo A-I synthesis in jejunal enterocytes after bile diversion is similar to that observed in fasted animals, although ileal enterocytes exhibit lower rates of synthesis. [167] Reinfusion of bile salts alone fails to prevent the decrease in ileal apo A-I synthesis. Apo B-48 synthesis, which is dramatically suppressed in jejunal and ileal enterocytes after bile diversion, is reexpressed in jejunal, but not ileal, enterocytes after intralumenal administration of sodium taurocholate. [195] The bile salt–dependent reexpression of apo B biosynthesis in rat enterocytes can be reproduced by infusion of lysolecithin or fatty acid alone, and it is apparent that this regulation is at a translational or posttranslational level because total apo B mRNA abundance is unaltered. [196] Studies examining intestinal microsomal triglyceride concentration indicate an association between the reexpression of jejunal apo B biosynthesis and microsomal triglyceride content, suggesting that a threshold level of triglyceride may be required for the stable elaboration of intestinal apo B-48. [196] Consistent with the dramatically reduced secretion of triglyceride after bile diversion, intestinal synthesis of apo A-IV is also reduced. [250] The phospholipid composition of lymph HDL during bile diversion is similar to that of non–bile-diverted controls, suggesting that intestinal HDL phospholipid is not derived from exogenous sources. [229] In the absence of dietary fat, the intestine contributes 11% to 40% of the total plasma triglyceride, 10% to 15% of apo B, 50% of plasma apo A-I, and almost 100% of apo A-IV. [157], [222], [249], [251], [252], [253] and [254] Under fasting conditions, intracellular apolipoprotein and VLDL particles can be localized to enterocytes, and newly synthesized apolipoprotein can be isolated from enterocytes and lymph. [145], [168], [255] Intestinal VLDL and HDL are also found in lymph during fasting, with the lipid presumably arising from bile and sloughed, digested enterocytes.

Effect of Phospholipid Absorbed phospholipid comes from bile (10–20 g/d) and from exogenous dietary phospholipid (5–10 g/d). Endogenous sources of lipoprotein phospholipid include de novo synthesized lecithin and the preformed intracellular pool of phospholipids. Lecithin is the predominant phospholipid in lipoproteins. Lecithin infusion results in the intracellular synthesis of triglyceride and lecithin and the incorporation of these lipids into secreted lipoproteins. Infusion of lecithin without triglyceride leads to newly synthesized triglyceride and subsequent VLDL secretion. [229], [256] Most of the infused phospholipid is reassembled into triglyceride through the intracellular hydrolysis of lysolecithin to glycerol-3-phosphate and subsequent reassembly to triglyceride through the phosphatidic acid pathway. [125], [257] Infusion of triglyceride and phospholipid similarly leads to newly synthesized chylomicron triglyceride, which is derived in part from the infused lecithin. Approximately 75% of the fatty acids in infused lecithin occur in lymph triglyceride. [256] The addition of lecithin during high rates of triglyceride infusion increases lymph triglyceride secretion, suggesting that adequate amounts of phospholipid must be available to serve as the surface coat for newly synthesized triglyceride to be secreted as a chylomicron. [258] If biliary lecithin is eliminated during high-dose triglyceride infusion, the effect on lipoprotein phospholipid is variable. Some researchers observed a decrease in secretion, but others reported no change. [229], [258], [259] and [260] At normal rates of triglyceride infusion, there is no decrease in lymph phospholipid secretion after bile diversion. [229], [258] Infusion of lecithin decreases cholesterol secretion. [229], [258] In fasting animals, the phospholipid compositions of VLDL and HDL in lymph resemble those in bile. [229] Elimination of bile that contains 16:0 fatty acids significantly decreases the 16:0 species from lymph lipoproteins in fasted animals. [229] In triglyceride-fed animals, the VLDL and HDL phospholipid compositions mirror the fatty acids in the infused triglyceride. [229] Bile diversion does not significantly affect the composition of lipoprotein phospholipid in fed animals.

Effect of Cholesterol Feeding Only a small percentage of cholesterol recovered in lymph is of exogenous origin. [248] In general, the consequence of increased dietary cholesterol is increased cholesterol content in all lymph lipoproteins. In monkeys, during the experimental infusion of diets with a high cholesterol content, there is an increase in thoracic duct total lymph cholesterol mass. [261] The amounts of free and esterified cholesterol increase, although there is preferential esterification of the absorbed (i.e., lumenal) cholesterol in the lymph chylomicron and VLDL fractions, which leads to a proportionally greater rise in esterified cholesterol than in free cholesterol in the lymph. [262] In rats chronically fed diets containing high levels of cholesterol, mesenteric lymph cholesterol redistributes to an intermediate-density lipoprotein fraction (1.006–1.030 g/mL), in which there appears a particle that is triglyceride- and cholesteryl ester–enriched, with apo A-I and apo B as the major apolipoproteins. [263] Alterations in the composition of ingested triglyceride may affect cholesterol absorption in the rat. Animals fed large concentrations of unsaturated fatty acids demonstrated increased cholesterol absorption, with the excess recovered in the lymph VLDL fraction. [222] These results have not been substantiated in controlled studies of humans. [264]

Metabolic Fate of Intestinal Lipoproteins

The small intestine synthesizes and secretes a variety of lipoprotein particles, principally triglyceride-rich lipoproteins—chylomicrons and VLDL—and HDL. The metabolic fate of each of these particles is distinct and has been the focus of considerable investigation. Chylomicrons enter the lymphatic circulation through fenestrations in the capillary endothelium and immediately undergo a series of modifications to their surface protein composition. The principal surface proteins of nascent intracellular intestinal chylomicrons are pro-apo A-I, apo A-IV, and apo B-48. After entering the lymph compartment, apo E, apo C-II, and apo C-III transfer from the surface of filtered plasma HDL by passive exchange, a process during which chylomicron apo A-I, apo A-IV, and phospholipid are transferred into HDL. [248] Most (50%–75%) of apo A-IV is transferred into the lipoprotein-free fraction of lymph and plasma, [241] where it displays fractional turnover kinetics distinct from those of HDL-associated apo A-IV. [265] As previously described, apo A-I is synthesized and secreted in the form of a stable propeptide containing a 6–amino acid prosegment. [161] This prosegment, which terminates with paired glutamine rather than paired basic residues, [161] is cleaved extracellularly in both lymph and plasma compartments by a metal-dependent protease. [266] The metabolic fate of chylomicron surface apolipoproteins A-I and A-IV appears to be distinctive. These apolipoproteins are thought to contribute substantially to the circulating pool of HDL apolipoprotein. In view of the importance of HDL as an epidemiologic marker of coronary artery disease susceptibility, [166] increased understanding of the physiological parameters governing chylomicron surface catabolism is of clinical importance. After completion of the surface exchange reactions (note that apo B-48 does not participate in these reactions and therefore can be used as a marker of an intestinally derived lipoprotein), chylomicrons are transported to the capillary beds of peripheral tissues, where the core triglyceride undergoes hydrolysis mediated by lipoprotein lipase. [267] Apo C-II is a critical cofactor in activating lipoprotein lipase, and its deficiency is associated with severe hypertriglyceridemia and chylomicron accumulation. [268] Apo C-II is synthesized in both liver and intestine in humans and circulates predominantly with triglyceride-rich lipoproteins. [269] After catabolism of the core triglyceride, the resulting chylomicron particle undergoes substantial modification; most of the surface apolipoproteins are lost, with the notable exception of apo E and apo B-48. This particle is referred to as a *chylomicron remnant* and is transported to a number of sites, including the liver, where it is thought to bind to a cell

surface receptor. The evidence suggests that apo E may be the cognate ligand. [270] This receptor displays characteristics distinct from the LDL (i.e., apo B, apo E) receptor. [270] Metabolic studies of patients with homozygous familial hypercholesterolemia and study of an animal model of familial hypercholesterolemia, in which there is also virtually complete deficiency of LDL receptors, demonstrate normal chylomicron uptake, indicating that at least two populations of lipoprotein receptors recognize apo E–containing particles. [271], [272] A cell surface receptor that resembles the LDL receptor and epidermal growth factor precursor has been identified and is referred to as the *LDL receptor–related protein (LRP)*. [273] LRP functions as an apo E receptor, has an estimated molecular mass of 503 kd, and is translated from a 15-kb transcript. [273] The tissue distribution of the LRP transcript (liver > brain > lung > intestine) suggests that the receptor—if it functions as a chylomicron remnant receptor—may play a more widespread role in tissue cholesterol delivery than previously supposed. Indeed, targeted disruption of LRP in mice leads to embryonic lethality, necessitating the use of the Cre-lox P recombinant system to inactivate LRP to ascertain its role in chylomicron remnant uptake. [274]

Hepatic uptake of triglyceride-rich lipoprotein remnant particles is also modulated by the presence of C apolipoproteins on the lipoprotein surface. Substantial inhibition of remnant uptake occurs with particles containing apo C-II, particularly isoform 2. [275] This finding, based on in vitro studies of the perfused rat liver, has been extended to a study of chylomicron catabolism in persons with apo C-III and apo A-I deficiency, in whom chylomicron catabolism was unusually rapid. [276]

The regulation of plasma HDL metabolism differs from that for chylomicron and LDL metabolism. Plasma HDL is directly secreted from the liver and intestine and is derived from the catabolism of triglyceride-rich lipoproteins. [226], [277], [278] The identification of a class B scavenger receptor (SR-B1) as the long-sought HDL receptor has generated considerable interest in HDL metabolism. [279] Another transporter family, related to the ABC-type transporters, has been identified in patients with low HDL levels as the defective gene in Tangier disease (see previous discussion of ABCA1 in section “ Brush Border Membrane Events”). [65], [66] and [67]

LIPID ABSORPTION IN MALABSORPTIVE STATES

The process of intestinal lipid absorption requires a critical interaction of several distinct elements, disturbances in any of which may become rate limiting to the overall process. A key feature of intestinal lipid absorption is the substantial functional reserve that compensates for minor limitations in some of the critical phases of this process and that limits the extent of malabsorption. For example, the enormous functional capacity of the exocrine pancreas for secreting lipase delays the appearance of lipid malabsorption in pancreatic insufficiency until relatively late in the destructive process. In infants with cystic fibrosis, the preservation of gastric lipase secretion limits the effects of pancreatic insufficiency.

The formation of lipid micelles is critical to effective absorption, and decreased intralumenal bile salt concentrations, resulting from uncompensated ileal losses or bacterial deconjugation in the proximal small intestine, produce malabsorption of lipid. Clinically, the extent of fat malabsorption may be complicated by the coexistence of several defects, such as mucosal damage and villus blunting, together with bile salt deconjugation in small intestine bacterial overgrowth. Processing and intestinal lipoprotein secretion by the enterocyte may be defective. In the setting of diffuse structural diseases, such as celiac sprue, defective enterocyte function can produce severe fat malabsorption.

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CHAPTER 19

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PROTEIN DIGESTION AND ASSIMILATION

The digestion and assimilation of proteins differ from that of carbohydrates and lipids in a number of aspects. Unlike the carbohydrates in the diet, which consist of quantitatively significant amounts of monosaccharides (e.g., glucose and fructose) and disaccharides (e.g., sucrose and lactose), in addition to polysaccharides (e.g., starch and glycogen), dietary amino acids are ingested predominantly as proteins. The amount of free amino acids and small peptides is very low in the normal diet. Dietary carbohydrates are absorbed by the enterocytes exclusively as monosaccharides. Therefore, the polysaccharides and disaccharides in the diet must be digested in the intestinal lumen completely to monosaccharides before absorption can occur. In contrast, the digestion of proteins in the intestinal lumen is incomplete, resulting in the generation of a mixture of free amino acids and small peptides in which peptides predominate. A major portion of protein digestion products is absorbed by the enterocytes as small peptides, which are subsequently digested to free amino acids inside the enterocytes. Thus, in addition to the specific transport systems in the intestine for monosaccharides, distinct transport systems exist in this organ for free amino acids and small peptides. Furthermore, unlike dietary carbohydrates, which consist predominantly of only three structurally different monosaccharides (glucose, galactose, and fructose), dietary proteins comprise a much greater number of structurally and physicochemically diverse amino acids. Consequently, the enzymes and transport systems involved in the digestion and absorption of proteins outnumber those involved in the digestion and absorption of carbohydrates. Moreover, unlike dietary lipids and their digestion products, which are highly hydrophobic, dietary proteins and their digestion products are largely hydrophilic. Therefore, bile salts, which play an obligatory role in the solubilization of dietary lipids to facilitate their digestion and assimilation, do not play any direct role in the digestion and assimilation of dietary proteins. However, bile salts and bile salt–facilitated fat digestion may influence protein digestion indirectly because bile salts and fat digestion products in the intestinal lumen are important modulators of cholecystokinin secretion, a process with profound influence on the secretion of pancreatic enzymes involved in protein digestion.

ASPECTS OF DIETARY PROTEINS

The recommended daily allowance for dietary proteins in humans, when expressed per kilogram of body weight, declines in an age-dependent manner. [1] In adults, this value is 0.75 g/kg; it is twice as much in newborns. The recommended daily allowance for dietary proteins increases during illnesses such as sepsis and during trauma; it also increases in women during pregnancy and lactation. The protein content of the average daily American diet is 70 to 100 g, which is more than adequate to meet the recommended daily allowance. Dietary proteins are required for two purposes: (1) to provide the amino acids that are nutritionally essential under all conditions (i.e., methionine, threonine, tryptophan, valine, isoleucine, leucine, phenylalanine, histidine, and lysine) and the amino acids that are conditionally essential under specific physiological and pathological conditions (i.e., cysteine, tyrosine, glutamine, and arginine); and (2) to provide nitrogen for the synthesis of nutritionally nonessential amino acids and other metabolically important nitrogen-containing compounds. The nutritive value of various dietary protein sources depends on their content of the nutritionally essential amino acids as well as on their digestibility. Proteins from animal sources have a high content of nutritionally essential amino acids. Egg and milk proteins are routinely used as reference standards for comparison of protein quality. Proteins from certain specific plant sources are “incomplete” because they lack or contain only limited amounts of some of the essential amino acids. However, by complementation with plant proteins, which contain the missing amino acids, it is possible to overcome this deficit in a complete diet. [2] Just as the essential amino acid content varies, the digestibility of different proteins also varies considerably. Protein digestibility is influenced by the type of protein and by the method of food processing before ingestion. Generally, plant proteins are less digestible than animal proteins. The relative contributions of animal proteins and plant proteins in the daily intake of dietary proteins differ markedly between developed and developing countries. [3] For example, in North America and Western Europe, animal proteins contribute about 60% to 70% to the total proteins in the diet. In contrast, this contribution is as low as 20% in Africa, the Middle East, and the Far East.

In addition to the proteins in the diet, substantial amounts of endogenous proteins enter the gastrointestinal tract and are digested and assimilated just like the dietary proteins. These endogenous proteins arise partly from the saliva and the gastric, biliary, pancreatic, and intestinal secretions and partly from desquamated epithelial cells of the gastrointestinal tract. The secretions account for about 20 to 30 g of endogenous proteins per day, and desquamated cells account for an additional 30 g per day. [4]

DIGESTION OF PROTEINS

The digestion of proteins occurs in two phases defined by the site of digestion along the gastrointestinal tract: a gastric phase and an intestinal phase. For the sake of convenience, the intestinal phase can be subdivided into a lumenal phase, a brush border phase, and an intracellular phase, again defined by the exact location of digestion in the intestine. The overall scheme of protein digestion in the gastrointestinal tract is depicted in Figure 19-1.

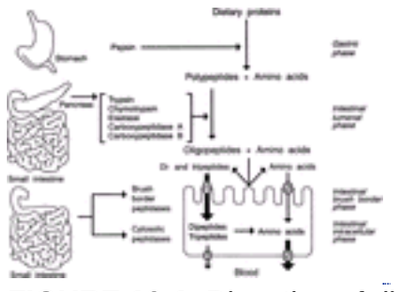
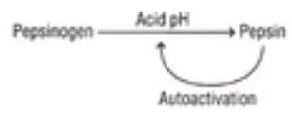


FIGURE 19-1. Digestion of dietary proteins and absorption of digestion products in the gastrointestinal tract. *1,* Transport system for dipeptides and tripeptides; *2,* transport systems for free amino acids.

Gastric Phase

Protein digestion begins in the lumen of the stomach with the action of proteases elaborated by the gastric cells. These enzymes, called *pepsins,* belong to the category of carboxyl proteases that are active at acid pH. The carboxyl groups of two anionic amino acid residues in the enzymes play an obligatory role in the catalytic activity. The enzymes are secreted as inactive precursors into the gastric lumen (i.e., pepsinogens), where they are autocatalytically activated to pepsins

under acidic conditions (Fig. 19-2). Two different pepsinogens are known to be present in humans. Pepsinogen A or I, the more abundant of the two forms, is secreted by chief cells found in the fundus and body of the stomach. The less abundant form, pepsinogen C or II (also known as *progastricsin*), is secreted by chief cells found throughout the entire stomach. There are five isoenzyme forms of pepsinogen A and two isoenzyme forms of pepsinogen C. Pepsinogens possess an amino-terminal region that is positively charged because of the high content of cationic amino acids. At neutral or alkaline pH, this region is folded in pepsinogens in such a way that it masks the catalytic site. This inactive state is stabilized by electrostatic interaction between the cationic amino-terminal region and the acidic amino acids in pepsinogens, which are ionized at neutral or alkaline pH. In the acidic environment of the gastric lumen, these acidic amino acids are protonated, so that electrostatic interaction is destabilized, uncovering the catalytic site. The unmasked active site removes the amino-terminal region, consisting of about 40 amino acids, from pepsinogens by autocatalytic digestion to generate the active forms of the enzymes, the pepsins. Pepsins A and C exhibit similar substrate specificity. They are endoproteases that hydrolyze internal peptide bonds in proteins and liberate large peptide fragments. Pepsins act preferentially on peptide bonds formed by the amino group of the aromatic amino acids phenylalanine and tyrosine, as well as by the branched-chain amino acid leucine (Fig. 19-3). The peptide bonds formed by the carboxyl group of these amino acids are also somewhat susceptible to digestion by pepsins. The action of these enzymes on proteins in the gastric lumen results in a mixture of large polypeptides, smaller oligopeptides, and small amounts of free amino acids.



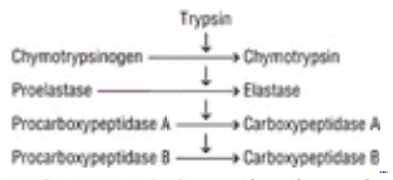


FIGURE 19-2. Activation of gastric and pancreatic zymogens.

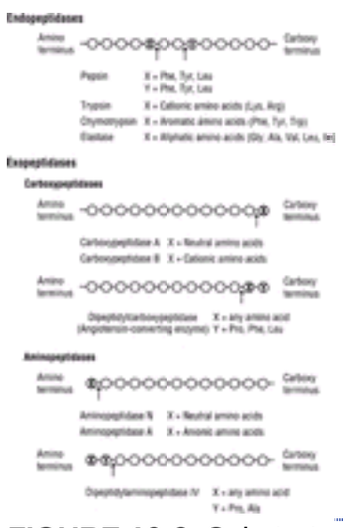


FIGURE 19-3. Substrate specificity of proteases and peptidases involved in the digestion of proteins in the gastrointestinal tract.

Gastric juice contains HCl and thus has a pH of less than 2. The acid pH aids the digestive action of pepsins in three ways: (1) Proteins are denatured, rendering them more susceptible to proteolytic attack; (2) pepsinogens are autocatalyzed to active pepsins; and (3) the catalytic mechanism mediated by the carboxyl groups of the two anionic amino acid residues in the active site of pepsins is effective. Pepsins are not catalytically active at a pH above 4. Therefore, the digestive action of pepsins on proteins is restricted to the stomach. The acidic stomach contents are neutralized by the pancreatic and intestinal secretions when they enter the duodenum, leading to the inactivation of pepsins.

The products of protein digestion by pepsins in the stomach lumen may play an essential role in modulating gastric functions, such as acid and pepsinogen secretion and gastric emptying. These products may also act as stimulants for the release of cholecystokinin in the duodenum, thus influencing pancreatic secretion. Despite the apparent functional consequences of the action of pepsins on proteins, the gastric phase is not essential for overall protein digestion inasmuch as protein digestion and assimilation are not significantly impaired by partial or total gastrectomy.

Intestinal Phase

The intestinal phase of protein digestion consists of three stages: the lumenal phase, the brush border phase, and the intracellular phase (see Fig. 19-1). The lumenal and the brush border phases take place before the protein digestion products are absorbed into the enterocytes across the brush border membrane. In the intracellular phase, the absorbed peptides are digested inside the enterocytes. Protein digestion comes to an end at this stage. The final products of digestion leave the enterocytes across the basolateral membrane and enter the portal circulation.

Lumenal Phase The lumenal phase of protein digestion in the small intestine is mediated by pancreatic proteases and peptidases. This phase begins in the duodenum, where the pancreatic juice enters the small intestine. The exocrine pancreas produces three endoproteases and two exopeptidases, and each of these enzymes is synthesized and secreted as an inactive precursor (i.e., a zymogen). The inactive forms of the three endoproteases are trypsinogen, chymotrypsinogen, and proelastase. The inactive forms of the two exopeptidases are procarboxypeptidase A and procarboxypeptidase B. Activation of these inactive zymogens is initiated in the duodenum by enteropeptidase (also known as *enterokinase*), an enzyme associated with the brush border membrane in the duodenum. Enteropeptidase is an integral membrane protein and consists of a heterodimer linked with disulfide bonds. [5] The heavy chain is anchored to the brush border membrane, and the light chain contains the catalytic site. The enzyme initially is synthesized by the enterocytes as a single polypeptide precursor and then is processed to the heterodimeric form by a hitherto unidentified mechanism. The only known substrate for enteropeptidase is trypsinogen. The action of enteropeptidase on trypsinogen results in the proteolytic removal of a hexapeptide from the amino terminus of trypsinogen, generating trypsin, the catalytically active form. This is the key step in the activation of pancreatic proteases because trypsin thus generated can activate trypsinogen autocatalytically and can also activate the remaining pancreatic zymogens (see Fig. 19-2). The action of trypsin leads to the conversion of chymotrypsinogen to chymotrypsin, proelastase to elastase, procarboxypeptidase A to carboxypeptidase A, and procarboxypeptidase B to carboxypeptidase B. Trypsin, chymotrypsin, and elastase are endoproteases capa- ble of hydrolyzing the internal peptide bonds in proteins and polypeptides. The substrate specificities of these enzymes are given in Figure 19-3. Trypsin hydrolyzes peptide bonds formed by the carboxyl group of cationic (i.e., basic) amino acids (lysine and arginine); chymotrypsin hydrolyzes peptide bonds formed by the carboxyl group of aromatic amino acids (phenylalanine, tyrosine, and tryptophan); and elastase hydrolyzes peptide bonds formed by the carboxyl group of aliphatic amino acids (glycine, alanine, valine, leucine, and isoleucine). Carboxypeptidase A and carboxypeptidase B are exopeptidases, capable of hydrolyzing the terminal peptide bonds on the carboxyl terminus of polypeptides. These two enzymes contain zinc at their active sites. Carboxypeptidase A cleaves carboxyl terminal peptide bonds if the amino acid at the terminus is a neutral amino acid (aromatic or aliphatic), and carboxypeptidase B cleaves carboxyl terminal peptide bonds if the amino acid at the terminus is a cationic amino acid (lysine or arginine) (see Fig. 19-3). These two enzymes thus release free amino acids from polypeptides. Pancreatic endoproteases

and exopeptidases function in a highly complementary manner to carry out the efficient digestion of proteins. Chymotrypsin and elastase generate oligopeptides that contain neutral amino acids at their carboxyl terminus, and these peptides become the preferential substrates for carboxypeptidase A. Trypsin generates oligopeptides that contain cationic amino acids at their carboxyl terminus, and these peptides become the preferential substrates for carboxypeptidase B. The action of these pancreatic enzymes on proteins in the intestinal lumen produces a mixture of oligopeptides and free amino acids, of which oligopeptides constitute a major portion (60%–70%) of the total amino nitrogen.

Brush Border Phase The amino acids generated by the concerted action of pancreatic enzymes on proteins are absorbed as such into the enterocytes across the brush border membrane. In contrast, the oligopeptides undergo further hydrolysis by the action of a battery of peptidases associated with the brush border membrane, resulting in a mixture of free amino acids, dipeptides, and tripeptides, before absorption. [6] The intestinal brush border membrane is particularly rich in aminopeptidase activity. Aminopeptidases are exopeptidases that are capable of hydrolyzing the terminal peptide bonds on the amino terminus of oligopeptides. This provides functional complementation to the carboxypeptidases present in the pancreatic juice. In addition to several aminopeptidases, the intestinal brush border membrane also contains endopeptidase and dipeptidase activities. The two major aminopeptidases associated with the intestinal brush border membranes are aminopeptidase N and aminopeptidase A. The substrate specificity of these two enzymes is given in Figure 19-3. Aminopeptidase N hydrolyzes the amino-terminal peptide bond of oligopeptides if the amino acid at the terminus is a neutral amino acid. Aminopeptidase A, in contrast, hydrolyzes the amino terminal peptide bond of oligopeptides if the amino acid at the terminus is an anionic amino acid. In addition to these enzymes, two other peptidases are associated with the intestinal brush border membrane. Dipeptidylcarboxypeptidase is an exopeptidase (also called *angiotensin-converting enzyme*) that hydrolyzes the peptide bond adjacent to the carboxyl-terminal peptide bond. Dipeptidylaminopeptidase IV is also an exopeptidase, but it hydrolyzes the peptide bond adjacent to the amino-terminal peptide bond. Thus, unlike aminopeptidase N and aminopeptidase A, which release free amino acids from oligopeptides, dipeptidylcarboxypeptidase and dipeptidylaminopeptidase IV release dipeptides from oligopeptides. The substrate specificities of these two enzymes are such that the dipeptides released are generally of the X-proline type, where *X* represents any amino acid (see Fig. 19-3). Animal studies have established unequivocally the essential role of dipeptidylcarboxypeptidase and dipeptidylaminopeptidase IV in the intestinal assimilation of oligopeptides. [7], [8], [9], [10] and [11]

Intracellular Phase The end products of protein digestion by gastric and pancreatic proteases and by peptidases associated with the intestinal brush border membrane comprise a mixture of free amino acids and small peptides. The peptides consist primarily of two to six amino acids. Of the total amino acid content in the intestinal lumen, the peptide-bound amino acids represent a predominant fraction. The absorption of free amino acids into the enterocyte across the brush border membrane is mediated by a number of distinct amino acid transport systems. Peptides consisting of two or three amino acids are absorbed intact across the brush border membrane by a specific peptide transport system. The peptides, once inside the enterocyte, are hydrolyzed to free amino acids in the cytoplasm by various intracellular peptidases. The peptidases exhibit a preference for dipeptides and tripeptides as their substrates. The similarity between the substrate specificities of the intracellular peptidases and the peptide transport system is of functional importance because the transport system makes the peptide substrates available for the intracellular peptidases by importing them across the brush border membrane from the intestinal lumen. The intracellular peptidases that have been characterized in detail include an aminotripeptidase, which releases the amino acid present at the amino terminus of tripeptides, and several dipeptidases, which show differential substrate specificities toward dipeptides containing specific amino acids. [6] Among the dipeptidases, iminodipeptidase (also called *prolidase*) is of special interest because of its restricted specificity toward dipeptides of the X-proline or X-hydroxyproline type. Peptide bonds formed by the imino acids proline and hydroxyproline in proteins are generally resistant to hydrolysis by proteases and peptidases involved in the digestion of proteins in the intestinal lumen. Consequently, the imino acids exist to a large extent as iminodipeptides in the lumen, which are transported intact into the enterocyte by the peptide transport system. These dipeptides are then efficiently cleaved by the intracellular prolidase. The amino acids, which are either transported as such into the cell in the free form or generated inside the cell by the action of intracellular peptidases, exit the cell across the basolateral membrane by way of specific amino acid transport systems to enter the portal circulation.

ABSORPTION OF PROTEIN DIGESTION PRODUCTS

Peptide Absorption

The intestinal brush border membrane possesses a specific transport system that accepts dipeptides and tripeptides as substrates. Free amino acids are not recognized by this system. The most interesting feature of this transport process is that it uses a transmembrane electrochemical H^+ gradient rather than a transmembrane electrochemical Na^+ gradient as the driving force. [12], [13] The physiological significance of this feature is obvious because there is evidence for the existence of an acid pH microclimate on the lumenal surface of the intestinal brush border membrane that creates a H^+ gradient across the brush border membrane in vivo. This acid pH microclimate is generated and maintained by the combined action of the Na^+/H^+ exchanger in the brush border membrane and Na^+,K^+-ATPase in the basolateral membrane of the enterocyte (Fig. 19-4). Another interesting and functionally important feature of the intestinal peptide transport system is its broad substrate specificity. This system accepts as substrates dipeptides as well as tripeptides consisting of neutral amino acids, anionic amino acids, and cationic amino acids. Apparently, there is only a single peptide transport system in the intestine that is capable of handling the large number of chemically and structurally diverse di- and tripeptides that are expected to be generated from the digestion of dietary proteins.

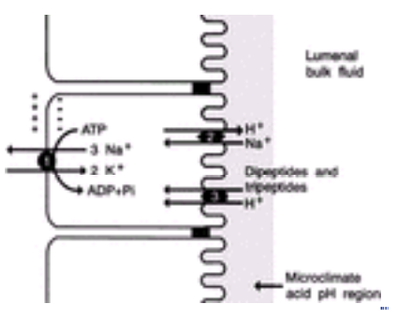


FIGURE 19-4. Generation of an electrochemical proton gradient across the intestinal brush border membrane. *1*, Na^+,K^+-ATPase; *2*, Na^+/H^+ exchanger; *3*, H^+/peptide cotransporter.

The peptide transporters from rabbit, rat, mouse, and human small intestine have been cloned. [14], [15], [16], [17] and [18] The human protein consists of 708 amino acids, with a predicted core molecular size of 79 kd. Hydropathy analysis of the primary amino acid sequence shows the presence of 12 transmembrane domains. The gene for this protein is located on human chromosome 13. This transporter is a member of a superfamily of H^+-coupled peptide transporters expressed in animals, yeast, bacteria, and plants. [19] In situ hybridization [20] and immunolocalization [21] studies in laboratory animals have shown that the peptide transporter is expressed in the small intestine (duodenum, jejunum, and ileum), but not in the esophagus, stomach, colon, or rectum. In the small intestine, the expression is restricted to the absorptive epithelium. Studies of cloned intestinal peptide transporters have established that the transporter recognizes a variety of neutral, anionic, and cationic dipeptides as substrates. [22], [23] and [24] This explains the broad substrate specificity of the intestinal peptide transport system. The mechanism of the transport process is a simultaneous translocation of H^+ and peptide substrate involving a single H^+-binding site on the protein. [25], [26]

Amino Acid Absorption

The absorption of free amino acids from the intestinal lumen into the enterocytes is mediated by amino acid transport systems expressed in the intestinal brush border membrane. [27] Several distinct amino acid transport systems have been functionally characterized in the intestinal brush border membrane, and these transport systems exhibit significant overlap in substrate specificity. The differences between the processes of peptide absorption and amino acid absorption are interesting to note in terms of the number of physiologically occurring amino acids and peptides and the number of transport systems that are involved in their absorption. About 20 different amino acids constitute dietary proteins. Absorption of these amino acids in the free form is mediated not by a single amino acid transport system but by several distinct amino acid transport systems. In contrast, the theoretically possible number of structurally different dipeptides and tripeptides arising from dietary proteins during the digestive process is several times greater than the number of structurally distinct amino acids (400 dipeptides and 8000 tripeptides). Nevertheless, a single peptide transport system mediates the absorption of all these diverse peptides. Another notable difference between the peptide and amino acid absorptive processes is in the energetics. The energy source for the peptide transport process is the electrochemical H^+ gradient; a Na^+ gradient is not directly involved. In contrast, several amino acid transport systems are dependent on the Na^+ gradient as the driving force. Some amino acid transport systems, however, are independent of any ion gradient.

The different amino acid transport systems known to be present in the intestinal brush border membrane, their substrate specificities, and their dependence on the Na^+ gradient are given in Table 19-1. The major transport system responsible for the absorption of neutral amino acids across the intestinal brush border membrane is the Na^+-dependent system B°. The molecular nature of this transport system has been elucidated. [28], [29] The human system B° protein is made up of 541 amino acids and is predicted to contain 10 transmembrane domains. The gene coding for this protein is located on human chromosome 19. Immunolocalization studies have established unequivocally that the system B° protein is restricted to the brush border membrane of the absorptive epithelial cells in the small intestine. [30] Glutamine is a high-affinity substrate for system B°, a fact bearing physiological significance because of the well-known relevance of this amino acid to the maintenance of intestinal structure and function and to the treatment and prevention of critical illness. [31], [32] and [33]

TRANSPORT SYSTEM	SUBSTRATES	NA^+-DEPENDENT
B°	Neutral amino acids	Yes
$B^{0,+}$	Neutral amino acids, cationic amino acids	Yes
X^-_{AG} or EAAT3	Anionic amino acids	Yes
IMINO	Imino acids	Yes
$b^{0,+}$	Neutral amino acids, cationic amino acids, and cystine	No
y^+ or CAT1	Cationic amino acids	No

CAT, cationic amino acid transporter; EAAT, excitatory amino acid transporter.

TABLE 19-1 Amino Acid Transport Systems in the Intestinal Brush Border Membrane

System $B^{0,+}$ is also present in the brush border membrane. This unique amino acid transporter mediates the Na^+- and Cl^--coupled electrogenic transport of neutral as well as cationic amino acids across the brush border membrane. [34] The gene coding for this recently cloned transporter is located on human chromosome X. [35], [36] Interestingly, this transporter is expressed predominantly in the ileum and the colon, not in the proximal part of the small intestine. [36] System $B^{0,+}$ is a transporter not only for amino acids but also for carnitine [37] and several nitric oxide synthase inhibitors. [36]

A separate transport system present in the intestinal brush border membrane is specific for anionic amino acids. X^-_{AG} (i.e., the transport system for the negatively charged amino acids aspartate and glutamate) is also Na^+ dependent. The protein, which has been cloned from rabbit small intestine, [38] contains 524 amino acids and 10 putative transmembrane domains. Its human ortholog also has been characterized. [39] Because this transporter is also expressed in glutamatergic neurons in the brain, where it functions in the neuronal reuptake of glutamate (an excitatory amino acid), system X^-_{AG} is currently classified as a member of the excitatory amino acid transporter (EAAT) family. The glutamate transporter expressed in the intestinal brush border membrane is known as EAAT3.

Another transport system in the intestinal brush border membrane recognizes neutral and cationic amino acids as high-affinity substrates. This system, $b^{0,+}$, is Na^+ independent, and interestingly, it also recognizes the disulfide amino acid cystine as a substrate. System $b^{0,+}$ is a heterodimer consisting of a light chain and a heavy chain. [40], [41] The light chain possesses 12 putative transmembrane domains and exhibits a membrane topology typical of most transport proteins. The heavy chain is responsible for directing the recruitment of the light chain specifically to the enterocyte brush border membrane. It may also have a modulatory role in the transport function. [42] The genes coding for the heavy chain and the light chain are located on human chromosomes 2 and 19, respectively. [43], [44]

System y^+ is a Na^+-independent amino acid transporter with a preference for cationic amino acids. [45] There are at least four subtypes within the cationic amino acid transporter (CAT) gene family: CAT1, CAT2, CAT3, and CAT4. Of these, only CAT1 is expressed in the intestine. This transporter, in contrast to system $b^{0,+}$, does not interact with cystine. System IMINO is a Na^+-dependent transporter with specificity toward the imino acids proline and hydroxyproline. The molecular identity of system IMINO remains unknown.

FATE OF ABSORBED PROTEIN DIGESTION PRODUCTS

At the end of the intracellular phase of protein digestion in the enterocyte, after the absorption of small peptides and free amino acids from the intestinal lumen across the brush border membrane, free amino acids constitute the predominant form of the protein digestion products in the enterocyte cytoplasm. It is probable, however, that a small but significant portion of the protein digestion products may exist in the form of small peptides that are absorbed by way of the brush border membrane peptide transporter but are resistant to hydrolysis by intracellular peptidases. Free amino acids in the cytoplasm of the enterocyte enter into various metabolic pathways, such as degradation, conversion into other amino acids, and incorporation into cellular proteins; they also enter the portal circulation by way of specific amino acid transport systems in the basolateral membrane. Glutamine, glutamate, aspartate, and arginine are preferentially used by the enterocyte as metabolic fuel.

The intestinal basolateral membrane possesses a set of amino acid transport systems that are different from those in the brush border membrane (Table 19-2). [27] The amino acid transport systems in the basolateral membrane function to export amino acids from the enterocyte into the portal circulation during the periods of feeding. They also participate in the import of amino acids from the portal circulation into the enterocyte for cellular metabolism when amino acids are not available from the intestinal lumen, such as between meals. Of the amino acid transport systems present in the basolateral membrane, systems y^+L and A are of particular importance. System y^+L is an amino acid exchanger that mediates the efflux of cationic amino acids from the intestinal cells into the blood coupled to the Na^+-dependent influx of neutral amino acids from the blood into the intestinal cells. [40], [41], [45] This transport system is also a heterodimer, consisting of a light chain and a heavy chain, the former mediating the actual transport function and the latter facilitating the recruitment of the light chain to the basolateral membrane. [41] System A is a Na^+-coupled transporter for neutral amino acids, including glutamine, that plays a role in the entry of amino acids from the blood into the intestinal cells for use in cellular metabolism. System A consists of at least three subtypes, amino acid transporter 1 (ATA1), ATA2, and ATA3. [46], [47] and [48] Of these, ATA1 and ATA2 are expressed in the small intestine. The intestinal basolateral membrane also possesses a peptide transport system that may or may not be identical to that in the brush border membrane. This transport system facilitates the exit of hydrolysis-resistant small peptides from the enterocyte into the portal circulation.

TRANSPORT SYSTEM	SUBSTRATES	NA^+-DEPENDENT
A	Neutral amino acids and imino acids	Yes
ASC	Small neutral amino acids (e.g., Ala, Ser, Cys)	Yes
asc	Small neutral amino acids (e.g., Ala, Ser, Cys)	No
L	Neutral amino acids	No
y^+L	Cationic amino acids	No
	Neutral amino acids	Yes

TABLE 19-2 Amino Acid Transport Systems in the Intestinal Basolateral Membrane

PHYSIOLOGICAL AND CLINICAL SIGNIFICANCE

Compelling evidence exists for the absorption of protein digestion products predominantly in the form of small peptides rather than free amino acids. Therefore, the physiological importance of the intestinal peptide transport system in the maintenance of protein nutrition is clearly evident. In addition, the intestinal peptide transporter has been shown to interact with several pharmacologically relevant drugs that possess peptide-like chemical structures. These drugs are efficiently absorbed from the intestinal lumen after oral administration by way of the peptide transporter. Consequently, the peptide transporter in the intestine is an important determinant of the bioavailability of orally active peptidomimetic drugs. Examples of peptidomimetic drugs that are absorbed in the intestine by way of the peptide transporter include several ß-lactam antibiotics (e.g., cephalexin, cefadroxil, cefradine, and ceftibuten), angiotensin-converting enzyme inhibitors (captopril and

lisinopril), the antitumor agent bestatin, inhibitors of renin and thrombin, and prodrugs such as valacyclovir, valgancyclovir, and a-methyldopa-Phe. [49]

The peptide transport process in the intestine also has potential for therapeutic applications. Peptide-based formula solutions have many advantages over amino acid–based elemental solutions for enteral nutrition. [50] The efficacy of peptide-based enteral solutions is facilitated by the intestinal peptide absorptive process. The osmolality of these solutions can be kept low while amino acid content and composition are maintained. Furthermore, amino acids are absorbed more efficiently and more evenly from peptides than from an equivalent mixture of free amino acids. Another clinical application of the peptide transport system is the practice of including heat-labile and water-insoluble amino acids such as glutamine, cysteine, and tyrosine in the form of peptides in enteral solutions to promote their absorption.

REGULATION OF AMINO ACID AND PEPTIDE ABSORPTION

The capacity of the small intestine to absorb protein digestion products changes significantly under various physiological and pathological conditions. The underlying mechanisms of these changes may be nonspecific, such as alterations in the absorptive surface area; or specific, such as alterations in the number and activity of particular transporters. The transport systems responsible for the absorption of amino acids and peptides are present in the small intestine even before birth in several animal species, including humans. The ability of the small intestine to absorb amino acids and peptides, when expressed per wet weight of intestinal tissue, is maximal at birth and during early development, decreasing thereafter with age. Several hormones have been shown to alter the intestinal amino acid and peptide transport systems. Somatostatin and vasoactive intestinal polypeptide inhibit these transport processes, whereas epidermal growth factor, neurotensin, cholecystokinin, and secretin enhance them. Studies of cultured cells indicate that various intracellular second messengers regulate the intestinal peptide transporter. In Caco-2 cells, a human intestinal cell line, the peptide transporter is stimulated by insulin [51] and inhibited by protein kinase C [52] and cyclic adenosine monophosphate (cAMP). [53] However, because there is evidence of direct coupling between the peptide transporter and the Na^+/H^+ exchanger via the transmembrane H^+ gradient in Caco-2 cells, [12], [54] it is not clear whether the observed effects of insulin, protein kinase C, and cAMP on peptide transporter activity are direct or mediated indirectly by alterations in the activity of the Na^+/H^+ exchanger.

In animal studies, the expression of the intestinal peptide transporter also has been shown to be modulated by dietary protein content and protein digestion products. [55], [56] Even though the peptide transporter is expressed along the entire small intestine, the diet-induced changes in the expression of the transporter are specific to certain regions. A high-protein diet increases the steady-state levels of the transporter-specific messenger RNA in the middle and distal regions of the small intestine. The expression of the brush border peptidases dipeptidylcarboxypeptidase and dipeptidylaminopeptidase IV, which release dipeptides from oligopeptides, are also enhanced by a high-protein diet, again only in the middle and distal regions of the small intestine. This coordinated regulation of the peptide transporter and the peptidases that generate the substrates for the transporter is likely to be of physiological importance. Similarly, the protein digestion products, particularly certain specific dipeptides and amino acids, also enhance the expression of the peptide transporter in the intestine.

PROTEIN-ENERGY MALNUTRITION

Protein-energy malnutrition represents the most common type of malnutrition seen primarily in developing countries. This condition is classified into two subtypes: kwashiorkor and marasmus. In kwashiorkor, intake of protein is inadequate, but energy intake in the form of carbohydrate and fat is adequate. In marasmus, the intake of both protein and energy is inadequate. The crucial clinical features that differentiate these two subtypes are the degree of body wasting and the presence or absence of edema. Kwashiorkor is characterized by edema and a body weight 60% to 80% of that expected for the given age. This condition is also associated with fatty liver. Inadequate protein intake in the presence of adequate energy in kwashiorkor results in decreased hepatic synthesis of blood proteins and ß-lipoprotein. Decreased protein levels in the blood cause edema, and decreased ß-lipoprotein in the liver causes defective handling of lipids, leading to fatty liver. Marasmus is characterized by the absence of edema and less than 60% of the expected body weight for the given age. Both conditions manifest a decreased cell-mediated immune response that reduces the ability to fight infection.

DEFECTS IN DIGESTION AND ABSORPTION OF PROTEINS

A number of defects, primary as well as secondary, associated with the impairment of protein digestion and assimilation are known to occur in humans (Table 19-3).

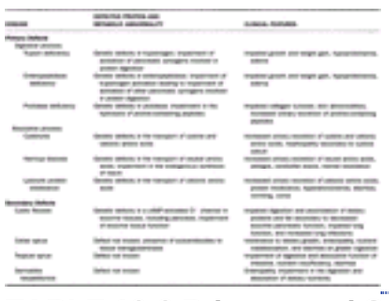


TABLE 19-3 Primary and Secondary Defects Associated with an Impairment of Protein Digestion and Assimilation

Primary Defects

Defects in the digestion of proteins can be caused by genetic diseases involving the expression and activity of any of the proteases or peptidases necessary for the digestive process. Cases of a genetic deficiency of pancreatic trypsinogen have been reported. [57], [58] Because trypsin is a key player in the activation of other pancreatic zymogens (e.g., chymotrypsinogen), a deficiency of trypsinogen results in a marked impairment of protein digestion, noticeable in early infancy. Infants with trypsinogen deficiency fail to gain weight and present with hypoproteinemia and edema. These infants improve dramatically if fed protein hydrolysates instead of intact proteins. Several cases have been reported of a genetic deficiency of enteropeptidase, [59] an intestinal enzyme responsible for the activation of trypsinogen. Infants with enteropeptidase deficiency present with symptoms similar to those observed in cases of trypsinogen deficiency, which include failure of growth, hypoproteinemia, and edema. Because a deficiency of enteropeptidase leads to impairment of trypsinogen activation and consequently to an impairment of the activation of other pancreatic protease zymogens, the digestion of proteins is markedly affected in these patients. Treatment with activated pancreatic enzymes results in improvement of protein digestion and assimilation. Genetic defects in enteropeptidase and trypsin cause impairment not only in protein digestion but also in the digestion of fat. This is because trypsin is obligatory for the activation of procolipase to colipase, a protein necessary for the hydrolytic action of pancreatic lipase on dietary triglycerides after emulsification with bile acids. Similarly, because pancreatic proteases play an essential role in the intestinal absorption of vitamin B_{12}, genetic defects in enteropeptidase and trypsin may also interfere with the absorption of this vitamin.

With the exception of enteropeptidase deficiency, no other documented inheritable defects in peptidases are associated with the intestinal brush border membrane in humans. However, a mutant rat strain is available that exhibits a genetic deficiency of dipeptidylaminopeptidase IV. [11] These mutant rats are apparently normal with no alteration in growth rate if fed a diet with protein constituents with a modest proline content. [11] However, if the dietary protein source is changed to gliadin, a proline-rich protein, the enzyme-deficient mutant rats experience a significant weight loss, whereas wild-type rats do not. These studies show that, as a result of the unique substrate specificity of dipeptidylaminopeptidase IV, a functional role for this enzyme becomes evident under physiological conditions only if the diet contains proline-rich proteins. These observations also point to the physiological advantage of the presence of multiple peptidases in the intestinal brush border membrane with significant overlapping substrate specificities.

Inheritable deficiency of the intracellular dipeptidase prolidase is known in humans. [60] In healthy persons, this enzyme is expressed not only in the intestine but also in several other tissues, such as the kidney; its functions are multiple, including an important role in collagen turnover. It is therefore not surprising that manifestations of prolidase deficiency are pleiotropic. However, no information is available on the intestinal assimilation of proteins in these patients. It is likely that prolidase deficiency does not lead to detectable alterations in the intestinal assimilation of proteins under normal dietary conditions, as seen in the mutant rat strain lacking dipeptidylaminopeptidase IV.

Defects in the absorption of protein digestion products can be caused by diseases involving the expression and activity of amino acid and peptide transport systems in the intestine. Several inheritable amino acid transport defects have been identified in humans. Cystinuria is an autosomal recessive disorder characterized by the increased excretion of cationic amino acids (lysine and arginine) and cystine. [61] The disease is associated with a defect in the renal reabsorption of these amino acids. A similar defect also occurs in the small intestine, leading to impaired absorption of cationic amino acids and cystine from the intestinal lumen. This suggests that an identical transport system is involved in the absorption of these amino acids in the kidney and the intestine. The primary clinical problem associated with the disease is nephropathy resulting from renal stones enriched in cystine. This disulfide derivative is poorly soluble in water (300 mg/L). In cystinuria, the concentration of cystine is greater than this solubility level in the renal tubule because of defective reabsorption, and cystine stone formation results. The amino acid transporter $b^{\circ +}$ is defective in cystinuria. This disease is caused by mutations in either the light chain [44] or the heavy chain [40] of the heterodimeric transporter $b^{\circ +}$. Thus, two genes, one on chromosome 2 (heavy chain) and the other on chromosome 19 (light chain), are associated with cystinuria. Because system $b^{\circ +}$ is present primarily in the brush border membrane of the intestinal and renal tubular epithelial cells, the transport defect in cystinuria is restricted to this membrane. Therapeutic strategies are directed toward facilitating cystine excretion in the urine by increasing urine volume, alkalinizing the urine to increase cystine solubility, and using penicillamine, which reacts with cystine to form a more soluble mixed disulfide.

Hartnup disease is also an autosomal recessive disorder of amino acid transport, but this condition is associated with impaired absorption of neutral amino acids (Ala, Ser, Cys, Thr, Gln, Asn, Val, Ile, Leu, Met, Phe, Tyr, and Trp) in the kidney and the intestine. [62] Consequently, this disease is associated with the hyperexcretion of several neutral amino acids in urine. The symptoms of the disease, which include photosensitive skin rash, ataxia, and neurological complications, are similar to those of pellagra (i.e., niacin deficiency). These symptoms occur despite an apparently normal dietary intake of niacin but disappear in response to niacin therapy. In humans, endogenous synthesis of niacin from the neutral amino acid tryptophan contributes significantly to normal niacin requirements. In Hartnup disease, the endogenous synthesis of niacin is most likely impaired because of defective intestinal absorption of tryptophan coupled with defective renal reclamation of tryptophan. Because people in industrialized nations generally consume a nutritious diet rich in protein, the disease is relatively benign, except for symptoms of niacin deficiency, cerebellar ataxia, and occasional psychological problems. However, in developing or underdeveloped nations, where suboptimal protein intake is common, the disease is characterized by more severe clinical symptoms, including mental retardation. The amino acid transport system B° has been suggested as one of the prime candidates for the amino acid transport defect seen in Hartnup disease because of its specificity toward neutral amino acids and its known expression in the intestine and the kidney. [28], [29] The amino acid transport defect in Hartnup disease is restricted to the brush border membrane of the intestinal and renal tubular epithelial cells. The finding that system B° is associated exclusively with the brush border membrane of these cells [30] supports the hypothesis that Hartnup disease is caused by defects in the gene coding for this transporter.

Interestingly, persons with cystinuria or Hartnup disease usually do not show obvious symptoms of protein malnutrition, despite the clearly established defects in the intestinal absorption of specific amino acids. This was puzzling until it was discovered that intestinal absorption of the affected amino acids (i.e., cationic amino acids in cystinuria and neutral amino acids in Hartnup disease) is normal if the amino acids are given in the form of dipeptides. [63], [64], [65] and [66] Because protein digestion products are absorbed across the intestinal brush border membrane mainly in the form of small peptides, these individuals obtain significant amounts of the affected amino acids through the peptide transport mechanism despite the amino acid transport defect in this membrane. These observations are of historical importance in peptide transport research because they provided early indisputable evidence that the transport mechanisms available for free amino acids and peptides in the intestine are different. [67]

Lysinuric protein intolerance is an autosomal recessive disorder caused by impaired absorption of the cationic amino acids lysine and arginine in the intestine and the kidney. [68] The absorption of ornithine, also a cationic amino acid, is impaired as well. Persons with this disease show decreased plasma levels and increased urinary excretion of these three amino acids. Deficiency of arginine and lysine, which are essential amino acids, causes impairment of protein synthesis, resulting in growth retardation and neurological complications. In addition, because arginine and ornithine are the amino acids involved in the urea cycle and hence in the elimination of ammonia, patients with this disease exhibit decreased urea cycle activity and consequently hyperammonemia. Dietary intake of protein induces diarrhea, vomiting, and, in extreme cases, coma. Treatment strategies, which are related to the prevention of hyperammonemia, include dietary protein restriction and ornithine supplementation. The intestinal and renal handling of cystine is not affected in lysinuric protein intolerance, indicating the difference between the molecular defects associated with this disease compared to cystinuria, which also involves the renal and intestinal absorption of cationic amino acids. The transport defect in lysinuric protein intolerance is restricted to the basolateral membrane in the intestinal and renal epithelial cells. [69] The transport of cationic amino acids across the brush border membrane of these cells is not affected. Linkage analysis [70] assigned the gene for lysinuric protein intolerance to the long arm of chromosome 14. The transport system defective in this disease is $y^{+}L$, as supported by its localization to the basolateral membrane of the intestinal and renal tubular epithelial cells and also by the location of the gene coding for the light chain of this heterodimeric system on chromosome 14. Mutations in this gene have been identified in patients with lysinuric protein intolerance. [71], [72]

Secondary Defects

Impairment of protein digestion and absorption occurs as a secondary defect in several diseases, including cystic fibrosis, celiac sprue, tropical sprue, and dermatitis herpetiformis. In cystic fibrosis, the primary defect occurs in the CFTR (cystic fibrosis transmembrane conductance regulator) protein, a cAMP-regulated Cl^{-} channel. The defective function of this protein leads to complications involving diverse organs, such as the lung, pancreas, gastrointestinal tract, and sweat glands. In the exocrine pancreas, the absence of the Cl^{-} channel in the apical membrane of pancreatic ductal epithelial cells results in failure to secrete salt and water into the duct. This leads to the retention of digestive enzymes in the pancreas and, ultimately, the destruction of pancreatic tissue. Pancreatic insufficiency associated with this disease causes impairment of the digestion and assimilation of protein and fat. Celiac sprue, or gluten-induced enteropathy, is characterized by an inability to tolerate gluten, a protein found in wheat and rye. The dietary intake of gluten induces abnormalities in the intestinal structure, characterized by blunting and flattening of the villi, which reduces the mucosal surface area and results in a deficiency of enterocyte-associated digestive enzymes and transport proteins. This causes a generalized impairment of the digestive and absorptive function of the intestine. Celiac sprue is a genetic disease, and immunoglobulin A antibodies to endomysium, a structure of the smooth muscle connective tissue, are specific indicators of the disease. The endomysial autoantigen has been identified as the tissue transglutaminase. [73] Interestingly, gliadin, which is the major protein component of gluten, is a preferred substrate for this enzyme. Tropical sprue is a serious gastrointestinal disease also characterized by a generalized impairment of the digestive and absorptive function of the intestine. A disease of unknown etiology, it occurs among the residents of tropical areas. The gastrointestinal manifestations of tropical sprue are strikingly similar to those observed in celiac sprue. Dermatitis herpetiformis is another malabsorption syndrome associated with blunting and flattening of the mucosal villi and consequently impaired digestion and absorption of dietary nutrients.

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CHAPTER 20

Charles H. Halsted and Bo L. Lönnerdal

VITAMIN AND MINERAL ABSORPTION

WATER-SOLUBLE VITAMINS

Folate and vitamin B_{12} (cobalamin) have complex and different mechanisms of absorption, and each has a broad range of clinical effects. Because these vitamins are interactive metabolically, many of the clinical signs of deficiency are similar, even though their structures, food sources, and modes of absorption are dissimilar (Table 20-1).

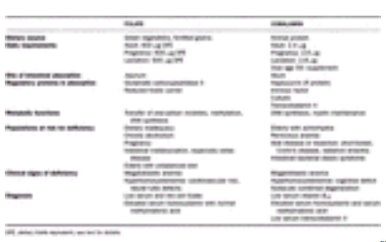

TABLE 20-1 Comparison of Folate and Cobalamin

Folate

Sources Folate in the form of oxidized folylmonoglutamate, or folic acid, contains a pteridine ring linked to *p*-aminobenzoic acid and glutamic acid. Folates appear in the diet and in endogenous storage as folylpolyglutamates with up to six additional glutamates in ?-peptide linkage to folylmonoglutamate. Whereas the term *folic acid* refers to the oxidized monoglutamyl form of the vitamin, reduction in the 5-, 6-, 7- and 8-positions and methylation at the 5-position of the pteridine ring produce 5-methyltetrahydrofolate (5-MTHF), the circulating form of the vitamin. Dietary sources of folate include green leafy vegetables (i.e., foliage), orange juice, grains (in particular, breakfast cereals that are fortified with folic acid), and organ meats such as liver. The Dietary Reference Intake (DRI; formerly referred to as the *Recommended Dietary Allowance [RDA]*) for folate from all sources is now quantified in dietary folate equivalents (DFEs). This term accounts for the fact that monoglutamyl folic acid that is marketed as a vitamin supplement or as a fortifier in the diet is 1.7 times more available than the natural food folates that occur as folylpolyglutamates in the diet. [1] In calculating DFEs, the amount of supplemental folic acid is multiplied by 1.7 and added to the amount of folate present in the mixed diet. The DRI for folate is set at 400 µg of DFE per day for adults, 600 µg/d during pregnancy, and 500 µg/d during lactation. Since 1998, all grain products in the United States have been fortified with folic acid at 1.40 mg/kg to ensure a daily intake equal to or more than the DRI for folate. This mandated fortification of the food supply has lowered the incidence of folate deficiency in the United States from 10% to less than 1% of the population. [2]

Folate Absorption and Homeostasis Folates are absorbed in the duodenum and upper jejunum. During the process of intestinal absorption, dietary folylpolyglutamates are hydrolyzed at the epithelial cell (enterocyte) brush border membrane, followed by active transport of the folylmonoglutamate derivative into the absorbing enterocyte. Studies that followed the movement of labeled compounds across the human jejunum during jejunal perfusion or when excreted in the urine after oral administration demonstrated the importance of the hydrolytic step, because the absorption of monoglutamyl folic acid was about 85%, compared to the 50% absorption of complex dietary folylpolyglutamates in adult human volunteers. [3] The 85:50, or 1.7:1, ratio of the absorption of the two different folates provides the basis for the DFE calculation. Two intestinal proteins—glutamate carboxypeptidase II (GCP II, formerly known as *folate conjugase* or *hydrolase*) and the reduced folate carrier (RFC)—are required, respectively, for the sequential hydrolysis of dietary folylpolyglutamates and the transport of folylmonoglutamate derivatives across the enterocyte brush border and basolateral membranes. GCP II has been characterized in pig and human intestine and expresses a brush border membrane protein with molecular size of 120 kd that contains a single 5' membrane-spanning domain and an extracellular catalytic domain. GCP II shares an identical sequence with two other proteins: brain *N*-acetylated a-linked acidic dipeptidase and prostate-specific membrane antigen. [4] In its functionally active enzyme form, purified human intestinal GCP II has an optimal pH of 6.5 and a K_m value of 0.6 µmol/L and is an exopeptidase that releases intermediate products of folylpolyglutamates in stepwise fashion ending with the folylmonoglutamate derivative. [5] By contrast, human RFC is a 65-kd brush border membrane protein with 12 membrane-spanning domains that operates at an optimal pH of 5.0 and a K_m value of 0.5 to 1.5 µmol/L. [6] Studies that used isolated intestinal brush border membrane vesicles defined a saturable carrier system for monoglutamyl folates with a preference for reduced and methylated forms and capable of either H^+ cotransport or anion exchange. During folate transport through the absorbing enterocyte, pteroylmonoglutamate is reduced and methylated, and 5-MTHF is transported across the intestinal basolateral membrane by the RFC to the portal vein and the liver. The uptake of 5-MTHF by hepatocytes involves both liver plasma membrane–binding and carrier-mediated transport into the cell. The binding step is regulated in part by the folate receptor or folate-binding protein (FBP), a 37-kd protein that operates at a pH of 6.5 to 7.5 and a K_m value in the nanomolar range. FBP is attached to the membrane by a glycosylphosphatidylinositol anchor. [7] 5-MTHF transport obeys the kinetics of the RFC, which also expresses its activity in the liver plasma membrane. [8] Within the hepatocyte, 5-MTHF uptake is followed by conversion to the folylpolyglutamate form for storage and metabolism. About 10% of the liver folate pool undergoes biliary secretion as 5-MTHF to an enterohepatic folate circulation, while much of the remaining passes into the systemic circulation. [9] The urinary excretion of 5-MTHF is regulated by filtration and then efficient reuptake by renal tubular epithelial cells via both FBP and RFC. [10] About 1% of the total body folate pool is excreted daily in the urine and 0.1% in the feces. [11]

Folate Functions Folates serve as substrates or cofactors in the transfer of single-carbon moieties in amino acid metabolism and in nucleic acid synthesis (Fig. 20-1). In the methyltransferase pathway, 5-MTHF, which is derived from both dietary and endogenous sources, is the substrate for the methionine synthase reaction that, with cobalamin cofactor, generates methionine from homocysteine. In the alternate salvage pathway, methionine is generated from betaine and homocysteine by way of the betaine homocysteine methyltransferase (BHMT) pathway. Methionine is converted to *S*-adenosylmethionine (SAM) by methionine adenosyltransferase (MAT), an enzyme produced by two genes; *MAT1A* expressed in the liver and *MAT2A* expressed in nonhepatic tissues, and in the liver in alcoholic hepatitis and carcinoma. [12] About 6 to 8 g of SAM is present in a healthy liver. Through many reactions that include DNA methylation and the synthesis of phosphatidylcholine, SAM is metabolized to *S*-adenosylhomocysteine (SAH), which is also generated from homocysteine through the reversible SAH hydrolase (SAHh) reaction. SAM regulates the synthesis of glutathione by up-regulating cystathionine ß-synthase and the homocysteine transsulfuration pathway. SAM also provides negative regulatory feedback to the methylene tetrahydrofolate reductase (MTHFR) reaction that converts 5,10-MTHF to 5-MTHF. Thus, adequate SAM ensures sufficient 5,10-MTHF as

substrate for thymidylate synthase, which ensures the nucleotide balance of deoxyuridine monophosphate and deoxythymidine monophosphate for DNA synthesis. According to these metabolic interactions, hyperhomocysteinemia, or elevated circulating homocysteine, results from either folate or cobalamin deficiency, but may also be caused also by genetic polymorphisms that reduce the activities of cystathionine ß-synthase methionine synthase, or MTHFR. Levels of SAM are reduced through decreased methionine production by methionine synthase or oxidative inactivation of MAT, [13] whereas SAH, a product of homocysteine, inhibits SAM methylation reactions. [14] The consequences of reduced levels of SAM include decreased DNA methylation and nucleotide imbalance with DNA instability, and decreased glutathione production and oxidative defense. Considerable evidence suggests that SAM deficiency is an integral component in the pathogenesis and clinical expression of alcoholic liver disease. [15]

FIGURE 20-1. Interactions of folate, methionine, and choline metabolism. *BHMT,* betaine homocysteine methyltransferase; *CBS,* cystathionine ß-synthase; *CDP-choline,* cytidine diphosphorylcholine; *choline-P,* phosphoryl choline; *DNMTs,* DNA methyltransferases; *dTMP,* deoxythymidine monophosphate; *dUMT,* deoxyuridine monophosphate; *GSH,* glutathione; *MAT,* methionine adenosyltransferase; *MS,* methionine synthase; *5-MTHF,* 5-methyltetrahydrofolate; *5,10-MTHF,* 5,10-methylenetetrahydrofolate; *MTHFR,* methylenetetrahydrofolate reductase; *PC,* phosphatidylcholine; *PE,* phosphatidylethanolamine; *PEMT,* phosphatidylethanolamine methyltransferase; *SAH, S*-adenosylhomocysteine; *SAM, S*-adenosylmethionine; *SAHh,* SAH hydrolase; *THF,* tetrahydrofolate; *TS,* thymidylate synthase.

Clinical Expressions of Folate Deficiency Because folate ultimately is required for maintaining nucleotide balance during DNA synthesis, its deficiency is expressed by increased apoptosis and by compensatory increased proliferation of cells with rapid turnover in the blood-forming bone marrow and in the intestinal mucosa. Bone marrow megaloblastosis and macrocytosis of red cells and enterocytes are a reflection of deficient DNA synthesis with production of immature and dysfunctional cells that translates clinically to macrocytic anemia and, potentially, diarrhea. As indicated in Figure 20-1, folate deficiency also affects the methionine metabolic cycle, resulting in hyperhomocysteinemia. In addition to megaloblastic anemia, several common clinical conditions have been linked to folate deficiency with abnormal DNA metabolism and hyperhomocysteinemia.

Anemia and diarrhea. The clinical expression of overt folate deficiency is megaloblastic anemia, with findings of macrocytic red blood cells with increased mean corpuscular volume and hypersegmented neutrophils in the peripheral blood. Macrocytic enterocytes were identified in small intestinal biopsy specimens from alcoholic patients with folate-deficient megaloblastic anemia. [16] Furthermore, experimental folate deficiency combined with ethanol intake in humans is associated with abnormal fluid and electrolyte transport in the intestine, which may explain in part the clinical finding of diarrhea in alcoholic patients with chronic folate deficiency. [17], [18]

Folate deficiency and neural tube defects. Neural tube defects are errors of fetal spinal cord development that occur within the first 2 weeks of pregnancy and result in anencephaly and spina bifida. Neural tube defects occur at a rate of 1 to 2 per 1000 live births and probably more frequently in spontaneous abortions. The association of neural tube defects with folate deficiency is based on findings that red cell folate levels in early pregnancy correlate with the risk for delivery of a child with a neural tube defect [19] and on the results of trials in which neural tube defects were prevented by using pre-pregnancy supplemental folic acid. [20] About 20% of infants with a neural tube defect and their mothers have a homozygous C677T mutation of the MTHFR gene, which together with dietary folate, regulates the level of 5-MTHF. [21] The advent of folic acid fortification of the U.S. food supply is expected to significantly reduce the risk for neural tube defects.

Cardiovascular disease. A metaanalysis of 15 studies of proven coronary artery disease showed that an increment of 5 µmol/L in the serum level of homocysteine above the normal range constitutes a coronary heart disease risk similar to an increase in total cholesterol of 20 mg/dL above normal. [22] A multicenter European survey showed that hyperhomocysteinemia exerts a 2.6- to 8.2-fold multiplicative effect on conventional coronary disease risks, including hyperlipidemia, smoking, and hypertension. [23] Hyperhomocysteinemia also is an established risk factor for carotid artery narrowing and occlusive strokes. [24] The results of ongoing prospective trials designed to demonstrate decreased cardiovascular risk with folic acid supplementation are pending.

Colon cancer. A prospective sigmoidoscopic and dietary survey of more than 20,000 health professionals found that the ingestion of diets high in folate was protective against the development of colonic adenomas, whereas a relatively low-folate diet together with the consumption of more than two alcoholic drinks per day increased adenoma risk twofold. [25] In a 6-year follow-up study of more than 40,000 male health professionals, the same investigators found a threefold increased risk of colorectal cancer in alcohol consumers ingesting a low-folate diet, whereas a high-folate diet was protective against colorectal cancer despite alcohol consumption. [26]

Causes of Folate Deficiency

Dietary deficiency. In a landmark self-experiment, removal of folate from the diet was followed by a fall in red blood cell folate and eventual megaloblastic anemia after 140 days. [27] Populations at risk for dietary folate deficiency include young, frequently dieting women, pregnant women who fail to take folate supplements, and the economically deprived, who often include elderly persons with degenerative diseases. Body stores of folate are sufficient for about 4 months; therefore, before folic acid supplementation of the U.S. food supply, the risk for dietary folate deficiency was high in these susceptible populations.

Intestinal malabsorption of dietary folate. Diseases of the intestinal mucosa, such as celiac disease and tropical sprue, affect both folate digestion by GCP II and transport by RFC. [3], [28] Many drugs affect folate absorption and contribute to the risk for folate deficiency. For example, sulfasalazine inhibits both the hydrolysis and transport of folylpolyglutamates, [29] and patients receiving this drug for ulcerative colitis should receive folic acid supplementation. Although the risks for folate deficiency from newer salicylate-releasing antiinflammatory drugs are not known, it has been shown in vitro that the inhibitory property of sulfasalazine on GCP II activity requires both the salicylate and sulfa components. [30] Whereas serum folate levels decrease after exposure to the anticonvulsant phenytoin (Dilantin), [31] a well-conducted study demonstrated that a therapeutic concentration of phenytoin in the intestinal lumen does not block folate absorption. [32]

Chronic alcoholism. Low serum folate levels occur in more than three fourths of chronic alcoholics, and megaloblastic anemia in at least one third. The diet of alcoholics is typically poor in folate; furthermore, alcohol impairs folate absorption and accelerates urinary folate excretion, and the alcohol metabolite acetaldehyde triggers an oxidation reaction that destroys the folic acid molecule. [33], [34] Because chronic liver disease reduces folate stores, the combination of these factors accelerates the risk for the development of folate deficiency in patients with alcoholic liver disease. [35]

Genetic abnormalities. A number of polymorphisms in folate regulatory genes have been described that are related to folate deficiency or increased folate requirement. Examples include C677T and A1298C polymorphisms in MTHFR that are associated with elevated homocysteine levels and an increased incidence of neural tube defects. [21], [36]

Assessment of Folate Deficiency Folate deficiency should be suspected in any anemic patient with an elevated mean corpuscular volume of red blood cells, and then confirmed by a low level of serum or red cell folate. Each of the latter tests can be performed by radioisotope binding assay. The serum folate level is more labile, whereas the red cell folate is a more accurate measure of tissue stores. Measurement of the plasma homocysteine level is a more sensitive functional assay for folate deficiency because 5-MTHF is the substrate for the methionine synthase reaction that converts homocysteine to methionine. However, because cobalamin is an essential cofactor for this reaction, its deficiency is also reflected by an elevated homocysteine level as well as by the same tissue expressions found in folate deficiency (see Fig. 20-1 and Table 20-1). Therefore, folate deficiency can be defined metabolically by a high homocysteine level and normal vitamin B_{12} status as described in the next section.

Vitamin B_{12} (Cobalamin)

Structure, Nomenclature, and Metabolic Functions Vitamin B_{12}, or cobalamin, contains a central cobalt atom surrounded by a planar corrin ring that is formed from four reduced pyrrole rings linked together and attached at a right angle to a nucleotide containing ribose, phosphate, and 5,6-dimethylbenzimadazole. In its active coenzyme forms, the cobalt atom is bound to specific moieties, resulting in methylcobalamin and adenosylcobalamin, whereas cyanocobalamin is the pharmaceutical and interchangeable form of the vitamin. There are only two known enzymatic reactions that require cobalamin and serve to bind it in tissues. Methylcobalamin is the cofactor for methionine synthase and participates in the transfer of the methyl group from 5-MTHF to homocysteine, rendering methionine and tetrahydrofolate (see Fig. 20-1). Thus, cobalamin deficiency disturbs folate metabolism and results in the same abnormalities of cellular regeneration as in folate deficiency, most commonly megaloblastic anemia, and in hyperhomocysteinemia. Adenosylcobalamin is involved in mitochondrial fatty acid metabolism as a cofactor for methylmalonyl-coenzyme A (CoA) mutase in the conversion of methylmalonyl-CoA to succinyl-CoA, so that cobalamin deficiency also uniquely results in elevation of serum methylmalonic acid. [37] In this section, the term *cobalamin* is used to describe dietary sources, metabolic pathways, and clinical manifestations of deficiency. The term *vitamin* B_{12} is used to describe the pharmacological and supplemental forms of the vitamin.

Dietary Sources and Requirements Because cobalamin originates from intestinal bacterial synthesis, it appears in the human diet exclusively bound to animal proteins. The usual sources of cobalamin include meat, fish, shellfish, poultry, eggs, milk, and milk products. The adult DRI is 2.4 µg/d (2.6 µg during pregnancy and 2.8 µg during lactation), which accounts for an average 50% absorption from all dietary sources. Because cobalamin absorption declines with aging, persons older than 50 years are advised to take supplements containing vitamin B_{12} or to ingest food that is fortified with vitamin B_{12}, such as most breakfast cereals. [37]

Intestinal Absorption, Metabolism, and Excretion of Cobalamin

Cobalamin absorption. The intestinal absorption of dietary animal protein–bound cobalamin involves gastric, upper intestinal, and ileal stages, and an enterohepatic circulation (Fig. 20-2). The initial two steps of the gastric phase require an acid pH. In the presence of gastric acid and pepsin, cobalamin is released from its protein bond and then transferred to R-binding protein (haptocorrin) that is present in saliva and gastric juice. The release of cobalamin and its transfer to R factor is optimal at pH 1 to 1.3, a level not achieved by achlorhydric individuals, who include most persons older than 65 years. Similarly, patients using proton pump inhibitors such as omeprazole do not absorb food-bound protein normally [38] and are presumably at risk for cobalamin deficiency. The third gastric step involves the secretion of an additional cobalamin-binding protein, intrinsic factor (IF), from acid-secreting parietal cells. However, IF is ineffective as a cobalamin binder at the acid pH of the healthy stomach, and it follows the haptocorrin-cobalamin complex into the duodenum. Here, the complex is degraded by pancreatic proteases, and at the neutral pH of the duodenum, cobalamin is quickly bound to the protease-resistant IF and in this state traverses the small intestine to the ileum.

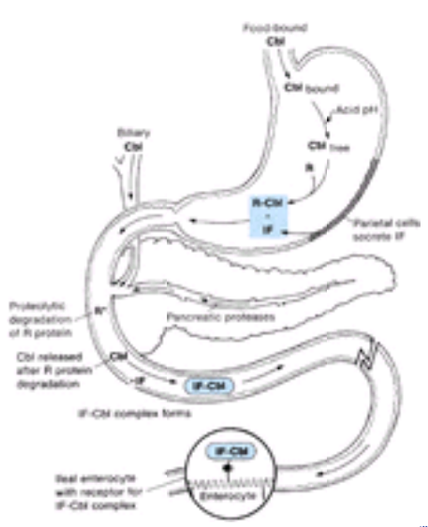

FIGURE 20-2. Sequential steps in the gastrointestinal absorption of dietary protein-bound cobalamin (*Cbl*). Gastric acid is required to liberate both methylcobalamin and adenosylcobalamin from dietary protein. In the acidic gastric environment, cobalamin is then bound to salivary R proteins, whereas gastric parietal cells secrete intrinsic factor (*IF*). After neutralization of the gastric contents in the upper duodenum and pancreatic protease digestion of the R-cobalamin complex, free cobalamin then binds to IF. After transfer to the terminal 50 cm of ileum, the IF-cobalamin complex interacts at the microvillus surface with a specific ileal receptor.

According to retrospective studies in patients with ileal resections of various lengths, at least 50 cm of the terminal ileum participates in the ileal transfer of cobalamin. [39] Ileal absorption of cobalamin requires several binding proteins and may take up to 6 hours for completion. The cobalamin-IF complex is bound to a specific membrane receptor on the ileal enterocyte in the presence of ionized calcium at a neutral pH. Characterization of the cobalamin-IF receptor, named *cubulin,* described a 460-kd glycoprotein that is present on the epithelial surface and colocates with an endocytic receptor, called *megalin,* in both ileal and renal tubular cells. [40] Subsequent transfer across the ileal enterocyte involves endocytic cleavage of the cobalamin-IF complex and then binding of cobalamin to transcobalamin II (TCII). TCII is a 43-kd protein with molecular homology to IF that is synthesized in ileal enterocytes and carries cobalamin out of the ileal mucosa to the circulation. [41] The TCII-cobalamin complex, or holotranscobalamin, accounts for 20% of all circulating cobalamin and is essential for its transport to all tissues. About half of the endogenous circulating pool of cobalamin, or 1.4 µg, is secreted into the bile daily, from which half again undergoes enterohepatic cycling and the other half, 0.7 µg/d, is excreted in the stool. Endogenous cobalamin is secreted into the duodenum bound to biliary haptocorrin; it is then released by pancreatic trypsin and rapidly bound to IF for subsequent passage to the ileal receptor site for intestinal reabsorption.

Clinical Expressions of Cobalamin Deficiency Because folate and cobalamin are required as substrate and cofactor for the methionine synthase reaction, the clinical expression of cobalamin deficiency as megaloblastic anemia and hyperhomocysteinemia is indistinguishable from that of severe folate deficiency. However, because cobalamin is also required in the methylmalonyl-CoA mutase reaction, its deficiency may be expressed in the nervous system as subacute combined degeneration of the posterior columns of the spinal cord, which is characterized by loss of position and vibratory sensation in the lower extremities. Cobalamin deficiency has more subtle and frequent effects on the central nervous system that include neuropsychiatric disorders such as ataxia, paresthesias, memory loss, cognitive dysfunction, and more severe dementia, in the absence of anemia. [42]

Causes of Cobalamin Deficiency Cobalamin deficiency has many potential causes owing to the complexity of the mechanisms of its absorption processes.

Dietary inadequacy. The daily requirement of cobalamin (2.4 µg/d) is very small compared to its body pool size (1–2 mg); therefore, it takes many years to become deficient from dietary inadequacy alone. Only true vegans are at risk for dietary cobalamin deficiency, as cobalamin is present in dairy products and in most multivitamins.

Abnormal gastric events. Because gastric acid is required for the release of food-bound cobalamin as the first step in cobalamin absorption, achlorhydric individuals are at risk for cobalamin deficiency. Studies have demonstrated that achlorhydria of aging is the most common cause of cobalamin deficiency in the United States. About 15% of ambulatory individuals older than 65 years are cobalamin deficient, according to the criteria of elevated methylmalonic acid and homocysteine levels and a serum cobalamin level of less than 300 pg/mL. [43] Gastric infection with *Helicobacter pylori* may predispose to more severe malabsorption of food-bound cobalamin. [44] Because cobalamin deficiency secondary to achlorhydria is caused by malabsorption of food-bound dietary cobalamin but not of unbound crystalline pharmaceutical vitamin B_{12}, its deficiency in elderly persons can usually be prevented by supplemental oral vitamin B_{12}. Similarly, gastric bypass surgery for obesity is associated with food-bound cobalamin malabsorption secondary to achlorhydria, and responds to oral doses of 350 µg of crystalline vitamin B_{12} supplement per day. [45] Pernicious anemia is an autoimmune disorder characterized by the absence of IF in the gastric juice and is associated with achlorhydria and hypergastrinemia. Antibodies to parietal cells can be found in 85% of cases, and blocking and binding antibodies to IF occur in 70% and 50% of cases, respectively. The type A atrophic gastritis of pernicious anemia involves the fundus and upper body of the stomach, in contrast to the more common type B atrophic gastritis of elderly patients, which involves the lower stomach and antrum. According to one survey, the rates of pernicious anemia among elderly men and women are 2.7% and 1.4%, respectively, with a somewhat greater incidence in African Americans than in whites. [46] Because patients with pernicious anemia lack IF, in addition to being achlorhydric, they can absorb neither crystalline cobalamin nor dietary food-bound cobalamin and must be treated with parenteral vitamin B_{12}, usually in monthly 1000-µg doses. Because IF is also required for the absorption of endogenous cobalamin in the enterohepatic circulation, cobalamin deficiency develops more rapidly in patients with untreated pernicious anemia than in strict vegans or elderly persons with achlorhydria.

Abnormal duodenal and jejunal events. Persistent duodenal hyperacidity may inactivate pancreatic trypsin and prevent the transfer of cobalamin from gastric haptocorrin (R factor) to IF; thus, it is not surprising that cobalamin deficiency is a recognized component of the Zollinger-Ellison syndrome. [47] Although cobalamin malabsorption was described in 50% of patients with pancreatic insufficiency, its deficiency is rare in this condition. [48] During passage through the jejunum, the IF-cobalamin complex is susceptible to bacterial cleavage or uptake, and cobalamin deficiency with macrocytic anemia is a recognized risk factor for bacterial stasis syndromes that occur with intestinal blind loops, strictures, and motility disorders such as scleroderma and diabetic visceral neuropathy. Intestinal infestation with the fish tapeworm *Diphyllobothrium latum,* which is found in rural Finland, also causes cobalamin deficiency through competition for the cobalamin-IF complex.

Abnormal ileal events. Because cobalamin-IF receptors are located on enterocytes in the distal 50 cm of ileum, diseases or surgical resection involving this length of small bowel are usually associated with cobalamin malabsorption and deficiency. These include Crohn's disease, radiation enteritis, severe celiac disease, and human immunodeficiency virus infection, and the short-bowel syndrome secondary to surgical removal of terminal ileum. Resection of the ileocecal valve increases the risk for cobalamin deficiency by permitting intestinal bacterial overgrowth. Because the terminal ileum is the site for both dietary cobalamin absorption and the reabsorption of endogenous cobalamin from its enterohepatic circulation, cobalamin deficiency may become evident within several months of surgical resection of more than 50 cm of terminal ileum. Two drugs, colchicine and *p*-aminosalicylic acid, may cause cobalamin deficiency by inhibiting its uptake by the ileal mucosa. [49], [50]

Genetic causes of cobalamin malabsorption. Congenital deficiency of IF results from the failure of its synthesis by gastric parietal cells. The gastric mucosa and acid secretion are normal and IF antibodies are absent in this condition. Cobalamin deficiency is usually apparent between the ages of 1 and 5 years. [51] Defective cobalamin transport by ileal enterocyctes (Imerslünd-Grasbeck syndrome) has been recognized most often in Finland and other Scandinavian countries. It is clinically evident in early childhood but has also been described in young adults. [52] These patients have normal gastric function and ileal histology. This autosomal recessive disease represents failure of synthesis or membrane insertion of cubulin and is expressed in both the ileum and renal tubule by the early onset of cobalamin deficiency and proteinuria. Congenital TCII deficiency is also autosomal recessive and has been described rarely in several families. It is expressed in the first year of life with pancytopenia and megaloblastic anemia. [52]

Measurements of Cobalamin Status Cobalamin deficiency should be suspected in any person who presents with macrocytic anemia and an elevated mean corpuscular red cell volume. However, because most cases of cobalamin deficiency are expressed as neuropsychiatric abnormalities, particularly as cognitive loss in the aged, it is important to suspect and detect deficiency even in the absence of hematologic abnormalities. Cobalamin is conventionally measured in the serum by a radioassay with high specificity and sensitivity. Normal values depend on the laboratory and generally range from 200 to 250 pg/mL. However, based on a comparative study in which serum elevation of the metabolite methylmalonic acid was used as the gold standard, an upper limit of at least 300 pg/mL has been recommended. [43] Although more expensive, the most accurate assessment of cobalamin status is by serum level measurement of its metabolites homocysteine and methylmalonic acid. [53] Cobalamin deficiency is distinguished by elevation of serum methylmalonic acid, as folate deficiency also elevates homocysteine owing to its role as substrate in the methionine synthase reaction. [54] Because metabolically effective circulating cobalamin is bound to TCII, measurement of serum holotranscobalamin is considered a more precise indication of cobalamin status than is the serum cobalamin assay. Difficulties in standardizing the holotranscobalamin assay may be resolved by the development of an immunodetection test. [55]

Clinical Tests of Cobalamin Absorption The etiology of cobalamin deficiency can be assessed by the Schilling test through measurements of urine excretion of label after oral administration of a physiological amount (0.5–2 µg) of radioactive ^{57}Co-cobalamin. Because a 1000-mg tissue-saturating ("flushing") dose of parenteral cobalamin is given 2 hours later, the cumulative appearance of the label in the 48-hour urine excretion is representative of the intestinal absorption of the radioactive cobalamin. To screen all possibilities for malabsorption, two approaches can be used. The food cobalamin test measures the first gastric phase of acid release of protein-bound cobalamin. [56] Practically, the egg yolk cobalamin absorption test (EYCAT) requires initial incubation of labeled cobalamin with egg yolk followed by oral administration and the flushing dose; normal results are usually greater than 2% excretion of the label. Although not universally available, only the EYCAT will distinguish cobalamin deficiency secondary to achlorhydria because achlorhydric patients are incapable of absorbing food-bound cobalamin but are capable of absorbing the widely available unbound crystalline form of cobalamin. The time-honored and widely available Schilling test of crystalline cobalamin absorption is designed to detect and define etiologies that include IF deficiency, as in pernicious anemia, ileal malabsorption, and bacterial stasis. Cobalamin deficiency should first be treated because ileal macrocytosis may impair cobalamin absorption. If the result of stage I of the Schilling test is normal (usually >9% recovery in a 48-hour urine collection), pernicious anemia is ruled out and cobalamin deficiency may be secondary either to diet, as in strict vegans, or to gastric achlorhydria, as occurs in the elderly. If the stage I result is abnormally low, pernicious anemia is confirmed by correction of the test result by provision in stage II of an exogenous source of IF together with the labeled cobalamin. A persistently abnormal stage II test result can be attributed to abnormal ileal absorption or intestinal bacterial competition for the cobalamin-IF complex. This issue can be resolved by the stage III test, in which the absorption of labeled cobalamin is improved or corrected after 2 weeks of oral antibiotic suppression of intestinal bacteria. Although based on physiological principles, the full battery of Schilling tests is cumbersome, requires accurate and complete patient urine collection, and therefore may produce false-positive results.

Other Water-Soluble Vitamins

Several other water-soluble vitamins are relevant to the practice of gastroenterology and hepatology. These include vitamin C (ascorbic acid), thiamin, pyridoxine, niacin, and riboflavin. Each is considered separately, with emphasis on requirement, mechanism of absorption, diagnostic tests, and clinical expression of deficiency (Table 20-2).

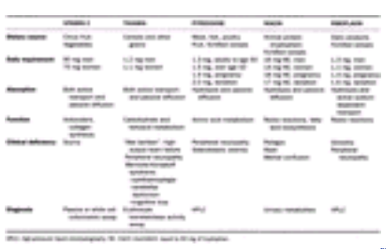

TABLE 20-2 Other Water-Soluble Vitamins

Vitamin C The term *ascorbic acid* derives from the antiscorbutic properties of vitamin C (prevention or treatment of scurvy). Vitamin C is found abundantly in citrus fruits and many vegetables. The DRI of vitamin C is 90 mg/d for men and 75 mg/d for adult women, with an additional 35 mg/d for smokers, in whom vitamin C turnover is increased. [57] Vitamin C functions as both a reducing agent and an antioxidant, as a cofactor for metalloenzymes, and as an electron donor in enzymes involved in collagen, carnitine, and catecholamine synthesis and in tyrosine metabolism. Vitamin C is absorbed equally well from natural foods and supplements by active, sodium-dependent transport at low concentrations and by passive diffusion at high concentrations. [58] The percentage of vitamin C absorbed decreases with increasing intralumenal concentrations and falls to less than 50% after the ingestion of 1g, with a resultant risk for osmotic diarrhea. [59] The maximal tissue saturation of vitamin C is achieved in healthy persons with oral intakes of 90 to 150 mg/d, and excess absorbed vitamin is metabolized to oxalate or excreted in the urine. In human metabolic turnover experiments in subjects deprived of vitamin C, the body pool decreased by 3% per day, with the development of signs of scurvy from the 55th day onward. [60] In modern medicine, scurvy is found in alcoholics, patients with severe celiac disease, severely anorectic patients, and persons who smoke heavily. The sequential clinical signs of scurvy relate to increased capillary fragility and include bruising of the skin, bleeding gums, and a characteristic perifollicular hemorrhagic rash in the lower extremities. In more extreme cases, scurvy results in joint effusions, vascular instability, and death. Vitamin C is assessed by a colorimetric assay; serum levels represent recent intake, whereas white blood cell levels represent tissue stores. [61]

Thiamin Thiamin, in its active form as thiamin pyrophosphate, is involved in carbohydrate metabolism as a coenzyme for pyruvate dehydrogenase and for the decarboxylation of a-ketoacids. Thiamin is present in fortified or whole grain cereals and breads, and in pork. The DRI for thiamin is 1.2 mg for men and 1.1 mg for women (1.4 g during pregnancy and lactation). [62] Thiamin is absorbed by a dual process in the jejunum. At low concentrations (0.2–2.0 mM), thiamin is absorbed by an active Na^+-dependent transport that involves phosphorylation, and at higher concentrations, it is absorbed by passive diffusion. [63] The body pool size of thiamin is about 30 mg, with a half-life of 9 to 18 days, and plasma and red cell levels are sustained by rapid urinary excretion of excess absorbed thiamin. Thiamin deficiency occurs during extreme malnutrition of any cause and is common in persons with chronic alcoholism because of a combination of poor intake and reduced absorption. Ethanol interferes with Na^+, K^+-ATPase–mediated active transport across the basal enterocyte membrane that drives thiamin absorption. [64] The clinical signs of thiamin deficiency are related to the heart and the nervous system. Classical wet beriberi, which often accompanies starvation, is characterized by cardiomyopathy with high-output cardiac failure, whereas the main sign of dry beriberi is progressive stocking-glove polyneuropathy of the lower extremities. Additional neurological features of thiamin deficiency occur mainly in chronic alcoholics and are classified under the Wernicke-Korsakoff syndrome as nystagmus and ocular paresis, cerebellar dysfunction with a wide-based gait, and global confusion. [65] Because body stores of thiamin are regulated by carbohydrate metabolism, the administration of intravenous glucose to a malnourished patient may precipitate an acute demand for residual endogenous thiamin with resultant overt thiamin deficiency made manifest by cardiac failure and confusion. Thiamin status is most accurately assessed by a functional assay of erythrocyte transketolase (ETKA) before and after the addition of thiamin pyrophsophate. [61]

Pyridoxine Pyridoxine, or vitamin B_6, is a coenzyme for many amino acid reactions and exists in six forms: pyridoxine, pyridoxal, pyridoxamine, and the phosphorylated forms of each. Its main food sources include meat, fish, poultry, noncitrus fruits, and fortified cereals. The DRI is 1.3 mg/d for adult men and women; it rises to 1.5 mg/d for those older than 50 years, 1.9 mg for pregnant women, and 2.0 mg for women who are lactating. [66] The phosphorylated forms of pyridoxine are absorbed by hydrolysis by brush border alkaline phosphatases, followed by passive transport of nonphosphorylated forms through the enterocyte. Phosphorylation is completed in the liver, and the principal circulating form is pyridoxal phosphate (PLP) in a complex with albumin. The body pool size of pyridoxine is 60 to 100 mg. Pyridoxine deficiency is one of the common features of chronic alcoholism and, owing to the role of pyridoxine in heme synthesis, presents with microcytic sideroblastic anemia, peripheral neuropathy, cheilosis, and glossitis. Pyridoxine deficiency in alcoholism is caused by hepatic degradation of the PLP-protein complex by acetaldehyde followed by rapid urinary excretion of the unbound vitamin. [67] The more than twofold elevation of serum aspartate aminotransferase relative to alanine aminotransferase characteristic of patients with alcoholic hepatitis may be caused by the greater requirement of the latter enzyme for PLP. [68] Because pyridoxine is an essential cofactor for cystathionine ß-synthase in the transsulfuration elimination pathway of homocysteine (see Fig. 20-1), its deficiency contributes to hyperhomocysteinemia in elderly patients and other susceptible individuals. Pyridoxine toxicity was described in women in whom severe sensory neuropathy of the hands and fingers developed after ingestion of 2 to 6 g/d for 2 months for premenstrual stress. [69] Plasma PLP can be measured by high-performance liquid chromatography (HPLC). [61]

Niacin Niacin is expressed biologically by its two coenzymes, nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP), which are involved in redox reactions, the oxidation of fuel molecules, and fatty acid biosynthesis. Dietary niacin requirements are based on nicotinic acid, nicotinamide, and tryptophan, which is metabolized to niacin such that 60 mg of tryptophan is equivalent to 1 mg of niacin. Dietary niacin is derived from animal protein as tryptophan, and from enriched or whole grain cereals and bread. The DRI of niacin is 16 mg of niacin equivalents (NEs) for men and 14 mg for women, with 18 mg for pregnancy and 17 mg for lactation. [70] Niacin is absorbed as NAD from the upper small intestine. Pellagra, the classical disease of niacin deficiency, is

brought about by insufficient dietary animal protein as a source of tryptophan and is manifested by mental confusion, a characteristic scaly red rash of sun-exposed areas, and diarrhea. Still prevalent in certain developing countries where animal protein is scarce, pellagra is occasionally identified in chronically alcoholic patients, usually in association with other vitamin deficiencies. Niacin in doses exceeding 1 g/d is often used in the treatment of hyperlipidemia. The common toxic side effects of flushing and exacerbation of peptic ulcer disease are greatly minimized by use of a controlled-release preparation. [71] Niacin status is assessed by measurement of the urinary ratio of its metabolites, *N*-methyl-2 pyridone-5 carboxamide and *N*-methylnicotinamide. [61]

Riboflavin Riboflavin, or vitamin B_2, exists in coenzyme forms as flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD), each of which is involved in redox reactions. The food sources of riboflavin include milk, other dairy products, and fortified cereals. The DRIs for riboflavin are 1.3 mg for men, 1.1 mg for women, 1.4 mg for pregnant women, and 1.6 mg for lactating women. [72] Dietary FAD and FMN are released from food protein by gastric acid, then hydrolyzed by intestinal phosphatases. [73] Intestinal riboflavin transport is regulated by a Na^+-independent, carrier-mediated system. [74] Riboflavin deficiency is manifested by intense glossitis, seborrheic dermatitis, and peripheral neuropathy and may occur with alcoholism, celiac disease, and anorexia of any cause. Riboflavin status is assessed by HPLC of urine or by colorimetric measurement of erythrocyte glutathione reductase before and after the addition of FAD. [61]

FAT-SOLUBLE VITAMINS

Vitamins A, D, E, and K are unlike the water-soluble vitamins by virtue of their diverse nonenzymatic functions, prolonged storage and greater risk for toxicity, and absorption processes similar to those of dietary lipids in general. Like dietary triglycerides, fat-soluble vitamins are solubilized in the duodenal lumen in the presence of bile and pancreatic enzymes, and are then maintained within the lipophilic core of mixed micelles. Pancreatic esterases release fat-soluble vitamins from their esters in the presence of bile salts. Together with triglyceride-derived fatty acids, fat-soluble vitamins are released from micelles at the enterocyte brush border membrane. The absorption of vitamins E and K is decreased in the presence of high doses of vitamin A, whereas vitamin A absorption may be enhanced in the presence of high doses of vitamin E. [75] At high doses, vitamins A and E can be absorbed directly from water-miscible emulsions. [76] Within proximal enterocytes, fat-soluble vitamins are incorporated into chylomicrons for secretion into lymphatics and ultimate uptake by the liver. Intestinal, biliary, and pancreatic diseases that affect dietary lipid absorption also decrease the absorption of fat-soluble vitamins and promote their deficiencies. Table 20-3 summarizes the sources, requirements, functions, subsequent mechanisms of absorption, and clinical features of each fat-soluble vitamin.

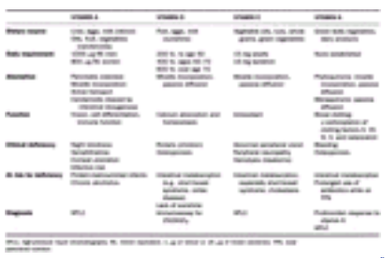

TABLE 20-3 Fat-Soluble Vitamins

Vitamin A

Vitamin A includes provitamin dietary carotenoid precursors of retinol and dietary retinol in its esterified form. Retinol is found exclusively in liver, eggs, and milk products; carotenoids are present in oils, fruits, and vegetables. Within the intestinal lumen, dietary retinyl esters are hydrolyzed by a specific pancreatic esterase to retinol, which is incorporated into micelles, then transported into enterocytes by a specific carrier protein. Transport is active and saturable at low concentrations, and passive at high concentrations. Within the enterocyte, retinol is transported by retinol-binding protein (RBP) to the smooth endoplasmic reticulum for reesterification, incorporation into chylomicrons, and subsequent transport as retinyl esters via lymphatics to the circulation and liver.

Dietary carotenoids undergo the same micellar incorporation as retinol and are passively transported across the cell membrane. Within the enterocyte, each molecule of pure ß-carotene is cleaved by ß-carotene dioxygenase to two molecules of retinol, which are esterified. Retinyl esters are stored in the liver as fat droplets in hepatic stellate cells, and after hydrolysis, they circulate as retinol bound to RBP. Carotenoids that reach the liver intact are transported with very-low-density lipoprotein (VLDL) and are converted to retinoids by tissue dioxygenases. [77]

Because the overall availability of purified ß-carotene in oil is only half that of retinol, the retinol activity equivalent (RE) of supplemental ß-carotene is considered to be 2:1. Considering the average 8% (1/12) availability of dietary ß-carotene in mixed vegetable and fruit diets, its RE is estimated at 12:1 × 2, or 24:1. In other words, after absorption and bioconversion, 24 µg of mixed dietary carotenoids is required to yield the same amount of retinyl esters as 1 µg of dietary retinol. [77] The DRIs for vitamin A are 400 to 700 µg RE for children, 1000 µg RE for adult men, and 800 µg RE for adult women. [77]

The biologic functions of vitamin A include the maintenance of vision through the conversion of retinal to rhodopsin, which enhances rod membrane potential for transmitting light signals to the optic nerve. Vitamin A is also essential for gene expression and cell differentiation during embryonic development, cell-mediated immune responsiveness, and spermatogenesis. The signs of vitamin A deficiency include failure of dark adaptation (night blindness), a follicular hyperkeratotic rash of the extremities, and impaired resistance to infections. Vitamin A deficiency occurs worldwide in protein-malnourished children, in whom night blindness and xerophthalmia develop with the appearance of conjunctival white deposits (Bitot spots), eventual corneal ulceration, scarring, and permanent blindness. Decreased hepatic RBP synthesis is a prominent feature of protein-calorie malnutrition and is the principal cause of vitamin A deficiency in malnourished children. International vitamin A supplementation programs have enhanced innate immune function and significantly reduced life-threatening infections in children worldwide. [78] According to levels in liver biopsy specimens, vitamin A deficiency is nearly universal in patients with alcoholic liver disease because of the effect of the microsomal CYP2E1 ethanol-oxidizing enzyme on enhancing the catabolism and biliary excretion of hepatic retinoids. [79] In the setting of chronic alcoholism, vitamin A deficiency may potentiate the development of esophageal squamous cell cancer. [80] On the other hand, the signs of supplemental vitamin A hepatotoxicity are enhanced by alcoholism, and excessive alcohol consumption interferes with the metabolism of supplemental ß-carotene and may enhance the propensity for lung cancer in alcoholic persons who smoke. [81] Vitamin A toxicity can occur with prolonged daily ingestion of 25,000 to 50,000 IU in easily obtained over-the-counter supplements and can result in liver failure, headache, vomiting, and a desquamating rash. [82]

Vitamin A status is measured by HPLC of plasma or serum by a method that is applicable to both vitamins A and E. RBP can be measured by radial immunodiffusion kit assay (The Binding Site, San Diego, California). The relative dose-response assay is used in field studies and consists of measuring RBP before and after a standard oral dose of vitamin A. Because apo-RBP accumulates in the liver during vitamin A deficiency, an increase of circulating RBP is indicative of vitamin A deficiency. [61] However, plasma RBP is also decreased in severe protein-calorie malnutrition, acute and chronic infection, and trauma. Liver biopsy is the gold standard for assessment of vitamin A status; however, it should be performed only if clear clinical indications of deficiency are present. The dichotomy of normal serum levels but low hepatic levels of vitamin A was demonstrated in a study of patients with liver disease of different etiologies. [79]

Vitamin D

Vitamin D is the generic term for a group of sterols that regulate calcium absorption and homeostasis. The forms of vitamin D include vitamin D_3 (cholecalciferol), which is the natural active form of the vitamin, and vitamin D_2 (ergocalciferol), its synthetic form. Vitamin D_3 is a provitamin that is synthesized in skin from endogenous 7-dehydrocholesterol on exposure to sunlight and is found in the diet in oily fish, egg yolks, and fortified milk. Persons living in temperate and tropical climates receive most of their vitamin D_3 by synthesis in the skin; vitamin D deficiency is more common in more northern or southern geographic areas, where sunlight is less constant. [83] The DRI for vitamin D is not precise and depends on the degree of exposure to the sun. The DRI for healthy adults ages 50 years and younger is 200 IU (equivalent to 5 µg/d), 400 IU/d for persons ages 50 to 70, and 600 IU/d for those older than 70 years. [84]

Like other fat-soluble vitamins, dietary vitamin D is absorbed after micellar solubilization in the upper intestine and by passive diffusion across the enterocyte membrane, then is incorporated into chylomicrons for entry into the circulation via lymphatics followed by uptake by the liver. [84] Two hydroxylation steps are required

for the activation of vitamin D. 25(OH)D_3 is synthesized in the liver and is both the principal circulating form and the substrate for 1a,25(OH)$_2D_3$, the active form of the vitamin that is synthesized in the proximal renal tubular cells. Renal 1-hydroxylase is tightly regulated by circulating levels of parathyroid hormone, calcium, and 1a,25(OH)$_2D_3$. Thus, 1a,25(OH)$_2D_3$ levels are increased by relative hypocalcemia that raises the level of parathyroid hormone. The main action of 1a,25(OH)$_2D_3$ is to maintain the circulating calcium level through regulation of its intestinal uptake, whereas postabsorptive circulating calcium regulates bone resorption by suppressing the release of parathyroid hormone. 1a,25(OH)$_2D_3$ also regulates gene transcription by interacting with vitamin D membrane receptors (mVDRs) and nuclear receptors (nVDRs) in many different tissues. [85]

Dietary vitamin D deficiency reduces calcium absorption, secondarily increasing parathyroid hormone and the risk for osteomalacia in adults or rickets in children, which is compounded by dietary calcium deficiency. On the other hand, osteoporosis in postmenopausal women and elderly men is caused by a lack of estrogen or androgen-derived estrogen, which increases bone resorption and thereby enhances serum calcium levels, resulting in reduced parathyroid hormone secretion and, thus, reduced renal production of 1a,25(OH)$_2D_3$. Consequently, calcium absorption and the serum levels of 25(OH)D_3 and 1a,25(OH)$_2D_3$ decline with aging, all of which contribute to the risk for bone loss in the elderly. In patients with malabsorption diseases characterized by decreased biliary secretion and micelle formation, such as primary biliary cirrhosis or the short-bowel syndrome, or by decreased mucosal lipid absorption, such as celiac disease, vitamin D and calcium are also malabsorbed, and combinations of osteomalacia and osteoporosis develop prematurely. For example, elevated gliadin antibody levels of celiac disease were tenfold more common among osteoporotic than nonosteoporotic Swedish adults. [86] Vitamin D toxicity can occur by excessive over-the-counter use of a vitamin D supplement and is characterized by hypercalcemia, nephrocalcinosis, and extremely elevated serum levels of 25(OH)D_3. [87]

Vitamin D deficiency is assessed by the findings of decreased circulating levels of 25(OH)D_3 and 1a,25(OH)$_2D_3$. [61] Decreased urinary calcium excretion, elevated serum parathyroid hormone, bone-derived alkaline phosphatase, and urinary hydroxyproline are all indicative of increased bone resorption in vitamin D deficiency. The rate of bone loss and the severity of osteoporosis can be partially alleviated by providing supplemental vitamin D in a dose of up to 800 IU/d together with supplemental calcium. [88] The amount of vitamin D required to treat deficiency in intestinal fat malabsorption syndromes is not established; one study demonstrated failure to increase serum levels of 25(OH)D_3 with single vitamin D_3 doses as high as 50,000 IU. [89] Nevertheless, it is worth providing large doses of vitamin D three times weekly to such patients while monitoring carefully for elevated levels of serum 25(OH)D_3 and urinary calcium excretion.

Vitamin E

Vitamin E comprises eight tocopherols, of which two, a- and ?-tocopherol, are significant to human nutrition. The predominant natural form of vitamin E is the *RRR* isomer of a-tocopherol. The major food sources of vitamin E are polyunsaturated vegetable and seed oils, whole grains, nuts, and green leafy vegetables. The DRIs of vitamin E are 6 to 11 mg for children, 15 mg for adults of all ages, and 19 mg for lactating women. [90]

The intestinal absorption of dietary vitamin E includes deesterification by pancreatic esterases, followed by bile-dependent incorporation into intralumenal micelles, subsequent passive diffusion into enterocytes, and incorporation into chylomicrons for transfer to the lymphatics and the circulation. After peripheral hydrolysis of chylomicrons, vitamin E returns to the liver with chylomicron remnants and then is transferred by hepatic a-tocopherol transfer protein (a-TTP) to *RRR*-a-tocopherol in combination with VLDL. After peripheral hydrolysis of VLDL, *RRR*-a-tocopherol is transferred to high- or low-density lipoprotein (HDL or LDL). [91] Subsequent transfer to tissues follows uptake of these lipoproteins. Plasma levels of a-tocopherol are determined by HPLC and vary according to the total plasma lipid concentration, [92] or, because a-tocopherol is transported with HDL and LDL, as a ratio to total cholesterol. [61]

The primary function of a-tocopherol is as a potent antioxidant of lipids. During antioxidant reactions, a-tocopherol accepts the free radical and becomes oxidized, and then is regenerated by vitamin C or glutathione. Vitamin E may also modulate cell-mediated immune function in elderly patients, [93] and may play a preventive role in coronary heart disease through its antioxidant effect on LDL. [94]

Vitamin E can be measured by a HPLC assay that is applicable also to vitamin A. [61], [95] Because vitamin E is ubiquitous in the diet, its deficiency occurs mainly in patients with malabsorptive disorders involving the biliary circulation, pancreas, and intestinal mucosa. Because their vitamin stores are lower, infants and children with biliary atresia or other causes of cholestasis acquire vitamin E deficiency more rapidly than do older children and adults with malabsorption diseases, including cystic fibrosis, cholestatic liver disease, short-bowel syndrome, and celiac disease. [96] Patients with chronic hemolytic anemia may become deficient in vitamin E owing to its overutilization, whereas acute hemolytic anemia may develop in preterm infants fed diets high in polyunsaturated fatty acids and pro-oxidant iron. Vitamin E deficiency is manifested also by neurological damage involving the posterior columns, cranial nerves, brainstem, and peripheral nerves, and by retinal damage, presenting clinically with loss of balance, peripheral neuropathy, or visual field defects. [97] A water-soluble form of vitamin E, *RRR*-a-tocopherol glycol (Aquasol E), is better absorbed than the dietary, fat-soluble natural vitamin and hence is more effective in the treatment or prevention of vitamin E deficiency in malabsorption diseases such as short-bowel syndrome. [76]

Vitamin K

Vitamin K incorporates dietary phylloquinone from plants and menaquinones that are synthesized by intestinal bacteria. The main dietary sources of phylloquinone are leafy green vegetables; dairy products are minor sources. There is no precise DRI, and the average intake of vitamin K is 120 mg in men and 90 mg in women. [98] Menaquinone is absorbed by passive diffusion from the distal ileum in the presence of bile salts, and less efficiently from the colon. [99] In contrast, dietary lipid-soluble phylloquinone undergoes micellar incorporation, after which it is transported into the enterocyte by active saturable transport system. [100] Comparative studies of the forms of vitamin K in the liver suggest that menaquinone from bacterial synthesis provides substantially less vitamin K than dietary phylloquinone.

Vitamin K facilitates the posttranslational ?-carboxylation of proteins involved in blood clotting: prothrombin and factors VII, IX, and X. The presence of ?-carboxyglutamic acid (Gla) residues is essential for calcium binding, as required in blood clotting. [101] Vitamin K also enhances ?-carboxylation of osteocalcin, and vitamin K deficiency contributes to osteoporosis, whereas vitamin K supplementation has been shown to prevent bone fractures. [102]

Vitamin K deficiency can be assessed by a prolonged prothrombin time that responds to parenteral vitamin K administration and by direct measurement of circulating phylloquinone by HPLC. [61] The clinical settings of vitamin K deficiency include the combination of dietary inadequacy and antibiotic use, such as may occur in patients with sepsis receiving total parenteral nutrition; patients with malabsorption syndromes, including hepatobiliary diseases associated with cholestasis, short-bowel syndrome, Crohn's disease, and celiac disease; and patients on prolonged antibiotic regimens. [103]

MINERALS

This section begins with discussion of the absorption of the macrominerals, calcium and magnesium, which always are present in biologic systems in the divalent form and do not undergo any redox reactions. Iron, the most abundant trace element, is discussed next. Iron occurs in several physiologically different forms, has complex absorption mechanisms, and undergoes several oxidation-reduction reactions during its normal metabolism. Finally, the absorptive mechanisms for zinc and copper are presented. Table 20-4 summarizes the dietary sources, requirements, absorptive mechanisms, functions, and clinical features of each of these minerals.

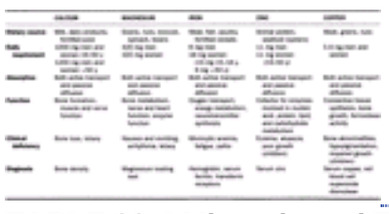

TABLE 20-4 Minerals and Trace Elements

Calcium

Sources and Bioavailability Although diet previously provided the major part of the daily calcium intake, recognition of the importance of calcium in osteoporosis and bone health has changed this pattern. Calcium supplements and fortified beverages and foods are now commonly used and are needed to meet the recently increased dietary requirements. Milk and other dairy products are good dietary sources and often provide up to 75% of dietary calcium. Meat products provide some calcium bound to protein, whereas calcium in plants usually is associated with organic anions, such as phytate and oxalate. The DRI is 1000 mg/d for adults (19–50 years), 1300 mg/d for teenagers and young pregnant and lactating women, [84] and 1200 mg/d for adults older than 50 years. The bioavailability of calcium from milk, dairy products, and meat is high, perhaps because the smaller peptides formed during digestion keep calcium in an absorbable, soluble form. [104] In contrast, phytate, uronic acid, and particularly oxalate, which are present in cereals, fruits, and vegetables, reduce the absorption of calcium. [105], [106] Fiber that contains phytate has been shown to have a negative effect on calcium absorption. However, the results have not been consistent and vary among different types of dietary fiber, possibly because of different contents of phytate. It appears that some forms of calcium supplements are less bioavailable than others. The most absorbable form is calcium citrate malate; calcium is somewhat less available from calcium carbonate and calcium triphosphate. [107] Although not directly affecting calcium absorption, high intakes of caffeine and protein have a negative effect on calcium balance by increasing urinary losses. [105] Furthermore, when supplements are taken with or close to a meal or if calcium-fortified foods are consumed, the absorption of calcium will be the net result of interactions between enhancers and inhibitors of calcium absorption present in the meal. Finally, there is strong homeostatic regulation of calcium absorption that may be more profound than the dietary factors discussed above.

Absorption and Homeostasis Calcium is absorbed both actively in the duodenum and passively by diffusion in the ileum. [106] Some absorption also occurs in the colon. [108] Calcium must be ionized at a low pH in the stomach. In patients with achlorhydria, the absorption of calcium from calcium carbonate is decreased compared to that in control subjects with normal gastric pH. [109] In this study, however, the achlorhydric patients absorbed calcium from skim milk to the same extent as subjects with normal stomach acidity, possibly because of the solubilizing effect of milk peptides, which is not as pH-dependent. Antacids can therefore also reduce calcium uptake. Active absorption via calcium-binding protein (CaBP) dominates at low levels of calcium intake, whereas passive diffusion makes a significant contribution to net calcium uptake at high levels of intake. [110] Active transepithelial calcium transport, which is largely localized to the upper part of the duodenum, is dependent on vitamin D (1,25[OH] $_2$D $_3$) and consists of three mechanisms. First, brush border uptake occurs down a steep electrochemical gradient from the lumen to the cytoplasm through calcium channels that are not voltage gated. [106] Second, calcium ions are transported through the enterocyte by carrier-mediated facilitated diffusion from the brush border membrane to the basolateral membrane. [111] This step has been described as rate-limiting in calcium absorption. Diffusion of calcium ions is very low in the absence of the cytosolic CaBP, calbindin D $_{9k}$, and transcellular calcium transport increases in a linear fashion according to the cellular content of calbindin. [112] Another CaBP, calbindin D $_{28k}$, is also induced by vitamin D, but it binds four calcium ions, whereas calbindin D $_{9k}$ binds two. [113], [114] In vitamin D–deficient cells, calcium enters the cell but stays at the brush border membrane, showing the strong need for calbindins for effective cellular translocation. [114] The third mechanism is extrusion of Ca^{2+} through the basolateral membrane, which is mediated by calcium ATPase and the Na^{+}/Ca^{2+} exchanger, occurs against an electrochemical gradient, and consequently requires energy. [115] Calcium interacts with the cytoplasmic region of ATPase and, through a phosphorylation-induced conformation change in the enzyme, is transported across the membrane via a channel formed by the transmembrane regions of ATPase. Vitamin D has some effect on both the uptake and extrusion phases by increasing the gene expression of membrane-bound calcium ATPase. [116] The passive absorption of calcium occurs by paracellular diffusion down a chemical gradient in all sections of the small intestine. [106] This pathway accounts for the major part of calcium absorption when intake is adequate or high, and is associated with down-regulation of the active transport of calcium. Passive diffusion of calcium also occurs in the colon. In rats, the contribution of the colon to net passive uptake of calcium in the entire tract has been estimated at 11%, whereas its contribution to active transport may account for 7%. [106] Similar values for humans are not available, but it has been shown that total calcium uptake by the colon in the human adult is substantial. [117] Homeostatic regulation of calcium absorption is achieved primarily by up- and down-regulation of active transport. [106] During pregnancy, calcium absorption increases significantly, which may be a response to the high fetal requirement. Lactation, however, does not appear to result in a similar increase in calcium absorption.

Disorders of Calcium Homeostasis Intestinal absorption appears to decrease with age, possibly because of lower circulating concentrations of 1,25(OH) $_2$D $_3$ or decreased responsiveness of the intestine to vitamin D. [118] Consequently, increased calcium intake is recommended for older persons to prevent bone loss. Gastrointestinal disease, such as celiac or Crohn's disease, often results in osteomalacia, usually as a consequence of vitamin D deficiency. [119] Cholestasis and ileal disease disrupt the enterohepatic circulation of bile salts, which in turn reduces micelle formation, leading to decreased lipid and vitamin D absorption. Increased fat excretion in the malabsorption diseases also causes losses of calcium bound to fatty acids. Gastric surgery often disturbs calcium absorption, resulting in osteomalacia as a late complication. [119] Disorders of thyroid hormone metabolism also affect calcium homeostasis through effects on parathyroid hormone. [120] In hyperthyroidism, calcium absorption is decreased secondary to suppression of parathyroid hormone secretion and its effect of renal hydroxylation of 25(OH)D $_3$, caused by increased bone resorption and serum calcium concentrations. In hypothyroidism, the hydroxylation of vitamin D to its active form 1,25(OH) $_2$D $_3$ in the kidney is lowered, resulting in lower calcium absorption. Similarly, renal disease can result in lower production of 1,25(OH) $_2$D $_3$ and thus decreased calcium absorption. Therefore, 1,25(OH) $_2$D $_3$ is used in the treatment of chronic renal disease. However, type I renal acidosis and nephrotic syndrome also cause calcium malabsorption, [121] but in these diseases, vitamin D administration is ineffective and treatment with alkali is required, suggesting a negative effect of acidosis on calcium absorption. Drugs can also affect calcium absorption. Whereas some drugs like glucocorticoids and phenytoin directly inhibit calcium absorption in the intestine, chlorothiazide and other diuretics increase the renal reabsorption of calcium and thereby decrease parathyroid hormone secretion and 1,25(OH) $_2$D $_3$ synthesis, indirectly reducing calcium absorption. Other drugs increase calcium absorption. Pharmacological doses of estrogen can enhance calcium absorption by reducing bone resorption and secondary hyperparathyroidism. [122] Vitamin D intoxication also increases calcium absorption. [123]

Magnesium

Sources and Bioavailability Magnesium is abundant in plants; whole grains, nuts, seeds, broccoli, spinach, and beans are good sources of magnesium. Dairy products and meat also contribute significantly to our daily magnesium intake. Recently, the intake of mineral water high in magnesium has become common, and mineral water is now a considerable source of magnesium for many individuals. The newly set DRI for magnesium is 420 mg/d for adult men, which is usually met by most men, and 320 mg/d for women, [124] whose magnesium intake is frequently less than the DRI. Based on disappearance data, the per capita availability of magnesium suggests an intake of about 350 mg/d, although this intake differs considerably from intake data that show about 180 to 200 mg/d for women and 250 to 300 mg/d for men. [125] Increased magnesium intake has been advocated for fertile women, particularly during pregnancy.

Absorption and Homeostasis Studies in experimental animals show that magnesium absorption occurs primarily in the ileum and the colon, whereas less is absorbed in the duodenum. [126] Human studies over a wide range of magnesium intakes show a saturable curve that is indicative of an active transport mechanism, as well as a linear function that is compatible with passive diffusion. [127] Studies in children also show evidence for carrier-mediated intestinal transport of magnesium. [128] Perfusion studies in humans show magnesium absorption in the jejunum and ileum, [129] and also substantial absorption in the colon. Absorption of magnesium in adults with a normal dietary intake of magnesium is about 21% in men and 27% in women. [130] Magnesium homeostasis is largely regulated by control of renal excretion, but increased absorption of magnesium in the intestine during low levels of dietary intake contributes to homeostasis.

Deficiency Magnesium deficiency secondary to low dietary intake is uncommon and difficult to diagnose because of a lack of reliable markers. [131] Serum or plasma magnesium is relatively tightly regulated and does not reflect tissue levels well. The magnesium content of mononuclear cells and urinary levels are not very reliable markers and are not used in clinical settings. [131] Loading tests are more reliable, but also more tedious. Congenital primary hypomagnesemia is an uncommon disorder that is correlated to a specific defect in the intestinal absorption of magnesium. [128], [132] Hypomagnesuria, hypocalcemia, hypokalemia, and tetany often occur in association with convulsions, [133] which are corrected with the administration of magnesium. The hypocalcemia of magnesium deficiency is caused by an inhibition of the exchange between bone calcium and serum magnesium, resulting in decreased responsiveness of osteoclast parathyroid hormone receptors. [134] Magnesium deficiency eventually inhibits parathyroid hormone secretion and thus decreases the renal production of 1,25(OH) $_2$D $_3$, thereby decreasing the intestinal absorption of calcium. Vitamin D does not correct the hypocalcemia, whereas correction of magnesium levels restores calcium levels to normal. Experimentally induced symptomatic magnesium deficiency in human volunteers consistently resulted in very low levels of plasma magnesium (10%–30% of control values), low urinary and fecal magnesium levels, hypocalcemia, and hypokalemia. [131] Gastrointestinal problems (nausea, vomiting), arrhythmia, and personality changes were common. Most subjects had abnormal neuromuscular function after 4 to 14 weeks of consuming the low-magnesium (~10 mg/d) diet. All symptoms vanished when magnesium was given, with serum magnesium normalizing quickly and serum calcium and potassium more slowly. Clinical conditions that contribute to magnesium depletion include malabsorption syndromes, [135] such as celiac disease, inflammatory bowel disease, and other immune diseases involving villous atrophy; intestinal bypass or resection; gastrointestinal infections; and renal dysfunction, such as may be caused by metabolic disorders, tubular disease, and nephrotoxic drugs and diuretics, and which may result in excessive magnesium losses. [136] Several endocrine disorders also cause magnesium depletion, [137] including diabetes mellitus, hyperparathyroidism with hypercalcemia, and hyperthyroidism. Alcoholism often leads to poor magnesium status, [138] and protein-calorie malnutrition has also been reported to involve impaired magnesium nutrition. [139] Magnesium depletion often occurs in uncontrolled clinical conditions. However, changes in magnesium concentrations in tissues and serum often reflect secondary or tertiary causes of depletion. [131] Potassium depletion, protein catabolism, and chronic acidosis all lower

cellular magnesium levels and, consequently, levels of magnesium in muscle and bone. [131]

Iron

Sources, Forms, and Bioavailability The human diet contains two fundamentally different forms of iron: heme iron and nonheme iron. These forms are absorbed and metabolized differently, and dietary factors have different effects on their absorption. [140] Heme iron is present as myoglobin and hemoglobin in meat, poultry, and fish. The absorption of heme iron is higher than that of nonheme iron. In iron-replete individuals, the efficiency of heme iron absorption is usually about 20% to 25%, whereas the absorption of nonheme iron is about 3% to 10%, depending on the diet. Heme iron absorption is relatively unaffected by dietary factors, whereas food components may have widely different effects on the absorption of nonheme iron. The presence of globin and heme as components of hemoglobin and myoglobin in animal protein in the diet has been shown to increase heme iron absorption. Similarly, a low iron status may enhance heme iron absorption, but only to a relatively modest degree. Ascorbic acid has a strong enhancing effect on nonheme iron absorption, [141] whereas phytate, tannins, and other polyphenols are inhibitory. [142] Calcium has been shown to inhibit iron absorption in some short-term radioisotope studies, [143] but not in others. [144] Long-term studies of infants, children, and lactating and postmenopausal women given high daily doses of calcium (500–1000 mg) for 6 to 12 months, however, failed to show any effect on iron status as assessed by hemoglobin or serum ferritin levels. [145], [146] and [147] It is therefore possible that homeostatic mechanisms adapt after an initial inhibitory effect of calcium on iron uptake, and no negative effect on iron status occurs. The DRI for iron is 8 mg/d for men and women older than 50 years; it is 15 mg/d for young women ages 15 to 18 years and 18 mg/d for older premenstrual women. [148]

Absorption and Homeostasis Heme and nonheme iron are absorbed via different pathways and are affected to different degrees by dietary components, physiological conditions, and iron status. The mechanisms underlying heme iron absorption are not yet fully understood. The uptake into the intestinal mucosal cell has been suggested to occur via putative "heme receptors" that have not yet been characterized, [149] or by intercalation of the hydrophobic heme group via the lipid bilayer of the brush border membrane. [150] Once within the cell, heme oxygenase breaks down the heme group, releasing iron for incorporation into intracellular ferritin or for export out of the cell. [151] Hence, once heme is broken down, the released iron becomes part of the nonheme iron pool. Several mechanisms involved in the mucosal regulation of nonheme iron absorption have been discovered (Fig. 20-3). Duodenal cytochrome *b* (Dcytb) was discovered in the small intestine of rats and found to reduce ferric iron (Fe^{3+}) to the more absorbable ferrous (Fe^{2+}) form. [152] It is known that rodents can absorb ferric iron well, supporting a role for this ferrireductase in iron absorption. Humans, however, do not absorb ferric iron well, [140] although ferrireductase activity has been measured in human duodenal mucosa. [153] Therefore, despite its presence in human small intestine, Dcytb may have only a limited role in iron absorption.

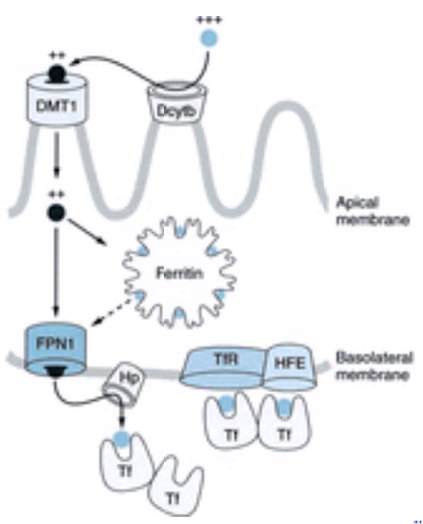

FIGURE 20-3. Suggested mechanisms for the regulation of iron absorption by the mucosal cell. *Dcytb*, duodenal cytochrome b; *DMT1*, divalent metal transporter 1; *FPN1*, ferroportin 1; *HFE*, hemochromatosis gene product; *Hp*, haephestin; *Tf*, transferrin; *TfR*, transferrin receptor.

Divalent metal transporter (DMT1; also called *DCT1* or *Nramp2*) is located in the brush border membrane and is involved in the regulation of iron uptake by the enterocyte. Its expression is up-regulated during iron deficiency and down-regulated in situations of iron excess. [154] The protein is primarily located in the villus tip cells, with less present in the crypt cells. The gene for this protein was found in mutant mice with microcytic anemia (mk), [155] and in the anemic Belgrade rat. [156] Originally, this protein was found by expression cloning in *Xenopus* oocytes, and several cations, including Fe^{2+}, Zn^{2+}, Cu^{2+}, Cd^{2+}, Co^{2+}, Mn^{2+}, Ni^{2+}, and Pb^{2+}, were found to be transported by DMT1. [157] However, it is not certain that oocytes with DMT1 perform the normal functions of a cell–that is, the functions that would occur in vivo. In another study, manganese was found to affect DMT1 expression in cultured Caco-2 cells, but zinc was far less effective. [154] Thus, under physiological conditions, this "divalent metal" transporter may transport only a few trace elements, while others are transported by other carriers. Inside the mucosal cell, the expression of ferritin is regulated by the intracellular concentration of iron. [158] Iron binds to iron regulatory proteins, which are sequence- and structure-specific RNA-binding proteins, which change conformation on binding iron. When the iron status is low, iron regulatory proteins a have high affinity for the iron-responsive element of ferritin mRNA and block protein synthesis. Thus, the rate of ferritin synthesis is low when the iron status is low. When iron is present in excess, the iron regulatory protein conformation does not allow binding to the iron-responsive element, and ferritin synthesis increases. In contrast to ferritin, transferrin receptor (TfR) mRNA contains five iron-responsive elements in the 3' UTR (untranslated region), [159] whereby its stability is regulated, and also a rapid turnover determinant. When the iron regulatory protein binds to the TfR mRNA, the interaction between ribonuclease and the rapid turnover determinant is hindered and TfR mRNA synthesis decreases. This coordinate regulation of ferritin and TfR synthesis, which occurs at the mRNA level, is an excellent means for the cell to respond to varying iron status. The TfR is located both in endosomal compartments and in the basolateral membrane, with the transferrin-binding structural feature being on the outside of the cell, allowing transferrin in portal blood to interact with the TfR. A novel protein capable of exporting iron out of the enterocyte was discovered simultaneously by three research groups and was called *ferroportin 1 (FPN1),* [160] *iron-regulated transporter 1 (IREG1),* [161] and *metal transporter protein 1 (MTP1),* [162] respectively. There is so far no consensus on which name should be used. This iron exporter is up-regulated during iron deficiency and down-regulated during iron excess, [161] and it is likely to be a key protein in normal mucosal iron homeostasis. A membrane-bound protein of 62 kd, it has ten transmembrane regions and contains iron-responsive elements. [162] Fe^{2+} must be oxidized to Fe^{3+} to become incorporated into transferrin in the portal vein. Hephaestin, a copper-dependent enzyme with activity similar to that of ceruloplasmin, was discovered by positional cloning in the sex-linked anemia (sla) mouse mutant, which has a block in intestinal iron transport. [163] This protein is likely to have some influence on human intestinal iron metabolism, but the human homolog has not yet been described, and its quantitative significance is unknown.

Disorders of Iron Homeostasis Hereditary hemochromatosis is an autosomal recessive genetic disorder of defective regulation of iron absorption that leads to severe iron overload in various tissues of the body, particularly the liver. [164] The gene frequency usually is about 1 in 300, but it varies with ethnicity and is highest among Northern Europeans. To prevent cirrhosis of the liver, patients are either phlebotomized regularly or given chelation therapy, usually with desferrioxamine. Studies of patients with hereditary hemochromatosis have revealed that the vast majority of these individuals (>95%) have a mutation in the HFE gene. [165] This gene is expressed at low levels in most tissues, including the small intestine, and appears to be involved in the appropriate down-regulation of iron absorption that normally occurs when iron stores are high. The HFE protein is similar to major histocompatibility complex (MHC) class I proteins. [166] A missense mutation (C282Y) affects a highly conserved cysteine residue, involving a disulfide bond within the MHC I proteins. This causes a defect in HFE protein trafficking and cell surface expression as a consequence of the inability of the defective HFE protein to bind ß $_1$-microglobulin. [167] HFE must interact with ß $_1$-microglobulin to regulate iron absorption, which is illustrated by the finding that iron overload develops in ß $_1$-microglobulin–deficient mice. [168] The HFE knock-out mouse shows all the signs of human hereditary hemochromatosis, which supports the critical role of this protein in the regulation of iron absorption. [169] HFE has been shown to bind directly to the transferrin receptor, [167] suggesting that the regulatory role of HFE may be mediated by an interaction with this protein. Sickle cell disease and ß-thalassemia are also genetic disorders that lead to abnormal iron metabolism. Whereas hereditary hemochromatosis is characterized by the preferential loading of iron into parenchymal cells, in sickle cell disease or ß-thalassemia, iron first accumulates in the reticuloendothelial cells. [170] Clinically, the primary cause of mortality in hereditary hemochromatosis is cirrhosis and hepatocellular carcinoma, whereas in ß-thalassemia, it is cardiomyopathy. [171] In comparison with patients who have ß-thalassemia, patients who have sickle cell disease are relatively spared the toxicity of iron overload. Whether these different outcomes are a consequence of differences in expression of the iron transporters in the intestine and liver has not yet been explored.

Deficiency and Excess Iron deficiency is the most common single nutrient deficiency worldwide, afflicting some 800 to 1000 million people in both less developed and industrialized countries. It is estimated that 500 to 600 million persons have iron deficiency anemia. In adults, this anemia results in impaired immune function and energy metabolism, producing lethargy. [172] In infants and young children, iron deficiency anemia also causes delays in cognitive function and motor development. [173] Whether these developmental delays are reversible is still not clear, but most studies suggest that they are irreversible. Prevention of iron deficiency is therefore a

high priority from a public health perspective. Iron deficiency usually is diagnosed by the serum ferritin concentration, which is directly proportional to body iron stores. When the body becomes depleted of iron, serum ferritin values decrease. A ferritin value below 12 µg/L usually is used as the cutoff for iron deficiency and has been shown to correlate with a lack of stainable iron in the bone marrow. If iron deficiency progresses, hematopoiesis becomes impaired, resulting in microcytic anemia. Because serum ferritin increases during infection and inflammation, resulting in a falsely high value, the circulating serum TfR level is now under evaluation as an additional test for iron deficiency. [174] When iron stores decrease, the cellular synthesis of TfR increases and is reflected in serum TfR concentrations. TfR is not affected by infection and may therefore better reflect tissue iron needs. [174] However, it is not yet known whether other factors affect the TfR, and furthermore, the several commercial kits yield very different results, so that interlaboratory comparisons are difficult. It may therefore take some time before this assay will become part of the clinical routine. Lack of dietary iron or poor iron bioavailability is by far the most common cause of iron deficiency, but chronic gastrointestinal blood loss, malabsorption secondary to mucosal disease (e.g., celiac disease), bypass of the proximal intestine, and achlorhydria can also lead to iron deficiency. Although iron deficiency is common in many populations, particularly children and fertile women, iron overload can also occur in a significant proportion of individuals. Besides hereditary hemochromatosis, iron-loading anemias, such as homozygous thalassemia and sideroblastic anemia, also cause iron overload. [175] Therefore, an overly zealous approach to providing iron to large population groups may harm a growing number of individuals with already large iron stores. Concern has also been raised that high iron stores, although not yet characterized as iron overload, can lead to increased free radical formation and chronic disease. It has been shown that Finnish men with elevated serum ferritin levels (>200 µg/L) have a significantly higher risk for coronary heart disease and non–insulin-dependent diabetes than men with normal (<200 µg/L) serum ferritin levels. [176], [177] Other studies yielded conflicting results, and a recent metaanalysis did not find a significantly increased odds ratio. [178] However, the likelihood of confounders in such epidemiologic studies is high, and further studies are needed to resolve this issue.

Zinc

Sources, Forms, and Bioavailability The daily dietary intake of zinc, which usually is about 10 to 15 mg, generally meets the requirement of men but is often marginal for women, particularly during pregnancy and lactation. [179] The DRI for men is 11 mg/d; for nonpregnant women, 8 mg/d; for young women ages 18 years and younger and for pregnant and lactating women, 13 and 14 mg/d, respectively; and for women ages 19 to 50 years and older than 50 years, 11 and 12 mg/d, respectively. [180] The best dietary sources of zinc are meat, meat products, and seafood. Whole grains and cereals also are relatively high in zinc, but bioavailability often is low because of the presence of phytate. [181] The use of zinc supplements has become more popular, particularly because of its perceived beneficial effect in the prevention and treatment of the common cold. Multivitamin/mineral tablets now often contain zinc, and some cereal products are fortified with zinc. Most forms of zinc are well absorbed, although zinc is less available from zinc oxide as a result of its low solubility. Phytates in cereals and legumes have a strong inhibitory effect on zinc absorption as a consequence of their chelating properties. [181] Because humans cannot digest phytate, zinc bound to phytate is unabsorbable and lost in the feces. Recent attempts to increase the bioavailability of zinc from human diets include alternative food processing (e.g., fermentation), phytase treatment, and the development of genetically modified cereals that are lower in phytate. [182], [183] and [184] All these approaches aim to lower the phytate content of the food. Calcium can lower zinc bioavailability, most likely by forming insoluble calcium-zinc-phytate complexes. However, under some circumstances, calcium can also increase zinc absorption. [185] The protein content of the diet is positively correlated to zinc absorption, most likely because of the formation of amino acids and small peptides that facilitate zinc uptake by the enterocyte. [186] Iron in high doses can reduce zinc absorption from oral supplements, [187] but this does not occur when both are given with a meal. [188] Thus, this interaction may be of some concern when multivitamin/mineral tablets are taken on an empty stomach, but not otherwise.

Absorption and Homeostasis The efficiency of zinc absorption from the diet usually is about 15% to 35% in adults. [189] Active transport dominates at low or normal intake, whereas passive diffusion contributes more significantly at high intake. [190] The extent of the homeostatic regulation of zinc in humans is not well known, but studies in experimental animals suggest that it occurs, although not to the same extent as for iron. [181] The mechanisms underlying the regulation of zinc absorption have long remained elusive. It was known that the low-molecular-weight (~6 kd) protein metallothionein (MT), which also binds copper and cadmium, [191] is present in the cytosol of the enterocyte. However, zinc induces MT only at very high intakes, so that this protein an unlikely regulator under physiological conditions. However, zinc-binding proteins likely play important roles in the trafficking of zinc across the mucosal cell. Zinc transporter 1 (ZnT-1) appears to be involved in the export of zinc across the enterocyte basolateral membrane, [192], [193] whereas ZnT-2 and ZnT-4 are involved in the flux of zinc in the endosomes, possibly regulating intracellular trafficking of zinc. These membrane transporters all have six transmembrane-spanning domains and a conserved histidine-rich region predicted to have a cytoplasmic loop that is likely to bind zinc. [192], [194] ZnT-1 was localized to the basolateral membrane, and ZnT-2 was found in acidic vesicles that accumulate zinc. [195] Overexpression of ZnT-1 supports its role in zinc export. [192] Although higher levels of ZnT-1 mRNA were found in zinc-supplemented rats, [193] direct functional evidence is still missing. ZnT-2 is more responsive to zinc than ZnT-1. Experiments showing zinc sequestration by endosomal vesicles during overexpression of ZnT-2 suggest that this transporter may be important for controlling intracellular transport of zinc by the enterocyte. [195] In rats, ZnT-2 mRNA levels closely correlate with zinc intake. ZnT-4 appears to colocalize with ZnT-2, suggesting some redundancy, but ZnT-4 mRNA is not responsive to changes in zinc status. All three transporters were found primarily in villus cells, much less frequently in crypt cells. The ileum was the major site for ZnT-1; ZnT-2 was found in rat duodenum and jejunum, and ZnT-4 in all parts of the small intestine. [193] Few studies to date have been conducted in intact animals, so that it is difficult to evaluate the physiological significance of these transporters in zinc absorption and homeostasis. Whether MT is involved in this regulation or plays more of a bystander role also needs to be evaluated further. MT expression was found to be closely correlated with ZnT-2 expression, and the ZnT-2 promoter may have multiple metal response elements, similar to the MT promoter. A superfamily of human zinc transporters has been found. [196], [197] Transfection of K562 cells with zinc importer protein (hZIP1 or hZIP2) increased the cellular uptake of zinc. Because these proteins are localized in the plasma membrane and because primarily zinc influx is affected, these transporters may also be responsible for zinc uptake by the enterocyte. Other transition metals decreased zinc uptake, suggesting that these proteins also may transport other cations. [196]

Deficiency and Excess Zinc is a cofactor for a wide diversity of enzymes and transcription factors. Severe zinc deficiency affects many metabolic pathways and tissues, impairing growth, development, reproduction, and immune function. [198] However, severe zinc deficiency is rare and usually is seen only in patients with acrodermatitis enteropathica; marginal zinc deficiency, in contrast, despite problematic status assessment, is likely to be common. [199] The diagnosis of milder forms of zinc deficiency is difficult in part because the plasma zinc level is affected by many factors other than zinc. [198] The response to zinc supplementation is therefore often used as an indicator of zinc status. Supplementation trials have shown positive effects on the growth of infants and children, [200] pregnancy outcome, [201] and diarrheal disease, [202] demonstrating that several population groups are vulnerable to subclinical zinc deficiency. This is particularly the case in developing countries, and strategies to prevent zinc deficiency worldwide are currently under way. The risk for zinc toxicity and the likelihood of achieving excessive dietary intakes of zinc are both low. [198] Patients at some risk for excessive zinc intake are those being treated with oral zinc supplements, such as for acrodermatitis enteropathica or Wilson disease (see section “ Copper”). The effects of such treatment do not appear to be acute; rather, the copper status is impaired with its long-term consequences.

Disorders of Zinc Homeostasis Acrodermatitis enteropathica is an inborn autosomal recessive disorder of zinc metabolism manifested by eczema, dermatitis, anorexia, compromised immune function, and poor growth. [203] The gene frequency is about 1 in 10,000. Symptoms usually occur when infants are weaned as a consequence of the low zinc intake and bioavailability from infant formula and weaning foods, combined with rapid growth. [204] Referral to a dermatologist is common, but the only effective treatment is zinc supplementation. Patients require daily doses of zinc, usually 30 to 50 mg, and higher doses during periods of rapid growth, such as at puberty. If doses are too high, however, copper deficiency can result, causing other clinical problems [205]; therefore, both the zinc status and the copper status of these patients must be monitored. The defective mechanism underlying acrodermatitis enteropathica is still not known. It is described as a disorder of zinc absorption; however, malabsorption may be a secondary problem resulting from mucosal damage caused by local zinc deficiency, as patients treated with zinc appear to absorb zinc normally. [205] It is likely instead that zinc efflux out of the enterocytes into the intestinal lumen is abnormally high, thereby causing an intracellular zinc deficiency. Whether the expression of any of the zinc transporters (ZnT-1, -2, -4; hZIP1, 2) is dysregulated in patients with acrodermatitis enteropathica has not yet been explored.

Copper

Sources and Bioavailability Copper is relatively abundant in meat, grains, and nuts; milk and dairy products are low in copper. [206] However, copper intakes are relatively close to the DRI (0.9 mg/d for men and women), [207] and some concern has been raised that copper status may be suboptimal in vulnerable groups. Environmental factors, such as soil, water source, fertilizer use, processing, and cooking, also affect the copper content of the diet. For example, tap water occasionally has high copper levels because of copper plumbing and well water sometimes has high copper levels, and certain groups, particularly infants and children, may have copper intakes that are excessive. [208] Thus, some concern regarding copper nutrition is warranted. Copper absorption from the diet usually is high, and few dietary components strongly affect its bioavailability. [209] High intakes of iron or zinc, however, may lower the copper status if the ratio between these elements and copper remains high for a long period. [210] Thus, multivitamin/mineral supplements that contain iron and zinc, but not copper, may negatively affect copper status. Ascorbic acid has been shown to lower copper absorption in animals by reducing well-absorbed divalent (cupric) to monovalent (cuprous) copper, which is not as well absorbed. [211] However, in human studies, young adult volunteers given high levels of ascorbic acid daily (600 mg) showed no change in copper status. [212] Whether even higher doses of ascorbic acid, such as those sometimes advocated for prevention of the common cold (>1000 mg/d), affect copper absorption and status is not yet known. Other organic acids in foods, such as lactic, citric, malic, and acetic acids, slightly enhance copper absorption, most likely by affecting its solubility by

chelation. [211]

Absorption and Homeostasis Copper absorption is mediated by both active transport and passive diffusion. [211], [213] Animal studies suggest that the duodenum is the major site of absorption, but some absorption also occurs in the stomach and ileum. [214] Human studies show that copper intake affects copper absorption. [215] Copper homeostasis is mediated in part by bile copper excretion; about 15% of the copper excreted in bile is reabsorbed through the enterohepatic circulation. [211] Cellular copper homeostasis is maintained by a delicate balance of copper uptake and efflux, which are mediated by highly conserved membrane proteins. [216] Copper-transporting P-type ATPases are involved in the intracellular trafficking of copper as well as in the export of copper out of the cell. [217] Cytoplasmic low-molecular-weight copper-binding proteins act as chaperones, steering copper ions to their incorporation into the proper apoproteins. [216] The expression of a human gene, *CTR1* (copper transporter 1), was shown to increase the cellular uptake of copper in yeast cells. [218] The involvement of Ctr1 in cellular copper metabolism was demonstrated by increased copper uptake by human fibroblasts transfected with the human *CTR1* gene. [219] The Ctr1 protein appears to be vital for normal copper metabolism because the *CTR1* knock-out mouse was embryolethal. [220] Mice heterozygous for *CTR1* exhibit tissue-specific defects in copper accumulation and in the activities of copper-dependent enzymes. Supplementation of the pregnant mice with copper did not rescue the homozygous knock-out embryos, suggesting that they cannot acquire copper because they lack the membrane-bound Ctr1 protein, and furthermore, there is no alternate transport system for copper. [220] Ceruloplasmin and hephaestin are copper-dependent enzymes involved in iron homeostasis (see previous section), and the heterozygous *CTR1* knock-out mice had decreased tissue levels of iron, most likely a consequence of impaired delivery of copper to its apoproteins. Interestingly, patients with the very rare genetic disorder aceruloplasminemia have normal copper metabolism but suffer from the consequences of impaired iron metabolism. [221] Studies of human intestinal Caco-2 cells in culture suggest two pathways of copper absorption. [222] At low levels, copper uptake and efflux are closely coupled, and little copper is stored in the cell. Conversely, at high levels, copper uptake and efflux are buffered by a large storage compartment. The molecular regulation of these events is not yet known but should be explored in light of the newly discovered transporters.

Disorders of Copper Homeostasis Menkes disease is an X-linked recessive disorder of copper absorption; copper deficiency occurs at a very young age. [223] The incidence of Menkes disease is 1 in 50,000 to 1 in 100,000 live births. Symptoms include pallor, lax skin, bone fractures, osteoporosis, altered pigmentation of skin and hair (secondary to low levels of melanin formation), growth failure, mental retardation, and microcytic anemia (secondary to low levels of ceruloplasmin/haephestin activity). Oral treatment with copper is ineffective, and patients often die at a young age. The gene was identified by work on mouse models of Menkes disease, [224] and the defective protein is one of the P-type ATPases, ATP7A or Menkes (MNK) protein, which is involved in cellular copper metabolism, particularly the export of copper out of the cell. [217], [225] Thus, copper is blocked in the enterocyte, and little copper is transported into the systemic circulation, resulting in severe copper deficiency. In Wilson disease, excessive amounts of copper accumulate in the body, particularly in liver and brain, and clinical symptoms include liver cirrhosis, eye lesions (Kayser-Fleisher ring), kidney malfunction, and neurological problems. [226], [227] Despite very high levels of copper in the liver, serum levels of copper and ceruloplasmin are low. Treatment has classically been chelation therapy with drugs such as penicillamine and trientine, but high oral doses of zinc (40–50 mg/d) are also effective because they inhibit copper absorption. [228] Wilson disease is an autosomal recessive disorder, occurring in 1 in 30,000 live births. This disorder of copper metabolism has also been shown to be caused by a defective transporter, in this case ATP7B or the WD protein. [229] Copper absorption per se does not appear to be dysregulated in these patients; rather, tissue copper metabolism, particularly in the liver, is affected, causing an excessive cellular accumulation of copper. The outcome for these patients, if treated, usually is good, although continuous monitoring of copper, zinc, and iron status is necessary.

Deficiency and Excess The significance of human copper deficiency was recognized by clinical studies of infants recovering from malnutrition. [230] Risks for copper deficiency include low iron stores in the liver of premature infants, a rapid growth rate, malabsorption syndromes, and increased copper losses, [231] but the deficiency usually is not precipitated unless the dietary intake of copper also is low. [208], [230] Low serum levels of copper and anemia, which is refractory to iron because of low ferroxidase/haephestin activities, are manifestations of copper deficiency and are accompanied by neutropenia, bone abnormalities, hypopigmentation of hair and skin, impaired growth, and an increased incidence of infections. [232] Decreased phagocytic capacity of neutrophils and impaired cell immunity affect the immune system. [233] Acute copper toxicity is rare and usually is caused by the consumption of contaminated foods or beverages, or by the accidental or deliberate ingestion of large quantities of copper salts. [234] Symptoms include nausea, vomiting, and diarrhea. Chronic toxicity is also rare but seems to occur in geographic clusters. Indian childhood cirrhosis has been reported in families that were consuming milk boiled or stored in brass or copper containers. [235] Children consuming such milk may ingest up to 1 mg of copper per kilogram per day, which is enough to explain the observed liver damage. Infants and children in the Austrian Tyrol were reported to have died of liver cirrhosis secondary to chronically high levels of copper intake. [236] In these cases, inheritance followed the typical pattern of a mendelian recessive trait, suggesting that these individuals were particularly sensitive to copper exposure. This was supported by the observation that many children who had received similar levels of copper had no liver damage. Sporadic cases have been reported in other areas, and some of these cases have occurred in consanguineous marriages. Cases were much more frequent in boys, and a genetic origin is possible. Whether a genetic disorder of copper metabolism is present in patients with liver cirrhosis is not known, but the possibility should be explored in light of the new findings of copper transporters in humans.

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CHAPTER 21

David H. Alpers and Samuel Klein

GENERAL NUTRITIONAL PRINCIPLES

BASIC NUTRITIONAL PRINCIPLES

Body Composition

The human body consists of 35 components that are organized into five levels of increasing complexity: atomic (e.g., nitrogen, potassium), molecular (e.g., water, protein), cellular (e.g., body cell mass, intra- and extracellular fluid), tissue (e.g., skeletal muscle, adipose tissue), and whole body (e.g., weight, height). A healthy, lean man is composed of 55% to 60% water, 15% to 20% fat, 15% to 20% protein (one half in skeletal muscle), 1% glycogen (four fifths in muscle, one fifth in liver), and 4% minerals. [1] Although sophisticated techniques are available to measure each body component, the definitions of some commonly used terms can be confusing. *Fat mass* represents all body triglycerides, which are present in adipose tissue, muscle, and liver. *Adipose tissue* is about 83% fat (e.g., triglyceride), 15% water, and 2% protein. *Fat-free mass* refers to total body mass minus total fat mass. *Lean body mass* is defined as total body mass minus adipose tissue. The body also can be divided into cellular and extracellular mass. *Body cell mass* is defined as the cellular components of all tissues (35%–45% of the body weight in normal men, 30%–40% in women) and can be measured by total exchangeable potassium. [2] *Extracellular mass* is defined as the heterogeneous group of tissues and fluids supporting the body cell mass.

Diet for Normal People

Many guidelines have been developed over the years for general use by the U.S. population for health maintenance and disease prevention. Although these have been published at different times and represent the input of a large number of experts with diverse interests, all expert panels have reported remarkably simple and consistent recommendations for healthy adults. Two of these guidelines are widely disseminated and are current. *Nutrition and Your Health: Dietary Guidelines for Americans, 2000* combines the guidelines of the U.S. Department of Agriculture and the U.S. Department of Health and Human Services. [3] This publication offers ten guidelines for a healthy diet (Table 21-1).

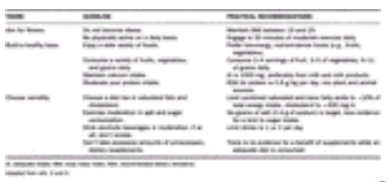

TABLE 21-1 Dietary Guidelines for Adult Americans, 2000

The *American Heart Association Dietary Guidelines Revision 2000* sets forth the recommendations of the American Heart Association. [4] This report recommends four goals for the population that fit well with the guidelines from the U.S. Department of Agriculture (see Table 21-1). The Healthy Eating Pattern goal suggests consuming a variety of fruits, vegetables, and grain products. The Healthy Body Weight goal suggests matching energy intake with energy needs and limiting the consumption of foods with a high caloric density. The Desirable Blood Cholesterol and Lipoprotein Profile goal limits intake of foods high in saturated fatty acids and cholesterol, substituting grains and sources of unsaturated fatty acids, such as vegetables, fish, legumes, and nuts. The Desirable Blood Pressure goal recommends limiting dietary salt intake and alcohol consumption, reinforcing the need for maintaining healthy body weight. Metaanalyses of randomized trials of the effect of reducing sodium on blood pressure concluded that a direct relationship exists for hypertensive persons. [5]

Vegetarian diets accomplish many of the recommendations because these diets lower the risk for the development of coronary artery disease, type II diabetes, and hypertension. [6] The recommendations listed in Table 22-1 are generalized for all Americans, but special considerations may be needed for African Americans and other minority groups. For example, the diets of middle-aged African Americans appear to be lower in calcium, iron (for women), folate, and zinc. [7] The condition of being overweight is more prevalent in African American women and in Hispanic Americans. Americans from the Asian and Pacific Rim countries tend to have diets lower in dairy products and calcium, although in second- and third-generation families, these differences are likely to disappear.

The approach to macronutrient intake of all the dietary recommendations is similar. [3], [4], [8], [9] and [10] The report on macronutrients of the Standing Committee on the Scientific Evaluation of Dietary Reference Intakes, Food and Nutrition Board, Institute of Medicine has been available since 2002. Until that time, the ecommendations for energy and protein intake were supplied by the 1989 tenth edition of *Recommended Dietary Allowances (RDAs)*. [10] All sources recommend maintaining a healthy weight. Standards for such weight have varied from those of the more restrictive *1979 Body Build Study* [11]—which does not allow for age-related weight changes and relates desirable body weight to a low mortality rate—to other measures of ideal weight, such as the RDAs, [10] the measures of the Fogarty Center Conference on Obesity, [12] and the average weights in the United States, which allow for slight increases with age. The guidelines suggest balancing energy intake and physical activity to maintain appropriate weight, and avoiding diets that are excessive or severely restricted in kilocalories. It is worthwhile to calculate the estimated resting energy expenditure (REE) and the activity-related energy requirement. REE is best estimated from the World Health Organization equation, but the Harris-Benedict equation has the advantage of greater simplicity, even though it is less accurate at the extremes of weight (Table 21-2). For estimates of overall energy use, the RDA figures are simple and useful (Table 21-3). These estimates of energy expenditure vary in both directions by 20%. Also, snacks and alcohol intake must be added to the estimated dietary intake.

Harris-Benedict Equation[33]	
Men = 66 + (13.7 × W) + (5 × H) − (6.8 × A)	
Women = 665 + (9.6 × W) + (1.8 × H) − (4.7 × A)	
Owen et al.[37]	
Men = 879 + (10.2 × W)	
Women = 795 + (7.18 × W)	
World Health Organization[48]	

AGE (y)	MALE	FEMALE
0–3	(60.9 × W) − 54	(61.0 × W) − 51
3–10	(22.7 × W) − 495	(22.5 × W) + 499
10–18	(17.5 × W) + 651	(12.2 × W) + 746
18–30	(15.3 × W) + 679	(14.7 × W) + 996
30–60	(11.2 × W) + 879	(8.7 × W) + 829
>60	(13.5 × W) + 987	(10.5 × W) + 596

W, weight (kg); H, height (cm); A, age (y).

TABLE 21-2 Formulas for Estimating Resting Energy Expenditure

LEVEL OF ACTIVITY	ACTIVITY FACTOR (× REE)	AVERAGE ENERGY EXPENDITURE* (kcal/kg per day)
Very light		
Men	1.3	31
Women	1.3	30
Light		
Men	1.6	38
Women	1.5	35
Moderate		
Men	1.7	41
Women	1.6	37
Heavy		
Men	2.1	50
Women	1.9	44

*Estimated from World Health Organization equations for median weights of persons ages 19 to 74 years; activity factor 1.0 = 24.0 kcal/kg for males, 23.2 kcal/kg for females.

REE, resting energy expenditure.

Adapted from ref. 10.

TABLE 21-3 Calculation of Energy Requirement

Regarding protein intake, the available guidelines recommend a moderate intake, not to exceed two times the RDA and not to provide more than 20% of total calories. There is no evidence that intake above the RDA (0.8 g/kg per day) is beneficial. Moreover, because most protein sources are rich in fat, a high protein intake would increase fat intake. All the guidelines recommend a diet with an abundance and variety of vegetables (3–5 servings) and fruits (2–4 servings), plenty of grains (6–11 servings), and sugar in moderation. They recommend the increased intake of complex carbohydrates and fiber as found in whole grain products, fruits, and vegetables. Such intake would provide 55% or more of the total daily energy intake. A corollary to the recommendations on carbohydrates is that the diet be low in fat, especially saturated fat, and cholesterol, to reduce the risk for atherosclerotic heart disease. In addition to a reduced total fat intake, the diet should have no more than 30% of calories from fat, no more than 10% of calories from saturated fat and *trans* fatty acids, and no more than 300 mg of cholesterol per day. The intake of polyunsaturated fats should be increased, up to 10% of total calories, because they have a cholesterol-lowering effect and are a rich source of vitamin E. This recommendation reflects the now large data sets that show favorable blood lipoprotein patterns in individuals who consume moderate amounts of total fats that are predominantly polyunsaturated. [3], [13] Finally, most guidelines suggest an Adequate Intake (AI) of calcium (1300 mg/d for adolescents, 1000 mg/d for adult men and nonpregnant women ages 19 through 50, and 1200 mg/d for adults older than 50 years). [13] Milk contains the highest concentration of calcium (280–300 mg/8-oz cup). There is as yet no convincing evidence that additional protective effects to prevent osteopenia are conferred by ingesting calcium over the recommended AI. Salt (NaCl) should be limited to 6 g/d or less, and alcohol to 1 oz of pure alcohol per day (1 oz is the equivalent of two cans of beer, two small glasses of wine, or two average cocktails).

The implementation of the published recommendations depends on two other federally supported guidelines. The RDA [10], [13], [14], [15] and [16] is the level judged to be high enough to ensure an adequate intake for the majority of the normal population, not the minimal intake necessary to avoid negative balance. The values for the RDA vary with age and gender. These values should not be confused with the U.S. RDAs, standards set by the U.S. Food and Drug Administration in 1973 for the purposes of food labeling for the entire population. The term *U.S. RDA* has been replaced by *Reference Daily Intake (RDI)* and *Daily Reference Value (DRV)*. The latter provided for total and saturated fat, cholesterol, total carbohydrates, dietary fiber, sodium, potassium, and protein—nutrients for which no U.S. RDA was available. The RDI and DRV are now the basis for the daily values that appear on all food labels, and reflect DRVs and RDIs for a 2000-calorie reference diet. The new recommended dietary intake levels are called *Dietary Reference Intakes (DRIs),* a collective term that includes the Estimated Average Requirement (EAR), the Recommended Dietary Allowance (RDA), the Adequate Intake (AI), and the Tolerable Upper Intake Level (UL) (Table 21-4). [13], [14], [15] and [16] These have been developed by the Institute of Medicine in the United States along with Health Canada. The EAR is the daily nutrient intake value estimated to meet the requirements of half of an age- and gender-specific group. It often is derived from balance studies and is used in setting the RDA, the intake level sufficient to meet the nutrient needs of most people within a given life-stage and gender group. The RDA also includes a factor to account for variation within the group, usually estimated at 10% (i.e., coefficient of variation, or CV). If not enough data are available to calculate an EAR, AI is used, based on observed intake consistent with good health. The UL is the maximum amount of a nutrient that can be ingested without posing a health threat and was included because so many nutrients are now ingested at levels far exceeding those possible from the diet.

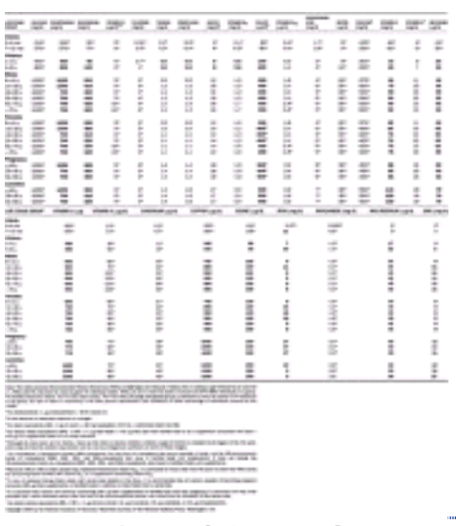

TABLE 21-4 Dietary Reference Intakes: Recommended Intakes for Individuals

These guidelines account for group and in some cases individual variability, but they are not meant for use in patients with either acute or chronic diseases. Rather, they should be used as baseline estimates for planning individual nutrient intake. Such adjustments are particularly important in diseases characterized by malabsorption or catabolism, in contrast to simple lack of intake. Confirmation of sufficient nutrient provision can then be monitored by tests specific for each macro- or micronutrient (see Chapter 53).

The food guide pyramid, recommended to implement the dietary guidelines in Table 21-1, has replaced the four basic food groups by dividing fruits and vegetables, to which are added breads (with cereal, rice, and pasta), milk products, and meat (with poultry, fish, dry beans, eggs, and nuts) to make five groups (Fig. 21-1). [17] The concept assumes that meals are built on a basis of grains, fruits, and vegetables, and supplemented with low-fat milk products and other protein sources in the meat group. The food pyramid comes with a warning to use fats and sweets sparingly. To limit fat and cholesterol intake, it is important to remember that ground beef, the single largest source of dietary fat in the United States, still contains more than half of its calories as fat after cooking; a single egg contains more than 200 mg of cholesterol, and 70% of the calories in regular cheese are from fat.

FIGURE 21-1. U.S. Department of Agriculture food guide pyramid. (From ref. [18].)

The practical aspects of providing advice using the food pyramid (or any other educational system) involves a knowledge of the number of servings in the diet, and what counts as one serving. The range of servings provided by the food pyramid are spread over the average range of energy intake, from 2200 to 2800 kcal/d (Table 21-5). The sizes of the servings are smaller than most people realize and do not include the addition of fats or oils in the calculation of daily energy intake. For example, one bread serving would be one slice of bread or 1/2 cup of cooked rice or pasta; one serving of vegetables is 1/2 cup of chopped, raw, or cooked vegetables; one piece of fruit or 3/4 cup of juice is one serving; 1 cup of milk or 11/2 to 2 oz of cheese is one serving; and 21/2 to 3 oz of cooked lean meat, poultry, or fish or 1/2 cup of cooked dry beans is one serving. [18]

TABLE 21-5 How Many Servings Are Needed Each Day?

Chemoprevention of Gastrointestinal Cancers One of the special applications of dietary recommendations for healthy people is to prevent gastrointestinal cancers. The general recommendations for such diets are similar to those that support health in the entire population. However, it has been estimated that about one third of all cancers are related to diet, and that most colorectal cancer in the United States might be prevented by dietary alterations. [19] Although epidemiologic data have suggested associations between overall diets or environment and the risk for cancer incidence or mortality, it has proved difficult to identify the dietary components that might influence such risks, and to demonstrate their benefit in a prospective fashion. [20], [21] and [22] Table 21-6 summarizes many of the data associating risks for cancer with dietary components. Very few of the associations reported have been convincing. When dietary components have been identified and tested prospectively, the data, in general, are negative, even when premalignant end points are examined, such as colorectal polyps. [23]

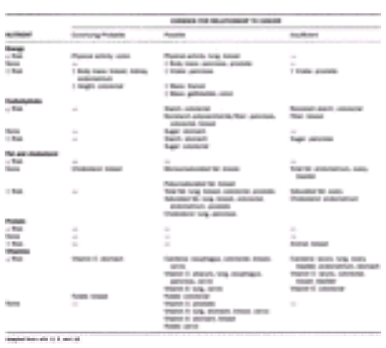

TABLE 21-6 Nutrition, Food, and Cancer Prevention

Epidemiologic evidence (case control and cohort studies) also suggests that people with cancer have a lower intake of raw, fresh, leafy green, or cruciferous vegetables, as well as raw or fresh fruits, especially citrus fruits. [24] Although the ingestion of such foods has increased in the United States, the average intake is less than 0.7 servings of vegetables or fruits per day. Moreover, a large cohort study (Iowa Women's Health Study of more than 40,000 women) showed no protective effect of vegetables or fruits on colorectal carcinoma incidence, except for garlic. [23] The antioxidant vitamins A, C, and E are among those compounds thought to be responsible for the possible effects of vegetables and fruits on carcinogenesis. The same large cohort study analyzed the effect of these three vitamins on colorectal cancer incidence and found that vitamins A and C had no effect. [23] An association of total (i.e., supplemental and dietary) and supplemental vitamin E with a decreased incidence was found, but dietary vitamin E intake alone was not correlated. Other studies have tested the addition of individual or combinations of vitamins (or ß-carotene) on the incidence of colorectal adenoma formation but have not found much efficacy. [19] The other dietary component that has been most extensively studied is fiber. Two extensive reviews have examined the descriptive and case control studies of the association of dietary fiber with colorectal cancer. [25], [26] Most studies showed some correlation, suggesting a protective effect of fiber. In the Health Professionals Follow-up Study involving men, a 64% reduction in cancer was noted in those with the highest quintile of fiber intake (>28.3 g/d) compared to those with the lowest intake (<16.6 g/d). [27] However, results from two large studies of women, the Nurses Health Study [28] and the Iowa Women's Study, [29] showed no effect. Prospective studies suggest that an effect of fiber may occur only in men. A number of interventional studies using fiber supplements have been performed, but the results are inconclusive. [19], [25], [26] The data regarding the effect of other dietary components on the incidence of colorectal cancer or adenomas are too fragmentary or incomplete to permit strong recommendations. [19] Some evidence suggests an effect of folate, and its role is being tested in interventional studies. Alcohol (>30 g/d) may increase the incidence of adenomas in the distal colon and rectum. Although many case control and cohort studies suggested that calcium protected against the development of colorectal cancer, other studies with larger cohorts showed no effect. The results of interventional studies are conflicting, and the end point used has been colonic cell proliferation, not adenoma or carcinoma incidence. In summary, some data implicate an increased intake of dietary fat and red meat and a decreased intake of vegetables, fruits, and fiber in colorectal carcinogenesis. The interventional recommendations include altering those components, which is precisely the recommendations for the diets to maintain the health of people in the United States. [3], [4] Because most people eat less vegetables, fruits, fiber, and calcium than what is needed to meet the RDAs, it is reasonable to promote these diets for general health.

Energy Metabolism

The human body continuously consumes energy for the maintenance of ionic and osmotic gradients, cell transport, nerve conduction, intermediary metabolism, biosynthesis, heat generation, and the performance of involuntary and voluntary mechanical work. Energy is provided largely by the mitochondrial production of high-energy phosphate bonds generated by the oxidation of fat, carbohydrate, and protein. After the hydrolysis of carbohydrates to simple sugars, fats to fatty acids and glycerol, and proteins to amino acids, most of these small molecules are converted to the acetyl unit of acetyl-coenzyme A (CoA), generating a small amount of ATP in the process. Acetyl-CoA is a common breakdown product of the three macronutrients. Acetyl-CoA, carrying most of the chemical energy of the original macronutrients, enters the citric acid cycle and undergoes oxidative phosphorylation, which are the final common pathways in the oxidation of food molecules (Fig. 21-2). Many amino acids enter the citric acid cycle as a-ketoglutarate or oxaloacetate rather than as acetyl-CoA.

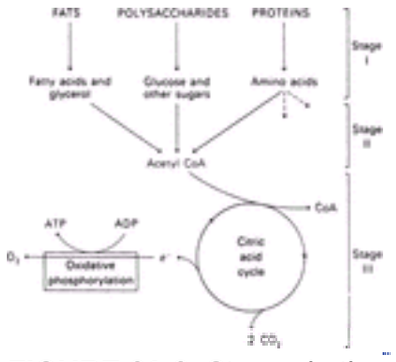

FIGURE 21-2. Stages in the extraction of energy from foodstuffs. (From Stryer L. Biochemistry, 3rd ed. New York: WH Freeman, 1988:325.)

A portion of the energy released during substrate oxidation is not used to perform work and is dissipated as heat. Therefore, energy production is traditionally measured in terms of heat production. One kilocalorie (kcal), equal to 4.184 kilojoules (kJ), is the amount of heat required to raise the temperature of 1 kg of water by 1°C. Normally, body temperature is carefully maintained within narrow limits so that heat production equals heat loss. Energy production can be determined directly (direct calorimetry) by measuring the transfer of heat from the body to water circulating in specially designed chambers or suits. Energy production also can be measured indirectly (indirect calorimetry) by measuring carbon dioxide (CO_2) production and oxygen (O_2) consumption, because the amount of heat produced during substrate oxidation is proportional to the amount of CO_2 produced and O_2 consumed. [30] The relationship between CO_2 production and O_2 consumption can be used to estimate the relative oxidation of different substrates. [31]

Dietary carbohydrates, fats, and proteins can be used as fuel soon after their ingestion, or they can be stored by the body for subsequent oxidation. Endogenous energy stores (Table 21-7), which are continuously being mobilized and oxidized, become a critical source of fuel during postabsorptive conditions and when energy intake is inadequate to meet energy demands. The largest source of endogenous energy is triglyceride in adipose tissue, which is uniquely designed to store fuel. Triglycerides have a high energy density and release 9.4 kcal/g when oxidized. Adipose tissue is composed almost entirely of triglycerides in an oil form, which constitutes 85% of adipocyte weight. In comparison, glycogen, the other major source of endogenous fuel, generates only 4.1 kcal/g on oxidation. Glycogen takes up a considerable amount of space because it is stored in liver and muscle tissue as a gel containing 2 to 4 g of water for every gram of glycogen. [32] The mobilization of adipose tissue yields 6 to 8 kcal/g, whereas the mobilization of glycogen yields only 1 to 2 kcal/g. The energy stored in the adipose tissue of a lean man can provide enough fuel for him to survive 2 months of total energy restriction, [33] whereas the energy present as liver glycogen is consumed within 24 hours of fasting. Certain cells and tissues, such as the brain, prefer glucose as a fuel, and others, such as bone marrow, erythrocytes, leukocytes, renal medulla, eye tissues, and peripheral nerve tissue, require glucose because they cannot oxidize fatty acids. None of the macronutrients are completely absorbed; some are excreted in the feces. Based on the average digestibility of fat (95%) and carbohydrate (97%), the digestible energy derived from fat is 9.0 kcal/g, and that from carbohydrate is 4.0 kcal/g.

TISSUE	FUEL	ENERGY (kcal)
Adipose tissue	Triglyceride	140,000
Muscle	Glycogen	2000
	Triglyceride	3000
Liver	Glycogen	300
	Triglyceride	500

TABLE 21-7 Body Energy Stores

Components of Energy Expenditure Total energy requirements include the sum of REE, the thermic effect of physical activity (TEPA), the thermic effect of feeding (TEF), and adaptive thermogenesis (AT). REE is the energy consumed while lying quietly awake in the postabsorptive state. Normally, REE accounts for about 70% of total daily energy expenditure. Energy requirements of different tissues and organs are heterogeneous, however. Energy consumption by the body's most metabolically active organs—the brain, liver, kidney, and heart—accounts for 60% of REE; these constitute only 5% of total body mass (Table 21-8). In contrast, adipose tissue, which accounts for about 20% and 30% of body weight in lean men and women, respectively, consumes less than 5% of REE.

	MASS		ENERGY EXPENDITURE	
TISSUE	g	% Total	kcal/d	% Total
Gut	2000	3	300	13
Brain	1400	2	400	18
Liver	1600	2.2	440	19
Heart	300	0.4	235	10
Kidneys	300	0.5	200	10
Adipose tissue	14,000	20	70	4
Skeletal muscle	28,000	40	400	18

TABLE 21-8 Postabsorptive Energy Requirements

REE is related to body weight across mammalian species; REE is proportional to the three-fourths power of body weight (weight $^{0.75}$). [34] Several equations have been used to estimate resting energy requirements in humans based on measurements of REE in normal subjects (see Table 21-2). [35], [36], [37] and [38] These equations generate values that are usually within 10% of measured values in normal healthy volunteers but are less accurate in persons who are at extremes of weight (i.e., very lean or obese) or who are ill. Starvation and severe hypocaloric feeding decreases the resting metabolic rate to values 15% to 20% below that expected for actual body size, whereas illness and injury can increase energy requirements. Physical activity usually accounts for 15% to 20% of total energy expenditure. The precise contribution of TEPA to total energy expenditure depends on the intensity and duration of activities. At rest, skeletal muscle accounts for 20% of total energy requirements. However, during moderate- to high-intensity aerobic exercise, energy consumed by working muscles can increase more than 50-fold, causing a 15-fold increase in total energy expenditure. The TEF represents the energy costs of digestion, absorption, transport, metabolism, and storage of nutrients, and it also may involve AT. Eating or infusing nutrients increases the metabolic rate by about 5% to 10% of the ingested or infused calories and depends on the specific foods consumed. Normally, 12% to 20% of the energy in ingested protein, 6% to 12% of carbohydrate energy, and 2% to 3% of fat energy is expended. AT is a proposed mechanism for wasting excess energy to maintain a constant body weight despite fluctuating amounts of energy intake, or for maintaining body heat during exposure to different environmental temperatures. [39] It has been proposed that energy expenditure and heat production for AT and TEF involve brown adipose tissue, a specialized, highly vascularized, thermogenic tissue innervated by sympathetic nerves. [40] Brown adipose tissue is packed with large mitochondria possessing an uncoupling protein that uncouples ATP synthesis from respiration. [41], [42] In this situation, the rate of substrate oxidization does not depend on the availability of ADP precursor, and the reaction can continue at high rates, permitting even small quantities of brown adipose tissue to increase heat production markedly. Radioimmunoassays for the uncoupling protein have demonstrated the presence of brown adipose tissue in humans of all ages [42]; other studies suggest that uncoupling protein expression also can be induced in white adipose tissue. Although it has been estimated that as little as 50 g of brown adipose tissue can contribute 10% to 15% of the energy turnover in humans, [41] the physiological importance of AT in energy metabolism in humans is not clear.

Proteins

Proteins are composed of amino acids joined together by peptide bonds. Twenty different amino acids are commonly found in human proteins. Differences in the sequences of amino acids in proteins permit diverse structures and functions; proteins serve as enzymes, carriers, receptors, hormones, and structural elements. The amino acid sequence determines the location of sites for covalent attachment of carbohydrate and ultimately determines the protein's three-dimensional configuration and specific function. Nitrogen is also present in the body in the form of free amino acids. Free amino acids are in a dynamic state with those being incorporated into tissue proteins, those undergoing catabolic reactions, and those used for the synthesis of other nitrogen-containing compounds. [43]

Protein Quality Protein quality is related to its ability to support metabolic homeostasis and growth, which is determined by amino acid content and bioavailability. Some amino acids (histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophan, valine, and possibly arginine) are considered essential because their carbon skeletons cannot be synthesized by the body. These amino acids must be consumed in the diet for normal function and survival. Other amino acids (glycine, alanine, serine, cysteine, cystine, tyrosine, glutamine, glutamic acid, asparagine, and aspartic acid) are nonessential because their carbon skeletons can be produced endogenously. In general, the greater the ratio of essential to nonessential amino acids, the better the quality of protein. The ability to digest protein and absorb its component amino acids also affects protein quality. True absorption ranges from 97% to 99% for proteins in meat, milk, and eggs, to 75% for proteins

in potatoes and navy beans. [44], [45] Protein bioavailability also can be affected by food preparation. For example, some lysine is lost by heating in the presence of reducing sugars. In contrast, heating increases the bioavailability of soy protein by inactivating the trypsin inhibitor present in soybeans. [46] In general, the proteins of egg, milk, fish, red meat, and poultry are high in biologic value, and the protein in wheat gluten is low. [44], [45], [47]

NITROGEN BALANCE Nitrogen balance is the difference between intake and output. Nitrogen is excreted primarily in the urine as urea, creatinine, porphyrins, ammonia, and uric acid. The relative proportions of these compounds can vary, but urea usually accounts for about 80% of urinary nitrogen. During fasting conditions, urinary nitrogen reaches a low level of about 2 mg/kcal of REE or about 40 mg/kg of body weight. Approximately 1 to 3 g of nitrogen is normally lost per day from fecal and other sources. [16] Fecal nitrogen losses reflect unabsorbed protein in the diet and in intestinal secretions and sloughed epithelial cells. The amount of endogenous protein that normally enters the intestinal lumen is about 50 g/d. Absorption of exogenous and endogenous protein is so efficient that fecal nitrogen is normally only 1 to 2 g/d. Minor amounts of nitrogen are lost through intact skin, nasal secretions, semen, menstrual fluid, and hair cuttings. Nitrogen balance can be used to estimate protein balance because about 16% of protein consists of nitrogen, and it is assumed that almost all body nitrogen is incorporated into protein or amino acids. A positive balance (i.e., intake greater than losses) represents a net increase in total body protein, whereas a negative balance (i.e., losses greater than intake) demonstrates net protein catabolism. One gram of nitrogen represents about 6.25 g of protein, which is equivalent to 30 g of hydrated lean body mass. Nitrogen balance is affected by protein intake and quality, energy intake, and nutritional status. Inadequate energy intake increases protein requirements. When protein intake is suboptimal, nitrogen balance can be improved by increasing energy intake. Therefore, nitrogen balance reflects both protein intake and energy balance. Most normal adults can maintain nitrogen equilibrium by ingesting 0.5 g of high-quality protein per kilogram of body weight per day. The range of recommended intake for adults is 0.5 to 0.8 g/kg per day, [10], [43], [48], [49] which provides a margin of safety to allow for decreased biologic availability and increased requirements in subsets of the population. The average protein in a Western diet has only 75% of the biologic value of egg protein. [43], [50] Intravenously administered amino acids are as effective in promoting nitrogen balance as oral protein. [51] Infancy is a time of intense growth, and the protein requirements per unit of body weight for infants are higher than those for adults. The normal infant also requires a higher proportion of essential to nonessential amino acids. [52] The growth spurt of adolescence, the only extrauterine period during which growth velocity increases, occurs between 10 and 13 years of age for American girls and between 12 and 15 years of age for American boys and contributes about 15% of final adult height and 50% of adult weight. [53] The guidelines for protein and calorie needs in infancy, childhood, and adolescence are summarized in Table 21-9. The nutritional demands of a normal pregnancy average 80,000 kcal (i.e., 300 kcal/d) and 950 g of protein (i.e., 3.5 g/d). The U.S. National Research Council RDA for pro-tein during pregnancy is 60 g/d, representing an increase of 10 g/d over the RDA for nonpregnant women 25 years of age or older. [10] For lactating women older than 25 years of age whose average daily output of milk is 850 mL, a protein intake of 65 g/d in the first 6 months and 62 g/d in the second 6 months is recommended by the National Research Council. [10] Slightly lower values are recommended for younger women. [10] Modified versions of these figures are available in the Dietacy Reference Intakes for Energy, Carbohydrates, Fiber, Fat, Protein and Amino Acids (macronutrients), 2002 (see legend to Table 21-9).

TABLE 21-9 Dietary Reference Intake Values for Energy and Protein in Individuals by Life Stage Group

Protein Metabolism Body proteins exist in a state of constant flux, with protein synthesis and breakdown occurring simultaneously. Normal daily protein turnover is 1% to 2% of total body protein and results largely from the degradation of muscle and hepatic proteins. Protein degradation involves the enzymatic hydrolysis of protein to its constituent amino acids. More than 75% of the amino acids released by protein breakdown are reused for the synthesis of new proteins; the remaining amino acids are oxidized. Proteases within cell lysosomes are responsible for most protein degradation. [54], [55], [56], [57] and [58] However, proteases are also found in plasma membranes and in the cytosol. The carbon skeletons of amino acids can be oxidized for energy or used for synthesis of glucose, ketone bodies, or fatty acids. [2] Nitrogen can be released as ammonia into the bloodstream and delivered to the liver, where it is converted to urea. The metabolism of amino acids involves the transfer of nitrogen between organs from the periphery to the liver (Fig. 21-3). The liver is a workhorse for amino acid metabolism and is the site of synthesis for urea and plasma proteins. It is the main site of catabolism for the essential amino acids, with the exception of the branched-chain amino acids leucine, isoleucine, and valine, which are degraded in muscle and kidney.

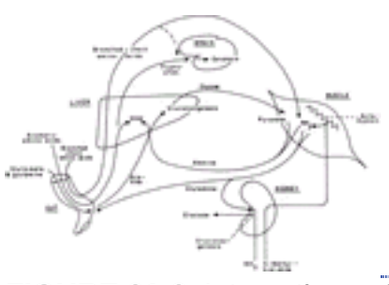

FIGURE 21-3. Interactions of organs in the metabolism of some major amino acids. (From Munro HN. Interactions of the liver and muscle in the regulation of metabolism in response to nutritional and other factors. In: Arias IM, Popper H, Schachter D, et al., eds. The liver: biology and pathobiology. New York: Raven Press, 1982:677.)

Skeletal muscle preferentially takes up the branched-chain amino acids after each meal and is the primary site of metabolism for these amino acids. Although leucine, isoleucine, and valine constitute only 8% of dietary amino acids, they make up 60% of the amino acids in the systemic circulation. [59], [60] When muscle proteins are catabolized, the branched-chain amino acids undergo transamination, yielding alanine, glutamine, and branched-chain keto acids. The keto acids are used by the muscle as fuel, and alanine and glutamine are exported and taken up predominantly by the liver and intestine, respectively. [59] These two amino acids account for more than 50% of the total amino acid nitrogen released from muscle. [60], [61] and [62] The kidneys also take up glutamine, which is the major substrate for renal ammonia production. [63]

Lipids

Lipids are a heterogeneous group of compounds that are soluble in organic solvents. Lipids include triglycerides (fat), sterols, glucolipids, phospholipids, and fat-soluble vitamins. These compounds serve as a source of energy, structural components of cell membranes, carriers of essential nutrients, and precursors for steroid hormone, prostaglandin, thromboxane, and leukotriene synthesis. Dietary lipids are composed mainly of triglycerides, which contain mostly saturated and unsaturated long-chain fatty acids with a 16- to 18-carbon chain length.

Lipid Metabolism The use of fat as a fuel requires the hydrolysis of triglyceride to free fatty acid and glycerol and the tissue uptake of free fatty acids for subsequent oxidation. Hormone-sensitive lipase within adipocytes hydrolyzes adipose tissue triglycerides and releases free fatty acids into the bloodstream, where they are bound to plasma proteins and delivered to other tissues. Lipoprotein lipase at the lumenal surface of the capillary endothelium hydrolyzes plasma triglycerides and releases free fatty acids for local tissue uptake. Fatty acids are transported across the cell membrane by passive diffusion, facilitated diffusion, and active transport. Membrane and cytosolic fatty acid binding proteins are important in transporting fatty acids across the cell membrane and in directing fatty acids from the cell membrane to different metabolic sites. This intracellular fatty acid transport system enhances fatty acid uptake by maintaining a fatty acid concentration gradient and prevents potentially toxic interactions between fatty acids and intracellular organelles. Long-chain fatty acids are delivered across the outer and inner mitochondrial membranes by a carnitine-dependent transport system. Inside the mitochondria, fatty acids are degraded by ß-oxidation to acetyl-CoA, which enters the tricarboxylic acid cycle (see Fig. 21-3). Ketone bodies are produced solely by the liver and are generated by the partial oxidation of fatty acids. Ketone body production increases when the rate of fatty acid production is much greater than the rate of fatty acid oxidation, such as during starvation or uncontrolled diabetes mellitus. In these conditions, ketone bodies become an important fuel and are released into the bloodstream for delivery to extrahepatic tissues. Ketone bodies represent a water-soluble fuel derived from water-insoluble fatty acids. Ketone bodies can cross the blood-brain barrier to replace glucose as the major fuel for the brain, sparing plasma glucose for consumption by other tissues. [64] The biosynthesis of fatty acids is mediated by fatty acid synthase, a multienzyme complex embodied in a single polypeptide chain. It

elongates the molecule by sequential addition of two carbon units and stops with the formation of palmitic acid, a 16-carbon fatty acid. The formation of malonyl-CoA from acetyl-CoA is the committed step in fatty acid biosynthesis and the most important step of regulation. The enzyme that catalyzes this step, acetyl-CoA carboxylase, is stimulated by citrate. Citrate is abundant when ATP and acetyl-CoA are abundant, a condition appropriate for fat synthesis. Palmitoyl-CoA, the end product of fatty acid synthesis, antagonizes the activation of acetyl-CoA carboxylase by citrate.

Essential Fatty Acids Most fatty acids can be synthesized by the liver, but humans lack the desaturase enzyme needed to produce the n-3 double bond (between carbons 3 and 4 counted from the methyl end) and the n-6 double bond (between carbons 6 and 7) in the fatty acid series. Essential fatty acids are important constituents of cell membranes and precursors of the eicosanoids. [65] Arachidonic acid (C20:4, n-6), a precursor of eicosanoids, prostaglandins, leukotrienes, prostacyclins, and thromboxanes, is synthesized from linoleic acid. [65], [66] Vegetable oils, such as corn, soybean, sunflower, peanut, and cottonseed oils, are rich sources of linoleic acid. [65], [66] Linoleic acid (C18:2, n-6) should constitute at least 2% and linolenic acid (C18:3, n-6, 9, 12) at least 0.5% of the daily energy intake to prevent the occurrence of essential fatty acid deficiency, usually manifested as a specific alteration in the plasma fatty acid profile and a skin rash. An elevated ratio of triene to tetraene (>0.4) is characteristic of essential fatty acid deficiency as a result of increased production of eicosatrienoic acid, a fatty acid containing three double bonds (i.e., triene) derived from oleic acid (C18:1), as well as from decreased arachidonic acid production, a tetraene derived from linoleic acid elongation. [67], [68] Essential fatty acid deficiency is rare in adult humans because of sufficient essential fatty acids stores in adipose tissue. However, continuous infusion of lipid-free total parenteral nutrition (TPN) can cause abnormalities of the triene-to-tetraene ratio within 10 days because of increased plasma insulin concentrations, which inhibit lipolysis and the release of essential fatty acids. [68]

Fish Oil Fish oils are ?-3 polyunsaturated fatty acids (PUFAs) found in marine animals, particularly fatty fish, such as herring, salmon, bluefish, and tuna. [69], [70] Epidemiologic studies suggest a possible protective effect of fish oils against cardiovascular disease. [71], [72] In sufficient doses, fish oil prolongs the bleeding time and decreases the production of the proaggregating substance thromboxane A_2. [73], [74], [75] and [76] The significant hypotriglyceridemic effects of fish oils have been confirmed repeatedly in healthy persons and in those with various hyperlipidemic states, but the long-term effects may be less striking. [75], [76], [77], [78], [79], [80], [81] and [82] The effects on serum cholesterol and low-density lipoprotein (LDL) levels have varied. [69], [82], [83] Animal models suggest that fish oils have an inhibitory effect on coronary atherosclerosis and intimal hyperplasia. [69], [73], [74] The ?-3 fatty acids generally suppress cellular inflammatory responses by changing the end products of eicosanoid synthesis. [69], [80], [84], [85] and [86] Dietary supplementation with fish oil suppresses the production by monocytes of the polypeptide cytokines interleukin-1 (IL-1) and tumor necrosis factor (TNF), suggesting an additional mechanism by which fish oils may exert an antiinflammatory effect. [84], [87]

Carbohydrates

Carbohydrates, which constitute most of the earth's organic matter, are important sources of metabolic fuel. In the United States, carbohydrates normally account for about 50% of ingested calories; approximately 60% is complex carbohydrate, primarily starch, and most of the remainder is sucrose and lactose. [88] About 10 to 20 g of indigestible carbohydrate (i.e., soluble and insoluble fibers) are consumed daily. They all undergo hydrolysis to yield glucose and other simple sugars. Some cells and tissues, such as erythrocytes, leukocytes, renal medulla, eye tissues, and peripheral nerve tissue, do not have the capacity for citric acid cycle activity and require glucose as a fuel for anaerobic glycolysis. The brain prefers glucose as a fuel. Daily glucose requirements include 40 g/d for anaerobic tissues and 140 g/d for the brain. [89]

Absorbed glucose that is not directly oxidized can be stored as energy in the form of glycogen or fat, which requires approximately 5% and 25%, respectively, of the original substrate oxidative energy potential. Glycogen is a branching, long-chain polymer of glucose molecules with water and electrolytes between the chains. It is found in most tissues but is stored significantly only in the liver and skeletal muscle. The primary function of hepatic glycogen, about 100 g in a normal adult, is to maintain blood glucose levels. Plasma glucose is an essential fuel for glucose-dependent tissues. Glycogen in skeletal muscle serves to supply glucose to the muscle itself during physical activity.

Glycolysis The conversion of glucose to pyruvate in the cytosol of cells is known as *glycolysis,* a process that results in the generation of ATP but does not require oxygen. Pyruvate represents a major metabolic junction; it can be reduced to lactate, transaminated to form alanine, or enter the mitochondria and undergo carboxylation to oxaloacetate or oxidative decarboxylation to acetyl-CoA (Fig. 21-4).

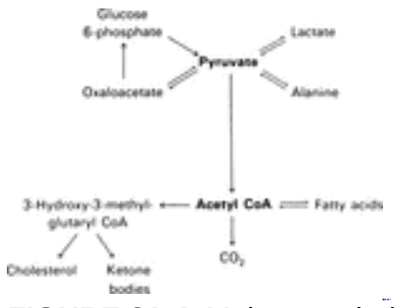

FIGURE 21-4. Major metabolic end products of pyruvate and acetyl-coenzyme A in mammals. (From Stryer L. Biochemistry, 3rd ed. New York: WH Freeman, 1988:633.)

Citric Acid Cycle and Oxidative Phosphorylation The citric acid cycle (i.e., tricarboxylic acid cycle, Krebs cycle) represents a series of reactions that occur in mitochondria. Carbohydrates, lipids, and amino acids enter the cycle after being metabolized to acetyl-CoA and are completely oxidized to CO_2 and water. Vital biosynthetic intermediates are produced by the cycle, and it plays a major role in gluconeogenesis, lipogenesis, and amino acid transamination and deamination. As acetyl-CoA is oxidized, reduced nicotinamide adenine dinucleotide (NADH) and reduced flavin adenine dinucleotide ($FADH_2$) are formed, which transfer the electrons to the respiratory chain in the inner mitochondrial membrane. In the mitochondria, the transfer of high-energy electrons from NADH or $FADH_2$ down the electron transport chain is coupled to the generation of ATP, a process known as *oxidative phosphorylation*. Glycolysis (i.e., anaerobic respiration) yields a net of only two ATPs per molecule of glucose, whereas aerobic metabolism (i.e., citric acid cycle and oxidative phosphorylation) yields 36 ATPs for each molecule of glucose oxidized.

Glucose Production Hepatic glycogenolysis is responsible for most of the glucose produced endogenously in the fed and postabsorptive states. Other mechanisms for glucose production are active and become critically important when hepatic glycogen is depleted, such as during prolonged starvation and endurance exercise. Gluconeogenesis is the process by which glucose is synthesized from noncarbohydrate precursors, lactate, glycerol, and most amino acids (principally alanine). Gluconeogenesis occurs primarily in the liver, but the kidneys also produce glucose, especially during prolonged fasting. The Cori and glucose-alanine cycles provide mechanisms for generating plasma glucose for glucose-dependent tissues from 3-carbon intermediates released from peripheral tissues. [90] The Cori cycle (or lactic acid cycle) resynthesizes glucose that has been partially metabolized to lactate by peripheral tissues. Lactate produced principally by muscle, erythrocytes, and adipose tissue is transported to the liver and kidneys, where it is converted to glucose (gluconeogenesis) and released into the bloodstream. The glucose-alanine cycle shuttles glucose from the liver to muscle and alanine from muscle to liver. In this cycle, pyruvate is transaminated to alanine, which is transported to the liver and converted to glucose, which is then returned to muscle through the bloodstream.

Fiber

The accepted definition of dietary fiber is plant cell wall components that resist digestion by enzymes of the small intestine, including both polysaccharides and noncarbodydrate components. The polysaccharide compounds in dietary fiber, which are the structural and matrix components of plant cell walls, consist primarily of cellulose, hemicelluloses, and pectins, [91] as well as fructooligosaccharides and resistant starches. Cellulose is a ß1-4 polymer of glucose and the main structural component of plant cell walls. Hemicellulose consists of branched polymers of pentose and hexose sugars. Other noncellulose polysaccharides include pectins, which are complex mixtures of colloidal polysaccharides, and several polysaccharides not associated with the cell wall, including mucilages and gums. These compounds are branched polymers containing many uronic acids that hold water and form gels. They are highly branched in growing plants and become less branched as the support structure develops. They act as adhesives and are insoluble in the unripe fruit, becoming soluble only as the fruit matures. Undigested oligosaccharides, such as those associated with flatus (e.g., stachyose and raffinose), are soluble and not included in the definition of fiber.

Fructooligosaccharides are mixtures of ß-D-fructose monomers linked by ß2-1 linkages. These molecules include inulin-type fructans (linear polymers) and levans (branched fructans) and are present in many edible plants, such as wheat grains and members of the onion family. The daily consumption of oligofructoses by North American populations is estimated at 1 to 12 g, slightly more in Western Europe. [92] The ß-C2 linkage makes these polymers resistant to hydrolysis by human digestive enzymes, and they are fermented and metabolized in the colon to short-chain fatty acids. This colonic fermentation produces a change in the microflora, enhancing

bifidobacteria and decreasing *Bacteroides* organisms, clostridia, and other anaerobes. Fructooligosaccharides are the best studied of the prebiotics, defined as "a nondigestible food ingredient that beneficially affects the host by selectively stimulating the growth or the activity of one or a limited number of bacteria in the colon and thus improves host health." [93]

Resistant starches (RSs) are defined as starches that enter the colon. RS1 is physically inaccessible starch because of the particle size or entrapment in food. RS2 and RS3 are resistant to amylase action because of the compact (unbranched) structure, by being either unbranched (RS2) or made retrograde (RS3)—that is, altered during processing. [94] Most resistant starches are produced during food preparation. Intake of such starches on a Western diet is estimated at 5 to 10 g/d.

Other components of dietary fiber include polyphenols (especially flavonoids) and other cell wall–associated nonpolysaccharide substances. Polyphenols are products of plant metabolism and range from single-ring phenols to highly polymerized compounds, such as tannins and lignins. Only lignins are currently included in the determination of dietary fiber, although they constitute about 12% of plant organic compounds. Lignins include a group of phenylpropane polymers of varying sizes, as they are continuously polymerized as the plant ages. They reinforce the cellulose support structure and inhibit microbial cell wall digestion. Lignins are thus resistant to all anaerobic digestion systems and are not partially metabolized in the colon, as are the cell wall polysaccharides. They represent only a small part of the human diet (~0.2%).

Phenolic acids and aldehydes, such as vanillin, are common, but the most common of the plant phenolics are flavonoids, consisting of two aromatic rings linked through three carbons that form an oxygenated heterocyclic ring. [95] Flavonoids and other polyphenols are ubiquitous in plants and beverages. They are found in tea and contribute to the bitterness of that and other beverages. [14] Polyphenols usually account for less than 1% of the dry matter of plants, but they can reach concentrations of 4000 to 7000 mg/mL in red wines and fruit juices. [95] The dietary intake of polyphenols in the United States is 1 to 1.1 g/d, with flavonoids accounting for about 4% of the total. Like other fiber components, polyphenols are degraded and their metabolites are absorbed in the colon, but the effect of these compounds on short-chain fatty acid production and microflora is dependent on the type of compound and the microorganisms present. Polyphenols bind proteins and precipitate them in the intestinal lumen, and they can decrease the absorption of nitrogen, fat, and some minerals, including iron.

Interest in polyphenols has generally focused on their antioxidant properties, particularly on carcinogens and LDL oxidation. Some evidence suggests that moderate consumption of tea, a rich source of flavonoids, may protect against several forms of cancer, cardiovascular diseases, and kidney stone formation. [96] The blacker the tea, the more the polyphenols have been oxidized, lowering the possible effective role of these compounds. Herbal teas are not true teas (*Camellia sinensis*) and have a much lower flavonoid content. Tea contributes more than 60% of dietary flavonoids, onions about 13%, and grapes, apples, red wine, and dairy products most of the rest. The consumption of one to two cups of tea a day has been associated with health benefits in epidemiologic studies, [96] including decreased mortality from stroke in men (50%) and from cancer of the mouth, pancreas, colon, esophagus, skin, lung, prostate, and bladder (20%–40%). These data show only associations, not causation, and the results must be confirmed by prospective intervention studies of the type that have failed to show an effect of total dietary fiber.

Phenolics can, under some conditions, act as pro-oxidants. [97] Their antioxidant properties are dependent on solubility and chelating potential, among other properties. Thus, it is not possible yet to recommend the consumption of large amounts of phenolics as foods or supplements until more data are available.

The heterogeneity of dietary fiber has inspired numerous classification schemes, including those based on source, chemistry, structure, water solubility, detergent solubility, physiochemical properties, and physiological actions. *Crude fiber* was a term commonly used until the early 1970s. It refers to the residue of plant material that remains when food is extracted by dilute acids and alkalis. Although crude fiber is the measurement still referred to in most food tables, it underestimates by 80% to 90% the amount of material in foods that is undigestible by human digestive enzymes. [98]

The physiological effects of dietary fibers on gastrointestinal function are complex because of their heterogeneity and the changes in the lumenal environment along the gastrointestinal tract. Combinations of fibers may have effects that differ from those of individual purified preparations, and the same purified fiber can have different effects depending on how finely or coarsely it is ground. The quantitative measurements of the fiber content of foods alone does not always allow prediction of their biologic action.

Many physical properties of dietary fiber are physiologically important, including hydratability, viscosity, ion exchange properties, and adsorptive capacity. *Hydratability* relates to the ability of a fiber to form viscous gels. It is a function of the physical and chemical composition of the fiber, including particle size, the age of the plant, and the chemical properties of the surrounding solvent. [99] Some dietary fibers, such as lignins and pectins, have a significant capacity to bind and exchange ions, particularly calcium, iron, magnesium, zinc, and phosphorus, and to adsorb materials such as bile salts, proteins, and bacterial cells. [99]

Fermentation of fiber by colonic bacteria generates volatile short-chain fatty acids, acetate, propionate, and butyrate, which serve as a systemic fuel and as the preferred energy substrate of colonocytes. [100] In general, water-insoluble fibers (e.g., wheat bran, bagasse) are less subject to fermentation and hold more water than do the water-soluble fibers (e.g., vegetable fiber, pectins, gums). Therefore, water-insoluble fibers have a greater effect on stool mass than water-soluble fibers. However, the ingestion of degradable fiber stimulates bacteria growth and generates a fecal mass composed largely of bacteria. Although the total number of bacteria can be affected by diet, there is no convincing evidence that dietary changes produce major changes in the composition of colonic microflora. [101]

The rate of gastric emptying and the rate of digestion and absorption are influenced by fiber components. Guar gum and pectins increase the viscosity of the chyme and slow gastric emptying, but particulate fibers (e.g., wheat bran) appear to promote more rapid gastric emptying. [98] Fiber can decrease or increase mouth-to-anus transit time depending on fiber type, particle size, and bulk-forming capacity. [102] Intestinal transit time and stool bulk are inversely related. A large particle size (e.g., coarse wheat bran) produces a greater increase in stool bulk and a greater decrease in transit time than does a small particle size (e.g., finely ground bran). The mechanism by which fiber decreases colonic transit time is unknown but may be related to an increase in colonic peristalsis secondary to increased fecal mass. [102]

Epidemiologic observations, made initially in Africa, led to the hypothesis that dietary fiber and increased stool output can decrease the risk for Western gastrointestinal diseases, such as colon cancer, diverticulosis, cholelithiasis, appendicitis, constipation, and hemorrhoids. [103] However, data from epidemiologic studies may be influenced by confounding variables; a cause-and-effect relationship between dietary fiber and disease prevention has not been proved. [99], [25], [26], [104] Dietary fiber may prevent colon carcinoma by several mechanisms: a decrease in colonic transit time, so that the time that colonic mucosa is exposed to carcinogens is shortened; adsorption of carcinogenic sterols or other carcinogens; dilution of potential carcinogens by increasing stool volume; and alteration of the relative number of anaerobic and aerobic bacteria in the colon. Fiber may prevent diverticula by increasing lumenal bulk, which decreases intralumenal pressures by maintaining a larger-diameter lumen and eliminating closed segments during contractions.

Several studies have suggested that wheat bran promotes the formation of a less lithogenic bile by expanding the pool of bile salts, increasing the amount of chenodeoxycholic acid in the bile salt pool and decreasing the levels of deoxycholic acid in the pool. Other studies suggest that only specific dietary fibers—pectins, but not lignins or oat bran—are capable of lowering the lithogenic index in healthy persons. [104]

The treatment of irritable bowel syndrome with a high-fiber diet (especially a diet that includes wheat bran and commercial fiber supplements) has produced conflicting results. [105] It has been suggested that those patients whose major symptom is constipation are the ones most likely to benefit from increased fiber intake. Several studies have shown that bran and other fiber supplements are effective in preventing constipation, but the side effects include flatulence, distention, and bloating, perhaps related to a long colonic residence time and bacterial fermentation. [106]

Fiber may prevent and treat hemorrhoids by decreasing straining during defecation. Straining causes engorgement of the vascular cushion lining the distal rectum and anal canal, making them more vulnerable to shearing stress. The passage of hard fecal masses through the anal canal exacerbates these shearing forces and displaces the vascular cushion caudally, where it may be trapped temporarily by contraction of the anal sphincter. [107] A trial of fiber is a reasonable initial therapeutic approach for many patients who have hemorrhoids.

Adverse Effects of Fiber Consumption A high-fiber diet is generally safe and well tolerated. Bloating and flatulence are the most common side effects. More serious adverse events, such as obstruction of the esophagus and small intestine and volvulus in the small bowel and sigmoid colon have been reported. [104] Increasing dietary fiber intake does not cause long-term mineral and trace element depletion in populations consuming adequate diets. [98], [103] The effects of fiber on minerals and trace elements are of greatest concern in populations whose diets may be unbalanced in the relative proportion of fiber and micronutrients. For example, dwarfism observed in Iranian children was found to be caused by zinc deficiency induced by eating an unleavened bread, which is high in fiber but also in phytate, which binds

zinc. [104]

Micronutrients: Minerals and Vitamins

The RDAs and AIs are based on the amount of a nutrient needed for an individual to avoid deficiency, or on the average daily amount that populations must consume to prevent deficiency (see Table 21-4). Statistically, the RDA is set as two standard deviations above the mean requirement, so that 97% of healthy persons are covered. The RDAs thus exceed the needs of many healthy persons. They are established only for healthy persons. RDAs were never intended as guidelines for therapy. For some nutrients (e.g., sodium, chloride, potassium), the evidence for daily requirements is much lower than the content of the average diet in the United States. The Food and Nutrition Board has provided an estimated minimal requirement for these nutrients (Table 21-10). New guidelines for these electrolytes will be published in the near future and are available through the National Academy Press (e.g., see ref. [16]). RDIs and DRVs were established in 1973 for the purposes of food labeling and are derived from the 1989 RDAs for micronutrients. [18] What appears on food labels are neither DRVs nor RDIs but daily values, reflecting the recommendations for a 2000-calorie reference diet.

AGE	WEIGHT kg	WEIGHT lb	SODIUM (mg)	CHLORIDE (mg)	POTASSIUM (mg)
Up to 5 mo	4.5	10	120	180	500
6–11 mo	8.9	20	200	300	700
1 y	11	24	225	350	1000
2–5 y	16	35	300	500	1400
6–9 y	25	55	400	600	1600
10–18 y	50	110	500	750	2000
Older than 18 y	70	154	500*	750	2000*

* The minimum requirement does not allow for prolonged losses by vomiting, diarrhea, or excessive dieting. The Food and Nutrition Board of the National Academy of Sciences has recommended a daily sodium intake of 6 g or less, or a sodium intake of 2.4 g, 4.8 times the minimum requirement. The National Research Council recommends a high intake of potassium-containing fruits and vegetables, even though the average U.S. adult diet contains 3.5 of g potassium, nearly twice the minimum requirement. This recommendation takes into account the supposed benefits of increased dietary potassium in hypertension and of dietary fiber in preventing colonic malignancy.

Adapted from ref. 10. New recommendations are expected to be published in 1990.

TABLE 21-10 Estimated Minimum Requirements for Healthy Persons for Sodium, Chloride, and Potassium

Minerals

Sodium. Sodium, the principal cation in extracellular fluid, is necessary for maintenance of intravascular fluid volume and membrane potentials. [18], [108], [109], [110] and [111] Total body sodium ranges from 48 to 60 mEq/g and is dependent on body size. A 70-kg man has about 83 to 97 g of sodium in his body, about one fourth of which is in the skeleton and cannot be exchanged. The kidney regulates sodium excretion by aldosterone action in the distal tubule in response to intravascular volume. Obligatory sodium losses are small compared with body stores. Minimal fecal and urinary losses per day are about 23 mg (1 mEq). There is no RDA for sodium at this writing, but the estimated minimal requirement for adults is 500 mg (22 mEq) daily. Because almost all sodium is absorbed, renal regulation of excessive sodium absorption is crucial. Sodium is abundant in foods, not only as the chloride salt but as the bicarbonate, glutamate, phosphate, caseinate, benzoate, nitrate, propionate, sulfate, and citrate salts, among others. It is also present in many condiments, such as catsup, meat tenderizer, prepared mustards, olives, pickles, sauces, butter, margarine, and salad dressings. Water softeners can increase the water content of sodium. Medications may contain sodium, although only a few contain enough to cause a problem. Deficiency from inadequate intake alone is rarely encountered. Increased losses from the gut (e.g., vomiting, diarrhea, or drainage) or kidney (e.g., diuresis, salt-wasting renal disease, adrenal insufficiency) or excessive perspiration are the usual causes. Approximate mean concentrations of sodium in various fluids (in millimoles per liter) are sweat, 30 to 70; saliva, 10 to 20; gastric juice, 70; bile, 145; pancreatic juice, 130; jejunal secretion, 115; ileal secretion, 100; and normal stool, 5.

Potassium. Potassium is the primary cation in intracellular fluid, in which its concentration is 140 to 160 mEq/L. [11], [109], [110], [111] and [112] The 2% of total body potassium present in the extracellular fluid is important in influencing resting membrane function, particularly in the cardiac muscle. The kidney is the major site of potassium excretion, which is normally regulated not by filtered load but by the action of aldosterone and systemic pH in the distal tubule. It is absorbed efficiently in the upper intestine but secreted in the colon, another aldosterone-sensitive tissue. Colonic secretion is aided by the electronegativity of the lumen. As with sodium, the amount lost in the stool is volume dependent. Because excretion is so highly regulated by the kidney, no RDA has been established at this writing, but the estimated minimum daily requirement for adults is 2000 mg (51 mEq), and most adults consume 50 to 150 mEq daily. Abundant food sources (i.e., >200 mg per portion) include meats and fish, vegetables (especially potatoes), nuts, fruits, and milk. Most salt substitutes use potassium chloride to replace sodium chloride and contain about 2000 mg of potassium per teaspoon.

Calcium. Calcium is the most abundant cation in the body. About 99% resides in bone; the other 1% is a crucial mediator for neural transmission, myocardial function, excitation and contraction of muscle, coagulation, cell division, maintenance of intercellular tight junctions, and enzyme function. [13], [113], [114], [115], [116] /SUP>and [117] Many factors affect the intestinal absorption of calcium: lumenal pH, lumenal binders, transit time, the presence in the diet of the few foods that are rich in calcium, and vitamin D status. [13], [116] The process is relatively inefficient in that only about 33% of the daily calcium requirement is absorbed by the intestine. There is an obligatory loss of calcium each day from the intestine and kidney. During periods of growth or new bone formation, the calcium requirement increases. The risk for a negative calcium balance is great, especially during childhood, adolescence, pregnancy, or lactation. The new RDA for adults is 1000 mg/d (see Table 21-4), but typical diets in the United States provide only about 750 mg/d. [13] Unlike many of the other new RDAs, these recommendations are meant not as nutrient requirements for individuals, but as reflections of national policy. Thus, they describe optimal intake for populations. The debate continues regarding the need for calcium guidelines based on the relationship between calcium intake and bone health. [118] The RDA was raised for adolescents to 1300 mg/d, based on increased bone growth during this life stage. The increased recommendation in older persons to 1200 mg/d is based on decreased calcium absorption in that group. Milk and dairy products are the richest source of calcium (providing about 60% of dietary calcium), and it is most bioavailable in those sources in the form of calcium citrate. Each 8-oz cup of milk contains about 280 to 300 mg of calcium. Green leafy vegetables are a good source, but the bioavailability is more irregular than that in milk products because the calcium is present as the phytate, oxalate, or other organic anion salt. These salts are poorly ionized and absorbed. The single universal requirement for calcium intake at each life stage without reference to intake of protein, sodium, or phytate has been challenged, based on the observation that calcium intakes are low in parts of the world where fracture rates are low, but protein intake also is low, whereas phytate intake is high. [119] Other rich dietary sources are fish with edible bones, such as sardines and salmon. Lumenal calcium is most actively transported in the duodenum, but in humans, most calcium is absorbed in the ileum. [116] About 150 to 300 mg is secreted into the lumen each day. [111] After glomerular filtration, about 98% of calcium is reabsorbed by the renal tubules. [117] Half of serum calcium is protein bound, 10% is complexed with anions, and 40% is ionized and physiologically active. [114] Normal levels of serum calcium are tightly regulated by the action of parathyroid hormone and vitamin D and are not related to total body stores, except when deficiency is severe. Bone density is not a sensitive indicator of calcium deficiency, but it is a measure of body stores and can detect a decrease in bone mass of more than 1% to 2%. [120] Ionized serum calcium is a measure of the hormonally regulated calcium concentration. Under most circumstances, ionized serum calcium reflects body stores, but when acutely altered, it may be low in the presence of normal total body calcium. [121]

Magnesium. Magnesium is the second most abundant intracellular cation. About 70% of the total is in bone, and the rest is in soft tissues. Less than 1% is in extracellular fluids, where 20% to 30% is bound to protein. [108] Serum magnesium levels do not closely mimic body stores. The magnesium concentration in cells is high, like that of potassium, but magnesium leaves the cells less readily. Magnesium is important for neuromuscular transmission, and it is an essential cofactor in many enzyme reactions, including oxidative phosphorylation and nucleic acid synthesis. It also plays a role in wound healing, myocardial contractility, membrane stability, and coagulation. [122], [123], [124] and [125] The RDA averages about 350 mg (29 mEq) for adults 400 to 420 mg for men and 310 to 320 mg for women. The typical adult diet in the United States provides 20 to 40 mEq/d. Food sources are well distributed, but nuts, cereals, seafood, meats, legumes, and green vegetables are rich food sources. [122], [123] Like that of calcium, the intestinal absorption of magnesium is relatively inefficient (i.e., 30%–40%), and most absorption occurs in the ileum. However, magnesium differs from calcium in that there is very little obligatory intestinal loss, so magnesium balance is maintained even with a very low dietary intake if no abnormal losses occur. Loss from the body occurs in regulated function, mostly through the kidney (2%–3% of the filtered load). [122], [123], [124] and [125] Serum magnesium is the standard for assessing body stores, but it is falsely elevated by hemolysis and does not always reflect either intracellular stores or active extracellular ionized magnesium. [18], [121]

Phosphorus. Phosphorus is the major intracellular anion (100 mmol/L). [125], [126] and [127] It is essential for normal membrane function, regulation of enzyme systems, and generation and storage of energy. It affects the delivery of oxygen to tissue by regulating the concentration of 2,3-diphosphoglycerate in red blood cells. However, 80% to 85% of total body phosphorus resides in bone. The requirements for phosphorus parallel those of calcium. The new RDA is only 700 mg/d for adults, somewhat below the DRI for calcium, although daily intakes in the United States range from 1000 to 1500 mg. Like magnesium, urinary phosphorus excretion does not reflect a low dietary intake, so balance studies are misleading. Moreover, the efficiency of phosphorus absorption varies with the food source and the dietary calcium-to-phosphorus ratio. In general, if protein intake is adequate, so is phosphorus intake. [13] Phosphorus is a constituent of all cells and is abundant in most foods, especially meats, dairy products, and carbonated beverages. The absorption of phosphorus requires hydrolysis of the organic phosphates in food by intestinal alkaline phosphatase and is promoted by the action of 1,25-dihydroxyvitamin D_3. Net absorption is 60% to 80% of ingested phosphorus. The kidney adjusts phosphate excretion over a wide range, so deficiency from low intake or malabsorption is rare. In fact, reabsorption can increase to 99.8% if the dietary intake of phosphorus is low. [125]

Iron. Unlike the those of the minerals previously discussed, the body stores of iron are not regulated by increased or decreased excretion, but rather by control of the rate of intestinal absorption, which is increased during deficiency. Daily losses normally occur from the gastrointestinal tract, skin, and urine. Fecal losses predominate, ranging from 6 to 16 mg/d, most of which is unabsorbed dietary iron. About 1 mg of endogenous iron is lost in the stool per day. Additional losses occur from the uterus in women, amounting to 0.5 to 1.0 mg/d averaged over a whole month. The requirements for iron per kilogram of body weight are highest during infancy (because of low body stores), periods of rapid growth (adolescence and pregnancy), and periods of excessive loss (menstruation). The RDA assigned by the 2001 DRI Committee for all age groups of men and postmenopausal women is 8 mg/d; the RDA for premenopausal women is 18 mg/d. [16] The recommended intake during pregnancy is 27 mg/d. At birth, even the child of an iron-deficient mother has normal stores because the fetus has priority for available iron. Milk is a poor dietary source of iron, but the AI for infants ages 0 to 6 months has been set at 0.27 mg/d, based on the daily amount of iron in ingested milk (~0.35 mg/L). It is assumed that milk intake and requirements correlate with body size. Therefore, this AI may not be sufficient for infants with a lower milk intake. Three months after birth, the requirement for iron increases because of growth, and the RDA of 11 mg/d for infants ages 7 to 12 months assumes that feedings complementary to milk are in place. It is during this period that the infant is most at risk for iron deficiency. During adolescence, the hemoglobin level rises 0.5 to 1.0 mg/dL per year, requiring 50 to 100 mg of iron per year, and 300 mg during adolescence. Food iron is available in a variety of red meats, nuts, seeds, and egg yolks. Milk products, potatoes, and fresh fruit are poor sources. Iron in vegetables varies according to growth conditions of the plant. Heme iron in vegetables is not so readily available for absorption as that in meat; it also requires reduction to the ferrous state (by ascorbic acid) for maximal absorption. However, heme iron accounts for only about 10% of dietary iron in the United States. The iron content of some foods (e.g., bread flour) is increased by fortification. Vegetarians are at risk for dietary iron deficiency because of the more limited absorption of nonheme iron. Iron is involved in many reactions as a cofactor for enzymes (heme or otherwise) and as a major constituent of heme as an oxygen-carrying cofactor. Iron is absorbed primarily in the duodenum and upper jejunum. The major transmembrane iron transporter is Nramp2, now called *divalent metal transporter DMT1*. [128] The absorption of inorganic iron is enhanced by gastric acid, ascorbic acid, and other organic acids, including the amino acids histidine, lysine, and cysteine, which form iron chelates. [16], [129] Absorption is decreased when iron forms an insoluble complex in the lumen with dietary phytates or phosphates, or with antacids or other medications. About 10% of iron is absorbed normally, but in deficiency this can increase to 30%. After absorption, iron is stored as ferritin in the liver, spleen, and bone marrow. The functional compartment of iron (hemoglobin, myoglobin) accounts for most of the total body iron, ranging from about 2180 mg to 2750 mg for average women and men, respectively.

Zinc. The body contains 1.5 to 2.5 g of zinc, so that it is the second most abundant trace mineral after iron. Although the turnover of isotopic zinc in adults is 6 mg/d, balance studies show that 12.5 mg of dietary zinc is needed to maintain positive balance. [16] The daily loss of 2.5 mg/d is mostly in feces, and absorption ranges from 20% to 40%, depending on the fiber content of the diet (inversely related). The 2001 DRI Committee report set the RDA at 9 and 8 mg for girls and boys ages 9 to 13 years, respectively, and at 8 and 11 mg/d for adult women and men, respectively. [16] The RDA for pregnancy is 13 mg for adolescents and 11 mg for women older than 18 years. One more milligram is added per day during lactation. Like other divalent cations, zinc is absorbed inefficiently along the entire length of the small intestine. A series of zinc transporters have been identified and are thought to be important in absorption, including ZnT-1, which is expressed in duodenal and jejunal villi. [130] Zinc absorption is decreased by lumenal binders (phosphate and others), as is the absorption of calcium and magnesium. [131] Inorganic iron, especially when ingested as a supplement, impairs inorganic zinc absorption. [132] Absorption can be enhanced, however, by animal proteins and sulfur-containing amino acids, as well as by hydroxy acids. [133] Zinc absorption may be increased during states that increase demand for the mineral, including infancy, pregnancy, and lactation. Zinc plays a critical role in the growth and function of cells. It is a cofactor for many enzymes that participate in the metabolism of carbohydrate, fat, and protein. [16] It is necessary for cell growth and proliferation, sexual maturation, reproduction, and dark adaptation and night vision, and it may play a role in wound healing and immune defenses. Finally, it may activate or inhibit enzymes, modify membrane functions, or bind to DNA transcription factors.

Copper. Copper is an essential trace mineral for humans. Estimates of copper requirements are based on balance studies, fecal and other losses, and absorption at each life stage. [16] Obligatory losses for adults are about 580 µg/d, and absorption averages about 25%. Most absorbed copper is excreted in bile, although some biliary copper is reabsorbed via an enterohepatic circulation. [134] When intestinal and biliary losses occur, the copper requirement increases. The AI for infants is based on intake of milk (120 µg/L) and is set at 200 µg for infants ages 0 to 6 months and 220 µg/d for infants ages 7 to 12 months. The RDA has been set at 900 µg/d for men and women 19 years of age or older. [16] The liver contains about one third of the body stores of copper, mostly in enzymes, including ceruloplasmin, cytochrome oxidase, superoxide dismutase, tyrosinase, lysyl oxidase, and histaminase. [135] Copper is important for normal skeletal and nervous system development, erythropoiesis, leukopoiesis, and iron absorption, and as an antioxidant. Copper absorption and intracellular metabolism are complex. A high-affinity copper transport protein, hCtr1, may transport copper across the apical enterocyte membrane. [136] The Menkes disease protein, MNK, is a membrane-associated P-type ATPase that is required for copper secretion into the portal vein. Hephaestin is a multicopper oxidase that is a membrane-bound analog of ceruloplasmin required for iron (but not copper) secretion from the intestine. [137] Absorbed copper is transferred in plasma to ceruloplasmin and albumin, and it is taken up by the liver via hCtr1-mediated transport. It is distributed to various cellular compartments by a series of small cytoplasmic copper chaperones. [136] In all tissue but the liver, MNK mediates copper efflux in the Golgi apparatus and subsequently in the plasma membrane, as the protein moves to the plasma membrane. In the liver, the Wilson disease protein, WD, is similarly placed and may serve similar functions.

Other trace minerals. Selenium is an essential part of the antioxidant system of glutathione peroxidase. [15], [138] Thus, its function is linked with that of vitamin E. Chromium potentiates the action of insulin at the cell receptor level and plays a role as a cofactor for insulin. [139] Manganese is a cofactor for many enzymes with widely varying functions, from superoxide dismutase to hydrolases and kinases, but a deficiency state in humans has not been identified. [16] Iodine is an essential component of the thyroid hormones and is thus integral to their function. [16], [18] Fluoride is concentrated in bones and teeth and is required for normal growth. The major source of fluoride is now fluoridated water. Although deficiency of most of these minerals occurs in humans, the occurrence of such deficiency is unusual in developed countries. For this reason, they are used primarily as TPN supplements. Vanadium, nickel, cobalt, tin, and silicon are considered essential in mammals because deficiencies have been produced experimentally, but human deficiencies have not been reported. [16] Other elements, including cadmium, lead, boron, aluminum, arsenic, mercury, strontium, and lithium, have not yet been proved essential. Most of these elements, with the exception of boron, are probably present in sufficient quantities as contaminants in TPN solutions. [140]

Vitamins

Thiamin (B_1). Thiamin is essential for the function of many enzyme systems and plays a major role in energy production. Thiamin pyrophosphate is a coenzyme in the oxidative decarboxylation of a-ketoacids to aldehydes, and it catalyzes transketolase activity in the pentose phosphate cycle. It is important for nucleotide synthesis and provides cofactors for fatty acid synthesis. Blood pyruvate levels increase with thiamin deficiency. The requirement usually is related to the intake of energy, especially carbohydrate. The RDA for adults is 0.5 mg for every 1000 kcal in the diet. [14] Allowances are based on the effects of varying dietary thiamin and the relationship with signs of clinical deficiency or with urinary excretion of thiamin or serum transketolase activity. Thiamin is synthesized by many plants. It is abundant in all foods and is added to many commercial baked products and cereals. Because thiamin is lost easily in the cooking and processing of food, its content in food varies according to preparation.

Riboflavin (B_2). Riboflavin is a major component of two essential coenzymes: flavin adenine dinucleotide (FAD) and flavin mononucleotide (FMN). It forms the active portion of these coenzymes that are involved in biologic oxidations. Riboflavin requirements are linked with protein or energy intake, like those for thiamin. However, unlike the requirements for thiamin, those for riboflavin are unrelated to food energy intake. The RDA is set at 1.1 mg/d and 1.3 mg/d for adult women and men, respectively. [14] Requirements have been assessed by measuring urinary excretion and by observing signs of deficiency. Riboflavin is available in all leafy vegetables, in meats and fish, and in milk and eggs. The average Western diet contains about 2.7 mg/d. The vitamin can be lost in food processing and by the action of ultraviolet light. Absorption is efficient from the small intestine, and riboflavin is excreted from the kidney unmetabolized.

Niacin (B_3). The term *niacin* encompasses nicotinic acid and its amide form, nicotinamide. It is a component of two coenzymes, nicotinamide adenine dinucleotide (NAD) and nicotinamide adenine dinucleotide phosphate (NADP), which participate in more than 50 metabolic reactions. Humans can synthesize about 1 mg of niacin from 60 mg of the amino acid tryptophan. Deficiency depends on the limited availability of niacin and tryptophan. [141], [142] The RDA is reported in niacin equivalents (NEs) (1 NE = 1 mg of niacin = 60 mg of tryptophan). The RDA has been estimated at 14 mg and 16 mg NE daily for women and men 14 years of age or older, respectively. [14] Average Western diets provide 16 to 24 NE/d. Nicotinic acid is present in most foods, except fats and oils. Meat, fish, and grain products are good dietary sources. Much is lost during grain processing, but it is added back. Niacin is well absorbed in the small intestine.

Pyridoxine (B_6). The term *vitamin B_6* refers to three naturally occurring pyridines: pyridoxine, pyridoxal, and pyridoxamine. All forms function similarly. Vitamin B_6 is essential for the function of many transaminases and amino acid decarboxylases. It is involved in the metabolism of all amino acids and in the synthesis of acetylcholine, porphyrin, arachidonic acid, dopamine, serotonin, and bile acids. Requirements are increased with a higher protein intake. The RDA for adults ages 19 to 50 years is 1.3 mg/d for average protein intake. Low levels of all three forms are present in all foods, with meats, fish, and grains good sources. The vitamin B_6 allowance has been estimated by using a ratio of 0.016 mg of vitamin per grams of protein ingested. Because energy and protein intake is lower in the older population, RDAs are increased for persons older than 50 years, and the estimates for women (i.e., 1.5 mg) are lower than those for men (i.e., 1.7 mg). [14] Vitamin B_6 is synthesized by microorganisms in the intestine, primarily in the colon, where the vitamin is not absorbed. Vitamin B_6 is rapidly absorbed in the small intestine and excreted in the urine as the metabolized product, 4-pyridoxic acid. Certain drugs are pyridoxine antagonists (e.g., isoniazid, hydralazine, penicillamine). A deficiency of vitamin B_6 and other B vitamins can occur in chronic alcoholics.

Folate (folacin, folic acid). *Folacin* is a generic term for compounds that have a structure and function similar to that of folic acid (pteroylglutamic acid [PGA]). The many forms differ in the degree of reduction of the double bonds in the ring structure (e.g., tetrahydrofolate), the presence of 1-carbon groups (e.g., methyltetrahydrofolate), and the number of glutamyl residues in the peptide chain (e.g., folate pentaglutamate). Folate functions as a carrier of 1-carbon groups from donor to recipient molecules and is necessary for the synthesis of nucleic acids, the initiation of protein synthesis, and the synthesis of acetylcholine and methionine. Three other vitamins—cobalamin, ascorbic acid, and niacin—are involved in converting folate to its active coenzyme forms. [14], [142] PGA is the form of folate used commercially, and it is a relatively poor substrate for dihydrofolate reductase. As a result, tissue utilization is much poorer for PGA than for natural methylated or reduced folates found in food. However, folate requirements have been based on replacement with PGA. The RDA for folate has been modified dramatically from the 1989 estimate of 200 µg/d for adults [10] to the current level

of 400 µg. [14] This change was the result of recognition of the role of folate in reducing the incidence of neural defects, [143] and the role of elevated serum homocysteine concentrations as a cardiovascular risk factor. [144] Dietary folate is now reported in folate equivalents, in recognition of the greater bioavailability of synthetic folic acid compared with natural folate. The RDA for women ages 14 to 50 years is 400 µg/d and should comprise synthetic folic acid plus dietary folate. During pregnancy, the amount of synthetic folic acid is specified at 400 µg/d. [14] The vitamin is abundant in citrus juices, enriched cereals and breads, legumes, liver, nuts, and green leafy vegetables. It occurs in food largely in the polyglutamate form, and its bioavailability is generally high. However, boiling, steaming, or frying can lead to significant losses. Folate is absorbed in the proximal small intestine by active transport. It is converted in the liver and other tissues to the 5-methyltetrahydrofolate form and is stored as polyglutamate. [145] Total body stores are relatively small and can be depleted in a few months if dietary intake is negligible. It can enter the serum or the bile and be reabsorbed, undergoing an enterohepatic circulation amounting to about 100 µg/d. In diseases that cause malabsorption, body stores of vitamin B_6 are lost more rapidly than in simple dietary deficiency.

Cobalamin (vitamin B_{12}). Cobalamin contains a cobalt atom to which are bound active groups, including hydroxy-, methyl-, and nitrocobalamin. The vitamin functions as a carrier for methyl and hydrogen groups. It is required for two enzyme reactions, methionine synthase and methylmalonic acid mutase, and thus participates in methionine and succinyl-CoA synthesis. The total body content of cobalamin is 2 to 2.5 mg, most of which is in the liver, and the half-life is 1.5 to 3.5 years. Thus, daily losses average about 1.3 µg/d. The RDA was set at 2 µg/d for adults, based on 70% absorption efficiency. [10] However, because 10% to 30% of adults older than 51 years may have protein-bound cobalamin malabsorption, the new RDA has been set at 2.4 µg/d. [14] Although this malabsorption probably is caused by reduced pepsin and gastric acid secretion, most elderly persons have some intrinsic factor. Thus, it is recommended by some experts that most intake in older persons be in the form of a dietary supplement, to ensure its adequacy. [146] The vitamin is synthesized only by bacteria and enters animal tissues after the ingestion of contaminated foods or production in the lumen. It is found only in animal products, including meat, fish, eggs, and milk. The average Western diet contains 5 to 15 µg/d. Cobalamin is relatively stable during cooking and processing. The absorption of cobalamin is complex. [147] In food, it is bound to enzymes from which it is released by gastric proteases. It is bound to haptocorrin in the stomach, released from haptocorrin by the action of pancreatic enzymes, and then bound to intrinsic factor, with which it is absorbed in the ileum by the process of receptor-mediated endocytosis. The receptor for the intrinsic factor–cobalamin complex, cubilin, is a large, multifunctional protein that also binds apolipoproteins. [148] In the enterocyte, the vitamin is released from intrinsic factor, bound to transcobalamin II, and delivered to the tissues complexed to that protein. It is released into the bile bound again to haptocorrin, and 5 to 10 µg/d undergoes an enterohepatic circulation. As with folate, body stores are lost more rapidly if malabsorption is present because endogenous as well as dietary cobalamin is lost.

Ascorbic acid (vitamin C). Ascorbic acid is an essential cofactor for several hydroxylation reactions and plays a key role in the synthesis of collagen. It may also function as an antioxidant for vitamins A and E, and it is involved in the formation of norepinephrine and serotonin. This role in neurotransmitter synthesis may explain the fatigue and weakness seen in scurvy. [149] A daily intake of 10 mg of ascorbic acid per day cures clinical signs of scurvy but does not maintain body stores. The previous RDA of 60 mg/d [10] produced wide fluctuations in plasma levels. The ascorbic acid requirement is also compounded by its possible chemoprotective value in some disorders (e.g., colon cancer, heart diseases, cataracts), at doses far in excess of those needed to prevent scurvy. Intake of 200 mg of ascorbic acid per day is needed to begin to saturate tissues, an intake that approximates the vitamin C content of diets (~225 mg) that allow maximal protective effects of the vitamin. [150] The new DRI values represent a compromise between the old RDA value and those needed for chemoprevention and are set at 75 mg/d and 90 mg/d for adult women and men, respectively. [14] During pregnancy and lactation, the RDA should be increased by 10 mg/d and 45 mg/d, respectively. The vitamin is especially concentrated in green vegetables and citrus fruits, although it is widespread among all foods. The food content depends on the state of ripeness and on the method of preparation because ascorbic acid is very sensitive to heating and oxidation. It is well absorbed in the small intestine by two sodium-dependent cotransporters (SVCT1 and SVCT2) at intakes of up to 180 mg/d. [151] Over that amount, the proportion appearing in the stool increases, and at high doses (>3–4 g/d), diarrhea can result.

Biotin. Biotin is a coenzyme for various carboxylases and is important in the metabolism of carbohydrate, protein, and fat. [152] It is produced by colonic bacteria and absorbed in the small bowel and colon by facilitated diffusion. Because of this endogenous production, an RDA has not been determined, but the AI for adults has been estimated at a DRI of 30 µg/d. [14]

Pantothenic acid. Pantothenic acid is a precursor of CoA, which is essential for the metabolism of fats, carbohydrates, and proteins and for the synthesis of steroids and porphyrins. [14, 153] It is widely distributed in foods, especially animal tissues, whole grain cereals, and legumes. Microflora may produce some pantothenic acid, but the data in humans are not clear. Although an RDA has not been established, the estimated AI for older adolescents and for adults is 5 mg/d. Pantothenic acid is well absorbed in the intestine and is excreted unchanged in the urine.

Vitamin A. *Vitamin A* is the collective term for vitamin A alcohol (retinol) and its related biologically active forms. It is essential for growth and development, the maintenance of epithelial cells, the stability of cell membranes, reproduction, and vision in dim light. [16, 154] The ß-carotenes are precursors of vitamin A and appear to have additional beneficial functions, probably as antioxidants. [155] The DRI allowance for vitamin A is based on many nutritional studies and amounts to 900 µg of retinol per day for men and 700 µg/d for women. [16] The recommendations for ß-carotene are even more complex because no reproducible biologic activities are available to establish adequate intake. Some epidemiologic studies show correlations between low serum levels (still in normal range), but no positive results in intervention studies. [156] Moreover, some carotenoids (e.g., lutein) are accumulated in the retina preferentially, whereas others (e.g., lycopene) lack provitamin A function but exhibit other biologic activity. The data were judged by the DRI Committee to be insufficient to make a recommendation on the required percentage of dietary vitamin A that must be derived from provitamin A carotenoids. [15] Vitamin A activity in foods usually is expressed in international units (IUs) and an equivalence established with retinol activity, accounting for both vitamin A and provitamin compounds in the diet. The previously accepted 6:1 equivalence of ß-carotene to vitamin A [10] has been questioned because of inefficient bioconversion of plant carotenoids. Thus, the new conversions are 1 retinol equivalent (RE) = 1 µg of all-*trans*-retinal, or 12 µg of ß-carotene, or 24 µg of mixed carotenes. One RE is also equivalent to 3.3 IU of retinol activity or 10 IU of ß-carotene activity. This means that older food tables have overestimated the vitamin A activity in foods. This change may be of especial importance in underdeveloped countries that rely mostly on vegetable products for their vitamin A. Animal products are rich sources, including liver, kidney, dairy products, and eggs. Carotenoids, especially ß-carotene, are found in green and yellow vegetables. There are more than 400 carotenoids in foods, only about 60 of which have provitamin A activity. In the United States, dairy products and margarines are supplemented with retinyl esters, and these products are the major dietary source of the vitamin. Vitamin A, ingested in the form of long-chain retinyl esters, is hydrolyzed to retinol by lipases and esterases in bile and pancreatic secretions or in milk. More than 80% of the vitamin is absorbed passively, reincorporated into retinyl esters, packaged into chylomicrons, transported in the lymphatic circulation, and stored in the liver, which controls its release. [16, 157] Carotene is absorbed passively, but less well than vitamin A (40%–60%). Most is hydrolyzed to retinol inside the enterocyte, and a small amount is absorbed intact. After vitamin A is released from the liver, it is transported in the plasma as a trimolecular complex with retinol-binding protein and transthyretin. [157, 158] If stores are adequate, any excess vitamin A is excreted in bile. A very small amount is excreted in urine along with other metabolites. About 10% of hepatic retinol is converted to retinoic acid, which in turn is conjugated with glucuronide and undergoes enterohepatic circulation. As in the case of folate, cobalamin, and 1,25-dihydroxyvitamin D, this enterohepatic circulation leads to the loss of endogenous as well as dietary vitamin A if malabsorption occurs.

Vitamin D. *Vitamin D* is the designation for a group of sterols and their metabolites that have antirachitic activity. [13] Cholecalciferol (vitamin D_3) is formed in the skin from 7-dehydrocholesterol by the action of ultraviolet light, and about 100 IU/d (10 µg = 400 IU) is produced in persons living in the temperate zone. [159] Ergocalciferol (vitamin D_2) is present in plants. Vitamin D is a provitamin, with the active metabolite being 1,25-dihydroxyvitamin D. The active vitamin promotes the intestinal absorption of calcium and phosphorus, in conjunction with parathyroid hormone. The vitamin is necessary for normal bone formation and regulates calcium and phosphorus metabolism in bone and kidney. [160] Because more than 90% of circulating 25-hydroxyvitamin D_3 is derived from skin and is endogenously produced, requirements have not been established for daily need. The current DRIs are based on AIs. Because vitamin D deficiency is more prevalent in older adults, the recommended AI for adults older than 50 years is twice that of younger adults (10 versus 5 µg/d, or 400 versus 200 IU), and for adults older than 70 years it is tripled (15 µg/d). [13] Vitamin D occurs naturally in foods of animal origin, such as fish liver oils, eggs, liver, and dairy products. Fortified foods now supply the major dietary sources, although most vitamin D is made endogenously. Vitamin D is absorbed in the small intestine along with other lipids, but it requires bile salt micelle formation for efficient absorption. It is transported in the lymph with chylomicrons and other lipoproteins. The serum content of vitamin D is not limited by the content of the specific vitamin D–binding protein, and the ingestion of excessive doses can produce toxic levels of vitamin D or its metabolites in the serum. Vitamin D is stored in fat depots (e.g., liver, adipose tissue) and muscle, and it is released slowly. The 25-hydroxyl group is added in the liver, and the 1-hydroxyl group in the kidney. 1,25-Dihydroxyvitamin D and other polar metabolites are excreted in the bile and undergo an enterohepatic circulation. Malabsorption disorders produce earlier deficiency because endogenous as well as exogenous vitamin is lost. Small amounts of the vitamin are excreted in the urine.

Vitamin E. The term *vitamin E* refers to two groups of lipid-soluble compounds, tocopherols and tocotrienes, that are found in plants. a-Tocopherol is the most active and abundant of these compounds. These compounds function as fat-soluble antioxidants and free-radical scavengers, in conjunction with the selenium/glutathione peroxidase system. [161] The RDA previously was based on the assumptions that the diet had no more than 0.1 ppm of selenium, average amounts of sulfur amino acids, a ratio of vitamin E to PUFAs of 0.4, and less than 1.5% linoleic acid in a diet containing 1800 to 3000 kcal. [10] Current recommendations also consider the possible role of vitamin E as an antioxidant in preventing disease, the increased intake of PUFAs in the United States, and the serum vitamin E values from NHANES III (Third National Health and Nutrition Examination Survey, 1998–1994). [15] Thus, the new DRIs are set about 50% higher than the 1989 levels (15 mg/d for adults). The vitamin is abundant in the lipids of green leafy plants, in vegetable oils, and in seeds. Foods high in PUFAs are also excellent sources of vitamin E. [15, 161] Vitamin E is absorbed passively from the small intestine with other lipids, and like that of other fat-soluble vitamins, its absorption requires bile salt micelles. Only about 40% of an oral dose is absorbed. The natural form is an acetate ester; bile salt–dependent pancreatic esterase is also required for absorption. There is no specific serum carrier for vitamin E. Because it is bound to LDL and other lipoproteins, serum levels are proportional to total lipids. The vitamin is delivered to the liver after the action of lipoprotein lipase, and from the liver it is delivered in lipoproteins to adipose tissue, where it is stored. Vitamin E destroys tissue peroxides that promote the oxidation of LDL [162] and prevents platelet adhesion by an antioxidant-independent mechanism. [163] These effects are the basis for the potential effect of the vitamin in preventing ischemic damage.

Vitamin K. The term *vitamin K* designates naphthoquinone compounds with antihemorrhagic activity. They are critical for the production of plasma clotting factors II (prothrombin), VII, IX, and X. Two

forms occur naturally: vitamin K_1 (phylloquinones) in green plants and vitamin K_2 (menaquinones) in bacteria. Colonic bacterial synthesis provides an estimated 2 µg/kg of body weight, and absorption of that source presumably occurs by backwash into the terminal ileum. The RDAs of 1989 were based on the function of the vitamin for clotting proteins, but the requirement may be greater for the nonhepatic vitamin K–dependent proteins, especially those in bone. [164] Because of the lack of data on which to estimate an average requirement, the current DRIs are based on AIs, set at 90 µg/d and 120 µg/d for adult women and men, respectively. [16] Therapeutic sources of vitamin K are synthetic compounds. The best food sources are green leafy vegetables, and they are considered to be the major source of the vitamin despite some endogenous production. Vitamin K is passively absorbed in the small intestine, a process that requires bile salt micelles and pancreatic enzymes. Unlike the other fat-soluble vitamins, vitamin K is not stored in large amounts in adipose tissue. The plasma form is carried on lipoproteins, but the storage form is primarily long-chain menaquinones. The vitamin is concentrated in the liver and is excreted in bile, stool, and urine. [16, 160] Vitamin K acts by carboxylating selected glutamic acid residues of proteins to form a-carboxyglutamic acid (Gla), which binds calcium. [165] The coagulation function of the vitamin K–dependent hepatic proteins (e.g., prothrombin and factors VII, IX, and X) and the function of the bone proteins (e.g., osteocalcin, matrix Gla protein) are proportional to the degree of carboxylation. Other vitamin K–dependent proteins of unknown function are present in other tissues.

ALTERED NUTRITIONAL STATES

Starvation

The metabolic response to starvation enhances survival by increasing the use of adipose tissue triglycerides as a source of fuel, preventing severe hypoglycemia, conserving lean tissue, and decreasing the metabolic rate. The duration of survival during starvation depends on the amount of body fat and lean body mass. In lean men, death occurs after approximately 60 days of starvation, [166] whereas obese persons can survive complete energy deprivation for more than 1 year without obvious adverse consequences. [167]

Marked metabolic adaptations occur within the first 24 hours of fasting. The mobilization of adipose tissue triglycerides, ketone body production, and the oxidation of plasma fatty acids increase, whereas hepatic glucose production and plasma glucose oxidation decrease. [168]

The rate of lipolysis of adipose tissue triglycerides increases because of a decrease in circulating insulin, an increase in plasma epinephrine concentration, and an increase in the lipolytic response to catecholamines. [169, 170] and [171] After 3 days of fasting, lipolysis of adipose tissue triglycerides increases to more than double the values observed after an overnight (i.e., 12-hour) fast. The increase in lipolysis and plasma fatty acid concentrations increases fatty acid oxidation. The increased delivery of free fatty acids to the liver in conjunction with a decrease in the plasma ratio of insulin to glucagon stimulates hepatic ketone body production. [172] The rate of ketogenesis is maximal by 3 days of starvation; plasma ketone body concentration increases 75-fold by 7 days. [173] Ketone bodies are water soluble and able to cross the blood-brain barrier. As plasma ketone body concentrations increase, ketone body oxidation by the brain increases; by 7 days of starvation, ketone bodies provide 70% of the brain's energy needs. [174] The shift in fuel use by the brain helps spare the limited supply of plasma glucose for glucose-requiring tissues.

Whole-body glucose production decreases by more than half during the first few days of fasting because of a marked reduction in hepatic glucose output. [175] Only 15% of hepatic glycogen stores remain after 24 hours of fasting. [176] Therefore, the contribution of gluconeogenesis from plasma precursors to total hepatic glucose output increases as the rate of hepatic glycogenolysis declines. As fasting continues, the conversion of glutamine to glucose in the kidney represents almost 50% of total glucose production.

Normally, about 70 g of amino acids are mobilized from protein stores and about 10 g of nitrogen is excreted in the urine. [177] During starvation, it is critical to slow down the rate of protein breakdown to prevent clinically significant protein losses. If protein breakdown proceeded at a normal rate throughout starvation, a potentially lethal amount of muscle protein would be catabolized in less than 3 weeks. The increase in ketone bodies [178] and starvation-induced inactivation of thyroid hormone (conversion of triiodothyronine [T_3] to reverse T_3) [179] directly inhibit muscle protein breakdown.

Energy expenditure is conserved during fasting because of a decrease in physical activity caused by fatigue and a decrease in the resting metabolic rate, which decreases by 10% to 15% at 7 days because of diminished size and function of metabolically active tissues, [180] increased conversion of active thyroid hormone to its inactive form, [181] and suppressed sympathetic nervous system activity. [182]

Maximum adaptation occurs as starvation continues. After 14 days of fasting, lipid, carbohydrate, and protein metabolism plateaus. Adipose tissue provides more than 90% of daily energy requirements. Muscle protein breakdown decreases to less than 30 g/d, causing a marked decrease in urea nitrogen production and excretion. Fluid requirements decrease because the diminished osmotic load from urea causes a decline in urine volume to 200 mL/d. Total glucose production decreases to about 75 g/d, providing fuel for glycolytic tissues (40 g/d) and the brain (35 g/d) while maintaining a constant plasma glucose concentration. Energy expenditure decreases by 20% to 25% after 30 days of fasting [183] and remains relatively constant thereafter despite continued starvation.

During the terminal phase of starvation, body fat mass, muscle protein, and the size of most organs are markedly decreased. The weight and protein content of the brain remains relatively stable throughout starvation. In rodent models, when fat stores are depleted, the energy derived from body fat decreases and muscle protein catabolism increases. Death occurs when 30% of muscle protein is lost. [184]

Metabolic Response to Illness and Injury

The metabolic response to illness and injury is characterized by hypermetabolism, negative nitrogen balance, insulin resistance and hyperglycemia, and increased mobilization and oxidation of adipose tissue triglycerides. These events are mediated by a complex cascade of endogenous mediators that cause a predictable physiological response. Increased production and secretion of the counterregulatory hormones (e.g., catecholamines, glucagon, and glucocorticoids) and cytokines are probably responsible for most of the observed responses to illness and injury.

In the 1930s, the classical work of Cutherbertson [185] on long-bone fractures provided the basis of our understanding of the metabolic response to injury. Cuthbertson demonstrated that the response to injury could be divided into two phases: the early ebb phase (12–24 hours after trauma) and the subsequent flow phase. The ebb phase is characterized by decreased blood pressure, oxygen consumption, cardiac output, and body temperature. The flow phase is characterized by hypermetabolism and increases in oxygen consumption, cardiac output, body temperature, and the urinary excretion of nitrogen, potassium, and phosphorus. Subsequently, Moore [186] divided the flow phase into the catabolic and anabolic phases. The restoration of tissue perfusion marks the beginning of the catabolic phase and lasts for days to weeks, depending on the severity of injury, medical intervention, and the premorbid health of the patient. This phase is characterized by catabolism, heat production, negative nitrogen balance, and hyperglycemia. [185, 186] It ends after volume deficits are corrected, infection is controlled, pain is eliminated, and oxygenation is restored. At this point, net anabolism may occur, resulting in a slow reaccumulation of protein and body fat.

Hormone and Cytokine Mediators During the flow phase, the secretion of several hormones is increased, and they act synergistically to generate alterations in intermediary metabolism. Catecholamines increase lipolysis and hepatic glucose production. Glucagon increases hepatic gluconeogenesis and glycogenolysis. Cortisol enhances protein breakdown and increases hepatic gluconeogenesis. Cytokines, produced by macrophages, lymphocytes, Kupffer cells, and endothelial cells, are also critical mediators of the metabolic response. TNF is believed to be the primary cytokine mediating many of the responses to infection and trauma, including fever, increased acute phase protein synthesis, protein catabolism, hypotension, decreased lipoprotein lipase activity, and metabolic acidosis. [187, 188, 189, 190] and [191] IL-1 and TNF act synergistically in promoting tissue injury and stimulating the release of counterregulatory hormones. [192] IL-1 causes fever, hypotension, and increased insulin and glucagon secretion, and it affects the concentrations of plasma divalent cations. IL-1 stimulates the liver to synthesize metallothioneins, which sequester zinc, and mediates the sequestration of iron in hemosiderin and ferritin, depriving invading organisms of these trace elements. [189, 190, 191, 192, 193, 194, 195, 196, 197, 198] and [199] IL-1 stimulates other portions of the acute phase response, such as fibrinogen and C-reactive protein production by hepatocytes and the release of lactoferrin by neutrophils. [193, 194] and [195] IL-1 enhances immunoglobulin production by B lymphocytes and is a potent stimulus for the synthesis and release of IL-2 by T lymphocytes. [193, 195, 200] IL-6 is also inducible by TNF and functions primarily as a stimulator of the hepatic acute phase protein response and lymphocyte proliferation. [192, 200] IL-8 is inducible by TNF and IL-1 and is a potent neutrophil chemoattractant. [200]

Energy Metabolism Increases in metabolic rate correlate directly with the severity of illness and injury. [201] For example, the REE increases after uncomplicated surgery by about 10%, after long-bone fractures by 20%, and after multiple trauma by 50%. The REE rarely exceeds twice the normal rate regardless of the extent of injury. [184] In fact, even patients with severe burns usually do not experience an increase in REE by more than 50% for any prolonged period. [202] In certain types of injury, a temporary resetting of the hypothalamic thermoregulatory set point is responsible for a 1° to 2°C elevation of body temperature known as *posttraumatic fever*. [187, 203] The REE increases about 12% for each 1°C increase in core body temperature. Because the central temperature set point is higher, the comfort temperature for an injured patient is elevated, and elevating the ambient temperature decreases the energy requirement. [184]

Protein Metabolism Illness or injury increases protein synthesis and protein breakdown. The increase in protein breakdown is greater than the increase in protein synthesis, resulting in negative nitrogen balance. Skeletal muscle is the major site of protein catabolism, but increased catabolism of extracellular proteins, including acute phase reactants, coagulation system proteins, and complement system proteins, also occurs. [193, 204, 205] and [206] Considerable protein synthesis is directed toward host defense, including phagocyte activity, hormones, cytokines, intracellular proteins,

immunoglobulins, complement, coagulation system proteins, and acute phase reactant glycoproteins. [193] The composition of amino acids released by muscle does not reflect the composition of muscle proteins. The branched-chain amino acids of skeletal muscle are metabolized within the muscle cell for energy, and transamination generates glutamine and alanine. As a result, alanine and glutamine, which constitute only 12% of muscle protein, make up 50% to 60% of the amino acids released into the plasma by muscle. Conversely, branched-chain amino acids make up 15% of the muscle protein but only 6% of the amino acids released. [193], [201] Glutamine is taken up and metabolized by the kidney at an accelerated rate, providing additional ammonium for excretion to help maintain acid-base balance in the face of the acidosis that frequently accompanies critical illness. [201] In addition to carrying amino groups from the periphery to the liver and kidney, glutamine serves as a major energy source for the lymphocytes, fibroblasts, and the gastrointestinal tract. [207] Alanine is captured by the liver for gluconeogenesis, and its nitrogen contributes to the increase in ureagenesis. [208] During severe illness, nitrogen losses may reach 20 to 40 g/d. This represents catabolism of 600 to 1200 g of lean body mass per day. Providing exogenous nitrogen and energy may [209] or may not [210] decrease the rate of catabolism, but it enhances protein synthesis and thereby decreases negative nitrogen balance. [207], [211]

Carbohydrate Metabolism The stress response is marked by hyperglycemia, in large part related to hepatic gluconeogenesis fueled by lactate, pyruvate, glycerol, alanine, and other glucogenic amino acids. Hepatic glucose production may exceed 500 g/d and is resistant to suppression by insulin. [212] Peripheral insulin resistance decreases skeletal muscle glucose uptake, which also contributes to hyperglycemia. [187] Glucose consumption by wounds and injured extremities is increased to provide fuel for inflammatory cells, such as fibroblasts, macrophages, and leukocytes. These cells metabolize glucose anaerobically and can release large quantities of lactate into the bloodstream, which is subsequently recycled to glucose by the liver.

Lipid Metabolism Fat is a major oxidative fuel in critically ill patients, even when exogenous carbohydrates are administered. The rate of lipolysis correlates directly with the severity of illness. [213] The increased delivery of fatty acids to the liver stimulates very-low-density lipoprotein (VLDL) production. However, the rate of VLDL secretion is not adequate to remove the excessive supply of fatty acids and thus contributes to hepatic fat accumulation. Hypertriglyceridemia can occur because of both increased VLDL production and decreased peripheral lipoprotein lipase activity and VLDL clearance.

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CHAPTER 22

Karen D. Crissinger and D. Neil Granger

GASTROINTESTINAL BLOOD FLOW

The ultimate function of the gastrointestinal tract is to assimilate nutrients and water from the external environment and make them available to cells throughout the body. Although the mucosal epithelium is largely responsible for extracting nutrients from the external environment, the blood and lymph circulations provide the conduits for transferring absorbed nutrients and water to the entire body. The vascular supply to the gastrointestinal mucosa is particularly well suited for the absorptive and secretory functions of this tissue in that it can accommodate a high rate of blood flow, has a large exchange surface area, and permits easy permeation of nutrients and water, yet largely retains proteins within the plasma compartment. This chapter summarizes current concepts regarding circulatory control and function in the gastrointestinal tract during normal physiological conditions, and it reviews current knowledge about pathophysiological mechanisms of intestinal injury resulting from periods of inadequate perfusion (i.e., ischemia).

ANATOMY OF THE GASTROINTESTINAL CIRCULATION

Extramural Vessels

Arteries In humans, the major arteries supplying the stomach and intestines are the celiac, superior mesenteric, and inferior mesenteric arteries. The celiac artery supplies the stomach, the first portion of the duodenum, a portion of the pancreas, and the liver. The superior mesenteric artery supplies the remainder of the pancreas and duodenum, the jejunum, the ileum, and the colon through two thirds of the transverse segment. The inferior mesenteric artery supplies the remainder of the colon and rectum, except the distal rectum, which is supplied by rectal arteries arising from the internal iliac arteries. Along the mesenteric border of the intestine, arterial and venous branches form multiple arcades, anastomose with one another, and provide a pathway for collateral blood flow. The arcades give rise to vasa recta, which branch to encircle the intestine and ultimately pierce the circular muscle. [1], [2] and [3]

Veins Blood from the stomach, pancreas, and intestines drains into the portal vein, except for that of the distal rectum, which drains into the internal iliac veins. The vessels that drain the intestines course within the mesentery, except those vessels supplying retroperitoneal portions. [1], [2] and [3]

Lymph Lymph vessels are closely associated with the arteries supplying the stomach and intestines, ultimately draining into the intestinal lymph trunk to the cisterna chyli, and then into the systemic circulation through the thoracic duct into the left subclavian vein. In humans, the lymphatic drainage of the distal portion of the intestines occurs by way of the left lumbar lymph trunk, which empties into the cisterna chyli separate from the drainage of the proximal gut. [1], [2] and [3]

Intramural Vessels and Microcirculation

Stomach In the human stomach, submucosal arterioles branch into capillaries at the base of the gastric glands, pass perpendicularly through the mucosa, form a lumenal capillary network, and drain into mucosal venules only at the most lumenal level of the lamina propria (Fig. 22-1). These venular branches converge on infrequent mucosal collecting venules, which then pass directly to the submucous venous plexus without receiving any direct capillary tributaries within the mucosa. [4]

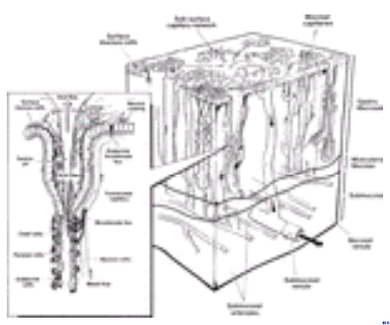

FIGURE 22-1. The vascular organization in gastric mucosa (**right**). The proposed mechanism for the vascular transport of HCO_3^- is toward the surface mucous cells from deeper within the mucosa (**inset left**). (From ref. [4].)

Small Intestine The major intramural arterial vessels are located in the deep submucosal plexus. Both arteries and veins in the deep submucosal plexus undergo extensive self-anastomosis. The capillaries of the muscular, submucosal, and mucosal layers are supplied by branches from the submucosal vascular network. [1], [2]

Villus microvascular architecture varies considerably among species (Fig. 22-2). [5] Human villi contain single, eccentrically located arterioles that pass to the villus tip, break up in a fountain-like pattern, and anastomose with eccentrically located venules that start at about 15% of the villus height below the villus tip. A tufted pattern of capillaries, derived from the tubular capillary plexus surrounding the epithelium of the crypts, supplies the basal 70% to 80% of the villus and also drains into the venule high in the villus (Fig. 22-3). [2], [6]

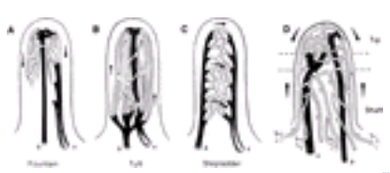

FIGURE 22-2. Models of mucosal microcirculation of the intestine. **A:** Fountain. **B:** Tuft. **C:** Stepladder patterns of villus blood supply. **D:** The dual pattern comprises both fountain and tuft components. *a*, arteriole; *v*, venule; *arrows*, principal direction of capillary blood flow in each pattern. (From ref. [5].)

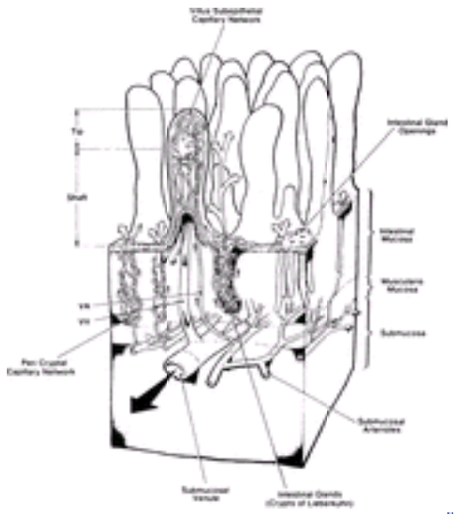

FIGURE 22-3. Mucosal microcirculatory patterns typical of human and rabbit small intestine. *VA*, villus arteriole; *VV*, villus venule. (Adapted from ref. [6].)

Anatomic requirements for countercurrent exchange include the presence of a countercurrent flow of blood and a short arteriolar-capillary distance. Although the existence of countercurrent exchange within the villi has been proposed, [7] controversy continues about whether these requirements are met in human villi. [5]
Colon The colonic mucosa is devoid of villi; therefore, the arterioles and their capillary branches pass to the epithelial surface between the crypts to form a network of capillary plexuses around the crypts. The colonic capillaries are much closer to the epithelial cells than are the villus capillaries in the small intestine. [8]

Intramural Lymphatic Circulation

The gastric lymphatics normally begin as a plexus of vessels immediately superficial to, within, and below the muscularis mucosae. The upper two thirds of the gastric lamina propria is normally devoid of lymphatics. [9]

Small intestine lymphatic vessels begin as centrally located, blind-ending vessels (i.e., lacteals) in the villi and are emptied when smooth muscle in the villus core contracts. These initial lacteals—similar in size to venous capillaries but with open endothelial intercellular junctions, few pericytes, and tenuous basement membranes—drain into a submucosal plexus connected to the collecting lymphatics. The lymphatics leave the intestine at the mesenteric border and pass through the mesentery in association with blood vessels. [2], [10]

Colonic lymphatics begin at the level of the muscularis mucosae, are smaller and more sparsely distributed than those in the small intestine, and do not penetrate higher than the bases of the crypts in humans. [11]

BLOOD FLOW REGULATION

Intrinsic Control

It is well recognized that blood flow in the gastrointestinal tract is normally maintained within narrow limits and changes in response to various functional stimuli. This ability to modulate blood perfusion in accordance with the moment-to-moment demands of the tissue has been attributed to intrinsic vasoregulatory systems. Several regulatory mechanisms have been proposed to explain vascular phenomena such as pressure-flow autoregulation and functional hyperemia in the gastrointestinal tract. Of these, metabolic, myogenic, and hormonal mechanisms are considered to be of the greatest physiological significance. The following section summarizes the available data implicating intrinsic factors in the regulation of gastrointestinal blood flow and the specific mechanisms that have been invoked to explain these vascular phenomena.

Pressure-Flow Autoregulation Pressure-flow autoregulation has been demonstrated in stomach, [29], [30] small intestine, [22], [31], [32], [33], [34], [35], [36], [37], [38] and [39] and colon. [12], [13], [40], [41] and [42] Autoregulation of blood flow in the gastrointestinal tract during fasting is not the intense phenomenon observed in kidney and brain; however, the results of several studies indicate that the intensity of autoregulation increases during periods of enhanced functional activity (Fig. 22-4). [14] Furthermore, it appears that the autoregulatory ability of the more metabolically active mucosal region of the intestine exceeds that of the whole organ. [43]

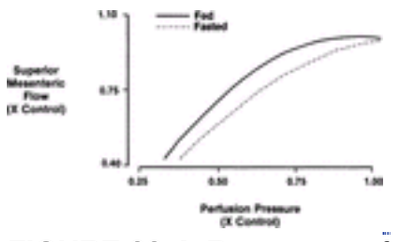

FIGURE 22-4. Responses of intestinal blood flow to step reductions in perfusion pressure in fed (*solid line*) versus fasted (*dashed line*) dogs demonstrate an increased intensity of autoregulation during enhanced functional activity. (From ref. [14].)

The correlation between metabolic rate and autoregulatory ability does not appear to apply to the neonate because autoregulation of blood flow is less intense or absent in neonatal intestine, which is more metabolically active than adult intestine. [44] Additionally, autoregulation is enhanced by hypoxia or ischemia [45] or by increasing intestinal oxidative metabolism in 1-month-old, but not in 3-day-old, piglet intestine. [46] One study suggests that autoregulation in the intestine is an endothelium-dependent phenomenon. [47] Although blood flow is not perfectly regulated with an arterial pressure of 100 to 50 mm Hg, oxygen uptake remains within normal limits over the same range of pressures. [12], [13] This autoregulation of gastrointestinal oxygen uptake is often cited as evidence that tissue oxygenation, rather than blood flow, is the controlled variable. As arterial pressure is reduced, vascular resistance falls, whereas perfused capillary density rises. These changes enhance both the convective and diffusive exchange of oxygen during the reduced pressure state. As a result, tissue oxygen tension tends to remain above the level at which oxygen availability limits mitochondrial oxygen consumption. Mathematical models of intestinal oxygen exchange predict that the contribution of capillary recruitment exceeds that of vasodilation in maintaining normal tissue oxygenation during acute hypotension. [48]
Reactive Hyperemia The term *reactive hyperemia* is used to describe the overshoot in blood flow that occurs after the release of arterial occlusion. All regions of the gastrointestinal tract exhibit a reactive hyperemia after brief (<5 minutes) periods of arterial occlusion. [49] The magnitude and duration of the hyperemic response are related to the extent and duration of the arterial occlusion, suggesting that the vascular response is caused by metabolite accumulation, oxygen deficiency, or both. The ability of the intestine to repay the oxygen debt incurred during vascular occlusion is largely determined by which vessel is occluded—that is, artery or vein. With arterial occlusion, repayment of the oxygen debt is inadequate, and the magnitude of the deficit is proportional to the duration of the arterial occlusion. [50], [51] Venous occlusions are associated with overpayment of oxygen in the postocclusion period, the magnitude of which is related to the duration of occlusion. It is suggested that arterial occlusions depress intestinal oxygen utilization, whereas venous occlusions enhance it. Increasing basal intestinal oxygen consumption by intraenteric placement of nutrients prolongs the reactive hyperemic response and increases the oxygen payback-to-debt ratio. [100] Laser Doppler studies indicate that mucosal and total blood flow consistently show reactive hyperemia in response to a 60-second occlusion, but the muscularis externa does not.
Alterations in Arterial Blood Gases and Hematocrit Alterations in arterial blood gases and hematocrit also affect gastrointestinal blood flow. Arterial hypoxemia increases blood flow and elicits capillary recruitment in denervated intestinal preparations. [52], [53] The vasodilation and increased perfused capillary density tend to minimize the reduction in oxygen uptake induced by the limited oxygen delivery. When blood flow is held constant, the intestine maintains oxygen consumption within 48% of control during arterial hypoxemia. When both blood flow and capillary density are free to increase, however, oxygen uptake remains within 26% of control despite the hypoxia. Similar to hypoxia, hypercapnia induces a marked relaxation of resistance vessels. [53] In contrast to hypoxia, hypercapnia causes the precapillary sphincters to constrict and the capillary density to decrease. Alterations in arterial hematocrit also influence gastrointestinal blood flow and oxygenation. [15], [16] and [17] An inverse linear correlation exists between the intestinal blood flow and the hematocrit, and a direct linear correlation between the arteriovenous oxygen difference and the hematocrit in both the intestine and stomach. The relation between intestinal oxygen uptake and hematocrit is parabolic (Fig. 22-5), [15] showing a maximal uptake at a hematocrit of 48.7%—that is, the optimal hematocrit. Intraenteric placement of nutrients increases the optimal hematocrit to 57.1%. In the stomach, the

optimal hematocrit is 38.2% during resting conditions, and it increases to 45.7% during pentagastrin stimulation. In the neonate, intestinal blood flow and oxygen uptake are not changed by alterations in hematocrit over a range between 10% and 54%, indicating that increases in oxygen extraction play a major role in maintaining a normal rate of oxygen utilization when intestinal oxygen delivery is reduced by lowering the hematocrit. [54]

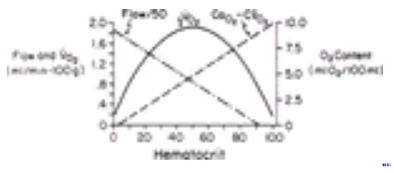

FIGURE 22-5. Effect of hematocrit on canine intestinal blood flow, oxygen consumption ($\dot{V}_{O_2}$), arterial oxygen content, and arteriovenous oxygen difference ($Ca_{O_2} - Cv_{O_2}$). (From ref. [15].)

Venous Pressure Elevation Venous pressure elevation has proved to be a useful perturbation for determining whether metabolic or myogenic mechanisms are involved in local vasoregulation. The metabolic hypothesis predicts that acute venous hypertension causes vasodilation and increased capillary density as a result of reduced blood flow and vasodilator accumulation. According to the myogenic hypothesis, vascular resistance increases and capillary density decreases during venous pressure elevation because of a rise in intravascular (intramural) pressure at the arteriolar and precapillary sphincter levels (Fig. 22-6). [49]

FIGURE 22-6. Metabolic and myogenic theories of intestinal blood flow regulation. (From ref. [49].)

Studies of the stomach, small intestine, and colon in adult animals indicate that vascular resistance rises in response to venous pressure elevation, [12], [13], [19], [25], [40], [41] and [42], [54], [55], [56], [57], [58], [59] and [60] findings consistent with a myogenic mechanism. In newborn animals, the intestinal vasculature dilates, rather than constricts, in response to venous hypertension, suggesting that metabolic factors are dominant in the hypermetabolic neonatal intestine. [24] Acute venous hypertension in the adult intestine elicits vasoconstriction as a result of rising in precapillary (arteriolar) resistance while postcapillary resistance falls. [61] Capillary exchange capacity increases in the stomach and the colon but decreases in the small intestine during venous hypertension. [49] These observations are consistent with the view that metabolic factors exert a greater influence on precapillary sphincters in the stomach and colon, whereas myogenic factors dominate in the small intestine. Despite the intense capillary derecruitment initiated by venous hypertension in the small intestine, oxygen extraction increases disproportionately to the reduced blood flow, and consequently, oxygen consumption rises. [56] The elevated intestinal oxygen utilization during venous hypertension has been attributed to increased villus motility. [62] There are conflicting reports regarding the influence of enhanced oxidative metabolism on the vascular responses to elevations in venous pressure. Some investigators have observed that increased oxygen demand significantly reduces or abolishes the rise in vascular resistance, [14], [42] whereas others have noted an exaggerated resistance response to venous pressure elevation. [63] Researchers uniformly agree that acute venous hypertension alters the distribution of blood flow within the bowel wall. [19], [64] As venous pressure is elevated, the percentage of total blood flow directed to the mucosa and submucosa is reduced, whereas the muscularis receives a larger fraction of the total blood flow. These observations indicate that the constriction of arteriolar and precapillary sphincter smooth muscles elicited by venous hypertension takes place in the mucosal and submucosal layers, and that the vasculature of the muscularis dilates in response to venous hypertension. Whereas exposure of the splanchnic circulation to an acute elevation in portal venous pressure is likely to elicit a myogenically mediated constriction of splanchnic arterioles, chronic portal hypertension tends to dilate the splanchnic vasculature. Furthermore, chronic portal hypertension has a significant impact on other regional vascular beds and on systemic hemodynamics. Blood flow to the gastrointestinal tract, kidneys, and skeletal muscle is significantly elevated. This presumably results from an increase in circulating vasodilators (e.g., glucagon) and a decrease in vascular sensitivity to vasoconstrictors (e.g., norepinephrine). The widespread dilation of arterioles results in a reduction of peripheral vascular resistance and a corresponding reduction of arterial blood pressure. In addition, cardiac output is elevated as a consequence of the increased venous return associated with the splanchnic and peripheral vasodilation. The elevated portal pressure results in the opening of portosystemic shunts to divert portal blood from the liver and reduce portal pressure. These shunts generally run along the esophagus (i.e., esophageal varices). The increase in portal pressure impairs venous drainage from the spleen into the portal vein, resulting in the accumulation of blood within, and distention of, the spleen (i.e., splenomegaly). [65], [66] and [67] Organ blood flow is determined by the arterial-venous pressure gradient and vascular resistance. It follows then that portal pressure is determined by portal venous inflow and portal venous resistance. The relationship between portal venous flow and portal pressure at a normal portal vascular resistance is depicted in Figure 22.7 as a solid line. In this instance, an increase in portal venous flow will produce a proportional increase in portal pressure (point A to point B; flow-induced portal hypertension). When portal vascular resistance is increased, the relationship between portal pressure and portal venous flow is shifted upward and to the left, as depicted by the dashed line. At any given portal venous inflow, an increased portal vascular resistance will result in an increase in portal pressure (point A to point C; resistance-induced portal hypertension). Portal pressure can be further increased by a concomitant increase in portal venous flow and portal vascular resistance (point A to point D; flow- and resistance-induced portal hypertension). Indeed, the latter situation appears to reflect the vascular changes that account for the elevated portal pressure observed in some experimental models of chronic portal hypertension, and it is likely to account for the portal hypertension associated with some forms of liver disease. With a portal vascular resistance that is 40% higher in the portal hypertensive than in the control state, it is predicted that increased portal inflow and increased portal vascular resistance account for 40% and 60% of the increase in portal pressure, respectively. [68]

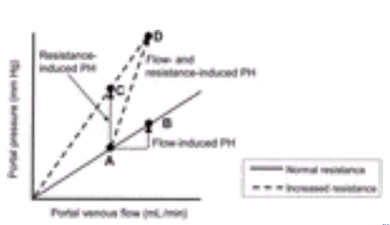

FIGURE 22-7. Hypothetical relationship between portal pressure and portal venous flow in the presence of a normal (*solid line*) or elevated (*broken line*) portal vascular resistance. Even when portal vascular resistance is normal, an increased portal blood flow (from point A to point B) can result in an increased portal pressure—that is, portal hypertension. An elevation in portal vascular resistance in the absence of increased portal blood flow (point A to point C) can also lead to portal hypertension. When both portal blood flow and vascular resistance are increased (point A to point D), the increase in portal pressure is more substantial. (From ref. [68].)

The portal hypertensive state leads to the development of collaterals (mostly along the esophagus; esophageal varices) to shunt blood from the congested portal vein, around the liver, to the systemic circulation (i.e., portosystemic shunting). Because a large proportion of portal venous blood bypasses the liver as a result portosystemic shunting, the hepatic degradation of different compounds, including circulating vasodilators, such as glucagon, is reduced. The diminished catabolism of circulating vasodilators increases their concentration in the plasma, allowing these agents to relax arteriolar vascular smooth muscle and reduce splanchnic vascular resistance. Another important action of some of the vasodilators that accumulate in chronic portal hypertension (e.g., glucagon) is to reduce the sensitivity of the splanchnic arterioles to vasoconstrictors such as norepinephrine, vasopressin, and angiotensin. The net result of the direct and indirect actions of the accumulated circulating vasodilators is increased splanchnic blood flow, which serves to perpetuate the portal hypertensive state. [65], [66] and [67]

Postprandial Hyperemia The term *postprandial hyperemia* is used to describe the increase in blood flow that occurs in response to a meal. The anticipatory-ingestion phase of digestion is characterized by transient increases in heart rate, cardiac output, and aortic pressure; however, gastrointestinal blood flow is either unchanged or slightly increased. [69], [70], [71], [72] and [73] These transient hemodynamic responses appear to be mediated by activation of the sympathetic nervous system, because they can be attenuated by adrenergic blocking agents. [71] In conscious animals, blood flow to the stomach and proximal bowel increases 30 to 90

minutes after ingestion of a meal. [69], [70], [71] and [72] Blood flow to the ileum increases 45 to 120 minutes postprandially, whereas colonic blood flow generally does not increase. [74], [75] and [76] Transient decreases in distal colon blood flow have been observed 30 minutes after a meal, a response attributed to tonic contractions produced by the gastrocolic reflex. [76] Blood flow in the superior mesenteric artery of conscious animals typically increases by 25% to 130% after ingestion of a meal. [69], [70] and [71], [77] The splanchnic vasodilation may last for 4 to 7 hours, depending on the nature and quantity of the meal. [78], [79] A smaller increase (10%–60%) in blood flow is observed in isolated bowel segments in adult animals after intralumenal placement of digested food or nutrient solutions. In similar preparations of isolated bowel segments in developing piglets, the response after placement of intralumenal nutrients is age-dependent. Total intestinal blood flow to the distal jejunoileum of 6- to 8-hour-old piglets that have never nursed does not increase after placement of lumenal nutrients, whereas that of all older age groups does increase with feeding. [80] Mucosal and submucosal blood flow does increase postprandially, but at the expense of flow to the muscularis and serosa. It remains unclear whether postprandial hyperemia is confined to those bowel segments that are exposed to chyme. In general, placement of nutrient solutions into one of two isolated segments in anesthetized animals increases blood flow to the segment containing the nutrient while not affecting blood flow to the adjacent segment. [81], [82], [83], [84] and [85] In conscious animals, however, ingestion of a meal produces hyperemia in distal bowel segments that are not yet exposed to chyme, suggesting that the hyperemia is a diffuse phenomenon. [23] The intramural responses to nutrient absorption are generally confined to the mucosal layer. [76], [84], [85] and [86] In some instances, blood flow to the external muscle layer decreases after intralumenal placement of nutrients. [80], [87] Studies in the rat small intestine suggest that postprandial hyperemia in this species occurs uniformly in all layers of the bowel wall. [88] Considerable effort has been devoted to defining the lumenal stimuli responsible for postprandial hyperemia. Although mechanical stimulation of the mucosa elicits hyperemia, chyme per se does not appear to produce the degree of mechanical stimulation necessary to increase intestinal blood flow. Similarly, lumenal placement of undigested food does not elicit hyperemia, whereas digested food significantly increases blood flow. [82] The latter observation indicates that hydrolytic products of food digestion initiate the hyperemia. The rise in lumenal osmolality that often accompanies a meal has received some attention, particularly in view of the fact that the intestinal vasculature dilates significantly in response to an increase in plasma osmolality. [89] Normally, the osmolality of intestinal chyme varies between 220 and 320 mOsm/kg, yet lumenal osmolalities in excess of 1500 mOsm/kg are needed to increase gut blood flow. [81], [90] A similar argument has been advanced to negate the theory of a role for pH changes in postprandial hyperemia in the jejunum and ileum; gut blood flow increases only when lumenal pH falls below 2.5. [91] Bile appears to play an important role in postprandial intestinal hyperemia. Ten percent gallbladder bile, the steady-state concentration in proximal bowel in the early postprandial period, does not increase jejunal blood flow, yet it appears to render glucose and long-chain fatty acids vasoactive. [92] Thirty-three percent gallbladder bile renders both short-chain fatty acids (e.g., caproic acid) and amino acids vasoactive and further enhances glucose-induced hyperemia. Although intralumenal placement of endogenous or synthetic bile does not have a direct vasoactive effect in the jejunum, bile more than doubles blood flow in the ileum. [82], [93] Bile acids are largely responsible for bile-induced hyperemia, an assertion supported by the observation that cholestyramine abolishes the vasodilator effects of endogenous bile on ileal blood flow. [93] Ingestion of protein-rich meals in humans and gastric placement of protein in conscious rats produces marked increases in splanchnic blood flow. In isolated loops of proximal small bowel, a protein-rich diet (64%) increases blood flow by the same extent as a carbohydrate-rich diet (68%). [94] Although hydrolyzed proteins are well-known to induce postprandial hyperemia, the specific hydrolytic products of protein digestion that mediate the response remain unknown. When postprandial concentrations of 16 different amino acids and three peptides are placed in the bowel lumen, blood flow fails to increase. [82] It has been suggested that fragments cleaved off protein molecules during hydrolysis may possess amino acid sequences similar to those found in vasoactive regulatory peptides normally produced in the intestinal mucosa. [95] After a meal, the lumenal concentration of glucose fluctuates between 28 and 222 mM. At these concentrations, glucose usually produces only a slight hyperemia (5%–10%) in dog and cat small intestine. [81], [82], [96], [97] An 18% to 21% increase in blood flow is produced by glucose in rat small bowel. [98] Glucose analogs have been used to define the contribution of absorptive and oxidative processes in carbohydrate-induced hyperemia. [99] Analogs of glucose that are neither transported nor metabolized (e.g., 2-deoxyglucose) do not increase blood flow, but 3-0 methylglucose, which is transported but not metabolized, produces a hyperemia that is about one-third that produced by glucose. Solubilized long-chain fatty acids appear to be the most potent lumenal stimulus of postprandial intestinal hyperemia. Oleic acid (10–20 mM) solubilized in 10% gallbladder bile produces a 20% to 60% increase in intestinal blood flow. [81], [92], [100] Recent evidence suggests that this increase in blood flow may be a response to epithelial cell injury. [101] Relatively little is known about the vascular response of the gut to the lumenal placement or ingestion of other dietary lipids. Short-chain fatty acids (e.g., caproic acid) do not alter blood flow, even in the presence of 10% gallbladder bile. Although lipids produce the greatest degree of intestinal hyperemia, the vascular responses elicited by protein and carbohydrate are not insignificant in that the three major dietary components of food—that is, fats, proteins, and carbohydrates–appear to act synergistically on blood flow when placed in the bowel lumen (Fig. 22-8). [102]

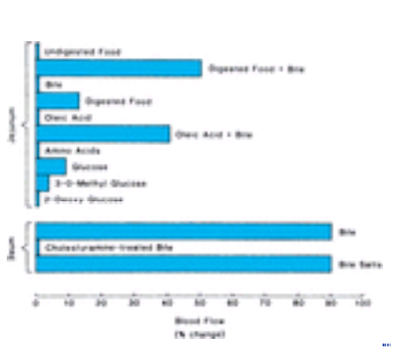

FIGURE 22-8. Effects of intralumenal placement of various constituents of chyme on intestinal blood flow. (From ref. [102].)

Modulators of Intrinsic Vasoregulation A number of chemical and physical factors have been proposed to explain intrinsic vasoregulatory phenomena in the gastrointestinal tract. These factors generally fall into one of three major categories: myogenic factors; chemicals directly linked to oxidative metabolism; and vasoactive peptides, hormones, and autocoids. The myogenic theory has been invoked to explain both pressure-flow autoregulation and the responses to venous pressure elevation in the intestine and stomach. The myogenic theory is based on the assumption that vascular wall tension is a controlled variable. According to this concept, arteriolar tension receptors modulate vascular smooth muscle tone in response to changes in microvascular transmural pressure. In accordance with the Laplace law, resistance vessels should dilate when vascular transmural pressure is decreased and constrict when it is increased; therefore, the myogenic theory predicts that vascular resistance should fall when arterial pressure is reduced. Similarly, this mechanism predicts the rise in intestinal vascular resistance that occurs during acute venous pressure elevation (Fig. 22-9). [19]

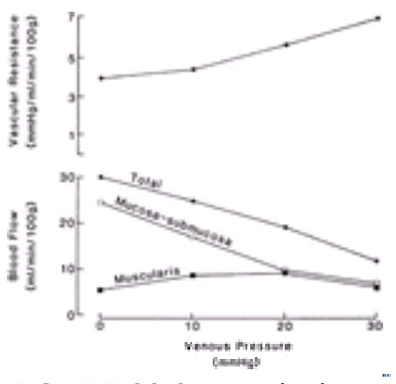

FIGURE 22-9. Intestinal vascular responses to acute venous hypertension in cats. The increase in vascular resistance and decrease in blood flow during acute venous pressure elevation are consistent with the myogenic theory of intrinsic vasoregulation. (Data from ref. [19].)

Because the intensity of all intrinsic vasoregulatory phenomena described in the gastrointestinal tract is significantly influenced by the oxidative requirements of the tissue, metabolic factors have received much attention. According to the metabolic theory of blood flow regulation, vascular resistance and precapillary sphincter tone are linked to the metabolic status of the tissue. Any condition that reduces oxygen delivery, increases oxygen demand, or both, will lead to a reduction in tissue oxygen tension and an accumulation of vasodilator metabolites in the immediate perivascular space. These changes cause relaxation of arteriolar and precapillary sphincter smooth muscle. The resulting increase in blood flow and capillary recruitment stabilize capillary oxygen tension (PO_2), increase the surface area for oxygen exchange, and decrease the capillary-to-cell diffusion distance. The ultimate effect of these changes is to maintain cell PO_2 above the critical level at which oxygen availability limits energy metabolism. The metabolic theory has been invoked to explain the vascular responses during pressure-flow autoregulation, reactive hyperemia, arterial hypoxemia, postprandial hyperemia, and venous pressure elevation. Both adenosine and tissue PO_2 have been proposed as mediators of metabolic vasoregulatory responses in the gastrointestinal tract. Adenosine is a powerful vasodilator when infused into the splanchnic circulation. [103] Adenosine accumulation in venous blood has been demonstrated during reactive and postprandial hyperemia in the small bowel. The role of adenosine in local vasoregulation

has been assessed with the use of substances such as theophylline, a competitive antagonist; the enzyme adenosine deaminase, which converts adenosine to inosine; and dipyridamole, an inhibitor of adenosine reuptake. These agents have been shown to attenuate or completely abolish the vasodilation associated with reductions in arterial pressure, release of an arterial occlusion, reactive hyperemia, acute and chronic hypoxia, and absorption of nutrients. [104], [105], [106], [107] and [108] For example, adenosine deaminase completely prevents absorptive hyperemia in rat and dog small intestine. [104], [107] Similarly, 8-phenyltheophylline abolishes pressure-flow autoregulation and reduces the reactive hyperemic response by 50% in the superior mesenteric artery of anesthetized cats. [109] Considerably less information is available regarding the role of tissue PO_2 in intrinsic vasoregulation. Mucosal PO_2 falls by about 50% during the absorption of glucose in the proximal small bowel, and the hyperemic response is quantitatively and temporally related to the decrease in mucosal PO_2. [88] Taurocholic acid produces a similar decrease (58%) in mucosal PO_2 and a 65% increase in blood flow in the ileum. Cholic acid does not affect either mucosal PO_2 or blood flow when placed in the ileal lumen. A large body of indirect evidence also suggests that the degree of tissue oxygenation influences intestinal vasoregulation. For example, it is well established that at any given oxygen uptake, the magnitude of the postprandial hyperemia is greater if the resting oxygen extraction (i.e., arteriovenous oxygen difference) is high; this indicates that the vascular response is influenced by the prevailing tissue PO_2. [36] Most of the hormones and peptides produced in the gastrointestinal mucosa act as vasodilators when infused into the splanchnic circulation. Because many of these substances are also released into the mucosal interstitium in response to a meal, it has been proposed that peptides and hormones mediate postprandial hyperemia. This hypothesis has received less attention because of studies demonstrating that gastrointestinal peptides and hormones are not vasoactive when infused into arterial blood to reproduce postprandial concentrations. Cholecystokinin, secretin, gastrin, and neurotensin do not produce vasodilation when infused, either alone or in combination, into the arterial supply of the proximal bowel at rates that reproduce postprandial hormone levels. [110], [111] In the distal ileum, however, neurotensin significantly increases blood flow at postprandial concentrations. [108] Although vascular infusion studies tend to argue against a role for gastrointestinal hormones and peptides in postprandial hyperemia, these experiments do not exclude the possibility that the peptides induce vasodilation by a paracrine action. Immunoneutralization techniques have been used to evaluate this concept. [112] Intravenous administration of antiserum directed against vasoactive intestinal polypeptide (VIP) produces a dose-related suppression of the absorptive hyperemia induced by the lumenal placement of bile-oleic acid. Antisera against cholecystokinin or substance P have no effect on absorptive hyperemia. These results indicate that VIP, which is released from mucosal neurons, mediates at least a fraction of postprandial hyperemia by acting as a paracrine agent. A variety of endogenous autocoids also have been implicated as local paracrine mediators in the regulation of intestinal blood flow. These substances are diverse in regard to structure and overall biologic activity, but they appear to have at least one property in common: they cause vasodilation of the gastrointestinal vasculature. Three members of this diversified group have been studied in relation to mediation of postprandial hyperemia: serotonin, histamine, and prostaglandins. Feeding or acid perfusion of the duodenum elevates serotonin levels in portal blood. [113] However, a serotonin antagonist, methysergide, does not reduce the magnitude of postprandial hyperemia. Intrajejunal placement of a mixed meal increases blood flow by 30%, a response that is attenuated by pretreatment with a histamine H_1 receptor antagonist, tripelennamine, but not with metiamide, a histamine H_2 receptor antagonist. [114] Because tripelennamine also blocks the food-induced increase in oxygen uptake, it remains unclear whether histamine directly dilates the vasculature during postprandial hyperemia or whether the blunted response to histamine H_1 receptor blockade reflects a reduction in oxidative metabolism. Studies of rat and dog jejunum suggest that endogenous prostaglandins may also play a role in postprandial hyperemia. [115], [116], [117] and [118] The cyclooxygenase inhibitors indomethacin and mefenamic acid greatly enhance food-mediated increases in blood flow and oxygen uptake, whereas arachidonic acid attenuates these responses. [115], [116] and [117] Jejunal production of prostaglandins I_2, E_2, and F_2 and thromboxane A_2 increases during nutrient absorption. Addition of arachidonate to food attenuates the release of the former two and enhances the release of the latter two substances, thereby attenuating food-induced jejunal hyperemia. [118] The available data suggest that prostaglandins are produced during the postprandial state, and these cyclooxygenase products tend to limit the increase in blood flow, either by inhibiting oxidative metabolism or by a direct vasoconstrictor action. Attention has been devoted to the potential role of endothelial cell–derived factors in the regulation of gastrointestinal blood flow. Vascular endothelial cells are known to produce agents that act on the underlying smooth muscle to promote protein phosphorylation, leading in turn to a decrease in smooth muscle tone. [119] Nitric oxide (NO) is a potent endothelial cell–derived vasodilator that is synthesized from L-arginine by the enzyme NO synthase. [120], [121] NO produced by endothelial cells diffuses to adjacent vascular smooth muscle, where it binds to soluble guanylate cyclase, which in turn increases the production of cGMP, with subsequent relaxation of smooth muscle (Fig. 22-10). [122] Endothelial cells contain both a constitutive and an inducible form of NO synthase. The constitutive form of the enzyme is Ca^{2+}/calmodulin– and NADPH-dependent, [123] and it produces NO in response to receptor (e.g., acetylcholine) or physical (e.g., shear stress) stimulation. [124], [125] The inducible form of NO synthase is increased in response to certain stimuli that act directly on endothelial cells (e.g., cytokines). The inducible form of the enzyme can produce larger quantities and a more sustained release of NO. The requirement of NO synthase for L-arginine appears to be absolute, in as much as L-arginine or analogs of L-arginine (N^G-monomethyl-L-arginine [L-NMMA], N^G-nitro-L-arginine-methyl ester [L-NAME]) do not act as substrates. Consequently, L-arginine analogs have been used in experimental animals to inhibit NO synthase. [123], [126] Inhibition of NO synthase has been used to demonstrate that endogenous NO mediates the intestinal hyperemic response to intralumenal bile-oleate, because NO synthase inhibition reduces the magnitude of the hyperemic response. [127]

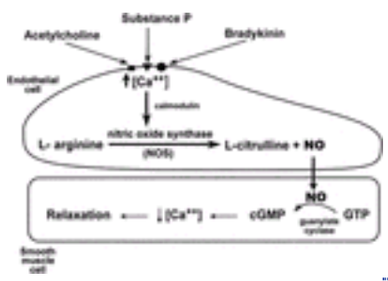

FIGURE 22-10. Mechanism of nitric oxide (NO)–mediated relaxation of vascular smooth muscle. NO generation by the endothelial cell enzyme NO synthase is induced by certain stimuli, such as acetylcholine, substance P, and shear stress. The endothelial cell–derived NO produced by these stimuli readily diffuses into the underlying smooth muscle cells, where it binds to and consequently activates soluble guanylate cyclase (sGC). The cGMP produced by sGC promotes smooth muscle relaxation by lowering intracellular calcium.

The mechanism for NO mediation of intrinsic regulation of gastrointestinal blood flow has not been well defined. In vitro studies using monolayers of cultured endothelial cells indicate that the rate of production of NO is influenced by physical forces such as shear rate and transmural pressure. [124], [125] The production of NO by endothelial cells appears to be directly coupled to shear rate, yet it is inversely related to transmural pressure. The relation between NO production and shear rate does not favor a role for NO in the gastrointestinal vasodilation associated with reductions in local arterial pressure (i.e., pressure-flow autoregulation); however, the increased NO production that would be elicited by a decline in endothelial cell transmural pressure may provide a chemical basis for the phenomenon of pressure-flow autoregulation. Nonetheless, it has been demonstrated that the autoregulatory capacity in the superior mesenteric artery is attenuated by NO, favoring a more profound influence of shear rate than transmural pressure on NO production in the vasculature of the superior mesenteric artery. [128] The contribution of NO to basal vascular tone in the gastrointestinal tract has been demonstrated with analogs of L-arginine. Inhibition of NO synthase reduces resting blood flow in the gastrointestinal tract. [129] The endothelium-dependent relaxation of gastric vascular smooth muscle by agents such as acetylcholine and bradykinin is also attenuated by inhibitors of NO synthesis. [126] In fetal [130] and developing [131] intestine, inhibition of NO synthase demonstrates that NO is an important regulator of basal intestinal vascular tone. The NO-cGMP axis in the regulation of intestinal vascular resistance is more important in 3-day-old than in 35-day-old swine. [131] A growing body of evidence indicates that NO mediates the gastrointestinal hyperemia associated with conditions as diverse as portal hypertension and central vagal stimulation. [132], [133] and [134] Inactivation of the NO that is basally produced by endothelial cells may also explain the increased vascular resistance that is observed after reperfusion of ischemic tissues. Superoxide rapidly and efficiently inactivates NO [135]; consequently, conditions associated with an accelerated production of superoxide [136] (e.g., ischemia and reperfusion) may abolish endothelium-dependent vascular tone and reduce blood flow. Interpretation of the blood flow responses to NO synthase inhibitors is complicated by the actions of these agents on circulating blood cells. The NO produced by endothelial cells appears to play an important role in preventing the intravascular adhesion and aggregation of platelets and leukocytes. [137], [138] and [139] Inhibitors of NO synthesis promote the adhesion and aggregation of platelets and leukocytes within the microcirculation. [139] A potential consequence of these events is increased vascular resistance secondary to obstruction of microvessels with platelet-leukocyte aggregates.

Extrinsic Control

Blood flow within the gastrointestinal tract is also influenced by extrinsic neurohumoral factors. Because the splanchnic organs receive about 25% of the cardiac output and contain about 25% of the total blood volume at rest, neurohumoral control of the splanchnic vascular bed can be an important component in the overall reflex control of the circulation, especially during periods of stress, such as exercise and shock. [140]

Neural Control Neural control of intestinal blood flow occurs predominantly by way of sympathetic noradrenergic nerves. [141] Extrinsic cholinergic nerves from the vagus do not supply the intestinal vasculature, [142], [143] but the myriad extravascular actions of parasympathetic stimulation in the gastrointestinal tract may indirectly affect blood flow. A local, intramural vasodilator nervous pathway also appears to exist within the intestine and causes an increase in intestinal blood flow after local electrical or mechanical stimulation of the mucosa. [144], [145] This response is blocked by tetrodotoxin or lidocaine, but not by autonomic denervation.

Autoregulatory Escape Autoregulatory escape (Fig. 22-11) [28] is a phenomenon in which stimulation of sympathetic nerves or intraarterial infusion of norepinephrine produces an initial intense vasoconstriction and decrease in blood flow, followed by a return (i.e., escape) of blood flow toward baseline levels despite continued nerve stimulation or norepinephrine infusion. Discontinuation of stimulation or infusion results in poststimulatory hyperemia.

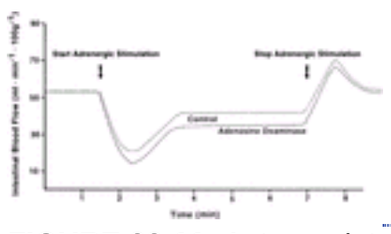

FIGURE 22-11. Autoregulatory escape from adrenergic stimulation in autoperfused piglet small intestine. Adenosine de-aminase pretreatment reduced the steady-state escape response to norepinephrine infusion, whereas pretreatment with chlorpheniramine, a histamine H_1 blocker, had no effect. (Data from ref. [28].)

Autoregulatory escape in the intestine was first described by Folkow and colleagues in 1964, [146] but the mechanism of the phenomenon is still not completely understood. Autoregulatory escape occurs only in arteriolar smooth muscle, not in venous smooth muscle. [146] It is unaltered by ß-receptor blockade [147] and administration of atropine. [148] In growing animals, there are conflicting reports regarding the ability of the intestine to escape from nerve stimulation. In one study, [149] the intestine of swine younger than 2 weeks of age did not undergo autoregulatory escape, whereas in another study, no difference in intestinal autoregulatory escape was observed between 3-day-old and 35-day-old piglets. [150] Three mechanisms commonly invoked to explain autoregulatory escape are redistribution of blood flow from the mucosa to the submucosa, adaptation of adrenergic receptors to continued nerve stimulation, and accumulation of vasodilator metabolites. The first possibility of blood flow redistribution has not been substantiated, as demonstrated by in vivo microscopic studies in which the vessels that initially constrict subsequently relax during the steady-state escape phase. [151] Adaptation of adrenergic receptors is also unlikely, because infusion of norepinephrine during the steady-state escape phase of sympathetic erve stimulation leads to further vasoconstriction and a second escape. [147] The most popular theory used to explain autoregulatory escape is that vasodilator metabolites accumulate during the initial vasoconstrictor phase, leading to arteriolar dilation and consequent restoration of blood flow toward normal. [147], [152], [153] and [154] The metabolic theory of intestinal vasoregulation states that tissue metabolism and arteriolar smooth muscle constitute a local control system that provides the necessary coupling between blood flow and tissue nutritional requirements. Any condition causing an imbalance between oxygen supply and demand will produce an outpouring of metabolites into the interstitial fluid. The metabolites then diffuse to the arterioles and precapillary sphincters to cause vasodilation and capillary recruitment. The increased blood flow or oxygen extraction restores oxygen supply to a level compatible with tissue oxygen demand. [155] Consistent with this theory is experimental evidence that the propensity for blood flow to escape from sympathetic vasoconstriction is significantly greater in the metabolically active mucosa than in the muscularis. [156] Adenosine appears to play at least a partial role in autoregulatory escape (see Fig. 22-11) [28]; however, a role for histamine [28], [148] and prostaglandins [148] has not been substantiated. Evidence also implicates a role for vasodilator peptidergic neurons in effecting autoregulatory escape after sympathetic nerve stimulation but not during infusion of norepinephrine. [157], [158] One neurotransmitter substance produced by these fibers is VIP. [112]

Circulating Vasoactive Substances Circulating vasoactive substances that affect gastrointestinal blood flow include adrenergic agents, vasopressin, and angiotensin. Norepinephrine, a predominantly a-adrenergic receptor stimulant, causes intestinal vasoconstriction, a decrease in capillary density, and a reduction in oxygen uptake. [154], [159], [160] With continuous intraarterial infusion, the intense initial vasoconstriction is followed by the return of blood flow toward control levels despite continued norepinephrine infusion (see previous section " Autoregulatory Escape"). Epinephrine can cause either a-receptor–mediated vasoconstriction at high doses or ß-receptor–mediated vasodilation at low doses, as well as a variable response in oxygen uptake. [154], [161], [162] Both vasopressin and angiotensin II are potent physiological vasoconstrictors that reduce blood flow and increase vascular resistance in all gastrointestinal organs. These agents cause generalized vasoconstriction, with a disproportionate selective reduction in mesenteric blood flow at doses that have been measured in pathophysiological states of hypotension. [163] Vasopressin causes a decrease in capillary density and a reduction in intestinal oxygen uptake, whereas angiotensin II reduces or does not affect [164] splanchnic oxygen uptake. In normal rat intestine, a-adrenergic and vasopressin activity account for most extrinsic vasoconstrictor tone, whereas vasopressin and angiotensin II account for most extrinsic vasoconstrictor tone in portal hypertensive rat intestine. [165] Renin-angiotensin and vasopressin systems are also involved in the intestinal vasoconstrictor response to hemorrhage and hypovolemia, and significant attenuation of this increase in vascular resistance occurs only when both systems are blocked simultaneously, [166], [167] even in the presence of an intact sympathetic system. Furthermore, angiotensin does not affect the mesenteric circulation indirectly through activation of the sympathetic nervous system or by promoting vasopressin release; it plays a physiologically important, direct role in the control of mesenteric blood flow after volume depletion. [168]

Oxygen Uptake–Blood Flow Relation: Functional Implications

Considerable attention has been devoted to the interaction between gastrointestinal blood flow and oxygen uptake, and the relevance of this interaction to mucosal function and integrity. Figure 22-12 depicts the observed relation between intestinal blood flow and oxygen uptake when blood flow is altered with a pump or by graded reductions in perfusion pressure. [20] Oxygen uptake remains virtually constant over a wide range of blood flow levels (i.e., it is independent of blood flow), and it is compromised only when blood flow reaches a critically low level. Below this level, oxygen uptake is dependent on blood flow. Resting blood flow in the small intestine, stomach, and colon is usually greater than the critical blood flow at which oxygen uptake is dependent on blood flow. [13], [18], [169], [210]

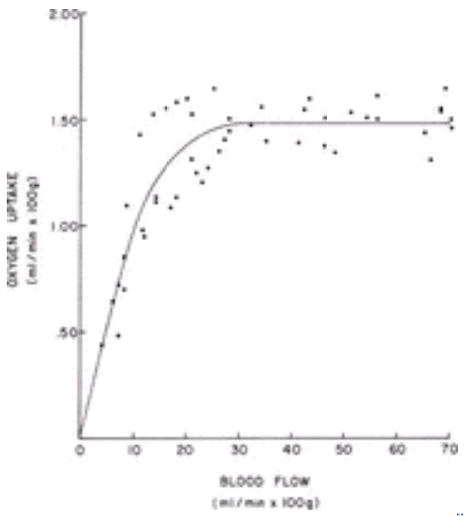

FIGURE 22-12. Relation between intestinal blood flow and oxygen uptake in feline jejunoileum when blood flow is altered with a pump or by graded reductions in perfusion pressure. (From ref. [20].)

The reduction in oxygen uptake that occurs when blood flow falls below a critical level can be explained in terms of the normal relation between mitochondrial oxygen consumption and cell PO_2. This relation predicts that oxygen uptake remains constant over a wide range of cell PO_2 levels, and uptake is reduced only when cell PO_2 falls to a low level, the critical PO_2. The resting cell PO_2 is normally well above the critical PO_2. Evidence indicates that graded reductions in blood flow produce concomitant reductions in cell PO_2 without altering oxygen uptake in the stomach. [164] At very low rates of blood flow, however, the rate of oxygen diffusion to the cells is so low that the intracellular PO_2 falls below the level required to maintain normal oxidative metabolism. The reduction in oxygen uptake observed at low blood flow

may simply reflect a depression in oxidative metabolism caused by the limited oxygen availability.

Many conditions alter the relation between oxygen uptake and blood flow such that oxygen uptake is dependent on blood flow even when it is increased above normal. These include lumenal distention, [170] alterations in hematocrit, [171] and devascularization. [40] All these conditions are thought to reduce tissue oxygenation severely in discrete or generalized regions of the stomach or gut. Lumenal distention and devascularization reduce tissue oxygenation by compromising blood flow, whereas a low hematocrit limits oxygen delivery, even at high blood flows. Normal physiological conditions can also influence the relation between oxygen uptake and blood flow. Stimulation or inhibition of oxidative metabolism will shift the plateau of the blood flow–oxygen uptake curve upward or downward, respectively (Fig. 22-13). [164] Stimulation of intestinal motility or enhancement of active transport raises the plateau of the blood flow–oxygen uptake curve. [172] Conversely, decreasing the temperature of isolated bowel segments lowers the plateau.

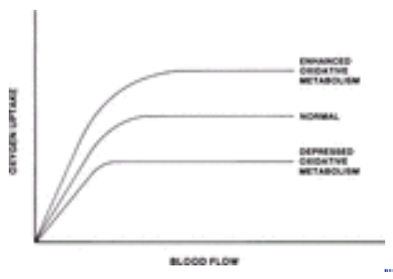

FIGURE 22-13. Relation between blood flow and oxygen uptake during stimulation or inhibition of oxidative metabolism. (From ref. [164].)

Another important aspect of the blood flow–oxygen uptake relation in the small bowel is that it will predict the influence of blood flow reductions on oxygen-requiring processes, such as absorption and secretion. For example, it has been shown that the reductions in glucose absorption produced by graded decrements in blood flow parallel the decline in oxygen uptake, [173] suggesting that oxygen availability limits solute transport when cell PO_2 falls.

Many vasodilators tend to increase intestinal oxygen uptake, irrespective of their effects on oxidative metabolism. These observations have been attributed to the use of preparations in which oxygen uptake is dependent on blood flow. If vasodilators are infused into these preparations, the increase in oxygen uptake would be expected to parallel the increase in blood flow. Vasodilators will not alter oxygen uptake in preparations in which oxygen uptake is independent of blood flow unless an effect is exerted on the oxidative metabolism. In general, vasoconstrictors decrease oxygen uptake in preparations exhibiting either a normal or abnormal relation between blood flow and oxygen uptake, unless the agent enhances oxidative metabolism. Vasoconstrictors that do not affect oxidative metabolism would be expected to decrease oxygen uptake by simply moving down the normal blood flow–oxygen uptake curve.

Another important determinant of the rate of oxygen exchange across capillaries is the effective capillary density. The number of capillaries perfused at any given time determines the flux of oxygen out of the capillary and into the tissue through an effect on diffusion parameters (i.e., surface area available for exchange and capillary-to-cell diffusion distances). The expected influence of capillary density on the relation between blood flow and oxygen uptake is shown in Figure 22-14. If capillary density within an organ is increased, the minimum blood flow required to attain the plateau of the blood flow–oxygen uptake curve will be lower (i.e., the curve will be shifted to the left) as a result of an increased capillary surface area and a reduced capillary-to-cell diffusion distance. [164]

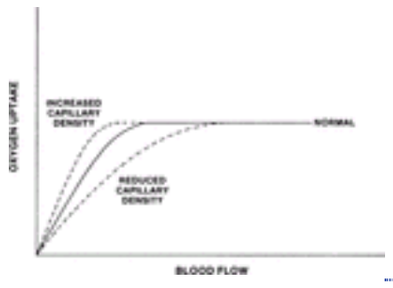

FIGURE 22-14. Relation between oxygen uptake and blood flow under conditions of increased or reduced perfused capillary density. (From ref. [164].)

The relation between blood flow and oxygen uptake has also been useful in explaining some of the reported observations regarding the influence of blood flow on gastric acid secretion. [174] The production of gastric acid is an energy-consuming process that results in an increase in gastric oxygen uptake. The consistent finding that acid secretion and oxygen consumption are highly correlated raises the question of whether acid secretion in the stomach is dependent on blood flow. The answer to this question largely depends on the surgical preparation used. In some preparations, a number of vessels supplying the stomach are occluded, and the end result is flow-dependent oxygen uptake. In this situation, an increase or decrease in blood flow is associated with a corresponding change in acid secretion. In normally perfused preparations, however, acid secretion and blood flow exhibit a relation similar to that observed between oxygen consumption and blood flow; acid secretion is dependent on blood flow at low blood flow rates and is independent at higher flow rates. This explains why vasoconstrictors tend to reduce gastric acid output and why some vasodilators (i.e., acid secretagogues) increase acid output whereas others (i.e., nonsecretagogues) do not. The controversy regarding the relation between gastric acid output and blood flow cannot be resolved entirely on the basis of tissue oxygenation. Most of the reports that demonstrate a relation between blood flow and acid secretion have relied on the aminopyrine clearance technique for measurement of blood flow. It is now well recognized that aminopyrine clearance reflects acid output by the stomach more accurately than it does blood flow to the stomach.

Recognition is increasing of the importance of determining both intestinal blood flow and oxygen uptake in developmental studies. Evaluation of age-related differences in intestinal blood flow has received significant attention because of a disease, neonatal necrotizing enterocolitis (NEC), and the belief that mesenteric ischemia plays a role in its pathogenesis. The capacity for regulation of blood flow does not provide information about the ability of the intestine for oxygen extraction when blood flow falls; therefore, conclusions based on blood flow data alone regarding the susceptibility of developing intestine to tissue hypoxia and subsequent mucosal injury may be erroneous. This concept is illustrated in Figure 22-15, in a study of the developmental cardiovascular and oxygenation responses to lumenal nutrients. [80] Blood flow does not increase with feeding in 1-day-old piglet intestine, but oxygen extraction increases so dramatically that oxygen uptake is not different among age groups (1 day–1 month) 30 minutes postprandially.

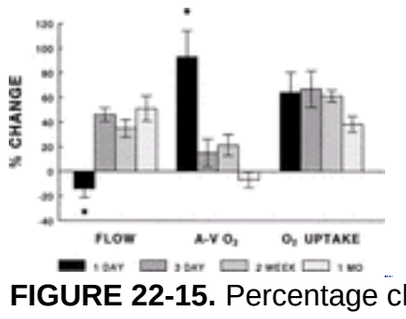

FIGURE 22-15. Percentage change from zero time in intestinal blood flow, arteriovenous oxygen ($A\text{-}V\,O_2$) content difference, and oxygen (O_2) uptake at 30 minutes after lumenal instillation of a predigested and bile acid–solubilized cows' milk–based artificial pig formula. * P <.05 for 1-day-old piglets versus older age groups. (From ref. [80].)

PHYSIOLOGY AND BIOCHEMISTRY OF ISCHEMIA

Ischemic damage to the intestine occurs when splanchnic blood flow falls to a level at which delivery of oxygen and other nutrients is insufficient to maintain oxidative metabolism and hence cell integrity. Blood flow to the gastrointestinal tract may be reduced during generalized nonocclusive ischemia (e.g., circulatory shock and congestive heart failure, especially in patients treated with cardiac glycosides) and in occlusive disorders (e.g., embolism, atherosclerosis, thrombosis) that primarily involve the mesenteric circulation. The mortality of acute mesenteric ischemia in adults has been reported at 70% to 90%, [175] primarily because of the difficulty of making an early diagnosis before bowel infarction occurs. Surgical intervention (e.g., embolectomy, intestinal resection) [176] and local intraarterial infusion of vasodilators (e.g., papaverine) [177] are used to treat acute mesenteric ischemia, but the mortality of this disease continues to be significant. Experimental nonocclusive mesenteric ischemia in dogs has been treated successfully with intravenously administered selective mesenteric vasodilators (e.g., urotensin I, sauvagine, and corticotropin-releasing factor), [178] thereby potentially obviating the risk of an indwelling angiographic catheter, but the use of these drugs in humans remains to be investigated.

Mesenteric ischemia also appears to play a role in NEC, a disease that predominantly affects the ileum and colon of premature infants, with an average 40% mortality, and can result in short-bowel syndrome and intestinal strictures. The pathogenesis of NEC is unknown, but enteral alimentation, infectious agents and immune factors, and mesenteric ischemia and tissue hypoxia have been rumored to be primary initiators of the disease. [179], [180]

Alterations of Intestinal Morphology with Ischemia

The response of the intestine to decreased blood flow can range from no damage to transmural necrosis, [181] and a gradient of sensitivity to ischemic injury has been demonstrated from the villus tips to the muscularis. [181], [182] and [183] Mesenteric ischemia is associated with characteristic mucosal lesions that progress from subepithelial edema within 30 minutes after total vascular occlusion, to loss of epithelial cells along the villus after 1 hour of total occlusion, to total loss of villi after 2 hours of occlusion. [181], [182] Within 30 to 60 minutes after total mesenteric artery occlusion, changes indicative of cellular failure appear, such as mitochondrial vacuolization and decreased oxygen uptake, loss of adenosine triphosphate, and release of lysosomal enzymes. [183]

Changes in Vascular and Mucosal Permeability with Ischemia

Increases in the capillary filtration coefficient and microvascular permeability in the small intestine have been observed after ischemia. [184] The osmotic reflection coefficient of ileal capillaries to plasma proteins decreases after 1 hour of ischemia and reperfusion; this is indicative of increased vascular permeability. Furthermore, the increase in permeability is derived from an increase in the number of large (200 Å) pores; the small-pore (50 Å) population is unaffected. In the intestine, the increase in the capillary filtration coefficient observed after ischemia and reperfusion is not solely a result of increased capillary surface area.

Increases in mucosal permeability induced by ischemia and reperfusion have been estimated based on the clearance of solutes ranging from 700 to 70,000 d. [26], [185] The ischemia/reperfusion–induced increases in mucosal permeability are dependent on both the duration and severity of the ischemic insult (Fig. 22-16). [186] Mucosal permeability increases significantly after 1 to 2 hours of mesenteric artery occlusion in adult animals.

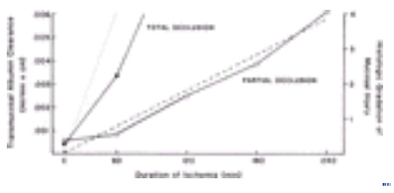

FIGURE 22-16. Comparison of quantitative morphologic data (*dotted* and *broken lines*) and mucosal albumin clearance results (*solid lines*). (From ref. [186] .)

In growing intestine, a similar correlation is observed between the duration of ischemia and the magnitude of the increase in clearance of 358-d ^{51}Cr-EDTA during reperfusion. [187] The extent of reperfusion-induced injury in the absence of lumenal nutrients is similar among groups of young swine ranging in age from 1 day to 1 month, but perfusion of the ileal lumen with cows' milk–based formula causes a significantly larger increase in mucosal permeability in 1-day-old swine than in all older age groups. [187] This increased permeability can be attributed to the lipid component of formula (Fig. 22-17). [188] Furthermore, lumenal perfusion with predigested and bile acid–solubilized premature human infant formula, combined with ischemia and reperfusion of the superior mesenteric artery, leads to an animal model of NEC in 1-day-old piglet intestine. The necrotic injury does not occur unless the lipid fraction of the formula is present, nor does it occur in 1-month-old animals. [189] The chemical composition of fatty acids appears to be the most important determinant of this lipid-associated mucosal injury. [190]

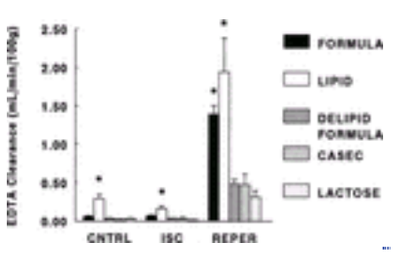

FIGURE 22-17. Comparison of ^{51}Cr-labeled EDTA clearance among nutrient solutions groups in 1-day-old piglet distal jejunoileum during 1 hour each of control (*CNTRL*), ischemia (*ISC*), and reperfusion (*REPER*). The nutrient solutions were all predigested and bile acid–solubilized and included cows' milk–based artificial pig formula, 5% corn oil, delipidated pig formula, 2.3% Casec (Mead Johnson Nutritionals), and 7% lactose. * P <.05 versus other nutrients within a time period. (From ref. [188] .)

Blood Flow, Oxygenation, and Ischemic Injury

Ischemic injury to the intestine occurs when blood flow is reduced to a level at which delivery of oxygen and other nutrients to the tissue is compromised. Although the correlation of tissue PO_2, mucosal blood flow, and mucosal injury has not been investigated, reduction of blood flow to levels that do not affect oxygen uptake are not associated with any evidence of mucosal damage in adult animals. [27] Furthermore, substantial increases in mucosal albumin clearance are not seen until the blood flow falls to levels at which oxygen consumption is reduced by approximately 50% (Fig. 22-18). [27]

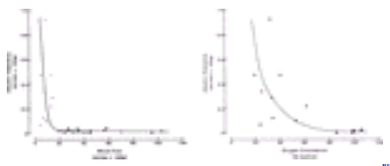

FIGURE 22-18. Relation between blood flow during control (*closed circles*) and ischemia (*open circles*) and mucosal albumin clearance during reperfusion of canine jejunum (**left**). Relation between intestinal oxygen consumption during control and ischemia and mucosal albumin clearance during reperfusion of canine jejunum (**right**). Substantial increases in albumin clearance were not seen unless blood flow was reduced to below 20 mL·min $^{-1}$·100 g $^{-1}$ or oxygen consumption was reduced to approximately one-half that of the control values. (From Bulkley GB, Kvietys PR, Parks DA, et al. Relationship of blood flow and oxygen consumption to ischemic injury in the canine small intestine. Gastroenterology 1985;89:852.)

Neonatal intestine, which has a limited capacity to maintain oxygen uptake during reductions in perfusion pressure, arterial hypoxia, and hemorrhage, and during the combined stresses of hypoxemia and feeding, ischemia/reperfusion, and reductions in perfusion pressure, has been shown to be more vulnerable to mucosal injury induced by ischemic cardiovascular stress than the intestine of older animals. [187], [188] and [189], [191], [192] This suggests, but does not prove, a relation between tissue hypoxia and mucosal injury.

The importance of collateral blood flow in the prevention of intestinal ischemia is well recognized in humans [193], [194] and [195] and adult animals. [21], [196], [197] Intestinal collateral blood flow may occur through anastomotic connections at several levels of vessel branching, including the main arterial trunks (i.e., celiac, superior, and inferior mesenteric arteries), [21], [193] extramural vessels (i.e., arterial arcades, marginal arteries), [197], [198] and intramural vascular plexuses located within the intestinal wall itself. [193], [198] Quantitative studies in adult animals have demonstrated that collateral channels among the major arterial trunks and between adjacent bowel segments both play a role in the prevention of intestinal ischemia. In the adult cat, perfusion through collateral vessels after occlusion of the superior mesenteric artery maintained flow to the small intestine and proximal colon to within 30% to 65% of preocclusion flow. [21] However, the efficiency of collateral perfusion by way of the celiac and inferior mesenteric arteries is substantially lower in dogs after superior mesenteric artery occlusion. [196] In adjacent segments of canine small bowel, collateral vessels maintain blood flow in one segment at approximately 55% of its control level when the artery to that segment is totally occluded. The percentage of collateral flow attributed to extramural vessels is 67%, whereas that attributed to intramural vessels is 33%. [197] In developing piglet intestine, after occlusion of a distal branch of the superior mesenteric artery, total wall and mucosal and submucosal blood flow is reduced by 70% in 1-day-old animals, compared to a 25% decrease in 1-month-old animals. This finding suggests that newborn rather than adult intestine may be at greater risk for ischemic injury because of poorly developed or inefficient collateral blood vessels.

Possible Mechanisms of Injury or Villous Necrosis

Ischemic injury in the intestine appears to be related, either primarily or secondarily, to the effects of tissue hypoxia. In some species, the hypoxic stress induced by ischemia is exacerbated by the presence of a countercurrent exchange mechanism. [7] A role for hypoxia is supported by the observation that intralumenal perfusion with oxygenated saline solution markedly attenuates mucosal injury during ischemia induced by hypotension, [199] whereas perfusion with nitrogenated saline solution does not attenuate injury. Possible mechanisms of mucosal injury induced by tissue hypoxia include depletion of high-energy phosphates necessary to produce protective substances, such as mucus, leading to increased susceptibility to the action of intralumenal proteases [200]; accumulation of histamine, leading to increased microvascular permeability [201]; production of metabolic acidosis, leading to release of lysosomal enzymes and cellular digestion [202]; conversion of xanthine dehydrogenase to xanthine oxidase, an enzyme that can produce cytotoxic oxygen-derived free radicals during reoxygenation [203]; and attraction of circulating granulocytes into the mucosa, or activation of resident leukocytes within the mucosa, with release of neutrophilic proteases and oxidants to initiate or propagate mucosal injury. [204]

It has been demonstrated that changes in the intestinal mucosa induced by circulatory shock lead to an increased vulnerability to the digestive action of trypsin and chymotrypsin. Inhibition of pancreatic proteases by aprotinin [205] or previous ligation of the pancreatic ducts [206] significantly attenuates ischemic mucosal injury. In addition, an intralumenal injection of trypsin exacerbates mucosal injury. [205] The digestive action is caused by enzymes already present along the intestinal wall before shock, because removal of the pancreas has no effect if the animal is subjected to shock immediately after pancreatectomy. [207] Inhibition of pancreatic elastase and bile salts, both of which contribute to the loss of protective brush border glycoproteins, [208], [209] decreases mucosal injury. It has been proposed that if impairment of mesenteric blood flow prevents the steady regeneration of these brush border glycoproteins, then the mucosa becomes accessible to the digestive action of the pancreatic endopeptidases present along the intestinal wall and lumen. [200] It is generally believed, however, that pancreatic proteases are not the primary mediators of ischemic injury in the small bowel.

A possible role for histamine in the pathogenesis of mucosal injury during intestinal ischemia arises from its known capability to increase microvascular permeability to macro-molecules [51], [201], [210] and the finding that the plasma histamine concentration is elevated after reperfusion of ischemic intestine. [211] Furthermore, inhibition of diamine oxidase, a histamine-catabolizing enzyme present in large quantities in the superficial epithelial cells of the intestinal mucosa, reduces survival in dogs [212] and rabbits [213] subjected to intestinal ischemia. It appears unlikely, however, that histamine is a mediator of ischemic injury in the small intestine, based on the failure of histamine H_1 and H_2 receptor antagonists to attenuate the increased vascular permeability resulting from ischemia and reperfusion of the intestine. [214] Histamine is also a potent vasodilator, and this effect should decrease, rather than increase, mucosal injury resulting from mesenteric ischemia. [161]

Because most lysosomal enzymes have an acid optimal pH, it has been postulated that ischemia-induced metabolic acidosis may stimulate the release of hydrolytic enzymes from lysosomes. Elevated plasma levels of lysosomal enzymes have been reported in association with acute mesenteric ischemia. [202] Although it has also been postulated that massive doses of corticosteroids administered to patients in a state of shock attenuate the hypoxia-induced lability of lysosomal membranes, [202] no well-controlled studies have demonstrated a beneficial effect of corticosteroid treatment of shock. The reperfusion-induced increase in microvascular permeability after mesenteric ischemia also is not attenuated by pretreatment with methylprednisolone. [214]

A role for reactive oxygen metabolites in the pathogenesis of injury associated with reperfusion of the ischemic bowel has received considerable attention during the past 20 years. Reactive oxygen metabolites play an important role in normal cellular metabolism, most notably as intermediates in mitochondrial and microsomal electron transport systems. [215] More than 90% of the molecular oxygen consumed by most cells is reduced by the mitochondrial electron transport chain to form water, and the remaining oxygen is metabolized to reactive oxygen species. Under normal conditions, tissues are protected from these oxygen-derived free radicals by the action of certain antioxidant enzymes and scavengers.

In adult animals, the digestive system is particularly well endowed with the enzymatic machinery capable of generating significant quantities of reactive oxygen metabolites. For example, the intestine and liver are the richest sources of xanthine oxidase, [216] an enzyme that catalyzes the production of both superoxide and hydrogen peroxide. Xanthine oxidase activity in the small intestine is located primarily within the mucosa, with a gradient of activity from villus tip to base. [217] In addition, the intestine contains a large, resident population of phagocytic cells (i.e., neutrophils, eosinophils, macrophages), which, when activated, produce considerable quantities of superoxide, hydrogen peroxide, and hypochlorous acid. [218] Oxidants generated by either xanthine oxidase or activated phagocytes can injure cells by a variety of mechanisms, including lipid peroxidation, degradation of the extracellular matrix, protein and carbohydrate decomposition, and DNA strand breakage. [215] Cellular enzymatic defense mechanisms against these oxidants include superoxide dismutase, which converts the superoxide anion to hydrogen peroxide and oxygen, and catalase and glutathione peroxidase, which detoxify hydrogen peroxide. [219] Another important oxidant defense is reduced glutathione, which serves both as a cosubstrate for the glutathione peroxidase–catalyzed decomposition of hydrogen peroxide and as a free radical scavenger. [215]

A large body of experimental data supports the hypothesis that reactive oxygen metabolites mediate the microvascular and mucosal permeability changes after reperfusion of the ischemic intestine and stomach in adult animals. [204], [220], [221], [222], [223] and [224] Increased intestinal vascular permeability produced by ischemia and reperfusion injury is not attenuated by antihistamines, indomethacin, or methylprednisolone, arguing against a role for histamine, prostaglandins, and lysosomal enzymes in this injury. [219] In contrast, superoxide dismutase (which scavenges superoxide anions), catalase (which detoxifies hydrogen peroxide), and dimethylsulfoxide (which scavenges hydroxyl radicals and decomposes hypochlorous acid) attenuate the vascular permeability changes observed after reperfusion of the ischemic intestine. [220]

A role for xanthine oxidase in the increased vascular permeability and morphologic changes induced by reperfusion has been proposed based on the following observations: attenuation of injury by pretreatment with allopurinol or pterin aldehyde, which are inhibitors of xanthine oxidase [221], [225], [226]; increased vascular and mucosal permeability during intraarterial infusion of hypoxanthine-xanthine oxidase, a superoxide anion–generating system [227]; attenuation of injury by soybean trypsin inhibitor, a substance that prevents the conversion of xanthine dehydrogenase to xanthine oxidase [222]; and attenuation of reperfusion injury by administration of a tungsten-supplemented, molybdenum-deficient diet, which inactivates xanthine oxidase. [228] In the intestine of developing piglets, however, the total absence of xanthine dehydrogenase/oxidase activity precludes its role in intestinal ischemia and reperfusion injury. [229]

Evidence to support a role for granulocyte-mediated injury after ischemia and reperfusion is also accumulating. Ischemia may lead to neutrophil activation, release or production of neutrophilic oxidants (e.g., superoxide, hydrogen peroxide, hypochlorous acid, *N*-chloramines) and proteases, and subsequent tissue injury. [218], [230], [231] and [232] A five- to sevenfold increase in myeloperoxidase activity, which is an index of granulocyte number, occurs during ischemia, whereas reperfusion induces an 18-fold increase in myeloperoxidase activity in feline intestine. [223] Both neutrophil depletion and prevention of neutrophil adherence significantly attenuate the increased intestinal microvascular permeability induced by ischemia and reperfusion in cat intestine, [204] suggesting that neutrophils, which migrate into the mucosa, mediate the injury produced by reperfusion of the ischemic bowel.

It has also been proposed that xanthine oxidase–derived oxidants may serve as chemoattractants for granulocytes in postischemic adult intestine. Allopurinol (an inhibitor of xanthine oxidase), [218] superoxide dismutase (a superoxide scavenger and inhibitor of neutrophil adherence), [218] catalase (a scavenger of hydrogen peroxide), [233] deferoxamine (an iron chelator), [233] dimethylthiourea (a hydroxyl radical scavenger), [233] and IB $_4$ (a CD18-specific monoclonal antibody inhibitor of neutrophil adherence) [234] all inhibit reperfusion-induced granulocyte accumulation in the small intestine.

Neonatal swine intestine contains significantly fewer resident granulocytes than does that of older piglets, a fact that does not support the theory that neonatal intestine is more vulnerable to oxidant-induced injury than that of older animals. [229] This lower number of resident granulocytes in the mucosa does not preclude the possibility of injury induced by resident granulocytes or granulocytes recruited into the tissue during ischemia or other stresses.

The view that leukocyte–endothelial cell adhesion plays an important role in the pathogenesis of ischemia and reperfusion injury, as well as in other inflammatory conditions of the gastrointestinal tract, [235], [236] has led to an increased interest in defining the factors that modulate leukocyte adherence and emigration in postcapillary venules. Intravital microscopic techniques have been used to monitor and quantify leukocyte–endothelial cell adhesion in mesenteric venules exposed to 60 minutes of ischemia and 60 minutes of reperfusion. [237] During the final 10 minutes of a 60-minute 80% reduction in mesenteric blood flow, the numbers of adherent and emigrated leukocytes increase by fourfold and threefold, respectively. At 60 minutes after reperfusion, sevenfold and eightfold increases in adherence and emigration are noted. Electron microscopic analyses of postischemic mesenteric venules reveal that more than 85% of the leukocytes that emigrate into the adjacent interstitial compartment are neutrophils. [238]

Several chemical mediators produced by endothelial or parenchymal cells have been implicated in the leukocyte–endothelial cell adhesion elicited by mesenteric ischemia and reperfusion. A growing body of evidence implicates xanthine oxidase–derived superoxide as a major mediator in this process. A role for superoxide is supported by reports that superoxide dismutase, whether administered before ischemia or after reperfusion, effectively reduces the number of adherent and emigrated leukocytes in mesenteric venules exposed to ischemia and reperfusion. [237], [239] Superoxide dismutase is also effective in attenuating the adherence of neutrophils to endothelial cell monolayers exposed to anoxia and reoxygenation. [239] However, superoxide dismutase is ineffective in altering neutrophil adhesion to biologically inert surfaces (e.g., plastic), indicating that endothelial cells are required for the enzyme's antiadhesive action. A similar antiadhesive action has been reported to occur with allopurinol, [237], [240] suggesting that xanthine oxidase is a likely source of the superoxide produced after reperfusion. Xanthine oxidase, rather than neutrophils, also appears to be responsible for generating the oxidants that mediate reperfusion-induced lipid peroxidation in intestinal mucosa. This contention is based on reports demonstrating that although both allopurinol and CD18-specific monoclonal antibodies prevent the reperfusion-induced increase in mucosal myeloperoxidase activity, only allopurinol prevents the rise in tissue conjugated dienes, an index of membrane lipid peroxidation. [241], [242]

The mechanism by which superoxide mediates reperfusion-induced leukocyte adherence and emigration within the mesenteric microcirculation has received much attention. The generation of superoxide on the surface of postcapillary venules of hamster cheek pouch by hypoxanthine-xanthine oxidase leads to leukocyte adherence, which can be prevented by superoxide dismutase. [243] Superoxide may promote leukocyte adherence by interacting with extracellular fluid to form a superoxide-dependent chemoattractant [244]; however, this possibility is not supported by studies demonstrating that exposure of intestinal interstitial fluid to superoxide does not generate a chemoattractant for neutrophils. [245] A more likely explanation for the proadhesive action of superoxide is that it inactivates an antiadhesion molecule that is normally produced by endothelial cells. NO, a product of L-arginine metabolism in endothelium that is rapidly inactivated by superoxide, may be such an endogenous antiadhesion molecule. NO donors are very effective in preventing the recruitment of leukocytes, platelet-leukocyte aggregation, and tissue injury associated with ischemia/reperfusion. [246] Inhibitors of NO production lead to a dramatic increase in the number of leukocytes adhering to and emigrating from mesenteric venules. [139] This adhesion response can be prevented or reversed by simultaneous exposure of venules to an NO synthase inhibitor and either L-arginine (but not D-arginine) or nitroprusside, which spontaneously generates NO. The observations that NO synthase inhibitors promote leukocyte adherence while superoxide dismutase reduces reperfusion-induced adherence are consistent with the view that the enhanced formation of superoxide by postischemic endothelial cells leads to NO inactivation and consequently results in enhanced leukocyte adhesion. Such a mechanism would explain why superoxide dismutase exerts an antiadhesive effect in postischemic tissues, because the enzyme would prevent inactivation of NO. NO also stabilizes mast cells through a mechanism that involves superoxide. [247]

Growing evidence indicates that in addition to protecting the microvasculature, endogenous NO regulates mucosal barrier integrity. [248] Inhibition of endogenous NO leads to increases in reperfusion-induced mucosal permeability, whereas exogenous sources of NO can reduce reperfusion-induced mucosal barrier dysfunction independently of alterations in intestinal blood flow. [249] Mucosal injury after hypothermic ischemia and reperfusion is also attenuated by exogenous sources of NO and therefore may be beneficial in intestinal allografts subjected to prolonged hypothermic ischemia. [250]

Catalase also has been shown to attenuate reperfusion-induced leukocyte adherence in mesenteric venules. [240] This observation is consistent with reports that demonstrate that hydrogen peroxide is produced by endothelial cell monolayers exposed to anoxia and reoxygenation, [251] and hydrogen peroxide promotes neutrophil adherence to cultured endothelial cells and in mesenteric venules. [252], [253] The levels of hydrogen peroxide required to promote adherence are well within the range of the hydrogen peroxide concentration produced by activated neutrophils. Both in vivo and in vitro studies indicate that hydrogen peroxide–induced neutrophil adherence is mediated by platelet-activating factor (PAF). [252], [253] PAF-receptor antagonists effectively attenuate the neutrophil adherence induced by hydrogen peroxide in feline mesenteric venules [234] and isolated canine carotid arteries. [254] The hydrogen peroxide–induced, PAF-mediated leukocyte adherence is prevented mostly by monoclonal antibodies directed against the common ß subunit of CD11/CD18.

The proposed role of reactive oxygen (hydrogen peroxide, superoxide) and nitrogen (NO) species in mediating the endothelium-dependent inflammatory responses observed in the gastrointestinal microcirculation after ischemia/reperfusion is summarized in Figure 22-19. Ischemia/reperfusion leads to an increased production of superoxide and hydrogen peroxide by the enzyme xanthine oxidase, with a corresponding reduced production of NO by endothelial NO synthase. The enhanced generation of oxidants results in the activation and deposition of complement, and phospholipase A $_2$–mediated production of leukotriene B $_4$ and PAF. Oxidants also mediate the initial expression of P-selectin by mobilizing the leukocyte rolling receptor from its preformed pool (Weibel-Palade bodies) in endothelial cells. Firm adhesion of leukocytes, which is mediated by ß $_2$-integrins (CD11/CD18), is induced by the engagement of activated complement, leukotriene B $_4$, and PAF with their receptors on rolling leukocytes. Sustained rolling and adhesion of leukocytes on endothelial cells is ensured by an oxidant-dependent synthesis of endothelial cell adhesion molecules, such as E-selectin and intracellular adhesion molecule-1 (ICAM-1). Oxidants, derived from either endothelial cells or leukocytes, elicit this biosynthetic response by activating specific nuclear transcription factors (e.g., nuclear factor-?B) that bind to the genes for these adhesion molecules. The inflammatory responses to ischemia/reperfusion are amplified by mediators released from mast cells and macrophages that normally reside near postcapillary venules. [255]

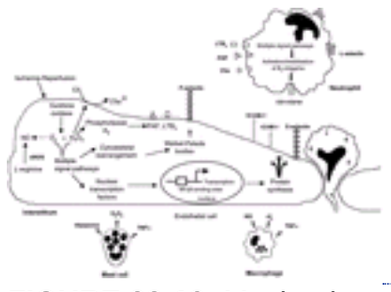

FIGURE 22-19. Mechanism proposed to explain the endothelium-dependent inflammatory responses observed in the postischemic microvasculature. *C5a*, complement 5a; *CD11/CD18*, leukocyte adhesion glycoprotein; *eNOS*, endothelial nitric oxide synthase; H_2O_2, hydrogen peroxide; *I/R*, ischemia/reperfusion; *ICAM*, intracellular adhesion molecule; LTB_4, leukotriene B_4; *NF-?B*, nuclear factor-?B; *NO*, nitric oxide; O^-_2, superoxide; *PAF*, platelet-activating factor; *TNF-a*, tumor necrosis factor-a; *VCAM*, vascular cell adhesion molecule. (From ref. 255.)

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CHAPTER 23

Mark W. Babyatsky and Daniel K. Podolsky

GROWTH AND DEVELOPMENT OF THE GASTROINTESTINAL TRACT

Structural and functional integrity of the mature gastrointestinal (GI) tract results from a developmental program that derives from many levels of regulation and interactions. During embryonic and fetal life, the GI tract and its accessory organs evolve from a single, nondescript tubular structure into constituent organs that exhibit unique structural, cellular, and functional specificities by the time of parturition. Final functional maturation, however, is completed only after birth. Recent gene targeting and deletion studies in mice and advances in understanding of the transcriptional control of growth and development have further defined the complex interactions that determine the cellular differentiation and growth patterns that result in the mature GI tract.

Although important characteristics in architectural organization are similar along the length of the GI tract, regional specialization is inherent to its development. Disruption of the normal sequence of developmental evolution in utero leads to a wide spectrum of anatomic disorders usually manifest during infancy. The GI tract undergoes dynamic renewal throughout life, with a continuing, if regionally variable, cycle of cellular proliferation, differentiation, and senescence. An appreciation of the mechanisms of ongoing self-renewal, apoptosis, and maturation is integral to an understanding of the pathogenesis of a number of neoplastic and inflammatory disorders.

This chapter first considers the structural development of the GI tract and pancreas; a discussion of functional maturation and its regulation follows. The latter also includes an overview of the processes controlling homeostasis of the epithelium in the adult, with an analysis of the transcriptional regulation of GI tract development, highlighting discoveries made through gene deletion or overexpression of specific genes in transgenic mice. Although much of our understanding of development has been achieved through the study of rodents, which demonstrate altricial development (i.e., maturational events concurrent with weaning), processes critical to human GI tract development are similar, even though human gastrointestinal development is precocious (i.e., functionally mature at birth). These processes, which unfold over the course of the normal 40-week gestation in humans, occur during the 21-day gestation of mice and rats. The relatively short period of gestation in these species makes them especially suitable for studies of developmental processes.

EMBRYOLOGY AND HISTOGENESIS

The human embryonic GI tract can first be recognized in the 4th week of gestation, when infolding creates an endoderm-lined tubular structure extending from the esophagus to the cloaca. The nascent GI tract is joined at its ventral region to the yolk stalk and allantois, which initially expand outside the embryo. During development, the GI tract is comprised of foregut (esophagus, stomach, and duodenum to the level of the ampulla of Vater), liver, pancreas, biliary tract, midgut (second portion of the duodenum to the proximal transverse colon), and hindgut (distal transverse colon to the proximal anal canal).

Esophagus

The foregut and respiratory tract are initially formed by a single tube at the proximal end of the primitive GI tract. [1] Subsequent development includes longitudinal division, resulting in the separation of respiratory and foregut structures, with the esophagus occupying a dorsal position by the 2nd month of gestation. The intimate relationship between the developing esophagus and tracheobronchial structures contributes to the potential for developmental anomalies (e.g., fistulae) involving connections between the esophagus and trachea. The esophagus can be distinguished from the stomach as early as the 4th week of gestation. [2] Rapid cephalad extension of the esophageal anlage leads to an elongated structure, which achieves its mature length relative to other structures by the 7th week of gestation. At the same time, rapid proliferation of the endodermal lining leads to near or actual occlusion of the lumen. Recanalization occurs through vacuolization of the endodermal cells, reestablishing the esophageal lumen by the 8th week of gestation. A similar sequence of proliferation, lumenal obliteration, and reestablishment of the lumen through vacuolization is found throughout the GI tract. In esophageal development, failure of recanalization may be a factor in the development of esophageal stenosis or atresia and may be associated with tracheoesophageal fistulae.

The esophagus is initially covered with a simple, cuboidal epithelium. By the 5th week of gestation, two layers of cuboidal cells and some neuroblasts dispersed in a developing layer of circular smooth muscle have developed. By 8 weeks of gestation, synaptic protein and glial supporting tissue can be demonstrated. [3] The external longitudinal layer of muscle develops later in gestation than the inner layer of circular muscle and is first appreciated during the 8th week. The epithelium becomes vacuolated during the 7th week of gestation, leading to the formation of a longitudinal channel, which forms the lumen when the vacuolization process is complete in the 8th week. In the 10th week of gestation, the esophageal epithelium is ciliated; subsequent proliferation and maturation lead to a stratified squamous epithelium typical of the adult esophagus by 22 weeks of gestation. Superficial glands may be found as early as 18 weeks of gestation, but the deep mucosal glands characteristically found in the mature esophagus are rare before birth and appear to develop during postnatal life.

The myenteric plexus can be demonstrated by gestational week 10, and mature ganglion cells by 13 weeks. [3] Extensive innervation of the esophagus is established before the onset of motor activity at 16 to 17 weeks of gestation, but maturation, reflected by an increase in neuron diameter and reduction of cell and fiber density, continues throughout gestation and into the 1st year of life. The cells that ultimately form the enteric nervous system migrate from the neural crest during the 1st trimester. [4] By the 8th week of gestation, neuroblasts have matured and become more numerous; synaptic protein and glial supporting tissue can be demonstrated asymmetrically penetrating the outer layers of the poorly differentiated muscular layer. [5] Neuropeptide immunoreactivity appears during the 11th week of gestation, when bombesin and neuropeptide Y can first be detected in the myenteric plexus. [6] Other neuropeptides are first expressed between the 13th and 18th weeks of gestation and, in order of appearance, include vasoactive intestinal polypeptide (VIP), galanin, substance P, somatostatin, metenkephalin, and calcitonin gene–related peptide (CGRP). Neuronal density peaks at 16 to 20 weeks of gestation, decreasing to adult levels in the 3rd trimester. In neonates and adults, the muscle of the proximal esophagus is striated, derived from the caudal branchial arches, and innervated by branches of the vagi. In the midesophagus, an intermediate zone exists after birth where neuropeptides are found in the smooth muscle circular layer but not in the longitudinal layer of striated muscle. Neuropeptide immunoreactivity can be detected in both muscle layers in the distal esophagus. The submucous plexus, absent in the adult human esophagus, is present by 13 to 19 fetal weeks, when protein gene peptide 9.5, a generalized neuronal marker, and VIP immunoreactivity can be detected.

The mature esophagus is a muscular organ throughout its length. The smooth muscle in the distal two thirds of the esophagus derives its innervation from the

splanchnic plexus. In mice, the entire esophagus is lined by smooth muscle until birth; skeletal muscle appears to be derived from differentiated smooth muscle cells by transdifferentiation during late fetal and early postnatal development, [7] commencing in the proximal esophagus at murine gestational day 16 and reaching the mature pattern of striated and smooth muscle on postnatal day 8. [8] However, the basic helix-loop-helix transcription factors that regulate skeletal muscle development (myogenic regulatory factor [Myf5]) are present at 12 to 13 days of mouse gestation, suggesting that endodermal cells of the foregut are committed to skeletal muscle differentiation even before smooth or skeletal cells become recognizable at 14 to 15 days of gestation. [9] However, gene disruption of Myf5 does not affect the development of smooth muscle, but it markedly delays the appearance of skeletal muscle by presumed transdifferentiation. [10] Cholinergic neurons are present before transdifferentiation occurs, but nicotinic receptor clusters do not appear until striated muscle is recognizable. Whereas targeted deletion of c-Ret or glial cell line–derived neurotrophic factor (GDNF) genes in mice leads to a lack of enteric neurons of the small intestine and colon with a phenotype similar to that of Hirschsprung disease (see section " Functional Maturation—Colon"), mice lacking Mash1, a transcription factor necessary for the development of enteric neurons of the esophagus, have no milk in their stomach and cannot relax their lower esophageal sphincter. However, transdifferentiation progresses normally; extrinsic neurons and extrinsic muscle development of the esophagus are normal in Mash1 null mice. [11]

Stomach and Duodenum

In the 4-week old human embryo, the stomach develops as a fusiform dilation of the foregut in the neck (Fig. 23-1A). [12] As it grows, the stomach descends into the abdomen by the 7th week of gestation. The gastric walls grow at disparate rates, creating the characteristic asymmetric shape as the dorsal border (which becomes the greater curvature) grows more rapidly than the ventral border (Fig. 23-1B). During the 6th week of gestation, the stomach undergoes a 90° clockwise rotation along its longitudinal axis so that the dorsal border lies to the left and the ventral border (which become the lesser curvature) moves to the right (Fig. 23-1C). As a result of this rotation, the left and right vagi largely supply the anterior and posterior areas of the stomach, respectively. The pyloric region of the stomach can be identified by the 3rd month of gestation. Although the shape of the stomach is suggestive of the adult gastric configuration by the end of the 7th gestational week, the cardia is still moving to the left of midline and the antrum is still moving to the right, so that the ultimate stomach contours are not reached until 8 to 9 weeks of gestation. [13] The greater curvature, lesser curvature, corpus, antrum, and pylorus can be clearly distinguished by 14 weeks. The duodenum forms from the distal foregut and cephalad midgut, which meet and fuse just distal to the area that becomes the ampulla of Vater. The duodenum grows rapidly, rotating to the right at the time of gastric rotation, creating a C-shaped loop in its retroperitoneal position by the 6th week of gestation. During this period of rapid growth, the duodenal lumen is temporarily obliterated by proliferation of the epithelium, but it is reestablished through vacuolization by the 8th week. Apoptosis of developing duodenal cells does not increase during duodenal recanalization and thus is not responsible for this process. [14] Failure of recanalization results in duodenal stenosis or atresia.

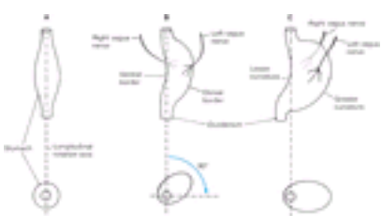

FIGURE 23-1. Schematic representation of the positional changes of the stomach. **A:** The stomach at 4 weeks. **B:** Rapid growth of the dorsal border, resulting in asymmetry. **C:** After a 90° clockwise rotation along the longitudinal axis, the left and right vagus nerves supply the anterior and posterior stomach, respectively. (Adapted from ref. [12].)

In gastric histogenesis, the stomach is lined by a stratified columnar epithelium composed of two to three cell layers in the 7th week of gestation. [15] Gastric pits, although rare at 7 weeks, begin to increase in number initially in the region of the lesser curvature, then in the greater curvature in the 8th week and the antral and cardiac regions by weeks 10 and 11. The glands first develop mucous neck cells, followed by parietal and chief cells. Ablation of parietal cells by the administration of ganciclovir to transgenic mice that express herpes simplex virus 1 thymidine kinase, driven by the H^+,K^+-ATPase promoter, is accompanied by ablation of the chief and mucous cells. [16] With cessation of ganciclovir, however, all cell types return, supporting the existence of pluripotent stem cells in the gastric mucosa. Targeted disruption of the H^+,K^+-ATPase gene a subunit in mice leads to complete inhibition of acid secretion, but parietal and chief cells and the H^+,K^+-ATPase ß subunit are preserved; gastric metaplasia with poorly differentiated ciliated cuboidal cells is seen. [17] In contrast, ablation of the ß subunit results in gastric mucosal hypertrophy, fewer parietal and chief cells, and a failure of delivery of the a subunit to the plasma membrane, in addition to achlorhydria and hypergastrinemia. [18] Omeprazole-induced hypochlorhydria, which blocks both H^+,K^+-ATPase subunits equally, results in a decreased parietal cell mass. [19]

Enteroendocrine cells can be detected by the 8th week of human gestation, and the fully differentiated spectrum of endocrine cell types can be detected by week 10, [20] suggesting a possible role in fetal development of the stomach. Substance P, neurokinin A, and CGRP appear first, followed by peptide YY cells, which are frequently colocalized with gastrin G cells after birth. During the first postnatal week, gastrin G cells appear in the gastric antrum while those in the pancreas disappear. Similarly, somatostatin D cells appear throughout the gastric mucosa, although they are detectable in the pancreas and duodenum in midgestation. Mice rendered deficient in Pdx1, a transcription factor that regulates multiple pancreatic and enteroendocrine genes, demonstrate a lack of G cells, but D cell expression is preserved. [21] Although earlier studies failed to demonstrate enterochromaffin-like (ECL) cells in the gastric mucosa before birth, histamine-containing cells appear by midgestation. [22] ECL cell proliferation does not accelerate until 1 to 3 weeks postnatally. Targeted disruption of the gastrin or gastrin/CCK-B receptor gene in mice does not affect ECL development, suggesting that gastrin, which stimulates ECL proliferation in mature animals and humans, does not play a role in early ECL development. [23], [24] and [25]

In the rat, primitive chief cells, which resemble mature chief cells in ultrastructure but, analogous to mucous neck cells, are capable of binding various lectins, are detectable from the 16th day of gestation until postnatal day 14. Gastric stem cells, although not yet clearly defined, appear to be located in the mucous neck region, producing the differentiated cell types that migrate toward the surface or base of the pits. [26] Ablation of parietal cells in transgenic mice by diphtheria toxin alters the balance between all the cell types, suggesting important developmental interactions between the cell lineages. [27]

Muc1, an epithelial mucin expressed in the mature upper GI tract, is also expressed at high levels early in rat gestation. [28] Muc1 mRNA is detected in the stomach by the 10th day of gestation, and protein is found by day 12. Initially, Muc1 lines the lumenal border of the gastric mucosa immediately after branching and differentiation of the gastric epithelial buds begin, suggesting a role for Muc1 in gastric epithelial development. In humans, Muc1-,-4, -5AC, -5B, and -6 mRNAs appear by 8 weeks of gestation. [29] Muc3 mRNA appears by 10.5 weeks, Muc2 not until 26 weeks of gestation. In adults, Muc1, -5AC, and -6 predominate; gastric carcinoma is associated with a fetal pattern of MUC gene expression. Spasmolytic polypeptide, a mucin-associated member of the trefoil family of peptides, [30] is abundantly expressed in fetal stomach before gastrin or somatostatin is expressed, [31] suggesting a role in gastric functional maturation.

Tyrosine kinases and phosphatases have been shown to play key roles in gastric development. Two families of receptor type tyrosine kinase genes, *elk/erk* and *esk/TTK,* are expressed at high levels in rat stomach from days 14 to 16 days of gestation, [32] decreasing after day 18, and are undetectable in adult rats. Protein tyrosine phosphatases may also be important in that PRL1 phosphatase expression correlates with the terminal differentiation of gastric zymogen cells and may play roles in the development of the esophagus, liver, and small intestine. [33] Both kinase and phosphatase genes are expressed early in development and reexpressed in a variety of gastric cancers.

Pancreas

The liver, biliary tract, and pancreas share a developmental origin in the ventral outpouching of the foregut. The pancreas emerges from the distal foregut as two buds at 9.5 days of gestation in the mouse and 10.5 days in the rat. In humans, the pancreas emerges from the hepatic diverticulum, which appears between the 3rd and 4th weeks of gestation and consists of the liver primordia and biliary system as well as the ventral pancreas. The dorsal bud is recognized first; shortly afterward, the ventral bud separates from the caudal-most region of the hepatic diverticulum (Fig. 23-2A,B). As the duodenum grows and rotates, the ventral bud migrates around the duodenum to fuse with the dorsal bud on the 11th day of mouse gestation and 5th week of human gestation, forming the uncinate process and inferior portion of the pancreatic head (Fig. 23-2C,D). Malrotation of the ventral bud results in an annular pancreas. The ventral duct also fuses with the distal portion of the dorsal duct

to form the duct of Wirsung, which becomes the main pancreatic duct emptying into the ampulla of Vater; the accessory duct of Santorini reflects the residual proximal dorsal duct (see Fig. 23-2C,D). Incomplete fusion of the two ductal systems results in pancreas divisum, which may be associated with later development of recurrent pancreatitis.

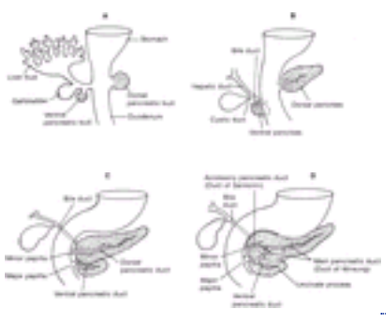

FIGURE 23-2. Successive stages in the maturation of the pancreas. **A:** The pancreas at 4 weeks. Note the location of the ventral bud arising from the primitive hepatobiliary system (i.e., hepatic diverticulum). **B:** The pancreas at 5 weeks. Note the rapid growth and elongation of the dorsal bud. **C:** The pancreas at 6 weeks. Note the migration of the ventral pancreatic bud to join the inferior portion of the dorsal bud. **D:** The pancreatic ductal system at 6 weeks. The main pancreatic duct joins the bile duct to enter the duodenum at the minor papilla. (Adapted from ref. [12].)

Early pancreatic development requires the interplay between endodermal and mesenchymal tissue. [34] Pancreatic fetal development appears to occur in three main stages. Initially, histological differentiation leads to recognition of distinctive pancreatic tissue on day 9.5 of gestation in mice and day 10.5 in rats, during which the ventral wall of the primitive gut endoderm begins to bulge into the surrounding mesoderm as part of the hepatic diverticulum (Fig. 23-3A). [35] During the subsequent 3 to 4 days of gestation, cytodifferentiation progresses, and low but significant levels of both exocrine and endocrine pancreas-specific gene products can be detected. [36] Beginning about day 15 of rat gestation, further differentiation leads to a rapid increase in rough endoplasmic reticulum, specific acinar enzymes, and islet cell hormones, as well as the appearance of zymogen and hormone granules. [37] After birth, the introduction of nutrients and weaning result in changes in the relative expression of pancreas-specific gene products, leading to the mature, terminally differentiated state. [38]

FIGURE 23-3. Schematic representation of pancreatic development. **A:** Mouse and rat development. **B:** Human development.

By the 8th day of mouse gestation, endodermal tissue in the regions of future pancreatic development is committed to pancreatic epithelial differentiation. Initially, morphogenesis of the pancreatic anlage occurs at two separate anatomic sites. The dorsal pancreatic bud appears first as a broad-based swelling immediately dorsal to the rudimentary stomach; the ventral pancreas appears 12 hours later on the opposite side of the gut, where the common biliary duct meets the ventral wall of the gut. [39] ^{3}H-thymidine incorporation studies have demonstrated that the appearance of the pancreatic buds occurs through a process of evagination and folding, as opposed to differential cell division and growth. By day 11 of mouse gestation, the pancreatic rudiment appears as a hollow epithelial bulb surrounded by condensed mesenchyme; single layers of cells face the lumen, linked together by junctional complexes. [40] At about the same time, the two primitive glands merge. During the next few days, rapid multiplication of these differentiated cells results in thickening of the pancreatic bud, which fills with tubular ductlike structures. At this stage, endocrine cells also appear scattered throughout the branching epithelium. By gestational day 15 in the mouse and days 16 to 17 in the rat, typical acini are evident. The endocrine cells pinch off from the pancreatic ductules, forming the islets of Langerhans.

In human development, the pancreas emerges histologically at the end of the 8th week of gestation as a collection of primitive epithelial tubules (Fig. 23-3B). [41] These structures subsequently branch, and their termini become surrounded by cell buds from which mature acini develop. During the 3rd month of gestation, secretory acini and islets of Langerhans are recognizable. At the beginning of the 2nd trimester, a lobular arrangement of the pancreas becomes visible, leading to the compact structure of the mature pancreas. Morphologic and functional development of ductal and acinar cells has been reported at 19 weeks of gestation. [42] Also at 19 weeks, the pancreatic mucin gene *MUC1* appears [43]; *MUC5B* appears at 12 weeks, and *MUC5* at 26 weeks. [44] *MUC3,* strongly expressed in adult pancreas, is not expressed during fetal life.

The acinar cell of the mature pancreas is normally a quiescent cell with a low mitotic rate. In fetal development, however, newly formed acinar cells maintain active cell division even after cytodifferentiation. [45] Antibodies specific for lumenal plasma membranes of acinar or duct cells of the mature exocrine pancreas demonstrate that fetal acinar cells undergoing rapid mitosis still express duct cell antigen. [46] Further, during regeneration after experimental pancreatitis, acinar cells transiently express ductal antigen, indicating a less differentiated state, until active cell division decreases.

Small Intestine and Colon

During the 5th week of gestation, the tubular midgut portion of the intestinal tract, joined to the yolk sac by the vitelline duct, rapidly elongates, extending ventrally into the body stalk. The vitelline duct, normally obliterated before birth, occasionally persists as a Meckel diverticulum. Between weeks 5 and 10, the small intestine extends through the umbilicus as the result of further elongation. During this period of rapid elongation, the midgut rotates 90° around the superior mesenteric artery in the dorsal mesentery. This rotation brings the proximal midgut to the right and the distal midgut to the left (Fig. 23-4A). As the midgut reenters the abdominal cavity in the 10th week of gestation, it undergoes a further 180° rotation (Fig. 23-4B). The proximal jejunum enters first and occupies the left side of the abdomen, and the ileum settles into the right side. The cecal swelling enters last, locating temporarily in the right upper quadrant just caudal to the right lobe of the liver (Fig. 23-4C). Between the 3rd and 5th months of gestation, the cecum descends into the right iliac fossa and becomes fixed to the posterior wall of the abdomen (Fig. 23-4D). As the liver increases in size, the ascending colon and the hepatic flexure become distinct from the transverse colon. The descending colon loses its mesentery and becomes anchored to the abdominal wall, leaving the sigmoid colon in its more caudal position on mesentery. The rectum arises independently from the remaining large intestine as a subdivision of the cloaca, separated from the urogenital sinus by the urorectal septum. The urorectal septum reaches the cloacal membrane during the 7th week of gestation, forming the perineum. During the 8th week, the rectum fuses with the colon, and the cloacal membrane forms the anal membrane, lost during the 9th week to establish communication with the amniotic space.

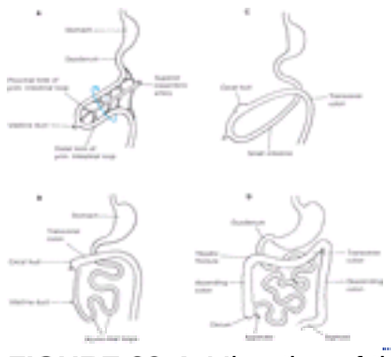

FIGURE 23-4. Migration of the intestinal loops. **A:** The intestine after a 90° rotation around the axis of the superior mesenteric artery, the proximal loop on the right and the distal loop on the left. **B:** The intestinal loop after a further 180° rotation. The transverse colon passes in front of the duodenum. **C:** Position of the intestinal loops after reentry into the abdominal cavity. Note the elongation of the small intestine, with formation of the small intestine loops. **D:** Final position of the intestines after descent of the cecum into the right iliac fossa. (Adapted from ref. [12].)

Histologically, the small and large intestines are lined initially by a simple cuboidal epithelium. As in the duodenum, epithelial proliferation during weeks 6 and 7 may lead to occlusion of the lumen. At 8 weeks, the epithelium is stratified; heparin sulfate, type IV collagen, and laminin, produced by epithelial cells, are detectable at the base of the undifferentiated epithelium, forming a basement membrane before further differentiation. [47] The lumen is reestablished during weeks 9 and 10, and a simple columnar epithelium reappears by the 12th week of gestation.

Villi form first in the proximal intestine during week 9 (Fig. 23-5), emerging in successively more distal regions over the ensuing days and completely formed by the end of week 13. In the stratified epithelium of the near-term fetal rat small intestine and colon, secondary lumina are thought to play a crucial role in villi formation. These lumina surround the main lumen and are joined by continuous tight junctions. The secondary lumina enlarge and eventually fuse with the main lumen, leaving a villus outpouching. Crypt formation begins in the 10th to 12th weeks of gestation, also progressing in a proximal-to-distal sequence. Postnatal development of absorptive enterocytes is associated with marked lengthening of the microvilli. [48]

FIGURE 23-5. Schematic representation of small intestine development. **A:** Mouse and rat development. **B:** Human development.

The four cell types found within the intestinal epithelium—absorptive columnar, goblet, enteroendocrine, and Paneth cells—appear to arise from a common progenitor cell. [49] Studies in chimeric and transgenic mice in which several cellular markers were used have demonstrated that the crypts in mature animals are clonal products of single progenitor cells. [50] However, crypts initially appear to be polyclonal; a single stem cell must become entrenched during development through undefined competitive mechanisms. Ablation of Paneth cells in transgenic mice with use of the cryptdin-2 gene promoter to drive the expression of diptheria toxin A or SV40 T antigen does not affect the expression or maturation of the other three cell types. Thus, Paneth cells, despite close anatomic association, are not critical for crypt stem cell function. [51]

The intestinal tube is initially surrounded by a layer of mesoderm that ultimately forms connective tissue, muscle, and serosa. As in gastric mucosa, enteroendocrine cells can be detected by the 8th week of gestation. Transgenic studies demonstrate that enteroendocrine cells share a developmental origin with endocrine cells of the pancreas. [52] Furthermore, secretin, peptide YY, and gastrin can de detected in fetal pancreatic islet cells. [53], [54] Mice with targeted disruption of the BETA2/NeuroD transcription factors, which normally bind an E-box motif crucial for enteroendocrine and pancreatic endocrine expression, fail to develop mature pancreatic islets and secretin- and CCK-producing cells in the small intestine, [55] and mice lacking the Pdx1 transcription factor demonstrate agenesis of the pancreas and a dramatic reduction in the number of small intestinal endocrine cells. [56] The circular muscle layer can be discerned by week 8; its appearance is followed closely by the emergence of the longitudinal muscle layer. The muscularis mucosae develops later but is present throughout the bowel by 20 weeks of gestation. The Auerbach plexus can be found by week 9 and the Meissner plexus by week 13, and Peyer patches emerge by about week 20.

Regional histogenetic differences during colon development have been observed. During late gestation in the rat (i.e., 18 days), the architecture of the proximal and distal colon is similar. However, the proximal but not the distal colon subsequently forms villuslike structures, although those disappear by 22 to 26 days after birth (Fig. 23-6). [57]

FIGURE 23-6. Schematic representation of colon development. **A:** Mouse and rat development. **B:** Human development.

FUNCTIONAL MATURATION

The structural features of the GI tract are well developed by the end of the 2nd trimester. In general, human GI tract functional development is precocious, with a wide spectrum of capabilities appearing well in advance of the time they are needed (Fig. 23-7), whereas many other species demonstrate altricial development, in which the development of many absorptive and enzymatic activities coincides with weaning, a process that begins in the 3rd week of life.

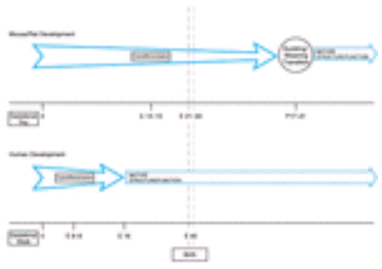

FIGURE 23-7. Comparative time course of major events in gastrointestinal development in rodents and humans. The digestive tract is structurally and functionally mature by the end of the first half of gestation in humans but reaches maturity in rodents only during the third postnatal week, following the suckling-weaning transition. *E,* embryonic day postconception; *P,* postnatal day.

Esophagus

Tagged erythrocyte isotope studies [58] and ultrasonography demonstrate that fetal swallowing occurs as early as the 11th week of gestation. The rate of fetal swallowing of amniotic fluid increases from 13 mL/d at 20 weeks' gestation to 450 mL/d in the presence of a mean amniotic fluid volume of 850 mL at term. [59] The role of swallowing in the regulation of amniotic fluid volume remains unclear, but fetal swallowing is important in GI tract development; fetal sheep [60] and rabbits [61] treated with esophageal ligation in the 2nd trimester demonstrate altered enterocyte morphology (including absence of microvilli), glycogen accumulation, and altered lysosomal morphology, changes similar to those observed in malnourished infants. Absorption of infused intraamniotic nutrients has been demonstrated in fetal rabbits, indicating that nutrient delivery via the fetal GI tract may be a potential treatment for intrauterine growth retardation. Sucking can be detected as early as 18 to 20 weeks' gestation, [62] although in preterm infants, sucking movements are generally feeble. By 34 to 35 weeks' gestation, a mature nutritive sucking pattern develops. The esophageal motility of preterm infants is characterized by poorly propagated, low-pressure biphasic contractions. After birth, swallowing in the full-term infant remains poorly coordinated for the first 12 hours, with both a high peristaltic rate and frequent nonperistaltic, simultaneous contractions throughout the length of the esophagus.

Ultrasonographic studies demonstrate the development of a functional lower esophageal sphincter during the 32nd week of human gestation, causing gastric enlargement by reduction of gastroesophageal reflux. [63] Lower esophageal sphincter pressures increase dramatically during the last trimester and again postnatally, [64] achieving adult levels by 3 to 6 weeks of age. Free gastroesophageal reflux is common postnatally and persists in up to 10% of infants for the 1st year. [65]

Stomach

Structural features of the stomach are also well developed by the end of the 2nd trimester, while functional maturation continues. Neither gastric secretory capacity nor gastric motility is fully developed at birth in either humans or rats. By the 27th to 28th week of gestation in humans, the gastric antrum exhibits only 20% to 25% of the motility reached by the term infant. [66] The gastric mucosa of most animals, including humans, is capable of secreting some acid before birth. [67] As noted, parietal cells are present before the end of gestation. In the rat, basal gastric acid secretion increases from the 19th to the 21st day of gestation, with maximal stimulation by pentagastrin at day 21. [68] However, acid secretion in the neonatal rat is very low and is insensitive to gastrin. [69] The pH of the gastric contents is nearly neutral in the term rat, remaining between pH 5 and 6 on day 10 after birth, falling to pH 4 on day 15, and reaching adult levels after weaning. [70] In humans, acid production, which is less than 50% of adult values during the first 3 months of life, reaches mature levels at 2 years of age.

Neonates are relatively insensitive to gastrin, evidenced by neonatal hypergastrinemia in humans, [71] dogs, [72] and rats. [73] Studies have revealed that the CCK-B receptor that binds gastrin is present before birth, [74] suggesting that the receptor is functionally immature. Compared to the acid response to gastrin, an early normal gastric acid response to cholinergic stimulation is present at birth, [75] whereas the gastric secretory response to histamine [76] does not mature until weaning. Produced by the parietal cell along with hydrochloric acid, intrinsic factor is detectable in human gastric mucosa by the 14th week of gestation; it increases rapidly after birth and achieves mature levels by postnatal day 10. The intrinsic factor receptor is expressed coordinately with its ligand and becomes restricted to the ileum at about 25 weeks' gestation. [77] Gastric lipase is expressed as early as 11 weeks of gestation, reaching adult levels by the 3rd postnatal month. [57]

Pepsinogen 1 immunoreactivity can be demonstrated in secretory granules of primitive chief cells of the murine fundic mucosa by the 16th day of gestation. [78] By postnatal day 13, pepsinogens 1 and 3 are present in almost equal amounts in rat fundic mucosa; pepsinogen 1 levels rise to adult levels after weaning. In humans, pepsinogen secretion is half of adult levels during the first 3 months after birth, rising to adult levels by 2 years of age. The mucous neck cell is the primary source of pepsinogen in the neonate; mature chief cells, which appear after birth, soon become the major source.

In the rat, concentrations of free cortisone rise immediately before the onset of weaning and peak during weaning. [59] In the neonate, injection of adrenocorticotropic hormone (ACTH) or glucocorticoids causes a precocious rise in gastric pepsinogen levels [79] and the number of functional gastrin receptors, [80] an early rise in antral gastrin concentrations, and an earlier induction of the acid secretion response to histamine, carbachol, and gastrin stimulation. Furthermore, cortisone induces a precocious rise in pepsinogen mRNA and a decrease in methylation of the pepsinogen genes, reflecting induction of chief cell differentiation. [81] Maturational glucocorticoid sensitivity disappears after weaning. Thyroxine also induces a precocious rise in pepsinogen activity, an effect that is additive with that of corticosterone. Estrogens reduce food intake, acid secretion, serum gastrin levels, and the number of gastrin receptors in the female or castrated male rat. [82]

Pancreas

Functional Development—Exocrine In contrast to the differentiation of endocrine cells (see next section), pancreatic exocrine cell differentiation follows morphogenesis. Exocrine gene transcription begins 24 hours after the pancreatic diverticulum is apparent. About 85% of the mature gland is comprised of the acinar cells of the exocrine pancreas, which synthesize and secrete large amounts of digestive enzymes, including amylase, lipase, trypsins, carboxypeptidases, and elastases. Although reverse transcriptase–polymerase chain reaction (RT-PCR) first detects carboxypeptidase A mRNA on day 10.5 of gestation in the mouse and amylase on day 12 (in the rat, carboxypeptidase A and elastase I are detected on days 12 to 13), immunostaining cannot detect amylase before visible differentiation of the acini and ducts at day 14.5. [83] The secretion of amylase and chymotrypsin is detected on day 15 of rat gestation, and that of trypsin, ribonuclease, elastase and lipase on day 18. The secretion of digestive enzymes is insensitive to known secretagogues until after birth, and fetal acini do not respond to intracellular messengers such as phorbol dibutyrate and calcium. Throughout the remainder of rat gestation, the levels of all digestive enzymes rise. After birth, lipase, ribonuclease, and elastase levels fall initially, rising to adult levels after weaning. [84] Trypsin and chymotrypsin mRNA levels rise at parturition, although chymotrypsin levels fall after weaning. Amylase mRNA concentrations do not change appreciably at birth but rise steadily after weaning. Enzymatic activity correlates well with the appearance of the various enzymes, with trypsin activity lagging behind the activities of the other digestive enzymes. [85] Acinar and ductular tissues have been assumed to derive from endodermal precursors. Destruction of exocrine cells with use of the elastase I promoter to drive diphtheria A toxin expression in transgenic mice also results in a decrease in ductular and islet mass. [86]

Functional Development—Endocrine Endocrine cells and specific endocrine gene products can be recognized even before pancreatic morphogenesis or before the appearance of exocrine-specific gene products, suggesting a possible role for these factors in pancreatic development. Somatostatin mRNA can be detected as early as day 7.5 of mouse gestation; insulin and glucagon mRNAs can be found at about day 9, gastrin on day 10, and pancreatic polypeptide on day 10.5. [87] Studies assessing the presence of the corresponding peptides have yielded conflicting results, probably because of the epitope variance in specificity of the antibodies used. However, it appears that the first mature peptides present are insulin and glucagon on day 9.5 of mouse gestation. A pancreatic polypeptide family member, probably peptide YY, is also expressed on day 9.5 and appears to be transiently coexpressed by all four differentiating endocrine cell types as they appear. Glucagon and insulin are frequently coexpressed in the same scattered individual cells from days 9.5 to 12.5 of mouse gestation. After a transient decrease in expression, insulin reappears at much higher concentrations in mature ß-cells around day 18.5 of gestation; glucagon expression reaches adult levels early in gestation, before the appearance of mature a-cells in typical islets. Gastrin is maximally expressed in late gestation and is switched off in the pancreas shortly after birth. [88] Somatostatin is first detected by immunocytochemistry on day 15.5 of mouse gestation; pancreatic polypeptide does not appear until after birth. As mature islets form toward the end of gestation, a sharp spatial segregation of their component cells occurs in rodents. In humans, the segregation of cell types is more subtle. Islet formation begins at gestational week 12; the islets initially derive from pancreatic ducts but lose their contact with the ducts by 17 to 20 weeks. The heterogeneity of endocrine cells in different segments of the mature mammalian pancreas may be secondary to the dual origin of the pancreas from ventral and dorsal buds. [89] The observation that cells expressing pancreatic polypeptide predominate in the lower head region of the mature pancreas suggests an embryological origin from the ventral pancreatic bud. [90] Although the pancreatic endocrine cell had long been thought to derive from the neural crest, [91] more recent data demonstrated that the endocrine cell arises from the

ductular cell, which also gives rise to the acinar cell. Tyrosine hydroxylase, an endocrine marker, is transiently expressed along with insulin and glucagon or both in the pancreatic duct on day 10 of mouse gestation. [92] The embryonic pancreatic duct retains the ability, when grown in vitro, to regenerate into a complete pancreas containing ductular, endocrine, and exocrine cells. [93] Quail-chick marker studies demonstrate that quail ectoderm and chick mesoderm explants grown in culture always produce pancreatic endocrine cells of chick type. [94] Mouse aggregation chimeras demonstrate that ß-cells of a single islet are derived from both chimeric components, suggesting that islets arise by the aggregation of individual cells rather than by the monoclonal expansion of cells. [95] Finally, the Pdx1 transcription factor, which transactivates the insulin and the somatostatin promoters, is expressed in endocrine cells of the pancreas and the duodenum, implying a common precursor for both types of endocrine cells. [96]

Regulation of Pancreatic Endocrine and Exocrine Development

Hormones and growth factors. The control of early development of all three structural components of the pancreas (i.e., acini, ducts, and islets) has long been thought to be dependent on mesenchymal factors. Although efforts in the 1970s to identify specific factors were unsuccessful, [97] more recent studies revealed that pancreatic epithelial rudiments isolated on day 11.5 of mouse gestation develop normal ductular, acinar, and endocrine structures when cultured in the presence of mesenchyme. [98] If the rudiments are placed in vivo under the renal capsule of a syngeneic mouse, no ducts or acini appear, but mature islets with the orientation of central insulin-expressing cells and peripheral glucagon-expressing cells develop. These studies demonstrate that at day 11.5 of mouse gestation, the developmental program for islet development is fully in place. Moreover, islets, as previously described, thought to derive from pancreatic ducts and ductules, can form in the complete absence of pancreatic ductal or mesenchymal tissue. Basement membrane components such as laminin and collagen type IV are important for pancreatic ductal development, whereas mesenchyme, previously thought to be necessary for the differentiation of all pancreatic constituents, is required only for acinar development. In further attempts to identify specific mesenchymal factors that may influence pancreatic differentiation, it was shown that pancreatic epithelial rudiments isolated at 12.5 days of mouse gestation and grown on rat tail collagen gels develop acini and small clusters of endocrine cells. [99] Epidermal growth factor (EGF) increases the ductular portion of the pancreas but decreases acinar tissue and leaves the relative amount of endocrine clusters intact. [100] EGF and its homolog, transforming growth factor-a (TGF-a), are expressed in developing rodent mesenchyme and in acinar and ductular cells; EGF receptors, which bind both factors, are also found on acinar cells and the apical surface of ductal cells. [101] Transgenic overexpression of TGF-a in the mouse pancreas results in ductular hyperplasia and interstitial fibrosis without an increase in acinar cell mass; acinar metaplastic changes of the ductules also occurs, evidenced by amylase expression and the appearance of zymogen and mucin granules. [102], [103] and [104] In contrast, TGF-ß $_1$ inhibits acinar development, enhances islet development, and has no effect on ductular tissue. [105] CCK and gastrin are two structurally and functionally related peptides that bind with different affinities to receptors; CCK has a higher affinity for the CCK-A receptor, gastrin for the CCK-B receptor. They exhibit distinct patterns of tissue localization and play important roles in the regulation of pancreatic growth. Pancreatic hypertrophy can be observed in animals fed a diet rich in soybeans, because the soybean trypsin inhibitor contained within is thought to abolish the usual inhibition of CCK by trypsin. Infusion of CCK or its analog, cerulein, increases pancreatic size, weight, DNA content, and acinar cell mass. [106] The use of specific CCK-A receptor antagonists confirms that CCK stimulates pancreatic growth in a dose-dependent manner, but it is not essential for pancreatic development. [107], [108] A strain of rats congenitally lacking the CCK-A receptor have a smaller pancreas than control rats. [109] As observed with CCK, gastrin induces a rapid rise in pancreatic RNA, DNA, and protein in healthy and in hypophysectomized rats. [110] However, neither endogenous hypergastrinemia nor hypogastrinemia induced by various mechanisms has any affect on rat pancreatic growth [111]; blockade with specific CCK-B antagonists similarly has no observable effect on growth of the rodent pancreas. [112] Gastrin plays more of a role in fetal pancreatic development, because fully processed CCK is expressed only at negligible levels during fetal life, whereas gastrin is exclusively expressed in the fetal pancreas. Hypergastrinemia or the persistence of pancreatic gastrin expression in the neonatal period is associated with nesidioblastosis. [113] Gastrin-overexpressing transgenic mice demonstrate no significant change in pancreatic morphology, but when they are cross-bred with TGF-a–overexpressing mice, their progeny demonstrate an increased islet cell mass and a dramatic reduction in TGF-a–induced ductular metaplasia. [114] Thus, the two peptides appear to modulate pancreatic development coordinately. The pancreatic regenerating (*reg*) gene may play a role in pancreatic growth, particularly in the development of ß-cells. Two nonallelic *reg* genes have been identified in humans, [115] rats, [116] and mice, [117] initially cloned from a cDNA library obtained after partial pancreatectomy. Expression of *reg* is noted after day 9 (*reg* I) or day 12 (*reg* II) in mouse gestation [118] and after 17 weeks of human gestation. Although *reg* expression rises dramatically after partial pancreatectomy, levels also rise with sham laparotomies. Similarly, despite correlation of *reg* expression with ß-cell mass, chronic glucose infusion, which induces ß-cell growth, is not associated with an increase in *reg* expression, [119] and fetal *reg* expression does not correlate well with ß-cell development. Other factors contribute to the regulation of pancreatic growth and development, but more studies are needed to define their precise roles. Secretin, expressed in fetal pancreas, stimulates pancreatic growth and may potentiate the stimulation of pancreatic growth by CCK. [120] The insulin-like growth factors (IGFs) types I and II are also expressed during fetal pancreatic development. [121] They stimulate fetal islet cell growth and are reactivated during pancreatic regeneration. [122] Targeted disruption of one of the alleles for mouse IGF-II results in a global growth-deficient phenotype without a specific pancreatic phenotype. [123] Although glucocorticoids inhibit overall pancreatic growth in adult rats, they increase pancreatic growth in fetal and suckling rats, [124] increase amylase activity in fetal rat pancreas, increase the accumulation of pancreatic enzymes in fetal rat explants, and enhance differentiation of the AR42J pancreatic acinar cell line. [125]

Small Intestine

The small intestine performs highly specialized functions responsible for most digestive and absorptive processes. As such, maturation of these complex functions can be arbitrarily analyzed according to specific nutrient groups.

Carbohydrate Digestion and Absorption Lactase–phlorizin hydrolase (LPH) activity can be detected early in the human fetus and is measurable by 12 weeks of gestation. [126] A significant increase in this activity occurs after the 24th week, and a late gestational surge is observed throughout the 3rd trimester. In rodents, this burst of activity is observed immediately before birth; it declines at weaning in association with a change in the primary carbohydrate sources, as a diet rich in maternal milk lactose is exchanged for a laboratory diet rich in sucrose and starch. Humans demonstrate a high level of LPH activity at birth, but the decline in activity does not correlate well temporally with weaning; humans are the only known species in which LPH activity does not fall in the postweaning period. Paradoxically, disaccharidase activities, such as those of sucrase-isomaltase, maltase, and trehalase, which should be physiologically unnecessary before weaning, are already present after 10 weeks of gestation at 60% to 70% of human adult levels, reaching adult levels by term. [127] In contrast, adult levels of disaccharidase activities in rodents are observed only during postnatal weaning. Sucrase-isomaltase and trehalase activities are undetectable before weaning but rise to adult levels by the 4th week of life. Glucoamylase, a microvillus enzyme, is also detected as early as the 10th week of gestation. Seventy percent of adult levels are achieved by 26 to 34 weeks of gestation. Salivary amylase can be detected after 20 weeks of gestation, and the level increases with fetal age. Proximal-to-distal gradients for LPH and other glucosidase activities can be demonstrated after 17 weeks of gestation, with maximal activities in the proximal jejunum and progressively less of these enzymatic activities along the more distal portions of the small intestine. [128] In all species studied, glucose transporters are present before birth, but fructose transporters appear only in the postnatal period. Uptake of glucose against a concentration gradient is demonstrable in the jejunum and ileum at 11 to 19 weeks of gestation in humans. [129] The longitudinal gradient in the level of glucose transport activity, well-documented in the mature intestine, occurs by 17 to 20 weeks of gestation. Active Na^+-dependent transport can be demonstrated after 19 days of gestation in fetal rat ileum. In the rodent, expression of SGLT1, a Na^+-dependent glucose transporter, remains unchanged after birth. Mutation of the SGLT1 gene results in severe glucose/galactose malabsorption and can result in overwhelming diarrhea in the newborn. [130] The facilitative transporter for glucose, GLUT2, and that for fructose, GLUT5, appear in rat fetal intestine before villus formation. [131] After birth, GLUT1 expression disappears and carbohydrate transporter levels are genetically programmed to match the species' natural diet. [132] The aldohexose transporters, which take up galactose and glucose with various affinities, demonstrate a shift to glucose transport corresponding to a decrease in dietary galactose in the suckling animal. [133] After weaning, fructose transport capacity rises sharply in rats and rabbits but not in cats; their adult diet continues to contain little fructose. Rats prevented from weaning still undergo the same steep rise in fructose uptake capacity, despite continued low fructose intake. [134] It is unclear whether the increase in absorptive capacity for hexose transport after birth reflects an increase in the number of transporters or an increased transporter efficiency.

Protein Digestion and Absorption Pancreatic trypsin and chymotrypsin activities depend on activation by enterokinase, and thus, enterokinase plays an important role in the early developmental stages of protein digestion. Enterokinase activity is only 6% of adult levels at 26 to 30 weeks of gestation and remains at 20% of adult levels even at the time of parturition. [135] Although trypsin can be detected at 20 weeks' gestation, its activity becomes measurable only at 28 weeks, reflecting its activation by enterokinase. Microvillar dipeptidase enzymes, which complete peptide digestion, are detected throughout the length of the small intestine in the 11-week-old fetus. Adult levels of dipeptidase activity have been demonstrated in the 14- to 16-week-old fetus. Concentrations of other microvillar dipeptidases are generally found in a longitudinal gradient, with the highest levels in the proximal intestine. Leucine aminopeptidase is an exception to this general organization, with distal activity twice that of proximal activity in the 16th week of gestation. Brush border and cytosolic peptidases reach adult levels in the neonate. Rat microvillar peptidase activity rises dramatically at weaning. Amino acid transporters also develop prenatally. [136] At birth, levels appear to be essentially equivalent to adult levels. Six different transporters for neutral and charged amino acids have been demonstrated during the 2nd trimester in the human fetus, [137] although the developmental pattern of uptake is not identical for all amino acids. In the fetus and neonate, macromolecular transport plays an important role in the digestion of proteins and lipids. In experimental animals, the small intestine epithelium appears to be more permeable to amino acids and peptides in the immediate postnatal period than in the mature intestine. Macromolecular tracers infused into the amniotic fluid or the intestinal lumen late in gestation are absorbed into enterocytes of humans, monkeys,

guinea pigs, and rats, reflecting a high rate of pinocytosis. [138] This process is extremely active in the first 2 weeks postnatally and decreases dramatically with weaning. The sites of absorption of different proteins vary in the rat. Intact immunoglobulins are transported in the jejunum but not in the ileum. [139] Nutritional proteins undergo pinocytosis in the ileum in a nonspecific manner. Pinocytosis of nutritional proteins and immunoglobulins decreases at weaning, although the adult rat can still absorb small amounts of intact protein. [140] In parallel with pinocytosis, enterocytes exhibit high levels of lysosomal proteases, such as cathepsins and other peptidases, during the first 2 weeks postnatally, but these levels fall thereafter. These intracellular enzymes provide a mechanism for protein digestion before the appearance of pancreatic proteolytic enzymes. Intact proteins also are absorbed in premature and term human infants during the first few months of life. Macromolecules may continue to cross the healthy adult small intestine, but the amounts are low compared to those in the newborn. Imperfect barrier function during the first months of life may play an important role in conferring a tolerance of or sensitivity to dietary proteins.

Lipid Digestion and Absorption The higher rate of fecal fat loss in neonates than in adults correlates with lower lipase and lower intralumenal bile acid concentrations. Lipase has long been detected in pancreatic extracts of 16-week-old human fetuses. Although levels rise significantly during the 3rd trimester, lipase activity in week 32 of gestation remains at only 50% of term levels, which are themselves only 10% of adult levels. The human neonate also has lingual lipase activity, which rises to adult levels by 2 years of age. The relatively low levels of lipase may explain the presence of unhydrolyzed triglycerides in neonate feces. In humans and rats, lingual lipase and maternal milk lipase aid in neonatal fat digestion. [141] Gastric lipase appears as early as 10 to 13 weeks of gestation. [142] Adult distribution of gastric lipase is established by 16 weeks' gestation and appears to be the major determinant of lipolytic activity in gastric aspirates of premature infants. Lingual lipase is produced in the serous glands of the tongue and appears to aid in the digestion of milk triglycerides in neonatal rats, mainly through hydrolysis in the stomach. In the rat, lingual lipase exists in small amounts at birth and increases markedly at weaning. Maternal milk lipase also plays a role in fat digestion. Synthesis of bile acids from cholesterol and conjugation with taurine and glycine can be demonstrated in organ culture in vitro with human liver tissue obtained from fetuses after 15 weeks of gestation. [143] Biliary secretion can be demonstrated as early as the 22nd week of gestation. Bile acid reabsorption occurs in the neonate by passive diffusion throughout the small intestine, but active Na^+-dependent ileal transport of bile acids does not occur until weaning in rats, rabbits, and humans. [144] Both secretion and reabsorption of bile acids are lower in suckling rats than in adults. Bile acid concentrations are initially too low to facilitate the formation of micelles. In the neonatal period, before the maturation of bile acid secretion and reabsorption and the mature production of pancreatic lipase, the suckling rats' small intestine exhibits increased permeability to both triglycerides and cholesterol. [145] The lipoproteins required for chylomicron production are abundant in the small intestine of the suckling rat, and chylomicrons can be formed and presumably transported into lymphatic channels. [146] After weaning, the dietary fat content decreases, pancreatic lipase activity matures, and fewer large lipid particles are seen in the enterocyte. Ileal bile acid absorption also begins at this time, reaching adult capacity after 1 month in the rat. [147] In human neonates, bile acid synthesis occurs at relatively high levels. However, because ileal resorptive mechanisms are not yet mature, a reduction in the bile acid pool results. In premature infants, this reduction is even more severe, and 10% to 20% of fat intake in formula-fed premature infants may not be absorbed. During the first 4 to 6 weeks of human life, intralumenal bile acid levels increase as absorptive mechanisms mature, leading to improved lipid absorption.

Small Intestine Immune System The GI tract, particularly the small intestine, contains a highly complex mixture of immune cell populations. The gut-associated lymphoid tissue (GALT) encompasses organized aggregates dominated by lymphocytes (Peyer patches) and a diffuse heterogeneous population of lymphocytes, monocytes or macrophages, and other cells, such as eosinophils and mast cells in the lamina propria. Intraepithelial lymphocytes are also scattered throughout the surface epithelium. Structures resembling Peyer patches can be demonstrated as early as 11 weeks of human gestation; by 14 weeks, $CD4^+$ and $CD8^+$ lymphocytes can be seen. [148] By the end of the 2nd trimester, Peyer patches histologically resemble the adult structure, indicating that antigen exposure or bacterial colonization is not necessary for their development; however, germinal centers do not form until after birth. [149] In transgenic mice carrying a null mutation for tumor necrosis factor-a (TNF-a), Peyer patches or lymph nodes do not develop, and splenic organization is markedly abnormal; if the 55-kd receptor for TNF-a is disrupted, lymph nodes and splenic tissue develop normally, but Peyer patches are still absent, suggesting that the 55-kd receptor provides specificity for Peyer patch development. [150] Other targeted mutations that preclude Peyer patch development in mice include knock-out of the inhibitory helix-loop-helix transcription factor Id-2, [151] lymphotoxins, [152] and the lymphotoxin-ß receptor. [153] In mice lacking Peyer patches, oral tolerance does not develop. [154] $ß_7$-Integrin–deficient mice have severely impaired GALT formation, [155] affecting the development of Peyer patches, the homing of lymphocytes, and small intestinal immune responses to experimental parasitic infection. [156] Targeted disruption of the homeodomain-containing transcription factor NKX2.3 in mice results in significant defects in intestinal development, as well as smaller Peyer patches and lack of the mucosal addressin cell adhesion molecule-1 (MAdCAM-1), normally responsible for B- and T-cell homing to peripheral lymphoid organs. [157] Lamina propria lymphocytes are first detected after 11 weeks of gestation. During fetal life, they consist of increasing numbers of scattered T and B cells. In contrast to aß T cells, ?d T cells, which make up 5% to 15% of small intestinal and 40% of colonic intraepithelial lymphocytes, can develop extrathymically as well as in the thymus. [158] Soon after birth, ?d T cells undergo clonal expansion, but with further maturation, they become clonally restricted and unique in each individual. Targeted deletion of ?d T cells in mice results in a lack of mucosal IgA-producing T cells but has no effect on aß T-cell development, which is thought to occur intrathymically. [159] IgA- and IgM-producing plasma cells are not found in the lamina propria until after birth and antigenic exposure. Intraepithelial lymphocytes appear at 11 and 12 weeks of gestation. [160] Fetal lamina propria lymphocytes are mostly $CD4^+$, as in the adult lamina propria, whereas fetal intraepithelial lymphocytes are often $CD4^-/CD8^-$; $CD8^+$ cells become more predominant postnatally. [161] Macrophages are present at 12 weeks, but their numbers increase greatly after birth. [162] Unlike mature colonic and ileal villus epithelium, fetal intestinal epithelial cells do not express MHC class II antigen, suggesting that antigen exposure may be important for its induction. In rats, suckling and germ-free animals have fewer intestinal lymphocytes than adults, and weaning, associated with intestinal maturation and increasing bacterial colonization, is also characterized by marked development of the mucosal immune system. [163] Cyclosporine, an inhibitor of T-lymphocyte activation, retards normal lymphocyte development in the small intestine. [164] Natural killer activity of intraepithelial and lamina propria lymphocytes is absent before birth, rising dramatically after weaning. [165]

Vitamins and Minerals Both passive and carrier-mediated absorption is increased in neonatal small intestine. Copper, iron, magnesium, and zinc are absorbed by the suckling rat small intestine in increased amounts, but rates of absorption decline to normal at weaning. Highly specific lactoferrin receptors for milk are present in human fetal and suckling small intestine. [166] Lead, cadmium, radium, plutonium, barium, and other toxic heavy metals are also absorbed more easily in the suckling than in the adult rat. Active transport of calcium occurs throughout the rat small intestine and colon before weaning but depends on mechanisms distinct from those in the mature mucosa, uniform throughout the intestine and not requiring vitamin D, in contrast to vitamin D–dependent uptake mechanisms concentrated in the duodenum that appear after weaning. Human neonates absorb iron, copper, calcium, zinc, and lead more efficiently than do adults. Although the mechanisms remain uncertain, these processes can facilitate lead intoxication. Inadequate bone mineralization is a common problem in premature human infants. This is not thought to result from a lack of absorptive capacity but from an insufficient supply of calcium in maternal or formula milk. Simple calcium supplementation can correct the calcium imbalance. Impaired absorption of fat-soluble vitamins is present in neonates and likely reflects the same pattern of impaired absorption of all lipids. Vitamin B_{12} absorption has been demonstrated in the rat neonate. [167] Intrinsic factor receptor activity is detectable throughout the small intestine and colon of the human fetus after 10 to 19 weeks, but it is present only in the distal ileum at the end of fetal development. [168] Folate absorption is impaired in the human neonate and infant compared with the adult. Biotin transport was found to be higher in the ileum than in the jejunum of suckling rats, equal in both parts of the small intestine in weanlings, and higher in the jejunum than in the ileum of adult rats. [169]

Regulation of Intestinal Maturation

Role of diet. During weaning, the infant GI tract is exposed to a dramatic change in dietary composition. Maternal milk is high in fat and low in carbohydrate, whereas a "conventional" diet includes relatively high amounts of carbohydrate and has a low fat content. The dominant carbohydrate source changes from lactose to a more varied mixture, dominated by sucrose and starch. Many of the maturational changes of the small intestine occur at weaning, leading to the supposition that dietary composition is an important factor contributing to these ontogenic changes. Although this association may be important in the rodent, in humans, dietary factors are not essential to the expression of many functional activities that emerge. LPH exhibits an accelerated surge before parturition, which suggests that the birth process (and possibly hormonal factors related to it), rather than a dietary challenge, regulates expression of this activity. Even in rodents, the importance of dietary components in triggering changes in various activities during weaning may be limited, and similar changes occur at almost the same time in rats prevented from weaning. [170] Premature weaning also elevates glucocorticoid levels, which can result in precocious digestive tract maturation. Other observations suggesting that dietary factors do not play a major role in small intestine development have been obtained through studies of intestinal explants. Despite subsequent implantation into the kidney or a subcutaneous space, or if cultured in vitro, the normal pattern of functional maturation has been observed. [171] Rat ileum that is bypassed surgically at 12 to 14 days of age expresses sucrase and maltase activity in the normal temporal framework. [172] Intestinal explants from 6-day-old rats cultured in the absence of hormones cannot be induced to precocious expression of sucrase-isomaltase or maltase. Although dietary factors may not fundamentally regulate the temporal sequence of GI tract development, they may modulate the process. A high-carbohydrate diet enhances intestinal glucose transport by increasing the number of glucose transporters along the crypt-villus axis. [173] Humans or rats fed a diet high in maltose or sucrose exhibit increased maltase or sucrase-isomaltase activity, but not increased LPH activity. Similarly, feeding lactose increases the level of brush border membrane LPH in rats, and prolonged suckling delays the usual decrease in LPH expression at weaning. [174] However, high levels of LPH mRNA persist after premature weaning. Malnourished suckling rats display delayed patterns of mucosal enzyme development, which can be reversed by refeeding. [175] Experimental studies have shown that bypassed segments of intestine maintain high levels of LPH, whereas the shortened segments of intestine left in continuity exhibit decreased LPH activity. [172] Clinically, a high-protein diet can stimulate greater pancreatic secretion of trypsin and lipase in premature infants. Dietary constituents other than nutrients may also modulate functional maturation. Pig, rabbit, and dog neonates in their first 24 hours after birth exhibit greater increases in small intestine weight, size, and DNA or protein content when fed colostrum than when fed an artificial diet. 173a Rat neonates fed colostrum also have increased small intestine DNA content and synthesis, although their intestinal weight does not vary from that of neonatal rats fed mature milk. 174a High concentrations of EGF occur in human, mouse, and rat milk. 175a EGF is found throughout the lumen of the GI tract of the rat and at

higher levels in suckling than in adult rats, correlating with the milk intake, implicating milk as an important source of EGF in the suckling period. [176] EGF retains biologic activity in the small intestine of suckling rats after ingestion of milk. Milk-borne or orally administered EGF is absorbed and distributed throughout the digestive tract. IGF-I is also found in human milk and colostrum. The peptide is protein-bound but released after treatment with acid. [177] Insulin, too, is found in colostrum and milk, and induces precocious sucrase-isomaltase activity in neonatal mice. Small amounts of glucocorticoids and thyroxine also are present in milk. [178] Corticosterone induces precocious intestinal expression of sucrase-isomaltase and maltase activity in adrenalectomized rats, but at higher concentrations than are found in maternal milk, a level similar to that of 18- to 20-day-old control rats, in which sucrase-isomaltase and maltase activities are normally expressed.

Hormones and peptide growth factors. Although nutrients in the diet may modify some ontogenic changes of the GI tract, lumenal or systemic hormones and peptide growth factors may play a more direct role. Among these factors, glucocorticoids have the best-documented impact on intestinal development. Tissue concentrations of free corticosterone rise 48 hours before the appearance of the enzymatic changes associated with weaning, suggesting that weaning may promote development indirectly by inducing changes in corticosteroid concentration. The administration of glucocorticoids to rats and mice during the suckling period prematurely decreases pinocytosis and levels of lactase and lysosomal hydrolases, and prematurely increases sucrase-isomaltase, maltase, trehalase, peptidase, pancreatic and salivary amylase, pepsinogen, and gastrin receptor levels. Glucocorticoids increase sucrase-isomaltase and LPH mRNA levels in rats, although enzyme activities appear also to be regulated by posttranscriptional events. [179] Age appears to be an important determinant of the response to glucocorticoids because glucocorticoids accelerate the rate of cellular proliferation of the suckling rat but not of the adult rat. In mice, dexamethasone induces maturation during the days immediately preceding weaning. [180] The concentration of glucocorticoid receptors in the intestinal mucosa peaks at the time of weaning in the rat, and hypophysectomy is associated with decreased intestinal sucrase-isomaltase activity in suckling rats; this can be normalized by the administration of cortisone. [181] Thus, during a limited period, glucocorticoids can influence maturation of the small intestine. Enzyme levels are not stimulated by glucocorticoids in rats adrenalectomized after 17 to 18 days of age. [182] Although adrenalectomy slows the rate of enzymatic changes at weaning in the rat, the changes commence at the same time and eventually reach the same levels as in sham-operated controls. [183] These studies suggest that the changes may be genetically programmed and are not glucocorticoid-dependent. Genetic programming would explain the preserved temporal development of sucrase-isomaltase, LPH, maltase, and ß-galactosidase activity in fetal rat and mouse intestine after transplantation into an adult. In humans, prenatal administration of glucocorticoids decreases the incidence of necrotizing enterocolitis in the neonate, presumably by promoting intestinal maturation, including the intestinal mucosal barrier. [184] Glucocorticoids also influence the differentiation of human fetal intestine in organ culture. [185] The neonates of mothers who received glucocorticoids in late pregnancy have a larger bile salt pool than do neonates of the same age whose mothers were not treated. [186] Thyroxine affects intestinal maturation, but interpretation of the mechanism is difficult because thyroxine also increases glucocorticoid levels. [187] Although hypothyroidism prevents or delays intestinal maturation, it also abolishes the developmental rise of corticosterone. The maturational delay in the expected rise in sucrase-isomaltase activity in hypothyroid fetuses can be reversed by the administration of glucocorticoids without thyroxine. Thyroxine and corticosteroids produce a synergistic response in the rat, enhancing the expression of small intestine enzymes before weaning. [188] However, the administration of cortisone results in only partial restoration of reduced jejunal lactase activity after hypophysectomy, whereas thyroxine alone fully restores this decline. Thyroxine administration alone also decreases LPH mRNA levels and increases intestinal alkaline phosphatase, but it has no effect on sucrase-isomaltase mRNA levels in rat small intestine. [189] Colostrum and maternal milk in many animal species, including humans, contain EGF. At weaning, EGF levels in mouse maternal milk decrease, and endogenous sources of EGF increase markedly. [190] EGF may have important trophic effects throughout the GI tract, but the lack of a precise temporal relation of declining maternal milk EGF concentrations to the augmented production from endogenous sources indicates that EGF is unlikely to play an essential role in the regulation of developmental changes. EGF administered orally increases cell growth in the intestine and pancreas of neonatal rats, even though EGF is largely degraded by proteolytic enzymes. [191] EGF in amniotic fluid is taken up in the small intestine by endocytosis in the 20-day-old rat fetus and may play a role in fetal intestinal development. [192] TGF-a appears to be the major ligand for the EGF receptor in fetal development and thus may contribute greatly to fetal GI tract development, until EGF expression increases at the end of gestation. [193] EGF receptors are most abundant at 8 to 12 weeks of gestation in the human fetus. [194] Gastrin also may play a role in GI tract development; it is found in detectable concentrations in fetal plasma and at high levels in neonatal plasma. Antrectomy performed on midgestation fetal sheep induces a 70% decrease in plasma gastrin levels, leading to reduced crypt and villus density, a lower crypt-to-villus ratio in the distal small intestine, and shorter gastric glands. Furthermore, mice bearing deletions of the gastrin [195] or the CCK-B/gastrin receptor genes have decreased numbers of parietal and ECL cells, resulting in atrophy of the corpus mucosa, particularly in the pit and basal regions. Bombesin, also known as *gastrin-releasing peptide (GRP),* stimulates gastric, colonic, and pancreatic cell growth in suckling rats. [196] Parenteral administration of bombesin during the suckling period of rabbits results in hypertrophy of the stomach, small intestine, and colon. A bombesin-like compound has been found in the breast milk of some mammals. [197] However, a physiological role for bombesin in intestinal maturation has not been proved. Insulin administered to suckling mice causes premature maturation of the small intestine. [198] These effects include precocious cessation of the intestinal macromolecular transport and induction of sucrase-isomaltase and other brush border enzyme activity in adrenalectomized suckling rats. Endogenous insulin levels rise during the weaning period in the rat, although no clear role has been established for insulin in the maturation of the GI tract. Prostacyclin prematurely increases sucrase-isomaltase and maltase activity in suckling rats, and these effects occur in the absence of glucocorticoids in adrenalectomized rats. Proglucagon-immunoreactive cells are first detected in the rat gut by day 14 of gestation. Proglucagon is processed to form glucagon or glicentin, oxyntomodulin, and glucagon-like peptide 1 (GLP-1) or GLP-2 in the small intestine and colon. GLP-2 stimulates cell proliferation and inhibits apoptosis in the small intestine and colon. [199] GLP-2 and its receptor are both expressed by day 18 of gestation in the rat, suggesting roles in gut maturation. Other gut peptides present during fetal development include galanin (day 15 of rat gestation), substance P and CGRP (day 18 of rat gestation), and VIP (week 9 of human gestation).

Effect of bacterial colonization. Bacterial colonization of the GI tract of human neonates occurs within a few days of delivery. Gut bacteria have no role in fetal development but may play a role in postnatal development. Disaccharidase activity of germ-free rats is greater than that of control rats after weaning. Introducing the cecal contents of control rats into the small intestine of germ-free rats causes disaccharidase levels to decrease to normal levels. [200] The intestine of the germ-free rat is also considerably thinner, with shallower crypts and a smaller mucosal surface area than those of control rats. The importance of bacterial colonization on intestinal development has been highlighted by the demonstration that colonization of germ-free mice with *Bacteroides thetaiotamicron,* an important constituent of both mouse and human intestinal microflora, induces many transcriptional changes associated with postnatal development— for example, adenosine deaminase, ornithine decarboxylase, the Na^+/glucose cotransporter SGLT1, colipase, and liver fatty acid–binding protein gene expression. [201] Other normal components of intestinal commensal bacteria, including *Bifidobacterium infantis, Escherichia coli* K12, or a “complete” complement of murine microflora, led to distinct transcriptional changes, suggesting that specific bacteria that colonize the intestine after birth play unique roles in postnatal intestinal maturation. After birth, Paneth cells play an important role in intestinal host defense against bacteria. Of interest, cryptdins 4, 5, and 6, lysozyme, and matrilysin, all implicated in mature Paneth cell antimicrobial properties, are expressed on the first postnatal day in mice, before crypt formation or Paneth cell differentiation. [202]

Colon

The functional maturation of the colon has not been studied as extensively as that of the small intestine. LPH is expressed transiently in newborn rat colon. Sucrase-isomaltase activity is found in the colon during fetal life, but disappears before birth. The fetal rat colon is capable of actively transporting glucose and alanine. However, Na^+,K^+-ATPase a and ß subunit mRNA levels are low in the rat colon before birth, increasing to adult levels by postnatal day 25. [203] Dexamethasone, but not aldosterone, induces a dose-dependent increase in Na^+,K^+-ATPase subunit mRNA levels. [204] Administration of glucocorticoids to the fetal rat colon enhances sodium absorption, an effect not seen in the rat small intestine. [205] Calcium, strontium, and magnesium are also absorbed in the colon at much higher rates in suckling than in adult rats.

Human colonic fetal tissue maintained in serum-free organ culture demonstrates no morphologic or enzymatic changes after the administration of hydrocortisone. EGF, however, causes a precocious drop in sucrase-isomaltase, maltase, and alkaline phosphatase activities. [191] TGF-a appears at 18 days of gestation in the distal fetal rat colon, before crypt formation, and by day 20 of gestation at the crypt bases. The proximal colon undergoes more complex structural changes at 20 days, including villi formation and expression of brush border hydrolases. [206] TGF-a is detectable in the lower half of the villi in the proximal colon from days 10 to 24 of postnatal development, during the establishment of adult proximal colonic architecture, and may play a role in the maturation of he distal and proximal colon to adultlike structures. Gastrin may also play a role in fetal colonic development, because the mature amidated peptide is expressed in fetal rat colon. Posttranslational maturation of colonic gastrin ceases in the postnatal period. [207]

Motility The enteric nervous system and muscle layers form early in gestation, but mature patterns of contractile activity and motility are not found until near term, leading to difficulties in enteral feeding of premature infants. The enteric nervous system, developing from neural crest precursors that migrate in a craniocaudal direction, can be detected in the stomach by week 7 of gestation and in the rectum by week 12. [208] In mice, transgenic studies with use of the dopamine ß-hydrolase promoter confirm neural colonization of the ileocecal junction at day 11.5 of gestation, and of the rectum the following day. [209] Studies of quail-chicken chimera reveal that vagal neural crest cells migrate toward the cranial end of the primitive gut and populate the entire bowel to the rectum, while sacral neural crest cells migrate from the caudal end to colonize the distal intestine to the level of the umbilicus. [210] Retroviral tracing methods show that sacral crest cells colonize the distal intestine only after the arrival of the vagal crest cells. [211] Mesenchyme is critical for maturation of the enteric nervous system. For example, laminin binding directs the migration of

neural crest cells to form neuronal processes and ganglia. [212] Axonal and dendritic process formation from crest-derived, colonized neuroblasts continues throughout gestation and postnatally through poorly defined interactions with local microenvironmental factors. Neuroblasts commit to the expression of specific neuropeptides along a specific ontogenic timetable. Cholinergic and serotonergic neurons develop first, followed by adrenergic and finally peptidergic neurons. [213] Hirschsprung disease, a relatively common familial and sporadic disorder occurring in 1 of 5000 live births, is defined by a lack of myenteric and submucosal ganglia affecting a variable length of colon extending proximally from the anus. This disease has become an important model for defining a complex multigenic disorder in which distinct and specific transcriptional and receptor-ligand pathways and interactions important in development result in a single phenotype—megacolon. Two major models of pathogenesis have emerged, accounting for the megacolon phenotype in both mouse models and human disease, although further genetic alterations continue to be elucidated. The first mutation associated with Hirschsprung disease was of the tyrosine kinase c-Ret. [214] Ret gene activation in transgenic mice leads to a loss of enteric neurons and megacolon. [215] GDNF and its other family members, neurturin, persephin, and artemin, bind to one of four glycosyl-phosphatidylinositol–linked receptors, GFRa1 to GFRa4, that activate Ret transduction. [216] Murine and human mutations of GDNF result in a colonic phenotype identical to c-Ret mutations. Furthermore, alterations of Pax3 or Sox10, transcriptional regulators of c-Ret, also demonstrate Hirschsprung-like megacolon. [217] In the Ret/GDNF/GFRa1 mutations, neural crest progenitors do not divide and thus cannot migrate to populate the enteric nervous system, although in humans, some residual Ret activity persists, and aganglionosis is limited to the distal bowel. [218] A second model involves disruption of endothelin-3 and its receptor, endothelin-B or endothelin-converting enzyme, which results from premature differentiation of migrating neural crest cells in fetal development with depletion of these progenitors before colonization of the distal colon, which occurs in a proximal-to-distal distribution. [219] In the latter model, proliferation of the progenitor cells is intact, but later stages of migration and differentiation cause a phenotype similar to the Ret mutation phenotype. Of note, GDNF-deficient mice demonstrate intact development of the interstitial cells of Cajal, which are responsible for normal slow wave activity of the GI tract, suggesting that the presence of enteric neurons or GDNF is not required for the ontogeny of the interstitial cells of Cajal. [220] However, the Kit receptor and its ligand, stem cell factor, are required for development of these pacemaker cells. [221] Most patients with Hirschsprung disease do not demonstrate mutations of the Ret or endothelin-3 pathways, suggesting that other factors play a role in the phenotype of this disorder. Alterations of enteric nervous system development at many different stages may account for the development of Hirschsprung disease, as exemplified by transgenic mice that overexpress *Hoxl-4* [222] or *Hoxa-4* [223] and exhibit aganglionic megacolon. Null mutation of *Hox11L.1* results in hypoganglionosis or neuronal intestinal dysplasia with a similar clinical phenotype. [224] Other genetic alterations may lead to megacolon, further enlightening our understanding of the development of the enteric nervous system. Like enteric nerves, enteric smooth muscle develops in a proximal-to-distal direction. Development of the outer circular muscle layer is detectable within the mesenchymal layer by week 8 of human gestation; the inner longitudinal layer is recognizable 2 to 3 weeks later. [225] However, contraction after electrical stimulation does not occur until the 3rd trimester. In premature infants, contraction pressures achieved at 25 weeks of gestation only approached only 60% of the levels reached at term. The mechanisms required for colonic defecation, including the coordinated contraction of colonic smooth muscle and the appropriate internal and external sphincter responses, are poorly developed until birth. Although 99% of term infants pass their first meconium stool within 48 hours after birth, infants born at 36 weeks' gestation may not pass their first stool until more than 1 week after birth. [226] Disorders of smooth muscle development affecting colonic motility, such as hollow visceral myopathy syndrome and megacystis microcolon hypoperistalsis, are quite rare compared to Hirschsprung disease, and factors important in their pathogenesis remain unknown. [227]

TRANSCRIPTIONAL REGULATION OF DEVELOPMENT

The application of powerful molecular biologic techniques has provided insights into the transcriptional control of development. An understanding of the mechanisms that regulate the transcription of tissue-specific genes allows a clearer understanding of development at its most fundamental level. Patterns of morphologic and functional development reflect the activation or suppression of expression of specific genes. Gene transcription, in turn, is determined by the presence of regulatory proteins (i.e., transcription factors). Transcriptional hierarchies provide a code of messages controlling crucial events in development, from the earliest stages to the terminal differentiation of maturing tissues. *Hox* genes are the most extensively studied class of genes in vertebrate development. In mice and humans, these genes are found in four clusters on different chromosomes and are similar to the Antennapedia/Bithorax cluster of genes in *Drosophila*. The latter are known to play homologous roles in differentiation and segmentation along the anteroposterior embryonal axis in insects. [228] More than 20 *Hox* genes are thought to play a role in GI tract development. [229] Targeted deletions of the *Hox* genes and other developmental genes in mice have shown that these genes are critical for many developmental programs. Some of these mutations are lethal in mice, and thus may not reveal multiple roles for specific transcription factors that may occur at later stages of development.

Within the GI tract and its appendages, a paradigm for transcriptional control of development was elucidated first for the liver, where a transcriptional hierarchy is essential for hepatic organogenesis and differentiation. [230] The hepatocyte nuclear factor 1 (HNF-1) family, C/EBP (CCAAT/enhancer-binding protein), the HNF-3 family, and HNF-4 have all been shown to be crucial for the expression of liver-specific genes and the development of the hepatocyte phenotype. Many of these factors are also expressed in the pancreas and intestine and appear to play similar roles in the development of these organs. For example, the a-amylase promoter contains binding sites for HNF-3ß or HNF-3? required for efficient expression of this pancreas-specific gene. In a null mutation of murine HNF-3ß, initial endoderm differentiation occurs, but no foregut structures form, and specific pancreatic transcription factors are underexpressed (Pax6) or undetectable (Isl1) (see section "Pancreas"). In contrast, hindgut formation is relatively normal in many mutant embryos. [231], [232] Although new data continuously provide a more integrated view of transcriptional control, the findings summarized in the following text demonstrate the power of available techniques to define the genetic control of gastrointestinal development.

Pancreas

The significance of transcription factors in the initiation of pancreatic differentiation and further commitment to endocrine, exocrine, and ductular epithelium has been appreciated. In addition, different factors signal dorsal compared to ventral pancreatic bud development. Sonic hedgehog (Shh) and Indian hedgehog (Ihh) are expressed early in both mouse and human intestinal and stomach development, in the lateral prospective intestinal part of the epithelium, not in prospective pancreatic tissue. When Shh is overexpressed in transgenic mice, pancreatic differentiation is impaired and intestinal development is enhanced. [233] Although, as previously discussed, pancreatic endocrine cells arise independently of neural crest cells, the two cell types share many developmental similarities, including expression of similar peptides and transcription factors. Furthermore, the region of the intestine fated to form the dorsal pancreatic bud is initially in direct contact with the notochord; notochord-derived fibroblast growth factor 2 (FGF-2) and activin-ß repress Shh and Ihh expression in this primitive region of the intestine, an essential event for early pancreatic differentiation. [234] The ventral pancreatic bud is never in contact with the notochord, and thus its early differentiation appears to be notochord-independent. Targeted deletion of the homeobox gene *Hlxb9* selectively blocks dorsal, but not ventral, pancreatic development. [235] Isl1, expressed in all adult islet cells, [236] first appears at 9 days of mouse gestation, immediately before the appearance of insulin- and glucagon-expressing endocrine cells. Isl1 is also expressed later in somatostatin and pancreatic polypeptide–expressing cells, immediately before expression of these peptides. Mesenchyme adjacent to the dorsal, but not ventral, bud of the primitive pancreas also expresses Isl1. In mice with targeted disruption of the Isl1 gene, ventral pancreatic epithelium and adjacent mesenchyme appear normal at 9.5 days of gestation. [237] In the dorsal bud, however, there is a complete lack of adjacent mesenchyme, and dorsal endocrine cells fail to appear, suggesting that Isl1 is required for dorsal mesenchyme and endocrine cell development. Isl1 is important for maximal expression of Pdx1. In turn, BETA2 (ß-cell E-box transactivator 2), a basic helix-loop-helix transcription factor, activates Isl1 expression. [238] BETA2 dimerizes with the ubiquitous E47 basic helix-loop-helix factor to transactivate the insulin promoter. [239] BETA3, homologous to BETA2, inhibits binding of BETA2 to the insulin E-box enhancer. [240]

Pdx1, or Ipf1, is a homolog of *Xenopus* XlHbox 8, a homeobox gene that plays a key role in *Xenopus* pancreatic and duodenal development. [241] Initially cloned as an activator of the insulin and somatostatin promoters, murine Pdx1 is first expressed in the dorsal endoderm of the primitive gut while it is still an open tube at 8.5 days of gestation. By day 9.5, Pdx1 expression is associated with the emerging dorsal and ventral pancreatic buds as well as with the duodenal endoderm between them. [242] Pdx1 expression is subsequently down-regulated but reappears in differentiating ß-cells during later gestation. Expression is maintained in adult mouse duodenal epithelial cells and islet ß-cells, where it transactivates the insulin gene. Mice with targeted deletion of *Pdx1,* as well as humans not expressing *Pdx1,* lack a pancreas and die soon after birth with elevated urinary glucose levels. [243] Closer examination of the *Pdx1*-deficient mice reveals that the initial stages of dorsal bud formation occur along with the appearance of insulin- and glucagon-expressing cells at the appropriate time. [244] The adjacent mesenchyme develops normally and is capable of directing normal development of pancreatic rudiments at the same stage of arrested development of the mutated mice, suggesting that development of the pancreatic mesenchyme is independent of Pdx1 expression and development of adjacent pancreatic epithelium. Pdx1 acts downstream of the initial specification of gut endoderm to a pancreatic fate. Following these initial changes, Pdx1 is required for the expression of genes that allow the pancreatic epithelial cells to grow, branch, and differentiate. The lack of Pdx1 in insulin- and glucagon-expressing cells indicates that the endocrine cell phenotype is not dependent on Pdx1, although late terminal differentiation of ß-cells and the maintenance of insulin gene expression in adulthood is Pdx1-dependent.

Following Pdx1-mediated stages of differentiation, Isl1, NeuroD/ß2, Pax4, Pax6, and NKX2.2 are expressed in both developing pancreatic and neuronal tissues, and are important for pancreatic endocrine commitment. [245] Parallel to neuronal development, pancreatic endocrine differentiation occurs through the notch signaling

pathway. Mice deficient for Dll1(delta-like 1), the intracellular mediator RBP-J?, or Hes (hairy and enhancer of split), as well as mice overexpressing ngn3 (neurogenin3) or Notch3, which repress notch signaling, demonstrate premature differentiation of pancreatic endocrine cells and depletion of pancreatic precursor cells. [246], [247] and [248] Moreover, targeted deletion of *ngn3* results in a pancreas without any endocrine cells, [249] further implicating the notch signaling pathway in pancreatic endocrine differentiation.

Exocrine cell differentiation is also controlled at the transcriptional level. Analysis of the elastase I promoter has identified a complex enhancer region and unique transcriptional controls in both exocrine and endocrine differentiation. In transgenic mice, the transcriptional enhancer of the rat elastase I gene lies proximal (-72 to -205) to the transcriptional start site and consists of three principal *cis*-regulatory elements. The A element, common to many exocrine pancreatic gene promoters, binds an acinar-specific transcription factor, pancreas transcription factor 1 (PTF1), and is itself sufficient to direct acinar-specific transcription of the elastase I gene. [250] PTF1, first expressed at 15 days of mouse gestation, may be responsible for the high level of acinar transcription of multiple genes at this stage of development. The B element of the elastase I promoter enhances the level of elastase I gene transcription in concert with the A element, but it is insufficient to activate transcription by itself through the binding of an unknown B element binding factor. The C element is required for optimal transcription of elastase I in acinar cells, but like the B element, it cannot activate transcription alone. [251] Future analyses of exocrine cell–specific genes will further elaborate the mechanisms of transcriptional control of pancreatic exocrine development.

Small Intestine and Colon

Transcriptional hierarchies responsible for tissue- and cell-specific expression exist along the length of the entire GI tract. As described above, normal gut tube formation is dependent on reciprocal interactions between the endoderm and the mesoderm. [252] HNF-3ß, which plays an important role in early gut endoderm formation, is capable of inducing transcription of *Shh,* a segment polarity gene, expressed in the earliest stages of chick gut formation in the primitive endoderm. [253] *Shh,* in turn, induces expression of members of the Abd-B class of *Hox* genes during early hindgut formation, perhaps through the induction of mesodermal *Bmp4,* a member of the TGF-ß superfamily. [254] *Hox* gene expression then plays a critical role in the formation of gastrointestinal morphologic boundaries. This is illustrated by mice carrying the null mutation for *Hoxd12* or *Hoxd13,* both of which demonstrate disorganization of the anorectal region, manifest by decreased rectal smooth muscle formation and defective morphogenesis of the internal anal sphincter. Transgenic mice overexpressing *Hoxl4* exhibit altered formation of hindgut structures, leading to the appearance of megacolon. [255] Mice with disruption of the *Hoxc4* gene die soon after birth secondary to complete blockage of the esophageal lumen and disorganization of the esophageal musculature. [256] Deletion of *Hoxa5* results in normal morphogenesis but delayed functional maturation of the small intestine, exhibited by lower brush border hydrolase expression at the time of weaning. [257] Gene deletions of other *Hox* family members should provide more insights into the role of these factors in gastrointestinal regionalization and morphogenesis.

Homeobox genes lying outside the *Hox* gene clusters also appear to be important in the regulation of differentiation of the digestive tract. Homologs of the *Drosophila* caudal gene likely play important roles in intestinal development. These genes, designated *Cdx1* and *Cdx2* in the mouse, are expressed early in development when endoderm differentiates into columnar epithelium within nascent villi. [258] *Cdx2* expression is observed proximally by day 9.5 of gestation in the mouse, *Cdx1* not until day 12.5 in the distal hindgut. [259] Postnatally, *Cdx2* is strongly expressed at the crypt-villus junction and into the villus, whereas *Cdx1* becomes more restricted to the proliferative area of the crypt, suggesting roles for these transcription factors in the patterning of the vertical intestinal axis. Inactivation of *Cdx2* in mice is embryonically lethal in homozygotes; heterozygotes demonstrate polypoid lesions predominantly in the proximal colon, which is the region of peak overlap between *Cdx1* and *Cdx2* expression. These lesions are predominantly hamartomas [260] or lesions composed of heterotopic stomach and small intestinal mucosa, [261] suggesting important roles for *Cdx2* in regional cellular differentiation. *Cdx1* homozygous null mice have grossly normal GI tracts, although the intestinal phenotype has not yet been studied in detail. [262]

Examination of the regulation of cell-specific gene products should elucidate the molecular mechanisms controlling development in the intestine. Of the multiple cell types found in the intestine, study of enterocyte-specific gene products, including sucrase-isomaltase, LPH, and the fatty acid–binding proteins, has yielded the first insights into transcriptional control. The *cis*-regulatory elements required for intestine-specific transcription are often found proximal to the transcriptional start site, in a manner similar to that for the elastase I gene. For example, the nucleotide sequence -103 to +28 of the rat intestinal fatty acid–binding protein is sufficient to limit expression to the enterocyte, [263] although elements outside this region do further modulate expression.

The sucrase-isomaltase gene is expressed along a vertical and horizontal gradient. [264] Studies using transgenic mice in which various regions of the sucrase-isomaltase promoter direct the growth hormone reporter gene have revealed that the sequence -8500 to +54 of the promoter directs transcription to all four intestinal cell types. Using -3524 to +54 limits transcription to enterocytes and endocrine cells, whereas using only -201 to +54 limits transcription almost exclusively to enterocytes. [265] Further analysis of the sucrase-isomaltase promoter in transient transfection assays revealed specific regulatory elements. [266] The SIF2 (sucrase-isomaltase footprint 1) and SIF3 elements are positive regulatory elements and bind HNF-1a and HNF-1ß with different affinities. [267] An increase in HNF-1a binding to the SIF3 element compared to HNF-1ß coincides with a marked induction of sucrase-isomaltase at weaning in mice. [268] The SIF1 element, a 22-nucleotide element located just upstream of the transcriptional start site, binds Cdx2. [269] Cdx2 transactivates sucrase-isomaltase and LPH transcription, [270] binds to multiple intestine-specific genes, [271] and induces enterocyte and goblet cell differentiation of undifferentiated small intestine epithelial IEC-6 cells, suggesting that Cdx2 plays a role in the initiation of intestinal differentiation in addition to maintaining the enterocyte phenotype. Cdx2 and HNF-1a act synergistically through direct protein-protein interaction to activate expression of the LPH gene by binding to their closely linked regulatory elements in the LPH promoter in the intestinal Caco-2 cell line. [272]

Laminin-1 stimulates small intestine villus differentiation through induction of Cdx2 expression. [273] Careful examination of the promoters of other intestine-specific genes, such as the trefoil peptides for mucus-expressing cells, cryptdin for the Paneth cells, and the various peptides expressed by enteroendocrine cells, should similarly provide new insight into gastrointestinal development. For example, keratinocyte growth factor (KGF) promotes goblet cell differentiation in a colonic cell line through the induction of a novel transcription factor, the goblet cell silencer inhibitor–binding protein (GCSI-BP), which may play an important role in intestinal goblet cell differentiation. [274]

As discussed previously, Pdx1 is important in islet cell development of the pancreas and enteroendocrine cell development of the upper small intestine. When Pdx1 is transfected into the rat small intestine cell line IEC-6, the cells become capable of producing multiple enteroendocrine peptides, including serotonin, CCK, gastrin, and somatostatin. Thus, the homeobox Pdx1 gene promotes enteroendocrine differentiation in enterocytes. [275]

The complexity of the transcriptional regulation of intestinal development has been further underscored through the analysis of factors controlling the expression of genes encoding regulatory factors such as Cdx1 and Cdx2. For example, the Wnt/ß-catenin signaling pathway induces expression of Cdx1 in rodent embryonic stem cells and endoderm. [276] TCF-4, a member of the TCF/LEF-1 (T-cell factor/lymphocyte enhancer factor 1) family of transcription factors, mediates the ß-catenin transactivation of the Cdx1 gene through several TCF-binding motifs located in the Cdx1 promoter. Mice homozygous for a targeted deletion of *Tcf4* reveal a decreased number of villi, a lower number of intervillus proliferating epithelial cells from which postnatal crypts derive, and a complete lack of enteroendocrine cells. [277]

The forkhead family of transcription factors is also expressed in the developing intestine in both the epithelium and surrounding mesenchyme. Mice with targeted deletions of the *Fkh6* mesenchymal gene demonstrate precocious fetal crypt development, particularly in the proximal small intestine. [278] Postnatally, crypts are enlarged, disorganized, and branched and form cell-lined cysts, suggesting that Fkh6 inhibits crypt cell proliferation. Although all four cell lineages are present in *Fkh6* mutant mice, there is a selective increase in the goblet cell population in the duodenum and jejunum. Other members of the forkhead family that are expressed in developing intestine include *HFH1* and *HNF3ß,* expressed in both the mesenchyme and epithelium of the fetus but restricted to proliferating crypt cells in the adult mouse, suggesting roles in intestinal development and proliferation. [279] *Nkx2-3* is similarly expressed in proliferating crypts, and targeted deletion resembles the *Fkh6* knock-out except that goblet cell numbers are not increased. [280] Bone morphogenetic proteins (BMPs) also have been implicated in intestinal development [281]; *Nkx2-3* knock-out mice are associated with markedly reduced expression of antimitotic *BMP2* and *BMP4*. [280]

The inhibitory transcription factors, Ids, are dominant-negative basic helix-loop-helix proteins that antagonize other helix-loop-helix transcription factors. Ectopic expression of Id-1 in transgenic mice with use of the fatty acid–binding protein promoter results in a subset of transgenic mice that manifest intestinal adenomas and Paneth cell loss. [282] Interestingly, transgenic mice heterozygous for a mutant allele of the adenomatous polyposis coli gene, Apc^{min}, develop multiple intestinal adenomas that may be secondary to altered phospholipase A_2 expression from Paneth cells. [283] However, diphtheria toxin A–mediated Paneth cell ablation in mice does not lead to the development of intestinal adenomas, suggesting complex interactions between Paneth cells and growth control in intestinal crypts. Mice

homozygous for targeted disruption of *Id1* demonstrate no obvious intestinal phenotype. [284]

GROWTH AND DIFFERENTIATION IN THE MATURE GASTROINTESTINAL TRACT

Cellular Components

Undifferentiated stem cells are present throughout the GI tract. In the small intestine, they give rise first to committed stem cells [285] and then to four distinct cell lineages: the Paneth cell, which migrates downward; the goblet; the enteroendocrine; and the most abundant columnar absorptive cell. The latter three migrate up the villus. The committed stem cells retain proliferative activity and reside in the middle zone of the crypt. By the 2nd postnatal week in rats, each crypt appears to be a monoclonal population derived from a single stem cell anchored three to five cell positions from the base of the crypt. Although there are no markers for stem cells, indirect evidence and mathematical modeling suggest that in the small intestine, stem cells are located at the third to fifth cell positions, whereas in the colon they are located at the first to second cell positions. Transplanted isolated crypts in immunocompromised mice develop all four cell lineages; however, these experiments have not yet led to purification of the elusive stem cell. [286]

An analogous organization may be present at other sites along the length of the GI tract. In the stomach, neck cells located in the middle third of the gastric glands serve as stem cells for the mucus-secreting cells in the pit, pepsinogen-secreting chief cells at the base of the gland, and parietal, enteroendocrine, and caveolated cells throughout the pit-gland gastric unit. The esophageal epithelium forms three successive layers—the basal layer (cuboidal cells and the putative stem cell), the intermediate layer (prickle cells), and the functional layer (stratified squamous cells). [287] In the small intestine, the adult villus columnar cell undergoes significantly greater differentiation than its fetal counterpart. In early fetal life, columnar cells contain short microvilli and large glycogen deposits. During later fetal life, the organelles of the columnar cells differentiate, with the appearance and subsequent disappearance of the meconium corpuscle system and the apical tubular system. Columnar cells exhibit still further morphologic evolution during continued gestation, including a reduction in glycogen content, the appearance of smooth endoplasmic reticulum, and the development of tall, uniform microvilli. [288] The mature villus cell is a highly polarized cell with a well-organized complex of endosomal compartments and markedly different apical and basolateral surface membranes. A gradient of functional differentiation is present in enterocytes along the vertical axis of the villus.

In the adult GI tract, the presence of certain secretory products has led to the identification of more than ten distinct enteroendocrine cell types. In the stomach, the mucous neck cell is thought to be the stem cell for the enteroendocrine cell. The primary enteroendocrine cells in the gastric antrum are the gastrin (G) cell, which secretes gastrin, and the D cell, which secretes somatostatin. The gastric fundus also contains the A cell, which secretes enteroglucagon, and the ECL cell, which secretes serotonin and histamine. The small intestine contains enteroendocrine cells that migrate toward the villus tip, differentiating during this migration, as evidenced by an increase in the number of cytoplasmic granules and a progressive loss of the ability to divide. Studies using transgenic mice that express intestine-specific transgene products because of the presence of villus-specific intestinal fatty acid–binding protein promoters have demonstrated the fundamental heterogeneity of enteroendocrine cell populations. [289] Transgenic mice containing the 5' flanking region of the rat liver fatty acid–binding protein ligated to growth hormone reporter gene demonstrate expression in most CCK-, GIP-, secretin-, and glucagon-producing small intestine cells, as well as 30% of serotonin- and substance P–expressing cells. [290] These mice demonstrate two populations of colonic enteroendocrine cells: one group expressing peptide YY, neurotensin, glucagon, and CCK, and another group expressing serotonin/substance P. A 1.6-kilobase region of the rat secretin promoter directing the growth hormone reporter reveals a close association between secretin- and CCK-expressing cells in the small intestine and secretin/neurotensin and glucagon/peptide YY cells in the colon.

Goblet cells express specific *MUC2* and *MUC3* genes in the small and large intestines, [291] and provide the structural backbone for the unstirred, continuous layer of mucus that lines the intestine. These cells also express intestinal trefoil factor (ITF/TFF3), [292] a lumenally stable peptide that protects against gastrointestinal injury. [293] , [294] The number of goblet cells increases from the duodenum to the ileum; they respectively constitute 4%, 6%, and 12% of all epithelial cells. [295]

Paneth cells, which play a role in host defense, are the only cell lineage deriving from the crypt stem cells that migrate downward toward the crypt base. Each mouse crypt contains 30 to 50 mature Paneth cells, the longest-lived lineage, surviving for 18 to 23 days before being phagocytosed by neighboring cells. Products of Paneth cells include lysozyme, phospholipase A_2, [296] TNF-a, guanylin, and cryptdins, homologs of leukocyte defensins that possess antibacterial activity. [297] Although morphologically indistinguishable, Paneth cells are heterogeneous along the longitudinal axis of the intestinal tract, based on the specific cryptdins they express. [298]

Tuft cells, first described in mouse gastric mucosa in 1955, are widely distributed in the GI tract. [299] Recognized by distinctive caveolae invaginating the apical surface between individual long microvilli, they appear to be highly differentiated cells that, in the small intestine, arise from stem cells in the lower portion of the crypts. They migrate upward and are extruded from the villus tip. In mouse colon, tuft cells have an estimated turnover time of 8.2 days. Although their function is unclear, tuft cells contain an extensive vesicular network, consistent with a role in absorption.

The intestinal mucosa also contains the distinctive M cell, which is restricted to the dome epithelium overlying the lymphoid follicles of Peyer patches in the ileum and other lymphoid follicles in the more distal gut. [300] These cells appear to be important in sampling the antigen milieu in the lumen. Ironically, they also appear to be the avenue of infection for several viral agents (e.g., reoviruses) that can breach the mucosal barrier. Cells resembling adult M cells are first identified overlying lymphocyte aggregates in the distal small intestine in the 17th week of gestation, and their appearance is temporally related to the emergence of these lymphoid aggregates. The surface membrane of the M cell is characterized by microfolds instead of microvilli, and by abundant vesicles, which may facilitate the transport of lumenal antigens, microorganisms, and other macromolecules to the underlying lymphoid tissue. [301] M cells appear flattened because of the underlying accumulation of lymphoid cells in the connective tissue. [302]

Cell Kinetics

The epithelium of the GI tract undergoes rapid turnover in which cell production in zones of proliferation is usually balanced by apoptosis and cell extrusion, creating a steady state under normal conditions. True growth occurs when the rate of cell proliferation is greater than the rate of cell loss. Only selective populations of cells in the GI tract retain proliferative capability. The compartmentalization of proliferating populations and the zonal concentration of the nonproliferating cells committed to terminal differentiation are features shared by all regions of the lumenal GI tract.

The kinetics of cell proliferation has been largely defined through analysis of labeled mitotic frequencies after the administration of ^{3}H-thymidine to identify the fractional mitotic index. This analysis allows a determination of the duration of the constituent component phases of the cell cycle: S phase, or DNA synthesis; G_1, a premitotic latent period; M, a period of mitosis; and G_2, a pre-DNA synthesis latent period (Table 23-1). Pulse labeling of mucosal explants further permits an estimation of mucosal turnover rates as cohorts of labeled cells complete mitosis and emerge from the zone of proliferation to traverse the vertical distance to the surface mucosa before subsequent loss.

TABLE 23-1 Summary of Kinetic Parameters in Human Gastrointestinal Mucosa

Esophagus As stratified squamous epithelium, the esophageal mucosa differs from that of the rest of the intestinal tract. However, as elsewhere in the GI tract, proliferation in the esophagus occurs in the basal layer. Turnover of the esophageal squamous epithelium takes 4 to 5 days in rodents and longer in humans. [303] After cell division, one daughter cell may remain in the basal layer, one may migrate toward the esophageal lumen, or both may migrate. The life span of the basal cell in the esophagus is 80 hours in rats and mice and approaches 8 days in humans. [304] The rate of cellular proliferation in the esophagus is about one-third that in the jejunum. [305] This squamous epithelium is penetrated at irregular intervals by papillae of the lamina propria. Mucous glands similar to those found in the cardia of the stomach are found at the level of the cricoid cartilage beneath the epithelium. Having no secretory or absorptive functions, the esophageal mucosa serves mainly in a

protective capacity and has no specialized cells.

Stomach The stomach serves mostly as a secretory organ and contains many more specialized cell types than the esophagus. The body and fundus are lined by an epithelium that includes mucus-secreting cells extending into gastric pits, with long glands extending deep into the gastric mucosa. These glands are also lined by parietal cells, which secrete acid and intrinsic factor, and chief cells, which secrete pepsinogen. The mucous neck cells lie in the middle third of the gastric glands. The gastric antrum contains deeper gastric pits than the rest of the stomach, and these are lined in part by cells that secrete an alkaline mucus. Enteroendocrine cells are predominantly found in the middle portion of the gastric glands among the mucus-secreting cells in the antrum, body, and fundus. The pattern of expression of a transgene containing promoter sequences of the fatty acid–binding protein gene (*FABP*), a gene used as a marker of gastrointestinal differentiation, in the stomach suggests that mixed surface mucous cell populations exist in the gastric pits, unlike the coherent vertical bands found in small intestine villi. [306] These findings imply that a single isthmus contains more than one stem cell or that migration and differentiation are not as rigorously coupled as they are in the small intestine. The mucous neck cells, the principal proliferating cells of the stomach, migrate to the lumenal surface, where most differentiate into mucus-secreting cells. The cell population above the zone of proliferation for the gastric mucous–epithelial cell is replaced every 2 to 3 days in humans. Migration rates from the proliferative zone in the glandular neck to the surface are comparable to those in the small intestine and colon. [307] During development in the first 4 postpartum weeks, rates of DNA synthesis are markedly higher while fractional cell loss is lower, leading to significant mucosal growth. Parietal cells are incapable of dividing and are replaced by migrating cells that differentiate. [308] They appear to have a long life span (e.g., 90 days in the mouse). Most G cells located in the antrum are derived from mitosis of other G cells, although some may arise from the differentiation of other gastric cells. [309] G-cell turnover varies widely among animal species but is as long as 2 to 4 months in mice.

Small Intestine The small intestine mucosa is composed of villi, which project into the lumen, and the surrounding crypts of Lieberkuhn, which project away from the lumenal surface. A single layer of columnar cells lines the crypts and villi. [310] As they migrate, the columnar cells differentiate into the other small intestine cell types. Enteroendocrine cells also migrate toward the villus tip. Mature goblet cells are located in the upper crypts and villi, and immature oligomucous cells are limited to the crypts. Paneth cells are located at the base of the crypts. Transitional cells with features common to columnar and goblet cells are also found in the crypts. At birth, the progeny of several uncommitted stem cells populate the crypts. However, by the end of second postnatal week in mice, a selection process of unknown mechanism, termed *purification,* enables a single stem cell to dominate, and the cell population in a single crypt is clonal in origin. [311] Progeny from that crypt migrate up adjacent villi (Fig. 23-8). Because four to ten crypts surround most villi, villi are composed of linear stripes of cells of different lineage. This may be especially true in the context of X-linked genes in females, which undergo random X inactivation, creating mosaicism for those gene products.

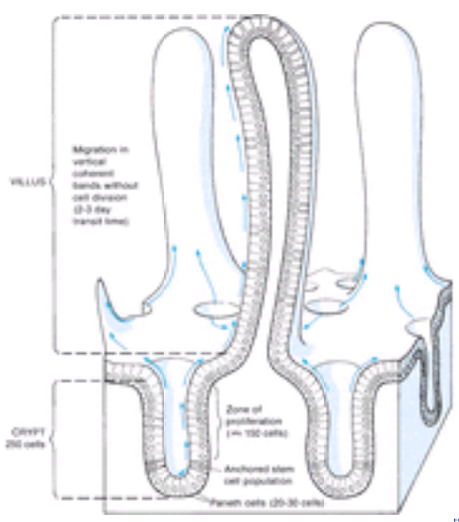

FIGURE 23-8. Schematic representation of the crypt-villus relation in the adult mouse small intestine. Each villus base is surrounded by 6 to 14 crypts, fewer proximally and more distally in the ileum (i.e., a horizontal gradient). The lower five cell positions contain a total of 40 to 50 cells that have an average cycle time of 26 hours and contain 20 to 30 nonproliferative Paneth cells. Anchored stem cells exist at the fifth position, with maximal rates of proliferation. The Paneth cells migrate from this position toward the villus tip and the crypt base. The upper portion of the crypt contains proliferating cells that undergo upward migration; 275 cells are delivered to the villus base from each crypt. This migration occurs in strict vertical, coherent bands toward the villus tip, where the cells are extruded.

In the organization of the intestinal crypt, true stem cells are present at a fixed position in the middle-to-lower region of the crypt cell, and a zone of clustered, actively proliferating progeny is just above in the midzone of the crypt. Clonal analysis reveals intermediate committed stem cells of two types: short-lived stem cells giving rise to one or two cell types and long-lived cells giving rise to all four cell types. [312] Columnar cell migration from the crypt base to the villus tip takes 5 to 6 days in the human proximal small intestine and 3 days in the human ileum. On average, these cells migrate one to two cell positions per hour. [313] As cells exit the crypt and enter the villus, they stop cycling and become trapped in the G_1 phase of the cell cycle as a result of down-regulation of cyclin D_1 and cyclin-dependent kinase 2. [314] After 2 to 3 days, they reach the villus tip, where, at least in the mouse, they shed at a rate of 1400 cells per villus per day. However, as ^{3}H-thymidine labeling has shown, a few scattered cells may remain for weeks. The shorter migration time in the distal than in the proximal small intestine may be secondary to the proximal-to-distal villus height–crypt depth gradient (i.e., the progressive decline in crypt depth and villus size through the length of the small intestine). Apoptosis, or programmed cell death, plays an important role in determining the architecture of intestinal epithelia. Two major apoptotic pathways have been identified. In the crypt, at the level of the stem and early committed stem cells, spontaneous apoptosis occurs at a low rate and regulates the number of cells entering the crypt-villus axis. [315] Mice bearing targeted deletion of *Bcl2* demonstrate increased levels of this spontaneous apoptosis in the crypt base of the colon, but not the small intestine. [316] In contrast, *Bax*-deficient and *p53*-deficient mice have normal levels of spontaneous crypt apoptosis. [317], [318] The trefoil peptide, ITF, inhibits crypt cell apoptosis through transactivation of the EGF receptor and phosphatidylinosotol 3-kinase (PI_3-K)-AKT pathways. [319], [320] Another pathway for spontaneous apoptosis in intestinal epithelia takes place at the villus tip, although apoptotic morphology is only rarely seen during the extrusion process. [321] This pathway is induced by activation of capsase 3, [322] but less is known about the role of specific apoptotic factors in the extrusion process. The cell cycle of the human small intestine basal crypt cell takes about 10 hours. Great variability exists, however; those cells that survive for weeks remain in a prolonged G_2 or G_0 (i.e., resting) phase. Neuroendocrine cells have cell cycles that last for several days, and Paneth cells turn over every 23.3 days, apparently being cleared from the crypt base by phagocytosis.

Colon The colon has a flat surface without villi, and cells appear to be extruded directly from the flat surface onto the bowel lumen. The proliferative zone is found in the basal half of the colonic crypts. Overall, the proliferative capacity of colonic crypts is comparable to that of the small intestine. However, proliferative activity appears to be greater in the distal colon than in the more proximal bowel. Crypts in the ascending colon have been estimated to contain 100 proliferating cells and produce six cells per hour, and the distal colonic crypts to contain 200 cells, producing 21 cells per hour. [323] Less is known about the organization of the stem cells and the zones of proliferation and differentiation in the colon than in the small intestine. However, observations of mice and colon tumors support a similar arrangement for colon and small intestine: an anchored stem cell giving rise to clonal populations of cells in a single crypt and the presence of a larger number of committed but proliferatively competent cells in the midportion of the crypt. The latter are presumed to give rise to the several cell types found in the normal colonic epithelium, most notably columnar absorptive cells and goblet cells. This suggestion is supported by the demonstration of clonality in colonic crypts and colonic tumors with the use of techniques to identify mosaicism of expression of X-linked markers in females and restriction fragment length polymorphism analysis. [324] The validity of the analogy is also supported circumstantially by the close ontogenetic similarity between colonic and intestinal epithelia with the identification of several colon cancer–derived cell lines (e.g., Caco-2, HT-29) that recapitulate the entire spectrum of small intestine epithelial cellular constituents when their growth conditions are properly manipulated. The proliferative zone is located in the bottom of the colonic crypt, with migration to the lumenal surface taking 3 to 8 days in humans and 2 to 3 days in rodents. [325] Great variability exists in the size and shape of the colon crypts along the length of the colon, and in the duration of the cell cycles of the proliferating crypt cell. [326] About 15% to 20% of cells in the crypt appear to be involved in DNA synthesis at any one time. In humans, the cell cycle duration varies from 58 hours in the descending colon to 25 hours in the cecum, with this variability a consequence of variability in the G_1 phase of the cell cycle. Cells migrate at somewhat less than one cell position per hour. Enteroendocrine cells undergo cell renewal more slowly, and populations appear to turn over in 35- to 100-day intervals. [327]

Cell Kinetics in Gastrointestinal Disorders Alterations of cell kinetics in a selected spectrum of gastrointestinal disorders provide insights into the mechanisms regulating cell growth. Inflammatory or destructive lesions of the intestinal tract induce proliferation or atrophy of the mucosa. In the esophagus, injury resulting from reflux esophagitis is characterized by a thickened basal layer in the involved portion of the esophagus, as demonstrated by increased ^{3}H-thymidine incorporation in the proliferative basal layer. In the stomach, stress-induced gastric erosions are associated with a decrease in gastric epithelial mitosis and thymidine labeling, and pernicious anemia is linked with a decrease in thymidine labeling, migration, and proliferation of gastric epithelial cells, and an increase in endocrine cell proliferation and gastric carcinoids secondary to hypergastrinemia. [328] Ethanol decreases the depth of gastric pits within 4 hours in the dog stomach, but mitotic activity increases at 20 to 24 hours with a prompt renewal of normal gastric mucosa. In the small intestine, ethanol decreases crypt and villus cell number and is associated with shorter villi in the jejunum. Ileal crypts reveal increased mitotic and thymidine kinase activity and an increased cell number in response to injury induced by alcohol. [329] In the

mucosa of patients with duodenal ulcer disease, a fall in thymidine incorporation and mitosis occurs, and this decrease in proliferation has been implicated in the pathogenesis of peptic ulcerations. Infection with *Helicobacter pylori,* a bacterium that colonizes gastric epithelium with great specificity, can result in either gastric atrophy or hypertrophy. Furthermore, the *Helicobacter* species can induce apoptosis of gastric epithelial cells by the stress-activated protein kinase SAPK/JNK pathway. [330] In the colon, repeated radiation injury leads to cell death. In mice with surviving cells, crypts are larger and have higher labeling indices than those in control mice. [331] Irradiation also alters epithelial cell morphology, and in humans results in the appearance of cuboidal epithelial cells and a decreased mitotic rate. After irradiation, early cell proliferative activity is seen at the bottom of the crypt, suggesting that this is where the putative stem cell resides. [332] Mice deficient in p53 demonstrate an absence of stress-induced apoptosis that would normally be observed 3 to 4 hours after ?-radiation. [333] Thus, acute apoptosis after radiation is p53-dependent. BCL2 also plays an important role in the apoptotic response to radiation in the colon, but not in the small intestine. Disorders of the GI tract that are precursors to malignancy are associated with various degrees of altered cell proliferation. In Barrett epithelium, for example, the number of proliferating cells is increased and some proliferative cells are present on the mucosal surface. Intestinal metaplasia is found and an increase in ^{3}H-thymidine incorporation occurs at the base of the villus-like columnar epithelium. [334] A similar pattern is observed in atrophic gastric mucosa with a selective increase in mitotic activity found in those glands demonstrating intestinal metaplasia. [335] Aberrant crypt foci, which on staining appear darker, larger, and thicker than normal crypts, cluster in aggregates and increase in size over time. [336] These foci grow by crypt fission and branching; they contain hyperproliferative epithelial cells and have a higher labeling index. [337] Although only a minor fraction of aberrant crypt foci undergo malignant transformation, these foci are recognized as an intermediate precursor of colorectal carcinogenesis. [338] In colonic adenomas, progressive expansion of the proliferative zone to the lumenal surface has been demonstrated. Abnormal retrograde migration from the mucosal surface also occurs, a pattern that is accentuated in colonic carcinoma. In the mucosa of patients with Gardner syndrome or familial polyposis, progressive expansion of the proliferative compartment occurs to the extent that mitosis may be seen anywhere in the mucosa. [339] Cell proliferation of flat colorectal mucosa is increased in patients with sporadic colonic tumors. Expansion of the proliferative zone toward the lumen has also been observed in ulcerative colitis. Higher rates of proliferation of the colonic mucosa appear to persist during the remittent stages of ulcerative colitis. Alterations in the cell cycle and resistance to apoptosis may also contribute to the development of colorectal carcinogenesis. Whereas low-level apoptosis occurs in the stem cell region of small intestinal crypts, apoptosis does not occur in the stem cell zone of colonic crypts. [340] In colonic adenomas and carcinomas, apoptosis is reduced. [341] In the goblet cells of “normal” colorectal mucosa in patients with colon cancer, the levels of apoptosis are reduced. [342] The APC (adenomatous polyposis coli) gene, mutated in familial adenomatous polyposis and some sporadic colorectal cancers, appears to be an important mediator of apoptosis resistance in colorectal carcinoma. [343]

Regulation of Proliferation and Differentiation

The functional importance of the constant cycle of proliferation, differentiation, and senescence in the GI tract is not entirely clear but may reflect a necessary means of protection from the constant exposure of mucosal cells to potentially injurious factors present in the lumen. Rapid replacement of the mucosal cells could be especially important in the colon, where the lumenal contents often include potent mutagens and carcinogenic substances. The high rate of cell turnover requires exquisite regulatory mechanisms to maintain mucosal homeostasis. Apoptosis, cell proliferation, and cellular commitment to differentiation must be balanced precisely. Myriad factors modulate this balance, including nutrients and other factors in the lumen and the proteins and hormones produced within the mucosa and at more distant sites.

Unlike the needs of other tissues in the body, the nutritional requirements of the GI tract mucosa are usually obtained directly from dietary components in the lumen. Lumenal substances, including nutrients and other factors, regulate growth and differentiation in three ways: by inducing direct effects on the absorptive cells before entering the systemic circulation, by stimulating systemic factors (e.g., trophic hormones), and by modulating motor activity. It has been suggested that the villus height–crypt depth gradient in the small intestine reflects the important contribution of lumenal nutrient concentration to mucosal growth. The greater villus height and crypt depth in the proximal small intestine may result in part from exposure to higher concentrations of nutrients, local growth factors, and pancreaticobiliary secretory products than in the more distal intestine. Transposing the ileal and jejunal loops reverses the gradient; the transposed ileal segment exhibits increased villus size, and the distally placed jejunal segment contains smaller villi. [344]

The role of lumenal factors in the maintenance of mucosal growth has been examined through the use of Thiry-Vella loops, in which intestinal segments with intact neurovascular connections are disconnected from the remainder of the bowel and the two ends are brought to the abdominal wall. [345] These loops may be used to study local effects of a variety of factors and to assay directly the importance of lumenal substances on mucosal growth and proliferation. After Thiry-Vella loops are formed from jejunal or ileal segments, hypoplasia of the intestinal mucosa is observed. Administration of liquid elemental diets into the bypassed segments in the dog induces mucosal hyperplasia. [346] However, comparable nutrient effects were not observed in similar studies performed in rabbits. [347] Furthermore, systemic interleukin-11 prevents the mucosal atrophy observed in Thiry-Vella loops. [348]

Small intestine resection leads to hypertrophy and dilation of the remaining small bowel, with epithelial cell hyperplasia, villus growth, increased cell migration rates, and increased absorptive capacity. [349] Local nutrition may be important in the development of hyperplasia after resection; dogs fed intravenously after jejunal resection do not show the same adaptive proliferation response observed in dogs fed orally. Intravenous feeding was also associated with decreased pancreatic secretion and reduced levels of several gastrointestinal hormones. The latter observations suggest that lumenal nutrients may exert their trophic actions directly or indirectly through the stimulation of hormones and other peptides, and through neural mediators, which modulate mucosal growth (i.e., neurocrine regulation). This concept is supported by the observation of gastric hypersecretion after small intestine resection in the dog, rat, and human, indicating that trophic factors such as gastrin could play a role in the adaptive response to small intestine resection by way of local (i.e., paracrine regulation) or systemic (i.e., hormonal regulation) action.

Other observations suggest that the importance of lumenal nutrients and other lumenal factors may be limited. Hyperplasia has been observed in ileal mucosa after resection of the colon. [350] Similarly, distal small bowel resection has been associated with hyperplasia of the more proximal small bowel, and resection of distal or proximal small bowel can result in hyperplasia of the gastric mucosa. [351] The mucosal hypoplasia observed in Thiry-Vella loops may be reversed by partial resection of any of the small intestine remaining in continuity. Additional evidence supporting the concept that systemic factors may be more important than the direct effect of the lumenal contents is derived from the study of paired rats in which cutaneous or vascular connections were established. In these studies, mucosal hyperplasia was observed in the small bowel of control (nonoperated) rats when connected to rats undergoing small bowel resection.

Glucose stimulates cell production and mucosal hyperplasia of the proximal small intestine when infused into surgically prepared sacs. [352] However, in the same system, galactose, methylglucoside, and NaCl, which are actively transported but not metabolized by the small bowel mucosa, have the same trophic effect as glucose. Mannose, which is not actively transported or metabolized by the small intestine, does not increase cell production or cause mucosal hyperplasia. These findings have led to the notion that the active transport workload itself may be the stimulus for growth in response to some lumenal factors; this is the functional workload hypothesis.

The importance of local nutrition in the regulation of intestinal growth is further discounted by the results of a series of experiments in which amino acid incorporation was studied. [353] Orally administered labeled amino acids were found to be incorporated into nonproliferating surface villus absorptive cells of the small bowel mucosa. However, amino acid incorporation in the proliferative zone of the crypt is slight unless the amino acids are provided by an intravenous route. These observations demonstrate a differential economy between the functionally active but mitotically inert differentiated villus cells and the actively proliferating crypt compartment. These results are consistent with the observation that lumenal nutrients can lead to villus hypertrophy but not true mucosal hyperplasia.

Fiber within the diet may have an important effect on mucosal proliferation. When cellulose is added to a low-fiber synthetic diet, an increase in DNA synthesis and content in gastric and colonic mucosa ensues, regardless of the protein or carbohydrate content of the diet. [354] Guar and pectin, two fibers that are poor bulking agents, are stimulants of growth in the colon, indicating that the effect of fiber on growth is not simply through bulk. [355] Kaolin, an excellent bulking agent, does not appear to have any effect on mucosal growth. Fibers that cause fermentation appear to stimulate colonic mucosal growth more than those that are inert, an effect possibly mediated through a decrease in lumenal pH in the colon. The proliferative effect of refeeding fermentable dietary fiber to starved rats is not seen in germ-free rats, indicating that the products of intestinal fermentation, and not fiber itself, stimulate intestinal growth. [356] Specifically, dietary fiber acts as the main substrate for the fermentation of short-chain fatty acids—butyrate, propionate, and acetate. [357] Of these, butyrate serves as the principal energy source for colonic epithelial cells. Further, butyrate stimulates cell proliferation of normal colonic crypts; reduction of lumenal butyrate levels leads to mucosal atrophy, which is reversible by the administration of butyrate. [358] In contrast, butyrate inhibits proliferation of neoplastic colonic epithelial cells by inhibiting DNA synthesis and arresting colonocytes in the G_1 cell cycle phase and induces differentiation and apoptosis in neoplastic, but not in normal, colonocytes. [359]

The polyamines spermidine and spermine, and their precursor putrescine, are present in the diet and synthesized in the gut lumen. After ligation of the small intestine in the rat, the concentrations of lumenal polyamines increase markedly in the proximal bowel, where a large increase in mucosal growth also occurs. [360] Polyamines

may contribute to the longitudinal gradient of growth observed in the small intestine. Moreover, infusion of dietary polyamines prevents the decrease in gastric and small intestine weight and in DNA, RNA, and protein content normally observed after antrectomy. [361] The trophic effect of polyamines on the gastric mucosa is independent of their ability to stimulate gastrin release, because comparable trophic effects are found after antrectomy.

Ornithine decarboxylase is the enzyme controlling the rate-limiting step in the formation of the polyamines spermine and spermidine. The activity of this enzyme is increased by many gastrointestinal proteins, by food, after partial resection, and during lactation. Difluoromethylornithine, an inhibitor of ornithine decarboxylase activity, decreases the proliferative response to lactation, intestinal obstruction, and feeding. [362] In response to feeding or refeeding, an increase in ornithine decarboxylase has been observed, predominantly in nondividing villus cells near the lumenal surface. In contrast, EGF leads to increased ornithine decarboxylase activity throughout the entire length of the crypt-villus unit. [363] Certain growth factors may increase ornithine decarboxylase activity in stem cells and influence intestinal growth through this mechanism. Polyamine synthesis and subsequent cellular proliferation after refeeding of cultured gastrointestinal crypt cells appears to be regulated by a calcium-activated, calmodulin-dependent process. [364]

Several gastrointestinal proteins with potential growth-modulating properties are present in the intestinal lumen and may exert growth regulation effects through interaction with the lumenal mucosal surface. EGF and EGF-related peptides, including TGF-a, interact with the intestinal lumenal surface. EGF is synthesized in the salivary glands and Brunner glands of the duodenum, a distribution consistent with the notion that this protein could contribute to the proximal-to-distal horizontal gradient of villus height–crypt depth found in the small intestine. [365] EGF is not present in serum in physiologically important amounts.

Gastrin is found in the intestinal lumen in large amounts. It is synthesized in the proximal GI tract, with the highest rates of production in the gastric antrum and smaller quantities in the duodenum and pancreas. When gastrin was infused into the ileum through a catheter, mucosal hyperplasia was observed downstream from the catheter in the absence of a demonstrable elevation of serum gastrin levels. However, when the same amount of gastrin was infused into the stomach, the peptide was degraded, and no trophic effect was observed in the gastric or small intestine mucosa. [366] These findings suggest that lumenal gastrin may not be present in sufficient quantities in the lumen under physiological conditions to play an important role in regulating mucosal growth and proliferation. However, gastrin exerts systemic trophic effects on the gastrointestinal mucosa (see section “ Gastrointestinal Peptides ”).

Prostaglandins appear to promote small intestine and gastric proliferation and growth, but some studies indicate that they may inhibit growth in the colon. Nonetheless, rats treated for 48 hours with 16,16-dimethylprostaglandin E_2 exhibit an increase in gastric and small intestine weight and an increase in ^{3}H-thymidine incorporation into the mucosa of the duodenum and colon. [367] Prostaglandins delivered by a gastric feeding tube produce an increase in antral weight and in small intestine weight and length. A similar study shows increased mucosal thickness throughout the digestive tract, most marked in the antrum, after administration of exogenous prostaglandins. [368] Prostaglandins are gastroprotective against alcohol, acid, hypertonic saline, and alkali at doses below those required to inhibit acid secretions. Furthermore, they prevent ethanol-induced decreases in gastric mucosal DNA, RNA, and protein content.

Pancreatic and Biliary Secretions Several proteins synthesized by the endocrine pancreas that are capable of trophic effects on the intestinal mucosa are also found in exocrine pancreatic secretions. These include gastrin, insulin, and glucagon. The concentration of these proteins from pancreatic secretions decreases along the length of the small intestine in parallel with a proximal-to-distal gradient of growth in the small intestine. Diversion of pancreatic and biliary secretions into the ileum causes ileal mucosal hyperplasia in the rat. [369] However, displacement of these secretions into the distal ileum does not result in hypoplasia or decreased growth of crypts or villi in those areas of the proximal intestine removed from exposure. [370]

Systemic Factors Several nongastrointestinal hormones are thought to play a role in the maintenance of intestinal growth. Hypophysectomy results in decreased weight, mitotic activity, villus height, and diameter of the small intestine in experimental animals. [371] Hypophysectomy also decreases the incorporation of thymidine and orotic acid into DNA and RNA in the intestinal mucosa and leads to reduced rates of protein synthesis. Hypophysectomy results in atrophy of parietal and chief cells in the gastric mucosa. However, hypophysectomy also markedly decreases appetite and food intake and may exert some of its effects through its impact on some of the lumenal factors discussed previously. Moreover, the various pituitary hormones removed by hypophysectomy may affect intestinal growth indirectly by undefined influences on various gastrointestinal proteins. Serum gastrin levels decrease by 60%, in parallel with decreases in antral gastrin content after hypophysectomy. Small intestine secretin content is also iminished in this setting. Adrenalectomy results in gastric and intestinal atrophy to a lesser degree than hypophysectomy. ACTH and glucocorticoids inhibit cell proliferation in the stomach and, to a lesser extent, in the duodenum. In other regions of the small intestine, glucocorticoids increase digestive and absorptive functions but decrease cell proliferation. [372] Glucocorticoids increase antral gastrin levels in the rat and prevent the decrease in antral gastrin content that occurs in hypophysectomized rats. It appears that corticosteroids promote the manifestations of functional differentiation while inhibiting proliferative activity. Growth hormone stimulates the growth of most tissues during development. Growth hormone stimulates mitosis in the duodenal crypts of hypophysectomized or thyroidectomized rats, and it leads to increased pancreatic DNA and RNA content and increased pancreatic weight in hypophysectomized rats. [373] However, growth hormone also leads to increased food intake, small intestine secretin content, and serum and antral gastrin levels in hypophysectomized rats. Antral G-cell proliferation and an increase in antral gastrin content have occurred in patients with acromegaly. Overexpression of bovine growth hormone in transgenic mice leads to an increase in villus height and small intestine mass. [374] This effect is not secondary to increased food intake, because diet-restricted mice and mice fed ad libitum demonstrate the same increases. In humans, growth hormone improves clinical parameters in Crohn’s disease, [375] perhaps through a trophic effect produced by intermediate modulation of several other growth factors. IGF-I, also known as *somatomedin C,* is expressed throughout the intestine and may mediate many of the actions of growth hormone on the GI tract. Thyroxine causes an increase in pancreatic weight and RNA content, but it does not increase DNA content. [373] Thyroxine does not increase the mitotic activity of duodenal crypts in hypophysectomized rats unless growth hormone is also administered.

Gastrointestinal Peptides Gastrin, the most potent stimulant of gastric secretion, is thought to have important trophic effects on the GI tract. Exogenous gastrin stimulates the growth of mucosa throughout the entire GI tract except in the esophagus and, possibly, the gastric antrum, where most of the peptide is produced. Gastrin has no trophic effects outside the intestinal tract, and within the intestinal tract, its effects appear to be limited to the mucosa. [376] In the stomach, gastrin increases the parietal cell population by shortening the time it takes fundic stem cells to differentiate into parietal cells. Pentagastrin also increases thymidine kinase activity in the rat gastric mucosa. The number of enteroendocrine cells, but not chief cells, is increased by gastrin in the stomach. Gastrin stimulates the production of its own receptors, as evidenced by a direct correlation between serum gastrin levels and gastrin binding capacity. [377] Proglumide, a competitive inhibitor of gastrin binding to its receptor, inhibits the trophic actions of gastrin on the stomach, duodenum, and colon. CCK also competes for binding to the gastrin receptor, and secretin blocks the gastrin receptor in a noncompetitive manner. Intravenous infusions of gastrin that maintain serum gastrin concentrations similar to those expected after a meat meal lead to increased DNA synthesis. Gastrin 17 and gastrin 34, the two most abundant forms of endogenous gastrin, are equal to pentagastrin in increasing DNA synthesis and content in duodenal and gastric mucosa. [378] Serum gastrin levels also parallel gastrointestinal growth responses after antrectomy or dietary manipulations. Marked increases in serum and antral gastrin levels caused by H^+,K^+-ATPase blockers are associated with a 25% to 30% increase in parietal, chief, and mucous cell numbers and a fivefold increase in ECL cells and thymidine incorporation. [379] Gastrin has also been postulated to promote growth of colorectal carcinoma through an autocrine mechanism. Gastrin mRNA has been detected in colon cancer cell lines, and serum gastrin levels are elevated in a subset of patients with colonic adenomas and carcinomas. [380] Gastrin receptors are present in colon cancer cell lines and tumors. [381] Studies using highly specific monoclonal antibodies reported that gastrin precursors that had not undergone full posttranslational modification were the predominant form of gastrin peptide in colorectal carcinoma. [382] Colonic cells may possess a distinct class of gastrin receptors that bind nonamidated gastrin instead of the classical amidated hormone. Amidated gastrin appears to stimulate cell proliferation by inducing c-Fos and c-Jun, whereas glycine-extended intermediates of gastrin stimulate transcription through early gene activation mediated by a distinct receptor. [383] Transgenic mice that selectively express elevated levels of incompletely processed gastrin demonstrate evidence of increased colonic mucosal proliferation in vivo. [384] The IGFs (types I and II) and their receptors are expressed throughout the GI tract. Administration of IGF-I to normal cats, growth hormone–deficient mice, rats after partial small bowel resection, or dexamethasone-treated rats increases gastrointestinal mass. [385] The trophic effect on IGF-I on the intestine is polyamine-dependent and can be blocked by difluoromethylornithine. IGF-II appears to play the dominant role in fetal growth and development, although adult intestinal epithelial cells express both IGF-II and its receptor. [386] VIP is structurally related to secretin. VIP similarly inhibits the effect of pentagastrin on the markers of gastrin and colonic mucosal proliferation but does not appear to have any significant intrinsic trophic effects. VIP also has no effect on the growth of pancreatic acinar cells in vitro. [387] GRP is homologous to the amphibian peptide bombesin. In the GI tract, GRP stimulates antral G-cell proliferation and increases gastric and colonic mucosal DNA content. [388] Its effects may be partly or completely mediated through the stimulation of gastrin release from the G cell. GRP reduces small intestine atrophy that results from ingestion of an elemental diet and improves the survival of patients with chemotherapy-induced enterocolitis. [389] Neurotensin, a tridecapeptide widely distributed throughout the GI tract, is a member of the pancreatic polypeptide family secreted by the N cell, the most abundant cell in the ileum. Neurotensin increases small intestine mucosal weight, DNA, and protein content in the rat, but it did not appear to have any effect on mucosal growth in the colon. [390] Neurotensin stimulates mucosal growth throughout the small intestine and increases levels of sucrase, maltase, and alkaline phosphatase in the small intestine. Infusion of peptide YY, a peptide synthesized in the pancreas and by enteroendocrine cells, inhibits CCK-stimulated pancreatic growth and may serve as a physiological inhibitor of pancreatic growth. [391] Peptide YY infusion also stimulates enterocyte proliferation in vitro, and when the effect of EGF on plasma levels of gastrin, enteroglucagon, and peptide YY was correlated with gut proliferation, only peptide YY levels were significantly elevated by EGF, indicating a possible role for peptide YY as a mediator of the trophic effects of EGF. Plasma levels of peptide YY are elevated after massive small bowel resection, and administration of peptide

YY is trophic for mouse small intestine and colon. [392] Neurotensin appears to bind the same receptor site as peptide YY in small intestine epithelial cells, but these receptors have a greater affinity for peptide YY than neurotensin. Neurotensin and peptide YY decrease VIP- or prostaglandin-stimulated cAMP levels in rat jejunal epithelial cells. Serum levels of EGF are very low, but intravenous EGF stimulates epithelial cell proliferation in humans, chickens, and mice. [393] Exogenous EGF stimulates DNA, RNA, and protein content of the gastric mucosa but may not stimulate growth of duodenal and colonic mucosa. After small bowel resection, EGF increases residual small intestine thickness and weight. [394] Within 1 week after surgical excision of submandibular glands, rats demonstrate lower jejunal and ileal but not colonic RNA and DNA content than sham-operated controls. Specific receptors exist for EGF in intestinal epithelial cells at the microvillar and basolateral surfaces. The detection of receptors in microvillus membrane preparations through binding of the labeled ligand has lent support to the notion that lumenal EGF may regulate intestinal epithelial growth, although the significance of this discovery is uncertain. The mitogenic activity of EGF in most responsive cells may be mediated through the regulation of extracellular matrix and ornithine decarboxylase. Some members of the EGF family of peptides, including amphiregulin and cripto, which are expressed in the colon, and heregulin, which is expressed in the stomach, pancreas, and small intestine, may also have mitogenic activity in the intestinal tract. [395] TGF-a, a structural homolog of EGF that binds to the same receptor, stimulates thymidine incorporation in many cell types. TGF-a, mRNA, and bioactive protein are demonstrable in human and rodent stomach and in small intestinal and colonic epithelium, and they may be the dominant physiological factor interacting with the EGF receptor in the distal GI tract. [396] Transgenic mice overexpressing TGF-a in the gastric mucosa demonstrate hyperplasia and hypertrophy similar to that seen in Ménétrier disease. [397] TGF-a also promotes restitution of gastric mucosa and of intestinal epithelial cell lines. TGF-a is expressed in a variety of esophageal and gastric carcinomas and colon cancer cell lines, leading to the supposition that TGF-a may contribute to autocrine neoplastic growth. The TGF-ß family of peptides plays key regulatory functions in a wide spectrum of biologic processes, including stimulation or inhibition of cell proliferation, cellular differentiation, embryological development, and formation of extracellular matrix. TGF-ß inhibits proliferation of all epithelial cell populations, including the intestinal epithelium, and overrides the effects of direct mitogens (e.g., EGF, TGF-a). [398] In the small intestine, the highest concentrations of TGF-ß mRNA are found in the crypts rather than in the villi, although the peptide may be more generally distributed. TGF-ß and its receptors are found in many colon cancer cell lines and appear to inhibit their growth. [399] Targeted disruption of the TGF-ß $_1$ gene in mice results in diffuse inflammation and ulceration of the stomach and intestine. [400] The ability of TGF-ß to promote collagen synthesis may be important in the formation of strictures in inflammatory bowel disease. In patients with inflammatory bowel disease, an increase in TGF-ß $_1$ mRNA levels secondary to increased TGF-ß expression in the lamina propria may contribute to stricture formation. [401] The fibroblast growth factor (FGF) family of peptides has not been examined extensively in the context of the GI tract, but it may play an important role in neovascularization, proliferation, and modulation of the functional phenotype of fibroblasts after gastrointestinal injury. Immunoreactive basic FGF has been detected in the small intestine and colon within the extracellular matrix adjacent to epithelial cells and blood vessels. [402] FGF receptors are also present in the GI tract. FGF that has been chemically modified for lumenal stability enhances ulcer healing, and immunoneutralized anti-FGF aggravates ulceration after injury in rodents. [403] Basic FGF demonstrates mitogenic activity in several human colon cancer cell lines, overriding the growth inhibitory effects of TGF-ß. [404] Keratinocyte growth factor (KGF), a member of the FGF family, increases intestinal crypt cell mitosis and protects against radiation-induced intestinal injury. [405] Hepatocyte growth factor (HGF) and its receptor, the c-Met protooncogene product, are expressed throughout the GI tract; HGF expression is greatest in the intestinal crypts. [406] HGF has been shown to be mitogenic for the liver, and it may also play a role in intestinal epithelial proliferation. [407] Somatostatin, which is found throughout the intestinal tract and pancreas, inhibits the release of many gut peptides, including gastrin. It also inhibits the action of gastrin and other gut peptides at their target cell. In addition to inhibiting gastrin release, somatostatin inhibits the stimulation of gastric mucosal growth by endogenous and exogenous gastrin. [408] It remains unclear whether somatostatin exerts its inhibitory effects on gastrointestinal growth solely through its inhibition of gastrointestinal peptides or whether it has direct effects on mucosal growth of its own, inducing apoptosis in intestinal crypts and villi. [409] High serum levels of enteroglucagon have been measured in several conditions associated with growth of the small bowel: adaptation after intestinal resection, celiac sprue, and tropical sprue. [410] After massive small bowel resection, ileal glucagon mRNA levels increase and intestinal L cells synthesize increased amounts of enteroglucagon. [411] Serum enteroglucagon levels correlate with the rate of crypt cell production in the distal small bowel but not in the colon. [412] Despite the correlation of enteroglucagon levels with mucosal growth, no evidence exists that enteroglucagon directly stimulates growth. The effects of pancreatic glucagon on intestinal growth are not well understood. Although glucagon has been shown to increase DNA synthesis and content in gastric and colonic mucosa, another study found that exogenous pancreatic glucagon decreases villus height along the rat small intestine. [413] Overexpression of GLP-2, one of the peptides derived from enteroglucagon, demonstrates a marked trophic effect on the intestine in transgenic mice. [414] The GI tract represents a dynamic model for the study of cell and tissue development, growth, and differentiation. Although insights into newly described growth factors and mechanisms of cell-to-cell communications and interactions have increased our understanding of the regulation of intestinal growth and development, the precise signals controlling these processes and the specific pathways of events, such as cell migration, adhesion, renewal, extrusion, and transformation, require further definition.

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CHAPTER 24

John M. Carethers and C. Richard Boland

NEOPLASIA OF THE GASTROINTESTINAL TRACT

The gastrointestinal tract, including the hollow organs of the gut, pancreas, liver, and biliary tree, is the site of more cancers and the source of more cancer mortality than any other organ system in the body. However, no simple etiologic explanation can unify all gut tumors.

The most notable feature of international cancer epidemiology is the wide variability of tumor incidence from country to country by organ site. For example, an esophageal cancer belt extends from northeastern China through central Asia into northern Iran. [1] In portions of these regions, the incidence of squamous cell carcinoma of the esophagus is more than 100-fold higher than that in adjacent low-incidence regions. In the United States, three- to fourfold differences in the incidence of esophageal cancer occur between the sexes and among races. [2] In Japan, the incidence of gastric carcinoma is about ten times higher than it is in the United States. Colorectal cancer occurs less commonly in Japan, but it is the most common gastrointestinal malignancy in North America, western Europe, and much of the industrialized world. Over the past decade, the incidence of colorectal cancer in Japan has risen to almost the same level as in the West. [3] These marked differences in cancer risk are not based on racial or genetic factors. When people migrate from a high-incidence region to a low-incidence region, the organ-specific rates of some cancers change to match that of the new region, usually within two generations. Collectively, the epidemiologic observations strongly indicate the importance of environmental factors in gastrointestinal carcinogenesis; however, individual genetic differences may influence the effects of these factors.

Worldwide, people are exposed to different types of foods, the gastrointestinal flora differs as a result of dietary and host factors, and certain mucosal infections occur in different frequencies. Gastrointestinal carcinogenesis, therefore, is a complex problem to understand and to modify. For example, tobacco smoke causes cancers of the oral and respiratory tracts, and by eliminating smoke from the environment, the risks of tumor development can be controlled. However, everyone consumes some kind of diet, and the elimination of specific items results in the introduction of replacement nutrients. Moreover, the diet consists of a complex combination of macronutrients (i.e., fat, carbohydrate, protein, fiber) and micronutrients (i.e., vitamins, minerals, other agents present in tiny concentrations) that affect cancer risk. Directly acting carcinogens or indirectly acting procarcinogens also are part of each person's diet. Mucosal infections with human papillomavirus and *Helicobacter pylori* probably contribute to the genesis of cancers of specific gastrointestinal organs.

Ultimately, changes at the genetic level can alter the growth characteristics of healthy gastrointestinal cells. Although many deleterious genetic events must occur for a normal cell to become malignant, inactivation of a gatekeeper gene or pathway specific to each tissue may be required to initiate the neoplastic process. [4] This concept is evident in familial syndromes such as familial adenomatous polyposis (FAP), in which thousands of benign colonic adenomas develop at a young age; it can also be applied to the development of sporadic colorectal cancer. After the initial gatekeeper alteration, successive genetic events propel the cell toward the malignant phenotype. The loss of the gatekeeper's function and the successive genetic events lead to an uncoupling of cellular growth control mechanisms that typically regulate a normal gastrointestinal cell. These include loss of cell cycle control checkpoints, abrogation of programmed cell death, and dysregulation of intracellular signaling pathways. Some genetic perturbations have been identified as markers for the presence of neoplasia as well as prognostic factors for malignancy, but their clinical use has not been fully evaluated.

Because our understanding of carcinogenesis in the gut is far from complete, it is necessary to understand the principles of regulation of normal, nonneoplastic gastrointestinal cell growth before considering the abnormal events. The mechanisms of tumor development appear to be distinct among the gastrointestinal organs, but general pathogenetic principles apply at the cellular and molecular level for most tumors.

NORMAL CELLULAR CONTROL MECHANISMS

Cell Cycle Checkpoints

The cell replication cycle consists of five phases. [5] Resting diploid cells not involved in active replication may be in the G_0 or G_1 phase. G_1 represents the postmitotic period before the initiation of new DNA synthesis as the cell prepares to divide. The cell may spend a variable period of time in G_1. Cells not involved in replication may be described arbitrarily as resting in the G_0 phase. After receiving the signal to divide, cells enter a phase of active DNA synthesis, the S phase. After duplication of the entire genome, the tetraploid cell pauses briefly in the G_2 phase before entering mitosis (M phase). After cell division into two identical daughter cells, the cell reenters the G_1 phase. As the genome becomes more complex with tumor progression, the S and M phases may be prolonged, altering the apparent fraction of cells involved in replication. This complicates a direct comparison of the proliferative activity of tumors with that of normal tissue.

Each transition from one phase of the cell cycle to the next is regulated by a family of cyclin-dependent kinases (CDKs) and their activating partners, the cyclins (Fig. 24-1). The CDK-cyclin complexes are positively and negatively regulated by phosphorylation. The formation and activation of a CDK-cyclin complex requires proper completion of the previous cell cycle phase before the subsequent phase is entered. [6], [7] If DNA is damaged by ultraviolet light in the G_1 phase of the cell cycle, passage into the S phase will not occur until the damage is repaired. Likewise, if DNA is not replicated with complete fidelity during the S phase, passage into G_2 and mitosis is prohibited until an exact copy of the DNA is duplicated. The CDK-cyclin complexes receive input from other cellular proteins that relay information regarding the integrity of each checkpoint. The product of the retinoblastoma tumor suppressor gene, *RB,* and p53 relay information to G_1 phase CDK-cyclins to regulate the initiation of DNA synthesis (S phase). [6] These cell cycle checkpoints are lost in oncogenesis. The CDK-cyclin complexes themselves may be altered in some tumors, and in the gastrointestinal tract, changes occur in the regulators that relay information to CDK-cyclin complexes. These genetic perturbations allow the neoplastic cell to cycle with disregard for mutations and chromosomal rearrangements, essentially establishing progeny that differ from the parent. This generation of genetic diversity is a form of genomic instability.

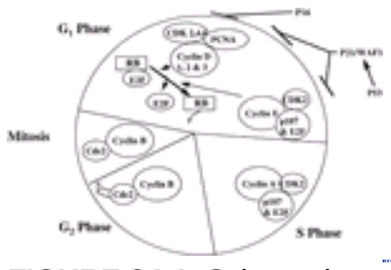

FIGURE 24-1. Schematic representation of the cell growth cycle. Transition through each phase of the cell cycle is regulated by cyclins and cyclin-dependent kinases (*CDK*s) to ensure completion of one phase before initiation of the subsequent phase. Each cyclin-CDK complex is positively and negatively regulated by phosphorylation. Inhibitors of CDKs include p21 and p16, both of which prevent transition to the S phase of the cell cycle. The transcriptional regulator E2F transactivates genes important for S phase entry. E2F is bound by the retinoblastoma protein (*RB*) until RB is phosphorylated, thus releasing E2F. Phosphorylation of RB is initiated and accelerated by cyclin D–dependent kinases and cyclin E–CDK2 complexes, respectively. *P,* phosphorylated; *PCNA,* proliferating cell nuclear antigen.

Programmed Cell Death

Deregulated proliferation is the sine qua non of the neoplastic process. An increase in the proliferation rate of cells can occur either by increased mitosis or by a decreased cell death rate. Programmed cell death is a physiological process that plays an essential role in normal tissue turnover and embryonic development. [8] This process has been shown to require energy, macromolecular synthesis, and de novo gene transcription. [8], [9] The morphologic changes of programmed cell death, first described in 1972 by Kerr and colleagues [10] and termed *apoptosis,* affect single scattered cells with characteristic changes in the nucleus (i.e., karyorrhexis), preservation of cytoplasmic organelles, cellular shrinkage, and the formation of apoptotic bodies. The initiation of programmed cell death may involve a sustained increase in intracellular calcium, which activates several calcium-dependent enzymes. [8] These enzymes, in turn, activate stored or newly synthesized transglutaminases (involved in the cross-linking of cytoplasmic proteins), proteases (e.g., caspases) that aid in disruption of the cytoskeleton and cell shrinkage, and endonucleases that cleave DNA into uniformly sized fragments. [8] Once activated, programmed cell death is irreversible. [9]

Activation of a cell death program is best illustrated by the normal turnover of the colonic epithelium. In healthy colonocytes, the surface epithelium is constantly renewed about every 6 days by cellular proliferation and differentiation of crypt cells. The proliferating compartment is in the lower third of the crypt, characterized by mitoses and upward migration from anchored stem cells. As cells move up the crypt, differentiation and maturation occur, and the cells lose their capacity to divide. Eventually, cells die and are shed. Programmed cell death of colonocytes thus maintains a uniform number of cells in the mucosa. Programmed cell death of colonocytes results, in part, from the activity of the adenomatous polyposis coli (APC) gene. [11] Other cellular proteins, such as those of the BCL2 gene family, may play a significant role in programmed cell death in the gastrointestinal tract.

Activation of programmed cell death is a mechanism involved in the treatment of gastrointestinal tumors. Both radiation and chemotherapy can induce apoptosis in tumors of the gastrointestinal tract, in addition to direct toxic tissue injury (i.e., necrosis). [8], [9] Apoptosis has been proposed to be the mechanism by which the nonsteroidal antiinflammatory agent sulindac and the cyclooxygenase-2 (COX-2) inhibitor celecoxib lead to the involution of colonic adenomas in patients with FAP. [12], [13] and [14] Interference with programmed cell death may be as important as deregulated cell proliferation in some gastrointestinal tumors, and its activation is the target of many therapeutic interventions.

Intercellular and Intracellular Growth Control Pathways

Cells that make up the tissues of the gastrointestinal tract are in contact with neighboring cells to form a tight epithelial barrier. In addition to functioning as a barrier, the epithelial junctions that interconnect neighboring cells distribute information about the cell's local environment, such as the presence of a contiguous cell (e.g., cell-cell adhesion and recognition at the zona adherens), and they may allow the direct passage of ions and small molecules through specialized pores called *gap junctions*. [15] Passage of ions and small molecules through gap junctions allows cells to be coupled and permits groups of cells to behave as a syncytium in regard to ion transfer. Cell-cell adhesion is primarily mediated through the intracellular adhesion molecule E (epithelial)-cadherin, which forms homeotypic interactions (i.e., with other E-cadherin molecules) and anchors the zona adherens to the actin cytoskeleton inside the cell. [16] Dysregulation of cell-cell adhesion disrupts normal intercellular communication and allows transformed cells to separate from each other and to spread locally, invading regional tissue. Proteins from the catenin family (a, ß, ?) bind to E-cadherin and to the actin cytoskeleton, but any signal relayed by the catenins for cellular adhesion is not currently known. Importantly, ß-catenin, a protooncogene that can transactivate genes associated with cellular proliferation, has its cytoplasmic levels regulated by the APC protein when its intercellular concentration rises (Fig. 24-2). [17], [18] and [19] APC targets ß-catenin for degradation; and if ß-catenin escapes this homeostatic regulation, it can move to the nucleus, where it associates with T-cell factor-4 (TCF-4) to transactivate genes that promote cell growth and prevent programmed cell death.

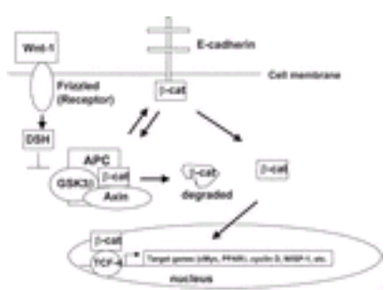

FIGURE 24-2. Adenomatous polyposis coli gene (*APC*) regulation of ß-catenin and its function as a tumor suppressor gene. Normal APC, in association with axin and glycogen synthase kinase-3ß (*GSK3ß*), binds, phosphorylates, and targets cytoplasmic ß-catenin (*ß-cat*) for degradation. Degradation of ß-catenin prevents its translocation into the nucleus, where it can initiate the transcription of genes by converting the transcriptional repressor T-cell factor-4 (*TCF-4*) to a transcriptional activator to promote cellular proliferation and prevent cell death. Cytoplasmic ß-catenin can bind to E-cadherin, usually in association with a-catenin. During embryogenesis, blockage of APC-mediated degradation can occur through the Wnt-1 signaling pathway (through the frizzled receptor and disheveled [*DSH*] signaling protein). Presumably, inactivation of APC would have the same consequences as Wnt-1 signaling.

Another mode of intercellular communication exists between epithelial cells and other cells that line the gastrointestinal tract. Growth factors are proteins that bind as a ligand to a receptor on the same (autocrine) cell or neighboring (paracrine) cell. Growth factors modulate growth characteristics of the cell. Once the growth factor is bound to its specific receptor, an intracellular signal is amplified and transmitted to the nucleus of the cell to induce or inhibit gene activation; this in turn modifies growth characteristics, metabolism, and ion transport. Although many growth factors use tyrosine phosphorylation at the start of their intracellular signaling pathway, other ligand-receptor complexes often use other pathways, such as serine/threonine phosphorylation, calcium and phosphoinositol formation, and the generation of cyclic nucleotides. Two important examples highlight the signal transduction process in normal and neoplastic growth in the gastrointestinal tract: the KRAS and transforming growth factor-ß $_1$ (TGFB1) signaling pathways.

The protooncogene Kirsten-RAS (*KRAS*) encodes an intracellular protein that binds the cyclic nucleotide guanosine tri- or diphosphate (GTP/GDP). On cell surface ligand-receptor activation (e.g., epithelial growth factor binding to its receptor), the KRAS protein is activated and exchanges the bound GDP for GTP (Fig. 24-3). Active KRAS molecules then interact with downstream intracellular effector molecules through an effector domain, conveying a growth response to the nucleus. To turn off this activity, KRAS is deactivated by hydrolysis of GTP to GDP, which returns KRAS to the inactive GDP-bound state. [20] This signal transduction process is perturbed with a mutant KRAS protein. Stabilization of mutant KRAS in its active state permits continuous signal transduction, allowing the cell to undergo unregulated proliferation. This process may be caused by mutations that inhibit the intrinsic GTPase activity of KRAS, allowing KRAS to stay in its active conformation, or by inducing an active conformation that does not require the binding of the guanine nucleotides. [20] Indeed, the most common mutations of *KRAS* in pancreatic and colon cancers involve codons 12, 13, and 61, corresponding to areas in the GTP/GDP binding domains of the KRAS protein. [20] In pancreatic cancers, mutant *KRAS* has

been reported in 95% of tumors, [21], [22] whereas it occurs in about half of colorectal cancers [23], [24] and [25] and is uncommon in gastric cancers. [26] It remains to be determined if other members of the RAS signaling pathway are involved in the pathogenesis of tumors with wild-type *RAS* alleles.

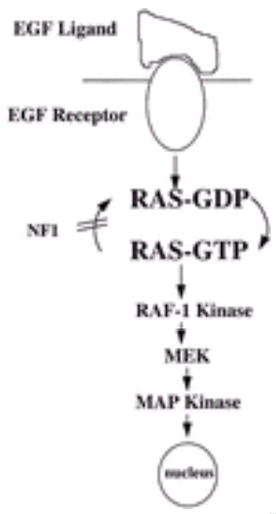

FIGURE 24-3. RAS intracellular signaling. Cell surface ligand binding to a receptor, such as epithelial growth factor (*EGF*) to the EGF receptor, triggers activation of GDP-bound RAS to GTP-bound RAS. RAS-GTP can activate subsequent steps in the intracellular signaling cascade (RAF-1 kinase, mitogen-activating protein [*MAP*] kinase kinase [*MEK*], and MAP kinase) to activate genes that promote cellular proliferation. The tumor suppressor protein neurofibromatosis 1 (*NF1*) is an activating protein for RAS, keeping RAS in the GTP-bound state. Activation of RAS by mutation allows continued signaling of this pathway.

TGFB1 is a ligand that inhibits the growth of gastrointestinal epithelial cells. [27] To exert its growth suppressive effect, TGFB1 in its latent form is activated by the insulin-like growth factor II receptor (IGF2R) present on the cell surface (Fig. 24-4). [28], [29] Activated TGFB1 then binds to TGFB1 type II receptor (TGFBR2), which subsequently phosphorylates (as a serine threonine kinase) the TGFB1 type I receptor (TGFBR1), forming a heteromeric complex of TGFB1, TGFBR2, and TGFBR1. [30], [31] The conjoined receptor complex then phosphorylates (as a serine threonine kinase) intracellular proteins that cascade to ultimately affect the transcription of growth-suppressing genes in the nucleus. In particular, SMAD2 and SMAD3 are phosphorylated by activated TGFBR1, which allows them to bind individually to SMAD4 and subsequently translocate to the nucleus. Inactivation of this pathway occurs in some gastrointestinal tumors. For example, many colonic and gastric tumors that exhibit microsatellite instability, a laboratory marker for the inactivation of the DNA mismatch repair (MMR) system, develop mutations in *TGFBR2*. [32], [33] and [34] Other tumors with microsatellite instability but with normal *TGFBR2* have mutations in *IGF2R*. [29] These mutations prevent expression of the receptor on the cell surface. The absence of TGFBR2 or IGF2R prevents activation and binding of the TGFB1 ligand and abrogates its growth-suppressive action. Some colon tumors without microsatellite instability have mutations in the kinase domain of TGFBR2. [35] Inactivation of the potential downstream signaling proteins SMAD4 and SMAD2 also has been found in gastrointestinal cancers. [36], [37] The key point is that essential growth-regulatory pathways may be deactivated at any one of several points in the cascade.

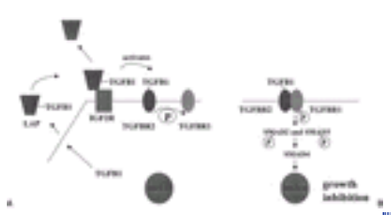

FIGURE 24-4. Transforming growth factor-ß $_1$ signaling. **A:** TGF-ß $_1$ (*TGFB1*) can be produced in an autocrine fashion by epithelial cells, and it is bound by a latency-associated peptide (*LAP*) once outside the cell. To activate TGFB1, LAP is cleaved by the insulin-like growth factor II receptor (*IGF2R*), leaving TGFB1 free to bind to the TGFB1 receptor type II (*TGFBR2*). This action, in turn, phosphorylates (P) the TGFB1 receptor type I (*TGFBR1*), which then forms a heteromeric complex with TGFBR2. **B:** Subsequent intracellular signaling to the nucleus occurs through the SMAD family of proteins, ultimately causing growth suppression. Mutations in *IGF2R, TGFBR2, SMAD2,* and *SMAD4* have been described that uncouple the growth-inhibitory effects of TGFB1 on the colonocyte.

Many of these growth-regulatory pathways are active simultaneously. They are often redundant and sometimes interconnected, but they typically provide a homeostatic balance for normal cellular growth. Alteration of one or more of these pathways may lead toward cellular proliferation and away from controlled growth and regulated cell turnover by programmed cell death, initiating and exacerbating transformation into neoplastic cells. The complexity of cellular growth mechanisms affords the neoplastic cell many opportunities for a pathway to become deregulated. It appears that more than one pathway is affected, which may be necessary for transformation. Specific pathway inactivations are more common in certain types or subtypes of tumors, creating opportunities for therapeutic intervention of gastrointestinal tumors as biotechnology advances.

MOLECULAR CARCINOGENESIS

Mechanisms by Which DNA Is Damaged

The current concept of carcinogenesis in the gastrointestinal tract begins with damage to normal cellular genes. This can occur through a variety of mechanisms. Mutations occur spontaneously because of predictable instability of the purine and pyrimidine bases themselves. The most common spontaneous mutation is depurination, which occurs after disruption of the *N*-glycosyl linkages between the purine base and the ribose chain. In another spontaneous event, deamination, cytosine is converted to uracil, or 5-methylcytosine to thymine. These single base mutations, if not repaired, lead to mispairing during the next round of replication. Both events are common and subject to revision by families of DNA repair enzymes.

Several environmental factors predictably damage DNA, including viral infections, chemical carcinogens, and radiation in the form of ultraviolet radiation, ionizing electromagnetic radiation such as x-rays and ?-rays, and particle radiation such as electrons, a particles, and heavy ions (Table 24-1). Radiation injury damages DNA by producing strand breaks. As an internal organ, the gastrointestinal tract is usually not exposed to sufficient radiation for it to be a major factor in gastrointestinal carcinogenesis. However, very high doses of x-rays administered under unusual circumstances have been implicated as a pathogenetic factor in some intestinal cancers. Viral oncogenesis has been documented in several nonhuman tumor models and in squamous cell carcinoma of the esophagus and anus in humans. [38], [39] JC virus, a DNA virus encoding a T antigen, is present in the gut and has been implicated in colorectal carcinogenesis. [40] There is evidence to suggest a role for viruses in the genesis of adenocarcinomas of the gastrointestinal tract, such as the role of hepatitis B and C viruses in causing hepatomas. Chemical carcinogens are thought to be most important in initiating adenocarcinoma in the gastrointestinal tract because of their access to the gut mucosa. This form of injury tends to produce mutations in single nucleotide bases.

Radiation Injury
Ultraviolet radiation
Especially important for skin
Most important wavelengths, 290–320 nm
Ionizing radiation
Electromagnetic radiation (e.g., x-rays, γ-rays)
Particulate radiation
Electrons, protons, neutrons, α particles, heavy ions
Viral Oncogenesis
Human papillomavirus
Squamous cell carcinoma of the esophagus
Anal carcinoma
Genital cancers
Epstein-Barr virus
Burkitt lymphoma
Nasopharyngeal carcinoma
Hepatitis B and C viruses
Hepatoma
Human T-cell leukemia viruses
Chemical Carcinogens
Important in a wide spectrum of cancers, including digestive, oral, pulmonary, urinary, cutaneous, and hematologic malignancies
Complex relation between host and environment involved
Numerous defensive or protective mechanisms at work

TABLE 24-1 Carcinogenic Agents

Single base pair mutations are characterized as *transitions* when they change one purine to another or change one pyrimidine to another. They are characterized as *transversions* when they produce a change between nucleotide classes. Losses of base pairs in the DNA sequence are called *deletions,* and gains are called *insertions*. Some single base pair changes are silent and do not result in amino acid changes. Mutations that change the amino acid in the protein product are called *missense mutations,* and they are characterized as conservative if the amino acid change occurs within the same class (i.e., from one neutral amino acid to another). Missense mutations can be serious if they result in the appearance or removal of charge, cysteines, phosphorylation or glycosylation sites, or other amino acids critical to the folding and function of the protein. Changes that create a stop codon within an open reading frame are called *nonsense mutations*. Nucleotide insertions and deletions (which do not occur in groups of three) create a frameshift, which commonly produces a series of missense and nonsense changes downstream on the DNA strand.

Chemical Carcinogenesis Chemical carcinogens are ubiquitous in the human diet but may not reach all the digestive organs in their active forms. [41], [42] Typically, proximate carcinogens are highly reactive, short-lived chemical compounds that bind nucleic acids, proteins, and other macromolecules near the site of their generation. Chemical carcinogens usually have a narrow range of host and tissue specificity. The microbial flora of the gastrointestinal tract and mucosal enzymes are important factors in the activation and inactivation of many carcinogens. For example, cycasin, an extract made from cycad nuts, produces intestinal tumors when administered to most rodents. [43] This compound is a glycoside that must be hydrolyzed by intestinal bacteria to produce methylazoxymethanol, which is unstable and spontaneously decomposes, giving rise to a reactive carbonium ion capable of methylating nucleic acids. Cycasin does not give rise to tumors in germ-free rodents because they lack the necessary glycosidases produced by the intestinal flora. The procarcinogen is never activated and is therefore unable to damage their DNA. For the same reason, cycasin is active only when administered orally and does not produce colon cancer after parenteral injection. Studies using derivatives of this compound have led to an understanding of the complexities involved in the metabolism of procarcinogens to the active, proximate mutagen. [44], [45], [46], [47] and [48]

Carcinogen Metabolism Although cancer is best characterized as an acquired disease based on environmental exposures, there are large individual differences in cancer susceptibility. [49] The best-known example of chemical carcinogenesis in humans is the relation between tobacco use and lung cancer. However, not all smokers are equally susceptible to cancer, even when differences in the number of cigarettes smoked are taken into account. The enzyme cytochrome P450 (CYP2EI) is responsible for the activation of many carcinogens, including many of those in tobacco smoke, and it is expressed heterogeneously throughout the population. One study indicated that the inheritance of specific alleles of the gene for this enzyme was associated with significantly different risks for lung cancer. [50] Although it was initially suggested that individuals who are rapid acetylators of potential carcinogenic compounds are at increased risk for colorectal cancer, there is little insight into this mechanism for increased risk for bowel cancer. [51], [52], [53] and [54] Nonetheless, it is reasonable to expect that differences exist in the ability to activate procarcinogens or inactivate proximate carcinogens that are important factors for cancer risk.

Mechanisms by Which Genes Are Damaged

Point Mutation The significance of point mutation in gene damage was first appreciated when it was found that animal tumor viruses carried mutated forms of cellular protooncogenes. Transfection of these mutated genes into NIH3T3 cells transformed the fibroblasts into cells that assumed a malignant morphology in vitro, no longer exhibited contact inhibition, and could form tumors when injected into nude mice. A transforming oncogene was first found in the Rous sarcoma virus, and later the *HRAS* gene was found to be oncogenic in this laboratory setting. [55], [56] *RAS* gene mutations have been demonstrated in about half of colorectal cancers and in most pancreatic cancers. [22], [24], [25] Point mutations may be produced in DNA by the administration of alkylating agents; for example, treatment of rats with the carcinogen 1,2-dimethylhydrazine (DMH) results in the alkylation of DNA throughout the genome. [57] After exposure to such an agent, the host attempts to repair damaged DNA, or in some instances, the cell responds to the damage by initiating programmed cell death. [8], [9] The action of a carcinogen triggers a dynamic response; the DNA is altered first, and the host then attempts to limit the impact of mutational damage, possibly sacrificing some damaged cells to prevent the replication of a damaged genome. Carcinogens may produce characteristic damage based on the chemistry of the interaction between the reactive compound and certain susceptible sites in purines and pyrimidines (Fig. 24-5). [58] A single genetic locus may be more likely to undergo a specific mutation, depending on the mutagen involved, as shown in Table 24-2. [59] The alkylator DMH preferentially methylates the O^6 position of guanine in DNA, although this is not the only site of damage. [60] Alkylation of guanine results in mispairing of DNA; the O^6-methylguanine acts like an adenine during replication, and the opposite strand pairs with a thymine (instead of cytosine) at replication. During the second round of replication, the newly inserted thymine pairs with an adenine, in place of the original guanine residue, resulting in an apparent guanine-to-adenine (G-to-A) mutation. Depending on the location of this mutation, it may have a trivial or profound effect on cell behavior. For example, when this change occurs in codons 12 or 13 of the *KRAS* gene, this signaling pathway is continuously activated, and a marked functional change occurs in the cell. Some mutations are silent, but the appearance of others results in a growth advantage for the cell, and the error is amplified through clonal expansion of the mutant cell.

TABLE 24-2 Mutagen-Specific DNA Damage in a Model Genetic Target

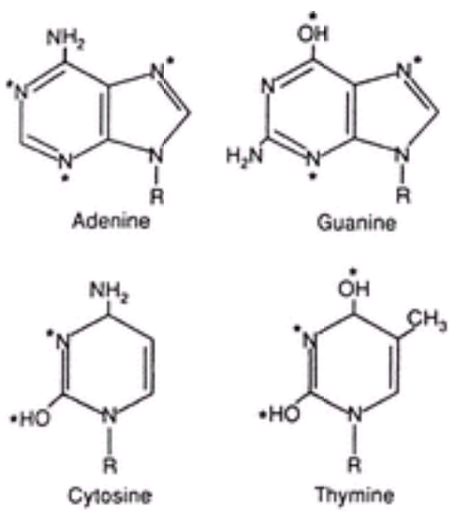

FIGURE 24-5. Alkylation sites in DNA bases. Specific sites on the purines adenine or guanine or the pyrimidines cytosine or thymine are targets for alkylation, as indicated by the asterisks. The adduct formation leads to mispairing during the next round of DNA replication. For example, O^6-methylguanine may mispair with

thymine instead of cytosine, and during the next cycle of DNA replication, the original guanine residue is replaced by adenine (i.e., a G-to-A transition mutation). Similarly, O^4-alkylated thymines lead to T-to-C transition mutations. Spontaneous deamination of cytosine results in uracil, which may mispair, and deamination of 5-methylcytosine creates a thymine residue (i.e., a one-step C-to-T transition mutation). Deamination of guanines and adenines creates xanthine and hypoxanthine, respectively. Thymine has no amino group to lose.

In rats treated with DMH, G-to-A mutations in *KRAS* develop in 66% of the colon cancers, indicating a strong selection for this mutation. [61] Other mutations may occur in locations that result in the inactivation of a gene essential for cell survival, and the cell is immediately eliminated. In the case of *KRAS,* mutations may occur anywhere in the gene. Mutational inactivation of the guanine nucleotide regulatory region of the *RAS* gene (at codons 12, 13, and 61) need occur in only one cell in the colon after carcinogen exposure because this cell will expand clonally, outgrow those around it, and create large numbers of progeny. Chronic inflammation is a common setting in which cancer develops in the gut, as in chronic esophagitis, chronic atrophic gastritis, and inflammatory bowel disease. Active forms of oxygen, such as hydroxyl and superoxide radicals, hydrogen peroxide, and singlet oxygen, arise as by-products of chronic inflammation and oxygen stress. These reactive species also mediate damage induced by ionizing and ultraviolet radiation. Reactive oxygen species create several forms of DNA damage, including strand breaks and single base changes. The two most common mutations induced by reactive oxygen molecules are the creation of 8-hydroxypurines, which result in misreading of those residues and adjacent pyrimidines. Because mispairing occurs in all four bases with almost equal frequency, it is not possible to predict what mutation this mechanism will induce.

DNA Rearrangement Gene expression may be modified by the gross rearrangement of DNA sequences. For example, in chronic myelogenous leukemia, a translocation occurs in which sequences from chromosome 22 are accidentally spliced in frame with the cellular oncogene *ABL* (i.e., the oncogene originally isolated from the Abelson murine leukemia virus and formerly designated v- *abl* or c- *abl*). The translocation creates a chimeric messenger RNA that translates into an abnormally large mutant ABL protein. [62] A similar rearrangement appears in Burkitt lymphoma, in which a reciprocal translation occurring between chromosomes 8 and 14 juxtaposes a portion of the immunoglobulin gene with the oncogene *MYC*. In this instance, *MYC* is not mutated, but the rearrangement causes an amplification of gene expression. [63] The protooncogene *BCL2* was first discovered in human B-cell follicular lymphoma as a t(14;18) chromosomal translocation, which placed *BCL2* under the control of the heavy immunoglobulin chain promoter. [64] Elevated levels of BCL2, a 26-kd protein, have since been found in adenocarcinomas of the colon and other tumors without this translocation. [65] BCL2 functions to inhibit programmed cell death and is often highly expressed in tumors resistant to chemotherapy. [66]

DNA Amplification DNA rearrangements can increase gene expression without the creation of additional copies of the gene. A normal cellular gene may also become a transforming gene through DNA amplification. This mechanism has attracted attention because resistance to methotrexate may be caused by amplification of the gene for dihydrofolate reductase. [67] Amplification of members of the *MYC* oncogene family are predictive of disease progression in neuroblastomas. [68] Some tumor cell lines have a large number of minichromosomes that exist independently as “double minutes” or are inserted into chromosomes (as homogeneously staining regions). Both represent examples of gene amplification. Amplification of *ERBB2* has been reported by several groups studying stomach cancers. [69], [70], [71] and [72] Other examples of amplified cellular oncogenes found in tumor cell lines include mutated *KRAS, MYB,* and *MYC*. [73], [74] and [75]

Altered Methylation of DNA Only a small percentage of the total genome is expressed in each tissue. One mechanism to control gene expression is to silence them through the stable, covalent methylation of the base cytosine at C-G sequences commonly found in promotors. DNA methylation is maintained by a specific methyltransferase, and the pattern of methylation is inherited in a stable way through successive generations of cells in a given tissue. A wide variety of genes are substantially hypomethylated in primary colorectal cancers in comparison with the adjacent normal mucosa. [76] These genes included the ?-globulin gene, which is normally hypomethylated only in blood precursor cells, and the ?-crystallin gene, which is normally hypomethylated only in embryonic lens tissue. [76], [77] Hypomethylation was found in even the smallest adenomas studied. The implications of hypomethylation are not clear. [78] Epigenetic alteration of DNA by methylation of promotor sequences of critical growth regulatory genes such as *p16* and *IGF2* have been described in microsatellite unstable tumors. [79], [80] The mechanism for promoter methylation is unknown, but there is evidence that the MMR system is not directly responsible for this activity. [80] The promotor of the MMR gene *hMLH1* has been shown to be methylated in about 10% of sporadic colorectal tumors (and is hypomethylated in surrounding normal tissue) with absence of *hMLH1* expression. [81] This indicates that DNA methylation is a mode of MMR gene inactivation in some sporadic tumors. The correlation between microsatellite instability and promoter methylation suggests that methylation abnormalities precede microsatellite instability in sporadic colorectal cancers, and that other genes with methylated promoters in microsatellite unstable tumors are additional targets of this poorly understood process.

DNA Deletion Cell proliferation is regulated by a family of genes known as *tumor suppressor genes*. These genes prevent inappropriate cell proliferation and regulate the number of cells in a tissue. Unlike oncogenes, these genes play a role in carcinogenesis through their inactivation. Even before the first tumor suppressor gene was identified, the presence of such genes was predicted by Knudson, [82] who proposed that some human cancers arise through a recessive mechanism in which both alleles of a tumor suppressor gene are independently inactivated. This concept was particularly important because it provided a framework for the inheritance of enhanced susceptibility to specific cancers. Loss of the first allele could occur as a dominantly inherited trait that is transmitted vertically in the germ line. Inactivation of the first allele can be phenotypically silent because the activity of the remaining second allele is sufficient to maintain the nonmalignant phenotype. However, such individuals are at extremely high risk for the development of cancer because only a single further inactivation mutation in a single cell may be required to trigger tumor development, usually at a young age. In contrast, the formation of sporadic tumors requires that two individual events occur independently, which decreases the likelihood of their occurrence in the same cell and increases the likelihood that they will occur later in life. Mutation of one allele followed by loss of DNA at the second allele (through chromosomal instability) is the “two-hit” mechanism for the complete inactivation of tumor suppressor genes.

Inactivation of DNA Repair

Nucleotide excision repair. DNA damaged by ultraviolet radiation, chemicals, and toxins is repaired by a complex of proteins that effectively excise the damaged DNA and religate the interrupted strand using the complementary DNA strand as a template. Collectively known as the *nucleotide excision repair complex,* the component proteins are designated *XP (xeroderma pigmentosum)* or *ERCC (excision repair cross-complementing)* in reference to the discovery of defective nucleotide excision repair in patients with xeroderma pigmentosum. [83] The XP and ERCC proteins are capable of recognizing and binding to damaged DNA, unwinding the DNA double helix by virtue of helicase activity, and excising 27 to 29 nucleotides containing the damaged DNA. [84], [85] and [86] The excision gap is then filled in by DNA polymerases. Some of the nucleotide excision proteins form an important transcription factor, TFIIH, which is involved in regulating the activity of all protein-encoding genes. [87], [88] Consequences of nucleotide excision repair defects include xeroderma pigmentosum, a disease characterized by sunlight-induced photodermatoses that result in skin cancers and neurological abnormalities. [84], [85] Cockayne syndrome and trichothiodystrophy, diseases characterized by neurological abnormalities and mental retardation without a major increase in skin cancers, are caused by mutations that impair the transcription function of nucleotide excision proteins. [84], [85] Malfunctioning nucleotide excision repair in cancers of the gastrointestinal tract has not been reported.

DNA mismatch repair. Unlike nucleotide excision repair, inactivation of the DNA MMR system is common in tumors of the gastrointestinal tract. Point mutations can occur in critical growth-regulatory genes and can accumulate in cells with a defective DNA MMR system (Fig. 24-6). The MMR system is responsible for maintaining replicative fidelity of DNA. Germ-line mutations in *hMSH2, hMLH1,* and *hMSH6* have been identified and clearly linked to hereditary nonpolyposis colorectal cancer (HNPCC), and somatic inactivation of *hMLH1* by promoter hypermethylation has been identified in sporadic tumors. [81], [89], [90], [91], [92] and [93] The contribution of *hPMS1* and *hPMS2* as a cause of HNPCC has not been proved, and no germ-line mutation in *hMSH3* has been identified in this syndrome. [94] The proteins that comprise the MMR system bind to nucleotide base-base mismatches of double-stranded DNA or loops of inaccurately replicated repetitive sequences (termed *microsatellite DNA*) and target the DNA area for excision, resynthesis, and ligation. [95], [96] The DNA sequences repaired by the MMR system have apparently escaped the normal editing function of DNA polymerase. Base mispairs, if not corrected by the MMR system, may cause nucleotide transitions or transversions, allowing a novel base to alter the authentic genetic sequence. [97] Such point mutations in genes that regulate cell growth may promote neoplastic growth.

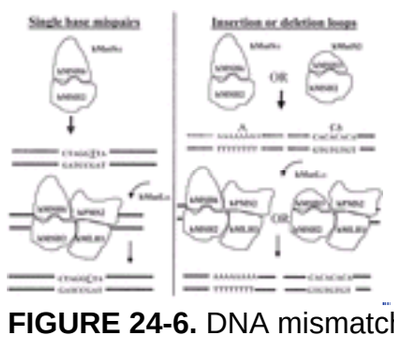

FIGURE 24-6. DNA mismatch repair (MMR). After DNA replication, mistakes that apparently have escaped the editing function of DNA polymerase are bound by the MMR proteins and targeted for repair. Single base mispairs and insertion or deletion loops of 1 base pair at microsatellite sequences can be recognized by hMutSa (heterodimer of hMSH2 and hMSH6), and insertion or deletion loops of greater than 1 base pair are recognized by hMutSß (heterodimer of hMSH2 and hMSH3). Subsequent recruitment of hMutLa (heterodimer of hMLH1 and hPMS2) to the altered DNA targets the area for excision, resynthesis, and ligation. Inactivation of the MMR proteins would permit these types of mutations to be passed to progeny cells.

Inherited Premutations Premutations may elevate familial cancer risk. [98] For example, a T-to-A mutation causing a substitution of lysine for isoleucine at codon 1307

in the *APC* gene is present in 6.1% of Ashkenazi Jews. [98] This missense mutation in itself is not thought to alter the function of the APC protein severely because most *APC* mutations are nonsense mutations, yielding a truncated protein. However, the T-to-A mutation converts the sequence AAATAAAA to $(A)_8$, a repetitive nucleotide tract that is relatively unstable during transcription and replication. In fact, the T-to-A premutation is associated with the acquisition of other nonsense mutations within and surrounding the $(A)_8$ tract, causing truncation of the APC protein. [98] This type of point mutation, which creates an intrinsically unstable repetitive DNA sequence, may lead to further mutations as a mechanism to inactivate gene function.

Genetic Mechanisms of Tumor Development

Gatekeeper, Caretaker, and Landscaper Genes Gatekeeper genes are cellular genes that directly regulate tumor growth by suppressing cell growth or promoting cell death. By definition, gatekeepers are tumor suppressor genes. It has been suggested that each human cell has only one or a few gatekeeper genes that are specific to that cell type. The esophagus, stomach, small intestine, colon, liver, and pancreas are different histologically, and the cells of these tissues express a different array of genes to perform their unique functions. The gastrointestinal organs probably do not share the same gatekeeper genes. Only the gatekeeper gene of the colon, *APC,* has been clearly identified. [99] Inactivation of a gatekeeper gene is the rate-limiting step for the initiation of tumor growth, and demonstrates a specific tissue distribution of that tumor. [4] As a tumor suppressor gene, inactivation requires alteration of the maternal and paternal alleles, fulfilling the Knudson two-hit hypothesis. [82] Restoration of the missing gatekeeper function in vitro leads to suppression of the neoplastic growth. Caretaker genes contribute to genomic stability, typically as part of a DNA repair process (e.g., *hMLH1* and *hMSH2*). Inactivation of a caretaker would lead to genomic instability by increasing the mutation rate of the cell, which may ultimately result in the mutation of a gatekeeper gene. This phenomenon might be likened to a constant and excessive exposure to mutagens. Therefore, caretakers indirectly promote neoplasia, and moreover they function as tumor suppressor genes. Restoration of caretaker function to a cancer cell will not affect its growth because these indirectly acting genes are not required for neoplasia. Defects in landscaper genes create an abnormal stromal environment for the epithelium, which contributes to neoplastic transformation. [100] For instance, chronic inflammation in ulcerative colitis affects the colonic epithelium, first with inflammation, but ultimately may cause neoplastic changes and progression to cancer. The regeneration that occurs to replace damaged epithelium may increase the probability of somatic mutations in an abnormal microenvironment. It has not yet been proved clearly that genetic defects in stromal cells are responsible for neoplastic changes in epithelial cells, but one could envisage alterations in intercellular signaling and cell-cell adhesion as mechanisms that influence surrounding cell behavior. Genetically, tumor initiation is caused by inactivation of both alleles of a gatekeeper gene. This may occur as the result of an inherited mutated allele (i.e., a germ-line mutation), coupled with somatic loss within a specific cell, or two somatic hits within a specific cell. For a caretaker gene to initiate neoplasia, both alleles must be inactivated; in addition, the resulting genomic instability must inactivate both alleles of the gatekeeper. It is believed that inherited susceptibility syndromes involving gatekeeper genes are associated with extremely high rates of cancer development, whereas inherited syndromes involving caretaker genes are associated with cancer rates lower than those of gatekeeper genes because some minimal number of additional mutations must occur. [4], [100] Inherited caretaker mutations would still be associated with cancer rates much higher than those in the general population.

Oncogenes The traditional concept of tumor initiation and promotion is inadequate to explain the full complexities of carcinogenesis. No single mutation or altered gene is sufficient to cause cancer in most cells. Multiple mutations, genetic deletions, and chromosomal rearrangements have been found in cancers that have been studied carefully. However, some single genes, when activated, can cause the phenotype of a cultured fibroblast cell line to become recognizably malignant. These initial cancer-causing genes were called *oncogenes,* and they were discovered by studying viruses that could transform chicken cells. [101] A number of oncogenes have been identified, and each can be activated by a point mutation that alters the activity of the gene product or by genetic rearrangement that results in an increase in gene expression (Table 24-3). [102]

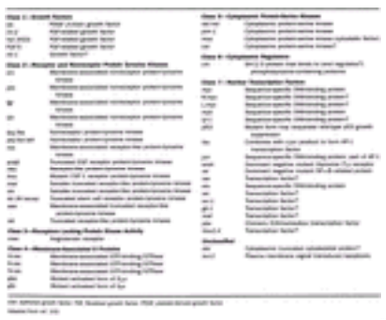

TABLE 24-3 Classification of Oncogenes and Gene Products According to Function

Some oncogenes produce growth factors (e.g., *SIS*), and when production is not regulated, the overproduced peptide ligand binds repeatedly and continuously to the receptor to cause unremitting stimulation. Other protooncogenes produce growth factor receptors (e.g., *ERBB2, FMS, KIT*). The most intensively studied protooncogenes are those involved in cellular signal transduction, including *SRC, RAS,* and *RAF*. These oncogenes are activated by point mutations that maintain the protein in a configuration such that signal transduction events are activated, regardless of whether the appropriate ligand or receptor is in place. Some of the protooncogenes, including *MYC, JUN,* and *FOS,* encode transcription factors, often nuclear phosphoproteins that interact with DNA and regulate gene transcription. Many of these genes were found because they had become incorporated into viral genomes, and because their expression had been deregulated by mutation or rearrangement, they created oncogenic viruses. In each case, these genes were capable of transforming cultured fibroblasts after infecting the cells. These oncogenes initially were designated v-oncogenes (e.g., v- *src*) because of their viral origin. Soon it was found that cellular protooncogenes could undergo activation in situ by point mutation or other genetic rearrangement and become oncogenic in the absence of a viral vector.

Tumor Suppressor Genes Boveri hypothesized as early as 1914 that the relentless growth of a malignant tumor was as likely to result from the inactivation or deletion of genes that inhibit proliferation as from enhanced activity of genes that promote growth. Tumors often have grossly abnormal karyotypes, and losses of chromosomal arms occur frequently. [103] Advances in molecular biology, specifically the use of restriction fragment length polymorphism and microsatellite analysis, have permitted the examination of specific chromosomal locations for genetic losses. Using such techniques, Vogelstein and colleagues [104] reported the widespread losses of genetic material in colorectal cancers and the deletion of specific genetic loci in most tumors. These investigators reasoned that identifiable hot spots for genetic loss would be the location of tumor suppressor genes relevant in the development of colorectal cancer. This hypothesis proved to be correct, and the concept has led to identification of a growing number of tumor suppressor genes in human cancer (Table 24-4). [105], [106] and [107] The mechanism of their activation may differ from gene to gene, but in all cases, it is the inactivation of the protein function that plays a role in carcinogenesis.

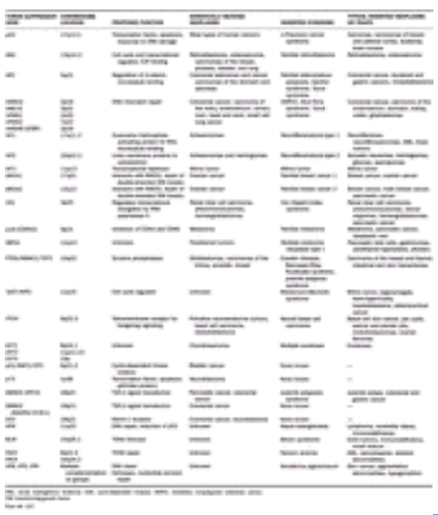

TABLE 24-4 Tumor Suppressor Genes Involved in Human Cancers

Perhaps the most important of these is the *p53* gene, which normally acts to prevent the cell from beginning new DNA synthesis or cell division. It serves as a critical regulatory gene to prevent inappropriate proliferation. [108] When DNA is damaged, the p53 protein becomes activated, which turns on the transcription of a variety of genes. These include activation of *WAF1/CIP1,* a gene encoding a 21-kd protein that inhibits cyclin-dependent kinase 2, thus arresting the cell in the G_1 phase of the cell cycle and preventing the replication of damaged DNA. [109], [110] and [111] p53 also stimulates the transcription of *GADD45,* and the protein gene product, in turn, complexes with proliferating cell nuclear antigen (PCNA), a necessary component for resynthesis of DNA after damaged DNA is removed. [112] By stimulating synthesis

of WAF1/CIP1 and GADD45, p53 effectively halts DNA replication and stimulates DNA repair. p53 protein also plays a critical role in programmed cell death, or apoptosis, when DNA is damaged, perhaps beyond repair. [109], [113], [114] and [115] Mutation of *p53,* therefore, would prevent cell death in the wake of DNA damage. Inactivation of *p53* in cancer involves two steps. First, one *p53* allele undergoes a missense point mutation, which typically occurs in a portion of the molecule involved in the binding to DNA. It is controversial how the presence of one wild-type together with one mutated copy of p53 protein alters the growth characteristics of a cell. However, most colon cancers have lost their wild-type *p53* gene, leaving a single mutated copy of the gene in the nucleus. [116] The peanut fungus *Aspergillus fumigatus* produces aflatoxin ß $_1$, which may cause hepatocellular carcinoma by mutating *p53* at a critical codon. [117] Other mechanisms have developed in certain cancers to inactivate p53 by interrupting its evolutionarily conserved regions (Fig. 24-7). These include the binding of viral proteins to p53 (i.e., JC virus or SV40 T antigen, human papillomavirus E6 protein, and adenovirus E1b protein), and binding of amplified cellular proteins to p53 (e.g., mdm2 oncoprotein in sarcomas). [40], [118] Mutant *p53* (with concomitant lack of normal *p53*) appears to be overexpressed in tumors, including colorectal tumors, and can be detected by immunohistochemical methods. [119] This overexpression is the result of enhanced stability of the mutant p53 protein, allowing its accumulation within the cell. [118], [119]

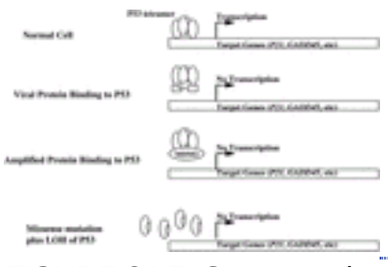

FIGURE 24-7. Some mechanisms for inactivating p53. The p53 protein binds as a tetramer to a p53-binding site in the promotor region of a target gene to activate genes involved in growth suppression and DNA repair. Viral proteins such as the SV40 T antigen, human papillomavirus E6, and adenovirus E1b can bind p53 protein to prevent its transactivating function. Similarly, amplified cellular proteins such as MDM2 can bind p53 protein to prevent promotor binding. In colon cancer, the typical p53 inactivation involves a missense mutation of one allele and loss of heterozygosity (*LOH*) of the second allele, leaving only mutated protein available. The mutated p53 protein may have limited or no binding to promotor sites to initiate transcription at target genes.

The tumor suppressor gene concept accommodates the notion of familial predisposition to cancer. The first example of this was found in familial retinoblastoma, in which there are germ-line mutations at the *RB1* locus. [105] The *RB1* gene has a tumor suppressor function in many tissues, including the gastrointestinal tract, but its inactivation is the limiting step (gatekeeper) only for the development of retinoblastoma. [120], [121] In familial retinoblastoma, every retinal epithelial cell contains one copy of a mutated *RB1* gene and one wild-type gene. The chance occurrence of a mutation in the wild-type *RB1* allele in any cell is sufficient to inactivate completely the tumor suppressive activity of the *RB1* gene. Because this event needs to occur in only a single cell to produce a tumor, the incidence of retinoblastoma is high in families with a mutated *RB1* allele. Essentially every child who inherits the mutated *RB1* allele is at risk for the development of bilateral tumors at an early age. In contrast, unaffected individuals require two inactivational hits in a single cell, one on each allele, for retinoblastoma to occur; accordingly, their sporadic occurrences are rare. Retinoblasts disappear from the eye after early infancy, so this disease is not seen later in life. Germ-line mutations at tumor suppressor gene loci may be associated with a normal phenotype at birth, but they are associated with very high risks for cancer. This is the outcome in Li-Fraumeni syndrome, in which there is a germ-line mutation in the *p53* gene; affected individuals are phenotypically normal at birth but develop breast cancers, sarcomas, and brain tumors at an early age. [105] An animal model for this disorder exists in mice made deficient for the *p53* gene by homologous recombination; these mice can survive with no copies of the *p53* gene, which underscores the redundancy and adaptability of the control over cell growth. [122] Two more recently discovered genes, *p63* and *p73,* have some functions similar to those of p53 protein, and perhaps redundant. [123], [124] and [125] Similar to p53, both p63 and p73 can form homooligomers, bind DNA, activate transcription from p53-responsive genes, and induce apoptosis. [125], [126] and [127] However, in contrast to p53, p63 and p73 give rise to multiple functionally distinct protein isoforms, some of which can function as “dominant negative” proteins, blocking the function of the corresponding full-length protein. [128] In addition, *p63* and *p73* are only rarely mutated in tumors and thus do not function as classical tumor suppressor genes. Furthermore, unlike *p53*-/- mice or patients with Li-Fraumeni syndrome, *p63*-/- and *p73*-/- mice are not tumor prone but instead manifest multiple developmental abnormalities. [129], [130] and [131] Germ-line *p63* mutations in humans cause ectrodactyly–ectodermal dysplasia–clefting (EEC) syndrome, an autosomal dominant disorder characterized by ectrodactyly, ectodermal dysplasia, and cleft lip with or without cleft palate. [132] Thus, although p63 and p73 have some overlapping functions and interactions with p53, they are not tumor suppressor genes like p53, and mutations do not pose the same high risk for cancer as p53 mutations.

Clonal Expansion A variety of genetic techniques have demonstrated that neoplasms are derived from a single cell, but most human cancers have multiple mutations, and it is unlikely that a single cell undergoes all these genetic changes at once. [133] The concept of multistage carcinogenesis assumes that gastrointestinal epithelium undergoes a constant barrage by factors that threaten to damage DNA, but the mutations occur one by one at critical locations in genes related to the control of cell growth or survival. Some of these mutations may inhibit the function of a gene essential for the life of the cell, which is lethal to that cell. Other mutations provide the cell with a slight growth or survival advantage over the rest of the cells in the tissue. In these instances, the mutated cells proliferate faster than the surrounding cells and gradually represent a larger proportion of the tissue. In other instances, a mutated cell may be less capable of repairing DNA damage, which facilitates the accumulation of mutations in succeeding generations. This expanded pool of cells continues to be susceptible to additional mutations. Because of the redundancy in the cellular mechanisms regulating proliferation, loss of any individual function is insufficient for the transformation of a normal cell. The accumulation of cooperating lesions is required to overcome the regulated cell growth. Periodically, an individual cell within an expanding clone experiences another mutation that adds to its growth or survival advantage, and the progeny of this cell then overgrow the population from which it originated. Successive waves of clonal expansion occur by the chance accumulation of new mutational events, which add to the survival advantage of the cell in an evolutionary manner. In early-stage neoplasms, there is evidence for only a small number of mutational events; later in neoplastic progression, however, the cell has a larger number of genetic lesions. [22], [134] Figure 24-8 demonstrates tumor progression as successive expansions of cellular clones with new growth characteristics.

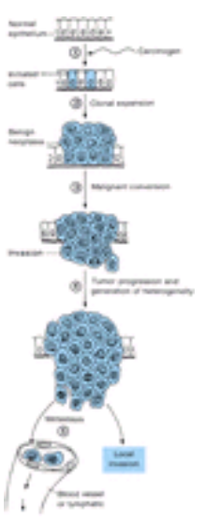

FIGURE 24-8. Tumor progression. This scheme illustrates the natural history of carcinogenesis. The first step represents a genotoxic event caused by a carcinogen. When a critical number of genes are mutated (the specific site and number remain speculative), a cell becomes neoplastic (step *1*). Clonal expansion gives rise to a benign neoplasm (step *2*). Unless additional events occur, a benign neoplasm may persist or grow indefinitely without becoming malignant. However, if additional genetic events occur, cells appear within a benign neoplasm that are capable of malignant behavior, such as invasion and metastasis (step *3*). A malignant neoplasm may invade locally and damage the host and surrounding organs. However, the genome of the malignant neoplasm is intrinsically unstable, and despite its monoclonal origin, the cancer becomes more heterogenous with time (steps *4* and *5*). Clinically, the generation of tumor cell heterogeneity increases the likelihood of metastasis to one or more distant sites.

Genomic Instability and Multistep Carcinogenesis Through the elegant work performed in the laboratories of Fearon and Vogelstein [134] and others, a conceptual interpretation of the genetic basis of colorectal cancer has been developed. This framework accommodates the growing evidence that most cancers develop slowly and that multiple genes are involved in the process. Certain types of genetic damage tend to occur early, and different types occur later. It is not yet clear how strictly the sequence of events is followed in most neoplasms, but not all cancers appear to accumulate every mutation or chromosomal deletion during their development. Some type of genomic instability is required to account for the mutations that occur in multistep carcinogenesis. In most colorectal cancers, the process could be called *chromosomal instability*. Chromosomal segments or entire chromosomes are deleted, duplicated, or rearranged, so that the normal diploid nucleus is changed

into an aneuploid one. A common finding in this pathogenic pathway is the allelic mutation of a tumor suppressor gene (e.g., *APC* or *p53*), followed by the loss of the remaining normal allele (termed *loss of heterozygosity,* or *LOH*). [104], [135] As an increasing fraction of allelic arms is lost, the prognosis for colorectal cancer worsens. Patients with tumors that have lost more than 20% of tested alleles are significantly more likely to die of their disease than those with tumors that have not, and this characteristic is a more powerful prognostic factor than the Dukes stage or the histological grade of the tumor. [135] It is not known what specific cellular function becomes disturbed, resulting in multiple LOH episodes throughout the genome. Chromosomal instability occurs in the smallest adenomas, suggesting that it occurs very early during colorectal neoplasia. [136] It has been hypothesized that structural changes to the chromosomes and abnormal mitoses are operative in the development of chromosomal instability. Mitotic checkpoint gene mutations, such as those found in *BUB1,* have been found, but only rarely in colorectal cancers. [137] A mutant APC protein, which is inactivated in the earliest stages of colonic neoplasia, is associated with defective chromosome segregation. [138], [139] Wild-type APC localizes to the kinetochore during mitosis and forms complexes with BUB1 and BUB3 mitotic checkpoint proteins. [139] Although mutant APC may contribute to ongoing chromosomal instability in a colonic cell, it is not clear how this causes the initial chromosomal instability responsible for APC inactivation. Other proteins, such as human securin, a protein necessary for completion of the anaphase portion of mitosis, may help maintain euploidy. [140] Telomerase dysfunction also has been implicated as a mechanism for chromosomal instability in mouse models. [141] Foreign proteins such as the T antigen from the polyomavirus JC Mad-1 strain have been found in colorectal cancers. [40], [142] T antigens are capable of inducing aneuploidy in cells and are known to inactivate important cell cycle proteins, such as p53 and RB. [40], [142] The mechanism for chromosomal instability remains under intense study. The early stages of carcinogenesis in the colon appear to occur slowly through the accumulation of mutations and LOH in a variety of loci. However, transition to the malignant stage is associated with widespread genomic instability and LOH throughout the genome, which accelerates the likelihood that tumor suppressor loci will be lost, including genes that may suppress metastatic behavior. Studies have shown that mutation of one *p53* allele coupled with LOH of the second allele coincides with the appearance of carcinoma within an adenomatous polyp, making *p53* the gatekeeper for malignant transformation in the colon. [143], [144] and [145] This model of multistep carcinogenesis reflects the consequences of gene dysfunction. A malignancy occurs when a sufficient combination of these mutated genes accumulates. In some instances, a sequential pattern for the occurrence of their metastasis may be required. The following sequence of events appears to be relevant to the chromosomal instability pathway of tumorigenesis in the human colon (Fig. 24-9). [134], [143] Hypomethylation of DNA occurs in the smallest adenomas and may be one of the early events in carcinogenesis, but the mechanism by which this occurs, the specific genes involved in the process, and its role in carcinogenesis are unknown. [76], [77] and [78] Mutations (and LOH) in the *APC* locus are also found in small adenomas, and these events have been linked to dysregulated proliferation on the basis of their role in FAP. [146] Mutations at the *APC* locus are typically nonsense mutations or insertions and deletions that produce frameshifts resulting in downstream stop codons. Mutations in the *KRAS* and *p53* genes are found in larger adenomatous polyps and are thought to play a mechanistic role in supporting neoplastic growth. Indeed, activation of KRAS seems to facilitate the exophytic growth of adenomatous polyps, as *KRAS* mutations are more rare in flat adenomas and cancers. [147]

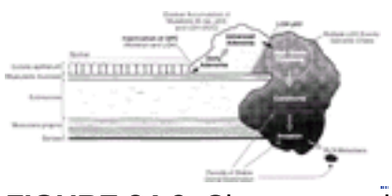

FIGURE 24-9. Chromosomal instability model of genetic events mediating neoplastic progression during colorectal tumorigenesis. This type of genomic instability is present in familial adenomatous polyposis and in about 85% of sporadic colorectal cancers in which segments or whole chromosomes are deleted, duplicated, or rearranged, forming aneuploid tumors. *APC* is inactivated by mutation and loss of heterozygosity (*LOH*) at the transition between normal colonic epithelium and adenomatous epithelium. Thereafter, successive rounds of clonal expansion occur, with accumulation of mutations (i.e., *KRAS, p53,* others) and LOH (i.e., chromosome 18q) that allow acceleration of growth and dominance over less-advantaged clones. *p53* inactivation by LOH of 17p13.1 first occurs in high-grade dysplasia, and defines the transition between benign and malignant neoplasia. Successive genetic events as well as prior genetic changes enable the colon cancer to invade and metastasize to regional lymph nodes (*RLN*s), and ultimately to distant organs such as the liver. (From ref. [143].)

Point mutations play a prominent role in the early stages of neoplastic progression, typically by inactivating the function of tumor suppressor genes. APC protein translation may actually halt after the creation of a nonsense mutation–induced gene. It has been hypothesized that mutant APC proteins may bind to the wild-type proteins and interfere with their function through a dominant negative mechanism. [148] In the case of *KRAS,* mutations inactivate the portion of the protein involved in halting signal transduction. [20], [149] In the case of *p53,* the mutations, which occur in a broad hot spot in the middle of the p53 protein, interfere with the ability of the protein to activate gene expression in the nucleus. [118] It is uncertain whether the mutant p53 protein can bind to and inactivate the wild-type protein and also act with a dominant negative mechanism, as suggested for APC. A second form of genomic instability that does not result in LOH events is involved in the genesis of about 12% to 15% of colon cancers. These tumors demonstrate multiple errors in DNA sequences throughout their genome, which results from a failure to edit errors made during DNA replication. [150], [151] and [152] This was first observed in microsatellite sequences, which are repetitive sequences that require the DNA mismatch repair system for proper replication because of an intrinsic tendency to be copied inaccurately. In certain tumors, minor changes occur in the DNA microsatellite length, termed *microsatellite instability*. Microsatellite sequences are ubiquitous and occur 100,000 times throughout the genome, and half or more may be mutated in tumors demonstrating microsatellite instability. Tumors with microsatellite instability demonstrate significantly less LOH than is commonly seen in colorectal cancers with chromosomal instability. Thus, either chromosomal instability or microsatellite instability develops as the type of instability within a tumor genome. [153] Microsatellite instability is more common in cancers of the proximal colon and is associated with diploid tumors and increased patient survival. [150], [154] This hypermutable phenotype appears to be the mechanism for rapid neoplastic progression in HNPCC and sporadic tumors that exhibit microsatellite instability. [152], [155] HNPCC and about 12% to 15% of sporadic colorectal tumors exhibit microsatellite instability. An analysis of repetitive sequences in tumor tissue led to the identification of other inactivated genes in these tumors, suggesting a second pathway for developing neoplasia. Each of these targeted genes contains a microsatellite within its (exonic) coding sequence, making it susceptible to mutation in the absence of DNA MMR activity. For example, a repeat of ten adenines in the *TGFBR2* gene has undergone frameshift mutation in 90% of colorectal tumors with microsatellite instability, inactivating this receptor, [27], [33] and the cell escapes growth suppression from the ligand TGFB1. In the remaining 10% of tumors with microsatellite instability, it is estimated that frameshift mutations of *IGF2R* at its polyguanine sequence uncouple TGFB1 growth suppression. [29] Another important target of microsatellite instability is *BAX,* a member of the BCL2 gene family. BAX heterodimerizes with BCL2 within the cell, and the mix of the heterodimer determines the cell's commitment to programmed cell death. *BAX* contains a polyguanine $(G)_8$ tract that is mutated in 50% of colorectal cancers with microsatellite instability, and in 64% of gastric adenocarcinomas with microsatellite instability. [156], [157] Mutation of *BAX* (by diminishing the ratio of BAX to BCL2) prevents programmed cell death. Other genetic targets identified include the MMR genes *hMSH3* and *hMSH6,* which may broaden and accelerate the accumulation of mutations. [158] A comprehensive search in which an online genetic database for coding microsatellites and subsequent colorectal tumor analysis were used revealed nine genes that were mutated in more than 20% of the tumors. [159] Aside from *TGFBR2, BAX,* and *hMSH3,* these included the activin type II receptor gene, *SEC63* (human homologue of a yeast DnaJ-like endoplasmic reticulum translocon component protein gene), *AIM2* (an interferon-inducible gene), NADH-ubiquinone oxidoreductase B14.5B subunit gene, *KIAA0977* (probable human homolog of the mouse embryonal protein *cordon-bleu*), and *PA2G4/EBP1* (homolog of the mouse cell cycle protein p38-2G4). [159] The role of these genes in the pathogenesis of tumors with microsatellite instability is not currently known. Several lines of evidence indicate that the Wnt signaling pathway, including the gatekeeper APC or other proteins in this signaling pathway, are affected in microsatellite-unstable tumors (see Fig. 24-2). Some *APC* mutations have been found in colorectal tumors with microsatellite instability. [160] In addition, ß-catenin, a protein that binds to and is normally degraded by the APC protein, undergoes a stabilizing mutation in microsatellite-unstable primary tumors and cell lines. [161] Activating mutations in ß-catenin signal the cell to proliferate incessantly, [162], [163] and the mutation prevents interaction with the APC protein and its subsequent targeting for degradation. TCF-4, to which ß-catenin binds as a partner in the nucleus for transcriptional activation, is mutated at its polyadenine site in 39% of primary colorectal cancers with microsatellite instability. [164] Simultaneous analysis of various components of the Wnt pathway in colorectal cancer indicates that there is frequently a perturbation in the Wnt signaling pathway, including mutations in APC, ß-catenin, TCF-4, or axin I (involved as a scaffold protein and negative regulator or Wnt signaling). [165], [166] Taken together, colorectal tumors exhibiting microsatellite instability appear to have initiated their growth through deregulation of Wnt signaling, which has the identical consequence as inactivating the gatekeeper APC. As described for the chromosomal instability pathway of tumorigenesis, inactivation of several genes likely promotes the progression of the neoplastic process in tumors with microsatellite instability, but a clear sequential pattern has not been elucidated. Mutations of *TGFBR2* and *BAX* appear to be late in the adenoma-carcinoma progression because these mutations occur in high-grade dysplasia at the interface between adenoma and carcinoma. [167], [168] A proposed scheme for the progression of microsatellite-unstable colorectal cancer is presented in Figure 24-10.

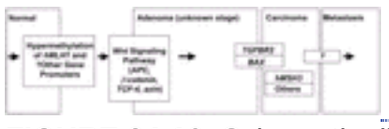

FIGURE 24-10. Schematic diagram of colorectal tumor progression in microsatellite unstable tumors. This type of genomic instability is present in hereditary nonpolyposis colorectal cancer (germ-line mutation of a DNA mismatch repair [MMR] gene) and in about 15% of sporadic colorectal tumors (hypermethylation of the *hMLH1* promoter). These tumors tend to be diploid because chromosomal arrangements typically do not occur. Biallelic inactivation of an MMR protein occurs to disrupt the DNA MMR system completely. The Wnt signaling pathway, including the gatekeeper protein APC and other proteins in this pathway, are deregulated. A number of genes with encoded microsatellites are targeted for mutation in microsatellite-unstable tumors, including *hMSH3, TGFBR2,* and *BAX*. *TGFBR2* and *BAX* mutations appear at the transition point during malignant conversion. Other mutated genes are likely to be involved in forming the final tumor and in mediating metastasis.

Much has been learned about multistep carcinogenesis through studies of colon cancer, and the principles may be applicable to cancers of many types. Multiple genetic events are required to overcome the redundancy in regulatory controls in the cell. These events accumulate during successive waves of clonal expansion and cooperate to produce tumor progression. The exact impact on growth regulation is not entirely known for any single genetic lesion. Because a small proportion of cancers lack the genetic events seen in most others, it is reasonable to speculate that there are additional mechanisms of tumor progression, and there is evidence for alternative pathways. [80], [147] One hypothesis involves widespread hypermethylation of gene promoters as a mechanism for epigenetic inactivation and gene silencing. [80] Indeed, sporadic colorectal cancers with microsatellite instability lose DNA MMR activity as a consequence of hypermethylation of *hMLH1*. [169], [170] Other genes commonly inactivated by hypermethylation at promoter CpG islands include the cell cycle regulator *p16,* the *THBS1* angiogenesis inhibitor, the estrogen receptor growth suppressor, the *TIMP3* metastasis suppressor, and the *O*[6]-methylguanine DNA methyltransferase DNA repair gene. [171] CpG islands are regions of DNA 0.5 to 2 kilobase pairs long, rich in cytosine-guanine dinucleotides, that are found in the 5' region of about half of all human genes. Methylation of cytosines within the CpG islands is associated with loss of gene expression and can be observed physiologically (as with X chromosome inactivation and aging), but also with neoplasia. A distinct pathway for colorectal cancer development, termed the *CpG island methylator phenotype (CIMP),* has been proposed. [172] In a study of colorectal cancers, the CIMP phenotype segregated with tumors that demonstrated microsatellite instability. [172] However, in a study of colonic adenomas, the CIMP and microsatellite instability pathways appeared to be distinct. [171] The CIMP phenotype seems to occur early in colorectal tumorigenesis, but the mechanism for its occurrence is not known.

TUMOR FORMATION AND BEHAVIOR

Molecular Histology

Much of the understanding of the genetics of gastrointestinal tract carcinogenesis has come from analyzing various histological stages of tumors. Neoplasia originates from a single cell that has developed a genetic alteration, and then grows at variable rates to larger sizes with differing histological morphology. In the colonic adenoma, there is continued mitosis and lack of cellular differentiation, so that the principal proliferative compartment moves from the base of the crypt to involve the entire crypt. Persistent replication of cells near the crypt surface, in concert with retarded cell maturation and extrusion, results in an increased number of replicating surface cells. Continued proliferation coupled with lack of cell death leads to a downward infolding of epithelial cells, which branch and interpose themselves between normal crypt elements. This process results in the characteristic branching glandular pattern of tubular adenomas. As the polyp continues to grow, the growth pattern of the underlying mesenchyme may be enhanced. [173], [174] If mesenchymal proliferation matches the rate of epithelial growth, resistance to continued epithelial proliferation is unimpeded, resulting in long, fingerlike projections of the glandular elements. This histological picture is characteristic of villous adenomas. Perhaps more commonly, an adenoma demonstrates a combination of these two histological types, a tubulovillous adenoma. Villous adenomas tend to be larger than tubular adenomas, consistent with the concept that the villous histology is associated with enhanced growth characteristics. Subsequently, some adenomas develop characteristics of carcinoma, giving rise to the malignant polyp. Malignant cells at some point develop the capacity to invade locally and the potential to spread to distant organs.

Each histological stage of neoplasia has been characterized genetically by analyzing the cells present in the specimen. Immunohistochemistry, RNA and DNA extraction, and in situ polymerase chain reaction are some of the techniques used to determine the genetic abnormalities of the cells. The sequential biologic process of tumor formation is now correlated to specific morphologic characteristics of tumors in their various stages. [143], [147]

The general features of the colorectal neoplastic model may be applicable to other gastrointestinal neoplasms. In esophageal, pancreatic, and hepatic neoplasms, it is difficult to obtain tissue samples at each stage of a multistep process. These tumors do not have a readily identifiable precursor lesion to obtain and study. Tumors of these organs, particularly of the esophagus and pancreas, often present in advanced stages, precluding an opportunity to obtain specimens at early stages of the disease.

Loss of Proliferative Control and Clonal Expansion

Proliferation is normally tightly regulated and confined to one compartment of the epithelial unit within the gastrointestinal tract. During periods of epithelial repair, this regulation is relaxed, and the rate of proliferation increases. When a sufficient number of cells has been produced, the rate of proliferation slows to match the rate of cell loss.

A chance genetic event may occur in one cell that has not yet been committed to programmed cell death, enabling it to undergo more cycles of replication than any neighboring cell. When this event affects the gatekeeper gene or pathway function, neoplasia is initiated. In the colon, the proliferative pool of cells migrates from its normally restricted site at the base of the crypt toward the apex of the crypt. The expanded pool of cells may be seen in magnified histological preparations of grossly normal-appearing colonic mucosa stained with methylene blue, termed *aberrant crypt foci*. [175] Aberrant crypt foci are collections of thickened epithelium with asteroid or oval-shaped glandular lumens. [176] The number of aberrant crypt foci appears to be higher in patients with a prior history of colon cancer, and highest in patients with FAP. [175] Most larger aberrant crypt foci do not appear to be neoplastic, although they contain mutations in the *KRAS* oncogene. [176], [177], [178], [179] and [180] However, dysplastic aberrant crypt foci have been reported to contain *APC* mutations. [177], [179] Dysplastic aberrant crypt foci may be the first histological counterparts that reflect loss of the gatekeeper gene function. The Wnt signaling pathway, of which APC is a part, appears to be critical for the initiation of colonic neoplasia and is the gatekeeper for colonocytes. [99], [146] Loss of the Wnt signaling pathway uncouples the cell from programmed cell death, the ability to regulate intracellular growth signaling, and recognition of cell-cell adhesion. [181], [182] Because these cells continue to proliferate but are incapable of invasion, they tend to expand locally and develop into an adenoma. Additional growth permits the formation of a polypoid lesion. This is a common stage in the development of colonic neoplasia but uncommon in the natural history of neoplasia of the esophagus, stomach, and pancreas.

The liberation from typical growth restraints allows the cell to grow at a faster rate than any surrounding cell. Subsequently, a new genetic event may occur in one of these cells, further accelerating growth. Each advantage gained in growth, beginning with the adenoma and subsequently progressing to carcinoma, occurs by successive waves of clonal expansion. Each clone gains its advantage by a genetic alteration within the cell that allows more successful proliferation than its predecessors. In the colon, it appears that mutations in the *KRAS* gene, and possibly the *p53* gene, participate in this process. Alterations in the KRAS signaling pathway may permit enlargement of the adenoma to an exophytic mass. [147] Flat cancers that have mostly been observed in Japan appear to lack *KRAS* mutations. [147], [183], [184] In microsatellite-unstable tumors, clonal expansion may be mediated in part by alterations in the Wnt signaling pathway and the TGFB1 signaling pathway, and by escape from BAX-mediated programmed cell death. [27], [33], [156], [157], [165], [166] In the gastrointestinal tract, the expanded pool of hyperproliferative cells undergoes new DNA synthesis in the hazardous environment of the gastrointestinal lumen. The unrestrained proliferation permits the clonal expansion of mutated cells, hastening the appearance of alterations that may provide a growth advantage.

The development of adenomas is a reversible biologic event in the colon, at least by gross appearance. Small, sporadic adenomatous polyps may spontaneously disappear, and small rectal adenomas often spontaneously disappear from patients with FAP after subtotal colectomy. [185] Patients who have hundreds of colonic adenomas may experience regression of these lesions after the administration of the nonsteroidal antiinflammatory drug sulindac or the COX-2 inhibitor celecoxib. [14], [186], [187] and [188] The mechanism underlying adenoma involution in response to sulindac or the COX-2 inhibitor celecoxib is the activation of programmed cell death. [12], [13] and [14] The incidence of gastrointestinal cancer is significantly reduced in patients taking aspirin and other nonsteroidal antiinflammatory drugs (see Chapter 91). Pharmacological therapies that may block neoplastic progression at this early stage are being evaluated.

Malignant Conversion

At least two genetic mechanisms permit destabilization of the cellular genome and the appearance of cancer. Both mechanisms involve the breakdown of a homeostatic mechanism by which the genome is faithfully reproduced.

Chromosome instability reflects a failure in the symmetric separation of sister chromosomes and results in LOH events throughout the genome. When LOH occurs at a tumor suppressor gene locus, additional clonal expansion is greatly favored. The inability to divide the tetraploid nucleus into two perfect diploid pairs at mitosis results in the random generation of aneuploid cells. Cells that accumulate extra chromosomal pieces become hyperploid, and those that lose chromosomal arms become hypoploid. Randomly occurring events that provide a growth advantage result in clonal expansion of the favored cell. Flow cytometry has been used to detect expanded clones with abnormal DNA contents. [189], [190] Highly aneuploid tumors exhibit increased virulence. [190], [191] The abnormal clones occur focally rather than diffusely throughout the tumor, and evidence suggests that metastatic lesions are derived from these aneuploid clones. [192] In the colon, the histological appearance of high-grade dysplasia coincides with the complete genetic inactivation of *p53* in chromosome-unstable tumors. [143]

A second type of genetic lesion capable of increasing the rate of mutation is inactivation of the DNA MMR system. This form of genomic instability leads to microsatellite instability, and allows an accumulation of mutations throughout the genome. Microsatellite instability leads to an increase in point mutations, as well as insertion/deletion mutations at simple repetitive DNA sequences (microsatellites). Cancer cells with microsatellite instability are typically diploid. [151] The polymerase chain reaction can detect the presence of the inaccurately copied microsatellite DNA. Because of the high mutation rate, it is probable that tumors with MMR defects may progress to carcinoma at a more rapid pace than other tumors. [152], [155] Malignant conversion in colorectal tumors with microsatellite instability involves mutation of *TGFBR2* and *BAX,* both of which appear in high-grade dysplasia at the interface of defining malignancy—the adenoma-to-carcinoma transition. [167], [168]

Malignant cells are capable of continued growth, invasion, and formation of distant metastases. The malignant phenotype is intrinsically unstable and gives rise to additional phenotypic diversity. Malignant tumors are dynamic by virtue of ongoing genomic instability. Some of the newly created cells lose proteins essential for survival and die. Other genetic events result in the generation of more virulent clones that overgrow and replace the parent tumor.

Metastasis

The metastasis of tumor cells is not a random or accidental process. For a tumor to metastasize, it must degrade the basement membrane and associated matrix components, migrate through the subtending connective tissue, enter into a lymphatic or blood vessel, migrate away from the parent tumor, avoid a gauntlet of naturally occurring defensive mechanisms, emigrate from the efferent vessel, lodge at a distant site favorable for its growth, and recruit a sufficient blood supply (Fig. 24-11). [193], [194] and [195] Metastasis occurs as a late event in the natural history of a tumor, because time is required for the gradual evolution of cells capable of all these behaviors.

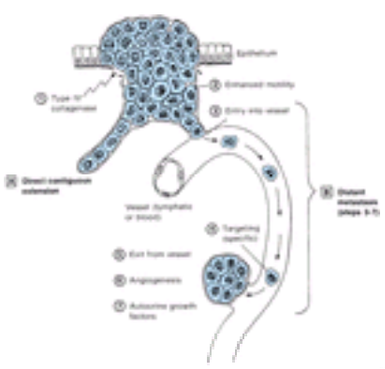

FIGURE 24-11. Tumor metastasis. Successful tumor metastasis requires several coordinated events. Specific collagenases are required to transgress the basement membrane (step *1*). The tumor may grow by direct contiguous extension into surrounding organs (**A**). Alternatively, tumor cells may enter a vascular structure for distant metastasis to occur (**B**). The spread of malignant cells is enhanced by the development of cellular motility (step *2*). Access to a vascular structure is permitted by the expansion of the growing tumor, the motile cells, and the ingrowth of blood vessels, which is stimulated by angiogenesis factors (steps *3* and *6*). Entry into the circulation is not sufficient to ensure distant metastasis. Specific targeting mechanisms and adaptations that would enhance survival at a distant site are required for distant metastatic spread.

The characteristics required for metastasis are quite specific. A malignant neoplasm, for example, may acquire the ability to digest the basement membrane and slowly expand into the submucosa, so that it is by definition malignant; however, it may not acquire other properties required for entry into the circulation or survival in a distant organ. Such a tumor is pathologically malignant and may become a bulky mass, but there may be a relatively long period during which it can be detected and successfully removed. Alternatively, a highly metastatic clone of cells emerging from a malignant tumor may give rise rapidly to a large number of metastatic units, and the host may suffer a brisk and relentless downhill clinical course.

Our insight into the metastatic process is insufficient to predict the clinical course of a malignancy by studying tumor cells in vitro. Tumor cells are frequently present in the portal and peripheral circulation at the time of colectomy for a colon cancer. [196], [197] However, the presence of cytologically identifiable tumor cells in the circulation has no prognostic significance because most of these cells are incapable of growing at a distant site and are susceptible to cytolysis by natural killer lymphocytes and other immunologic mechanisms. Clumps of cells appear to be more likely to survive as a metastasis, perhaps because individual cells in the clump have only some of the functions required for successful metastasis, but the entire “social unit” is capable of growing communally.

Tumor cells require a blood supply to survive. Oxygen can normally diffuse 100 to 200 µm from a blood vessel, which is roughly four to ten cell diameters, depending on the tumor. To facilitate their growth at metastatic sites, tumors stimulate the growth of capillaries. Tumor-associated angiogenesis appears to be mediated by a variety of different tumor growth factors, including angiogenesis factors. The presence of such growth factors can be demonstrated by the tumor’s ability to recruit endothelial cells to form capillaries in vitro. [198] Inhibition of vascular endothelial growth factor in mouse models of colon cancer blocks tumor growth and induces apoptosis by inhibiting angiogenesis and endothelial cell survival. [198] Evidence suggests that the expression of transcripts in the endothelium of tumor blood vessels differs from that in the endothelium of nontumor vessels. [199] Therapies to interfere with tumor angiogenesis are under development, including COX-2 inhibitors and other drugs. [200], [201]

The sites to which malignant cells metastasize may appear to be random, based purely on the blood supply of an organ, but the process is more complicated, and tumor cell targeting to distant organs may be mediated by specific cell membrane receptors. For example, although tumor cells may be present in the portal blood or peripheral circulation at the time of removal of a colon cancer, hepatic metastases occur only in some instances. Metastases may be present in the lung, bone, and other distant sites, such as the brain, even in the absence of hepatic metastases. Tumor cells can migrate through the hepatic sinusoids without establishing a malignant deposit in the liver.

Not all cells in a primary tumor have identical metastatic potential. [202] From a primary tumor, it is possible to clone cells with an enhanced capacity for metastasis and a propensity to metastasize to specific organs. For example, certain experimental tumor cells form lung metastases in rodents, but others do not. When these metastatic colonies are removed from the lungs, and their numbers are expanded in culture and reinjected by tail vein, each successive cycle produces a larger number of pulmonary metastases. The propensity of the highly metastatic cell line to grow in the lung appears to result from a specific interaction between the injected cells and the pulmonary capillary bed. Lung lesions are seen even after left ventricular injection of the cells, in which the lung is not the first capillary bed, or after transplantation of lung tissue to a subcutaneous location. By means of the same technique, other cell lines can be developed with an enhanced propensity to metastasize to the liver or to specific sites within the brain. [202], [203] and [204] Site-specific metastasis appears to be mediated by cell surface membrane glycoproteins. Removal of the glycoproteins from the tumor cell surface membranes interferes with the specificity of metastasis. By fusing membrane vesicles from metastatic cells to

those of nonmetastatic cells, it is possible to confer homing specificity to the recipient cells.

Primary colon cancer differs from metastatic colon cancer in that metastatic colon cells express sialylated mucin-associated carbohydrate structures that may play a role in adhesion to the target endothelial glycoproteins and the target cell basement membrane. [205] Other carbohydrate structures found on colon cancer cells appear to bind to molecules such as laminin, a normal component of the basement membrane.

Metastasis Genes

The genetic basis of metastasis is complex and incompletely understood. One gene has been identified that is associated with distant metastasis in colorectal carcinoma. The *NM23* gene (also designated *NME*) seems to function as a metastasis suppressor gene, inasmuch as it was deleted in distant metastases from colorectal cancer. [206] Patients without allelic deletions of this gene have a significantly better prognostic outlook. The cellular mechanism by which the *NM23* locus confers metastatic capability is uncertain, but transfection of the gene into certain tumor cell lines inhibits their ability to migrate in response to chemoattractants. [207] *NM23* gene mutation has been reported from human colorectal cancer metastases, suggesting that it may undergo a two-step inactivation, like other tumor suppressor genes. [208]

Other potential metastasis suppressor genes may be located on the long arm of chromosome 18. Chromosome 18q is lost in 80% of patients with stage IV colorectal cancer. [209] The deleted in colorectal cancer (DCC) gene, located on chromosome 18q21, was one such metastasis suppressor gene. *DCC* was identified after a study of commonly deleted regions on chromosome 18q in colorectal cancers. [210] *DCC* encodes a unique protein with homology to neural cell adhesion molecules and other related cell surface glycoproteins. This similarity has led to the hypothesis that the DCC gene product is involved in cell-to-cell adhesion and cell matrix interactions, which may be important in preventing tumor growth, invasion, and metastases. Expression of DCC was reportedly absent in most colorectal cancers metastatic to the liver, but it was lost in only a minority of nonmetastatic cancers. [211] However, germ-line inactivation of the murine homolog of *DCC* failed to support a tumor-suppressive function for its protein. [212]

Additional genes on chromosome 18q, such as the TGFB1 signaling molecules *SMAD2* and *SMAD4,* and other genes elsewhere may possess metastasis suppressor function, but further genetic and biochemical studies are required to resolve this issue. [213] Numerous genes participate in the metastatic phenotype, and their accumulation within malignant cells is responsible for the broad range of virulent behaviors seen in cancer.

CLINICAL MARKERS OF NEOPLASIA

Biologic Tumor Markers and Oncofetal Proteins

Some gastrointestinal cancers acquire and elaborate cell surface determinants that recapitulate structures transiently expressed during normal fetal development. These oncofetal proteins are typically glycoproteins and can often be detected in the plasma. Carcinoembryonic antigen (CEA) was first detected with antibodies developed after rabbits were immunized with colorectal cancer cells; these antibodies are largely directed to the carbohydrate moieties of the glycoprotein. [214], [215] Subsequently, elevated levels of CEA were demonstrated in patients with colorectal cancer and other cancers. However, the limitations of CEA measurements in the population as a whole have precluded the use of CEA as a screening or diagnostic marker. Poor sensitivity and specificity in asymptomatic populations render it less than useful for detecting early, potentially curable cancers. [216] Other conditions besides colorectal cancer are associated with elevated plasma levels of CEA, including cancers of the stomach, pancreas, and liver, nonmalignant diseases of the liver such as alcoholic hepatitis, inflammatory bowel disease, and tobacco use. [217] CEA levels measured at the time of diagnosis may be correlated to the prognosis of colorectal and other cancers (i.e., the higher the CEA elevation, the more advanced is the cancer stage at diagnosis). [218], [219] Nonetheless, this information cannot be used to predict outcome.

Other oncofetal proteins have been used to screen for the presence of gastrointestinal cancer. As with CEA, poor sensitivity and specificity and elevated levels in nonmalignant conditions have limited their usefulness in screening. The determinants for most markers reflect modifications of blood group antigens (e.g., Lewis blood group) and mucin glycoproteins. Table 24-5 shows some tumor markers and their associated cancers.

TABLE 24-5 Examples of Oncofetal Antigen Markers in the Gastrointestinal Tract

Genetic Tumor Markers

Cancer cells develop as a consequence of multiple genetic insults that affect normal growth regulatory mechanisms. Detection of mutations in tissue samples would therefore suggest the presence of cancer or precursor neoplasia. Techniques in molecular biology, largely through use of the polymerase chain reaction, allow the detection of minute amounts of tumor cells or free nucleic acid in biologic samples. For instance, *KRAS* mutations have been detected in DNA purified from feces, but benign and malignant colorectal tumors could not be differentiated. [220] In addition, *KRAS* mutations have been detected in the stool of patients with pancreatic adenocarcinoma as well as chronic pancreatitis. [221] The detection of *KRAS* mutations in feces as a means of differentiating neoplasia from inflammation is difficult, and differentiating between colonic and pancreatic neoplasia likewise is problematic. *KRAS* mutations also have been detected in the plasma of patients with colorectal cancer. [222] The detection of *KRAS* mutations in pancreatic juice may eliminate the colon as a source of the mutation, but still cannot distinguish benign from malignant disease. [223]

Mutation in *p53* has been associated with poor prognosis in several malignancies, including colon cancer, [224] and loss of normal *p53* signals the transition from benign to malignant colorectal neoplasia. [143] Tumors with mutations in *p53* tend to respond poorly to therapy, whereas malignancies that rarely have *p53* mutations (e.g., Wilms tumor, testicular cancer, acute lymphoblastic leukemia) often respond to therapy. [225] The finding of *p53* mutations in the urine of patients with bladder cancer and in the stool of patients with colorectal cancer suggests that detection may eventually be useful for screening. [226], [227] The presence of mutated *p53* suggests the presence of a neoplastic process; however, it may not indicate loss of the second, normal *p53* allele that is seen in malignant transformation. Thus, distinguishing a benign adenoma from colorectal cancer, as with *KRAS,* may be impossible. The costs and utility of advanced molecular diagnostics in colon cancer in comparison with those of conventional screening methods have not been addressed. Current oncology clinical practice guidelines do not recommend the routine use of tests for the *p53* tumor suppressor gene or the *KRAS* oncogene in screening for colorectal cancer. [228]

Detection of naked DNA by microsatellite analysis of the serum [229] and plasma [230] of patients with head and neck cancer and of patients with small cell lung cancer, respectively, indicates that these tumors release large amounts of DNA into the blood. For the most part, microsatellite alterations in the circulating tumor DNA were similar to those in the primary tumor. Enrichment of the tumor DNA in the blood may occur. Aberrantly methylated *hMLH1* promoter DNA has also been detected in the serum of patients with microsatellite-unstable tumors. [231] LOH can be detected in the serum, which requires higher concentrations of DNA than that needed for the detection of microsatellite instability. [232] However, plasma or serum microsatellite alterations in benign or nonneoplastic tumors have not been identified.

The status of chromosome 18q in a tumor may have prognostic significance for patients with colorectal cancer. Rates of survival associated with stage II (Dukes B2) colorectal cancer and LOH at chromosome 18q were similar to those associated with stage III cancer. [233] The survival rates of patients with stage II cancer and no LOH at chromosome 18q were similar to those of patients with stage I cancer. The status of chromosome 18q was determined from the resected tumor and suggested that patients with stage II disease and LOH at chromosome 18q should be offered chemotherapy because patients with stage III disease have been shown to derive survival benefit from chemotherapy. [234] It is not known what genes on chromosome 18q may be metastasis suppressor genes that, if lost from the tumor, may allow

spread to the regional lymph nodes (stage III) or distant organs. [213]

The test with the best specificity may be the detection of *APC* mutations in fecal DNA. [237] Mutations in *APC* generally initiate colorectal neoplasia, and detection of such mutations would be expected to segregate with neoplasia. In a feasibility study, *APC* mutations were detected in 57% of patients with polyps or cancer, but in none of the control patients without colorectal neoplasia. [235] A digital protein truncation test, in which multiple samplings of fecal DNA were amplified by the polymerase chain reaction and later subjected to an in vitro transcription translation assay, was used in the study. The test is based on the identification of abnormal proteins synthesized from mutant genes; future improvements in proteomics may increase the sensitivity of this assay. This approach will not detect defects in other proteins of the Wnt signaling pathway.

Assaying multiple genetic targets as part of one complete assay is another approach to detect neoplasia. There is some genetic heterogeneity in colorectal tumorigenesis that may extend to other cancers of the gastrointestinal tract. One group assayed mutations of *KRAS, p53,* and *APC* genes, *BAT-26* (a microsatellite instability marker), and highly amplifiable long-strand DNA from human fecal material. [236] *Long-strand DNA* refers to DNA that has not been degraded during apoptosis; it may be present in cells that have escaped programmed cell death. With use of the panel of multicomponent markers, the sensitivity for detecting neoplasia was 82% for adenomas and 91% for cancers. [236] When *KRAS* was excluded from the analysis, the specificity for detecting neoplasia increased from 93% to 100%, with a positive predictive value for colorectal neoplasia of 100% and a negative predictive value of 85%. [236]

The use of genetic tumor markers for screening and determining survival is currently limited. As our understanding of tumor biology progresses and the sophistication of biotechnology improves, genetic tumor markers will likely play a role in the management of cancer patients.

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CHAPTER 25

Alex Novogrudsky and Andrew G. Plaut

THE BACTERIAL FLORA OF THE HEALTHY GASTROINTESTINAL TRACT

Interest in gastrointestinal bacteria can be traced back hundreds of years, but serious inquiry into the composition of the gut microflora and its impact on health began late in the 19th century. [1] Soon after bacteria were identified as agents of disease, bacterial masses living in the human intestine were looked on with suspicion. Why these bacteria were bad was hard to define for early clinician-scientists, but it was clear that the microflora was well worth changing, or abolishing altogether. The methods used included medications, physical devices such as high-volume enemas, and fermented foods such as yogurt. To this day, attempts to change the microflora persist, and well-controlled clinical studies on selected strains of *Lactobacillus* and *Saccharomyces* that are able to colonize the human intestine show some therapeutic benefit. Beyond the use of such probiotics, however, modifications of the enteric microflora by strategies such as enemas are useless and certainly risky reminders of the history of this discipline. Characterization of the gut microflora has encouraged the development of selective decontamination methods for the preparation of persons undergoing alimentary tract surgery and for patients receiving intensive care, but a recent review of ten studies indicates that this strategy is of limited utility. [2]

Serious study of the gut microflora first required reliable methods of culture, particularly for the study of the anaerobic bacteria that dominate the normal commensal gut microflora (*commensal:* “together at the table”). Anaerobes, which do not grow or are killed in the presence of oxygen, were first cultured in the laboratory of Louis Pasteur in 1877, and despite continued refinement of methods since that time, difficulties in handling these oxygen-sensitive species persist. Laboratory culture made it possible to carry out taxonomy of the dominant gut bacteria, an effort now on firmer footing with the use of methods of molecular biology to characterize chromosomal and plasmid DNA. These methods not only refine identification but also can establish the relationships among bacterial species and allow tracking of microorganisms as they are transmitted from one person to another. [3] The use of methods such as 16S ribosomal RNA-based molecular phylogeny as well as nucleic acid amplification technology can even identify microorganisms that until recently defied all attempts at in vitro culture—a good example being the identification of *Tropheryma whippelii* as the cause of Whipple disease. [4] The highly sensitive polymerase chain reaction and the availability of primers specific for highly conserved regions of bacterial 16S ribosomal DNA enable the detection of DNA derived from gut bacteria even in blood specimens from healthy individuals. [5] Such culture-independent methods may in time reveal that some illnesses of still uncertain etiology are caused by infectious microorganisms, some of which may originate in the bowel flora.

Beyond the significant advances already made in culture and taxonomy, the most recent chapter in the study of gut bacteria brings the surprising experimental finding that bacteria colonizing the small intestinal mucosa can signal the host mucosa epithelium to undergo surface modifications. In rodent models of bacterial colonization, bacteria not only have a high specificity for the cell or tissue they colonize but also may actually induce changes in host cell surfaces that improve the microbial nutritional microenvironment. These changes brought about by commensal bacteria may strengthen the resistance of the mucosa to infection, providing a rational basis for the use of probiotics as health supplements and therapy. Studies in experimental animals indicate that colonization is of mutual benefit for host and microbe; the host provides nutrients, a controlled temperature, and a favorable physical environment for bacterial replication, while the bacteria provide specific nutrients needed by the host and possibly protect against attachment and mucosal injury by pathogenic bacteria. As discussed later in this chapter, the very existence of a colonizing microflora is now regarded as essential for the development of key components of the mucosal immune system.

COLONIZATION OF THE HUMAN GUT

True colonization requires that entering bacteria bind to tissues, that they proliferate at the site of binding, that their progeny efficiently reattach, and that they withstand mechanical and other defensive mechanisms of the host. An alternative to direct binding to host cells is for incoming bacteria to bind to microbes already colonized, a process referred to as *coaggregation*. Few microorganisms meet the stringent criteria of colonization, including many strains currently used as probiotic agents. Anaerobes also depend on redox potentials in the gut that are low enough for their growth, conditions met in the colon through oxygen utilization by aerobic species. Successful colonization is independent of the number of microbes ingested, which is why early attempts to force colonization by large inocula were ineffective.

Microorganisms that colonize the normal gut are abundant, taxonomically complex, and markedly uneven in distribution, but the entire commensal microflora appears to consist solely of bacteria and a few yeasts. When collected by careful techniques, fluids or mucosal washes from the esophagus, stomach, and jejunum contain no more than 10^6 [6] bacteria per milliliter, and these relatively few microbes are likely to come from food or from the oral cavity, which itself has a rich bacterial population. Bacteria can proliferate in the upper gut if the gastric pH rises, if gut motility is abnormal, or if sluggishly emptying diverticula or fistulae offer a site for microbial persistence, the basis for bacterial overgrowth syndromes. Otherwise, the upper gut has no persuasive tissue-binding sites for commensal microbes. The same is not true for pathogens; *Helicobacter pylori* can attach and survive in the stomach, and *Vibrio cholerae* securely binds to G_{M1} ganglioside on jejunal epithelial cells, where it proliferates and produces cholera toxin. Why nonpathogens fail to find a niche in the upper reaches of the digestive tract is something of a puzzle, although the nutritional needs of a mass of microorganisms in the upper gut may leave the host at a competitive disadvantage.

The Gut of Nursing and Weaned Children

The gastrointestinal tract of a healthy newborn is not colonized, but a complex microflora is established within weeks after birth. Fecal enterobacteria, streptococci, and staphylococci appear within days, and by oxidative metabolism they reduce the redox potential to encourage rapid colonization by strict anaerobes. In breast-fed babies, about 90% of the fecal bacteria are *Bifidobacterium* species, the expansion of which is further encouraged by specific growth factors in human milk. Enterobacteriaceae and enterococci are present in low numbers, but *Bacteroides* species, staphylococci, and lactobacilli are absent. Within weeks of weaning an infant to food or cows’ milk–based formula, *Bacteroides* species, enterococci, lactobacilli, and clostridia appear, a change likely more a consequence of a profound reduction in the numbers of *Bifidobacteria* species (no longer promoted by milk growth factors) than of changes in the mucosa or growth conditions. This new

microflora dominates throughout adult life, and major changes do not occur without the use of antibiotic drugs.

The Gut of Healthy Adults

The lumenal contents of the distal ileum and colon and the feces of adults contain about 10^{11} to 10^{12} colony-forming units per gram, a value approaching the theoretical limits for packing of bacteria into a cubic centimeter. As indicated earlier, coliforms, enterococci, clostridia, and lactobacilli are all found in a normal fecal sample, but the most abundant species by far are the strictly anaerobic *Bacteroides* organisms and anaerobic lactic acid bacteria (Table 25-1). Although aerobic or facultative coliforms are easily cultured (hence the use of coliform counting to detect contaminated water), anaerobes, which are far more difficult to culture and recover, outnumber aerobes 1000- to 10,000-fold in the human colon.

BACTERIA	COMMENTS
GRAM-POSITIVE COCCI	
Peptostreptococcus	Formerly *Peptococcus*; anaerobic counterpart of the streptococci
Streptococcus	Colonic species are mainly serogroup D
Enterococcus	Formerly with serogroup D; species include *E faecalis* and *E faecium*
Gemella	Closely related to oral streptococci
GRAM-POSITIVE RODS	
Clostridium	Forms spores; more than 80 species
Actinomyces	No spores, filamentous; strongly prefer anaerobic growth
Bifidobacterium	No spores; dominate colonic microflora of nursing babies
Lactobacillus	No spores; native forms include *L acidophilus*, *L fermentum*, *L plantarum*
Eubacterium	
GRAM-NEGATIVE RODS	
Bacteroides	*B fragilis* strongly dominates the adult colonic microflora
Campylobacter	
Porphyromonas	Includes many organisms formerly classified with the *Bacteroides* (e.g., *P gingivalis*)
Prevotella	Includes many organisms formerly included with the *Bacteroids* (e.g., *P melaninogenica*)
Shigella	
Enterobacteriaceae	Includes *Escherichia coli*, *Citrobacter*, *Enterobacter*, *Shigella*, *Salmonella*, *Serratia*, *Proteus*, *Providencia*, *Klebsiella*, and *Yersinia* species
GRAM-NEGATIVE COCCI	
Veillonella	Closely related to aerobic Neisseriaceae

Note: Strict anaerobes are in **bold** type; others are microaerophilic or facultative.

TABLE 25-1 A Guide to the Facultative and Strictly Anaerobic Bacteria in the Human Colon

The nutritional requirements of *Bacteroides* species are easily met from dietary and host sources. *Bacteroides* organisms are saccharolytic, producing enzymes that cleave glycosidic linkages in the abundant complex polysaccharides such as dietary xylans, glucans, and pectins that reach the colon.[6,7] The same enzymes also attack *O*-linked oligosaccharides on mucin proteins produced by the gut epithelium; therefore, *Bacteroides* species cleave both endogenous and exogenous glycopolymer substrates, reducing all to mono- or disaccharides for efficient utilization as nutrients. The complex classification of *Bacteroides* organisms is reflected in recent changes in both genus and species designation (see Table 25-1). The predominant colonic *Bacteroides* species is *B fragilis;* the corresponding oral species have been reassigned to the genus *Prevotella* or *Porphyromonas*.[8] Species names can obscure close relationships; for example, *Bifidobacterium bifidum* in the infant colon is gram-positive, but a close relative of the gram-negative *Bacteroides* species that dominates the colonic flora in adult life. *Clostridium difficile* is a strict anaerobe that often colonizes infants, who are seemingly protected from its enterotoxin. A remarkable clinical observation is that most persons never have a serious anaerobic infection throughout life despite the great abundance of anaerobes in the oral cavity and colon. This cannot be attributed solely to inefficient transmissibility of these species from person to person, as shown by the highly efficient transmission of anaerobes such as *C difficile* in the hospital setting.

Fecal lactobacilli are facultative or strict anaerobes and produce very large amounts of lactic acid (and somewhat less acetic acid) from carbohydrate substrates. The feces contain 10^{4} to 10^{8} colony-forming units of lactobacilli per gram, levels somewhat below the 10^{7} to 10^{8} colony-forming units of Enterobacteriaceae, such as *Escherichia coli,* per gram. The introduction of lactobacilli as probiotics[9] to treat antibiotic-associated diarrhea and to maintain general health is an old idea that has been vigorously revived through the development of strains that colonize more efficiently.

Quorum Sensing: How Bacteria Count Their Numbers

Quorum sensing is a recently identified and well-characterized mechanism through which bacteria communicate with one another. Originally defined in *Vibrio* species living in marine environments, quorum sensing involves the microbial control of gene expression in response to cell density and is found among both gram-positive and gram-negative bacteria.[10] The sensing processes of gram-negative bacteria are based on the production of extrasignaling molecules. Termed *autoinducers,* these molecules are acylated homoserine lactones that communicate cell density via receptor-based signaling to which the same or similar species are capable of responding. It has been shown that the biosynthetic pathway of one of the autoinducers (termed *AI-2*) is identical in the gut bacteria *E coli* and *Enterococcus faecalis* and in the pathogens *Salmonella typhimurium* and *V cholerae,* suggesting that it may be a general signal for bacterial communication. Studies of enterohemorrhagic *E coli* (EHEC) O157:H7 show that several of its key virulence mechanisms and basic physiological functions involve genes regulated by quorum sensing through AI-2, a product of the *luxS* gene.[11] Interspecies communication based on quorum sensing has not been proved to occur in the human gut, but this mechanism could allow a given bacterium to know its cell density and relative numbers in a complex microbial environment, and to use this information to express, or suppress, genes controlling many metabolic processes.

THE MICROBIAL FLORA AND THE HOST EPITHELIUM

The Bacterial Microflora and the Mucosa

The precise physical relationships of commensal microbes to the mucosa of the healthy human colon are not known in detail, although the proximity of certain species such as *Bacteroides* to their preferred substrates has been studied in depth.[8] The study of similarly colonized sites, such as the oral cavity, which is more easily accessed for such research, shows it to be a complex interplay of bacterial species. For example, streptococci colonize the newly cleaned enamel surface of the tooth covered by the proteinaceous acquired pellicle within hours to days, but after several weeks, they are replaced by anaerobes that dominate the microbial flora. This sequential and predictable arrival of colonizing bacteria involves mutual dependency; electron microscopy of developing bacterial masses on the tooth surface shows that in-coming new species often bind not to host tissues but to a dissimilar but already adherent microbe. The study of similar microbial relationships in the ileum and colon is more challenging, not only because this part of the gastrointestinal tract is inaccessible but also because the underlying colonized epithelium undergoes continual renewal.

Microbial Attachment to Host Cells

Microbial adherence is a critical determinant of colonization and is host-, tissue-, and cell-specific. By definition, adherence easily overcomes mechanical factors such as fluid flow and motility, which is made vividly clear by the tight attachment of pathogens such as *V cholerae* to the epithelium of the self-cleaning upper gut. This raises the question of why the large surface area of the human small intestine is so free of colonizing bacteria, and suggests that innate defense mechanisms such as cationic defensin peptides[12] and adaptive mucosal immune mechanisms are truly protective. Evidence that bacteria cannot find enough nutrients in the small bowel to permit colonization is not persuasive, but unsuitable pH and oxidation-reduction potential at this site definitely limit the growth of anaerobic species.

Most detailed information on bacterial attachment derives from studies of enteric and urinary tract pathogens whose attachment to host epithelial cells involves either type I or type IV pili. Pili are assembled from pilin subunits into a rigid fibrillar rod that extends far enough from the bacterial surface to overcome what otherwise would be mutually repellent negative charges on the microbial and epithelial cell membranes. Pilus assembly involves multiple microbial proteins and transport systems, similar to those involved in bacterial entry into cells during infection.[13] Originally considered a largely passive process, bacterial adherence induces metabolic events in both host and bacterial cells. For example, both can sense when binding has occurred, as shown with *E coli* (of colonic origin) that expresses type I pili encoded by the gene fimA. Once bound to bladder epithelial cells, bacterial pilus assembly is quickly interrupted, a process dependent on the expression of a bacterial protease (DegP) that degrades unassembled pilus subunits in the periplasmic space.[14] Mouse models of bladder infection by enteric coliforms also show that host cells sense bacterial pilus attachment; adherence induces a signaling cascade in the epithelial cell that brings about DNA fragmentation, caspase production, cell apoptosis, and,

finally, shedding of the cell from the epithelium. This orchestrated cell death in response to pilus-based signaling is likely to be host protective, limiting proliferation of pathogenic microorganisms on mucosal surfaces. Whether binding of commensal bacteria to gut epithelial cells regulates colonization is not yet clear.

Changes Induced by Colonizing Microorganisms

Experimental models have been developed that explore changes in the host epithelial cells induced by colonizing bacteria. [15] This research began with the observation that ileal epithelial cells of conventionally reared (i.e., colonized) mice continue to produce surface fucosylated glycans after weaning, but the mucosa of germ-free animals does not. Also, fucosylated glycans reappear in germ-free animals if a normal mouse flora is introduced. To refine these observations, *Bacteroides thetaiotamicron,* an anaerobe that typically colonizes the mouse (and human) ileum and colon, was introduced into mice as the sole colonizing species, and when present at sufficient numbers, it was able to signal the host cell continually to produce fucosylated glycans. [16], [17] *B thetaiotamicron* accomplishes this through the combined function of its L-fucose operon and a transcriptional repressor (FucR) whose activity is determined according to the levels of fucose in the environment. If the fucose level is low, FucR is unbound, and transcription of a protein that stimulates the epithelial cell to synthesize flucosylated glycans proceeds. These glycans can then be used by the bacterium as a source of energy and may promote the entry of other commensals that can metabolize fucose.

Follow-up experiments have shown that such induced changes are not limited to glycan production. [16] In gnotobiotic animals, careful manipulation of colonizing species is possible. Laser-capture microdissection and real-time quantitative reverse transcriptase–polymerase chain reaction (qRT-PCR) have been used to probe the changes in epithelial cells after colonization with *B thetaiotamicron*. Colonization increased the ileal levels of Na^+/glucose cotransporter mRNA and induced changes in the proteins involved in lipid absorption, such as pancreatic lipase–related protein-2, colipase, fatty acid–binding proteins, and apo A-IV. Changes in proteins involved in gut defense also occurred under the influence of *B thetaiotamicron,* including a 2.6-fold increase in the polymeric immunoglobulin receptor, increases in decay accelerating factor (an apical epithelial inhibitor of complement-mediated cytolysis), and increases in other proteins postulated to be important in mucosal integrity. Somewhat more relevant to the human gut, *E coli* K12 and *Bifidobacterium* species also induced changes in the expression of colipase in the mouse model. These experimental results are remarkable because they show that commensal bacteria can directly modify epithelial cell function, opening a new chapter in understanding the biologic properties of the gastrointestinal microflora. Such host cell changes may have important implications in disease and may help explain the value of the normal flora in defense against enteric pathogens.

METABOLIC ACTIVITY OF COLONIZING BACTERIA

General Biochemical Properties

The biologic activities of the colonic microflora are as diverse as their number, and commensal bacteria are no less active in modifying ingested proteins, carbohydrates, lipids, and medications than are host enzymes themselves. Clinically relevant biochemical modifications are those of bile acids, mucins, drugs, and steroids, and the biochemical mechanisms involved are largely hydrolysis, dehydroxylation, and chemical reduction. [18] Hydrolysis involves attack on glycosidic bonds in diverse compounds such as conjugated bile acids, estriol-3-glucuronide, bilirubin glucuronide, and digoxin, to name only a few. Potential carcinogens such as diethylstilbestrol or 1-nitropyrene are detoxified in the liver by the addition of a glucuronide residue, but subsequent deconjugation by bacterial glucuronidase to the free form can restore carcinogenic activity, as occurs with carcinogenic substances that already occur as glycosides in nature. One well-characterized and clinically important deconjugation involves the bacterial removal of glycine and taurine residues from bile acids. These amino acids are ionized at physiological pH, increasing the amphipathic properties of the bile acids, which are essential for their role in the assimilation of dietary fat and fat-soluble vitamins in the small intestine. The principal deconjugating microbial enzyme is ß-glucuronidase, highly active in bacteria such as *E coli* and *Clostridium* species, although levels are relatively low in lactobacilli and bifidobacteria. [19] Colonic bacteria also remove and modify hydroxyl and keto groups on sterols, including those comprising steroidal drugs. [20]

Interaction among Microbial Species

Competition for essential nutrients, production of substances that inhibit bacterial growth, suppression of bacterial adherence, and quorum sensing are all examples of microbial interactions in the gut, some of which contribute to the ability of the normal flora to prevent enteropathogenic infections. Early investigators noted that coliforms in continuous or static flow cultures in a reducing environment inhibited the growth of *Shigella flexneri,* a finding attributable to successful competition by *E coli* for growth-limiting carbon sources, mainly glucose. [21] /SUP>The extremely high efficiency with which essential nutrients are taken up by all bacteria must be considered when low levels of essential nutrients are invoked as a control on colonization. [22] Toxic metabolic end products of normally colonizing bacteria, such as alcohols, short-chain fatty acids, H_2S, and H^+, probably also inhibit pathogens, [23], [24] as does lactic acid produced by resident *Lactobacillus* and *Bifidobacterium* species in the colon. A low pH inhibits the growth of many anaerobic and facultative pathogenic species, which possibly explains the efficacy of probiotic lactobacilli as a treatment for persistent colitis caused by *C difficile*. Conversely, short-chain fatty acids produced by anaerobic species in the colon are known to inhibit the growth of *S flexneri* and *S typhimurium*. [25] Specific antimicrobial substances known as *bacteriocins* are proteins produced by streptococci and gram-negative enteric bacteria, but a persuasive case for their role in defending against proliferation of pathogenic microorganisms has not been made. [26] Another proposed role for the colonizing microflora in preventing the entry of pathogens is adherence inhibition, which in principle suggests that occupation of the cell surface by a colonizing commensal microflora physically prevents access of pathogens to the mucosa. How this might occur is unclear [27] and again illustrates that relatively little is known about the physical interaction of the microorganisms in the colonic environment.

HEALTH BENEFITS OF THE NORMAL MICROFLORA

Production of Short-Chain Fatty Acids

Short-chain fatty acids have a vital role in nutrition of colonic epithelial cells. [28] They are derived from the fermentation of dietary carbohydrate and nonstarch polysaccharides by bacteria in the cecum and ascending colon. Most short-chain fatty acids arise from water-soluble polysaccharides (e.g., pectin, psyllium) rather than insoluble fibers (e.g., cellulose and lignin). The most important short-chain fatty acids for colonic cell energy consumption are acetate C2, propionate C3, and butyrate C4, with butyrate being the most abundant and most efficiently utilized. Short-chain fatty acids of bacterial origin vary in lipid solubility depending on their state of ionization, which is an important influence on their transport into colonocytes. Once in the colonocyte, short-chain fatty acids provide epithelial cell nutrition, regulate cellular proliferation, and play a role in regulating regional blood flow of the colonic mucosa. Propionate has been reported to inhibit cholesterol synthesis and may be responsible for the effects of dietary fiber on plasma cholesterol levels. In the small intestine, short-chain fatty acids have also been noted to increase the growth and differentiation of enterocytes. Given the many properties of short-chain fatty acids, their use in therapy for diseases such as diversion colitis, ulcerative colitis, radiation proctitis, and pouchitis has at times been strenuously advocated, but most controlled studies show modest therapeutic benefit at best.

?-Carboxylation of Glutamic Acid Residues

Vitamin K (the naphthoquinones) is found in leafy green vegetables as K_1 (phylloquinone), but the K_2 (menaquinone) form is synthesized by intestinal bacteria. Vitamin K_1 is required for the conversion of certain glutamic acid residues of coagulation factors and of prothrombin to the ?-carboxyglutamate form, essential modifications that promote calcium binding, which is required for the proper function of these proteins. Profound vitamin K deficiency increases the coagulation time, particularly in newborns before the intestinal microflora has been fully established. Less commonly, vitamin K deficiencies are seen in obstructive jaundice (vitamin K absorption is bile acid–dependent) and after the prolonged use of antibiotics that reduce the number of vitamin K–synthesizing bacteria in the bowel.

Mucosal Immunity

Mouse models of colonization and immunity show that colonizing bacteria promote the full development of the mucosal immune system. [29] T cell–deficient mice or mice that lack costimulatory factors for adaptive immunity have unexpectedly high immunoglobulin A (IgA) antibody levels in the gut, suggesting that a primitive T cell–independent mechanism is important in establishing the mucosal immune system. Experiments have shown that the B cells producing these IgA antibodies originate in the peritoneal cavity, [30] and the antibodies are directed against antigens of the commensal microflora. For example, pathogen-free mice colonized with 10^8 colony-forming units of the commensal *Enterobacter cloacae* have gut secretory IgA antibody specific for these bacteria, whereas completely germ-free animals do not. Furthermore, animals in which gut-associated lymphoid tissue (Peyer patches, dendritic cell network, or B-cell follicles) was rudimentary or absent were found to have nearly normal IgA-producing B cells in the lamina propria, whereas animals without lymphoblasts, which are incapable of establishing a B-cell lineage, did not. These results show that a primitive intestinal IgA-producing mechanism solely driven by antigens exists within the colonizing microflora, that its assembly is not dependent on organized lymphoid tissue, and that an IgA response to

bacterial proteins occurs only if the intestine is colonized, not germ-free. These results add to early observations showing that monocontamination of germ-free animals changes gut histology from that of the germ-free state (tall, thin villi; shallow crypts; few lamina propria lymphocytes; sparse Peyer patches) to that of the normal state, a condition that has been referred to as *controlled inflammation*.

Probiotics

The value of feeding capsules or solutions containing living yet safe bacteria to treat gastrointestinal illness is an old idea that now has regained a foothold in clinical medicine. [31] Interest has been renewed because of increasing concern about the overuse of antimicrobial drugs, the availability of safe microorganisms for use as probiotics, and the evidence that newer strains of these microorganisms successfully colonize the gastrointestinal tract, or at least persist for weeks after feeding stops. [32] The microorganisms in current use are *Lactobacillus acidophilus, Lactobacillus rhamnosus* (e.g., *Lactobacillus* GG), *Lactobacillus casei,* and various *Bifidobacterium* species. The criteria for use are safety, ability to survive and adhere to sites in the human gut, validated benefit, compatibility with food and food processing, and anticipated antagonism to recognized pathogens. Because of the unique colony morphology and metabolic properties of certain of these strains, such as *Lactobacillus* GG, they are readily identified in fecal culture, so that their ability to colonize is easily estimated. Aside from certain yeast probiotics that cause rare pediatric infections, these organisms are generally regarded as safe. Probiotic bacteria produce organic acids, peroxide, and antibacterial peptides in vitro, which helps explain their clinical benefits, but exactly how probiotics interrupt ongoing infectious and noninfectious diseases of the gut is uncertain. The hypothesis that "good bacteria drive out bad" has not been proved, and the induction of host epithelial cell change (discussed earlier) may be involved. Several controlled trials show that probiotics are useful in treating *C difficile* infection, gut infections related to human immunodeficiency virus, pouchitis, traveler's diarrhea, and inflammatory bowel disease.

INFLUENCE ON HORMONES AND DRUGS

The great variety of bacteria in the colonic microflora can modify many foreign compounds and medications that reach the colon. The most widely known beneficial action of bacteria on a drug involves sulfasalazine, which is used in the treatment of inflammatory bowel disease. This medication is formulated with an aminosalicylate moiety coupled to sulfapyridine by an azo bond, and cleavage of this bond by bacterial enzymes delivers the aminosalicylate directly to the inflamed colonic mucosa. Bacterial modifications of drugs can be troublesome, as an early and well-documented example demonstrates—the cardiac glycoside digoxin is metabolized to pharmacologically inactive metabolites such as dihydrodigoxin by the enteric bacterium *Eubacterium lentum*. [20] Another example is the dehydroxylation of 3,4-dihydroxyphenylalanine (DOPA) to *m*-hydroxyphenylacetic acid, which decreases the bioavailability of the drug. [33] Bacteria also modify many other drugs that enter the colon, but few such changes have had a demonstrable impact on drug bioavailability. Enzymes produced by the intestinal microflora can also aromatize, dehydroxylate, hydroxylate, deconjugate, reduce, or epimerize sterols in the gastrointestinal tract. Deconjugation modifies the gastrointestinal absorption (and subsequent renal excretion) of estrogens; the urinary levels of 12 estrogens in pregnant women were 33% to 38% lower in those treated with ampicillin, [34] presumably because a reduction in colonic bacteria resulted in less extensive deconjugation of these sterols. Patients depending on cyclic estrogen and progesterone oral contraceptives may experience higher failure rates and breakthrough bleeding when taking antibiotics. [35], [36]

The biochemical modifications of certain environmental compounds by gut bacteria can generate carcinogenic substances. Many procarcinogens in the environment and in products such as food preservatives, dyes, and dietary additives are subject to changes by bacterial ß-glycosidases, ß-galactosidases, nitroreductases, and azoreductases that generate mutagens and other tumor promoters. One of many examples is cycasin, a naturally occurring glycoside of methylazoxymethanol in the seeds and roots of cycad plants. Cycasin is decidedly carcinogenic and when fed to healthy rats can lead to the development of liver tumors, renal sarcomas, squamous cell carcinomas, and colonic adenocarcinoma; however, tumors do not develop in germ-free rats fed the same glycoside. [37] Because this aglycone methylazoxymethanol is tumorigenic irrespective of gut colonization, it is clear that the parent cycasin must be hydrolyzed (by ß-glycosidase) to the carcinogen. In addition to natural procarcinogens, other carcinogenic substances, such as *N*-hydroxyfluorenylacetamide [38] and 1-nitropyrene, [39] which are first detoxified in the liver by the addition of a glucuronide moiety, can be converted back to carcinogens when attacked by gut bacterial ß-glucuronidase. Textile dyes such as trypan blue, [40] methyl yellow, and methyl orange [41] and industrial pollutants such as *p*-nitrobenzoic acid [42] are not carcinogenic in their original form, but they are metabolized to mutagens by microbial nitro- and azoreductases.

THE NORMAL MICROFLORA IN CLINICAL ILLNESS

Bacterial Overgrowth Syndromes

The most compelling evidence of the vigorous metabolic activity of the bowel flora is the proliferation of colonic anaerobes in the upper gut as a result of motility disorders, autonomic neuropathy, diverticula of the jejunum, pancreatic insufficiency, partial bowel obstruction, fistulae, or achlorhydria. [43] The number of anaerobes in the upper gut of such patients exceeds 10^6/g (or mL) of lumenal content, and *Bacteroides* organisms and anaerobic lactobacilli and enterococci are the dominant species, as they are in the colon. These microorganisms are avid for dietary cobalamins (vitamin B_{12}), and their cleavage of trace-labeled bile acids, fatty acids, xylose, lactulose, and glucose forms the basis of breath tests for bacterial overgrowth. Anaerobes living in the upper gut have access to bile acids, and, as discussed earlier, deconjugation removes a major hydrophilic element from these polar compounds. In addition, bacterial 7-dehydroxylases remove hydroxyl groups from bile acid sterols to convert cholic to deoxycholic acid, and chenodeoxycholic to lithocholic acid. Modifications of bile acids impair their function because the loss of water-soluble components limits their capacity to form mixed micelles involved in absorption of dietary fat and fat-soluble vitamins. Although most of the metabolic consequences of stagnant loop syndromes can be treated by reducing the number of bacteria with antimicrobial drugs, recovery may be delayed because bacterial overgrowth induces mucosal injury that is only slowly repaired.

Hepatic Encephalopathy

The commensal gut microflora has a major role in perpetuating hepatic encephalopathy through the production of ammonia and unidentified compounds that are involved in brain dysfunction. About 40% of the urea produced by the liver is hydrolyzed by urease of the normal microbial flora in the digestive tract, [44] and this ammonia is the preferred source of nitrogen for most enteric microorganisms. Antibiotics were originally used to reduce colonic ammonia production in liver disease, but a more effective and certainly safer strategy is to use oral lactulose to acidify the colon contents, promoting conversion of ammonia (NH_3) to the ionized and less completely absorbed ammonium ion (NH_4^+). Acidification of the colon by lactulose is itself dependent on bacterial metabolism of lactulose to lactic acid.

Methylmalonic Acidemia

The normal bacterial microflora may participate in rare clinical problems associated with inborn errors of metabolism. An example is methylmalonic acidemia, an inherited metabolic defect that causes mental retardation, episodic vomiting, lethargy, and protein intolerance and results from mutations in the host enzyme propionyl-coenzyme A (CoA). [45] The enzyme deficiency prevents the conversion of methylmalonic acid to methylmalonyl-CoA, and the accumulated methylmalonic acid is shunted to the production of propionic acid, a toxic metabolite. Bacterial propionic acid production in the colon simply adds to the toxic load, which can be reduced by treatment with metronidazole.

INNATE AND IMMUNOLOGIC REACTIVITY

Tolerance—The Permissive Response

Although the intestinal immune system can launch a robust protective immune response to enteric pathogens, nonresponsiveness (tolerance) typifies the reaction of the gut to commensal microorganisms. [46] It is important to note that immune tolerance is not indifference, but rather an active immune response that does not produce mediators of protection. But how does the immune system distinguish between pathogenic and commensal microorganisms? Active research on this subject has relevance for several unexplained gastrointestinal diseases.

Mucosal defense involves a series of complex systems that include so-called innate immunity (mediated in part by a-defensins, phospholipases, lactoferrin, and lytic enzymes), [47] adaptive immunity (in the gut, largely IgA and to a lesser extent IgM antibodies), and cellular immunity. The innate immune system is primitive, coordinated through germ line–encoded receptors expressed on the surface of neutrophils, macrophages, and surface epithelial cells at the host-environment boundary. Phagocytes engulf and kill invading pathogens, present microbes in the context of major histocompatibility complex (MHC) class II molecules to T cells, and activate the synthesis of inflammatory cytokines. In contrast, the adaptive immune response is characterized by somatic gene rearrangements and diversification that ultimately lead to millions of antigen-recognizing receptors on the surface of B and T cells. Bacteria survive through their ability to evade these defensive mechanisms.

As is emphasized below, infection by pathogenic microorganisms results in the epithelial cell production of nuclear factor-?B (NF-?B), a proinflammatory cytokine that enters the cell nucleus to trigger transcription of the gene encoding interleukin-8 (IL-8) and other inflammatory cytokines. Nonpathogens do not elicit such a response; indeed, binding of commensal bacteria to epithelial cells can block the NF-?B response by increasing the cytoplasmic levels of I?B, a protein that binds cytoplasmic NF-?B, preventing its translocation to the nucleus. Cell binding of commensal microbes is thought to increase I?B levels by delaying addition of the polyubiquitination and phosphorylation markers that normally foster intracellular degradation of this protein. [48], [49] Despite recent insights into how commensal bacteria forestall inflammatory responses, many details are still lacking. It has been proposed that a specific subset of dendritic cells in the intestinal mucosa may phagocytose apoptotic enterocytes together with their bound commensal microflora, inducing a state of systemic tolerance to these microbes. [50] This tolerogenic potential of the gut points to a possible opportunity to treat or prevent autoimmune disease, a well-studied example being the prevention of experimental neurological diseases similar to multiple sclerosis by feeding myelin basic proteins. [51]

Toll Receptors

The innate immune system relies on the identification of distinct features unique to pathogenic bacteria known as *pathogen-associated molecular patterns (PAMPs),* and the receptors that recognize them are termed *pattern-recognition receptors (PRRs)*. PAMPs, which are produced by the pathogen and are essential for the survival of the organism (and are therefore conserved), signal the presence of a pathogen to the host cell. Examples of PAMPs are lipopolysaccharides, teichoic acids, and peptidoglycans in bacteria, and mannans in yeast. [52], [53]

On the mammalian side of this interaction are the Toll-like receptors (TLRs), proteins originally identified in *Drosophila* that trigger the production of defensive, antimicrobial peptides. Different TLRs are activated by different pathogens, allowing *Drosophila* to discriminate, for example, between fungal and bacterial pathogens. [54], [55] Production of the protective peptides in *Drosophila* involves transcription factors very similar to NF-?B that activate tumor necrosis factor-a and IL-12 in mammalian species. [56], [57] In mammals, a family of TLRs have an important role in the induction of the adaptive immune system. [58] TLR4, the first mammalian form to be identified, induces an NF-?B signaling pathway similar to its homolog in *Drosophila,* activating genes that trigger T-cell recruitment. The microbial ligands recognized by mammalian cells have not all been identified, but each activates a different TLR. [59], [60] However, unlike *Drosophila* TLRs, each of which can discriminate among microbial species, mammalian TLRs appear to recognize a range of PAMPs from numerous microorganisms.

Although the details of human TLR specificity are incomplete, binding of microbial products initiates signals in the host cell that activate intracellular receptors designated *NOD1* and *NOD2,* lipopolysaccharide-binding proteins that have important functions in innate immune defense. NOD2 has been implicated in inflammatory bowel disease, with the finding that mutations in the gene encoding human NOD2 (on chromosome 16) can be correlated with the presence of Crohn's disease. [61], [62] Because NOD2 activates NF-?B production in response to lipopolysaccharide, and mutations in the protein may result in under- or overproduction of inflammatory mediators, a role for NOD2 mutations in Crohn's disease is of major importance in helping to understand the sustained inflammation of this disorder. Active research in this area will help define patterns of responsiveness of the normal gut to its commensal microflora.

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CHAPTER 26

Paul B. Watkins

DRUG METABOLISM IN THE LIVER AND INTESTINES

GENERAL PRINCIPLES

The absorption of drugs into the body most often involves passive diffusion through lipid membranes, usually via the digestive tract but occasionally through the skin or lungs. For this reason, drugs that enter the body typically are fat-soluble (lipophilic). Once inside the body, lipophilic drugs tend to sequester in the body's fat stores and bind in blood to plasma proteins. Drugs that are extensively bound to proteins are generally not readily eliminated in urine. If lipophilic drugs are excreted in bile, they tend to be reabsorbed from the digestive tract and thus undergo enterohepatic cycling. For these reasons, most drugs that gain entry into the body would remain there for a very long time if it were not for the body's ability to convert (metabolize) drugs into more water-soluble, and hence more readily excreted, metabolites. The liver is the major organ involved in drug metabolism. For some xenobiotics, the small intestine also plays a major role.

DISCOVERY OF DRUG-METABOLIZING ENZYMES

A major breakthrough promoting the study of drug metabolism was the development of techniques to isolate the endoplasmic reticulum from whole pieces of liver tissue. During this process, endoplasmic reticulum breaks up into small spheres called *microsomes*. It was discovered that under certain experimental conditions, liver microsomes produce most of the metabolites generated from drugs (and other xenobiotics) by the liver in vivo. The reactions characterized generally involved insertion of an oxygen atom into the drug (usually a hydroxylation) or covalent binding (conjugation) of the drug to a polar ligand such as glucuronic acid, sulfate, or glutathione. Many drugs first had to undergo hydroxylation to produce a reactive site on the molecule suitable for conjugation reactions. This typical sequence of events led to the division of xenobiotic metabolism into two categories: phase 1 (often hydroxylation reactions) and phase 2 (conjugation).

It is now appreciated that most (but not all) of what was described as phase 1 microsomal metabolism is the result of the activity of a large family of enzymes termed *cytochromes P450*. This somewhat awkward name derived from the fact that when first isolated, these enzymes appeared to be chemically similar to mitochondrial cytochromes. The *P* is an abbreviation for *pigment,* reflecting the fact that these enzymes are red in color (because of the presence of heme in the enzyme) and largely account for the reddish brown color characteristic of microsomes. Finally, it was noted that these enzymes intensely absorb light with a 450-nm wavelength (under certain experimental conditions). The cytochromes P450 are now usually referred to simply as *P450s* or *CYPs* (pronounced *sips*).

P450s AND DRUG METABOLISM

The vast majority of drugs in clinical use today are metabolized by liver P450s. A recent realization is that there are relatively few P450s important for drug metabolism. [1] Each P450 represents the product of a unique gene. The P450s involved in drug metabolism fall predominantly into three gene families, now termed *CYP1, CYP2,* and *CYP3*. [2] Within each P450 family, subfamilies are designated by capital letters. Each subfamily generally contains multiple members, designated by arabic numerals usually reflecting the order in which they were discovered. In general, orthologous P450s (the equivalent form in different species) are given a different final arabic numeral. For example, the term *CYP3A4* applies only to an enzyme present in human liver, whereas the related enzymes in other species have different numeric designations (e.g., CYP3A2, CYP3A6).

It is widely believed that the P450s involved in drug metabolism evolved as a mechanism to protect the body from potentially harmful chemicals, *xenobiotics,* present in the environment or in the diet. [3] According to this theory, plants produce potentially toxic xenobiotics to become inedible to insects and higher life forms. In addition, spoilage of food often involves bacteria and fungi capable of producing potentially toxic xenobiotics. The introduction into the environment of each new xenobiotic in turn created selection pressure for insects and animals to develop specific P450s capable of metabolizing and rapidly eliminating that xenobiotic (if the food source could not be easily avoided).

A list of the major P450s involved in human drug metabolism appears in Table 26-1. The most abundant single P450 in human liver is CYP3A4, and it has been estimated that this specific P450 may be involved in the metabolism more than 50% of drugs used clinically. [4] Only a few of the many substrates for CYP3A4 are listed in Table 26-1.

TABLE 26-1 Characteristics of Major Human Liver P450s

Some of the principles of P450 metabolism are shown in Figure 26-1 and Figure 26-2. Drugs either passively diffuse, or are actively transported, into the liver during passage through sinusoidal blood (illustrated by drug A in Fig. 26-1). Once inside the liver, drugs diffuse to the particular P450s capable of metabolizing them. With

some drugs, a single P450 is involved in most of the metabolism. The P450 inserts an oxygen atom onto the drug, usually in the form of a hydroxyl group. In some cases, however, the immediate product of P450 catalysis is unstable, and rearrangement of the molecule (such as *N*-dealkyation) then occurs, resulting in different end products. After the P450-catalyzed reaction, the resultant metabolite typically undergoes phase 2 conjugation. The conjugated metabolite is frequently secreted back into the space of Disse, now more water-soluble and less protein-bound, and hence more readily eliminated in urine by the kidneys. Alternatively, the metabolite can be secreted into the bile canaliculus to be excreted into the small intestine in bile, now less likely to undergo enterohepatic cycling because of its reduced lipophilicity. In many and probably most instances examined to date, P450-mediated metabolism is rate-limiting in the elimination of the drug. [5]

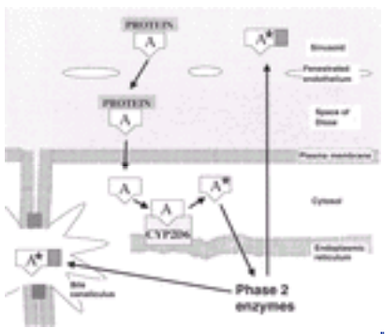

FIGURE 26-1. Metabolism of a drug in the hepatocyte. Lipophilic drugs, such as drug A, exist in sinusoidal blood largely bound to proteins. Drugs can diffuse through the endothelial cell fenestrations into the space of Disse while still protein bound. Drugs then enter the hepatocyte by active transport or passive diffusion. Once inside the cell, a drug typically encounters a specific P450 (CYP2D6 in this example) capable of binding to the drug and converting it to more water-soluble and chemically reactive metabolites. The resultant metabolite may be secreted from the hepatocyte without undergoing additional metabolism (not shown). In most instances, however, the P450-generated metabolite will undergo conjugation to a polar ligand in a reaction catalyzed by phase 2 enzymes. The resultant conjugated metabolite is then secreted into the biliary canaliculus for elimination in bile, or secreted back into sinusoidal blood to be excreted in urine.

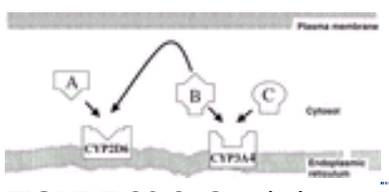

FIGURE 26-2. Catalytic specificity of P450s. Drugs are metabolized by a specific P450 only if they can fit appropriately into the substrate-binding site of the enzyme. In this example, drug A can be metabolized by CYP2D6, but not by CYP3A4. Drugs B and C can each be metabolized by CYP3A4, but only drug B can be metabolized by both CYP2D6 and CYP3A4. The principle of competitive inhibition between two substrates is illustrated by drugs B and C. If both are present in the hepatocyte, the metabolism of each may be reduced simply because both drugs cannot occupy the enzyme active site at the same time.

The concept of specificity of P450s is shown in Figure 26-2. In many instances, a single P450 represents the major pathway of metabolism of a drug. This is shown by drugs A and C, which can bind to, and be metabolized by, only CYP2D6 and CYP3A4, respectively. In many cases, however, a drug can be metabolized by more than one P450, and this is illustrated by drug B. Commercial vendors now supply recombinant expressed P450s that allow rapid identification of P450s capable of metabolizing a given drug. As will be discussed, knowing which P450(s) metabolizes a drug is useful information because it can often predict drug interactions and the extent of interpatient variability in disposition of the drug. For this reason, many pharmaceutical companies now use high-throughput technology to identify the P450s that metabolize compounds early in development and use this information to select lead candidates for further development. [6], [7]

The concept of specificity has allowed the use of some select drugs to quantify the activities of specific P450s in subjects in clinical trials. [8] Typically, the probe drug is administered intravenously or orally, and the concentration of the parent drug, or of a metabolite, is measured in urine, blood, breath, or saliva at a fixed time point. Some of the more commonly used probe-based tests are listed in Table 26-2.

P450	PROBE	ANALYTE
CYP1A2	Caffeine	Plasma, breath, urine, saliva
CYP2D6	Dextromethorphan	Urine, saliva
	Debrisoquine	Urine
CYP2C19	S-Mephenytoin	Urine
	Omeprazole	Urine
CYP2E1	Chlorzoxazone	Plasma
CYP3A4	Erythromycin	Breath
	Midazolam	Plasma

TABLE 26-2 Some Probe-Based Tests for P450s

The interindividual differences in the activity profile of the liver P450s are large, and in many instances, this variation appears to account for interpatient differences in the pharmacokinetics of medications. [4] In some cases, probe-based tests (see Table 26-2) have been shown to predict the clearance of specific drugs. For example, a caffeine-based test that measures CYP1A2 activity (caffeine breath test) has been shown to predict the clearance of tacrine. [9] In addition, an erythromycin-based test that measures CYP3A4 activity (the erythromycin breath test) has been shown to predict the clearance of cyclosporine [10] and docetaxel. [11] The variation among patients in the activities of specific liver P450s can reflect both genetic and nongenetic factors.

Genetic Factors Influencing P450 Activity

Genetic mutations in part explain why there are large interindividual differences in the activities of many of the P450s. [12], [13], [14] and [15] For example, both CYP2D6 and CYP2C19 are termed *polymorphic* enzymes because the population can be divided into distinct groups based on the relative activity of these enzymes. [16] About 5% of whites completely lack CYP2D6 activity and therefore are CYP2D6 “poor metabolizers.” Patients who are CYP2D6 poor metabolizers have been shown to have increased sensitivity to the effects of several drugs, including some medications commonly used to treat cardiac arrhythmias, psychosis, and depression. The poor metabolizer phenotype results from the simultaneous inheritance of two of multiple known functionally defective CYP2D6 alleles. [17] The incidence of CYP2D6 poor metabolizers varies substantially across different ethnic populations. [16] For example, fewer than 1% of Japanese are poor metabolizers.

The CYP2C19 poor metabolizer phenotype also occurs in approximately 5% of whites, but in up to 20% of Asians. [16] CYP2C19 poor metabolizers have been shown to have higher blood levels of omeprazole than others when treated with usual therapeutic doses. [18] This is expected because omeprazole relies on CYP2C19 for elimination (see Table 26-1). This appears to explain why CYP2C19 poor metabolizers have a higher cure rate for *Helicobacter pylori* infection when treated with omeprazole-containing regimens. [19] As is the case with CYP2D6, the poor metabolizer phenotype also results from the inheritance of two of several different functionally defective CYP2C19 alleles. In general, CYP2D6 and CYP2C19 poor metabolizer phenotypes behave as autosomal recessive traits, with heterozygotes having a metabolic capability that is intermediate between those of typical normal and poor metabolizers.

Allelic mutations resulting in diminished catalytic function have been described for other cytochromes P450, including CYP2C9 [20] and CYP2A6, [21] although individuals completely lacking these catalytically active enzymes have not yet been recognized. In addition, genetic mutations of unclear functional significance have been identified in CYP3A4 [22] and CYP2E1 [23] genes. Approximately 20% of whites and 50% of African Americans express a P450 closely related to CYP3A4, termed

CYP3A5. [24] It was originally believed that CYP3A5 was a minor liver P450 even in those who express the enzyme, but newer data suggest the liver content of CYP3A5 may be comparable to that of CYP3A4 in some individuals. [24] The functional significance of the CYP3A5 polymorphism is largely unexplored.

In one instance, genetic factors can cause an abnormally high level of activity of a P450. Approximately 2% of whites have gene duplication of CYP2D6, resulting in an “ultrarapid” metabolizer phenotype. [13] These individuals exhibit unusually rapid clearance of at least some CYP2D6 substrates, and this can account for therapeutic failure of some medications. The incidence of ultrarapid metabolizers in nonwhite populations is currently under investigation.

Nongenetic Factors Influencing P450 Activity

Inflammation and alterations in nutritional status can affect the activities of liver P450s. [25] In addition, many drugs can alter the activities of P450s, resulting in drug interactions. [26] Some drugs are known to inhibit the activity of specific P450s, reducing the elimination of drugs that require metabolism by that P450. Inhibition of P450 activity can occur by a variety of mechanisms, [27] but the most common form of inhibition reflects simple competition between two drugs for metabolism by the same P450. This is shown schematically in Figure 26-2, in which drugs B and C are each capable of binding to, and being metabolized by, CYP3A4. Hence, metabolism of drug B by CYP3A4 may be reduced in the presence of drug C, simply because drug C is physically interfering with the ability of drug B to bind to the active site on CYP3A4. Likewise, drug B could potentially inhibit the metabolism of drug C in an analogous fashion. Whether two drugs competing for metabolism by the same P450 will result in a clinically important interaction depends on a number of factors. These include the intrahepatocyte concentrations of each drug, the relative affinities of each drug for binding to the enzyme active site, and the importance of the P450 to the overall elimination of the drug. Additional important variables are the relative safety (therapeutic index) of the drugs involved and whether the metabolites generated by the P450 are pharmacologically active.

Some clinically important interactions involving inhibition can be inferred from Table 26-1. For example, patients treated with tricyclic antidepressants will have elevated tricyclic blood levels and possibly associated toxicity if certain selective serotonin reuptake inhibitors (SSRIs) are added to their treatment regimen (without lowering the dose of tricyclics). This results from the inhibition of CYP2D6 by SSRIs. [28] In addition, cyclosporine blood levels can rise to toxic levels in organ transplant recipients who receive concomitant treatment with erythromycin or ketoconazole, largely because of inhibition of CYP3A4. [29] Many interactions between drugs used in patients with human immunodeficiency virus (HIV) infection involve P450 inhibition. [30]

Some medications are such potent and selective inhibitors of specific P450s that they can be used in experimental settings to pharmacologically “knock out” the enzyme in a human subject. The drugs most frequently used to inhibit specific P450s are noted in Table 26-1. If elimination of a drug is not affected by the inhibitor, then it can be assumed that the target P450 does not represent a major pathway for elimination. In some cases, these inhibitors have very high affinity for the enzyme active site and largely prevent the enzyme from metabolizing other drugs on a competitive basis (illustrated in Fig. 26-2). For example, a single dose of quinidine or ketoconazole largely competitively inhibits the activities of CYP2D6 and CYP3A4, respectively. [31] Other selective inhibitors are not competitive inhibitors but rather “mechanism-based” inhibitors. [27] Mechanism-based inhibition is irreversible and occurs when the inhibitor is converted by the target P450 to a metabolite that noncompetitively inactivates the enzyme (often by covalently binding to the enzyme protein). This appears to be the mechanism by which a single therapeutic dose of disulfiram (Antabuse) results in greater than 90% inhibition of CYP2E1 activity that lasts for hours. [32] The effect of disulfiram is specific in that the catalytic activities of other major P450s are not affected (as measured by suitable probes). [32] Another example of a potent and selective mechanism-based inhibitor is the antibiotic troleandomycin, which, when administered in the usual therapeutic dose, produces greater than 90% reduction in liver CYP3A4 activity that also persists for hours. [33] Although it has been less well studied in vivo than disulfiram, data obtained in human liver microsomes suggest that the effect of troleandomycin is specific for CYP3A4 and that other major P450s are not inhibited. [34] Examples of other mechanism-based inhibitors include the CYP1A2 inhibitor furafylline [35] and the CYP2A6 inhibitor methoxsalen. [36]

In addition to inhibition, a medication can sometimes result in an increase in the activity of a particular P450. In most cases, this “induction” results from increased hepatocyte concentrations of a specific P450. Some medications that can induce P450s are listed in Table 26-1. Induction of P450 activity can occur by several mechanisms, but most commonly reflects an increase in the rate of transcription of the corresponding gene. In some instances, the cellular receptor involved in transcriptional activation has been identified. [37] For example, induction of CYP3A4 by rifampin and antiseizure medications appears to involve a nuclear receptor termed the *human pregnane X receptor (PXR),* [38] also termed *steroid xenobiotic receptor (SXR)* or *pregnane-activated receptor (PAR)*. The inducer binds the receptor, and after forming a heterodimer with another receptor (RXR), it binds to regulatory elements in the CYP3A4 gene. This results in increased transcription of the CYP3A4 gene, which in turn results in increases in the hepatocyte concentration of CYP3A4. [37] It appears that PXR also mediates induction of CYP2C9 (and probably CYP2B6) by rifampin and certain antiseizure drugs. [39] PXR has been proposed to be the major xenobiotic sensor present in humans, orchestrating a host response to rid the body of foreign substances. [40] PXR has also been shown to be activated by lithocholic acid, and activation of PXR results in up-regulation of genes involved in bile acid secretion. [41] It seems likely that phenobarbital and rifampin relieve pruritus in cholestasis by activating PXR. Polymorphisms in PXR have been identified, but their functional significance is not known. [42]

Induction of CYP1A2 also involves a receptor, termed the *aryl hydrocarbon (Ah) receptor*. [43] Aryl hydrocarbons in cigarette smoke and the drug omeprazole bind to the Ah receptor, mediating transcription of the CYP1A2 gene and increased production of the enzyme. Some P450s, such as CYP2D6 and CYP2A6, do not appear to be inducible.

Examples in which induction causes clinically significant drug interactions can also be deduced from Table 26-1. When transplant recipients are treated with rifampin or certain antiseizure medications, blood levels of cyclosporine can fall to subtherapeutic levels, largely because of induction of CYP3A4. [29] These patients are at risk for organ rejection unless their daily dose of cyclosporine is increased. Likewise, individuals treated with rifampin or antiseizure drugs are at risk for therapeutic failure of oral contraceptives (CYP3A4) and warfarin (CYP2C9). [26] Induction can also be used to assess the contribution of an inducible P450 to the disposition of a drug. For example, if treatment of subjects with rifampin does not increase clearance of a drug, it is unlikely that CYP3A4 or CYP2C9 are major pathways for metabolism.

Role of P450s in Drug-Induced Liver Injury

Most drugs that are capable of causing liver toxicity appear to do so through the generation of toxic metabolites. [44] As discussed above, P450s generally convert drugs to metabolites that are safely eliminated from the body. However, under certain circumstances, P450s can generate reactive and potentially toxic metabolites. Indeed, the identical enzymes (see Table 26-1) involved in the safe metabolism of drugs are those that have been most frequently implicated in the production of hepatotoxic metabolites. In general, P450s are expressed in highest concentration in zone 3 hepatocytes, and this in part accounts for the predominance of pericentral necrosis in some forms of drug-induced liver injury (such as that caused by acetaminophen). [45] Interspecies differences in P450 catalytic activities and regulation probably contribute to the imperfect ability of preclinical animal studies to identify human hepatotoxins. [46]

The most studied example of production of a hepatotoxic metabolite by P450s involves acetaminophen liver injury. Acetaminophen is believed to cause toxicity in the liver by production of the *N*-acetyl benzoquinone amine metabolite NAPQI. Studies with recombinant human liver enzymes suggested that NAPQI could be produced by several P450s, including CYP2E1, CYP3A4, CYP1A2, and CYP2A6. [47], [48] Investigators have addressed the relative importance of each of these enzymes in human studies by using inducers and inhibitors of specific P450s. NAPQI formed from acetaminophen in the liver is conjugated to glutathione and eliminated as various thiol metabolites in urine. Hence, the total production of NAPQI can be estimated from the production of thiol metabolites eliminated in urine. It has been shown that the urinary excretion of thiol metabolites is not increased in subjects pretreated with the CYP1A2 inducer omeprazole [49] or the CYP3A4 inducer rifampin, [50] so that a prominent role of these enzymes is unlikely (see Table 26-1). However, pretreatment of subjects with the CYP2E1-specific inhibitor disulfiram resulted in a 69% reduction in the production of NAPQI. [50] Hence, although in vitro studies indicate the potential involvement of multiple P450s, in vivo studies in humans indicate that a single P450, CYP2E1, accounts for most of the NAPQI formed after therapeutic doses of acetaminophen.

Some other observations have suggested a role for CYP3A4 in acetaminophen toxicity. There have been rare case reports of severe acetaminophen toxicity in patients treated with antiseizure medications, [51] which induce CYP3A4 but are not inducers of CYP2E1. In one series of patients with acute liver failure, [52] patients with acetaminophen liver injury who had been receiving treatment with antiseizure drugs appeared to have a worse outcome than other patients (although a more current report [53] from the same institution did not verify these findings). Studies in rodents have also suggested a role for CYP3A enzymes in the production of NAPQI. [54] However, marked differences between humans and rats in the catalytic activities of P450s involved in NAPQI production have been assessed in vitro. Moreover, in three separate studies, NAPQI production from acetaminophen did not increase in individuals treated with antiseizure medications known to induce CYP3A4. [55], [56] and [57] Of course, these human studies involved administration of relatively low (therapeutic) doses of acetaminophen, and it is therefore possible that CYP3A4

contributes to NAPQI production in an overdose situation.

Chronic consumption of ethanol appears to increase susceptibility to acetaminophen hepatotoxicity. [58] Ethanol is a recognized inducer of CYP2E1 (see Table 26-1), and ethanol induction of CYP2E1 therefore provides an attractive explanation for incremental risks in ethanol consumers. However, early animal [59] and human [60] studies did not show increases in NAPQI formation when acetaminophen was given during or immediately after the ingestion of ethanol. Indeed, these studies suggested that ethanol consumption actually reduces the rate of production of NAPQI, protecting the liver from toxicity. These observations have now been explained. [61] Ethanol is a substrate for CYP2E1 (see Table 26-1). When ethanol is present in the body in substantial concentrations, a large proportion of the CYP2E1 binding sites in the liver are occupied by ethanol. [62] Ethanol binds CYP2E1 with high affinity and is slowly metabolized by the enzyme. Hence, the rate of production of NAPQI from acetaminophen is reduced when ethanol is present. This is illustrated in Figure 26-3. An additional important finding is that when ethanol is bound to CYP2E1, the enzyme is stabilized against degradation, so that its intracellular half-life is increased. [63] Prolonged intoxication therefore results in an accumulation of (ethanol-inhibited) CYP2E1. With very high ethanol intake, induction of CYP2E1 may involve increased enzyme synthesis as well as stabilization. [64] When ingestion of ethanol is stopped and ethanol is cleared from the liver, the accumulated CYP2E1 is no longer inhibited, and the aggregate CYP2E1 activity is increased above normal levels. [61] However, the stabilization against degradation is also reversed, resulting is a relatively rapid fall of enzyme activity to the baseline value (before ethanol consumption). When acetaminophen was given to ethanol-treated subjects immediately after the ethanol had cleared from the body, increased production of NAPQI was demonstrated. [61] This effect of inhibition followed by transient induction is mimicked by some other substrates of CYP2E1, including isoniazid. [65] This may account for reports of enhanced susceptibility to acetaminophen toxicity in patients treated with isoniazid. [66], [67]

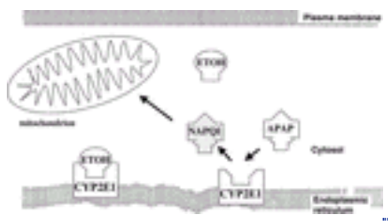

FIGURE 26-3. Effect of ethanol on the formation of the *N*-acetyl benzoquinone amine metabolite NAPQI from acetaminophen. The hepatotoxic metabolite is chiefly formed by CYP2E1. NAPQI appears to result in toxicity at least in part by causing mitochondrial toxicity. Ethanol binds to and is metabolized by CYP2E1. This is the presumed basis for inhibition of NAPQI formation while an individual is intoxicated. However, the ethanol-bound CYP2E1 is stabilized against intracellular degradation and therefore accumulates during prolonged periods of intoxication. When the individual stops drinking and ethanol leaves the liver, the accumulated CYP2E1 becomes catalytically active, causing an increase in NAPQI formation from acetaminophen. The effect is short-lived because the CYP2E1 is no longer stabilized against degradation once not bound to ethanol.

Techniques similar to those used in acetaminophen studies have been used to explore the role of P450s in liver injury produced by halothane. Halothane undergoes both reductive and oxidative metabolism in the liver. [68] The oxidative pathway of halothane metabolism has been implicated in producing a reactive metabolite (a trifluoroacetyl intermediate) that can bind covalently to proteins in the liver cell. These modified proteins are believed to stimulate an immunologic response, causing the severe form of liver injury associated with halothane. [69]

Pretreatment of patients with the CYP2E1 inhibitor disulfiram produced a 70% inhibition in the plasma and urine levels of trifluoroacetic acid, the stable product of the oxidative pathway. Methoxsalen pretreatment also produced a small reduction in trifluoroacetyl production, whereas troleandomycin pretreatment was without effect. [70] These studies indicate that the major enzyme involved in the oxidative metabolism of halothane is CYP2E1, followed by CYP2A6.

A variety of other studies have been performed to determine the role of specific P450s in liver disease produced by other drugs. [71], [72] and [73] In a few cases, positive results have been obtained. Five individuals who had completely recovered from liver toxicity caused by perhexiline (an antianginal medication used in Europe) were given debrisoquine to determine their CYP2D6 activity (see Table 26-2). [74] Four of the five individuals were characterized as CYP2D6 poor metabolizers. This frequency was significantly greater than anticipated (5% in whites). Perhexiline is believed to be metabolized by CYP2D6, and it is speculated that individuals deficient in CYP2D6 activity accumulate the drug in the liver. Toxicity could then be produced either by the parent drug, or might result from shunting metabolism to non-CYP2D6 pathways capable of generating reactive metabolites.

Patients who have recovered from chlorpromazine-induced liver injury have been reported to have both a reduced capacity for sulfoxidation of chlorpromazine [75] and an unusually high rate of hydroxylation of the drug. [75] The enzymes involved in sulfoxidation of chlorpromazine are not known, but the hydroxylation pathway appears to be catalyzed primarily by CYP2D6. A logical but as yet untested hypothesis is that individuals predisposed to chlorpromazine-induced liver injury are both ultrarapid metabolizers of CYP2D6 (because of gene duplication) and poor sulfoxidators (reflecting an as yet unidentified gene).

In most instances to date, however, attempts to link susceptibility to liver toxicity to variation in activity of specific P450s have failed. For example, there do not appear to be links between genetic deficiency in CYP2D6 activity and susceptibility to liver toxicity from several CYP2D6 substrates, including metoprolol, amitriptyline, and amodiaquine. [76] In addition, serum alanine aminotransferase elevations during treatment with the anti–Alzheimer disease drug tacrine are not associated with abnormal activity of CYP1A2, [77] even though this enzyme represents the major route of metabolism of tacrine and CYP1A2 has been implicated in the formation of a reactive metabolite from tacrine. [78] A final example is the nonsteroidal antiinflammatory drug diclofenac, which has been rarely associated with severe liver injury. [79] The major pathway of metabolism of diclofenac (and most nonsteroidal antiinflammatory drugs) is 4-hydroxylation catalyzed by CYP2C9. [80] However, attempts to link the risk for liver injury to functional mutations in CYP2C9 have been unsuccessful. [81]

Our limited success to date in using knowledge of P450s to predict those at risk for drug-induced liver injury probably reflects multiple factors. [44] Most of the hypotheses that have been tested involve examination of the major P450 involved in the overall metabolism of the drug in question. Toxic metabolites are often the result of minor pathways of metabolism that can be difficult to identify. Most importantly, the rate of production of a toxic metabolite by a given P450 is just one of many variables that are likely to determine whether toxicity actually occurs in a given patient.

Anti-P450 Antibodies and Drug-Induced Liver Injury

Liver disease caused by several different drugs is associated with circulating antibodies to P450s. [82], [83] Examples of anti-P450 antibodies are shown in Table 26-3. The current concept of the formation of these antibodies is that a P450 produces a metabolite so reactive that it binds directly to the P450, essentially immediately after the metabolite is formed. Antibodies are formed if this metabolite-drug adduct is antigenic and escapes from the cell to interact with immune cells. Controversy continues over whether anti-P450 antibodies actually mediate an immune attack on the liver, because no one has yet convincingly shown that they can cause liver injury in a living animal model. It remains possible that the antibodies are an epiphenomenon, appearing only after the antigens are released into circulation as hepatocytes are lysed by other mechanisms. However, cell lysis may not be necessary for antigen recognition because P450s appear to be present in low abundance on the outside of the liver plasma membrane. [83] Regardless of whether they mediate drug-induced liver injury, these antibodies can be used in immunochemical techniques to identify which specific human P450s are involved in generating the reactive metabolite, even if the structure of the reactive metabolite is unknown. The presence of anti-P450 antibodies can also be very helpful in establishing a drug as the cause of liver injury, although such tests are not widely available.

DRUG	MEAN FOLD INCREASE IN AUC*	REFERENCE
Simvastatin	13.5	96
Lovastatin	15.0	132
Midazolam	1.5	133
Triazolam	1.5	134
Saquinavir	2.0	135
Nifedipine	3.0	136
Carbamazepine	1.5	137
Buspirone	9.2	138
Cyclosporine	1.6	139

* Area under the blood concentration versus time curve divided by that observed when drug taken with water or orange juice.

TABLE 26-3 Some Drugs Affected by Grapefruit Juice

Intestinal P450s

P450 enzymes are detectable throughout the human digestive tract, including the esophagus, stomach, small bowel, and colon. [84] The concentration of total P450 enzymes is highest in the jejunum and gradually declines to lower levels in the ileum and colon. The major P450 in small bowel enterocytes is CYP3A4, accounting for at least 70% of the total P450s. [85], [86] and [87] CYP3A4 has been detected in the esophagus, stomach, and colon, but at much lower levels than in enterocytes. [85], [86] Other P450s detected in the digestive tract include CYP3A5, CYP2C9, CYP2C19, and CYP1A1. [88] Notably, CYP2C8, CYP2B6, and CYP1A2 have not generally been detected and appear to be liver-specific enzymes. In most cases studied, P450s are not found in the intestinal crypt cells, but they appear, probably as a result of transcriptional activation of the corresponding genes, as the epithelial cells migrate out of the crypt toward the villus tip (Fig. 26-4). This location is appropriate for the metabolism of xenobiotics in the gut because absorption is thought to occur chiefly in the upper part of the villus.

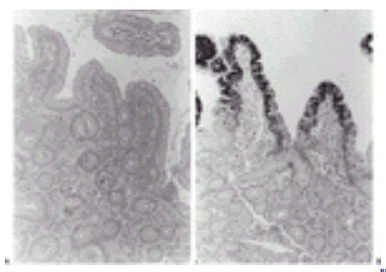

FIGURE 26-4. Induction of CYP3A4 in the small bowel of a patient treated with rifampin. This patient underwent duodenal biopsy (**A**) before and (**B**) after treatment with rifampin for 7 days. The biopsy specimens were fixed in formalin, sectioned, and subjected to immunoperoxidase staining after exposure to an antibody that specifically reacts with CYP3A4. CYP3A4 was detected only in mature enterocytes (*dark stain*) and was clearly induced by treatment with rifampin. In these sections, the antibody dilution was selected to maximally demonstrate induction. Lower dilutions of antibody revealed the presence of CYP3A4 in the mature enterocytes before treatment with rifampin (not shown). (From ref. [87].)

To date, the only intestinal P450 shown to catalyze significant drug metabolism is CYP3A4. This was originally demonstrated by intrajejunal administration of the CYP3A4 substrates cyclosporine [89] and midazolam [90] during the anhepatic phase of the liver transplant operation. In each case, up to 50% of the detectable drug absorbed into portal blood had been converted into metabolites characteristically produced by CYP3A4. Other studies have simultaneously administered unlabeled and stable labeled CYP3A4 substrates by intravenous and oral routes, respectively. If it is known that the drug is completely absorbed, it is possible to quantify intestinal metabolism. Such studies have shown that the intestine is a major site for metabolism of verapamil [91] and midazolam. [92]

The importance of intestinal CYP3A4 has also been highlighted by drug interactions produced by grapefruit juice. [93] A growing number of drugs have been shown to have enhanced oral availability when consumed with grapefruit juice (versus water or orange juice; see Table 26-3). This interaction has been shown to result from both competitive and mechanism-based inhibition of CYP3A4. In addition, the enterocyte content of CYP3A4 actually falls within hours after ingestion of a single glass of grapefruit juice, [94], [95] presumably because the inactivated enzyme undergoes rapid intracellular degradation. [95] The effect of grapefruit juice on the disposition of drugs is quite prolonged and is still detectable with some drugs up to 3 days after ingestion of a single glass of juice. [96] This presumably reflects the time required to replace the lost enzyme. The ingredients in grapefruit juice responsible for CYP3A4 inhibition appear to be compounds called *furanocoumarins* (also called *psoralens*). [95], [97] Furocoumarins are not present in most other types of fruit juice. Grapefruit juice does not appear to inhibit liver CYP3A4, at least when consumed in usual quantities. [93], [94] This presumably reflects insufficient concentrations of furanocoumarins reaching the liver after absorption. Grapefruit juice can therefore be used experimentally to “knock out” intestinal CYP3A4 (although it may affect some drug transporters as well). [98], [99] If grapefruit juice does not affect the oral availability of a drug, it can be assumed that intestinal CYP3A4 is not contributing much to the first-pass metabolism.

The aggregate CYP3A4 in the intestine is probably less than 10% of that in the liver. [100] For significant metabolism to occur in the intestine, it seems likely that a drug must be very rapidly metabolized by CYP3A4, be administered in relatively small amounts, or have a prolonged absorption phase (i.e., prolonged contact with the enzyme). It has been proposed that the physiological role of intestinal CYP3A4 may be primarily to limit entry into the body of potentially harmful dietary compounds. One study [101] suggested that CYP3A4 may convert aflatoxin ß $_1$ to reactive metabolites that bind to macromolecules within enterocytes. This, in effect, traps the metabolites within the cells. Because the enterocytes are normally sloughed into the digestive tract, the aflatoxin metabolites are eliminated in stool.

Intestinal CYP3A4 may also represent a significant pathway for the systemic clearance of some CYP3A4 substrates. This was suggested by the demonstration that an average of 8% of a dose of midazolam perfused through the mesenteric artery was metabolized in the intestine on a single pass. [90] In one patient, 25% of the systemically administered midazolam was metabolized in the intestine. [90]

In the general population, it appears that large differences in the activity of CYP3A4 are found in the intestine, just as in the liver. [102] There does not appear to be coordinate expression of the CYP3A4 gene in liver and intestine. [102] For example, a person can simultaneously have a relatively low level of CYP3A4 activity in the liver and relatively high levels of expression of the same gene in the enterocytes (and vice versa). Given the effects of grapefruit juice discussed above, it seems likely that as yet unidentified dietary factors may account, at least in part, for this discordance. In rats, for example, ethanol treatment and removal of iron or selenium from the diet results in the reduction in intestinal P450 activity but has little effect on P450 activity in the liver. [103], [104] and [105]

No natural dietary components have yet been reported to up-regulate the expression of CYP3A catalytic activity in the intestine of any species, including humans. However, in rats and humans, intestinal CYP3A enzymes have been shown to be inducible by at least some of the medications that also induce these enzymes in liver (see Fig. 26-4). [87] PXR is present in enterocytes and likely mediates this induction. It is likely that drug interactions involving CYP3A4 substrates that have previously been attributed to induction or inhibition of CYP3A4 in the liver may largely reflect these processes in the intestine.

The intestinal activity of CYP1A1 appears generally to be low but is greatly influenced by diet. When healthy volunteers are maintained on a semisynthetic diet, a significant fall in the catalytic activity characteristic of CYP1A1 is noted in biopsy specimens of intestinal mucosa. [106] In humans, polycyclic hydrocarbons that result from charcoal broiling can dramatically induce CYP1A1 in the intestine. [107], [108] In addition, CYP1A1 mRNA and catalytic activity are significantly induced in the intestine of volunteers treated with the ulcer medication omeprazole. [109] No studies have directly shown a significant contribution of intestinal CYP1A1 to first-pass metabolism of drugs. However, in one clinical study involving the consumption of charcoal-grilled hamburger, investigators found an inverse correlation between intestinal CYP1A1 expression and levels of polycyclic aromatic hydrocarbon protein adducts in circulating lymphocytes. [108] These investigators suggested that induction of CYP1A1 in the small intestine may be protective by reducing the absorption of polycyclic aromatic hydrocarbons.

PHASE 2 ENZYMES

In general, phase 2 enzymes have not been as well characterized as the P450s. This in part reflects the fact that phase 2 enzymes are generally colorless (so that purification is more difficult) and tend to lose catalytic activity during purification. The best-studied phase 2 enzymes are the uridine 5'-diphosphate (UDP)

glucuronosyltransferases (UGTs), the glutathione *S*-transferases (GSTs), and the sulfotransferases, which catalyze conjugation to glucuronic acid, glutathione, and sulfate, respectively.

UDP Glucuronosyltransferases

Conjugation to glucuronic acid results in enhanced water solubility; for this reason, glucuronide conjugates are usually readily excreted into urine. Glucuronide conjugates are also excreted into bile and, because of their water solubility, generally pass in the stool. In addition, glucuronidation generally results in reduced pharmacological activity. However, glucuronides can occasionally retain pharmacological activity. An unusual example of this is morphine glucuronide, which actually has roughly 50-fold greater opioid activity than morphine itself. [110] In addition, some glucuronides (particularly acylglucuronides) have been shown to be reactive molecules, capable of covalent binding to proteins. [111] For example, a glucuronide metabolite of diclofenac has been shown to covalently bind multiple proteins in the hepatocyte, and this covalent binding may contribute to the hepatotoxicity rarely associated with this drug.

Like the P450s, the UDP glucuronosyltransferases arise from a multigene family of microsomal enzymes, some of which are inducible. [112], [113] In humans, 15 UGT cDNAs have been identified comprising two gene families, UGT1 and UGT2. There are two gene subfamilies within the human UGT2 family (A and B) and one (A) within the human UGT1 family. The UGT1 family contains eight enzymes (designated by arabic numerals), and the UGT2 family contains seven enzymes. All UGT1A proteins have the identical 245 carboxyl-terminal amino acid sequence, and this reflects a process called *exon sharing*. There is considerable catalytic specificity of UGTs toward drugs, although such characterization is progressing relatively slowly. One problem has been that unlike the P450s, which exist on the outside of microsomes, the UGTs are located on the inside of microsomes. It is therefore often necessary to permeabilize microsomes to assess UGT activity, and this may introduce artifacts.

Multiple polymorphisms of the UGT1 family have been demonstrated. [114] The best studies of polymorphisms involve UGT1A1, which is the enzyme responsible for bilirubin conjugation. [112] Mutations in the UGT1A1 gene have been shown to account for Gilbert and Crigler-Najjar (types 1 and 2) hyperbilirubinemia. The implications of UGT polymorphisms in drug metabolism or hepatotoxicity are generally unknown. However, it has been proposed that a polymorphism in UGT1A6 may influence susceptibility to acetaminophen liver toxicity. [115] Anticonvulsants have been shown to induce some UGTs and may account for some drug interactions, particularly in patients treated for HIV infection. [30] The receptors involved in induction of UGTS are not currently known.

Multiple UGTs are expressed in the intestinal tract, [113] and in some cases, these are the identical enzymes expressed in the liver. However, some liver UGTs do not appear to be expressed in the digestive tract (e.g., UGT2B4), and other UGTs are expressed in intestine but not in liver (e.g., UGT1A8). It has been proposed that UGTs present in the colon may provide a protective function by glucuronidating compounds that are deconjugated by colonic bacteria. [113]

Glutathione *S*-Transferases

The GSTs involved in liver or intestinal drug metabolism are cytosolic enzymes belonging to eight separate gene families: a, µ, ?, and ?. [116] (Microsomal GSTs also exist, but these are generally not involved in drug metabolism.) Each family of cytosolic GSTs has several members designated by arabic numbers. For example, GSTM1 is the first of five members of the µ gene family. GSTs are expressed in many organs in addition to liver and intestine. The complements of GSTs present in liver and intestine are not identical. GSTs typically function to detoxify electrophilic metabolites of drugs, such as NAPQI produced from acetaminophen. In general, the reactions they catalyze are not rate-limiting in drug elimination. An exception may be busulfan, which requires glutathione conjugation catalyzed by GSTA1 for elimination. Studies have suggested that busulfan oral availability is limited by GST1A1 activity in the intestine. [117] Children appear to have a higher level of intestinal GST1A1 activity, and this has been proposed as the basis for an increased dosing requirement of busulfan in young children. [118]

Bile contains millimolar concentrations of glutathione, and it is believed that this serves to prevent intestinal cells from becoming depleted of substrate for GSTs. [119] A recent study has suggested that GSTs may exist outside enterocytes, within the overlying mucus, to detoxify electrophils present in the gut lumen. [120]

Polymorphisms in GSTs are common. [116] GSTM1 and GSTT1 are absent in 40% and 15% of whites, respectively. Ultrarapid metabolizers of GSTM1 substrates have also been identified, and this appears to reflect gene duplication. Because depletion of cellular glutathione enhances susceptibility to the toxicity of electrophilic metabolites, such as NAPQI, it is logical to assume that polymorphisms in GSTs could contribute to susceptibility to certain types of drug-induced liver disease. This appears to be the case for the anti–Alzheimer disease treatment tacrine; in patients receiving this drug who lack both GSTM1 and GSTT1, the incidence of treatment-associated alanine aminotransferase elevations appears to be increased. [121]

Sulfotransferases

Human liver and intestine are capable of sulfating a variety of xenobiotics by means of sulfotransferases, which are divided into five gene families. [122] A universal nomenclature has not yet been established for these cytosolic enzymes. The catalytic activity of the sulfotransferases varies considerably among individuals, and polymorphisms in specific sulfotransferases have been identified. Some sulfotransferases are inducible, at least in liver. For example, one study suggested that the loss of efficacy of birth control pills during treatment with antiseizure medications or rifampin might be caused by induction of sulfation, not just induction of CYP3A4 (see Table 26-1). [123]

ROLE OF TRANSPORTERS

It has been recently appreciated that, in addition to phase 1 and 2 enzymes, the disposition of drugs can be governed by the activities of proteins that function to transport the parent drug or its metabolites. [124] Some transport proteins function to secrete phase 1 or phase 2 metabolites from the hepatocyte into bile, [125] or into the intestinal lumen after they are formed in the enterocyte. [126] Such transporters are therefore sometimes collectively termed *phase 3*. The best-studied drug transporter is p-glycoprotein (Pgp), the product of the multidrug resistance gene MDR1. [127] Pgp is present in the biliary canaliculus, where if functions to pump some drugs and their metabolites into bile. Pgp is also present in the brush border of the enterocyte, where it secretes some drugs or metabolites into the lumen of the small bowel. Many, but not all, substrates for Pgp are also substrates for CYP3A4. In addition, Pgp is inducible by many of the drugs that induce CYP3A4, and this appears to be because activated PXR stimulates transcription of both genes. [128] Drug interactions that can be explained by the induction of CYP3A4 in liver or intestine may therefore also result from induction of Pgp in these organs. For example, induction of Pgp likely contributes to the interaction between rifampin and cyclosporine, a substrate for both CYP3A4 and Pgp. [10] Likewise, some drugs, such as ketoconazole, that inhibit CYP3A4 probably also inhibit Pgp.

Differentiating the contribution of Pgp from that of CYP3A4 in the disposition of a drug or in drug-drug interactions is not straightforward and is an active area of investigation. This is in part because CYP3A4 and Pgp may function synergistically. For example, mice lacking functional Pgp (because of targeted disruption of the mouse MDR genes) have higher rates of CYP3A-mediated demethylation of intravenously administered erythromycin. [129] This appears to result from increased hepatocyte concentrations of the parent drug secondary to decreased biliary secretion. In the intestine, Pgp may also function to increase CYP3A4-mediated metabolism by prolonging the absorption phase (i.e., increasing the duration of exposure of the drug to the enzyme), or by removing primary metabolites from the enterocyte, reducing secondary metabolism. [130], [131]

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CHAPTER 27

David A. Brenner and Richard A. Rippe

PATHOGENESIS OF HEPATIC FIBROSIS

OVERVIEW: CIRRHOSIS AND FIBROSIS

Cirrhosis is defined by its histological features, consisting of nodules of regenerating hepatocytes and the abnormal deposition of connective tissue in and around the nodules (fibrosis) (Fig. 27-1). Although the injury that initiated the process of regeneration and fibrosis may cease (and histological or laboratory evidence of inflammation and injury disappear), the fibrosis may be fixed and irreversible. By obliterating sinusoidal fenestrations, obstructing the space of Disse, and replacing vascular channels, fibrotic tissue increases resistance to blood flow. The resultant elevation of sinusoidal pressure causes shunting of portal flow around the liver, both exaggerating cellular dysfunction and creating the devastating consequences of portal hypertension. In addition, excess fibrosis prevents nutrient and metabolite exchange, further exacerbating the ramifications of the fibrotic response. Thus, liver fibrosis is a sine qua non of cirrhosis and directly responsible for many of its clinical features.

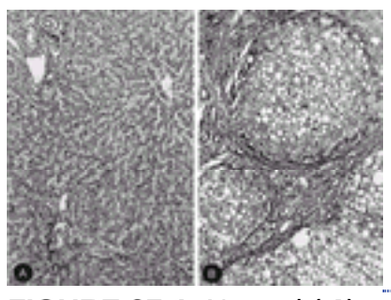

FIGURE 27-1. Normal (**A**) and cirrhotic (**B**) human liver. (From Brenner DA, Rippe RA. Pathogenesis of hepatic fibrosis. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Many different diseases may result in the common “end stage” of cirrhosis (Table 27-1). The spectrum of clinical abnormalities characterizing the cirrhotic stage of these diseases depends more on the extent of fibrosis, as well as the presence or absence of ongoing injury, than on the cause. Because so few of the underlying causes of cirrhosis are treatable, much attention has been focused on understanding, and potentially reversing, the generic process of fibrogenesis itself. However, it is unlikely that all diseases causing cirrhosis do so through the same initiating mechanism. For example, chronic hepatitis B, chronic ethanol abuse, and hemochromatosis may all result in cirrhosis. However, the liver in chronic hepatitis B characteristically demonstrates a lymphocytic infiltrate, whereas alcoholic liver disease is marked by hepatic infiltration with neutrophils, and infiltrates are absent in hemochromatosis. Furthermore, there are subtle differences in the specific content or location of deposited scar tissue in cirrhosis of different causes. Such differences may reflect differences in pathogenesis as well. If so, therapies successfully designed to limit fibrogenesis in one disease state may fail to work in another. One must bear this in mind when considering data obtained in any one model of hepatic fibrogenesis.

Infectious
Viral hepatitis (hepatitis B, C, D; cytomegalovirus infection)
Bacterial: brucellosis, syphilis
Parasitic: schistosomiasis
Environmental
Drugs: methyldopa, methotrexate, isoniazid, ethanol
Toxins: carbon tetrachloride, pyrrolidizine alkaloids
Metabolic
Hemochromatosis
Wilson disease
α_1 Antitrypsin
Glycogen storage disease
Gaucher disease
Other
Sarcoidosis
Chronic biliary obstruction
Cardiac cirrhosis

TABLE 27-1 Some Causes of Liver Fibrosis

This chapter reviews our current understanding of the pathogenesis of hepatic fibrosis and highlights those elements of the process for which intervention has been attempted. Conceptually, the process of hepatic fibrosis must, at some point, consist of the stimulation of excessive extracellular matrix (ECM) molecule synthesis by one or more populations(s) of cells (or an increase in the population of ECM-producing cells), or a decrease in the rate of ECM degradation or turnover.

HEPATIC CONNECTIVE TISSUE

For a parenchymal organ to maintain its cellular performance, a supporting framework of connective tissue must be present. The connective tissue may anchor the parenchymal cell, provide tensile strength for the tissue, control access to microorganisms or macromolecules, and act as a reservoir for growth factors. The nature and density of their ECM markedly affect the phenotypic features of many cells. Culturing hepatic stellate cells (HSCs), for instance, on laminin, type I collagen, or plastic can markedly change their shape, the type of ECM molecules they secrete, and their rate of production. [1], [2] and [3] At most epithelial-mesenchymal interfaces, this connective tissue is organized as a basement membrane. For instance, bile duct epithelium rests on a basement membrane. However, the hepatocyte is the only cell of ectodermal or endodermal origin in direct contact with plasma. Neither the hepatocyte nor the sinusoidal endothelial cells have the support of a complete basement membrane, so blood passing through the fenestrated sinusoidal endothelium need cross only the space of Disse to reach the microvilli of the hepatocyte cell surface. Thus, the connective tissue stroma of the normal liver has the difficult task of maintaining and supporting the structure of the organ without compromising the direct access of blood to the hepatocytes. By light microscopy, the only readily apparent connective tissue stroma in the liver is the basement membrane of the biliary ductules and blood vessels, and some less organized connective tissue within the portal tracts. However, immunohistochemical and electron microscopic studies have revealed that a complex low-density array of ECM molecules exists within the space of Disse. The primary constituents of this wispy sinusoidal stroma (as well as of basement membranes elsewhere) are glycoproteins, proteoglycans, and collagens. [4] Because liver collagen content markedly increases with

fibrogenesis, its biosynthesis represents a natural target for therapeutic intervention in liver fibrosis.

COLLAGEN SYNTHESIS

Collagen is the most abundant family of proteins in the vertebrate body. Collagen types I, III, IV, V, VI, XIV (undulin), and XVIII (endostatin) are the primary collagen types that have been identified in the liver. [5], [6] and [7] All collagen types share certain fundamental structural features. They are triple helical molecules, each strand of which is itself a helical polypeptide chain rich in proline and hydroxyproline and bearing a glycine residue at every third position. These strands, or a-chains, aggregate to form a triple helix interrupted in some collagen types by globular domains. The triple helices then cross-link to form collagen fibrils, each type with unique physiochemical properties.

Type I collagen forms large fibrils and is the major ECM protein of bone, tendon, skin, ligaments, fascia, and arteries. It is also the major collagen type synthesized in the fibrotic liver, and its production serves as a model for understanding collagen synthesis. Type I collagen undergoes a complex biosynthetic pathway (Fig. 27-2). [8], [9], [10], [11] and [12] The type I collagen molecule is composed of two a $_1$(I)-chains and one a $_2$(I)-chain that are produced by two separate genes located on different chromosomes. These genes are coordinately regulated in a tissue-specific, developmental, and inducible manner. Generally, twice as much of the a $_1$(I) collagen gene is transcribed as of the a $_2$(I) collagen gene. Once the two collagen genes are transcribed, the mRNAs are processed in the nucleus.

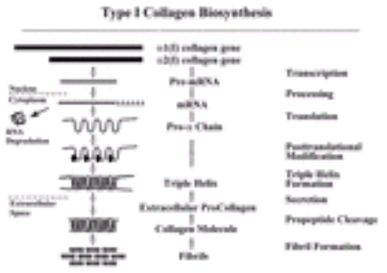

FIGURE 27-2. Type I collagen biosynthesis.

One of the consequences of HSC activation is a dramatic increase in the production of type I collagen. The increase results from a two- to threefold increase in the transcription rate of this gene, coupled with a 16- to 20-fold increase in the stabilization of the mRNA of the gene (Fig. 27-3). [13] The stabilization of the a $_1$(I) collagen mRNA is a complex process. One aspect of the stabilization process involves the binding of aCP2, an RNA-binding protein, to conserved elements located at the 3' end of the mRNA molecule. Interactions between aCP2 and poly-A binding protein decrease mRNA degradation. A conserved stem-loop structure located in the first exon of the a $_1$(I), a $_2$(I), and a $_1$(III) collagen genes interacts with a 120-kd unidentified protein. [14] This protein may also interact with aCP2 and with the cap-binding protein, a protein that binds at the 5' end of the mRNA molecule. [9] These interactions contribute to increased stability of the a $_1$(I) mRNA molecule and increase translation of the protein.

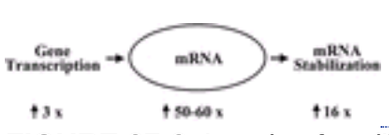

FIGURE 27-3. Levels of a $_1$(I) collagen mRNA increase following hepatic stellate cell activation.

Once processed, the mature mRNA molecule is transported to the cytoplasm, where it is translated into protein. Collagen I mRNAs code for “prepropeptide” chains, meaning that the translated products have “leader” sequences serving to guide the newly made molecules into the endoplasmic reticulum. Further, amino- and carboxyl-terminal “propeptides” are present on each chain. Inter- and intrachain disulfide bond formation in these globular terminal regions is necessary for a-chain aggregation. After removal, propeptides may regulate collagen gene transcription.

Leader sequences are cleaved as the prepropeptide a-chains enter the rough endoplasmic reticulum where important posttranslational modifications occur. Hydroxylation of some prolyl and lysyl residues to hydroxyproline and hydroxylysine is catalyzed by three hydroxylases, each requiring oxygen, a-ketoglutarate, ferrous iron, and ascorbate as cofactors. [15] Within the triple helical region, every third proline residue is hydroxylated, unlike every lysine residue present in the polypeptide chain. These reactions occur only when the prolyl or lysyl residue occupies specific positions in the a-chain and are a prerequisite for the a-chain interactions resulting in the triple helical procollagen molecule. [15] This posttranslational modification stabilizes the formation of the triple helical structure. [15] Additionally, some hydroxylated prolyl or lysyl residues are glycosylated. [11], [16] The procollagen molecule passes through the Golgi apparatus, and propeptides are enzymatically cleaved off at the time of collagen secretion into the extracellular space, which requires stable triple helix formation for maximal efficiency. Free propeptides may remain within the cell or between cells in the matrix, or they may enter the circulation, from which they are cleared by the kidneys and by endothelial cell uptake. [17] Once the propeptides are cleaved, spontaneous aggregation of mature collagen molecules takes place to form fibrils. For full tensile strength, the fibrils require covalent intramolecular and intermolecular cross-linkage. These extracellular cross-linkages are enzyme-catalyzed reactions. Finally, concomitant with collagen synthesis is collagen degradation, both intra- and extracellularly. The mechanisms controlling these pathways are poorly understood, but in some systems, most of the transcribed collagen is never deposited.

Several aspects of collagen synthesis deserve emphasis. First, the intracellular posttranslational hydroxylation of proline and lysine is required for pro-a-chain aggregation into triple helices. [11] Interruption of hydroxylation reduces the formation of procollagen. Extracellular lysyl oxidase activity is required for collagen cross-linking and stable fibril formation. [18] Inhibition of this enzyme results in unstable collagen. Ultimately, at least nine posttranslational enzymes are required for collagen production, so that opportunities for therapeutic intervention are numerous.

Hepatic Basement Membrane Proteins

Connective tissue organized as basement membrane is found throughout the body. The primary molecular constituents are collagen, glycoproteins, and proteoglycans. [4] Within the liver, these molecules make up the large vascular, bile duct, and neural basement membranes. Although the molecular details of organization are unclear, their structure results in a 20- to 60-nm-thick lamellar unit distinctly recognizable by electron microscopy.

Type IV collagen is the major collagen of basement membrane. It is an unusual collagen type in that the propeptides are not cleaved off on secretion, and the molecules cross-link at these globular domains to form a network resembling “chicken wire.” [19], [20] The major glycoprotein of the basement membrane is laminin, a large cruciform molecule bearing domains that bind to type IV collagen, fibronectin, heparin sulfate, nidogen (also called *entactin*), and specific cell surface proteins. [20] Laminin represents a family of large glycoproteins sharing some structural features and playing important roles in guiding normal embryogenesis and perhaps tissue repair. [20], [21] Nidogen, a smaller glycoprotein, is usually found bound to the center of the laminin molecule, where it may mediate binding to other matrix proteins. [22] Proteoglycans are a heterogeneous group of proteins, grouped together by their sulfated carbohydrate groups. The primary epithelial basement membrane proteoglycan is heparin sulfate, with polymers of *N*-acetylglucosamine and uronic acid attached to a protein core. [23] Proteoglycans may contribute to the basement membrane structure through binding sites for cell surface proteins or other ECM molecules. Thus, the structure and constituents of hepatic basement membranes resemble those of basement membranes elsewhere in the body. However, as already noted, the sinusoidal endothelial cells and hepatocytes lack basement

membranes, and it is the accumulation of sinusoidal fibrosis that characterizes hepatic cirrhosis.

Non–Basement Membrane Proteins

Although the molecules mentioned thus far are the primary constituents of basement membrane, they are also found in sparser, less organized fashion throughout the portal tract stroma and the space of Disse. Laminin and collagen IV, for instance, may be demonstrated by immunohistochemical staining to be discontinuously present throughout the space of Disse. However, other important hepatic ECM molecules, such as proteoglycans from the chondroitin sulfate/dermatan sulfate group, the glycoprotein fibronectin, and the major fibrillar collagens of the liver, types I and III, are not prominent in the basement membrane complex. These are the molecules in the space of Disse that make up the insoluble portion of the hepatocytes' ECM.

Approximately 40% of normal liver collagen is type I collagen. It is found in the stroma surrounding portal tracts, central veins, and the liver capsule, where it is probably the blue material visualized by trichrome staining. However, scattered strands may also be detected by immunohistological techniques along the sinusoids in the space of Disse, particularly at points of sinusoidal branching. Another 40% of hepatic collagen is type III, forming slightly smaller fibrils that distribute together with type I collagen. Collagen types V and VI are much less abundant in liver tissue. [24] Type V collagen is probably a pericellular collagen, acting perhaps as an "ectoskeleton" around cells. It represents no more than 2% of liver collagen. Type VI collagen is the rarest collagen in the liver and appears to form microfibrils, loosely connecting bundles of larger fibrillar collagen types I and III. [25]

Glycoproteins found in the space of Disse include laminin and the large glycoprotein fibronectin. A number of fibronectin types arise from alternative splicing of a single gene product. [26], [27] All have two subunits joined by disulfide bonds, and each type bears domains binding fibrin, heparin, bacteria, denatured collagen, cells, and probably other moieties as well. Fibronectin is found in the portal stroma and capsule, but it also may be identified in continuous contact with hepatocytes as the most prominent component of ECM within the space of Disse. A circulating form of fibronectin may lodge in the ECM, but deposition following local secretion also occurs.

Cell surface receptors have been identified for collagens, fibronectin, laminin, and proteoglycans, including integrins, dystroglycan, receptor tyrosine kinases or phosphatases, and syndecans. Integrins, the best studied of the cell matrix receptors, are a family of proteins that serve as transmembrane links between extracellular ligands and the cell cytoskeleton. Integrins are heterodimer molecules that contain one a-chain and one ß-chain. The several different a- and ß-chains allow for a wide variety of combinations of a-chains and ß-chains and multiple possibilities for ligand recognition. The primary collagen-binding integrins are a $_1$ß $_1$ and a $_2$ß $_1$. [2], [28], [29] and [30]

CELLULAR SOURCES OF EXTRACELLULAR MATRIX PROTEINS

Each of the liver cell populations may play a role in the production of ECM molecules, either directly by secretion or indirectly through the production of cytokines or mediators of inflammation. Knowledge of the cellular origins of these molecules could be very important therapeutically. For instance, because normal ECM production is critical to normal tissue function throughout the body, ideal therapy for liver fibrogenesis entails the delivery of antifibrogenic drugs only to liver cells producing excessive amounts of ECM proteins.

Although a variety of hepatic cells are capable of synthesizing collagen, a preponderance of experimental evidence incriminates the HSC as the main collagen-producing cell during hepatic fibrogenesis. [4], [5], [31], [32], [33] and [34] HSCs are located in space of Disse in close proximity to hepatocytes on one side and endothelial and Kupffer cells on the other side. HSCs (also called *fat-storing cells, Ito cells,* or *hepatic lipocytes*) are desmin-positive perisinusoidal cells that are the primary cell in the body responsible for vitamin A storage in the form of retinyl esters. These cells have the capacity to synthesize several ECM proteins and glycoproteins, including at least five collagen types, fibronectin, laminin, entactin, tenascin, and several proteoglycans. [35], [36], [37], [38], [39], [40], [41] and [42]

Another source of excessive ECM deposited in the liver is the portal fibroblast. In primary biliary cirrhosis, excess deposed collagen is characteristically located around the fibroblasts surrounding the portal tracts. [43]

Modulation of the ECM by a complex interaction of cytokines acting in paracrine fashion has provided insight into liver fibrogenesis. Cells producing these cytokines may have an indirect effect on the ECM by their secretion. Many of these modulating cells are of hematopoietic origin. For example, the degranulation of platelets may provide a rich source of transforming growth factor-a (TGF-a). This factor may influence both the proliferation and ECM production of fibroblasts, hepatocytes, and HSCs. Platelet-derived growth factor and TGF-a–like activity can both be derived from platelets, but their impact on fibrogenesis is less clear. Because platelets accumulate in the vicinity of injury, the paracrine effects of these compounds may be important in the fibrogenesis that follows some forms of chronic liver injury.

The resident macrophages of the liver, the Kupffer cells, probably along with itinerant monocytes and other hepatic cell types, are a rich source of cytokines and chemokines. [44], [45], [46] and [47] In addition to TGF-ß, macrophages can secrete tumor necrosis factor-a (TNF-a), interleukin-1 (IL-1), prostaglandins, interferons, and collagenase. Complex interactions can result from these substances under various conditions. Cytokines can either regulate the synthesis of other cytokines or can modify the actions of other cytokines at the target cell. Thus, macrophages may have an important role in fibrogenesis, but whether they augment or attenuate fibrogenesis is unclear. In many hepatic fibrotic diseases, a clear inflammatory process is accompanied by a polymorphonuclear infiltrate. However, not all hepatic fibrotic conditions are associated with this situation. In liver fibrosis induced by carbon tetrachloride (CCl_4), a clear fibrotic response develops, but without a polymorphonuclear infiltrate. Similar phenomena, less well studied, probably pertain to lymphocytes passing through the hepatic sinusoids. Neutrophils may also play a role in some states of hepatic fibrogenesis. Neutrophils may be responsible for local tissue destruction through the secretion of reactive oxygen species leading to accumulation and degranulation of platelets. Specific chemotactic substances, such as leukotriene B_4, can recruit and activate macrophages. Additionally, neutrophils produce collagenases that may have important local effects. [48], [49] Thus, the role of the neutrophil in liver fibrosis is probably a consequence of its ability to cause injury and activate cytokine-producing cells.

MATRIX-DEGRADING ENZYMES

More is known about the constituents of the ECM structure than about the distribution and activity of ECM-degrading enzymes in the liver. Nevertheless, their importance is implied by the dynamic state of normal ECM structure. For instance, whereas fractional turnover rates for collagens in organs are generally 3% to 5% per day, some tissues may have rates as high as 10%. [50] Collagen degradation can take place either inside or outside the cell. The proportion of degradation occurring intracellularly before secretion ranges from 10% to 90% in various cell culture systems, apparently through degradation in lysosomes or endoplasmic reticulum. Furthermore, phagocytosis and intracellular degradation of collagen after secretion are well described, so that more than one route to intracellular degradation exists. The role of these pathways in states of active fibrogenesis is unknown. Whether similar intracellular degradation of other ECM molecules occurs is also unknown.

The matrix metalloproteinases (MMPs) are a family of at least 20 enzymes that degrade matrix proteins and are thus considered to be important in mediating matrix turnover. [51] They are secreted as inactive proenzymes and require proteolytic activation. Several enzymes can activate the pro-metalloproteinase; however, plasmin is the most common. [51], [52] and [53] The MMPs are regulated by several mechanisms, including transcriptional regulation, activation of the latent proenzyme, and extracellular inhibition by specific tissue inhibitors of metalloproteinases (TIMPs). The MMPs can be divided into four groups based on their substrate specificity. [52] Representing the first group are the collagenases, MMP-1 and MMP-2. These enzymes are involved in degrading fibrillar collagen, including collagen types I, II, and III. They cleave the triple helical structure of collagen, allowing for subsequent degradation of the molecule by other MMPs. [54] The second group of MMPs are the stromelysins, which degrade collagen types II, IV, IX, X, and XI, denatured collagens (gelatin), laminin, proteoglycans, and fibronectin. [54] The third group of MMPs include the gelatinases: MMP-2 (gelatinase A) and MMP-9 (gelatinase B). These enzymes degrade gelatins, elastin, and collagen types IV, V, VII, X, and XI. [54] The fourth group of MMPs represent a miscellaneous group that includes membrane-type MMPs, which degrade types I and III collagen, proteoglycans, and fibronectin. [52] The activity of active MMPs is regulated by a group of four proteins, the TIMPs. [52] TIMPs irreversibly bind to active MMPs to prevent their proteolytic activity. [54] In addition to regulation at the level of transcription, translation, and activation, these metalloproteinases may be affected by inhibitors such as a $_2$-macroglobulin, a circulating liver-produced protein. [54] Thus, a complicated interaction of factors controls normal and hepatic ECM degradation.

MMP-2, type IV collagenase/gelatinase, is a metalloproteinase produced by fibroblasts, macrophages, and HSCs. [55], [56] MMP-2 degrades denatured collagen as well

as collagen types IV and V. On activation both in vitro and in vivo, HSCs express type IV collagenase/gelatinase. [57], [58], [59] and [60] In vivo, the expression of MMP-2 may disrupt the basement membrane, which includes type IV collagen. Fibroblasts, endothelial cells, HSCs, and macrophages produce MMP-13 (also called *stromelysin* or *transin*), a metalloproteinase with broad activity, capable of enhancing the removal of propeptides from collagen I and III, thereby enhancing procollagenase activation, and digesting proteoglycans, collagen IV, laminin, fibronectin, and denatured collagen. [61], [62]

THE EXTRACELLULAR MATRIX IN CIRRHOSIS

The ECM has been examined in cirrhotic humans and in several animal models of hepatic fibrogenesis. In general, during fibrogenesis, collagen mRNA levels increase about fourfold, and liver collagen content increases about 10- to 20-fold. [4], [24], [63], [64] Lysyl oxidase activity increases as well.

Type III collagen is the first to increase after liver damage. [65] However, type I collagen becomes the dominant hepatic collagen type, exceeding collagen III by a ratio of 4:1, and may eventually account for 60% to 70% of total hepatic collagen. [4], [66], [67] Laminin mRNA levels also rise, and liver laminin content accordingly increases. [31], [68], [69], [70] and [71] The normal neural, ductular, and vascular basement membranes may remain undisturbed, but ECM molecules accumulate in the space of Disse, with fibronectin preceding type I collagen, type IV collagen, and laminin (Fig. 27-4). [69], [72], [73] Gradually, a continuous ECM accumulates between hepatocytes and endothelial cells, obliterating sinusoidal fenestrations and preventing the exchange of nutrients and metabolites between the hepatocytes and the hepatic sinusoid. This "capillarization of the sinusoids" and "collagenization of the space of Disse" precede the development of the signs and symptoms of chronic liver disease. Additionally, an early hallmark of alcohol-induced hepatic fibrosis is the deposition of excess ECM in the terminal hepatic venule.

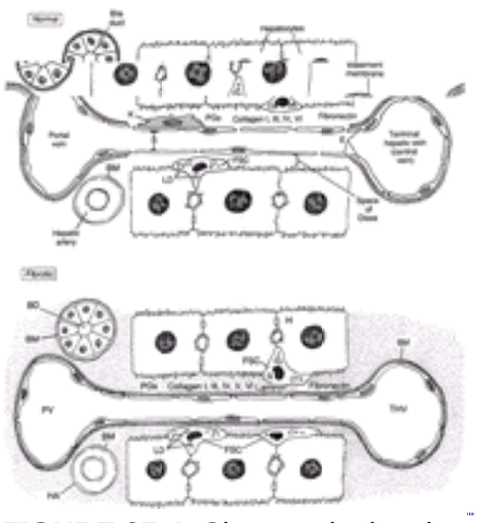

FIGURE 27-4. Changes in the sinusoidal space between normal and fibrotic liver. *BD,* bile duct; *BM,* basement membrane; *E,* endothelial cells; *FSC,* fat-storing (stellate) cells; *H,* hepatocyte; *HA,* hepatic artery; *K,* Kupffer cell; *LD,* lipid droplets; *PG,* proteoglycans; *PV,* portal vein; *S,* sinusoid; *THV,* terminal hepatic (central) vein. (From Brenner DA, Rippe RA. Pathogenesis of hepatic fibrosis. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

MMP (collagenase) activity has been reported to increase early during fibrogenesis, presumable less so than collagen deposition. Indeed, it has been suggested that excessive or inappropriate matrix degradation may be among the causes of fibrosis by eliminating or distorting the normal extracellular ECM "cues" guiding the normal expression of ECM genes. Later, collagenolytic activity declines and a new steady state is reached. Expression of the TIMPs is also increased during liver fibrosis. [74], [75] However, it is noteworthy that in a number of models of fibrogenesis, prolonged exposure to the injurious agent is required to achieve cirrhosis, whereas transient injury is followed by gradual resolution of the fibrosis. [76], [77] and [78] Possible explanations are that the gradual loss of collagenolytic activity with prolonged fibrogenesis leads to progressive fibrosis, or that the increase in type I collagen deposition occurring during fibrogenesis renders collagenolytic activity decreasingly effective in maintaining normal matrix integrity.

Mechanisms of Increased Collagen Production

Oxidative stress is an important factor in the development and progression of liver fibrosis of different causes, including alcohol ingestion, viruses, iron or copper overload, cholestasis, and hepatic blood congestion. In oxidative stress, highly reactive oxygen intermediate species (ROIs) are generated. The ROIs can oxidize many proteins, but lipids represent a major target. The oxidized molecules can have effects detrimental to cellular metabolism. The role of the oxidation of lipids, lipid peroxidation, has been extensively studied in liver fibrosis.

Lipid Peroxidation Lipid peroxidation is associated with the tissue injury and fibrogenesis of several pathological disorders, including atherosclerosis, iron overload, and porphyria, and also with ethanol-, bleomycin-, and CCl_4-induced liver toxicity. The polyunsaturated fatty acids of phospholipids in cell membranes are highly susceptible to attack by free radicals, and subsequent reactions with oxygen generate lipid peroxides. Lipid peroxides may be further oxidized to aldehydes, such as 4-hydroxy-2,3-nonenal (HNE) and malondialdehyde (MDA). Peroxidation resulting in these highly reactive aldehydes stimulates collagen gene expression in cultured fibroblasts. [79], [80] Incubating these cultures with exogenous MDA, a major product of lipid peroxidation, induces a similar increase in collagen gene expression. Furthermore, decreasing the basal level of lipid peroxidation in these cultures with a-tocopherol (vitamin E) prevents lipid peroxidation and blocks the stimulation of collagen production and collagen gene expression by ascorbic acid. Thus, it may be that the "direct" effects of ascorbate to increase collagen production actually reflect the effects of ascorbate-enhanced lipid peroxidation. The stimulation of lipid peroxidation with ascorbic acid, acetaldehyde, or MDA stimulates collagen gene transcription, and inhibition of lipid peroxidation with vitamin E or methylene blue inhibits transcription. HNE also increases collagen production in cultured HSCs. [81], [82] and [83] The stimulatory effect of lipid peroxidation products on collagen expression that occurs in vitro is also seen in vivo.

Ethanol-Induced Fibrosis Excess ethanol consumption represents the number one cause of liver fibrosis in the United States. Ethanol-induced liver fibrosis is strongly associated with oxidative stress. The fibrotic effect of ethanol is caused in part by the generation of ROIs (including 1-hydroxyethyl) via cytochrome CYP2E1. [84] Inhibiting CYP2E1 reduces the formation of ROIs and the generation of lipid peroxidation products in ethanol-fed animals. [85], [86] The generated ROIs are believed to affect HSCs directly, resulting in increased ECM production. In addition, ROIs can activate Kupffer cells, which may increase proinflammatory and profibrogenic cytokine production. ROIs also induce the formation of lipid peroxidation products from membrane lipids, of which HNE and MDA are the major species produced. Both HNE and MDA levels and their modified protein adducts are increased in alcoholic patients. [87] HNE and MDA levels are increased, along with collagen deposition, after the treatment of chronic alcoholism. [88] Alternatively, ROIs from ethanol can be generated by NADPH (reduced nicotinamide adenine dinucleotide phosphate) oxidase in Kupffer cells. [89] Ethanol also promotes fibrogenesis through its primary metabolite, acetaldehyde. The mechanism by which acetaldehyde increases collagen transcription is unknown. Acetaldehyde covalently binds to proteins via Schiff bases, especially with the a-amino group of lysines, forming acetaldehyde-protein adducts. These adducts can be detected in the liver of ethanol-fed rats, and antibodies to acetaldehyde adducts are present in the serum of most alcoholic individuals. [90], [91], [92], [93], [94] and [95] Perhaps the formation of adducts of acetaldehyde with proteins involved in transcriptional regulation is the mechanism by which collagen gene transcription increases. Acetaldehyde is a highly reactive compound that can form aldehyde adducts with many proteins, often altering their function. Acetaldehyde stimulates collagen expression in fibroblasts and in cultured HSCs. [96], [97] and [98] This stimulatory effect may be caused by acetaldehyde increasing the binding of transcription factors to the promoters of the collagen genes. [99], [100] and [101] The activation of c-jun nuclear kinase (JNK) appears to be involved in the mediation of the increase in collagen expression by acetaldehyde by increasing transcription factor binding to the collagen promoter. [102], [103] The interaction of acetaldehyde with xanthine oxidase may also promote lipid peroxidation. Differences in acetaldehyde metabolism, either genetically influenced or acquired, result in a range of hepatic acetaldehyde concentrations. If acetaldehyde plays a role in alcohol-induced fibrogenesis, then perhaps fibrosis develops only in those individuals with high hepatic concentrations of acetaldehyde. This could explain the predisposition of some, but not all, alcoholic individuals to the development of hepatic fibrosis. Acetaldehyde may also be an important mediator of hepatic fibrogenesis in nonalcoholic liver disease. High levels of acetaldehyde are generated in the CCl_4-injured rodent liver after the administration of ethanol (an endogenous source of acetaldehyde). CCl_4 liver injury promotes the excessive accumulation of acetaldehyde from ethanol by decreasing acetaldehyde production via an alternative degradation pathway. This observation provides support for the commonly held belief that patients with nonalcoholic liver disease should not drink alcohol.

Iron and Copper Overload Iron overload in hemochromatosis and copper overload in Wilson disease are other causes of ROI formation and fibrosis. The accumulation of iron results in the formation of reactive oxygen through the Fenton reaction. This leads to the oxidation of internal membrane lipids of the mitochondria, microsomes, and lysosomes; the resultant generation of lipid peroxidation products (HNE and MDA) and associated protein adduct formation in turn lead to cell injury caused by functional problems with cellular structures. [104], [105] and [106] Studies have shown that increased iron increases collagen gene expression. [107], [108] The role of oxidative stress in the development of iron-induced fibrosis has been confirmed in studies in which antioxidants, such as vitamin E and silybin, are used to reduce the levels of lipid peroxidation products and the development of fibrosis. [109], [110] Increased iron thus increases lipid peroxidation, causing irreversible cellular damage. This can activate Kupffer cells that produce ROIs and fibrogenic cytokines that stimulate the HSCs to produce excess ECM components. The molecular mechanisms responsible for iron-induced collagen gene expression are unknown; however, increased binding of Sp1 to the collagen gene promoter has been shown in HSCs following iron treatment. [111] The mechanisms by which iron overload causes fibrosis have been studied more than those for copper overload. Copper overload results in a chronic inflammatory response accompanied by significant fibrosis that can eventually lead to cirrhosis. Excess copper increases lipid peroxidation products that can be reduced with antioxidant (vitamin E) treatment. [112]

Transforming Growth Factor-ß TGF-ß belongs to a superfamily of growth hormones and cytokines that affects growth and tissue patterning in many tissue types. TGF-ß superfamily members regulate the transcription of several genes, including those involved with regulating the cell cycle, cell adhesion molecules, homeobox genes, and extracellular matrix proteins. TGF-ß represents a family of five peptide hormones (TGF-ß $_{1–5}$) that are primarily produced and secreted by inflammatory cells and platelets. Members of the TGF-ß family play important roles in several physiological processes, including embryogenesis, wound repair, inflammation, carcinogenesis, immunosuppression, and fibrogenesis. The exogenous administration of TGF-ß induces fibrosis in lung, kidney, and liver, [113] and TGF-ß is thus considered to be a potent fibrogenic cytokine. During experimental models of hepatic fibrosis, including bile duct ligation, CCl_4 administration, and schistosomiasis, a prolonged increase in TGF-ß expression is observed. [114], [115] Patients with alcohol- or virus-induced cirrhosis also exhibit increased TGF-ß mRNA levels. TGF-ß induces fibrogenesis by multiple mechanisms. It increases the production and decreases the degradation of ECM proteins, so that an overall net increase in the deposition of these proteins results. TGF-ß increases the expression of ECM proteins such as type I collagen and fibronectin and increases type I collagen synthesis in cultured HSCs. [116], [117] The increase is the consequence of an increase in both the transcription rate of these genes and posttranscriptional regulation. Three potential TGF-ß–responsive elements have been identified in the a_1(I) collagen gene. [10] An increase in TGF-ß receptor expression occurs following HSC activation, which may allow an autostimulatory loop to enhance the TGF-ß effects in HSCs. TGF-ß decreases the expression of Sp3, a negative regulator of transcription. This may allow for increased binding of Sp1 to the collagen gene promoter, thereby increasing collagen gene expression, as Sp1 is a potent transactivator of the a_1(I) collagen gene. [118], [119] TGF-ß also increases the levels of plasminogen activator inhibitor (PAI-1) and TIMPs, both of which inhibit the degradation of ECM components. [120] In addition, TGF-ß inhibits the production of MMP-1 (type I collagenase) and MMP-3 (stromelysin). TGF-ß also increases the rate of HSC activation. [121] This in vitro analysis is corroborated by in vivo analysis, which has shown increased levels of TGF-ß mRNA in liver biopsy specimens from patients with chronic fibrosing liver disease that was positively correlated with fibrosis. [122] TGF-ß was localized by immunohistochemical staining to areas of hepatic fibrosis in biopsy specimens of patients with cirrhosis. [122], [123] TGF-ß plays a central role in the production of pathological ECM deposition in the liver. HSCs in the liver respond to TGF-ß by increasing the secretion of collagen and fibronectin into the ECM. TGF-ß functions in part by stimulating transcription of the collagen and fibronectin genes. TGF-ß increases the transcription of type I procollagen genes in HSCs. [116], [117] An increase in fibronectin mRNA is also seen following TGF-ß treatment in HSCs. The increased transcription of collagen and fibronectin genes results in increased production of these proteins. TGF-ß is secreted as a homodimer latent peptide that is subsequently activated to an active form by the dissociation of the inhibitory latency-associated peptide. The active form of TGF-ß interacts with three specific cell surface receptors: RI, RII, and RIII (Fig. 27-5). Type III TGF-ß receptors contain short cytoplasmic tails and do not mediate TGF-ß signal transduction, but they may bind TGF-ß and present it to type II receptors. Types I and II receptors contain serine/threonine kinases that are directly responsible for cellular signaling of TGF-ß. Active TGF-ß initially binds to RII, which then recruits and phosphorylates the RI receptor. This initiates a specific intracellular signaling process. The SMAD proteins, and in particular SMAD2, SMAD3, and SMAD4, mediate the intracellular signaling of TGF-ß. [124], [125] Three main classes of SMAD proteins exist: the receptor-regulated SMADs (R-SMADs: SMADs 1, 2, 3, 5, and 8), the common-partner SMAD (co-SMAD: SMAD4), and the inhibitory SMADs (I-SMADS: SMADs 6 and 7).

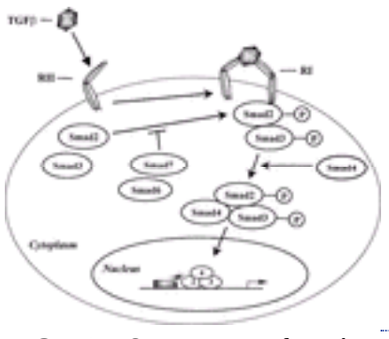

FIGURE 27-5. Transforming growth fac- tor-ß signaling.

The importance of SMAD proteins in the cell is demonstrated in knock-out animals. Both SMAD2 knock-out and SMAD4 knock-out result in embryonic lethality in mice. SMAD2 knock-out mice show a lack of anterior-posterior specification and do not develop mesoderm; SMAD4 knock-out mice fail to grow because of reduced cell proliferation, and they demonstrate gastrulation defects. SMAD3 knock-out mice are viable, survive to adulthood, and are fertile, but they are smaller, exhibit limb malformations, have a defective immune system, and develop colon cancer at approximately 6 months of age. Fibroblast cultures isolated from the SMAD3-deficient mice show an inability to form SMAD complexes on the DNA and fail to induce transcription from a TGF-ß–responsive promoter. SMAD3 is not required for HSC activation, but SMAD3 is required for maximal type I collagen gene expression in HSCs. [126] The HSC response to TGF-ß appears to wane over time in culture. During the early stages of HSC activation, these cells are fully responsive to TGF-ß; however, over time, fully activated HSCs lose their TGF-ß responsiveness. [127] This apparently is caused by a loss of TGF-ß–dependent SMAD2 phosphorylation. The molecular mechanisms underlying this unresponsiveness are not well understood.

ACTIVATION AND PROLIFERATION OF FIBROGENIC CELLS

An increase in the number of collagen-producing cells, including HSCs and fibroblasts, as a consequence of proliferation, migration, or transformation is a mechanism contributing to hepatic fibrogenesis.

In tissue injury and fibrogenesis, platelets may recruit collagen-producing cells by releasing two of their factors, platelet-derived growth factor (PDGF) and TGF-a. TGF-a and PDGF are chemoattractants for fibroblasts, smooth muscle cells, and monocytes. Furthermore, PDGF induces proliferation of fibroblasts, smooth muscle cells, and HSCs. [128] Monocytes, macrophages, and neutrophils may also be recruited by leukotriene, by the cell attachment segment of fibronectin, and by a polar lipid generated by hepatocytes metabolizing ethanol. [129] Activated monocytes, including Kupffer cells, are capable of generating fibroblast and HSC growth-promoting factors, including IL-1, PDGF, and fibroblast growth factor. [121], [130]

Regardless of etiology, hepatic injury induces the HSC to undergo a complex transformation or “activation” process into a myofibroblast-like cell (reviewed in ref. [34]; Fig. 27-6). Following a fibrogenic stimulus, the HSC undergoes activation with morphologic changes, including the loss of vitamin A stores and the appearance of rough endoplasmic reticulum. Early activation is also characterized by the expression of smooth muscle a-actin (which is why these cells have been called *myofibroblast-like cells*), cytokine receptors on the cell surface, and type IV collagenase that disrupts the normal basement membrane. If the fibrogenic stimulus persists, the cells continue to lose their vitamin A stores and become responsive to growth factors, of which PDGF is the most potent, and to fibrogenic cytokines, of which TGF-ß is the most potent. This amplifies the cell population and increases fibrogenesis. HSC activation is also associated with additional changes in the pattern of gene expression. These include the synthesis of several modulators of ECM homeostasis that ultimately alter the delicate balance between matrix synthesis and degradation and lead to fibrosis.

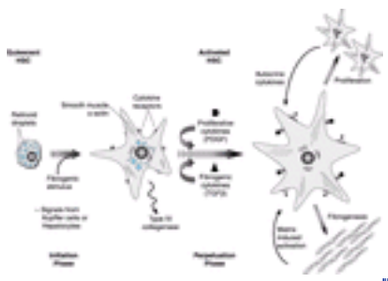

FIGURE 27-6. Hepatic stellate cell activation.

The stimuli that are responsible for HSC activation probably include signals derived from hepatocytes, Kupffer cells, and sinusoidal endothelial cells. The Kupffer cell plays a critical role in the development of liver fibrosis. Depletion of the Kupffer cell, the resident macrophage in the liver, with gadolinium chloride significantly reduces fibrosis in experimental animal models of fibrosis. [131] Endotoxin may be a key factor in the activation of the Kupffer cell. [132] Once activated, the Kupffer cell generates significant levels of ROIs. These molecules are believed to contribute to HSC activation either directly or through the generation of lipid peroxidation products. Antioxidants block HSC activation in oxidative stress conditions associated with high levels of ROIs. [133] TGF-ß, produced by activated Kupffer cells and activated HSCs, amplifies the fibrogenic response of the HSC. TGF-ß receptors on the cell surface are expressed by activated HSCs but not quiescent HSCs. [134] Autocrine stimulation of TGFß expression enhances activated HSC-mediated fibrogenesis. Expression of a fibronectin splice isoform that is expressed during early liver injury primarily by the sinusoidal endothelial cell can stimulate HSC activation. [73]

Two major events result in the HSC becoming a major contributor in liver fibrosis. These are the development of an active fibrogenic state and the amplification of the activated HSC population. Fibrogenesis is associated with an increased production of type I collagen as well as other ECM proteins and an increased production of TIMPs. Together, these changes in gene expression result in an imbalance of ECM biosynthesis, with a net increase in matrix deposition. The increased synthesis of type I collagen following HSC activation is caused by an increase in the transcriptional rate of the collagen genes, a stabilization of the collagen mRNA, and increased translation of the collagen mRNA. [9], [13], [14], [135] TGF-ß is a potent fibrogenic cytokine for the activated HSC. The activated HSC secretes TGF-ß and expresses TGF-ß receptors on the cell surface. This results in an autoregulatory loop to amplify matrix deposition. TGF-ß also increases the synthesis of other ECM constituents and decreases MMP expression. [120]

Amplification of the HSC population represents another major event in HSC activation that significantly contributes to the fibrogenic response. PDGF, the most potent proliferative cytokine known for the HSC, binds to a specific tyrosine kinase cell surface receptor. Binding of PDGF to its receptor initiates phosphorylation signaling cascades that leads to increased DNA synthesis and cell proliferation. PDGF activates the focal adhesion kinase (FAK)–phosphatidylinositol 3-kinase (PI3-K)–protein kinase B (AKT) signaling pathway transducing mitogenic signals for the cell and is critical for cellular chemotaxis. [136] PDGF also activates the ras-raf-MEK-ERK and MAPK-JNK signaling pathways transducing mitogenic signals for the cell. [137] PDGF also increases the migration of activated HSCs. [136]

Integrins also can activate the proliferative signaling pathway. The activated HSC responds to additional extracellular signals mediated through cell-matrix interactions. Depending on the composition of the ECM, different signals are transmitted into the cell that differentially regulate gene expression. Isolated HSCs grown on a basement membrane–like matrix, Matrigel, become activated at a much slower rate than cells grown on a type I collagen matrix or a plastic surface. [3] HSCs when grown on plastic increase collagen production and decrease collagenase production. [74], [138], [139]

The status of the extracellular environment is monitored by integrins, transmembrane proteins that associate with focal adhesion complexes. HSCs express a $_1$ß $_1$- and a $_2$ß $_1$-integrins on their cell surface. [28] These proteins sense the status of the extracellular environment and initiate signaling through the focal adhesion complex, a group of proteins that include FAK. Integrin binding to ECM components leads to integrin clustering, resulting in the activation of the focal adhesion complex that involves phosphorylation of FAK as well as other focal adhesion–associated proteins. Binding of a $_1$ß $_1$ to types I, III, and IV collagen activates the extracellular signal–regulated kinases 1 and 2 (ERK1, ERK2), which down-regulates type I collagen gene expression. [140] Another collagen receptor, discoidin domain receptor 2, is also expressed by activated HSCs. [141]

A key question surrounding the resolution of hepatic fibrosis is the fate of the amplified, activated HSC population. Recent evidence indicates that this cell population is cleared from the recovering liver by apoptosis. [78], [142] Nuclear factor-?B (NF-?B) expression protects the activated HSC from apoptosis. [143] Activation of PI3-K and AKT is also protective against apoptosis in several cell types.

MONITORING THE PROGRESSION OF FIBROSIS

The standard method to assess the degree of fibrosis and monitor its progression in patients is the liver biopsy. However, because this procedure is invasive and expensive and carries significant risk, noninvasive methods to assess the degree of fibrosis, especially the effects of antifibrotic treatment, are being explored. Several potential serum markers are being investigated, either singly or as a series, to monitor the level of fibrosis. These include collagen type IV, collagen type VI, collagen type XIV (undulin), the amino-terminal propeptide of collagen III (PIIINP), laminin, tenascin, hyaluronan, MMP-2, MMP-9 complexed with TIMP-1 (MMP-9/TIMP-1), and TIMP-1. Although not developed as a standard clinical test, increases in fibrosis correlate with serum levels of collagen IV, hyaluronan, TIMP-1, and PIIINP. [144]

ANTIFIBROTIC THERAPIES

Several stimuli are known to induce liver fibrosis (see Table 27-1). All these stimuli ultimately lead to activation of the HSC, resulting in a fibrogenic response. Therefore, therapeutic approaches to treat liver fibrosis directly may include (1) inhibiting HSC activation, (2) treating the consequences of HSC activation, and (3) inducing apoptosis of the activated HSCs (Table 27-2).

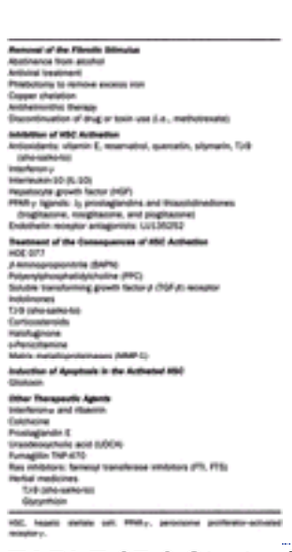

TABLE 27-2 Strategies for Antifibrotic Therapies

Because liver fibrosis is associated with HSC activation, a general approach to treat liver fibrosis may involve inhibiting activation of the HSC. Oxidative stress is associated with several animal models of liver fibrosis, including iron overload, CCl_4 intoxication, and excess ethanol administration. Oxidative injury causes injury to hepatocytes and other cells, resulting in the formation of ROIs that lead to the formation of lipid peroxidation products. Either ROIs or lipid peroxidation products can

interact with the HSC, resulting in cellular activation. Some antioxidants are efficient scavengers of ROIs. Vitamin E, the best-studied antioxidant, inhibits iron-induced fibrosis and decreases lipid peroxidation, HSC activation, and type I collagen gene expression. [145] Other antioxidants used to block oxidative stress include reservatrol, [146] quercetin, [146] silymarin, [147] and prostaglandin E_2. [148], [149] Sho-saiko-to (TJ-9), a traditional medicine composed of seven herbal components, has been used in Japan as a therapy for liver fibrosis. In vivo, TJ-9 has been shown to suppress fibrosis in animal models, and in cultured HSCs, TJ-9 inhibits cell activation and proliferation, and the oxidant burst. [150] Natural extracts are also being investigated for antifibrotic properties. In addition to TJ-9, glycyrrhizin, an extract of licorice root, has been shown to slow the progression of hepatitis C to hepatocellular carcinoma in animals and humans. [151] It is thought to inhibit I?B binding to the active NF-?B molecule.

Interferon-? inhibits HSC activation in culture and decreases ECM gene expression. In animal models of fibrosis, interferon-? decreases fibrosis induced by CCl_4 or schistosomes. IL-10 has also been shown to decrease fibrosis in patients with chronic hepatitis C who do not respond to interferon. Although no change in hepatitis C virus mRNA levels were noted, liver biopsy specimens showed decreased inflammation and fibrosis. [152] IL-10 probably decreases the proinflammatory helper T cell type 1 (Th1) response, leading to reduced fibrosis. Interestingly, a more severe fibrosis develops in IL-10 knock-out mice when they are challenged with a fibrogenic stimulus. Treatment of culture-activated HSCs with IL-10 also reduces type I collagen gene expression. [153] Colchicine and corticosteroids have been shown to inhibit fibrosis, probably by antiinflammatory mechanisms; however, the use of these drugs is controversial. [154]

Hepatocyte growth factor (HGF) is mitogenic for hepatocytes and other cell types. HGF is involved in hepatocyte development and regeneration and possesses anti-apoptotic activity in hepatocytes. In a rat animal model of liver cirrhosis induced by dimethylnitrosamine, administration of the human HGF gene into skeletal muscle induced high levels of circulating human and endogenous rat HGF. In addition, reduced levels of TGF-ß hepatocyte apoptosis, HSC activation, and fibrosis were noted, resulting in a complete resolution of cirrhosis and decreased mortality. [155]

Peroxisome proliferator–activated receptor-? (PPAR-?) represents another target for antifibrotic therapy aimed at inhibiting HSC activation. This nuclear receptor regulates gene expression associated with lipid metabolism. Although quiescent HSCs express PPAR-?, expression of PPAR-? decreases following HSC activation. Treatment of activated HSCs with PPAR-? ligands, prostaglandin J_2, and thiazolidinediones (troglitazone, rosiglitazone, and pioglitazone) restores PPAR-? expression and inhibits HSC proliferation and chemotaxis. A suppression of the activated phenotype, as assessed by decreased type I collagen and smooth muscle a-actin expression, occurs. [156], [157] and [158] Thus, PPAR-? ligands appear to inhibit HSC activation, leading to a decrease in the fibrogenic response.

Endothelin (ET) receptor antagonists may be another class of drugs with potential use as antifibrotic agents. Quiescent HSCs express primarily ET_A receptors, whereas activated HSCs express primarily ET_B receptors. [159] LU135252, an ET_A receptor antagonist, has been shown to decrease biliary fibrosis in the bile duct ligation animal model based on histological analysis, total liver collagen content, and mRNA expression for a $_1$(I) collagen, TIMP-1, and TIMP-2. [160], [161] and [162]

The complex biosynthetic pathway for type I collagen allows for multiple potential therapeutic targets (see Fig. 27-2). Prolyl 4-hydroxylase catalyzes the formation of hydroxyproline at every third amino acid, which is a specific posttranslational modification to the collagen chains that are required for the formation of a stable triple helical structure of collagen. Inhibitors of prolyl 4-hydroxylase include HOE077 and S4682. In animal models of fibrosis induced by CCl_4 treatment, a choline-deficient diet, or heterologous serum, simultaneous administration of HOE077 inhibited hydroxyproline content and histological liver fibrosis. HOE077 also inhibits stellate cell activation in culture [163] and in vivo. [163], [164] and [165] This may be the predominant mechanism for its antifibrotic effect. The potent and irreversible prolyl 4-hydroxylase inhibitor S4682 decreases hepatic fibrosis in the CCl_4 rat model, [166] with a concomitant decrease in morbidity, documented by improved liver function parameters and decreased ascites. Lysyl oxidase is responsible for cross-linking the secreted collagen triple helices in the extracellular space to form the characteristic collagen fibrils. This cross-linking protects the deposited collagen from nonspecific protease degradation. Expression of lysyl oxidase increases in cirrhosis. [167] A potent inhibitor of lysyl oxidase, ß-aminopropionitrile (BAPN), decreases the tensile strength of connective tissues; however, this drug is associated with skeletal deformities and aneurysms.

Polyenylphosphatidylcholine (PPC) can block collagen production and increase collagenase activity in cultured HSCs. [168] In vivo, PPC inhibits fibrosis in ethanol-induced fibrosis in baboons and in CCl_4-treated rats. [154] Because of the encouraging in vivo data, clinical trials are in progress. Halofuginone has also been shown to inhibit collagen production in thioacetamide-induced liver fibrosis in rats and may inhibit HSC activation. [169]

Activated HSCs are responsive to the fibrogenic cytokine TGF-ß and to the proliferative cytokine PDGF. A soluble TGF-ß receptor has been developed that consists of the extracellular portion of the TGF-ß type II receptor linked to human immunoglobulin G. Administration of the soluble TGF-ß receptor has been shown to significantly reduce experimentally induced liver fibrosis. [170] A group of compounds, indolinones, have been used to target the PDGF receptor. These compounds bind to the catalytic site of the PDGF receptor kinase, preventing downstream signaling. [171] A semisynthetic analog of fumagillin, TNP-470, inhibits the progression of fibrosis in both CCl_4- and dimethylnitrosamine-induced liver fibrosis by blocking cell cycle progression, thereby inhibiting HSC proliferation. [172]

The ECM is degraded by the MMP family of proteins, which have specific extracellular proteins as their substrates. Overexpression of MMP-1 (interstitial collagenase) can reduce collagen deposition following thioacetamide-induced fibrosis in the rat and thus represents another avenue for antifibrotic therapy. [173]

A final approach to treat liver fibrosis is to induce apoptosis in the HSC. In theory, this would lead to the reduction in the number of fibrogenic cells in the liver and consequently reduce ECM deposition. After a fibrogenic stimulus is removed in experimental cirrhosis, activated HSCs undergo apoptosis, not reversion to the quiescent phenotype. It has been shown that NF-?B, an inducible transcription factor, is protective against apoptosis in the HSC. [143] Therefore, blocking NF-?B activity should induce apoptosis in this cell. Accordingly, gliotoxin has been shown to inhibit NF-?B activity and induces apoptosis in the HSC. [174]

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CHAPTER 28

Rebecca Taub

HEPATIC REGENERATION

The liver exhibits unusual properties of regeneration after partial hepatectomy or toxic injury. This evolutionarily conserved response is a logical adaptive response of organisms because the liver is the major detoxifying organ of the body and first in line to be injured by ingested toxins. Partial hepatectomy, the model that most clearly demonstrates the regenerative capacity of the liver, was first described by Higgins and Anderson in 1931. [1] It has been noted that after a partial hepatectomy in which two thirds of a rodent liver, including the left lateral and medial lobes, is removed intact, the remnant liver enlarges until the mass of the liver is restored, whereupon the process stops. [2], [3], [4], [5], [6] and [7] The mass of liver is reconstituted within 7 days, most recovery occurring within 3 days, whereupon the cells again become quiescent. Recovery from toxic liver damage and recovery from hepatitis require similar types of hepatic regeneration, the major difference from hepatectomy being that these insults cause hepatic necrosis and apoptosis. Central questions about the process of liver regeneration remain, including the following (1) What signals trigger the early events in regeneration? (2) How are the architecture and function of the liver retained during regeneration? and (3) What signals are responsible for turning off the growth response once the mass of the liver is reconstituted? Using molecular-cellular approaches and genetic animal models, investigators have begun to dissect the critical factors and identify the important regulatory proteins that control this process. [8]

Many growth factors, including hepatocyte growth factor (HGF), epidermal growth factor (EGF), transforming growth factors (TGFs), insulin, glucagon, and cytokines (tumor necrosis factor [TNF-a], interleukin-1 [IL-1], and interleukin-6 [IL-6]), have been implicated in regulating this process, but the mechanisms remain poorly understood. Thus, both cytokine-dependent and -independent pathways are critical for liver regeneration (Fig. 28-1).

FIGURE 28-1. Model for growth factor pathways postulated to regulate liver regeneration after injury or partial hepatectomy. Proposed model of growth factor and cytokine activation of transcriptional cascade and DNA synthesis during liver regeneration. After partial hepatectomy, either gut-derived cytokines or cytokines released de novo activate hepatic nonparenchymal cells, resulting in increased production of tumor necrosis factor-a (TNF-a), interleukin-1 (IL-1), and IL-6. Cytokines and growth factors are responsible for the activation of nuclear factor-?B (NF-?B), signal transducer and activator of transcription-3 (STAT3), and perhaps CCAAT/enhancer-binding protein-ß (C/EBP-ß) and other factors in remnant hepatocytes. As a result of the activation of these hepatocyte transcription factors, the primary growth-response program (immediate-early genes, glucose regulation) is initiated, finally leading to DNA synthesis.

Liver regeneration plays an important role in several clinical settings. For the most part, these settings involve instances in which liver cells have actually been damaged. Animal models in which hepatocytes are directly damaged (galactosamine, carbon tetrachloride [CCl_4] treatment) result in regeneration of the liver, but in addition to mature undamaged hepatocytes, which participate in the process, progenitor cells are recruited. [9] In humans, understanding liver regeneration will help explain how the liver responds to toxic damage by agents such as alcohol and acetaminophen in overdose, or infections like hepatitis. Additionally, increasing numbers of liver transplants are being performed, including living related donor and small-for-size transplants. Successful transplants require at least some liver regeneration and repair, and it is essential to understand the biologic and molecular bases for liver cell growth.

BASIC CHARACTERISTICS OF LIVER REGENERATION

The most commonly studied model of liver regeneration is the two-thirds partial hepatectomy model in rodents. In the partial hepatectomy model of liver regeneration, remnant liver cells are not injured because there is no wound or site of injury. The word *regeneration* is a misnomer because the lobes of the liver that are removed do not grow back, unlike the limbs in amphibian models. Instead, a hyperplastic response occurs that involves the replication of virtually all the cells in the remnant liver. Thus, in this model, liver regeneration does not require the recruitment of progenitor cells, but it involves replication of the mature functioning cells of the liver. Once the mass of the liver has been reestablished, proliferation stops. The process is compensatory because the size of the resultant liver is determined by the demands of the organism.

Cell division is rarely seen in hepatocytes in the normal adult liver, which are considered to be in the G_0 phase of the cell cycle. [2], [4] It has been shown that after partial hepatectomy, approximately 95% of hepatic cells that are normally quiescent rapidly reenter the cell cycle. In the rat liver, the rate of DNA synthesis in hepatocytes (S phase) begins to increase after about 12 hours and becomes maximal at about 24 hours. The induction of DNA synthesis occurs later in nonparenchymal liver cells. Subsequent cycles of DNA synthesis are decreased because complete restoration of liver mass requires only 1.6 cycles of DNA synthesis in all cells. In the mouse liver, the peak in DNA synthesis is later, 36 to 40 hours, and somewhat variable between strains. The onset of DNA synthesis is well synchronized and proceeds in a portal to central direction in hepatocytes. The incidence of mitosis (M phase) is lower than predicted based on the number of hepatocytes undergoing DNA synthesis. The general belief is that the ploidy of liver cells and percentage of binucleated cells change with rounds of DNA synthesis. [10] However, studies have shown that hepatocytes have incredible replicative function. In hepatocyte transplantation studies, only a few hepatocytes are required to restore the liver mass completely. [11], [12] The nonparenchymal liver cells, which constitute approximately 40% of liver cells, undergo DNA replication later: Kupffer cells, 48 hours; endothelial cells, 96 hours; biliary epithelial cells, 48 hours. Most of the increase in liver mass is seen by 3 days, and mass restoration is complete in 5 to 7 days. [13]

Liver regeneration following massive hepatocyte necrosis or apoptosis induced by hepatic toxins such as CCl_4 or Fas ligand also involves the proliferation of hepatocytes, but the cell cycle response is not as synchronized. As would be expected, dramatic changes in liver architecture take place during liver regeneration, whether after partial hepatectomy or liver necrosis. Induction of novel forms of fibronectin, cell adhesion proteins, and other basement membrane proteins is observed. Changes in gap junctions occur transiently, and re-formation of the normal architecture occurs after the restoration of mass. Little is known about the regulation of the complex process of reorganization of liver architecture.

Replicative Potential of Hepatocytes

Partial hepatectomy induces replication of 95% of the hepatocytes in the liver. However, only one to two rounds of replication are required for complete restoration of liver mass. These studies do not explore the full replicative potential of hepatocytes. Studies in which hepatocytes are serially transplanted into recipient damaged livers have clearly demonstrated that at least some populations of hepatocytes have a very high replication capacity, not unlike that of stem cells in other tissues. Original studies in urokinase plasminogen activator transgenic mice estimated that repopulation of the liver could be achieved by a small number of transplanted hepatocytes undergoing 12 to 18 rounds of replication. [11], [14] Subsequently, in another liver injury model caused by tyrosinemia, it was found that serial transplantations of hepatocytes resulted in at least 69 cell doublings, a massive expansion of cells equivalent to that of hematopoietic precursor cells. [12]

Apparently, the replicative capacity of hepatocytes is in part regulated by the ability of hepatocyte chromosomal telomeres to remain in a replication-competent state. Mice with mutations in the telomerase RNA (mTR) gene as a consequence of gene knock-out are normal. [15] However, after six generations, telomerase dysfunction occurs and affects highly proliferative tissues such as bone marrow and gut. Liver development is normal in these mice. When crossed with the Alb-UPA transgene that causes increased hepatocyte turnover, late generation mTR-/- livers fail to provide sufficient replicative hepatocytes to overcome the high hepatocyte turnover. Increased hepatocyte apoptosis is also observed. After partial hepatectomy, mTR-/- livers demonstrate delayed regeneration and reduced progression through mitosis because of the formation of aberrant mitotic spindles.

In response to certain types of injury, normal hepatocytes are unable to replicate. In cases in which agents such as dipin, retrorsine, or galactosamine have diminished the replicative capacity of most normal hepatocytes, a population of cells termed *oval cells* that are found in ductular regions proliferate to replace the hepatic parenchyma. [16] Studies suggest that even these oval cells may be derived from hepatocytes that have somehow escaped the injury. [17] Other liver repopulation and transplantation studies suggest that bone marrow stem cells may have the capacity to differentiate along hepatocyte lineages. [18] It is not clear whether transition from a bone marrow precursor into a hepatocyte is a rare event or the means by which the liver replenishes its hepatocyte pool in the face of certain types of injury.

MECHANISMS OF LIVER REGENERATION

Experiments performed by Moolten and Bucher [19] provided the first evidence that circulating growth factors are present in the serum of hepatectomized rats that induce hepatocyte replication in parabiosed nonhepatectomized animals. Much of the early work into the initiating signals of liver regeneration focused on isolated hepatocytes in which hepatocyte growth factors were first identified. These studies led to the identification of several potential hepatocyte growth factors, such as HGF and TGF, and antiproliferative factors, such as TGF-ß, that might have important roles in regulating hepatocyte growth during liver regeneration. However, it was difficult to prove that any of these factors had a critical role in liver regeneration itself. Molecular studies involving the analysis of gene expression cascades in the regenerating liver began to provide insights into signaling pathways that are rapidly activated in the remnant liver after hepatectomy. Specific transcription factors, such as nuclear factor-?B (NF-?B), signal transducer and activator of transcription-3 (STAT3), AP-1, and others, were shown to be activated in remnant hepatocytes rapidly, within minutes after a partial hepatectomy. The finding of activated transcription factors provided clues to the presence of cytokine-dependent (TNF-a, IL-6) and -independent (HGF, TGF, and unknown) pathways that might be important regulators of the regenerative response. [3], [6], [7] However, which of the many signals and induced gene products are critical for liver regeneration is not apparent from the large array of activated gene products.

Regeneration studies performed in mice harboring gene knock-outs have highlighted the importance of specific genes (Table 28-1). [8], [20], [21], [22], [23], [24], [25] and [26] Unfortunately, knock-out of some of the genes of greatest interest resulted in embryonic lethality. For example, HGF, c-Jun, and NF-?B/p65 are all required for normal liver development, and therefore studies of regeneration in these models are not possible. Nonetheless, the finding of apoptosis in the developing liver in these gene knock-outs provides evidence for the critical importance of these proteins in the liver. In the case of HGF, this is particularly frustrating because HGF has long been felt to be a critical regulator of liver regeneration. [2] It has been difficult to demonstrate that HGF is active during the initial phases of regeneration. Studies indicate that treatment with HGF promotes regeneration and hepatocyte proliferation and that it reduces hepatic apoptosis and fibrosis and CCl_4-mediated injury. [27], [28] Ultimately, other models in which HGF or Met receptors are conditionally eliminated from the liver after birth will allow for an assessment of the requirement for HGF during regeneration.

Gene deletions in mice that result in either abnormal liver development or defective liver regeneration:

Defective Liver Development
HGF
p65/TNF-a-ß
c-Jun
XBP-1
Defective Liver Regeneration
IL-6
TNFR1
iNOS
CREM
C/EBP-ß
Keratin-8
Plasminogen
Telomerase

C/EBP, CCAAT/enhancer-binding protein; CREM, cAMP regulatory element; HGF, hepatocyte growth factor; IL-6, interleukin-6; iNOS, inducible nitric oxide synthase; NF-a-B, nuclear factor-a-B; TNF, tumor necrosis factor; TNFR1, tumor necrosis factor receptor type 1.

TABLE 28-1 Mouse Genetics: Defining Molecular Pathways in Liver Development and Regeneration

Several of the gene knock-out models showing impaired liver regeneration involve cytokine-dependent pathways. For example, it appears that TNF-a is required for the induction of IL-6 after partial hepatectomy, and inducible nitric oxide synthase (iNOS), which helps prevent liver injury after hepatectomy, is a possible target gene of these cytokines. [20], [21], [26] TNF-a, IL-6, and normal iNOS regulation are all required for normal liver regeneration. The availability of mutant mice that are deficient in specific subunits of NF-?B makes it possible to directly test the significance of NF-?B during liver regeneration. It has been shown that mice that are missing the NF-?B/p65 gene through gene targeting die during embryogenesis, apparently because of the sudden death of hepatocytes at approximately 15 days of gestation. [29] The regenerative response of p50-deficient mice is largely normal because of a compensatory increase in the level of the NF-?B/p65 subunit as a consequence of reduced levels of the inhibitor I?B-a. [30]

CCAAT/enhancer-binding protein-ß (C/EBP-ß), a leucine zipper transcriptional factor that is considered to be a cytokine-regulated factor, is also required for normal liver regeneration. [22] How the activity of C/EBP-ß is regulated by extracellular signals after hepatectomy is not known. There is some support for the hypothesis that the relative levels of pro-proliferative forms of C/EBP-ß are important in supporting liver growth and balance antiproliferative forms and C/EBP-a, an antiproliferative factor. Interestingly, as described below, C/EBP-ß regulates a set of genes after hepatectomy distinct from those regulated by IL-6. [3] By examining which gene expression pathways are altered in each of the knock-out mouse models, it will ultimately be possible to determine the development of the various signal transduction pathways that lead to liver regeneration.

Induction of Primary Growth-Response Genes and Latent Transcription Regulatory Proteins

One way to address the central questions about the process of liver regeneration is by determining the critical molecules that regulate this process. Therefore, a number of studies were performed to determine what genes are expressed de novo in the remnant liver at the onset of liver regeneration. [31], [32] The cells in the normal liver are in the quiescent, nonproliferative G_0 phase of the cell cycle (see Fig. 28-1). Within minutes after a partial hepatectomy, in response to signals that initiate liver regeneration, these cells reenter the cell cycle in the G_1 phase. When this happens, signals transmitted to the nuclei of the liver cells result in transcriptional activation of primary growth-response genes, or “immediate-early genes.” [33], [34], [35], [36] and [37] Many of these genes (e.g., fos, myc, jun) encode proteins that have been identified as “protooncogenes” because somatic mutations or overexpression of these genes can lead to malignant transformation. In an unmutated form, these proteins are important for normal cell growth and cell cycle progression. Because many immediate-early genes encode proteins that are important for cell growth control, one can imagine how expression of the genes results in a cascade of events leading to DNA synthesis (S phase) and cellular division.

Several years ago, it was shown that more than 70 growth-response genes are activated in the remnant liver during the early stages of liver regeneration, [31], [38] and

that number has been expanded during subsequent years to more than 100. [39] In most cases, these genes are not expressed in the normal liver. However, within minutes to hours of a partial hepatectomy, the level of their expression in the remnant liver increases dramatically. Examination of the temporal course of expression of genes whose expression is activated during liver regeneration defined the boundaries between proliferation and cessation of growth (Fig. 28-2). In these studies, the genes themselves become “reporters” because the fact that they are being expressed indicates that growth-regulatory signals are being transmitted within the remnant liver cells. During regeneration, the major growth period of the liver ends at 60 to 72 hours after hepatectomy, when the liver has already more than doubled in size. There are two major patterns of growth-regulated gene expression. In the rat regenerating liver, *growth-regulated genes* demonstrate elevated expression throughout the entire growth phase, and expression returns to normal after about 3 days. *Cell cycle–regulated genes* show a sharp peak of expression that coincides with the G_1 phase of the hepatocyte cell cycle, including the first round of replication, and a second smaller round of replication that occurs 48 hours after the partial hepatectomy.

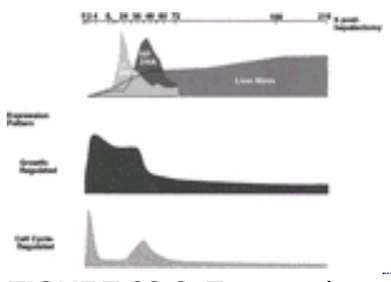

FIGURE 28-2. Temporal course and representative patterns of regulated gene expression during rat liver regeneration. The pattern of gene expression is indicated for growth-regulated genes (e.g., ß-actin) and cell cycle–regulated genes (e.g., insulin-like growth factor binding protein-1 [*IGFBP-1*]). Expression after the growth phase (CCAAT/enhancer-binding protein-a [C/EBP-a]) occurs when the gene is down-regulated during the period of maximal growth (not shown). Hours after partial hepatectomy and patterns of DNA synthesis in hepatocytes (*H*) and nonparenchymal cells (*NP*) and reaccumulation of liver mass [30] are indicated.

Not all these genes are immediate-early genes. A number of genes are first expressed a few hours after the initial induction of immediate-early genes. Termed *delayed-early genes,* these genes are regulated at a transcriptional or posttransciptional level by products of the immediate-early gene. In this way, a cell cycle regulatory cascade of gene and protein expression allows cells to progress through the G_1 phase of the cell cycle. Ultimately, changes in levels of cyclins and their regulatory kinases, the cyclin-dependent kinases (CDKs), by both transcriptional and posttranscriptional regulation allow for transition through the late phases of G_1 (D cyclins, CDK4, CDK6) into the S phase (cyclin E, CDK2) and later phases of the cell cycle (cyclin A, cyclin B). [40] Cyclin D/CDK4 complexes are important for progression beyond a late G_1 restriction in part because they phosphorylate retinoblastoma (RB) and E2F factors. The cyclin-associated inhibitors p21 and p27 are likely to be important for both progression through the cell cycle and inhibition of unregulated cell proliferation.

Rapid Activation of Latent Transcription Factors

In addition to the contribution of the liver-specific transcription factors in modulating liver function and growth in the regenerating liver, several transcription factors are activated de novo in the remnant liver. Of particular interest are those factors that are latent in normal liver cells and activated within minutes after the partial hepatectomy by the earliest signals that trigger regeneration. In other words, these proteins or transcription factors are activated in the first few minutes after hepatectomy by posttranslational modifications. Unlike immediate-early genes or primary growth-response genes, they are not activated by new mRNA synthesis and translation. In fact, these transcription factors actually turn on the transcription of the primary growth-response genes. Investigators reasoned that understanding the signal transduction pathways responsible for the activation of such transcription factors would provide insight into what growth factor and cytokine signals trigger liver regeneration.

For example, two different DNA binding complexes, NF-?B and STAT3, are present in an inactive form in the normal liver. [41], [42], [43] and [44] Both are downstream of cytokine signaling pathways. They are activated within minutes to hours after hepatectomy in the remnant liver cells, thus enabling these factors to turn on the expression of the primary growth-response genes. Because these factors are activated by the earliest signals that trigger liver regeneration, understanding the mechanism by which they are activated has provided clues to the initiating signals. These factors are not turned on in all growing cells, and therefore they could account for some liver-specific aspects of growth in the regenerating liver.

NF-?B is a transcription factor complex that forms when a 50-kd protein subunit and a 65-kd protein subunit unite to form a transcriptional complex in the nucleus of the cell. [45] Normally, the p65 subunit resides in the cytoplasm of hepatocytes in an inactive form bound to its inhibitor I?B-a (Fig. 28-3). After a partial hepatectomy, a signal that may be generated by a variety of growth factors and cytokines, including TNF-a, IL-1, and others, results in the dissociation of I?B-a from p65. This occurs because the I?B kinase (IKK) complex phosphorylates I?B-a, thereby leading to its degradation and release from p65. [16] Phosphorylation of p65 also occurs, and the net effect is that the p65 is free to move into the nucleus of cells, where it can complex with the p50 subunit, forming an active NF-?B transcription complex. This leads to the transcriptional activation of genes that are controlled by a ?B DNA binding site in their promoter. During liver regeneration, the signal leading to NF-?B activation occurs within a few minutes after the partial hepatectomy and is gone by 1 to 2 hours, when NF-?B levels are again virtually undetectable. In the regenerating liver, activation of NF-?B occurs in hepatocytes, but it may also occur in nonparenchymal cells, such as Kupffer and endothelial cells, that also have receptors for various cytokines.

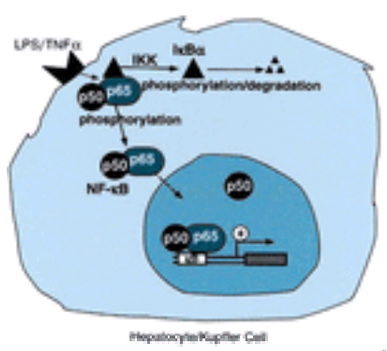

FIGURE 28-3. Model of nuclear factor-?B (NF-?B) activation in the hepatocyte and Kupffer cell after hepatectomy. Several different cytokines (transforming growth factor-a [TNF-a]) and LPS released into the portal circulation from the intestine signal via their extracellular receptor, resulting in phosphorylation by I?B kinase complex (IKK) and degradation and dissociation of I?B-a from cytoplasmic p65/p50. Subsequently, free, newly phosphorylated p65/p50 moves into the nucleus, where it is able transactivate target genes.

Another class of transcription factors that is rapidly activated in the remnant liver after a partial hepatectomy is the STAT family. These factors were first discovered as part of the intracellular response to a variety of different extracellular signals mediated by interferons, cytokines, and growth factors. [46], [47] However, the STAT factor that is specifically activated in the regenerating liver is STAT3, which has been shown to be induced in liver cells by IL-6, LPS (an endotoxin, probably via IL-6 activation), and EGF (Fig. 28-4). [3], [48], [49] After one of these factors is bound to its receptor, a Janus tyrosine kinase (JAK) is activated. This kinase phosphorylates the STAT3 factor on a specific tyrosine residue, resulting in its nuclear translocation and the ability to transactivate target genes.

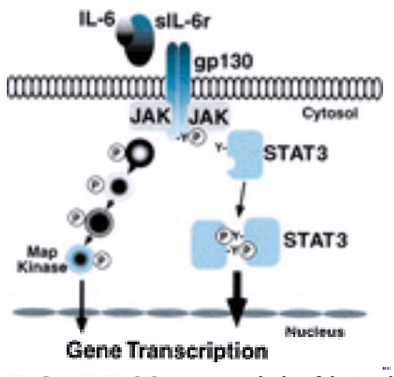

FIGURE 28-4. Model of interleukin-6 (IL-6) signaling. IL-6 binds to its soluble receptor complexed with the gp130 receptor. This results in the activation of a tyrosine kinase in the Janus kinase (JAK) family that phosphorylates cytoplasmic signal transducer and activator of transcription-3 (STAT3). Phosphorylated, dimerized STAT3 moves into the nucleus, where it is able to bind to and transactivate target gene promoters, resulting in increased gene transcription. IL-6 is also able to signal via the mitogen-activated protein kinase (MAPK) pathway.

During liver regeneration, the activation of STAT3 is quite dramatic, although later and more prolonged than NF-?B activation, showing peak activity between 1 and 3 hours and continued activity as late as 8 hours after hepatectomy. [20], [41] During liver regeneration, the major cytokine responsible for activating STAT3 is IL-6. The STAT3 response is specific for liver regeneration because little STAT3 activation is seen following sham surgery. STAT transcription factors are potent transcriptional activators that can both directly and indirectly lead to transcriptional activation of a large number of genes. Many of these genes, such as jun and fos, themselves encode transcription factors that make up part of the AP-1 complex. AP-1 activity is also induced early during liver regeneration, in part via new protein synthesis and in part via activation by early signals such as TNF-a. One may imagine that if STAT3 plays a role in the activation of these factors, it may be initiating a transcriptional cascade within the regenerating liver that ultimately leads to transition through G_1 and entry into S phase. Mutant mice that contain gene deletions for NF-?B, IL-6, and TNF receptors and conditional STAT3 knock-outs allow the significance of STAT3 and NF-?B activation during liver regeneration to be tested.

LIVER FUNCTION DURING LIVER REGENERATION

Interplay between Hepatic Transcription Factors and Growth-Induced Transcription Factors

The liver is a vital organ and must continue to function as its cells proliferate during liver regeneration. In most cells, such as the hematopoietic lineages, proliferation is not compatible with differentiated function. How the liver is able to function while undergoing regeneration is an important question in liver biology. By examining the immediate-early gene response in the regenerating liver, it was possible to find clues to how the liver is able to function as it regenerates. Although many immediate-early genes are expressed in common in the regenerating liver and other growing cells, approximately one third of the genes are expressed at a high level or specifically in growing liver cells. Both hepatocytes and the nonparenchymal cells of the remnant liver express most of these genes, but the expression of a subset of genes is limited to hepatocytes. [38] Several of these liver-restricted immediate-early genes encode proteins that are involved in the gluconeogenic response of the liver.

The induction of gluconeogenic genes represents an adaptive response of the liver whereby the remnant liver that comprises only one third of the original mass compensates by producing enough glucose for the whole organism. Liver-specific transcription factors have an important role in determining the level of liver-specific functions, including glucose production. They act by regulating the expression of genes encoding liver-specific enzymes (e.g., metabolic enzymes) and liver-specific secreted proteins (e.g., albumin). The adaptive response of the liver during regeneration that allows for the maintenance of metabolic homeostasis is accomplished by an interplay between growth-induced transcription factors and preexisting liver-specific transcription factors that regulate the differentiated functions of the hepatocyte.

After hepatic injury or stress, gluconeogenic and acute phase response genes are rapidly up-regulated to restore metabolic homeostasis and limit tissue damage. The gluconeogenic genes encoding phosphoenol pyruvate carboxykinase (PEPCK) and glucose 6-phosphatase (G6Pase) are induced, [50], [51] while, as others have noted, [52] the genes encoding proteins that oppose gluconeogenesis (glucokinase) are down-regulated during liver regeneration. This parallels acute hormonal changes that occur in the regenerating liver when insulin levels rapidly fall and glucagon levels increase. [53] Glucokinase is induced by insulin and down-regulated by glucagon; PEPCK and G6Pase show the opposite regulation.

In addition, regulation of the liver-restricted insulin-like growth factor binding protein-1 (IGFBP-1) gene is dramatically altered by changes in the metabolic state and hepatectomy in a manner analogous to that of the gluconeogenic genes. IGFBP-1 mRNA is induced more than 100-fold in the regenerating liver and also induced at a high level during fasting and insulin deficiency. [54] The IGFBP-1 gene thus provides an appropriate reporter to assess the transcriptional milieu that leads to activation of gluconeogenic genes during regeneration. Normally, the IGFBP-1 gene in the liver is regulated in the basal state by hepatic nuclear factor-1 (HNF-1), a homeodomain containing hepatic transcription factor that plays a prominent role in the regulation of many hepatic genes. The level of HNF-1 does not change appreciably during liver regeneration. During liver regeneration, IL-6 is in part responsible for induction of the IGFBP-1 gene. [55] This up-regulation is accomplished by physical interactions between growth-induced IL-6–regulated transcription factors, STAT3 and AP-1 (c-Fos, c-Jun), and the resident hepatic transcription factor, HNF-1. Activation by this complex of transcription factors is mediated through the HNF-1 DNA binding site immediately upstream of the IGFBP-1 promoter site (Fig. 28-5). [55] Similar regulation is observed when G6Pase and a-fibrinogen promoters are used, indicating that HNF-1/IL-6/STAT3/AP-1–mediated transactivation of hepatic gene expression is a general phenomenon after liver injury. These results demonstrate that the two classes of transcription factors, growth-induced (STAT3 and AP-1) and tissue-specific (HNF-1), can interact as an adaptive response to liver injury to amplify the expression of hepatic genes important for the homeostatic response during organ repair. In this way, the liver is able to maintain metabolic function despite the loss of two thirds of its functional mass.

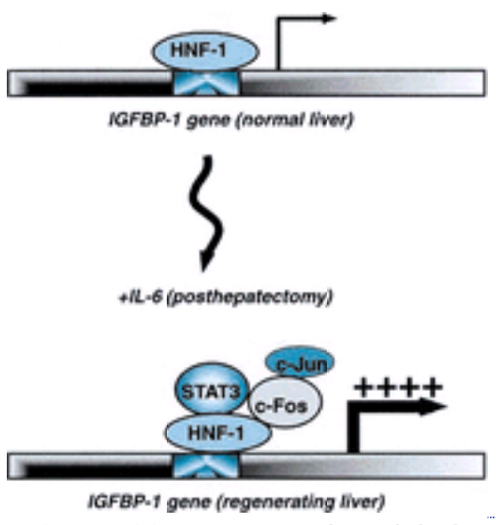

FIGURE 28-5. Proposed model of cooperative interaction between a growth-induced (c-Fos, c-Jun, signal transducer and activator of transcription-3 [STAT3]) and constitutive (hepatocyte nuclear factor-1 [HNF-1]) hepatic factor to up-regulate the level of a liver-specific target gene (insulin-like growth factor binding protein-1 [IGFBP-1]) after hepatectomy.

Regulation of Liver-Specific Transcription Factors

The levels of many hepatic transcription factors remain unchanged after partial hepatectomy, and this continued expression provides the regenerating liver with some of its maintained functional capacity. [56], [57] However, the liver-restricted transcription factors C/EBP-a (CCAAT/enhancer-binding protein) and C/EBP-ß show inverse

regulation during liver regeneration, so that the less antiproliferative form C/EBP-ß is relatively up-regulated and antiproliferative C/EBP-a is down-regulated during the phase of rapid liver growth. C/EBPs (including C/EBP-a and C/EBP-ß) are leucine zipper transcription factors that bind as homodimers or heterodimers to the recognized DNA sequence in the promoter of some liver genes, such as albumin, PEPCK, and many others. [58], [59], [60], [61], [62] and [63] The importance of C/EBP-a in controlling gluconeogenic genes was established by the finding that mice that are genetically missing the gene for C/EBP-a die at birth of hypoglycemia because of a lack of expression of PEPCK, G6Pase, and perhaps other metabolic enzymes. [64], [65] This finding directly connects C/EBP-a to the regulation of these genes. However, the C/EBPs have also been shown to have an antiproliferative effect, particularly C/EBP-a. [6], [22] Thus, during liver regeneration, an apparent paradox occurs because of the simultaneous need for gluconeogenic gene expression positively controlled by C/EBP-a and proliferation that is opposed by C/EBP-a. How is this accomplished?

During liver regeneration, an inverse regulation of C/EBP-a and C/EBP-ß proteins results in up to a sevenfold increase in the a/ß DNA binding ratio between 3 and 24 hours after hepatectomy that may have an important impact on target gene regulation. [62] However, total C/EBP binding activity in the remnant liver cell nuclei remains relatively constant during the 7-day period after hepatectomy. Both differentiation-specific liver functions and proliferation coexist in the same liver cells. This is unusual because in most cell types, growth and differentiation are uncoupled and mutually exclusive. The persistent expression of C/EBP-a and C/EBP-ß isoforms predicts that C/EBP proteins contribute to the function of hepatocytes during physiological growth, and that significant amounts of these proteins do not inhibit the progression of hepatocytes into S phase of the cell cycle. The importance of the induction of C/EBP-ß during liver regeneration was further demonstrated by studies in which C/EBP-ß-/- mice were subjected to partial hepatectomy (see below); it was shown that C/EBP-ß is important for the regenerative response and the expression of some cell cycle regulatory genes and proteins.

CYTOKINE-DEPENDENT AND -INDEPENDENT PATHWAYS IN REGENERATION

Both NF-?B and STAT3 are frequently activated in cells by cytokines. Cytokines bind to their cellular receptors and generate intracellular signals that lead to transcription factor activation. [66] The finding that these transcription factors are induced during liver regeneration raised the interesting possibility that cytokines released after a partial hepatectomy either from the gut or from neighboring Kupffer and other liver nonparenchymal cells may act on hepatocytes to stimulate the activation of the latent NF-?B and STAT3 transcription factors. C/EBP-ß, which increases during liver regeneration, has also been linked to activation by cytokines. [67], [68]

Studies linking cytokines, particularly TNF-a, IL-1, and IL-6, to liver regeneration were relatively limited before studies in knock-out mice demonstrated a requirement for TNF-a and IL-6 in the regenerative response. [69], [70] Other studies suggested that circulating endotoxin LPS derived from the gut increases after a partial hepatectomy, and that animals that have been rendered germ free or that are deficient in the LPS response demonstrate blunted liver regeneration. [40] However, high levels of these cytokines, seen in CCl_4-induced liver damage or sepsis, may actually be detrimental to liver function and recovery. [71] The well-modulated levels of cytokines seen following partial hepatectomy are necessary for normal liver regeneration, and the excessive levels of cytokines seen in pathological processes may lead to liver damage. [72]

Normal liver regeneration after partial hepatectomy requires the IL-6 cytokine (see model, Fig. 28-1). [20] After partial hepatectomy, IL-6-/- livers exhibit impaired liver regeneration, characterized by liver necrosis and failure, a blunted DNA response in hepatocytes, and discrete G_1 phase abnormalities, including absence of STAT3 activation and selective abnormalities in gene expression. Treatment of IL-6-/- mice with a single preoperative dose of IL-6 returns STAT3 binding, gene expression, and hepatocyte proliferation to near normal and prevents liver damage, establishing IL-6 as a critical component of the regenerative response after partial hepatectomy. Most of the actions mediated by IL-6 occur within the first few hours after hepatectomy and lead to progression through the early to middle phases of G_1 and ultimately to DNA synthesis and mitosis. TNF-a signaling is also required for a normal proliferative response after hepatectomy, and this effect appears to be mediated largely by the ability of TNF-a to induce IL-6. [21] Specific cell cycle progression pathways and patterns of induced gene expression are dependent on IL-6, whereas others are normal in the absence of IL-6. [39]

To determine the percentage of immediate-early genes regulated by IL-6 after partial hepatectomy, the gene expression program in the IL-6+/+ and IL-6-/- livers at 2 hours after hepatectomy was examined with a cDNA array representing 588 highly regulated mouse genes. [39] Thirty six percent of the 103 immediate-early genes are induced differently in IL-6+/+ and in IL-6-/- livers, implying IL-6 regulation. IL-6 treatment of the IL-6-/- mice in the absence of hepatectomy induces a much smaller set of genes in the liver, suggesting that IL-6 cooperates with other hepatectomy-induced factors to activate the large number of genes. The mitogen-activated protein kinase (MAPK) pathway is activated in IL-6+/+ livers early during regeneration but is remarkably delayed in IL-6-/- livers. This pathway has been shown to be critical for cell growth in general and for liver regeneration specifically. [73] Defective liver regeneration may be explained by the large number of gene activation pathways altered in IL-6-/- livers and further supports the finding that IL-6 is necessary for normal liver regeneration. However, IL-6 is not a complete growth factor in that it does not stimulate much hepatic proliferation in the absence of hepatectomy. Therefore, other pathways must be activated in addition to the IL-6/TNF-a pathway for regeneration to proceed normally.

Both C/EBP-ß and IL-6 are involved in cytokine activation pathways during the acute phase response in the liver, and IL-6 is increased in the cytokine activation network in liver regeneration. As in IL-6-/- livers, regeneration is impaired in C/EBP-ß-/- livers, but the genes and pathways affected are distinct from those regulated by IL-6. [22] The mechanism by which C/EBP-ß is activated and induced during liver regeneration remains unknown. In some early studies, it was felt that IL-6 regulated the level of the C/EBP-ß leucine zipper transcription factor, and conversely, that C/EBP-ß was important for the induction of IL-6. Studies of C/EBP-ß-/- and IL-6-/- animals indicated no positive correlation between IL-6 and C/EBP-ß levels. In fact, IL-6 levels are elevated in C/EBP-ß-/- animals. [22] Moreover, some genes are induced relatively normally in both C/EBP-ß-/- and IL-6-/- livers after hepatectomy, further indicating the complexity of the regenerative response.

Both C/EBP-ß-/- and IL-6-/- mice show impaired responses after hepatectomy characterized by a DNA synthetic defect. The defect in IL-6-/- livers is more severe in that the animals exhibit increased morbidity and mortality and evidence of liver necrosis. Mass restitution occurs but is delayed in IL-6-/- animals that survive the surgery, whereas liver mass restitution is not impaired in C/EBP-ß-/- livers. Mass restitution largely occurs as a function of progression through the G_1 phase of the cell cycle and may not be different in livers that show primarily a defect in S phase entry, as in the C/EBP-ß-/- livers. [74] In IL-6-/- livers, all these defects can be corrected by a single dose of IL-6. On the other hand, treatment of C/EBP-ß-/- animals after hepatectomy with IL-6 actually worsens the outcome; the animals become ill. This result is compatible with the observation that C/EBP-ß-/- animals actually have elevated levels of IL-6, and the hypothesis that C/EBP-ß and IL-6 mediate their effects through largely nonoverlapping intracellular signals.

These differences are particularly well demonstrated by examining the genes that are expressed differently in IL-6-/- and C/EBP-ß-/- livers after hepatectomy. A number of the same genes have been examined in the two animal models. The only gene that shows reduced induction in both models is SAP, an acute phase gene that is known to be regulated by both IL-6 and C/EBP-ß. [75] For example, IL-6-/- livers show particular defects in genes encoding AP-1 DNA binding proteins (c-fos, JunB, LRF-1) and c-myc. These transcription factors are purported to be important for the regulation of cyclin D_1 expression, which is predictably abnormal in IL-6-/- livers. On the other hand, AP-1, c-myc, and cyclin D_1 genes are normally induced in C/EBP-ß-/- livers. Gluconeogenic genes are normally regulated in IL-6-/- livers but not in C/EBP-ß-/- livers, in which they are elevated to compensate for the hypoglycemia. The HGF gene has been shown to be regulated by both IL-6 and C/EBP-ß in vitro, [76] but normal induction of HGF mRNA is observed in both IL-6-/- and C/EBP-ß-/- livers. Unique among the genes that were studied is PRL-1, which shows normal induction in both C/EBP-ß-/- and IL-6-/- livers after hepatectomy.

Gene expression studies have helped determine which aspects of hepatic regeneration are independent of cytokine activation pathways involving IL-6, TNF-a, and C/EBP-ß. As indicated, C/EBP-ß regulation may be independent of these cytokines but still linked to a cytokine response. It was determined that PRL-1, a gene that encodes a nuclear protein tyrosine phosphatase, is induced equally well in both IL-6-/- and C/EBP-/- livers after partial hepatectomy. [3] The Egr-1 transcription factor is responsible in part for the activation of the PRL-1 gene. [77] Like STAT3 and NF-?B, Egr-1 is induced in the regenerating liver in the absence of de novo protein synthesis and reflects activation by a cytokine-independent pathway in liver regeneration. As yet, it is not clear which growth factor signal during liver regeneration is responsible for the activation of Egr-1 and genes that are regulated normally after hepatectomy in C/EBP-ß-/- and IL-6-/- livers. Further analysis of gene expression patterns in these and other gene knock-out mice with impaired liver regeneration will provide insight into regulatory pathways, actions of specific growth factors (HGF and TGF), and cross talk between cytokine-dependent and -independent pathways.

The relationship between normal liver proliferation after hepatectomy and the acute phase response is interesting. The same cytokines that may promote normal liver regeneration are also activated during the acute phase response. The acute phase response, a first line of defense against wounds and infection, results in high levels of some circulating proteins produced by the liver. [78], [79] Although an acute phase response occurs in the regenerating liver after hepatectomy, it is of relatively

low grade in comparison with the response induced by LPS or turpentine injection. [80] Although common intracellular pathways mediated by cytokines may be seen in both liver regeneration and the acute phase response, the end results are quite different. In liver regeneration, the response is mitogenesis; in the acute phase response, the production of acute phase reactants leads to wound healing.

Interleukin-6–Induced Liver Regeneration after Liver Injury: Reduced Hepatocyte Proliferation and Persistent Injury

In addition to hepatectomy, IL-6-/- livers have been shown to respond abnormally to a variety of liver injury models, including CCl_4, ischemia/reperfusion, fas activation, and bile duct ligation. [20], [81], [82], [83], [84], [85] and [86] As such, IL-6-/- mice provide an excellent model system for examining the relationship between liver regeneration, repair, and ultimately fibrogenesis. Several studies have examined the role of IL-6 in liver injury following ischemia/reperfusion. In one study, both hepatectomy and ischemia were simultaneously applied. It was found that hepatic ischemia in combination with hepatectomy significantly reduces the mitogenic response in mouse livers. [84] Treatment with IL-6 improves the mitogenic response in the face of ischemic injury back to the normal level. In warm ischemia/reperfusion models, IL-6 is protective against ischemic injury, and IL-6-/- livers show increased injury. In this model, TNF-a results in injury, as antibodies to TNF-a are able to reduce ischemic injury to a similar degree as administration of IL-6. [82] A program of gene expression, activation of STAT3 and NF-?B, and a high degree of hepatocyte proliferation similar to that in the hepatectomy model have been observed in transplanted rodent livers subjected to prolonged warm ischemia, [83] indicating that IL-6–dependent hepatic regeneration occurs following ischemic liver injury.

Studies supporting TNF-a as one of the factors that promotes liver regeneration indicate that one of the actions of TNF is to regulate the expression of IL-6. TNF receptor type 1 (TNFR1) knock-out mice have defects in liver regeneration very similar to those of IL-6 knock-outs, including major reductions in STAT3 activation and IL-6 mRNA levels. [21] These animals are deficient in the TNFR1, which appears to mediate most of the effects of TNF-a in the liver. The DNA synthetic response of TNFR1-/- livers is restored by treatment with IL-6, so that the need for TNF-a is bypassed. This therapeutic intervention could have important consequences because it has been shown that too much TNF-a is deleterious in the liver and contributes to liver injury. A notable difference between TNFR1-/- and IL-6-/- livers is that induction of NF-?B after hepatectomy is relatively normal in IL-6-/- but virtually absent in TNFR1-/- livers. This is presumably because TNF-a is directly responsible for activating NF-?B in the liver after hepatectomy. Because IL-6 is a known NF-?B target gene, the reduction of active NF-?B provides a mechanism by which IL-6 and STAT3 levels are reduced in TNFR1-/- livers after hepatectomy.

Although IL-6 and TNF appear to be beneficial and required for liver regeneration after a partial hepatectomy, the need for these factors has not been as clearly established after liver injury, a more common regenerative stimulus. A number of studies have suggested that TNF-a increases liver injury after toxic damage. For example, TNFR1-/- livers are less susceptible to liver failure and fulminant hepatitis after damage with LPS and galactosamine. [87] In addition, studies have shown that blocking TNF-a with anti–TNF-a antibodies reduces the level of hepatic injury after treatment with the toxin CCl_4. [71] Several of these cytokines, including both IL-6 and TNF-a, have been implicated as profibrogenic in chronic liver injury leading to cirrhosis. Studies indicate that TNFR1-/- livers regenerate less well than wild-type livers after CCl_4 treatment, but the effect on the degree of liver injury was not as clear. [88] TNF-a itself may increase the degree of liver injury because the level of TNF-a after toxic damage is in relative excess, and the amount of IL-6 produced is relatively deficient to maximally stimulate regeneration and repair.

IL-6 also mitigates acute toxic liver injury. [81] CCl_4 is a hepatotoxin that causes direct hepatocyte injury by altering the permeability of cellular, lysosomal, and mitochondrial membranes. [89] Highly reactive free radicals are also formed from the metabolism of CCl_4 by cytochrome P450 2E1 of the hepatocyte, causing centrizonal necrosis. CCl_4 causes not only primary liver necrosis but also hepatocyte apoptosis. [90], [91] Following acute CCl_4 treatment, increased hepatocellular injury and defective regeneration with significant blunting of STAT3 and NF-?B activation and reduced hepatocyte DNA synthetic and mitotic responses are seen in IL-6-/- mice. [81] CCl_4 treatment, unlike partial hepatectomy, is followed by increased hepatocyte apoptosis in IL-6-/- livers. Pretreatment with IL-6 before CCl_4 reduces acute CCl_4 injury and apoptosis and accelerates regeneration in both IL-6+/+ and IL-6-/- livers.

Two major defects are seen in IL-6-/- livers subjected to a single dose of CCl_4. First, despite more extensive injury, IL-6-/- livers show reduced hepatocyte proliferation. Unlike the hepatectomy model, in which additional IL-6 given to wild-type mice has little impact on the course of regeneration, treatment with IL-6 clearly accelerates the proliferative response in both IL-6+/+ and IL-6-/- livers. Peak entry into S phase occurs at 36 rather than at 48 hours. This response further defines the role of IL-6 as a cell cycle progression factor. The second major finding in IL-6-/- livers is a dramatic increase in the degree of liver injury following CCl_4 toxicity (roughly two-thirds greater than in the wild type). Treatment with IL-6 has a profound impact on both IL-6-/- and IL-6+/+ livers in reducing the amount of CCl_4-induced injury. IL-6 treatment also reduces the number of apoptotic hepatocytes in both IL-6-/- and IL-6+/+ livers.

In another model of liver injury associated with high levels of TNF-a, the concanavalin A T-cell activation liver model, antibodies to TNF-a reduced injury, as did injection of recombinant IL-6. [92] In addition to decreasing CCl_4-induced liver injury, IL-6 decreases hepatocyte apoptosis, a component of CCl_4-induced liver injury. In this model, it is not clear whether reduction in apoptosis is the consequence of an overall reduction in injury or whether IL-6 exerts a primary anti-apoptotic effect that serves to rescue hepatocytes. Fas ligation, a purer model of hepatocyte apoptosis than CCl_4 toxicity, provides more insight into mechanisms by which IL-6 reduces hepatocyte apoptosis. Il-6-/- mice are more susceptible to Fas-mediated death than wild-type animals. [93] The direct anti-apoptotic effects of IL-6 are demonstrated in vitro as IL-6 decreases Fas-mediated apoptosis in both IL-6-/- and IL-6+/+ primary hepatocyte cultures, suggesting that IL-6-/- hepatocytes have a preexisting defect in anti-apoptotic pathways. After Fas activation, IL-6-/- livers show evidence of both proximal and distal alterations in the apoptotic pathways, including elevated caspase 8 and caspase 3 activation–associated fragments and loss of cytochrome *c* staining. IL-6-/- livers show reduced preexisting protein expression of the anti-apoptotic factors Bcl-2 and Bcl-xL, as well as more rapid degradation of FLIP following Fas treatment that appears to be posttranscriptionally regulated (Fig. 28-6).

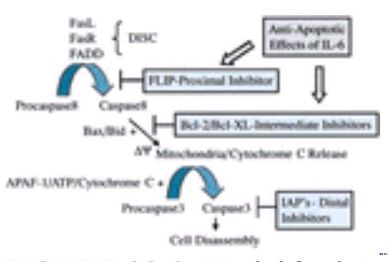

FIGURE 28-6. Model for interleukin-6 (IL-6) protection against Fas-mediated liver apoptosis. IL-6 may protect the liver from apoptosis after Fas agonist treatment by regulating several anti-apoptotic factors in the Fas signaling cascade. As shown in the model, when bound by its ligand, Fas receptor forms trimers and binds the adapter protein FADD. FADD can then bind, via a death effector domain, the zymogen form of caspase 8 (FLICE/MACH). Caspase 8 then oligomerizes and activates through self-cleavage. Caspase 8 is an initiator caspase and proximal in the apoptotic cascade. If inactivated by FLIP (I-FLICE), the apoptotic signal will be terminated, and the cell survives. IL-6 maintains the FLIP protein level in the liver after Fas agonist treatment and prevents it from rapid degradation. Caspase 8 activates a complex and poorly understood process that culminates in mitochondrial membrane depolarization (mitochondrial permeability transition) and the translocation of cytochrome *c* from the inner mitochondrial membrane to the cytoplasm. Cytochrome *c* binds to apoptotic protease-activating factor-1 (Apaf-1) and in the presence of ATP activates caspase 3. This executioner caspase cleaves intracellular substrates, resulting in the characteristic morphologic and biochemical changes of apoptosis. [104], [105] Bcl-2 and Bcl-xL are anti-apoptotic proteins that function to maintain mitochondrial membrane integrity and prevent the release of cytochrome *c*. Bcl-xL has also been shown to inhibit the association of Apaf-1 to downstream caspases. [106] Likely through posttranscriptional mechanisms, IL-6 establishes an adequate level of Bcl-2 and Bcl-xL proteins in the liver cells to render them more resistant to Fas-mediated apoptosis. (From ref. [93].)

Cytokine-Independent Pathways: Roles of Hepatocyte Growth Factor and Transforming Growth Factor-a

In addition to cytokine-dependent pathways, several growth factors are felt to play an important role in promoting cellular replication during liver regeneration. Two of the most important based in part on in vitro studies in isolated hepatocytes and in part on in vivo studies are TGF-a and HGF. Like IL-6, HGF regulates a variety of processes in the liver in addition to being a direct stimulant of hepatocyte proliferation. [2] HGF is a potent inducer of DNA synthesis in hepatocytes in culture but also alters morphology and motility of cells. In vivo, HGF is synthesized in nonparenchymal cells and acts on hepatocytes in a paracrine fashion. It has been proposed that

HGF is activated by proteases after hepatectomy. Antibodies to HGF at the time of CCl_4 injection block the regenerative response. In this study, the degree of injury (and therefore the requirement for regeneration) was not assessed. [94] It has been shown that infusion of HGF into rodents reduces the level of CCl_4-induced hepatic injury. [27], [28] The effect on specific signaling pathways leading to regeneration has not been studied. In a rat model of liver cirrhosis produced by dimethylnitrosamine, HGF produced after gene transfections into skeletal muscles induces a high plasma level of human HGF, resulting in activation of the c-Met/HGF receptor. [27] The increase in TGF-ß $_1$ normally associated with dimethylnitrosamine is reduced, but it is not clear whether this is a direct effect of HGF or results from the fact that liver injury is less severe in the HGF-infused animals. HGF infusion inhibits fibrogenesis and hepatocyte apoptosis. It produces resolution of fibrosis in an already cirrhotic liver and improves the survival rate of rats. Again, as in the case of IL-6, conclusions cannot be drawn regarding whether the effect of HGF on cirrhosis is limited to its effect on hepatocyte injury or whether it reduces stellate cell activation directly.

The effects of HGF on liver injury are very similar to those of IL-6, but thus far, attempts to link these two cytokine/growth factors have failed. Although HGF may be directly up-regulated by IL-6 treatment in vitro, no change in HGF signaling or HGF mRNA levels is observed in IL-6-/- livers. [20] Moreover, although abnormal regulation of plasminogen that regulates HGF activity is seen in IL-6-/- livers, [39] no difference in HGF-mediated signaling has been detected. It remains to be seen whether the similar effects of IL-6 and HGF on hepatocyte apoptosis, injury, and regeneration are mutually exclusive yet overlapping.

The genetic evidence for a role for TGF-a in liver regeneration is complicated by the fact that there are multiple TGF ligands and receptors in the liver. [16] TGF-a knock-out mice show normal liver regeneration, and TGF-a receptor mutation is associated with embryonic lethality. The presence of multiple ligands for the TGF-a receptor makes it difficult to assess the importance of this strong hepatocyte mitogen in the process of liver regeneration.

LIVER REGENERATION IN HUMAN DISEASE

Many types of liver disease, including hepatitis and cirrhosis, are accompanied by some degree of liver regeneration. In part, this is compensatory as the liver attempts to restore liver mass lost as a result of the pathological process. However, in addition, the disease itself may be accompanied by elevations of cytokines and transcription factors that promote hepatocyte and nonparenchymal cell proliferation. These pathophysiological responses of the liver may exacerbate the disease process, leading to increased fibrogenesis in cirrhosis and the accumulation of mutations in proliferating hepatocytes and progenitor cells that ultimately lead to hepatomas.

It has been found that the X protein and MHBs ^{t} transactivators encoded by the hepatitis B virus induce the NF-?B transcription factor in infected hepatocytes, perhaps by modulating the oxidative state of the cells. [95], [96] and [97] Increased NF-?B may result in the initiation of a growth program and prevent apoptosis. The recruitment of immune cells as a result of increased cytokine production (e.g., IL-8) leads to additional increases in circulating cytokines, which may act as mitogens in both hepatocytes and nonparenchymal cells, leading to fibrogenesis and cirrhosis. [96] Thus, a pathological state of the liver, such as hepatitis, may inappropriately activate pathways that are normally part of a physiological response to reestablish liver mass.

The number of liver transplants performed is growing. Several centers are dramatically increasing the number of living related donors and examining the possibility of using split-liver transplants to increase the availability of organs. [98], [99] In instances in which a small-for-recipient liver is used, a hyperplastic response clearly must occur in the recipient, similar to the type of regeneration that occurs after a partial hepatectomy. Even in an appropriately sized liver transplant, because of the cellular damage that occurs in the donor liver before transplantation, a certain amount of cellular replication and repair may be required after transplantation for normal liver function to be established. Many cases of primary graft failure in liver transplantation result not from the immunologic response but from malfunction of the donor liver cells. Studies suggest that liver regeneration is an important component of the survival of the transplanted liver. [100], [101] and [102] In successfully transplanted livers, and in short-term ischemia when shunting has been performed, a rapid induction and return to normal of the immediate-early genes, jun and fos, occurs, similar to the kinetics of expression seen during liver regeneration. A high percentage of hepatocytes undergo DNA replication, [83] further supporting a role for regeneration in transplant survival. If cytokine release is important for liver recovery after transplantation, cytokine release may also be somewhat deleterious in that it may activate an immune response within the transplanted liver. However, agents that block the immune response, such as steroids, may also hamper the regenerative response by blocking NF-?B or STAT3 activation. [103] Thus, an understanding of the extent of liver regeneration, and the degree of cytokine and growth factor activation after liver transplantation, is clearly needed.

CONCLUSIONS

Liver regeneration after partial hepatectomy represents a physiological response of the body to the loss of liver mass. Based on a molecular analyses of regeneration, it is now better understood how the liver is able to regenerate and have the functional capacity that is required to maintain metabolic homeostasis during proliferation. In pathological processes such as fulminant hepatic failure and sepsis, in which hepatic function may be lost, it is now possible to assess parameters of liver function, such as the levels of hepatic gluconeogenic enzymes and mRNAs and transcription factors necessary to maintain adequate liver function. In regeneration, a correct balance of liver-specific transcription factors such as the C/EBPs is maintained, whereas in hepatic dysfunction, one can predict that the levels of these factors will be abnormally low. Studies will allow for a better understanding of the mechanism by which liver function is lost and may ultimately lead to therapeutic interventions.

Cytokines (TNF-a, IL-1, IL-6), which may be derived from liver nonparenchymal cells (Kupffer and endothelial cells), have an important role in promoting normal liver regeneration. Although more difficult to demonstrate in genetic models, HGF, TGF-a, and other growth factors are likely to be required for normal regeneration. However, it seems clear that the levels of these cytokines and growth factors must be well modulated for the outcome to be effective regeneration rather than liver damage. For example, abnormally high levels of cytokines, seen during pathological processes such as toxic damage to the liver, lead to liver injury and cell death. In other processes, like cirrhosis and hepatitis, high levels of cytokines may lead to an aberrant proliferative response that ultimately leads to fibrosis and hepatomas. HGF and IL-6 have pleiotropic effects in the liver, including growth-promoting, anti-injury, and anti-apoptotic effects. They are more beneficial in inducing proper regeneration than TNF-a, which may have both growth-promoting and growth-inhibiting properties. In processes leading to liver damage, it may be possible to modulate the levels of these cytokines therapeutically to obtain the correct balance of specific cytokines, thus promoting restoration of mass and minimizing injury.

The complexity of the regenerative response, which requires several parallel signals, is highly protective to the organism. If IL-6 alone could induce hepatic hyperplasia, then any condition in which IL-6 was elevated would be detrimental to the animal. For example, during an acute phase response, elevated IL-6 levels aid recovery from injury, but the liver does not undergo rapid expansion. Animals overproducing hepatic IL-6 show high levels of STAT3 and other acute phase genes, but their livers are histologically normal and show little increase in proliferation. However, the introduction of an additional signal, overproduction of the soluble receptor for IL-6 in their livers within the portal spaces, overwhelms the growth-inhibiting signals, and hyperplastic changes, adenomas, and endothelial cell proliferation develop in the livers. [86] Thus, under normal physiological signals, and in response to stress, homeostasis is maintained. A loss of liver mass, through either surgery or toxic damage, is required to provide the complex signals, both cytokine-dependent and -independent, that are required for liver regeneration.

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CHAPTER 29

Douglas A. Drossman and Lin Chang

PSYCHOSOCIAL FACTORS IN THE CARE OF PATIENTS WITH GASTROINTESTINAL DISORDERS

BIOPSYCHOSOCIAL MODEL OF ILLNESS AND DISEASE

Experienced physicians recognize that a clinical approach that focuses entirely on the physical or organic nature and origins of disease has limitations. Even sophisticated diagnostic methods such as endoscopy, manometry, and imaging are not sufficient to explain the degree and variability or even the presence of many gastrointestinal symptoms, or the patient's behavior in the face of these symptoms. Furthermore, such data are insufficient to plan treatment. [1] In other words, abnormalities of structure and function, the *disease,* does not explain the patient's experience of ill health, the *illness*. [2] Failure to link the physical (i.e., disease) and the psychological (i.e., illness) components will reduce the likelihood for more effective treatments.

Consider the following case:

Case Example

A 42-year-old man describes severe chronic epigastric burning and occasional associated mild lower abdominal discomfort that he has had for 10 years. An upper gastrointestinal barium series reveals a hiatal hernia, and barium enema examination demonstrates a "spastic colon." H_2 antagonists and antacids provide some relief of his symptoms, but they recur over time. Upper gastrointestinal (GI) endoscopy reveals a small linear erosion in the distal esophagus, but no evidence of Barrett esophagus. A 24-hour pH study demonstrates significant gastroesophageal reflux. High-dose proton pump inhibitors relieve the epigastric burning, but soon afterward, the patient reports worsening of his lower abdominal discomfort, rectal fullness associated with alternating diarrhea and constipation, and a sensation of incomplete evacuation and bloating.

His mother had had ill-defined upper gut symptoms. The patient reported that stress exacerbates both his upper and lower abdominal and rectal symptoms. He also admits to an obsessive preoccupation that his symptoms will worsen. Results of a colonoscopy with colonic biopsies are normal. When he turned 40, he felt that his life was "not where he wanted to be." He is not married and has no children. He is an unpublished novelist and writer for television. He later discloses that some of his anxiety concerning his rectal symptoms is related to being sexually abused in childhood by a distant relative. He never discussed this with his family or close friends. He feels nervous, caught in a "vicious cycle" of increasing symptoms, recurrent anxiety, and then further worsening of the symptoms.

The key, in terms of treatment, is to break the cycle, both medically and behaviorally. A low-dose tricyclic antidepressant, a short course of a benzodiazepine, and cognitive-behavioral psychological treatment with relaxation training are recommended as a means to reduce the anxiety and maladaptive thought patterns that may be perpetuating his illness experience.

This case illustrates several points. Organic (gastroesophageal reflux disease [GERD]) and functional (IBS) bowel symptoms are present, but in both conditions, the symptoms are influenced by psychological factors [3] and exacerbated by stress. Furthermore, an obsessive preoccupation that his symptoms will worsen further amplifies the perceived symptoms. Notably, following successful treatment of the reflux symptoms, his IBS symptoms worsened, thus perpetuating the illness condition. Other psychosocial influences include the perception of "failure" at age 40, symptom identification stemming from his mother's long-standing GI illness, and a history of childhood trauma. All these factors, including the history of childhood sexual abuse, can be addressed in therapy, thereby reducing the anxiety component exacerbating his symptoms.

The physician helped the patient by interpreting his clinical condition as a "vicious cycle," thus communicating that both psychosocial and medical issues are important. By acknowledging the legitimacy of the symptoms while communicating the influencing effects of psychosocial factors on them, the physician could then institute a multicomponent treatment. The patient was now more accepting of both pharmacological and psychological treatments.

To integrate the interacting roles of psychosocial and medical factors, a biopsychosocial model needs to be considered (Fig. 29-1). [1] Thus, a biologic event, such as the development of Crohn's disease or human immunodeficiency virus (HIV) infection, can affect cellular and organ function, the person, family, and society, while a psychosocial change, such as the death of a spouse or abuse, affects not only the person's psychological status and illness behavior, but also cellular immunity and possibly susceptibility to or activity of the disease. [4]

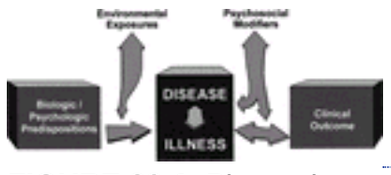

FIGURE 29-1. Biopsychosocial model–conceptualization. Both biologic and psychosocial predispositions (e.g., genetic influences on disease susceptibility and behavior) contribute to the expression of disease and illness. Furthermore, environmental exposures (e.g., infection, family influences on attitudes toward illness), as well as existing psychosocial modifiers (e.g., concurrent psychiatric diagnosis, life stress, social support, coping style), ultimately determine the clinical outcome. (From ref. [1].)

The biopsychosocial model [1, 5] explains the clinical variability among individuals with a given medical condition and provides a framework to integrate both biologic and psychosocial processes and to optimize diagnosis and patient care. In contrast, the biomedical model, which assumes a disease-based single etiology, places "blinders" on the physician, so that psychosocial data either are not recognized or are perceived as irrelevant.

INTERACTIONS OF BIOLOGIC AND PSYCHOSOCIAL FACTORS IN GASTROINTESTINAL ILLNESS

Support for a role for psychosocial factors in GI illness has lagged for the following reasons [6, 7, 8, 9 and 10]:

1. The traditional, biomedical model is inherently slow to change. [7]
2. Only recently have newer techniques (e.g., ambulatory monitoring of motility, visceral pain assessment, standardized psychiatric diagnostic criteria, health-related quality-of-life assessment) been integrated with multidisciplinary research methods. [8]
3. The complexity of psychosocial investigation makes data difficult to obtain and interpret. [8]
4. Research in brain-gut neurophysiology (e.g., positron emission tomography [PET], functional magnetic resonance imaging [fMRI]) is only beginning. [1]

Psychophysiological Factors

Healthy subjects frequently report abdominal discomfort or bowel dysfunction when upset or distressed, [11] and it is well established that stressful stimuli can produce disturbances in intestinal vascularity, secretion, motility, and pain perception. [9] In addition, disturbances in bowel function can reciprocally affect emotional centers in the brain, such as the locus ceruleus. [12]

Stress can also heighten an individual's perception of a physiological stimulus, as in the preceding case example. In patients with GERD, acute psychological stress produced more heartburn, autonomic arousal, and anxiety in patients with preexistent anxiety without a change in the degree of acid reflux (pH probe). [3] Therefore, the increased heartburn resulted from enhanced perception of visceral sensation rather than a physiological increase in acid reflux.

Psychoimmunologic Factors

During the past 25 years, we have learned that environmental stress, via the central nervous system (CNS), influences immune function, susceptibility to disease, and regulation of disease activity. [4] For example, altered in vitro responsiveness to mitogens and antigens, reduced lymphocyte-mediated cytotoxicity and delayed hypersensitivity, diminished skin graft rejection and graft versus host reactivity, and suppressed antibody response have been noted after stressful experiences such as bereavement and marital disruption or stress-inducing experiments in college students. [9] In a study of 394 healthy subjects, there was a significant ($P < .005$) dose-response association between psychological stress and the frequency of respiratory infections in response to the intranasal inoculation of virus. [13] Anxious medical students taking examinations seroconverted to hepatitis B vaccine later than those less anxious, and those with greater social support developed higher antibody titers to the vaccine. [14] These immune effects may be environmentally conditioned. Mucosal mast cells have been classically conditioned to secrete an immune mediator (protease II) in response to an audiovisual cue. [15] By linking the taste of saccharine to cyclophosphamide, immune function was suppressed in NZB-NZW mice, thereby delaying the development of murine lupus erythematosus and prolonging survival. [16]

Finally, behavioral treatment may have clinical consequences. A stress management program of patients with stage I and II malignant melanoma demonstrated a 25% increase in the number of natural killer (NK) cells and in NK cytotoxic activity 6 months after treatment in the stress management group. Six years later, the treatment group had significantly lower mortality, with a trend toward fewer tumor recurrences than the control group. [17]

The Brain-Gut Axis

The brain-gut axis describes the bidirectional neural pathways that link cognitive and emotional centers in the brain with the neuroendocrine centers, the enteric nervous system, and the immune system. [18, 19] For example, extrinsic (vision, smell) or enteroceptive (emotion, thought) information has, via neural connections from the CNS, the capability to affect gastrointestinal sensation, motility, and secretion. Conversely, viscerotopic effects (e.g., nociception) reciprocally affect central pain perception, mood, and behavior. [18]

Modulation of visceral afferent information is discussed in detail in Chapter 38. It occurs at multiple levels:

1. CNS modulation of enterochromaffin cells and immune cells can change the sensitivity of peripheral sensory nerve endings in the gut wall.
2. Descending pathways from the brainstem to the dorsal horn of the spinal cord modulate the amount of information that ascends from the periphery to the brain. [20]
3. Ascending arousal systems from the brainstem to the thalamus and cortex determine how the information is consciously perceived. [10, 21]

The activation of these modulatory systems is dependent on peripheral and, importantly, central events, such as acute gut inflammation [18] or chronic inflammation. [22, 23] Anxiety or recall of aversive memories can enhance, [24] while distraction, hypnosis, and relaxation can decrease, the perception of painful events. [25, 26]

Regulation of these anatomic connections occurs via various brain-gut neurotransmitters. Neurotransmitters are not site specific; rather, they lead to varied effects on gastrointestinal, endocrine, and immune function, as well as neural function and human behavior. For example, the enkephalins can variably affect pain control, [27] GI motility, [28] feeding activity, [29] emotional behavior, and immunity. [30] Cholecystokinin (CCK) increases small intestinal motility, slows gastric emptying, and has a central satiety effect. [31] Meal-associated secretion of CCK may influence bulimia nervosa. [32] 5-HT receptors in both brain and gut are implicated in functional GI disorders, [33, 34] migraine headache, [35] alcoholism, and disorders of mood. [36] Lymphocytes, mast cells, and macrophages contain receptors that respond to neurotransmitters and neuropeptides (e.g., endorphins, vasoactive intestinal peptide, substance P), and this provides the basis for stress and emotion to influence gastrointestinal inflammation and immune function. [37] Nitric oxide can increase the expression of protooncogenes (e.g., c- *fos*) involved in the transcriptional control of genes that encode the production of other neuropeptides (e.g., dynorphin), and this can lead to semipermanent changes in brain or gut function, including the development of visceral sensitization. [38] Corticotropin-releasing factor (CRF), which has central and peripheral receptors, can delay gastric emptying (via CRF-2 receptors), [39] accelerate colonic motility (via CRF-1 receptors), [40, 41] and lower rectal perceptual thresholds. [42]

Dysregulation of the Brain-Gut System in Functional Gastrointestinal Disorders

A unifying hypothesis to explain the functional GI disorders [43] is that they result from dysregulation of "brain-gut" neuroenteric systems. When compared with healthy subjects, these patients exhibit, with considerable variation, increased motor reactivity to several stressors, including balloon distention, food, various peptides, and physical and psychological stressors. [10, 44] Furthermore, patients with functional esophageal, [45, 46] gastroduodenal, [47] and bowel [48] disorders have decreased pain

thresholds to balloon distention and other stimuli (visceral hypersensitivity).

Evidence supports a prominent role of stress in the pathophysiology and clinical presentation of functional GI symptoms. [8], [49] In a predisposed individual, stress can result in permanent, irreversible increased responsiveness of central stress circuits and vulnerability to the development of functional and affective disorders. Gwee and colleagues [50] found that chronic life stressors predict the development of IBS symptoms following gastroenteritis. Alterations in central stress circuits affect the perception of incoming visceral signals [51] as well as output systems to the gut resulting in motility, sensory, and immunologic disturbances, which may be manifested as pain, diarrhea, and constipation. The CNS and spinal cord are able to up-regulate and down-regulate visceral sensation [18] and respond to environmental or psychological stress, personality, or emotions to regulate gut function. Such explains dichotomous symptoms in these disorders: both constipation and diarrhea, or disturbed motility without pain, or even pain without disturbed motility ("altered perception of normal function"). It also explains how psychosocial trauma, such as a history of physical or sexual abuse, [52] or a poor coping style, such as "catastrophizing," [53] profoundly affects symptom severity, daily function, and health outcome.

Recent studies using PET and fMRI indicate that cerebral function differentiates IBS patients from normals. [54], [55] and [56] Using distal colonic stimulation, two studies demonstrated a greater activation in IBS patients of the anterior cingulate (caudal anterior cingulate or rostal midcingulate) cortex, a brain region concerned with organizing the most appropriate behavioral response to sensory stimuli, while taking into account its affective component. [57] Modulation of this region by hypnotic suggestion was associated with changes in the subjective intensity and unpleasantness of a somatic pain stimulus. [26], [57] These findings suggest that IBS patients may fail to use CNS down-regulating mechanisms in response to incoming or anticipated visceral pain. Instead, they may activate areas of the brain that amplify pain perception.

Stress typically produces analgesia of the body, but it has been shown to result in visceral hyperalgesia in recently developed animal models that mimic some pathophysiological features of IBS. [58] These support the observation that early life stress and trauma can influence the development of functional GI disorders later in life. Al-Chaer and colleagues [59] demonstrated that colonic irritation in neonatal rats results in chronic visceral hypersensitivity that persists into adulthood even after the inflammation has resolved. In another rat model, neonatal stress (maternal separation) results in permanent changes in the CNS, which are associated with depression-like behaviors, increased fearfulness, and hypothalamic-pituitary-adrenal responsiveness to stressors and a predisposition to the development of visceral hyperalgesia, somatic hypoalgesia, and increased colonic motility in response to psychological stress. [58], [60]

Implications of Brain-Gut Activity in Inflammatory Bowel Disease

Inflammation may be modulated by disruption of brain-gut pathways. [19], [61] The hypothalamic-pituitary-immune axis is critical to the body's homeostatic regulation of stress, physiological functioning, and disease susceptibility. [19], [62] Through this system, stress influences the immune response, [63] as shown in Figure 29-2. [19]

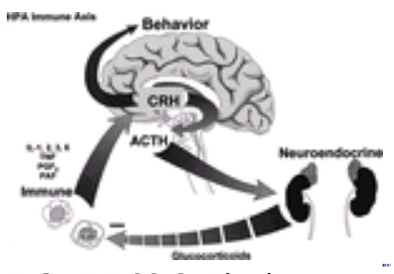

FIGURE 29-2. The immune–central nervous system–endocrine feedback system. Stimulation by immune cytokines and inflammatory mediators facilitates a hypothalamic-pituitary-adrenal response, beginning with release of corticotropin-releasing hormone (*CRH*). This in turn leads to the release of adrenocorticotropic hormone (*ACTH*), which stimulates glucocorticoid release from the adrenal glands. Finally, the glucocorticoids suppress inflammation (*broken arrow*) and the immune response, thereby completing the negative feedback circuit. Homeostasis is maintained through the dynamic and integrated responses of these systems in response to intrinsic or extrinsic forces (e.g., stress, infection, other illnesses) that can disturb the steady state. *IL-1,* interleukin-1; *PAF,* platelet-activating factor; *PGF,* prostaglandin F; *TNF,* tumor necrosis factor. (From ref. [19].)

Chronic psychological stress appears to be capable of exacerbating previously quiescent ulcerative colitis. [64] However, short-term stressful events (past 6 months), perceived stress (past month), or depressive symptoms do not. This suggests that the long-term effects of psychoimmune factors may modify the disease activity in inflammatory bowel disease (IBD). [62] Although psychosocial stressors may not initiate the inflammation, they may dysregulate the immune response, [19], [61] thereby increasing disease activity. The association of long-term stress with the exacerbation of IBD is likely mediated by hypothalamic-pituitary-immune axis, as demonstrated in various animal modules. [19]

The brain and gut are interdependent, and this is best understood from a biopsychosocial framework. Thus, it is no longer rational to try to discriminate whether GI symptoms are gut-related or psychiatric. The clinician's task is to determine the degree to which central and peripheral factors affect the illness condition.

The Experience of Illness and Its Consequences

Illness behavior is determined by sociocultural norms [65] and family beliefs, as well as personality, loss, a history of abuse and other major stressors, daily "hassles," and one's previous experiences with illness. The disease and the patient's behavior affect several health outcomes: symptoms, degree of psychological distress, quality of life, health care visits, and the development of iatrogenic complications (see Fig. 29-1). [8], [52], [66], [67] and [68] The outcome can also be modified through certain coping strategies, [53], [67], [69] by adequate social support networks, [70] and by the effectiveness of the physician-patient interaction. [71], [72] and [73]

Because attention to psychosocial features is likely to improve the patient's clinical condition, clinicians should assess the contribution of these factors to the GI illness and apply this to daily care. The following sections focus on how clinicians can accomplish this.

ESTABLISHING A THERAPEUTIC PHYSICIAN-PATIENT RELATIONSHIP

Obtaining high-quality clinical information and optimally effecting the plan of care require an effective physician-patient relationship. Physicians may feel "at odds" caring for patients with chronic illness for several reasons:

1. Their biomedical training limits their ability to seek out and obtain psychosocial data.
2. They may believe that psychosocial factors are "soft" data and not relevant.
3. They may perceive they have little time to integrate these factors into the diagnostic and treatment plan.
4. They are dissatisfied with the course of treatment.
5. They have a sense of not "connecting" with the patient or the patient's problem(s).

Why do physicians and patients sometimes not "connect"? [73] Some patients may seem demanding when they expect physicians to make them better or contact them frequently with requests. Experience suggests that patients may be responding to feelings of ineffectiveness in managing their own illness, and this may generalize to feeling that they are not in control of their lives. So the expectation for care is off-loaded to the physician. However, we physicians also have expectations and needs as caregivers. The degree to which they are met affects our attitudes and behaviors in administering care. Our responsibility is not to pick up an unachievable burden of responsibility for the patient's care, nor to push it back to the patient ("You'll have to learn to live with it"). Instead, we should share the responsibility to achieve realistic goals. We should facilitate a process in which patients can regain their self-esteem by validating that their feelings are legitimate. We then should help patients to regain control (and self-esteem) by finding ways for them to learn to help themselves as we work to reduce symptom distress and provide support and hope. We also must be vigilant to establish care that is rendered in a time-efficient manner: to work from priorities rather than address all the problems, and to

establish regular brief visits over time.

The physician-patient relationship is evolving from a traditional model, in which the physician assumes a dominant role, to a more negotiated, patient-centered or "participatory" arrangement. [74], [75] Although physicians must take charge of acute disease and medical emergencies, they must encourage shared responsibility for chronic illness. [72], [76] This approach offsets any tendency for patients to assume passive and dependent roles that impair psychological well-being, and it relieves physicians from taking on too much responsibility. Furthermore, this approach is associated with greater patient and physician satisfaction and an improved clinical outcome. [77]

The following guidelines are recommendations for establishing an effective physician-patient interaction [72], [78]:

1. **Listen actively.** Clinical data are obtained through an active process of listening, observing, and facilitating. It is a question of focus—hearing the patient's story, rather than attempting to "fit" the story into a specific disease. It also involves paying attention to the patient's tone of voice, facial expressions, and "body language," and it need not take extra time.
2. **Validate the patient's feelings.** Physicians must validate any feelings of shame or embarrassment that may arise when patients disclose personally meaningful information. Avoid making personal judgments or closing communication with quick reassurances or solutions. For example, statements such as "Don't worry, it's nothing serious" or "Your problem is due to stress" are frequently perceived by patients as dismissive or judgmental. Patients usually do perceive their condition as serious, and for various reasons, [79] they may not consider it to be caused by stress.
3. **Provide empathy.** Empathy involves demonstrating an understanding of the patient's distress while maintaining an objective and observant stance. An empathic statement would be, "I can see how difficult it has been for you to manage with your pain." This communicates the physician's understanding of the issues from the patient's perspective. Providing empathy improves patient satisfaction and adherence to treatment. [80], [81]
4. **Identify the agenda(s)—the patient's expectations and concerns.** The reason(s) for the patient's visit (e.g., "What led you to see me at this time?" "What are your concerns and expectations?") identify the patient's expectations and increase satisfaction with care. [82] Possible reasons include the following: (1) new or exacerbating factors (e.g., dietary change, concurrent medical disorder, side effects of new medication); (2) personal concern about a serious disease (family death or cancer); (3) environmental stressors, including recent losses or abuse; (4) psychiatric comorbidity, particularly depression or anxiety; (5) impaired daily function, with inability to work or socialize; (6) a "hidden agenda," such as narcotic or laxative abuse, pending disability, or litigation. Once the patient's agenda has been obtained, the physician must also communicate an agenda and work toward a mutually specified set of goals.
5. **Limit the discussion of psychological issues to what is consistent with the patient's beliefs.** Many patients are unable or unwilling to report feeling states or acknowledge a relationship between psychological stresses and their symptoms. [79], [83] Therefore, patients may reject the offer of "insight" about the possible role of psychological factors in their illness. The information provided must be consistent with the patient's frame of reference. For example, a behavioral intervention can be explained as a way to help the patient cope with the psychological distress or dysfunction resulting from the illness, and a tricyclic antidepressant can be offered as a centrally acting analgesic.
6. **Don't overreact.** Some patients interact in ways that are perceived as dependent, demanding, or adversarial, such as making frequent phone calls or visits for narcotics, disability, or diagnostic studies. Physicians may *overreact* by becoming angry, performing unneeded studies, or overmedicating. [84] It helps to understand these behaviors as ineffective communications rather than patient problems, and to "tune in" on inner thoughts and feelings ("What is it about this patient's response that makes me feel irritated?") before acting impulsively. For example, when addressing the influence of stress on IBS symptoms, a physician may react defensively to a patient who replies, "Do you mean this is in my head?" by saying, "Oh no, I didn't mean that. This is a real problem in your intestines." Or the physician may become angry: "Well, it is due to stress, and you'll have to learn to live with it." However, the patient has not been given the opportunity to elaborate on the underlying concern, and these statements reduce the patient's ability to appreciate the symptoms as an integrated effect of mind and body. A more appropriate response (after the patient elaborates on the reasons for this concern) would be, "I can see how you *might* feel that way. But I know that the symptoms you're having are real and are due to your irritable bowel. Also, psychological stress, just like overeating or changes in activity or lifestyle, are part of everyone's life, and it makes bowel symptoms act up more in IBS. I'm prepared to work with you to find out all the things that make your symptoms worse, and we can use that information to help manage your symptoms."
7. **Provide education.** Education involves (1) eliciting the patient's understanding and concerns related to the condition, (2) addressing them, (3) providing information that is consistent with the patient's frame of reference or knowledge base, and (4) checking the patient's understanding of what was discussed.
8. **Reassure.** Patients may fear serious disease or surgery and feel helpless, and they need reassurance. The physician should (1) identify the patient's worries and concerns, (2) acknowledge and validate them, (3) respond to the specific concerns, and (4) avoid "false" reassurances ("Don't worry, everything's fine"), particularly before the medical evaluation is complete, because the patient may view this as a doctor's lack of commitment. For example, for the patient with IBS who is worried about an underlying cancer, the physician might say, "I can understand your concern about the possibility of cancer. However, the symptoms you have and the results of the studies are typical of IBS, and this does not turn into cancer. If new symptoms develop, we'll address them, and when you turn 50 (even if you don't have symptoms), we'll also begin the recommended screening program for polyps and early cancer."
9. **Set realistic, shared goals.** A primary goal is to focus the care on improving daily function and quality of life, rather than on attempting to achieve complete relief of symptoms or cure, which is rarely possible. If a patient expects a cure, the physician should acknowledge an awareness of the patient's expectations but indicate that this is not likely. Efforts are better directed toward achieving relief of symptoms and improving daily function and quality of life. This approach leads to achievable shared goals and frees the physician from the unrealistic burden of taking full responsibility for the patient's well-being. The responsibility for the treatment of chronic illness should be assumed by the patient and facilitated by the doctor.
10. **Negotiate.** The patient and physician must agree on the treatment. After an adequate evaluation has been performed, and with the patient's full understanding of the condition, the physician should ask about the patient's experience and understanding of and interest in various treatments, and then provide choices (rather than directives) that are consistent with these beliefs. If the patient does not believe that stress exacerbates symptoms, then stress reduction techniques are not likely to work and should not be initially recommended. Treatment options that harmonize with the patient's beliefs will be accepted. Negotiation is particularly important when an antidepressant is recommended, or when a referral to a psychologist for pain management techniques is offered.
11. **Help the patient take responsibility.** Patients who continually rely on the physician to make treatment decisions and on family and friends to take care of personal responsibilities, and who frequently worry about their illness or inappropriately seek disability, are abrogating their responsibility for the treatment. Patients need to participate actively in their health care, and this can be communicated in several ways. For example, rather than asking the patient, "How is your pain?" one might say, "How are you managing with your symptoms?" The former question leaves the responsibility for dealing with the pain with the physician, whereas the latter assumes the patient's involvement. Another method includes using a diary. [78] This allows the patient to identify exacerbating factors so as to be able deal with them, or to offer treatment approaches with a discussion of their risks, so that the patient can make the choice. If a treatment does not work, then the patient and physician can consider other options together. The responsibility for treatment should be assumed by the patient and facilitated by the physician.
12. **Reinforce health-promoting behaviors.** Patients with chronic illness may receive attention from family and friends and obtain privileges (e.g., time off from work or other responsibilities, disability) by focusing on symptoms and communicating suffering, and this may reduce the patient's motivation toward achieving health. Therefore, the physician has a role to reinforce more healthful behaviors by minimizing diagnostic strategies, providing symptomatic treatment when needed, and rewarding the patient's ability to adapt to and function in the presence of the illness. For example, if the patient reports numerous symptoms, maintain a neutral manner and do not feel compelled to ask the patient to elaborate on all of them. If the information suggests a need for further medical evaluation, act accordingly; however, if not, redirect the inquiry toward management: "I see, and have you still been able continue your household activities?" If the patient discusses positive steps taken towards new activities, nonverbal reinforcement can encourage healthful behaviors; the physician can engage the patient to elaborate by leaning forward, nodding, and smiling. Because patients may not self-initiate behaviors because of feelings of ineffectiveness, it is important for the physician not to set the patient up to fail. For example, if the patient proposes to begin an exercise program or take on a new hobby, do not say, "That's a great idea, I'm sure you can do it." For if the task is not accomplished, the patient may perceive it as a failure. The physician might say, "Given your pain, it will be a difficult task, but well worth the effort. I'm pleased to hear that you're thinking of ways to get yourself better."
13. **Establish limits.** For some patients, limits with regard to frequent phone calls, unexpected or lengthy visits, or unrealistic expectations for care must be maintained. Some physicians, in their desire to help, may extend the care beyond what feels right, and they subsequently feel trapped when these expectations continue. When this occurs repeatedly, the physician might consider and address whether the patient's behavior reflects certain psychosocial agendas (e.g., concerns about loss or rejection, need to be in control, somatization). Physicians should also assert their needs in terms of time and availability. Not to do so poses the risk of the physician feeling angry or helpless, and this is counter-therapeutic. The physician should present personal needs in a way that is not perceived as rejecting or belittling. For example, setting time limitations on visits can be accomplished by scheduling brief but regular appointments of a fixed duration, rather than by extending the time of a particular visit. Patients should be encouraged to bring up the most important concerns first. When the appointment is about to end, the patient should be reminded that about 5 minutes are left. If time runs out and the patient continues to talk, the physician can indicate nonverbally that the visit has ended (e.g., close up the chart, shift to get up) and mention that what is not finished can be brought up the next time. If the patient still continues to speak, the physician can quietly state that the visit has ended and begin to stand up. If the physician is consistent in this approach, the patient will accept the time-structured visit, provided that the physician maintains a commitment to continuing care. Similarly, frequent unneeded phone calls not requiring immediate intervention are addressed by politely reducing the length of the discussion and referring the issue to the next scheduled appointment. Demands for narcotics can be addressed as, "I appreciate how bad the pain is, but narcotics are not indicated and can be harmful because they are addicting

and may harm bowel function and increase pain. I think we need to identify other, more effective treatments for your pain."

14. **Maintain continuity of care.** An important therapeutic effect takes place when the patient learns that the physician will "be there." This involves a belief that the physician is interested and approachable rather than that the physician is constantly available. Establishing ongoing but brief appointments for patients is also helpful. When the symptoms are particularly difficult to manage, or when coexisting psychological difficulties are recognized, concurrent treatment by a psychologist, psychiatrist, or pain center may be needed.

Once established, an effective physician-patient relationship provides the basis for the efficient acquisition of medical and psychosocial data and a mutually satisfying plan of care.

OBTAINING PSYCHOSOCIAL DATA THE INTERVIEW

The medical interview is the *process* of obtaining data to diagnose, treat, and determine prognosis. It is a natural form of inquiry, although all too often the physician's style may interfere with accurate data collection. When properly carried out, the interview also facilitates the development of an effective physician-patient relationship.

Sample Interviews

Observe the following hypothetical example [1]:

Interview 1 The patient is seated, looking down and with shoulders slumped, as the physician enters the room and sits down.

1. Doctor: "Mr…. uh… *(looks at chart)*… Jones? How can I help you?" Patient: *(speaking slowly, looking down)* "Well, it's this aching… right here… *(points to upper abdomen)*… and diarrhea. I've had it for weeks and…" *(briefly looks up at doctor)*…
2. Doctor: *(writing in chart)* "Epigastric pain and diarrhea… any other symptoms?" Patient: "Well, this achi… this pain makes me not want to eat… and I'm sick to my stomach and tired… I live alone now, and…"
3. Doctor: "Hmm… does eating cause the pain and give you diarrhea?" Patient: "Uh, maybe, I guess…"
4. Doctor: "Can you tell me more about the diarrhea?" Patient: "Well, it's loose… *(pause)*… uh, what do you want to know?"
5. Doctor: "How often do you go?… and is there any blood in the stool?" Patient: "About two or three times a day; no, I don't see any blood."
6. Doctor: "Any other symptoms?… fever, weight loss, or vomiting?" Patient: "Yes."
7. Doctor: *(more hurried)* "Well, fever, weight loss, or what?" Patient: "Don't think I've had any fever. I've lost about 10 pounds, and I'm so tired… don't feel like doing anything." Later, in the interview:
8. Doctor: "Well, it seems like you've had these symptoms for a while. What I'd like to do now is examine you and then order some tests… to make sure there's nothing serious… but from what you've said, I don't think that'll be the case. Uh… is there anything you know that could be causing this?… Are you under any stress?" Patient: Well, I'm having a lot of pressure at work."
9. Doctor: *(smiles, closes chart)* "I can understand that."

Later, the physician obtained blood and stool samples, ordered a small bowel series, and performed a small bowel biopsy and flexible sigmoidoscopy. The results were negative, and the doctor reassured the patient and gave him a diagnosis of IBS. This physician believed he had obtained enough data to make a medical diagnosis. However, the questioning style raises some doubt as to the accuracy and completeness of the medical and psychosocial data, and it is likely that the patient was not satisfied in presenting his concerns. Table 29-1 describes several interviewer behaviors that can influence the quality of the data.

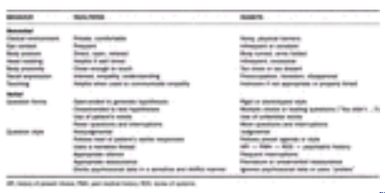

TABLE 29-1 Physician Behaviors Influencing Accurate Data Collection

Several difficulties with the above interview are to be noted:

1. The doctor does not know the patient's name when he greets him (1).
2. Writing in the chart interferes with attention to the patient (2).
3. He interrupts the patient (2, 3, 4, 7).
4. He restates the patient's "aching" as "pain" (2).
5. He asks vague (4) and multiple closed-ended questions (3, 5, 6, 7) that seem to confuse the patient. In general, closed-ended questions are less efficient than a combination of open and closed questions in obtaining medical information. [85]
6. He does not attend to or acknowledge the patient's nonverbal communication (poor eye contact, slumped posture) (1).
7. He does not respond to the patient's verbal cues relating to psychosocial issues (2, 7).
8. He maintains control of the interview, focusing on disease-related questions, without eliciting the patient's agenda (2, 3, 4, 5, 6, 7).
9. He communicates a mixed message about the cause of the illness—that is, he plans to order tests, although he prematurely reassures the patient (8).
10. He "probes" for psychosocial factors only after the medical data have been obtained, implying that the problem is psychological (8).
11. He communicates a lack of interest in exploring psychosocial issues (9).

Consider the differences with another interview style:
Interview 2

1. Doctor: "Hello, Mr. Jones… *(handshake)*… I'm Doctor Smith. How can I help you?" Patient: *(speaking slowly)* "Well, it's this aching… right here… *(points to upper abdomen)*… and diarrhea… I've had it for weeks and… *(briefly looks up at doctor who is attentive)*… I just feel bad."
2. Doctor: "So, you have aching and diarrhea… anything else?" Patient: "Well, I don't feel like eating… and I'm sick to my stomach and tired."
3. Doctor: *(after brief pause)* "Yes?" Patient: *(continues)* "I've lost 10 pounds since this began… *(pause)*… and it keeps me from working."
4. Doctor: *(speaking more slowly)* "So this is affecting your work?" Patient: "Yes… *(briefly looks up as if to say something and then quickly down)*… I worry if it's something serious."
5. Doctor: *(nods, maintains silence)*… Patient: *(continuing)* "I live alone now." *(long silence, nervously looking up and then down)*…
6. Doctor: *(leans forward slightly, looking concerned, and speaks slowly and deliberately)* "Mr. Jones, you seem a bit tense now… Is there anything else you'd like to tell me?" Patient: *(looking down)* "Well,… *(pause)*… I had a roommate who had similar symptoms." *(pause)*…
7. Doctor: *(speaking more softly)* "So you thought you may have caught something? *(patient nervously looks up and then down)… (pause)*… Why do you think that?" Patient: "Well, he was a very close friend… *(long silence, begins to wipe eye and then continues)*… first they didn't know what was wrong… then they found he had AIDS." *(starts to sob)*…
8. Doctor: *(moves closer and after long period of silence)* "I can understand how difficult this has been for you. You've lost someone very close to you and are also concerned about your own health, and it's all affecting your ability to work… *(pause)*… I'd now like to ask you a few more questions about your illness, and then I'll examine you. Whatever we find, we'll need to talk more about what can be done to help you. Is there anything else you'd like to tell me?" Patient: *(looks up)* "No… *(brief smile)*… Thank you, Doctor."

In this interview, the physician obtained additional data that expand the differential diagnosis: The patient may have been exposed to HIV, and depressive symptoms related to the possibility of acquiring the disease and the loss of his close friend may also be contributing to the symptoms. All these possibilities need to be addressed. As noted in Table 29-1, several facilitative behaviors used by this physician are worthy of comment [86]:

1. He readjusts the tempo of his questioning to match that of the patient (4, 6, 7).
2. He uses silence and brief facilitating statements to encourage the patient to elaborate (3, 5).

3. He uses nonverbal behaviors to communicate interest and support (1, 6).
4. He encourages the patient to elaborate by restating his earlier responses (2, 4, 7) without introducing bias.
5. He elicits psychosocial data by facilitation rather than direct inquiry (e.g., probing). This is accomplished nonverbally (e.g., silence, attentiveness [5]) and by a more open-ended interview style that encourages the patient to elaborate on previous cues (4, 7).
6. He acknowledges (and validates) the patient's mood disturbance (6).
7. He simultaneously obtains both psychosocial and medical data. This is done through more open-ended inquiries that explore the symptoms and their psychosocial context (4, 7, 8).
8. He ends by summarizing the patient's concerns and commits himself to help regardless of cause (8). The focus is on the patient's well-being rather than solely on the diagnosis and treatment of disease.

Interview Technique

These vignettes illustrate several important points [87]:

1. The physician seeks to understand the illness from the *patient's perspective* and uses the knowledge to reorient the data into disease-related and behavioral categories. [88]
2. The physician has to have a flexible technique. [89] For example, more open-ended questions are used for patients with unexplained or chronic symptoms, whereas more directed questions are required for medical emergencies. The physician needs to slow his pace to permit a more passively responding patient to elaborate, and to use a more directed style if the patient is garrulous or controlling.
3. The psychosocial and biomedical data are obtained concurrently rather than sequentially. [86] This saves time, yields quality information, and avoids shifting between medical and psychosocial issues that are experienced together. Furthermore, the patient learns to accept the legitimacy of this association, and the physician's interest in hearing about it. It is poor interview technique to avoid psychosocial issues by overly controlling the questions. [90], [91]
4. Addressing the patient's thoughts, concerns, and emotional state facilitates a therapeutic relationship. Being open to communication and caring behavior are major factors in patient adherence to treatment, satisfaction, and continuation of the therapeutic relationship. [77], [92], [93]

UNDERSTANDING THE DATA

Once the medical and psychosocial data have been obtained, their relative contributions to the illness must be determined. Several questions (Table 29-2) can help prioritize these data. [78]

1. Does the patient have an acute or chronic illness?
2. What is the patient's life history of illness?
3. Why is the patient coming now?
4. Is there a history of unresolved major loss or trauma?
5. What are the patient's perceptions and expectations?
6. Does the patient exhibit abnormal illness behavior?
7. What is the impact of the illness?
8. Is there a psychiatric diagnosis?
   a. Depression
   b. Anxiety Disorder
   c. Somatization-Somatoform Disorders
   d. Factitious Disorder
9. Are there cultural or ethnic influences?
10. How does the family interact around the illness?
11. What are the patient's other psychosocial resources?
12. How far do you go in the workup?
13. When do you obtain a mental health consultation?

TABLE 29-2 Questions to Consider When Obtaining and Using Psychosocial Data

Does The Patient have an Acute or Chronic Illness?

Regardless of cause, the longer an illness continues, the more likely psychosocial processes contribute. Medical patients with long-standing unexplained pain are unlikely to have a specific medical disease diagnosed, and psychological assessment usually identifies contributing psychosocial factors. [94]

What Is the Patient's Life History of Illness?

The frequency of other physical complaints and health care visits is an important diagnostic and prognostic indicator. Patients with a history of multiple illnesses and frequent physician visits are more likely to continue to seek health care during future illnesses [68] and have a higher level of life stress and psychosocial disturbances that will require behavioral interventions for improvement. [68] Conversely, patients without a history of medical complaints are likely to require further medical investigation for new symptoms.

Why Is the Patient Coming Now?

With nonemergent symptoms, it is important to determine what factors influenced the patient's decision to seek medical care. These factors are not always volunteered and often must be elicited in a skillful and sensitive manner. For example, asking, "Why did you come now?" may be interpreted as more rejecting than asking, "Was there anything else that led you to see me?" Table 29-3 [95] lists reasons why patients may seek medical care and offers suggestions for treatment.

REASON	PHYSICIAN'S APPROACH
Recent symptom exacerbation	1. Identify exacerbating factors (e.g., diary). 2. Encourage patient to modify them.
Fear of serious disease	1. Determine underlying concerns (e.g., recent family death/illness, prodding by spouse). 2. Offer reassurance. 3. Avoid premature or false reassurance ("There's nothing wrong ... it's nerves").
Environmental stressor(s)	1. Identify stressor(s). 2. Determine patient's insight and ability to work toward changing stressor(s). 3. Determine best treatment approach: a. Wait for resolution. b. Counsel patient. c. Suggest stress reduction techniques. d. Refer for psychological counseling.
Psychological distress	1. Is there a psychiatric diagnosis? a. Anxiety b. Depression c. Somatization 2. Consider psychopharmacological treatment. 3. Consider psychiatric consultation.
Functional impairment	1. Determine change in functional status. 2. Set treatment goal to improve function rather than relief of symptoms.
"Hidden agenda"	1. Determine the hidden agenda: a. Narcotics b. Laxative abuse c. Disability d. Keep sick role privileges (work/home) 2. Clarify limits of your role in treatment.
Social/cultural factors	1. Determine if a. Physician visits are for social support. b. There are cultural effects (e.g., "roots," folk remedies). c. There is a need to legitimize to family/friends. 2. Adapt treatment to be consistent with patient beliefs or expectations: a. Set up brief, regular appointments. b. Permit "folk" remedies if not harmful.

TABLE 29-3 Why Is the Patient Coming to the Doctor?

Is There a History of Unresolved Major Loss or Trauma?

Unresolved losses (e.g., the death of a parent or spouse), personally meaningful operations (e.g., hysterectomy, ostomy), or interference with the outcome of a pregnancy (abortion, stillbirth), may be associated with symptom exacerbations or difficult adjustments to medical disease soon after these events, on their anniversary, or during the holiday season. [96], [97] The physician should realize that illness sometimes serves as an unconscious adaptive mechanism to minimize the

reality of the loss. [98] Although patients may initially deny this association, the loss and its effects (which may require psychological adjustment or counseling) can be acknowledged without linking them to the symptoms.

A history of physical or sexual abuse in medical patients, [99] and particularly in GI patients, [100] is associated with a poor health outcome: more severe pain, symptoms (somatization), health care use, and surgery. [52], [101] These effects of trauma may be related to an increased awareness of bodily sensations, comorbid psychosocial disturbances or poor coping strategies, or a reduction of the pain threshold by peripheral or central mechanisms. [102] The case of the male patient with GERD and IBS presented at the beginning of this chapter illustrates this point.

It is important for physicians to consider prior abuse, particularly among patients with chronic unexplained symptoms. Patients do not usually volunteer this type of information. [101] A history of abuse should be considered in patients with any of the following: severe chronic pain (particularly abdominal or pelvic), severe constipation (pelvic floor dyssynergia), eating disorders (bulimia nervosa, morbid obesity), a history of multiple operations, sexual dysfunction, and severe psychological disturbances (somatization, dissociation or borderline or multiple personality disorder, posttraumatic stress disorder, substance abuse). [100]

It is important to obtain this history in a sensitive and caring manner because further counseling or support groups may facilitate psychosocial adjustment. [100] The patient should first be given the opportunity for voluntary disclosure: “Is there anything else you would like to discuss that you think is important?” If the patient says, “Things were pretty bad then,” elaboration should be encouraged. If no history of abuse is volunteered, the patient might be asked, “Are there any experiences we haven’t discussed that have been particularly painful or difficult?” A negative response is satisfactory; however, a mixed response is still concerning. The patient can be asked more directly, “As you may know, it’s not uncommon for women to have been emotionally, physically, or sexually victimized at some time… has that ever happened to you?”

If the patient denies a history of abuse but the nonverbal behavior is incongruent, it is best to register that information for future inquiry and say no more. If the patient acknowledges abuse, remain nonjudgmental and provide support and gentle encouragement to continue. However, learning the details of the experience is not as important as sustaining an empathic environment for possible future disclosure. When a history of trauma is acknowledged, it is important to inquire whether the patient would like to discuss these thoughts and feelings further with others having similar experiences, or with professionals trained to address the issue further.

What Are the Patient’s Perceptions and Expectations?

Case Example [62]

A 27-year-old homemaker with chronic, moderately active ulcerative colitis for 13 years and high-grade epithelial dysplasia on colonoscopic biopsy on two occasions refused colectomy. During several unsuccessful attempts to “educate” regarding the “medical” reasons for the procedure, she expressed the fear that she would be unable to have children; she and her husband were childless. When these unrealistic fears were addressed, she consented.

This vignette illustrates the need to identify patient misperceptions or unrealistic expectations. Important questions to ask include, “What do you think is causing this problem?” “What are your concerns or fears about this illness?” “What kind of treatments do you think you should receive?” and “What do you hope I will be able to do for you?” [103] Patients who respond, “That’s why I came to you” or “You’re the doctor” should be encouraged to assume greater responsibility for their health care (see below).

Some patients with chronic illness may have unrealistic expectations of another diagnosis or a cure and must be encouraged to state their expectations to work toward realistic treatment goals. Many patients believe that a negative evaluation means that the illness is imagined or psychiatric, which may lead to further requests for unneeded diagnostic tests. This issue needs to be addressed in a manner that does not undermine the patient. For example, consider a patient with chronic abdominal pain who urgently requests that another unnecessary computed tomogram be obtained to rule out a cancer. [72] Rather than trying to “convince” the patient that “nothing is wrong” (by pathological standards), the physician should acknowledge the concern that a cancer must not be overlooked, and that many patients with chronic pain have “real” pain not identified by imaging. [72] Then, the physician can state that based on the clinical data, this test is not indicated but might be performed if, for example, the results of certain laboratory tests became abnormal. Avoid ambiguous behaviors that increase doubts or fears (e.g., ordering studies to convince the patient or “just to be sure”). A commitment to maintain vigilance for new developments and ongoing care reduces the need to perform unneeded diagnostic studies or administer unnecessary treatments.

Does the Patient Exhibit Abnormal Illness Behavior?

The varying patterns of how symptoms are perceived, evaluated, and acted on are designated *illness behaviors*. [104], [105] These are determined by current and prior experiences with illness, the attitudes and behaviors of family, friends, culture, and society, and the patient’s personality, current psychological state, and coping style. *Maladaptive* illness behavior [106], [107] should be considered when there is (1) disability disproportionate to detectable disease, (2) a relentless search for validation of disease, (3) placement of control and responsibility for health care with the physician, (4) a continued sense of expectation to be cared for by others, (5) a tendency to avoid health-promoting roles, and (6) adoption of and (7) display of behaviors oriented toward exemption from work or family obligations, avoidance of stressful situations, or dependence on the physician, with abrogation of personal responsibility in the plan of care.

What Is the Impact of the Illness?

Physicians must decide whether, when, and to what degree medical or behavioral intervention is necessary. Particularly in cases of chronic illness, this decision is best made based on the effect the illness has on the patient’s daily life. For example, pain that leads to decreased activity or ongoing loss of time at work will require more intervention than pain that does not interfere with the performance of daily activities.

Health-related quality of life (HRQOL) incorporates the patient’s perceptions, experiences, and functional status during an illness. [108] This evaluation differs from disease assessment in that psychosocial (e.g., daily function, recreation, sexual function) *and* symptom- or disease-related factors are considered, and its validity rests with the patient rather than on biologic or physician-based standards. Research in HRQOL is growing for gastrointestinal disorders [109], [110], [111] and [112] and postsurgical outcomes. [113], [114]

Is There a Psychiatric Diagnosis?

Psychological illness, such as depression and anxiety, are the common underrecognized diagnoses frequently associated with physical symptoms. [115], [116] A psychiatric diagnosis will modify the patient’s experience of medical disease. Proper disease management may also include antidepressant or antianxiety drugs or behavioral interventions.

In recent years, psychiatric diagnosis has become standardized through the use of the *Diagnostic and Statistical Manual of Mental Disorders,* published by the American Psychiatric Association and currently in its 4th edition (DSM-IV). [117] Because many psychiatric diagnoses (anxiety, panic, somatization) rely on gastrointestinal symptoms, care should be taken not to overdiagnose a psychiatric disorder when the symptoms can be explained by a GI condition (e.g., IBS or IBD).

Depression Depression exists on a clinical continuum ranging from a normal emotional response to loss (the “blues”), to a more intense expression of mood (depressive symptoms), to a collection of signs and symptoms (syndrome of depression), to a severe state of neurochemical disturbance (disease). Similarly, the therapeutic interventions for depression (e.g., “tincture of time,” counseling, pharmacotherapy) are determined by the degree and persistence of the disorders on this continuum. The DSM-IV [118] lists several diagnostic categories of depression: *Major Depression, Single (296.2x)* or *Recurrent (296.3x), Dysthymic Disorder (300.40),* and *Depressive Disorder Not Otherwise Specified (311). Major Depression* is characterized by one or more episodes lasting for at least 2 weeks of depressed mood or diminished interest or pleasure in activities and associated symptoms that may include the following: change in weight, sleep disturbance, psychomotor agitation or retardation, loss of energy, feelings of worthlessness or inappropriate guilt, decreased concentration, and recurrent thoughts of death. *Dysthymic Disorder* is a more chronic (lasting longer than 2 years) disturbance of depressed mood with the same associated symptoms. Finally, *Depressive Disorder Not Otherwise Specified* includes depressive features that do not meet more specific criteria for a mood disorder. It is important to rule out other conditions, including other medical or

psychiatric disorders (e.g., dementia), uncomplicated bereavement, and effects of drugs (corticosteroids, cimetidine, alcohol, benzodiazepines, and a variety of antihypertensive agents) that may mimic or exacerbate an affective disorder. Primary depression may be manifest more in terms of somatic complaints or pain than mood disturbance. If a patients exhibits loss of interest (anhedonia), a depressed affect may not be required to make the diagnosis of depression. [118] Treatment may include antidepressant drugs, psychological (e.g., cognitive-behavioral) therapy, or both. Patients with chronic pain and disability usually have comorbid depression [119], [120] and, not infrequently, a history of loss or abuse. Depending on the severity, treatment may include psychological care by the medical physician or a mental health professional, or antidepressant drugs. Using the previously described interview style, the physician may determine that the patient is depressed and may elicit contributing factors, such as childhood abuse. If there is a long-standing history of pain and the negative studies to date preclude further extensive diagnostic evaluation, the focus should now be on treatment and adaptation to the chronic disorder. Patients may be offered psychological counseling to help them cope with the condition. If this is refused, antidepressant medication may be prescribed to reduce pain and associated symptoms, such as fatigue and poor sleep. Monthly follow-up visits oriented toward improvement in function may be scheduled. Although the sleep disturbance, fatigue, and sense of hopelessness may improve, the long-standing history of pain and chronic illness may preclude complete pain relief. Treatment should therefore be directed toward improved function and adaptation to the chronic illness.

Anxiety Disorders Anxiety is perceived as an impending threat or danger from an unknown or irrational source. It may be associated with a specific stimulus (e.g., phobia), or it may not (e.g., “free-floating” anxiety). It may cause autonomic activation (“flight-fight” response), with symptoms of breathlessness, palpitations, chest or abdominal discomfort, diaphoresis, and diarrhea. An anxiety disorder should be considered in the differential diagnosis of patients presenting with GI symptoms. However, symptoms of anxiety not related to the psychiatric disorder can be caused mainly by the effects of disruptive GI symptoms, such as diarrhea. *Panic Disorder (300.01)* [121] may be the most common anxiety disorder leading to medical visits. It is characterized by recurrent unexpected episodes of panic associated with any the following for at least 1 month: persistent concern about having additional attacks, worry about the implications of the attack, or a change in behavior related to the attacks. At least four or more of the following must be present to define a panic attack: palpitations, sweating, trembling, sensation of shortness of breath or smothering, choking sensation, chest discomfort, nausea or abdominal distress, dizziness or lightheadedness, feelings of derealization, fear of losing control or going crazy, fear of dying, paresthesias, or chills or hot flushes. Another anxiety disorder is *Generalized Anxiety Disorder (300.02),* which is characterized by chronic (lasting at least 6 months) unrealistic or excessive anxiety about life circumstances (e.g., finances, health) that are difficult to control and associated with three or more of the following: restlessness or feeling on edge, being easily fatigued, difficulty concentrating, irritability, muscle tension, and sleep disturbance. Similar disorders include *Panic Disorder with Agoraphobia (300.21)* (symptoms lead to avoidance of places or situations), *Simple (300.29)* or *Social (300.23) Phobias, Posttraumatic Stress Disorder (309.89),* and *Obsessive-Compulsive Disorder (300.30)*. Benzodiazepines are effective in the short-term treatment of anxiety disorders [122]; however, withdrawal rebound is a risk, and tricyclic antidepressants [123] and selective serotonin reuptake inhibitors [124] are also effective and favored for long-term use. Relaxation exercises with respiratory control and cognitive maneuvers improve the patient’s sense of control and help reduce the frequency and severity of panic attacks. [125], [126] and [127]

Somatization-Somatoform Disorders Somatization is the tendency to experience and communicate psychological distress as physical symptoms that are misinterpreted as serious physical illness. *Somatization Disorder (300.81)* is characterized by numerous physical complaints beginning early in life. With this presentation, a prior history of abuse should be considered. [128] A history of many unexplained physical complaints leads to medical visits and impaired social functioning. The complaints must include the following: at least four pain symptoms (e.g., head, abdomen, back, joints, chest), two GI symptoms (e.g., nausea, bloating, vomiting, diarrhea), one sexual symptom (e.g., sexual indifference, irregular menses, vomiting in pregnancy), and one pseudoneurological symptom (e.g., impaired balance, paralysis, hallucinations, loss of sensation, blindness). Other somatoform disorders to consider are *Conversion Disorder (300.11), Hypochondriasis (300.70), Undifferentiated Somatoform Disorder (300.81),* and *Pain Disorder*. Medical or psychiatric diagnoses, or drugs that explain these symptoms, must be excluded. Treatment usually involves behavioral interventions that help reduce the patient’s tendency to communicate via bodily complaints. Antidepressants may be empirically tried as a means to increase the symptom threshold.

Factitious Disorder Patients who have *Factitious Disorder with Predominantly Physical Signs and Symptoms (300.19)* [129] simulate illness (e.g., surreptitiously ingest laxatives or anticoagulants, inject stool, produce false elevation of temperature) in a manner not likely to be discovered. Unlike the motives of patients who malinger, consciously feigning illness for obvious gain (e.g., to obtain drugs, be excused from work, be placed on disability), the motives of these patients are more enigmatic; they usually are attempting to obtain certain benefits (e.g., to be taken care of) in the sick role. An important subgroup of patients with Factitious Disorder are GI patients with laxative-induced factitious diarrhea. [130], [131] Because patients will deny the use of laxatives, the diagnosis usually is based on detecting the substance by stool examination. Phenolphthalein-containing laxatives (e.g., Ex-Lax and Correctol, recently removed from U.S. products) turn pink on alkalinization of the stool or urine. The anthraquinones (e.g., senna, cascara) cause melanosis coli, which can be detected at sigmoidoscopy, and the osmotic laxatives (e.g., sodium phosphate) can be detected by stool electrolyte analysis. In addition, increasing the stool volume with urine or water can be detected by a high stool urea concentration or a low stool osmolality, respectively. If the patient’s activities are a threat to life, a room search may be ethically justified. The findings must be disclosed in a nonpunitive manner, and only when the diagnosis is certain. The problem should be presented as a difficult-to-control compulsive disorder that requires treatment; a confrontation because of unacceptable behavior should be avoided. If the patient refuses psychiatric help, the physician can continue to monitor the medical status. Patients with depressive features may benefit from antidepressants.

Are Cultural or Ethnic Influences Involved?

Cultural or ethnic beliefs may modify clinical illness or the patient’s interactions with traditional health care systems. Strong cultural beliefs regarding illness exist in many ethnic groups, including African Americans, Hispanics, Native Americans, and Gypsies. [132] Physicians should inquire about the patient’s beliefs regarding the onset, pathophysiology, expected course, and desired or expected treatment. [65] When possible, the patient’s illness model should be considered in planning treatment, and doing so may lead to effective interventions.

How Does the Family Interact Around the Illness?

For the most part, early family experiences with illness are associated with some attention and support, although with an orientation toward recovery and health, and usually this carries through to adult life. However, undue attention to illness during childhood may affect later illness behaviors, such as health care visits and costs. [133], [134]

In certain “psychosomatic families,” attention to an illness serves to maintain family harmony. When tensions arise in emotionally linked (“enmeshed”) families, an afflicted child (with diabetes, asthma, inflammatory bowel disease, or anorexia nervosa) is conditioned to focus on the illness, thereby diverting attention from the family distress. [135] This conditioning may inhibit the child’s later ability to achieve autonomy, and the pattern may continue even into adulthood. If the patient is married, at some time the patient and spouse should be seen together. Inquiry should be made about how the spouse perceives and responds to the patient’s illness. When healthy family behaviors are seen, the members can be recruited into helping the patient toward recovery.

What Are the patient’s Other Psychosocial Resources?

Psychosocial factors that promote health (by providing social support, such as churches, recreational clubs, and community organizations) play a role in “buffering” the adverse effects of stress on physical and mental illness and need to be identified. Patients with good social support experience a sense of control over their illness and report lower stress levels than with less social support. [136], [137] Coping, defined as “efforts, both action-oriented and intrapsychic, to manage (i.e., master, tolerate, minimize) environmental and internal demands and conflicts, which tax or exceed a person’s resources,” [138] is another mediating psychosocial factor that promotes health. Among GI patients, a maladaptive coping style such as “catastrophizing” exacerbates symptoms and psychological distress, adversely affects quality of life, and increases physician visits and the risks of surgery. [53], [139]

How Far Do You Go in the Workup?

Abnormal findings on the physical examination or abnormal results of screening tests (e.g., blood in stool, fever, abnormal liver chemistries, abdominal mass) will determine whether further diagnostic studies are needed. [140] In their absence, and particularly in cases with psychosocial difficulties, it is wise to follow the illness over time before making diagnostic decisions. A common pitfall is to overdo diagnostic studies when the physician is uncertain and when the patient insists that “something be done.” [73], [84]

When Do You Obtain A Mental Health Consultation?

When psychosocial factors (e.g., psychiatric diagnosis, psychosocial trauma, impaired functioning) strongly contribute to an illness, a mental health onsultation should

be considered. The consultant will help determine whether pharmacological or additional psychological treatments are needed.

TREATMENT

Psychopharmacotherapy

Psychopharmacological agents may be used to treat patients with moderate to severe functional GI disorders, and as an adjunct in the treatment of patients who have other GI diseases with or without evidence of a primary psychiatric disorder.

Antidepressants The rationale for the use of antidepressants, either tricyclic antidepressants (TCAs; e.g., amitriptyline, imipramine, desipramine), selective serotonin reuptake inhibitors (SSRIs; e.g., fluoxetine, sertraline, paroxetine, citalopram), or less frequently novel antidepressants (e.g., venlafaxine, mirtazapine), may be based on several possible mechanisms and includes the following: (1) treatment of psychiatric comorbidity (with full therapeutic doses) associated with IBS, (2) alteration of gastrointestinal physiology (e.g., visceral sensitivity, motility, and secretion), [141], [142] and [143] and (3) reduction of central pain perception arising from afferent signals in the gut, possibly via facilitation of descending corticofugal pain modulator pathways. [144], [145] The choice of an antidepressant will often depend on its known side effects. The *TCAs* appear to be effective in GI and other conditions. [8], [146] In addition to primary actions on noradrenergic and serotonergic receptors, they also have antimuscarinic and antihistaminic effects leading to symptoms of orthostatic hypotension, constipation, sicca syndrome, sedation, fluid retention, cardiac arrhythmias, and weight gain. Amitriptyline and doxepin have strong sedative effects, so that a single nighttime dose can promote sleep. It has been shown that poor sleep quality is associated with increased bowel symptoms, [147] and therefore these agents can potentially improve both GI and extraintestinal symptoms. However, desipramine or nortriptyline may be chosen if less anticholinergic and sedative effects are desired. The TCAs also decrease phase III propagation velocity and increase orocecal transit time, [142] and clinically they can reduce diarrhea symptoms in patients with IBS. [148] Although full antidepressant doses of TCAs are less frequently used today to treat psychiatric conditions (mainly because of their side effects), smaller doses are widely used to treat a wide variety of chronic pain conditions and functional disorders, [146] including IBS. At lower doses than those usually used to treat depression (up to 75 mg nightly), amitriptyline has been found to be significantly more effective than placebo in reducing abdominal pain and producing global improvement in patients with IBS. [146], [149] Antidepressants have neuromodulatory and analgesic properties, which may benefit patients independently of the psychotropic effects of the drugs. Neuromodulatory effects may occur sooner and at lower doses than those used to treat depression (e.g., 10 to 25 mg of amitriptyline or 50 mg of desipramine). Treatment with TCAs should begin with low doses (e.g., 25 mg/d) and be increased as needed up to full therapeutic doses. The *SSRIs* have not been adequately studied in the treatment of patients with GI disorders and are more expensive, but empirically they help reduce painful symptoms and improve general well-being. [144] They may be particularly helpful for patients with coexistent anxiety, paniclike symptoms, or obsessional behaviors. [124] In addition, they are safer and better tolerated than the TCAs and produce little risk from overdose. Their side effects include nausea, agitation, sleep disturbance (insomnia more often than somnolence), vivid dreams, sexual dysfunction (delayed ejaculation or anorgasmia), night sweats, and possibly weight loss (particularly with fluoxetine). Especially with sertraline or fluoxetine, up to 15% of patients experience diarrhea. However, paroxetine has more antimuscarinic effect than the other SSRIs and may be associated with constipation. [124] Most patients will benefit from a single morning dose (10–20 mg of fluoxetine or citalopram, 50 mg of sertraline, 20 mg of paroxetine, 50 mg of luvoxamine). However, lower doses may be required for the elderly or for patients with liver disease because of the prolonged half-life of the metabolites in these cases. In addition, because of their high affinity for the cytochrome P450 system (particularly paroxetine), the SSRIs should be used with caution if given with TCAs, benzodiazepines, anticonvulsants, antiarrhythmics, or warfarin. [124] Some psychiatrists take advantage of this effect by adding a low dose of an SSRI when patients show an incomplete response to a TCA. Within the SSRI class, fluoxetine may be selected when poor compliance is an issue because the drug has a long half-life; citalopram has been considered to have the lowest side effect profile and may be of particular benefit because of its ability to relieve symptoms via peripheral effects on colonic tone and sensitivity in IBS, and paroxetine has more anticholinergic effect and therefore may be selected for patients with significant diarrhea. Table 29-4 compares low- and high-dose TCAs and SSRIs. In addition, other, novel antidepressants are available; for example, mirtazapine has a potentially beneficial 5-HT $_3$ receptor–blocking effect and should be considered for patients with poor sleep, an inability to gain weight, and diarrhea. Venlafaxine, which produces combined serotonin and norepinephrine uptake inhibition, has been shown to increase stimulated pain thresholds [150] and has been suggested for the treatment of certain chronic painful disorders. [151]

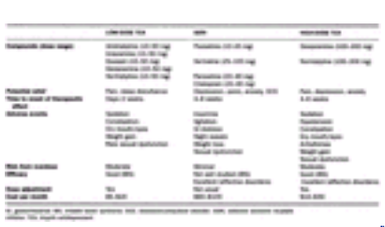

TABLE 29-4 Psychotropic Agents

Most clinicians choose an antidepressant when symptoms of abdominal discomfort are frequent or severe or lead to a loss of usual daily function. It should be continued for 6 to 12 months before tapering is considered. To encourage compliance, patients should be informed that the medication is not addictive and acts as a central analgesic that controls pain in various medical conditions by reducing visceral afferent function or facilitating descending pain inhibitory pathways. However, it also treats depressive symptoms induced by the illness. [78] Patients should also be informed that it can take up to several weeks to work, and that side effects, if they occur, abate over 1 to 2 weeks. A poor clinical response may be caused by noncompliance or a dose that is lower than adequate. [152]

Anxiolytics The benzodiazepines (e.g., diazepam, lorazepam, alprazolam) are most frequently used to treat the experience and behavioral effects of anxiety and stress-induced symptom exacerbations. In view of their weak treatment effects and a potential for physical dependence and interaction with other drugs, benzodiazepine anxiolytics should be prescribed with caution and for a limited period of 1 to 3 weeks. Overall, a benefit for patients with chronic GI disturbances remains to be proved. Also, benzodiazepines may be contraindicated for patients with chronic pain and depression because they decrease serotonin levels and lower pain thresholds, and by stimulating ?-aminobutyric acid receptors, they may actually contribute to depression. [153] Newer antianxiety agents, such as buspirone, do not act on the benzodiazepine receptor, so they may have fewer short- and long-term side effects. [154] Preliminary evidence suggests that 5-HT $_1$ agonists such as buspirone may have a role in decreasing GI symptoms because of their relaxing effects on visceral organs. [155] However, it is not clear whether central or peripheral effects mediate the therapeutic effects in IBS, and further studies are needed.

Antipsychotic Drugs Also called *major tranquilizers* or *neuroleptics,* the phenothiazines (e.g., chlorpromazine) and butyrophenones (e.g., haloperidol) and newer agents (e.g., risperidone) produce improvement in disordered thought, perception, and behavior among psychotic patients. They are of limited value for medical patients, although have been used for nighttime sedation (phenothiazines) and to treat acute agitation or alcohol withdrawal.

Opiates These agents have little or no role in treating patients with chronic pain or psychosocial disturbances because of their potential for tachyphylaxis, abuse, and dependency. Furthermore, they may produce “narcotic bowel syndrome,” leading to worsening abdominal pain and secondary pseudo-obstruction with ileus and vomiting. [156], [157]

Psychological Treatment

Referral for psychological treatment can be proposed as part of a multicomponent treatment package to help patients better manage their symptoms or to address psychosocial difficulties (e.g., abuse, loss) that may interfere with daily function and ability to cope with the illness. In general, these treatments are reserved for patients with moderate to severe symptoms, particularly if they experience psychological distress. However, patients must perceive the treatment as relevant to personal needs. One study found that patients who do not recognize an association between stress and GI symptoms are less likely to respond to psychological treatment. [158]

Some patients may be reluctant to see a psychologist or psychiatrist because they do not understand the benefits of a referral, viewing it as a rejection by the medical physician because of a psychiatric rather than a physical problem. It helps to explain that the mental health professional is a member of a treatment team involved in the patient’s overall management. For patients who do not associate stressors with symptom exacerbation, the physician should indicate that psychological treatment also helps to reduce the psychological distress associated with the symptoms. Furthermore, the psychiatrist may also recommend a pharmacological agent to help with pain control. Continued medical care is essential, even in cases in which the primary focus of treatment involves psychological techniques.

In recent years, specific treatments based on psychological methods to achieve symptom reduction or help patients adapt to their medical condition have become more widely used and accepted by patients. Most of these treatments have been administered to patients with IBS, although they may be potentially beneficial for

patients with other GI conditions.

In a review of the best-designed studies of the treatment of IBS, psychological treatment was superior to conventional medical therapy in 8 of 13 studies. [8] In 7 of 8 studies, psychological treatment continued to be superior to placebo during follow-up (duration, 9–40 months), indicating the these methods have lasting value. The choice of treatment will depend on the patient's requirements, available resources, and the experience of the therapist. The most commonly used psychological treatments in the management of functional GI symptoms are summarized below.

Cognitive-Behavioral Treatment Cognitive-behavioral treatment identifies maladaptive thoughts, perceptions, and behaviors and attempts to "reframe" or modify them to increase control of symptoms. A symptom diary can demonstrate that symptoms are triggered by stressors or lead to negative cognitions (e.g., fear of dying or of cancer, feelings of hopelessness or lack of control, "catastrophizing"). [78] Controlled studies have shown a benefit [8] and a reduction in health care costs [159] for cognitive or behavioral techniques in the treatment of IBS.
Stress Management Stress management is usually taught in small groups in which education and relaxation techniques are provided. Such a treatment program was shown to be superior to a phenothiazine/tricyclic combination (Motival) for IBS. [160]
Dynamic (Interpersonal) Psychotherapy This type of treatment proposes that psychological and physical distress is exacerbated by difficulties in interpersonal relationships, leading to feelings of anxiety. [158], [161] Addressing the psychological issues helps the patient achieve symptom control, which in turn helps prevent recurrences of bowel symptoms. Trials have shown greater and sustained improvement in physical and psychological symptom scores in psychotherapy groups than in support control groups in refractory IBS [158] and functional dyspepsia. [162]
Hypnotherapy Hypnotic induction involves eye fixation, hand levitation, and other techniques to deepen the hypnotic state and increase the subject's openness to progressive muscle relaxation and "gut-directed" hypnotherapy. [163], [164] Patients who undergo hypnosis have fewer bowel symptoms and an improved sense of well-being in comparison with patients in psychotherapy or controls, [163] even after 18 months. [165] Its effect is associated with reduced colonic contractile activity [166] and normalized thresholds for pain caused by distention with a rectal balloon. [167]
Relaxation (Arousal Reduction) Training Reduction in skeletal muscle tension helps counteract the physiological effects of stress or anxiety by decreasing autonomic arousal, tension, and anxiety, and possibly gut motility. Transcendental meditation, mindfulness meditation, and yoga are methods used to achieve these goals.

Behavior Modification

Withdrawal from Narcotics Narcotic abuse is a common problem in patients with chronic pain and other functional GI disorders. Outpatient efforts to reduce narcotic medication gradually are frequently unsuccessful. One approach [168] involves the gradual reduction of a narcotic given on a noncontingent basis in an inpatient setting. The patient must understand and agree to all aspects of the protocol, except for the percentage and timing of the dose reduction. The method involves gradually reducing the usually prescribed dose of the narcotic, generally by giving a long-acting agent (e.g., methadone) over several days and using frequent dosing (to avoid withdrawal effects) in a noncontingent fashion. Clonidine (0.1 mg orally twice daily, or as a patch) is given to minimize withdrawal effects [169] and to help reduce GI symptoms. [170] In addition, a long-acting benzodiazepine is given during the withdrawal phase, and a TCA is begun to provide long-term analgesia if pain is a problem.
Bowel Retraining Bowel retraining can be recommended for patients with habit constipation resulting from poor bowel training or laxative abuse. The technique assumes that the patient has lost the normal physiological response (the "call to stool") and has no underlying organic disturbance in bowel function. All stimulant laxatives are withdrawn or discontinued. The patient is prescribed a high-fiber diet or a daily fiber supplement and an osmotic cathartic (e.g., lactulose or sorbitol). The fiber is not taken if the patient shows evidence of a markedly delayed transit time. The patient eats breakfast and drinks coffee or tea to stimulate the bowels, and then sits for 15 to 20 minutes on the commode. While on the commode, the patient should read for relaxation. There should be no obligation to "perform"; the effort is to identify a time when bowel function can naturally resume. If the patient does not have a bowel movement in 48 to 72 hours, an enema is taken.

CONCLUDING COMMENT: PHYSICIAN CONSIDERATIONS

Physicians have certain desires and expectations in their work [73]: to make a definitive (and at times challenging) diagnosis, to experience intellectual "closure"—an understanding of the medical issues based on existing knowledge, to facilitate a gratifying treatment response (or cure), to feel accomplished (and sometimes praised) in their work, and to establish a mutually gratifying physician-patient relationship.

Unfortunately, these expectations are not met with all patients. In patients with functional (e.g., IBS, functional dyspepsia) or organic (e.g., IBD, GERD, pancreatic or liver disease) GI disorders, psychosocial factors may influence symptoms and behavior in ways that are not understood in terms of the disease or treatable by disease-specific measures. [1] This uncertainty may lead some physicians to feel ineffective in their diagnostic and treatment efforts, [171] and these feelings can be heightened by patients who continue to suffer, and who fail to praise or even criticize the physician.

Some summary guidelines can be offered to help physicians feel more satisfied with their care of patients under these conditions:

1. Make an effort to understand the illness from the patient's perspective, including the psychosocial correlates of the illness.
2. Accept and acknowledge the degree of discomfort or suffering that patients may report.
3. Once the diagnostic issues are clear, accept the uncertainty inherent to medical practice and do not overreact to the patient's distress.
4. Be aware of the "process" of interaction with the patient. "It's not what you do, but how you do it that makes a difference."
5. Refocus the treatment from "cure" or rapid recovery to symptom management and adaptation to illness, including improvement in psychosocial function and quality of life.
6. Work toward achieving mutually determined long-term goals (e.g., return to work, exercise program).
7. Attempt to increase the patient's responsibility ("empowerment") for the plan of care.
8. For patients with seemingly unrealistic needs and demands, determine if they are important from the patient's perspective, and if so, attempt to find a compromise. If they are unrealistic (e.g., undergoing surgery for chronic pain), clearly present your views and limitations in a respectful manner.
9. Remain aware of your thoughts and feelings in the interaction, which will help avoid decisions that later may be regretted (*furor medicus*). [172]
10. Set up and adhere to realistic goals regarding personal time and effort.
11. Finally, some physicians may feel unable or unwilling to work in this manner with patients. They continue to feel frustrated in the effort, and to continue benefits neither the patient nor the physician. When this occurs, appropriate referral is recommended.

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CHAPTER 30

Nicholas J. Talley and Gerald Holtmann

APPROACH TO THE PATIENT WITH DYSPEPSIA AND RELATED FUNCTIONAL GASTROINTESTINAL COMPLAINTS

Dyspepsia is a frequent reason for consultation in primary care and gastrointestinal practice. With the widespread availability and use of endoscopy, it has become evident that a structural explanation is found in only a minority of patients with dyspepsia. The application of appropriate diagnostic and therapeutic strategies in patients with either uninvestigated dyspepsia or documented functional (nonulcer) dyspepsia continues to be a challenge in clinical practice. This chapter aims to summarize the basis for a rational approach to the management of such patients.

CLINICAL PRESENTATION OF DYSPEPSIA

Symptoms

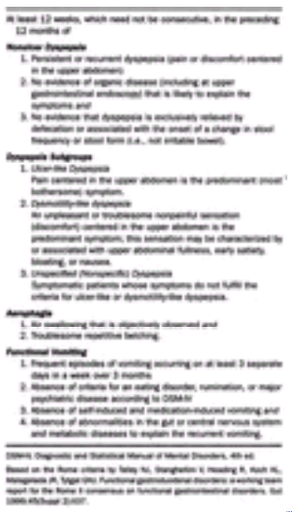
The term *dyspepsia* is derived from the Greek roots *dys* (“bad”) and *peptein* (“digestion”). Based on the consensus opinion of an international panel of clinical investigators, it is defined as persistent or recurrent pain or discomfort centered in the upper abdomen [1]; however, the definition does not exclude those who also have symptoms elsewhere. *Discomfort,* a subjective, negative feeling in the upper abdomen that does not reach the level of pain, may be characterized by one or more symptoms: early satiety, postprandial fullness, bloating, and nausea. *Heartburn,* a retrosternal burning discomfort that rises up toward the neck, is considered distinct from dyspepsia. Dyspepsia is not restricted to meal-related symptoms because patients with peptic ulcer disease often report pain that is not related to a meal. [1] Occasional episodes of dyspepsia often occur in otherwise healthy individuals, but clinically relevant dyspepsia is a relapsing or chronic condition, present for at least 12 weeks in the prior year. [2]

Diagnostic tests do not find a cause for dyspepsia in approximately 60% of studied patients. [1], [3], [4] The term *functional (or nonulcer) dyspepsia (NUD)* refers to relapsing or chronic dyspepsia in patients who lack an identifiable cause for their symptoms after a routine clinical diagnostic workup. [1], [5] Currently, patients with histological gastritis (or duodenitis) are not excluded from the NUD category because a link between these abnormalities and symptoms has not been convincingly established. NUD remains a somewhat confusing, although widely used, medical term. Some authorities who use it infer that the symptoms should resemble those of ulcer disease, [5] whereas others use the label to refer to symptoms that are not at all like those of an ulcer. [4]

The lack of a structural lesion does not mean that the symptoms are not real. Indeed, the lack of a clear association between structural lesions and symptoms has been noted in several other gastrointestinal (GI) disorders. For example, endoscopic studies have found that gastric and duodenal ulcer recurrences are frequent in asymptomatic subjects, [6], [7] and esophagitis and peptic ulcers were found in about 8% and 4% of totally asymptomatic subjects, respectively. [3]

Subgroups of Dyspepsia

Because NUD presumably affects a heterogeneous group of patients, attempts have been made to categorize them based on symptom clusters (Table 30-1). [1], [4], [8] Patients with NUD may be subdivided into at least the following symptom subgroups: ulcer-like, dysmotility-like, and unspecified dyspepsia (see Table 30-1). [1], [5], [8], [9]


At least 12 weeks, which need not be consecutive, in the preceding 12 months of

Rome/new Dyspepsia

1. Persistent or recurrent dyspepsia (pain or discomfort centered in the upper abdomen)
2. No evidence of organic disease (including at upper gastrointestinal endoscopy) that is likely to explain the symptoms and
3. No evidence that dyspepsia is exclusively relieved by defecation or associated with the onset of a change in stool frequency or stool form (i.e., not irritable bowel).

Dyspepsia Subgroups

1. Ulcer-like Dyspepsia
   Pain centered in the upper abdomen is the predominant (most troublesome) symptom.
2. Dysmotility-like dyspepsia
   An unpleasant or troublesome nonpainful sensation (discomfort) centered in the upper abdomen is the predominant symptom; this sensation may be characterized by or associated with upper abdominal fullness, early satiety, bloating, or nausea.
3. Unspecified (Nonspecific) Dyspepsia
   Symptomatic patients whose symptoms do not fulfill the criteria for ulcer-like or dysmotility-like dyspepsia.

Aerophagia

1. Air swallowing that is objectively observed and
2. Troublesome repetitive belching.

Functional Vomiting

1. Frequent episodes of vomiting occurring on at least 3 separate days in a week over 3 months
2. Absence of criteria for an eating disorder, rumination, or major psychiatric disease according to DSM-IV
3. Absence of self-induced and medication-induced vomiting and
4. Absence of abnormalities in the gut or central nervous system and metabolic diseases to explain the recurrent vomiting.

DSM-IV, Diagnostic and Statistical Manual of Mental Disorders, 4th ed.

Based on the Rome criteria by Talley NJ, Stanghellini V, Heading R, Koch KL, Malagelada JR, Tytgat GNJ. Functional gastroduodenal disorders: a working team report for the Rome II consensus on functional gastrointestinal disorders. Gut 1999;45(Suppl 2):II37.

TABLE 30-1 Criteria for the Clinical Categorization of Functional Gastrointestinal Disorders (Rome II)

The categorization of dyspepsia was introduced for the purposes of better targeting treatment to symptoms; for example, patients with dysmotility-like symptoms should respond best to prokinetics, whereas patients with ulcer-like dyspepsia should respond best to antisecretory therapy. Even though this categorization is widely used, its validity is a matter of debate. Several studies using standardized symptom questionnaires observed considerable overlap (10%–20%) and a lack of stability among the different dyspepsia categories. [8], [9], [10] and [11], [12] Previous categorizations generally focused on the presence or absence of specific symptoms, [1], [4] but the severity or predominance of pain versus discomfort may be more relevant, as is discussed in the section on pathogenesis. [2]

Misclassification of Gastroesophageal Reflux Disease as Dyspepsia Whether reflux-like dyspepsia should be considered a part of the spectrum of NUD and what symptoms should be included in this subgroup has been vigorously debated. [1], [4] Despite a lack of endoscopic evidence of esophagitis, pathological acid reflux is detectable in a subset of patients with reflux-like symptoms whether or not they have epigastric pain or discomfort. [13] Heartburn is considered to be of value in identifying true esophageal reflux. [1], [4], [5], [8] The absence of typical heartburn reliably indicates the absence of gastroesophageal reflux disease (GERD) [5], [14]; when present, heartburn is an insensitive indicator of reflux disease in comparison with 24-hour esophageal pH monitoring. [5], [14], [15] At least 20% of patients clinically classified as having NUD have pathological acid reflux, which may explain their symptom complex. [15] Other data suggest that a subset of patients with NUD who respond to acid-lowering therapy have frequent short reflux episodes. [16], [17] It is reasonable to conclude that patients who predominantly have typical reflux symptoms along with dyspepsia should be considered to have GERD, rather than NUD, until it can be proved otherwise. These patients often respond to a trial of acid suppression. Because the term *reflux-like dyspepsia* has resulted in substantial confusion, it should be abandoned. [1], [4]

Functional Vomiting, Rumination, and Aerophagy Dyspepsia and reflux are not the only symptom clusters arising from the foregut (see Table 30-1). *Aerophagy* is an excessive unconscious swallowing of air that results in belching and discomfort. These patients usually report transient improvement of symptoms after belching. [1] Postprandial belching from physiological air swallowing is normal and occurs up to three to four times per hour. Pathological aerophagy is suggested by a specific history and can be confirmed by observation. Because excessive gas is probably not present, it has been suggested that either disturbed upper GI tract motility or psychopathology accounts for the symptoms, but this has not been carefully studied. [1] Patients with *functional vomiting* have frequent episodes of vomiting, but clinical evaluation does not yield a structural or metabolic cause for the symptoms. Gastroparesis and chronic idiopathic intestinal pseudoobstruction should be excluded by appropriate testing (see Chapter 65 and Chapter 73), as should anorexia nervosa, bulimia nervosa, and panic attacks by the clinical assessment. *Rumination* in adults without mental retardation or neurological disease is an eating disorder, not a functional GI disorder. It is characterized by the regurgitation of recently swallowed food that occasionally may be rechewed and reswallowed. These patients may present with a history of considerable weight loss. [18] Symptoms usually do not occur during sleep and are sometimes falsely described or inappropriately labeled as chronic vomiting. [18] Reflux esophagitis and esophageal motor disorders must be excluded, but the history or observation is virtually diagnostic.

Irritable Bowel Syndrome Irritable bowel syndrome (IBS), which affects about one in six persons in the general population, is characterized by recurrent abdominal discomfort or pain and an erratic disturbance of defecation. [8], [10], [11] A positive diagnosis of IBS can be made based on a careful history. [1], [19] Of patients with NUD, at least one third have concurrent symptoms of IBS. [10], [11], [20], [21] Investigators using factor analysis have identified distinct symptom groupings consistent with IBS, NUD, and GERD in the population, although overlap exists. [22], [23] and [24] Because many patients do not mention bowel dysfunction unless specifically asked, an explicit inquiry should be made because the presence of concurrent IBS will influence management (see Chapter 86).

DYSPEPSIA IN THE GENERAL POPULATION AND CLINICAL PRACTICE

Prevalence and Incidence in the Population

Population-based studies from around the world have yielded prevalence rates of dyspepsia ranging from 7% to 60%. [24], [25], [26], [27], [28], [29], [30], [31], [32], [33], [34] and [35] This wide range is unlikely to reflect true variability in prevalence, but probably is explained by methodological differences. Indeed, when two different countries were studied with exactly the same methodology, similar prevalence rates for dyspepsia were observed. [8], [10]

Although prevalence has been extensively studied, few data are available for incidence. One 12- to 20-month population-based study from Olmsted County, Minnesota, identified an onset of symptoms compatible with NUD of approximately 9%, while the same number of subjects who initially reported symptoms became symptom free. [36] In a Swedish study, the calculated 3-month incidence of dyspepsia in previously asymptomatic subjects was under 1%. [21]

Dyspepsia in Clinical Practice and Health Care–Seeking Behavior

Although fewer than 50% of subjects with dyspepsia and IBS seek medical care, conservative estimates suggest that these disorders account for 2% to 5% of all consultations in family practice. [8], [10], [30], [37] Up to one third of these patients eventually may be referred to a gastroenterologist. [37] Although there is an association between the perceived intensity of abdominal (and extraintestinal) symptoms and health care–seeking behavior in NUD, [8], [10] psychological distress also may affect the utilization of health care services. [38], [39] Patients with dyspepsia have more concerns about the possible seriousness of their symptoms and are especially worried about cancer. [40] Moreover, consulters with dyspepsia have poorer coping capabilities, which may magnify their dyspeptic and psychological symptoms. [41]

CAUSES OF DYSPEPSIA

In patients with dyspepsia presenting for investigation, the four major causes of symptoms are chronic peptic ulcer disease, gastroesophageal reflux (with or without esophagitis), malignancy, and NUD. [13], [42], [43], [44], [45] and [46]

Structural Gastrointestinal Disease

The most important structural cause of symptoms to consider is *peptic ulcer disease* because definitive therapy is now available. [47] Symptoms discriminate poorly between peptic ulcer disease and NUD. [35], [48] Indirect evidence for the absence of ulcer disease includes the absence of *Helicobacter pylori* infection on noninvasive testing and no recent nonsteroidal antiinflammatory drug (NSAID) use, but normal upper GI endoscopy findings while the patient is off antisecretory therapy is the only defined diagnostic approach.

In Western countries, the prevalence of *gastric cancer* in previously uninvestigated dyspepsia has been reported to be 1% to 2%, but in persons younger than 55 years of age in most Western countries, gastric cancer is a rare cause of dyspepsia. [3], [49], [50] and [51] Nevertheless, the possibility of a gastric neoplasm must be considered for at least two reasons: Delayed diagnosis of early gastric cancer adversely affects patient prognosis, [52] and patient concerns about cancer promote health care–seeking behavior. [40]

At ultrasonography, *biliary tract disease* (gallstones) may be found in 1% to 3% of patients with dyspepsia but usually is incidental and not a cause of symptoms. [53], [54]

Although the prevalence of gallstones increases with age and is three times greater in female patients, there is no clear age-related increase in the prevalence of NUD and no gender difference. Cholelithiasis causes biliary pain, which is typically severe, constant pain in the epigastrium or right upper quadrant that persists for hours and occurs episodically. [55] In the absence of characteristic biliary pain, it is unlikely that gallstones are the cause of dyspepsia. [56], [57] and [58] Obtaining an accurate history is therefore key in differentiating biliary tract disease from NUD.

Chronic pancreatitis or *pancreatic carcinoma* may cause symptoms that occasionally are confused with those of NUD. [59], [60] Patients often have vague symptoms of insidious onset, but postprandial pain is usual that may radiate through to the back; these patients also may have a history of risk factors for pancreatitis, such as excess alcohol use. Weight loss, nausea, and diarrhea (steatorrhea) are typically late symptoms. Serum amylase and lipase levels are usually normal, but there may be glucose intolerance. In suspected cases, ultrasonography (including endoscopic ultrasonography), helical computed tomography (including thin sections of the pancreas), or magnetic resonance cholangiopancreatography (MRCP) will usually aid in the diagnosis.

Endoscopy-Negative Gastroesophageal Reflux

Because 60% of patients with true GERD are endoscopy negative, gastroesophageal reflux should be strongly suspected, despite the absence of esophagitis at endoscopy, in NUD patients with epigastric burning pain or discomfort that radiates up toward the throat and is relieved by antacids. [61], [62] Twenty percent of dyspeptic patients who were free of peptic ulcer and gallstones had abnormal esophageal acid exposure times. [15] Between 12% and 40% of patients with otherwise unexplained dyspepsia were found to have pathological gastroesophageal reflux [15], [16] and [17], [63], [64]; such patients should not be misclassified as having NUD.

Even if esophageal acid exposure lies within the conventional normal range, symptoms in some cases may be linked to reflux episodes. In a study of 771 consecutive patients referred for 24-hour esophageal pH monitoring, esophageal exposure to acid was normal in 60%, but 71% of these patients reported symptoms during reflux episodes. [16] In one of eight patients with normal esophageal exposure to acid, a significant association was found between symptoms and reflux episodes. The symptom cluster in these patients, which included belching, bloating, and nausea, did not differ between those with and those without pathological GERD. These results are consistent with the concept that *esophageal hypersensitivity* to acid is one of the major pathophysiological explanations of dyspepsia in the absence of pathological levels of acid reflux. [65], [66] Indeed, contact of the esophageal mucosa with acid (without induction of heartburn) causes a cerebral cortical response that is detectable with functional magnetic resonance imaging [67]; hence, esophageal acidification may alter the central processing of visceral afferents.

In patients with difficult-to-manage NUD, 24-hour esophageal pH testing (including an assessment of symptom association with acidification) may be of value to confirm nonerosive reflux disease. The symptomatic response to a short therapeutic trial with a high-dose proton pump inhibitor (PPI) has been advocated as a diagnostic measure for GERD patients, and this test does have good sensitivity and specificity. [68] However, the response to PPI therapy may be misleading in patients with suspected NUD because of the marked spontaneous fluctuations of symptoms in the condition.

Drug-Induced Dyspepsia

NSAIDs, including the cyclooxygenase-2 (COX-2)–specific NSAIDs, represent the most important cause of drug-induced dyspepsia. [69] Other drugs that may produce upper abdominal symptoms include iron or potassium supplements, digitalis, theophylline, and oral antibiotics, especially erythromycin and ampicillin. [70] Reducing the dose or discontinuing drug therapy usually relieves dyspepsia in such cases.

Psychiatric Disorders

Patients with constant pain do not have NUD; rather, these patients usually have a chronic pain syndrome. Similarly, patients with multisystem complaints of abdominal symptoms are more likely to have depression or somatoform disorder and should not be mislabeled as having NUD; these patients can benefit from formal psychiatric evaluation. Rarely, panic disorder can present with episodic upper abdominal distress. Eating disorders should be considered in any young patient presenting with significant weight loss in addition to dyspepsia. [70]

Other Disorders

Diabetes mellitus with underlying autonomic neuropathy can cause postprandial fullness, early satiety, nausea, and vomiting, but symptoms correlate poorly with gastroparesis. Poor glycemic control or psychiatric disorders may be of causal importance. [71] Furthermore, diabetic radiculopathy of the thoracic nerve roots can cause upper abdominal pain. [70] Metabolic disturbances (e.g., *hypothyroidism, hypercalcemia*) can produce upper GI distress. *Ischemic heart disease* sometimes presents with upper abdominal pain induced by exertion. *Intestinal angina* should be considered in older patients, particularly smokers; it typically presents with postprandial pain associated with a fear of eating and significant weight loss. [70] The prevalence of *celiac disease* in patients with dyspepsia may be approximately twice that in the general population in some parts of the world; [72] serologic screening (e.g., antiendomysial antibody) should be considered to allow diagnosis of an eminently treatable disease.

Malignancies such as colon cancer (e.g., involving the transverse colon), gastric lymphoma or sarcoma, esophageal cancer, pancreatic cancer, and ampullary cancer may cause upper abdominal distress that may initially be confused with NUD, although this occurrence is rare. [70] *Infiltrative diseases* of the stomach, including eosinophilic gastritis, Crohn's disease, sarcoidosis, tuberculosis, and syphilis, also may very rarely produce dyspepsia. [73] *Ménétrier's disease* often presents with upper abdominal symptoms; atrophic gastritis is an asymptomatic condition.

Abdominal wall pain from *muscle strain, nerve entrapment,* or *myositis* can be confused with NUD. Characteristically, there is localized tenderness that on palpation reproduces the pain, and the tenderness may be increased by tensing the abdominal muscles. [74]

PATHOGENESIS OF NONULCER DYSPEPSIA

Disturbed Motor Function

During the past 15 years, a role for gastric and intestinal dysmotility in the pathogenesis of NUD has been based on the observation that approximately 40% of patients seen at tertiary referral centers with NUD have delayed gastric emptying of solids, [75], [76], [77], [78], [79], [80], [81], [82], [83], [84], [85] and [86] and a similar number have antral hypomotility after meals. [80], [81] The prevalence of gastric motility disturbances in patients with NUD seen in the primary care setting is unknown but may be lower.

A link between symptoms and gastroparesis in NUD, however, is not always detected. [85], [86] An Italian study of more than 300 patients with NUD reported delayed gastric emptying in only one third and a clear association between delayed gastric emptying (radioisotope technique) and symptoms. [83] The probability of having delayed gastric emptying was more than doubled in female patients with severe postprandial fullness and was increased fourfold in female patients who reported vomiting. However, a larger study of patients with dysmotility-like dyspepsia assessed by a ^{13}C-octanoic acid gastric-emptying breath test failed to confirm these findings. [87] Thus, symptoms and delayed gastric emptying appear to be at best weakly linked.

Gastric antral hypomotility, characterized by either a decreased frequency or decreased amplitude of phasic pressure waves, is not a specific finding in NUD; such abnormalities have been observed in patients with peptic ulceration as well as in those with other diseases. [81], [88], [89] Prolonged ambulatory monitoring of small intestinal motility of patients with NUD has documented symptoms associated with burst activity and retrograde or nonpropagated phase III activity in some. [90] It is unlikely that this technique will identify specific motility patterns that are closely linked to dyspeptic symptoms. [91]

In patients with otherwise unexplained nausea, an elevated slow wave frequency with either a regular (*tachygastria*) or irregular (*tachyarrhythmia*) rhythm has been observed on electrogastrography (EGG). [92], [93] In 72 patients with NUD, the EGG findings were abnormal in 50% with delayed gastric emptying and in 22% with normal emptying. [93] Because gastric arrhythmias also have been documented in patients with severe nausea resulting from GERD [94] and other diseases, [95] the clinical significance of EGG abnormalities in NUD remains in question.

More recently, the concept of impaired fundus relaxation has been suggested as a potential mechanism of symptoms in NUD. After a meal, the fundus normally relaxes, preventing an uncomfortable sensation. [96] A failure of fundic relaxation has been associated with early satiety and weight loss in NUD (Fig. 30-1). [96] This study was conducted in highly selected patients; [96] weight loss is rare in NUD. Interestingly, this functional abnormality could be abolished by sumatriptan, a 5-HT 1 receptor agonist, and symptoms improved. [97] Further studies are needed to confirm this intriguing concept.

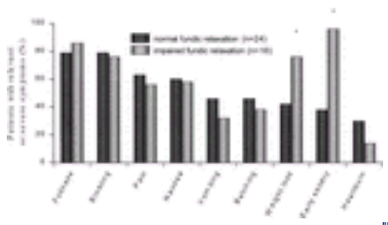


FIGURE 30-1. A study of postprandial fundic relaxation in 40 patients with nonulcer dyspepsia; gastric accommodation was impaired in 40% and was significantly associated with early satiety and weight loss. (From ref. [96] .)

Disturbed Sensory Function

In 1973, Ritchie [98] described lowered sensory thresholds to rectal balloon distention in a subset of patients with IBS, a disorder that frequently overlaps with NUD, and this finding was confirmed by others. [99] , [100] and [101] Moreover, a significant reduction in rectal sensation thresholds could be induced in patients with IBS who had normal baseline thresholds, but not in controls, [102] suggesting a disorder of visceral afferent function. Similarly, patients with NUD have a decreased threshold for the perception of mechanical gastric fundic and duodenal distention (Fig. 30-2). [103] , [104] , [105] , [106] and [107] Furthermore, sensory thresholds could be lowered in patients with NUD by intraduodenal lipid, but not glucose, infusion. [108] , [109] This phenomenon may explain the induction of symptoms after fatty meals reported by some patients with NUD. Nevertheless, approximately 50% of patients with NUD have normal gastric perception thresholds, and there is considerable overlap between healthy controls and patients, raising questions about the exact relevance of gastric hypersensitivity. [110] It has been shown that gastric mechanical thresholds normally increase (or adapt) after repeated distention or exposure to aspirin, but this does not occur not in patients with NUD, [111] , [112] suggesting that sensory dysfunction is one key abnormality in NUD.

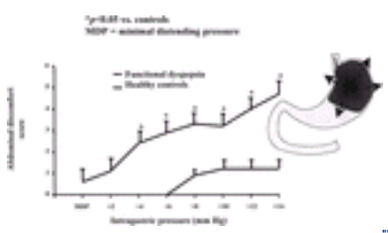


FIGURE 30-2. Gastric sensory thresholds assessed by a gastric barostat in patients with functional (nonulcer) dyspepsia and in healthy controls. (Adapted from Mearin F, Cucala M, Azpiroz F, et al. The origin of symptoms on the brain-gut axis in functional dyspepsia. Gastroenterology 1991;101:999.)

Although most studies have observed lowered gastric mechanosensory thresholds, the data regarding small intestinal sensory thresholds in NUD are more controversial. [113] Decreased thresholds for intestinal balloon distention have been observed in patients with NUD, although the method used (a latex balloon) may not have produced a well-standardized distention stimulus. [114] In contrast, Spanish studies [115] found isolated gastric but not intestinal hyperalgesia in NUD, and selective jejunal hyperalgesia in patients with IBS. [116] More recent studies, however, clearly identified lowered duodenal sensory thresholds in a group of patients with NUD that were independent of coexisting intestinal dysmotility (Fig. 30-3). [106] , [107]

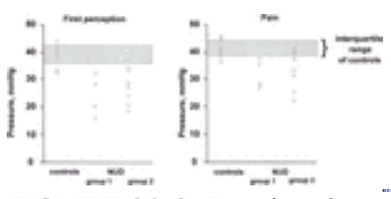


FIGURE 30-3. Duodenal sensory thresholds measured with a barostat placed in the third portion of the duodenum in healthy controls and in patients with nonulcer dyspepsia (NUD). Group 1 had abnormal intestinal reflexes, and group 2 did not. Note that patients with nonulcer dyspepsia in groups 1 and 2 had significantly lower first perception and maximally tolerated pressures with the barostat, and this was similar in those with or without altered intestinal reflexes. (From ref. [107] .)

A key question is whether lowered sensory thresholds in the stomach and duodenum are pathognomonic for NUD. The answer now appears to be no. Both abnormal rectal and esophageal sensory thresholds have been reported in patients with either IBS or NUD. [117] Furthermore, duodenal sensory thresholds were significantly lower in patients with either NUD or IBS and in patients with both conditions than in healthy controls. [20] Altered mechanical visceral sensory function is therefore most likely a nonspecific marker for functional GI disorders rather than an abnormality that directly explains the site and type of symptoms.

Although there are well-defined experimental animal models for the study of visceral hyperalgesia, [118] , [119] and [120] little is known about the mechanisms in humans. It is unknown whether altered sensation results from disturbed thresholds at the GI tract level (i.e., alterations of the mucosal mechanoreceptors, muscle, serosa, or supporting mesentery) or in the afferent nerve terminals, the vertebral ganglia, the spinal cord, the brainstem, and the cerebral cortex. Chronic gastric inflammation caused by *Helicobacter pylori* infection may theoretically be a cause of lower mucosal sensory thresholds, although no consistently significant differences in sensory thresholds between *H pylori*–positive and— negative patients with NUD and healthy controls have been observed. [107] , [121] , [122] and [123]

Mechanical or chemical stimulation of the dorsal root causes visceral hyperalgesia, [124] and it has been speculated that visceral hyperalgesia is associated with altered central projection of central (dorsal horn) neurons. [124] , [125] and [126] The prevalence of extraintestinal complaints, such as back pain and headache, is increased in NUD and IBS, [127] suggesting that central processing abnormalities may be linked also to the pathogenesis of symptoms. Positron emission tomography has revealed differences in cerebral blood flow between IBS patients and healthy controls anticipating rectal distention. [128] Such experiments have not been performed in patients with NUD, but it is reasonable to postulate that similar disturbances may exist in NUD.

Vagal and spinal afferents mediate visceral sensation. Altered vagal function in patients with NUD and IBS has been observed, [129] , [130] including an increase in the area of the antrum in fasting patients with NUD and diabetes mellitus in comparison with healthy subjects. [131] Symptoms of bloating and postprandial fullness are commonly reported by patients after vagotomy, and these symptoms may be linked to an impairment of vagally mediated fundic relaxation. [132] The normal fundic relaxation in response to a meal was observed to be significantly diminished in patients with a truncal vagotomy and in patients with NUD in comparison with healthy

controls. [133] Moreover, other indirect data suggest that the vagus nerve can inhibit the transmission of sensory information, and this inhibition may be impaired in NUD. [134]

Duodenogastric Reflux

Duodenogastric reflux has been postulated to be of importance in the pathogenesis of NUD. There is now convincing evidence that duodenogastric reflux is not more common in those with NUD than in controls, and it does not appear to be related to either symptoms or antral hypomotility. [89], [135], [136] and [137]

Gastric Acid

Basal and peak rates of acid output do not differ between patients with NUD and appropriate controls. [138], [139] However, acid secretion in response to gastrin-releasing peptide (GRP), which simulates the postprandial state, has been shown to be significantly increased in *H pylori*–infected duodenal ulcer patients and *H pylori*–infected healthy volunteers, [140] and more importantly, GRP-stimulated acid secretion was also significantly higher in *H pylori*–positive patients with NUD than in *H pylori*–positive healthy volunteers (Fig. 30-4). [140] Overall, about 50% of the patients with NUD had a disturbance of GRP-stimulated acid secretion similar to that of the patients with duodenal ulcer. Unfortunately, in the population studied, the background rate of peptic ulcer disease is particularly high, and this may have inadvertently contaminated the NUD group. [140] Furthermore, *H pylori*–negative NUD cases were not studied. Thus, the relevance of acid dysregulation in symptom generation in *H pylori*–infected patients with NUD remains to be determined.

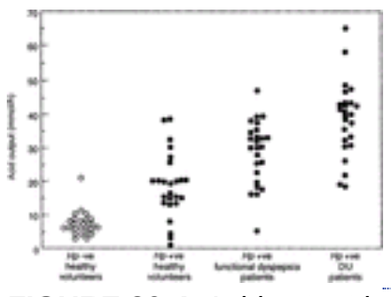


FIGURE 30-4. Acid output in response to gastrin-releasing peptide in *H pylori (HP)*–negative healthy volunteers, *H pylori*–infected healthy volunteers with functional (nonulcer) dyspepsia (*NUD*), and *H pylori*–infected patients with duodenal ulcer (*DU*). Note that the patients with NUD had a significantly increased acid output that was similar in a subset to that of patients with DU. (From ref. [140] .)

Whether NUD patients have gastric mucosal hypersensitivity to acid in NUD is not clear. George and colleagues [141] found that only 6 of 18 patients with NUD, 15 of whom had histological gastritis, had a positive symptom response to gastric instillation of acid, whereas 8 other patients had symptoms during the infusion of saline solution. Pentagastrin injection led to increased reports of pain in patients with NUD, although a histamine H_2 antagonist did not significantly reduce the pain. [142] Because pentagastrin may have other effects in addition to the stimulation of acid secretion, and acid secretion may not be blocked sufficiently by a histamine H_2 antagonist, these data are inconclusive. Esophageal sensory thresholds have been observed to decrease in normal controls during esophageal acid instillation, [143] suggesting that physiological esophageal acid reflux may trigger a decrease in sensory thresholds. In fasting dyspeptic patients, the clearance of exogenous acid infused into the duodenal bulb was shown to be impaired and was linked to the induction of nausea. [144]

Postinfectious Dyspepsia

H pylori infection is the most common cause of histological gastritis in humans [145], [146] and is found in approximately 40% of patients with NUD. [146], [147], [148] and [149] However, it is also common in asymptomatic subjects. [150] Because the prevalence of *H pylori* infection increases with age in those with and without NUD, [146], [149] studies reporting a higher prevalence of *H pylori* infection in patients with NUD [147], [151], [152], [153] and [154] have been criticized for including inappropriate comparison groups or failing to adjust for age, race, and socioeconomic status. [152], [154] In addition, there is a lack of convincing evidence to link *H pylori* with any specific symptom. [153], [154], [155], [156] and [157] In a systematic analysis that included 30 relevant observational studies involving approximately 3400 patients with NUD, and 11 additional studies with more than 6400 patients with uninvestigated dyspepsia, an association between *H pylori* infection and NUD was not confirmed. [158] The authors concluded that based on the available evidence, there is no strong association between *H pylori* infection and dyspepsia, but the evidence was insufficient to confirm or refute the existence of a modest association. Another issue in weighing any link between *H pylori* and NUD is biologic plausibility. Although chronic gastric inflammation is a potential cause of visceral hyperalgesia, [152] two studies failed to find a significant difference in sensory thresholds between *H pylori*–positive and–negative patients. [107], [121]

Thus, a causal relationship between *H pylori* colonization and NUD remains to be proved. However, the data do not exclude a role for *H pylori* infection as an initiating event in NUD in a predisposed individual, imilar to that proposed for postinfectious IBS. [159], [160] Similarly, a small subgroup of patients with NUD characterized by early satiety, nausea, weight loss, and impaired fundic accommodation may develop symptoms following a short episode of infectious gastroenteritis. [161]

Duodenitis

The prevalence of histological duodenitis in patients with NUD is between 14% and 83%, [162], [163] whereas the prevalence of macroscopic duodenitis in asymptomatic patients is 10%. [164] A duodenal ulcer may eventually develop in 48% of patients with duodenitis. [164], [165] It has been claimed that endoscopic irrigation of the duodenum with hydrochloric acid induces symptoms in patients with duodenitis, but unfortunately controls have not been not evaluated. [166] Based on the available evidence, erosive duodenitis appears to fall more within the spectrum of chronic duodenal ulcer disease and probably should not be considered part of NUD.

Psychosocial Factors and Alterations of the Central Nervous System

Individuals with NUD have been shown to be more psychologically disturbed, in terms of being more anxious and depressed, [167], [168], [169], [170], [171] and [172] and to score higher on measures of neuroticism [173] and somatization [127], [174], [175] and [176] than healthy controls. Other studies suggest that patients with NUD are no more psychologically disturbed than patients with organic bowel disease. [167], [177] In a case control study, Talley and colleagues [169] found similar levels of control of anger, anxiety, and unhappiness and of total emotional control over negative reactions in patients with NUD and control subjects. This subject remains controversial.

Most of these studies have included only individuals with NUD presenting for medical care. These studies, therefore, do not rule out the hypothesis that rather than causing symptoms, anxiety and depression simply make patients with NUD more likely to present their symptoms to a physician. [10] However, one large recent study was unable to confirm this concept. [178] Furthermore, in a follow-up study of patients with duodenal ulceration and NUD, abnormal personality patterns appeared to normalize after disappearance of the abdominal symptoms, [179] suggesting that psychological disturbances in some patients with NUD may be a consequence of the symptoms rather than of causal importance. Alternatively, both dyspepsia and psychological disturbances may be caused by another, unknown common environmental or genetic factor.

If psychological factors directly contribute to the development of symptoms in some patients with NUD, the precise mechanisms remain to be elucidated. In depressed patients, the hormonal response to a serotoninergic challenge is diminished. [180] A similar abnormality of central 5-hydroxytryptaminergic pathways may also exist in the functional GI disorders. Indeed, the prolactin response to buspirone, an azaspirone that stimulates central serotoninergic-1A receptors, was found to be significantly greater in patients with NUD than in healthy controls, and the difference correlated with the degree of delayed solid phase gastric emptying assessed scintigraphically. [180] These results require confirmation, however, because D-fenfluramine, a selective stimulus to central 5-hydroxytryptaminergic pathways, increased plasma prolactin and cortisol concentrations to a similar extent in patients with IBS and in controls, suggesting that central 5-hydroxytryptaminergic

pathways function normally in IBS. [181]

The role of stress in the pathogenesis of NUD remains controversial. Acute stress may result in decreased gastric contractility that precedes the onset of symptoms. [182], [183] and [184] It is not known, however, whether chronic dyspeptic symptoms are explained by such mechanisms. Indeed, patients with NUD, with or without antral hypomotility, have normal autonomic and humoral responses to experimental stress. [82] The stress of major life events, such as bereavement or divorce, has also been linked to NUD, although further studies are needed to evaluate this important area. [185], [186] Most studies have not focused on long-term stressors, and it is questionable whether experimental laboratory stressors reflect the stress of real life. Examinations, as an example of a real-life stressor, induced dyspeptic symptoms in medical students that were associated with personality traits, including anxiety. [187] Another study evaluated major life event stress in 100 patients with NUD, 100 patients with duodenal ulcer, and 100 healthy controls; the patients with NUD reported significantly more stressful life events. [188] It is notable, however, that most studies have not focused on long-term stressors and have not controlled for recall bias; abdominal symptoms may prompt patients more often to remember events that occurred in the past related to the occurrence of symptoms.

A potential link between childhood or adult sexual, emotional, or verbal abuse and functional GI disorders is based on studies in patients with IBS [189] and population-based surveys. [190] When standard criteria were applied, a surprisingly high prevalence (26%) of abuse in IBS patients (41% in women and 11% in men) was reported by subjects in a U.S. community. [190] Similarly, dyspepsia and heartburn were both significantly associated with abuse. [190] A causal link, however, has not been established, and notably, subjects with a history of abuse are more likely to seek medical treatment. [190]

Environmental Factors

The long-term use of NSAIDs causes asymptomatic mucosal lesions in up to 60% and ulcers in 10% to 30% of subjects treated; however, the role of aspirin and NSAIDs in NUD is not clear. Whereas acute dyspepsia can be induced by the ingestion of these drugs, including low doses of aspirin [9], [10] and COX-2 inhibitors, [69], [191] it is uncertain how often they cause chronic symptoms. [192], [193], [194], [195] and [196] In a random sample of 1644 residents in Olmsted County, Minnesota, aspirin (odds ratio, 1.8) and smoking (odds ratio, 1.5), but not alcohol (odds ratio, 0.9), were associated with dyspepsia. [196] However, after adjustments for nongastrointestinal somatic complaints, these environmental factors were no longer significant. [196] Most investigations have consistently failed to demonstrate that smoking and alcohol are important risk factors in NUD. [192], [193], [194], [195] and [196]

Some patients with NUD and IBS report specific food intolerances, but a convincing relationship between diet and chronic dyspepsia remains to be demonstrated. [197], [198] In one study, the main difference in eating pattern noted between NUD and controls was that a significantly lower percentage of patients with NUD regularly ate three meals per day. Also, dyspepsia patients in both groups associated certain eating habits and the consumption of specific foods with exacerbations of dyspeptic symptoms, which led to food avoidance in 80% of both groups. [198] In a cross-sectional survey from the United Kingdom, 20% of the population reported food intolerance, but food intolerance, as assessed by a double-blind, placebo-controlled food challenge, was confirmed in only 20%. [199] It is therefore unlikely that food intolerance is a major cause of symptoms in otherwise unexplained dyspepsia.

Coffee and decaffeinated coffee stimulate acid secretion. [194] Although the number of cups of coffee ingested has not been linked to NUD, [192], [193] coffee induced symptoms in 53% of patients with NUD and in 22% of healthy controls. [193] Whether coffee acts as a direct irritant or precipitates gastroesophageal reflux in NUD is unknown. [200]

A Proposed Disease Model

NUD is likely to be a multifactorial disorder. A subset of patients have atypical gastroesophageal reflux but are mistakenly labeled as having NUD. A small group (perhaps those at risk for future peptic ulceration) may have *H pylori*–induced dyspepsia that totally resolves after cure of the infection, because mucosal inflammation resets afferent pathways and symptoms persist despite healing of the gastritis. Acute resolving viral or bacterial infections in childhood or adulthood may be important in initiating visceral hypersensitivity or fundic failure of accommodation. These patients may be genetically predisposed to the development of an irritable gut, perhaps related to abnormal “hard wiring” in the enteric nervous system or higher centers (e.g., the personality trait neuroticism). A number of external factors then may be able to induce or modulate visceral hypersensitivity and altered intestinal reflexes, with or without secondary motor abnormalities, that lead directly to symptoms. It seems less likely that stress or personality traits alone can cause NUD, but they may promote abnormal processing of disturbed afferent signals, which then are interpreted as symptoms. Moreover, psychological status, stress, cultural and family factors, social support, and early childhood experiences are likely to influence the interpretation of symptom severity and health care seeking. Although speculative, this synthesis best fits current data. These concepts will of course have to be modified as new information becomes available.

DIAGNOSTIC APPROACH TO THE PATIENT WITH UNINVESTIGATED DYSPEPSIA

Identifying Patients with Structural Disease as a Cause of Dyspepsia

The diagnostic workup of patients with dyspepsia can be time-consuming and costly if undirected. Therefore, attempts have been made to limit diagnostic procedures to patients at higher risk for structural, potentially life-threatening disease. Age, symptom patterns, and *H pylori* status all have been evaluated as predictors of structural disease.

Age The risk for having a structural cause of symptoms increases with age. [3], [43], [44], [201], [202] A cutoff of 45 years has been traditionally applied in dyspepsia management strategies because the risk for gastric cancer in Western countries is extremely low in patients younger than 45 years. [43], [44] Patients older than 45 years with new-onset dyspepsia should undergo upper GI endoscopy. Because esophagogastroduodenoscopy (EGD) yielded only three gastric cancers in a sample of more than 7000 dyspeptic patients younger than 45 years without sinister symptoms, [49] the age of 50 or 55 years has been proposed by some as a more practical cutoff. [50], [203]

Pattern of Symptoms A long duration of symptoms in younger patients makes cancer an unlikely cause of symptoms. Dividing uninvestigated patients into ulcer-like, dysmotility-like, and nonspecific dyspepsia subgroups does not appear to identify subjects at higher risk for a structural lesion. [35], [48], [204], [205] and [206] Similarly, individual symptoms are of poor discriminating value. [206], [207] In subjects without alarm features, such as weight loss, bleeding, dysphagia, and recurrent vomiting, and who are younger than 45 years, gastric cancer is a very rare finding. [203]

H pylori *H pylori* infection and traditional NSAID use are strongly associated with peptic ulcer disease. [208] Restricting endoscopy to younger patients who test positive for *H pylori* by locally validated serology or a ^{13}C- or ^{14}C-urea breath test, to NSAID users, and to patients older than 45 years reduces the need for endoscopy by 20% to 40% without causing a relevant number of peptic ulcers or gastric cancers to be missed. [208], [209] This strategy, however, has several limitations; the accuracy of serology can be highly variable, and although peptic ulcer disease is declining overall in the developed world, an increasing proportion of patients have *H pylori*– and NSAID-negative ulcer disease. [210]

Endoscopy

Peptic ulcer disease, reflux esophagitis, and gastric or esophageal cancer are the most important conditions that must be ruled out to establish a firm diagnosis of NUD. Upper GI endoscopy remains the gold standard test and is superior to upper GI radiography. [211]

Not all structural abnormalities identified at endoscopy are clinically meaningful. In a study from Switzerland, reflux esophagitis or gastric and duodenal ulcers were found in 7% of asymptomatic subjects and in 24% of patients. [202] Similarly, in a large Norwegian population study, only peptic ulcers and duodenitis were definitely associated with dyspeptic symptoms, whereas other endoscopic and histological findings were not linked to dyspepsia. [3]

It is notable that some patients with a history of peptic ulceration continue to have dyspepsia even after cure of *H pylori* infection and in the absence of a persisting ulcer, which suggests that NUD has developed. [212], [213] If up to 50% of patients with duodenal ulcer have dyspeptic symptoms after apparent cure of their disease, [212] it can be speculated that the mechanisms causing ulcer dyspepsia are also linked to the pathophysiology of NUD.

Other Diagnostic Tests

Abdominal ultrasonography has a low yield and should not be ordered routinely. [35], [58], [70] Whereas endoscopy may yield pathological findings in about 50% of new patients with dyspepsia, an extended functional workup (including 24-hour esophageal pH recording, testing of gastric emptying, biliary scanning, and testing of lactose tolerance) will find abnormalities in only 50% of the remaining patients. [13] The true gain with these additional tests, however, is small. For example, identification of delayed gastric emptying by scintigraphy is unlikely to influence whether or not a prokinetic is prescribed because it has not been established that patients with delayed emptying respond better than those with normal emptying. [214] Similarly, although lactose deficiency may be present, dyspeptic symptoms often fail to respond to lactose withdrawal. A list of recommended tests to establish a diagnosis in dyspepsia is presented in Table 30-2.

Useful
1. A careful history that elicits the symptoms, and a relevant physical examination
2. Upper gastrointestinal endoscopy during a symptomatic period off acid suppression
3. Routine hematologic and biochemical tests (full blood count, ESR or CRP, serum glucose measurement, liver function tests, electrolytes and creatinine, calcium, thyroid function)
Optional
Helicobacter pylori testing
Ultrasonography of the gallbladder, liver, and pancreas
24-Hour esophageal pH testing
Uncertain Clinical Value
Gastric-emptying study
Fundus relaxation postprandially (e.g., by SPECT or ultrasound)
Electrogastrography
Gastroduodenal manometry
Water or nutrient load test

CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; SPECT, single photon emission computed tomography.

TABLE 30-2 Diagnostic Studies to be Considered in the Patient with Suspected Nonulcer Dyspepsia

MANAGEMENT STRATEGIES

The initial management options are outlined in Table 30-3. Because only a few patients with dyspepsia have peptic ulcer, and even fewer have cancer, the American College of Physicians recommends empiric medical therapy (e.g., a histamine H_2 blocker) for 4 weeks for younger patients (i.e., younger than 45 years) without alarm features (option 1 in Table 30-3). [211] The use of PPIs has been extended to dyspepsia in clinical practice. The empiric therapy approach has been criticized, however, because it may promote the prolonged use of inappropriate medications, weaken the value of subsequent investigations, cause serious side effects (rarely), mask symptoms of serious disorders, and mask malignancy at subsequent endoscopy. [215], [216] Furthermore, if there is a high likelihood of symptom recurrence after the end of treatment, most patients will be investigated anyway, and resources will not be conserved. Lastly, this recommendation also fails to take into account the fact that dyspeptic patients with *H pylori*–related peptic ulcer usually have a potentially curable disease.

Exclude symptomatic gastroesophageal reflux, irritable bowel syndrome, and other conditions by history and examination.
Determine whether endoscopy is indicated immediately:
Age >50 y and new symptoms
Alarm symptoms or signs (e.g., weight loss, recurrent vomiting, bleeding)
If patient is young (<50 y) and no alarm features:
Option 1
Empiric therapy initially (e.g., antisecretory agent with optional endoscopy if fails to respond or promptly relapses)
Option 2
Test for *Helicobacter pylori* status (serology or breath test or stool antigen test).
If *H pylori* positive, empiric anti–*H pylori* treatment; if fails, go to option 1.
If *H pylori* negative, go to option 1.
Option 3
Prompt endoscopy for all patients.
Diagnose ulcer, cancer; treat appropriately.
Positively diagnose nonulcer dyspepsia and treat accordingly.

TABLE 30-3 Management Options for Uninvestigated Chronic Dyspepsia

Prompt endoscopy (option 3 in Table 30-3) has long been accepted as the gold standard approach. A large Danish randomized trial compared prompt endoscopy followed by targeted treatment with histamine H_2 receptor antagonists versus empiric histamine H_2 receptor antagonist treatment with diagnostic endoscopy reserved for those who failed therapy. [217] Two thirds of those randomized to empiric treatment eventually underwent endoscopy during the 1-year follow-up. It is noteworthy that the diagnosis of two cases of esophageal cancer was delayed 1 month by empiric treatment. After 1 year, the dyspeptic symptoms and functional status had improved similarly in both groups. Nevertheless, a significantly greater number of patients in the empiric treatment group were dissatisfied with their management and had lost more time from work, although this may reflect patient expectations before entry into the trial.

A number of management trials have now shown that empiric *H pylori* testing and treatment lead to outcomes similar to those observed after prompt endoscopy (option 2 in Table 30-3). The test and treat *H pylori* strategy was associated with a two-thirds reduction in subsequent endoscopies performed within 1 year. [218] In one controlled trial, fewer patients were satisfied with the *H pylori* test and treat strategy, [219] but other data suggest that this is not of major clinical concern. [218] In contrast, *H pylori* testing and referral of positive patients for endoscopy (versus acid suppression) was not cost-effective, and the outcomes were similar. [220] Thus, the *H pylori* test and treat strategy for dyspeptic patients younger than 45 years, with the use of office-based serology testing, appears to be safe and may result in lower costs than initial endoscopy with similar clinical outcomes. [218] For success, the test and treat strategy relies on a sufficient background prevalence of *H pylori* and peptic ulcer disease; the approach will fail if the prevalence of *H pylori* infection falls below 10% in the population. [215]

Endoscopy remains the gold standard approach and can be more cost-effective than other management strategies if the cost is below U.S. $500.00. [221]

Therapeutic Approach in Documented Nonulcer Dyspepsia

Placebo Several pharmacological treatments are now available for patients with NUD (Table 30-4), and most have been compared to placebo in controlled trials. The response to placebo in NUD ranges from 30% to 60%. [222], [223] This placebo response, however, may not reflect a nonspecific effect of treatment but rather spontaneous regression of the disease. Indeed, the course of NUD typically is characterized by relapsing and remitting symptoms, but during a 1-year period, more than 70% of patients will continue to have symptoms. [36], [224], [225] A major issue in the treatment of patients with NUD is improvement of the long-term outcome; unfortunately, most trials have focused on short-term results only. Moreover, the quality of the trials in this field has been highly variable. A systematic review in 1996 of all published trials concluded that most suffered from important weaknesses in study design and execution, such as failure to include validated outcome measures. [222]

First Line
Histamine H_2 receptor blocker*
Proton pump inhibitor
Anti-*Helicobacter pylori* therapy
Second Line
Tricyclic antidepressants (low dose)
Prokinetic (cisapride,† metoclopramide, domperidone)
5-HT_1 agonists (e.g., buspirone, sumatriptan)
Simethicone
Sucralfate
Uncertain or Unknown Efficacy
Promising new visceral analgesics
Serotonin type 3 (e.g., ondansetron, alosetron) or type 4 receptor antagonists
Promising new prokinetics
New serotonin type 4 agonists (e.g., tegaserod)
Gonadotropin-releasing hormone analogs
Somatostatin analogs
Unlikely to Be Beneficial
Antacids
Prostaglandin analogs
Motilinomimetics
Anticholinergics/antispasmodics
Nitrates

* Commonly prescribed, but trial data equivocal (although metaanalyses positive).

† Restricted access because of rare life-threatening cardiac side effects (long QT syndrome).

TABLE 30-4 Pharmacological Treatment for Nonulcer Dyspepsia

Antacids/Simethicone Randomized controlled studies have failed to show a significant benefit of antacid use over placebo. [226], [227], [228] and [229] Antacids are probably most effective in those with undiagnosed gastroesophageal reflux. [230] The potential role of gas retention and air swallowing in NUD was indirectly examined in one study, which observed a significantly greater improvement in symptoms during treatment with simethicone than with cisapride, [231] but more data are needed.

Acid Inhibition Controlled trials testing histamine H_2 receptor antagonists have yielded conflicting results. [222], [226], [228], [230], [232] These might be explained in part by the relatively small sample sizes, and thus inadequate study power, and the heterogeneity of the study populations. Metaanalysis suggested a benefit of histamine H_2 receptor antagonists over placebo, but only selected trials could be included, and studies with patients who had GERD symptoms were not specifically excluded. [232], [233], [234] and [235] The efficacy of the selective muscarinic blocking agent pirenzepine (still available in many countries, but not the United States) is questionable. [230], [233] It is notable that this drug has a lower acid-inhibitory potency than histamine H_2 blockers and has anticholinergic effects that decrease lower esophageal sphincter pressure. Reports have described greater relief of symptoms during treatment with a PPI than with placebo or ranitidine in uninvestigated dyspepsia [236] and NUD. [237] Several large placebo-controlled studies [238], [239] and [240] have provided further evidence that inhibition of acid secretion with a PPI relieves symptoms in a subgroup of patients with NUD. However, the therapeutic gain over placebo is modest and may in part be explained by contamination of the trials with GERD patients. Moreover, those with dysmotility-like dyspepsia may not respond better to a PPI than to placebo (Fig. 30-5). [239] A greater response to PPI than to placebo was observed in *H pylori*–infected patients in one trial, [237] but this was not confirmed by other, methodologically sound trials. [239], [240]

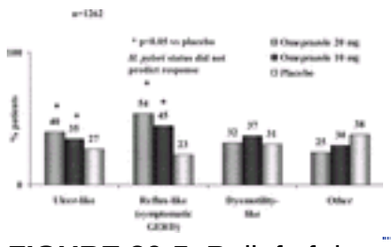


FIGURE 30-5. Relief of dyspepsia with a proton pump inhibitor versus placebo in nonulcer dyspepsia according to dyspepsia subgroup. *Ulcer-like,* predominant epigastric pain; *dysmotility-like,* nonpainful predominant symptom; *reflux-like,* symptomatic reflux. (Data from ref. [239].)

Cytoprotection Sucralfate stimulates mucosal prostaglandin synthesis and release of cytokines and has cytoprotective properties in experimental models. [241] One placebo-controlled study [242] reported significant improvement of symptoms in 77% of patients treated with sucralfate, versus 56% of patients in the placebo group. An open study that directly compared sucralfate with ranitidine [243] noted superior symptom improvement in patients treated with sucralfate. Other studies of NUD failed to detect a significant effect of sucralfate compared with placebo. [243], [244] Whether this drug is truly superior to placebo remains uncertain. Rebamipide, a cytoprotective drug available in the Far East, was not superior to placebo in NUD. [223] Misoprostol is also not efficacious in NUD. [245]

Prokinetic Drugs Two classes of prokinetic drugs have been widely used in NUD: the dopaminergic receptor blockers metoclopramide and domperidone, and cisapride, a 5-HT_4 agonist that also has 5-HT_3 antagonist actions. Indeed, many studies have demonstrated significant improvement of symptoms during treatment with these prokinetic agents, [84], [230], [233], [246], [247] although not all the results have been positive. [246], [248], [249] A comparison of cimetidine and cisapride demonstrated a significantly better response to therapy with the prokinetics, [250] and a trial comparing cisapride to metoclopramide favored cisapride. [251] There is reasonable evidence that cisapride is superior to placebo in NUD, [230], [233] but many of the published trials have had major design limitations, and a funnel plot suggested that the apparent beneficial effects of prokinetics may be the result of publication bias. [235] Large uncontrolled studies suggest that up to 80% of patients with uninvestigated dyspepsia respond to cisapride, but patients responding do not appear to be characterized by a specific symptom profile. [252] The mechanisms by which prokinetics relieve symptoms is uncertain; alterations of gastric emptying have not been convincingly directly linked to an improvement of symptoms, but few studies have assessed gastric emptying before and after therapy. [246] The safety profile of the current prokinetics limits their use. Metoclopramide can induce side effects through its central antidopaminergic mechanism; these include dystonic reactions, drowsiness, increased prolactin levels, and rarely, particularly in the elderly, tardive dyskinesia. Cisapride has been withdrawn because of QT prolongation and life-threatening arrhythmias (including sudden death). Domperidone, available in Canada and Mexico, also increases prolactin levels and may be proarrhythmic. Tegaserod, a well-tolerated partial 5-HT_4 agonist with no cardiac toxicity, is being tested in NUD. Erythromycin, a motilin agonist that initiates phase III activity in the antrum and small bowel, [253] has not been evaluated in NUD, and the drug often induces nausea. A new motilinomimetic, ABT-229, was not superior to placebo in NUD patients with or without delayed gastric emptying, and higher doses caused worse outcomes. [254] The cholecystokinin receptor antagonists loxiglumide and dexloxiglumide accelerate gastric emptying and may be of benefit in NUD, but development of cholelithiasis is a potential limitation. [255], [256]

Fundus-Relaxing Drugs It has been suggested that fundic relaxation following a meal is impaired in a subset of patients with NUD who have early satiety. [96] Hence, drugs that relax the gastric fundus may have therapeutic potential in the syndrome. [97], [257] Administration of the 5-HT_{1A} receptor agonist sumatriptan has been shown to induce relaxation of the gastric fundus, permitting larger intragastric volumes to accumulate before thresholds for perception or discomfort are reached. [97] Other fundus-relaxing drugs include buspirone (a 5-HT_{1A} agonist), clonidine (an a-adrenergic agonist), and citalopram (a selective serotonin reuptake inhibitor). Randomized controlled trials are needed to confirm beneficial treatment effects, but preliminary data are promising. [257], [258]

Treatment Targeting *H pylori* Most of the earlier studies of the effects of *H pylori* eradication in patients with NUD had important methodological limitations (i.e., lack of randomization or lack of a placebo control, application of inadequate outcome measures, failure to eradicate infection, lack of adequate follow-up after therapy, or inadequate study power). [147], [259] More recent studies have assessed the long-term outcome of eradication therapy, but the results have been mixed despite greater methodological rigor. [260], [261], [262], [263], [264], [265], [266], [267], [268], [269], [270], [271] and [272] Three large, properly designed studies failed to demonstrate a significant benefit of *H pylori* eradication, [269], [271], [272] whereas one observed a small but statistically significant benefit of *H pylori* eradication (Fig. 30-6). [270] However, the latter study showed a remarkably low placebo response rate, perhaps because peptic ulcers actually developed in a high proportion of those with persistent *H pylori* infection during follow-up. Although follow-up endoscopy was not routinely undertaken, a duodenal ulcer was found in four of six symptomatic patients who failed *H pylori* eradication and underwent endoscopy. [270] The metaanalyses have produced contradictory conclusions; one suggested a small but significant effect of *H pylori* eradication therapy in NUD, [273] whereas another failed to detect a relevant benefit but was less comprehensive. [274] Long-term outcome studies are not available.

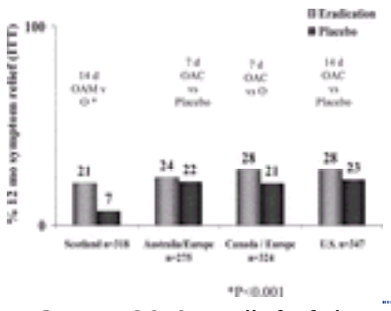


FIGURE 30-6. Relief of dyspepsia at 12 months after randomization to *H pylori* eradication therapy or a control group (omeprazole or placebo in four high-quality trials). *A,* amoxicillin; *C,* clarithromycin; *M,* metronidazole; *O,* omeprazole. (Data from the following trials: Scotland, [270] Australia/Europe, [271] Canada/Europe, [269] and the United States. [272])

H pylori eradication should be considered for patients with NUD. Perhaps 1 in 15 will be cured with just one course of therapy, [273] but most will not respond. The disadvantages include side effects of therapy, a lack of improvement after treatment, which is conducive to patient dissatisfaction, and the potential (albeit controversial) small risk for the development of esophagitis after treatment. These issues must be explained to and considered by any patient contemplating eradication therapy for NUD. [275]

Visceral Analgesic Drugs Agents that reduce gut visceral hypersensitivity are currently being assessed in NUD and IBS. [276] In two double-blinded, placebo-controlled trials, fedotozine (a peripheral ?-receptor agonist) was superior to placebo in relieving epigastric pain and nausea and registered better global symptom scores. [277], [278] Although the effect was statistically significant, the differences were numerically small, and the drug is no longer under study. In recent years, the potential role of serotonin receptors in modulating gut sensation has been recognized. [276], [279] 5-HT $_3$ receptors appear to play a role in the altered nociception that occurs during gastrointestinal inflammation, [280] and 5-HT $_3$ antagonists may modulate peripheral gut sensory nerve transmission. [281] One study observed a reduction of concomitant dyspepsia symptoms in patients with IBS during treatment with ondansetron, a 5-HT $_3$ antagonist. [282] Another trial, with alosetron, demonstrated a modest benefit in NUD over placebo. [283] However, the development of severe constipation and ischemic colitis in patients with IBS led to the withdrawal of alosetron. Leuprolide acetate, a gonadotropin-releasing hormone analog, has been shown in two randomized placebo-controlled trials to improve symptom scores during a 3-month period in women with severe functional GI complaints. [284], [285] Its mode of action is unknown, but ovarian hormone release is reduced, and such hormones may modulate GI smooth muscle function. The role of the gonadotropin-releasing hormone analogs remains to be established; moreover, their inconvenient mode of administration as well as significant side effects resulting from chemical castration is likely to restrict their application substantially.

Antispasmodic Drugs Because pyloric or antral spasm has not been documented in NUD, it is not surprising that neither dicyclomine nor trimebutine was more efficacious than placebo in small crossover studies. [230] Anticholinergic agents combined with a benzodiazepine have not been tested in NUD; such drugs should be used sparingly if at all because of their potential for habituation.

Antinauseant Drugs Antinausea drugs include the prokinetics and 5-HT $_3$ antagonists (discussed above) and the antihistamines and phenothiazines (e.g., prochlorperazine). Benzodiazepines may also help reduce nausea by their sedative effects. Although the therapeutic gain with nonspecific antinauseants has not been formally tested in patients with NUD, the histamine H $_1$ antagonists dimenhydrinate and cyclizine decrease gastric dysrhythmias and may be worth a trial. [286]

Antidepressant Drugs The data on the efficacy of antidepressants and other psychotropic agents in NUD are very limited. However, in one placebo-controlled trial, the antidepressant mianserin (a combined 5-HT $_2$, 5-HT $_3$, and a $_2$-adrenergic antagonist) was superior to placebo in a heterogeneous group of patients with functional gastrointestinal disorders. [287] Furthermore, the tricyclic antidepressant imipramine at a relatively low dose of 50 mg daily was clearly superior to placebo in patients with noncardiac chest pain. [288] A study of seven patients reported significantly less severe symptoms after 4 weeks of treatment with amitriptyline, but interestingly, the symptom improvement during treatment was not associated with a normalization of the perceptual responses to gastric distention. [289] In a metaanalysis, tricyclic antidepressants were of benefit in IBS (including NUD). [290] There are as yet no data on the effects of selective serotonin reuptake inhibitors (SSRIs) in patients with NUD. [291] Regarding the use of antidepressants, it is important to note that tricyclics (but not the SSRIs) appear to be effective at doses well below those currently used for the treatment of depressive disorders, and they appear to be effective in NUD, even in patients without obvious psychiatric abnormalities. In patients with symptoms resistant to standard therapy, a trial of a low-dose tricyclic antidepressant or an SSRI is justified.

Complementary and Alternative Medicine Limited placebo-controlled trials have reported improvement of symptoms in NUD and IBS during treatment with herbal preparations. [292], [293] The findings of these studies must be confirmed independently. Major problems of alternative medicines are related to a lack of standardization of the compounds, the inclusion of multiple potentially active extracts, and lack of knowledge about their long-term safety and precise mechanisms of action. Acupuncture and acupressure can reduce chemotherapy-induced and postoperative nausea and vomiting but have not been tested in NUD. [2] The value of gastric pacing in NUD is also unclear. [2]

Psychological Therapies Few controlled trials have evaluated the efficacy of psychological therapies in NUD. Haug and colleagues [294] observed significantly greater improvement of symptoms in patients treated with cognitive psychotherapy than in a control group that received no specific treatment. In a randomized trial of patients with NUD receiving either psychodynamic interpersonal psychotherapy or supportive therapy, small but significant advantages were observed in the psychotherapy arm compared with the controls in both the gastroenterologists' and the patients' total symptom scores; 1 year after treatment, the symptom scores were similar (Fig 30-7). [295] Another trial suggested that hypnotherapy may be efficacious in NUD. [296] However, trials of psychological treatment have generally been suboptimal, and direct comparisons with pharmacological therapies (including cost-effectiveness data) are lacking. [297]

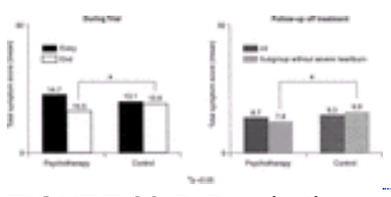


FIGURE 30-7. Psychotherapy in functional dyspepsia: results from a randomized controlled trial of 95 patients who failed conventional therapy at the end of treatment and 12 months later. Psychotherapy was superior to supportive therapy at the end of treatment, but scores 1 year after therapy were similar (except in a subgroup analysis of those without severe heartburn). (From ref. [295].)

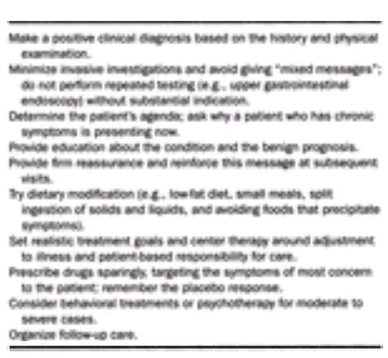
Treatment of Aerophagy Treatment of excessive unconscious air swallowing is difficult. Stress reduction (e.g., relaxation or cognitive-behavioral therapy) and dietary modifications (avoiding sucking on sweets or chewing gum, eating slowly, encouraging small swallows at meal time, and avoiding carbonated beverages) sometimes may help. Simethicone and activated charcoal are not of established value. Tranquilizers or prokinetics sometimes may be of benefit. [1]

Management Guidelines

A management algorithm is shown in Figure 30-8.

FIGURE 30-8. Management algorithm for patients presenting with new-onset dyspepsia who have not been previously evaluated. *GERD,* gastroesophageal reflux disease; *Hp, Helicobacter pylori*; *IBS,* irritable bowel syndrome; *NSAID,* nonsteroidal antiinflammatory drug; *PPI,* proton pump inhibitor. (Modified from American Gastroenterological Association medical position statement. Gastroenterology 1998;114:579.)

Patients with New-Onset (Uninvestigated) Dyspepsia The medical history is important. If typical reflux symptoms, heartburn, or acid regurgitation is the predominant complaint, a diagnosis of gastroesophageal reflux rather than NUD should be made and appropriate treatment instituted. A short trial of high-dose PPI therapy may be useful as a diagnostic test. Similarly, if bowel dysfunction is linked directly to the epigastric pain or discomfort, a diagnosis of IBS should lead to reassurance, explanation, dietary modification, and selective pharmacological therapy or stress management, which constitute the mainstay of treatment for patients with IBS (see Chapter 86). Patients older than 45 years with unexplained dyspepsia and those with alarm symptoms should undergo prompt upper GI endoscopy, and management should be based on the findings. In patients without alarm features, management should depend on the degree of uncertainty that both patient and physician are willing to accept. The use of a locally validated noninvasive *H pylori* test and initiation of anti– *H pylori* treatment in infected subjects constitute a reasonable initial approach, unless *H pylori* is uncommon in the background population. [215] Anti– *H pylori* therapy should relieve symptoms in most patients with peptic ulcer and substantially eliminate the ulcer diathesis. Noninvasive testing, however, may result in an inappropriate number of subjects being unnecessarily treated. Moreover, symptoms may recur even in patients cured of peptic ulcer disease [212]; reflux disease, for example, may be unmasked in some patients. [275] No agent has been approved by the Food and Drug Administration for use in NUD. If empiric therapy is considered in the uninvestigated patient who is negative for *H pylori,* the major first-line drugs remain histamine H_2 blockers or, increasingly, PPIs. Patients who fail to respond within 8 weeks or who rapidly relapse may require endoscopy. In patients with documented NUD after endoscopy, a positive clinical diagnosis and firm reassurance remain the key steps in management, especially if the principles of management outlined in Table 30-5 are adhered to.



Make a positive clinical diagnosis based on the history and physical examination.
Minimize invasive investigations and avoid giving "mixed messages"; do not perform repeated testing (e.g., upper gastrointestinal endoscopy) without substantial indication.
Determine the patient's agenda; ask why a patient who has chronic symptoms is presenting now.
Provide education about the condition and the benign prognosis.
Provide firm reassurance and reinforce this message at subsequent visits.
Try dietary modification (e.g., low-fat diet, small meals, split ingestion of solids and liquids, and avoiding foods that precipitate symptoms).
Set realistic treatment goals and center therapy around adjustment to illness and patient-based responsibility for care.
Prescribe drugs sparingly, targeting the symptoms of most concern to the patient; remember the placebo response.
Consider behavioral treatments or psychotherapy for moderate to severe cases.
Organize follow-up care.

Peyrin-Biroulet L.

TABLE 30-5 Management Principles in Nonulcer Dyspepsia

Patients Who Fail to Respond Failure to respond to treatment may raise two questions: Is the diagnosis of NUD correct, and was the treatment chosen appropriate? If a diagnostic workup has not been done, testing (including upper GI endoscopy and occasionally other investigations as outlined above) should be performed. Once structural lesions have been definitely excluded, treatment should be adjusted (e.g., a short trial of high-dose acid suppression, with dose reduction if successful). Combination therapy (e.g., an antispasmodic plus an antisecretory agent) or simethicone or sucralfate is not established as beneficial but may be worth a trial. If early satiety is a prominent symptom, consider a trial of buspirone, sumatriptan, or clonidine. Another option is to initiate low-dose treatment with a tricyclic antidepressant or an SSRI in full dose. With such therapy, it is important to inform the patient about potential side effects to ensure compliance.

Patients with Recurrent or Relapsing Symptoms In most patients, symptoms improve after an initial course of treatment. Because NUD and related functional disorders are characterized by chronic relapsing symptoms, it is highly probable that symptoms will recur after the end of treatment. No long-term strategy has been tested adequately in these patients. If symptoms have responded initially to a specific treatment but recur (usually after weeks or even months), another course of the same treatment is justified. Some patients have frequent symptom relapses that affect their quality of life, and in such cases, intermittent drug treatment can be helpful. Alternatively, consider prescribing on-demand therapy (self-directed therapy only when symptoms develop); this strategy has not been tested in NUD but is successful in nonerosive GERD. [298] The patient also should be advised to return for reevaluation if the pattern of symptoms changes or the symptoms do not respond to therapy.

Intractable Nonulcer Dyspepsia A small group of patients, usually concentrated at tertiary referral centers, do not respond to therapeutic measures, and their quality of life is affected considerably. [299] Rare causes of dyspepsia must be excluded, but once a comprehensive evaluation has been undertaken, repeated diagnostic testing should be avoided because this undermines patient confidence. The patient with intractable symptoms can benefit from an ongoing relationship with a physician who demonstrates a commitment to the patient's well-being. The physician should set realistic goals. For example, although improvement in functional status and quality of life is usually possible, complete symptom relief is usually not, and the patient must be made aware of the realities. The patient should be encouraged to become a partner in all management decisions. Regular brief visits will provide useful psychosocial support. It is important to rule out comorbid major psychiatric disease. Specific behavioral therapy (e.g., psychotherapy or cognitive-behavioral therapy) can reduce anxiety levels and encourage health-promoting behaviors in patients with intractable complaints, giving them greater control over their symptoms. In patients who require continuous therapy, drug holidays should be used to confirm that the therapy is still of value.

PROGNOSIS

A firm diagnosis of NUD after endoscopy is generally a safe one; few patients are later found to have an ulcer or other structural disease. Population-based studies have demonstrated a considerable turnover of subjects with abdominal symptoms; up to one third may lose their symptoms for years. [36] Patients with NUD seen at tertiary centers represent a subgroup with relatively intractable symptoms, and in this group, evidence that spontaneous remissions occur is only anecdotal. Whereas treatment may relieve symptoms for a limited period, cure cannot be expected.

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CHAPTER 31

Ray E. Clouse

APPROACH TO THE PATIENT WITH DYSPHAGIA OR ODYNOPHAGIA

SYMPTOM DEFINITIONS

Dysphagia and odynophagia are common complaints that usually merit prompt investigation and management. The term *dysphagia,* derived from Greek roots, literally means to “eat badly” or “with difficulty.” In common practice, dysphagia has become an umbrella term encompassing the sensations (short of pain) associated with abnormal bolus transit from mouth to stomach, as well as other signs or symptoms accompanying abnormal transit. In a sense, dysphagia has syndromic overtones. The term *odynophagia,* from Greek roots meaning to “eat with pain,” refers more specifically to pain during any component of the swallowing process. The two terms are not synonymous and may reflect different underlying disorders, yet they have overlapping differential diagnoses.

Dysphagia is represented by two principal types: oropharyngeal dysphagia and esophageal dysphagia. The differentiation is more than semantic; this convenient segregation reflects differences in pathophysiology, differential diagnosis, investigation, and management. For the purposes of this chapter, the initiation of swallowing is considered to occur at the pharyngeal phase, when velopharyngeal closure, laryngeal elevation and closure, opening of the upper esophageal sphincter, tongue loading and pulsion, and pharyngeal clearance result from highly coordinated central events. Bolus preparation and movement during the oral phases of swallowing are discussed in less detail. Abnormalities preceding the pharyngeal phase can be detected by bedside evaluation and radiologic imaging, if required.

Patients with *oropharyngeal dysphagia* typically describe food lodging or sticking in the back of the throat or cervical esophageal region and accurately localize the site of swallowing error as demonstrated radiologically. [1] The symptom may be perceived as low as the level of the suprasternal notch, but oropharyngeal dysphagia rarely is referred to more distal locations. [2] Hesitation with swallowing, frequent and repeated attempts at swallowing, and throat clearing may accompany the dysphagia. Not only does the patient sense or demonstrate abnormal pharyngoesophageal transit, but tracheobronchial aspiration may occur (with coughing or choking during or after swallows), reflecting a failure of coordination of bolus transit and laryngeal closure. Related symptoms include a rough or dysphonic voice after eating; hoarseness may reflect the underlying neuromuscular disorder. Poor neuromuscular coordination or high pressure within the bolus from mechanical obstruction can disrupt protection of the nasopharynx and result in nasopharyngeal regurgitation. Consequently, the spectrum of symptoms associated with oropharyngeal dysphagia can be dramatic and severe. These associated symptoms often are absent, and characteristics of the dysphagia itself commonly are used for establishing the diagnosis. Such characteristics, besides location of the symptom, include onset of dysphagia within 1 second of swallowing, inability to swallow any liquids or solids once a food bolus is lodged, and expectoration rather than regurgitation of the bolus. [3]

Esophageal dysphagia reflects disorders of the esophageal body and esophagogastric junction as well as anatomic areas abutting these regions, such as the gastric cardia and mediastinum. Obstructing lesions in the proximal esophageal body can produce symptoms mimicking oropharyngeal dysphagia; more distal processes produce esophageal dysphagia with distinctive characteristics. The most telling feature differentiating esophageal from oropharyngeal dysphagia is the sensing of abnormal bolus transit at a retrosternal site. Unfortunately, distal esophageal processes can refer symptoms proximally to the cervical esophageal region and suprasternal notch in nearly 30% of cases. [2], [4], [5] This fact alone is responsible for most of the clinical difficulty in segregating types of dysphagia by means of historical features. In contrast to oropharyngeal dysphagia, esophageal dysphagia is not immediate, typically is not associated with tracheobronchial aspiration, and is more likely to allow further ingestion of liquids to alleviate the dysphagia sensation. Additionally, patients with esophageal dysphagia are more likely to try repeated or forceful swallowing to dislodge or advance food boluses. When bolus impactions occur, patients with esophageal dysphagia regurgitate foamy, bland secretions or ingested liquids that have been retained above the impacted food.

Both dysphagia and odynophagia occur with or shortly after the initiation of a swallow. Pain and discomfort in the neck or retrosternal region present between swallows occur through different mechanisms and reflect an expanded differential diagnosis. The *globus sensation,* a sense that something is lodged continuously in the throat, must be differentiated from dysphagia or odynophagia before an improper investigation is undertaken. [6] The globus sensation typically is sensed in the midline at the laryngeal level, but it can lateralize in as many as 20% of patients. [7] The sensation classically is reported as a “lump in the throat,” but a feeling that a foreign body, sharp object, or food particle is lodged also is a compatible description. [6] Most notably, the globus sensation does not interfere with swallowing; although 20% of patients note something abnormal during food swallows, the original symptom abates during the process. [8] Globus may accompany a variety of disorders also producing dysphagia, such as gastroesophageal reflux disease (GERD) and distal esophageal motility disorders, and up to 45% of the general population may have intermittent symptoms resembling globus. [9] The persistent symptom, however, most often reflects a functional gastrointestinal disorder. [10], [11] A summary of the features differentiating functional globus from oropharyngeal dysphagia is provided in Table 31-1. *Xerostomia* also can be confused with oropharyngeal dysphagia if the medical history is not obtained carefully.

TABLE 31-1 Features Distinguishing Oropharyngeal Dysphagia and Functional Globus

MECHANISMS RESPONSIBLE FOR SYMPTOM PRODUCTION

Sensory information from the pharyngeal and very proximal esophageal regions is carried by cranial nerves V, X, and XI. Both discomfort and pain can be elicited from the posterior pharynx and hypopharynx by noxious mucosal stimulation. Pressure on the pharyngeal surfaces is readily perceived, and minor alterations in bolus pressure dynamics may be sufficient to trigger dysphagia in some conditions. Proximal esophageal distention just distal to the upper esophageal sphincter results in upper sphincter hypertonicity, but symptoms have not been linked directly to this motor response. [12] The nasal and tracheobronchial symptoms associated with the oropharyngeal dysphagia syndrome are provoked by noxious stimulation of cranial nerve V innervating nasal passages and activation of the cough reflex, respectively. Little is known about the perception of oropharyngeal dysphagia in patients with normal swallowing dynamics.

Esophageal sensation occurs via vagal and spinal afferent pathways modulating motor activity, although noxious stimuli are carried almost exclusively through spinal afferents. [13], [14] At the spinal level, afferents from each esophageal region are widely distributed and overlap innervation from other organs, such as the heart. [15] This convergence of input contributes to the similarity in the presentations of esophageal and cardiac symptoms. [16] Fine perception of abnormal bolus movement normally is not present in the esophageal body, and retention of a nonobstructing solid bolus in the distal esophagus is poorly recognized. [17] As further evidence, capsule-shaped pH transmitters can be attached to the distal esophageal mucosa and produce few symptoms. [18] Consequently, factors beyond bolus retention must be necessary for dysphagia in most cases. Distention of esophageal regions proximal to a lodged bolus may be one important factor. Increased intraesophageal pressure encountered in achalasia before the esophagus becomes markedly dilated may stimulate the sensation of dysphagia. [19] Motility alterations in the esophageal body in response to transient obstruction also may have a sensory counterpart, but it is difficult to attribute dysphagia to specific esophageal body motor patterns alone. Both hypomotility and hypermotility ranging from mild to marked are encountered during the routine manometric evaluation of patients not reporting dysphagia at the time of the investigation. The exception is the achalasia patient, whose intraesophageal pressure is increased during the study. [19], [20]

In some instances, esophageal dysphagia is produced by mucosal damage. Mucosal inflammation may be partly responsible for the nonobstructive dysphagia seen with GERD. [21] Similarly, mucosal damage overlying strictures or tumors may augment sensation through the effects of inflammation on visceral sensitivity. [22] In patients who have rings or strictures with little or no associated mucosal injury, dysphagia occurs when food boluses completely and transiently obstruct. Symptoms are corroborated radiographically with brief impaction of radiopaque solid boluses in the narrowed region. Symptoms are alleviated with dissolution of the bolus, and dysphagia is not reproduced with liquids or solids that pass freely. Consequently, with many esophageal abnormalities producing dysphagia, symptoms do not occur until the lumenal diameter is less than 13 mm, a size at which conventional food boluses (e.g., bread, meat) become at least transiently impacted. [23] Persistent obstruction from food bolus impactions produces additional uncomfortable symptoms arising from proximal esophageal regions.

Sensitivity to these mechanisms varies considerably from patient to patient. The variation in part may reflect hypersensitivity or hyposensitivity accompanying specific conditions or disease processes. [13] Aging, connective tissue diseases (particularly those overlapping with scleroderma), diabetes, and other conditions associated with esophageal hypomotility are associated with hyposensitivity to experimental stimuli, such as acid instillation and balloon distention. [13], [24], [25] and [26] Although it has not been established, such patients may be poorly sensitive to transit abnormalities and underreport dysphagia. Likewise, as the esophagus develops secondary hypomotility and dilates in response to chronic obstruction, the sense of dysphagia dissipates in compensation. In contrast, hypersensitivity to acid and distention stimuli in the functional esophageal syndromes and spastic disorders may help explain the esophageal dysphagia occurring in these conditions. [27], [28] A sense of abnormal transit is reported, yet convincing evidence of an obstructing lesion or mucosal injury often is lacking. [6]

Odynophagia only occasionally accompanies dysphagia and appears to occur through separate mechanisms. With the exception of pain accompanying acute esophageal obstruction, odynophagia requires afferent input not resulting from abnormal bolus transit alone. In the esophageal phase of swallowing, odynophagia may require direct invasion or irritation of intramural nerves, as occurs with malignancies or deep inflammatory processes. Sensory stimulation resulting from some forms of esophagitis also can be responsible for this symptom (e.g., caustic or infectious esophagitis).

DIFFERENTIAL DIAGNOSES

The approach to the patient with dysphagia or odynophagia begins with a consideration of the differential diagnoses. Diagnostic possibilities vary markedly between oropharyngeal and esophageal dysphagia, further emphasizing the importance of characterizing dysphagia by type.

Oropharyngeal Dysphagia

Symptoms of abnormal transit in any part of the gut can be produced by disturbed motor function or by structural lesions. Oropharyngeal dysphagia is a manifestation of motor dysfunction rather than of structural disease specific to the oropharynx or its neighboring structures in a ratio of approximately 4:1. [29] Motor dysfunction results from a variety of neurological, striated muscle, and systemic disorders (Table 31-2). Structural lesions are less commonly responsible for oropharyngeal dysphagia, but they can be intrinsic or extrinsic to the pharyngoesophageal region—indenting, invading, or encircling the structures. Many of the responsible disorders listed in Table 31-2 have additional manifestations that assist in limiting the differential diagnosis.

TABLE 31-2 Causes of Oropharyngeal Dysphagia

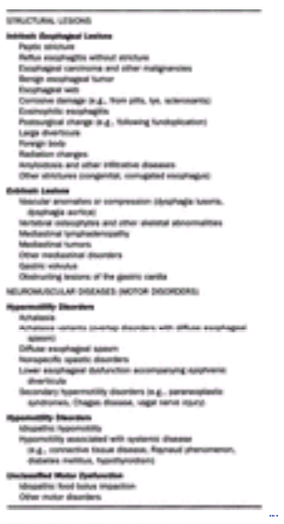
Despite the range of diagnostic possibilities, most patients presenting with oropharyngeal dysphagia have had a cerebrovascular accident—Parkinson disease being responsible for much of the remainder. [30] Either clinical or videofluoroscopic evidence of swallowing dysfunction can be found in more than 50% of patients after acute stroke, and chest infection (implying tracheobronchial aspiration) occurs in nearly a third. [31], [32], [33] and [34] Dysphagia typically dissipates during the 6 months following the event, but symptoms and signs of oropharyngeal dysfunction persist in 10% to 15% and gradually appear in a small percentage initially free of dysphagia. [35] Laryngopharyngeal sensory deficits accompanying motor dysfunction and thereby impairing the appropriate response to aspiration may be factors in the poor outcome of stroke victims with oropharyngeal dysphagia. [36] Some patients with documented swallowing dysfunction have none of the classical symptoms of oropharyngeal dysphagia, and pulmonary or nutritional complications dominate because of diminished sensation from nerve damage or a reduced level of consciousness.

Pharyngolaryngeal cancer surgery is an important structural cause of difficult oropharyngeal dysphagia. Symptoms result from obstruction at the pharyngoesophageal junction or reduced tongue driving force, in the case of resection at the base of the tongue. [37] The importance of the tongue as a pressure generator in the process of swallowing is highlighted in these patients. The esophagus is anchored at the cricoid cartilage, and osteophytes at the fourth through seventh cervical vertebrae can sufficiently impede normal physiology to produce dysphagia. [38] This degree of advanced osteophyte formation is typical of diffuse idiopathic skeletal hyperostosis, or Forestier disease, a disorder that affects 12% of the population and can manifest clinically as dysphagia. [39], [40] Careful documentation with videofluoroscopy of the role of skeletal abnormalities in altering swallowing dynamics and bolus transit is necessary before the symptom can be attributed to this condition. [39], [41]

Esophageal Dysphagia

In contrast to oropharyngeal dysphagia, wherein motility disorders top the differential diagnosis, esophageal dysphagia is most often caused by structural lesions. As with oropharyngeal dysphagia, the range of possibilities is broad (Table 31-3). Esophageal strictures associated with GERD are common, increase in frequency with age, and are not associated with a significant heartburn history in 25% of patients. [42] Nonobstructive dysphagia is reported to some degree in nearly a third of patients

undergoing preoperative evaluation for antireflux surgery. [43] Distal esophageal rings, particularly mucosal rings at the squamocolumnar junction (Schatzki rings), also are common; some indentation of the barium column is detected at this level during radiographic evaluation in as many as 14% of individuals. [23], [44] Malignant esophageal neoplasms typically present with dysphagia. The diagnosis of esophageal adenocarcinoma is rapidly increasing, particularly in middle-aged white men. Other structural explanations listed in the table are uncommon; vascular anomalies are rare. [45], [46] and [47] The ability of proximal gastric disorders (e.g., gastric volvulus) to produce this presentation should be considered in the appropriate setting when the cause of esophageal dysphagia is unclear. [48]



STRUCTURAL LESIONS
Intrinsic Esophageal Lesions
Peptic stricture
Reflux esophagitis without stricture
Esophageal carcinoma and other malignancies
Benign esophageal tumor
Esophageal web
Corrosive damage (e.g., from pills, lye, sclerosants)
Eosinophilic esophagitis
Postsurgical change (e.g., following fundoplication)
Lesga diverticula
Foreign body
Radiation changes
Amyloidosis and other infiltrative diseases
Other strictures (congenital, corrugated esophagus)
Extrinsic Lesions
Vascular anomalies or compression (dysphagia lusoria, dysphagia aortica)
Vertebral osteophytes and other skeletal abnormalities
Mediastinal lymphadenopathy
Mediastinal tumors
Other mediastinal disorders
Gastric volvulus
Obstructing lesions of the gastric cardia
NEUROMUSCULAR DISEASES (MOTOR DISORDERS)
Hypermotility Disorders
Achalasia
Achalasia variants (overlap disorders with diffuse esophageal spasm)
Diffuse esophageal spasm
Nonspecific spastic disorders
Lower esophageal dysfunction accompanying epiphrenic diverticula
Secondary hypermotility disorders (e.g., paraneoplastic syndromes, Chagas disease, vagal nerve injury)
Hypomotility Disorders
Idiopathic hypomotility
Hypomotility associated with systemic disease (e.g., connective tissue disease, Raynaud phenomenon, diabetes mellitus, hypothyroidism)
Unclassified Motor Dysfunction
Idiopathic food bolus impaction
Other motor disorders

TABLE 31-3 Causes of Esophageal Dysphagia

Esophageal dysmotility can be classified in two predominant categories, hypomotility and hypermotility (Fig. 31-1). [13], [27] Hypomotility of either the smooth muscle esophageal body (also called *ineffective esophageal motility* [49], [50]) or lower esophageal sphincter (LES) represents failure of the contractile mechanisms through neurogenic or myogenic processes. The outcome is feeble or absent esophageal peristalsis and loss of resting tone in the LES. Systemic disorders associated with hypomotility have been mentioned previously, but most hypomotility is idiopathic. [51], [52] These processes predispose to GERD, and impairment of bolus clearance often is conspicuous. Nevertheless, dysphagia typically is not attributed to hypomotility, even when severe, without careful exclusion of structural explanations that include reflux esophagitis.

FIGURE 31-1. A method of categorizing dysmotility in the distal esophagus and lower esophageal sphincter (LES) based on the principal type of motor dysfunction. Hypermotility may result from inhibitory nerve deficiency or an imbalance between inhibitory and contractile influences. Some patients have a mixture of hypomotility and hypermotility features and cannot be classified solely into one branch of the scheme. (From ref. [27].)

In contrast, hypermotility results from deficient inhibitory nerve influence. [53] Nonpropulsive, uncoordinated contractions disrupt bolus transit; incomplete LES relaxation produces functional obstruction and dysphagia. Consequently, achalasia, diffuse esophageal spasm, and related patterns of severe hypermotility belong in the differential diagnosis (see Fig. 31-1). [13], [27] A variety of nonspecific spastic disorders, including high-amplitude peristaltic contractions (“nutcracker esophagus”), other vigorous contraction wave abnormalities, and isolated hypermotility features of the LES (increased basal pressure or incomplete relaxation), have been found in patients with otherwise unexplained dysphagia. [27], [54], [55] Transit disruption is less common with these findings, and dysphagia may be related to the hypersensitivity that often accompanies the disorders. [27] Spastic disorders typically have no underlying pathological explanation, and their presence neither precludes a functional diagnosis for the presenting complaints nor excludes the possibility that associated psychological factors are contributing to symptoms. [6], [27], [56], [57] Nevertheless, because esophageal dysphagia can represent serious structural explanations, a thorough investigation is required before a functional diagnosis is assigned. Less classified motor abnormalities also have been reported (e.g., lower esophageal muscular rings). [44], [58]

Odynophagia

Pain during the oropharyngeal phase of swallowing has been attributed to a variety of processes, particularly malignancies, foreign body ingestion, and mucosal inflammation and ulceration. Oropharyngeal odynophagia may indicate deeper invasion of sensory nerves or extrinsic structures, and a careful evaluation for structural lesions should be undertaken. Odynophagia occurring later in the swallowing process most commonly is caused by specific types of mucosal damage: caustic injury and infection. It also accompanies tumors and other processes associated with deep mural injury, such as radiation damage, deep peptic ulceration, and idiopathic ulcer (in immunosuppressed patients)—processes more likely to produce chronic or persistent symptoms. Odynophagia rarely results from uncomplicated reflux disease. Chest pain is reported by up to half of patients with achalasia at some period during the course of the illness. [59], [60] This pain typically is not directly related to swallowing, runs a course separate from that of dysphagia, is influenced in a limited fashion by treatment for achalasia, and usually is not confused with purer presentations of odynophagia. [60]

The most common causes of acute esophageal odynophagia in immunocompetent subjects are infection with *Candida albicans* or herpes simplex virus and pill-induced injury. [61], [62] Esophagitis with other infectious causes can have a similar presentation; these include bacteria, fungi, cytomegalovirus, and varicella-zoster virus. [63] In fact, the entire range of opportunistic esophageal infections encountered in immunosuppressed individuals may be responsible for this symptom (see Chapter 61). Whereas herpes simplex virus is the most common viral cause of esophagitis in immunocompetent subjects, cytomegalovirus is a more common offender in the immunosuppressed. [64] In this latter group, however, *Candida* remains the most common cause of infectious esophagitis. [63] Pill-induced caustic injury to the esophagus has been associated with more than 70 medications, including aspirin, potassium supplements, tetracycline, ferrous sulfate, quinidine, alendronate, and nonsteroidal antiinflammatory drugs. [62], [65], [66] Tissue injury to the esophagus from caustic medications can be deep, and odynophagia that becomes severe over 3 to 4 days is a common component of the presentation (see Chapter 63).

APPROACH TO THE PATIENT WITH DYSPHAGIA

Dysphagia is a common clinical problem. Nearly one in five subjects older than 50 years of age describes the symptom in epidemiologic surveys. [67], [68] Dysphagia is the second most common indication for endoscopy in the United States, [69] and a mechanism responsible for the symptom can be found in most cases, even those initially labeled psychogenic. [70], [71] Consequently, a systematic approach to the differential diagnosis that results in a well-planned investigation is important. A variety of algorithms have been recommended, but each depends on a carefully obtained medical history as the initial step. [72], [73], [74] and [75] The physical examination is more likely to provide useful information in patients with oropharyngeal dysphagia.

The Medical History and Physical Examination of the Patient with Dysphagia

Establishing the type of dysphagia, either oropharyngeal or esophageal, is of primary importance. Differentiating historical features have been outlined in the opening paragraphs of this chapter. Patients with oropharyngeal dysphagia may have symptoms of oral dysfunction that further support the diagnosis (e.g., poor handling of the food bolus with drooling and spillage, piecemeal swallowing, and dysarthria). When the type of dysphagia remains unclear, discerning the outcome after the ingestion of specific foods or liquids can be helpful. Asking for a description of recent meals and resulting symptoms may keep the patient focused on the specific events of swallowing. Additionally, offering the patient a small quantity of water to swallow during the interview may clarify the medical history through first-hand observation.

Certain features in the medical history can help narrow the differential diagnosis of oropharyngeal dysphagia. Proximal dysphagia for solid foods alone is suggestive of restrictive processes, such as strictures, webs, and tumors. Odynophagia in association with oropharyngeal dysphagia also has implications (see below). A sudden onset of symptoms suggests an acute neurological insult, and a short but progressive course typifies malignancy, whereas an insidious presentation reflects inflammatory processes, such as myopathies. Delayed expectoration is suggestive of a retaining pharyngeal diverticulum.

Historical evidence and physical findings corroborating a neurological, muscular, or systemic disease should be sought when oropharyngeal dysphagia is suspected, with the large differential diagnosis outlined in Table 31-2 taken into account. Inquiries should be made about any past history of cerebrovascular accidents or neurological disability that preceded or accompanied dysphagia, and a comprehensive neurological examination is essential (for a further elaboration of the physical findings accompanying oropharyngeal dysphagia of neurological causes, see Chapter 59). The medication history should be reviewed for agents that can either cause or exacerbate dysphagic symptoms, including those with adverse neuromuscular effects (e.g., sedatives, narcotics, muscle relaxants) or that cause xerostomia (e.g., anticholinergics, antihistamines, antidepressants). [76] The neck should be examined for masses, thyromegaly, or lymphadenopathy. The presence and degree of tracheobronchial aspiration (including aspiration pneumonia) should be extracted from the history to determine the severity of the dysphagia syndrome. Likewise, limitations of food intake and their impact on weight should be assessed based on the available information. Determining the need and urgency for nonoral feeding is a primary component of the oropharyngeal dysphagia evaluation. [77]

If esophageal dysphagia is suspected, the medical history can help narrow the differential diagnosis and direct the investigative approach. [78] Features or established diagnoses of systemic disorders associated with esophageal dysphagia (see Table 31-3), particularly connective tissue diseases and Raynaud phenomenon, should be sought. [79], [80] Historical aspects favoring structural diagnoses lead to early endoscopy, biopsy, and dilation. Features favoring a motility disorder may indicate barium radiography and manometry, either early in the evaluation or for completion of the workup. Although presentations of the disorders listed in Table 31-2 are quite variable from patient to patient, certain features of common diagnoses can be helpful in establishing the initial impression (Fig. 31-2). [78] Structural esophageal lesions typically result in dysphagia after ingestion of solid foods but not after liquids alone. Bulky foods of larger caliber, such as meats and bread, more reliably reproduce the symptom. In contrast, precipitants of dysphagia from esophageal motility disorders are more variable.

FIGURE 31-2. Features of the medical history that help focus the differential diagnosis of common causes of esophageal dysphagia.

At least 95% of patients with achalasia have dysphagia, but the course frequently is nonprogressive. [59] This may leave a large number of patients without a diagnosis, an undesirable outcome in light of the treatment options available for this disorder. Because heartburn also can be reported by achalasia patients, some are followed for years with a presumptive diagnosis of GERD before the correct diagnosis is established. [81], [82] Conventional investigative tests such as endoscopy or barium radiography were not helpful prospectively in as many as a third of achalasia patients eventually presenting for manometry, with which the diagnosis was secured. [82] A careful medical history can enhance suspicion of achalasia. Dysphagia typically is for liquids as well as solid foods, although one third of patients may report dysphagia for solid foods alone. Regurgitation of esophageal contents is reported by 60% to 90% of subjects, a feature that can be differentiated from gastroesophageal regurgitation by the bland, nonacidic characteristics of the regurgitated material and the ability of the patient to reswallow the bolus without aversion. Maneuvers performed by the patient during meals to increase intraesophageal pressure and enhance esophageal emptying frequently can be elicited (e.g., straightening the back, raising the arms above the head, and forceful swallowing). [13], [59] The ability to retain and regurgitate large quantities of ingested food or foamy saliva from a dilated esophagus also characterizes achalasia.

Approach to the Patient with Oropharyngeal Dysphagia

The evaluation of oropharyngeal dysphagia has three important components: (1) determining the structural and physiological abnormalities responsible for symptoms, (2) discovering the underlying disorders (including neurological and muscular disorders) responsible for these findings, and (3) evaluating the safety and practicality of oral feeding. [77], [83] The second of these is largely determined from the history and physical findings, as described previously, in conjunction with appropriate laboratory investigation. The other two are most commonly and effectively accomplished by following a protocol for videofluoroscopic swallowing evaluation—referred to as a *modified barium swallow*. [1], [73], [84] The examination is structured to detect and analyze functional impairment of the swallowing mechanism and provides evidence for each of the four categories of oropharyngeal swallowing dysfunction: (1) inability to initiate or excessive delay in the initiation of pharyngeal swallowing, (2) aspiration of the ingestant, (3) nasopharyngeal regurgitation, and (4) residue of ingestant within the pharyngeal cavity after swallowing. [73] Because it is widely available, characterizes swallowing dysfunction well, and can examine the short-term effects of swallowing interventions (compensatory or corrective swallowing strategies, diet modifications), the modified barium swallow presently is considered the most applicable initial test in patients with a compatible presentation. [71]

Videofluoroscopy also can detect structural abnormalities potentially responsible for symptoms, but it is less satisfactory than endoscopic evaluation. Transnasal fiberoptic pharyngoscopy/laryngoscopy (nasoendoscopy) is the optimal method for the direct inspection of mucosal surfaces in the region, and when combined with a swallowing assessment protocol, it provides at least indirect evidence of all categories of swallowing dysfunction except possibly nasopharyngeal regurgitation. [85], [86], [87] and [88] Nasoendoscopy or indirect laryngoscopy typically is performed early in the evaluation of oropharyngeal dysphagia and is especially important if the symptoms suggest a mucosal lesion. Nasoendoscopy also can be used to assess sensory function by applying pulses of air to the mucosa innervated by the superior laryngeal nerve and evoking glottic adduction. [88], [89] and [90] Thus, in centers where nasoendoscopy combined with a swallowing assessment can be performed, the approach may provide much of the same information as videofluoroscopy and has the unique advantages of portability, repeatability without radiation exposure, and technical success in patients with a reduced sensorium. [87], [91], [92] This approach is less effective than videofluoroscopy in determining the cause of swallowing dysfunction, and nasoendosopy does not obviate the need for videofluoroscopy in all subjects—providing merit to each test in clinical practice. In contrast, manometry has contributed little to the routine evaluation of the patient with oropharyngeal dysphagia despite its capacity to quantify contraction strength. [93], [94] It is complementary to videofluoroscopy when the two are performed concurrently, and this approach or possibly even manometry alone can be helpful in differentiating obstruction at the level of the upper esophageal sphincter from weak pharyngeal propulsive forces. [95], [96]

In the systematic assessment of patients with oropharyngeal dysphagia, a specific diagnosis of the underlying cause will be detected in many instances, and the mechanism responsible for the symptom will be detected in nearly all. [71] Important therapeutic recommendations can arise from the evaluation, even when a specific diagnosis is not established. The algorithm shown in Figure 31-3 demonstrates how these outcomes are accomplished. If local lesions are detected by direct mucosal visualization, further study of swallowing mechanisms may not be required. The videofluoroscopic examination may uncover rings, webs, strictures, posterior pharyngeal diverticula, or tumors that are amenable to specific surgical or endoscopic interventions. However, oropharyngeal dysphagia should not be attributed to

some common radiologic findings without careful consideration of alternative explanations. Such findings include cricopharyngeal bars, prominent vertebral osteophytes, and lateral pharyngeal diverticula. The first of these is found in more than 15% of evaluated dysphagic patients, correlates poorly with the symptomatic state, and precludes detection of the true explanation for dysphagia in the vast majority if alternative explanations are not pursued. [97], [98] Some authorities believe that dysphagia should not be attributed to a cricopharyngeal bar unless no other explanation is available and coexistent pharyngeal dysfunction is present to accentuate the importance of the bar. [73] The swallowing evaluation also may reveal disorders responsive to cricopharyngeal myotomy or other, less invasive approaches that reduce upper sphincter pressure. [99] The success of myotomy is greatest in situations in which the cricopharyngeal opening is limited but pharyngeal function is preserved. [100] Examples include Zenker diverticulum, primary cricopharyngeal dysfunction, and postcricoid stenosis. Success in neurogenic dysphagia can reach 60%, but predictors of response are not readily available. [73] Reviews of this treatment in relation to specific underlying disorders should be consulted. [73], [101], [102]

FIGURE 31-3. Approach to the patient with oropharyngeal dysphagia. The modified barium swallow (videofluoroscopic swallowing evaluation) is a key element in the evaluation of most patients, providing useful information for both diagnosis and management. Nasoendoscopy accompanied by a swallowing protocol is an alternative method to reach similar end points. *EGD,* esophagogastroduodenoscopy. (Adapted from ref. [73].)

The detection of aspiration during videofluoroscopy is a predictor of pneumonia risk and future hospitalization, although false negatives also are common with this test in patients with stroke and other forms of neurogenic dysphagia. [33], [103], [104] Aspiration during nasoendoscopic swallowing assessment has similar predictive value, as does bedside evaluation examining oxygen desaturation with water swallowing. [91], [105] As a minimum, compensatory swallowing strategies are required, and oral feeding may be prohibited. If aspiration is demonstrated with all food consistencies despite compensatory maneuvers, nonoral feeding typically is instituted; this approach may not prevent subsequent aspiration pneumonia, however. [106], [107] and [108] The videofluoroscopic study or nasoendoscopy with swallowing evaluation also is used to direct specific compensatory or corrective swallowing strategies that may reduce the risk for aspiration. [91] Swallowing therapy techniques that have been employed are listed in Table 31-4, and of these, the evidence most strongly favors the efficacy of diet modifications in reducing the risk for subsequent aspiration pneumonia. [73], [109] Although many of the maneuvers listed in Table 31-4 affect the occurrence of aspiration during a brief swallowing evaluation, such as the modified barium swallow, their actual ability to reduce subsequent morbidity is less established. Nevertheless, potential benefits at low cost drive the recommendation for their use. [73], [77], [110] After all, the first-year mortality caused by aspiration pneumonia in patients with stroke approaches 20%, and subsequent annual mortality from this complication remains 10% to 15%. [30]

CATEGORY OF TECHNIQUE	SPECIFIC EXAMPLE
Swallowing maneuver	Supraglottic swallow
	Supersupraglottic swallow
	Effortful swallow
	Mendelsohn maneuver
Postural adjustment	Head tilt
	Chin tuck
	Head rotation
	Head rotation with extrinsic pressure on thyroid cartilage
	Lying on side, elevation
Facilitatory techniques	Strengthening exercises
	Biofeedback
	Thermal stimulation
	Gustatory stimulation
Dietary modification	Thickening liquids
	Thinning liquids

From refs. 1, 161, 162; table adapted from ref. 73.

TABLE 31-4 The Spectrum of Swallowing Therapy Techniques

Other interventions for oropharyngeal dysphagia depend on the specific diagnosis that is derived from this initial evaluation, and the reader should refer to discussions of the disorders at other locations in this text. Certain combinations of videofluoroscopic findings can help determine the underlying disorder correctly. [30] Some systemic or neurological disorders that produce oropharyngeal dysphagia can be treated, and treatment results in a reduction of swallowing symptoms, although this response is not uniform. For example, reduction in dysphagia from Parkinson disease is inconsistent with levodopa treatment despite improvement in other neurological features of the illness. [111], [112]

Approach to the Patient with Esophageal Dysphagia

Endoscopy has become the primary tool for investigating esophageal dysphagia, not only for its accuracy, but also for its therapeutic potential. [113] Barium radiography once was recommended as the initial test in all patients with dysphagia, but the popularity of this approach is waning. The safety of endoscopy, its high diagnostic yield, and its provision for mucosal biopsy and dilation of commonly encountered structural lesions favor its use over radiographic screening when esophageal dysphagia is suspected. Likewise, barium radiography is not sufficiently sensitive to the range of tissue damage resulting from GERD, a common cause of esophageal complaints. [114] Barium radiography with fluoroscopy does retain a role in the evaluation of esophageal dysphagia (Fig. 31-4). Strictures that can be overlooked at endoscopy, particularly those having a lumenal diameter of more than 10 mm, are detectable. [115], [116] and [117] Barium studies are more sensitive than endoscopy to distal rings and to achalasia and can fully establish the level of dysphagia if symptoms are reproduced during the study. [115], [118] The sensitivity of barium radiography for motor disorders and structural lesions is further enhanced if barium-impregnated solids are employed. [119], [120] Although the barium swallow may be required ultimately to establish a diagnosis, its benefits do not overshadow the utility of endoscopy in the typical case, and the American Gastroenterological Association in a practice guideline has recommended endoscopy as the initial investigative test in esophageal dysphagia. [113] Achalasia suspected from the outset is an exception. [113] Compatible radiographic findings can be confirmed by manometry, and endoscopy can be reserved until the treatment plan for newly established achalasia is designed (see Chapter 59).

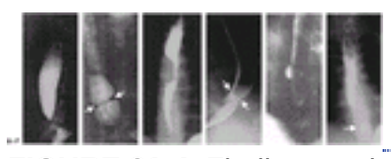


FIGURE 31-4. Findings on barium radiography can help establish the diagnosis in patients with esophageal dysphagia and unrevealing endoscopy. **A:** Absent peristalsis and barium retention above a closed lower sphincter suggest achalasia in this patient with minimal esophageal dilation. **B:** A lower esophageal (Schatzki) ring (*arrows*) causing intermittent dysphagia was not appreciated at endoscopy. **C:** Findings at endoscopy were completely normal in this patient with mediastinal adenopathy producing extrinsic compression of the midesophagus. **D:** A stricture with a lumenal diameter of 12 mm (*arrows*) was not appreciated on the initial

endoscopic evaluation. **E:** This epiphrenic diverticulum was overlooked at endoscopy, yet once discovered, it prompted manometric evaluation, and a severe distal spastic disorder was discovered. **F:** Endoscopy was unrevealing in a patient with a medical history confusing for oropharyngeal versus esophageal dysphagia, but a barium pill (12.5-mm diameter) lodged at the esophagogastric junction (*arrow*) reproduced symptoms and led to reevaluation and dilation.

Once structural explanations for esophageal dysphagia are excluded, the evaluation should proceed with esophageal manometry. [121] Manometry is sensitive to conventional hypermotility and hypomotility disorders but will not establish a conclusive diagnosis or have treatment implications in all patients with otherwise unexplained esophageal dysphagia—even when food bolus impactions have occurred. Nevertheless, manometry is indicated if only to detect the reasonable proportion of patients with achalasia who would be overlooked by other investigative tests. [82], [121] Manometry occasionally fails to establish the diagnosis of achalasia because of seemingly normal LES relaxation, arguably the most important diagnostic criterion; this artifactual error can be reduced if a sleeve sensor is used, if an averaged value is recorded for LES relaxation nadir pressure, or if high-resolution manometric methods are employed. [122], [123] and [124] Manometry also will detect spastic disorders, all of which may be associated with dysphagia. [27] Although the demonstration of a direct relationship between symptoms and these motor disorders often is less convincing, the findings may help direct therapeutic trials. [27]

Dysphagia to some degree is the most common complication of antireflux surgery, its occurrence predicted with any reliability only by the presence of preoperative dysphagia. [125] The technical skill of the surgeon is more important than the degree of existing hypomotility in producing this adverse outcome. [126] Early symptoms, even when severe (<5% of patients), may dissipate within 8 to 12 weeks after the operation with resolution of local inflammatory response. [127], [128] Periodic dilation with a 16- to 20-mm-diameter bougie or balloon may suffice. [128] Persistent dysphagia, occurring to at least mild degree in up to 24% of patients, [129] most often is related to suboptimal surgical technique. In cases requiring reoperation, transdiaphragmatic migration of the fundoplication, slipped or misplaced fundoplication, twisted fundoplication, or residual paraesophageal hernia can explain this symptom. [130] Physiological failure of the procedure with persistent reflux is reported following a small percentage of operations, and reflux-related explanations for dysphagia must be excluded.

Multivariate analysis of manometric features in postoperative dysphagia suggests that both the degree of obstruction and the degree of peristaltic competency participate in symptom production, both delaying transit across the lower sphincter. [131], [132] The last may be of particular importance. [132] In practice, endoscopy and barium videofluoroscopy are the principal evaluation tools, the emphasis being on detecting an abnormal postoperative appearance and on evaluating the degree of retention. [133] Manometry has a limited role unless used in conjunction with videofluoroscopy or high-resolution techniques, procedures that remain primarily investigational. [19], [132] Dilation for persistent dysphagia is effective in half of subjects when the fundoplication is intact; evidence of slipped fundoplication predicts a poor response, and reoperation can be avoided in some with pneumatic dilation. [129], [134]

Patients with suspected esophageal dysphagia who remain without a diagnosis following endoscopy, barium radiography, and esophageal manometry probably benefit little from additional testing. Ambulatory pH monitoring to define nonobstructive dysphagia from endoscopy-negative reflux disease may have a role that could be obviated with a therapeutic trial of antireflux treatment. When additional evaluation is desired, further efforts at reproducing dysphagia during videofluoroscopy with the use of barium-impregnated liquids or solids might be considered. Reproducing dysphagia in the absence of a transit abnormality helps establish a functional diagnosis and provides alternative therapeutic approaches.

Symptomatic Treatment of Dysphagia

A thorough investigation of dysphagia reveals a diagnosis in a large proportion of cases, and therapy is directed at the specific diagnosis. In some instances, empiric interventions are employed for both diagnosis and treatment. A specific explanation for oropharyngeal dysphagia cannot be found in a reasonable minority of patients with this syndrome, even when abnormalities in swallowing physiology are defined by the modified barium swallow. Compensatory or corrective swallowing strategies and dietary manipulations are the principal empiric interventions (outlined above), aimed at reducing aspiration risk and avoiding nonoral feeding. The empiric use of botulinum toxin or cricopharyngeal myotomy is ill-advised without convincing evidence of restrictive obstruction at the upper sphincter level because of the potential for added morbidity. [100], [135], [136] Botulinum toxin injection can reduce all indicators of swallowing dysfunction in appropriately selected patients, but the response durability still is being evaluated. [137] Balloon catheter or Savary dilation of the cricopharyngeus more recently has been reported for patients with primary cricopharyngeal dysfunction, and the morbidity associated with the approach is low. [138], [139] There is no evidence that this less invasive approach is effective for oropharyngeal dysphagia unless similar selection criteria are used as for the other techniques. Other empiric therapies are not offered routinely in the management of oropharyngeal dysphagia.

For esophageal dysphagia, empiric strategies include dilation, antireflux therapy, and treatments aimed at sensorimotor dysfunction. The value and safety of dilation with a large-caliber (=14-mm-diameter) bougie or balloon when no abnormality is detected at endoscopy or by videofluoroscopy can be debated, as dilation is not without risk. [113], [140] Dysphagia was significantly reduced in a small group of patients with no obstructive lesions who were randomized to dilation with a 16.7-mm Maloney dilator versus those who underwent dilation with a small-caliber (8.7-mm-diameter) bougie. [141] The benefits were sustained in 80% of initial responders. Empiric use of an 18-mm dilator in an uncontrolled study of patients with esophageal dysphagia for solid foods yet negative endoscopic and radiologic evaluations produced immediate response in 95% of subjects and a sustained response for nearly 2 years in 68% of the initial responders. [142] By comparison, dysphagia resolved in only 12% of the patients with dysphagia for both solid foods and liquids when managed in the same fashion. Although the risk for perforation or significant bleeding approximates 1% when standard dilation is performed for a variety of benign indications, [140] the complication rate is undoubtedly lower when empiric dilation is performed. These findings suggest that dilation with a large-caliber dilator is reasonable as a therapeutic trial in patients with significant symptoms from unexplained esophageal dysphagia, especially if the clinical presentation resembles that of a stricture or ring.

Antireflux therapy, specifically antisecretory therapy, probably is offered to many patients with investigation-negative esophageal dysphagia, but a reasonable estimate of its efficacy is unknown. Esophageal prokinetics, such as the 5-HT $_4$ agonist cisapride, increase midesophageal contraction pressures and could conceivably benefit patients with unexplained esophageal dysphagia, [143] but serviceable reports to support this possibility are unavailable. Glucagon inhibits LES contraction and has been used acutely in patients with idiopathic food bolus impactions. The limited benefits of this approach may be related to the negative effects of glucagon on esophageal body motility. [144] Smooth muscle relaxants, such as nitrates, calcium channel blockers, and peppermint oil, effectively decrease LES pressure or reduce distal esophageal contraction amplitudes and improve transit symptoms in achalasia. [145], [146], [147] and [148] These agents have been tried in patients with spastic disorders with limited benefit. Most trials have studied effects on pain rather than dysphagia, and benefits may be better for transit symptoms. Antidepressants, particularly the tricyclic antidepressants, reduce many unexplained esophageal symptoms, including those associated with spastic esophageal disorders. [6], [27] These agents may alter esophageal sensation or the central processing of visceral afferent input. [6], [149], [150] Although most measured outcomes from antidepressant use have focused on pain, the general cluster of esophageal symptoms, including dysphagia, appears to be responsive when the symptoms represent a functional esophageal disorder. [151], [152] Antidepressants typically are reserved for patients without significant transit delay, with no other explanation for dysphagia on thorough investigation, and in whom enhanced tolerance to the symptom would be a suitable outcome.

APPROACH TO THE PATIENT WITH ODYNOPHAGIA

Odynophagia is an uncommon medical problem in comparison with dysphagia, but its presence usually indicates a significant underlying and definable disorder. The symptom can range from dull discomfort to intense pain that interferes with any swallowing attempt.

The Medical History and Physical Examination of the Patient with Odynophagia

Determining the presence of odynophagia is important in all patients who present with esophageal complaints. The most important characteristic is the temporal relationship of pain to swallowing: early, in the oropharyngeal phase, or later, in the esophageal phase. The characteristics used in differentiating types of dysphagia are helpful in making this determination. When odynophagia is described as accompanying the oropharyngeal phase, malignancy, foreign body, infection, or other causes of inflammation become more likely. Exposure histories that predispose the patient to mucosal irritation, such as prior radiation therapy or corrosive ingestion, should be elicited. A history of tobacco and alcohol use, risk factors for head and neck malignancy, should be recorded, and the physical examination should proceed with these diagnostic possibilities in mind. Nasoendoscopy or indirect laryngoscopy is essential when odynophagia is present in this swallowing phase.

When odynophagia originates in esophageal locations, the medical history should focus on factors predisposing the patient to opportunistic infection, including systemic illnesses, immunosuppressive treatment, and antibiotic use. Symptoms of esophageal disease develop in as many as 30% to 40% of patients with acquired

immunodeficiency syndrome (AIDS) at some point in the course of the illness. [153] The possibility of foreign body ingestion and the use of caustic medications should be carefully explored. Ingestion of pills at bedtime, while reclining, or with little water intake should be determined. The use of caustic medications may not readily be recalled, for example, by young patients using doxycycline for acne management. A list of caustic medications that have been associated with esophageal damage is provided in Chapter 63.

Investigation of Odynophagia

Whether oropharyngeal or esophageal in location, odynophagia typically requires a careful examination of the muscosal surface with direct visualization and biopsy if necessary. This is accomplished with nasoendoscopy, indirect laryngoscopy, or esophagoscopy depending on the nature of the odynophagia. The investigation is altered if a foreign body is the suspected cause of oropharyngeal odynophagia. [154], [155] and [156] Plain films of the neck with soft tissue technique, including lateral views, are indicated, but many foreign bodies are radiolucent (see Chapter 63). Sharp or pointed objects lodged at or above the cricopharyngeus should be removed with use of the laryngoscope or by someone experienced in direct laryngoscopy. [154], [155] Blunt objects retained in this region, such as coins, mandate protection of the airway during removal. In some instances, the foreign body may have become dislodged, leaving residual odynophagia from laceration. Nevertheless, inspection is required, and oral contrast studies (which can interfere with direct mucosal inspection) are not indicated in managing oropharyngeal or esophageal foreign bodies. [154]

The approach to esophageal odynophagia depends on a combination of the acuity of the presentation, the suspected diagnosis, and the setting (immunocompetent or immunocompromised host). Chronic processes often indicate invasion of or extension of mucosa-based processes into the deep esophageal layers. Both radiologic imaging (chest radiography, computed tomography, barium radiography) and endoscopy may be needed to establish the diagnosis and the extent of esophageal or mediastinal involvement. In immunocompetent patients with this presentation, initial testing is driven by the suspected diagnosis, and algorithms defining the most expedient or cost-effective approach are not available.

Provided a history of foreign body ingestion is absent, acute esophageal odynophagia in immunocompetent subjects most often reflects infectious or pill-induced esophageal damage. Evidence for either can be detected by endoscopy or barium radiography, and despite the recognized radiologic patterns of mucosal damage in these forms of esophagitis, [157], [158] endoscopy is more sensitive to mucosal damage. Because of the high incidence of esophageal complaints and disease in AIDS patients, management approaches have been studied. *Candida* infection is very common, and empiric treatment of AIDS patients with oral thrush is recommended. [63], [159] Further investigation, including biopsy, is delayed until a poor response to empiric treatment is determined. Barium radiography is not worthwhile if diagnostic testing is required. A randomized trial comparing barium radiography with endoscopy in symptomatic patients with human immunodeficiency virus infection confirmed the higher sensitivity and utility of the latter technique. [160] In general, cultures taken at the time of endoscopy are less useful than histological study of biopsy specimens because of their inability to differentiate colonization from the pathogen responsible for infection, their lower sensitivity, and the delay in obtaining results. Treatment for odynophagia in this setting is directed by the specific diagnosis (see Chapter 61).

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CHAPTER 32

Donald O. Castell and Philip O. Katz

APPROACH TO THE PATIENT WITH UNEXPLAINED NONCARDIAC CHEST PAIN

Recurring unexplained chest pain located in the substernal area in patients in whom a cardiac etiology has been excluded has been a source of clinical uncertainty for more than a century. Although this phenomenon is frequently called *noncardiac pain*, we prefer the term *unexplained chest pain (UCP)* because even patients with normal coronary arteries occasionally have evidence of myocardial ischemia (i.e., microvascular angina or “syndrome X”). [1], [2] Unexplained retrosternal chest pain is a problem commonly seen in clinical practice, causing anxiety for both patient and physician because of the uncertainty regarding possible underlying coronary artery disease.

UCP remains a difficult and challenging problem for the gastroenterologist because these patients are often referred for evaluation of the esophagus once a cardiac etiology has been excluded. Up to 13% of people in a survey of U.S. households reported chest pain of unknown cause, and up to 44% of these respondents were seeking care for their pain. [3] The differential diagnosis centers around ruling out cardiac disease because this is the most serious cause of pain. The importance of excluding a cardiac etiology is reinforced by a study showing that when cardiac disease is ruled out, almost no cardiac fatalities occur during 7-year follow-up. [4] Coronary artery and esophageal disease coexist; 50% of patients with proven angina secondary to coronary atherosclerosis may have gastroesophageal reflux disease (GERD). [5] Establishing an esophageal origin of chest pain with a high degree of certainty results in less frequent pain and fewer visits to the physician. [6]

After coronary artery disease has been ruled out, a search for an esophageal cause, particularly GERD, should be aggressively pursued because this condition is eminently treatable. GERD can be diagnosed with a therapeutic or diagnostic trial of medication or by an abnormal finding on ambulatory pH monitoring with symptom correlation. The yield of endoscopy is low in the evaluation of chest pain compared to that in typical reflux disease. [7] If GERD is excluded, the presence of an esophageal motility abnormality or visceral hypersensitivity can be pursued with manometry and provocative testing with edrophonium [8] or balloon distention. [9]

OVERVIEW AND SCOPE OF THE PROBLEM

Approximately 1,500,000 million patients per year undergo cardiac catheterization for recurring chest pain. [10] In 30% or more of these, the findings are normal. Many other patients undergo a noninvasive cardiac evaluation that is considered to exclude coronary artery disease, leaving at least 450,000 new patients with unexplained chest pain seen annually in the United States. Our own review of 635 patients seen during a 10-month period in the emergency department at the Graduate Hospital (Philadelphia) with substernal chest pain showed that for 75%, the final diagnosis was noncardiac (Fig. 32-1). A recent multicenter review of more than 10,000 patients evaluated in emergency departments for substernal chest pain revealed that 55% had no evidence of any cardiac abnormality when followed for at least 6 months. [11] The cost of caring for these patients is exceptionally high, likely to be more than $2 billion a year.

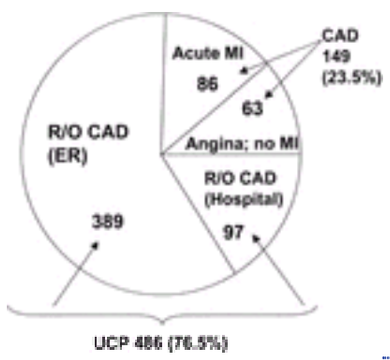

FIGURE 32-1. Discharge diagnosis for 635 consecutive patients seen in the emergency department at Graduate Hospital for acute substernal chest pain. Coronary artery disease (*CAD*) was excluded (*R/O*) in 76.5% either in the emergency department evaluation or after brief admission to the hospital. The diagnosis of these patients remains unexplained chest pain (*UCP*). CAD was diagnosed in the hospital as either acute myocardial infarction (*MI*) or persistent angina in a total of 23.5% of the patients.

DIFFERENTIAL DIAGNOSIS

The clinical history often does not easily differentiate coronary from esophageal pain in patients with UCP. In one study, 25% of patients with chest pain evaluated by a cardiologist were given a mistaken diagnosis of coronary artery disease based on the clinical presentation compared to the final angiographic findings. [12] Gastroesophageal reflux can be induced during exercise [13] and present with exertional chest pain that mimics angina pectoris. In fact, changes in ST-T waves indistinguishable from those of coronary chest pain may be seen during exercise testing.

If the history shows other key esophageal symptoms, such as heartburn, dysphagia, regurgitation, and odynophagia, an esophageal cause should be suspected. Heartburn is strongly predictive of GERD in these patients. Features that also suggest an esophageal origin include pain lasting more than 2 to 3 hours, pain that does not radiate laterally, meal-related pain, pain that is relieved by antacids, or pain that awakens the patient. None of these are 100% specific, however, and further evaluation is always required; cardiac disease must be ruled out.

The absence of esophageal symptoms decreases the likelihood of an esophageal cause of chest pain. However, up to 50% of patients who prove to have coronary artery disease will have one or more symptoms typical of esophageal disease, and many will have concurrent coronary artery disease and GERD. [14] It is well-known that mitral valve prolapse and esophageal abnormalities can coexist. As many as 80% of patients with mitral valve prolapse have been shown to have abnormal esophageal motility, often consistent with that seen in diffuse esophageal spasm. [15]

Because the history cannot definitely differentiate cardiac from esophageal pain, a cardiac evaluation is recommended as the initial approach in patients with unexplained substernal pain. The nature of the workup is determined by numerous factors, including the patient’s age and other cardiac risk factors. Although a noninvasive workup with an electrocardiogram, exercise testing, and perhaps echocardiography may be reasonable in ruling out cardiac disease in a person younger than 40 years, coronary angiography remains the gold standard. It is our policy to leave this decision to cardiac specialists.

Patients with normal coronary arteries or insignificant (<50%) lumenal narrowing have an excellent prognosis; a mortality rate of less than 1% from cardiac causes was seen in a 7-year follow-up study. [4] A normal angiogram allows the physician to reassure patients that their recurring chest pains are not life threatening. Despite

this reassurance, most patients continue to have recurring chest pain and a compromised lifestyle, and many continue to believe that they have heart disease even after coronary artery disease has been ruled out. [16]

The direct cost of caring for these patients was estimated to be $4,000 per patient or more than $300 million dollars per year in 1988. [8] It is likely that the actual number of patients and the total costs have become considerably higher as health care costs have risen dramatically. Because of this continued morbidity and cost, providing an accurate and thorough diagnosis is important in caring for these patients.

Despite the difficulty of determining the cause from the history, evaluation after cardiac disease has been excluded is centered on ruling out diseases of the upper gastrointestinal tract, biliary tree, chest wall, and pulmonary system that may present with chest pain.

DIAGNOSTIC APPROACH

A suggested diagnostic approach is illustrated in Figure 32-2. Peptic ulcer disease and biliary tract disease rarely present with chest pian. Therefore, the frequency of these abnormalities in patients with UCP is quite low. In the absence of a history of coexistent right upper quadrant pain after meals (biliary colic), jaundice, or classical ulcerlike dyspepsia, a routine evaluation for these diseases is not warranted. As noted above, endoscopy is usually unrevealing.

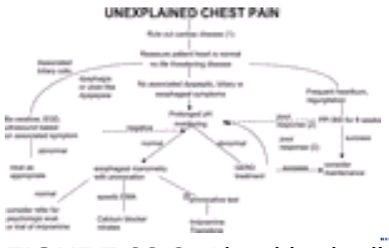

FIGURE 32-2. Algorithmic diagnostic/therapeutic approach to patients with unexplained chest pain. Initial evaluation to exclude cardiac disease depends on (1) testing individualized for each patient, usually in consultation with a cardiologist, and (2) ambulatory pH monitoring with esophageal and intragastric electrodes while the patient is on therapy to evaluate the adequacy of gastric acid suppression and control of reflux before an alternative diagnosis is considered. Spastic esophageal motility abnormality (*EMA*) includes diffuse esophageal spasm, nutcracker esophagus, and hypertensive lower esophageal sphincter. A therapeutic trial with a proton pump inhibitor (*PPI*) given twice daily before meals (*BID*) is particularly indicated in patients with associated symptoms suggesting gastroesophageal reflux disease (*GERD*).

If the patient has dysphagia, consider esophagogastroduodenoscopy/barium swallow as the initial approach to evaluate the patient for stricture, mass lesion, or other obstruction, such as a ring. If heartburn is present, the diagnosis of GERD is more likely, and early endoscopy is probably not necessary because endoscopic esophagitis is seen in fewer than 10% of patients with UCP. [7] In the absence of dysphagia, endoscopy may not be useful. Abdominal ultrasonography should be used judiciously in these patients because biliary disease may be asymptomatic (i.e., “silent” gallstones), and experience suggests that in the absence of biliary colic, cholecystectomy is not helpful.

Ambulatory pH monitoring is the preferred technique to document reflux and demonstrate a correlation between reflux and symptoms. Several investigations have shown that up to 50% of patients with UCP have an acid-induced esophageal abnormality: increased esophageal acid exposure, a high symptom index (a correlation of episodes of reflux with chest pain) without an increased frequency of reflux (the “acid-sensitive” esophagus), or exercise-induced reflux. [5] /SUP>, [7] , [17] Studies have suggested that a trial of proton pump inhibitor (PPI) therapy may be more appropriate (see section “ Treatment”).

Esophageal manometry has been used extensively in the evaluation of patients with UPC; however, its usefulness is debated. About 25% to 30% of patients have some type of abnormal esophageal motility during stationary manometry (Fig. 32-3) [18] ; however, few have pain during motility testing, so that it is difficult to establish the clinical significance of an abnormal test result. We believe it is useful to attempt to induce the patient’s pain by using various provocative tests. The most widely used approach is to induce pain with an intravenous injection of edrophonium (Tensilon; Hoffman-LaRoche, Nutley, New Jersey). In up to 30% of patients, typical chest pain is reproduced when a 10-mg dose is given by intravenous push. [8] The high-amplitude contractions induced by edrophonium are not the cause of the pain; normal subjects do not have pain, and the patients’ motility changes are the same as those in normal subjects. This suggests heightened visceral sensitivity (of unknown cause). Edrophonium does not cause coronary artery narrowing or affect the pulmonary circulation; therefore, when the pain is reproduced, the esophagus can be implicated as the source. The sensitivity of provocative testing may be increased by using progressive balloon distention. Patients with UCP have a lower threshold for pain during balloon distention than normal subjects—that is, they experience pain with a lesser degree of distention. [9]

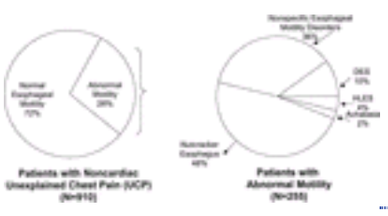

FIGURE 32-3. Esophageal motility abnormalities identified in 255 of 910 patients with unexplained chest pain following exclusion of coronary artery disease. *DES*, diffuse esophageal spasm; *HLES*, hypertensive lower esophageal sphincter. (From ref. [18] .)

PATHOGENESIS

As noted above, it is likely that as many as 50% of patients with UCP have GERD; in one placebo-controlled study, 80% of patients with documented GERD had a clinical response to omeprazole. [19] Once GERD has been excluded, the pathogenesis of UCP remains elusive. The high-amplitude contractions, such as those seen in “nutcracker esophagus,” have been suggested by some to indicate an esophageal cause; however, they have been suggested by others to represent a clinically unimportant finding or a secondary response to stress-induced panic. In a study in which ten subjects with UCP underwent simultaneous measurement of intralumenal pressure and pH and ultrasonographic imaging of the esophagus for 24 hours, [20] muscle thickness was assessed by ultrasonography during chest pain and asymptomatic baseline and control periods. Sustained esophageal contractions averaged 68 seconds. Five patients had a positive chest pain response to edrophonium provocation, and all had sustained esophageal contractions during the pain. Sustained esophageal contractions of shorter duration (mean, 24 seconds) were seen in a few asymptomatic periods. Healthy subjects and patients without chest pain did not have sustained contractions. Muscle thickness was increased during both chest pain and asymptomatic periods. Thus, the magnitude of the contraction was not predictive of the occurrence of pain, which corroborates the findings of stationary esophageal manometry.

It has been suggested that UCP is a disorder of visceral hypersensitivity. We evaluated the role of repeated esophageal acid sensitization in the production of chest pain. [21] Esophageal acid exposure in the 30 minutes preceding episodes of chest pain associated with reflux (pH < 4) was compared with acid exposure in the 30 minutes preceding episodes of chest pain not associated with reflux. In 51 patients, 129 pain events were recorded; 34 events were associated with episodes of reflux, and 95 were not. In the episodes of chest pain associated with reflux, significantly more total esophageal acid exposure time was observed in the 30 minutes preceding the index event than in those pain events not associated with reflux (6.8 ± 1.9 minutes versus 0.6 ± 0.6 minutes, $P < .001$). Also, more individual episodes of reflux occurred in the 30 minutes preceding the index pain event in the 34 events with a positive symptom correlation than in the 95 events with a negative symptom correlation.

We have also found that a shorter duration of esophageal acid infusion is needed to induce chest pain when the time interval since a previous infusion is shorter. [22] In an older study in which a modified Bernstein test was used, a repeated infusion at a pH of 1 or 1.5 resulted in a greater frequency of chest pain than a single infusion. [23] In addition, prior acid infusion into the esophagus reduces the volume threshold at which chest pain is first perceived when an intraesophageal balloon is inflated. [24] These studies support other evidence that a hypersensitive esophagus may be an important variable in the production of chest pain.

TREATMENT

Recurring substernal chest pain, when it remains truly unexplained after evaluation by numerous health care providers, produces considerable anxiety for the patient. With the background of a negative cardiac evaluation, treatment begins with reassurance to the patient that the heart is normal and that a life-threatening illness is not present. If an esophageal abnormality is found and the patient is made aware that the esophagus is the cause of pain, the patient is better able to accept the symptoms and experiences fewer limitations in daily life as a result of the pain. [6]

It is common practice to treat UCP secondary to GERD with traditional antireflux therapy. Uncontrolled observations with both histamine H_2 receptor antagonists and PPIs have suggested that this is an effective approach. The only double-blinded, randomized, placebo-controlled trial [19] compared omeprazole and placebo in 36 patients with UCP and GERD documented by 24-hour ambulatory pH testing. Patients received either omeprazole (20 mg twice daily) or placebo for 8 weeks and kept a diary of the frequency and severity of daily chest pain scored on a scale of 0 (none) to 10 (worst possible). This study confirmed clinical experience in that a significant decrease in chest pain was achieved in 81% of patients with omeprazole, versus 6% with placebo (Fig. 32-4); both the number of days with chest pain (39% ± 7.2% versus 10% ± 6.9%, $P = .006$) and the severity of pain (40.7 ± 8.1 versus 14.8 ± 8.2, $P = .03$) were reduced. This study included both male and female patients (female-to-male ratio of 2:1), and almost all had normal findings on endoscopy (90%). The study supports the use of PPIs in higher doses in patients with UCP who have known nonerosive GERD, the typical population seen in clinical practice. The study also reinforces the need and value of excluding GERD in this population.

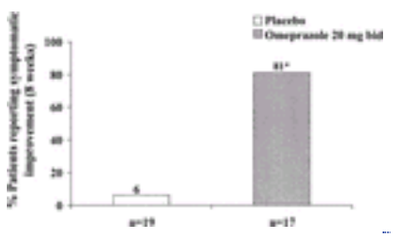

FIGURE 32-4. Percentage of 36 patients with unexplained chest pain and abnormal findings on pH monitoring reporting symptomatic improvement after 8 weeks on omeprazole therapy versus placebo. (From ref. [19] .)

The Omeprazole Test

Fass et al. [25] examined the value of empiric therapy in a randomized, double-blinded, placebo-controlled trial of a short course of high-dose omeprazole as a diagnostic test for GERD in 37 patients with UCP (the “omeprazole test”). Coronary artery disease was excluded by cardiac catheterization or a noninvasive workup and consultation with a cardiologist. To be enrolled, patients had to have chest pain at least three times a week and be able to record symptoms accurately. Before the study, all participants underwent upper gastrointestinal endoscopy and 24-hour ambulatory pH monitoring and kept a diary of the frequency and severity of chest pain. Patients were randomized to either placebo or omeprazole (40 mg in the morning and 20 mg in the evening) for 7 days, followed by a crossover after a 2-week washout period during which baseline symptom assessment was repeated. Of the 37 patients, 23 (62.2%) were classified as positive for GERD and 14 as negative for GERD, based on the presence of erosive esophagitis or abnormal results of 24-hour pH monitoring without calculation of a symptom index.

The result of the treatment trial (the omeprazole test) was considered positive for GERD if the baseline chest pain score was reduced by more than 50% after treatment with omeprazole or placebo. A positive omeprazole result was noted in 18 (78%) of the GERD-positive patients and in only 2 (14%) of the GERD-negative patients, which translated to a sensitivity of 78.3% (95% confidence interval [CI], 61.4–95.1) and a specificity of 85.7% (95% CI, 67.4–100) in comparison with upper gastrointestinal endoscopy and ambulatory pH monitoring for the diagnosis of GERD. An economic analysis performed later estimated that this 1-week omeprazole trial could save $573.00 per patient with UCP, assuming that the patient would be evaluated by endoscopy, esophageal manometry, and ambulatory pH monitoring. Such a treatment trial could result in an 81% reduction in the number of endoscopies and a 79% reduction in the use of ambulatory 24-hour esophageal pH monitoring in this patient population.

This is the best-designed study in which researchers have attempted to investigate this difficult issue. It was blinded, randomized, and controlled with placebo. All patients received all diagnostic and therapeutic interventions. However, the population studied was almost exclusively male (unusual for UCP, in which the gender distribution is usually 50:50), and the frequency of endoscopic esophagitis was higher (35%, or eight patients) than in other large studies of patients with UCP (<10%). [7] , [19] It is particularly important to be aware of the frequency of pain in these patients. All had chest pain at least three times a week. The frequency of heartburn was high, so that the pretest probability of GERD was high. In general, patients with UCP have pain less frequently (often less than once a week) and have less frequent heartburn, and a 1-week trial therefore may not be sufficient to evaluate improvement. Whether a longer trial would suffice has not been studied. The study does not allow us to examine treatment after the omeprazole test. It remains to be shown whether this PPI test will be useful in those patients with less frequent pain and whether the results can be extrapolated to women with chest pain.

It is not clear what is the best dosage of PPI to use. Higher doses appear to be needed than are used to treat patients with heartburn. Whether a PPI twice daily (before breakfast and dinner) or a regimen including a PPI twice a daily plus a histamine H_2 receptor antagonist at bedtime [26] provides the “best” outcome of aggressive acid suppression is not known.

The sensitivity and specificity of prolonged ambulatory pH monitoring are the subject of considerable debate. Beedassy et al. [21] evaluated the results of monitoring 104 consecutive patients with UCP and normal coronary arteries documented by catheterization. The percentage of time that the pH was less than 4 in patients (upright, recumbent, and total) and the symptom correlation with a pH of less than 4 (symptom index) were reviewed. In 51 (45%) of the patients, the percentage of time when the pH was less than 4 during the study was abnormal. Patients with a high symptom index (27) were more likely to have an abnormal pH result. These results confirm those of previous studies showing that pH monitoring is the most sensitive traditional diagnostic test in patients with UCP.

Fass et al. [28] evaluated the ability of 24-hour ambulatory esophageal pH monitoring to predict a reduction in chest pain with treatment in 23 patients with GERD-related UCP. Twenty-two of the patients were men, and all had had at least three episodes of chest pain a week for a minimum of 3 months before the study. Of the 23 patients, 15 (64%) had abnormal results of 24-hour pH monitoring based on the time of esophageal acid exposure at a pH of less than 4. No comment was made about a positive symptom index. All were treated with omeprazole (40 mg in the morning and 20 mg in the evening). A significant correlation ($r = 0.51$, $P < .02$) was found between the results of 24-hour ambulatory esophageal pH monitoring and the change in symptom intensity score with treatment. This study suggests the value of prolonged ambulatory monitoring, both from the standpoint of the frequency of an abnormal study and its potential value in predicting the response to treatment. A positive symptom index during pH-metry should provide the best predictor of outcome of PPI therapy.

If patients do not respond to a trial of antireflux therapy, an evaluation with prolonged ambulatory pH monitoring performed with dual esophageal and gastric pH electrodes while therapy is continued is the procedure of choice. [29] This allows assessment of pH control and symptom correlation. In the event that gastric pH control is incomplete, especially overnight, [30] esophageal acid exposure continues, [31] or symptoms persist in association with continued reflux, higher-dose PPI therapy or the combination of a PPI twice daily plus a histamine H_2 receptor antagonist at bedtime can be considered. If the results of pH testing are negative, further evaluation should include esophageal motility testing and provocation with edrophonium (and balloon, if available). Patients without heartburn or regurgitation (or other symptoms to suggest GERD) should be evaluated earlier with pH monitoring. Patients successfully treated with acid suppression should be considered for long-term maintenance with PPI therapy, although no specific study has addressed this issue.

Other Treatments

Many patients with UCP do not have GERD. Although in 25% to 35% a motility abnormality is detected on esophageal manometry and in approximately 20% pain is provoked by stimulation with edrophonium, treatment of this group has been difficult. At least one placebo-controlled study suggests that a calcium channel blocker may be effective for patients with nutcracker esophagus. [32] Nitroglycerin has been shown to reduce the pain and simultaneous contractions in patients with diffuse esophageal spasm. [33], [34] A more recent uncontrolled study has suggested that endoscopic injection of botulinum toxin may be effective in these patients. [35] Double-blinded, placebo-controlled studies are limited in number and have shown minimal success; however, these agents should be considered in patients with abnormal esophageal contractions.

A 50-mg dose of imipramine at bedtime results in significant improvement in patients with UCP who do not have reflux disease. [36] Anxiolytics [37] or the antidepressant trazodone (100–150 mg/d) [38] has been shown to be helpful in decreasing symptoms associated with abnormal esophageal contractions. It is likely that these agents, particularly imipramine, act to alter visceral sensation. [39] The reports lend support to the suggestion that pain or hypersensitivity in these patients is mediated by abnormal visceral nociception. Many patients have high scores on somatization and depression scales during psychological testing, so the agents may also relieve these symptoms.

Prakash and Clouse [40] have reported a long-term experience of tricyclic antidepressants in 21 outpatients with UCP, normal coronary arteries, and no or an incomplete response to antireflux therapy. Patients were observed for a minimum of 6 months (median, 2.7 years; range, 0.8–8.6 years), and all received tricyclic antidepressants continuously. Five patients had had abnormal pH readings, and aggressive PPI or surgery had failed in all. Only six (28.6%) had an anxiety or affective disorder. Seventeen (81%) showed improvement or complete remission during initial treatment with 20 to 75 mg (median, 50 mg) of amitriptyline, imipramine, nortriptyline, or desipramine daily. Only 3 of the 17 whose condition improved required a change because of side effects.

In follow-up, five (29%) patients discontinued treatment after 0.5 to 2.7 years with long-term relief of chest pain. In one, the pain recurred. Seven (41.2%) were in treatment almost continuously for a mean of 2.6 ± 0.6 years. Five patients discontinued medication (despite pain relief) because of side effects. Overall, 13 (76.5%) of 17 experienced sustained improvement (>6 months). None of these 13 required further investigation or treatment for chest pain during follow-up.

Several potentially important points are made. Long-term tricyclic agents are tolerated well by these patients, and pain is relieved. Although these observations are uncontrolled, they offer some promise for patients in whom antireflux therapy fails or who do not have GERD, which underscores the importance of a thorough evaluation. Patients do not have to have psychiatric disturbances to respond to these agents. Regardless of their mechanism, the agents offer promise. They are inexpensive and generally well tolerated.

Another interesting treatment modality was tested by Van Peski-Oossterbaan and colleagues. [41] Seventy-two patients with UCP (normal coronary arteries) and without major depression, psychosis, organic brain syndrome, or use of psychoactive substances were randomized to no treatment or to cognitive-behavioral therapy (4 to 12 weekly sessions of 45 to 60 minutes each), during which the patients were instructed in relaxation techniques, counseled, and shown how to identify defined irrational beliefs about UCP. The patients were evaluated at 6 and 12 months for frequency of chest pain and use of medical services. A clear advantage of the cognitive-behavioral treatment was found in comparison with no treatment, inasmuch as 50% of those treated were free of chest pain at 6 months, versus 6.1% of those untreated (P <.001). Forty-eight percent were free of pain at 12 months in the treated group, versus 13% of those untreated (P <.001). Visits to their general practitioners were unchanged. Patients appeared to have high levels of baseline functioning and were educated. The study was not blinded. Nevertheless, the result reinforces those of other case reports and the clinical impression that nonpharmacological intervention may be of value in selected patients. This time-intensive method should not be ignored as definitive treatment.

SUMMARY

Published studies support the clinical impression that PPIs are the drug of choice for GERD-related UCP. The recent literature encourages the use of a trial of a PPI as a useful diagnostic test for GERD in patients with UCP who have chest pain more than three times a week and other symptoms suggestive of GERD. Tricyclic antidepressants and behavior modification appear useful in patients who do not have GERD, underscoring the importance of a thorough assessment in these patients. Further research is needed to elucidate the respective role of esophageal sensory and contraction abnormalities in the pathogenesis of this complex syndrome.

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CHAPTER 33

Grace H. Elta

APPROACH TO THE PATIENT WITH GROSS GASTROINTESTINAL BLEEDING

Gastrointestinal (GI) bleeding is a common clinical problem requiring more than 300,000 hospitalizations annually in the United States. The annual rate of hospitalization for upper GI bleeding has been estimated at 36 to 102 patients per 100,000 members of the general population and is twice as common in men as in women. [1], [2] Lower GI bleeding is less common; the estimate from a large health maintenance organization of cases of acute lower GI bleeding leading to hospitalization was 20 per 100,000. [3] The rates of acute upper and lower GI bleeding are both increased in patients taking aspirin, and the risk appears to dose related. [4], [5] Calcium antagonists, which also inhibit platelet aggregation, may increase GI bleeding, [6], [7] whereas nitrovasodilator drugs may decrease the risk. [8]

During the last 10 years, hospitalization days have significantly decreased, and most patients with upper GI bleeding undergo endoscopy within 24 hours after admission. [9] Of these endoscopic procedures, 20% to 35% include endoscopic hemostatic therapy. An important change in the management of nonvariceal upper GI bleeding has been the use of upper GI endoscopy to assess the risk for rebleeding, allowing outpatient care of selected patients with significant cost savings. [10], [11] Because the cost of a hospital bed accounts for most of the hospital cost of providing care for patients with upper GI hemorrhage, shortening the length of stay or eliminating the need for hospitalization can significantly lower costs. A comparison of the costs of care for patients with acute upper GI bleeding showed that gastroenterologists provide more cost-effective care than internists or surgeons. [12] The cost savings associated with a shortened length of stay reflected the time interval between endoscopy and discharge; the time to endoscopy did not differ among services.

Mortality rates from upper GI hemorrhage vary from 3.5% to 7% in the United States, [1], [2] and [3], [13] although a large study from the United Kingdom reported a mortality rate of 14%. [14] The mortality rate for lower GI bleeding has been reported at 3.6%; although similar to upper GI bleeding, it is markedly higher in patients who begin bleeding after hospitalization. [3] The early and accurate diagnosis of patients with severe bleeding can facilitate therapeutic maneuvers, leading to lower mortality rates. [15] This trend appears to have developed with the use of therapeutic endoscopic techniques. [16]

CLINICAL PRESENTATION

The overall strategy for management of GI bleeding starts with an assessment of the patient to determine how much blood has been lost and whether the bleeding is ongoing. This is followed by resuscitation because the dire consequences of GI bleeding are those of shock. A brief history and physical will often determine whether the source of bleeding is in the upper or lower GI tract and whether the bleeding has ceased, which happens in 80% of patients. The need for urgent versus elective endoscopy can then be determined (although in upper GI bleeding, urgent endoscopy has been proposed for all patients to facilitate triage and decreased resource utilization). If endoscopy is to be performed electively, empiric medical therapy for the most likely diagnoses can be started to decrease the risk for rebleeding. Once a definitive diagnosis is made, specific medical, endoscopic, angiographic, or surgical therapy can be performed.

Patient Assessment

The first step in assessing the bleeding patient is to determine the urgency of the situation. Agitation, pallor, hypotension, and tachycardia may indicate shock requiring immediate volume replacement. Patients with severe blood loss may actually have bradycardia rather than tachycardia secondary to vagal slowing of the heart. [17] Shock occurs when blood loss approaches 40% of the total blood volume. If no evidence of hypotension is found, the orthostatic vital signs can help diagnose lesser degrees of intravascular volume depletion. Postural hypotension with a decrease in systolic blood pressure of 10 mm Hg or more usually indicates at least a 20% reduction in blood volume. In the acutely bleeding patient, intravenous access should be established. If the patient has signs of shock or continues to bleed, a large-bore central intravenous line is useful. Blood samples for an assessment of hematocrit, platelets, and coagulation factors and for blood typing and cross-matching should be sent immediately to the laboratory.

The initial hematocrit obtained for a patient with acute bleeding poorly reflects the degree of blood loss. Because the hematocrit is expressed in terms of erythrocyte volume as a percentage of the total blood volume, it does not drop until the blood volume has been restored. This repletion of blood volume from extravascular fluid begins immediately but takes 24 to 48 hours to equilibrate completely. An acutely bleeding patient can be evaluated more effectively by monitoring the blood pressure and pulse and looking for gross evidence of ongoing bleeding than by assessing laboratory tests.

Resuscitation

Patients with severe acute GI bleeding require admission to an intensive care unit. Fragile patients with a history of cardiopulmonary disease may require measurement of capillary wedge pressure. The intravascular volume should be replenished with normal saline solution to prevent the consequences of shock while blood is being cross-matched for transfusion. This allows adequate circulation of the remaining erythrocytes. The oxygen-carrying capacity of blood can be maximized by administering supplemental oxygen. In rapidly bleeding patients, oxygen availability is markedly decreased during early hemorrhage, reflecting primarily a decreased cardiac output. The decrease in oxygen-carrying capacity of the blood caused by a decrease in hemoglobin is less important. Metabolic acidosis has been measured in patients with acute hemorrhage, reflecting poor tissue perfusion. [18] Close attention to vital signs, urine output, and central vascular pressure is mandatory.

The specific criteria that define when a patient requires transfusion vary according to the age of the patient, whether concomitant cardiopulmonary disease is present, and whether the bleeding continues. In general, the hematocrit should be maintained above 30% in elderly patients and above 20% in young, healthy patients. With continued evidence of bleeding, the decision to transfuse cannot be based on the hematocrit alone. Unstable vital signs and gross evidence of active bleeding, such as hematemesis, bright red blood in the nasogastric aspirate, or hematochezia, are better requisites for transfusion. The hematocrit is a poor index for following the patient's need for additional transfusions. The plasma volume after acute GI bleeding is often overexpanded by intravenous fluids; the immediate posttransfusion hematocrit may underestimate the final value. Overuse of transfusions is probably more common than underuse.

In general, packed erythrocytes are the preferred form of blood transfusion. Whole-blood transfusions should be reserved for the unusual circumstances of massive blood loss and rapid, high-volume replacement, which increases the need for coagulation factor replacement. Preferably, blood volume has already been replenished with saline solution by the time banked blood is available. The use of packed cells also spares components for the blood bank. If the results of coagulation tests are abnormal, as is the case in many patients with cirrhosis, fresh-frozen plasma and platelets may also have to be administered. Even patients with initially normal coagulation factors and platelet counts eventually need plasma and platelet transfusions if they are transfused repeatedly. Patients who require massive transfusions (>3000 mL) should receive warmed blood to prevent decreases in body temperature. [19] Rarely, in massively transfused patients, calcium supplementation may be

necessary to counter the effects of calcium-binding agents in banked blood.

Location of Bleeding

In patients with obvious upper GI bleeding who present with hematemesis, a nasogastric tube should be placed to further assess the rate of ongoing blood loss. When upper GI bleeding is suspected, as in the patient with melena or with a history of previous epigastric symptoms or disease, a nasogastric tube aspirate demonstrating blood confirms the upper tract as the source. However, the nasogastric aspirate may be a negative in a patient with duodenal bleeding presenting with melena when a competent pylorus prevents duodenogastric reflux. [20] A negative nasogastric aspirate does not preclude the upper gut as the source of bleeding. Melena usually indicates an upper GI source (i.e., above the ligament of Treitz), although bleeding may be from the small bowel or proximal colon. Melena occurs when hemoglobin is converted to hematin or other hemochromes by bacterial degradation. This can be produced experimentally by the ingestion of as little as 100 to 200 mL of blood. [21] If the volume of a lower GI hemorrhage is too small to cause hematochezia but sufficient to supply enough hemoglobin for degradation, and if colonic motility is sufficiently slow, bleeding from the small bowel or proximal colon may cause melena. This is an uncommon occurrence because small bowel bleeding is rare and colonic sources bleed slowly, causing Hemoccult-positive stools, or bleed rapidly enough to cause hematochezia.

Another indication of an upper GI source of bleeding is a mildly elevated level of blood urea nitrogen (BUN). [22] Some of this azotemia is caused by the absorption of blood, but the experimental ingestion of blood results in lower elevations in BUN of shorter duration, suggesting that part of the azotemia is secondary to hypovolemia. [23] A study has confirmed that the BUN-to-creatinine ratio is higher in upper GI than in lower GI bleeding but is a poor discriminator. [24] Testing for occult blood in nasogastric aspirates is rarely necessary because the blood is often obvious. The one occasion on which occult blood testing is helpful is when a coffee ground appearance of the aspirate may be produced by some foods. A simple positive test for occult blood may merely indicate nasogastric tube trauma. When occult testing of gastric aspirates is used, it is important not to rely on standard stool kits, which may yield false-negative results in acidic solutions. [25]

Hematochezia usually indicates a lower GI source. However, 11% of patients with rapid bleeding from an upper GI source pass bright red blood rectally because of rapid GI transit. [26] Placement of a nasogastric tube and an endoscopic examination should be considered if there is any question about the location of bleeding in a patient with hematochezia.

ACUTE UPPER GASTROINTESTINAL BLEEDING

Upper GI bleeding is a common clinical problem, causing 10,000 to 20,000 deaths each year in the United States. Forty-four percent of hospitalizations for upper GI bleeding are for patients older than 60 years of age. [1] Approximately 80% of upper GI bleeding episodes are self-limited and require only supportive therapy. [27] Patients with continued or recurrent bleeding have mortality rates of 25% to 30%. [1], [27]

Prognostic Indicators

Several prognostic indicators in upper GI bleeding have been identified. The most important is the cause of bleeding. Variceal hemorrhages have much higher rebleeding and mortality rates than other conditions. Mortality from variceal hemorrhage during the initial hospitalization is at least 30%, with rebleeding rates of 50% to 70%. [28] A reduction in the mortality rates associated with variceal bleeding would lower the overall mortality of upper GI bleeding because varices account for approximately 10% of all bleeding episodes. [29]

Stigmata of recent bleeding that can be visualized at endoscopy, such as active arterial spurting, oozing of blood, a visible vessel, or a fresh or old blood clot, are important predictors of outcome in peptic ulcer bleeding (Table 33-1). A visible vessel is described endoscopically as an elevated, dark red, blue, or gray mound that protrudes from the ulcer crater and is resistant to washing. The endoscopic diagnosis of a visible vessel has been validated by pathological correlation in a group of gastric ulcer patients who required surgical resection, although this visible vessel is actually an organizing clot plugging a side hole in the bleeding artery located just below the ulcer base. [30] The evolution of the endoscopic appearance of visible vessels, or sentinel clots, has been described as an initial large, red clot that becomes darker and smaller with time, eventually replaced with a white plug of fibrin and platelets that finally disappears. [31] The dark, small sentinel clot that is not oozing, also described as a bare visible vessel, and older stigmata in the form of a flat, black eschar or white clot, have lower rates of rebleeding. [32] Controversy regarding the evolution of vessel color and the variable risks associated with the color of the visible vessels continues. [33]

STIGMATA OF HEMORRHAGE	INCIDENCE (%)	REBLEEDING (%)
Spurting arterial bleeding	8	85–100
Nonbleeding visible vessel	17–50	18–55 (mean, 43)
Adherent clot (no visible vessel)	18–26	24–41
Other stigmata	12–18	5–9
No stigmata	10–36	0

Adapted from Johnston JH. Endoscopic risk factors for bleeding peptic ulcer. Gastrointest Endosc 1990;36:516.

TABLE 33-1 Stigmata of Hemorrhage and the Risk for Rebleeding

Despite the lack of uniform endoscopic descriptions and risks for rebleeding depending on the type of visible vessel, the presence of a visible vessel in an ulcer crater at endoscopy predicts an increased risk for required surgical intervention and increased mortality. [34] The incidence of rebleeding in patients who have ulcers with visible vessels is up to 50%, whereas no rebleeding is observed in patients with no stigmata of recent bleeding. [35] When endoscopy is performed within 6 to 24 hours after admission, visible vessels are found in 20% to 50% of bleeding ulcers.

The identification of predictors of recurrent hemorrhage may direct the need for therapeutic endoscopic techniques, which can lower the mortality rate for patients with ulcers that have stigmata of recent bleeding. [36] Which stigmata indicate a requirement for therapy in patients with bleeding ulcers? More than 30 randomized trials have been reported, and although results have varied, most authorities recommend treatment of actively bleeding (i.e., spurting or oozing) visible vessels and nonbleeding visible vessels that are raised and cannot be washed off. [37] Adherent clot should be aggressively washed off to assess a possible underlying vessel if there is clinical evidence of major hemorrhage, such as hypotension, significant hematemesis, or a transfusion requirement of more than two units.

Other prognostic indicators include the following:

Severity of the initial bleed. Severity is assessed by the transfusion requirement, the presence of bright red blood in the nasogastric aspirate, or the presence of hypotension (Table 33-2). [38]

NASOGASTRIC ASPIRATE	STOOL COLOR	MORTALITY RATE (%)
Clear	Red, brown, black	10
Coffee grounds	Brown or black	10
	Red	20
Red blood	Black	10
	Brown	20
	Red	30

Adapted from Silverstein FE, Gilbert DA, Tedesco FJ. The national ASGE survey on upper gastrointestinal bleeding. Gastrointest Endosc 1981;27:73.

TABLE 33-2 Prognostic Value of the Severity of Upper Gastrointestinal Bleeding

Age of the patient. Patients older than 60 years have been shown to have higher mortality rates than their younger counterparts, although this indicator may not be independent from concomitant disease. [1]
Concomitant disease. Diseases such as chronic renal failure and severe cardiopulmonary disease affect the ultimate outcome.

Onset of bleeding during hospitalization. Patients who begin to bleed while hospitalized have a mortality rate of 25%, whereas the rate is only 3.7% for patients who start to bleed before admission. [2]
Giant ulcers. Patients with giant ulcers (diameter >2.0 cm) have mortality rates as high as 40%. [39]
Emergency surgery. Patients requiring emergency surgery have a surgical mortality as high as 30%, compared with 10% for those undergoing elective surgery.

Diagnostic Approach

Patients with self-limited, minor bleeding and other more serious medical problems may not require endoscopy. However, for most patients, even those with relatively minor bleeding, an accurate diagnosis or localization of the source is desirable to direct further management.

History and Physical Examination As initial resuscitative measures are being implemented, a history should be taken and a physical examination performed. Even experienced gastroenterologists can guess the cause of bleeding only 50% of the time after a careful history and physical examination. However, the history may raise specific diagnostic possibilities. A history of peptic disease or dyspeptic symptoms suggests ulcer bleeding. The recent use of nonsteroidal antiinflammatory drugs (NSAIDs) must always be determined. A history of ingesting alcohol or a caustic substance is important to obtain. A history of cirrhosis or symptoms of cirrhosis such as ascites may suggest the need for urgent endoscopy to diagnose variceal bleeding. Other medical problems, such as prior aortic graft surgery, coagulopathies, cancer, or recent nose bleeds, may suggest likely diagnoses. Physical examination of the skin may provide diagnostic clues. Stigmata of cirrhosis, evidence of underlying malignancy (e.g., Kaposi sarcoma), or hereditary vascular anomalies may be revealed. The finding of lymphadenopathy or abdominal masses may suggest malignancy. Abdominal tenderness in the epigastrium is common in peptic disease. Hepatic or splenic enlargement may indicate liver disease or certain malignant disorders. When patients present with upper GI bleeding, a rectal examination may indicate the magnitude of blood loss by demonstrating maroon stool or melena in patients with severe bleeding or normal-colored stool in patients with minimal or recent bleeding.
Endoscopy Endoscopy has replaced barium contrast studies for the diagnosis of upper GI bleeding. The greater accuracy and therapeutic potential of endoscopy generally makes it the diagnostic procedure of choice. [40] Diagnostic endoscopy is viewed as a safe and simple procedure by patients and physicians, although morbidity rates of 1.0% and mortality rates of 0.1% have been reported. Endoscopy is contraindicated for an uncooperative patient or a patient with a suspected perforated viscus. Endoscopy can locate precisely the site of bleeding when bleeding continues or the stigmata of bleeding persist. In patients with massive hemorrhage, the source of bleeding occasionally cannot be discerned by endoscopy. In patients whose bleeding has stopped and in whom no stigmata of bleeding remain, a significant lesion seen on endoscopy (e.g., a clean ulcer base) is the presumed source. If more than one lesion or no lesion is identified, a definitive diagnosis cannot be made, and these patients must be restudied if they bleed again. Up to 24% of patients presenting with melena will have a nondiagnostic upper GI endoscopy, with the most common source of bleeding being the right side of the colon. [41] It is important not to mislead those caring for the patient by overstating the certainty of the localization or diagnosis of the bleeding site. The timing of the diagnostic endoscopy depends on the severity and suspected cause of the hemorrhage. Patients who fail to stop bleeding with simple supportive care require urgent endoscopy to guide further therapeutic techniques. Patients with underlying cirrhosis should undergo endoscopy as close to the bleeding episode as possible because they often have more than one source of potential hemorrhage and the diagnosis of bleeding varices alters future approaches to treatment. For most patients whose bleeding ceases, diagnostic endoscopy can be postponed for 24 hours without seriously altering diagnostic accuracy or clinical outcome. [42]
Angiography Angiography is used as a diagnostic examination for acute upper GI bleeding only if endoscopy has failed. The bleeding must be arterial and at a rate of at least 0.5 to 0.6 mL/min to detect extravasation. Angiography represents a therapeutic alternative for the delivery of intraarterial vasopressin in stress gastropathy or for embolization of bleeding ulcers or neoplasms in inoperable patients. Angiography may be used to diagnose difficult cases of recurrent GI bleeding from an unknown source. It provides an accurate diagnosis for 50% to 75% of patients but is associated with a serious complication rate of about 2%. Complications of angiography are related to catheter placement (e.g., dissection, thrombosis, false aneurysm) or to the contrast material (e.g., allergic reactions, renal failure). When embolic occlusion of vessels is used, the complication rate increases because of ischemic necrosis and perforation, although the use of minicoils instead of fluid embolization agents has reduced the risk. [43]

Causes and Therapy

The three major causes of upper GI bleeding are peptic ulcer disease, gastric erosions, and varices (Table 33-3). Their distribution depends on the patient population studied. In all endoscopic series, no diagnosis is made in 10% to 15% of patients, and as many as 20% to 30% of patients have more than one diagnosis. The endoscopically examined patients with no diagnosis of disease have an excellent prognosis.

DIAGNOSIS	PERCENTAGE OF TOTAL DIAGNOSES (%)
Duodenal ulcer	24.3
Gastric erosions	23.4
Gastric ulcer	21.3
Varices	10.3
Mallory-Weiss tear	7.2
Esophagitis	6.3
Erosive duodenitis	5.8
Neoplasm	2.9
Stomal ulcer	1.8
Esophageal ulcer	1.7
Miscellaneous	6.8

Adapted from Silverstein FE, Gilbert DA, Tedesco FJ. The national ASGE survey on upper gastrointestinal bleeding. Gastrointest Endosc 1981;27:73.

TABLE 33-3 Final Diagnosis of the Cause of Upper Gastrointestinal Bleeding in 2225 Patients

Peptic Ulcer Bleeding Duodenal, gastric, and stomal ulcers account for about 50% of upper GI bleeding episodes. Although several effective therapies have been developed for peptic ulcer disease during the past 20 years, these advances have had little or no impact on hospitalization rates for bleeding ulcers. Perhaps the reason is that ulcers can bleed without a prior history of peptic symptoms. [44] There also may be an increase in upper GI bleeding in the elderly because of the widespread use of NSAIDs, including aspirin. [45] Ulcers that are located high on the lesser curve of the stomach or on the posteroinferior wall of the duodenal bulb are more likely to rebleed. [46] Bleeding tends to occur when an ulcer erodes into the lateral wall of a vessel. The vessel often loops up to the floor of the crater and commonly protrudes with an aneurysmal dilatation. A vessel with an eccentric breach is thought to be more likely to be associated with continued or recurrent bleeding than a transected vessel because retraction contraction of a severed vessel is an important mechanism of hemostasis. [47] Patients with continued or recurrent ulcer bleeding have increased mortality rates. [48] Therapy is therefore directed at stopping bleeding and preventing recurrent bleeding.
Cessation of bleeding: endoscopic methods. The physician should perform lavage with tap water at room temperature for the important task of monitoring the rapidity of bleeding. Although lavage with iced saline solution and levarterenol has been suggested for vasoconstriction, controlled trials have not shown therapeutic benefit, and ice water increases patient discomfort. [49] A large-bore orogastric tube should replace the diagnostic nasogastric tube when cleaning for subsequent endoscopy is needed. Aliquots of 100 to 500 mL of water are instilled and removed, preferably by gravity drainage to prevent extensive suction trauma. For patients with persistent or recurrent hemorrhage from peptic ulcer disease, endoscopic control of bleeding, which is safer than emergency surgery, should be attempted. [50] Endoscopic methods can be divided into thermal and nonthermal (i.e., injection therapy) types. They are described in detail in Chapter 148. Numerous randomized controlled trials have documented the efficacy of injection therapy for bleeding peptic ulcers and for nonbleeding visible vessels. [51], [52] Nonthermal methods include injection of sclerosing agents such as alcohol or ethanolamine, injection of vasoconstrictors such as epinephrine, or injection of normal saline solution. [53] Because the choice of agent does not seem to alter efficacy, it is presumed that the mechanism of action of injection therapy is local tamponade. In contrast to thermal therapy, in which precise localization of the bleeding vessel is desirable, injection therapy is feasible even in an actively bleeding patient in whom visualization may be difficult. A spray of 3% hydrogen peroxide has been suggested as a means of clot dissolution to allow endoscopic visualization. [54] Comparisons of thermal and injection therapies suggest that they are equally effective. [55], [56] and [57] Some endoscopists use a combination of injection and thermal therapy, with initial injection to slow the bleeding or “clear the field” followed by coagulation of the identified vessel. Thermal methods include the neodymium:yttrium-aluminum garnet (Nd:YAG) laser, the heater probe, the argon plasma coagulator, and multipolar electrocoagulation. In this technique, direct probe pressure is used to tamponade the bleeding vessel, followed by raising the tissue temperature to coagulate and seal the vessel. Controlled trials of multipolar electrocoagulation show efficacy in reducing further bleeding, transfusion requirement, and need for surgery in actively bleeding ulcers and in nonbleeding visible vessels. [58], [59] A similar technique that uses pure thermal energy is the heater probe, which has also been efficacious in the treatment of bleeding ulcers and nonbleeding visible vessels. [60], [61] The major advantage of these two devices is that they are portable and relatively simple to use. The Nd:YAG laser is as effective as the heater probe and multipolar electrocoagulation, but its immobility, requirement for trained support personnel, and greater equipment expense reduce its attractiveness. [62] The argon plasma coagulator is the newest member of the thermal armamentarium. Although not as well studied, it appears to be equally as effective as other thermal methods. [63] Although most studies have not shown a reduction in mortality rates with endoscopic therapy, [64] a metaanalysis of numerous trials suggested a reduction, although this reached statistical significance

only for laser therapy. [16]

Cessation of bleeding: when endoscopy and medical therapy fail. After endoscopic treatment to control bleeding from peptic ulcers, bleeding recurs in 15% of patients. [48] A randomized controlled trial of repeated endoscopic treatment versus surgery for patients with recurrent ulcer bleeding concluded that endoscopic re-treatment is superior to surgery. [65] If hemorrhage is not stopped or if bleeding recurs again, surgery should be considered early because the risk for death increases as the patient becomes more unstable. Peptic ulcer bleeding is effectively treated with surgery, and this is safer than other therapeutic alternatives, such as angiography. Unlike bleeding from gastric erosions, bleeding from ulcers is not effectively stopped with intraarterial vasopressin, presumably because of the large size of the bleeding vessel in peptic ulcer disease. [66] Embolization by means of an angiographic catheter with polyvinyl alcohol particles or microcoils can be successful but requires significant expertise and causes complications. [67] This procedure should be reserved for the patient who is too unstable to undergo surgery.

Prevention of recurrent hemorrhage: pharmacological therapy. Most peptic ulcer rebleeding occurs within the first 3 days after presentation. The therapeutic goal is to prevent clot dissolution and allow healing of the underlying lesion. In vitro data show that coagulation and platelet function are better at neutral pH. [68] Although clot is not dissolved by acid, it is dissolved by gastric juice, suggesting that pepsin degradation may be important. [69] Because the activity of pepsin is pH dependent, it is reasonable to assume that a clot cannot dissolve if the gastric juice pH is high. Historically, multiple trials using histamine H_2 antagonists agents failed to demonstrate any improvement in survival for patients with upper GI bleeding. [70] Early studies of more potent acid suppression with continuous intravenous histamine H_2 antagonists or with proton pump inhibitors (PPIs) also failed to show efficacy. [71], [72] However, both of these studies included all bleeding ulcer patients, the majority of whom are at low risk for rebleeding, so that an insufficient number of patients may have been studied. In a comparison of omeprazole and placebo in high-risk ulcer patients with bleeding stigmata at endoscopy who were not treated endoscopically, high-dose omeprazole (40 mg twice daily) significantly lowered the rates of further bleeding and surgical intervention. [73] Although it is unlikely to replace endoscopic therapy, this study demonstrated the efficacy of potent acid suppression, perhaps through the stabilization of clotting activity. A placebo-controlled trial of high-dose infusion omeprazole after endoscopic treatment of bleeding peptic ulcers demonstrated a substantial reduction in the risk for rebleeding. [74] Potent acid suppression with high-dose intravenous or oral proton pump inhibition is now the mainstay of medical treatment for peptic ulcer bleeding (Table 33-4).

Decreases rebleeding and need for surgery	High-dose proton pump inhibitors
Efficacy not proven	Somatostatin Prostaglandins Tranexamic acid Intravenous vasopressin
Not effective	Histamine H_2 antagonists

TABLE 33-4 Medical Therapy for Peptic Ulcer Bleeding

Other drugs, such as somatostatin, [75], [76] prostaglandins, [77] tranexamic acid (an antifibrinolytic agent), [78] and intravenous vasopressin, [79] have not been proved to alter the course in bleeding ulcer patients. Mucosal protective agents such as sucralfate have not been studied in bleeding patients. It has been demonstrated that in patients without stigmata of recent hemorrhage, immediate refeeding and subsequent discharge are as safe as delayed refeeding for 36 hours. [80] The prevention of rebleeding by therapeutic endoscopic methods in high-risk ulcers with stigmata of bleeding is widely accepted. [81] However, some issues remain. First, the lack of standardized definitions of the various stigmata of recent hemorrhage and sufficient knowledge about each of their natural histories continues to be a problem. [82] Second, therapeutic endoscopy adds to the risk of the endoscopic procedure, with a risk for precipitating bleeding as high as 20% and with perforation occurring in as many as 1% of patients. Third, therapeutic endoscopy adds to the cost of treatment unless it successfully decreases the need for surgery or shortens the hospital stay and therefore must be applied judiciously.

Hemorrhage from Gastric Erosions The term *gastritis* should be reserved for the pathologist; *gastric erosions* or *gastropathy* should be used by the endoscopist. Histologically, gastritis is defined by epithelial distortion and inflammatory cell infiltrate, which may be chronic (i.e., predominantly plasma cells) or acute (i.e., polymorphonuclear cells). There may be biopsy evidence of severe histological gastritis with a normal endoscopic appearance. [83] This type of gastritis is not associated with upper GI hemorrhage. Endoscopically, gastropathy is defined by the gross appearance of mucosal hemorrhages, erythema, and erosions. An erosion is technically a break in the mucosa that does not cross the muscularis mucosae. Practically, most endoscopists define an erosion as an area of adherent hemorrhage or a defect in the mucosa with a necrotic base that is less than 3 to 5 mm in size and without significant depth. This type of gastric erosion may lead to upper GI bleeding and has several different causes. [84]

Drug-induced gastropathy. Gastropathy induced by aspirin or other NSAIDs is common. In almost all normal volunteers challenged with aspirin, mild hemorrhagic gastropathy develops that involves the proximal or entire stomach within 24 hours. [85] The bleeding associated with this acute damage is minimal and only rarely clinically apparent. If the aspirin is continued, adaptation and healing occur. In a smaller percentage of individuals continually exposed to NSAIDs, chronic erosive gastropathy, predominantly involving the antrum, or frank ulcer disease eventually develops. [86] This common type of erosive gastropathy is usually a self-limited disease that heals rapidly after removal of the offending agent. Interventional treatments such as therapeutic endoscopy or surgery are rarely required. Bleeding from NSAID-induced gastric erosions usually resolves spontaneously, and most of the serious upper GI bleeding associated with NSAID intake is the result of NSAID-induced peptic ulcers. [87] Although they have not been well studied in the treatment of bleeding from NSAID-induced gastric erosions, the use of high-dose PPIs seems reasonable. A more important issue in the management of NSAID-induced gastropathy is prophylaxis. Prostaglandins have been shown to be effective in preventing acute erosions from NSAIDs and in preventing chronic ulcers. [88] Although it has been difficult to demonstrate prevention of serious ulcer sequelae, such as hemorrhage or perforation, [89] a large study of more than 8500 patients did show a 40% reduction in serious NSAID-induced upper GI complications by misoprostol. [90] Prophylactic treatment with histamine H_2 blockers appears effective in duodenal disease but not in the stomach, [91] and sucralfate is not effective. [88] Omeprazole prevents damage in healthy volunteers [92] and is more effective than ranitidine for preventing ulcers. [93] It remains unclear who should receive prophylaxis because significant upper GI bleeding does not develop in most patients on NSAIDs, and routine prophylaxis with prostaglandins or PPIs would be extremely expensive. It seems reasonable to recommend additional treatment with prostaglandins or PPIs for the patient who has already demonstrated significant upper GI bleeding but requires continued NSAID intake. Additionally, treatment of concomitant *Helicobacter pylori* infection also decreases the risk for recurrent bleeding. [94] Another alternative is to change the NSAID to a less damaging agent, such as enteric-coated aspirin, which has been shown to cause fewer ulcers and less gastropathy in normal volunteers and patients with chronic rheumatic diseases. [95] Although less well studied, it has been suggested that the nonacetylated salicylates may also be gastric sparing. [96] Much better studied are the new specific cyclooxygenase-2 (COX-2) inhibitors, which have been shown to decrease not only gastric erosions but also upper GI tract bleeding. [97] An uncommon cause of drug-induced erosive gastropathy is hepatic artery pump chemotherapy. This treatment may cause hemorrhagic gastropathy, duodenitis, and frank ulcer disease. [98] The pathogenesis is presumably direct tissue injury from the chemotherapeutic agents or from ischemia secondary to catheter placement. Little is known about the management of this entity.

Gastropathy related to alcohol intake. Historically, it was thought that alcohol ingestion caused gastric erosions. In animal models, absolute alcohol causes severe hemorrhagic gastropathy, although lower doses of intragastric alcohol can produce adaptive cytoprotection. [99] These lower doses are closer to those obtained in human alcohol use. Alcohol consumption is a risk factor for upper GI bleeding only when it is excessive, four or more drinks per day. [100] Perhaps the historical "alcohol-induced gastritis" was actually the result of portal hypertension in patients with alcoholic liver disease. A recent endoscopic evaluation of GI hemorrhage in alcoholics found the causes to be peptic ulcer disease and disorders related to portal hypertension. [101]

Portal gastropathy. Portal gastropathy has been described as a diffuse erythematous, reticular, or mosaic pattern of the gastric mucosa. [102] The endoscopic spectrum of portal gastropathy includes a snakeskin appearance, with more severe cases having small areas of intense erythema (e.g., scarlatina rash), frank petechiae, or multiple bleeding spots. Vascular ectasia may be present throughout the stomach or show an antral predilection. Hyperdynamic congestion has been the presumed cause, [103] with a controversial role suggested for prior variceal sclerotherapy. [104] Increased gastric blood flow without congestion may also exist. [105] Although mild cases of portal gastropathy have a nonspecific appearance and no clinical consequences, the more severe variants are associated with overt and chronic bleeding. [106] The prevalence of portal gastropathy in patients with cirrhosis is 80%, with acute bleeding occurring in only 2.5% and chronic bleeding in 11%. [107] Portal gastropathy does not respond to treatment with acid suppression, although potent acid suppression has not been studied. Decreasing the portal pressure with propranolol is efficacious in the prevention of rebleeding. [108], [109] Portal systemic shunting in the unusual patient who has continued significant blood loss has been effective. [110] A transjugular intrahepatic portal systemic shunt (TIPS) would likely have similar efficacy.

Stress gastric erosions. An important cause of gastric erosions that cause major hemorrhage is illness severe enough to require an intensive care unit hospitalization, or "stress gastritis." It occurs in patients with respiratory failure, hypotension, sepsis, renal failure, thermal burns, peritonitis, jaundice, or neurological trauma. [111] The risk for bleeding in an individual patient varies with the number of such conditions. Endoscopic evidence of gastropathy is found in almost all intensive care unit patients, although only 2% to 10% of these patients have significant bleeding. [112] All treatment modalities for significant bleeding from stress erosions are associated with high failure rates and significant morbidity. Endoscopic therapy is usually the first and safest choice, although it has not been specifically studied in stress gastric erosions. The presence of multiple bleeding sites precludes its use in some patients. In contrast to the treatment of bleeding ulcers, angiographic control of gastric mucosal bleeding reportedly has a good success rate, perhaps because of the small vessel size in these superficial lesions. Intraarterial vasopressin controls hemorrhage in 80% to 90% of successfully catheterized patients with gastric erosion. [113] Unfortunately, even skilled angiographers can catheterize only 75%

of patients. Intravenous infusion of vasopressin has not been as well studied but is also reported to be effective. [114] The operative mortality rate is extremely high for patients bleeding from stress gastric erosions, and rebleeding after surgery is common; surgery is reserved as a last alternative. [115] The major emphasis in the management of stress gastric erosions is prophylaxis. Although there has been some controversy about the cost-effectiveness of treating all patients at risk, [116] a review of all studies strongly supports routine prophylaxis for all patients ill enough to be in an intensive care unit. [117] The use of high-dose antacids, [118] histamine H_2 blockers, [119] or sucralfate [120] in patients at risk has decreased the incidence of bleeding. Misoprostol is as effective as high-dose antacids in preventing stress erosions, although both caused diarrhea in 25% of patients. [121] Because high-dose antacids cause considerable side effects and nursing inconvenience, intravenous histamine H_2 blockers, PPIs, and sucralfate are more popular. Sucralfate administered through the nasogastric tube also appears as effective as high-dose antacids and is cheaper than intravenous histamine H_2 blockers. [122] However, a large multicenter trial showed a significantly lower rate of clinically important bleeding with ranitidine than with sucralfate. [123] This study also challenged a previous suggestion that agents such as sucralfate, which improve the mucosal defense without altering intragastric pH, may result in lower rates of nosocomial pneumonia in patients on respirators. [124] The validity of the hypothesis that gastric bacterial overgrowth occurs with acid-reducing therapy and that endotracheal intubation establishes a pathway for colonization of the respiratory tract continues to be questioned. [123], [125]

Esophageal Varices The first episode of upper GI hemorrhage from esophageal varices is associated with a mortality rate of 21% to 50%, and in years past, two thirds of these patients died within 1 year. [28] These bleak mortality figures may have improved slightly in more recent studies, [126] although a variable delay in trial entry makes comparison of mortality rates among studies difficult. [127] This high mortality rate attests to the difficulty of managing acute variceal bleeding and preventing further bleeding. It also may explain why so many therapeutic alternatives have been proposed. Most patients with varices have underlying cirrhosis, and this contributes to the high mortality rate; 40% die of associated medical problems. It has been estimated that one fourth to one third of patients with cirrhosis hemorrhage at least once from varices. [128] Despite the increasing array of therapeutic options for variceal hemorrhage, there has been little change in long-term survival.

Determinants of variceal rupture. Portal hypertension must be present with pressures of 12 mm Hg or greater for varices to develop. However, the degree of pressure elevation does not correlate with the risk for rupture. [129] Portal pressures may be similar in patients with no evidence of varices and in those with large varices. Once a threshold portal pressure develops that permits varices, other factors control their formation and their risk for rupture. It has been suggested that directly measured intravariceal pressure may correlate better with the risk for hemorrhage. [130] The portal pressure measured within 48 hours after the bleeding event may also correlate better with bleeding. A higher risk for rebleeding has been demonstrated in patients admitted with variceal hemorrhage who have higher portal pressures. [131] Esophagitis has not been shown to predispose to variceal bleeding, even though intuitively it seems reasonable that erosions on top of esophageal varices may cause a vessel to bleed. This remains somewhat controversial. [132] Gastroesophageal reflux as measured by a pH probe is not more common in patients with a history of variceal bleeding than in controls. [133] As in other types of GI bleeding, the recent use of aspirin predisposes to a first episode of variceal bleeding. [134] An important predictor of variceal hemorrhage is the size of the varices. Several studies have shown that large varices are more likely to bleed than small ones. [135] Wall tension is a function of diameter and wall thickness, and it is not surprising that larger varices are more likely to rupture. Another endoscopic finding of value in predicting variceal bleeding is the appearance of the vessel wall. Certain colors of varices are thought to indicate impending hemorrhage. The red color sign is the result of microtelangiectasia of the varix. Variants of this sign are red wale marks, which look like whip marks; cherry red spots 2 mm in diameter; hemocystic spots, which are round, crimson projections larger than 4 mm that look like blood blisters; and diffuse redness. All the red color signs and a blue color are thought to be risk factors for bleeding. Red color signs have been correlated with increased variceal pressure as assessed by direct needle puncture. [136] When examined by endoscopic ultrasonography, hemocystic spots appear as saccular aneurysm projections on the variceal surface. [137] The white nipple sign on a varix, thought to be a platelet-fibrin plug, is considered diagnostic of previous bleeding but not predictive of rebleeding. [138] The presence of cutaneous vascular spiders also correlates with a risk for hemorrhage from esophageal varices. [139] A prognostic index for variceal hemorrhage based on the three variables of variceal size, presence of red wale marks, and a modified Child classification of underlying liver disease was able to identify subsets of patients with a 1-year incidence of bleeding ranging from 6% to 76%. [140] The value of these predictors of variceal rupture depends on the usefulness of prophylactic therapy for variceal bleeding (Table 33-5).

Factors correlated with risk for bleeding	Variceal size
	Red marks on varices
	Severity of liver disease
Probably correlated with bleeding	Intravariceal pressure
	Portal pressure during hospitalization for bleeding
	Cutaneous spider angiomata
Not correlated with bleeding	Esophagitis
	Degree of portal pressure elevation over 12 mm Hg

TABLE 33-5 Predictors of Variceal Rupture

Management of acute variceal hemorrhage. Variceal bleeding is often the most rapid type of upper GI hemorrhage. The emphasis in acute management is on resuscitation. More than 90% of episodes of variceal bleeding cause a drop in the hematocrit to below 30%, so that transfusions are required. However, as in upper GI hemorrhage with other causes, 70% to 80% of cases resolve without specific intervention. Urgent endoscopy is indicated for a patient with a suspected variceal source because the methods of treating the acute bleeding episode and preventing recurrent bleeding differ from those for upper GI bleeding with other causes. In more than half to two thirds of patients with cirrhosis who present with bleeding, the sources are nonvariceal, and many of these patients have more than one lesion. [141] For this reason, early endoscopy is mandatory to determine the site and cause of bleeding. In the acutely bleeding patient, intravenous octreotide, a synthetic analog of somatostatin, is often begun as soon as the diagnosis is made. This newer agent has replaced vasopressin because of its ease of use, although vasopressin has been the historical treatment to which octreotide has been compared. The efficacy of both of these agents has been difficult to demonstrate convincingly. Compared to placebo, somatostatin has had conflicting results, [142], [143] although it has been shown to be equally efficacious with terlipressin and vasopressin. [144], [145] Several studies combining sclerotherapy with octreotide (or vapreotide) versus sclerotherapy alone found less bleeding with combination therapy and no difference in mortality rates. [146], [147], [148] and [149] Historically, vasopressin was used for splanchnic arteriolar vasoconstriction resulting in decreased portal pressure, although it remains controversial whether this effect is maintained in the face of severe hemorrhage. [150] Although early trials appeared to show marginal efficacy for vasopressin, [151] cardiovascular complications were common. This led to the concomitant use of nitroglycerin, which both decreased complications and improved efficacy. [152] An alternative to vasopressin for control of variceal hemorrhage is a synthetic analog of vasopressin, terlipressin, which is reportedly more effective than placebo or vasopressin although not yet available in the United States. [153] When combined with nitroglycerin, it is as effective as balloon tamponade [154] and may be as effective as sclerotherapy. [155] Pharmacological constriction of the lower esophageal sphincter with metoclopramide lowers intravariceal pressure, [156] although this is more effective when combined with intravenous nitroglycerin. [157] Only limited data are available regarding metoclopramide in the treatment of bleeding varices. [158] After supportive medical therapy and octreotide infusion have been started in the patient with variceal bleeding, the next step in management is urgent band ligation or sclerotherapy. If a skilled endoscopist is not available or, rarely, if the bleeding is too rapid to permit endoscopy, balloon tamponade is indicated. Variceal band ligation is a superior alternative to sclerotherapy for acutely bleeding patients and for the prevention of rebleeding (see Chapter 147) (Fig. 33-1). Ligation of varices has a significantly lower complication rate than sclerotherapy [159], [160], [161], [162] and [163] and may further lower the rebleeding rate [159], [160], [162] and improve survival. [159] Variceal ligation also requires fewer sessions to achieve variceal obliteration and is now technically simpler with multiple-shoot devices. [164] The combination of band ligation and sclerotherapy compared to ligation alone has had conflicting results. Some studies show no benefit for the combined therapy and a higher complication rate than that for ligation alone. [165], [166] Other studies suggest that combined therapy may be more effective at variceal eradication, [167], [168] with a decrease in recurrent bleeding. [169] Although emergent sclerotherapy has a success rate for controlling bleeding of 85% to 95%, [170], [171] it is now relegated to rapidly bleeding patients in whom visualization is insufficient for band ligation or to patients with varices too small to ligate. A new ligation technique in which tiny detachable snares are used to treat esophageal varices has been developed [172] and appears comparable to band ligation therapy. [173]

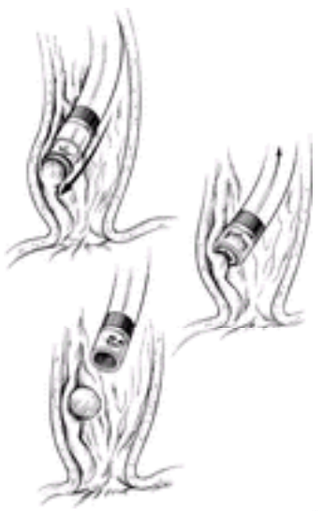

FIGURE 33-1. Endoscopic band ligation involves the application of a small suction chamber to a standard endoscope tip, which allows the varix to be pulled into the stretched band. A trigger device then releases the band around the varix base, causing subsequent necrosis and scarring.

Emergency sclerotherapy in cirrhotics with upper GI hemorrhage is associated with bacteremia, and the administration of prophylactic antibiotics after emergency endoscopy decreases the incidence of clinical infections. [174] Antibiotic prophylaxis is now recommended for all patients with cirrhosis who present with upper GI bleeding. [175] When endoscopic and medical therapy fails to control variceal bleeding, balloon tamponade is indicated as a temporizing maneuver. Balloon tamponade with the Sengstaken-Blakemore tube is effective in achieving hemostasis in 70% to 90% of patients. [176] Unfortunately, hemostasis is often temporary, with rebleeding occurring in 30% to 50% of patients after the balloon is deflated. An adapted Sengstaken-Blakemore tube with an esophageal aspiration port or the Minnesota tube with a built-in esophageal port is probably the most popular device (Fig. 33-2). [177] The complication rate of balloon tamponade is 10% to 30%; complications include esophageal perforation, aspiration pneumonia, malfunction requiring replacement, chest pain, gastric erosion, and agitation. [178] Endotracheal intubation before balloon insertion has been recommended to decrease complications. [179]

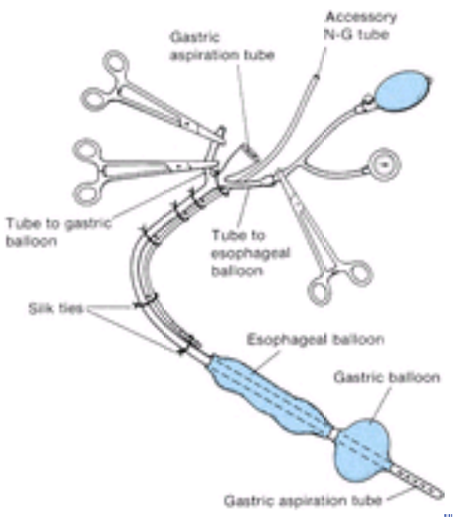

FIGURE 33-2. Modified Sengstaken-Blakemore tube. Also available is the Minnesota tube, which has a built-in esophageal port. (Adapted from ref. [177].)

TIPS, a nonsurgical method of creating portosystemic shunting, has gained popularity. [180] TIPS has been used to manage acute variceal hemorrhage and prevent rebleeding. An expandable metallic stent is placed between the hepatic and portal veins within the liver by means of an angiographically guided catheter (Fig. 33-3). Technical success is achieved in 93% to 96% of patients, with a 10% to 15% procedure complication rate and a 25% incidence of hepatic encephalopathy. [181], [182] In one third of patients, TIPS stenosis or occlusion develops by 1 year of follow-up, although this is successfully treated by redilation or placement of an additional stent. TIPS has been proposed as an effective bridge to liver transplantation because it avoids surgery in patients with advanced liver disease. [183]

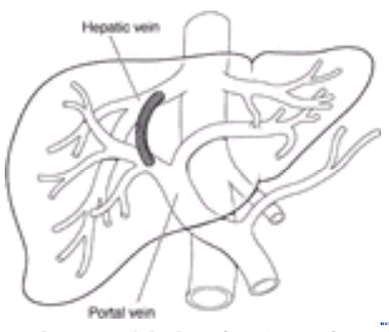

FIGURE 33-3. The transjugular intrahepatic portosystemic shunt (TIPS) is a metal expandable stent that is placed angiographically between branches of the hepatic and portal veins to create a nonsurgical shunt between the portal and systemic venous systems.

The surgical shunting of portal blood to the systemic circulation to control ongoing hemorrhage from varices is reserved for patients resistant to other therapies and has become quite uncommon since the advent of TIPS. The mortality rate for these emergency shunts is as high as 50% to 80%, which has dampened enthusiasm for the procedure. [184] This mortality rate is in contrast to that of surgery to prevent recurrent variceal bleeding. Although a better survival rate for emergency shunting has been reported, the study may have included patients who would have been controlled with other, simpler techniques at other centers. [185] Staple gun transection of the esophagus has been advocated as the simplest type of emergency surgery for active variceal bleeding and has been proposed as the salvage procedure of choice after failure of acute sclerotherapy. [186] However, because of lower postsurgical rebleeding rates, portosystemic shunts continue to be favored by many surgeons.

Prevention of recurrent variceal hemorrhage. One third of patients surviving a variceal hemorrhage rebleed within 6 weeks. [28] Death from bleeding occurs in 40% to 60% of these patients. Prevention of recurrent hemorrhage is an important part of therapy. Therapies to prevent rebleeding include band ligation or sclerotherapy, medical therapy with propranolol or nitrates, TIPS, and rarely surgical shunts. An improvement in the mortality rate is particularly difficult to demonstrate with all these methods, suggesting that the risk for death is more likely correlated to the severity of the underlying liver disease than to rebleeding. Endoscopic therapy is thought to be superior to medical management for the prevention of rebleeding, although controversy continues on this point. Controlled trials of ß $_1$-blockade compared with sclerotherapy showed lower rebleeding rates with sclerotherapy, [187] and a metaanalysis confirmed this superiority, although there was no difference in survival. [188] Combination therapy with nadolol and nitrates proved superior to sclerotherapy, with fewer complications and a lower rebleeding rate. [189] Since variceal banding has replaced sclerotherapy at most centers for the prevention of rebleeding, the combination of nadolol plus nitrates must be compared to band ligation before the ultimate role of medical versus endoscopic therapy for the prevention of rebleeding is determined. The risk for rebleeding is greatest in the first few weeks after the initiation of a endoscopic therapy regimen, before the varices are obliterated. Propranolol has been advocated for the reduction of rebleeding rates during endoscopic sclerotherapy before variceal obliteration, although the results have been conflicting. [190], [191] A study of variceal ligation versus ligation plus nadolol and sucralfate showed that the combination of ligation and medical therapy decreased both variceal and nonvariceal bleeding. [192] Subcutaneous octreotide has also been combined with endoscopic therapy to prevent rebleeding. [193] Propranolol was proposed more than 20 years ago to prevent rebleeding in patients who had already presented with variceal hemorrhage. [194] Propranolol decreases portal pressures in laboratory animals and humans, although the response is not uniform. [195] Despite early controversy, subsequent studies and several metaanalyses show efficacy with propranolol in doses that reduce the heart rate by 25%. [196] Nadolol is a popular alternative to propranolol because it has to be administered only once daily and is not metabolized by the liver. Isosorbide mononitrate has been shown to be as effective as propranolol [197] and has been used in patients who either cannot tolerate propranolol or do not respond to it satisfactorily. The combination of propranolol and isosorbide mononitrate enhances portal pressure reduction compared to propranolol alone [198] and has better efficacy in the prevention of rebleeding. [199] Despite some early concerns about the effect of nitrates on renal function and ascites, the combination of nadolol and isosorbide-5-mononitrate appears safe. [200] TIPS has

been compared to sclerotherapy and band ligation for the prevention of rebleeding. [201], [202], [203] and [204] TIPS has also been compared to a combination of endoscopic and medical therapy with either sclerotherapy plus propranolol or band ligation plus propranolol. [205], [206] Most of these trials showed less rebleeding in the patients receiving TIPS, although survival rates and costs were similar and the incidence of encephalopathy was higher with TIPS treatment. It appears that once the acute variceal bleeding episode has been controlled with endoscopic treatment, the choice for prevention of rebleeding is either further band ligation to achieve obliteration, band ligation plus medical therapy, TIPS, or medical therapy alone with a combination of nadolol and isosorbide mononitrate. Surgical shunts decompress the portal system by diverting blood into the systemic circulation. Several types of shunts have been used, including end-to-side portacaval, side-to-side portacaval, mesocaval, and splenorenal shunts. Multiple studies have confirmed their efficacy in preventing rebleeding. [207] Unfortunately, encephalopathy developed postoperatively in 10% to 40% of patients. This led to the development of the distal splenorenal (i.e., Warren) shunt, which spares portal blood flow to the liver (Fig. 33-4). The proponents of this shunt continue to report lower rates (4%–15%) of encephalopathy. [208] This has been confirmed in some controlled trials, [209], [210] although other studies comparing nonselective and distal splenorenal shunts have shown similar rates of operative mortality, late mortality, incidence of encephalopathy, and shunt occlusion. [211], [212] It appears that the incidence of encephalopathy is more related to the severity of underlying liver disease than to the type of surgery performed. Some surgeons reserve the selective shunt for patients with preserved hepatopetal (i.e., intestine-to-liver) blood flow, although this may be unnecessary. A partial portacaval shunt (i.e., small-diameter or H-graft) has also been advocated as a mechanism to preserve hepatic portal blood flow and decrease postoperative encephalopathy. [213], [214] This shunt reportedly does not compromise subsequent liver transplantation, although it may have a higher incidence of occlusion and encephalopathy than the distal splenorenal shunt. [215]

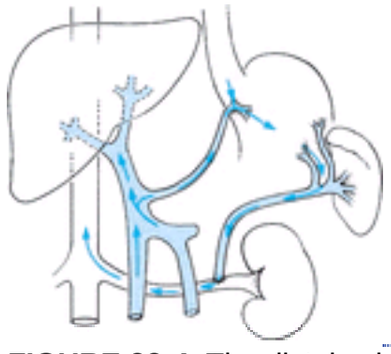

FIGURE 33-4. The distal splenorenal shunt was designed to prevent diversion of gut venous flow to the liver.

Despite the efficacy of all shunts in preventing rebleeding, controlled trials have not shown improved mortality rates. [216] A randomized comparison of surgical shunt with the effects of sclerotherapy showed similar survival and efficacy in the prevention of variceal rebleeding, with sclerotherapy having lower costs. [217] Most physicians favor endoscopic therapy as the initial therapy, with surgery reserved for the patients who fail an adequate attempt at variceal obliteration. For patients who are transplantation candidates or have poor hepatic function, TIPS is generally preferred to surgical shunt. For patients with well-preserved hepatic function, controversy continues about the choice of endoscopic treatment or surgery. [218], [219] Two other surgical methods of portal decompression have been used. The Sugiura procedure involves esophageal transection with paraesophagogastric devascularization. [220] Many modifications of this esophageal transection and devascularization procedure exist. The results of the Sugiura procedure in patients with nonalcoholic liver disease have been excellent, but some surgeons have documented high rebleeding rates. [221] A procedure that has not been extensively studied is splenopneumopexy. [222] This can be performed in patients with diffuse splanchnic venous thrombosis when alternative shunts are not technically possible. It involves resection of part of the left diaphragm with apposition of the abraded surfaces of the spleen and left lung to allow decompressive collaterals to form. Hepatic transplantation is the accepted treatment for otherwise healthy patients with end-stage liver disease. When these patients present with bleeding varices, management of the bleeding should avoid abdominal operations, which makes transplantation more difficult.

Prophylactic treatment of variceal hemorrhage. Because 30% of cirrhotics experience variceal hemorrhage and the mortality rate associated with even one episode is very high, prevention is important. Prophylactic portacaval shunts have successfully prevented bleeding but do not improve survival. [223] The benefit of not bleeding may be offset by the operative morbidity and mortality, and this surgical treatment has completely fallen out of favor. Less invasive methods of prophylactic therapy include endoscopic and medical treatment. It was hoped that identification of a group of cirrhotics at high risk for variceal bleeding (e.g., those with large varices with red color signs) would select a subset of patients who would benefit from preventive sclerotherapy. However, results of multiple trials continue to be contradictory, with most studies failing to show benefit. [224], [225], [226], [227], [228] and [229] Prophylactic sclerotherapy is not routinely recommended, although the safer procedure of band ligation has been advocated. [230] A recent comparison of band ligation to propranolol for the prophylaxis of variceal bleeding in patients with grade 3 or 4 varices showed superiority for endoscopic treatment. [231] Confirmation of this result is needed before band ligation can be routinely recommended. Propranolol or nadolol is the least invasive therapy of the prophylactic alternatives and therefore the most attractive, but results of controlled trials have been controversial. [232] However, several controlled studies and two metaanalyses concluded that nonselective ß-adrenergic antagonists have a beneficial effect in prophylaxis of a first variceal hemorrhage. [233], [234] and [235] This relatively complication-free treatment can be recommended for patients with large or moderately sized varices. Cirrhotic patients with small or no varices should be followed for the development of varices. The best clinical predictor of the presence of varices is splenomegaly or a platelet count below 88,000/mm^3. [236], [237] A prospective screening study suggests that patients should undergo endoscopy annually if the initial endoscopy reveals small (grade 1) varices, and that screening every other year is adequate for patients with no varices. [238] In the 3% to 20% of patients who cannot tolerate propranolol, isosorbide-5-mononitrate is an effective alternative. [197] Because combination therapy with propranolol and isosorbide mononitrate is more effective in reducing portal pressure, [198] it may become the standard for prophylaxis of first variceal hemorrhage.

Gastric Varices Gastric varices usually accompany esophageal varices, although they may occur alone. In a study of 568 patients with portal hypertension, primary gastric varices were present in 20% of patients, and secondary gastric varices, which developed after treatment of esophageal varices, occurred in 8% of patients. [239] Risk factors for hemorrhage from gastric fundal varices are similar to those for esophageal variceal bleeding: larger variceal size, the presence of red marks, and relatively severe underlying liver disease. [240] Several classification schemes that depend on the variceal location have been suggested, [239], [241] and it appears that the various subsets have different natural histories and responses to treatment. [239], [242] Gastric varices that develop after sclerotherapy for esophageal varices are referred to as *secondary,* although the propensity for this to occur is controversial. [243] Gastroesophageal varices that cross the gastroesophageal junction along the lesser curvature are most common and often disappear spontaneously with the obliteration of esophageal varices. Gastroesophageal varices that cross the junction along the greater curvature are less common but are associated with a higher incidence of bleeding and are much less likely to disappear after esophageal variceal obliteration. Isolated gastric varices that do not connect with esophageal varices usually occur in the gastric fundus and are the most difficult to treat endoscopically. [244] Isolated gastric varices located elsewhere in the stomach or duodenum (i.e., ectopic varices) are more commonly secondary varices, occur most often in the antrum, and rarely bleed. Isolated gastric varices in the fundus may be the result of splenic vein thrombosis, which can be verified with angiography. These patients are best treated with simple splenectomy that adequately decompresses their varices. They have an excellent prognosis because they do not have underlying liver disease. [245] Splenic vein thrombosis may occur as a complication of pancreatitis secondary to contiguous inflammation from the body and tail of the pancreas. [246] Bleeding gastric varices commonly are associated with large esophageal varices and are the result of underlying liver disease. When patients with gastroesophageal varices bleed, they are more likely to bleed from their esophageal varices, although bleeding gastric varices are associated with higher transfusion requirements. [239] Acute sclerotherapy to stop bleeding is reportedly effective for all types of gastric varices. [247] Sclerotherapy to prevent rebleeding has been advocated for gastric varices that cross the esophageal junction but persist after esophageal variceal obliteration, [239] although sclerotherapy in the stomach may have a higher complication rate. Alternative endoscopic treatments for gastric varices include injection of thrombin, [248] snare ligation, [249] and a combination of sclerotherapy with percutaneous transhepatic obliteration. [250] Injection of cyanoacrylate glue is reportedly effective [251], [252] and may be superior to sclerotherapy with ethanolamine. [253] Shunting with TIPS [254], [255] or surgery may be the best treatment for preventing rebleeding from gastric varices, especially from isolated fundal varices.

Mallory-Weiss Tear Mallory-Weiss tears occur near the gastroesophageal junction in the gastric or esophageal mucosa. They are caused by retching, perhaps with forceful gastric mucosal prolapse as identified at endoscopy. [256] They account for 5% to 10% of cases of upper GI hemorrhage. [257] There is usually a history of vomiting foodstuffs before hematemesis, although bleeding can occur with the first emesis. Many patients with Mallory-Weiss tears have a history of alcohol intake or have portal hypertension. [258] The bleeding usually resolves with conservative management, although endoscopic therapy may be required. [259] Rarely, patients with rebleeding or uncontrollable hemorrhage require oversewing of the bleeding mucosa or angiographic treatment.

Esophagitis and Esophageal Ulcers Esophagitis and esophageal ulcers account for approximately 8% of cases of upper GI hemorrhage. The primary cause of these lesions is peptic reflux, but other causes include irradiation, infectious esophagitis associated with pathogens such as *Candida* or herpesvirus, pill-induced damage, and sclerotherapy-induced ulcers. The presentation of bleeding esophageal lesions is similar to that of peptic ulcer disease. Persistent or recurrent bleeding should be treated aggressively with therapeutic endoscopic or angiographic techniques because esophageal lesions are less amenable to surgery than peptic ulcer disease. When the ulcer is caused by sclerotherapy, the physician must be certain of the source of bleeding because recurrent varices are managed differently.

Sucralfate has been suggested for the treatment of these chemically induced ulcers, although a controlled trial did not prove efficacy. [260] Omeprazole was found effective in the treatment of sclerotherapy-induced ulcers in an uncontrolled trial. [261]

Erosive Duodenitis Hemorrhage resulting from erosive duodenitis is closely related to duodenal ulcer bleeding but is usually less severe because the lesions are shallower and involve smaller vessels. It accounts for approximately 5% of cases of upper GI hemorrhage. The hemorrhage usually occurs in patients with a history of peptic ulcer disease or similar risk factors. Bleeding from duodenitis is almost always self-limited, rarely requiring therapeutic endoscopic intervention.

Neoplasms Neoplasms of the stomach, esophagus, or duodenum are uncommon causes (2%–5%) of upper GI hemorrhage. Bleeding from these lesions is usually self-limited, and treatment is ultimately in the hands of the oncologist or surgeon. If persistent or recurrent bleeding occurs in a patient unsuitable for surgical resection, endoscopic therapy or angiographic arterial embolization may be used. [262], [263] Intraarterial vasopressin is not usually effective because of the large size of the bleeding vessels.

Angiodysplasia and Gastric Antral Vascular Ectasia Vascular ectasia or angiodysplasia, which occurs less commonly in the stomach or duodenum than in the colon, is the cause of upper GI bleeding in 5% to 7% of patients (see Chapter 130). [27], [264] Often found in patients of advanced age, it has been associated with chronic renal failure, hereditary hemorrhagic telangiectasia (Osler-Weber-Rendu syndrome), and prior radiation therapy. [265], [266] Angiodysplasia has also been associated with aortic valve disease, although this remains controversial. [267] The diagnosis is usually made endoscopically by visualizing small, punctate, bright red, vascular mucosal lesions. Controlled studies of therapeutic alternatives are not available, but most clinicians would first attempt endoscopic coagulation techniques. These have been successful, although associated with high rebleeding rates in patients with hereditary (Osler-Weber-Rendu) lesions. [268] In cases of vascular ectasia associated with chronic renal failure and its attendant prolonged bleeding time secondary to platelet dysfunction, estrogen-progesterone therapy has been reported beneficial. [269] Similar efficacy has been reported in patients with normal renal function and chronic GI blood loss from vascular ectasia. [270] This suggests that the abnormal platelet function present in renal failure may not be a prerequisite for estrogen-progesterone treatment. One controlled crossover trial confirmed this efficacy. [271] An unusual variant of gastric vascular ectasia is the watermelon stomach, [272] also called *gastric antral vascular ectasia (GAVE)*. The endoscopic appearance is a jagged column of vessels that run along the top of longitudinal rugal folds, traversing the antrum and converging on the pylorus. This vascular aggregate resembles the stripes on a watermelon. Endoscopic biopsy or resected specimens show dilated mucosal capillaries with focal thrombosis and fibromuscular hyperplasia of vessels in the lamina propria. The condition has been associated clinically with hypochlorhydria, systemic sclerosis, and portal hypertension. [273], [274] Although GAVE occurs in patients with cirrhosis, it is distinct from portal gastropathy and does not appear to respond to lowering portal pressures. [275] Endoscopic coagulation treatment with the Nd:YAG laser, multipolar electrocoagulation, argon plasma coagulator, or heater probe may be useful. [276] Estrogen-progesterone treatment has also been effective in decreasing blood loss, [277] even in patients with underlying cirrhosis. [278]

Aortoenteric Fistula Aortoenteric fistulae usually involve the aorta but occasionally arise from branches of the celiac axis. Most aortoenteric fistulae are secondary to prior aortic Dacron graft surgery, [279] although they may occur as primary fistulae caused by atherosclerotic vessels or more rarely mycotic aneurysms, tuberculosis, or syphilis. [280] Aortoenteric fistulae almost always involve the third portion of the duodenum, although they may rupture into the jejunum, ileum, stomach, and colon. In patients with Dacron grafts, the fistula usually arises from the proximal portion of the graft and may be associated with false aneurysms. The classical clinical presentation is a “herald” bleed that occurs and stops spontaneously hours or occasionally weeks before the exsanguinating hemorrhage. A high index of suspicion is necessary to make the diagnosis because the fistula is difficult to discern by x-ray film, endoscopy, and angiography. Arteriography is often not helpful and may delay surgery. Abdominal computed tomography may demonstrate evidence of the fistula. If there is a history of aortic Dacron graft surgery in a patient presenting with GI hemorrhage, endoscopy should be performed to rule out other causes of bleeding, and the endoscopist should attempt to reach the third portion of the duodenum in an effort to visualize the fistula. If not identified, a fistula should be the presumed source of bleeding, and the patient should undergo surgery.

Hematobilia and Hemosuccus Pancreaticus *Hematobilia* is defined as hemorrhage into the biliary tract from any cause. Hemorrhage traversing the pancreatic duct has been called *hemosuccus pancreaticus,* although it is often included with hematobilia because it exits the ampulla. The mortality associated with hemorrhage from both these sites is significant (20%–50%). The most common cause of hematobilia is prior liver or biliary tree trauma, including prior percutaneous liver biopsy or transhepatic cholangiography. Extrahepatic or intrahepatic aneurysms of the hepatic artery or its branches are often caused by the trauma and may communicate with the bile ducts. Angiographic treatment with embolization is usually effective. [281] Less common causes of hematobilia are extrahepatic or intrahepatic tumors, gallstones, and cholecystitis. Hemosuccus pancreaticus represents bleeding from peripancreatic blood vessels into a pancreatic duct. Blood emanates from digested peripancreatic pseudoaneurysms or veins that rupture into a pseudocyst or from true aneurysms of the peripancreatic vessels that rupture into pancreatic parenchyma and ducts. [282] This usually occurs in patients with a history of chronic pancreatitis and pseudocysts. The diagnosis may be made by endoscopy, with visualization of blood coming from the papilla, although it is easily missed when the bleeding has ceased. Angiography is indicated to define the bleeding site and may be used for treatment by embolizing the vessel. [283] If embolization therapy is not successful, surgery may be required.

Dieulafoy Disease The Dieulafoy lesion is defined as a ruptured, thick-walled artery that is larger than other surrounding submucosal vessels, with little or no associated ulceration. The cause of bleeding is not thought to be a primary ulcerative process but rather pressure erosion of the overlying epithelium by this ectatic vessel. [284] The Dieulafoy vessel usually occurs in the fundus, although up to 34% of the lesions develop outside the stomach. [285] Endoscopically, it appears as a round mucosal defect with a protruding artery at the base. Patients present with hematemesis or melena without any relevant history. Endoscopic injection therapy, electrocoagulation techniques, band ligation, and hemoclipping have been successful in stopping bleeding in most patients, although surgery is occasionally required. [285], [286]

Factitious Bleeding or Bleeding from Nongastrointestinal Sources Some patients present with hematemesis or melena that does not originate from a GI source. Usually, it is caused by swallowing blood from epistaxis, hemoptysis, or oral lesions. These diagnoses are best determined by a careful history and physical examination. Endoscopy to rule out GI sources may be required if the diagnosis remains uncertain. Rarely, patients present with factitious bleeding. They may cause themselves to bleed by venopuncture and then swallow the blood before presentation. A high index of suspicion is necessary to make this diagnosis.

ACUTE LOWER GASTROINTESTINAL BLEEDING

Lower GI bleeding is defined as bleeding from a source below the ligament of Treitz. The annual incidence of lower intestinal bleeding is estimated to be 20 to 27 per 100,000 adults at risk. [3] When patients hospitalized for GI bleeding are identified, lower GI sources account for one fourth to one third of all bleeding events. [287], [288] Lower GI bleeding is more common in men than in women, and the incidence rate increases with age, with more than a 200-fold increase noted from the third to the ninth decade of life. [3] Overall mortality rates for lower GI bleeding are consistently below 5%, which is lower than mortality rates for upper GI bleeding.

Diagnostic Approach

The diagnosis should be sought in all patients with lower GI bleeding unless their overall prognosis is too poor to warrant further tests. In most patients whose bleeding ceases spontaneously, an elective colonoscopy after routine preparation is indicated. Patients with continued bleeding require urgent diagnosis. If a perianal or rectal source is suspected, simple proctoscopy can be performed quickly and may provide the diagnosis. For most colonic bleeding, a more thorough examination is required. Unless the bleeding is massive, rapid intestinal lavage allows adequate preparation for urgent colonoscopy. For patients who are bleeding too rapidly for a cleaning preparation or when urgent colonoscopy is not available, angiography is indicated. As with upper GI bleeding, most angiographers prefer prior radiolabeled nuclide scans to demonstrate active bleeding and direct the examination, [289] although the value of this practice has been questioned. [290] Before angiography, nasogastric lavage and even upper GI endoscopy should be considered to rule out an upper GI source of bleeding.

History and Physical Examination A thorough history and physical examination often point to the correct diagnosis. For example, a prior diagnosis of hemorrhoids or inflammatory bowel disease is important. Symptoms that are associated with bleeding, such as abdominal pain or diarrhea, suggest specific diagnoses. A recent history of anorexia or weight loss or an abdominal mass found during physical examination may indicate an underlying malignancy.

Colonoscopy Colonoscopy has replaced the barium enema study in the diagnostic evaluation of lower GI bleeding. Several series have demonstrated the superior diagnostic sensitivity of colonoscopy, even in comparison with double-contrast barium enema studies. When the indication for the diagnostic study is GI bleeding, a colonoscopy is usually indicated regardless of the result of the barium study because false-negative results occur in 10% to 20% of barium studies, and abnormal findings on a barium study lead to colonoscopy for biopsy or therapeutic maneuvers. For these reasons, most clinicians favor colonoscopy as the primary examination. In the occasional patient in whom complete colonoscopy is not technically feasible or the colonoscopy is nondiagnostic, a barium study is helpful. Patients with rapid, ongoing blood loss require diagnostic angiography or urgent colonoscopy after purge. Although the traditional view is that colonoscopy in patients with severe hematochezia is impractical because of inadequate visualization, colonoscopy is feasible and useful after prior rapid cleaning. [287], [288] Urgent colonoscopy made a final diagnosis of colonic lesions in 74% to 90% of patients. In one series that included prior upper GI endoscopy, upper GI lesions were diagnosed in 11%, presumed small bowel lesions in 9%, and no lesion in 6% of 80 patients with ongoing hematochezia. [26] This diagnostic accuracy is better than that of arteriography.

Radionuclide Scans The site of GI bleeding can be localized by scanning for extravasation of intravascular radioactively labeled blood. Technetium Tc 99m sulfur colloid scans are obtained shortly after injection. The use of erythrocytes labeled with ^{99m}Tc-pertechnetate allows repeated scanning over 24 to 36 hours after injection to detect intermittent bleeding. These techniques can reveal bleeding when the rate of blood loss is as low as 0.5 mL/min and have no associated morbidity. In comparison with surgical pathology, they have a sensitivity of 88% for correct localization of the bleeding site. [291] The major disadvantage of radionuclide scans is that they localize the bleeding to an area of the abdomen but do not diagnose the specific location or the responsible lesion. For this reason, radionuclide studies are

often used to determine which patients have sufficient ongoing bleeding to warrant angiography, although their value as an angiography screen has been questioned.[292] However, they may allow more selective angiographic studies, decreasing the dye load. In the rare situation in which massive hemorrhage makes endoscopy impossible, angiography should be obtained immediately and not be delayed by prior radionuclide scans.

Angiography In the rapidly bleeding patient, angiography offers accurate diagnosis and therapy. For patients in whom active bleeding cannot be demonstrated, some advocate an aggressive diagnostic angiographic approach based on pharmacological techniques with heparin or streptokinase.[293] When dye extravasation is not demonstrated, angiography can lead to a presumptive diagnosis such as angiodysplasia. Rare small bowel lesions such as arteriovenous malformations or neoplasms may be demonstrated. Selective arterial embolization with polyvinyl alcohol particles or microcoils is beginning to replace intraarterial vasopressin in the treatment of lower GI hemorrhage.[294] Although intraarterial vasopressin is effective in 70% to 90% of cases, bleeding recurrence is high and the elderly may not tolerate the cardiovascular complications of vasopressin.[295] The use of 3F coaxial catheters allows placement of the microcatheter through an outer 5F diagnostic catheter close to the extravasating artery to deliver an embolic agent, thereby lowering the risk for bowel ischemia.[296] Microcoils are becoming the embolic agent of choice because they are easy to inject and provide reliable arterial occlusion, and flow distal to the coil occlusion can be maintained. Patients who fail angiographic therapy require surgery.

Causes and Therapy

The diagnostic certainty regarding the source of acute colonic bleeding is problematic in much of the literature on lower GI bleeding.[297] A definitive diagnosis is made when endoscopic or angiographic evidence of active bleeding is found, or endoscopic evidence of stigmata of recent bleeding. A presumptive diagnosis is made when a tagged red blood cell scan is positive and colonoscopy shows a potential bleeding site in the area of the positive scan. Historically, the two major causes of acute lower GI bleeding were thought to be diverticulosis and angiodysplasia. However, more recent studies utilizing colonoscopy for diagnosis report angiodysplasia much less often (Table 33-6).[3],[288] Diverticulosis remains the most frequent cause of gross lower GI bleeding, although diverticula are also found incidentally in up to 66% of patients with other bleeding sources. In contrast to patients with upper GI bleeding, patients with lower GI bleeding are less likely to present with shock and have lower transfusion requirements. Like upper GI bleeding, lower GI bleeding stops spontaneously in approximately 80% of cases.

DIAGNOSIS	PERCENTAGE OF TOTAL DIAGNOSES (%)	MEAN (%)
Diverticulosis	20–55	33
Angiodysplasia	3–37	8
Cancer/polyp	8–30	19
Colitis*	6–22	18
Anorectal	0–9	4
Others†	3–14	8
Unknown	1–25	16

* Includes inflammatory bowel disease, infectious colitis, ischemic colitis, radiation colitis, vasculitis, and inflammation of unknown etiology.

† Includes postpolypectomy bleeding, aortoenteric fistula, trauma from fecal impaction, and anastomotic bleeding.

Adapted from ref. 297.

TABLE 33-6 Final Diagnosis of Major Lower Gastrointestinal Bleeding from Seven Studies

Diverticular Bleeding Diverticular bleeding occurs in only 3% of patients with diverticulosis. However, it is the most common cause of major lower GI hemorrhage because of the high prevalence of diverticulosis in the Western world (see Chapter 87). Despite the left-sided preponderance of diverticula, angiographic studies have demonstrated that 70% of bleeding diverticula occur in the right side of the colon.[298] In contrast, studies in which colonoscopy was used in a limited number of patients suggest that bleeding diverticula are more likely to be left-sided.[299],[300] Although most patients with diverticular hemorrhage have always been managed nonoperatively,[301] colonoscopic treatment of bleeding further decreases the need for surgical intervention.[304] Diverticula are usually located in the colonic wall at the sites of penetration of nutrient vessels. Bleeding presumably results from a colonic artery that penetrates into the dome of the diverticulum. The artery ruptures into the diverticular sac and causes copious bleeding. Clinical evidence of associated diverticulitis or inflammation is usually not present, and vessel rupture is thought to be the result of pressure erosion. Diverticular bleeding presents with acute, painless, maroon to bright red hematochezia, although melena may occur. Diverticulosis is not thought to be a cause of Hemoccult-positive stool or slow bleeding.[302] If the initial bout of diverticular bleeding ceases spontaneously, no further therapy is indicated because bleeding does not recur in most patients. Of the 75% to 80% of patients in whom bleeding ceases, 65% to 75% will not have a recurrence, and 25% to 35% will have repeated episodes of diverticular hemorrhage.[3],[303] Urgent colonoscopy after purge with endoscopic treatment of diverticular bleeding for patients with active bleeding or stigmata of bleeding has been shown to be highly effective in decreasing the need for surgical intervention compared to historical controls.[304] Endoscopic treatment has not been compared to angiographic intervention, which is reported to be effective in 93% of patients in whom embolization was possible and in 76% on an intention-to-treat basis.[305] Any patient in whom endoscopic or angiographic control of diverticular bleeding fails should undergo urgent surgery to remove the portion of the colon bearing the bleeding site. Additionally, patients with recurrent diverticular bleeding should undergo elective surgery if their general medical condition and anticipated life span warrant such aggressive therapy. Accurate preoperative localization of the bleeding site with either angiography or colonoscopy reduces postoperative rebleeding rates by directing the resection to the appropriate segment of colon. Subtotal colon resection is recommended for patients with recurrent bleeding but no demonstration of a bleeding site and may be associated with lower rebleeding rates than limited colon resection without increasing morbidity or mortality.[306] Surgical mortality rates for recent series are between 5% and 10%.[307]

Angiodysplasia Vascular ectasia, or angiodysplasia, is a common cause of acute major lower GI hemorrhage and slow intermittent blood loss (see Chapter 130). Of 80 patients with lower GI angiodysplasia, 46% presented with acute hemorrhage and 54% presented with chronic or occult blood loss.[308] The percentage of cases of acute lower GI bleeding that have been attributed to angiodysplasia varies widely in the literature, from 10% to 40%.[309],[310] Most vascular ectases are degenerative lesions associated with aging. Two thirds of patients with colonic angiodysplasia are older than 70 years of age. These lesions are different from the congenital vascular lesions that occur throughout the GI tract in various age groups. Angiodysplastic lesions are usually multiple, less than 5 mm in diameter, and involve primarily the cecum and right side of the colon. A clinical association with aortic value stenosis is recognized,[311] although the validity of the association continues to be controversial.[312] It has been reported that aortic valve replacement decreases bleeding frequency.[313] Acquired von Willebrand disease secondary to aortic stenosis, which is corrected after valve replacement, has been the hypothesized link between bleeding angiodysplasia and aortic valve disease.[314] The pathogenesis of angiodysplasias is unknown, but one theory is that repeated, partial, intermittent obstruction of the submucosal veins where they pierce the muscle layers of the colon leads to dilation and tortuosity of the vessels.[315] Eventually, the entire arteriolar-capillary-venular unit dilates, creating a small arteriovenous communication. The predilection for the right side of the colon of these degenerative lesions may reflect the greater tension in the cecal wall than in the rest of the colon. The diagnosis of vascular ectasia can be made by colonoscopy or angiography. A sensitivity of 70% for helical computed tomographic angiography has been reported in the diagnosis of colonic angiodysplasia by the demonstration of vessel accumulation in the colon wall, an early-filling vein, and an enlarged supplying artery.[316] The diagnostic sensitivity of colonoscopy is 80% to 90%, and colonoscopy has the advantage of therapeutic potential.[26] Naloxone may enhance the appearance of both normal colonic vasculature and ectasias.[317] The sensitivity of angiography for angiodysplasia appears to be much lower, although no data based on pathology as the gold standard are available. The earliest angiographic sign is a densely opacified, dilated, tortuous, slowly emptying intramural vein. A vascular tuft represents a more advanced lesion, and an early-filling vein reflects an arteriovenous communication and is a late sign.[318] All diagnostic modalities frequently identify the lesions without demonstrating active bleeding. Because active bleeding is infrequently identified and because these lesions appear to be common in the elderly without a history of significant blood loss, a definitive diagnosis is difficult. Nevertheless, if no other source of GI bleeding is identified in a patient with recurrent or persistent GI bleeding sufficient to require transfusions or cause significant anemia, the presence of angiodysplasia is an indication for treatment. Angiographic embolization and endoscopic techniques of hemostasis are both successful for controlling continued gross hemorrhage.[319] Electrocoagulation techniques, including heater probe, hot biopsy with monopolar coagulation, multipolar coagulation, Nd:YAG laser, and the argon plasma coagulator, have all been used successfully for the treatment of bleeding angiodysplasia. Rebleeding rates of 10% to 30% have been reported.[320] Options then include further endoscopic therapy or surgery. Complications of endoscopic therapy are uncommon but include induction of bleeding and perforation. A retrospective comparison of medical therapy, coagulation techniques, and surgery showed a significant decrease in transfusion requirements for all three types of management of angiodysplasia.[321] Specimen injection techniques have demonstrated incidental angiodysplastic lesions in as many as 50% of surgically resected colons from autopsy specimens.[322] It seems prudent to reserve endoscopic therapy for lesions causing significant blood loss or anemia. For chronic or recurrent bleeding caused by angiodysplasia, estrogen-progesterone therapy is successful in decreasing transfusion requirements. If therapeutic colonoscopy and estrogen-progesterone therapy fail to prevent recurrent bleeding, a hemicolectomy or colectomy is indicated, depending on the localization of these lesions.

Neoplasms and Postpolypectomy Bleeding Benign and malignant neoplasms of the colon are common and, like diverticula and angiodysplastic lesions, occur predominantly in the elderly. They usually present with small degrees of intermittent bleeding or Hemoccult-positive stools. However, neoplastic lesions are the cause of acute lower GI bleeding in 2% to 26% of cases.[287] The diagnosis is made by colonoscopy, and the treatment is surgical or colonoscopic excision. Small bowel tumors are rare but may be diagnosed by small bowel x-ray films or enteroclysis. Occasionally, angiography may be required to make the diagnosis. A history of

intermittent small bowel obstruction is a clue to a small bowel tumor as the cause of lower GI bleeding. Postpolypectomy bleeding accounts for 2% to 5% of cases of acute lower GI bleeding. [3], [288] A review of patients with postpolypectomy hemorrhage noted that half of the patients required transfusions; the presentation time from polypectomy ranged from 0 to 17 days, with a median of 5 days. [323] Most patients had been on NSAIDs/aspirin or anticoagulants. Endoscopic therapy successfully treats more than 95% of patients. [324]

Perianal Disease Hemorrhoids and anal fissures are the most common causes of minor intermittent lower GI bleeding. Most young and middle-aged persons with rectal bleeding do not even seek medical care. [325] Only rarely is the amount of bleeding severe enough to cause iron deficiency anemia or acute and severe enough to mandate transfusions. Massive hemorrhage from simple hemorrhoids is rare. Bleeding is usually from internal hemorrhoids and is painless. The characteristic clinical history is the presence of bright red blood on the toilet tissue or around the stool but not mixed in the stool. Bleeding often occurs with straining or the passage of hard stool. A similar history is common in patients with bleeding from anal fissures, with the exception that anal fissures are often painful. Because rectal polyps and carcinomas may present with a similar bleeding history, patients should be evaluated with flexible sigmoidoscopy, including retroflexion of the flexible sigmoidoscope in the rectum to examine the proximal anal canal. Careful external examination of the external anal canal is also necessary. Perianal disease is treated with sitz baths, bulk-forming agents, avoidance of straining, and ointments or suppositories. It is unknown whether actual therapeutic benefit is obtained with locally applied medications containing lubricants and hydrocortisone, but many patients report symptomatic relief. When bleeding or other symptoms continue to be troublesome, hemorrhoidal banding, coagulation techniques, or surgery may be indicated. [326]

Meckel Diverticulum Meckel diverticulum is the most frequent congenital anomaly of the intestinal tract, with an incidence of 0.3% to 3.0% in autopsy reports. It develops from incomplete obliteration of the vitelline duct, leaving an ileal diverticulum. Approximately 50% of these diverticula contain normal ileal mucosa, and most of the remaining 50% contain gastric mucosa or may occasionally contain duodenal, colonic, or pancreatic ectopic mucosa. The gastric mucosa is capable of acid secretion, which can result in ulceration of adjacent ileal mucosa. Most Meckel diverticula remain asymptomatic and do not require surgical excision when discovered incidentally. [327] Bleeding, the most common complication, usually occurs in childhood, although bleeding may rarely occur in adults. [328] Patients present with painless bleeding that may be dark or bright red, although its appearance is classically described as “currant jelly.” The diagnosis can be made by scanning with radioactively labeled technetium, but false-negative results are not uncommon, and false-positive results have also been reported. [329] It has been suggested that the administration of pentagastrin or cimetidine before the scan may improve sensitivity of the test. [330], [331] Barium filling of the diverticulum may occur, especially with an enteroclysis. Mesenteric angiography may demonstrate the site of bleeding. Surgical excision is the treatment of choice.

Inflammatory Bowel Disease Bleeding from inflammatory bowel disease is usually minimal to moderate, although it reportedly accounts for 2% to 6% of all cases of acute lower GI bleeding. [3], [297] A review of acute major GI hemorrhage in inflammatory bowel disease suggests that it is much more common in Crohn’s disease than in ulcerative colitis. [332] However, acute lower GI bleeding accounts for only 1% of hospital admissions for Crohn’s disease. Surgery is required for treatment in 20% to 35% of cases. [333]

Colitis Secondary to Ischemia, Infections, or Irradiation *Ischemic colitis* is a common entity in the elderly (see Chapter 131). It is usually caused by “low-flow states” and small vessel disease rather than by large vessel occlusion. Any segment of the colon may be involved, although the most common areas are the splenic flexure, descending colon, and sigmoid colon. The typical presentation is mild, cramping abdominal pain localized to the lower left side, followed within 24 hours by rectal bleeding or bloody diarrhea. The blood loss is characteristically minimal, although ischemic colitis causes 3% to 9% of all cases of major lower GI bleeding. [3], [297] Plain abdominal films may show the classical “thumb-printing” lesion of the colon. The diagnosis is best made by colonoscopy and biopsy. Most cases resolve spontaneously with observation and medical support. Surgery is reserved for the rare circumstance of clinical deterioration with fever and a rising leukocyte count or persistent hemorrhage. *Infectious colitis* caused by *Campylobacter jejuni, Salmonella* species, *Shigella* species, invasive *Escherichia coli* or *E coli* 0157:H7, or *Clostridium difficile* often presents with bloody diarrhea. The degree of blood loss is rarely significant. The diagnosis is made by sigmoidoscopy or colonoscopy with biopsy and stool culture. Treatment is not required or is determined by the specific pathogen. *Radiation-induced colitis* is a chronic or recurrent problem that may follow irradiation immediately or several years later (see Chapter 132). The blood loss is rarely massive but may cause iron deficiency or a need for intermittent blood transfusions. The diagnosis is based on a history of prior irradiation with endoscopic biopsy confirmation. Medical treatment with bulk-forming agents or sulfasalazine has not been successful. Intrarectal instillation of formalin will stop bleeding in 75% to 80% of patients. [334] Endoscopic coagulation treatment with the Nd:YAG, argon, or KTP (potassium [K] titanyl phosphate) laser, bipolar electrocoagulation, and treatment with the heater probe have also been successful in 65% to 90% of patients. [335], [336], [337] and [338] These endoscopic treatment modalities appear safer and technically easier than formalin instillation. Decreased transfusion requirements have also been reported after treatment with estrogen-progesterone and hyperbaric oxygen. [339], [340] Surgical intervention is difficult because of the radiation damage to local tissue, with substantial morbidity reported. [341]

Intussusception Intussusception may present with maroon stools and is almost always accompanied by cramping abdominal pain. Uncommon in adults, it usually has a leading point, such as a polyp or malignancy. The diagnosis may be suggested by plain abdominal films and the finding of a sausage-shaped mass during physical examination. Barium enema studies may be useful for diagnosis and in children may be used for therapeutic reduction. The treatment of intussusception in adults is usually surgical.

Portal Hypertension and Rectal Varices Ileal or colonic varices, which tend to occur around ostomies, [342] may present with massive lower GI bleeding. [343] The diagnosis is often made by angiography, and the treatment is decompression of the portal hypertension by surgery, TIPS, or propranolol and nitrates. Rectal varices may also present with gross lower GI bleeding. Variceal ligation has been used to stop bleeding, although decompression of portal hypertension is probably superior for preventing rebleeding. Multiple colonic vascular ectases in patients with portal hypertension can cause hematochezia or Hemoccult-positive stools; the condition is called *portal colopathy*. [344]

Other Causes Several rare causes of lower GI bleeding deserve brief mention. Solitary rectal ulcer, which may be associated with an internally prolapsing rectal mucosa, causes lower GI bleeding, although the bleeding is rarely massive. [345] Aortoenteric fistulae, not associated with prosthetic grafts, have been described in the ileum and colon. The diagnosis is usually made by angiography, and the treatment is surgery. NSAIDs predispose patients to lower GI bleeding by two mechanisms: causing small bowel or colon ulcers [346], [347] and interfering with platelet function. [348], [349] Metronidazole may be effective in decreasing GI blood loss from NSAID-induced enteropathy. [350]

BLEEDING FROM AN UNKNOWN SOURCE

There are always a few unfortunate patients with chronic bleeding (obscure-occult) or recurrent acute bleeding (obscure-overt) in whom a diagnosis cannot be made despite upper and lower GI x-ray studies, endoscopy, and angiography. It has been estimated that in as many as 5% of patients, a source of bleeding cannot be identified despite extensive examination. [351] The cause of GI bleeding from an obscure origin in most patients is thought to be vascular ectasia. [352] Unfortunately, many of these lesions are too small to be detected by angiography and can be missed or not reached by endoscopy. [353] They usually are degenerative lesions that develop with aging. Rarely, arteriovenous malformations or vascular fragility syndromes may be associated with elastic tissue disorders, such as pseudoxanthoma elasticum and Ehlers-Danlos syndrome, or with skin-associated vascular anomalies, such as heredity hemorrhagic telangiectasia or blue rubber bleb syndrome (see Chapter 49 and Chapter 130). [354], [355] and [356]

The failure to make a diagnosis in some patients with chronic or recurrent bleeding reflects the relative inaccessibility of the small bowel. In young patients, a radionuclide scan for Meckel diverticulum is valuable. The most useful approach for identifying small bowel sources of bleeding is enteroscopy, which may replace a “second look” upper GI endoscopy. [357] Small bowel enteroscopy by the “pull” or Sonde technique has been successful in diagnosing the cause of bleeding from obscure origins in 25% to 40% of patients whose condition has eluded all other diagnostic tests. [358], [359] This is a tedious and time-consuming test for both patient and physician; in three fourths of the patients, the ileum is reached after an average intubation time of 6 hours. Other drawbacks include the lack of intervention capability, inability to view the lumen completely (estimated at 50%–70%), and lack of availability of the technique. Much more commonly applied is “push” enteroscopy with a pediatric colonoscope or a modified, longer upper GI endoscope (see Chapter 139). This has the advantage of allowing therapeutic maneuvers and is widely available. [360], [361] Diagnostic yields of 30% to 45% are reported. [362], [363] Up to half of the diagnoses made at push enteroscopy are within the reach of a standard upper GI endoscope, demonstrating the value of repeated expert upper GI endoscopy. [364] Push enteroscopy and Sonde enteroscopy have been combined to increase the diagnostic yield to 58%. [365] Capsule endoscopy may become important in visualizing the portion of the small bowel that is not within reach of the push enteroscope.

Enteroclysis, or a small bowel infusion x-ray study, provides better radiographic images of the small bowel than traditional small bowel follow-through. Unfortunately, it is an uncomfortable examination requiring intubation of the patient through the mouth, with fluoroscopic positioning of the tip of the tube at the duodenojejunal junction. Barium is then rapidly injected to fill the small bowel. Methylcellulose and water with or without parenteral glucagon are also given to achieve a double-contrast effect. [366], [367] A combination of push enteroscopy and enteroclysis, in which the enteroclysis tube is inserted on withdrawal of the enteroscope, provides a better diagnostic yield in a more comfortable and convenient single diagnostic setting. [368] Diagnostic angiography in the patient who is not actively bleeding may also reveal vascular anomalies or small bowel tumors not identified by other tests. [369]

Intraoperative small bowel enteroscopy has a high success rate in identifying bleeding lesions, although the operative morbidity may be significant, [370] and after seemingly appropriate surgery, rebleeding occurs in 26% of patients overall and in 39% of patients with small bowel vascular abnormalities. [371], [372] Intraoperative

angiography with methylene blue injection and intraoperative scintigraphy have also been used in locating small bowel angiodysplastic lesions. [373], [374]

Push enteroscopic cauterization of small intestinal angiodysplasias has been shown to be effective in decreasing blood transfusion requirements. [375], [376] If this is unsuccessful or not feasible because multiple lesions are present, surgical resection of the involved segments may be necessary. A trial of estrogen-progesterone therapy for the possible underlying diagnosis of vascular ectasia may be worthwhile, even in some patients without a definite diagnosis. Unfortunately, the condition of a few patients with bleeding from an unknown source defies diagnostic efforts, or they patients are too ill for surgery and are relegated to receive transfusions as needed.

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CHAPTER 34

David A. Ahlquist

APPROACH TO THE PATIENT WITH OCCULT GASTROINTESTINAL BLEEDING

By definition, occult gastrointestinal (GI) bleeding is hidden and not apparent on stool inspection. It occurs commonly, but a test is required for its detection. Elevated fecal blood levels are found in about 1 of 20 adults on prevalence screens [1], [2], [3] and [4] and probably occur episodically in all persons. Although often insignificant, occult GI bleeding may herald a health-threatening lesion arising at any level from mouth to rectum. As such, the clinician is challenged when faced with occult GI bleeding.

The critical metabolic sequela of occult GI bleeding is iron deficiency. Iron deficiency afflicts 20 million people in the United States alone, and its global prevalence is estimated at 15%. [5], [6] It is the most common cause of anemia and often results from occult GI blood loss. [7] The management of iron deficiency depends on the cause, and a GI origin should always be considered.

Fecal occult blood testing is widely used to screen for colorectal cancer and to evaluate iron deficiency and anemia. A rational assessment and treatment of occult GI bleeding are based on an appreciation of the pathophysiology of occult blood loss, available diagnostic tools, and therapeutic principles.

QUANTIFYING BLOOD LOSS

Given a typical daily stool mass of 150 g (150 mL) and a circulating hemoglobin (Hb) of 15 g/dL, the following equivalences can be calculated: 2 mL of fecal blood loss per day = concentration of 2 mg of Hb per gram of stool = total daily Hb loss of 300 mg = daily iron loss of 1 mg.

Depending on the rate and site of GI bleeding, enterocolic transit time, efficiency of lumenal Hb metabolism, and degree of fecal mixing, blood loss may be visually gross or occult. At one extreme, large amounts of blood may be lost into the proximal GI lumen and remain occult. A bolus of more than 150 mL of blood in the stomach or cecum is required to consistently produce melena or hematochezia. [8], [9] Fecal Hb concentrations may approach those of circulating blood without being visibly apparent. [10] On the other extreme, a mere drop of blood from anorectal lesions may be visible as a bright red streak on the stool surface.

Fecal blood is not simply present or absent; the levels range in a continuum from normal to pathologically elevated. Fecal blood loss quantified in healthy volunteers averages 0.5 to 1.5 mL/d (0.5–1.5 mg of Hb per gram of stool), [11], [12] and [13] with levels below 2 mL/d found in 95% (Fig. 34-1).

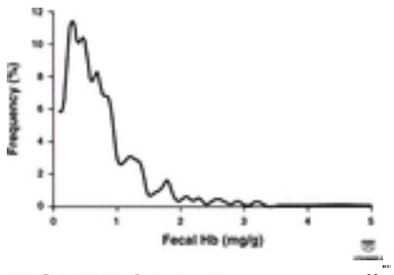

FIGURE 34-1. Frequency distribution of fecal occult blood levels in 900 asymptomatic subjects undergoing colorectal cancer screening as quantified by the HemoQuant assay. In more than 95%, the fecal hemoglobin levels are below 2 mg/g, which is considered to be the upper limit of normal.

GASTROINTESTINAL HEMOGLOBIN METABOLISM

The degree of Hb disassembly during GI transit and fecal storage affects measurements because available fecal blood tests target different components of the Hb molecule (Table 34-1). An appreciation of enterocolic Hb metabolism helps to clarify test limitations and is useful in selecting a test appropriate to the clinical indication.

TABLE 34-1 Effect of Gastrointestinal Bleeding Site on Metabolic Fate of Intralumenal Hemoglobin

Heme is cleaved from Hb by enzymes in the upper gut. [14] Some heme, probably less than 15%, is absorbed intact in the proximal small intestine. [15], [16], [17], [18] and [19] Heme absorption may increase threefold in iron deficiency [20] and decrease after gastrectomy. [21] In the colon, from 1% to 99% of heme is converted by bacteria to porphyrins. [16], [22] Heme-derived porphyrins escape detection by guaiac tests. [10], [13], [18]

The globin chains of Hb are digested by upper gut proteases. Because immunoassays for Hb are directed against globin antigens, they are insensitive for upper GI bleeding. [23], [24] and [25] Immunoassays fail to detect ingested blood in quantities up to 100 mL. [24], [26] Hb globins are also metabolized by colonic bacteria, and immunoassays are less likely to detect bleeding from the right side of the colon than from the left. [27] Furthermore, immunoreactivity is progressively lost during fecal storage.

IRON METABOLISM AND DEFICIENCY

Occult GI bleeding may be compensated for by increased iron absorption or mobilization of body iron stores, which in turn stimulates erythropoiesis. However, if iron loss from occult bleeding chronically exceeds intestinal iron absorption, iron stores become depleted, and iron deficiency with attendant anemia and other metabolic sequelae ensues. The time required for iron deficiency to develop depends on the rate and chronicity of occult bleeding.

Iron Absorption

Ordinarily, 1 to 2 mg of iron is absorbed daily, which represents about 10% of that in a typical Western diet. [28] Both heme and elemental forms of iron are absorbed primarily in the duodenum and proximal jejunum. Elemental iron absorption may be increased up to tenfold by iron deficiency, anemia, hypoxia, liver disease, increased erythropoiesis, and an elevated lumenal iron concentration. [5], [28], [29] It is facilitated by reducing substances, bile, pancreatic juice, gastric acid, and a nonacidic gastric modulator. [5], [30], [31], [32] and [33] Iron absorption is impeded by various mucosal diseases of the upper small intestine and by certain lumenal substances, such as phytates in cereals, tannic acid in tea, and oxalates, that chelate iron. [28], [29], [34], [35] and [36] Selective iron malabsorption occurs rarely secondary to an inherited defect of transferrin function [37] or to an acquired autoimmunity to the transferrin receptor. [38]

Iron Loss

A physiological mechanism does not exist for iron excretion. Under normal circumstances, absorption of dietary iron (average, 1–2 mg/d) equals or just exceeds iron loss through normal daily occult GI bleeding (average, 1 mg/d). Trace amounts of iron are also lost through cutaneous desquamation and intestinal epithelial sloughing. Menorrhagia often contributes to iron deficiency in premenopausal women. [39] Although excessive bleeding can occur from the airway, from the urinary tract, or from multiple phlebotomies, these are uncommon causes of iron deficiency, and, if present, clinically apparent. [5] Excessive iron loss through occult GI bleeding, the most common cause of iron deficiency, is the most difficult to recognize. As a general rule, daily GI blood losses of at least 5 to 10 mL are required to overcome compensatory absorptive increases and lead to a negative iron balance.

Iron Storage

Most body iron, about 75%, is stored in metabolically active forms, with roughly 70% in Hb and 5% in myoglobin and various tissue enzymes. [5] The remaining 25% of body iron is metabolically inactive and stored in a soluble complex with ferritin or in a particulate state as hemosiderin, especially in hepatocytes and in the reticuloendothelial cells of the liver, bone marrow, and spleen. [5] A small amount of ferritin is present in plasma and correlates well with total body iron stores. [40]

Metabolic Consequences of Iron Deficiency

Iron is present in all human cells and is of vital importance for oxygen transport and many metabolic functions. This ubiquitous metal is essential for nearly half of the enzymes of the Krebs and tricarboxylic acid cycles, for DNA synthesis, and for many other cellular processes. [5], [41] It is not surprising that the clinical and metabolic manifestations of iron deficiency are so generalized. The fatigue associated with iron deficiency should be recognized as a pancellular phenomenon and results not only from anemia [42], [43] but also from dysfunction in nonhematologic tissue. [44], [45]

Characteristic mucocutaneous findings may be present (see section “ Clinical Manifestations of Blood Loss”). Gastric atrophy with achlorhydria and a spruelike small intestinal lesion with malabsorption may occur with iron deficiency and resolve with iron replacement. [46], [47], [48] and [49] Iron deficiency has also been causally linked to certain behavioral, immunologic, and developmental abnormalities. [50], [51], [52], [53] and [54]

ETIOLOGY OF BLOOD LOSS

Many of the lesions that cause gross bleeding (see Chapter 33) can also cause occult bleeding. The mechanisms of occult GI bleeding necessarily involve a disruption of the epithelium and subepithelial blood vessels and can be categorized as inflammatory, infections, vascular, neoplastic, and other (Table 34-2). The prevalence of responsible lesions varies by age and geography. For example, occult bleeding in infants commonly arises from milk-induced enteritis, [52] but in the elderly from peptic ulcer disease or neoplasms. [7], [13], [55], [56] Although rare in temperate climes, hookworm infestation is the most common cause of harmful occult bleeding in tropical and subtropical regions. [57]

Inflammatory Causes	**Tumors and Neoplastic Causes**
Acid peptic disease	Primary GI cancer at any site
Large hiatal hernia (Cameron erosions)	Metastases to GI tract
Crohn’s disease	Large polyp at any site
Chronic ulcerative colitis	Lymphoma
Mild enterocolitis	Leiomyoma
Whipple disease	Leiomyosarcoma
Sprue	Lipoma
Eosinophilic gastroenteritis	Other
Meckel diverticulum	**Drugs**
Solitary colon ulcer	Nonsteroidal antiinflammatory drugs
Other	Other
Infectious Causes	**Miscellaneous Causes**
Hookworm	Long-distance running
Strongyloidiasis	Extra-GI bleeding
Ascariasis	Benign infiltrative lesions
Tuberculous enterocolitis	Factitious cancer
Amebiasis	Artifact
Other	
Vascular Causes	
Angiodysplasia and vascular ectasia	
Gastroesophageal varices and congestive gastropathy	
Hemangiomata	
Blue rubber bleb nevus syndrome	
Watermelon stomach	
Other	

GI, gastrointestinal.

TABLE 34-2 Disorders that May Present as Occult Gastrointestinal Bleeding with or without Iron Deficiency

Inflammatory Causes

In Western countries, erosions or ulcerations of the esophagus, stomach, and duodenum are the most common GI lesions associated with occult bleeding and iron deficiency (see Chapter 60 and Chapter 66). Iron deficiency has been attributed to such acid peptic disease in 30% to 70% of adult cases and is often asymptomatic. [7], [24], [25], [55], [56] Acid peptic disease, particularly reflux esophagitis, [58] may also underlie iron deficiency in children. Most peptic disease is associated with drug use (see below) or *Helicobacter pylori* gastritis (see Chapter 68). In some populations, *H pylori* disease is pandemic and contributes to widespread iron deficiency. [59], [60] Studies suggest that the iron deficiency resulting from *H pylori* infection may occur in the absence of erosions or ulcers and resolves with antibiotic therapy. [61]

The association between large diaphragmatic hernias and iron deficiency anemia has long been known. [62], [63] and [64] In up to 10% of iron-deficient patients, a large diaphragmatic hernia is found. [7], [55] Blood loss is caused by longitudinal mucosal erosions (Cameron erosions) located in the proximal stomach and is thought to be secondary to mechanical trauma resulting from breathing. [65], [66]

Many inflammatory GI conditions of the small bowel and colon may cause occult bleeding (see Chapter 83 and Chapter 85). Bovine milk may produce occult bleeding and iron deficiency in infants as a consequence of inflammation throughout the gut, which resolves after the ingestion of milk is stopped. [67] Meckel diverticulum may cause occult bleeding in children and young adults. [52] Crohn's disease and chronic ulcerative colitis uncommonly present with occult bleeding alone. [7], [55] Occult blood loss may occur with celiac sprue, Whipple disease, eosinophilic gastroenteritis, radiation enteritis, and solitary colon ulcer.

Infectious Causes

Whereas occult bleeding may accompany many acute infectious enterocolitides (see Chapter 74), it may be chronic and lead to iron deficiency in such conditions as GI tuberculosis (see Chapter 75), amebiasis (see Chapter 125), ascariasis (see Chapter 126), and *H pylori* gastritis (see above). However, the most common infectious cause of harmful occult bleeding worldwide is hookworm (see Chapter 126).

Several hundred million people are infected with hookworm globally, and the prevalence in some tropical countries exceeds 80%. [57] The major and often the only manifestation is iron deficiency, which exacts an enormous socioeconomic toll in lost work productivity. Daily occult fecal blood loss averages more than 12 mL in most hosts, may exceed 100 mL, correlates with the hookworm burden, and drops precipitously after use of a vermifuge. [57], [68]

Vascular Causes

Varices are occasionally incriminated as the source of occult bleeding and have been reported in up to 3% of patients with iron deficiency. [7], [13] The attendant congested mucosa in the gastroduodenum [69] or colon [70] may account for occult blood loss with portal hypertension.

Vascular malformations (see Chapter 130) are found in up to 6% of adults with iron deficiency anemia [7], [24], [25], [55], [56] and are a common explanation for occult bleeding of obscure origin. [71], [72] and [73] GI bleeding remains occult in many patients with acquired or hereditary (e.g., hemorrhagic telangiectasia [Osler-Weber-Rendu syndrome]) vascular malformations. [74] An increasingly recognized and endoscopically treatable vascular lesion is watermelon stomach, which characteristically presents with iron deficiency anemia in older women. [75], [76] Clinically significant occult bleeding has been described from other vascular lesions, including post-radiation telangiectases [77] and lesions associated with the blue rubber bleb nevus syndrome, [78] scleroderma, [79] Turner syndrome, [80] and Klippel-Trenaunay syndrome. [81]

Tumors and Neoplasms

GI tumors are second only to peptic disease as a cause of occult bleeding leading to iron deficiency in adults in Western countries. [7], [24], [25], [55], [56] Because of the importance of fecal occult blood testing in the detection of colorectal adenocarcinoma and adenomas, these lesions are discussed separately (see section "Occult Bleeding Patterns" under "Fecal Blood Screening for Colorectal Neoplasia"). Colorectal cancer is the most common malignant lesion to cause occult bleeding in Western countries, followed by primary cancers in the stomach, esophagus, and ampulla. [7], [13], [24], [25] /SUP>, [55], [56], [82] Lymphomas, lipomas, leiomyomas, leiomyosarcomas, hamartomas, juvenile polyps, and metastatic lesions to the gut can also produce occult bleeding (see Chapter 80, Chapter 89, and Chapter 91).

Drugs

Any ingestant that directly or indirectly injures the GI tract may cause abnormal occult bleeding. Ethanol may lead to hemorrhagic gastritis only at high concentrations, [83], [84] and the contribution of social drinking to occult bleeding is probably minor. Anticoagulants appear to unmask bleeding from preexisting lesions rather than produce bleeding per se. [85], [86] Certain antibiotics, potassium preparations, antimetabolites, and other drugs may damage the epithelium, but few data on occult bleeding are available.

Aspirin and related nonsteroidal antiinflammatory drugs (NSAIDs) are responsible for most cases of drug-induced occult GI bleeding, owing to widespread use. In the United States, more than 30 billion NSAID tablets are consumed annually. [87] Except for acetaminophen and sodium salicylate, all NSAIDs may cause blood loss. [88], [89] At therapeutic doses, some NSAIDs induce more bleeding than others. [89] The new cyclooxygenase-2 inhibitors are much less toxic to the mucosa than conventional NSAIDs. [90], [91] NSAIDs most commonly ulcerate the stomach but may injure the GI tract at any level and cause bleeding. [89], [92] NSAID gastroenteropathy results from the inhibition of epithelial cyclooxygenase and loss of mucosa-protective prostaglandins. [93] The NSAID dose influences the amount of mucosal injury and blood lost. [85], [87] Those taking therapeutic amounts of aspirin lose on average 2 to 5 mL of blood daily, and some more than 30 mL/d. [85], [87], [94] Fecal blood levels are rarely elevated in those taking low-dose aspirin for cardiovascular prophylaxis. [86], [95]

Miscellaneous Causes

Iron deficiency may develop in long-distance runners and compromise performance. [96] Occult GI bleeding appears to be a major cause. [97], [98] In elite class runners, both gastric and colonic erosions have been identified and have been shown to resolve when running is stopped. [99], [100] Occult GI bleeding is less likely to occur with other endurance activities. [101], [102]

Fecal occult blood levels may be elevated in patients who swallow blood from tracheobronchial, dental, oronasopharyngeal, or factitious sources. Infiltration by benign conditions such as endometriosis, [103] amyloidosis, [104] and splenosis [105] may cause hemorrhage and chronic occult bleeding. Stools sampled from toilet water may be contaminated by blood in the urine or menstrual blood. [106] Finally, numerous substances other than blood may react with guaiac and other tests to produce false-positive results (see section "Fecal Blood Testing").

CLINICAL MANIFESTATIONS OF BLOOD LOSS

Symptoms

Occult GI bleeding in most cases is clinically silent and unsuspected. However, predominant manifestations of the underlying GI disease may be responsible for occult bleeding. Also, characteristic symptoms and signs of iron deficiency may occur secondary to chronic occult blood loss from any cause. Fatigue with exercise intolerance is the most frequent symptom of iron deficiency and can be disabling. [41], [42], [43], [44] and [45] Rapid palpitations and exertional dyspnea are also common features of advanced iron deficiency. Less common overt sequelae of iron deficiency may be present; for example, iron deficiency may contribute to a restless leg syndrome. [107]

Pica, or compulsive eating, may occur in roughly 50% of patients with iron deficiency. [108], [109] Pagophagia, or ice eating, is the most frequent form, and some afflicted persons have consumed more than 9 kg of ice daily. Other variants of pica include ingestion of soil (geophagia), laundry starch, brittle or crunchy foods ("gooberophagia"), and chalk. Such bizarre behavior resolves with iron repletion.

Physical Findings

Telltale findings may suggest both the presence and cause of iron deficiency. Papilledema, hearing loss, cranial nerve palsies, and retinal hemorrhages occur rarely with severe iron deficiency and

may resolve with iron repletion. [5], [110] More characteristic of iron deficiency are various epithelial abnormalities. Fingernails and toenails may become brittle, longitudinally furrowed, or spooned, [111] changes called *koilonychia*. Glossitis may occur with erythema and loss of the lingual papillae. [112] Scaling or fissuring of the lips, called *cheilitis,* and atrophic rhinitis may also result from iron deficiency. The association of esophageal webs with iron deficiency is known as *Paterson-Kelly syndrome* or *Plummer-Vinson syndrome*. [113], [114] These proximal esophageal webs may cause dysphagia, are more common in women, and may resolve with iron therapy. [5], [115]

ASSESSMENT AND DIAGNOSTIC STRATEGIES

Occult GI bleeding may be evidenced either indirectly by laboratory confirmation of iron deficiency or directly by measurement of fecal blood. Neither approach is infallible, and they should be viewed as complementary. Iron deficiency would not be present if enteric blood loss were quantitatively minor or of short duration. Negative fecal blood test results may be caused by intermittent bleeding or analytic limitations of the tests. It is important to appreciate the strengths, limitations, and complementarity of the laboratory techniques used to establish occult bleeding. The subsequent GI evaluation should be tailored to the clinical setting.

Blood and Bone Marrow Tests for Iron Deficiency

Although it is a late-stage manifestation, hypochromic microcytic anemia is often the first clue to the presence of iron deficiency. [5] The characteristic erythrocyte morphology on peripheral smear is now more commonly detected by automated calculations that reveal a low mean corpuscular Hb concentration (MCHC) and low mean corpuscular volume (MCV). Anisocytosis, or variability in cell size, is also common in iron deficiency anemia and is reflected by an elevated red cell distribution width (RDW). However, these red cell changes are present in fewer than 70% of persons with iron deficiency anemia, [116] and they may occur in patients with anemia of chronic illness, thalassemia, or sideroblastic anemia. [5], [116], [117] The status of iron stores must be evaluated to determine the cause of microcytic anemia.

Unlike iron deficiency, other causes of hypochromic microcytic anemia are associated with normal or increased tissue iron stores. [5], [116], [117] Although typically decreased in iron deficiency, serum iron levels and transferrin saturation correlate rather poorly with marrow iron stores, are influenced by many medical conditions, and are also commonly low in anemia of chronic illness, the most troublesome differential diagnosis. [5], [116], [117] In contrast, serum ferritin levels correlate well with tissue iron stores and better discriminate iron deficiency anemia from other types of anemia. [5], [117] Serum ferritin levels fall well before anemia develops, and a low serum ferritin level is pathognomonic for iron deficiency. Inflammatory, malignant, liver, and renal disease can elevate serum ferritin and uncommonly cause a falsely normal serum level when marrow iron stores are absent. [5], [116], [117] Under such conditions, erythrocyte ferritin, [118] serum transferrin receptor, [119] or erythrocyte protoporphyrin [120], [121] levels may more accurately reflect body iron stores. Iron deficiency occasionally presents with normal levels of ferritin or other blood markers, and a bone marrow study is indicated if uncertainty remains. Although the gold standard for iron deficiency is the absence of bone marrow hemosiderin by Prussian blue staining, this invasive procedure is seldom required.

Fecal Blood Testing

Four fecal occult blood tests (guaiac, immunochemical, heme-porphyrin, and radiolabeled erythrocyte tests) are commercially available, each with advantages and disadvantages.

Guaiac Tests Guaiac preparations have been employed to detect blood for more than a century [122] and remain the most widely used type of fecal blood test. Guaiac-impregnated pad tests, such as Hemoccult, were developed in the 1960s. [123] These pad tests are elegantly simple, inexpensive, and highly portable, and a sophisticated laboratory is not required for interpretation. Guaiac, a leuco-dye, is a colorless compound that becomes colored in the presence of adequate peroxidase-like substances (e.g., Hb) and hydrogen peroxide. There appears to be no consistent fecal Hb level above which guaiac test results become positive and below which they remain negative. [10], [13], [123], [124], [125] and [126] In general, the rate of fecal blood loss must exceed 10 mL/d before a Hemoccult test result is positive at least half the time. [10], [13], [124], [125] Guaiac test results may be positive in stools with less than 1 mg of Hb per gram [10] or remain negative in those with more than 80 mg/g. [10], [127] Hemoccult sensitivity is enhanced by wetting the fecal smear before adding the peroxide catalyst [128] or by hydrating the stool before smearing. [10] Also, the native water content of stools affects Hemoccult reactivity—wetter stools are more likely to yield positive results than drier stools. [10], [13] Another major explanation of the variability in guaiac test reactivity is degradation by the fecal flora of heme to porphyrin, which does not possess peroxidase-like activity. This accounts for the relative insensitivity of guaiac tests for occult bleeding arising from the right side of the colon and more proximal gut. [13], [16], [22], [24], [129], [130] Conversion of heme to porphyrin continues during fecal storage and causes a corresponding fall in Hemoccult positivity. [10], [131] Thus, guaiac tests tend to underestimate enteric blood loss, especially with bleeding from proximal lesions or with stored specimens. Several factors besides wet stools can produce chemical false-positive reactions. Certain fruits and vegetables have peroxidase-like activity, and diet restriction reduces Hemoccult false positives. [132] Whether oral iron causes guaiac false positives is debated. [133], [134] Other factors reported to cause a color reaction include sucralfate, [135] cimetidine, [136] halogens, [137] and toilet bowl sanitizers. [138] Factors that inhibit guaiac reactivity, in addition to dry stools and heme degradation, include ascorbic acid, [139] antacids, [140] heat, acid pH, and defective reagents. [141] Maneuvers to increase guaiac sensitivity, such as hydrating the smeared fecal aliquot before testing or reconfiguring reagent concentrations, often succeed at the expense of specificity. [128], [142], [143]

Immunochemical Tests Immune detection of fecal blood, studied for more than three decades, is relatively inexpensive and simple to perform. [23], [24], [25], [26] and [27] The antihemoglobin or anti-albumin antibodies used in these assays do not react with nonhuman blood, diet peroxidases, or medications, [144], [145] and burdensome dietary preparations are avoided. The metabolism of antigens during transit or storage compromises the immune detection of fecal blood. These tests detect as little as 0.3 mg of blood added to a stool, but they fail to detect fecal Hb after the ingestion of 20 to 100 mL of blood. [23], [26], [130] It is not surprising that immunochemical tests are less likely to detect upper GI than colorectal bleeding. [23], [24] and [25], [130] Although immunochemical tests promise to improve specificity, reactions to certain fecal constituents may cause false positives at rates comparable to those of guaiac tests. [26], [146], [147] Like guaiac tests, immunochemical tests are qualitative, variably sensitive, and affected by the anatomic site of bleeding and by storage. Immunochemical tests have also been developed to detect blood leukocyte proteins, such as calprotectin [148] and lactoferrin. [149] However, such tests are insensitive for the detection of GI bleeding per se and correlate poorly with hemoglobin-directed fecal blood tests. [150]

Heme-Porphyrin Assay The quantification of fecal blood provides meaningful diagnostic information because the predictive value of an elevated level depends on the degree of elevation. [151], [152] A heme-porphyrin–based assay, [10], [16] HemoQuant (Mayo Medical Laboratories, Rochester, Minnesota; SmithKline BioSciences, Van Nuys, California; Nichols Laboratory, Los Angeles, California), is quantitative, noninvasive, specific for heme, chemically sensitive, and suitable for automation. HemoQuant involves the fluorometric assay of heme and heme-derived porphyrin. Unlike guaiac and immunochemical tests, the HemoQuant test includes that fraction of heme already degraded to porphyrin during fecal storage [13] or enterocolic transit. [10], [13], [16], [22] Because of this feature, its sensitivity for proximal GI bleeding is higher than that of guaiac and immunochemical tests. [13], [24] The heme-porphyrin test may be affected by red meat, but not by other dietary peroxidases, medications, or fecal contaminants. [10], [13], [16], [18] HemoQuant results are calculated as Hb equivalents and reported as milligrams of Hb per gram of stool. Fecal Hb concentrations from 0.01 to 500 mg/g are accurately measured, [10], [11], [12], [13], [14], [15] and [16] and values below 2 mg/g are considered normal. [13], [151], [152] HemoQuant has been validated by the recovery of blood added to stools, [10] recovery of ingested blood, [16], [18], [24], [130] and close correlation with other quantitative assays. [16], [17] HemoQuant is currently performed for commercial use in reference laboratories only, and the inevitable 2- to 4-day delay in results necessitated by specimen shipment is a distinct inconvenience.

Radiolabeled Erythrocyte Technique The fecal recovery of intravenously injected ^{51}Cr-labeled erythrocytes has been used to quantify enteric blood loss for more than four decades. [11], [12] After chromium enters the gut lumen, its reabsorption is negligible, and therefore it has proved to be a valid quantitative marker for GI bleeding. However, some have questioned its accuracy at very low levels of bleeding because of the biliary excretion of free chromium. [153] The test is expensive, requires 3 to 5 days of whole-stool collections, and is impractical for large-volume routine use. Slow enterocolic transit may cause falsely low results. [154]

Specimen Collection and Sampling Accurate fecal blood testing requires control of each step of the testing process. The common practice of sampling stools from the toilet water introduces potential measurement error caused by leaching of blood into the water [138] or by contamination with menstrual or urinary blood, [106] urinary reducing substances, [137], [139] or toilet bowl sanitizers. [138] The use of a collection device prevents these artifacts. [138] Because blood is nonuniformly distributed within the fecal specimen, [155] testing of multiple aliquots or multiple stools reduces sampling error. [151], [156] Guaiac-based tests have been developed to detect blood in the toilet water or on stool wiped from the perineum, but few published data are available. Reports on the testing of stool obtained from digital examination are conflicting. [157], [158] and [159] Some studies suggest that adequate stool is obtained in fewer than half of digital examinations [159] and that the digital approach lowers test specificity. [160]

Test Selection No single fecal blood test is appropriate for all applications. Test selection should be guided by a judicious consideration of test performance characteristics, availability, and cost relative to the clinical indication (Table 34-3). A simple, inexpensive, qualitative test may be preferred to screen for colorectal bleeding. Immunochemical or guaiac tests are appealing for such screening because they preferentially detect colorectal bleeding. The heme-porphyrin assay offers no advantage over these simpler tests in screening for colorectal lesions. [143], [161] However, to evaluate iron deficiency or anemia, the heme-porphyrin assay is most useful because it accurately quantifies lumenal blood loss regardless of the site of bleeding. [13], [24], [130] The radiolabeled erythrocyte technique yields similar quantitative information, [17] but it is time-consuming, much more expensive, and logistically burdensome. Guaiac and immunochemical tests would appear less suitable in the evaluation of anemia because they are qualitative and insensitive for upper GI bleeding. [13], [24], [130], [152] In a study of anemic patients with hemorrhagic lesions in the proximal gut, the heme-porphyrin test detected 88%, guaiac testing 26%, and immunochemical testing of stools 2%. [24]

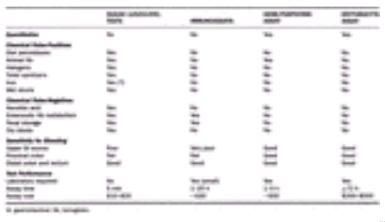

TABLE 34-3 Comparative Features of Fecal Occult Blood Tests

Patient Evaluation

In practice, abnormal occult GI bleeding comes to the attention of clinicians after a positive fecal blood test result has been obtained for a patient undergoing colorectal cancer screening, after iron deficiency anemia has been encountered, and, less commonly, after fecal blood tests have been applied to investigate a patient presenting with GI symptoms. The diagnostic strategy should be tailored according to the setting.

Abnormal Fecal Blood Test Results in an Asymptomatic Patient without Anemia This common situation is encountered almost exclusively as a result of colorectal cancer screening because there are very few other reasons to routinely check fecal occult blood levels (see section " Fecal Blood Screening for Colorectal Neoplasia"). The major responsibility is to exclude a colorectal cancer, although this finding can be expected in only 2% to 10% of cases with positive test results. [1], [2], [3] and [4] Most agree that colonoscopy and other structural approaches are more sensitive and specific for colorectal neoplasia, [161], [162] but these interventions are also invasive and more expensive. Because of the relatively lower prevalence in most Western countries of malignancies proximal to the colon, physicians could justifiably stop their evaluation after colon studies in the absence of symptoms or anemia. [163] However, a good case can be made for additionally checking the serum ferritin level because this is inexpensive, introduces essentially no morbidity, and may point to an indolent proximal GI lesion. If the ferritin level is low, esophagogastroduodenoscopy (EGD) is the preferred next step because the false-negative rate of barium roentgenography of the stomach for peptic and other lesions is so high (see Chapter 138, Chapter 139, Chapter 148, and Chapter 152). If this result is negative, a radiograph of the small intestine is appropriate. If all GI studies are unrevealing, periodic hematologic follow-up is in order. Finally, in populations with a high prevalence of gastric or esophageal cancer, it may be prudent to routinely extend the evaluation to the upper gut if the results of colorectal studies are negative. The common practice of having all patients admitted to the hospital undergo fecal occult blood testing is of uncertain value. Utilization studies indicate that such testing is inappropriate in most cases because of an absence of valid indications, the presence of serious comorbidities, and inadequate patient preparation. [164], [165]

Occult Bleeding in the Anemic Patient Unless menorrhagia, gross hematuria, frequent blood donations, or another source of extraintestinal blood loss is clinically apparent, most patients found to have iron deficiency with or without anemia should undergo an aggressive GI evaluation. Because a GI lesion is found in most men and postmenopausal women with new-onset iron deficiency anemia, [7], [55], [56], [166] it can be argued that fecal blood testing is superfluous and that GI studies should be performed regardless of fecal blood test results. However, nutritional inadequacy, iron malabsorption, or extraintestinal bleeding may be the critical etiologic factor in some patient groups with iron deficiency anemia. Fecal blood testing may be helpful to assess the contribution of enteric blood loss in selected groups with iron deficiency, including children, menstruating women, postgastrectomy patients, immigrants from underdeveloped countries, strict vegetarians, frequent blood donors, and patients with steatorrhea. For example, menstruating women with new-onset iron deficiency often demonstrate occult fecal blood loss and, subsequently, are found to harbor hemorrhagic GI lesions. [167] Thus, it should not be assumed that menstrual blood loss alone accounts for iron deficiency in this large subset of patients. Most adults with new-onset iron deficiency anemia are found to have culprit hemorrhagic lesions in the proximal gut (Table 34-4). [24], [25], [167], [168], [169], [170] and [171] However, roughly 15% of patients with lesions seen on EGD also have potential sources of blood loss on colonoscopy. [25], [167], [168], [169], [170] and [171] Most upper GI lesions in patients with iron deficiency are peptic in nature, whereas most colorectal lesions are neoplastic. [7], [24], [25], [167], [168], [169], [170], [171] and [172]

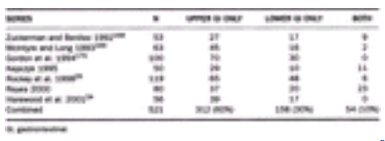

TABLE 34-4 Anatomic Site of Occult Gastrointestinal Bleeding in Patients with Iron Deficiency Anemia

A practical schema for evaluating adults with iron deficiency anemia and occult GI bleeding is shown in Figure 34-2. The large majority of lesions causing occult bleeding in anemic patients can be demonstrated by routine endoscopic procedures. At many centers, extended or push enteroscopy is used only if the results of the initial evaluation by routine EGD, colonoscopy, and small bowel radiography are negative; endoscopically treatable lesions, especially vascular ectasia, are not uncommonly discovered. [71], [163], [173] Some advocate extended upper GI endoscopy rather than conventional EGD for the initial evaluation as a more cost-effective strategy. [62], [72], [174] For immigrants from tropical or subtropical regions, a stool examination for hookworm or other parasite ova should be considered early in the evaluation of anemia.

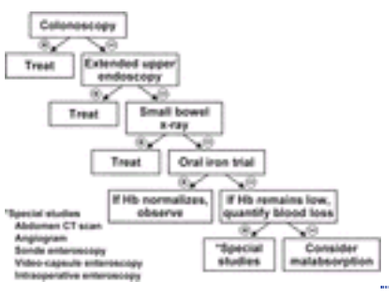

FIGURE 34-2. Diagnostic schema for asymptomatic adult patients with iron deficiency anemia and occult gastrointestinal bleeding. *CT,* computed tomography; *Hb,* hemoglobin.

Because further studies for obscure bleeding sources are more invasive, expensive, and less revealing of lesions, it is often justifiable to observe a response to iron therapy before further studies are undertaken. NSAIDs should be strictly avoided during such a trial. Anemia will resolve and no serious pathology will emerge over time in most such patients. [175] In patients who fail to respond to iron therapy, confirmation of GI blood loss with valid fecal blood testing is desirable before further evaluation. If GI bleeding is established, special diagnostic studies may prove helpful, including universal (Sonde) enteroscopy, [176] angiography (see Chapter 130 and Chapter 159), abdominal computed tomography, and, in rare situations, surgical exploration with intraoperative endoscopy. [73] Laparoscopically assisted panenteroscopy may provide a less invasive future alternative to intraoperative endoscopy. [177] A pill-sized videocapsule device may allow noninvasive interrogation of parts of the small intestine that cannot be reached by conventional endoscopes. [178] Radioisotope blood loss scans have no role in the evaluation of occult bleeding.

Occult Bleeding in the Patient with Gastrointestinal Symptoms Few data are available on the use of fecal blood testing to evaluate patients with GI symptoms in the absence of iron deficiency or anemia, although some suggest that a positive stool result increases the likelihood that organic disease is present. [179] The sequence of diagnostic testing should be initially directed to the anatomic level suggested by the symptoms. Site-specific symptoms in patients with iron deficiency are predictive of abnormalities in the corresponding portion of the bowel. [62], [166] Clinical judgment is required to determine the extent of evaluation in such instances.

FECAL BLOOD SCREENING FOR COLORECTAL NEOPLASIA

Fecal blood screening for colorectal cancer is widely practiced, and U.S. surveys suggest that roughly 30% of adults have undergone such screening. [180], [181] and [182] This section deals only with fecal testing, not with other approaches to the detection of colorectal neoplasia (see Chapter 89, Chapter 91, Chapter 140, Chapter 152, and Chapter 155).

Occult Bleeding Patterns

Fecal blood levels vary widely in colorectal cancer [13, 128, 151, 155, 161, 183] and commonly fall within the normal range (Fig. 34-3). Occult bleeding increases with neoplasm size, surface ulceration, and stage. [13, 161, 184, 185, 186] and [187]

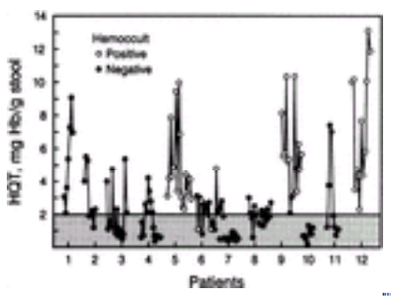

FIGURE 34-3. Occult blood levels in stools collected for 2 weeks from 12 patients with asymptomatic primary colorectal adenocarcinoma as determined by HemoQuant (*HQT*) and Hemoccult. The shaded zone below a HemoQuant level of 2 mg of hemoglobin per gram of stool represents the conventional normal range. (From ref. [151].)

Although it is well established that symptomatic colorectal polyps may bleed, [129] less is known about occult bleeding with asymptomatic polyps. Quantitative studies have shown fecal blood distributions to be comparable in asymptomatic groups with and without adenomas. [126, 161, 183] Roughly 5% of patients with endoscopically proven polyps are guaiac positive, [161, 186] a positivity rate similar to that of the general population. Although many polyps found on fecal blood screening are probably incidental, [161, 184] about 20% of large adenomas may be associated with elevated fecal blood levels. [161] Fecal blood appears to be a poor marker for adenomas, especially small ones.

Test Validity

Validity is a function that combines sensitivity, specificity, and predictive value, and it varies with the techniques used and populations studied. Validity must be high if a screening test is to perform effectively and efficiently. The sensitivity, or detection rate, ranges in patients with symptomatic colorectal cancer from 40% to 97%. [13, 27, 144, 180, 186, 187] Rigorous prospective studies comparing fecal blood levels against structural gold standards (e.g., colonoscopy) yield average sensitivity estimates for widely used guaiac tests of about 26% (range, 22%–50%) for asymptomatic colorectal cancers (Table 34-5). [161, 189, 190, 191, 192, 193, 194] and [195] The newer immunochemical and guaiac tests may yield somewhat higher cancer detection rates. [142, 143, 196] However, fecal blood levels from patients with asymptomatic colorectal neoplasms often fall within the normal range (Fig. 34-3), and detection by any fecal blood test is not possible if neoplasms do not bleed.

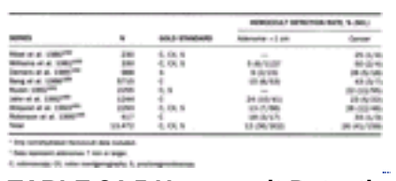

TABLE 34-5 Hemoccult Detection Rates for Asymptomatic Colorectal Neoplasms Based on Comparison against Structural Gold Standards in the Screening Setting*

Screening specificity, the rate of negative tests in those without cancer, ranges from 90% to 98%, [1, 2, 3] and [4, 180] which translates to false-positive rates of 2% to 10%. The positive predictive value, or probability that a positive test result will yield a colorectal cancer, depends on both test specificity and cancer prevalence. For commonly used guaiac tests in the screening setting, the positive predictive value in colorectal cancer averages 5% (range, 2%–18%). [1, 2, 3] and [4, 161, 180] Most positive test results yield no pathology and are considered false positives.

Many asymptomatic colorectal cancers are unfortunately missed by fecal blood screens, and most positive screening test results are caused by trivial conditions other than cancer. Sensitivity can be increased only by reducing specificity, and vice versa (see Fig. 34-4).

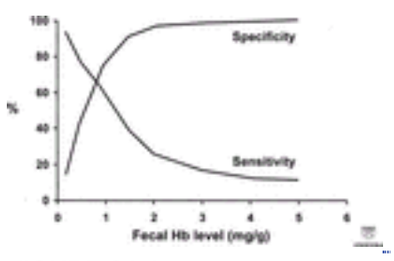

FIGURE 34-4. Fecal hemoglobin screening for colorectal cancer detection showing inverse relationship between sensitivity and specificity. Quantitative stool testing was performed in 2293 persons who also underwent colorectal evaluation by diagnostic gold standards. (From Ahlquist DA, Gilbert JA. Novel fecal markers for colorectal cancer screening: future considerations. Dig Dis 1996;14:132.)

Test Compliance

Screening effectiveness requires not only a high level of test validity but also a compliant population. Operationally, the neoplasm detection rate is the product of test sensitivity and the compliance rate in the target population. Compliance rates have been as low as 19% in general populations and as high as 97% in carefully instructed outpatients using a stool collection device, [138, 180, 197] but compliance averages 30% to 60% in community-based programs. [180, 197] Compliance varies with demography, educational effort, and setting. [197, 198] Physician compliance in screening programs also varies. [180, 199]

Test Effectiveness and Cost

Fecal occult blood screening reduces colorectal cancer mortality by 15% to 30% when regularly applied for more than 10 years (Fig. 34-5). [1, 2] and [3] Because of low detection rates for precursor adenomas, such screening has little influence on cancer incidence. [1, 2] and [3, 200] The benefit is achieved primarily by the detection of early-stage cancer. [201] Screening is ineffective when test intervals exceed 2 years [202, 203] or when compliance is low. [204, 205] and [206]

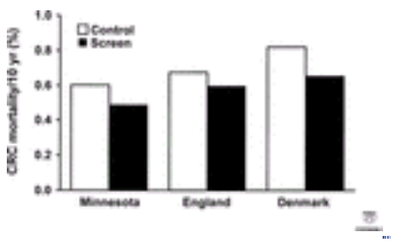

FIGURE 34-5. Colorectal cancer mortality during 10 to 14 years in controlled trials with Hemoccult screening: screened versus control groups. [1], [2] and [3]

Based on mathematical models, [204] the cost-effectiveness of fecal blood testing is comparable to that of other approaches to colorectal cancer screening. [207], [208] However, the measures of effectiveness are lowest with fecal blood testing, and its comparable cost-effectiveness ratio is carried by lower estimated costs. [201] Program costs of fecal blood screening are strongly leveraged by the false-positive rate and the resultant high cost of downstream colonoscopies.

Guidelines for Colorectal Cancer Screening

Because colorectal cancer is so prevalent in the United States, its incidence increases sharply after age 50 years, and its early detection confers a favorable prognosis, the American Cancer Society and the National Cancer Institute recommend annual fecal blood testing alone or in combination with sigmoidoscopy every 5 years for average-risk persons 50 years of age or older. If fecal blood testing is to be practiced for cancer detection, clinicians should be aware of the test limitations and inform their patients accordingly, understand the common causes of occult GI bleeding, and follow a rational strategy in evaluating positive test results.

Future Approaches to Stool Screening

In comparison with structural screening approaches, such as colonoscopy, stool testing has several advantages. Stool testing is noninvasive, requires no cathartic preparation, and can be done without a formal health care visit. Markers with performance characteristics better than occult blood could improve the effectiveness and appeal of stool screening. Exfoliated markers represent rational alternative candidates. [209], [210] Unlike blood, exfoliated markers enter the colorectal lumen continuously with cell shedding, which enhances sensitivity, and have the potential to be unique to neoplastic tissue, which enhances specificity. Neoplasm-associated DNA alterations are a particularly promising class of exfoliated markers, given the known genetic defects in colorectal cancer and the exquisitely sensitive assay techniques available.

Early clinical studies with DNA-based stool testing suggest good sensitivity and specificity for colorectal cancer [211], [212] and apparently much higher rates of polyp detection than are achieved with fecal blood testing. [211] However, large comparative studies in representative general populations must be conducted to corroborate these findings before practice implementation can be justified.

THERAPEUTIC CONSIDERATIONS

The treatment of abnormal occult GI bleeding is dictated by the clinical setting. The critical first step in the management of newly discovered occult bleeding is to exclude a serious underlying lesion. For patients with positive screen test results but normal structural colorectal studies, it is often appropriate to check whether iron stores are low. If concomitant iron deficiency exists, then an aggressive search for the GI bleeding source is warranted. Specific medical or extirpative therapy of the lesion may not only prevent lesion-associated morbidity but also definitively stop further iron loss (see other chapters for lesion-specific treatments). For patients with persistent bleeding from diffuse or inaccessible vascular malformations, nonspecific medical treatment with systemic estrogen, aminocaproic acid, or somatostatin has been attempted. [213]

Whether the bleeding source is correctable or not, a cornerstone of treatment is the replenishment of iron stores. Some patients with chronically bleeding lesions, such as radiation enteritis or Cameron erosions, [65], [77] may be appropriately managed with long-term iron replacement alone because satisfactory medical cures are lacking and surgical approaches may be associated with unwanted morbidity. Uncommonly, blood transfusions are indicated initially in those with severe anemia and cardiovascular compromise.

Oral iron therapy with ferrous sulfate tablets is the preferred approach in most patients with iron deficiency because it is cheap, effective, and usually well tolerated. Other oral preparations include ferrous fumarate, ferrous gluconate, and preparations with added ascorbic acid or other absorption enhancers. [5] Dietary iron replacement is usually inefficient and impractical. For example, it has been calculated that a daily consumption of 5 kg of red meat would be required to provide 60 mg of iron, [117] which is the amount contained in one 325-mg tablet of ferrous sulfate. A maximal adult dose of ferrous sulfate is 325 mg three times daily, and absorption is not appreciably increased with higher doses. [5] With iron replacement, a peak in reticulocytosis occurs after 7 to 10 days, the Hb level usually normalizes within 2 months, and epithelial abnormalities resolve within months. It should be emphasized that the Hb level normalizes well before body iron stores are repleted. [6] If blood loss is not ongoing, satisfactory iron replacement usually requires 3 to 6 months. The serum ferritin level can be used to monitor the adequacy of iron replenishment.

Side effects of oral iron develop in 10% to 20% of patients, appear to be dose-related, are similar for the different preparations, and result in discontinuation in about 8%. [214] In a large prospective study, [214] the side effects most commonly reported included constipation in 13%, diarrhea in 6%, heartburn or epigastric pain in 5%, and nausea in 4%. The side effects of oral iron therapy can often be avoided simply by lowering the dose to one tablet daily or every other day. Caution should be taken not to overreplace body iron stores because excess iron may cause oxidative stress and contribute to a long-term risk for inflammatory, cardiovascular, and neoplastic disease. [54], [215], [216] There is no justification for iron supplementation in iron-replete individuals.

Parenteral iron is indicated in patients with iron malabsorption and in those who do not comply with or cannot tolerate oral iron. [217] Because parenterally administered iron salts are toxic, iron must be injected in a complexed form. Parenteral iron preparations vary in their bioavailability, degradation kinetics, dosimetry, and side effects. [5], [218], [219] The iron complex is taken up by reticuloendothelial tissues and slowly converted to bioavailable iron. Fatal idiosyncratic reactions occur rarely with parenteral iron dextran. [5] In about 10% of patients, a serum sickness-like illness develops with fever, myalgias, arthralgias, lymphadenopathy, and urticaria. [5] As a guiding principle, oral iron is equally effective in correcting iron deficiency anemia and is safer in most cases. [5], [217]

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CHAPTER 35

Dean L. Antonson, James K. Madison, and Renée L. Young

APPROACH TO THE PATIENT WITH UNINTENTIONAL WEIGHT LOSS AND EATING DISORDERS

Body weight is determined by the interplay of calorie intake, activity level, and metabolic rate. Significant alterations involving any of these factors may result in weight loss. This chapter focuses on the patient who presents with unexplained weight loss (i.e., the extent to which any of these factors has been affected is not readily apparent). The first part of the chapter explores unintentional weight loss caused by evolving disease states; the second part focuses on intentional weight loss resulting from eating disorders. Eating disorders present initially as unexplained weight loss because these patients tend to hide their eating behaviors. The patient with unintentional weight loss is not dieting to lose weight, whereas the patient with an eating disorder is in relentless pursuit of this goal. A careful and thorough diet history, therefore, will allow one to begin to separate these patients. A thoughtful and deliberate investigative approach to testing is required for the former patient. Simple screening laboratory tests and referral to an eating disorder center are required for the latter.

UNINTENTIONAL WEIGHT LOSS

Unexplained weight loss in patients is one of the common enigmas presented to the physician. Patients experiencing weight loss are typically alarmed because of underlying fears of malignancy or other serious diseases. Physicians realize that unintentional weight loss is a nonspecific manifestation of many conditions. The evaluation of this problem challenges the physician by its broad range of diagnostic possibilities and seemingly endless differential diagnoses. Cost-efficient medicine makes an efficient diagnostic strategy essential.

Body weight tends to remain constant for long periods of time, although fluctuations in weight on a day-to-day basis are common and reflect deviations in food, water, and salt intake. Weight loss is generally defined as continued loss of more than 5% of body weight, and it is especially of concern when progressive and persistent.

Weight loss is a nonspecific clinical sign with multiple causes. Six major studies of involuntary weight loss have been published since 1981 (Table 35-1). [1], [2], [3], [4], [5] and [6] The causes of weight loss varied widely; approximately 50% of cases could be attributed to cancer or other diseases, [1], [2], [3], [6] and 20% to 60% to psychiatric conditions. [4], [5] The mortality rates of patients presenting with unexplained weight loss ranged from 9% to 25% in the first year and were 38% in 2 years. [1], [2] and [3] In the first three studies cited, mortality rates were lower in patients without defined physical causes of weight loss. [1], [2] and [3] The conclusions from these six studies are the following:

1. Malignancy is not the most common cause of weight loss and is usually diagnosed early in the evaluation period.
2. In most cases of unexplained weight loss, the cause is established initially without extensive evaluation.
3. Psychiatric causes are common, especially in the elderly population.

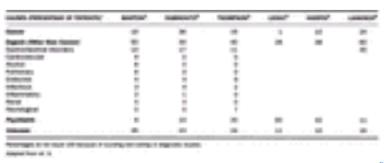

TABLE 35-1 Clinical Studies Evaluating Causes of Unintentional Weight Loss

Etiology

The differential diagnosis of unintentional weight loss is lengthy and, most simply, can be categorized physiologically as follows:

1. Decreased caloric intake because of an altered perception of smell or taste, anorexia, nausea, or an altered perception of satiety
2. Acceleration or alteration of metabolism caused by tumor cells, excess thyroid hormones, or exercise
3. Increased loss of calories in the urine or stool secondary to diabetes mellitus or malabsorption. [7]

Morley [8] has categorized the major causes of weight loss in the geriatric population as either social, psychological, medical, or age-related (Table 35-2). Robbins [9] has published a mnemonic consisting of the nine *D*s of weight loss in the elderly population (Table 35-3); Wise and Craig [10] have added a tenth *D* for “don’t know.”

Social
Isolation
Poverty
Inadequate education
Lack of transportation
Unavailability of preferred foods
Urban decay
Psychological
Depression
Schizophrenia
Bereavement
Bulimia
Manipulation
Sociopathy
Late-life mania
Dementia
Conversion reaction
Anorexia nervosa
Anxiety
Alcoholism
Late-life paranoia
Excessive burden of life (food refusal)
Medical
Increased metabolism—hyperthyroidism, pheochromocytoma, Parkinson disease
Anorexia—drugs, abdominal ischemia, cancer, hyperparathyroidism
Swallowing problems—dysphagia, cerebrovascular accident
Malabsorption—gluten enteropathy
Increased metabolism and anorexia—COPD, cardiac cachexia
Age-Related
Impaired olfactory sensitivity
Appetite suppression
Impaired taste sensitivity

COPD, chronic obstructive pulmonary disease.
Adapted from ref. 7.

TABLE 35-2 Causes of Weight Loss in the Geriatric Population

Dentition
Dysgeusia
Dysphagia
Diarrhea
Disease (chronic)
Depression
Dementia
Dysfunction
Drugs
Don't know

Adapted from refs. 8 and 9.

TABLE 35-3 Ten *D* s of Weight Loss in the Elderly Patient

Medical Causes

Cancer. Malignancy is frequently, but erroneously, thought to be the most common cause of weight loss in the patient without other signs and symptoms (see Table 35-1). [11] However, this diagnosis should be considered early because of the gravity of cancer. Mechanisms of weight loss resulting from malignancy vary, and often more than one factor is present. Anorexia is common in most types of cancer. The ectopic production of hormones such as bombesin, which produces anorexia by sending satiety messages to the central nervous system, [8] or interleukin [1] or tumor necrosis factor (cachectin), another anorectic agent, [12], [13] may cause anorexia. Increased metabolism may play a role in some cancers (e.g., lymphoma, leukemia).

Infection and renal, pulmonary, and cardiac diseases. *Infection* with human immunodeficiency virus (HIV) should be considered, especially in patients in high-risk groups. Fungal infections, tuberculosis, parasitic infections (including amebic abscess), and subacute bacterial endocarditis are often hidden infections that can be associated with weight loss. An early manifestation of *uremia* is anorexia. Patients with *end-stage lung disease* or chronic obstructive pulmonary disease frequently experience increased dyspnea when eating and hence quit eating or limit their intake. The use of accessory muscles of respiration in pulmonary diseases consumes excess calories and potentiates weight loss. *Congestive heart failure* may lead to weight loss through increased metabolic demands, loss of appetite, dietary restrictions, and an inability to find palatable foods that are low in sodium and heart healthy. Atherosclerotic disease can lead to abdominal angina from decreased blood flow to the intestinal tract during digestion.

Endocrine diseases. Unexplained weight loss is one of the hallmarks of undiagnosed or untreated *diabetes mellitus*. This weight loss is usually accompanied by a simultaneous increased intake of food and water intake. *Hyperthyroidism* frequently is associated with weight loss, increased appetite, a diet high in carbohydrates, and a general increase in food intake. In "apathetic" hyperthyroidism, weight loss and weakness (especially proximal myopathy) may predominate. [11] Traditionally, *hypothyroidism* presents with weight gain; however, it may also present with weight loss when anorexia and apathy are the dominant clinical features. Less common endocrine causes of weight loss include *pheochromocytoma* (catecholamine release), *panhypopituitarism* and *adrenal insufficiency* (lack of cortisol support), and *hyperparathyroidism* (increased metabolism and anorexia).

Gastrointestinal diseases. Mechanisms of weight loss in gastrointestinal disease are often multifactorial, including anorexia, obstruction with vomiting, fear of eating, malabsorption, inflammation, and compression of organs, as in massive splenomegaly. Gastroesophageal reflux disease may present with weight loss secondary to peptic strictures, dysphagia, or a fear of eating. Dysphagia may be related also to achalasia, esophageal or gastric cancer, or a cerebrovascular accident or other neurological condition, such as Parkinson's disease or amyotrophic lateral sclerosis. Benign ulcer disease is a common cause of anorexia and weight loss. Palmer [14] reported that 80% of 650 patients with gastric ulcers lost 10 or more pounds and 55% complained of weight loss. The old adage that patients with gastric ulcers lose weight because eating causes pain and patients with duodenal ulcers gain weight because food soothes the pain is not always true. Peptic ulcers may present also with nausea and vomiting, especially when associated with considerable inflammation. Delayed gastric emptying or gastroparesis may present with weight loss secondary to abdominal fullness, nausea, vomiting, distention, and decreased appetite. Atrophic gastritis and subsequent vitamin B_{12} deficiency can lead to loss of appetite. Constipation can cause anorexia and is frequently identified in the elderly. [15] Diabetic enteropathy, cholelithiasis, cholecystitis, inflammatory bowel disease, hepatitis, end-stage liver disease, pancreatitis, esophageal dysmotility, and malabsorption resulting from celiac sprue or other mucosal abnormalities all must be included in the differential diagnosis of weight loss.

Connective tissue diseases. Patients with scleroderma may have swallowing problems or a more diffuse intestinal dysmotility causing weight loss. Systemic lupus erythematosus and rheumatoid arthritis may present with nausea, anorexia, and malaise, resulting in poor oral intake and weight reduction.

Medications. The side effects of drugs and drug-drug interactions cannot be overemphasized, especially in patients taking multiple medications. The weight loss associated with medications can be secondary to decreased appetite, anorexia, nausea, gastroparesis, or diarrhea. Antiretroviral therapy in acquired immunodeficiency syndrome (AIDS) is a good example of this.

Age-Related Factors By the age of 65, approximately half of all Americans have lost a portion of their teeth and have chewing problems. [16] Absence of teeth, ill-fitting dentures, and pain during eating are predictors of significant involuntary weight loss. [16] Age-related impairment of olfactory and taste sensitivity may lead to decreased food intake. Kamath [17] has reviewed studies that may support a decline in sensitivity to taste with increasing age, although it is difficult to determine the role of the many variables that affect taste. Routine oral hygiene has been shown to improve sensitivity to sweet and salty tastes and increase appetite in some patients. [17] Zinc deficiency is sometimes found in the elderly and can be associated with ageusia. [18] Early satiety in elderly patients has been associated with a fall in the opioid feeding drive and hypersensitivity to cholecystokinin. [19] General medical problems such as arthritis, visual impairment, cardiovascular disease, and dementia make it increasingly difficult for elderly people to shop for and prepare food.

Social Causes Humans tend to consume more calories in social situations. Social isolation may lead to weight loss secondary to decreased food intake. Social isolation may be caused by a lack of transportation, poverty, hearing impairment, or other medical ailments that make people reluctant to enter social situations. Elderly patients may experience a unique social situation that is frequently unrecognized (i.e., not being given food choices appropriate for their ethnic background in their care facility). Furthermore, elderly patients are expected to eat with other patients in a care facility; frequently, this assembly includes demented patients with bizarre eating habits. In such an environment, patients may lose the urge to eat. [8]

Psychiatric and Behavioral Causes *Alcoholism* may be diagnosed at almost any age and become a problem at any point in life. Although alcoholism is frequently associated with depression, they are independent risk factors for weight loss. The diagnosis of alcoholism is often difficult to make, and frequently, a vague complaint such as weight loss may be the only physical sign of the problem. *Depression* can lead to apathy, weight loss, and anorexia, and it frequently presents as a vague complaint without the patient being aware that anything is wrong emotionally. Weight loss as a symptom of depression is more common in the elderly than in the younger population, and depression is the most treatable of the psychiatric disorders of later life. [20] The loss of a loved one may cause depression. The reduced food intake associated with the grieving process results in ketosis, and ketone bodies further suppress the appetite, triggering a vicious cycle. [8] The importance of screening patients for depression, especially elderly patients with weight loss, cannot be overemphasized. Several standard tools are available for quick checks for depression.

Neurological Causes The prevalence of various gastrointestinal symptoms among patients with *Parkinson disease* has been delineated by Edwards and associates. [21] A high frequency of defecatory dysfunction, constipation, dysphagia, and disordered salivation has been described. *Dementia* prevalence increases with age and has been estimated to be 2% to 3% in patients between 65 and 79 years old. [18] Appetite disturbances and apathy clinically appear as cognitive function deteriorates. Patients lose the ability to prepare meals, do the grocery shopping, and eat independently. A recent study in patients with Alzheimer disease actually demonstrated that weight loss precedes mild to moderate dementia, and early weight loss is therefore unlikely to be a consequence of the patient's inability or unwillingness to eat. [22] Weight loss in institutionalized patients with Alzheimer disease is commonly related to a marked decrease in staff time spent feeding the patient, compared to time spent by family members feeding the patient. [8] Weight loss in demented patients is also related to an inability to recognize the need to eat, apraxia of swallowing, picas, and ingestion of nonnutritious substances. [8] The weight loss seen in patients with Alzheimer disease is not related to an elevation in resting energy

requirements. [23]

Diagnosis

Documentation An elaborate workup of weight loss is hardly indicated without adequate documentation of the weight loss. Perception of weight loss may be influenced by the rate of weight change, gender, original body size, and underlying disorders present. [24] In up to 50% of patients who report weight loss, the loss is not corroborated by medical records or the family. [1] The most accurate form of documentation is a previous medical record, but if the record is unavailable, a patient's report of a previous weight or change in clothing size that is confirmed by others is sufficient documentation.
Evaluation A directed, complete history and physical examination are key to early diagnosis, efficient evaluation, and initiation of treatment. In the study by Marton and associates, [1] half the patients studied had a chief complaint that pointed to disease of a specific organ. The history should explore symptoms, past medical history, prior surgeries, and cigarette use; it should quantify alcohol intake and the current and past use of medications, and it should include a thorough review of organ systems. The history should also include a search for psychiatric causes, including alcoholism and depression. A standard testing tool should be employed for detecting depression. Potential social, psychological, and physical factors should be identified. The physical examination should begin with attention to the patient's mood, overall appearance, and affect. A complete physical examination includes inspection of the skin and lymphatics; auscultation of the cardiovascular and pulmonary systems; examination of the joints and bony skeleton, breasts, and abdomen; a genital and pelvic examination; examination of the rectum and prostate; and neurological evaluation with a Mini-Mental Status check. A complete examination encompasses not only auscultation but also palpation. Auscultation for bowel sounds and bruits is incomplete without palpation for masses or hepatosplenomegaly. Diagnostic testing should first be directed at clues or areas of concern found during the history and physical examination. If no clues are unearthed during these examinations, simple diagnostic testing is indicated. Tests that are usually of benefit are listed below, but the cost-effectiveness of this initial screening remains to be established.

1. Complete blood cell count
2. Serum chemistries, including tests of renal and hepatic function and measurement of calcium and albumin
3. Urinalysis
4. Chest x-ray film
5. Thyroid-stimulating hormone assay
6. HIV testing if any risk factors are present
7. Erythrocyte sedimentation rate.

In the absence of localizing symptoms or signs or findings on the laboratory work listed above, routine screening for cancer is indicated, as recommended by the American Cancer Society.

1. Fecal occult blood testing of stools (age 40 or older)
2. Flexible sigmoidoscopy/barium enema or colonoscopy (age 50 or older)
3. Cervical Papanicolaou smear in women
4. Mammography (women past the age of 40 years)
5. Prostate-specific antigen (in men age 50 or older).

If the results of these tests are normal and cancer is still suspected, abdominal and pelvic computed tomography may be warranted.

Treatment and Follow-up

If the results of this initial testing are negative, then a period of careful waiting and watching is in order. In the study of Marton and colleagues, [1] more than 65% of the patients who did well during follow-up had completely normal findings on screening tests. In contradistinction, none of the patients who did poorly had normal findings initially. This waiting period is unlikely to result in an adverse outcome because organic disease is rarely found in patients with normal findings on the history, physical examination, and screening laboratory tests.

Another productive approach is to evaluate the nutritional intake and consider the patient's food preferences and any abnormal attitudes toward eating. Offering favorite foods or snack foods may be sufficient to halt weight loss. Avoidance of special medical diets (salt, fat, or caloric restrictions) may actually be of more benefit to the patient with weight loss than strict compliance with the diet.

Early intervention is the key to a successful outcome. In general, the treatment of weight loss must be directed at treating the underlying disease process. Regardless of the cause, when weight loss is severe and manifestations of malnutrition are seen, nutritional supplementation may be required. Liquid supplements are available that are nutritionally complete. Many patients, however, find these supplements less than palatable.

INTENTIONAL WEIGHT LOSS

Anorexia Nervosa

Historical Perspective The first clinical description of anorexia nervosa appearing in the medical literature was published by Richard Morton, physician to King James II, in his textbook of medicine in 1689. [25] During the next 200 years, additional case reports of anorexia nervosa were recorded. [26] In the early 1900s, Simmonds attempted to ascribe the etiology of anorexia nervosa to pituitary insufficiency, [27] a theory that was later disproved by Sheehan. [26], [27] Major advances in our understanding of anorexia nervosa did not occur until the 1960s, when Dr. Hilde Bruch began extensively studying patients with anorexia nervosa and identified the three common hallmarks of the anorexic patient: a distorted body image, an inability to interpret hunger and satiety, and a paralyzing sense of ineffectiveness. [26], [28] Furthermore, and perhaps more important, she recognized that a true loss of appetite did not occur in anorexia nervosa but rather a preoccupation with food and eating. [26] The revolution in our understanding of anorexia nervosa, begun by Bruch and redefined further by Russell [29] and others, has allowed us to define this disorder clearly, gain insight into its multifactorial etiology, and establish effective treatment.
Epidemiology Anorexia nervosa in Western societies has increased dramatically in the last third of the 20th century. Five well-controlled studies from both Europe and the United States have allowed us to quantify this increase. [29] When the incidence of anorexia nervosa was examined through sequential decades from the 1940s through the 1980s in the same population, it was found in Sweden to be increased fivefold, in Scotland fourfold, in Switzerland threefold, and, in two reports from the United States, twofold and fourfold. [29] Increased incidence rates for anorexia nervosa are now also being reported from many other countries. Rates in Japan are equal to or greater than those in the United States, and increased rates have also been reported from China, Spain, Argentina, and Fiji. [30] When the incidence rates are normalized and specific at-risk populations of girls and women between 16 to 25 years of age are examined, incidences range from 30 per 100,000 to as high as 156 per 100,000, making anorexia nervosa the third most common illness in female adolescents and young adults, exceeded only by obesity and asthma. [31] Because of the chronicity of anorexia nervosa, the prevalence rates are several-fold higher than the incidence rates for any specific population. The most commonly quoted prevalence rates in the United States for anorexia nervosa are from 0.5% to 1%. [32] When more broadly defined, the lifetime prevalence rates for anorexia nervosa have been reported to be as high as 3.7%. [30] Similar prevalence rates have been reported from England and Western Europe. [31] The incidence of anorexia nervosa in males is 5% to 10% that in females. The dramatic increase in eating disorders is thought to represent only the tip of the iceberg. Consistent with the current trends of health consciousness in the United States and to a lesser extent in Europe, dieting behaviors among adolescent girls and young women have profoundly increased. The incidence and prevalence of severe dieting behaviors that fail to meet the specific criteria for anorexia nervosa are reported as between 5% and 10% of young females. [31], [33] Paralleling this rise has been a concomitant increase in body image dissatisfaction. In one U.S. study, half of all underweight adolescent girls wanted to lose weight, and 80% claimed that they were unhappy with their present weight. [34] Overall, one half to two thirds of adolescent girls and young women perceived themselves as too fat, although only 15% were documented to be overweight. [34] In a recent study from the Centers for Disease Control and Prevention, almost 50% of adolescent girls were involved with dieting, whereas only 15% of boys were similarly concerned. [32] Forty percent of these patients felt very negatively about their body image, with the greatest displeasure being focused on their hips, waist, and thighs, a consistent finding in patients with eating disorders. [32] Although most early studies reported that anorexia nervosa occurs primarily in white, middle- and upper-class females, more recent studies have documented that anorexia nervosa is present in all ethnic groups and at all socioeconomic levels. [31], [33], [35] In general, eating disorders are more prevalent in Native Americans, equally prevalent in Hispanics and whites, and less common in blacks and Asians. [30] Anorexia nervosa is especially prominent in persons engaging in appearance-related sports and occupations, such as acting, ballet dancing, modeling, and gymnastics. The risk for the development of anorexia nervosa is greatest between the ages of 15 and 25 years, although

extension into age groups as young as 9 or 10 years is being reported more frequently. Numerous risk factors for the development of anorexia nervosa have been identified. In addition to age, gender, and the strong influence of Western culture, factors include the onset of dieting behavior, excessive exercise, the presence of a physical illness that results in initial weight loss, and the presence of characteristic personality features, such as low self-esteem, obsessive-compulsive and avoidant personality traits, body image dissatisfaction, and body image distortion. [31] In a small percentage of patients, especially those who present under 14 years of age, anorexia nervosa may develop in association with a streptococcal infection. [36] Genetic factors are also prominent; the incidence of eating disorders is increased in the first-degree relatives of patients with anorexia nervosa and in monozygotic twins. [31], [34] The incidence of anorexia nervosa in second-generation immigrants from underdeveloped countries is increased to rates similar to those in the new geographic location, a finding that highlights the importance of cultural factors. [31]

Etiology Although the etiology and pathogenesis of anorexia nervosa are unknown, a certain commonality exists in all the theories about the factors responsible for the development of anorexia nervosa. A succession of factors are thought to be involved: predisposing factors (individual, familial, and sociocultural), precipitating factors (stressors, diet, illness, and weight loss), and perpetuating factors (cognitive reinforcement and effects of starvation). [37] Although no single theory has yet been accepted, all theories propose that antecedent events and variables predispose the susceptible adolescent or young adult to pursue the goal of weight loss. Both positive and negative reinforcements allow the disorder to progress. Conflicts, stresses, and failure result in low self-esteem and dissatisfaction, and the perfectionistic tendencies of these individuals then funnel the need to control some aspect of their life toward dieting and weight loss (thinness being equated with success). An ever-encompassing spiral of weight loss, fear of weight gain, intensified dieting, body image dissatisfaction, and the effects of starvation lead to an uncontrollable progressive weight loss with an increasing feeling of success and control.

Physical Evaluation It is difficult for the patient in whom anorexia nervosa is developing to hide her disease. The rather typical appearance of marked starvation and cachexia, along with noted muscle wasting, a decrease in subcutaneous tissue, and a prepubescent body habitus, are easily identified. Vital signs frequently demonstrate hypothermia, with body temperatures 1° to 2°F below normal, bradycardia with a resting pulse rate as low as 40/min, and hypotension with blood pressures as low as 90/60 mm Hg. Moderate to marked acrocyanosis is almost uniformly present. Skin and hair changes are seen, including a yellow discoloration of the skin secondary to carotenemia, thinning of the scalp hair, and the development of lanugo hair, particularly over the face, upper arms, and back. The skin may also be dry, pale, and atrophic. Deep tendon reflexes are decreased on neurological examination. Despite the cachectic appearance, individuals with anorexia nervosa often appear energetic, although on rare occasions profound degrees of lethargy are present. Lethargy occurring in the individual with anorexia nervosa is of concern and may be the harbinger of early organ failure. It is uncommon for the individual with anorexia nervosa to present to the physician complaining of the physical changes that have resulted from starvation. Rather, the patient with anorexia nervosa will present with complaints that include menstrual irregularity, depression, lethargy, or repeated sports injuries. Additionally, complaints may include weakness, palpitations, chest pain, abdominal pain, or frequent headaches.

Laboratory Evaluation Despite the profound starvation and emaciation, most serum laboratory and chemistry values remain normal. Rarely do serum laboratory studies reflect the level of starvation present. The most common electrolyte abnormalities include hypokalemia, hyponatremia, and a mild metabolic alkalosis. [38] Classically, the individual with anorexia nervosa demonstrates what has been termed the *sick euthyroid* state, with low levels of triiodothyronine (T_3), thyroxine (T_4), and thyroid-stimulating hormone (TSH). This pattern is easily differentiated from hypothyroidism, in which the TSH levels are markedly increased. An additional hallmark laboratory finding in patients with anorexia nervosa is a low sedimentation rate, often recorded as either 0 or 1. This feature allows easy differentiation of anorexia nervosa from other prominent inflammatory processes that may result in significant weight loss. The laboratory and radiologic abnormalities commonly seen in patients with anorexia nervosa are listed in Table 35-4.

Hypokalemia, hyponatremia, metabolic alkalosis
Low thyroid tests, low T_3, T_4, TSH
Low erythrocyte sedimentation rate
Mild leukopenia, relative lymphocytosis
Hypercholesterolemia
Mild increase in hepatic aminotransferases
Mild increase in creatinine
Elevated cortisol
Microcardia
Widened sulci, enlarged ventricles
Decreased pituitary size
Hypoglycemia
Carotenemia
Hypomagnesemia
Anemia, thrombocytopenia
Low phosphorus
Mild increase in serum proteins
Low alkaline phosphatase
Decreased FSH, LH, and prolactin
Electrocardiographic changes: decreased P-wave amplitude, prolongation of PR interval, ST-T wave changes, junctional rhythm
Decreased zinc levels
Metabolic acidosis

FSH, follicle-stimulating hormone; LH, luteinizing hormone; T_3, triiodothyronine; T_4, thyroxine; TSH, thyroid stimulating hormone.

TABLE 35-4 Laboratory and Radiologic Abnormalities in Anorexia Nervosa

Medical Complications The extensive array of medical complications resulting from anorexia nervosa is listed in Table 35-5. These conditions are generally attributable to progressive starvation and emaciation. The most important ones are discussed below.

TABLE 35-5 Medical Complications of Anorexia Nervosa

After suicide, cardiac arrhythmias are the most common cause of death in patients with anorexia nervosa. [34], [38] Arrhythmias occurring in the face of profound bradycardia are most worrisome and carry a significant risk for tachyarrhythmias and sudden death. [34] Additionally, significant prolongation of the QT interval is thought to be an ominous sign predictive of a significant risk for sudden death. [38] Starvation causes both acute and long-term effects on bone mineralization and development. Poor bone growth, reduced bone turnover, and osteoporosis occur to some degree in all patients and may become severe. [38] The bone density measurements of 50% of patients with anorexia nervosa are two standard deviations below the mean. [39] Osteoporosis that develops after puberty is reversible. [38] Severe malnutrition and osteoporosis in early adolescence, however, may lead to a stunting of linear growth, particularly when associated with primary amenorrhea. [40]

Menstruation and reproductive function are significantly affected by anorexia nervosa. The pulsatile release of luteinizing hormone is diminished, pituitary gland output in response to gonadotropin-releasing hormone stimulation is reduced, feedback to estrogen is lost, and multifollicular changes occur within the ovaries. [38] Reproductive function is also affected; 10% of a large sample of anorexic women demonstrated infertility problems. [38] In this same cohort, the rate of infant prematurity was increased twofold, and perinatal mortality was increased sixfold. [38] Structural changes within the brain are common in patients with anorexia nervosa and are thought to be secondary to the profound effects of starvation. [41] Computed tomography and magnetic resonance imaging of the brain have demonstrated widening of the sulci and an increase in ventricular size. [41] Following weight restoration, white matter and cerebrospinal fluid volume slowly return to normal. Gray matter volume deficits may persist for years, may not fully return to normal, and may be associated with persistent deficits in neuropsychological parameters. [30]

Diagnostic Criteria Table 35-6 summarizes the revised diagnostic criteria for anorexia nervosa as listed in the *Diagnostic and Statistical Manual of Mental Disorders,* 4th edition (DSM-IV). [42]

1. Refusal to maintain body weight over a minimally normal weight for age and height (e.g., weight loss leading to a maintenance of body weight <85% of that expected; or failure to make expected weight gain during a period of growth leading to body weight <85% of that expected).
2. Intense fear of gaining weight or becoming fat, even though underweight.
3. Disturbance in the way in which one's body weight or shape is experienced, undue influence of body weight or shape on self-evaluation, or denial of the seriousness of the current low body weight.
4. In postmenarcheal females, amenorrhea—that is, the absence of at least three consecutive menstrual cycles. (A woman is considered to have amenorrhea if her periods occur only following hormone [e.g., estrogen] administration.)

Restricting type: During the current episode of anorexia nervosa, the person has not regularly engaged in binge-eating or purging behavior (i.e., self-induced vomiting or the misuse of laxatives, diuretics, or enemas).

Binge-eating/purging type: During the currentepisodes of anorexia nervosa, the person has regularly engaged in binge-eating or purging behavior (i.e., self-induced vomiting or the misuse of laxatives, diuretics, or enemas).

From ref. 42.

TABLE 35-6 Diagnostic Criteria—Anorexia Nervosa

Treatment Because of the complexity and interweaving of the psychological, nutritional, and medical problems associated with anorexia nervosa, treatment must be structured to incorporate all three of these areas. Reversal of the significant starvation that has preceded treatment is the most important goal of initial therapy. Feedings are reestablished by increasing the caloric intake to approximately 200 to 250 calories above the caloric intake at the time of presentation. The caloric intake is then sequentially increased by 250 to 300 calories every 4 to 5 days to achieve a steady weight gain of approximately 1.5 kg/wk for the hospitalized patient and 0.75 to 1.0 kg/wk for the outpatient. Meals must be supervised closely and patients supplemented with liquid enteral products for food items not consumed. A slow, steady weight gain to 100% of ideal body weight for individuals younger than 16 years and to 90% to 100% of ideal body weight for individuals older than 16 years is set as the goal. Ideal body weight determinations are obtained from the Centers for Disease Control or National Center for Health Statistics standard growth charts for pediatric patients younger than 16 years and from one of the standard height and weight tables, such as the 1983 Metropolitan height and weight tables, for older adolescents and young adults. The nutritionist must play a prominent role not only in supervising meals, but also in addressing and resolving the patient's unusual thoughts regarding food, eliminating food rituals and fears, and removing the misconceptions about fat and calories that have often developed. Involvement by the nutritionist in overcoming the patient's body image distortion and dissatisfaction is extremely important. Because of the hypermetabolic phase of early refeeding, during which time a portion of the calories ingested is expended as heat, the nutritionist may also be called on to assist the anorexic patient in condensing calories. Daily intakes may at times reach 4000 to 5000 calories to maintain weight gain. Medical treatment primarily focuses on nutritional rehabilitation in conjunction with the dietitian. In the first several days of treatment, any electrolyte abnormalities present must be corrected. Hypokalemia is particularly common in patients with anorexia who are purging by vomiting or using laxatives. Potassium supplementation may be required if potassium levels are below 3 mEq/L. Constipation is managed primarily with the use of psyllium and milk of magnesia; stimulant cathartics should be avoided. Gastric fullness and gastric bloating are frequent and generally resolve with 3 to 4 weeks of refeeding, but if severe, a promotility agent may be required. Most other changes associated with starvation self-correct with slow refeeding and do not require significant medical management. Hospitalization may be required if the patient presents initially with a body weight at or below 70% to 75% of the ideal weight, or if hemodynamic compromise, significant electrocardiographic abnormalities, dehydration, electrolyte abnormalities, profound weakness, mental confusion, or failure to progress with intensive outpatient management is noted. Only rarely are enteral tube feedings or hyperalimentation necessary. Regardless of the refeeding approach, extreme caution must be taken to avoid a too rapid or aggressive replenishment of calories, especially in severely underweight patients. This can precipitate the refeeding syndrome, caused by the rapid depletion of already low levels of phosphorus, and result in severe fluid retention, cardiac arrhythmias, congestive heart failure, muscle weakness, delirium, seizures, or death. [43] The psychotherapeutic and psychopharmacological treatment for both anorexia nervosa and bulimia nervosa is discussed at the end of the chapter.

Prognosis: Mortality and Morbidity Morbidity and mortality from anorexia nervosa remain significant. Earlier mortality rates in the 5% to 10% range have now been demonstrated to represent an early mortality of 5% and a late mortality that may be as high as 13% to 20%. [34] Suicide remains the No. 1 cause of death, followed by cardiac arrhythmias, infections, gastrointestinal complications, and emaciation. [38] In a longitudinal study, rates of death were 10 times and rates of suicide 58 times those expected after 11 years. [44] Morbidity rates are estimated to be 25%, with morbidity reflecting the multiple organ system dysfunctions associated with starvation. [45] The most common of these include osteoporosis, renal insufficiency, and infections. [38] Studies are beginning to appear on the long-term prognosis for patients with anorexia nervosa. Rates for full recovery are widely variable, ranging from 32% to 71% after 20 years. [34] Based on a large review of studies with follow-up for at least 4 years after initial treatment, about 44% of patients had outcomes rated as good, 28% variable, and 24% poor; 5% died. [30] Psychiatric comorbidities are also beginning to be recognized. Follow-up studies of adolescents have demonstrated that 30% continue to have problems with affective disorders and 43% with anxiety disorders. [34] Other common persisting comorbid illnesses include obsessive-compulsive disorder, avoidant personality disorder, and substance abuse. [30]

Bulimia Nervosa

Historical Perspective Although reports of engorgement and induced vomiting have been recorded since ancient times, the development of bulimia nervosa is of relatively recent origin. The first clinical description of bulimia nervosa was provided by Gerald Russell in 1979 [46] and was further defined in the *Diagnostic and Statistical Manual of Mental Disorders,* 3rd edition (DSM-III) in 1980. The term *bulimia,* however, was first used by Travisa in 1398 to mean "immoderate appetite." [47] Gradually, the term became associated with gluttonous overeating and induced vomiting and was included as a disorder in a text by Janet based on his classical work on neurosis in 1903. [47], [48] The psychiatric disorder of bulimia nervosa, however, should be distinguished from the gluttony and induced vomiting (bulimia) that were part of the social rituals of earlier times, not a maladaptive behavior used to control calorie consumption and promote weight loss. The key central issues of bulimia nervosa, including loss of control, low self-esteem, and guilt, are not apparent in these early descriptions.

Epidemiology Like that of anorexia nervosa, the incidence of bulimia nervosa has increased dramatically during the last 20 years. This is a disorder that occurs primarily in adolescents and young adult women, with a male-to-female ratio between 1:10 and 1:20, similar to that for anorexia nervosa. The overall incidence of bulimia nervosa is higher than that of anorexia nervosa and is estimated to range from 2% to 5% for female high school and college students. [49] Some series have reported incidences as high as 20%. [49] Prevalence rates for bulimia nervosa range from 1% to 10% in most studies. [32], [34], [50] The incidence of individuals demonstrating some but not all of the symptoms of bulimia nervosa suggests that 17% to 19% of women of college age engage in these behaviors. [49]

Etiology The pathogenesis of bulimia nervosa, like anorexia nervosa, is multifactorial, encompassing familial, developmental, social, cultural, physiological, and genetic factors. There is a strong association of bulimia with affective disorders (depression and dysthymia), and a strong family history of affective disorders in bulimic patients (as high as 50% in first-degree relatives) has been noted. [51], [52] Additionally, alcoholism and drug addiction are prevalent, with alcoholism being reported in 50% to 60% of first- and second-degree family members. [53] Families of patients in whom bulimia nervosa develops are described as having high levels of conflict with unstructured and ambivalent lines of authority, highly achievement oriented, and lacking expressivity. [54] Affective instability and low self-esteem are the hallmark personality features of the individual with bulimia nervosa. [54] As in anorexia nervosa, comorbid psychiatric illnesses such as depression or dysthymia (50%–75%), substance abuse (30%–37%), personality disorders, especially borderline personality disorder and avoidant personality disorder (42%–75%), and bipolar disorder (4%–6%), are frequently present. [30] Initially, a high index of sexual abuse was reported to be a precipitating factor, but more recent studies have shown that the incidence of sexual abuse in patients with bulimia nervosa is no higher than that in adolescents presenting with other psychiatric disorders. [55]

Sociocultural factors are quite prominent precipitating features in bulimia nervosa, as in anorexia nervosa. Bulimic individuals are also in pursuit of thinness. They equate thinness with success and control, and this is compounded by an equally strong societal view of obesity as a stigma. Unlike the individual with anorexia nervosa, in whom dieting leads to malnutrition, decreasing mental function, decreasing self-esteem, and the subsequent need to further intensify dieting, the individual with bulimia nervosa exhibits a divergent pattern. The poorly self-controlled and impulsive individual in whom bulimia nervosa develops cannot override the strong urge of hunger, loses control and binges on food, feels a loss of control resulting from this behavior, and further intensifies dieting. When dieting becomes unsuccessful, purging behavior ensues. Although initially purging is followed by a sense of relief, the bulimic individual begins to loathe the loss of self-control resulting from the purging behaviors and attempts to intensify dieting. A positive reinforcing loop is thus established, resulting in repetitive cycles of binging and purging. Characteristically, the binge episode lasts for 1 to 2 hours, during which a large number of calories are ingested, with the average intake estimated to be 4000 calories. [34]

Physical Evaluation Most bulimic individuals are of normal weight, and suspecting or establishing the diagnosis based on a routine physical examination is difficult. Bulimic individuals remain quite secretive about their behaviors, concealing them until their difficulties with loss of control lead to a desire to confront it. Unlike the individual with anorexia nervosa, the bulimic individual perceives her difficulties as problematic and will often seek help or assistance, particularly when the disorder progresses to the point that most of her daily thoughts and actions are controlled by the process. Before this happens, the bulimic individual may seek medical attention for one of the associated signs or symptoms of the disorder, including fluid retention (with a request for diuretics), abdominal fullness, frequent headaches, chest pain, constipation, hematemesis, and dental problems. On physical examination, three distinct abnormalities may be recognized that are pathognomonic for bulimia nervosa. These include calluses or scarring over the dorsum of the hand or fingers used to induce vomiting (Russell sign), hypertrophy of the salivary glands (sialoadenitis), and erosion of the dental enamel (perimolysis) (see Chapter 50). Up to 70% of bulimic individuals will be noted to have dental caries, and an even greater percentage will demonstrate perimolysis on careful examination, [56] particularly involving the lingual and occlusal surfaces. A chronic sore throat is frequently reported, and on physical examination the posterior pharyngeal areas are often erythematous.

Laboratory Evaluation On laboratory testing, the bulimic individual often demonstrates electrolyte abnormalities, most commonly hypokalemia, hyponatremia, and

hypochloremic alkalosis. [57] Profound electrolyte abnormalities with potassium levels below 3.0 mEq/L can be seen with laxative or diuretic abuse. Chronic dehydration is also frequently present. The laboratory and radiologic abnormalities commonly seen in patients with bulimia nervosa are listed in Table 35-7.

Hypokalemia, hyponatremia, hypochloremic alkalosis
Chronic dehydration
Metabolic acidosis
Elevated cortisol
Increased prolactin levels
Low magnesium levels
Hypoglycemia
Azotemia

TABLE 35-7 Laboratory and Radiologic Abnormalities in Bulimia Nervosa

Medical Complications Although the weight of most bulimic patients remains normal, the yo-yo dieting behavior associated with binging and purging is generally accompanied by a state of semistarvation. This results in a constellation of side effects and common medical complications quite similar to those seen in anorexia nervosa (Table 35-8). In addition to the medical complications in bulimia nervosa that result from starvation, the purging behaviors cause gastrointestinal side effects, including abdominal and epigastric pain, hematemesis, persistent sore throat, and constipation. Electrolyte abnormalities are frequently seen with laxative abuse and may be quite prominent. The number of laxatives taken varies from three or four per day to 150 per day. The long-term use of stimulant cathartics may ultimately result in a cathartic colon, with the patient requiring extensive medical and possibly surgical management.

[illegible]

TABLE 35-8 Medical Complications of Bulimia Nervosa

Electrocardiographic abnormalities are common in patients with bulimia nervosa. Particularly worrisome are those occurring in association with profound hypokalemia, which can result in arrhythmias and sudden death. Cardiac arrhythmias are the leading cause of death in bulimia nervosa. [34]

Diagnostic Criteria Table 35-9 details the revised diagnostic criteria for bulimia nervosa listed in DSM-IV. [42] Two subtypes of bulimia nervosa have now been described to help clarify the diagnosis in those patients who have mixed features of both anorexia and bulimia.

1. Recurrent episodes of binge eating. An episode of binge eating is characterized by both of the following: (1) eating, in a discrete period of time (e.g., within any 2-hour period), an amount of food that is definitely larger than most people would eat in a similar period of time and under similar circumstances; and (2) a sense of lack of control over eating during the episode (e.g., a feeling that one cannot stop eating or control what or how much one is eating).
2. Recurrent inappropriate compensatory behavior in order to prevent weight gain, such as self-induced vomiting, misuse of laxatives, diuretics, enemas, or other medications, fasting, or excessive exercise.
3. These behaviors both occur, on average, at least twice a week for 3 months.
4. Self-evaluation is unduly influenced by body shape and weight.
5. The disturbance does not occur exclusively during episodes of anorexia nervosa.
Purging type: During the current episode of bulimia nervosa, the person has regularly engaged in self-induced vomiting or the misuse of laxatives, diuretics, or enemas.
Nonpurging type: During the current episode of bulimia nervosa, the person has engaged in other inappropriate compensatory behaviors, such as fasting or excessive exercise.

From ref. 42.

TABLE 35-9 Diagnostic Criteria—Bulimia Nervosa

Treatment Most of the medical complications resulting from bulimia nervosa will slowly resolve once the purging behaviors have been controlled, and they do not require significant medical management. If electrolyte abnormalities are severe, and particularly if potassium levels are below 3.0 mEq/L, potassium supplementation will be required during the first few days of treatment. A dental consultation should be obtained for most of the individuals who have used vomiting as a means of purging. The most troublesome medical complication is the development of pseudo-Bartter syndrome, which can follow the abrupt discontinuation of diet pills or laxatives in patients who have significantly abused these substances. [58] Chronic laxative and diuretic abuse leads to chronic dehydration, activating the renin-angiotensin-aldosterone system and resulting in hyperaldosteronism. Significant fluid retention and peripheral edema can develop within 2 to 7 days after these drugs have been discontinued. Diuretic therapy is frequently required for the treatment of severe edema, with slow tapering of the diuretics over 4 to 12 weeks, while aldosterone levels normalize. Potassium supplementation is frequently required in conjunction with diuretic administration. The nutritional rehabilitation of the patient with bulimia nervosa focuses on the need to stop the purging behaviors, dispel the myths and inaccurate notions that have developed regarding food, calories, and fat, and overcome body image distortion.

Prognosis: Morbidity and Mortality The mortality rates for patients with bulimia nervosa within 2 to 5 years after diagnosis continue to remain at 5%. [34] Between 50% and 60% of individuals will demonstrate recovery over this same period of time, with a relapse rate between 30% and 50%. [34] In an extensive review of several follow-up studies by Woodside, [34] 2 years after treatment, 20% to 25% of individuals were well, 10% to 15% had minor recurrent difficulties, 15% had major recurrent difficulties, 15% were chronically ill, and 20% to 25% had fully relapsed. In one large study, 6 years after successful treatment, 60% of patients demonstrated a good, 29% an intermediate, and 10% a poor outcome, and 1% had died. [30] The most prominent factor associated with failure to remain well was concomitant alcohol or drug use. [34]

PSYCHOTHERAPY AND PSYCHOTROPIC MEDICATIONS FOR ANOREXIA NERVOSA AND BULIMIA NERVOSA

Successful treatment of anorexia nervosa and bulimia nervosa must address core issues such as distorted beliefs about weight and eating, distorted body image, fear of weight gain, perfectionist self-criticism, interpersonal mistrust, a sense of ineffectiveness, and poor self-regulation. Before intervening, the treatment team should evaluate the importance of these issues to the individual patient. Other issues often include deficits in assertiveness, poor impulse control, fears of the demands of maturity, and poor conflict management skills. A comprehensive evaluation also examines family factors that may contribute to eating disordered behavior, such as emotional overinvolvement, poor role boundaries within the family, lack of conflict resolution, and age-inappropriate intrusiveness.

The initial assessment also should determine whether the patient's symptoms warrant inpatient or some other intensive therapy rather than outpatient care. The presence of additional significant psychopathology decreases the patient's chances for successful completion of low-intensity protocols. Of particular concern is the coexistence of major depression, alcoholism or other substance use disorders, or personality disorders involving poor impulse control, self-injurious behaviors, or extreme distortions in interpersonal relationships. Overt psychotic symptoms, severe dissociative reactions, or symptoms that include assaultive behavior often must receive first priority in treatment planning. A careful psychodiagnostic interview combined with appropriate psychological testing is particularly valuable at this stage.

Hospital-based programs for eating disorders typically are used for those patients with the most severe symptoms or with the complications described above. For anorexic patients below 70% to 75% of their ideal body weight, outpatient programs are not usually feasible, and even partial hospital programs may not offer the intensity of treatment necessary to ensure initial compliance and progress. Clearly, those anorexic patients who are medically compromised or require tube feeding are not appropriate candidates for outpatient care. In the authors' experience, younger patients with a more recent onset tend to show more signs of medical instability at higher weights than do older patients whose condition is more chronic. Because these younger patients also have the greatest opportunity to recover from such potential long-range problems as osteoporosis, we advocate more aggressive treatment with hospitalization at somewhat higher weights. Among both anorexic and bulimic patients, failure to achieve appropriate eating patterns and weight on an outpatient basis should be regarded as a reason to move the patient to more intensive care, such as partial hospital, residential, or inpatient treatment. The presence of suicidal tendencies, substance abuse/dependency, or other significant psychological symptoms in addition to the eating disorder also warrants a higher level of care.

Psychopharmacology

The use of psychotropic medication to treat eating disorders continues to be a very active field. Unfortunately, studies have failed to demonstrate the effectiveness of any psychotropic medication for treating anorexia nervosa. In clinical experience, some patients with anorexia respond to drug therapy; however, this usually reflects the selection of a psychoactive agent targeted at psychopathology other than the anorexia. In brief, psychotropic medication should be used with anorexic patients only when the target of the intervention is some concurrent symptom. The most significant caveat in this area is that severely underweight patients can display other symptoms, particularly depression, anhedonia, and lethargy, simply as a result of their starvation. The general consensus is that treating such depressive symptoms when they are a consequence of starvation is not appropriate clinical management.

In contrast to the results with anorexia, numerous controlled studies have shown effectiveness for the use of antidepressants in decreasing bulimic symptoms. Patients treated with imipramine, [59] desipramine, [60] and fluoxetine, [61] among others, have shown a decreased frequency of binging, purging, and a variety of other measures of bulimic behavior. The effectiveness is often related to the dose, and some studies have shown that moderately long trials of the medication are more effective than brief treatments. Potential problems with using this approach exclusively in treating bulimia include patient noncompliance and the fragility of the effect after medication is discontinued. The results of studies in which earlier classes of antidepressants were used suggest that a smaller percentage of patients recover receiving medication alone compared with psychotherapy. [59]

Family Therapy

Family therapists have applied a variety of methods to the treatment of eating disorders. Structural family therapy [62] and systemic [63] and behavioral methods [64] have been demonstrated to be effective in patients with eating disorders. However, no studies have isolated specific factors that would lead to the selection of family therapy for a particular patient. When the initial assessment identifies specific skill deficits within the family, methods developed to intervene with these deficits are applied. In presenting recommendations for family therapy, it is important that the family members understand that the recommendation is being made so that they may participate in the patient's recovery—not because the treatment team believes that the family caused the eating disorder. Much resistance and fear are generated when the family members perceive that they are viewed as the villains of the piece rather than as potential heroes.

A significant concern is that only one study has compared the effectiveness of family with another form of treatment (i.e., individual psychotherapy for the treatment of anorexia and bulimia). [65] Family therapy was found to be differentially beneficial for patients with anorexia who were younger than 18 years of age and whose disorder had been present for less than 3 years. Older patients with anorexia did significantly better with individual therapy. Patients with bulimia did not show differential effects and did not show much improvement with either therapy method used in this study. In the absence of larger and more complete studies, this finding at least signals the need to use family therapy in a circumspect manner in dealings with adult patients. It does not speak to the use of marital therapy.

Cognitive-Behavioral Therapy

The most comprehensively documented psychotherapy for bulimia is cognitive-behavioral therapy. Treatment may be delivered in either group or individual formats. The purpose of this form of therapy is to alter behavioral patterns, distorted beliefs, and dysfunctional self-perceptions that promote eating disordered behavior. The therapist helps the patient test and invalidate existing assumptions and thoughts about weight and body size. Ultimately, the patient learns to challenge mistaken beliefs and attitudes about herself. Fairburn [66] has demonstrated that this approach can produce success rates as high as 70% in the outpatient treatment of bulimia. These rates appear durable for up to 2 years of follow-up. Similar strong results have been reported for the group treatment of bulimia by Mitchell and colleagues. [59] Cognitive-behavioral therapy has been shown to be superior to behavior therapy as well as placebo. In studies assessing the effectiveness of cognitive-behavioral therapy compared to antidepressant medication alone and the combination of antidepressants and cognitive-behavioral therapy, the cognitive-behavioral therapy is usually, [59] but not always, [60] superior to antidepressants alone and is equal to the combination treatment. [59] With anorexia, the evidence is less powerful, but it remains persuasive enough that cognitive-behavioral therapy should be considered the gold standard of treatment for this disorder.

Interpersonal Therapy

Interpersonal therapy was originally developed as a treatment for depression and has been applied successfully in this area. [67] It has demonstrated effectiveness that cannot be statistically differentiated from that of cognitive-behavioral therapy. [68] The care with which these studies were conducted and the clarity of the results suggest that interpersonal therapy be given serious consideration in the treatment of bulimia, even though more research must be conducted. In interpersonal therapy, the focus is not on weight or body image issues per se. Rather, the therapist helps the patient evaluate interpersonal skills, conflicts with significant people, difficulty in role transitions, and loss. The emphasis is on solving interpersonal problems and patterns of relating to others rather than on changing thoughts. At this time, no research-based criteria are available for recommending interpersonal therapy versus cognitive-behavioral therapy for a particular patient.

We recognize that many other psychotherapeutic approaches have been applied to the treatment of eating disorders. This review has been selective, emphasizing those treatments that are most commonly used in the field or whose effectiveness is supported by clear data. The most significant role for the health care provider in supporting patients during their recovery from an eating disorder is to dispel pessimism regarding treatment. Although old methods of treatment produced discouragingly low rates of recovery, modern treatment studies indicate that eating disorders are very treatable and that many people can expect improvement or recovery.

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CHAPTER 36

Judith Korner, Samuel Klein, and Louis J. Aronne

APPROACH TO THE PATIENT WITH OBESITY

Obesity is an overabundance of adipose tissue that arises from an excess of energy intake compared to expenditure. Obesity is not simply the result of gluttony and a lack of willpower. Rather, each individual inherits a set of genes that control appetite and metabolism, and a genetic tendency to gain weight that may be exacerbated by environmental conditions such as food availability, level of physical activity and individual psychology and culture.

Obesity is defined in terms of body mass index (BMI), which provides an index of the relationship between weight and height. The BMI is calculated as weight (in kilograms) divided by height (in square meters), or as weight (in pounds) times 703 divided by height (in square inches). The primary classification of overweight and obesity relates to the BMI and the risk for mortality (Table 36-1). [1] The prevalence of obesity in adults in the United States without coexisting morbidity increased from 12% in 1991 to 17.9% in 1998. Gastroenterologists are in a unique position to manage this disease because of the multiple gastrointestinal conditions associated with increased body weight. This chapter reviews the molecular mechanisms that regulate food intake and body weight, and the comorbidities, in particular gastrointestinal complications, associated with excess body weight. The clinical approach to the patient with obesity is also outlined.

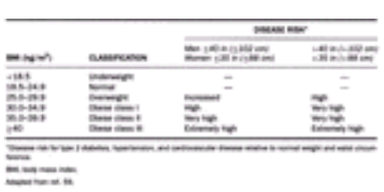

TABLE 36-1 Classification of Weight by BMI, Waist Circumference, and Associated Disease Risk*

MOLECULAR CIRCUITRY OF BODY WEIGHT REGULATION

Multiple factors determine how much we eat and how much we weigh. Weight gain occurs when dietary energy intake is not in balance with daily energy expenditure, and obesity represents an interplay between diet, physical activity, environment, age, and genetics. The estimated heritability of the BMI is between 50% and 90%—that is, the susceptibility to obesity is determined largely by genetic factors, but the expression of these genes is influenced by our environment. In today's modernized societies, the access to large quantities of high-calorie, high-fat foods and the decreased demand for physical activity are responsible for the marked increase in the prevalence of obesity. [2]

The discovery of leptin and other genes responsible for obesity in rodents has made a considerable impact on our understanding of body weight regulation and may lead to exciting advances in the treatment of human obesity. Leptin (derived from the Greek *leptos,* meaning "thin") is a hormone produced by fat cells that circulates at levels proportional to body fat content. Leptin crosses the blood-brain barrier to bind to its receptor in the hypothalamus, thereby activating signals that inhibit food intake and increase energy expenditure. When leptin is given to leptin-deficient mice, obesity and metabolic abnormalities such as hyperglycemia, hyperinsulinemia, and hypercortisolemia are reversed. [3] However, the initial hypothesis that human obesity results from a deficiency of leptin has not been upheld. In fact, most obese humans have high circulating levels of leptin, and only a few individuals with severe obesity have been identified with either a congenital leptin deficiency or a mutation in the leptin receptor gene. [4] In early clinical trials, high doses of subcutaneously administered leptin, often associated with reactions at the injection site, had only a modest effect in reducing body weight. [5] Therefore, it appears that obese persons are "leptin resistant," and if a defect in the leptin pathway exists in human obesity, it is probably a post-receptor defect in the subsequent leptin signaling pathway.

Leptin is also present in the gastric mucosa of humans within secretory granules of endocrine and chief cells. [6] Release of leptin into the systemic circulation and into gastric juice is stimulated by intravenous infusions of pentagastrin or secretin. Leptin receptor is also present in gastric epithelium. These findings raise the possibility that leptin produced in the stomach plays a paracrine, endocrine, and/or exocrine function in energy homeostasis.

Leptin is known to affect the gene expression of other neurohormones involved in energy balance (Fig. 36-1). [7] Neuropeptide Y (NPY) stimulates food intake and decreases energy expenditure. The expression of messenger RNA for NPY is inhibited by leptin. Conversely, leptin stimulates the gene expression of peptides that have opposite characteristics. a-Melanocyte-stimulating hormone (a-MSH), a peptide derived from proopiomelanocortin (POMC), and cocaine- and amphetamine-regulated transcript (CART) are hypothalamic peptides that are positively regulated by leptin and produce anorexia.

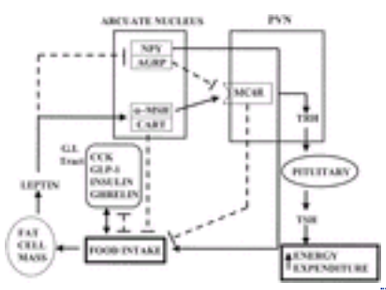

FIGURE 36-1. Interaction of leptin and hypothalamic neuropeptides in controlling energy homeostasis. Food intake increases fat cell mass, which results in an

elevation of circulating leptin levels. Leptin stimulates the synthesis of cocaine- and amphetamine-regulated transcript (*CART*) and the production of a-melanocyte-stimulating hormone (*a-MSH*) in the arcuate nucleus of the hypothalamus, which decreases food intake via melanocortin-4 receptor (*MC4R*) signaling in the paraventricular nucleus (*PVN*). Activation of MC4R also increases energy expenditure via signaling through thyrotropin-releasing hormone (*TRH*) and thyrotropin (*TSH*). Leptin inhibits the production of agouti-related protein (*AGRP*), which allows for more effective signaling of a-MSH at MC4R. Leptin also inhibits the synthesis of neuropeptide Y (*NPY*). Thus, the result of increased leptin levels is stimulation of anorectic (CART and a-MSH) and inhibition of anabolic (AGRP and NPY) signaling pathways to decrease food intake and increase energy expenditure. Gastrointestinal peptides also modulate food intake through peripheral and central pathways. (*Arrows*, stimulatory; *dashed lines*, inhibitory.) *CCK*, cholecystokinin; *GLP-1*, glucagon-like peptide-1.

The melanocortin system, in particular, is under intensive investigation because of strong evidence in both rodents and humans of its control of energy homeostasis. [2] There are five different receptors for a-MSH, two of which, melanocortin-3 receptor (MC3R) and melanocortin-4 receptor (MC4R), are primarily expressed in the brain. Some of the metabolic effects resulting from the stimulation of MC4R are decreased food intake, stimulation of thyrotropin-releasing hormone (TRH), and an increase in energy expenditure. Mutations in MC4R are found in approximately 1% to 5% of patients with a BMI above 40. In addition, rare mutations have been reported in the genes for POMC and the enzyme that processes the POMC protein to a-MSH that are associated with severe childhood obesity. Further evidence for the importance of the melanocortin pathway stems from the identification of agouti-related protein (AGRP). AGRP is a hypothalamic neuropeptide that stimulates food intake in the rat through antagonism of the interaction of a-MSH at MC4R (see Fig. 36-1); expression of the AGRP gene is suppressed by leptin.

Gastrointestinal tissues also mediate important communication to the brain about energy intake through neural signaling and endocrine pathways. Cholecystokinin (CCK), which is released in response to dietary fat, enhances nutrient absorption by slowing gastric emptying and stimulating gallbladder contraction, and inhibits food intake during a meal via the afferent vagal system. [8] Glucagon-like peptide-1 (GLP-1) inhibits gastrointestinal motility, hunger, and food intake. [9] Ghrelin is a peptide found in the hypothalamus and stomach. [10] Intracerebroventricular injection of ghrelin stimulates feeding in the rat and increases body weight through stimulation of NPY and AGRP neurons. Apolipoprotein A-IV is a glycoprotein synthesized by the human intestine in response to fat absorption. [11] Apo A-IV causes central inhibition of food intake and is likely to be involved in the short-term and possibly long-term regulation of food intake. The role of these peptides in the regulation of body weight in humans is under intense study and has important implications for the design of therapeutic agents for obesity.

COMORBIDITIES OF OBESITY

Gastrointestinal Dysfunction

Most large epidemiologic studies have found that symptoms of gastroesophageal reflux disease (GERD) are more common in obese than in lean persons. In a prospective study of 1224 patients referred for upper gastrointestinal endoscopy, reflux esophagitis was found in 16% of the patients and hiatus hernia in 20%. [12] A coexisting hiatus hernia was found in 68% of the patients with reflux esophagitis. Overweight was most pronounced in those with mild to moderate esophagitis, whereas in patients with severe esophagitis, the body weight was normal. These results support the view that adiposity is associated with both sliding hiatus hernia and reflux esophagitis, and that hiatus hernia may play a role in the development of GERD. In contrast, a study of morbidly obese subjects found no significant correlation between body weight, BMI, or waist-hip circumference and reflux. [13]

Although increased intra-abdominal pressure induced by excessive abdominal girth may predispose obese persons to reflux, we are unaware of any randomized controlled trials that have evaluated whether weight loss decreases reflux symptoms. In one study, conducted in lean persons, diet-induced weight loss correlated directly with a decrease in reflux symptoms. [14] However, alterations in diet, rather than a decrease in body weight, may have been responsible for the beneficial effects because even a modest weight loss of 2 to 3 kg was associated with a marked improvement in the symptom score. In studies of patients with class I [15] or class III [16] obesity who had symptoms of GERD, diet-induced weight loss did not relieve symptoms or improve 24-hour esophageal pH values. In contrast, the gastric bypass procedure has consistently been shown to decrease GERD symptoms. [17], [18] In fact, GERD symptoms often resolve immediately after surgery before significant weight loss occurs, suggesting that the elimination of acid or bile reflux, rather than a decrease in weight, is responsible for the beneficial effect. Although some studies have found that vertical banded gastroplasty and gastric banding, which increase resistance to flow through the proximal pouch, do not alter the lower esophageal sphincter pressure or increase episodes of reflux, [19] severe gastroesophageal reflux can occur after vertical banded gastroplasty. [20]

Studies of the relationship between gastric emptying and obesity have yielded conflicting results. Gastric emptying has been reported as accelerated, delayed, or unchanged in the obese. [21] Rapid gastric emptying may result in decreased satiety signals from the intestine and may be a contributing factor in the pathogenesis of obesity.

Gallbladder Disease

Obesity, particularly in women, is an important risk factor for gallbladder disease. [22] Data from the Nurses Health Study demonstrated that the risk for symptomatic gallstones increases linearly with the BMI. Obese women (BMI >30 kg/m [2]) had a twofold and extremely obese women (BMI >45 kg/m [2]) a sevenfold excess risk for symptomatic gallstones in comparison with lean women. [23] The risk for gallstones also increases during weight loss because of increased bile cholesterol supersaturation, enhanced cholesterol crystal nucleation, and decreased gallbladder contractility. The incidence of new gallstones is approximately 30% in obese patients who experience rapid weight loss after treatment with a very-low-calorie diet (<600 kcal/d) or gastric surgery. [24], [25] and [26] Ursodeoxycholic acid therapy at a dose of 600 mg/d provides maximal gallstone prevention. Prophylactic therapy should be considered for patients who are expected to achieve rapid weight loss exceeding 1.5 kg or 1.5% of body weight per week.

Pancreatitis

It is not known whether obese patients are at increased risk for gallstone pancreatitis. However, most studies have found that overweight and obese patients are at higher risk for the development of local complications and severe pancreatitis. [27], [28] Obesity also increases the risk for respiratory insufficiency and mortality. It is possible that excess fat deposited in the peripancreatic and retroperitoneal spaces predisposes obese patients to peripancreatic fat necrosis and subsequent local and systemic complications.

Liver Disease

Obesity is associated with a spectrum of liver abnormalities that can be classified by alterations in liver histology (macrovesicular steatosis, steatohepatitis, fibrosis, and cirrhosis) and are now known collectively as *nonalcoholic fatty liver disease (NAFLD)*. [29] Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) are the liver enzymes most commonly elevated in obese patients, but enzyme levels usually do not exceed two times the upper limit of normal and often do not correlate with the severity of the histological abnormalities. Dieting itself often causes a transient increase in serum transaminase concentrations and a decrease in serum alkaline phosphatase concentration during early weight loss. In a histological analysis of liver biopsy specimens from overweight and obese patients who had abnormal liver biochemistries but no overt findings of liver disease or evidence of acquired, autoimmune, or genetic liver disease, 20% of the patients had septal fibrosis alone and 10% had septal fibrosis and “silent” cirrhosis. [30] In a review of the data from a series of studies, 40% to 100% of the patients with nonalcoholic steatohepatitis were obese. [31] Data from autopsy studies, investigations of obese patients undergoing obesity surgery, and cross-sectional analyses of liver biopsy samples suggest that steatosis occurs in about 75%, steatohepatitis in about 20%, and cirrhosis in about 2% of obese patients. [32], [33] and [34]

The pathogenesis and natural history of NAFLD are not well understood. Most patients are asymptomatic, but some may have fatigue, malaise, and vague abdominal discomfort. Hepatomegaly has been reported in up to three fourths of patients with NAFLD. In contrast to the ratio of AST to ALT in patients who have alcohol-induced steatohepatitis, this ratio is usually less than 1 in patients who have NAFLD. In studies that followed patients for up to 7 years, liver disease continued to progress in about 40% of patients, and cirrhosis developed in about 10%. In most patients with simple steatosis, the clinical course was benign, whereas in those with steatohepatitis, fibrosis, and cirrhosis, clinical sequelae of severe liver disease were more likely to develop. Furthermore, obesity increased the risk for fibrosis and cirrhosis in patients with alcoholic liver disease [35] and hepatitis C. [36]

Although weight loss is often recommended as therapy for obese patients with NAFLD, it is not known whether weight loss affects long-term outcome and reduces the risk for progressive disease. A gradual loss of 10% or more of body weight can correct abnormal liver chemistries and decrease liver size, fat content, and features of

steatohepatitis. However, rapid weight loss after gastric surgery, very-low-calorie diets, or fasting decreases the hepatic fat content but can induce hepatic inflammation and exacerbate steatohepatitis. The use of metformin has been shown to reverse fatty liver disease in insulin-resistant obese mice, but this potential therapeutic approach has not been evaluated in humans. [37]

Colon Cancer

Overweight and obesity are associated with an increased risk for esophageal, gallbladder, and colorectal cancers. [38] Men whose BMI is above the fourth quintile have an odds ratio of 1.96 for the development of colon cancer, and women with a BMI above the fourth quintile have an odds ratio of 1.54. [39] The effect of the BMI is strongest for distal cancers and in patients with a family history of colorectal cancer.

Dyslipidemia and Cardiovascular Disease

Obesity is an independent risk factor for cardiovascular disease, including coronary heart disease, congestive heart failure, stroke, and peripheral vascular disease. Increased body weight and abdominal obesity predispose individuals to elevated levels of total cholesterol, low-density lipoprotein (LDL) cholesterol, triglycerides, apolipoprotein B (a marker for small, dense LDL), insulin, and plasminogen activator-1 and to low levels of high-density lipoprotein (HDL) cholesterol. [40] The cluster of hypercholesterolemia, hypertriglyceridemia, hyperuricemia, impaired glucose tolerance, insulin resistance, diabetes, and hypertension in various combinations is known as the *metabolic syndrome* or *syndrome X* and is commonly associated with abdominal fat accumulation. Most studies have found that weight loss decreases serum triglyceride, total cholesterol, and LDL cholesterol concentrations and increases the serum HDL cholesterol concentration. [41] At 2 years, a sustained weight loss of 5% is sufficient to maintain the reduction in serum triglyceride concentrations, whereas serum total and LDL cholesterol levels revert toward baseline if at least a 10% weight loss is not maintained. [42]

Data from the Third National Health and Nutrition Examination Survey (NHANES III) demonstrated that the age-adjusted prevalence of hypertension is more than twofold higher in obese men and women (42% and 38% prevalence rates, respectively) than in lean men and women (~15% prevalence rate in both men an women). Weight loss results in a reduction of systolic and diastolic blood pressure; however, a study of patients after gastric surgery has raised questions about the duration of this effect. [43]

Type 2 Diabetes Mellitus

Body mass index, abdominal fat distribution, and weight gain are important risk factors for type 2 diabetes mellitus. Data from NHANES III showed that the prevalence of diabetes increases from 2% in those with a BMI between 25 and 29.9 kg/m ^{2} to 13% in those with a BMI of 35 kg/m ^{2} or higher. The Nurses Health Study found that the risk of diabetes begins to increase even in women of “normal” weight when the BMI exceeds 22 kg/m ^{2}. Weight loss of 5% improves glycemic control in most obese diabetic patients. In addition, sustained weight loss is effective in preventing the development of new cases of diabetes in obese persons.

HISTORY AND PHYSICAL EXAMINATION

The initial evaluation of an obese patient starting a weight loss regimen includes a history, physical examination, and laboratory studies. A history of eating disorders or purging is a relative contraindication to treatment, and referral to a specialist in these areas is appropriate. The patient’s current level of physical activity and understanding of nutrition should be assessed. Diseases associated with weight gain, such as polycystic ovarian disease, hypothyroidism, and Cushing syndrome, should be ruled out or treated. In addition to the gastrointestinal complications of obesity, the physician should search for other complications, such as hypertension, type 2 diabetes, hyperlipidemia, coronary heart disease, osteoarthritis of the lower extremities, gout, and certain cancers. Depression, and the drugs used to treat depression, may cause weight gain. The physician should be aware of the symptoms and signs of sleep apnea: loud snoring, brief awakenings, sleeping in the sitting position, daytime fatigue, morning headaches, or polycythemia on the laboratory evaluation. If signs of sleep apnea are present, the patient should be referred to a pulmonologist or sleep specialist. Assessment of the patient should include a calculation of the BMI. The distribution of adipose tissue is an important consideration in addition to the BMI. An abdominal distribution of adipose tissue is characterized by an increase in waist circumference, which is measured by placing a measuring tape in a horizontal plane at the level of the iliac crest without compressing the skin. A waist circumference greater than 35 inches (88 cm) in women and 40 inches (102 cm) in men is considered an independent risk factor for type 2 diabetes and cardiovascular disease (see Table 36-1).

TREATMENT

The primary target of treatment should be those individuals at medical risk because of their weight (Table 36-2). A realistic treatment goal is usually a loss of 5% to 10% of the initial body weight over 6 to 12 months followed by long-term maintenance of the reduced weight. Most cardiovascular risk factors are decreased even at this modest level of weight reduction [1]; however, patients with cholelithiasis and osteoporosis must be warned that these conditions may be aggravated by weight loss. A summary of the steps in evaluating and treating the overweight and obese patient is presented in Table 36-3. *The Practical Guide. Identification, Evaluation, and Treatment of Overweight and Obesity in Adults* [44] is a useful source that details the appropriate management of the overweight and obese patient.

TABLE 36-2 Treatment Options

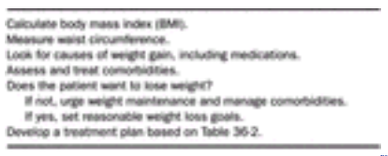

TABLE 36-3 Steps in Evaluating and Treating the Overweight and Obese Patient

Lifestyle Modifications

Calorie-Restricted Diets To induce weight loss, a calorie deficit must be achieved. General guidelines for healthful eating, such as a reduction in fat, sugar, and alcohol intake, should be emphasized. Balanced diets should contain a minimum of 1000 to 1200 kcal/d and provide adequate amounts of all essential nutrients. A diet that is individually planned to create a deficit of 500 kcal/d should produce a weight loss of about 1 lb weekly. The step I diet, consisting of approximately 55% carbohydrate, 15% protein, 30% fat (8%–10% saturated fatty acids, <300 mg of cholesterol per day), and 2.4 g of sodium daily, will also decrease other risk factors, such as high blood cholesterol and hypertension. Water intake should be at least six to eight 8-oz glasses per day. Supplementation with a multivitamin can be considered. A calcium intake of 1000 to 1500 mg/d is particularly important for individuals at risk for osteoporosis. Calories derived from fat are easier to consume in excess because they are “denser” (9 cal/g versus 4 cal/g for protein and carbohydrate) and easily hidden in foods, and they increase food palatability. In general, energy consumed as fat causes less satiety than energy consumed as carbohydrate. Fat-modified foods lower the total fat intake but are an aid to weight loss only if they are also low in calories. Studies suggest that because individuals tend to consume a constant weight of food, a diet low in energy density is likely to lead to a reduction in energy intake. [45] A diet low in energy density is usually low in fat and high in water content, complex carbohydrates, and fiber. Fruits, vegetables, and grain products are foods low in energy density. However, foods high in energy density are not always high in fat content. For example, dry foods such as fat-free pretzels or crackers are high in energy density. The glycemic index of a food, defined as the area under the glucose response curve after consumption of 50 g of

carbohydrate from a test food divided by the area under the curve after consumption of 50 g of carbohydrate from a control food such as white bread or glucose, has been proposed as an important factor in regulating energy intake. [46] Starchy foods such as refined grain products and potatoes generally have a high glycemic index, whereas vegetables, legumes, and some fruits generally have a low glycemic index. In one study, subjects given a diet with a low glycemic index in the morning consumed less food later in the day in comparison with subjects given a diet with a high glycemic index. [47] Although the glycemic index requires further study before it is universally accepted by nutrition professionals, in our opinion, it does play a role in individual dietary prescription.

Exercise Physical activity is a key component of any long-term weight maintenance program. A moderate amount of physical activity uses approximately 150 calories of energy per day. Some examples of moderate activity include walking 2 miles, pushing a stroller 11/2 miles in 30 minutes, bicycling 5 miles in 30 minutes, and swimming laps for 20 minutes. [44] Patients who are physically limited by obesity or arthritis may start with water exercises, bedside stretching, or seated activities. In general, patients should build up to at least a 30- to 45-minute exercise regimen, daily if possible, and the exercise should be of the greatest intensity that is safe for their own level of fitness.

Behavior Therapy

The goal of behavior therapy is to overcome barriers to compliance with a regimen of diet and physical activity. Behavior therapy may consist of self-monitoring, stimulus control, reinforcement, and stress management. *Self-monitoring* includes recording dietary intake (food choices, amounts, times), exercise, and changes in body weight. *Stimulus control* helps the patient to identify and change cues associated with eating too much and exercising too little. *Reinforcement* encourages the attainment of goals that are difficult to achieve by providing rewards other than food when goals are reached. *Stress management* helps the patient cope with stressful events by developing outlets other than eating for reducing stress. Evaluating setbacks and determining how to do better next time can break the chain of negative thinking and self-punishment when lapses occur.

Medications that May Cause Weight Gain and Alternatives

Many commonly prescribed medications are known to cause weight gain as a side effect (Table 36-4). When antiobesity medications are being considered, attention should be paid to the use of such medications and the possibility of switching the patient to others that may induce weight loss. Metformin is often the best first-line drug in the obese diabetic patient because it may induce weight loss and improve the body composition and lipid profile. Acarbose and miglitol, which are a-glucosidase inhibitors, may also induce weight loss. Most mood stabilizers and antidepressants, particularly the tricyclics, cause weight gain, although bupropion may induce weight loss. An anticonvulsant, topiramate, has been shown to induce weight loss, in contrast to the weight gain often associated with other agents, and case reports have appeared of its being used to mitigate the weight gain associated with the new antipsychotic agents, such as olanzipine.

Antidepressants, lithium
Neuroleptics
Insulin, sulfonylureas, thiazolidinediones
Antiepileptics
Antihistamines
Corticosteroids
Hormonal contraceptives
Progestational steroids
β Adrenergic blockers
Phenothiazines

Adapted from ref. 59.

TABLE 36-4 Drugs that May Promote Weight Gain

Pharmacotherapy

The National Heart, Lung, and Blood Institute supports the use of pharmacotherapy for patients who have not achieved weight loss and maintenance goals by means of initial therapy with lifestyle modification (see Table 36-2). Two agents are currently approved by the U.S. Food and Drug Administration for long-term use, sibutramine and orlistat (Table 36-5).

DRUG	DOSE	ACTION
Sibutramine	• 5, 10, 15 mg • 10 mg orally once a day to start • May be increased to 15 mg or decreased to 5 mg	• Inhibits norepinephrine, dopamine, and serotonin reuptake • Reduces appetite and increases thermogenesis
Orlistat	• 120 mg orally with meals	• Inhibits intestinal lipases • Decreases fat absorption

From ref. 44.

TABLE 36-5 Characteristics of Pharmacotherapies Approved for Long-Term Use in the Treatment of Obesity

Sibutramine Sibutramine has been shown to achieve and maintain weight loss for as long as 2 years. [48] The therapeutic effect of sibutramine is based on inhibition of the reuptake of norepinephrine, serotonin, and dopamine and activation of the respective neurotransmitter receptors, which results in decreased food intake. [49] In addition, sibutramine may attenuate the decrease in resting metabolic rate that accompanies caloric restriction and weight loss. [50], [51] In a 1-year placebo-controlled trial, 65% of the patients who received 15 mg of sibutramine daily lost more than 5% of their body weight, versus 29% of the patients who took placebo, and 39% lost more than 10% of their body weight, versus 8% who achieved the same mean weight loss in the placebo-treated group. [52] Patients who lose at least 4 lb in the first 4 weeks of treatment are more likely to achieve a weight loss of at least 5% within 6 months. [53] The most common adverse effects reported in clinical studies of sibutramine have been headache, dry mouth, constipation, and insomnia. [52], [53] and [54] Small mean increases in both diastolic and systolic blood pressure (2 mm Hg at doses of 10 and 15 mg) and pulse rate (3 to 6 beats/min) are associated with sibutramine treatment and occur most frequently during the first 8 weeks of treatment. Routine clinical monitoring of vital signs is therefore recommended.

Orlistat Orlistat binds to gastric, pancreatic, and carboxyl ester lipases in the gastrointestinal tract and blocks the action of these lipases on dietary triglycerides and vitamin esters. The inhibition of fat digestion decreases micelle formation and the absorption of long-chain fatty acids, cholesterol, and certain fat-soluble vitamins. The percentage of malabsorbed fat is directly related to the drug dose in a curvilinear fashion. At a dose of 360 mg/d (120 mg three times daily with meals), approximately 30% of ingested triglyceride is excreted in stool, which is near the maximum plateau value. [55], [56] Several prospective randomized controlled trials have evaluated the efficacy of orlistat therapy in initiating and maintaining weight loss. [43] In one study, after 2 years, 57.1% of orlistat-treated patients maintained a loss of more than 5% of body weight, compared with 37.4% of placebo-treated patients. [55] The most common side effects of orlistat are related to its action on gastrointestinal lipases: abdominal pain, fecal urgency, liquid stools, flatulence with discharge, oily spotting, and incontinence. Long-term treatment with orlistat can affect the homeostasis of certain fat-soluble vitamins. During 1- and 2-year clinical trials of orlistat, vitamin concentrations, usually of vitamins D and E and ß-carotene, fell below normal limits in approximately 5% more orlistat- than placebo-treated subjects. These abnormalities resolved rapidly with vitamin supplementation. It is recommended that all patients who are treated with orlistat be given a daily multivitamin supplement that is taken at a time when orlistat is not being ingested. Orlistat can have medically significant effects on the absorption of lipophilic medications if both drugs are taken simultaneously. Concomitant use of orlistat in a patient taking cyclosporine or warfarin requires careful monitoring of the appropriate parameters. A theoretical concern is that long-term orlistat therapy may increase the risk for specific gastrointestinal diseases, such as gallstones and colon cancer. However, an increased incidence of these diseases has not been observed in subjects who have completed 1- and 2-year clinical trials.

Drugs Approved for Short-Term Use Noradrenergic agents, such as phentermine and diethylpropion, have been shown to be better than placebo in relatively small-scale, short-term studies. No large-scale, long-term studies of the effects of these compounds on weight loss, their health benefits, or their side effects have been performed, although they have been available for more than 30 years. Therefore, if these drugs are used for longer than 3 months, the patient should be informed that such use is “off label” and has not been studied.

Over-the-Counter Products A myriad of products are available and are marketed as “nutritional supplements.” Some, like chromium picolinate, chitosan, L-carnitine, and hydroxycitric acid, have no proven efficacy to recommend their use. Others, like the combination of ephedrine and caffeine often seen in “fat-burning products,” have been shown to be effective but are not safe for unsupervised use because of the risk for tachycardia and hypertension.

Surgery for Weight Management

Obesity surgery should be considered for patients with a BMI above 40 or with a BMI between 35 and 40 if they have failed other methods of treatment and serious obesity-related complications are present. [1], [57] The overall mortality rate for bariatric surgery is approximately 1% but may be higher in individuals with comorbidities. Operative complications such as wound dehiscence, anastomotic leak, stomal stenosis, and thrombophlebitis occur in about 10% of patients. [56] Gastroplasty, also known as *stomach stapling,* involves constructing a 15- to 30-mL pouch along the lesser curvature of the stomach. A Silastic ring or band of polypropylene is used to restrict the outlet size. A modification of this procedure involves the use of an adjustable band that wraps around the proximal stomach to create a small pouch. The outlet of the pouch can be adjusted percutaneously by injecting saline solution into a reservoir, which inflates the band and restricts the size of the outlet. Patients who undergo these procedures feel full after eating very small amounts of food and may vomit if they continue to eat. In general, 70% of patients maintain a loss of 20% or more of total body weight during 5-year follow-up. Complications of laparoscopic gastric banding include stomal stenosis, staple line disruption, increased gastroesophageal reflux, band slippage, esophageal dilatation, erosion of the band into the stomach, infection, and leaks leading to band deflation.

The gastric bypass procedure, also known as *Roux-en-Y gastric bypass,* is the most commonly performed procedure. It involves constructing a small proximal gastric pouch that empties into a segment of jejunum brought up to the gastric pouch as a Roux-en-Y limb. Increasing the length of the limb from 75 to 150 cm increases weight loss but also long-term side effects and nutrient malabsorption. More weight is lost with the gastric bypass procedure than with gastroplasty. Complications of the gastric bypass procedure include stomal stenosis, marginal ulcers, staple line disruption, dilation of the bypassed stomach, internal hernias, dumping syndrome, and malabsorption of nutrients. [43] Supplemental calcium to prevent osteoporosis and a multivitamin containing vitamin B_{12} and folate are recommended daily. Iron supplementation, occasionally requiring intravenous administration because of malabsorption, may be necessary for menstruating women. Magnesium and zinc supplements are required in some individuals. Although technically challenging, laparoscopic gastric bypass surgery is now being performed. Stenosis of the gastric outlet, requiring dilation, and leaks are more common than after the open procedure, but postoperative recuperation and the incidence of abdominal hernia are reduced.

Partial biliopancreatic bypass with duodenal switch is occasionally performed in the most obese patients. This procedure allows the consumption of normal amounts of food and is more effective than the gastric bypass procedure, producing up to 75% weight loss, but it is associated with a greater number of malabsorptive and gastrointestinal side effects.

CONCLUSION

Obesity is a common chronic disorder associated with multiple gastrointestinal comorbidities. Current nonsurgical treatments for obesity produce mean losses of 5% to 10% of body weight, enough to relieve many of these comorbidities. Diet, exercise, and behavior therapy form the cornerstone of treatment for obesity. In certain individuals, the addition of pharmacotherapy or surgery is indicated if a more conservative approach has failed.

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CHAPTER 37

William L. Hasler

APPROACH TO THE PATIENT WITH NAUSEA AND VOMITING

Nausea and vomiting are nonspecific symptomatic responses to a variety of conditions. *Nausea* is the subjective sensation of an impending urge to vomit, usually perceived in the throat or epigastrium. *Vomiting* is the forceful ejection of upper gut contents from the mouth. Although usually preceded by nausea, vomiting may occur without nausea. *Retching* may precede vomiting but involves no discharge of gastric contents from the mouth.

Nausea and vomiting should be distinguished from other symptoms. *Regurgitation* is the effortless return of gut contents into the mouth without nausea or somatic muscle contractions. *Rumination* (*merycism*) is regurgitation with rechewing and reswallowing of food, often multiple times after a meal. *Anorexia* is a loss of appetite, which may or may not be associated with nausea. *Early satiety* is a sensation of gastric fullness before meal completion. Nausea may be part of the general complaint of *indigestion,* which includes discomfort, heartburn, anorexia, and bloating.

SOCIOECONOMIC IMPACT

Nausea and vomiting have significant a socioeconomic impact on patients' lives. Most patients with nausea resulting from acute enteric illness restrict their activities. [1] Pregnant women with first-trimester nausea and vomiting report increased fatigue, sleep disturbances, and irritability, which impair family and social interactions. [2], [3] and [4] Nausea and vomiting after cancer chemotherapy and in the postoperative period reduce the time spent on leisure activities and recreation, household tasks, and socializing and increase the level of hardship to family members. [5], [6] and [7] Chemotherapy-evoked nausea restricts functioning more than the associated vomiting. [5] When queried, 32% of patients scheduled for general anesthesia said they would pay extra money out of pocket to avoid postoperative nausea and vomiting. [8]

Patients, employers, and the health care industry incur significant expenses as a consequence of nausea and vomiting. Acute enteric infection reportedly increased medical expenses by $1.25 billion and cost $21.8 billion in lost productivity in the United States in 1980. [9] Two British studies estimated that 8.5 million working days per year are lost because of nausea of pregnancy, and that severely affected women miss a mean of 62 hours of work while pregnant. [10], [11] Nausea and vomiting after chemotherapy decrease employee productivity and increase the health care costs of prolonged hospitalization and home nursing time. [12] Postoperative nausea and vomiting cost outpatient surgical centers a mean of $415 per patient. [13] Another British study calculated that nausea and vomiting after surgery prevent the performance of 96 to 576 surgical procedures per year per center as a consequence of increased recovery room stays and extra nursing effort. [14] In anorectal surgery, recovery room costs are lower with local anesthesia plus intravenous sedation than with general anesthesia because of a reduced incidence of postoperative nausea and vomiting. [15]

Despite the pronounced socioeconomic effects of these sometimes disabling symptoms, available evidence suggests that the impact of current therapies on health-related and monetary consequences of nausea and vomiting is inadequate. In a survey of 1413 outpatients receiving chemotherapy, the routine use of potent serotonin receptor antagonists produced a downward trend in the frequency of vomiting. [16] However, the average duration of posttreatment nausea increased significantly. A second study of 600 patients receiving chemotherapy also reported increases in nausea despite the widespread use of newer antiemetic agents, emphasizing the need for ongoing research into novel agents to treat severe nausea and vomiting. [17]

PATHOPHYSIOLOGY

Vomiting results from a coordinated interaction of neural, humoral, somatic muscular, and gastrointestinal myoelectric and muscular phenomena. What is known about the pathways mediating vomiting comes from studies involving the ablation of neural structures in experimental animals. The mechanisms that result in nausea are less well understood. Whereas vomiting may be generated in decerebrate animals, nausea requires the activation of selected cerebral cortical sites.

Activation of the Emetic Response

Vomiting may be initiated by stimuli acting on structures in the central nervous system and peripherally (Fig. 37-1). The area postrema on the dorsal surface of the medulla at the caudal aspect of the fourth ventricle is believed to represent the "chemoreceptor trigger zone" and is responsive to a broad range of neurochemical activators. [18] Other central nervous system and peripheral afferent sites mediate emesis after other stimuli.

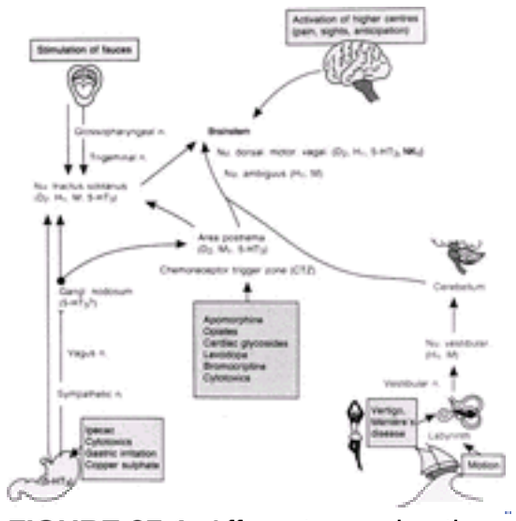

FIGURE 37-1. Afferent neural pathways and receptor populations involved in vomiting, as determined by experimental animal studies. A series of brainstem structures coordinates vomiting after activation of cortical, oral, vestibular, or peripheral afferents of the chemoreceptor trigger zone (area postrema). (Adapted from Mitchelson F. Pharmacological agents affecting emesis. A review [part I]. Drugs 1992;43:295.)

Area Postrema as a Receptor of Neurochemical Emetic Stimuli Because of its location and lack of a tight blood-brain barrier, the area postrema is an interface to receive input from the brain and peripheral nerves and to sample chemical activators in the bloodstream and cerebrospinal fluid. [19] Several observations confirm the role of the area postrema as the site of action for many stimuli. The dopamine receptor agonist apomorphine evokes vomiting when administered systemically or directly to the area postrema, whereas ablation of the structure prevents apomorphine-induced emesis. [18] Drugs (digoxin, opiates, nicotine, ergot alkaloids, some cancer chemotherapies), metabolic disorders (uremia, diabetic ketoacidosis, hypoxemia, hypercalcemia), radiation therapy, and bacterial toxins induce vomiting by area postrema activation. [20], [21] Neurotransmitters localized to the area postrema include dopamine, serotonin, norepinephrine, enkephalins, ?-aminobutyric acid, and substance P. Receptor subtypes characterized in the structure include dopamine D_2, muscarinic M_1, histaminergic H_1, serotoninergic 5-HT_3, and vasopressinergic subtypes.

Other Central Neural Activators of Emesis The central nervous system is the source of emesis in certain settings. Cerebral pathways mediate vomiting from noxious odors, visual stimulation, somatic pain, and unpleasant tastes. [21] Brainstem vestibular nuclei activate emesis in motion sickness, Ménière disease, and labyrinthine tumors or infections via area postrema–independent pathways. [22] Muscarinic M_1 and histaminergic H_1 pathways play important roles in motion sickness. [23] Motion stimuli increase hypothalamic and brainstem histamine levels, and irrigation of the ear with cold or hot water evokes hypothalamic histamine release. [23], [24] It is postulated that motion stimuli activate H [1] receptors in the brain and brainstem that directly coordinate the somatic and gastrointestinal events of emesis. [23] Anticholinergic agents binding to neural M_1 receptors may prevent motion sickness by enhancing habituation responses to motion stimuli. [23]

Peripheral Afferent Neural Activation of Emesis Gastric irritants such as copper sulfate, salicylate, *Staphylococcus* enterotoxin, and antral distention activate afferent nerves projecting from the stomach. [25], [26] and [27] Nongastric intraperitoneal stimuli that evoke emesis include distention of the colon, small intestine, and bile ducts, peritoneal inflammation, and mesenteric vascular occlusion. Stimuli also may originate from the pharynx and heart. These observations explain emesis induced by peritonitis, mesenteric ischemia, and myocardial infarction. Vomiting induced by hypertonic saline solution or intragastric copper sulfate is mediated by the activation of gastroduodenal mucosal afferent fibers, whereas vomiting with gastric distention involves the activation of smooth muscle afferent fibers. [27], [28] Emetic responses to many peripheral stimuli are mediated by vagal pathways. Vagotomy prevents emesis after intragastric copper sulfate and reduces vomiting provoked by the chemotherapeutic agent cisplatinum and by abdominal irradiation. [25], [29], [30] and [31] Splanchnic afferent pathways may modulate emesis after selected peripheral stimuli. [32] Furthermore, there is evidence of plasticity in peripheral afferent neural pathways that activate emesis. [33] In cats, the threshold for vomiting induced by a selective 5-HT_3 receptor agonist is increased by acute vagotomy plus splanchnicectomy. [33] However, if vagotomy is performed 14 to 51 days after splanchnicectomy, the thresholds for induction of emesis are reduced, indicating the recruitment of new neural pathways. Most afferent vagal fibers project to the brainstem nucleus tractus solitarius, although some fibers project to the area postrema or to the dorsal vagal motor nucleus. [34] The nucleus tractus solitarius is believed to initiate the cascade of events associated with emesis. [35] Ablation studies demonstrate that many peripheral stimuli act independently of the area postrema. [18] Serotoninergic pathways play pivotal roles in mediating the effects of many peripheral stimuli. Serotonin synthesized by enterochromaffin cells or myenteric neurons is believed to act on 5-HT_3 receptors on vagal afferent fibers and in the area postrema and nucleus tractus solitarius. [36] 5-HT_3 antagonists reduce the vomiting evoked by cisplatinum and copper sulfate, indicating the importance of 5-HT_3 receptors in mediating vomiting evoked by peripheral stimulation. [36] In some species, activation of the 5-HT_4 subtype may be more important for the emetic response to peripheral stimulation. [37]

Central Neural Coordination of the Act of Vomiting

Multiple brainstem sites mediate the act of vomiting in response to emetic stimuli from the activator sites described above. Immunohistochemical studies show increased expression of c-fos protein in the area postrema, the nucleus tractus solitarius, the dorsal vagal and phrenic nuclei, the medullary nuclei involved in control of respiration, the hypothalamus, and the amygdala after emetic stimulation. [38] The nucleus tractus solitarius receives projections from the area postrema, abdominal vagus, and labyrinths and has been postulated to serve as a common focus for initiating the vagus-dependent gastrointestinal motor and myoelectric aspects of emesis. [34], [35]

The nucleus tractus solitarius also projects to the Botzinger complex in the region of the medullary retrofacial nucleus, which regulates respiration; the nucleus ambiguus, which supplies the laryngeal and pharyngeal musculature; and the trigeminal, facial, and hypoglossal nuclei, which serve the facial and tongue muscles. [39] Neurons of the medullary reticular formation interact with the nucleus ambiguus, the dorsal motor nucleus of the vagus, and the respiratory nuclei. [40] Ablation of the reticular formation prevents emesis in dogs, confirming the importance of this structure. [41] Evidence has shown that nonrespiratory neurons in the reticular area drive respiratory premotor neurons in the ventral respiratory group. [42] During retching, firing rates in pre–Botzinger complex inspiratory neurons nearly disappear, establishing the interrelation of respiratory and nonrespiratory pathways. [43] These findings are consistent with the postulate that output from the medullary reticular formation with modulatory output from the nucleus tractus solitarius coordinates neuronal firing in each of these nuclei to regulate the somatic muscular components of emesis. [44] Once activated, these brainstem structures provide stimulation to the diaphragm via cervical spinal pathways and to the external musculature via the thoracolumbar spinal nerves. The nucleus tractus solitarius also sends ascending projections into the hypothalamus and limbic regions, suggesting activation of cerebral regions regulating the behavioral aspects of emesis. [45]

Investigations also suggest a prominent role for brainstem tachykinin NK_1 pathways in mediating emesis. The most prevalent tachykinin, substance P, induces vomiting when injected into the brainstem. [46] Peripheral administration of NK_1 antagonists block emetic responses and prevent the retching burst firing patterns in brainstem neurons in response to emetic stimulation. [47], [48], [49] and [50] Furthermore, injections of selective NK_1 receptor antagonists into the area postrema, medial solitary nucleus, and dorsal vagal complex also prevent vomiting. [51], [52] and [53] Other transmitters likely modulate the emetic response. The nucleus tractus solitarius and vagal nuclei are rich in catecholamines, enkephalins, somatostatin, vasopressin, and thyrotropin-releasing hormone. Furthermore, selective serotonin 5-HT_{1A} receptor agonists block vomiting evoked by motion sickness, nicotine, cisplatinum, and copper sulfate, suggesting a wide ranging role for 5-HT_{1A} receptors in the emetic response. [54]

Peripheral Responses to the Initiation of Vomiting

Somatic Muscular Events Associated with Vomiting Emesis results from a stereotypical series of somatic muscle actions. During retching, inspiratory thoracic, diaphragmatic, and abdominal muscles contract concurrently against a closed glottis. [55] The resulting high positive intra-abdominal pressure forces gastric contents into the esophagus and herniates the gastric cardia into the thorax. [56] A high negative intrathoracic pressure prevents expulsion of lumenal contents into the mouth, even though the upper esophageal sphincter may relax during retching. [57] In contrast to the pressures during retching, both the intra-abdominal and intrathoracic pressures are positive during vomiting because of a lack of diaphragmatic contraction in the crural region that permits transmission of the high positive abdominal pressure into the thorax. [58] Synchronous contractions of the inspiratory and expiratory muscles contribute to the reversal of intrathoracic pressure during expulsion. During emesis, the high positive intrathoracic pressure represents the force for expulsion of lumenal contents into the mouth. [56] Oral propulsion of the vomitus is

facilitated by movement of the hyoid bone and larynx up and forward. [59] Glottic closure prevents pulmonary aspiration of gut contents. Elevation of the soft palate prevents passage of the vomitus into the nasal cavities. Before emesis, the patient may hyperventilate. However, with vomiting, ventilation is suppressed except for small inspirations between expulsions. [60]

Gastrointestinal Myoelectric and Motor Events Associated with Nausea and Vomiting Gastrointestinal motility is regulated by oscillatory electrical activity, known as the *slow wave,* that controls the maximal frequency and direction of contractions. [61] In the stomach, a pacemaker in the gastric body generates slow waves at 3 cycles per minute (cpm) that propagate to the pylorus. Small intestinal slow waves originate in the proximal duodenum at 11 to 12 cpm. [62] Patients with nausea from pregnancy, gastroparesis, functional dyspepsia, or motion sickness exhibit abnormalities of gastric slow wave frequency, including accelerated (tachygastria) or decelerated (bradygastria) cycling. [63], [64], [65], [66] and [67] Gastric dysrhythmias may result from single ectopic pacemaker sites that overdrive the rest of the stomach, or from several foci with uncoupling of electrical propagation. [68] The causal relationship between gastric dysrhythmias and nausea is unproven. In some conditions, slow wave rhythm is disturbed before or simultaneously with the first report of symptoms, suggesting a possible pathogenic role. [66], [67], [69], [70] However, other stimuli that produce nausea do not disrupt the normal slow wave, indicating that dysrhythmias are not necessary to experience nausea. Motor phenomena that occur before retching include relaxation of the lower esophageal sphincter and proximal stomach, and contraction of the cricopharyngeus and cervical esophagus. [71] With vomiting, gut myoelectric and motor events are more pronounced. Before emesis, intestinal slow waves are abolished and followed by bursts of electrical spikes that propagate orally. [72] A retroperistaltic contractile complex (retrograde giant contraction) begins in the mid small intestine and propagates into the stomach, causing enterogastric reflux. [73] Investigations involving autotransplantation of intestinal segments indicates that these motor phenomena are controlled by extrinsic innervation. [74] After the retrograde giant contraction has passed, phasic intestinal contractions persist for several minutes and are followed by motor quiescence. [73] Subsequent pyloric closure, antral contraction, and lower esophageal sphincter relaxation facilitate expulsion of the intragastric contents. Retroperistaltic contractions of the cervical esophageal body and pharynx also have been described. [59], [71] Blockade of these motor events with atropine or vagotomy does not prevent emesis, indicating they are not strictly needed for vomiting to occur; rather, they enhance the evacuation of lumenal contents. [73], [75] The neuronal pathways that mediate the gut motor events associated with emesis are incompletely characterized. The ability of atropine to block retrograde contractions indicates a role for vagal cholinergic innervation. [73] Centrally, a role for NK_1 receptor mediation was suggested by a study in rats in which injection of substance P into the dorsal vagal motor nucleus produced gastric relaxation that was attenuated by vagotomy or by pretreatment with an NK_1 receptor antagonist. [76]

Endocrine and Autonomic Responses to the Initiation of Nausea and Vomiting Levels of circulating hormones and neurotransmitters are altered with nausea and vomiting. Vasopressin levels increase during motion sickness and electrical vagal stimulation and after apomorphine and cisplatinum administration. [77], [78] and [79] Intravenous vasopressin at supraphysiological doses evokes nausea in humans, suggesting the hormone may contribute to symptom production, whereas in monkeys, vasopressin V_1 antagonists prevent motion-induced emesis. [80], [81] However, the ability of apomorphine to produce vomiting in patients with central diabetes insipidus suggests that vasopressin is not critical for the induction of emesis. [82] Levels of other transmitters, such as epinephrine, ß-endorphin, growth hormone, cortisol, adrenocorticotropic hormone, and prolactin, change during nausea and vomiting; however, their pathogenic importance is uncertain. [83] Autonomic neural disturbances result from the proximity of brainstem nuclei mediating vomiting to those controlling respiratory, vasomotor, cardiac, and salivary activity. With nausea, pallor, diaphoresis, hypersalivation, defecation, tachycardia, or hypotension may be noted. Elevated parasympathetic vagal activity, as measured by increased beat-to-beat heart rate variability, has been observed in women with ovarian cancer experiencing nausea after chemotherapy. [84] Bradycardia and blood pressure variation may occur with retching or vomiting.

Investigation of the Pathophysiology of Nausea

Because of a lack of a suitable animal model, the neural structures that mediate the sensation of nausea are poorly understood. It has been postulated that nausea results from lesser activation of the same pathways that produce emesis. Some investigators consider measurable changes in autonomic function or hormone release to correlate with the experience of nausea in animal models. [85] However, most of these phenomena persist in decerebrate animals incapable of perceiving gastrointestinal sensations. Food avoidance has been proposed as a measure of nausea, but the validity of this model is unconfirmed. [86] Several investigations provide insight into cerebral cortical regions stimulated during nausea in human models. Activation of the inferior frontal gyrus in proportion to the degree of nausea has been demonstrated by magnetic source imaging in healthy human volunteers exposed to vestibular stimulation or apomorphine. [87] This finding is consistent with observations of electroencephalographic changes in temporofrontal regions during motion sickness. [88]

DIFFERENTIAL DIAGNOSIS

The differential diagnosis of nausea and vomiting is extensive and includes a broad range of pathological and physiological conditions affecting the gut, peritoneal cavity, central nervous system, and endocrine and metabolic functions (Table 37-1).

TABLE 37-1 Differential Diagnosis of Nausea and Vomiting

Medications

Drug reactions are among the most common causes of nausea and vomiting and usually present soon after the initiation of therapy. Emetogenic drugs may exert their effects by actions on distinct anatomic sites within the central and peripheral nervous systems. Medications that act on the area postrema include antiparkinsonian agents (e.g., L-dopa, bromocriptine), nicotine, digoxin, and opiate analgesics. [20] Nonsteroidal antiinflammatory drugs and erythromycin activate peripheral afferent pathways, most likely vagal. [89] Other medications that cause nausea and vomiting include cardiac antiarrhythmics, antihypertensives, diuretics, oral antidiabetics, oral contraceptives, and gastrointestinal medications such as sulfasalazine.

The most extensively studied medications that evoke emesis are the cancer chemotherapeutic agents. Risk factors for chemotherapy-induced nausea include a low level of social functioning, prechemotherapy nausea, female sex, and the administration of highly emetogenic chemotherapy. [90] Emesis from chemotherapy may be acute, delayed, or anticipatory. Potent agents that induce acute vomiting within hours of administration include cisplatinum, nitrogen mustard, and dacarbazine, all of which increase plasma serotonin levels or the urinary excretion of 5-hydroxyindoleacetic acid (5-HIAA), a serotonin metabolite. [91], [92] In contrast, less emetogenic agents, such as cyclophosphamide, evoke little rise in plasma serotonin or urinary 5-HIAA. [91] Cisplatinum raises ileal tissue serotonin levels and increases the number of serotonin-immunoreactive cells in animal models. [92] Cisplatinum-evoked emesis is reduced by vagotomy, suggesting mediation by peripheral afferent pathways. [30] However, the inability of the peripherally active 5-HT_3 receptor antagonist zatosetron-QUAT to block cisplatinum-induced emesis when given intravenously, coupled with a significant antiemetic effect when administered within the intracerebral ventricles, indicates a central neural component to vomiting evoked by cisplatinum. [93]

In contrast to acute chemotherapy-evoked emesis, delayed and anticipatory vomiting are mediated by serotonin-independent pathways. Delayed emesis after

cisplatinum is not associated with urinary 5-HIAA excretion and is poorly controlled by 5-HT $_3$ receptor antagonist treatment. [91], [94] In dogs, 5-HT $_4$ receptors may mediate the delayed phase of chemotherapy-induced emesis. [95] In another study, increased norepinephrine production was noted during the delayed phase after cisplatinum, suggesting the participation of other neuroendocrine mediators. [96] Anticipatory nausea and vomiting occur in 25% of patients by the fourth course of cancer chemotherapy, more often in younger patients. [97] The development of anticipatory symptoms is dependent on anxiety and coping mechanisms. [97], [98] Patient expectations strongly predict the onset of anticipatory nausea but not anticipatory vomiting. [99] Anticipatory emesis is poorly controlled by antiemetic medications and is best managed by relaxation therapy, systematic desensitization techniques, and anxiolytic medications such as benzodiazepines.

Infectious and Toxic Causes

Infectious illness produces nausea and vomiting, usually of acute onset. Acute enteric illness occurs most often in children younger than 3 years of age but exhibits a second peak incidence in the third decade of life. [1] It occurs at a rate of 1.2 infections per person per year, commonly in the autumn and winter. Viral gastroenteritis may be caused by rotaviruses, reoviruses, adenoviruses, and the Hawaii, Snow Mountain, and Norwalk agents. [100], [101], [102], [103] and [104] Bacterial infection with *Staphylococcus aureus, Salmonella, Bacillus cereus,* and *Clostridium perfringens* also produces nausea and vomiting, in many cases via toxins that act on the area postrema. [105], [106] and [107] The enterotoxin elaborated by *S aureus* has been characterized and is distinct from enterotoxins A through E and the toxic shock syndrome toxin 1. [108] Nausea in immunocompromised patients may result from gastrointestinal infection with cytomegalovirus or herpes simplex virus. [109], [110] Nongastrointestinal infections that produce nausea include otitis media, meningitis, and hepatitis.

Disorders of the Gut and Peritoneal Cavity

Gut and peritoneal disorders represent prevalent causes of nausea and vomiting. Gastric or small intestinal obstruction produces prominent nausea that may be relieved by vomiting. Gastric obstruction often is intermittent, whereas small intestinal obstruction usually is acute and associated with abdominal pain. Superior mesenteric artery syndrome is a rare condition in which the duodenum is compressed by the overlying superior mesenteric artery as it arises from the aorta, causing anatomic obstruction. [111] This syndrome is seen in patients who have had profound weight loss, recent surgery, or prolonged bed rest. Other rare mechanical causes of nausea and vomiting include subacute gastric volvulus and antral webs. [112], [113]

Motility disorders such as gastroparesis and chronic intestinal pseudoobstruction produce nausea secondary to an inability to clear retained food. Gastroparesis is caused by systemic diseases including diabetes mellitus, scleroderma, systemic lupus erythematosus, polymyositis-dermatomyositis, and amyloidosis. [114], [115] Diabetic gastroparesis commonly results from long-standing poor glycemic control but is not necessarily associated with a poor prognosis. [116], [117] Animal models suggest that gastric stasis occurring secondary to diabetes may be a consequence of loss or damage to the interstitial cells of Cajal, which regulate the pacemaker activity of the stomach. [118] Delayed gastric emptying of solids is demonstrable in two thirds of scleroderma patients with abnormal esophageal motility. [119] Gastroparesis develops in fewer than 5% of patients who undergo vagotomy and gastric drainage. [120] In patients with the acquired immunodeficiency syndrome, human immunodeficiency virus infection may contribute to delays in gastric emptying. [121] Pancreatic adenocarcinoma produces gastroparesis by unknown mechanisms. [122] Gastric ischemia is a rare cause of gastroparesis, especially in women. The median arcuate ligament syndrome produces gastroparesis by compression of the celiac axis. [123] Chronic intestinal pseudoobstruction may be hereditary or result from systemic diseases similar to those that produce gastroparesis. Paraneoplastic intestinal pseudoobstruction is reported with some malignancies, most commonly small cell carcinoma of the lung. [124]

Nearly one third of patients with gastroparesis exhibit no underlying systemic disease to explain their symptoms. Some cases of idiopathic gastroparesis are preceded by prodromal symptoms such as diarrhea, fever, headache, or myalgias, consistent with a viral etiology. The agent responsible is rarely identified; however, one report in children detected rotavirus in 8 of 11 cases. [125] In general, postviral gastroparesis confers a better prognosis than nonviral idiopathic disease, with a shorter duration of illness, less severe symptoms, and a better quality of life. [125], [126] Women with idiopathic gastroparesis often report prior physical or sexual abuse, similar to observations in patients with irritable bowel syndrome. [127] Many patients with gastroparesis exhibit disruption of slow wave rhythmicity, suggesting that pacemaker abnormalities may induce some of the motor findings and symptoms in these individuals. [65]

Nausea and vomiting may be prominent in patients with functional (nonulcer) dyspepsia, especially of the dysmotility subtype. Although many patients with functional dyspepsia exhibit abnormalities of gastric motor or myoelectric function, these abnormalities correlate poorly with symptoms and may not be pathogenic in such cases. [128] Nausea also may be a consequence of gastroesophageal reflux; however, many affected individuals also exhibit delayed gastric emptying, providing another cause for their symptoms. [129]

Abdominal disorders not involving the gut lumen may produce nausea and vomiting. Inflammatory conditions such as pancreatitis, appendicitis, and cholecystitis activate afferent pathways from the peritoneum. Biliary colic in the absence of inflammation produces nausea by activating afferents through distention of the biliary tree. Fulminant hepatic failure results in nausea, presumably because of the retention of unknown emetic toxins and increased intracranial pressure.

Central Nervous System Causes

Many emetic stimuli produce symptoms by action on the cerebrum or brainstem. Increased intracranial pressure from malignancy, infarction, hemorrhage, infection, or congenital causes may produce emesis with or without associated nausea. Studies in canine models demonstrate maximal induction of emesis at an intracranial pressure of 80 mm Hg. [130] Emotional responses to unpleasant smells, tastes, or memories may induce vomiting. Psychogenic vomiting most commonly presents in young women, often with a history of psychiatric illness or social difficulties. [131] Other psychiatric conditions associated with nausea include anxiety disorders, depression, anorexia nervosa, and bulimia nervosa.

Labyrinthine disorders such as labyrinthitis, tumors, and Ménière disease produce nausea and vomiting, often with vertigo. Motion sickness is evoked by repetitive movements that activate the vestibular nuclei and is associated with extensive autonomic activation that produces pallor, diaphoresis, and salivation. [132] Space sickness, experienced by most astronauts in zero gravity, is related to motion sickness but may not exhibit the associated autonomic phenomena. [133]

Endocrine and Metabolic Causes

Endocrine and metabolic causes of nausea include uremia, diabetic ketoacidosis, hyper- and hypoparathyroidism, hyperthyroidism, and Addison disease. It has been postulated that uremia, diabetic ketoacidosis, and hypercalcemia activate the area postrema; however, thyroid and parathyroid disease also disrupt normal gastrointestinal motor activity. [20]

First-trimester pregnancy is the most common endocrine cause of emesis, occurring in 50% to 70% of pregnant women. On average, symptoms begin 39 days after the last menses, peak in the ninth gestational week, and last 35 to 45 days. [11], [134] Although nausea of pregnancy is commonly referred to as *morning sickness,* only 1.8% of women report symptoms restricted to the morning, whereas 80% experience symptoms lasting all day. [134] Nausea of pregnancy is more common in women who are primigravidas, younger, less educated, housewives, and overweight. [135] The positive relationship between nausea of pregnancy and preconception body mass is postulated to serve a beneficial role by suppressing maternal tissue synthesis and partitioning nutrients in favor of the fetus. [136] In general, first-trimester nausea is not deleterious to either the fetus or mother. Rather, it has been associated with reduced incidences of miscarriage, congenital heart defects, and fetal demise. [137], [138] However, in less developed regions where malnutrition is prevalent, nausea and vomiting may have nutritional consequences in the later stages of pregnancy that increase the risk for poor pregnancy outcomes. [139] Furthermore, women with more severe or prolonged symptoms may deliver infants with reduced birth weights. [140] Hyperemesis gravidarum, a condition of intractable vomiting complicating 1% to 5% of pregnancies, may produce dangerous fluid and electrolyte abnormalities. The cause of nausea of pregnancy is unknown but is believed to be hormonally related. Although symptoms parallel levels of ß-human chorionic gonadotropin, no clear pathogenic role has been discerned for this hormone. [141] Intolerance of oral contraceptives is strongly associated with nausea of pregnancy, suggesting potential roles for estrogen and progesterone. [142] One recent investigation has reported an association between elevated maternal serum prostaglandin E $_2$ levels and nausea of pregnancy. [143] Acute fatty liver of pregnancy, a condition distinct from first-trimester nausea, produces severe third-trimester vomiting and can be complicated by liver failure, disseminated intravascular coagulation, and fetal or maternal death. [144]

Radiation-Induced Vomiting

Radiation therapy for malignancy produces emesis by effects on both the structure and function of the gut. The incidence of nausea and vomiting is dependent on the region that is irradiated, being as high as 80% when the upper abdomen is included in the radiation field. [145] Involvement of serotoninergic pathways is indicated by the ability of 5-HT $_3$ receptor antagonists to reduce nausea and vomiting evoked acutely by abdominal radiation therapy.

Postoperative Nausea and Vomiting

Nausea and vomiting complicate 17% to 37% of surgical operations, more commonly in women. [146], [147] Postoperative nausea and vomiting occur more frequently after general anesthesia than with regional nerve blocks and correlate with the duration of surgery. The condition occurs more often after abdominal and orthopedic surgery than with laparoscopic or other extra-abdominal operations and is exacerbated by concurrent use of opiate agents.

Cyclic Vomiting

Cyclic vomiting syndrome is characterized by discrete episodes of intractable emesis with intervening asymptomatic periods. The mean age at onset is 5 years, and the condition affects both sexes equally. Affected children typically have eight or nine attacks per year with a mean duration of 1 to 3 days. [148], [149] Two thirds of children miss more than 10 days of school per year. [149] There are strong associations of cyclic vomiting with migraine headaches, motion sickness, gastroesophageal reflux, psychological symptoms, atopy, and prior forceps delivery. [149] Attacks may be precipitated by stress or infections. Cyclic vomiting syndrome also has been observed in adults, who infrequently report the onset of symptoms during menstrual periods or pregnancy or after ingesting a large meal. [150]

The etiology of cyclic vomiting is unknown and is likely multifactorial. Affected children are at increased risk for the development of migraine headaches as they enter adolescence, and there often is a strong family history of migraines. [151], [152] Children with migraine-associated cyclic vomiting syndrome report milder emetic episodes, more abdominal pain, more headache with photophobia, and increased psychological stress. [152] Other proposed causes of cyclic vomiting syndrome in subsets of patients include food allergy and mitochondrial fatty acid oxidation disorders, such as short- and medium-chain acyl-CoA dehydrogenase deficiency and late-onset glutaric acidemia type II. [153], [154]

Miscellaneous Conditions

Posterior myocardial infarction produces nausea via diaphragmatic irritation. Nausea may occur in congestive heart failure from passive congestion of the liver and gut. Excess ethanol intake induces vomiting by local action on the gut and by action on the brainstem. Acute graft versus host disease is the dominant cause of nausea and vomiting in bone marrow transplant recipients, most commonly occurring within the initial months after the transplant. [110] Jamaican vomiting sickness occurs after the ingestion of unripe akee fruit. [155] Excess vitamin intake as well as extended fasting or starvation also may cause nausea.

HISTORY AND PHYSICAL EXAMINATION

Historical Features

A detailed history provides useful diagnostic information about the cause of unexplained nausea and emesis. Acute vomiting (1–2 days) most often results from infection, a medication or toxin, or the accumulation of endogenous toxins, as in uremia or diabetic ketoacidosis. Chronic vomiting (>1 week) usually results from long-standing medical or psychiatric conditions.

Timing of Nausea and Vomiting The timing of symptoms provides clues to the underlying disease. Vomiting soon after a meal occurs with gastric obstruction from ulcer disease or malignancy. Psychogenic vomiting also occurs soon after eating, but most patients control the act of emesis until a convenient receptacle is available. Patients with gastroparesis may report nausea within 5 minutes after eating; however, most have symptoms more than 1 hour after a meal. Nausea and vomiting from inflammatory conditions such as cholecystitis and pancreatitis may present in the first postprandial hour. In some cases of esophagitis or ulcer disease, nausea may abate with eating. Vomiting early in the morning may be reported in first-trimester pregnancy, uremia, and chronic alcoholism.
Character of the Vomitus The characteristics of the vomitus can assist in the diagnosis. The return of undigested food suggests achalasia or a Zenker diverticulum. Vomiting of partly digested food hours or days after ingestion suggests gastric obstruction or gastroparesis. Bile in the vomitus excludes obstruction proximal to the ampulla of Vater, whereas blood or coffee-ground material suggests a process with mucosal damage, such as an ulcer or malignancy. However, retching and vomiting can induce hematemesis from Mallory-Weiss tears across the gastroesophageal junction. Voluminous clear acidic vomitus suggests gastric hypersecretion from Zollinger-Ellison syndrome. Feculent emesis occurs in distal intestinal or colonic obstruction, intestinal bacterial overgrowth, and gastrocolic fistulae. Odorless emesis may be noted with gastric achlorhydria.
Symptoms Associated with Nausea and Vomiting Abdominal pain is noted with ulcer disease, intestinal obstruction, and inflammatory disorders such as cholecystitis and pancreatitis. Vomiting may relieve nausea and pain in ulcer disease and intestinal obstruction but has no effect in inflammatory conditions. Diarrhea, fever, or myalgias are suggestive of enteric infection. Weight loss and malnutrition occur with chronic illness, but psychogenic vomiting rarely produces weight loss. Central nervous system lesions or meningitis are suggested by headaches, visual changes, altered mentation, and neck stiffness. In these disorders, emesis may be effortless, there may be no nausea, or emesis may be projectile. Labyrinthine diseases present with tinnitus or vertigo. Chest pain, dysphagia, or jaundice suggests pregnancy or cardiac disease, esophageal disease, or hepatobiliary disease, respectively. Associated symptoms also indicate the severity of the underlying condition and help to direct treatment. Prolonged vomiting may produce significant fluid and electrolyte loss that may be manifested as postural lightheadedness, rapid heart rate, and mouth dryness.
Complications of Nausea and Vomiting Gastrointestinal hemorrhage may result from retching-induced mucosal tears across the gastroesophageal junction, known as *Mallory-Weiss tears*. A more severe complication, Boerhaave syndrome, occurs when retching or vomiting produces complete esophageal rupture with subsequent mediastinitis or peritonitis. In patients with impaired consciousness, emesis may be complicated by pulmonary aspiration of acidic material leading to severe chemical pneumonitis.

Physical Examination Findings

The physical examination assists in the diagnosis and management of nausea and vomiting. Fever suggests infection or inflammation. Tachycardia or orthostatic hypotension indicates dehydration, which is supported by finding a loss of skin turgor or dry mucous membranes. Skin examination may show sclerodactyly in scleroderma or jaundice in hepatobiliary disease. Oral examination may reveal loss of dental enamel, common in bulimia. Adenopathy raises the possibility of a neoplasm. Hepatomegaly also is found in malignancy as well as in benign hepatic disease. An absence of bowel sounds signifies ileus, whereas high-pitched hyperactive bowel sounds with a distended abdomen are consistent with intestinal obstruction. A succession splash on side-to-side movement is found in gastric obstruction or gastroparesis. If abdominal tenderness or guarding is found, an inflammatory or infectious process such as an ulcer, cholecystitis, pancreatitis, or peritonitis is considered. Gross or occult fecal blood on rectal examination should prompt evaluation for an ulcer or neoplasm. On neurological examination, focal signs, papilledema, and impaired mentation suggest a central nervous system process, whereas nuchal rigidity is consistent with meningitis. Asterixis is present in metabolic diseases such as uremia and hepatic failure. Patients with gut motility disturbances may exhibit peripheral or autonomic neuropathy.

LABORATORY STUDIES AND DIAGNOSTIC TESTING

A thorough history and examination provide the information necessary to diagnose and treat most patients with nausea and vomiting. However, some patients require blood studies, structural evaluations, or assessment of gut function for diagnosis and management.

Laboratory Testing

Laboratory tests are ordered based on the findings of the history and physical examination. With long-standing symptoms or dehydration, the serum electrolytes can show hypokalemia or an elevated blood urea nitrogen relative to the creatinine level. Metabolic alkalosis may result from a loss of hydrogen ions in the acidic vomitus and contraction of the extracellular space from dehydration. A complete blood cell count can rule out anemia from inflammation or blood loss, leukocytosis from an inflammatory source, or leukopenia from a viral infection. Chronic blood loss also may be suggested by low serum levels of iron and iron saturation of transferrin.

Hypoalbuminemia is seen in some chronic diseases and in conditions with gut protein loss. Amylase, lipase, and liver chemistries are obtained for suspected pancreatic or hepatobiliary disease. Endocrine and metabolic causes can be assessed through serum pregnancy testing, thyroid function testing, and measuring the levels of blood urea nitrogen, creatinine, glucose, ketones, calcium, and plasma cortisol. Specific serologic markers can be obtained for presumed collagen-vascular diseases such as lupus, whereas results of tests for antineuronal nuclear antibodies are positive with paraneoplastic intestinal pseudoobstruction (usually from small cell lung carcinoma). [124] Meningitis is confirmed by lumbar puncture.

Structural Evaluation

Structural investigation may be needed to exclude organic disease as a cause of emesis. Flat and upright abdominal radiographs are obtained as a screening examination. Small intestinal air-fluid levels with an absence of colonic air suggests intestinal obstruction, whereas diffuse lumenal distention with absent bowel sounds is consistent with ileus or chronic pseudo-obstruction. Subdiaphragmatic free air indicates visceral perforation. Suspected small intestinal obstruction may be further evaluated, documented, and characterized by dedicated small intestinal contrast radiography. Water contrast agents such as Gastrografin provide reasonable degrees of accuracy in the exclusion of obstructions that require operative intervention. [156] However, barium provides superior anatomic characterization of partial obstruction. If symptoms are intermittent and the index of suspicion for intermittent obstruction is high, enteroclysis may provide a more detailed assessment of small intestinal lumenal processes. [157] With this technique, the proximal intestine is intubated under fluoroscopic guidance and barium and methylcellulose are perfused to provide a double contrast image. If colonic obstruction is a consideration, colonoscopy or barium enema radiography should be performed before small intestinal contrast studies. Upper gastrointestinal endoscopy or contrast radiography may be performed in cases of suspected ulcer disease or outlet narrowing. Endoscopy affords the advantage of providing biopsy capability of abnormal mucosa. Endoscopy also may suggest gastroparesis, demonstrating retained food in the absence of outlet obstruction. For suspected pancreaticobiliary disease, ultrasonography or computed tomography (CT) may be useful. Biliary scintigraphy may provide findings suggesting acute cholecystitis. CT or magnetic resonance imaging (MRI) of the head is indicated if the presentation suggests a central nervous system cause. If bowel ischemia is a consideration, mesenteric angiography or MRI of the mesenteric flow may be diagnostic.

Studies of Gastrointestinal Motor and Myoelectric Activity

When lumenal obstruction has been excluded, functional causes of nausea such as gastroparesis and pseudoobstruction are entertained. The clinician may elect to treat empirically with a medication designed to stimulate gut motility. Alternatively, testing of gut motor and myoelectric activity can be performed to characterize the functional defects.

Quantification of Gastric Emptying Gastroparesis is diagnosed by demonstrating delayed gastric emptying of an ingested meal. Scintigraphic measures of the emptying rates of liquid (e.g., ^{111}In-DTPA [diethylenetriamine pentaacetic acid] in water) or solid (e.g., ^{99m}Tc-sulfur colloid in eggs) radionuclides are the most commonly employed tests of gastric emptying (Fig. 37-2). [158] Under normal conditions, liquids empty with a half-time of between 8 and 28 minutes. Emptying of solids exhibits an initial lag phase, in which little label is passed into the intestine, followed by a linear emptying phase with evacuation of 40% to 80% of the tracer from the stomach at 2 hours. Some clinicians have proposed extending scintigraphic testing to include the 4 hours after eating to enhance the diagnostic accuracy of the test. [159] Solid phase emptying is affected earlier and to a greater extent than liquid emptying in gastroparesis; thus, solid phase scans are more sensitive for diagnosis.

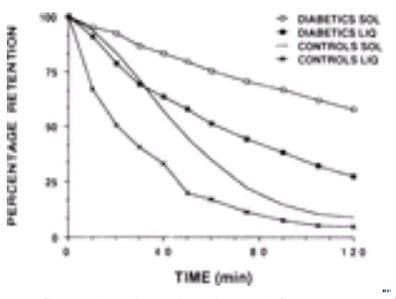

FIGURE 37-2. Gastric-emptying profiles of solids and liquids in diabetics with gastroparesis are delayed in comparison with those of healthy volunteers. (From Urbain JL, Vantrappen G, Janssens J, et al. Intravenous erythromycin dramatically accelerates gastric emptying in gastroparesis diabeticorum and normals and abolishes the emptying discrimination between solids and liquids. J Nucl Med 1990;31:1490.)

Other means of quantifying gastric emptying are under investigation. Investigations measuring gastric dimensions with ultrasound have reported highly reproducible results and demonstrable abnormalities in patients with known gastroparesis. [160], [161] However, ultrasound is less reliable for assessing the emptying of solid meals, so that the test is less useful in patients with mild gastroparesis. MRI can assess emptying of both liquid and solid phases of a meal; however, its cost precludes its widespread use at this time. [162], [163] and [164] Single photon emission computed tomography (SPECT) has been used to measure volumes in the proximal and distal stomach after eating. [165] In addition to determining emptying, this technique offers the potential to assess defects in gastric fundic relaxation. A promising new technology to quantify gastric emptying is ^{13}C breath testing, which involves ingestion of a nonradioactive ligand bound to a digestible nutrient and offers the potential for use in populations in whom scintigraphy is relatively contraindicated (e.g., children and pregnant women). With this technique, ^{13}C-octanoate, ^{13}C-acetate, or ^{13}C-*Spirulina platensis* ingested with a solid meal is delivered into the upper intestine at a rate dependent on the rate of gastric emptying. [166], [167] Digestion and absorption of the ingested compound liberate ^{13}C-carbon dioxide, which diffuses across the intestinal mucosa and is expired in the breath. Results of breath testing correlate reasonably well with those of gastric scintigraphy, although falsely abnormal results are possible in malabsorptive or pulmonary disease. [167], [168] and [169] Some studies suggest that optimal measurements of emptying rates by breath testing may require 6 to 8 hours, thus impairing the clinical utility of the technique. [166], [169]
Gastrointestinal Manometry When scintigraphy provides an incomplete diagnosis of the cause of nausea, gastrointestinal manometry can be performed. Because of the discomfort involved and the personnel required, this test is performed only in specialized centers. Under fasting conditions, the stomach and small intestine exhibit a pattern known as the migrating motor complex (MMC), which consists of three phases that are repeated every 90 to 120 minutes. [170] The most propulsive phase, phase III, clears undigested debris from the upper gut. Absence of gastric phase III, found in some cases of gastroparesis, predisposes to bezoar formation. Loss of intestinal phase III may lead to bacterial overgrowth. The postprandial period exhibits irregular, continuous contractions. [171] The absence of this fed pattern in the stomach correlates with delayed solid phase gastric emptying. [172] In addition to defining gastric dysmotility, manometry can characterize small intestinal disturbances (Table 37-2). In intestinal pseudoobstruction, barium radiography may show intestinal dilatation and delayed transit. Small intestinal manometry provides more specific information regarding the neuropathic or myopathic nature of the condition. Pseudo-obstruction resulting from enteric nerve dysfunction, as in the familial visceral neuropathies or early scleroderma, produces intense, uncoordinated bursts of motor activity with loss of normal MMC and fed motor activity. [173] Smooth muscle dysfunction, as in the familial visceral myopathies or late scleroderma, produces very low amplitude contractions. [174] Intermittent bursts of phasic contractions separated by periods of motor quiescence, a pattern known as the *minute rhythm,* have been observed in patients with small intestinal obstruction, although similar patterns are seen in neuropathic pseudoobstruction and irritable bowel syndrome. [175] Finally, the demonstration of brief, simultaneous pressure increases in all recording sites is diagnostic of rumination syndrome and eliminates the need for further investigation. [176]

MANOMETRIC FINDING	CHARACTERISTICS	DISEASES
Intestinal bursts	Irregular phasic contractions of normal amplitude; failure to develop normal fasting or fed patterns	Visceral neuropathies Scleroderma (early) Amyloidosis (early) Paraneoplastic pseudoobstruction Chagas disease Brainstem lesion Neurofibromatosis
Intestinal hypomotility	Reduced-amplitude phasic contractions	Visceral myopathies Scleroderma (late) Amyloidosis (late) Polymyositis-myositis Muscular dystrophy Myotonic dystrophy
Minute rhythm	Intermittent phasic bursts separated by periods of motor quiescence	Mechanical obstruction Neuropathic pseudoobstruction
Simultaneous contractions	High-amplitude contractions seen simultaneously in all gastric and small intestinal channels	Rumination syndrome

TABLE 37-2 Manometric Findings in Small Intestinal Dysmotility Syndromes

There are limitations to gastrointestinal manometry. Abnormal motor patterns are observed occasionally in healthy individuals. A small number of normal people do not exhibit MMC activity during a 4- to 6-hour fasting recording period, or because of technical reasons, they do not exhibit a fed motor pattern. [177] Consequently, some investigators recommend monitoring intestinal motility for 24 hours in ambulatory fashion to compensate for physiological variability in MMC periodicity. [178] When the diagnosis of neuropathic or myopathic pseudo-obstruction is entertained, a full-thickness intestinal biopsy specimen obtained surgically may be required to document degeneration of nerve or muscle layers. Nevertheless, in selected clinical settings, manometry provides important information. Proposed indications for manometric testing include evaluation of unexplained nausea and vomiting, characterization of the neuropathic or myopathic nature of a dysmotility syndrome, and demonstration of whether dysmotility in a patient with constipation or gastroesophageal reflux is generalized. [179] Additionally, in children, the absence of phase III activity predicts the inability to tolerate enteral nutrition and suggests consideration of parenteral feedings. [180] Finally, and perhaps most importantly, the documentation of normal motility is inconsistent with intestinal pseudoobstruction and suggests that the diagnostic evaluation should be redirected to other etiologies. [181]

Electrogastrography Gastric myoelectric disturbances are diagnosed in referral centers by electrogastrography (EGG), a technique by which slow waves are measured with cutaneous electrodes overlying the stomach. Gastric electrical rhythms are recorded for 1 to 2 hours under fasting and fed conditions, during which unwanted signals from the heart, diaphragm, and intestines are filtered out. Recordings in healthy individuals exhibit a uniform 3-cpm rhythm that increases in amplitude after eating. In nauseated patients, symptoms may relate to abnormally rapid (tachygastria) or slow (bradygastria) EGG rhythms or to an absence of the postprandial signal increase (Fig. 37-3). Some investigators correlate the absence of the fed amplitude increase with delayed gastric emptying of solids. [182] However, other studies suggest that EGG and gastric scintigraphy define different patient populations and are complementary in evaluating the patient with unexplained nausea. [183] One group has observed that nauseated diabetics with EGG abnormalities experience more severe symptoms than those with normal EGG patterns, suggesting that dysrhythmic activity may be a marker of more advanced functional impairment of the stomach. [184] The performance of EGG is associated with pitfalls, including movement artifacts and noise, which can interfere with accurate interpretation. [185] Proposed indications for EGG include evaluation of unexplained nausea and vomiting, noninvasive testing to predict gastroparesis, and exclusion of generalized gastrointestinal dysfunction in patients with constipation or gastroesophageal reflux. [179] EGG has been shown to be of value in predicting the development of postoperative retching and nausea in children undergoing fundoplication for gastroesophageal reflux. [186]

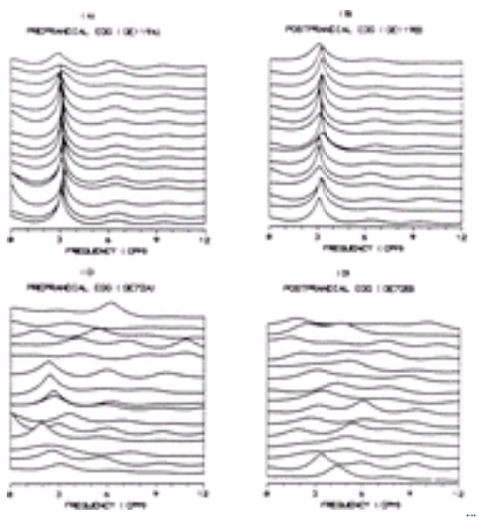

FIGURE 37-3. Frequency spectra of electrogastrographic recordings from an individual with normal gastric emptying and a patient with delayed gastric emptying. Under fasting conditions, the rhythm is regular at 3 cycles per minute (cpm) in the individual with normal emptying (**A**), whereas the patient with delayed emptying exhibits dysrhythmic activity with no dominant frequency (**C**). After eating, the person with normal emptying continues to show 3-cpm activity with increased signal amplitude (**B**), whereas the patient with delayed emptying shows a chaotic slow wave rhythm (**D**). (From Chen JD, Lin Z, Pan J, McCallum RW. Abnormal gastric myoelectrical activity and delayed gastric emptying in patients with symptoms suggestive of gastroparesis. Dig Dis Sci 1996;41:1538).

Approach to Functional Testing of Gastrointestinal Motor and Myoelectric Activity There currently is a lack of consensus regarding the clinical utility of measurements of gastric emptying, gastroduodenal manometry, and EGG in the investigation of nausea and vomiting because of a dearth of well-controlled outcome investigations. In the only large retrospective evaluation of the clinical applicability of gastric-emptying measurement, gastric scintigraphy was found not to influence clinical decision making or alter treatment options. [187] However, the investigation included many patients who were referred for symptoms other than nausea and vomiting, such as postoperative dumping syndrome. Diagnostic findings of intestinal manometry altered treatment recommendations in approximately 20% of patients in one study; however, many individuals were referred for evaluation of lower gastrointestinal symptoms. [188] In a more recent investigation, a diagnosis obtained on gastroduodenal manometry led to new therapy in 13% of cases, a new diagnosis in 15%, and referral to another specialist in 8%. [189] A positive clinical impact was observed in only 29% of patients. To date, there are no publications reporting an impact of EGG on adult patient outcome. Therefore, performance of functional testing in the majority of patients with unexplained nausea and vomiting is based on the ordering practices of each individual physician and on the “need to know” a specific diagnosis. Better-defined guidelines for the rational use of these studies await carefully controlled investigation.

PRINCIPLES OF MANAGEMENT

The care of the patient with nausea and vomiting involves assessment of the etiology and the severity of the condition, with prompt initiation of therapy to prevent complications.

Indications for Hospitalization

The first decision in managing the patient with vomiting is to determine if intravenous resuscitation is needed. Poor skin turgor or orthostatic pulse or blood pressure changes indicate that more than 10% of body fluids have been lost, mandating intravenous infusion of saline solution (0.45% or 0.9%). Potassium supplements may be started for hypokalemia if urine output is adequate. Intravenous fluids can be given in the emergency department or outpatient clinic if the patient is expected to resume adequate oral intake at home. If the prospects for oral replenishment are uncertain, hospitalization should be considered. Patients with mechanical obstruction or ileus may benefit from nasogastric suction. The threshold for hospitalization is lowered for diabetics, patients with concurrent diarrhea, persons with other chronic debilitating disease, and the very young or old, as these patients become rapidly dehydrated.

Dietary and Nonmedicinal Considerations

If the patient can return home, dietary advice should be given. Because liquids empty more rapidly from the stomach than solids, a predominantly liquid diet is recommended. [158] Frequent small meals are prescribed because large volumes may worsen symptoms. Lipids are potent inhibitors of gastric emptying, so that foods

rich in fats should be avoided. [190] Studies suggest that diets preferentially high in protein content may benefit patients with nausea of pregnancy and motion sickness. [191] In patients with gastroparesis, reducing indigestible dietary fiber is recommended to prevent organization of the residue into a bezoar. [192] Finally, medications that inhibit motor function should be discontinued if possible.

Aggressive control of blood glucose is advocated for nauseated patients with long-standing diabetes mellitus. Although the Diabetes Control and Complications Trial documented the beneficial effects of tight glycemic control on objective parameters of peripheral nerve function, the utility of intensive insulin therapy in reducing symptomatic manifestations of gastroparesis in diabetic patients is unknown. [193] Nonetheless, because acute hyperglycemia has profound disruptive effects on gastric motor and myoelectric activity, it is reasonable to strive for near-euglycemic plasma levels. [194]

Medical Therapy for Nausea and Vomiting

Medical or surgical treatment of nausea and vomiting should be directed at the underlying disease process whenever possible. Nevertheless, many patients benefit from the initiation of medications designed to suppress emesis and correct aberrant gastrointestinal function.

Antiemetic Medications Antiemetic drugs acting on central nervous system muscarinic, cholinergic, histaminergic, dopaminergic, or serotoninergic receptors reduce symptoms in many cases (Table 37-3). The use of these agents should be tailored to the underlying emetic stimulus. Antihistamines such as meclizine and dimenhydrinate are useful for vomiting associated with labyrinthine disorders, such as motion sickness or labyrinthitis. [195] They also are effective in treating nausea associated with uremia and the postoperative state. Antihistamines are safe, although sedation and dryness of the mouth may limit their use in some settings. Newer, less sedating antihistamines such as astemizole have limited antiemetic activity. [196]

TABLE 37-3 Characteristics of Antiemetic Medications

Drugs such as scopolamine that antagonize muscarinic receptors in vestibular pathways also are effective in motion sickness when given orally or transdermally. [197] Side effects of antimuscarinic agents, such as dryness of the mouth and eyes, sedation, impaired concentration, headaches, constipation, and urinary retention, limit their usefulness in some individuals. Dopamine receptor antagonists, including phenothiazines (prochlorperazine, chlorpromazine) and butyrophenones (droperidol, haloperidol), are the most commonly prescribed antiemetics. These agents act on area postrema dopamine D_2 receptors and are effective in treating emesis resulting from gastroenteritis, medications, abdominal irradiation, surgery, and toxins as well as some chemotherapeutic agents. [198] Antidopaminergic agents produce many central nervous system side effects, including drowsiness, insomnia, anxiety, mood changes, confusion, dystonic reactions, parkinsonian symptoms, and irreversible tardive dyskinesia. Through effects on the pituitary, antidopaminergics may induce hyperprolactinemia, leading to breast engorgement, galactorrhea, and sexual dysfunction. Other rare side effects include blood dyscrasias and jaundice. Many antidopaminergics have effects on other receptor subtypes and provoke antihistaminic and antimuscarinic side effects as well. Serotonin 5-HT_3 receptor antagonists are now the most widely used agents in the prophylaxis of chemotherapy-induced emesis; however, they frequently are given in other conditions that produce nausea and vomiting. In patients with persistent unexplained nausea and vomiting, 5-HT_3 receptor antagonists may produce small reductions in symptoms. [199] Patients with nausea secondary to human immunodeficiency syndrome also experience some benefit from this class of drug, as may individuals with bulimia nervosa. [200], [201] Side effects from the 5-HT_3 antagonists include constipation, headaches, and rare elevations in liver chemistries.

Prokinetic Agents for Gastroparesis and Intestinal Dysmotility Gut motility disorders may respond to drugs that stimulate gastric emptying and intestinal transit (Table 37-4). These agents are often given before meals to reduce postprandial gastric stasis, with a dose given at bedtime to effect gut clearance of undigested residue during sleep.

MEDICATION	MECHANISM OF ACTION	COMMON SIDE EFFECTS
Commonly Used Agents for Gastroparesis		
Metoclopramide	5-HT_4 receptor facilitation of acetylcholine release from enteric nerves Dopamine receptor antagonism 5-HT_3 receptor antagonism (weak)	Sedation Anxiety Mood disturbances Sleep disruption Dystonic reactions Tardive dyskinesia Galactorrhea Sexual dysfunction
Erythromycin	Motilin receptor agonist	Abdominal pain Nausea and vomiting Diarrhea
Agents of Limited Prescription in Gastroparesis		
Cisapride	5-HT_4 receptor facilitation of acetylcholine release from enteric nerves Direct smooth muscle stimulant 5-HT_3 receptor antagonist (weak)	Cardiac arrhythmias Abdominal cramps Diarrhea
Domperidone	Peripheral dopamine receptor antagonist	Galactorrhea/gynecomastia Sexual dysfunction
Bethanechol	Muscarinic receptor agonist	Abdominal pain Salivation Nausea Diaphoresis
Agents for Small Intestinal Dysmotility		
Octreotide	Somatostatin receptor analog	Diarrhea Altered glycemic control (diabetics) Gallstones Thyroid disease
Leuprolide	Gonadotropin-releasing hormone analog	Amenorrhea Osteoporosis

TABLE 37-4 Prokinetic Medications for Gastrointestinal Dysmotility Syndromes

Metoclopramide is a substituted benzamide that acts via serotonin (5-HT_4) receptor facilitation of gastric cholinergic transmission and via dopamine (D_2) receptor antagonism in the stomach and brainstem. [202], [203] The prokinetic properties of metoclopramide are limited to the proximal gut; thus, the drug is useful for gastroparesis but not small intestinal or colonic dysmotility. Metoclopramide possesses antiemetic activity as well as motor stimulatory action, providing additional symptom control. Because of central antidopaminergic side effects, such as agitation, drowsiness, dystonias, and irreversible tardive dyskinesia, metoclopramide is poorly tolerated in up to 20% of patients. Hyperprolactinemia as a consequence of dopamine antagonism in the pituitary may cause impotence, galactorrhea, and amenorrhea. Selected macrolide antibiotics exhibit potent prokinetic effects in the upper gut. Erythromycin induces antroduodenal contractions and promotes gastric emptying through action on receptors for the hormone motilin, the physiological regulator of the MMC. The abilities of atropine and vagal cooling to block the effects of erythromycin indicate that vagal cholinergic pathways participate in its prokinetic effects. [204], [205] and [206] Some patients with gastroparesis and pseudoobstruction respond to erythromycin, but this agent has a narrow dose range of efficacy, which limits its utility. Low doses have no effect, whereas high doses induce abdominal pain, nausea, and diarrhea through the induction of intense motor spasms. [207], [208] and [209] Studies of investigational erythromycin analogs without antimicrobial effects show similar motor excitatory effects. [210], [211] and [212] Clarithromycin also stimulates gastroduodenal motility in healthy humans, suggesting potential for treating gastroparesis. [213] Domperidone, a benzimidazole derivative not presently available in the United States, is a dopamine D_2 receptor antagonist with prokinetic efficacy in gastroparesis comparable to that of metoclopramide. [214] Furthermore, the drug produces significant improvements in quality of life in affected patients. [215], [216] The drug crosses only into central nervous system regions with porous blood-brain barriers, such as the area postrema, and thus exhibits a side effect profile superior to that of metoclopramide. [217], [218] Because of its lack of central toxicity, domperidone is ideal for treating gastrointestinal symptoms in patients with Parkinson disease, who often must take dopaminergic drugs to control their somatic motor symptoms. [219] Although dystonic reactions are not observed with domperidone, hyperprolactinemia is still a problem because of the incomplete blood-brain barrier of the anterior pituitary. Other prokinetic agents have been largely abandoned because of unacceptable side effect profiles. Cisapride, a benzamide derivative acting on 5-HT_4 receptors to facilitate myenteric acetylcholine release, demonstrated clinical benefits in patients with gastroparesis and intestinal pseudoobstruction. [203], [220], [221] However, numerous deaths were attributed to cisapride, mainly because of induction of cardiac arrhythmias, including torsade de pointes. [222], [223] As a consequence, the drug was withdrawn from the market in 2000 except for selected low-risk patients. Bethanechol, a muscarinic receptor agonist with direct smooth muscle excitatory effects, is a potent stimulant of gut phasic contractions, but much of this motor activity is not propagative, and therefore the drug is a poor prokinetic. [224] Furthermore, side effects, including abdominal cramps, salivation, diaphoresis, and nausea, are prominent; thus, bethanechol is rarely used to manage gut motility disorders. Direct comparisons between the different prokinetic agents have not been made except in rare instances. A metaanalysis of published placebo-controlled clinical trials of metoclopramide, erythromycin, domperidone, and cisapride suggested

that erythromycin is most potent at stimulating gastric emptying, whereas domperidone and erythromycin are most effective at reducing symptoms. [225] Many patients with gastroparesis become refractory to standard prokinetic therapy and represent clinical challenges. In these individuals, combination treatment with two prokinetic drugs may provide superior relief. [226] Furthermore, drug administration by alternate routes may enhance their efficacy. Metoclopramide exhibits significant potency when administered subcutaneously. [227] Intranasal metoclopramide reduces vomiting after moderately emetogenic chemotherapy, suggesting that this experimental formulation may have future utility as well. [228] Agents with selective prokinetic effects on the small intestine may have some utility in certain patients with intestinal pseudo-obstruction. The somatostatin analog octreotide induces propagative small intestinal motor patterns in patients with pseudo-obstruction secondary to scleroderma and, when given over 3 weeks, reduces nausea, vomiting, abdominal discomfort, and objective measures of intestinal bacterial overgrowth. [229] Investigations indicate that octreotide may produce prolonged symptomatic improvement (>6 months) in some patients with pseudo-obstruction. [230] The gonadotropin-releasing hormone analog leuprolide evokes propagative gastric and small intestinal complexes in subsets of patients with intestinal dysmotility and may reduce nausea, bloating, and defecation disturbances in these individuals. [109], [231] Because leuprolide produces amenorrhea and may lead to osteoporosis, recommending this drug for an upper gut motility disorder should be judiciously considered.

Treatment of Emesis Resulting from Cancer Chemotherapy and Radiation Therapy Extensive investigation has focused on antiemetic regimens to prevent or treat nausea and vomiting complicating cancer chemotherapy. Most programs include multiple medications that act on distinct receptor sites. [232] For highly emetogenic agents such as cisplatinum, prophylactic regimens usually include 5-HT $_3$ receptor antagonists such as ondansetron and granisetron. [30], [93], [233] In contrast, 5-HT $_3$ receptor antagonists are less effective for delayed emesis after chemotherapy or with less emetogenic chemotherapeutic agents. [91], [233] Metoclopramide in high doses is useful for the prophylaxis of chemotherapy-induced emesis, possibly stemming from weak 5-HT $_3$ receptor antagonism. [234] Other antidopaminergic agents such as prochlorperazine and domperidone are used with efficacy with some forms of chemotherapy. Corticosteroids exhibit potent antiemetic effects against chemotherapy via unknown mechanisms. [235] In contrast to the 5-HT $_3$ receptor antagonists, the corticosteroids may provide the greatest benefit in reducing delayed chemotherapy-induced emesis. [236], [237] Intravenous benzodiazepines such as lorazepam are included in antiemetic regimens because they produce sedation and reduce anticipatory nausea. [238] Cannabinoids such as tetrahydrocannabinol and nabilone have efficacy in chemotherapy-induced emesis prophylaxis comparable to or slightly better than that of antidopaminergics; however, these drugs produce severe side effects such as somnolence, ataxia, syncope, seizures, and hallucinations, which are prominent in the elderly. [239], [240] Two thirds of the respondents to a recent survey of practicing oncologists indicated they would prescribe marijuana less than once monthly if it were rescheduled, in large part because of concerns about its toxicity. [241] The mechanisms of the antiemetic effects of cannabinoids are unknown, although tetrahydrocannabinol reduces prostaglandin synthesis and stimulates the endogenous production of endorphins. [242] Radiation therapy, both to abdominal and extraabdominal sites, may induce profound nausea and vomiting. As with chemotherapy-induced symptoms, 5-HT $_3$ receptor antagonists exhibit efficacy superior to that of metoclopramide in radiotherapy-evoked emesis. [243], [244] Similar to their actions with chemotherapy, the 5-HT $_3$ receptor antagonists produce better control of radiation-induced vomiting than of nausea. [243]

Therapy of Postoperative Nausea and Vomiting Several antiemetic regimens have been proposed to prevent or control postoperative nausea and vomiting. In several studies, the dopamine receptor antagonist droperidol and the 5-HT $_3$ receptor antagonists have shown efficacy in preventing postoperative symptoms. [245], [246] and [247] Furthermore, the combination of these agents appears to be superior to either drug alone. [245], [246] Other drug classes shown to reduce postoperative nausea and vomiting include the corticosteroids and the a $_2$-adrenoceptor agonist clonidine. [248], [249] and [250]

Miscellaneous Antiemetic Therapies Other medications have been shown in small, often uncontrolled trials to be efficacious in selected emetic disorders. Tricyclic antidepressants are used for nausea associated with depression. Recently, tricyclics have been found to be effective in patients with functional nausea and vomiting as well as in adults with cyclic vomiting syndrome. [150], [251] Emesis in children with cyclic vomiting often is relieved by antimigraine therapies such as sumatriptan. [152], [252] Patients with nausea resulting from opiate withdrawal can respond to opiate antagonists such as naloxone. The a-adrenoceptor agonist clonidine has been reported to benefit some patients with gastroparesis. [253] A new class of drugs that act as antagonists on tachykinin NK $_1$ receptors shows promise in treating both nausea and vomiting resulting from a broad range of conditions. NK $_1$ antagonists were initially used to control chemotherapy-induced emesis; in contrast to the 5-HT $_3$ receptor antagonists, they relieved delayed emesis and nausea as well as acute emesis. [254], [255] and [256] Subsequently, efficacy for NK $_1$ receptor antagonists was demonstrated in postoperative nausea and vomiting as well as motion sickness. [257], [258] and [259] Selected alternative and traditional nonmedicinal therapies have been promoted for treating nausea and vomiting. Ginger has been advocated for patients with postoperative nausea, motion sickness, nausea of pregnancy, and chemotherapy-induced nausea. [260] Vitamin B $_6$ (pyridoxine) also may be of benefit in some women with nausea of pregnancy. [261] Acupuncture, acupressure, and acustimulation have been suggested for nausea of pregnancy, postoperative nausea, motion sickness, and chemotherapy-induced emesis. [262], [263], [264], [265] and [266] Transcutaneous electrical nerve stimulation and electrical stimulation of the vestibular system have been shown to reduce chemotherapy-induced nausea and postoperative nausea, respectively. [267], [268] Finally, hypnosis has been proposed for the prevention of anticipatory nausea in chemotherapy patients. [269]

Surgical and Endoscopic Management of Nausea and Vomiting

For nausea and vomiting that are refractory to dietary measures and medications, the treatment options are limited. In diabetic gastroparesis, intermittent enteral feedings through a jejunostomy tube can reduce symptoms, decrease the number of hospitalizations, and improve the nutritional status. [270] Some patients benefit from the additional placement of a gastrostomy to vent trapped gas and secretions from the stomach. [271] Anecdotal reports suggest that pyloric injection of botulinum toxin may relieve symptoms in patients with diabetic gastroparesis, presumably by reducing associated pylorospasm. [272] In patients with intestinal pseudoobstruction in whom enteral feedings exacerbate symptoms, home intravenous hyperalimentation provides essential nutrition. In patients with pseudoobstruction, decompression stomata also may reduce symptoms when prokinetic drugs are ineffective. [273]

Reports suggest that some gastroparesis patients may derive benefit from electrical stimulation of the stomach. Gastric pacing at a rate of 3 cpm via electrodes implanted in the gastric serosa accelerates gastric emptying and reduces symptoms in patients with diabetic or idiopathic gastroparesis. [274] The U.S. Food and Drug Administration has approved a gastric neurostimulator as a humanitarian device for patients with diabetic or idiopathic gastroparesis. This device, which delivers low-energy electrical pulses at a rate of 12 cpm, improves gastric emptying only modestly; however, uncontrolled trials have reported 80% reductions in nausea and vomiting. [275], [276] Future electrical stimulators may offer the capability of delivering current sequentially at several sites in the stomach, providing greater coordinated propagation of the depolarizing stimulus. [277]

Surgical resections generally are of limited value in patients with refractory emesis. The main exception is gastroparesis resulting from prior vagotomy and gastric drainage surgery. In these patients, completion gastrectomy relieves symptoms, presumably by eliminating gastric retention. [278], [279] and [280] However, poor outcomes have been described in this setting for individuals with severe nausea who require parenteral nutrition and who retain solid food in the stomach. [281] Although gastric drainage procedures and resections generally do not provide symptom relief in patients with idiopathic gastroparesis or gastroparesis secondary to systemic disease, case reports have documented the effectiveness of total gastrectomy in selected patients with severe refractory gastroparesis. [282]

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CHAPTER 38

Pankaj Jay Pasricha

APPROACH TO THE PATIENT WITH ABDOMINAL PAIN

> We look away from each pain, in the waiting sadness, Hoping it will pass away. Yet they form our winter's Bower, darkly filling our senses with green, one Of our internal seasons, but not merely season, But place, dwelling, defense, foundation, home.
>
> –Rainier Maria Rilke, *The Duino Elegies*

Pain is a paradox, as reminded by the haunting words of the Austro-German poet Rilke. When acute, it serves to warn the organism about potentially noxious agents in the environment; when chronic, it can so dominate the clinical picture that it can no longer be regarded as a sentinel sensation subservient to an underlying disease but instead assumes the characteristics of a disease state by itself. Further, it is also unique among sensations in that it can evoke, and to some extent be invoked, by complex alterations in the psychosocial state of human beings. An understanding of the underlying complex biologic, psychological, and social factors is therefore critical to a rational and satisfactory approach to the patient with pain.

NEUROBIOLOGY OF PAIN

Anatomic Pathways

Peripheral Pathways Normally, visceral pain begins with the stimulation of the peripheral nerve endings (located in the serosal, muscular, and mucosal layers of the gut) of a special class of spinal sensory neurons (nociceptors) whose cell bodies lie in the dorsal root ganglion (Fig. 38-1). After leaving the viscus they innervate, these nerve fibers run with the sympathetic fibers (as mesenteric nerves) and pass through, without interruption, one of several prevertebral autonomic plexuses associated with the corresponding visceral artery (e.g., celiac, hepatic, superior mesenteric). Thereafter, the nociceptor fibers travel within the regional splanchnic nerve until they reach the paravertebral sympathetic chain. Here they finally part company with the sympathetic nerves, taking the white rami communicans to the spinal nerve and thereafter to their eventual termination in the dorsal horn of the spinal cord. Although nociceptive fibers, like other extrinsic afferent nerves, run together with the sympathetic nerves for a greater part of their length, they are not considered part of the sympathetic nervous system. The sympathetic system does not therefore convey pain, but it may participate in the sensitization of nociceptor peripheral terminals under certain conditions. [1] Similarly, most of the sensory information carried by the parasympathetic vagal pathways is physiological; however, observations suggest that these nerves may also serve a modulatory role in nociception as well as in the autonomic, behavioral, and emotional response to it. [2], [3]

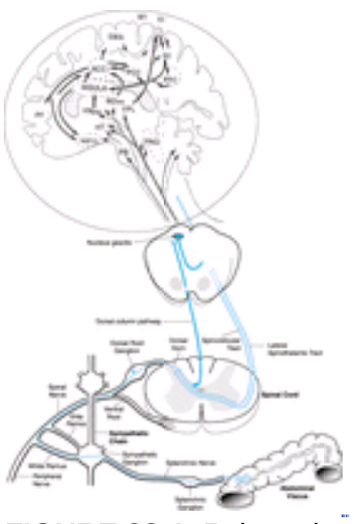

FIGURE 38-1. Pain pathways. (**Bottom**) Classic neuronal pathways, such as the spinothalamic and spinoreticular tract, mediating abdominal visceral pain sensation leave the dorsal horn, cross the midline, and ascend to higher centers. In the postsynaptic dorsal column pathway, nociceptive information from visceral organs is relayed to cells near the central canal. These postsynaptic dorsal horn cells send their axons in the midline of the dorsal column to synapse in the nucleus gracilis. The pathway then crosses the midline in the lower brainstem to ascend to the ventral posterolateral nucleus of the thalamus. (**Inset**) Schematic of ascending pathways, subcortical structures, and cerebral cortical structures involved in processing pain. *ACC,* anterior cingulate cortex; *AMYG,* amygdala; *HT,* hypothalamus; *MDvc,* ventrocaudal part of the medial dorsal nucleus; *PAG,* periaqueductal gray; *PB,* parabrachial nucleus of the dorsolateral pons; *PCC,* posterior cingulate cortex; *PF,* prefrontal cortex; *PPC,* posterior parietal complex; *S-1* and *S-2,* first and second somatosensory cortical areas; *SMA,* supplementary motor area; *VMpo,* ventromedial part of the posterior nuclear complex; *VPL,* ventroposterior lateral nucleus. (*Inset* from ref. [10].)

Spinal Connections After entering the spinal cord, afferent fibers make contact with neurons in the gray matter, which is conventionally described in terms of topographic regions or laminae. Whereas somatic nociceptor afferents mainly synapse in laminae I and II at their corresponding ipsilateral segmental level, visceral nociceptive afferents are more widely distributed, synapsing not only at more than one segmental level above and below the level of entry (up to six spinal segments), but also in several laminae at the same level, ipsilaterally and contralaterally. [4]

Ascending Pathways According to conventional teaching, spinal gray matter neurons mediating abdominal visceral pain cross the midline to the contralateral side and then travel cephalad within monosynaptic ascending pathways in the ventrolateral quadrant of the spinal cord (e.g., the spinothalamic, spinoreticular, spinobrachial, spinomesencephalic, and other tracts), to synapse within several thalamic and reticular formation nuclei of the pons and medulla [5] (see Fig. 38-1). However, findings also suggest that a prominent, and perhaps predominant, component of visceral pain may be relayed in the postsynaptic dorsal column (PSDC) of the spinal cord [6] (see Fig. 38-1). This pathway arises from spinal neurons adjacent to the central canal (lamina X) and ascends in the midline ipsilaterally. The PSDC pathway is polysynaptic in that there is at least one more relay station (the gracile nucleus for neurons originating in sacral spinal levels and in the cuneate and gracile nuclei for thoracic-level neurons) before it reaches a higher center. Neurons from these nuclei, in turn, project to the contralateral ventral posterior lateral

nucleus in the thalamus. Evidence for the functional importance of this pathway is derived from physiological studies as well as from patients with intractable pelvic cancer pain in whom a limited midline myelotomy to interrupt the PSDC resulted in complete or near-complete relief. [7], [8]

Supraspinal Structures and Circuits Involved in Pain Ascending pathways from the spinal cord relay pain to the thalamus and other subcortical organs and then to higher centers. From a functional perspective, these circuits can be viewed as either "sensory-discriminative" or "affective-cognitive." [9], [10] The former is responsible for precisely characterizing the noxious stimuli according to their location and nature. The latter is responsible for early autonomic and emotional responses accompanying pain including arousal, fear, and escape. Subsequently, it also invokes many important aspects of the clinical pain experience such as mood, attention, memory, and ability to cope and tolerate, as well as the characteristic feeling of "unpleasantness" and emotions about the long-term consequences of pain (e.g., "suffering"). Sensory-discriminative function appears to be served by laminae II and other regions in the neck of the dorsal horn and the ventroposterolateral and ventroposteromedial nuclei of the thalamus with projections to the somatosensory cortex. The affective-cognitive component of pain involves the superficial lamina I neurons, parabrachial region of the pons with projections to the ventroposterior inferior and the posterior division of the ventromedial thalamic nuclei, the central nucleus of the amygdala, the ventromedial hypothalamus, the insula, and the anterior cingulate cortex (ACC). Other structures are also involved in the maintenance of these circuits. One of these, the posterior parietal cortex, integrates visual and other sensory cues with nociceptive inputs from the somatosensory cortex and relays this information to the ACC. Another, the insular cortex, is concerned with monitoring the overall threat to the body and determines the destructive significance of pain. The insular cortex is also closely connected with the ACC, which, in turn, receives connections from the prefrontal cortex. Pain unpleasantness, if sustained, eventually engages areas in the prefrontal cortex, which appears to be the region mainly responsible for reflection on future implications of pain and planning and prioritizing the responses to pain as well as determining the secondary pain affect. The ACC therefore appears to occupy a critical position, integrating information about the immediate environmental threat from the parietal cortex with the emotional and behavioral response plans originating in the prefrontal cortex and, subsequently, directing attention and response priorities. In addition to those described previously, several other pathways also carry nociceptive information from the spinal cord to various regions in the brainstem. [11] Some of these regions, such as the reticular formation, may be involved in general arousal and motivational functions. Others, such as the parabrachial nucleus of the pons and the periaqueductal gray, are also well connected to the affective-cognitive circuits described earlier and together modulate the cognitive, emotional, behavioral, and autonomic responses to pain and the "unpleasantness" of the sensation. It is apparent therefore that a pain center as such does not exist in the brain. Instead, it may be more useful to view pain being perceived by a complex neural network (the *neuromatrix*), [12] with two components, one providing fine sensory information about the sensation and the other producing the emotional, cognitive, and behavioral response. This concept helps us to understand why chronic pain is such a complex phenomenon that can so profoundly affect an individual's sense of well being as well as to explain how a variety of physical and emotional stressors can affect the perception of pain.

Descending Pathways The functional connections between the spinal cord and higher centers run both ways (Fig. 38-2). Most of the descending pathways are inhibitory and may serve to provide negative feedback to strong painful input; however, excitatory connections also exist. One of the most important centers involved in descending modulation of nociception is the periaqueductal gray (see earlier), which connects caudally to the spinal cord by the nucleus raphe magnum and raphe spinal pathways. It also receives descending projections from several of the other structures involved in the affective-cognitive aspects of pain including the limbic system and hypothalamic regions and thus may assist in coordinating a "dampening" response. Other centers participating in descending inhibition include pontine nuclei such as the locus ceruleus and subceruleus. [5]

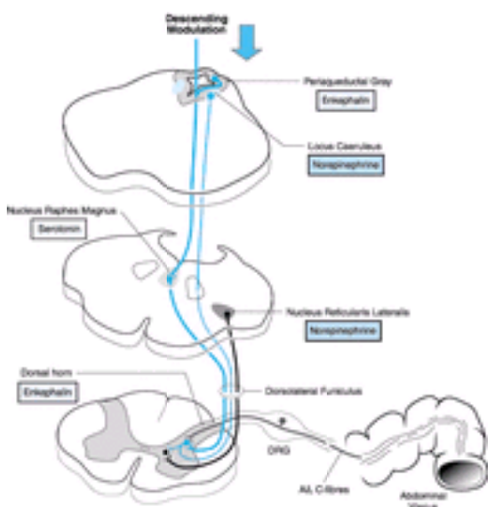

FIGURE 38-2. Descending pathways involved in modulation of nociception and their principal neuromediators (see text for details). (Adapted from Russo CM, Brose WG. Chronic pain. Annu Rev Med 1998;49:123.)

Cellular, Molecular, and Neurochemical Substrate of Pain

Nociceptive sensory neurons consist of slow unmyelinated C fibers and fast myelinated Ad fibers belonging to the primary afferent nociceptor [13] (Fig. 38-3). Both Ad and C fibers are polymodal and respond to both mechanical and thermal stimuli, with C fibers also responding to noxious chemical stimuli such as acid and capsaicin, the pungent component in hot chili peppers. Most visceral C-type nociceptors contain neuropeptides such as substance P (SP) and calcitonin gene–related peptide (CGRP) and are dependent on trophic support by nerve growth factor (NGF). [14]

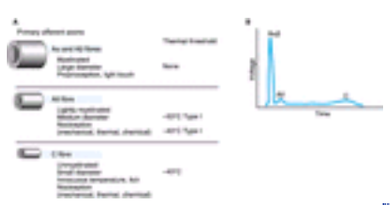

FIGURE 38-3. Different nociceptors detect different types of pain. (**A**) Peripheral nerves include small-diameter (Ad) and medium- to large-diameter (Aa,ß) myelinated afferent fibers, as well as small-diameter unmyelinated afferent fibers (C). (**B**) The fact that conduction velocity is directly related to fiber diameter is highlighted in the compound action potential recording from a peripheral nerve. Most nociceptors are either Ad or C fibers and have different conduction velocities (6–25 and approximately 1.0 m s^{-1}, respectively). (Adapted from ref. [13].)

The nociceptor has to perform three key tasks: transduction of noxious stimulus to an electrical signal, conduction of that electrical signal from the peripheral to the central end of the nociceptor, and, finally, encoding and relaying that signal to second-order neurons in the form of synaptically transmitted chemicals (Fig. 38-4). Signal transduction presents a unique challenge for nociceptors because, unlike other sensory stimuli (e.g., light), noxious stimuli can take one of a variety of diverse forms including heat, pressure, and chemical injury. In general, nociceptors convert noxious stimuli to an electrical response through specialized receptors. Some of these receptors are "promiscuous" in that they are capable of responding to more than one type of noxious stimulation. The best example of this is the receptor for capsaicin, vanilloid receptor 1 (VR $_1$). [13], [15] This receptor is expressed by nociceptive primary afferents, and it responds to and appears to integrate several noxious stimuli produced during tissue injury, including heat, local tissue acidosis, and several pro-algesic metabolites. Activation of the receptor results in a cationic, calcium-preferring current that leads to depolarization of the membrane. Acid and heat are both thought to function as endogenous ligands of this receptor, and evidence also points to a potential role for other biologically active compounds such as anandamide and related lipid metabolites. Further, other pro-algesic agents such as bradykinin or ATP, while independently capable of stimulating nociceptors, may also potentiate VR $_1$ responsiveness by lowering the threshold for activation to the range of normally nonpainful thermal stimuli (i.e., normal body temperature). This diverse portfolio of responses has led the current view of VR $_1$ as a "molecular integrator" of several chemical and physical stimuli that elicit pain.

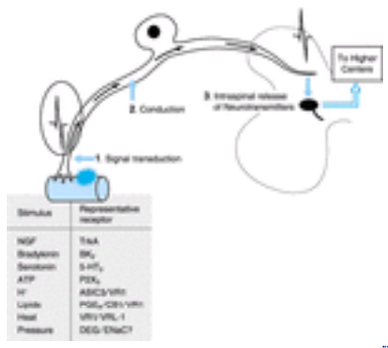

FIGURE 38-4. The primary nociceptor and its functions. Sensation from the peripheral organ is carried by fibers whose origin is in cell bodies within the dorsal root ganglia. The central projections of these neurons, in turn, relay information to second-order neurons in the spinal cord that transmit this information upward to the brainstem and higher centers by distinct pathways. This diagram illustrates the three basic components of pain signaling at the level of the first-order neuron in the nociceptive pathway. A painful stimulus is translated into electrical activity, which is conducted centrally and eventually results in release of neurotransmitters at the central synapses, with stimulation of second-order neurons. (Table data from ref. [13].)

In contrast to thermal and chemical receptors, little is known about the receptors capable of transducing mechanical stimuli. In addition to specific receptors responsive to physiological mechanical stimuli (e.g., the degenerin ion channel family), [13], [16] pressure changes in tissue may be signaled by the release of chemicals such as ATP, which then interact with chemical receptors such as the P2Y and P2X receptors. [17]

Thermal, mechanical, or chemical stimuli, acting through specific receptors, induce a change in the membrane potential of the nociceptor terminal called a *receptor* or *generator* potential. When this exceeds the threshold for activation, an action potential is triggered. Activation thresholds are under a variety of influences including the ionic concentration of the environment, as well as the activity of several intrinsic membrane currents (K^+, Na^+, Ca^{2+}). Attention has been focused on sensory neuronal voltage-gated Na^+ currents (VGSCs), particularly on the tetrodotoxin (TTX)-resistant channels, SNS (PN-3) and SNS2 (NaN), that are expressed predominantly in small sensory neurons. [18] TTX-resistant channels recover from inactivation more rapidly, and their presence is associated with the ability to fire long trains of action potentials and hence contributes to an increased excitability.

Once generated, action potentials are conducted centripetally to the spinal terminals of the nociceptors, where they initiate neurotransmitter (principally glutamate, SP, and CGRP) and thereby relay nociceptive information to second-order neurons. The early response to relatively mild stimuli is transmitted by glutamate acting on the a-amino-3-hydroxy-5-methylisoxazole-4-propionic acid (AMPA) and kainate ligand-gated ion channels. [19] If the stimulus is sustained or intense enough, it causes release of SP, which acts on the NK_1 receptor [20] to produce a correspondingly more intense postsynaptic response, an effect that, in turn, is boosted by another glutamate receptor, the *N*-methyl-D-aspartate (NMDA) receptor. CGRP, which is coexpressed with SP, also plays a role in nociceptive transmission, with effects that are less well understood. [21] Finally, other glutamate receptors such as the metabotropic (mGluR) receptors are also brought into play at higher levels of noxious stimulation and appear to play a critical role in central sensitization, along with the NK_1 and NMDA receptors (see later).

In addition to the “classic” neurotransmitters described earlier, several others may also be involved in spinal nociceptive processes. These include excitatory neuropeptides such as neurokinin A and brain-derived neurotrophic factor (BDNF), possibly inhibitory ones such as somatostatin and galanin, vasoactive intestinal polypeptide (VIP), and nitric oxide. [22], [23] and [24] Finally, many receptors that act at the peripheral ends of nociceptors (e.g., VR_1, P2X, and prostaglandin E_2 receptors), are also expressed receptors on the central ends, suggesting a role for their ligands in spinal modulation of nociception. [13]

The dorsal horn cannot be considered a simple way station, but rather a critical point where peripheral signals may be enhanced or attenuated by caudally directed neural connections from higher centers (see Fig. 38-2) before they are relayed cranially. Descending inhibitory neurons counteract the excitation from the periphery, mainly by the release of glycine and ?-aminobutyric acid (GABA). $GABA_B$ receptors act mainly presynaptically, whereas $GABA_A$ receptors, like glycine receptors, modulate dorsal horn neurons postsynaptically. [23] Although serotonin (5-HT) is also thought to be an important participant in descending inhibition, its effects are not straightforward, because of the multiplicity of receptors, some of them with conflicting actions on nociception. $5\text{-HT}_{1B/D}$ receptors probably mediate selective inhibition of nociceptive neurons, whereas other receptors have either mixed (5-HT_{2C}) or excitatory effects (5-HT_3). [23] A closely related inhibitory system involves norepinephrine, whose effects on nociception are largely mediated by a $_2$-adrenergic presynaptic receptors. [23] The spinal cord also contains high concentrations of opioid receptors (μ,d,?, in order of abundance), with the majority presynaptic in location, that is, on primary afferents. [22] Opioid activation of these receptors leads to a hyperpolarization of the neurons and inhibition of the release of excitatory neurotransmitters such as SP and glutamate. In this context, cholecystokinin (CCK) appears to share an intimate and complex relationship with opioids. CCK, acting through the CCK-B receptor, effectively antagonizes the analgesic effects of opioids. [25] Other mediators found in the dorsal horn with potentially antinociceptive effects include acetylcholine, adenosine, nociceptin, and a variety of others whose roles yet to be fully understood. [26]

Thus, the dorsal horn is the site of a complex interaction between a variety of neurotransmitters and their receptors derived from or located on afferent fibers, intrinsic dorsal horn neurons, or descending (modulatory) fibers. This interaction determines the nature and intensity of the signal that travels up the ascending pain pathway to higher centers.

PAIN AND SENSITIZATION

Most pain, if lasting more than a few minutes, occurs on a background of a potentiated or “sensitized” nociceptive system. [19] This is an extremely important concept to understand because it can help to explain many commonly encountered clinical situations. The stimuli that cause pain also produce tissue injury or inflammation; this, in turn, results in the local accumulation of several factors that amplify the activity of peripheral nociceptors as well activate previously dormant neurons (“silent nociceptors”). Tissue injury may also produce *phenotypic switching,* a phenomenon in which normally nonnociceptive fibers (e.g., Aß) display changes in neurotransmitter content and central innervation pattern that allow them also to participate in pain signaling. [27] Together, these phenomena are termed *peripheral sensitization* and lead to an “afferent barrage” that eventually produces central sensitization or an increase in the responsiveness of dorsal horn neurons. [28] The gain of the entire system is therefore reset upward, with the result that noxious stimuli now elicit a pain response that is much greater when compared with the normal state, a phenomenon termed *hyperalgesia.* A further characteristic of the sensitized state is called *allodynia,* a phenomenon in which innocuous or physiological stimuli are perceived as painful. These manifestations of sensitization are graphically illustrated in Figure 38-5. As an example, one can therefore postulate that patients with painful chronic pancreatitis exhibit pancreatic neuronal sensitization and may experience mechanical allodynia: pain in response to physiological changes in intraductal pressure, which would otherwise have not been perceived. Similarly, subsequent minor flares of inflammation in such patients could also cause the associated pain to be felt as severe, rather than mild (hyperalgesia).

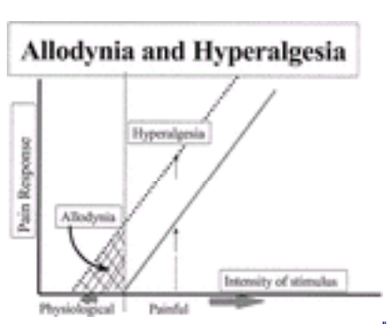

FIGURE 38-5. Basic concepts of pain sensitization, as illustrated in a theoretical stimulus-response curve. The *solid line* on the **right** represents a hypothetical control

population, whereas the *broken one* on the **left** represents the response in a sensitized population (e.g., patients with pancreatitis). The *broken vertical line* represents the threshold for painful stimulation in the control population. The sensitized population experiences pain in response to stimulation that is in the nonpainful (physiological range) for the control population, a phenomenon known as allodynia (*shaded area*). Hyperalgesia refers to a response to painful stimulation that is greater than the control population (*arrows*).

Pain sensitization results from both early posttranslational changes as well as later transcription-dependent changes in effector genes; these occur in both nociceptors and dorsal horn neurons. These changes can be initiated by a variety of factors in inflamed tissue including ions (K^+, H^+), amines (5-HT, histamine), kinins (bradykinin), prostanoids (prostaglandin E_2), purines (ATP), cytokines (tumor necrosis factor, interleukin-1 or 6), nitric oxide, and caloric activity (heat). [29] In the short-term, several of these agents, acting through specific receptors or cellular messengers, initiate a chain of events that lead to an increase in the phosphorylated state of critical ion channels, such as the nociceptor-specific TTX-resistant sodium channel SNS, thus enhancing excitability (Fig. 38-6). These early sensitization events are sustained and are further amplified by transcriptional up-regulation of several of the foregoing channel proteins hours to days later after the onset of tissue injury or inflammation. [19] This process is exemplified by the effects of NGF, whose levels are increased in variety of inflammatory states including chronic pancreatitis. [30] Increased tissue levels of NGF result in increased expression of various genes important for nociception (see Fig. 38-6), thus amplifying and perpetuating the sensitized state. [31] Similar paradigms of sensitization can be used to illustrate the processes underlying central sensitization, including early posttranslational and late, transcriptional events [19] (Fig. 38-7).

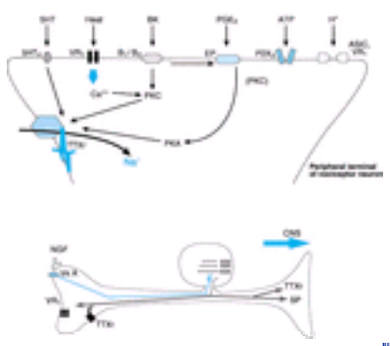

FIGURE 38-6. (**Top**) Peripheral sensitization: early events. Various biologic mediators comprising the inflammatory “soup” interact with specific receptors expressed on the peripheral ends of sensory neurons. Some of these (e.g., noxious heat) can activate neurons directly, whereas the majority can acutely sensitize the neuron, that is, make it more excitable by changes in intracellular calcium and phosphorylation states that result in greater activation of critical ion channels such as the tetrodotoxin-resistant sodium channel (TTXr). *ASIC,* acid sensing ion channel; *BK,* bradykinin; *5 HT,* 5-hydroxytryptamine; *PGE2,* prostaglandin E_2; *PKA,* protein kinase A; *PKC,* protein kinase C; *VR1,* vanilloid receptor 1. (**Bottom**) Model for “late” sensitization of sensory neurons. Neurotrophins such as nerve growth factor (NGF) are released in response to inflammation and cause up-regulation of genes critical for nociception such as ion channels such as VR1, TTXr (expressed in the periphery) and neurotransmitters such as brain-derived neurotrophic factor (BDNF) and substance P (SP) released centrally. (From Woolf CJ, Costigan M. Transcriptional and posttranslational plasticity and the generation of inflammatory pain. Proc Natl Acad Sci U S A 1999;96:7723.)

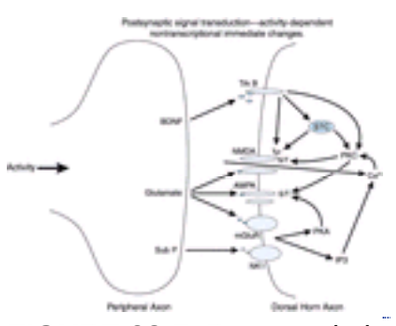

FIGURE 38-7. Posttranslational changes within dorsal horn neurons after release of transmitters from C fiber central terminals. These transmitters/neuromodulators act on receptors and ion channels in the dorsal horn to activate protein kinases that phosphorylate membrane-bound NMDA and AMPA receptors and alter their functional properties, increasing membrane excitability and thereby eliciting central sensitization. (From Woolf CJ, Costigan M. Transcriptional and posttranslational plasticity and the generation of inflammatory pain. Proc Natl Acad Sci U S A 1999;96:7723.)

Sensitization, Visceral Hyperalgesia, and “Functional” Pain Syndromes

The concepts discussed earlier provide a biologic basis for the perpetuation of the pain state in inflammatory visceral conditions such as pancreatitis and may also be valid in conditions such as irritable bowel syndrome (IBS) or other functional pain syndromes such as nonulcer dyspepsia. [24], [32] Patients with IBS experience pain at distention pressures or volumes that produce, at best, normal internal sensation in healthy volunteers (allodynia); they also experience more severe discomfort at noxious distention pressures or volumes (hyperalgesia). [24] Experimental evidence suggests that both inflammatory and motor events could theoretically set in motion a cascade of events leading to sensitization in this condition. Thus, mucosal inflammation accompanying gastrointestinal infections can produce visceral hyperalgesia by both peripheral sensitization and central hyperexcitability. [33] Motor events in the gut, if prolonged and abnormally excessive, may also result in a state of sensitization: repeated distention of the colon in humans (to 60 mm Hg) has been shown to increase the area of pain referral in the short term. [34] Further, significant numbers of patients develop IBS-like symptoms after a bout of acute infectious gastroenteritis. [35] According to one theory, the lack of overt inflammation or disruption of tissue architecture in patients with IBS is explained by the fact that the initiating event was transient but left persistent changes in its wake that resulted in peripheral and central hyperalgesia. In such a sensitized state, even normal contractile events could be perceived as painful (i.e., allodynia). Host factors and genetic susceptibility may determine which patients eventually develop the clinical syndrome.

Although there is reasonable evidence to suggest nociceptive sensitization in these syndromes, the relative contributions of central and peripheral factors remain known. A very active area of research includes functional imagines studies of the brain in response to noxious visceral stimulation. Although the implications of these findings remain to be fully understood, these studies are important in showing objective evidence of altered processing of noxious information in patients with functional bowel pain. [35]

NOCICEPTION, SUFFERING, AND ILLNESS BEHAVIOR: THE BIOPSYCHOSOCIAL CONTINUUM OF THE PAIN EXPERIENCE

It is important to understand that *nociception,* the detection of tissue damage by specialized transducers attached to C and Ad fibers, is not synonymous with pain, and increased afferent signaling to the central nervous system by itself is not necessarily enough to make a patient with chronic pain seek medical attention. Nociception can lead to pain *perception,* but this can also occur in the absence of nociception resulting from damage to the peripheral or central nervous system. Pain perception, in turn, can cause *suffering* (a combination of anxiety, fear, stress, uncertainty, and loss of loved objects). [36] *Illness behavior,* another important concept, reflects how pain is acted on by an individual person and results from a complex mixture of physiological (e.g., pain intensity/severity or associated features), psychological (e.g., mental state, stress, mood, coping style, prior memories or experiences with pain), and social (e.g., concurrent negative life events, attitudes and behavior of family and friends, perceived benefits such as avoidance of unpleasant duties) factors. [37] The biologic basis of these processed can be partially explained within the known framework of the nociceptive process described earlier. Thus, the well-known exacerbation of pain in patients with stressful life events may result from changes in central descending modulatory systems and perhaps as well by alterations in sympathetic discharge locally. [24]

It is important therefore to understand that the patient with chronic pain represents a dysregulation or dysfunction of a system that is, in effect, a continuum of

biopsychosocial factors (Fig. 38-8). In a given patient, the primary disturbance may disproportionately affect one component of the spectrum, and it is the wise physician's task to identify this without losing sight of the whole.

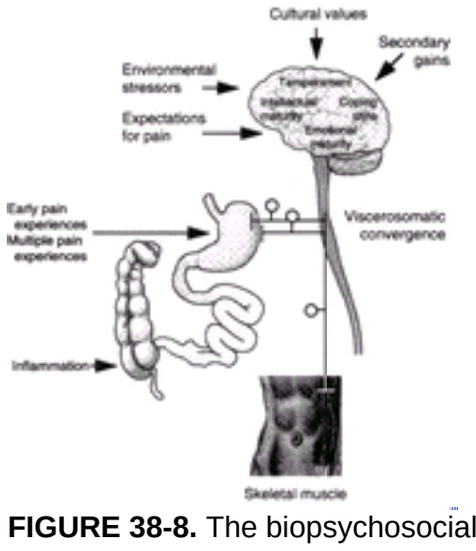

FIGURE 38-8. The biopsychosocial model of chronic abdominal pain. In this illustration, inflammation in the colon is shown as initiating pain and sensitization, providing nociceptive input to the central nervous system. Thereafter, psychological, developmental, and social/environmental factors alter the "experience" of pain. (From Hyams JS, Hyman, PE. Recurrent abdominal pain and the biopsychosocial model of medical practice. J Pediatr 1998;133:473.)

CLINICALLY IMPORTANT PHYSIOLOGICAL CHARACTERISTICS OF ABDOMINAL PAIN

Abdominal pain in the abdominal region can arise from the internal organs and their immediate peritoneal lining (visceral pain) or from the parietal peritoneum or the muscular and other layers of the abdominal wall (somatic pain). At the onset at least, visceral pain has certain distinct characteristics, reflecting the peculiar nature of visceral innervation. [28] First, it is not evoked equally from all organs (solid ones are less sensitive than hollow organs, and capsular or serosal coverings are more sensitive than the organs themselves), because not all structures receive equal nociceptive innervation. Second, visceral afferents are relatively few (as compared with somatic structures) and diverge extensively in the central nervous system, with the result that the pain tends to be poorly localized. Third, acute visceral pain is often accompanied by autonomic disturbances such as changes in blood pressure and heart rate, pallor and sweating, and visceral motor phenomena such as vomiting and diarrhea. In addition, intense emotional responses and anxiety can occur. Finally, and perhaps most importantly from a physician's perspective, most clinically significant forms of visceral pain are referred to somatic areas, a phenomenon that is discussed next in greater detail.

A patient with "pure" visceral pain is seldom seen by the physician, because this phase usually lasts only a few hours. When it begins, it is felt in the midline in the epigastric, periumbilical, or hypogastric regions, reflecting the ontogenic origin of the involved organ from the foregut, midgut, or hindgut, respectively (Fig. 38-9). Even observant patients may find it difficult to describe as a real pain, and they may perceive it as a deep and dull discomfort instead. This phase of pain is typically limited, and if the underlying insult persists, referred pain sets in later. Referred pain is perceived in overlying or remote superficial somatic structures such as skin or abdominal wall muscle, with the site varying according to the involved visceral organ. Referred pain is more helpful in determining the site of the underlying disorder than the original pure visceral pain, which tends to be perceived in the midline regardless of the organ involved (see Fig. 38-9). Further, referred pain is sharper and assumes several of the characteristics of pain of somatic origin.

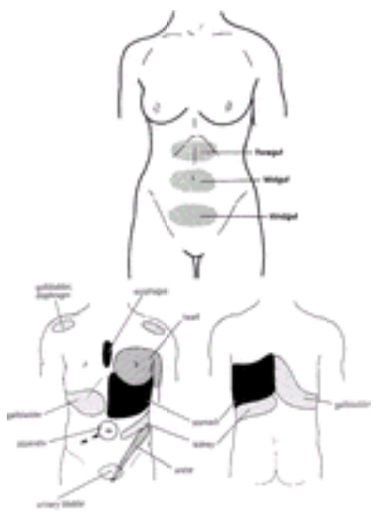

FIGURE 38-9. True visceral and referred pain patterns. (**Top**) Approximate levels of the abdomen where "true" visceral pain is felt, according to its source. (From Mulholland MW. Approach to the patient with abdominal pain. In: Yamada T, ed. Textbook of gastroenterology, 3rd ed. Philadelphia: JB Lippincott, 1999:826.) (**Bottom**) Important skin areas for referral of visceral pain. (From Snell RS. Clinical anatomy for medical students, 6th ed. Philadelphia: Lippincott Williams & Wilkins, 2000.)

Referred pain can occur without hyperalgesia of the somatic structure (simple irradiation) or can be accompanied by sensitization of these structures with resulting hyperalgesia. [38] The physiological basis of referred pain is incompletely understood, and no one theory is completely satisfactory. Nevertheless, familiarity with these theories is important to understand the clinical implications of these phenomena (Fig. 38-10).

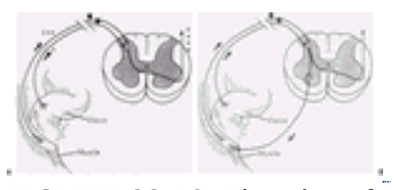

FIGURE 38-10. Theories of referred pain from a visceral organ, using skeletal muscle as the site of referral. (**Left**) The convergence-facilitation theory states that painful visceral insults produce an "irritable focus" in the corresponding spinal cord segment, thus facilitating or amplifying signals from somatic structures whose nerves converge on the same spinal neurons. According to this theory, therefore, the referred hyperalgesia is mainly central. (**Right**) The reflex-arc theory states that viscerocutaneous or visceromuscular reflexes traveling through the spinal cord induce neurogenic changes in the muscle or skin that result in a local painful state. According to this theory, referred hyperalgesia can be viewed as peripheral. (From ref. [38].)

CAUSES AND CLASSIFICATION OF ABDOMINAL PAIN

Gastrointestinal viscera are relatively insensitive to stimuli such as light touch, pinching, cutting, and even burning, compared with cutaneous and other somatic structures. [39] Instead, typical painful visceral stimuli include changes in length-pressure relationships within hollow organs (contractions and/or distentions), stretching of capsules, ligaments, or mesenteric attachments, inflammation with local production of sensitizing chemical and biologic agents, and ischemia, particularly when it affects visceral musculature. An appealing approach to the classification of pain, used widely in the somatic literature, is based on putative neurophysiological mechanisms. According to this, pain can be either *nociceptive* or *neuropathic;* [40] the former is the result of persistent stimulation of peripheral nociceptors by local injury and/or inflammation), whereas the latter is independent of nociceptor stimulation, implying changes in the pain pathways (either peripheral or central) that result in persistent but aberrant signaling. The pain of some acute gastrointestinal conditions such as pancreatitis or cholecystitis can be considered to be predominantly nociceptive. Somatic neuropathic pain syndromes include poststroke pain (central) or diabetic neuropathy (peripheral); whether there is a visceral counterpart remains unknown. The chronicity of pain is also important: acute pain serves a useful biologic function warning the organism of impending injury, whereas pain that persists beyond the expected course of an injury to heal appears to have outlived its utility and can result in significant secondary morbidity in the form of physical, emotional, or socioeconomic stresses on the patient, family, and society. [40] These issues can often so dominate the overall clinical picture that the pain is often dismissed by physicians as “psychogenic,” “functional,” “not organic,” or, worse still, “malingering.” In fact, most patients with chronic abdominal pain are not malingerers or hypochondriacs; however, their “psychosocial burden” can be overwhelming. The challenge for clinicians is to recognize and address this without losing track of the fundamental nature of the problem.

CLINICAL ASSESSMENT OF ABDOMINAL PAIN

History

A carefully taken history often holds the key to an accurate diagnosis of abdominal pain. Several critical features need to be elicited.

Site The phenomenon of referred pain (see earlier) renders it difficult to make a diagnosis of visceral pain by site alone. Nevertheless, such an approach is commonly used with most clinicians dividing the abdomen into four quadrants (Table 38-1). These general, if crude, inferences have been supported by studies using balloon insufflation in various organs in conscious patients. [39], [41] There is considerable overlap between the pain from various organs such as the bile ducts and the proximal small intestine. Further caution is warranted when dealing with patients with chronic painful conditions (including IBS) who often have distorted patterns of pain rendering when compared with healthy volunteers. [34]

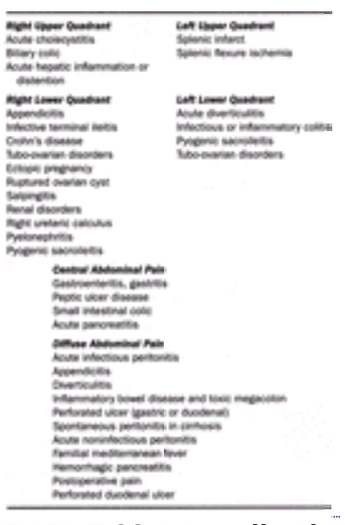

TABLE 38-1 Localization of Common Causes of Acute Abdominal Pain

Temporal Characteristics Among the most important of these characteristics are the nature of onset and the pattern of variation with time (intermittency and periodicity). Immediate pain is suggestive of an acute obstruction of a hollow viscus (e.g., bile duct obstruction by a stone), perforation (e.g., free perforation of a duodenal ulcer), or a catastrophic ischemic condition such as acute mesenteric ischemia. The more common situation is a relatively gradual onset of pain; the transition from a “glowing ember” to a “raging flame” may take hours or days, depending on the underlying condition and is typical of inflammatory conditions such as appendicitis, diverticulitis, pancreatitis, and cholecystitis. Abrupt cessation of pain can occasionally occur spontaneously and should suggest the relief of an obstructed organ (e.g., stone passage or resolution of a volvulus). In many cases, however, the pain is intermittent in that it wanes for varying intervals of time, only to wax to its original intensity subsequently. Such pain is typical of colic, usually intestinal in origin. Biliary pain, although traditionally also labeled as colic, shows less variability than commonly thought. *Periodicity* refers to a long duration (weeks to months) of pain-free intervals. Such periodicity is seldom regular (unless pain is pelvic and related to the menstrual cycle) and used to be characteristic of conditions such as peptic ulcer disease (in the days before effective therapy was available). Other examples include patients with recurrent urinary stones, patients with IBS (in which the pain often varies with periods of psychosocial stress and associated disturbances in bowel function), and a long list of less common conditions (Table 38-2).

TABLE 38-2 Some Causes of Intermittent Abdominal Pain

Character and Intensity of Pain These attributes are influenced by the social and educational background of the patient as well as past personal experience. This may account for the fact that rating the severity or describing the nature of the pain seldom helps in distinguishing the cause of the pain. For example, the pain associated with peptic ulcer disease has been described by such apparently contradictory terms as aching, gnawing, sharp, burning, tearing, and squeezing. Furthermore, patients often ascribe different meanings to common descriptors. As an example, many patients often associate colic with any diarrheal illness. From a medical perspective, however, the pain of colic refers to a characteristic wavelike buildup in intensity culminating in severe pain often associated with other symptoms such a sweating, nausea, and dizziness. [42] The pain of colic is the result of visceral obstruction and peristaltic contractions associated with increased intralumenal pressure and generally is similar in character regardless of the organ involved (Table 38-3).

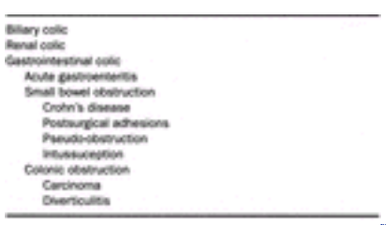

TABLE 38-3 Common Causes of Colicky Abdominal Pain

Relieving and Aggravating Factors A relationship between fluctuations in pain and physiological gastrointestinal activity suggests a link to a hollow viscus (rather

than musculoskeletal causes) and may point toward a specific diagnosis. Thus, pain with swallowing (*odynophagia*) almost invariably points to an esophageal lesion. The pain of duodenal ulcer tends to improve with food or antacid use, whereas gastric ulcer pain may be worsened by food intake. The specific nature of the food that either relieves or aggravates pain is seldom of diagnostic value, contrary to popular belief. Thus, the relationship between fatty foods and biliary pain or between spicy foods and peptic ulcer pain is dubious at best. [43] Relief after vomiting suggests a pyloric or proximal small bowel lesion. Colonic pain or distress may be relieved by a bowel movement, particularly in patients with IBS. Conversely, anorectal conditions such as proctitis or fissures may be aggravated by bowel movements. Retroperitoneal processes, including pancreatitis, tend to be somewhat relieved by maneuvers that increase the volume of this space (sitting up and bending forward). Visceral pain by itself often induces restlessness in the patient, but when parietal or somatic structures become involved, aggravation by motion, coughing, or straining is characteristically noted. Most inflammatory conditions of visceral organs are associated with varying degrees of systemic reaction including anorexia, malaise, and, perhaps, fever.

Physical Examination

The patient with acute abdominal pain is best dealt with as an emergency, possibly surgical in nature (see Chapter 40). In patients with more chronic pain, vital signs are less likely to be disturbed. Systemic examination may provide useful clues to the diagnosis such as purpural rashes, pallor, jaundice, edema, and other obvious signs of a more generalized disorder (Table 38-4).

PHYSICAL FINDING	RELATED CONDITION
Jaundice	Choledocholithiasis
	Gallstone pancreatitis
	Liver congestion or inflammation
Purpural or retinal cytoid bodies	Autoimmune process
Distended abdomen	Bowel obstruction
	Ascites
Palpable mass	Hernia
	Neoplasm
Focal neurological finding	Nerve root compression
	Vertebral body fracture
Anal fissure	Crohn's disease
Dark red "port-wine" urine	Acute intermittent porphyria
Occult blood in stool	Bowel inflammation
	Peptic ulcer disease
	Gastrointestinal cancer
Positive Carnett test	Abdominal wall hernia
	Cutaneous nerve entrapment
	Myofascial pain syndromes
	Rectus sheath hematoma
	Rib tip syndrome

Note: From Zackowski SW. Chronic recurrent abdominal pain. Emer Med Clin North Am 1998;16:877.

TABLE 38-4 Clues to the Etiology of Abdominal Pain on Clinical Examination

Subsequently, a careful examination of the abdomen is made. It begins with inspection: note is made of surgical scars and abdominal distention. This is followed by palpation, which should be performed with warm hands and should begin away from the area of maximal tenderness. This will often identify localized masses, free peritoneal fluid such as ascites, or areas of tenderness. It can also determine whether there is localized or diffuse peritoneal inflammation. Eliciting rebound tenderness, a staple of medical school teaching, is generally to be discouraged because not only may it be unreliable, [44] but also, because of its painful nature, it may compromise the patient's further cooperation. Auscultation of the abdomen may help in ascertaining the general state of the bowels (it is seldom helpful to auscultate all four quadrants). Peritonitis is associated with hypoactive or absent bowel sounds; hyperactive sounds may be heard in patients with infectious gastroenteritis. Infrequent, prolonged rushes of high-pitched or "tinkling" peristalsis are often heard over the distended loops of bowel seen with intestinal obstruction.

Rectal examination in male patients and pelvic examination in female patients may occasionally reveal signs of local peritonitis in the area. The presence of occult blood in the stools may provide a clue to an intralumenal lesion such as peptic ulcer.

When no obvious clinical or laboratory clues to a specific disease process are seen, the Carnett test may help to determine whether chronic intermittent abdominal pain arises from the abdominal wall or has an intra-abdominal origin. [45], [46] If a tender spot is identified, the patient is asked to raise his or her head, thus tensing the abdominal musculature. If there is greater tenderness on repeat palpation, the Carnett test is positive and suggests a cause in the abdominal wall (Table 38-5). Conversely, diminished tenderness may suggest an intra-abdominal process.

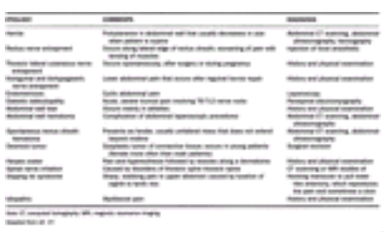

TABLE 38-5 Etiology of Abdominal Wall Pain

Differential Diagnosis

The differential diagnosis of "abdominal pain" is immense. Familiarity with common gastrointestinal diseases and their natural history is a prerequisite for an accurate approach to the patient, and clinical suspicion should form the basis for further testing. These diseases are dealt with in their entirety in other chapters; nevertheless, it is important to point out certain specific clues to such conditions.

Gastroduodenal Pain Gastric pain is usually felt in the midepigastric region but can also be perceived in the left upper quadrant and occasionally in the chest. [39] Peptic ulcers are one cause of *dyspepsia,* which is syndrome of epigastric pain, discomfort often accompanied by bloating, and early satiety, as well as other symptoms. Many conditions can cause a similar clinical picture including disorders of the pancreas or biliary tree, gastric malignant disease, gastroesophageal reflux, and use of several medications including antibiotics. When no underlying cause is found, the term *nonulcer* or *functional dyspepsia* is used. This is dealt with extensively in Chapter 30.

Biliary Pain Biliary pain, whether arising from the gallbladder or the bile duct, is felt as a dull sensation in the midline (epigastrium) initially; with the development of inflammation or if the pain continues, it is often referred to the right upper quadrant and occasionally to the right infrascapular region. Acute obstruction of the biliary tree is almost always painful, although not necessarily "colicky" in the true sense. A gradual dilation of the bile duct, as from a distal ampullary or pancreatic cancer, may remain painless throughout its course.

Pancreatic Pain Pancreatic pain usually either results from inflammation (acute or chronic) or is caused by a neoplasm. The pain is typically felt as severe and deep in the midline, and it is often associated with referred pain in the left upper quadrant. It can also radiate to the back, reflecting the proximity of the pancreas to the underlying vertebral column. Further, peripancreatic inflammation can often track to different locations in the abdomen, causing pain in remote sites, including the lower abdomen or chest.

Liver Pain Liver pain results from acute stretching or distortion of the liver (Glisson capsule), which can be caused by inflammation, vascular engorgement, or rapidly expanding lesions under the surface. [39] Deep parenchymal lesions of the liver, including tumors, are typically painless, as are chronic processes such as simple fatty liver or chronic viral hepatitis.

Splenic Pain Splenic lesions cause pain only when the capsule is stretched. The usual causes of splenomegaly (hematologic disorders and malignant diseases, portal hypertension) develop slowly and are usually painless. More acute causes including trauma can, however, result in pain that is felt in the left upper quadrant.

Small Bowel Pain Small bowel pain can result from nonobstructing lesions such as inflammation, ulcers, or neoplasms. It is aggravated by food intake and is typically felt as a dull periumbilical or other midline discomfort; occasionally, it can be felt in either flank or in the back. [39] A more dramatic form of small bowel pain is intestinal colic, which reflects intense muscular activity usually proximal to an obstruction but also seen in patients with acute gastrointestinal infections. Ischemia, acute or chronic, can also cause small bowel pain, including the rare but classic “abdominal angina” seen with chronic obstructing vascular lesions of the intestine, resulting in avoidance of eating because of the aggravation of pain.

Large Bowel, Rectal, and Pelvic Pain The large bowel has dual nociceptive innervation. Pain afferents, through the hypogastric nerves, run with sympathetic nerves to the spinal cord at T10-L2; in addition, they course with parasympathetic pelvic nerves to spinal cord S2-S4. [39] Inflammatory or neoplastic lesions of the colon can cause pain, which can be similar to small bowel pain, particularly if it arises in proximal segments. Pain in the distal colon is felt in the lower abdomen, reflecting its anatomic origin. Again, unless referred pain sets in or the parietal peritoneum is involved, the pain is felt as dull and in the midline. Despite the origin of the word, colic is usually not a typical manifestation of obstructing lesions of the colon, because this organ is relatively fixed and is capable of dilating significantly. However, inflammatory or infectious lesions of the colon can give rise to what is often described by patients as “spasms”, which typically are associated with and are usually relieved (at least temporarily) by bowel movements. *Rectal lesions* can also cause the symptom of *tenesmus,* a term that signifies frequent and painful desire to evacuate the bowel that is often not successful or leaves the patient with a feeling of incomplete evacuation. In this regard, it is analogous to the urinary symptom of strangury. It usually signifies proctitis (idiopathic or infectious) but can also be caused by rectal cancers or occasionally thrombosed hemorrhoids. Another peculiar and incompletely understood rectal syndrome is that of *proctalgia fugax,* which is a transient, but often recurrent and usually severe ache in the rectal region that typically wakes the patient up at night. Pain arising in the lower abdomen may also arise in the pelvic organs; such pain is often difficult to distinguish from colonic pain unless it is accompanied by disturbances in menstruation or by dyspareunia (painful coitus). However, in many instances no obvious cause is found, and such patients present as much of a dilemma to gynecologists as they do to gastroenterologists.

Abdominal Wall Pain Pain arising primarily in the abdominal wall can result from a heterogeneous and poorly defined group of conditions whose description remains largely anecdotal (see Table 38-5). [47] The diagnosis is suggested when the pain is superficial, when it is localized to a small area that is usually significantly tender, when it is associated with dysesthesia in the involved region, and when Carnett’s sign (see earlier) is positive. If there is an obvious scar from previous surgery or injury, a diagnosis of entrapment neuropathy (with or without a “neuroma”) is often entertained. In the absence of a scar, the pain is postulated to arise from “myofascial trigger points” thought to represent abdominal cutaneous nerve entrapment resulting from a fibrous ring in the rectus muscle. [48] Other causes of such pain include epigastric hernias that typically occur in patients lifting heavy weights, possibly resulting in a tear in the linea alba in the midline. These hernias are associated with the protrusion of peritoneal fat (epiplocele) or abdominal wall fascia but rarely become large enough to permit entry of a hollow viscus. Although they have been associated with considerable pain, their prevalence and contribution to chronic abdominal wall syndromes are unclear. Spigelian hernias, between the external and internal oblique muscles, can also produce similar pain, but this is at the level of the arcuate line. “Trigger” point injection with local anesthetics is popularly regarded as a useful method to distinguish abdominal wall pain from that of visceral origin. [49] However, this can be misleading if the referred component of a visceral pain has dominated the clinical picture. Such a phenomenon may also account for some of the observed inaccuracy in the predictive value of the abdominal wall tenderness test. [50]

Rare and Obscure Causes of Abdominal Pain Rare causes of abdominal pain [51] (Table 38-6) include disorders that primarily affect visceral nerves rather than the organs themselves, such as acute intermittent porphyria, chronic poisoning with lead or arsenic, or diabetic radiculopathy. Women taking oral contraceptives may experience mysterious attacks of abdominal pain that can sometimes be related to mesenteric venous thrombosis.

TABLE 38-6 Rare or Obscure Causes of Abdominal Pain

Diagnostic Workup

Even though acute abdominal pain is traditionally equated with a surgical diagnosis, fewer than 5% of adults presenting with acute abdominal pain will require hospital admission, and even fewer will ever need surgery. [52] Indeed, in a substantial proportion of patients with acute abdominal pain, no definite abdominal disorder will be found to account for the symptoms. [53] Similarly, in the absence of obvious clinical or laboratory clues, it is relatively unusual to uncover a new pathophysiological basis for patients with chronic abdominal pain who have previously been seen by other physicians. [54] In the absence of specific indications, further diagnostic tests (e.g., endoscopic retrograde cholangiopancreatography) are rarely helpful in the patient with chronic abdominal pain lasting longer than 6 months. [55] Laparoscopy is often considered in such patients, particularly if a clinical suspicion of “adhesions” exists. However, adhesions are very common in women, even in the absence of prior surgery, [56], [57] and are found in equal proportion in patients complaining of pelvic pain and those with other complaints. [58] Laparoscopy for chronic pain for adhesions or other lesions therefore seldom leads to a specific diagnosis and even less often results in a change in management. [59]

If enough tests are done, there is a considerable statistical probability of uncovering some abnormalities. However, these abnormalities can be minor and of questionable relevance to the patient’s complaints (e.g., minimal changes in the secondary branches of the pancreatic duct). Before acting on these abnormalities, the prudent physician must carefully reevaluate the patient (Table 38-7) and consider the possibility that the patient may have functional bowel syndromes including the functional abdominal pain syndrome (FAPS).

Clinical Issues
Does the patient:
Have pain that has persisted for 3 months or longer?
- Despite appropriate interventions?
- In the absence of progressive diseases?

Have unrealistic expectations of
- Health care provider?
- Treatment offered?

Complain about previous health-care providers?
Have a history of substance abuse?
Display pain behaviors?
- Grimacing
- Rigid or guarded movement

Legal and Occupational Issues
Is litigation pending?
Is the patient receiving disability compensation?
Was the patient employed before the onset of the pain?
Is there a job to which the patient can return?
Does the patient have a history of frequent changes in occupation?

Psychosocial Issues
Do other family members also suffer from chronic pain?
Excessive depression or inappropriate moods present?
Major stressful event before onset of pain exacerbation?
- High levels of marital or family conflict?

History of childhood abuse or other sexual trauma?
Activities given up because of pain?
- Plans for renewed or increased activities if pain is reduced?

Note: Adapted from Turk DC, Okifuji A. Pain: assessment of patients’ reporting of pain: an integrated perspective. Lancet 1999;353:1784.

TABLE 38-7 Questions to Assess a Patient with Pain: Red Flags for Referral to Specialists in Functional Bowel Disease

FUNCTIONAL ABDOMINAL PAIN SYNDROME

It has become evident that patients whose chronic abdominal pain remains unexplained despite a careful evaluation share enough common features to warrant consideration of a distinct syndrome, the FAPS [60] (Table 38-8). The lack of association with physiological gastrointestinal events such as eating or defecation distinguishes it from other functional disorders such as IBS. FAPS is relatively uncommon, found in only about 2% of respondents, and appears to increase with age;

women account for more than two thirds of the patients. [61] Nevertheless, patients with FAPS are heavy users of medical resources and have high morbidity. [62]

At least 6 months of 1. Continuous or nearly continuous abdominal pain, 2. No or only occasional relationship of pain with physiological events (e.g., eating, defecation, or menses); and 3. Some loss of daily functioning; and 4. The pain is not feigned (e.g., malingering); and 5. Insufficient criteria for other functional gastrointestinal disorders that would explain the abdominal pain.

TABLE 38-8 Diagnostic Criteria for Functional Abdominal Pain Syndrome

Etiology and Pathophysiology

Psychological, social, and cultural factors play a major role in the presentation of these patients. Like those with other more "organic" forms of chronic pain such as pancreatitis or cancer, patients with FAPS frequently suffer from depression, anxiety, sleep disturbances, withdrawal, decreased activity, fatigue, loss of libido, and morbid preoccupation with the chronic pain. [63], [64] and [65] These patients are often functionally impaired at many levels including work, family, and social settings. [66] Unlike patients with other forms of chronic pain, however, many of these patients have experienced abdominal pain in childhood, and they often have relatives with past or present abdominal pain. [66] A history of previous physical or sexual abuse is very frequent, reported in up to one half of these patients in some series. [67] The onset of pain may also be associated with the death of a relative, spouse, or other important figure. [62]

Illness behavior may also be aberrant in these patients. Pain may serve many functions in these patients, including a means of atonement, derailment of success, and a morbid replacement for loss of relationships. Finally, it is possible that because of positive reinforcement, actions that mitigate pain, such as decreasing physical activity or the use of medications, will often continue even after the inciting event (e.g., the acute pain symptom) is resolved. At the same time, other behaviors (e.g., appearing uncomfortable and helpless, not reporting for work) may be reinforced by many of the outcomes that result from the gains the patient may feel he or she is obtaining. These gains may include increased or renewed attention and empathy from within the family structure, avoidance of the stresses associated with work and family responsibilities, and monetary compensation. Thus, in susceptible patients, a set of behaviors becomes established that contributes to the maintenance of chronic pain.

Although our understanding of FAPS is rudimentary at best, it is helpful to view the contribution of these factors in terms of the biopsychosocial paradigm discussed previously in this chapter. In this context, FAPS can be viewed as representing a dysfunction of perception, coping, or response strategies resulting in a patient who has clearly lost a sense of well-being to pain.

Clinical Features

Chronic abdominal pain may occur in bouts lasting from several hours to days or weeks, but even between severe exacerbations, there is residual pain, albeit not as severe or incapacitating. The abdominal pain is often described in vague terms, sometimes through unusual, idiosyncratic, or even bizarre language. [62] The lack of association with physiological events such as eating or defecation and the maintenance of weight also suggest FAPS. Although nocturnal pain is often assumed to be "organic," functional pain may also sometimes awaken patients from their sleep. [68] Pain in locations other than the abdomen is also common in these patients, as are various somatic complaints. [69], [70]

Physical examination provides many pointers to this diagnosis but is not itself conclusive. Suggestive clues include absence of the autonomic activation (e.g., tachycardia, diaphoresis), inconsistent tenderness, discrepancy between tenderness elicited with pressure from the stethoscope and that from the examining hand, clutching of the physician's arm during the examination, and the "closed eyes sign" (characterized by the patient's keeping his or her eyes closed, often with a fixed, beatific smile, during abdominal palpation). [71] Other clues at the time of the interview may provide insight into social impairments contributing to the clinical presentation. These include the constant presence of a spouse or parent who assumes responsibility as a mediator between the physician and the patient, suggesting family "enmeshment" that may be contributing to illness behavior.

Management

A gastrointestinal cause of the symptoms is rarely found through subsequent investigations (including surgical exploration) or follow-up visits in these patients. [70], [72] Yet, many physicians feel compelled to embark on an ever more invasive and expensive series of tests in hope of uncovering the magical but elusive explanation for the patient's symptoms. Such activity ("furor medicus" [73]), in fact, may be counterproductive because it reinforces the patient's conviction that there is something wrong to account for the pain that, if only found, could be successfully corrected. For this and other reasons, pain may become a central feature in the lives of these patients, leading to a "pain career." [62] Often, when a "cause" is discovered, it is not clear whether it is truly the source of the pain, an incidental finding or epiphenomenon, or a consequence of the treatment, often surgical, used in an attempt to treat the original complaint. [74] In most of these cases, the presumed cause of pain will have been diagnosed and treated, only to see the pain remain or a new type of pain to manifest itself elsewhere. Patients with chronic intractable abdominal pain are rarely substantially free of pain after one or more years of follow-up, [62] emphasizing the importance of focusing on adaptation to the pain rather than cure. Given our current state of knowledge about this syndrome, it is unrealistic to talk about a cure for FAPS. Palliation is therefore an appropriate goal, and, in most patients, it is achievable. In general, the therapeutic approach to FAPS is similar to the multifactorial approach to other forms of chronic pain described later, with greater emphasis on the psychosocial dimensions.

TREATMENT OF ABDOMINAL PAIN

Whenever possible, healing of the underlying tissue insult or injury provides the most satisfactory approach to the control of pain. In many patients with pain of suspected gastrointestinal origin, however, the underlying condition may not be reversible or even identifiable. Pain management then often becomes the primary therapeutic issue and requires familiarity with some general principles. In patients with acute pain, once the need for immediate surgical exploration is ruled out, attention is quickly turned toward effective pain relief, typically in the form of potent opiate drugs such as morphine (5 to 10 mg) or the equivalent dose of meperidine. Although analgesics are often withheld in patients with acute abdominal pain for fear of "masking" the diagnosis, this concern has not been validated by any scientific studies and probably dates to the era when high doses of morphine (30 mg or more) were used. [75] If ongoing pain relief is required over a period of several days, narcotics are best administered using patient-controlled devices. Although intermittent use of narcotics (either as needed or at fixed intervals) is commonly practiced, inadequate dosing or frequency can lead to unnecessary patient discomfort.

Patients with chronic abdominal pain present a far greater challenge. The multifactorial and complex nature of chronic pain has made it clear that no single set of drugs, skills, or techniques can address all the issues that affect the quality of life in most patients. Ideally, these different dimensions require a multidisciplinary approach, requiring the help, at various times or in parallel, of pain-management physicians (internists, anesthesiologists, neurologists, or neurosurgeons), clinical psychologists, nurses, and, often, vocational rehabilitation specialists and nutritionists. However, the disadvantage of such an approach, even when available, is that it may lead to fractionated care; this, in turn, can lead to poor compliance on the patient's part when faced with therapeutic aspects that may be initially difficult to accept. Therefore, it is important for the gastrointestinal physician to remain engaged as the patient's gatekeeper; this role also requires a broad understanding of the management of chronic pain. Such knowledge will facilitate appropriate referral to other specialists, promote a more meaningful interaction with them, and maintain a strong and supportive patient-physician relationship, instead of leading to feelings of abandonment and other negative attitudes. A critical factor in the success of this role is the establishment of a relationship with the patient over several visits, [76] while providing reassurance and empathy. A successful outcome, although still difficult, is almost impossible without such a relationship firmly in place. Because many patients may have chronic abdominal pain or functional bowel syndromes, it is very important for the practicing gastroenterologist to understand the interaction between psychosocial factors and disease in producing gastrointestinal symptoms and illness-related behavior. These aspects are discussed in greater detail in Chapter 29.

Pharmacological Management of Chronic Pain

Narcotic Analgesics Gastroenterologists are loath to use traditional nonsteroidal antiinflammatory drugs, for fear of inducing gastrointestinal side effects, as well as the general feeling that they are less effective in the treatment of chronic abdominal pain than in the relief of acute pain. [77] Indeed, when mild pain chronic pain necessitates analgesic use, these drugs are often bypassed in favor of the weak opioids, propoxyphene or codeine. However, neither of these drugs is particularly potent, although their efficacy be enhanced with the addition of acetaminophen. [78] More severe pain requires stronger analgesics such as morphine, methadone, or transdermal fentanyl. Opioids are thought to be more effective in nociceptive pain (see earlier) than in neuropathic pain, which may require considerably higher doses. Meperidine is generally believed to be the drug of choice for patients with pancreatitis because of its lesser tendency to cause sphincter of Oddi spasm; however, this has been shown to be true only at subanalgesic doses. [79], [80] Because meperidine is more likely to produce other side effects, it is seldom used for chronic pain management. Opioid analgesics share a common adverse effect profile in general: constipation, nausea, sedation, respiratory depression, and depression of gastrointestinal motility when used on a long-term basis ("narcotic bowel"). However, the problem most physicians fear is the potential for addiction. Nevertheless, in patients with diseases such as chronic pancreatitis, these drugs can and should be used judiciously, using fairly rigid guidelines or protocols for narcotic use (Table 38-9). Patient selection remains of paramount importance; the use of narcotics in patients with functional bowel pain is hardly ever a good idea.

1. The patient has a clear understanding that opioids are being used for a limited term in the first instance.
2. Only one practitioner takes responsibility for the opioid prescription.
3. Opioid prescription is contingent on certain agreed obligations or goals being met by the patient, such as return to work or alteration of inappropriate behaviors. This could take then form of a written contractual arrangement.
4. Unauthorized demands for emergency injectable opioids will not be tolerated, although some provision can be made for "rescue analgesia" for brief exacerbations of pain.
5. The patient understands that opioid dosage compliance will be checked at various random intervals, which may include drug screens and blood samples.
6. Physicians must be prepared to terminate the arrangement if the goals are not met or if there is evidence of misuse, even though this may lead to a confrontational meeting with the patient.

Note: From Gourlay GK. Clinical pharmacology of the treatment of chronic non-cancer pain In: Committee IASP, ed. Pain 1999: an updated review. Seattle: IASP Press, 1999:433.

TABLE 38-9 Guidelines for Narcotic Use in Patients with Chronic Noncancer Pain

Antidepressants Tricyclic antidepressants (TCAs) have a proven track record in the management of chronic functional visceral pain. A metaanalysis of antidepressant treatment for functional gastrointestinal disorders concluded that these agents were quite effective, with an NNT of 3.2 (i.e., three patients needing treatment for one patient to experience symptom improvement) and a relatively large improvement in pain (about 0.9 standard deviations). [81] However, these drugs have not been fully embraced by either gastroenterologists or their patients. The latter often fear being stigmatized as having "mental" problems, a misperception that is perpetuated if a clear rationale for the use of these drugs is not presented by the prescribing physician. Effective analgesic doses of these drugs are significantly lower than those required to treat depression, and there is reasonable evidence to conclude that the beneficial effects of antidepressants on pain occur independently of changes in mood. However, in this regard, diminution of anxiety and restoration of mood and sleep patterns can be considered desirable. Most of these drugs act on the presynaptic pump, inhibiting the uptake of the neurotransmitters (principally serotonin and norepinephrine) and hence increasing their availability in the synaptic cleft. The principal mode of action is believed to result from modulation of central nervous system monoamine neurotransmitter levels, although a peripheral mechanism, possibly involving an opiate-like mechanism, may also contribute. [82] TCAs are well absorbed after oral administration, undergo extensive first-pass hepatic metabolism with long elimination half-lives (measured in days and weeks), and have active metabolites, some of which are also marketed as drugs. In addition to the putative analgesic effects on monoamine uptake, several of the older agents have anticholinergic (muscarinic) and antihistaminic (both H_1 and H_2) properties that contribute to their side-effect profile. Other serious side effects include cardiac conduction problems including a prolonged QT interval and dysrhythmia. Selective serotonin reuptake inhibitors (SSRIs), such as paroxetine, sertraline, and fluoxetine, which are currently the mainstay in the treatment of depression, have fewer side effects and have been advocated particularly for patients with functional constipation because these drugs can increase bowel movements and can even cause diarrhea. However, they have been less well evaluated in the management of pain than TCAs; at the present time, the somatic pain literature suggests that the efficacy of these agents for chronic pain is equivocal at best. [83] Newer antidepressants may hold more promise in this regard. [83] These include the serotonin and norepinephrine reuptake inhibitors such as venlafaxine, which inhibits the uptake of both norepinephrine and serotonin almost equally. Before antidepressants are prescribed, it is important to assess the psychological profile of the patient, because this may be important in determining the choice of therapy. If the patient is depressed, then it may be more appropriate to use full antidepressant doses of a drug that also has analgesic properties. This could be either a TCA with a low side effect profile or perhaps one of the newer agents discussed earlier (not an SSRI). If the patient is already taking an antidepressant, but this does not have proven analgesic activity (e.g., an SSRI), consideration should be given to switch to one that does. If the patient is not depressed, it is critical to spend some time explaining the scientific rationale for the use of antidepressants, with an attempt to separate the analgesic effects clearly from the antidepressant effects. The most appropriate agent to start with in this setting is one of the TCAs. A typical regimen would start with 25 mg of amitriptyline or nortriptyline (an even lower dose such as 10 mg may be considered in the elderly patient). This is given at night and will almost immediately begin helping with disturbed sleep pattern that often accompanies chronic pain. Daytime sedation may occur, but tolerance develops rapidly. Tolerance to the antimuscarinic effects may take longer, and it is important to advise the patients about this. In the absence of significant side effects, the dose of the antidepressant is gradually increased until adequate benefit is achieved or the upper limit of the recommended dose is reached. It is also important to tell the patient that the analgesic effect may take several days to weeks to develop and that, unlike conventional analgesics, the drug is not to be taken on an as-needed basis but on a fixed schedule. A trial of at least 4 to 6 weeks at a stable maximum dose is recommended before the drug is discontinued. If a particular TCA is clearly not found to be effective under these circumstances, one may consider switching to an antidepressant of another class, such as nefazodone, mirtazapine, or venlafaxine.

Other Neuropsychiatric and Miscellaneous Drugs Unlike the antidepressants, the use of neuroleptics (fluphenazine and haloperidol), antiepileptic drugs (phenytoin, carbamazepine, gabapentin, valproic acid, and clonazepam), clonidine (an a_2-adrenergic agonist), baclofen (a $GABA_B$ receptor agonist), and the NMDA receptor antagonists ketamine and dextromethorphan in chronic pain management is controversial, with equivocal evidence of efficacy and the lack of a clear biologic basis. [84], [85], [86], [87] and [88] Although the rationale for the use of these drugs is appealing (see the discussion earlier in this chapter), their use in gastrointestinal pain disorders is limited by gastroenterologists' lack of familiarity with these agents, inadequate or no controlled studies, and the side effects. Finally, mention must be made of the use of benzodiazepines, which are frequently prescribed for patients with chronic pain including insomnia, anxiety, and muscle spasm. Although these drugs are useful in these settings for short-term use, there is a significant risk of dependence on these drugs, and there is little if any evidence that they have any real analgesic effect.

Neural Blockade

Theoretically, interruption of the pain pathways should provide relief of pain that is peripheral in origin. This has led to the development of various techniques, both for diagnostic and therapeutic purposes. [89] However, interruption of the peripheral ends of the nociceptive nerves may not relieve pain if impulses continue to be generated at the proximal end, including the dorsal root ganglia. Further, if interruption of peripheral input is incomplete, altered spinal processing of nonnociceptive fiber input will continue to cause allodynic pain (see earlier). Even if interruption is complete, a component of central sensitization could be present in certain pain states that could result in continuous pain signaling despite the absence of peripheral input. Variability in these biologic factors, combined with inconsistencies in blockade techniques, may explain why, for instance, celiac plexus blocks may work for pain arising from pancreatic cancer but are generally ineffective in patients with chronic pancreatitis (see later).

Despite these limitations, it is clear that neurolytic techniques are valuable for certain subsets of patients, particularly those with cancer; celiac neurolysis may be effective in up to 85% of patients with pancreatic cancer pain and in about 70% of patients with pain from nonpancreatic cancers. [90] By contrast, use of these techniques to relive nonneoplastic pain, such as the pain of chronic pancreatitis, is not routinely recommended because of low efficacy (50% or less) and the short duration of relief (around 2 months) even in those patients who initially respond. [91]

Techniques of neurolysis vary considerably with respect to choice and concentration of agents (e.g., alcohol or phenol), method of injection (guided by plain radiography, fluoroscopy, computed tomography, or ultrasound). Endoscopic ultrasound has been used in patients with pancreatic cancer pain, with results that appear comparable to those of standard percutaneous techniques. [92] There appears to be little basis to favor one particular technique over another, and the choice of procedure should be individualized depending on local expertise and the patient's preference.

Local neural blockade has been used with good effect in some patients with chronic pain secondary to abdominal wall causes. [47], [93] Once a trigger point has been identified by digital examination, a small amount of lidocaine (1%) is injected at site of greatest tenderness elicited by the tip of the needle. Although the response may be short-lived, this can serve as a valuable therapeutic trial. Further, many patients obtain long-lasting relief after one or two injections alone. For patients whose relief is temporary, a 1:1 mixture of lidocaine and steroids (e.g., triamcinolone) can be used. More ablative chemicals (e.g., phenol) are best left to the anesthesiologist to

administer.

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CHAPTER 39

William L. Hasler

APPROACH TO THE PATIENT WITH GAS AND BLOATING

NORMAL PHYSIOLOGY

The gastrointestinal tract of a normal human contains less than 200 mL of gas, whereas daily gas expulsion averages 600 to 700 mL. [1] On average, healthy men pass flatus 14 times per day, although as many as 25 daily expulsions are considered normal. Larger flatus volumes are passed after meals, but flatus production persists during sleep at a slower rate. [1] The principal gases in flatus are nitrogen, oxygen, carbon dioxide, hydrogen, and methane, and they show significant inter- and intraindividual variability (Fig. 39-1). [2] Most hydrogen production results from bacterial metabolism of ingested carbohydrates and endogenous glycoproteins from sloughed enterocytes. Methane also is produced in 40% of the population by the bacterial breakdown of carbohydrates. Carbon dioxide in flatus derives from fermentation of carbohydrates, fats, and proteins. Gases produced by colonic bacteria (hydrogen, methane, and carbon dioxide) represent 74% of flatus. Flatus odor correlates with hydrogen sulfide concentrations; other sulfur-containing gases in flatus include methanetriol and dimethylsulfide (see Fig. 39-1). [3] Another prominent metabolic pathway in the colon is the consumption of hydrogen to form acetate from carbon dioxide.

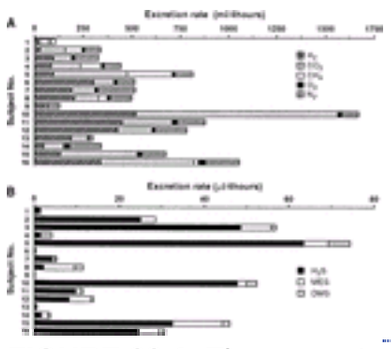

FIGURE 39-1. The amounts of major gases responsible for flatus volume (**A**) and odor (**B**) passed during a 4-hour period are shown for each of 16 healthy individuals. Quantitatively important gases include hydrogen (H_2), carbon dioxide (CO_2), methane (CH_4), oxygen (O_2), and nitrogen (N_2). Prominent sulfur-containing gases include hydrogen sulfide (H_2S), methanetriol (*MES*), and dimethylsulfide (*DMS*). (From ref. [2].)

Several factors modulate gas production by colonic bacteria. Age, gender, and the methane-producing capabilities of a given individual's flora do not significantly affect flatus frequency, although men produce more aromatic flatus than women. [4] Major shifts in the production of methane or a reduction of sulfates occurs spontaneously, indicating the inconstancy of bacterial flora populations. Variability in bacterial hydrogen production reflects changes in methane production or mixing of the stool within the colon. [5] Reductions in hydrogen production develop during daily lactose feeding in patients with lactase deficiency, indicating dietary modulation of the gut flora metabolic pathways. [6]

CLINICAL SYNDROMES

Gas in the gastrointestinal tract is responsible for several clinical phenomena. Eructation, or belching, is the retrograde expulsion of esophageal or gastric gas from the mouth. Involuntary belching after eating is caused by the release of swallowed air after gastric distention and may be exacerbated by foods that reduce lower esophageal sphincter tone. The *Magenblase syndrome* is defined as epigastric fullness and bloating relieved by belching. Manometric studies of eructation show decreases in lower esophageal sphincter tone followed by upper esophageal sphincter relaxation. [7] Flatulence is the volitional or involuntary release of gas from the anus. Manometric studies performed during flatulence demonstrate propagated colon contractions and increased rectal pressure coupled with early anal sphincter relaxation. [8] Bloating is the perception of retained excess gas within the gut lumen. Bloating is experienced monthly by 15.9% of individuals in the United States, whereas 21.8% experience abdominal pain or discomfort. [9] Women are more likely to experience bloating or distention than men (19.2 versus 10.5%). Although some conditions give rise to increased intestinal gas volumes, many patients with bloating exhibit normal amounts of gas within the gastrointestinal tract. [10] Other symptoms experienced by patients with complaints of excess gas include abdominal pain, halitosis, anorexia, early satiety, nausea, loud borborygmi, and constipation.

PATHOGENESIS

Gas and bloating are reported in a number of disorders. These gaseous symptoms may result from excess gas production, abnormal gas transit with generation of retarding motor patterns, and abnormal perception of normal amounts of gas within the gut.

Carbohydrate Maldigestion

Maldigestion and malabsorption of carbohydrates commonly are associated with intestinal gas production. Substances that cause gaseous symptoms include simple and complex carbohydrates and dietary fiber. Unabsorbed carbohydrates are propelled to the colon, where they serve as nutrient substrates for enteric bacteria. Unlike carbohydrate handling by mammalian tissues, bacterial carbohydrate metabolism liberates hydrogen gas and short-chain fatty acids. Flatulence is the initial symptom reported; borborygmi and bloating develop with greater degrees of malabsorption, and abdominal pain and diarrhea develop with the highest levels. Carbohydrate maldigestion and malabsorption may result from loss of enterocyte enzymes in normal intestinal mucosa, inability to transport a poorly absorbed sugar in an otherwise healthy individual, or an organic disorder of the intestinal mucosa, such as celiac disease.

Maldigestion and Malabsorption of Simple Sugars The most common carbohydrate maldigestion syndrome, lactose intolerance, results from insufficient levels of

enterocyte lactase, which hydrolyzes ingested lactose into glucose and galactose. Lactase deficiency is present in 21% of Caucasian Americans but is more prevalent in individuals of African (75%), Hispanic (51%), and Native American (79%) background. [11] Absence of intestinal lactase is rare, so that most individuals can tolerate small amounts of milk products. In individuals who consider themselves severely lactose intolerant, symptoms attributable to lactase deficiency are uncommon unless more than 240 mL of milk is ingested per day. [12] However, in most lactase-deficient individuals, symptoms develop after they ingest 1 L of milk or more. Other simple sugars also may lead to gas and bloating. Furthermore, nonabsorbable sugars such as lactulose accelerate small intestinal transit, further increasing gastrointestinal symptoms. [13] Increased gas production long has been recognized to result from the consumption of fruits or juices. Only 60% of fructose, a sugar in fruits and soft drinks, is absorbed in the normal human intestine. As little as 37.5 g of fructose can produce significant symptoms in some individuals (Fig. 39-2). [14] Sorbitol, a natural substance in fruits and an artificial sweetener in diet foods, is malabsorbed by 43% of Caucasians and 55% of nonwhites. [15] Severe sorbitol intolerance is reported in 32% of non-Caucasians versus 4% of Caucasians. Some subjects are susceptible to as little as 5 g of sorbitol, whereas nearly all develop severe symptoms with 20 g. [16] Consumption of chocolate containing isomalt produces greater increases in breath hydrogen than chocolate with sucrose, and xylitol also is malabsorbed to variable degrees. [17] In fact, 2% to 4% of sucrose is malabsorbed by healthy individuals, although most of this is salvaged in the colon. [18]

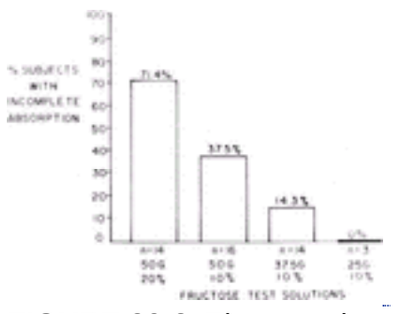

FIGURE 39-2. The prevalence of incomplete fructose absorption as a function of the ingested dose as defined by an increase in breath hydrogen to 20 parts per million or more. (From ref. [14].)

Finally, hereditary syndromes of intolerance of specific sugars are described. Sucrase-isomaltase deficiency is a rare condition presenting with symptoms of carbohydrate malabsorption after sucrose ingestion. Symptoms typically, but not always, begin in infancy. The disorder is inherited in autosomal recessive fashion and results from one of several defects in the sucrase-isomaltase gene. [19]

Maldigestion of Complex Carbohydrates and Fiber Ingested complex carbohydrates also may be poorly assimilated. The average amounts malabsorbed from 100-g meals are 20 g for baked beans, 7 to 10 g for wheat, oats, potatoes, and corn, and 0.9 g for rice (Fig. 39-3). [20] Comparisons of pH and concentrations of lactic and volatile fatty acids in cecal fluid and expelled feces indicate that most unabsorbed starch is metabolized in the colon. Whole grains produce five times more hydrogen than refined flours. There is no evidence that significant adaptation develops with respect to the volumes of gas expelled in persons habitually ingesting a diet rich in beans. Nondigestible oligosaccharides, such as stachyose, raffinose, and verbascose, are abundant in beans and legumes. Flour derived from soy beans low in these oligosaccharides produces less gas than flour derived from standard soybeans. [21]

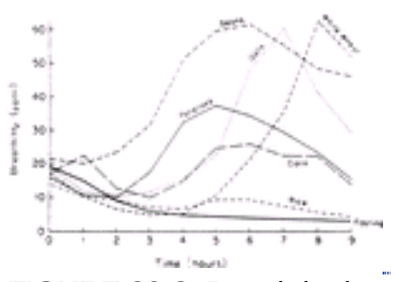

FIGURE 39-3. Breath hydrogen excretion patterns from healthy volunteers after ingestion of different complex carbohydrate-containing foods. Beans, oats, and wheat produce the greatest amount of gas, whereas rice provokes little hydrogen production. (From ref. [20].)

Fiber intake correlates with flatus production in many individuals, although some studies have found no association of gas and bloating with ingestion of psyllium or methylcellulose. [4] In one investigation, fiber-free diets reduced daily gas expulsion from 700 to 200 mL. [1] The intrinsic flora may contribute to bloating after fiber ingestion; individuals who produce low levels of methane experience more symptoms than those who produce high levels of methane. [22] However, incubation of bran with human feces produces only 10% as much hydrogen and carbon dioxide as incubation of lactulose. A clinical correlate of this laboratory finding is a report that fiber supplements increase feelings of bloating but not flatus frequency, whereas lactulose increases both. [23]

Small Intestinal Bacterial Overgrowth

The stomach and small intestine are relatively sterile compared to the colon. Small intestinal bacterial overgrowth complicates several obstructive disorders of the gut, including adhesions, Crohn's disease, radiation enteritis, ulcer disease, and malignancy. Small intestinal diverticula predispose to bacterial overgrowth as a consequence of lumenal stasis, whereas patients with vagotomy acquire intestinal colonization because of hypochlorhydria. The ability of proton pump inhibitors to induce symptomatic bacterial overgrowth is uncertain. Increased small intestinal bacterial counts are prevalent during treatment with omeprazole; however, symptoms commonly are not observed. [24] Patients with functional gut disorders, including intestinal pseudo-obstruction, exhibit overgrowth because of an impairment of gut clearance mechanisms. Such patients frequently show loss of the activity fronts of the migrating motor complex, the fasting pattern responsible for clearing undigested debris from the small intestine. Forty-three percent of cases of diabetic diarrhea are attributed to bacterial overgrowth, mostly from altered motor function. [25] Disorders that increase bacterial delivery to the upper gut, such as cologastric fistulae and coprophagia, may overwhelm normal defenses against infection. Reports suggest that a subset of patients with irritable bowel syndrome may have small intestinal bacterial overgrowth as a cause of symptoms. [26] In some of these individuals, antibiotics relieve gastrointestinal complaints. Finally, bacterial overgrowth is considered in the differential diagnosis of unexplained symptoms in the elderly. Abnormalities of intestinal motor function are believed to underlie development of the condition in older individuals.

Small bowel bacterial overgrowth is defined by the presence of more than 10 [5] colony-forming units per milliliter in intestinal fluid samples. The mean number of bacterial species infesting the upper intestine is 4.6 ± 0.8. The main organisms cultured are *Streptococcus, Escherichia coli, Lactobacillus,* and *Bacteroides*. [27] In addition to producing gas, bloating, diarrhea, abdominal discomfort, and nausea, bacterial overgrowth leads to nutrient malabsorption and weight loss. Other complications include the development of metabolic bone disease and an increased risk for spontaneous bacterial peritonitis in cirrhotic patients. [28]

Dysmotility Syndromes

Conditions that alter gut motor function may produce prominent gas and bloating. Bloating and fullness commonly are reported by individuals with gastroparesis resulting from gastric retention of solids, liquids, and gases. Patients with fat intolerance and rapid gastric emptying also have bloating. [29] Fundoplications for gastroesophageal reflux involve wrapping the gastric fundus around the lower esophagus, thus reducing the ability to belch or vomit. In the early postoperative period, up to 73% of patients experience bloating. [30] Two years later, this gas-bloat syndrome has abated in 71% of individuals. Preoperative delay in gastric emptying is a risk factor for gas-bloat syndrome. Patients with chronic intestinal pseudo-obstruction experience gas and bloating and exhibit small intestinal dilation because of delayed gas transit and small intestinal bacterial overgrowth. Causes of gastroparesis and intestinal pseudo-obstruction include diabetes mellitus, scleroderma, amyloidosis, familial conditions, paraneoplastic syndromes, and endocrine disease. Many cases are idiopathic in nature. Bloating also is reported by patients with chronic constipation.

Acute processes producing adynamic ileus also are associated with prominent gas and bloating. Ileus may involve the entire gut or may be localized, as in acute colonic pseudo-obstruction. Causes of acute ileus include systemic infection or inflammation, the postoperative state, medications, generalized or localized gut hypotension, metabolic disturbances, and cardiovascular disease.

Functional Bowel Disorders

Gas and bloating are prevalent complaints of patients with functional bowel disorders, such as irritable bowel syndrome (IBS) and functional dyspepsia. In one investigation of IBS, bloating was rated the most bothersome symptom by 60% of patients, whereas only 29% considered pain to be most intrusive. [31] Bloating was experienced on 28% of days in another study of IBS, whereas pain was reported on 33% of days. [32] Of the symptoms related by IBS patients, episodes of bloating and pain lasted the longest, on average 5 days. Among different IBS subtypes, patients with constipation-predominant IBS experience greater bloating than those with diarrhea.

The pathophysiology of gas and bloating in the functional bowel disorders is poorly understood. Some have attempted to ascribe these symptoms to anatomic causes. Early publications made reference to a splenic flexure syndrome with air trapping in that region. [33] More recent examinations of digitized computer images of radiographs have identified increases in gas bubbles in patients with functional disorders; however, gas amounts correlate poorly with symptoms. Computed tomography in IBS patients shows increases in lateral abdominal profiles, which have been attributed to changes in gut smooth muscle tone. [34] Others have postulated that the increased distention of IBS results from weak abdominal muscles, recent weight gain, a low position of the diaphragm, or exaggerated lumbar lordosis.

Abnormal fermentation has been proposed as a possible cause of gas and bloating in the functional bowel disorders. Different studies have reported variable responses to dietary exclusion of lactose, sorbitol, and fructose. [35] One study observed a poor correlation of breath hydrogen excretion with pain and bloating in patients with IBS, whereas another noted increased hydrogen production that was responsive to dietary exclusion of certain starches. [36] One group observed increased hydrogen and methane production after lactulose administration in patients with functional dyspepsia and *Helicobacter pylori* infection, suggesting potential pathogenic roles for bacterial populations outside the colon. [37]

Abnormal gastrointestinal motor and sensory functions likely play an important role in the pathogenesis of gas and bloating in IBS. The transit of radioactively labeled bran from the ileum to the colon is faster in IBS patients with bloating than in healthy volunteers. [38] In argon washout studies, patients with functional abdominal pain retain normal amounts of intestinal infused gas. [10] However, more gas refluxes from the small intestine into the stomach, indicating abnormal gas transit. An investigation of distal duodenal gas perfusion reported increased pain in IBS patients without alterations in small intestinal motility, suggesting a primary abnormality of visceral afferent function. [39] More recently, in a study using jejunal perfusion of gases physiologically similar to venous proportions, IBS patients exhibited abnormal gas retention with increases in abdominal distention and symptoms, whereas healthy volunteers tolerated large volumes of gas without retention or symptom development. [40]

Miscellaneous Causes

Most upper gastrointestinal air in healthy individuals accumulates as a result of aerophagia, which can be worsened by gum chewing, smoking, or oral irritation. During aerophagia, negative intrathoracic pressure pulls air into the esophagus across an open upper esophageal sphincter. Patients who have undergone laryngectomy experience gaseous symptoms as a consequence of swallowing air to facilitate esophageal speech. Patients with ulcer disease, gastroesophageal reflux, or biliary colic exhibit aerophagia and chronically belch in an effort to reduce their symptoms. Intestinal obstruction may produce bloating, although other symptoms usually are present. Endocrinopathies such as hypothyroidism may result in gaseousness as part of a broader symptom presentation. Patients with inferior myocardial infarction can experience excess eructation. [41] Finally, medications (anticholinergics, opiates, calcium channel antagonists, antidepressants) produce gas through effects on gut motility.

EVALUATION OF THE PATIENT

History and Physical Examination

Patients with excess gas may report symptoms consistent with functional disease but that also can result from a structural abnormality. Thus, the clinician must search for clues that suggest an organic cause. Relief of symptoms with defecation or flatus is consistent with IBS, as is the absence of symptoms that awaken the patient from deep sleep. In contrast, vomiting, fever, weight loss, nocturnal diarrhea, rectal bleeding, or steatorrhea indicates probable organic disease. Conditions that predispose to small intestinal bacterial overgrowth should be determined by history. The use of medications that delay gut transit should be questioned. Assessment of ethnic background and family history can determine the risk for carbohydrate maldigestion syndromes, such as lactase deficiency. Finally, a history of anxiety or other psychiatric disease raises the possibility of aerophagia or a functional bowel disorder.

A dietary history may correlate symptoms with specific foods. Ingestion of legumes, fruits, unrefined starches, and lactose-containing foodstuffs should be addressed. The clinician should ascertain if soft drinks containing fructose or diet foods and gums containing sorbitol are being consumed. Activities that involve excess swallowing, such as gum chewing, smoking, and chewing tobacco, predispose to aerophagia.

The physical examination findings usually are normal in patients with excess gas, but anxiety, hyperventilation, and air swallowing may be evident as an initial impression. Findings suggestive of organic disease include sclerodactyly with scleroderma, dermatitis herpetiformis in celiac disease, peripheral or autonomic neuropathy with dysmotility syndromes, and cachexia, jaundice, or palpable masses with malignant obstruction. Abdominal inspection may reveal scars from prior fundoplication, vagotomy, or other operations that can cause adhesions. Auscultation assesses for absent bowel sounds with ileus or myopathic dysmotility, high-pitched bowel sounds with intestinal obstruction, or a succussion splash in gastric obstruction or gastroparesis. Abdominal percussion and palpation may reveal tympany and distention in the patient with mechanical obstruction or intestinal dysmotility. The presence of shifting dullness or a fluid wave indicates ascites rather than excess intestinal gas. A rectal examination is performed to exclude occult fecal blood, which would suggest lumenal inflammation or neoplasm.

In general, patients with gas and bloating do not experience serious sequelae of their condition. However, in some patients with complete mechanical intestinal obstruction or toxic megacolon secondary to inflammatory colitis, lumenal perforation may develop and can be life-threatening. Patients with acute colonic pseudo-obstruction are at risk for perforation secondary to thinning of the cecal wall. Rarely, pneumatosis cystoides intestinalis develops in patients with excess hydrogen-producing bacteria in the small intestine. [42] Other complications of organic disease occur rarely and generally are manifestations of the underlying disease rather than of the gas itself. Case reports have appeared of explosions resulting from ignition by tobacco smoking of feculent gas expelled by eructation in patients with gastrointestinal obstruction and bacterial overgrowth. [43] Colonic explosions infrequently have been reported in patients undergoing colonoscopy with mucosal cautery. These rare complications result from inadequate bowel cleaning or the use of mannitol or sorbitol purging solutions, which generate hydrogen and methane to concentrations approaching 30% to 45%. [44]

Laboratory and Structural Testing

Screening laboratory tests assist the clinician in excluding organic disease. The determination of a normal complete blood cell count, electrolyte, glucose, albumin, and total protein levels, and sedimentation rate excludes most inflammatory or neoplastic conditions. In some individuals, a determination of calcium and phosphate levels, parameters of renal and thyroid function, liver chemistries, and fasting morning cortisol levels may be needed. Amylase may be elevated in patients with ischemic gut segments. Patients with diarrhea should collect stools for ova and parasites to rule out giardiasis. Endomysial or tissue transglutaminase antibody levels can be used to screen for celiac disease. [45] If these are positive, the diagnosis is confirmed by intestinal mucosal biopsy. Other serologies of value in selected patients include antinuclear antibodies and scleroderma antibodies to evaluate for possible rheumatologic disease, and antinuclear neuronal antibodies to screen for paraneoplastic visceral neuropathy. [46]

Imaging techniques may be needed to test for disorders that produce mechanical obstruction or functional gas retention. Flat and upright abdominal radiographs may reveal diffuse distention consistent with ileus or pseudo-obstruction, diffuse haziness in ascites, or air-fluid levels in the patient with intestinal obstruction. Distinguishing ileus from obstruction may not be accomplished by plain radiography alone. Contrast enema radiography can detect colonic or distal small intestinal obstruction. Small intestinal contrast radiography can evaluate for partial gastric outlet or small intestinal obstruction. Upper or lower gastrointestinal endoscopy facilitates the identification and biopsy of lesions producing partial blockage. Small bowel barium studies also can crudely quantify intestinal transit and assess motor patterns in patients with possible chronic intestinal pseudo-obstruction. If the index of suspicion for partial obstruction is high, enteroclysis may provide a more detailed assessment of small intestinal lumenal processes. [47] With this technique, the proximal intestine is intubated under fluoroscopic guidance, and barium and

methylcellulose are perfused to provide a double contrast image. Ultrasonography or computed tomography can provide useful information regarding the causes of gaseous distention and exclude processes such as ascites, which may be misinterpreted as abdominal gas.

Functional Testing

When the results of structural testing are unrevealing, tests of gut function may provide insight into the cause of gas and bloating. Techniques are available to quantify gut transit, assess carbohydrate absorption, and measure flatus production.

Tests of Gut Motor Function In suspected gastrointestinal dysmotility, gastric-emptying scanning or gastrointestinal manometry may be considered. Scintigraphic measures of the emptying of liquid (^{111}In-DTPA [diethylenetriamine pentaacetic acid] in water) or solid (^{99m}Tc-sulfur colloid in eggs) radionuclides are the most commonly performed tests of gastric emptying. Scintigraphy also has been used to assess rates of small intestinal or colonic transit. [48] Alternatively, radiopaque marker techniques are easily performed to diagnose slow transit constipation. [49] In chronic intestinal pseudo-obstruction, small intestinal manometry provides information about the neuropathic or myopathic nature of the condition. Pseudo-obstruction resulting from enteric nerve dysfunction, as in the familial visceral neuropathies or early scleroderma, produces intense, uncoordinated bursts of motor activity with loss of normal migrating motor complex (MMC) and fed motor activity. [50] Smooth muscle dysfunction, as in the familial visceral myopathies or late scleroderma, produces low-amplitude contractions. Intermittent bursts separated by periods of motor quiescence, a pattern known as the *minute rhythm,* has been observed in patients with small intestinal obstruction, although similar patterns are seen in neuropathic pseudo-obstruction and IBS. [51] Manometry does not provide precise characterization of the underlying pathology in some cases. Thus, a full-thickness intestinal biopsy specimen obtained surgically may be required to document degeneration of nerve or muscle layers.

Breath Testing Hydrogen breath testing may be used to confirm carbohydrate maldigestion or malabsorption as a cause of gas and bloating. This technique relies on the ability of lumenal bacteria to produce hydrogen during the metabolism of ingested substrates and the inability of human tissue to utilize similar metabolic pathways. Expired breath samples commonly are obtained before and for 2 hours after the ingestion of an aqueous solution of the sugar that is presumed to be malabsorbed or maldigested. Proper breath holding followed by immediate exhalation reduces the variability in hydrogen levels from 28% to 10%. [52] Increases in breath hydrogen by more than 20 parts per million within 120 minutes of lactose ingestion distinguishes biopsy-proven lactase deficiency from lactase sufficiency with a sensitivity of 90%. [53] Hydrogen excretion after lactose ingestion correlates well with ymptoms of carbohydrate maldigestion, whereas lactose tolerance testing with measurement of plasma glucose concentrations shows a poor correlation. [54] Hydrogen measurement may have to be extended to 10 hours if testing for maldigestion of complex carbohydrates such as starch is being performed. Even for lactose, some have proposed extending hydrogen measurements to 5 to 7 hours to increase the sensitivity and specificity of the test. [55] Children with sucrose intolerance may be tested for sucrase-isomaltase deficiency with sucrose hydrogen breath testing. Some patients may be tested for fructose or sorbitol malabsorption with hydrogen breath testing, although normal values for these tests are less well defined. Hydrogen breath testing also has been used to test for small intestinal bacterial overgrowth. Elevations in fasting breath hydrogen or early rises within 30 minutes after substrate ingestion suggest overgrowth. The most commonly used sugar for breath hydrogen testing in suspected bacterial overgrowth is glucose, which provides diagnostic sensitivities and specificities of 60% to 90%. Others have proposed using lactulose or rice as substrates, but one study reported only 17% to 33% sensitivity for detecting bacterial overgrowth by these methods. [56] False-negative results of breath tests occur in patients with few hydrogen-producing bacteria, whereas false-positive results occur with rapid transit of the ingested carbohydrate to the colon. Other centers have relied on ^{14}C- or ^{13}C-labeled substrates, with measurement of breath ^{14}C-carbon dioxide or ^{13}C-carbon dioxide, although special facilities are necessary for these analyses. A comparison of glucose hydrogen breath testing and ^{14}C-xylose breath testing noted superior sensitivity for the hydrogen measurements in the detection of bacterial overgrowth. [57] When the diagnosis is in doubt, the gold standard remains quantitative culture of duodenal or jejunal secretions. [58] Bacterial counts above 10 ^{5} colony-forming units per milliliter are diagnostic of bacterial overgrowth. Finally, hydrogen breath testing has been used to quantify orocecal transit in suspected chronic intestinal pseudo-obstruction. Unabsorbed substrates such as lactulose are used for this purpose. The transit time is measured from ingestion until an increase in breath hydrogen, representing the onset of colonic bacterial metabolism, is observed (Fig. 39-4). This method has significant limitations. It often is difficult to determine when the increase in hydrogen production ensues. Furthermore, lactulose itself accelerates small intestinal transit. [59] Finally, erroneous values are obtained in patients with small intestinal bacterial overgrowth.

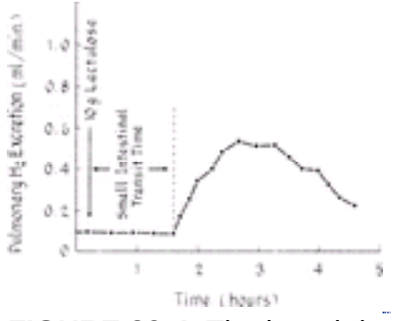

FIGURE 39-4. The breath hydrogen excretion profile for a healthy volunteer after ingestion of 10 g of lactulose. The short intestinal transit time is quantified by the time required to detect an increase in expired hydrogen. (From ref. [13].)

Flatus Analysis In some research institutions, flatus analysis is performed to gain insight into the processes responsible for excess flatulence. Testing consists of counting the number of flatus passages over 24 hours to determine if increased flatulence is present (normal, <20 daily). [60] The expelled gas is then analyzed to determine if it is rich in nitrogen, indicating aerophagia, or gases such as carbon dioxide, hydrogen, and methane, suggesting increased colonic production. Such scrutiny has been used to direct the treatment of patients with severe flatulence. [60]

PRINCIPLES OF MANAGEMENT

The management of the patient with gas and bloating depends on the cause of the symptoms. Structural abnormalities such as mechanical obstruction may require surgery. Patients with excess eructation from gastroesophageal reflux can derive benefit from acid-suppressive medications. For other causes of gaseous symptoms, several recommendations can be made, including dietary modifications and the prescription of nonmedication and medication therapies.

Dietary and Nonmedication Therapy

Dietary measures may reduce gas and bloating in selected patients. Elimination of lactose may produce improvements in individuals with lactase deficiency. Most lactase-deficient persons tolerate small amounts of lactose (0.5–7 g), indicating residual lactase activity. [61] When consumed in yogurt with active cultures, lactose may be better tolerated because of the presence of bacterial ß-galactosidase. Hydrogen production from yogurt is only one third of that from milk. [62] The pasteurization of yogurt reduces its lactase activity, increasing breath hydrogen concentrations by 39%. However, pasteurized yogurt containing up to 20 g of lactose is tolerated by lactase-deficient individuals, suggesting that some of the benefits of yogurt stem from factors other than its enzymatic capabilities. Fermented milk products containing *Lactobacillus acidophilus, Bifidobacterium* species, and *Lactobacillus bulgophilus,* developed to increase lactase levels, reduce bloating in patients with lactose intolerance. [63] Children with sucrase-isomaltase deficiency also benefit from dietary modifications with elimination of sucrose. In some individuals, sucrose can slowly be reintroduced into the diet because the colon possesses some capacity for fermentation of the sugar.

Many IBS patients are postulated to have symptoms resulting from carbohydrate intolerance. The efficacy of lactose restriction in IBS patients with lactase deficiency is unimpressive, with only a subset experiencing symptom benefits. [64] Similarly, 40% of patients experience symptom relief after exclusion of fructose and sorbitol. [35] This value is similar to responses to placebo in controlled trials of IBS; thus, it is uncertain whether the result is clinically significant. Others have proposed the initiation of low-gas diets that exclude many complex carbohydrates. [65] This diet has been reported to reduce flatus frequencies by 50% in some patients (Table 39-1). [66]

VISCEROSENSORY SYMPTOM (N = 443)	INDICATED BY PATIENT, % (No.)	SINGLE MOST BOTHERSOME SYMPTOM, % (No.)
Extra-abdominal symptoms		
Pain behind chest bone	14 (61)	3 (11)
Pressure behind chest bone	11 (52)	3 (13)
Nausea	49 (229)	3 (15)
Bloating symptoms		
Sensation of gas	76 (357)	22 (98)
Sensation of bloating	66 (308)	5 (24)
Sensation of fullness	56 (253)	17 (75)
Bloating with distention of the belly	58 (262)	16 (73)
Incomplete evacuation		
Sensation of fullness in the rectum after a bowel movement	32 (143)	3 (15)
Abdominal pain		
Belly pain	60 (227)	29 (129)

Note: The prevalence of visceral sensory symptoms and the single most bothersome symptom during the past 2 weeks according to the patient are shown.

From ref. 50.

TABLE 39-1 Prevalence of Symptoms in Irritable Bowel Syndrome

In some instances, the foods themselves may be modified to decrease their propensity to produce gas. Soaking cowpeas and yam beans for 12 hours and cooking for 30 minutes eliminates most malabsorbed oligosaccharides, reducing raffinose from between 0.71% and 6.86% to between 0.04% and 0.40%, and stachyose from between 2.38% and 4.14% to between 0.12% and 0.72%. [67] Gamma irradiation to levels used for insect disinfestation (0.25–0.75 kGy) also reduces raffinose in mung beans. [68]

Lifestyle changes and other nonmedication treatments are offered to selected individuals. Many cases of excess eructation stem from aerophagia, which can be controlled by cessation of gum chewing and smoking. The chronic belcher may benefit from observation in a mirror to emphasize the role of air swallowing. Gas-trapping undergarments have been proposed for individuals with excessive malodorous flatus. The best-characterized device consists of an airtight Mylar brief with a charcoal-lined cushion and has been reported to absorb more than 90% of offensive gases (Fig. 39-5). [3]

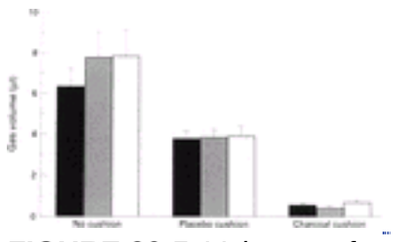

FIGURE 39-5. Volumes of sulfur-containing gases escaping into the surrounding environment when gas was instilled at the anus in the presence of an activated charcoal cushion, a placebo cushion, and no cushion. Values are shown for hydrogen sulfide (*black bars*), methanetriol (*shaded bars*), and dimethylsulfide (*white bars*). (From ref. [3].)

Medication Therapy

A number of medications have been proposed to treat gas and bloating. However, few controlled trials have confirmed their efficacy versus placebo. Thus, many recommendations regarding their utility are based on studies in healthy individuals or are grounded in the anecdotal experience of clinicians who care for these patients.

Enzyme Preparations Enzyme preparations are advocated to facilitate the breakdown of food residues incompletely digested by intrinsic enzymes in patients with gas and bloating. The best-characterized exogenous enzymes are the ß-galactosidase (lactase) preparations used for lactose intolerance. In adults, lactase supplements reduce hydrogen excretion and bloating, cramps, and flatulence after the ingestion of lactose. [69] Similarly, in lactose-intolerant children, lactase tablets reduce hydrogen levels after a lactose challenge from 60 to 7 parts per million, with corresponding reductions in pain, bloating, diarrhea, and flatulence. [70] Small studies have reported differential efficacy of different lactase preparations, but this finding must be confirmed in larger trials. [71] Other enzyme compounds are used to prevent gaseous symptoms. Hydrogen production and bloating and cramping decrease when children with sucrase-isomaltase deficiency are given sacrosidase, a liquid derived from *Saccharomyces cerevisiae* that contains 6000 IU of sucrase activity per milligram of protein. [72] Bacterial a-galactosidase derived from *Aspergillus niger* (Beano) reduces flatulence in healthy volunteers who consume meatless chili rich in oligosaccharides. [73] However, most indigestible fiber does not contain galactose; thus, this agent is less likely to produce benefit in patients with gas from causes other than a legume-rich diet. Encapsulated pancreatic enzymes reduce bloating, gas, and fullness after high-calorie, high-fat meals in healthy individuals without reducing breath hydrogen or methane. [74] However, studies of pancreatic supplements have not been performed in patients with gas and bloating to test if they have broader utility.

Adsorbents and Agents to Decrease Surface Tension Some agents are proposed to reduce distention by a defoaming action or by direct adsorption of excess gas. Simethicone promotes rupture and liquid drainage from thick foam films. One study reported decreases in breath hydrogen in healthy subjects who received simethicone; however, a second study observed no reduction in hydrogen with simethicone in a randomized, double-blinded trial. [75] Simethicone reduced symptoms to a greater degree than cisapride in an investigation of patients with functional dyspepsia. [76] Another study reported superior relief of gas-related abdominal discomfort in patients with diarrhea given the combination of loperamide and simethicone in comparison with either agent alone. [77] Activated charcoal has been used to treat both the volume of gas produced and its odor. Charcoal reduced flatulence and breath hydrogen after the consumption of gas-producing meals in one study. [78] In a second controlled investigation, charcoal decreased bloating, cramps, and hydrogen after lactulose in separate populations in the United States and India. [79] However, another study showed no reductions in hydrogen or flatulence with charcoal after the ingestion of baked beans. Similarly, the effects of activated charcoal on flatus odor are uncertain. In dogs, charcoal reduces fecal hydrogen sulfide production. [80] Ingestion of charcoal in combination with *Yucca schidigera* and zinc acetate reduces the number of flatulence episodes with bad odor by 86%. However, an investigation in humans did not observe any effect of charcoal on sulfur-containing gases responsible for flatus malodor. [81] Bismuth compounds also have been promoted to reduce flatus volume and odor. Tripotassium dicitrate bismuth, bismuth subsalicylate, and bismuth subnitrate inhibit in vitro fermentation of lactose-enriched feces. Studies of flatulent patients observed a reduced fermentation of raffinose with chronic bismuth subsalicylate therapy. Furthermore, fecal homogenates from individuals on bismuth subsalicylate for 3 to 7 days showed impaired hydrogen sulfide release, suggesting a role for this agent in controlling flatus odor. [82]

Antibiotics Antibiotics provide benefit to patients with small intestinal bacterial overgrowth. Elderly patients treated for bacterial overgrowth exhibit increases in weight and body mass index. Several inexpensive antibiotics reduce symptoms of bacterial overgrowth, including tetracycline and metronidazole. In patients with systemic sclerosis, ciprofloxacin produced superior symptom control versus trimethoprim. [83] In a second study, amoxicillin-clavulanic acid and cefoxitin effectively treated more than 90% of strains responsible for small intestinal bacterial overgrowth. [27] A third investigation observed 70% reductions in breath hydrogen after norfloxacin. [84] More recently, researchers have focused on nonabsorbable antibiotics that are bacteriocidal without entering the systemic circulation. In separate studies, rifamaxin produced greater reductions in hydrogen excretion and symptoms than activated charcoal and chlortetracycline. [85] Antibiotics have been proposed as primary therapy for IBS. The importance of endogenous flora in IBS is suggested by the evaluation of a population followed after antibiotic treatment for *H pylori* infection. [86] In persons treated with antibiotics, IBS developed less frequently than in those who did not receive antibiotics. In a poorly controlled follow-up of IBS patients with purported bacterial overgrowth, antibiotics produced symptom relief in 48%. [26] A more recent controlled trial of IBS patients with positive hydrogen breath test results after lactulose observed benefits in 50% of individuals after a 10-day course of neomycin, versus 17% with placebo. [87]

Prokinetic Medications Drugs that stimulate gut propulsion theoretically should benefit patients with gas and bloating secondary to gastrointestinal dysmotility. In addition to reducing nausea and vomiting, metoclopramide decreases fullness and bloating in patients with diabetic gastroparesis. Similarly, the peripheral dopamine receptor antagonist domperidone relieves bloating as well as nausea and heartburn in Parkinson's disease patients with delayed gastric emptying. [88] The recently withdrawn serotonin 5-HT $_4$ receptor agonist cisapride decreases eructation in patients with gastroesophageal reflux. Likewise, some but not all placebo-controlled studies report reduced bloating in functional dyspepsia patients treated with cisapride. In one investigation, this improvement was restricted to patients with normal

rather than delayed gastric emptying. [89] In contrast to its therapeutic efficacy in gastric dysmotility, metoclopramide provides no benefit to patients with postoperative ileus. [90] Other prokinetics may selectively target the small intestine and colon. Cisapride was shown to accelerate orocecal transit and eliminate bacterial colonization in cirrhotic patients with bacterial overgrowth. In studies of IBS patients, cisapride produced no significant reductions in bloating or flatulence, although one investigation reported relief of distention. [91] The somatostatin analog octreotide reduces breath hydrogen after glucose in scleroderma patients with intestinal pseudo-obstruction and bacterial overgrowth. [92] When administered over 20 to 33 weeks, the combination of the motilin receptor agonist erythromycin and octreotide decreased symptoms in patients with chronic intestinal pseudo-obstruction. [93] The 5-HT $_4$ receptor agonist tegaserod accelerates small intestinal and ascending colon transit in patients with constipation-predominant IBS. [94] Investigations with tegaserod in constipated IBS patients report at least temporary reductions in bloating associated with the syndrome. [95]

Probiotics and Alternative Therapies The aim of probiotic therapy is to replace pathogenic colonic organisms with ingested innocuous strains. Probiotics also may have additional mechanisms of action. *Lactobacillus plantarum* increases fecal carboxylic acid and decreases blood fibrinogen levels; the importance of these effects is unknown. [96] *Lactobacillus casei* strain GG reduces bloating, diarrhea, and taste disturbances associated with antibiotic therapy of *H pylori* infection. However, in IBS patients with bloating, *Lactobacillus* GG did not reduce abdominal pain, fecal urgency, or bloating, although it did promote a trend to improved fecal consistency in patients with diarrhea. [97] A second study observed significant reductions in flatulence but not bloating during 4 weeks of treatment with *L plantarum,* which was associated with decreased fecal enterococci in comparison with placebo treatment. [98] Although these reports suggest possible benefits of microbial therapy, probiotics should be considered experimental until larger double-blinded trials are performed to confirm their efficacy. Other alternative therapies have been proposed for gas and bloating. Hypnotherapy reduces bloating and flatulence and improves quality of life in IBS patients and has been described as treatment for intractable eructation. [99] In an open trial, acupuncture relieved bloating and improved general well-being in a small group of patients with IBS. The addition of auricular plaster therapy to acupuncture at the Zusanli point promoted faster recovery of normal peristalsis in patients with postoperative ileus than in a control group. [100]

Surgical Management

Operative therapies for gas and bloating usually are considered only for patients with the most refractory cases of severe organic disease. Percutaneous endoscopic gastrostomy is effective in selected cases of gas-bloat syndrome after fundoplication. [101] Excision of small intestinal diverticula reduced symptoms and vitamin B_{12} malabsorption in a patient with small intestinal bacterial overgrowth. Selected patients with localized intestinal pseudo-obstruction may benefit from resection of the dysfunctional bowel segment. Patients with more extensive intestinal pseudo-obstruction may experience symptom relief after a venting jejunostomy. [102] Similarly, some patients with acute colonic pseudo-obstruction may require surgical or radiographic insertion of a decompressing cecostomy to prevent rupture of the organ. Finally, some individuals with advanced pseudo-obstruction require surgical or radiographic placement of indwelling central venous catheters for home total parenteral nutrition.

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CHAPTER 40

Michael W. Mulholland and John F. Sweeney

APPROACH TO THE PATIENT WITH ACUTE ABDOMEN

The term *acute abdomen* describes a syndrome of sudden abdominal pain with accompanying symptoms and signs that focus attention on the abdominal region. It is clinically useful to limit discussion to cases in which the pain has been present for less than 24 hours. Associated symptoms such as nausea, vomiting, constipation, diarrhea, anorexia, abdominal distention, and fever are often present and sometimes confuse the overall clinical picture. Although operative therapy is not required for all cases of acute abdomen, unwarranted operative delay can have serious, potentially fatal consequences. Successful management of affected patients is based on the following: a careful initial assessment, incorporating history taking and physical examination; delineation of clinical priorities; and concurrent resuscitation, diagnosis, and therapy. The most common causes of the acute abdomen are listed in Table 40-1 and Table 40-2.

Gastrointestinal	
Appendicitis	
Perforated peptic ulcer	
Intestinal obstruction	
Intestinal perforation	
Intestinal ischemia	
Colonic diverticulitis	
Meckel diverticulitis	
Inflammatory bowel disease	
Pancreatic, Biliary, Hepatic, and Splenic	
Acute pancreatitis	
Acute cholecystitis	
Hepatic abscess	
Ruptured or hemorrhagic hepatic tumor	
Acute hepatitis	
Acute cholangitis	
Splenic rupture	
Urologic	
Ureteral stone	
Pyelonephritis	
Retroperitoneal	
Aortic aneurysm	
Retroperitoneal hemorrhage	
Gynecologic	
Ruptured ovarian cyst	
Ovarian torsion	
Ectopic pregnancy	
Acute salpingitis	
Pyosalpinx	
Endometritis	
Uterine rupture	
Abdominal Wall	
Rectus muscle hematoma	

Note: Adapted from Mulholland MW, Debas HT. In: Kelley WN, ed. Textbook of internal medicine, 3rd ed. Philadelphia: Lippincott-Raven, 1997:599.

TABLE 40-1 Abdominal Causes of Acute Abdomen

Thoracic	
Myocardial infarction	
Acute pericarditis	
Lower lobe pneumonia	
Pneumothorax	
Pulmonary infarction	
Hematologic	
Sickle cell crisis	
Acute leukemia	
Neurologic	
Herpes zoster	
Tabes dorsalis	
Nerve root compression	
Metabolic	
Diabetic ketoacidosis	
Addisonian crisis	
Acute porphyria	
Hyperlipoproteinemia	
Drug-Related	
Lead toxicity	
Narcotic withdrawal	

Note: Adapted from Mulholland MW, Debas HT. In: Kelley WN, ed. Textbook of internal medicine, 3rd ed. Philadelphia: Lippincott-Raven 1997:600.

TABLE 40-2 Extra-abdominal Causes of Acute Abdomen

NEUROANATOMY OF ABDOMINAL PAIN

Nerve fibers that mediate painful visceral stimuli have cell bodies in dorsal root ganglia. Processes of first-order sensory neurons pass through spinal nerves to white rami communicantes and then to sympathetic ganglia, which parallel the spinal cord. Nerve processes then travel from sympathetic ganglia through the splanchnic nerves to reach intra-abdominal splanchnic ganglia. The splanchnic ganglia are concentrations of nervous tissue surrounding the origins of the celiac, superior mesenteric, and inferior mesenteric arteries. Sensory nerve fibers traverse splanchnic ganglia without synapse and accompany blood vessels to reach their target organs. Second-order neurons transmit afferent impulses centrally through the lateral spinothalamic tract on the side opposite to the dorsal root containing the nerve cell body. Neurons in the lateral spinothalamic tract project to the thalamus, where they synapse with tertiary neurons mediating sensory and discriminatory aspects of pain. Tertiary neurons project to the somatosensory cortex. Some secondary neurons in the lateral spinothalamic tract synapse in the brainstem with neurons

projecting to the limbic system. Almost all nerve fibers that transmit painful sensations travel in association with sympathetic nerves. As a result, although 80% to 90% of vagal fibers are afferent, interruption of parasympathetic innervation by vagotomy does not block abdominal pain.

During normal embryological development, abdominal organs receive bilateral sympathetic innervation, with input to each organ originating from several adjacent spinal levels. As a result, visceral pain is sensed as a midline, poorly localized sensation. The location of the pain is determined by the developmental origin of the affected organ (Fig. 40-1). Visceral pain originating from organs derived from the foregut is sensed in the epigastrium. Foregut structures include the stomach, liver, biliary system, pancreas, spleen, and duodenum. Pain from midgut structures usually is experienced in a periumbilical distribution, whereas organs derived from the hindgut project painful sensations to the hypogastrium and lower midline. The embryonic midgut develops into the jejunum, ileum, appendix, and colon to the level of the midtransverse colon. Hindgut structures include the distal transverse, splenic flexure, descending, and sigmoid portions of the colon.

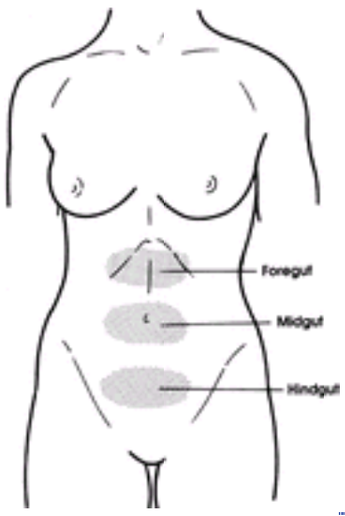

FIGURE 40-1. Distribution of visceral pain perception based on embryological origin of diseased organ.

Neuronal pathways that conduct visceral abdominal pain are composed of slowly conducting C-type fibers. Nociceptive fibers may be stimulated by distention, forceful muscular contraction, mesenteric traction or torsion, or certain noxious chemicals. In hollow organs, such as the small intestine, sensory fibers are found within the muscular wall. In solid organs, such as the liver, nociceptive fibers are limited to the capsule of the organ. For this reason, mass lesions of the liver parenchyma usually do not cause pain until increasing size causes stretching of the Glisson capsule or traction on the organ.

The peritoneum provides a continuous layer over the inner surface of the abdominal wall, reflecting onto the intraperitoneal viscera. Although visceral and parietal peritoneal surfaces have a common mesenchymal derivation, their patterns of innervation are entirely different. The visceral peritoneum is supplied by C-type nociceptive fibers, which transmit painful visceral sensations as outlined earlier. The parietal peritoneum is supplied by rapidly conducting A-type nerve fibers. A-type nerve fibers have small receptive fields and produce sharp, well-localized, highly discriminated sensations. The parietal peritoneum is somatically innervated in a unilateral dermatomal distribution. Somatic peritoneal nerves transmit painful impulses in response to changes in temperature or pH and in response to pressure or incision. Somatic pain caused by stimulation of the parietal peritoneum usually is perceived as intense and constant.

Physiological properties of the peritoneum, including particulate and microbial absorption and circulation of intraperitoneal fluid, may affect perception of noxious stimuli. The peritoneal cavity is the largest extravascular space in the body, with an estimated surface area of 1.7 m^2 in adults. Although the entire peritoneal surface can participate in water and low-molecular-weight solute exchange, removal of particulate matter occurs only along the undersurface of the diaphragm. In this area, specialized lymphatic vessels are present beneath the mesothelial layer covering the diaphragmatic surface. Large pores, or *lacunae,* serve as channels for passage of peritoneal particulate matter up to 10 µm into the lymphatic circulation. Negative intrathoracic pressure during inspiration draws fluid into the lymphatic channels; valves in thoracic lymphatic channels prevent reflux during expiration.

The removal of fluid by means of diaphragmatic lymphatics causes movement of fluid within the peritoneal cavity. Experimentally, injection of contrast material into the right iliac fossa is followed by accumulation of contrast in the right paracolic region, in both subhepatic spaces, and in the pelvis. [1] The patterns of intraperitoneal circulation correspond to movement of contaminated material after rupture of an abdominal viscus and to subsequent abscess formation. Because the parietal peritoneum is sensitive to chemical irritants, movement of fluid may cause pain to be sensed in an area distinct from the site of the disorder. As an example, perforated duodenal ulcer may cause pain in the right lower quadrant as a result of movement of gastric contents along the right paracolic gutter.

A distinctive feature of visceral abdominal discomfort is its association with pain perceived at an extra-abdominal site unrelated to the injured organ. This phenomenon, called *referred pain,* is associated with intra-abdominal or retroperitoneal pathology and usually is described as a dull ache near the body surface. In general, referred pain is sensed in a cutaneous area that has its somatic innervation derived from the same spinal segments that supply visceral afferents to the diseased organ. Patterns of referred pain are often stereotypical enough to be diagnostically useful. Gallbladder inflammation frequently is associated with pain referred to the right scapula. Subdiaphragmatic inflammation causes pain referred to the area supplied by cervical dermatomes 3, 4, or 5. Pain from pancreatic disease may be sensed in the posterior midline in the region of the first lumbar vertebra (Fig. 40-2).

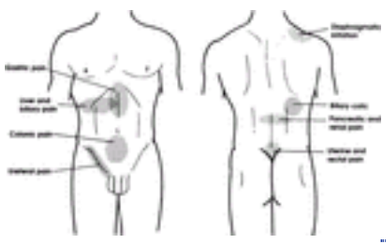

FIGURE 40-2. Common patterns of referred pain in patients with acute abdominal processes. (From Mulholland MW, Debas HT. Approach to the patient with acute abdomen. In: Kelley WN, ed. Textbook of internal medicine, 3rd ed. Philadelphia: Lippincott–Raven, 1997:620.)

ASSOCIATED GASTROINTESTINAL SYMPTOMS

Visceral abdominal pain is distressing, yet difficult for patients to describe. As a result, patients and physicians may focus on associated gastrointestinal symptoms that almost invariably accompany acute visceral pain.

Anorexia accompanies almost all acute abdominal processes but is not specific to any pathological process. Anorexia is usually present in cases of acute appendicitis, and its absence should place that diagnosis in question. Anorexia is also common in the initial phases of acute cholecystitis. The symptom is found less frequently with urologic or gynecologic causes of acute abdominal pain.

Emesis occurs as a result of reflex stimulation of the medullary vomiting center in the initial phases of acute abdominal processes. Reflex vomiting is not progressive and does not relieve unpleasant symptoms. In contrast, vomiting resulting from mechanical intestinal obstruction is recurrent and progressive. Vomiting caused by intestinal obstruction may cause intravascular volume depletion and prerenal azotemia. Bacterial proliferation proximal to the obstruction causes the vomitus to

become feculent.

Abdominal distention associated with acute abdominal pain usually signifies accumulation of swallowed gas in the bowel as a result of mechanical obstruction or ileus. Fluid secreted into the bowel lumen proximal to a mechanical obstruction also may contribute to abdominal distention. Mechanical obstruction may be associated with hyperactive or high-pitched bowel sounds, but the auscultatory finding of diminished bowel sounds is not a reliable sign differentiating ileus from mechanical obstruction. A truly silent abdomen in the presence of distention and tenderness is an ominous sign of bowel necrosis and peritonitis.

Constipation may be a sign of previous health habits (e.g., diet), a disease process (e.g., diverticulitis), or the development of a complication (e.g., perforation). The cessation of intestinal movements or flatus coinciding with the development of acute abdominal pain is more properly referred to as *obstipation.* Obstipation is associated with both mechanical obstruction and functional ileus.

Watery diarrhea associated with acute abdominal pain suggests acute gastroenteritis or infectious colitis. Bloody diarrhea in the context of the acute abdomen may be associated with exacerbation of chronic inflammatory bowel disease, with mesenteric ischemia, or occasionally with mesenteric venous thrombosis.

HISTORY

Treatment for patients with acute abdominal processes differs fundamentally from care delivered to patients with long-term complaints. The potential for pathological processes to be rapidly progressive, and for serious adverse consequences to result from therapeutic delay, places a time constraint on diagnosis and treatment. An accurate diagnosis should lead promptly to specific therapy. A complete and accurate history and physical examination are the most important requirements for success.

The treating physician first should focus on the nature and timing of the abdominal pain. The pattern of onset and the progression of pain provide valuable clues to cause. The pain associated with perforation of a duodenal ulcer or rupture of an abdominal aortic aneurysm is incapacitating, begins suddenly, and quickly reaches peak intensity. Because the onset of pain is so dramatic, affected patients may be able to provide detailed information about the time of onset or their activities at that moment. In contrast, pain associated with appendicitis increases over a period of 1 to several hours. Similarly, pain caused by acute cholecystitis increases over hours before reaching a steady intensity. The duration of painful symptoms is equally important. Biliary colic typically lasts for several hours before rapidly resolving, presumably as a result of the offending stone's dislodging from the cystic duct. Pain caused by acute pancreatitis is unrelenting. Initially, patients with mechanical small bowel obstruction may feel remarkably well between episodes of intense and debilitating colic.

Prior symptoms may implicate preexisting disease. A history of epigastric pain relieved by food may indicate prior duodenal ulceration, whereas previous biliary colic is common in patients presenting acutely with cholecystitis or pancreatitis. Previous abdominal operations influence both the occurrence and the manifestations of acute abdominal processes. Adhesions resulting from previous intraperitoneal operation are the most frequent cause of mechanical obstruction of the small intestine. Patients who have undergone hysterectomy, appendectomy, or pelvic colonic resection are at greatest risk of adhesive obstruction.

PHYSICAL EXAMINATION

The physical examination should be conducted in a systematic and unhurried manner. A complete abdominal examination requires unhindered visualization of the area between the nipples and the midthigh, anteriorly and posteriorly. The examination should begin with observation of the patient's expression and behavior. A patient with serious intraperitoneal disease usually has an anxious, pale face. Sweating, dilated pupils, and shallow breathing are common. In the presence of chemical or bacterial contamination of the peritoneum, the patient tends to lie immobile, to minimize movement of inflamed viscera against the parietal peritoneum. Knees may be flexed, the abdomen scaphoid, breathing shallow. Inhaling deeply or coughing aggravates the pain. With ureteral colic or mesenteric ischemia, by contrast, the patient may appear restless, with frequent changes in posture in an attempt to relieve discomfort. During inspection, the location of all surgical scars, masses, external hernias, and stomas should be determined.

Auscultation should precede abdominal palpation. All four quadrants should be auscultated for tone and quantity of bowel sounds and the presence of vascular bruits. Bowel sounds are considered absent only if no tones are heard over a 2-minute period of auscultation.

Next, the abdomen should be palpated. To determine areas of tenderness and the vigor with which palpation may be pursued, it is useful first to ask the patient to demonstrate the point of maximal discomfort. Palpation should begin in the abdominal quadrant farthest from the area of suspected disease. Gentle pressure to elicit tenderness and muscular resistance then ensues. Progressively deeper palpation is attempted to delineate masses. Intentional efforts to reproduce abdominal pain by deep palpation and rapid release of pressure, termed *rebound tenderness,* are not helpful and should not be attempted. Production of rebound tenderness provides no information that is not available through gentle examination, causes the patient to guard voluntarily, and eliminates the possibility of meaningful serial abdominal examinations. The best evidence of a localized inflammatory process is demonstration of *point tenderness,* caused by the movement of parietal peritoneum against the inflamed surface of a diseased viscus. Point tenderness should be sought by palpation in the area of maximal discomfort but also may be elicited by grasping the patient's hips and gently rocking the pelvis; the movement of inflamed peritoneum is presumed to cause pain. The stethoscope also may be used to palpate the abdominal quadrants.

Every patient must undergo a digital rectal examination. If an inflamed appendix lies deep within the pelvis, point tenderness sometimes may be elicited only by palpation through the right rectal wall. Stool should be tested for guaiac positivity. In females, manual and speculum vaginal examinations are required; vaginal secretions should be obtained for Gram stain and culture. All external stomas, wounds, and fistulae should be explored digitally.

CONFOUNDING FACTORS

Female Gender

Virtually all series of surgically treated patients with acute abdominal pain report that diagnostic inaccuracy is greatest in young women. [2] Detailed sexual and menstrual histories are crucial; a pregnancy test is mandatory in every female patient of childbearing age who is evaluated with acute abdominal pain. Gynecologic causes of acute abdominal pain include ectopic pregnancy, ruptured or twisted ovarian cyst, acute salpingitis, and tuboovarian abscess. On pelvic examination, ectopic pregnancy may present as a tender, unilateral, adnexal mass. Inflammatory disease of the fallopian tubes and uterus produces physical findings of local cervical tenderness, bilateral pelvic pain with displacement of the cervix, adnexal mass, or cervical discharge. All secretions from the cervical os should be submitted for aerobic and anaerobic cultures. Fluid within the pouch of Douglas can be aspirated and examined for blood or bacteria. When the distinction between acute appendicitis and salpingitis is difficult, pelvic ultrasound can be diagnostic. [3] The pelvic organs and appendix also may be observed laparoscopically, with laparoscopic appendectomy as a therapeutic option.

Pregnancy

The most common cause of the acute abdomen in pregnancy is acute appendicitis, occurring in 1 of 6000 pregnancies. [4] Diagnosis of acute abdominal pain in pregnancy is complicated because uterine enlargement displaces organs from their normal anatomic positions. For example, the appendix is moved progressively from the right lower quadrant toward the right upper quadrant (Fig. 40-3). As a consequence, point tenderness associated with appendicitis is sensed in a position more cephalad than usual. In addition, the appendix is moved away from the abdominal wall by the uterus, so parietal peritoneal irritation may be absent. Pregnant women should be evaluated as aggressively as nonpregnant patients; although therapy may be more cumbersome in pregnancy, the indications for operation are usually unchanged. Operative delay that permits progression of acute appendicitis to appendiceal perforation often is followed by premature labor. [5] In one study, three fetal deaths (43%) occurred among seven premature deliveries. [4] In the same series, no fetal or maternal mortality was noted in a group managed without appendiceal perforation. The best guarantee for a living, healthy baby is a healthy mother.

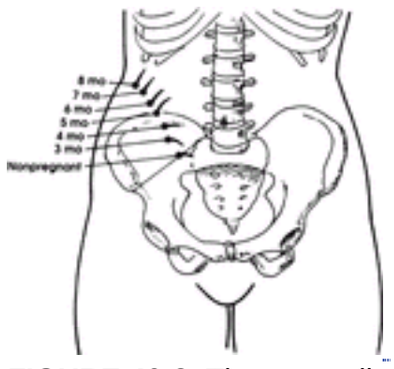

FIGURE 40-3. The appendix is displaced upward and to the right by the uterus during pregnancy. Movement of the appendix during pregnancy may cause the pain of acute appendicitis to be sensed in an atypical position.

Age

The diagnosis of acute abdominal pain is most difficult at the two extremes of age. Communication is limited in infants, and the temporal progression of symptoms may be impossible to elicit. The abdominal wall in infants is not well developed muscularly, and guarding in response to underlying inflammatory processes is diminished. Because fever and leukocytosis are common accompaniments of many childhood illnesses, their diagnostic usefulness is reduced. In contrast, fever and elevated leukocyte counts are much less common in elderly patients, even in the presence of advanced disease. Physical signs of intraperitoneal inflammation likewise are diminished in older patients.

Immunosuppression

Patients who are immunosuppressed may not exhibit appropriate clinical responses to acute intraperitoneal inflammatory processes. Included in this category are patients with the acquired immunodeficiency syndrome, patients receiving long-term corticosteroid therapy, and those undergoing chemotherapy. At special risk are recipients of solid organ transplants. The physical signs of peritonitis (e.g., pain, muscular guarding, rebound) may not be apparent, and fever is often absent. Intraperitoneal infection may progress to septic shock as the first manifestation of disease. Leukocytosis is usually present but decreased; a shift to immature leukocyte forms is apparent. Early use of radiologic evaluation, especially computed tomography (CT), is appropriate.

A syndrome of diffuse abdominal pain, fever, and diarrhea has been reported in patients with severe neutropenia (<100 neutrophils/mL). Patients with acute leukemia or aplastic anemia and those undergoing bone marrow transplantation are at greatest risk. The pathological entity termed *neutropenic colitis* is characterized by diffuse mucosal ulceration, invasive infection with enteric microorganisms, and sepsis. [6] The ascending colon is the intestinal segment most often affected, although the other portions of the colon and the terminal ileum also may demonstrate ulceration. Cecal edema may be demonstrated by CT. Operation is indicated if perforation is suspected, but recovery correlates more closely with rebound of neutrophil counts than with surgical treatment.

Recent Laparotomy

Acute abdominal processes are difficult to evaluate after recent laparotomy. Abdominal pain is universally present but much decreased if laparoscopic techniques were used. Leukocytosis is common in the early postoperative period. Numerous sources for fever exist, including pulmonary atelectasis, urinary infection, phlebitis, and infection in the surgical incision. In evaluating abdominal pain in the postoperative period, it is important to consider the signs and symptoms in relation to the time after operation. Uncomplicated recovery should be accompanied by a steady progression: lessening abdominal pain, resolving biochemical abnormalities, predictable recovery from ileus, and increasing mobility. Failure to achieve this progress warrants investigation. The most common causes of abdominal sepsis after operation relate to inadequacies of surgical technique, such as anastomotic dehiscence. Acute acalculous cholecystitis also may occur in the postoperative setting.

CAUSES OF ACUTE ABDOMEN IN ADULTS

Appendicitis

Acute appendicitis is the most common cause of the acute abdomen in the United States and should be included in the differential diagnosis for every patient presenting with acute abdominal pain. About 250,000 appendectomies are performed in the United States annually, with 2000 deaths resulting from complications of the disease. [7] One in 15 persons develops appendicitis during his or her lifetime.

Appendicitis develops as a result of obstruction of the appendiceal lumen by fecalith or appendiceal calculus or by hyperplasia of submucosal lymphatic tissue. In the initial stages, the pathogenesis of appendicitis resembles that of bowel obstruction. Typically, the first symptom is upper midline or periumbilical pain. Discomfort develops over 1 to several hours and is followed by anorexia, nausea, and vomiting. If the appendiceal lumen remains occluded, inflammatory changes, initially confined to the appendiceal mucosa, become transmural. Contact of the inflamed serosal surface of the appendix with the parietal peritoneum is associated with a change in the pain pattern: a shift to the right lower quadrant. The usual presentation of midline abdominal pain moving to the right lower quadrant is seen less frequently in older patients, in whom diffuse pain and nonlocalized tenderness are more common. Early diagnosis is also more difficult in children, and, as a consequence, perforation and infectious complications are more frequent. [8]

Approximately 2% to 3% of patients with appendicitis have an abdominal mass at initial evaluation. [9] The mass, formed by inflamed omentum and adherent loops of intestine, signifies the development of a phlegmon or an abscess secondary to appendiceal perforation. Perforation also should be suspected if the patient has been symptomatic for more than 24 hours, if the temperature exceeds 38°C, or if the leukocyte count is greater than 15,000 cells/mL.

Abnormal gas patterns on abdominal radiograph, including right lower quadrant ileus or diffuse small bowel air-fluid levels, are present in 62% of patients with uncomplicated appendicitis, in 71% of those with periappendiceal phlegmon, and in 97% of those with gangrenous appendicitis or perforation. [7] An appendicolith is visible on abdominal flat plate in 7%. Pneumoperitoneum is distinctly uncommon, present in less than 1% of cases.

Appendicitis is a clinical diagnosis, supported by carefully selected laboratory and radiologic studies. Patients with compatible historical features and physical examinations do not need additional diagnostic studies, and immediate surgical exploration is appropriate. Ancillary tests are reserved for atypical or equivocal presentations.

Abdominal sonography may be helpful in patients in whom the clinical features of appendicitis are ambiguous. [10] The technique depends on graded compression of the abdomen with the ultrasonographic transducer to displace gas-filled loops of bowel. The appendix is visualized in the shape of a round target with the anechoic lumen surrounded by a hypoechoic and thickened (>2 mm) appendiceal wall. For nonperforated appendicitis, ultrasonography has been reported to have a diagnostic accuracy of 93%, with a negative predictive value of 97%, in selected series. [11], [12] Muscular rigidity accompanying perforated appendicitis prevents adequate abdominal compression and significantly reduces ultrasonographic accuracy. Low diagnostic sensitivity in this group is less important, however, because the need for surgical management is usually clinically obvious. CT scanning is a reliable method for differentiating periappendiceal phlegmon from abscess, and it may be useful in nonoperative management of periappendiceal mass. [13], [14] and [15] CT has a 97% sensitivity and a 91% specificity for acute appendicitis associated with mass. [13] CT may also provide useful information on a broad spectrum of diseases that may mimic appendicitis. [16]

Patients presenting with acute appendicitis not complicated by abdominal mass should undergo emergency appendectomy. The overall perioperative mortality rate among patients with appendicitis is about 0.5%. Although fewer than 0.2% of patients with nonperforated appendicitis die, the mortality rate rises tenfold with the occurrence of perforation. Mortality is clearly related to age, with the highest mortality rates occurring in patients aged more than 50 years. Overall mortality rate is less than 1% for patients less than 50 years of age, 4.5% for those 51 to 70 years of age, and 20% or greater for patients aged 71 to 90 years. [17]

Septic complications occur with increased frequency in the presence of perforation. The wound infection rate is 1.8% when a noninflamed appendix is removed, increasing to 8.5% for inflamed appendices, and 17% with appendiceal perforation. [18] Pelvic abscess occurs in about 15% of patients after resection of a perforated

appendix.

In 10% to 20% of patients explored for presumed appendicitis, a normal appendix is discovered at laparotomy. A literature review of 13,848 cases showed a linear correlation between the rate of appendiceal rupture and diagnostic accuracy. [19] As diagnostic accuracy increases, so does the rate of perforation. If operation is used earlier, diagnostic accuracy decreases, as does the rate of perforation. A negative appendectomy rate of 10% to 15% is widely considered a reasonable compromise. In 30% of cases in which the appendix is not inflamed, a different, specific cause of abdominal pain is identified. Mesenteric lymphadenitis, Meckel diverticulitis, cecal diverticulitis, pelvic inflammatory disease, ectopic pregnancy, and ileitis lead a long list of diseases that can mimic appendicitis.

An appendiceal mass or abscess may be treated by both operative and nonoperative means. [13], [19], [20] Early operative intervention is not favored by most surgeons treating appendiceal abscesses or phlegmons. Initial treatment should include ultrasonographic or CT evaluation, percutaneous drainage of periappendiceal fluid collections, and antibiotics. [21] With resolution of periappendiceal inflammation, elective interval appendectomy is performed. Interval appendectomy is associated with a 0.2% mortality rate and a major complication rate of only 1.2%. [22] Laparoscopic approaches for appendectomy have been developed, adapted to children and adults, and widely disseminated. [23], [24] and [25] The advantages of laparoscopic treatment of appendicitis relative to open appendectomy are not as clear-cut as the advantages of laparoscopy in the treatment of gallstone disease. Laparoscopic appendectomy is associated with a longer operative time, a small reduction in hospital stay, and an earlier return to normal activities. Wound complications are slightly more frequent after laparoscopic appendectomy. Laparoscopic appendectomy is more expensive than open treatment. [26], [27]

Perforated Duodenal Ulcer

Although hospitalization rates for peptic ulceration have declined since 1970, the overall rate of duodenal ulcer perforation has not decreased. Furthermore, the introduction of newer antisecretory drugs has not affected rates of perforation. In two thirds of affected patients, perforation is the first manifestation of duodenal ulcer disease. Whereas the overall rate of peptic ulcer perforation has not decreased, the patient profile has changed, with affected patients being increasingly elderly. Currently, 63% of patients are more than 60 years old, and 44% are older than 70 years. [28], [29] Elderly patients, who more frequently have coexisting medical illnesses, also have higher morbidity and mortality rates.

The patient usually experiences sudden, severe epigastric pain, followed by diffuse abdominal pain. Somatic pain is caused by chemical irritation of the peritoneum by acidic gastric contents. Subphrenic collection of gastric content is associated with right scapular radiation of pain. Respiration worsens symptoms; the patient typically seeks to minimize movement. Physical examination reveals a quiet abdomen with marked muscular rigidity and epigastric tenderness. If perforation has occurred within less than 24 hours, low-grade fever and tachycardia are often present, but hypotension is unusual. Laboratory examinations reveal leukocytosis; mild hyperamylasemia is attributed to resorption of duodenal contents from the peritoneal cavity. Upright abdominal films demonstrate pneumoperitoneum in 80% of cases (Fig. 40-4). If pneumoperitoneum is absent, water-soluble contrast examination may be used to demonstrate perforation.

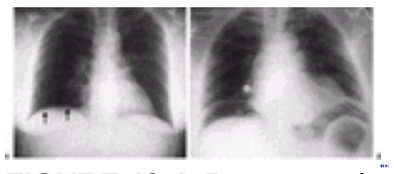

FIGURE 40-4. Pneumoperitoneum accompanying perforated duodenal ulcer. Upright radiographs from two patients reveal (**A**) minimal pneumoperitoneum (*arrows*) and (**B**) massive pneumoperitoneum.

The empty, acid-secreting human stomach contains 102 to 103 organisms, with lactobacilli and aerobic streptococci predominating. Because the initial bacterial inoculum is low, septic consequences associated with duodenal perforation are closely related to the length of time that elapses before definitive treatment. [30] Peritoneal fluid samples obtained within 6 to 12 hours of perforation are culture positive in fewer than 50% of cases; by 24 hours, more than two thirds are positive. After 24 hours, coliform species predominate.

Nonoperative management of documented duodenal ulcer is justified only in unusual circumstances. Operative treatment of perforated duodenal ulcer has four goals: patient safety, peritoneal debridement, closure of the perforation, and alteration of the ulcer diathesis so the risk of recurrent ulceration is minimized. Risk factors that predict operative mortality include serious concurrent medical illness, preoperative shock, and long-standing perforation (i.e., longer than 48 hours). [31] If these factors are absent, definitive ulcer operations may be performed with predictably low mortality and acceptable morbidity. In the presence of one or more risk factors, simple omental patch closure is an expedient and safer alternative to definitive ulcer surgery. [32] The long-term prognosis for patients with perforated acute ulcers is similar to that for patients with perforated chronic ulcers at follow-up of 5 to 6 years. [33] Minimally invasive techniques for treatment of duodenal perforation have been developed. [34], [35]

Laparoscopic omental patch closure may be combined with postoperative anti- *Helicobacter* therapy. This treatment plan assumes that secure closure of the perforation can be obtained and that most ulcers are caused by *H pylori.* Initial results are promising. Minimally invasive approaches to perforation are likely to become standard practice for the future. [36]

Obstruction of the Small Intestine

Obstruction of the small intestine accounts for about 5% of acute surgical hospitalizations; despite significant advances in anesthetic and surgical techniques, the mortality rate still ranges from 3% to 7%. In the United States, 70% to 80% of cases of small bowel obstruction result from postoperative adhesions. In one fourth of cases, adhesive obstruction is preceded by a gynecologic operation, and in another fourth, obstruction follows total or segmental colectomy. After adhesions, in decreasing order of incidence, primary or metastatic carcinoma, external hernias, regional enteritis, and internal hernias are causes of small bowel obstruction. Less common conditions include previous abdominal or pelvic radiation, intussusception, endometriosis, volvulus, and congenital abnormalities.

Intestinal colic is the cardinal symptom of obstruction of the small intestine. Initially, periods of crampy pain centered in the midabdomen may be interspersed with relatively pain-free intervals. If intestinal distention is unrelieved, pain becomes unrelenting. Vomiting is progressive. Bacterial overgrowth in the obstructed segment causes the nature of the vomitus to change with time, becoming feculent. Complete small intestine obstruction is associated with obstipation, although gas and stool distal to the point of obstruction may continue to pass for a short interval after initial symptoms.

The most important diagnostic consideration with these patients is to differentiate partial from complete obstruction. Evaluation begins with plain abdominal radiographs. Complete obstruction is characterized by dilated loops of small intestine with air-fluid levels and no visible gas within the colon (Fig. 40-5). Patients with partial obstruction usually show clear evidence of gas in the colon above the peritoneal reflection in addition to dilated loops of small intestine.

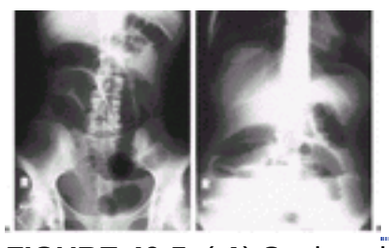

FIGURE 40-5. (**A**) Supine abdominal radiograph of a patient with small bowel obstruction demonstrates dilated loops of small intestine. (**B**) Upright radiograph of

same patient demonstrates air-fluid levels in obstructed segments of the small intestine.

If the diagnosis of complete small intestinal obstruction is unequivocal, additional radiologic studies to demonstrate the site of obstruction or the nature of the obstructive process are neither appropriate nor necessary in most patients. Barium contrast radiography is useful to clarify the diagnosis in patients with atypical symptoms or nondiagnostic plain abdominal films, to differentiate paralytic ileus from mechanical obstruction, and to evaluate selected subsets of patients in whom nonoperative management would be desirable. [37] The last group includes patients with multiple laparotomies for adhesive obstruction, those suffering obstruction in the immediate postoperative period, patients with known intraperitoneal carcinomatosis, patients who have undergone extensive abdominal radiation therapy, and those with Crohn's disease. CT is rapidly displacing traditional contrast radiography in evaluation of small bowel obstruction. [38] CT scanning reveals a point of transition between dilated bowel proximal to the obstruction and collapsed distal bowel. Associated findings, such as tumor masses or hernia defects, may be visualized and may provide additional diagnostic information not available by contrast radiography.

If initial diagnostic studies indicate partial small intestine obstruction, nonoperative management consisting of nasogastric decompression and intravenous hydration is appropriate. Seventy-five percent of patients treated in this manner have clinical and radiographic improvement within 24 hours. An additional 15% respond within 48 hours. Failure to achieve clinical improvement by 48 hours is an indication for operative treatment. There is no evidence that long intestinal tubes are more effective than standard nasogastric tubes in achieving intestinal decompression.

In contrast to partial obstruction, complete small intestine obstruction is an indication for emergency laparotomy because of the risk associated with the development of intestinal strangulation. [39] The presence of intestinal necrosis triples operative mortality rates and increases the incidence of serious postoperative complications fivefold to tenfold. In patients with complete obstruction of the small intestine, preoperative diagnosis of strangulation cannot be made or excluded by any currently available radiologic test, clinical parameter, or combination of parameters or by experienced clinical judgment. In one prospective evaluation, preoperative clinical indices and judgments of senior attending surgeons were assessed in 51 consecutive patients about to undergo laparotomy for complete small bowel obstruction. [40] The signs usually considered diagnostic—continuous abdominal pain, fever, peritoneal irritation, leukocytosis, and acidosis—were not sensitive, specific, or predictive for strangulation. Senior surgeons, aware of all preoperative data, correctly predicted strangulation in only 10 of 21 affected patients (48% sensitivity). Nonoperative treatment of complete small intestine obstruction entails a calculated risk (30%) of delayed definitive treatment of intestinal ischemia. [40]

Resuscitation before operative intervention should focus on the following: correction of hypoxemia caused by respiratory restriction secondary to abdominal distention; replacement of intravascular volume deficits caused by losses through vomiting, bowel wall edema, and extravasation of fluid into the peritoneal cavity; and correction of serum electrolyte abnormalities. Because of intralumenal bacterial overgrowth and the potential for septic complications associated with intestinal obstruction, broad-spectrum antibiotics should be administered preoperatively. Gastric decompression is necessary to lessen the risk of pulmonary aspiration during anesthetic induction.

Patients who have been treated for cancer and who subsequently develop obstruction of the small intestine present a special challenge. [41] It is important to realize that 25% to 40% of such patients do not have recurrent or metastatic cancer as the cause of their obstruction. For these patients, lysis of adhesions or herniorrhaphy provides long-term relief. If localized recurrent or metastatic tumor is the cause of obstruction, significant palliation may be obtained by operative treatment. Advanced metastatic tumor causing small intestine obstruction, exemplified by ovarian carcinomatosis, has a dismal prognosis and may be a contraindication to operative treatment. [42]

As with other causes of acute abdomen, elderly patients affected by small bowel obstruction tend to present with more advanced disease and with fewer laboratory and physical findings. In one report, 35% of patients older than 70 years who developed intestinal gangrene had no abnormal physical signs or laboratory parameters. [43] Resultant diagnostic uncertainty in elderly patients is associated with operative delay and a 40% increase in perioperative morbidity.

Minimally invasive approaches have been adapted to the treatment of small intestinal obstruction. [44], [45] Intestinal distention may limit abdominal insufflation in some cases, but laparoscopic enterolysis is associated with more rapid return of bowel activity and shorter hospitalization.

Colonic Diverticulitis

Acute abdominal processes involving the large intestine and emergency surgical procedures used to treat colonic diseases are associated with higher mortality and morbidity rates than are diseases in any other organ system. The treatment of acute diverticulitis illustrates many of these difficulties.

Colonic diverticula are extremely common and are clearly age related in the United States. By the seventh decade, more than 50% of the population can be demonstrated to harbor colonic diverticula. The sigmoid segment is involved in 90% of cases and is the only site of diverticulosis in half. Isolated right colonic or cecal diverticula are present much less frequently, demonstrated in about 2% of examinations. Although diverticulosis is common, symptomatic diverticulitis develops in few patients with colonic diverticula; lifetime risk approximates 5%. [46]

The signs and symptoms associated with acute diverticulitis depend on the colonic segment involved, the degree of pericolonic inflammation, and the development of extracolonic complications. Classically, patients present with left-sided lower abdominal pain, fever, and obstipation. Nausea and vomiting are usually not prominent. Mild abdominal distention is variably present. Leukocytosis is so common that in its absence the diagnosis should be questioned. Physical examination may reveal a palpable, tender mass in the left lower quadrant, but the absence of a mass does not exclude the diagnosis of diverticulitis.

Acute diverticulitis is a clinical diagnosis. Therapy, consisting of parenteral antibiotics and bowel rest, should be initiated empirically if appropriate symptoms exist. In 85% of cases, symptoms abate within 3 days of initiating treatment. [47] After patients become asymptomatic, radiographic or endoscopic evaluation of the colon should be performed to confirm the diagnosis of diverticulitis and to exclude other diseases, specifically colonic adenocarcinoma. If symptoms do not subside promptly, or if a complication of diverticulitis is suspected, additional radiologic evaluation is indicated.

Plain abdominal radiographs, usually obtained in symptomatic patients, do not reveal any abnormality in 70% of cases. Although findings consistent with diverticulitis are demonstrated in approximately 60% of cases by barium enema, complications of diverticulitis can be detected in only 30% to 40%. The most common findings are localized extravasation of contrast material, indicating a contained perforation, or a fixed, narrowed segment of colon. Because of the volume of contrast used and the pressure under which it is infused, barium enema carries a definite risk of exacerbating diverticulitis.

When diverticular complications are suspected, CT has been increasingly used as the initial test. [48], [49] CT has several advantages in this clinical situation: it causes much less discomfort than a barium enema, the risk of perforation is negligible, and extracolonic complications of diverticulitis can be evaluated. Pathological changes of diverticulitis detectable by CT include intramural colonic thickening, mesenteric edema, pericolonic and mesenteric abscesses, phlegmon formation, and pneumoperitoneum. [50] If intravenous contrast is not administered, visualization of contrast material within the bladder is presumptive evidence for colovesical fistula. CT examination has a sensitivity of 65% for uncomplicated disease, with a 90% to 100% sensitivity for detecting diverticulitis complicated by perforation, abscess, or fistula. [49], [51]

The ability of CT to define the complications of acute diverticulitis, such as pericolonic abscess, allows for CT-guided percutaneous needle aspiration and catheter drainage in properly selected patients. [52], [53] The goal of percutaneous drainage of diverticular abscesses is to minimize the need for colostomy formation and to optimize the conditions for resection with primary anastomosis while not compromising patient safety. Patients with generalized peritonitis, pneumoperitoneum, or colonic obstruction secondary to diverticulitis are not candidates for this treatment approach. In addition, small pericolic or intramesenteric abscesses that could be resected en bloc with the sigmoid colon are not appropriately treated with percutaneous drainage.

A 2.5- to 5-mm (8- to 16-French) catheter usually is used. A sinogram should be obtained through the catheter within 48 hours of placement to evaluate abscess cavity size and to demonstrate fistulization to the colon. Catheter injection should be repeated periodically to evaluate progress. CT-guided percutaneous drainage of peridiverticular abscesses is feasible in about 75% of patients. In a selected series of patients, catheter drainage was associated with resolution of sepsis in 89%. [54] Fistulous communication with the colon was demonstrated radiologically in 47% but was feculent in only 16%. In 74% of treated patients, single-stage sigmoid resection with primary anastomosis was performed subsequently without complication.

The presence of pneumoperitoneum, generalized peritonitis, or colonic obstruction associated with diverticular abscess is an indication for emergency operation. Operative goals include resection of the diseased segment of colon and peritoneal debridement. In the presence of active intraperitoneal infection or a mechanically unprepared bowel, resection of the diseased segment, formation of a proximal colostomy, and closure of the rectum (i.e., Hartmann procedure) comprise the safest surgical option. [55] Intestinal continuity is restored 2 to 3 months after resolution of the septic process, often laparoscopically. [56]

Acute Cholecystitis

Impaction of a gallstone in the cystic duct causes biliary colic, characterized by pain in the epigastrium or right upper quadrant of the abdomen. Biliary colic begins gradually before reaching a plateau of steady pain, which typically lasts several hours. Discomfort relents as the stone dislodges. The pain of biliary colic is severe and is frequently accompanied by nausea and vomiting. Symptoms classically are associated with the ingestion of a fat-containing meal, although they may also begin during fasting.

Continued obstruction of the cystic duct initiates an inflammatory response in the gallbladder wall. The presence of obstruction plus inflammation distinguishes acute cholecystitis from simple colic. Because of continuing ductal obstruction, the pain of acute cholecystitis may persist for many hours to days. If the inflammatory process progresses to involve the serosal surface of the gallbladder, adjacent parietal peritoneum is irritated. In this instance, pain becomes more intense and more clearly localized to the right upper quadrant. Movement of an inflamed gallbladder against the parietal peritoneum during breathing may inhibit deep inspiration, producing a positive Murphy sign. Accompanying fever is usually low grade. Most patients with uncomplicated cholecystitis have mild leukocytosis, with leukocyte counts ranging from 12,000 to 16,000 cells/mm [3]. Levels of serum bilirubin, alkaline phosphatase, and amylase may be nonspecifically elevated, usually not more than twice normal.

Although 95% of patients with acute cholecystitis have calculous biliary disease, the demonstration of cholelithiasis in patients with abdominal pain does not confirm the diagnosis of cholecystitis. Asymptomatic gallstones are present in 30% to 40% of persons older than 40 years of age, and only 15% to 35% of patients who have symptoms consistent with cholecystitis are proven to have the disease. Because of the high frequency of symptomless gallstones, additional tests are necessary to support the clinical diagnosis.

Ultrasonography is the preferred initial screening test for acute cholecystitis. Ultrasound has advantages of cost, portability, and ease of performance. In addition to detecting gallstones with 95% sensitivity, the procedure also provides information regarding other organs in the upper abdomen. Secondary findings that support the diagnosis of acute cholecystitis include gallbladder wall thickening, distention of the gallbladder, pericholecystic fluid, and sonographic lucency within the gallbladder wall. [57] Thickening of the gallbladder wall greater than 3 mm predicts acute cholecystitis with more than 90% accuracy, but this finding must be interpreted within the clinical context, because wall thickening has also been reported with portal hypertension, hypoalbuminemia, and ascites, as well as after gallbladder contraction when patients have not fasted.

A sonographic Murphy sign is elicited by placing the ultrasonographic transducer over the gallbladder and applying pressure or asking the patient to inspire deeply. In the presence of gallstones, a positive sonographic Murphy sign predicts acute cholecystitis in 90% of cases.

Cholescintigraphy is based on iminodiacetic compounds that are taken up by hepatocytes and secreted into bile. Technetium 99 labeling permits visualization after intravenous injection. The upper abdomen is imaged for 2 to 3 hours after injection, and delayed images may be obtained up to 24 hours. In normal patients, the liver is imaged first, followed by the gallbladder, common bile duct, and duodenum. If the common bile duct and duodenum are visible but the gallbladder is not, cystic duct obstruction is presumed to be present. If neither the gallbladder nor the common bile duct can be seen, common duct obstruction may be present. Severe hepatocellular disease and hyperbilirubinemia also may produce this pattern.

The gallbladder cannot be seen by cholescintigraphy in half of patients with chronic cholecystitis, even if the cystic duct is not acutely obstructed. False-positive findings also may result from failure to fast, alcoholic liver disease, and long-term parenteral nutrition. Because of these limitations, cholescintigraphy usually is reserved for patients with normal ultrasonography but strong clinical suspicion of acute cholecystitis and for those with gallstones who have no ancillary ultrasonographic findings of cholecystitis. [58]

Optimal management of patients with acute cholecystitis is laparoscopic cholecystectomy, both open operative and laparoscopic approaches are available in properly selected patients. [59], [60], [61] and [62] A detailed discussion of laparoscopic cholecystectomy may be found in Chapter 100 and Chapter 161.

Mesenteric Ischemia

Acute visceral ischemia is a prototypical, common, and frequently misdiagnosed cause of acute abdomen in adults. The four major ischemic syndromes are mesenteric embolism, acute mesenteric thrombosis, low-flow mesenteric ischemia, and iatrogenic mesenteric ischemia. Each syndrome is life-threatening, with mortality rates exceeding 50%. Early recognition, aggressive resuscitation and monitoring, and directed, specific therapy are essential.

Mesenteric embolism accounts for about 50% of cases of acute visceral ischemia. The embolic source is typically cardiac and is derived from mural thrombus associated with atrial fibrillation or myocardial infarction. Embolism to visceral vessels of atheromatous debris from the thoracic aorta is much less common.

Mesenteric embolism is accompanied by sudden severe epigastric and midabdominal pain. Forceful vomiting and evacuation of stool commonly follow the onset of pain. The general physical examination may reveal an irregularly irregular pulse of atrial fibrillation or a cardiac murmur. Early after embolization, physical examination of the abdomen may be entirely unremarkable; a classic presentation is severe abdominal pain out of proportion to physical findings. Abdominal distention, guarding, and absence of bowel sounds are associated with intestinal infarction and imply disease progression.

No laboratory tests are pathognomonic for mesenteric embolism or visceral ischemia. Hemoconcentration, leukocytosis, and acidosis, like definite physical findings, indicate advanced disease. Electrocardiography may confirm cardiac conduction abnormalities suspected on physical examination. Diagnosis depends on emergency diagnostic angiography. Emboli to the superior mesenteric artery (SMA) typically lodge at branch points of the artery distal to its origin. The inferior pancreaticoduodenal artery and the middle colic artery, the first two branches of the proximal SMA, may be spared with the embolus impacted distally (Fig. 40-6). After diagnosis, systemic heparinization is instituted, vigorous resuscitation with central cardiac monitoring is begun, and the patient is taken for emergency operation. Surgical therapy involves isolation of the proximal SMA and extraction of the embolus through a longitudinal arteriotomy. After restoration of flow, an assessment of intestinal viability is performed, and frankly necrotic bowel is resected.

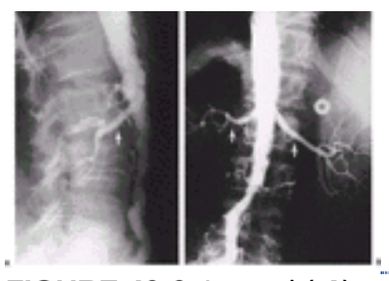

FIGURE 40-6. Lateral (**A**) and anteroposterior (**B**) emergency visceral angiograms demonstrate the absence of celiac and superior mesenteric artery filling in a patient with acute mesenteric thrombosis. Patent renal arteries are indicated by *arrows.*

Mesenteric thrombosis causes acute visceral ischemia after atherosclerotic narrowing of the SMA exceeds a critical level or becomes complete. Although acute thrombosis can occur without antecedent symptoms, a history of postprandial abdominal pain and weight loss, termed *chronic intestinal angina,* may exist. Unlike lesions resulting from mesenteric embolism, those associated with thrombosis tend to occur at the origin of the SMA from the aorta. Acute symptoms, abdominal

physical findings, and laboratory tests are similar to those listed for mesenteric embolism. Diagnosis relies on emergency visceral angiography. Emergency revascularization procedures involve construction of an arterial conduit, usually with an autogenous vein, from the aorta to the vessel distal to the point of obstruction.

Nonocclusive visceral ischemia occurs in low-flow states that cause intestinal vasoconstriction. Shock, decreased cardiac output, dehydration, hypovolemia, and inappropriate use of vasoconstrictive or inotropic agents can cause nonocclusive mesenteric ischemia. Patients typically have a history of recent myocardial infarction, congestive heart failure, or arrhythmias before development of intestinal ischemia. Visceral angiography reveals severe vasoconstriction of mesenteric vessels, which typically have a pruned appearance (Fig. 40-7).

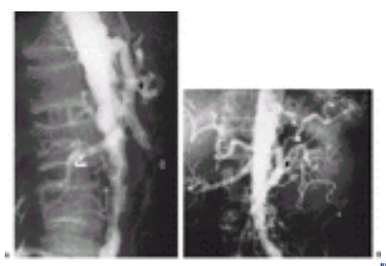

FIGURE 40-7. Lateral (**A**) and anteroposterior (**B**) postoperative angiograms from the patient shown in Figure 40-6 show the prosthetic graft (*black arrow*) supplying the superior mesenteric artery (*open arrow*). The renal artery (*curved arrow*) is similar to that shown in Figure 40-6.

Treatment of nonocclusive mesenteric ischemia requires restoration of intravascular volume and hemodynamic stability, elimination of contributing pharmacological agents, and efforts to relieve mesenteric vasospasm. Papaverine infusion through catheters placed angiographically into the SMA has been reported to reverse vasoconstriction. Peripheral infusion of glucagon (2–4 mg/hour) also has been used for this purpose. Operative therapy is not therapeutic in nonocclusive mesenteric ischemia and is reserved for resection of necrotic bowel.

Iatrogenic mesenteric ischemia occurs most commonly after angiographic procedures or operations on the aorta. Angiography may cause intestinal ischemia by dislodging atheroma from a diseased vessel wall or by vessel dissection or intimal flap formation. Aortic aneurysm resection is the operation most often associated with visceral ischemia because of involvement of the inferior mesenteric artery with the aneurysm. Clinically apparent colonic injury occurs in 1% to 2% of cases. Sigmoid colonic ischemia is heralded by bloody diarrhea; the diagnosis may be confirmed by urgent flexible sigmoidoscopy.

Abdominal Aortic Aneurysm

Acute symptoms caused by abdominal aortic aneurysm are usually a result of expansion or rupture or, less commonly, are secondary to dissection, distal embolism, or thrombosis. Through mechanisms that are not well understood, acute expansion of the aortic wall before rupture is associated with severe pain in the back, flank, or abdomen. Back pain is also typical of small tears, which produce leakage of blood into the retroperitoneum before catastrophic intraperitoneal hemorrhage. About 20% of ruptured abdominal aortic aneurysms present with these prodromal symptoms.

The classic clinical presentation of ruptured abdominal aortic aneurysm is diffuse abdominal pain, hypotension, and pulsatile abdominal mass. Pain may radiate to the back, flank, or groin. Profound shock implies free intraperitoneal leakage of blood. The most frequent diagnostic errors result from failure to palpate an abdominal mass, and the most frequent misdiagnosis is myocardial infarction.

If ruptured abdominal aortic aneurysm is suspected, additional diagnostic tests are not required. Subsequent resuscitation and therapy should take place in an operating room where immediate laparotomy can be performed. The postoperative mortality rate is about 50%. Acute renal failure, colonic ischemia, and lower extremity ischemia are common postoperative complications.

OPERATIVE THERAPY FOR ACUTE ABDOMEN

The treatment of patients with an acute abdomen has changed fundamentally since the 1980s. The proliferation of diagnostic and therapeutic modalities has provided greater flexibility in the treatment of these patients. The therapeutic hierarchy now includes endoscopic interventions, invasive radiologic procedures, and laparoscopy, in addition to supportive medical care and laparotomy. The proper treatment of these patients requires a sophisticated understanding of the power and limitations of each approach, as well an appreciation of the correct sequence for their application. For example, whereas supportive medical therapy is still the mainstay for treatment of acute biliary pancreatitis, endoscopic papillotomy is appropriate for selected subgroups of patients. Similarly, endoscopic detorsion is the accepted initial therapy for acute sigmoid volvulus, with operative treatment reserved as a secondary option. In most cases, peridiverticular abscess may now be treated by CT-guided percutaneous drainage, converting what was formerly an emergency laparotomy into elective colectomy. Laparoscopic cholecystectomy has become standard treatment for acute cholecystitis; conversion to open cholecystectomy is reserved for when the laparoscopic approach is anatomically difficult or unsafe. These approaches are detailed in other chapters of this text and are not discussed further here. Nonetheless, laparotomy is still required in many patients with acute abdominal disease.

After the initial history and physical examination, a basic question must be answered: Is immediate operation necessary? Immediate intervention is required if the suspected pathological process is progressive and potentially fatal. This criterion is met with intraperitoneal hemorrhage, for example, ruptured abdominal aortic aneurysm or ruptured ectopic pregnancy. In these circumstances, laparotomy becomes a method of diagnosis as well as a means of treatment.

If immediate operation is not immediately necessary, the next question becomes: Will operation ultimately be necessary? If initial evaluation reveals pneumoperitoneum, radiologic evaluations to differentiate perforated duodenal ulcer from perforated colonic diverticulum are not required; the processes may be distinguished easily at operation. In fact, the time required for additional diagnostic tests may be detrimental if therapeutic delay permits the disease process to progress. Instead, preoperative efforts should be expended to prepare the patient for anesthesia and operation. If the need for operation is uncertain, additional tests to define precisely the diagnosis are justified. As the processes of diagnosis and resuscitation proceed, a timetable is essential; worsening clinical evaluation mandates earlier surgical intervention.

Preparation for Operative Therapy

After a decision for operative intervention has been made, expeditious efforts to prepare the patient are undertaken. The first priority is assessment of operative risk.

General anesthesia has an associated risk, even without a surgical procedure. Improvements in anesthetic technique and agents have decreased mortality from approximately 1:10,000 in the 1950s to about 1:100,000 or less for healthy patients in 1997. About 50% to 75% of deaths directly attributable to anesthesia are the result of human error and usually are related to improper airway management: unrecognized esophageal intubation, unrecognized extubation, and unrecognized disconnection from the ventilator with resultant inadequate ventilation. The routine use of capnometry and pulse oximetry, in addition to other noninvasive monitors, has reduced significantly the incidence of problems associated with intubation.

Coronary Artery Disease Perioperative cardiac events are the leading cause of death after laparotomy, and much of the preoperative risk assessment is directed toward detecting the presence of ischemic heart disease. Preoperative congestive heart failure, recent myocardial infarction, and unstable angina are significant risk factors for postoperative cardiac complications. Diabetes mellitus, atherosclerotic peripheral vascular disease, and hypertension also confer risk but less than congestive heart failure or unstable angina. For patients with valvular heart disease, perioperative cardiac risk is proportionate to the severity of associated congestive heart failure, pulmonary hypertension, and arrhythmias. Stable angina and advanced age alone do not appear to be significant predictors of perioperative cardiac morbidity. Previous coronary artery bypass surgery appears to lower the risk of adverse postoperative cardiac events. The resting 12-lead electrocardiogram is the best screening test for ischemic heart disease, although the evaluating physician must be aware that resting electrocardiography may not detect all patients with ischemic heart disease and is not sensitive for prior subendocardial myocardial infarction. Whereas stress evaluations may be appropriate for elective surgical procedures, this level of sophistication is not available in emergency cases. A history of prior myocardial infarction should be sought in every case. Studies have

demonstrated that the risk of repeated infarction is related to the time elapsed since the previous myocardial infarction, with the incidence of reinfarction at 6% after 6 months. [63] Reinfarction for patients undergoing emergency noncardiac surgery usually occurs within 48 hours and causes extreme morbidity, with an associated mortality of 20% to 50%. [64] Patients with three-vessel or left main coronary artery occlusion are at increased risk. Intra-abdominal procedures lasting longer than 3 hours are associated with increased rates of reinfarction. Aggressive invasive hemodynamic monitoring and pharmacological intervention can reduce the risk of postoperative reinfarction.

Congestive Heart Failure The presence of congestive heart failure is a strong predictor of postoperative cardiac morbidity, and elective surgical procedures should be deferred if it this condition detected. Patients with ventricular dysfunction are at high risk during emergency laparotomy because they are intolerant of the intravascular fluid shifts associated with inflammatory disease processes and the myocardial depression associated with anesthetic agents. When emergency laparotomy is necessary, aggressive perioperative management should attempt to optimize cardiac output.

Hypertension Hypertension is the most common chronic medical condition in surgical patients. The incidence of intraoperative hypotension and myocardial ischemia is increased for patients with uncontrolled hypertension relative to those with well-controlled hypertension. When operation is elective, hypertension should be treated preoperatively to the patient's normotensive levels, and antihypertensive medications should be continued postoperatively. Hypertensive patients have exaggerated responses to painful stimuli during general anesthesia.

Pulmonary Disease Preexistent pulmonary disease is important in planning anesthetic management during emergency laparotomy. Pulmonary disease may be categorized as acute or chronic and restrictive or obstructive. Restrictive disease, defined by reduction of lung volumes, may be caused by processes that are intrinsic or extrinsic to the lungs. Intrinsic restrictive disorders that are commonly encountered in the context of emergency laparotomy include adult respiratory distress syndrome. Extrinsic causes of restrictive pulmonary disease include obesity and chest wall deformity. Safe management of patients with restrictive pulmonary disease requires recognition and alterations in ventilatory techniques but less preoperative preparation than for obstructive disease. Obstructive disease is characterized by reduced flow rates on pulmonary function tests. Obstructive pulmonary disease can be either chronic (emphysema) or acute (asthma). For most patients, obstructive pulmonary disease has a reversible component; this contribution to obstruction should be treated preoperatively by bronchodilators and therapy to mobilize secretions. Asthmatic patients frequently have reactive airway disease, in which the introduction of the endotracheal tube may induce bronchospasm during anesthetic induction and emergence. Because of the increased risk of postoperative complications, patients with preexisting pulmonary disease should be extubated only when they meet adequate extubation criteria, based on preoperative functional data. Alterations of pulmonary mechanics and resulting postoperative complications are greatest after upper abdominal surgery. Vital capacity and functional residual capacity are reduced, with lowest levels in the first 2 days postoperatively. Therapy with incentive spirometry seeks to restore functional residual capacity to preoperative levels. In patients with pulmonary compromise, the use of epidural narcotics for postoperative pain control is an important advance, allowing earlier extubation for patients with intrathoracic and upper abdominal surgery and earlier restoration of pulmonary function to preoperative levels.

Obesity Obesity represents a major comorbid factor for emergency laparotomy. Morbid obesity is defined as twice ideal body weight. Obese patients exhibit restrictive pulmonary defects as a result of reduced functional residual capacity; this deficit is exacerbated by the supine position necessary for laparotomy. Because of reduced functional residual capacity, obese patients frequently desaturate on anesthetic induction. Obese patients also have increased rates of pulmonary hypertension, right-sided heart failure, and occult coronary artery disease. Gastroesophageal reflux, a result of increased intra-abdominal pressure, increases risk of aspiration on induction of anesthesia. Fatty tissues may cause prolonged effects of lipid-soluble anesthetics.

Preoperative Resuscitation

When diseases that cause intraperitoneal hemorrhage are suspected (e.g., ruptured abdominal aortic aneurysm or ruptured ectopic pregnancy), preoperative resuscitation should be conducted within the operative suite. For these diseases, restoration of intravascular volume requires control of the source of hemorrhage, and laparotomy should proceed immediately. Most other causes of the acute abdomen have an inflammatory or infectious pathogenesis, with potential systemic disturbances in cardiovascular, pulmonary, and renal function. In these instances, preoperative resuscitative efforts must be undertaken before anesthetic induction. No patient should undergo anesthetic induction while he or she is suffering from septic shock. Effective intravascular volume should be restored by infusion of crystalloid solutions. Electrolyte abnormalities, commonly observed with repeated vomiting or small bowel obstruction, should be addressed. Coagulation abnormalities, if present, may require correction by transfusion of plasma factors or platelets. Preoperative administration of intravenous antibiotics is appropriate in almost all cases. Initial antibiotic choice should be dictated by the presumptive diagnosis and anticipated associated organisms. For example, perforation in acute appendicitis is accompanied by intraperitoneal spillage of colonic organisms, including gram-negative species and anaerobes such as *Clostridium perfringens.* Second- or third-generation cephalosporins are appropriate preoperatively. Subsequent antibiotic therapy can be guided by the findings at operation and by intraoperatively obtained peritoneal cultures.

Preoperative and Intraoperative Monitoring

Patients undergoing general anesthesia are routinely monitored by pulse oximetry, exhaled ventilatory capnometry, and electrocardiography. An esophageal or precordial stethoscope and esophageal temperature probe are standard. Stimulating electrodes can be applied to monitor the level of muscular paralysis. A Foley catheter is an atraumatic and inexpensive means to monitor renal perfusion.

Additional, invasive hemodynamic monitoring is appropriate for selected patients undergoing emergency laparotomy. Invasive monitoring is appropriate for patients with significant preexisting cardiopulmonary dysfunction and for those with evidence of preoperative shock or hemodynamic instability. Right or left ventricular dysfunction, pulmonary hypertension, or hemodynamically significant valvular disease are clear indications for insertion of a flow-directed pulmonary artery catheter (Swan-Ganz catheter). The catheter travels through the central venous circulation and the right heart to the pulmonary artery. Right atrial and pulmonary artery pressures can be measured directly. Left atrial pressure is approximated by the wedged pulmonary artery pressure. Thermodilution can be used to measure cardiac output; systemic and pulmonary vascular resistances then can be calculated. In most instances, the decision to insert a pulmonary artery catheter should prompt insertion of a radial artery catheter to provide direct measurement of arterial pressure and access to arterial blood samples.

Diagnostic Laparoscopy for Acute Abdomen

Diagnostic laparoscopy is a safe and accurate tool that can be used to rule out acute intra-abdominal disease, thus avoiding the need for a laparotomy. This technique is especially appealing in critically ill patients, in whom the morbidity and mortality associated with a nontherapeutic laparotomy can be substantial. In many cases, definitive therapy can be also undertaken using minimally invasive techniques. Moreover, if conversion to a formal laparotomy is required, the information obtained from the diagnostic laparoscopy allows the surgeon appropriately to place and minimize the size of a laparotomy incision.

Although diagnostic laparoscopy can be undertaken using local anesthesia and intravenous sedation, as a general rule the procedure should be undertaken in the operating room with the patient under general anesthesia. Patient positioning and manipulation during the procedure, airway control, and immediate conversion to laparotomy are all facilitated by general anesthesia. Patients should be placed in the supine position, with their arms tucked. This allows the surgeon to stand toward the head of the operating table when laparoscopic evaluation or treatment of pelvic structures is being undertaken. An orogastric tube and Foley catheter should be inserted to decompress the stomach and bladder, respectively, minimizing the risk of an injury to either structure during the procedure. Access to the abdomen is obtained by an "open" approach using a small periumbilical incision. The abdomen is then insufflated to 15 mm Hg with carbon dioxide. Once adequate pneumoperitoneum is established, additional trocars for instrumentation can be placed under direct vision as needed. A general inspection in a systematic fashion of all four abdominal quadrants is undertaken using a 30° or 45° laparoscope. Any free fluid that is encountered should be sent for Gram stain and culture. Exposure of upper abdominal structures is assisted by placing the patient in the reverse Trendelenburg position (head-up tilt). Placing the patient in a severe Trendelenburg position, which retracts the small bowel cephalad out of the pelvis, facilitates exposure of pelvic structures. Finally, tilting the operating table laterally from side to side aids with exposing the right and left paracolic gutters of the abdominal cavity. Atraumatic graspers are used to inspect the liver, gallbladder, stomach and duodenum, colon, appendix, and pelvic organs. The small intestine should be inspected from the ileocecal valve to the ligament of Treitz. Opening the gastrocolic ligament allows one to access the pancreas and posterior stomach for inspection.

Performance of Laparotomy

The placement of incision is influenced strongly by the presumed pathological process and by the certainty of diagnosis. Strong suspicion of acute appendicitis dictates exploration by a small transverse incision in the right lower quadrant. The oblique and transversus muscles are separated along the direction of their fibers to expose the peritoneum. Open cholecystectomy usually is performed through a right subcostal incision. In many instances, the precise diagnosis is not known at the time of laparotomy; in these cases, a vertical midline incision may be preferred. A midline laparotomy incision allows access to all four quadrants of the abdomen and permits mobilization of intraperitoneal structures to expose retroperitoneal organs. Midline incisions do not interfere with placement of ostomies or drains.

A careful, systematic abdominal exploration ensues after incision. The intraperitoneal organs are examined sequentially, usually starting in the upper abdomen with the stomach, the first portion of the duodenum, the liver, and the gallbladder. The ligament of Treitz is located to the left of the midline beneath the transverse colon,

and the small bowel is inspected from the jejunum to the ileocecal valve. The transverse and sigmoid segments of the colon can be inspected without additional dissection, as can the anterior surfaces of the ascending and descending colon. The greater omentum and visceral and parietal peritoneal surfaces are easily visualized. A thorough inspection of retroperitoneal organs requires additional dissection. The posterior surface of the stomach and the pancreas may be exposed by dividing the gastrocolic omentum, entering the lesser sac. The posterior surfaces of the ascending or descending colon may be viewed directly only after mobilization from the retroperitoneum.

The peritoneal cavity ordinarily contains a minute amount of free fluid, enough to lubricate the peritoneal surfaces. Increased amounts of fluid may represent inflammatory exudate, ascites, and chylous leakage. Peritoneal fluid should be sampled for bacterial and fungal culture and Gram stain. White blood cell count and differential, determination of fluid amylase and protein content, and cytology may also be performed. Organ masses are noted, and an attempt is made to distinguish inflammatory and neoplastic masses. The omentum and peritoneal surfaces are inspected for mass involvement; intraoperative biopsy usually is performed more easily from these sites. Organ perfusion is assessed clinically, based on color, tissue turgor, and the presence of pulsatile flow in small mesenteric vessels. When these clinical signs are equivocal, intraoperative Doppler ultrasonography and intravenous fluorescein may be used to provide additional information. Lymph node enlargement should be noted and may prompt intraoperative biopsy.

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CHAPTER 41

Robert W. Summers

APPROACH TO THE PATIENT WITH ILEUS AND OBSTRUCTION

Ileus is a pathophysiological state of inhibited motility in the gastrointestinal tract that may be temporary (reversible) or permanent. The adjectives *paralytic* and *adynamic* emphasize the activation of inhibitory mechanisms. Motor paralysis and paresis may be better terms than ileus because they describe the physiological malfunction more clearly, but neither enjoys wide use. The term *functional obstruction* is a possible alternative, but it is confusing because “functional” could imply a psychological component to some, as in the functional bowel disorder, and “obstruction” implies an anatomic impediment to flow; neither is intended, however.

Pseudoobstruction is often used in describing a chronic abnormality of function simulating mechanical obstruction but having no anatomic cause. Sudden massive idiopathic dilation of the colon is called *acute colonic pseudoobstruction,* or Ogilvie syndrome in more common use. *Toxic megacolon* is a special sort of ileus in which severe transmural inflammation produces atony of the colonic muscle, and at the same time the mucosal barrier is disrupted, resulting in systemic toxemia.

Obstruction implies blockage of the gut at one or more sites; the adjective *mechanical* emphasizes the anatomic nature of the problem. *Obturation* is a synonym that implies that the process is intralumenal. The obstruction is complete if there is complete inability of intestinal contents to pass through the digestive tract, and it is partial if passage continues but is impaired. Obstruction is described as *simple* if the lumen is occluded at only one location. The term *closed loop obstruction* is used if the lumen is obliterated at two sites. This situation is often accompanied by impairment of the blood supply. If the blood supply is inadequate to maintain the viability of the gut, the term *strangulated obstruction* is used to describe the condition.

PATHOPHYSIOLOGICAL CONSIDERATIONS

The pathophysiology of simple and strangulated obstructions is summarized in Figure 41-1.

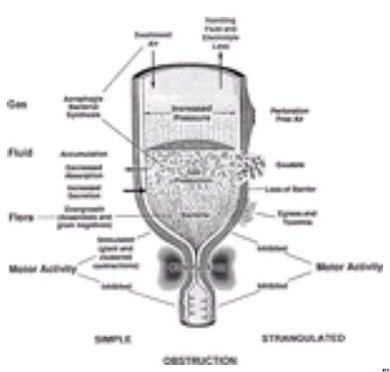

FIGURE 41-1. Pathophysiology of simple (**left**) and strangulated (**right**) obstruction in the small intestine.

Changes in Blood Flow

Strangulated obstruction, a potentially lethal event, impairs blood flow and requires urgent recognition and management. Few investigators have studied the problem in humans because of the invasive methods required to measure blood flow and the critical nature of the condition. The major threat to life in closed loop obstruction is impairment of blood flow in large vessels. The vascular supply can be compromised through external compression of the bowel or its mesentery by adhesions, hernial orifices, tumors, torsion (i.e., volvulus), or intussusception. Except in closed loop obstruction, an increase in intralumenal pressure causes local ischemia only as a late event. Pressures reach only 8 to 10 cm H_2O in simple obstruction and are not high enough to produce transmural ischemia. [1] Total mesenteric blood flow actually increases in experimental simple obstruction, and an increase in muscle blood flow occurs during heightened motor activity because some blood is shunted from mucosa to the muscle. [2]

The mucosa has the greatest metabolic need for blood and is the most sensitive to ischemia. Within minutes after vascular occlusion, damage to the tips of the villi is manifest by sloughing of the epithelial cells. [3] If ischemia continues for 30 to 60 minutes, the villi are almost completely denuded, and only the crypt epithelium is preserved. The major early consequences of these changes are impairment of the basic transport processes, loss of the protective barrier to gut bacteria and their toxic products, and exudation and hemorrhage into the lumen. [4] Failure to recognize and treat strangulated obstruction inevitably leads to infarction, necrosis, and perforation of the bowel wall with peritonitis or sepsis and a mortality rate of 30%.

Ileus can result from arterial blockade caused by mesenteric vasculitis, atherosclerosis, or emboli. The converse is not the case; that is, ileus does not impair large arterial blood flow, and the intralumenal pressures generated with ileus are insufficient to compromise mucosal blood flow.

Changes in Bowel Flora

The clinical picture in ileus and obstruction is affected by the bowel flora. The flora of the stomach, jejunum, and proximal ileum are mainly gram-positive and facultative organisms. Organisms are present in low concentrations (about 10^3–10^4 organisms/mL of fluid). Aerobic lactobacilli, streptococci, staphylococci, and fungi

are normal flora in this region, whereas anaerobes and coliforms are rare and are present in even lower concentrations. [5] The organisms in the colon are drastically different in numbers and kind. About 10^{9} to 10^{12} organisms/g of feces exist in the colon, constituting up to 40% of the fecal dry weight. Most (99%) are non–spore-forming anaerobic rods, including *Bacteroides* species, lactobacilli, and enterobacteria. [6] About 1% are aerobic gram-negative rods, mostly coliforms. Distal ileal flora represents a mixture of upper intestinal and colonic flora. The concentrations are commonly 10^{6} to 10^{7} organisms/mL.

Mechanisms regulating gut flora involve primarily gastric acidity and propulsive motility. Large numbers of bacteria normally are destroyed by the acid milieu in the stomach, but, if the pH is high because of achlorhydria or antisecretory drugs, higher concentrations of organisms are found in the upper intestine. [7] Normal motility clears both nutrients and organisms from the intestine, but if propulsive activity is impaired, stasis and bacterial overgrowth occur. Within a few hours after complete obstruction, the contents of the proximal bowel become malodorous and feculent because of a marked increase in anaerobic organisms, especially *Bacteroides* and gram-negative species. [8]

In partial obstruction (e.g., ileal strictures from Crohn's disease) or impaired motility (e.g., diabetic autonomic neuropathy or scleroderma), intestinal stasis promotes bacterial overgrowth and malabsorption. Excessive lumenal organisms cause mild mucosal injury, excessive gas formation, catabolism of nutrients with formation of short-chain fatty acids, and protein deprivation. Deconjugation of bile acids by bacteria impairs micelle formation and causes steatorrhea. Vitamin B_{12} deficiency develops from bacterial binding of the vitamin B_{12} intrinsic factor complex.

The most serious consequence of increased lumenal bacteria occurs in strangulated obstruction. The ischemia compromises the integrity of the bowel wall's defense barrier. The mucosa is especially susceptible to anoxia and necrosis, which lead to hemorrhage and increased permeability with transudation of toxic, infected intralumenal fluid across the bowel into the peritoneum, regional nodes, and mesenteric circulation. This exudate is particularly lethal if clostridial bacteria and their exotoxins are present in the fluid. Enterotomy without antibiotic coverage leads to a high rate of postoperative wound infection.

Less is known about changes in bowel flora with ileus, but it is likely that qualitative and quantitative changes occur that are similar to those seen with mechanical obstruction. Bacterial overgrowth occurs, with impaired propulsive motility. [9]

Changes in Bowel Contents

Absorption and secretion of fluid and electrolytes take place in both small and large intestines. The duodenum and jejunum accomplish high-capacity absorption, whereas the ileum and colon provide high efficiency. With intestinal obstruction, fluid and electrolytes accumulate proximal to the obstructive site. Isotopic studies of water and electrolyte flux during the first 12 hours after obstruction demonstrate both reduced net absorption and increased secretion of water, sodium, and potassium. [10] As obstruction is prolonged, failure of absorption and enhanced secretion of water and electrolytes increase still further, and net absorption becomes net secretion. Neural reflexes, activated by stretch receptors, induce secretion. Bacterial overgrowth contributes to the enhanced secretion, partly through the metabolism of ingested nutrients, but the exact mechanism is not known. [11] In contrast, absorption of water and electrolytes continues in the obstructed colon, resulting in the conversion of the liquid ileal effluent to a solid fecal mass. Fluid and electrolyte fluxes with ileus have not been studied satisfactorily but probably are not greatly different from normal.

In addition to increased fluid, intestinal gas adds to the abdominal distention and the gas-filled loops routinely observed on plain abdominal radiographs. The origin of the gas is mainly from swallowed air in both ileus and obstruction. Gas does not accumulate to any degree if a cervical esophagostomy is performed in experimental obstruction, and analysis of the gas composition reveals high concentrations of nitrogen from swallowed air. If oral intake continues, bacterial fermentation produces gases (e.g., carbon dioxide, hydrogen, methane) from ingested nutrients, adding to distention.

Bowel distention contributes significantly to the patient's discomfort. Abdominal pain is much more severe in obstruction than it is in ileus. Distention also may compromise respiration, especially in patients with cardiorespiratory problems, through impairment of diaphragmatic motion. Finally, the lumenal sequestration of fluid and electrolytes, loss of absorptive capacity, and vomiting contribute to dehydration and circulatory insufficiency through fluid loss from the extracellular and intravascular compartments.

Changes in Motility

Marked changes in motor activity occur with obstruction of the small intestine or colon (Fig. 41-2). In experimental obstruction of the intestine, the segment distal to the obstruction is inhibited immediately, but contractions increase proximally. [12] Accumulated fluid, gas, and nutrients raise intralumenal pressure and wall tension, stimulating stretch receptors and enhance the peristaltic reflex. The increased motor activity proximally is prevented by atropine and thus is cholinergically mediated. [13] With more prolonged obstruction, periods of quiescent motor activity lasting 1 minute or longer are interspersed with intense regular 10- to 11-minute pressure waves, some of which occur only at one recording site and others of which migrate aborally. Clustered contraction patterns often are seen in the normal fasting state, but they are not normal after a meal. They produce the intermittent colic and borborygmi that patients with obstruction typically experience after eating. [14] These migrating clustered contractions are typical of partial as well as complete obstruction after a meal, but they are not diagnostic because they occur in other conditions, including pseudoobstruction. [15] If the obstruction is allowed to continue, the quiescent periods become progressively longer, and motor activity throughout the bowel is gradually reduced through activation of inhibitory intestinointestinal reflexes.

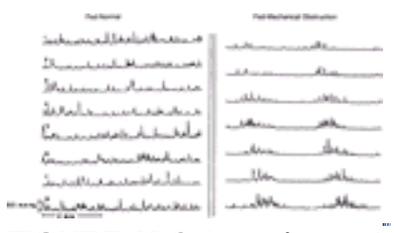

FIGURE 41-2. Irregular contractions normally occur evenly after a meal (**left**). In the obstructed intestine, clusters of contractions occur postprandially at intervals of 1 to 2 minutes, with intervening periods of quiescence (**right**).

In experimental obstruction, almost no contractile activity occurs in the segment of small bowel aborad to the obstruction throughout the period of obstruction. The mechanism of this descending inhibition is not established. It is not caused by an absence of intestinal contents, because contractions do not increase after chyme is infused. [13] It is not reversed by a- or ß-adrenergic antagonists, nor is it overcome by cholinergic agonists. It may be caused by activity of the nonadrenergic, noncholinergic inhibitory (nitric oxide) nerves, but this hypothesis is unproven.

Colonic obstruction also causes changes in motility that occur more slowly as a result of the larger accommodative volume. [16], [17] If the right colon is obstructed, both the segment proximal to the obstruction and the distal colon exhibit reduced contractions. If the left colon is obstructed, however, motor activity increases throughout the large bowel, exhibiting a pattern of clustered contractions similar to that seen in small bowel obstruction.

Mechanisms involved in adynamic ileus may be neurogenic, myogenic, or humoral. They may include either excessive inhibition or deficient excitation. The least understood mechanisms are the reflex mechanisms and muscle failure, whereas most is known about circulating substances because many are easily measurable. Any of the following may reduce or abolish motor activity: blood-borne toxins, drugs, circulating hormones, and abnormalities in mineral balance, acid-base balance, and oxygen supply.

Postoperative ileus has been studied more than some other causes of motor paralysis and may be a prototype for some of the other conditions. Somatic inhibitory reflexes are activated as soon as the parietal peritoneum is entered. After an operation, gastrointestinal motor failure ensues for several days; food and fluids are not tolerated, and no flatus or stool is passed per rectum. Anesthetic agents themselves inhibit motility. [18] Studies of motor activity in animals showed that contractions return first in the small intestine 3 to 6 hours after laparotomy, in the stomach after 24 hours, and in the colon after several days. [19], [20] and [21] Analogous human

studies confirm that the colon is slowest to recover, and activity of the right colon returns several days before that of the sigmoid colon. [22], [23] Because motility patterns do not return entirely to normal as soon as contractions return, normal digestive function is delayed. Postoperative ileus persisting longer than 3 to 4 days suggests that a complication exists. The factors influencing the duration of ileus are varied. Manipulation of the abdominal organs delays return of function longer than does a laparotomy alone. Chemical or physical irritants in the peritoneal cavity, such as bile, blood, or acid, delay recovery even longer. Low-grade sterile peritonitis may stimulate the somatic inhibitory reflexes. Bowel dilation from gas and fluid accumulation activates visceral afferents and an inhibitory reflex arc. [24], [25] Opioid narcotics reduce propulsive motility patterns and augment inhibitory reflexes. If none of these is present, a mechanical obstruction may exist. Abdominal computed tomography and small bowel enteroclysis are the most helpful studies to differentiate between obstruction and motor paresis. [26]

The mediators involved in postoperative ileus are unknown. Adrenergic inhibition has been thought to be the cause, but this concept does not explain why the process lasts several days. Plasma concentrations of catecholamines and norepinephrine turnover are increased temporarily after anesthesia, laparotomy, and intestinal manipulation, [27], [28] and the return of migrating motor complexes is faster if an operative splanchnicectomy is performed. The use of epidural anesthesia, which blocks efferent sympathetic nerves, does not shorten ileus or the time of first passage of gas or feces, however, and the benefits of adrenalectomy, demedullation, or adrenergic antagonists are controversial. [19], [29], [30] These findings suggest that mechanisms other than spinal reflexes play a major role in development and maintenance of postoperative ileus. [31]

Metabolic Consequences and Systemic Effects

One of the first systemic consequences of obstruction or ileus is fluid, electrolyte, and acid-base imbalance. The type of disturbance is largely dependent on the anatomic site of the block. With gastric outlet obstruction, repeated emesis of clear fluid high in hydrochloric acid and potassium chloride leads to metabolic alkalosis with hypokalemia and hypochloremia. With distal duodenal or proximal jejunal obstruction, alkaline biliary and pancreatic secretions are lost, producing metabolic acidosis. The volume of emesis is less with distal intestinal obstruction, but vomiting is less likely to decompress the bowel; therefore, the colicky pain and abdominal distention are more severe. Vomiting and dehydration are uncommon in colonic obstruction, but distention and pain may be intense. If the ileocecal valve is incompetent, colonic obstruction more closely resembles ileal obstruction; if the valve is competent, however, gas accumulates rapidly from both swallowed air and bacterial fermentation. The pressure inside the colon may rise rapidly and cause rupture, most commonly in the cecum. The tensile strength of the cecum is relatively low, and mucosal microcirculation is more tenuous than in the small bowel. By the law of Laplace, tension in the wall is greatest where the radius is the greatest. Emergency decompression of the colon is required if the diameter of the colon rapidly enlarges to greater than 12 cm, because of the risk of perforation.

The same metabolic consequences may occur as a result of ileus, although in most situations ileus is caused by metabolic abnormalities. Vomiting, changes in flora, and alterations in absorption and secretion may produce a clinical situation identical to that seen in simple obstruction. Ischemic complications and sepsis are uncommon, however.

With closed loop obstruction and strangulation, a systemic inflammatory response occurs as a result of the release of a wide range of substances from necrotic bowel. Cyclooxygenase and lipoxygenase metabolites, slow-reacting substance, a variety of kinins, histamine and serotonin, lysosomal enzymes, and free radicals all probably play a role in the fever, leukocytosis, fluid shifts, and shock that may precede or coexist with the developing sepsis. Finally, organ failure and death may ensue with one or more of the following: metabolic encephalopathy, acute renal insufficiency, hepatic failure, high-output shock and myocardial dysfunction, adult respiratory distress syndrome, and disseminated intravascular coagulation.

HISTORY

The clinical findings in ileus and obstruction depend on the anatomic site (Table 41-1). The physician should attempt to determine the anatomic site involved and to ascertain the underlying cause. For management decisions, it is important to know the duration of the process and whether complications such as peritonitis, perforation, sepsis, or strangulation are present. The main symptoms of obstruction are crampy, spasmodic abdominal pain, vomiting, borborygmi, abdominal distention, and obstipation.

TABLE 41-1 Clinical Features of Ileus and Obstruction Dependent on Anatomic Site

Partial obstruction causes intermittent crampy abdominal pain occurring after meals, especially after eating fiber-rich foods. If the patient eats a low-residue diet or avoids eating entirely, symptoms are reduced or avoided. As the obstruction becomes increasingly complete, the pain intensifies. True intestinal colic is dull, crampy, or squeezing, and it occurs in waves of severe pain followed by waning intervals of rest in 1- to 10-minute cycles. Bowel sounds may be so loud that they can be heard by others in the room (i.e., borborygmi). With ileus, pain is much less intense and may be sensed only as a feeling of pressure or fullness. Patients may observe abdominal distention or bloating.

As the obstruction becomes more advanced, nausea, vomiting, and pain are more frequent and more severe. Pain and distention are relieved by vomiting in proximal but not distal obstruction. See Table 41-1 for other site-specific features of obstruction. With complete obstruction and protracted vomiting, dehydration and orthostatic hypotension cause reduced urine output. Patients also develop obstipation and fail to pass flatus. Patients with fecal impaction often have paradoxic diarrhea.

To investigate the underlying causes of obstruction, inquiry should be made about the following: previous operations, which may indicate adhesions; previous episodes of obstruction; bulges on the abdominal wall or in the groin, which may indicate hernias; previous cancers or polyps; abdominal irradiation; inflammatory bowel disease; peptic ulcer; gallstone disease; pancreatitis; foreign body ingestion; and diverticular disease. A psychiatric history (e.g., ingestion of foreign bodies) and a family history of polyps or cancer may provide helpful clues to the underlying diagnosis. Exaggeration of pain during menses suggests endometriosis.

Hypomotility and paralytic ileus most often are caused by the following: metabolic, electrolyte, and acid-base disorders; pharmacological inhibition of motility; or primary or secondary neuropathies or myopathies. Careful family, medication and illicit drug, endocrine, and immunologic histories are important in discovering the underlying cause. The physician should be alert to thyroid and parathyroid disorders, hypokalemia, diabetes mellitus, scleroderma, heavy-metal poisoning, and porphyria.

The development of constant unremitting localized pain, fever, chills, rigors, and a general sudden worsening of the clinical state all suggest ischemia and infarction. Waiting for such signs to develop before acting is poor clinical judgment, however. Strangulation and necrotic bowel may exist with none of these features being present. A high index of suspicion and aggressive management are important practices to adopt if mortality is to be kept low. If the clinical condition changes and any of these systemic symptoms appear, urgent steps must be taken to search for and treat potentially ischemic or necrotic bowel.

PHYSICAL EXAMINATION

In mechanical obstruction, the patient may be doubled over, holding the abdomen and writhing restlessly in bed, frequently changing position, and vomiting or retching. Intervals of relief may occur during which the patient may lie still. With ileus, the pain is less severe in the face of moderate abdominal distention, but it is more steady and unrelenting, and the patient usually lies quietly in bed.

Some physical findings may provide clues to the underlying cause of the obstruction or ileus (Table 41-2).

PHYSICAL SIGNS	DIAGNOSIS
Abdominal scars	Adhesions
Pyoderma gangrenosum, erythema nodosum	Ulcerative colitis, Crohn's disease
Buccal, palmar, plantar pigmentation	Peutz-Jeghers syndrome
Cullen, Grey, Turner sign	Hemorrhagic pancreatitis
Cutaneous atrophy, hyperpigmentation	Chronic pancreatitis, abdominal radiation
Vesicles, bullae, scars, pigmentation	Porphyria
Neurofibromas	Von Recklinghausen disease
Acanthosis nigricans	Gastrointestinal malignant diseases
Butterfly dermatitis	Lupus erythematosus
Atrophy, telangiectasias, variable pigmentation	Scleroderma, dermatomyositis

TABLE 41-2 Cutaneous Findings That Are Clues to the Cause of Obstruction

Carefully palpate the umbilicus, the lower trunk, and both inguinal and femoral areas in every patient with suspected obstruction, to detect external hernias. Incarcerated femoral hernias are especially easy to overlook in the obese patient. Hernias sometimes can be manually reduced, relieving the obstruction and avoiding the hazards of strangulation. Careful palpation is also imperative to detect hepatosplenomegaly, liver masses, or other intra-abdominal masses that suggest malignant neoplasms. Tender lumps may suggest an abscess from Crohn's disease or diverticulitis, an intussusception, or an ischemic incarcerated loop of bowel. Localized tenderness occurs at the lateral margin of the rectus abdominis with a lateral ventral or spigelian hernia. Metastatic nodes in the umbilicus (Sister Mary Joseph nodule) indicate peritoneal carcinomatosis, whereas nodes in the inguinal region may occur with colorectal or other neoplasms. A careful rectal and pelvic examination always should be done to find rectal or vaginal tenderness, as well as masses and fecal or barium impaction. For example, obturator and sciatic hernias are discovered only by rectal examination, and a Blumer shelf indicates metastatic cancer.

Resonant percussion or tympany occurs in both ileus and obstruction because of entrapped intestinal or colonic gas. Shifting dullness or a puddle sign occurs if there is free abdominal fluid and may suggest associated malignant or inflammatory ascites, as in complicated pancreatitis, bowel necrosis, or early tuberculous peritonitis.

Auscultation is important in differentiating obstruction from ileus. With ileus, the abdomen is almost completely silent. Rare low-pitched gurgles or weak tinkles are heard, along with transmitted heart tones and moving water if the patient changes position. In obstruction, bowel sounds become louder, higher pitched, and hyperactive unless the obstruction is prolonged for several days or is complicated by ischemia, necrosis, or peritonitis. These sounds have a musical, tinkling, or metallic quality and occur in clusters or rushes, coinciding with colic. The examiner may need to listen for 10 to 15 minutes to hear abnormal sounds. In recent-onset proximal obstructions, these sounds may occur every 3 to 5 minutes or up to 15 minutes if obstruction is in the distal bowel or is more prolonged or complicated by strangulation.

Repeated examinations are essential because complications may develop with time and may produce changing physical findings. If bowel sounds disappear, or if signs of peritoneal irritation, such as low-pitched rubs, muscle guarding, rigidity, or rebound tenderness, intervene, the bowel is probably ischemic, and rapid intervention is imperative. Patients with fevers, hypotension, rigors, or signs of sepsis develop ominous findings that often signal a life-threatening complication.

DIFFERENTIAL DIAGNOSIS AND DIAGNOSTIC STRATEGIES

Causes of Mechanical Obstruction

The causes of mechanical obstruction are varied and may be divided into extrinsic lesions, intrinsic lesions, and intralumenal objects (Table 41-3). In adults, the most common causes of obstruction are adhesions and hernias in the small bowel and cancer in the colon. It may not be possible to diagnose the cause of the obstruction preoperatively, but a careful clinical examination and appropriate studies usually yield a diagnosis and aid in management decisions.

Extrinsic Lesions
Adhesions and congenital bands
Hernias
External hernias
Internal hernias
Diaphragmatic hernias
Pelvic hernias
Volvulus
Gastric
Midgut
Cecal
Sigmoid
Extrinsic masses
Benign or malignant tumors
Abscesses
Aneurysms
Hematomas
Endometriosis
Intrinsic Lesions
Benign and malignant neoplasms
Adenocarcinomas
Lymphomas, lymphosarcomas
Carcinoid tumors
Inflammatory conditions
Tuberculous enteritis, Crohn's disease
Strictures secondary to IKDI, nonsteroidal antiinflammatory drugs, and ischemia
Intrinsic Lesions (continued)
Radiation injury, caustic ingestants
Eosinophilic gastroenteritis, ameboma
Diverticulitis, pelvic inflammatory disease
Intussusception
Congenital defects
Hypertrophic pyloric stenosis, annular pancreas
Intestinal atresia/agenesis
Malrotation/volvulus
Intestinal duplication, mesenteric cysts
Meckel diverticulum
Hirschsprung disease
Hematoma
Abdominal trauma
Thrombocytopenia
Henoch-Schönlein purpura
Intralumenal Objects
Meconium ileus
Barium impaction
Fecal impaction
Gallstone ileus
Gastric bezoars
Foreign bodies

TABLE 41-3 Causes of Mechanical Obstruction

Extrinsic Lesions Extrinsic masses can compress the bowel or mesentery and cause obstruction. Adhesions are the most common cause of small intestine obstruction in adults, but they rarely obstruct the colon. They most commonly occur after gynecologic procedures and operations on the small and large intestine and uncommonly occur after gastric and biliary surgical procedures. Adhesions may occur from a few days to 10 or 20 years after an operation. They may occur without a history of an operation, usually after infection (e.g., peritonitis) or irradiation. Adhesive bands form and contract with time, entrapping a loop of bowel. They often cause a closed loop obstruction, commonly associated with strangulation. Congenital bands may occur in association with malrotation (i.e., Ladd bands) or in the absence of any known cause.

Hernias. Hernias may cause either simple obstruction or closed loop obstruction; strangulation is common in incarcerated hernias (i.e., those that cannot be reduced) because the blood supply is compromised by the hernial ring. External hernias protrude through the abdominal wall, are palpable as tender masses, and are often reducible. Internal hernias are not palpable and usually are identified at the time of operation. Diaphragmatic hernias rarely cause obstruction unless they are paraesophageal. Uncommon pelvic hernias may be palpable but require careful pelvic, rectal, and perineal examination and are most often recognized at operation as the cause of bowel obstruction. Chapter 121 includes a complete discussion of hernias.

Volvulus. A *volvulus* is an abnormal torsion of a segment of bowel, usually colon, producing a closed loop obstruction and occlusion of the blood supply. It involves the sigmoid colon in 70% to 80% of cases and the cecum in 10% to 20% of cases. Typically, the onset of pain is sudden, severe, and followed rapidly by severe abdominal distention. A tender mass may be palpated, and characteristic abdominal plain radiographic findings are often present (Fig. 41-3). Reduction is possible by barium enema or colonoscopy, but the volvulus is likely to recur. Volvulus of the small intestine occurs in newborns but is rare in adults. Volvulus of the stomach often is associated with large defects in the diaphragm, congenital malrotation, or large paraesophageal hernias.

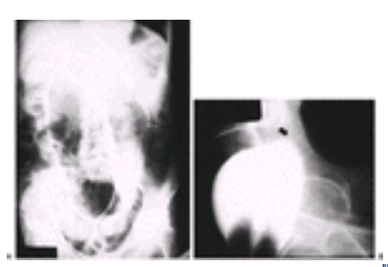

FIGURE 41-3. (**A**) Plain abdominal radiograph shows colonic obstruction secondary to sigmoid volvulus. (**B**) Barium enema reveals a tapered cutoff at the site of the

volvulus (*arrow*).

Intrinsic Lesions

Tumors. Tumors can narrow or obstruct the lumen, or they may be the leading point of an intussusception. Neoplasms causing obstruction may be benign or malignant. If malignant, they may be primary or metastatic, although metastatic tumors usually tether and fix the bowel rather than involve the lumen. Although uncommon, primary malignant tumors of the small bowel are most often carcinoids, lymphomas, or adenocarcinomas. Adenocarcinoma is the most common cause of colonic obstruction.

Inflammatory or ischemic processes. Inflammatory or ischemic processes involving the bowel wall produce strictures, lumenal narrowing, muscle dysfunction, and impaired transit. Blunt trauma to the abdomen can produce intramural hemorrhage and compromise of the lumen. Hematomas also may occur as a result of severe thrombocytopenia or clotting disorders with or without the vascular fragility seen, for example, in Henoch-Schönlein purpura.

Intussusception. Intussusception exists when a leading segment of bowel invaginates into an accepting segment. In older children and adults, an intrinsic bowel lesion, such as a tumor or Meckel diverticulum, usually initiates the process. The inner, advancing invaginated segment is called the *intussuscipiens,* and the outer, accepting invaginating segment is termed the *intussusceptum.* Initially, the inner walls become edematous because of lymphatic obstruction. As the process advances, venous obstruction, infarction, and necrosis follow.

Intralumenal Objects Barium from an upper gastrointestinal series can cause a barium impaction in the colon if it is not evacuated promptly. Excessive extraction of water from the barium occurs if there is prolonged colonic stasis. This is often caused by an underlying motility disorder, such as scleroderma, chronic intestinal obstruction, or colonic pseudoobstruction. [32] Obstructing fecal impaction may result from severe chronic constipation, a variety of drugs, such as narcotics or antipsychotics, an underlying colonic carcinoma, or diverticulitis. Large gallstones occasionally erode through the gallbladder wall into the duodenum or, more rarely, into the colon or stomach. Some of these stones are large enough to obstruct the bowel, most commonly the distal ileum. This condition is misnamed gallstone ileus and may be suspected if dilated loops of bowel proximal to a small bowel obstruction are associated with air in the biliary tree from the cholecystoduodenal fistula. Finally, objects from the stomach may produce intestinal obstruction if they pass into the intestine, such as gastric bezoars, ingested foreign bodies, and iatrogenically introduced bodies.

Causes of Adynamic Ileus

Ileus is never primary, and a search for the etiologic disorder is essential to achieve success in management. Because the clinical picture in ileus can be so similar to that in mechanical obstruction, diagnostic considerations for both disorders must proceed concurrently. Long-standing or complicated mechanical obstruction may even terminate with ileus. The underlying causes of mechanical obstruction are overt, simple, anatomic, and almost always conclusively apparent. Conversely, the underlying mechanisms in acute motor paralysis are occult, complex, and rarely completely understood. Causes of adynamic ileus are outlined in Table 41-4. The conditions associated with ileus often can be defined, but a great deal is yet to be learned before the mechanisms are understood and rational therapy can be applied. Conversely, treatment usually is successful through management of the underlying cause.

Intra-abdominal Causes	Extra-abdominal Causes
Reflex inhibition	Reflex inhibition
Laparotomy	Craniotomy
Abdominal trauma	Rib, spine, or pelvic fractures
Renal transplantation	Myocardial infarction
Inflammatory conditions	Coronary bypass
Perforated viscus or penetrating wounds	Open heart surgery
Bile peritonitis	Pneumonia, pulmonary embolus
Chemical peritonitis	Burns
Intraperitoneal hemorrhage	Black widow spider bites
Toxic megacolon	Drug-induced
Familial Mediterranean fever	Anticholinergic/ganglionic antagonists
Acute pancreatitis	Opiates
Acute cholecystitis	Chemotherapeutic agents
Celiac disease	Tricyclic antidepressants
Inflammatory bowel disease	Phenothiazines
Acute irradiation injury	Metabolic abnormalities
Abdominal irradiation	Septicemia
Infectious processes	Electrolyte imbalance
Bacterial peritonitis	Heavy metal poisoning (lead, mercury)
Appendicitis	Porphyria
Diverticulitis	Uremia
Herpes zoster virus	Diabetic ketoacidosis
Anorectal herpes simplex virus	Sickle cell disease
Ischemic processes	Pulmonary failure
Arterial insufficiency	
Venous thrombosis	
Mesenteric arteritis	
Strangulation obstruction	
Retroperitoneal processes	
Ureteropelvic stones	
Pyelonephritis	
Retroperitoneal hemorrhage	
Pheochromocytoma	
Malignancy (Ogilvie syndrome)	

Note: Adapted from ref. 34.

TABLE 41-4 Causes of Adynamic Ileus and Acute Colonic Pseudo-obstruction

Chronic intestinal pseudoobstruction clinically mimics mechanical obstruction but is not caused by an anatomic blockage. It may result from primary neuropathic or myopathic disorders, which are often familial. It also may develop secondary to a variety of other disorders (see Chapter 73).

The term *acute colonic pseudoobstruction* implies that nonobstructive massive dilation of the colon is temporary and reversible. The condition is commonly called Ogilvie syndrome, although this term is technically incorrect. Ogilvie described chronic dilation in association with invasion and destruction of the celiac axis and semilunar ganglion by retroperitoneal malignant disease. [33] Its underlying causes are multiple and essentially the same as those compiled for ileus (see Table 41-4). [34] Numerous mechanisms must be operative for such a range of associated conditions. Impaired motility or aerophagia each plays an important role, but which comes first and which dominates are uncertain. It is likely that the massive dilation initiates autoinhibitory reflexes that perpetuate the problem.

Toxic megacolon occurs in severe inflammatory bowel disease and in other forms of colitis, including bacillary or amebic dysentery. The exact pathogenesis remains an enigma, but it is likely that transmural inflammation adversely affects the neuromuscular apparatus. Inhibitory drugs, such as anticholinergics, opiates, and antidiarrheals, probably have an important deleterious effect and must be avoided or discontinued.

ASSESSMENT

Biochemical and Hematologic Tests

Laboratory studies have limited usefulness in the diagnosis of mechanical obstruction, but they are critical in its management. Early in the course of obstruction, all blood tests are usually normal. Except for laboratory evidence of infection, inflammation, or tumors, biochemical and hematologic tests only rarely aid in establishing the cause of a mechanical obstruction. In contrast, because of the metabolic abnormalities that commonly produce or are associated with ileus, the laboratory is essential in discovering the underlying cause of ileus and in its management. In milder conditions, measurement of serum electrolytes, blood urea nitrogen, and creatinine is helpful in assessing fluid balance and the presence and severity of dehydration. As vomiting and dehydration persist, hemoconcentration increases, and hemoglobin, hematocrit, and serum albumin levels rise. Leukocytosis is common in infectious and inflammatory disorders. In more severe cases, measurement of arterial blood gases is also necessary to assess acid-base balance. More proximal obstructions cause greater acid-base imbalance, whereas the more distal obstructions cause greater electrolyte disorders. Abnormalities of serum calcium, magnesium, and phosphate may contribute to ileus and, if abnormal, should be corrected.

If the bowel is ischemic or infarcted, a variety of enzymes may leak across the intestine and be released into the circulation. Pancreatic and hepatic enzyme elevations may be encountered, but no laboratory test is a reliable indicator of infarction. In addition, enzyme levels may be elevated because of concurrent primary pancreatic or hepatic disease. Sepsis also may contribute to these abnormalities as well as to elevations in bilirubin and in serum and urine phosphate. Most of the abnormalities manifest late in the course of the disease. [35], [36], [37] and [38] In summary, even though abnormalities in laboratory tests may occur because of intestinal infarction, they are neither sensitive nor specific in this setting. Analysis of ascites can be very helpful in establishing diagnosis or in discovering complications. Analyses should include cell counts, albumin (subtract ascites albumin from simultaneous serum albumin to obtain serum/ascites albumin gradient), total protein, glucose (with simultaneous blood glucose) lactate, amylase, and cultures in blood culture media.

Radiologic Studies

Plain Films If the clinical evaluation suggests either obstruction or ileus, radiographic examination is extremely helpful to confirm the diagnosis, to differentiate ileus

from obstruction, to localize the level of an obstruction, and to contribute to an understanding of the underlying cause. [39], [40] The first studies to be selected include posteroanterior and lateral chest radiographs and upright and supine films of the abdomen without contrast. One study found that a supine abdominal film and an upright frontal chest radiograph identified pertinent abnormalities in 98% of cases and were sufficient for screening. [41] The chest radiograph is important to detect pneumonia or other extra-abdominal processes as a cause of ileus, to evaluate cardiorespiratory status preoperatively, and to discover free air or other subdiaphragmatic abnormalities. If an upright chest film is not physically or technically possible, a cross-table lateral film of the abdomen with the left side down may demonstrate free peritoneal air. It is important to wait 5 to 10 minutes after the patient assumes this position to allow migration of small amounts of air to the paracolic gutter. The abdominal radiographs demonstrate the amount and distribution of gas and fluid in the gastrointestinal tract. The jejunum lies in the left upper and central abdomen, the ileum lies in the right central and lower abdomen, and the colon occupies the flanks and right iliac fossa. Some radiologists recommend a prone abdominal view instead of or in addition to a supine view, because it allows intestinal gas to fill the left colon and rectum, improving the assessment of colonic obstruction. [42] Normally, there is almost no air in the small intestine and only scattered gas bubbles and feces in the colon. In early or incomplete small bowel obstruction, gas and fluid begin to accumulate, causing the lumen to become dilated. Air, fluid, and feces persist in the colon, and without contrast it is difficult to determine the level of obstruction or even to differentiate obstruction from localized ileus. With total lumenal blockage, more gas and fluid accumulate proximal to the level of the obstruction, and the lumen becomes widely dilated (often >3.5 cm in the jejunum). The valvulae conniventes of the small intestine produce markings across the entire lumenal diameter (Fig. 41-4). In the upright or decubitus position, multiple air-fluid levels occur with a stepladder pattern (i.e., different levels in adjacent loops). Distal to a complete obstruction, the bowel, including the colon, empties and collapses within 12 to 24 hours. If air persists in the colon, its diameter is less than that of the small bowel, which is evidence of a partial obstruction.

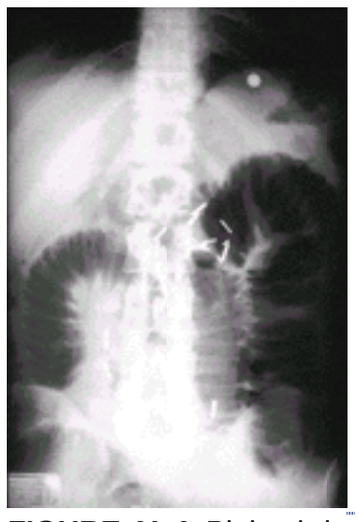

FIGURE 41-4. Plain abdominal radiograph shows multiple dilated loops of intestine indicating small bowel obstruction caused by adhesions from previous surgery. No gas is present in the colon.

With colonic obstruction, most or all of the air and fluid accumulate in the colon proximal to the obstruction if the ileocecal valve is competent. The haustra cause incomplete indentations in the contour of the wall, producing a scalloped effect. Distal to the obstruction, the colon and rectum become free of gas and feces. If the ileocecal valve is incompetent or absent, gas and fluid are seen throughout both the proximal colon and the small bowel. Except in proximal colonic obstruction, the diameter of the transverse colon is greater than that of the small bowel. Early in the course of strangulation, no features clearly distinguish the radiologic findings from those of simple obstruction. If strangulation is far advanced, the necrotic bowel loses its mucosal contour and becomes splayed by edema, exhibiting a thumbprinted appearance. Air may be seen in the bowel wall (i.e., pneumatosis), in branches of the portal venous system, or free within the peritoneal cavity, signifying perforation. In ileus, gas and fluid accumulate differently throughout the gastrointestinal tract (Fig. 41-5). Loops of mildly distended bowel may develop proximal or adjacent to the site of an acute inflammatory process such as appendicitis or pancreatitis. These loops are involved by localized ileus and are called *sentinel loops.* If peritonitis coexists with ileus, the bowel wall becomes thickened, and the properitoneal fat line becomes obscured. If ascites is present, it produces a hazy, ground-glass density throughout the abdomen. In acute colonic pseudoobstruction, the entire colon becomes dilated, but the cecal diameter is usually the greatest.

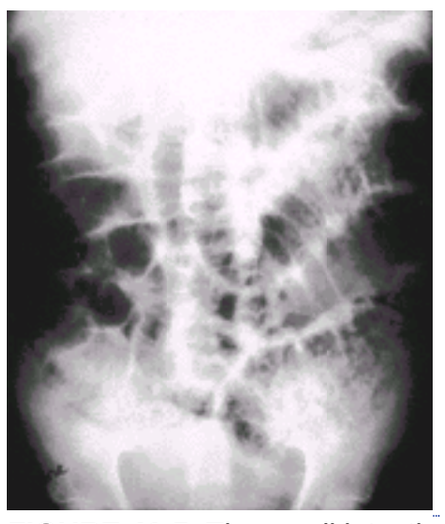

FIGURE 41-5. The small intestine is dilated and distended with gas; a small amount of gas is also present in the colon. These findings suggest ileus, caused in this case by jejunal ischemic necrosis.

In the differentiation of ileus from obstruction, the degree of intestinal distention, the amount of intralumenal fluid and gas, and the distribution pattern of air-fluid levels in the upright or decubitus position are important features to compare. Obstruction usually causes more fluid and gas accumulation than ileus, and air-fluid levels tend to be longer and more pronounced. Intestinal gas accumulates in both conditions, but in ileus more gas is in the colon than the small bowel. The pattern is reversed in small bowel obstruction. A stepladder appearance can occur in either obstruction or ileus. If multiple air-fluid levels appear in a string-of-beads pattern, however, a high-grade partial or complete obstruction of the small bowel is likely. This obstruction is caused by small air bubbles trapped in the superior recesses of splayed-out valvulae conniventes in an upright or decubitus position while the dilated loop is filled with large amounts of fluid. Supine films, conversely, may show little or no gas in the small intestine. If the diagnosis is inconclusive, sequential films often help to define it.

Contrast Studies The differentiation of ileus from mechanical obstruction is not always possible with plain abdominal films, and the presence or absence of obstruction can be determined only by using contrast media. Barium provides better contrast and detail than water-soluble media and therefore is usually the agent of choice. If there is a question of bowel viability or perforation, however, barium should be avoided because it would create an intense inflammatory reaction if leaked into the peritoneal cavity. Contrast studies can usually differentiate complete obstruction from partial obstruction and from paralytic ileus and can aid in the selection of therapy. Small bowel enteroclysis should be the method of choice to detect early or subtle structural disorders and most accurately to document normal status. Some authors recommend abandoning the conventional small bowel follow-through method. [43] When enteroclysis is not available, the follow-through can be improved by adding the peroral pneumocolon or gas-enhanced double-contrast study. One study used a 6-hour radiograph after oral Gastrografin (diatrizoate and iodine) to provide information in clinically equivocal intestinal obstruction. When the contrast agent was in the colon in 6 hours, almost 90% of the patients could be managed nonoperatively. [44] If there is any doubt about the site of obstruction, barium should not be given orally until colonic obstruction is excluded by barium enema or colonoscopy. When orally administered barium accumulates proximal to a colonic obstruction, water continues to be extracted from colonic contents, and the barium becomes inspissated and impacted. It can be removed only at the time of operation, which is hazardous because of potential spillage. It is acceptable to give small amounts of barium orally or by intubation in small bowel obstruction after colonic obstruction is excluded. Net secretion in the small bowel keeps the barium in a liquid suspension, and if necessary it can be removed through an intralumenal tube. In small bowel obstruction, barium provides superior definition to water-soluble media, which are hyperosmolar and become diluted by secretion and mixing with intestinal contents, thus reducing contrast and definition. Barium also should be used for retrograde studies unless there is a possibility of perforation. A nonionic contrast agent, such as Amipaque (metrizamide), is isotonic with blood. Therefore, it is the contrast agent of choice whenever a water-soluble agent is needed; however, high cost usually limits its clinical application to neonates and young children.

Other Imaging Procedures Angiography is sometimes useful in diagnosing abdominal tumors, but other modalities have generally replaced this method. It is still useful in suspected mesenteric ischemia and infarction. Ultrasonography has a limited role in both obstruction and ileus because the gas-blocking from dilated loops of bowel prevents imaging beyond the tissue-gas interface. Computed tomography may be very helpful in establishing the diagnosis and in locating the obstruction

anatomically if other imaging studies are inconclusive. Establishing the diagnosis with certainty preoperatively is not always necessary or even desirable if a needed operation is delayed by waiting for the test.

Other Studies

The role of esophagogastroduodenoscopy in ileus and obstruction is limited, as is enteroscopy. Benign ulcers, strictures, and carcinomas are the most commonly encountered lesions. However, the placement of stents can be useful in palliation of pancreatic cancer that partially obstructs the duodenum. Similarly, colonoscopy can be of value in the diagnosis and management of sigmoid volvulus, obstructing cancers, and inflammatory strictures (see section “ Nonoperative Therapy”).

Endoscopic procedures play a limited role in diagnosis of small and large intestinal obstruction but may be more important in therapy. Brushings and biopsies can provide definitive histological diagnosis to plan treatment. However, endoscopy is increasingly used to decompress and devolvulize the stomach and colon or to relieve obstruction through the use of expandable metallic stents.

An abdominal paracentesis should be done if fluid is present and the cause of obstruction or ileus is not clear. The presence of blood, bile, amylase, proteins, leukocytes, bacteria, or malignant cells is helpful in making therapeutic decisions.

THERAPEUTIC CONSIDERATIONS

Therapeutic Strategy

Decision making in acute obstruction requires repetitive, careful patient observations, well-informed clinical judgment, and close communication with a skilled surgeon. After the initial evaluation and establishment of the presence of obstruction or ileus, fluid, electrolyte, and acid-base resuscitation should begin. Adjustments in the composition and rate of administration are made as soon as the relevant laboratory tests become available. Decompression of the distended bowel is instituted at the same time by nasogastric aspiration. The next decision to be made is whether to use urgent operative therapy, expectant operative therapy, or nonoperative therapy.

Acute complete bowel obstruction is a surgical emergency. The objective of treatment is to relieve the mechanical impediment to propulsion, and operative therapy is most often the means to that end. Ileus is managed by treating the underlying cause, and operative therapy should be avoided entirely unless some other intra-abdominal catastrophe requires laparotomy.

Replacement of Fluids

Correction of fluid, electrolyte, and acid-base imbalances must be guided by the level and duration of obstruction and measurements of the hematocrit; of serum sodium, potassium, chloride, bicarbonate (HCO_3^-), blood urea nitrogen, and creatinine; and of arterial blood gases and pH. Bedside estimates of dehydration are inaccurate, although orthostatic hypotension and tachycardia support the need to begin therapy. If severe hypovolemia exists, restoration of intravascular fluid volume should be monitored by measurements of urinary output, central venous pressure, and arterial blood pressure. If heart failure, chronic obstructive pulmonary disease, or renal failure is a concurrent problem, a Swan-Ganz catheter should be used to monitor the function of the left ventricle.

Fluid replacement should be based on estimates of previous deficits, daily maintenance requirements, and current losses. Mild to moderately severe dehydration may vary from 4% to 8% of total body weight. Half of this deficit should be given during the first 24 hours and half during the second 24 hours. Maintenance requirements average 1500 to 2000 mL in an afebrile 70-kg person with normal renal function. Continued losses through vomiting or nasogastric suction can be measured to allow calculation of total fluid replacement and caloric loss. A balanced salt solution, such as lactated Ringer solution, should be used to correct the deficit.

In patients with metabolic alkalosis from gastric outlet obstruction, fluid replacement should begin with isotonic sodium chloride. As soon as adequate urine output is assured, potassium chloride should be given because renal potassium loss is great in gastric outlet obstruction. The potassium loss can be measured in a 24-hour urine sample. Hydrochloric acid can be given intravenously, but it is rarely needed. In metabolic acidosis, 2 mmol/L $NaHCO_3$ or one sixth molar sodium lactate should be given. Large amounts of sodium HCO_3^- ($Na\,HCO_3^-$) given as ampules of 44.6 mmol/L may produce intracellular volume depletion. If correction of acidosis with $NaHCO_3$ is too rapid, cerebrospinal fluid pH may fall rapidly and worsen neurological symptoms. Overcorrection of arterial pH above 7.4 shifts the hemoglobin dissociation curve to the left and reduces delivery of oxygen to the tissue by increasing the affinity of hemoglobin for oxygen. Slower restoration of pH to 7.4 may be desirable; if there is no liver dysfunction, sodium lactate may be preferred over $NaHCO_3$. Net HCO_3 deficit resulting from gastrointestinal loss may be roughly calculated from plasma HCO_3 concentration (plasma [HCO_3^-]) as follows: 24 mEq/L - plasma HCO_3^- × 0.6 body weight (kg). $NaHCO_3$ should be given if the arterial pH is less than 7.1. One half of the calculated deficit should be given to raise the plasma HCO_3^- to 16 mEq/L after 12 to 24 hours. If nothing is allowed by mouth for several days, some provision must be made for nutrition.

Decompression of the Bowel

There is value in reducing gaseous distention. It is distressing for the patient, causing pain and respiratory embarrassment. Distention induces secretion, causes nausea and vomiting, and may result in aspiration, particularly during induction of anesthesia. Nasogastric suction is appropriate in both obstruction and ileus, but the use of long indwelling intestinal tubes, such as the Cantor tube or Miller-Abbott tube, is controversial and is not recommended routinely. [45] The long tubes are difficult and sometimes impossible to pass without endoscopic guidance. [46] The balloons may expand to dangerous volumes during passage, may predispose to intussusception, or produce intestinal obstruction themselves, and they are probably no more effective in reducing distention than a nasogastric tube. They may be tried if strangulation is unlikely, if nasogastric suction is ineffective, or if long-term nonoperative therapy is planned. Experience is accumulating with temporary endoscopic decompression of colonic obstruction. A flatus tube can be passed over an endoscopically directed guidewire through a distal pinhole lumen. [47] Alternatively, the obstruction can be relieved using a through-the-scope balloon-dilating catheter or a laser or an expandable metallic stent. [48, 49]

Principles of Surgical Treatment

As a general rule, acute complete mechanical obstruction of the small intestine should be relieved as soon as preoperative resuscitation is adequate and nasogastric decompression is established. In the presence of complete obstruction, strangulation cannot be excluded by clinical criteria, and delay in operation may be fatal. [50] These reasons led to a valid clinical dictum, “don’t let the sun set on a small bowel obstruction.” The reason for urgency is mostly that the mortality associated with ischemic bowel complicating obstruction is high, and this problem can be detected with certainty only at operation. An operation provides the ultimate procedure to detect strangulation and to accomplish definitive treatment. Antibiotics almost always are given prophylactically because an enterotomy in an unprepared bowel has a high incidence of wound infection, and sepsis is a frequent complication whenever blood flow is compromised. Coverage must be provided for anaerobes and gram-negative organisms. Laparoscopic management of small bowel obstruction is possible in about 60% of patients. Its use allows quicker recovery, early discharge, and cost savings. [51]

When strangulation is encountered from any cause, the grossly necrotic bowel is resected, and the viability of the adjacent bowel must be assessed. This is difficult if not impossible to do on the basis of gross observation of the tissue. Two important new intraoperative aids to improve accuracy are Doppler ultrasonography and fluorescein dye injection. After restoration of mesenteric circulation, a handheld Doppler probe sends and receives sound waves to assess blood flow. [52] The technique is easy to perform and determines viability accurately, as confirmed by a second-look operation. [53] Intravenous fluorescein can be quantitatively detected in the bowel through the use of a perfusion fluorometer. This method is also highly accurate and useful in determining viability, especially in longer segments. [54, 55] Nonperfused segments are resected, followed by an end-to-end anastomosis. Primary neoplasms of the small intestine are rare but require adequate resection with lymph node dissection if the lesion is malignant. Metastatic tumors are better bypassed than resected.

In partial small bowel obstruction, immediate operative therapy is not usually necessary, and antibiotics are of no proven benefit. There is time to establish the severity and underlying cause with relevant diagnostic tests. If the patient continues to pass stool and flatus and if air persists in the colon, strangulation is unlikely, and expectant therapy is usually appropriate. If any worrisome signs should arise, such as fever, rebound tenderness, leukocytosis, or unexplained hyperamylasemia, laparotomy is indicated. If an external hernia can be found, gentle attempts should be made to reduce it, with avoidance of excessive force and watching for evidence

of infarction or perforation after successful reduction.

Immediate operation is also usually not indicated if the patient has a history of multiple previous episodes of bowel obstruction, multiple abdominal operations with extensive adhesions, extensive abdominal radiation therapy, bacterial peritonitis, Crohn's disease, or carcinomatosis with widespread metastases. In these situations, strangulation is less likely the result of fixation of the bowel. Operative procedures in this setting are exceedingly difficult because of dense adhesions that increase the risk of inadvertent enterotomies, with possible wound contamination, infection, and postoperative intestinal fistulae. In patients who have been treated surgically for abdominal malignant disease, obstruction should be considered to be a result of adhesions and should be managed operatively. If metastatic lesions are known to exist, obstructive episodes usually are managed nonoperatively. [56] If nonoperative therapy is chosen and decompression does not produce marked clinical improvement and resolution of bowel distention in 24 to 48 hours, successful nonoperative therapy is unlikely, and surgical intervention is indicated. [45], [57] If distention resolves, the patient may be managed by follow-up; if signs of bowel infarction appear at any time, however, abdominal exploration is urgently indicated. The value of immediate operation compared with tube decompression and close observation for 48 hours in the treatment of small bowel obstruction remains controversial. The mortality in patients with ischemic bowel is about 30%, and the high risks of this potential complication combined with the difficulty in prompt detection argue for early operative therapy in most settings.

Colonic obstruction usually develops over several days, but it almost always requires surgery. The principles of correcting dehydration and electrolyte balance, decompressing the bowel, and administering antibiotics before colonic surgery always apply. Nasogastric suction may or may not be effective in reducing the distention. Resuscitation reduces operative risk, and emergency surgery is not necessary unless there is peritonitis, perforation, or respiratory embarrassment from excessive distention.

The two most common obstructing colonic lesions are cancer and diverticulitis. The two may be difficult or impossible to differentiate on clinical or radiologic grounds, and they may coexist. If there is any doubt about the diagnosis of diverticular disease after radiologic or endoscopic studies, a resection should be done. Sometimes the cancer is apparent only after careful histological examination. If the problem presents as an acute obstruction, proper bowel cleansing is often not possible. A two-stage operation consisting of resection and diversion, followed by reanastomosis at a later date, has been the standard practice because primary anastomoses had a high rate of anastomotic leaks, wound contamination, and abscess formation. [58] However, resection and primary anastomosis are increasingly used unless the patient is severely ill or has peritonitis from perforation or infarction. Postoperative infections are reduced by intraoperative irrigation of the bowel. [59], [60] and [61] The management problems with diverticular disease are similar to those with cancer. If perforation and abscess are present, the diseased bowel often is resected, and the proximal segment is temporarily diverted. [62]

Nonoperative Therapy

Nonoperative therapy may be helpful in certain benign and malignant disease processes. Endoscopy can be useful not only for the diagnosis of ileus or obstruction but also for definitive therapy. Graded pneumatic balloon catheters can accomplish brusque dilation of obstructing lesions. This approach has been especially helpful in Crohn's disease. [63], [64] and [65] Radial cuts can be made in narrow, short strictures and membranes. Frequently, volvulus of the sigmoid colon and, less often, of the cecum can be reduced by aspirating the gas with a colonoscopically placed tube. Another technique that may be tried in sequence is to exert gentle hydrostatic pressure with a barium enema or other rectally introduced contrast medium. The risk of perforation or necrosis cannot be ignored when either of these procedures is used. If these procedures are successful, frequent examinations should continue for several days to watch for signs of infarction. Recurrence with both cecal and sigmoid volvulus usually requires elective repair. Resection rather than fixation is indicated if there is evidence of ischemia, and some surgeons recommend it in all cases. [66] Adhesions or radiation-induced strictures are uncommon causes of obstruction in the colon. Endoscopic treatment with hydrostatic or pneumatic balloons sometimes can relieve the obstruction, thus obviating the need for emergency surgery in some cases, although recurrences demand definitive surgical therapy. [67]

Snares, heater probes, bipolar cautery devices, and lasers can be used to heat and remove or coagulate obstructing tumors. The necrotic tissue sloughs and provides a larger opening in the lumen. Relief of obstruction resulting from colon cancer is increasingly achieved with self-expandable stent endoprostheses. A stent may be inserted under direct colonoscopic observation and fluoroscopic control as an adjunct to operative preparation in patients with obstructing cancers. Temporary relief of obstruction permits preoperative mechanical and antibiotic preparation, allowing for a one-stage procedure instead of a temporary colostomy. Such an approach is increasingly successful and can provide significant cost savings by avoiding a second surgical procedure. Unfortunately, most patients presenting with obstruction already have advanced disease, but stents also may provide long-term palliation. The procedure may be complicated by perforation or stent displacement. Stent placement also is being investigated for management of neoileal stenosis resulting from recurrent Crohn's disease after an end-to-end anastomosis. [68], [69] Palliative relief of obstruction sometimes can be accomplished in inoperable colon cancer through the use of laser coagulation. [49], [70], [71] However, the yttrium-aluminum-garnet laser, balloon dilation, and chemical sclerosis techniques are falling out of favor because their use has a high rate of complications. [72] The laser partially vaporizes and coagulates the tumor; the necrotic tissue sloughs, and the lumen is restored. Although perforation is a risk, this procedure may be preferable to surgical therapy in elderly patients with other serious medical problems. In some patients with advanced cancer, intestinal obstruction may not be amenable to operative therapy because of high morbidity, mortality, or complication rates or unresectability. Obstructive symptoms often can be controlled pharmacologically, but in a few cases, mostly in patients with proximal obstruction, endoscopic or surgical venting gastrostomy or jejunostomy can provide relief of nausea, vomiting, and colic. Anticholinergic, antispasmodic, and antisecretory drugs, such as hyoscine sulfate, can be effective in relieving colicky pain and the frequency and volume of emesis, but they may cause agitation or other side effects, especially in elderly patients. Octreotide 0.3 mg daily reduced nausea and vomiting more than hyoscine butylbromide 60 mg and may be helpful in refractory cases. [73] Antiemetic agents such as haloperidol can be helpful in reducing nausea and vomiting; they can be given subcutaneously by a pump for extended periods, whereas this is not possible with most phenothiazines. Morphine is probably the most effective analgesic drug if pain is a major factor.

The therapy for ileus is directed primarily toward treatment of the underlying or associated disease. Sometimes the underlying cause is not apparent, and the ileus resolves spontaneously with supportive care. Resolution of the associated condition is accompanied by return of intestinal or colonic motor activity. Drugs inhibitory to motility should be withdrawn if possible. In most cases, however, a thorough search for the underlying cause is the key to successful management. The simple step of patient positioning is critically important. When the patient lies supine, the gas stays in the anterior-located transverse colon, but it quickly moves to the left colon when the patient lies on the right side. The gas moves to the rectosigmoid when the patient becomes supine and into the rectum when the patient is prone or in the knee-chest position.

In the postoperative period, temporary ileus is expected; if it persists for more than a few days, the routine causes of ileus should be evaluated, beginning by correction of metabolic abnormalities and withdrawal of any drugs that inhibit motility, especially narcotics. If evidence of impaired propulsion appears after the ileus has resolved and the patient has begun oral intake, a new mechanical obstruction must be suspected, and proper steps must be initiated to document and treat the problem. Obstructions from early adhesions tend to be fibrinous and rarely compromise the vascular supply. Most patients respond to conservative therapy and do not require a second operation. [74], [75]

In acute colonic pseudoobstruction, fluid and electrolyte imbalances must be corrected; nothing is allowed by mouth, and nasogastric suction is instituted. The patient must be examined frequently to assess abdominal girth, tenderness, and signs of peritoneal irritation or sepsis. As in intestinal ileus, treatment must be directed toward any underlying cause, such as sepsis, and inhibitory drugs should be withdrawn. Gentle enemas may aid in evacuation of the distal bowel, and plain abdominal radiographs should be obtained regularly to determine changes in colonic diameter. If the condition can be anticipated or recognized early, it is likely that massive dilation can be prevented with supportive care. Although 12 cm is often stated as the upper limit of colonic diameter for which medical management can be continued, a rapidly expanding colonic diameter is stronger evidence of impending perforation and a more important indicator of risk than a single numeric value. Furthermore, the risk of perforation appears to correlate better with the duration of dilation than with the diameter. [76] If there is any question whether a mechanical obstruction is present, a contrast enema or colonoscopy is mandatory before resorting to drug therapy. If an ominous degree of dilation appears, some method of rapid decompression must be instituted because perforation, massive peritoneal soilage, and fatal sepsis may result. A surgical cecostomy has been the standard therapy and rarely may be indicated if all other measures fail. Colonoscopic deflation has been recommended enthusiastically, with a reported success rate of 75% to 100%. [77], [78] The use of limited air and carbon dioxide insufflation and a large-bore aspiration catheter dragged alongside the colonoscope with a snare may facilitate the procedure. If the procedure is to be of long-term benefit, the decompression catheter should be left for continuous aspiration. Colonoscopy has its own inherent risk of perforation, especially in a poorly prepared colon, and the recurrence rate approaches 50%. The latest approach involves the slow intravenous infusion of a cholinergic agonist. Neostigmine is given slowly over 3 to 5 minutes with mandatory cardiovascular monitoring; at a dose of 2.5 mg in 100 mL saline. Decompression by passage of flatus occurs in 3 to 30 minutes in greater than 90% of cases. [79], [80] and [81] Acute colonic pseudoobstruction may not be as hazardous as previously thought, and dilation usually resolves spontaneously with conservative positioning and nonendoscopic and nonsurgical treatment (86%). [82] Present information supports initial conservative therapy of acute colonic pseudoobstruction, but careful clinical monitoring continues to be mandatory because perforation remains a disastrous complication. If the problem is unresponsive or worsens, pharmacological therapy should be tried. Colonoscopic deflation should be the next step, followed

by operative cecostomy in the event of failure.

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CHAPTER 42

Don W. Powell

APPROACH TO THE PATIENT WITH DIARRHEA

GENERAL EPIDEMIOLOGY

Diarrheal diseases affect developed nations quite differently than they do developing nations. In developed nations, diarrheal disorders are primarily of economic significance. Total costs (direct plus indirect) in the United States were estimated as more than $4 billion (year 2000 dollars) for acute diarrheas and $2 billion for chronic diarrhea plus inflammatory bowel disease (IBD). [1] Other investigators have estimated the cost of all food-borne illnesses in the 1980s as more than $28 billion per year. [2] Although diarrheal diseases are a minor cause of death in industrialized nations, they are among the leading causes of death and morbidity in developing countries. Since the early 1980s, global death rates from diarrheal diseases, mostly affecting infants and small children, have decreased from 5 to 10 million per year to 3 to 4 million. [3], [4]

Diarrhea related to acquired immunodeficiency syndrome (AIDS) is covered elsewhere (see Chapter 124), as are the details of infectious diarrhea (see Chapter 74, Chapter 75, and Chapter 88) and many specialized forms of diarrhea (see Chapter 56, Chapter 76, Chapter 77, Chapter 78 and Chapter 79, Chapter 83, Chapter 85, Chapter 86, Chapter 95, and Chapter 125, Chapter 126, Chapter 127, Chapter 128, Chapter 129 and Chapter 130).

GENERAL DEFINITION

Normal stooling frequency ranges from three times a week to three times a day. [5], [6] Diarrhea was defined by Doctors Roux and Ryle in 1924 as “the too rapid evacuation of too fluid stools.” [7] A decrease in stool consistency or fluidity and stools that cause urgency or abdominal discomfort are more likely to be termed *diarrhea* by patients than increases in frequency alone. [5], [8] The physician or clinical investigator often chooses to define diarrhea as a physical sign (24-hour excretion weight or volume) rather than a symptom. Healthy children and adults have daily stool weights of less than 200 g, [9] and infants have a daily stool weight of less than 10 g/kg. [10] Even though stool consistency probably best defines diarrhea, it cannot be easily measured. Thus, stool weights in excess of 200 g/24 h may be the most easily obtainable, objective definition of diarrhea. This definition may cause misdiagnosis in 20% of patients with loose stools of less than this weight. [8] Twenty-seven percent of both men and women report one loose stool in the previous month. [11] Because stool composition varies from 60% to 85% water, [8], [12], [13] this definition implies that diarrhea is a disease of intestinal water and electrolyte transport.

A stool pattern of increased frequency, with either no change in consistency or a 24-hour stool output of less than 200 g, often accompanies motility disorders (irritable bowel syndrome [IBS]) or anorectal disease (proctitis). Some term this *pseudodiarrhea. Incontinence* (which is not necessarily diarrhea) is defined as the involuntary release of rectal contents. It is a relatively common (2%–10% of the population) and increases with age. It is reported to occur in 50% of nursing home patients. [14], [15] and [16] Incontinence is more common if stool is liquid, and it is present more often in the setting of abnormal neuromuscular function (abnormalities of afferent sensation or efferent motor function) or pelvic problems (trauma to or disease of the sphincter or pelvic muscles). [14], [16] Thus, it is more common in elderly patients, in persons in poor health, and in postpartum women.

Aside from water content, the factors governing normal stool weight and consistency are not completely understood. *Consistency* is best defined as the ratio of fecal water to the water-holding capacity of insoluble solids. [8] Fiber content and bacterial mass, which make up one half of the dry weight of stool, are probably the major components of these fecal solids. [17] Increasing fiber (wheat bran) ingestion from 10 to 26 g/d increases average stool weight from 100 to 149 g/d and increases average stool frequency from less than once to twice per day. [9] The weight of women’s stool is less than that of men by one half, reflecting differences in dietary fiber intake or hormonal factors. [9] There is some controversy whether the ovarian cycles do [9] or do not [18] influence stool weight and consistency. Both exercise [19] and stress [20] increase stool weight, presumably by decreasing jejunal or colonic fluid and electrolyte absorption. Even personality factors correlate with stool weight. [6] Finally, intralumenal bile acid content may influence stool weight, because the ingestion of cholestyramine [21] or aluminum hydroxide, [22] potent bile acid-binding substances, induces constipation. As described later, abnormal stool weight (>200–300 g/24 h) is almost always the result of a failure of intestinal solute and fluid absorption or actual solute and fluid secretion.

PATHOPHYSIOLOGY OF DIARRHEA

Abnormal Motor Function

In the early 20th century, diarrhea was thought to be caused primarily by increased gastrointestinal motility. Although gastrointestinal motility plays a role in diarrhea (see Chapter 11, Chapter 73, and Chapter 86), most of the diarrheal conditions that have been studied have shown alterations of both intestinal fluid and electrolyte transport and the induction of propagative forms of intestinal motility. What is less clear is whether there is diarrhea caused specifically and only by abnormal motility, a condition termed *diarrhée motrice.* The diarrhea caused by autonomic dysfunction in patients with hexosaminidase B deficiency (Sandhoff disease) may be such a type of diarrhea. [23] Perhaps the “pseudodiarrhea” of anorectal disease and the rapid small intestine transit of IBS are other examples of motility-induced diarrhea. Most clinicians, however, would agree with the concept of the diarrhea spiral shown in Figure 42-1.

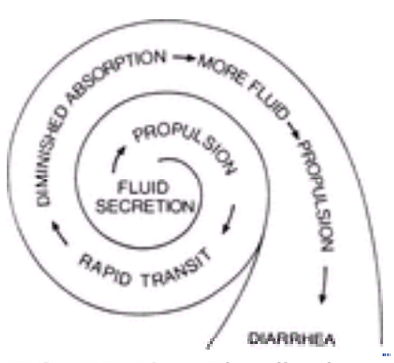

FIGURE 42-1. The diarrhea spiral. An increase in the amount of intralumenal fluid resulting from abnormalities in intestinal fluid and electrolyte transport initiate propulsive activity both by reflex and as a result of the same pathophysiological processes that disturbed epithelial function. Rapid transit follows with decreased contact time and perhaps also with diminished surface area. This causes diminished absorption, more intralumenal fluid, more propulsion, and finally diarrhea. (From Read NW. Diarrhée motrice. Clin Gastroenterol 1986;15:657.)

Abnormal Fluid and Electrolyte Transport

Decreased Absorption As shown in Table 42-1, entering the duodenum each 24 hours is 8 to 10 L of fluid containing 800 mmol sodium (Na^+), 700 mmol chloride (Cl^-), and 100 mmol potassium (K^+). Two liters of this duodenal load is derived from the diet; the remainder comes from secretions of the salivary glands, stomach, liver, pancreas, and the duodenum itself. The small intestine normally absorbs all but 1.5 L of this fluid, and this is the volume characteristically presented to the colon. The colon, in turn, absorbs all but approximately 100 mL of this fluid, which contains approximately 3, 8, and 2 mmol of Na^+, K^+, and Cl^-, respectively. Although the maximum absorptive capacity of the small intestine remains undefined, the capacity of the normal adult human colon is 4 to 5 L/24 h. [24] Theoretically, diarrhea can result from decreased absorption by either the small intestine or the colon. If either deranged epithelial transport mechanisms or the presence of nonabsorbable solutes in the intestinal lumen reduce the absorptive capacity of the small intestine by 50%, the daily volume of fluid then presented to the normal colon (approximately 5 L) would exceed its absorptive capacity. A stool excretion of up to 1000 mL would result, which, by definition, is diarrhea. Alternatively, if the colon is deranged so it cannot absorb even the 1.5 L normally presented to it by the small intestine, then diarrhea (stool volume >200 mL/24 h) would result.

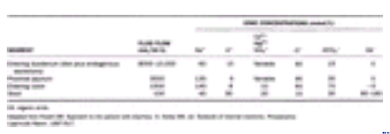

TABLE 42-1 Organ Physiology of Human Intestinal Water and Solute Transport

Increased Secretion In the decade after 1965, scientists rediscovered and documented the ability of the intestine to secrete as well as to absorb fluid and electrolytes, and the result was an entirely new concept: intestinal secretion as a pathophysiological mechanism of diarrhea. The subsequent discoveries that neurotransmitters, hormones, bacterial enterotoxins, inflammatory mediators, and cathartics all stimulated intestinal Cl^- and water secretion through changes in intracellular cyclic adenosine monophosphate, cyclic guanosine monophosphate, or ionized calcium (Ca^{2+}) further developed this concept (see Chapter 14). Although initially it was thought that bacterial enterotoxins cause secretion only by a direct effect on enterocyte receptors, 50% or more of the intestinal secretion initiated by bacterial enterotoxins in vivo comes about from stimulation of receptors on enterochromaffin cells that release hormones that activate the enteric nervous system, secondarily stimulating the enterocyte. [25], [26] Furthermore, inflammatory mediators (adenosine, histamine, serotonin [5-HT], hydrogen peroxide, platelet-activating factor, leukotrienes, and prostaglandins) released from immune cells (mucosal mast cells and resident phagocytes such as eosinophils, macrophages, and neutrophils) and mesenchymal cells (myofibroblasts, endothelium, and smooth muscle) in the lamina propria and submucosa are also capable of

initiating intestinal secretion. [27], [28] These mediators may directly stimulate the enterocyte and may also activate the enteric nervous system. Studies of intestinal electrolyte and water transport have demonstrated that the intracellular messengers also inhibit electrically neutral NaCl absorption, in addition to stimulating electrogenic Cl^- and bicarbonate (HCO_3^-) secretion by enterocytes of the small intestine and colon. Because the direction of net fluid movement is in response to the net direction of solute movement, a submaximal secretory stimulus gives the appearance of an inhibition of absorption rather than stimulated secretion.

Normal Intestinal Physiology

There are two general categories of diarrheal pathophysiology—malabsorption and secretion—also referred to as osmotic and secretory diarrheas, respectively. To understand this concept, it is necessary to consider how a normal intestine alters intralumenal ionic concentrations and osmolality and, subsequently, how these parameters are perturbed by secretory agonists or by the presence of nonabsorbable (osmotic) solutes.

The fasting intestinal flow rate in healthy humans averages approximately 2.5 mL/min in the upper jejunum and 0.4 to 0.9 mL/min across the ileocecal valve. [24] After meals, flow rates depend on the rate of gastric emptying, the rates of pancreatic and biliary secretion, and the osmolality of the ingested meal. Rates may approximate 20 to 50 mL/min in the upper jejunum and 5 to 10 mL/min across the ileocecal valve. Regardless of whether a subject ingests a hypotonic meal, such as a steak with an osmolality of 230 mOsm/kg H_2O, or a hypertonic meal, such as milk and a doughnut with an osmolality of 630 mOsm/kg H_2O, the very permeable duodenum allows the movement of water and electrolytes into or out of the lumen, rendering the meal approximately isotonic by the time it reaches the proximal jejunum (Fig. 42-2). At this point, the electrolyte content is essentially that of plasma (Fig. 42-3). Furthermore, the volume of this meal is augmented by gastric, pancreatic, biliary, and duodenal secretions such that the 313-mL milk and doughnut meal expands to 1200 mL and the 645-mL steak meal approaches 2000 mL by the time it reaches distal duodenum or proximal jejunum. However, the high-carbohydrate hypertonic meal is handled differently from the high-protein hypotonic meal. The rapid digestion of starches and lactose into sugars presents a large osmotic load to the proximal small bowel, and considerable amounts of fluid must passively enter to equilibrate the osmolality. After ingesting this meal, it may be at the midjejunum before efficient absorption of the fluid and electrolytes begins. In contrast, after ingesting the hypotonic steak meal, absorption begins virtually in the duodenum (see Fig. 42-2). In either case, as the chyme moves toward the colon, the electrolyte concentrations in the lumenal fluid remain approximately those of plasma, except for Cl^-, which is reduced to concentrations of 60 to 70 mM, and HCO_3^-, which is increased to a similar concentration, as the result of the Cl^- and HCO_3^- transport mechanisms residing in the small intestine (see Fig. 42-3).

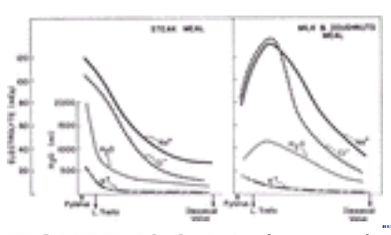

FIGURE 42-2. Intralumenal volumes and electrolyte content in the human small intestine after a hypotonic (steak) meal and a hypertonic (milk and doughnut) meal. (From ref. [12] and Fordtran JS, Locklear TW. Ionic constituents and osmolality of gastric and small intestinal fluids after eating. Am J Dig Dis 1966;11:503.)

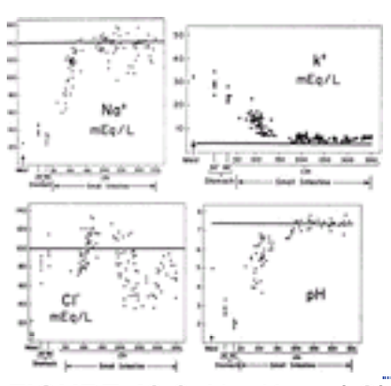

FIGURE 42-3. Na, K, and Cl concentrations and pH values in human gastric and small intestine fluid after a steak meal. The horizontal line in each figure indicates normal plasma concentrations or pH. (From ref. [12].)

In the colon, the amiloride-sensitive Na^+ transport mechanism of the colonocyte and the low epithelial permeability allow this segment to extract Na^+ and fluid very efficiently from the contents. In fact, the crypts of the distal colon may act like suction devices to extract fluid from the stool efficiently, thus forming solid feces. [29] As a result, the Na^+ content of stool decreases to approximately 30 to 40 mM, and poorly absorbed divalent cations such as magnesium (Mg^{2+}) and Ca^{2+} are concentrated to values of 5 to 100 mM, depending on diet (see Table 42-1). K^+ values increase from 5 to 10 mM in the small bowel to 75 to 90 mM in stool as the result of both the lumen negative electrical potential difference, which favors the movement of cations from blood to lumen, and the active K^+ secretory mechanisms present in the colon. [30] The anion concentrations in the intestinal lumen change drastically in the colon. Bacterial degradation of carbohydrate (i.e., unabsorbed starches, sugars, and fiber) creates short-chain fatty acids that attain concentrations of 80 to 180 mM. [31], [32] At colonic pH, these are present as organic anions, mainly acetate, propionate, and butyrate. Depending on the concentrations and quantities of organic anions created, stool pH may decrease to 4 or lower. Even though colonic bacteria degrade carbohydrates and increase the concentration of organic anions and therefore the number of osmotically active particles, the osmolality of stool, if measured as soon as it is passed, is approximately that of plasma, 280 to 310 mOsm. [31], [32], [33] and [34] If stool osmolality is measured hours to days after passage, even if it has been stored in deep freeze, the osmolality may increase to greater than 350 mOsm because of continued bacterial degradation of stool carbohydrate in the collecting container.

Pathophysiology of Osmotic Diarrheas

Contrast the events just described with what transpires after a lactase-deficient subject ingests a lactose test meal, [35] or a physiologically normal subject ingests a nonabsorbable solute such as polyethylene glycol (PEG) or Mg^{2+}. [33] Although the same dilution of the meal occurs in the duodenum (Fig. 42-4), the lactase-deficient subject is unable to reabsorb the fluid because the unabsorbable molecule lactose is not metabolized to absorbable glucose and galactose. Similarly, the osmotic activity of ingested PEG, which is nonabsorbable, or poorly absorbable Mg^{2+} causes fluid entry into the small bowel, rendering the intralumenal solutions isosmotic with plasma. Intralumenal Na^+ concentrations drop to less than 80 mM, and the permeable jejunum cannot absorb Na^+ against such a steep lumen-to-plasma gradient. What happens to the chyme when it reaches the colon depends on the nature of the unabsorbed solute. If the solute is nonmetabolizable (i.e., PEG or Mg^{2+}), some Na^+ and water may be absorbed by the colon, which can concentrate Na^+ from lumenal concentrations of less than 30 mM, but it cannot absorb all the excess stool water. In fact, there is a linear relationship between the ingested osmotic load of PEG and stool water output (stool weight). If the unabsorbed solute is a carbohydrate (i.e., lactulose or lactose) that can be metabolized by colonic bacteria, it effects stool weight differently. The disaccharide is metabolized to short-chain fatty acids (organic anions) that obligate retention of inorganic cations, significantly increasing the number of osmotically active particles in the colon. This increases the solute load, promoting the movement of more fluid into the colon. Although some of the organic anions and fluid are absorbed as they traverse the colon, the unabsorbed carbohydrate, the organic anions with their obligate cations, and fluid are excreted in the stool. [33]

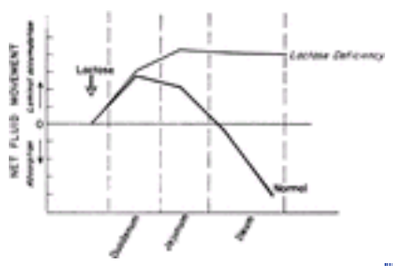

FIGURE 42-4. Changes in fluid absorption and secretion in the small intestine of normal and lactase-deficient subjects after a lactose meal of 50 g/m ^{2} body surface area in 400 mL of distilled water. (From Binder HJ, Bayless TM, Whiting DS. Pathophysiology of diarrhea. Unit VIIB. In: The undergraduate teaching project in gastroenterology liver disease. Timonium, MD: American Gastroenterological Association, 1979; data from ref. [35].)

With ingestion of either unabsorbable solute (i.e., Mg^{2+} or PEG) [33] or unabsorbed carbohydrate (i.e., lactulose [33] or, in some persons, lactose [35]), a considerable amount of the osmolality of stool results from the nonabsorbed solute, so there is a significant difference, or gap, between stool osmolality and the sum of the electrolytes in the stool (see section " Stool Weight, Response to Fast, and Stool Osmotic Gap").

The osmotic solutes causing osmotic diarrheas can be derived from either exogenous (ingested) or endogenous sources and may result from congenital or acquired malabsorptive disease. The causes are listed in Table 42-2 and are discussed in more detail in later sections.

Exogenous
Laxatives
Polyethylene glycol/saline (Golytely), $Mg(OH)_2$ (Milk of Magnesia), $MgSO_4$ (Epsom salts), Na_2SO_4 (Glauber or Carlsbad salt), Na_2PO_4 (neutral phosphate), lactulose
Antacids
Those containing MgO or $Mg(OH)_2$
Dietetic foods, candy or chewing gum, and elixirs containing sorbitol, mannitol or xylitol
Acarbose (α-glucosidase inhibitor)
Olestra, orlistat (lipase inhibitor)
Miscellaneous drugs
Long-term ingestion of colchicine, cholestyramine, neomycin, paraaminosalicylic acid, biguanides, methyldopa
Endogenous
Congenital
Specific malabsorptive diseases
Disaccharidase deficiencies (lactase, sucrase-isomaltase, trehalase)
Glucose-galactose or fructose malabsorption
Generalized malabsorptive diseases
Abetalipoproteinemia and hypobetalipoproteinemia
Congenital lymphangiectasia
Enterokinase deficiency
Pancreatic insufficiency (cystic fibrosis or Schwachman syndrome)
Acquired
Specific malabsorptive diseases
Postenteritis disaccharidase deficiency
Generalized malabsorptive diseases
Pancreatic insufficiency (alcohol), bacterial overgrowth, celiac sprue, rotavirus enteritis, parasitic diseases (giardiasis, coccidiosis), metabolic diseases (thyrotoxicosis, adrenal insufficiency), inflammatory disease (eosinophilic enteritis, mastocytosis), protein-calorie malnutrition, short-bowel syndrome, jejunoileal bypass

TABLE 42-2 Classification of Osmotic or Malabsorptive Diarrhea

Pathophysiology of Secretory Diarrheas

In secretory diarrheas, particularly in the fasting state, the major solutes in the lumen are Na^{+}, K^{+}, Cl^{-}, and HCO_3^{-}. It is the failure to absorb these electrolytes, their active secretion, or both that determines the amount of fluid entering or leaving the colon. In secretory diarrheas, therefore, the stool osmolality will be very close to twice the concentrations of stool Na^{+} plus K^{+}.

As is the case with osmotic causes of diarrhea, the stimuli causing secretory diarrhea may be of exogenous origin or they may be endogenous (Table 42-3). Endogenous substances such as dihydroxy bile acids [36] and dietary long-chain fatty acids may cause secretory diarrhea, [37], [38] particularly if they have been hydroxylated by intestinal microflora. [39] The important concept here is that there are "malabsorbed" endogenous detergents that may lend an element of secretory diarrhea to otherwise malabsorptive diseases. The detergent stool softener dioctyl sodium sulfosuccinate [40] and the cathartic ricinoleic acid (castor oil) [41] are examples of exogenous long-chain fatty acids that stimulate colonic secretion.

Exogenous
Laxatives
Phenolphthalein, anthraquinones, bisacodyl, oxyphenisatin, senna, aloe, ricinoleic acid (castor oil), dioctyl sodium sulfosuccinate
Medications
Diuretics (furosemide, theophilline): asthma medication (theophylline); thyroid preparations; methionine
Cholinergic drugs; glaucoma eye drops and bladder stimulants (acetylcholine analogs or mimetics); myasthenia gravis medication (cholinesterase inhibitors); cardiac drugs (quinidine and quinine); gout medication (colchicine); prokinetic drugs (metoclopramide); antihypertensives (ACE inhibitors); H_2 blockers (ranitidine); antidepressants (selective serotonin reuptake inhibitors); antineoplastics; chenodeoxycholic acid
Prostaglandins (misoprostol); 5-aminosalicylic acid (sulfasalazopyrin); gold (may also cause colitis); NSAIDs
Toxins
Metals (arsenic); plant (mushroom, e.g., Amanita phalloides); organophosphates (insecticides and nerve poisons); seafood toxins (ciguatera, scombroid poisoning, paralytic, diarrhetic, or neurotoxic shellfish poisoning); coffee, tea, or cola (caffeine and other methylxanthines); ethanol; monosodium glutamate
Bacterial toxin
Staphylococcus aureus, Clostridium perfringens and C botulinum, Bacillus cereus
Gut allergy without histologic change
Endogenous
Congenital
Microvillus inclusion disease
Congenital chloridorrhea (absence of Cl^{-}/HCO_3^{-} exchanger)
Congenital Na diarrhea (absence of Na^{+}/H^{+} exchanger)
Congenital enterocyte heparan sulphate deficiency
Bacterial enterotoxins
Vibrio cholerae, toxigenic Escherichia coli (LT and ST), Campylobacter jejuni, Yersinia enterocolitica, Klebsiella pneumoniae, Clostridium difficile, Staphylococcus aureus (toxic shock syndrome)
Endogenous laxatives
Dihydroxy bile acids and long-chain fatty acids, especially hydroxylated ones
Hormone-producing tumors
Pancreatic cholera syndrome and ganglioneuromas (VIP); medullary carcinoma of thyroid (calcitonin and prostaglandins); mastocytoses (histamine); villous adenoma (secretagogue unknown, ? prostaglandins)
(LT, heat-labile toxin; ST, heat-stable toxin; VIP, vasoactive intestinal polypeptide.)

TABLE 42-3 Classification of Secretory (Deranged Electrolyte Transport) Diarrhea

Pathophysiology of Inflammatory Diarrheas

Some diarrheas are characterized by enterocyte damage and death, with minimal to severe inflammation, in which both malabsorption and secretion occur. As described in Chapter 14, the paradigm of neuroimmunophysiology allows us to postulate how the adherent and invasive microorganisms of infectious diarrhea, as well as idiopathic inflammatory diarrheas such as IBD, cause diarrhea. [26], [27] and [28] After initial colonization, adherence, or epithelial invasion by microorganisms, or the initiated inflammation of IBD, there is a release of various cytokines such as interleukin-1 (IL-1), tumor necrosis factor-a (TNF-a), and chemokines such as IL-8 from the epithelium and subepithelial myofibroblasts (Fig. 42-5). IL-8 is a potent chemotactic molecule that activates resident phagocytes and recruits new phagocytes to the lamina propria. If the chemotactic substance released by the epithelium (IL-8) or by lumenal microorganisms (chemotactic peptides) is also sufficiently concentrated in the lumen of the bowel, neutrophils may cross the epithelium to form a crypt abscess. [42] Release of mediators such as prostaglandins, leukotrienes, platelet-activating factor, and hydrogen peroxide from phagocytes induces intestinal secretion by acting on the enterocyte and also by activating enteric nerves. [25], [26], [43], [44] and [45]

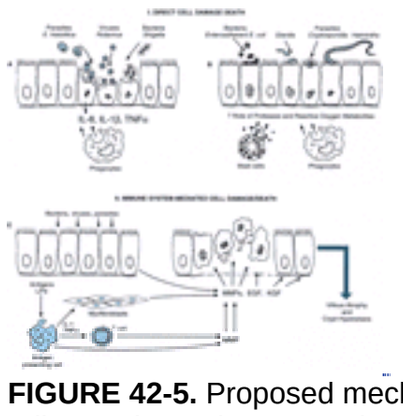

FIGURE 42-5. Proposed mechanism of enterocyte damage and death. Direct damage as a result of (**A**) enterocyte penetration and (**B**) secretion of toxic products by adherent bacteria or parasites. There may be a role for immune cell products in the damage also (see **C**). Indirect (immune system-mediated) damage or death as a result of (**C**) activation of macrophages and dendritic cells (antigen-presenting cells), T-lymphocytes, and myofibroblasts. These cells secrete matrix metalloproteinases (*MMPs*), epidermal growth factor (*EGF*), and keratinocyte growth factor (*KGF*), which remodel the tissue, resulting in villous atrophy and crypt hyperplasia.

There are three general mechanisms of inflammatory diarrhea. Most are accompanied by enterocyte brush border damage and some by enterocyte cell death with ulceration. [45]

1. Lumenal or invading microorganisms or parasites may directly damage or kill enterocytes [46] (see Fig. 42-5 *A*).
2. If the infecting microorganism is complex, such as a nematode, then intestinal anaphylaxis will play an important role in the diarrhea (see Fig. 42-5 *B*). Infection with complex worms causes mild inflammatory enteritis, but it is accompanied by the development of IgE and IgG antibodies against the worm. With continued infection or reinfection, the antibodies cross-link IgE or IgG receptors on the mast cells, resulting in the explosive release of mast cell inflammatory mediators such as histamine, adenosine, prostaglandins, and leukotrienes. This pathophysiological response in the gut is not unlike that experienced in the upper airway in allergic rhinitis or asthma, in which there is an initial anaphylactic response followed by inflammation. The resulting intestinal secretion washes out, [45] and the muscle contraction physically expels the nematode from the intestine. [47]
3. Immunologic mechanisms may cause enterocyte damage and death by products released from polymorphic neural leukocytes, epithelial macrophages, and T-lymphocytes.

In all these situations, epithelial cells, macrophages, and subepithelial myofibroblasts may release matrix metalloproteinases that attack the basement membrane and interstitial matrix molecules. [48], [49], [50] and [51] The result is exfoliation of the epithelium and subsequent remodeling of the matrix with the development of villous atrophy and crypt hyperplasia in the small intestine [48] and an attenuated surface but hyperplastic irregular regenerative crypts in the colon. [52] The culmination of these events may be the presence of damaged or immature cells on rudimentary or nonexistent villi of the small intestine and on the surface of the colon. These immature absorptive cells have poor disaccharidase and peptide hydrolase activity, reduced or absent Na^+-coupled sugar or amino acid transport mechanisms, and reduced or absent NaCl absorptive transporters. Conversely, the crypt cells and the new, immature villus or surface cells maintain their ability to secrete Cl^- (and perhaps HCO_3^-). At the same time, the release of inflammatory mediators from the inflammatory cells of the lamina propria stimulates secretion from the hyperplastic crypt and immature villus or surface cells. Immune-mediated vascular damage may cause protein to leak from capillaries. If severe ulceration has occurred, exudation from capillaries and lymphatics may contribute to the diarrhea. After damage, epithelial restitution and proliferation begin secondary to release of prostaglandins and growth factors, such as transforming growth factor, hepatocyte growth factor, keratinocyte growth factor, epidermal growth factor, and fibroblast growth factor, from the epithelial cells, immune cells, and myofibroblasts. [50], [51], [53] These processes repair the epithelial surface. Repeated bouts of inflammation may lead to fibrosis rather than repair. [54] Lymphocyte and neutrophil activation also release IL-1 and tumor necrosis factor-a in the blood, and their action on the brain accounts for some of the systemic effects of severe inflammation (fever, malaise, anorexia, and obtundation). These cytokines also activate corticotropin-releasing factor in the brain that stimulates the pituitary-adrenal axis and initiates the glucocorticoid stress response.

There are four general categories of inflammatory diarrhea: infection, hypersensitivity, cytostatic (anticancer) agents, and idiopathic (possibly autoimmune) diseases (Table 42-4).

With Minimal to Moderate Inflammation
Infections
Bacteria (enteroadherent or enteropathogenic *Escherichia coli*)
Viruses (rotavirus and Norwalk agent, human immunodeficiency virus)
Parasites (*Giardia*, *Cryptosporidium*, *Isospora*, *Cyclospora*, *Ascaris*, *Trichinella*)
Mixed organism (tropical sprue, bacterial overgrowth)
Cytostatic (anticancer) agents
Chemotherapy (mucositis)
Radiation therapy (acute or chronic radiation enteritis, radiation sickness)
Hypersensitivity
Nematode infestation, food allergy
Idiopathic or autoimmune
Microscopic (lymphocytic) and collagenous colitis, Canada-Cronkhite syndrome, graft versus host disease
Medications
Nonsteroidal antiinflammatory drugs; simvastatin; ticlopidine; cimetidine; lansoprazole; gold (Auroitin); cyclosporine
With Moderate to Severe Inflammation With or Without Ulceration
Infections
Destruction of enterocytes (*Shigella*, enteroinvasive *E coli*, *Entamoeba histolytica*, hookworm)
Penetration of mucosa (*Salmonella*, *Campylobacter jejuni*, *Yersinia enterocolitica*, *Mycobacterium avium* complex, Whipple disease)
Hypersensitivity
Celiac sprue, milk or soybean protein hypersensitivity, eosinophilic gastroenteritis, nematode infestation
Drug-induced colitis (gold, methyldopa)
Idiopathic or autoimmune disorders
Ulcerative colitis or proctitis, Crohn's disease, lymphoma

TABLE 42-4 Classification of Inflammatory Diarrheas

ACUTE DIARRHEAS: DEFINITION

Acute diarrheas are defined as those of less than 2 to 3 weeks' duration and, at most, 6 to 8 weeks' duration. Although they have many causes, the most common acute diarrheas are those caused by infectious agents and drugs or chemicals.

ACUTE INFECTIOUS DIARRHEAS

Epidemiology

Developing Versus Developed Countries The most important epidemiologic factor for the diagnosis and management of acute infectious diarrhea is whether the diarrhea is occurring in developing countries or in developed countries. [1], [2], [3] and [4] In 1995, acute infectious diarrhea caused more than 3 million deaths worldwide. This mortality occurred in children younger than 5 years of age in developing nations, where two thirds of the world's population live in extreme poverty, and in areas of rapid urbanization, crowded substandard housing with inadequate sewage disposal and inadequate water supplies, insufficient food with lack of refrigeration, poor education (particularly in regard to personal hygiene), and a fundamental lack of access to health care. [2] In addition to socio-economic status and hygienic practices, in developing countries there is a relationship between diarrhea incidence and ambient air temperature, [55] which may explain the higher incidence of diarrhea in tropical emerging nations. Deaths occur in developed nations as well. The incidence of infant diarrheal deaths in the United States decreased by 75% from 1968 to 1991, [56] but diarrhea was still the principal cause of death for more than 500 children each year. [2], [56] Twenty to 40 of these deaths resulted from rotavirus infection. [57] Reported diarrheal death rates in the United States are probably underestimated, but they may be as high as 5000/year from food-borne infections alone. [58] The death

rate for children and young adults in the United States is 2 to 3/100,000 persons and is approximately 15/100,000 persons for persons more than 74 years old. [59], [60] Thus, even though there are far more episodes of diarrhea in children compared with adults, the elderly are truly the age group at risk for death from diarrheal diseases in this country. In developing countries, children have 100 to 1500 episodes per 100 child-years, and approximately 10% of the episodes result in dehydration requiring therapy. [2] In the United States, children have an average of two to three episodes per year. In the total population of the United States, food-borne illness alone accounts for 76 million episodes of diarrhea and 325,000 hospital admissions per year. [58] With improvement in sanitation and education in the United States, rates of parasitic diarrheas have declined, but, in the 1990s, in those diseases with person-to-person transmission (e.g., rotavirus) or food- or water-borne transmission (e.g., *Salmonella* or *Campylobacter* organisms), the incidences doubled. [61] Preliminary data from the United States Centers for Disease Control and Prevention (CDC) indicate that the rates of *Salmonella, Shigella, Listeria, Escherichia coli,* and *Yersinia* infections (but not *Vibrio* infection) have declined by 20% to 30%, most likely as the result of increased attention to food safety.

Sporadic Diarrhea Most infectious diarrheas are acquired by fecal-oral transmission, which can occur by way of water, food, or person-to-person contact. Although microorganisms can be spread by any of these vehicles, certain classical relations exist between vehicle and disease (Table 42-5). Water systems may be contaminated by human waste as the result of poor sewage systems or by animal feces from farm run-off material. Diarrheal disease results if the water is not properly purified. Beef, pork, or poultry may be the source of infection if it is poorly cooked. More commonly, infection comes from contamination of food-preparing surfaces by organisms present on these meats; the organisms then are spread to uncooked foods, such as salads, that are prepared on these same surfaces. Transmission also comes from minute inoculation of certain foods that is followed by bacterial proliferation because the food is either poorly refrigerated or inadequately heated before serving. This commonly occurs at picnics, banquets, or restaurants. Person-to-person transmission can occur through aerosolization (rotavirus), through contamination of hands (*Clostridium difficile*), or through contamination of fomites. [62], [63] Spread also comes from sexual activity, particularly oral-genital or oral-anal contact.

TABLE 42-5 Epidemiologic Clues to Infectious Diarrhea

Patients with infectious diarrhea complain of nausea, vomiting, and abdominal pain and have watery, malabsorptive diarrhea or dysentery (bloody diarrhea and fever) (see Chapter 74, Chapter 75, and Chapter 88). Both large and small bowel infections can present with any combination of these signs and symptoms, but certain patterns tend to occur characteristically (Table 42-6). Certain enteric infections are accompanied by systemic symptoms that may be clues to their diagnosis or account for delayed, often severe, complications [64], [65], [66], [67], [68], [69], [70], [71], [72], [73] and [74] (Table 42-7). Conversely, patients with systemic diseases (see Table 42-6) may have diarrhea as a significant component of the illness, either because the gastrointestinal mucosa is the portal of entry, as in hepatitis, listeriosis, and legionellosis, or because of the systemic release of inflammatory mediators, such as the interleukins or bradykinin, that cause intestional secretion.

TABLE 42-6 Correlations Between Pathophysiology and Symptoms of Infectious Diarrhea

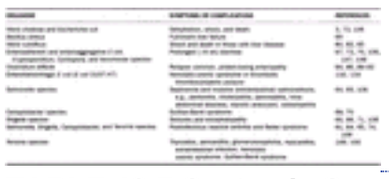

TABLE 42-7 Systemic Symptoms and Complications of Enteric Infections

Food- and Water-Borne Diarrheas Estimates of the incidence of food-borne illness in the United States range from 6 to 81 million cases per year [58], and the World Health Organization (WHO) believes the incidence to be 300 times more frequent than suggested by reported cases. Emerging food-borne diseases over the past several decades include the enteritides serotype of *Salmonella* and *Campylobacter jejuni.* Deaths from *E coli* O157:H7 and *Cyclospora* infections in the United States, United Kingdom, and Japan, as well as the deaths related to bovine spongiform encephalology (mad cow disease) in the United Kingdom, have produced a crisis in public confidence regarding government-regulated food safety. [75], [76] Various factors relating to the industrialization of food consumption and eating habits are responsible for the surge in food-borne illness. [2], [76] Food poisoning occurs not only from invasive organisms but also from toxin producers (see Table 42-5). The infectious food poisonings can occur with ingestion of either fin fish or shellfish. [77], [78], [79], [80] and [81] Fish can become contaminated in their own environment (especially the filter-feeding bivalve molluscs such as muscles, clams, oysters, and scallops), or fish can be contaminated by food handlers. [80] The infectious organisms contaminating seafood include invasive organisms such as hepatitis virus, Norwalk virus, *Salmonella, Shigella, Campylobacter,* and *Plesiomonas,* and watery diarrhea–producing organisms such as *Aeromonas, Clostridium botulinum,* and *Vibrio cholerae*. [80] Organisms that are specific for seafood are the noncholera vibrios, such as *V parahemolyticus,* which causes either watery or bloody diarrhea, and *V vulnificus,* which causes watery diarrhea, but also fatal septicemia especially in those with liver disease. [82], [83] Food poisoning also occurs with chemicals such as monosodium glutamate, heavy metals or insecticides, and natural toxins found in mushrooms or seafood. [77], [78], [79] and [80] Ciguatera, diarrhetic, paralytic, neurotoxic, amnestic, and pfiesteria fish poisonings are all caused by dinoflagellates ingested by fish or shellfish. [77], [78], [79], [80] and [81] Puffer fish poisoning is caused by a toxin (tetrodotoxin) contained only in this fish, and scombroid poisoning comes from the breakdown of flesh in certain fish with the release of histamine.

High-Risk Groups

Antibiotic-Associated Diarrheas Diarrhea may occur in up to 20% of patients receiving broad-spectrum antibiotics, but only a portion (30%–50%) of these diarrheas are the result of *C difficile* infection (pseudomembranous colitis) (Table 42-8). [84], [85], [86] and [87] *C difficile*–negative diarrhea has been ascribed to antibiotic-induced abnormalities in colonic carbohydrate fermentation, with resulting decreased short-chain acid production, [87] to overgrowth of fungi (*Candida* species), [88] or to overgrowth of other, unidentified microorganisms. Non– *C difficile* diarrhea is usually mild and self-limited and clears either spontaneously or in response to cholestyramine therapy. *C difficile* colitis, however, can cause severe diarrhea that has significant morbidity and mortality and may present with a relapsing course after seemingly successful therapy with metronidazole or vancomycin. [89], [90], [91], [92] and [93] Antibiotic-associated diarrhea can follow treatment with any antibiotic, although broad-spectrum antibiotics such as clindamycin and third-generation cephalosporins are particularly problematic. [85], [86], [94] The disease can be nosocomial and is increasing in frequency. The incidence of *C difficile* and other antibiotic-associated diarrheas may be reduced by the concomitant administration of probiotics. [93] Relapses of *C difficile* colitis not responding to repeated courses and tapering regimens of antibiotics [89] may respond to long-term cholestyramine therapy, [90] intravenous immunoglobulin infusion, [92] or administration of normal, donated stool through the colonoscope. [91]

Antibiotic Use
Outpatient clinics
Hospital
Recent Travel
Developing nations
Peace Corps workers
Campers (ground water)
Homosexuals, Prostitutes, and Intravenous Drug Users
Gay bowel syndrome
Acquired immunodeficiency syndrome
Day-Care Facilities
Children
Secondary contacts (family members)
Institutions
Mental institutions
Nursing homes
Hospitals

TABLE 42-8 High-Risk Groups for Infectious Diarrhea

Traveler's Diarrhea Not only are North American travelers to developing countries at high risk of acute infectious diarrhea, but so are travelers on airplanes and cruise ships, where errors in food preparation can lead to common-source epidemics. [80] Bacterial agents account for 85% of traveler's diarrhea, with enterotoxigenic *E coli* (ETEC), *Shigella, Campylobacter, Aeromonas, Plesiomonas, Salmonella* and noncholera vibrios leading the list. [95], [96] Prevention through education or chemoprophylaxis (doxycycline, trimethoprim-sulfamethoxazole, or fluoroquinolones) and both fluid replacement and antibiotic treatments are discussed in more detail in Chapter 56.

Sexually Transmitted Diarrheas Men who have sex with men and prostitutes are apt to develop infectious diarrhea through the oral-fecal route. "Gay bowel syndrome" is discussed in Chapter 88, and AIDS diarrhea is covered in detail in Chapter 124.

Day-Care Diarrhea Diarrhea is extremely prevalent in the more than 6 million children in the United States attending day care, and it usually involves those organisms that colonize at a low inoculum dose (e.g., *Shigella, Giardia, Cryptosporidium*) or those that are spread easily (e.g., rotavirus, astrovirus, adenovirus). [63], [97] However, almost any organism can be isolated in outbreaks of day-care diarrhea. The mechanism of transmission in day-care centers is person-to-person contact by way of fecal contamination of hands and fomites (e.g., toys, surfaces in diaper changing areas, bathroom tap, and flush handles). The secondary attack rate from day-care diarrhea ranges between 10% and 20%, representing an important source of infection for parents and siblings as well. [63], [98]

Diagnosis of Acute Infectious Diarrheas

Differential Diagnosis An algorithm for the evaluation of acute infectious diarrheas is given in Figure 42-6. This approach relies heavily on consideration of the epidemiology of acute infectious diarrhea as described earlier and on ruling out other causes of acute watery or bloody diarrhea. [99]

FIGURE 42-6. Algorithm for the diagnostic approach to acute diarrhea. (Adapted from Aranda-Michel J and Giannella R. Acute diarrhea: a practical review. Am J Med 1999;106:670.)

The differential diagnosis of acute watery diarrhea includes the food toxins, drugs, and medications listed in Table 42-2 and Table 42-3. Although these drugs are the most common culprits, any medication can cause diarrhea. In children, acute appendicitis may be misdiagnosed as "acute gastroenteritis," thus delaying surgery. [100] The differential diagnosis of acute bloody diarrhea in adults includes infectious causes, superior mesenteric arterial or venous thrombosis or ischemic colitis (see Chapter 131), IBD (see Chapter 83), and drug-induced colitis (see Chapter 85). Although the patient's age and the presence of manifestations of atherosclerosis may suggest mesenteric vascular insufficiency or ischemic colitis, older people also may have more severe clinical manifestations with invasive infectious agents. Radiographically, enterohemorrhagic *E coli* infection can mimic ischemic colitis, with submucosal hemorrhage presenting as thumbprinting on the flat plate of the abdomen. Ulcerative proctitis or colitis and Crohn's disease can present with an acute course that suggests infectious enterocolitis, or vice versa. Although the colonoscopic and radiographic appearance of the various invasive enteritides can mimic IBD with aphthous-like ulcers, segmental colitis, or pancolitis, colonic biopsy will yield histological hallmarks that differentiate IBD from infectious diarrheas. *C difficile* infection may have the classic pseudomembranous enterocolitis appearance by radiography or endoscopy, but on occasion the pseudomembrane is not present, particularly in pancytopenic patients undergoing cancer chemotherapy, and then the disease looks more like IBD. On rare occasions, the opposite mistake is made, and blood-filled macrophages in the stool of a patient with ulcerative colitis are mistaken for blood-filled trophozoites of amebiasis, or chronic ischemic colitis masquerades as pseudomembranous colitis. To complicate matters further, the colonic mucosa inflamed with ulcerative colitis appears to be more susceptible to colonization by pathogenic enteric bacteria. *Salmonella, Campylobacter,* or *C difficile* infection may accompany IBD and may confuse the diagnosis and treatment. Drugs that may induce colitis indistinguishable from ulcerative colitis include gold (administered for rheumatoid arthritis) and methyldopa (for hypertension).

Laboratory Diagnosis of Infectious Diarrheas The cost of making the diagnosis of infectious diarrhea and of delivering specific (antibiotic) therapy is high, especially for a disease that is common and is usually mild and self-limited. Because fewer than 10% of stool specimens are positive for pathogens, [99], [101] the cost of a single positive stool culture is quite high (more than $1000), if stools are cultured indiscriminately. [102], [103] However, the cost can be reduced to $30 per culture if only *Campylobacter, Salmonella,* and *Shigella* are sought and if only liquid stools are cultured. Special rules apply to the culture of stool in hospital-acquired diarrhea [104], [105] (see section "Infectious Nosocomial Diarrhea"). The use of antibiotics in infectious diarrhea is also controversial. [82], [99], [106] Therefore, the questions revolve around who should undergo a diagnostic evaluation, who should be treated and when, and whether treatment should consist of only symptomatic therapy or symptomatic therapy plus specific antibiotics. The algorithm in Figure 42-6 uses discriminating symptoms to determine whether fecal specimens should be sent for laboratory diagnosis. [99], [103] Fecal leucocyte stains [107] are not useful for inpatients [108] and have poor sensitivity for ruling out invasive infectious diarrhea. [109] Stool excretion of granulocyte marker proteins is being evaluated as perhaps a better surrogate for fecal leukocytes. [110], [111], [112] and [113] In addition, certain organisms that can cause diarrhea are not generally identified by routine diagnostic methods. For example, certain causes of bloody diarrhea, such as *Yersinia, Plesiomonas,* and enterohemorrhagic *E coli* O157:H7, may require specific special culture techniques or can be identified only with type-specific antisera. [114] Similarly, *Aeromonas, Cryptosporidium, Cyclospora, Microspora,* and noncholera *Vibrio* organisms, which cause watery diarrhea, may require special laboratory attention for culture and identification. The enterotoxigenic, enteropathogenic, enteroinvasive, enteroadherent, and enteroaggregative *E coli* organisms are diagnosed only by methods available in research laboratories. Therefore, the physician may need to communicate the presumptive diagnosis to the laboratory. The enteroadherent bacteria and certain parasites such as *Giardia* and *Strongyloides* may be difficult to detect in stool and may best be diagnosed by intestinal biopsy. *C difficile* is best diagnosed by toxin assay, [84], [85] and [86] although the rapid enzyme-linked immunosorbent assays commonly used in hospitals have a sensitivity of only 70% to 90%, so false-negative results can occur. [115] Finally, 20% to 40% of all acute infectious diarrheas remain undiagnosed even with the application of all laboratory techniques.

Treatment of Acute Infectious Diarrheas

The treatment of diarrhea can be divided into symptomatic therapy (fluid replacement and antidiarrheal) and specific antimicrobial therapy. [116], [117] and [118] Because death in most instances of acute diarrhea is caused by dehydration, a cardinal principle in the management of any diarrhea consists of assessment of the degree of dehydration and replacement of fluid and electrolyte deficits. [99], [118], [119] Severely dehydrated patients, particularly those with altered mental status, should be rehydrated with intravenous Ringer lactate or saline solutions to which additional K^+ and $NaHCO_3$ may be added as necessary. Alert patients should be given oral replacement solution (ORS). Whereas experience in developing countries has demonstrated the efficacy of ORS in treating severe dehydrating diarrhea, ORS use in developed countries has lagged behind. This may account in part for some of the morbidity and mortality still observed in the United States. [119], [120], [121] and [122] In mild

to moderate dehydration, ORS can be given to infants and children in volumes of 50 to 100 mL/kg over a period of 4 to 6 hours; adults may need to drink up to 1000 mL/h. [120], [122] After the patient is rehydrated, ORS is given at rates equaling stool loss plus insensible losses until the diarrhea ceases. The WHO solution is endorsed for use in both rehydration and maintenance therapy worldwide, although some are concerned that the high Na^+ content (90 mM) may lead to hypernatremia and the high osmolarity (311 mOsm/L) may worsen diarrhea. Consequently, many clinicians recommend alternating full-strength WHO ORS with equal volumes of water or giving a mixture of two parts ORS with one part water or formula during the postrehydration, maintenance phase of ORS therapy. Studies are emerging proving the superiority of hypotonic ORS solutions (Na 75 mM, osmolarity 245 mOsm/L). [120], [123]

The addition of amino acids to glucose-based ORS or the substitution of rice gruel or cereal for glucose has created “super ORS” solutions that may be even better than the conventional WHO solution. [118], [124] A newer concept for rehydration is to add amylase-resistant starch (pectin) to the ORS. This starch escapes digestion and absorption in the upper bowel and is then broken down to short-chain fatty acids by bacteria in the colon, where it promotes fluid absorption. [125], [126] Clearly, the WHO solution and the commercial ORS solutions are superior to Gatorade, Coca-Cola, or fruit juices, which were used in the past for rehydration in this country (Table 42-9). ORS can be used safely with loperamide antidiarrheal medications in traveler’s diarrhea. [127]

SOLUTION	Na (mM)	K (mM)	Cl (mM)	CITRATE (mM)	GLUCOSE* (mM)
WHO solution	90	20	80	30	111 (20)
Reduced osmolarity ORS	75	20	65	10	75 (13.5)
Rehydralyte	75	20	65	30	139 (25)
Pedialyte	45	20	35	30	139 (25)
Resol	50	20	50	34	111 (20)
Ricelyte	50	25	45	34	(30)
Gatorade	23.5	<1	17		(40)
Coca-Cola	1.6	<1		13.4†	(100)
Apple juice	<1	25			(120)
Orange juice	<1	50		50	(120)
Chicken broth	250	8		0	0

* Values in parentheses represent grams of carbohydrate.

† Rice syrup solid rather than glucose.

Adapted from Dr. John D. Levine MM. Treatment of diarrhea. Infect Dis Clin North Am 1988;2:719.

TABLE 42-9 Composition of Oral Replacement Solutions for the Treatment of Diarrhea

A devastating effect of recurrent diarrheal diseases is malnutrition. At one time, complete or partial bowel rest was recommended during acute diarrhea. It is now clear that patients with acute diarrhea should be fed, not starved. [128], [129] Continued breast-feeding or half-strength formula for infants is recommended. Ad libitum diets low in fiber, Na^+-rich soups, and foods with high sugar content are preferred for older children and adults.

Bismuth subsalicylate (Pepto-Bismol, Procter & Gamble, Cincinnati, OH) is safe and efficacious in bacterial infectious diarrheas. [99], [116], [117] and [118] It may not have antidiarrheal activity in viral diarrhea, raising the question whether its main effect in traveler’s diarrhea relates to its antibacterial or antiinflammatory action or perhaps to the ability of its clay vehicle to adsorb enterotoxins. Kaolin-pectin or smectic preparations may be minimally effective. [130], [131] Because of the possibility of worsening the colonization or invasion of the organism by paralyzing intestinal motility and evidence that the use of motility-altering drugs may prolong microorganism excretion time, neither opiates nor anticholinergic drugs are recommended for infectious diarrheas. [99], [117], [118] However, it has been shown that loperamide can be both useful and safe in traveler’s diarrhea, provided it is not given to patients who have high fever or to those with blood or pus in the stool, especially when the drug is administered concomitantly with effective antibiotics. [99], [117], [118] The anxiolytics and antiemetics that decrease sensory perception may make symptoms more tolerable and are generally safe.

Antibiotic therapy in the infectious diarrheas is controversial. [82], [99], [106], [117], [118] There are certain infectious diarrheas in which treatment is recommended: shigellosis, cholera, traveler’s diarrhea, pseudomembranous enterocolitis, parasitic infections, and sexually transmitted diseases. Patients with mild disease and those who are clearly improving may not need antibiotic treatment. There are other diarrheas in which treatment may not be indicated because there is no effective therapy: viral diarrhea and cryptosporidiosis. Treatment of *E coli* O157:H7 infection is not recommended at present because current antibiotics do not appear to be helpful, and the incidence of hemolytic uremic syndrome may be greater after antibiotic therapy. [132], [133] There are several diseases in which the indications are less clear but treatment is usually recommended: infection with the noncholera vibrios, prolonged or protracted infection with *Yersinia,* early in the course of campylobacteriosis, *Aeromonas* and *Plesiomonas* infections, and nursery outbreaks of enteropathogenic *E coli* diarrhea. Regardless of the cause of infectious diarrhea, patients should probably be treated if they are debilitated with malignant disease, are immunosuppressed, have an abnormal cardiovascular system or valvular, vascular, or orthopedic prostheses, have hemolytic anemia (especially if salmonellosis is involved), or are extremely young or old. Treatment is also advised for those with prolonged symptoms and those who relapse. These guidelines were developed for patients with salmonellosis, [134] but they are useful guidelines for all infectious diarrheas. Chapter 74, Chapter 75, Chapter 88, Chapter 125, and Chapter 126 outline the antimicrobials of choice for the various infections, as do references [82], [106], and [116], [117] and [118]. Although there is evidence that early treatment alters the course of infectious diarrhea, [134] it usually it takes 3 to 5 days after obtaining stools before specific organisms can be grown and identified. If treatment was warranted while awaiting laboratory diagnosis, the quinolones (e.g., ciprofloxacin), which have efficacy against most enteric infections, are the treatments of choice. [82], [106], [116], [117] and [118] Trimethoprim-sulfamethoxazole is second-line therapy in this setting. If the symptom complex suggests *Campylobacter* infection, erythromycin should be added. Prolonged diarrhea suggesting giardiasis or bacterial overgrowth should be treated with a course of metronidazole even if stools are negative for cysts.

PROLONGED INFECTIOUS DIARRHEAS

Severe Protracted Diarrhea in Infants and Children

Although classically a postinfectious diarrhea syndrome in infants and children of developing countries, [135], [136] severe protracted diarrhea can occur in milder forms (*postenteritis syndrome*) [137] or in a severe form in developed countries as well. Postulated causes of the syndrome are enteroadherent bacterial infection, uncleared cryptosporidial or *Cyclospora* infection, bacterial overgrowth, or infection with unknown organisms, which lead to disaccharidase deficiency in mild cases and profound generalized malabsorption in severe forms. [138], [139] Severe malnutrition and death (mortality up to 50%) can ensue. [140] Treatment includes dietary lactose exclusion in mild disease [137] and controlled feeding or total parenteral nutrition in those severely affected. [129] Metronidazole, tetracycline, trimethoprim-sulfamethoxazole, folic acid, and zinc therapy also may be of help. [140], [141] Where this disease ends and tropical sprue begins is uncertain; the difference may be only the age of the patient. [139]

Tropical Sprue

This disease of unknown origin affects those in certain tropical parts of the world, including the Indian subcontinent and Asia, the West Indies, northern part of South America, parts of Central America, and central and southern Africa (see Chapter 75). It can occur in visitors residing in these areas for as short a time as 1 to 3 months. Its acute onset suggests an infectious origin, perhaps with a coccidia protozoa, or another unknown infection that either persists or in some way leads to brush border damage and bacterial overgrowth with perpetuation of the disease. [138] Small bowel histology may show minimal villus blunting and inflammatory infiltrate or may reveal severe villus atrophy and crypt hyperplasia. Abnormal pancreatic exocrine function may occur in tropical sprue as it does in celiac sprue. [142] If the patient is removed from the tropical areas and the mucosal change is mild, the disease may remit spontaneously. [143] A combination of tetracycline and folic acid is effective therapy.

Persistent Diarrhea in Travelers

Protracted diarrhea lasting more than 3 to 4 weeks has been seen in up to 10% of returned travelers. [143], [144] Undiagnosed infection with bacteria or protozoal organisms that typically causes long-lasting diarrhea may be the cause (see section that follows), and stool cultures and evaluation for ova and parasites as well as antimicrobial treatment may lead to cure. Because these patients usually have already received routine traveler’s diarrhea antibiotics (e.g., trimethoprim-sulfamethoxazole or quinolones) tetracycline or metronidazole, which may be effective against enteroadherent *E coli,* protozoal organisms, and

nonspecific bacterial overgrowth, may be given when the results of stool evaluations for pathogens are negative. [144], [145] The pathophysiological mechanisms proposed here are similar to those proposed for severe protracted diarrhea and tropical sprue (see earlier). The point at which protracted traveler's diarrhea ends and tropical sprue begins may be a matter of obtaining a biopsy specimen for small bowel histology.

Protracted Infectious Diarrhea in Adults

In the United States, some organisms typically cause a prolonged course of diarrhea. Organisms that are difficult to diagnose and are known to cause protracted or prolonged diarrheas include enteropathogenic (enteroadherent) *E coli,* [145] *Giardia,* [146] *Amoeba,* [82] *Cryptosporidium,* [147] *Aeromonas,* [72], [148] and *Yersinia enterocolitica.* [149], [150] *C difficile* infection is often difficult to clear, and five or more relapses have been observed. [85], [86], [89], [90], [91] and [92] Patients with chronic diarrhea should have these diagnoses excluded. If none of these organisms are found, a therapeutic trial of metronidazole or trimethoprim-sulfamethoxazole may be indicated. [143], [144]

Infectious Diarrhea–Induced Irritable Bowel Syndrome and Brainerd or Epidemic Chronic Diarrhea

As many as 25% of patients will experience IBS–like symptoms (pain, bloating, urgency, sense of incomplete evacuation, loose stools) for 6 months or longer after documented infectious diarrhea. [151], [152], [153] and [154] The pathophysiology is thought to be unresolved, mild intestinal inflammation. In some patients, the syndrome may cause acquired bile acid malabsorption, and these patients may respond to cholestyramine therapy (see section " Bile Acid Diarrhea"). [153], [154] and [155]

Severe and prolonged diarrhea after raw milk ingestion was reported in outbreak form in Brainerd, Minnesota in the 1980s. [156] Patients developed prolonged chronic watery diarrhea with weight loss and were often mistakenly diagnosed as having IBS. It also has been reported in other states in association with raw milk ingestion or untreated water ingestion. Many of these patients have microscopic inflammation on colonic biopsy. The disease occurs in sporadic form and probably represents a severe form of infectious diarrhea–induced IBS, as discussed earlier. It may also be the same as *chronic idiopathic diarrhea* discussed in the section "Chronic Diarrheas."

NOSOCOMIAL DIARRHEAS

Diarrhea is either the first or second most common nosocomial illness [157] among hospitalized patients and those residing in chronic care facilities for the retarded, the mentally disturbed, or the elderly. Often this is a hidden problem, known only by the nurse's aide who changes the bed sheets. In the intensive care setting, it occurs in 30% to 50% of patients, [15], [158] and in chronic care facilities more than one third of patients have a significant diarrheal illness each year. [159], [160] This is a multifactorial condition whose recognized causes are discussed in the following subsections.

Fecal Impaction

Clinical lore has it that the most common cause of diarrhea in hospitalized or institutionalized patients is fecal impaction. Such paradoxical diarrhea and incontinence appear to be most common in patients with dementia or psychosis. [161] Although the validity of this clinical impression remains uncertain, performing a rectal examination and perhaps a flat and upright abdominal radiograph in such patients can certainly be recommended.

Medications and Weight-Reducing Agents

Any of a patient's medications may initiate diarrhea. However, certain medications are more apt to cause diarrhea than others [162] (see Table 42-3 and Table 42-4). Antibiotic-associated diarrhea (see earlier) is certainly the most common manifestation of drug-related diarrhea. Orlistat, a lipase inhibitor used to treat obesity, and acarbose, an a-glucosidase inhibitor used to treat diabetes, can both cause diarrhea. [162] Olestra, a nonabsorbable fat substitute used for frying foods, may also cause diarrhea at doses greater than 40 g/d. [163]

Elixir Diarrhea

Drugs such as theophylline or KCl made up in liquid formulations (elixirs) may cause diarrhea because of the high content of sorbitol used to sweeten the elixir. [164] This is the iatrogenic equivalent of "chewing gum diarrhea" (see section " Sorbitol and Fructose Diarrhea"). Patients receiving medications in the liquid form through feeding tubes may receive more than 20 g of sorbitol daily.

Enteral Feeding

An important but poorly understood cause of diarrhea is tube feeding, particularly in the critically ill patient. [165], [166] and [167] Up to 35% of patients receiving tube feeding develop diarrhea. Various pathophysiological factors are hypothesized: bacterial contamination of the enteral formula; administration of hypertonic solutions that cause diarrhea by inducing a form of "dumping syndrome"; administration of lactose-containing formulas to lactase-deficient subjects; administration of sorbitol-containing elixirs; administration of low-Na^+ formulas that result in considerable blood-to-lumen Na^+ diffusion in addition to fluid movement if the formula is hypertonic as well; and hypoalbuminemia in malnourished patients, a condition that alters the oncotic or Starling forces in the gut capillaries, thus preventing absorption or inducing secretion. Although there are experimental studies that support all these possibilities, there are no clear data supporting any single or group of pathophysiological processes (see Chapter 53). Furthermore, more mundane causes of diarrhea (e.g., concomitant medication, *C difficile* infection) can also occur in these patients.

Infectious Nosocomial Diarrhea

Patients in mental institutions have high incidences of infection by bacterial pathogens, protozoan parasites (*Entamoeba histolytica* and *Giardia*) and helminths. [160], [168] Infectious diarrheas are also common in acute-care hospitals; these illnesses account for more than 20% of nosocomial infections and are second only to respiratory infections on pediatric wards. [160] The rates are particularly high in intensive care settings (8 cases per 100 admissions). [169] In the intensive care setting, a role has been postulated for tube feeding as a source of infection in combination with histamine (H_2) blockers, which eradicate the gastric acid barrier. The most common cause in the past was infection with *Salmonella* species, and it still occurs. [170] However, since 1980, *C difficile* has accounted for more than 50% of hospital cases. [171] Although shigellosis still occurs, it is now a rare nosocomial infection in hospitals. [172] The likelihood of a nosocomial infection caused by *Salmonella* or *Shigella* organisms in the tertiary hospital is so rare that routine cultures for *Salmonella* and *Shigella* and ova or parasite examinations are not cost effective and should not be ordered if diarrhea begins 3 to 4 days after hospital admission, provided the patient is not more than 65 years old and has no preexisting diseases, the patient does not have AIDS or neutropenia, there is no evidence of a nosocomial outbreak, and the patient has no nondiarrheal manifestation of infection (e.g., fever, abdominal pain, erythema nodosum). This dictum is known as the *modified 3-day rule*. [104], [105]

Immunosuppressed patients are another important group susceptible to nosocomial diarrhea. Viral infections (rotavirus, astrovirus, adenovirus, and coxsackievirus) may be important causes of nosocomial infectious diarrheas in bone marrow transplant units. [169], [173] In this setting, infectious diarrhea must be differentiated from the diarrhea of graft versus host disease.

Outbreaks of hemorrhagic *E coli* and *C difficile* infections have been recognized in hospitals and nursing homes. [94], [174], [175] Some of the strokes, injuries from falls, and even myocardial infarctions occurring in nursing home settings could be caused by the hypovolemia and toxic state induced by these nosocomial diarrheas. Infection control measures and restricting use of broad-spectrum antibiotics may reduce the incidence of nosocomial *C difficile* diarrhea. [176]

Hospital-acquired diarrheas may be a causative factor in other nosocomial hospital infections, such as infection of the urinary tract. [177] The impact of nosocomial diarrhea on the duration and cost of hospitalization and on morbidity is probably substantial.

Cancer Treatment

The incidence of acute, mild diarrhea with chemotherapy or radiation therapy is quite high, approaching 100% with some agents or irradiation regimens. [178] Radiation therapy also causes chronic diarrhea (see Chapter 132). Nausea, vomiting, and diarrhea are dose- and age-related phenomena. Chemotherapy diarrhea is more likely with specific forms of chemotherapy, such as the following: 5-fluorouracil, irinotecan (CPT-11), interferon-a2A, topotecan, and IL-2. [178], [179] and [180] The combination of 5-fluorouracil plus leucovorin causes severe watery diarrhea. [178], [179] The incidence of diarrhea with IL-2 therapy approaches 80%. [180] Radiation may induce diarrhea either through damage to segments of bowel during pelvic irradiation or through damage to the entire bowel if high-dose, total body radiation is received (see Chapter 132). Total-body radiation at low doses (1.5 Gy) causes only nausea and vomiting; watery or bloody diarrhea ensues at total-body doses of greater than 6 Gy. Pelvic irradiation over a period of 4 weeks with doses of 3 to 4 Gy also may cause diarrhea. Current treatment for both chemotherapy- and radiation-induced diarrhea is symptomatic and includes antimotility drugs and cyclooxygenase blockers. [178] Octreotide may be helpful in severe chemotherapy- or radiation-induced diarrhea. [178]

RUNNER'S DIARRHEA

Gastrointestinal disturbances including anorexia, heartburn, nausea, vomiting, cramps, urgency, and diarrhea are quite common in those who exercise vigorously, particularly marathon runners and triathletes. [181] Watery, self-limiting diarrhea may occur in 10% to 25% and is particularly common (40%–70%) in women runners. The mechanisms operative in runner's diarrhea are unclear but may involve release of gastrointestinal hormones such as gastrin, motilin, or vasoactive intestinal polypeptide (VIP) or release of inflammatory mediators such as prostaglandins. [182], [183] A role for ischemia has been postulated because of the occurrence of ischemia colitis in marathon runners. [184], [185] Many treatment regimens have been used, but none have been studied thoroughly. Because mouth-to-cecum transit time is either normal or delayed with exercise, [186] antimotility drugs may not help, but they may be tried. Nonsteroidal antiinflammatory agents (NSAIDs) are taken by many runners, but it is not clear whether they help in this condition.

CHRONIC DIARRHEAS: DEFINITION, CLASSIFICATION, AND EPIDEMIOLOGY

Definition and Classification

Chronic diarrheas are those of at least 4 weeks' duration and, more certainly defined, 6 to 8 weeks' duration. They fall into three categories: osmotic (malabsorptive) diarrhea (see Table 42-3), secretory diarrhea (see Table 42-4), and inflammatory diarrhea (see Table 42-5). It would be convenient clinically if the character of the stool correlated well with the pathophysiology; that is, if an obviously steatorrheic stool occurred with all cases of malabsorptive diarrhea, watery stool only with secretory diarrheas, or bloody stool only with inflammation. Unfortunately, this is not the case. Carbohydrate malabsorption causes watery diarrhea, as does inflammation that is not severe enough to cause intestinal ulceration. Inflammation also may cause malabsorption; for example, celiac sprue, whose hallmark is intestinal malabsorption, is actually caused by inflammation. [187] Furthermore, malabsorptive diseases often have an element of intestinal secretion. [188] For clinical purposes, it is reasonable to classify diarrheas as steatorrhea, watery diarrhea, or inflammatory diarrhea, realizing that these categories are mixed with regard to pathophysiology. Such a categorization directs the physician to certain diagnostic algorithms.

Epidemiology

Although specific prevalence and incidence figures for the major diarrheal diseases are provided in the chapters on those specific entities, the precise incidence or prevalence of chronic diarrhea is not known because the appropriate population studies have not been performed. The best estimate is that chronic diarrhea occurs in approximately 5% of the United States population. [189], [190]

STEATORRHEA (MALABSORPTIVE DISEASES)

Although the three major nutrients—fat, carbohydrate, and protein—all may be malabsorbed, clinical symptoms usually follow from malabsorption of either carbohydrate or fat. [189], [190], [191] and [192] Protein or amino acid malabsorption (azotorrhea) occurs but is not clinically recognized unless it is severe enough to cause malnutrition or unless specific amino acid transport defects cause congenital systemic disease (see Chapter 19 and Chapter 77). Malabsorption of electrolytes and water is also part of the pathophysiology of malabsorptive diarrheas. The gut's limited ability to absorb high concentrations of divalent ions (i.e., Mg^{2+}, sulfate [SO_4], and phosphate [PO_4]) results in clinically evident diarrhea if these ions are ingested in excess. Nonetheless, the generalized malabsorptive diseases present as steatorrhea. Therefore, an understanding of fat absorption is necessary to understand malabsorptive diseases. Based on the normal physiology of fat absorption (see Chapter 18), fat malabsorption can be divided into three broad categories: intralumenal maldigestion, mucosal malabsorption, and postmucosal malabsorption related to lymphatic obstruction. The diseases listed in Table 42-2 can be allocated to one or more of these three general categories.

Intralumenal Maldigestion

Cirrhosis and Bile Duct Obstruction Bile duct obstruction from cancer of the pancreas can cause steatorrhea, presumably through both pancreatic and bile salt insufficiency. There is a 25% to 100% incidence of mild steatorrhea in patients with cirrhosis. This may result from inadequate micelle formation from bile salt insufficiency; [193], [194] however, secondary factors, including malnutrition, portal hypertension, bacterial overgrowth, and drugs (e.g., neomycin), also may play a role. In both cancer of the pancreas and severe liver disease, diarrhea is not usually a significant clinical problem; the fat malabsorption is usually mild. Certainly, the weight loss in these two conditions is multifactorial.

Pancreatic Exocrine Insufficiency Chronic pancreatitis may cause weight loss because of anorexia or because of fear that eating will initiate pain by activating pancreatitis. After at least 90% of the exocrine secretory capacity of the pancreas is lost, chronic pancreatic exocrine insufficiency supervenes, and malabsorption leads to continued weight loss in spite of a good to excellent appetite (see Chapter 95). Increased intestinal transit time may add to the poor intralumenal digestion and malabsorption. [195] Up to 70% of patients with pancreatic calcification have chronic pancreatitis severe enough to cause malabsorption. Major degrees of pancreatectomy also cause fat malabsorption that is poorly responsive to pancreatic enzyme replacement. [196] Cystic fibrosis is a childhood equivalent of chronic pancreatic insufficiency, but the weight loss in this disease is probably caused as much by the anorexia of chronic infection [197] as it is by the malabsorption induced by pancreatic enzyme and bile acid deficiencies. [198] Schwachman syndrome is another pediatric cause of pancreatic insufficiency, although some patients improve with age. [199] Somatostatinoma is a rare pancreatic islet tumor with highly variable symptoms that may present with gallstones, diabetes, and diarrhea. [200] It is the one neuroendocrine tumor in which the diarrhea is caused by steatorrhea rather than by intestinal secretion. Presumably, the steatorrhea is secondary to inhibition of pancreatic secretion.

Mucosal Malabsorption

Drugs The chronic ingestion of drugs such as colchicine, [201] neomycin, [202] paraaminosalicylic acid (PAS), [203] and the fenamate class of NSAIDs induces steatorrhea by enterocyte damage. Cholestyramine causes mild steatorrhea by binding bile acids. [204]

Infectious Diseases Parasites can cause malabsorption through brush border damage, particularly the protozoa *Giardia, Cryptosporidium,* and *Isospora* [205], [206] and [207] and the helminth *Strongyloides*. [208] Because these are treatable infections, they must be sought and rigorously excluded by stool examination or small intestine biopsy. Whipple disease and *Mycobacterium avium-intracellulare* complex are infectious agents that cause malabsorption. [209], [210] Unfortunately, *Mycobacterium avium-intracellulare* complex responds poorly to treatment (see Chapter 124). Chronic enteric infections, such as giardiasis associated with the agammaglobulinemias or cryptosporidiosis accompanying AIDS, also cause malabsorption. [211]

Autoimmune Enteropathies

Autoimmune enteropathy has been described in both newborn infants and children [212] and adults. [213] The histology of the small bowel resembles celiac disease, with subtotal or complete villous atrophy and increased lamina propria inflammatory cells, but test results for antigliadin and antiendomysial antibodies (AEAs) are negative, and patients do not respond to a gluten-free diet. Circulating autoantibodies have been found in these patients directed against the enterocytes, goblet cells, smooth muscle, thyroid, islet cells, and parietal cells and against the hemidesmosomes of epithelial cells (the same autoantibodies as in bullous pemphigoid). These patients require glucocorticoids and azathioprine or cyclosporine for control of their disease.

Nongranulomatous chronic idiopathic enterocolitis, also known as *ulcerative jejunitis,* presents with more severe inflammation than autoimmune enteropathy, manifesting superficial ulcerations in addition to the villous atrophy. [214] This disease is prominent in the small intestine but may involve the colon in half the patients. Patients with nongranulomatous chronic idiopathic enterocolitis may have an abrupt onset of their diarrhea and weight loss in early or middle adult life, and they often develop complications such as obstruction, perforation, and hemorrhage. Mortality is high (approximately 30%), although the patients appear to respond to corticosteroids and immune suppression therapy.

Immunoproliferative small intestinal disease is a collective term for immune enteropathies developing in patients in the Middle East or Mediterranean countries who have diseases otherwise known as *a-heavy-chain disease* or *Mediterranean lymphoma*. [215] Like patients with the autoimmune enteropathies, these patients present with malabsorption and sprue-like intestinal histology. a-Heavy-chain disease tends to be benign, but it may progress to malignant Mediterranean lymphoma. A common clonal origin of lymphocytes can be detected in both. Current views are that immunoproliferative small intestinal disease is one end of the spectrum of B-cell maltomas. [215]

T-cell lymphomas of the bowel may also be the cause of malabsorption and sprue-like histology, sometimes with intestinal ulcers (see section “ Celiac Sprue”). [216]

Mastocytosis and Eosinophilic Gastroenteritis Infiltrative immune system diseases include systemic mastocytosis [217], [218] and eosinophilic gastroenteritis, [219] in which gross distortion of the mucosa is associated with fat malabsorption. On occasion, steatorrhea may be profound, and these patients present with a sprue-like syndrome. In other patients, the watery diarrhea, systemic flushing, abdominal pain, tachycardia, and protein-losing enteropathy overshadow the steatorrhea.
Celiac Sprue The use of highly specific antibody tests for celiac disease—AEAs and antitransglutaminase (anti-tTG) antibodies, which have 95% to 100% sensitivity and specificity—have revolutionized our concepts of this disease. [220], [221], [222], [223], [224] and [225] Instead of being a rare (1 in 10,000 population) disease with a classical “malabsorption/malnutrition” presentation, screening studies suggest that the disease may be as common as 1 in 200, with many subclinical manifestations (Table 42-10). Some symptoms result from intestinal inflammation or malabsorption; other presentations occur as a result of coexisting autoimmunity. Furthermore, certain unrelated diseases are common to the human leukocyte antigen (HLA) phenotypes that are prevalent in patients with celiac disease: HLA DQ2, B8, DR3 and, rarely, DR5 or 7. [220], [221], [224]

TABLE 42-10 Clinical Presentations of Celiac Sprue

The term *sprue* comes from the Dutch *spruw,* which means *thrush,* in recognition of the extraintestinal manifestation of oral aphthae. These nonabsorptive/nonmalnutrition, extraintestinal manifestations may be an important clue to the disease (see Table 42-10). Currently, physicians do a poor job of diagnosis of celiac disease: only 50% of patients consider that they were diagnosed “promptly”; 27% consulted two or more gastroenterologists before the diagnosis; and only 30% to 50% consider their physician knowledgeable about diagnosis and treatment. [226] Given the frequency of celiac disease in certain associated disorders, screening should be considered for groups in which the incidence of celiac disease had been shown to be greater than 2.5%: first-degree family members [221], [224] and patients with type 1 diabetes, [227] selective immunoglobulin A deficiency, [228] autoimmune thyroid disease, [229] Sjögren syndrome, [230] Down syndrome, [231] IBS, [232] cerebellar ataxia, [233] unexplained osteoporosis, [234] epilepsy with posterior cerebral calcification, [235] lymphocytic gastritis, [236] and microscopic colitis. [237] Many other diseases in Table 42-10 have incidence approximating 2.5%, and patients with these diseases may, in the future, be candidates for screening. Patients with Sjögren or Down syndrome may not show intestinal disease on biopsy. [230], [231] It is not clear, therefore, whether there is a certain incidence of false-positive test results in these patients or whether they have occult disease. [231] Celiac disease appears to have a significant adverse effect on menses, fertility, pregnancy, miscarriage rate, intrauterine growth rate, and eventual birth weight. [238] Low birth rates have also been reported when it is the father who has celiac disease. [239] Patients with celiac disease have a 300-fold relative risk of small bowel T-cell lymphoma and probably a severalfold incidence of small bowel adenocarcinoma, [224], [228], [240] so patients presenting with these tumors should be screened for celiac disease as well. Some of these patients with T-cell lymphomas will present as having celiac disease unresponsive to gluten-free diet (refractory sprue). [240], [241] Patients labeled as having refractory sprue may have been misdiagnosed as having celiac disease and really suffer from autoimmune enteropathy. [241] The treatment of true “refractory sprue” is with steroids and immunosuppressive drugs. [241]
Dermatitis Herpetiformis This form of skin disease is associated with sprue-like intestinal morphology in 70% to 80% of cases. [242] The specificity and sensitivity of anti-tGT antibodies in this disease is unclear, with reports ranging from 50% [243] to 90%. [244] This blistering skin disease, which is characterized by IgA deposits in the dermal papilla, usually responds to dapsone, whereas the mucosal lesion responds to a gluten-free diet. The diet also seems to have a beneficial effect on the skin lesion, and about 50% of diet-adherent patients are able to stop dapsone medication. [242], [243] Cyclosporine may be useful in patients resistant to conventional therapy. [245] These patients may experience many of the same “extraintestinal” complications as patients with celiac disease, including thyroid disease [246] or lymphoma. [247]
Whipple Disease This is a systemic infectious disease (see Chapter 75) caused by an actinomycete, *Trophermyma whipplei,* [248] and classically involving middle-aged men (male-to-female is 5:1). [209], [210] The peak incidence occurs at ages 40 to 50 years, but it is reported in infants and in octogenarians. It presents with all the signs and symptoms of severe mucosal disease but has some additional characteristics: arthralgias in 65%, chills and fever in up to 40%, hypotension (blood pressure <110/60 mm Hg) in 70%, lymphadenopathy in more than 50%, and, most important, involvement of the central nervous system in a plethora of ways. [209], [210] A unique neuromuscular syndrome caused by central nervous system involvement is oculofacioskeletal myorhythmia. [210]
Lipoproteinemias Abetalipoproteinemia and hypobetalipoproteinemia are rare defects in chylomicron formation caused by abnormalities in microsomal transfer proteins or to molecular defects in apolipoprotein B itself. [249] Both conditions present with steatorrhea, acanthocytic red cells, ataxia, and retinitis pigmentosa (see Chapter 18 and Chapter 77). Patients with Tangier disease (absence of apolipoprotein A-I and apolipoprotein A-II) have yellow-orange streaks and spots in the tonsils and colonic mucosa. They may have diarrhea but not steatorrhea.

Postmucosal Obstruction

Intestinal lymphangiectasia can be either congenital or acquired in association with trauma, lymphoma, carcinoma, or Whipple disease. [250] This condition causes protein-losing enteropathy with significant steatorrhea. [250], [251] It is the classic form of postmucosal obstruction malabsorption. The unique clinical presentation—malabsorption of fat with loss of protein and lymphocytes, but normal absorption of carbohydrates—relates to the obstructed lymphatics channels, which are the route of absorption for fat and for the recovery of lymphocyte and protein-laden lymph. The absorption of carbohydrates and amino acids takes place by way of the portal circulation and remains unaffected. Immune deficiency, both humoral and cellular, may result. [252] Octreotide has been used successfully to treat this disease. [253]

Mixed Causes of Steatorrhea

Bacterial Overgrowth Stasis syndromes cause steatorrhea as well as an inflammatory and secretory form of diarrhea in patients with anatomic bowel obstruction (e.g., Crohn’s disease), small bowel diverticulosis, and motility disorders or in the elderly. Steatorrhea results from deconjugation of bile salts, causing poor micelle formation. However, brush border injury, mucosal inflammation, hydroxylation of fat with resulting fatty acid diarrhea, and changes in intestinal motility all play a role in this disease. Surgical correction of obstruction, antibiotics for bacterial overgrowth, and stimulation of motility with octreotide or prokinetic agents may improve symptoms, depending on the cause (see Chapter 78).
Short Bowel Syndrome Extensive intestinal resection that leaves less than 200 cm of jejunum-ileum remaining represents another complicated, multifactorial form of steatorrhea resulting from the lack of sufficient absorptive surface, decreased transit time, and diminished bile salt pool [254] (see Chapter 79). It is part of a larger syndrome called *intestinal failure,* which also includes parenchymal bowel disease (e.g., Crohn’s disease) and motility disorders in which nutrition is in peril. [255] The diarrhea is heightened by the osmotic effect of nonabsorbed solutes, by gastric hypersecretion, perhaps by bacterial overgrowth, and conceivably even by intestinal secretion. Glutamine-rich diets, exogenous growth factors, and long-acting octreotide hormones show promise as new treatments. [256], [257]

Metabolic Diseases Diseases such as thyrotoxicosis, [258] adrenal insufficiency, [259] autoimmune polyglandular syndrome, [260] protein-calorie malnutrition [261] and prolonged fasting [262] may result in malabsorption through different mechanisms. Thyrotoxicosis may simply shorten transit time and disturb the intralumenal phase of the fat absorption. Adrenal insufficiency appears generally to disturb intralumenal and mucosal absorption, as do protein-calorie malnutrition and prolonged fasting, which also cause villus atrophy. The malabsorption of polyglandular syndrome type I is related to a deficiency of cholecystokinin-producing enteroendocrine cells. As with liver disease, the clinical picture usually overshadows the diarrhea and malabsorption, and certainly the weight loss in these conditions is only partly related to the malabsorption.

WATERY DIARRHEAS

Ingestion of Nonabsorbable Solutes

Magnesium-Induced Diarrhea Persons ingesting significant amounts of magnesium-based antacids or high-potency multimineral-multivitamin supplements may have significant diarrhea with stool weights up to 2000 g/24 h. [263], [264] Occasionally, magnesium-containing laxatives are a cause of surreptitious diarrhea. [263], [264] and [265] Magnesium in tube-feeding preparations may play a role in the diarrhea of patients receiving high-volume liquid feedings. [266] These diarrheas are diagnosed with measurements of stool Mg^{2+} and osmotic gap (see section " Evaluation of Severe or Elusive Diarrhea ").

Sodium Anion Diarrheas Nonabsorbable sodium anion laxatives such as Na_2PO_4 (neutral phosphate) or Na_2SO_4 (Glauber or Carlsbad salt) and high concentrations of SO_4 in naturally occurring drinking water [267] induce osmotic diarrhea. When these substances are ingested factitiously, these diarrheas may be difficult to detect because these substances do not result in a calculated osmotic gap on stool analysis (see section " Evaluation of Severe or Elusive Diarrhea ").

Carbohydrate Malabsorption

Sorbitol and Fructose Diarrhea Carbohydrate malabsorption may be either specific or generalized (see Table 42-3). Diarrhea can result from the long-term ingestion of dietetic foods, candy, chewing gum, or medication elixirs that are sweetened with unabsorbable carbohydrates such as sorbitol (chewing gum and elixir diarrhea). [268], [269] Sorbitol and fructose are also present in pears, prunes, peaches, and apple juice, [270] and excessive ingestion of these foods results in diarrhea as well. Long-term ingestion of drugs that cause malabsorption of fat (discussed earlier) also causes carbohydrate malabsorption. Fructose may be malabsorbed if ingested in high concentrations, particularly if it is ingested alone and not as a component of sucrose. [271], [272] Primary fructose malabsorption also has been documented secondary to defects in the GLUT 5 transport system. [273] Toddler's diarrhea in children may be secondary to drinking large amounts of fructose-containing fruit juice, [270], [274] and colic in infants has been postulated to be caused by carbohydrate malabsorption. [275] Occasional adult diarrhea also appears to be related to ingestion of large volumes of fruit juice or soft drinks that are sweetened with fructose-containing corn syrup.

Glucose-Galactose Malabsorption and Disaccharidase Deficiencies Congenital absence of enterocyte brush border carbohydrate hydrolases and transport proteins may cause diarrheas because of various disaccharidase deficiencies: lactase, sucrase-isomaltase, and trehalase. These are discussed in more detail in Chapter 77. Lactose intolerance usually presents in childhood or adolescence, but it may not be recognized in adults. [276] The high-risk groups for lactase deficiency include Asians and Native Americans (90% prevalence), African Americans, Jews, Hispanics, and southern Europeans (60%–70% prevalence). However, lactase deficiency should be considered in cases of unexplained watery diarrhea, especially if accompanied by abdominal cramps, bloating, and flatus, even in people not considered to be among the high-risk groups, [277], [278] because a 10% to 15% prevalence of lactase deficiency can be expected in northern or western Europeans and their American descendants. A trial of a lactose-free diet, a breath hydrogen test, or a lactose absorption test may be diagnostic. Disaccharidase deficiency can occur secondary to intestinal insults such as IBD or celiac disease and can last for months. [279], [280] Some patients with lactase deficiency are misdiagnosed as having IBS. [278], [281] Conversely, many truly lactase-deficient patients attribute intestinal symptoms to lactose intolerance when, in fact, they can tolerate reasonable amounts of lactose and probably do have IBS. [281], [282] Patients with low trehalase activity report abdominal symptoms on ingestion of mushrooms, which contain high levels of trehalose. [283]

Rapid Intestinal Transit As much as 50 g of a normal 200-g carbohydrate diet may be unabsorbed by the normal small intestine and passed into the colon, where it is metabolized by colonic flora. [284], [285] Diets high in carbohydrate and low in fat may cause more carbohydrate malabsorption and osmotic diarrhea because the low fat content allows rapid gastric emptying and rapid small intestine motility. A primary intestinal motility abnormality in which the migrating motor complex is not disrupted by eating resulting in a continuation of the propagative motility pattern may be the cause of carbohydrate malabsorption and osmotic diarrhea in some children with toddler's diarrhea. [286] Similar abnormalities of the migrating motor complex have been reported in patients with the painless diarrhea variant of IBS, and a rapid orocecal transit time has been demonstrated. [287], [288] Carbohydrate wastage from rapid transit may be part of the pathophysiology of diarrhea in thyrotoxicosis [289] and ulcerative colitis. [290] Because carbohydrate is metabolized to H_2 and CO_2 by colonic bacteria, the symptoms of excess flatus, abdominal bloating, and cramping abdominal pain may be important clues to the diagnosis of carbohydrate malabsorption.

Prior Surgery

Bile Acid Diarrhea Three types of bile acid–induced diarrhea are proposed type 1, which results from severe disease, resection, or bypass of the distal ileum; type 2, or primary bile acid malabsorption (see Chapter 77); and type 3, in which bile acid malabsorption follows upper abdominal surgery, either truncal vagotomy or cholecystectomy. [291] Ileal disease, resection, or bypass (e.g., because of Crohn's disease or postoperative adhesions) allows dihydroxy bile salts to escape absorption. If concentrations higher than 2 mmol are attained in the colon, intestinal secretion and diarrhea ensue. [36] Because fasting prevents gallbladder contraction, so large boluses of bile do not enter the intestine, type 1 bile acid diarrhea commonly disappears on fast. This form of diarrhea can be recognized by the history of previous ileal surgery or the presence of ileal disease. Bile acid diarrhea must be differentiated from fatty acid diarrhea, which occurs if ileal disease or resection involves such a large segment of ileum (>100 cm) that hepatic synthesis cannot maintain an adequate intralumenal bile salt pool. [292], [293] Under these circumstances, steatorrhea ensues, and fatty acid–induced intestinal secretion complicates the picture. It is important to differentiate these two related syndromes because bile acid diarrhea responds to bile salt binders such as cholestyramine, but the diarrhea of fatty acid malabsorption does not and may worsen with such therapy. Therapy for fatty acid diarrhea is a low-fat diet that is supplemented with medium-chain triglycerides to prevent severe weight loss. Bile acid malabsorption has been associated with active or previous infections of the ileum, [153], [294] after radiation treatment, [295] and with motility disturbance. [296], [297] Type 2 bile acid diarrhea, or primary bile acid malabsorption, may be congenital or acquired. The acquired variety is described as a disease of excess bile acid loss responsive to cholestyramine, but not associated with other types of ileal dysfunction. [298] It may occur as the result of absence of bile acid receptors or transport proteins (see Chapter 77) similar to findings in congenital type 2 bile acid diarrhea. [299] Patients have been found with documented bile acid malabsorption who have [300] or do not have [301] histological abnormalities of the terminal ileum, including subtotal villus atrophy and crypt hyperplasia. Some investigators believe that this is a common cause of diarrhea-predominant IBS, and some of these patients clearly have cholestyramine-responsive diarrhea. [154], [155], [302] There are also well-studied patients with bile acid malabsorption and normal stool fat excretion; however, the diarrhea of these patients did not respond to cholestyramine. [295], [296] This appears to be a form of idiopathic diarrhea in which bile acids are malabsorbed, but the diarrhea may not be caused by the bile acids. Measured increases in fecal bile acids in patients with postcholecystectomy diarrhea suggest that it is one of the type 3 bile acid malabsorption syndromes. [303], [304] and [305] It is unclear why interruption of gallbladder storage would lead to increased bile acid wastage. Although many patients respond to cholestyramine, some do not, raising the question whether other pathophysiological mechanisms are involved in this form of diarrhea.

Postvagotomy Diarrhea Truncal vagotomy combined with some type of drainage procedure was previously the most common operation for peptic ulcer disease. It is accompanied by diarrhea in 20% to 30% of patients. [306], [307] Because the incidence of diarrhea is much less after selective or superselective vagotomy, a vagus-mediated discoordination of gastric motility, intestinal secretion, or intestinal absorption may be involved. [308], [309] The idea that bile acids play an important role in the diarrhea accounts for its classification as a type of bile acid diarrhea. [308] The treatment for this condition is not always rewarding. Motility-altering drugs (opiates and anticholinergics) or cholestyramine may benefit some patients. [308], [310] In addition, celiac sprue may make its first appearance after gastric surgery or vagotomy; it is a diagnosis that is treatable and should not be missed. [311]

Functional Watery Diarrheas (Irritable Bowel Syndrome)

Among patients defined as having IBS, a few (approximately 25%) have a predominant symptom complex of painless diarrhea. [312], [313] Over the years, the size of this category of IBS becomes smaller as new conditions are discovered such as occult lactose intolerance, collagenous or microscopic/lymphocytic colitis, primary fructose malabsorption, rapid transit with carbohydrate-wasting diarrhea, primary bile acid malabsorption (type 2), food hypersensitivities, and postinfectious diarrhea-IBS. Adult celiac disease is now added to the list; it has been shown to occur in 10% of patients with IBS who meet the Rome criteria for diagnosis. [232] All these examples should give the clinician pause before attributing the painless diarrhea variant of IBS to any psychosocial cause, particularly in men, in whom IBS is

rare in the first place. Perhaps such patients are better labeled as having idiopathic chronic diarrhea.

TRUE SECRETORY DIARRHEAS

Endocrine Tumor Diarrheas

Carcinoid Syndrome Patients with metastatic carcinoid tumors of the gastrointestinal tract or, rarely, primary nonmetastatic carcinoid tumors of the bronchial epithelium, may experience a syndrome that includes the following: watery diarrhea; cramping abdominal pain with borborygmus; episodic flushing; skin changes including telangiectasia, cyanosis, and pellagra-like skin lesions; bronchospasm with asthma attacks and dyspnea; and cardiac murmurs, usually related to right-sided valvular lesions. [314], [315] and [316] The symptoms are caused by secretion of 5-HT, histamine, catecholamines, kinins, prostaglandin, and tachykinins (e.g., substance P) by the tumor mass. All these agents, excluding catecholamines, are potent intestinal secretagogues (see Chapter 14). Up to one third of these patients do not report flushing episodes, and the pellagra-like skin changes and heart murmurs may take some time to develop to clinical appearance. Therefore, this disease should be considered in patients with secretory diarrhea, even if the patient does not have the classic history or physical examination findings.
Gastrinoma Zollinger-Ellison syndrome develops from sporadic, gastric-producing tumors, except in about 20% of cases, when it is part of multiple endocrine neoplasia syndrome 1 (MEN1). [317] Although 70% to 90% of patients with Zollinger-Ellison syndrome present with pain and develop peptic ulcers at some time during the course of their disease, diarrhea also occurs in 25% to 75% of patients and may precede the ulcer symptoms. [317] Furthermore, in 10% of patients, diarrhea may be the major pathophysiological manifestation of the disease. The diarrhea is not strictly an intestinal secretory diarrhea. [318] It is caused in part by high volumes of hydrochloric acid (HCl) secretion, and it can be reduced by nasogastric aspiration or effective antisecretory therapy. [319] Maldigestion of fat owing to inactivation of pancreatic lipase and precipitation of bile acids because of the low pH may also play a role. Because Zollinger-Ellison syndrome is the most common of the neuroendocrine tumors, it must be definitively ruled out as a cause of secretory diarrhea. Rarely, this syndrome can develop from non–small cell lung cancer [320] or ovarian mucinous cystadenomas. [321]
Vipoma or Watery Diarrhea-Hypokalemia-Achlorhydria Syndrome Non–beta-cell pancreatic adenomas (pancreatic endocrine tumors) secrete a host of peptides, including VIP, pancreatic polypeptide, peptide histidine isoleucine, and occasionally secretin, gastrin inhibitory polypeptide, neurotensin, calcitonin, and prostaglandins. [322], [323] Of these, only VIP has been found to be elevated in virtually all patients with watery diarrhea-hypokalemia-achlorhydria (WDHA) syndrome. Because infusions of this hormone can produce all the symptoms, it seems likely that VIP is the primary mediator of this syndrome. Vipoma may therefore be a reasonable and perhaps a more descriptive name than pancreatic cholera or WDHA syndrome. Patients with this tumor have secretory diarrhea, with 70% of patients having more than 3 L of stool per day and virtually all having more than 700 mL/d. Diarrhea with 10 to 20 L of stool per 24 hours has been reported. With high levels of circulating VIP, all segments of the intestine may secrete Na^+, K^+, Cl^-, and HCO_3^-, as well as water, thus accounting for the dehydration, hypokalemia, and acidosis that may accompany this disease. [324], [325] Abdominal pain is not an important symptom of this disease. Patients exhibit flushing (20%) and hypercalcemia (without hyperparathyroidism) occurs in more than 70% of patients, probably caused by the tumor release of neuroendocrine products. Other features of the syndrome include achlorhydria, hypokalemia, hypomagnesemia, enlarged gallbladder, hypokalemic myopathy or nephropathy, hyperglycemia, and lacrimal gland hypersecretion (tearing). In the pediatric age group, vipomas may present as neural crest (sympathetic chain) tumors—ganglioneuromas, neuroblastomas, neurofibromas, and pheochromocytomas. Some of these tumors secrete VIP and produce secretory diarrheas that resolve after the tumor is removed. [326], [327] A rare patient has pancreatic islet tumors and watery diarrhea with normal VIP levels, suggesting that some other peptide, such as pancreatic polypeptide, neurotensin, calcitonin, or prostaglandins, may cause the intestinal secretion. [326]
Medullary Carcinoma of the Thyroid This cancer may present in sporadic form or, in 25% to 50% of patients, as part of MEN2 with pheochromocytomas and hyperparathyroidism. [328], [329] MEN2 is caused by activation of the cellular oncogene *RET,* and mutations of these gene are found in a subset of patients with sporadic medullary carcinoma. [330] Watery (secretory) diarrhea is a prominent part of the syndrome. The diarrhea is thought to be caused by the secretion of calcitonin by the tumor; however, these tumors also elaborate other secretagogues, such as prostaglandins, VIP, substance P, and sometimes 5-HT or kallikrein. [331] Although studies in some patients have shown small intestine secretion, [331] others have shown severely shortened colonic transit time. [332] Therefore, the pathophysiology in this disease may not always be a straightforward secretory one. Usually, by the time watery diarrhea occurs (30% of cases), it indicates metastasis with poor prognosis.
Glucagonoma Patients with glucagon-secreting pancreatic islet tumors present with diabetes (90%), a form of eczematous skin rash called *migratory necrolytic erythema,* and, occasionally, glossitis, cheilitis, mild diarrhea (25%), psychiatric or neurological aberrations, and thromboembolic propensities. [333] The cause of the diarrhea in these patients is unclear.

Nonendocrine Malignant Diseases

Villous Adenomas Villous adenomas of the rectum or rectosigmoid may cause a secretory form of diarrhea with K^+ loss. [334], [335] and [336] Diarrhea in the range of 500 to 3000 mL/24 h has been recorded. Tumors that are capable of causing such secretory diarrhea are usually large—more than 3 to 4 cm in diameter and often as large as 10 to 12 cm. [336] Although the cause of the secretion may be intrinsic to the nature of this neoplastic epithelium, secretagogues such as prostaglandins have been found in both the tumor and rectal effluent of such patients, [337], [338] and [339] and indomethacin administration reduces the diarrhea in some patients. [338], [340]
Systemic Mastocytosis If mast cell proliferation is limited to the skin, it is termed *urticaria pigmentosa*. [217] If it involves the bones, liver, spleen, lymph nodes, and gastrointestinal tract, it is known as *systemic mastocytosis.* The diarrhea of systemic mastocytosis may be continuous and accompanied by steatorrhea secondary to infiltration of the mucosa and the resulting villus atrophy. [217], [218] However, the diarrhea may be intermittent and may be associated with flushing, tachycardia, hypotension, and, occasionally, headache, cognitive disorders, nausea and vomiting, peptic ulcers, syncope, itching, and urticaria, which may be provoked by alcohol ingestion. [217] In this form of the syndrome, histamine or another mast cell mediators such as prostaglandin D_2 may be the secretagogue responsible, by either stimulating gastric acid secretion (much as in Zollinger-Ellison syndrome) or having a secretory effect on the intestine. Antihistaminics (H_1 blockers), H_2 blockers or proton pump inhibitors, cyclooxygenase inhibitors, and disodium cromoglycate may be helpful in treatment. [217], [218], [341] Blockade of mast cell mediator receptor or of mast cell degranulation may reduce all these symptoms and the diarrhea, but not the steatorrhea, which may be better treated with corticosteroids. [217], [218], [341]

Factitious Diarrhea

Approximately 15% of patients referred to secondary or tertiary centers for diarrhea [342] and 25% of patients with proven secretory diarrheas [343], [344], [345], [346], [347] and [348] are found to be surreptitiously ingesting either laxatives or diuretics. These patients present with severe chronic watery diarrhea, often with abdominal pain, weight loss, nausea, and vomiting, sometimes with hypokalemic myopathy and acidosis. Occasionally, they have severe protein-losing enteropathy as well. They have 10 to 20 bowel movements per day, with 24-hour stool volumes in the range of 300 to 3000 mL, and may have nocturnal diarrhea as well. The most common drug causing this syndrome in the United States is probably bisacodyl. [349] Anthraquinones, which include senna, cascara, aloe, rhubarb, frangula, and danthron, are other abused laxatives. [348], [350] Osmotic laxatives such as Na_2SO_4, Na_2PO_4, $MgSO_4$, and magnesium citrate are occasionally used. [347], [348] Because there is no readily available assay for dioctyl sodium sulfosuccinate (the docusate salts), one of the more common laxatives, its frequency of use in this syndrome is uncertain. Some patients ingest large quantities of diuretics.

More than 90% of these patients are women. It is unclear why they ingest these drugs to the point of requiring hospitalization. There appear to be two different clinical syndromes: [345], [351]

1. Women younger than 30 years of age in whom some elements of eating disorders (e.g., anorexia nervosa and bulimia) appear to be part of the psychic abnormality
2. Middle-aged to elderly women with histories of extensive medical care who seem to gain some kind of secondary benefit from the sick role and the attendant personal attention

Many of these latter patients are health care workers, such as nurse's aides, and this is particularly true among the few men who present with this syndrome. On confrontation, patients may either deny the drug ingestion and leave the physician's care or admit the aberrant behavior and submit to psychiatric care. After laxatives are discontinued, these patients develop edema as a result of secondary hyperaldosteronism or pseudo-Bartter syndrome, [352] but it subsides spontaneously within 1 to 2 months if left untreated. Another complication of chronic laxative abuse is the development of ammonium urate renal calculi, presumably secondary also to the dehydration and acidosis of severe diarrhea. [353]

This syndrome also exists in pediatrics, where it has been called *Munchausen syndrome by proxy* or *Polle syndrome*. [354], [355] (Polle was Baron von Munchausen's son, who died at an early age of unknown causes.) In these circumstances, it is a form of child abuse in which the guilty parent, usually the mother, again derives some

kind of secondary gain from the extensive hospitalizations and evaluations of the child. The frequency of this factitious disorder as a cause of severe diarrhea is high enough to warrant laxative screening to rule out this syndrome before initiating extensive medical evaluation for the other causes of diarrhea.

Chronic Idiopathic Diarrhea and Pseudopancreatic Cholera Syndrome

Patients in whom extensive evaluation for a cause of secretory diarrhea is negative, including a search for hormone-secreting tumors and laxative and drug ingestion, are said to have either *chronic idiopathic diarrhea* or *pseudopancreatic cholera syndrome,* depending on whether the fasting stool volumes are less than or greater than 700 mL/24 h, respectively. [356], [357] Some of the patients with this syndrome are found to have microscopic/lymphocytic colitis, Brainerd diarrhea, or type 2 bile acid diarrhea variant. Others could be ingesting laxatives or drugs for which assays are not readily available. These cases defy diagnosis, and, importantly, these patients outnumber those with neuroendocrine secretory tumors. If no diagnosis is revealed, symptomatic therapy with bile salt–binding drugs, opiates, or anticholinergic medications may be tried. Follow-up studies suggest that in most of these patients, the diarrhea is self-limited and disappears spontaneously in 6 to 24 months. [357] One current hypothesis is that these patients may be suffering from severe, postinfectious IBS, that is, Brainerd diarrhea (see section “ Prolonged Infectious Diarrheas”).

Diabetic Diarrhea

The most common gastrointestinal symptom in patients with diabetes of either type is constipation (25% of patients) and not diarrhea (2.5%–3.7%). [358], [359] Nevertheless, up to 20% of young to middle-aged people with type 1 diabetes that has been poorly controlled for more than 5 years (particularly men between 20 and 40 years of age), may have profuse watery, urgent diarrhea, often occurring at night with incontinence. [360], [361] These patients usually have severe neuropathy and often have both nephropathy and retinopathy. Some patients have exocrine pancreatic insufficiency or bacterial overgrowth secondary to the motility disturbances of the autonomic neuropathy. Because patients with type 1 diabetes may have concomitant celiac disease, [227] the single most important test in these patients, at least in terms of diagnosing a potentially treatable disease, is the AEA or anti-tTG serology tests. The most common cause of “diabetic” diarrhea in those with type 2 disease is therapy with metformin. [358]

Unfortunately, most diabetic patients with severe diarrhea do not have a treatable cause of steatorrhea, and the cause of their diarrheal condition is unknown. Animal studies suggest that diabetes induces specific sympathetic denervation of the bowel. [362], [363] This leaves unopposed cholinergic tone, which impairs fluid and electrolyte absorption or actually stimulates frank intestinal secretion. It is on this basis that clonidine, a specific a $_2$-adrenergic agent, has been recommended as a treatment for diabetic diarrhea. [364] Diarrhea may improve when patients take this drug, although it is uncertain whether this response is the result of the nonspecific antisecretory effect of a $_2$-adrenergic agents or whether the response confirms the proposed sympathic denervation. These patients with neuropathy frequently have impaired anal sphincter function, and this contributes to their incontinence. [358], [365] Octreotide therapy may be helpful. [366]

Alcoholic Diarrhea

Binge drinking of alcohol causes a brief episode of diarrhea that usually lasts less than 1 day. This may result from acute damage to both the microvasculature and the epithelium [367], [368] and [369] and is accompanied by alterations in water, electrolyte, and nutrient absorption. [370]

Patients with chronic alcoholism often have severe watery diarrhea that persists for days or even weeks after hospitalization. Physiological abnormalities described in patients with alcoholism include more rapid oral-cecal and colonic transit, [371], [372] decreased intestinal disaccharidases, decreased bile secretion (particularly in those with cirrhosis), and decreased pancreatic secretion. [351] Folate or vitamin B $_{12}$ deficiency or protein malnourishment also may play a role. With abstinence, renourishment, and replenishment of vitamin deficiencies, most patients’ diarrhea slowly improves.

Congenital and Neonatal Diarrheas

Several causes of congenital diarrhea have been documented: a congenital short bowel syndrome [373]; a primary form of ileal dysfunction with bile acid malabsorption, also called *familial microvillus atrophy* [299], [300], [374]; congenital insulin-dependent diabetes mellitus with secretory diarrhea [375]; congenital chloridorrhea [376], [377] and [378]; congenital Na diarrhea [379], [380] and [381]; microvillus inclusion disease [382], [383]; congenital enterocyte heparan sulphate deficiency [384]; “tufting” enteropathy and epithelial dysplasia [385]; mitochondrial encephalomyelopathies with diarrhea, immune dysregulation, polyendocrinopathy, enteropathy, X-linked syndrome (IPEX) [386]; and Satoyoshi syndrome. [387] Some of these diseases are discussed in more detail in Chapter 77.

INFLAMMATORY DIARRHEAS

Inflammatory Bowel Disease

Patients with either Crohn’s disease of the small or large intestine or ulcerative colitis have diarrhea with stool volumes usually less than 1 L/24 h that frequently, but not always, improve with fasting. Decreased Na^+, Cl^-, and water absorption or frank secretion can be demonstrated in both the small intestine and colon of patients with IBD. [26], [27] and [28], [45], [388] Patients with severe ulcerative colitis may have water and electrolyte secretion in the unaffected small intestine, suggesting the presence of circulating secretagogues. [388] The abnormalities of transport are caused by inflammatory mediators, such as histamine, prostaglandins, leukotrienes, platelet-activating factor, and cytokines (interleukins), that are released from mast cells, phagocytes, and mesenchymal cells (see section “ Pathophysiology of Inflammatory Diarrheas”). The abnormalities of electrolyte transport brought about by these secretagogues are further enhanced by a damaged absorptive surface epithelium and even denuded mucosa with leakage of plasma or blood into the lumen. Effective treatment of IBD with salicylates, steroids, antibiotics, antiinflammatory drugs, and immune mediating drugs is accompanied by reductions in the levels of inflammatory mediators, improved histology, and reduction in diarrhea. [389]

Eosinophilic Gastroenteritis

Infiltration of the gastrointestinal tract of either adults or children with eosinophils is a recognized clinical entity that is accompanied by diarrhea. [390], [391] and [392] Diarrhea occurs in 30% to 60% of patients with eosinophilic enteritis regardless of whether the eosinophils are infiltrating the mucosa, the muscle, or the serosal layers of the gut. [392] Peripheral eosinophilia is present in 75% of these patients. The disease may involve the entire gastrointestinal tract from esophagus to anus, or it may be isolated to the colon. Abdominal pain, nausea, vomiting, and weight loss are other prominent symptoms of this disease. Steatorrhea and protein-losing enteropathy are present in 10% to 30% of these patients. A few patients with peripheral eosinophilia, but no evidence of gastrointestinal infiltration, appear to have symptoms similar to those with gastrointestinal involvement. [392] The cause of this disease is unknown, but approximately 50% of patients have atopic (allergic) histories. Food allergy is suspected in these patients, but elimination diets are only occasionally successful. [391] Parasites, particularly *Strongyloides,* should be rigorously ruled out before making a diagnosis of eosinophilic gastroenteritis. [393] Steroids remain the mainstay of therapy. [390], [391] and [392] Sodium cromoglycate may be useful. [394]

Milk and Soy Protein Allergy

Intolerance to cow’s milk and soy protein is a well-established cause of enterocolitis in infants. [395], [396] and [397] The disease involves both the small intestine and the colon and may present within the first 6 months of life with either acute or gradual onset of vomiting and diarrhea, occasionally with bloody stools caused by ulcerative proctocolitis. Approximately 50% of the patients who are allergic to one of these proteins are also allergic to the other. In older children, milk allergy can present with constipation. [398]

Food Allergy

Dietary hypersensitivity is clearly recognized in infants and adults (see earlier), and systemic anaphylaxis has long been recognized in association with peanuts and seafood ingestion. [399] Diarrhea, constipation, and gastroesophageal reflux as a result of food allergy are increasingly recognized. [400], [401] (see Chapter 129).

Commonly suspected allergens include milk, eggs, seafood, nuts, artificial flavors, and food coloring. [399], [400]

Microscopic Colitides

Collagenous colitis and lymphocytic colitis are two diseases grouped together as atypical or microscopic colitides. [402], [403] and [404] Microscopic or lymphocytic colitis seems to be equally prevalent in men and women, whereas collagenous colitis occurs 10 times more often in middle-aged or elderly women. These diseases are categorized as either inflammatory diarrheas, because intraepithelial lymphocytes and lamina propria lymphocytes are prominent, [405] or as secretory diarrheas, because intestinal secretion is present in the disease. [406] In many cases, the secretory process is mild, and diarrhea stool volumes may return to normal with fasting. [356] Other patients with more severe diarrhea continue to have elevated stool volumes on fasting. It is now clear that many patients (perhaps 15%) with lymphocytic colitis have celiac sprue. [407] Increased lumenal prostaglandin levels suggest that this inflammatory mediator is being released by the subepithelial immune cells and is causing the diarrhea in this disease. [406] An epidemiologic relationship with long-term NSAID use also has been reported. [408] In addition to gluten sensitivity, bile has been proposed as the trigger for such prostaglandin release. Sensitivity to these agents may be the reason that symptoms disappear with fecal stream diversion. [409] Bismuth subsalicylate, 5-aminosalicylates, steroids, immunosuppressants, octreotide, calcium channel blockers, diverting ileostomy, and gluten-free diet have all been successful as therapies. [402], [403], [404] and [405], [407]

Chronic watery diarrhea also occurs in patients whose small intestine and colonic biopsies reveal microscopic eosinophilic infiltration in the crypt region with a normal surface or villous epithelium. Such patients have been labeled as having *pericrypt eosinophilic enterocolitis*. [410] Fifty percent of these patients have a collagen-vascular disease. The diarrhea responds well to corticosteroids.

Protein-Losing Enteropathy

Severe protein loss through the gastrointestinal tract occurs in a variety of disease states: [411], [412], [413] and [414]

Infection: *C difficile* infection, *Salmonella* infection, enterocolitis, shigellosis, viral gastroenteritis, parasite infestation, bacterial overgrowth, Whipple disease
Diseases with mucosal erosion or ulcerations: gastritis, gastric cancer, collagenous colitis, IBD
Diseases marked by lymphatic obstruction: congenital intestinal lymphangiectasia, sarcoidosis, lymphoma, mesenteric tuberculosis, as a sequela of surgical correction of congenital heart disease with Fontan operation, long-term peritoneal dialysis
Mucosal diseases without ulceration: Ménétrier disease, sprue, eosinophilic gastroenteritis, amyloidosis
Immune diseases: systemic lupus erythematosus or food allergies, primarily to milk

Although the protein losses caused by ulceration with denudation of mucosal capillaries and lymphatics and resulting from obstructed lymphatics are easy to comprehend, new knowledge about the immune vascular injury in other sites (e.g., lung, kidney) allows a better appreciation of this phenomenon in the gastrointestinal tract. The condition usually responds to corticosteroids or other antiimmune therapy.

Chronic Radiation Enterocolitis

Although acute radiation diarrhea is common (see previous discussion), patients receiving pelvic radiation for malignant diseases of the female urogenital tract or the male prostate may develop chronic radiation enterocolitis 6 to 12 months after total doses of radiation greater than 4 to 6 Gy (see Chapter 132). The terminal ileum, cecum, and rectosigmoid are the segments usually involved because they are fixed in the pelvis and therefore may receive the full brunt of the weekly radiation dosages. The histology is one of obliterative arteritis, occasionally with lymphangiectasia, partial villus atrophy, fibrosis, and strictures. With time, severely bleeding rectal telangiectasias develop in many patients. The diarrhea may be caused by bile acid malabsorption if the ileum is involved, by bacterial overgrowth if small intestine strictures occur, or by chronic inflammation of the small intestine and colon. [415] Antiinflammatory drugs such as sulfasalazine and corticosteroids have been tried with little success; occasionally, cholestyramine and NSAIDs may help, as may opiate antidiarrheal medications. The bleeding rectal telangiectasia may respond to electrocoagulation, laser ablation, or intrarectal formalin [416] (see also Chapter 132).

Miscellaneous Diseases

Although *acute mesenteric arterial* or *venous thrombosis* presents as acute bloody diarrhea, *chronic mesenteric vascular ischemia* may present as watery diarrhea with spotty endoscopic inflammation (see Chapter 131) that can be mistaken for a symptom of IBD. Chronic infections including gastrointestinal *tuberculosis* [417] and *histoplasmosis* [418] present with diarrhea that may either be bloody or have characteristics of a secretory process. Immunologic diseases such as *Behçet disease* [419] may have diarrhea as a symptom. There seems to be geographic heterogeneity among these patients, with those in Japan having more gastrointestinal involvement than those in Turkey or Israel. [420] *Churg-Strauss syndrome* may present with diarrhea. [421] Diarrhea is the hallmark of acute *graft versus host disease* after allogeneic bone marrow transplantation. [422] The triad of dermatitis, hepatic cholestasis, and enteritis with diarrhea define this disease, and the volume of diarrhea has even been proposed as part of clinical staging. *Neutropenic enterocolitis* is ileocolitis occurring in neutropenic patients with leukemia. Some cases of this may be caused by *C difficile* infection. [423] The *Cronkhite-Canada syndrome,* usually listed under the polyposes because of the characteristic retention (inflammatory) polyps, has most of the hallmarks of an immune disorder, and severe gastrointestinal protein loss and diarrhea are present. [424], [425] Long-term NSAID use can lead to colonic ulcers (*NSAID colitis*) with diarrhea or right-sided colonic weblike strictures (*diaphragm disease*). [426]

CLINICAL EVALUATION OF CHRONIC DIARRHEA

The goal of the gastroenterologist in evaluating a patient with chronic diarrhea is to make a definitive diagnosis as quickly and inexpensively as possible. This section outlines an approach to diagnosis of adult patients; the diagnostic approach in infants and children may be different. [211], [427] Experienced clinicians [189], [190], [428], [429] and [430] suggest that 75% to 80% of chronic diarrheas can be diagnosed by an expert history and physical examination, coupled with certain screening and focused laboratory examinations (Fig. 42-7). This expert opinion is backed by clinical studies. [431], [432] The remaining 25% of patients with severe or elusive diarrhea may need hospitalization and extensive testing. Three tests—quantitative stool fat, colonoscopy with biopsy, and the response to fasting with measurement of stool volume and osmotic gap—lead to a definitive diagnosis in most of these remaining patients. [189], [190], [428], [429] and [430]

FIGURE 42-7. Approach to the evaluation of chronic diarrheal disease.

Malabsorption

History and Physical Examination The causes of generalized malabsorption are outlined in Table 42-2, and the signs and symptoms in Table 42-10, Table 42-11 and Table 42-12. [189], [190], [191] and [192], [433] The approach to evaluation is shown in Figure 42-8. Mild degrees of malabsorption may be entirely asymptomatic and may not result in the classic gastrointestinal manifestations of flatulence, bulky or greasy foul-smelling stools, and weight loss. For these reasons, malabsorption sometimes presents as an aberration of one of the other body systems (see Table 42-10 and Table 42-11).

TABLE 42-11 Correlation of Clinical Manifestions, Pathophysiology, and Laboratory Findings in Malabsorptive Processes

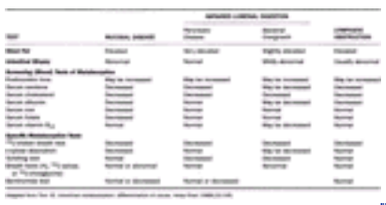

TABLE 42-12 Comparison of Laboratory Results in the Three Types of Malabsorption: Mucosal Disease, Impaired Intralumenal Digestion, and Postmucosal or Lymphatic Obstruction

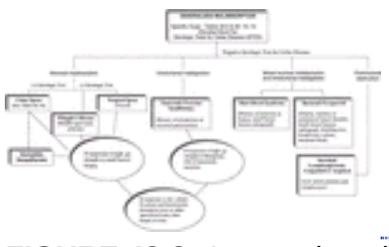

FIGURE 42-8. Approach to the evaluation of malabsorption.

Clues that help differentiate clinically severe mucosal disease, such as celiac sprue or Whipple disease, from the malabsorption caused by exocrine pancreatic insufficiency are the presence of cytokine-induced anorexia, lethargy, and malaise as well as extraintestinal signs and symptoms in patients with intestinal mucosal disease. Severe abdominal pain from repeated bouts of pancreatitis may be a useful clue to pancreatic insufficiency. The character of the stool may be a useful differentiating symptom. The fat content of the stool in pancreatic insufficiency is considerably higher than in mucosal disease (see Table 42-12). [434] At body temperature, the fat may be present as oil, and the oil may separate from the stool, manifesting as oily seepage from the anus or as oil droplets floating in the toilet bowl after passage of the steatorrheic stool. Patients with pancreatic insufficiency rarely have flatulence and bloating, perhaps because salivary or gastric amylase ameliorates somewhat the carbohydrate malabsorption. Stools tend to float and are difficult to flush in malabsorptive diseases as a result of the gas content, which is caused by carbohydrate malabsorption and subsequent fermentation and gas formation, and not fat malabsorption. [435] Total serum protein concentrations are usually lower in intestinal mucosal diseases, most likely because of concomitant protein-losing enteropathy, and anemia is more common because of both iron and folate malabsorption. The understanding of an association of certain diseases or syndromes with subclinical celiac disease (see Table 42-10 and section “Celiac Sprue”) requires knowledge of those diseases and conditions in which screening for celiac disease is appropriate.

Screening Tests for Malabsorption

Blood tests. Tests that may yield important clues to the presence of significant malabsorption include a complete blood count, prothrombin time, serum protein determination, and alkaline phosphatase (see Table 40-12). These may signal the presence of iron, folate, vitamin B_{12}, vitamin K, or severe vitamin D malabsorption with osteomalacia. Serum carotene, cholesterol, albumin, serum iron, folate, and vitamin B_{12} determinations give additional clues to the presence of malabsorption, and these usually point to intestinal mucosal disease. Serum carotene may be low simply from poor intake, but not usually less than 50 µg/dL, as is commonly seen in severe malabsorption.

Serologic tests for celiac disease. Patients suspected of having celiac disease, either because of signs and symptoms or malabsorption or resulting malnutrition or because of concomitant diseases in which there is a high incidence of celiac disease (see Table 42-10 and Chapter 76), should be screened with serologic tests for celiac disease. AEA tests are highly sensitive (90%) and specific (90%–95%). [221, 222, 223] and [224, 231] Simple, accurate tests for anti-tTG antibodies (both IgA and IgE subclass) using guinea pig or human tissue transglutaminase are being developed that are even more sensitive and specific (95%–100%) and will diagnose the disease in patients who are IgA deficient, a not uncommon concomitant condition in those with celiac disease. [222, 223, 225] Screening with AEA testing alone can underestimate celiac disease by as much as 11%. [222] Use of any IgA test alone will fail to diagnose those with IgA deficiency. Patients with Down syndrome and Sjögren syndrome may have false positive test results. [231]

Radiography (Malabsorption) Radiology should be viewed as an adjunct to the diagnosis of malabsorption and not as a primary test. A flat plate of the abdomen may demonstrate pancreatic calcification. Pancreatic ultrasound, computed tomography, and endoscopic retrograde cholangiopancreatography can be used if necessary to confirm the presence of pancreatic disease as a cause of malabsorption. Small intestine disease also can be revealed by barium contrast radiographs, [436] either an upper gastrointestinal series with small intestine follow-through or, preferably, an enteroclysis examination (see Chapter 152). Previous gastric surgery, gastrocolic fistulae, blind loops from previous intestinal anastomoses, small intestine strictures, multiple jejunal diverticula, and abnormal intestinal motility that could lead to bacterial overgrowth may be demonstrated. Certain diseases may present radiographically as uniform thickening of the valvulae conniventes (e.g., amyloidosis, lymphoma, Whipple disease); uniform or patchy diseases such as lymphoma or lymphangiectasia also may be seen. Patients with sprue show dilation of the small intestine with little mucosal abnormality. Segmentation of the barium column also occurs in sprue or in any of the malabsorptive or severe secretory diarrheas if there is sufficient intralumenal fluid to cause precipitation or flocculation of the barium. Endoscopic ultrasound has been reported to be useful in raising the suspicion of celiac disease. [437]

Upper Gastrointestinal Endoscopy and Biopsy Upper gastrointestinal endoscopy with distal duodenal biopsy should be undertaken if diagnostic clues or positive celiac disease serology suggest small bowel mucosal malabsorption, especially in patients with proven steatorrhea (see later). Even a negative biopsy result can be helpful if it redirects the clinician to the pancreas. Occasionally, inflammatory disease of the upper small bowel is evident on upper endoscopy and biopsy.

Endoscopic appearance. The presence of scalloping or a reduced number of duodenal folds or the presence of a mosaic pattern or nodularity has a positive predictive value in the 95% to 100% range for celiac disease in adults [438] and children. [439] However, other diseases may have similar appearances: eosinophilic enteritis, tropical sprue, giardiasis, human immunodeficiency virus–related enteropathy, graft versus host disease, and amyloidosis. [440, 441] and [442] The addition of magnification endoscopy and chromoendoscopy may be useful in the diagnosis of villous atrophy. [443, 444] Push enteroscopy may be useful in the evaluation of “refractory sprue,” [445] but it does not add much to the evaluation of routine malabsorption. [446]

Duodenal histology. When history, physical examination, and screening tests (especially serologic tests for celiac disease) strongly suggest malabsorption, and certainly when steatorrhea is documented, small intestine biopsy is probably the most cost-effective way to determine whether the steatorrhea is caused by intestinal or pancreatic sources. Endoscopic grasp biopsies are usually as good as small bowel biopsy instruments such as the Rubin-Quentin multipurpose tube, the Crosby-Kugler capsule, or the Watson or Carey capsule for distal duodenal biopsy. [447] The capsule technique is still used in infants because of its safety and the ease of passing it orally in the suckling child. [448, 449] and [450] There are a few diseases in which small bowel biopsy is absolutely diagnostic (Table 42-13). Diseases that have a patchy distribution within the gastrointestinal tract may be missed on small bowel biopsies. Our newer understanding of celiac disease as a mild, occult, or latent disease requires new histological rigor for diagnosis of this disease. [451] The presence, location, and number of intraepithelial lymphocytes are becoming important criteria for the diagnosis of subtle celiac disease, although intraepithelial lymphocytes are also seen in other immune-mediated diseases (e.g., tropical sprue, food allergy, autoimmune enteropathy). [452, 453] Miscellaneous lesions not included in Table 42-13 that may be discovered with small bowel biopsy include benign lymphoid hyperplasia, milk or soy protein hypersensitivity, lipid storage diseases such as Fabry or Niemann-Pick disease, histoplasmosis, capillariasis, cytomegalovirus enteritis, schistosomiasis, and Waldenström macroglobulinemia. [454, 455] and [456]

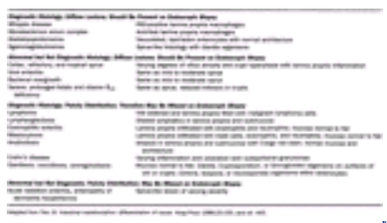

TABLE 42-13 Diagnostic Reliability of Peroral Small Intestine Biopsy

Stool Tests for Malabsorption

Collection and appearance. Early examination of stools for ova and parasites should be considered before beginning 24-hour stool collections or obtaining barium radiographs, although this is not always cost effective. The presence of barium interferes with the ability to find ova or parasites, even 1 to 3 weeks after the procedure. Outpatient qualitative tests on single stool collections and quantitative tests, usually requiring hospitalization and 48- to 72-hour stool collections, can be useful in defining the cause of diarrhea. Stool can be collected in the outpatient area for analysis. The patient can be given preweighed paint cans and toilet bowl inserts to facilitate separation of stool from urine. The stool is transferred from the insert to the can and kept cold in an inexpensive Styrofoam ice chest containing dry ice or one of the refreezable "blue-ice" packs. Collections are made for 48 hours, or better still, for 72 hours. An important adjunct to diagnosing the cause of diarrhea is to look at the stool. The bloody stool of gut inflammation is not easily confused with watery diarrhea. Similarly, the diagnosis of watery diarrhea to the exclusion of steatorrhea can be made with 80% to 95% accuracy. [457] However, patients with steatorrhea sometimes have watery diarrhea, and then stool appearance and weight can be misleading. [457], [458] Thus, it is usually wise to measure stool fat. It is also important to be sure your patient is not taking orlistat for treatment of obesity or consuming large quantities of food that has been fried in olestra, [459] because these agents can cause or simulate steatorrhea.

Qualitative fecal fat tests. The usual intake of fat in the typical North American diet is 100 to 150 g/d, mostly as triglycerides (see Chapter 18). Previously, it was thought that 93% to 97% of this fat was absorbed and that this percentage capability extended up to an intake of 200 g/d. [460] This relationship was recognized not to hold for neonates, whose stool fat may normally exceed 10% of intake. [10] It is now clear that the normal coefficient of absorption of dietary triglycerides is 99%, but for phospholipid of endogenous sources (e.g., bile, sloughed enterocytes, bacteria), it is only 90% or so. Therefore, 5 to 6 g of normal fecal fat is unabsorbed phospholipid from the 40- to 50-g endogenous pool, and 1 g is derived from the diet. [461] Thus, if total stool fat exceeds the normal 7 g/24 h, it is abnormal. Stool fat content exceeding 6 to 7 g/24 h or triglyceride content exceeding 1 g/24 h can be detected by a simple qualitative fecal fat determination with approximately 90% sensitivity and 90% specificity. [462], [463] and [464] The test loses some sensitivity with mild steatorrhea (fecal fat in the range of 6–10 g/24 h). There are also false-negative results if fat intake is inadequate; therefore, it is wise to ensure that the patient is ingesting a normal diet (approximately 100 g fat/24 h). False-positive results can occur if mineral oil laxatives, rectal suppositories (cocoa butter), or orlistat are given to the patient before stool collection or if the patient ingests large quantities of olestra. To perform the qualitative stool fat examination, stool is mixed with water and alcohol and is heated, and then Sudan 3 or 4 stain is added to stain for neutral fat (triglycerides). Fatty acid soaps can be dissociated by mixing glacial acetic acid and Sudan stain with the stool and heating it to boiling. A positive test is recorded if there are more than 100 orange-red globules of fat at least 6 to 75 µm in diameter (bigger than red blood cells) per medium power (40×) field. There is a positive correlation between the number of droplets and the fat content of the stool and a less clear relationship between the size of the droplets and the fat content. [464] The accuracy of the test also depends on the experience of the laboratory personnel, and there seem to be fewer and fewer laboratories that perform this test well. Because of the problems of completely collecting infants' stools, various investigators have studied the validity of the steatocrit test, a gravimetric method that is performed on random stool samples. [465], [466] and [467] The test correlates well with other fecal absorption tests in infants and children and, because of its simplicity, needs to be investigated in adolescents and adults.

Quantitative fecal fat tests. The quantitative fecal fat determination is the standard against which all malabsorption tests are compared. Homogenized stool specimens are analyzed by the techniques of Van de Kamer, [468] which involve the saponification of wet feces with alcohol and sodium hydroxide, liberation of the fatty acids with HCl, extraction with petroleum ether, and then titration of the fatty acids. A spectroscopic method of fat determination has been reported. [469] The major problem with the quantitative fecal fat analysis is ensuring that an adequate stool specimen has been collected. Because a patient's bowel movements may vary from day to day, this is possible only with a 72-hour collection. Even so, fewer than 50% of patients pass more than 90% of ingested stool markers in 3 days. [470] Because of the skills necessary to perform it accurately, the Sudan quantitative test for fecal fat seems to be disappearing from hospital laboratories; the 3-day fecal quantitative fat determination remains the single best test for detection of steatorrhea. Although stool fat excretion for 24 hours may be the same for mucosal and pancreatic disease, mucosal disease usually causes more water malabsorption or outright secretion. [434] The result is stool fat that is excessive but is diluted by the excess fluid. Consequently, the *stool fat concentration* (grams of fat/100 g wet stool weight) is usually less than 9.5% in mucosal diseases such as celiac sprue and greater than 9.5% in chronic pancreatic insufficiency. Other studies have questioned the value of this refinement of the fecal fat test. Although it has been repeatedly confirmed that a fecal fat excretion of more than 7 g/24 h exceeds by more than two standard deviations of the mean normal fat excretion, it is important to realize that diarrhea of secretory or osmotic origin can wash fat from the bowel lumen and can simulate steatorrhea. Therefore, if diarrhea is severe (>800 g stool/24 h), stool fat excretion can be as high as 14 g/24 h. [471] Problems arise because patients with mild forms of true steatorrhea have stool fat excretion in the range of 8 to 14 g/24 h. Obviously, patients with diarrhea whose stool fat excretion falls in this borderline range could have secretory, osmotic, or malabsorptive diarrhea, and these patients need more definitive evaluation.

^{14}C-Triolein Breath Test for Fat Maldigestion/Malabsorption Measurements of the respiratory excretion of labeled CO_2 after oral administration and metabolism of radioactive carbon-labeled substrates, or of H_2 after administration of carbohydrates, can be used to assess fat, carbohydrate, and bile salt malabsorption or bacterial overgrowth. [472] All CO_2 breath tests, both those using ^{14}C and those using the stable isotope ^{13}C, are based on the idea that either intralumenal (bacterial) or cellular (intermediary) processes convert the substrate into CO_2, which is excreted in expired air. The stable isotope (^{13}C) tests require mass spectrometric detection techniques. After enzymatic hydrolysis by pancreatic proteins and micelle formation with bile salts, triglycerides are absorbed within the first 150 cm of jejunum. If a triglyceride such as triolein, labeled with ^{14}C at the carboxyl groups, is given with 20 to 30 g of nonlabeled triolein (to promote biliary emptying), it should be efficiently absorbed and essentially completely metabolized to CO_2 and water, and the $^{14}CO_2$ should appear in the breath. This forms the basis for the ^{14}C-triolein breath test. [473], [474] and [475] In patients with impaired fat absorption, there is a reduction in the $^{14}CO_2$ excreted. Either cumulative or peak values provide discrimination, with 85% to 95% sensitivity and specificity. False-positive results are obtained if there is poor gastric emptying (e.g., in diabetes or after gastric surgery), if there are conditions that lead to dilution of $^{14}CO_2$ (e.g., obesity, hyperlipidemia, and ascites), if there is impaired metabolism (e.g., severe liver disease), or if there is CO_2 retention (e.g., chronic lung disease). False-negative results (increased CO_2 production) can theoretically be seen in fever, starvation, and hyperthyroidism. The amount of radioactivity is approximately the same as that received in a chest film and is confined almost exclusively to the lungs. The usefulness of the test is limited by the false-positive results in healthy persons (20% in IBS) and the false-negative results in patients with mild degrees of fat malabsorption.

D-Xylose Absorption Tests D-Xylose is a five-carbon sugar that is incompletely absorbed in the duodenum and jejunum and is metabolized by intralumenal bacteria, so it can be used as a test of both bacterial overgrowth and mucosal disease [476], [477] Xylose is not completely metabolized; therefore, its excretion into urine or its concentration in blood after ingestion of a standardized dose has been used for more than 30 years as a small intestine absorption test. [477] After an overnight fast, either 5 or 25 g is given orally. Patients with normal renal function therefore excrete more than 4 g after a 25-g dose. The test has poor sensitivity in mild mucosal disease, although sensitivity can be increased by obtaining blood values after the oral dose. A blood level of greater than 25 mg/dL 1 hour after the 25-g dose, or greater than 20 mg/dL 1 hour after the 5-g dose, can be expected. [476] Because the test depends on normal renal function, it is less useful in elderly patients in whom the glomerular filtration rate is reduced, or in patients with known renal disease. Because it can be distributed into sequestered fluids, it is less useful in patients with ascites. Indeed, increased portal pressure alone has been shown to decrease xylose absorption. Furthermore, NSAIDs impair either absorption or urinary excretion. It has false-positive and false-negative rates of 20% to 30%. It should be considered in those mucosal diseases in which a patchy distribution of the histological lesion may invalidate the endoscopic biopsy (see Table 42-13). A D-xylose breath test has been proposed. [478]

Pancreatic Function Tests

Tubeless pancreatic function tests. Pancreatic chymotrypsin cleaves free paraaminobenzoic acid (PABA) from the synthetic peptide *N*-benzoyl-L-tyrosyl-paraaminobenzoic acid; this is the principle behind the *Bentiromide test*. If 500 to 1000 mg is administered orally and normal pancreatic chymotrypsin is present, the free PABA should be absorbed, conjugated in the liver, and excreted in the urine. With severe pancreatic insufficiency, less than 57% of the PABA is excreted in 6 hours. [479], [480], [481] and [482] Unfortunately, severe intestinal mucosal disease, renal disease, diabetes, and severe liver disease all diminish PABA absorption or excretion in the absence of pancreatic insufficiency, and the test is not very sensitive in patients with mild pancreatic insufficiency. Furthermore, several ingested substances, including medications (e.g., acetaminophen, benzocaine, lidocaine [Xylocaine], procaine, sulfa-containing antibiotics, chloramphenicol, chlorothiazide, furosemide) as well as certain foods (prunes and cranberries), interfere with the laboratory determination. [482] To overcome some of these limitations, the *pancreolauryl test* was developed. [479] Fluorescin is conjugated with two molecules of lauric acid and is given orally to be hydrolyzed by pancreatic cholesterolase. The fluorescin release is absorbed and excreted in the urine. It has many of the same limitations as the bentiromide test. [479], [480]

Blood tests of pancreatic function. The *amino acid consumption test* is based on the idea that a stimulated healthy pancreas will deplete plasma amino acids to

make zymogens to a greater extent that than will a damaged pancreas. The specificity of this test is very low, although the sensitivity seems to be high. [480] Low plasma levels of *apolipoprotein B-48* after a 40 g lipid meal have been shown to indicate chronic pancreatitis with exocrine insufficiency. [483], [484] The usefulness of this determination as a diagnostic test remains to be proven.

Pancreatic stimulation tests. After duodenal intubation with a double-lumen tube (one lumen having an aspiration port to remove contaminating gastric juice), the pancreas can be stimulated with intravenous secretin, or cholecystokinin, or by an intragastric liquid meal containing fat, protein, and carbohydrates (Lundh meal). [479], [485], [486] Pancreatic juice is aspirated from the duodenal port, and the serially collected samples are analyzed for volume, HCO_3^- concentration, and, in some laboratories, pancreatic enzymes (lipase, colipase, trypsin, or chymotrypsin). An HCO_3^- concentration lower than 90 mM suggests chronic pancreatitis, and in research laboratories there are normal values for specific pancreatic enzyme concentration or output. Although the test may be accurate and sensitive, it requires intestinal intubation and is time consuming. Therefore, this test is not uniformly used clinically and may be better suited for research protocols.

Fecal pancreatic enzymes. Measurements of fecal chymotrypsin, trypsin, lipase, and elastase have all been proposed as tests for pancreatic insufficiency and have sensitivity and specificity in the 80% to 90% range. [479], [480]

Tests for Bacterial Overgrowth

Schilling test. Vitamin B_{12} (cobalamin) is not absorbed unless it is bound to intrinsic factor, and intrinsic factor cannot bind if salivary and gastric R proteins are not degraded by the pancreatic proteolytic activity in the upper small intestine. Approximately 50% of patients with severe pancreatic insufficiency are found to have impaired vitamin B_{12} absorption measured with the Schilling test. Obviously, this is not very specific because of diminished vitamin B_{12} absorption in patients with pernicious anemia, bacterial overgrowth, severe mucosal intestinal disease, or extensive ileal disease or resection. The Schilling test may be useful in distinguishing between bacterial overgrowth and mucosal disease in the setting of an abnormal D-xylose test, especially if the test reverts to normal after oral antibiotic treatment. [487]

Bacterial growth from duodenal aspirates. The growth of more than 10^6 bacterial colonies per milliliter of duodenal fluid suggests bacterial overgrowth. Care should be taken to collect samples anaerobically, and they should be plated for aerobic and anaerobic cultures immediately. [488] The test has not been standardized, and contamination with oropharyngeal bacteria can be a problem.

H_2 and ^{14}C breath tests for bacterial overgrowth. The breath hydrogen test can be used to diagnose bacterial overgrowth and has the advantage of not using radioactive isotopes. However, up to 20% of people are colonized by bacteria that do not produce H_2; thus, the sensitivity and specificity of this test may be poor (see Chapter 78). The *^{14}C–D-xylose breath test* is said to be more specific for bacterial overgrowth, with 85% of patients having an increase in exhaled $^{14}CO_2$ within 60 minutes of ingesting 1 g of ^{14}C–D-xylose. [489] Because most of the ^{14}C-labeled xylose is absorbed in the small intestine, less reaches the colon, where it can be metabolized to CO_2 by bacteria confounding interpretation of the test. The test can be complicated by delayed gastric emptying. Champions of the test claim almost 100% specificity and sensitivity and an even greater reproducibility than that found in duodenal intubation with culture [489], but others report little advantage over glucose breath hydrogen tests. [490] The *cholyl-^{14}C-glycine breath test* is based on the rationale that conjugated (glycine or taurine) bile acids are reabsorbed passively throughout the jejunum and actively absorbed in the terminal ileum. [491], [492] Bacterial overgrowth hydrolyzes the peptide bond, releasing the ^{14}C-labeled glycine, which is then absorbed, metabolized to $^{14}CO_2$, and exhaled in expired air. Unfortunately, the test is not specific, and it gives similar results in patients with terminal ileal disease or resection and may be misleading in severe mucosal disease.

Watery Diarrheas

History and Physical Examination Useful clues to the cause of watery diarrheas include the volume of diarrhea, the number of stools, the presence of nocturnal diarrhea, incontinence, and a history of hypokalemia (Fig. 42-9). A detailed dietary and medication history, history of past surgeries, and positive Rome criteria for IBS may facilitate a diagnosis of diarrheas caused by ingested substances, carbohydrate malabsorption, bile acid diarrhea, postvagotomy status, or IBS. These diarrheas often respond, at least partially, to fasting.

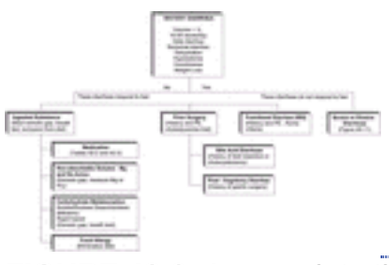

FIGURE 42-9. Approach to the evaluation of watery diarrheas.

Conversely, a history of 10 to 20 bowel movements per day, particularly nocturnal bowel movements with incontinence, should suggest the secretory diarrheas, as should documented electrolyte abnormalities (hypokalemia and acidosis) or dehydration, particularly if accompanied by systemic symptoms such as flushing. The presence of long-standing diabetes or alcoholism obviously is necessary for a diagnosis of these forms of diarrhea. Weight loss is not usually a component of most of the watery diarrheas, except for advanced neuroendocrine tumors and mastocytosis. Physical examination is helpful only if the thyromegaly of medullary carcinoma, the cutaneous flushing of the neuroendocrine tumors, and systemic mastocytosis are evident, or if there are the dermatographism of systemic mastocytosis and the migratory necrolytic erythema of glucagonoma. [323] The concomitant presence of peptic ulcer should suggest gastrinoma or systemic mastocytosis. Scars from previous surgery raise the question of postvagotomy diarrhea or terminal ileal resection, causing bile acid diarrhea. Autonomic dysfunction (e.g., postural hypotension, impotence, gustatory sweating) is almost invariably present in diabetic diarrhea. More details of clues to the diagnosis of pancreatic endocrine tumor syndromes are given in Chapter 97.

Screening Tests for Watery Diarrheas There are no screening tests for these diarrheas. Elimination diets (therapeutic trials) for the carbohydrate diarrheas and food allergy may be used.

Radiography (Watery Diarrhea)

Contrast radiographs. Routine films of the gastrointestinal tract are not usually helpful in the diagnosis of watery diarrheas unless there is evidence of a previous vagotomy, extensive small bowel resection or cholecystectomy, the presence of a tumor (carcinoid or villous adenoma), or a bowel filled with fluid (endocrine tumor). The radiographic appearance of an ahaustral colon with focal areas of spasm (pseudostrictures) may suggest chronic laxative abuse (the cathartic colon). [493]

Special radiologic tests. Suspected pancreatic endocrine tumors and metastatic medullary carcinoma of the thyroid can be accurately localized with a combination of abdominal ultrasound, abdominal computed tomography, magnetic resonance imaging, and selective abdominal angiography, along with radiologic somatostatin receptor screening [494], [495] and endoscopic ultrasound [496] (see Chapter 67 and Chapter 97). Medullary carcinoma of the thyroid lesions have also been localized with octapeptide (^{111}In-OTPA)-cholecystokinin analog scanning. [497] Somatostatin receptor scintigraphy has proven to be both sensitive and useful in the diagnosis and evaluation of Zollinger-Ellison syndrome. [494]

Flexible Sigmoidoscopy or Colonoscopy and Biopsy Patients with severe watery or elusive diarrhea should have colonoscopy to rule out villous adenomas of the rectosigmoid and biopsy to rule out microscopic or collagenous colitis, mastocytosis, or early Crohn's disease. Colonoscopy also may reveal melanosis coli secondary to chronic anthracene laxative use, and this can be confirmed by biopsy. Melanosis coli can develop as early as 4 months after starting to use such laxatives and may require a year after discontinuance to resolve. [498], [499] Investigators have suggested that IBD alone can cause melanosis coli. [500] Terminal ileal biopsy may give an indication of type 2 bile acid diarrhea, Crohn's disease, or infectious disease. [501] Colonoscopy and biopsy have proven very useful for diagnosis in the tertiary referral setting. However, they have been shown to have poor yield as *initial tests* in patients presenting with diarrhea and a normal gross appearance of the colon. [502], [503] Biopsy can be useful to distinguish acute self-limited (infectious) colitis from IBD. [504]

Stool Tests for Watery Diarrhea

Fecal carbohydrate tests and fecal pH. Tests used for the determination of urine sugar can be applied as qualitative tests for stool carbohydrate, although these tests have not been standardized. [189] Urine dipsticks will give a positive reaction to glucose, but not to other sugars. Clinitest tablets (Bayer Corp., Mishawaka, IN) will give a positive reaction to glucose, galactose, fructose, maltose, and lactose, but not to sucrose, lactulose, sorbitol, or mannitol. Carbohydrate malabsorption can be detected by measuring pH on a homogenized stool sample. The breakdown of unabsorbed carbohydrate by colonic bacteria to short-chain fatty acids reduces stool pH to less than 5.3 in pure carbohydrate malabsorption. However, in generalized malabsorptive disease, stool pH is more than 5.6 and is usually 6.0 or greater. [31], [32]

Laxative screen. To screen for laxative abuse in factitious diarrhea, it is necessary to send stool or urine to specific reference laboratories for qualitative tests for emetine (a component of ipecac), bisacodyl, castor oil, or anthraquinone. [189], [342], [343], [344], [345], [346], [347], [348], [349], [350] and [351], [505] Screening is not available for the docusate salts (dioctyl sodium sulfosuccinate), which are present in many of the over-the-counter cathartics. Stool SO_4, PO_4, and Mg analysis detects those factitious diarrheas caused by osmotic cathartics. Although there is controversy regarding the ethics and legality of a room search for laxatives and diuretics, most

believe that it is warranted because of the expense and risk to the patient of the extensive evaluation. [189] Certainly, a room search can be a test with the highest diagnostic yield for surreptitious laxative abuse. [342], [343], [344], [345] and [346]

Tests for Carbohydrate Malabsorption

Lactose tolerance test. To detect either congenital or acquired lactase deficiency, 50 g of lactose is administered, and plasma glucose is measured at 1 and 2 hours. A plasma glucose increase of more than 20 mg/dL is a normal response. [276] Patients who lack lactase and therefore the ability to split lactose into glucose and galactose have little increase in plasma glucose. This test does not have good sensitivity and has largely fallen out of favor because of the more sensitive and simple breath H_2 lactose test (see later).

H_2 breath test for carbohydrate malabsorption. H_2 breath tests can be used to study carbohydrate malabsorption or bacterial metabolism because the sole source of H_2 in the mammal is bacterial fermentation. In patients in whom therapeutic trial of carbohydrate-restricted free diet is inconclusive, breath H_2 testing may be indicated. [189], [472], [506] Because H_2 is produced only by bacterial fermentation of carbohydrates, increased breath H_2 excretion after a carbohydrate challenge can be used to uncover small intestine bacterial overgrowth, disaccharidase deficiency, and even excess carbohydrate wastage that might occur with motility disorders. [507] Patients with small intestine mucosal disease and pancreatic exocrine insufficiency also may have mild carbohydrate malabsorption. Bacterial overgrowth of the small bowel may cause an early peak of increased H_2 production within 2 hours of giving a carbohydrate meal. In disaccharidase deficiency, small intestine mucosal disease, or pancreatic insufficiency, the peak in H_2 may come later, between 3 and 6 hours after ingestion, when the carbohydrate reaches the colonic bacteria. [508], [509] The increase in H_2 excretion by patients with pancreatic insufficiency can be reduced by concomitant administration of pancreatic enzymes. Therefore, depending on what kind of carbohydrate malabsorption is sought, an oral dose of lactose (0.25 to 1.0 g/kg body weight), glucose (50 g), lactulose (10 g), fructose (1 g/kg), or rice flour (100 g) may be given after an overnight fast. To test for lactose intolerance, a lactose dose of 25 g is frequently used, although a 50-g test dose may be more sensitive and a 12.5-g dose more specific. [506] A breath H_2 test using D-xylose has been proposed. [478] Breath H_2 tests must be standardized for each carbohydrate and dose, but generally an increase of over 20 ppm in exhaled H_2 over baseline values within the first 3 to 8 hours of ingestion is diagnostic. It is important to realize that the breath H_2 test measures only carbohydrate malabsorption, and 10% to 20% of people have a gut flora incapable of producing H_2. [472]

Tests for Bile Acid Malabsorption

Cholyl-^{14}C-glycine breath test. This test can be used also to identify impaired ileal absorption of bile acids, although it is neither specific nor sensitive. [491], [492] The measurement of cholyl-^{14}C-glycine excreted in the stool may increase the sensitivity but adds to the difficulty of performing the test.

^{75}SeHCA Test. Because selenahomotaurocholic acid (SeHCA) is an analog of taurocholic acid and has a similar enterohepatic circulation, it may be of value in detecting ileal dysfunction as a cause of bile acid diarrhea. ^{75}Se-labeled HCA is given orally, and the patient is scanned with the gamma camera; those who retain less than 34% of the administered dose after 3 days are considered to have increased bile acid loss. [510], [511] and [512] This test has not been approved for use in many countries.

Serum HCO concentration. With impaired bile acid absorption, cholesterol 7a-hydroxylase increases in the liver, and serum levels of 7a-hydroxy-4-cholesten-3-one (HCO) rise. These levels have been shown to correlate well with the ^{75}SeHCA test and thus may become useful for diagnosing bile acid malabsorption. [513]

Blood and Urine Hormone Levels Endocrine tumors such as carcinoids, gastrinoma, vipoma, medullary carcinoma of the thyroid, systemic mastocytosis, and glucagonoma can be diagnosed by demonstrating elevated blood levels of 5-HT or urinary 5-hydroxyindoleacetic acid (5-HIAA) and elevated serum levels of gastrin, VIP, calcitonin, histamine, somatostatin, and glucagon, respectively (see Chapter 97). These may be obtained through commercial (e.g., ARUP Laboratory, Salt Lake City, UT) or research laboratories (the National Institutes of Health, Bethesda, MD). There is little diagnostic usefulness in measuring other hormones secreted by these tumors. [189], [514], [515] These tests must be viewed cautiously because no endocrine cause of diarrhea is found in a significant percentage of those with elevated blood hormone levels. The finding of an elevated blood hormone level may signify the presence of incurable metastatic disease. [514] Provocative tests such as pentagastrin stimulation for the diagnosis of Zollinger-Ellison syndrome or calcitonin-secreting tumors are addressed in Chapter 67 and Chapter 97.

Inflammatory Diarrheas

History and Physical Examination The important clinical manifestations of inflammatory diarrheas are the signs and symptoms of inflammation and the effects of severe chronic protein loss (Fig. 42-10). Fever with acute or chronic abdominal pain, particularly if it is localized to either right or left lower quadrants, may be an important clue to IBD ileitis or colitis. Eosinophilic, allergic, and immunologic enteritis usually causes diffuse pain and tenderness. Severe protein-losing enteropathy is manifested either as peripheral edema, ascites, or anasarca in the absence of liver disease or proteinuria. Diarrhea in these inflammatory diseases may be meager (e.g., the pseudodiarrhea of proctitis), or it may be fairly severe. Exsanguinating hemorrhage, as in fulminant colitis, or moderately severe secretory watery-type diarrhea, as in graft versus host disease, can occur. Milder watery diarrheas may occur in microscopic or collagenous colitis and eosinophilic gastroenteritis. Crohn's disease, radiation enteritis or colitis, and milk or soy protein allergy may present with either watery or bloody diarrhea.

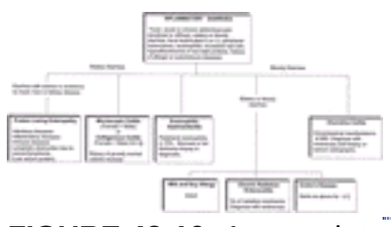

FIGURE 42-10. Approach to the evaluation of inflammatory diarrheas.

Systemic manifestations of inflammatory disease may be prominent in these patients. These include oral aphthous ulcers, polymigratory arthritis, uveitis, and various dermatitides, including erythema nodosum, pyoderma gangrenosum, and the palpable purpura of vasculitis. As is the case with the mucosal malabsorptive disease with severe subepithelial inflammation, anorexia can be an important cause of the weight loss.

Screening Tests for Inflammatory Diarrheas Peripheral blood findings of leukocytosis, eosinophilia, elevated sedimentation rate, C-reactive protein, hypoalbuminemia, or low total serum proteins suggest the presence of inflammation, hypersensitivity, or severe protein-losing enteropathy. Low serum albumin and globulins should always alert one to the possibility of protein-losing enteropathy. The hallmark of the inflammatory diarrheas, however, is the presence of blood, either gross or occult, or leukocytes in the stool. Stool examination for leukocytes should use methylene blue stain and not Gram stain for best results. [107] Fecal leukocyte tests have poor sensitivity for ruling out inflammatory diarrheas [108], [109] and are gradually being supplanted by tests of granulocyte markers such as lactoferrin and calprotectin (see later).

Radiography (Inflammatory Diarrheas)

Contrast radiographs. Plain films or contrast examinations may show diagnostic evidence of IBD or the inflammation that may be present with eosinophilic gastroenteritis or radiation enterocolitis. [516] Edema of the valvulae conniventes may suggest protein-losing enteropathy. Early or mild gut inflammation may be missed entirely by radiography. Computed tomography may also be used for the detection of inflammatory disease of the colon. [517]

Special radiologic tests. Indium-labeled leukocyte scans may be useful in detecting bowel inflammation not evident by endoscopy or contrast radiography. [111], [518], [519] Imaging with ^{99m}Tc antigranulocytic antibodies [520] and ^{123}I-IL-2 [521] has shown promise.

Endoscopy and Biopsy Clues to the diagnosis of any of the inflammatory diseases of the colon and terminal ileum can be made earliest with colonoscopy and biopsy. Thus, colonoscopy and biopsy become very useful, if not mandatory, tests. Upper endoscopy and biopsy can be useful to diagnose gastroduodenal inflammatory diseases.

Fecal Lactoferrin and Calprotectin Tests In specific laboratories, stool excretion of fecal lactoferrin or calprotectin (constituents of leukocytes) can be used as a quantitative index of fecal leukocyte loss. [110], [111] It seems to have reasonable sensitivity (=60%), specificity (80%–90%), and cost effectiveness. [112], [113]

Tests for Enteric Protein Loss

^{51}Cr-albumin clearance. Measurements of enteric protein loss are sensitive tests for inflammatory diarrhea. This can be accomplished by measuring 24-hour stool excretion or clearance of ^{51}Cr-labeled albumin. Clearance is determined by 24-hour stool excretion divided by mean serum activity, just as in the creatinine clearance calculation. [522], [523] More recently, endogenous labeling of albumin can be accomplished by injecting 50 µCi of $^{51}CrCl_3$ 2 days before collecting 24- to 72-hour stool samples. A sample of blood is drawn during the stool collection to determine serum activity and calculate clearance.

a_1-Antitrypsin clearance. a_1-Antitrypsin clearance of native endogenous proteins, particularly, has proven to be as sensitive as ^{51}Cr-labeled protein clearance and avoids the administration of radioactive material. [522], [523], [524] and [525] The principle of the clearance studies is the same as with radioactive albumin. a_1-Antitrypsin is measured in serum and stool either by radial immunodiffusion or by laser nephelometry with monospecific antisera. Stool determination requires an aliquot of lyophilized stool to be extracted with saline. When combined with a protein pump inhibitor, it can be used to diagnose protein-losing gastropathy. [526]

^{99m}Tc-albumin scintography. The sequestration of the albumin in the protein-weeping bowel can be made by ^{99m}Tc-albumin scintography. This may become a useful, noninvasive test for protein-losing enteropathy. [527], [528]

Evaluation of Severe or Elusive Diarrhea

To evaluate severe or elusive diarrheas, measurements of stool fat, electrolytes, and osmolality on timed stool collections, combined with observing the response of stool volume to fasting, are very useful (Fig. 40-11). A 48- to 72-hour stool collection must be obtained while the patient is on a 100-g fat diet and the collection repeated while the patient has fasted. The fasting portion of the collection must be performed in the hospital; however, the nonfasting part of the collection can be obtained on an outpatient basis. The stool should be analyzed for appearance, weight, quantitative fecal fat, electrolytes (Na^+, K^+, and if thought necessary, Cl^-, PO_4, and Mg^{2+}), osmolality, fecal pH, and laxative screen.

Stool Weight, Response to Fast, and Stool Osmotic Gap There are many theoretical and practical problems encountered in using the response to fasting and feeding and the measurements of stool osmotic gap to diagnose the cause of diarrhea. [31], [32], [33] and [34], [189], [529], [530], [531], [532] and [533] First, in some diseases, the ingestion of a drug or feeding itself initiates the secretory phenomenon. Under these circumstances, the fasting stool may return to normal, and yet the pathophysiology is that of a secretory diarrhea. Examples are the patients who factitiously ingest laxatives and some patients with microscopic (lymphocytic) or collagenous colitis. Second, the stool specimen may be accidentally contaminated with urine or purposefully diluted with water. Third, there may be malabsorptive diseases that also have a secretory component. For example, in celiac sprue there is secretion by the small intestine, and malabsorbed fat also can initiate active secretion in the colon. Similarly, in viral diarrhea, there may be simultaneous secretion by the hyperplastic crypts and malabsorption by the damaged villus epithelium. Nonetheless, there are some general responses of stool volume to fast in the various diarrheas, especially the watery diarrheas, as shown in Figure 42-9 and Figure 42-11. Steatorrheic stools are usually less than 700 g, and stool weight returns to normal on fasting. Stool osmotic gap measurements are often indeterminate in these patients. Inflammatory diarrheas are variable in weight, but usually less than 1000 g, and have a variable response to fasting. As is the case in steatorrhea, the stool osmotic gap is not usually helpful.

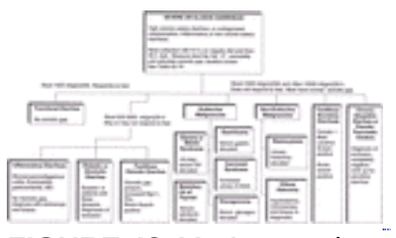

FIGURE 42-11. Approach to the evaluation of severe or elusive diarrheas.

The measurements of stool electrolytes for calculation of osmotic gap are useful only as guidelines for the classification of diarrhea. [189], [190], [265], [529], [530], [531], [532] and [533] In practice, to calculate the stool osmatic gap, it is not correct to use measured stool osmolality; it may be falsely altered by bacterial degradation of carbohydrate after the stool is passed. Therefore, the use of normal plasma osmolality, 290 mOsm/kg H_2O, is recommended because newly passed stool is essentially isosmotic with plasma. Na^+ and K^+ concentrations must be measured in the stool and then multiplied by 2, to account for the obligate (mainly organic) anions in the stool. The osmotic gap, or the difference between stool osmolality (or 290 mOsm) and 2 × [Na^+ + K^+] concentrations, should normally be less than 125 and is usually less than 50 (Table 42-14). This number takes into account unabsorbed divalent cations (Ca^{2+} and Mg^{2+}) and ammonium and their obligate anions.

TABLE 42-14 Stool Osmotic Gap as a Guide to the Pathophysiology and Diagnosis of Diarrhea

In secretory diarrheas, the solute causing the movement of water from blood to bowel lumen is composed of the secreted Na^+ and K^+ molecules. Thus, in secretory diarrheas, twice the sum of the stool Na^+ plus K^+ concentrations will approximate stool osmolality. Furthermore, unless the secretory stimulus is being ingested (e.g., laxatives), the diarrhea and stool weight should be only minimally or moderately reduced if the patient fasts; fasting stool weight should remain higher than 200 g/24 h. In general, therefore, if stool Na^+ concentrations are greater than 90 mM and the osmotic gap is less than 50, then a secretory diarrhea is present (see Table 42-14). Conversely, if stool Na^+ is less than 60 mM and the osmotic gap is greater than 125, then it is likely to be an osmotic form of diarrhea. In osmotic diarrhea, it is the ingestion of nonabsorbable (or nonabsorbed) solutes that displaces Na^+ from the stool and causes the osmotic gap and the diarrhea. Osmotic diarrhea should disappear if the patient fasts, and stool weights should return to values of less than 200 g in 24 hours. Stools with Na^+ concentrations of 60 to 90 mM and calculated osmotic gaps of 50 to 100 can result from either secretory or malabsorptive abnormalities or from diseases that have as their pathophysiological basis some element of both secretion and malabsorption. [31], [34], [189], [529], [530], [531], [532] and [533] Stool osmolality, measured by the freezing point depression method, may have some utility in diagnosis because a low stool osmolality suggests contamination of the stool inadvertently with urine, or purposefully with water in the case of factitious diarrhea [31], [34], [189], [529], [530], [531], [532] and [533] (see Table 42-14). In patients suspected of having magnesium-induced diarrhea, fecal analysis reveals stool Mg^{2+} concentrations greater than 45 mM (usually greater than 100 mM) or Mg^{2+} output greater than 15 mmol/24 h. In patients in whom the magnesium ingestion is innocently motivated, the diarrhea disappears on fasting. In those ingesting magnesium surreptitiously, or in an infant or child being purposefully fed magnesium, stool analysis demonstrates an osmotic gap greater than 100 mOsm/kg H_2O, and the excess stool weight and osmotic gap do not disappear on fasting. [263], [264], [265] and [266] In patients suspected of having sodium anion–induced diarrheas, the stool Na^+ concentration is 90 mM or greater, and twice the concentrations of Na^+ + K^+ equal approximately 290 mOsm (see Table 42-14). This type of diarrhea mimics secretory diarrhea, even though it is an osmotic diarrhea, because the stool Na^+ content is high (>90 mM), and there is not an osmotic gap. This type of diarrhea can be diagnosed, in the absence of a history of ingestion of Na_2SO_4- or Na_2PO_4-containing substances, by determining stool Cl^- concentration. If nonabsorbable anions have been ingested, they displace stool Cl^-, and the resulting stool Cl^- value is usually less than 20 mM. [31], [189] There are technical and practical problems with using stool analysis as a guide to the diagnosis of diarrhea. [31], [34], [529], [530], [531], [532] and [533] Some hospitals are unable to measure stool electrolytes and osmolality, and therefore an aliquot of the homogenized stool sample must be sent frozen on dry ice to a reference laboratory for analysis. Therefore, the response to fast may be a more practical way to distinguish secretory from osmotic diarrheas. [529]

ANTIDIARRHEAL THERAPY

Mechanisms of Action

Antidiarrheal agents can be divided into two categories: agents useful for mild to moderate diarrheas (Table 40-15) and those helpful in secretory and other severe diarrheas (Table 42-16). A major drawback of current antidiarrheal drugs is that some have no antisecretory activity; for example, the bulk-forming agents only increase the consistency of stool and do not decrease its elaboration. [534] Other antidiarrheal agents have only mild proabsorptive or antisecretory action. [535], [536] and [537] Although in vitro and in vivo studies in experimental animals suggest that many of the agents listed in Table 42-15 and Table 42-16 have proabsorptive or antisecretory activity, most of them are lacking this effect in human studies, or else it is minimal. Most of the current antidiarrheal agents have their activity by altering the intestinal motility. Bismuth salicylates, loperamide, clonidine, phenothiazine, and somatostatin have mild antisecretory activity, but they also cause dilation of the small intestine and colon and decrease peristalsis. [535], [536] and [537] The opiates also cause disordered contractions of the distal large bowel and increased anal sphincter tone. [538], [539], [540], [541] and [542] The sum of these effects is to trap fluid within the intestine and to put it in contact with the mucosa for a greater period of time, allowing more complete absorption.

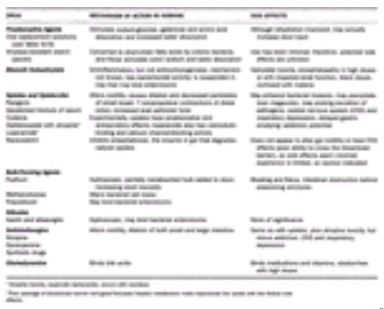

TABLE 42-15 Antidiarrheal Agents for Mild to Moderate Diarrheas

TABLE 42-16 Agents Helpful in Secretory and Other Severe Diarrheas*

More recently discovered (or rediscovered) effective proabsorptive agents include the enkephalinase inhibitors, [543] and newer or natural preparations of amylase-resistant starch or pectin. [126] The enkephalinase inhibitor racecadotril (Acetorphan) inhibits the breakdown of natural endogenous opiates (enkephalins) by intestinal tissue, including the intestinal epithelia. It is effective against experimental secretory diarrheas, and its proabsorptive effect is prevented by naloxone, indicating the role of endogenous opiates in its proabsorptive action. [544], [545] and [546] Pectins are a form of amylase-resistant starch that are not hydrolyzed by amylase and therefore are not absorbed in the small intestine. They are converted to short-chain fatty acid by colonic bacteria, and these short-chain fatty acids stimulate colonic water and electrolyte absorption. [125] (see later). Other potential new antisecretory agents are the new 5-HT $_3$ receptor antagonists [547] and enteric nervous system neurotransmitter antagonists such as antisubstance P or VIP. [3]

Agents for Mild Diarrhea

Bismuth subsalicylate [99], [102], [118], [548] and loperamide preparations [99], [102], [118], [540], [549] continue to prove effective and safe in mild to moderate diarrhea of all types (see section “ Treatment of Acute Infectious Diarrheas”). The antimobility efforts of the opiates listed in Table 42-15 may be symptomatically useful in mild diarrheas, but they may result in inadequate fluid replacement therapy in severe diarrheas if one is relying on stool output as a gauge for replacing fluid losses. The problems may be especially significant in infants and children. Furthermore, the antimotility effects are not desired if the diarrhea is caused by microbiologic organisms because stasis may enhance their invasion even to the degree of causing a narcotizing enterocolitis. [550] These drugs also delay subsequent clearance of the microorganisms from the bowel, thus increasing carriage time. Clinical evidence also suggests that such drugs are dangerous in severe IBD, reportedly precipitating the development of toxic megacolon in severe ulcerative colitis. Therefore, although they appear to be safe, their use must be tempered in severe secretory diarrheas, in invasive traveler’s diarrheas and in severe IBD.

The newer antienkephalinase, racecadotril, has been shown to be effective in controlled trials in children [544], [545] and adults. [551], [552] Similarly, the use of amylase-resistant starch (pectin) has been effective in controlled trials of treatment of protracted infectious diarrheas. [125], [126] Because experience with these newer agents is limited, potential side effects are unknown. 5-HT antagonists have proven useful in diarrhea-predominant IBS, [547] but the occurrence of ischemic colitis during their use has resulted in temporary removal from the market and severe use restrictions.

Agents for Secretory Diarrhea

The use of drugs with potentially serious side effects (see Table 40-16) can be justified for treatment of severe secretory diarrheas such as the diarrheas of carcinoid syndrome and neuroendocrine tumors, diabetic diarrhea, graft versus host disease, 5-fluorouracil–leucovorin diarrhea, short bowel syndrome, and AIDS diarrhea. [535], [536] and [537], [553], [554], [555], [556], [557], [558], [559], [560], [561], [562], [563] and [564] The somatostatin analog octreotide appears to have its major antisecretory effect in carcinoid syndrome and in some other neuroendocrine tumors because it inhibits hormone secretion by the tumor. It also may prevent some of the other clinical manifestations of hormone release (e.g., flushing, tachycardia, skin rash). Unfortunately, patients with large tumor burdens may escape from the drug, and then their management may require debulking surgery or agents such as phenothiazine or calcium channel blockers, which have serious side effects. Octreotide may not be helpful in the diarrhea of medullary carcinoma of the thyroid [557] and it is of only limited usefulness in short bowel syndrome [555], [561] and the refractory diarrhea of AIDS. [559], [564] Furthermore, resistance may develop if neuroendocrine tumors are treated for a long period. [560] The newer long-acting preparations, Sandostatin-LAR (Novartis) and Lanreotide-PR (Ipsen-Biotech), have made therapy with this drug more convenient for the patient. [556], [563]

Although clonidine is very useful in the diarrhea of opiate withdrawal and sometimes in patients with diabetic diarrhea, particularly those who already have severe postural hypotension that cannot be made much worse by the agent, such side effects become a limiting factor. [364] Lithium carbonate, bromocriptine, nicotinic acid, and berberine have all been reported to be useful in certain secretory diarrheas. [535], [536] and [537]

Indomethacin, a cyclooxygenase blocker that inhibits prostaglandin production, may occasionally be useful in neuroendocrine tumor and in patients with irritable bowel and possible food allergy (see previous discussion), but it may be harmful in IBD. Indomethacin is most useful in patients with acute radiation diarrhea, AIDS diarrhea, and the diarrhea accompanying villous adenomas of the rectum or colon. The response of these diarrheal diseases to specific cyclooxygenase-2 agents has not been reported.

Agents for Inflammatory Diarrhea

In IBD, glucocorticoids have an effect within 72 hours on both prostaglandin and leukotriene production, [565] and they can be shown to have a proabsorptive effect on the intestine within 5 hours of administration. [565], [566] and [567] Thus, in IBD, steroids are both antiinflammatory and antidiarrheal.

Newer treatment options for inflammatory diarrhea include drugs with immune suppression and immunomodulation activity such as 6-mecaptopurine, methotrexate, cyclosporine, infliximab, thalidomide, and budesonide. [389], [568] These agents can be considered to be antidiarrheal medications by virtue of being antiinflammatory agents. Probiotics show promise in inflammatory diarrheas. A *probiotic* is defined as a viable microbial dietary supplement that affects the host in a beneficial way through its effects on the intestinal tract. [569], [570] Several different species are available: *Lactobacillus* GG, *L acidophilus, Bifidobacterium* species, and *Saccharomyces boulardii* are some of those more widely used. [93], [570], [571] They have been proposed and explored in trials for both prevention and, in some instances, treatment of antibiotic-associated diarrhea, viral diarrhea, bacterial diarrhea, lactose intolerance, IBD, and others. [93], [572], [573] and [574] It is too early to tell whether these probiotics will have efficacy. For every positive trial, there seems to be a negative outcome in another study. This issue will be clarified over the next few years.

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CHAPTER 43

Arnold Wald

APPROACH TO THE PATIENT WITH CONSTIPATION

Although constipation is a common gastrointestinal complaint in clinical practice, [1] some uncertainty exists as to the precise definition of the term. Patients complain of constipation if they think that they defecate too infrequently or with too much effort, if their stools are too hard or too small, if defecation is painful, or if they have a sense of incomplete evacuation. This lack of objectivity has contributed to the controversy concerning the incidence, pathogenesis, and treatment of constipation and defecation disorders. Furthermore, the availability of over-the-counter laxatives and their long-term and often inappropriate use may result in laxative dependence, may damage the bowel, and may lead to problems where none previously existed.

DEFINITIONS

Although no single definition is applicable to all constipated persons, it is useful to review the most common definitions to provide guidelines for clinical practice. *Bowel frequency* lends itself to quantification and is a convenient measurement with which to evaluate patients or to survey large populations. Because population surveys have found that most people consuming a Western diet defecate at least three times per week, [2], [3] *constipation* has been defined as a frequency of defecation of twice weekly or less. Other smaller surveys suggest that the lower limit of normal is five times per week, varying according to sex and race [4]; however, this has not been widely adopted as the standard. Furthermore, frequency alone may not be a sufficient criterion because many constipated patients complain of excessive straining at defecation, with or without hard stools, even though frequency of defecation is within the normal range. *Difficulty during defecation* is highly subjective and difficult to quantify. Attempts to do so have used expulsion of water-filled balloons, solid spheres of different volumes, [5], [6] and synthetic stool with the use of radiologic or scintigraphic techniques. [7], [8] These techniques have methodological shortcomings and cannot be used for large population surveys because of their invasive nature or exposure to radioactivity. Even more problematic are measurements of *stool weight* and *consistency.* Normal stool weight varies widely among individuals, with considerable day-to-day fluctuation [9]; stool consistency is unpleasant to measure and difficult to quantify. [10] The clinician should use a combination of subjective and objective criteria as a starting point in assessing such complaints. These include a frequency of defecation less than three times weekly, alone or in conjunction with subjective complaints of difficulty with defecation, [11] especially if there has been a distinct change in regular bowel habit.

SOCIOECONOMIC AND MEDICAL CONSEQUENCES

The costs of constipation are considerable. In the United States, more than $800 million are spent for laxatives; many of these drugs are used unnecessarily, and some may be harmful. Constipation accounts for about 20,000 hospitalizations per year and 2.5 million office visits. [12] No data exist regarding additional costs generated as a result of medical evaluations, diagnostic studies, surgery, and absences from work related to constipation. [13]

Management of problems secondary to constipation adds to the economic burden of this condition. For example, chronic constipation may be associated with urinary tract infections, [14], [15] and in children it may be associated with enuresis and vesicoureteral reflux, as well as fecal soiling. [16] These conditions often improve or disappear with treatment of constipation.

Fecal impaction resulting from constipation is the most common cause of fecal soiling in institutionalized elderly persons. Together with urinary incontinence, it is a contributing factor in the removal of elderly persons from their homes. Chronic constipation also may lead to pudendal nerve damage and fecal incontinence in middle-aged and older women. [17], [18] In more advanced cases, rectal prolapse may result.

In young women, chronic severe constipation is associated with an increased incidence of unnecessary surgery, particularly appendectomy, hysterectomy, and ovarian cystectomy. [19] The reasons for this are unknown. In older persons, dilated loops of colon may result in volvulus or ischemic colitis. Stercorous ulcers with bleeding or perforation constitute a hazard in patients with fecal impaction. [20] Finally, there is some evidence that links constipation with colon and rectal cancer in women. [21]

PATHOPHYSIOLOGICAL CONSIDERATIONS

Constipation may be conceptually regarded as disordered movement through the colon or anorectum because transit through the more proximal regions of the gastrointestinal tract is normal in most constipated patients. From a pathophysiological viewpoint, impairment of large intestine transit can occur because of a primary motor disorder, in association with many diseases, or as a side effect of many drugs (Table 43-1 and Table 43-2). Diseases associated with constipation include metabolic and endocrine disorders and those neurogenic disorders that affect the gastrointestinal tract.

Metabolic and Endocrine Disorders
Diabetes mellitus
Hypothyroidism
Hypercalcemia, hypokalemia
Pregnancy
Porphyria
Panhypopituitarism
Pheochromocytoma
Glucagonoma
Neurogenic Disorders
Peripheral
Hirschsprung disease
Chagas disease
Neurofibromatosis
Ganglioneuromatosis
Autonomic neuropathy
Hypoganglionosis
Intestinal pseudoobstruction (myopathy, neuropathy)
Central
Multiple sclerosis
Spinal cord lesions
Parkinson disease
Shy-Drager syndrome
Trauma to nervi erigentes
Cerebrovascular accidents
Collagen Vascular and Muscle Disorders
Systemic sclerosis
Amyloidosis
Dermatomyositis
Myotonic dystrophy

TABLE 43-1 Secondary Causes of Functional Constipation

Analgesics
Anticholinergics
Antispasmodics
Antidepressants
Antipsychotics
Antiparkinsonian drugs
Cation-Containing Agents
Iron supplements
Aluminum (antacids, sucralfate)
Calcium (antacids, supplements)
Barium sulfate
Metallic intoxication (arsenic, lead, mercury)
Neutrally Active Agents
Opiates
Antihypertensives
Ganglionic blockers
Vinca alkaloids
Anticonvulsants
Calcium channel blockers
$5\text{-}HT_3$ antagonists

TABLE 43-2 Drugs Associated with Constipation

Chronic illnesses often lead to physical and mental impairments that can produce or exaggerate constipation. This condition may be further exacerbated by inactivity or physical immobility, which can lead to fecal retention. A bedridden patient may be unable to respond to defecatory signals because of inadequate toileting arrangements; as a result, fecal retention may lead to megarectum, diminished rectal sensation, and fecal impaction. Other factors that may contribute to constipation in the bedridden patient include underlying illness, medications, and dietary inadequacies. Generalized weakness or striated muscle diseases, such as dermatomyositis, may result in significant constipation because of poor expulsion efforts. [22]

Metabolic and Endocrine Disorders

The most common of the endocrine disorders that cause constipation are diabetes mellitus and hypothyroidism. Constipation was reported by 60% of an unselected clinic population of diabetic patients. [23] Although constipation may occasionally be severe, in our experience symptoms tend to be mild and responsive to relatively simple measures. Constipation associated with hypothyroidism is also usually mild and improves with thyroid replacement therapy, but it can present with life-threatening megacolon in patients with myxedema. [24] Constipation is said to be common during pregnancy, [25] and some women report that they are constipated immediately before menstruation. [26] Alterations of progesterone and estrogen may be responsible because pregnancy is also associated with decreased lower esophageal sphincter pressures, [27] delayed orocecal transit times, [28] and increased gallbladder volume. [29] Similar changes in orocecal and gallbladder function have been documented during the luteal phase of the menstrual cycle. [30] Although delayed colonic transit also has been reported during the luteal phase, [31] one study failed to substantiate such changes in a nonconstipated population. [32] Other less common endocrinopathies that cause constipation are listed in Table 43-1.

Neurogenic Disorders

Because colonic and anorectal motor functions are coordinated by both the enteric nerves and the extrinsic innervation of the sympathetic and parasympathetic nerves (see Chapter 11), diseases of the central and peripheral nervous systems are often associated with constipation (see Table 43-1).

Disorders of Extrinsic Innervation The distal colon is supplied by parasympathetic innervation derived from the sacral nerves that pass through the pelvis and enter the bowel wall in the lower part of the rectum. [33] Transection of these nerves or lesions in the sacral cauda equina may produce constipation associated with hypomotility, colonic dilation, decreased rectal tone and sensation, stasis of the distal colon, and impaired defecation. [34], [35] Similar findings occur in patients with injury to the lumbosacral spine, [36] in patients with meningomyelocele, [37] and after low spinal anesthesia. [38] Constipation also may occur with high spinal cord damage, but in these patients colonic reflexes are intact, and defecation can often be triggered by digital stimulation of the anal canal. [39] Although the motor response of the sigmoid colon after a meal is reduced in persons with high spinal injuries, responses to pharmacological stimuli are normal. [40] Anal sphincter pressures are normal, but rectal sensation may be impaired, and rectal compliance is reduced in patients with high spinal injuries. [40], [41] The prevalence of constipation in patients with multiple sclerosis is high and is associated with neurogenic dysfunction of other organ systems. [42] Constipation may be exacerbated by physical inactivity or by use of medications with constipating side effects. Severely disabled and constipated patients with multiple sclerosis demonstrate absent colonic motor responses after eating a meal as well as high pressure/volume colonmetrograms that probably result from interruption of normal cortical inhibition of colonic motor activity. [43] Other investigators have found prolonged colonic transit and anorectal changes suggestive of rectosphincteric dyssynergia (see discussion later in this chapter), leading to poor rectal evacuation. [44] Whether these findings apply to most patients with multiple sclerosis with bowel dysfunction is unknown.

Disorders of the Enteric Nervous System

Hirschsprung disease. The classic form of this disorder is characterized by obstipation from birth and colonic dilation proximal to a contracted nonpropulsive segment of distal bowel. Rectal examination reveals an empty rectal vault, but the abdomen is markedly distended by a colon filled with stool. Barium enema discloses the characteristic findings in the distal colon and rectum. In contrast to normal function, the internal anal sphincter does not relax after rectal distention [45] and, indeed, often contracts. The absence of this rectosphincteric inhibitory reflex is universal in Hirschsprung disease, [46] reflecting the absence of intramural ganglion cells of both the submucosal and myenteric plexuses; this results from a developmental arrest of caudal migration of neural crest cells from the notochord during embryonic development. Characteristic histochemical findings include increased acetylcholinesterase and catecholamine activity. [47], [48] In addition, neurons containing hydroxytryptamine are absent, [49] and there is a depletion of inhibitory neurotransmitters such as substance P and vasoactive intestinal polypeptide. [50], [51] The demonstration by manometry of the rectosphincteric inhibitory reflex excludes Hirschsprung disease from diagnostic consideration. However, rectal biopsies to document the absence of neurons must be obtained to confirm the diagnosis. Tissue specimens may be obtained by suction biopsy techniques or by a full-thickness biopsy of the rectal wall. In addition to absent neurons, increased acetylcholinesterase staining also can be useful to establish the diagnosis. [47] Although most patients with Hirschsprung disease are diagnosed by 6 months of age, some patients have a clinically milder disease, and the diagnosis may be delayed well into adulthood. [52], [53] Although some of these patients have a very short aganglionic segment, clinical severity correlates poorly with the length of the aganglionic segment. Some investigators have postulated the existence of ultrashort-segment Hirschsprung disease, in which the disease is limited to the anal canal. [21] Because this area normally contains no ganglia, the rectal biopsy results are not useful, and barium study results are normal. Therefore, the diagnosis rests entirely on the manometric demonstration of an absent rectosphincteric inhibitory reflex. However, the most common reasons for failure to elicit internal sphincter relaxation are technical. The recording instrument must be calibrated correctly, settings must be of adequate sensitivity, the probe must be properly positioned, and balloon inflation of the rectum must be of sufficient volume, particularly in patients with megarectum. In addition, the rectum should be emptied before manometry because relaxation may not be elicited if the rectum is filled with stool. Finally, there often is no inhibitory reflex in premature or low-birth-weight neonates, in whom the reflex may take up to 2 weeks to appear. [54] The existence of ultrashort-segment Hirschsprung disease is therefore doubtful.

Other neurological diseases. Constipation has been reported in patients with both decreased [55] and increased [56] numbers of ganglion cells in the colon, although

quantification of ganglion cells in biopsy specimens is difficult. [21] There also have been reported cases of a condition termed *zonal colonic aganglionosis,* in which discrete areas of the colon are devoid of enteric neurons. [57] These disorders may be congenital or acquired secondary to a vascular accident. Abnormal-appearing enteric neurons in the large intestine have been described as a paraneoplastic phenomenon in patients with generalized intestinal pseudo-obstruction and in patients with severe chronic constipation associated with slow transit (see section " Severe Constipation in Young to Middle-Aged Adults"). The role of long-term laxative use in producing damage to the enteric neurons is unproven. [58], [59] Diagnosis requires special staining and labor-intensive quantification of neurogenic elements of colon specimens [60]; it is also susceptible to interpretive bias. Autonomic neuropathy may be associated with constipation, [61] and secondary autonomic dysfunction is believed to underlie constipation in patients with diabetes mellitus. [22] Diabetic patients with constipation may have delayed or absent [62] gastrocolonic response after a meal.

Idiopathic Constipation

Most constipated patients have no obvious cause to explain their symptoms but are presumed to have an underlying disorder of colonic or anorectal motor function. The precise categorization of distinct subtypes of idiopathic constipation is somewhat difficult because of inconsistent definitions and methodologies. However, it is possible to classify patients with idiopathic constipation into several broad groups based on age at presentation, symptoms, duration of complaint, colonic transit characteristics, and anorectal sensorimotor function.

Idiopathic Childhood Constipation Chronic constipation in childhood is multifactorial in origin and involves both psychological and physiological factors. [63] Although severe behavioral problems may occur and should be identified, idiopathic constipation in children is not synonymous with psychogenic constipation; indeed, behavioral abnormalities are usually mild in affected children and may be secondary to bowel dysfunction. [64], [65] Constipation in children is often associated with fecal impaction and dilation of the rectum and sigmoid colon. [66] Many children have slow colonic transit, usually localized to the distal colon and rectum, [67] which suggests either voluntary withholding behavior or abnormal anorectal function. Although many constipated children complain that they do not sense an urge to defecate, demonstration of rectal sensory impairment has been inconsistent. [68], [69] and [70] In one report, impaired perception of sustained distention of a rectal balloon persisted for as long as 3 years in five of eight children who had been successfully treated. [71] If confirmed, such findings could, in part, explain the high frequency of clinical relapse in successfully treated children. However, another study found that rectal sensory impairment had little effect on therapeutic outcome. [70] Studies of anal sphincter pressures also have been contradictory: resting anal canal tone has been reported to be increased, [68], [72], [73] decreased, [74] or similar [70] compared with that in nonconstipated children. Relaxation of the internal anal sphincter is usually normal, [70] and its absence suggests short-segment Hirschsprung disease. Finally, up to 63% of constipated children with fecal soiling fail to relax the puborectalis and external sphincter muscles when they are asked to defecate, [70], [75] a phenomenon that has been termed *anismus* or *rectosphincteric dyssynergia.* This may be a learned behavior acquired at an earlier age, for example, if attempts to evacuate a large fecal bolus were associated with discomfort or an anal fissure (see section " Behavioral Approaches"). Rectosphincteric dyssynergia could lead to further retention of stool, continuing the vicious cycle of events. However, the contribution of this finding to the pathogenesis or maintenance of constipation in children appears to be negligible. [76]

Severe Constipation in Young to Middle-Aged Adults Chronic constipation in this age group is predominantly confined to women. Abdominal pain is uncommon, and megacolon is rare. Patients may complain of infrequent defecation, excessive straining when defecating, or both, and the condition often fails to improve with fiber supplements or mild laxatives. There are several subtypes within this group of patients who, despite similarities with respect to their clinical pictures, may be distinguished by studies of bowel function and psychological profiles. Approximately 30% of patients who consult a physician for complaints of infrequent defecation and who are unresponsive to therapeutic intervention have normal colonic transit. Patients with normal transit constipation may consciously or unconsciously deny that they defecate and often exhibit evidence of increased psychosocial distress. [77] Because these patients do not complain of abdominal pain, they cannot be diagnosed as having irritable bowel syndrome, but their psychological profiles are similar to those of patients with irritable bowel syndrome. [77], [78] Some patients with normal transit constipation demonstrate abnormalities of anorectal sensory and motor function that are indistinguishable from those in patients with slow transit constipation (see section " Colonic Transit Studies"); the relation of these findings to the patients' complaints is unclear. The remaining 70% of patients with severe constipation exhibit slow colonic transit. Many have *colonic inertia,* defined simply as the delayed passage of radiopaque markers through the proximal colon (Fig. 43-1). Unlike patients with normal colonic transit, those with slow transit have significantly lower psychological distress scores that are similar to those of nonconstipated patients with other gastrointestinal disorders. [77]

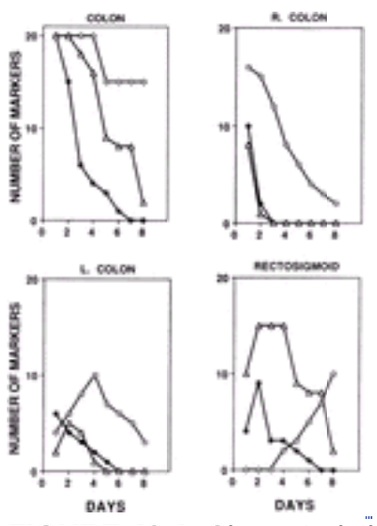

FIGURE 43-1. Characteristic transit patterns of 20 radiopaque markers through the colon during an 8-day period in three groups of constipated patients. With normal transit (*closed circles*), there is rapid disappearance of markers from the colon. In colonic inertia (*open circles*), there is prolonged transit through the right and left colon segments with delayed appearance in the rectosigmoid colon. In outlet obstruction (*open triangles*), transit is normal in the right and left colon, but stagnation occurs in the rectosigmoid colon. (Adapted from Wald A. Colonic transit and anorectal manometry in chronic idiopathic constipation. Arch Intern Med 1986;146:1713.)

Controversy exists concerning the validity of the concept of colonic inertia. Some investigators [39] have argued that colonic transit can vary considerably in the same person on different days and is affected by diet and the menstrual cycle (potentially important in young women). Furthermore, criteria for colonic inertia are imprecise, because colonic stasis could occur as a result of decreased propulsion (hypomotility) or increased distal motility and retropulsion (hypermotility) of markers. Our experience suggests that the menstrual cycle affects colonic transit of markers minimally, if at all, [32] and that significantly slow colonic transit is sufficiently reproducible for clinical purposes. However, one should reserve the term *colonic inertia* for cases in which transit in the proximal colon is delayed without evidence of retropulsion of markers from the left colon; the use of multiple markers of different shapes and shorter sampling intervals may be helpful. Perhaps colonic scintigraphy to measure transit in these patients may help to clarify terminology and pathophysiology. [79] It is not entirely clear which pathophysiological mechanisms are important in patients with colonic inertia. At rest, colonic motility in these patients appears to be similar to that in normal control subjects, [80], [81] and [82] but several studies have demonstrated no increase of motor activity after meals [83] or after administration of bisacodyl. [80], [81] Such findings suggest that abnormalities of the enteric nerve plexus or colonic smooth muscle may exist. Certain histological abnormalities have been demonstrated in resected colon specimens from such patients. [84] These abnormalities include decreased numbers of neurons and axons as well as nuclear abnormalities in the ganglia. Other investigators have demonstrated abnormalities of the contractile properties of colonic smooth muscle. [85] Patients with slow transit constipation have been demonstrated to have decreased numbers of interstitial cells of Cajal, which are thought to play a critical role in intestinal motility. [86] Patients with severe colonic inertia may have symptoms and abnormalities in other areas. These include esophageal dysmotility, [82] delayed transit through the small intestine, [87] and a high incidence of bladder dysfunction and urinary symptoms. [15], [88] Although such disturbances suggest possible neurogenic dysfunction, the precise mechanisms of colonic disturbances remain to be defined. The term *outlet obstruction* [89] has been used to designate a form of idiopathic slow transit constipation in which markers progress normally through the proximal colon but stagnate in the rectum (see Fig. 43-1). This pattern is not specific and may be seen in children with Hirschsprung disease or idiopathic constipation, in nonambulatory or infirm elderly persons with fecal impaction, [90] in patients with megarectum, [91] and in young and middle-aged adults with idiopathic slow transit constipation who demonstrate abnormal responses of the pelvic floor muscles during defecation. [92] This last entity provides another plausible mechanism by which constipation may be produced or exacerbated. Normally, defecation involves the coordinated relaxation of the puborectalis and external anal sphincter muscles together with increased intra-abdominal pressure and inhibition of colonic segmenting activity, which propels stool toward the rectum. In patients with rectosphincteric dyssynergia, ineffective defecation appears to be associated with failure to relax, or inappropriate contraction of, the puborectalis and external anal sphincter muscles. [70], [75], [92], [93] This narrows the anorectal angle and increases the pressures of the anal canal, so evacuation is less effective. Such patients also have difficulty in expelling spheres, water-filled

balloons, or artificial stool from the rectum, compared with most nonconstipated subjects. [75], [92], [94] Because relaxation of these muscles involves cortical inhibition of the spinal reflex during defecation, this pattern may represent a conscious or unconscious act. It can be modified, as has been demonstrated by studies in which biofeedback has normalized defecation patterns in both children and adults with this abnormality. [95], [96] and [97] The pathogenesis of rectosphincteric dyssynergia is not completely understood but is probably multifactorial. Because it can often be modified with operant conditioning training, it is probably an acquired, learned dysfunction rather than an organic or neurogenic disease. Studies indicate that rectosphincteric dysfunction occurs in constipated patients with normal transit and can occur in patients with colonic inertia as well as in those with outlet obstruction. [39] The presence of rectosphincteric dyssynergia can be established in various ways, ranging from simple to sophisticated. During the rectal examination, the contraction of the puborectalis and external anal sphincter muscles can be felt by asking the patient to strain. During anorectal manometry, if the patient is asked to expel the manometer, the characteristic normal pattern is one in which intrarectal pressure increases and external sphincter pressure decreases; in rectosphincteric dyssynergia, in contrast, external pressure increases during attempted expulsion of the manometer. This also can be demonstrated with electromyographic recordings of either or both muscles; normally, electromyographic activity is inhibited during defecation, but it increases with rectosphincteric dyssynergia. Finally, defecography has been advocated to evaluate the expulsion of thickened barium from the rectum radiographically. [7], [98], [99] and [100] In rectosphincteric dyssynergia, the anorectal angle either does not widen or actually narrows during attempts to expel the barium, so little or no expulsion occurs. [21], [101] In our experience, there is agreement on the diagnosis of rectosphincteric dyssynergia between manometry and defecography in approximately 67% of cases. [102] A review of the literature concluded that defecography was of unproven value in evaluating anorectal function and had poor interobserver reliability. [103]

Irritable Bowel Syndrome Constipation in patients with irritable bowel syndrome is most often seen in young to middle-aged adults and is more common in women than men. It is differentiated clinically by the presence of abdominal pain, especially in the lower abdomen, and by the passage of small, hard stools, often with a sense of incomplete evacuation and excessive straining. Not uncommonly, patients also complain of abdominal bloating, flatulence, and upper gastrointestinal as well as nongastrointestinal symptoms. [104] These include heartburn, dysphagia, nausea, genitourinary complaints, back pain, and, in women, dyspareunia. Patients with irritable bowel syndrome frequently demonstrate evidence of increased psychosocial distress. [79] Colonic transit times in these patients are often normal or only modestly slowed. [105] Suggestive findings on colonic radiographs and patterns of colonic dysmotility have been described. These changes are more fully described in Chapter 73 and Chapter 86.

Constipation in the Elderly Some elderly patients with chronic constipation had similar complaints when they were younger. Others develop constipation because of colonic or systemic disorders or as a side effect of medications. There are few data to suggest that aging significantly affects colonic motor function. [106], [107] However, laxatives are used more commonly by elderly women, [106] and physician visits for constipation are more frequent in persons 65 years of age and older. [12] This may be because elderly people often define constipation as the need to strain to defecate, rather than infrequency of bowel movements. [108] Population surveys that rely on bowel frequency alone [2] significantly underestimate the prevalence of constipation in the elderly population. Fecal impaction is a significant problem in elderly patients who are institutionalized. [107] Mental confusion, immobility, or inadequate toilet arrangements may cause such persons to ignore or not act on the urge to defecate, and the fecal bolus may become too large or uncomfortable to pass. The development of megarectum often leads to blunting of rectal and anal sensation that persists even after disimpaction. [90] This predisposes such patients to reaccumulation of feces unless a scrupulous toileting program or periodic evacuation is instituted. Fecal impaction is the most common cause of spurious diarrhea and of fecal incontinence in institutionalized elderly persons. [109]

Megarectum and Megacolon Although only a few patients with constipation have megacolon or megarectum, most patients with a dilated colon or rectum have constipation or defecatory difficulties. In addition, megarectum and megacolon may occur separately or together. [39], [110] Although radiographic criteria exist to diagnose these entities, [111] radiologic assessment does not always correlate with manometric evaluation of rectal elasticity. [66] Idiopathic megacolon may be divided into primary (congenital) megacolon and secondary (acquired) disease. *Primary megacolon* is thought to be associated with neurogenic dysfunction, although histological changes may not be evident without specialized neurohistological staining. [60] In contrast, *secondary megacolon* and megarectum often develop later in life and may occur in response to chronic fecal retention. These patients have increased rectal compliance and elasticity, blunted rectal sensation, an increased threshold, and decreased depth of internal anal sphincter relaxation. [91] To avoid misdiagnosing Hirschsprung disease by anorectal manometry, effective rectal cleansing and larger volumes of rectal distention are often necessary to induce internal anal sphincter relaxation in these patients. Megarectum is generally associated with fecal impaction and soiling, which often occur in children and in physically and mentally impaired elderly persons. [109] In addition, megarectum can occur in Hirschsprung disease, meningomyelocele, and other lesions of the lumbosacral cord and in patients with poor toileting routines. The sensory and motor abnormalities associated with megarectum may be reversible with appropriate therapy in many patients, but this has not been established in the elderly, and abnormalities may persist long after successful treatment in children. [71]

EVALUATION OF CONSTIPATION

History

Evaluating complaints of constipation involves a careful delineation of its duration and characteristics and a review of any previous diagnostic studies and treatment. This information should be obtained with knowledge of the many potential causes of constipation previously detailed.

A critical question concerns the onset and duration of the complaint. Constipation that is present from birth or the neonatal period is most certainly congenital in origin, whereas onset in later life suggests an acquired disorder. A recent change in bowel habit demands a workup for organic disorders, especially in adults, whereas complaints of several years' duration or longer are more likely to be caused by functional disorders.

Next, establishing the nature of the symptoms is important in suggesting various categories of bowel dysfunction and in addressing the specific concerns of the patient. Inquiry concerning frequency of defecation and defecatory difficulties such as excessive straining, discomfort, or sense of incomplete evacuation of the rectum may help to identify specific areas of concern. A complaint of small or hard stools is subjective; it may be helpful to ask the patient to draw a typical stool and an "ideal" one, to determine whether misperceptions or unrealistic expectations exist. The presence of pain or bleeding with defecation should be noted.

Abdominal pain or bloating in association with constipation leads to consideration of the diagnosis of irritable bowel syndrome. Upper gastrointestinal symptoms such as dysphagia, heartburn, early satiety, or vomiting also suggest irritable bowel syndrome or a more diffuse disorder of gastrointestinal function. Genitourinary symptoms, although not specific for constipation from any particular cause, may indicate a central or peripheral neurogenic disorder. Inquiry concerning laxative use and its duration is important, as are questions concerning similar gastrointestinal complaints in other family members and parental views on laxatives and bowel habit. Finally, a gentle but careful assessment for evidence of affective disorders, dysphoria, emotional distress (e.g., litigation, psychological counseling), and the use of mood-altering drugs helps to establish potential contributing factors to subjective complaints in the patient. Questionnaires such as the SCL-90-R [112] may be helpful in this regard; it takes less than 15 minutes to complete, is acceptable to most patients, and correlates well with the Minnesota Multiphasic Personality Inventory. These instruments should be used as an adjunct to careful history taking and not as substitutes for it. In children, inquiry should be made concerning nightmares, enuresis, school performance, and intrafamilial tensions. A history of bowel disturbances in the family should be elicited because there is an increased incidence of constipation and defecatory difficulties in one or both parents of children with encopresis. [113]

Physical Examination

Physical examination includes a search for evidence of nongastrointestinal diseases that can cause or exacerbate constipation. Particular attention should be given to a careful neurological examination, including an assessment of autonomic function and abdominal palpation for evidence of bowel distention, retained stool, or previous surgical procedures.

Anorectal and perineal examinations should search for perineal disease or deformity, abnormal location of the anal orifice, atrophy of the gluteal muscles, and rectal prolapse. Digital examination can elicit the pain of an anal fissure, detect a fixed stenosis of the anal canal, assess tone and strength of the anal canal at rest and with squeeze, or detect the presence of a rectal mass or fecal impaction. While the patient strains, the physician should look for the presence of rectosphincteric dyssynergia, a rectocele bulging anteriorly into the vagina, perineal descent, or rectal prolapse. Gaping of the anal canal when the examiner pulls the puborectalis muscle posteriorly or immediately on withdrawing the finger from the anal canal suggests denervation of the external anal sphincter. Perineal sensation should be assessed, and reflex contraction of the anal canal after pinprick of the perianal area (*anal wink*) also can be used to test neurological function of the perineal areas.

Studies of Colorectal Structure

Studies of colorectal structure are important to exclude organic disease. However, they provide little information about colonic and anorectal function and are

generally overused in patients with long-standing symptoms.

Flexible sigmoidoscopy is a technique to assess bowel mucosa and intralumenal characteristics. It can be used to identify lesions that narrow or occlude the bowel and can detect *melanosis coli,* a brown-black discoloration of the bowel mucosa that is produced by lipofuscin deposits in the lamina propria with long-term use of anthraquinone laxatives. [114] Flexible sigmoidoscopy also can be used to estimate bowel diameter and to detect the featureless characteristics of the distal bowel in patients with long-standing laxative abuse.

If patients complain of constipation of short duration or a recent change in bowel habit, *barium radiographs* are an important complement to sigmoidoscopy to detect organic causes and also to diagnose megacolon and megarectum. Contrary to studies that have found a poor relationship between rectal size on radiography and rectal elastic properties, [66] some investigators have found reasonably good correlation between these two parameters. [91], [102] However, barium enema provides limited information about colonic transit and motor function in most patients with chronic constipation. [115] On balance, studies of anorectal manometry and compliance provide more useful measurements of anorectal function than do barium studies in patients with functional constipation.

Barium radiographs do show the characteristic denervated bowel segment with proximal dilation of the colon in classic Hirschsprung disease. In such circumstances, bowel cleansing should not be ordered, so the characteristic changes will be accentuated. Complete filling of the colon is not necessary for diagnosis of this disorder. Plain films of the abdomen can be useful in the evaluation and treatment of chronic constipation. They can detect significant stool retention in the pelvic colon or throughout the entire colon, can suggest the diagnosis of megacolon, and can be used to monitor the adequacy of bowel cleansing of patients with fecal retention if there is uncertainty on physical examination.

Rectal biopsies are useful only in patients with suspected Hirschsprung disease. Suction biopsy samples should be obtained at least 3 cm above the distal portion of the internal anal sphincter to exclude the disease. For patients with idiopathic constipation, rectal and colonic biopsy samples with the use of endoscopic forceps are thought to be too superficial to be of clinical utility. However, some studies have demonstrated that mucosal levels of serotonin, [116] 5-hydroxyindoleacetic acid, [116] and substance P [117] are characteristically altered in patients with idiopathic constipation. If confirmed, analysis of biopsy specimens could be useful in the diagnosis and investigation of motor disorders of the colon.

Studies of Colonic and Anorectal Function

Function tests are usually reserved for patients with severe idiopathic constipation who fail to respond to relatively simple therapeutic measures (see section “Diagnostic Strategies”). They are also useful in defining the patterns of bowel function in various subgroups of patients with constipation, to identify potentially useful therapeutic strategies.

Colonic Transit Studies A colonic transit study is most useful in evaluating a patient with severe constipation whose major complaint is that of infrequent defecation. Although the methods vary somewhat, the principle of the test is the same. The patient ingests a high-fiber diet (20–30 g/d) while abstaining from the use of laxatives, enemas, and medications that may affect bowel function. Radiopaque markers are ingested, and their transit through the colon is monitored by abdominal radiographs until at least 80% of the markers have passed or a defined period of time has elapsed (usually 6 to 8 days). Transit is then correlated with bowel habit before and during the test. The original technique described by Arhan and colleagues [118] involved the ingestion of 20 radiopaque markers cut from a 5.25-mm (16-French) radiopaque Levin tube. Abdominal radiographs were obtained at 24-hour intervals; markers were counted in the right, left, and rectosigmoid colons, as defined by certain anatomic landmarks, and could be followed as they moved distally until expelled. The markers in each segment each day were totaled and multiplied by a factor of 1.2 to obtain segmental and total transit times. The original technique has been modified to reduce exposure to radioactivity and the number of patient visits to the radiology suite. Daily doses of markers are given for 2 days, [32] 3 days, [119], [120] or 6 days, and radiographs are obtained at various intervals (Fig. 43-1 and Table 43-3). Using fast-film techniques with up to 110 keV, radiation exposure can be significantly reduced and sufficient detail is obtained to identify the markers and anatomic landmarks.

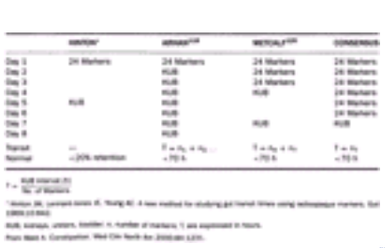

TABLE 43-3 Colonic Transit Studies

Notwithstanding differences in patients with respect to cultural practices, diet, and other factors that may affect colonic transit, studies of physiologically normal subjects on diets containing 20 to 30 g of fiber per day indicate an upper limit of normal for most adults of approximately 70 hours. [32], [119], [120] Although transit times differ between men and women, these differences are not important for clinical purposes. In addition, there are no significant differences between the two phases of the menstrual cycle, [32] which is an important consideration because women are more commonly evaluated for severe constipation. Despite some limitations of the test, [103] colonic marker studies are helpful instruments in evaluating patients with severe intractable constipation, and these tests have prognostic and therapeutic implications as well. Patients with normal colonic transit who complain predominantly of infrequent defecation may consciously or unconsciously misrepresent or misperceive bowel habit and, as a group, exhibit higher levels of psychological distress than do patients with slow transit constipation. [78] In contrast, patients with painless constipation and slow colonic transit have less psychological dysfunction. [78] Patients with the pattern of colonic inertia often respond poorly to therapeutic intervention. [121] In general, therefore, patients with slow transit may have a physiological basis to their symptoms, and those with normal transit may have a significant psychological or behavioral component to their complaints, although this view may be simplistic. A newer method measures colonic transit by scintigraphy. [79] This technique involves ingestion of a resin containing a radioisotope followed by nuclear imaging at various intervals. Although it appears promising as an investigational tool, it provides no more clinical information than does the conventional marker study. [122] In addition, the invasiveness of the procedure and the need for nuclear medicine facilities limit its clinical utility. Finally, one can assess bowel transit by collecting and examining stools radiographically after the administration of radiopaque markers or by inspection of the stools after ingestion of liquid markers. Because these techniques are unpleasant for both patients and technicians and can quantitate only total gastrointestinal transit rather than assess segmental colonic transit, they are not widely performed.

Anorectal Manometry Anorectal motility studies may provide useful information in many patients with severe constipation. The most useful parameters are rectal sensation, viscoelasticity, relaxation of the internal anal sphincter, and defecatory patterns produced on attempted expulsion of the apparatus. Satisfactory measurements of anal sphincter responses can be obtained with open-tipped perfused catheters, direct on-line pressure transducers, or air-filled balloons of various sizes and configurations. If rectal sensation is impaired, as in many patients with severe constipation, it is helpful to know whether impairment is associated with increased rectal compliance or megarectum. Compliance can be measured by sequentially inflating a balloon placed in the rectum and measuring pressures at each level of distention. Pressures should be corrected by subtracting those obtained by inflation outside the patient. The investigator should note the volume at which the first urge to expel the balloon occurs and also the symptoms elicited with increasing inflation of the balloon. Patients with constipation associated with irritable bowel syndrome often have low compliance and tolerate distention poorly, in striking contrast to patients with megarectum. [91] The absence of internal anal sphincter relaxation strongly suggests Hirschsprung disease; in the presence of megarectum, it is important to use larger volumes of rectal distention and to evacuate the rectum to avoid an inappropriate diagnosis of aganglionosis. Manometry has been used to assess anorectal patterns during attempted expulsion. Intrarectal pressures give some indication of intra-abdominal pressures generated during expulsion, whereas pressure recordings in the anal canal indicate relaxation or inappropriate contraction of the external anal sphincter or gluteal muscles. [70] Manometry can be supplemented by electromyographic (EMG) studies with the use of surface recordings of the external anal sphincter or by electrodes on the surface of an anal plug. However, concern has been expressed that results of studies performed in the laboratory may be primarily laboratory artifacts and that true dyssynergia is rare. [123]

Studies of Defecation Defecation can be evaluated by both radiographic and nonradiographic techniques, and these studies may be especially useful in patients who complain of excessive straining during defecation or who employ digital manipulation to facilitate evacuation. These tests vary in their complexity and in the information they provide, and each has potential limitations. [124] *Defecography* is a technique in which barium thickened to a consistency that approximates stool is introduced into the rectum. Evacuation of the barium is monitored by fluoroscopy and videotape while the patient sits on a specially constructed commode. Assessment of the anorectal structures, including the anorectal angle, are obtained at rest and during defecation; also, anatomic abnormalities such as rectoceles and intussusceptions that are not observed at rest may be apparent during defecation. However, emptying is semiquantitative, subjective, and liable to bias. It is probable that many patients feel embarrassed by the nature and setting of the procedure; such inhibitions can potentially result in an abnormal study. Defecography results

should be interpreted cautiously because anatomic abnormalities such as rectocele and intussusception are not uncommon in normal volunteers without constipation. [99], [125] More experience with this technique is needed before definitive decisions can be made on the basis of radiographic findings. [103] Scintigraphic techniques using radioisotope-labeled artificial stool can provide quantitative information concerning rectal emptying but provide little or no anatomic information. [8] Expulsion of water-filled balloons or solid spheres of different sizes and volumes [126] provides some information about expulsion but no information about anatomic changes or relations in the anorectum.

DIAGNOSTIC STRATEGIES

Most chronically constipated patients do not require extensive diagnostic studies beyond a careful history and physical examination and the appropriate exclusion of systemic or gastrointestinal causes for their complaints. Functional evaluation of the colon and anorectum should be reserved for patients who fail to respond to initial simple therapy and who express continued dissatisfaction with their bowel habit. Most patients with constipation-predominant irritable bowel syndrome do not require extensive diagnostic testing and rarely complain of persistent constipation beyond 3 to 6 months' duration.

The functional evaluation of chronic severe constipation begins with carefully defining the complaint and choosing studies that are most likely to yield diagnostic information concerning that complaint (Fig. 43-2). Intuitively, it may not seem important to measure colonic transit if a patient defecates several times per day but does so only with excessive straining or digital manipulation. Likewise, performing anorectal manometry would appear to add little to the evaluation of a patient who claims to have a bowel movement once every 8 days but has normal colonic transit. However, it has been shown that symptoms do not discriminate among physiological subgroups of patients with severe idiopathic constipation. [127] Thus, the workup in patients with severe constipation should be similar regardless of presenting symptoms.

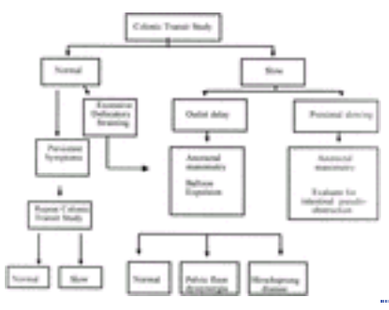

FIGURE 43-2. Algorithm for evaluation of chronic severe constipation that has not responded to dietary measures or fiber supplements. (From ref. [13].)

For the patient with infrequent defecation who fails to respond to initial therapy, a 2-week, prospectively obtained bowel diary with measurement of colonic transit time is the single most useful diagnostic study to obtain. [128] A normal study together with a defecation frequency within the normal range eliminates the need for further diagnostic tests of gastrointestinal function and may serve to reassure both physician and patient that colorectal function is not seriously impaired. In contrast, significant slowing of colonic transit suggests the need for additional studies to characterize various aspects of gastrointestinal motor function further (see Fig. 43-2).

Patients with *colonic inertia* usually have persistent complaints and respond poorly to medical therapy. [77] Anorectal manometry should be performed to characterize sensorimotor function of the bowel such as rectal sensation and viscoelasticity. Appropriate studies of upper gastrointestinal motor function should be obtained to look for evidence of gastrointestinal pseudo-obstruction (see Chapter 65 and Chapter 73) because such information may have both prognostic and therapeutic implications (see section " Colonic Inertia "). In contrast, the pattern of *outlet delay* suggests a more localized problem of the anorectum that can be further characterized by anorectal manometry, defecography, and a search for anatomic or functional abnormalities. The finding of normal anorectal function in a patient with outlet obstruction may suggest possible withholding behavior and is common in children.

For the patient who complains of *excessive defecatory straining* in the absence of an organic cause, both colonic transit studies and studies of anorectal function may be helpful. Normal anorectal motility and defecography may help to reassure patients that there is nothing seriously wrong from a functional standpoint; many of these patients exhibit high levels of psychological distress similar to that of patients with normal transit constipation. [129] The importance of anatomic abnormalities in such patients remains to be established.

TREATMENT CONSIDERATIONS

Much has been written about the treatment of constipation. There is general agreement that selecting therapeutic strategies requires understanding of the whole patient, fiber supplements should be added to the diet, establishing proper toileting arrangements can help certain patients, and long-term use of stimulant laxatives should be judicious. Treatment should be individualized, taking into consideration the age of the patient, duration and severity of constipation, potential contributing factors, and the patient's concerns and expectations. Although this approach is reasonable, there is little objective evidence to support it, in part because of the paucity of controlled trials and the heterogeneity of the population who consult for constipation. In addition, short-term results do not always reflect long-term outcome, and there is probably a significant placebo component to most therapeutic approaches. Broadly speaking, nonsurgical treatment can be separated into several categories: dietary approaches such as fiber supplementation; behavioral approaches such as habit training, contingency management, and biofeedback; and pharmacological approaches. Surgery has a role in selected patients with severe constipation in whom abnormal bowel function can be ameliorated by operative intervention.

Dietary Approaches

Dietary adjustments are the first line of intervention for most adults with constipation. Inadequate intake of dietary fiber is widely believed to contribute to constipation in industrialized nations because high fiber consumption in other parts of the world is generally associated with the daily passage of several bowel movements of considerable volume. [130] Indeed, increased consumption of dietary fiber by nonconstipated persons increases stool weight and frequency of defecation and decreases gastrointestinal transit time. [131] Although constipation appears to result from dietary fiber inadequacy in some persons, there is no evidence that constipated patients in general consume less fiber than do nonconstipated persons. [39] This is not to say that fiber supplementation should not be attempted. Many constipated patients do respond to increases in fiber intake to between 20 and 30 g per day, especially when the fiber is accompanied by water supplementation. [132]

Fiber components are not equivalent in their ability to modify stool characteristics and bowel habit. For example, wheat bran is most effective in increasing stool weight, followed by fruits and vegetables, oats, mucilages, corn, cellulose, soya, and pectin. [131] In animal studies, wheat bran accelerated colonic transit, whereas cellulose produced no change compared with fiber-free controls; pectin, sugar, and oat bran had variable effects on different regions of the colon. [133] The bulking effect of fiber is not so much related to water retention capabilities as to mechanical factors and colonic microbial ecology and interaction with intralumenal contents. Fiber may serve as a substrate for colonic bacteria and thus may increase stool bulk by proliferation of bacteria and production of gases that are trapped in the stool. [131], [134] Microbial breakdown products such as short-chain fatty acids also may stimulate colonic motility. [129] The net effect is increased stool bulk and shortened colonic transit in many persons. [135]

It is reasonable to recommend a high-fiber diet for all ambulatory adult patients who have constipation without megacolon (Table 43-4). It is of greatest help in patients with low fiber intake and in patients with constipation-predominant irritable bowel syndrome, in whom fiber intake should be increased gradually, to avoid undue cramping and bloating. [131] Fecal impactions should be removed before initiating fiber supplementation. Patients with obstructive lesions anywhere in the gastrointestinal tract should not be given fiber supplements. Fiber is not indicated in patients with megacolon or megarectum, especially if such patients are confined to bed, are demented, or have neurogenic constipation. Such patients are better managed by reducing colonic contents and instituting periodic timed evacuation.

	AMOUNT OF SERVING (g)	AMOUNT/100 g OF FOOD
Breakfast Cereals		
All-Bran	9.9	26.70
Corn Flakes	2.8	11.00
Rice Krispies	1.4	4.51
Shredded Wheat	3.0	12.31
Special K	1.7	5.41
Breads		
White bread	0.8	2.71
Whole wheat	2.4	8.51
Fruits		
Apple	3.2	1.41
Banana	5.9	1.71
Peach	2.1	2.38
Pear	3.1	2.41
Strawberry	3.3	2.12
Nuts		
Brazil	5.4	7.71
Peanut	5.7	9.31
Peanut butter	2.1	7.51
Vegetables		
Broccoli	5.6	4.11
Cabbage	1.9	2.81
Cauliflower	2.5	1.81
Lettuce	0.8	1.51
Carrot	3.7	3.20
Baked beans	18.6	7.31
Peas	11.3	6.31
Tomato	3.0	1.41

From Yang P, Banwell JG. Dietary fiber: its role in the pathogenesis and treatment of constipation. Pract Gastroenterol 1986;10:28.

TABLE 43-4 Food Sources of Dietary Fiber

Behavioral Approaches

Habit training and contingency management are often employed in children with idiopathic constipation with or without soiling. [63] The goal is to achieve regular evacuation, to prevent buildup of stool and fecal soiling. A similar approach may be useful in patients with neurogenic constipation, with or without soiling.

Before embarking on a behavioral approach, the patient's colon must be disimpacted and evacuated effectively. This can be accomplished with twice-daily saline or tap water enemas for 3 days and can be monitored by palpating the abdomen or obtaining an abdominal radiograph. Occasionally, a child may have to be hospitalized if cleansing cannot be achieved at home. Colonic evacuation may be accomplished by having constipated children drink a balanced electrolyte solution containing polyethylene glycol 3350 in amounts totaling 5 to 19 L. [136]

After bowel cleansing, polyethylene glycol, 8.5 to 34 g in 240 mL fluids or lactulose 15 to 30 mL/d, is given to produce at least one stool per day. In addition, the child is instructed to use the bathroom after breakfast or dinner to take advantage of meal-stimulated increases in colonic motility. If there is failure to defecate after 2 days, a cleansing enema is given to prevent recurrence of fecal impaction. After defecation occurs regularly for 2 to 3 months, weaning from the polyethylene glycol or lactulose is attempted gradually. Positive reinforcement is given for successful toileting, and punishment for failure is prohibited. Contact with the child and at least one parent should be maintained regularly.

These behavioral approaches have achieved success rates of up to 78% in children, [65], [137], [138] although relapses are not uncommon. [65], [139] The relative importance of each component of such strategies is unknown. [63] Treatment failures have been traditionally attributed to patient and family behavioral disturbances and noncompliance, but underlying disturbances of bowel function may play a role in some children. [68], [69], [70], [71], [72], [73], [74] and [75]

Another behavioral approach is the use of biofeedback to correct inappropriate contraction of the pelvic floor muscles and external anal sphincter during defecation. Studies have used EMG recordings concurrently with anal plugs [95] or anorectal manometers to monitor external and sphincter pressures during attempted expulsion of the apparatus. [96], [97] The patient watches the recordings of EMG or pressure responses and is told to modify inappropriate responses through trial-and-error efforts. Although this approach is useful in adults, a large study found that biofeedback therapy did not result in higher success rates than did conventional therapy alone. [76]

Despite the widespread belief that childhood encopresis is secondary to behavioral disturbances, the importance of behavioral factors is uncertain. Controversy centers on differences in the reported frequency and severity of behavioral disorders in encopretic children and whether they precede soiling or occur as a result of the underlying bowel dysfunction. Inconsistencies among various studies may be caused by differences in study populations: children with encopresis who were evaluated in psychiatric settings exhibited significant psychological dysfunction, whereas those referred to medical settings were less likely to do so. Several studies have reported only mild behavioral dysfunction and unremarkable toilet experiences in encopretic children treated in medical settings. [64], [137], [140] In contrast, others have reported more anxiety, less tolerance for demands, more submission in the face of peer aggression, less control of aggressive impulses, and more academic difficulties among encopretic children than among matched controls. Landman and colleagues [141] found that children with encopresis tended to feel less in control of positive life events and had lower self-esteem than children with other chronic illnesses such as recurrent abdominal pain, enuresis, or headache. Although other investigators made similar observations, they concluded that such personality characteristics developed because of negative experiences associated with soiling. [142], [143] Consistent with this interpretation, Levine [65] found that successfully treated encopretic children became better adjusted and exhibited no evidence of symptom substitution as long as 3 years after therapy. These observations imply that poor coping skills, low self-esteem, and passivity are potentially reversible with successful treatment of fecal soiling.

Pharmacological Therapy

The use of laxatives is deeply rooted in medical and social traditions, [144] and vast amounts are consumed in the Western world, [12] especially by elderly persons. [109] Laxatives are classified into five groups on the basis of their presumed mode of action (Table 43-5).

LAXATIVE	USUAL ADULT DOSE*
Bulk-forming Laxatives	
Natural (e.g., psyllium)	7 g/day
Synthetic (e.g., methylcellulose, polycarbophil)	4–6 g/day
Emollient Laxatives	
Mineral oil	15–45 mL/day
Hyperosmolar Laxatives	
Polyethylene glycol	8–25 g/day
Lactulose	15–30 mL/day
Sorbitol (70%)	15–30 mL/day
Glycerine	3 g suppository
Saline Laxatives	
Magnesium hydroxide	2400 mg (30 mL)
Magnesium citrate	200 mL
Stimulant Laxatives	
Castor oil	15–60 mL
Diphenylmethanes	65–130 mg
Phenolphthalein	30 mg
Bisacodyl	10 mg suppository
Anthraquinones	
Aloe (casanthranol)	30–60 mg
Cascara sagrada	2–5 mL
Senna	17–34 mg

* Oral except where indicated otherwise.

From Wald A. Constipation. Med Clin North Am 2000;84:1231.

TABLE 43-5 Laxatives for the Management of Constipation

The *bulk-forming laxatives* comprise natural (psyllium) or synthetic polysaccharides or cellulose derivatives that act in a manner similar to that of fiber naturally contained in the diet. Because fluid intake should be increased with these preparations, [132] they should be used cautiously in patients who require severe fluid restriction. Sugar-free bulk laxatives may contain aspartame (NutraSweet) and are contraindicated in patients with phenylketonuria.

Emollient laxatives consist of mineral oil and docusate salts. Mineral oil can be given orally or by enema; it penetrates and softens the stool. Although some mineral oil is absorbed and deposited in the liver, spleen, and mesenteric lymph nodes, [145] there are no described harmful effects. Because mineral oil may decrease the

absorption of fat-soluble vitamins A, D, and K, it should be administered between meals. Aspiration with lipid pneumonia is well described; therefore, mineral oil is contraindicated in patients with esophageal dysmotility or dysphagia and in elderly or debilitated patients and should not be given at bedtime. It is an unattractive agent in view of the availability of other effective products.

Docusate salts are anionic surfactants that lower the surface tension of stool to allow mixing of aqueous and fatty substances. This action softens stool to permit easier defecation. These agents also stimulate intestinal fluid and electrolyte secretion by increasing mucosal cyclic adenosine monophosphate. [146] Although not absorbed, they alter intestinal mucosal permeability and increase the absorption of other laxatives, such as mineral oil, phenolphthalein, and danthron. Placebo-controlled studies failed to demonstrate changes in stool water content, stool weight, frequency of defecation, or colonic transit times after administration of docusates in the recommended doses. [147]

Hyperosmolar agents include polyethylene glycol and nonabsorbable sugars such as lactulose and sorbitol. Sorbitol and lactulose are degraded by colonic bacteria to low-molecular-weight acids that increase stool acidity and osmolarity and lead to accumulation of fluid in the colon. Doses should be adjusted to reduce abdominal bloating and flatulence and to modulate defecation. Sorbitol and glycerin are given intrarectally and may produce rectal irritation. Polyethylene glycol electrolyte solutions are most often given for bowel cleansing before colonoscopy or before institution of bowel programs. One formulation consists of polyethylene glycol without electrolytes that is given in varying doses on a daily basis. Formulations with or without electrolytes have proven effective when given on a daily basis. [148], [149]

Saline laxatives contain relatively nonabsorbable cations and anions that exert an osmotic effect to increase intralumenal water content. Magnesium increases intestinal motor activity; because an appreciable amount of magnesium may be absorbed, these agents should be avoided in patients with renal insufficiency because of the danger of magnesium toxicity. [150] Other side effects include hypocalcemia in children and mineral imbalances. Saline laxatives can also be administered by enemas or suppositories.

Stimulant laxatives consist of castor oil, anthraquinones (cascara sagrada, senna, casanthranol, and danthron), and diphenylmethanes (phenolphthalein and bisacodyl). Castor oil is hydrolyzed by intestinal lipases to ricinoleic acid, which stimulates intestinal secretion, decreases glucose absorption, and increases intestinal motility. [151]

The anthraquinone laxatives increase fluid and electrolyte accumulation in the distal ileum and colon through incompletely understood actions. [12] All are absorbed from the small intestine to some extent (especially danthron) and are metabolized by the liver. Anthraquinones are converted to a pharmacologically active state by contact with intestinal microorganisms. Pathological changes in the colon produced by long-term anthraquinone use include melanosis coli, a benign and reversible condition. Although smooth muscle atrophy and damage to the myenteric plexus were suggested in earlier studies, [152] there is no evidence that anthraquinones given in clinically relevant doses cause enteric damage in either experimental animals or humans. [58], [59]

Approximately 15% of phenolphthalein is absorbed from the small intestine and undergoes an enterohepatic circulation, which explains its often long duration of action. It acts directly to stimulate colonic motor activity and inhibits glucose and sodium absorption to increase intralumenal fluid content. Side effects include fixed-drug skin eruptions, erythema multiforme, and photosensitive bullous skin lesions. Fatalities have been reported in allergic patients who were rechallenged with the drug. [144]

In 1997, the United States Food and Drug Administration (FDA) reclassified phenolphthalein as “not generally recognized as safe and effective” after a study in rodents found an increased incidence of nongastrointestinal neoplasms. [153] Subsequently, most phenolphthalein-containing laxatives were voluntarily withdrawn in the United States. However, these animals were given doses 10 to 1000 times the human dose, in terms of body surface area. Studies of persons who use laxatives have not suggested that the risk of these cancers is increased in humans. [154] Similarly, concerns about a possible relationship between melanosis coli and the development of colonic neoplasms [155], [156] were not substantiated in a prospective case control study. [157]

Bisacodyl is structurally similar to phenolphthalein and exhibits similar actions on small intestine fluid accumulation and colonic motor activity. [80] Because the drug is a gastric irritant, tablets are enteric coated and should not be broken or chewed. [144]

The use of drugs to enhance colonic transit by increasing propulsive motor activity has been hampered by limited knowledge of the control of various aspects of colonic motility (see Chapter 11) and the pathophysiology of severe idiopathic constipation. *Cholinergic agents* such as bethanechol have been used with little success and exhibit moderate side effects in patients with colonic inertia. *Cholinesterase inhibitors* such as neostigmine may have fewer side effects, but there is no evidence of increased efficacy, [40] except in acute megacolon. [153]

Prokinetic agents are drugs that stimulate gastrointestinal motor activity to enhance transit of intralumenal contents. Metoclopramide has been used to treat upper gastrointestinal motor disorders but exerts little effect on colonic motility and is not effective in constipated patients. [13] Cisapride appeared to enhance transit through the proximal colon and has been shown to stimulate colonic motility and improve rectal sensation in chronically constipated patients. [154], [155] Although some studies suggest that clinical improvement occurred with the drug, [154], [155] and [156] others report highly variable and often disappointing results in patients with severe idiopathic constipation. Cisapride was withdrawn by the FDA because of the risk of cardiac toxicity. Some patients with severe constipation have been treated successfully with misoprostol, a prostaglandin used to prevent peptic ulcers associated with nonsteroidal antiinflammatory drugs. [158], [159] Another small study reported that colchicine (0.6 mg three times daily) was effective in patients with refractory constipation. [160] Further studies are needed to assess the possible efficacy of these agents in severely constipated patients.

There has been intense interest in the pharmacological actions of serotonin (5-hydroxy-tryptamine [5-HT]) agonists on gastrointestinal motility. 5-HT $_4$ agonists stimulate intestinal motility, in part by facilitating enteric cholinergic transmission. [161], [162] Clinical trials indicate that several 5-HT $_4$ receptor agonists accelerate colonic transit time and improve symptoms in patients with constipation predominant irritable bowel syndrome or constipation. [163], [164]

It has been hypothesized that motility disorders leading to chronic constipation are caused by excessive endogenous opioids. Several patients with severe constipation have been treated successfully with intravenous naloxone, an opioid receptor antagonist. [165] As shown by scintigraphic techniques, naloxone accelerates transit in various parts of the colon in normal volunteers. [166] Preliminary studies suggest that oral naloxone, compared with placebo, may increase stool volumes in elderly constipated persons. [167] Such agents have been used successfully to treat constipation associated with long-term use of narcotic agents. [168]

Surgical Treatment

With several important exceptions, the indications for surgical intervention in managing patients with chronic constipation are somewhat controversial. The generally agreed indications and contraindications for surgery are discussed in the next section.

Hirschsprung Disease Surgery is the treatment of choice for all forms of this disease but varies according to the length of the aganglionic segment. In patients with short-segment or ultrashort-segment disease, anal myotomy, in which the internal sphincter and a varying length of rectal smooth muscle are incised, can be very beneficial. [169], [170] In the classic form of the disease, more extensive procedures are used to overcome the obstructing effect of the aganglionic segment. These include the following: removal of the segment, as in the Swenson operation [171]; bypassing it, as in the Duhamel operation [172]; or using the endorectal pull-through techniques of Soave [173] and Boley. [174] The choice of surgical technique depends on the surgeon, but excellent results have been reported for all of them. [175], [176] For good results to be obtained, thorough cleansing of the colon before surgery is mandatory. [170]

Colonic Inertia In selected patients with colonic inertia, subtotal colectomy with ileorectal anastomosis can be dramatically beneficial in ameliorating incapacitating symptoms that are unresponsive to medical management. [177], [178] and [179] Limited resection of the colon, however, produces unsatisfactory results and a high rate of anastomotic leaks. [177] Radiologic or manometric studies should be performed preoperatively to document that the disorder is confined to the large intestine and to establish normal esophageal, gastric, and small intestine motor function. Nevertheless, abnormal gastroduodenal manometry does not necessarily predict failure of colectomy, [180] and normal studies do not guarantee success. Patients who have evidence of a more extensive dysmotility disorder can be anticipated to have less satisfactory results, [181] as do those with coexisting psychiatric disorders. [182] Such procedures should be done only if colonic distention is life-threatening or severely incapacitating. There is a fairly high complication rate, and patients with bloating and abdominal pain are less likely to have improvements with surgery. [183]

Rectocele Surgical therapy consists of reducing or eliminating the pouch. Because rectoceles are common in nonconstipated persons, care must be taken before attributing defecatory problems to this entity. Ideally, one should observe improved rectal evacuation after the patient exerts finger pressure on the posterior wall of the vagina during defecography. Rectocele repairs often do not alleviate symptoms of difficult defecation.

Rectal Intussusception and Prolapse Surgical therapy consists of various resuspension procedures and abdominal rectopexy with sigmoid resection. [170] However, most patients who undergo surgery experience no improvement in their defecatory problems. [184] As with rectoceles, rectal intussusceptions are not uncommon in nonconstipated persons, and their presence in constipated patients should not imply causation.

Rectosphincteric Dyssynergia Surgery for this disorder is contraindicated because patients receive no benefits from posterior division of the puborectalis muscle, and fecal incontinence commonly occurs postoperatively. [185]

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CHAPTER 44

Raphael B. Merriman and Marion G. Peters

APPROACH TO THE PATIENT WITH JAUNDICE

Jaundice (icterus) is the yellow discoloration of skin, sclera, and mucous membranes caused by the excessive accumulation of bilirubin pigments. It is usually apparent when bilirubin levels exceed 3 mg/dL or 50 µmol/L. [1] Hyperbilirubinemia results from one or more derangements in the complex multistep process that balances bilirubin production with bilirubin clearance. Consequently, jaundice can indicate a disorder in bilirubin metabolism, hepatic function, or biliary disease or combinations thereof. The optimal approach to the patient with jaundice demands precise localization of the site of disordered bilirubin handling and appropriate diagnostic tests. A review of the causes and management of jaundice logically begins with a clear understanding of the structure, formation, uptake, and conjugation of bilirubin, its clearance through the liver, and its subsequent excretion into the biliary tree and gastrointestinal tract.

Cholestasis is characterized by the constellation of physiological, morphologic, and clinical manifestations that result from the impairment of the bile excretory system in the liver and biliary tree. [2], [3] Reduced bile flow resulting in the accumulation of conjugated bilirubin, bile salts, and cholesterol in the blood is the hallmark of cholestasis. However, an impairment of organic ion transport, decreased excretion of biliary lipids, a release of liver plasma membrane enzymes, and a decrease in intestinal bile also occur. In patients with cholestasis, total serum bilirubin may be normal while serum alkaline phosphatase and bile acids are elevated. [4] *Bile acids* or bile salts are soluble, amphipathic end products of cholesterol metabolism formed in the pericentral hepatocytes and accounting for approximately 85% of the constituents of bile. [5] The primary bile acids (formed in the hepatocyte) chenodeoxycholic and cholic acids are the most common. The secondary bile acids are formed by anaerobic bacterial dehydratase conversion of primary bile acids to lithocholic and deoxycholic acids, respectively. Primary and secondary bile acids account for 99% of the bile acids in human bile. Another bile acid, ursodeoxycholic acid, so termed after its isolation from bile of the polar bear, is present in trace amounts. The bile acids' hydroxyl groups are water soluble and thus hydrophilic, whereas the opposite pole is relatively hydrophobic, giving bile acids their detergent properties. The hydrophilic activity of bile salts increases as the number of hydroxyl groups increases. Thus, lithocholic acid is more hydrophobic than chenodeoxycholic acid and cholic acid, with ursodeoxycholic acid the most hydrophilic. Bile acids are absorbed in the distal small intestine and are returned to the liver mostly protein bound and resecreted, constituting the enterohepatic circulation. The bile acid pool of 2 to 3 g in adults cycles several times with each meal, and although bile acid synthesis averages 0.3 g/d, total bile acid secretion is 12 to 18 g/d. Bile acids are critical for cholesterol elimination, by conversion of cholesterol to bile acids, by stimulation of biliary cholesterol and phospholipid secretion, and by micellar solubilization of cholesterol and phospholipids within bile, ultimately facilitating fecal elimination. Bile acids also induce bile flow, and through negative feedback, they can inhibit bile and cholesterol biosynthesis. Bile is a major conduit for the elimination of other endogenous and exogenous pigments including porphyrins and urobilins, in addition to the elimination of most rare metals (copper, zinc, manganese, chromium, selenium, molybdenum, and cadmium and organic anions). [6] In the small intestine, bile acids promote dietary lipid absorption through the formation of mixed micelles.

BILIRUBIN

Structure and Formation

Bilirubin, a yellow tetrapyrrole pigment, is the principal end product of the degradation of the heme moiety of hemoglobin and other hemoproteins. The catabolism occurs largely within the reticuloendothelial cells of the liver, spleen, and bone marrow. The heme iron protoporphyrin IX ring is opened by oxidation of the a-bridge carbon by heme oxygenase, present in many cell types, particularly Kupffer cells, macrophages, hepatocytes, and renal epithelial cells. The reaction forms biliverdin, a green tetrapyrrolic pigment, and equimolar quantities of carbon monoxide, which binds to hemoglobin and is excreted through the lungs (Fig. 44-1). Biliverdin is then rapidly reduced to bilirubin by biliverdin reductase. Bilirubin IXa, derived from protoporphyrin isomer IX with cleavage at the a bridge, is the predominant bile pigment in plasma, where its concentration generally does not exceed 1 mg/dL (17µmol/L) in healthy people. [7] Hydrogen bonds stabilize the molecule in a folded, ridge-tile conformation restricting exposure of the molecule's polar groups to aqueous solvents. These structures form the basis of bilirubin's hydrophobic behavior and slow (indirect) diazo reactivity. [8] Opening of the heme ring at non–a-carbon positions would lead to isomeric pigments that are more water soluble and do not require conjugation for efficient excretion in bile and urine. However, although heme catabolism generates almost exclusively a single form of bilirubin, other stereoisomers are formed from this by exposure to light as the pigment circulates through peripheral tissues. Being more polar, these photoisomers do not require conjugation for excretion in bile. Their formation is clinically important in phototherapy of neonatal jaundice, in which they accelerate bilirubin elimination but otherwise have no known significance. [8] With the exception of bilirubin IXß found in meconium, significant quantities of these isomers are not detectable.

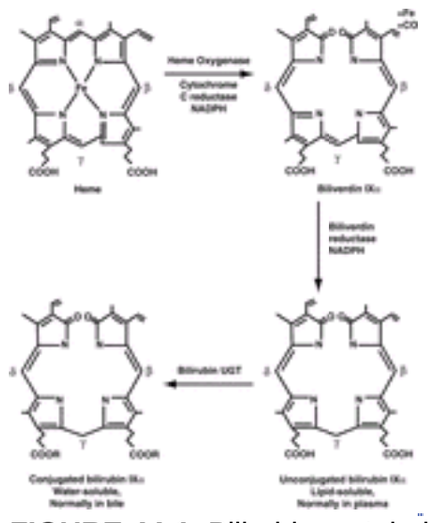

FIGURE 44-1. Bilirubin metabolism. The first step in the degradation of the heme group to bilirubin is the cleavage of its a-methene bridge to form biliverdin, a reaction catalyzed by heme oxygenase. The enzyme is a monooxygenase, and O_2 and NADPH are required for the cleavage reaction. The reaction releases CO, iron, and biliverdin. The CO binds to hemoglobin and is excreted through the lungs, and most of the iron is reincorporated into new heme proteins. The second step in heme catabolism is the reduction of biliverdin IXa to bilirubin IXa by biliverdin reductase and NADPH. The result is unconjugated bilirubin. After bilirubin IXa is formed, it is released into the bloodstream, where it rapidly binds to plasma proteins, especially albumin. These complexes are poorly filtered by the glomerulus. Bilirubin is rendered more soluble in the liver by conjugation: the attachment of sugar residues such as glucuronates to its propionate side-chains. This reaction requires the enzyme bilirubin UDP glucuronosyltransferase (bilirubin UGT).

Most bilirubin (80%) derives from senescent and sequestered erythrocytes. [1] The remainder is largely derived from the degradation of hepatic and renal heme, which constitutes a cytosolic pool of heme destined for incorporation into specific heme proteins including cytochrome P450, peroxidase, and catalase. One third of the bilirubin derived from the hepatic heme proteins passes directly into bile as conjugated bilirubin, and the rest refluxes into plasma. [9] Myoglobin heme, because of its slow turnover, contributes less than 1% to bilirubin formation. Normally, less than 3% of bilirubin is derived from ineffective erythropoiesis in the bone marrow, but this fraction may be considerably increased in hemoglobinopathies, congenital and acquired dyserythropoietic anemias, and megaloblastic anemia. Normal daily human bilirubin production averages 4 mg/kg body weight, equivalent to approximately 250 mg of bile pigment.

Unconjugated bilirubin is normally insoluble in water. In bile, however, it is solubilized by weak interactions with mixed micelles of bile salts and other lipids. [6] When the capacity for bilirubin solubilization is exceeded in bile, the unbound pigment may precipitate as a calcium salt and may form bilirubin pigment stones. Most bilirubin formed peripherally is transported in plasma tightly bound to albumin. The concentration of unbound bilirubin remains very small until the bilirubin concentration exceeds 35 mg/dL. [10] Increased unbound bilirubin pigment may cross cell membranes resulting in tissue injury, a key factor in the pathogenesis of bilirubin encephalopathy and kernicterus in the newborn. [11] In its albumin-bound state, bilirubin is poorly filtered at the glomerulus, limiting its renal elimination. Therefore, the conjugation of bilirubin by hepatocytes facilitates the excretion in bile of a potentially toxic substance.

Uptake, Binding, and Conjugation

The hepatocyte has evolved specialized mechanisms to facilitate the uptake of the nonpolar, tightly albumin-bound molecule across the sinusoidal membrane, to maintain solubility by binding to cytosolic proteins and to convert it to more polar compounds by conjugation, permitting subsequent excretion. Initially, albumin spontaneously dissociates from its bilirubin ligand, the principal determinant of which is the unbound bilirubin concentration. [12] Subsequent hepatocyte uptake of unconjugated bilirubin is often considered a facilitated process involving several transporters, possibly including liver-specific forms of the organic anion transporting polypeptide (OATP) family, a sodium-independent transport system, [13] although the identity of bilirubin transporters for uptake remains controversial and uncertain. Once inside the hepatocyte, hydrophobic bilirubin is maintained in solution by binding nonenzymatically to the cytosolic fatty acid binding proteins and ligandins (isoforms of proteins of the glutathione *S*-transferase gene family [GST]). [14] GSTs are important in creating intracellular gradients, in determining bilirubin's movement, in compartmentalization, and in presentation to microsomes for conjugation.

The conjugation of bilirubin in the endoplasmic reticulum is the physiological process whereby bilirubin is rendered soluble and more hydrophilic for ultimate excretion into the canaliculus. Conjugation is achieved by the esterification of one or both of the propionic side-chains through the addition of polar groups. Glucuronic acid esterification predominates, but xylose or glucose esterification can occur. Bilirubin is conjugated with uridine 5'-diphospho (UDP)-glucuronic acid by bilirubin UDP-glucuronosyltransferase (bilirubin UGT, also known as UGT-1A1). This produces bilirubin monoglucuronides and subsequently the diglucuronide. Normal human bile principally contains bilirubin diglucuronide (70%–90%) and smaller amounts of monoglucuronides (5%–25%). Their proportions change when the ratio of unconjugated bilirubin to available enzyme is increased (with an increased bilirubin load in hemolysis or reduced enzyme activity in Gilbert syndrome), favoring the formation of monoglucuronides. [15] It is becoming apparent that there are many distinct UGT isoforms. However, in humans, conjugation of bilirubin is exclusively carried out by the substrate specific microsomal enzyme, UGT-1A1 encoded by the UGT-1 gene subfamily that is located on chromosome 2. [16] Apart from bilirubin, other substrates for glucuronidation include drugs such as testosterone, estradiol, 1,25-dihydroxyvitamin D_3, digoxin, and propranolol, in addition to thyroid hormones and catecholamines. Understanding of the sequence of bilirubin UGT and subsequent identification of mutations have facilitated the understanding of several disorders characterized by deficient bilirubin conjugation, such as Gilbert syndrome and Crigler-Najjar syndrome.

Clearance and Excretion

Hyperbilirubinemia results from a combination of increased bilirubin turnover or decreased bilirubin clearance. Isotope labeling studies permit an estimation of daily hepatic bilirubin clearance but are available only in a research setting. [7] Hepatic extraction of unconjugated bilirubin from plasma is 4.6 µg/min/kg under normal conditions. Almost 40% refluxes unaltered back into plasma, as does a small proportion of esterified bilirubin. The proportion of unconjugated to conjugated can change from more than 9:1 in nondisease states to 1:1 in cholestatic disease.

The generation of bile flow is highly regulated and involves the coordinated action of several transporter proteins located at canalicular and sinusoidal domains in the hepatocyte. The canalicular ATP-dependent export pumps include the multidrug resistance 1 P-glycoprotein (MDR1), the phospholipid multidrug resistance 3 P-glycoprotein (MDR3), the canalicular bile salt export pump (BSEP), and the canalicular multispecific organic anion transporter (MRP2 or cMOAT), all members of the ATP binding cassette family, in addition to several putative ATP-independent transport systems. More recent advances have provided an enhanced understanding of these canalicular excretory processes. [13] The transporters are the rate-limiting step in bile salt export. Conjugated bilirubin is excreted across the canalicular membrane by a saturable process capable of functioning against a concentration gradient of 40- to 100-fold. Bilirubin monoglucuronides and diglucuronides are excreted into bile by the transporter MRP2. This carrier is also responsible for excreting leukotriene 4, some hydrophobic bile acids, oxidized glutathione, indocyanine green, sulfobromophthalein, and cholecystographic contrast. Definitive evidence of a potential sensitive membrane transporter is currently lacking.

After conjugated bilirubin reaches the bile ducts, it flows into the gallbladder, where sodium, bicarbonate, and water are actively absorbed. Cholecystokinin coordinates gallbladder contraction and sphincter of Oddi relaxation, permitting the controlled flow of bile into the small intestine to participate in the digestive process. Conjugated bilirubin is not absorbed from the gastrointestinal tract. However, it may be deconjugated and degraded by bacterial ß-glucuronidases and other enzymes in the terminal ileum and colon to form a large variety of urobilinogens. Some urobilinogen is reabsorbed from the colon resulting in small plasma concentrations. Most of this is re-excreted by the liver, although the kidney filters a small proportion. Increased urine urobilinogen may represent increased bilirubin formation and subsequent enterohepatic circulation of urobilinogen or, alternatively, decreased hepatic clearance of urobilinogen and therefore does not distinguish between hemolysis and liver disease. [17] In circumstances of complete biliary obstruction, jaundice may be apparent in the absence of urobilinogens because of the lack of delivery of bilirubin to the intestine. Urine urobilinogen levels may rise before jaundice becomes clinically apparent. Because unconjugated bilirubin is very tightly bound to albumin, the free fraction is very small and consequently never appears in the urine. Bilirubin conjugates are much less tightly bound to albumin. Therefore, in the presence of cholestasis, bilirubin conjugates formed in the hepatocyte are diverted back into the circulation, and the larger unbound fraction is filtered in the kidney. Thus, urine bilirubin is an absolute indication of conjugated hyperbilirubinemia. In the neonate, increased ß-glucuronidases and deconjugation may contribute unconjugated bilirubin to the physiological jaundice of the newborn. In the colon, the unstable urobilinogens are oxidized to urobilins, orange pigments that are excreted in the feces. Bilirubin itself is rarely found in human feces unless intestinal flora is underdeveloped or altered by antibiotics. Although the reticuloendothelial system has the capacity to metabolize heme from hemoglobin to form up to 1500 mg of bilirubin daily, a substantial increase in hemoglobin

turnover is necessary before an increase in bilirubin levels is apparent. Consequently, jaundice seldom results from hemolysis unless it is severe or accompanied by hepatic dysfunction. [18]

Measurement

The upper limit of normal for total plasma bilirubin concentration is 1.0 to 1.5 mg/dL (17 to 26 µM), with indirect-reacting bilirubin 0.8 to 1.2 mg/dL (14 to 21 µM) and direct-reacting bilirubin usually less than 0.2 mg/dL (largely a technical artifact); the ranges reflect variations in analytical methods. [19] The distribution of plasma bilirubin in the population is not normal, but it is skewed toward the upper end values. Males have slightly higher values than females. Unconjugated bilirubin IXa accounts for 95% of circulating isomers, and bilirubin monoesters and diesters account for about 4%. [20]

Diazo Method Accurate measurement of total bilirubin and its fractions in serum is a crucial first step in the approach to a patient with jaundice. [21] The formation of diazo derivatives of bilirubin, as originally described by van den Berg in 1913, provides the basis for most clinical methods of quantifying bilirubin. [7] The addition of the diazo reagent (e.g., diazotized sulfanilic acid) ultimately resulted in the generation of two azopyrromethane molecules from the tetrapyrrole, the amount of which formed could be determined spectrophotometrically. Van den Bergh and Müller first noted in 1916 that conjugated bilirubin reacted rapidly with the diazo reagent. The direct-reacting bilirubin (a proxy for conjugated bilirubin) and total bilirubin were measured, and indirect-reacting bilirubin (a proxy for unconjugated bilirubin) was then calculated by subtraction. However, it became apparent that measurements of the two pigment fractions lacked specificity and often did not reflect the actual concentrations of conjugated and unconjugated bilirubins for many technical reasons, including the exquisite sensitivity of the reaction conditions to small changes in pH, light exposure, and storage temperature. This was especially a problem when accurate pigment fractionation was most needed, that is, at low concentrations of total bilirubin (<5 mg/dL), to differentiate hemolysis from unconjugated hyperbilirubinemia.
Delta Bilirubin Glucuronides are chemically reactive metabolites. In adults, with prolonged cholestasis, especially if it is accompanied by renal failure, a fraction of the bilirubin conjugates reacts nonenzymatically and irreversibly with serum albumin to give yellow biliproteins in which bilirubin is covalently bound to albumin. [22] This fraction increases with the duration of cholestasis. Designated *delta bilirubin,* it retains its direct-reacting properties. Chromatographic studies had indicated the presence of this fourth form of bilirubin in serum, and its existence was subsequently confirmed after separation by high-performance reversed-phase liquid chromatography. [23] Because delta bilirubin is irreversibly bound to albumin, it is not filtered at the glomerulus, and its half-life reflects that of albumin (14 days). Consequently, it remains in plasma long after other bilirubin esters have been eliminated. Clinically, this is suggested by persistent conjugated hyperbilirubinemia in the absence of bilirubinuria and is particularly evident in the recovery phase of cholestasis. [24] Delta bilirubin can be calculated as follows: delta bilirubin = total bilirubin - (conjugated + unconjugated bilirubin).
Dry Film Method The adoption of dry film technology represented a major advance in accuracy and is now standard in most clinical laboratories. [25] Using the Ektachem or Fuji dry chemistry slides, diazo coupling proceeds on a buffered porous layer, and the azo derivatives become bound to a cationic polymeric mordant, resulting in a spectral change. Measurements are made at two different wavelengths to account for spectral interferences. Protein-conjugated bilirubin is retained on only one of the slides and therefore can be removed from the sample being measured. One slide is used to measure total bilirubin, and a second slide measures both unconjugated bilirubin and conjugated bilirubin, the difference being delta bilirubin. The test is accurate, reproducible, and amenable to automation, and the method correlates well with high-performance reversed-phase liquid chromatography for the fractionation of serum bilirubin species.

HYPERBILIRUBINEMIA

Hyperbilirubinemia results from a disruption at one or more steps in the complex metabolic pathways previously described, extending from excess bilirubin formation, altered uptake, defective conjugation, and diminished excretion into the biliary tree and subsequently into the gastrointestinal tract. Bilirubin levels correlate directly with bilirubin production and inversely with hepatic bilirubin plasma clearance. Practically, hyperbilirubinemia is classified as excess of either conjugated (direct-reacting) or unconjugated (indirect-reacting) bilirubin. This distinction is crucial to guide the evaluation of the patient, the differential diagnoses to be considered, and subsequently the optimal investigative approach. A pure increase in unconjugated bilirubin is usually indicative of defective conjugation. A parallel increase in both fractions in proportion to normal is usually indicative of increased pigment production. A normal or increased unconjugated bilirubin level with a markedly increased conjugated bilirubin level occurs in hepatobiliary disease. [20] Mixed conjugated and unconjugated hyperbilirubinemia may occur in the setting of multiple transfusions postoperatively, in fulminant liver failure with hemolysis resulting from Wilson disease, and in bacteremia. Many patients with chronic hemolytic disorders such as sickle cell anemia or thalassemia have underlying liver disease from transfusion-related iron overload or transfusion-related viral hepatitis, resulting again in a mixed pattern of hyperbilirubinemia.

Unconjugated Hyperbilirubinemia

Overproduction of bilirubin may occur with excess destruction of red cells, with ineffective erythropoiesis, or with increased catabolism of heme compounds in the liver. Hemolysis may be intravascular (heme in plasma), extravascular (in cells of the reticuloendothelial system), or a combination of both. The bone marrow is capable of an eightfold increase in erythrocyte production in times of chronic hemolysis, with a maximum achievable steady-state bilirubin turnover of approximately 40 mg/kg/d, resulting in a serum unconjugated bilirubin level of 4 mg/dL. [19] With normal hepatic function, a 50% reduction in erythrocyte survival does not result in jaundice. [26] Even in the context of more severe hemolysis, unconjugated bilirubin levels rarely exceed 4 to 5 mg/dL unless there is concomitant impairment of hepatic clearance.

Hemolysis and Ineffective Erythropoiesis Hemolysis and ineffective erythropoiesis are the only relevant causes of bilirubin overproduction in humans. Hemolysis, even if severe, usually results in mild hyperbilirubinemia (<5 mg/dL) unless there is coexistent liver disease, which may contribute to concomitant conjugated hyperbilirubinemia. Hemolytic anemia may result from inherited disorders of the red cell membrane (sickle cell anemia), of hemoglobin (thalassemia), or of red cell enzymes (glucose-6-phosphate dehydrogenase or pyruvate kinase deficiency). Acquired hemolytic conditions may result from ABO blood group incompatibility, iron or vitamin B_{12} deficiency, and lead toxicity. Ineffective erythropoiesis shortens red cell life span and can be associated with hyperbilirubinemia. Impaired hepatic blood flow from congestive heart failure, cirrhosis, and portacaval shunting decreases bilirubin delivery to the liver and decreases clearance with resulting increased unconjugated hyperbilirubinemia. Other causes include hypothyroidism and hyperthyroidism, sepsis, and drugs (e.g., isoniazid, a-methyldopa, phenothiazines, nonsteroidal antiinflammatory drugs, rifampicin, sulfonamides, thiazides, and ribavirin).
Bilirubin Uridine Diphosphate Glucuronosyltransferase Deficiencies These disorders are characterized by abnormalities of the bilirubin UGT enzyme UGT-1A1 and consequently are manifest by unconjugated hyperbilirubinemia. Whereas Gilbert syndrome is common, the other disorders remain rare, but their study has provided invaluable insights into the complexities of bilirubin metabolism.
Gilbert syndrome. This condition is a characterized by unconjugated hyperbilirubinemia occurring in the absence of overt hemolysis or liver disease. [27] Typically, there are mild, chronic, or intermittent episodes of jaundice that increase with intercurrent illness, fasting, stress, fatigue, menses, and ethanol and nicotinic acid intake. Three to 8% of the population is affected, with a male predominance. It is usually inherited in an autosomal recessive pattern and is rarely manifest before puberty. Patients are generally asymptomatic, and nonspecific symptoms of fatigue and abdominal pain do not correlate with bilirubin levels. Mildly lemon-tinged sclera are usually the only physical findings. About 50% of patients may have mild hemolytic anemia of uncertain origin. [27] Serum bilirubin concentrations are usually higher than 3 mg/dL but may increase to 5 to 8 mg/dL during stress. However, bilirubin levels may fluctuate in anyone, and it may be difficult to differentiate these from the higher end of the normal spectrum. Other hepatic biochemical indices are normal. A presumptive diagnosis can be made in a patient with isolated, asymptomatic, unconjugated hyperbilirubinemia if a careful history and physical examination do not suggest alternate diagnoses. Provocative fasting studies to confirm the diagnosis are seldom indicated or useful. Routine use of liver biopsy is not indicated. The significance of reports of increased lipofuscin pigment within the centrilobular cells is uncertain. Fasting serum bile acid levels are normal, in contrast to the hyperbilirubinemia of liver disease. Bilirubin UGT activity is reduced to approximately one-fourth of normal. [28] Patients have reduced bilirubin diesters and increased bilirubin monoglucuronides. Molecular studies have indicated that the phenotype results from different mutations. One mutation is an expansion of thymidine-adenine (TA) repeats in the promoter region of the *UGT-1A1* gene (autosomal recessive) for which homozygosity is predicted in 16% of whites. Another mutation is found in exon 1A1 of the same gene, occurring in Japanese and Asian populations (autosomal dominant negative), and is associated with serum bilirubin levels in the range of 3 to 10 mg/dL. [29] The genetic variation described in Gilbert syndrome may lead to pharmacological variation in drug glucuronidation and may explain the occasional unexpected drug toxicities reported. [29], [30] and [31] The use of pharmacological agents that induce hepatic microsomal enzymes such as phenobarbital, which normalizes plasma unconjugated bilirubin levels and correcting ratios of monoesters and diesters but does not increase UGT-1A1 activity, leaves questions regarding phenobarbital's true mode of action. Gilbert syndrome is a benign condition, and no therapy is routinely required.
Crigler-Najjar syndrome. This syndrome is characterized by unconjugated hyperbilirubinemia, which may occur from birth. [27], [32] Type I is a rare autosomal recessive disorder and is characterized by a complete deficiency of the enzyme bilirubin UGT. It results from a mutation in one of the five exons of the gene encoding bilirubin UGT. Mutations may occur in exon 1A1 or exons 2 to 5 of the *UGT-1* gene locus encoding the enzyme UGT-1A1, resulting in complete absence of the enzyme in type I. Crigler-Najjar type I bile contains only traces of bile conjugates. Heterozygotes have normal bilirubin levels. Infants develop severe unconjugated hyperbilirubinemia within 7 days of birth, followed by kernicterus and bilirubin encephalopathy if prompt therapy is not initiated. Death follows within 18 months if untreated. Treatment

consists of the rapid initiation of phototherapy with exchange transfusions. Type I does not respond to phenobarbital. Orthotopic liver transplantation is curative. Reports have indicated temporary success with isolated hepatocyte transplants. [33] Type II (Arias disease) is an autosomal dominant disorder with variable penetrance that results in various levels of unconjugated hyperbilirubinemia. Type II is caused by a variety of mutations in exons 1A1, 2, and 5 of the *UGT-1* gene locus, the functional consequence of which is partial inactivation of bilirubin UGT. Affected persons have about 10% of normal bilirubin UGT activity. Bilirubin monoconjugates and even some diconjugate are present in bile. Jaundice often is not apparent until the second year of life. Bilirubin levels rarely exceed 20 mg/dL. Therapy with phenobarbital results in a decrease in serum bilirubin by 30% but is usually unnecessary unless there is a risk of kernicterus.

Conjugated Hyperbilirubinemia

Conjugated hyperbilirubinemia is defined by a conjugated bilirubin level greater than 30% of the total bilirubin level. [18], [26] Broadly, causes may be congenital, familial, or acquired (Table 44-1).

TABLE 44-1 Causes of Conjugated Hyperbilirubinemia

Congenital Hyperbilirubinemias

Rotor syndrome. First described in 1948, this is a rare, asymptomatic, benign, congenital disorder that is manifested as conjugated or mixed hyperbilirubinemia. [34] It is an autosomal recessive disorder that becomes apparent in childhood. Total bilirubin levels are usually 2 to 5 mg/dL, which are greater than 50% conjugated bilirubin, with bilirubinuria and exacerbation of hyperbilirubinemia during illness. Other liver tests, including bile acids, are normal, as is liver histology. Although the result of an oral cholecystogram is normal, radionuclide scans show absent or markedly delayed excretion. It is unclear whether the primary defect is impaired secretion or impaired storage of bilirubin. Urinary coproporphyrins are markedly increased, especially type I.

Dubin-Johnson syndrome. This is an autosomal recessive syndrome resulting in impaired ATP-mediated transport of bilirubin diglucuronide as well as other organic ions. [35] Bilirubin refluxes back into plasma, and hyperbilirubinemia results. Total bilirubin levels are usually between 2 and 5 mg/dL; other liver tests are normal. Patients usually present after puberty, and exacerbations occur during intercurrent illness and with some drugs. The secretory defect in patients with Dubin-Johnson syndrome results in abnormal radionuclide scans and oral cholecystographic studies. Bile salt secretion is normal. Although total urinary coproporphyrin levels are normal, there is an increase in type I levels in the urine and a decrease in type III levels. Liver histology reveals a darkly pigmented liver that is black on gross inspection. The importance of diagnosing Rotor or Dubin-Johnson syndrome is to establish a diagnosis, to assure the patient that there is not a more serious problem, and to avoid repetitive testing for more serious disorders, including porphyrias.

Familial Cholestasis The disorders termed *progressive familial intrahepatic cholestasis* (PFIC) have disparate pathogeneses but share the following common features: chronic persistent hepatocellular cholestasis usually leading to cirrhosis; exclusion of other metabolic and anatomic defects; autosomal recessive inheritance patterns; and characteristic combined clinical, biochemical, and histological features. All these disorders share a defect in the generation of bile flow. The identification, cloning, and characterization of bile transport proteins have allowed a more precise definition of the diverse PFIC syndromes. [36], [37] PFIC-1 describes syndromic forms of defects in the *FIC1* gene. Byler disease is the best known and is named after Jacob Byler, of Amish ancestry. Patients typically have recurrent and later persistent cholestasis, evolving to cirrhosis. Clinically, the disease is characterized by jaundice, steatorrhea, growth retardation, hepatosplenomegaly, and watery diarrhea. Biochemically, although bilirubin, alkaline phosphatase, and serum bile acids are elevated, ?-glutamyltransferase and biliary bile acid concentrations are low. Mutations in the *FIC1* gene in PFIC-1 relate to a member of the subfamily of P-type ATPases, involved in aminophospholipid transport. [38] Byler syndrome describes non-Amish kindreds with a disorder similar to Byler disease but with linkage to other genetic loci.

Benign recurrent intrahepatic cholestasis. Mutations in the *FIC1* gene have also been identified in some patients with benign recurrent intrahepatic cholestasis (BRIC), which has different phenotypic manifestations. BRIC is a familial disorder characterized by recurrent episodes of painless intrahepatic cholestasis, beginning in early childhood or adulthood. [39] Episodes last weeks to months and may be associated with steatorrhea and weight loss, but they resolve spontaneously without permanent liver injury. Biochemical alterations are similar to those of Byler disease. Mutation analyses suggest that phenotypic differences in PFIC-1 and BRIC relate to quantitative differences in FIC1 protein function. [38] PFIC-2 designates a group of patients with phenotypic features similar but not identical to those with PFIC-1. In PFIC-2, the initial presentation and progression are more severe. Liver biopsy often shows evidence of giant cell hepatitis. The disorder was mapped to mutations in a gene encoding the canalicular bile salt export protein (BSEP), a gene related to the MDR family of the ATP-binding cassette transporter superfamily. [40] This results in defective canalicular excretion of bile acids with accumulation within hepatocytes and ongoing injury that is not responsive to treatment with ursodeoxycholic acid. PFIC-3 shares similar features with PFIC-1 and PFIC-2, except for elevated serum levels of ?-glutamyltransferase and extensive bile ductular proliferation. Patients have less jaundice, often present later in life, and may respond to ursodeoxycholic acid. Mutations have been described in the MDR3, a canalicular phospholipid transporter, [41] the absence of which permits toxic bile acid accumulation and exposure to hepatocytes and cholangiocytes. Other rare syndromes that may mimic PFIC are 3-ß-OH steroid dehydrogenase deficiency and cholestasis with lymphedema or hypertrichosis. [36]

Choledochal cystic disorders. These disorders, including Caroli disease, are inherited anomalies of the biliary ducts. Caroli disease involves the congenital dilation of the segmental intrahepatic biliary tree, whereas choledochal cysts involve the cystic dilation of the common bile duct. Patients can present at any age with jaundice or with symptoms of cholangitis, such as fever and right upper quadrant pain. Choledochal cysts are three times more frequent in female patients. Jaundice in a patient with Caroli disease or a choledochocele should raise suspicion of cholangiocarcinoma because these patients are at increased risk of developing such tumors.

Hepatocellular Diseases Hyperbilirubinemia may accompany acute or chronic hepatocellular disease and must be considered in the differential diagnosis and distinguished from diseases whose manifestations are predominantly cholestatic. All hepatocellular diseases may present as cholestasis, especially when severe, and they constitute the most common causes of intrahepatic cholestasis. Therefore, cholestatic forms of viral hepatitis and alcoholic liver disease are more common than primary sclerosing cholangitis or primary biliary cirrhosis. These hepatocellular diseases are described individually elsewhere in this book. Viral hepatitis may have a prolonged cholestatic phase after acute infection. Patients may come to medical attention only during this phase of lowered serum aminotransferases and high bilirubin, with or without prolonged prothrombin time. Patients with elevation in prothrombin time resulting from poor absorption will respond to parenteral vitamin K, whereas those with submassive necrosis or fulminant hepatic failure will not. Even patients with hepatitis A, which does not ever become chronic, may rarely have a prolonged cholestatic phase up to 18 months after acute infection. [42] Bilirubin may be elevated in 10% to 20% of patients with alcoholic hepatitis, and usually right upper quadrant pain, fever, and leucocytosis accompany this condition. [43] Finally, with end-stage liver disease of any cause, bilirubin may become elevated. However, physical signs of cirrhosis should be readily apparent. Patients with cholestatic immune-mediated liver diseases such as primary biliary cirrhosis, primary sclerosing cholangitis, and autoimmune cholangiopathy are cholestatic early (elevated serum alkaline phosphatase) but develop jaundice only later in the course of disease. [44], [45] Drug hepatotoxicity may produce a broad spectrum of liver injury that results in jaundice, cholestasis, or bile duct damage. [46]

Infiltrative Disorders These disorders may also result in cholestasis. When jaundice occurs in patients with primary or metastatic tumors, much of the liver is usually replaced, and the prognosis is poor. [47] However if a tumor obstructs a major bile duct, such as in cholangiocarcinoma, jaundice may occur earlier. Diffuse lymphomatous infiltration occurs with Hodgkin disease and periportal lymphadenopathy with non-Hodgkin lymphoma, and both may cause jaundice. Granulomatous hepatitis has a diverse etiology, and patients may occasionally present with jaundice. [48], [49] In systemic amyloidosis, either primary or secondary, histological evidence of hepatic involvement is common but usually clinically quiescent. However, a subgroup may develop marked cholestasis, which often portends a poor prognosis. [50]

Renal Disease The association between renal dysfunction and cholestatic jaundice is long established. A postoperative fall in glomerular filtration rate is observed in 60% to 75% of patients undergoing surgery for obstructive jaundice. [51] Acute renal failure may occur in 8% to 10% of those patients and is a contributing cause of

death in 70% to 80% of those who develop it. [52] The pathophysiology is complex and multifactorial, and not surprisingly, no clearly effective therapeutic strategy has emerged. Intrahepatic cholestasis, with or without jaundice, can occur as a paraneoplastic manifestation of renal cell carcinoma (Stauffer syndrome) in the absence of liver metastases. [53]

Infection Infections may cause jaundice through extrahepatic obstruction (e.g., ascariasis), granulomatous inflammation (e.g., tuberculosis), or, more commonly, through intrahepatic cholestasis. [54] The predominant mechanism of sepsis-associated cholestasis (especially gram-negative sepsis) is related to endotoxins derived from the bacterial cell wall, which are potent inducers of proinflammatory cytokines tumor necrosis factor-a and interleukin-1 (IL-1) and IL-6. [55] A disproportionate elevation of bilirubin to alkaline phosphatase and aminotransferases is suggestive of sepsis-related cholestasis. [56] Persistent or increasing hyperbilirubinemia with ongoing infection correlates with the worst outcome. Patients infected with human immunodeficiency virus (HIV) infection are susceptible to various hepatic processes that are related to immunosuppression or are associated with the risk factors of homosexuality and parenteral drug abuse. These disorders include hepatic granulomas, cytomegalovirus hepatitis, multimicrobial HIV cholangiopathy, Kaposi sarcoma, and lymphoma. The differential diagnosis also includes other opportunistic infections and neoplasms, as well as concomitant chronic viral hepatitis B, C, and D and drug-related (including antiretroviral) hepatotoxicity. [57] Systematic evaluation and treatment are necessary to ensure that morbidity and mortality are minimized and quality of life and medical care costs are optimized. [58]

Bone Marrow Transplantation Bone marrow transplant recipients may develop jaundice for many reasons. Venoocclusive disease is a common cause of hyperbilirubinemia, especially after autologous bone marrow transplantation, [59] and it is associated with increased morbidity and mortality. Other complications include drug- and chemotherapy-induced hepatitis, sepsis-related liver injury, and recurrent disease. Acute or chronic graft versus host disease affects as many as half of long-term transplant survivors and is associated with elevated alkaline phosphatase levels and a poor prognosis.

Total Parenteral Nutrition Total parenteral nutrition (TPN) is associated with a spectrum of hepatobiliary complications that are a major cause of morbidity and mortality and increase with the duration of therapy. Cholestasis is the most common hepatic complication in infants receiving TPN. Although cholestasis is less common in adults, steatosis and steatohepatitis in addition to biliary sludge and gallstone formation predominate. [60] The cause of TPN-induced cholestasis is uncertain but is likely multifactorial. [61], [62] Direct hepatotoxicity may result from individual TPN components including excess lipids and amino acids such as tryptophan and peroxides. Cholestasis may be related to specific nutritional deficiencies in TPN of substances such as taurine and methyl-donor molecules. Defective or altered bile acid secretion may be relevant and may have therapeutic implications. The lack of enteral intake and inadequate stimulation of the enterohepatic circulation and gut function lead to diminished cholecystokinin release and stimulation of the gallbladder. This has numerous potential pathological consequences including gallbladder and bile stasis. Gut hypomotility and small bowel bacterial overgrowth results with production and absorption of lithocholic acid, a cholestatic bile salt, and enhanced endotoxin absorption, often exacerbated by concomitant sepsis. Liver biochemical tests are of limited use as sensitive or specific indicators of TPN-related hepatobiliary disease. Liver biopsy is rarely necessary. Jaundice may reflect the development of biliary sludge (50% incidence at 4 to 6 weeks of TPN and present in 100% of patients after more than 6 weeks) and calculous or acalculous biliary tract disease (the former occurring particularly with preexisting ileal disease). With long-term TPN, a rising alkaline phosphatase level may reflect the development of intrahepatic cholestasis or, alternatively, may reflect progression of TPN-associated steatohepatitis to decompensated cirrhosis. [63] Approaches that may ameliorate the hepatobiliary impact include the minimization of duration of TPN, cycling of TPN, optimization of caloric components, and reduction of excess lipids and amino acids. Attempted limited enteral intake, selective gut decontamination, and the use of ursodeoxycholic acid and cholecystokinin may further limit the TPN injury.

APPROACH TO THE PATIENT WITH JAUNDICE

The investigation of a patient with jaundice begins with a thorough review of the history of presentation, medication use, past medical history, examination, and evaluation of liver-related laboratory tests. With an understanding of the pathophysiology of cholestasis, a systematic approach to the jaundiced patient can be applied. Identification of the correct diagnosis can lead to an appropriate therapeutic intervention. The implications of jaundice in certain conditions can be life-threatening, and thus a timely diagnosis is important. Several questions must be answered initially:

1. Is the elevated bilirubin conjugated or unconjugated? In general, most jaundiced patients will not have isolated unconjugated hyperbilirubinemia.
2. If the hyperbilirubinemia is unconjugated, is it caused by increased production, decreased uptake, or impaired conjugation?
3. If the hyperbilirubinemia is conjugated, is the problem intrahepatic or extrahepatic?
4. Is the process acute or chronic?

Patients with conjugated hyperbilirubinemia usually have acquired disease, and the physician must identify an intrahepatic or obstructive cause. Acute disease can usually be differentiated from chronic disease by the patient's history, physical examination, and laboratory tests. For example, clinical evaluation may demonstrate xanthelasmas, spider angiomata, ascites, or hepatosplenomegaly. Laboratory evidence of chronic disease may consist of a hypoalbuminemia, thrombocytopenia, and a prolonged prothrombin time that is not corrected with vitamin K administration.

Chronic cholestasis may arise from such diseases as cirrhosis, primary sclerosing cholangitis, primary biliary cirrhosis, or carcinoma, or it may result from drugs. Patients with chronic cholestasis do not usually have hepatitis or gallstones. [64] The presence of fever, right upper quadrant pain, tenderness, hepatomegaly, and new-onset bilirubinuria usually indicates acute disease. An adult with asymptomatic, isolated unconjugated hyperbilirubinemia who is not taking any drugs and has no evidence of hemolysis probably has Gilbert syndrome and can be monitored with bilirubin determinations for 12 months. If no abnormality develops, no further evaluation is needed.

The patient's history, physical examination, and laboratory tests are crucial to the diagnosis of hyperbilirubinemia. [65] Physicians are 80% to 90% accurate in diagnosing extrahepatic disease by these means, but obstruction is often overdiagnosed. [66] When first evaluating a patient with hyperbilirubinemia, the physician must make a quick assessment of the emergency of the situation. Fever, leukocytosis, and hypotension point to ascending cholangitis, which requires immediate therapy. Asterixis, confusion, or stupor may indicate severe hepatocellular dysfunction or fulminant hepatic failure and mandates immediate therapy. After immediate life-threatening causes of hyperbilirubinemia have been excluded, a systematic approach to the patient helps to make the diagnosis.

Diagnosis by Age of the Patient

The most common causes differ according to the age of the patient. In neonates, physiological jaundice is overwhelmingly the most common cause of jaundice, with biliary atresia, infections (TORCHES), and metabolic diseases together accounting for less than 25% of cases. In adolescents, Gilbert syndrome and viral hepatitis account for 80% of cases, with the remainder including toxins, drugs, and autoimmune and biliary tract diseases. A careful history will elicit drug use or abuse, risk factors for viral hepatitis, amenorrhea associated with autoimmune hepatitis, or symptoms associated with biliary tract disease.

In young adults, viral hepatitis is the most common cause of jaundice, followed by biliary tract disease, alcoholic liver disease, and autoimmune diseases. Genetic diseases (hemochromatosis and Wilson disease) may present solely as elevation in aminotransferases but rarely as cholestasis at this age, and evaluation should include ceruloplasmin, urine copper, ferritin, and transferrin saturation in these patients.

In contrast, malignant disease accounts for half of the cases of jaundice seen in elderly patients. Drug toxicity is common with increasing polypharmacy in the elderly and should always be included in the history of these patients with jaundice. Multiple drugs have been associated with hyperbilirubinemia, whether hepatocellular (acetaminophen), cholestatic (anabolic steroids), or mixed (sulfonamides). Ten percent of abnormal liver tests in hospitalized patients are associated with drug toxicity. Autoimmune disease has a second peak in the elderly and should be sought. Antinuclear antibodies may be weakly elevated in healthy elderly patients, and other laboratory evidence of autoimmune disease should be sought (elevated IgG in autoimmune hepatitis, IgM in primary biliary cirrhosis). Biliary tract disease, viral hepatitis, and alcoholic liver disease also occur in elderly patients

Neonatal and Childhood Jaundice

The neonate is predisposed to the development of jaundice because of an increased production of bilirubin, reduced bilirubin UGT levels, and impaired excretory ability. Physiological jaundice (with a serum bilirubin of 5 to 6 mg/dL) occurs in full-term infants in the first few days of life and is more common in premature infants, usually resolving over several weeks. Exaggerated physiological jaundice is characterized by hyperbilirubinemia up to 17 mg/dL, but higher levels in a term infant should prompt a search for a pathological cause. [11] The consequences and optimal treatment of neonatal hyperbilirubinemia are controversial. [67] Neonates should have follow-up examinations 2 to 5 days after delivery, and jaundiced infants require early measurement of total bilirubin. Phototherapy is the mainstay of therapy. Exchange transfusions are rarely necessary.

Neonatal infection, birth trauma, and hypothyroidism may compound hyperbilirubinemia. Increased bilirubin production occurs in certain racial groups such as Native

Americans. Inherited defects in red cell enzymes such as glucose-6-phosphate dehydrogenase deficiency, red cell structural defects such as elliptocytosis, or blood group incompatibility may result in pathological jaundice. Icterus is neonates may be related to Gilbert syndrome. In Crigler-Najjar syndrome type I, bilirubin encephalopathy may occur in the first few days or months of life, whereas in Crigler-Najjar syndrome type II, bilirubin levels are rarely greater than 20 mg/dL. Neonates, particularly if they are not feeding well or are exclusively breast-fed, have lower levels of intestinal bacteria. As a result, bilirubin deconjugated in the gut is not reduced to urobilinogen and excreted but undergoes enhanced enterohepatic circulation. Later-onset breast milk jaundice is seen in 0.5% to 1% of newborns during days 4 to 10 after birth. Unconjugated bilirubin levels may rise to 10 to 20 mg/dL. The infant remains well, interruption of breast-feeding will result in a rapid decline in bilirubin levels, and resolution occurs by 4 to 6 weeks. [68] Transient familial neonatal hyperbilirubinemia (Lucy-Driscoll syndrome) results from a serum inhibitor of bilirubin UGT that disappears by 2 weeks of life.

Severe neonatal hyperbilirubinemia has neurotoxic sequelae in the form of bilirubin encephalopathy. *Kernicterus* describes the pathological findings of yellow staining and necrosis of neuronal cells of the basal ganglia, hippocampal cortex, and subthalamic area, and clinically, it may follow an acute or chronic course with potentially debilitating outcomes. Early hospital discharge and a less aggressive approach to treatment may have contributed to a possible recent re-emergence of this disorder. [69], [70] The free fraction of unconjugated bilirubin crosses the blood-brain barrier, especially if it is impaired by infection, prematurity acidosis, or hyperoxia. The neurotoxic effects are related to the concentration of bilirubin in the brain and the duration of exposure, although serum bilirubin levels correlate poorly with encephalopathy in nonhemolytic hyperbilirubinemia. Hypoalbuminemia and drugs that displace bilirubin from albumin further exacerbate the toxicity.

In the older neonate and infant, the diagnostic algorithm must reflect knowledge of the age-specific onset of disorders and their predominant presenting features, combined with the use of selected diagnostic tests. [71] Whereas jaundice may be obvious, more subtle manifestations of chronic cholestasis may exist such as the consequences of fat-soluble vitamin deficiency (neurological and hematologic), xanthomata, and growth failure. In the initial evaluation of an infant with cholestasis, if hepatic synthetic function is impaired, causes of liver failure must be considered such as neonatal iron storage disease, tyrosinemia, galactosemia, fructosemia, and mitochondrial cytopathies. In the absence of liver failure, other metabolic causes such as a $_1$-antitrypsin deficiency and cystic fibrosis and viral causes such as herpes must be evoked.

Anatomic causes of cholestasis that may be corrected surgically must be distinguished. Biliary atresia is the single most common cause of neonatal cholestasis and accounts for 50% to 60% of pediatric liver transplantations. Two phenotypes are apparent: the less common (congenital) form presents in the first few weeks of life, and the more common phenotype (acquired form) presents in the first to second month of life. Numerous theories attempt to provide rational etiologic explanations. [72] Jaundice, pale stools, and hepatomegaly are the dominant clinical features. Rapid diagnosis is essential for definitive surgical intervention. Portoenterostomy results in successful biliary drainage in 80% of patients, but it must be performed as early as possible. In infants, idiopathic neonatal hepatitis is the second most common cause of cholestasis and refers to a heterogeneous group of disorders that includes infectious, toxic, metabolic, and genetic defects in bile acid synthesis. [36]

Alagille syndrome is a cause of cholestasis before the age of 6 months. The five main features are hepatic, cardiovascular, vertebral, and ocular and a peculiar facies. [73] Expression is variable, and the first two manifestations are most common. Jaundice may be intermittent, but cholestasis is persistent, with its attendant complications. Histologically, paucity of interlobular bile ducts is typical. The mode of transmission is autosomal dominant with reduced penetrance. Mutations in the coding sequence of the *JAGGED1* gene have been identified in a large proportion of patients with the Alagille syndrome. *JAGGED1* codes a ligand for the NOTCH1 receptor, implicated in a fundamental mechanism controlling cell fate during embryogenesis.

Liver Disease in Pregnancy

Liver disease in pregnancy includes disorders unique to pregnancy and those coincident with or exacerbated by pregnancy. [74], [75] and [76] Normal changes during pregnancy include a lower mean serum albumin (3.1 g/dL), resulting from hemodilution, and a higher alkaline phosphatase level (two to four times normal), mainly of placental origin. Fibrinogen, transferrin, and cholesterol levels are also increased. Serum aminotransferases, bilirubin, serum bile acids, and ?-glutamyltransferase levels are unchanged. Spider angiomata and palmar erythema may be seen in more than 60% of pregnant women. [77] The differential diagnosis of hepatic disorders of pregnancy is dependent on the time of onset in relation to the stage of gestation and the type of symptoms and signs. Careful history of present and past pregnancies, as well as parity, including a family history of complicated pregnancies, may provide important clues to diagnosis.

Liver Disease Unique to Pregnancy

Hyperemesis gravidarum. Intractable nausea and vomiting in the first trimester are key features of hyperemesis gravidarum, which may result in dehydration, electrolyte disturbances, and malnutrition. More common in young women with a multiple gestation, it may be associated with reduced fetal wastage. Mild elevations of alkaline phosphatase and hyperbilirubinemia (<4 mg/dL) occur, although aminotransferases may rarely increase markedly. Treatment includes hydration and careful management of electrolytes and nutrition.

Intrahepatic cholestasis of pregnancy. Intense, often debilitating pruritus is the hallmark of intrahepatic cholestasis of pregnancy (ICP), which typically begins in the third trimester. [78] Although rare in most countries, the endemic occurrence in Chilean Indians and Scandinavians suggests a strong genetic predisposition in some persons. ICP recurs in 45% to 70% of subsequent pregnancies and with the use of birth control pills. It is more common in twin pregnancies. In some women, defects in the MDR3 transporter involved in bile salt secretion may contribute to a subset of women with ICP who also have an increased ?-glutamyltransferase levels. [79] Hyperbilirubinemia, up to 5 mg/dL, is detected in 20% of cases, with minimal elevations in hepatic alkaline phosphatase. An increase in total serum bile acids (often up to 100-fold) with pruritus is highly suggestive of ICP. [80] Fetal morbidity is increased because of premature births (19%–60%) and intrapartum fetal distress (22%–33%). The perinatal mortality of 10% to 11% is poorly predicted by maternal disease severity or conventional antepartum fetal testing. [78] Limited controlled data suggest that early treatment with ursodeoxycholic acid is safe, improves pruritus, lowers bilirubin and aminotransferases, normalizes some bile acid ratios, and may contribute to improved fetal outcome. [81], [82] and [83] Early delivery for fetal distress, preferably after establishment of fetal lung maturity, is usually followed by complete resolution.

Other disorders. In the third trimester (and rarely in the postpartum period), three other disorders unique to pregnancy have predominantly hepatocellular patterns of injury with elevated aminotransferases. If severe, they may be associated with cholestasis and significant maternal and perinatal morbidity and mortality. [74], [75], [84] *Acute fatty liver of pregnancy* is characterized by a spectrum of disease that may extend from mild disease to fulminant hepatic failure. Histology reveals a microvesicular steatosis. In some cases, inherited disorders of mitochondrial ß-oxidation of long-chain fatty acids are evident in the fetus and mother. [85] *Preeclampsia* is a multisystem disorder associated with proteinuria, hypertension, peripheral edema, and hyperreflexia. It is common (5%–7%) among primigravidas, especially those with a multiple gestation. Hepatic involvement indicates severe disease and increased risk of eclampsia. Serum aminotransferases may be massively increased. Acute, severe, right upper quadrant pain may raise the specter of complicating hepatic infarction or rupture. *HELLP syndrome,* characterized by hemolysis, elevated liver enzymes, and low platelet counts, may occur in a subset of patients with preeclampsia/eclampsia or acute fatty liver of pregnancy. Indeed, it may be difficult to distinguish the latter three disorders because they share many overlapping features. [75] However, prompt recognition and expedited delivery remain the cornerstone of management of the more severe cases. Liver biopsy is reserved for patients in whom the result will affect immediate obstetrical management. Oil red O staining of fresh tissue is required for confirmatory diagnosis of microvesicular steatosis. Complete recovery usually follows delivery.

Diseases Coincident with Pregnancy Disorders that are exacerbated by pregnancy include acute hepatitis E virus in travelers from endemic areas, where maternal mortality may approach 29% in the third trimester. Herpes simplex hepatitis may be more common in pregnancy; it has a high mortality rate, and prompt antiviral therapy improves survival. Some patients with Dubin-Johnson syndrome may experience asymptomatic, isolated, conjugated hyperbilirubinemia during the third trimester. A hypercoagulable state exists in pregnancy and can be associated with the development of Budd-Chiari syndrome. Pregnancy is associated increased gallstone formation, and extrahepatic cholestasis from choledocholithiasis or biliary sludge may be encountered, extending into the postpartum period. Surgical management may be appropriate only in the second trimester. For women with known severe chronic liver disease, amenorrhea may be present. However, autoimmune hepatitis and primary biliary cirrhosis are usually better controlled in pregnancy because of increased endogenous glucocorticoid production. Patients with portal hypertension may develop variceal bleeding, and there is a higher degree of fetal wastage. [86]

Postoperative Jaundice

Jaundice is uncommon after elective abdominal surgery (<1%), but it may occur in up to 17% of patients who are in the intensive care unit or after major surgery. The cause is multifactorial, and liver failure is rarely associated with the jaundice. Specific treatment is not usually required. Postoperative jaundice may be classified into three major categories. [87]

1. Bilirubin overproduction may result from hemolysis of transfused blood or absorption of hematomata; hemolysis may be related to administered drugs, prosthetic valves, or underlying hemolytic anemias; or Gilbert syndrome may occur. One liter of transfused blood will generate 5 g of bilirubin. Ten percent of red blood cells within a transfused unit are hemolyzed within 24 hours of storage.

2. Hepatocellular dysfunction may result from a diverse group of insults [88]: anesthesia, ischemia, drugs, and TPN. Ischemia, related to cardiogenic or noncardiogenic shock, may be apparent only after careful examination of the anesthetic records. Most forms of anesthesia reduce hepatic blood flow. Anesthetic drugs, especially the halogenated hydrocarbon anesthetics such as halothane, typically cause acute hepatitis within 21 days of initial exposure. Seventy-five percent of patients have an accompanying fever, and 20% to 60% of patients have peripheral eosinophilia. [89] Recurrent exposure may result in icterus within 7 days. Features predictive of an adverse outcome include age greater than 60 years, obesity, multiple exposures, short latent period to the development of jaundice, serum bilirubin more than10 mg/dL, and prothrombin time longer than 20 seconds. A careful review of drugs (including TPN) employed in the perioperative period is essential. Posttransfusion hepatitis is now rarely encountered because of screening of the donor population. Preexisting liver disease is an important risk factor and may be exacerbated by an operative procedure, potentially leading to decompensation if the patient has cirrhosis. Ascites is frequent in cirrhotic patients who decompensate after surgery as a result of poor volume handling. Jaundice with abnormal biochemical test results is commonly seen in patients who have bacteremia, especially with intra-abdominal sources of infection. The mechanisms are uncertain and may relate to direct infection of the liver, endotoxin production, or hemolysis. [90]
3. Acalculous cholecystitis is a cause of jaundice in hospitalized patients, especially after vascular surgery, trauma, or burns. The mortality is high, usually related to comorbid conditions. Proposed etiologic mechanisms include bile stasis, infection, and gallbladder ischemia. Patients affected are often volume depleted, receiving opiates and hyperalimentation. A high index of suspicion is necessary in a hospitalized patient with right upper quadrant pain, fever, leukocytosis, and cholestatic hepatic biochemical abnormalities. Ultrasound is usually the favored and feasible imaging study, and typical sonographic features include a thickened wall, pericholecystic fluid, intramural gas, and sloughed mucosal membrane. [91] Treatment should consist of antibiotics and possible cholecystostomy.

Any evaluation of postoperative jaundice should include a careful review of the operative notes, blood products requirements, microbiology culture results, and medications. In patients with cholestasis, an ultrasound interrogation of the biliary tree should be performed.

Cholestasis after Liver Transplantation

Cholestasis occurring after liver transplantation may be categorized as related to conditions occurring early, within 6 months of transplantation or late, after 6 months from transplantation, though overlap may occur. [92] Early cholestasis may be related to the condition of the donor liver, vascular and biliary problems, infections, and acute rejection. Functional cholestasis related to donor liver preservation injury (cold or rewarming ischemia or reperfusion injury) peaks between 10 and 16 days postoperatively and usually resolves with supportive management. Prolonged ischemia, especially cold type, is associated with an increased risk of later biliary strictures. Bacterial infection may result in cholestasis and mandates a systematic evaluation of potential sources, particularly the biliary tree. Cholestasis may result from viral infection, especially by cytomegalovirus, chiefly within 3 months of transplantation. Direct viral culture, the faster shell vial culture technique, or histological documentation of the cytopathic effects and microabscesses may confirm the diagnosis. Most transplant recipients experience at least one episode of acute cellular rejection, although a predominant cholestatic pattern of liver enzyme abnormalities is uncommon. Complex drug regimens used after transplantation predispose to drug-related cholestasis. Cyclosporine, tacrolimus, and azathioprine (but not mycophenolate mofetil) have been implicated, in addition to sulfonamides. The integrity of the bile ducts is dependent on the hepatic arterial supply. Subacute or incomplete hepatic arterial thrombosis can result in chronic ductal ischemia, bacteremia from cholangitis, and ultimately nonanastomotic biliary strictures.

Cholestasis after 6 months may result from chronic rejection, recurrent disease, and bile duct damage. Chronic rejection is characterized histologically by bile duct atrophy and loss leading to progressive cholestasis and graft failure that is unresponsive to antirejection therapy. Primary biliary cirrhosis may recur in 17% of patients. [93] Fibrosing cholestatic hepatitis is an early aggressive posttransplantation form of recurrent viral hepatitis, especially hepatitis C. It rapidly leads to graft failure and is characterized histologically by intrahepatic cholestasis with perisinusoidal fibrosis and minimal inflammation. [94] Retransplantation is often necessary.

DIAGNOSTIC APPROACH

Figure 44-2 and Figure 44-3 depict algorithms useful in the differential diagnosis and evaluation of a patient with jaundice, respectively. The initial step is to determine whether the jaundice is conjugated or unconjugated. The causes of conjugated hyperbilirubinemia range anatomically from the ampulla of Vater to the hepatocyte. If the history and physical examination do not provide a clue to the cause, the initial approach should be a right upper quadrant ultrasound scan to evaluate the liver, the biliary system, and the porta hepatitis.

FIGURE 44-2. Differential diagnosis of the jaundiced patient.

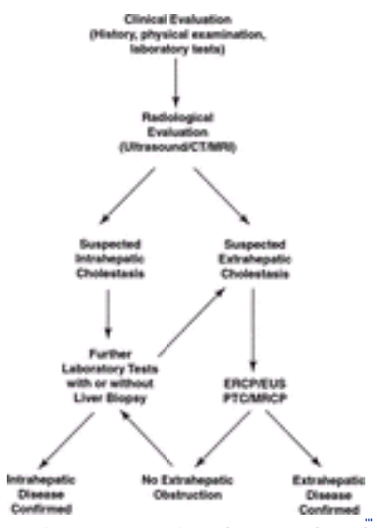

FIGURE 44-3. The evaluation of the jaundiced patient.

History

A family history of liver disease, alcohol and drug history, sexual history, transfusion history, and nutrition history are important clues to the possible cause of hyperbilirubinemia. Exposure to environmental toxins, persons with jaundice, drugs (e.g., prescription, nonprescription, and intravenous drugs and nutritional herbal supplements), and outbreaks or epidemics in the community should be sought. Previous liver biochemical tests are valuable, as is a history of biliary or pancreatic disease.

Constitutional symptoms, such as fever, chills, weight loss, and flulike symptoms, are also important clues. For instance, shaking chills or fevers point toward

cholangitis or bacterial infection and should steer the clinician away from viral hepatitis. Abdominal pain may indicate pancreatic disease, especially if it radiates to the back, but it is otherwise not helpful. [95] Viral hepatitis can cause a right upper quadrant ache, but it is not usually described as a pain. Weight loss, anorexia, nausea, and vomiting are not helpful signs because most patients with hepatobiliary disease or obstruction have anorexia and some weight loss. [26] The absence of weight loss does not rule out a diagnosis of malignant disease. Pruritus can be associated with both intrahepatic cholestasis as well as biliary obstruction, especially if the latter lasts longer than 3 to 4 weeks.

The patient's age may be helpful in constructing a differential diagnosis, as discussed earlier. Patients younger than 30 years old are more likely to have acute parenchymal disease. Patients older than 65 years are more likely to have stones or malignant diseases. Patients between 30 and 50 years of age are more likely to have chronic liver disease. In general, children and young adults are more likely to have viral hepatitis. Women older than 30 years and men older than 50 years are likely to have biliary tract disease in the form of stones or carcinoma, respectively. In middle adulthood, cirrhosis and drug effects are common causes. Men are more likely to develop cirrhosis secondary to alcohol use, pancreatic cancer, hepatocellular carcinoma, or hemochromatosis. Women are more likely to have primary biliary cirrhosis, gallstones, and chronic active hepatitis.

Physical Examination

Physical examination should include examination of the liver and examination for evidence of chronic liver disease and extrahepatic disease. Jaundice is differentiated from other abnormalities in skin color, such as hypercarotenemia, uremic pigmentation, picric acid ingestion, or quinacrine therapy because only bilirubin stains the sclera, because of its high affinity for elastin. [26] Later, and especially if the jaundice is severe, the skin color be greenish. The general appearance of the patient may suggest chronic liver disease with cachexia, muscle wasting, palmar erythema, Dupuytren contracture, leukonychia, parotid enlargement, skin pigmentation (from melanin), or xanthelasmas. Chest evaluation may reveal gynecomastia, spider nevi, or dilated veins.

The size and consistency of the liver may be helpful. A shrunken, nodular liver may allow one to identify cirrhosis, and a palpable mass may indicate an abscess or malignant tumor. If the liver span is greater than 15 cm, the physician should consider fatty infiltration, congestion, other infiltrative diseases, or malignant disease. Liver tenderness may denote acute disease but is generally not helpful. The presence of a friction rub or bruit suggests malignant disease. Spider angiomata, palmar erythema, and distended abdominal veins in a patient with jaundice indicate cirrhosis. [95] Ascites in the presence of jaundice usually indicates cirrhosis, although it can be seen with malignant disease and severe acute disease, such as viral or alcoholic hepatitis. Splenomegaly may be seen in patients with infections, infiltrative diseases, viral hepatitis, or cirrhosis. A palpable, distended gallbladder suggests biliary obstruction, often malignant in origin. Asterixis is an unusual finding, except in fulminant hepatic failure and end-stage liver disease.

Laboratory Tests

Laboratory tests can confirm suspicions formed during the history and physical examination. The clinician must differentiate conjugated from unconjugated hyperbilirubinemia by using total and direct bilirubin assays. Patients with unconjugated hyperbilirubinemia should be evaluated for evidence of hemolysis, which includes a reticulocyte count, examination of the peripheral smear, serum level of lactic dehydrogenase, and haptoglobin levels. An abnormality in any of these values may lead the clinician to look for evidence of ineffective erythropoiesis, such as vitamin B_{12} deficiency, lead toxicity, thalassemia, or sideroblastic anemia. When hemolysis is excluded in patients with unconjugated hyperbilirubinemia, most asymptomatic healthy patients have Gilbert syndrome.

For conjugated hyperbilirubinemia, initial serum tests should include assays of serum aminotransferases and alkaline phosphatase. These test results can differentiate hepatocellular from cholestatic disease. Bilirubin levels consistently less than 5 mg/dL are not seen in obstruction, unless early, but they are common in patients with cirrhosis. [95] Bilirubin levels greater than 20 mg/dL in the presence of normal renal function, especially in elderly patients, should make the clinician suspect malignant biliary obstruction.

Although neither aspartate aminotransferase nor alanine aminotransferase levels are specific for liver disease, they rarely rise to more than 300 IU/mL in any other diseases. [26] An acute myocardial infarction can elevate the serum aspartate aminotransferase level because of necrotic cardiac muscle. In a jaundiced patient, serum aminotransferase levels less than 300 IU/mL are seen in alcoholic hepatitis and drug-induced injury, although these levels also are seen in chronic liver disease and obstruction. Levels greater than 400 IU/mL indicate hepatocellular injury, and levels greater than 1000 IU/mL usually indicate acute hepatitis, drug hepatotoxicity, or prolonged hypotension. A level of serum alanine aminotransferase greater than the aspartate aminotransferase level suggests viral hepatitis or nonalcoholic steatohepatitis.

Second-line tests for jaundice help to evaluate the patient for evidence of obstruction and exclusion of bone disease (e.g., ?-glutamyltransferase, 5'nucleotidase, leucine aminopeptidase), specific liver diseases (e.g., antimitochondrial antibody, hepatitis serologies, a_1-antitrypsin, iron levels, ceruloplasmin), malignant disease (e.g., a-fetoprotein), and autoimmune phenomena (e.g., immunoglobulins, sedimentation rate, antinuclear antibody). Bile acids are increased in virtually all forms of hepatobiliary disease and are not usually useful in determining the cause of jaundice (except in inherited disorders of bilirubin metabolism, when they are normal).

The alkaline phosphatase level can signal the cause of hyperbilirubinemia. If the alkaline phosphatase level is normal, extrahepatic obstruction is unlikely, with the exception of early acute obstruction. [95] If the alkaline phosphatase level is more than three times the minimal abnormal level, cholestasis or extrahepatic obstruction probably exists. The level of alkaline phosphatase may be elevated disproportionately compared with bilirubin in partial biliary obstruction or early intrahepatic cholestasis (e.g., primary biliary cirrhosis or primary sclerosing cholangitis) because the large reserve of nonobstructed parenchyma remains intact to excrete bilirubin. The elevation in alkaline phosphatase and bile acids reflects enzyme shedding in the hepatocyte and altered permeability in the biliary tree. [2], [96] If the alkaline phosphatase and bilirubin levels are markedly elevated, a common bile duct stone should be excluded. Therefore, the alkaline phosphatase level is a more sensitive test for biliary obstruction than bilirubin.

Bilirubin remains normal until most of the bile ducts are obstructed. ?-Glutamyltransferase is found in the liver, pancreas, heart, and lungs and is elevated in a multitude of disorders. It is elevated in patients with hepatobiliary disease, alcohol intake, pancreatitis, chronic lung disease, renal failure, diabetes, and congestive heart failure and as a result of a variety of drugs.

Albumin levels and prothrombin time should be determined to assess liver function. Protein levels help to differentiate acute from chronic liver disease. Elevated globulin with hypoalbuminemia supports the diagnosis of cirrhosis, as does failure of the prothrombin time to correct after oral or parenteral administration of vitamin K. A trial of vitamin K should be given and administered parenterally to ensure adequate absorption. Hypercholesterolemia is often seen in patients with cholestasis.

Urine tests that signal cholestasis or elevated bilirubin include urinary urobilinogen and urinary conjugated bilirubin. In acute hyperbilirubinemia, jaundice can lag behind bilirubinuria. [26] The renal threshold for conjugated bilirubin is 1 mg/dL, which is less than that needed to produce clinical jaundice. False-positive urine test results occur with salicylates, and phenothiazines and false-negative results occur if the urine is not analyzed promptly. A positive urine test result is a sensitive indicator of conjugated hyperbilirubinemia and should prompt further investigation. [97]

Noninvasive Tests

After the history, physical examination, and laboratory tests are obtained, an informed choice must be made regarding further diagnostic tests. These tests incur substantial cost and therefore should not be ordered indiscriminately for any abnormal biochemistry value. These diagnostic tests can be invasive or noninvasive. Noninvasive tests include ultrasound, computed tomography (CT), magnetic resonance imaging (MRI), and technetium 99m (^{99m}Tc)–labeled iminodiacetic acid derivative scans.

Ultrasound Ultrasound is the first test used to detect biliary obstruction. The diagnostic accuracy ranges from 77% to 94%, and the result is most accurate when the bilirubin level exceeds 10 mg/dL. Ultrasound allowed a correct diagnosis in 86% of 35 icteric patients. [98] In cases of acute obstruction, it may take 4 hours to 4 days for ducts to dilate, and the ducts of some patients with partial or intermittent obstruction may not dilate. Not all patients with dilated ducts have obstruction, and conversely, between 24% and 40% of patients with common bile duct stones have normal-sized bile ducts. Ultrasound has a sensitivity of up to 77% and specificity of 83% to 95% for bile duct obstruction. [99] The variability in sensitivity reflects limitations from overlying bowel gas, site and size of the stones, and presence or absence of duct dilation. Ultrasound is inconsistent in determining the site of obstruction, partly because of its inability to see the distal duct well in 30% to 50% of patients. [100], [101] and [102] Despite these caveats and the fact that ultrasound is operator dependent, it remains the preferred initial screening test for evaluating biliary obstruction.

Computed Tomography CT imaging has a sensitivity of 60% to 90% and results rely less on the operator's proficiency. CT is not impeded by fat. However, CT scanning is more expensive than ultrasound and requires intravenous contrast medium in many instances. Unless the patient is obese, ultrasound should be the initial test of choice. [103]

Magnetic Resonance Imaging MRI relies on the physical properties of unpaired protons in tissues to generate images, without use of ionizing radiation. [104], [105] MRI is generally insufficiently sensitive or specific in the assessment of diffuse liver disease, except for the assessment of fat and iron. It appears that MRI is more sensitive and specific than CT with contrast for the detection and evaluation of focal and malignant lesions. Using the tissue contrast inherent in the technique, MRI avoids the potentially nephrotoxic contrast agents used with CT imaging. This is particularly relevant in patients with jaundice or cholestasis. The MRI characteristics of stationary and mobile liquids makes MR cholangiopancreatography (MRCP) and MR angiography powerful noninvasive diagnostic tools. Bile duct calculi are seen particularly well with MRCP. With MRI, imaging of the biliary tree is feasible both proximal and distal to the site of obstruction. This is particularly valuable after inadequate or unsuccessful endoscopic retrograde cholangiopancreatography (ERCP), [106] although its use is more difficult to justify if there is a high probability of the need for therapeutic intervention.

Radiolabeled Technetium-Sulfur Colloid This is rapidly taken up by the reticuloendothelial system. The image generated depends on blood flow and the functional state of the reticuloendothelial system. Radionuclide imaging is an excellent means of detecting cystic duct obstruction. It is the test of choice if acute cholecystitis with cystic duct obstruction and biliary leakage is suspected, but it has little value in differentiating intrahepatic from extrahepatic causes of cholestasis. The hepatic iminodiacetic acid scan, which makes use of ^{99}Tc-labeled iminodiacetic acid scan, has a 15% to 30% false-negative rate for detecting intrahepatic lesions. The scan is not a sensitive means of detecting ductal dilation because the image seen depends on the concentration of tracer. Di-isopropyl and *p*-isopropyl iminodiacetic acid are tracers that allow biliary tree visualization in the presence of jaundice. The test result is normal if the gallbladder, common bile duct, and small intestine are seen within 60 minutes. False-negative tests occur with prolonged fasting, TPN, and bilirubin levels greater than 5 mg/dL.

Invasive Tests

Invasive tests currently used include ERCP, percutaneous transhepatic cholangiography (PTC), endoscopic ultrasound (EUS), and liver biopsy. ERCP and PTC use cholecystographic dye and radiography to visualize the biliary tree. They are excellent tests to verify ductal dilatation and permit concomitant therapeutic intervention.

PTC visualizes the biliary tree in 90% to 100% of patients with dilated ducts and localizes the site of obstruction in 90% of cases. To perform the test safely, PTC usually requires a prothrombin time of less than 16 seconds, a platelet count greater than 50,000, and the absence of ascites. Minor complications occur in 30% of patients. Major complications, including sepsis, bleeding, biliary leak, pneumothorax, arteriovenous fistula, hematoma, abscess, and peritonitis, occur in 1% to 10% of patients who undergo PTC.

ERCP can localize the site of obstruction in more than 90% of patients. It is particularly helpful in diagnosing patients with common duct stones. Because it has therapeutic capabilities, it allows some patients to avoid surgery. ERCP is also helpful if a stricture resulting from chronic pancreatitis is suspected. The major reasons for nonvisualization of the biliary tree during ERCP are prior surgery (e.g., Roux-en-Y loop) and an inability to cannulate the sphincter of Oddi. The morbidity rate is 2% to 3%, somewhat less than with PTC. The most common complications are pancreatitis, bleeding, and cholangitis. The rate of sepsis is less than 1% if prophylactic antibiotics are given when an obstruction is suspected.

EUS combines endoscopy with real-time, high-resolution ultrasound and provides excellent sonographic visualization of the biliary tree without bowel gas interference. EUS is superior to ultrasound and CT for diagnosing bile duct stones. [107] EUS is comparably accurate but safer and less expensive than ERCP when evaluating patients with suspected choledocholithiasis. If available, EUS should be considered, particularly if there is a contraindication to ERCP or if prior ERCP was unsuccessful. [108]

The decision to pursue ERCP, PTC, or EUS should depend on the presumed site of obstruction, the presence of coagulopathy or ascites, and the local expertise of the radiologists and gastroenterologists. PTC and ERCP rarely are used in combination. Benign strictures should be differentiated from cholangiocarcinoma, which often requires cytologic analysis or biopsy of the lesion.

Liver Biopsy

If high-grade extrahepatic obstruction has been excluded or hepatocellular disease is strongly suspected, a liver biopsy should be performed. [109] Liver biopsy can correct 20% of errors in clinical diagnosis. Liver biopsy kits usually include the Jamshidi (suction) needle, Klatskin needle, or Tru-Cut needle. Complications after liver biopsy occur with an incidence ranging from 0.1% to 3.0%. [110], [111] The nature of liver biopsy complications warranting hospital admission are pain, hypotension, hemoperitoneum, hemobilia, pneumothorax or hemothorax, and intrahepatic arteriovenous fistula. Overall, this is a safe procedure, but the risk-to-benefit ratio needs to be carefully assessed and explained to the patient. The use of ultrasound before the procedure may reduce the complication rate. [112] Patients with coagulopathy, thrombocytopenia, or ascites may require blood products or an alternative route for biopsy, such as the transjugular approach. [113] For patients with renal insufficiency or patients who were taking warfarin (Coumadin), an ultrasound-guided liver biopsy with desmopressin acetate or a gelatin foam plug may reduce the risk of serious bleeding complications.

In the workup of a patient with hyperbilirubinemia, a liver biopsy can be useful if other diagnostic tests are unrevealing. This diagnostic strategy is essential. If the ERCP result is nondiagnostic, a liver biopsy should be performed. Five percent of the cases of extrahepatic cholestasis are diagnosed by liver biopsy because of inadequate clinical suspicion of obstruction or an inability to visualize the ducts adequately. [114] For 15% of cases, a liver biopsy is not helpful in determining the cause of the hyperbilirubinemia. If the clinician's level of suspicion is high and the ultrasound scan is negative, a cholangiogram should be performed.

The decision tree that the clinician follows depends to a great extent on pretest probability. If there is a low suspicion of extrahepatic obstruction and the ultrasound scan is negative, further evaluation of possible dilated ducts probably is not warranted. Clinical instinct should not be ignored, however, if the radiographic tests do not confirm the physician's suspicions. Judgment based on the patient's history and physical examination is dependable in evaluating patients with jaundice. Diagnostic accuracy with subsequent adequate care relies on the judicious use of appropriate confirmatory tests and radiographic studies.

COMPLICATIONS OF CHOLESTASIS

Pruritus

Pruritus is commonly associated with cholestasis and may limit activity, cause anxiety, disturb sleep patterns, and result in secondary skin infection. The presence and severity of pruritus in cholestasis do not necessarily correlate with the degree of cholestasis. The pathogenesis of cholestasis-associated pruritus is uncertain. Formerly, proposed accumulated endogenous pruritogens of cholestasis interacting with cutaneous nerve endings were implicated. More recently, the concept that pruritus is of central origin, mediated by endogenous opioid or serotonin ligands, has gained credence. [115], [116] Conventional agents have not been adequately, objectively evaluated using reliable and validated outcome measures, [117] but they are generally safe, modestly effective, and usually the agents of first choice. Studies have suggested the efficacy of opioid antagonists. [118], [119] and [120] Table 44-2 outlines current management options for pruritus.

Topical Therapy
Lower bathing water temperature and use fewer or lighter clothing and bed coverings
Minimize dry skin by using moisturizing soaps (e.g., Dove) and applying topical moisturizers liberally (e.g., Eucerin cream)
Anion-Exchange Resins
Cholestyramine or colestipol: start with 4 g (one scoop or packet) PO bid, starting before and after breakfast, and increasing to six packets or scoops daily, separated from other medications by 2 h
Bile Salts
Ursodeoxycholic acid, 15 mg/kg/d PO qhs
Doxepin
25–50 mg PO daily
Hepatic Microsomal Enzyme Induction
Rifampin, 300–600 mg PO daily
Opioid Receptor Antagonists
Naltrexone, 12.5 mg PO daily, increasing slowly to 50–100 mg PO daily
Naloxone and nalmefene are only commonly available for parenteral use

TABLE 44-2 Management of Pruritus of Cholestasis

Hepatic Osteodystrophy

Hepatic osteodystrophy is the metabolic bone disease that occurs in patients with chronic liver disease, particularly cholestatic disease. [121] Hepatic osteodystrophy encompasses both osteoporosis and osteomalacia, although the former process is usually dominant. In cholestasis-associated osteoporosis, the precise pathophysiological roles of vitamin D receptor polymorphisms, osteoblast trophic factors such as insulin-like growth factor type I, and osteoprotegerin, an osteoclast regulator, are been actively explored. [121] Increased cytokine activation involving IL-1, IL-6, and tumor necrosis factor may be important in modifying osteoclast or osteoblast function. [122] Osteomalacia results from impaired bile salt–assisted intestinal absorption of ingested vitamin D and consequently calcium and phosphate, and it is inversely associated with the degree of steatorrhea and cholestyramine use. The management of hepatic osteodystrophy is outlined in Table 44-3 and involves the early identification through bone mineral density screening by dual-energy x-ray absorptiometry of all patients with cholestatic liver disease. [123] Intervention with antiresorptive medications is indicated for those with symptomatic fractures or confirmed osteoporosis based on bone density measurement. [124] The use of cyclical etidronate prevented bone loss in patients with primary biliary cirrhosis who were cotreated with corticosteroids and bile salts for 1 year. [125]

Baseline DEXA Scanning
Normal (T/Z Score): <1 SD below the mean
Osteopenia: 1–2.5 SD below the mean
Severe osteoporosis: >2.5 SD below the mean plus one or more fragility fractures
Laboratory Testing
Baseline: 25-OH vitamin D level (normal range 10–55 mg/mL), calcium, phosphate, thyroid function tests, intact parathyroid hormone (free serum testosterone in men and estradiol and luteinizing hormone in women)
Treatment
(Relative contraindication: patients with history of renal stones)
Calcium, 1–1.5 g qd
Before transplantation: calcifediol (Calderol), 20–50 μg three times weekly
After transplantation: calcifediol (Calderol), 20–50 μg weekly
If coexistent renal disease, calcitriol (Rocaltrol), 0.25–0.5 μg PO qd
Estrogen replacement, if no contraindications exist
If bone density worsens rapidly or if there is evidence of severe osteoporosis or symptomatic fractures, treat with bisposphonates or calcitonin
Monitoring
DEXA
For osteoporosis: Repeat 6 mo after treatment initiated and then yearly
For normal study: Repeat every 2–3 y
Repeat 25-OH vitamin D and 24-h urine calcium yearly and treat accordingly

DEXA, dual-energy x-ray absorptiometry; SD, standard deviation.

TABLE 44-3 Management of Hepatic Osteodystrophy

Fat-Soluble Vitamin Deficiency

This condition is common in patients with prolonged cholestasis, and management is outlined in Table 44-4. Replacement will depend on the extent of deficiency and response to therapy. Care should be taken to inform patients of the risks and benefits of therapy, including the need for assessment of hypercalciuria after therapeutic intervention.

Monitoring Fat-Soluble Vitamins
25-OH vitamin D level (normal range, 10–55 mg/mL) (if renal disease, check $1,25\text{-}(OH)_2$ vitamin D)
Vitamin A level (normal range, 360–1200 μg/L)
Vitamin E level, with fasting total lipid profile (normal range, 5.5–17.0 mg/L) (total lipids = cholesterol plus triglycerides [in grams]. To calculate vitamin E level: serum vitamin E [mg]/total lipid [g]. If ≥0.8, normal; if <0.6, supplement)
Vitamin K: measure prothrombin time (normal range, 11.4–13.2 s)
Replacement
Vitamin D: calcium, 1–1.5 gm PO qd
Calcifediol (Calderol) 20–50 μg PO three times weekly, before transplantation (coexistent renal disease, use calcitriol [Rocaltrol] 0.25–0.5 μmg PO qd)
Vitamin A: β-carotene 15 mg (25,000 U vitamin A) PO qd or Aquasol A 50,000 U IM qd for 2 wks
Vitamin E: Liquid E (D-α-tocopherol), water-soluble, 100 IU PO qd
Vitamin K: 10 mg subcutaneously daily, for 3 d, then monthly if cholestatic
Evaluation of Therapy
After 3 mo: 24-h urine calcium (normal range, 50–250 mg/d)
Yearly: 25-OH vitamin D ($1,25\text{-}(OH)_2$ vitamin D if renal disease), vitamins A and E, and prothrombin time

TABLE 44-4 Management of Fat-Soluble Vitamin Deficiency in Prolonged Cholestasis

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CHAPTER 45

Richard H. Moseley

APPROACH TO THE PATIENT WITH ABNORMAL LIVER CHEMISTRIES

The approach to the patient with abnormal liver chemistries is not governed by any well-defined diagnostic algorithms. Instead, a systematic approach to patients with suspected underlying liver disease involves a thorough understanding of the diverse panel of available measurements of liver function and serum markers of hepatobiliary disease. From this panel, a group of indices most appropriate to the particular clinical problem is selected. A single test is rarely sufficient in the approach to most clinical problems. The selection process is, however, facilitated by several distinct patterns of hepatocellular injury. Because diagnostic tests are imperfect, they are usually discussed in terms that allow assessment of their diagnostic value. The *sensitivity* of a test is defined as the likelihood of an abnormal test result in patients known to have a disease, and *specificity* is defined as the likelihood of a normal test result in patients known to be free of the disease. The *false-positive rate* is the likelihood of an abnormal test result in patients without the disease (1- specificity), and the *false-negative rate* is the likelihood of a normal test result in patients known to have the disease (1- sensitivity). Thus, sensitivity and the false-negative rate evaluate a diagnostic test in patients with disease, and specificity and the false-positive rate evaluate a test in patients without disease. [1] This chapter discusses representative and commonly used tests and offers guidelines in the interpretation of results.

CLINICAL EVALUATION

As in most disease states, an accurate history is critical in the approach to the patient with laboratory evidence of liver disease. Although systemic symptoms of liver disease, such as anorexia, weight loss, chills and fever, nausea, and vomiting, are nonspecific and are typically of little help in the differential diagnosis, valuable information can be elicited by questions regarding family history, use of prescription drugs and over-the-counter medications, alcohol consumption, use or abuse of illicit substances, exposure history, sexual and menstrual history, occupational or environmental history, travel history, past surgery (including anesthesia records, if available), and transfusion history.

A family history of jaundice may be present in Gilbert syndrome, Dubin-Johnson syndrome, Rotor syndrome, benign recurrent intrahepatic cholestasis, and hereditary hemolytic states such as hereditary spherocytosis. Familial forms of chronic intrahepatic cholestasis, such as arteriohepatic dysplasia (Alagille syndrome), and of cirrhosis have been well described. Hemochromatosis, Wilson disease (hepatolenticular degeneration), and a $_1$-antitrypsin deficiency are examples of liver diseases transmitted by an autosomal recessive mode of inheritance; genetic factors may also play a role in other hepatobiliary disorders, including primary sclerosing cholangitis, primary biliary cirrhosis (PBC), and autoimmune hepatitis.

Given the relatively nonspecific presentation of drug-induced liver disease, drug-related hepatic injury may not be immediately suspected in a patient with impaired liver function. Difficulties in diagnosis are compounded by the unknown hepatotoxicity of newly introduced agents. Nevertheless, the possibility of drug-induced liver injury should be considered in all patients with a seemingly nonspecific change or worsening in liver chemistries, and such considerations are aided by a complete drug history. Alcohol and nonprescription medication use is an important part of this inquiry. Alcohol intake should be quantified and expressed, if possible, in terms of grams per day of alcohol (daily consumption in milliliters × 0.79 × percentage of alcohol in the form ingested). A threshold for the development of cirrhosis of 80 g/d for 15 years has been described in male patients with alcoholism, [2] and it is likely that a lower threshold exists in women. [3] Acetaminophen hepatotoxicity, secondary to induction of the cytochrome P450–dependent pathway of acetaminophen metabolism by ethanol or to low hepatic glutathione stores in the malnourished patient with alcoholism, is being increasingly recognized. [4] Patients with alcoholism who present with jaundice and profoundly abnormal serum transaminase levels should always be questioned regarding the use of acetaminophen. A high incidence of aspirin-induced hepatotoxicity, which appears to correlate with serum salicylate levels, has been observed in patients with rheumatic diseases, including juvenile rheumatoid arthritis and systemic lupus erythematosus. [5] Hypervitaminosis A is a well-recognized clinical syndrome associated with hepatic injury, intracranial hypertension, and desquamative dermatitis. [6] Although most cases of hepatic injury from vitamin A have occurred with massive long-term intakes, hepatotoxicity may be potentiated by ethanol, severe hypertriglyceridemia, and renal failure, and it may occur at vitamin doses as low as 4000 IU/day. [6] Cases of hepatitis after administration of folk remedies, such as germander (*Teucrium chamaedrys*), Jin Bu Huan Anodyne Tablets (*Lycopodium serratum*), and chaparral, document the need for specific queries about ingestion of herbal products and the use of other unconventional forms of therapy, particularly in the setting of hepatic injury of unknown origin. [7]

A history of recent ingestion of raw oysters or steamed clams should suggest infection with the hepatitis A virus (HAV), although specific risk factors that have been associated with HAV infection within the United States also include homosexual contact [8] and contact with children attending day-care centers. [9] In contrast to the well-recognized problem of nosocomial hepatitis B virus (HBV) infection, nosocomial outbreaks of hepatitis A have received little attention. [10] Questions directed at determining the source of water for patients are occasionally relevant, because private water supplies contaminated with sewage have often been implicated in outbreaks of hepatitis A. HBV infection should be suspected in patients with abnormal liver chemistries and a history of exposure to or contact with jaundiced persons, syringes or needles (including tattoo paraphernalia), or blood or blood products. Intravenous drug users and male homosexuals appear to be particularly susceptible to chronic HBV infection. The development of abnormal liver chemistries in the healthy-appearing HBV carrier warrants strong consideration of superinfection with the delta agent (hepatitis D virus, or HDV). In countries with low endemism for HBV, such as the United States, HDV infection is found chiefly among parenteral drug users. [11] Hepatitis C virus (HCV) is parenterally transmitted, and a higher prevalence occurs in persons with frequent exposure to blood, including patients who received multiple transfusions before 1992, patients with hemophilia, and current and former intravenous drug users. [12] Occupational exposure to HCV from needle-stick accidents [12] and nosocomial spread of HCV [13] should also be considered. In contrast to HBV, HCV transmission by sexual or close physical contact appears to be inefficient and uncommon. [12] Recent travel to areas endemic for viral hepatitis should be noted; water-borne outbreaks of hepatitis E have been clearly

documented in Southeast Asia and the Indian subcontinent. Abnormalities in smell (dysosmia) and taste (dysgeusia) may be noticed by patients afflicted with viral hepatitis. Arthritis, abrupt in onset and with a strong predilection for proximal interphalangeal joints, has been observed during the prodromal phase in approximately 20% of patients with HBV infection. [14] Abnormal liver chemistries may also be a direct manifestation of illicit drug use. The use of the stimulant 5-methoxy-3,4-methylenedioxymethamphetamine (MDMA), or Ecstasy, has been associated with acute hepatitis and fulminant hepatic failure. [15]

Sexually transmitted diseases are an important cause of abnormal liver chemistries, and a sexual history should be included in the evaluation of such patients. Efforts to obtain accurate historical information are usually compromised by apprehension felt on the part of the interviewer rather than the patient. Relevant historical elements include information regarding whether the patient is currently sexually active, the number of sexual partners the patient has had in the preceding 6 months, whether the patient's sexual partners are of the same or different sex, whether the patient has recently had a new sexual partner, and the sites of sexual exposure. A sexual history in the female patient should always include information on contraceptive use, which has been associated with intrahepatic cholestasis, hepatic adenoma, and hepatic vein thrombosis (Budd-Chiari syndrome). A menstrual history may reveal the presence of secondary amenorrhea, a frequent complication of chronic liver disease. Infertility may be a presenting symptom in women with autoimmune hepatitis. [16]

Although the use of hepatotoxins such as carbon tetrachloride, chloroform, and trinitrotoluene has diminished, liver injury associated with accidental and occupational exposure to workplace chemicals remains a significant problem. Although an itemized list is beyond the scope of this text, exposure to industrial and environmental hepatotoxins may be elicited by a thorough occupational history. Examples include the following: trichloroethylene, a commonly used solvent in dry cleaning, which can cause acute centrilobular hepatitis; vinyl chloride, which is used in the plastics industry and is associated with the occurrence of hepatic angiosarcomas; arsenic, used in insecticide sprays by vineyard workers and implicated in chronic liver diseases, including noncirrhotic portal hypertension and hepatic angiosarcoma; and 2-nitropropane, used in industrial construction, highway maintenance, ship building, and plastic production and implicated in instances of fulminant hepatic failure. [17]

The liver may be a target organ in a vast array of systemic disorders. In particular, but by no means exclusively, the presence of coexistent cardiac, pancreatic, or inflammatory bowel disease should be considered in the evaluation of any patient with abnormal liver chemistries. Right-sided congestive heart failure, hypotension, and shock are well-recognized causes of abnormal liver chemistries. Prolongation of the prothrombin time, often disproportionately to other signs of liver dysfunction, is the most common abnormality in patients with congestive heart failure, although elevations in serum bilirubin (primarily of the unconjugated form and rarely more than 3 mg/dL) and in serum aminotransferases can also occur. [18] Clinically inapparent left-sided heart failure or cardiac tamponade may present with a picture like that of acute or chronic hepatitis. [19] Hemochromatosis, in turn, may present as a congestive cardiomyopathy [20] or as hypogonadism, arthropathy, diabetes, or hyperpigmentation. Distal common bile duct stenosis is a well-described complication of chronic alcoholic pancreatitis to be considered in the setting of anicteric alkaline phosphatase elevations of a persistent nature. [21] The biliary tree may be similarly affected in cystic fibrosis. [22] Hepatobiliary manifestations of inflammatory bowel disease of clinical import occur in up to 10% of patients. [23] Hematologic disorders, such as polycythemia rubra vera, myeloproliferative disorders, and paroxysmal nocturnal hemoglobinuria may predispose to hepatic vein thrombosis. Hemoglobinopathies, such as sickle cell anemia and thalassemia, have been implicated as risk factors for pigment stone formation. Bacteremia, particularly with gram-negative organisms or *Staphylococcus aureus,* should be considered in any ill person with direct and total serum bilirubin values that are disproportionately elevated in comparison with levels of alkaline phophatase and aspartate aminotransferases. [24] Bilirubin elevations may become manifest before the clinical recognition of infection, and persistent or progressive hyperbilirubinemia despite antiinfective therapy portends a poor prognosis and may warrant institution of additional therapeutic agents. [25] Leptospirosis should be regarded with a high index of suspicion in the febrile patient with both hepatic and renal abnormalities and a history of potential contact with animal urine or water. Renal cell carcinoma may present with abnormalities in liver chemistries, primarily elevated alkaline phosphatase levels, in the absence of hepatic metastases (nephrogenic hepatic dysfunction syndrome). [26] Liver diseases peculiar to gravid women include intrahepatic cholestasis of pregnancy, toxemia, and acute fatty liver of pregnancy, although viral hepatitis is the most common cause of jaundice during pregnancy. [27] Membranoproliferative glomerulonephritis, chronic lymphocytic sialoadenitis, and essential mixed cryoglobulinemia have been associated with chronic HCV infection. [28] A high prevalence of elevated serum aminotransferases has been reported in patients with gluten-sensitive enteropathy; in most cases, these abnormalities resolve with a gluten-free diet. [29] Rarely, abnormal serum aminotransferases may be the sole feature that leads to the diagnosis. [29] Endocrine disorders are rare but recognized causes of abnormal liver function tests. Hyperthyroidism, independent of congestive heart failure or concomitant unrelated liver disease, can result in jaundice and a prolonged prothrombin time [30]; serum aminotransferases and alkaline phosphatase are typically less than 250 IU/L and less than threefold elevated, respectively, although exceptions have been reported. [30] Elevated serum aspartate aminotransferase levels reported in hypothyroidism are secondary to enzyme release from muscle. [31] Moderate elevations in serum aminotransferases and vague constitutional symptoms may suggest the diagnosis of Addison disease. [32]

The nature of, and indications for, previous abdominal surgical procedures should be fully ascertained. If it is available, information concerning the gross appearance of the liver at the time of operation may prove valuable. In the postoperative patient, surgical and anesthesia records should be carefully reviewed for the inhalational agent administered, the presence and duration of intraoperative hypotension, and the amount of blood product support required. Hepatic injury has been observed with most of the halogen-substituted inhalation anesthetics (e.g., halothane, methoxyflurane, enflurane); it presents initially with fever and is followed by the appearance of jaundice, with or without eosinophilia, after a latent period of several days. [33] Transfusions, particularly of stored blood, can be a factor in the development of postoperative jaundice. Progressive liver disease, including cirrhosis, has been described as a late complication of jejunoileal bypass surgery. [34] Biliary strictures, retained and recurrent stones, or papillary stenosis should be considered in the diagnosis of the abnormal liver chemistries in the patient who has undergone cholecystectomy.

Generalized pruritus may be a presenting symptom in patients with liver disease, particularly cholestatic syndromes. The exact mechanism responsible for this often disabling symptom is unclear. Despite the often favorable response to oral cholestyramine, a bile acid binding agent, there is no apparent correlation between either serum or tissue levels of bile acids and the degree of pruritus. [35] Clinical experience suggests that pruritus in the jaundiced patient is often nocturnal and is most pronounced on the palms and soles. Muscle cramps, also occurring frequently at night, and affecting largely the gastrocnemius muscle and small muscles of the foot, are common in patients with cirrhosis. [36]

The presence, or absence, and character of abdominal pain may provide some clues in the approach to establishing a cause for abnormal liver chemistries. In contrast to the intense and rapidly developing right upper quadrant abdominal pain of acute extrahepatic obstruction, such as occurs in choledocholithiasis, the pain associated with acute viral hepatitis can be best described as a heavy or dragging sensation. Pain from primary and metastatic tumors of the liver may be distinguished by its dull or boring character, although hemorrhage into the tumor may result in the sudden onset of severe pain.

Physical findings of some discriminative value in the patient with abnormal liver chemistries include stigmata of chronic liver disease (e.g., spider angiomata, palmar erythema, parotid gland enlargement, gynecomastia, Dupuytren contracture, testicular atrophy), hepatomegaly and liver consistency, splenomegaly, gallbladder distention, and abdominal tenderness. However, poor interobserver agreement for several of these clinical signs has been reported [37]; for other signs, such as Dupuytren contracture, the correlation with chronic liver disease is poor. [38] Although the degree of hepatomegaly can vary widely in all forms of hepatobiliary disease, a liver span greater than 15 cm is more often associated with passive congestion from right-sided heart failure or neoplastic and infiltrative processes (e.g., amyloidosis, myeloproliferative disorders, hepatic steatosis, and glycogen and lipid-storage disorders). [39] A pulsatile liver may be encountered in tricuspid insufficiency. A hepatic bruit or friction rub should alert the examiner to the possibility of an underlying hepatocellular carcinoma; alternatively, a friction rub can occur with a hepatic abscess or in acute cholecystitis. [40] A hepatic bruit may also be auscultated in acute alcoholic hepatitis. The presence of sunflower cataracts and of Kayser-Fleischer rings (golden-brown or greenish discoloration of the Descemet membrane in the limbic region of the cornea, initially appearing at the superior corneal quadrant) should be sought either with the unaided eye or with slit-lamp ophthalmoscopy, even if the latter finding is no longer considered pathognomonic for Wilson disease. [41] Conjunctival suffusion, with or without hemorrhage, should suggest leptospirosis. A Murphy sign, or inspiratory arrest during deep palpation of the right upper quadrant, is highly suggestive of acute cholecystitis. Punch or fist percussion tenderness can also be elicited in acute cholecystitis (and in acute hepatocellular injury) and may help to differentiate hepatobiliary from pleural-based pain. A distended gallbladder, detected by either inspection or palpation, may be a presentation of malignant obstruction of the common bile duct (the Courvoisier sign).

Jaundice, manifested by yellow pigmentation of the skin, mucous membranes, and sclerae, typically requires a serum bilirubin concentration of greater than 3 mg/dL for detection. Artificial light makes detection at low levels more difficult. Ingestion of foods rich in carotene (e.g., carrots) and lycopene (e.g., tomato juice); drugs such as Atabrine, quinacrine, or busulfan; or toxins such as picric acid may result in similar skin discoloration that is readily differentiated from jaundice by the absence of scleral icterus.

In addition to jaundice and excoriations resulting from pruritus, skin manifestations of potential aid in the differential diagnosis of patients with abnormal liver chemistries include the hyperpigmentation associated with PBC and hemochromatosis, the xanthomata and xanthelasmas present in chronic cholestasis, and the

hypertrichosis of periorbital and malar regions and eczematoid dermatitis of sun-exposed areas in porphyria cutanea tarda.

HEPATIC FUNCTION TESTS

Laboratory determinations that reflect hepatic disease are collectively called *liver function tests.* However, only some are true measurements of hepatic function, and the use of this descriptive term should be discouraged. Tests that examine the ability of the liver to excrete substances into bile, particularly organic anions, fall within this strict definition, as do laboratory assessments of the synthetic and metabolic capacity of the liver.

Bilirubin

Because bilirubin is an endogenous organic anion, derived primarily from the degradation of hemoglobin from senescent erythroid cells, tests of bilirubin metabolism are important in the assessment of hepatic function. [42] Photometric determination of the azo derivatives obtained by reaction of plasma with the diazonium ion of sulfanilic acid (the diazo, or van der Bergh, reaction) separates bilirubin into two fractions, a water-soluble direct-reacting conjugated form and a lipid-soluble indirect-reacting form representing unconjugated bilirubin. Normal plasma total bilirubin concentrations in boys and men are significantly higher than in girls and women, and virtually all the bilirubin normally present in serum is in the unconjugated fraction. Hyperbilirubinemia, clinically manifested as jaundice, can accordingly be classified as either predominantly unconjugated or predominantly conjugated, simply by subtracting direct from total serum bilirubin to estimate indirect, or unconjugated, bilirubin. Increased production of bilirubin, impaired transport into hepatocytes, and defective bilirubin conjugation within the hepatocyte characterize disorders associated with unconjugated hyperbilirubinemia. Up to 85% of total serum bilirubin is in the unconjugated form in these disease states. [39] Along with the rate of hemolysis, the ability of the liver to conjugate bilirubin determines the degree of unconjugated hyperbilirubinemia observed. Even in severe hemolytic disorders, total serum bilirubin rarely exceeds 5 mg/dL in the presence of normal hepatic function. [39] Unconjugated hyperbilirubinemia may also be observed in disease states that interfere with the delivery of bilirubin to the liver, such as congestive heart failure, or in the presence of portosystemic shunts. In contrast, in disorders with impaired intrahepatic excretion of bilirubin (the rate-limiting step in overall bilirubin metabolism) and in extrahepatic obstruction, a conjugated hyperbilirubinemia is observed. Typically, in these settings, more than 50% of the serum bilirubin is in the direct-reacting form. [39]

A direct-reacting fraction of bilirubin that is apparently covalently bound to albumin has also been identified; it is termed *albumin-bound bilirubin* or *delta bilirubin.* [43] This complex represents a significant fraction of total bilirubin in patients with either hepatocellular or cholestatic forms of jaundice if hepatic excretion of conjugated bilirubin is impaired, but it is not present in disorders associated with a predominant unconjugated hyperbilirubinemia. During recovery from jaundice, albumin-bound bilirubin tends to persist in plasma because the complex is minimally filtered by the kidney. [43] This provides an explanation for the slow resolution of jaundice in convalescent patients with otherwise apparently normal liver function.

Urine bilirubin is invariably conjugated bilirubin and is encountered only in conditions in which serum levels of direct or conjugated bilirubin are elevated. The tea-colored appearance of urine caused by the presence of bilirubin must be differentiated from similar discoloration caused by hemoglobinuria and myoglobinuria. Prolonged storage before testing may produce false-negative results; phenothiazine administration can cause false-positive findings. [39] Bilirubinuria may precede the clinical appearance of jaundice, largely because of the low (<1.0 mg/dL) renal threshold for conjugated bilirubin.

Serum Bile Acids

Two primary bile acids, cholic and chenodeoxycholic acid, are synthesized in the liver from cholesterol and are converted by intestinal bacteria to the secondary bile acids, deoxycholic and lithocholic acid. Chenodeoxycholate can also be transformed into the tertiary bile acid, ursodeoxycholate. Serum bile acid determination in the assessment of patients with liver disease has been advocated. Although serum bile acid levels are almost always elevated in moderate to severe liver disease, poor diagnostic sensitivity in patients with mild liver disease has prevented widespread application. [44] The finding of normal fasting levels of cholic acid conjugates may, however, be helpful in supporting a diagnosis of Gilbert syndrome in patients with unconjugated hyperbilirubinemia. [45] Higher sensitivity of serum bile acid levels, as compared with conventional tests, has also been demonstrated in the detection of patients with cirrhosis, [46] reflecting decreased first-pass elimination resulting from portosystemic shunting. [47] Elevated serum levels of bile acids in these patients may also have prognostic implications. [48] The increase in serum bile acids that uniformly occurs as a result of diminished hepatic uptake or biliary excretion may be absent in patients with coexisting ileal disease that interferes with the intestinal phase of the enterohepatic circulation of bile acids. Conversely, small intestinal bacterial overgrowth may elevate serum bile acid levels. [49] Furthermore, serum bile acid determination appears to be of no value in assessing the patency of surgical portosystemic shunts. [50]

Dye Tests

Sulfobromophthalein (BSP) is a cholephilic organic anion previously used to assess hepatic function. The only current clinical application of the BSP plasma disappearance test is in the diagnosis of the inherited conjugated hyperbilirubinemic states, Dubin-Johnson syndrome and Rotor syndrome. [51] Anaphylactic reactions have been reported after BSP injection, largely limiting studies of BSP uptake and biliary excretion to research settings.

Indocyanine green (ICG) is a less toxic dye with hepatic uptake and excretion characteristics similar to those of BSP. However, because of greater hepatic clearance, ICG appears to be a less sensitive indicator of mild hepatic dysfunction than BSP. [52] Negligible removal by extrahepatic tissues makes ICG an ideal indicator of hepatic blood flow. [53]

Clotting Factors

Liver disease is a common cause of impaired coagulation. Normal serum activities of the vitamin K–dependent coagulation-factor proenzymes (factors II, VII, IX, and X), as assessed by the one-stage prothrombin time, depend on both intact hepatic synthesis and adequate intestinal absorption of lipid-soluble vitamin K. Vitamin K is required for the posttranslational formation of ?-carboxyglutamyl residues that are essential for physiological activation of the factors. [54] Prolonged prothrombin times can be observed both in hepatocellular disorders that impair hepatic synthetic function, such as hepatitis and cirrhosis, and in cholestatic syndromes that interfere with lipid absorption. Hepatocellular injury can be differentiated from cholestatic causes of prothrombin time prolongation by the parenteral administration of vitamin K [55]; intact hepatic function is established by an improvement in prothrombin time greater than 30% within 24 hours of administration. A prolonged prothrombin time may occur in the absence of liver disease, such as in consumption coagulopathies; anticoagulant, antibiotic, or cholestyramine use; steatorrhea; and, rarely, dietary deficiency of vitamin K. Correction of the abnormal prothrombin time by parenteral vitamin K is observed in these conditions. Prolongation of prothrombin time in acute hepatocellular injury signifies severe hepatocellular necrosis, may antedate other manifestations of hepatic failure, and is associated with a poor prognosis. [56] Similarly, in chronic liver disease, a prolonged prothrombin time carries a poor long-term prognosis. [57] Plasma concentrations of individual proteins may be useful clinical guides; because of its short half-life, factor VII is considered the best index of severity of liver disease and of prognosis. [58] A characteristic pattern of hemostatic abnormalities occurs in patients with severe liver dysfunction; it consists of a low plasma fibrinogen level, a prolonged prothrombin time, and a normal or prolonged partial thromboplastin time. A hepatoma-associated dysfibrinogen, similar to fetal fibrinogen, has been described that produces prolonged prothrombin, thrombin, and reptilase times and inhibition of normal plasma coagulation. [59]

Albumin

Albumin is quantitatively the most important of several plasma proteins formed in the liver. Accordingly, measurement of total concentration of serum albumin is a useful test of hepatic synthetic function. The relatively long half-life of serum albumin (20 days) makes the serum albumin level a better index of severity and prognosis in patients with chronic liver disease than in patients with acute hepatic injury, in whom levels are usually normal or only minimally depressed. [52] Nutritional factors, namely, the availability of amino acids, are critical determinants of the rate of albumin synthesis. [60] Moreover, alterations in serum albumin levels may reflect not only disturbances in synthesis but also changes in the rate of catabolism, dilution by expanded plasma volume (e.g., in cirrhosis), or enhanced loss from the gastrointestinal tract or kidneys. Serum albumin levels are also decreased during normal pregnancy. [61] The shorter half-life (1.9 days) of prealbumin, a glycoprotein synthesized by the liver with a faster electrophoretic migration than albumin, was exploited to demonstrate that serum prealbumin levels may be a sensitive index of liver function after acetaminophen overdose. [62]

Immunoglobulins

Although measurement of serum globulins does not fulfill the operational definition of a liver function test, the hypergammaglobulinemia that is commonly observed in patients with liver disease indirectly represents functional impairment of the reticuloendothelial cells of the hepatic sinusoids. [52] Nondiagnostic immunoglobulin abnormalities can be detected in most acute and chronic forms of liver disease, with drug-induced and extrahepatic cholestasis being notable exceptions. Although there is considerable overlap, hypergammaglobulinemia greater than 3.0 g/dL in a patient with chronic hepatitis is more consistent with autoimmune liver disease than with viral hepatitis. Rarely, the hypergammaglobulinemia in autoimmune hepatitis may be so pronounced that it causes the hyperviscosity syndrome. [63] A predominant rise in the IgA fraction is observed in hypergammaglobulinemia associated with alcoholic cirrhosis; a disproportionate elevation of IgM is a feature that differentiates PBC from other liver diseases, specifically chronic active hepatitis, that are associated with prominent hypergammaglobulinemia. [64] However, a specific diagnosis is rarely established by quantitative determinations of immunoglobulins. The demonstration of hyperglobulinemia on serum protein electrophoresis is only a clue to the presence of chronic liver disease. [52] Conversely, hypoglobulinemia should suggest protein-losing enteropathy.

Lipoproteins

The pivotal role the liver plays in normal lipoprotein and cholesterol metabolism is reflected in the characteristic finding of abnormal lipoproteins and mild hypertriglyceridemia in acute forms of hepatocellular injury. Decreases in hepatic lecithin-cholesterol acyltransferase activity appear to account for the absence of a and pre-ß bands on lipoprotein electrophoresis commonly associated with acute viral hepatitis and alcoholic hepatitis. [65] Alterations in hepatic triglyceride lipase activity may result in the characteristic elevation in low-density lipoprotein triglyceride, which, in turn, gives rise to the broad ß electrophoretic band observed in these disorders. [66] Abnormalities in serum lipoproteins in chronic forms of liver disease are a reflection of the degree of ongoing liver injury. [52] Hypocholesterolemia is observed in acute and chronic forms of hepatic insufficiency and is associated with a poor prognosis. [67] In contrast, cholestasis is associated with hypercholesterolemia, as a result of increases in unesterified cholesterol, and the appearance of an abnormal lipoprotein, lipoprotein-X. [68] The origin of lipoprotein X appears to be biliary vesicles destined for canalicular secretion that in the setting of cholestasis are instead transcytosed to the sinusoidal membrane and released into the blood. [69]

Tests of Hepatic Metabolism

Drug metabolism is another critical hepatic function, and liver disease is frequently associated with impaired drug metabolism. The most widely performed tests of hepatic metabolic capacity are the antipyrine clearance determination and the aminopyrine demethylation breath test. Antipyrine is a minor analgesic that, on the basis of rapid and complete absorption from the gastrointestinal tract, distribution in total body water, and minimal nonhepatic elimination, would seem to qualify as an ideal probe for studies of hepatic drug metabolism. Impaired antipyrine metabolism by the cytochrome P450 oxidase system, however, appears to be more a reflection of chronic active liver disease; the extent of impairment correlates well with serum albumin and prothrombin time determinations and with the degree of necrosis and inflammation on liver biopsy. [70] Little or no impairment in antipyrine metabolism is observed in patients with acute hepatitis or well-compensated cirrhosis. [70] The aminopyrine breath test avoids the need for multiple blood determinations. [^{14}C]aminopyrine is demethylated, through a [^{14}C]formaldehyde intermediate, to $^{14}CO_2$, which is measured in expired air. Single-sample, 2-hour breath $^{14}CO_2$ determinations (expressed as a percentage of the administered dose) are significantly decreased in patients with both acute and chronic hepatocellular injury but are normal to minimally decreased in patients with intrahepatic and extrahepatic cholestasis without hepatocellular injury. [71] The aminopyrine breath test has also been used as a prognostic test in patients with alcoholic hepatitis, a value of greater than 1% of the administered dose correlating with improved 3-week survival. [72] Additional measurements of functional hepatic mass include the galactose elimination capacity, which demonstrates significant correlation with albumin synthesis in patients with cirrhosis, [73] the caffeine and phenacetin breath tests, and measurement of the hepatic conversion of lidocaine to its primary metabolite, monoethylglycinexylodide. Although these quantitative tests may be noninvasive predictors of hepatic histology, interindividual differences in the metabolism of a single drug and intraindividual differences in the metabolism of different drugs make interpretation difficult, and it is unlikely that these tests will supplant percutaneous liver biopsy, for example, in the diagnostic approach to the patient with liver disease.

There is increasing evidence to suggest that susceptibility to hepatotoxic drug reactions or disease states is related to a genetically determined capacity for oxidative metabolism. [74] Noninvasive tests have been developed that identify significant interpatient differences in hepatic concentrations and activities of certain forms of cytochrome P450 that underlie this polymorphism in oxidative drug metabolism. [75] The high incidence of impaired sulfoxidation in patients with PBC [76] may be a pathophysiological manifestation of this polymorphism.

SERUM MARKERS OF HEPATOBILIARY DYSFUNCTION

As has been discussed, routine biochemical laboratory tests are not true indices of hepatic function. Instead, they serve as markers of hepatobiliary dysfunction resulting from hepatocellular necrosis, cholestasis, or infiltrative processes.

Aminotransferases (Transaminases)

Aspartate aminotransferase (AST) and alanine aminotransferase (ALT), measured by the serum glutamic-oxaloacetic transaminase (SGOT) and serum glutamic-pyruvic transaminase (SGPT) tests, respectively, are important markers of hepatocellular injury. Whereas AST can be found in various tissues, notably cardiac and skeletal muscle, kidney, and brain, ALT is limited primarily to the liver. Within the liver cell, AST is present in two isozymic forms, one in mitochondria and one in the cytosol, [77] but ALT is localized to the cytosol. In normal serum, most of the AST activity is accounted for by the cytosolic isoenzyme. [78] Normal serum ALT levels vary according to sex and body mass index, with higher values found in male patients and in persons whose body mass index is greater than 23. [79] Given the tissue distribution of these two enzymes, elevations of serum ALT are a more specific reflection of hepatocellular disease than are elevated serum AST levels. The latter is more often elevated after acute myocardial infarction, and elevations in ALT levels that occur in this setting are commonly the result of hepatic ischemia brought on by extensive myocardial injury, congestive heart failure, or cardiogenic shock. [80] Patients with idiopathic inflammatory myopathies, such as polymyositis and dermatomyositis, may, however, present with elevated ALT levels in association with elevated creatine phosphokinase as a reflection of muscle, and not hepatic, injury. [81] The highest serum elevations of both enzymes are seen in patients with viral, toxin-induced, and ischemic hepatitis, whereas smaller (<300 U/L) elevations relative to the degree of histological necrosis are usually encountered in alcoholic hepatitis. [80] The AST/ALT ratio in serum is also regarded as a useful indicator, with a ratio greater than 2 being highly suggestive of alcohol-induced hepatic injury. [82] In contrast, in patients with acute or chronic viral hepatitis or extrahepatic biliary obstruction, an AST/ALT ratio of less than 1 is typically observed, although a correlation between an AST/ALT ratio greater than 1 and the presence of underlying cirrhosis has been described in patients with chronic hepatitis B infection. [83] An AST/ALT ratio of greater than 1 in the setting of nonalcoholic chronic liver disease should raise suspicion regarding underlying cirrhosis, but in the presence of cirrhosis, the AST/ALT ratio may be less useful in differentiating alcoholic from nonalcoholic forms of liver disease. Several mechanisms have been proposed for the disproportionate elevation of serum AST levels in alcoholic liver disease. Hepatic ALT activity in this setting is diminished to a greater extent than hepatic AST activity. [84] Pyridoxal 5'-phosphate is necessary for the activity of both aminotransferases, and there may be enhanced sensitivity of hepatic ALT to alcohol-induced pyridoxine deficiency. [85] Preferential alcohol-induced injury to mitochondria enriched in AST is an alternative hypothesis. Impaired plasma clearance of AST by sinusoidal cells [86] may play a role in the relative increase in serum AST levels observed in cirrhosis. [83]

Although these indices of hepatocellular injury are not predictive of histological findings, serial determinations of serum AST and ALT levels may reflect the extent of hepatocellular injury and are useful in following the progression of liver disease. However, decreases in AST and ALT levels in serum may be either a sign of recovery from an acute injury or, particularly in the case of fulminant hepatic failure, an indication of limited hepatic reserve after overwhelming hepatocyte necrosis. In addition, hypoaminotransferasemia in patients with chronic renal failure who are undergoing hemodialysis has been found to obscure the diagnosis of HCV infection. [87] False elevations of AST have been observed in patients receiving *para*-aminosalicylic acid and erythromycin, and AST may, rarely, exist as a macroenzyme by forming a complex with immunoglobulin, leading to an otherwise unexplained elevation in serum AST activity. [88]

Serum levels of the mitochondrial isoenzyme of aspartate aminotransferase (mAST) and the ratio of mAST to total AST have been reported to be specific and sensitive markers of chronic alcoholism. [89] Until these and other new markers of alcohol abuse, such as desialylated transferrin, are better evaluated, the ?-glutamyltransferase determination, in combination with mean corpuscular volume and serum AST levels, remains the recommended biochemical indicator of recent alcohol abuse. [90]

Other Enzyme Markers of Hepatocellular Injury

Within the liver, the mitochondrial enzyme, glutamate dehydrogenase (GDH), is preferentially localized in centrizonal hepatocytes. The observation that alcohol exerts a toxic effect predominantly on mitochondria in centrizonal hepatocytes may account for the finding, in a large series of patients with alcoholism, that serum GDH determination was more useful than serum AST levels in diagnosing patients with histologically documented alcoholic hepatitis. [91] Serum GDH levels have also been reported to be increased in the congestive hepatopathy observed in acute right-sided heart failure. [91] However, the specificity of GDH as a marker of pericentral hepatocellular necrosis in alcoholic liver disease is open to question, [92] and serum GDH determinations have not received widespread application.

Alkaline Phosphatase

In the liver, alkaline phosphatase appears to be an integral enzyme of the exterior surface of the bile canalicular membrane. [93] Although hepatocellular injury invariably results in increases in serum aminotransferase activity, significant (fourfold or greater) elevations of serum alkaline phosphatase activity are typically observed in patients with cholestatic syndromes. Lesser increases in serum alkaline phosphatase levels lack specificity and may be present in all forms of liver disease. The major mechanism underlying these elevations is increased synthesis, through enhanced mRNA translation, [94] of hepatic alkaline phosphatase, rather than impaired biliary secretion of the enzyme. The mechanisms by which increased hepatic alkaline phosphatase activity leads to elevations in serum activity are less clear. Alkaline phosphatase contained within the bile canalicular membrane may be solubilized by bile acids that accumulate during cholestasis; these, in turn, alter the permeability characteristics of the intercellular tight junctions. [95] Alternatively, the distribution of hepatic alkaline phosphatase activity may be altered, again by the high intrahepatic concentrations of bile acids in patients with cholestasis, so it is found in all domains of the hepatocyte plasma membrane and enters serum directly from the plasma membrane. [95]

Alkaline phosphatase activity can also be demonstrated in bone, placenta, intestine, kidney, and leukocytes. Liver and bone are the predominant sources of serum alkaline phosphatase activity in normal subjects, with less than 20% derived from the intestine. Elevations of the intestinal isoenzyme occur in several disorders, including chronic renal failure, as well as in individuals secreting the ABH red blood cell antigen and in those of B and O blood groups. [96] In pregnancy, a substantial fraction may be derived from the placenta. Low levels of serum alkaline phosphatase have received comparatively less attention but may be encountered in hypothyroidism, pernicious anemia, zinc deficiency, and congenital hypophosphatasia. Decreased serum alkaline phosphatase levels have also been observed in acute hemolytic anemia complicating Wilson disease. [97] Benign familial elevation of serum alkaline phosphatase in a pattern suggesting autosomal dominant inheritance has been reported. [98] Ectopic production of an alkaline phosphatase isoenzyme (Regan isoenzyme) occurs in patients with cancer, and elevations in serum alkaline phosphatase levels may, therefore, be observed in the absence of bony or hepatic metastasis. [99] Similarly, patients with stage I and II Hodgkin disease, osteomyelitis, and congestive heart failure have been found to have marked elevations in serum alkaline phosphatase levels in the absence of hepatic involvement. [100]

Other Enzyme Markers of Cholestasis

Although the alkaline phosphatase isoenzymes exhibit varied susceptibility to heat inactivation and separation is possible with polyacrylamide gel electrophoresis, [101] alternative approaches to determine the source of an elevated serum alkaline phosphatase level are commonly used. Serum ?-glutamyltransferase determination establishes the hepatic origin of an elevated alkaline phosphatase level by virtue of its localization within the hepatobiliary tree, as well as kidney, pancreas, and intestine. Alcohol ingestion produces elevated serum enzyme levels, presumably by enzyme induction, and this finding has been invoked as a sensitive marker of chronic alcohol consumption that occurs independently of any liver damage. [102] However, sensitivity varies from 30% to 80%, depending on the population studied, and elevated serum ?-glutamyltransferase levels are also encountered in pancreatic disorders, myocardial infarction, uremia, chronic obstructive pulmonary disease, rheumatoid arthritis, and diabetes mellitus, as well as in patients using microsomal enzyme-inducing drugs such as barbiturates, phenytoin, and warfarin.

Determination of serum 5'-nucleotidase or leucine aminopeptidase levels fulfills a role similar to that of serum ?-glutamyltransferase determination. Despite their presence in a wide variety of other body tissues, elevated enzyme levels in the nonpregnant patient are specific for hepatobiliary disease and correlate well with elevated alkaline phosphatase levels of hepatic origin. Serum leucine aminopeptidase levels are elevated in pregnancy, [103] and conflicting data exist concerning 5'-nucleotidase levels in pregnancy. In patients with cancer, elevated 5'-nucleotidase levels are a sensitive marker in the diagnosis of metastatic disease to the liver. [104] A normal 5'-nucleotidase level does not exclude liver disease in the setting of an elevated alkaline phosphatase level, because these enzyme markers may not increase in parallel in early or mild hepatic injury. [105]

Lactate Dehydrogenase

Although commonly available, measurement of total serum lactate dehydrogenase (LDH) has limited diagnostic specificity for hepatocellular disease, and fractionation of serum LDH to determine levels of the isoenzyme of hepatic origin (LDH_5) is rarely indicated. Moderate elevations of serum LDH are frequently encountered in hepatocellular disorders such as viral hepatitis and cirrhosis and are less common in cholestatic disorders. However, marked elevations of serum LDH are observed in ischemic hepatitis, and an ALT/LDH ratio of less than 1.5 may differentiate ischemic hepatitis from acute viral hepatitis. [106]

DISEASE-SPECIFIC MARKERS

The laboratory tests outlined earlier alert the physician to the presence of hepatobiliary disease. In the sections that follow, additional markers of specific disorders are discussed.

Viral Serology

Diagnosis of acute hepatitis A is based on serologic detection of HAV-specific IgM antibody (anti-HAV IgM). Seropositivity first becomes detectable at the onset of clinical illness and is invariably present at the onset of jaundice. This serologic marker typically persists for 120 days, far exceeding both clinical and biochemical resolution of illness, and prolonged periods of seropositivity (>200 days) have been observed. [107] Nevertheless, it is best regarded as a marker of acute or recent HAV infection. In contrast, anti-HAV IgG is present primarily in convalescent sera and persists for long periods after infection, perhaps for life.

Certain serologic tests are available to establish a diagnosis of HBV infection. Hepatitis B surface antigen (HBsAg) is the first marker detectable in serum, preceding elevations in serum aminotransferases as well as the onset of symptoms. This antigenemia typically lasts for 1 to 2 months in self-limited infections. The titer of HBsAg, although not routinely reported, appears to be inversely related to the degree of hepatic inflammation. Persistence of HBsAg beyond 24 weeks is associated with a chronic carrier state, although persistence of HBV may also occur in the absence of any conventional serologic marker. [108]

Antibody to core antigen (anti-HBc) is detected in serum approximately 2 weeks after the appearance of HBsAg; typically, a window or lag period then occurs before the appearance of specific antibody to HBsAg (anti-HBs). During this period, and in the 10% of patients who do not manifest detectable levels of HBsAg, anti-HBc may be the only detectable serologic marker of recent infection with HBV. The highest titers of anti-HBc occur in patients with the longest periods of HBsAg positivity. Antibody to the core antigen of HBV of the IgM class (anti-HBc IgM) is the most sensitive marker of acute hepatitis B. [109] The specificity of anti-HBc IgM as a test, however, is lessened by its persistence, at low levels, in some patients with chronic active hepatitis B. [110] In acute hepatitis B, the hepatitis B e antigen (HBeAg) and the other direct marker of viral replication, HBV DNA, are detectable in serum shortly after HBsAg. However, tests of these markers of active viral replication should be reserved for patients with chronic hepatitis B where they are used in determining and monitoring treatment. Testing for HBV DNA is also useful in the identification of HBV as the cause of liver disease in HBsAg-negative patients, including patients with fulminant hepatitis B, who have early clearance of HBsAg, patients with cryptogenic cirrhosis [111] and hepatocellular carcinoma, [112] and patients infected with HBV mutants. [113]

Diagnosis of blood-borne non-A, non-B hepatitis previously relied on the serologic exclusion of HAV, HBV, and other hepatotrophic viruses such as cytomegalovirus, herpes simplex, and Epstein-Barr virus. With the identification of HCV, initial screening now relies on detection of circulating antibody to HCV using enzyme-linked immunosorbent assays (ELISAs). Hypergammaglobulinemia and connective tissue disorders are associated with false-positive results, and recombinant immunoblot assay can be used to confirm ELISA results. [114] False-negative ELISA results may occur in immunocompromised patients and patients undergoing long-term hemodialysis. [115] Qualitative HCV RNA by polymerase chain reaction confirms the diagnosis and is particularly useful in patients with acute hepatitis C, before an

antibody response is detectable, and in monitoring during and after treatment. Genotyping and quantitative HCV RNA tests are performed only before treatment.

Hepatitis D should be considered in any HBsAg-positive patient with acute or chronic hepatitis, especially if the disease is severe or the patient is in a high-risk group. Serologic confirmation of hepatitis D coinfection or superinfection is accomplished by testing for HDV antigen (HDV RNA) and anti-HDV antibodies. A minimum of one acute and one convalescent serum sample should be assayed, because anti-HDV antibodies can be transient, can be present at low titer, and can appear late in infection. Total (predominantly IgG) antibodies to HDV are usually detectable in high titer in superinfections. Persistence of anti-HDV IgM typically predicts progression to chronic HDV infection. Detection of HDV RNA by reverse transcriptase–polymerase chain reaction is the method of choice for the diagnosis of ongoing HDV infection. [11]

Acute hepatitis E infections have increasingly been reported in the United States in persons returning from international travel. [116] Diagnosis rests on detection of antibodies to hepatitis E viral antigens (anti-HEV).

Immunologic Tests

Immunologic abnormalities occur in a wide spectrum of liver diseases. The antinuclear antibody (ANA) reaction in autoimmune hepatitis is of the homogenous pattern by immunofluorescence, and a titer of 1:160 or higher is usually required for diagnosis. Antibodies to double-stranded (native) DNA occur in more than 40% of patients with autoimmune hepatitis. [117] However, these antibodies are also present in similar percentages in patients with acute and chronic hepatitis B, suggesting that they represent a response to DNA released from hepatocyte necrosis. [118] Antimitochondrial antibodies (AMA) are present in more than 90% of patients with PBC and in about 25% of patients with autoimmune hepatitis and drug-induced liver injury. [119] An AMA titer higher than 1:40, even in the absence of serum alkaline phosphatase elevation or symptoms, is strongly suggestive of PBC. [120] Four major mitochondrial antigens related to PBC have been described: M2 antigen on the inner mitochondrial membrane, identified as the dihydrolipoamide acyltransferase of the branched-chain a-keto acid dehydrogenase complex, [121] and M4, M8, and M9 antigens on the outer mitochondrial membrane. he specific profile of antibodies to these antigens in patients may have clinical and prognostic importance. [122] Antibodies to the soluble Ro antigen [123] (often present in patients with Sjögren syndrome and systemic lupus erythematosus) and anticentromere antibodies [124] have also been identified in patients with PBC, particularly those with extrahepatic autoimmune disorders such as sicca syndrome and limited scleroderma, respectively.

Anti–smooth muscle antibodies, reactive to S actin, may be detected in up to 70% of patients with autoimmune hepatitis, in approximately 50% of patients with PBC, and, occasionally, in patients with acute viral hepatitis. [125] They may be the only immunologic marker present in 26% of patients with autoimmune hepatitis. [126] The presence of anti–liver/kidney microsomal antibodies (anti-LKM1), along with absent or low-titer antiactin or antinuclear antibodies, serves to identify a subset of patients with idiopathic autoimmune hepatitis characterized by a more aggressive course and a predominance in young women. [127] The antigen to which anti-LKM1 is directed has been identified as the polymorphic cytochrome P450 isozyme, CYP2D6. [128] This form of autoimmune hepatitis, termed type 2, is most prevalent in western Europe and is rare in the United States. [126] Similarly, anti-LKM2 antibodies directed against cytochrome CYP2C9 have been described in patients with hepatitis and concomitant administration of the diuretic tienilic acid (ticrynafen). [128]

Unlike PBC, a specific autoantibody useful in the diagnosis of primary sclerosing cholangitis has not been identified. Although more than 80% of patients with primary sclerosing cholangitis have detectable perinuclear antineutrophil cytoplasmic antibodies, considerable overlap with autoimmune hepatitis limits its use as a diagnostic tool. [129]

Human leukocyte (HLA) antigens have been associated with a wide spectrum of liver diseases. Specifically, HLA-B8 and HLA-DRw3 have been associated with autoimmune hepatitis. [130] However, the increased frequencies of these HLA haplotypes are neither absolute nor diagnostic. Although HLA typing provides information on gene frequencies, it is neither routinely performed nor recommended.

Copper Storage Parameters

Determination of the serum concentration of ceruloplasmin, a copper transport protein in plasma, is particularly useful in the diagnosis of Wilson disease. Although it is not directly involved in the pathogenesis of this autosomal recessively inherited copper storage disorder, low levels of ceruloplasmin (<20 mg/dL) are found in approximately 90% of homozygotes and in about 10% of heterozygotes. In contrast, serum ceruloplasmin is typically elevated in PBC, another disorder associated with increased hepatic copper concentrations. [131] Increased serum levels in this disorder and other forms of liver disease reflect the role of ceruloplasmin as a nonspecific acute phase reactant. Accordingly, normal values may occasionally be observed during the chronic active hepatitis phase of Wilson disease. [132] Pregnancy and exogenous estrogen administration may also lead to elevated values for this protein. [133] Likewise, hypoceruloplasminemia may result from the diminution in hepatic synthetic function observed in non-Wilsonian fulminant hepatic injury, [134] in chronic hepatitis, [135] and, less commonly, in severe malnutrition, other protein-losing states, and Menkes syndrome. [133] A high incidence of low ceruloplasmin levels has been described in otherwise healthy members of a single family; this benign disorder has been termed *hereditary hypoceruloplasminemia* to differentiate it from Wilson disease. [136] In contrast, an autosomal recessive disorder of iron metabolism has been described in which serum ceruloplasmin is absent. [137] Excessive iron deposition, predominantly in the brain, liver, and pancreas in patients with aceruloplasminemia, leads to movement disorders and diabetes and may be mistaken for nonclassical forms of Wilson disease. [137]

Alternative diagnostic tests in Wilson disease include determinations of urinary copper excretion, serum copper levels, and quantitative hepatic copper content in a liver biopsy specimen. In almost all patients with symptomatic Wilson disease, urinary copper excretion exceeds 100 µg/24 h. Total serum copper is composed of a predominant (~90%) fraction that is irreversibly bound to ceruloplasmin and a small fraction that is loosely bound to albumin and to amino acids. The latter, termed the *free* or *nonceruloplasmin copper,* is calculated by subtracting the amount of copper associated with ceruloplasmin (0.047 mmol/mg) from the total amount of serum copper. Normally, this value is less than 10 µg/dL; it is markedly elevated in Wilson disease, although total serum copper concentrations may be normal, given the profound decrease in ceruloplasmin-bound copper. [138], [139] Ceruloplasmin-bound serum copper can also be calculated by multiplying the ceruloplasmin concentration (in mg/dL), determined by an oxidase assay, by three. [139] Because copper is preferentially excreted in bile, caution is advisable in the interpretation of the results of these two tests in patients with chronic cholestasis or cirrhosis. Patients with untreated Wilson disease have hepatic copper levels greater than 250 µg/g dry weight.

Despite the discovery of the gene for Wilson disease, designated *ATP7B,* molecular diagnosis by direct mutation analysis has been complicated by the lack of a single dominant mutation among the more than 60 disease-specific mutations identified. [140] However, analysis of inheritance of highly polymorphic satellite markers surrounding the *ATP7B* gene has been useful in confirming or refuting Wilson disease in family members of affected patients. [141], [142]

Iron Storage Parameters

Measurements of serum iron level and total iron-binding capacity (or transferrin) are useful in the diagnosis of hereditary hemochromatosis (HHC). Transferrin is normally 20% to 45% saturated, and both the serum iron level and percentage of saturation of transferrin are elevated early in the course of this disorder. A transferrin saturation of 45% or greater is a useful screening threshold because it identifies 98% of affected persons while producing few false-positive results. [143] However, these tests have a relatively low degree of specificity in patients with liver disease; increased serum iron levels, with normal transferrin saturation, are commonly observed in patients with alcohol-induced liver injury. [144] Acute elevations in serum iron levels have also been observed in acute viral hepatitis. [145] Transferrin synthesis is inversely correlated with total body iron stores; hence transferrin levels decrease in iron overload and increase in iron deficiency. [146] Factors other than iron balance, however, affect transferrin levels. Transferrin levels are decreased in inflammation and chronic disease and are increased during pregnancy and with estrogen therapy. [146] False-positive elevations in transferrin saturation are frequently observed if a nonfasting specimen is obtained. [146] Assays of serum ferritin may more closely estimate hepatic and total body iron stores, [147] and elevated serum ferritin levels are commonly observed early in the course of HHC, even before there is any histological evidence of liver injury. [148] Ascorbic acid deficiency in patients with iron overload may lead to inappropriately low serum ferritin levels, [149] and several families have been described in which asymptomatic relatives of patients with HHC had normal serum ferritin levels despite evidence of moderate hepatic iron overload. [150] Because serum ferritin is an acute phase reactant, other forms of hepatocellular necrosis and systemic infection can be associated with elevated serum ferritin levels disproportionate to body iron stores. [151] Furthermore, in chronic hepatitis, elevated serum iron and ferritin levels in the absence of increased hepatic iron stores most likely represent increased release of iron from damaged hepatocytes. [152] Increases in serum vitamin B_{12} levels reflect release from necrotic hepatocytes and may be used to interpret elevated serum ferritin levels. [153] As a general guideline, in the presence of inflammation, ferritin levels of less than 50 µg/L usually reflect iron deficiency, and levels greater than 500 µg/L, in association with a transferrin saturation of more than 50%, usually represent HHC. A fasting transferrin

saturation level threshold associated with the homozygous genotype of more than 62% in men and more than 50% in women was exceeded in 4% of men and in 8% of women heterozygous for HHC on the basis of HLA typing. [154] For these reasons, quantitative determination of tissue iron concentration on liver biopsy had been previously required; a hepatic iron index was calculated by dividing the hepatic iron concentration (in mmol/g dry weight) by the patient's age (in years); an index value greater than 1.9 was considered diagnostic of HHC. [155] However, with the identification of the *HFE* gene and a mutation that leads to the substitution of tyrosine for cysteine at position 282 (C282Y) associated with HHC, [156] the role of liver biopsy in patients with hemochromatosis has been modified. Liver biopsy is no longer required for the diagnosis and instead is now used to assess the degree of liver injury. In certain C282Y homozygous patients, liver biopsy may be avoided even for this purpose, because age less than 40 years, serum ferritin levels less than or equal to 1000 µg/L, the absence of hepatomegaly, and normal serum AST and ALT levels reliably predicted the absence of severe fibrosis. [157], [158]

a-Fetoprotein

A sensitive radioimmunoassay for a-fetoprotein (AFP), a major serum protein during fetal life, is employed in the screening for primary hepatocellular carcinoma. About 70% to 90% of patients with hepatocellular carcinoma have elevations in serum AFP, and significant elevations are also observed in patients with germ cell tumors, other gastrointestinal malignant diseases, and nonneoplastic hepatic disorders such as autoimmune, viral, and alcoholic hepatitis and PBC. [159] To enhance the specificity of this test in the diagnosis of hepatocellular carcinoma, a minimum concentration for positivity of 400 ng/mL has usually been assumed, [160] although this arbitrary cutoff may exclude up to one third of patients with biopsy-proven hepatocellular carcinoma. A monoclonal radioimmunoassay may improve the specificity of AFP screening. [161] Although not widely available, assays for des-?-carboxyprothrombin, an abnormal prothrombin, may also be useful in the detection of primary hepatocellular carcinoma. [162]

a $_1$-Antitrypsin

a $_1$-Antitrypsin is a 52-kd glycoprotein that is synthesized in the liver and, to a lesser extent, in monocytes and macrophages, [163] and migrates in the a $_1$-globulin fraction on serum protein electrophoresis. Normal serum levels (150 to 350 mg/dL) may increase postoperatively and in association with inflammation, malignant disease, pregnancy, or estrogen therapy. The principal function of this protein is the inhibition of leukocyte elastase. The single gene coding for the synthesis of a $_1$-antitrypsin is contained within a 10-kb segment of five exons on chromosome 14. [164] More than 25 codominantly expressed alleles have been described at this locus, and the normal phenotype for the protease inhibitor (Pi) system has been designated Pi MM by electrophoretic mobility. Persons homozygous for the electrophoretically slowest of the genetic variants of this protein, designated Pi ZZ, exhibit markedly decreased serum a $_1$-antitrypsin levels and are predisposed to the early onset of chronic active hepatitis and cryptogenic cirrhosis. [165] Heterozygotes (Pi MZ) demonstrate serum levels that are 50% to 60% of normal values. [166] The inability of the hepatocyte to process and secrete the Z protein, which differs from the normal M protein by a single amino acid substitution, [167] results in the characteristic presence of periodic acid-Schiff (PAS)-positive diastase-resistant globules in periportal hepatocytes on percutaneous liver biopsy. The diagnosis of a $_1$-antitrypsin deficiency should be entertained in a patient with a hepatocellular injury pattern to liver chemistry abnormalities if an absent a $_1$-globulin peak is observed on serum electrophoresis and is confirmed by serum a $_1$-antitrypsin activity determination and genetic Pi typing.

Serum Ammonia

Urea formation in the liver, through the Krebs-Henseleit cycle, is required for the disposal of the toxic product of nitrogen metabolism, ammonia. Elevated serum ammonia levels are often observed in both acute and chronic forms of liver disease. The striking elevations seen in fulminant hepatic failure are the result of impaired conversion of ammonia to urea in the setting of severe hepatocellular necrosis, whereas the hyperammonemia present in patients with cirrhosis and portal hypertension primarily reflects portosystemic shunting of ammonia derived from colonic bacteria. [168] Additional factors that influence the level of serum ammonia in patients with cirrhosis include intestinal production of ammonia by bacterial deamination of blood or dietary protein, renal production of ammonia by glutaminase in response to metabolic alkalosis or hypokalemia, intestinal production of ammonia from urea by urease-forming bacteria in the setting of diminished renal function, and hepatic production of ammonia from amino acids in response to increased glucagon secretion. [169] Although routinely determined in patients with suspected hepatic encephalopathy and used as an index of the success of therapy, serum ammonia levels only roughly correlate with the degree of encephalopathy. [169] Hyperammonemia and encephalopathy in the absence of liver disease have been reported in patients with urea cycle enzyme deficiencies, after ureterosigmoidostomy, and in a patient with a neurogenic bladder infected with urease-producing bacteria. [170] Serum ammonia determination is best regarded merely as an aid in the differential diagnosis of encephalopathy, and serial determinations have little role in clinical practice.

Additional abnormalities that can be observed in hepatic encephalopathy include decreased serum levels of branched-chain amino acids and elevated serum levels of aromatic amino acids, methanethiol, and short-chain fatty acids. [171] This serum amino acid profile contrasts with that observed in severe autoimmune hepatitis, in which both aromatic and branched-chain amino acid levels are increased, and it does not distinguish between patients with and without clinical manifestations of hepatic encephalopathy. [171]

Liver Biopsy

Unlike most of the laboratory tests discussed earlier, a predictive value cannot be assigned to a specific morphologic feature observed with a liver biopsy. Yet liver biopsy can be extremely useful in the diagnostic approach to the patient with abnormal liver chemistries. Proper biopsy interpretation is assisted by the availability of all clinical, biochemical, immunologic, and radiographic data to correlate histological features with an etiologic diagnosis. As a general rule, direct forms of liver injury tend to cause predominant centrizonal necrosis, immunologically mediated forms of hepatocyte injury are localized to the periportal regions, and cholestatic liver injury can be recognized by the accumulation of canalicular bile and feathery degeneration of hepatocytes in the absence of a significant inflammatory infiltrate. Major applications of liver biopsy, other than in the evaluation of a patient with persistently abnormal liver chemistries, include the following: establishing the diagnosis in patients with unexplained hepatomegaly; in patients with suspected systemic disease, such as tuberculosis, sarcoidosis, or fever of unknown origin [172]; and in patients with suspected primary or metastatic carcinoma. Contraindications to percutaneous liver biopsy include an uncooperative or unstable patient, ascites, right-sided empyema, and suspected hemangioma or echinococcal cyst. Transjugular liver biopsy is an alternative to the percutaneous approach in patients with severe coagulopathy and massive ascites. [173]

GENERAL APPROACH

Extensive numbers of tests have been described in the preceding pages and in other chapters. Initially, nonhepatic causes of any observed abnormalities must be considered (Table 45-1). The dilemma then faced is in selecting a proper diagnostic approach to the patient with suspected liver disease. Liver disease can be classified into four major types: the cholestatic, hepatocellular, and immunologic forms of injury and infiltrative processes. Depending on the target of the immune response, immunologic injury results in either a cholestatic picture (if the bile ducts are preferentially involved, as in PBC) or a hepatocellular form of injury (if the primary insult is to the hepatocyte membrane, as in viral and autoimmune hepatitis). Cholestasis can be further categorized as either a functional defect in bile formation at the level of the hepatocyte (intrahepatic cholestasis) or a structural impairment in bile secretion and flow (extrahepatic cholestasis). Evaluation is aided by the presence of these relatively discrete patterns of liver injury and tests of discriminative value in the detection of these patterns. Routinely, the results of the following tests should be determined in all patients with suspected liver disease before disease-specific markers are sought:

- Serum aminotransferase (AST and ALT) activity
- Serum alkaline phosphatase
- Serum total and direct bilirubin
- Serum total protein, with albumin and globulin fractionation
- Prothrombin time

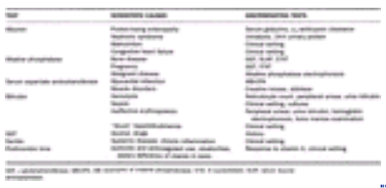

TABLE 45-1 Nonhepatic Causes of Abnormal Liver Chemistries

Patterns of abnormalities typical of the various forms of hepatobiliary injury emerge from this battery of tests, as outlined in Table 45-2. Additional diagnostic information may be provided by testing for disease-specific markers, as listed in Table 45-3. Further laboratory evaluation of any patient with evidence of chronic (>6 months) elevation in serum aminotransferases should, at the minimum, include the following tests:

- Serum protein electrophoresis
- Serum ferritin
- ANA and anti–smooth muscle antibodies
- Serum ceruloplasmin
- Hepatitis B viral serology
- Antibody to HCV

TABLE 45-2 Routine Biochemical Tests in the Patient with Idealized Hepatobiliary Disease

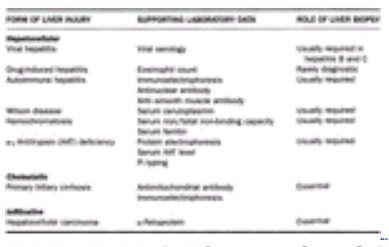

TABLE 45-3 Diagnosis of Selected Hepatobiliary Disorders

The cause of the chronic elevation in serum aminotransferases is suggested by these and other tests, as outlined in Figure 45-1. The management of minor elevations in serum ALT levels, for example, of less than 6 months' duration is less defined, although in asymptomatic persons this may represent clinically insignificant fluctuations and may resolve over time. [174]

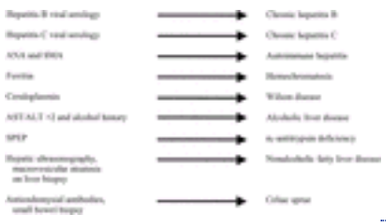

FIGURE 45-1. Evaluation of chronic serum aminotransferase elevations.

The differential diagnosis of a patient with abnormal liver chemistries consistent with cholestatic injury (i.e., elevations of serum alkaline phosphatase and bilirubin levels, with or without moderate elevations of ALT or AST) represents a formidable clinical challenge. Hyperbilirubinemia resulting from extrahepatic biliary obstruction, in contrast to that observed in acute and chronic forms of hepatocellular injury, tends to level off over time and rarely exceeds levels of 35 mg/dL in the absence of oliguria or hemolysis. The mechanism for this plateau effect appears to be related to altered bilirubin metabolism, including enhanced renal excretion of conjugated bilirubin. In addition, a daily increase of 1.5 mg/dL in total serum bilirubin is characteristic of extrahepatic biliary obstruction. [175] Nevertheless, none of the routine biochemical laboratory tests can reliably differentiate intrahepatic cholestasis from extrahepatic biliary obstruction. Furthermore, within 24 to 48 hours of acute extrahepatic obstruction, profound elevations in serum ALT and AST levels may be observed, followed by a rapid decline. [176], [177] An elevated serum bilirubin level in the absence of elevations of AST or alkaline phosphatase levels is an unusual finding and more commonly is a marker of underlying cardiac disease than any liver or biliary tract disease. [178]

Although there are no symptoms or signs that are pathognomonic for intrahepatic or extrahepatic forms of cholestasis, a history of previous biliary tract surgery, the presence of abdominal pain or significant weight loss, palpable gallbladder or abdominal mass, fever or other signs of cholangitis, and an elevated serum amylase level should point to an extrahepatic cause such as choledocholithiasis, pancreatitis, cholangiocarcinoma, or carcinoma of the pancreas. Partial obstruction or obstruction involving only a portion of the intrahepatic biliary tree may result in an elevated serum alkaline phosphatase level in the absence of hyperbilirubinemia. Conversely, a normal alkaline phosphatase level in a jaundiced patient strongly rules against the presence of extrahepatic biliary obstruction. [39] Fever and right upper quadrant abdominal pain may occur in drug-induced cholestasis and may lead to confusion if a detailed drug history is not available. A cholestatic biochemical profile with fluctuating levels of serum alkaline phosphatase in a patient with inflammatory bowel disease should suggest primary sclerosing cholangitis. Marked cholangiographic changes in the presence of advanced histological stage may, however, occur without a concomitant increase in serum alkaline phosphatase activity. [179] In the setting of acute pancreatitis, marked elevations in ALT and ?-glutamyltransferase levels point to a biliary origin. [180]

Additional laboratory tests, even liver biopsy, may not further differentiate between intrahepatic and extrahepatic cholestasis. Moreover, in drug-induced cholestasis, one of the more common causes of intrahepatic cholestasis, discontinuation of the offending drug may not be immediately followed by resolution of the cholestatic picture. As discussed in greater detail in other chapters, the most direct approach to the differential diagnosis of cholestatic injury is the use of abdominal ultrasonography to assess bile duct size, followed, if biliary dilatation is present, by endoscopic retrograde cholangiopancreatography (ERCP) or percutaneous transhepatic cholangiography (PTC). Biliary tract obstruction of short duration may not be accompanied by detectable dilatation of the bile ducts, and, when there is a strong clinical suspicion of an extrahepatic cause of cholestasis, ERCP or PTC may be indicated even in the presence of a normal ultrasonographic examination. [181]

Isolated elevation of serum alkaline phosphatase, confirmed by serum leucine aminopeptidase, 5'-nucleotidase, or ?-glutamyltransferase to be of hepatic origin, is strongly suggestive of an infiltrative process, whether a localized disorder (i.e., PBC) or a systemic granulomatous disease, such as sarcoidosis, miliary tuberculosis, coccidioidomycosis, histoplasmosis, brucellosis, Q fever, or a drug reaction (e.g., allopurinol, quinidine), or, alternatively, the first indication of metastatic carcinoma to the liver. A greater than threefold elevation in serum alkaline phosphatase levels in patients with cirrhosis should raise concern for the development of hepatocellular carcinoma. The triad of an elevated serum alkaline phosphatase level, detectable titers of AMA, and an elevated serum IgM level in a middle-aged woman is of considerable discriminative value in the diagnosis of PBC. However, patients with the clinical and pathological features of PBC but with negative AMA and positive

ANA tests are becoming increasingly recognized. In the largest series of patients with this disorder, best termed *autoimmune cholangiopathy,* [182] no significant differences were observed in gender or in the laboratory findings of AMA-positive and AMA-negative patients. In a patient with a history of malignant disease, particularly breast or colon cancer, the presence of an elevated serum alkaline phosphatase level warrants an evaluation for metastases. The absence, in this setting, of diagnostic findings on abdominal ultrasound, computed tomography of the abdomen, technetium sulfur colloid nuclear imaging, and invasive tests such as ERCP, is one of the major indications for percutaneous or laparoscopic liver biopsy. A decision tree that can be used in the approach to the patient with an elevated alkaline phosphatase level is provided in Figure 45-2.

FIGURE 45-2. Diagnosing the patient with elevated serum alkaline phosphatase (*AP*) levels. *CT*, computed tomography; *ERCP*, endoscopic retrograde cholangiopancreatography; *GGT*, serum ?-glutamyltransferase; *5'NT*, 5'nucleotidase.

Although other features, such as serum bilirubin levels and prolongation of the prothrombin time, may vary and correlate with the severity of the injury, elevated serum aminotransferases are characteristically associated with hepatocellular forms of injury and reflect release of intracellular enzymes from hepatocytes undergoing necrosis. As a general rule, aminotransferase levels greater than 400 U/L are indicative of hepatocellular injury. In contrast, milder degrees of serum aminotransferase elevation (<300 U/L) are of little diagnostic benefit because they are seen in cholestatic disorders as often as in acute and chronic hepatocellular disease. Establishing the cause of hepatocellular necrosis usually requires more information than routine laboratory results provide.

As discussed previously, the nature and degree of aminotransferase elevation may be helpful in differentiating alcoholic hepatitis from ischemic, viral-induced, or drug-induced hepatitis. Leukocytosis can be a prominent feature associated with alcoholic hepatitis. Ischemic liver injury is, at times, indistinguishable from acute viral hepatitis. A disproportionate and marked elevation in serum aminotransferases in the setting of generalized malaise, anorexia, jaundice, and tender hepatomegaly characterizes both disorders. However, measures to improve hepatic blood flow (e.g., correction of hypotension or congestive heart failure) are accompanied by a faster fall in serum aminotransferase levels than is observed in the course of acute viral hepatitis. [183] Normal values are occasionally found within 48 to 72 hours in cases of ischemic hepatitis. Associated mild to moderate renal impairment also suggests ischemic hepatitis. [183] Features suggestive of drug-related hepatotoxicity include a history of recent institution of therapy and indirect evidence of a hypersensitivity reaction, such as rash, arthralgia, or eosinophilia. Profoundly abnormal aminotransferase activities, with preservation of an elevated AST/ALT ratio, are observed in patients with alcoholism and acetaminophen hepatotoxicity, [184] and a positive response to questions regarding the use of acetaminophen mandates measurement of the blood level of this drug. The liver is commonly involved in patients with typhoid fever. Lower peak serum ALT and AST, higher peak serum alkaline phosphatase, an ALT/LDH ratio of less than 4, fever, and relative bradycardia may help to distinguish *Salmonella* hepatitis from acute viral hepatitis. [185] A high index of suspicion is required in the diagnosis of Wilson disease, and, although occasional patients may be diagnosed at older ages, [186] ceruloplasmin levels should be routinely determined in patients younger than 40 years of age with hepatic (variable elevations in serum aminotransferases in the presence of mild hyperbilirubinemia and hypoalbuminemia suggesting HBsAg-negative chronic active hepatitis, autoimmune hepatitis, or cryptogenic cirrhosis) and neurological abnormalities. Although a reduced serum ceruloplasmin level may be observed in other disorders, as has been discussed, its presence in an asymptomatic person with elevated serum aminotransferase levels is highly suggestive of Wilson disease. Hemolytic anemia, with or without low alkaline phosphatase activity, [97] and hypouricosemia are additional clues to the diagnosis of this disorder. However, establishing such a diagnosis in the setting of fulminant hepatic failure is difficult. The specificity of alkaline phosphatase to total bilirubin ratio of less than 2 in distinguishing fulminant hepatic failure secondary to Wilson disease from that resulting from other causes is open to question. [187]

Furthermore, the cause of elevated serum aminotransferases may vary considerably depending on the patient population examined, with ischemic hepatitis predominating in highly developed countries and viral and toxic hepatitis more likely in developing countries. [188] Chronic hepatitis of unknown origin may occur in a substantial proportion of asymptomatic patients with mild to moderate elevation of serum aminotransferases who live in areas with a low burden of hepatotropic viruses and genetic disorders. [189]

The extent of hepatic dysfunction in any form of injury is routinely assessed by the prothrombin time and serum albumin concentration. Prolongation of the prothrombin time without improvement after parenteral vitamin K administration and a serum albumin concentration lower than 3 g/dL both reflect the worse prognosis and greater severity of underlying liver disease.

Several studies performed before the identification of HCV addressed the issue of evaluating asymptomatic patients with moderate elevations in serum aminotransferase levels. [190], [191] Apart from determinations of ferritin, ceruloplasmin, a $_1$-antitrypsin, and markers for HBV, blood tests had little discriminative value. A high incidence of obesity and regular alcohol use was demonstrated in these patients. Fatty infiltration, consequently, was the most common finding on liver biopsy; however, histological findings of chronic persistent or active hepatitis in 20% of patients in one study [189] lent support for the use of percutaneous liver biopsy in the diagnostic approach to patients with persistently elevated levels of serum aminotransferases. Furthermore, examination of a select population of patients with unexplained chronic serum ALT elevations, in whom viral, alcohol-related, or drug-related disease was excluded, demonstrated histological features of chronic active hepatitis, including cirrhosis, in more than two thirds of patients. [192] Steatohepatitis was the most frequent alternative diagnosis, with clinical and laboratory findings, including seropositivity for ANAs, indistinguishable from those of patients with chronic active hepatitis. [192]

Previously considered to be a disorder predominantly seen in middle-aged, obese women with hyperglycemia or hyperlipidemia, nonalcoholic steatohepatitis (NASH), part of the spectrum of nonalcoholic fatty liver disease, is now recognized to exhibit a broader clinical profile. [193], [194] Diagnosis relies on the presence of macrovesicular steatosis and parenchymal inflammation with or without Mallory bodies, fibrosis, and cirrhosis in liver biopsy specimens in the absence of a history of excessive alcohol consumption, [193], [194] although the role of liver biopsy in this disorder is controversial. Abnormal serum aminotransferases are the most common biochemical abnormalities in this disorder, although there is a lack of association between biochemical abnormalities and the degree of steatosis. In general, serum aminotransferases are only mildly to moderately elevated; fewer than one third of patients have serum AST or ALT levels greater than threefold elevated. [193], [194] In the absence of cirrhosis, the AST/ALT ratio is invariably less than 1.0. [193] In contrast to patients with alcoholic liver disease, patients with NASH usually have an AST/ALT ratio less than 1, [195] and an AST/ALT ratio greater than 1 is a significant predictor of severe liver fibrosis or cirrhosis. [195], [196] Serum albumin levels are normal; hyperbilirubinemia is unusual, and, when present, levels are less than 2 mg/dL. [193], [194] Increased transferrin saturation and serum ferritin levels have been described but most likely result from underlying necroinflammatory activity. [193], [194] Although not widely available, a normal ratio of desialylated transferrin to total transferrin helps to distinguish patients with NASH from patients with excessive alcohol ingestion. [197] Weight reduction in obese patients may lead to improvement in serum aminotransferase abnormalities. [198]

With the development of serologic tests for HCV, up to 17% of volunteer blood donors with asymptomatic elevations in serum ALT were found to have evidence of HCV infection. [199] Therefore, determination of antibody to HCV is now routinely performed as part of the evaluation of patients with elevated serum ALT and AST levels. Asymptomatic patients who test positive for anti-HCV antibodies often demonstrate chronic liver disease on liver biopsy even in the absence of serum ALT elevations. However, liver biopsy need not be performed in antibody-positive patients with normal ALT levels whose serum HCV RNA evaluation is negative. [200]

The role of liver biopsy in asymptomatic patients with chronic liver chemistry abnormalities in whom specific liver disorders have been excluded by serologic evaluation is not well established. Although a liver biopsy may establish a diagnosis, frequently NASH, the results of the liver biopsy are infrequently different from the presumptive prebiopsy diagnosis, and no proven therapy is available for the majority of these patients. [201], [202] Therefore, the decision to perform a liver biopsy under these circumstances needs to be individualized, taking into account the risks and benefits of this procedure. Effective therapy may soon become available for NASH,

[203] but until that time, liver biopsy is unlikely to have a significant impact on patient management.

Patients who have undergone liver transplantation are frequently evaluated for abnormal liver chemistries. The differential diagnosis is quite extensive and includes acute cellular rejection, hepatic artery thrombosis, infectious complications such as cytomegalovirus and opportunistic infections, drug-induced hepatotoxicity such as cyclosporine-induced cholestasis, and biliary complications. Abnormal liver chemistries in the transplant recipient may also herald recurrence of the primary liver disease. For example, autoimmune hepatitis may recur in up to 33% of patients who undergo liver transplantation for chronic disease. [204] Liver biopsy and Doppler ultrasonography are frequently required because a specific pattern of biochemical changes corresponding to a particular diagnosis cannot be usually identified. [205]

In conclusion, the diagnostic tests discussed in this chapter suggest but rarely provide a specific diagnosis in a patient with suspected liver disease. Nevertheless, information obtained from these tests should facilitate the efficient and proper use of other noninvasive and invasive tests such as ultrasonography, computed tomography, radionuclide hepatobiliary scanning, PTC, ERCP, liver biopsy, and laparoscopy.

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CHAPTER 46

Bruce A. Runyon

APPROACH TO THE PATIENT WITH ASCITES

The word *ascites* is of Greek derivation *(askos)* and means *bag* or *sack.* The word is singular and is used to refer to the condition of pathological fluid accumulation within the abdominal cavity. The adjective *ascitic* is used to describe the fluid itself.

CAUSES OF ASCITES AND MECHANISMS OF ASCITES FORMATION

Table 46-1 lists the causes of ascites formation in a prospective series of 901 paracenteses performed on the general medical and gastroenterology/hepatology wards of two academic institutions. [1] Cirrhosis and alcoholic hepatitis cause most cases of ascites in these settings; only 15.9% of patients had a cause other than chronic parenchymal liver disease.

CAUSE	NUMBER	(% OF TOTAL)
Chronic parenchymal liver disease (cirrhosis and alcoholic hepatitis)	758	84.1
"Mixed" (portal hypertension plus another cause, e.g., cirrhosis and peritoneal carcinomatosis)	42	4.7
Heart failure	24	2.7
Malignancy without another cause	23	2.6
Tuberculosis without another cause	6	0.7
Fulminant hepatic failure	6	0.7
Pancreatic	4	0.4
Nephrogenous ("dialysis ascites")	2	0.2
Miscellaneous*	36	3.9

* Includes biliary ascites and chylous ascites resulting from lymphatic tears, lymphoma, and cirrhosis.
from ref. 1.

TABLE 46-1 Causes of Ascites in a Series of 901 Samples

Ascites Formation in Liver Disease

Portal hypertension appears to be a prerequisite for ascites formation in the setting of liver disease, even in fulminant hepatic failure. [2] Three theories of ascites formation have been developed. The *underfill theory* postulates that an alteration in oncotic-hydrostatic balance leads to loss of intravascular fluid into the peritoneal cavity with resulting intravascular underfilling. This activates plasma renin, aldosterone, and the sympathetic nervous system and results in renal sodium retention. The observation that patients with cirrhosis have intravascular *hyper* volemia rather than *hypo* volemia led to the *overflow theory.* In this hypothesis, primary renal sodium retention is proposed to lead to intravascular hypervolemia and resulting overflow of fluid into the peritoneal cavity. The most recent theory, the *peripheral arterial vasodilation theory,* includes components of both prior theories. [3] This newest theory proposes that portal hypertension leads to vasodilatation, which causes decreased effective arterial blood volume. These events are hypothesized to characterize early compensated cirrhosis, before ascites formation. As the natural history of the disease progresses, neurohumoral excitation increases, more renal sodium is retained, and plasma volume expands; this leads to overflow of fluid into the peritoneal cavity. The underfill theory is proposed to be operative early, and the overflow theory is proposed to be operative late in the natural history of cirrhosis, according to the vasodilation theory. [3]

In general, the kidneys retain sodium in response to hypovolemia to regain a euvolemic state. In patients with advanced cirrhosis, however, the kidneys retain sodium despite hypervolemia. The paradox of avid renal sodium retention in the presence of hypervolemia is explained, at least in part, by the lack of a good sensor for intravascular volume. In general, the kidneys rely on blood pressure to assess volume status indirectly; when doing so, hypotension equates with hypovolemia. Patients with advanced cirrhosis are hypotensive. [4] The kidneys retain sodium in response to this hypotensive, vasodilated state.

Since popularization of the vasodilation theory, there has been an international search for the mediator of the vascular dilation. Many substances have been investigated and ruled out, and nitric oxide appears to be the currently favored mediator. [5]

In summary, although the exact sequence of events that occur between development of portal hypertension and renal sodium retention is not entirely clear, it appears that portal hypertension leads to an increase in nitric oxide levels. Nitric oxide, in turn, mediates the splanchnic and peripheral vasodilation associated with portal hypertension. The neurohumoral excitation state (involving the renin-aldosterone system, sympathetic nervous system, vasopressin, and endothelin-1) of advanced cirrhosis is an attempt to maintain the perfusion pressure in the presence of the nitric oxide–mediated hypotension. [4] Fluid forms in the abdomen when the lymph filtering across the hypertensive sinusoid and then across the Glisson capsule exceeds the diaphragmatic lymphatics' ability to reabsorb the escaping fluid. [3]

Ascites Formation in Other Diseases

"Mixed" Ascites Approximately 5% of patients will have "mixed" ascites: that is, underlying portal hypertension as well as a second cause for ascites formation, such as cirrhosis plus peritoneal tuberculosis or cirrhosis plus peritoneal carcinomatosis (see Table 46-1). [1] A clue to the presence of a second cause of ascites formation in a patient with obvious cirrhosis is an inappropriately high ascitic fluid lymphocyte count. The assumption that a patient with cirrhosis can have only one cause for ascites formation could lead to a missed diagnosis of a curable but potentially fatal disease such as tuberculous peritonitis.
Malignant Disease Fortunately, cancer is an uncommon cause of ascites formation, but unfortunately most patients with malignancy-related ascites survive only a few weeks after the onset of fluid retention. [6] An exception to this very short life expectancy is the patient with ovarian carcinoma, who may respond to debulking surgery and chemotherapy. [7] Not all malignancy-related ascites results from peritoneal carcinomatosis; the characteristics of the ascitic fluid and the treatments vary

depending on pathophysiology of ascites formation [8] (Table 46-2).

SUBTYPE	PREVALENCE (%)
Peritoneal carcinomatosis alone	53.3
Massive liver metastases alone	13.3
Peritoneal carcinomatosis and massive liver metastases	13.3
Hepatocellular carcinoma with portal hypertension	13.3
Malignant chylous ascites	6.7

Adapted from ref. 6.

TABLE 46-2 Subtypes of Malignancy-Related Ascites and Their Prevalence

The mechanism of ascites formation in patients with malignant ascites depends on the location of the tumor. Peritoneal carcinomatosis appears to cause ascites formation by “exudation” of proteinaceous fluid from tumor cells lining the peritoneum and entry of extracellular fluid into the peritoneal cavity for reestablishment of oncotic balance. [8] In patients with massive liver metastases, liver replacement by tumor or occlusion of portal veins with tumor emboli leads to portal hypertension; then ascites forms as it does in patients with parenchymal liver disease and portal hypertension. In the United States, most patients with hepatocellular carcinoma have underlying cirrhosis and portal hypertension; some of these patients do not develop fluid retention until the tumor becomes relatively large and replaces a significant percentage of the liver parenchyma. [6] Alternatively, tumor-induced portal vein thrombosis or arteriovenous fistulae within the tumor may contribute to the patient’s portal hypertension and predispose to fluid retention. Chylous ascites resulting from malignant disease appears to be caused by lymph node involvement by tumor and rupture of chyle-containing lymphatics. [6] Tumor can occlude the hepatic veins and can lead to ascites formation.

Tuberculous Peritonitis In the United States, tuberculous peritonitis was a rare disease in the past but has become more common as the human immunodeficiency virus epidemic has evolved. [9] More than one half of patients with tuberculous peritonitis in the United States are found to have underlying cirrhosis, usually alcoholic in origin. [10] As in peritoneal carcinomatosis, tuberculous peritonitis probably causes ascites formation because of “exudation” of proteinaceous fluid by tubercles lining the peritoneum and entry of extracellular fluid into the peritoneal cavity for reestablishment of oncotic balance. At peritoneoscopy, the diffuse studding of the peritoneum by these lesions substantiates the plausibility of this mode of pathogenesis. Presumably, *Coccidioides* (a very cause form of inflammatory ascites) results in ascites formation by the same mechanism as tuberculosis. [11]

Heart Failure Ascites is currently an uncommon complication of heart disease. [12] , [13] It forms in high-output and low-output heart failure. [14] In the former situation, decreased peripheral resistance appears to initiate salt and water retention, whereas in the latter condition, a diminished cardiac output is the first event. [14] Both these initial events lead to a decreased effective arterial blood volume and subsequent activation of the vasopressin, renin-aldosterone, and sympathetic nervous systems. In turn, renal vasoconstriction and sodium and water retention occur. [14] Fluid then weeps from the congested hepatic sinusoids as lymph, as in the formation of cirrhotic ascites.

Pancreatic Ascites This rare form of ascites develops as part of severe acute (even hemorrhagic) pancreatitis or as a result of pancreatic duct rupture or leakage from a pseudocyst as a complication of chronic pancreatitis. [15] Patients with this form of ascites may also have underlying cirrhosis. Pancreatic ascites may occasionally be complicated by bacterial infection. Pleural effusions (left-sided, usually) may be associated. Ascites forms in this situation either by leakage of pancreatic juice into the peritoneal cavity or by a “chemical burn” of the peritoneum. Extracellular fluid then enters to reestablish oncotic equilibrium. The ascitic fluid retains the unique characteristics of the source fluid (e.g., high amylase concentration), modified to some degree by the added extracellular fluid. [15]

Fulminant Hepatic Failure Ascites may develop as a manifestation of acute liver failure in viral hepatitis. Because fulminant liver failure is uncommon, however, the total number of patients with ascites who have acute liver failure is small (see Table 46-1). The ascites that forms in this setting has a high (=1.1 g/dL) serum-ascites albumin concentration gradient indicating portal hypertension. [1] Ascites thus presumably forms in fulminant hepatic failure by mechanisms similar to those of the fluid that forms in parenchymal liver disease. [1]

Biliary Ascites Bile can accumulate in the peritoneal cavity when the gallbladder, bile duct, or gut ruptures, or it can develop after biliary surgery. This is an uncommon form of ascites. Biliary ascites is most commonly the result of rupture of the gallbladder, usually a complication of gangrene of the gallbladder in elderly men. [16]

Lymphatic Tear After extensive retroperitoneal dissection, as in distal splenorenal shunt or radical pelvic lymphadenectomy for testicular carcinoma, lymphatics may be transected and may leak lymph for variable periods. [17] The formation of ascites in this condition is similar to that of malignant chylous ascites, that is, lymphatic leak. The presence or absence of chyle in the ascitic fluid depends on where the tear is in the lymphatic system—in chyle-containing channels or not.

Miscellaneous Causes of Ascites In sexually active, otherwise healthy young women with fever and inflammatory ascites, chlamydial infection must be very high in the differential diagnosis. Chlamydia apparently now causes more cases of Fitz-Hugh-Curtis syndrome than does the gonococcus. [18] Chlamydia peritonitis is one of the few curable causes of ascites formation. Nephrogenous ascites develops in patients who are undergoing hemodialysis. [19] On careful evaluation, many of these patients are found to have underlying chronic liver disease, which may be the reason they develop fluid overload more readily than patients without liver disease who are undergoing dialysis. Evaluation of patients with nephrogenous ascites may include peritoneoscopy, which will assist with the differential diagnosis and will confirm or rule out tuberculosis and cirrhosis. [19] The proper treatment of this condition is uncertain, and the prognosis is poor. [19] Nephrotic syndrome is always listed as a cause of ascites as if it were common, but in fact it is rare in adults. [20] Usually, a second cause of ascites formation is also present. [20] It is postulated that, in nephrotic syndrome in the absence of another cause of fluid retention, loss of protein (in particular albumin) in the urine leads to decreased effective arterial blood volume, activated vasopressin, renin-aldosterone, and sympathetic nervous systems with resulting renal sodium and water retention. [14] Continuous ambulatory peritoneal dialysis causes an iatrogenic form of ascites that is usually under the management of nephrologists. The major problem is infection, which occurs approximately once per patient-year of treatment. Urine may accumulate in the peritoneal cavity as a result of trauma, as a complication of renal transplantation, or in the newborn—a condition known as “urine ascites.” [21] Reabsorption of urea can lead to “pseudorenal failure.” [21] Serositis with ascites formation may complicate systemic lupus erythematosus. [22] This form of ascites has been reported to respond to steroid therapy. [22] Pathogenesis presumably involves inflammation of the peritoneum, with resultant exudation of proteinaceous fluid into the cavity. In recent years, most cases of ascites caused by ovarian disease are the result of peritoneal carcinomatosis. [7] Meig syndrome—ascites and pleural effusion caused by benign ovarian neoplasms—is no longer a common cause of ascites formation (see Table 46-1). Ascites in patients with myxedema appears to be cardiac ascites, related to the subtle heart failure these patients develop. [23] As in other forms of cardiac ascites, this ascitic fluid is high in protein and has a high albumin gradient. [23] , [24] Treatment of the thyroid insufficiency cures the fluid retention. Patients with either acute or chronic Budd-Chiari syndrome frequently develop ascites from portal hypertension and venous outflow obstruction. The protein concentration of this fluid is variable, but the albumin gradient is high.

EVALUATION OF THE PATIENT WITH ASCITES

History

Most ascites results from liver disease, and much of the liver disease in the United States is caused by alcohol abuse, chronic hepatitis C, and obesity. [25] Many patients have more than one insult speeding liver injury. Ascites frequently develops as a part of the patient’s first decompensation of alcoholic liver disease, either alcoholic hepatitis or cirrhosis. Patients with alcoholic liver disease who intermittently cease or reduce alcohol consumption may experience ascites in a cyclic fashion. These cycles may be separated by many years and tend to parallel alcohol consumption. [26] In contrast, patients who develop ascites with nonalcoholic liver disease tend to be persistently troubled by ascites after its onset, probably because of the late stage at which ascites forms and the lack of effective therapy of nonalcoholic liver disease short of liver transplantation. When the patient has a very long history of stable cirrhosis and then develops ascites, the possibility of superimposed hepatocellular carcinoma should be considered.

Patients with ascites should be questioned about risk factors for liver disease, including alcohol use, intravenous drug use, transfusions, acupuncture, tattoos or other body piercing, and origination from an endemic area for hepatitis. The exposure may be so remote in time that the patient may not remember it initially or may not believe that a long-forgotten needle exposure is the cause of the liver disease decades later.

Obesity has become a recognized cause of a progressive form of liver disease—nonalcoholic steatohepatitis—that can progress to cirrhosis. [27] A decade of obesity can be an additive risk factor for cirrhosis in patients with alcoholism. [28]

Patients who have a history of cancer and who develop ascites should be suspected of having malignant ascites. Similarly, patients who have no risk factors for liver disease and who lose a significant amount of weight during ascites formation may harbor a neoplasm. Patients with cardiac ascites often have a past history of heart disease. [13] Some patients with alcoholism who develop ascites have alcoholic cardiomyopathy, rather then liver disease. Tuberculous peritonitis is usually manifested by fever and some abdominal discomfort. More than one half of patients with tuberculous peritonitis in the United States have underlying cirrhosis. [10] Patients who

develop ascites in the setting of known diabetes or nephrotic syndrome should be suspected of having nephrotic ascites; these patients have anasarca. Ascites developing in a patient with lethargy, cold intolerance, or change in the skin and voice should be considered as possibly resulting from myxedema. Connective tissue diseases may be complicated by ascites as a manifestation of serositis. [22]

Physical Examination

Increasing abdominal girth may be caused by ascites, massive hepatomegaly related to alcoholic hepatitis without fluid retention, gaseous distention of the bowel, obesity, or massive ovarian tumor without free fluid. Percussion of the flanks will rapidly determine which patients have ascites. If an experienced examiner detects no flank dullness, there is little or no ascites present (90% accuracy). [29], [30] If flank dullness is detected, the patient should be rolled into a partial decubitus position to determine whether the air-fluid interface shifts. Flank dullness was found to be a more helpful test than the puddle sign or the fluid wave in two prospective studies. [29], [30]

Physical findings that are very suggestive of the presence of significant liver disease include blotchy palmar erythema localized predominantly to the hypothenar eminence and large, three-dimensional pulsatile vascular spiders. These findings may be difficult to detect in patients with very dark pigmentation. Similarly, the presence of pathologically large abdominal wall collateral veins suggests that portal hypertension is present. Nearly 100% of patients with cirrhosis and ascites and relatively light pigmentation have at least one of the foregoing findings (Runyon BA, personal observation).

The presence of large veins on the flanks and back of the patient suggests inferior vena cava block, as in a web lesion or malignant obstruction. A firm nodule in the umbilicus, the *Sister Mary Joseph nodule,* is not common but is very suggestive of peritoneal carcinomatosis originating from a gastric, pancreatic, or hepatic primary tumor. A pathological left-sided supraclavicular node (the *Virchow node*) suggests the presence of cancer in the upper abdomen. The neck veins of patients with ascites should always be examined for distention in pursuit of a cardiac origin of ascites. Surprisingly, not all patients with cardiac ascites have peripheral edema, and most do not have rales. When patients with cirrhosis have peripheral edema, it is usually in the lower extremities only. Conversely, patients with nephrosis and those with cardiac failure may have anasarca.

Ascites may be semi-quantitated using the following system: 1+ is detectable only by careful examination, 2+ is easily detected but of relatively small volume, 3+ is large ascites but not tense, and 4+ is tense ascites.

Abdominal Paracentesis

Indications Abdominal paracentesis is the fastest and perhaps the most cost-effective method of diagnosing the cause of ascites formation. [31] In the past, *diagnostic paracentesis* was underused in the assessment of patients with ascites, in part because of concern regarding complications of the procedure itself. However, more recent studies have documented the safety of this procedure. [32], [33] Moreover, in view of the prevalence of ascitic fluid infection at the time patients with ascites are admitted to the hospital, *an admission surveillance paracentesis* is important for detection of infection. [34] Therefore, it is appropriate to sample ascitic fluid in all patients with *new*-onset ascites (as inpatients or outpatients) and in all patients with ascites admitted to the hospital. Paracentesis should be repeated (for outpatients or inpatients) if signs or symptoms of infection such as abdominal pain or tenderness, fever, encephalopathy, renal failure, acidosis, or peripheral leukocytosis develop. [31], [34]

Technique My technique of paracentesis has changed in recent years, in part because of the increased use of therapeutic paracentesis. [35] In addition, the percentage of patients with ascites who are obese has increased. Although the midline is avascular (unless there is an unusual collateral vein there), the abdominal wall is thinner in the lower quadrants compared with the midline. More fluid can be obtained from a supine patient with a lower quadrant paracentesis than from the midline. These factors have led me to perform more lower quadrant paracenteses in recent years and fewer midline procedures. Needles inserted too close to abdominal wall scars may enter the bowel, which may be fixed to the wall. [32] If there is a midline scar, the needle should be placed several centimeters from the scar, thus favoring a lower quadrant approach. If a lower quadrant is chosen, it is necessary to avoid placing the needle too near the liver or spleen and to avoid entering a subcutaneous collateral vein or the inferior hypogastric artery. A site located on either flank two finger-breadths cephalad and two finger-breadths medial to the anterior superior iliac spine is usually safe and successful. Because many of these patients are taking lactulose, the bowel may be distended with gas. The cecum distends more than other segments of the bowel. Therefore, the left side is usually tapped to avoid a distended cecum. There must be dullness in the site selected for needle entry. This usually requires that the patient be placed in the supine or lateral decubitus position. I prefer to use standard metal (no trocar, no plastic) 1.5-inch needles—22-gauge needles for diagnostic paracenteses and 15-gauge needles for therapeutic paracenteses (see section “ Therapeutic Paracentesis”). “Spinal needles,” that is, 3.5-inch needles, are needed only 6% of the time, only in the setting of a large panniculus. [32] Multiple-hole blunt (~15-gauge) needles with sharp removable trocars are now available and have been found to be safe. [36] Large-bore trocars are avoided because they leave large puncture wounds that leak ascitic fluid or cause other complications when they are placed inadvertently into vessels or the bowel. After selecting the insertion site and needle, the operator disinfects the skin with an iodine solution. The skin and subcutaneous tissue should be infiltrated with a local anesthetic. Sterile gloves should be used when the operator is actually obtaining the fluid. The sterile paper packaging of the gloves can be used as a sterile field on which to place syringes, needles, and so forth. The manner in which the needle is inserted is crucial for a safe successful paracentesis. Use of a “Z-tract” minimizes leakage of fluid after the procedure, but there is confusion about how to accomplish a Z-tract. It does *not* involve “zig-zagging” the needle in the subcutaneous tissue; this may cause laceration of vessels and hemorrhage. To create a Z-tract, the operator uses one gloved hand to move the skin (~2 cm in any direction) in relation to the deep abdominal wall while inserting the paracentesis needle, which is attached to the syringe. The other gloved hand is used to hold the syringe and pull on the plunger simultaneously; the inexperienced operator finds it difficult to manipulate the syringe with only one hand. The skin is not released until the needle has penetrated the peritoneum and fluid flows. When the needle is eventually removed, the skin slips back into its original position and seals the leak. If the needle is inserted directly into the fluid without a Z-tract, the fluid will leak out easily because of a straight line of flow. During insertion, the needle should be advanced in approximately 5-mm increments. A slow incremental insertion allows the operator to see a “flash” of blood if he or she enters a vessel; then the needle can be withdrawn before there is further damage. A slow insertion also allows the bowel to move away from the needle before the needle enters the bowel. The syringe that is attached to the needle *should not* be aspirated continuously during the entire insertion. If this is done, bowel or omentum may be suctioned to the end of the needle as soon as the needle enters the peritoneal cavity, thus giving the appearance of a “dry tap.” Therefore, the needle should be inserted approximately 5 mm, then the syringe aspirated for a few seconds while the needle is stationary, then advanced, then aspirated, and so on, until the sensation of entering the peritoneum is felt and fluid is aspirated. A slow insertion also allows time for the elastic peritoneum to “tent” over and be pierced by the needle. The quantity of fluid removed depends on the goal of the paracentesis. The requirement for optimal ascitic fluid culture plus cell count and “chemistries” (see section “ Ascitic Fluid Analysis”) is at least 25 mL.

Contraindications and Risks There are very few real contraindications to paracentesis. Because most patients with cirrhotic ascites have coagulopathy, [32] coagulopathy is not a contraindication unless there is primary fibrinolysis or clinical disseminated intravascular coagulation with clinically evident bleeding; these conditions occur less than once per 1000 paracenteses, in my experience. There is no cutoff of coagulation parameters beyond which paracentesis should not be performed. Patients with cirrhosis but without fibrinolysis or disseminated intravascular coagulation do not bleed seriously from needle sticks unless a blood vessel is entered. [32], [33] Some physicians give prophylactic fresh frozen plasma or platelets routinely before paracentesis in patients with cirrhosis and coagulopathy. This practice is not supported by data. [32], [33] A prospective study regarding paracentesis complications reports very low morbidity. [32] There were no deaths or infections related to paracentesis in this study. Complications included abdominal wall hematomas both large (in 2 of 229, 0.9%) and small (in 2 of 229, 0.9%). Seventy-one percent of the patients who underwent paracentesis had an abnormal prothrombin time, which was prolonged (=5 seconds) in 21%. [32]

Ascitic Fluid Analysis

Gross appearance. Most ascitic fluid is transparent and yellow tinged. Very dilute fluid in anicteric patients with cirrhosis may resemble water. Patients with deep jaundice have bile-stained ascitic fluid, but their fluid is less bile stained than their serum. [16] The opacity of most cloudy ascitic fluid specimens results from the presence of many neutrophils. This fluid has a shimmering effect when it is held in a glass tube in front of a bright light. Neutrophil counts lower than 1000/mm [3] may not be very cloudy, but counts greater than 50,000/mm [3] have a purulent consistency. About 20% of cirrhotic ascitic fluid is not transparent, but it is not frankly milky either. In this condition, called *opalescent ascites,* the cloudiness of the fluid is the result of triglyceride. [37] The lipid will usually stratify in the refrigerator over a 48- to 72-hour interval. A minimum of 10,000 red cells/mm [3] is required for ascitic fluid to appear pink; smaller concentrations result in a clear or “smoky” appearance. Ascitic fluid with a red cell count greater than 20,000/mm [3] is distinctly blood tinged. Most bloody ascitic fluid occurs in the setting of a “traumatic tap” in a patient with cirrhosis; this fluid is heterogenously bloody, and the fluid clots. In contrast, nontraumatic (or remotely traumatic) bloody ascitic fluid is homogenously red and does not clot because it has already clotted and lysed. Ascites in the setting of tuberculous peritonitis is seldom bloody. Overall, only 22% of malignancy-related samples are bloody. [6] Half of samples from patients with hepatocellular carcinoma are bloody, but only about 10% of samples from patients with peritoneal carcinomatosis have this appearance. [6] Pancreatic ascites may appear tea colored or even black, presumably because of the denaturing effect of the pancreatic enzymes on ascitic fluid red cells.

Which ascitic fluid tests are worth ordering? Screening tests are performed on the initial specimen as well as other tests that are appropriate, based on the probability of the presence of a cause for ascites formation other than cirrhosis. Subsequent tests may be performed (usually necessitating another paracentesis)

based on the results of the initial analysis and incoming clinical information. However, because most specimens will represent uncomplicated cirrhotic ascites, no further tap will usually be needed. Based on cost-effectiveness analysis, I have developed a list of “routine, optional, and unusual” tests (Table 46-3). The strategy of test ordering is discussed in the following sections.

ROUTINE	OPTIONAL	UNUSUAL
Cell count	Gram stain	Tuberculosis smear and culture
Albumin	Glucose	
Culture in blood culture bottles	Lactate dehydrogenase	Cytology
	Amylase	Triglyceride
Total protein		Bilirubin

TABLE 46-3 Laboratory Data to be Obtained on Ascitic Fluid

Cell count. The cell count is the single most helpful test to order. If only one drop of fluid is obtainable, it should be sent for cell count. One needs only 10 µL for a standard manual hemocytometer count. The fluid should be submitted in an EDTA tube to prevent aggregation of the cells; clumped cells make an accurate count nearly impossible. An ascitic fluid culture is difficult to interpret without a cell count. The upper limit of “normal” total white blood cell count in uncomplicated cirrhotic ascites is 500 cells/mm [3]. [38] However, during a 10-kg diuresis in a patient with cirrhosis, the mean ascitic fluid white cell count triples from approximately 300 to approximately 1200 cells/mm [3]. [39] The upper limit cutoff of absolute polymorphonuclear leukocyte (PMN) count is usually stated to be 250 PMN/mm [3]. [38] Fortunately, the PMN count remains relatively constant during diuresis, presumably because of the short half-life of PMNs. [39] Therefore, the 250 cutoff pertains even at the end of diuresis. Most cases of bloody ascites result from traumatic paracentesis in a patient with cirrhosis. If a paracentesis is traumatic, only 1 PMN per 250 red cells can be attributed to blood contamination of ascites and only 1 lymphocyte per 750 red cells. [39] Any inflammatory cause of ascites and any inflammatory complication of ascites can elevate the ascitic fluid white cell count. The most common cause of an elevated count is spontaneous bacterial peritonitis (SBP), in which the total white cell count is elevated as well as the absolute PMN count. The percentage of PMNs is almost always greater than 50% and is usually greater than 70%. In tuberculous peritonitis and peritoneal carcinomatosis, there is usually a predominance of lymphocytes. The white cell count in chylous ascites may be increased because of leakage of lymphocytes with the chyle. The red cell counts in cardiac ascites may be increased because of leakage of red cells across the congested liver, whereas red cells may be increased in chylous ascites as a result of leakage from lymph channels.

Albumin. The serum-ascites albumin gradient categorizes ascites better than the total protein concentration. [1], [40], [41] To calculate this gradient, the albumin concentrations of serum and ascitic fluid specimens are measured, and the ascitic fluid value is simply subtracted from the serum value. The specimens should be obtained relatively simultaneously—within a few minutes or hours. In addition, if the patient’s condition is so unstable that portal pressure is altered, as in shock, the albumin gradient will be less reliable. Accuracy of the albumin assay at low albumin concentrations is important. Usually, the laboratory’s albumin standards must be diluted to assess and ensure linearity of the assay at low range. The albumin gradient’s utility is based on the concept of oncotic-hydrostatic balance. [41] If a patient has a high portal pressure, there must be a high oncotic gradient to match. Of the serum proteins, albumin has the most effect on oncotic pressure of serum. There are large differences between serum and ascitic fluid albumin concentrations in patients with portal hypertension (Table 46-4 and Fig. 46-1). If serum albumin minus ascitic fluid albumin is greater than or equal to 1.1 g/dL (11g/L), the patient has portal hypertension with 97% accuracy [1] (see Fig. 46-1). The higher the gradient, the greater the portal pressure. [41] Conversely, if serum albumin minus ascitic fluid albumin is less than 1.1 g/dL, the patient does not have portal hypertension (see Fig. 46-1).

HIGH GRADIENT (i.e., ≥1.1 g/dL)	LOW GRADIENT (i.e., <1.1 g/dL)
Cirrhosis	Peritoneal carcinomatosis
Alcoholic hepatitis	Tuberculosis (without cirrhosis)
Cardiac ascites	Pancreatic ascites (without cirrhosis)
Massive liver metastases	Biliary ascites (without cirrhosis)
Fulminant hepatic failure	Nephrotic syndrome
Budd-Chiari syndrome	Ascites in patients with connective tissue disease
Portal vein thrombosis	
Veno-occlusive disease	Ascites from bowel obstruction or infarction
Acute fatty liver of pregnancy	
Myxedema	
“Mixed” ascites	

TABLE 46-4 Classification of Ascites by Serum-Ascites Albumin Concentration Gradient

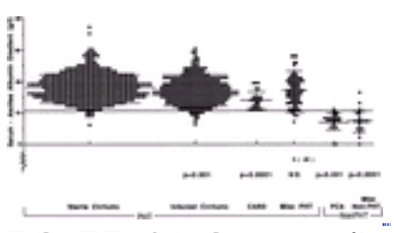

FIGURE 46-1. Serum-ascites albumin gradient, classified by the presence or absence of portal hypertension. Statistical comparisons are to the sterile cirrhotic group by unpaired *t*-test. *NS,* not significant; *CARD,* cardiac ascites; *Misc PHT,* miscellaneous portal hypertension–related; *PCA,* peritoneal carcinomatosis; *Misc Non-PHT,* miscellaneous non–portal hypertension related. Mean ± standard deviation bars are included as well as a horizontal line at 11 g/L (1.1 g/dL), the threshold for portal hypertension. (From ref. [1].)

High-albumin gradient and *low-albumin gradient* should replace the terms *transudative* and *exudative* in the description of ascites (see section “ Total Protein”). [1], [40], [41] The albumin gradient is approximately 97% accurate in categorizing ascites patients into those with and those without portal hypertension (see Fig. 46-1). [1] However, if the albumin assay is not accurate at low range, errors will occur. If a patient with cirrhosis has a serum albumin less than 1.1 g/dL (this occurs very rarely), the gradient will be falsely low. If the patient is in shock, the gradient will be low because the portal pressure is low. In most laboratories, lipid interferes with the albumin assay; therefore, patients with chylous ascites may have a falsely high albumin gradient. Patients with “mixed” ascites—that is, portal hypertension plus another cause of ascites formation—will have a high-albumin gradient, as expected. [1] The presence of tuberculosis or peritoneal carcinomatosis would not be expected to lower the portal pressure. The albumin gradient is simply a reflection of the portal pressure. Cirrhosis is the most common cause of a high-albumin gradient, and the most common cause of a low-albumin gradient is peritoneal carcinomatosis; however, there are other causes of high and low gradients (see Table 46-4). The albumin gradient need only be performed on the first paracentesis in a given patient; it need not be repeated on subsequent analyses.

Culture. The sensitivity of bacterial culture in detecting spontaneous infection in neutrocytic ascites varies dramatically depending on the method of culture used; “conventional” cultures detect approximately 40% of cases, whereas bedside inoculation of blood culture bottles with ascites results approximately 90% detection of bacterial growth in neutrocytic ascites. [42] Without fail, all prospective studies have demonstrated the superiority of the blood culture bottle method over the conventional method. [42], [43] The problem with the conventional culture is that it is designed to detect bacterial growth in polymicrobial infections that involve very high concentrations of bacteria. SBP is essentially always a monomicrobial infection with a median bacterial count of only one organism per milliliter. [42] Blood culture bottles were designed to detect monomicrobial infections involving small quantities of bacteria and are ideal for culturing ascites in suspected SBP. [42] However, not all blood culture bottles are alike. Some permit inoculation of only a few milliliters of fluid; others actually suppress growth of some bacteria. Using bottles that permit inoculation of 10 to 20 mL of ascites appears to be optimal. [42] Bedside inoculation of the blood culture bottles is preferable to delayed inoculation of the bottles in the microbiology laboratory. [43]

Total protein. Ascitic fluid was classified in the past as an exudate when the protein level was greater than or equal to 2.5 g/dL; occasionally, a cutoff of 3.0 g/dL was used. However, the exudate/transudate system of ascitic fluid classification is fraught with problems. The ascitic fluid protein level in cirrhotic ascites is entirely dependent on serum total protein concentration and portal pressure. [41] Therefore, a patient with cirrhosis with a relatively high serum protein level will have a relatively high ascitic fluid protein level without cancer or tuberculosis. Nineteen percent of uncomplicated cirrhotic ascites samples have greater than 2.5 g/dL of protein. [44] During a 10-kg diuresis, ascitic fluid total protein doubles from 1.4 to 2.9 g/dL; 67% of patients with cirrhotic ascites develop a protein level greater then 2.5 g/dL at the end of diuresis. [39] Contrary to popular belief, ascitic fluid total protein concentration does not increase during SBP, and patients with the lowest protein levels in ascites are the most prone to develop SBP. [45], [46] Almost one third of patients with malignant ascites have portal hypertension resulting from massive liver metastases or hepatocellular carcinoma as the cause of ascites formation; their fluid total protein is less than 2.5 g/dL. [6] In cardiac ascites, the protein concentration is essentially always greater than 2.5 g/dL. [13] The accuracy of an ascitic fluid total protein level greater than or equal to 2.5 g/dL in detecting an exudative cause of ascites formation was determined to be only 55.6% in a study of 901 samples (Table 46-5 and Fig. 46-2). [1] A cutoff of 3.0 g/dL yielded essentially an identical

accuracy—56.0% (Runyon BA, Montano AA, Akriviadis EA, et al., personal observations). Modified pleural fluid exudate criteria were also assessed for accuracy in detecting an exudative cause for ascites formation in this study. Exudate criteria included an ascitic fluid lactate dehydrogenase (LDH) level greater than 2.33 μkat/L (>140 U/L), ascitic fluid/serum (AF/S) total protein ratio greater than 0.5, and a ratio of AF/S to LDH greater than 0.6. Specimens with zero or one criterion were classified as transudates, and specimens with two or three criteria were classified as exudates. The accuracy of these criteria in detecting an exudative cause for ascites formation was determined to be only 57% (see Table 46-5; Fig. 46-3) (Runyon BA, Montano AA, Akriviadis EA, et al., personal observations).

PARAMETER	ACCURACY (%)
Serum-ascites albumin gradient in detecting portal hypertension	96.7
Ascitic fluid total protein concentration >2.5 g/dL in detecting exudate	55.6
LDH, AF/S LDH, and AF/S total protein criteria in detecting an exudate	57.0

AF/S, ratio of ascitic fluid to serum; LDH, lactate dehydrogenase.

TABLE 46-5 Accuracy of Ascitic Fluid Parameters

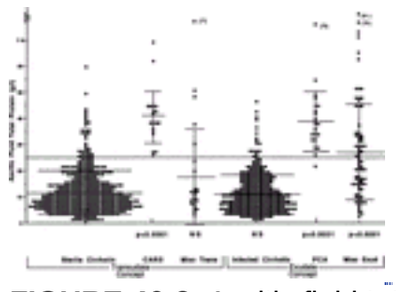

FIGURE 46-2. Ascitic fluid total protein in ascites of various types, classified by transudate versus exudate. Statistical comparisons are with the sterile cirrhotic group by unpaired *t*-test. *NS,* not significant; *CARD,* cardiac ascites; *Misc Trans;* miscellaneous transudative; *PCA,* peritoneal carcinomatosis; *Misc Exud,* miscellaneous exudative. The miscellaneous transudative category included fulminant hepatic failure, chylous cirrhotic ascites, and nephrotic and nephrogenous ascites. The miscellaneous exudative category included patients with mixed ascites (if one of the causes was exudative), infected fulminant hepatic failure samples, massive liver metastases, hepatocellular carcinoma, infected cardiac ascites, chylous lymphoma-related ascites, tuberculous peritonitis, pancreatitis, chlamydia peritonitis, and secondary bacterial peritonitis in the absence of cirrhosis. Mean ± standard deviation bars are included as well as a *horizontal line* at 25 g/L (2.5 g/dL), the threshold for exudate. (From ref. [1].)

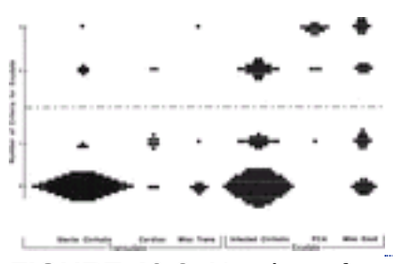

FIGURE 46-3. Number of exudate criteria in ascites of various types, classified by transudate versus exudate. *Misc Trans,* miscellaneous transudative; *PCA,* peritoneal carcinomatosis; *Misc Exud,* miscellaneous exudative. The *horizontal line* separates specimens satisfying zero or one exudate criteria from those satisfying two or three exudate criteria.

In summary, whether defined by total protein alone or by modified pleural fluid exudate criteria, the concept of transudate versus exudate is not very useful in ascitic fluid analysis. The serum-ascites albumin gradient is superior. Despite its shortcomings, the ascitic fluid total protein level is of some value. Oral quinolones have been shown to help prevent SBP in inpatients with ascites with low protein concentrations. [47] In addition, measurement of total protein, glucose, and LDH in ascites has been reported to be of value in distinguishing SBP from gut perforation into ascites [48], [49] (Fig. 46-4). Patients with ascitic fluid that is neutrocytic and meets two out of the following three criteria are unlikely to have SBP and warrant immediate evaluation to determine whether gut perforation into ascites has occurred: total protein greater than 1g/dL, glucose less than 50 mg/dL, and LDH greater than the upper limit of normal for serum. [48], [49]

FIGURE 46-4. Algorithm for differentiating spontaneous from secondary bacterial peritonitis. (From ref. [49].)

Glucose. Because of its small molecule weight, glucose enters ascites readily. Therefore, the ascitic fluid glucose concentration is similar to that of serum unless glucose is being consumed in the peritoneal cavity by white blood cells or bacteria. If the gut perforates (e.g., perforated ulcer or diverticulum) into ascitic fluid, the glucose concentration usually is low and can be as low as 0 mg/dL. [48], [49]

Lactate dehydrogenase. LDH is a much larger molecule than glucose and enters ascitic fluid less readily than does glucose. The ratio of AF/S to LDH is approximately 0.40 in uncomplicated cirrhotic ascites; in SBP, presumably because of neutrophil release of LDH, the ascitic fluid LDH level rises such that the mean ratio is 0.9±0.3. [48] If the LDH ratio is more than 1.0, LDH is being produced in or released into the peritoneal cavity; usually because of infection or tumor.

Amylase. The mean ascitic fluid amylase concentration is 42 ± 44 IU/L in uncomplicated cirrhotic ascites, and the AF/S ratio of amylase is approximately 0.4. [15] The ascitic fluid amylase concentration rises above this mean in the setting of pancreatitis or gut perforation. In pancreatic ascites, the ascitic fluid amylase concentration averages approximately 2,000 IU/L, and the AF/S ratio in pancreatic ascites averages approximately 6.0. [15]

Gram stain. Although a Gram stain of ascites is frequently ordered when SBP is suspected, careful inspection of the centrifuged sediment of 50 mL of ascites is only 10% sensitive in visualizing bacteria in early-detected SBP; a Gram stain of uncentrifuged fluid is positive in only 7%. [42] Approximately 10,000 bacteria/mL are required for detection by Gram stain; the median concentration of bacteria in SBP is only one organism/mL. [42] A Gram stain of ascites is analogous to a Gram stain of blood in bacteremia; it is positive only when there is overwhelming infection. The Gram stain is most helpful in ruling in or ruling out free perforation of the gut into ascites, in which sheets of multiple bacterial forms are found. A syringe of fluid must be submitted to the microbiology laboratory when requesting a Gram stain, in addition to cultures in blood culture bottles.

Smear and culture for tuberculosis. The direct smear of ascitic fluid has only 0% to 2% sensitivity in detecting *Mycobacteria.* [10], [50] I have seen only one positive ascitic fluid tuberculous smear, and it was falsely positive. Culture of ascitic fluid is only about 40% sensitive. [10], [50] Peritoneal biopsy is 64% to 83% sensitive. [10], [50] The sensitivity of peritoneoscopy in detecting tuberculous peritonitis approaches 100%. [10], [50] Tuberculous peritonitis may mimic the culture-negative variant of SBP, but mononuclear cells predominate in tuberculous peritonitis, rather than PMNs in the setting of SBP. The adenosine deaminase activity of ascitic fluid has been proposed as a useful nonculture method of detecting tuberculous peritonitis; however, patients with cirrhosis and tuberculous peritonitis usually have falsely low values. [10]

Cytology. Cytology smears are reported to be 58% to 75% sensitive in detecting “malignant ascites.” [51] However, only about two thirds of patients with malignancy-related ascites have peritoneal carcinomatosis; essentially 100% of patients with peritoneal carcinomatosis will have viable malignant cells exfoliating into their ascitic fluid and will have positive ascitic fluid cytologic studies. [6] The remaining one third of patients with malignancy-related ascites have massive liver

metastases, chylous ascites from lymphoma, or hepatocellular carcinoma; these patients have negative cytology. [6] The percentage of false-positive cytologic examinations approaches zero. [51] Hepatomas metastasize to the peritoneum infrequently enough to be the subject of case reports. [52] Flow cytometry occasional detects evidence of malignant disease when cytology is negative. [53] Some cytology laboratories prefer that specimens be submitted in fixative; others prefer fresh, unfixed specimens. It is best to determine the specifics of submission with each local laboratory to maximize the sensitivity of the cytology. In addition, submitting specimens in very small quantity (only a few milliliters) or submitting an unfixed specimen late on a Friday night may handicap the pathologist's ability to assist in the diagnosis of malignancy. I submit at least 50 mL of fluid for cytologic examination on the rare occasion that I request this expensive test. Malignancy-related ascites may have an elevated PMN count (16% of cases in one series), presumably because dying tumor cells may attract neutrophils into the fluid. [6] Usually in this setting, there is no predominance of PMNs; this helps in differentiating this condition from SBP.

Peritoneal biopsy. The ascitic fluid cytology is so sensitive in detecting peritoneal carcinomatosis that peritoneal biopsy is not needed in the search for tumor. The best indication for peritoneal biopsy is in the setting of suspected tuberculous peritonitis. However, one half of patients with tuberculous peritonitis in the United States have underlying cirrhosis with portal hypertension and intra-abdominal collateral vessels. Blind Cope needle or Abrams needle biopsy of the peritoneum in the setting of portal hypertension risks fatal laceration of a collateral vessel. Direct visualization of the peritoneum by peritoneoscopy is preferable to blind biopsy in the cirrhotic patient with suspected tuberculous peritonitis. Culture of the biopsy for mycobacteria is the most sensitive and specific test for tuberculous peritonitis. [10]

Triglyceride. If the ascitic fluid is milky, a triglyceride level should be obtained. Chylous ascites has a triglyceride content greater than 200 mg/dL and usually greater than 1000 mg/dL. [54] About 20% of sterile cirrhotic ascitic fluid specimens are slightly cloudy, but the cloudiness is not the result of an elevated cell count. [37] The triglyceride concentration of these specimens is approximately 60 mg/dL, compared with approximately 20 mg/dL for clear cirrhotic ascites; these specimens are called opalescent. [37]

Bilirubin. Brown ascites should be tested for bilirubin concentration. An ascitic fluid bilirubin level greater than the serum level suggests bowel or biliary perforation into ascites. [16], [49]

pH. Ascitic fluid pH was proposed in the past as a helpful test in differentiating infected or malignant ascites from uncomplicated cirrhotic ascites. Unfortunately, the studies that purported to validate the pH were small (only 6 to 18 infected patients per study) and did not use optimal culture technique. In two more recent, larger studies, the pH was not found to be helpful. [55], [56] Ascitic fluid pH appears to be simply an indirect measurement of the presence of PMNs. It also requires special processing; that is, it must be sent immediately to the laboratory on ice in a plastic syringe without anticoagulant. It is simpler and more cost effective to measure the PMN concentration directly. [55]

Lactate. The ascitic fluid lactate concentration has also been proposed as a marker of infection or tumor. However, lactic acid metabolism in ascites is linked to pH changes by the same pathophysiology. Lactate has the same problems as pH as an ascitic fluid test. It is not a helpful or cost-effective test for the same reasons that pH determination is not a helpful or cost-effective test. [55]

Carcinoembryonic antigen. Measurement of carcinoembryonic antigen (CEA) in ascitic fluid has been proposed as a helpful test in detecting malignant ascites. [57] However, the study that validated CEA did not have a subgroup of patients with malignancy-related ascites and did not study a large number of patients. No subsequent studies have been reported. In the study of CEA in ascites, all benign fluids had CEA levels less than 10 ng/mL, whereas CEA was greater than 10 ng/mL in 14 of 29 (48%) patients with malignant ascites. However, cytology was positive in only 12 of 29 (41%). Such insensitivity of cytology makes interpretation of the study difficult. CEA may be of some utility in ascitic fluid analysis, but its precise value remains unclear.

Ascitic fluid "humoral tests of malignancy". Several tests, such as fibronectin, cholesterol, a $_1$-antitrypsin, cyclic AMP, and glycosaminoglycans determinations, were proposed as useful in detecting malignant ascites. The basic premise of these studies was that the ascitic fluid cytology is too insensitive to be the only test used in detecting malignancy-related ascites. Unfortunately, these studies did not have proper controls and did not recognize that there are several subgroups of malignancy-related ascites, nor did they appreciate that the cytology would be helpful only in detecting peritoneal carcinomatosis. Therefore, patients with negative cytologies also have negative "humoral tests of malignancy." [58] In addition, patients with high-protein, noncirrhotic ascites essentially always have false-positive "humoral tests of malignancy." [6], [58] Therefore, these tests are more confusing than helpful.

Serum Analysis Measurement of serum albumin concentration is mandatory to determine the albumin gradient. If ascitic fluid infection is suspected, blood should be cultured in addition to ascitic fluid even if the patient is not febrile (only approximately two thirds of patients with SBP have fever). Calculations of protein and LDH ratios in the differential diagnosis of ascites have not been as helpful as hoped. Application of pleural fluid exudate criteria to ascitic fluid simply is not useful (see Fig. 46-3). Patients with systemic bacterial infection and cirrhosis do not convert glycogen to glucose normally and may develop profound peripheral hypoglycemia (as low as 5 mg/dL); if the ascitic fluid glucose is especially low in a patient with neutrocytic ascites, measurement of a "stat" serum glucose may explain the ascitic fluid hypoglycemia and may result in emergency administration of concentrated glucose intravenously. Measurement of serum bilirubin or triglyceride concentration may be of value in interpreting the ascitic fluid counterparts of these tests. Testing the serum for a-fetoprotein concentration may be of value in detecting hepatoma in the patient with compensated cirrhosis who suddenly develops ascites; the ascitic fluid a-fetoprotein is always lower than the serum value in a given patient. [6]

Conclusion The history and physical examination will usually provide a working diagnosis of the cause of ascites formation. Ascitic fluid analysis combined with testing of blood will usually provide a definitive diagnosis (see Table 46-3). Although the ascitic fluid cell count is the single most helpful ascitic fluid test in determining which patients need empiric antibiotic treatment and in distinguishing inflammatory from noninflammatory conditions, the albumin gradient is the test used to classify ascites into high- and low-albumin gradient categories (see Table 46-4 and Table 46-5 and Fig. 46-1, Fig. 46-2 and Fig. 46-3). Abdominal paracentesis assists in the diagnosis and treatment of patients with ascites. Performance of this procedure with appropriate analysis of the fluid is perhaps the safest and most cost-effective way to diagnose the cause of ascites formation and to rule in or rule out ascitic fluid infection.

Radiologic Assessment

Many physicians order radiologic tests in addition to appropriate testing of the fluid in the initial evaluation of the patient with ascites. Abdominal ultrasound may be of value in differentiating obesity from ascites or in detecting small-volume ascites. In the patient with ascites and multiple abdominal scars, ultrasound may be of value in determining where to insert the paracentesis needle safely. [35] Ultrasound may detect ovarian or pancreatic or mesenteric masses in the patient whose ascitic fluid analysis suggests peritoneal carcinomatosis.

Ultrasound may also be of value in detecting hepatocellular carcinoma, and ultrasound with Doppler can be diagnostic of hepatic vein or portal vein thrombosis. Worldwide, ultrasound is the most commonly used imaging modality in patients with ascites. In many parts of the world, hepatologists perform the examinations themselves.

Computed tomographic studies may complement ultrasound and may be of additional value in detecting the increased density of the liver compared with the spleen in hemochromatosis.

In the patient with infected ascites, if the initial ascitic fluid analysis meets criteria for gut perforation (see Fig. 46-4), [49] and especially if large numbers of different bacteria are noted on Gram stain, emergency plain and upright abdominal films should be obtained to determine whether free air is present. If there is free air, the patient probably needs laparotomy. If no free air is present, emergency water-soluble gut contrast studies should be performed. If the patient is young, a water-soluble upper gastrointestinal contrast study should be performed first, to look for extravasation from a perforated duodenal ulcer. If the patient is elderly, a water-soluble contrast enema should be performed first, to look for extravasation from a perforated colonic diverticulum. If extravasation is found, emergency surgical intervention should take place. The plain abdominal film may also be of value on rare occasions in detecting calcification of metastatic colonic carcinoma or ovarian neoplasms.

COMPLICATIONS OF ASCITES

Infection

Spontaneous Bacterial Peritonitis Ascitic fluid infection can be classified into five categories (Table 46-6). [34] An abdominal paracentesis must be performed before a diagnosis of ascitic fluid infection can be made. Simply culturing blood and making a clinical diagnosis of peritonitis are not adequate. The prototypic form of ascitic fluid infection is SBP: positive ascitic fluid culture (essentially always pure growth of a single organism) and an elevated ascitic fluid absolute PMN count greater than or equal to 250 cells/mm [3], without an evident intra-abdominal source of infection that requires surgical treatment.

CATEGORY	ASCITIC FLUID ANALYSIS
Spontaneous bacterial peritonitis	PMNs ≥250/mm³, single organism
Culture-negative neutrocytic ascites	PMNs ≥250/mm³, negative culture
Secondary bacterial peritonitis	PMNs ≥250/mm³, usually multiple organisms
Monomicrobial bacterascites	PMNs <250/mm³, single organism
Polymicrobial bacterascites	PMNs <250/mm³, multiple organisms

PMNs, polymorphonuclear leukocytes.

TABLE 46-6 Classification of Infected Ascites

Setting. For all practical purposes, SBP occurs only in the setting of liver disease, usually severe liver disease. [34], [59] Ninety-five percent of patients with SBP have an elevated serum bilirubin, 81% are clinically jaundiced, and 98% have an abnormal prothrombin time. [60] The liver disease may be chronic, as in alcoholic cirrhosis, subacute, as in alcoholic hepatitis, or acute, as in fulminant hepatic failure. SBP is most common in alcoholic cirrhosis (with or without chronic hepatitis C), the most common cause of ascites formation in the United States. All forms of cirrhosis, however, have been reported to be complicated by SBP. SBP is commonly encountered in patients with known hepatitis B–related liver disease and superimposed acute hepatitis. [34] Ascites is a prerequisite to development of SBP, but the fluid may not always be clinically detectable. It is unlikely that SBP precedes ascites formation. Usually, this infection develops in patients at the time of their largest ascites volume. Nephrotic ascites regularly was complicated by SBP in the preantibiotic era, but the current use of diuretics and antibiotics has made SBP uncommon in this setting. [20] Cardiac ascites rarely is complicated by SBP, and patients with peritoneal carcinomatosis hardly ever develop spontaneous peritonitis. [59], [61] About half of SBP episodes are detected at the time of admission to the hospital; the remainder develop after admission. [60]

Pathogenesis. The pathogenesis of SBP has become clearer. Most of the organisms that cause SBP are of enteric origin. [42] It had been assumed in the past that virulent gut flora simply migrated transmurally to infect ascitic fluid. However, transmural migration of bacteria has been documented only in the setting of loss of mucosal integrity. [62] If organisms could easily traverse the gut wall, polymicrobial infections would be the rule, rather the exception. In addition, the flora of the gut differs from the flora of SBP—anaerobes and enterococci predominate in the former, and *Escherichia coli, Klebsiella pneumoniae,* and pneumococci are the most common isolates in the latter. [34] "Translocation" of bacteria from the lumen of the gut to mesenteric lymph nodes is more likely to be one of the first steps in the pathogenesis of SBP, rather than transmural migration of bacteria from the gut directly into ascites. The normal gut mucosa functions as a selective filter; gram-negative aerobic bacteria translocate more readily than gram-positives and anaerobes. [63] Cirrhotic rats have been shown to translocate bacteria from the gut to mesenteric lymph nodes much more commonly than normal rats translocate; the gut mucosa in the setting of cirrhosis is "leakier" than normal. [64] Overgrowth of an organism in the gut is one of the most predictable precursors of translocation. [63] Intestinal bacterial overgrowth has been shown to precede translocation in cirrhotic rats. [65] Bacteremia may be an additional factor in the pathogenesis of SBP. Cirrhotic rats contain a localized infectious process, such as pneumonia, less well than do normal rats. They also develop bacteremia after intratracheal exposure to pneumococci and have more prolonged bacteremia and more fatalities when ascites is present compared with cirrhotic rats without ascites and compared with normal rats. [66] Therefore, it is not surprising that bacteremia is common in patients with severe liver disease. [67] Fifty percent of patients with SBP have bacteremia at the time of diagnosis of peritoneal infection. [60] Patients with cirrhosis with ascites usually have serum complement deficiency. [68] Because complement is required for the opsonization (coating for phagocyte-mediated killing) of most virulent organisms, complement deficiency would be expected to predispose to bacterial infections, including bacteremia. [68] Furthermore, neutrophil and reticuloendothelial system dysfunctions are also common in cirrhosis. [69], [70] Such abnormalities in host defense against infection would be expected to lead to frequent and prolonged bacteremia. Thus, in the setting of cirrhosis, mesenteric lymph colonization or portal bacteremia would be expected to lead to peripheral bacteremia. In turn, this systemic bacteremia could lead to bacterascites as bacteria-laden lymph forms from blood and leaks across the Glisson capsule (Fig. 46-5). Because the ascitic fluid of patients with cirrhosis is a significant dilution of their complement-deficient serum, and because the opsonic activity (endogenous antimicrobial activity) of human ascitic fluid correlates directly with the fluid's protein concentration, many of these patients have no ability to control or eradicate colonization of their ascitic fluid. [68] The protein concentration of ascitic fluid does not change with development of spontaneous infection. [45] Patients with low-protein ascites, such as less than 1 g/dL, have been shown to be particularly prone to SBP. [46] Patients with deficient ascitic fluid opsonic activity have been shown to be predisposed to SBP. [71]

FIGURE 46-5. Proposed pathogenesis of spontaneous bacterial peritonitis.

In summary, the gut mucosa of cirrhotic animals appears to be abnormally permeable to bacteria. Intestinal bacterial overgrowth of a specific organism leads to translocation of that organism. Bacteria readily enter mesenteric lymph or portal blood and could then access the peripheral circulation. Lymph that forms from bacteria-laden blood in the liver then enters ascitic fluid. Ascitic fluid that is deficient in antimicrobial properties is the fertile soil in which SBP develops.

Signs and symptoms. Eighty-seven percent of patients with SBP reported in one study had signs or symptoms of infection. [72] Prevalence of the most common signs and symptoms from a review of 489 patients was as follows: fever, 69%; abdominal pain, 59%; and mental status change, 54%. [73] However, the clinical evidence of infection may be very subtle, as in mild hepatic encephalopathy that is apparent only as a change in personality noted by the patient's family.

Prevalence. When abdominal paracentesis is performed routinely in patients with ascites at the time of admission to the hospital (whether or not they have symptoms of infection), up to 27% are found to have SBP or a variant thereof. [34], [74] Approximately 15% of hospitalized cirrhotic patients with ascites are found to have SBP, as specifically defined, on hospital admission. [34] However, the use of selective intestinal decontamination (see section " Prevention") has reduced the prevalence of ascitic fluid infection. [75] To maximize survival, it is important to perform paracentesis on hospital admission so infection can be diagnosed and treated early. In addition, paracentesis should be repeated during hospitalization if any deterioration occurs, including pain, fever, mental status change, renal failure, acidosis, peripheral leukocytosis, or gastrointestinal bleeding. There are no data on the frequency of SBP in outpatients. Fatal infection may be a common cause of death of cirrhotic patients who die at home.

Flora. Approximately 80% of SBP is caused by *E coli,* streptococci (mostly pneumococci), and *Klebsiella.* [42], [73] Anaerobes cause approximately 1% of SBP. [42] Many reported cases of anaerobic SBP are polymicrobial and probably represent misdiagnosed secondary peritonitis. [49] Presumably, the infrequency of anaerobic SBP results from the relatively high PO_2 of ascites, approaching that of arterial blood. The prevalence of pneumococcal SBP has increased as ascitic fluid culture methods have improved. [42]

Risk factors. Patients with cirrhosis are unusually prone to bacterial infections because of multiple defects in immune defense (see section " Pathogenesis"). [34] In a prospective study performed in Brazil, 47% of 170 cirrhotic patients during a single hospitalization had bacterial infection of various types ranging from asymptomatic urinary tract infections to fatal sepsis. [76] Bacteriuria is twice as common in cirrhotic patients compared with matched controls and is more common in patients with SBP compared with uninfected patients with ascites. [77] Perhaps it is this high frequency of colonization of cirrhotic patients with bacteria and the inability of these immunocompromised patients to localize infection that lead to bacteremia and colonization of ascites. Paracentesis itself appears to cause ascitic fluid infection only when the paracentesis needle enters the bowel inadvertently during attempted paracentesis. Fortunately, this is very unusual (see the later discussion of polymicrobial bacterascites); only 10 cases were found among 1578 paracenteses in a retrospective study. [78] Gastrointestinal hemorrhage has been linked to development of spontaneous bacteremia and SBP. [79] The mechanism of this predisposition is not entirely clear, but it probably involves an ischemia-reperfusion–related increase in translocation. In addition, invasive procedures performed in relation to gastrointestinal bleeding, such as endoscopy or intravascular catheter placement, may also predispose to bacteremia. Even simple, seemingly innocuous procedures such as bladder catheter insertion in the setting of a bladder infection may cause bacteremia. Certainly, leaving a catheter in place for several days guarantees cystitis; urosepsis frequently follows in cirrhotic patients because of their inability to localize infections.

Diagnosis. By definition, a positive ascitic fluid culture and an ascitic fluid PMN count greater than or equal to 250 cells/mm [3] are required before a diagnosis of SBP is made. A clinical diagnosis of ascitic fluid infection is not adequate; paracentesis must be performed. A high index of suspicion of SBP and a low threshold for performing a paracentesis are required for early diagnosis of this infection.

Survival. The infection-related mortality has decreased dramatically, from approximately 100% in the 1960s to 5% in a 1991 study. [80] This improved survival is probably a reflection of earlier detection of infection as well as the avoidance of nephrotoxic antibiotics in patients with cirrhosis. Many patients are cured of their infection and yet die of liver or renal failure or gastrointestinal bleeding.

Treatment. Patients with ascitic fluid PMN counts greater than or equal to 250 cells/mm [3] in a clinical setting compatible with SBP should be treated empirically with

antibiotics. Patients with hemorrhage into ascites, peritoneal carcinomatosis, pancreatic ascites, or tuberculous peritonitis may have an elevated PMN count that is not related to SBP. They do not require empiric treatment. If the situation is initially unclear, however, treatment should be given until the diagnosis is clarified. Usually, patients with uninfected neutrocytic ascitic fluid (except those with hemorrhage) have a predominance of lymphocytes in their ascitic fluid differential; this helps to distinguish them from patients with SBP, in whom PMNs predominate. Patients with bloody ascites should have a "corrected" PMN count calculated—one PMN is subtracted from the total absolute PMN count for every 250 red blood cells. [39] Such corrections of the PMN count may result in a negative number, that is, less than zero. Patients with bloody ascites do not need antibiotics unless their corrected PMN count is greater than or equal to 250 cells/mm [3]. Patients with suspected SBP require broad-spectrum therapy until the results of susceptibility testing are available. The recommendation for choice of empiric treatment has changed over the years. In the past, aminoglycosides were used. Now we know that gentamicin has an unpredictable volume of distribution in patients with ascites and that serum creatinine (and even the creatinine clearance) is a poor index of the glomerular filtration rate in patients with ascites. [81] Therefore, it is very difficult to give appropriate loading or maintenance doses of gentamicin in these patients. Even if overdosing is avoided, up to 73% of cirrhotic patients develop nephrotoxicity. [82] Gentamicin and gram-negative infections appear to be synergistic in leading to kidney damage. [83] Because of the difficulties in dosing and the apparent nephrotoxicity of aminoglycosides, the use of these drugs was simultaneously abandoned in the treatment of cirrhotic patients in the early 1980s in many liver units around the world. Cefotaxime, a third-generation cephalosporin, was shown to be superior to ampicillin plus tobramycin in a controlled trial. [84] This drug covered 98% of the flora (including pneumococci), was more efficacious, and did not result in superinfection or nephrotoxicity. [84] This drug, or a similar third-generation cephalosporin, probably is the treatment of choice of suspected SBP. For cefotaxime, 2 g intravenously every 8 hours is very effective. [80], [85] A randomized controlled trial demonstrated a survival advantage if albumin is given intravenously (1.5 g/kg body weight at the time of detection of infection and 1 g/kg on day 3 of treatment) in addition to cefotaxime compared with cefotaxime alone. [86] The albumin appears to help prevent the renal failure that can develop in the cirrhotic patient with bacterial infection. After susceptibility testing results are available, a narrower-spectrum drug can usually be substituted. Pneumococci are sensitive to penicillin, and most *E coli* species are sensitive to ampicillin. Ceftriaxone has not been compared with cefotaxime in a randomized trial, but it appears to be effective if given in high enough dose, such as 2 g intravenously every 12 hours. [87] Amoxicillin-clavulanic acid is effective in treated SBP but is unavailable for parenteral use in the United States. [88] Oral antibiotic (ofloxacin, 400 mg twice daily for a mean duration of 8 days) treatment of SBP has been studied and found to be as effective as intravenous cefotaxime. [89] However, only "good risk" patients were entered into the randomized trial. Thirty-nine percent of patients with SBP who were screened for entry were excluded because of ileus, gut bleeding, shock, azotemia, or encephalopathy. This study was performed on inpatients. There is no information on outpatient treatment of SBP. A 2-day course of intravenous ciprofloxacin followed by 5 days of oral ciprofloxacin was found to be as effective as 7 days of intravenous ciprofloxacin in a randomized trial. [90] Optimal duration of therapy is becoming clearer. The ascitic fluid culture becomes negative after one dose of cefotaxime in 86% of patients. [49] The ascitic fluid PMN count, in general, drops exponentially after treatment is started such that the PMN count after 48 hours of therapy is always less than the pretreatment value in SBP treated with appropriate antibiotics. [49] A randomized controlled trial of 5 days versus 10 days of treatment documented that short-course treatment is as effective as long-course therapy. [80] When the setting, organism, and clinical response to treatment are typical, follow-up paracentesis is not necessary, and 5 days of treatment are usually enough. [91] Patients have even undergone liver transplantation after only 4 days of treatment with good results. [92]

Recurrence. All series of patients with SBP report recurrences. A prospective study, reported in 1988 from Barcelona, documented a 69% recurrence rate at 1 year. [93] An ascitic fluid protein concentration less than 1.0 g/dL was the best predictor of recurrence, just as it is a good predictor of a first episode. [46], [93] In view of this impressive recurrence rate, patients who survive SBP and who are otherwise candidates for liver transplantation should be considered high priority for liver transplantation before they have a fatal recurrence of SBP.

Prevention. The impressive recurrence rate of SBP has led to multiple trials of antibiotic prophylaxis. Oral quinolone prophylaxis (norfloxacin 400 mg/d) has been documented to prevent gram-negative bacterial infections in patients who have survived one SBP episode, in patients with low-protein ascites, and in patients with fulminant hepatic failure. [75] Norfloxacin (400 mg/d orally twice daily for 7 days) has been documented to prevent gram-negative bacterial infections in inpatients with variceal bleeding. [75] Trimethoprim-sulfamethoxazole orally 5 days per week and ciprofloxacin (750 mg) once per week have also been shown to prevent bacterial infections in high-risk patients with cirrhosis. [94] One double strength tablet of trimethoprim-sulfamethoxazole daily also appears to be effective. In an animal model, trimethoprim-sulfamethoxazole improved survival in the setting of cirrhosis with ascites. [95] Use of norfloxacin to prevent a recurrence of SBP has been calculated to be cost effective. [96] Unfortunately, use of antibiotics leads to selection of resistant flora and can lead to infection by resistant flora. This has been documented in patients and rats with cirrhosis. [97], [98] Although oral norfloxacin reduced ascitic fluid infection by approximately 60% in cirrhotic rats, treated rats developed enterococcal overgrowth in the gut, and, when they developed infection, enterococcus was cultured from their ascitic fluid and mesenteric lymph nodes. [98] In addition, norfloxacin did not improve survival in the animal model. [98] Many liver units are giving their transplant candidates continuous quinolone prophylaxis. There are no data to support this practice, unless the patient has had at least one SBP episode. [93] A retrospective study has shown that an important risk factor for posttransplant fungal infection is total dose of quinolone. [99] More data are needed, preferably a controlled trial, before we know whether to use quinolones liberally in patients awaiting liver transplantation. While we wait for the results of more studies, it is appropriate to use norfloxacin or trimethoprim-sulfamethoxazole once daily for prevention of recurrence of SBP and in inpatients with an ascitic fluid protein concentration lower than 1 g/dL and to use one of these agents twice daily for 7 days for patients with cirrhosis (with or without ascites) and significant gastrointestinal bleeding. [75] Some physicians give prophylactic antibiotics to cirrhotic patients at the time of variceal sclerotherapy. However, a randomized trial has shown that this is not necessary. [100] With use of short sclerotherapy needles and clean water for the endoscope, the risk of bacteremia is so low that prophylactic antibiotics are not warranted. In addition, variceal banding is even less likely to lead to bacteremia, and banding should replace sclerotherapy in view of its superior efficacy.

Spontaneous Bacterial Peritonitis Variants SBP variants include monomicrobial nonneutrocytic bacterascites (MNB), culture-negative neutrocytic ascites, secondary bacterial peritonitis, and polymicrobial bacterascites (see Table 46-6). [49], [72], [78] /SUP>, [101] The adjective *monomicrobial* is used to distinguish this variant of ascitic fluid infection from polymicrobial bacterascites (see later). In the older literature, MNB was either grouped with SBP or was called asymptomatic bacterascites. However, many patients with bacterascites have symptoms; therefore, the modifier *asymptomatic* does not seem appropriate. The flora of MNB is similar to that of SBP. [42], [72] Because of the mortality (22%–43%) associated with monomicrobial bacterascites and the finding that MNB can progress to SBP in a matter of minutes, treatment appears to be warranted in some patients. [72], [102] Because the PMN count is not elevated (by definition) in MNB, it is reasonable to use empiric antibiotics, such as cefotaxime 2 g intravenously every 8 hours, for patients with cirrhotic ascites who have *convincing signs or symptoms of infection* regardless of the PMN count in ascitic fluid. Empiric treatment can be discontinued after only 2 to 3 days if the culture remains negative. For patients who have asymptomatic monomicrobial bacterascites, paracentesis should be repeated for cell count and culture. If the PMN count has risen to 250/mm [3] or more, treatment should be started. Patients who have developed no PMN response and no clinical evidence of infection do not require treatment. These are the patients who have probably eradicated the colonization by their own immune defenses. [73] The diagnosis of culture-negative neutrocytic ascites (CNNA) is made when

1. The ascitic fluid culture grows no bacteria.
2. The ascitic fluid PMN count is greater than or equal to 250 cells/mm [3].
3. No antibiotics have been given (even a single dose usually makes the culture negative).
4. There is no other explanation for an elevated PMN count, such as hemorrhage into ascites, peritoneal carcinomatosis, tuberculosis, or pancreatitis (see Table 46-6). [101]

Previously, the PMN criterion for CNNA was the presence of more than 500 cells/mm [3]; however, a value of greater than or equal to 250 cells/mm [3] was supported by a prospective study and is now viewed as the most appropriate criterion. [55] Inadequate culture technique explains the majority of episodes of CNNA. [42] In hospitals that use the older "conventional" culture method, negative cultures of infected ascites are common. Because improper culture techniques result in negative cultures, one of the most important methods to reduce the prevalence of CNNA in a hospital that is still using the conventional method of culture is to convince the microbiology laboratory to convert to the optimal method of culture—bedside inoculation of blood culture bottles with ascitic fluid. [42], [43] Patients who have CNNA despite good culture technique, in general, have spontaneously resolving SBP. This does not occur very often, but it is a real phenomenon. [73] Initially, one does not know that the culture is destined to be "no growth" in a patient with this variant of ascitic fluid infection. Therefore, empiric treatment should be given. When the preliminary report demonstrates no growth, it is helpful to repeat the paracentesis to assess the response of the PMN count to therapy. A decline in PMN count confirms a response to treatment and probably warrants a few more days of therapy. A stable PMN count, especially if there is not a predominance of PMNs, indicates that a nonbacterial (or mycobacterial) cause of the neutrocytosis is present. Biopsy and culture for tuberculosis may be appropriate. Secondary bacterial peritonitis is surgical peritonitis in patients with ascites. [49] Secondary peritonitis is treated with antibiotics *and* surgery, whereas SBP is always treated *only* with antibiotics. [34], [49] Performing a laparotomy in SBP or treating secondary peritonitis only with antibiotics usually results in the death of the patient. [49] Secondary bacterial infection of ascitic fluid can occur in any patient with ascites, even patients with noncirrhotic ascites. [49] Host defenses have little bearing on outcome when the gut perforates into ascitic fluid; the patient will die without surgical intervention. The possibility of secondary peritonitis should be *considered* in any ascites patient with peritonitis (see Fig. 46-4). However, because of the rarity of SBP in peritoneal carcinomatosis or cardiac ascites, a surgical source of peritonitis should be *presumed* in patients with these diagnoses until proved otherwise. [59], [61] Surprisingly, even with free perforation of the colon into ascitic fluid, patients do not develop a classical surgical abdomen. [49] Peritoneal signs require contact of inflamed visceral and parietal peritoneal surfaces. This does not happen when there is a large volume of fluid present. Therefore, clinical signs and symptoms do not separate patients with secondary peritonitis from those with SBP. Gut perforation can be suspected and pursued if ascitic fluid analysis

meets two of the following three criteria (see Fig. 46-4): total protein greater than 1 g/dL, glucose less than 50 mg/dL, and LDH greater than 225 mU/mL (or more than the upper limit of normal for serum). [49] Multiple organisms are cultured from ascitic fluids in the setting of a perforated viscus except for gallbladder rupture. [16], [49] If multiple organisms and PMNs are seen on Gram stain, the likelihood of perforation is very high. Brown ascitic fluid with a bilirubin concentration greater than 6 mg/dL and greater than the serum level is indicative of biliary or gut (especially upper gut) perforation into ascites. [16] The initial ascitic fluid analysis is very helpful in delineating which patients are likely to have ruptured gut. As noted earlier (see section "Radiologic Assessment"), these patients need an emergency radiologic evaluation, that is, within minutes, to confirm and localize the gastroduodenal or colonic site of rupture. If perforation is documented, emergency intervention is mandatory to maximize survival; survivors have been reported. [48], [49] Patients with nonperforation secondary peritonitis tend not to have a diagnostic initial ascitic fluid analysis. [48], [49] Fortunately, it is less urgent to make the diagnosis of secondary peritonitis in nonperforation peritonitis compared with perforation peritonitis. Therefore, there may be time to evaluate the response of the ascitic fluid culture and PMN count to treatment. These parameters have been shown to be helpful in distinguishing secondary from spontaneous peritonitis. [48], [49] The best time to perform a single repeat paracentesis to assess response is after 48 hours of treatment. [49] At 48 hours, essentially every patient with SBP who has been treated with an appropriate antibiotic will have a PMN count lower than the pretreatment value and the culture will be negative; in contrast, in secondary peritonitis, the culture remains positive, and the PMN count rises. [49] Patients suspected of having secondary peritonitis require broader-spectrum empiric antibiotic coverage than those with SBP, in addition to an emergency evaluation to assess the need for surgical intervention (see earlier). Cefotaxime and metronidazole together provide excellent initial empiric therapy of suspected secondary peritonitis while the radiologic workup is under way. [49] Polymicrobial bacterascites should be suspected when

1. The paracentesis is difficult because of ileus or a traumatic tap.
2. Stool or air is aspirated into the paracentesis syringe.
3. Multiple organisms but no PMNs are seen on the Gram stain (see Table 46-6). [78]

Polymicrobial bacterascites is essentially diagnostic of inadvertent gut perforation by the paracentesis needle. [78] Fortunately, this variant of ascitic fluid infection is the rarest; only 10 cases (0.6%) were found among 1578 paracenteses in a retrospective study. [78] Surprisingly, needle perforation of the bowel is relatively well tolerated. Only 18% (2) of 11 patients with needle perforation of the gut into ascitic fluid developed peritonitis; only 0.07% (2) of 2774 paracentesis procedures caused peritonitis. [78] Neither of these two patients required laparotomy, and none died because of the paracentesis-related peritonitis. [78] If needles larger than 22 gauge had been used, the results may have been different. It appears that patients with low-protein ascitic fluid are at most risk of developing a PMN response and clinical peritonitis related to needle perforation of the gut. [78] Most of the patients with high-protein ascites, that is, more than 1 g/dL, did not even receive antibiotics and yet did well. However, most physicians would probably feel uncomfortable withholding antibiotic treatment if needle perforation is suspected. If a decision to treat is made, anaerobic coverage should be included, such as cefotaxime and metronidazole. If a decision not to treat is made, follow-up paracentesis is helpful in following the PMN count and culture. If the number of organisms does not decrease or a PMN response occurs, antibiotic treatment should be initiated.

Tense Ascites

Some patients ignore their growing abdomens and do not seek medical attention until they can no longer breathe comfortably because of the pressure the fluid exerts on the diaphragm. This condition is called *tense ascites.* Truly tense ascites requires urgent treatment—therapeutic paracentesis. Tense ascites can be drained (even 20 L or more) without untoward hemodynamic effect. [103], [104], [105] and [106] Although discarding all the fluid obtained from patients who have difficulty synthesizing protein may be problematic, therapeutic paracentesis of cirrhotic ascites is less problematic than older textbooks of medicine would lead the reader to believe. The myth of paracentesis-related hemodynamic disasters was based on observations in small numbers of patients, probably coincidences. [104]

Patients who develop tense ascites are frequently the least compliant with or the most refractory to conventional therapy. Careful attention should be given to the education of these patients regarding the chronicity of their disease as well as diet and diuretic therapy.

Abdominal Wall Hernias

Abdominal wall hernias (umbilical and inguinal) are common in patients with ascites and may cause serious complications. In one study, 17% of cirrhotic patients with ascites were found to have umbilical hernias on admission. [107] During follow-up, 14% of these patients' hernias incarcerated, 35% developed skin ulceration, and 7% ruptured. [107] Unfortunately, attempts at repair of the hernias are fraught with complications. If the patient is a candidate for liver transplantation and the hernia is not thin walled or incarcerating, hernia repair can be postponed until the time of transplant.

If hernia repair is planned other than at the time of transplant, the ascites should be medically removed preoperatively, because the hernia recurs in 73% of patients who have ascites at the time of hernia repair but in only 14% of patients who have no ascites at the time of repair. [108] If skin ulceration develops, surgery should be performed on a semiemergency basis. Emergency surgery should be performed for hernia incarceration or rupture.

Hepatic Hydrothorax

Small pleural effusions are common in patients with cirrhotic ascites. They are usually unilateral and right-sided, but occasionally they may be bilateral with the right-side predominating. Only rarely is there a unilateral left-sided effusion; tuberculosis and pancreatic ascites are more common causes of left-sided effusions. [109] When the effusion is large and obscures most of the right lung, it is referred to as *hepatic hydrothorax.* This complication tends to occur in patients who are the least compliant with or the most refractory to therapy. This condition is thought to result from a congenital weakness in the membranous portion of the right diaphragm, which ruptures because of the elevated intra-abdominal pressure associated with ascites. [110] Occasionally, this results in sudden shortness of breath associated with decompression of the abdomen. On rare occasions, no ascites is detected in the presence of a huge pleural effusion; most likely this is caused by a one-way valve mechanism in the diaphragm.

The main symptom associated with hepatic hydrothorax is shortness of breath. Infection of this fluid is unusual. [111] When it does occur, it is usually a result of SBP.

The fluid resembles ascites, but the analysis is not identical to that of the ascitic fluid because the pleural fluid is in a system with different pressures than the portal bed. The pleural fluid protein is usually 0.75 to 1.0 g/dL higher than that of ascitic fluid from the same patient. [110]

Treatment of hepatic hydrothorax is usually more difficult than expected. Some authors have recommended tetracycline sclerosis using a chest tube. However, chest tube insertion with suction has been reported to lead to serious fluid and protein depletion and death in two patients. [112] Once a chest tube is inserted, too often it becomes very difficult to remove. Clamping the tube may cause fluid to leak around the tube's insertion site. Peritoneovenous shunt can be considered in the patient with large-volume ascites, but it is frequently accompanied by perioperative complications and shunt failure (see later). Direct surgical repair of the defect can be considered, but too often these patients are not good operative candidates. Sodium restriction and diuretics are the most effective conservative form of therapy of hepatic hydrothorax. A treatment for diuretic-resistant ascites, transjugular intrahepatic portasystemic stent shunt (TIPS), has been reported to be effective for hepatic hydrothorax (see later). [110] Thorascopic suturing of the diaphragmatic defect has been reported to be effective in an uncontrolled study. [113]

TREATMENT OF THE PATIENT WITH ASCITES

Not all forms of ascites respond to the same treatment. Therefore, the correct diagnosis of the cause of ascites formation is important. The diagnosis is usually apparent based on the history, physical examination, and the ascitic fluid analysis.

Ascites Not Related to Portal Hypertension

The most common form of low-albumin gradient ascites is *peritoneal carcinomatosis.* Peripheral edema in these patients responds to diuretics, but the ascites usually does not respond to diuretics; edema-free patients treated with diuretics lose only intravascular volume without loss of ascites. [8] The mainstay of treatment of peritoneal carcinomatosis is therapeutic paracentesis. [7], [103] Except for patients with ovarian carcinoma, patients with peritoneal carcinomatosis live only a matter of weeks; therefore, the total number of paracenteses required to minimize symptoms is not great. Patients with ovarian cancer may have a good response to debulking and chemotherapy. [7] Peritoneovenous shunts are said to be effective in malignant ascites and are associated with less morbidity than in cirrhotic ascites [114]; however, in view of the very short life-expectancy of these patients, hospitalization for installation of a shunt may not be appropriate.

Nephrotic ascites is rare and is also notable in that it is perhaps the only form of nonportal hypertension–related ascites that does respond to salt restriction and diuretics. [20]

Tuberculous peritonitis requires antituberculous therapy; there is no point in using diuretics unless the patient has concomitant portal hypertension from cirrhosis. *Pancreatic ascites* may resolve spontaneously or may respond to somatostatin infusion, but if it results from a duct leak it may require endoscopic stenting or operative intervention. [115] A *postoperative lymphatic leak* may also resolve spontaneously, but on occasion it may require surgical intervention or peritoneovenous shunting. [17] *Chlamydia peritonitis* is cured by tetracycline therapy. [18] *"Nephrogenous" ascites* (dialysis ascites) may respond to vigorous dialysis or kidney transplantation. [19]

Portal Hypertension–Related Ascites

Treatment of the Underlying Liver Disease The first step in treating portal hypertension–related ascites is to treat the underlying liver disease. In many patients in the United States, this requires convincing the patient to stop drinking alcohol. Patients with two liver insults, such as alcohol plus hepatitis C, should cease alcohol consumption. With time and healing of the reversible component of alcoholic liver disease, the ascites may resolve or at least convert from refractory to nonrefractory. [26] Patients with autoimmune chronic active hepatitis, iron-storage disease, or Wilson disease should receive the specific therapy for those diseases. By the time patients with nonalcoholic liver disease develop ascites, however, the amount of reversible component to their disease is minimal, and liver transplantation must be considered.

Determination of the Precipitating Cause of Ascites Formation In the initial management of ascites, it is also of value to determine the precipitating cause of ascites formation. Frequently, ascites accumulates during an episode of dietary indiscretion or discontinuation of diuretics; patients may decide that soup or whole milk is exactly what they think they need, and will consume large amounts of these high-salt foods. Education regarding these matters may help to prevent future hospitalizations.

Sodium Balance and the Importance of Dietary Sodium Restriction The importance of sodium balance in the management of patients with fluid overload cannot be overemphasized. [31, 116, 117] Water follows sodium passively. Sodium that enters the body must be excreted, or water will be retained. Nonurinary (gut, respiratory, skin) sodium excretion is only approximately 10 mmol/d in patients with cirrhosis and ascites. [118] Therefore, if sodium intake can be limited to 10 mmol/d and urinary excretion is 0 mmol/d, sodium balance is achieved and weight is stable. If urinary excretion of sodium is greater than 10 mmol/d beyond dietary intake, weight loss occurs. If urinary excretion of sodium is less than 10 mmol/d beyond dietary intake, weight gain occurs. Patients with no urine sodium excretion can be maintained free of ascites if they will follow a 10 mmol/d sodium diet. However, a 10 mmol/d sodium diet is not practical outside of a metabolic ward. Diuretics are used to increase urinary sodium excretion to permit a more liberal diet and yet achieve weight loss and a reduction in fluid overload (see later). [31, 116, 117] Twenty-four-hour urinary sodium measurements are useful in patients with portal hypertension–related ascites to assess the degree of sodium avidity, to monitor response to diuretics, and, indirectly, to assess compliance with diet. [116, 120] Even outpatients can collect complete specimens in 67% of instances if they are given written and verbal instructions, a 3-L container, and written orders for testing. [120] A "spot" urinary sodium concentration test may be a satisfactory substitute for a 24-hour collection. One study has shown that if the random urine sodium concentration is greater than the potassium concentration, 24-hour excretion is greater than 78 mmol/d in 95% of instances. [120] If diuretic treatment progresses satisfactorily, one does not need to know the urinary sodium excretion. However, if diuretic therapy is not progressing satisfactorily, this can result from inadequate natriuresis, failure of the patient to restrict sodium intake properly, or both. [117, 118] Monitoring 24-hour urinary sodium excretion (or urinary sodium/potassium ratios) and daily weight will usually clarify the problem. The completeness of 24-hour collections can be monitored by measurement of creatinine excretion. Male patients with cirrhosis should excrete 15 to 20 mg/kg creatinine body weight/d, and women should excrete 10 to 15 mg/kg/d. Patients who gain fluid weight despite excreting more than 78 mmol/d of sodium in the urine or despite urinary sodium/potassium ratios greater than 1.0 are eating more sodium than 88 mmol/d. [117, 118] In this setting, attention should be focused on diet education, not second-line treatment.

Diet In portal hypertension–related ascites, weight change is directly and predictably related to sodium balance. Dietary sodium restriction is, therefore, essential. The patient and cook (if different from the patient) should be educated by an enthusiastic dietitian. The more contact the dietitian has with the patient and cook, the better. One patient-dietitian encounter is not enough. Although a diet containing 500 mg (22 mmol)/d sodium is feasible in a hospital setting, it is an unrealistic goal for most outpatients. The advantage of a less-restricted, 2000 mg (88 mmol)/d hospital diet is the opportunity to establish a consistent diet and matching diuretic regimen that will continue to be effective in the patient's home environment. I use an 88 mmol/d sodium diet routinely. [117, 118]

No Indiscriminate Fluid Restriction Indiscriminate fluid restriction is inappropriate. Fluids need not be restricted unless serum sodium drops to less than 120 mmol/L. [31] Unless the decline in sodium is very rapid, cirrhotic patients do not have symptoms from hyponatremia until the sodium is well below this level. To restrict the fluids of everyone serves only to alienate patients, nurses, and dietitians. There is no evidence that fluid restriction speeds diuresis. It is the sodium restriction that is important.

Diuretics Spironolactone is the single best diuretic for patients with cirrhosis and ascites, starting with a minimum of 100 mg/d. Controlled trials have documented the superiority of spironolactone over furosemide for single-agent treatment. [121] Amiloride, starting at 10 mg/d, is more expensive (approximately fourfold) but does not cause painful gynecomastia, as spironolactone occasionally does. Spironolactone remains the mainstay of treatment of patients with cirrhosis and ascites. The half-life of spironolactone and its active metabolites is up to 5 days; it makes no sense to administer the drug multiple times per day. [122] Single daily doses of pills are most appropriate and enhance compliance. Because of the drug's long half-life, it takes almost 1 month for a patient to reach steady state regarding spironolactone effect. [122] It takes 2 weeks for any effect to be seen. In general, spironolactone *and* furosemide are used together to minimize the interval between starting therapy and natriuresis. The initial doses are 100 mg/d spironolactone and 40 mg/d furosemide, then increasing each drug simultaneously as needed. If weight loss or urine sodium concentration is inadequate, diuretics are increased up to a "ceiling" dose of 400 mg/d spironolactone and 160 mg/d furosemide. The ratio of spironolactone and furosemide can be adjusted to correct serum potassium problems. One hundred–milligram spironolactone pills are available. Patients with intrinsic kidney disease, such as patients with diabetes or IgA nephropathy, may become hyperkalemic on less spironolactone than patients without kidney disease. Combined with a 2000 mg/d sodium diet, this regimen will achieve successful diuresis in more than 90% of cirrhotic patients. [123] Intravenous furosemide causes an acute reduction of glomerular filtration rate in patients with cirrhosis; I avoid using intravenous diuretics if at all possible. [124] If rapid weight loss is desired, therapeutic paracenteses should be performed (see later). For patients who have massive edema, there is no limit to the diuretic-induced daily weight loss; once the edema has resolved, 0.5 kg/24 h is probably a reasonable maximum. [125] If patients develop encephalopathy, serum sodium less than 120 mEq/L with fluid restriction, or serum creatinine greater than 2.0 mg/dL, diuretics are usually stopped, and the situation is reassessed. Potassium abnormalities are seldom prohibitive because of the ability to adjust the ratio of the diuretics or to discontinue spironolactone in patients with intrinsic renal disease. Diuretic treatment will fail in many of the patients who develop serious complications of this therapy, and these patients will require second-line therapy. Prostaglandin inhibitors, such as nonsteroidal antiinflammatory drugs, should not be used in patients with ascites because they curtail diuresis, they may promote renal failure, and they commonly cause gastrointestinal bleeding. [126, 127] Complete removal of ascites may not be readily obtainable. However, theoretically, the concentration of ascitic fluid through diuresis increases the fluid's opsonic activity tenfold and may be of value in attempting to prevent SBP. [128] Because the waiting times for liver transplantation are 18 to 24 months in some parts of the United States and because 50% of patients with cirrhosis and ascites will die in 24 months, patients should be considered for liver transplantation after they have developed ascites (Fig. 46-6). [129]

FIGURE 46-6. Algorithm for the treatment of patients with cirrhosis and ascites. *TIPS,* transjugular intrahepatic portasystemic stent shunt. (From ref. [31].)

Refractory Ascites

Patients in whom inpatient diuretic treatment fail are labeled diuretic-resistant or refractory and are treated with second-line therapy. *Failure* is defined as minimal or no weight loss despite maximum tolerable doses of diuretics with urine sodium excretion approaching 0 mmol/d. [117, 118, 130, 131]

Liver Transplantation Patients with refractory ascites have a 50% 6-month survival and should be even more highly prioritized for liver transplantation, if they are otherwise good candidates for the procedure. [132] Other options for diuretic-resistant patients include therapeutic paracenteses, TIPS, peritoneovenous shunts, and surgical portacaval shunt (see Fig. 46-6).

Therapeutic Paracentesis Until the late 1940s, therapeutic paracentesis was essentially the only available therapy for ascites. [116] In the early 1950s, diuretics became available coincidentally with reports of complications of therapeutic paracentesis. [116] Therapeutic paracentesis rapidly fell out of favor as a treatment option for patients with ascites. In the 1980s, interest was renewed in therapeutic paracentesis. Scientific data regarding large-volume (5-L) paracentesis were reported. [133] Patients were treated with daily paracentesis followed by colloid infusion or a single paracentesis without intravenous colloid infusion. These patients tolerated large-volume paracenteses very well, just as patients have done since the time of the ancient Greeks. [116] In fact, if minor asymptomatic changes in electrolytes and serum creatinine are considered complications of therapy, therapeutic paracentesis is reported to be safer than diuresis, based on the randomized controlled trial performed

in Barcelona. [106] If the goal of treatment is to achieve complete removal of ascites while the patient is still in the hospital, paracenteses are also faster than diuresis. [106] Technically, performing a therapeutic paracentesis is similar to performing a diagnostic paracentesis, except a larger-bore needle (15–16 gauge), is used and much greater amounts of fluid are removed. Collapse-resistant tubing is used to connect the paracentesis needle to 2-L vacuum bottles. Vacuum speeds the paracentesis; 5 L can be removed in 20 to 40 minutes. Newer paracentesis needles and other equipment items are now available in the United States (see section " Technique"). The removal of 5 L of fluid by therapeutic paracentesis is considered "large volume." [106] Total paracentesis, that is, *removal of all ascites, even more than 20 L,* can be performed safely. [133] Patients with tense ascites and avid sodium retention represent a small subset of patients with ascites, however. Because more than 90% of these patients respond to a routine sodium-restricted diet and diuretics, routine medical therapy is the appropriate therapy of this large majority. [31], [117], [118], [123] In addition, therapeutic paracentesis lacks the ascitic fluid opsonin-conserving advantage of diuresis; removing and discarding ascitic fluid opsonins by means of therapeutic paracentesis could theoretically predispose patients to SBP. [134] It is my opinion that serial paracenteses should be reserved for patients who really need them, that is, the patients with truly diuretic-resistant ascites. These patients have a 24-hour urine sodium that approaches zero despite maximum doses of diuretics.

Albumin versus no albumin. In the first of their therapeutic paracentesis studies, the Barcelona group administered intravenous albumin (approximately 10 g/L of ascites removed) after each 5-L paracentesis. [106] In a subsequent study, these investigators randomized patients who received therapeutic paracentesis to albumin versus no albumin. [135] Again, if minor changes in electrolytes and serum creatinine are considered complications of therapy, the no-albumin group developed significantly more complications. Patients who did not receive albumin developed significant increases in renin and aldosterone, indicating evidence of volume depletion. However, the electrolyte imbalance and plasma renin increases were asymptomatic, and risk of progression to renal failure or death was not different between groups. [135] In addition, 31% of patients in this study received paracentesis for tense ascites in the absence of diuretic therapy. [135] Many of these patients could have been diuretic sensitive. In contrast to the results in Barcelona, a more recent study did not demonstrate the plasma renin activity rise after a single 4- to 6-L paracentesis in patients who were truly resistant to diuretics. [136] The key to explaining the difference between these studies may be found in a study demonstrating that patients with advanced cirrhosis have "circulatory hyporeactivity," whereas patients with less advanced disease are more sensitive to changes in intravascular volume. [137] It is possible that patients with truly diuretic-resistant ascites do not develop changes in plasma hormones after paracentesis in the absence of albumin infusion because of their "circulatory hyporeactivity," whereas patients with diuretic-sensitive ascites do develop changes in these hormones. The subset of patients who develop a rise in plasma renin after paracentesis has been shown to have a poor prognosis. [138] However, no study has demonstrated a survival advantage for patients who have received albumin after paracentesis. There has only been indirect evidence linking albumin infusion to a survival advantage; serial paracenteses without albumin infusion can lead to a rise in plasma renin in patients with tense ascites (but not necessarily diuretic-resistant ascites), and a rise in plasma renin is associated with a poor prognosis. [135], [138] This indirect evidence does not prove the need for postparacentesis albumin infusion. [139], [140] More convincing data involving appropriate subgroups of patients (e.g., truly diuretic-resistant patients) and regarding clinically relevant issues (e.g., survival) rather than asymptomatic laboratory abnormalities are required before albumin or other plasma expanders can be recommended. In addition, the proponents of albumin infuse one half of the volume of albumin immediately after the procedure and the remainder 6 hours later. [141] This approach converts an otherwise rapid outpatient procedure into an all-day clinic visit or a brief hospitalization. Albumin is also quite expensive and has become more expensive as its use has increased. The decision whether to infuse albumin is important from financial and logistical points of view.

Transjugular Intrahepatic Portasystemic Stent Shunt An interventional radiologic procedure that reduces portal pressure, TIPS, may be efficacious for treatment of patients with diuretic-resistant ascites. [142] This procedure consists of the following:

1. Inserting a 60-cm-long metal needle from the right jugular vein into the hepatic vein
2. Creating a fistula between the high-pressure portal system and the low-pressure hepatic venous system
3. Dilating the fistula
4. Inserting a stent into this shunt
5. Expanding the stent.

TIPS can dramatically decrease portal pressure and can reduce the tendency to retain sodium. [142] However, it does worsen the hyperdynamic circulation and lower blood pressure. These opposing effects of TIPS on factors that aggravate sodium retention would be predicted to lead to a nonuniform response of patients to this procedure. In addition, patent shunts have an approximately 25% chance of causing hepatic encephalopathy, some of which is very refractory to medical therapy. [143] Stenosis and occlusion occur in up to 50% of patients and lead to repeat cannulations and balloon dilations of the stent. [143] Conversely, use of medical-grade carbon dioxide as a contrast medium has reduced the contrast nephropathy associated with the procedure and reduced morbidity. Two randomized controlled trials of TIPS for this indication have had conflicting results. [144], [145] However, the larger and more recent study demonstrates a survival advantage compared with therapeutic paracenteses. [145] This has led to renewed enthusiasm for this procedure. A multicenter trial in the United States is nearing completion.

Peritoneovenous Shunts In the mid-1970s, the peritoneovenous shunt was promoted as a new "physiological" treatment in the management of ascites. Reports of complications of shunt insertion and shunt failure rapidly diminished enthusiasm for this treatment. Reported complications include pulmonary edema, variceal hemorrhage (from fluid overload), disseminated intravascular coagulation, thromboembolic phenomena including superior vena cava thrombosis, pseudocyst formation, and peritoneal fibrosis. [146] Unfortunately, shunt failure from thrombosis continues to be a serious problem. Most shunts clot in less than 1 year's time. "Second-generation" shunts, such as Denver shunts, have not reduced the shunt failure rate resulting from thrombosis. [147] Two large-scale randomized trials have documented no improved survival in patients with shunts compared with medically treated controls. [123], [148] Removal of most of the ascitic fluid intraoperatively before shunt insertion, with use of perioperative antiplatelet therapy, has reduced the incidence of disseminated intravascular coagulation, and replacement of ascites with saline and administration of intraoperative intravenous furosemide have decreased the incidence of pulmonary edema. [149] Peritoneovenous shunting is reserved for the very small group of patients in whom both diuretic *and* paracentesis therapy fail and who are not candidates for TIPS or liver transplantation.

Hepatorenal Syndrome

Hepatorenal syndrome is one of the terminal complications of advanced liver disease, most commonly occurring in patients with cirrhosis or alcoholic hepatitis. It has been the source of much confusion and has perhaps received more attention than it deserves. It is simply the mode of exodus of patients with advanced liver disease, when they do not develop a faster cause of death. The kidney is an innocent bystander in a very distorted milieu; transplanting a kidney from a patient with hepatorenal syndrome into a patient without liver disease and transplanting a normal liver into a patient with hepatorenal syndrome both lead to resolution of renal failure in the recipient. [150], [151] The concept that patients "die of hepatorenal syndrome" is erroneous. These patients are dying of liver failure *with* renal failure rather than dying *of* renal failure.

Pathophysiology Hepatorenal syndrome is the terminal phase of the spectrum of functional renal impairment that is characteristic of cirrhosis. As liver disease worsens, renal function deteriorates such that patients progress from compensated cirrhosis to diuretic-sensitive ascites to diuretic-resistant ascites and finally to hepatorenal syndrome. [152] This progression is relatively silent, with fluid retention and azotemia providing the evidence that the process is occurring. Vasodilation-related hypotension worsens as liver disease advances and progressive renal ischemia occurs.

Diagnosis The International Ascites Club has developed the following criteria for the diagnosis of hepatorenal syndrome:

1. Advanced chronic or acute liver failure with portal hypertension
2. Serum creatinine greater than 1.5 mg/dL or creatinine clearance lower than 40mL/min
3. The absence of shock, bacterial infection, nephrotoxic drug exposure, or rapid loss of fluid through the gut or kidneys
4. No sustained improvement in renal function with withdrawal of diuretics and plasma expansion with 1.5 L of isotonic saline
5. Proteinuria less than 500mg/d and no ultrasound evidence of obstructive uropathy or parenchymal renal disease. [152]

There is a misconception that hepatorenal syndrome is usually characterized by oliguria. This concept has been perpetuated despite data to the contrary. Actually, oliguria in hepatorenal syndrome is usually an immediate premortem phenomenon. Oliguria may be a manifestation of the renal failure associated with shock or exposure to a nephrotoxin. This acute tubular necrosis is frequently misdiagnosed as hepatorenal syndrome, but, as indicated earlier, these insults to the kidney preclude the diagnosis of hepatorenal syndrome (Fig. 46-7). When actual urine volumes were measured sequentially before and during evolution of hepatorenal syndrome in a study published in 1964, "… oliguria was the rule only during the late azotemic phase" when 52% of patients produced less than 500 mL/d. [153] In the preazotemic phase, no patient had a urine volume of less than 500 mL/d; in the early azotemic phase, only 11% of patients had a urine volume of less than 500 mL/d. [153]

FIGURE 46-7. Algorithm for the diagnosis and treatment of patients with suspected hepatorenal syndrome.

Hepatorenal syndrome usually develops in patients with cirrhosis and long-standing ascites. A prospective study documented an 18% 1-year and a 39% 5-year cumulative probability of hepatorenal syndrome in this setting. [154] Patients with cirrhosis who develop renal failure in the absence of prior ascites have a cause of renal failure other than hepatorenal syndrome. Much has been written about urinalysis in the differential diagnosis of azotemia in the setting of cirrhosis. Usually, the criteria for distinguishing one condition from another are placed in a table with no reference to the original data. One original study that has been quoted was published in 1937 at a time when diagnoses such as cholemic nephrosis were used. [155] It is difficult to find useful original data. In general, the urine protein is less than 500 mg/d, [152] and the urine sediment is "inactive" with only an occasional cast. In contrast, in acute tubular necrosis, the sediment is more "active" with more casts and cellular debris. If white blood cells are prominent in the urine, cystitis or even urosepsis may be present; bacterial infection in the patient with cirrhosis is regularly complicated by hepatorenal syndrome. [156] If red blood cells are present in large numbers along with protein but in the absence of white cells, the patient may have parenchymal renal disease resulting from IgA nephropathy. [157] Even the urine sodium concentration of less than 10 mmol/L, which is so often quoted as an important component of the diagnosis of hepatorenal syndrome, is not as useful as one would hope. Patients with urine sodium greater than 10 mmol/L have been reported. [158] Patients who have hepatorenal syndrome and who develop superimposed acute tubular necrosis may have a urine sodium concentration lower than 10 mmol/L. It has been my experience that many, if not most, patients who are labeled as having hepatorenal syndrome actually have a different diagnosis, most commonly diuretic-induced azotemia. Shock and nephrotoxin exposure are probably the next most common causes of azotemia that masquerades as hepatorenal syndrome. Parenchymal renal disease such as IgA nephropathy or diabetic nephropathy have also been misdiagnosed as hepatorenal syndrome. The relative stability of the azotemia and the abnormalities in the urinalysis help to distinguish parenchymal renal disease from hepatorenal syndrome. Before a patient is labeled as having hepatorenal syndrome, the International Ascites Club criteria should be fulfilled.

Treatment Patients suspected of having hepatorenal syndrome, based on serum creatinine greater than 1.5 mg/dL in the setting of advanced liver disease (see Fig. 46-7), should undergo the following:

1. Withdrawal of diuretics and nephrotoxic drugs
2. Urinalysis with microscopy and sodium concentration
3. Renal ultrasonography
4. Intravenous fluid challenge with 1.5 L of isotonic saline or perhaps 50 g of albumin.

A central line could be placed for monitoring of filling pressures, but this is seldom done unless there has been massive loss of fluid or there are plans for hemodialysis (see later). Serum urea and creatinine are monitored daily to assess the response over time. If the urea and creatinine continue to rise, there are three main therapeutic options:

1. Infusion of a vasoactive agent
2. Transjugular intrahepatic portosystemic stent shunt
3. Liver transplantation.

Clearly, liver transplantation is not seriously considered if there are psychosocial contraindications. If the patient is a candidate for liver transplantation and has not undergone evaluation, this should be urgently initiated. Transplantation is the most definitive treatment for hepatorenal syndrome. [151] However, a vasoactive agent is usually given whether transplantation is a possibility or not, because patients wait days or even weeks for an available organ. Intravenous dopamine by continuous infusion was used in this setting in the past, but parenteral octreotide and oral midodrine have been used with better efficacy, according to a study with historical controls. [159] In this study, octreotide was started at a dose of 100 µg subcutaneously three times daily and was increased to 200 µg three times daily. Midodrine was given at a dose of 7.5 mg three times daily and was increased as needed to 12.5 mg three times daily. Midodrine can theoretically cause hypertension, but I have not encountered this as a problem. These patients are very hypotensive and remain hypotensive on this regimen at these doses. Octreotide can also be given by continuous infusion, at a dose of 25 to 50 µg/h. [160] The optimal duration of treatment is not clear. If the azotemia does not improve within a few days of starting treatment, there is no point in continuing it. If there is a response, it appears to be useful to continue treatment for a few weeks. Patients have received it for months. If there is a response but treatment is stopped too soon, azotemia will recur. If diuretics are restarted during this treatment, azotemia will recur. Stable or improving renal function in patients with serum creatinine less than 2.0 mg/dL appears to be required before diuretics can be safely restarted, in my experience. In general, use of vasoactive agents simply "buys some time" for recovery of some reversible component of the liver (or unrecognized renal) injury. If there is no reversible component, this form of treatment may be futile or only of transient benefit. Patients awaiting liver transplantation should probably remain on the regimen until an organ is available. Anecdotally, this regimen can prevent the need for dialysis. Other vasoactive agents, such as terlipressin or ornipressin, have been used in Europe but are not available in the United States. TIPS has been reported in nonrandomized studies to reverse hepatorenal syndrome. [161] In view of the importance of portal hypertension in the pathogenesis of the process and the ability of TIPS to lower portal pressure, this form of treatment is appealing. However, randomized trials are needed. Many treatment options and anecdotes of success in treating hepatorenal syndrome have been disappointing when they are studied scientifically. Diuretic-induced azotemia masquerading as hepatorenal syndrome will respond to simple removal of diuretics. If another treatment is given in addition and the patient is (incorrectly) labeled as having hepatorenal syndrome, the additional treatment may be inappropriately given credit for the success. If TIPS is used for treatment of hepatorenal syndrome, the amount of iodinated contrast used during the procedure should be minimized. TIPS has a notoriously high mortality in the setting of alcoholic hepatitis and should be avoided in these patients; a new nomogram has been developed to predict 90-day survival after TIPS. [162] The peritoneovenous shunt failed to improve survival in patients with hepatorenal syndrome in the Veterans Administration randomized trial and is no longer advocated for treatment of this condition. [123] Hemodialysis is regularly discussed among the treatment options for patients with hepatorenal syndrome but is seldom used, except immediately before liver transplantation. Patients with this degree of liver failure are difficult to dialyze because of hypotension, and survival is very poor without eventual liver transplantation. [163] Continuous venovenous hemofiltration is better tolerated but very expensive because a dialysis nurse must be at the bedside continuously. [164] As indicated earlier, liver transplantation is the most definitive treatment for patients with hepatorenal syndrome. [151] Survival can be similar to that of patients who undergo liver transplantation without renal failure. [165]

SUMMARY AND CONCLUSIONS

The development of ascites in a patient who has cirrhosis and who is otherwise a good candidate for liver transplantation should lead to evaluation for transplantation. Patients should be rapidly prioritized for liver transplantation once they develop refractory ascites. Diet education and diuretics are the mainstays of treatment for patients who await transplantation and for those who are not candidates for the procedure. Serial therapeutic paracenteses are effective in controlling ascites in patients who are truly diuretic resistant. TIPS is a promising option for treatment of diuretic-resistant ascites, but its final place in our treatment armamentarium remains to be discerned.

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CHAPTER 47

Umaprasanna S. Karnam and K. Rajender Reddy

APPROACH TO THE PATIENT WITH A LIVER MASS

With the increasing use of imaging modalities for evaluation of various abdominal conditions, more *liver masses* are being recognized. These lesions may be small or large and may or may not be the cause of patient's symptoms. They can occur in patients with no underlying liver disease or in those with recognized chronic liver disease. They challenge the clinical acumen of many a clinician, the novice as well as the expert. Although most of the incidentally noted masses are benign, a definitive diagnosis has to be established to reassure the patient and also to be certain that a radiologically observed lesion is not malignant. In an era of cost-effective medical practice, it behooves the physician to use the most appropriate diagnostic tools associated with minimal morbidity to reach an accurate diagnosis. This chapter is devoted to the most common liver masses found in clinical practice and the advantages and disadvantages of the various diagnostic modalities, and it addresses the approach to evaluation of liver masses in special circumstances. Liver masses can be of epithelial or mesenchymal origin, and they may be benign or malignant (Table 47-1).

BENIGN TUMORS	MALIGNANT TUMORS
Epithelial Tumors	
Hepatocellular adenoma	Hepatocellular carcinoma
Bile duct adenoma	Cholangiocarcinoma
Biliary cystadenoma	Biliary cystadenocarcinoma
Mesenchymal Tumors	
Infantile Hemangioendothelioma	Epitheloid Hemangioendothelioma
Cavernous hemangioma	Angiosarcoma
Benign mesenchymoma	Embryonal sarcoma
Angiomyolipoma	Fibrosarcoma
Leiomyoma	Leiomyosarcoma
Teratoma	Hepatoblastoma
Tumorlike Lesions	
Focal nodular hyperplasia	
Nodular regenerative hyperplasia	
Biliary hamartoma	
Von Meyenburg complex	
Mesenchymal hamartoma	
Inflammatory pseudotumor	
Miscellaneous Lesions	
Simple hepatic cyst	
Metastatic liver disease	
Lymphoma	
Focal fatty change	
Liver abscess	
Echinococcal cyst	

TABLE 47-1 Lesions Presenting as Liver Masses

CAVERNOUS HEMANGIOMA

This is the most common benign mesenchymal hepatic tumor, and the prevalence ranges from 3% to 20%. [1] These lesions vary from less than 1 cm to greater than 20 cm. Giant hemangiomas have been defined as lesions that are larger than 4 cm. [2] Most often, they are noted incidentally and are of no major clinical consequence. They are rarely diagnosed in children or adolescents, and they are mostly detected in women between the third and fifth decades of life. Women more often than men have these lesions(ratio, 2 to 6:1), and the lesions tend to be larger and more numerous in women than in men of similar age. Up to 10% of patients exhibit multiple hepatic hemangiomas, and very few of them may have such lesions in other sites of the body, such as in the brain. [3]

The origin of hemangiomas is not clear. Most consider these lesions benign congenital hamartomas with growth resulting from progressive ectasia. Cavernous hemangiomas have been observed to have estrogen receptors, an observation that is in concordance with accelerated growth of cavernous hemangiomas during pregnancy and with use of supplemental estrogens (as in the use of oral contraceptives). [4], [5] and [6]

An incidental mass on abdominal imaging is the most common clinical resentation. The lesion is usually less than 5 cm in diameter, is located in the right lobe, and is asymptomatic. Large lesions tend to be symptomatic, particularly those greater than 10 cm, in which symptoms have been reported in up to 90% of the patients. The second most common clinical presentation is abdominal pain, generally in the right upper quadrant. The pathogenesis of pain may result from intralesional hemorrhage, localized thrombosis, pressure or distention of Glisson capsule, or torsion of a pedunculated hemangioma. It is important to recognize that patients could have another disorder causing the abdominal pain, and attributing any abdominal pain to a detected hepatic hemangioma is not always appropriate. A rare complication of large cavernous hemangiomas is Kasabach-Merritt syndrome, which is characterized by bleeding from disseminated intravascular coagulation (secondary to consumption coagulopathy within the hemangioma). [7] Rupture, either spontaneously or induced by trauma, is a rare yet much feared complication of a large hemangioma. [8], [9] In such cases, patients present with hypotension or shock, severe abdominal pain, and distention. Some patients may have elevated transaminases, prolongation of prothrombin time, and evidence of disseminated intravascular coagulation. Rarer presentations include obstructive jaundice, gastric outlet obstruction, hemobilia, inflammatory pseudotumor, and caval compression. [10], [11] and [12]

Laboratory tests are generally normal, except in patients with complications such as Kasabach-Merritt syndrome or obstructive jaundice. Imaging holds the key to establishing a definitive diagnosis. Ultrasonographic appearance of this lesion is of a solitary, well-demarcated, hyperechoic and homogeneous mass in up to two thirds of patients. Larger lesions may be more heterogeneous. The sensitivity of ultrasonography for diagnosing this entity ranges from 60% to 75%, and specificity ranges from 60% to 80%. [3], [13] Hemangiomas, on unenhanced computed tomography (CT) scans, are nearly isodense to large blood vessels. Dynamic CT is better than a standard CT in detecting cavernous hemangioma because the latter may fail to delineate the lesion during the phase when the lesion becomes isodense with the liver. After a rapid intravenous bolus injection of contrast medium, hemangiomas show nodular peripheral enhancement that is isodense with large vessels, and they show progressive centripetal filling in over time. Large hemangiomas (>5 cm) may have a central necrotic area or scar that may rarely calcify. Small hemangiomas may demonstrate rapid and uniform enhancement mimicking a hypervascular tumor. Whereas hemangiomas remain hypodense to blood vessels on portal venous and delayed phases, other benign as well as malignant masses become hypodense to both liver and blood vessels. The sensitivity and specificity of dynamic CT scan to delineate cavernous hemangioma range from 75% to 85% and from 75% to 90% respectively. [14], [15] Low signal intensity on T1-weighted image, high signal intensity on T2-weighted images, and multiple intralesional lobulations are characteristic findings on magnetic resonance imaging (MRI) scans of these lesions and are of help when other imaging modalities are not definitive (Fig. 47-1). Although MRI is expensive, it has the highest sensitivity (up to 95%) and

specificity (up to 95%) in the diagnosis of hepatic hemangiomas. [16] An additional advantage of MRI is that a gadolinium-enhanced study, as opposed to a CT scan, can be done in patients with renal insufficiency or iodine allergy. Further refinements of MRI technology may make it an even better tool in the detection and characterization of very small hemangiomas. Even within a cirrhotic liver, larger hemangiomas can usually be diagnosed with CT or MRI scanning, although in patients with progressive cirrhosis, hemangiomas are likely to decrease in size and to become more fibrotic, and they are difficult to diagnose radiologically and pathologically. Single photon emission CT (SPECT) has sensitivity and specificity similar to those of MRI for lesions greater than 3 cm in diameter and is less expensive, but it fails to detect smaller lesions in deeper hepatic parenchyma. SPECT is also of limited value for lesions close to the heart or major intrahepatic vessels. The current role of SPECT may be confirmatory, by clarifying equivocal findings of ultrasonography or CT. [17], [18], [19] and [20]

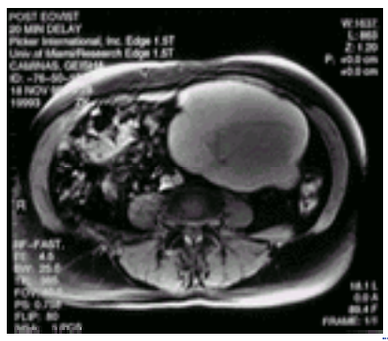

FIGURE 47-1. Characteristic appearance of hemangioma on T2-weighted MRI scan.

Laparoscopy may have to be done in a few cases to see the surface of the liver to detect these lesions. Biopsy is used as the last resort because of the potential risk of hemorrhage or rupture. Biopsies obtained with a 20- to 22-gauge needle under CT or ultrasonic guidance can be safely done, and a definitive diagnosis can be established. [21], [22] Angiography is very seldom used to diagnose these lesions.

Most hemangiomas follow a benign course, and most people live and die *with the* tumor rather than *of it*. [6] Treatment or follow-up is not indicated for asymptomatic lesions that are less than 5 cm in diameter. Follow-up may be needed in patients with asymptomatic hemangiomas greater than 10 cm in diameter. Resection is reserved for symptomatic lesions, rapidly enlarging lesions, and those that are greater than 15 cm in diameter at initial presentation, although the indication for resection based on size alone has been controversial. [23] Enucleation or resection is the surgical treatment of choice, but it may not be possible in patients with extensive hilar involvement or with multiple hemangiomas, in whom other options have to be considered. Arterial embolization has been used to control acute bleeding and to reduce blood loss at surgery. It has also been used in patients with one or only a few tumors whose vascular anatomy is unfavorable for resection but amenable to embolization. Ligation of the hepatic artery, hepatic irradiation, and intralesional steroid use have also been reported. Nonsurgical therapies should be considered only in symptomatic patients in whom the lesions are not amenable to surgery. Liver transplantation has rarely been undertaken in patients with large, unresectable tumors or in those with extensive and multiple tumors. Overall, surgical resection is indicated for fewer than 2% of diagnosed hemangiomas. [24], [25], [26], [27], [28], [29] and [30] Kasabach-Merritt syndrome can be managed by medical, radiologic, or surgical means (resection of hemangioma or liver transplantation). [31], [32]

FOCAL NODULAR HYPERPLASIA

Focal nodular hyperplasia (FNH) is the second most common benign lesion of the liver. The reported incidence of this lesion is between 2.5% and 8%. [33] This is a nodular hyperplastic lesion and has a large female predominance (female-to-male ratio of 6 to 8:1), and the average age of presentation is between 30 and 50 years. FNH represents 2% of hepatic tumors in children and is usually diagnosed between the ages of 2 and 5 years. Like hepatic hemangioma, FNH rarely causes symptoms and most often is discovered during an evaluation for abdominal pain. There are two theories regarding the pathogenesis of FNH, the vascular and the clonal. An anomalous artery supplying the lesion divides, forms a star shape, and separates the parenchyma into nodules. [34] The clonal theory holds that the cells in FNH lesion may arise from one pluripotent progenitor cell capable of differentiating into hepatocytes and biliary epithelial cells. The relationship between oral contraceptives and the occurrence of FNH has been controversial and is believed not to exist. [35], [36] and [37] Further, no correlation has been observed between the length of oral contraceptive use and the size or number of FNH lesions. However, under the influence of high hormonal states such as pregnancy or the use of oral contraceptives, the lesions may grow. [38], [39]

The lesion is most often single, subcapsular, and located in the right lobe. Multiple lesions may be seen in up to 20% of cases. Most of the lesions are 3 to 5 cm, although lesions may range from 1 mm to 19 cm. Symptoms occur more frequently in women using oral contraceptives, which could be the result of an increase in size of the lesions under the influence of these drugs. Rupture leading to hemorrhage is very rare, and malignant transformation is not a feature. Whereas physical examination is generally unremarkable, hepatomegaly and a palpable mass may be seen in a minority of patients. Histological features of FNH include a characteristic central scar with large, tortuous arteries in a matrix of connective tissue, areas of abundant intranodular bile duct proliferation, and the absence of central veins or portal triads in the nodules.

On ultrasonography, FNH is isodense to the surrounding liver tissue. Doppler scans may reveal hypervascularity secondary to enlarged arteries in the lesion. CT scan using a triple-phase protocol reveals the characteristic findings: isodense lesion in the noncontrast phase, hyperdense lesions in the arterial phase, and isodense lesion in the portal phase. FNH on an MRI is an isointense homogenous lesion on T1-weighted image and isointense to hyperintense on T2-weighted images. Contrast-enhanced MRI is probably more sensitive than CT in demonstrating the characteristic central scar, but otherwise it shows the same features as the latter (Fig. 47-2). The presence of Kupffer cells, which are capable of concentrating colloid in FNH, causes it to enhance on technetium-99m (^{99m}Tc) sulfur colloid scans. Thus, scintigraphy may help in differentiating FNH from adenomas. An MRI scan with the hepatobiliary contrast agent gadobenate dimeglumine, which demonstrates prolonged enhancement on 3-hour delayed scan, is considered very specific for FNH. The characteristic central scar may be identified in up to 20% of cases on ultrasonography, 60% on CT scans, and 78% on MRI. Angiography is rarely used to establish the diagnosis and has been used to assess arterial anatomy as part of preoperative evaluation. The combination of three-phase ^{99m}Tc diisopropyl iminodiacetic acid, ultrasound, and contrast CT, is 82% sensitive, 97% specific, and 90% accurate, as confirmed in resected lesions. Patients with lesions negative on this combination of tests and ^{99m}Tc red blood cell SPECT should undergo surgery to rule out an adenoma or malignant tumor. A needle liver biopsy is often unhelpful in the diagnosis because it may not demonstrate the features of nodularity with the central scar, which is the sine qua non for definite diagnosis, and display of such features requires a wedge biopsy. Further, these lesions are vascular and may present an added risk of bleeding. [40], [41], [42], [43], [44] and [45]

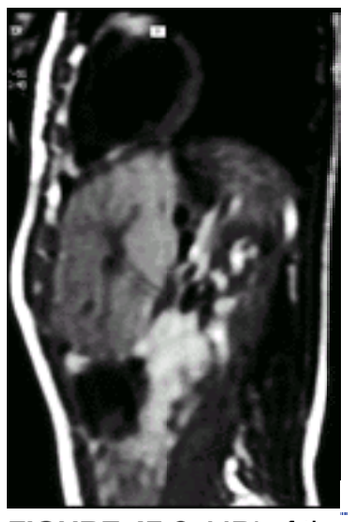

FIGURE 47-2. MRI of the liver showing a characteristic central scar of focal nodular hyperplasia.

Although surgery is not done in patients with asymptomatic FNH lesions, enucleation or wedge resection is undertaken in severely symptomatic patients or in those with rapidly enlarging lesions. Angiographic embolization and hepatic artery ligation have been done for unresectable lesions. In patients with FNH who are taking oral contraceptives, it is prudent either to discontinue these drugs in favor of other forms of contraception or to perform serial ultrasonography each year to ensure that the lesions are not increasing in size. The risk of developing symptoms and bleeding during pregnancy is remote. If there are special circumstances, serial hepatic sonography may be done during and after pregnancy. [46] /SUP>

NODULAR REGENERATIVE HYPERPLASIA

Nodular regenerative hyperplasia (NRH) is a benign proliferative process in which the normal hepatic parenchyma is replaced by diffuse regenerative nodules of hepatocytes. NRH is relatively common, and autopsy studies show a prevalence of 2%. [47] , [48] There is no clear-cut gender predilection, and NRH has been reported in children as well. The nodules vary in size from 0.1 to 1 cm. NRH has been associated with lymphoproliferative disorders, rheumatoid arthritis, primary biliary cirrhosis, bone marrow transplantation, anabolic steroids, Budd-Chiari syndrome, liver and renal transplantation, polyarteritis nodosa, hereditary hemorrhagic telangiectasia, systemic mastocytosis, amyloidosis, toxic oil exposure, partial hepatectomy, Felty syndrome, and hepatocellular carcinoma, in addition to others. [49] , [50] , [51] , [52] , [53] and [54] There are two theories regarding the pathogenesis of NRH. The vascular theory postulates that the basic lesion is portal venous obstruction that results in atrophy of hepatocytes in zone III. This leads to a compensatory proliferation of hepatocytes from portal region, which then form regenerative nodules. [55] Portal hypertension may result from compression of the intrahepatic portal radicles by the regenerating nodules. The second theory postulates that NRH is a generalized proliferative disorder of the liver. [56] Attesting to this is the observation that three-fourths of patients with NRH and hepatocellular carcinoma have liver cell dysplasia. The question of NRH as a predisposing factor to hepatocellular carcinoma is still not definitively answered.

The clinical presentation of patients with NRH varies from an incidental finding in asymptomatic patients at one end of the spectrum to hepatic failure at the other extreme. Portal hypertension with ascites may occur in 5% to 8% of patients, and esophageal varices may occur in up to 13%. [57] Patients may progress to liver failure, and liver transplantation has been performed in a few instances, although recurrence of NRH after liver transplantation has been known to occur. [58]

Hepatosplenomegaly may be noted on clinical examination, and nonspecific elevations in hepatic biochemical tests have been observed. NRH may be suspected when a patient presents with symptoms of portal hypertension and a liver biopsy fails to show cirrhosis or is interpreted as normal. Histological findings of NRH include regenerating hepatocytes, curvilinear compression of the central lobule, and lack of fibrous scar that may not be detected on a needle biopsy; laparoscopy with direct visualization of the liver or an open liver biopsy may be needed in certain cases.

Ultrasonography reveals hepatic nodules, which are isoechoic or hyperechoic. On CT scan, the nodules are nonenhancing and hypodense. Features of portal hypertension such as varices and ascites may be seen. Enlargement of the caudate lobe, peripheral hepatic hypoperfusion, and narrowing or obstruction of the hepatic veins or vena cava suggest Budd-Chiari syndrome as a cause of NRH. MRI is more accurate than CT in demonstrating vascular patency and may show more nodular lesions. The large regenerative nodules are usually hyperintense on T1-weighted images and hypointense on T2-weighted images. The scan may also show hypervascular enhancement, thus helping to distinguish NRH from cirrhotic regenerative nodules and hepatocellular carcinoma. ^{99m}Tc-sulfur colloid scintigraphy may be normal or may demonstrate uptake in nodules of larger size. [40] , [41] and [42]

HEPATIC ADENOMA

Hepatic adenoma is a lesion characterized by the benign proliferation of hepatocytes. Hepatic adenoma is found predominantly in young or middle-aged women. These lesions are usually solitary, and a few patients have multiple lesions. Up to one third of lesions are larger than 10 cm. [59] Rare before the introduction of oral contraceptives, the incidence of these tumors increased after 1960s, and they were observed more commonly in patients using high-dose estrogens, and for twice as long, when compared with controls ($P < 0.001$). [60] The incidence appears to have decreased, and this change may result from a reduction in the dose of estrogens in oral contraceptives. Although the annual incidence of hepatic adenoma is 1 to 1.3 per million in women who have never used oral contraceptives, it increases exponentially to 34 per million in long-term users of oral contraceptives. Hepatic adenomas are also seen in glycogen storage disease, diabetes mellitus, pregnancy, and use of androgens. Multiple adenomas may occur in patients with glycogen storage diseases types 1 and 3 (incidences of 50% and 25%, respectively) at an earlier age and have a male predominance. [61] Malignant transformation occurs at a faster rate in this subset of patients. Single or multiple adenomas may also arise in men and children without any predisposing conditions. There is some evidence that hepatic adenoma is derived from a single clone of hepatocytes. Altered cadherin expression (normally mediates calcium-dependent cell-to-cell adhesion) has been described in a few cases of hepatic adenoma. [62]

Hepatic adenoma is frequently symptomatic, with the most common symptom being epigastric or right upper quadrant pain; 30% of the patients may have severe and sudden pain. Bleeding within the lesion and tumor necrosis may be responsible for abdominal pain. Chronic, vague intermittent abdominal pain, hepatomegaly, and an abdominal mass may be other features. Symptoms of severe pain and signs of hypotension and shock may herald intraperitoneal or subcapsular hemorrhage, which could be potentially fatal. Perimenstrual flare of symptoms has also been noted. [63]

Laboratory parameters are usually normal. Sonography reveals a well-demarcated, smooth lesion with variable internal echogenicity, and Doppler studies may show venous signals within the lesion (Fig. 47-3). CT demonstrates most adenomas as sharply demarcated masses with smooth borders (Fig. 47-4). In contrast to FNH, hepatic adenoma may be heterogenous because of the presence of hemorrhagic necrosis or fat. When hepatic adenoma occurs in association with glycogen storage disease, the initial image may appear hypodense to the surrounding parenchyma. CT scan may demonstrate calcification, fat, and hemorrhage and tumor capsule, in a minority of patients. MRI has a low signal on T1-weighted images, and the lesion heterogeneously enhances on T2-weighted images. Recent hemorrhage or increased glycogen may increase the T1 signal, whereas central necrosis appears as low signal intensity on T2. Even though adenomas contain well-differentiated hepatocytes and may form bile, they characteristically lack the bile ductules to excrete bile. ^{99m}Tc sulfur colloid scintigraphy may show some traces of uptake in hepatic adenoma, but in contrast to FNH, there is no delayed excretion. One characteristic of hepatic adenoma seen on all imaging modalities is the well-encapsulated features of the adenoma with well-defined borders. Angiography has been performed to differentiate hepatic adenoma from hepatocellular carcinoma, with vascular leaking, arteriovenous shunting, and portal venous invasion suggesting the latter. [64] , [65]

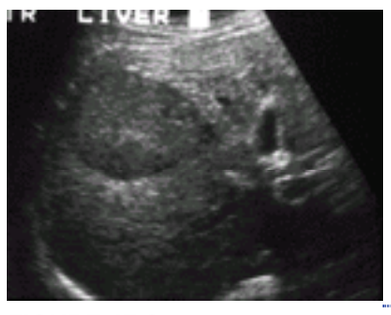

FIGURE 47-3. Ultrasonogram showing the well-encapsulated character of hepatic adenoma.

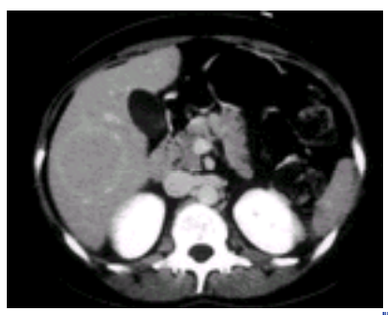

FIGURE 47-4. CT scan showing the characteristic appearance of a hepatic adenoma.

Liver biopsy to make a diagnosis is not preferred if adenoma is clinically suspected because of the risk of bleeding from these vascular lesions. Further, these lesions may have a focus of a malignant lesion that may not be evident on a biopsy done from a particular area. [66], [67] Risk of malignancy and symptomatic complications mandate surgical removal of suspected hepatic adenomas. In women taking oral contraceptives, a brief period of observation after removal of estrogens may be prudent, because adenomas have been reported to regress when patients discontinue oral contraceptive use, starting as early as 6 months. [68] However, a disadvantage is that these lesions may rarely de-differentiate into hepatocellular carcinoma even after they have been observed to regress radiologically. Avoidance of oral contraceptives permanently is necessary. In female patients whose adenomas are not resected, pregnancy should be undertaken with caution and with an understanding that there is an increased risk of tumor growth, hemorrhage, and fatality. Surgery is safe in the elective setting, with a mortality rate of less than 1%, but the rate increases when intraperitoneal bleeding has already occurred. [69] Arterial embolization has been used to control bleeding, to reduce the tumor size preoperatively, and to relieve symptoms in patients who are not candidates for operation. Multifocal adenomas may require orthotopic liver transplantation. In glycogen storage disease, adenomas may regress with nocturnal continuous feeding, but orthotopic liver transplantation is likely to correct the primary metabolic defect and should be considered. If liver transplantation is not feasible in patients with glycogen storage disease, twice yearly ultrasonography is recommended to monitor these lesions. Patients with glycogen storage disease and hepatic adenoma have not shown changes in number or size of these lesions during pregnancy.

The condition of liver adenomatosis (more than 10 adenomas) is a distinct entity, and no association exists between it and either oral contraceptive use or glycogen storage disease. These patients are at greater risk of bleeding, rupture, and malignant transformation of the lesions. This entity should be a diagnosis of exclusion because metastatic disease and multifocal hepatocellular carcinomas are more common causes of multiple solid liver masses. Diagnosis hinges on histology, and orthotopic liver transplantation may be considered in these patients. [70], [71]

CYSTIC LESIONS

Biliary Cystadenoma

Biliary cystadenoma and its malignant counterpart, cystadenocarcinoma, are the most commonly encountered primary cystic neoplasms of the liver. This lesion is more common in women, ranges in size from 2.5 to 28 cm, and can occur at extremes of age, although the mean age of presentation is usually in the fourth decade. [72] These lesions usually arise in the liver parenchyma, although rarely they arise from the extrahepatic biliary tree or gallbladder. There is no association between biliary cystadenoma and oral contraceptive use. The most common presenting symptom is right upper quadrant abdominal discomfort. Imaging studies show a focal mass with thick walls, cystic spaces, and septations. Malignant transformation to cystadenocarcinoma has been described in up to 25% of cases, and no imaging modality can clearly distinguish the benign from the malignant lesion. Hence complete surgical excision is the treatment of choice in patients with suspected cystadenomas. [72], [73]

Solitary Hepatic Cyst

These lesions have a prevalence of 4% and are usually discovered incidentally in women (female-to-male ratio, 4:1). The cyst is commonly found in the right hepatic lobe, and only a few of the lesions are larger than 5 cm in diameter. Although lesions are often asymptomatic and noted incidentally, patients infrequently have the clinical presentation of right upper quadrant abdominal pain, and this pain usually occurs in patients with cysts larger than 5 cm in diameter. Intracystic hemorrhage, infection, and neoplasia are some of the reported complications. Asymptomatic solitary hepatic cysts are best managed conservatively. Laparoscopic unroofing of accessible lesions is the preferred treatment of symptomatic solitary cysts. Several therapeutic approaches have been used for large simple cysts, including needle aspiration, with or without injection of sclerosing solution, internal drainage with cystojejunostomy, wide unroofing, and varying degrees of liver resection. [74], [75]

Echinococcal Cyst

Echinococcal (hydatid) cysts of the liver are caused by the larval form of *Echinococcus granulosus,* acquired usually from infected dogs. These cysts are fluid-filled structures limited by a parasite-derived membrane, which contains germinal epithelium. Hydatid cysts of the liver are uncommonly encountered in the United States, and patients are often asymptomatic. Symptoms may result from the mass effect of an enlarging cyst or from complications such as intraperitoneal leakage, infection, or biliary obstruction. Communication with the biliary tree is found in at least 25% of patients. Minor leakage of cyst content into the peritoneal cavity may produce a mild acute allergic reaction that may manifest with urticaria and abdominal discomfort. In cases of major rupture, patients often have acute peritoneal signs and associated acute anaphylaxis that may be fatal. Echinococcal cysts most often present as a hepatic mass with a typical appearance on abdominal imaging, coupled with confirmatory serologic testing by indirect hemagglutination or enzyme-linked immunosorbent assay. These tests have a sensitivity of 85% to 100% and a specificity of 88% to 96% for echinococcal infection. Ultrasonography, CT, and MRI typically demonstrate a multilocular cystic lesion with daughter cysts or calcification of the cyst walls. In complicated disease, the images may also show communication of the cyst with the biliary system or external leakage of cyst material. Examination of the stool is unhelpful, because fecal eggs are not present in the human host. The Casoni skin test has largely been abandoned because of its low specificity and potential danger of a severe local allergic reaction. Asymptomatic cysts that have a completely calcified wall and are associated with a negative or low indirect hemagglutination test may contain no active scolices. These cysts have a very low likelihood of complications and should probably be treated conservatively. In the rest of the cases, the treatment of choice consists of either surgery or percutaneous drainage. Medical therapy with mebendazole or albendazole is reportedly not effective in most cases of hydatid disease. However, the use of these drugs has been advocated as a preoperative measure. Therapeutic approaches consist of surgical evacuation and resection or excision of the cysts, with prior irrigation by hypertonic saline as a scolicidal agent. Other scolicidal agents that have been tried for this purpose include formalin, hydrogen peroxide, silver nitrate, and absolute alcohol. It has been suggested that percutaneous drainage combined with albendazole therapy may be an effective and safe alternative to surgery for the treatment of uncomplicated hydatid cysts of the liver, although this approach has not been widely accepted in North America. [76], [77]

Bile Duct Hamartoma

Also known as bile duct adenoma and von Meyenburg complexes, this benign focal lesion is characterized by disorganized proliferation of bile ductules in a matrix of fibrocollagenous stroma. The lesions are almost always asymptomatic, less than 1 cm in diameter (although rarely they may coalesce into larger lesions), and seen on the surface of the liver during laparoscopy or laparotomy. They may be mistaken for metastases or microabscesses. [78] On CT scan, they appear as hypodense lesions on either nonenhanced or contrast-enhanced studies. The further addition of MRI in the evaluation of these lesions is of little value. There is no risk of malignant degeneration, and no treatment is warranted. Von Meyenburg complexes (biliary microhamartomas) are 2 to 4 mm in diameter

and can be considered the diminutive counterpart of biliary hamartoma. These are often multiple and may occur in association with fibrocystic disease of the liver such as polycystic liver disease, congenital hepatic fibrosis, and Caroli disease. [79]

Mesenchymal Hamartoma

An extremely rare benign tumor thought to arise from the portal tract mesenchyme, it usually presents in boys before 3 years of age; cases have been reported in adults as well. A few case reports suggest a relationship between mesenchymal hamartoma and embryonal sarcoma of the liver. It usually presents with abdominal enlargement or a mass. Congestive heart failure (secondary to arteriovenous shunting) and ascites (secondary to cyst rupture) are uncommon presenting features. The lesion is usually large, single, and well demarcated. It has a smooth surface, and it is soft and fluctuant when palpated at surgery. Lesions may calcify and regress in size. Ultrasound scans show cystic locules that are isoechoic or anechoic, and CT scans show a low-attenuation multilocular mass with septations. In general, these lesions have low signal intensity on T1-weighted images and high signal intensity on T2-weighted images. Large and symptomatic tumors may be resected. [80], [81] and [82]

HEMANGIOENDOTHELIOMA

This entity includes the *infantile form,* which is the most common benign hepatic tumor in children, [83], [84], [85] and [86] and the *epitheloid form,* which is a malignant lesion of adults. [87], [88] The former presents in infancy with mass effect, hepatomegaly, cardiac failure, or consumptive coagulopathy (Kasabach-Merritt syndrome). Cutaneous hemangiomas are found in the majority of cases (in contrast to the epithelioid variant), and other associated entities include renal agenesis, patent ductus arteriosus, myelomeningocele, and atrial septal defect. Epithelioid hemangioendothelioma presents in adults with hepatomegaly, abdominal pain, and low-grade fever, and patients may have normal hepatic biochemical tests. Its development has been associated with the use of contraceptive steroids, exposure to polyvinyl chloride, arsenic, and radiocontrast medium (Thorotrast). Sonographic appearance is variable, and the lesions may be hypoechoic or hyperechoic. Unenhanced CT scans usually reveal a low-attenuation mass, and calcification may be noted in 50% of cases. Large tumors demonstrate peripheral pooling of contrast material on arterial and early portal venous images before filling in from the periphery. The lesions are hypointense on T1-weighted images and hyperintense on T2-weighted images. The enhancement after gadolinium administration is similar to that on contrast CT scan. [89], [90] On all imaging modalities, the infraceliac aorta is abnormally small, related to shunting of a large part of cardiac output into the tumor through the celiac vessel. The infantile type is sometimes responsive to medical therapy (steroids) or hepatic artery embolization. Surgical resection is resorted to when other measures fail. Overall, this entity carries a favorable prognosis. In contrast, the epithelioid variant is poorly responsive to medical therapies, and resection or liver transplantation is favored. [91], [92]

ANGIOMYOLIPOMAS

Angiomyolipomas are solitary, benign hepatic tumors that occur predominantly in women between the fourth and seventh decades of life. This lesion is composed of varying proportions of fat, as well as smooth muscle with thick-walled blood vessels. This lesion is also considered to be a choristoma (a mass of maldeveloped tissue not usually found in the organ). [93] Unlike its counterpart in the kidney, hepatic angiomyolipomas are rarely associated with tuberous sclerosis. Large tumors may be symptomatic, with abdominal pain. The tumor stains positively with antibodies to HMB-45 (a marker of melanocytic differentiation), and this characteristic feature aids in its diagnosis. [94] Ultrasonography reveals a homogeneously hyperechoic mass. CT is more specific for the diagnosis, and most lesions show a density component of -20 Hounsfield units on CT, indicative of adipose tissue (Fig. 47-5). Because the fat component can range from 5% to 90% in the lesion, it may be difficult to differentiate these tumors from hepatocellular carcinoma. Angiography may reveal a hypervascular lesion. Malignant transformation is unknown, and resection is the treatment for symptomatic or suspicious lesions. [93], [95]

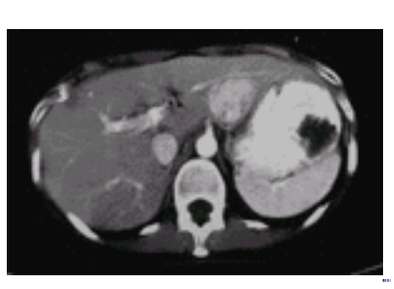

FIGURE 47-5. CT scan showing a variegated appearance suggestive of hepatic angiomyolipoma.

FOCAL FATTY INFILTRATE

Focal fatty infiltrate is an ill-defined lesion, seen on imaging, resulting from macrovesicular steatosis involving contiguous acini with no distortion of acinar architecture. These lesions may be single or multiple and may mimic malignant tumors. Focal fatty infiltrate is associated with alcoholism, obesity, malnutrition, total parenteral nutrition, chemotherapy, hypertriglyceridemia, diabetes mellitus, and acquired immunodeficiency syndrome. Nonspecific abnormalities in hepatic biochemical tests have been observed. Ultrasonography reveals a hyperechoic lesion with ill-defined borders. On CT scans, the lesions are hypodense and sharply demarcated, and the demonstration of normal caliber vessels traversing the lesion on helical CT is characteristic. MRI shows a hyperintense lesion on T1-weighted images. Because the Kupffer cells are not affected, ^{99m}Tc sulfur colloid scintigraphy is not helpful. A liver biopsy may be needed to rule out malignancy definitively. Resolution of the lesions on correction of the underlying medical problem has been observed.

INFLAMMATORY PSEUDOTUMOR

Inflammatory pseudotumor represents less than 1% of all benign focal lesions of the liver and presents primarily in middle-aged men with fever, jaundice, weight loss, abdominal pain, and malaise. Although cultures of the lesion are negative, this entity is thought to result from a localized infection. Leucocytosis, elevated erythrocyte sedimentation rate, and normal a-fetoprotein values help in differentiating it from a neoplastic process. Sonography reveals a mosaic pattern. CT scans show an irregular, clearly demarcated lesion that does not enhance with contrast. Spontaneous regression has been reported in a few instances. When there is any doubt about the diagnosis, resection is in order. [96], [97]

Hepatocellular carcinoma, cholangiocarcinoma, and meta- static diseases of the liver are discussed in other portions of this textbook.

LAPAROSCOPY

Laparoscopy is a minimally invasive modality that has the advantage of direct visualization of the liver. Advances in technology make it an attractive option when results of noninvasive techniques are equivocal and when the clinician is reluctant to embark on exploratory laparotomy. Laparoscopy is currently done as an outpatient procedure. In addition to visualizing the lesions present on the surface of the liver, it also provides valuable information about the rest of the liver parenchyma (e.g., fatty liver or cirrhosis). Peritoneal involvement secondary to metastatic disease can be appreciated even when CT and MRI findings are negative. Biopsies of hepatic lesions can be obtained under direct visualization, thus increasing the accuracy of diagnosis.

CLINICAL APPROACH

> In medicine we have diagnosis, which is a matter of faith; prognosis, which is a matter of hope; and treatment, which is only too often an affair of charity. But the greatest of these is diagnosis. For without a proper diagnosis, it is impossible to forecast the course and outcome of a disease or to treat it satisfactorily.
>
> Sir Robert Hutchison

The foregoing quotation is very apt in the evaluation of liver masses. The approach to a liver mass is predicated on several factors such as the age and gender of the patient, the mode of presentation, methods used in diagnosis, the presence of underlying liver disease, and the degree of certitude desired by the clinician and the patient. The diagnosis and therapy of a liver mass have to be tailored to a given case, and rigid adherence to any protocol may be detrimental. The algorithms in this chapter serve as a broad framework, and the astute clinician can *doctor* this approach to

reach a favorable outcome. In some situations, referral to a tertiary care center would be prudent.

Advances in imaging technology have increased the precision of establishing a definitive diagnosis. In a prospective study, preoperative diagnoses was established by means of clinical history, serum tumor markers, sonography, and spiral CT scan in 160 patients having 225 focal liver lesions. [98] MRI, angiography, and lipiodol CT were added to the foregoing tests when deemed necessary. All patients underwent surgery, and a definitive pathological diagnosis was obtained for all the lesions. The preoperative diagnosis was confirmed in 98.2% of the lesions, and the indications for resection were correct in 97.5%.

The sensitivity, specificity, positive predictive value, negative predictive value, and accuracy of this approach for benign lesions, for cholangiocarcinoma, and for metastases were also high (Table 47-2). Thus, clinical acumen with appropriate radiologic studies does provide a high degree of accuracy in the appropriate diagnosis of lesions in the liver and should be reassuring to patients. However, exceptions do occur, and histological examination, either by a needle biopsy or by wedge resection, may ultimately be needed before a definitive procedure. *A simplified algorithm for evaluation of focal liver masses is presented in* Figure 47-6.

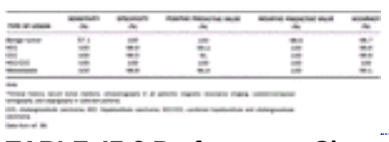

TABLE 47-2 Performance Characteristics of Diagnostic Workup* in Evaluating Liver Mass Lesions

FIGURE 47-6. A simplified algorithm for evaluation of patient with a liver mass.

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CHAPTER 48

Peter C. Hayes and Kenneth J. Simpson

APPROACH TO THE PATIENT WITH FULMINANT (ACUTE) LIVER FAILURE

Fulminant (acute) liver failure (FLF) is a relatively uncommon condition characterized by hepatocyte necrosis and disruption of liver function in proportion to the degree of liver cell death. With extensive hepatocellular necrosis comes the development of clinically obvious hepatic encephalopathy, which defines the clinical condition of FLF. The challenge for physicians is to recognize this condition before the onset of encephalopathy, thus allowing early transfer of patients to specialized units that can offer the prospect of liver transplantion for those patients who are deemed to have a poor prognosis. The most common causes of FLF are acetaminophen poisoning and acute viral hepatitis or presumed viral hepatitis (non-A, non-B, non-C hepatitis). The development and clinical introduction of a bioartificial liver will, one hopes, lead to an improvement in the prognosis of this devastating condition.

The chapter first discusses the definition, pathogenesis, and etiology of FLF. Thereafter, the clinical management is divided into pre–intensive care unit (ICU) and ICU management of patients with FLF. Subsequently, liver transplantation and developments in transplantation are discussed, as well as the available bioreactors for use in patients with FLF.

FLF is the consequence of severe hepatic injury. This results in a sudden deterioration in hepatocellular function and leads to a characteristic clinical syndrome with hepatic encephalopathy and, in severe cases, multiorgan failure and death. There are many causes of FLF, which vary in their relative contribution in different parts of the world. Supportive therapy allows both functional and structural regeneration of the liver after FLF, without the development of chronic liver disease. However, in certain cases, the prognosis is grave and is greatly improved by emergency liver transplantation. One hopes that, with the development and refinement of bioartificial liver systems, hepatocyte transplantation, and gene therapy, this reliance on emergency liver transplantation will eventually be of historical interest only.

DEFINITIONS

The original clinical syndrome of FLF was defined by the Fulminant Hepatic Failure Surveillance Study in 1970 as a “potentially reversible condition, the consequence of severe liver injury, in which the onset of hepatic encephalopathy was within the 8 weeks of the first symptoms of illness and in the absence of pre-existing liver disease.” [1] Subsequently, there has been considerable debate about refining this definition. Patients developing hepatic encephalopathy within 8 to 24 weeks of the onset of symptoms were defined as having “late-onset hepatic failure.” [2] Several authors have suggested that the interval between the first detection of jaundice and the onset of hepatic encephalopathy would allow more accurate timing of the duration of illness. Indeed, the duration of illness, as defined by the interval between the onset of jaundice and the development of hepatic encephalopathy, has been shown to be of prognostic significance in patients with FLF that is not induced by acetaminophen. [3] However, there is a lack of agreement regarding the crucial time period between the onset of jaundice and the development of hepatic encephalopathy. Workers from the Hospital Beaujon, in Paris, France use the term FLF to describe the onset of encephalopathy within 2 weeks of the development of jaundice and subfulminant hepatic failure when hepatic encephalopathy develops between 2 weeks and 3 months from the development of jaundice. [4] Others at King’s College Hospital, in London, England have suggested that FLF should be replaced by the term “acute liver failure” and subcategorized into three groups: hyperacute liver failure, in which the interval between jaundice and encephalopathy is 7 days or less; acute liver failure, for cases in which the interval is 7 to 28 days; and subacute liver failure, for cases in which the interval is 5 to 12 weeks. [5] Such a classification of FLF has the advantage that certain causes, complications, and prognoses are more common in the different subgroups. For example, hyperacute liver failure is most commonly seen after acetaminophen poisoning, and it has a high frequency of cerebral edema, but paradoxically it is associated with a relatively good prognosis.

A clinically relevant problem is how to define the concept of a pre-encephalopathy stage of FLF or subfulminant hepatic failure. Many more patients show clinical and biochemical evidence of severe liver injury but never develop hepatic encephalopathy. In these patients, who may not be managed by hepatologists, it is important to anticipate and monitor closely for the development of hepatic encephalopathy. The ability to predict which patients with severe liver injury will deteriorate clinically with the development of encephalopathy will allow earliest referral to a specialized liver center and will avoid the known dangers of transporting patients with encephalopathy. This pre-encephalopathy stage of FLF or subfulminant hepatic failure has been defined as “severe acute hepatic failure” and as “impending” or “incipient” FLF. [6] In France, this condition is diagnosed whenever the prothrombin ratio falls to less than 50% of normal during the course of an acute liver disease, and does not require the presence of clinical encephalopathy. [7] Even using widely accepted criteria in the United Kingdom for transfer of patients with acetaminophen poisoning to specialized liver centers, data from our own unit have shown that only 50% of these patients will subsequently develop hepatic encephalopathy.

Throughout this chapter we use the designation FLF, and for the purposes of discussion, we use the original definition of Trey and Davidson, namely, the development of hepatic encephalopathy within 8 weeks of the onset of symptoms in a patient without preexisting liver disease. The term *subacute hepatic failure* is used to define the syndrome whereby hepatic encephalopathy develops within 8 weeks to 6 months of the onset of symptoms. These definitions were used by Lucey and colleagues when discussing transplant listing and by Schiodt and colleagues in the Acute Liver Failure Study Group. [8], [9]

DIFFERENTIAL DIAGNOSIS

The constellation of clinical and biochemical features including hepatic encephalopathy, jaundice, transaminitis, and coagulopathy often makes the diagnosis of FLF. However, the distinction from decompensated chronic liver disease may be difficult, especially if there is a prolonged clinical course with the development of ascites. Careful review of all the patient’s medical records may reveal previous evidence of liver abnormality. The presence of the cutaneous stigmata of chronic liver disease, the clinical or radiologic detection of splenomegaly, and the endoscopic appearances of esophageal varices are more suggestive of chronic liver disease in patients who have only had jaundice for less than 2 weeks. Ultrasound often fails to differentiate between collapse and cirrhosis, but it may be useful in identifying uncommon causes of FLF such as Budd-Chiari syndrome and metastatic malignant disease. Occasionally, liver biopsy is required. Because of the coagulopathy, this is most safely performed by the transjugular route. At the same time, measurement of the hepatic venous pressure gradient may suggest the occurrence of portal hypertension, which is uncommon in patients with a short history of jaundice and encephalopathy. The liver biopsy, as well as excluding cirrhosis, may be helpful in providing some prognostic information. In one study, hepatic necrosis greater than 70% was associated with a poor prognosis. [10] /SUP>Certain other clinical situations may be confused with FLF. Some patients with severe intra-abdominal sepsis or pancreatitis may develop a syndrome very similar to FLF. Tropical diseases such as falciparum malaria and dengue fever may be confused with FLF.

INCIDENCE AND ETIOLOGY

Accurate data on the incidence of FLF in populations throughout the world is limited. It has been suggested that, in the United States, there are 2000 deaths from acute viral hepatitis annually. [11] Alternatively, using International Classification of Diseases codes on death certificates, approximately 5000 potential deaths from FLF were identified in the United States, between 1980 and 1988, with an incidence of 3.5 deaths per million population. [11] Examination of the National Hospital Discharge Survey between 1988 and 1990 gave an estimate of hospitalizations for FLF at 31.2 per

million population. [11] The distribution of causes of FLF is different in different countries throughout the world: In general, viral hepatitis is the most common cause of FLF in developing countries, and in the West, toxic drug reactions and acetaminophen poisoning are the more common causes (Table 48-1).

CAUSE	INVESTIGATION
Acetaminophen poisoning	Acidosis, early renal failure, very high transaminases (>5000), detectable acetaminophen
Idiosyncratic drug reactions	Positive serology
Budd-Chiari syndrome	Ultrasound, CT scan, MRI, hepatic venogram
Wilson disease	Slit-lamp examination, hemolytic anemia, relatively normal transaminases, ceruloplasmin, urinary and serum copper
Hepatic malignancy	Ultrasound scan, CT scan, MRI, transjugular biopsy
Autoimmune hepatitis	Immunoglobulins, antinuclear factor, anti–smooth muscle antibody
Viral Hepatitis	
Hepatitis A	IgM anti-HAV
Hepatitis B	IgM anti-HB core
Hepatitis D	HDV, RNA, IgM anti-HDV, HDV antigen

Note:
CT, computed tomography; HAV, hepatitis A virus; HB, hepatitis B; HDV, hepatitis D virus; IgM, immunoglobulin M; MRI, magnetic resonance imaging.

TABLE 48-1 Fulminant Liver Failure: Causes and Helpful Diagnostic Investigations

Acetaminophen Toxicity

Acetaminophen is the most common single cause of FLF in the United States. [9] The relative frequency has increased from 20% (1994 to 1996) to 36% (1998 to 1999), but it still does not reach the numbers seen in the United Kingdom (between 70% and 80%). Changes in the packaging of acetaminophen and restriction in availability may have reduced its relative frequency, but the early data are still contradictory, and acetaminophen remains the most common cause of FLF in the United Kingdom. [12], [13] Acetaminophen poisoning is less common in Australia and other European countries and is rarely encountered in India. [14], [15] In contrast to the United Kingdom, where more than 90% of cases are associated with attempted suicide, in the United States, most patients (64%) report accidental toxicity. [16], [17] Other reports have highlighted the potential fatal ingestion of therapeutic doses of acetaminophen, which may induce FLF in patients taking enzyme-inducing drugs (e.g., phenobarbitone or isoniazid) or in those who have been fasting or who consume excessive amounts of alcohol.

The diagnosis of acetaminophen-induced FLF is suggested by the presence of markedly elevated serum transaminases (>100 times normal), acute renal failure, metabolic acidosis, and hypophosphatemia. In some cases, acetaminophen may still be detectable within the blood because, as liver injury progresses, the plasma clearance of acetaminophen is reduced. As discussed later, the prognosis of patients with acetaminophen toxicity may be difficult to assess, especially in patients with staggered or accidental overdoses. However, the spontaneous rates of survival are higher in acetaminophen-induced FLF compared with other causes.

The hepatotoxic affects of acetaminophen are dependent on its metabolic activation by several members of the cytochrome P450 system producing the highly reactive intermediate, *N*-acetyl-*para*-benzoquinoneimine (NAPQI). NAPQI is normally conjugated with glutathione, but levels accumulate once cytoplasmic glutathione becomes depleted. However, the exact mechanism by which NAPQI leads to hepatocyte death is not clear. Interesting studies in animals have implicated a central role for Kupffer cells; depleting Kupffer cells with gadolinium chloride completely protects against the liver injury in vivo in the rat. [18] Other workers have suggested a role for Fas-mediated apoptosis. [19]

Acute Viral Hepatitis

Acute viral hepatitis causes 10% of cases of FLF in the United States. A similar frequency has been reported in the United Kingdom. [14] Viral hepatitis is the most common cause of FLF in France (50%) and is the cause of FLF in most cases in India. [15] Hepatitis B virus (HBV) is the most commonly isolated, and its relative frequency is determined by the carrier rate in a certain country. The reported frequency in the United States (5% of cases of FLF) fell slightly between 1994 to 1996 and 1998 to 1999, perhaps reflecting increasing vaccination rates. [14], [20] Approximately 1% of symptomatic patients with acute hepatitis B are at risk of developing FLF. [11] The risk is higher in women and after cytotoxic chemotherapy. [21] FLF associated with hepatitis B can occur during seroconversion to anti-HBe positivity and with coinfection or superinfection with hepatitis D virus (HDV). Superinfection with HDV is more likely to result in FLF because the antigen (HBsAg) essential for HDV replication is already present. The presence of IgM anti-HBc antibody at high titer is diagnostic. In up to 20% of patients, HBsAg is lacking, and HBV DNA is undetectable; these findings may be associated with improved prognosis. [21] In a few patients without any serologic markers of HBV infection, HBV DNA has been detected by the polymerase chain reaction in liver tissue. [22] The detection of HDV RNA, IgM anti-HDV, or HDV antigen suggests superinfection or coinfection with HDV. Patients surviving HDV-associated FLF become chronic carriers of both HDV and HBV. This situation is far more common after HDV superinfection and frequently leads to severe chronic hepatitis and cirrhosis. [23] The prognosis of HBV-associated FLF is generally good. [3]

Several factors are important in the development of FLF resulting from HBV or HDV infection. FLF is more common in chronic carriers of HBV after recent withdrawal of antineoplastic or immunosuppressive drugs and in those superinfected with HDV. The size of the viral inoculum is also important. There is some debate regarding the importance of infection with HBeAg-deficient mutants. [24], [25] HBV is not generally considered to be cytopathic, and with the early loss of HBsAg, it is generally accepted that FLF related to HBV is associated with a vigorous host immune response toward the virus.

Hepatitis A virus (HAV) and hepatitis E virus (HEV) are both RNA viruses that are enterically transmitted and can cause occasional epidemics. HAV is distributed throughout the world and has a similar relative frequency in causing FLF in the United States compared with HBV. Acute hepatitis A causes FLF in up to 0.4% of symptomatic patients. [26] Approximately 100 deaths per year occur in the United States from HAV-associated FLF, and it is more common in patients who are infected when they are more than 40 years old and in patients with chronic liver disease, especially HCV infection. [27], [28] Detection of IgM antibody to HAV is diagnostic. The prognosis of FLF caused by HAV infection is relatively good. [3] HEV has a more restricted distribution compared with HAV. HEV infection is endemic in Asia, the Middle East, and North Africa. Cases of FLF associated with HEV infection reported from the developed countries of North America and Europe are usually in patients who have traveled to endemic areas within the previous 1 to 2 months. Rarely are cases reported in patients without a history of travel to endemic areas. Mortality from HEV infection is similar to that from HAV infection. However, the risk of FLF is particularly increased in pregnant women, especially those in the third trimester. [29] Mother-to-fetus intrauterine transmission has been reported to occur, and severe liver necrosis may affect the infant. [30] The finding of IgM anti-HEV antibody in serum is diagnostic of acute HEV infection. The pathogenic mechanisms leading to FLF in patients with HAV or HEV infection are poorly understood. Both viral and host immunologic factors are likely to be important.

The risk of developing FLF after infection with hepatitis C virus (HCV) is not clear. The occurrence of HCV RNA in patients with FLF is related to the geographic area, 45% in Taiwan and 50% in Japan, but 0% in France, the United Kingdom, and the United States. [31] It may be difficult to exclude an acute exacerbation of a hitherto unrecognized chronic hepatitis C infection. FLF has been reported in chronic carriers of HCV after coinfection or superinfection with HBV or after withdrawal of chemotherapy.

Several other viruses have been reported to cause FLF, including herpes simplex virus, varicella zoster, cytomegalovirus, Epstein-Barr viruses, human herpes virus 6, human parvovirus B19, and the RNA viruses responsible for viral hemorrhagic fevers. Severe liver injury and FLF may accompany adenoviral infection in immunocompromised persons and are major obstacles to the use of these viruses as vectors for the delivery of gene therapy.

Large proportions of patients in most reported series of FLF have no identifiable cause for developing FLF. Such patients are classified as having an indeterminate cause. However, many have a viral prodrome, and the term *non-A, non-B hepatitis* became popular. With the further recognition of HCV and HEV, such patients are more correctly now described as contracting *non- A–E hepatitis*. Intensive investigation over the years has led to the identification of several potential candidate viruses. The agent designated GBV-C or hepatitis G can be transmitted by blood transfusion, but it is not associated with FLF or any other form of liver disease. [32] The TT virus was characterized more recently from a patient with posttransfusion hepatitis. This DNA virus can infect the liver, but the degree of infection is not related to hepatic histological activity, and evidence of replication within the liver is lacking. Early studies suggested that TT virus may be associated with non-A–E FLF, but

subsequent studies did not confirm its pathogenic role. [33] Other potential candidates as causal agents in non-A–E hepatitis include SANBAN and SEN-V viruses; further studies of these agents are eagerly awaited.

Idiosyncratic Drug Reactions

Although severe hepatotoxicity is rare, many drugs have been reported to cause liver injury and occasionally FLF. Drug reactions as a cause of FLF vary in relative frequency from 2% in the United Kingdom to 16% to 17% in France and the United States. [20] A high index of suspicion is often required to make the association between drug ingestion and the development of FLF. However, certain drugs are more commonly associated with both hepatotoxicity and FLF. Antituberculosis therapy (isoniazid and especially pyrazinamide) is an important cause of FLF and may potentiate acetaminophen toxicity. [34] Halothane and its derivatives can induce immune-mediated liver injury and can potentially lead to FLF. Halothane hepatitis occurs up to 28 days after exposure and is more common in female patients and those who have received multiple halothane anesthetic administrations. [35] This condition has become a much less frequent cause with the advent of newer anesthetic agents. Sulfonamides, for example sulfasalazine, can cause FLF, which may recur with exposure to other sulfonamide-containing drugs. Nonsteroidal antiinflammatory drugs such as piroxicam and bromfenac have been reported to cause FLF. Other drugs implicated in causing FLF include fialuridine and 2,3-dideoxyinosine. These drugs may induce mitochondrial dysfunction and hepatotoxicity. Idiosyncratic drug reactions are associated with a poor prognosis. [3]

Reye syndrome is characterized by rapidly progressive hepatic encephalopathy associated with cerebral edema and hepatic steatosis. This syndrome is most common in children, but a few adult cases have been reported. [36] There is a strong association with the use of salicylates, and a reduction in consumption of these drugs has led to a fall in incidence of this disorder. [37] Mitochondrial toxicity has been implicated in the pathogenesis of Reye syndrome. The mortality rates are high in patients who develop coma, but early recognition of the syndrome and specialist management have led to a reduction in death rates. [38]

Several investigators have reported hepatotoxicity and FLF after ingestion of herbal remedies, available without prescription. The use of such agents is commonplace, and patients may not necessarily volunteer to their physicians that they have been consuming these remedies. Herbal remedies often contain multiple compounds, which do not remain constant, and therefore it may be difficult to isolate the responsible component. Wall germander is hepatotoxic, and FLF requiring emergency liver transplantation has been reported in some cases. [39] Pennyroyal oil causes dose-dependent acute hepatitis and occasionally FLF, which may be successfully treated with *N*-acetylcysteine. [40] Many other herbal remedies have been associated with acute hepatitis or FLF. FLF has also been reported after occupational or accidental ingestion of several industrial solvents such as carbon tetrachloride, trichloroethylene, and 2-nitropropane. Ingestion of food contaminated with aflatoxin and acute poisoning with yellow phosphorus can also lead to FLF.

Illicit drugs may cause severe hepatotoxicity and FLF. Several cases have been reported after ingestion of cocaine or ecstasy (3,4-methylenedioxymetamphetamine). FLF after ecstasy ingestion can occur either with or without an associated hyperpyrexia syndrome (rarely, exertional heat stroke can lead to FLF). The case fatality rate is high, and some survivors after emergency liver transplantation have been reported. [41]

Amanita mushroom consumption has been reported as a cause of FLF from continental Europe and the United States. About three medium-sized mushrooms can deliver a lethal dose of amanitoxins. FLF occurs late in the clinical course. Severe vomiting and diarrhea lasting 1 to 4 days follow an initial asymptomatic phase of up to 12 hours. After recovery from these symptoms, the liver injury becomes apparent, with massive elevation of the serum aminotransferases and the development of jaundice, coagulopathy, and hepatic encephalopathy. In the largest series published, the mortality rate was 22%, and it was especially high in patients with markedly prolonged prothrombin times. Emergency liver transplantation can be successful, but prognostication in such patients is exceedingly difficult.

Other Causes

Obstruction to the hepatic veins as occurs in Budd-Chiari syndrome can rarely lead to FLF. Abdominal pain and the early development of ascites (which is exudative in the early stages) are characteristic of Budd-Chiari syndrome, but the absence of these symptoms and signs does not exclude the diagnosis. Hepatic venous thrombosis is more common in female patients. [42] There is a high frequency of an associated coagulation abnormality, for example, lupus anticoagulant or a primary myeloproliferative disorder. Ultrasound, computed tomography, and hepatic venography are helpful in confirming the diagnosis. Early intervention with emergency mesocaval or mesoatrial shunt, transjugular intrahepatic portosystemic stent shunt placement, or emergency liver transplantation, followed by lifelong anticoagulation may be successful. Pyrrolizidine alkaloids present in some herbal remedies or teas can induce venoocclusive disease of the liver and can present with FLF. Antineoplastic chemotherapy, radiation, and graft versus host disease may result in venoocclusive disease.

Rarely, patients with Wilson disease present with FLF. Such a presentation may occur in patients newly diagnosed with Wilson disease or in those who have been taking D-penicillamine treatment for several years after interruption of therapy. Examination of the genetic variants occurring in this condition has failed to identify a specific genetic mutation in patients with FLF in Wilson disease. Coombs-negative hemolytic anemia accompanies the release of copper from damaged hepatocytes into the systemic circulation and contributes to the very high bilirubin levels seen in these patients. Occasionally, the serum transaminases are normal, but more often they are only modestly increased. Kayser-Fleischer rings and diminished serum ceruloplasmin may be absent in 50% and 10% of patients, respectively. The plasma copper (especially the free copper) and urinary copper levels are elevated, and the 24-hour urinary copper excretion is increased. [43] The prognosis of FLF secondary to Wilson disease is poor, and liver transplantation is often required. [3] A prognostic index derived from the serum aspartate aminotransferase, bilirubin, and prothrombin time may indicate those with a fatal outcome, who would be best treated with liver transplantation, compared with those who could survive with D-penicillamine treatment. [44]

FLF is a rare finding in patients with hepatic metastasis. Hepatomegaly and ascites are almost always found, and most commonly the primary tumor is breast carcinoma. Malignant melanoma and nasopharangeal carcinomas have also been reported to cause FLF. Infiltration of the liver by leukemia, Hodgkin's disease, or non-Hodgkin's lymphoma may present with FLF. [45] Hepatomegaly is a prominent feature, and marked increases in circulating lactate concentrations have been reported. Most often, the patient has a previous history of the tumor, and very rarely it is the first manifestation of the disease. In patients with acute leukemia or lymphoma, intensive chemotherapy may lead to regression of the FLF.

Occasionally, pregnancy may be associated with FLF. The two main syndromes leading to this clinical picture are acute fatty liver of pregnancy and hemolysis with elevated liver enzymes and low platelet (HELLP) syndrome. [46] Such syndromes are often associated with clinical features of preeclampsia including hypertension, peripheral edema, and proteinuria, and early symptoms of the liver disease include polydipsia, nausea, and vomiting. The delivery of the child results in the regression of the liver disease, although severe bleeding may occur after delivery.

In a small number of patients, hypoxic liver cell necrosis leads to FLF. [47] This is associated with a high mortality rate because the underlying precipitating cause is most commonly a cardiac arrhythmia or myocardial infarction. Other reported causes include pulmonary emboli and acute cardiac tamponade. Rarely, hypoxic liver cell necrosis follows severe and profound hypoxemia.

MANAGEMENT

The management of patients with FLF is mainly supportive, that is, avoiding complications and correcting abnormalities while allowing the greatest potential for recovery of liver function to take place. The only definitive treatment that alters the natural history of the disease is liver transplantation, as discussed later. *N*-acetylcysteine, used in the setting other than as an antidote to acetaminophen poisoning, may fall into the category of definitive (nonsupportive) therapy, but its effect is small and controversial (see later). Artificial liver support systems currently are of unproven benefit but clearly represent the most exciting treatment option for the future.

Early (Pre-ICU) Management

Cardiovascular Manifestations FLF is characterized by hyperdynamic circulation with tachycardia, hypotension, increased cardiac output, and low peripheral vascular resistance. This clinical picture resembles that seen in sepsis, and separation of these two syndromes may be difficult; the two often coexist. Use of inotropes to maintain the mean arterial pressure around 80 mm Hg or higher is generally thought desirable to maintain tissue, especially renal, perfusion, although some patients will maintain reasonable cardiovascular stability with judicious volume support alone. Traditionally, inotropes are reserved for use in the ICU and are discussed later. Early on, if adequate urine output is maintained, this can be used as a good clinical indicator of satisfactory tissue perfusion. However, because renal failure is common in FLF, particularly that caused by acetaminophen poisoning, poor urine output cannot be taken as indicating renal hypoperfusion. Placement of a line to monitor the central venous pressure is important to ensure adequate filling of the circulation. Peripheral vasoconstriction, which may be present earlier in the course of the disease, is usually a good indicator of volume requirement at this stage. Because the prothrombin time is such a useful marker of hepatic function, the administration of fresh frozen plasma before inserting lines is generally avoided. The internal jugular vein is usually the best site for cannulation because compression can be applied if bleeding occurs. Low-dose dopamine was shown to be ineffective in maintaining renal function in critically ill patients in a reasonably large randomized study, [48] and there is no evidence that it is beneficial in maintaining renal function in FLF. Terlipressin, which can be given by bolus

injection rather than by infusion, may be valuable as an inotrope, but experience is limited.

Metabolic Effects The most important metabolic abnormality to anticipate is hypoglycemia. Ten percent dextrose should be given through the central line to prevent this condition, and testing of blood glucose should be carried out every 2 hours, especially if the patient becomes drowsy. Acidosis is common, particularly in patients with acetaminophen poisoning, and it has prognostic significance. A pH of 7.3 or less in patients who have been fully resuscitated from a cardiovascular perspective is associated with a 90% mortality in acetaminophen poisoning and is one of the major factors in the King's College criteria for poor prognosis (Table 48-2). [3] Hypophosphatemia is common, but correction of this condition should be cautious in renal failure because levels may rise rapidly. Hypokalemia accompanying respiratory alkalosis may be marked and should be corrected.

Acetaminophen Poisoning
Arterial pH <7.3 (H^+ >50 nmol.l^{-1})
OR
Three of the following:
Prothrombin time >100 s
Serum creatinine >3.41 mg/L
Encephalopathy grade III/IV
Causes Other than Acetaminophen Poisoning
Prothrombin time >100sec
OR
Three of the following:
Unfavorable cause (e.g., drugs, non-A, non-B) jaundice for >7 d
before encephalopathy age <10 or >40 y
Prothrombin time >50 s
Serum bilirubin >300μmol/L

TABLE 48-2 Indicators of Poor Prognosis in Fulminant Hepatic Failure

Renal Effects Hepatorenal syndrome is common in FLF, particularly in acetaminophen poisoning, because acetaminophen is directly toxic to the kidney. Indeed, a few patients will develop renal failure alone after acetaminophen poisoning. Urine output should be closely monitored in patients with acute liver injury, which requires urinary catheterization. Renal function must be monitored by serum creatinine, rather than by urea, because urea is dependent on hepatic synthesis and values may be misleadingly low. Creatinine should be measured daily, and twice daily once it is more than 1.70 mg/L, because it may increase rapidly. A serum creatinine level of more than 3.41 μg/L is a marker of poor prognosis in acetaminophen-induced FLF (see Table 48-2).

Hematologic Manifestations One of the best markers of acute liver injury is the prothrombin time, and this should be measured daily and more frequently once it is longer than 50 seconds. Because the prothrombin time is a valuable marker of prognosis, clotting factors should not be given prophylactically but only if there is bleeding. If clotting factors have been given, the length of time it takes for the prothrombin time to revert to its untreated level is unpredictable. A prothrombin time longer than 50 seconds in non–acetaminophen-induced FLF and a prothrombin time more than 100 seconds in acetaminophen-induced FLF are indices used in the King's College criteria for poor prognosis. [3] An increase in the prothrombin time between day 3 and day 4 after the ingestion and a single prothrombin time greater than 180 seconds may also have some prognostic significance. These prognostic markers may be influenced by other drugs that induce acidosis (e.g., aspirin) and by not taking the acetaminophen as a single overdose. Staggered overdoses or paracetamol taken during a therapeutic misadventure will not conform to these prognostic indicators, and in these patients it is notoriously difficult to establish an accurate prognosis and to make the decision confidently to proceed to liver transplantation. To standardize prothrombin time reports among patients with FLF, the international normalized ratio (INR) has been advocated. However, others have suggested that the INR is not valid in patients with liver disease, especially those with FLF, and it is best to express the prothrombin time as an activity percentage. Gastrointestinal bleeding is reduced by the prophylactic use of H_2-antagonists or sucralfate. [49], [50] Thrombocytopenia is common in FLF, and the platelet count may decrease to less than 30 × 10^9/L. Platelets should be given only in patients with active bleeding because clotting factors in the platelet replacement packs will affect the prothrombin time.

Infection Sepsis is common in FLF, and the prophylactic administration of antibiotics such as ciprofloxacillin is recommended once encephalopathy develops. Blood cultures should be taken daily, and bacteremia should be treated appropriately and early. This is important, because sepsis is a contraindication to liver transplantation.

Neurological Features Encephalopathy is a cardinal feature of FLF and should be actively assessed and monitored because it may be subtle in the early stages and may progress rapidly. Traditionally, encephalopathy is divided into 4 grades: slow mentation (grade 1), agitation (grade 2), permanent somnolence (grade 3), and coma (grade 4). This grading is important because grades 3 and 4 have prognostic significance (see Table 48-2). Patients are generally transferred to ICU once grade 3 encephalopathy develops because assisted ventilation is often required. Raised intracranial pressure (ICP) and cerebral edema generally are complications of patients with grade 4 encephalopathy. The disadvantage of this classification of encephalopathy is that it is somewhat crude and not generally known by personnel in other medical disciplines who may be involved in the management of these patients, such as intensive care specialists and anesthesiologists. The Glasgow coma scale may be useful in discussing the neurological state of patients and should probably be more widely used.

***N*-Acetylcysteine** This agent has been known for years to be an effective antidote for acetaminophen poisoning if it is administered within 18 hours of the overdose. In the United Kingdom, it is generally given intravenously. Oral administration of this drug or methionine will reduce the risk of liver injury, but vomiting may complicate drug delivery. However, as well as the action of this drug as an antidote, it was reported by the King's College group that administration of this agent after this period improved outcome for both acetaminophen-induced and non–acetaminophen-induced FLF. [51] The mechanism of this possible benefit is unclear. Improved tissue oxygen delivery has been proposed as one mechanism, but this was based on a standard methodology that is now questioned, and further work demonstrated that oxygen consumption is unchanged by *N*-acetylcysteine administration. [52], [53] There is no universal agreement that late (>18 hours after the overdose) administration of *N*-acetylcysteine is beneficial in FLF, particularly in the non-acetaminophen group of patients. The drug is generally well tolerated, however, apart from occasional urticarial and allergic reactions, and many clinicians give the drug in the absence of alternative therapy. If given, it is generally administered until the patient is clearly recovering or dies.

Late (ICU) Management

Cardiovascular Manifestations By the time the patient with FLF requires to be transferred to ICU, usually because of deteriorating encephalopathy or for renal replacement therapy, hypotension is generally apparent, with a systolic blood pressure lower than 100 mm Hg and a mean arterial pressure lower than 70 mm Hg. Norepinephrine is the most commonly used inotrope, started at a dose of 0.05 $\mu g.kg^{-1}.min^{-1}$ and increasing to achieve a mean arterial blood pressure higher than 80 mm Hg. Adequate blood pressure is important to maintain satisfactory renal perfusion in these patients, who are frequently oliguric or anuric, and to maintain an adequate cerebral perfusion pressure. Peripheral perfusion may be compromised by norepinephrine administration, especially in the relatively hypovolemic patient. The insertion of a pulmonary artery catheter to monitor right-sided and left-sided cardiac pressures is valuable for optimizing fluid replacement.

Ventilation Pulmonary problems in FLF may include aspiration pneumonia, noncardiogenic pulmonary edema, and the adult respiratory distress syndrome as part of multiorgan failure. Artificial ventilation is generally initiated because of deepening encephalopathy, but oxygen requirements commonly increase because of an increased metabolic rate secondary to the hyperdynamic state and cytokine release.

Intracranial Pressure Raised ICP leading to cerebral edema is a common cause of death in FLF. Recognition of raised ICP by such clinical features as pupillary dilation, elevated blood pressure, bradycardia, and opisthotonos are insensitive and delayed features, and many clinicians believe that insertion of an ICP monitor is indicated despite the recognized risks of insertion in a patient with coagulopathy and thrombocytopenia. Most clinicians prefer to treat deranged coagulation before insertion of an ICP transducer, and it is desirable to wait until a prothrombin time of more than 100 seconds in acetaminophen poisoning and more than 50 seconds in non-acetaminophen cases is exceeded, so clotting factor replacement does not interfere with critical decision-making steps, particularly listing for liver transplantation. Once these prothrombin levels have been exceeded, clotting factor and platelet replacement therapy may allow safer insertion of an ICP monitor. With such monitoring, the management of raised ICP is greatly facilitated. An ICP greater than 20 mm Hg for more than 10 minutes is associated with cerebral edema and should be treated. The choice of intracranial monitoring system is debatable; epidural, subdural, and parenchymal systems are available. All are associated with some risk of intracranial bleeding. The risk of hemorrhage may be lower with epidural devices, but most centers use fiberoptic catheter systems placed subdurally for greater accuracy and ease of bedside insertion. Other noninvasive methods for detecting cerebral edema such as computed tomography are insensitive in this setting, only provide a snapshot at one moment in time, and require moving the patient from the ICU setting, which is undesirable and possibly dangerous. Management of the ICP is valuable, but the cerebral perfusion pressure is probably a more useful parameter. It is derived from the mean arterial pressure minus the ICP and ideally should be higher than 60 mm Hg. A value lower than 40 mm Hg is likely to lead to cerebral damage, and many clinicians believe that a cerebral perfusion pressure of less than 40 mm Hg for more than 60 minutes is a contraindication to liver transplantation. [54] A further useful measurement in monitoring the neurological status in these patients is to record cerebral oxygen consumption. This may be achieved by measurement of cerebral blood flow together with arterial and jugular bulb oxygen contents, but simply using a jugular bulb catheter to measure jugular venous oxygen saturation provides a reasonable guide to whether luxury perfusion or cerebral ischemia is associated with raised ICP and helps therapy to be appropriately targeted. Mannitol therapy has been shown to be effective in controlling ICP in this setting and is the treatment of choice. [55] However, repeated administration may be required, and efficacy reduces with time. Hyperosmolality may occur, and plasma osmolality should be measured. The response to mannitol administration should be monitored, and further treatments or second-line therapies should be used if necessary. Second-line therapy with thiopentone is less commonly used in patients with head injury, and evidence of efficacy in FLF is limited. More recently, work from our unit has shown that moderate hypothermia is effective in controlling ICP. [56] Animal data support this clinical observation, but more work is needed to establish whether this finding is translated into improved survival. A further important neurological complication of FLF is epileptic seizures. These may be masked by sedation and are easily overlooked. One study in which prophylactic phenytoin therapy was administered suggested that this was of benefit in reducing the prevalence of cerebral edema. [57] Continuous electroencephalographic monitoring for epileptiform activity should be routine.

Renal Effects Renal failure is common in FLF, and most patients will require renal replacement therapy at some time during their illness. Continuous venovenous hemofiltration is preferable to intermittent filtration or dialysis, which may lead to an increase in ICP. Because of coagulopathy, prostacyclin is often used in preference to heparin as the anticoagulant in the hemofiltration system, although in some cases no anticoagulant is necessary. Filtration against bicarbonate buffer may be used in place of lactate buffer, but with continuous hemofiltration, extrahepatic lactate clearance is sufficient to avoid a significant increase in plasma lactate. Correction of acidosis, rather than lactate decrease, is the criterion of effectiveness. After recovery of hepatic unction or liver transplantation, renal support may need to be continued for weeks as the hepatorenal failure or acute tubular necrosis recovers. The eventual renal recovery is almost invariably complete.

Infection By the time patients with FLF are admitted to the ICU, prophylactic antibiotics should have been started. The most common infections are with gut- or skin-derived organisms, and antibiotics should be broad-spectrum agents such as cefotaxime and amoxicillin-clavulinic acid combinations. Daily blood cultures and culture of urine and sputum should be performed. Sepsis may be difficult to identify because of the similarity between the cardiovascular changes of FLF and sepsis, and a low threshold of suspicion must be maintained. Suspected infection in patients receiving antibiotics, especially after 5 days in an ICU, should lead to the empiric introduction of antifungal therapy. Many clinicians administer prophylactic fluconazole from the time of admission to the ICU.

Liver Transplantation It is generally in ICU that liver function deteriorates to the level at which prognosis can be determined (see Table 48-2). Patients in whom the poor prognosis criteria are not met will generally survive with full supportive therapy, but 80% to 90% of those who reach these criteria will die without liver transplantation. As well as the King's College and the coagulation criteria discussed earlier, Apache II and III scores have been assessed to identify patients with acetaminophen-induced acute liver failure with a poor prognosis. An Apache II score greater than 15 soon after admission may be useful in the early identification of patients with a poor prognosis and has an advantage over the King's College criteria in that intensive care specialists are familiar with its use. Overall, however, the predictive power of the two scoring systems is similar. [58] These poor prognosis scores are generally taken as the transplantation criteria as long as other medical or psychiatric contraindications are not present. Because patients may deteriorate very rapidly once the criteria are met, listing for liver transplantation should not be delayed. Other important testing required for urgent listing for liver transplantation such as human immunodeficiency virus status should therefore be carried out before the criteria are met to avoid unnecessary delay. Matching between donor and recipient with regard to size is not so important as before because of the ability to reduce graft size. However, transplantation across blood groups, with the exception of blood group AB recipients, has generally been abandoned because of poor results. Overall, the results of liver transplantation for FLF are not as good as for chronic liver disease. Most centers achieve 12-month survival figures of around 60% compared with 80% for patients who undergo liver transplantation for cirrhosis. One of the main reasons for this probably is that patients go to the operating room while they are critically ill, and anything that can be done to stabilize their clinical condition while they wait for a donor to become available is likely to affect results beneficially. Interventions aimed at improving the patient's condition, albeit temporarily, have been promoted as a bridge to transplantation. These include high-volume plasmapheresis, moderate hypothermia, hepatectomy, and use of a molecular adsorbent recirculating system. This last intervention is currently under study in numerous centers, and benefit has been reported in both acute and chronic liver failure. However, results of randomized studies will be needed to evaluate its role properly. The most promising therapeutic approach as a bridge to transplantation is probably the use of bioartificial liver support devices.

Liver Support Devices Treatment strategies using liver support devices designed to help patients with FLF have either been those aimed at detoxifying the blood or those aimed at providing liver-derived factors. The former includes hemofiltration, charcoal hemoperfusion, and plasmapheresis. None of these has proven to be of value in clinical practice. The latter measures require liver cells, and although extracorporeal hepatic perfusion has been attempted, incorporation of hepatocytes into a filtering system that contains a framework to support the cells (a bioreactor) physically as a bioartificial liver support device is the most attractive. This approach still requires much research and must be subjected to clinical trials before it can be advocated in clinical practice. At present, the choice of hepatocyte represents the major problem. Ideally, the liver cell should be human, highly differentiated, and readily available in large numbers. The critical functions necessary for a hepatocyte to work well in a bioartificial liver remain to be discovered. If bioartificial liver support devices do prove to be effective in clinical practice, it is hoped that, as well as being a bridge to transplantation, they may allow some patients to recover sufficient native liver function to survive without liver transplantation.

OVERVIEW

FLF is an uncommon medical emergency with many possible causes. Reliable criteria have been established that predict prognosis, and patients in the poor prognosis group are unlikely to survive without definitive intervention. At present, the only treatment shown to prolong survival is liver transplantation, but it is to be hoped that liver support devices and, in particular, bioartificial liver systems may in the near future prolong survival, both by allowing more patients to survive to liver transplantation or by bridging the period of acute liver failure until native liver function returns.

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CHAPTER 49

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SKIN LESIONS ASSOCIATED WITH GASTROINTESTINAL AND LIVER DISEASES

Many diseases can involve both the skin and gastrointestinal tract. Primary disorders of the gut such as inflammatory bowel disease can initially manifest in the skin, and primary skin disorders such as pemphigus vulgaris may present with mucosal lesions. It is important that specialists in both disciplines be aware of how cutaneous lesions may be reflective of gastrointestinal diseases. This chapter reviews the skin lesions that may be associated with specific gastrointestinal complaints or known pathological processes. If a dermatologic lesion or diagnosis is suspected, dermatologic consultation is suggested to assist in diagnosis and management.

INFLAMMATORY BOWEL DISEASE

Chronic nonspecific ulcerative colitis and Crohn's disease are associated with a host of immunologic abnormalities. Although the role of circulating immune complexes in producing extraintestinal manifestations has been debated (see Chapter 7, Chapter 83, and Chapter 129), it is not surprising that numerous reactive inflammatory vascular dermatoses occur in patients with these diseases.

Erythema: Generalized, Localized, and Annular

Reactive inflammatory vascular dermatoses comprise a spectrum of inflammatory blood vessel reactions that occur in association with internal disease. [1] These range from generalized or local erythema, (macular blanchable lesions characterized histologically by simple vasodilation and mild inflammation) to urticarial lesions, erythema multiforme, and vasculitis characterized by palpable purpuric lesions with histological leukocytoclasis and fibrinoid necrosis of blood vessel walls. Table 49-1 summarizes the various types of vessel-based inflammatory lesions that may occur in patients with gastrointestinal disease.

TABLE 49-1 Erythematous Vessel-Based Eruptions in Patients with Gastrointestinal Disease

Urticaria and Angioedema

Urticaria is a raised, peau d'orange cutaneous vascular reaction characterized by dermal edema and erythema in which, by definition, individual lesions resolve within 24 hours. [1] Similar lesions occurring in a deep dermal or subcutaneous location are called *angioedema.* Urticaria is a common condition, affecting up to 20% of people at least once in their lifetime, often in young adulthood. In patients with inflammatory bowel disease, [2] urticaria is usually drug induced or results from some cause other than bowel disease. Hereditary or acquired C1 esterase deficiency is associated more with angioedema than urticaria; abdominal pain caused by mucosal edema may occur. Chronic urticaria affects primarily middle-aged women and is characterized by the occurrence of daily hives (individual lesions still resolve within 24 hours) for longer than 6 weeks. Two specific conditions, urticarial vasculitis and a syndrome of urticarial lesions occurring in the setting of a serum sickness–like illness lasting longer than 24 hours, are not, by definition, true urticaria. A skin biopsy should be performed on urticarial lesions that last longer than 24 hours, to exclude vasculitis. The cause of chronic urticaria in an individual patient may be difficult to establish. Several practical approaches have been published. [3] Treatment of urticaria consists of an attempt to eliminate the cause and to relieve signs and symptoms with around-the-clock antihistamine therapy.

Erythema Multiforme

Erythema multiforme is an acute, self-limited, mucocutaneous syndrome characterized by targetoid cutaneous lesions that may be accompanied by serum sickness–like signs and symptoms (e.g., fever, arthralgias). [4] *Erythema multiforme minor* refers to a form of the syndrome in which patients have a finite number of acrally located typical target lesions and, in general, lack mucosal lesions and other symptoms. *Erythema multiforme major* represents a more severe disease, and it is termed *Stevens-Johnson syndrome* if the patient has significant mucosal involvement. [4] Patients with *toxic epidermal necrolysis,* which may overlap with severe Stevens-Johnson syndrome, have a burnlike appearance. Toxic epidermal necrolysis is usually drug induced and may be associated with a 30% rate of mortality.

Well-documented causes of acute erythema multiforme include drugs (e.g., sulfonamides, phenytoin, penicillin), herpes simplex, *Mycoplasma* infections, and other infections (e.g., tuberculosis), as well as a host of underlying diseases. [4] Typical erythema multiforme has been described in patients with inflammatory bowel disease. [2] Complications of erythema multiforme major may affect the gastrointestinal tract and may be severe in toxic epidermal necrolysis. Esophagitis and esophageal stricture have been reported. There is considerable evidence that most cases of recurrent erythema multiforme minor are induced by recurrent herpes simplex infections. [4]

Therapy for erythema multiforme has not been assessed in a double-blind trial. Systemic corticosteroids are often prescribed for adults with erythema multiforme major in whom infection has been excluded. Ophthalmologic consultation for eye involvement and aggressive topical care are required to prevent secondary infection.

Erythema Nodosum

Erythema nodosum is characterized by the occurrence of tender, nonulcerative nodules, most often on the legs, resulting from inflammation in the subcutaneous fat (Fig. 49-1; see Color Fig. 49-1). Usually, these nodules last from 3 to 6 weeks and affect the anterior lower legs in a symmetric distribution. Patients may have associated signs and symptoms, including fever, malaise, arthritis, and arthralgias. [5]

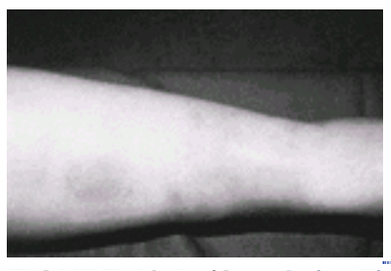

FIGURE 49-1. (See Color Fig. 49-1.) The tender nodules of erythema nodosum occurred in association with ulcerative colitis.

Erythema nodosum has been well established as an extraintestinal manifestation of ulcerative colitis, Crohn's disease, and infectious colitis, such as that caused by *Yersinia enterocolitica.* [1] The incidence of erythema nodosum in patients with ulcerative colitis was 7% in one large series, but the incidence in Crohn's disease is lower. [5] Other well-documented conditions associated with erythema nodosum include drugs (e.g., estrogen, sulfonamides), infections (e.g., by bacteria, viruses, fungi, chlamydia, acid-fast bacteria, spirochetes), malignant diseases, and other diseases, including Behçet disease and sarcoidosis. [5] An immune complex–mediated mechanism affecting vessels in the septal panniculus has been proposed, although the exact pathogenesis of erythema nodosum remains unknown.

Treatment of erythema nodosum in patients with inflammatory bowel disease is directed at the underlying disease. Nonsteroidal antiinflammatory drugs or even acetaminophen may alleviate the systemic symptoms.

Vasculitis

The term *leukocytoclastic vasculitis* has been used to refer to a form of vasculitis affecting postcapillary venules with the histopathological findings of endothelial swelling, neutrophilic invasion of blood vessel walls, leukocytoclasis (karyorrhexis of nuclei of neutrophils), extravasation of erythrocytes, and fibrinoid necrosis of blood vessel walls. [6] It presents as palpable purpura and may go by other names such as small vessel venulitis, Henoch-Schönlein purpura (which is actually a subgroup with IgA-containing immune complexes), allergic angiitis, and hypersensitivity vasculitis. Larger vessel involvement characterizes polyarteritis nodosa, granulomatous vasculitis (e.g., Wegener syndrome, Churg-Strauss syndrome), and giant cell arteritis.

Small vessel vasculitis has been proposed as a cutaneous model for circulating immune complex–mediated vessel damage. [6] The central and peripheral nervous system, synovium, pleura, pericardium, gastrointestinal tract, and kidney may act as filters for complexes with the similar end result of necrotizing lesions. [6]

From 39% to 61% of patients with necrotizing venulitis do not have a precipitating cause that can be identified. Drugs, infections (e.g., hepatitis B, streptococcal infection), and diseases associated with immune complexes (e.g., collagen vascular diseases, cancers, chronic active hepatitis) account for most of the identified associations. Crohn's disease [7] and ulcerative colitis [8] have both been clearly described in association with necrotizing venulitis and occasionally with granulomatous vasculitis. [9]

Prognosis in all forms of vasculitis depends on the internal organ involvement. Treatment should be directed at the underlying disease. Severe vasculitis may require acute therapy with plasmapheresis or high-dose therapy with systemic corticosteroids, or both. Low-dose weekly methotrexate, azathioprine, or, for larger vessel vasculitis, cyclophosphamide may be important adjuvant therapies. Oral colchicine or sulfone drugs (e.g., dapsone) may be used for primarily cutaneous vasculitis. [6]

Erythema Elevatum Diutinum

Erythema elevatum diutinum is a chronic fibrosing dermatosis characterized by juxtaarticular, persistent, symmetric, firm, tender, red to yellow-brown papules and plaques on the extensor surfaces of extremities (Fig. 49-2; see Color Fig. 49-2). It is believed to be caused by immune complexes. Early histological features include leukocytoclastic vasculitis, subepidermal fibrin deposition, and dermal papillary edema. A storiform proliferation of dermal spindle cells with fibrosis and foci of neutrophils or neutrophilic vasculitis characterize older lesions. Besides inflammatory bowel disease and celiac disease, it has been associated with rheumatoid arthritis, pyoderma gangrenosum, diabetes mellitus, paraproteinemias, and dermatitis herpetiformis, as well as chronic infections including streptococcal, mycobacterial, and human immunodeficiency virus infections. [10] Oral dapsone therapy is the treatment of choice.

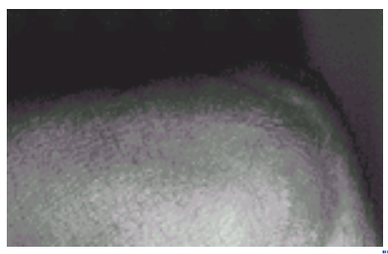

FIGURE 49-2. (See Color Fig. 49-2.) Typical lesions over joints in a patient with erythema elevatum diutinum.

Pyoderma Gangrenosum

Pyoderma gangrenosum is a misleading designation for an idiopathic ulcerating cutaneous disease. Theories range from a forme fruste of vasculitis to a cell-mediated reactive process. [6] Typically, fully evolved lesions have a dusky purple, undermined border and heal with cribriform scarring (Fig. 49-3; see Color Fig. 49-3). [6] Lesions may demonstrate the *pathergy phenomenon:* the rapid extension of the lesion after trauma such as from surgical debridement. Controversy exists about the exact histology of pyoderma gangrenosum, perhaps because older or partially treated lesions are often examined by biopsy. [6]

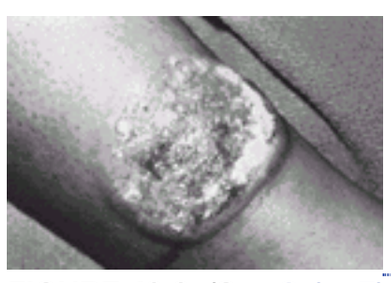

FIGURE 49-3. (See Color Fig. 49-3.) Pyoderma gangrenosum in a patient with Crohn's disease.

The diagnosis of pyoderma gangrenosum is a clinical diagnosis of exclusion. Bacterial, fungal, mycobacterial, and other infections, as well as vasculitis, squamous cell carcinoma, iododerma, bromoderma, and factitial disease, must be excluded histologically with appropriate special stains and cultures from biopsy specimens. More than 50% of patients with typical pyoderma gangrenosum do not have an associated disease. The association with inflammatory bowel disease, however, is one of the best known and should be considered and excluded in every patient with pyoderma gangrenosum. [11] Other associated conditions include chronic active and chronic persistent hepatitis, polyarthritis including rheumatoid arthritis, and myeloproliferative disorders.

Aggressive therapy is often required because the disease can spread rapidly and can predispose patients to sepsis or inanition and even death. Systemic corticosteroids in high doses, immunosuppressive agents, and sulfones are the mainstays of treatment. Anti–tumor necrosis factor antibody therapy may be useful.

Sweet Syndrome

Sweet syndrome, also known as acute febrile neutrophilic dermatosis, is characterized by the abrupt onset of tender cutaneous erythematous papules and plaques (Fig. 49-4; see Color Fig. 49-4). [15] Fever and peripheral blood neutrophilia also may occur. Disease onset frequently follows a nonspecific infection of the respiratory or gastrointestinal tract and is believed to represent a hypersensitivity reaction. Sweet syndrome has been reported in association with acute infections, myelodysplastic and other malignant diseases, and inflammatory bowel disease. [16] Histological specimens show a neutrophil vascular reaction with dermal inflammatory neutrophilic infiltrates and leukocytoclasia with no fibrinoid change in vessel walls.

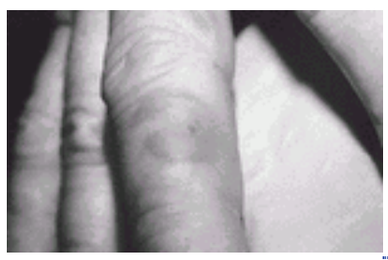

FIGURE 49-4. (See Color Fig. 49-4.) Tender erythematous papules and plaques on the upper extremities in a patient with Sweet syndrome.

Cutaneous lesions are typically located on the upper extremities, head, and neck. They may ulcerate, become purulent, and resemble lesions of pyoderma gangrenosum. Many dermatologists view Sweet syndrome and pyoderma gangrenosum as representing a continuum along a spectrum of disease, and considerable overlap between these two diseases occurs. [15] Like pyoderma gangrenosum, the disease responds to therapy with prednisone or other immunosuppressive agents, although often therapy with oral dapsone alone is adequate.

Oral Manifestations

As many as 30% of patients with ulcerative colitis have oral lesions, including aphthae and angular stomatitis, [2] whereas specific lesions (i.e., granulomas) of Crohn's disease affect the mouth and lips more rarely. Granulomatous lip lesions of Crohn's disease may be confused with Melkersson-Rosenthal syndrome, a condition in which granulomatous cheilitis occurs in association with facial nerve palsy. Granulomatous oral lesions may be nodular or show a cobblestone pattern. [12] *Aphthae* (canker sores) are nonspecific mucosal lesions that may be the most common oral lesions in patients with inflammatory bowel disease (Fig. 49-5; see Color Fig. 49-5). Simple aphthae occur in up to 25% of healthy persons. Patients with numerous, almost constant, oral aphthae (complex aphthosis) should be evaluated to exclude inflammatory bowel disease [13] and Behçet disease.

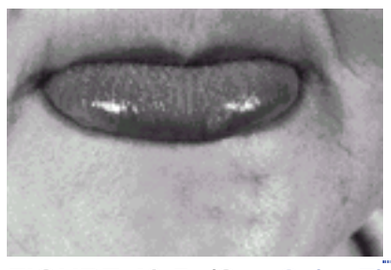

FIGURE 49-5. (See Color Fig. 49-5.) Aphthous ulcer involving the tongue in a patient with Behçet disease.

Angular cheilitis (perléche and angular stomatitis) may occur as a manifestation of nutritional deficiency or, more commonly, as a manifestation of oral candidiasis. *Pyostomatitis vegetans* is a papular eruption of the oral mucosa that produces a cobblestone and eroded appearance. This distinctive oral disease is associated with Crohn's disease or ulcerative colitis in most patients. [14]

Miscellaneous Skin Lesions

Although certain reactive dermatoses occur more commonly in patients with inflammatory bowel disease, many reports exist of single-case associations of a given dermatosis with Crohn's disease or ulcerative colitis. Metastatic Crohn's disease, which is the occurrence of noncaseating granulomas remote from the gastrointestinal tract in patients with Crohn's disease, has been well described at a host of cutaneous sites. [17] Lesions are usually ulcers, papules, or nodules. Pustular vasculitis, which can occur with inflammatory bowel bypass syndrome, is discussed later (see section " Bowel-Associated Dermatosis-Arthritis Syndrome"). Details of other less common associations can be sought in review articles, [2], [7] as well as in classical reports of large patient population surveys. [18], [19] Table 49-2 provides an overview.

Erythemas (including annular erythemas)
Urticaria
Erythema nodosum
Necrotizing venulitis
Larger vessel necrotizing vasculitis
Pustular vasculitis
Pyoderma gangrenosum
Oral lesions
Specific granulomas (Crohn's disease only)
Aphthosis
Angular cheilitis
Pyostomatitis vegetans
Metastatic Crohn's disease
Finger clubbing
Acquired acrodermatitis enteropathica (zinc deficiency)
Striae
Epidermolysis bullosa acquisita
Psoriasis
Exfoliative erythroderma
Vitiligo
Lichen nitidus
Lichen planus

TABLE 49-2 Some Cutaneous Associations of Crohn's Disease and Ulcerative Colitis

RHEUMATOLOGIC DISEASES

Rheumatoid Arthritis and Systemic Lupus Erythematosus

A review of skin manifestations of rheumatoid arthritis and systemic lupus erythematosus is beyond the scope of this review. Gastrointestinal involvement is usually related to vasculitis and is similar to that described earlier. Chapter 128 describes the gastrointestinal manifestations of rheumatic diseases.

Dermatomyositis

Dermatomyositis is an idiopathic inflammatory disorder that affects primarily skin and muscle and may manifest gastrointestinal symptoms (see Chapter 128). The cutaneous lesions are a violaceous poikiloderma (i.e., hyperpigmentation and hypopigmentation, telangiectasia, and epidermal atrophy) that occurs over the eyes (heliotrope sign), knuckles (Gottron sign), and extensor surfaces. There is a photosensitivity component to the eruption. Periungual telangiectasia and cuticular dystrophy are also typical. As many as 25% of patients with primary idiopathic polymyositis, and a smaller percentage of those with dermatomyositis, have dysphagia caused by involvement of striated muscles of the pharynx and esophagus. [20] Dermatomyositis or polymyositis may occur as a paraneoplastic syndrome in adults; therefore, the adult patient with dermatomyositis or polymyositis should be evaluated for occult malignant disease. Studies should include a complete history, physical examination, chest radiograph, and screening laboratory tests, as well as a breast and pelvic examination, Papanicolaou smear, and mammography in women and a prostate examination in men. [21] Because gastrointestinal tumors may be a cause, the rectum, colon, pancreas, and stomach may need evaluation as well.

Scleroderma

Scleroderma is a cutaneous or multisystem disease of unknown origin. A localized cutaneous form (morphea) is characterized by plaque, gyrate, generalized, or linear dermal sclerosis. The systemic form may be considered as a continuum of disease severity, the extremes of which are the milder CREST variant (calcinosis, Raynaud's phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia) and a more severe, progressive systemic sclerosis. [22] Most patients with systemic scleroderma have the CREST variant. Although patients with CREST can become critically ill with any of the varied manifestations of systemic scleroderma, they generally are middle-aged or older women who have a chronic course and die with, not of, their disease. Esophageal reflux should be treated to reduce esophageal fibrosis. Sclerosis occurs acrally and periorally and can affect the trunk. A salt-and-pepper postinflammatory pigment change is particularly noticeable over sclerotic areas in darker-skinned patients. Telangiectasias are described as being boxlike or matlike and occur on the hands, face, oral mucosa, and other sites. [23] The progressive systemic sclerosis type of systemic scleroderma occurs with an explosive onset. The dermal sclerosis is dramatic, but CREST features are often absent or are present to a lesser degree. The systemic and gastrointestinal features of scleroderma are discussed in Chapter 59, Chapter 73, and Chapter 128.

Behçet Disease

Behçet disease is a complex multisystem disease first described by and named after a Turkish dermatologist. [6] Because there is no pathognomonic laboratory test for the diagnosis, several sets of diagnostic criteria have been proposed. The O'Duffy criteria require that patients without inflammatory bowel disease or collagen vascular disease must have oral aphthosis (canker sores) plus at least two of the following: genital aphthae, synovitis, posterior uveitis, cutaneous pustular vasculitis, and meningoencephalitis. [24] The exclusion of patients with inflammatory bowel disease is crucial because patients with bowel disease may have recurrent aphthosis and (especially those with HLA-B27 antigen) are best considered as having the Reiter disease/HLA-B27 spectrum of diseases rather than Behçet disease. [25]

The oral lesions in Behçet disease are aphthae and can easily be distinguished from the psoriasiform oral lesions of Reiter disease. [25] Genital aphthae resemble oral aphthae (Fig. 49-6; see Color Fig. 49-6). Recurrent herpes simplex must be excluded by culture or by immunoperoxidase study of biopsy material. Cutaneous pustular vasculitis lesions show either leukocytoclastic vasculitis or a neutrophilic vascular reaction histologically. [26] Other lesions have been described that are similar to those seen in erythema nodosum, Sweet syndrome, or pyoderma gangrenosum.

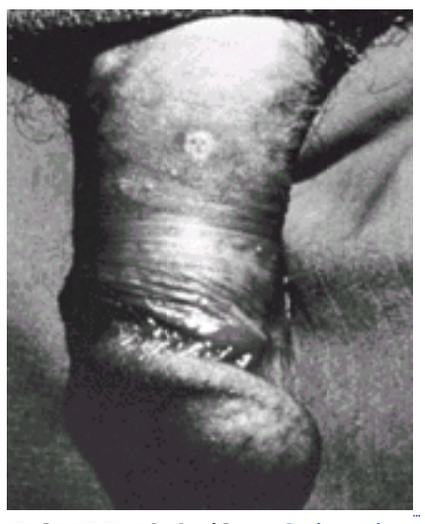

FIGURE 49-6. (See Color Fig. 49-6.) Early genital aphtha showing features of pustular vasculitis in a patient with Behçet disease.

Certain ocular changes have been described in these patients, although posterior uveitis (i.e., retinal vasculitis) is the most important manifestation. The synovitis produces an asymmetric, migratory, nonerosive oligoarthritis. Many neurological manifestations have been reported. Vascular involvement such as aneurysms, arterial occlusions, venous occlusions, and varices may occur, and they can prove fatal. The principal gastrointestinal manifestations are aphthae, which can occur at any gastrointestinal site. Changes similar to those in inflammatory bowel disease have been described, but, as has been discussed, these may occur in HLA-B27–positive patients with a Reiter-like illness and aphthae.

Bowel-Associated Dermatosis-Arthritis Syndrome

Approximately 20% of patients in one report developed a characteristic syndrome after jejunoileal bypass surgery [27] that included pustular vasculitic cutaneous lesions, arthritis, and serum sickness–like features. Clinically and histopathologically, the skin lesions resembled the pustular vasculitis lesions seen in Behçet disease. This operation has essentially been abandoned; however, we expanded the concept to include patients who have not had bypass surgery but who have either inflammatory bowel disease or the occurrence of a blind loop after Billroth II surgery. [28]

The cutaneous lesions occur in crops, with each episode lasting 1 to 2 weeks and occurring at intervals of 1 month to several months (Fig. 49-7; see Color Fig. 49-7). Lesions similar to those seen in erythema nodosum may occur. Fever, myalgias, chills, flulike symptoms, gastrointestinal upset, and arthralgias or nonerosive arthritis are often associated. Circulating immune complexes have been postulated to mediate this syndrome, [27] and serum enhancement of neutrophil migration may mediate the cutaneous lesions. [29]

FIGURE 49-7. (See Color Fig. 49-7.) Bowel-associated dermatosis-arthritis syndrome. Pustular vasculitis lesions occurred in a patient with a blind loop after Billroth II surgery.

The bowel bypass syndrome can be cured by restoration of normal bowel anatomy (see Chapter 79). Treatment of the underlying disease is necessary to control the syndrome in patients with inflammatory bowel disease. Systemic antibiotic therapy with metronidazole, tetracycline, or erythromycin has proved beneficial in controlling cutaneous and systemic manifestations of this syndrome. Systemic corticosteroid therapy and oral dapsone are also effective therapies. [6]

Amyloidosis

Amyloidosis is a group of syndromes characterized by the extracellular deposition of an abnormal protein with certain staining properties and electron microscope features. Primary amyloidosis, also called light-chain–related systemic amyloidosis, [30] often occurs in patients with multiple myeloma. Macroglossia, cutaneous papules, plaques, and nodules occur with pinch purpura (Fig. 49-8; see Color Fig. 49-8). Patients with primary amyloidosis have frequent gastrointestinal, cardiac, renal, and other multisystem involvement. Skin biopsy of a cutaneous lesion in primary amyloid is virtually diagnostic of the disease.

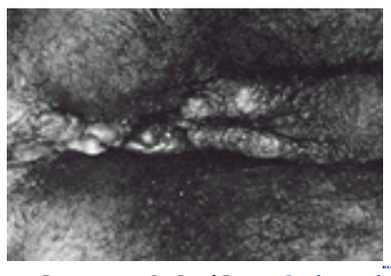

FIGURE 49-8. (See Color Fig. 49-8.) Amyloidosis. Note the perirectal amyloid nodules in this patient with multiple myeloma.

Patients with secondary (AA) amyloidosis do not have cutaneous involvement. [31] This pattern is associated with underlying disease, such as chronic inflammatory bowel disease, tuberculosis, leprosy, chronic osteomyelitis, or familial Mediterranean fever. Biopsy of normal skin reveals amyloid deposits in fewer than one third of patients, whereas small bowel or colonic mucosal biopsy samples are positive in more than 75% to 85% of patients.

SKIN DISEASE AND GASTROINTESTINAL BLEEDING

Vascular Abnormalities

Osler-Weber-Rendu Disease (Hereditary Hemorrhagic Telangiectasia) This autosomal dominant trait is characterized by telangiectasis, aneurysms, and arteriovenous malformations that can affect mucocutaneous areas as well as internal organs. [32] The mucocutaneous hallmark for the syndrome is 1- to 3-mm telangiectatic mats resembling those seen in patients with scleroderma (Fig. 49-9; see Color Fig. 49-9). These lesions can affect the lips, tongue, face, hands, chest, and feet [32] (see Chapter 130).

FIGURE 49-9. (See Color Fig. 49-9.) Typical telangiectasia of Osler-Weber-Rendu disease.

Blue Rubber Bleb Nevus Syndrome *Blue rubber bleb nevus syndrome* is a rare autosomal dominant condition with blue, subcutaneous, rubbery, compressible, sometimes painful nodular vascular malformations that are up to 10 cm in diameter. [33] These vascular malformations are not true hemangiomas. They occur in the small intestine or colon but also have been observed in the oropharynx, nasopharynx, esophagus, stomach, peritoneal cavity, mesentery, liver, lung, glans penis, eye, and central nervous system. [34] Gastrointestinal bleeding and intussusception may occur (see Chapter 130).

Kaposi Sarcoma Most publications since the early 1980s regarding *Kaposi sarcoma* have focused on the epidemic form of the disease, associated with the acquired immunodeficiency syndrome (AIDS) [35] (see Chapter 51). An endemic form has long been recognized in Africa, and the third variant is the classic type, which occurs primarily in persons of Mediterranean ancestry. The tumor in all groups is derived from proliferating endothelial cells. The cutaneous lesions vary from reddish-purple macules to large vascular tumors. In the slowly progressive classic form of the disease, lesions begin on the feet or lower legs of affected patients (who are typically elderly men) and extend proximally, becoming associated with severe peripheral edema. The most common site of internal involvement, but usually not a prominent feature, is the gastrointestinal tract. Sites affected, in order of decreasing frequency of involvement, are the small intestine, stomach, esophagus, and colon. [36] Extensive hemorrhage or partial bowel obstruction can occur. Other organs that may be affected include lymph nodes, larynx, spleen, liver, and lungs. Most patients with the classic form of Kaposi sarcoma die with, and not of, their disease. In contrast, the Kaposi sarcoma associated with AIDS is far more aggressive clinically (see Chapter 51).

Malignant Atrophic Papulosis (Degos Disease) *Malignant atrophic papulosis* (Degos disease) is a rare, idiopathic, usually lethal disease with characteristic lesions that affect the skin, gastrointestinal tract, and central nervous system. [37] The cutaneous lesions are discrete, painless papules with umbilicated porcelain-white centers surrounded by telangiectatic rims. Histologically, these lesions reveal an atrophic epidermis with a wedge-shaped dermal scar that is broader at the top than at the bottom. [38] Gastrointestinal involvement may lead to massive gastrointestinal bleeding and to death (see Chapter 130). Current evidence is against an immunopathogenesis in this disease. [38] There is no effective therapy, and visceral involvement may be fatal.

Diseases of Connective Tissue

Pseudoxanthoma Elasticum *Pseudoxanthoma elasticum* is a genetic disorder characterized by cutaneous and visceral manifestations that result from alterations to elastic fibers. Four genetic subtypes have been described, including both autosomal recessive and dominant inheritance patterns. [39] These types have a variety of ocular, cardiovascular, and gastrointestinal manifestations. The cutaneous lesions consist of yellow papules and plaques that resemble chicken skin (Fig. 49-10; see Color Fig. 49-10). These lesions begin early in life and may progress to affect the buccal mucosa, neck, axillae, and other flexures. Histologically, lesions show calcification of middermal elastic fibers. Angioid streaks are the characteristic retinal finding in patients affected with pseudoxanthoma elasticum but are not specific. The entire cardiovascular system is at risk for vascular calcification.

FIGURE 49-10. (See Color Fig. 49-10.) Pseudoxanthoma elasticum. Note the chicken skin–like appearance of the axillary skin.

Gastrointestinal involvement may present with recurrent upper or lower gastrointestinal hemorrhage (see Chapter 130). Endoscopy reveals the characteristic yellow cobblestone changes in the bowel. The pathogenesis remains unknown. The treatment is aimed at complications, but no therapy exists to alter the connective tissue abnormality. Prognosis depends on the subtype of disease.

Ehlers-Danlos Syndrome *Ehlers-Danlos syndrome* is a heterogenous group of genetically inherited syndromes characterized by defective collagen. More than 10 distinct subtypes exist, with various autosomal or X-linked inheritance patterns and with diverse abnormalities of collagen production. [40] The skin may be both hyperextensible and fragile. Wound healing is delayed and may produce cigarette burn–like scars or "fishmouth" scars as well as pseudotumors over joints. Purpura caused by easy bruising and hyperextensibility of joints are prominent features of Ehlers-Danlos syndrome. Visceral involvement is varied. Ocular and dental abnormalities are common. Large arteries may be affected by abnormal collagen, and rupture may occur, especially during pregnancy. Gastrointestinal perforation is prevalent in patients with type IV Ehlers-Danlos syndrome who have defective type III collagen and translucent skin with numerous ecchymoses. In addition to bowel perforation and gastrointestinal bleeding, these patients have a predisposition toward rupture of blood vessels; therefore, the prognosis is particularly grave [41] (see Chapter 130).

Cutis Laxa (Generalized Elastolysis) *Cutis laxa* is a heterogeneous group of autosomal and X-linked disorders characterized primarily by abnormalities of elastic fibers. [42] The skin appears to be too large for the body. It sags and has a wrinkled, aged appearance. Gastrointestinal diverticula and hernias occur. [42] The genitourinary tract also may be affected. Typical corkscrew-shaped blood vessels are seen on arteriography. Progressive pulmonary involvement may lead to chronic obstructive pulmonary disease, cor pulmonale, and premature death.

Neurofibromatosis (Von Recklinghausen Disease) *Neurofibromatosis* is inherited in an autosomal dominant fashion. Cutaneous features such as café au lait macules, axillary freckles, and cutaneous neurofibromas are hallmarks of this disease. Neurofibromas may occur throughout the gastrointestinal tract in 25% of patients. [43] The tongue, gallbladder, stomach, and jejunum are more commonly affected than the esophagus or colon. Ulceration, bleeding, volvulus, obstruction, perforation, and intussusception may occur. Malignant transformation of neurofibromas in the gut is not common, but it is well reported. Surgery is often required for symptomatic gastrointestinal involvement.

SYNDROMES ASSOCIATED WITH GASTROINTESTINAL POLYPS

Gardner Syndrome (Familial Adenomatous Polyposis)

In 1953, Gardner refined his description of a dominantly inherited triad of osteomas, epidermoid cysts, and colorectal polyposis that predisposed to colonic adenocarcinoma. [44] *Gardner syndrome* occurs as part of a spectrum that includes familial polyposis coli (no extracolonic lesions) and Turcot syndrome (features of Gardner syndrome plus central nervous system neoplasms) [45] (see Chapter 90).

Patients with Gardner syndrome develop multiple epidermoid cysts in their early teenage years. These cysts occur in a generalized distribution, especially on the scalp, face, and extremities [46] (Fig. 49-11; see Color Fig. 49-11). Desmoid tumors, which represent locally aggressive but nonmetastasizing tumors of fibrous tissue,

often affect the mesentery or the abdominal wall.

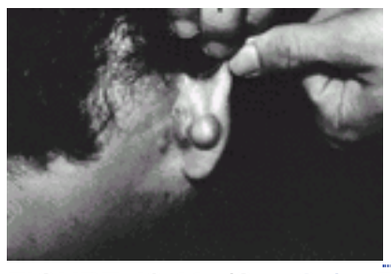

FIGURE 49-11. (See Color Fig. 49-11.) Typical epidermal inclusion cyst of Gardner syndrome. This patient had many other cystic nodules, particularly on the scalp.

Peutz-Jeghers Syndrome

Peutz and, later, Jeghers described a condition with an autosomal dominant inheritance pattern characterized by small intestinal hamartomatous polyps and hyperpigmented macules of the lips. [47] Although most polyps occur in the jejunum, they also occur in the ileum, stomach, duodenum, and colon (see Chapter 90). Polyps are present in childhood and may predispose to intussusception or to gastrointestinal bleeding.

The melanotic macules of Peutz-Jeghers syndrome begin in infancy. They affect the lips most commonly, but they also may affect the palms, soles, digits, periorbital skin, anus, and buccal mucosa. They histologically resemble ephelides (freckles). All except the buccal lesions fade with age. [48]

Cronkhite-Canada Syndrome

The *Cronkhite-Canada syndrome* is a sporadically occurring disorder of adults defined by the tetrad of cutaneous hyperpigmentation, alopecia, onychodystrophy, and intestinal polyposis. [49] The gastrointestinal signs and symptoms (e.g., diarrhea, abdominal pain, weight loss, anorexia) often precede the onset of the hair, nail, and cutaneous manifestations by several months (see Chapter 90).

The predominant cutaneous lesions are macular hyperpigmented macules that coalesce to form plaques that are distributed primarily on the upper extremities but may occur on most cutaneous sites. [50] Vitiligo occasionally may occur. Onycholysis, onychoschizia, or onychomadesis may affect all 20 nails. Widespread alopecia is nonscarring and may resemble alopecia areata. [50] The tongue may be fissured.

Cowden Disease

Cowden disease, also called multiple hamartoma syndrome, is inherited in an autosomal dominant fashion. Mucocutaneous markers for the syndrome include facial and oral mucosal papules that histologically represent tricholemmomas (benign hair-derived tumors). [51] The oral lesions produce a cobblestone appearance. A scrotal tongue is often associated. Acral keratoses resemble acrokeratosis verruciformis. [50] Patients also may have lipomas, hemangiomas, neuromas, and café au lait macules.

Gastrointestinal hamartomatous polyps occur throughout the gastrointestinal tract, with the majority affecting the rectum and sigmoid colon. The polyps are not premalignant; however, up to one third of patients with Cowden disease develop a malignant disease, usually of the breast or thyroid. [51]

Muir-Torre Syndrome

Muir and Torre each reported the association of sebaceous neoplasms and multiple visceral carcinomas. [52], [53] The inheritance pattern is autosomal dominant. Cutaneous markers for the syndrome include sebaceous neoplasms (hyperplasia, adenoma, epithelioma, and carcinoma), basal cell carcinomas, and keratoacanthomas. [50]

Adenomatous colonic polyps are not a universal feature of the syndrome, but one half or more of these patients do have polyps, and some develop colonic adenocarcinomas. Urogenital, hematologic, breast, and other malignant diseases may occur. [54] Patients should be screened frequently for colonic polyps, colonic carcinoma, and other malignant diseases.

Acrochordons and Colonic Polyps

In 1982, Klein and associates reported an increased incidence of *acrochordons* (skin tags) in patients with acromegaly observed for colonic polyps [55] and suggested that patients with more than six acrochordons could reasonably be targeted for evaluation for colonic polyps. [56] Several subsequent studies confirmed the correlation between acrochordons and colonic polyps, [57] although this was refuted in an autopsy study. [58] Most agree that the association is of no prognostic significance (see Chapter 91).

CUTANEOUS SIGNS OF GASTROINTESTINAL MALIGNANCY

Metastases and Direct Extension

Metastatic nodules of gastrointestinal adenocarcinoma are usually firm, pink, dermal to subcutaneous masses most likely to occur on the abdomen or pelvic area. The primary lesion has usually already been diagnosed by the time metastases develop. Biopsy of such a lesion helps to diagnose the primary site in fewer than 50% of patients. [59] An indurated nodule of the umbilicus (the Sister Mary Joseph nodule) may be the initial clinical presentation of an underlying (usually advanced) abdominal malignant disease, most commonly adenocarcinoma of the stomach or large bowel. Rarely, direct involvement of the lymphatics in the abdominal or thigh region draining a primary site can lead to extensive dermal infiltration, presenting as a sclerotic plaque. Occasionally, such a plaque can be quite erythematous and can mimic cellulitis (carcinoma erysipeloides).

Cutaneous Lesions that Should Raise Suspicion of Possible Gastrointestinal Malignancy

Extramammary Paget disease is considered to be a cutaneous adenocarcinoma, probably of apocrine gland origin. [60] It presents insidiously as an erythematous scaling or lichenified patch, often with surface erosion or crusting. Typically, the site has been treated with topical corticosteroids or antifungals without response, and the diagnosis is made by clinical suspicion and skin biopsy. If it occurs in the perianal area, it should raise suspicion of a possible underlying rectal or cloacogenic carcinoma [61] (see Chapter 91 and Chapter 92). The frequency of underlying visceral carcinoma has been reported at up to 86%, although a more recent review showed that only 25% of patients with perianal Paget's disease had a concurrent carcinoma of rectal origin. [61] Treatment of Paget's disease is usually by surgery (Mohs micrographic or wide excision) or radiation therapy, but the lesion frequently recurs.

Acanthosis nigricans presents as a smooth, thickened, and dark appearance to the skin in body fold areas, most often in the axilla and around the neck (Fig. 49-12; see Color Fig. 49-12). If extensive, it may lead to a striking appearance, with thickening of the palms and soles and oral lesions of hyperkeratosis. Acrochordons (or skin tags) also may occur in affected sites. This clinical presentation is most often seen without malignant disease in obese patients, in certain insulin-resistant diabetes syndromes, as a familial trait, and with certain medications.

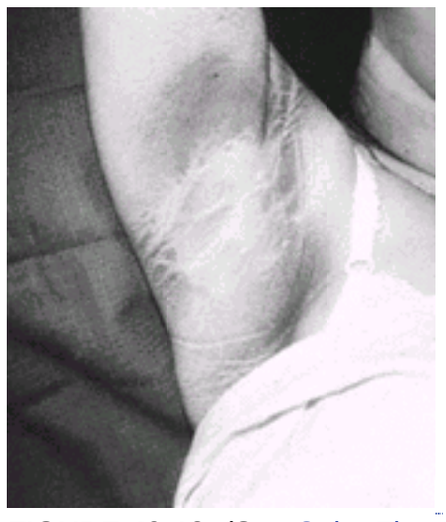

FIGURE 49-12. (See Color Fig. 49-12.) Acanthosis nigricans in a nonobese adult patient.

Suspicion for malignancy-associated acanthosis nigricans should arise if the condition occurs with recent onset in a nonobese patient who does not have diabetes. Biopsy of the skin is not helpful. Attention should be focused on the possibility of gastric adenocarcinoma, although other intra-abdominal and pelvic sites also should be evaluated for possible malignant disease. [62], [63] The malignant disease appears concomitantly in 61% of patients, but in almost 20%, the acanthosis nigricans may precede the diagnosable tumor by years. In these patients, frequent surveillance is necessary. In a patient known to have had an abdominal adenocarcinoma who then develops acanthosis nigricans, a vigorous search for recurrence or metastatic disease should be undertaken. The mechanism of malignant acanthosis nigricans is thought to be related to peptide growth factors released by the tumor. [64] Remission of the skin lesions occurs with removal of the tumor. There is no good dermatologic or cosmetic treatment for acanthosis nigricans.

Keratoderma of the palms and soles (tylosis) is a nonspecific presentation of diffuse or punctate yellow hyperkeratosis of the palmar and plantar surfaces. There are multiple causes, and it is only in patients with long-standing skin lesions and a family history of cancer that the question of gastrointestinal malignant disease should be raised. The strongest association has been with esophageal squamous cell carcinoma, and in the Howel-Evans syndrome it may occur in an autosomal dominant pattern, with up to 95% of family members developing esophageal carcinoma by 65 years of age [65] (see Chapter 62). In such kindreds, regular evaluations with barium swallow and esophagoscopy are warranted. [66] Other kindreds have been described with punctate keratodermas and carcinoma of the colon or pancreas. There are individual case reports of tylosis occurring in association with esophageal or bronchial carcinoma without a family history. In most of those cases, the palmar-plantar thickening was of recent onset without any other skin disease. The diagnosis of malignant disease was usually apparent at the time of initial presentation and evaluation.

Generalized erythroderma is a nonspecific cutaneous reaction pattern with a large differential diagnosis of possible causes, including primary skin diseases, adverse drug reaction, and internal malignant disease (especially lymphoma). The possibility of esophageal or other occult carcinoma should be raised in an erythrodermic patient with compatible symptoms. [68]

Hypertrichosis lanuginosa acquisita refers to the sudden development of diffuse, fine, downy hair on the face and trunk. It is extremely rare but is strongly associated with internal malignant disease. It has been seen most often with carcinoma of the lung or colon and, less commonly, of the pancreas and gallbladder. [68]

Patients with the *Paterson-Brown-Kelly (Plummer-Vinson) syndrome* present with mucocutaneous findings of brittle, spoon-shaped nails (koilonychia), atrophic tongue, and angular stomatitis (see Chapter 31 and Chapter 58). Dysphagia and iron deficiency complete the syndrome. Carcinoma of the postcricoid area of the esophagus occasionally occurs in long-standing cases. [69]

Flushing episodes may be the presenting sign of *carcinoid syndrome,* which has been seen primarily with carcinoid tumors of the small intestine with liver metastases. The flushing is a bright red color on the face and upper chest, occasionally involving the entire trunk and extremities, lasting from 10 to 30 minutes. After repeated episodes, permanent, rosacea-like telangiectasia and edema develop. Episodes may be provoked by food intake, alcohol, and emotional stress. The mechanism of the flushing is related to contributions by various vasoactive and neuropeptides, as discussed in more detail in Chapter 80. The best current treatment for carcinoid flush is octreotide, which is effective in up to 90% of cases. [70] Similar flushing episodes can occur in patients with multiple endocrine neoplasia. Pellagra-like skin lesions and cutaneous metastases have rarely been reported in patients with carcinoid tumors.

Necrolytic migratory erythema is the specific descriptive term for the cutaneous lesions of the glucagonoma syndrome, which occurs with a glucagon-secreting islet cell tumor of the pancreas (see Chapter 97). The cutaneous eruption typically affects perioral, lower abdominal, and perineal sites with a configurative erythema, erosions, and superficial necrosis and scale. Clinically, the condition resembles zinc deficiency. Patients often have angular cheilitis and a red, swollen tongue. The clinical presentation consisting of glucose intolerance, weight loss, psychological changes, and anemia is more important than skin biopsy in making the diagnosis. [71] Skin lesions may respond to intravenous amino acid supplementation, to octreotide or its analog, or to treatment of the underlying tumor. [72], [73] Necrolytic migratory erythema has occasionally been reported in the absence of a pancreatic tumor, but a vigorous search for a tumor is needed whenever typical skin lesions occur in a patient with other suggestive symptoms.

BULLOUS DISEASES OF THE SKIN THAT CAN DIRECTLY AFFECT THE GASTROINTESTINAL TRACT

Epidermolysis Bullosa

Epidermolysis bullosa (EB) is a spectrum of inherited mechanobullous diseases in which blisters occur spontaneously or at sites of friction or trauma. Classification of clinical subtypes of EB has been based on inheritance patterns, the presence or absence of scarring, and, more recently, the electron microscopically determined level of the blistering. Both dominant and recessive forms of dystrophic EB occur. Onset of skin lesions occurs at birth or shortly thereafter, and a dermatologic diagnosis of EB usually has been made before the patient presents to a gastroenterologist. Rarely, pyloric atresia has been found in infants with junctional or recessive dystrophic EB, apparently resulting from mucosal blistering and scarring in utero. The major sites of gastrointestinal involvement, most often occurring in the recessive dystrophic form, are the oral cavity, esophagus, and anal areas [74], [75] (see Chapter 63).

Esophageal lesions may become symptomatic at any time in the first 3 decades of life. Dysphagia is the most common symptom and may reflect the presence of esophageal bullae or weblike scarring, both of which can cause partial or total obstruction. Most such lesions occur in the cervical esophagus near the cricopharyngeal area. Anal involvement, with bullae and subsequent scarring, may lead to constipation. Lesions in this location are also prone to severe secondary infection. Nutritional deficiency is common in severe EB. EB acquisita is an uncommon late-onset disease with skin blistering and scarring at trauma sites and sometimes causing extensive, scarring oral and esophageal lesions. [76] There is no family history of EB in these patients, which occasionally has occurred in association with inflammatory bowel disease.

Pemphigus Vulgaris

Pemphigus vulgaris is an immunologic blistering disease that commonly presents with oral erosions before the onset of cutaneous lesions. Skin or mucosal biopsy findings of an acantholytic blister and direct and indirect immunofluorescence microscopic evidence of IgG in the intraepidermal space confirm the diagnosis. Symptomatic esophageal involvement is rare (see Chapter 63). [77] Pemphigus involving the lower gastrointestinal tract can present with hemorrhage. [78]

Bullous Pemphigoid

Bullous pemphigoid is another immunologic blistering disease in which older patients present with tense bullae. A biopsy specimen of the lesion shows a subepidermal blister, and IgG is found in the basement membrane zone in a linear pattern on direct and indirect immunofluorescence microscopy. Mucosal involvement is less common in bullous pemphigoid than in pemphigus (see Chapter 63), occurring in the mouth in about one third of pemphigoid patients and less often in the esophagus or anus. [79] There have been case reports of bullous pemphigoid occurring coincident with gastrointestinal malignant disease, particularly carcinoma of the colon.

SKIN CONDITIONS ASSOCIATED WITH ABDOMINAL PAIN

Porphyrias

Variegate porphyria is the most common porphyria in which patients present with skin lesions and episodic abdominal pain. The typical skin lesions (the same as are seen in porphyria cutanea tarda; see section "Cutaneous Signs Associated with Liver Disease") are noninflamed blisters and erosions at sun-exposed sites, particularly the backs of the hands. Scarring with milia formation, hyperpigmentation, facial hypertrichosis, and sclerosis may occur. Photoprotection is of minimal benefit because the patients are exquisitely sensitive to light, and avoidance of inducing drugs is important. Hereditary coproporphyria is even more rare but also may have photosensitivity as a component.

Fabry Disease

The skin lesions in *Fabry disease,* a rare lysosomal storage disease, are punctate, 1-mm, red to blue angiokeratomas that do not blanch. They are most densely distributed from the midabdomen to the knees. Lesions have their onset from childhood to adolescence. Decreased or absent sweating also may occur. The gastrointestinal manifestations of this disease are covered in Chapter 127.

Mastocytosis

Mastocytosis is an uncommon disorder of mast cell proliferation. The most common skin lesions are urticaria pigmentosa, with childhood onset. These are usually reddish-brown macules, but solitary nodules may occur. On stroking, these lesions typically itch, and a raised wheal or vesicle occurs (the Darier sign). Much less common is a telangiectatic variant or an erythroderma that occurs in adults. Skin biopsy confirms the increased numbers of mast cells in the dermis. Cutaneous symptoms may be controlled with antihistamines. Topical cromolyn and systemic psoralen in combination with ultraviolet A irradiation (PUVA) have been reported to be helpful in some patients. The gastrointestinal manifestations of mastocytosis are discussed in Chapter 42 and Chapter 128.

Herpes Zoster

T7 through L1 dermatomal involvement of *herpes zoster* may cause abdominal pain before the onset of typical grouped vesicular lesions. Involvement of sacral nerve roots can produce constipation and pain during defecation. [81] Postherpetic neuralgia can cause persistent, recurrent pain in the involved dermatome.

Rosacea

Rosacea is a chronic inflammatory condition characterized by central facial flushing, telangiectasias, acneiform lesions, and possible complication of nasal thickening called *rhinophyma* (Fig. 49-13; see Color Fig. 49-13). Several reports have described a possible relationship between *Helicobacter pylori* infection of the gastric mucosa and rosacea; however, no large case control studies have been performed to date to prove a causal relationship. [82] Rosacea is commonly treated with topical or oral antibiotics, with the highest response rate usually obtained with oral tetracycline.

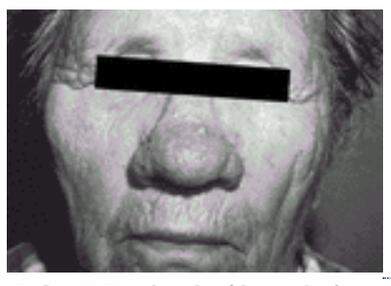

FIGURE 49-13. (See Color Fig. 49-13.) Characteristic telangiectasias, acneiform papules, and rhinophyma in a patient with rosacea.

CUTANEOUS SIGNS ASSOCIATED WITH LIVER DISEASE

Jaundice

This yellow discoloration of the skin, mucous membranes, and sclerae occurs because of the accumulation of bilirubin and its metabolites in these areas. In liver disease, it usually results from impaired excretion of conjugated (direct) bilirubin and becomes clinically evident at blood levels greater than 2.5 mg/dL. It can occur in almost any type of liver disease. [83] Treatment of the underlying liver condition will cause the jaundice to resolve. Ultraviolet light can resolve neonatal hyperbilirubinemia.

Pruritus

Pruritus, which is often severe and debilitating, can occur in cholestasis, cirrhosis, chronic hepatitis C, primary sclerosing cholangitis, and other forms of liver failure. [84] Patients with liver disease–associated itching often have elevated bile acids, bilirubin, alkaline phosphatase, and cholesterol, but the degree of itching does not generally correlate with the degree of elevation of these values. In fact, as the liver disease progresses, the itch may disappear, and this sign often heralds liver failure.

Pruritus may be the only symptom of cholestasis of pregnancy, which has associated increased fetal morbidity and mortality. A study of 8 women with 13 pregnancies complicated by cholestasis revealed only 1 normal delivery, 8 stillbirths, and 3 cases of fetal distress, 1 of which resulted in fetal death. Three of these women had subsequent pregnancies with cholestasis treated with ursodeoxycholic acid and had healthy deliveries. [85]

Treatment of liver-related itch involves measures to decrease the pruritus as well as measures to target the underlying liver disease (e.g., interferon for hepatitis C or surgery for common duct stones). Topical and systemic corticosteroids, PUVA therapy, and sedating antihistamines can decrease pruritus. Opiate antagonists, such as naloxone, nalmefene, and naltrexone decrease itching, suggesting that a central opioid may be involved in the mechanism of pruritus. Symptoms resembling those of opiate withdrawal may occur with the use of these drugs, and naltrexone can be hepatotoxic. [86]

Other forms of cholestatic itch are treated empirically, because the pathogenesis of this itch is unknown. First-line agents are bile acid sequestering agents such as cholestyramine and colestipol, which are resins that bind bile acids in the bowel and thereby reduce blood levels. They are moderately effective in decreasing itching, but are poorly tolerated because of gastrointestinal discomfort. These agents may work by a means other than just decreasing bile acids, because they also decrease itching associated with uremia and polycythemia vera. It is typical for patients to respond initially, but many become somewhat resistant thereafter. Ursodeoxycholic acid is actually a bile acid that lowers endogenous bile acid secretion into the gut. It is the only specific therapy that causes both a biochemical and survival benefit in

patients with primary biliary cirrhosis. [87] Rifampicin can decrease the pruritus of cholestasis, but it must be used with care, because it can be hepatotoxic. [88] Serotonin antagonists such as ondansetron also work in some cases. [89]

Polyarteritis Nodosa

Polyarteritis nodosa is a reaction pattern characterized by tender palpable purpuric nodules and livedo (Fig. 49-14, see Color Figs. 49-14) most commonly on the anterior legs, often with associated fever, arthralgias, malaise, and fibrinoid necrotizing arteritis histologically. Renal insufficiency and microaneurysms may occur. Hepatic arteritis was seen in 100% of patients with polyarteritis nodosa at autopsy in one study. [90] Intrahepatic hemorrhage is also reported. [91] Polyarteritis nodosa can occur in association with hepatitis B and, less commonly, hepatitis C and infections with parvovirus B19 and human immunodeficiency virus. It is often treated with immunosuppressive medications such as corticosteroids, which could exacerbate the underlying infection.

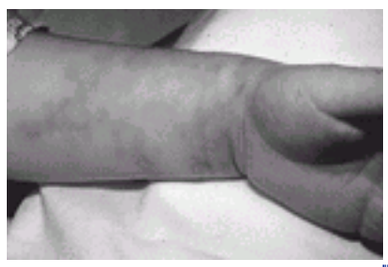

FIGURE 49-14. (See Color Fig. 49-14.) Polyarteritis nodosa with characteristic livedo reticularis and tender nodules.

Cryoglobulinemia

Cryoglobulins are immune complexes that can form as a reaction to drugs or infections. They can deposit within blood vessels and can create palpable purpuric nodules similar those seen in polyarteritis nodosa, with similar systemic complaints. Signs and symptoms are worsened by cold exposure and may include the Raynaud phenomenon or acral skin infarcts (Fig. 49-15; see Color Fig. 49-15). Hepatitis C is probably the leading cause of essential mixed cryoglobulinemia (type II or III forms). [92] Patients with hepatitis B infections have detectable cryoglobulins, usually of mixed type, in around 15% of cases. [93] Persistent chronic hepatitis A infection with cholestasis may result in cryoglobulinemic vasculitis. [94] Cryoglobulins resolve with treatment of underlying liver disease.

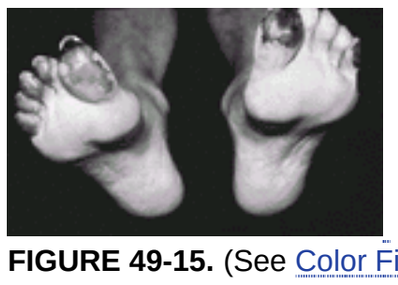

FIGURE 49-15. (See Color Fig. 49-15.) Cryoglobulinemia often results in acral vasculitic infarcts.

Papular Acrodermatitis of Childhood

This condition, also known as *Gianotti-Crosti syndrome,* is associated with hepatitis B, Epstein-Barr infection, and other viral infections in children. [95] It is characterized by asymptomatic erythematous papules that arise on the extremities, may occur on the face, and typically spare the trunk. These lesions tend to last 2 to 4 weeks and resolve on their own.

Lichen Planus

Lichen planus is a mucocutaneous condition characterized by pruritic, purple, polygonal papules with surmounting Wickham striae and lacy reticulate white mucosal plaques. It can occur in conjunction with hepatitis C in significantly higher prevalence than in normal controls. [96] Lichen planus can also be seen in primary biliary cirrhosis and many other liver diseases. Most often, however, there is no underlying disease. Treatments include systemic or topical corticosteroids, dapsone, PUVA therapy, methotrexate, and thalidomide.

Porphyria Cutanea Tarda

Porphyria cutanea tarda is characterized by subepidermal bullae, milia, and scarring most often on the dorsum of the hands, with hypertrichosis and hyperpigmentation of the face, photosensitivity to visible light (Soret band, 410 nm), and iron overload. It is often acquired in chronic liver diseases such as alcoholic or viral hepatitis and cirrhosis. Hepatitis C RNA has been detected in up to 91% of patients with porphyria cutanea tarda. [97] Most often, the predisposition is an inherited, dysfunctional enzyme known as uroporphyrinogen decarboxylase, which is important in porphyrin metabolism, and the liver disease potentiates expression of porphyria cutanea tarda. Treatments include phlebotomy, antimalarials, deferoxamine, and thalidomide.

Hypermelanosis

Several theories exist regarding why patients with chronic liver disease such as cirrhosis may develop hypermelanosis, which usually manifests as a diffuse or patchy grayish mucocutaneous pigmentation. It may occur because of increased levels of adrenocorticotropic hormone or melanocyte-stimulating hormone. As a result, the melanin content of the skin may be increased, and giant melanosomes may be present. Hemosiderin may also be directly deposited in the skin in iron overload conditions such as hemachromatosis.

Vascular Changes

Some of the vascular changes of liver disease are thought to occur because of altered estrogen metabolism. Spider angiomata, which are small arteriovenous malformations characterized by a central feeding vessel and radiating telangiectasias, are often seen in chronic liver disease. They are most common on the face, upper trunk, and distal upper extremities. Palmar erythema may be patchy or diffuse.

Nail Changes

Terry's nails is a term for an opaque whitish discoloration of the nails, with the distal third being pink. This discoloration can be seen in cirrhosis, but it is not specific to liver disease. Azure lunula (blue discoloration of the half-moon distal part of the nail matrix) is seen in hepatolenticular degeneration (Wilson disease), which is the result of abnormal copper metabolism. Clubbing may also occur in liver disease.

Other Cutaneous Manifestations

Urticaria, urticarial vasculitis, other forms of vasculitis, erythema multiforme, erythema nodosum, and pyoderma gangrenosum—all covered elsewhere in this chapter—can occur in liver disease, especially in patients with hepatitis B infection. Acute hepatitis B infection can produce a serum sickness–like reaction with fever, arthralgias, polyarthritis, urticaria, and vasculitis.

PANCREATIC DISEASE

Periumbilical purpura (Cullen sign) and left flank purpura (Grey Turner sign) are caused by dissection of blood along fascial planes from retroperitoneal bleeding of acute hemorrhagic pancreatitis. Presentation of tender, red, subcutaneous nodules, occurring at any site but most commonly on the pretibial region of the extremities, should raise the diagnostic possibility of *panniculitis* (Fig. 49-16; see Color Fig. 49-16). *Pancreatic fat necrosis* may feel fluctuant, but lesions rarely ulcerate or discharge fluid. [98] Pancreatic fat necrosis may be seen as the presenting sign in 22% of patients with pancreatitis or in up to 65% of patients with pancreatic carcinoma. Excisional biopsy (not punch biopsy) is indicated to confirm the diagnosis of panniculitis. The clinical differential diagnosis includes superficial migratory thrombophlebitis, which may be seen in pancreatic, gastric, or other cancers and in Behçet disease.

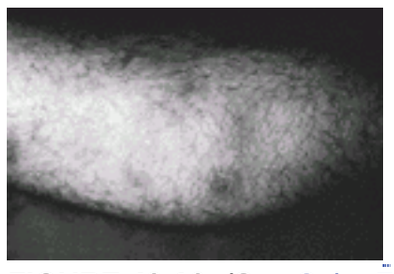

FIGURE 49-16. (See Color Fig. 49-16.) Panniculitis presented as tender erythematous nodules in a patient with pancreatitis.

The eruption of multiple yellow-red papules, predominating over extensor surfaces or buttocks, suggests a diagnosis of eruptive xanthomatosis. These lesions occur in the presence of hypertriglyceridemia and type IV or V hyperlipoproteinemia and may develop in a patient with pancreatitis, particularly in the setting of diabetes mellitus.

DERMATOLOGIC DISORDERS ASSOCIATED WITH MALABSORPTION

Dermatitis herpetiformis is an uncommon blistering disease characterized by intense pruritus and multiple grouped small vesicles distributed symmetrically over the body (Fig. 49-17; see Color Fig. 49-17). The scalp and extensor surfaces are prominently involved, and the itching and excoriation may be so severe that intact blisters are not conspicuous. Diagnosis is made by skin biopsy, which shows a subepidermal blister with a neutrophilic infiltrate of the dermal papillae. Direct immunofluorescence of perilesional skin confirms the diagnosis of dermatitis herpetiformis with the demonstration of IgA at the dermal-epidermal juncture or within the dermal papillae. [99], [100]

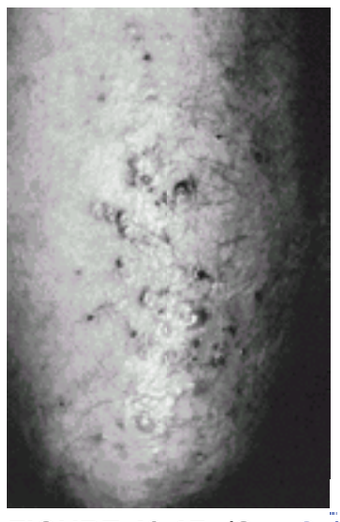

FIGURE 49-17. (See Color Fig. 49-17.) Dermatitis herpetiformis. Both elbows are involved. Note the intact vesicles and multiple crusted (i.e., excoriated) lesions.

Gastrointestinal manifestations may not be clinically prominent in patients with dermatitis herpetiformis who initially present to a dermatologist. Nevertheless, almost all patients have gluten sensitivity of the small intestine (see Chapter 76). Steatorrhea is observed in 20% to 30% of patients, and anemia may occur secondary to malabsorption (e.g., of folate or iron). Even in the absence of gastrointestinal symptoms, strict observance of a gluten-free diet leads to improvement of the skin disease (which may take 6 to 24 months of careful diet restriction) and minimizes the need for medication. [101], [102] Gluten challenge may cause a flare of the skin lesions in patients whose condition was previously controlled by diet.

Therapy of dermatitis herpetiformis with one of the sulfones, usually diaminodiphenylsulfone (dapsone) or with sulfapyridine, leads to prompt relief of symptoms and halting of lesion formation (within hours to a few days). Oral dapsone is often started at a dose of 100 to 200 mg daily in adults and is titrated according to the clinical response and with the goal of minimizing side effects.

Malabsorption can occur with other skin diseases. Patients with extensive inflammatory skin disease, such as exfoliative erythroderma or atopic eczema, develop secondary malabsorption, termed *dermatopathic enteropathy.* [103] This is usually manifested as mild steatorrhea and is often asymptomatic; resolution is prompt with treatment of the skin disease. The mechanism is unclear, but it does not appear to result from gluten sensitivity. Malabsorption also can occur as a result of abnormal motility or vascular compromise in systemic sclerosis and systemic vasculitides. Skin manifestations of chronic malabsorption are largely nonspecific and reflect altered nutritional status.

NUTRITIONAL DISORDERS

Although classically, the cutaneous manifestations of nutritional deficiency have been separated with regard to specific nutrients, there is usually considerable overlap, and many of the skin signs are nonspecific. [104] See Chapter 17, Chapter 18, Chapter 19 and Chapter 20 and Chapter 53 for further information.

Protein Malnutrition

Marasmus (total starvation) in the United States is usually limited to severely ill patients. Such patients have dry, loose skin, loss of subcutaneous fat, and thin, slowly growing hair, and, occasionally, prominent facial lanugo hair and hyperkeratosis around hair follicles. Absence of edema separates marasmus from kwashiorkor. In *kwashiorkor* (carbohydrate excess with protein deficiency), there may be hypopigmented and hyperpigmented scaling patches with peeling of the epidermis over edematous sites, joints, and flexural areas (flaky paint appearance). Purpura, which also may occur, results in mixed coloration. The hair may demonstrate alternate bands of dark and light pigmentation that correlate with episodes of inadequate protein intake (flag sign). Zinc and vitamin deficiencies usually accompany these

malnutrition states and undoubtedly contribute to the cutaneous findings. [105]

Essential Fatty Acid Deficiency

Essential fatty acid deficiency can occur in patients with fat malabsorption from any cause. It has been less common in patients who have been receiving fat-free parenteral nutrition because parenteral lipid supplementation is now routine. [106] Cutaneous lesions are typically diffusely distributed, erythematous scaling patches. There may be alopecia and traumatic purpura. Linoleic acid is the major fatty acid involved, and there is direct evidence of its role in the repair of the skin lesions. Older studies suggested that topical application of inoleic acid, as sunflower or safflower oil, could correct the total deficiency, but more recent evidence stresses the importance of systemic replacement.

Vitamin Deficiencies

Vitamin A deficiency may occur with fat malabsorption or liver disease. The classical change is that of multiple keratotic follicular papules over the extremities (phrynoderma), usually with generalized dry skin. Keratinizing metaplasia may occur on mucosal surfaces. Skin improvement occurs slowly with replacement therapy. *Vitamin A excess* may be more common than deficiencies, especially with the use of synthetic retinoids and supplemental vitamin A intake. Alopecia, fine erythematous scaling (sunburn appearance), cheilitis, skin fragility, and brittle nails will slowly resolve after the excess is corrected.

There are many similarities in the cutaneous manifestations of deficiency of the B vitamins riboflavin, pyridoxine, and niacin. Seborrheic dermatitis appearing over the face, prominent involvement of the perioral and groin areas, and a smooth tongue are common features. In *pellagra* (niacin deficiency), there may be painful erosive and pigmented scaling patches, especially in sun-exposed sites. When this occurs on the upper chest, it is termed a *Casal necklace.* Pellagra still occurs in those with inadequate intake (e.g., patients with alcoholism, elderly persons) and in patients with severe malabsorption. It can occur also in patients with carcinoid, or in those receiving therapy with isoniazid, 6-mercaptopurine, or 5-fluorouracil. Skin lesions respond dramatically to niacin replacement therapy.

Cutaneous manifestations of *vitamin B_{12} deficiency* are uncommon. Generalized hyperpigmentation, which may specifically involve palms, soles, nails, and white hair, alopecia areata, or vitiligo may occur in association with pernicious anemia. *Folic acid deficiency* may present with an unusual lemon-yellow skin color.

Vitamin C deficiency still occurs fairly often, especially in elderly persons and in patients with alcoholism who have inadequate dietary intake. Purpura and petechiae, perifollicular hemorrhage with corkscrew-shaped hairs, and gingival erosions around teeth are the prominent features. Mucosal lesions respond first to replacement of vitamin C, followed by slower resolution of purpura.

Zinc Deficiency

Zinc deficiency, or acrodermatitis enteropathica, may be genetic or acquired. [107] The cutaneous manifestations are acrally distributed (hands, face, feet, anogenital region) and are typically erythematous, scaling, vesiculopustular, or eroded plaques. There may be alopecia, impaired wound healing, and stomatitis. Secondary candidal infection is common and should also be treated. Zinc replacement, pancreatic enzyme supplementation, and 8-hydroxyquinoline have been used in the therapy.

PERINEAL SKIN LESIONS

There are several skin lesions that occur in the perianal or perineal areas that may be encountered in the course of examination or endoscopy procedure (Table 49-3) (see Chapter 92). Vesicles or erosions on an erythematous base should suggest herpetic lesions. Herpes simplex is often recurrent in this site, and in immunocompromised patients, lesions may be persistent, crusted erosions or ulcers. Herpes zoster usually presents acutely as dermatomal grouping of vesicles. Perianal ulcers and erosions also may occur in Crohn's disease, in amebiasis, in primary or secondary syphilis, or in nontreponemal venereal disease. Flesh-colored pedunculated papules in the perineal area often are condylomata acuminata (venereal warts). Lesions, if they are few, usually respond well to therapy with cryosurgery, electrofulguration, surgical or laser excision, or application of topical podophyllin. Refractory lesions may respond to intralesional or systemic interferon.

Erythema with Scale or Maceration
Contact dermatitis
Seborrheic dermatitis
Psoriasis
Candidiasis
Dermatophytosis
Secondary syphilis
Extramammary Paget disease
Nutritional deficiencies
Bowen disease
Vesicles, Erosions, and Ulcers
Herpes simplex
Herpes zoster
Impetigo (streptococcal, staphylococcal)
Syphilis (primary or secondary)
Chancroid
Deep fungal, acid-fast bacilli, protozoal infections
Bullous pemphigoid
Pemphigus
Ecthyma
Nodules, Tumors, and Ulceration
Condylomata acuminata (i.e., warts)
Hidradenitis suppurativa
Squamous or basal cell carcinoma
Crohn's disease
Carcinoma: metastatic or direct extension
Kaposi sarcoma
Granulomatous herpes simplex

TABLE 49-3 Perineal Skin Lesions (Practical List of Common Causes)

Erythema with scaling or maceration in the gluteal fold and perianal area raises the major considerations of candidal infection, psoriasis, or seborrheic dermatitis, irritant contact dermatitis (from fecal soiling), or bacterial (streptococcal or staphylococcal) impetigo. Clinical presentation, potassium hydroxide preparation of skin scrapings, and culture are helpful in differentiating these diagnoses. If lesions do not respond to appropriate therapy for one of these diagnoses, biopsy may be indicated to rule out extramammary Paget's disease. Idiopathic pruritus ani is a diagnosis that should be made only after a primary dermatosis, such as those just listed, has been excluded. It is a common complaint, particularly in middle-aged men, and psychological factors have been emphasized in some reviews. [108], [109] The key to effective therapy of pruritus ani is eliminating underlying triggering factors, including infections, irritants, allergies, and anorectal disorders (e.g., rectal fissures, prolapse). Careful cleaning measures, dietary changes, and treatments ranging from topical corticosteroids to intralesional steroids or cryosurgery have been used successfully. Treatment is most successful if the symptoms have been present for less than 2 years.

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CHAPTER 50

John Rabine* Timothy T. Nostrant

ORAL MANIFESTATIONS OF GASTROINTESTINAL DISEASE

Many gastrointestinal disease states are associated with oral findings (Table 50-1). This chapter briefly addresses the disorders in which oropharyngeal pathology is most common.

TABLE 50-1 Oral Lesions Associated with Gastrointestinal Diseases

GASTROESOPHAGEAL REFLUX DISEASE

The supraesophageal manifestations of gastroesophageal reflux disease (GERD), including reflux laryngitis and reflux-associated asthma, have gained increasing attention. Dental injuries, predominantly erosions, are also very common. Erosions are defined as circumscribed, punched-out, discolored regions on the surface of a tooth corresponding to enamel loss (Fig. 50-1) and can be differentiated from caries, which are dark defects with jagged edges. [1] Gastric acid exposure has been the proposed mechanism of dental erosions because of its association with salivary loss, [2] bulimia, [3] and hiatal hernias. [4] Reflux has been documented in 83% of patients with erosions referred by their dentists for pH monitoring. [5]

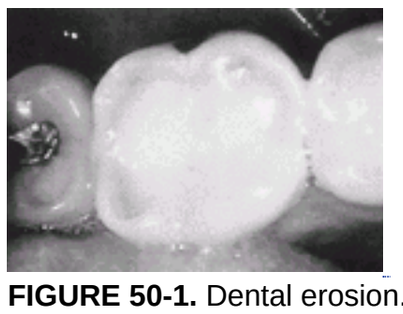

FIGURE 50-1. Dental erosion. Circumscribed yellowish regions correspond to exposed dentin in areas where enamel has been lost during repeated exposure to gastric contents in a female patient with bulimia.

The true incidence of dental erosions in patients with GERD is unclear, but at least 20% of patients with reflux have been found to have erosions, [6], [7] and more than 50% of patients with proven reflux by pH monitoring have been shown to have dental erosions. [5] Extrinsic acid exposure (e.g., citrus fruits, soft drinks, vinegar, chewable vitamin C tablets) is more likely to induce erosions on the labial surfaces of upper anterior teeth, whereas intrinsic acid from GERD is more likely to harm the lingual surfaces of teeth. [1], [8], [9], [10] and [11]

The treatment of GERD-induced dental erosions is obviously aimed at limiting reflux and oral acid exposure. Patients should avoid acidic foods and drinks, allow antacids to dissolve in the mouth during symptomatic reflux events, and avoid brushing their teeth immediately after a reflux event. Chewing gum (sugarless) can be used to stimulate salivary flow between meals, and medications that reduce salivary flow should be avoided. Patients with xerostomia resulting from medications, previous surgery, or previous radiation therapy should drink water frequently or use a saliva substitute. Patients with erosions should be under the care of a dentist, who can provide a polyethylene mouth guard at night to limit dental exposure to acid, topical fluoride pastes, and restorative dental procedures once the underlying reflux disorder has been addressed by the gastroenterologist.

Laryngeal abnormalities are also very common with supra-esophageal acid reflux. Symptoms that suggest reflux laryngitis include hoarseness, throat clearing, globus sensation, halitosis, and prolonged vocal cord warm-up. [12] Barium esophagram, upper gastrointestinal endoscopy, and standard esophageal manometry are diagnostic in fewer than 25% of cases, whereas results of 24-hour pH monitoring are positive for distal reflux in 80% and for proximal reflux in 30%. Reflux laryngitis can be induced with minimal acid exposure. Small amounts of acid applied to the vocal cords only three times per week will induce significant cord edema and inflammation. [13] Therefore, microaspiration of small, undetectable amounts of acid may cause laryngitis, which explains both the need for strong acid suppression for effective treatment and the low number of abnormal pH studies in those patients who do respond to treatment. [14], [15] Proximal acid reflux is cleared more slowly than distal acid reflux, and acid may reflux into the oral cavity when none is present distally. Dual pH monitoring is crucial for a diagnosis in patients with supra-esophageal

symptoms.

The gastroenterologist may be able to identify reflux laryngitis during routine upper gastrointestinal endoscopy because the larynx is well visualized. [16], [17] The posterior glottis, including the posterior edges of the vocal cords and the mucosa overlying the arytenoid cartilage, are seen best during inspiration, when the vocal cords are abducted. The vocal cords are better examined during phonation, when the cords are adducted. Classical posterior laryngitis consists of red arytenoid tissue and "piled-up" interarytenoid mucosa, but these features are seen in the minority of patients with reflux laryngitis (Fig. 50-2). [13] Laryngeal edema is the most common finding but may be easily overlooked. Table 50-2 summarizes the laryngoscopic findings of reflux disease. [16]

Reflux laryngitis
Vocal cord nodules
Polypoid degeneration
Contact ulcers/granulomata
Laryngeal stenosis
Proximal laryngospasm
Laryngeal cancer

TABLE 50-2 Laryngeal Findings of Gastroesophageal Reflux Disease

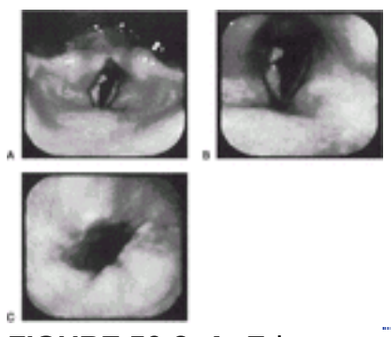

FIGURE 50-2. A: Edema and erythema of the vocal cords and posterior glottis (reflux laryngitis). **B:** Vocal cord granuloma. **C:** Distal reflux esophagitis in the same patient as in **A** and **B**.

Studies have defined the benefits of acid suppression for reflux laryngitis. A 12-week course of omeprazole clearly improved laryngoscopic findings except for granulomata. [18] An initial response rate of 65% was seen in 22 patients treated with 40 mg of omeprazole daily for 8 weeks. Relapse occurred in 38% after omeprazole was stopped. Abnormal pH studies were documented in the majority of nonresponders, and the response was not improved with twice-daily omeprazole. [19]

POLYPOSIS SYNDROMES

Gardner Syndrome

The oral manifestations of diseases associated with intestinal polyposis, such as Gardner syndrome, may be the first clue to the diagnosis.

Multiple mutations of the adenomatous polyposis coli (APC) gene result in multiple forms of Gardner syndrome. Bone tumors, soft tissue tumors, lipomas, desmoids, and sebaceous cysts may be seen (Fig. 50-3). Dental abnormalities, including unerupted teeth, supernumerary teeth, and dentigerous cysts, may predispose these patients to early and profound tooth loss. [20]

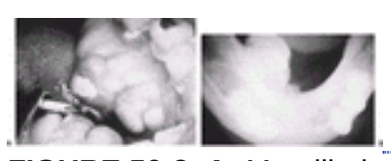

FIGURE 50-3. A: Mandibular osteoma of Gardner syndrome. **B:** Radiograph of the mandible in the same patient. (From Bresalier RS. Neoplasia of the colon and rectum. In: Feldman M. Gastroenterology and hepatology: the comprehensive visual reference. Philadelphia: Current Medicine, 1997.)

Peutz-Jeghers Syndrome

Small intestinal hamartomatous polyps are the classical finding in Peutz-Jeghers syndrome, an autosomal dominant disease with variable penetrance. Pigmentation abnormalities with melanin deposition on mucosal surfaces are the visual hallmark. Typically appearing in the first year of life, small black or blue-gray macules form on the lips, perioral region, and buccal mucosa but may develop on any mucocutaneous surface, including the fingers and toes. Perioral lesions often fade with age, while those in the mouth or on the extremities may become more apparent. [21] Overall, the risk for malignancy in multiple organs can be as high as 30%, and patients require periodic surveillance.

Cowden Disease

Multiple hamartoma syndrome, or Cowden disease, is a rare polyposis syndrome in which facial trichilemmomas and oropharyngeal papillomatous growths appear in the second decade. [22], [23] Gastrointestinal tract malignancy is not increased, but a higher incidence of extraintestinal malignancy is noted, particularly breast and thyroid cancers. Patients' overall risk for malignancy and requirement for surveillance are similar to those for patients with Peutz-Jeghers syndrome.

BLEEDING DISORDERS

See Chapter 49 and Chapter 130.

Osler-Weber-Rendu Syndrome

Hereditary hemorrhagic telangiectasia, or Osler-Weber-Rendu syndrome, is an autosomal dominant disorder characterized by widespread mucocutaneous telangiectases of the face, ears, lips, oral cavity, nasopharynx, and hands. Major sources of bleeding are the nose and stomach, although colonic bleeding and oral bleeding are recorded. [20] Similar telangiectases can be seen in scleroderma and CREST (*c*alcinosis cutis, *R*aynaud phenomenon, *e*sophageal motility disorder, *s*clerodactyly, and *t*elangiectasia) syndrome, so the clinical features of these disorders can be used to differentiate them from Osler-Weber-Rendu syndrome. [24]

Blue Rubber Bleb Nevus Syndrome

Blue rubber bleb nevus syndrome is characterized by multiple blue-colored, cavernous hemangiomata of the skin, oropharynx, and gastrointestinal tract that may

rupture and bleed. The skin and gastrointestinal tract are affected more commonly than the mouth. [20], [25] Histologically, these lesions consist of large, thin-walled vascular spaces with vessels lined by hyperplastic endothelium and separated by a scant network of elastin-deficient connective tissue. [26]

Pseudoxanthoma Elasticum

Pseudoxanthoma elasticum is a rare form of elastic tissue degeneration producing thin vascular abnormalities in the gastrointestinal and genitourinary tracts that bleed.

IMMUNODEFICIENCY STATES

Acquired Immunodeficiency Syndrome

Oral infections are frequently associated with the human immunodeficiency virus and cause significant disability in affected patients from anorexia, decreased oral intake, weight loss, and malnutrition. Common oral findings are listed in Table 50-3.

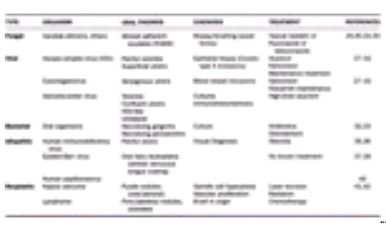

TABLE 50-3 Immunodeficiency States (AIDS)

Graft-versus-Host Disease

Graft-versus-host disease is a significant cause of morbidity in patients who have undergone bone marrow transplantation. It generally occurs within the initial 3 to 4 weeks after transplantation and may present as mucositis, dermatitis, enteritis, and hepatic dysfunction. Chronic graft-versus-host disease often develops in patients with acute graft-versus-host disease and typically presents at 3 months after transplantation. Clinically, patients have symptoms suggestive of sicca complex and scleroderma. Painful oral mucositis and upper gastrointestinal dysmotility may develop, the latter resulting in gastroesophageal reflux and bacterial overgrowth. In addition to more aggressive immunosuppression, patients with graft-versus-host disease require nutritional and fluid support, as well as therapy for any infectious complications.

COLLAGEN VASCULAR DISEASE

Scleroderma is the classic collagen vascular disease associated with oral manifestations. Perioral skin tightening, telangiectases of the lips and tongue, and dysgeusia are common findings. Sclerosis of the esophagus and small intestine may result in the sequelae of reflux disease and malabsorption from bacterial overgrowth, respectively. The sicca complex, characterized by decreased salivary and lacrimal gland secretion, may be seen in numerous connective tissue diseases but is classically associated with rheumatoid arthritis (Sjögren's syndrome). Dental caries, tongue and buccal fissures, and swallowing difficulties may all develop. Based on the presence of serum and salivary antibodies to human T-lymphocyte virus-1 (HTLV-1), this virus has been implicated in the pathogenesis of Sjögren's syndrome. [43] Oral manifestations are not common in dermatomyositis, but oral ulcers, leukoplakia-like lesions, and gingival telangiectases have been reported in association with this disorder. [44]

NUTRITIONAL DEFICIENCIES AND MALABSORPTION

As summarized in Table 50-4, vitamin deficiencies may result in numerous epithelial changes in the oropharynx (Fig. 50-4, Fig. 50-5 and Fig. 50-6). Such deficiencies are usually associated with malabsorption or with inadequate intake in the setting of chronic illness. Rarely, a deficiency stems from competition for normal body stores, such as vitamin B_{12} deficiency in *Diphyllobothrium latum* infestation. The oropharyngeal manifestations of the malabsorptive states depend on the specific nutrient deficiency. A sore mouth (niacin deficiency), cheilitis and glossitis (B vitamin and folate deficiencies), stomatitis and oral ulceration (B vitamin, folate, and niacin deficiencies), and bleeding (vitamin K deficiency) may all develop. White patches on the tongue reflect vitamin A deficiency. The hypermetabolism of tryptophan to serotonin with decreased niacin production can cause pellagra in patients with carcinoid syndrome.

TABLE 50-4 Vitamin/Mineral/Trace Metal Deficiencies

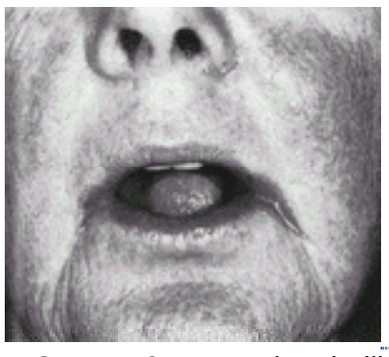

FIGURE 50-4. Angular cheilitis. Inflammation at the angles of the mouth is commonly associated with deficiencies of B vitamins, such as riboflavin (vitamin B_2), niacin (vitamin B_3), and pyridoxine (vitamin B_6). Secondary *Candida* or *Staphylococcus* infection may develop. (From Habif TP. Superficial fungal infections. In: Baxter S, ed. Clinical dermatology: a color guide to diagnosis and therapy, 3rd ed. St. Louis: Mosby, 1996:400.)

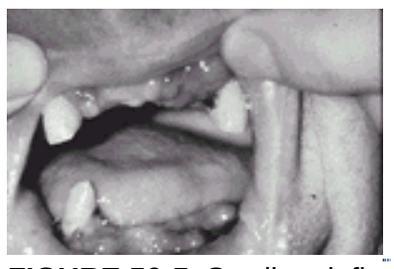

FIGURE 50-5. Swollen, inflamed, bleeding gums and poor dentition in a case of scurvy. (From Briggaman R. Clinical nutrition I slide set. Bethesda, MD: American Gastroenterological Association, 2000.)

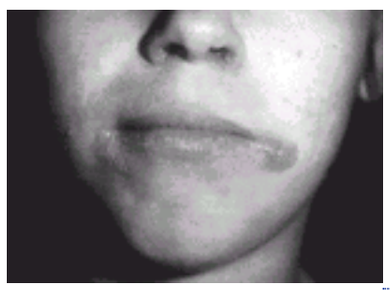

FIGURE 50-6. Zinc deficiency may present as a scaly, erythematous rash at the angles of the mouth and the nasolabial folds in addition to stomatitis and tongue coating. (From Grossman M. Clinical nutrition I slide set. Bethesda, MD: American Gastroenterological Association, 2000.)

INFLAMMATORY BOWEL STATES

Celiac Disease

Cheilitis, gingivitis, and small vesicular ulcers of the tongue and oral mucosa are seen in celiac sprue. [20] The oral cavity may burn. Numerous small white ulcerations may develop on the dorsum of the tongue, and swelling of the tongue can result in a “strawberry” appearance.

Crohn’s Disease and Ulcerative Colitis

Oral lesions are extremely common in inflammatory bowel disease (IBD), with a prevalence of 4% to 30%, and are more frequently associated with Crohn’s disease than with ulcerative colitis. [45], [46] A prevalence of 0.5% was noted in a tertiary care center specializing in oral dermatology. [47] In addition to extraintestinal inflammation, malabsorption and subsequent malnutrition may develop in IBD secondary to rapid transit, stricture formation, enterocolonic fistulae, severe mucosal loss, and bacterial overgrowth. Deficiencies of folate, iron, vitamin B_{12}, and the fat-soluble vitamins are most common. Trace metal deficiencies may develop in patients with IBD who are on total parenteral nutrition without adequate replacement.

Crohn’s Disease The oral lesions of Crohn’s disease typically reflect active intestinal disease, although they may precede or follow clinically evident intestinal flares. [20] Aphthous linear labial ulcers, pseudopolyps, and labial or buccal swelling have all been reported. [20], [47] The various lesions frequently heal spontaneously but may persist for several years, making it difficult to define a correlation between oral manifestations and intestinal disease activity. [48], [49] Nonetheless, some have proposed that an eruption of aphthous ulcers actually predicts an impending flare of Crohn’s disease. [50] The pathogenesis of aphthous lesions is unclear, but it has been suggested that lymphocytes activated against enterobacterial toxins may release inflammatory mediators; diminished immunoglobulin A secretion may also be involved. [48], [49], [50], [51] and [52] Although not specific for IBD, aphthous ulcers are the most common oral manifestation of Crohn’s disease, seen in up to 20% of patients with this disease. [20] The distinct, shallow white ulcers may occur singly or be widespread throughout the oral cavity (Fig. 50-7). Gingival hyperplasia and nodularity are not uncommon, and a cobblestone pattern may appear on the palate. A cobblestone pattern may also appear on the buccal mucosa when smooth nodules are present. Oral candidiasis has been seen in up to 5% of patients with Crohn’s disease, a feature not found in patients with ulcerative colitis within the same trial. [46] In one trial assessing the treatment of oral lesions in patients with Crohn’s disease, complete remission of the lesions after treatment with systemic steroids or azathioprine was noted in only 50%, whereas topical steroids induced a remission in 58% of cases. [53]

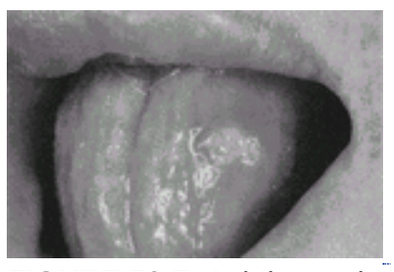

FIGURE 50-7. Aphthous ulcer. Aphthae appear as minute, shallow white ulcers along mucous membranes. Although they may occur in healthy individuals, multiple or persistent lesions suggest an underlying diagnosis such as inflammatory bowel disease or Behçet disease. (From Allison MC. Diagnostic picture tests in gastroenterology. London: Mosby-Wolfe, 1991.)

Ulcerative Colitis Aphthous stomatitis and ulcers may be seen in patients with ulcerative colitis, although ulcers are more typical of Crohn’s disease. Cheilitis may also occur and has been associated with scaling perioral skin and cervical adenopathy. [20], [45] In pyostomatitis, also known as *pyoderma vegetans,* deep pustules may progress to deep ulcers, classically on the lower lip but occasionally also on the buccal mucosa. Like many oral manifestations of IBD, this lesion often develops during a colitis flare and resolves with aggressive IBD management. Although not classically thought of as affecting the oral cavity, pyoderma gangrenosa of the tongue has been reported and is characterized by deep ulceration and necrosis. It has been suggested that all these lesions involve an immunologic reaction, such as circulating antigen-antibody complexes. [20] Unfortunately, many oral manifestations may persist or recur even after total colectomy. Although systemic steroids may relieve these lesions, topical or locally injected steroids sometimes lead to a more rapid and profound resolution.

Behçet Disease

Behçet disease is characterized by orogenital ulceration, ocular manifestations, and arthritis. Common problems include diarrhea, anorexia, bloating, and colorectal ulceration. Oral lesions consist of small or very large aphthous ulcers or herpetiform-like ulcers that tend to involve the palate, pharynx, and esophagus and can cause significant pain and dysphagia. Recurrent oral lesions may be the only manifestation of Behçet disease for several years. [54] Based on the clinical symptoms and appearance of the ulcers, IBD, herpes simplex virus, and Reiter syndrome all must be considered in the differential diagnosis. The oral lesions of Behçet disease are treated with topical steroids, nystatin, and tetracycline. [20] Thalidomide has been suggested as an alternative therapy, with an excellent response rate noted in one early trial. [55]

MISCELLANEOUS DISORDERS

The oral findings and diagnostic considerations in lead poisoning, amyloidosis, and Plummer-Vinson syndrome are listed in Table 50-5.

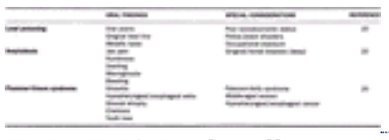

TABLE 50-5 Miscellaneous Disorders

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*The views expressed in this article are those of the author and do not reflect the official policy or position of the U.S. Air Force, U.S. Department of Defense, or U.S. Government.

CHAPTER 51

Edward N. Janoff and Phillip D. Smith

APPROACH TO GASTROINTESTINAL PROBLEMS IN THE IMMUNOCOMPROMISED PATIENT

Gastrointestinal illnesses are an important complication of immunodeficiency diseases and immunosuppressive therapy. Such illnesses may be severe, prolonged, or even fatal, whereas the same illnesses in immunocompetent persons are typically mild and transient. Most, but not all, gastrointestinal illnesses in immunocompromised persons are infections. [1], [2], [3] and [4] During the past 40 years, these infections have received increasing investigative attention, first in adults with hypogammaglobulinemia and children with congenital immunodeficiency syndromes, and then in patients with malignancies or on immunosuppressive drugs. In addition, the spectrum of conditions for which immunosuppressive drugs are used has expanded to include a wide array of autoimmune conditions, rheumatic diseases, malignancies, and organ, bone marrow, and stem cell transplantation. Distinguishing the effects of immunosuppressive drugs from those of infection and graft-versus-host disease has been a major challenge. Most recently, the pandemic caused by human immunodeficiency virus type 1 (HIV-1) has broadened the spectrum of gastrointestinal pathogens associated with immunosuppression and focused attention on effective clinical evaluation and antimicrobial therapy. Thus, although the numbers of immunocompromised hosts and identifiable enteric microorganisms have increased substantially, the efficient recognition and effective treatment of gastrointestinal illnesses and immune dysfunction are now the basis for increased optimism in the evaluation and management of gastrointestinal disease in immunocompromised persons.

The high incidence of gastrointestinal illnesses in immunocompromised patients underscores the fundamental role of systemic and mucosal immunity in host defense. Although these anatomically distinct systems may serve different functions and are composed, in part, of phenotypically and functionally distinct cell populations, systemic immunity and mucosal immunity represent an integrated and dynamic system of host defense. Evidence for the critical role of cellular and humoral defense mechanisms in providing protection against enteric pathogens is the strong association between HIV-1–induced immunosuppression and gastrointestinal infections. An array of other immunocompromising conditions also predispose the gastrointestinal tract to disease. These conditions are classified as primary or secondary (Table 51-1). Primary immunodeficiency disorders, which are uncommon, are caused by congenital defects in the T, B, or phagocytic cells. In contrast, secondary immunodeficiency disorders are more common than primary disorders and are acquired. Identification of the underlying primary or secondary immunodeficiency is important because the relative risk for associated gastrointestinal illness and the spectrum of such illnesses vary with each deficiency (see Table 51-1).

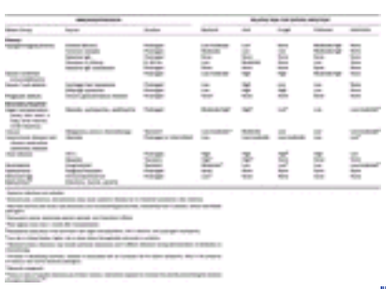

TABLE 51-1 Immunosuppression Disorders and Relative Risk for Enteric Infection

Because the most common gastrointestinal manifestation of immunodeficiency is infection, this chapter focuses on the approach to esophageal and enteric infections in patients with immunocompromising conditions. The first section of the chapter presents the clinical clues that suggest an underlying immunodeficiency in patients with enteric disease. The second section describes the gastrointestinal syndromes associated with immunodeficiency, and the third section reviews the specific pathogens associated with discrete immunologic defects.

EVALUATION OF UNDERLYING IMMUNE DYSFUNCTION ASSOCIATED WITH GASTROINTESTINAL ILLNESS

Clinical and Microbiologic Clues

The clinical history provides some of the most important clues that an underlying immunodeficiency disorder may be the cause of a gastrointestinal illness, particularly an infection. Features of the history that indicate the possibility of an immune disorder are listed in Table 51-2. Foremost among the clinical features is prolonged or severe diarrheal illness. Microbiologic features of the identified pathogen also may indicate a possible underlying immune disorder. Table 51-3 summarizes these features. [5], [6], [7], [8], [9], [10], [11], [12], [13], [14], [15], [16], [17], [18], [19] and [20] Once a primary or secondary immunodeficiency syndrome is suspected, a simple and sequential diagnostic evaluation of the patient’s immune function and the intestinal syndrome can be initiated.

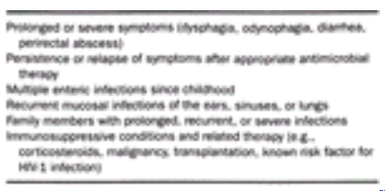

TABLE 51-2 Features of the History that Suggest an Underlying Immunodeficiency Condition

Presence of an opportunistic pathogen
Recurrence of a common enteropathogen (e.g., *Campylobacter jejuni* in children and adults with immunoglobulin deficiencies)
Presence of more than one opportunistic or common enteric pathogen
Persistence of an enteropathogen despite appropriate antimicrobial therapy
Simultaneous systemic and gastrointestinal infection with the same pathogen (e.g., *Streptococcus bovis* in colon cancer patients and *Salmonella* species, *Shigella* species, and *Campylobacter jejuni* in AIDS patients)

TABLE 51-3 Microbiologic Features of Enteric Infections that Suggest an Underlying Immunodeficiency Condition

Initial Evaluation

The physical examination findings, often underappreciated, also may provide information to suggest that an immunologic defect is present. Findings that may be associated with immunocompromising diseases or conditions are listed in Table 51-4. Clinicians should search for these features when an immune deficiency is suspected. After the history has been taken and the physical examination performed, the stool should be examined for enteric pathogens. The protocols for the identification of enteric pathogens in immunocompromised hosts are discussed in the chapters on the specific pathogens and the chapter on the gastrointestinal manifestations of AIDS. The most important tests used to detect pathogens in stool from immunodeficient subjects are listed in Table 51-5. Colonoscopy or esophagogastroduodenoscopy may be helpful for inspecting the mucosa and obtaining tissue for histopathological evaluation.

FINDINGS	IMMUNODEFICIENCY CONDITION
Ataxia, ocular telangiectases	Ataxia-telangiectasia
Facial abnormalities	DiGeorge syndrome
Oral thrush	Neutropenia, cell-mediated immunodeficiency secondary to chronic steroid use, HIV-1 infection, chemotherapy
Oral ulcers	Neutropenia, HIV-1 infection, HIM
Chronic periodontitis	Neutropenia, neutrophil chemotactic defects
Paucity of lymphoid tissues (nodes, tonsils)	X-linked agammaglobulinemia
Edema	Protein-losing enteropathy, nephrotic syndrome
Focal or diffuse adenopathy	HIV-1 infection, lymphoid malignancies, ± CVH, HIM
Splenomegaly	CLL, CML, CGD, ± CVH, early HIV-1 infection, HIM
Seborrheic dermatitis	HIV-1 infection, malnutrition
Petechiae or ecchymoses	Thrombocytopenia with granulocyte leukemias, chronic steroid use
Eczema	Wiskott-Aldrich syndrome, IgA deficiency, hyper-IgE

CGD, chronic granulomatous disease; CLL, chronic lymphocytic leukemia; CML, chronic myelogenous leukemia; CVH, common variable hypogammaglobulinemia; HIM, immunoglobulin deficiency with increased IgM.

TABLE 51-4 Findings on Physical Examination that Suggest the Presence of an Underlying Immunodeficiency Condition

TEST	PATHOGENS
Microscopic evaluation of stool for ova and parasites	*Giardia lamblia*, *Strongyloides stercoralis*, *Entamoeba histolytica*, *Cyclospora*
Culture (stool)	*Campylobacter* species, *Shigella* species, *Salmonella* species, *Aeromonas* species
Culture and/or toxin detection in stool (cytotoxin assay or ELISA for toxin A and B)	*Clostridium difficile*
Acid-fast stain (stool)	*Cryptosporidium*, *Mycobacterium* species, *Isospora belli*, *Cyclospora*
Histology (intestinal tissue)	Cytomegalovirus, herpes simplex virus, fungi, microsporidia
Immunocytochemistry (intestinal tissue)	Cytomegalovirus, herpes simplex virus
Electron microscopy (stool)[223]	Adenovirus, Norwalk virus/calcivirus, astrovirus, microsporidia
Polymerase chain reaction (stool)[223]	Norwalk virus/calicivirus
ELISA (stool)[223]	Rotavirus
Culture (stool)[223]	Adenovirus, rotavirus

*Excluding HIV-1–infected persons.

ELISA, enzyme-linked immunosorbent assay.

TABLE 51-5 Examination of Stool and Intestinal Tissues for Enteric Pathogens in Subjects with Diarrhea and Suspected Immunodeficiency*

Laboratory Analysis

When the history, physical examination, or presence of an unusual enteric pathogen suggests that the patient may be immunocompromised, selected laboratory tests will usually identify the underlying immunologic defect. [21] Although elegant and expensive immunologic tests are available, the laboratory analysis should begin with a complete blood cell count and differential (Table 51-6). The results of this routine test provide important information on the numbers of neutrophils and lymphocytes, particularly the presence of neutropenia and lymphopenia. Certain lymphoid malignancies, such as chronic lymphocytic leukemia, can be efficiently excluded when the number of lymphocytes is not increased. The levels of total serum protein and albumin are helpful in suggesting whether chronic liver disease or hematologic diseases, such as multiple myeloma or hypogammaglobulinemia, may be present. Thereafter, specialized or more complicated immunologic tests are helpful, depending on the suspected underlying immunodeficiency (see Table 51-6).

	ABNORMAL RESULT	IMMUNODEFICIENCY CONDITION
Routine Tests		
Complete blood cell count	Neutropenia	Cyclic neutropenia, chemotherapy, myelodysplasia, drug reaction, HIM
	Neutrophilia	Leukemoid reaction, normal response to infection (especially bacterial), steroids
	Lymphopenia[a]	SCID, HIV-1 infection, malnutrition, lymphoid malignancy, steroids, chemotherapy, ± CVID
	Lymphocytosis	Lymphoid malignancy (e.g., CLL)
	Thrombocytopenia	Wiskott-Aldrich syndrome
Total protein and albumin	Low albumin with high globulins	Chronic liver disease, malnutrition
(Total protein = albumin + γ globulins)	Normal albumin with high globulins	Multiple myeloma, polyclonal gammopathy (including HIV-1, CVID)
	Normal albumin with low globulins	Hypogammaglobulinemia, CLL
Specialized Tests		
Cellular		
Skin testing for delayed-type hypersensitivity (especially to Candida)	No response	Food dysfunction, lack of prior exposure to antigens
Lymphocyte subsets (flow cytometry)		
T cells	Low CD4 with normal-high CD8 percentage and number	HIV-1 infection
	Low CD4 and CD8	Inherited SCID, steroids, DiGeorge syndrome
	Increased with normal CD4/CD8 percentages	Inherited agammaglobulinemia
B cells	Increased number	CLL
	Reduced number	Inherited agammaglobulinemia, inherited SCID, CVID, X-linked immunodeficiency with hyper-IgM (no surface IgD or IgG), selective IgA deficiency
	Normal number	
Humoral		
Serum protein electrophoresis	Normal albumin with increased polyclonal gamma globulins	Polyclonal gammopathy (including HIV-1, CVID, HIV)
	Normal albumin with increased monoclonal gamma globulins	Multiple myeloma, benign monoclonal gammopathy
	Normal albumin with low gamma globulins	Hypogammaglobulinemia
Quantitative immunoglobulins (nephelometry)	Low level of isolated IgA, IgM, or IgG subclass	Selective IgA deficiency, ataxia-telangiectasia (IgA and IgG2)
	All Ig classes low	Hypogammaglobulinemia, CLL
	Increased IgM with low IgG and IgA	HIM
	Low IgM with increase IgG and IgA	Wiskott-Aldrich syndrome
	One Ig class high, others low	Multiple myeloma (IgG or IgA, Waldenström macroglobulinemia (IgM), HIM
	Increased IgA and IgE	Wiskott-Aldrich syndrome, late HIV-1
Specific antibody assays (ELISA)		
Hemagglutinin titers (anti-A, anti-B)	Low	Low IgM from any cause
Response to vaccine antigens and natural (infectious)	Low	Primary immunodeficiency (B and/or T cell)
Response after vaccination (protein and polysaccharide antigens)	Low	Overall B-cell dysfunction
Antipolysaccharide antibodies	Low	Selective unresponsiveness to encapsulated organisms, asplenia (+), IgG2 deficiency
Antibodies to HIV-1 (ELISA, immunoblot)	Positive	HIV-1 infection

*Normal number of total lymphocytes should be age-adjusted in children. Lymphopenia more often detects secure number of T cells.

CMD, chronic granulomatous disease; CLL, chronic lymphocytic leukemia; CVH, common variable hypogammaglobulinemia; ELISA, enzyme-linked immunosorbent assay; HIM, immunoglobulin deficiency with increased IgM; HIV, human immunodeficiency virus; SCID, severe combined immunodeficiency syndrome.

TABLE 51-6 Laboratory Results Suggestive of Potential Immunodeficiency

GASTROINTESTINAL SYNDROMES

Diarrhea

Diarrhea is the prototypical manifestation of intestinal disease in the immunocompromised host. Diarrhea is typically defined as the passage of three or more loose or watery stools per day for more than 2 days. In a healthy population, the vast majority of episodes of infectious diarrhea are acute in onset and self-limited in duration (<2 weeks). Chronic or prolonged diarrhea is defined as lasting 30 days or longer in adults (14 days in children). [22], [23] In the immunocompetent host, prolonged diarrhea is caused predominantly by noninfectious conditions, such as inflammatory bowel disease, celiac sprue, laxative abuse, endocrinopathies, malignancy, short bowel syndrome, eating disorders, functional illness, and surgically induced bacterial overgrowth. [24], [25] and [26] Even when a cause cannot be determined, diarrhea that lasts longer than 30 days in immunocompetent persons resolves spontaneously in most cases. [27] In contrast, in immunocompromised persons, an array of infectious pathogens are the most common cause of diarrheal illness; these are listed in Table 51-7.

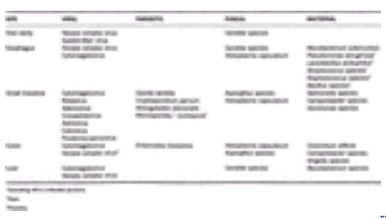

TABLE 51-7 Gastrointestinal Pathogens in Immunocompromised Subjects*

The presence of diarrhea in close contacts, such as family members, household contacts, sexual partners, or community members, suggests that the illness is caused by an environmentally acquired pathogen rather than an underlying immunodeficiency. For example, in 1986, an outbreak of prolonged diarrhea in otherwise healthy persons occurred in Brainerd, Minnesota. [28] No cause of the diarrhea could be determined in this and several similar outbreaks, referred to as *Brainerd diarrhea,* nor was immunodeficiency implicated as a predisposing factor. [23] In other outbreaks, epidemiologic evaluation resulted in the identification of a common pathogen. In this regard, the broad community involvement in outbreaks of diarrheal illness induced by *Cyclospora* [29], [30], [31], [32] and [33] and *Cryptosporidium* [34] did not support immunodeficiency as a factor in the vast majority of symptomatic patients. However, symptoms were more severe and persisted longer in the subset of individuals who were immunosuppressed. Thus, broad community exposure to certain pathogens may uncover underlying immunodeficiency in selected individuals. Similarly, although persistent diarrhea may develop in a small minority of travelers after their return, [35] chronic diarrhea may develop in immunocompromised persons after exposure to microorganisms such as *Cryptosporidium* during their travels.

Clinical clues also help distinguish diarrhea in persons with underlying immunodeficiency from diarrhea in otherwise healthy hosts. In the immunocompromised patient, diarrheal illness is frequently prolonged or recurrent and often quite severe, whereas in the immunocompetent host, it is typically self-limited and mild. In some patients, prolonged diarrhea is the initial clue suggesting the presence of an immunodeficiency disorder, such as HIV-1 infection. Chronic diarrheal illness in immunocompromised patients may be associated with vomiting and abdominal pain. For example, vomiting and abdominal cramps in bone marrow transplant recipients with diarrhea often predicts an infectious enteritis. [36] Other symptoms that may accompany diarrhea in an immunocompromised person include anorexia, nausea, vomiting, gastrointestinal bleeding, fever, weight loss, dehydration, and malnutrition. Bloody diarrhea with fever, although uncommon in the immunocompromised host, should suggest the possibility of cytomegalovirus (CMV) infection when cell-mediated immunodeficiency is present. [37], [38] and [39] Although bloody diarrhea is uncommon in persons with intact immune function, the leading cause of such diarrhea in immunocompetent persons is *Escherichia coli* O157:H7 infection, which appears not to occur with greater frequency in immunocompromised persons. [40] Children are particularly susceptible to the dehydration and malnutrition that accompany prolonged or severe diarrhea. Diarrhea that is associated with recurrent sinopulmonary, skin, oral, or blood infections is strong clinical evidence of an underlying primary immune deficiency. Another important clue that suggests the presence of an underlying immunodeficiency condition is persistence or relapse after appropriate antimicrobial therapy. Whereas the immunocompetent host usually clears an appropriately treated enteric illness, immunocompromised patients frequently experience recurrence of symptomatic infection despite therapy. Thus, recurrent *Giardia lamblia*–associated diarrhea after appropriate antimicrobial therapy suggests the possibility of underlying hypogammaglobulinemia. [41] Recurrent infections with *Salmonella* species, *Shigella* species, and *Campylobacter jejuni,* which are rare in immunocompetent persons, should suggest immunosuppression secondary to HIV-1 infection. [9], [10], [11], [12] and [13], [15], [16], [17], [18], [19], [20], [21], [22], [23], [24], [25], [26], [27], [28] and [29]

Chronic diarrhea greatly diminishes a person's sense of well-being and interferes with the activities of daily living, social interactions, and work. In HIV-1–infected patients, the presence of two loose or watery stools per day for at least 1 month is predictive of a marked reduction in the quality of life. [42] Infectious gastrointestinal disease in immunocompromised patients also may be associated with shortened survival. For example, the mortality rate of bone marrow transplant recipients with infectious gastroenteritis is fourfold higher than that of uninfected patients. [36] Among liver transplant recipients, CMV disease involving any organ is associated with a fourfold increase in the relative risk for death within 1 year after transplantation. [43] However, because gastrointestinal disease in immunocompromised persons is in itself rarely fatal, the associated shortened survival likely reflects the severity and consequences of immune dysfunction, such as associated bacterial and fungal sepsis. [44], [45], [46] and [47] Similarly, HIV-1–infected infants have an 11-fold increased risk for death from diarrhea, which is usually persistent and often preceded by recurrent episodes of acute diarrheal illness. [48] In HIV-1–infected adults, CMV infection of the colon can lead to necrotizing colitis, perforation, and death. [49], [50], [51] and [52] Of note, most infections related to HIV-1 and the acquired immunodeficiency syndrome (AIDS), including CMV and *Cryptosporidium* infections and nonspecific chronic diarrhea, have decreased dramatically in the United States and Europe since the introduction of highly active antiretroviral therapy (HAART). [53], [54] and [55] Drug-related side effects of HAART are now among the most common causes of enteric symptoms in this population (reviewed in ref. [53]).

Oropharyngeal and Esophageal Symptoms

In the immunosuppressed patient, discomfort, pain, and burning sensations in the oral cavity are important symptoms of oropharyngeal opportunistic infections. The most common pathogens causing such symptoms are *Candida albicans,* herpes simplex virus (HSV), and Epstein-Barr virus (EBV). *C albicans* causes (1) pseudomembranous, exudative plaques (thrush) on any oral tissue; (2) erythematous, atrophic lesions on the palate and tongue; (3) adherent hyperplastic lesions (hypertrophic candidiasis); and (4) erythematous fissures at the corners of the mouth (angular cheilitis). [56] These lesions are easily distinguished from HSV vesicular gingivostomatitis [57], [58] and EBV hairy leukoplakia (unilateral or bilateral adherent, white or gray patches on the lateral margins of the tongue and less commonly other regions of the tongue). [59] These oropharyngeal infections are associated with a spectrum of immunosuppressive states and disorders, notably chemotherapy, AIDS, and malignancy.

Dysphagia and odynophagia in the immunocompetent person are usually caused by acid peptic disease, malignancy, or neuromuscular disease. However, in the immunocompromised patient, dysphagia and odynophagia most often indicate the presence of an esophageal infection. Either of these symptoms, frequently both, affects the majority of patients with infectious esophagitis secondary to *Candida* species, HSV, CMV, *Mycobacterium* species, and HIV-1. [60], [61] and [62] Neither dysphagia nor odynophagia, however, is specifically associated with a particular infection [8], [63]; therefore, these symptoms cannot be used to differentiate one esophageal infection from another. In addition, retrosternal chest pain may accompany infectious esophagitis in immunocompromised persons, but this symptom is also nonspecific.

In the immunocompromised patient with dysphagia or odynophagia, the presence of oropharyngeal thrush or HSV gingivostomatis suggests the presence of *Candida* or HSV esophagitis, respectively. In four series of HIV-1–infected patients, between 71% and 100% of those with thrush had *Candida* esophagitis. [64], [65], [66] and [67] In the largest series, which consisted of 110 patients, the presence of thrush had a sensitivity of 53% and a positive predictive value of 77% for *Candida* esophagitis. [67] However, symptomatic infection of the two sites can occur independently. Symptomatic HSV infection of the oropharynx, presenting as either vesicular lesions or ulcerative gingivostomatitis, [68], [69] has been reported in 29% of patients with HSV esophagitis. [60] Heartburn may accompany infectious esophagitis but is less frequent and should suggest gastroesophageal reflux disease. Thus, because symptomatic infectious esophagitis occurs rarely in immunocompetent persons with normal

esophageal structure and function, [70], [71], [72], [73] and [74] the development of infectious esophagitis in a previously healthy subject is strong clinical evidence that an underlying immunodeficiency condition is likely and that the patient should undergo an immunologic evaluation. An exception to the association between infectious esophagitis and an immunodeficiency disorder is the presence of *Candida* esophagitis in persons with severe insulin-dependent diabetes mellitus, which may reflect hyperglycemia-induced phagocyte dysfunction. [75] Infectious esophagitis in the immunocompromised patient also may be accompanied by nonspecific gastrointestinal symptoms, such as nausea and vomiting. [60] These symptoms occur far more commonly in patients with CMV and HSV esophagitis than in those with fungal or bacterial esophagitis. [60], [76] An additional clue that the esophagitis may be caused by CMV is an association of abdominal pain and diarrhea with the esophageal symptoms. [60], [76], [77] When dysphagia or odynophagia occurs in an immunocompromised patient with cough, fever, and weight loss, esophagitis secondary to mycobacterial infection should be considered. [78], [79] and [80] Food aversion and weight loss may accompany prolonged esophageal symptoms, particularly in cases of CMV and mycobacterial esophagitis, which are less responsive to therapy than *Candida* and HSV esophagitis.

Gastrointestinal Bleeding

Gastrointestinal bleeding occurs infrequently in immunocompromised patients. The most common cause of esophageal bleeding in such patients is HSV-induced mucosal ulceration. In a review of 100 HIV-1–infected patients with esophageal disease, 6% of the patients experienced esophageal bleeding, which in the majority of patients was caused by HSV-induced mucosal ulceration. [61] Up to 30% of immunocompromised patients with HSV esophagitis are reported to have esophageal bleeding. [81] Typically, bleeding from HSV esophagitis is slow and limited, but massive bleeding has been reported. [82] Rarely, bleeding secondary to HSV esophagitis may occur in the absence of esophageal symptoms. [83] Esophageal bleeding presenting as hematemesis has been reported in patients with esophagitis caused by CMV, [83], [84] but bleeding secondary to CMV esophagitis is rare, at least in patients with AIDS. [61], [62], [77] Although pathogens other than CMV, HSV, and *Mycobacterium* species seldom cause esophagitis in immunocompromised patients (see Table 51-7), *C albicans* has been reported to cause esophageal bleeding, albeit rarely. [85], [86]

Intestinal bleeding in an immunocompromised patient should alert the physician to the possibility of infectious enteritis or colitis. Similar to esophageal bleeding, intestinal bleeding in immunocompromised persons occurs infrequently. However, in contrast to esophageal bleeding, which is most commonly caused by HSV, intestinal bleeding in immunocompromised persons, such as patients with AIDS, is more commonly caused by CMV. [49], [87], [88] Bleeding from CMV-induced intestinal ulcerations is typically slow and rarely severe. [89] In a prospective study of 44 HIV-1–infected patients with CMV colitis, bleeding occurred in 10% of them. [88] Similarly, among ten heart and heart-lung transplant recipients with CMV gastrointestinal disease, two patients had endoscopically detected gastric bleeding and another had intractable colonic hemorrhage. [90] In solid organ transplant recipients, the highest risk for the development of significant CMV-induced complications occurs between the fourth and twelfth weeks after transplantation, especially in those who receive organs from CMV-seropositive donors. [91], [92] The possibility that intestinal bleeding in an immunocompromised person is caused by CMV is increased when diarrhea, abdominal pain, and fever accompany the bleeding. In the immunocompromised patient who is neutropenic, bloody diarrhea should raise the possibility of typhlitis or necrotizing enterocolitis. [93], [94] and [95]

Abdominal Pain

Abdominal pain frequently accompanies intestinal and colonic infections, particularly when intense mucosal inflammation is present. However, abdominal pain is nonspecific and is not helpful in distinguishing one type of enteric infection from another. Like other clinical manifestations of infection, abdominal pain may be diminished in immunosuppressed patients receiving cytotoxic or corticosteroid agents. Immunosuppressed patients are susceptible to the same common causes of abdominal pain as immunocompetent persons, including peptic ulcer disease, biliary tract disease, appendicitis, pancreatitis, diverticulitis, and inflammatory bowel disease. As summarized in Table 51-8, immunocompromised patients also experience abdominal pain as a result of immunosuppression-associated processes, including CMV disease, perforation secondary to neoplasm or infection, typhlitis, cholecystitis caused by opportunistic pathogens, chemotherapy-induced bleeding, pancreatitis caused by certain drugs, hepatosplenic candidiasis, and tissue infiltration by tumor or lymphoma. In the setting of compromised host defense mechanisms, these processes can progress more rapidly to a medical catastrophe than they can in immunocompetent persons, which underscores the importance of early diagnosis and therapy.

IMMUNOCOMPETENT AND IMMUNOCOMPROMISED	IMMUNOCOMPROMISED ONLY
Peptic ulcer disease	Cytomegalovirus disease
Biliary tract disease	Colonic perforation caused by cytomegalovirus, *Candida* species, *Histoplasma capsulatum*, lymphoma
Appendicitis	Typhlitis
Pancreatitis	Cholecystitis caused by cytomegalovirus, *Cryptosporidium parvum*, *Isospora belli*, *Microsporidium* species, *Pneumocystis carinii*
Diverticulitis	Bleeding secondary to cytotoxic drug-induced thrombocytopenia
Inflammatory bowel disease	Pancreatitis caused by immunosuppressive and antiviral drugs
Colonic perforation	Graft-versus-host disease
	Hepatosplenic candidiasis
	Tissue infiltration by tumor or lymphoma
	Venoocclusive liver disease

TABLE 51-8 Causes of Abdominal Pain in the Immunocompromised Host

Enteric disease caused by CMV is frequently accompanied by abdominal pain in immunocompromised patients, particularly HIV-1–infected patients and transplant recipients. Such pain is often described as constant, intense cramping in the lower quadrants [49], [90] and differs in quality from the pain associated with biliary tract disease and intestinal obstruction, which typically fluctuates in intensity. Severe epigastric pain from CMV pancreatitis has been reported in HIV-1–infected patients. [96] When abdominal pain in a patient with HIV-1 infection suddenly increases, CMV-induced perforation of the colon or distal small intestine should be included in the differential diagnosis. [50], [97], [98] Other causes of perforation in immunocompromised persons include diverticular disease, [99] lymphoma, [100], [101], [102], [103] and [104] and, rarely, *Candida* species [103], [104] and *Histoplasma capsulatum* infections. [105] Typhlitis or necrotizing enterocolitis, as discussed below, should also be included in the differential diagnosis of right lower abdominal pain in neutropenic patients. [106]

Cholecystitis and cholangitis occur in immunocompromised patients secondary to cholelithiasis and the same gram-negative pathogens that cause biliary tract disease in immunocompetent persons. However, in HIV-1–infected patients, these conditions, as well as sclerosing cholangitis and papillary stenosis, are also associated with biliary tract infection by *Cryptosporidium,* CMV, *Enterocytozoon bieneusi, Septata intestinalis, Isospora belli,* and *Pneumocyctis carinii*. [107], [108], [109], [110] and [111] The presence of thrombocytopenia expands the differential diagnosis of abdominal pain in the immunocompromised patient because platelet counts below 20,000/mm ^{3} can cause bleeding into the intestinal wall, retroperitoneum, or abdominal wall. [112], [113] Drug-induced pancreatitis should be considered in immunosuppressed patients on L-asparaginase, the antiretroviral agent didanosine, or corticosteroids. HIV-1 protease inhibitors may be associated with renal stones and secondary abdominal pain. Hepatosplenic candidiasis occurs when the liver is seeded with *Candida* species during disseminated candidiasis in neutropenic patients, usually as a result of therapy for acute leukemia or lymphoma. [114] Typically, patients have right upper quadrant pain and tenderness accompanied by nausea and fever. Venoocclusive disease of the liver is an important consideration when a patient presents within 20 days after bone marrow transplantation with right upper quadrant pain, tenderness, hepatomegaly, hyperbilirubinemia, and sudden weight gain secondary to ascites. [115], [116] and [117] High-dose cytoreductive therapy and underlying liver disease increase the risk for the development of severe veno-occlusive disease, which is reported to occur in 15% of marrow transplant patients. [117]

Anorectal and Perianal Disease

Anorectal disease is one of the most important gastrointestinal complications of immunosuppression, affecting 5% to 7% of neutropenic patients with leukemia or lymphoma. [118], [119] The importance of this complication is underscored by the devastating morbidity and high level of mortality in unrecognized and inappropriately treated cases of anorectal disease in immunocompromised patients. The various manifestations of anorectal disease in neutropenic patients include abscess, fissure,

fistula, and infected hemorrhoids. Typically, lesions present as perianal pain, tenderness, induration, and fever, usually without fluctuation or purulence. Extensive tissue necrosis and breakdown have been reported in many patients. [118] Frequent clinical evaluation is mandatory because lesions can progress rapidly, causing urinary retention and peritonitis, and fascial extension to the genitalia is possible. [118] Culture of material obtained from the lesions by either surgical drainage or needle aspiration usually yields multiple bacteria, with anaerobes the most common organisms. [118], [119] and [120] The high rates of recurrence (12%) and mortality (18%) associated with perianal disease in neutropenic patients managed nonoperatively emphasize the importance of surgical drainage. [119] Unless drainage occurs spontaneously, early operative incision, drainage, and debridement have been associated with prompt clinical improvement and increased survival. [118], [119], [121], [122] and [123] One group, however, has achieved control of anorectal infections in 88% of neutropenic patients with an antibiotic regimen that includes both an aminoglycoside and an agent with anaerobic coverage. [120] Thus, a reasonable approach to the management of anorectal disease in neutropenic patients is to institute immediately a trial of antibiotic treatment with an aminoglycoside and an antibiotic having specific activity against anaerobes during the initial medical and surgical evaluation. When fluctuation, necrosis, local progression of infection, or continued fever is present or when an abscess fails to drain spontaneously, the lesion should be promptly incised, drained, and debrided.

Perianal ulcerative disease can be an incapacitating problem in patients with cell-mediated immunodeficiency. In most patients, this problem is caused by HSV infection, which typically begins as small vesicular eruptions that coalesce and progress to frank ulceration. [124] The lesions are painful and macerated, and they may become secondarily infected. Ulcerative perianal lesions caused by HSV were among the infectious processes that heralded the emergence of AIDS. [125] The infection generally responds to acyclovir, but long-term therapy is often necessary. [126]

SPECIFIC IMMUNODEFICIENCY SYNDROMES AND PATHOGENS

Spectrum of Infections

The risk for acquiring a specific enteric pathogen and the type of pathogen acquired are directly related to the type of immune defect (see Table 51-1). The severity and duration of the immune defect also influence host susceptibility to enteric infections. These infections, which involve predominantly the esophagus, small intestine, and colon and less frequently the liver and oral cavity, are caused by the viral, parasitic, fungal, and bacterial pathogens listed in Table 51-7. Traditionally, infectious agents are regarded as opportunistic pathogens when they consistently cause severe, chronic, or frequent gastrointestinal disease in immunocompromised but not immunocompetent persons, and as nonopportunistic pathogens when they cause treatable infections in immunocompetent and immunocompromised persons. However, because many nonopportunistic pathogens, such as *Salmonella* species, *Shigella* species, and *C jejuni,* cause persistent or recurrent infection in immunocompromised persons, the distinction is not particularly helpful in assessing gastrointestinal infections in this population.

Three pathogens, *G lamblia, Cyclospora cayetanensis,* and *Clostridium difficile,* are noteworthy because they can cause prolonged diarrhea in immunocompetent as well as immunosuppressed persons, and they therefore do not necessarily suggest the presence of an immunodeficiency. Infection with the protozoan *G lamblia* is most often associated with diarrhea, abdominal cramps, bloating, flatulence, and weight loss. A history of exposure to mountain, lake, or stream water, close contact with children in day care centers, or recent travel is common. *G lamblia* also is associated with hypogammaglobulinemia (see below). *Cyclospora,* another protozoan parasite, causes prolonged diarrhea in immunocompetent persons and is endemic in Peru [30], Nepal, and the Caribbean. [29], [127], [128] *Cyclospora* is associated with prolonged diarrhea in AIDS patients. Patients with *C difficile* infection nearly always have a history of recent treatment with antibiotics or chemotherapy, particularly older or hospitalized patients. Although treatment with metronidazole is effective in more than 90% of cases, up to 15% of immunocompetent patients relapse and require repeated treatment, particularly if the inciting antimicrobial agents are continued. [129] In contrast, high rates of relapse with *Salmonella* species and *Shigella* species are indicative of an impaired immunologic response, as in HIV-1–infected persons. [130] Relapse of *C jejuni* infection also occurs in HIV-1–infected patients and patients with hypogammaglobulinemia. [5], [6] and [7], [131] Other bacterial, viral, and parasitic infections rarely cause chronic diarrhea in immunocompetent persons.

In the esophagus, the most commonly identified pathogens in immunocompromised persons are *C albicans,* HSV, and CMV. Two or more pathogens are often present, as has been reported in 20% of HIV-1–infected patients with symptomatic esophagitis. [8] Much less common causes of infectious esophagitis in immunocompromised patients are certain bacteria, [132] including *Mycobacterium tuberculosis,* [133], [134] *Streptococcus* species, [135] and *Lactobacillus acidophilus.* [136] Bacterial esophagitis has been described in bone marrow transplant recipients and in patients with hematologic malignancies, neutropenia, and HIV-1 infection and can be an infrequent source of occult sepsis. Esophagitis caused by *Cryptosporidium* species, [137] *Leishmania* species, [138] and *H capsulatum* [139] has been reported in HIV-1–infected persons but is rare. Fortunately, this persistent pathogen can be treated effectively.

Specific Syndromes

Hypogammaglobulinemia Common variable hypogammaglobulinemia (CVH), also referred to as *common variable immunodeficiency,* represents a group of uncommon (<1 per 100,000 population) disorders usually inherited in an autosomal manner. CVH is characterized by the inability of B cells to differentiate normally into immunoglobulin-secreting cells, which results in a marked reduction in the levels of IgG, IgM, and IgA and immunologic abnormalities that include autoimmune conditions and lymphoproliferative disorders. [140] Recurrent sinopulmonary infections far outnumber intestinal infections in persons with CVH. Atrophic gastritis with achlorhydria may predispose to enteric disease, often with malabsorption. Unlike congenital X-linked agammaglobulinemia, CVH typically manifests during the second or third decade, occurs equally often in men and women, and is infrequently associated with severe enteroviral infections. Among patients with CVH, *G lamblia* infection is the most common cause of symptomatic diarrheal illness. [41], [141] The parasite is identified in up to 90% of hypogammaglobulinemic subjects with chronic diarrhea, but in only 10% of those without diarrhea. [41] Infection with *C jejuni* and *Salmonella* species and bacterial overgrowth are also causes of recurrent diarrheal illness in patients with CVH. Children with hypogammaglobulinemia and chronic diarrhea also have a higher incidence of intestinal infections with *G lamblia, C jejuni, C difficile,* and rotavirus. [5], [142] Importantly, increased rates of *G lamblia* infection are not associated with selective IgA deficiency [41], [141] or cell-mediated immune deficiencies, including AIDS. [143], [144] and [145] The relapse rate for giardiasis after antimicrobial therapy is approximately 15% in both immunocompetent subjects and patients with AIDS, [143] which contrasts with the substantially higher relapse rate in hypogammaglobulinemic subjects. [146] Chronic giardiasis also occurs commonly in patients with nodular lymphoid hyperplasia of the small intestine and immunoglobulin deficiency. [147], [148] and [149] *G lamblia*–associated diarrhea in hypogammaglobulinemic patients is prolonged, with one reported case lasting 10 years, [150] and is often accompanied by malaise, nausea, anorexia, weight loss, and steatorrhea. Thus, prolonged infection with *G lamblia,* the presence of *G lamblia* and other enteric bacterial pathogens or rotavirus, and recurrent giardiasis after antimicrobial therapy should suggest the possibility of underlying hypogammaglobulinemia. Selective IgA deficiency is the most common disorder of immunoglobulin production, affecting between 1 per 500 and 1 per 1000 population, but associated intestinal disease is uncommon. X-linked agammaglobulinemia is the most severe immunoglobulin disorder. Children with this B-cell deficiency experience very high rates of chronic *G lamblia* infection as well as devastating systemic manifestations of chronic enteroviral infections. The latter include enteroviral meningoencephalitis and paralytic poliomyelitis following either natural infection or oral immunization with live vaccine. Among the fewer than 150 cases of immunoglobulin deficiency with hyper-IgM, up to half experience chronic diarrhea, in part related to *Giardia* and *Cryptosporidium* infection. [151] Patients with selective IgG subclass deficiencies, particularly decreased IgG2, often show impaired responses to polysaccharide antigens and an increase in sinopulmonary infections and disease but no appreciable predisposition to intestinal infections.

Transplantation-Associated Gastrointestinal Illness Gastrointestinal disease is an important cause of morbidity and mortality in recipients of bone marrow and solid organ (kidney, liver, heart, lung) transplants. Marrow transplant recipients experience more frequent and more severe gastrointestinal disease than do solid organ recipients. This difference is mainly a consequence of the more profound neutropenia and cell-mediated immune deficiency and the occurrence of graft versus host disease in marrow recipients. In the largest study to date of the incidence, etiology, and outcome of diarrheal illness after marrow transplantation, 43% of 126 transplant recipients experienced an acute diarrheal illness. [152] Graft-versus-host disease accounted for 48% of the diarrheal illnesses, whereas intestinal infections with astrovirus, adenovirus, CMV, rotavirus, *C difficile,* or *Aeromonas* accounted for 13% of the illnesses; no cause could be identified in 39% of the cases. Acute graft-versus-host disease usually begins 3 to 4 weeks after transplantation and presents as anorexia, nausea, vomiting, watery diarrhea and abdominal pain, and gastrointestinal bleeding. [153], [154] The latter may involve the esophagus, stomach, and small intestine and is usually caused by CMV infection and vascular ectasias (stomach). [155] When rash and jaundice are also present, graft-versus-host disease is likely. However, because the gastrointestinal symptoms are nonspecific, the diagnosis of acute graft-versus-host disease is confirmed by histological examination of endoscopically obtained biopsy samples, which typically show single-cell apoptosis of the epithelium, crypt distortion and abscesses, and a periglandular infiltrate. [156], [157] Chronic graft-versus-host disease typically occurs 3 to 12 months after marrow transplantation. The major gastrointestinal manifestations of chronic graft-versus-host disease include mucositis of the oral cavity, dysphagia secondary to esophageal desquamation and stricture, and small bowel dysmotility associated with bacterial overgrowth. [158], [159] and [160] In contrast to marrow recipients, solid organ recipients experience gastrointestinal complications predominantly secondary to infections. Among lung transplant recipients, for example, operative and nonoperative gastrointestinal complications developed in 51%. [161] The nonoperative complications included esophagitis, gastritis, colitis, enteritis, peptic ulcer disease, gastrointestinal bleeding, cholecystitis, diverticulitis, and Ogilvie syndrome; 65% of these processes were caused by infections, excluding cholecystitis and diverticulitis. The frequency and severity of infections diagnosed in transplant recipients vary considerably, depending on the factors listed in Table 51-9. As

previously stated, the enteric infections of bone marrow transplant recipients are more frequent and more severe than those of solid organ transplant recipients. The length of time since the transplant operation is closely related to the development of gastrointestinal complications. [152], [161], [162] During the first month after transplantation, marrow recipients are vulnerable to fungal and bacterial infections as a consequence of cytoreductive therapy, whereas during the same period, organ recipients are more susceptible to the complications of surgery and surgery-related procedures. One month after marrow transplantation, acute graft-versus-host disease should be included in the differential diagnosis of diarrhea, as discussed previously. Between 1 and 6 months after transplantation, both marrow and solid organ recipients are highly susceptible to gastrointestinal infections caused by viruses, including CMV, adenovirus, HSV, varicella-zoster virus, and rotavirus (children); fungi, such as *Candida* species and *H capsulatum* [163], [164]; bacteria, particularly *C difficile*; and less commonly parasites, such as *Cryptosporidium*. [165], [166] and [167] CMV disease is an important complication of transplantation, affecting up to 25% of organ transplant recipients [47], [91], [92]; it causes major gastrointestinal morbidity [90] and is associated with a marked increase in mortality among transplant recipients. [43] The presence of graft-versus-host disease in marrow transplant recipients also significantly increases the risk for CMV disease. [168] After 6 months, gastrointestinal manifestations of chronic graft-versus-host disease may develop in marrow and organ transplant recipients. The susceptibility to an array of enteric pathogens also increases after 6 months (see Table 51-7). In addition, marrow and organ transplant recipients are susceptible to the gastrointestinal complications of chemotherapy and radiation therapy and to typhlitis.

Type of transplantation
Time since transplantation
Prevalence of potential pathogens in the environment
Prevalence of latent infections in the donor and host before transplantation
Level of immunosuppression
Availability of diagnostic tests
Rigor of diagnostic evalution for enteric pathogens

TABLE 51-9 Factors that Influence the Development and Management of Gastrointestinal Infections in Transplant Recipients

Pathogens that are endemic in certain regions may be more common in transplant recipients. For example, *Strongyloides stercoralis,* which is more prevalent in the southeastern part of the United States, has been reported in renal transplant recipients in that region. [169], [170] Similarly, *H capsulatum* infections are more common in transplant recipients in the midwestern and southeastern parts of the United States. Although amebiasis and malaria are common in immunocompetent persons in the tropics, *Entamoeba histolytica* and *Plasmodium falciparum* infections do not appear to be more common in organ (renal) transplant recipients in the same region. [171] In addition, the prevalence of latent infections in the donor and host before transplantation strongly influences the development of infection by the same pathogen after transplantation. For example, among marrow and liver transplant recipients, the rates of symptomatic CMV infection are highest when the donor is seropositive and lowest when both the donor and recipient are seronegative. [172], [173] and [174] The relative risk for death 1 year after transplantation is affected by the presence of latent CMV infection. One year after liver transplantation, the relative risk for death was highest (4.56) when the donor was seropositive for CMV and the recipient seronegative. [43] In addition, the more profound the immunosuppression, as in marrow transplant recipients, the more severe the infections. The rate of infectious complications is also affected by the type of immunosuppressive regimen. Compared with heart transplant recipients who received conventional immunosuppressive therapy, recipients who received cyclosporine had lower rates of complications and morbidity from CMV and HSV infection. [91] Finally, the availability of diagnostic tests and the rigor with which patients are evaluated for the presence of enteric pathogens strongly influence the detection of gastrointestinal pathogens in immunocompromised patients.

Neutropenia A reduction in the absolute number of circulating neutrophils to less than 1000/mm [3] is associated with increased susceptibility to infections. When the level of neutrophils declines to less than 500/mm [3], the normal host defense against endogenous gastrointestinal flora is substantially impaired. Neutropenic patients are far more likely to present with fever and sepsis caused by gram-negative bacteria and with fungemia than with primary gastrointestinal syndromes. Importantly, the intestinal flora is the source of most cases of bacteremia in neutropenic patients. [175], [176] Because the risk for the development of bacteremia in neutropenic patients colonized with potential gram-negative bacterial pathogens is 17- to 174-fold greater than the risk in noncolonized patients, some clinicians advocate surveillance cultures of mucosal and other surfaces. [175] Three gastrointestinal syndromes may complicate neutropenia: oral infections, anorectal infections, and typhlitis. The oral manifestations of neutropenia include gingivitis, periodontitis, and oral ulcers. These complications occur most commonly when the neutrophil count declines below 500/mm [3]. [177], [178], [179], [180] and [181] Anorectal disease is an important and life-threatening complication of neutropenia and has been discussed previously. Typhlitis, an important cause of abdominal pain in neutropenic patients, is discussed next.

Typhlitis Typhlitis, also referred to as *necrotizing enterocolitis, neutropenic enterocolitis,* and the *ileocecal syndrome,* is an infrequent complication of chemotherapy-induced neutropenia in patients with leukemia or lymphoma. [93], [94] and [95], [106], [182], [183] In a minority of cases, the neutropenia is caused by the hematologic disease itself, such as aplastic anemia or multiple myeloma. A clinical condition consistent with typhlitis also has been reported in HIV-1–infected patients, except that neutropenia may be absent. [184], [185] Typically, patients with typhlitis present with right lower quadrant pain, diarrhea, lower gastrointestinal bleeding, and fever. Typhlitis has been associated with *Clostridium septicum* sepsis in several cases, [93], [94], [186] suggesting that local infection with this gas gangrene–producing bacterium may be involved in the pathogenesis of the syndrome in some patients. The syndrome should be recognized early because the characteristic cecal inflammation may progress to life-threatening necrosis and perforation, in which case surgical intervention is imperative. [187], [188], [189] and [190] However, some patients with typhlitis, particularly those with HIV-1 infection, have been managed conservatively with broad-spectrum antibiotics, bowel rest, fluids, and close radiographic and computed tomographic monitoring. [185]

***Candida* Diarrhea** *Candida* species are present in the gastrointestinal tract of most immunocompetent adults, and elderly persons appear to harbor more *Candida* species than younger persons. [191], [192] In the vast majority of such individuals, the fungus is a nonpathogenic commensal. Although pathogenic gastrointestinal candidiasis was previously thought to be associated exclusively with the mycelial phase of the organism, [193] accumulating clinical evidence suggests that the yeast phase may be associated with a diarrheal illness in certain clinical settings. [194], [195], [196], [197], [198], [199] and [200] Patients with *Candida*-associated diarrhea are usually elderly, malnourished, and either critically or chronically ill and do not have other detectable enteric pathogens. The diarrhea is typically prolonged, of the secretory type (volumes often exceed 2 L/d), nonbloody, nonmucoid, and without accompanying symptoms of fever, nausea, or vomiting. [200] Many patients with *Candida*-associated diarrhea have a history of treatment with multiple antibiotics or chemotherapy. Tetracycline therapy in particular is associated with high rates of gastrointestinal colonization with *Candida,* but colonization in this setting is not consistently associated with diarrheal illness. In patients with *Candida*-associated diarrhea and no other detectable pathogens, nystatin or fluconazole therapy may be associated with clearing of the *Candida* and usually, but not invariably, resolution of symptoms. [197], [200], [201]

Small Bowel Bacterial Overgrowth In healthy immunocompetent persons, proximal small intestine fluid usually contains 10 [4] bacteria per milliliter or less; these are predominantly gram-positive aerobes and are similar to the microflora of the stomach and oral cavity. [202], [203] and [204] The presence of small bowel bacterial overgrowth, defined as more than 10 [4] predominantly gram-negative anaerobes of colonic origin per milliliter of small bowel fluid, is a well-recognized complication of small intestinal anatomic abnormalities, motility disorders, reduced acid secretion, and chronic pancreatitis and cirrhosis. [205] Small bowel bacterial overgrowth also has been reported in two populations of immunocompromised or potentially immunocompromised hosts: persons with hypogammaglobulinemia and the elderly. Studies performed before the widespread availability of sophisticated microbiologic techniques showed that some patients with hypogammaglobulinemia were affected with small bowel bacterial overgrowth. [206], [207], [208], [209], [210] and [211] Among such patients, however, the diagnosis of bacterial overgrowth was confounded by several factors: clinical conditions (e.g., achlorhydria and anatomic abnormalities) associated with excessive numbers of bacteria in the proximal small intestine, [209], [210] variations in the rigor with which fluids were anaerobically collected, [206], [207] and [208] and a definition of bacterial overgrowth based on excessive numbers of oral cavity–type bacteria in some cases. [210] Selective IgA deficiency appears not to be associated with bacterial overgrowth by colonic anaerobes. [209], [210] and [211] Among elderly subjects, small bowel bacterial overgrowth with colonic-type bacteria has been detected in 10% of subjects 50 to 74 years of age and in 64% of subjects older than 75 years of age with chronic diarrhea, anorexia, or nausea and no predisposing condition. [212] Malabsorption and undernutrition attributed to small bowel bacterial overgrowth in the elderly [213], [214] and [215] have been effectively corrected with antibiotic therapy. [216] In some elderly subjects, however, small bowel bacterial overgrowth may be a benign condition that does not warrant treatment. [217], [218] Nevertheless, the presence of bacterial overgrowth in some elderly persons without other predisposing conditions may reflect the immune dysfunction that sometimes accompanies advanced age. Regarding the mucosal immune system, such dysfunction includes altered differentiation or homing of antigen-stimulated B cells, impaired initiation or regulation of local antibody production, and a decline in macrophage production of certain effector products (e.g., nitric oxide), collectively referred to as *immunosenescence*. [219], [220] and [221] Thus, larger studies are needed to characterize small bowel bacterial overgrowth fully in subjects who have hypogammaglobulinemia or are at an advanced age and to determine the most sensitive and specific noninvasive tests for clinically diagnosing significant overgrowth. Until such information is available, small bowel bacterial overgrowth should be included in the differential diagnosis of chronic diarrhea in persons who have immunoglobulin deficiency or are at an advanced age without detectable enteric pathogens and treated with appropriate antibiotics when the syndrome is present.

SUMMARY

In summary, gastrointestinal complications are a major problem in patients with primary (genetically determined) and secondary (acquired) immunodeficiency diseases and conditions. The most important of these complications include esophagitis, infectious diarrhea, gastrointestinal bleeding, typhlitis, and anorectal

disease, which can be severe and, in some cases, life-threatening. The dramatic increase in the number of persons with primary and secondary immunodeficiency diseases increases the likelihood that physicians in all specialties will encounter immunocompromised patients with gastrointestinal problems. Consequently, familiarity with the gastrointestinal manifestations of the various immunosuppressive diseases and conditions should facilitate prompt diagnosis and, when possible, effective treatment.

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CHAPTER 52

Ellen M. Zimmermann and Gregory M. Christman

APPROACH TO GASTROINTESTINAL AND LIVER DISEASES IN THE FEMALE PATIENT

Gastrointestinal diseases in women, particularly in pregnant female patients, have unique diagnostic and therapeutic implications and warrant specific consideration. Diseases such as hyperemesis gravidarum and intrahepatic cholestasis of pregnancy are unique to pregnancy, whereas other common gastrointestinal diseases, including gastroesophageal reflux disease, may be more severe during pregnancy. Chronic diseases such as inflammatory bowel disease often initially present in women of childbearing age. In addition, it is important to consider the impact of pregnancy on the course and therapy of preexisting chronic gastrointestinal and liver diseases even before conception. This chapter reviews the gastrointestinal diseases associated with pregnancy and the gynecologic diseases relevant to the evaluation of gastrointestinal symptoms.

PHYSIOLOGY OF PREGNANCY

Physiological Changes in Normal Pregnancy

Profound physiological changes occur during pregnancy and affect nearly every bodily system (Table 52-1). The complex hormonal changes that occur during pregnancy are key to understanding the physiological alterations associated with pregnancy. Three hormones are relevant to changes in gastrointestinal physiology (Fig. 52-1). Human chorionic gonadotropin (hCG) is initially derived from the primitive trophoblast and later from the placenta. [1] Levels of serum hCG peak in the first trimester. Measurement of the ß subunit of hCG is widely used as a conventional immunologic pregnancy assay. hCG plays a major role in maintaining the corpus luteum of the menstrual cycle until the placenta is capable of synthesizing adequate progesterone to support pregnancy. [1] hCG may also regulate steroid production in the fetus and in the placenta [2], [3] and appears to have immune-modulating and thyroid-stimulating activities. [4] Because levels of serum hCG peak in the first trimester, when nausea is common, it has been implicated in the pathogenesis of nausea and vomiting of pregnancy and hyperemesis gravidarum, although direct evidence supporting a pathogenic mechanism is lacking. [5]

SYSTEM	CHANGE IN PREGNANCY	COMMENTS
Metabolic System		
Basal body temperature	Increase	
Oxygen consumption	Increase	20%–30% increase
Cardiovascular System		
Cardiac output	Increase	30%–50% increase; peak effect at 20–24 weeks of gestation
Stroke volume	Increase	
Heart rate	Increase	
Peripheral resistance	Decrease	
Blood volume	Increase	40%–50% increase; mostly plasma
Plasma volume	Increase	45%–55% increase; dilutional anemia common
Red cell mass	Increase	20%–30% increase
Urinary System		
Renal pelvic diameter	Increase	Hydronephrosis common
Ureteral diameter	Increase	Hydroureter common
Glomerular filtration rate	Increase	30%–50% increase; peak second trimester
Renal blood flow	Increase	40% increase
Respiratory System		
Respiratory rate	No change	
Total lung capacity	No change	Volumes redistributed second half of pregnancy
Tidal volume	Increase	By 200 mL
Residual volume	Decrease	By 300 mL because of enlarging uterus
Gastrointestinal System		
Lower esophageal sphincter tone	Decrease	Progressive decrease to term
Gastric emptying time	Increase	
Intestinal transit time	Increase	
Colonic transit time	Increase	
Gastric acid secretion	Increase	
Hepatobiliary System		
Gallbladder emptying time	Increase	
Liver size	No change	
Liver histology	Subtle change	Subtle, nonspecific, including cytoplasmic fat vacuoles

TABLE 52-1 Physiological Changes during Pregnancy

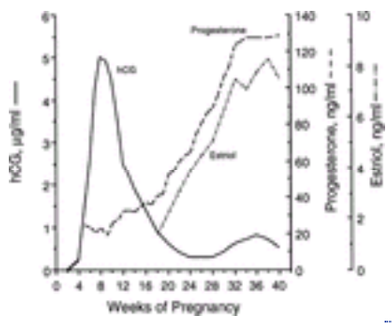

FIGURE 52-1. Changes in the gestational hormones human chorionic gonadotropin (hCG), progesterone, and estriol during pregnancy. (Data from Jaffe RB. Protein

hormones of the placenta, decidua, and fetal membranes. In: Yen SSC, Jaffe RB, eds. Reproductive endocrinology: physiology, pathophysiology and clinical management, 2nd ed. Philadelphia: WB Saunders, 1986:761. Johansson ED. Plasma levels of progesterone in pregnancy measured by a rapid competitive protein binding technique. Acta Endocrinol (Copenh) 1969;61:607. Tulchinsky D, Hobel CJ, Yeager E, et al. Plasma estrone, estradiol, estriol, progesterone, and 17–hydroxyprogesterone in human pregnancy. I. Normal pregnancy. Am J Obstet Gynecol 1972;112:1095.)

Progesterone and estrogen are the major steroid hormones synthesized during pregnancy. Progesterone is synthesized from cholesterol precursors initially by the ovary and later by the placenta. Several estrogens are synthesized by the placenta, including estradiol, estrone, and the major estrogen specific to pregnancy, estriol. Serum concentrations of progesterone and estriol increase with advancing gestation. Progesterone has a profound effect on uterine smooth muscle, maintaining the myometrium in a relatively relaxed state, and has a similar effect on the smooth muscle of other systems, including the gastrointestinal, renal, and pulmonary systems. Progesterone acts directly on smooth muscle by inhibiting calcium release from intracellular stores without an apparent effect on influx of extracellular calcium. [6], [7] Progesterone may also modulate immune mechanisms responsible for the maintenance rather than the rejection of fetal tissue. Estrogens have a potent ability to increase uteroplacental blood flow, an effect that may be mediated by prostaglandins. [8] Estrogens have also been implicated in changes in smooth muscle physiology, although studies suggest it is a less potent smooth muscle relaxant than progesterone.

Many of the physiological changes occurring during pregnancy result in alterations of the normal values of routine laboratory tests (Table 52-2). Some of the changes in values seen during pregnancy result from the confounding effects of several factors. For example, despite the rise in albumin synthesis during pregnancy, the plasma albumin level decreases during the second trimester because of the increase in plasma volume.

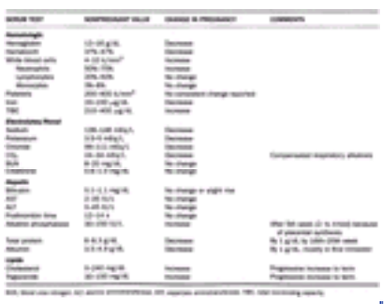

TABLE 52-2 Changes in Common Laboratory Tests during Pregnancy

Drug Metabolism in Pregnancy

Pregnancy induces changes in drug absorption, metabolism, distribution, and elimination. Because of the dynamic nature of these changes, it is often difficult to predict which factors will have a significant impact on drug pharmacokinetics. [9], [10] Knowledge of the changes in drug metabolism can assist physicians and patients in the safe and effective management of medications prescribed during pregnancy. Drug absorption may be affected by the changes in gastrointestinal function that occur during pregnancy. [11], [12] Delayed gastric emptying may delay peak drug levels. [13] Prolonged intestinal transit time may increase the absorption of some relatively water-insoluble drugs or increase the metabolism of drugs by intestinal wall enzymes. [14] Decreased gastric acid secretion may change the gastric pH and alter drug solubility.

Changes in body fluid compartments during pregnancy can profoundly affect drug distribution. [11] Increases in maternal plasma volume and the enlarging fetal compartment increase the volume of distribution of many drugs, altering maternal plasma drug concentrations and elimination half-life. The progressive decrease in maternal albumin concentration causes a corresponding increase in unbound drug fraction. Because pharmacological efficacy and toxicity are related primarily to the concentration of unbound drug, pregnancy-induced changes in protein binding may have important clinical implications, especially for acidic drugs that are highly protein-bound. [11] The 50% increase in the maternal glomerular filtration rate that occurs during pregnancy [15] results in the increased elimination of renally excreted drugs.

The changes in hepatic drug metabolism have not been rigorously studied during pregnancy. 17-Ethynyl estradiol decreases cytochrome P450 activity. [16] On the other hand, drug levels of phenytoin (Dilantin) and carbamazepine (Tegretol) decrease in pregnancy, presumably because of increased P450 activity. [17] Progesterone induces certain cytochrome P450 enzymes and other mixed function oxidase enzymes. [18], [19] These hormone activities can have confounding effects on drug metabolism and make drug levels difficult to predict.

Placental and fetal factors influence the effect of maternal medications on the developing fetus. Although most chemicals enter the fetal circulation by simple diffusion, some cross the placenta by facilitated or active transport processes. [20] The placenta contains enzymes capable of drug oxidation, reduction, hydrolysis, and conjugation. [21] Placental cytochrome P450 enzymes may play a role in fetal protection in the bio-oxidation of xenobiotics, including drugs. [22] Fetal tissues are also capable of metabolizing drugs. Although this contributes little to drug clearance, which is largely effected by maternal systems, it may contribute to fetal toxicity through the accumulation of toxic drug metabolites. This mechanism may be particularly important for polar drugs, which do not readily cross the placenta back to the maternal circulation. [11] Fetal albumin concentrations are below maternal levels early in pregnancy and then rise progressively. Early in pregnancy, fetal unbound drug concentrations may be high, even when maternal drug concentrations are low, resulting in toxicity to the fetus. [23] This potential toxicity during the first trimester, the period of major organogenesis, underlies the recommendations for the conservative use of medications during early pregnancy.

Fetal malformations occur in 3% to 6% of pregnancies. [24], [25] Drugs are probably responsible for only about 1% to 5% of fetal malformations; most malformations result from unknown causes. [25], [26] and [27] Even though the teratogenic effects of drugs account for a minority of fetal malformations, the consequences of a single unrecognized teratogen can be devastating. Unfortunately, few studies have been conducted of medication during pregnancy, and most information on the teratogenic potential of medications is derived from animal studies. [28] The Food and Drug Administration has established five drug categories to assist patients and physicians in the decision about drug use in pregnancy (Table 52-3). [9] The risk categories are based on the degree to which available information has ruled out risk of a drug to the fetus, balanced against the potential benefits of the same drug to the patient. Critics argue that excessive reliance on the results of animal studies can lead to inaccurate and misleading conclusions. The difficulties of extrapolating the results of animal studies to humans are well-known, the most notable example being thalidomide, which was not teratogenic in animal studies. [28] More accurate and reliable methods of determining human teratogenic potential are being studied, [29] and methods of determining patients' genetic predisposition to teratogenicity are being sought. [30] Table 52-3 lists current information and the pregnancy risk category for commonly used gastrointestinal drugs. [9], [10], [31]

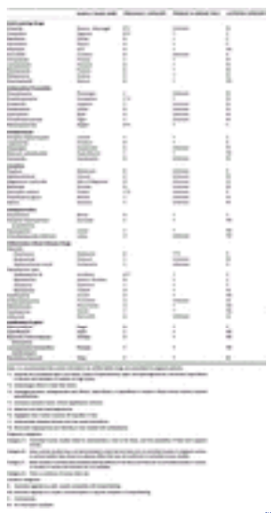

TABLE 52-3 Commonly Used Gastrointestinal Drugs in Pregnancy and Lactation

GASTROINTESTINAL SYMPTOMS IN PREGNANCY

Gastrointestinal symptoms are common during pregnancy. Symptoms such as nausea, constipation, and reflux are common and usually mild, and they rarely require diagnostic tests or drug therapy. Occasionally, however, serious diseases arise during pregnancy, and a reluctance to perform diagnostic tests can significantly delay diagnosis. Studies suggest that endoscopic procedures are safe during pregnancy and should be considered for the evaluation of serious symptoms. [32], [33]

Nausea and Vomiting and Hyperemesis Gravidarum

Nausea and vomiting have been estimated to occur in 50% to 90% of all pregnancies and begin in the first trimester in most patients. [34], [35] It is generally held that nausea and vomiting predict a good pregnancy outcome because the incidence of stillbirths and miscarriages is lower in women experiencing these symptoms. [35], [36] Nausea and vomiting do not affect birth weight and have not been associated with an increased incidence of congenital malformations.

Intractable vomiting that occurs early in gestation, causing dehydration, electrolyte disturbances, or nutritional deficiencies, is called *hyperemesis gravidarum*. [5] The timing, epidemiology, and clinical features are similar to those of the common, milder form of nausea and vomiting in pregnancy, suggesting that nausea during pregnancy may be part of a spectrum, with hyperemesis gravidarum representing the extreme form. The average incidence of hyperemesis gravidarum is 3.5 cases for every 1000 deliveries. [37], [38]

Pathophysiological Considerations Several lines of evidence support a role for the steroid hormones estrogen and progesterone in the pathogenesis of nausea and vomiting of pregnancy. First, a strong correlation has been found between nausea and vomiting and intolerance to oral contraceptives. [35] Second, epidemiologic studies show a higher incidence of nausea and vomiting in nulliparous, nonsmoking, and obese women, all of whom have higher levels of circulating and urinary estrogens. [39] Estrogen and progesterone both have relaxant effects on gastrointestinal smooth muscle. [40] Steroid hormones, particularly progesterone, may prolong gastric emptying and intestinal transit times, which may predispose the patient to nausea and vomiting. Pregnant patients with nausea and vomiting have gastric slow wave dysrhythmias (i.e., tachygastria and bradygastria) [41] and instability of fasting electrical activity and an altered electrical response to food ingestion. [42] The gastric slow wave dysrhythmias can be induced in asymptomatic, nonpregnant patients by administering progesterone alone or in combination with oral estrogen at doses that result in serum hormone levels near the levels of pregnancy. [43] On the other hand, nausea and vomiting are most common in the first trimester, whereas steroid hormone levels peak near term. In addition, estradiol and progesterone levels do not differ between asymptomatic pregnant patients and pregnant patients with nausea and vomiting. [44] These latter observations have led to the hypothesis that it is the rapid changes in gestational hormones early in pregnancy that induce nausea and vomiting in susceptible patients. [45] Levels of hCG peak during the first trimester, at a time when symptoms of nausea and vomiting are commonly recognized. It is controversial whether hCG levels are higher in pregnant patients with nausea and vomiting than in asymptomatic pregnant patients. [46] Women with molar pregnancies have extremely high hCG levels and a higher incidence of nausea and vomiting than women with healthy intrauterine pregnancies. However, hCG levels do not differ between patients who have molar pregnancies with nausea and vomiting and patients who have molar pregnancies without nausea and vomiting. Transient elevations in serum thyroxine levels, especially serum free thyroxine, have been documented in as many as 70% of pregnancies complicated by hyperemesis gravidarum. [38], [47], [48] and [49] Hyperthyroxinemia resolves in the second trimester. Because hCG has inherent thyroid-stimulating activity, it has been suggested that high hCG levels in the first trimester are responsible for the hyperthyroxinemia. [38] It has been postulated that hCG and thyroxine cause maternal nausea and alter the nutrient balance favoring placental growth. [50] *Helicobacter pylori* infection has been found to be more common in patients with hyperemesis gravidarum than in controls, and antibiotics have significantly relieved symptoms of nausea and vomiting in *H pylori*–positive patients. [51], [52] and [53]
Clinical Features, Diagnosis, and Treatment Hyperemesis gravidarum typically begins during the sixth week of gestation and improves in the second trimester. Sixty percent of patients with hyperemesis gravidarum may have symptoms beyond 20 weeks, whereas women with the milder form rarely have symptoms beyond 20 weeks. The sequelae of the intractable vomiting of hyperemesis gravidarum include fluid and electrolyte disturbances, dehydration, weight loss, ketosis, and acetonuria. Laboratory abnormalities, including elevations in aspartate aminotransferase (AST), alanine aminotransferase (ALT), bilirubin, amylase, and lipase, may be seen. [54] Rare complications include neurological abnormalities, retinal hemorrhage, Mallory-Weiss tear, and acid aspiration syndrome. [37], [54] The management of common nausea and vomiting in pregnancy depends on the severity of the condition. Inappropriate weight gain or weight loss may indicate a more serious disease and warrants close monitoring. Patients may be advised to eat frequent small meals and to avoid easily identifiable situations that precipitate nausea and vomiting. Rarely is drug therapy required for nausea and vomiting in early pregnancy, but when needed, antihistamines and phenothiazines are commonly used. [55] Hospitalization is often the key to successful therapy of hyperemesis gravidarum. Fluid and electrolyte therapy is necessary to reverse abnormalities. Enteral or parenteral hyperalimentation may be necessary in extreme cases. [56], [57] The use and therapeutic value of pharmacological therapy in hyperemesis gravidarum is controversial. Phenothiazines and antihistamines have been used with some success, [58] although their safety in pregnancy is not clearly established. The serotonin receptor antagonist ondansetron was no better than promethazine for the treatment of hyperemesis gravidarum. [59] It is reasonable to consider *H pylori* infection in severe cases of hyperemesis gravidarum, although treatment in pregnancy is not straightforward because of medication and safety issues. In general, hyperemesis gravidarum has no adverse effect on pregnancy outcome. [60], [61]

Gastroesophageal Reflux

Heartburn is a common symptom of pregnancy, estimated to affect as many as 60% to 70% of pregnant women. [62] The prevalence of heartburn increases with gestational age. [62], [63] Patients with prepregnancy heartburn and increased parity are at increased risk for experiencing heartburn during pregnancy. Heartburn during pregnancy does not appear to affect maternal health or the well-being of the fetus. [64]

Pathophysiological Considerations Two mechanisms have been proposed to explain the pathophysiology of heartburn in pregnancy: decreased lower esophageal sphincter (LES) pressure and the mechanical effects of the gravid uterus. Other potentially important factors, such as relaxations, mechanisms of esophageal acid clearance, and sensory neuronal mechanisms, have not been established. Women with heartburn during pregnancy appear to have LES pressures lower than those of nonpregnant controls or pregnant women without heartburn. [65] Early in pregnancy, asymptomatic pregnant patients had normal LES pressures compared with nonpregnant patients; however, pregnant patients exhibited blunted responses to pharmacological challenges with edrophonium, methacholine, and pentagastrin, [66] suggesting that LES competence is impaired early in pregnancy, before the development of symptoms. LES pressures progressively fell when studied at 12, 24, and 36 weeks of gestation and returned to normal 1 to 4 weeks after delivery. [65] The perceived importance of mechanical factors in the development of heartburn during pregnancy has diminished in the past decade as more interest has focused on hormonal changes affecting the LES. [67] Theories regarding mechanical factors are based on the positive correlation between the prevalence of heartburn and the size of the gravid uterus. Proposed mechanical factors include loss of the intra-abdominal LES segment, alterations of anatomic structures surrounding the LES, hiatal hernia formation, and delayed gastric emptying. [68]
Clinical Features, Diagnosis, and Treatment A compatible history is usually sufficient to make the diagnosis of reflux in pregnancy. Confirmatory tests are rarely needed; radiographic studies should be avoided. Endoscopy can be performed safely during pregnancy but is indicated only if complications such as stricture or ulceration are suspected. Treatment regimens must consider drug side effects and fetal well-being. A standard antireflux regimen emphasizing frequent small meals

with no food before bedtime, elevating the head of the bed, and avoiding bothersome foods is effective for most patients. Antacids and sucralfate are considered safe when used in the second and third trimesters (see Table 52-3). [69] Little is known regarding the safety of histamine H_2 receptor antagonists during pregnancy. All histamine H_2 receptor antagonists readily cross the placenta. [70] Cimetidine has an antiandrogenic effect in animals [71], [72] but has been safely used to treat peptic ulcer disease and to prevent aspiration at the time of cesarean section. [73] Ranitidine does not have antiandrogen effects. [72] A prospective cohort study showed no increased risk for premature birth or teratogenicity when histamine H_2 blockers were used in the first trimester. [74] Promotility drugs such as metoclopramide, cisapride, domperidone, and proton pump inhibitors have been used to treat reflux associated with pregnancy, although experience is limited and controlled studies are unavailable. [69], [75], [76]

Constipation

Pregnancy is often associated with constipation. In one study, 31% of women were constipated during pregnancy, most commonly during the first trimester and again late in gestation. [77] In another study, 11% of women reported decreased bowel movement frequency during pregnancy, but only 1.5% of this group required laxative treatment. Fifty-five percent did not have any change in their bowel habits, and 54% reported increased frequency. [78]

Pathophysiological Considerations It is postulated that constipation results from bowel hypomotility related to the smooth muscle relaxant effects of estrogen and progesterone. Several studies have shown an increased gastrointestinal transit time during pregnancy, especially during the second and third trimesters. [79], [80] and [81] The increased gastrointestinal transit time was correlated with increased serum progesterone levels and returned to normal as the levels normalized after delivery. [79], [80] The transit time was prolonged during the luteal phase of the normal menstrual cycle when the progesterone level was high compared with the transit time during the follicular phase when the progesterone level was low. In pregnant women, mean plasma motilin concentrations were reduced with fasting and after a glucose load or a mixed meal, with a return to normal within 1 week after delivery. [82] Other factors contributing to the constipation of pregnancy include pressure on the rectosigmoid by the gravid uterus, increased colonic absorption of sodium and water, and prenatal vitamin supplementation with iron.
Clinical Features, Diagnosis, and Treatment Constipation during pregnancy usually does not warrant extensive evaluation. Serious illnesses such as bowel obstruction may present as constipation and can usually be excluded by clinical evaluation. Increased dietary fiber and fluid intake provide symptomatic relief of constipation. If constipation persists, supplemental fiber in the form of psyllium appears to be safe. [69], [76] Other laxatives have not been studied sufficiently, but several are widely used without apparent adverse effects, including bisacodyl, senna, and docusate stool softeners (see Table 52-3).

Acute Abdominal Pain

The differential diagnosis of acute abdominal pain includes all nonobstetrical causes as well as causes specific to pregnant women (Table 52-4). Multiple factors complicate the diagnosis of acute abdominal pain during pregnancy. First, the differential diagnosis of acute abdominal pain changes as pregnancy progresses. For example, ectopic pregnancy must be considered strongly in the first trimester, but other obstetrical causes are more common in the second and third trimesters. Second, anatomic landmarks are shifted by the gravid uterus. Toward the end of gestation, the appendix may be located in the right upper quadrant, often causing confusion between appendicitis and acute cholecystitis (Fig. 52-2). Third, the diagnosis can be delayed by reluctance to perform diagnostic tests during pregnancy, especially tests involving ionizing radiation. The abdominal wall becomes less reactive, probably because of the effects of gestational hormones and the enlarging uterus, and the usual diagnostic signs of rebound tenderness, guarding, and rigidity are less commonly elicited. In the early stages, nonobstetrical pain is often attributed to obstetrical causes, resulting in a delayed diagnosis. The combined effect of these factors is to delay the diagnosis of acute abdomen in pregnancy to the point at which it is often made only after the patient has become extremely ill. Pregnancy outcomes and management recommendations are being reevaluated in light of the increasing use of laparoscopic techniques. [83], [84], [85] and [86]

Nonobstetrical Causes
Acute appendicitis
Acute cholecystitis
Acute pancreatitis
Hepatic rupture
Intestinal obstruction
Adnexal torsion
Sickle cell crisis
Obstetrical Causes
Ectopic pregnancy
Abruptio placentae
Red degeneration of a uterine myoma
Uterine rupture

TABLE 52-4 Causes of Acute Abdomen in Pregnancy

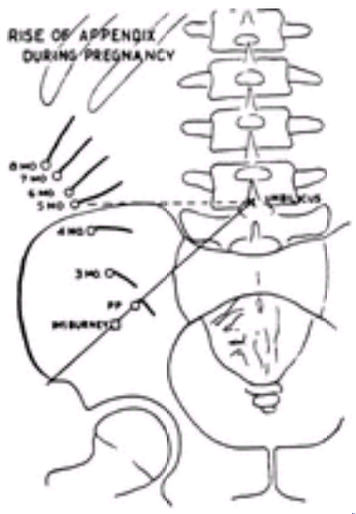

FIGURE 52-2. The position of the appendix changes during pregnancy. (From Baer JL, Reis RA, Arens RA. Appendicitis in pregnancy with changes in position and axis of the normal appendix in pregnancy. JAMA 1932;98:1359.)

Nonobstetrical Causes

Acute appendicitis. Appendicitis develops in approximately 1 in 2,000 pregnant women. [87], [88] Appendicitis is the most common nonobstetrical condition requiring surgery during pregnancy. Symptoms are generally the same as in nonpregnant patients, but the location of abdominal tenderness differs because the enlarging uterus displaces the appendix cephalad (see Fig. 52-2). Guarding, rebound tenderness, and fever all appear to be less common in pregnant patients with acute appendicitis. [89], [90] The differential diagnosis includes ligamentalgia, ovarian torsion, ectopic pregnancy, and pyelonephritis. [91] The physiological leukocytosis of pregnancy reduces the diagnostic utility of this common sign of inflammation. [89] Pyuria and hematuria may be more common, occurring in 10% to 29% of cases, because of the proximity of the appendix to the retroperitoneal ureter. [89] Local perforation and peritonitis may not be immediately evident because the uterus can form a medial wall that contains the abscess. The abscess can stimulate uterine contractions and premature delivery. Transvaginal ultrasonography is a safe and effective means of imaging the pelvis and may be helpful, especially early in pregnancy. [91] Early surgery is indicated because maternal and fetal mortality rates increase with the development of peritonitis. [89] Delayed diagnosis is probably responsible for the higher rates of appendiceal perforation and mortality associated with acute appendicitis in pregnancy. Laparoscopy appears to be as safe as laparotomy. [92]
Acute pancreatitis. The incidence of acute pancreatitis in pregnancy is estimated to be between 1 in 4,000 and 1 in 12,000 pregnancies. [93] It is unclear if this incidence is higher in pregnancy than in a matched nonpregnant population. Most cases occurring in pregnancy are associated with cholelithiasis. [94] Other causes of pregnancy-associated acute pancreatitis include hyperparathyroidism [95] and alcoholism. [96] Acute pancreatitis during pregnancy also occurs in the setting of an underlying lipoprotein disorder. Most women have a modest increase in serum triglycerides during the third trimester [97] because of a direct effect of estrogen on liver lipoprotein synthesis and a decreased clearance of triglycerides secondary to hormonal suppression of lipoprotein lipase activity in the liver and adipose tissues. [98], [99]

This increased triglyceride concentration is usually of little clinical consequence, but significant hyperlipidemia may develop in women with inherited lipoprotein disorders and can result in xanthomatosis and acute pancreatitis. Early recognition is key because patients can be managed successfully with a low-fat diet. [100], [101] Acute pancreatitis in pregnancy carries a high risk for maternal and fetal mortality. The early use of total parenteral nutrition is warranted to maintain maternal and fetal nutrition and to avoid a catabolic state in the pregnant woman. [102] Symptomatic biliopancreatic gallstone disease as a cause of pancreatitis can be managed surgically or endoscopically (see section " Cholelithiasis"). [94], [103], [104] Pseudocysts have been reported to complicate acute pancreatitis in pregnancy, and conservative management is generally recommended until after delivery. [105]

Intestinal obstruction. Intestinal obstruction occurs in between 1 in 2,500 and 1 in 3,000 pregnancies. [106] Most cases occur in patients who have had a prior operative procedure, most commonly appendectomy or gynecologic surgery, and presumably result from pressure on preexisting adhesions by the enlarging uterus. [107], [108] Obstruction is most common in the third trimester and least common in the first. [109] Symptoms of obstruction in pregnant women are similar to those of nonpregnant patients. A single supine and upright abdominal x-ray film may be diagnostic, but the diagnosis is confirmed more commonly by the progression of x-ray findings over several hours. The management principles are similar to those for intestinal obstruction in the nonpregnant patient. Operative therapy is generally required for small bowel obstruction. Cecal or sigmoid volvulus can be successfully treated with endoscopic detorsion. [108] The maternal mortality rate is 10% to 20%. Fetal mortality appears to be even higher secondary to fetal hypoxia and acidosis. [110], [111]

Adnexal torsion. An ovarian cyst in a twisted adnexa can cause an acute abdomen during pregnancy. [112] As with appendicitis, the localization of the pain may not be classical because of the enlarged uterus. A prompt diagnosis and surgical intervention are necessary to avoid tissue necrosis. In the case of a dermoid cyst, prompt surgical therapy decreases the risk for rupture, which can produce a severe generalized peritonitis. [113] Adnexal torsion is often misdiagnosed as acute appendicitis, with the correct diagnosis made at laparotomy. The ultrasonic appearance may differentiate complex and fluid-filled ovarian cyst from an inflamed appendix. [112]

Sickle cell anemia. Patients with sickle cell anemia may present with severe abdominal pain during pregnancy. [114], [115] Severe sickle cell crisis requires treatment, as in the nonpregnant patient, and is associated with a high maternal and fetal mortality rate. [116]

Obstetrical Causes

Ectopic pregnancy. The most common obstetrical cause of an acute abdomen in the first trimester is ectopic pregnancy. History, pelvic examination, measurement of the serum ß-hCG level, culdocentesis, ultrasonography, and laparoscopy can all be valuable in establishing the diagnosis. Medical and surgical therapies are available (see section " Ectopic Pregnancy" for a more detailed discussion).

Abruptio placentae. Abruptio placentae is an important cause of severe abdominal pain during late gestation, occurring in about 8 in 1,000 pregnancies. [117] Abruption occurs when the placenta prematurely separates from its uterine attachment. Classically, patients present with pain, uterine irritability and tenderness, and vaginal bleeding. The diagnosis can be more difficult if blood is retained in the uterus. Ultrasonography may demonstrate a retroplacental blood collection, and amniocentesis may yield bloody amniotic fluid. The diagnosis should be considered if severe, unremitting pain develops during labor. Abruptio placentae may be self-limited or may cause fetal demise and catastrophic maternal complications, including massive hemorrhage, disseminated intravascular coagulopathy, and renal cortical necrosis. Treatment depends on the severity of the abruption but generally consists of blood replacement, correction of disseminated intravascular coagulopathy if present, and expeditious delivery. [118], [119]

Red degeneration of a uterine myoma. Red degeneration of a uterine myoma is caused by a hemorrhagic infarction of a uterine fibroid. Focal acute pain may be associated with low-grade fever and ileus. The diagnosis should be considered in patients with pain and fibroids in whom other diagnoses have been excluded. [118]

Uterine rupture. Uterine rupture generally occurs late in pregnancy in patients with a prior history of cesarean section or myomectomy. Uterine rupture may occur in as many as 1% in patients attempting vaginal delivery after cesarean section with a lower uterine segment incision. The rupture may occur spontaneously or during labor and involves dehiscence of the prior surgical scar with or without expulsion of the fetus into the abdominal cavity. Traumatic uterine rupture usually occurs in the setting of a motor vehicle accident. Expulsion of the fetus usually results in fetal demise and intra-abdominal bleeding. Treatment is surgical. [91]

GASTROINTESTINAL AND LIVER DISEASES IN PREGNANCY

Inflammatory Bowel Disease

The majority of women with inflammatory bowel disease (IBD) are of childbearing age. Recent studies support an optimistic view regarding fertility and the course of pregnancy for patients with IBD. [120], [121] and [122]

Effects on Fertility Little evidence supports fertility problems in patients with ulcerative colitis; two control studies showed no decrease in fertility. [123], [124] and [125] In Crohn's disease, the existing data are not as straightforward. Two case control studies reported decreased fertility in patients with Crohn's disease. [126], [127] Infertility appears to correlate with disease activity and is inversely related to nutritional status. Measures of childlessness, infertility, fecundity, and methods of birth control suggest that in some cases reduced fertility may be the patient's choice rather than a consequence of the disease. [124] Early data were conflicting as to whether infertility was more common with large or small bowel Crohn's disease, [128], [129] although in a recent controlled study, the site of intestinal involvement did not appear to be a factor. [126]

Effects on Pregnancy Outcome Patients with ulcerative colitis deliver healthy offspring, with rates of congenital abnormalities, spontaneous abortion, and stillbirth that approximate those in the general population. [123], [127], [130], [131], [132] and [133] Some studies suggest that spontaneous abortion and stillbirth are increased in patients with severe colitis, especially when the onset of colitis is during pregnancy. [134] Data suggest that patients with severe colitis during pregnancy can be managed successfully with parenteral hyperalimentation. [134], [135] and [136] Patients with Crohn's disease that is mild or in remission at the time of conception appear to have normal pregnancies. [132] However, in patients with active Crohn's disease at the time of conception, the incidence of stillbirth, low birth weight, spontaneous abortion, and premature delivery is increased. [131], [137], [138] and [139]

Effects of Pregnancy Data support the classical concept that IBD that is quiescent at the onset of pregnancy is likely to remain quiescent throughout pregnancy. Patients with an exacerbation of ulcerative colitis brought into remission during pregnancy are also likely to do well for the remainder of pregnancy. However, patients with active disease at the time of conception are likely to experience worsening of their disease. [132], [139] Pregnancy does not appear to change the relapse rate of ulcerative colitis or Crohn's disease. [131], [134], [137], [140], [141] Evidence suggests that the course of ulcerative colitis during a single pregnancy may not necessarily predict its course during subsequent pregnancies. Patients with IBD may experience a marked improvement in their symptoms during pregnancy but then a flare of their disease postpartum.

Effects of Drug Therapy Data suggest that pregnancy occurring during disease remission is more likely to result in a normal full-term baby. A patient who is in a remission that can be sustained without drug therapy should remain off therapy. However, most patients can achieve a satisfactory remission only with medical therapy. It is important to consider the potential side effects of the drugs used and their potential harm to the fetus (see Table 52-3). Sulfasalazine can cross the blood-brain barrier, theoretically causing kernicterus by displacing bilirubin from serum albumin. Fortunately, because sulfasalazine is poorly absorbed and has a low affinity for binding protein, neonatal jaundice is quite uncommon. [142] Sulfasalazine therapy has not been associated with an increase in prematurity, birth weight, or spontaneous abortion. [143], [144] The incidence of developmental defects is only 1%, which is similar to the incidence in the general population. [143] Sulfasalazine is present in small amounts in breast milk. [145], [146] However, no side effects of sulfasalazine have been reported in nursing infants. It is important to recommend that patients on sulfasalazine take a folate supplement because sulfasalazine inhibits the transport and metabolism of folic acid. The 5-aminosalicylates avoid some of the theoretical concerns of sulfasalazine. No known complications of 5-aminosalicylates in pregnancy have been reported. [147] Currently, there is no justification for switching pregnant patients from sulfasalazine to a 5-aminosalicylate. Male patients with IBD who desire fertility should be given 5-aminosalicylates because sulfapyridine, one of the metabolites of sulfasalazine, can decrease sperm count and motility. [148] Steroids have been widely used for many diseases during pregnancy and have not been shown to increase the risk for low birth weight, spontaneous abortion, or fetal abnormalities despite an increased incidence of these findings in animals treated with high doses of steroids. In a series of 287 patients with IBD treated with corticosteroids, the frequency of fetal complications was lower than in the general population. [130] It appears that patients who become pregnant in the setting of severe active Crohn's disease are at risk for complications regardless of the treatment, and it seems prudent to use corticosteroids when needed in moderate or severe disease to induce a remission. Metronidazole in high doses is tumorigenic and teratogenic in animals. It is present in breast milk, and its effect on the human fetus is unknown. Therefore, metronidazole should be avoided during pregnancy. [31] Cyclosporine is embryotoxic and fetotoxic in animals but has been used safely in patients with severe ulcerative colitis. [149] Azathioprine and 6-mercaptopurine are being used increasingly for primary therapy of IBD and for their steroid-sparing effect. These drugs have well-known teratogenic effects in animals. Nevertheless, the drugs have been used safely in more than 1000 renal transplant patients who became pregnant. [150] Even though patients with IBD are on relatively low doses of immunosuppressant therapy, such therapy is not routinely recommended during pregnancy. [151] The risks and benefits of these drugs must be considered with the patient before conception. Infliximab has been used safely in pregnancy, although data are scant. [152]

Peptic Ulcer Disease

Peptic ulcer disease and its complications are rare during pregnancy. [153], [154] It has not been clearly established whether the incidence truly differs between pregnant

and nonpregnant women because peptic ulcer disease is uncommon in all women of childbearing age. [5] Peptic ulcer disease may be underdiagnosed in pregnancy, thus confounding these statistics. Peptic symptoms may more likely be attributed to reflux, and like reflux symptoms, they may improve with antacid therapy. Moreover, diagnostic tests are performed less often in pregnant patients. The symptoms of patients with known peptic ulcer disease usually improve during early gestation, and they appear to be protected from peptic symptoms until late in the third trimester. Other studies report disappearance of ulcer symptoms in 98% of pregnant women, although by the end of the third postpartum month, symptoms may recur in almost 50%.

Complications of peptic ulcer disease during pregnancy, such as perforation, hemorrhage, and obstruction, are extremely rare; however, when they occur, they do so late in the third trimester or early in the postpartum period. [155], [156] During pregnancy, such complications may become life-threatening for both mother and child. All reports of fetal and maternal mortality from complications of peptic ulcer disease precede the widespread use of histamine H_2 receptor antagonists. The treatment of peptic ulcer disease is based on symptoms and a risk-benefit analysis of drug therapy. A cohort study showed no increased risk for premature birth or teratogenicity when histamine H_2 blockers were used in the first trimester. [155] Cimetidine has been used without adverse effects during pregnancy but is not recommended because of antiandrogen effects. [72] Antiandrogen effects have not been demonstrated with ranitidine or other histamine H_2 receptor antagonists. Histamine H_2 receptor antagonists and proton pump inhibitors are listed in Table 52-3 and are discussed in the section “Gastroesophageal Reflux.” [157] Little information is available on the diagnosis or treatment of *H pylori* infection in pregnancy.

Cholelithiasis

Asymptomatic gallstones are found in 4.5% of pregnant women. [158], [159] Cholelithiasis is also more common in women with a history of prior pregnancy. [160], [161]

Pathophysiological Considerations The changes in gallbladder function and bile composition that occur during pregnancy favor gallstone formation. Gallbladder size increases during pregnancy, [162], [163] and [164] with no corresponding change in common bile duct diameter. [165] Gallbladder emptying appears to be impaired and residual volume increased in pregnancy, possibly because of the smooth muscle relaxant effects of progesterone. During the second and third trimesters, a progressive decrease in the rate of chenodeoxycholate synthesis with no change in the rate of cholic acid synthesis results in an altered ratio of bile salts that is characteristic of pregnancy and hormonal therapy. The hepatic secretion rate of cholesterol into bile remains constant during pregnancy. [166] The decreased total bile salt pool in the later half of pregnancy increases the fractional concentration of cholesterol, creating favorable conditions for gallstone formation. [166]
Clinical Features, Diagnosis, and Treatment The signs and symptoms of biliary colic, acute cholecystitis, and cholelithiasis are similar to those in nonpregnant patients. The differential diagnosis includes other causes of acute pain in pregnant women (see Table 52-4). As in nonpregnant patients, the diagnosis is based on typical symptoms, laboratory findings, and the presence of gallstones. Ultrasonography is the preferred method for detecting gallstones, although ultrasonographic examination may be more difficult during late gestation because of gallbladder displacement. Oral cholecystography and isotope scans should be avoided. It is unknown whether the frequency of complications of cholelithiasis is higher in pregnant than in nonpregnant women. Cholecystitis is second only to appendicitis as an indication for surgery in pregnant women. Spontaneous perforation of the gallbladder associated with cholecystitis has been reported. [167] The approach to therapy is similar to that in the nonpregnant patient. The treatment of uncomplicated biliary colic is generally conservative, with analgesics and antiemetics. Surgery is generally indicated for patients with acute cholecystitis. Classically, the second trimester is considered the safest time for surgery because the risk for spontaneous abortion is increased during the first trimester, and the risk for premature labor in the third. [168] Various data suggest that cholecystectomy is safe at any stage of pregnancy. [103], [104], [169] Laparoscopic cholecystectomy is the standard and is associated with fewer complications than open laparotomy. [170], [171] Pancreatitis caused by choledocholithiasis during pregnancy can be serious; high morbidity and mortality rates underlie the recommendations for early intervention. Endoscopic retrograde cholangiopancreatography has been used as a bridge to postpartum cholecystectomy in patients with complications of choledocholithiasis. [172]

Viral Hepatitis

Acute viral hepatitis probably occurs in fewer than 1% of gestations, although the true incidence is unknown. [173], [174] and [175] The clinical manifestations and laboratory findings in acute hepatitis A, B, and C (caused by the hepatitis A, B, and C viruses, respectively) are similar to those in nonpregnant women. The onset is most common in the third trimester. Fulminant viral hepatitis and liver failure can occur but are rare. Recent reports of hepatitis E in India and the Middle East suggest that acute hepatitis E in pregnant women is a more severe disease than in nonpregnant women, and the mortality rate is higher than that associated with other forms of acute viral hepatitis. [176], [177] Acute viral hepatitis of any cause is associated with an increased rate of fetal loss and premature birth. Vertical transmission of hepatitis viruses from acutely infected mothers has been reported but is rare. Infants exposed to hepatitis A are given immune serum globulin and hepatitis A vaccine at delivery. [174], [178] Infants exposed to hepatitis B receive hepatitis B immune globulin and hepatitis B vaccine. [179] No prophylaxis against the vertical transmission of hepatitis C or E is available.

The effect of chronic maternal hepatitis B on pregnancy outcome and maternal well-being depends on the degree of liver dysfunction. Patients with normal liver function have a good prognosis, whereas the disease of women with significant liver dysfunction may worsen, creating a risk for spontaneous abortion, premature delivery, and perinatal death. [180], [181] Hepatitis B virus can be transmitted vertically from chronically infected mothers during pregnancy or at delivery. The risk for vertical transmission increases with the maternal viral load and replication rate and may be as high as 90%. In addition, chronic hepatitis B develops in 90% of infected infants. [182], [183] and [184] Infants exposed to the hepatitis B virus should be treated with hepatitis B immune globulin and hepatitis B vaccine. Immunoprophylaxis is highly effective in preventing both hepatitis B and hepatitis D transmission. [185], [186] and [187] The high incidence of chronic hepatitis B in exposed offspring and the efficacy of hepatitis B immunoprophylaxis underlie the current recommendations for universal screening for hepatitis B surface antigen (HBsAg) in all pregnant women during the third trimester. [179]

Women with chronic hepatitis C generally have mild liver disease with normal or nearly normal liver function; pregnancies in women positive for hepatitis C virus are generally uncomplicated. [188] In many women, the transaminase levels normalize, often concurrently with an increase in the hepatitis C viral load. [189], [190] The cause of these changes is unclear but may be related to immunologic changes occurring during pregnancy. Hepatitis C can worsen after pregnancy. [191] Hepatitis C may be a risk factor for cholestasis of pregnancy. [192]

Hepatitis C virus is transmitted vertically in approximately 5% of deliveries from hepatitis C virus RNA–positive mothers. [189], [193], [194], [195] and [196] Breast-feeding does not appear to be a mode of transmission, even though hepatitis C virus may be detected in breast milk. [189], [193], [194] Although there is no known effective immunoprophylaxis for hepatitis C, it is encouraging that viremia in infected infants may be only transient. [189], [194] Prenatal screening for hepatitis C virus is not yet routine but may be reasonable given that women positive for hepatitis C virus cannot be reliably identified by history or examination. [197], [198] and [199] The safety and efficacy of anti–hepatitis C virus therapy in pregnant women or their infected offspring are unknown. [200], [201]

Intrahepatic Cholestasis of Pregnancy

Intrahepatic cholestasis of pregnancy (IHCP) is a benign cholestatic disorder that usually develops in the third trimester and disappears abruptly after delivery. IHCP occurs in fewer than 1% of pregnancies in the United States. The incidence varies geographically and seasonally, and the overall incidence has decreased in the past 20 years. [202], [203]

Pathophysiological Considerations The etiology of IHCP is poorly understood, but hormonal, environmental, and genetic factors likely contribute. The most widely accepted hypothesis is that IHCP is an inherited sensitivity to the cholestatic effects of estrogen. In almost half of women in whom IHCP develops, pruritus or cholestasis also develops when they use oral contraceptives. [204], [205] and [206] Female relatives of index cases are at risk. [207], [208] IHCP is probably related to other genetic defects in bile salt synthesis or transport. [209] IHCP is associated with mutation of MDR3, a gene encoding a biliary canalicular phospholipid translocator and causing some cases of familial intrahepatic cholestasis. [210], [211] Environmental factors such as low dietary levels of selenium have been suggested. [212]
Clinical Features, Diagnosis, and Treatment IHCP typically causes pruritus in the third trimester that generally progresses until delivery or termination of the pregnancy. The pruritus is often severe. IHCP tends to recur during subsequent pregnancies at a rate of 60% to 70%. [213] After delivery, the pruritus quickly resolves within 24 to 48 hours, and biochemical abnormalities and histological changes resolve over weeks to months. Long-term follow-up suggests a higher incidence of cholelithiasis and gallbladder disease but not of chronic liver disease. [214], [215] and [216] The consequences for the fetus can be serious, and close surveillance of the pregnancy is warranted. The rates of premature labor and neonatal death are higher than expected, [215], [217], [218], [219], [220] and [221] although a more favorable outcome has been reported more recently. [222], [223] As expected, the serial markers of cholestasis, including bilirubin and alkaline phosphatase, are usually elevated, as are the

serum levels of bile salts and cholic acid and the Bromsulfophthalein retention time. Also, the markers of hepatocellular function, including AST and ALT, may be elevated several fold, mimicking the patterns seen in viral hepatitis. The timing and clinical course are usually diagnostic, and liver biopsy is generally not necessary for a diagnosis. The histopathological features of IHCP include central lobular cholestasis, canalicular bile plugs, and bile pigment within hepatocytes. Little inflammatory reaction or hepatic necrosis is seen. The portal tracts and intralobular bile ducts are normal. Histological changes and symptoms resolve after delivery. Therapy is directed at relieving the pruritus. Ursodeoxycholic acid relieves symptoms and improves laboratory values [224], [225], [226], [227] and [228]; the fetal outcome may also be improved. [229] Symptoms may respond to oral steroids [230] or cholestyramine. [231], [232] *S*-edenosyl-L-methionine, which antagonizes the cholestatic effects of estrogen, [233], [234] may be effective, particularly when combined with ursodeoxycholic acid. [235]

Acute Fatty Liver of Pregnancy

Acute fatty liver of pregnancy (AFLP) is a serious, rare, idiopathic liver disorder of the third trimester. The incidence is likely less than one case per million deliveries, although epidemiologic data are scant. [236], [237], [238], [239] and [240] Milder cases of the disease are now being reported, suggesting that AFLP is a spectrum and may be more common than has been appreciated. [241], [242] AFLP is more common in first pregnancies, twin pregnancies, and pregnancies in which the fetus is male.

Pathophysiological Considerations The cause of AFLP is unknown. It is related to other diseases in the group collectively termed *hepatic microvesicular steatoses*, which also include Reye syndrome, Jamaican vomiting sickness, valproic acid hepatotoxicity, tetracycline hepatotoxicity, and medium-and long-chain acyl-CoA dehydrogenase deficiency. [237], [243] Unlike IHCP, AFLP does not have a strong genetic component, and it does not tend to recur in subsequent pregnancies. The characteristic histological findings are infiltration of hepatocytes with microvesicular fat and pleomorphic and abnormal mitochondria. Based on the histological findings, a metabolic disorder of lipid metabolism or mitochondrial function has been suggested. The microvesicular fat consists principally of triglycerides and free fatty acids and is contained in non–membrane-bound cytoplasmic droplets. Potentially toxic levels of free fatty acids have been found. [244] Several rare defects in fatty acid oxidation have been diagnosed in infants born to mothers with AFLP, [245], [246] the most common being long-chain 3-hydroxyacyl-CoA dehydrogenase (LCHAD) deficiency. [247], [248], [249] and [250]

Clinical Features, Diagnosis, and Treatment The disease generally begins near 35 weeks of gestation with nonspecific symptoms such as nausea, fatigue, malaise, vomiting, and abdominal distress. Clinical signs of liver dysfunction and, in severe cases, liver failure, including jaundice, hepatic encephalopathy, and bleeding, develop 1 to 2 weeks later. Physical findings are minimal, with right upper quadrant tenderness the only consistent finding. The liver is generally nonpalpable. Altered mental status and ascites may appear late in the disease. Hypertension, proteinuria, and preeclampsia are seen in 21% of patients. AFLP and HELLP syndrome (see next section) are distinct clinical entities but may coexist. Complications of fulminant hepatic failure, including cerebral edema, hemorrhage, renal failure, and infection, can occur in AFLP and portend a poor prognosis. Maternal and fetal mortality rates are high; outcomes are improved with the prompt initiation of delivery. [173], [251], [252] Delivery is often complicated by severe postpartum hemorrhage. The signs and symptoms gradually resolve over weeks, and liver function returns to normal. Chronic liver disease is not seen. [253], [254] and [255] The AST, ALT, bilirubin, and alkaline phosphatase levels are universally elevated, with a mixed cholestatic and hepatocellular picture. Antithrombin III levels are also elevated, with patients exhibiting other abnormalities consistent with disseminated intravascular coagulation. Leukocytosis is universal. The serum liver chemistries may be only modestly elevated, underestimating the degree of liver dysfunction. As in other forms of hepatic failure, hypoglycemia, impaired synthesis of clotting factors, and hepatic encephalopathy are poor prognostic signs. Renal dysfunction occurs and occasionally requires dialysis. AFLP may be confused with acute viral hepatitis, preeclampsia-related liver disease, and tetracycline-induced fatty liver. Viral hepatitis may occur at any time during pregnancy, whereas AFLP occurs in the third trimester. Serologic testing for hepatitis A, B, or C virus may indicate a viral cause, and AST and ALT values tend to be much higher in acute viral hepatitis than in AFLP. Preeclampsia-related liver disease is always associated with hypertension and proteinuria, which are usually absent in AFLP. Tetracycline-associated liver disease can cause a syndrome that is virtually identical to AFLP. Previously, high doses of tetracycline were used to treat urinary tract infections and pyelonephritis; however, recognition of this potential problem and the use of alternative antibiotics have virtually eliminated this disease. The therapy for AFLP is supportive, and prompt termination of the pregnancy is generally advocated; successful liver transplantation has been performed in patients with fulminant hepatic failure and AFLP.

Preeclampsia-Associated Liver Disease and HELLP Syndrome

Preeclampsia is a common complication of mid to late pregnancy and is characterized by hypertension, proteinuria, and edema. [256] Preeclampsia develops in 5% to 10% of all pregnancies and is more frequent in young primigravidas and women with lupus anticoagulant. [257] Progression to seizures is the hallmark of true eclampsia and is rare. [258] The liver is involved in 10% of women with preeclampsia; liver involvement is more common in eclampsia. [259], [260] and [261] Liver involvement is the primary cause of death in 15% to 20% of fatal cases. [262], [263], [264] and [265]

Pathophysiological Considerations Two pathophysiological conditions have been described. The first, a moderately severe disorder of hepatocyte necrosis associated with thrombocytopenia, is termed *HELLP syndrome* (*h*emolysis, *e*levated *l*iver enzymes, and *l*ow *p*latelets). The second is *acute liver hemorrhage and rupture*. The histopathological findings in preeclampsia-associated liver disease include (1) deposition of fibrin along the sinusoids and portal tract capillaries, hepatic arterials, and portal veins; (2) periportal and portal tract hemorrhage; and (3) ischemic necrosis. In severe cases, diffuse areas of hemorrhage and necrosis are seen. Large hematomas form beneath the capsule and may rupture. Hepatic necrosis is often associated with necrosis in other organs, including the brain and kidney.
Clinical Features, Diagnosis, and Treatment Thrombocytopenia and increased AST and ALT levels are hallmarks of the HELLP syndrome. Renal dysfunction is virtually universal. Hemolysis is due to microangiopathic hemolytic anemia. Ascites is found in 10% of women with HELLP syndrome and is associated with a risk for congestive heart failure and adult respiratory distress syndrome. The abnormalities may begin suddenly and without warning. Close monitoring and rapid delivery are warranted in most cases. [266], [267] Treatment guidelines include close monitoring of the mother's platelet count, coagulation parameters, and disseminated intravascular coagulation screening panel, as well as AST and ALT levels. Hypertension is treated with magnesium sulfate, and fetal lung maturity is monitored. Patients with postpartum HELLP may benefit from corticosteroids. [268], [269] and [270] Preeclampsia and HELLP syndrome are associated with intrauterine growth retardation, sudden fetal death, and perinatal instability. [271], [272] Survival of the fetus is increased with prompt delivery. Maternal mortality is low (2.4%), possibly because of prompt delivery and rapid recovery. Liver necrosis generally heals rapidly after delivery, with little evidence of chronic liver disease. Women are at risk for recurrent preeclampsia and HELLP syndrome in subsequent pregnancies. Hepatic rupture resulting in serious hemorrhage occurs late in pregnancy or up to 48 hours postpartum. Many but not all cases are associated with the HELLP syndrome. [273] The sudden onset of right upper quadrant pain often heralds the onset of hemorrhage and rupture. Rupture often presents with sudden abdominal distention and shock but may rarely evolve over several days. [274], [275] Chest and shoulder pain are sometimes noted. Abdominal computed tomography is the most sensitive and specific method of detecting hepatic hemorrhage or rupture. Paracentesis will confirm intra-abdominal hemorrhage, and results will be negative in patients with contained hematomas. Supportive therapy is recommended for patients with contained hemorrhage, with emergency laparotomy if rupture of the Glisson capsule is suspected. Angiographic embolization has been performed with success. [276] Overall maternal and fetal mortality rates in hepatic rupture are high. Healing of contained hemorrhages occurs slowly, and recurrence in subsequent pregnancies is rare. [277]

Other Conditions

Spontaneous rupture of the spleen has been reported in pregnancy. [278] Sudden left upper quadrant pain associated with signs of intravascular volume depletion suggests the diagnosis. Most ruptures occur in the third trimester. Emergency surgery is indicated.

Rupture of a splenic artery aneurysm is rare, but as in splenic rupture, the risk for maternal and fetal mortality is high. [279] Patients may present with intravascular volume depletion with or without abdominal pain. Angiography confirms the diagnosis, and surgery is indicated.

Acute granulomatous peritonitis has been described as a cause of abdominal pain after cesarean section. [280] The mechanism appears to be premature rupture of fetal membranes or meconium spillage into the peritoneal cavity during cesarean section.

EVALUATION OF WOMEN WITH PELVIC PAIN

This section reviews the gynecologic diagnoses that should be considered in the evaluation of women presenting with acute and chronic abdominal pain (Table 52-5).

Acute Presentation
Pelvic inflammatory disease
Ectopic pregnancy
Mittelschmerz
Ruptured ovarian cyst
Corpus luteum
Ovarian endometrioma
Adnexal torsion[a]
Chronic Presentation
Endometriosis[b]
Ovarian cancer
Dysmenorrhea

[a]May present rarely as chronic pain.
[b]May present rarely as acute pain.

TABLE 52-5 Gynecologic Causes of Abdominal Pelvic Pain in Women of Reproductive Age

Pelvic Inflammatory Disease

Acute pelvic inflammatory disease (PID) is an inflammatory and infectious process of the upper genital tract, including the fallopian tubes, ovaries, and surrounding tissue. The incidence of PID is highest among women in their late teens and early twenties, with an annual rate of 1.5% to 2% in this age group. [281] The risk factors for the development of PID include sexual activity (especially with multiple partners), the presence of a contraceptive intrauterine device (IUD), and prior pelvic surgical instrumentation. [282] PID is less likely to develop in women using oral contraceptives because of the thickening effect of progestins on cervical mucus, which results in a more effective barrier to infectious organisms. [283]

Cervicitis, frequently caused by the sexually transmitted organisms *Chlamydia trachomatis* and *Neisseria gonorrhoeae* often precedes the development of PID. [284] PID commonly develops immediately before or after menstruation because of the physiological loss of the cervical mucus mechanical barrier around the time of menses and because the growth of *N gonorrhoeae*, particularly virulent gonococcal phenotypes, may be stimulated during the secretory phase (second half) of the menstrual cycle. [285] PID results from the ascent of cervicovaginal bacteria along the mucosal surfaces of the uterus and fallopian tubes. Salpingitis (tubal infection) is the most characteristic feature of ascending infection. Infectious organisms may also migrate into the peritoneal cavity, causing peritonitis, intra-abdominal abscess formation; in rare advanced cases, organisms may spread hematogenously.

Patients with PID classically present with the triad of fever, lower abdominal pain, and vaginal discharge. Less acute cases may present with a history of continuous lower abdominal discomfort exacerbated by movement and intercourse; the presence of fever and vaginal discharge varies. In patients with salpingitis, bimanual examination commonly reveals marked tenderness over the region of the cervix, uterus, and both adnexa. Unilateral tenderness occurs in fewer than 10% of women with PID, and when present, it should prompt an investigation for other diagnoses. A Gram stain of cervical secretions may help determine the presence of gonococcal cervicitis by demonstrating leukocytosis and gram-negative intracellular diplococci. Cervical secretions should be assessed for *N gonorrhoeae* and *C trachomatis* by direct culture, fluorescent antibody smear, or enzyme-linked immunoassay. [284] Culdocentesis and pelvic ultrasonography may help exclude other causes. Diagnostic laparoscopy may expedite an accurate diagnosis, especially if appendicitis or ectopic pregnancy is suspected.

Most patients with PID, including patients with tubal-ovarian or pelvic abscesses, can be successfully treated with antibiotic regimens as recommended by the Centers for Disease Control: parenteral cefoxitin plus oral doxycycline or intravenous clindamycin plus gentamicin. [285], [286] and [287] When PID is associated with an IUD, the IUD should be removed.

Ectopic Pregnancy

The incidence of ectopic pregnancy has been steadily rising since 1970. In the United States, approximately 2% of pregnancies are ectopic. [288], [289] Ectopic pregnancy remains the leading cause of maternal mortality despite improved treatments and earlier diagnosis. Risk factors for ectopic pregnancy include a history of PID, previous pelvic surgery, use of progestin-only oral contraceptives, use of an IUD, previous ectopic pregnancy, and use of advanced reproductive technologies, including menopausal gonadotropin-induced ovulation and in vitro fertilization and tubal embryo transfer. Based on the parallel rise in the incidence of ectopic pregnancy and PID during the past two decades, PID is felt to be a major factor in the development of subsequent ectopic pregnancy. [290] One proposed mechanism is that PID causes damage to the ciliated portion of the fallopian tubes, resulting in abnormal transport of the ovum. Ectopic pregnancies may occur in a variety of extrauterine sites, including the fallopian tubes, ovary, cervix, and abdominal cavity. The fallopian tubes are by far the most common extrauterine implantation site.

Patients commonly present with abdominal pain, amenorrhea or abnormal uterine bleeding, and symptoms of early pregnancy, including breast tenderness and nausea. The possibility of intra-abdominal bleeding should always be entertained in patients presenting with syncope, orthostatic hypotension, or pain referred to the shoulder as a result of irritation of the diaphragm from hemoperitoneum. Although virtually all patients have evidence of abdominal or adnexal tenderness, only 50% of patients have an adnexal mass palpable on pelvic examination. Intra-abdominal bleeding may be rapidly determined by culdocentesis, although this may occasionally yield false-negative or false-positive results. Early diagnosis of ectopic pregnancy is facilitated by the hCG assay, measurement of the progesterone level, and high-resolution transvaginal ultrasonography. [289], [291], [292] and [293]

Surgery is the standard therapy for diagnosed or suspected ectopic pregnancy, although newer medical regimens are effective in selected cases. The stability of the patient, size of the ectopic pregnancy, and the patient's desire for future fertility determine the therapeutic approach. In cases of severe hemodynamic compromise, immediate exploratory laparotomy should be performed. In more stable settings, therapeutic laparoscopic options are considered. Selected patients with smaller, unruptured ectopic pregnancies may be treated medically with methotrexate. [294], [295] and [296]

Midcycle Ovulatory Pain

Pain resulting from intraperitoneal bleeding associated with the physiological rupture of an ovarian follicle can range from a mild sensation of discomfort experienced on a monthly basis to severe pain prompting emergency evaluation. Ovulatory pain (i.e., mittelschmerz) is often rather sudden, sharp, and localized to one of the lower quadrants. It usually subsides over a period of several hours. The patient's menstrual history frequently reveals regular menstrual cycles with midcycle pain. Occasionally, normal ovulation may be associated with intraperitoneal bleeding and irritation sufficient to promote symptoms of severe pain, including peritoneal signs, and other diagnoses, such as adnexal torsion, PID, and acute appendicitis, must be excluded. Ovulatory pain in patients receiving anticoagulation, with clotting disorders, or on large doses of nonsteroidal antiinflammatory agents may be more severe because of increased bleeding from the ruptured follicle. When patients have severe symptoms, consideration should be given to ovarian suppression with oral contraceptives.

Ruptured Ovarian Cysts

Acute abdominal pain may be caused by a ruptured corpus luteum cyst with hemorrhage or by rupture and leakage of an ovarian endometrioma. The diameter of a normal corpus luteum commonly varies between 2 and 4 cm. Enlargement and cystic change of the corpus luteum are common and generally painless unless rupture or bleeding into the cystic cavity occurs, with sudden distention of the capsule. With hemorrhage, degeneration of the corpus luteum and subsequent loss of ovarian progesterone and estrogen secretion result in the onset of menstrual bleeding that is often irregular and abnormal. Like ovulatory pain, symptomatic rupture of a corpus luteum is more common in patients on anticoagulant therapy or in patients taking large doses of nonsteroidal antiinflammatory agents.

Hemorrhage into the corpus luteum with subsequent rupture generally occurs late in the menstrual cycle, often immediately before menstruation. Patients may present with concomitant lower quadrant pain, delayed menses, abnormal uterine bleeding, and a slightly enlarged, tender adnexal mass. It is unusual for hemorrhage to result in an acute drop in the hemoglobin level or affect volume status, but significant hemoperitoneum may occur and be evident on culdocentesis.

Ovarian Torsion

Although torsion occasionally occurs in a normal adnexa, ovarian or adnexal torsion is most often associated with ovarian neoplasms, fallopian tube tumors, or paratubal structures, such as paramesonephric cysts. The most common ovarian neoplasms leading to ovarian torsion in women of reproductive age are the benign cystic teratoma and the common fibroma. [257], [258] Teratomas are often recognized by palpable adnexal enlargement on pelvic examination and a semisolid echogenic ultrasonic pattern, reflecting their frequent makeup of sebaceous material, hair, and dedifferentiated tissue. Pain from torsion of an ovarian tumor often presents as

sharp, severe discomfort that is brief, irregular, and repetitive if the torsion is intermittent and self-correcting. It is common for patients presenting with ovarian torsion to describe an acute onset of discomfort after rising or stooping. If rotation around the torsion axis is sufficient, vascular compromise develops, and relief from the discomfort is not obtained. With necrosis of ovarian tissue, the pain may become less precise in location and dull in character, and over a period of 4 to 6 hours, sufficient inflammation of the parietal peritoneum may develop to produce a generalized peritonitis with associated nausea, vomiting, and ileus. Fever often appears late in this process. Although ovarian torsion and necrosis generally present as a rapidly developing acute process, torsion and necrosis of an ovarian neoplasm can occasionally be walled off by omentum or bowel, so that the patient's discomfort becomes low-grade and chronic.

Dysmenorrhea

The term *dysmenorrhea* refers to pelvic pain associated with menstruation. Dysmenorrhea is a spectrum ranging from mild physiological discomfort to severe, debilitating symptoms. Dysmenorrhea is characterized as primary if it has no organic cause and in this case usually begins 2 to 3 years after menarche. Dysmenorrhea is secondary if it is caused by pelvic pathology such as endometriosis, PID, or congenital abnormalities. [297]

Dysmenorrhea appears to be a combined result of hormonal effects, prostaglandins, myometrial activity, and psychological factors. [297] Progesterone stimulation of previously estrogen-primed endometrium and myometrium is necessary for the development of primary dysmenorrhea. Progesterone increases the synthesis of prostaglandins F_{2a} and E_2, which can be found in higher levels in endometrium and in menstrual fluid in patients with dysmenorrhea. [297], [298] Endometrial release of leukotrienes C_4, D_4, and E_4 is also increased. [297] Prostaglandins appear to mediate symptoms of discomfort by enhancing uterine contractility and sensitizing pain fibers to both mechanical and chemical stimuli. [299] Patients with secondary dysmenorrhea have acquired causes of menstrual discomfort, such as abnormalities that inhibit the outflow of blood, including endometrial polyps, pedunculated submucous myomas, and cervical stenosis. Cervical stenosis most commonly results from prior cervical instrumentation, including conization or therapeutic abortion. Secondary dysmenorrhea also may be caused by IUDs and by endometriosis. Adenomyosis, which is an extension of the glandular portion of the endometrium into the underlying myometrium, occurs primarily in multiparous women in their late thirties and early forties. It may cause increased menstrual flow and a symmetrically enlarged uterus. Pelvic endometriosis and its association with secondary dysmenorrhea are discussed in the next section.

Education, including an understanding of normal ovarian physiology, is an important part of the therapy for primary dysmenorrhea. Medical therapy is indicated for patients in whom symptoms of dysmenorrhea interfere with normal activities. Nonsteroidal antiinflammatory medications such as ibuprofen inhibit prostaglandin production and are effective in most patients. Oral contraceptives suppress ovulation and are also effective in the treatment of primary dysmenorrhea. The treatment of secondary dysmenorrhea depends on the cause. Uterine anomalies leading to outflow obstruction should be corrected to prevent retrograde menstruation, which predisposes to the development of endometriosis. Endometrial polyps and pedunculated submucous myomas may be resected surgically.

Endometriosis

Endometriosis is an important cause of lower abdominal pain and gastrointestinal symptoms in women of reproductive age. [300] Endometriosis is not found before menarche and is rare during menopause. The actual prevalence is unknown, but the condition is found in approximately 3% of all fertile women, 20% to 40% of infertile women, and as many as 50% of patients with pelvic symptoms.

Endometriosis is characterized by the ectopic growth of hormonally responsive endometrium outside the uterus. The most common location for endometriosis is the pelvic peritoneum in close proximity to the fallopian tube, cul-de-sac, ovaries, and uterosacral ligament. [301] The most widely accepted mechanism by which endometriosis develops is retrograde menstruation through the fallopian tube into the peritoneum, with subsequent growth of the displaced cells in the pelvis. [302] Retrograde menstruation may be a frequent and perhaps universal occurrence. [303] It is not known why implantation and growth of ectopic endometrium occur in some women. Endometriosis at distant sites such as the pericardium or pleurae is best explained by the vascular dissemination of endometrial cells.

Risk factors for endometriosis include heavy, frequent, or prolonged menstrual periods and increased estrogen exposure. [304] Endometriomas appear to be monoclonal in origin. [305] Data support a genetic basis for endometriosis in a small percentage of overall cases. Genes for factors modulating estrogen exposure have been studied, most significantly aromatase P450. [306] Candidate genes involved in detoxification, including glutathione *S*-transferase (GST) and *N*-acetyltransferase 2 (NAT2), have also been studied. Proinflammatory cytokines, including interleukin-1 (IL-1), IL-6, IL-8, and tumor necrosis factor-a (TNF-a), have been implicated in the pathogenesis. [307]

Endometriotic implants have a variety of phenotypic appearances, including the classical nodular "powder burn" implants, red lesions with a vesicular appearance, and papular implants with a whitish, yellow, or nonpigmented appearance. There is little correlation between the phenotypic appearance of a lesion and the specific symptoms or response to therapy. However, the deeply infiltrating lesions are the ones most often associated with pain. [308]

It has been estimated that endometriotic involvement of the gastrointestinal system occurs in as many as 37% of women with documented pelvic endometriosis. [309] Endometriosis involving the gastrointestinal tract can be localized by frequency of occurrence to the rectosigmoid (72%), rectovaginal septum (13%), small bowel (7%), cecum (4%), appendix (3%), and other intestinal sites (0.5%). [310], [311] and [312] Most gastrointestinal lesions are limited to superficial pinpoint involvement over the rectosigmoid and do not cause significant symptoms. Occasionally, a larger surface area of the bowel wall may be involved with a deeply invasive lesion that is frequently accompanied by extensive fibrosis. The characteristic gross appearance of an endometrioma of the bowel wall is that of an indurated, ill-defined tumor with a glistening gray color on cross section. [313] The tumor and associated fibrosis may be circumferential and narrow the lumen, resulting in symptoms of bowel obstruction. The lesions are usually confined to the serosal and muscular layers, although they may project into the lumen as a polypoid mass covered by intact mucosa. Their appearance on barium study may closely mimic that of carcinoma. Rarely, cancer can arise in endometriosis. [314]

Patients with endometriosis may present with the hallmark symptom of pain and cramps beginning at or slightly before the onset of menses. Pain also may be constant and unrelated to menses. Patients often relate progressively worsening dysmenorrhea and a poor response to prostaglandin synthetase inhibitors. Patients may note that symptoms improve during pregnancy or with oral contraceptive therapy.

Gastrointestinal symptoms are common in patients with endometriosis. The most common gastrointestinal symptoms are rectal pain, constipation, and painful defecation. Symptoms may be referred as a consequence of involvement of the uterosacral ligaments and rectovaginal septum or of compression of the rectum by enlarging endometrial implants in the rectovaginal septum. Lesions within the bowel may result in symptoms of large bowel obstruction. [313] Rarely, endometrial implants may invade the mucosa, resulting in cyclic rectal bleeding at the time of menses. [315], [316] Other rare gastrointestinal complications associated with endometriosis include intussusception, [317] small bowel obstruction, [317], [318] hemorrhagic ascites, [319] protein-losing enteropathy, [320] and bowel perforation. [321] Appendiceal endometriosis is usually asymptomatic and found incidentally, but it may result in perforation or intussusception. [322] Superficial implants on the serosal surface of the bowel rarely cause symptoms.

Patients with endometriosis may have nodular thickening and tenderness along the uterosacral ligaments, on the posterior surface of the uterus, and in the cul-de-sac. Pelvic examination around the time of menstruation can be particularly revealing because nodularity and tenderness tend to be more conspicuous at that time. Because endometriosis tends to involve structures in the posterior cul-de-sac, pelvic findings are best appreciated on rectovaginal examination.

The evaluation of intestinal tract endometriosis if often difficult and inexact. Radiographic studies are useful in demonstrating external compression, angulation, and stricture. However, these findings are nonspecific, and it may be impossible to exclude malignancy based on the radiographic appearance alone. Endoscopic evaluation may be valuable to exclude malignancy and to diagnose the rare occurrence of mucosal endometriosis resulting in hematochezia. Endoscopic ultrasonography may be useful in documenting the depth of invasion. [323] Preoperative evaluation by endoscopy or radiography is advised in all patients with bowel symptoms preoperatively. If intestinal involvement is documented, preoperative bowel preparation can be performed.

The treatment of endometriosis involves the pharmacological suppression of ovarian function or surgical ablation or removal of the lesions. Pharmacological agents used in the medical suppression of endometriosis include combination oral contraceptives, progestational agents, danazol, and gonadotropin-releasing hormone analogs. [324] A variety of laparoscopic and nonlaparoscopic surgical options are available. [325], [326] and [327]

Ovarian Cancer

Ovarian cancer the most common gynecologic malignancy in the United States. [328] It is most prevalent in perimenopausal and postmenopausal women. The survival rate has improved but remains low, possibly because of advanced local spread of the disease at the time of diagnosis in most patients. [329] The risk for the development of ovarian cancer is four times higher for women in the United States than for those in Japan. [330] The risk for Japanese women emigrating to the United States increases, strongly suggesting an environmental factor influencing disease development.

The cause of ovarian cancer is unknown. Epithelial neoplasms constitute 80% to 90% of cases, with the remainder being germ cell and sex cord/stromal cell tumors. [331] Risk factors for the development of ovarian cancer include a history of late childbearing, early menarche and late menopause, and infertility. [332] Three to five percent of cases are familial, with three hereditary syndromes identified: (1) breast and ovarian cancer, (2) HNPCC (Lynch II), and (3) site-specific ovarian cancer. [328], [333] Most cases of familial breast and ovarian cancer are linked to the BRCA1 gene. [334], [335] Repeated ovulatory stimulation is postulated to be a factor in the malignant transformation of ovarian epithelium. Supporting this hypothesis is the observation that ovarian cancer is more common in nulliparous women and less frequent in oral contraceptive users. [336]

Ovarian cancer usually spreads by contiguous invasion and by diffuse intraperitoneal seeding, commonly involving the serosal surfaces of the fallopian tubes, adjacent uterus, bladder, and rectum. Diffuse involvement of the intestinal serosa often results in severe gastrointestinal dysfunction. Intestinal loops may become rigid and inflexible because of encasement by diffuse serosal tumor nodules. Extensive invasion of the myenteric plexus may interrupt normal peristalsis, causing segmental intestinal atony. Encasement of the intestinal wall can lead to lumenal narrowing and obstruction. The incidence of malignant ascites in patients with ovarian cancer ranges from 35% at the time of initial surgical diagnosis to more than 62% in patients dying of ovarian cancer. When a postmenopausal woman presents with ascites, the possibility of ovarian malignancy must always be entertained. Although dissemination beyond the peritoneal cavity occurs infrequently, even in advanced cases, ovarian cancer has the capacity to metastasize to distant lymph nodes and organs. The most common distal sites include the lung and liver.

Patients usually present with nonspecific symptoms, including urinary symptoms, constipation, and abdominal pain. They may present with malignant ascites, abdominal or pelvic masses, or symptoms of gastrointestinal obstruction. Pain is usually a symptom of late-stage ovarian cancer but may be associated with ovarian torsion or rupture early in the course of the disease. When the malignancy involves adjacent pelvic and abdominal sites, traction or compression of the abdominal viscera may result in chronic discomfort.

Ultrasonography is useful in differentiating ovarian neoplasms from enlarged ovarian cysts. [337] Ultrasonography, computed tomography, and nuclear magnetic resonance imaging can also document evidence of tumor spread, such as ascites and metastasis to the liver and aortic lymph nodes. The finding of ureteral dilation may indicate retroperitoneal lymphatic involvement. The level of the tumor marker CA-125 may assist in differentiating malignant epithelial ovarian carcinomas from benign masses in postmenopausal women. In premenopausal women, an elevated CA-125 level may be seen in other conditions, such as endometriosis or liver disease. In addition, advanced ovarian cancer may occur in the absence of CA-125 at any age. [338] Surgical staging and resection/debulking are an important part of the treatment for ovarian cancer. [331] Patients with borderline and highly differentiated localized tumors have a good prognosis and do not require chemotherapy. Platinum-based chemotherapy regimens are recommended in most invasive cases. [339] The prognosis is poor for patients with recurrent disease.

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CHAPTER 53

David H. Alpers and Samuel Klein

APPROACH TO THE PATIENT REQUIRING NUTRITIONAL SUPPLEMENTATION

NUTRITIONAL ASSESSMENT

Most methods currently used to evaluate nutritional status are aimed at identifying specific nutrient deficiencies or protein-energy malnutrition. The diagnosis and treatment of specific nutrient deficiencies are usually straightforward. For example, potassium deficiency can be identified by measuring the serum potassium concentration and can be corrected by oral or intravenous potassium supplementation. In contrast, the diagnosis of protein-energy malnutrition is more complicated. Commonly used indicators of nutritional status have been validated by linking nutritional indicators with clinical outcome; patients with abnormal indicators have a worse outcome than patients with normal indicators. However, all indicators are influenced by illness or injury, so it is difficult to separate the contribution of malnutrition from that of the illness itself. Therefore, if nutritional status linked to the severity of disease, patients who are “malnourished” may have a poor outcome simply because they are sicker than patients who are “well nourished.” Moreover, nutritional therapy may not improve outcome unless malnutrition is an independent contributor to adverse medical events.

Nutritional Assessment Techniques

Specific Nutrient Deficiency A careful history and physical examination, routine blood tests, and selected laboratory tests based on the history and physical examination findings can be used to diagnose specific macronutrient, mineral, vitamin, and trace mineral deficiencies (Table 53-1, Table 53-2 and Table 53-3). [1]

TABLE 53-1 Some Physical Signs of Nutritional Deficiency

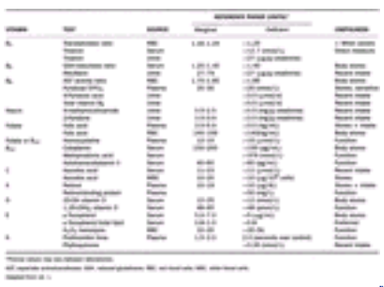


TABLE 53-2 Clinical Laboratory Tests for Detection of Vitamin Deficiency

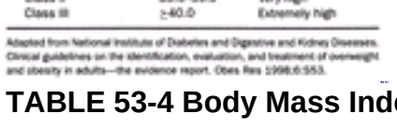


TABLE 53-3 Laboratory Detection of Micronutrient Mineral Deficiency

Protein-Energy Malnutrition Ideally, nutritional assessment is used to identify those patients who will benefit from nutritional support. However, the indicators currently used to assess protein-energy malnutrition have been validated by determining their relationship to clinical outcome rather than to the response of patients to nutritional therapy.

Body mass and composition. In general, unintentional weight loss is associated with an adverse clinical outcome. [2] However, the assessment of changes in body weight can be confounded by errors in recall and changes in body water. Anthropometry, the measurement of triceps and subscapular skinfold thickness, provides an index of body fat and muscle mass, which is compared with the values in standard tables. Although the interpretation of these data may be limited by differences in age and ethnicity between patients and the standard population, interrater variability, and hydration, markedly abnormal values (below the 5th percentile) usually predict a poor clinical outcome. Body muscle mass can also be assessed in the clinical setting by measuring the urinary creatinine excretion. [3] Patient values can be compared with those in tables indicating an expected amount of creatinine excretion in relationship to height in subjects consuming a meat-free diet. [1] The body mass index (BMI), which is obtained by dividing weight (kilograms) by the square of the height (square meters), can help identify patients at increased risk for medical complications (Table 53-4).

WEIGHT CATEGORY	BMI (kg/m^2)	RISK
Extremely underweight	<14.0	Extremely high
Underweight	14.1–18.4	Increased in smokers, chronic illness
Normal	18.5–24.9	Normal
Overweight	25–29.9	Increased
Obesity		
Class I	30.0–34.9	High
Class II	35.0–39.9	Very high
Class III	≥40.0	Extremely high

Adapted from National Institute of Diabetes and Digestive and Kidney Diseases. Clinical guidelines on the identification, evaluation, and treatment of overweight and obesity in adults—the evidence report. Obes Res 1998;6:553.

TABLE 53-4 Body Mass Index As a Measure of Associated Disease Risk

Serum protein concentrations. The concentration of several plasma proteins, particularly albumin, correlate with clinical outcome; a low plasma albumin concentration is associated with an increased incidence of medical complications. [4], [5] Illness or injury, however, rather than malnutrition per se, is responsible for hypoalbuminemia in sick patients. [6] Inflammation and injury decrease albumin synthesis, increase albumin degradation, and increase albumin transcapillary losses. Specific gastrointestinal, renal, and cardiac diseases can increase albumin losses through the gut or kidney. Wounds, burns, and peritonitis cause losses from surface tissues. Nutritional support itself does not usually alter serum albumin levels. [7] Serum concentrations of transferrin, retinol-binding protein, and thyroxine-binding prealbumin are also affected by inflammation, so that they are unreliable as a measure of protein-energy malnutrition.

Immune system function. The total circulating lymphocyte count and delayed cutaneous hypersensitivity to skin test antigens have been used as clinical indicators of malnutrition. Although depression of circulating lymphocytes (<200/mm [3]) and anergy assessed by delayed cutaneous hypersensitivity are associated with a poor clinical outcome, they are not specific for nutritional deficiency and are affected by specific diseases, injury, and certain drugs.

Multivariate analysis. Multiple regression analyses of selected indicators of nutritional status have been used to provide a score for individual patients and thereby identify those at increased risk for medical complications. [8], [9]

Subjective global assessment. The subjective global assessment involves a focused history and physical examination to determine the adequacy of nutrient intake and the possible effect of malnutrition on body composition and organ function (Table 53-5). [10], [11] In addition, it includes an analysis of the patient's future ability to ingest adequate nutrients. The findings of the history and physical examination are subjectively weighted to rank patients as well nourished, moderately malnourished, or severely malnourished. Patients who are judged to be severely malnourished are at high risk for medical complications and adverse outcome. [10], [12]

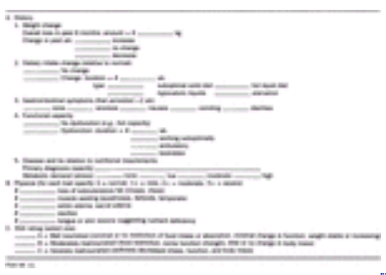


TABLE 53-5 Subjective Global Assessment (SGA) of Nutritional Status

There is no "gold standard" for determining nutritional status. All current approaches are influenced by the severity of illness and have been validated by assessing clinical outcome rather than by nutrition-specific parameters. Therefore, current nutritional assessment techniques identify sick patients. Although it is often assumed that "malnourished" patients will benefit from nutritional support, this assumption has not been documented in prospective randomized clinical trials. [13]

CHOOSING THE ROUTE FOR NUTRITIONAL SUPPORT

The decision to initiate nonvolitional nutrition support is complex and involves a clinical assessment of the patient's nutrient intake, body fat and protein mass, protein and energy requirements, severity of illness, and anticipated duration of inadequate volitional intake. Once it has been decided that nonvolitional nutrition support is indicated, the route of nutritional support must be considered. The usual course is to choose the oral or enteral route when this approach can be used effectively and safely. There are several reasons for this choice. First, enteral nutrition may provide nutrients that are not available from parenteral nutrition. The best example of this principle is the provision of short-chain fatty to the colonic mucosa via the bacterial degradation of fiber or unabsorbed carbohydrate. The provision of this important colonic energy source may maintain colonic function. [14] Second, it is felt that lumenal nutrients may prevent atrophy in the small intestinal mucosa. Atrophy occurs in rats fed by parenteral nutrition, but the importance of this phenomenon in humans is still uncertain. [15] Finally, enteral feeding may be safer than parenteral nutrition. [16], [17] and [18]

If oral or enteral feeding is chosen, intestinal motility and absorptive surface must be adequate. Other disorders that interfere with enteral feeding include severe diarrheal illnesses, extensive inflammatory bowel disease, acute gastrointestinal bleeding, conditions that will require abdominal surgery in the near future, obstructing small bowel or colon lesions, and acute inflammation of the pancreas or biliary tract. A number of guidelines have been developed that provide useful and practical information for the clinician. [19], [20] The guidelines for use in specific disorders, such as acquired immunodeficiency syndrome (AIDS), are practical modifications of the generalized guidelines, with special emphasis on the problems of feeding patients with that disorder. [21] However, the clinician should not rely on a

set pattern of guidelines for decisions regarding nutritional support. The published guidelines should be used as resource material to aid in the clinical judgment required for each patient.

ENTERAL NUTRITION

The availability of so many different types of whole-food diets and defined formulas makes it possible for many hospitalized patients to receive adequate nutrient intake by oral or enteral tube feedings.

Whole-food diets include a regular hospital diet and diets modified in either consistency (e.g., clear liquid, full liquid, pureed, and soft diets) or nutrient content (e.g., low-residue, low-fat, low-sodium, low-protein, and high-fiber diets). In many cases, nutrient intake can be increased by encouraging the ingestion of ordinary food and by avoiding missed meals and unpalatable diets. Many patients eat more if they are given assistance at mealtime and provided with desired foods and if some food supplied by relatives and friends is allowed.

Defined formulas are commercially made products with a known “defined” nutrient composition (Table 53-6). These formulas can be divided into five categories: feeding modules, monomeric formulas, oligomeric formulas, polymeric formulas, and disease-specific formulas. Feeding modules consist of single nutrients, such as protein (intact protein, hydrolyzed protein, or crystalline amino acids), carbohydrate (glucose polymers), or fat (long-chain or medium-chain triglyceride), that can be used to supplement a specific diet or be part of a modular enteral system composed of several nutrient modules.

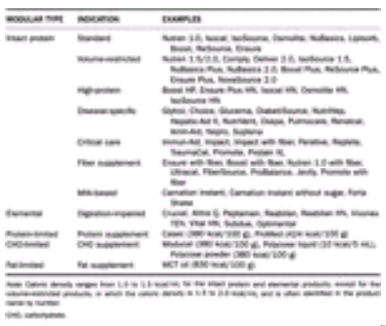


TABLE 53-6 Classification of Commercially Available Enteral Nutrition Products

Monomeric (elemental) formulas contain nitrogen in the form of free amino acids and usually only small amounts of fat (<5% of total calories). Therefore, these formulas can be useful in patients who require an extremely low-fat diet. However, monomeric formulas are hyperosmolar, unpalatable, and more expensive than polymeric formulas, and in the absence of pancreatic insufficiency, they are not absorbed better than oligomeric or polymeric formulas. [22]

Oligomeric (semi-elemental) formulas contain hydrolyzed protein in the form of small peptides and sometimes free amino acids. Fat represents 5% to 40% of energy content as either long-chain triglycerides or a combination of long-chain and medium-chain triglycerides. Data from several studies suggest that oligomeric diets can reduce diarrhea and other gastrointestinal side effects in critically ill patients [23] and in patients receiving abdominal irradiation and chemotherapy. [24], [25] However, the purported clinical benefits of oligomeric formulas require confirmation in large prospective clinical trials before precise recommendations regarding their use can be made.

Polymeric formulas contain nitrogen in the form of whole proteins. These formulas can be categorized into three groups: blenderized food formulas, milk-based formulas, and lactose-free formulas. Milk-based formulas usually contain milk as a source of protein and fat and tend to be more palatable than other defined diets. Although milk-based formulas can be problematic for lactose-intolerant patients, they are often tolerated when infused continuously to decrease the load of lactose delivered to the intestine at any one time. Lactose-free formulas are the most commonly used polymeric formulas in hospitalized patients, and many modifications in protein and fat content are available. Disease-specific formulas have been designed for patients who have specific illnesses, including hepatic insufficiency, renal insufficiency, pulmonary insufficiency, diabetes, and severe metabolic stress. In general, the clinical superiority of these formulas over less expensive, standard polymeric enteral formulas remains controversial. However, branched-chain amino acid–enriched formulas may permit greater nitrogen intake in selected patients with chronic protein-intolerant hepatic encephalopathy (see section “ Nutritional Support in the Hospitalized Patient with Gastrointestinal Disease ”), and renal failure formulas contain much smaller amounts of sodium, potassium, phosphorus, and magnesium than standard formulas.

A positive caloric balance is necessary to optimize and sometimes to achieve positive nitrogen balance and to incorporate amino acids into new protein. To achieve this goal, the ingestion of about 25 to 35 kcal/g of protein (~150–200 kcal/g of nitrogen) is required. This ratio is based on the derivation of 10% to 15% of caloric needs during catabolism from protein degradation.

Oral Rehydration Therapy

Patients with protracted vomiting or diarrhea can lose excessive amounts of fluid and electrolytes. Oral rehydration therapy enhances sodium and water absorption by stimulating sodium/glucose cotransport in the small intestine. Oral rehydration therapy has been demonstrated to have considerable clinical benefits in patients with severe gastrointestinal fluid and mineral losses, such as those with cholera-induced diarrhea, high-output ostomies, and short bowel syndrome. [26], [27], [28], [29], [30], [31] and [32]

A variety of oral rehydration solutions are currently available (Table 53-7). The World Health Organization developed a solution based on the results of studies in patients with cholera. [31] The glucose component of an oral rehydration solution should be provided at concentrations between 70 and 150 mmol/L. Higher concentrations of glucose can be deleterious because the increase in osmolality may reduce water absorption. Making the solution hypotonic by replacing glucose with rice solids or other polymeric forms can decrease stool output compared with isotonic solutions. [24], [25], [26] and [27] In patients with short bowel syndrome ending in a jejunostomy, net sodium and fluid balance is related to jejunal length. Patients with less than 100 cm of jejunum are usually net secretors, whereas those with more than 100 cm of jejunum are usually net absorbers. [32] The use of oral rehydration therapy can markedly decrease ostomy output and convert some patients from net secretors to net absorbers. In these patients, the consumption of electrolyte replacement drinks containing 60 mmol or less of sodium per liter causes a negative sodium and fluid balance, whereas the balance becomes positive after the ingestion of solutions containing 90 mEq/L and is maximal at a concentrations of 120 to 160 mEq/L. [33] Sport drinks and commercially made defined liquid formula diets do not contain an adequate amount of sodium for maximal fluid absorption in patients with severe short bowel syndrome and a jejunostomy.

TABLE 53-7 Characteristics of Selected Oral Rehydration Solutions

Tube Feeding

General Principles Enteral tube feeding is useful in patients who have a functional gastrointestinal tract but who cannot or will not ingest adequate nutrients. Many

approaches to tube feeding are possible (e.g., nasogastric, nasoduodenal, nasojejunal, gastrostomy, jejunostomy, pharyngostomy, esophagostomy tubes) and depend on physician experience, clinical prognosis, anticipated duration of feeding, patency and motility of the gut, risk for aspiration of gastric contents, and patient preference. Short-term feeding (<6 weeks) can be achieved by the placement of a soft, small-bore nasogastric or nasoenteric feeding tube. These tubes are made of silicone or polyurethane and can be left in place without the tissue irritation or pressure necrosis caused by larger polyvinyl chloride tubes. Many patients are able to eat with the tube in place, which permits the use of tube feeding to supplement oral intake. Nasogastric feeding is usually the most appropriate route, but orogastric feeding can be used in cases of nasal injury or gross nasal deformity. Nasoduodenal or nasojejunal feeding is useful in patients with gastroparesis. In addition, by taking advantage of the barrier function of the pyloric and gastroesophageal sphincters, nasoenteric feeding may decrease the incidence of aspiration in high-risk patients. However, the prevention of aspiration by feeding beyond the pylorus has not been proved in clinical trials. Most patients who require long-term tube feeding (>6 weeks) should have a gastrostomy or jejunostomy tube placed. These tubes can be placed endoscopically, radiologically, or surgically, depending on the clinical situation and local expertise (see Chapter 146). Percutaneous endoscopic gastrostomy can be performed within 30 minutes and is successfully completed in more than 90% of attempts. [34] Gastrostomy tubes can be placed percutaneously without endoscopy by inserting the catheter directly into the stomach via a peel-away sheath introduced over a previously placed J-wire guide. [35] This approach is particularly useful in patients with an obstructing lesion of the esophagus or hypopharynx that prevents passage of an endoscope or a gastrostomy tube bumper. A summation of data from several large series suggests that percutaneous endoscopic gastrostomy tube placement is associated with mortality in 0.5%, major complications (peristomal leakage with peritonitis, necrotizing fasciitis of the anterior abdominal wall, gastric hemorrhage) in 1%, and minor complications (minor wound infections, stomal leaks, tube extrusion or migration, aspiration, gastrocolic fistula, ileus, fever) in 8% of patients. [36] Jejunal tube placement can be achieved by threading a tube through an existing gastrostomy or by percutaneous endoscopic jejunostomy in patients with previous partial or total gastrectomy. [37], [38], [39], [40] and [41] Surgical gastrostomy is more expensive than percutaneous endoscopic gastrostomy unless the tube is placed during an operation performed for another reason. A jejunostomy tube may also be placed at the time of abdominal surgery and consists of a subserosal tunnel or needle catheter jejunostomy. Gastrostomy and jejunostomy tubes can also be placed by using laparoscopic techniques.

Administration of Tube Feeding Patients with feeding tubes in their stomach can often tolerate intermittent gravity feedings, in which the total amount of daily formula is divided into four to six equal portions that are infused by gravity over 30 to 60 minutes. The patient's upper body should be elevated by more than 30° during and for at least 1 hour after feeding. Intermittent feedings are useful for patients who cannot be positioned with the head of the bed continuously elevated or who require greater freedom from feeding. However, patients who experience nausea and early satiety with bolus gravity feedings may require continuous infusion at a slower rate. Patients who have gastroparesis often tolerate gastric tube feedings when they are started at a slow rate (e.g., 10 mL/h) and advanced by small increments (e.g., 10 mL/h every 8–12 hours). However, patients with severe gastroparesis require passage of the feeding tube tip past the ligament of Treitz. Continuous infusion should always be used when feeding directly into the duodenum or jejunum to avoid distention, abdominal pain, and dumping syndrome.

Complications Mechanical, metabolic, and gastrointestinal complications can occur in patients receiving tube feedings. Although the placement of nasogastric tubes is usually safe, tubes can be misplaced, particularly in unconscious patients. Intubation of the tracheobronchial tree has been reported in up to 15% of patients, and intracranial placement can occur in patients with skull fractures. Feeding tubes can cause nasopharyngeal erosions, pharyngitis, sinusitis, otitis media, pneumothorax, and gastrointestinal tract perforation. Small-diameter nasogastric tubes (e.g., 8F or 9F) are usually used to lessen nasopharyngeal irritation. These small tubes often become occluded by inspissated feedings or pulverized medications given through the tube. Frequent flushing of the tube with 30 to 60 mL of water and avoiding the administration of pill fragments whenever possible help maintain patency. Many techniques for unclogging tubes have been published and include the gentle infusion of small quantities of carbonated beverage, pancreatic enzymes, and 95% ethyl alcohol into the tube. In addition, commercially made products can be obtained that either dissolve or mechanically remove the obstruction. Metabolic complications, such as hypokalemia, hyponatremia, hypophosphatemia, and hyperglycemia can occur. [40] Patients should be monitored for metabolic abnormalities, particularly severely malnourished patients during the initial refeeding period. Gastrointestinal side effects of tube feedings include nausea and vomiting, pulmonary aspiration, abdominal pain, and diarrhea. Diarrhea is the most common complication and occurs in 30% to 50% of critically ill patients receiving tube feedings. Diarrhea is often caused by antibiotic use [41] and the use of liquid medications and elixirs that contain nonabsorbable carbohydrates, such as sorbitol. [42] Patients with normal gastrointestinal tract function can tolerate larger volumes of tube feedings. In one study, tube feeding did not cause diarrhea in healthy subjects until it was given at rates exceeding 200 mL/h. [43] Several case reports have also documented an association between small bowel necrosis and jejunal feedings in critically ill patients. [44] Most of these patients were receiving pressors to maintain an adequate blood pressure, suggesting that the increased oxygen requirements associated with feeding can cause ischemia in patients whose intestinal blood flow is unable to increase.

PARENTERAL NUTRITION

In some patients, enteral feedings are either contraindicated or cannot be provided in sufficient quantities to meet nutritional requirements. In these cases, parenteral nutritional support can be a valuable adjunctive and sometimes life-saving therapy. Certainly, patients unable to ingest "adequate" nutrients for a "prolonged" period of time require nutritional therapy to prevent the adverse effects of malnutrition. However, the precise definitions of "adequate" and "prolonged" are not clear and are likely to differ from patient to patient depending on the amount of body energy stores and lean body mass, the presence of preexisting medical illnesses, and the level of metabolic stress. The use of parenteral nutrition is often recommended if enteral intake has been, or is anticipated to be, inadequate for 5 to 10 days. [45] However, carefully performed, well-designed prospective randomized clinical trials to support the efficacy of this approach for many clinical situations are few in number. [46] Therefore, the use of parenteral nutrition requires a careful integration of data from pertinent clinical trials, clinical expertise in the illness or injury being treated, a reasonable estimate of the anticipated duration of inadequate food intake, clinical expertise in nutritional therapy, and input from patients and their families.

Central Parenteral Nutrition

General Principles The ability to use a central vein to supply nutrient requirements for growth and maintenance was realized more than 30 years ago when catheters were inserted into the superior vena cava of beagle dogs to provide the sole source of nutrients for 72 to 256 days. [47] Today, the use of central parenteral nutrition (CPN), also known as *total parenteral nutrition (TPN),* is commonplace. Although percutaneous infraclavicular subclavian vein catheterization with advancement of the catheter tip to the junction of the superior vena cava and right atrium is the most commonly used technique for catheter placement, many other approaches have been successfully performed when the subclavian vein is not accessible, including internal jugular, basilic, saphenous, and femoral veins and even thoracotomy with direct insertion into the right azygos vein [48] or the right atrial appendage. [49] In addition, peripherally inserted central venous catheters can be used to provide CPN. In this approach, insertion by a physician is not required, and the technique cannot cause pneumothorax or traumatic injury to arteries or nerves, but it is associated with an increased risk for thrombosis if the catheter tip is placed proximal to the superior vena cava. [50] Parenteral nutrient solutions provide all the basic nutrient requirements, including fluids, proteins, carbohydrates, fats, minerals, trace elements, and vitamins (Table 53-8). The lipid component can be piggybacked to the primary nutrient mixture, or a total nutrient admixture can be prepared, which reduces handling costs and potential breaks in sterility. However, total nutrient admixtures prevent visible inspection for particulate matter. Invisible calcium and phosphorus precipitates have caused fatal pulmonary emboli in patients receiving parenteral nutrition. [51] Therefore, pharmacy standards regarding physical-chemical compatibility should be closely followed in all patients receiving CPN, and in-line filters should be used with all parenteral nutrient solutions. Even a careful inspection of clear nutrient solutions cannot detect small microprecipitates, which can obstruct very small pulmonary capillaries. [52]

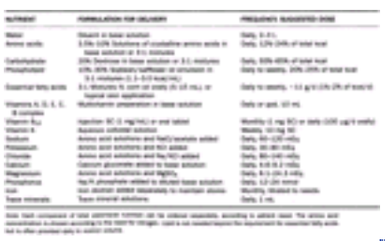


TABLE 53-8 Delivery of Nutrients During Total (Central) Parenteral Nutrition

The specific formulation prescribed for a patient depends on the patient's estimated nutrient requirements and ability to tolerate specific nutrients without adverse effects. The patient's protein, energy, and fluid requirements are the most important initial considerations in designing the appropriate parenteral formulation. Protein requirements are met by infusing standard solutions composed of crystalline amino acids containing 40% to 50% essential and 50% to 60% nonessential amino acids, usually with little or no glutamine, glutamate, aspartate, asparagine, tyrosine, and cysteine. Most hospitalized patients require between 1.0 and 1.5 g of protein per kilogram of body weight per day. Some amino acid solutions have been modified for specific disease states, such as those enriched in branched-chain amino acids, advocated for patients with hepatic encephalopathy, and those containing 67% to 100% essential amino acids, advocated for patients with renal insufficiency. However, the clinical superiority of these formulations over standard amino acid solutions has not been well documented in clinical trials. [53], [54] The major sources of energy are glucose and lipid. However, infused amino acids are also oxidized and should be included in the estimate of energy provided as part of the parenteral formulation. Glucose (dextrose) is usually the predominant energy source in CPN formulations and is the required fuel for erythrocytes, white blood cells, bone marrow, and renal medulla (~40 g/d) and the preferred fuel for the brain (~120 g/d). Each gram of hydrated dextrose provides 3.4 kcal and is readily oxidized unless

excessive amounts are infused. In stable postoperative patients, increasing the rate of glucose infusion up to, but not more than, 7 mg/kg per minute increases the rate of glucose oxidation. [55], [56] Lipid emulsions contain soybean oil or a combination of soybean and safflower oil triglycerides, egg yolk phospholipids as an emulsifying agent, and glycerin to maintain isosmolarity. These emulsions provide energy and are a source of essential fatty acids, linoleic and linolenic acids. Lipid calories are as effective as additional glucose calories in conserving body nitrogen economy and supporting protein metabolism once absolute tissue requirements for glucose have been met. [57], [58] The optimal percentage of calories that should be infused as fat is not known. At least 5% of total calories should be given as lipids to prevent essential fatty acid deficiency. The rate of lipid emulsion infusion should probably not exceed 0.7 kcal/kg per hour because most complications associated with lipid emulsion infusions have been reported when more than 1.0 kcal/kg per hour (0.11 g/kg per hour) was provided. [59] Lipid emulsion infusion can cause pulmonary dysfunction, [60] hepatic phospholipidosis, [61] impaired immune system function, [62] pancreatitis, [63] decreased platelet aggregation, [64] fat overload syndrome, [65] and hypersensitivity reactions. [66] Together, carbohydrate and lipid should provide 25 to 35 kcal/g of protein to optimize incorporation of amino acids into new protein.
Complications Mechanical, infectious, metabolic, and gastrointestinal complications are associated with the use of CPN. Administering CPN under the guidance of experienced teams decreases the incidence of most complications. [67] Percutaneous insertion of a central venous catheter can damage local structures and cause pneumothorax, brachial plexus injury, subclavian and carotid artery puncture, hemothorax, thoracic duct injury, and chylothorax. In addition, the catheter can be sheared off, become occluded, or cause subclavian vein or superior vena cava thrombosis. [68], [69] and [70] Catheter-related sepsis is the most common serious complication in patients receiving CPN; even when meticulous care is provided, catheter-related sepsis occurs in up to 4% of hospitalized patients. [71] Multiple-lumen central catheters that are used for CPN are associated with an increased incidence of infections compared with single-lumen catheters. [71] Catheter-related sepsis should be considered in all patients in whom a fever or leukocytosis develops while they are receiving CPN. The catheter tunnel and exit site should be carefully inspected, and any exit site drainage should be cultured. Blood cultures should be obtained from the central line and a peripheral vein. The presence of subcutaneous infection along the catheter tunnel or clinical toxicity (e.g., hypotension) without another known source of infection is an indication for immediate catheter removal. If no evidence of tunnel infection or clinical toxicity is found, the catheter can remain in place, and empiric antibiotic therapy should be started through the central venous catheter. Blood cultures should be obtained again in 48 to 72 hours to ensure clearance of bacteremia. The catheter should be removed from patients who have blood cultures that are positive for fungus or *Staphylococcus aureus,* polymicrobial infection, persistent or recurrent catheter-related bacteremia, or persistent fever without another suspected source of infection. The metabolic complications observed in patients receiving parenteral nutrition are usually caused by inappropriate nutrient administration resulting in nutrient excesses or deficiencies or both. A blood glucose level above 200 mg/dL should be avoided because it is associated with leukocyte and complement dysfunction and may increase the risk for infection. [72] Blood glucose goals for most patients are 100 to 200 mg/dL initially and 100 to 150 mg/dL when stable. Blood glucose should be kept below 120 mg/dL in pregnant patients to avoid the complications of gestational diabetes and large-for-gestational-age births. Metabolic bone disease, including osteopenia and osteomalacia, has been observed in patients receiving long-term (>3 months) CPN. [73] Several hepatobiliary abnormalities have been observed in patients receiving CPN and are usually more severe and more frequent in infants than in adults. [74] Hepatic abnormalities include alterations in liver biochemistries, steatosis, steatohepatitis, lipidosis and phospholipidosis, cholestasis, fibrosis, and cirrhosis. Most hepatic abnormalities occur early and are transient, but in a small subset of patients receiving long-term (>16 weeks) CPN, progressive and more serious liver disease develops. [75] Biliary abnormalities include acalculous cholecystitis, gallbladder sludge, and cholelithiasis and usually occur in patients receiving relatively prolonged (>4 weeks) courses of CPN. Acalculous cholecystitis has been reported in about 5%, [76] cholelithiasis in about 30%, and gallbladder sludge in up to 100% [77] of patients receiving prolonged CPN. The pathogenesis of acalculous cholecystitis is unclear. Gallbladder stasis is an important contributing factor in the pathogenesis of gallbladder sludge and stones. Maintaining regular gallbladder contractions by either enteral feedings or cholecystokinin injections prevents gallbladder sludge and gallstone formation. [78], [79]

Peripheral Parenteral Nutrition

Peripheral parenteral nutrition (PPN) consists of a mixture of nutrients containing a final concentration of 5% to 10% dextrose, 2% to 5% amino acids, electrolytes, and vitamins and minerals. This crystalloid solution is mixed with a 10% or 20% lipid emulsion as an all-in-one admixture, or the lipid emulsion can be piggybacked to the distal port of the intravenous infusion line. So that adequate calories can be given in a reasonable volume, the lipid component usually provides approximately half of the total calories.

The use of a peripheral vein to provide nutritional support avoids many of the mechanical and infectious complications of CPN but is associated with a high risk for thrombophlebitis, which occurs in up to 94% of patients. [80] However, the use of PPN has become possible in many patients since safe isotonic lipid emulsions have been developed and a better understanding of the causes of PPN-induced thrombophlebitis has been acquired. Several factors are important in the pathogenesis of thrombophlebitis in patients receiving PPN: (1) the osmolality, pH, and lipid content of the PPN solution, and the presence of particulate matter; (2) the diameter, length, and composition of the catheter; (3) the duration, rate, and volume of the infusion; (4) the diameter and anatomic position of the vein; and (5) the insertion technique. [81], [82] Adherence to the following principles can increase the life of a single infusion site to more than 10 days in many patients: (1) Provide at least 50% of total energy as a lipid emulsion, add 500 to 1000 U of heparin and 5 mg of hydrocortisone per liter of solution, and add sodium hydroxide if needed to achieve a pH of 7.4; (2) insert a fine-bore 22- or 23-gauge polyvinyl pyrrolidine-coated polyurethane catheter in as large a vein as possible in the proximal forearm with use of a sterile technique; (3) place a 5-mg glyceryl trinitrate ointment patch over the infusion site; (4) infuse the PPN solution with a volumetric pump and keep the total infused volume below 3500 mL; and (5) filter the solution with an in-line 1.2-µm filter.

Home Parenteral Nutrition

The ability to provide parenteral nutrition at home was first demonstrated in 1970. [83] In 1992, approximately 40,000 patients (120 per 1 million residents) received home parenteral nutrition (HPN) in the United States, [84] a rate tenfold higher than the prevalence rate in Europe (1 per million to 12 per million residents). [84], [85] and [86] Patients with benign diseases have a much better outcome than those with terminal illnesses. Most HPN patients with benign gastrointestinal diseases achieved complete rehabilitation; only 5% of the total HPN days were spent in the hospital. [84] Approximately 5% of patients died each year of HPN-associated complications. [87] In contrast, more than half of the patients receiving HPN who had cancer or AIDS died within 6 months, and more than 80% had died at 1 year.

HPN is usually given through an implantable subcutaneous port or a catheter inserted in the subclavian vein and tunneled subcutaneously to exit on the anterior chest. Nutrient formulations are infused for 8 to 12 hours overnight to allow patients to be active during the day. Intravenous lipids may not be necessary for patients who eat and are able to absorb adequate amounts of fat, whereas others may receive 20% to 30% of their calories as a lipid emulsion. The protein, energy, electrolyte, vitamin, and trace mineral requirements are normal unless the disease process increases requirements or losses.

The most serious common complication is catheter-related sepsis, most often caused by *S aureus* or *Staphylococcus epidermidis*. In addition, venous thrombosis, chronic liver disease, cholelithiasis, and metabolic bone disease are associated with the prolonged use of HPN.

MICRONUTRIENT DEFICIENCY AND TREATMENT

Tissues that proliferate rapidly (skin, oral and gastrointestinal mucosa, hair, bone marrow) or in which metabolic rates are high (muscle, gastrointestinal mucosa) are at greatest risk for signs of deficiency at an early stage. Clues regarding deficiency come from a history of reduced intake or increased losses (or of conditions that make those situations more likely), physical examination findings, and results of laboratory tests that ideally determine body stores of the nutrient. The signs and symptoms of nutritional deficiency in adult patients are listed in Table 51-1. The laboratory values for the major vitamins and minerals are listed in Table 51-2 and Table 51-3. A more detailed discussion of these nutrients and the assessment of their status can be found in general references. [1], [88], [89]

Sodium

Deficiency Sodium depletion usually presents with symptoms of dehydration because of concomitant water loss. Severe depletion presents with nausea, vomiting, exhaustion, cramps, seizures, and ultimately cardiovascular collapse. On the physical examination, mucosal xerosis, sunken eyeballs, and mental confusion can be present, but none of these are specific findings. In older patients, other explanations are often present for all these signs. [90] Orthostatic hypotension is often used to estimate the intravascular volume and is defined as a systolic blood pressure below 20 mm Hg or a diastolic blood pressure below 10 mm Hg within 3 minutes after standing, with or without a rise in the heart rate of 5 to 10 beats/min. [91] The signs of total body sodium excess are weight gain and edema. The presence of either of these would not reflect sodium deficiency. If sodium deficiency is the result of increased gastrointestinal or cutaneous losses, the urine osmolality is increased and the urine sodium concentration is less than 10 mEq/L. If sodium deficiency is the result of diuretic therapy, the urine osmolality is decreased and the urine sodium concentration is more than 10 mEq/L.
Assessment The serum sodium concentration primarily reflects water, not sodium, balance. Thus, any serum concentration is possible when deficiency is present, depending on the relative deficits of water and sodium. If hyponatremia is present but the cause is not apparent, the serum osmolality should be measured. In

hospitalized patients, hyponatremia is most often the result of dilution by excess free water, not depletion of sodium. The free water excess is usually caused by cardiac, hepatic, or renal insufficiency or by overly enthusiastic fluid administration and is accompanied by edema and weight gain. When normovolemia is present, the hyponatremia may be caused either by inappropriate secretion of antidiuretic hormone or by displacement of sodium osmoles with elevated glucose or urea as osmoles.

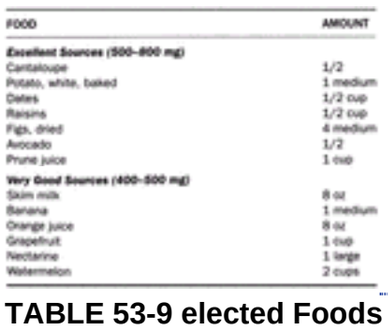
Treatment When signs of volume depletion are present, at least a 10% reduction in extracellular fluid volume has occurred. [92] The sodium deficit can be estimated as follows: Na deficit (mEq) = ([Na desired] - [Na observed]) × 0.6 × weight (kg). Sodium is usually replaced with NaCl (1 g = 17 mEq of Na), either orally, enterally, or parenterally. Symptomatic hyponatremia (e.g., seizures, lethargy) is treated with intravenous 0.9% NaCl or occasionally with hypertonic (3% or 5%) NaCl. Sodium infusion should be designed to increase the serum concentration by 1 mEq/h until it reaches 120 mEq/L. Too rapid restoration has been associated with permanent neurological deficits resulting from central pontine myelinolysis. When sodium depletion is accompanied by hypernatremia or normonatremia, the appropriate amount of water should be supplied along with the sodium to achieve or maintain a normal serum sodium concentration. Chronic losses from diarrhea, vomiting, or ileostomy should be treated in adults with an oral rehydration solution that provides a carbohydrate source along with adequate sodium, so that Na/solute–coupled cotransport can occur and maximize the rate of sodium repletion. These solutions are discussed in the section “Enteral Nutrition.” Sodium toxicity can result from excessive administration of NaCl. Symptoms of toxicity include vomiting, diarrhea, peripheral vascular collapse, respiratory depression, and death.

Potassium

Deficiency Hypokalemia (<3.5 mEq/L) rarely is caused by decreased intake but can result from shifts between the extra- and intracellular compartments of the body occurring while the total body potassium remains normal. This shift can be caused by alkalosis and the administration of glucose or insulin. Alternatively, decreased body stores result from increased renal excretion or intestinal losses, with subsequent shift of potassium from extra- to intracellular compartments. Other causes of renal losses include mineralocorticoid excess, some antibiotics, and rarely renal tubular acidosis. In pure deficiency states, each decrease of 1 mEq/L corresponds to a loss of 200 to 300 mEq of body potassium. The loss of 5% to 10% of body stores is usually tolerated without symptoms, but when serum levels fall below 3.0 mEq/L, weakness, paresthesias, confusion, cramps, myalgias, and cardiovascular abnormalities can occur. [93]

Assessment The normal serum level of potassium is 3.5 to 4.5 mEq/L and reflects both body stores and the availability of glucose as an energy source. Red blood cell hemolysis or extreme leukocytosis without hemolysis can result in spurious hyperkalemia. Total body potassium deficit cannot be estimated accurately. Treatment should be provided if symptoms are attributed to the hypokalemia, or if other conditions, such as chronic liver disease, are present that would exacerbate the hypokalemia. Alkalosis tends to lower the serum potassium concentration, whereas acidosis raises it. Thus, alkalosis can be treated first before the addition of potassium salts, but the same should not be done when acidosis is present.

Treatment Oral replacement should be used if possible because this allows the serum potassium concentration to rise slowly in equilibrium with the intracellular compartment. Either food (Table 53-9) or supplement (Table 53-10) sources should be used. The administration of 20 to 40 mEq at a time up to four times a day is safe. The chloride salt is preferred when renal or gastric losses account for the hypokalemia. When hypokalemia is secondary to diarrhea, one of the alkalinizing salts (e.g., citrate, acetate, bicarbonate) is preferred. The slow-release, matrix, and microencapsulated forms are better tolerated than liquid preparations. The slow-release forms have reduced the chance for small bowel ulcers and strictures but may not be useful for patients with short bowel syndrome. For intravenous therapy, the infusion rate should not exceed 20 mEq/h unless cardiac monitoring is included. The chloride salt is usually best for intravenous therapy, except when phosphate is used for treating the associated hypophosphatemia of diabetic ketoacidosis. In the presence of acidosis, potassium acetate is the best choice. Hyperkalemia should be avoided during therapy because levels above 8.0 mEq/L carry a severe risk for cardiac dysfunction, including bradycardia, asystole, and ventricular fibrillation. Skeletal muscle weakness, areflexia or hyperreflexia, paralysis, and fatigue may also occur at these high levels.



FOOD	AMOUNT
Excellent Sources (500–800 mg)	
Cantaloupe	1/2
Potato, white, baked	1 medium
Dates	1/2 cup
Raisins	1/2 cup
Figs, dried	4 medium
Avocado	1/2
Prune juice	1 cup
Very Good Sources (400–500 mg)	
Skim milk	8 oz
Banana	1 medium
Orange juice	8 oz
Grapefruit	1 cup
Nectarine	1 large
Watermelon	2 cups

TABLE 53-9 elected Foods Rich In Potassium

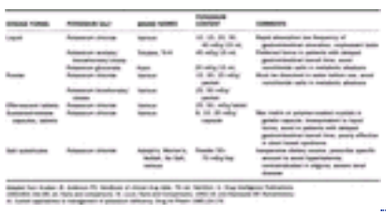


TABLE 53-10 Oral Potassium Products Comparison Chart

Calcium

Deficiency The conditions that cause hypocalcemia requiring calcium supplementation include vitamin D deficiency, failure of vitamin D synthesis or action, hypoparathyroidism, hypomagnesemia, severe acute pancreatitis, malabsorption syndromes, neoplasms with osteoblastic metastases, and medications, including aminoglycosides, cisplatin, calcitonin, furosemide, mithramycin, phosphates, and anticonvulsants. [94] Acute manifestations include tetany, altered myocardial function and arrhythmias, hyperreflexia, paresthesias, seizures, mental status changes, and choreoathetotic movements. The electrocardiogram shows prolonged QT intervals with normal QRS complexes. Chronic deficiency may not be associated with hypocalcemia. With a total body content of 100 g and a net daily loss of 100 mg/d, a 20% decrease in body stores that could be easily detected would develop in 2000 days, or about 6 years. Osteopenia usually presents with fractures or bone pain. However, patients identified as being at high risk for the development of osteopenia should begin treatment well before this occurs. Risk factors include cigarette smoking, family history of osteoporosis, sedentary lifestyle, long-term glucocorticoid therapy, long-term anticonvulsant therapy, early surgical or natural menopause (<50 years), and testicular failure (secondary to drug therapy or disease).

Assessment Recent intake or absorption is assessed by 24-hour urinary calcium. [88] At levels of absorption above 2 mg/kg (corresponding to an intake of 6 mg/kg), urinary calcium equals absorbed calcium. When absorption is low, urinary calcium levels do not decline proportionally, unlike the levels of sodium, potassium, magnesium, and phosphorus, all of which are better absorbed with better-regulated urinary excretion. Urinary calcium is used primarily to determine that absorption is low and to follow the patient after oral replacement is begun, to ensure that absorption becomes normal and that hypercalciuria does not ensue. Urinary calcium excretion of more than 300 mg/d is an indication to reduce the dose of calcium (or vitamin D). Body stores of calcium are assessed by bone densitometry, with many centers now using dual-energy x-ray absorptiometry. The accuracy of this method is equal to that of quantitative computed tomography and is sensitive to changes of 9% to 12% in bone mass. [95] Results must be compared with those of age- and gender-matched controls and are useful for diagnosis and following the response to therapy. Bone densitometry should be used to screen all patients at high risk for the development of osteopenia and to guide treatment decisions for selected postmenopausal women, but it is not recommended for routine screening. Serum calcium and alkaline phosphatase levels do not become abnormal until total body calcium depletion is quite severe.

Treatment The treatment of acute symptomatic hypocalcemia is required immediately to prevent laryngospasm, seizures, and cardiac arrhythmias. A total of 200 to 300 mg of elemental calcium should be given intravenously over 5 minutes as 1 g of calcium chloride or 2 to 3 g of calcium gluconate. This should be followed by an infusion of calcium of 0.5 to 2.0 mg/kg per hour. [94], [96] The serum calcium should normalize in 6 to 12 hours while the vital signs and electrocardiographic activity are monitored. Patients with chronic hypocalcemia or osteopenia on the basis of poor intake or absorption should be treated with oral supplements of up to 2.5 to 3.0 g/d given in divided doses. The best dietary source of calcium is milk, and this can be used if clinically tolerated. Otherwise, a number of oral calcium salts are available that vary in elemental calcium content (Table 53-11). [97] Calcium carbonate is used most often because of the high percentage of elemental calcium, but it is rather insoluble. If the urinary calcium does not increase with calcium carbonate, another, more soluble salt (e.g., calcium gluconate) may be tried, with or without the addition of vitamin D. People who take calcium supplements may be at risk for ingesting lead, even though the amount of lead in bone meal has dropped as a consequence of a decrease in the levels of environmental lead from gasoline and paint. When tested by the Natural Resources Defense Council, many of the 20

major brands of calcium supplements marketed in California contained enough lead to increase the average intake of 5 to 10 µg by 50%. [98] The supplements tested that were lowest in lead content (<0.5 µg per dose) were Children's Mylanta, Posture-D High Potency Calcium with Vitamin D, and Tums 500 Calcium Supplement.

SALT	PERCENTAGE OF ELEMENTAL CALCIUM (%)	TABLET SIZE (mg)	AMOUNT OF CALCIUM PER TABLET (mg)
Calcium carbonate	40		
Caltrate		1500	600
Os-Cal 500		1250	500
Tums		500	200
Calcium phosphate	38		
Posture		800	300
Calcium acetate	25	1000	250
Calcium citrate	21		
Citracal		950	200
Calcium lactate	13	650	85
Calcium gluconate	9	500	45
Calcium glubionate	6.5	1800/5 mL	115/5 mL

TABLE 53-11 Selected Oral Calcium Supplements

When malabsorption is present, the doses of vitamin D used are large (25,000–50,000 IU given from once a week to daily). Alternatively, the more soluble forms of vitamin D, especially 1,25-dihydroxyvitamin D, may be effective. Treatment programs must be individualized and monitored closely. Thiazides can decrease the urinary loss of calcium (thereby rendering the urinary determination less meaningful) buy also increase the urinary loss of magnesium. Thus, their use in patients with osteopenia must be carefully considered. Supplemental calcium may predispose some individuals to kidney stones but decreases the risk for oxalate stones in patients with short bowel syndrome. The development of calcium toxicity in patients with malabsorption is rare but can occur in patients with osteopenia and normal absorption, especially if they are also receiving supplemental vitamin D. Hypercalcemia can cause anorexia, nausea, constipation, polyuria, fatigue, weakness, confusion, lethargy, decreased deep tendon reflexes, shortened QT and prolonged PR interval and QRS complex duration, and cardiac arrest. [96]

Magnesium

Deficiency Symptomatic magnesium deficiency is usually the result of decreased absorption or increased losses in the urine or from the gastrointestinal tract, especially in patients with extensive small bowel resection. [99] Excessive urinary loss, usually caused by decreased tubular reabsorption, occurs in a variety of circumstances. Medications promote renal loss, including diuretics, aminoglycosides, cyclosporine, amphotericin B, cisplatin, and digitalis. [100], [101] Increased renal losses also occur in hypercalcemia, volume expansion, tubular dysfunction, acute and chronic alcoholism, diabetes mellitus, hyperparathyroidism, and hypophosphatemia. Hypomagnesemia caused by a shift of magnesium from the extracellular to the intracellular compartment may result from feeding, treatment of diabetic ketoacidosis, parathyroidectomy, acute pancreatitis, and correction of acidosis in renal failure. Hypomagnesemia is often associated with and can be a cause of hypocalcemia, hypokalemia, and hypophosphatemia. [101] Hypomagnesemia can present as tremor, myoclonic jerks, ataxia, tetany, ventricular dysrhythmias, tachycardia, hypotension, cardiac arrest, apathy, depression, delirium, hallucinations, and psychosis. [102] The signs and symptoms of hypomagnesemia are often a consequence of the hypocalcemia resulting from the same condition (e.g., malabsorption) or of the decreased parathyroid hormone secretion that occurs when hypomagnesemia is severe.

Assessment In the absence of excess fecal or renal losses, urinary excretion reflects intake and absorption of magnesium. [89] When the serum magnesium is very low in patients with short bowel syndrome, the urinary magnesium excretion may also be low and perhaps more sensitive than the serum magnesium. [103] Serum magnesium concentrations do not correlate well with total body stores, but it is reasonable to assume that depletion is present when the serum concentration is less than 1.0 mg/dL. [101] Because magnesium is primarily an intracellular ion, the best indicator of depletion may be a measurement of the content in white cells, although only a few studies have compared this value with those in other tissue pools. [89] A load test for magnesium depletion has been described, but its usefulness has not been satisfactorily documented.

Treatment Moderate to severe deficiency should be treated parenterally, especially if tetany or ventricular arrhythmias are present. An intravenous loading dose of 4 to 8 mmol should be given first, followed by 25 mmol/d thereafter until the plasma magnesium is above 0.4 mmol/L. [104] If hypocalcemia or hypokalemia is present, magnesium should be replaced first. Repletion is best achieved with slow, continuous infusions to avoid rapid increases in urinary excretion. Replacement should be monitored closely with serial serum concentrations and evaluation of deep tendon reflexes. Hypermagnesemia (>3.0 mg/dL) should be avoided because vasodilation, nausea, vomiting, drowsiness, lethargy, hypotension, bradycardia, depressed deep tendon reflexes, and a prolonged PR interval can develop. Patients with mild deficiency or ongoing magnesium losses should receive foods rich in magnesium or an oral supplement. A variety of magnesium salts are available, but many of them, especially the oxide and hydroxide, are very insoluble (Table 53-12). Because magnesium is poorly absorbed, it is best to give it in frequent small doses. Potassium-sparing diuretics such as spironolactone decrease urinary magnesium losses and may be useful, especially in patients who are also hypokalemic. However, some patients will require parenteral supplements. Outpatients unresponsive to oral therapy can be treated with intramuscular injections of 2.0 mL of 50% magnesium sulfate solution (8.1 mEq) in each buttock at intervals as needed.

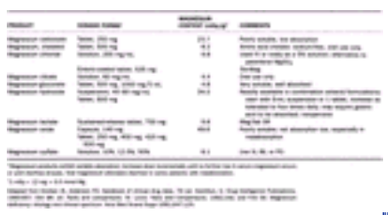


TABLE 53-12 Magnesium Products Comparison Chart

Phosphorus

Deficiency Most patients in whom clinical evidence of hypophosphatemia develops have an underlying wasting condition, [105] such as malabsorption, malnutrition of any cause, cancer, or chronic alcoholism. Causes of hypophosphatemia include increased urinary losses, shifts of serum phosphorus into the intracellular compartment, and reduced absorption. Absorption is decreased with the use of phosphate-binding antacids, vitamin D deficiency, and parathyroid hormone deficiency. Renal loss is increased in metabolic alkalosis, diuretic therapy, renal tubular dysfunction, osmotic diuresis, hyperparathyroidism, and long-term corticosteroid therapy. [104] When carbohydrate is administered or when phosphate-rich lean tissue is restored and insufficient phosphorus is available, serum levels can fall. [104] Signs and symptoms of hypophosphatemia include hemolytic anemia (serum phosphorus concentration <0.5 mg/dL), rhabdomyolysis (<1.0 mg/dL), and other complications that occur with lesser degrees of depletion, including platelet dysfunction, metabolic acidosis, peripheral neuropathy, central nervous system dysfunction, metabolic encephalopathy, and cardiac failure. Most cases of hypophosphatemia appear to be associated with intercompartmental shifts of phosphorus rather than with decreases in the total body pool, although this distinction is not easy to make.

Assessment The normal serum phosphorus concentration is 2.5 to 4.5 mg/dL. Levels lower than 1.0 mg/dL usually cause symptoms. [104], [105]

Treatment If hypophosphatemia is the result of therapy with phosphate-binding medications (antacids, sucralfate), stopping the medication is sufficient. If oral or enteral intake is possible, supplementation for other causes of a serum phosphate level below 2.5 mg/dL can be provided with milk (1000 mg or 32 mmol/quart of skim milk) or with other oral preparations in doses of 2 to 3 g/d (e.g., Neutra-Phos K or Uro-KP Neutral with 250 mg or 8.0 mmol per capsule or dose of powder). Most of the oral preparations, however, cause diarrhea (secondary to lactose intolerance in the case of milk and the phosphate content in the others), which may limit their use in patients with malabsorption. Intravenous supplementation should be limited to those who cannot take oral preparations or who have severe hypophosphatemia (<1.0 mg/dL). Initially, the dose used should be 2.5 to 5.0 mg of elemental phosphorus per kilogram administered over 6 to 8 hours in normal saline solution, not to exceed 7.5 mg/kg every 6 to 8 hours over an extended period to avoid hypocalcemia or sodium or potassium overload. The exact amount needed to restore serum phosphorus cannot be predicted because severe hypophosphatemia may occur with normal body stores of phosphorus. When hypocalcemia or renal failure is present, great caution should be used in administering phosphate by any route, but especially intravenously. Too rapid administration can cause calcium phosphate precipitation in renal tubules and other tissues.

Iron

Deficiency For clinicians, the sequelae of the last stage of iron deficiency, when stores are depleted and anemia occurs, are equated with iron deficiency. However, the role of delivering oxygen to the tissues is dependent not only on hemoglobin concentration but also on the ability of iron-containing proteins in mitochondria to

process the oxygen. Some evidence demonstrates alterations in the activity of iron-containing respiratory chain enzymes in iron-deficient animals. [106] Such changes may account for the symptoms more prevalent in children (i.e., anorexia, decreased resistance to infection, and reversible protein-losing enteropathy) and for other features not easily explained by anemia, such as angular stomatitis, atrophic lingual papillae, koilonychia, and behavioral or cognitive changes. [107] Cognitive changes have been correlated with low hemoglobin levels in dieting adults, although it is not clear whether the iron was causative. [108] When anemia develops, weakness and pallor predominate.

Assessment The hemoglobin and hematocrit are the initial and best screening tests, [109] but the results should be interpreted with caution. The patient's age and gender have to be considered in an assessment of the iron status of any individual. Moreover, hemoglobin concentrations can be altered by dehydration or overhydration, chronic inflammation or infection, protein-calorie malnutrition, cobalamin or folate deficiency, hemoglobinopathies, and pregnancy. Thus, hemoglobin is at best an indirect index of the iron status. The ferritin levels in serum are directly proportional to marrow iron stores and inversely proportional to transferrin levels, with each milligram per liter representing 10 mg of storage iron. Low levels of serum ferritin (<15 ng/mL) in the presence of anemia nearly always reflect decreased iron stores, and values up to 35 ng/mL are very suggestive. [1] However, ferritin levels can be elevated in cases of acute and chronic inflammation, cobalamin or folate deficiency, leukemia or lymphoma or other tumors, alcohol intake, and hyperthyroidism. Thus, values between 15 and 100 ng/mL can be seen in patients with iron deficiency. Serum iron levels alone are not a good measure of the iron status because they do not reflect a stable body pool. About 35 mg is turned over in the plasma each day, and levels can change rapidly in response to acute inflammation. In iron deficiency, serum transferrin levels rise while iron levels fall, so that the percentage of saturation falls below 15%. When chronic inflammation, infection, or liver disease is present, serum transferrin levels fall. Transferrin synthesis is down-regulated at the level of translation by iron, so that transferrin levels are elevated in iron deficiency. Thus, a low percentage of saturation in the presence of low transferrin levels does not reflect iron deficiency but rather the anemia of chronic inflammation. Table 53-13 outlines the predictive value of the usual tests relative to the stage of iron deficiency.

TEST	STAGE OF IRON DEFICIENCY (Predictive Value, %) I	II	III	IV
Bone marrow iron stain	100	100	100	100
Serum ferritin (μg/L)	100	100	100	100
Zinc protoporphyrin (μmol/mol of heme)	0	100	100	100
Transferrin saturation (%)	0	71	78	96
Hemoglobin (g/L)	0	0	100	100
MCV (μm^3)	0	0	22	100
MCH (pg)	0	0	33	100

Note: The prevalence rates of patients presenting in each stage of deficiency are about 24%, 23%, 15%, and 38%, respectively, for stages I through IV.

MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin.

Modified from Hastka J, Lasserre J-J, Schwarzbeck A, et al. Laboratory tests of iron status: correlation or common sense? Clin Chem 1996;42:5.

TABLE 53-13 Predictive Values of Laboratory Tests in Different Stages of Iron Deficiency

How to proceed when ferritin values are above 35 ng/mL is not clear, and transferrin receptors have been suggested as a possible aid. [110] The amount of uptake of transferrin into cells is regulated by alterations in the number of transferrin receptors on the surface of cells and is dependent on the intracellular concentration of iron, cell proliferation status, and metabolic need. The highest concentration of transferrin receptors on cells is on those with the greatest need for iron, such as reticulocytes. A truncated form of the receptor is found in human plasma bound to transferrin, and its content is elevated in iron deficiency, even if mild and of recent onset. The levels of transferrin receptor are not affected by inflammation or liver disease, and theoretically, the levels can be used to distinguish iron deficiency from the anemia of chronic disease. However, receptor levels are also increased in ß-thalassemia, hemolytic anemia, sickle cell anemia, polycythemia, myelofibrosis, and chronic lymphocytic leukemia. [111], [112] The value of the transferrin receptor in iron deficiency has yet to be corroborated by a widespread experience, although a radioimmunoassay detecting human soluble receptor is now commercially available.

Treatment Oral iron is available in a wide variety of preparations as a single nutrient or in combination with other vitamins and minerals. The amount used is not important so long as it is adequate to replenish body stores, with an assumption of 20% absorption at the start of treatment for iron deficiency. The approximate percentages of the preparations as elemental iron are as follows: sulfate anhydrous, 30%; sulfate $7{\cdot}H_2O$ hydrated, 20%; fumarate, 33%; and gluconate, 11.6%. The dose should be calculated based on the elemental iron content. A standard dosing schedule is ferrous sulfate (65 mg of elemental iron) two to three times a day between meals. Side effects include nausea, indigestion, diarrhea, and abdominal cramping and can limit the amount that can be ingested. Side effects may be less common when slow-release forms are used, but the iron may be made less available by delaying release to beyond the duodenum, the site of maximal absorption. The total iron deficit can be determined roughly by calculating the amount needed to replenish hemoglobin, with an additional gram for repleting tissue stores. Iron deficit (mg) in males = body weight (lb) × [15 - hemoglobin (g/dL)] + 1000. A hemoglobin value of 12 or 13 g/dL can be used for females. If 10% absorption throughout therapy is assumed, the total dose required can be calculated. Once the daily tolerable supplement is determined for a patient, the length of treatment necessary to replete the stores can be calculated. Oral preparations should be continued for 1 to 3 months after the hemoglobin level is restored to allow for restoration of other tissue stores. Parenteral iron is used only when oral iron is not tolerated or effective. Iron dextran (50 mg/mL) is available for intramuscular or intravenous use. The total dose required to replete stores is based on the patient's weight and hemoglobin level:

$$\text{Total Iron (mg)} = 0.3 \times \text{Body Weight (lb)} \times 100 - \frac{\text{Hemoglobin(g/dL)} \times 100}{14.8}$$

The intravenous route is safe when the iron is diluted in normal saline solution and administered over 2 to 3 hours. [113] A test dose of 25 mg should be given initially, and the daily dose should not exceed 100 mg if one wishes to minimize the risk for side effects. However, medium-dose (100–400 mg) or high-dose (500–1000 mg up to full replacement) regimens can be used safely and more conveniently, provided adequate precautions are taken and the patient is observed during the infusion. [113] Anaphylaxis is extremely rare when iron dextran is given in this way. However, the patient should be closely supervised and parenteral iron given with caution anyone with asthma or allergy. Epinephrine should be available for acute hypersensitivity reactions, and antihistaminics can be given for symptoms of urticaria, rashes, sweating, dizziness, headache, nausea, and fever. Most patients with Crohn's disease who require parenteral iron will respond, but erythropoietin can enhance the response. [114]

Zinc

Deficiency Zinc deficiency is difficult to identify and thus is probably underdiagnosed. Overt deficiency in otherwise healthy individuals is not reported in the United States, but the growth of children can be limited by inadequate dietary zinc. [115] Patients with gastrointestinal disease (Crohn's disease, short bowel syndrome), cystic fibrosis, pancreatic insufficiency, or chronic diarrhea from any cause are at increased risk for zinc deficiency because of increased losses in the stool and decreased intake. [116] Diarrheal stool contains about 17 mEq of zinc per liter, in excess of the Recommended Dietary Allowance (RDA). Other conditions predisposing to zinc deficiency include cirrhosis, sickle cell anemia, pregnancy, pica, and penicillamine treatment. [115] Clinical manifestations include growth retardation (in children), skin lesions on the face and limbs (which can vary from moist and pustular to seborrheic and acneiform), alopecia, diarrhea, apathy, night blindness, and possibly poor wound healing and loss of taste. However, the single double-blinded study of the effect of zinc supplements on taste and smell dysfunction did not support a role for zinc. [117]

Assessment None of the available methods reliably reflect either recent intake and absorption or body stores. The fasting plasma concentration is most often used, but it correlates poorly with total body zinc except when the patient is severely deficient. Most zinc in blood is in red cells, so that even small amounts of hemolysis can alter plasma zinc. Most plasma zinc is bound tightly to a $_2$-macroglobulins or loosely to albumin. Thus, hypoproteinemia can lower zinc levels. The plasma level may be falsely low because it can be decreased by many other factors, such as stress, infection, polypharmacy (displacing zinc from albumin), and corticosteroids. [1], [89] Levels below 50 μg/mL are often associated with some symptoms, and patients with levels below 30 μg/mL nearly always have some manifestation of deficiency. Metallothionein is a zinc/copper–binding protein that is reduced in zinc deficiency, and its red cell content has been low when zinc plasma levels were normal, perhaps reflecting a functional zinc pool. [118] If these results are reproducible, this test might prove useful in the early detection of zinc deficiency. At present, however, if zinc deficiency is suspected, it is best diagnosed by the symptomatic response to zinc therapy.

Treatment Zinc is available as a single nutrient and as a component of many multivitamin/mineral preparations. Zinc supplementation should be provided to persons at risk for the development of deficiency, as well as to those with symptomatic deficiency. [119] Zinc sulfate (67 or 220 mg) contains 15 or 50 mg of elemental zinc. Thus, unlike a single dose of other minerals, a single dose of zinc exceeds the RDA, and oral therapy is usually sufficient, even though absorption is only about 20%. Parenteral zinc therapy is also available as the sulfate or chloride. Zinc is safe to use; the minimum oral toxic dose is 500 mg/d. [1] Long-term ingestion of high doses (450 g/d) can induce copper deficiency. [120] Zinc supplements have reduced the incidence, prevalence, duration, and severity of diarrhea and pneumonia in children from developing nations, presumably because the availability of dietary zinc in such regions is limited. [121]

Copper

Deficiency Copper deficiency in adults is rare. It can occur in patients receiving TPN without copper supplementation and in patients treated with chelating agents, such as D-penicillamine, with or without large doses of zinc. [122] Premature infants, malnourished or malabsorbing patients, and persons with Menkes syndrome, an inherited defect in copper absorption, are at higher risk for the development of deficiency. [123] Neutropenia, anemia, diarrhea, and scurvylike bone changes can occur. [124] Copper supplementation should also be considered for patients with increased gastrointestinal fluid losses, especially from chronic biliary tract fistulae, because biliary secretion is the major excretory route for copper. [122]
Assessment The laboratory assessment of the copper status is difficult because serum concentrations do not correlate well with tissue levels. Because 80% of copper is bound to ceruloplasmin (each milligram binds 3.3 µg of copper) and the rest to other proteins, the level does not correlate with intake. [125] However, tissue copper levels are quite stable and change slowly. Thus, when deficiency occurs, the plasma level often falls earlier than the tissue levels. Normal levels are increased by estrogens or oral contraceptives, pregnancy, and acute and chronic infections. Levels are decreased in nephrosis, Wilson disease, and any cause of protein malnutrition. Free copper levels are more instructive than total levels and can be estimated by calculating bound copper [bound copper = ceruloplasmin (µg/dL) × 3] and subtracting it from the total. Values below 25 µg/dL are normal. [126] This calculation is more sensitive in detecting elevated copper levels (as in Wilson disease) than in detecting copper deficiency.
Treatment Oral supplementation can be provided by the copper sulfate in multivitamin/mineral preparations, which contain 0.4 mg of elemental copper per milligram of copper sulfate. The daily dose of 2 to 3 mg is adequate to treat deficiency in adults. The suggested maintenance intravenous dose is 0.3 to 1.5 mg/d, but the dose should be reduced to 0.15 mg/d for patients with cholestasis. [127] To treat deficiency, 2 to 7 mg/d has been given intravenously. There is a large margin of safety in using copper supplements, but acute oral ingestion of more than 15 mg of elemental copper produces nausea, vomiting, diarrhea, abdominal cramps, and mucosal ulceration. At larger doses, hemolysis, gastrointestinal bleeding, azotemia, and jaundice can occur. The treatment of toxicity includes gastric lavage and 1 g of D-penicillamine per day. [128]

Thiamin (B_1)

Deficiency Deficiency may be caused by decreased intake or increased tissue utilization, or by a combination of the factors. Thus, the usual clinical settings include pregnancy, chronic alcoholism, malabsorption syndromes, chronic nausea and vomiting, prolonged febrile illnesses, and chronic renal dialysis. The total daily requirement is usually more than 1 mg/d in adults, and the total stores are only about 30 mg, so deficiency can develop fairly rapidly. In parts of the world where polished rice is the staple cereal in the diet, beriberi (weakness, paresthesias, high-output cardiac failure) is seen. In the United States, where food thiamin is abundant, the deficiency syndrome is often seen in conjunction with deficiency of other B vitamins. Symptoms and signs include peripheral neuropathy, limb weakness, cerebellar dysfunction, subacute necrotizing encephalomyelopathy, and Wernicke encephalopathy (apathy, confusion, ataxia, photophobia, nystagmus, paralysis of upward gaze). Signs of deficiency may worsen if glucose is given without thiamin. If lactic acidosis is also present, signs of heart failure may develop. [129] Thiamin deficiency should be considered in the differential diagnosis of lactic acidosis.
Assessment Recent intake or absorption, but not body stores, is correlated with urinary excretion. The amount excreted is higher during childhood, when growth is rapid, so the result (micrograms of thiamin per gram of creatinine) must be compared with the values in age-matched controls. Body stores are measured either directly in blood or serum by high-performance liquid chromatography or by the erythrocyte transketolase assay, which measures a thiamin-dependent enzyme activity. The activity coefficient of the enzyme is determined by the activity in the presence and absence of added thiamin. In deficiency states, more stimulation occurs, and the activity coefficient is above 1.25. [89]
Treatment Thiamin HCl is available as a single nutrient in oral and parenteral forms and as a constituent of almost all multivitamin preparations. Adults with mild deficiency should receive 10 to 20 mg intramuscularly or 25 to 50 mg orally twice daily for 1 week, followed by an oral maintenance dose of 2 to 5 mg/d. Critically ill patients, especially those with central nervous system manifestations of deficiency, should receive doses of 50 to 100 mg twice daily intravenously for 3 days, followed by oral supplementation of 5 to 30 mg/d until a normal diet is resumed. The thiamin should be given before any carbohydrate is administered to avoid enhancing the deficiency. Because thiamin deficiency is often associated with deficiencies of other B vitamins, multiple vitamins should be given. Rare responses to parenteral thiamin include feelings of warmth, tingling, pruritus, nausea, sweating, and anaphylactic reaction. [130] It has been suggested that thiamin improves energy levels during exercise and in the elderly, and cognition in Alzheimer disease, but the few studies performed do not support these conclusions. [131]

Riboflavin (B_2)

Deficiency Riboflavin deficiency usually occurs along with deficiencies of other B vitamins, especially in patients with malabsorption and chronic alcoholism. Early symptoms relate to oral or eye lesions. Angular stomatitis (maceration and fissuring of the mucocutaneous junction at the angles of the mouth), cheilosis (inflammation of the lips), glossitis, geographic tongue, seborrhea-like dermatitis, pruritus, photophobia, corneal vascularization, and visual impairment may occur. The differential diagnosis of the lip lesions includes poorly fitting dentures with malocclusion, allergy (e.g., lipstick, tooth paste), and iron deficiency anemia. Other vitamin B deficiencies can cause the tongue lesions. Some of these symptoms occur in vitamin B_6 deficiency because the oxidase necessary to produce functional vitamin B_6 is riboflavin-dependent. [132]
Assessment Urinary excretion correlates well with intake because the vitamin is excreted unmetabolized. With fasting or prolonged bed rest, urinary excretion can be falsely elevated. Levels below 40 µ/g of creatinine signify very low intake in an adult. Body stores of riboflavin are assessed by measuring the riboflavin-dependent enzyme erythrocyte glutathione reductase in the blood. An activity coefficient above 1.2 (stimulated/control activity) signifies deficiency. [89]
Treatment Deficiency can be treated with 5 to 10 mg/d orally, along with other vitamins. [133] When malabsorption is present, a prophylactic dose of 3 mg/d is recommended, usually as part of a multivitamin preparation. Riboflavin may cause a yellow-orange appearance to the urine but is remarkably nontoxic.

Niacin (B_3)

Deficiency Because of the current niacin supplementation of grains and breads, the classical deficiency syndrome of pellagra (dermatitis, dementia, diarrhea) is rare in the United States. Deficiency is seen most often in patients with chronic alcoholism, malabsorption syndromes, or carcinoid syndrome, in which a large amount of tryptophan (niacin precursor) is converted instead to serotonin. Pellagra rarely occurs in the carcinoid syndrome, and when it does, it can be overcome with oral niacin. Hartnup disease is associated with a defect in tryptophan absorption and can also predispose to deficiency. [134] Isoniazid therapy can lead to deficiency because hydrazines form adducts with pyridoxal phosphate. The last enzyme in the conversion of tryptophan to nicotinic acid, kynureninase, requires pyridoxal, thereby linking the deficiency of these two vitamins. Moreover, isoniazid resembles nicotinic acid and acts as an inhibitor. In niacin deficiency, the dermatitis occurs over exposed areas and is scaly. Painful tongue and angular stomatitis are seen but may be caused in part by accompanying riboflavin deficiency. Diarrhea is probably caused by the direct effect of niacin deficiency on epithelial cell function. Central nervous system dysfunction includes irritability, headache, insomnia, psychosis, hallucinations, and seizures.
Assessment The urine content of a niacin metabolite, *N*-methylnicotinamide, reflects intake and absorption and is less than 0.5 mg/g of creatinine in adults with deficient intake. No test is available for body stores of niacin, although the results of one study in young men suggested that an erythrocyte ratio of nicotinamide adenine dinucleotide (NAD) to nicotinamide adenine dinucleotide phosphate (NADP) below 1.0 might reflect deficiency. [89]
Treatment Isolated niacin deficiency is unusual in the United States, so treatment with other B vitamins often accompanies niacin replacement. [134] Depending on their severity, the symptoms of pellagra respond to oral doses of 100 to 500 mg of niacin per day. [134] Mental, gastrointestinal, and oral symptoms clear rapidly, but the resolution of skin lesions may require weeks to months. Large doses of nicotinic acid (1–3 g/d) used to treat hypercholesterolemia can cause flushing, pruritus, burning sensations, nausea, vomiting, heartburn, diarrhea, dizziness, and tachycardia. Hyperglycemia and increased serum transaminase and bilirubin levels can occur at doses as low as 750 mg of nicotinic acid per day. [135]

Pyridoxine (B_6)

Deficiency Because the vitamin is abundant in foods and is produced by colonic bacteria, dietary deficiency is rare. It is most often reported as a result of treatment with pyridoxine antagonists, especially isoniazid, hydralazine, oral contraceptives, dopamine, and D-penicillamine. Rarely, deficiency occurs in chronic alcoholism or malabsorption, usually in association with deficiencies of other B vitamins. The clinical manifestations include peripheral neuropathy, seborrheic dermatitis around the eyes and nasolabial folds, and oral lesions (angular stomatitis, cheilosis, glossitis), similar to those seen in riboflavin and niacin deficiencies. Seizures and sideroblastic anemia may also occur. [136] Hyperhomocystinuria has been identified as an independent risk factor for vascular disease and may be caused in part by a deficiency of cystathionine synthase, the vitamin B_6–dependent enzyme that catalyzes the conversion of homocysteine to cystathionine. When serum homocysteine levels were elevated (>16.3 µmol/L), lower levels of plasma pyridoxal 5-phosphate, cobalamin, and folic acid were found, and supplementation with all three vitamins

normalized the homocysteine levels. [137] However, it is premature to recommend the use of these supplements to alter atherogenesis in this subset of patients with hyperhomocystinemia. [138]

Assessment Recent intake is reflected by the urinary excretion of metabolites. A low intake correlates with excretion of less than 500 µg of 4-pyridoxic acid per day. Body stores are measured by the erythrocyte aminotransferase index, with a determination of enzyme activity in the presence and absence of pyridoxal phosphate. Deficiency is correlated with an erythrocyte aspartate aminotransferase (E-AST) index above 2.2. Alternatively, pyridoxal phosphate can be measured in whole blood (50–120 nmol/L) or plasma (>30 nmol/L), although the levels associated with deficiency have not been precisely determined. [89]

Treatment Pyridoxal phosphate is available in both oral and parenteral forms. Suspected deficiency should be treated with 50 to 150 mg/d orally, especially when neuropathy is present. A multiple vitamin preparation containing 2 to 5 mg of pyridoxine should be added to provide the other B vitamins. Some experts suggest such a supplement for all patients on isoniazid and similar inhibitors, whereas others suggest supplements only for those patients at high risk for the development of neuropathy. When pyridoxal phosphate is ingested in large amounts (2–6 g/d), a peripheral neuropathy may develop. [139] Pyridoxine antagonizes levodopa by stimulating the decarboxylation of dopa to dopamine. Therefore, patients taking levodopa should limit their pyridoxine intake to less than 5 mg/d. [140]

Folate

Deficiency Folate deficiency is associated with poor socioeconomic and dietary conditions, especially with chronic alcoholism (usually >80 g/d) and malabsorption syndromes. Alcohol can increase urinary folate excretion. [141] Deficiency also results from competition for absorption (sulfasalazine), long-term anticonvulsant therapy (usually years), increased utilization (pregnancy, hemolytic anemia, leukemia, chronic myelofibrosis), and inhibition of dihydrofolate reductase (the enzyme that converts folate to the active coenzyme) by methotrexate, trimethoprim, and pyrimethamine. [142] If folate stores are normal initially, symptoms will appear in about 4 months. The primary manifestation of classical folate deficiency is macrocytic anemia, often accompanied by thrombocytopenia and leukopenia. Other symptoms may include glossitis, diarrhea, fatigue, and possibly (but uncommonly) neurological signs. Two other significant forms of folate deficiency occur in the absence of classical evidence of deficiency. Women consuming too little folic acid during early pregnancy are at increased risk for delivering children with neural tube defects. The Dietary Reference Intake Committee has confirmed the recommendation of the U.S. Department of Health and Human Services that all women capable of becoming pregnant ingest 400 µg of synthetic folic acid daily. [142] Hyperhomocystinemia can be caused by folate deficiency if it is not accompanied by elevated methylmalonic acid levels. High levels of homocysteine are correlated with an increased risk for myocardial infarction in about 10% of patients. [138] Although the folate levels are normal, the hyperhomocytinemia can be corrected by supplemental folate, but it is not clear that the risk for heart disease is altered. The epidemiologic data implicating homocysteine as a factor in cardiovascular risk are strong, but data from some prospective studies have been less consistent. [138], [143] Although a low folate status is a strong determinant of elevated total homocysteine levels, the folate status was not associated with an increased risk for coronary atherosclerosis. [144]

Assessment Because folate deficiency develops during decreased intake or absorption, the first laboratory abnormality to develop is a decrease in the serum folate (2–6 ng/mL), which is sensitive to changes in intake. [89] Values below 2 ng/mL are usually associated with megaloblastic anemia and decreased tissue reserves. Because about 50% of serum folate is bound to albumin, hypoalbuminemia can produce falsely low serum folate levels. Body stores of folate can be measured by red cell folate more accurately than by serum folate. Because both red and white cells contain much more folate than serum does, hemolysis or leukemoid reaction can falsely elevate the serum folate. When both serum (<2 ng/mL) and red cell folate (<140 ng/mL) are low, folate deficiency is the cause. When cobalamin deficiency is present, folate may not be well utilized in 15% to 25% of cases, so serum folate rises and red cell folate falls. Multilobed polymorphonuclear cells and macrocytic anemia develop after tissue folate levels fall. [89] Serum homocysteine levels above 30 µmol/L are consistent with folate deficiency if the methylmalonic acid levels are normal.

Treatment Oral folate supplements are available in tablets up to 1 mg, but the unreduced pteroylglutamic acid is the form used. Most of this form is excreted unchanged or after degradation, so the dose of this supplement cannot be compared with the real requirements. The parenteral route is used only if oral therapy is not possible or malabsorption is severe. A reticulocyte response is seen in 3 to 5 days, and a peak response occurs in 5 to 10 days. The 400-µg dose found in multivitamin preparations is usually adequate as maintenance therapy for patients with malabsorption or alcoholism. For patients on sulfasalazine or with hemolytic anemia, a dose of 1 mg/d is suggested. Folate is remarkably nontoxic. Very large doses (>100 times the RDA) can precipitate convulsions in patients treated with phenytoin. [145] Giving folate to a patient with cobalamin deficiency corrects the anemia but does not prevent irreversible neurological damage. The Tolerable Upper Intake Level (UL) for folate of 1000 µg/d has been challenged because all but eight cases of masking neurological progression in vitamin B_{12} deficiency occurred in patients taking more than 5 mg of folate per day. [146]

Cobalamin (B_{12})

Deficiency A dietary deficiency of cobalamin occurs only in lacto-ovo-vegetarians who do not consume any food of animal origin. The most common causes in adults are gastric lesions (especially the atrophic gastritis associated with pernicious anemia or gastrectomy), lesions or resection of the terminal ileum (especially in Crohn's disease), and bacterial overgrowth. Gastric pathology accounts for well over half the cases. An increasingly common cause of low serum cobalamin levels is AIDS. The significance of the low levels is not always clear, but some patients manifest subtle alterations in mental and cognitive abilities. [147] Deficiency also occurs rarely in patients with severe chronic pancreatitis, congenital deficiency of the carrier protein transcobalamin II (TCII), or an inability to utilize food-bound cobalamin in the absence of other gastric pathology. [148] Symptoms are insidious and develop over 2 to 3 years, sooner if malabsorption is the cause. A sore tongue, paresthesias, anorexia, loss of taste, diarrhea, dyspepsia, hair loss, impotence, irritability, and psychiatric illness (e.g., depression) can be present. Numbness and tingling, especially in the lower extremities, can progress to loss of vibratory sensation, loss of coordination, muscle weakness, and atrophy, and memory disturbances can develop. [149] Macrocytic anemia with megaloblastic bone marrow and multilobed nuclei in the polymorphonuclear leukocytes are frequently, but not always, present. The macrocytic anemia must be differentiated from chronic liver disease and from hypothyroidism. Many patients, especially those who are elderly, now present with neurological symptoms and signs in the absence of anemia assessment The diagnosis of cobalamin deficiency is usually suspected on the basis of hematologic findings. [150] The serum concentration of cobalamin usually correlates with body stores. Unlike the folate concentration, the cobalamin concentration in red cells is not higher than that in serum, so hemolysis is not a cause of falsely high values. In humans, only a small proportion of cobalamin is carried on TCII, the protein that delivers the vitamin to the tissues. Thus, there can be a stage at which the serum level is normal (cobalamin is bound to haptocorrin) but tissue levels are low. Falsely high levels can be seen in leukocytosis (release of haptocorrin, increasing the total serum binding capacity) and in acute liver disease (release of body stores). Conversely, serum levels can be low when body stores are normal. Protein deficiency can lower serum levels without affecting tissue delivery if the TCII levels are normal. Levels are low in pregnant women because of dilution and redistribution of binding proteins. Serum cobalamin levels below 150 pg/mL (110 pmol/mL) are always associated with deficiency if causes of falsely low levels are not present (folate or protein deficiency, pregnancy). Levels between 150 to 200 pg/mL should be considered suspect and should lead to further testing with methylmalonic acid and homocysteine levels, correlation with abnormal hematologic or neurological findings, or both. A subtle presentation of cobalamin deficiency is especially likely in elderly patients. [151] In doubtful cases, a therapeutic trial with cobalamin is safe, and reversal of the abnormal findings is diagnostic. Biochemical tests for cobalamin deficiency can clarify cases in which the serum cobalamin levels combined with the clinical picture are not definitive. The workup should also include a careful neurological examination. The serum can be assayed for metabolites that increase when the function of the two cobalamin-dependent enzymes is impaired: methylmalonyl-CoA mutase (methylmalonic acid increased) and methionine synthase (homocysteine increased). Folate deficiency leads to an increase in homocysteine levels (>20 µmol/L) but not in methylmalonic acid levels, whereas cobalamin deficiency increases methylmalonic acid (>390 nmol/L) as well. [152] Methylmalonic acid levels can be elevated in the absence of cobalamin deficiency when renal insufficiency is present. Because TCII is the protein that delivers cobalamin to tissues but accounts for less than 10% of serum cobalamin binding, low holo-TCII concentrations have been reported as the earliest sign of negative cobalamin balance. [89] This test is not used routinely. The selection of the various tests for cobalamin deficiency depends on the stage of deficiency suspected (Table 53-14).

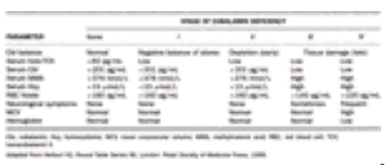


TABLE 53-14 Laboratory Tests in Sequential Stages of Cobalamin Deficiency

Because abnormal serum measurements of cobalamin metabolism precede the late manifestations of tissue damage, it is recommended that populations at risk be screened. These include strict vegetarians, persons older than 65 years of age with decreased food intake, and patients with any of the following: unexplained neurological/psychiatric symptoms or anemia, long-term use of proton pump inhibitors, autoimmune diseases, AIDS, previous gastric surgery, Crohn's disease of the ileum, and malabsorption from any cause. Screening should be done with serum holo-TCII measurements if available, either alone or in conjunction with serum cobalamin. If serum cobalamin alone is used and values below 350 pg/mL are obtained, metabolite assays should be performed if cobalamin deficiency is suspected clinically. The combination of a serum cobalamin value below 350 pmol/mL and an elevated methylmalonic acid concentration has a specificity of 98% for cobalamin

deficiency. [153] Intake or absorption should be assessed only when clinically indicated. If gastric atrophy is demonstrated by biopsy, or if gastrectomy is known from the history or the roentgenographic findings, it is not usually necessary to confirm the absence of intrinsic factor by a Schilling test. Urinary excretion of less than 5% labeled cobalamin is diagnostic of malabsorption and of more than 10% labeled cobalamin is normal, but many results are indeterminate (5%–10%). [1], [89] This lack of sensitivity is also a factor in deciding whether to proceed with the test. The two major causes of a falsely normal Schilling test result are an erroneous value (intertest variability can be as high as 30%–50%) and malabsorption of food cobalamin, presumably because of an inability of gastric proteases to liberate the vitamin. The Schilling test must be performed with labeled food cobalamin (usually in liver or scrambled eggs) to make the diagnosis because the standard Schilling test uses free cobalamin. However, the commercially available versions of the test have not been uniformly standardized.
Treatment If cobalamin deficiency is caused simply by decreased intake without malabsorption, 3 to 6 µg of oral cobalamin per day will suffice. However, if malabsorption is present, either because of lack of intrinsic factor or of intestinal disease, then losses of up to 10 µg/d must be allowed for. Deficiency is treated with 100 to 1000 µg/d for 5 to 10 days, followed by at least 300 µg/mo given by subcutaneous injection indefinitely. Alternatively, oral cobalamin can be given (1000 µg/d) to take advantage of the inefficient (~1%) absorption of the vitamin in the absence of intrinsic factor. [154] Increased well-being is noted within 24 hours after the initiation of treatment, painful glossitis improves in 48 hours, and reticulocytosis begins in 5 to 7 days. Serum folate falls rapidly. The reversal of neurological findings may take 6 months or more. No significant toxicity has been reported with therapeutic doses of oral or parenteral cobalamin.

Ascorbic Acid (C)

Deficiency Scurvy develops in 2 to 3 months if the diet is deficient in ascorbic acid. It occurs in the United States only rarely, usually in cases of chronic alcoholism, malabsorption, or food faddism. Early symptoms are weakness, lassitude, irritability, aching joints and muscles, and weight loss. Later, perifollicular hyperkeratotic papules appear on the buttocks, thighs, and legs, followed by petechiae on the lower legs. [155] In advanced deficiency, the gums become swollen, red, and spongy and hemorrhaging occurs, especially from the gums and in the skin and muscles. Anemia is common. No deficiency syndrome other than scurvy has been reported.
Assessment Plasma ascorbate levels reflect recent intake. Thus, a low serum concentration precedes clinical scurvy. [89] Low levels do not necessarily denote scurvy, however. Levels can be falsely low in patients with chronic inflammatory diseases, in cigarette smokers, after severe emotional stress, and in women taking oral contraceptives. [89] Levels below 0.2 mg/dL (11 µmol/L) denote deficient intake or absorption; that level is reached in 3 to 5 months. Leukocyte ascorbate concentration is better correlated with body stores (<150 µmol/L represents a high risk for deficiency), but the assay is technically difficult and requires large blood samples. Because the diagnosis of scurvy should be made quickly, plasma ascorbate is the initial test of choice.
Treatment Scurvy may respond to as little as 10 mg of vitamin C per day, but doses between 60 to 100 mg/d and 250 mg four times daily are recommended to replenish body stores. Large doses of ascorbic acid (2–6 g/d) have been reported to cause diarrhea, promote the formation of renal oxalate stones, increase the excretion of basic drugs by acidifying the urine (e.g., tricyclic antidepressants), decrease the excretion of acidic drugs (e.g., aspirin), and interfere with many laboratory tests, including the fecal tests for occult blood that depend on oxidation of the substrate for a positive reaction. However, the reports of clinical adverse effects with supplemental vitamin C have largely not been substantiated in normal healthy populations. [156] On the other hand, a metaanalysis of the six largest supplementation trials for rhinovirus infection found no evidence of benefit. [157] Epidemiologic studies have noted an inverse relationship between serum levels and coronary artery disease, hypertension, cataracts, and carcinomas, but no prospective studies have been performed. [1] It is premature to recommend supplemental vitamin C for these conditions.

Biotin

A dietary deficiency of biotin is very rare, [142], [158] but it can be caused by excessive consumption of raw egg white, which contains avidin, a biotin-binding glycoprotein. The effects of deficiency include anorexia, nausea, dermatitis, alopecia, mental depression, and organic aciduria. Symptoms disappear with doses of 0.15 to 0.3 mg parenterally, or with 0.2 to 10 mg/d orally for a few days. No adverse effects of such treatment have been reported.

Pantothenic Acid

Definite clinical deficiency has not been reported in humans, probably because the vitamin is so abundant in foods. [142] The “burning feet” syndrome seen in malnourished patients may respond to pantothenic acid, but it is unclear whether this represents a specific deficiency. If deficiency is suspected, 10 mg of calcium pantothenate can be used.

Vitamin A

Deficiency Vitamin A deficiency is usually the result of decreased intake or of fat malabsorption. Contributing factors include impaired conversion of carotenoids to vitamin A (in mucosal disease), decreased storage capacity (in liver disease), decreased levels of serum transport proteins (in liver disease or protein malnutrition), and increased urinary losses (in cancer, tuberculosis, or urinary tract infections). [159] Inadequate intake is rare in the United States, but the number of new cases of corneal disease caused by vitamin A deficiency worldwide approaches 1 million each year. [160] It takes about 2 years to deplete hepatic stores. Symptoms of deficiency are night blindness, xerophthalmia, follicular hyperkeratosis, altered taste and smell, increased cerebrospinal fluid pressure, and increased infections. [161] When zinc deficiency is also present, the effect on visual adaptation may be magnified.
Assessment The intake of both carotene and vitamin A is reflected in the serum levels. Decreased intake is reflected in serum vitamin A levels between 10 and 20 µg/dL. [89] A level below 10 µg/dL indicates deficiency. Low carotene levels are meaningful only if carotene is being ingested in the diet because there are no body stores of carotene. In severe liver disease and chronic infection, serum vitamin A levels may fall because retinol-binding protein is not produced in normal amounts. However, carotene levels tend to rise because less carotene is converted to vitamin A in these conditions. Vitamin A can be elevated with normal carotene levels in patients ingesting excess amounts of vitamin A or in patients on chronic hemodialysis because of a decreased conversion of retinol to retinoic acid. Elevated carotene levels with normal retinol are seen in persons ingesting excess amounts of carotene and also in patients with anorexia nervosa, hypothyroidism, hyperlipidemia, or the hypercholesterolemia of diabetes. [89]
Treatment Vitamin A is provided in the form of free retinol. Deficiency states respond to oral doses of the daily vitamin between 5000 and 50,000 IU. Intramuscular delivery can also be used. [1] Because of their antioxidant properties, carotenoids have been implicated in many chronic diseases often linked with vitamins E and C, the other vitamin antioxidants. [155] However, no convincing evidence is yet available to support supplementation of any antioxidant vitamins, carotenoids, or preformed vitamin A to prevent malignancies or cataracts. Doses of vitamin A above the RDA are contraindicated in pregnant women because of the potential for teratogenicity. The minimum daily dose that is toxic to patients without malabsorption is 25,000 to 50,000 IU. Toxicity is correlated with serum levels of vitamin A above 1000 µg/dL. Symptoms of chronic toxicity include dry mouth and mucous membranes, skeletal pain, increased cerebrospinal fluid pressure, alopecia, anorexia, irritability, hepatic dysfunction, exophthalmos, and hypercalcemia. Carotene cannot be converted to retinol fast enough for even large doses to produce retinol toxicity.

Vitamin D

Deficiency Vitamin D deficiency secondary to inadequate intake is rare in the United States, in part because milk products are fortified with the vitamin. Even persons who do not consume dairy products usually obtain adequate sunlight exposure to prevent vitamin D deficiency. However, in elderly persons with little exposure to sunlight, decreased vitamin D intake or synthesis along with inadequate calcium intake may contribute to a high incidence of bone fractures. [162] Deficiency should be considered in any patient with steatorrhea (malabsorption of dietary vitamin and polar metabolites via the enterohepatic circulation) or severe liver (decreased 25-hydroxylase activity) or kidney (lack of 1a-hydroxylase) disease, as well as in patients with Crohn’s disease or previous ileal resection. [163] Vitamin D deficiency causes hypocalcemia and hypophosphatemia, which result in increased parathyroid hormone secretion and bone demineralization. In time, this sequence leads to osteomalacia in adults and rickets in children and can be an unrecognized factor in acute hip fracture. [164] Tetany results from severe hypocalcemia, and muscle weakness is correlated with hypophosphatemia and depletion of muscle phosphate. Patients with risk factors for the development of osteopenia (cigarette smoking, family history, sedentary lifestyle, long-term glucocorticoid therapy, long-term anticonvulsant therapy, early menopause, testicular failure) should be followed even more carefully if they are at risk for vitamin D deficiency. Long-term alcohol abuse is also an overlooked cause of osteoporosis in men. [165]
Assessment The 25-hydroxyvitamin D level is low when body stores, intake, or endogenous production of the vitamin is low. [166] Thus, the level is satisfactory for assessing vitamin D status with respect to deficiency and toxicity. The production of 25-hydroxyvitamin D is not closely regulated and rises or falls as its substrate is made available. The concentration in serum is 5 to 10 times that in other tissues except for adipose tissue. It is bound in serum to a binding protein that is normally only 5% saturated, and the half-life is long (24 hours). Thus, recent exposure to sunlight or increased oral intake is reflected in the serum level. Levels are low in dietary deficiency, decreased absorption, lack of sunlight, prematurity, and severe liver disease; low levels are also associated with drugs that alter the metabolism (e.g., anticonvulsants). High levels are seen in growing children, conditions in which parathyroid hormone levels are elevated, sarcoidosis, and some forms of idiopathic hypercalciuria. Patients at risk for deficiency should be followed with periodic measurements of the serum 25-hydroxyvitamin D level, as well as bone density and 24-hour urinary calcium measurements, to assess the status of calcium stores and intake (see section “ Calcium”). 1,25-Dihydroxyvitamin D can also be

measured in the serum. The production of this isoform is closely regulated, but by extracellular ionized calcium, not by vitamin D stores, unless they are very low. The level of this vitamin correlates with vitamin D function more than with stores. Serum 1,25-dihydroxyvitamin D levels are low in profound vitamin D deficiency, chronic renal disease, hypoparathyroidism, vitamin D–resistant rickets type I, and osteolytic conditions not caused by increased parathyroid hormone levels (cancer, hyperthyroidism). Primary hyperparathyroidism, vitamin D–resistant rickets type II, and pregnancy are conditions in which levels are elevated. Hypervitaminosis D elevates 25-hydroxyvitamin D levels markedly, but 1,25-dihydroxyvitamin D levels are little affected. Table 53-15 provides suggested guidelines for evaluating the vitamin D status.

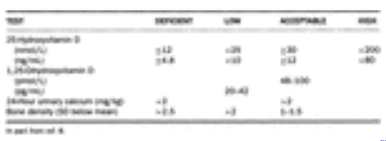


TABLE 53-15 Suggested Guidelines for Evaluating Vitamin D Status

Treatment Oral vitamin D does not maintain the vitamin D status as well as endogenous vitamin D; the latter is released more constantly from the skin and is not converted so rapidly to other isomers by the liver, to which virtually all orally administered vitamin D is exposed following absorption. Nonetheless, oral supplements are needed sometimes, especially in patients with fat malabsorption, uremia, long-term corticosteroid use, and possibly postmenopausal osteoporosis, although its proper place in the management of osteoporosis is still uncertain. Adults with vitamin D deficiency should receive 0.1 to 0.2 mg (4000– 8000 IU) of cholecalciferol daily by mouth. Much larger doses should be used if malabsorption is present, although the exact dose needed may vary widely. Theoretically, the more polar forms, 25-hydroxyvitamin D (calciferol) and 1,25-dihydroxyvitamin D (calcitriol), should be useful in patients with malabsorption, [167], [168] but the absorptive advantage is probably not sufficient to justify the great increase in cost. The efficacy of treatment should be assessed by periodic measurement of the 25-hydroxyvitamin D level, with the dose adjusted to keep the serum level within normal range. In patients with malabsorption, one should determine that the function of vitamin D has also been restored—that is, that calcium absorption has been normalized—by bringing the 24-hour urinary calcium into the normal range. The risk for toxicity in patients with malabsorption is extremely slight. In other patients, hypercalcemia and hypercalciuria may develop. The estimated minimal toxic daily oral dose for healthy adults is 0.125 mg (5000 IU). [1], [162] Nausea, anorexia, itching, polyuria, abdominal pain, constipation, bone pain, metallic taste, and dehydration may be present early, followed by other manifestations of hypercalcemia. Vitamin D toxicity is reversible if renal damage is not severe.

Vitamin E

Deficiency Deficiency of vitamin E is uncommon in humans. Persons at risk include newborns and premature infants, patients with fat malabsorption or biliary obstruction, and food faddists. Vitamin E does not cross the placenta well and is poorly absorbed in newborns. Severe fat malabsorption in cystic fibrosis, abetalipoproteinemia, short bowel syndrome, biliary obstruction, and excessive mineral oil ingestion can lead to deficiency. [169] Unsteady gait, tremor, weakness, ophthalmoplegia, pigmentary retinopathy, and proprioceptive impairment have been noted in adult malabsorptive syndromes, along with red cell hemolysis. A similar progressive neurological syndrome has been reported in children with cholestatic liver disease. [170]
Assessment Deficiency can be documented by a low serum a-tocopherol concentration (<11.6 µmol/L or 5.0 µg/mL). [89] Because the vitamin is carried in serum by lipoproteins, the plasma concentration of vitamin E is low in any hypolipidemic state. Therefore, the ratio of a-tocopherol to total lipid (triglyceride + cholesterol) is a more accurate indicator of vitamin E status than is the vitamin level alone. [171] Ratios of more than 0.8 mg of a-tocopherol per gram of total lipid or more than 0.22 mg of a-tocopherol per gram of cholesterol are normal. Ethane and pentane are generated through the peroxidation of n-3 and n-6 fatty acids, respectively. Breath ethane has been used to evaluate the vitamin E status and follow therapy in children because the level rises with deficiency. [172]
Treatment Large oral doses of D-a-tocopherol acetate, up to 600 mg (800–900 IU) per day, are needed, especially in patients with malabsorption. A water-soluble form of the vitamin (D-a-tocopherol-PEG-1000 succinate) is available over the counter as Liqui-E. [173] This compound forms micelles when given in doses above 25 mg/kg per day and may assist in the absorption of other fat-soluble vitamins. The results of the use of vitamin E to treat chronic diseases have been mixed, promising for preventing heart disease, but not for cancer. [1], [155] The American Heart Association consensus statement does not recommend the routine use of vitamin E at this time. [174] No consistent ill effects are seen with up to 3200 IU (2112 mg) per day in healthy persons. However, at doses from 100 to 1100 mg/d, oxidation of vitamin K to its active form can be inhibited, and high-dose vitamin E may be problematic in patients with bleeding disorders. However, the use of 800 to 1200 IU/d had no effect on the prothrombin time of patients on warfarin therapy. [131]

Vitamin K

Deficiency Nutritional deficiency is uncommon in adults because of the large amount in foods and because bacteria synthesize the vitamin in the intestinal lumen. Deficiency is most commonly the result of fat malabsorption, often compounded by poor intake and diminished liver function or decreased bile excretion. [175] Inhibition of bacterial biosynthesis by broad-spectrum antibiotics can be important. Deficiency is manifested by easy bruising and clotting abnormalities.
Assessment The most commonly used clinically applicable test for vitamin K deficiency is the one-stage prothrombin time. The clotting factors tested (II, V, VII, and IX) are all responsive to vitamin K except for factor V. Factor VII has the shortest half-life and is the usual rate-limiting factor. The prothrombin time does not test vitamin K stores and is abnormal when deficiency is present, when the synthesis of clotting factors is impaired by liver disease, or when factors are consumed in intravascular coagulation. Thus, the test is nonspecific. Response of the prothrombin time to 5 to 10 mg of parenteral vitamin K for 2 to 3 days confirms the presence of a deficiency state. Determination of the urinary a-carboxyglutamic acid (Gla) level may be even more sensitive because it tests a function of vitamin K, and the level can be low when blood coagulation is normal. [176] Plasma levels of Gla proteins may be useful in detecting deficiency of vitamin K but have not been widely used. These proteins include undercarboxylated prothrombin (PIVKA-II) and carboxylated/undercarboxylated osteocalcin. [8]
Treatment Long-term therapy with doses titrated to the prothrombin time (~2 mg daily or every other day) should be restricted to patients with malabsorption. The vitamin is available for subcutaneous or intramuscular injection at 2 or 10 mg/mL. Vitamin K is not a component of any multivitamin preparation and must be prescribed individually.

NUTRITIONAL SUPPORT IN THE HOSPITALIZED PATIENT WITH GASTROINTESTINAL DISEASE

The major purpose of nutritional therapy is to prevent or correct specific nutrient deficiencies and to prevent the adverse effects of protein-energy malnutrition. Nutritional support has also been proposed as a primary therapy for patients with certain gastrointestinal diseases, such as inflammatory bowel disease and hepatic encephalopathy. In this section, we review the available data evaluating the clinical efficacy of nutritional support in patients with gastrointestinal diseases. Whenever possible, only the data from prospective randomized clinical trials are considered because this approach is the most reliable technique for evaluating the usefulness of therapy.

Short Bowel Syndrome

Patients who have undergone massive intestinal resection often require nutritional support for optimal function and survival. The length of the remaining jejunum, the presence of the ileocecal valve, and whether the colon is intact are critical determinants of the need for nutritional support. [177] In general, patients with a jejunostomy and less than 90 cm of jejunum, or those with a colon in continuity but less than 50 cm of jejunum, are likely to require parenteral nutrition. Providing nutrients with continuous nighttime tube feedings [178] or fluid and electrolytes with oral rehydration therapy [33] (see section “ Oral Rehydration Therapy”) can sometimes decrease or eliminate the need for parenteral therapy.

Inflammatory Bowel Disease

Enteral Nutrition Three metaanalyses of published prospective randomized clinical trials concluded that enteral nutrition is not as effective as corticosteroids. In addition, no benefit of elemental over non-elemental formulas was noted. [179], [180] and [181] The true clinical efficacy of enteral nutritional therapy has not been evaluated because no study has compared patients receiving dietary therapy with an untreated control group. However, the overall remission rate reported with enteral nutrition therapy is about 60%, [179], [180] which is higher than the range of 20% to 40% reported for placebo treated patients with moderate disease. [182], [183]
Parenteral Nutrition Data from several prospective randomized clinical trials found that “bowel rest” is not necessary to achieve clinical remission in patients with active Crohn’s disease; patients who received TPN plus bowel rest fared no better than those receiving TPN plus oral/enteral feeding. [184], [185] A beneficial effect of TPN in patients with Crohn’s disease or ulcerative colitis has not been demonstrated in prospective randomized clinical trials. No significant differences in clinical response rates were found in hospitalized patients with severe disease exacerbations who were randomized to receive TPN and steroid therapy or an oral/enteral diet

and steroid therapy. [186], [187] and [188] Nonetheless, TPN may be useful in patients with severe inflammatory bowel disease who cannot eat because of prolonged ileus or who might benefit from bowel rest because of severe diarrhea or anticipated surgery.

Specific Nutrient Therapy Fish oil supplementation may produce a modest decrease in the activity of ulcerative colitis, [189], [190] may help to lower the steroid dose, and may prevent early but not late relapse, [191] but it is not effective by itself. Two prospective randomized clinical trials of maintaining clinical remission in patients with Crohn's disease have shown both success [192] and failure. [193] Short-chain fatty acids have not been shown to be effective for rectal disease in ulcerative colitis. [194]

Gastrointestinal Fistulae No prospective randomized clinical trials have evaluated the efficacy of TPN, but "bowel rest" may improve the clinical outcome of selected patients with fistulae. Before the use of TPN, mortality in patients with gastrointestinal fistulae was caused by malnutrition, fluid losses, and peritonitis. A retrospective analysis of patients with small bowel fistulae showed an improved outcome for those who received nutritional support. [195] Anastomotic fistulae at sites of recent resection in Crohn's disease may close permanently with TPN and bowel rest, although octreotide therapy may also be necessary for success in some patients. [196] However, fistula closure is not well maintained after an oral diet is reinstituted in patients with active disease at the origin of the fistula, or with post-fistula obstruction.

Growth Failure in Children TPN or oral/enteral supplemental feedings can initiate catch-up growth, resulting in marked increases in height and weight, in children with inflammatory bowel disease and growth failure. [197], [198], [199] and [200] In fact, even intermittent enteral feeding can improve growth. [201], [202]

Acute Pancreatitis

Most patients with acute pancreatitis are unable to eat because oral feeding can cause abdominal pain and increase serum amylase and lipase concentrations. Nonetheless, nutritional support is not useful in patients with mild or moderate pancreatitis because of the rapid resolution of symptoms and ability to begin oral feeding within several days. In addition, no benefit of parenteral nutrition compared with enteral nutrition has been demonstrated in patients with mild or moderate pancreatitis, [203] and in one study, the use of TPN was associated with a higher prevalence of catheter-related sepsis and insulin requirements. [204] However, nutritional support may be necessary in about 10% to 20% of patients with pancreatitis who have severe and complicated disease when a prolonged period of starvation is anticipated.

It has been shown that it is possible to provide enteral feeding to patients who have mild to moderate [203] or severe [205], [206] pancreatitis, without causing an exacerbation of symptoms, when feeding is delivered distal to the ligament of Treitz into the upper jejunum. Moreover, in prospective randomized clinical trials that compared enteral with parenteral nutritional therapy in patients with severe pancreatitis, those who were randomized to receive enteral nutrition showed a greater reduction in acute phase response proteins, a greater improvement in disease severity scores, and fewer medical complications than those given parenteral nutrition. [205], [206] However, it is not known whether enteral feeding was beneficial or whether parenteral feeding was harmful because no study included an unfed control group.

Providing a portion of total calories as lipid can help prevent hyperglycemia in glucose-intolerant patients with pancreatitis. However, it is important to ensure adequate triglyceride clearance because hypertriglyceridemia induced by infusion of fat emulsions can cause a relapse of pancreatitis. [63]

Liver Disease

Alcoholic Hepatitis One prospective randomized clinical trial compared PPN with an anabolic steroid, oxandrolone, or the combination for 21 days and found no change in mortality but improved Child-Pugh scores in those receiving PPN plus oxandrolone. [207], [208] In another study, nutritional therapy improved liver function and reduced early mortality (1 and 6 months) in a subgroup with moderate "malnutrition" or liver disease. [209] No improvement was noted in the group with severe disease. It is clear that these markers correlate with the severity of liver disease and with clinical outcome. [210]

Alcoholic Cirrhosis Several prospective randomized clinical trials showed that enteral nutrition in patients hospitalized for complications of cirrhosis led to an improved clinical status. [211], [212] and [213] A single study showed that enteral feeding of a branched-chain amino acid formula reduced mortality. [214] However, the ad lib diet group ingested only about half the calories of the enterally fed group, and the control group had an unusually high mortality rate (47%).

Hepatic Encephalopathy In a metaanalysis of nine prospective randomized clinical trials evaluating the use of TPN enriched with branched-chain amino acids in patients with acute encephalopathy, [53] recovery from encephalopathy was improved during short-term (7–14 days) treatment. However, the variability in mortality rates was so great that no conclusion could be reached regarding that outcome measure. No benefit of branched-chain amino acid therapy was detected in the one study that compared a group that received branched-chain amino acid–enriched TPN with a control group that received standard amino acids. Studies of the use of enteral solutions containing branched-chain amino acids in patients with chronic encephalopathy have yielded conflicting results. However, the studies with the largest number of patients showed clinical benefits in protein-intolerant patients. [215]

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CHAPTER 54

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GENETIC COUNSELING FOR GASTROINTESTINAL PATIENTS

P>Medical science, like medical economics, is in the midst of a major revolution. A large part of that revolution derives from the science of genetics. In past years, clinical genetics belonged to the practice of pediatrics and concerned primarily rare inherited diseases. Genetics was of minor practical importance to physicians in adult medicine. That era can now be considered historical, and gastroenterology, as much or more than any clinical specialty, is evidence of this.

The etiology of many gastrointestinal (GI) diseases, both inherited and acquired, both malignant and benign, is now understood at the DNA level. Additionally, many diseases that were once considered single entities are now known to be of heterogeneous etiology. Causes often include inherited abnormalities, environmental factors, and complex interactions between the two. Relevant examples of GI disease include colon cancer, inflammatory bowel disease, many liver conditions, and even peptic ulcer disease.

This new science of genetics has quickly translated into medical applications, including new methods of disease screening, susceptibility testing, diagnostics, and therapeutics. The clinical application of genetic knowledge is expected to expand dramatically with the recent sequencing of the entire human genome. All this change requires that the physician have a thorough understanding of modern genetics. Only in this way can the application of the many genetic advances be realized in the clinic.

An important part of the clinical application of genetics is patient education by the health care team. Many disease discussions now must involve genetics. Patient education is particularly important when genetic testing is being considered. Both patient and physician must understand the impact of genetic testing on medical management, future health, insurance, employment, and psychosocial issues. Each of these issues must be addressed in making the decision to proceed with genetic testing. For many diseases, the incorporation of formal genetic counseling into the evaluation and treatment process is becoming a practical necessity.

This chapter reviews the basic and clinical genetics needed by the practicing gastroenterologist, both to understand emerging relevant genetic discoveries and to apply them in the clinical setting. The principles of genetic counseling are then presented, followed by issues specific to genetic testing. Such knowledge is also imperative to enable the practitioner to educate the patient toward informed clinical decisions and to decide when formal genetic counseling is needed. Many of the examples used relate to genetic counseling and testing for inherited colorectal polyp and cancer conditions because much knowledge and experience have been accumulated in this area. However, with an increased understanding of other pediatric and adult conditions, such as celiac disease, irritable bowel disease, and pancreatitis, genetic counseling and testing for these conditions may soon become mainstream.

GENETIC KNOWLEDGE FOR CLINICAL PRACTICE

Basic Concepts of Molecular Genetics

Structure of DNA and RNA The most basic components of genetic material are the nucleotides that make up deoxyribonucleic acid (DNA) and ribonucleic acid (RNA), which in turn are the building blocks of the genome. Each nucleotide has a backbone composed of a 5-carbon sugar (deoxyribose for DNA and ribose for RNA), a phosphate, and one of several different nitrogen-containing rings known as *bases*. The bases are either pyrimidine or purine compounds. Pyrimidines include cytosine (C), thymidine (T), and uracil (U), and purines include adenine (A) and guanine (G). Nucleotides are joined together by a phosphodiester link between the 5' and 3' carbon atoms of the sugars to make a strand of DNA or RNA. Strands are described and schematically viewed in the 5' to 3' orientation because enzymes almost always work in that direction. DNA exists as a double-stranded helix in the nucleus of cells. Each DNA strand is oriented in complementary, but opposing, 5' to 3' directions. The strands are held together by noncovalent and very specific interactions; adenine pairs only with thymidine of the opposing strand and cytosine with guanine. The opposing or complementary pair of DNA nucleotide bases is referred to as a *base pair*. RNA, on the other hand, is single-stranded and is found in the nucleus and cytosol of the cell.
Replication, Transcription, and Translation DNA is replicated for cell division, called *mitosis,* or for gamete (egg and sperm) formation, called *meiosis*. Replication proceeds by way of a protein complex referred to as *DNA polymerase*. Errors in DNA replication are monitored and repaired by mismatch repair (MMR) protein complexes, a function that maintains the integrity of the DNA. RNA synthesis is the first step in protein production. RNA is synthesized from a single strand of DNA, which serves as the template. Synthesis is accomplished by a complex of proteins called *RNA polymerase*. The process of making RNA from DNA takes place in the nucleus and is called *transcription*. In RNA, the pairing of bases is slightly different in that uracil pairs with adenine and cytosine with guanine. Although the primary function of RNA is to encode proteins, RNA itself is also a part of ribosomes, transfer RNAs, and small ribonucleoprotein particles. The primary RNA transcript contains three domains: (1) regulatory elements, which are involved in controlling the protein synthetic process; (2) exons, which contain coding sequences, or those parts of the RNA that are actually translated into amino acids and eventually protein; and (3) introns, which are interspersed between the coding sequences but are themselves noncoding and thus not translated into amino acids (Fig. 54-1).

FIGURE 54-1. Organization of genetic material. DNA is the double-stranded molecule that contains the genetic code and includes coding (exons) and noncoding (introns) segments. In a multiple-step process, DNA is transcribed into RNA and spliced into messenger RNA (mRNA), which contains regulatory elements and exons but no introns. The mRNA is translated into a series of amino acids that combine to form a specific protein. Proteins are the molecules that function within cells.

Before the RNA leaves the nucleus, enzymes remove all the introns, and the resulting messenger RNA (mRNA) molecule contains only the regulatory elements and a

continuous stretch of coding sequence to be translated into protein. In the cytosol, the mRNA strand is read in sets of three nucleotides, called *codons*, each of which is translated into a specific amino acid. As reading proceeds, these amino acids are linked together as a polypeptide chain by the ribosome complex. In a manner analogous to the 5' to 3' directionality of nucleic acids, amino acids are added to the carboxyl-terminal end of a growing polypeptide chain, resulting in an “amino” to “carboxyl” orientation. Each of the codons is translated into one of 20 amino acids or a stop codon. The stop codon tells the ribosome that it is at the end of the protein. The genetic code is very precise but is also “degenerate,” meaning that multiple codons can encode a single amino acid. For example, the genetic code for the amino acid valine can be GUU, GUC, GUA, or GUG.

Types of Mutations Many types of genetic mutations can result in a defective protein and thereby cause a genetic or inherited disorder. Many genetic changes, however, do not result in genetic disorders but rather are part of normal genetic variation. Such changes are called *polymorphisms* and are typically seen in at least 1% of the population. Polymorphisms can be completely inconsequential or can have a phenotype with subtle consequences (e.g., differences in drug metabolism or minor variations in disease susceptibility). When new genetic changes are identified in symptomatic or presymptomatic individuals, it is important to clarify whether the variant is a deleterious (disease-causing) mutation or a polymorphism. Understanding the basis of the genetic changes is important because it significantly influences the type of molecular diagnostic testing that should be considered.

Amino acid changes. The first major class of mutations consists of amino acid changes in the coding sequencing. Nucleotide errors can result in three categories of amino acid changes: silent, missense, and nonsense mutations. Mutations are said to be *silent* when the altered codon encodes the same amino acid as the normal nucleotide sequence. For example, if a GUU is changed or mutated to a GUC, it still encodes the amino acid valine. Mutations are called *missense* when the substitution results in a changed codon that encodes a different amino acid. As an example, if a GUA is changed to a CUA, the resulting amino acid is changed from a valine to a leucine. Finally, nucleotide changes are called *nonsense mutations* when the alteration results in a “stop” signal codon instead of an amino acid codon. Stop codons include the base triplets UAA, UAG, and UGA. In the wrong place, these codons result in a truncated or shortened protein.

Deletions and insertions. The second class of mutations includes deletions and insertions, whereby nucleotide(s) are deleted from or added to the DNA sequence. If the changes are within the exons or coding portions of the DNA, they often result in a shift of the triplet reading frame and are thus called *frameshift mutations*. The triplet code “downstream” from the insertion or deletion (also said to be “3-prime” or “after” the insertion or deletion) is shifted out of phase, and the result is a nonsense code that may code for erroneous amino acids or a stop codon. Frameshift mutations are therefore also categorized as nonsense mutations (Fig. 54-2). In most frameshift situations, an accidental “stop” codon is encountered in the mRNA downstream of the mutation within the frameshift. Thus, insertions and deletions causing frameshifts almost always result in truncated or shortened proteins. The exception to this result occurs when the deletion or insertion of nucleotides is in a multiple of 3, whereby the triplet reading sequence stays in-frame, with only the insertion or deletion of amino acids. This error can still be deleterious, however, because the loss or gain of amino acids can alter protein product and function.

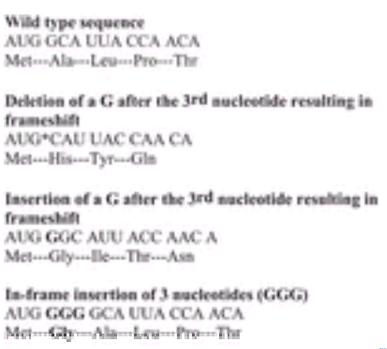


FIGURE 54-2. Nucleotide insertion and deletion mutations. Nucleotide insertions and deletions may cause in-frame or frameshift mutations. The deletion or insertion of nucleotides in multiples of other than 3 base pairs causes frameshift mutations that result in major alterations to the amino acid sequence. The insertion of nucleotides in multiples of 3 does not cause frameshift mutations but does cause alterations in amino acid sequence.

Splice site and regulatory mutations. Mutations also occur outside the coding sequence, or in the intron area. Introns contain specific nucleotide sequences immediately next to the exons that dictate exactly where the introns are to be excised. The sequence is a GT on the 5' end of the intron and an AG on the 3' end before the next exon. These sequences are conserved (i.e., not altered). Changes that do occur are called *splice site mutations*. The sequence of four to ten nucleotides into the intron on both the 5' and 3' ends is important as well, but not as rigorously conserved. Some mutations in these extended regions of introns can also result in improper splicing. When splice site mutations occur, an exon is often skipped, and a protein with a deleted segment is the result. A protein normally is encoded from a number of exons, so the result is a partial protein. Additionally, frameshifts can occur when the exons combine improperly, as previously described. Mutations can also occur in the regulatory segments of the DNA, although these are often difficult to identify. These types of changes can result in a failure of proteins to be expressed at the correct levels, in the correct tissues, or at the correct time.

Large deletions, duplications, translocations, and inversions. The last class of mutations consists of large deletions, duplications, translocations, and inversions of DNA. These typically arise during gamete formation as the consequence of an uneven exchange of DNA between chromosomes and can be passed down through generations. A *deletion* involves loss of genetic material, and *duplication* occurs when one section of genetic information is repeated more than once. *Translocations* occur when a section of DNA moves to another location on the same chromosome or onto a different chromosome. In an *inversion,* a section of DNA inverts itself into the opposite orientation. In each of these situations, the genetic changes can result in loss of expression of the gene, expression of a mutated or truncated protein, misregulation of the gene, or generation of a novel hybrid protein. Some of these mutations can be detected by cytogenetic methods that look at chromosomes. Large alterations in DNA, for example, can be detected by examining the Giemsa banding patterns of metaphase chromosomes for correct number and size (karyotyping) or by probing chromosomes with fluorescent DNA markers (fluorescent in situ hybridization, or FISH). The more subtle changes can be detected by Southern blotting methods.

Human Genome Project

The Human Genome Project was conceived in the 1980s, and an international multicenter program was organized in 1990. The program was designed to construct detailed genetic and physical maps of the human genome, to determine the complete nucleotide sequence of human DNA, to localize all genes within the human genome, and to sequence genomes of other model organisms. In 2001, a draft of the entire human genome was completed. [1], [2] Based on this initial sequence framework, it is estimated that the human genome contains 3 billion base pairs and 30,000 to 40,000 protein-coding genes. The number of protein-coding genes is significantly lower than the previously predicted 100,000 genes, but alternative splicing of RNA accounts for much of the protein complexity.

Data and research tools from the Human Genome Project are available to the public and can be assessed through the National Center for Biotechnology Information (NCBI) Web site. The NCBI Web site is a user-friendly resource that integrates multiple sources of public genome information (http://www.ncbi.nlm.nih.gov/genome/guide/human/). At this Web site, you can search for specific genetic markers or genes and also browse through a genomic region while using the information resources that you select. Most of the genetic loci that are displayed link to detailed descriptions of the gene or marker. Within the NCBI site is a useful search and retrieval program with which proteins and nucleotide sequences can be accessed (http://www.ncbi.nlm.nih.gov/Entrez/). The actual assembly of the draft genome sequence is located at http://genome.ucsc.edu/.

The Human Genome Project also recognizes the ethical, legal and social implications of the acquisition and use of the genetic knowledge by individuals and society, and it is driving policy discussions in both the public and professional arenas.

Genetic Tools and Technologies

The cooperative effort of the Human Genome Project, together with extensive genetic work in numerous laboratories, has given rise to powerful new tools and technologies for genetic research and clinical application. Familiarity with these tools is important for the practice of medicine as clinical genetic offerings continue to emerge.

Genetic Markers

Short tandem repeats. One tool developed throughout the 1990s is a set of genetic markers placed in order along chromosomes and spanning the entire human genome. Many of these markers are unique regions of the genome that contain variable numbers of nucleotide repeats resulting in variable length of the DNA segments in the human population. These are referred to as *short tandem repeats (STRs)* or *variable nucleotide tandem repeats (VNTRs)*. STRs can be repeats of any length, but the most utilized are the dinucleotide repeats (CACACA…) and the tetranucleotide repeats (AGATAGATAGAT…). Genotyping of individuals at these markers is used extensively in genetic linkage studies to search for new familial disease genes. This tool has thereby dramatically changed the pace of gene discovery because the entire genome is now systematically scanned by using these markers. Linkage by means of these markers is used in the clinical setting to identify mutant gene carriers when a specific mutation cannot be found.

Single-nucleotide polymorphisms. More recently, 1.4 million *single-nucleotide polymorphisms (SNPs)* have been identified within the human genome. [3] A SNP is a site on the DNA where a single base pair varies from person to person. SNPs can also be used as genetic markers for linkage and association studies, described later. Researchers estimate that 60,000 of these SNPs are within exons. In addition, a small portion of SNP variants encode amino acid changes (0.12%–0.17%) and may have functional consequences. Thus, a small fraction of SNPs are not only markers but may contribute to certain diseases.

Genetic Technologies

The DNA microchip. The DNA microchip is a new research tool with anticipated use in clinical genetic diagnosis in the near future. A DNA microchip contains sub-nanoliter amounts of DNA spotted on a solid surface, such as a silicon wafer or a glass microscope slide. Thousands of different DNA fragments can be spotted onto a single microchip, and each fragment is analyzed as an independent data point. Under appropriate thermodynamic conditions, DNA or RNA will bind to its specific complementary strand. Typically, the DNA or RNA to be tested is tagged with a fluorescent or radioactive label. Because RNA is readily degraded, it is copied into DNA, thereby called *complementary DNA (cDNA),* with the use of an enzyme discovered in viruses called *reverse transcriptase*. Many thousands of DNA sequences with known mutations are spotted onto a test microchip. DNA or cDNA from a test sample, possibly DNA from a cancer specimen, is then applied to the microchip. If binding to a specific sequence occurs, fluorescence is observed and the cancer in question contains the specific mutation on the chip. DNA microchips can be used to determine if genomic DNA has a mutation or if there are multiple copies of DNA in a sample (gene amplification), and to investigate differences in gene expression levels (microarray).

Microarray technology. Microarray technology is a method to examine simultaneously all the messenger RNA that is expressed or suppressed in a certain tissue, such as a polyp. A window is thereby obtained on the cellular pathways that are playing an active role in the tissue. The technique of microarrays involves the quantitative hybridization of a large panel of known cloned genes or synthetic oligonucleotides with the total cDNA derived from a particular cell or tissue type, such as the polyp. The overall hybridization to the microarray provides a comprehensive profile of the relative RNA message levels for all genes represented in the microarray chip. Although this technology is still in its infancy, it offers great potential for the molecular characterization of both normal and pathological tissues and of clinical diagnoses.

Gene Discovery Methods

A number of methodologies are used to identify the particular genes involved in inherited conditions. This work, called *gene discovery,* has been a major focus of molecular genetics for the past 15 years. The results have been rather remarkable, in that genes for most of the major inherited diseases have now been identified. Much work in gene discovery is now focused on less penetrant genes that appear to play a role in inherited susceptibility to many common diseases.

Some basic vocabulary of the human genome is important in understanding gene discovery work. The human genome is composed of 22 pairs of chromosomes, called *autosomes,* plus two sex chromosomes, XX for females and XY for males. Chromosomes segregate independently during gamete formation. The location of a gene or genetic marker on a chromosome is called a *locus*. Each person has two copies of each chromosome and consequently two copies of each gene (or set of genetic material), a maternal copy and a paternal copy. The two copies can vary, and each alone is called an *allele*. Multiple alleles of each genetic element may exist in the population, but an individual is limited to two alleles at each locus, a paternal allele and a maternal allele.

Linkage Studies Linkage analysis investigates whether specific genetic markers segregate with a certain disease or characteristic in a family. In a dominant genetic condition, one set of genetic markers near the gene responsible for the syndrome is passed from an affected parent to his/her offspring with a 50% probability. By following the unique pattern of genetic markers from affected parent to affected child, but not from affected parent to unaffected child, linkage to the genetic disorder can be established. In a recessive genetic condition, the set of genetic markers is followed from both parents, and the combination of two sets of genetic markers in offspring segregates with the genetic condition. In this case, both parents are usually clinically unaffected. The multiple applications of the basic concept underlying linkage analysis are described in later sections. Linkage analysis is used to find new genes causative of a genetic disorder. This requires obtaining blood specimens for DNA analyses from a large family or multiple unrelated families with the disorder in question. It is then clinically determined which family members have the disorder, a process called *phenotyping*. Next, the DNA of family members is analyzed, a process called *genotyping,* with multiple genetic markers across chromosomal regions. This may include all chromosomes when a candidate genomic region is not already identified. When certain genetic markers of known chromosomal location segregate with the disease in question, the disease gene is thereby known to be in that chromosomal location or at that locus. The statistical analysis used in this method is called *logarithm of the odds,* or *lod score*. The lod score compares the likelihood of a real association with the likelihood of chance association. Lod scores of 3.0 or higher are considered significant. A genetic marker within 5 centimorgans (cM, about 5 million base pairs of DNA) of a disease gene is considered useful for genetic counseling. Identification of the disease gene typically requires a better resolution of the locus because more than 50 genes can be contained within a 5-cM span.

Sibling Pair Studies Sibling pair studies are being increasingly used to identify common disease genes, such as those for inflammatory bowel disease, coronary heart disease, diabetes, and colon cancer. Hundreds of siblings, both of whom have the disease in question, are identified and undergo genetic analysis. As in the family linkage studies, DNA from the sibling pairs is analyzed with multiple genetic markers in the human genome. The linkage analysis involves the identification of genetic loci where the population of affected siblings co-inherit a significantly higher average number of alleles than would be expected by normal mendelian inheritance.

Association Studies In contrast to family or sibling pair studies, association studies are population based. Association studies investigate differences in allele frequencies between persons with a specific disorder (cases) and individuals without the disorder (controls). If an allele frequency is statistically different in the case population, it is considered to have a causative role in the disorder. This role may be direct, in which the allele is a causative change, or indirect, as the consequence of proximity of the allele to another disease gene. The relative risk imposed by an allele associated with a disorder can be calculated from the magnitude of the difference in allele frequency between the case and control populations.

Mutation Detection

Use of Mutation Detection Once genes and their locations have been identified, mutation detection is often possible for both research and the diagnosis of clinical disease. Mutation detection is the identification of the specific disease-causing mutation in a person or family. Most inherited conditions arise from multiple different mutations in the same gene (allelic heterogeneity). However, a separate and distinct mutation segregates in each family with a particular disease. The success of identifying this causative mutation in an index case (i.e., a case clinically known to have the disease) varies depending on the disease. Once a mutation has been found in an index case, other family members can be tested for the presence or absence of that specific mutation with nearly 100% accuracy. If a mutation cannot be found in the index case, however, then all family members must be treated as potentially having the disease gene. Alternatively, linkage analysis can be used to indicate the likelihood that individual family members carry the mutant gene.

Specific Techniques Used in Mutation Detection

Sequencing. Sequencing is a standard method used to detect mutations. Sequencing is the process of determining the order of nucleotide bases (A, C, G, and T) for all the coding regions and certain intron regions of the gene of interest. Although sequencing is the gold standard for clinical genetic testing, it has limitations, including high cost and failure to detect some genetic rearrangements, deletions, and mutations in regulatory regions. Currently, sequences can be resolved on stretches of DNA 500 to 800 base pairs in length. Sequencing is often more effective when complemented with an initial DNA screening method that detects the presence of a mutation and narrows the DNA area to be sequenced. The next three methods described represent such screening methods. Once one of these methods indicates a DNA variant, sequencing is required to confirm the change and determine if it is a normal variant in the human population or a disease-causing mutation.

Conformation-specific gel electrophoresis (CSGE). CSGE, also called *heteroduplex analysis (HA),* is one method used to complement sequencing. Numerous copies of the specific region of genomic DNA, from 150 to 300 base pairs in length, are synthesized by the polymerase chain reaction (PCR), then denatured and renatured. If a sequence variation in the two alleles of genomic DNA is present, some strands will renature as a hybrid of the wild type and the variant DNA strands, called a *heteroduplex*. When analyzed by gel electrophoresis, the hybrid DNA will migrate through the gel matrix at a different rate and can be visualized as a distinct band on the gel. CSGE is thought to detect more than 90% of mutations.

Single-strand conformation polymorphism (SSCP). Like CSGE, SSCP initially requires PCR amplification of a specific region of DNA. The resulting double-stranded DNA is then denatured and quickly renatured so that some strands are not able to find their complementary strand before renaturation. This results in a population of single-stranded DNA folded on itself, forming a secondary structure. The renatured DNA is next run on a gel, and DNA variants that vary from the normal sequence form unique secondary structures that migrate at a different rate within the gel matrix. This method has been successfully used to detect mutations; however, it is only 60% to 95% effective and is dependent on the DNA sequence being examined and the laboratory conditions used.

Denaturing-gradient gel electrophoresis (DGGE). DGGE is similar to CSGE, but it differs in that the double-stranded DNA is run through a gel matrix that has an increasing gradient of urea and formaldehyde, chemicals that denature DNA. Variations in DNA sequence will denature under different stringencies and consequently migrate differently in the gel matrix. The DNA can be visualized, and differences (mutations) are detected. DGGE can detect as many as 95% of sequence changes.

Protein truncation test (PTT). PTT uses in vitro methods to synthesize protein from the DNA in question. Viable cells from the subject are required to generate a full-length cDNA of the gene. If a nonsense mutation is present (i.e., one that results in a shortened protein), two bands are observed on the electrophoresis gel, one for the normal protein and one for the shortened protein. PTT fails to detect changes in cases of a large deletion, or if the truncation is at the very start or the very end of the gene or gene segment. In the clinical setting, PTT is used to indicate the presence of a truncating mutation, and it may be followed by sequencing to determine

the exact mutation.
Linkage analysis. Linkage analysis is used not only in gene discovery but also clinically to determine the likelihood that a person at risk carries the mutant gene. Linkage analysis can be used if the gene location is known but a specific mutation has not been identified in the family. The method is identical to that described in the section “Gene Discovery Methods” and is informative in 90% to 95% of families that are able to submit the appropriate samples from multiple members for testing.
Site-specific mutation analyses. Once a specific mutation has been identified in an index case of a family, “site-specific mutation analyses” are used to examine the presence or absence of that particular mutation in other family members. A number of PCR-based methods are used, often specific to the mutation found in the family. The various techniques are not described here, but of note, these techniques are all highly accurate and much less expensive than the original mutation identification in the index case of a family. Familiarity with the types of mutations that occur in specific diseases is important and determines the specific test ordered. Site-specific mutation analysis for specific mutations, for example, is performed for hemochromatosis and cystic fibrosis, whereas complete gene sequencing are performed for the diagnosis of familial adenomatous polyposis (FAP).

Types of Genetic Disorders

Genetic disorders can be categorized into four major groups. Knowledge of the types of genetic disorders is important to understanding the etiology, occurrence, and recurrence risks of inherited diseases.

Chromosomal Disorders Chromosomal disorders occur when the chromosomal number or structure differs from that normally expected. A person with a chromosomal disorder may have an extra chromosome (e.g., Down syndrome, in which an individual has three copies of chromosome 21) or lack a chromosome (e.g., Turner syndrome, in which a female subject has one instead of two X chromosomes). Chromosomal disorders also include duplication, absence, or structural rearrangement of large segments of chromosomes (e.g., DiGeorge sequence). Examples of chromosomal disorders and their GI manifestations can be found in Table 54-1. If a chromosomal disorder is suspected, karyotype or other analysis can be performed on blood lymphocytes.

CHROMOSOMAL DISORDERS	GASTROINTESTINAL MANIFESTATIONS
Duplication of 3q	Umbilical hernia, omphalocele
Deletion of 9p	Inguinal and/or umbilical hernia, diaphragmatic hernia
Deletion of 11q	Pyloric stenosis, inguinal hernia
Microdeletion of 22q 11.2 (DiGeorge sequence)	Esophageal atresia, velopharyngeal insufficiency, imperforate anus, diaphragmatic hernia
Trisomy 13 (Patau syndrome)	Omphalocele, heterotropic pancreatic or splenic tissue, incomplete rotation of colon, Meckel diverticulum, enlarged gallbladder, diaphragmatic defect
Trisomy 18 (Edwards syndrome)	Inguinal or umbilical hernia, malposed or funnel-shaped anus, hypoplastic diaphragm, Meckel diverticulum, heterotopic pancreatic and/or splenic tissue, omphalocele, incomplete rotation of colon, pyloric stenosis, extrahepatic biliary atresia, hypoplastic gallbladder, gallstones, imperforate anus
Trisomy 21 (Down syndrome)	Tracheal stenosis with hourglass trachea and midtracheal absence of tracheal pars membranacea, tracheoesophageal fistula, duodenal atresia, omphalocele, pyloric stenosis, annular pancreas, Hirschsprung disease, imperforate anus

TABLE 54-1 Examples of Chromosomal Disorders with Associated Congenital Gastrointestinal Manifestations

Additionally, chromosomal mosaicism is possible, in which an individual’s genetic makeup comprises two or more populations of cells that vary in chromosomal content. Mosaicism occurs when one cell sometime later in embryonic development acquires a mutation during mitosis. Any future cells (clonal population) derived from this one stem cell will contain the same genetic error. Consequently, the individual will have both normal and abnormal cells and is mosaic for the chromosomal disorder. Mosaicism may or may not affect the phenotype of the individual or pose a risk to his/her offspring.
Mendelian Disorders Mendelian disorders are named after Gregor Mendel, an Austrian monk who lived in the mid-19th century and is considered to be the father of genetics. Mendelian disorders are caused by mutations in single genes and can be passed to offspring in an autosomal dominant, autosomal recessive, or sex-linked fashion. Although a mutant gene may cause the condition, the severity of the disease may be influenced by environmental factors. Examples of single-gene disorders affecting the GI tract are listed in Table 54-2 and Table 54-3, and the rules of inheritance are discussed in a later section.

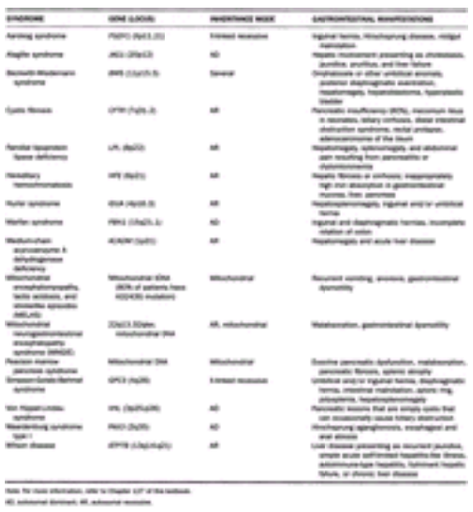


TABLE 54-2 Examples of Monogenetic (Mendelian) Disease with Congenital Gastrointestinal Manifestations

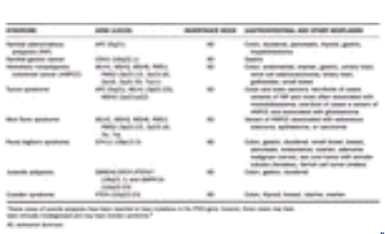


TABLE 54-3 Hereditary Cancer Syndromes Predisposing to Increased Risk for Gastrointestinal Neoplasms

Mitochondrial Disorders Mitochondria, organelles found in the cytoplasm of a cell, produce adenosine triphosphate (ATP), an energy-rich molecule, through the oxidative phosphorylation process. Mitochondria have their own DNA molecules, distinct from nuclear DNA, and these code for genes important in oxidative phosphorylation. Mutations in the mitochondrial genes cause several genetic conditions, including Pearson marrow pancreas syndrome (see Table 54-2). Mitochondria are maternally inherited through the mitochondria in the cytoplasm of the egg. Sperm have few mitochondria; none of them enter the egg during fertilization and thus are not passed on to offspring. A female affected with a mitochondrial disorder will pass the disease to all her offspring (100% transmission), whereas no offspring will inherit the disorder from an affected male (Fig. 54-3).

FIGURE 54-3. Typical pedigree showing mitochondrial inheritance. Males are represented by *squares* and females by *circles.* All offspring of an affected female will inherit the condition, whereas none of the children of an affected male will.

Multifactorial/Complex Disorders Although much is understood about chromosomal, mendelian, and mitochondrial disorders, these conditions reflect only a small proportion of inherited diseases. Multifactorial/complex disorders are more common and currently less well understood. As their name implies, these conditions are caused by a combination of factors through gene-gene interactions or gene-environment interactions. Twin studies can be used to assess the quantitative impact of gene and environmental influences on diseases. One twin study, for example, found that approximately 35% of colon cancers arise from an inherited predisposition. [52] The known inherited colorectal cancer predisposition syndromes constitute 5% to 10% of the familial aggregation, implying that additional colon cancer susceptibilities are yet to be defined.

Modes of Inheritance

Mendelian (monogenetic) disorders include those passed to offspring through autosomal dominant, autosomal recessive, and sex-linked forms of inheritance. As previously discussed, humans have two copies (alleles) of each gene located on the autosomes. If both alleles at the locus are the same, the individual is homozygous for that gene. If two different alleles are present at a given locus, the individual is a heterozygote.

Autosomal Dominant The basis of autosomal dominant inheritance is that an alteration in *one* of the two copies of the gene leads to disease. One allele has a mutation, and the corresponding allele on the homologous chromosome is normal. Autosomal dominant conditions express themselves in the heterozygous state. Figure 54-4 shows a typical autosomal dominant pedigree. The rules of autosomal dominant inheritance are as follows: An affected individual has a 50% chance of passing on the mutant gene and a 50% chance of passing on the normal gene to each offspring.
The condition appears to be transmitted vertically (i.e., multiple generations are affected).
Both male and female parents can pass on the mutant gene to both male and female offspring (i.e., male-to-male transmission occurs).
An affected individual has an affected parent and generations are not skipped (exceptions are new mutation and incomplete penetrance).

FIGURE 54-4. Typical pedigree showing autosomal dominant inheritance. Males and females are equally affected. Vertical transmission of the condition is apparent.

Autosomal Recessive Autosomal recessive conditions require that an individual inherit *two* copies of a mutant gene at a given locus, one disease-causing allele from each parent (Fig. 54-5). Autosomal recessive conditions express themselves in the homozygous state. Individuals who are heterozygous for a recessive condition (i.e., have only one mutant allele) typically do not manifest symptoms of the condition and are referred to as *carriers*. In some conditions, carrier status affords a selective advantage (e.g., carriers of sickle cell anemia are resistant to malaria). The rules of autosomal recessive inheritance are as follows: A mating between two carrier parents results in a 25% chance that they will have an affected (homozygous) child, and a 75% chance that they will have a normal child. Of the phenotypically normal children, two thirds will be carriers and one third will be homozygous normal.
The condition appears to be transmitted horizontally (i.e., multiple individuals in one generation can be affected).
Males and females are affected equally.

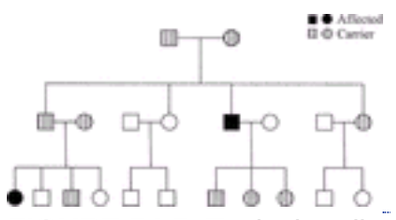


FIGURE 54-5. Typical pedigree showing autosomal recessive inheritance. Males and females are equally affected. An individual must inherit two copies of a mutant gene at a given locus to be affected.

Sex-Linked Sex-linked disorders involve the sex-determining X and Y chromosomes. The difference in sex chromosome composition between males and females is the basis for understanding X-linked conditions. Men who inherit an abnormal gene on the X chromosome (the X chromosome in males is always inherited from the mother) will be affected with the associated condition. Because women have two X chromosomes, the inheritance is similar to that of autosomal dominant and recessive conditions. In X-linked dominant conditions, one mutant allele is sufficient to cause the condition in females, whereas two mutant alleles are required to cause the condition in X-linked recessive disease in females. A typical pedigree showing X-linked recessive inheritance appears in Figure 54-6. X-linked recessive disorders are more common than X-linked dominant conditions. The rules of X-linked recessive inheritance are as follows: Affected men are common, whereas affected women are not (unless an affected man and a carrier woman produce offspring).
All the daughters of an affected male will be carriers. None of his sons will be affected because fathers pass on a Y chromosome to their sons, not an X chromosome (no male-to-male transmission).
Male offspring of carrier women have a 50% chance of being affected and a 50% chance of being unaffected.
Female offspring of carrier women have a 50% chance of being a carrier and a 50% chance of having two normal alleles.
Because affected men have carrier daughters, who in turn have affected sons, the condition appears to “skip” generations.
Rarely with certain X-linked recessive conditions, female carriers display mild symptoms of the disease (e.g., fragile X syndrome).

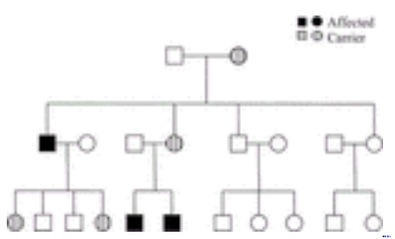


FIGURE 54-6. Typical pedigree showing X-linked recessive inheritance. Males are more likely to be affected, whereas females are more likely to be carriers.

Other Rules of Genetics Knowledge of the basic modes of inheritance is important in assessing occurrence and recurrence risks for patients. Other genetic concepts must also be applied to provide accurate education and risk assessment to individuals and their families.
Delayed age at onset. FAP demonstrates the feature of delayed age at onset. Children born with the FAP genotype have one mutant copy of the *APC* gene on chromosome 5 and typically do not exhibit any of the phenotypic characteristics of the condition in childhood (see Table 54-3). In FAP, the average age at polyp development is 16 years. The presence of a deleterious genotype does not necessarily lead to a phenotypic expression from birth.
Penetrance. *Penetrance* refers to the likelihood of disease expression if the disease-causing gene is present. For example, individuals with FAP are thought to have nearly a 100% chance of developing colon polyps and subsequent colon cancer if prophylactic colectomy is not performed, and therefore the penetrance of FAP is

100%. The likelihood of an individual with hereditary nonpolyposis colorectal cancer (HNPCC) developing colon cancer in his/her lifetime is close to 80%; therefore, the penetrance of colon cancer in HNPCC is 80%. Every clinical feature of a genetic condition has a penetrance level.

Nonpenetrance. Fundamental to the interpretation of the family history is an awareness of nonpenetrance. Nonpenetrance is illustrated by the individual with a mutant genotype in whom the disease or condition never develops. Nonpenetrance may complicate the interpretation of the family history, especially in the case of conditions inherited in an autosomal dominant pattern, which one would expect to observe in all successive generations. Nonpenetrance is also illustrated by the individual who dies of other causes before the disease develops or who undergoes prophylactic surgery.

New mutation. The clinician must also consider the possibility of a new mutation. Mutations in human genes may occur randomly during gamete maturation or very early in embryogenesis. A mutation will be replicated and become a part of every cell in the body of the affected individual. Every genetic condition has an associated new mutation rate. For example, although reports of new mutations in the genes associated with HNPCC are rare, 30% of individuals in whom FAP is newly diagnosed and do not have a previously known family history of FAP represent new mutations (i.e., neither parent has an *APC* mutation).

Pleiotropy. Many genes, including mutated *APC* genes that cause FAP, exhibit pleiotropy. Pleiotropy occurs when a mutated gene has more than one phenotypic effect on the body. For example, in an individual with FAP, colonic polyps, desmoid tumors, osteomas, or thyroid cancer can develop. Although colon cancer is the greatest risk, it is not the only phenotypic effect. The other findings are said to be pleiotropic manifestations of the mutant gene.

Variable expressivity. Variable expressivity is the concept that the same genetic condition is expressed to a different extent in different individuals. In some individuals with HNPCC, for example, multiple cancers, including colon cancer, uterine cancer, and urinary tract cancer, develop, whereas only colon cancer may develop in other individuals. Gene expression can be modified by genes, environmental factors, and other unknown factors.

Genetic heterogeneity and locus heterogeneity. Critical to identifying the possible etiology of a clinical presentation is the awareness that similar phenotypes may have different genetic causes. For example, although pancreatitis is strongly associated with environmental factors such as alcohol use or gallstones, mutations in three genes have been shown to be associated with pancreatitis. These include the genes for cystic fibrosis (*CFTR*), cationic trypsinogen (*PRSS1*), and a trypsin inhibitor (*PST1* or *SPINK1*). Table 54-4 outlines several GI diseases and their genetically heterogeneous causes. The term *locus heterogeneity* indicates that mutations in any one of several related genes may result in the same genetic disorder. Although FAP is not considered to have locus heterogeneity (all cases of FAP are thought to be caused by mutations in the *APC* gene), HNPCC is a condition caused by mutations in any one of several DNA mismatch repair genes (*MLH1, MSH2, MSH6, PMS1, PMS2*). Knowledge of the genetic conditions that have locus heterogeneity is essential in ordering the proper genetic test.

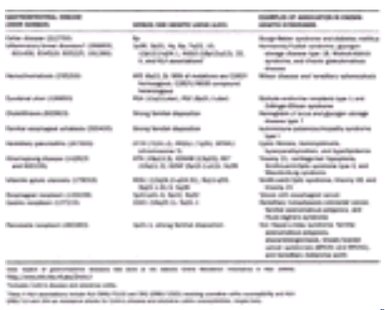


TABLE 54-4 Gastrointestinal Disease Displaying Genetic Heterogeneity

Allelic heterogeneity. One contribution to variable expressivity is allelic heterogeneity. The term *allelic heterogeneity* simply means different mutations in alleles of the same gene. These different mutations may result in different and distinct phenotypes. A good example is FAP, in which some features are related to the location of the mutation in the *APC* gene. Mutations in the 5' or 3' (beginning or end) region of the *APC* gene are generally associated with an attenuated version of FAP (also called *attenuated adenomatous polyposis coli*). In this condition, fewer polyps form at later ages than in typical FAP. [4] In contrast, mutations located between codons 1250 and 1464 are associated with a diffuse (>5000 polyps) phenotype. [5] An ongoing area of study involves the correlation of genotype to phenotype and understanding genes well enough to know the molecular and health impact of mutations in certain areas of the gene.

Compound heterozygosity. Compound heterozygosity occurs when an individual has two different disease-causing alleles at a specific locus. Individuals with compound heterozygosity can be affected with an autosomal recessive condition such as cystic fibrosis. The *CFTR* gene is responsible for cystic fibrosis when both alleles contain mutations. Seventy percent of all *CFTR* mutations are caused by deletion of 3 base pairs at position 508 of the gene, commonly called the *?508 mutation*. Many individuals with cystic fibrosis have two ?508 mutations and are homozygotes. Individuals who have a ?508 mutation on one allele and a different mutation, such as the R117H mutation, on the second allele are compound heterozygotes. Different disease-causing mutations can affect the phenotype of an individual, as explained in the section “Allelic Heterogeneity,” and this holds true for autosomal recessive conditions. For example, individuals with cystic fibrosis who have two ?508 mutations almost always have pancreatic insufficiency, whereas compound heterozygotes with one ?508 mutation and one R117 mutation are less likely to have pancreatic insufficiency.

Somatic versus germ-line mutations. In the field of cancer genetics, it is important to understand the concept of somatic versus germ-line mutations. Somatic mutations are acquired mutations. Only the daughter cells of the somatically mutated cell carry the mutation. Somatic mutations are not found in cells in other parts of the body and cannot be passed on to offspring. Germ-line mutations are inherited mutations or those that occur at the beginning of embryogenesis, so that they are present in every cell of the body. An individual with a germ-line mutation can pass the mutation on to his/her offspring. Mutations of the *APC* gene, for example, may be germ-line or somatic. Germ-line *APC* mutations are present in every cell of the colon (and body), so that colonic polyposis and FAP develops and the disease is passed on to offspring. Somatic *APC* mutations in a colonic epithelial cell appear to be the first step in the development of a sporadic polyp. Only the daughter cells carry the somatic mutation. A single polyp eventually forms from this clone of cells. The mutation is not found in other cells of the colon or body, however, and therefore cannot be passed on to offspring.

PRINCIPLES OF GENETIC COUNSELING

Definition

The first official definition of genetic counseling was proposed and adopted by the American Society of Human Genetics (ASHG) in 1975. The practice was defined as follows:

> Genetic counseling is a communication process that deals with the human problems associated with the occurrence or risk of occurrence of a genetic disorder in a family. This process involves an attempt by one or more appropriately trained persons to help the individual or family to: (1) comprehend the medical facts including the diagnosis, probable course of the disorder, and the available management, (2) appreciate the way heredity contributes to the disorder and the risk of recurrence in specified relatives, (3) understand the alternatives for dealing with the risk of recurrence, (4) choose a course of action which seems to them appropriate in view of their risk, their family goals, and their ethical and religious standards and act in accordance with that decision, and (5) make the best possible adjustment to the disorder in an affected family member and/or to the risk of recurrence of that disorder. [6]

Modern genetic counseling practice continues to encompass these core components. Additionally, as the repertoire of clinically available molecular tests continues to grow, patients and their families should be educated about available genetic tests together with the risks and benefits of genetic testing.

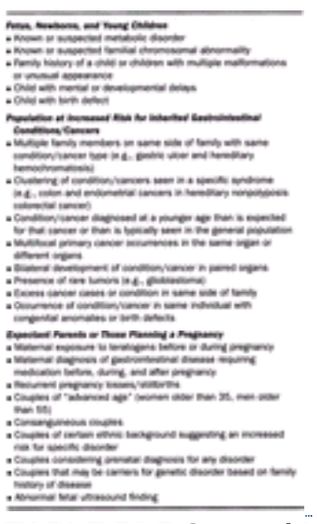
Genetic Providers

Medical geneticists (PhD or MD), advanced practice nurses with genetic certification (MSN), and genetic counselors (MS) are specialty-trained clinical genetic practitioners who provide education, a diagnosis based on the clinical findings or results of genetic testing, therapeutic management and options, and psychosocial support throughout the duration of care for the patient at risk for or affected with a genetic disorder. Genetic providers also stay abreast of the social-ethical, legal, ethnic-cultural, and financial issues that surround genetic testing.

Although the majority of genetic services are in obstetrical care (>60%), other subspecialties have quickly developed because of the expanding knowledge of the genetic etiology of human disease. Subspecialty areas include cancer, neurogenetics, psychiatric, and pediatric counseling. In genetics clinics, a consultation may require a multidisciplinary team comprising PhD/MD geneticists, surgeons, genetic counselors, nurses, nutritionists, and other practitioners of health care disciplines to address the patient’s complex needs and manage the pleiotropic effects of a genetic condition. Some gastroenterologists may have specialty training or knowledge to provide genetic counseling, whereas others may find it valuable to consult with or refer to genetic practitioners. A directory of national and international genetic counselors and geneticists can be found at the Web sites http://www.nsgc.org/resourcelink.asp and http://www.faseb.org/genetics/memb-dir/ashg.htm, respectively. The following is an overview of clinical genetic counseling for the gastrointestinal patient.

Components of Clinical Genetic Counseling

Indications for Counseling Any patient with a GI condition for which a genetic etiology is suspected or known is an appropriate candidate for genetic counseling. The genetic counseling session is collaborative in nature because of the tremendous amount of education and personal decision making involved for the patient. [7], [8] Family members are often seen conjointly with the patient because of the hereditary nature of the disease. The patients who undergo genetic counseling for most GI diseases with a known genetic etiology can generally be grouped by age or special populations, and although the list is not exhaustive, the common considerations for a genetic consultation are summarized in Table 54-5.



Fetus, Newborns, and Young Children
- Known or suspected metabolic disorder
- Known or suspected familial chromosomal abnormality
- Family history of a child or children with multiple malformations or unusual appearance
- Child with mental or developmental delays
- Child with birth defect

Population at Increased Risk for Inherited Gastrointestinal Conditions/Cancers
- Multiple family members on same side of family with same condition/cancer type (e.g., gastric ulcer and hereditary hemochromatosis)
- Clustering of condition/cancers seen in a specific syndrome (e.g., colon and endometrial cancers in hereditary nonpolyposis colorectal cancer)
- Condition/cancer diagnosed at a younger age than is expected for that cancer or than is typically seen in the general population
- Multifocal primary cancer occurrences in the same organ or different organs
- Bilateral development of condition/cancer in paired organs
- Presence of rare tumors (e.g., glioblastoma)
- Excess cancer cases or condition in same side of family
- Occurrence of condition/cancer in same individual with congenital anomalies or birth defects

Expectant Parents or Those Planning a Pregnancy
- Maternal exposure to teratogens before or during pregnancy
- Maternal diagnosis of gastrointestinal disease requiring medication before, during, and after pregnancy
- Recurrent pregnancy losses/stillbirths
- Couples of "advanced age" (women older than 35, men older than 55)
- Consanguineous couples
- Couples of certain ethnic background suggesting an increased risk for specific disorder
- Couples considering prenatal diagnosis for any disorder
- Couples that may be carriers for genetic disorder based on family history of disease
- Abnormal fetal ultrasound finding

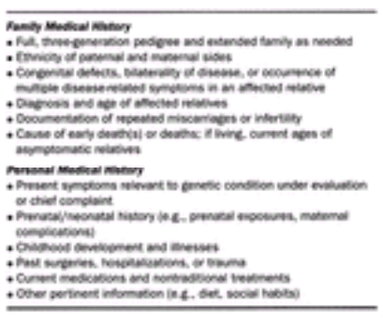
TABLE 54-5 General Indications for Genetic Consultation Based on Needs of Special Populations

Counseling for conditions involving fetuses, newborns, or young children. Fetuses, newborns, or young children who present with congenital GI malformations may have chromosomal abnormalities or single-gene mutations. For example, 25% of fetuses with cystic fibrosis and 12% of fetuses with chromosomal disorders present with an echogenic bowel seen on ultrasonography during the second trimester. It is currently estimated that more than half of all birth defects have a genetic origin. Some types of GI dysfunction may be seen as part of a syndrome or display genetic heterogeneity (see Table 54-2, Table 54-3, and Table 54-4). Pyloric stenosis is such a condition. It is an associated feature in some genetic syndromes but is also seen as an isolated condition. Single-gene mutations for isolated pyloric stenosis have been mapped to several chromosomal regions.

Counseling for adults and for cancer predisposition. Although some predominantly GI diseases of young adolescent/adult population have been linked to genes or genetic loci, most GI diseases for which genetic testing is commercially available relate to cancer predisposition. A small but significant number of colon cancers (5%–10%) belong to a distinct class of inherited colorectal cancer predisposition syndromes (see Table 54-3). [9] Lastly, expectant mothers or women considering pregnancy who may be exposed to teratogens (external agents such as GI medications) that may affect fetal GI or other organ development will benefit from a genetic consultation.

Personal and Family Medical History

Pedigree. At the core of genetic counseling is a family "pedigree." A pedigree is a schematic diagram of familial generations, including personal and family medical history, in which standardized symbols are used. [10] Pedigrees aid geneticists in evaluating for the inheritance pattern of disease and therefore should include both the family history and the personal medical history of the patient (Table 54-6). [11] The information gathered is pertinent to propose the differential diagnoses of genetically related GI disorders, to provide risk assessment, to determine which diagnostic test to order, to interpret genetic test results accurately, and to guide individualized medical intervention. When genetic tests are unavailable or declined by the patient, the family history remains valuable in providing targeted surveillance and aiding in management decisions. [12]



Family Medical History
- Full, three-generation pedigree and extended family as needed
- Ethnicity of paternal and maternal sides
- Congenital defects, bilaterality of disease, or occurrence of multiple disease-related symptoms in an affected relative
- Diagnosis and age of affected relatives
- Documentation of repeated miscarriages or infertility
- Cause of early death(s) or deaths; if living, current ages of asymptomatic relatives

Personal Medical History
- Present symptoms relevant to genetic condition under evaluation or chief complaint
- Prenatal/neonatal history (e.g., prenatal exposures, maternal complications)
- Childhood development and illnesses
- Past surgeries, hospitalizations, or trauma
- Current medications and nontraditional treatments
- Other pertinent information (e.g., diet, social habits)

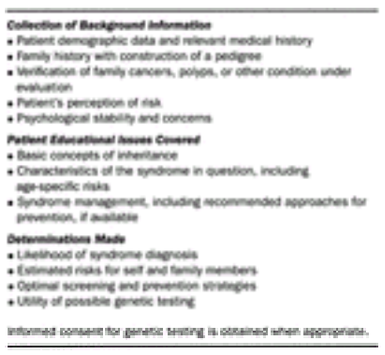
TABLE 54-6 Suggested Components of Personal and Family History

Accuracy and perceptions. Generally, individuals report accurate medical diagnoses for first-degree relatives (parents, children, siblings) and second-degree relatives (grandparents, aunts, uncles, grandchildren), but reports for distant relatives may be less accurate. [13], [14] Confirmation of the information provided by the patient through medical records collection and review can be insightful and necessary. Subtle differences detected from pathology records or a repeated consultation of a previous histopathological report can change the diagnosis, affect appropriate medical recommendations, and necessitate different genetic testing or screening. For example, it is common for family members to know if a close relative has colonic polyps, but it is unusual for the patient to know whether the polyps are adenomatous, hyperplastic, or hamartomatous. Knowledge of the histology of polyps is highly pertinent when a family is evaluated for an inherited colon cancer condition (see Table 54-3). A review of the family history information during clinic visits can also provide an opportunity to learn about the patient's health beliefs. An example of beliefs would be, "Aunt Mary got cancer because she has a stressful job," and an example of family dynamics would be, "My brother and I haven't spoken since I was diagnosed with cancer."

Risk Assessment and Physical Evaluation High, moderate, and general population (average) risks are defined as part of the counseling evaluation. Risk assessment involves a discussion of the likelihood of a possible genetic component and its implications. An individual or family that fulfills one or more of the criteria listed in Table 54-5 is generally considered at increased risk and appropriate for genetic counseling to identify the role of contributing hereditary factor(s) and provide risk assessment. Genetic testing ultimately defines the genetic risk with precision. The results of genetic testing can direct those with the inherited condition in question to proper management, and at the same time allow those without the condition to avoid unnecessary medical screening and management procedures. Depending on the condition suspected, a physical examination can be extremely important in determining the likelihood it is present. FAP is a good example. The presence of associated extracolonic manifestations, such as osteomas, fibromas, and congenital hypertrophic retinal pigment epithelium, in a patient at risk makes the diagnosis much more likely. The family and personal history, environmental risk factors, and complete physical examination are collectively meaningful in an assessment of the probability that a genetic disease is present. Mathematical predictive models that utilize this information are emerging to assist with risk assessment and to predict the likelihood of finding a mutation with genetic testing for some conditions. Interestingly, some insurance companies already rely on these probability models to determine eligibility for the coverage of molecular testing costs. The general rule often followed by genetic providers is that it is appropriate to offer available diagnostic genetic testing to individuals whose chance of having a genetic mutation is 10% or more.

Education and Genetic Discrimination

Content of patient genetic education. Patient education consists of information about the nature and etiology of the disorder, its natural history and prognosis, the mode of inheritance, the occurrence or recurrence risks, the recommended prevention, screening and treatment options, the availability of research studies, and the possibilities for prenatal diagnosis (Table 54-7). Helping patients understand the basic principles of genetics and disease can dispel myths. It is not uncommon for individuals with a strong family history of a condition to develop their own beliefs about the inheritance, such as "The disease only affects males in my family" when the disease is actually autosomal dominant. Very common is an association with physical features, such as "Everyone in my family with red hair gets cancer."



Collection of Background Information
- Patient demographic data and relevant medical history
- Family history with construction of a pedigree
- Verification of family cancers, polyps, or other condition under evaluation
- Patient's perception of risk
- Psychological stability and concerns

Patient Educational Issues Covered
- Basic concepts of inheritance
- Characteristics of the syndrome in question, including age-specific risks
- Syndrome management, including recommended approaches for prevention, if available

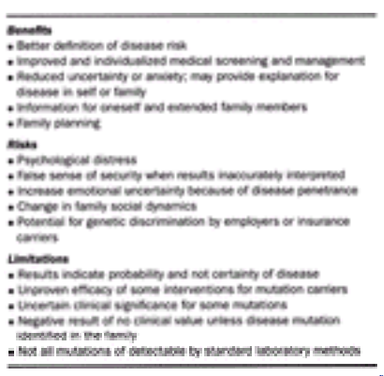
Determinations Made
- Likelihood of syndrome diagnosis
- Estimated risks for self and family members
- Optimal screening and prevention strategies
- Utility of possible genetic testing

Informed consent for genetic testing is obtained when appropriate.

TABLE 54-7 Areas to Be Covered by Genetic Counseling

Informed consent for genetic testing is obtained when appropriate. In a discussion of the quantitative risks of health problems associated with an inherited condition, it is important to recognize that many patients do not understand statistical terminology. It may often be better, for example, to state that eight of ten individuals with HNPCC will acquire colon cancer in their lifetime, rather than that the lifetime risk of associated colon cancer is 80%. It may also be useful to compare risks with the risks in the general population. For example, although 6% of Americans will acquire colon cancer in their lifetime, the risk of individuals with HNPCC for the development of colon cancer is increased approximately 13-fold. Risk communication geared towards the patient's educational background and personal experience will improve understanding and facilitate compliance with the recommended surveillance.

Issues, benefits, and risks of genetic testing. In a discussion of the option of genetic testing, education should include applicability, cost, turnaround time, and the personalized meaning of results. It should also include the benefits, risks, and limitations of testing (Table 54-8). Particular concerns of the patient are inevitably the insurance issues and the employment and social discrimination that may follow a genetic diagnosis. Consumer education by the genetic provider is an approach that may involve many practitioners and can often facilitate the decision regarding genetic testing for the patient. [15], [16] and [17] Additionally, individuals who are considering genetic testing need to be informed about current federal and state legislation regarding genetic privacy and the potential for genetic discrimination. [18] Presently, 42 states have enacted some level of protection for genetic privacy, but fewer states have legislative protection in place for employment, disability insurance, and life insurance (www.nhgri.nih.gov/Policy_and_public_affairs/Legislation/insure.htm). There are some federal safeguards for genetic information. One example is the Health Insurance Portability and Accountability Act of 1997 (Kennedy-Kassebaum Bill), which precludes the use of genetic information as the basis for a preexisting condition when a person is asymptomatic. This law also prohibits insurers from using genetic information to exclude a person from group coverage or individually charging higher premiums within a group. In the context of employment, the Americans with Disabilities Act of 1990 now includes "genetic or medically identified potential of or predisposition toward a physical or mental impairment that substantially limits a major life activity" as a definition of a disability. To date, no cases have been prosecuted with this act as legal grounds. Loopholes exist in these federal and states laws, and the potential remaining risk for the misuse of genetic information by third parties is unclear.



Benefits
- Better definition of disease risk
- Improved and individualized medical screening and management
- Reduced uncertainty or anxiety; may provide explanation for disease in self or family
- Information for oneself and extended family members
- Family planning

Risks
- Psychological distress
- False sense of security when results inaccurately interpreted
- Increase emotional uncertainty because of disease penetrance
- Change in family social dynamics
- Potential for genetic discrimination by employers or insurance carriers

Limitations
- Results indicate probability and not certainty of disease
- Unproven efficacy of some interventions for mutation carriers
- Uncertain clinical significance for some mutations
- Negative result of no clinical value unless disease mutation identified in the family
- Not all mutations of detectable by standard laboratory methods

TABLE 54-8 Benefits, Risks, and Limitations of Genetic Testing

At the time of this writing, it appears that despite the cited laws, some risk for genetic discrimination remains, even though it appears to be very low and often not different from that of an individual with a clinical diagnosis or known familial risk. Nonetheless, it is the obligation of the provider to outline appropriately the risks and benefits of a genetic diagnosis. In many cases, the medical benefit of a genetic diagnosis far outweighs the risks, but in some instances, the reverse may be true. Thus, an informed and supportive discussion of the issues is extremely important and will allow patients to reach decisions on genetic testing that are appropriate for them and their families. Lack of such a discussion may well lead to inappropriate decisions based on lack of knowledge, fear, and misconception. Education about familial risks and genetic results not only provides the setting for appropriate testing decisions but can also promote long-term patient satisfaction and adherence to medical management. [19]

Psychosocial Issues and Support

Burden of a genetic diagnosis. Genetic information may sometimes be psychologically overwhelming, akin to a cancer diagnosis. The patient may display emotional responses through a variety of protective psychological mechanisms: denial, anger, guilt and shame for passing on a disease to children, despair, grief, and depression. The most important initial step for the provider is acceptance of the patient's responses and vulnerabilities. [20], [21] An environment of trust and openness can then be provided so that short- and long-term psychological support can be given. The genetic diagnosis frequently delineates risks that differ widely from the patient's beliefs or perceived risks. Challenges for the provider thus also include the reconciliation of unexpected news with long-held beliefs and ideas.

Decision to pursue genetic testing. The decision of whether or not to pursue genetic testing is often connected to the psychological issues. Several studies carried out before the implementation of clinical genetic testing noted high acceptance rates and an intention to pursue testing when it became available. [22], [23], [24], [25], [26] and [27] However, the acceptance of genetic testing since it became available has been lower than originally expected. [28], [29] Reasons for the low acceptance rate include costs, perceived lack of medical benefits, perceived risks, personal goals, ethnicity, gender, and fear of genetic discrimination. Each of these issues is inextricably intertwined with patient psychology in addition to the disease and risks at hand. [26], [30], [31], [32], [33], [34], [35] and [36]

Psychological assessment associated with genetic counseling. In view of the many psychological issues surrounding genetic diagnosis and testing, a thoughtful psychological assessment, usually informal but sometimes formal, should be included in pretest or posttest counseling. Pretest counseling prepares both patient and clinician for receiving the test results. Counseling should assess the patient's perception of genetic testing results, in addition to the psychological and social effects of testing on the patient and extended family members. The decision to test should complement the patient's personal and family goals, ethical and religious standards, and medical goals. In some cases, psychological intervention may require additional referrals to psychiatrists, clinical psychologists, support groups, or counseling groups.

Return of results. Because testing has implications not only for the testing candidate but also for extended family members, a discussion during pretest or posttest counseling regarding how to share test results with other family members or children at risk is appropriate. Results disclosure is usually presented in person to ensure a precise understanding of the test results, to provide appropriate support for the psychological consequences surrounding the results, and to outline individual surveillance or medical options that derive from the results. Disclosure of results by telephone or through a primary health care provider as an intermediary is generally not considered sufficient—again, akin to disclosing a cancer diagnosis in this way.

Reproductive Counseling Gastroenterologists caring for women who are either pregnant or of childbearing age and being treated for chronic GI disease must be familiar with the topic of reproductive counseling. A major topic in reproductive counseling is teratogens. A teratogen is defined as an environmental agent, such as a GI medication, that is capable of causing either structural or functional abnormalities in the developing fetus. In an assessment of potential risk to a pregnancy, an environmentally induced malformation depends on several measures. These may include the time of exposure during pregnancy, the dosage of the drug, the duration of exposure to the drug, individual susceptibility, the potential interaction with other environmental agents, placental transport, species differences, and pharmacokinetics and metabolism of the agent. [37] In counseling a pregnancy at risk for teratogenicity, it is important to compare the risk of 2% to 5% in the general population that any pregnancy will result in a major birth defect, regardless of family history and exposures, with the potential for increased risk based on currently known teratogenic exposure(s). If discontinued before pregnancy, most drugs generally pose no increased risk to the developing fetus. Moreover, the developmental period at which exposure occurs can be a predictor of the type of embryonic damage. Typically, medications taken during the preimplantation phase, or the first 2 weeks after conception, exhibit the all-or-none phenomenon. This should not be misinterpreted to mean that malformations cannot be induced at this stage. Rather, embryonic lethality resulting in an unrecognized pregnancy is more likely than a surviving fetus with anomalies. [37] The period of organogenesis (gestational day 18 through about day 60) is when most major and gross anatomic and functional development occurs, and therefore this is the time when fetal exposure to teratogens is most likely to result in severe malformations. During the second and third trimesters of pregnancy, damage to the genitourinary system and the brain is more likely. The Food and Drug Administration (FDA) has developed a rating system that combines what is known regarding the teratogenicity of a given drug and the potential benefit of that drug to a pregnant woman. *Physicians' Desk Reference* provides a similar rating scale. Table 54-9 lists some commonly prescribed GI medications and their FDA ratings for use in pregnancy. Additionally, several comprehensive and up-to-date resources provide detailed information on teratogenicity and pregnancy risk to clinicians (Table 54-10).

TABLE 54-9 Commonly Prescribed Gastrointestinal Medications and the Food and Drug Administration Use-in-Pregnancy Rating System and Lactation Studies

TABLE 54-10 Database Resources for Teratology Information

Developing Technology From our experience, a frequently asked question during genetic counseling pertains to gene therapy. Gene therapy involves introducing genetic material into human cells with the goal of achieving a preventative or therapeutic effect. [38] To date, gene therapy has had limited success. Gene therapy was initially considered useful, but has not been accomplished, for monogenic conditions such as cystic fibrosis and familial hypercholesterolemia. Gene therapy is now under investigation for use in treatment of more complex genetic and infectious diseases. The main challenges are finding safe, reliable and efficient vehicles through which genetic material can be transferred to the target cells in the body. An additional challenge involves having the replaced gene express itself in the target cells at the right time and quantity to be beneficial to the patient. Lists of clinical trials currently underway can be found at www4.od.nih.gov/oba/rac/clinicaltrial.htm and at www.wiley.co.uk/genetherapy.

GENETIC TESTING

Definition and Clinical Value

Genetic testing has recently become a part of the standard care for a number of GI conditions and is currently the most common topic of genetic counseling in the GI patient. *Present genetic testing* usually refers to testing for the presence of inherited conditions and involves analyzing DNA, RNA, chromosomes, proteins, or certain metabolites to detect changes related to a genetic disorder. GeneClinics is a federally funded and continually updated database of clinical and research laboratories that perform molecular analyses for inherited genetic diseases (http://www.genetests.org/). The discoveries of the Human Genome Project will inevitably lead to an increasing array of genetic tests for the diagnosis of both unusual and common GI disorders. Less commonly, genetic testing involves an analysis of specific tissue to assess for acquired or somatic mutations, such as the genetic testing performed as an adjunct in staging certain cancers. The information can be used to acquire a better understanding of the molecular and biologic nature of a particular disease so that treatment can be targeted. This type of genetic testing is not discussed here.

Classification of Approaches to Genetic Testing

Genetic testing falls into several classifications, discussed in the next sections. Although genetic testing is a powerful tool in guiding the clinical care of the at-risk or affected individual, attention should be given to its current limitations. Currently, the main limitation of genetic testing is that the sensitivity and specificity of testing methodologies for most conditions are not 100%. A negative or indeterminate result in an individual without a prior known family-specific mutation does not indicate the absence of a deleterious mutation for a suspected syndrome or the lack of a genetic etiology. Such misinterpretations are potentially fatal when medical management ceases for an at-risk individual. [39]

Diagnostic or Initial Genetic Testing Diagnostic testing is genetic testing performed in a person suspected of having an inherited disease, the mutation for which has not been sought or identified previously in relatives. An example would be a 40-year-old with ten adenomatous polyps but no known family history of FAP or attenuated FAP. Finding an *APC* mutation would confirm the diagnosis of FAP or attenuated FAP and would allow the clinician to proceed with appropriate screening and therapy. Failure to identify a mutation, however, is not helpful because mutations are not invariably found initially. Thus, the diagnosis remains unclear and may or may not be caused by an *APC* mutation. Clinicians are thus left to use their best clinical judgment.

Presymptomatic Testing or Testing When the Disease Mutation Is Known Knowledge of a specific disease mutation in a family allows family members at risk for the disease to undergo presymptomatic testing, the second classification of genetic testing. Presymptomatic testing involves testing healthy family members for the presence or absence of a specific mutation previously identified in an affected family member. A positive test result in a presymptomatic family member indicates that the person being tested carries the mutation and therefore has a genetic diagnosis of the disease in question. Appropriate screening and surveillance can then be planned. A negative test result (i.e., failure to find the mutation known to be segregating in the family) means that the person tested does not have the disease that is present in the family. Further screening or management of the disease in question is unnecessary for the person with a negative test result in this situation. A note of caution here is that a positive genetic diagnosis is not equivalent to a clinical diagnosis. Many features of the disease in question may or may not appear, depending on the observed penetrance of each of the features. This concept is frequently misunderstood by patients and requires careful attention. Retrospective study, for example, has shown that many patients thought a positive result of a test for FAP meant that they had colon cancer. Precise genetic counseling is thus extremely important at this stage. The patient must be educated about the likelihood of the development of certain disease features and must understand that appropriate management can prevent many of the consequences of the condition. A genetic diagnosis of FAP, for example, implies a 100% risk for eventual polyposis, but colon cancer can virtually always be prevented with appropriate therapy. A genetic diagnosis of HNPCC confers an 80% lifetime risk for colon cancer (not 100%), which likewise can almost always be prevented through appropriate screening.

Carrier Testing Carrier testing is performed for autosomal recessive and sex-linked conditions to learn if an individual is a heterozygous carrier of a gene mutation. Carrier testing may be appropriate for unaffected individuals with a relative who has an autosomal recessive or sex-linked condition. Additionally, members of different ethnic groups may be more likely to be gene carriers because of increased carrier frequencies in specific populations, and carrier testing is routinely offered to these groups (see section “ Genetic Screening for the General Population ”).

Prenatal Testing In prenatal testing, the health status of a fetus is evaluated during pregnancy. This may be desired because of an abnormal result of a screening test, such as ultrasonography, or because of an increased risk associated with the family history or maternal age. Fetal cells are obtained through procedures such as amniocentesis and chorionic villus sampling. Karyotyping to look for chromosomal disorders, or mutation analysis to look for a specific gene mutation known to be present in the family, may be applied to these cells. Many health, ethical, and social issues surround prenatal testing and require careful attention of the genetic counselor and the health team.

Newborn Screening In newborn screening, genetic testing is performed shortly after birth to learn if an infant is at an increased risk for having a specific genetic condition, the consequences of which could be prevented or ameliorated by immediate treatment. All states in the United States now require screening for several genetic conditions, most of which are metabolic (Table 54-11).

SCREENING TESTS	GASTROINTESTINALLY RELATED SYMPTOMS
Phenylketonuria	Neonatal vomiting
Galactosemia	Diarrhea, hepatomegaly, vomiting, progressive liver dysfunction leading to cirrhosis if untreated
Maple syrup urine disease	Pancreatitis, vomiting
Homocystinuria	Inguinal hernia, fatty changes in liver, pancreatitis
Sickle cell disease	Cholelithiasis, splenomegaly, splenic syndrome
Cystic fibrosis	Pancreatic insufficiency, meconium ileus in neonates, biliary cirrhosis, distal intestinal obstruction syndrome, rectal prolapse, adenocarcinoma of the ileum
Tyrosinemia (type I)	Hepatosplenomegaly, hepatic cirrhosis, hepatocellular carcinoma, pancreatic islet hypertrophy, ascites, jaundice, vomiting, paralytic ileus, diarrhea, melena

TABLE 54-11 Newborn and Prenatal Screening

Social Issues Surrounding Genetic Testing

The recent availability of genetic testing for many diseases adds a new dimension to clinical care and genetic counseling. Position statements on genetic testing have emerged and serve as valuable references to guide clinicians in routine and special situations. Additionally, issues surrounding genetic testing are being extensively debated by medical scientists, by legislative bodies, and in the courts.

Informed Consent The informed consent process is uniformly accepted by professional genetic societies as an integral part of genetic testing and should be an essential part of pretest counseling before any molecular testing is undertaken. [40], [41] This is a consensus opinion of the Task Force Committee on Informed Consent. Key elements of the informed consent include a description of the potential risks and benefits of genetic testing, current limitations of testing, and an elicitation of the patient's and family's expectations, concerns, and risk perceptions (Table 54-12). Guidelines are also available for genetic testing in special circumstances, such as presymptomatic testing in minors and potential adoptees. [42], [43] and [44]

The patient should have a working understanding of the following issues:
Medical Issues
• The syndrome in question, including inheritance, risks, screening guidelines, and disease management
• Medical care possibly better directed with genetic diagnosis
• Compliance with surveillance guidelines necessary for benefit
• Prevention possibly improved with knowledge and compliance
Genetic Issues
• Interpretation and implications of a positive, a negative (dependent on whether familial mutation is known), and an indeterminate genetic test result
• Risk to self and family members better defined if genetic testing successful
• Methods of testing and the accuracy associated with each testing option
• Alternatives to genetic testing
Psychological Issues
• Failure to detect a mutation in the first person tested in family: frustration, anxiety, disappointment, possible relief
• Positive test: coping with possible anger, anxiety, responsibility guilt, illness, self-image issues
• Negative test in a member of a family with a known mutation: possible relief, decreased worry for self and offspring, survivor guilt
Social/Economic Issues
• Failure to detect a mutation in the first person tested in family: no information for self, children, and other at-risk relatives
• Positive test: possible insurance, employment, and social discrimination
• Negative test in a member of a family with a known mutation: possible resolution of insurance problems, family relationships possibly positively or negatively affected
Special Issues for Testing in Children
• Protection of children's rights
• Self-image problems
• Possible problems with parent-child relationships, sibling-sibling relationships
• Social stigmatization
Other Issues
• Cost of test, insurance coverage, and genetic discrimination
• Confidentiality of results

TABLE 54-12 Informed Consent for Genetic Testing

Genetic Testing for Children Because of the potential for adverse psychological effects resulting from stigmatization and other harms, [20], [45] genetic testing in children should be considered only if medical benefit is clearly possible. [42], [46] FAP testing by the age of 10 to 12 years is one such example; screening and preventive measures are indicated by that age in carriers, whereas screening is unnecessary in noncarriers. [47] A parent's intense desire to be certain that a child does not have FAP at a younger age, however, may not justify the potential social stigmatization and self-image issues a child may have to faced if FAP is actually diagnosed. When the testing of minors is being considered, both the parents and health care professionals need to weigh the potential medical benefits against the psychosocial well-being of the child very carefully. The decision-making capacity of a child should also be considered, and whether advocacy for testing is truly on the behalf of the child's interests. Age-appropriate education for the child undergoing testing is also a necessity. Continuing education and follow-up as the child matures can focus on information relevant for life decision making.

Testing of Adopted Persons The American Society of Human Genetics (ASHG) recommends that the guidelines for the testing of adoptees (newborns and children) be consistent with those for genetic testing performed in all children of similar age (as discussed previously). Thus, the timing of genetic testing should be primarily justified for reasons such as the initiation of preventive or therapeutic medical management through early diagnosis. [44] Additionally, the ASHG recommends that a relevant family history and medical history for the adoptee be accessible and updated so that this information can be shared by all parties involved, including the adoptee, the biologic parents, and the adoptive parents when medically appropriate. Currently, however, only Wisconsin, mandates the collection of medical and genetic information on adoptees. [43]

Duty to Warn Relatives Recently, the duty to warn at-risk relatives about the ramifications of inherited disease has been heavily debated. [48] Two legal cases have concluded that at minimum, there appears to be at least a duty to warn the patient about the familial implications of genetic disease. In one of the cases, it was further declared that "reasonable steps be taken by the provider to assure that the information reaches those likely to be affected or is made available for their benefit" (*Pate v Threlkel,* 661 So 2d 278 [Fla 1995] and *Safer v Estate of Pack,* 677 A2d 1188 [NJ Super Ct App Div 1996]). The disclosure of information regarding oftentimes treatable and curable inherited disease to offspring and other relatives is still debated and unrefined. [49]

Who Should Be Tested First?

In a family believed to have an inherited syndrome, initial genetic testing to find the causative mutation should be offered to the person most likely to carry the mutant gene. In an FAP family, for example, genetic testing would start with a person clinically known to have the disease. This individual is considered the ideal "testing candidate." A negative test result in this case would be interpreted as a failure to find a disease-causing mutation in that person, and would simply mean that genetic testing cannot be used in the family and that other family members must be managed on the basis of the clinical findings. Another example would be a kindred meeting the Amsterdam criteria for HNPCC. The youngest person with colon cancer would be the testing candidate because that person would be most likely to carry the mutant gene. Further genetic testing in the family would be irrelevant if a mutation could not be found in that person, whereas finding a mutation would allow testing in other family members with nearly 100% accuracy. Thus, if a familial mutation is detected by genetic testing, a negative result in a member of this family is interpreted as a true negative—that is, this individual has not inherited the condition.

An important issue to keep in mind is that finding a mutation in the child of a living parent often indicates that a parent also has the mutation. This situation can be challenging when the parent has not consented to know his/her genetic status, has not learned about genetic testing, and possibly does not want such information at all. Assessing the family dynamics as well as encouraging family decision making regarding genetic testing should therefore be part of the pretest counseling, although ultimately genetic testing is the individual's choice.

Because most testing methodologies require a fresh blood sample, the first person tested must also be living. DNA banking is frequently considered when the death of an individual key to the genetic diagnosis in a family is imminent. DNA banking is the process of obtaining a biologic specimen, usually blood, from which DNA can be extracted. A laboratory will store the DNA for a certain length of time for a specified cost so that it will be available for genetic testing or testing that becomes available in the future. However, before the sample is drawn, informed consent for DNA banking is necessary outlining what it is permissible to do with the sample and who has authority over it after the death of the donor. After the death of the affected individual, individuals who have permission to access the sample may request genetic testing so that the information can be used by at-risk family members.

Regulatory Issues

Clinical genetic testing is presently controlled by specific regulations and oversights implemented to ensure that tests are performed properly. In the United States, the Clinical Laboratory Improvement Amendments of 1988 oversee all testing on human samples for health assessment or for the diagnosis, prevention, or treatment of

disease. [50] Genetic test results to be returned to the patient should be performed in a Clinical Laboratory Improvement Amendments–approved laboratory.

Impact of Genetic Testing on Clinical Management

Genetic testing results are frequently used to direct screening in a family to those who are gene carriers. Precise screening recommendations are emerging for many of the genetic conditions, in part because of the ability to determine accurately who has the inherited disease in question (see Chapter 90 for present examples).

Chemoprevention is an emerging approach that may eventually be of great benefit to those at increased risk for colon cancer. The use of celecoxib (Celebrex), a cyclooxygenase-2 inhibitor, to induce the regression of adenomatous polyps in FAP patients who still have a rectum after colectomy has been approved by the FDA. Aspirin, other nonsteroidal antiinflammatory drugs, folate, calcium, and estrogens have shown promise in decreasing the incidence of adenomatous polyps and colon cancer. Genetic testing may well direct how such therapies are applied by precisely defining the precancerous conditions.

The choice of surgery for colon cancer may be affected by genetic testing. A right hemicolectomy, for example, is appropriate for most cases of cecal cancer, whereas a subtotal colectomy is indicated if a person is known to have a mutation of HNPCC. Prophylactic colectomy is also being discussed in that condition, as prophylactic gastrectomy is for inherited gastric cancer that arises from mutations of the *CDH1* gene. [51]

Pharmacogenetics is the study of variations in drug uptake, binding, distribution, metabolism, and excretion based on inherited determinants. As information emerges, genetic testing may be a major factor in medication choice and dosing.

Genetic Screening for the General Population

General population screening is commonly encountered in prenatal diagnosis or newborn screening because early medical intervention prevents disease or reduces its severity. A number of state-supported screening programs for genetic diseases with GI involvement are outlined in Table 52-11; the panel of screening tests varies by state.

Presently, population-wide screening for most genetic diseases is not available or even indicated. Screening of the general population for a particular disease could be justified only in the case of a highly penetrant condition for which effective medical treatments were available. Founder mutations or a high prevalence of disease carriers supports population genetic screening for certain diseases, such as sickle cell anemia, in certain countries and ethnic groups. On the other hand, population screening for FAP is impractical in view of the cost of screening and the rarity of the condition. Population screening may become more applicable and attractive in the future, however, as high-prevalence genes that affect common disease susceptibility are elucidated.

SUMMARY AND CONCLUSIONS

The genetic revolution and its emerging implications to clinical medicine make it necessary for the gastroenterologist to have a basic understanding of genetics and to know when genetic testing is a part of disease management. Furthermore, the far-reaching effects of genetic diagnoses on patients require the physician not only to have a medical understanding of inherited disease but also a familiarity with the psychosocial issues surrounding genetic testing. Only then can proper education and advice be given to patients as inherited conditions and genetic testing are considered in the clinical setting.

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CHAPTER 55

David W. Crabb

APPROACH TO THE PATIENT WITH DRUG OR ALCOHOL DEPENDENCY

Americans commonly abuse drugs or alcohol; approximately 14 million Americans abuse alcohol, [1] tens of millions have tried cocaine and marijuana, a million may have tried anabolic steroids, and 700,000 are addicted to heroin. The results of urine tests to detect drug use are positive in 3% to 16% of preemployment screens. [2] Because patients deny drug use, its occurrence is nearly always underestimated, leading to significant clinical problems. Drugs have effects that simulate pathological conditions, and hospitalized users may undergo withdrawal syndromes. Patients with chronic abdominal pain are also at risk for becoming addicted to narcotics. [3] Therefore, it is important for physicians to be alert for occult alcohol and drug abuse, to understand the biology of drug dependence, to recognize personality disorders in patients susceptible to drug addiction, and to know how drug abuse can mimic and complicate other illnesses. It is also important to recognize that some prevention programs have been shown to be effective [4] and that drug and alcohol abuse is treatable, albeit with less success than could be hoped for. We have entered an era of pharmacotherapy for drug and alcohol abuse. [5] This chapter is concerned with the neurobiology and clinical presentations of traditional drugs of abuse, such as alcohol, sedative-hypnotics, narcotics, stimulants and hallucinogens, and cannabis, as well as tobacco, laxatives, diuretics, and anabolic steroids.

The three general responses to drugs of abuse are tolerance, dependence, and withdrawal. Most drugs produce tolerance with continued use, so that the abuser must increase the dose to obtain the desired effects. Three forms of tolerance are recognized. Pharmacokinetic tolerance results from the induction of drug-metabolizing enzymes, usually in the liver, and increased rates of drug metabolism. Pharmacodynamic tolerance denotes decreased activity of the drug in the central nervous system (CNS) and may result from decreased receptor number or action, as well as the plasticity of the nervous system. Tolerance may even develop in the setting of the first exposure of the nervous system to a drug, and this may contribute to addiction. Conditioned tolerance is the ability of individuals to perform better only in an environment in which they have previously used the drug. Conditioning also plays an important role in drug craving because environmental cues, which are stimuli that the abuser associates with the psychic effects of the drug, can trigger the emotional memory of drug use and craving for the drug. Responses to conditioned cues appear to persist much longer than the physical abstinence syndrome. Tolerance is often but not invariably accompanied by dependence. Physical dependence is an adaptive state manifested by intense physical disturbances that develop when the administration of the drug is suspended. Psychic dependence is a condition in which a drug produces a feeling of satisfaction and a psychic drive that requires periodic or continuous administration of the drug to produce pleasure or to avoid discomfort. [6] However, this dichotomy may not have a valid neurobiologic basis. If dependence involves neural systems such as the sympathetic outflow tracts, the withdrawal syndrome will be physiologically violent, with sympathetic overactivity. Dependence involving inhibitory systems may be followed by withdrawal seizures. However, if the dependent systems are those mediating pleasure and reward, withdrawal symptoms may be limited to depression, dysphoria, and drug craving. The current *Diagnostic and Statistical Manual of Mental Disorders,* 4th edition (DSM-IV) criteria for psychoactive substance dependence and abuse are presented in Table 55-1 and Table 55-2. [7] These definitions apply to any substance. They emphasize the behaviors leading to increased consumption of drugs or alcohol, as well as withdrawal and tolerance.

A maladaptive pattern of substance use, leading to clinically significant impairment or distress, as manifested by three (or more) of the following, occurring at any time in the same 12-month period:
1. Tolerance, as defined by either of the following:
   a. A need for markedly increased amounts of the substance to achieve intoxication or desired effect.
   b. Markedly diminished effect with continued use of the same amount of the substance.
2. Withdrawal, as manifested by either of the following:
   a. The characteristic withdrawal syndrome for the substance (refer to criteria A and B of the criteria sets for withdrawal from the specific substances given in DSM-IV).
   b. The same (or a closely related) substance is taken to relieve or avoid withdrawal symptoms.
3. The substance is often taken in larger amounts or over a longer period than was intended.
4. There is a persistent desire or unsuccessful efforts to cut down or control substance use.
5. A great deal of time is spent in activities necessary to obtain the substance (e.g., visiting multiple doctors or driving long distances), use the substance (e.g., chain-smoking), or recover from its effects.
6. Important social, occupational, or recreational activities are given up or reduced because of substance use.
7. The substance use is continued despite knowledge of having a persistent or recurrent physical or psychological problem that is likely to have been caused or exacerbated by the substance (e.g., current cocaine use despite recognition of cocaine-induced depression, or continued drinking despite recognition that an ulcer was made worse by alcohol consumption).

From American Psychiatric Association. Diagnostic and statistical manual of mental disorders, 4th ed. Washington, DC: American Psychiatric Association, 1994:181.

TABLE 55-1 Criteria for Psychoactive Substance Dependence

A. A maladaptive pattern of substance use leading to clinically significant impairment or distress, as manifested by one (or more) of the following occurring within a 12-month period:
   1. Recurrent substance use resulting in failure to fulfill major role obligations at work, school, or home (e.g., repeated absences or poor work performance related to substance use; substance-related absences, suspensions, or expulsions from school; neglect of children or household).
   2. Recurrent substance use in situations in which it is physically hazardous (e.g., driving an automobile or operating a machine while impaired by substance use).
   3. Recurrent substance-related legal problems (e.g., arrests for substance-related disorderly conduct).
   4. Continued substance use despite having persistent or recurrent social or interpersonal problems caused or exacerbated by the effects of the substance (e.g., arguments with spouse about consequences of intoxication, physical fights).
B. The symptoms have never met the criteria for substance dependence for this class of substance.

From American Psychiatric Association. Diagnostic and statistical manual of mental disorders, 4th ed. Washington, DC: American Psychiatric Association, 1994:182.

TABLE 55-2 Criteria for Substance Abuse

DRUGS OF ABUSE AND BRAIN REWARD SYSTEMS

The initial studies of the action of drugs of abuse focused on identifying the cellular receptors through which they, like most drugs, act. It is now the goal of drug abuse research to identify the neural systems and intracellular signaling pathways that are activated or inhibited by individual drugs. Drugs of abuse alter activity within the brain reward system, [8], [9] which consists of the mesoaccumbens system, which in turn is comprised of dopaminergic neurons of the ventral tegmental area (VTA) projecting to the nucleus accumbens (NAc) and the limbic system (Fig. 55-1). Simplistically, brain reward/pleasure is associated with increased activity of dopaminergic neurons in the VTA, which release dopamine in the NAc. Many actions of drugs of abuse are mediated by drug-induced increases in dopamine levels in the NAc. Cocaine and amphetamines increase dopamine levels by inhibiting dopamine uptake or by acting as dopamine agonists in the NAc. Indeed, dopamine transporter–deficient mice are insensitive to the motor-activating effects of cocaine. [10] Opiates, [11] barbiturates and benzodiazepines, [8] and cannabis [12] interact with inhibitory receptors (μ-opioid receptors, ?-aminobutyric acid [$GABA_A$] receptors, and tetrahydrocannabinol [THC] CB_1 receptors, respectively) and are presumed to inhibit pathways that tonically inhibit dopamine neurons in the VTA. This disinhibition results in increased dopamine release in the NAc. Similarly, phencyclidine (PCP) is an *N*-methyl-D-aspartate (NMDA) receptor antagonist and is presumed to stimulate dopamine release by blocking an NMDA-stimulated system that normally inhibits dopamine neurons in the VTA. Nicotine probably stimulates nicotinic acetylcholine receptors present on VTA neurons, leading to increased dopamine release in the NAc. [13] The action of ethanol is perhaps the most complex. It can stimulate inhibitory $GABA_A$ receptors (similar to sedative-hypnotics), inhibit excitatory NMDA receptors (similar to PCP), and stimulate inhibitory opioid pathways. [14] Thus, ethanol probably inhibits pathways that inhibit dopamine neurons in the VTA, leading to increased dopamine release in the NAc. It is important to note that the chronic abuse of these drugs also has long-term effects on these neurons, [11] and that these changes are important in the development of tolerance, withdrawal, and craving. Although the abuse potential of these drugs may depend on their shared ability to activate dopaminergic reward systems, their other pharmacological effects seen following acute and chronic use may differ, as is described in the following sections.

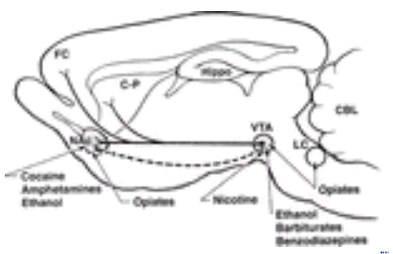


FIGURE 55-1. Anatomy of the mesoaccumbens reward system in the rat and probable sites of action of drugs of abuse. Dopaminergic pathways from the ventral tegmental area (*VTA*) are shown as *solid lines*; the pathway from the nucleus accumbens (*NAc*) is shown as a *dashed line*. The sites of drug action are inferred from neurochemical and intracranial self-administration studies. *CBL*, cerebellum; *C-P*, caudate-putamen; *FC*, frontal cortex; *Hippo*, hippocampus; *LC*, locus ceruleus. (Modified from ref. [8].)

PRINCIPLES OF TREATMENT

The treatment of drug and alcohol abuse revolves around several principles. [15], [16] First, abuse of multiple substances must be suspected so that the proper detoxification program is used. Detoxification is the separation of the patient from the drug(s) with the prevention of severe withdrawal syndromes (particularly those associated with alcohol, sedative, and opiate abuse). Detoxification is followed by counseling that attempts to generate insight and to teach the patient how to avoid high-risk situations predisposing to relapse. The goal of complete abstinence is central and is generally monitored by urine testing. A better monitoring test is a significant need in alcohol treatment. Pharmacological therapy includes naltrexone for alcoholism, opiate antagonists for opiate abuse, experimental treatments for cocaine abuse, and nicotine substitution for tobacco. [5] It is obvious, however, that the use of these drugs requires a motivated patient. These therapies are discussed in the following sections.

INDIVIDUAL DRUGS OF ABUSE

The actions of drugs of abuse are summarized in Table 55-3. Discussions of diagnosis and treatment are found in several books, [15], [16] and methods of detecting the drugs are described in the section “Diagnosis of Patients with Suspected Substance Abuse.”

Depressants	
Ethanol	Fluidize membranes (ethanol), induce cytochrome P450 (ethanol and barbiturates)
Barbiturates	Facilitate Cl^- conductance by effects on $GABA_A$-barbiturate-benzodiazepine-receptor-Cl^- channel
Benzodiazepines	Inhibit NMDA and voltage-sensitive calcium channels (ethanol)
Opiates	Interact with μ-opioid receptors, increasing K^+ conductance, reducing cAMP and Ca^{2+} influx
Cannabis	Interacts with cannabinoid receptor, reducing cAMP levels
Stimulants	
Cocaine	Blocks uptake of dopamine and norepinephrine
Amphetamines	Release norepinephrine, inhibit monoamine uptake, activate serotoninergic and dopaminergic receptors
Phencyclidine	Blocks dopamine uptake, increases dopamine release, anticholinergic, interacts with σ-opioid and NMDA receptors
Hallucinogens	Sympathomimetic, inhibit serotoninergic neurons
Nicotine	Activates ganglionic nicotinic receptors
Anabolic Steroids	Activate muscle and central nervous system (?) androgen receptors, increase growth hormone (?)
Laxatives	Stimulate intestinal motility and ion secretion, mechanisms not clearly established
Diuretics	Inhibit electrolyte reabsorption in renal tubules

GABA, γ-aminobutyric acid; NMDA, *N*-methyl-D-aspartate.

TABLE 55-3 Pharmacological Actions of Drugs of Abuse

Ethanol

Pharmacology The biologic actions of ethanol are remarkably diverse. Ethanol interacts with microdomains of neuronal proteins, especially ion channels. [14], [17] In particular, $GABA_A$ and NMDA receptor–associated ion channels are thought to mediate many psychomotor effects of ethanol. Ethanol at low concentrations (20–100 mmol/L) stimulates chloride uptake by isolated brain vesicles [17], [18] and augments the action of GABA at the benzodiazepine-barbiturate-$GABA_A$ receptor chloride channel. [14] Only a subset of $GABA_A$ receptors are so affected, possibly as a result of different receptor complex subunit compositions. [14], [17] This effect is blocked by Ro15-4513, a benzodiazepine receptor inverse agonist. Ro15-4513 also appears to antagonize certain behavioral effects of low-dose, but not high-dose, ethanol. [18] Flumazenil, a benzodiazepine antagonist, does not influence ethanol intoxication. Thus, ethanol may selectively enhance GABAergic transmission. This is consistent with the antianxiety effects of ethanol and the cross-tolerance between ethanol, barbiturates, and benzodiazepines. Tolerance at this receptor may result from membrane changes because the receptor number does not change. [19] Calcium channels are also important mediators of ethanol effects. The NMDA receptor is a glutamate-operated calcium channel. Low doses of ethanol inhibit NMDA-induced increases in neuronal intracellular calcium and resulting depolarization. [14], [20], [21] and [22] Ethanol also inhibits neuronal voltage-sensitive calcium channels. [23], [24] Tolerance to the effects of ethanol on both types of calcium channels develops in some brain regions with chronic use, possibly as a result of increased numbers of the channels. [24] Additional actions of ethanol include alterations of protein kinase activity (e.g., protein kinases A and C). Prolonged ethanol use leads to pharmacokinetic tolerance, in part by induction of the ethanol-metabolizing cytochrome P450 isoenzyme CYP2E1 and other cytochromes, [25], [26], [27] and [28] resulting in up to 30% faster ethanol metabolism. These cytochromes also metabolize a number of drugs

more rapidly in the chronic alcohol user [29]; those of greatest clinical relevance include warfarin, sulfonylureas, phenytoin, rifampin, diazepam and other hypnotics, and propranolol.

Clinical Features and Diagnosis The lifetime risk for alcohol-related problems is 20% to 25% for men and 5% for women; skid row, homeless alcoholics are in the minority. [1] The risk for alcoholism involves a genetic component, as has been demonstrated by adoption studies [30], [31], [32] and [33] and twin studies, [34], [35] whereas the inherited predisposition of some alcoholics to drinking problems is highly sensitive to environmental factors. Alcohol dehydrogenase and aldehyde dehydrogenase variants are the only genes that have been associated to date with an altered risk for alcoholism. [36], [37] and [38] Features of the alcohol dependence syndrome are listed in Table 55-4. [39] The most important diagnostic tool is a careful history elicited from the patient and family members regarding alcohol use and any related difficulties. This can be supplemented with the CAGE test, [40] a simple, nonthreatening list of questions; patients are asked if they have felt the need to *c*ut down on drinking, are *a*nnoyed by questions about their drinking behavior, feel *g*uilty about their drinking and its effects, and need an *e*ye opener (i.e., an early morning drink) to feel better. Two or more positive responses are highly suggestive of alcohol abuse, with a 93% sensitivity and 76% specificity. Increased values of the mean corpuscular volume, uric acid, triglycerides, or ?-glutamyl transpeptidase should suggest occult alcohol abuse, [41] but none of these tests is especially sensitive or specific for alcoholism. Two promising markers are acetaldehyde-protein adducts and carbohydrate-deficient transferrin. The adducts form in liver [42], [43] and blood [44], [45] by the condensation of acetaldehyde with proteins. Carbohydrate-deficient transferrin is formed in the liver during heavy drinking and can be detected immunologically or electrophoretically. [46] The test seems to be highly specific for alcohol consumption in excess of 60 g/d and has a sensitivity of about 90%. [46]

1. Narrowing of the drinking repertoire
   With increased severity of dependence, there is little variability of drinking pattern between drinking days and occasions.
2. Salience of drinking behavior
   Negative social, family, and/or health consequences will not deter drinking behavior. Highest priority is given to securing alcohol.
3. Subjective awareness of a compulsion to drink
   Severely dependent alcoholics are aware of an impaired capacity for moderate drinking. In the past, this characteristic was called "loss of control" drinking; in some cultures, it is characterized by an inability to abstain.
4. Tolerance
   With increased severity of dependence, and prior to the development of severe liver damage, alcoholics are less sensitive to the effects of alcohol.
5. Physical dependence
6. Relief avoidance drinking
   With increased severity of dependence, drinking occurs earlier in the day (e.g., upon awakening) in order to avoid the discomfort of "morning-after" symptoms.
7. Rapid reinstatement of the syndrome with recurrent drinking

From ref. 39.

TABLE 55-4 Alcohol Dependence Syndrome

Treatment The treatment of alcoholism requires detoxification on an inpatient basis [47] or on an outpatient basis in the absence of major medical illnesses or severe withdrawal symptoms. [48] The alcohol withdrawal syndrome is the consequence of adaptation of the neuronal membranes and ion channels to ethanol. During withdrawal, inhibitory GABAergic transmission may be reduced, and the excitatory effects of glutamate- and voltage-dependent calcium influx may be increased, [49] resulting in neural overactivity. In the first day or two of abstinence, tremor and tachycardia occur. Hallucinations are experienced by 10% to 20% of patients. Seizures occur somewhat later, and subsequently delirium tremens occurs in a minority of patients. This delirium is characterized by hallucinations and severe sympathetic overactivity, manifested by tachycardia, fever, dilated pupils, hypertension, and diaphoresis. The efficacy of barbiturates or benzodiazepines in preventing alcohol withdrawal symptoms is based on the augmentation of GABAergic transmission and neural inhibition. [50] Subsequent to detoxification, intensive counseling, behavior modification, and involvement with Alcoholics Anonymous and other support groups are helpful. The outcome of therapy may be better predicted by the characteristics of the patient than by the specific type of therapy. [1], [51] Individuals with intact support (i.e., those who are married, have a stable job, have no antecedent psychopathology, and are middle or upper class) have abstinence rates of up to 60% at 12 to 18 months after treatment, whereas unemployed individuals in the lower economic class have an abstinence rate of only 30%. Monitoring alcohol consumption with carbohydrate-deficient transferrin during treatment appears to be useful. [52] Pharmacological agents help maintain sobriety. [5], [53] Serotonin reuptake inhibitors (e.g., fluoxetine, zimeldine) are ineffective. [53] Alcohol-sensitizing drugs (e.g., disulfiram, cyanamide) are relatively ineffective. They act by inhibiting aldehyde dehydrogenase, so that acetaldehyde accumulates if ethanol is consumed, causing nausea, tachycardia, facial flushing, and hypotension. However, in the largest controlled trial, [54] disulfiram-treated, placebo disulfiram–treated (1-mg tablets), and control groups had nearly identical 1-year abstinence rates (approximately 20%). The oral opiate antagonist naltrexone reduces alcohol craving and relapse, [53], [55], [56] although it does not increase the rate of complete abstinence. [53] It is approved for this indication. The drug can cause hepatotoxicity and is contraindicated in patients with acute hepatitis. A number of studies in Europe have shown that acamprosate (calcium acetylhomotaurinate, a homologue of GABA) improves abstinence rates and duration of abstinence. [57], [58] Drugs on the horizon include nalmefene, [59] an opioid antagonist without the liver toxicity of naltrexone, and the 5-HT $_3$ receptor antagonist ondansetron, which may be particularly valuable in early-onset alcoholism, [60] as this subgroup has been suggested to have abnormalities of serotoninergic systems. Ondansetron may also have additive effects with naltrexone. [61] In alcoholics with coexistent psychiatric diagnoses, specific psychiatric and pyschopharmacological therapy is indicated.

Sedative-Hypnotics

Pharmacology Barbiturates and benzodiazepines interact with the GABA $_A$ receptor and augment the inhibitory action of GABA. [62], [63], [64] and [65] This explains the dangerous interaction between alcohol and these sedatives. Users of benzodiazepines develop tolerance to the sedative effects of the drugs but not to the anxiolytic effects. Barbiturates, but not benzodiazepines, induce hepatic microsomal enzymes and pharmacokinetic tolerance. [66]

Clinical Features and Treatment Barbiturates and benzodiazepines are used for sedation and muscle relaxation and to treat seizures, anxiety, and panic disorders. High doses of these drugs cause emotional lability, slurred speech, and ataxia. Rarely, the drugs induce delirium with incoherence, disorientation, and disturbances of perception. Benzodiazepines are ineffective in reducing pain, and some suggest that they may in fact lower the pain threshold. [67] They are therefore best avoided in patients with chronic pain. Dependence on barbiturates can develop after about a month of use at high doses; benzodiazepine dependence requires substantially longer use, probably about 20 weeks. Abrupt withdrawal from these drugs causes tremor, dysphoria, insomnia, hyperreflexia, anxiety, and, in severe cases, seizures. [66], [68] Barbiturate withdrawal in particular can be as dangerous as delirium tremens. Safe withdrawal can be achieved by reducing the dose of the barbiturate or benzodiazepine by about 10% per day. [15], [16] Persons who are tolerant of or dependent on benzodiazepines may experience seizures if they are given flumazenil. This drug does not, however, reverse sedation from or overdoses of barbiturates, ethanol, or opiates.

Opiates

Pharmacology Opiate abuse is probably as old as alcohol abuse. The Greek word *opion* means “juice of the poppy,” as does the Dutch word *doop,* the roots of the English words *opium* and *dope,* respectively. The effects of intravenous opiates include an initial intense euphoria that is followed by a dreamlike, tranquil state. Opiates act through μ receptors, which have a high affinity for morphine, other alkaloid opiates, and ß-endorphin. [8], [11], [69] Activation of μ receptors inhibits neural activity by increasing K^+ conductance and inhibiting adenylyl cyclase and Ca^{2+} channels through G-protein interactions (see Table 55-3). [8], [11], [70], [71] Chronic use of opiates does not reduce the number of neural μ receptors; rather, tolerance involves the increased expression of adenylyl cyclase and cAMP-dependent protein kinase A and the activation of slowly depolarizing Na^+ channels. [11] This has been best studied in the locus ceruleus, a sympathetic control center. [70] As a result, within 10 hours after discontinuation of a short-acting drug like heroin, the noradrenergic neurons of the locus ceruleus become hyperactive, leading to the autonomic features of opiate withdrawal. It appears that the cAMP-responsive element binding protein (CREB), a nuclear transcription factor, is a central participant in opiate dependence, as CREB knock-out mice have a greatly attenuated withdrawal syndrome. [72] Most opiates are metabolized in the liver. Morphine is glucuronidated; heroin is deacetylated to morphine and then glucuronidated. Meperidine has a significant first-pass metabolism of an oral dose and undergoes oxidation to meperidinic acid or *N*-demethylation to normeperidine. The latter is proconvulsant [73] and can accumulate in chronic renal failure. The half-life of meperidine is prolonged from 3 to 6 hours in cirrhosis. Codeine (3-methoxymorphine) undergoes less first-pass metabolism than other opiates, which accounts for its usefulness for oral administration. It is metabolized to morphine (10%) or norcodeine (10%) or is glucuronidated before renal excretion. Of the opiates used for the treatment of diarrhea, loperamide has the advantage of a very low incidence of CNS effects (including euphoria), in part because of its concentration in the gastrointestinal tract. A large fraction is excreted in the stool. Very high doses of loperamide have been used for severe diarrhea, possibly acting through mechanisms not involving opiate receptors. [74] Diphenoxylate can induce dependence at doses only five times the usual dose. For this reason, atropine was added to diphenoxylate (Lomotil) to reduce

the likelihood of abuse. The constipating effects of 45 mg of codeine, 5 mg of diphenoxylate, and 2 mg of loperamide are equal. Conversely, the opiate antagonist naloxone has been reported to increase fecal output in a patient with idiopathic constipation and intestinal pseudo-obstruction [75]; this has not been shown to help the majority of such patients. Oral naloxone may also be of use in treating the constipation caused by opiates; because it has a high rate of first-pass metabolism, it does not antagonize the central analgesic effects. [76]

Clinical Features and Diagnosis Heroin is the drug of choice of most narcotic addicts; however, prescription drugs like codeine, methadone, and oxycodone are also widely abused. Fentanyl is abused by medical personnel, and it and its illicit derivatives, such as 3-methylfentanyl, have such a high degree of potency that overdose is a serious risk. [77] Methylphenyltetrahydropyridine (MPTP) is a synthetic derivative of meperidine that destroys dopaminergic neurons in the substantia nigra. Its use caused an outbreak of toxic parkinsonism in the 1970s. [78] Tolerance to miosis or constipation does not develop, so these remain clues to opioid abuse. Requests for escalating doses of codeine for the control of diarrhea therefore indicate worsening of the underlying disease or abuse of the drug. Fully developed withdrawal symptoms result from overactivity of the sympathetic nervous system and include lacrimation, rhinorrhea, dilated pupils, piloerection (hence the term *cold turkey*), sweating, yawning, hypertension, tachycardia, and fever. Hallucinations, tremor, and delirium are not typical and suggest that the patient is also withdrawing from another drug.

Treatment The patient is first withdrawn from opiates. [15], [16] Withdrawal symptoms can be blocked by clonidine, which is given in 0.1- to 0.3-mg doses four times a day. [15], [16] This drug acts on presynaptic a_2-adrenergic receptors; like μ receptors, these are coupled by G proteins to K^+ channels and thereby inhibit neural activity in the locus ceruleus. [79], [80] Methadone can also be used to wean the patient from shorter-acting opiates. Relapse can be blocked by the chronic use of methadone or the antagonist naltrexone. Naltrexone can cause dose-dependent liver injury. [81] Unfortunately, patients maintained on these drugs often abuse other drugs, especially alcohol and cocaine. [82] Newer medications reported to reduce relapse include levomethadyl acetate (L-a-acetylmethadol, LAAM) and buprenorphine (a μ-opioid partial agonist). [83], [84] These drugs are longer-acting and may be given three times a week rather than daily, as is the case for methadone. Buprenorphine may be associated with a lower risk for physical dependence and have weaker opiate effects, and overdose is less likely.

Cocaine and Other Stimulants

Pharmacology Cocaine hydrochloride is used nasally (i.e., snorting) or intravenously. The base (“free base,” “crack”) is stable to heat. It is vaporized by heating it in a pipe and is absorbed from the pulmonary circulation, and it reaches the brain very rapidly. The neuronal effects of cocaine are biphasic. [85] It initially blocks the reuptake of dopamine, increasing its concentrations in the NAc [10], [86], [87] and activating both D_1 and D_2 receptors. [69], [71] Presynaptic feedback, probably involving dynorphins and opioid receptors, [10] subsequently reduces the release of dopamine. The late decrease in dopamine release may induce postcocaine dysphoria and facilitate craving. Cocaine is metabolized by hydrolytic and *N*-oxidative pathways. Hydrolysis inactivates the psychomotor effects of the drug and generates nonhepatotoxic intermediates, benzoylegconine and methoxyegconine. *N*-oxidative metabolism generates a series of *N*-hydroxyl and nitroxide intermediates. These intermediates are hepatotoxic. [88] However, cocaine hepatotoxicity [89], [90] is probably underrecognized owing to the high prevalence of viral hepatitis and alcohol abuse among these patients. Alcohol enhances cocaine hepatotoxicity by inducing cytochrome P450s of the *N*-oxidative pathway. [88] Amphetamines are active orally or intravenously. Derivatives used on the street include methamphetamine (“crystal”), methylenedioxyamphetamine (MDA), methylenedioxymethamphetamine (MDMA, “ecstasy”), para-methoxyamphetamine (PMA), methylenedioxyethylamphetamine (MDEA), and methyldimethoxyamphetamine (DOM). MDMA is a neurotoxin that may have delayed consequences. [91] Amphetamines release norepinephrine in the CNS and may activate dopamine receptors, and the hallucinogenic derivatives also activate serotonin receptors (see Table 55-3). [92] However, their major effect, like that of cocaine, is at the dopamine transporter, where they cause reverse transport of dopamine back into the synaptic cleft. Metabolism of amphetamines involves *p*-hydroxylation, *N*-demethylation, deamination, and conjugation. Because all the amphetamines are weak bases, their renal excretion is greatly increased by acidification of the urine.

Clinical Features and Diagnosis The effects of cocaine, amphetamines, and other stimulants are similar and result from sympathetic activation (pupillary dilation, tachycardia, and hypertension), [85] so that users are predisposed to cardiac arrhythmia or infarction, acute hypertensive crises, aortic dissection, myocarditis, and seizures. Necrotizing angiitis is associated with intravenous methamphetamine use. [93] Additional complications are intestinal ischemia, disseminated intravascular coagulation, rhabdomyolysis, and possibly perforating ulcers. [94], [95] Use during pregnancy can cause microcephaly, growth retardation, intrauterine cerebral infarction, and hemorrhage. [96], [97] and [98] Amphetamines can produce persistent hallucinations and delirium. The most common medical complication of intravenous cocaine abuse is the acquisition of hepatitis B or C.

Treatment Withdrawal from stimulants produces a mild physical abstinence syndrome characterized by depression, fatigue, disturbed sleep, and intense craving. Stimulant abusers are often detoxified as outpatients, and abstinence is documented by mandatory urine testing. The craving for cocaine is treated by intensive counseling and may be helped by pharmacological therapy. The tricyclics imipramine and desipramine, by blocking the reuptake of amine transmitters, may reduce dysphoria and improve sobriety, [99], [100] and [101] but controlled trials have yielded mixed results. Mazindol, [102] fluoxetine, [103] carbamazepine, [104] methylphenidate, lithium, [99] and bupropion [105] lack efficacy. Phenytoin [106] and amantadine [107] may help to maintain abstinence. The fact that cocaine abuse remains pervasive indicates that current therapy is inadequate. Auricular acupuncture has been reported to reduce cocaine use. [108] Another form of treatment is extinction therapy, in which the patient is allowed to watch videotapes depicting the use of cocaine or to handle drug paraphernalia without the reward of cocaine. It is hoped that this treatment will extinguish conditioned cues. Those with more severe problems, such as criminal involvement, may be treated for 6 to 12 months in therapeutic communities. Stimulant users commonly abuse other drugs with relaxing properties, such as heroin, alcohol, marijuana, barbiturates, and benzodiazepines, which complicates their treatment. Disulfiram has also been reported to reduce the use of cocaine by individuals treated with buprenorphine [109] or methadone [110] for combined heroin and cocaine addiction. It has been suggested that disulfiram inhibits dopamine ß-hydroxylase and reduces the conversion of dopamine to norepinephrine, thus maintaining neuronal dopamine.

Phencyclidine

Phencyclidine, or phenylcyclohexylpiperidine (“angel,” “angel dust,” “peace,” “zombie,” “supergrass rocket fuel,” “hog,” other names), can be taken by any route, including smoking. The pharmacological actions of PCP include potent activation of NMDA receptors, inhibition of dopamine reuptake (similar to the action of cocaine), stimulation of dopamine release, blockade of cholinergic receptors, and possibly interaction with opioid receptors (see Table 55-3). [111] It induces euphoria at low doses, and stimulates the sympathetic nervous system, hyperactivity, and hallucinations at higher doses. The hallucinations are frequently auditory, and the behavior of the abuser can closely resemble paranoid schizophrenia. [111] Intoxicated subjects have vertical and horizontal nystagmus, evidence of sympathetic overactivity, numbness and an increased pain threshold, ataxia, and dysarthria. The drug is hydroxylated and conjugated before excretion; its elimination is accelerated by acidifying the urine and by nasogastric suction. PCP is commonly misrepresented and sold on the street as lysergic acid diethylamide (LSD), mescaline, psilocybin, or THC. Other abused drugs that stimulate NMDA receptors include ketamine (used in pediatric and veterinary anesthesia and known on the street as “cat Valium,” “K,” “special K,” “super acid,” “vitamin K,” and other names) and dextromethorphan (DXM, “robo”). The latter is present in over-the-counter cough medications, so that the effects of high doses of these medications include the effects of the antihistamines and sympathomimetics as well.

Hallucinogens

Hallucinogens, including LSD, PCP, and amphetamine derivatives, are sympathomimetics, producing hypertension, pupillary dilation, tachycardia, and hyperreflexia as well as disturbances of perception. LSD acts presynaptically as a 5-HT_{1A} receptor agonist in the dorsal raphe nuclei to inhibit neural firing (see Table 55-3). LSD and hallucinogenic amphetamines are 5-HT_2 receptor agonists as well. [92] Users risk loss of control and experience recurrent drug-induced perceptions (flashbacks) and terrifying hallucinations (bad trips). Hallucinogens offer an interesting example of the dissociation of tolerance and dependence; tolerance appears to develop rapidly (after three to four doses) by unknown mechanisms, but there appears to be little physical or psychic dependence or craving.

Inhalants

Inhalants include a number of chemically distinct compounds that produce vapors that can be inhaled, resulting in CNS effects. Examples include solvents in paint and gasoline (toluene, benzene, methylene chloride, hexane), nitrous oxide (used as a propellant for canned whipped cream), and trichloroethylene (found in spot removers). Inhalants are often the first drugs abused by children, with as many as 6% of fourth graders trying these substances. The acute effects are similar to the CNS depressant effects of alcohol, including disorientation, slurred speech, incoordination, and irritability. A “sudden sniffing death syndrome” is recognized that may be the result of asphyxia. Longer-term toxicity includes demyelination, brain atrophy, toxic hepatitis, and renal dysfunction (including renal tubular acidosis). In addition, volatile nitrites, which cause vasodilation, are abused in the hope of enhancing sexual performance.

Club and Date Rape Drugs

These drugs are discussed together because of the circumstances of their use rather than their chemical similarity. Club drugs include MDMA, flunitrazepam (Rohypnol), ?-hydroxybutyrate (GHB), and ketamine. They are commonly used by teens and young adults at raves and trances (all-night dances). MDMA has been discussed in the section “Cocaine and Other Stimulants.” Rohypnol, GHB, and ketamine have largely CNS depressant effects. Rohypnol (“rophies,” “roofies,” “roach,” “rope”) is a benzodiazepine. Clonazepam, another benzodiazepine, is sometimes sold as Rohypnol. GHB (“liquid ecstasy,” “somatomax,” “scoop”) is abused for euphoric, sedative, and anabolic effects. It cannot be sold over the counter, but a related compound, ?-butyrolactone, can still be purchased. This substance is converted to GHB in the body. Each of these drugs is odorless, colorless, and tasteless. They can be added to beverages to sedate unsuspecting victims before sexual assault.

Cannabis

The active constituent of marijuana is ? ^{9}-THC. [112] THC binds to a membrane receptor [113] that is linked to G_{i} and G_{0} proteins; hence, stimulation of the receptor inhibits the formation of cAMP and has indirect effects on ion channels. [114] Regions of receptor density are found in the basal ganglia, hippocampus, cortex, and cerebellum. [115] The fatty acid derivative arachidonylethanolamide (anandamide) is an endogenous ligand for the THC receptor. [116], [117] THC causes euphoria, relaxation, subjective intensification of perception, alteration of the sense of time, and impaired psychomotor function. THC also causes vasodilation, which is responsible for tachycardia and conjunctival injection, and stimulation of appetite. Paradoxically, a rare cannabis arteritis has been reported in young men, manifested as distal arterial lesions and venous thrombosis, similar to Buerger disease. [118] The drug is metabolized by hydroxylation to the active compound 11-hydroxy-? ^{9}-THC and then to the inactive compound 8,11-dihydroxy-? ^{9}-THC. Cannabis dependence is recognized in the DSM-IV, [7] and withdrawal symptoms of restlessness, insomnia, and nausea have been observed. Users with a prior history of schizophrenia may be at increased risk for the precipitation of psychosis. Attacks of anxiety or paranoia occur in about 5% of episodes of cannabis use. Prolonged use of cannabis is reported to result in an amotivational syndrome characterized by preoccupation with drug use, passivity, and decreased drive and memory. [16] Treatment is based on counseling and must include suspicion of abuse of multiple drugs.

Tobacco

Pharmacology The major pharmacologically active compound in tobacco is, of course, nicotine, [119] which is thought to act on nicotinic cholinergic receptors in the mesoaccumbens system. This alkaloid is absorbed through the oral mucosa and the lungs. Smokers unconsciously regulate the amount of nicotine they absorb by altering how they smoke; therefore, nicotine intake is little changed by use of low-nicotine cigarettes or by a reduction in the number of cigarettes smoked. [119] Nicotine has a short half-life (1–2 hours) and is extensively metabolized in the liver by CYP2A6 to cotinine, which can be measured to monitor tobacco use. Nicotine increases sympathetic outflow, resulting in tachycardia, vasoconstriction (which can reduce the rate of absorption of subcutaneous insulin), hypertension, and mental arousal. Stimulation of Renshaw cells may inhibit anterior horn cells and cause muscle relaxation. Smoking also induces hepatic drug-metabolizing enzymes, resulting in increased rates of metabolism of imipramine, desmethyldiazepam, lidocaine, oxazepam, pentazocine, propranolol, and theophylline. The altered metabolic rate declines toward normal within 2 weeks of abstinence.

Clinical Features and Treatment Tobacco is surprisingly addictive. [120] It has been suggested that variants of the dopamine D_{2} receptor, the dopamine transporter, and CYP2A6 may contribute to the risk for addiction. Smoking during pregnancy is a common cause of premature delivery and low-birth-weight infants. Nicotine is concentrated in the tissues of the fetus, and the elevated levels of carbon monoxide in the blood of these women may reduce oxygen delivery to the fetus. Tolerance to nicotine, although partial, occurs very rapidly (within several hours), and a withdrawal syndrome consists of restlessness, irritability, anxiety, impatience, and inability to concentrate. These symptoms can be mitigated by clonidine. [121] Withdrawal from cigarettes is facilitated by alternative nicotine delivery systems. Controlled trials of nicotine replacement, used in combination with counseling, have shown an improvement in the rate of abstinence at 6 months (20%–30%) compared with placebo (10%–20%). [122], [123] Nicotine gum and patches are available over the counter. The transdermal systems cost about twice as much as the chewing gum, with similar response rates. Additional formulations of nicotine, including nasal spray and inhalers, are now available by prescription. The nicotine delivered by these systems should be given cautiously to patients sensitive to increased sympathetic tone (e.g., those with a recent myocardial infarction, cardiac arrhythmias, or angina pectoris), and use of these agents must be coupled with counseling. Cigarette smokers have perhaps a twofold increased risk for peptic ulcer, discussed in detail in Chapter 66. Aside from the use of nicotine replacement, other medications have been assessed for activity in helping patients to stop smoking. Systematic reviews indicate that nortriptyline and bupropion increased cessation rates. [124], [125] Anxiolytics have not been shown to be effective. Some data suggest that bupropion reduces the weight gain that often accompanies smoking cessation. [126] Women appear to have unique risks associated with smoking. More men have quit smoking during the past decades than women, so that the prevalence of smoking is now roughly equal between men and women. Furthermore, nicotine replacement does not seem to be as effective for women, and women are more likely to gain weight when they stop smoking.

Anabolic Steroids

Pharmacology The names of commonly used anabolic steroids are listed in Table 55-5. Although it is difficult to obtain controlled data, their use in combination with intensive weight training probably increases lean muscle mass and strength. They may induce an anabolic state by interacting with muscle androgen receptors, by antagonizing glucocorticoid receptors, or by inducing central effects of increased aggressiveness that makes training more effective. [127] Growth hormone secretion may be increased in some persons using the drugs.

Bolasterone	Methyltestosterone[†]
Boldenone*[‡]	Nandrolone[‡]
Clostebol	Norethandrolone
Dehydrochlormethyltestosterone	Oxandrolone[†]
Fluoxymesterone	Oxymesterone
Mesterolone	Oxymetholone[†]
Methandienone[†]	Stanozolol[†‡]
Methenolone[†]	Testosterone[‡]
Methandrostenolone[†‡]	Trenbolone*[‡]

*Veterinary preparation.
[†]Oral agent.
[‡]Used parenterally.

TABLE 55-5 Anabolic Steroids Used by Athletes

Clinical Features and Diagnosis Anabolic steroids are widely used among athletes to augment training, particularly in power sports such as weight lifting and football, [128] as well as among individuals who simply wish to improve their appearance. [129] Six percent of male high school students and 1% of female students acknowledge use of anabolic steroids. These individuals often share needles and may abuse other drugs. [129], [130] Typically, oral (C-17 alkylated steroids) and injected drugs are used together in cycles of 4 to 12 weeks during heavy training, at doses that are orders of magnitude higher than those used for medical indications. [131] The side effects of androgenic steroids have largely been reported for patients receiving long-term, low-dose therapy. Other side effects may be caused by intermittent high-dose administration. Among competitive power lifters assumed to have used anabolic steroids, increased premature mortality has been observed. [132] The liver may be affected by any of the C-17 alkyl androgens, [133] with minor, reversible elevations of alkaline phosphatase and transaminases and, rarely, jaundice. More serious problems include liver cell hyperplasia, liver cell adenoma, hepatoma, and angiosarcoma. [133] Interestingly, liver malignancies have regressed in some instances after the steroid was discontinued. Peliosis hepatis (blood-filled cysts communicating with the sinusoids) may complicate steroid use and can cause death from bleeding. Metabolic effects of androgens include increases in low-density-lipoprotein (LDL) cholesterol and decreases in high-density-lipoprotein (HDL) cholesterol, occasionally associated with myocardial infarction and stroke in steroid users. [134] Prostatic hypertrophy and, very rarely, prostatic carcinoma have been reported. Testicular atrophy and decreased spermatogenesis, although reversible, result from the suppression of gonadotropin secretion. Some athletes inject human chorionic gonadotropin (hCG) to prevent testicular atrophy. [135] Estradiol is increased by aromatization of the androgens, and gynecomastia occasionally occurs. Premature closure of the epiphyses may develop in adolescents, [136] and irreversible masculinization may develop in women. [137], [138] Lastly, the use of androgenic steroids may induce neuropsychiatric disturbances. Surveys have uncovered significant mood changes, psychotic episodes, and aggressive behavior during steroid use; the occurrence of dysphoria and depression after steroid use suggests addictive potential. However, under controlled conditions, the manic effects of testosterone affect only a small proportion of men. [139] Steroid abuse should be suspected because of the muscular development of the patient and should be considered in athletes with hypercholesterolemia, abnormal liver serum liver chemistries or hepatomegaly, and psychiatric disturbances. Needle marks are often visible on the thighs or buttocks of these patients and obviously are markers of a risk for hepatitis and human immunodeficiency virus (HIV) infection. Abusers of anabolic steroids commonly abuse other substances, such as stimulants (e.g., pseudoephedrine, phenylpropanolamine, ephedrine, clenbuterol [140]), diuretics (to help them lose weight), beta-blockers (to reduce hand tremor in accuracy competitions such as archery), and peptide hormones (e.g., growth hormone or erythropoietin), [135], [140] as well as other illicit drugs.

BULIMIA AND SUBSTANCE ABUSE

Eating disorders, such as anorexia nervosa and variants of binge eating (i.e., bulimia), are recognized to be quite prevalent. [141] These disorders are discussed in detail in Chapter 35 (for the diagnostic criteria, see Table 35-7 and Table 35-9). Bulimia patients commonly abuse other substances. About 61% abuse laxatives, 50% use over-the-counter diet pills, typically sympathomimetics, and 33% use diuretics for weight loss. [142] More than one third admit to alcohol or drug abuse. An eating disorder (and associated drug abuse) may be suspected in young women with the signs and symptoms listed in Table 35-8. [143] Both anorexic and bulimic patients are at increased risk for suicide. The therapy for these conditions centers on counseling. Placebo-controlled, double-blinded studies have demonstrated the effectiveness of tricyclic antidepressants [144], [145] and fluoxetine [146] for bulimia, whereas these drugs are of no proven benefit for anorexia. [147] Long-term follow-up of bulimic patients suggests that although many recover, a substantial fraction continue to have problems at 10 years. [148]

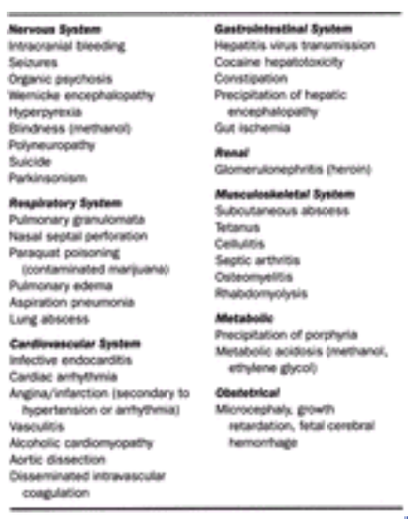
Laxative abusers are predominantly female, and the detection of laxative abuse should strongly suggest bulimia. [142] The laxatives may be taken to reduce distention after a binge or because of the perception that their use speeds weight loss. However, cathartics result in at most a 10% reduction in energy absorption. [149] The typical patient has multiple complaints, including weakness, nausea or vomiting, diarrhea or constipation, and abdominal pain. Cutaneous signs such as clubbing, skin eruptions from phenolphthalein use, and hyperpigmentation have also been observed. [150] Laxative abuse is one of the more common causes of chronic unexplained diarrhea referred to a university medical center study. [151] The diagnosis rests on detection of the laxative in the stool (alkalinization to detect phenolphthalein) or urine (bisacodyl, danthron, rhein, and phenolphthalein); methods are available to determine the latter by thin-layer chromatography. [152], [153] In some cases, a room search may be necessary. Although it is important to make this diagnosis to prevent unnecessary testing, long-term improvement in these patients is not often seen. Factitious diarrhea is also discussed in Chapter 42.

Other substances abused by bulimic patients include diuretics (confirmed by blood or urine testing for furosemide and thiazides), thyroid hormone preparations, and syrup of ipecac (emetine) to induce vomiting. The latter has been linked to the development of skeletal and cardiac myopathy. [154]

DIAGNOSIS OF PATIENTS WITH SUSPECTED SUBSTANCE ABUSE

The clinical features of drug use are summarized in Table 55-6. Medical complications can be seen with any injected drug and may be the first clue to substance abuse (Table 55-7). They include skin infections, tetanus, septicemia, infectious endocarditis (most commonly staphylococcal tricuspid endocarditis), venous thrombosis and pulmonary embolism, pulmonary granulomata, focal sclerosing glomerulonephritis, and acquisition of hepatitis B, C, or D or HIV infection. The threat of transmission of malaria may be the original reason why heroin is commonly mixed with quinine. [155] Illicit drugs may be adulterated with other compounds, resulting in additional toxicity, [155] or may not be the substance anticipated. Even the history of use of a particular drug must therefore be viewed skeptically and polydrug abuse assumed.

TABLE 55-6 Recognition of Drug Intoxication and Overdose



Nervous System
Intracranial bleeding
Seizures
Organic psychosis
Wernicke encephalopathy
Hyperpyrexia
Blindness (methanol)
Polyneuropathy
Suicide
Parkinsonism

Respiratory System
Pulmonary granulomata
Nasal septal perforation
Paraquat poisoning (contaminated marijuana)
Pulmonary edema
Aspiration pneumonia
Lung abscess

Cardiovascular System
Infective endocarditis
Cardiac arrhythmia
Angina/infarction (secondary to hypertension or arrhythmia)
Vasculitis
Alcoholic cardiomyopathy
Aortic dissection
Disseminated intravascular coagulation

Gastrointestinal System
Hepatitis virus transmission
Cocaine hepatotoxicity
Constipation
Precipitation of hepatic encephalopathy
Gut ischemia

Renal
Glomerulonephritis (heroin)

Musculoskeletal System
Subcutaneous abscess
Tetanus
Cellulitis
Septic arthritis
Osteomyelitis
Rhabdomyolysis

Metabolic
Precipitation of porphyria
Metabolic acidosis (methanol, ethylene glycol)

Obstetrical
Microcephaly, growth retardation, fetal cerebral hemorrhage

TABLE 55-7 Medical Complications of Drug Abuse

Alcohol use within 6 to 12 hours can be detected by analysis of breath, blood, or urine. Alcohol is eliminated at a rate of about 100 mg/kg per hour, but this rate varies substantially among individuals. There is a great need for a test for chronic alcohol abuse. Detection of the other drugs of abuse is complicated by variable amounts of drugs used and attempts by the user to avoid detection (e.g., by adulterating urine samples) (Table 55-8). Detection strategies can be divided into two phases, screening and confirmation. [156], [157] Routine toxicologic screening tests, usually based on thin-layer chromatography and used for patients suspected of taking a drug overdose, are the least sensitive. More sensitive screening tests are generally immunologic tests: radioimmunoassay, enzyme-multiplied immunoassay technique (EMIT), or fluorescence polarization immunoassays (see Table 55-8). These are widely available and are inexpensive. Positive screening tests are then confirmed, generally by gas chromatography–mass spectrometry (GC-MS), which is almost absolutely specific and extremely sensitive. [77], [158], [159] Establishing drug abuse also entails careful collection of the urine sample to prevent substitution or adulteration and attention to the chain of custody required to prove that a given patient has in fact been using the drug detected. Urine drug testing requires informed consent of persons older than 18 years of age except in medical emergencies.

1. Document quality of specimen: check temperature, color, pH, and specific gravity of urine to detect adulteration; check for presence of blood.
2. Document chain of custody.
3. Be aware of approximate duration of positive urine screening test results after drug use:
Alcohol: 6–12 hours.
Barbiturates and benzodiazepines: several days.
Opiates: 2–5 days.
Cocaine: 2–4 days.
Amphetamines: 2–4 days.
Marijuana: up to 30 days.
PCP: up to 8 days.
4. Be alert to false-positives:
Barbiturates and benzodiazepines: ibuprofen.
Opiates: poppy seeds (morphine), dextromethorphan.
Amphetamines: ephedrine, phenylpropanolamine, pseudoephedrine, other sympathomimetics.
Marijuana: ibuprofen, naproxen, fenoprofen (older screening tests), passive smoke.
5. Be alert to false-negatives:
Adulteration of sample: addition of acids, bases, benzalkonium chloride (Visine), soap.
Dilution of urine (includes use of diuretics for this purpose).
Short time elapsed after ingestion.
Intentional acidification of urine to speed elimination of amphetamine or PCP before test; alkalinization of urine to slow excretion during the testing period.
Ibuprofen may interfere with the derivatization step in GC-MS confirmation for marijuana.
Screening tests generally not available for LSD, mescaline, psilocybine, designer drugs.

GC-MS, gas chromatography-mass spectrometry; LSD, lysergic acid diethylamide; PCP, phencyclidine.

Compiled from refs. 2,77, and 158.

TABLE 55-8 Guidelines for Using Urinary Screening Tests for Drug Abuse

Benzodiazepines and barbiturates present few difficulties in identification. Tests for opiates may yield positive results for morphine if the patient has consumed poppy seeds; however, there can be no doubt of the origin of 6-monoacetylmorphine, a metabolite of heroin identifiable by GC-MS. Dextromethorphan, found in certain cough and cold medications, usually does not give a false-positive result at ordinary doses [158] and can be positively identified by GC-MS. [77] Fentanyl, methylfentanyl, and LSD may present problems of detection because they are so potent and are used at low doses. [77] Cocaine metabolites can be detected for up to 48 hours after use. The detection of amphetamines and PCP in screening tests is complicated by cross-reactions with over-the-counter medications. Several sympathomimetics can give positive results on amphetamine screening: ephedrine, pseudoephedrine, phenylpropanolamine, phentermine hydrochloride, fenfluramine hydrochloride, and others. [158] Both ? [9] -THC and PCP are lipophilic and can be detected in the urine for many days after use because they redistribute from fat stores. Phenylcyclohexyl pyridine (PHP), a compound related to PCP, can be synthesized from pyrrolidine instead of pyridine and cannot be detected by currently available urine screening assays. Occasional false-positive results have been observed for PCP screening tests when the patient was using the decongestant combinations Rondec or Dimetapp.

Anabolic steroid use can be documented by GC-MS and measurement of serum gonadotropins. Testosterone use is detected by finding increased ratios of testosterone to epitestosterone in urine. Epitestosterone is an endogenous steroid produced independently of testosterone. Surreptitious use of diuretics can be detected by measuring blood furosemide or urinary thiazide concentrations. [160]

MANAGEMENT OF ACUTE DRUG OVERDOSE OR TOXICITY

Table 55-6 summarizes the pharmacological effects of several classes of drugs of abuse. It is important to ensure that the apparently intoxicated individual does not have an additional medical problem, such as alcoholic hypoglycemia, head injury, meningitis, Wernicke or hepatic encephalopathy, seizures, or intracranial bleeding. [161] The overdose may have been accidental, as in the case of body packers who smuggle drugs by swallowing packages of the substances; if the packages do not pass spontaneously, surgical removal may be required. [162] Abused drugs are commonly adulterated with other compounds, such as quinine or lidocaine, but these are rarely the cause of acute toxicity. [155] Patients in coma need circulatory and respiratory support and care to avoid pressure complications. [161] A specific diagnosis should be made if possible, recalling that multiple drug abuse is the rule. Any suspicion of opiate overdose warrants a trial of 0.4 to 2 mg of naloxone intravenously, and benzodiazepine overdose responds to flumazenil; however, the latter drug may precipitate seizures. Specific antagonists are not otherwise available. Excretion of amphetamines and PCP is increased by acidifying the urine, achieved by infusion of ammonium chloride. Attempted suicide or homicide should be kept in mind. Seizures, agitation, or delirium caused by cocaine, stimulants, and PCP can be managed with diazepam or haloperidol and by reducing sensory stimulation.

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CHAPTER 56

Samuel L. Stanley, Jr.

ADVICE TO TRAVELERS

The advent of the jet age has revolutionized international travel. Persons now can be transported from one corner of the world to another in hours rather than days or weeks. Greater accessibility and lower prices for international travel have created a booming business, and it has been estimated that foreign travel and tourism now account for more than 25% of all world trade. [1] As more people travel, the importance of preventing disease has become increasingly recognized. The population at risk is significant. Annually, more than 25 to 30 million persons travel from nontropical regions into regions with malaria. [2] About 24 million persons from the United States alone travel into regions where hepatitis A is highly endemic each year. [3] In addition to infectious diseases, travelers may be at increased risk for a number of other problems, including jet lag, high-altitude sickness, excessive sun exposure, venous thrombophlebitis, and accidental death.

The goal of the physician is to minimize the traveler's risk for illness. To accomplish this goal, physicians must rely on educating travelers about specific risks and methods to reduce them, appropriate vaccinations and prophylactic medications, and the provision of medicines to treat certain illnesses that may arise during travel. The key to providing these services is the pretravel office visit, a visit that optimally should take place at least 6 weeks before the trip. At this visit, travel risks can be assessed, appropriate counseling offered, prophylactic medications dispensed, and vaccinations initiated. This chapter reviews the important components of the pretravel history, discusses noninfectious risks to travelers, provides a detailed description of the vaccination options available to travelers, discusses the problem of malaria prevention, describes current approaches to preventing travelers' diarrhea, and concludes with a brief synopsis of how to approach the problem of diarrhea in the returning traveler. Issues that may be of special interest to the gastroenterologist include the problem of prevention of travelers' diarrhea in persons with altered gastric acidity, the diagnostic approach to diarrhea in the returned traveler, and the use of vaccines and prophylactic medications in the immunocompromised patient (such as patients who have undergone liver transplantation).

PRETRAVEL HISTORY

The important components of the pretravel history are outlined in Table 56-1. A detailed itinerary represents a critical first step. The exact dates of travel, all countries to be traversed, and the length of stay in each country are the first components needed. Knowing the exact route of travel is important because even a layover at the airport in a country where yellow fever is endemic can entail a requirement for vaccination at subsequent destinations. The duration of the stay in a country is especially critical because the risk for contracting many of the infectious diseases (typhoid fever, hepatitis A) increases with longer periods of exposure. The planned itinerary of the traveler within each country is also important. In some regions of the world (e.g., much of Southeast Asia), malaria has been eliminated from major urban areas, but it remains a significant risk in rural areas. High-altitude destinations in otherwise malarious countries may be associated with no risk for malaria but present the potential problem of altitude sickness. Where ground travel is involved, even the timing of travel may be important; in countries where mosquito-borne illnesses are endemic, travel from dusk to dawn presents a higher risk for disease than do daytime excursions.

Detailed itinerary
Destination and route of travel
Duration of stay
Urban versus rural
Type of travel (e.g., business, tourist, Peace Corps, missionary)
Planned diet
Accommodations
Activities/work
Health profile
Immunization status
Underlying chronic diseases—immunocompromised?
Pregnancy status
Current medications

TABLE 56-1 Important Elements of the Pretravel History

Many important items fall under the rubric of the type of travel planned. The 1-week business trip involving accommodations at a four-star hotel in a major city in Southeast Asia presents significantly fewer health risks for a given traveler than the 1-year stay for the volunteer at a village hospital in sub-Saharan Africa. The traveler's planned activities should be established. Whether travel will involve primarily day trips to frequently visited tourist sites or will be a safari into the heart of the bush country directly affects the relative risks for disease. An expedition or job in which frequent contact with animals is expected is an indication for the rabies vaccine. The nature of the planned accommodations and how and where water and food will be obtained during the trip bear directly on the risk for insect-borne diseases and fecally-orally transmitted diseases, respectively.

Once the details of the trip have been established, a health profile of the traveler should be obtained. Important details include any underlying medical conditions that may create special problems during travel. Persons who are taking medications should ensure that they have an adequate supply for the duration of the trip, with an extra amount in case of loss or travel delays. Patients with chronic lung disease may have problems with hypoxemia in the reduced cabin pressure (similar to an altitude of 5000–8000 feet) of airplanes at maximum cruising altitude (30–40,000 feet above sea level). [4] In the case of persons whose arterial oxygen pressure is likely to be less than 50 mm Hg during the flight, supplemental oxygen can be obtained, but special arrangements with airlines must be made (usually at least 48 hours in advance). Although the leading cause of death in travelers from the United States to other countries is a cardiovascular event (49% of reported fatalities in one study), [5] travel for patients with stable heart disease is generally not medically contraindicated. However, persons with uncontrolled heart failure, unstable angina, or a recent (within 4 weeks) myocardial infarction should avoid travel if possible.

Travelers who have a pacemaker should have it evaluated before travel, and like all persons with cardiac disease, they should travel with a copy of their electrocardiogram. Because pacemakers may trigger metal detection devices at airports, travelers should carry a letter from their physician stating that they have a cardiac pacemaker, along with the model number and type of pacemaker.

Diabetic persons need to recognize that travel may disrupt routines in exercise, meals, and sleep, and hence change insulin requirements. Diabetic travelers should

carry a full supply of insulin and syringes adequate for longer than the expected duration of the trip as well as at least one bottle of regular insulin. Because customs officers of some countries may be suspicious of travelers carrying syringes and needles, they should carry a signed note from a physician stating that they require needles and syringes for the treatment of diabetes. Snacks should be carried at all times, and sugar cubes or candy for the management of hypoglycemia should be readily available. A travel companion who is educated about the symptoms and signs of hypoglycemia can be invaluable.

Women of reproductive age must be questioned about whether they are currently pregnant or are considering conception during the period of travel. Certain vaccines are contraindicated in pregnant women, as are some of the antibiotics used to treat or prevent travelers' diarrhea and some of the medications used to prevent chloroquine-resistant malaria. In addition, certain vacation activities, such as scuba diving and immersion in hot tubs, pose direct risks to the fetus and should be avoided. Plane travel is generally considered safe until week 36 of gestation, but pregnant women should be cautioned to move around the cabin whenever it is safe to do so to reduce the risk for thromboembolic disease associated with venous stasis. Finally, activities and destinations that take the pregnant traveler far from adequate emergency medical care should be avoided. Special recommendations for infants and children are beyond the scope of this chapter; the reader is referred to the article by Barnett and Chen for a review. [6]

Other persons also may require specialized recommendations. Immunocompromised individuals generally should not receive vaccines containing live viruses, and certain countries may restrict the entry of persons infected with the human immunodeficiency virus (HIV). Patients with gastrointestinal disease whose gastric acidity is reduced as a result of either gastric surgery or treatment with histamine H_2 blockers or antacids are more susceptible to enteric pathogens and may be candidates for prophylactic antibiotics to prevent infection. The physician needs to be aware that persons with malabsorptive states may require dosage adjustments or medication alternatives for commonly used drugs.

GENERAL PRECAUTIONS

Water

With few exceptions, water in developing countries should be considered contaminated and should not be used for drinking or brushing teeth or consumed in the form of ice. Locally bottled water also should be considered suspect, as should iced tea, and unpasteurized dairy products should be strictly avoided. Alcohol should not be relied on to sterilize the water in ice cubes or nonalcohol components in mixed drinks. Safe beverages generally include brand name carbonated drinks, wine, and hot coffee or hot tea that has been prepared with boiling water. It has been said that there is no such thing as a bad bottle of beer, and this appears to be true worldwide. Wet cans and bottles should be wiped clean and dried before use. When water is the only available beverage, boiling represents the best method for sterilization. Because most organisms are killed at 100°C, simply bringing water to a boil is adequate for sterilization. [7], [8] Chemical sterilization of water with halogens (chlorine or iodine) is less effective but can be used when boiling is impossible. A standard protocol would be 0.1 mL (about 2 drops) of 5% chlorine bleach in a liter of water, or 0.2 mL (about 4 drops) of 2% tincture of iodine in a liter of water. The water should set for at least 30 minutes at room temperature before it is used. Halogenation is less effective in cold or turbid water, and iodine should be avoided in persons with known iodine allergy, patients with unstable thyroid disease, and pregnant women. Commercially produced kits that use iodine resins and filters to purify water are available.

Food

Food can be the source of enteric pathogens, tissue parasites, and microbial toxins. The old adage "Boil it, peel it, cook it, or forget it" should be the traveler's mantra when confronted with dietary choices in developing countries. Food should be well cooked and served hot to avoid the possibility of bacterial growth from contamination after food preparation. Raw foods should be avoided, and the many diseases associated with shellfish (e.g., cholera, hepatitis A) and the fact that shellfish often are consumed uncooked or poorly cooked make them particularly risky for the traveler. [9], [10] Fruits, such as bananas and oranges, that can be peeled are safe, but uncooked fresh vegetables should not be consumed. Vegetables with a high water content, such as lettuce, are particularly risky, and salads probably are a common source of enteric pathogens. Ciguatera fish poisoning is a threat from fish caught in Caribbean and Indo-Pacific waters in tropical latitudes. Whereas the source of the food (expensive restaurants, private homes) does not guarantee safety, consuming food from street vendors is most kindly described as adventurous and more accurately described as foolhardy. In one study of students in Mexico, persons who purchased food from street vendors were three times more likely to acquire diarrhea than those who had never frequented street vendors. [11]

Accidents

Although much of the emphasis in ensuring safe travel focuses on infectious diseases, accidents represent the most common cause of travel deaths in persons under 40 years of age and the second most common cause of death overall. [5] In a study of U.S. travelers to Mexico, half of 396 deaths were injury related. [12] It should be emphasized that in both studies, the proportion of death resulting from injury was significantly higher in the group of travelers than in all age groups within the United States. Travelers need to be counseled about this increased risk for accidental injury and death and reminded that travel is not an excuse to forget the need to drive all motor vehicles carefully, adhere to the rules for water safety, and the danger of mixing activities with alcohol consumption.

Jet Travel

Travelers should be reminded to keep well hydrated and to move about the airplane cabin when it is safe to do so to reduce the risk for venous stasis and thromboembolic disease. Although not life-threatening, the disruption of sleep and waking cycles that occurs with traversing multiple time zones can be a significant problem for the business traveler and the tourist. When possible, some adjustment of the sleep cycle and timing of meals to the time zone of the travel destination before departure may be useful in mitigating jet lag. Short-acting benzodiazepines (e.g., triazolam) can be used to facilitate sleep during long (>6 hours) plane flights and to induce sleep at the appropriate bedtime hours after arrival. These agents have potential side effects that may be worse in elderly patients. Retrograde amnesia has been reported and may be associated with concurrent alcohol consumption. [13]

High Altitude

Travelers who find themselves at an altitude of 3000 m above sea level or higher are at risk for high-altitude illness (mountain sickness). If the ascent is gradual, the body can acclimatize, and symptoms may be minimal or absent. When the ascent is rapid or immediate (as in the case of the traveler who flies into a high-altitude city), symptoms of headache, fatigue, mild dyspnea, and insomnia are more likely to develop. With further ascent, life-threatening conditions, such as high-altitude pulmonary edema and high-altitude cerebral edema, can develop. The key to prevention is gradual ascent (300–500 m/d), acclimatization at each new altitude, and descent to lower elevations when symptoms arise. Acetazolamide, an inhibitor of carbonic anhydrase with some diuretic action, appears to be somewhat effective in preventing mild to moderate cases of high-altitude illness. The drug should be taken at a dose of 125 to 250 mg three times a day beginning 24 hours before the ascent and continued for 48 to 72 hours after the high elevation has been reached. Persons who are allergic to sulfa should not take acetazolamide.

Sunstroke/Sunburn

Sun and specifically ultraviolet radiation exposure may be particularly intense at high altitudes and at tropical latitudes, and the typical "sun and surf" or "sun and ski" vacation puts travelers at risk for sunburn. Hats and sunglasses are desirable, and sunscreens should be mandatory for travelers. The sunscreen should be formulated to prevent damage from ultraviolet B radiation (mainly responsible for sunburn) and ultraviolet A radiation (mainly responsible for photoallergic reactions), and it should have a sun protection factor (SPF) of at least 15. Overexertion by travelers unaccustomed to the intense heat and humidity of tropical climates can also put them at risk for sunstroke. A reminder about these dangers during the pretravel visit is important.

Insects

Some of the most dangerous infectious diseases worldwide are transmitted by insect vectors, including malaria, yellow fever, sleeping sickness, and rickettsial infections. In addition, insect contact can result in reactions ranging from local skin lesions to anaphylaxis and death. The risk for insect contact can be greatly reduced by wearing appropriate clothing, using bed netting, reducing nighttime exposures, and judiciously applying insecticides and insect repellents. Clothing that is light-colored and that covers as much of the body as possible is desirable (long-sleeved shirts, pants tucked into socks and boots); for maximum resistance, clothing should be treated with a permethrin-containing insect spray. Permethrin repels and kills ticks, mosquitoes, and chiggers, and permethrin-impregnated bed netting represents a significant barrier to mosquitoes. To protect exposed areas of skin, an insect repellant containing *N,N*-dimethyl- *m*-toluamide (DEET) should be used. Because of the risk for systemic toxicity from skin absorption, formulations containing high concentrations of DEET should not be used by infants, children, pregnant

women, and persons with desquamating skin conditions. [14]

Sexually Transmitted Diseases

A number of studies indicate that travel increases a person's risk for sexually transmitted diseases (STDs). [15], [16] and [17] People away from home may feel free from everyday restraints and engage in activities that they would not consider under normal circumstances. Long-term residents or business travelers in foreign countries appear to have a high rate of sexual involvement with native peoples. [18] Because STD rates are higher in many developing countries, travelers can put themselves at risk for a variety of diseases, including HIV infection, hepatitis B, syphilis, gonorrhea, chancroid, herpes simplex, and granuloma inguinale. The widespread heterosexual transmission of HIV in sub-Saharan Africa, areas of South America, and many countries in Asia has led to a high prevalence of HIV infection in the general population and an extraordinarily high HIV prevalence among commercial sex workers. [19] Travelers need to be counseled that abstinence is the only absolutely sure way to avoid STD. If abstinence is not considered an option, travelers should be informed that the proper use of condoms can decrease the risk for acquiring STD. Because the failure rates of latex condoms made in the United States appear to be lower than those of condoms manufactured abroad, [18] travelers should be advised to take along an adequate supply of condoms for their trip.

VACCINATIONS

Vaccines for the traveler can be divided into three major categories. The first includes vaccines required for entry into certain countries. The only vaccine that can be required by countries as a prerequisite for entry under the International Health Regulations adopted by the World Health Organization is the yellow fever vaccine. In response to disease outbreaks or special circumstances, individual countries occasionally adopt different rules, and the traveler always is cautioned to contact an authority such as the Centers for Disease Control and Prevention (CDC) on their hotline (404-332-4555) or the embassy or consulate of the destination country before travel. The second category comprises vaccines that are not required for entry into a country but are recommended because of specific health risks within that area. Examples in this category would be the typhoid fever vaccine and the vaccine to prevent Japanese encephalitis. The final category is made up of vaccines that are not specific for travelers but should be administered if the traveler's vaccination status is not up to date before the trip. An example of this latter category would be vaccination against tetanus. The detailed immunization history obtained in the pretravel visit may reveal gaps in vaccine coverage, and the physician should take advantage of this opportunity to provide all necessary vaccines for the traveler. Vaccine dosing regimens are outlined in Table 56-2.

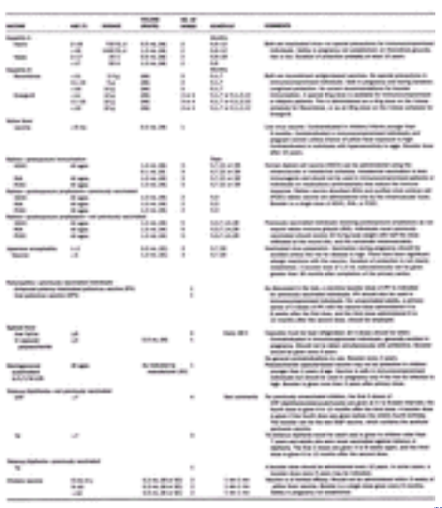

TABLE 56-2 Immunization Schedules for Vaccines That May Be Administered to International Travelers

Required Vaccines

Yellow Fever Yellow fever is caused by a small RNA virus of the flavivirus family and is transmitted by the bite of *Aedes* and *Haemagogus* mosquitoes. It is confined to countries in tropical South America and Africa and is not found in Asia (Table 56-3). Epidemics within South America and Africa continue to occur, and it is estimated that hundreds of thousands of persons in these areas may be infected yearly. [20] Clinical manifestations range from a mild febrile illness to a fulminant, sometimes fatal disease with hepatic, renal, and myocardial damage. Fortunately, the number of cases of yellow fever reported in travelers has been extraordinarily low, but this may, in part, reflect widespread use of the yellow fever vaccine in this population. Prevention of the disease is based on avoiding mosquito bites and provision of the yellow fever vaccine. Yellow fever vaccine is composed of attenuated live yellow fever virus (strain 17D) and is estimated to provide protection in more than 95% of vaccinated persons. Protective immunity lasts for at least 10 years after the single-dose vaccination. The vaccine is extremely safe, and side effects of vaccination are unusual. The most serious complication reported has been encephalitis (fewer than 20 cases), which was seen almost exclusively in infants younger than 4 months of age. [21] For this reason, the vaccine is not administered to children younger than 6 months of age, and vaccination usually is deferred until they are at least 9 months old. Persons with egg allergies should not receive the vaccine, and its use should be avoided in pregnant women unless they are felt to be at high risk for disease. In this case, the known benefit of vaccination would outweigh a potential (but undocumented) risk to the fetus. It is preferable that immunocompromised patients not receive the yellow fever vaccine because of the theoretical risk for vaccine-induced encephalitis. In the case of persons who wish to travel to a country where yellow fever is endemic, but whose travel plans put them at low risk for yellow fever, a medical waiver of vaccination form can be provided to satisfy entry requirements. When the immunocompromised person must travel to an area of endemicity where the risk for yellow fever transmission is high, it is probably prudent to administer the vaccine, but the potential risks and benefits should be discussed in detail with the traveler.

COUNTRIES IN AFRICA	COUNTRIES IN SOUTH AMERICA
Angola	Bolivia
Burkina Faso	Brazil
Cameroon	Colombia
Democratic Republic of Congo	Ecuador
Gabon	French Guiana
Gambia	Peru
Ghana	Venezuela
Guinea	
Liberia	
Sierra Leone	
Sudan	
Zambia	

Adapted from Centers for Disease Control and Prevention. Health information for international travel 1999–2000. Atlanta, GA: Department of Health and Human Services, 1999.

TABLE 56-3 Countries Where Yellow Fever Is Endemic

Recommended Vaccines

Hepatitis A Hepatitis A is an enterically transmitted viral disease that is highly endemic in most of the developing world (Fig. 56-1). Travelers from developed countries to these regions of endemicity are at significant risk for the development of hepatitis A. Symptomatic hepatitis A is the most common serious illness in travelers and is probably the most frequent medical reason for a premature return to the United States by persons who had planned a long stay in a developing country. [22], [23] Published attack rates of 0.1% to 1% for unprotected travelers contracting hepatitis A during a 2- to 3-week stay in an area of endemicity, [24] and the development of symptomatic disease in 1 in 300 travelers during a 1-month stay, illustrate the risk. [25] Hepatitis A virus is transmitted by the fecal-oral route and can be spread through person-to-person contact or by the consumption of fecally contaminated water and food. The prevention of hepatitis A virus infection is based on the avoidance of potentially contaminated food and water and the administration of the hepatitis A vaccine. Both currently available preparations are purified, formalin-inactivated, whole-virus vaccines that appear to be highly effective in preventing symptomatic hepatitis A. In a prospective study undertaken in Thailand, 40,000 primary school children were randomized to receive either the hepatitis A vaccine or the recombinant hepatitis B vaccine. The protective efficacy of the hepatitis A vaccine in preventing symptomatic hepatitis A was 95%. [26] A second inactivated hepatitis A vaccine showed 100% efficacy in preventing symptomatic

hepatitis A in a randomized, placebo-controlled trial involving 1000 children in an upstate New York community with frequent outbreaks of hepatitis A. [27] A protective antibody response probably is obtained within 2 or 3 weeks after the initial dose of either vaccine. [3] Firm evidence regarding the total duration of protection after completion of the two- or three-dose vaccine regimen is not available, but based on the observed rate of decrease in antibody levels, protective immunity may persist for 10 years or longer. [28] The vaccines are well tolerated; mild tenderness at the injection site is the most frequently reported side effect.

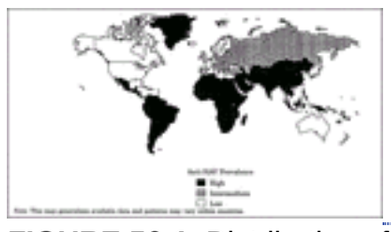

FIGURE 56-1. Distribution of hepatitis A virus prevalence. The prevalence of antibodies to hepatitis A virus (*anti-HAV*) in populations within countries is shown. (From Centers for Disease Control and Prevention. Health Information for International Travel 1999–2000. Atlanta, GA: Department of Health and Human Services, 2000.)

Because of its efficacy and excellent safety profile, inactivated hepatitis A vaccine probably should be administered to all travelers to areas where hepatitis A is endemic, including the Caribbean and Mexico. The role of passive immunization with immune serum globulin (administered as a single dose of 0.02 mL/kg) in preventing hepatitis A infection is now limited to providing protection for persons who have not received the vaccine and will be entering areas of endemicity before protective levels of antibody can develop after vaccination (2 to 3 weeks after the initial dose of vaccine). In this instance, both the hepatitis A vaccine and immune serum globulin can be administered into separate sites in the deltoids, and although the ultimate titer of anti–hepatitis A virus antibodies obtained may be lower than that obtained with vaccination alone, it still appears to be adequate for protection. [29]

Hepatitis B The prevalence of hepatitis B infection in the population of many developing countries is in excess of 8%. The virus is transmitted by contact with infected blood or through sexual activity. Risk factors for blood transmission are working in the health care field, any exposure to needles (intravenous drug use, tattooing), and receiving a transfusion of blood not screened for hepatitis B virus. For most travelers, the risk for hepatitis B is relatively low, but the following persons should be vaccinated: anyone working in a health care–related area who has not been vaccinated previously, anyone contemplating a stay of longer than 6 months in an area where the disease is highly endemic, anyone who may be sexually active with members of the local population in an area of endemicity, and anyone who has a health condition and may require hospitalization in an area of endemicity. The preparations available in the United States are recombinant vaccines consisting of highly purified viral surface antigens expressed in yeast. Vaccination requires three or four doses, with the final dose scheduled for 6 months or 1 year after the primary dose. Optimal protection is not achieved until after the final (third or fourth) dose; for travelers who cannot complete the full schedule before departing, however, it is worthwhile to initiate the vaccine series because some protection is conferred after the first and second doses. The vaccine appears to be highly effective, with more than 90% of vaccinated persons acquiring high anti–hepatitis B surface antigen antibody titers after the third vaccine dose, and 80% to 95% of persons protected against symptomatic hepatitis B based on the findings of multiple double-blinded, placebo-controlled clinical trials of the vaccine. [30], [31], [32] and [33] For persons who obtained initial antibody titers above 10 MIU/mL, complete protection against symptomatic infection was provided during a 10-year follow-up period. The overall duration of protective immunity is not yet known, but it is believed to be long, [34] and there are no current solid recommendations for boosters after completion of the primary three- or four-dose vaccine series. Side effects of vaccination are rare, and the vaccine is safe for use in immunocompromised persons, who may have defects in antibody responses after immunization; therefore, it may be worthwhile to check anti–hepatitis B surface antigen antibody titers after completion of the vaccine series to determine whether protective levels have been achieved.

Rabies Rabies remains a significant health problem in many areas of the world, and it is conservatively estimated that 35,000 rabies deaths occur annually. [35] The greatest risk for travelers is in areas where canine rabies remains highly endemic, which includes the Indian subcontinent, Nepal, the Philippines, Thailand, Sri Lanka, Vietnam, Guatemala, Peru, Colombia, Ecuador, El Salvador, and parts of Mexico. Canine rabies also can be found in many other countries in South America and Asia and in much of Africa. Travelers who will be staying for more than 30 days in an area where rabies is a significant threat should receive preexposure vaccination with the human diploid cell rabies vaccine (HDCV), purified chick embryo cell vaccine (PCEC), or rabies vaccine adsorbed (RVA). [36] Preexposure vaccination may provide protection in cases of unrecognized exposure to rabies virus or delayed postexposure prophylaxis. It must be emphasized to the traveler that preexposure vaccination does not eliminate the need for additional therapy after a rabies exposure, but it does reduce the number of postexposure vaccine doses and obviates the need for rabies immune globulin (RIG). The HDCV, PCEC, and RVA vaccines are inactivated vaccines; hence, they are not contraindicated for immunocompromised individuals or pregnant women at risk. In immunocompromised persons, neutralizing antibody titers should be determined 2 weeks after the final dose; if they are inadequate (complete neutralization in the rapid fluorescent focus inhibition test at a minimum of a 1:25 dilution), another booster dose should be given. [36] Chloroquine, and possibly other antimalarials being taken for antimalarial prophylaxis, may reduce the antibody response to the rabies vaccine, and the more immunogenic intramuscular administration schedule of the vaccine should be used when the patient is taking drugs that can interfere with the immune response. Side effects noted with the rabies vaccine include pain and erythema at the injection site and mild systemic reactions such as fever, nausea, abdominal pain, muscle aches, and dizziness. An immune complex–mediated reaction, with urticaria, pruritus, fatigue, and sometimes angioedema, has developed in a few persons receiving booster vaccinations with rabies vaccine. [37]

Japanese Encephalitis Japanese encephalitis is a mosquito-borne flavivirus infection that occurs in Japan, China, Korea, India, and much of Southeast Asia. The disease is transmitted by *Culex* mosquitoes, and the highest prevalence is highest in rural areas, although transmission in cities has been reported. In countries in temperate regions, a marked seasonal variation is noted, with most cases seen in the summer. In tropical latitudes, cases occur throughout the year. Most infections probably do not result in symptoms, but in the estimated 1 in 300 infected persons in whom encephalitis develops, the case fatality rate approaches 25%. [38] From 1978 to 1992, 24 cases of Japanese encephalitis in travelers (including 11 Americans) visiting or residing in Asia were reported. [38] The prevention of Japanese encephalitis is based on taking measures to prevent mosquito bites and, for some persons, administering the inactivated Japanese encephalitis virus vaccine in a three-dose regimen. The vaccine licensed in the United States appears to be about 90% effective in preventing disease, but it has been associated with a significant number of allergic reactions. Urticaria, angioedema, and anaphylaxis appear to occur at a rate of between 15 and 62 per 10,000 vaccine doses, and persons who have a history of severe allergic reactions to other allergens appear to be at higher risk for severe allergic reactions to the vaccine. [38] These allergic responses have occurred as late as 2 weeks after the administration of a dose of vaccine, so that that it may be prudent to complete the course of vaccination at least 2 weeks before travel is begun. Because of the significant number of allergic reactions and the relatively low risk for Japanese encephalitis in most travelers to Asia, the vaccine should be limited to travelers to areas where the Japanese encephalitis virus is endemic and who will be staying at least 1 month, who are likely to have significant exposure in rural areas, and who will be in an area of endemicity in temperate zones during the appropriate (summer) season.

Typhoid Fever Infection with *Salmonella typhi* remains a significant problem in much of the developing world. Of the approximately 500 cases of typhoid fever reported in the United States yearly, about 60% are acquired during foreign travel. [39] The risk for typhoid fever in travelers to areas where the disease is highly endemic, such as India and Peru, is estimated to be as high as 100 cases per 1 million travelers. [39] Typhoid fever can be prevented with strenuous efforts to avoid ingesting potentially contaminated food and water and with vaccination against typhoid fever. Vaccination is generally indicated for anyone traveling to a developing country where typhoid fever is endemic and exposure to contaminated food or water may be unavoidable. Three vaccines are available to prevent typhoid fever: the older, parenterally administered inactivated vaccine; a live attenuated oral vaccine; and a newly licensed acellular parenteral vaccine. There are probably no indications for using the older inactivated vaccine, which, although effective, is associated with significant injection site and systemic reactions. The oral vaccine consists of an attenuated (Ty21a) strain of *S typhi* and has been demonstrated to be effective (67% protection) in large-scale clinical trials, [40] probably providing protection for 5 years. [41] The oral vaccine should not be administered to immunocompromised persons or to pregnant women. Because dosing at home is required, careful instructions must be given to the traveler to ensure compliance. The vaccine should not be given to persons who are taking antibiotics, and it should be administered before travelers begin malaria prophylaxis with mefloquine. The new acellular vaccine, which contains polysaccharide components, is administered in a single parenteral injection and provides 2 years of protection against disease. [42] It is especially useful for persons who have medical contraindications to the oral vaccine, for travelers who do not have time to complete the oral vaccine dosage schedule before travel, and for those who cannot easily swallow capsules or may have other compliance problems.

Meningococcal Disease Travelers to destinations where epidemics of meningococcal disease are frequent (much of sub-Saharan Africa during the months of December to June) or countries with current epidemics of meningococcal disease should receive the meningococcal vaccine. Because of outbreaks in Saudi Arabia associated with the yearly arrival of pilgrims to Mecca (the hajj), Saudi Arabia now requires proof of meningococcal vaccination for all pilgrims. The quadrivalent polysaccharide vaccine available in the United States provides protection against serotypes A, C, Y, and W135. No vaccine that prevents serotype B disease is available. Protective immunity appears to last at least 3 years; the vaccine is less effective in children younger than 4 years of age. Significant side effects of vaccination are rare, and although the safety of the vaccine in pregnancy has not been established, available clinical data do not suggest any adverse effects of vaccination on the mother or fetus.

Cholera Cholera, caused by *Vibrio cholerae* O group 1 or O group 139, is associated with significant morbidity and mortality in many developing countries. The risk that travelers who follow basic precautions about avoiding contaminated food (especially uncooked or inadequately cooked fish and shellfish) and water will contract cholera is extremely low, however. The currently available cholera vaccines (prepared from killed bacteria) are only 50% effective in preventing disease, and the duration of protective immunity is no more than 6 months. For these reasons, the CDC does not recommend cholera vaccination for most travelers. [43] Persons with

impaired gastric acidity [44] or impaired immune systems who will be spending a significant amount of time in areas where the disease is highly endemic are at higher risk, however, and are candidates for the complete cholera vaccine series. Although cholera vaccination cannot be required for entry under the International Health Regulations, local authorities in certain countries may request documentation of cholera vaccination. In this case, a single dose of cholera vaccine will satisfy the requirement. Reactions to the vaccine include pain and erythema at the injection site and occasionally systemic symptoms such as fever, malaise, and headache.

Vaccines That May Be Updated

Poliomyelitis Significant progress is being made in the worldwide eradication of poliomyelitis, but foci of infection in developing countries in Africa, Asia, and the Indian subcontinent still present a risk (albeit extremely low) to the traveler. For persons who have completed the primary immunization schedule, a one-time booster dose before travel should be administered. It is best given in the form of the enhanced potency inactivated vaccine (IPV) rather than the oral polio vaccine (OPV) to minimize the risk for transmission of vaccine-associated disease. [45] If possible, the vaccination of pregnant women should be deferred; if immediate protection against poliomyelitis is needed, the current recommendation is for OPV. For adults who are uncertain about their vaccination status or who know they were never vaccinated, a complete series with IPV should be administered. If time does not permit a complete series, alternative schedules are available. [45]
Tetanus/Diphtheria Outbreaks of diphtheria in former states of the Soviet Union have served as a potent reminder of the ability of diphtheria to reemerge when breakdowns in public health result in inadequate immunization levels in the population. Diphtheria also remains a significant health problem in many developing countries worldwide. Tetanus is a global health problem that is completely preventable by adequate immunization. Adults and children older than 7 years of age who never were vaccinated against diphtheria and tetanus should receive primary immunization with the tetanus/diphtheria toxoid formulation for adult use (Td). The standard recommendation is that previously vaccinated persons should receive a booster immunization if more than 10 years has elapsed since completion of the primary immunization series or the last booster dose. More frequent Td booster schedules (every 5 years) appear to increase the number of adverse reactions to the vaccine. Because a booster immunization within the past 5 years obviates the need for a booster immunization in the event of a tetanus-prone wound, it may be prudent to vaccinate on the 5-year schedule patients who will not have access to good health care and will be in areas where the sterility and safety of injections are questionable.
Measles The risk for exposure to measles overseas is probably relatively high, and it is recommended that persons traveling abroad be immune to measles. Adults born before 1957 are presumed to be naturally immune to measles and generally are not considered for vaccination. For adults born in or after 1957 who have not received two doses of measles vaccine, a single dose of live attenuated measles vaccine should be administered before travel unless vaccination is specifically contraindicated (e.g., pregnancy, immune compromise in some cases, severe egg allergies). The measles, mumps, rubella (MMR) vaccine has been administered safely to HIV-infected individuals, and it is recommended that measles-susceptible HIV-infected travelers be vaccinated regardless of their current disease status. [46]
Other Vaccines Influenza occurs in the winter in temperate zones and year-round in tropical regions. The same groups that receive influenza vaccine during the influenza season in the United States (the elderly, patients with chronic diseases, health care workers) should be considered for vaccination before travel to an area at risk. Pneumococcal vaccine should be administered to travelers older than 65 years of age and persons with chronic illnesses.

PROTECTION AGAINST MALARIA

Malaria is caused by protozoan parasites of the *Plasmodium* genus, which are transmitted to humans by the bite of female *Anopheles* mosquitoes. It is estimated that more than 100 million people are infected with malaria worldwide, with 1 to 2 million deaths yearly, primarily in infants and children, attributable to malarial infection. Malaria in humans is caused by *Plasmodium falciparum, Plasmodium vivax, Plasmodium ovale,* and *Plasmodium malariae*. *P vivax* is probably the most prevalent cause of malaria worldwide, but *P vivax*–associated disease is rarely fatal. *P falciparum* is associated with the highest levels of parasitemia (the percentage of erythrocytes invaded), and it can cause fatal disease. The mortality rate in nonimmune adults for untreated *P falciparum* malaria is about 25%. [47] The risk of unprotected travelers to malarious areas is significant. From 1980 to 1992, more than 2500 cases of *P falciparum* malaria were reported to the CDC. Travel to sub-Saharan Africa was associated with the highest risk for disease (82% of reported cases); 8% of cases were acquired in Asia, 5% in the Caribbean and South America, and 5% in other regions. In 1992, 910 cases of malaria were reported in the United States and U.S. territories. Most were caused by *P vivax* (51%) and *P falciparum* (33%); few cases of *P malariae* (4%) or *P ovale* (3%) infection were reported. [48]

The prevention of malaria in travelers is based on two critical components. First, it is vitally important that persons in malarious areas take precautions to avoid the bite of mosquitoes (see section " Insects"). The second critical component is chemoprophylaxis, the use of antimalarial agents to prevent the establishment of infection. Chemoprophylaxis for malaria has become increasingly complicated as a result of the development of drug resistance in *P falciparum* and *P vivax* parasites. For years, chloroquine taken on a weekly schedule served as safe and effective prophylaxis against all forms of malaria; however, the past three decades have seen a spread of chloroquine-resistant *P falciparum* malaria from foci in Southeast Asia and South America to most of the rest of the malarious regions of the world. As shown in Figure 56-2, chloroquine-sensitive malaria is now confined to just a few areas of the world. The ineffectiveness of chloroquine has greatly complicated chemoprophylactic regimens. It is important for the physician to recognize that recommendations for malaria chemoprophylaxis for travelers to a given region of the world can change rapidly based on the emergence of drug-resistant strains, and physicians must keep up to date (through the CDC or other agencies) on the latest recommendations. The drug currently used for most travelers from the United States to malarious areas is mefloquine, which is taken on a weekly dosing schedule and, in large-scale studies, appears to be effective and safe (Table 56-4). [49] The most significant side effects associated with mefloquine are neuropsychiatric, but these effects are seen most frequently at the doses used for the treatment of malaria, not for prophylaxis. Mefloquine is not recommended for persons with a history of epilepsy or severe psychiatric conditions and is not recommended for patients with cardiac conduction abnormalities. The available data suggest that mefloquine is probably safe in pregnancy and should be the drug used for prophylaxis in pregnant women or in women who may become pregnant in areas where exposure to chloroquine-resistant malaria cannot be avoided. For persons who cannot take mefloquine or who are traveling to the few regions of the world where mefloquine-resistant *P falciparum* has been reported, doxycycline, taken daily, is an effective alternative regimen (see Table 56-4). A new agent, a combination of atovaquone and proguanil (Malarone), has appeared safe and effective in early trials and represents an alternative for those who cannot take mefloquine or doxycycline. [50]

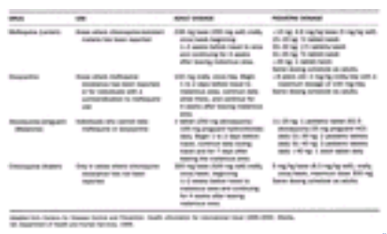

TABLE 56-4 Drugs Used for Malaria Prophylaxis

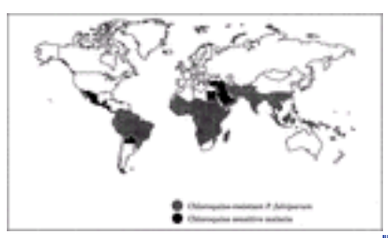

FIGURE 56-2. Distribution of malaria and chloroquine-resistant *Plasmodium falciparum.* (From Centers for Disease Control and Prevention. Health Information for International Travel 1999–2000. Atlanta, GA: Department of Health and Human Services, 2000.)

When travelers take precautions to prevent mosquito bites and are compliant with an appropriate chemoprophylactic regimen, the risk for acquiring malaria is probably low, but not nonexistent. For this reason, travelers should be counseled about the symptoms of malaria (fever, headache, malaise, fatigue, muscle aches, gastrointestinal upset) and told to seek medical care if they develop. Some physicians provide curative treatment regimens, with instructions for the traveler to take

the drug if symptoms of malaria develop. The traveler's ability to make the correct diagnosis is probably low, [51] however, and the side effects of drugs used in curative doses should make this a rarely used option.

OTHER INFECTIOUS DISEASES THAT POSE A RISK

Dengue fever is a viral disease transmitted by *Aedes* mosquitos. Symptoms of dengue fever include high fever, frontal headache, joint and muscle aches, nausea, and vomiting. A rash may be seen 3 to 5 days after the onset of illness. Dengue fever has now become the most common arbovirus disease of humans, and it is estimated that millions of cases occur each year. Epidemics have occurred throughout most of the tropics, including many countries in South America, Africa, and Asia. The disease is present in Mexico and the Caribbean, two major destinations for American tourists. The prevention of dengue fever is currently based solely on the avoidance of mosquito bites by means of the strategies outlined above.

Hepatitis E virus infection has been identified in a few American travelers. Epidemic and endemic hepatitis E transmission occurs in much of Asia, North Africa, and rural areas of central Mexico. The prevention of hepatitis E in travelers is based on the avoidance of potentially contaminated food and water.

TRAVELERS' DIARRHEA

Travelers' diarrhea affects 30% to 50% of travelers to developing countries. [52] Although the disease is generally mild and self-limited, it can severely disrupt travel plans. A recent survey of almost 800 American travelers found that 46% had an episode of diarrhea during their trip, and diarrhea was one of the major reasons individuals sought medical care on their return to the United States. [53] Long-term travelers also are afflicted. A study of 36 Peace Corps volunteers in Guatemala revealed that each individual had on average seven episodes of diarrhea during a 2-year stay. [54] Diarrheal episodes accounted for 10% of the 23,689 person-days of illness in the study.

Enterotoxigenic *Escherichia coli* (ETEC) is the most common cause of travelers' diarrhea worldwide, but *Shigella* species also account for a significant proportion of cases, and *Campylobacter* species are being increasingly recognized as a problem among travelers to Asia. Among bacterial pathogens, *Aeromonas, Plesiomonas,* and *Vibrio* species also are occasionally isolated. [55] Rotaviruses and the Norwalk virus may account for 5% to 15% of cases of travelers' diarrhea. [56] Whereas parasitic causes are relatively uncommon, *Giardia* species are seen in 5% of cases, [57] and a newly recognized protozoan pathogen, *Cyclospora cayetanensis,* is an occasional cause of travelers' diarrhea. [58], [59] *Cryptosporidium parvum* is seen relatively infrequently as a cause of travelers' diarrhea, but it can be a particular problem for immunocompromised individuals. Microsporidia have been identified in the stool of some travelers with diarrhea, but whether these are clinically important is unclear. [60] *Entamoeba histolytica* is an unusual cause of travelers' diarrhea but must be considered in anyone with bloody stools. The etiologic agents of travelers' diarrhea vary by geographic region. In an analysis of 432 military personnel in whom diarrhea developed during Operation Desert Shield, an enteropathogen was isolated in about half, with 29% of these having ETEC, 25% *Shigella* species (mostly *S sonnei*), 7% *Salmonella* species (not *S typhi*), and only 2% *Campylobacter* species. [61] In contrast, among American military personnel deployed in Thailand, more than half of those in whom diarrhea developed were infected with *Campylobacter* species, whereas only a few cases of diarrhea caused by *Shigella* species or ETEC were detected. [62]

Most cases of travelers' diarrhea occur between 5 and 15 days after arrival. [63] The illness is heralded by malaise, anorexia, and abdominal cramps, followed by watery, usually nonbloody, diarrhea. In some cases, nausea and vomiting may be a prominent component, whereas a low-grade fever is present in about 33% of cases. Most bouts of travelers' diarrhea are self-limited, with resolution after 4 to 6 days, but in a significant number of travelers, disease is longer-lasting (6–10 days). Persons with gastric hypoacidity and immunosuppressed patients are probably at greater risk for the development of travelers' diarrhea. [63] These epidemiologic findings are supported by studies that show that gastric hypoacidity leads to a reduction in the infectious dose of *V cholerae* and *Salmonella* species. [44], [64] Travelers with ulcerative colitis, Crohn's disease, or celiac sprue appear to be at greater risk for severe cases of travelers' diarrhea, which may exacerbate their underlying disease. [65], [66] Many infections, such as those caused by *Shigella, Salmonella,* and *Cryptosporidium* species, may be much more severe in immunocompromised patients.

Travelers' diarrhea is contracted by the ingestion of fecally contaminated food or water; hence, the first line of defense for the traveler is care in selecting food and beverages, as previously outlined. Whereas stringent precautions can reduce significantly the chances of contracting travelers' diarrhea, they do not completely eliminate the risk. For this reason, two other measures can be used. The first approach is chemoprophylaxis with antibiotics to prevent diarrhea from developing. An alternative approach is to provide the traveler with antibiotics to be taken when diarrhea develops. Benefits and risks are associated with each of these approaches. Prophylactic antibiotics represent the most effective approach to preventing diarrhea in travelers. Agents tested for their ability to prevent the onset of diarrhea in travelers include doxycycline, trimethoprim-sulfamethoxazole (TMP-SMX), and quinolones. Doxycycline was studied in Kenya, Morocco, and Mexico and showed efficacy of 81% to 86% in preventing diarrhea. [67] The development of widespread resistance to doxycyline in *Shigella,* ETEC, and other enteropathogens, however, has effectively ended the use of doxycyline for this indication. TMP-SMX has been tested in Mexico, where a single-dose regimen conferred 95% protection during a 2-week period. [68] The use of TMP-SMX has been curtailed by the development of resistant organisms in areas of endemicity, and the possibility of severe side effects (allergies and rarely Stevens-Johnson syndrome) should limit the use of TMP-SMZ for this indication. The most commonly used agents currently are quinolones. Norfloxacin in a daily dose of 400 mg had a protective efficacy of 93% in Egypt, and of 88% in a study in Mexico. [65], [69] A single daily dose of ciprofloxacin (500 mg) had a protective efficacy of 94% in a study in Tunisia. [70] In areas where ETEC and *Shigella* and *Salmonella* species predominate as causes of travelers' diarrhea (most of the developing world), a quinolone represents the drug of choice for travelers if antibiotic prophylaxis is used or for the treatment of travelers' diarrhea. This is not the case for travelers to Thailand and neighboring regions in Asia, where *Campylobacter* species are an important cause of travelers' diarrhea. [62], [71] Many isolates of *Campylobacter* species in these areas are resistant to quinolones, and azithromycin, a new, broad-spectrum macrolide that is effective against *Campylobacter* species and other enteric pathogens, can be used at a dose of 500 mg/d as prophylaxis or as treatment by travelers to these regions. [72] As a cautionary note, it should be stated that a few isolates of azithromycin-resistant *Campylobacter* species have been recognized in Thailand. [62]

The advantage of prophylactic antibiotics is their high level of efficacy in preventing disease, which in most cases ensures that travel will not be interrupted by the development of diarrhea. The disadvantages relate to the possibility of side effects of the antibiotics and concerns about the selection of antibiotic-resistant organisms. For these reasons, the routine use of antibiotic prophylaxis to prevent travelers' diarrhea is generally not recommended. Because travelers' diarrhea is usually a relatively benign and self-limited illness, the potential risks of any prophylactic therapy must be low to justify its use. There are two settings in which prophylactic antibiotics may be useful. First, for the short-term traveler (2 weeks or less in an area of endemicity) whose vacation or business schedule would be severely disrupted by a bout of diarrhea, the potential benefits of prophylactic antibiotics may outweigh the risks. This should be discussed in detail with the traveler before prophylactic antibiotics are prescribed. Second, persons who cannot tolerate a bout of travelers' diarrhea because of a medical condition or who are at increased risk for acquiring the disease (e.g., immunocompromised persons, those with gastric hypoacidity, and those with underlying gastrointestinal diseases) are reasonable candidates for longer-term prophylaxis.

Some authorities recommend chemoprophylaxis with bismuth subsalicylate. [73], [74] However, this is only moderately effective (approximately 65%) in preventing diarrhea and can be associated with side effects related to both the salicylate and bismuth components. For this reason, we do not routinely advocate the use of bismuth subsalicylate for the prevention of travelers' diarrhea.

The most widely used approach to travelers' diarrhea is probably the provision of antibiotics to be used by the traveler if and when diarrhea strikes. It needs to be emphasized before travel that self-treatment regimens are not appropriate for the traveler with bloody diarrhea, severe abdominal pain, or high fever. In these cases, medical care should be sought immediately. A reasonable current recommendation is to provide a 3-day course of a quinolone for travelers to most developing countries. Travelers to Thailand and surrounding countries in Southeast Asia are provided with a 3- to 5-day course of azithromycin instead. The patient is told to begin the antibiotic when diarrhea starts and to continue the treatment for 3 days. Studies have demonstrated that a single dose of ciprofloxacin (500 mg taken orally) can reduce the duration of travelers' diarrhea from 50 hours to 24 hours and reduce the number of liquid stools from 11.4 to 5.0. [75] Forty-eight hours after a single dose of fleroxacin (400 mg orally), 67% of travelers had stools of normal consistency, compared with 37% of travelers receiving a placebo. [76] These studies suggest that single-dose antibiotic therapy with one of the quinolone regimens is effective treatment for travelers' diarrhea and also can be recommended for self-treatment by travelers. Whether the addition of the antimotility agent loperamide to antibiotic treatment is beneficial is unclear. The combination of the antimotility agent loperamide and TMP-SMX was superior in reducing the duration of diarrhea to either agent alone in a study undertaken in Mexico. [77] In a placebo-controlled, randomized trial comparing the combination of loperamide and ciprofloxacin with ciprofloxacin alone, a slight but statistically insignificant benefit of the combination was noted during the first 24 hours after the initiation of therapy, but only in individuals infected with ETEC. [78] This may reflect a greater efficacy of the quinolone regimen in controlling diarrhea in comparison with the TMP-SMX regimen. [78] Because the addition of loperamide was not associated with any adverse effects in either study, it probably is

reasonable to continue to offer loperamide as an adjunctive treatment for the symptomatic relief of travelers' diarrhea, pending the results of further studies of its efficacy. For the traveler who cannot be given one of the suggested antibiotics, bismuth subsalicylate also has been used to treat diarrhea and has been shown to shorten the course of illness in placebo-controlled studies. [79] The same cautions about the potential side effects of ingesting large quantities of salicylate in prophylactic bismuth subsalicylate are even more applicable to the higher doses recommended for treatment.

Finally, the most important component of self-treatment is the replacement of the fluid and electrolytes lost during diarrhea. The best approach is the use of an oral rehydration solution, such as the World Health Organization oral rehydration salts solution. Packets are available at stores or pharmacies in most developing countries, and when dissolved in boiled or chemically treated water, a safe solution is created containing sodium chloride (3.5 g/L), potassium chloride (1.5 g/L), glucose (20.0 g/L), and trisodium citrate (2.9 g/L).

Returning Traveler

Watery diarrhea occurring in travelers within the first few days after return usually represents travelers' diarrhea that was contracted before the return trip. [24, 80, 81] The initial approach should be that used for the self-treatment of travelers' diarrhea (provision of a 3- to 5-day course of a quinolone or azithromycin and possibly loperamide with appropriate fluid replacement). The diarrhea should resolve in 24 to 48 hours with therapy. It is critical to remember that diarrhea can be a prominent symptom of malaria, and the traveler returning from malarious areas who has fever and diarrhea must have blood smears examined to exclude *Plasmodium* infection. Watery diarrhea that occurs later after return or that persists longer than 10 days despite antibiotic therapy is most commonly caused by *Giardia lamblia* infection. [81] Stools (preferably three) should be examined for parasites, including *Giardia, Cryptosporidium* (especially in immunocompromised individuals), *Cyclospora,* and *Isospora* species (see Chapter 125). *E histolytica* has been reported as cause of nondysenteric chronic diarrhea, but even if not frankly bloody, stools are almost always guaiac positive in amebiasis. Stool cultures should be obtained, and testing for *Clostridium difficile* should be performed. If no pathogen is detected and a trial of antibiotics (quinolone or azithromycin) was not previously administered, it should be initiated at this time. If antibiotics were used previously or are now unsuccessful, presumptive treatment for *Giardia* infection with metronidazole (250 mg taken orally three times a day for 5 to 7 days) is indicated. [82] If the diarrhea fails to respond to metronidazole, a gastrointestinal evaluation should be performed, including upper gastrointestinal endoscopy, small bowel biopsy, examination of smears for parasites, cultures for enteropathogens, quantitative culture for small bowel overgrowth, and examination of biopsy specimens for evidence of tropical sprue. [82] Flexible sigmoidoscopy and biopsy of the rectal mucosa also may be useful in this setting. Studies of absorptive function may help to localize disease to the small bowel. [82] If the clinical symptoms and biopsy specimen findings are consistent with tropical sprue, treatment with tetracycline (250 mg four times daily for 6 weeks) given with folate (5 mg daily) is indicated. If no cause is found for the diarrhea after this extensive workup, the traveler should be advised to try symptomatic treatment and should be reassured that most cases of chronic diarrhea eventually resolve.

The diagnostic and therapeutic considerations differ somewhat for the returning traveler with bloody diarrhea, and the pace of the workup should be accelerated. *Shigella, Campylobacter,* and *Salmonella* species are the most likely initial possibilities, and amebiasis is a consideration in this setting. Stool culture, with a search for the aforementioned three agents in addition to enterohemorrhagic *E coli* and *C difficile* (if there is a history of prior antibiotic ingestion), and examination of three stools for ova and parasites or a specific enzyme-linked immunosorbent assay (ELISA) for amebiasis also should be performed. After the stools have been collected, empiric therapy with a quinolone or azithromycin can be initiated pending the results of culture and microscopy. If the stool is negative for enteropathogens, including *E histolytica,* flexible sigmoidoscopy to examine the rectal and colonic mucosa is indicated. Samples for culture and microscopy for *E histolytica* should be obtained, and serologic testing for amebiasis should be performed. Biopsy specimens of abnormal tissue should be examined for amebiasis and for noninfectious causes of bloody diarrhea, including inflammatory bowel disease. It must be remembered that travel can unmask previously unrecognized or dormant intestinal diseases, such as inflammatory bowel disease, irritable bowel syndrome, celiac sprue, and lactose intolerance.

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CHAPTER 57

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COMPLEMENTARY AND ALTERNATIVE MEDICINE IN GASTROENTEROLOGY

One of the major trends in health care during the past decade has been the marked growth in the use of complementary and alternative medicine (CAM). Two widely used definitions of CAM are “those health care and medical practices that are not currently an integral part of conventional (Western) medicine,” and “those interventions neither taught widely at U.S. medical schools nor generally available in U.S. hospitals.” During the 1990s, the federal government created the National Center for Complementary and Alternative Medicine (NCCAM), as well as the Office of Dietary Supplements. Moreover, versions of *Physicians’ Desk Reference* have been created for both herbal therapies and dietary supplements. [1], [2] CAM has been grouped within five major areas by the NCCAM. The greatest proportion of human as well as experimental animal and in vitro research relating CAM to gastrointestinal (GI) diseases has evaluated biologically based therapies (the focus of this chapter). Other forms of CAM include manipulative and body-based methods, energy therapies, mind-body interventions, and alternative medical systems (Table 57-1).

TYPE OF CAM	EXAMPLES
Biologically based therapies	Herbal therapies
	Special diets to promote health
	Orthomolecular therapies (e.g., megavitamins)
	Biologic therapies (e.g., shark cartilage)
Alternative medical systems	Oriental medicine
	Maintaining proper balance of Qi
	Traditional Indian medicine, Ayurveda
	Homeopathy (dilute treatments more potent)
Mind-body interventions	Meditation
	Prayer
	Mental healing
	Art, dance, and music therapy
Manipulative and body-based methods	Osteopathy
	Massage therapy
	Chiropractic
Energy therapies	Qigong
	Touch therapy to maintain body's energies and balance
	Bioelectromagnetically based therapies

TABLE 57-1 Types and Examples of Complementary and Alternative Medicine

The increasing interest in CAM was highlighted in a study by Eisenberg and colleagues [3] that demonstrated a high prevalence of CAM use and a large sum of out-of-pocket dollars spent on CAM in the United States in 1990. A follow-up survey documented trends in CAM use in the United States between 1990 and 1997. [4] CAM use increased from 36% to 46% during that 7-year period. Of great importance, almost 40% of patients did not disclose their CAM use to their traditional physicians. In 1997, approximately 15 million adults took prescription medicines in conjunction with herbal remedies or high-dose vitamins, and total out-of-pocket costs for CAM were estimated to be at least $27 billion. Patients reported equal confidence in their CAM providers and their traditional medical doctors. [5] A 2001 survey found that approximately half of American adults were taking nonprescription dietary or mineral supplements. [6] Moreover, approximately one in six Americans reported regular use of a supplement such as ginseng, amino acids, or over-the-counter hormones. CAM users tend to be highly educated, are often white (non-Hispanic), and frequently use CAM therapy for chronic diseases. CAM use in chronic illness may result from frustration with the lack of efficacy of conventional therapy and the fact that CAM allows patients some control over their therapy. Thus, it should not be surprising that CAM is frequently used by patients with chronic GI diseases. Moreover, many users of CAM (especially dietary supplements) emphatically believe in the usefulness of this therapy. Indeed, when asked what they would do if a government agency said that the supplement they used most often was ineffective, 71% of regular users reported they would still continue to take the supplement. [6] Because of all these factors, it is important that health care providers be knowledgeable about CAM, including both potential benefits and side effects. This chapter reviews CAM use as it applies to major GI disease processes.

CAM USE IN LIVER DISEASE

Use of CAM in liver disease appears to parallel overall widespread CAM use in the United States, as indicated by data presented at the 1999 Complementary and Alternative Medicine Symposium on Chronic Liver Disease at the National Institutes of Health. [7] We will review milk thistle (silymarin), glutathione prodrugs, SAMe,

vitamin E, lecithin, herbals, and glycyrrhizin in liver disease.

Silymarin (Milk Thistle)

Silymarin is the active ingredient extracted from *Silybum marianum* (“milk thistle”). The plant is a member of the daisy family, and the leaves have prominent white “milky” veins. [8] This agent protects animals from multiple types of experimental liver injury induced with carbon tetrachloride, acetaminophen, iron overload, bile duct obstruction, and, very importantly, mushroom poisoning. [8], [9] and [10] Its hepatoprotective effects are highlighted by its use in the treatment of *Amanita* mushroom poisoning. Studies in several animal species have documented the efficacy of this agent in mushroom poisoning, both in pretreatment phases and following ingestion. [8], [9] and [10] Moreover, several cases have been reported in which dramatic results were achieved in patients up to 3 days after accidental ingestion.

In the United States, silymarin is probably the most widely used form of CAM in the treatment of liver disease. Clinically, it may have hepatoprotective effects in various forms of toxic hepatitis, fatty liver, cirrhosis, ischemic injury, and virus-induced liver disease. [8], [9] It has antioxidant activities, protects against lipid peroxidation, and has antiinflammatory and antifibrotic effects. Large controlled trials of silymarin have been performed in Europe, with variable results. Ferenci and colleagues [11] evaluated 170 patients with cirrhosis in a treatment program (140 mg three times daily) that had a mean duration of 41 months. They observed a positive beneficial effect, especially in patients with alcoholic cirrhosis and in those with milder disease (Child class A). On the other hand, a study by Pares and colleagues [12] found no beneficial effects (150 mg of silymarin three times daily) in a study of 200 patients with alcoholic cirrhosis, some of whom also had hepatitis C. Both of these trials had major shortcomings, including high dropout rates and compliance issues. Despite these limitations, silymarin is likely to remain one of the most popular forms of CAM therapy for liver disease because it has a good safety profile and has been extensively investigated in multiple forms of experimental liver injury in animals, and some positive results have been reported in humans.

Glutathione Prodrugs

Glutathione is a tripeptide that is synthesized from glutamate, cysteine, and glycine. Glutathione, in its reduced form, is the main nonprotein thiol in the cells and has an important role in the detoxification of electrophils and protection against reactive oxygen toxicity. [13] Glutathione cannot be taken up by hepatocytes, but a number of pharmacological agents have been devised to enhance intracellular pools. The most widely used include *N*-acetylcysteine (NAC) and 2-oxothiazolidine-4-carboxylic acid (OTZ), with NAC available as a supplement. [14]

Glutathione prodrugs have been used extensively in virtually every known experimental model of hepatotoxicity with beneficial results. [13], [14] The glutathione prodrug NAC (given as Mucomyst) is the standard therapy for acetaminophen toxicity in humans. [15] Maintaining adequate hepatocyte glutathione levels has been documented to prevent acetaminophen liver injury. However, the information on glutathione prodrugs in prospective randomized studies in other forms of liver disease is very limited. Unfortunately, the initial promise of increased efficacy of using reduced glutathione (GSH) prodrug/antioxidant therapy in combination with interferon for hepatitis C appears not to have held up under scrutiny. [16] Glutathione prodrugs can directly affect the hepatocyte, and they can also positively modulate proinflammatory cytokine production with inhibition of cytokines such as tumor necrosis factor (TNF) and interleukin-8 (IL-8). [17] This presumably occurs through inhibition of the oxidative stress–sensitive transcription factor nuclear factor-?B (NF-?B), which plays a central role in lipopolysaccharide (LPS)-stimulated TNF production. Pena and colleagues [18] demonstrated that the GSH prodrug procysteine can increase whole-blood GSH and inhibit monocyte TNF and IL-8 production when given intravenously to patients with stable cirrhosis. Similarly, procysteine was shown to protect against alcohol-induced liver injury in the intragastric ethanol feeding model. [19] Thus, GSH prodrugs can, in theory, decrease oxidative stress in hepatocytes and attenuate proinflammatory cytokine production in Kupffer cells to inhibit hepatotoxicity in processes such as alcoholic liver disease (Fig. 57-1).

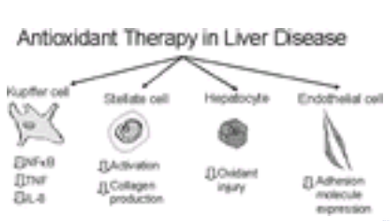

FIGURE 57-1. Most forms of complementary alternative medicine therapy in liver disease have antioxidant effects. These theoretically decrease proinflammatory cytokine production, decrease stellate cell activation and subsequent collagen production, attenuate oxidant injury in hepatocytes, and decrease endothelial cell adhesion molecule expression. *IL-8,* interleukin-8; *NF-?B,* nuclear factor-?B; *TNF,* tumor necrosis factor.

***S*-Adenosylmethionine**

S-Adenosylmethionine (SAMe, adomet) is an obligatory intermediate in the conversion of methionine to cysteine in the hepatic trans-sulfuration pathway. The conversion of methionine to SAMe involves transfer of the adenosyl moiety of ATP to the sulfur atom of methionine and is catalyzed by methionine adenosyltransferase (MAT). SAMe is a precursor for the synthesis of polyamines, choline, and GSH. SAMe is also the major methylating agent for a vast number of molecules via specific methyltransferases. [20]

Patients with many types of both acute and chronic liver disease have elevated plasma methionine levels, markedly delayed clearance of an oral methionine load, and decreased hepatic MAT activity. Because SAMe is a precursor of GSH synthesis, its deficiency may result in the GSH deficiency reported in the liver and serum of patients and experimental animals with liver disease. Because GSH is required for the optimal expression of MAT activity in liver, hepatic deficiency of MAT may in part be a consequence of GSH deficiency. Hepatic-specific MAT is sensitive to oxidative stress, and it is likely that the subnormal hepatic MAT activity reported in alcoholic liver disease is a result of oxidation of the active site through increased oxidative stress. [21]

Subnormal hepatic SAMe levels have been reported in various experimental models of liver injury, and SAMe therapy in rats attenuates liver injury secondary to cholestasis, ethanol, carbon tetrachloride, fatty liver, acetaminophen, or galactosamine. [20] MAT A1 (liver-specific MAT) knock-out mice have been generated. [22] Initially, they are highly susceptible to another “stress,” such as a choline-deficient diet. Later, fatty liver and periportal inflammation develop, which may have a great relevance not only to alcoholic liver disease but also to nonalcoholic steatohepatitis (NASH). Lastly, very late in life, hepatic cancer develops in some mice. MAT deficiency modulates the expression of many genes, especially those involved in proliferation/growth, and normal MAT A1 activity appears to be important for maintaining the hepatocyte in a “differentiated” state. [22], [23] Studies from our laboratories have shown that SAMe down-regulates production of the cytotoxic proinflammatory cytokine TNF in animal models of liver injury and in peripheral blood monocytes or macrophage cell lines in vitro. [24] Mato and colleagues [25] reported that patients with alcoholic liver cirrhosis who were randomized to receive SAMe (1200 mg/d orally) for 2 years had decreased rates of liver mortality/liver transplantation (16% versus 30%) compared to a placebo-treated group. SAMe has also been used successfully in various types of cholestatic liver disease. Thus, substantial animal studies and emerging clinical data indicate that SAMe may be useful in several forms of liver disease. Some potential beneficial effects of SAMe include increased GSH production, improved mitochondrial function, decreased proinflammatory cytokine production, and enhanced methylation reactions.

Vitamin E

Vitamin E is a potent chain-breaking antioxidant that is widely used as a nutritional supplement. In several types of liver disease, depressed serum and hepatic levels of vitamin E have been documented. [26]

Vitamin E has been used extensively with hepatoprotective effects in experimental models of liver injury, such as that induced by carbon tetrachloride or ischemia. [27], [28] and [29] Vitamin E has been used in human liver disease with some success, with probably the most compelling data generated in children with NASH. [30] Obese children were given vitamin E (400–1200 IU) in an open-label pilot study, and all children with NASH demonstrated an improvement in liver enzymes without losing weight. Vitamin E has multiple potential beneficial effects, probably the best recognized being membrane stabilization. [28] Vitamin E–treated peripheral blood monocytes and Kupffer cells demonstrated reduced NF-?B activation and TNF production. [17] Vitamin E also inhibits hepatic stellate cell activation and collagen

production, thus inhibiting fibrosis (see Fig. 57-1). [31]

Polyenylphosphatidylcholine (Lecithin)

Polyenylphosphatidylcholine (PPC) is a lipid extract from soybeans that has been shown to have hepatoprotective effects, especially in alcoholic liver disease. [32, 33] PPC (specifically, dilinoleoylphosphatidylcholine, or DLPC) was shown to prevent the development of septal fibrosis and cirrhosis in alcohol-fed baboons, and to stimulate the release of collagenase activity by cultured hepatic stellate cells. DLPC was also shown to decrease stellate cell activation in rats. [32, 33] The effect of this agent was subsequently extended to two different models of experimental liver fibrosis (CCl_4 and heterologous albumin) in which DLPC was shown not only to attenuate liver fibrosis, but also to accelerate its regression. DLPC was also shown to decrease the hepatic levels of lipid peroxidation metabolites (4-hydroxylnoneneal and F2 isoprostanes) in ethanol-fed animals (rats and baboons), [34] suggesting that DLPC possesses antioxidant properties that may contribute to its antifibrotic and cytoprotective effects. These animal studies of DLPC have led to a Veterans Administration cooperative study evaluating its effects in patients with early alcoholic liver disease, the results of which should influence our use of this CAM agent.

Oriental Herbals

Herbals are widely used for a variety of chronic inflammatory processes, such as rheumatoid arthritis, as well as chemoprevention of cancer. Green tea and green tea polyphenols down-regulate proinflammatory cytokine production (e.g., TNF) and block endotoxin lethality and endotoxin-induced hepatotoxicity in galactosamine-sensitized rats. [35] Other major polyphenols are made from grape seeds; they also have antioxidant/antiinflammatory properties and have been used to protect experimental animals from liver injury.

TJ9 is commonly used in China and Japan and comes from an aqueous extract from the roots of a variety of plants. This agent has been shown to be hepatoprotective/antibiotic in certain animal models of liver injury and to down-regulate proinflammatory cytokine production. [7, 36] It is frequently used to treat both hepatitis B and hepatitis C in the Orient. Compound 861 is an aqueous extract of ten defined herbs that is reported to have very potent antifibrotic activity. It has been used extensively in China as an antifibrotic agent and has been reported actually to decrease fibrosis in sequential liver biopsy specimens from patients with hepatitis B. [7, 37] It has also been shown to be effective in CCl_4-induced liver injury and to have antiproliferative effects on stellate cells in vitro. One of the major problems with herbals is that they are actually a combination of agents that are poorly characterized and not highly reproducible from one lot to the next. The potential uses and risks of herbals have recently been reviewed, and summaries are available in the Cochrane Reviews. [7, 8, 38]

Glycyrrhizin

Glycyrrhizin is an extract made from licorice roots that has been used in a variety of GI disease processes ranging from peptic ulcer disease to liver disease. Glycyrrhizin has been reported to improve liver enzymes in a variety of patients with chronic hepatitis, including hepatitis C. [39] A retrospective study of intravenous glycyrrhizin therapy in patients with chronic hepatitis C showed approximately a 2.5-fold reduction in the risk for hepatocellular carcinoma. [40] Although glycyrrhizin has potential beneficial effects, it also has aldosterone-like properties that predispose to electrolyte imbalance and fluid retention, and therefore we do not recommend that our patients use this drug. [41]

CAM USE IN PANCREATIC DISEASE

Currently, effective therapies for pancreatic disease, especially chronic pancreatitis, are limited. Relief of abdominal pain, malabsorption, and diabetes are the main areas of focus, with abdominal pain being the most difficult symptom to treat. Because pancreatic enzyme replacement therapy is ineffective in controlling pain [42] and large randomized studies evaluating endoscopy or surgery are lacking, [43] patients and providers are actively seeking alternative therapies for chronic pancreatitis, including acupuncture, herbal therapies, and antioxidant therapy.

Acupuncture

Acupuncture has been used for decades in traditional Chinese medicine. The Chinese believe that the interaction of yin and yang creates the vital life force known as Qi (Chee) and that diseases occur when the flow of Qi is disrupted (excessive or deficient). Qi flows through the body in meridians or channels and can be accessed via acupoints. Modern functional magnetic resonance imaging studies have shown reproducible, regionally specific, and quantifiable effects on relevant structures in the brain when acupoints are stimulated. [44] However, in the only randomized prospective study of 23 patients with nonalcoholic chronic pancreatitis, neither electroacupuncture nor transcutaneous electrical nerve stimulation was as effective as medical therapy. [45]

Antioxidants

The increased use of antioxidants as an alternative therapy for chronic pancreatitis is based on the compelling evidence of persistent oxidant stress in patients with chronic pancreatitis, [46] in addition to increased lipid peroxidation and altered glutathione metabolism. [47] Furthermore, regardless of the cause of injury, oxidative stress is felt to be pivotal in the progression to parenchymal pancreatic tissue destruction. [48] Serum antioxidant profiles of patients with acute and chronic pancreatitis, and of those with hereditary pancreatitis, reveal lower plasma concentrations of selenium, vitamin A, vitamin E, xanthine, ß-carotene, ß-cryptoxanthine, and lycopene in comparisons with control subjects. [49, 50] Additionally, selenium deficiency in chronic pancreatitis has been well documented, with some patients experiencing relief of chronic pain after selenium supplementation. [51]

The histological hallmarks of chronic pancreatitis include loss of parenchyma, fibrosis, blockage of pancreatic ducts with protein plugs that calcify, and variable degrees of inflammatory cell infiltration with a predominance of lymphocytes. Cytokines and chemokines released from these cells promote ongoing inflammation. Elevated proinflammatory cytokines have been found in the serum, [52] peripheral blood monocytes, [53] and more recently in the pancreatic tissue [54] and pure pancreatic juice [55] of patients with acute and chronic pancreatitis. Cytokine response predicts disease severity and outcome, supporting the rationale for the antioxidant/cytokine blocking therapies now under development. In a prospective trial of ten patients with chronic and acute recurrent pancreatitis of various causes, supplementation with a complex containing vitamin E, vitamin C, and organic selenium resulted in significant pain relief and a decreased need for hospital admission during the year of treatment in comparison with the year before treatment. [56] In a 20-week, double-blinded, placebo-controlled, switch-over trial of supplementation with selenium, ß-carotene, vitamin C, vitamin E, and methionine, most patients reported fewer attacks with significantly less background pain while on treatment. [57] A retrospective cross-sectional audit of 94 consecutive patients with chronic pancreatitis on antioxidant therapy followed for an average of 30 months showed a decrease in the requirements for surgical intervention, in the number and intensity of painful attacks, and in absences from work. [58] Large prospective placebo-controlled trials are needed to confirm these preliminary observations.

CAM USE IN PEPTIC ULCER AND NONULCER DYSPEPSIA

Peptic ulcer disease remains prevalent despite the recognition of its association with *Helicobacter pylori* infection and the availability of potent antisecretory medications. For thousands of years, alternative therapies were used to provide symptom relief and promote ulcer healing, especially in Indian and Chinese traditional medicine.

Acupuncture

Acupuncture has been used extensively in China and the former Soviet Union for the treatment of peptic ulcer. The physiological basis of acupuncture effects on acid secretion has been well studied in animals and humans. [59] In a prospective randomized study of human volunteers, electroacupuncture, but not sham acupuncture or simple acupuncture, significantly reduced vagally stimulated acid secretion. [60] Clinical trials reporting efficacy for acupuncture in humans with gastric or duodenal ulcer have been criticized for poor study design and lack of randomization.

Acupuncture may be useful for persons with nonulcer or functional dyspepsia. These patients have been shown to be hypersensitive to gastric distention, and somatic stimulation with transcutaneous electric stimulation can reduce the discomfort associated with gut distention. [61] Electroacupuncture modulates gastrointestinal motility

by significantly increasing the percentage of regular gastric slow waves as measured by electrogastrography (EGG) [62] and by influencing duodenal peristalsis. Several small uncontrolled trials claim efficacy for acupuncture in subjects with nonulcer dyspepsia. [63], [64] Future controlled randomized trials are needed to clarify whether patients with nonulcer dyspepsia or documented gastroduodenal dysmotility can benefit from acupuncture.

Herbal Therapies

Sangre de grado is derived from the sap of several Amazonian trees of the *Croton* genus (*C dracanoides, C planostigma, C lechleri*). It is commonly used in tropical South America to treat a variety of GI illnesses, including infectious diarrhea, gastritis, and peptic ulcer, and it is also used topically for wounds, bites, and insect stings. [65] It is inexpensive and widely available, unlike many of the Western pharmaceuticals. Its mechanism of action is unknown, but its efficacy has been attributed to potent antibacterial properties. Despite the high prevalence of *H pylori* infection in South America, no current studies have focused on sangre de grado and *H pylori* eradication per se.

In a randomized clinical trial of 508 patients with endoscopically proven gastric or duodenal ulcers and documented *H pylori* infection, the Chinese herb Chuanjia Weidan was compared with cimetidine. Ulcer healing rates and symptom reduction were comparable, but the *H pylori* clearance rate was 75% with Chuanjia Weidan therapy, versus 6% for the cimetidine-treated group. Ulcer recurrence rates were also higher for the cimetidine treated group. [66] A preliminary screening of more than 200 different Chinese herbal medicines revealed that 38 of the preparations had growth-inhibiting effects on *H pylori* organisms cultured from 136 subjects with antral infection. [67]

Other botanicals and herbals with reported efficacy and extensive use in the treatment of humans with peptic ulcer include flavonoids (quercetin, silymarin, naringin, anthocyanosides, sophoradin derivatives), saponins, tannins, gum guar and mucilages (myrrh), licorice, aloe gel, goldenseal, and capsicum (chili). [68] Proposed mechanisms of action include inhibition of prostaglandins, cytoprotection, antiinflammatory or immune modulatory effects, altered nociception, and suppression of *H pylori*. However, well-designed randomized controlled trials supporting these claims are lacking.

CAM USE IN IRRITABLE BOWEL SYNDROME

Irritable bowel syndrome (IBS) is one of the functional bowel disorders characterized by chronic or recurrent abdominal pain and disturbed defecation without structural or biochemical abnormalities. Although the pathogenesis of IBS remains unknown, the documented altered bowel motility patterns and exaggerated pain perception to visceral distention (“visceral hyperalgesia”) suggest neurohormonal or autonomic dysfunction as a likely cause. [69], [70]

Treatments for IBS generally focus on symptom reduction; however, no single treatment is reliably effective. Rather, multiple regimens combining dietary and lifestyle modification, normalization of bowel habits, antispasmodics when indicated, and anxiolytics or antidepressants are used. Often, an initial approach is to increase dietary fiber; however, clinical controlled studies of fiber therapy have revealed conflicting results. [71] Because of the limited efficacy of conventional treatments for IBS, many patients seek alternative therapies. A questionnaire completed in a physician’s waiting room in Britain revealed that one in five patients with IBS had used some form of alternative therapy. Also, 41% were agreeable to consultation with alternative medicine practitioners, compared with only 6% of patients with “organic” upper GI diseases. Of alternative medicines used at home, herbal remedies were most common. [72] However, most people with IBS symptoms do not seek formal medical care, and the use of alternative therapies in these individuals will remain difficult to quantify unless surveys of large cross sections of the population are developed and administered.

Acupuncture

Acupuncture therapy may benefit patients with IBS through a variety of mechanisms, including its effects on GI motility and autonomic function. Coffin and colleagues [61] examined the effects of transcutaneous electrical nerve stimulation of the hand during balloon distention of the stomach and duodenum and found that it induced somatosensory stimulation and raised the discomfort threshold. Kunze and colleagues [73] found a greater relief of symptoms with real (31%) than with sham (17%) acupuncture in patients with IBS. Among ten patients with IBS, Diehl and colleagues [74] reported a good response to acupuncture in four and a partial response in five, with one nonresponder. In another small study of seven patients given acupuncture for IBS, all reported an improved sense of general well-being and less abdominal bloating, but no change in abdominal discomfort or bowel patterns. [75]

Commonly Used Herbal Therapies

Essential oils are known for their carminative and antispasmodic properties and thus have been used for various GI ailments. Peppermint oil is derived from steam distillation of the arial parts of flowering *Mentha X piperita L*. Its active component is menthol, which has pharmacological properties similar to those of calcium channel antagonists. [76] Peppermint oil has been shown to relax smooth muscle from isolated human colonic cells. In a review of eight randomized controlled trials, peppermint oil use was associated with relief of IBS symptoms. However, only one trial selected patients based on the Manning diagnostic criteria for IBS, and many studies had methodological flaws, such that the authors of this metaanalysis recommended additional well-designed studies before definitive judgment about the use of peppermint oil in IBS could be made. [77]

Ginger root (*Zingiber rhizoma, Zingiber officinale*) has been used for thousands of years in Asia to treat multiple GI ailments, including nausea, dyspepsia, cholera, and diarrhea, as well as motion sickness, postoperative vomiting, and hyperemesis gravidarum. The pharmacological properties of ginger include antioxidant effects and inhibition of prostaglandins. It also increases intestinal tone and peristalsis. Ginger supplementation enhanced charcoal meal transport in mice, with effects felt to be similar to or slightly weaker than those of metoclopramide and domperidone. [78]

Licorice (*Glycyrrhiza glabra, Glycyrrhiza uralensis*) is used to treat functional dyspepsia, bronchitis, and peptic ulcer. It has expectorant properties and antiinflammatory and antiplatelet effects, and it increases prostaglandin levels. Overuse can result in salt and water retention, hyperkalemia, hypertension, and edema. Many drug-drug interactions are also possible.

Derived from the plant *Hypericum perforatum,* St. John’s wort is the most commonly used herb worldwide to treat depression and anxiety. In clinical studies comparing St. John’s wort with various serotonin reuptake inhibitors, it was equally effective for the treatment of depression and had fewer side effects. [79] The active ingredient of St. John’s wort, hypericin, acts by inhibiting the reuptake of norepinephrine, serotonin, and dopamine. Controlled trials of St. John’s wort use in IBS have not been reported.

Perhaps because of their agreeable flavor and aroma, or the ceremony of making and sharing a beverage with others, the commercial use of herbal teas is increasing. The following have reported benefits for the GI tract and their use is felt to be safe, but controlled clinical trials of efficacy have not been published. Chamomile (*Matricaria camomilla, Chamaemelum nobile*) is felt to have GI antispasmodic, antiinflammatory, and antimicrobial activities, in addition to anxiolytic and slight sedative effects. The licorice-tasting herbs fennel (*Foeniculum vulgare*) and anise (*Pimpinella anisum*) are touted to help expel gas and aid digestion. Lemon balm (*Melissa officinalis*) has mild sedative, carminative, and antispasmodic effects, and fragrant raspberry leaf (*Rubus idaeus,* the “pregnancy tea”) is reported to relieve morning sickness, nausea, and dyspepsia. [80]

Chinese Herbal Agents

In traditional Chinese medicine, rather than single agents being used, multiple herbs are combined for their synergistic properties. In a randomized placebo-controlled trial of 176 patients with IBS according to the Rome criteria, those receiving treatment with individualized herbs (n = 58) or a standard formulation of Chinese herbs (n = 63) reported significant relief of bowel symptoms and quality of life in comparison with those receiving placebo (n = 55). No greater benefit for those receiving individualized herbs rather than standard herbs during the 4-month study was found. However, only the subjects who had received individualized herbs showed a sustained relief of symptoms at 14 weeks after completion of the study. The authors concluded that further investigation of Chinese herbal medicine as a treatment for IBS should be undertaken. [81]

Indian Herbal Agents

The bilva (*Aegle marmelos correa*) plant from the Indian Ayurvedic system of medicine has been used to treat chronic constipation, flatulence, diarrhea, and colic. Yadav and colleagues [82] randomized patients with IBS to one of three groups: standard treatment (clidinium bromide, chlordiazepoxide, and ispaghula), herbal therapy (bilva plus the sedative herb *Bacopa monniere Linn*), or placebo. Both active treatment groups significantly improved compared with the placebo group. A subanalysis revealed the standard therapy was most beneficial for patients with pain as a predominant symptom, whereas the Ayurvedic herbs were most beneficial for those with diarrhea. At 6-month follow-up, relapse rates did not differ between the active treatment and placebo groups.

CAM USE IN INFLAMMATORY BOWEL DISEASE

Inflammatory bowel diseases (IBD) exhibit a large degree of heterogeneity, both in disease expression and in response to established therapies. The toxicity and relatively low efficacy of many standard medical therapies impel some patients with IBD to seek alternative therapies, including probiotics, nutritional supplements, and botanical products. Clinical evaluations of alternative therapies suggest therapeutic benefits, and studies of probiotic organisms and helminthic parasites provide new insights into the effects of host and environmental factor interactions on GI inflammation.

Probiotics

Probiotics are defined as “preparations or products containing viable, defined microorganisms in sufficient numbers, which alter the microflora (by implantation or colonization) in a compartment of the host and exert beneficial health effects.” [83] Probiotic bacteria stimulate nonspecific host resistance to microbial pathogens and enhance humoral immune responses to those pathogens. [84] They also alter the composition of the lumenal flora by producing lactic acid, bacteriocins, and other antimicrobial peptides that are efficacious against pathogens.

Lactobacterium species, in general, perform well against pathogenic bacteria without inhibiting other probiotic organisms. Most species adherent to the small bowel belong to the *Lactobacillus* genus and perform well as potential probiotics. *Lactobacillus salivarius* was noted to compete exceptionally well against pathogenic bacteria without inhibiting other beneficial strains of lactobacilli or bifidobacteria isolated from the human ileum. [85]

Often, GI inflammation leads to imbalances of the intestinal microflora. [86] Some patients with ulcerative colitis have high numbers of pathogenically adhesive and enterohemorrhagic *Escherichia coli* organisms. [87] The presence of pathogenic *E coli* strains is reciprocally related to the number of nonpathogenic *E coli* bacteria, [88] suggesting that augmentation of the normal flora with nonpathogenic strains may be helpful. A randomized blinded study of a nonpathogenic strain of *E coli* (Nissle 1917) [89] demonstrated efficacy equal to that of 5-acetylsalicylic acid (5-ASA) in maintaining remission of symptoms and preventing an active flare of ulcerative colitis. A larger study designed to compare rates of efficacy between therapeutic arms compared the remission rates for treatment with *E coli* (Nissle 1917), mesalazine (1.2 g/d), and a combination of both. [90] This study reported equivalent remission rates; however, large numbers of patients relapsed in all arms, perhaps because of low doses of 5-ASA in the control and combination arms. In an open-label pilot trial, patients with Crohn’s disease treated with 5-ASA were offered a novel probiotic, *L salivarius,* when they needed a change in therapy. Colonization of the GI tract by this organism promoted a systemic antibody response and, if they were elevated before treatment, a decline in TNF levels. [91] These trials suggest efficacy equivalent to that of 5-ASA; however, further clinical trials designed to test the therapeutic benefits of probiotics rigorously are needed to confirm these preliminary observations.

Studies in mice reveal that probiotic administration leads to the competitive displacement of intestinal pathogens and the engagement of cell membrane receptors that modulate signaling events for cytokine synthesis. [83] Mice bred to be deficient in IL-10 have no colonic inflammation while maintained in isolation, but they acquire colitis on exposure to environmental bacteria. On exposure to the environment, IL-10-/- mice showed an increase in the number of bacteria adhering to and translocating across the colonic mucosa in comparison with age-matched controls. After environmental exposure, colonic *Lactobacillus* levels steadily increased over 16 weeks to match control levels, but colonization in the IL-10-/- mice did occur with a different species of *Lactobacillus*. These investigators then repopulated the colons of the IL-10-/- mice by using a rectal enema containing *Lactobacillus reuteri,* followed by daily rectal swabs with the same organism. At 4 weeks, colonic levels of *L reuteri* were similar in the IL-10-/- and control mice. Repopulation with *L reuteri* was associated with a decrease in levels of anaerobic bacteria adhering to and translocating across the colonic mucosa, in addition to attenuation of the colonic inflammation. [92] In another study, intragastric instillation of a genetically altered IL-10–secreting *Lactococcus lactis* probiotic organism caused a 50% reduction in dextran sulfate–induced colitis, preventing the onset of colitis in IL-10-/- mice following exposure to environmental bacteria. [93]

Helminthic Parasites

A theory recently advanced suggests that tolerance to lumenal bacterial antigens (helper T cell type 2 [Th2] mucosal conditioning) may be conferred via exposure to helminthic parasites. [94] The decline in helminthic infestations in the United States during the past 60 years has been offered as an explanation for the rise in the incidence of Crohn’s disease during the same period. [94] A naive Th0 cell begins to secrete IL-2 and proliferate when presented with a specific antigen. With prolonged antigen exposure, the cytokine profile can polarize to either a Th1 (interferon-?, TNF-a) or Th2 profile (IL-4, IL-5, IL-10, IL-13). [95], [96] Immune response to helminthic infection generally elicits a strong Th2-type reaction, whereas patients with Crohn’s disease mount a Th1 response to lumenal antigens. Persons harboring helminths are more apt to mount a diminished Th1 response when challenged with other antigens. [94] Mice exposed to *Schistosoma mansoni* before trinitrobenzene sulfonic acid and ethanol were protected from the development of experimental colitis. [97] In IL-10-/- mice inoculated with the helminths *Heligmosomoides polygyrus* and *Trichuris muris,* far less inflammation developed than in those given a sham inoculation. [94] These data suggest a possible link between decreasing rates of helminthic infections in developed nations and the rising incidences in Crohn’s disease, and may offer another method by which immune modulation can be used to treat patients with this chronic disease.

Prebiotics

Prebiotics, described as “nondigestible food ingredients that beneficially affect the host by selectively stimulating the growth and/or activity of one or a limited number of beneficial bacteria in the colon, thereby improving the host’s health,” [98] provide an additional mechanism for influencing the population dynamics of the intestinal microflora. The most beneficial prebiotics reach the cecum, where they can promote the growth of lactic acid–producing microorganisms. Recovery of a high proportion of the chicory *Fructans inulin* and its hydrolysate, oligofructose, from ileostomy outputs supports their ability to reach the cecum. [99] Prebiotics are present in significant amounts in a wide variety of fruits and vegetables and are fermented by colonic bacteria to lactic and short-chain carboxylic acids. [100] Other oligomers with prebiotic potential are lactulose and such sugar alcohols as mannitol and xylitol. Additionally, prebiotics such as oligofructose may function in a primary antibacterial role by augmenting the growth of beneficial probiotic strains, as seen in competition experiments between strains of *E coli, Clostridium perfringens,* and *Bifidobacterium infantis*. [99]

Transforming Growth Factor-ß

Therapy with enteral nutrition has been shown to be efficacious in Crohn’s disease. A specific dietary component, transforming growth factor-ß (TGF-ß), has been studied individually for its applicability as an IBD treatment. TGFs are multifunctional polypeptides found in milk. An oral polymeric diet rich in TGF-ß was studied in 29 children with Crohn’s disease. Seventy-nine percent achieved remission within 8 weeks of therapy, with two treatment failures that required surgical intervention or steroid therapy. Laboratory indicators of inflammation decreased with therapy, and a follow-up colonoscopy revealed endoscopic improvement in about 50% of cases, with some demonstrating complete mucosal healing.

Green Tea

Green tea extracts contain high levels of antioxidants. The polyphenol fraction accounts for most of the antioxidant potential and includes the compounds (-)-epicatechin (EC), (-)-epigallocatechin (EGC), (-)-epicatechin-3-gallate (ECG), and (-)-epigallocatechin-3-gallate (EGCG). [35] Green tea polyphenols are more potent antioxidants that either vitamins E or C and have potent antiinflammatory properties. [101] Green tea polyphenols have been shown to inhibit intestinal inflammation in the IL-2 knock-out mouse model of IBD. [35] An important potential mechanism for treating IBD, independent of antioxidant properties, relates to the effects of the green tea polyphenol fraction on NF-?B, a nuclear activating protein. [102] In a dose-dependent fashion, EGCG inhibits the effects of inflammatory mediators on NF-?B by inhibiting the phosphorylation of I?B by I?B kinase (IKK). When bound to I?B, NF-?B cannot migrate to the nucleus and bind with cytokine promoter sequences,

interrupting the proinflammatory cascade. [102]

Fish Oil

Arachidonic acid metabolites, particularly leukotriene B_4 (LTB_4), are important components in the inflammatory processes associated with IBD. Arachidonic acid metabolites cause increased vascular permeability, vascular dilation, and neutrophil recruitment. Serum LTB_4 levels are increased in patients with ulcerative colitis. [103] Eicosapentaenoic acid (EPA), a component of fish oil, can reduce the formation of LTB_4 by serving as a competitive substrate for 5-lipoxygenase, thereby producing a less potent metabolite, LTB_5. Controlled trials of fish oil in patients with ulcerative colitis have demonstrated trends toward efficacy. The data of Hawthorne and colleagues [104] suggested a steroid-sparing effect from 4.5 g of EPA per day in patients with ulcerative colitis. Belluzi and colleagues [105] found that an enteric-coated preparation of 1.8 g of EPA per day maintained remission in patients with Crohn's disease. Fish oil therapy is limited by the induction of an unpleasant body odor and the need for large doses.

Nicotine

Tobacco use has a well-described dichotomous effect in IBD. Although smoking may aggravate Crohn's disease, patients with ulcerative colitis may experience disease onset or a flare of their existing disease with smoking cessation. [106] This prompted investigators to evaluate the use of nicotine supplementation in the treatment of ulcerative colitis. In a randomized double-blinded placebo-controlled study of nicotine patches (average, 17 g/d) for active ulcerative colitis, a statistically significant response was observed in comparison with placebo. [107] Two subsequent studies with the nicotine patch as monotherapy showed no statistical improvement in remission rates, [108] but a study by Sandborn and colleagues [109] did demonstrate a significant improvement in clinical parameters. Because of poor tolerance of the side effects of nicotine, nicotine patches might be considered as an alternative therapy for former smokers with ulcerative colitis who find their disease exacerbations difficult to control with standard medications.

Soybean Derivatives

Two soybean derivatives have shown promise as antiinflammatory agents in laboratory animals. Genistein, a principal soy isoflavone, induced a mild improvement in endoscopic findings in guinea pigs with a chemically induced colitis. [110] A soybean extract with a high concentration of a potent protease inhibitor (Bowman-Birk inhibitor) attenuated the colitis caused by dextran sodium sulfate in mice. [111] Researchers at the University of Pennsylvania are exploring the efficacy of Bowman-Birk inhibitor in humans with ulcerative colitis. [101]

Short-Chain Fatty Acids

Colonic aerobic bacteria act to salvage energy for the host by fermenting undigested food materials to short-chain fatty acids, which in turn are absorbed by colonocytes. [92] Butyrate has been shown to inhibit the production of cytokines as well as the activation of NF-?B. [112] Initial trials of short-chain fatty acids demonstrated efficacy, but subsequent trials showed mixed results, possibly because of poor compliance. An open-label study investigated the effects of *Plantago ovata* (an alternative source of butyrate) on 12-month remission rates, either alone or in combination with standard 5-ASA therapy. Remission rates were similar for the groups. [112] A subsequent abstract evaluated butyrate yields from a variety of fibers, comparing *P ovata* seeds, husks, and other forms of fiber. All produced significant amounts of butyrate, but butyrate production from the seeds trended toward a higher output. [113]

CAM USE IN DIARRHEAL STATES

Antibiotic-associated diarrhea occurs in approximately 20% of hospitalized patients receiving antibiotics, with up to 33% of cases related to *Clostridium difficile*. [114], [115] The other 66% experience symptoms related to alterations in the bacterial production of short-chain fatty acids. [116] Antibiotic-associated diarrhea was reduced from 27% in a placebo group to 9% in a group taking the probiotic *Enterococcus faecium*. [117] Other organisms shown to be efficacious in antibiotic-associated diarrhea include *Lactobacillus* GG, *Lactobacillus acidophilus* and *Lactobacillus bulgaricus* administered in combination, and *Saccharomyces boulardii*. [116] Worldwide, infectious diarrhea remains a leading cause of morbidity and mortality in infants and children. Numerous studies have investigated the potential of many probiotic and prebiotic agents for the treatment of these conditions. Trials with *Lactobacillus* GG and *S boulardii* used as prophylactic therapy for diarrheal infections demonstrated small overall reductions in diarrhea in comparison with placebo, although some subgroups experienced particularly substantial benefit. The evaluation of probiotics as abortive agents for diarrheal illnesses has been carried out mainly in children, who are at the greatest risk for serious illness and death. *E faecium* and *S boulardii* relieve symptoms in acute diarrheal illnesses, and *Lactobaccillus* GG decreases the rate of rotaviral diarrhea and viral shedding. [118]

Traditional remedies for diarrhea and various intestinal disorders include the ingestion of pectin and green bananas, which serve as a supply of amylase-resistant starch. [119] These starches pass through the small intestine undigested, so that they can undergo fermentation in the colon into short-chain fatty acids. [120] The presence of short-chain fatty acids in the colon stimulates salt and water absorption and also has a trophic effect on colonic and small bowel mucosa. [121] When dietary pectin and green bananas were blended into a rice-based diet and used to treat children with persistent diarrhea resulting from a variety of infectious and noninfectious causes, a statistically significant reduction in stool weight and relief of symptoms were noted in the treated children in comparison with the children on control diets. [119]

SAFETY ISSUES

The safety of CAM agents is not well defined, in large part because of a lack of regulatory guidelines. In 1994, Congress passed the Dietary Supplement Healthy and Education Act (DSHEA), which exempted dietary supplement producers from many Food and Drug Administration (FDA) regulations. Consequently, dietary supplements are under less government scrutiny than over-the-counter nonprescription drugs. Thus, it is critical that both consumers and health care workers understand that the labeling of CAM products is not necessarily correct, that products may not be pure, and that the advertised dose may not be correct.

Concerns about safety relate to both product content and potential harm. Multiple studies have documented great variability in the composition of herbal products/supplements, with some products containing only a small fraction of the advertised dose. [122], [123] and [124] This product variability is probably an unavoidable aspect of many Chinese herbal compounds, and traditional prospective clinical trials difficult are difficult to design because of the inherent variability in the therapeutic agent. Toxicity induced by CAM products has been reported. [123], [125] Hepatotoxicity is one of the most frequently reported side effects of CAM products (e.g., germander and pyrrolizidine alkaloid–containing compounds), sometimes leading to liver transplantation or death. [125], [126] and [127] Unfortunately, few prospective studies have evaluated the incidence/prevalence of toxicity. In a retrospective study evaluating liver enzymes in all patients receiving Chinese drug therapy in a German hospital, clinically important liver enzyme elevations occurred in about 1% of patients. [128] In all patients, liver enzyme levels improved or normalized after discharge, even though almost half of these patients continued to take their Chinese medications. Because this was a hospital-based study, the herbal medications may have been more carefully scrutinized than they are in general practice.

Some CAM products are contaminated with drugs/toxins. [122] A classical example is the report of a young woman who presented with nausea, vomiting, and an irregular heart beat after she initiated an oral regimen of dietary supplements for "internal cleaning." The patient was found to have an elevated digoxin level, and her herbal medications were contaminated with plant extracts containing a cardiac glycoside (*Digitalis lanata*). [129] Importantly, some Chinese patent medicines purposely contain small amounts of prescription drugs (e.g., antibiotics, ephedrine, methyltestosterone). [122], [130] These drugs may also be contaminated with heavy metals (e.g., lead, arsenic, mercury). [131] CAM agents can interact adversely with traditional medications. An example is the potential interactions of St. John's wort with cyclosporine and indinavir. [132] This interaction was highlighted when cardiac transplant patients underwent rejection shortly after starting St. John's wort, presumably because of the induction of cytochrome P450 3A4 by the St. John's wort. [133] It is important to instruct all transplant patients (and other patients on immunosuppressive therapy) to inform their physician before starting any CAM agent. Other more common side effects can also be seen with relatively safe agents such as St. John's wort, including photosensitivity, especially in virally infected patients. A variety of herb-anesthesia interactions are possible (e.g., garlic, gingko, St. John's wort), such as bleeding, and the American Society of Anesthesiologists recommends that patients discontinue all herbal medicines 2 to 3 weeks before elective surgery and inform their physicians about their CAM use. [134] Lastly, the public can be easily misinformed about CAM agents and inadvertently take an unintended product with a

subsequent adverse outcome.

CONCLUSION

In conclusion, CAM is already being widely used by our patients. Some forms of CAM therapy are based on substantial scientific rationale (e.g., SAMe for liver disease, certain green tea polyphenols as antiinflammatory therapy). Some forms of CAM have yielded very positive clinical trial data (e.g., acupuncture for nausea and vomiting). However, because a great potential for toxicity/drug interactions also exists, both the patient and health care provider need to be optimally educated about CAM. In addition to the previously mentioned versions of *Physicians' Desk Reference,* the Web sites of the Cochrane Database System Reviews (http://www.cochrane.org/) and the NCCAM (http://nccam.nih.gov/) are valuable sources for updated information.

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CHAPTER 58

H. Worth Boyce, Jr. and Gregory A. Boyce

ESOPHAGUS: ANATOMY AND STRUCTURAL ANOMALIES

Clinicians who assume responsibility for the diagnosis and treatment of esophageal disorders must have a knowledge base of esophageal anatomy. Recognition of pathological alterations in their earliest stages and optimal interpretation of every test of esophageal function and morphology can be accomplished only through awareness of normal anatomy and its variants. As understanding of the manifestations of esophageal disease advances, a knowledge of esophageal anatomy will be increasingly recognized as essential to clinical success.

EMBRYOLOGY

During the fourth week of embryologic development, a small diverticulum forms on the ventral surface of the foregut adjacent to the pharyngeal gut. This tracheobronchial diverticulum separates gradually from the dorsal foregut through the esophagotracheal septum, with eventual separation into the trachea and esophagus. [1] With cranial growth of the embryonic body, the esophagus elongates rapidly. During the sixth week of gestation, the circular muscle coat and ganglion cells of the myenteric plexus form. During the seventh week, blood vessels enter the submucosa.

The esophageal epithelium rapidly proliferates and almost completely fills the lumen in the seventh and eighth weeks, leaving residual channels within the nearly occluded lumen. [2] A single esophageal lumen returns in the tenth week, leaving a superficial layer of ciliated epithelial cells.

In the fourth month, ciliated epithelial cells are replaced by stratified squamous epithelium, a process that continues until birth. Residual islands of ciliated epithelium at the proximal and distal ends of the esophagus remain and give rise to esophageal glands. [3]

ADULT ANATOMY

Gross Anatomy

The adult esophagus is a flattened muscular tube that arises proximally at the pharyngoesophageal junction (i.e., C5-6 vertebral interspace) and courses through the posterior mediastinum to end at the gastroesophageal junction (i.e., T10 vertebral level). The esophageal lumen can distend to approximately 2 cm in anteroposterior diameter and up to 3 cm in lateral diameter. The length of the adult esophagus is variable but ranges from 18 to 26 cm. [4] The cervical esophagus extends from the pharyngoesophageal junction to the suprasternal notch and is about 4 to 5 cm long. At this level, the esophagus is surrounded by the trachea anteriorly, the vertebral column posteriorly, and the carotid sheaths and thyroid laterally.

The thoracic esophagus passes just posterior to the tracheal wall and courses right posterior to the aortic arch (i.e., T4 vertebral level) and posterior to the tracheal bifurcation and left main stem bronchus. At the T8 vertebral level, the esophagus turns left and crosses anterior to the aorta at the level of the diaphragmatic hiatus. At the T10 vertebral level, the esophagus passes through an elliptical opening (hiatus) in the muscular diaphragm and enters into the cardiac portion of the stomach at an oblique angle.

The abdominal portion of the esophagus, or the submerged segment, is short, varying in length from 0.5 to 2.5 cm. [5] At this level, the left lobe of the liver lies anteriorly, the caudate lobe of the liver lies to the right, the fundus of the stomach is to the left, and the right crus of the diaphragm and aorta lie posteriorly. The borders of the esophageal hiatus are formed by the diaphragmatic crura and median arcuate ligament, if present. The crura arise from the first four lumbar vertebrae, intervertebral discs, and anterior longitudinal ligament. The fibers of the left and right crura pass upward and anteriorly to form the muscle borders of the hiatal ring and then insert into the transverse ligament of the central tendon of the diaphragm. [6] The crural origin of the muscle margins of the esophageal hiatus is variable. [7], [8] and [9]

At the level of the diaphragm, the esophagus is surrounded by collagen and elastic fibers of the phrenoesophageal membrane. This membrane extends from the hiatal margin to insert into the circumference of the esophagus both above and below the diaphragm. [10], [11] It is most pronounced in infants. With age, the esophagus is less firmly fixed to the hiatus, and fat appears between the fibers. [12] This membrane is lacking in patients with long-standing hiatal hernia. [13]

The esophageal mucosal surface is rather homogeneous in color and topography. The color is a pinkish gray from the cricopharyngeus to the squamocolumnar junction (Fig. 58-1). Small, linearly oriented mucosal vessels may be seen on close inspection with good illumination. These and many other vessels are apparent when the esophagus is distended, as in achalasia. In achalasia, the vascular pattern of the esophagus is difficult to differentiate from that of the rectum. The squamocolumnar mucosal junction is detected easily in normal subjects by the abrupt disappearance of this vascular pattern and the dramatic color change to the reddish orange, slightly granular mucosa of the gastric cardia. [14] This mucosal junction normally is located at or below the level of the diaphragmatic hiatus.

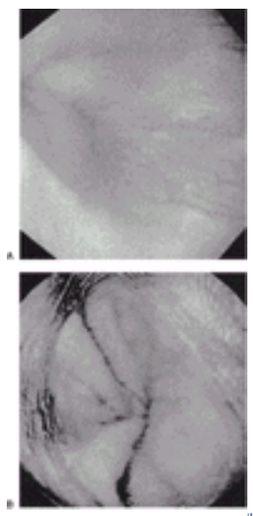

FIGURE 58-1. (See Color Fig. 58-1.) **A:** This view of the normal squamocolumnar junction reveals a slightly serrated contour, a distinct color difference between the two types of mucosa, and linear esophageal vessels that disappear at the mucosal junction. The slightly elevated gastric mucosal folds contact the squamous margin. **B:** The squamocolumnar junction has been better defined by dark Lugol iodine staining of the glycogen-containing squamous cells. This normal junction is located just below the diaphragmatic hiatus.

The esophagogastric muscular junction is represented intralumenally by the cephalic margin of the longitudinal mucosal folds in the gastric cardia. [14], [15] These folds are best seen when a hiatal hernia is present. [14]

Blood Supply

The arterial blood supply to the esophagus is largely segmental, with little vascular overlap (Fig. 58-2). The cervical esophagus is supplied mainly by branches of the inferior thyroid artery. Branches of other arteries, such as the common carotid, subclavian, vertebral, and ascending pharyngeal, may provide additional blood supply. The thoracic esophagus is supplied by branches of the aorta and the right intercostal and bronchial arteries. The abdominal esophagus is supplied by branches of the left gastric, short gastric, and left inferior phrenic arteries. Devascularization and ischemia of the esophagus are of concern during resection operations because of the segmental nature of the blood supply.

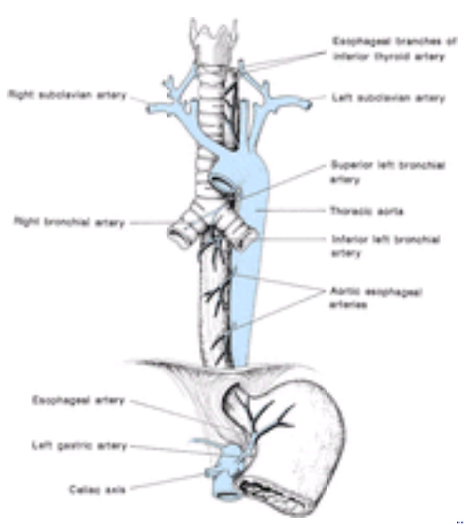

FIGURE 58-2. Arterial system of the esophagus.

The venous anatomy of the esophagus has been well defined (Fig. 58-3). Fine intraepithelial channels drain into a subepithelial superficial venous plexus. This plexus drains into deep intrinsic veins in the submucosa. At the level of the gastroesophageal junction, the superficial venous plexus and deep intrinsic veins communicate with their gastric counterparts. [16]

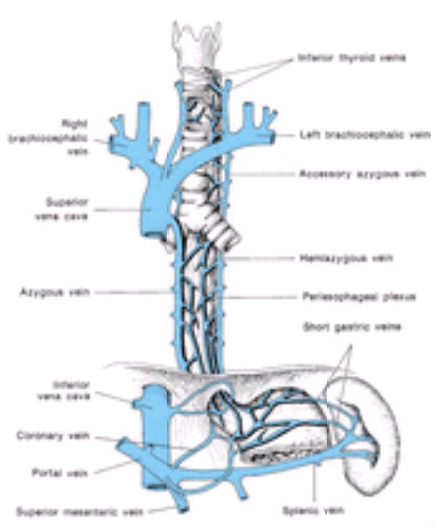

FIGURE 58-3. Venous drainage of the esophagus.

Perforating veins connect the deep intrinsic veins to adventitial veins. At the level of the cervical esophagus, the adventitial veins drain into the inferior thyroid vein, deep cervical vein, vertebral vein, and peritracheal venous plexus.

At the thoracic level, adventitial veins drain into the azygous vein on the right, the hemizygous vein on the left, and the intercostal veins when the hemizygous vein is absent. At the gastroesophageal junction, the portal systemic circulation involves venous drainage of the esophagus, stomach, pancreas, spleen, diaphragm, and retroperitoneum. Anatomic studies have suggested a high-pressure watershed region between the portal and azygous systems at the region of the gastroesophageal junction that is prone to venous dilation in portal hypertension. [17] In portal hypertension, the deep intrinsic veins in the submucosa and subepithelial superficial venous plexus dilate, protrude into the lumen, and may markedly stretch and thin the epithelial surface, forming esophageal varices. [16] In patients with portal hypertension, these vessels serve as collateral channels to provide a route of return for portal blood to the systemic circulation. If pressure and flow increase sufficiently, these dilated veins, or varices, may spontaneously rupture and lead to severe bleeding. The precise pathophysiology of variceal bleeding is not known, but the explosion theory (i.e., rupture from high pressure) seems to be favored over the erosion theory (i.e., mucosal destruction by refluxed gastric contents). Variceal bleeding is from the distal 6 to 8 cm of the esophagus in nearly all cases. Varices may extend up to about the aortic arch level, the upper limit of the venous drainage of the lower esophagus by way of the azygous vein system into the superior vena cava. Varices in a location cephalic to this level have been termed *“downhill” varices*. The

“downhill” description refers to the anatomic site of venous obstruction above the level of the varices, in either the cervical venous system or the superior vena cava. The varices then develop below the site of venous obstruction, whereas typical distal esophageal varices develop in a location cephalic to or above the portal venous flow.

Innervation

The vagus nerve supplies only parasympathetic innervation to the esophagus, although below the neck, the vagus nerve carries a mixture of parasympathetic and sympathetic nerve fibers (Fig. 58-4). The cervical esophagus is innervated by the recurrent laryngeal nerves, which arise from the vagus. Branches of the vagus nerves and the left recurrent laryngeal nerve innervate the upper thoracic esophagus. The left and right vagus nerves intertwine with sympathetic fibers to form the esophageal plexus. [18] , [19] Out of the esophageal plexus, the anterior and posterior vagus trunks form at a variable distance above the diaphragm. [18] Below the diaphragm, the anterior (i.e., left) vagus trunk splits into anterior gastric branches and hepatic branch. The posterior (i.e., right) vagus trunk splits into posterior gastric branches and a branch to the celiac plexus. Sympathetic innervation is supplied by the superior cervical ganglion, sympathetic chain, major splanchnic nerve, thoracic aortic plexus, and celiac ganglion.

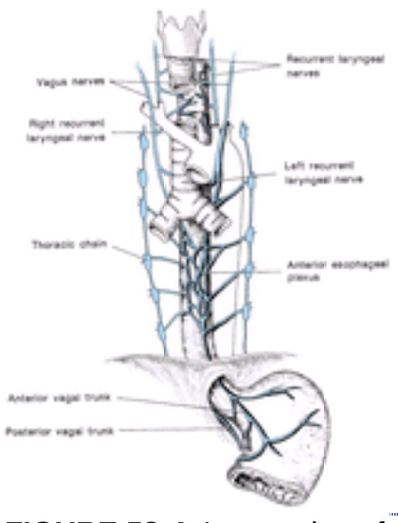

FIGURE 58-4. Innervation of the esophagus.

Lymphatics

A dense network of lymph vessels is found in the mucosa and submucosa of the esophageal wall (Fig. 58-5). These vessels travel a variable distance longitudinally before penetrating the muscular wall and draining into adventitial lymph nodes. The deep cervical lymph nodes drain the proximal esophagus. At more distal levels, the lymph vessels drain into the adjacent lymph node chain. Internal jugular, tracheal, tracheobronchial, posterior mediastinal, and pericardial nodes drain adjacent esophageal segments. Unlike the arterial supply, the lymphatic drainage of the esophagus is not segmental. Multiple interconnections exist between nodal chains. This arrangement accounts for the frequent wide intramural and mediastinal lymphatic spread of esophageal carcinoma.

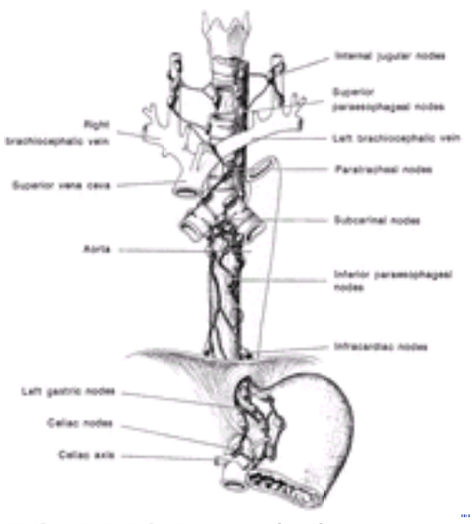

FIGURE 58-5. Lymphatic system of the esophagus.

HISTOLOGY

Light Microscopy

The esophageal wall is composed of four layers: mucous membrane, or tunica mucosa; submucosa, or tunica submucosa; muscularis externa, tunica muscularis, or muscular propria; and adventitia, or tunica adventitia (Fig. 58-6). [20] , [21] The absence of a serosal layer allows esophageal malignancies to spread readily and makes esophageal anastomosis and surgical repair more difficult.

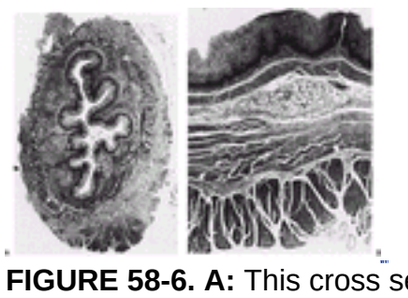

FIGURE 58-6. A: This cross section (×2.5) from the middle third of the esophagus has a mixture of skeletal and predominantly smooth muscle in the muscularis propria. The submucosal glands are clearly shown. At one point, there is an esophageal cardiac gland in which a small focus of glandular epithelium interrupts the squamous mucosa. This is a normal finding, seen in at least 1% of all esophagi. **B:** Longitudinal section of esophageal wall (×10). (Courtesy of Rodger C. Haggitt, M.D., Seattle, WA.)

The innermost layer, the mucous membrane, is composed of nonkeratinized squamous epithelium supporting connective tissue called the *lamina propria* and a thin

layer of smooth muscle called the *muscularis mucosae*. The squamous epithelium is composed of a basal cell layer known as the *stratum basale,* a prickle cell layer known as the *stratum intermedium,* and a superficial layer known as the *stratum superficiale*. The inner border of the epithelium is irregular because of protrusions of the lamina propria called *dermal papillae* or *rete pegs*. The basal cell layer is composed of basophilic, cylindrical cells that have the capacity to divide and replenish the superficial layers. [20]

The esophagus contains cells that are a part of the gut-associated lymphoid tissue. Cytotoxic T cells (i.e., intraepithelial lymphocytes) and Langerhans cells (i.e., macrophages) are found in the squamous epithelium. [22] Helper T cells and B lymphocytes are seen primarily in the lamina propria. [20] Intraepithelial melanocytes and argyrophil cells can be found in the basal cell layer. [23]

The muscularis mucosae is composed of smooth muscle cells that separate the lamina propria from the submucosa (see Fig. 58-6). The submucosa consists primarily of loose connective tissue. The vascular network known as the *Heller plexus,* a nerve plexus, mucin-secreting glands, lymph follicles, and lymphocytes are located at the level of the submucosa.

Striated muscle fibers of the inferior pharyngeal constrictor and cricopharyngeus muscles overlap with striated circular muscle fibers of the cervical esophagus at the level of the C5-6 vertebral interspace. This corresponds to the level of the physiological upper esophageal sphincter segment, of which the cricopharyngeus appears to be the major component. [24], [25], [26] and [27]

The muscular wall of the esophagus is composed of an inner circular and an outer longitudinal layer, the inner circular layer being the thicker of the two (see Fig. 58-6). The first centimeter of the proximal esophagus is striated muscle alone, the muscle of the next 6 to 8 cm is mixed (i.e., both striated and smooth), and the remaining length is all smooth muscle. In situ, longitudinal muscle fibers run in an elongated spiral. Circular muscle fibers run in an elliptical course, with some fibers leaving their bundle to join higher or lower bundles. [27]

Below the diaphragm and proximal to the angle of His (i.e., the abdominal or submerged segment), an area has been described in fixed gastroesophageal specimens in which the inner circular muscle layer thickens and the fibers become semicircular and interlaced. Oblique fibers of gastric type from the greater curve are also found. [28], [29] It has been suggested that this area corresponds to the physiological lower esophageal sphincter, but by most observations the sphincter lies proximal to this level just cephalic to the diaphragmatic hiatus. [14], [15]

The myenteric plexus, or Auerbach plexus, is found between the inner circular and outer longitudinal muscle coats. The adventitial layer consists of connective tissue with networks of nerve plexus, vascular structures, and elastic fibers. Other specialized elements can be seen in the esophageal wall. Islands of gastric mucosa, sebaceous glands, taste buds, and foci of hyperplastic epithelial cells with intranuclear glycogen (i.e., glycogenic acanthosis) have been described. [30], [31], [32] and [33] The latter condition is commonly seen during video-esophagoscopy as focal white elevations, several millimeters in diameter, scattered randomly at any level of the esophagus, and it has no clinical significance. Glycogenic acanthosis stains more intensely with Lugol iodine chromo-endoscopy than the surrounding typical mucosa.

Electron Microscopy

With electron microscopy, basal cells appear as oblong or cuboidal structures with central nuclei. The cytoplasm contains mitochondria, small Golgi complexes, free ribosomes, lysosomes, and little endoplasmic reticulum. No glycogen is seen in these cells. As these cells mature and leave the basal layer, they become larger and more flattened, with cellular constituents similar to those of basal cells. They constitute the prickle cell layer, or stratum intermedium. Glycogen is present in these cells, and membrane-coating granules believed to play a role in cell cohesion are seen in the superficial prickle cell layer. [34], [35] The presence of glycogen in the superficial mucosal cells accounts for their brownish black staining by Lugol iodine solution as applied for chromo-endoscopy. In the superficial layer, the squamous epithelial cells are more flattened and oriented parallel to the surface. The cell membrane becomes more prominent, and the cell edges may overlap. Membrane-coating granules are present. Acid and neutral mucosubstances are found on all layers of epithelial cells. Acid mucosubstances are present in larger amounts on superficial cells and may play a protective role. [36], [37] Microplicae seen on surface cells by scanning electron microscopy may function in holding the mucus in place. [38]

With electron microscopy, the submucosal nerve plexus is seen as an irregular network near the inner coat of the muscularis externa. [39] In the lower esophageal sphincter region, nerve endings are seen as multiple varicosities with close contact to differentiated smooth muscle cells (i.e., Cajal cells) that play a role in the initiation and coordination of contraction. [40]

DEVELOPMENTAL ANOMALIES

Tracheoesophageal Fistula and Atresia

During embryogenesis, the process of elongation and separation of the trachea and esophagus can be disrupted. If fusion of the tracheoesophageal septum is incomplete, a tracheoesophageal fistula (TEF) results. If elongation outstrips foregut cell proliferation, ventral or dorsal cells may form tracheal tissue and esophageal atresia may develop, [1] either with or without associated TEF. Five basic types of TEF and atresia have been described (Fig. 58-7). Esophageal atresia with lower-pouch fistula is by far the most common. [41], [42], [43], [44], [45] and [46] Hydramnios and prematurity are common in infants with atresia or TEF. [47], [48] and [49] Up to 50% of infants may have other associated congenital anomalies. [49]

FIGURE 58-7. The spectrum of esophageal atresia, tracheoesophageal fistula, and congenital stenosis. Atresia with lower pouch fistula (third from left) is by far the most common anomaly.

Symptoms vary with the type of tracheoesophageal anomaly. In infants with atresia alone, the diagnosis is often made after birth. Unswallowed saliva fills the mouth and nostrils, and formula is regurgitated. Infants with proximal fistula exhibit respiratory distress and cyanosis during feedings. In infants with atresia and distal fistula, excessive salivation and regurgitation occur along with cyanosis and pneumonia secondary to reflux of gastric contents. If both proximal and distal fistulae are present, the proximal fistula usually is the cause of most symptoms. The fifth type, isolated TEF or H-type fistula, leads to cough and choking with feedings, recurrent pneumonia, and intermittent abdominal distention. In rare cases, this anomaly is initially diagnosed in adulthood. Adult patients present with a history of recurrent pneumonia and bronchiectasis. [50], [51], [52] and [53]

TEF suspected by the clinical signs and symptoms and can be diagnosed by cautiously introducing radiographic contrast through a catheter. This will delineate the atretic segment and the location of the fistula. In patients with H-type TEF, repeated contrast examinations may be necessary before the fistulous tract is demonstrated. [54]

The surgical trend has been toward early primary fistula repair, with staged repair reserved for infants with respiratory distress or severe associated congenital anomalies. Repeated bougienage may be used to lengthen the atretic segments, allowing subsequent primary anastomosis. [55], [56] Delayed colon interposition is used if primary anastomosis is impossible. The primary determinants of survival are the coexistence of other congenital anomalies and the severity of associated pulmonary disease. [57], [58]

Significant complications may occur after a successful repair of esophageal atresia. [59]

Congenital Esophageal Stenosis

Congenital esophageal stenosis is rare, estimated to occur once in 25,000 live births (see Fig. 58-7), and is thought to result from failure of the normal embryonic separation of trachea and esophagus. [60]

Little information is available about possible pathophysiological mechanisms that contribute to congenital esophageal stenosis. One study of the histological and immunohistochemical features found in esophagi from two young adults has provided several unique observations. [61] Quantitative experiments compared the numbers of myenteric neurons and amount of fibers in the circular muscle. Congenital esophageal stenotic esophagi showed infiltration of neutrophils in the myenteric plane, without any increase in collagen. NADPH-diaphorase histochemistry showed a significant reduction of myenteric nitrinergic neurons (7 ± 2.3 versus 2.7 ± 1.8 neurons per high-power field) and fibers at the circular muscle. Other peptidergic neurons studied were not significantly reduced in congenital esophageal stenosis. The specific total lack of oxide inhibitory innervation may be an important mechanism in the pathogenesis of stenosis and aperistalsis of the esophagus in the disorder. [62]

Stenoses caused by cartilage, residual respiratory epithelium, and muscular wall maldevelopment have been described (Fig. 58-8). [62], [63], [64] and [65] Unlike atresia and TEF, congenital stenosis often is not diagnosed until later in childhood, and several cases have been reported in adults. [66], [67] and [68] Symptoms include regurgitation, prolonged eating time, and dysphagia with recurrent solid bolus impaction. An esophagram usually demonstrates segmental stenosis in the middle third of the esophagus. With adequate insufflation during endoscopy or by barium during contrast esophagrams, the appearance of multiple rings with normal overlying mucosa can be seen in most cases (see Color Fig. 15-11 in *Atlas of Gastroenterology,* 3rd ed.). Segmental resection has been advocated for these patients; however, properly performed bougienage has been reported to be safe in children and adults. [60], [68] Esophageal stenosis may result when tracheobronchial remnants remain within the esophageal wall after repair soon after birth. [69]

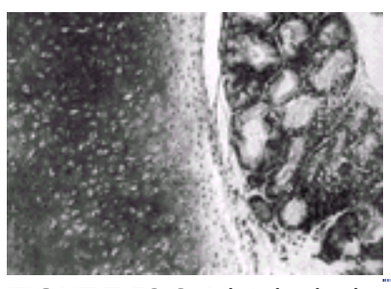

FIGURE 58-8. Histological section from the esophageal wall of a resected segment of congenital stenosis. Note the cartilage (**left**) and tracheal glands (**right**). (From Ishida M, Tsuchida Y, Saito S, et al. Congenital esophageal stenosis caused by tracheobronchial remnants. J Pediatr Surg 1969;4:340.)

Congenital Esophageal Duplication

Congenital esophageal duplications, tubular or cystic, represent about 15% of digestive duplications. They develop in the third week of embryonic development because of failure of vacuoles to coalesce properly, preventing recanalization of the esophageal lumen. As a result, a cyst or parallel tubular channel forms within the esophageal wall. [70] Duplications of the gastrointestinal tract have three common characteristics: (1) They are contiguous with some segment of the gastrointestinal tract; (2) they are lined by alimentary epithelium; and (3) smooth muscle is present in their walls. Tubular duplications are rare and may be associated with other congenital abnormalities, mainly of the spine. [71] They may be asymptomatic or present with dysphagia. Tubular duplications may communicate at both ends with the esophageal lumen or be closed at one end, [72], [73], [74] and [75] but more than 80% are cystic and do not communicate with the lumen. Most patients present before 1 year of age, but as many as 25% to 30% of cases are first diagnosed in adults. The most common presenting symptom is dysphagia, but the diagnosis is most often made as an incidental finding in patients studied for other reasons. [71] The presence of a posterior mediastinal, soft tissue mass on the chest x-ray film is the usual finding.

Congenital Duplication Cyst

Cystic duplications are less rare; they constitute 0.5% to 2.5% of esophageal tumors. They are the second most common benign esophageal tumor, with leiomyoma being the most common. [76], [77] They occur within the wall of the esophagus and are surrounded by two layers of smooth muscle. They are lined by squamous columnar, cuboid, pseudostratified, or ciliated epithelium. [78], [79] Sixty percent arise in the distal third of the esophagus, 17% in the middle third, and 23% in the upper third. [80], [81]

Cysts in the distal two thirds of the esophagus may result in symptoms of dysphagia, epigastric discomfort, retrosternal pain, cough, dyspnea, or regurgitation, although many are diagnosed without being symptomatic. [82] Cysts located posterior to the heart have been associated with cardiac arrhythmias. [83] Intraspinal cystic extensions can cause neurological deficits that may be the initial sign of these disorders. [84] In one report, intraspinal anomalies coexisted with mediastinal masses in almost 25% of patients, and these anomalies were often asymptomatic initially. [85]

Gastric cysts, inclusion cysts without a smooth muscle wall, bronchogenic cysts, and neuroenteric cysts also rarely are found in the esophageal wall. [80]

Congenital duplication cysts can be seen on chest radiographs as posterior or middle mediastinal masses. On barium esophagram, a smooth, curved displacement of the esophagus is seen without the sharp, steplike proximal and distal margin seen with a leiomyoma.

Computed tomography can be helpful in determining the location, size, and anatomic relation to other organs. [86], [87] At endoscopy, a soft, compressible bulge into the esophageal lumen without overlying mucosal abnormality can be seen. [88] Endoscopic ultrasonography can define the structure of a duplication cyst. [89]

Surgical resection is usually recommended for definitive treatment and pathological diagnosis. [90], [91] Marsupialization has been used for the treatment of large cysts, and needle aspiration has been used to relieve tracheal compression from a duplication cyst. [92], [93] Although rare, the development of malignancy within a tubular or cystic duplication has been reported. [94], [95] and [96]

Bronchopulmonary Foregut Malformation

Bronchopulmonary foregut malformations are pulmonary sequestrations with a patent congenital communication to the upper gastrointestinal tract. [97], [98], [99], [100] and [101] Bronchopulmonary foregut malformations develop when cell rests with respiratory potential arise from the esophagus caudal to the lung bud or when a portion of the lung bud arises from the dorsal esophagus rather than the ventral trachea. [98] The tract within the sequestered pulmonary lobe typically involutes because of outgrowth of its blood supply; incomplete involution of the tract leads to a gastrointestinal tract communication. [99] Bronchopulmonary foregut malformations are most commonly seen in the lower lobes. The arterial supply and venous drainage are variable. [100], [101] Up to 40% of children with communicating bronchopulmonary foregut malformations have associated congenital anomalies. [102] The clinical presentation in infants is respiratory distress that is exacerbated with feedings. Congestive heart failure also may occur. In adults and older children, recurrent pneumonia, bronchiectasis, hemoptysis, gastrointestinal bleeding, and dysphagia may develop. [100] Contrast esophagram, bronchography, and angiography are used for diagnosis and surgical planning.

Aortic Arch Vessel Abnormalities

It has been estimated that 3% of the population have a congenital abnormality of the aortic arch vessels, but only rarely does this result in symptomatic compression of the esophagus. [103] In the embryo, the foregut is surrounded by vascular structures of the branchial arches. Normally, portions of the branchial arches obliterate to form the great vessels and aortic arch. Abnormalities in developmental obliteration of the branchial arches may lead to vascular compression of the trachea and esophagus. [104]

The term *dysphagia lusoria,* literally translated from the Latin *lusus naturae* ("trick of nature"), [105] is used to describe symptomatic esophageal compression resulting from any vascular anomaly of the aortic arch, but it usually results from an aberrant right subclavian artery. Symptoms of this anomaly may occur at the onset of semisolid feedings, later in childhood, or in adult life. With this anomaly, the right subclavian artery arises from the left side of the aortic arch and courses obliquely upward and posteriorly, compressing the esophagus (Fig. 58-9 *A*). It is estimated that aberrant right subclavian artery occurs in 0.5% to 1% of the population, with only 10% of these affected individuals having symptoms related to compression. [106] During esophageal endosonography performed in 3334 patients, an aberrant right subclavian artery was identified in 12 (0.36%). None of the patients had symptoms of this entity. [107] This is the vascular anomaly that most typically causes symptoms later in life, [108] although most patients with this condition do not have symptoms.

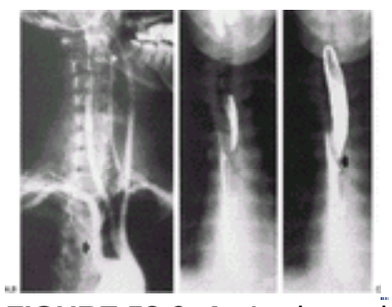

FIGURE 58-9. A: Angiography reveals an aberrant right subclavian artery (*arrow*) arising from the aortic arch. **B:** Barium esophagram in the same patient reveals oblique esophageal compression (*arrow*) by the aberrant right subclavian artery posterior to the esophagus.

Barium esophagram demonstrates an oblique filling defect just above the level of the aortic arch (see Fig. 58-9B). Aortic arch angiography has been the standard for diagnosis, although computed tomography and magnetic resonance imaging may be used for more precise delineation of this anomaly. Magnetic resonance imaging is particularly sensitive for this disorder and permits examination in both coronal and sagittal planes. Endoscopy may demonstrate a posterolateral pulsatile compression of the lumen. The right radial pulse may be weakened or obliterated by endoscopic compression of the vessel or during bougienage. [109], [110] Correction is performed in children by division and ligation of the aberrant artery. In adults, reanastomosis to the ascending aorta is performed to avoid the development of a subclavian steal syndrome. [111], [112] For patients with intractable dysphagia, a cervical approach has been reported to be successful, with ligation of the aberrant artery near its root and then connected to the right carotid artery. [113] Surgery is not always needed because many adult patients can easily tolerate minor degrees of dysphagia by simply modifying their diet. Asymptomatic persons not recognized to have this vascular anomaly pose a potential problem to the surgeon who operates on the neck, especially during a thyroidectomy. [105] In these patients, the inferior laryngeal nerve is displaced from its usual position and can easily be injured if not carefully identified.

Esophageal compression by an anomalous vertebral artery and right aortic arch with constricting left ligamentum arteriosum has been reported in adults and can be successfully repaired surgically. [114], [115] Other vascular anomalies are causes of tracheoesophageal compression in infants, and these require early surgical intervention. [116], [117]

Heterotopic Gastric Mucosa

Heterotopic gastric mucosa (inlet patch) can be seen occasionally in the esophagus during careful endoscopic inspection. Autopsy studies of infants and children indicate the prevalence of ectopic gastric mucosa as 7.8%. The majority of these are in the proximal one third of the esophagus. [118] Endoscopic studies reveal a prevalence of 2.8% to 10% of consecutive endoscopies. [119], [120] and [121] Inlet patches have been reported to occur in 5.6% of patients with Barrett esophagus but in none with achalasia. [122] It is provocative and interesting to speculate that this relationship has a pathogenetic significance.

At endoscopy, these areas of gastric mucosa typically are small, distinct patches of reddish orange or salmon-colored mucosa in the proximal 3 cm of esophagus (see Color Fig. 15-5 in *Atlas of Gastroenterology,* 3rd ed.). One report describes a patient with heterotopic mucosa in the left piriform sinus and cervical esophagus associated with a fistula. [123] This heterotopic mucosa, referred to as an *inlet patch* if it is found in the cervical esophagus, [119] can occur throughout the esophagus and can have a linear appearance. [124] Inlet patches can be dramatically demonstrated by injecting topical dilute iodine solution (i.e., Lugol) through an endoscopic cannula to stain the squamous mucosa around their margin. On biopsy, corpus-fundic or antral-type mucosa is seen, sometimes containing parietal cells capable of acid secretion. [125] Complications of TEF and adenocarcinoma arising from heterotopic gastric mucosa have been reported. [126], [127] and [128] An apparently increased risk for peptic esophagitis has been noted in biopsy specimens taken from squamous mucosa adjacent to these patches of columnar epithelium, some of which contain parietal cells. The degree of histological change typical of esophagitis appears to be inversely related to the distance from the margin of columnar epithelium at which the specimen is taken. [129] Other reported complications include cervical webs or rings. [130] Transendoscopic thermal ablation of an inlet patch combined with high-dose omeprazole therapy has been shown to allow replacement of the inlet patch by normal squamous mucosa, with resolution of related symptoms. [131]

Esophageal Rings and Webs

Mucosal Rings The lower esophageal mucosal ring, or "B" ring, was initially described by Templeton in 1944. [132] In 1953, Ingelfinger and Kramer [133] and Schatzki and Gary [134] independently described the association of lower esophageal mucosal rings with dysphagia. Lower esophageal mucosal rings are located at the level of the squamocolumnar mucosal junction. These rings consist of mucosa and submucosa and are covered by squamous mucosa on the proximal side and either columnar mucosa or several millimeters of squamous mucosa on the distal or gastric side. [135], [136] The true ring is circumferential, symmetric from all radiographic angles of view, and 3 mm or less in thickness (Fig. 58-10). Most lower esophageal mucosal rings are asymptomatic; however, they may be a cause of intermittent dysphagia and typically present in patients older than 40 years of age. [137], [138]

FIGURE 58-10. A: Barium esophagram demonstrates a lower esophageal ring at the upper end of a hiatal hernia pouch. **B:** Lower esophageal "B" or Schatzki ring. The ever-present hiatal hernia is shown below the ring. The ring margin is smooth, less than 3 mm thick, and without inflammation or evidence of fibrosis. **C:** Retroview into a small hiatal hernia, with the gastric folds effaced by inflation, reveals the appearance of a "B" or Schatzki ring with squamous mucosa over the caudal margin of the ring. The proximal, intrinsic muscular, segment of the lower esophageal sphincter is snugly closed around the endoscope (**top center**). This 12-mm

segment of squamous mucosa between the endoscope and the squamocolumnar junction that normally lies within the diaphragmatic hiatus represents the crural segment of the lower esophageal sphincter, displaced cephalad by the hiatal hernia.

The severity of symptoms depends on the diameter of the ring. Rings larger than 20 mm in diameter usually are asymptomatic, rings 13 to 20 mm in diameter cause variable degrees of dysphagia depending on type and size of bolus, and rings less than 13 mm in diameter regularly cause solid food dysphagia. [139], [140] True rings usually are not associated with inflammation, although serial esophagrams have confirmed unexplained development and progressive narrowing of true lower esophageal mucosal rings. [141], [142] and [143] A good deal of misunderstanding in the literature and in practice has resulted from confusing true rings with ringlike strictures caused by acid reflux. On esophagram, a lower esophageal mucosal ring appears as a thin (<3.0 mm) transverse circumferential ridge above the hiatus of the diaphragm. To visualize the ring, the region of the esophagogastric junction must be adequately distended (see Fig. 58-10 *A*). By definition, a hiatal hernia is always present. Mucosal or Schatzki rings are not present in patients with columnar cell–lined (Barrett) esophagus. Performing a Valsalva maneuver during the esophagram is helpful in demonstrating a ring. [144] Symptomatic mucosal rings are reproducible and do not disappear during radiographic examination. Barium esophagram with a prone full-column technique is more sensitive than double-contrast radiography or endoscopy in detecting many lower esophageal mucosal rings. [145] The use of a barium tablet or marshmallow bolus may further improve the sensitivity of the barium esophagram to correlate dysphagia with the ring. The endoscopic characteristics of lower esophageal rings and the associated findings should be understood by all endoscopists (see Fig. 58-10B,C). [146] These rings are different from ringlike strictures and are usually treated differently by dilation. Symptomatic lower esophageal rings should be treated (see Chapter 145). Most patients can be effectively treated with passage of a 16.5- to 20-mm (50F–60 F) Maloney dilator. [147] For very tight rings, some clinicians use the standard gradual peroral sequential dilation technique rather than passage of a single large-diameter bougie.

Muscular Rings The lower esophageal muscular ring, or "A" ring, occurs about 1.5 cm proximal to the squamocolumnar junction at a level that corresponds to the most cephalic segment of the lower esophageal sphincter. [14], [148] Muscular rings are seen most often in patients with esophageal motor disorders, gastroesophageal reflux, and hiatal hernia. The most common symptom associated with muscular rings is intermittent dysphagia. On barium esophagram, muscular rings are smooth, symmetric narrowings that are broader (4–5 mm) in longitudinal extent than mucosal rings. They may vary in caliber during radiographic examination and may disappear with full distention. [144] The treatment of symptomatic muscular rings by passing a 16.5- to 20-mm (50F–60F) Maloney dilator is helpful, but only for a limited time. Injecting botulinum toxin A into a hypertensive muscular ring also often provides symptomatic relief for a limited time. [149], [150] and [151]

Webs Esophageal webs are thin (1–2 mm) transverse membranes of squamous epithelium (see Color Fig. 15-7 in *Atlas of Gastroenterology,* 3rd ed.). They usually occur in the cervical esophagus, may originate from either wall, and are rarely circumferential. They may be multiple. Cine radiography performed for symptoms of dysphagia detected esophageal webs in 7% of patients in a large series. [152] Cervical esophageal webs have been reported to occur in association with heterotopic gastric mucosa, perhaps as the result of chronic injury caused by local acid production by oxyntic cells in the heterotopic gastric tissue. [130] The association of postcricoid webs with iron deficiency anemia (i.e., Plummer-Vinson or Patterson-Kelly syndrome) is rare. [152], [153], [154] and [155] However, this group of patients may be at increased risk for the development of pharyngeal and cervical esophageal carcinoma. [156], [157] Most webs are asymptomatic. Intermittent solid food dysphagia is the usual complaint in symptomatic patients. Midesophageal webs are rare and typically present with dysphagia. They may be single or multiple and are believed to be of congenital origin. [158], [159], [160], [161] and [162] They may be confused with congenital esophageal stenosis or the so-called ringed esophagus. [66], [68] If symptomatic, they are best treated with bougienage. Treatment with transendoscopic incision or surgical resection has been reported but is rarely necessary. [159], [160], [161], [162], [163] and [164]

PHARYNGOESOPHAGEAL DIVERTICULA

Pharyngoesophageal diverticulum was first described by Ludlow in 1769. [165] Zenker and Ziemssen subsequently reviewed the world literature in 1877–hence the association of Zenker's name with this condition. [166] Pharyngoesophageal or Zenker diverticula form from a protrusion of the posterior hypopharyngeal mucosa between the oblique fibers of the inferior pharyngeal constrictor and the transverse fibers of the cricopharyngeus (i.e., triangle of Killian) proximal to the esophagus. Debate has long continued regarding the mechanisms for the formation of pharyngoesophageal diverticula. [167] Evidence suggests that during swallowing, high hypopharyngeal pressures occur in some people because of poor compliance of the upper esophageal sphincter, and this may be important in the development of Zenker diverticula. [168] Associated neurological disorders, usually substantiated by cranial computed tomography or magnetic resonance imaging, have been found in up to 83% of cases. Basilar brain and brainstem lesions are most common. Peripheral neuropathies are found in some cases. [169]

Patients typically present after 50 years of age with a duration of symptoms ranging from weeks to years. Symptoms include dysphagia for both solids and liquids, regurgitation of undigested food, cough, and halitosis. If a large diverticulum is present, gurgling in the neck or a bulge in the left side of the neck can be seen during eating. Aspiration pneumonia or significant weight loss can occur. The diagnosis is obtained by barium swallow with lateral views of the pharyngoesophageal junction (Fig. 58-11). Caution must be used during the passage of nasogastric tubes or endoscopes because of the risk for inadvertent perforation of the diverticulum. Endoscopy is important for adequately evaluating the diverticulum. Passage of the endoscope over an endoscopically or fluoroscopically placed intraesophageal guidewire may be necessary in some instances to intubate the esophagus safely.

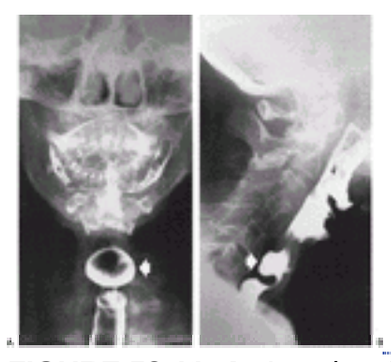

FIGURE 58-11. A: Anterior view of barium-filled Zenker diverticulum (*arrow*). The narrowed segment just distal to the diverticulum is the lumen of the cricopharyngeus. **B:** Lateral view of Zenker diverticulum (*arrow*) in the same patient. The prominent closure of the cricopharyngeus muscle (upper esophageal sphincter) is shown just distal to this diverticulum.

The usual approach to symptomatic pharyngoesophageal diverticulum is surgery. [170], [171] A dynamic videofluoroscopic examination or a manometric study of pharyngeal motility should be performed before cricopharyngeal myotomy to ensure adequate pharyngeal function. Cricopharyngeal myotomy is the primary surgical method in all cases, with either resection of the diverticulum or diverticulopexy. Diverticulectomy is preferred for diverticula larger than 3 cm, but in patients of advanced age or with significant comorbidity, diverticulopexy may be preferable therapy. Surgical morbidity and recurrence rates vary widely in reported surgical series.

Endoscopic therapy of Zenker diverticula has been tried with different techniques since 1917, but because of technical difficulties and complications, none has been considered ideal. [172] Techniques have included use of a diathermy knife, surgical stapling gun, and carbon dioxide laser diverticulotomy. [173], [174] and [175] The newest endoscopic method is performed with prior antibiotic prophylaxis, intravenous sedation, and topical anesthesia. The septum between the posterior wall of the proximal esophagus (cricopharyngeus muscle) and anterior wall of the Zenker diverticulum is incised with a needle-knife papillotome and electrical monopolar cautery. [176], [177] The patient may resume oral intake 18 hours after the procedure. In about two thirds of cases, a second procedure is needed to complete the myotomy for good dysphagia relief. Patients with chronic cough are not good candidates because they are at higher risk for cervical emphysema. Other complications, such as bleeding, stenosis, and fistula, are unusual. [176]

Postoperatively, radiographic recurrence appears to be more common than symptomatic recurrence. [177] Squamous cell carcinoma can complicate a long-standing pharyngoesophageal diverticulum. In a series of 1249 patients seen during a 53-year period, squamous cell carcinoma was found in 0.4%. [178] With barium swallow, carcinoma may appear as a persistent filling defect in an otherwise smooth-walled diverticulum. [179] Spindle cell carcinoma and benign tumors also have been reported to arise in pharyngoesophageal diverticula. [179], [180] Simple diverticulectomy has been recommended for localized tumor, with pharyngolaryngectomy reserved for more extensive disease. The 5-year survival for reported cases is 14%. [181]

ESOPHAGEAL DIVERTICULA

Midesophageal and Epiphrenic Diverticula

Diverticula also may occur in the middle or distal esophagus. It used to be thought that midesophageal diverticula developed secondary to traction from contiguous mediastinal inflammation and adenopathy (e.g., fungal, tuberculous). Evidence now indicates that these diverticula probably develop secondary to motility disorders.[182] In children, retained esophageal foreign bodies may be a cause.[183] Midesophageal diverticula can be large and wide-mouthed, small, or ultiple. The diagnosis is typically made by barium swallow (Fig. 58-12).

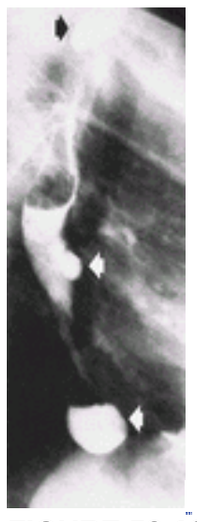

FIGURE 58-12. Barium radiograph demonstrates three diverticula: Zenker diverticulum (*upper arrow*), midesophageal diverticulum (*middle arrow*), and barium-filled epiphrenic diverticulum (*lower arrow*). This occurrence of three types of diverticula in the same patient is distinctly rare.

Distal esophageal or epiphrenic diverticula usually develop secondary to motility disorders, including achalasia. Dysphagia and chest pain are probably caused by the underlying motility disorder.[184] An epiphrenic diverticulum is seen on barium swallow as a smooth lateral pouch, usually protruding from the right wall just proximal to the lower esophageal sphincter (see Figure 58-12).

Endoscopically, a diverticulum appears as a smooth pouch off the axis of the esophageal lumen. As it enlarges, the mouth of the diverticulum may open in direct line with the axis of the proximal esophagus. If this occurs, food and fluid first enter the diverticulum and then enter the distal esophagus by spillover. Food impaction leads to enlargement of the diverticulum, regurgitation, dysphagia, and retrosternal discomfort. Dilation over a guidewire under fluoroscopy provides partial and temporary symptom relief in some patients with diverticula. Small diverticula are often asymptomatic, but transthoracic diverticulectomy may be needed for regurgitation and pulmonary sequelae. Botulinum toxin A injection of the lower esophageal sphincter has been effective in providing excellent relief of dysphagia, although of limited duration, in patients with epiphrenic diverticula associated with achalasia and other motility disorders.[150] The evolution of laparoscopic surgery now includes therapy for epiphrenic diverticula. A thorough presurgical evaluation, including barium esophagram, esophagoscopy, and esophageal manometry, should be carried out in all cases. Divertulectomy, accompanied by lower esophageal sphincter myotomy plus some degree of fundoplication, has been successfully accomplished via the laparoscopic transhiatal approach without complications in several cases.[185]

Esophageal Intramural Pseudodiverticulosis

Esophageal intramural pseudodiverticulosis is a rare condition, first described by Mendl and colleagues in 1960,[186] in which multiple small pseudodiverticula form in the wall of the esophagus by dilation of the excretory ducts of the submucosal esophageal glands.[187], [188]

Most patients present with chronic dysphagia.[186], [187] Esophageal strictures are seen in 70% to 90% of patients, and esophageal manometric abnormalities have been found in two thirds of those studied. Esophageal candidiasis has been described in up to 50% of reported cases; however, its role in the development of esophageal intramural pseudodiverticulosis is unknown.

The relation of corrosive injury of the esophagus to intramural pseudodiverticulosis has been emphasized.[189] A report of 14 cases of this entity in 59 patients with corrosive esophageal injury noted that an esophageal stricture was a constant association. No correlation was found between the length of the stricture and the number of diverticula, and the diverticula regressed in number or disappeared altogether after the stricture was dilated.

The treatment of esophageal intramural pseudodiverticulosis is directed toward underlying conditions. Dilation of strictures, antireflux therapy, and calcium channel blockers have been reported to relieve symptoms.[189], [190]

ESOPHAGEAL HIATAL HERNIAS

Hiatal hernia, one of the most common maladies of humans, is covered in detail in Chapter 60 and Chapter 71.

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CHAPTER 59

Peter J. Kahrilas and John E. Pandolfino

MOTILITY DISORDERS OF THE ESOPHAGUS

One of the many complexities of the esophagus is that it encompasses the anatomic and physiological transition between two distinct regions, the oropharynx and the gut. The oropharynx is composed of striated muscle, controlled by the cerebral cortex and medulla, and is capable of precise tactile sensation; the distal esophagus is composed of smooth muscle, controlled by the vagus nerve and enteric nervous system, and is capable only of notoriously imprecise sensation. Between these end points, a gradual transition occurs within the esophagus. It follows from this arrangement that proximal esophageal motor dysfunction closely mimics that of the pharynx, whereas distal dysfunction is more akin to that observed in the remainder of the alimentary tract. Recognizing this natural grouping, this chapter on motility disorders of the esophagus first considers oropharyngeal dysphagia, which includes most physiological aberrations of the upper esophageal sphincter (UES) and proximal esophagus, and then focuses on purely esophageal motor disorders, which include considerations mainly relevant to the distal smooth muscle esophagus.

OROPHARYNGEAL SWALLOWING DISORDERS

Oropharyngeal dysphagia is associated with high morbidity, mortality, and cost. Estimates of the prevalence of dysphagia among persons older than 50 years of age range from 16% to 22%. [1], [2] Within health care institutions, it is estimated that up to 13% of hospitalized patients [3] and 60% of nursing home residents [4] have feeding problems, most of which are attributed to oropharyngeal dysfunction, as opposed to esophageal dysfunction. The consequences of oropharyngeal dysphagia are severe: dehydration, malnutrition, aspiration, choking, pneumonia, and death. In fact, mortality of nursing residents with dysphagia and aspiration can be as high as 45% over 1 year. [5] As our population continues to age, oropharyngeal dysphagia will become an increasing problem associated with complex medical and ethical issues.

Mechanics of Swallow

Oropharyngeal swallowing begins with an oral phase that is then followed by a pharyngeal phase. The oral phase of swallowing is largely voluntary and is highly variable depending on, for example, taste, environment, hunger, and motivation. Disorders of the oral phase of swallowing occur with many conditions characterized by global neurological dysfunction such as head trauma, cerebral tumors, or chorea. Detailed discussion of these conditions can be found in texts on swallow evaluation and therapy. [6], [7] The pharyngeal phase of swallowing is the complex oropharyngeal contractile event referred to as the *swallow response.* Afferent sensory fibers capable of triggering the pharyngeal swallow travel centrally through the internal branch of the superior laryngeal nerve (from the larynx) and through the glossopharyngeal nerve (from the pharynx). [8] These sensory fibers converge before terminating in the medullary swallow center. The location and architecture of the medullary swallowing center have been extensively investigated. [9] The evolving model is summarized in Figure 59-1. [10] The clinical significance of this neuronal architecture is that swallowing is relatively resistant to disturbance by diseases affecting brain centers higher than the medulla. However, medullary motor neuron diseases such as bulbar poliomyelitis or amyotrophic lateral sclerosis can lead to severe dysfunction.

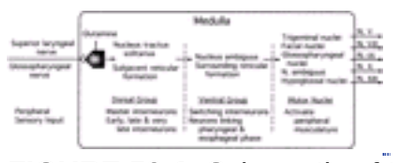

FIGURE 59-1. Schematic of the neural organization of the pharyngeal swallow response. Sensory input enters the medulla at and below the nucleus of the solitary tract, and motor output exits through five cranial nerves. The presumed function of each group of neurons is itemized beneath each anatomical grouping. The “master neurons” that establish the timing of sequential excitation of specific cranial motor nuclei are believed to be among the dorsal group, located in and around nucleus tractus solitarius. Once activated, the master neurons can establish the entire motor sequence of the swallow without further sensory input. Also among the dorsal group are interneurons activated at specific times within the swallow pattern, corresponding to the activity of specific groups of pharyngeal and esophageal muscles. Depending on the temporal relationship of neuronal activity with the onset of deglutition, these medullary neurons are classified as “early,” “late,” or “very late” neurons. The second group of interneurons located in and around the nucleus ambiguus probably function as switching neurons to relay the swallowing orders from the dorsal pattern generators to the various motor neuron pools involved in enacting the muscular response. (From Kahrilas PJ. Functional anatomy and physiology of the esophagus. In: Castell DO, ed. The esophagus, 2nd ed. Boston: Little, Brown, 1995:1.)

Although understood physiologically as the patterned activation of motor neurons and their corresponding motor units, swallowing is clinically evaluated in mechanical terms, specifically, the effect of this motor activity on the configuration of the oropharyngeal cavity. The anatomic complexity of the oropharynx is best evaluated by videofluoroscopic or cineradiographic analysis. The major events within the pharyngeal swallow are illustrated by representative x-ray images and three-dimensional reconstructions in Figure 59-2. Evident in this figure, swallowing results in the transient geometric rearrangement of pharyngeal structures from a respiratory to an alimentary pathway that is normally accomplished and reversed within 1 second. The overall swallow response can be subdivided into several closely coordinated actions:

- Nasopharyngeal closure by elevation and retraction of the soft palate
- UES opening
- Laryngeal closure
- Tongue loading (ramping)
- Tongue pulsion
- Pharyngeal clearance.

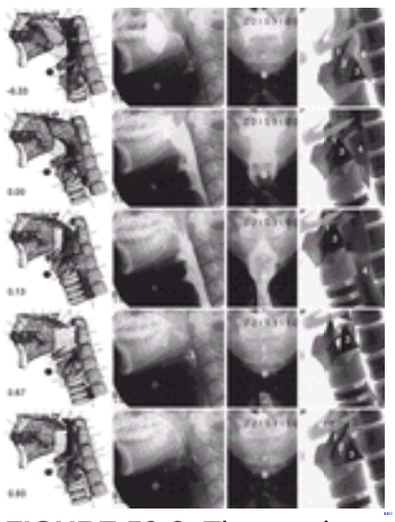

FIGURE 59-2. The oropharyngeal swallow as imaged by videofluoroscopy and is reconstructed in three dimensions with computer graphics. From **left** to **right,** each horizontally arranged group of images contains the three-dimensional reconstruction of the pharyngeal cavity and surrounding structures, the lateral radiographic appearance of the pharynx during a 10-mL barium swallow, the corresponding posteroanterior radiographic appearance, and a magnified view of the hypopharynx at the instant in time indicated at the **left.** Time 0.00 is the instant of upper esophageal sphincter (UES) opening; the entire sequence of events transpires within 1 second. The metal sphere under the chin is used to correlate among images. In the magnified hypopharyngeal reconstructions, *1* is the epiglottis, *2* is the laryngeal vestibule, *3* is the arytenoid cartilage, *4* is the esophagus, and *4'* is the pyriform sinus after closure of the UES. Note the importance of laryngeal elevation during the pharyngeal reconfiguration and synchrony of UES opening with laryngeal vestibule closure.

Precise coordination of these actions is an obvious imperative, and early attempts at defining that coordination focused on timing these individual elements of the swallow response relative to each other. However, that timing is affected both by volition and by the volume of the swallowed bolus, making it difficult to establish a universal temporal reference among elements. A detailed analysis of the coordination among swallow events concluded that there is, in fact, constant coordination at the beginning and end of the swallow but variability in how long the alimentary pharyngeal configuration persists before reverting to the respiratory configuration. [11] Thus, to construct a time line of the biomechanical events within the swallow and still preserve the constancy of the most stereotyped aspects of the swallow, onset events must be timed from the beginning and offset events from the end, as illustrated in Figure 59-3.

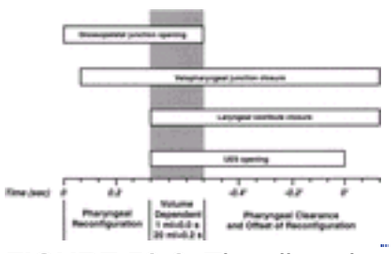

FIGURE 59-3. Time line showing the coordination and volume-induced modifications in the timing of events within the pharyngeal swallow. Each *horizontal bar* depicts the period during which one of the oropharyngeal valves is in its swallow configuration, as opposed to its configuration during respiration. Events at the onset and offset of pharyngeal reconfiguration bear a fixed time relation to each other regardless of swallow bolus volume. The stereotypy of these phases is demonstrated by referencing onset events from *time 0,* counting forward, and offset events from *time 0',* counting forward or backward, respectively. This timing scheme defines the volume-dependent, middle portion of the time line (*shaded*) that has a value of 0 for 1-mL swallows and 0.2 seconds for 20-mL swallows. Thus, the alteration in the timing of the swallow response with larger-volume swallows occurs by prolonging the persistence of pharyngeal reconfiguration without changing the synchrony of events at onset or offset. (From Kahrilas PJ, Lin S, Chen J, Logemann JA. Oropharyngeal accommodation to swallow volume. Gastroenterology 1996;111:297.)

The most fundamental anatomic reconfiguration required to transform the oropharynx from a respiratory to a swallow pathway is to open the inlet to the esophagus and to seal the inlet to the larynx. As suggested in Figure 59-2 and Figure 59-3, these events occur in close synchrony. On examining Figure 59-2, it is apparent that the larynx exhibits substantial axial mobility, and the UES is obligated to move with the larynx because the cricopharyngeus attaches to the lateral aspects of the cricoid cartilage and the lamina of the cricoid underlies the anterior wall of the sphincter. The mechanical determinants of UES opening are laryngeal elevation and anterior traction on the hyoid. [12], [13] The mechanical determinants of laryngeal vestibule closure, which is almost exactly synchronized with UES opening, are laryngeal elevation and anterior tilting of the arytenoid cartilages against the base of the epiglottis. [7] Thus, analyzing the efficacy of either of these events inevitably focuses on laryngeal elevation, which is greater and persists longer for larger volume swallows. [14] UES relaxation occurs at roughly the same degree of elevation regardless of swallow volume, and it precedes sphincter opening by about 0.1 second. What changes with swallow volume is the persistence of laryngeal elevation above this critical value, consistent with the scheme of Figure 59-3. It is important to recognize the distinction between UES relaxation and UES opening. UES relaxation results from cessation of excitatory neural input while the larynx is elevating. Once the larynx is elevated, UES opening results from traction on the anterior sphincter wall caused by contraction of the suprahyoid and infrahyoid musculature that also results in a characteristic pattern of hyoid displacement. [12], [13] Both the diameter and the duration of sphincter opening increase with increased bolus volumes.

The two main determinants of bolus transport out of the oropharynx are the actions of the tongue and of the pharyngeal constrictors. In the case of the pharyngeal constrictors, the propagated pharyngeal contraction has similar propagation and vigor regardless of bolus volume. [15] However, the propagated pharyngeal contraction is more involved with the process of clearance than of bolus propulsion; it strips the last residue from the pharyngeal walls. Tongue motion, conversely, varies substantially with bolus volume, suggesting that it has a cardinal role in determining differences in bolus propulsion among swallow volumes. [16]

UES closure coincides with the arrival of the propagated pharyngeal contraction, as evident by the fixed time relationship between these events. [13] However, the contractile activity of the sphincter has an added dimension as well, exhibiting increased electromyographic activity during laryngeal descent. [17] The magnitude of this deglutitive UES contractile activity is further augmented by either sphincteric or proximal esophageal distention. The net result is a grabbing effect in which reflexive contraction of the sphincter and laryngeal (UES) descent complement each other to clear residue from the hypopharynx. [18] This clearing function probably acts to minimize the risk of post-swallow aspiration by preventing residual material from adhering to the laryngeal inlet when respiration resumes.

Evaluation and Classification of Oropharyngeal Dysphagia

The evaluation of patients with presumed oropharyngeal dysphagia should focus on five fundamental questions:

1. Does the patient describe dysphagia, as opposed to globus sensation or xerostomia?
2. Is the dysphagia oropharyngeal or esophageal in origin?
3. Is the dysphagia secondary to a structural or functional disorder?
4. Is there an underlying related or causative disorder?
5. Should therapy be directed toward the underlying cause or the dysphagia itself?

These questions can usually be answered with a careful history and physical examination. However, further diagnostic tests may be needed to determine both the cause of oropharyngeal dysphagia and the proper treatment.

The patient history is crucial in the evaluation of oropharyngeal dysphagia. Major objectives of the history are to differentiate oropharyngeal dysphagia from esophageal dysphagia, xerostomia, or globus sensation. Whereas patients almost invariably accurately recognize the locus and consequence of oropharyngeal dysphagia, they mistakenly identify the neck as the locus of bolus hang-up with esophageal dysphagia about 30% of the time. Therefore, elicitation of symptoms such

as aspiration, coughing, nasopharyngeal regurgitation, or drooling are of great value in distinguishing oropharyngeal dysphagia from proximally referred esophageal dysphagia.

Distinguishing oropharyngeal dysphagia from globus sensation can be particularly vexing. Unlike dysphagia, which occurs only during swallowing, globus sensation is prominent between swallows. Patients relate the nearly constant sensation of having a lump in their throat or feeling a foreign object caught in their throat. In some instances, globus sensation is associated with reflux symptoms and in others with substantial anxiety; that anxiety is sometimes thought to be etiologic (globus hystericus). Unfortunately, studies have failed to define an objective anatomic or physiological cause for globus sensation, and we are left with the crucial data being in the history; globus sensation persists regardless of the act of swallowing.

Physical examination may help to identify features of the underlying systemic or metabolic disorder or to localize the neuroanatomic level and severity of a causative neurological lesion when present. The patient's general condition should be assessed to determine the degree that dysphagia is affecting fluid and nutritional status. Patients may require hydration and correction of electrolyte abnormalities before a thorough examination is undertaken. Examination of the oral cavity, head, and neck for masses, lymph nodes, goiter, and evidence of previous surgery or radiation therapy will help to define structural abnormalities associated with dysphagia. Neurological examinations may indicate cranial nerve dysfunction, neuromuscular disease, cerebellar dysfunction, or an underlying movement disorder. Of note, contrary to popular belief, the gag reflex is not predictive of pharyngeal swallowing efficiency or aspiration risk. The gag reflex is absent in 20% to 40% of physiologically normal adults. [19]

If the cause of oropharyngeal dysphagia is not readily apparent after initial evaluation, further diagnostic studies are indicated. Because the management implications are so different, *the first task in the evaluation of suspected oropharyngeal dysphagia is to distinguish between structural and functional etiologies.* Structural abnormalities that may result from trauma, surgery, tumors, caustic injury, congenital anomalies, or acquired deformities are identified by endoscopic or radiographic examination. Functional abnormalities can be attributable to dysfunction of intrinsic musculature, peripheral nerves, or central nervous system control mechanisms. Endoscopy may be performed either transorally or transanally to identify tumors, webs, or hypopharyngeal diverticula. Barium studies may define areas of obstruction and hypopharyngeal diverticula; however, they add little structural information to endoscopic studies.

After structural defects have been excluded, videofluoroscopic evaluation of swallowing is used for a functional evaluation. This approach is frequently referred to as a modified barium swallow, which Logemann [7] described as a protocol composed of a series of swallow tasks. Images are obtained in a lateral projection, framed to include the oropharynx, palate, proximal esophagus, and proximal airway. These images are then evaluated with respect to four major categories of oropharyngeal dysfunction:

1. Inability or excessive delay in initiation of pharyngeal swallowing
2. Aspiration
3. Nasopharyngeal regurgitation
4. Residue of the ingestate within the pharyngeal cavity after swallowing.

Furthermore, the procedure allows for evaluation of the efficacy of various compensatory dietary modifications, postures, and swallowing maneuvers in compensating for observed swallowing dysfunction.

Intralumenal manometry can quantify the strength of pharyngeal contraction, the completeness of UES relaxation, and the relative timing of these events. When coupled with concurrent videofluoroscopy, it may provide useful complementary information regarding UES dysfunction. High intrabolus pressures may distinguish impaired UES opening from impaired UES relaxation and weak pharyngeal contractions as a cause of oropharyngeal dysphagia.

Structural Causes of Oropharyngeal Dysphagia

Implicit in the mechanical description of swallowing summarized earlier is that normal swallowing is associated with minimal outflow resistance from the oropharynx. Identification of obstructing lesions that cause dysphagia will lead to specific management in most cases. For example, cervical webs and pharyngeal or cricopharyngeal strictures or tumors are indications for surgery, dilation, antineoplastic therapy, or some combination of these treatments. In the setting of benign lesions such as strictures or postcricoid webs, simple dilation is safe and effective. Cervical osteophytes, if prominent, can also cause obstructive dysphagia. Because the posterior wall of the pharynx is so closely opposed to the anterior aspect of the cervical vertebrae, cervical osteophytes result in an anterior bulging into the hypopharynx that can make passage of a normal-sized bolus difficult. [20] The most common structural abnormalities of the hypopharynx associated with dysphagia are hypopharyngeal diverticula and cricopharyngeal bars. Given the interplay between both structural and functional defects involved in these two disorders, further consideration is merited.

Hypopharyngeal Diverticula and Cricopharyngeal Bars Acquired hypopharyngeal diverticula are most common in men after the age of 60 years. The most frequent site of herniation is Killian's dehiscence between the oblique fibers of the inferior pharyngeal constrictor and the cricopharyngeus muscle in the midline posteriorly, this being the location of a Zenker diverticulum (Fig. 59-4). [21] Other locations of acquired pharyngeal diverticula include the lateral slit separating the cricopharyngeus muscle from the fibers of the proximal end of the esophagus through which the recurrent laryngeal nerve and its accompanying vessels run to supply the larynx, the location of penetration of the inferior thyroid artery into the hypopharynx, and the junction of the middle and inferior constrictor muscles. The unifying theme of these locations is that they are sites of potential weakness of the muscular lining of the hypopharynx. Hypopharyngeal diverticula are often asymptomatic until they enlarge sufficiently to store a significant amount of food or liquid. In most instances, symptoms are dysphagia, halitosis, post-swallow regurgitation, or even aspiration of material from the pharyngeal pouch.

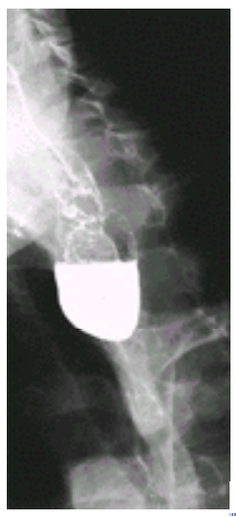

FIGURE 59-4. Radiograph of a large Zenker diverticulum partially filled with barium. Although the point of herniation is midline posterior, the diverticulum necessarily migrates laterally in the neck because there is no potential space between the posterior pharyngeal wall and the vertebral column.

Hypopharyngeal diverticula have been hypothesized to result from delayed UES relaxation, failure of relaxation, or premature contraction. [22] However, few data and considerable contradiction accompany each of these hypotheses. A more plausible explanation for the development of diverticula is that they form as a result of a restrictive myopathy associated with diminished compliance of the cricopharyngeus muscle. Surgical specimens of cricopharyngeus muscle strips from 14 patients with hypopharyngeal diverticula demonstrated structural changes that would decrease UES compliance and opening. [23] The cricopharyngeus samples from these patients had "fibro-adipose tissue replacement and (muscle) fiber degeneration." Thus, although the muscle relaxes normally during a swallow, it cannot distend normally, resulting in the appearance of a cricopharyngeal indentation, or bar, during a barium swallow (Fig. 59-5). Diminished sphincter compliance necessitates increased hypopharyngeal intrabolus pressure to maintain transsphincteric flow through the smaller UES opening. The increased stress on the hypopharynx from the increased intrabolus pressure may lead to the formation of hypopharyngeal diverticula.

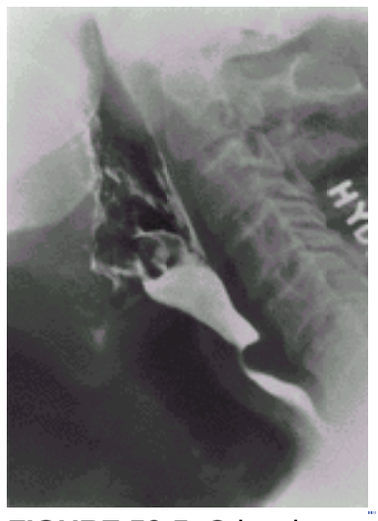

FIGURE 59-5. Cricopharyngeal bar in a patient with oropharyngeal dysphagia. The posterior indentation of the barium column is caused by a noncompliant cricopharyngeus muscle. (Courtesy of Dr. Richard Gore.)

Management of Structural Oropharyngeal Dysphagia The treatment of hypopharyngeal diverticula is cricopharyngeal myotomy with or without diverticulectomy. Cricopharyngeal myotomy reduces both the resting sphincter tone and resistance to flow across the UES. Resting UES tone is reduced to approximately 50% of baseline, suggesting that the derived benefit is from increased compliance at the UES. One study found that the compliance of the sphincter after diverticulectomy with myotomy was restored to normal in five patients after surgery, as indicated by normal hypopharyngeal intrabolus pressure during swallowing. [24] Good or excellent results can be expected in 80% to 100% of patients with Zenker diverticula treated by transcervical myotomy combined with diverticulectomy or diverticulopexy. [25] There are instances in which a limited procedure would be adequate, but a definitive approach to the problem of pulsion diverticula should involve both diverticulectomy and myotomy. Diverticulectomy alone risks recurrence because the underlying stenosis at the level of the cricopharyngeus is not remedied. Similarly, myotomy alone risks not solving the problem of food accumulation within the diverticula, with attendant regurgitation and aspiration. Small diverticula may, however, disappear spontaneously after myotomy. Whether a cricopharyngeal bar in the absence of a diverticula requires treatment is less clear. Certainly, if dysphagia exists and combined fluoroscopic and manometric analysis demonstrates reduced sphincter opening in conjunction with elevated upstream intrabolus pressure, there is good rationale for treatment. Anecdotal evidence suggests that, in this situation, simple dilation with a large-caliber bougie may be efficacious, and this is certainly a reasonable treatment option before myotomy.

Functional Causes of Oropharyngeal Dysphagia

Primary neurological or muscular diseases involving the oropharynx can be associated with dysphagia. Thus, whereas esophageal dysphagia usually results from esophageal diseases, oropharyngeal dysphagia is frequently the result of neurological or muscular diseases, with oropharyngeal dysfunction being just one pathological manifestation. Although the specifics of the diseases vary, the net effect on swallowing can be analyzed according to the mechanical description of the swallow outlined earlier. Table 59-1 summarizes the mechanical elements of the swallow along with the manifestation and consequence of dysfunction and representative pathological conditions in which they are likely encountered. Some of the more distinct pathological entities are discussed in the following sections.

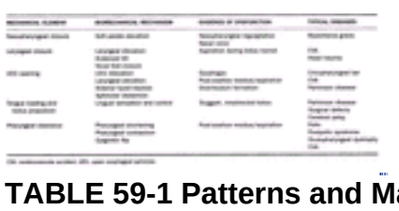

TABLE 59-1 Patterns and Manifestations of Oropharyngeal Dysphagia

Neurological Causes Neurological diseases can damage the neural structures requisite for either the afferent or efferent limbs of the oropharyngeal swallow. Because there is nothing unique to neurons that control swallowing, their involvement in disease processes is usually random. Furthermore, in most instances, functions mediated by adjacent neuronal structures are concurrently involved. Virtually any disease of the central nervous system can potentially cause dysphagia. The following discussion focuses on neuropathological processes with the best-characterized patterns of dysphagia.

Cerebrovascular accidents. Aspiration pneumonia has been estimated to have a 20% death rate in the first year after a *stroke* and 10% to 15% each year thereafter. [26] It is usually not the first episode of aspiration pneumonia but the subsequent recurrences over several years that will eventually cause death. [27] The ultimate cause of aspiration pneumonia is dysphagia leading to aspiration, which can occur by numerous mechanisms: absence or severe delay in triggering the swallowing reflex (swallow apraxia), reduced lingual control, and unilateral or bilateral weakened laryngopharyngeal musculature. [7] Conceptually, these causes can be divided into motor or sensory impairments. Although sensory and motor impairment frequently coexist, each is addressed separately. Consistent with the neuronal architecture outlined in Figure 59-1, cortical strokes are less likely to result in dysphagia than are brainstem strokes. [28] In a report of 100 consecutive patients, 37 of 86 (43%) who sustained an acute cerebral infarct experienced dysphagia when they were evaluated within 4 days of the event. [28] However, 86% of these patients were able to swallow normally 2 weeks later. Two-dimensional scalp topographic maps of the pharyngeal muscles obtained by transcranial magnetic stimulation suggests that recovery occurs as a result of contralateral areas' taking over the lost function. [29] Thus, dysphagia is more prevalent among patients incurring larger strokes or in patients who have had prior infarcts. The sensory cues required for eliciting the pharyngeal swallow are unclear. In pathological circumstances in which one or the other neural substrate of the afferent signal has been damaged, patients may experience a relative inability to initiate a swallow. A study of sensory acuity in the supraglottic and pharyngeal regions in patients who had suffered strokes revealed either unilateral ($n = 9$) or bilateral ($n = 6$) sensory impairment of moderate to severe degree in all 15 patients studied. [30] Impaired sensation in these critical areas likely results in swallow apraxia. An interesting compensatory strategy for this sensory defect is to accentuate oropharyngeal stimulation during eating to facilitate achieving the threshold for triggering the medullary swallowing center. [7] Preliminary data suggest that the sour bolus is particularly effective in some patients with swallow apraxia, and this provides some hope for the treatment of this disabling condition. [7]

Poliomyelitis. Most cases of *poliomyelitis* involve only the spinal cord; however, the fatality rate from bulbar disease far exceeds that of spinal disease, with the most common fatal complication being respiratory depression. In addition to its dreaded effect of respiratory depression, bulbar poliomyelitis is also associated with dysphagia. In one analysis of the persistent sequelae of bulbar poliomyelitis, 28 of 47 patients (60%) had recurrent or continued involvement of the pharynx 17 or more months after their acute illness. [31] Speech and swallowing dysfunctions are the result of weakness of the levator veli palatini muscle, pharyngeal constrictors, and hyolaryngeal elevators. [32] Neurologists have observed an increasing number of patients with new paretic symptoms traceable to their remote poliomyelitis infection 30 to 40 years earlier. The slowly progressive postpolio muscular atrophy may occur in muscles that were clinically unaffected by the acute illness. [33] One investigation studied 13 patients with postpolio dysphagia and demonstrated palatal, pharyngeal, and laryngeal weakness. [34] Over half of the patients evaluated demonstrated silent aspiration, suggesting that the clinician should maintain a low threshold for evaluating such patients with videofluoroscopy. Accumulating evidence suggests that at least some of these patients with postpolio syndrome respond to pyridostigmine (Mestinon), a cholinomimetic agent.

Amyotrophic lateral sclerosis. *Amyotrophic lateral sclerosis* is a progressive neurological disease characterized by degeneration of motor neurons in the brain, brainstem, and spinal cord. Specific symptoms are dependent on the locations of affected motor neurons and the relative severity of involvement. When the degenerative process involves the cranial nerve nuclei, swallowing difficulties ensue. Oropharyngeal dysfunction characteristically begins with the tongue and progresses to involve the pharyngeal and laryngeal musculature. Patients experience choking attacks, become dehydrated or malnourished, and incur aspiration pneumonia. The decline in swallowing function is progressive and predictable, invariably leading to gastrostomy feeding. Some patients die as a consequence of their swallowing dysfunction in conjunction with respiratory depression. [35]

Parkinson disease. Although only 15% to 20% of patients with *Parkinson disease* complain of swallowing problems, more than 95% have demonstrable defects videofluoroscopically. [7], [36] The disparity between these numbers suggests that patients compensate effectively during the early stages of the disease and complain of dysphagia only when it becomes severe. Abnormalities of the oral phase of swallowing include repetitive lingual pumping before initiation of a pharyngeal swallow, piecemeal swallowing, and oral residue after the swallow. [7], [36] Patients may also exhibit a delayed swallow response and a weak pharyngeal contraction, resulting in vallecular and pyriform sinus residue after each swallow. [36] Combined manometric and fluoroscopic data suggest this to be related to the combination of incomplete UES relaxation and a weakened pharyngeal contraction. [36] Although no controlled trials are available, dysphagic symptoms have improved after therapy with levodopa

and carbidopa. [37], [38]

Tumors. Medullary or vagal *tumors* are potentially debilitating with respect to swallowing. Astrocytomas are the most common histological subtype affecting adults, whereas medulloblastomas are the most common type encountered in children. [39] The relative inaccessibility of the medulla to surgery usually means that substantial morbidity is incurred during attempted resection or palliation. Unilateral lesions of the vagus nerve can result in hemiparesis of the soft palate and pharyngeal constrictors, as well as of the laryngeal musculature. Surgical manipulation of this region can even result in complete loss of the pharyngeal swallow response. [40] The recurrent laryngeal nerves can be injured as a result of thyroid surgery, poliomyelitis, aortic aneurysms, pneumonectomy, primary malignant tumors of the mediastinum, or metastatic lesions to the mediastinum. Owing to its more extensive loop into the chest, the left recurrent laryngeal erve is more vulnerable to involvement with mediastinal node malignancy. Unilateral recurrent laryngeal nerve injury results in unilateral adductor paralysis of the vocal cords. This defect can result in aspiration during swallowing because of impaired laryngeal closure. Although some reports state the contrary, [41] it is probably rare to have any primary pharyngeal dysfunction associated with recurrent laryngeal nerve injury.

Muscular Diseases Primary muscular diseases involving the oropharynx are associated with dysphagia reflective of the pattern of involvement. Nasal voice and nasopharyngeal regurgitation indicate either weakness or paresis of the soft palate elevators, and tongue weakness can cause poor control of the bolus within the mouth. Post-swallow residue in the valleculae or hypopharynx reflects an ineffective, presumably weakened pharyngeal contraction. Aspiration suggests either weakened laryngeal elevators or post-swallow residue that is then aspirated after the swallow sequence is completed. As with neurological disorders, virtually any disorder affecting skeletal muscle can result in dysphagia. The following discussion focuses on a few of the better characterized entities.

Oculopharyngeal dystrophy. *Oculopharyngeal muscular dystrophy* is a syndrome characterized by progressive dysphagia and palpebral ptosis first described by Taylor in 1915. [42] He noted that all of his afflicted patients reaching age 50 years died of starvation resulting from pharyngeal paralysis. [42] The disease is now known to be a form of muscular dystrophy inherited as an autosomal dominant disease with occurrences clustered in families of French-Canadian descent. Genetic studies of an afflicted family indicate linkage to chromosome 14, perhaps involving the region coding for cardiac a or ß myosin heavy chains. [43] Oculopharyngeal dystrophy affects the striated pharyngeal muscles and the levator palpebrae. Other forms of muscular dystrophy occasionally affect the pharyngeal constrictors but rarely, if ever, is this a dominant manifestation. The first symptom of oculopharyngeal dystrophy is usually ptosis, which develops slowly and eventually dominates the patient's appearance. Dysphagia may begin before the ptosis but is more often manifest simultaneously with it or a few years later. The dominant functional abnormalities are weak or absent pharyngeal contraction with hypopharyngeal stasis. [44] Dysphagia progresses slowly but may ultimately lead to starvation, aspiration pneumonia, or asphyxia.

Myotonic dystrophy. *Myotonic dystrophy* is a rare disorder characterized by prolonged contraction and difficulty in relaxation of involved skeletal musculature. Dysphagia is a common complaint among affected patients. Early investigations suggested that the dysphagia was the result of myotonia of the cricopharyngeus with resultant overflow into the airway during attempted swallowing, [45] but subsequent manometric and radiographic investigations have not confirmed this. [46] The experience in more recent investigations was that even though only half of the patients studied complained of dysphagia, motor abnormalities could be demonstrated in every patient studied. The pattern of abnormality was of a weakened pharyngeal contraction, absent peristalsis in the striated muscle esophagus, and diminished or absent peristalsis in the smooth muscle portion of the esophagus. No study has demonstrated myotonia in any part of the esophagus. Thus, the consequences of the disease are identical to other forms of muscular dystrophy; poor pharyngeal clearance with risk of aspiration during the swallow if there is concurrent weakness of the laryngeal elevators or after the swallow, when the substantial pharyngeal residue may fall into the reopened airway.

Myasthenia gravis. *Myasthenia gravis* is a progressive autoimmune disease characterized by high circulating levels of acetylcholine receptor antibody and destruction of acetylcholine receptors at neuromuscular junctions. Musculature controlled by the cranial nerves is almost always involved, particularly the ocular muscles. Dysphagia is prominent in more than one third of patients with myasthenia gravis, and, in unusual instances, it can be the initial manifestation of the disease. [7] In mild cases, dysphagia may not be evident until after 15 to 20 minutes of eating. Classically, manometric studies reveal a progressive deterioration in the amplitude of pharyngeal contractions with repeated swallows. Peristaltic amplitude recovers with rest or after the administration of 10 mg edrophonium chloride. [47] In more advanced cases, the dysphagia can be profound and associated with nasopharyngeal regurgitation and nasality of the voice, even to the extent of being confused with bulbar amyotrophic lateral sclerosis or brainstem stroke. [48]

Management of Functional Oropharyngeal Dysphagia Management of functional causes of oropharyngeal dysphagia begins with definition of the aberrant physiology along the lines summarized in Table 59-1. This is most easily accomplished with a videofluoroscopic swallowing study. After definition of the patient's swallowing dysfunction, four specific issues pertaining to management of oropharyngeal dysphagia can be addressed:

1. Identification of an underlying systemic disease
2. Characterization of a disorder amenable to surgery or dilation
3. Identification of a specific pattern of dysphagia amenable to swallowing therapy
4. Assessment of aspiration risk.

Identifying underlying disease. A potential outcome of the swallowing evaluation is the identification of an underlying neuromuscular, neoplastic, or metabolic disorder that will dictate specific management. For example, dysphagia can be the presenting symptom in patients with myopathy, myasthenia, thyrotoxicosis, motor neuron disease, or Parkinson disease. In each instance, identification of the underlying disease will result in specific treatment. Whether identification and treatment of the underlying disorder improve swallowing function depends on both the natural history of the disease and the existence of effective treatment.

Disorders amenable to surgery. In marked contrast to the high efficacy observed with structural cricopharyngeal disorders, the efficacy of myotomy in neurogenic dysphagia is variable. Furthermore, most series evaluating the efficacy of myotomy in neurogenic dysphagia are uncontrolled, without specific outcome measures. Thus, although an overall favorable response rate in excess of 60% is reported, there are currently no validated criteria for preoperative selection. Theoretically, the functional limitation in these circumstances is of pharyngeal propulsion, and the potential benefit of myotomy is less obvious. [49]

Specific patterns of dysphagia amenable to swallowing therapy. After characterization of a patient's swallow dysfunction, the radiographic study should proceed to test selected compensatory or therapeutic treatment strategies. Compensatory treatments include postural changes, modifying food delivery or consistency, or the use of prosthetics. For instance, head turning can eliminate aspiration or pharyngeal residue by favoring more functional structures in patients with hemiparesis. [7] Similarly, diet modifications can reduce the "difficulty" of the swallow. [7] Therapeutic strategies are designed to alter the physiology of the swallow, usually by improving the range of motion of oral or pharyngeal structures using voluntary control of oropharyngeal movement during swallow. Depending on the severity of the impairment, level of motivation, and global neurological intactness, defective elements of the swallow can be selectively rehabilitated. For a detailed description of the techniques and limitations of swallow therapy, the reader is referred to treatises on the topic. [7], [25]

Detection of severe aspiration. Oropharyngeal dysphagia associated with aspiration is responsible for an estimated 40,000 deaths a year from aspiration pneumonia. [30] Videofluoroscopy is believed to be the most sensitive test for detecting aspiration that is not evident by bedside evaluation in 42% to 60% of patients. However, despite the logical association between deglutitive aspiration and the subsequent development of pneumonia, this sequence is not inevitable. In fact, available data suggest that radiographic aspiration has a positive predictive value of only 19% to 68% and a negative predictive value of 55% to 97% for pneumonia. [25] Nonetheless, the balance of evidence suggests that detection of aspiration is a predictor of pneumonia risk, and its detection dictates that compensatory swallowing strategies, nonoral feeding, or corrective surgery be instituted. Whether nonoral feeding eliminates the risk of aspiration is controversial. A provocative finding by Croghan and associates [5] was that, in 22 patients with radiographic aspiration, pneumonia and death were more frequent among patients who received feeding tubes. This finding suggests that aspiration of oral secretions may be important in determining pneumonia risk and has led some clinicians to consider procedures such as tracheostomy to protect the airway.

Xerostomia Although not an esophageal motility disorder, *xerostomia* (the subjective feeling of oral dryness) is covered briefly, given its association with dysphagia. Xerostomia is usually associated with reduced salivary secretion. The normal, unstimulated salivary flow rate should be 0.3 to 0.5 mL/min, whereas flow rates of less than 0.1 mL/min are consistent with xerostomia. The most common causes of xerostomia are drugs, autoimmune diseases including Sjögren syndrome and rheumatoid arthritis, and radiation therapy. Symptoms associated with xerostomia are as follows: pain, burning, and soreness of the oral mucosa, especially the tongue; difficulty in mastication, swallowing, and speech; impairment of taste; painful oral ulcers; difficulty in wearing dentures; increased dental caries; and increased frequency or volume of fluid intake. [50] The dysphagia associated with xerostomia is primarily attributable to the absence of lubricating properties of saliva. Therapy ranges from attempts to increase secretion with sugarless gum, sour lozenges, or pharmacological agents such as pilocarpine and cholinergic agents. Patients without stimulated salivary response can try commercially available saliva substitutes. [51]

ESOPHAGEAL MOTILITY DISORDERS

Neuromuscular function of the esophagus (motility) has the consistent objective of emptying the esophagus despite intrusions from above or below: primary esophageal peristalsis empties swallowed material from the esophagus; secondary peristalsis eliminates air or fluid refluxed from the stomach; the UES contracts during inspiration to exclude inspired air from the digestive tract; and elements of the gastroesophageal junction contract during transient increases of intra-abdominal pressure, preventing gastroesophageal reflux. A basic characteristic of *esophageal motility disorders,* conversely, is failure in preserving esophageal emptiness. Retained material within the esophagus, or the excessive entry of material into the esophagus, is abnormal. Such dysfunction can be categorized as disorders of peristalsis or of sphincter competence. The main dysfunction of sphincter competence occurs with gastroesophageal reflux disease (GERD; see Chapter 60). This discussion focuses on less common causes of sphincter dysfunction and on disorders of peristalsis. However, before considering these functional aberrations,

consider our present understanding of the mechanics of esophageal peristalsis.

Mechanics of Peristalsis

Esophageal Body Both the inner circular and outer longitudinal muscle layers of the proximal esophagus are composed of striated muscle and are controlled by somatic motor fibers from lower motor neurons in the nucleus retrofacialis and the nucleus ambiguus. Axons of these lower motor neurons course through the vagus nerve through the recurrent laryngeal nerve. The thoracic esophagus is mostly composed of smooth muscle receiving innervation from preganglionic neurons in the dorsal motor nucleus of the vagus. Vagal fibers synapse on myenteric plexus neurons situated between the circular and longitudinal muscle layers, rather than directly at neuromuscular junctions. Although the relationship between morphology and function of the neurons remains undetermined, vagal stimulation can either excite or inhibit esophageal musculature, depending on the intensity of the stimulus used. [52] Excitatory neurons mediate contraction of the circular and longitudinal muscle through cholinergic receptors. Inhibitory neurons affect predominantly the circular smooth muscle through nitric oxide and exert a progressively prolonged inhibition at more distal esophageal loci. Esophageal peristalsis commences as the pharyngeal contraction traverses the UES and progresses along the esophagus at 2 to 4 cm/s. Whereas primary peristalsis is that initiated by a swallow, secondary peristalsis can be elicited in response to esophageal distention at any level with air, fluid, or a balloon. [53] Primary peristalsis is dependent on activation of the swallowing center and vagal pathways. Peristalsis in the striated muscle esophagus is a function of sequential firing of efferent vagal neurons, and, thus, in this segment both primary peristalsis and secondary peristalsis are blocked by bilateral cervical vagotomy. Although primary peristalsis in the smooth muscle segment of the esophagus is also blocked by bilateral cervical vagotomy, secondary peristalsis is preserved and can be initiated by esophageal distention or vagal nerve stimulation. The progression of primary peristalsis in the smooth muscle segment is governed by the gradient of excitatory and inhibitory influence. Mechanical inhibition or latency results from membrane hyperpolarization through nitric oxide release, [54] whereas cholinergic nerves cause depolarization and decreased latency. Cholinergic influence is most prominent proximally and progressively decreases distally, whereas the inhibitory influence increases distally. [55] The mechanical correlate of peristalsis is of a stripping wave that milks the esophagus clean from its proximal to distal end (Fig. 59-6). The velocity of movement of the stripping wave corresponds closely with that of the manometrically recorded contraction such that the point of the inverted “V” seen fluoroscopically at each esophageal locus occurs with the upstroke of the pressure wave. [56] Detailed analyses of the vigor and propagation of esophageal peristalsis have concluded that progression through the tubular esophagus is not seamless. Rather, there is a distinct transition zone between the striated and smooth muscle esophagus characterized by low peristaltic amplitude, slight delay in progression, and an increased likelihood of failed transmission. [57] This transition zone becomes quite evident when peristaltic amplitude and progression are plotted topographically. The topographic analysis reveals not only the proximal transition zone, but also a segmental characteristic of peristaltic progression through the smooth muscle esophagus with two distinct contractile segments followed by the lower esophageal sphincter (LES), which contracts with vigor and persistence quite dissimilar to the adjacent smooth muscle esophagus. [58]

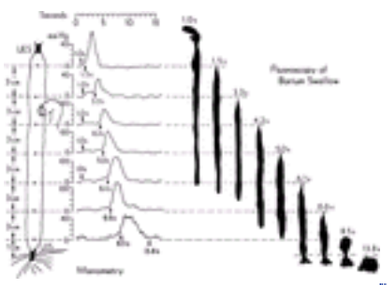

FIGURE 59-6. The relationship between manometric and fluoroscopic recordings of esophageal peristalsis during concurrent manometric and video recording of a 5-mL liquid barium swallow. The tracings from the video images of the fluoroscopic sequence on the **right** show the distribution of the barium column at the times indicated above the individual tracings and by *arrows* on the manometric record. In this example, a single peristaltic sequence completely cleared the barium bolus from the esophagus. Pharyngeal injection of barium into the esophagus occurs at the 1.0-second mark. The entry of barium causes distention and a slightly increased intralumenal pressure, indicated by the *downward pointing arrows* marked *1.0 s*. Shortly thereafter, esophageal peristalsis is initiated. During esophageal peristalsis, lumenal closure and hence the tail of the barium bolus passed each recording site concurrent with the onset of the manometric pressure wave. Hence, at 1.5 seconds, the peristaltic contraction had just reached the proximal recording site, and barium had been stripped from the esophagus proximal to that point. Similarly, at 4.2 seconds, the peristaltic contraction was beginning at the third recording site, and, correspondingly, the tail of the barium bolus was located at the third recording site. Finally, after completion of the peristaltic contraction (time 13.8 seconds), all the barium has been cleared into the stomach. (From ref. [56] .)

The longitudinal muscle of the esophagus also contracts during peristalsis, with the net effect of transiently shortening the structure by 2 to 2.5 cm. [59] Similar to the pattern of circular muscle contraction, longitudinal muscle contraction is propagated distally as an active segment at a rate of 2 to 4 cm/s. [60] The segment of contracting longitudinal muscle precedes but overlaps with the contracting segment of circular muscle. Thus, within a given esophageal segment, the contractions of the longitudinal and circular muscle are slightly out of phase with each other. Another crucial feature of the peristaltic mechanism is deglutitive inhibition. A second swallow, initiated while an earlier peristaltic contraction is still progressing in the striated muscle esophagus, causes rapid and complete inhibition of the contraction induced by the first swallow. [61] Deglutitive inhibition is secondary to hyperpolarization of the circular smooth muscle and is mediated by noncholinergic neurons in the myenteric plexus. An experimental model illustrating deglutitive inhibition in the tubular esophagus by the creation of an artificial high-pressure zone was described by Sifrim and colleagues [62] (Fig. 59-7). The artificial high-pressure zone is created by distending the esophageal lumen with a balloon and recording intralumenal pressure between the balloon and the esophageal wall. Once the high-pressure zone is established in the normally flaccid tubular esophagus, deglutitive inhibition can be demonstrated throughout the length of the esophagus, commencing concurrently with the pharyngeal swallow. [62]

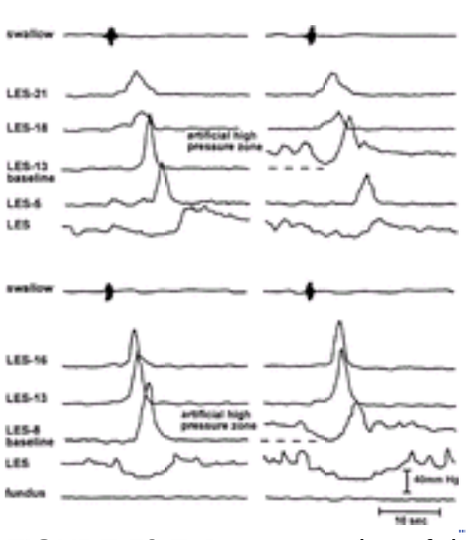

FIGURE 59-7. Demonstration of deglutitive inhibition in the tubular esophagus by the creation of an artificial high-pressure zone. The **top tracing** is a swallow marker, and the position from which each of the other tracings was obtained is referenced to the lower esophageal sphincter (LES). The artificial high-pressure zone was created 13 cm above the LES in the **upper panel** and 8 cm above the LES in the **lower panel** by inflating a balloon within the esophagus and interposing the manometric sensor between the wall of the esophagus and the balloon. The **tracings on the left** are before balloon inflation, whereas the **tracings on the right** are with the balloon inflated and the high-pressure zone developed. The contraction within the artificial high-pressure zone is inhibited concurrently with the pharyngeal swallow, much the same as with the LES illustrated in Figure 59-8. (From ref. [62] .)

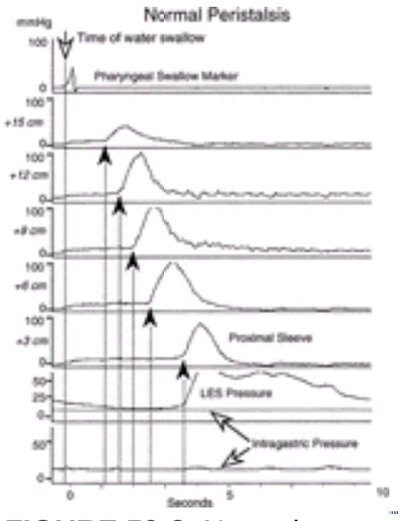

FIGURE 59-8. Normal manometric recording of an esophageal peristaltic contraction. The timing of the swallow is indicated by the first *vertical dotted line.* The *italicized numbers* to the **left** of the pressure calibrations indicate the position of each recording site relative to the midpoint of the sleeve sensor that is centered within the lower esophageal sphincter (LES). The onset of the peristaltic contraction at each esophageal recording site is indicated by a *solid arrowhead* and a *vertical dotted line*. Propagation of the peristaltic contraction is judged by the timing of the onset of the pressure complex at each recording site because this is the event indicative of lumenal closure. The propagation velocity of the peristaltic contraction is not uniform along the length of the esophagus, varying from 3 to 6 cm/s. LES pressure is recorded by a sleeve transducer that eliminates artifactual relaxation caused by esophageal shortening during swallow. LES relaxation is measured relative to intragastric pressure, indicated on the LES tracing by a *horizontal dotted line*. LES relaxation commences at the time of the pharyngeal swallow.

Lower Esophageal Sphincter Physiologically, the LES is a 2- to 4-cm-long segment of tonically contracted smooth muscle. Manometric studies reveal both axial and radial asymmetry. [63] Endoscopic ultrasound suggests that axial asymmetry may be explained by variable muscle thickness, [64] whereas radial asymmetry is likely the result of the surrounding diaphragmatic crus. [63] The LES is innervated by vagal and sympathetic efferents, which synapse on the myenteric plexus neurons. Basal LES pressure results from myogenic tone, and modification of that tone occurs through excitatory and inhibitory neurohormonal influences. The tonic behavior of the LES has been attributed to a depolarized state with the presence of continuous spike-burst activity. [65], [66] Relaxation of the LES occurs with swallowing, esophageal distention related to refluxed material or incomplete emptying, and transient LES relaxation (tLESR). Swallow-induced LESR occurs less than 2 seconds after initiation of swallowing and concurrently with deglutitive inhibition that traverses the smooth muscle esophagus. As evidenced by the manometric recording of normal peristalsis illustrated in Figure 59-8, LESR commences before the onset of the peristaltic contraction in the proximal esophagus. The LESR induced by primary peristalsis and pharyngeal stimulation is mediated by vagal efferents and is abolished by bilateral cervical vagotomy. [67] Esophageal distention of either the striated or smooth muscle esophagus induces LESR along with secondary peristalsis. It is presumed that distention-induced LESR in the striated muscle portion is centrally mediated, whereas that in the smooth muscle is mediated by the myenteric plexus. Relaxation may occur transiently without swallowing or esophageal contraction. These tLESRs are thought to be an important mechanism in the pathogenesis of GERD. Proximal gastric distention is the major stimulus for generating tLESRs. Distention stimulates gastric mechanoreceptors in the proximal stomach activating afferent fibers projecting to the nucleus of the solitary tract. Interactions with as yet undefined interneurons then result in activating vagal efferents in the dorsal motor nucleus of the vagus that stimulate prolonged LES and crural diaphragm inhibition. This reflex is also vagally mediated and can be abolished by vagotomy. [68]

Evaluation and Classification of Esophageal Dysphagia

Dysphagia is a fundamental symptom of esophageal disorders. Esophageal as opposed to oropharyngeal dysphagia is suggested by the absence of associated aspiration, cough, nasopharyngeal regurgitation, dry mouth, pharyngeal residue after swallow, or concurrent neuromuscular dysfunction. Conversely, the associated conditions of heartburn, esophagopharyngeal regurgitation, chest pain, odynophagia, or intermittent esophageal obstruction suggest esophageal dysphagia. However, an important limitation of the patient history with esophageal dysphagia is that patient identification of the location of obstruction is of limited accuracy. Specifically, a distal esophageal obstruction caused by an esophageal ring or achalasia will often be sensed as cervical dysphagia such that patients correctly locate distal dysfunction only 60% of the time. [69] Because of this subjective difficulty in distinguishing proximal from distal lesions within the esophagus, an evaluation for cervical dysphagia should encompass the entire esophagus.

Another important consideration in patient management is that esophageal motility disorders are much less common than mechanical or inflammatory causes of dysphagia: tumors, strictures, rings, and peptic, pill, or infectious esophagitis. Historical points suggestive of a motor disorder are difficulty with both solids and liquids as opposed to only solids, which is more suggestive of mechanical obstruction. However, as will become evident in the ensuing discussion, the functional consequences of mechanical or inflammatory disorders can exactly mimic those of primary motility disorders. Thus, as with oropharyngeal dysphagia, an esophageal motility disorder should be considered *only after exclusion of these more common diagnoses* by endoscopic or radiographic examination.

As outlined earlier, the physiological elements of esophageal peristalsis are the coordinated inhibition and excitation of the circular and longitudinal muscle layers of the esophagus. It follows that pathophysiological processes can involve dysfunction of deglutitive inhibition (including sphincter relaxation) or of the propagated contraction. Furthermore, because esophageal motor disorders are not usually diagnosed on histopathological grounds, their diagnosis depends on defining these functional aberrations. Table 59-2 illustrates an attempt at categorizing esophageal motility disorders on the basis of regional function of deglutitive inhibition and sequenced excitation. On examining Table 59-2, there are few well-defined esophageal motility disorders, and, as one could predict, in some circumstances it can be very difficult to choose definitively among the diagnostic possibilities. However, such is the state of the art. Furthermore, in the instance of spastic disorders, there is considerable debate over the clinical significance of some manometric findings. [70] With these considerations in mind, the following discussion emphasizes disease entities with clear diagnostic or therapeutic implications, rather than manometric phenomena of unproved significance. The exception is GERD which, although highly significant, is discussed in Chapter 60.

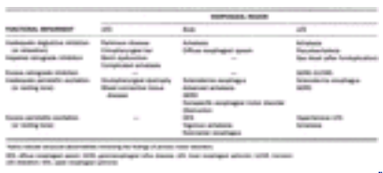

TABLE 59-2 Patterns of Esophageal Dysmotility*

Esophageal Manometry The clinical use of manometry is in defining the contractile characteristics of the esophagus in an attempt to identify pathological conditions. Manometric evaluation of the tubular esophagus assesses the success rate, rate of progression of the contractile complex, and characteristics of the contractile complex (amplitude, duration, repetitive contractions). *Failed peristalsis* is defined as the absence of contraction after a swallow or a contraction that proceeds only part way down the esophagus and either disappears or ends with a simultaneous contractions. Each of these outcomes results in incomplete esophageal emptying. [56] The progression rate of peristaltic contraction is measured by identifying the timing of the initial upstroke of the peristaltic contraction at adjacent recording sites and knowing the distance between these sites. Progression rates greater than 6.25 cm/s are associated with poor esophageal emptying and are considered a simultaneous contraction. [71] Contractile amplitude is considered hypotensive if it has an amplitude of less than 35 mm Hg because this, too, is associated with impaired esophageal emptying. [56] A typical convention for scoring hypertensive peristalsis is that the mean amplitude of contractile complexes is greater than 180 mm Hg, but this upper limit has varied among investigators. Repetitive contractions are multipeaked complexes. By convention, a contraction is scored as multipeaked if the valley between the peaks is at least 10 mm Hg less than and at least 1 second after the preceding peak. Both the magnitude of LES pressure and LES sphincter relaxation characteristics are determined from manometry. The three most widely used techniques are a stationary sleeve sensor, a rapid pull-through of side-hole sensors across the sphincter during suspended respiration, and a station pull-through of a side-hole sensor recording pressure activity for 30 to 60 seconds at 1 cm increments as the catheter is withdrawn. LES pressure varies by up to 25 mm Hg with the method of measurement used and with whether midrespiratory or end-expiratory LES pressure is reported. In addition to the varied methodology, the minute-to-minute variations of LES secondary to normal physiological events, such as the migrating motor complex, make it extremely difficult to establish normal ranges. Therefore, the most meaningful statement that can be made regarding an isolated measurement of LES pressure is that it is abnormal to have an extremely low value (<10 mm Hg). Relaxation characteristics can be evaluated using a sleeve

sensor, during a station pull-through, or after repositioning a side hole at the optimal position. A sleeve sensor is most suited to evaluate LESR, given that esophageal shortening occurs with swallowing. Deglutitive LESR has been characterized using a sleeve sensor in normal control subjects, in patients with GERD, in patients with achalasia, and in patients with diffuse esophageal spasm (DES). This system reliably records LESR characteristics and has a high diagnostic value in suspected achalasia. [72] Clinically, manometry can potentially aid in the diagnosis and management of esophageal syndromes involving dysphagia, chest pain, or GERD and in defining multisystem diseases that have esophageal dysmotility as one component. Unfortunately, along with pathophysiologically important abnormalities, manometry also detects insignificant aberrations of esophageal motility that have no proven relevance to the symptoms or management of patients with esophageal syndromes. This limited specificity and low prevalence rates of significant disorders in symptomatic patients make it a poor screening test, and, thus, it should never be the initial diagnostic test performed.

Achalasia

Achalasia is the most easily recognized and best-defined motor disorder of the esophagus. First recognized more than 300 years ago, the disorder was initially labeled cardiospasm, reflecting the observation that it was caused by a functional obstruction of the esophagus at the cardiac sphincter with no obstructing lesion evident in autopsy specimens. The first reported case was treated by passing a piece of carved whalebone with a sponge affixed to the distal end through the esophagus to facilitate esophageal emptying after meals. [73] That patient apparently sustained himself in this fashion for 15 years. During the next 2 centuries, there were sporadic reports of cases similarly treated with crude ramrods or dilators. In 1937, Lendrum [74] proposed that the functional esophageal obstruction in this syndrome resulted from incomplete relaxation of the LES and renamed the disease achalasia ("failure to relax"), ushering in our current concept of the disease.

Epidemiology Achalasia is a rare disease with an estimated incidence of 1 per 100,000 population per year in the United States and Europe. [75], [76] The incidence in Zimbabwean blacks may be much lower, raising the issue of racial or environmental predisposition. [77] The disease affects both sexes equally and usually presents in adult life, most commonly between the ages of 25 and 60 years. [78] Because achalasia is a chronic condition, its prevalence greatly exceeds its incidence. Estimates of the prevalence of achalasia in Europe range from 7.1 per 100,000 in Wales to 13.4 per 100,000 in Ireland. [79] Reports of familial clustering of achalasia raise the possibility of genetic predisposition; however, the data on this are inconclusive. Achalasia has been reported in monozygotic twins, [80] in siblings, [81] and in children of affected parents. [82] However, other reports of the occurrence of achalasia in only one of a pair of monozygotic twins speak against strong genetic determinant. [83] Emphasizing this point, a survey of 1012 first-degree relatives of 159 patients with achalasia identified no affected relatives. [84]

Neuropathology Achalasia is characterized by failure of the LES to relax completely with swallowing and aperistalsis in the smooth muscle esophagus. The resting LES pressure is elevated in about 60% of cases. If there are nonperistaltic, spasmlike contractions in the esophageal body, the disease is referred to as *vigorous achalasia.* These physiological alterations are thought to result from damage to the innervation of the smooth muscle esophagus (including the LES). Proposed neuroanatomic changes responsible for achalasia include loss of ganglion cells within the myenteric (Auerbach) plexus, degeneration of the vagus nerve, and degeneration of the dorsal motor nucleus of the vagus. Of these possibilities, the loss of ganglion cells is best substantiated. Several observers reported fewer ganglion cells and ganglion cells surrounded by mononuclear inflammatory cells in the smooth muscle esophagus of patients with achalasia. [85], [86] One report additionally noted ganglion cell degeneration extending into the proximal stomach in half of 34 specimens analyzed. [87] The degree of ganglion cell loss parallels the duration of disease such that ganglion cells are almost absent in patients afflicted for 10 years or longer. [88] A morphologic study of 42 esophagi resected from patients with advanced achalasia revealed diminished myenteric ganglion cells and inflammation within the myenteric plexus in all cases. [89] The ultimate cause of ganglion cell degeneration in achalasia is unknown; however, there is increasing evidence consistent with an immune-mediated process. Immunohistochemical analysis of the myenteric infiltrate in patients with achalasia revealed that most inflammatory cells are CD3/CD8-positive lymphocytes that express T-cell internal antigen-1 (TIA-1), indicating that these cells are either resting or activated cytotoxic T cells. [90] In addition to this histological support, there is also an association with the class II human leukocyte antigen antigens DQW1 [91] and DQB1. [92] Achalasia may also be associated with degenerative neurological disorders such as Parkinson disease. Patients with both achalasia and Parkinson disease were noted to have Lewy bodies (intracytoplasmic hyaline or spherical eosinophilic inclusions) in the degenerating ganglion cells of the myenteric plexus. [86] Physiological studies also provide evidence of at least partial postganglionic denervation of esophageal smooth muscle in achalasia. Such damage can potentially affect excitatory (cholinergic) ganglionic neurons, inhibitory (nitric oxide ± vasoactive intestinal polypeptide) ganglionic neurons, or both. Consider first the excitatory ganglionic neurons. Muscle strips from the circular layer of the esophageal body of patients with achalasia contract when they are directly stimulated by acetylcholine but fail to respond to ganglionic stimulation by nicotine, indicating a postganglionic excitatory defect. [93] An in vivo demonstration of smooth muscle sensitivity to acetylcholine is the Mecholyl test, in which the injection of this acetylcholine analog evokes profound contraction of the otherwise flaccid achalasic esophagus. [94] This exaggerated response has been interpreted as a demonstration of Cannon's law of denervation supersensitivity. However, it is likely that loss of excitatory innervation is variable among patients with achalasia. Partial preservation of the postganglionic cholinergic pathway is suggested by the observations that the LES pressure in patients with achalasia increases after administration of the acetylcholinesterase inhibitor edrophonium and decreases after administration of the muscarinic antagonist atropine. [95] These observations are crucial to understanding why botulinum toxin may have therapeutic benefit in achalasia (see later). Regardless of excitatory ganglionic neuron impairment, it is clear that the inhibitory ganglionic neurons are necessarily impaired as an early manifestation of achalasia. Functionally, these neurons are responsible for deglutitive inhibition (including LESR) and for the timing of propagation of esophageal peristalsis; their absence offers a unifying hypothesis for the key physiological abnormalities of achalasia: impaired LESR and aperistalsis. It is increasingly evident that the inhibitory ganglionic neurons use nitric oxide as a neurotransmitter. [96] In support of this conclusion, patients with achalasia have been shown to lack nitric oxide synthase in the gastroesophageal junction. [96] Furthermore, vasoactive intestinal polypeptide may be a cotransmitter in these neurons, and immunohistochemical studies have demonstrated a marked reduction of vasoactive intestinal polypeptide–staining neurons in patients with achalasia. [97] A multitude of evidence supports impaired physiological function of postganglionic inhibitory innervation in the smooth muscle esophagus of patients with achalasia. Animal models of achalasia have been established using nitric oxide inhibitors such as N^{G}-nitro-L-arginine. [98] Muscle strips from the LES of patients with achalasia do not relax in response to ganglionic stimulation as they do in normal controls. [93] Cholecystokinin octapeptide, which normally stimulates the inhibitory ganglionic neurons, thereby reducing LES pressure, paradoxically increases the LES pressure in patients with achalasia. [99] Impaired inhibitory innervation of the tubular esophagus above the LES is more difficult to demonstrate than within the sphincter itself because of the absence of resting tone in this region. However, in a clever experiment, Sifrim and colleagues [100] used an intraesophageal balloon to create a high-pressure zone in the tubular esophagus that then relaxed with the onset of deglutitive inhibition. This deglutitive relaxation in the esophageal body was absent in early, nondilated cases of achalasia.

Clinical Presentation Clinical manifestations of achalasia may include dysphagia, regurgitation, chest pain, weight loss, and aspiration pneumonia. All patients have solid food dysphagia; most patients also have variable degrees of liquid dysphagia. The onset of dysphagia is usually gradual, with the duration of symptoms averaging 2 years at presentation. [78] Dysphagia severity fluctuates, but eventually it plateaus. Predictably, regurgitation occurs when large amounts of food are retained in the dilated esophagus. The regurgitant is often recognized as food that has been eaten hours, or even days, previously. It tends to be nonbilious, nonacidic, and mixed with copious amounts of saliva. Chest pain is a frequent complaint early in the course of achalasia, occurring in approximately two thirds of patients. [101] Its cause is unknown, but it is speculated to be related to the occurrence of esophageal spasms or to the process of esophageal dilation associated with disease progression. Treatment of achalasia is less effective in relieving chest pain than it is in relieving dysphagia or regurgitation. However, unlike dysphagia or regurgitation, chest pain may improve or disappear over time. [101] An estimated 10% of patients with achalasia have bronchopulmonary complications as the result of regurgitation; in some instances, it is these complications rather than dysphagia that prompts them to seek medical care. [102] Other interesting, but fortunately rare, symptoms of achalasia are airway compromise and stridor as a result of the dilated esophagus compressing the trachea. [103] This occurs secondary to dysfunction of the belch reflex either because of neural degeneration or because esophageal dilation prevents activation of stretch receptors within the esophageal wall. [104] It is paradoxical that many patients with achalasia complain of heartburn, even after the onset of dysphagia. [105] Although reflux may be a common sequela of the treatments for achalasia, it seems physiologically inconsistent to have dysphagia from impaired LESR and reflux from excessive LESR simultaneously. In support of this skepticism, ambulatory 24-hour esophageal pH studies of patients with achalasia have shown only periods of esophageal acidification caused by the bacterial fermentation of retained food in the esophagus rather than discrete gastroesophageal reflux events. [106] Furthermore, prolonged LES recordings have shown a complete absence of tLESRs in patients with achalasia. [107] However, there are occasional exceptions to this, evident from a well-documented case of a patient with achalasia with intact tLESR despite the absence of deglutitive LESR. [108]

Radiographic and manometric findings. Barium swallow radiography (Fig. 59-9) or esophageal manometry (Fig. 59-10) can demonstrate the physiological abnormalities of achalasia. The characteristic x-ray finding is of a dilated intrathoracic esophagus with impaired emptying, an air-fluid level, absence of a gastric air bubble, and an LES that tapers to a point giving the distal esophagus a beaklike appearance. Occasionally, an epiphrenic diverticulum, immediately above the LES, is observed with achalasia. [109] With long-standing achalasia, the esophagus may assume a sigmoid configuration, and in some instances an air-fluid level, mediastinal widening, and outline of the dilated esophageal wall are even evident on a plain chest film. The characteristic radiographic findings depend on esophageal dilation. Because dilation is not always present in achalasia, sensitivity of the radiographic examination is limited. [76]

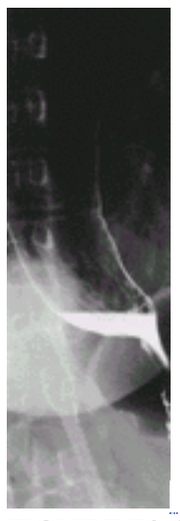

FIGURE 59-9. Characteristic barium swallow in idiopathic achalasia. Note esophageal dilation with an air-fluid level and the smooth tapering at the gastroesophageal junction. Radiographic findings can be much more subtle in the early phases of the disease.

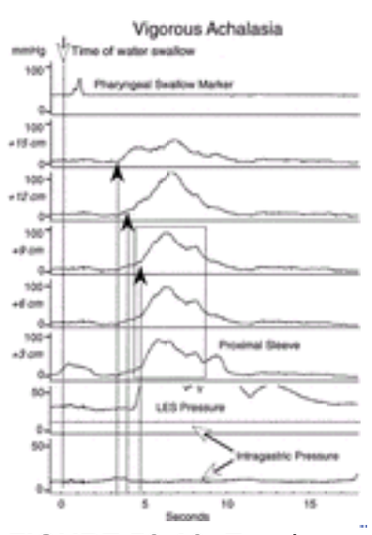

FIGURE 59-10. Esophageal manometric recording in a patient with idiopathic achalasia, in this case vigorous achalasia and a nondilated esophagus. The timing of the swallow is indicated by the *first vertical dotted line*. The *italicized numbers* to the **left** of the pressure calibrations indicate the position of each recording site relative to the midpoint of the sleeve sensor that is centered within the lower esophageal sphinter (LES). Although there is a propagated contraction in the proximal (striated muscle) esophagus, there is no propagated contraction in the smooth muscle esophagus, as indicated by the simultaneous onsets of the contraction waves. The *rectangle* surrounds a section of the tracing demonstrating isobaric waves across three esophageal recording sites. During this period of the recording, those three recording sites are contained within a “common cavity” sealed from above and below by higher-amplitude, lumen-obliterating contractions. The resting LES pressure is 20 mm Hg relative to intragastric pressure (indicated by the *horizontal dotted line*) and demonstrates minimal deglutitive relaxation.

The defining manometric features of achalasia are aperistalsis and incomplete LESR. In cases of achalasia established by all available clinical criteria, the defining manometric features are present in more than 90% of patients. An analysis of manometric LES parameters in patients with achalasia found that impaired LESR by itself (to a nadir value greater than 12 mm Hg) had a 92% sensitivity and 94% specificity for the detection of achalasia. [72] However, even though manometry is the current standard, there is manometric heterogeneity in achalasia such that manometric findings must sometimes be combined with additional clinical data to detect atypical cases, such as patients with only short segments of aperistalsis or quantifiable normal LESR. [108] Some patients exhibit higher-amplitude (>60 mm Hg) simultaneous repetitive contractions in response to swallows, thereby defining the variant known as vigorous achalasia. These patients in particular tend not to have esophageal dilation, making them difficult to diagnose radiographically. [110] In a comparison among diagnostic modalities, one prospective study showed that achalasia was suggested by the radiologist in only 21 of 33 patients who were given this diagnosis at manometry. [76] Other manometric features (increased resting LES pressure, increased intraesophageal baseline pressure, or isobaric waveforms) provide supportive evidence to improve the suspicion rate of achalasia.

Endoscopic findings. Endoscopy is relatively insensitive in the detection of achalasia; endoscopists prospectively suggest the correct diagnosis in fewer than one third of patients. [76] Typical endoscopic findings include retained food or saliva and dilation and atony of the esophageal body. With progressive dilation and stasis, erythema, friability, and superficial ulcerations may be seen. Whitish plaque covering the epithelial surface may be seen as the result of *Candida* overgrowth; this is usually asymptomatic. The achalasic LES has a pinpoint appearance and does not open with air insufflation. Nonetheless, the instrument should easily pass through the sphincter into the stomach with minimal pressure. Resistance, or a feeling of stiffness as the endoscope crosses the gastroesophageal junction, should immediately raise the suspicion of malignant disease (see section “ Pseudoachalasia”). Equivocal mucosal abnormalities of the gastroesophageal junction, evident on forward viewing or retroflexed inspection, should always undergo biopsy because of the possibility of pseudoachalasia.

Differential Diagnosis The differential diagnosis of achalasia includes both other esophageal motility disorders with functional attributes overlapping those of achalasia and diseases of distinct pathophysiology that duplicate the functional consequences of achalasia. With respect to other motility disorders, inspection of Table 59-2 suggests many similarities between esophageal spasm and achalasia, especially vigorous achalasia. In fact, the only distinction between these entities is the demonstration of incomplete LESR in vigorous achalasia. Thus, it has been speculated that in some instances esophageal spasm (and vigorous achalasia) may represent an early stage of achalasia and may evolve into full-fledged achalasia. Vantrappen and colleagues [111] first speculated on this relationship and reported several cases demonstrating just such an evolution. However, as discussed later in this chapter, spastic disorders of the esophagus are a heterogeneous lot, and a few, at best, are part of this continuum. With respect to diseases of distinct pathophysiology that duplicate the functional consequences of achalasia, the main considerations are Chagas disease and pseudoachalasia. These disorders are considered in some detail because they may resemble achalasia so closely that conventional diagnostic tests are misleading. A rare genetic achalasia syndrome has also been described; *familial adrenal insufficiency with alacrima* is inherited as an autosomal recessive trait that manifests itself with the childhood onset of autonomic nervous system dysfunction including achalasia, alacrima, sinoatrial dysfunction, abnormal pupillary responses to light, and delayed gastric emptying. [112]

Chagas disease. Although achalasia is almost universally idiopathic in North America, the disease can be closely mimicked by esophageal involvement in *Chagas disease,* which is endemic in areas of central Brazil, Venezuela, and northern Argentina. Cases have also been reported in southern Texas. Chagas disease is spread by the bite of reduvid (kissing) bug, which transmits the culprit protozoan, *Trypanosoma cruzi.* After infection, an acute septicemic phase of the illness develops that varies in severity from being so mild as to go unnoticed to being so severe as to be fatal. [113] The chronic phase of the disease develops many years after infection and results from destruction of autonomic ganglion cells throughout the body, including the heart, gut, urinary tract, and respiratory tract. Chronic cardiomyopathy with conduction system disturbances and arrhythmias is the most common cause of death. The digestive tract organs most often affected are the esophagus, duodenum, and colon. The severity of esophageal dysfunction is directly proportional to the degree of intramural ganglion cell loss; abnormal peristalsis is first detectable after 50% of ganglion cells are destroyed and esophageal dilation only after 90% are destroyed. [113] Paralleling this, the initial dysfunction is confined to the esophageal body, with LES dysfunction occurring late in the course of the disease. [113] The most useful clinical distinction between idiopathic achalasia and esophageal involvement in Chagas disease is evidence of other tubular organ involvement (megaureter, cardiomyopathy, megaduodenum, megacolon, megarectum) in the latter. Otherwise, patients with Chagas disease have the same clinical, radiographic, and manometric characteristics as those with idiopathic achalasia. The diagnosis of Chagas disease is confirmed by a serologic test using complement fixation or polymerase chain reaction. The treatment of the achalasia syndrome in Chagas disease is similar to that for idiopathic achalasia. Treatment of the infection itself is of limited efficacy in the acute phase of the disease and is of no proven efficacy in chronic disease.

Pseudoachalasia. Neither the radiographic nor the manometric features of achalasia are specific for idiopathic achalasia or achalasia associated with Chagas disease; tumor-related *pseudoachalasia* accounts for up to 5% of cases with manometrically defined achalasia. Pseudoachalasia is more likely with progressive age (>50 years), abrupt onset of symptoms (<1 year), and early weight loss in excess of 7 kg. [78] However, even though these criteria make pseudoachalasia more likely, they still have poor predictive value in the individual patient. [114] Tumor infiltration (especially carcinoma in the gastric fundus) can completely mimic the functional impairment seen with idiopathic achalasia [115] (see Table 59-2). It is because of this potential pitfall that a thorough anatomic examination including endoscopy should be done as part of the diagnostic evaluation of every new case of achalasia. A clue to the presence of pseudoachalasia on the endoscopic examination is more than the slightest resistance of passage of the endoscope across the gastroesophageal junction. In idiopathic achalasia, the endoscope should pop through with only

gentle pressure required. If suspicion of pseudoachalasia persists, endoscopic biopsy, computed tomography, magnetic resonance imaging, or endoscopic ultrasound should be considered for further evaluation, depending on the special circumstances. Adenocarcinoma of the gastroesophageal junction accounts for more than one half of pseudoachalasia cases, with a myriad of tumors and miscellaneous conditions accounting for the remainder. Within the spectrum of malignant diseases, pancreatic cancer, oat cell tumors, hepatoma, bronchogenic tumors, esophageal squamous cell carcinoma, prostate tumors, and lymphoma have been reported. [78] These tumors produce an achalasia syndrome by infiltrating the wall of the esophagus at the gastroesophageal junction, in essence causing a malignant obstruction at the LES with proximal esophageal dilation. [115] Similarly, pseudoachalasia has also been reported to result from esophageal infiltration by amyloid, [116] eosinophilic gastroenteritis, [117] and sarcoidosis. [118] Although often speculated in the literature, it is less certain, and certainly much less common, that an achalasic syndrome occurs as a paraneoplastic syndrome without direct tumor stenosis of the gastroesophageal junction. [78]

Management Because the underlying neuropathological condition cannot be corrected, treatment of achalasia is directed at compensating for the functional abnormalities and preventing complications. The main functional abnormality in achalasia is poor esophageal emptying, and this is treated by reducing the LES pressure. With reduced LES pressure, gravity promotes esophageal emptying. Peristalsis rarely returns with therapy. LES pressure can be reduced by pharmacological therapy, forceful dilation, or surgical myotomy. Pharmacological treatments, on the whole, are not very effective, perhaps indicated more as temporizing maneuvers than as definitive therapies. The definitive treatment of achalasia is still disruption of the LES either surgically (Heller myotomy) or with a pneumatic dilator. The optimal approach is an issue of debate, given the lack of randomized controlled trials with accepted criteria for assessing efficacy. Thus, there is no alternative but to compare therapies based on the numerous uncontrolled series that have been published using a variety of qualitative end points as indications of efficacy. Table 59-3 summarizes the pooled estimate of response rate of medical and surgical treatment data in achalasia. Data in the table are uncontrolled and were consecutive series collected both prospectively and retrospectively. Because there was no uniformity in assessment of efficacy among trials, the proportion with a good-to-excellent response was extracted from each study, regardless of criteria. Only studies including more than ten patients and with a follow up of 1 year were tabulated.

TABLE 59-3 Pooled Estimate of Response Rate of Achalasia Treatments across Referenced Studies*

Pharmacological therapy. Smooth muscle relaxants such as nitrates or calcium channel blockers, administered sublingually immediately before eating, may offer partial relief of the dysphagia accompanying achalasia by reducing the resting sphincter pressure. Anticholinergics, [119] amyl nitrite, [120] sublingual nitroglycerin, [121] theophylline, [121] and ß $_2$-agonists [121] have also been tried, with variable results. The greatest reported experience has been with isosorbide dinitrate and nifedipine. [122] Isosorbide dinitrate (Isordil), 5 to 10 mg sublingually before meals, reduces the resting LES pressure by 66% for 90 minutes, with clinical efficacy paralleling the magnitude of LES response. [123] A 19-month trial of this therapy has been reported to provide marked or complete relief of dysphagia. [123] Side effects, particularly headache, are common. Placebo-controlled trials have not been reported. Calcium channel blockers (diltiazem, nifedipine, verapamil) reduce the LES pressure by 30% to 40% for more than an hour with potential benefit in achalasia. [124], [125] The greatest clinical experience has been with nifedipine (Procardia), which can be administered at 10 to 30 mg sublingually (capsules are crushed in the mouth) 30 to 45 minutes before meals. The benefit of nifedipine, 30 to 40 mg sublingually per day, was assessed in a group of 29 patients with early achalasia (before esophageal dilation) in a placebo-controlled trial. Nifedipine was significantly better that placebo (which had no significant benefit), yielding good to excellent results in 70% of patients with achalasia who were followed-up for 6 to 18 months. [122] However, subsequent placebo-controlled crossover trials found only minimal clinical improvement with nifedipine. [126], [127] Limiting side effects of nifedipine use are flushing, dizziness, headache, peripheral edema, and orthostasis. Sildenafil is another smooth muscle relaxant that has been shown to decrease the LES pressure and deglutitive relaxation pressure in patients with achalasia by blocking phosphodiesterase type 5, the enzyme that destroys cyclic guanosine monophosphate induced by nitric oxide. A double-blind placebo-controlled trial revealed that 50 mg of sildenafil caused a significant decrease in LES pressure and relaxation pressure when compared with placebo. [128] The effect peaked at 15 to 20 minutes after administration and persisted for less than 1 hour.

Botulinum toxin injection. Pasricha and associates [129] first reported the effects of botulinum toxin on the LES in patients with achalasia in 1994. These investigators found that intrasphincteric injection of botulinum toxin decreased LES pressure by 33% and improved dysphagia in 66% of patients for a 6-month period. Botulinum toxin works by irreversibly inhibiting the release of acetylcholine from presynaptic terminals, effectively eliminating the neurogenic component of LES pressure. However, because this effect is eventually reversed by the growth of new axons, it is not surprising that the effect of botulinum toxin is not long lasting. Although most patients initially experience a good response, there is minimal continued efficacy at 1 year (see Table 59-3). [130], [131], [132] and [133] The technique for administering botulinum toxin involves injecting a total of 80 U into four quadrants of the LES with a sclerotherapy catheter. Doses higher than 100 U have not been shown to be more effective. [134] Studies comparing botulinum toxin with pneumatic dilation suggests that the added expense for repeated injection outweighs the potential economic benefits of the added safety, unless life expectancy is 2 years or less. [135] Thus, this therapeutic option should be mainly reserved for elderly or frail patients who are poor risks for more definitive treatments.

Pneumatic dilation. Therapeutic dilation for achalasia requires the forceful distention of the LES to a diameter of approximately 3 cm to disrupt the circular muscle of the sphincter partially and to effect lasting reduction of LES pressure. Dilation with an endoscope or with standard bougies (=60 F) provides only very temporary benefit. Only balloon dilators specifically designed to treat achalasia achieve adequate diameter for lasting effectiveness. The basic element of an achalasia dilator is a long, noncompliant, cylindrical balloon that can be positioned fluoroscopically (Rigiflex dilator [Microvasive, Millford, MA]) or endoscopically (Witzel dilator [Medi-Globe, Tempe, AZ]) across the LES and then inflated to a characteristic diameter in a controlled fashion using a handheld manometer. The technique of pneumatic dilation is variable among practitioners in terms of patient preparation, parameters of balloon inflation, and postdilation monitoring. There is general agreement that pneumatic dilation can be done on an outpatient basis with the patient under conscious sedation. In patients with substantial esophageal retention, it is useful to have them on a liquid diet for one or more days before the procedure. Reported balloon inflation pressures range from 360 mm Hg (7 psi) to 775 mm Hg (15 psi), with periods of inflation ranging from several seconds to 5 minutes. [136], [137] Although there is minimal methodological consistency among authors, a cautious approach of beginning with relatively low inflation pressures or a smaller-diameter dilator (3.0 to 3.5 cm) is fairly universal. The major complication of pneumatic dilation is esophageal perforation; mortality is very rare. [138] Most series report an incidence of esophageal perforation during pneumatic dilation of between 1% and 5%. [102], [136] Because most perforations are readily evident within a few hours of the procedure, patients should be observed closely for signs of an esophageal leak for 3 to 6 hours after pneumatic dilation. Alternatively, some practitioners routinely obtain a fluoroscopic examination of the esophagus after pneumatic dilation to make sure that perforation has not occurred. Usually, a water-soluble contrast agent is given first, followed by barium. If the perforation appears small and confined, or intramural, conservative management consisting of close observation while allowing the patient nothing by mouth and administering intravenous antibiotics is appropriate. [102] If any substantial perforation has occurred, or if worsening pain and fever occur during observation of what was thought to be a small perforation, surgical repair should be pursued quickly. Patients with perforation from pneumatic dilation that is recognized and promptly treated surgically (within 6 to 8 hours) have outcomes that are comparable to those of patients undergoing elective Heller myotomy. [139] The best predictor of efficacy after a pneumatic dilation is the postdilation LES pressure; neither sphincter relaxation nor peristaltic function is significantly changed. LES pressure values of less than 10 mm Hg are associated with prolonged remission, whereas patients with values greater than 20 mm Hg derive little benefit from the procedure. [140] It is neither unusual nor dangerous to repeat a pneumatic dilation two to three times in instances of an unsatisfactory result. The clinical efficacy of dilation has been reported to range between 32% to 98% (see Table 59-3). [130] Patients having a poor initial result or rapid recurrence of symptoms have diminished likelihood of responding to additional dilations. [141] /SUP>Subsequent response to surgical myotomy is not influenced by the history of previous dilations. [142]

Heller myotomy. The surgical objectives in the treatment of achalasia are to reduce the LES pressure enough to eliminate dysphagia but not enough to allow gastroesophageal reflux. Current procedures are variations on the esophageal myotomy described by Heller in 1913. [143] In Heller's operation, anterior and posterior myotomies were performed through either a laparotomy or a thoracotomy. Although clearly efficacious, open Heller myotomy is associated with considerable morbidity related to thoracotomy, which led most patients to pursue pneumatic dilation as the initial intervention. However, adoption of the laparoscopic approach for achalasia surgery has led many practitioners to use surgery as an initial intervention because this technique offers similar efficacy to the open procedure and substantially reduced morbidity. Surgical series of patients with achalasia who were treated with myotomy reported good to excellent results in 62% to 100% of patients, with persistent dysphagia troubling fewer than 10% of patients overall (see Table 59-3). [130] The appeal of myotomy is that it offers a more predictable method of reducing LES pressure than does pneumatic dilation. [144] Studies suggest that laparoscopic and thoracoscopic approaches are associated with similar efficacy and reduced morbidity when compared with myotomy through thoracotomy or laparotomy. [130], [145], [146], [147], [148], [149] and [150] Patti and associates [147] reported the clinical outcomes of 168 achalasic patients who underwent thoracoscopic myotomy (35 patients) or laparoscopic Heller myotomy accompanied by a partial fundoplication (133 patients). There were no deaths in this series, and only eight patients required reoperation. Relief of dysphagia was obtained in 93% of the patients who underwent the laparoscopic Heller myotomy and in 85% of those treated with thoracoscopic myotomy. Not only did the laparoscopic approach improve dysphagia more effectively, but also it was associated with shorter hospital stay and less postoperative gastroesophageal reflux. Historically, postmyotomy reflux in patients with achalasia was particularly severe. [151] However, with the availability of proton pump inhibitors, reflux is usually easily controlled, making these complications very unlikely. Thus, laparoscopic Heller myotomy combined with a partial fundoplication has

become the preferred surgical procedure for achalasia. Occasionally, patients fail to respond to dilation and myotomy and require more extensive surgical procedures. In extremely advanced or refractory cases of achalasia, esophageal resection with gastric pull-up or interposition of a segment of transverse colon or small bowel may be the only surgical option. [152], [153] Indications for this intervention include unresolvable obstructive symptoms, cancer, and perforation during dilation. Although excellent long-term functional results can be achieved, the reported mortality of this surgery is about 4%, consistent with the mortality rate of esophagectomy performed for other indications.

Medical versus surgical treatment. Although pharmacological therapy is simple and safe, it has become increasingly clear that this should be reserved for use as a temporizing measure while more definitive therapy is being considered. Thus, practically speaking, the choice is between pneumatic dilation and laparoscopic Heller myotomy as the primary therapy for achalasia. However, there are no prospective controlled trials comparing these treatments. One controlled trial exists comparing pneumatic dilation with myotomy through thoracotomy. That study reported 95% symptom resolution with myotomy and 51% symptom resolution in the dilation group, but the study was criticized for the methodology of pneumatic dilation used. [154] Most studies report symptom resolution in approximately 70% of patients with pneumatic dilation, substantially higher than the 51% reported in the controlled trial but still substantially lower than that reported in uncontrolled series of laparoscopic Heller myotomy (see Table 59-3). Furthermore, although laparoscopic Heller myotomy is invasive, its morbidity is low. Conversely, pneumatic dilation has a perforation rate that averages 3%. Even though these patients do well if the perforation is recognized and is operated on promptly, they require a thoracotomy. Thus, it appears that, in expert hands, laparoscopic Heller myotomy is likely the optimal initial therapy when one considers both the safety and the efficacy of the procedure.

Risk of squamous cell cancer. Numerous series report cases of squamous cell carcinoma developing in the achalasic esophagus, [155], [156] and the relative risk of developing squamous cell cancer has been estimated to be 33-fold relative to the nonachalasic population. [157] The pathogenesis of the carcinoma is obscure, but stasis and mucosal irritation may be precipitating factors. The tumors develop many years after the diagnosis of achalasia. Because the tumors often arise in a greatly dilated esophagus, symptoms can be delayed, and the neoplasms are large and advanced at the time of detection. These considerations raise the issue of surveillance endoscopy in patients with achalasia to detect early squamous cell cancer. However, an elegant analysis by Sandler and colleagues [158] of a database encompassing the entire Swedish population of 1062 patients with achalasia suggests that, after discounting incident carcinomas, the overall squamous cell cancer risk for patients with achalasia compared with age-matched controls is 17-fold, making for a 0.15% incidence of cancer among the patients with achalasia. The authors calculated that, if surveillance endoscopy were to be done annually, 406 examinations would need to be done in men and 2220 in women before a single potentially treatable tumor was found.

Spastic Disorders

The concept of *esophageal spasm* is credited to Hamilton Osgood, [159] based on his 1889 description of six patients with episodic chest pain and dysphagia: “sudden and often intense constriction in the epigastrium… with an arrest of food at the cardiac orifice.” From the time of that report until the present day, further insight into the entity of esophageal spasm has paralleled the development of imaging or measurement technology that could detect it. “Diffuse spasm of the lower part of the esophagus” was first described radiographically in 1934, [160] and the first manometric descriptions of spasm came in the late 1950s. [161] Introduction of the term *diffuse esophageal spasm* and our present concept of this entity followed the description in 1967 by Fleshler [162] of a “clinical syndrome characterized by symptoms of substernal distress or dysphagia or both, the roentgenographic appearance of localized, nonprogressive waves (tertiary contractions), and an increased incidence of nonperistaltic contractions recorded by intralumenal manometry.”

In broad terms, DES as described by Fleshler represents a disorder of peristalsis. However, the esophagus of the patient with DES usually retains its ability to propagate normal primary peristaltic waves most of the time, suggesting that the responsible neuromuscular disorder must be more subtle than with achalasia. Partly because of this fact, the criteria for diagnosing DES (which is the best accepted of proposed disorders of peristalsis) remain variable and confusing. [163] Furthermore, along with the evolution of manometric techniques capable of accurately recording intralumenal esophageal contractile activity came the description of other minor aberrations of peristalsis (nutcracker esophagus, hypertensive LES, and nonspecific esophageal motor disorder) to which symptoms, especially chest pain, may be attributed. [164] Despite this proliferation of literature, whether these entities have any clinical significance remains controversial. The heterogeneity among patients, the absence of specific pathological features, and the absence of well-defined clinical implications caution against considering them akin to achalasia. Strict criteria for defining a disease would stipulate that the peristaltic abnormality cause a major alteration of esophageal physiology demonstrable by both manometric and nonmanometric measures, that it be temporally associated with symptoms, and that its correction result in symptomatic improvement. [165] If these criteria are applied to these aberrations of peristalsis, only severe DES would be judged significant. [70], [165] With this caution in mind, consider what has been learned of spastic disorders of peristalsis.

Neuromuscular Pathology The esophageal muscularis propria and myenteric plexus are not readily accessible for biopsy; the result is that little material is available for histopathological examination. Furthermore, because patients with spastic disorders of the esophagus rarely undergo esophageal surgery and the diseases are not fatal, making for a paucity of pathological specimens; the limited information that does exist pertains only to DES, as opposed to the minor aberrations of peristalsis. The most striking reported pathological change is diffuse muscular hypertrophy or hyperplasia, mainly of the distal two thirds of the esophagus. Muscular thickening of up to 2 cm has been reported in patients with clinical and manometric evidence of DES. [166] However, there are other well-documented cases of spasm in which esophageal muscular thickening was not found at thoracotomy, [167] and still other instances of patients with muscular thickening not associated with DES symptoms. [168] Similarly, little evidence of neuropathology has been reported; diffuse fragmentation of vagal filaments, increased endoneural collagen, and mitochondrial fragmentation have been described, but the significance of these findings is unclear. [166]

Pathophysiology of Spastic Disorders of Peristalsis Despite the absence of defined histopathology, physiological evidence implicates myenteric plexus neuronal dysfunction in spastic disorders of the esophagus. During peristalsis, vagal impulses reach the entire smooth muscle segment of the esophagus simultaneously and activate myenteric plexus neurons between the longitudinal and circular muscle layers. [169] Ganglionic neurons then intervene between the efferent vagal fibers and the smooth muscle, belonging to either an inhibitory population (using nitric oxide as a neurotransmitter) that hyperpolarize the muscle cell membrane and inhibit contraction or to an excitatory (cholinergic) population that depolarizes the membrane thereby prompting contraction. Thus, the instantaneous activity of the musculature at each esophageal locus is determined by the balance between these controlling influences from the myenteric plexus. Experimental evidence suggests heterogeneity among patients with spastic disorders such that some primarily exhibit a defect of inhibitory interneuron function, whereas in others the defect is of excess excitation. Two in vivo experiments implicated a defect of myenteric plexus inhibitory interneuron function in the genesis of simultaneous contractions in the distal esophagus. Behar and Biancani [170] timed the propagation of a swallow-induced contraction in physiologically normal persons and in a group of patients whose dominant manometric abnormality was of a simultaneous contraction in the distal 10 cm of esophagus. Figure 59-11 illustrates the key finding of that work. Within the proximal (striated muscle) esophagus, the two groups exhibited similar contraction propagation, consistent with this timing’s being the result of the sequenced activation of motor units by vagal efferents programmed within the medullary swallowing center. However, once entering the smooth muscle segment, the patients’ contractions diverged from those of the normal subjects and resulted in a simultaneous contraction in the distal esophagus. The duration and amplitude of contraction at each esophageal locus were normal, but the progressive delay of initiation of that contraction, a function attributable to increasing influence of inhibitory interneurons in the distal esophagus, was absent. Furthermore, when these persons swallowed twice within a 5-second interval, there was no deglutitive inhibition of the first peristaltic contraction within the smooth muscle esophagus, as is observed in normal subjects. Another experiment demonstrating impaired deglutitive inhibition in DES comes from work using an artificial high-pressure zone within the distal esophagus, as illustrated in Figure 59-7. Patients with motor disorders characterized by rapidly propagating or simultaneous contractions exhibited only partial relaxation of the artificial igh-pressure zone, proportional to the impairment of propagation velocity. [100] Taken together, these findings strongly suggest that one potential neuropathological process in DES is a selective, intermittent dysfunction of myenteric plexus inhibitory interneurons.

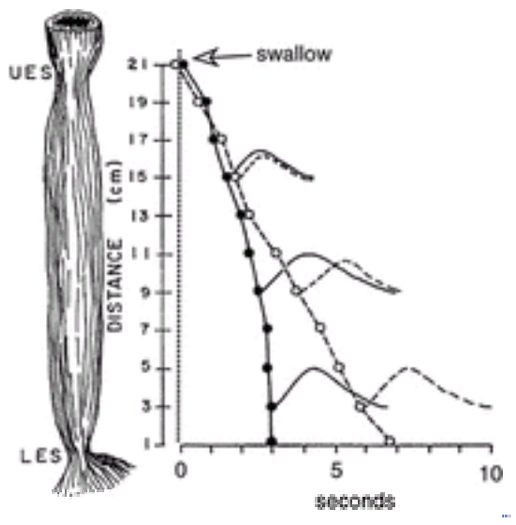

FIGURE 59-11. Rate of propagation of an esophageal contraction in persons with normal peristalsis (*open circles*) and in a group of patients with a motor disorder characterized by dysphagia and simultaneous contractions in the distal esophagus (*filled circles*). The propagation is similar between the two groups in the proximal (striated muscle) esophagus but diverges in the distal (smooth

muscle) esophagus. Because the prolonged latency in the distal esophagus is believed to be a function of postganglionic inhibitory interneurons with the myenteric plexus, this finding suggests selective dysfunction of these cells. (From ref. [170].)

As outlined earlier, dysfunction of the excitatory population of interneurons within the myenteric plexus, or abnormal excitability of the muscle itself, can also cause an aberration of peristalsis. Several lines of evidence suggest instances of excess excitation in response to hormonal or cholinergic stimulation. Representative of such an abnormality is a second group of patients in the analysis of Behar and Biancani [170] whose main manometric abnormality was of frequent spontaneous distal esophageal contractions. These patients had normal propagation latency, as defined by the parameters of Figure 59-11, but a significantly longer and higher amplitude contraction at each locus within the smooth muscle. [170] On examining the categorization of disorders of peristalsis outlined in Table 59-4, these individuals would be labeled as having nutcracker esophagus. Patients with peristaltic disorders characterized by excess excitation demonstrate heightened sensitivity to stimulation with cholinergic agents, [171], [172] the cholinesterase inhibitor edrophonium, [173] pentagastrin, [174] and ergonovine. [175] An electromyographic correlate of this excitability is found from bipolar ring electrode recordings from the distal esophagus. [176] Whereas physiologically normal persons uniformly exhibited spiking activity before each esophageal contraction, patients with DES exhibited spike-independent spontaneous esophageal contractions.

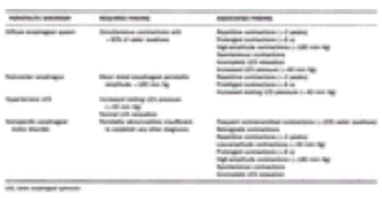

TABLE 59-4 Manometric Criteria for Disorders of Peristalsis

The foregoing discussion suggests that the physiological abnormalities of patients with spastic disorders are heterogeneous, but all are characterized by an imbalance between the excitatory and inhibitory influences on the esophageal smooth muscle. The suggestion of an impairment of the pathway of deglutitive inhibition is particularly interesting in that it places DES in a pathophysiological continuum with achalasia, consistent with documented case reports of patients undergoing this evolution. [111] Furthermore, there are marked similarities between vigorous achalasia and DES; both are characterized by simultaneous contractions of the esophageal body; the only differences being involvement of the LES and constancy of the disorder in vigorous achalasia. As in achalasia, the simultaneous contractions typifying DES impair bolus transit through the esophagus, potentially explaining the associated dysphagia. [177] Considerable attention has been focused on the clinical significance of the disorders of peristalsis other than DES described in Table 59-4, particularly *nutcracker esophagus.* Esophageal bolus transit is normal in this condition, [178] and no associated neuromuscular pathology is recognized. A few instances in which patients with nutcracker esophagus were subsequently diagnosed with achalasia have been reported, [179], [180] but considering the rarity of this observation and the high prevalence of nutcracker esophagus, the association is most likely explained by chance. No correlation has been shown between the severity of nonspecific peristaltic abnormalities and the severity of symptoms. [181] Long-term observation has shown neither a parallel in clinical course with manometric findings nor consistency in manometric diagnosis over time. [182] A therapeutic trial with nifedipine showed that significant reduction of wave amplitude in patients with nutcracker esophagus was not accompanied by a significant reduction in chest pain. [183] Conversely, a controlled trial of trazodone produced significant symptomatic improvement but no change in manometric findings. [184] One controlled trial demonstrated an association of symptomatic and manometric improvement with diltiazem, but the correlation for individual patients was not reported. [185] Finally, provocation of nonspecific motility abnormalities with cholinergic stimulation has not proven useful in detecting patients who are subsequently responsive to pharmacological treatment. [186], [187] In summary, it has been difficult to determine a direct relevance of nonspecific motility disturbances to either symptoms or function, making their detection of no generalizable clinical value.

Epidemiology No population-based studies exist on the incidence or prevalence of esophageal motility disorders other than achalasia. Thus, the only way to estimate the incidence or prevalence of spastic disorders is to examine the data on populations at risk and to reference the observed frequency of spastic disorders to the incidence of achalasia which, as detailed earlier, is about 1 per 100,000 population. Populations at risk for motility disorders are patients with chest pain or dysphagia, so it is among these patients that extensive manometric data have been collected. As detailed in Table 59-5, summarizing representative studies of at least 30 subjects, manometric abnormalities are prevalent among these groups. [155], [164], [188], [189], [190], [191], [192], [193], [194], [195], [196] and [197] Asymptomatic volunteers (usually younger than the patients) were evaluated to define normal values in five of these studies. [155], [189], [196], [197] and [198] The manometric pattern of achalasia was not detected in any volunteer. Similarly, although DES is more variably defined than is achalasia, [163] simultaneous contractions after at least 30% of swallows were not found in any volunteer. [196]

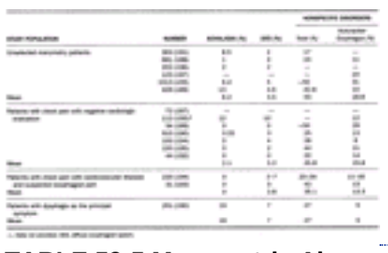

TABLE 59-5 Manometric Abnormalities in Patients with Chest Pain or Dysphagia

Most manometric findings among the patients in Table 59-5 fall under the categories other than DES in Table 59-4. Of the group, increased wave amplitude (nutcracker esophagus) is the most commonly detected pattern, accounting for a large portion of patients in the nonspecific disorders category. Variability in the prevalence values of the nonspecific disorders in Table 59-5 is partly attributable to a lack of uniform definitions of the motility disorders listed as well as to the inhomogeneity of patient populations studied. [70]

Clinical Presentation The major symptoms of spastic disorders are dysphagia and chest pain; regurgitation is infrequent compared with achalasia. The prevalence of chest pain is constant among the spastic disorders listed in Table 59-4, but the prevalence and severity of dysphagia are greater with DES. [181] Among the patients with chest pain as a dominant symptom, there is a high prevalence of psychiatric diagnosis, particularly anxiety and depression. [199] Evidence also exists suggesting a lower visceral pain threshold in this group; balloon distention and esophageal compliance studies reproduce esophageal pain at low distending volumes without corresponding motor changes. [200], [201] Finally, symptoms compatible with the irritable bowel syndrome may be seen in more than 50% of these patients. [202] Dysphagia for both liquids and solids is reported by 30% to 60% of patients with spastic disorders. [181], [190] Dysphagia is usually intermittent, sometimes related to swallowing specific substances or liquids at extreme temperatures. Dysphagia is usually not progressive, and weight loss is rare. In some instances, patients with spastic disorders experience episodes of esophageal obstruction while eating that persists until relieved by emesis; these instances are probably related to simultaneous prolonged DES-type contractions. More commonly, the dysphagia reported by patients with spastic disorders does not prevent or prolong eating. Intermittent substernal chest pain is reported by 80% to 90% of patients with spastic disorders. [181], [190] Esophageal chest pain is very similar to angina; usually described as crushing or squeezing, often radiating to the neck, jaw, arms, or midline of the back. Pain episodes may last from minutes to hours, but swallowing is usually not impaired during these episodes. Severe pain episodes may require narcotics or nitroglycerin for relief, further confusing the distinction between esophageal and cardiac pain. The mechanism producing pain is poorly understood; transient ischemia, lumenal distention, and altered visceral sensitivity have all been hypothesized. [200], [203], [204] More likely, the mechanism responsible for esophageal pain is variable among individual patients.

Endoscopic, radiographic, and manometric findings. Spastic motor disorders of the esophagus have no pathognomonic endoscopic features. However, endoscopy is useful in the evaluation of patients with dysphagia or suspected esophageal pain to identify either structural lesions or reflux esophagitis. Typically, spastic disorders are defined by manometry or barium radiography, the abnormal motor events are usually intermittent, and there are no uniform radiographic or manometric definitions. In the unequivocal case, nonperistaltic, high-amplitude, prolonged contractions are seen during esophageal manometry, and these are associated with the patient's experiencing pain (Fig. 59-12). These unequivocal cases are probably associated with a defect in the neuronal architecture of the myenteric plexus, placing them along the continuum of vigorous achalasia and achalasia (see Table 59-2). [100] The LES typically functions normally. Radiographically, "corkscrew esophagus," "rosary bead esophagus," pseudodiverticula, or curling are indicative of DES (Fig. 59-13). It must be emphasized, however, that neither tertiary contractions (nonperistaltic, simultaneous esophageal contractions) on radiographs nor occasional simultaneous contractions on manometry are pathognomonic of esophageal spasm; both may be seen in asymptomatic persons. [164]

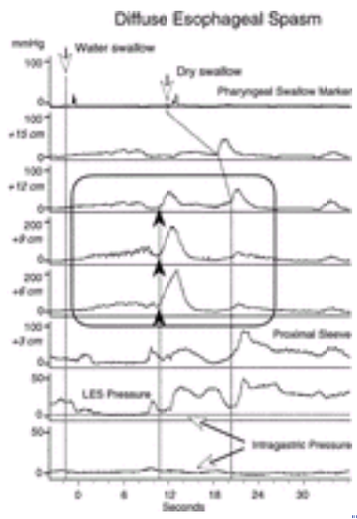

FIGURE 59-12. Esophageal manometric recording in a patient with symptomatic diffuse esophageal spasm. The timing of the swallow is indicated by the *first vertical dotted line*. The *italicized numbers* to the **left** of the pressure calibrations indicate the position of each recording site relative to the midpoint of the sleeve sensor that is centered within the lower esophageal sphincter (LES). The resting LES pressure is 20 mm Hg relative to intragastric pressure (indicated by the *horizontal dotted line*) and demonstrates normal deglutitive relaxation. The *rectangle* surrounds a section of the tracing demonstrating high-amplitude, prolonged, simultaneous, repetitive contractions in the smooth muscle esophagus. The most prominent simultaneous contraction is indicated by the *solid arrowheads* connected by a *vertical dotted line*. After the subsequent dry swallow, there is a propagated contraction in the proximal (striated muscle) esophagus, but this also terminates with simultaneous contraction waves in the smooth muscle. This patient's symptoms were dysphagia and chest pain.

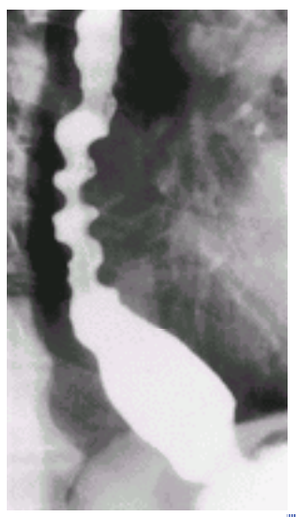

FIGURE 59-13. Corkscrew esophagus on barium radiography in a patient with symptomatic diffuse esophageal spasm.

One hypothesized explanation for the limited sensitivity of esophageal manometry in detecting spasm is that abnormal motor events escape detection because of sampling error. Because both chest pain and spasm are sporadic events, the likelihood of documenting both in a brief bedside study seems slim. This perception fostered the development of ambulatory esophageal manometry systems capable of longer recording intervals with the hope of correlating spontaneously occurring symptoms with abnormal esophageal motility. Unfortunately, despite multiple examinations of this technology, no consensus exists among investigators regarding scoring techniques or on the utility of this technology in establishing abnormal esophageal motility as the cause of "noncardiac" chest pain. One study, using combined ambulatory manometry and pH studies on a highly selected group of patients acutely hospitalized for attacks of chest pain, showed that up to 43% of patients have pain associated with episodes of acid reflux, but there was little additional yield from the manometric data. [205] Thus, at present, ambulatory manometry remains an investigational technique, with scoring criteria and symptom-association criteria remaining to be established. Variability in results evident in published trials can easily be explained by differences in patient populations studied and analysis schemes used. Furthermore, the evolving picture suggests that motor abnormalities play a less significant role than previously suggested in chest pain, and GERD disease plays a much more significant role. [206] Bancewicz and associates [191] found that intensive antireflux therapy was the most useful therapy for a large group of symptomatic patients without a specific motor disorder. Similarly, in another study, Achem and colleagues [207] found that antireflux therapy benefited patients with unexplained chest pain, regardless of the presence or absence of nonspecific motor abnormalities.

Differential Diagnosis Although abnormal peristalsis can potentially cause dysphagia and chest pain, significant abnormalities are rare, as suggested by the data in Table 59-5. Furthermore, esophageal dysmotility is not life-threatening, as is angina pectoris, the pain of which it can closely mimic. Features suggesting an esophageal as opposed to a cardiac etiology include the following:

- Pain that is nonexertional and prolonged
- Pain that interrupts sleep
- Pain related to meals
- Pain relieved with antacids
- The presence of additional esophageal symptoms of heartburn, dysphagia, or regurgitation.

However, each of these characteristics still exhibits overlap with cardiac pain in some instances. Thus, an esophageal origin of chest pain should be considered only after careful consideration and evaluation of potential cardiopulmonary causes. Furthermore, even within the spectrum of esophageal diseases, neither chest pain nor dysphagia is specific for a spastic disorder because both symptoms are also characteristic of common esophageal disorders including peptic or infectious esophagitis. *After these more common diagnostic possibilities have been excluded* by appropriate radiographic evaluation, endoscopic evaluation, and in some instance, a therapeutic trial of antisecretory medications, spastic disorders should be considered as the cause of the still unexplained symptoms.

Treatment Ironically, the medical therapies of esophageal spasm are similar to those of coronary artery disease, a disease with which it can be confused. However, despite the dogma of treatment with smooth muscle relaxants, there is a paucity of data on the medical treatment of esophageal spasm. Long-term outcome studies of the medical treatment of DES with smooth muscle relaxants are not available. Nitrates, [208] calcium channel blockers, [209] and hydralazine [210] have all been shown to be beneficial in small trials. In addition, botulinum toxin injected at the esophagogastric junction has also been used with some success in patients with nonachalasic esophageal spasm. [211] The only double-blind placebo-controlled trial showing efficacy with medical therapy was in the case of the anxiolytic trazodone, suggesting that reassurance and control of anxiety are important therapeutic goals. [184] Consistent with this conclusion, successful management of symptoms associated with spastic motility disorders have also been reported using behavioral modification programs and biofeedback. [212] Although the rationale is unclear, esophageal dilation with standard mercury-filled dilators has also been suggested as a therapy for dysphagia or chest pain in patients with spastic disorders. However, in the only controlled trial of this therapy, dilation with an 8-mm ("placebo") dilator was as effective as an 18-mm dilator in producing transient symptom relief. [213] Alternatively, pneumatic dilation has been used in patients with DES and severe dysphagia. [102], [214] In one practitioner's experience, 45% of patients with DES noted relief, compared with 80% of patients with achalasia. [102] In another series of nine patients with DES and LES dysfunction, dysphagia but not chest pain was improved during 37 months of observation. [214] However, it is not clear what distinguishes that group of patients from patients with vigorous achalasia, implying that patients likely to derive benefit from pneumatic dilation are those with mixed features of achalasia and DES, as described by Vantrappen and colleagues. [111] If dysphagia becomes so severe that weight loss is observed or if pain becomes unbearable, surgical therapy consisting of a Heller myotomy across the LES with proximal extension of the incision to include the involved area of spasm should be considered. [215], [216] However, there are no controlled studies of this treatment in patients with well-defined DES, and the indication for this procedure is extremely rare. In summary, at this point, the therapy of esophageal spastic disorders is as poorly defined as are the spastic disorders themselves. Clearly, most esophageal pain is attributable to reflux, irrespective of the presence or absence of minor motility abnormalities, and antireflux therapy should be extensively pursued before attempting therapy of a spastic disorder. Within the universe of motility disorders, achalasia stands alone as a well-defined

disease. Partially overlapping achalasia is DES, a disorder characterized by a severe peristaltic defect with functional and symptomatic consequences in some cases. Interestingly, DES most likely responds to specific therapy when it exhibits characteristics indicative of vigorous achalasia, implying that it is part of a pathophysiological continuum in these cases (Fig. 59-14). Alternatively, episodic esophageal spasm may be a manifestation of GERD, a disorder for which very effective therapy exists.

FIGURE 59-14. The universe of esophageal motility disorders. Some overlap exists among adjacent entities, as suggested by the Venn diagram. The attributes indicated on the **left** are increasingly relevant going down the diagram, whereas those at the **right** are increasingly applicable moving up the diagram. (From Kahrilas PJ. Esophageal motility disorders: current concepts of pathogenesis and treatment. Can J Gastroenterol 2000;14:221.)

With respect to other esophageal motility disorders, the current therapy is as poorly defined as are the disorders themselves. Although in extreme circumstances these disorders (nutcracker esophagus, hypertensive LES, and nonspecific disorders) may mimic DES with regard to symptoms and can exhibit profound esophageal hypercontractility, there are no significant controlled data suggesting that these patients benefit from antispasmodic therapy. In view of the unproven value of detecting these manometric aberrations, practice guidelines do not support pursuing these findings in the evaluation of patients with chest pain. Rather, until the meaning of these aberrations of esophageal motility are clarified, we should not overlook therapy aimed at the most common esophageal disorder, GERD, or more global conditions such as depression or somatization neurosis that are coexistent in these patients

Esophageal Involvement in Systemic Disease

Esophageal dysmotility occurs as a manifestation of several disease processes with the potential to affect smooth muscle or the autonomic nervous system. Included are collagen-vascular diseases, infiltrative disease such as amyloidosis or malignant diseases, metabolic disorders such as thyrotoxicosis and diabetes, and caustic injury as occurs with GERD. However, esophageal involvement is well characterized only with collagen-vascular diseases, diabetes, and GERD. Dysmotility associated with GERD is discussed in Chapter 60; the following discussion focuses on esophageal involvement in the collagen-vascular diseases and diabetes.

Scleroderma *Scleroderma* causes diffuse fibrosis and degenerative changes in the skin and synovium. The disease also has the potential to involve the heart, kidneys, lungs, intestines, and esophagus. White women between 30 and 50 years of age are the most commonly afflicted. Two forms of scleroderma are recognized: progressive systemic sclerosis with diffuse scleroderma (the more fulminant form with early involvement of internal organs) and the CREST syndrome (calcinosis, Raynaud phenomenon, esophageal dysfunction, sclerodactyly, and telangiectasia). The basic disease process is of smooth muscle atrophy with subsequent fibrosis. With either form of scleroderma, the esophagus is involved in 75% to 85% of cases. [217], [218] Pathological changes are confined to the smooth muscle portion of the esophagus resulting in aperistalsis and atony of the LES. [219] The main clinical manifestations of scleroderma esophagus are dysphagia and heartburn. Symptomatic patients usually have Raynaud phenomenon, but the severity of the esophageal disease does not covary with the disease severity in other organs. [220] The prevalence of erosive esophagitis may be as high as 60%, with reported cases of Barrett metaplasia and Barrett adenocarcinoma. [221], [222] Dysphagia may be attributable to poor peristalsis or to a stricture complicating the peptic esophagitis (seen in as many as one third of patients). [222] Radiographic findings typically consist of a slightly dilated aperistaltic esophagus and free reflux. Wide-mouth diverticula of the esophagus, similar to those described in the intestines, have also been seen. [223] Manometric abnormalities consist of a hypotensive or absent LES pressure, hypotensive to absent distal esophageal peristalsis, and normal proximal esophageal peristalsis. [217] There is no specific treatment for the esophageal involvement in scleroderma. Gastroesophageal reflux should be identified and treated commensurate with its severity. Strictures may require frequent dilation, and in rare cases of intractable esophagitis, antireflux surgery may be required. [224]

Other Collagen Vascular and Connective Tissue Diseases Esophageal symptoms are uncommon in *systemic lupus erythematosus,* although 25% to 35% of unselected patients have the manometric findings of hypotensive peristalsis and a hypotensive LES. [225], [226] *Mixed connective tissue disease* exhibits a mixture of clinical features found in scleroderma, polymyositis, and systemic lupus erythematosus and is characterized by high titers of a circulating antibody for a nuclear ribonucleoprotein antigen. More than 60% of patients with mixed connective tissue disease have esophageal involvement defined by cineradiography, [227] and up to 82% of patients have manometric findings. [225] Abnormalities of both smooth and skeletal muscle are found. In the largest report to date, 5 of 17 patients had a manometric pattern consistent with scleroderma, and 10 patients had aperistalsis of the entire esophageal body along with low pressures in both the UES and LES. [225]

Diabetes Mellitus More than 60% of patients with *diabetes* with peripheral or autonomic neuropathy and an occasional patient without neuropathy have esophageal manometric abnormalities. [228] Reported manometric abnormalities associated with diabetes include hypotensive peristalsis, frequent failed peristalsis, hypotensive LES with impaired deglutitive relaxation, simultaneous contractions, and repetitive contractions. [228], [229] and [230] The significance of these abnormalities is uncertain because most of these patients are asymptomatic. [228] Histological and pharmacological studies suggest that the esophageal abnormalities are secondary to the degenerative effects of diabetes mellitus on the autonomic nervous system, rather than smooth muscle dysfunction.

Presbyesophagus Manometric studies from the 1960s on men more than 80 years old suggested that aging was associated with failed peristalsis, decreased LESR, and an increase in spontaneous contractions. [231] Symptoms correlated poorly with these findings, leading to introduction of the term *presbyesophagus* to describe incidental asymptomatic peristaltic abnormalities in elderly patients. In 1974, a study of patients who were more than 70 years old and who were carefully screened to rule out diabetes or neurological disease found no increased incidence of peristaltic abnormalities in elderly persons compared with young control subjects. [232] The only difference found was decreased peristaltic amplitude in the elderly group, suggesting that age in of itself does not lead to peristaltic dysfunction. Thus, cases of “presbyesophagus” are likely related to comorbid conditions commonly encountered in elderly patients.

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CHAPTER 60

Joel E. Richter

GASTROESOPHAGEAL REFLUX DISEASE

Gastroesophageal reflux disease (GERD) results from the failure of the normal antireflux mechanism to protect against frequent and abnormal amounts of gastroesophageal reflux (GER), that is, the effortless movement of gastric contents from the stomach to the esophagus. GER is not itself a disease, but a normal physiological process. It occurs in virtually everyone, multiple times everyday, especially after large meals, without producing either symptoms or signs of mucosal damage. In contrast, GERD is a spectrum of disease usually producing symptoms of heartburn and acid regurgitation. Most patients have no visible mucosal injury at the time of endoscopic examination (nonerosive GERD), whereas others have esophagitis, peptic strictures, Barrett esophagus, or evidence of extraesophageal diseases such as chest pain, pulmonary symptoms, or ear, nose, and throat symptoms. GERD is a multifactorial process, one of the most common human diseases, and of economic importance, contributing to the expenditure in the United States of 4 to 5 billion dollars per year for antacid medications.

EPIDEMIOLOGY

The prevalence of GERD differs, depending on whether the analysis is based on symptoms (primarily heartburn) or signs (i.e., esophagitis) of disease. When based on symptoms, GERD is common in Western countries. For example, in a nationwide population-based study by the Gallup Organization in the United States, 44% of the population reported heartburn at least once a month. [1] More convincing data were obtained by Locke and colleagues, [2] who mailed out 2200 validated self-report questionnaires to a predominantly white population residing in Olmsted County, Minnesota. The prevalence of heartburn and acid regurgitation in the past 12 months was noted to be 42% and 45%, respectively. Frequent symptoms (at least weekly) were reported by 20% of respondents, with an equal gender distribution across all ages. The majority reported that heartburn was of moderate severity and had a duration of 5 years or more, and only 5.4% reported a physician visit for reflux complaints within the previous year. More variable prevalence rates for symptomatic GERD have been reported from Europe, ranging from 5% in Switzerland to 27% in Finland. [3]

Conversely, the true prevalence of esophagitis is very difficult to define because healthy persons rarely undergo endoscopic procedures. Studies suggest that 7% of persons in the United States have erosive esophagitis, whereas European studies identify prevalence rates ranging from 2% to 10%. [4] Analyses of the gender ratio of GERD show nearly equal proportions of affected men and women, but a male predominance occurs in esophagitis and Barrett esophagus. [4] Increasing age is an important factor in the prevalence of GERD complications, probably the result of cumulative acid injury to the esophagus over time. [5]

In contrast, the prevalence of GERD and its complications is relatively low among residents of Africa and Asia. For example, a cross-sectional study in Singapore reported prevalence rates for reflux symptoms of 7.5% in Indians, 0.8% in Chinese persons, and 3.0% in Malays. [6] Possible reasons for the lower GERD prevalence includes low dietary fat, lower body mass index, and lower maximal acid output related to infection with *Helicobacter pylori.* [7], [8]

The prevalence of GERD is increasing in Western countries. El-Serag and Sonnenberg [9] observed opposing time trends in the prevalence of peptic ulcer disease and GERD in the United States: rates of peptic ulcer and gastric cancer fell between 1970 and 1995; the prevalence of GERD and esophageal adenocarcinoma rose significantly. The authors speculated that the decreasing prevalence of *H pylori* may be playing a contributory role to the increasing prevalence of GERD. Data suggest that *H pylori*–induced gastritis involves both the antrum and corpus, affecting the parietal cells and thus reducing acid secretion and elevating gastric pH. [8] This may have a protective influence on the esophageal mucosa in patients susceptible to GERD. Long-term epidemiologic studies will be required to address this question more appropriately.

Other noteworthy epidemiologic observations about GERD are that it is more common in whites than in African Americans or Native Americans, [4] family clustering has been reported, [10] and it is rarely the cause of death. [4] GERD, however, is associa- ted with considerable morbidity and with complications such as esophageal ulcerations (5%), peptic stricture (4% to 20%) and Barrett esophagus (8% to 20%). Furthermore, GERD as a chronic disease significantly impairs quality of life. As compared with other chronic medical conditions, the impairment of quality of life resulting from GERD is similar to, or even greater than, that resulting from arthritis, myocardial infarction, heart failure, or hypertension [11] (Fig. 60-1).

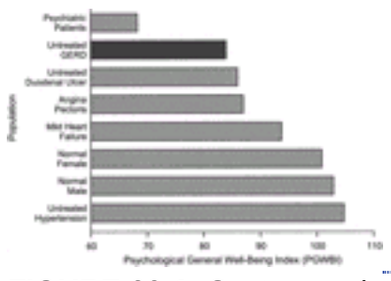

FIGURE 60-1. Gastroesophageal reflux disease (GERD) and quality of life. GERD has a greater negative impact on the sense of well-being than many common chronic diseases including hypertension and angina. The only illness having a greater effect than GERD on quality of life is psychiatric disease. (Data from ref. [11].)

PATHOPHYSIOLOGY

The pathophysiology of GERD is complex and results from an imbalance between defensive factors protecting the esophagus (antireflux barriers, esophageal acid clearance, tissue resistance) and aggressive factors from the stomach contents (gastric acidity and volume and duodenal contents). The intermittent nature of symptoms and esophagitis in many patients suggests that the aggressive and defensive forces are part of a delicately balanced system.

Antireflux Barriers

The first tier of the three-tiered esophageal defense against acid damage consists of the *antireflux barriers.* This is an anatomically complex region that includes the intrinsic lower esophageal sphincter (LES), the diaphragmatic crura, the intra-abdominal location of the LES, the phrenoesophageal ligaments, and the acute angle of His (Fig. 60-2).

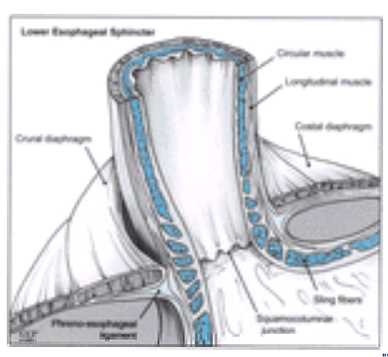

FIGURE 60-2. Anatomy of the gastroesophageal junction illustrating the major elements of the antireflux barrier. (From ref. [17].)

The LES is a tonically contracted segment of distal esophagus about 3 to 4 cm in length. [12] It is the major component of the antireflux barrier and is capable of preventing reflux even when it is completely displaced from the diaphragmatic crura because of a hiatus hernia. [12] The proximal margin of the LES is normally about 1.5 to 2.0 cm above the squamocolumnar junction, whereas the distal segment, about 2 cm in length, lies within the abdominal cavity. [13] This location of the distal LES contributes to the maintenance of gastroesophageal competence during intra-abdominal pressure events. Resting LES pressure ranges between 10 to 30 mm Hg and includes a generous reserve capacity, because a minimal basal LES pressure in the range of 5 to 10 mm Hg usually prevents GER. [14] The LES maintains a high-pressure zone by the intrinsic tone of its muscle and by cholinergic excitatory neurons. [15] There is considerable diurnal variation in basal LES pressure; it is lowest after meals and highest at night. It is also influenced by certain circulating peptides and hormones, foods (particularly fat), and numerous drugs (Table 60-1). During swallowing, LES relaxation (LESR) occurs for 5 to 10 seconds, thus permitting esophageal peristalsis to sweep the swallowed bolus into the stomach.

	INCREASE PRESSURE	DECREASE PRESSURE
Hormones	Gastrin Motilin Substance P	Secretin Cholecystokinin Somatostatin Vasoactive intestinal polypeptide
Neural agents	α-Adrenergic agonists β-Adrenergic antagonists Cholinergic agonists	α-Adrenergic antagonists β-Adrenergic agonists Cholinergic antagonists
Foods	Protein	Fat Chocolate Peppermint
Miscellaneous	Histamine Antacids Metoclopramide Domperidone Prostaglandin $F_{2\alpha}$ Cisapride	Theophylline Prostaglandins E_2 and I_2 Serotonin Meperidine Morphine Dopamine Calcium blockers Diazepam Barbiturates

TABLE 60-1 Substances that Modulate Lower Esophageal Sphincter Pressure

Anatomically, the LES lies within the hiatus created by the right crus of the diaphragm, and it is anchored by the phrenoesophageal ligaments, which inserts at about the level of the squamocolumnar junction (see Fig. 60-2). [16] Developmentally, the crural diaphragm arises from the dorsal mesentery of the esophagus and is innervated separately from the costal part of the diaphragm. It is inhibited by esophageal distention, during vomiting, and in association with transient LESRs, but not during swallowing. The crural diaphragm provides extrinsic squeeze to the intrinsic LES, contributing to resting pressure during inspiration and also augmenting LES pressure during periods of increased abdominal pressure such as coughing, sneezing, or bending. [17] Crural contractions impose rhythmic pressure increases of about 5 to 10 mm Hg on the LES pressure recording. [18] During deep inspirations and some periods of increased abdominal straining, these changes may reach 50 to 150 mm Hg. [19]

The oblique entrance of the esophagus into the stomach creates a sharp angle on the greater curve aspect of the gastroesophageal junction, the angle of His. This angle has been shown in cadavers to create a flap valve effect that contributes to gastroesophageal junction competency. [20]

Mechanisms of Reflux

Transient Lower Esophageal Sphincter Relaxations This is the most common mechanism underlying GER and also accounts for the reflux of gases during belching. Figure 60-3 illustrates an example of a transient LESR and highlights differences from swallow-induced LESRs: transient LESRs are not associated with antecedent pharyngeal contractions, are unaccompanied by esophageal peristalsis, persist for longer periods (>10 seconds) than swallow-induced LESRs, and are always accompanied by inhibition of the crural diaphragm. [21] Transient LESRs account for nearly all reflux episodes in healthy persons and for 50% to 80% in patients with GERD, depending on the severity of associated esophagitis (Fig. 60-4). [22] However, one study suggested that low basal LES pressure, rather than transient LESRs, may be the primary mechanism of GER in patients with nonreducible hernias. [23] Transient LESRs are not always associated with GER. In healthy persons, about 40% to 60% of transient LESRs are accompanied by reflux episodes, compared with 60% to 70% in patients with GERD. [15], [22], [24], [25] The rate of transient LESR is increased by gastric distention, whether by gas or a meal, by stress, and, to a lesser extent, by subthreshold (for swallowing) stimulation of the pharynx. [26] Under normal circumstances, meals are the major stimuli for transient LESRs, but the importance of specific foods is unknown. [27] Transient LESRs are inhibited by the supine position, sleep, general anesthesia, and vagal cooling. [26] Various drugs also impair transient LESRs including cholecystokinin A antagonists, anticholinergic drugs, morphine, somatostatin, nitric oxide inhibitors, 5-hydroxytryptamine $_3$ (5-HT $_3$) antagonists, and d-aminobutyric acid $_B$ (GABA $_B$) agonists. [28] Current evidence indicates that transient LESRs are mediated through vagal pathways. [26] Gastric distention activates mechanoreceptors in the proximal stomach adjacent to the gastric

cardia that send signals to the brainstem center through vagal afferent pathways. The structured sequence of motor events including LESR, inhibition of the crural diaphragm, and contractions of the esophageal body suggest that this process occurs in a programmed manner, probably controlled by a pattern generator within the vagal nuclei. The motor arm is in the vagus nerve and shares common elements with swallow-induced LESR. [30]

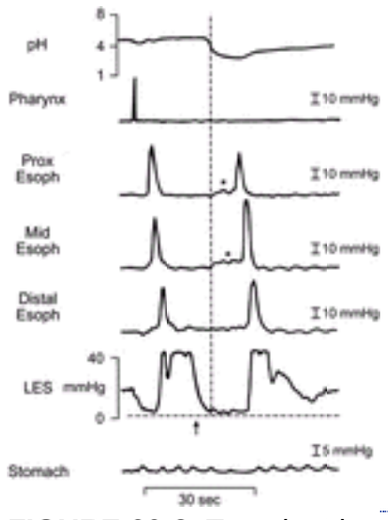

FIGURE 60-3. Transient lower esophageal sphincter (LES) relaxation (*arrow*) occurring immediately after the completion of a swallow-induced LES relaxation. Note the substantially longer duration of the transient LES relaxation (TLESR) compared with the preceding swallow-induced LES relaxation. A reflux episode (*dotted line*) associated with a common cavity (*asterisk*) occurs after complete LES relaxation has been achieved and is cleared by secondary peristalsis. (From ref. [26].)

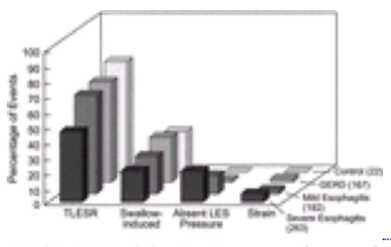

FIGURE 60-4. Proportion of reflux episodes in the spectrum of gastroesophageal reflux disease (*GERD*) occurring by the mechanisms of transient lower esophageal sphincter relaxation (*TLESR*), swallow-induced LES relaxation, absent basal LES pressure, and straining in the presence of low LES pressure (Data from ref. [22].)

Swallow-Induced Lower Esophageal Sphincter Relaxations About 5% to 10% of reflux episodes occur during swallow-induced LESRs. Most of these episodes are associated with defective or incomplete peristalsis. [22], [28] During a normal swallow-induced LESR associated with normal peristalsis, reflux is uncommon because of the absence of concomitant crural diaphragm relaxation, the relatively short duration of LES relaxation (5 to 10 seconds), and the prevention of reflux by the oncoming peristaltic wave (see Fig. 60-3). Reflux during swallow-induced LESR is more common in the presence of a hiatus hernia because of pooling of gastric liquids in the hernia sac and the absence of any residual diaphragmatic support during the LESR. [31]

Hypotensive Lower Esophageal Sphincter Pressure Stress reflux and free reflux are two mechanisms by which GER can be associated with diminished LES. [14], [22] *Stress reflux* results when a relatively hypotensive LES is overcome and is "blown open" by an abrupt increase in intra-abdominal pressure from coughing, straining, or bending. Stress reflux is unlikely when the LES pressure is greater than 10 mm Hg. *Free reflux* is characterized by a fall in intraesophageal pH without an identifiable change in intragastric pressure, and it usually occurs when the LES pressure is 0 to 4 mm Hg. Reflux as the result of low or absent LES pressure is uncommon. It is found mostly in patients with severe esophagitis, in whom it may account for up to 23% of reflux episodes (see Fig. 60-4), and rarely in patients without endoscopic evidence of esophagitis. [14], [22], [24], [25] The mechanisms of a low LES pressure are poorly understood. The presence of a hiatus hernia reduces LES pressure because the intrinsic support of the crural diaphragm is lost. [12] Some LES weakness may be secondary to impairment of the excitatory cholinergic pathways to the LES as a result of esophagitis. Induction of experimental esophagitis in cats affects the release of acetylcholine and lowers LES pressures; changes that are reversible on healing of the esophagitis. [32] However, healing of esophagitis in humans is rarely accompanied by an increase in LES pressure. [33]

Hiatus Hernia

P>The relationship between *hiatus hernia* and GERD remains controversial. Mainstream opinion has shifted widely from one that virtually equated hiatus hernia with GERD to one that denied it a causal role. Currently, both epidemiologic and physiological data confirm the importance of the hiatus hernia in patients with more severe esophagitis, peptic stricture, or Barrett esophagus. Hiatus hernias, identified endoscopically or radiologically, are reported in 54% to 94% of patients with reflux esophagitis; a rate strikingly higher than in the healthy population. [34]

The functional impact of the hiatus hernia has been clarified by elegant combined manometry and videofluoroscopic studies that show that hiatus hernia impairs LES function through several mechanisms as well as impairing esophageal clearance (Fig. 60-5). [31], [35], [36] Reflux is worse in patients who have a nonreducible as opposed to a reducible hiatus hernia. Nonreducing hernias are those in which the gastric rugal folds remain above the diaphragm between swallows. [36] Furthermore, statistical modeling has revealed a significant interaction between hiatus hernia and LES pressure, such that the likelihood of GER is increased as basal LES pressure decreases, an effect that is substantially amplified by the presence of a hiatus hernia and as hernia size increases. [12]

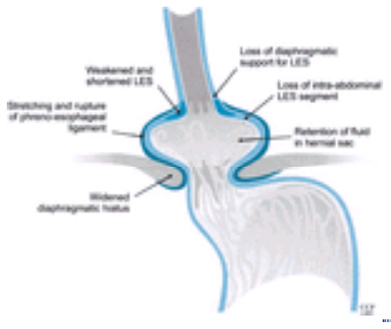

FIGURE 60-5. The impact of a hiatus hernia on the antireflux barrier. (From ref. [29].)

Displacement of the LES from the crural diaphragm into the chest reduces basal LES pressure and shortens the length of the high-pressure zone primarily because of the loss of the intra-abdominal LES segment. [35] Both these effects are caused by the loss of the extrinsic support of the diaphragmatic crura resulting in increased GER. [23] Hiatus hernia virtually eliminates the increase of LES pressure that occurs during straining and may increase the triggering of transient LESRs during gastric insufflation with gas. [35], [37] Large, nonreducible hernias also impair esophageal acid clearance because of increased tendency for reflux to occur from the hernia sac during swallow-induced LESRs. [31], [36]

The origin of hiatus hernia remains unclear. Familial clustering of GERD [10] suggests the possibilities of inherited muscle weakness in this area. Animal studies propose that reflux itself causes esophageal shortening promoting the development of a hiatus hernia. [38] Other studies find an association with obesity [39] and lifting of

heavy weights, [40] raising the possibilities that over time, chronic intra-abdominal stressors may weaken the esophageal hiatus and may cause the development of a hiatus hernia. This theory is attractive because it helps to reconcile the increased prevalence of hiatus hernias as the population ages. [34]

Esophageal Acid Clearance

The second tier against reflux damage is *esophageal acid clearance.* Reflux events determine the frequency and extent that gastric contents enter the esophagus, but esophageal acid clearance time determines the duration the mucosa is exposed to acid and probably the severity of acid damage. Esophageal acid clearance involves two related but separate processes: *volume clearance,* which is the actual removal of the reflux material from the esophagus, and *acid clearance,* which is the restoration of normal pH in the esophagus after acid exposure through titration with base, rather than true removal of the refluxed material.

Volume Clearance Esophageal peristalsis operates to clear the acid volume in both the upright and supine positions, but it is inoperative during deep rapid eye movement sleep. [45] Helm and colleagues [41] showed that one or two primary peristaltic contractions will completely clear a 15-mL fluid bolus from the esophagus. Primary peristalsis is elicited by swallowing, which occurs with a frequency of once per minute in awake subjects, regardless of whether reflux occurs. Secondary peristalsis, initiated by esophageal distention from acid reflux, is much less effective in promoting clearance of refluxate, thus offering only an ancillary protective role. Peristaltic dysfunction, that is, failed peristaltic contractions and hypotensive (<30 mm Hg) peristaltic contractions that incompletely empty the esophagus, increases in frequency with the severity of esophagitis. Kahrilas and colleagues [42] found that the prevalence of peristaltic dysfunction rose from 25% in patients with mild esophagitis to more than 50% in patients with severe esophagitis (Fig. 60-6). Failed peristalsis results in very poor volume clearance because the foci of feeble contractions clear most, but not all, of the refluxate from the esophagus. Whether esophagitis leads to peristaltic dysfunction or whether an underlying motility disorder predisposes to the development of reflux disease is not clear. Animal studies find that esophageal dysmotility associated with active esophagitis is reversible, but that associated with stricture or extensive fibrosis is not. [32] Clinical observations suggest that impaired motor function does not revert to normal after either effective medical or surgical therapies. [33]

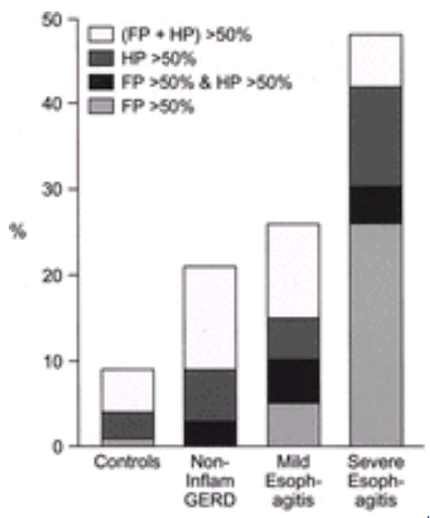

FIGURE 60-6. The prevalence of peristaltic dysfunction in controls and in patients with increasing severity of gastroesophageal reflux disease (GERD). The criteria for motility abnormalities was the occurrence of either failed, nontransmitted primary peristalsis (*FP*) or hypotensive peristalsis (<30 mm Hg) in the distal esophagus (*HP*) with more than half of the swallows. Note the increased prevalence of peristaltic dysfunction as the severity of GERD increases. (From ref. [42].)

Gravity contributes to bolus clearance when reflux occurs in the upright position. At night, when patients are supine, this mechanism is not operative unless the head of the bed is elevated. This time-honored treatment of GERD markedly improves acid clearance time and is most beneficial in those patients with aperistalsis (i.e., scleroderma). [43]

Salivary and Esophageal Gland Secretions *Saliva* is the second essential factor required for normal esophageal clearance of acid. Saliva has a pH of 6.4 to 7.8 and therefore is a weak base compared with the acidic gastric contents. [44] The high rate of spontaneous swallowing results in saliva production of approximately 0.5 mL per minute. Although saliva is ineffective in neutralizing large volumes of acid (5 to 10 mL), it can neutralize small residual amounts of acid remaining in the esophagus after the volume of refluxed material has been cleared by several peristaltic contractions. [41] The importance of swallowed saliva is supported by findings that increased salivation induced by oral lozenges or bethanechol is associated with a significant decrease in acid clearance time. In contrast, suction aspiration of saliva is accompanied by a marked prolongation of esophageal clearance, despite the presence of normal peristaltic contractions. [44] Physiological or pathological compromises of salivation may contribute to GERD. Diminished salivation during sleep explains why nocturnal reflux episodes are associated with markedly prolonged acid clearance times. [45] Similarly, chronic xerostomia is associated with prolonged esophageal acid exposure and esophagitis. [46] Cigarette smoking may promote GER. This was originally attributed to the effects of nicotine on lowering LES pressure, but more recent studies suggest that cigarette smokers have hyposalivation, which may also prolong esophageal acid clearance. [47] Finally, the esophagosalivary reflex may be impaired in patients with reflux esophagitis. This is a vasovagal reflex demonstrated by perfusing acid into the esophagus, thereby stimulating increased salivation. [48] This reflex may explain the symptoms of water brash (copious salivation) observed in some patients with reflux disease. The esophagosalivary reflex is very active in healthy persons, with a doubling or tripling of the salivary flow rate on exposure to acid. However, this reflex is diminished in patients with esophagitis and in those with strictures. [48] In addition to the role of saliva, dilution and neutralization of residual acid are achieved by the aqueous bicarbonate (HCO_3^-)-rich secretions of the esophageal submucosal glands. These glands have been identified in the opossum as well as in the human esophagus. [49] /SUP>Reflux of acid into the esophageal lumen stimulates these glands and helps to neutralize the acid, even if swallowing does not occur. [50]

Tissue Resistance Although clearance mechanisms minimize acid contact time with the epithelium, even healthy persons may have their esophagus exposed to acid 1 to 2 hours during the day and sometimes at night. Nevertheless, only a few persons experience symptomatic GER, and even fewer suffer GERD. This is the result of a third tier for esophageal defense, known as *tissue resistance.* Tissue resistance is not a single factor, but a group of dynamic mucosal structures and functions that interact to minimize mucosal damage from the noxious gastric refluxate. [51] Conceptually, tissue resistance can be subdivided into preepithelial, epithelial, and postepithelial factors. The preepithelial defense in the esophagus, in contrast to the stomach and duodenum, is poorly developed. There is neither a well-defined mucous layer nor a buffering capacity by the surface cells to secrete HCO_3^- into the unstirred water layer. This results in a lumen-to-surface pH gradient in the esophagus of 1:10, in contrast to the stomach and duodenum, where the gradient can range from 1:1000 to 1:10,000. [52] The epithelial defenses in the esophagus consist of both structural and functional components. Structural components include the cell membranes and intercellular junctional complexes of the esophageal mucosa. This structure is a 25- to 30-cell-thick, nonkeratinized squamous epithelium functionally divided into a proliferating basal cell layer (stratum basalis), a midzone layer of metabolically active squamous cells (stratum spinosum), and a 5- to 10-cell-thick layer of dead cells (stratum corneum). The esophageal mucosa is a relatively “tight” epithelium with resistance to ionic movement at the intercellular as well as the cellular level as the result of both tight junctions and the matrix of lipid-rich glucoconjugates in the intercellular space. [53] The functional components of tissue resistance include the ability of the esophageal epithelium to buffer and extrude hydrogen ions (H^+). Intracellular buffering is accomplished by negatively charged phosphates and proteins, as well as HCO_3^-. When the buffering capacity is exceeded and intracellular pH falls, it has the capacity actively to remove H^+ from the cells. This is possibly by the action of two transmembrane proteins, one a sodium (Na^+)/H^+ exchanger and the other a Na^+-dependent chloride (Cl^-)/HCO_3^- exchanger. [54], [55] After reflux-induced cell acidification, these transporters restore the intracellular pH to neutrality by exchanging H^+ for extracellular Na^+ or by exchanging Cl^- for extracellular HCO_3^-, respectively. Additionally, esophageal cells contain within their membrane a Na^+-independent Cl^-/HCO_3^- exchanger that extrudes HCO_3^- from the cytoplasm when the intracellular pH is too high. [54] When the epithelial cells are no longer able to maintain intracellular pH, they lose their ability to regulate volume, edema occurs, and balloon cells develop. The postepithelial defense is provided by the esophageal blood supply. Blood flow delivers oxygen, nutrients, and HCO_3^- and removes H^+ and carbon dioxide, thereby maintaining normal tissue acid-base balance. Blood flow to the esophageal mucosa increases in response to the stress of lumenal acid. [56] Cellular injury also stimulates cell proliferation, [57] which results in thickening of the basal cell layer of the epithelium. Unlike the stomach, in which superficial mucosal injury is repaired in hours, the esophagus repairs itself more slowly over days to weeks.

Gastric Factors

Gastric factors (particularly gastric volume and certain aggressive factors found in the refluxate) are potentially important in the production of reflux esophagitis. Gastric volume is determined by the

basal acid secretion rate, concomitant *H pylori* infection, duodenogastric reflux, and the rate of gastric emptying. Increased gastric volume not only provides more gastric contents available for reflux, but also increases the rate of transient LESRs.

Gastric Acid Secretion The primary importance of *gastric acid* is indisputable in the production of reflux esophagitis, but its mechanism may involve activation of pepsin rather than direct damage from acid alone (Fig. 60-7). In animal studies, acid causes minimal injury at a pH of less than 3.0, primarily by protein denaturation. However, the combination of acid and even small amounts of pepsin disrupts the mucosal barrier resulting in increased H^+ permeability, histological changes, and gross hemorrhage. [58] Complementing the animal studies, a series of clinical reports showed that patients with various grades of esophagitis, including Barrett esophagus, have increased frequency and duration of esophageal exposure to gastric refluxate of pH lower than 4. [59], [60] Conversely, perfusing the esophagus of animals with a pepsin solution of pH 7.5 produces minimal mucosal disruption or changes in permeability. [58] These observations are the cornerstone of acid suppressive therapy in the treatment of GERD.

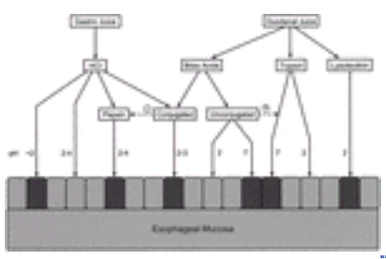

FIGURE 60-7. The postulated injurious agents responsible for esophageal mucosal damage. Mucosal injury is illustrated by the *darker boxes* representing the epithelial surface.

Some studies have suggested that patients with reflux, especially those responding poorly to conventional antisecretory therapy, may have higher rates of acid secretion than controls. [61] However, most evidence finds no abnormality of gastric acid secretion in patients with GERD. For example Hirschowitz [62] compared the gastric secretion of 115 patients with esophagitis with 508 age-, gender- and disease-matched control patients without esophagitis. Fasting basal or maximal secretion of either acid or pepsin was the same in both groups, and the severity of esophagitis was not related to any of these factors. Factors that reduce gastric acid secretion naturally, for example, concomitant infection with *H pylori,* especially if it is the cagA $^+$ virulent strain, may protect from the development of severe esophagitis and Barrett esophagus. [63], [64] *H pylori* infection, particularly infection with this virulent strain, is a biologic *antisecretory agent* that lowers gastric acidity. It produces severe corpus gastritis and accelerates the progression to multifocal atrophic gastritis and intestinal metaplasia, with concomitant lower acid output (Fig. 60-8). In addition, the bacteria produce ammonia that acts as a powerful neutralizing agent at elevated pH conditions. [8], [65] The corpus mucosa returns to normal when the *H pylori* infection is cured, increasing acid secretion and possibly contributing to the reports of esophagitis after successful treatment of *H pylori* infection. [66], [67] The consequences of long-term normalization of parietal cell function and return to higher intragastric acidity is unknown, but they could promote the development of more severe GERD, Barrett esophagus, and adenocarcinoma in Western populations. [8]

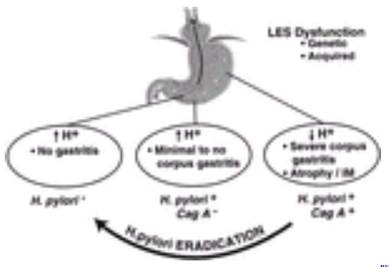

FIGURE 60-8. The acidity and volume of the gastric refluxate are modulated by the presence or absence of *Helicobacter pylori* and the cagA $^+$ virulence strain through the development of chronic corpus gastritis and gastric atrophy or intestinal metaplasia. Among persons susceptible to gastroesophageal reflux disease (GERD), the more acidic refluxate may increase the probability of esophagitis, Barrett esophagus, and adenocarcinoma, whereas the less acidic refluxate may protect against these complications of GERD. Conceivably, *H pylori* eradication could shift this delicate balance and could increase the potential for the development of more severe forms of GERD. (From ref. [8].)

Duodenogastric Reflux Bile acids have been implicated in the development of esophagitis, especially in the presence of increased *duodenogastric reflux.* Studies in animals demonstrate that conjugated bile acids produce their greatest injury in the presence of acid and pepsin, whereas trypsin, deconjugated bile salts, and the conjugated bile salt taurodeoxycholate are more damaging in the absence of acid [68] (see Fig. 60-7). Several surgical reports have suggested that duodenogastric reflux into the esophagus is frequent and may predispose to complications of GERD. [59], [69] However, accurate measurement of duodenogastric reflux is difficult. Duodenogastric reflux may be indirectly estimated by ambulatory pH studies using an esophageal pH of less than 7 to indicate *alkaline reflux.* [70] However, the reliability of this indirect marker is now questioned by newer techniques, which either spectrophotometrically measure bilirubin, the most common pigment in bile, [71] or measure esophageal impedance of the flow of liquids and gases independent of pH. [72] These studies show that acid reflux and bile reflux increase together across the spectrum of GERD, making it nearly impossible to incriminate one agent over the other in the development of esophagitis. [73] In addition, aggressive acid suppression with proton pump inhibitors (PPIs) decreases both acid and duodenogastric reflux probably by decreasing the volume of gastric contents available to reflux into the esophagus. [74] Finally, the absence of membrane microvesiculation and intracellular bile deposits in human esophageal biopsies, two distinctive morphologic features of experimental acid-bile salt injury, also argue against an important role for bile salts in GERD. [75]

Delayed Gastric Emptying The importance of *delayed gastric emptying* in the pathogenesis of GERD is controversial. Early studies observed a delay in the gastric emptying of solids in up to 50% of patients with reflux. [76] However, methodological problems including conducting the study with the patient supine, scanning only in the anterior position, and unstable radioisotopes may have influenced the results of these studies. More recent studies of patients in the upright position, in whom scanning was done both anteriorly and posteriorly, found only a 6% to 38% rate of delayed gastric emptying, regardless of the severity of the esophagitis. [77], [78] Nevertheless, delayed gastric emptying may be a major factor contributing to GERD in some groups, such as diabetic patients with autonomic peripheral neuropathy.

ASSOCIATED CONDITIONS

Certain medical and surgical conditions can predispose a person to GERD. The most common is *pregnancy;* 30% to 50% of pregnant women complain of heartburn, especially in the first trimester. Pregnancy increases the risk for reflux by the relaxing effects of circulating estrogens and progesterones on LES pressure. [79] Although symptoms may be severe, esophagitis is uncommon, and this type of “situational” GERD is cured with childbirth.

Up to 90% of patients with *scleroderma* have GERD as the result of smooth muscle fibrosis causing low LES pressure and weak or absent peristalsis. Severe disease is common; up to 70% of patients have esophagitis, many have peptic strictures, and Barrett esophagus and carcinoma of the esophagus have been reported. [80]

Unlike the previous two conditions that are characterized by LES dysfunction, hypersecretion of acid and increased gastric volume are the major factors causing GERD in patients with the *Zollinger-Ellison syndrome.* In these patients, the esophagitis and complications may be more difficult to treat than the ulcer disease. [81] After Heller myotomy, 10% to 20% of patients may develop GERD. [82] Finally, prolonged *nasogastric tube intubation* may contribute to the development of reflux esophagitis, in part because acid tracks orad along the tube and because the tube mechanically interferes with LES barrier function. [83]

CLINICAL MANIFESTATIONS

Classical Reflux Symptoms

Heartburn is the classical symptom of GERD, with patients generally reporting a burning feeling, rising from the stomach or lower chest and radiating toward the neck, throat, and occasionally the back. [84] Usually, it occurs postprandially, particularly after large meals or the consumption of spicy foods, citrus products, fats, chocolates, and alcohol. Recumbency and bending over may exacerbate heartburn. When heartburn dominates the patients' complaints, it has very high specificity (89%), but low sensitivity (38%) for GERD as diagnosed by abnormal 24-hour esophageal pH testing. [85] The diagnosis of GERD usually is based on the occurrence of heartburn on 2 or more days a week, although less frequent symptoms do not preclude the disease. [86] Although this symptom is an aid to diagnosis, the frequency and severity of heartburn do not predict the degree of esophageal damage. Heartburn is caused by acid stimulation of sensory nerve endings in the deeper layers of the esophageal epithelium. These nerve endings are normally protected by a relatively impermeable epithelium, but with epithelial changes caused by reflux, they may be stimulated by H^+ or spicy foods. [87]

Other common symptoms of GERD are acid regurgitation and dysphagia. The effortless regurgitation of acidic fluid, especially after meals and exacerbated by stooping or recumbency, is highly suggestive of GERD. [85] Among patients with daily regurgitation, the LES pressure usually is low, many have associated gastroparesis, and esophagitis is common. For these reasons, acid regurgitation may be more difficult to control medically then classical heartburn complaints. Dysphagia is reported by more than 30% of patients with GERD. [88] It usually occurs in the setting of long-standing heartburn, with slowly progressive dysphagia primarily for solids. Weight loss is uncommon because patients have good appetites. The most common causes are a peptic stricture or Schatzki ring, but other causes include severe esophageal inflammation alone, peristaltic dysfunction, and esophageal cancer arising from Barrett esophagus.

Less common reflux-associated symptoms include water brash, odynophagia, burping, hiccups, nausea, and vomiting. [89] *Water brash* is the sudden appearance in the mouth of a slightly sour or salty fluid. It is not regurgitated fluid, but rather secretions from the salivary glands in response to acid reflux. [44] *Odynophagia,* pain on swallowing, can occasionally be seen with severe ulcerative esophagitis. However, its presence should raise the suspicion of an alternative cause of esophagitis, especially infections (candidiasis, herpes) or pills (tetracycline, potassium chloride, quinine, vitamin C, alendronate).

In contrast to the previously described symptomatic presentations, some patients with GERD are asymptomatic. This is particularly true in elderly patients because of decreased acidity of the reflux material or decreased pain perception. [5] Many elderly patients present first with complications of GERD because of long-standing disease with minimal symptoms. For example, up to one third of patients with Barrett esophagus are insensitive to acid at the time of presentation. [90]

Extraesophageal Manifestations

It has been suggested that GER may be the cause of a wide spectrum of conditions including noncardiac chest pain, asthma, posterior laryngitis, chronic cough, recurrent pneumonitis, and even dental erosion. [91] Some of these patients have classical reflux symptoms, but many are "silent refluxers," contributing to problems in making the diagnosis. Furthermore, it may be difficult to establish a causal relationship even if GER can be documented by testing (e.g., pH studies), because patients may simply have two common diseases without a cause-and-effect relationship.

Chest Pain GER-related *chest pain* may mimic angina pectoris. The chest pain is usually described as squeezing or burning, substernal in location, and radiating to the back, neck, jaw, or arm. It often is worse after meals, awakens the patient from sleep, and may worsen during periods of emotional stress. Heavy exercise, even treadmill testing, may provoke GER. [92] Reflux-related chest pain may last minutes to hours, often resolves spontaneously, and may be eased with antacids. Most patients with GERD-induced chest pain have heartburn symptoms. [93] Early studies suggested that spastic motility disorders were the most common esophageal cause of chest pain. However, more recent studies using ambulatory esophageal pH and pressure monitoring suggest that about 25% to 50% of patients with noncardiac chest pain have GERD. [94] Overall, these series of reports found that 41% of patients had abnormal 24-hour pH test results, whereas 32% had chest pain that was clearly associated with acid reflux. Patients with coronary artery disease commonly have coexisting esophageal diseases, but the evidence that GER causes ischemic pain is controversial. [95] The mechanism for GERD-related chest pain is not clearly understood and probably is multifactorial, related to H^+ ion concentration, volume and duration of acid reflux, and secondary esophageal spasm.

Asthma and Other Pulmonary Diseases The association of GERD and *pulmonary diseases* was recognized by Sir William Osler, [96] who recommended that asthmatic patients should "learn to take their large daily meal at noon to avoid nighttime asthma which occurred if they ate a full supper." More recent studies suggest the coexistence of the two diseases in up to 80% of asthmatic patients, irrespective of the use of bronchodilators. [97], [98] GERD should be considered in asthmatic patients who present in adulthood, those without an intrinsic component, and those not responding to bronchodilators or steroids. [99] Up to 30% of patients with GERD-related asthma have no other esophageal complaints. Other pulmonary diseases associated with GERD include aspiration pneumonia, interstitial pulmonary fibrosis, chronic bronchitis, bronchiectasis, and possibly cystic fibrosis, neonatal bronchopulmonary dysplasia, and sudden infant death syndrome. Proposed mechanisms of reflux-induced asthma are either aspiration of gastric contents into the lungs with secondary bronchospasm or activation of a vagal reflex from the esophagus to the lungs causing bronchoconstriction. Animal [100] and human [101] studies report bronchoconstriction after esophageal acidification, but the response tends to be mild and unpredictable. In contrast, intratracheal infusion of even small amounts of acid induces profound and reproducible bronchospasm in cats. [100] The reflux of acid into the trachea as compared with the esophagus alone predictably caused marked changes in peak expiratory flow rates in asthmatic patients. [102] Although either mechanism may be responsible for reflux-induced asthma, most patients probably suffer from intermittent microaspiration.

Ear, Nose, and Throat Diseases GERD may be associated with a variety of laryngeal conditions and symptoms, of which *reflux laryngitis* is perhaps the most common. [103], [104] These patients present with hoarseness, globus sensation, frequent throat clearing, recurrent sore throat, and prolonged voice warmup. Ear, nose, and throat signs attributed to GERD include posterior laryngitis with edema and redness, vocal cord ulcers and granulomas, leukoplakia, and even carcinoma. These changes usually are limited to the posterior third of the vocal cords and interarytenoid areas, both in close proximity to the upper esophageal sphincter. GERD is the third leading cause of chronic cough (after sinus problems and asthma), accounting for 20% of cases. [105] *Dental erosion,* defined as the loss of tooth structure by chemical processes not involving bacteria, can be caused by GER in healthy persons and in patients with bulimia. [106] Despite the association between ear, nose, and throat diseases and GERD, overt esophagitis usually is absent, and most patients have only mild reflux symptoms, if any. [103] Microaspiration of gastric contents is the most likely cause of these complaints. Animal studies find that the combination of acid and pepsin is very injurious to the larynx. [103] Human studies report that proximal esophageal acid exposure, especially at night while sleeping, is significantly increased in patients with laryngeal symptoms and signs. [107]

DIAGNOSTIC EVALUATION

Many tests are available for evaluating patients with suspected GERD. These tests are often unnecessary because the classical symptoms of heartburn and acid regurgitation are sufficiently specific to identify reflux disease and to begin medical treatment. However, this may not always be the case, and the clinician must decide which test to choose to arrive at a diagnosis in a reliable, timely, and cost-effective manner, depending on the information desired (Table 60-2).

Tests for Reflux
Intraesophageal pH monitoring
Ambulatory bilirubin monitoring
Radionuclide ^{99m}Tc scintiscanning
Barium esophagram
Tests to Assess Symptoms
Empiric trial of acid suppression
Intraesophageal pH monitoring
Acid perfusion (Bernstein) test
Tests to Assess *Esophageal* Damage
Endoscopy
Esophageal biopsy
Barium esophagram
Tests to Assess *Pathogenesis*
Esophageal manometry
Gastric analysis
Radionuclide ^{99m}Tc scintiscanning

*Order of presentation represents the order of diagnostic usefulness.

TABLE 60-2 Commonly Used Tests for Assessing the Presence, Mechanism, and Consequences of Gastroesophageal Reflux Disease*

Empiric Trial of Acid Suppression

The simplest and most definitive method for diagnosing GERD and assessing its relation to symptoms (either classical or atypical) is the empiric trial of acid suppression. Unlike other tests that only suggest an association (e.g., esophagitis at endoscopy or positive symptom index on pH testing), the response to antireflux therapy ensures a cause-and-effect relationship between GERD and symptoms. Therefore, it has become the "first" test used in patients with classical or atypical reflux symptoms without "alarm" complaints. The popularity of this approach was aided by the introduction

of the PPIs, which, unlike the histamine $_2$ receptor antagonists (H $_2$RAs), could drastically reduce the amount of acid reflux into the esophagus. Symptoms usually respond to a PPI trial in 7 to 14 days. If symptoms disappear with therapy and then return when the medication is stopped, GERD may be assumed.

In the reported empiric trials with heartburn, the initial dose of PPI was high (e.g., omeprazole 40 to 80 mg/d) and was given for not less than 14 days. A positive response is defined as at least 50% improvement in heartburn. Using this approach, the PPI empiric trial had a sensitivity of 68% to 83% for determining the presence of GERD. [108, 109] In noncardiac chest pain, Fass and colleagues [110] found that a 7-day trial of omeprazole, 40 mg in the morning and 20 mg at night, had a sensitivity of 78% and specificity of 86% for predicting GERD, when compared with traditional tests. Likewise, Ours and colleagues [111] found omeprazole, 40 mg twice a day for 2 weeks, to be a very reliable method for identifying acid-related cough. Empiric trials using a 2- to 4-month regimen of PPIs taken twice a day also are commonly used in patients with suspected GERD-associated asthma and GERD complaints related to the ear, nose and throat.

An empiric trial of PPIs for diagnosing GERD has many advantages. The test is office based, is easily performed, is relatively inexpensive, is available to all physicians, and avoids many needless procedures. For example, Fass and associates [110] showed a savings of more than $570 per average patient because of a 59% reduction in the number of diagnostic tests performed for noncardiac chest pain. Disadvantages are few, including a placebo response and uncertain symptomatic end point if symptoms do not resolve totally with extended treatment.

Endoscopy

Upper *endoscopy* is the current standard for documenting the type and extent of mucosal injury to the esophagus (see Chapter 138). It identifies the presence of esophagitis and excludes other causes of the patient's complaints. However, only 40% to 60% of patients with abnormal esophageal reflux by pH testing have endoscopic evidence of esophagitis. Thus, the sensitivity of endoscopy for GERD is 60% at best, but it has excellent specificity, at 90% to 95%. [112]

The earliest endoscopic signs of acid reflux include edema and erythema. Neither finding is specific for GERD, and both are very dependent on the quality of endoscopic visual images. [112] More reliable are the findings of friability, granularity, and red streaks. Friability (easy bleeding), occurring with gentle pressure on the mucosa, results from the development of enlarged capillaries near the mucosal surface in response to acid. Red streaks may extend upward from the esophagogastric junction along the ridges of the esophageal folds. In studies evaluating these stigmata, nearly all patients had GERD. [113] With progressive acid injury, erosions develop (Fig. 60-9A, see also Color Fig. 60-9A). These are characterized by shallow thinning of the mucosa associated with a white or yellow exudate surrounded by erythema. Commonly located just above the esophagogastric junction, erosions may be either single lesions or coalesced regions. Typically, they occur along the tops of mucosal folds, areas most prone to acid exposure. Erosions may also be caused by nonsteroidal antiinflammatory drug use, heavy smoking, and infectious esophagitis. [113] Ulcers reflect more severe esophageal damage. They penetrate the mucosa, tend to have either a white or yellow discolored base, and may be seen either isolated along a fold or surrounding the esophagogastric junction.

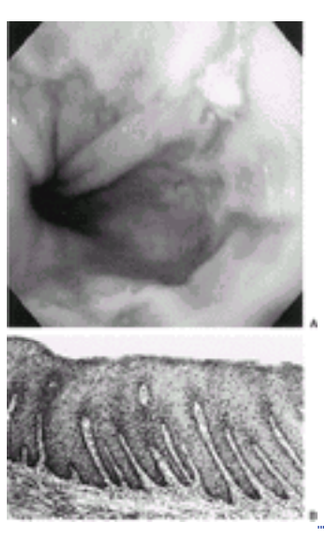

FIGURE 60-9. (See Color Fig. 60-9A.) Endoscopic and histological signs of esophagitis. (**A**) Two linear erosions extending proximally from the squamocolumnar junction at the proximal border of a hiatal hernia. This would be classified as grade II esophagitis by the Savary-Miller and Hetzel systems and as grade B by the Los Angeles scale. (**B**) Reparative changes secondary to reflux disease characterized by basal cell hyperplasia (*dark cells* crowding the papillae) and marked elongation of the rete pegs (*pale papillae* with fingerlike projections).

Endoscopic grading of GERD depends on the endoscopist's interpretation of these visual images. Unfortunately, there exists no standard classification scheme for endoscopic findings. Instead, several grading systems are available, but none are completely satisfactory (Table 60-3). In Europe, the most popular scheme is the *Savary-Miller classification,* [114] which is based on degree of mucosal erosions. In the United States, the Hetzel and Los Angeles systems are most popular. The *Hetzel system* grades severity not by the number of erosions but by the area of mucosal injury. [115] In the *Los Angeles system,* the number, length, and location of mucosal breaks determine the degree of esophagitis. [116] These different classification systems diverge the most when defining the subtlest degree of injury. When erythema, edema, and an indistinct Z-line are included, the sensitivity of diagnosing GERD rises at the expense of specificity.

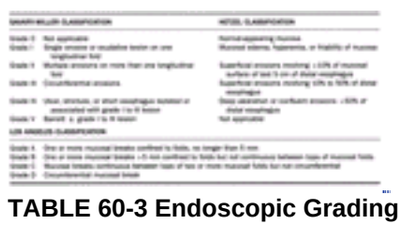

TABLE 60-3 Endoscopic Grading Systems for Esophagitis

Most patients with GERD are treated initially without endoscopy. The important exception is the patient experiencing alarm symptoms: dysphagia, odynophagia, weight loss, and gastrointestinal bleeding. With such symptoms, endoscopy should be performed early to rule out other entities such as infections, ulcers, cancer, or varices.

The role of endoscopy in GERD in the absence of alarm symptoms is more controversial and is evolving in the era of PPI therapy. Initially, endoscopy was used to place patients into two groups—those with nonerosive or mild disease and those with severe erosive disease—and to direct their treatment more precisely. However, this practice is now less popular with the use of PPIs as the first line of therapy for GERD. Because these drugs treat both groups equally well, early endoscopy has less impact on the choice of therapy. Currently, the most important reason for performing endoscopy in patients with GERD is to identify peptic strictures or Barrett esophagus. Using this rationale, most patients with chronic GERD need only one endoscopic examination while they are receiving therapy.

Esophageal Biopsy

The ability to obtain tissue during endoscopy is very important. *Biopsies* of the esophagus help to identify reflux injury, exclude other esophageal diseases, and confirm the presence of complications, especially Barrett esophagus (see Chapter 142). Microscopic changes indicative of reflux may occur even when the mucosa appears normal endoscopically. [117] In patients with classical esophagitis, biopsies are usually not taken unless they are needed to exclude other diagnoses such as neoplasm, infection, pill injury, or bullous disease. When Barrett esophagus is suspected, biopsies are

mandatory and are best done when esophagitis is healed.

The most sensitive histological markers of GERD are reactive epithelial changes characterized by an increase in the basal cell layer greater than 15% of the epithelium thickness or papilla elongation into the upper third of the epithelium (see Fig. 60-9B). These changes represent increased epithelial turnover of the squamous mucosa. Papilla, or rete peg, height increases as a result of loss of surface cells from acid injury, whereas basal cell hyperplasia is indicative of mucosal repair. Unfortunately, these changes are also noted in up to 50% of healthy persons when biopsies are taken from the distal 2 to 3 cm of the esophagus. [118] Hence, the changes are sensitive markers for GERD but have poor specificity.

Acute inflammation characterized by the presence of neutrophils and eosinophils is very specific for esophagitis. [118] Acid reflux injury to the vascular bed of the esophagus releases vasoactive substances that promote edema and migration of neutrophils and eosinophils into the area. Neutrophils are specific for acute esophagitis but are an insensitive marker, being present in only 15% to 40% of patients with GERD. [119] Eosinophils are found more often on biopsy (19% to 63% of subjects) but are less specific, present in up to 33% of healthy adults. [120] Interestingly, the sensitivity and specificity of eosinophils in children are much stronger, reflecting the lack of eosinophils in the juvenile inflammatory response. [118, 121]

Esophageal pH Monitoring

Ambulatory intraesophageal pH monitoring is now the standard for establishing pathological reflux (see Chapter 150). [122, 123] The test is performed with a pH probe passed nasally and positioned 5 cm above the manometrically determined LES. The probe is connected to a battery-powered data logger capable of collecting pH values every 4 to 6 seconds. An event marker is activated by the subject in response to symptoms, meals, and body position changes. Patients are encouraged to eat normally and to pursue regular daily activities. Monitoring is carried out usually for 18 to 24 hours. Reflux episodes are detected by a drop in pH to less than 4. Commonly measured parameters include the percentage of total time that the pH is less than 4, the percentage of time upright and supine that the pH is less than 4, the total number of reflux episodes, the duration of longest reflux episode, and the number of episodes longer than 5 minutes. [122] The total percentage of time that the pH is less than 4 is the most reproducible measurement for GERD, with reported upper limits of normal values ranging from 4% to 5.5%. [123] Ambulatory pH testing can discern positional variations in GER, meals, and sleep-related episodes and helps to relate symptoms to reflux events (Fig. 60-10). As the result of its reliability for measuring GER across normal activities, ambulatory pH testing has replaced other older studies, such as the standard acid reflux (Tuttle) test and radionuclide scintigraphy.

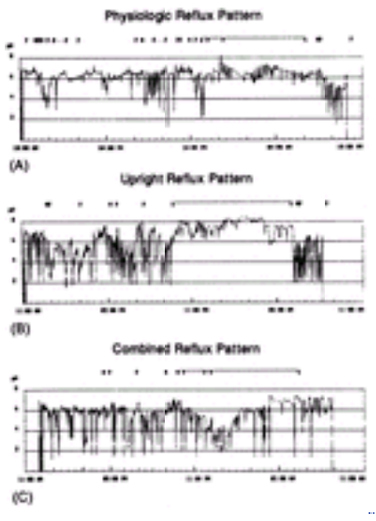

FIGURE 60-10. Common patterns of 24-hour esophageal pH monitoring. **(Top)** The physiological pattern of gastroesophageal reflux (*GER*) in healthy persons. Reflux is noted after meals (*m*) but not while asleep (*s*). A reflux episode is defined as a pH drop to less than 4. **(Middle)** Upright reflux pattern with extensive GER during day but not at night. These patients have frequent symptoms, but esophagitis is uncommon. **(Bottom)** Combined pattern with GER during day and at night (one episode lasted 3 hours). Most of these patients have esophagitis.

One important problem with esophageal pH monitoring is that there exists no absolute threshold value that reliably identifies pathological GER. Validation studies comparing the presence of esophagitis with abnormal pH test report sensitivities ranging from 77% to 100% with specificities from 85% to 100%. [123] However, these patients rarely need pH testing; rather, the patients with normal endoscopic findings and suspected reflux symptoms should benefit most from ambulatory pH monitoring. Unfortunately, the data are much less conclusive in this group, with considerable overlap between controls and patients with nonerosive reflux. [123] Other drawbacks of pH testing include possible equipment failure, the pH probe's missing a reflux event because it is buried in a mucosal fold, and false-negative studies resulting from dietary or activity limitations from poor tolerability of the nasal probe.

An important advantage of ambulatory esophageal pH monitoring is its ability to record and correlate symptoms with reflux episodes over extended periods. For this indication, it has essentially replaced the shorter acid perfusion (Bernstein) test. Because only about 10% to 20% of reflux episodes are associated with reported symptoms, different statistical analyses have evolved attempting to define a significant association between these two variables including the symptom index, symptom sensitivity index, and symptom association probability. [124] Unfortunately, no studies to date have defined the accuracy of any of these symptom scores in predicting response to therapy. Therefore, pH testing and symptom correlation can define an association between complaints and GER, but only treatment trials address the true definition of a causal relationship.

Definite clinical indications for ambulatory pH monitoring have been established. [123] *Before fundoplication,* pH testing should be performed in patients with normal endoscopic findings to identify the presence of pathological reflux. If esophagitis is present, pH testing is not necessary because the disease has been established. *After antireflux surgery, persistent or recurrent symptoms* warrant repeat pH testing. In these situations, pH monitoring is performed with the patient discontinuing all antireflux medications (PPIs for 1 week, H_2RAs for 2 days). Esophageal pH testing is particularly helpful in the evaluation of patients with *reflux symptoms resistant to treatment with normal or equivocal endoscopic findings*. For this indication, pH testing is usually done in patients receiving therapy to define two populations: those with and those without continued abnormal esophageal acid exposure times. The group with persistent GER needs intensification of the medical regimen, whereas those patients with symptoms and adequate acid control have another cause of their complaints. Finally, ambulatory pH testing may help in *defining patients with extraesophageal manifestations of GERD*. In this situation, pH testing is usually done with additional pH probes placed in the proximal esophagus or pharynx. [125] Initially, most of these studies were done when patients were not taking antireflux medications, to confirm the coexistence of GERD; however, this does not guarantee symptom causality. Therefore, the current approach is to treat the patients aggressively with PPIs first and to reserve pH testing only for those patients not responding after 4 to 12 weeks of therapy. [126]

Barium Esophagram

The *barium esophagram* is an inexpensive, readily available, and noninvasive esophageal test (see Chapter 152). It is most useful in demonstrating structural narrowing of the esophagus and in assessing the presence and reducibility of a hiatal hernia. Subtle findings such as Schatzki rings, webs, or minimally narrowed peptic strictures are often seen only with an esophagram; they are missed by endoscopy, which may not adequately distend the esophagus. This test, which involves consuming a 13-mm radiopaque pill or marshmallow along with the barium liquid, is the most sensitive for detecting esophageal narrowing, with values reported between 95% and 100%. [127] By giving the patient in the prone oblique position swallows of barium, the barium esophagram also allows good assessment of peristalsis and is helpful preoperatively in identifying a weak esophageal pump. [128]

The ability of the barium esophagram to detect esophagitis varies considerably. Although sensitivities of 79% to 100% have been reported with moderate to severe esophagitis, mild esophagitis is usually missed. [128] Barium testing also falls short when addressing the presence of Barrett esophagus. Barium studies can identify GER when contrast moves in a retrograde fashion from the stomach into the esophagus. If this occurs spontaneously, repeatedly, or to a significant degree into the middle or proximal esophagus, the test is positive, but it has a sensitivity of only about 40% for defining GERD. [129] Provocative maneuvers such as leg lifting, coughing, the Valsalva maneuver, or the water-siphon test can be used to elicit stress reflux. Although these tests can improve the sensitivity of

the barium esophagram, some argue that they also decrease its specificity. [129], [130]

The barium esophagram is primarily used in evaluating the patient with GERD with new-onset dysphagia because it can define subtle strictures and rings as well as assess motility. Conversely, endoscopy is preferred in the patient with recurrent dysphagia known to have a stricture or for the assessment of esophagitis or Barrett esophagus.

Esophageal Manometry

Esophageal manometry allows accurate assessment of LES pressure and relaxation, as well as peristaltic activity including contraction amplitude, duration, and velocity (see Chapter 150). However, esophageal manometry is generally not indicated in the evaluation of the patient with uncomplicated GERD because most of these patients have a normal resting LES pressure. [42] It is an integral component of pH testing to define the LES location accurately, a task poorly performed by endoscopy, fluoroscopy, or the pH pull-through technique. Esophageal manometry is an essential test in the preoperative evaluation of patients for antireflux surgery. [131] A normal LES pressure does not preclude surgery for the reasons discussed, yet occasionally an alternative diagnosis such as achalasia or scleroderma is made, which may change the clinical approach. Most importantly, the presence of ineffective peristalsis characterized by either low-amplitude (<30 mm Hg) peristaltic contractions or frequent failed peristalsis [132] suggests a weak esophageal pump. In these patients, a loose 360° fundoplication or an incomplete fundoplication will minimize the risk of postoperative dysphagia.

Miscellaneous Tests

Radiolabeled technetium-99m sulfur colloid scintiscanning is useful as a semiquantitative test for detecting GER. [133] After instilling 300 mL of radioisotope in saline through a nasogastric tube into the stomach, gamma counts over the esophagus are obtained in the supine position before and after provocation with abdominal compression. Although test specificity approaches 90%, the sensitivity is quite variable, from 14% to 90%. [134]

The *acid perfusion (Bernstein) test* is useful for detecting the relationship of symptoms to esophageal acidification. [135] The study is done with the patient upright with a nasogastric tube positioned in the midesophagus. Initially, normal saline is infused at 120 drops/min for 5 to 15 minutes, followed by an infusion of 0.1 N hydrochloric acid. If symptoms develop with acid infusion, saline is reinfused to assess symptom relief. Symptoms during acid infusion, but not saline infusion, constitute a positive test. The sensitivity of the Bernstein test for GERD ranges from 32% to 100%, and its specificity ranges from 40% to 100%. [136] In clinical practice, 24-hour esophageal pH testing has generally replaced both these tests.

Bile reflux can be measured using *ambulatory esophageal bilirubin monitoring* (Bilitec: Medtronics, Minneapolis, MN), [71] which uses the spectrophotometric property of bilirubin, the most common pigment in bile. As in pH testing, a fiberoptic light source is introduced into the esophagus with a data collection system worn on a waist belt. A spectrophotometer measures the wavelength absorption at 450 nm (bilirubin) and at 565 nm (reference) every 8 seconds. An integrated microcomputer calculates the difference of the absorbances, which is directly proportional to the bilirubin concentration in the sample. [71], [73] This allows a pH-independent assessment of duodenogastroesophageal reflux, which is preferable to the older method employing an esophageal pH of more than 7. [70] In the future, ambulatory measurements of esophageal impedance, [72] which measures the electrical activity of liquid and gas moving up and down the esophagus, combined with pH monitoring may be the preferred technique for measuring nonacidic reflux.

DIFFERENTIAL DIAGNOSIS

Symptoms associated with GERD may be mimicked by other esophageal and extraesophageal diseases including achalasia, Zenker diverticulum, gastroparesis, gallstones, peptic ulcer disease, functional dyspepsia, and angina pectoris. These disorders usually can be identified by failure to respond to aggressive antisecretory therapy and by diagnostic tests such as endoscopy, barium esophagram, esophageal manometry, ultrasound, nuclear emptying studies, and various cardiac tests. Although GERD is the most common cause of esophagitis, other causes (esophagitis, infections, or radiation esophagitis) need to be considered in cases that are difficult to manage cases and in older or immunocompromised patients.

CLINICAL COURSE

The clinical course of reflux esophagitis depends to a great extent on whether the patient has erosive or nonerosive GERD on initial presentation. Furthermore, patients tend not to cross over from one group to another unless they are treated medically or surgically: in follow-up ranging from 6 months to more than 5 years, only 15% of patients with nonerosive disease evolved over time to having esophagitis or complications of GERD. [137], [138]

Nonerosive Reflux Disease

Although early studies from tertiary referral centers suggested that nearly half of patients with GERD had esophagitis, [139] studies carried out in community practices reveal that up to 70% of the patients with GERD had a normal endoscopic examination. [140], [141] Furthermore, another community-based study of antacid users found that 53% of patients with GERD had nonerosive disease, and two thirds of the remaining had only minimal erosive changes at endoscopy. [142] Endoscopy-negative patients with GERD are more likely to be female, younger, thin, and without hiatal hernia. Despite their mild mucosal damage, these patients demonstrate a chronic pattern of symptoms with periods of exacerbation and remission. [143]

Nonerosive GERD is suspected by the presence of typical reflux symptoms with a normal endoscopic examination and is confirmed by the patient's response to antisecretory therapy. When performed, 24-hour esophageal pH monitoring identifies three distinct subset of patients with nonerosive disease. First, there are the patients with abnormal acid exposure time who are usually responsive to antisecretory therapy. Second are the patients with normal reflux parameters but a good relationship between acid reflux episodes and symptoms. This group represents 30% to 50% of patients with nonerosive GERD [179] and has "functional heartburn." [143] These patients probably have heightened esophageal sensitivity to acid and are less likely to respond to antireflux therapy. [144] The third group is characterized by normal acid exposure times and poor symptom correlation. Despite sometimes having classical reflux symptoms, other diseases such as achalasia, gastroparesis, bile reflux, or functional dyspepsia are the cause of their symptoms. Overall, patients with nonerosive GERD do not respond to antireflux treatments as well as do patients with erosive GERD, probably because these three subsets are not carefully defined before treatment. [143]

Erosive Reflux Disease

The clinical course of patients with *erosive esophagitis* is more predictable and is associated with complications of GERD. Controlled studies have shown that in the absence of ongoing maintenance therapy, up to 85% of patients with erosive GERD will have a relapse within 6 months, and the relapse rate is highest in those with the more severe grades of esophagitis. [145], [146] This observation, however, should not prevent at least one attempt to withdraw medication, because 20% of patients remain in remission for up to 1 year, especially those with milder esophagitis grades. Although the natural history of untreated erosive GERD is well studied, two European studies suggest that these patients are more prone to reflux complications. In a Finnish study, 20 patients with erosive GERD treated with lifestyle changes, antacids, and prokinetic drugs were followed up for a median of 19 years. Fourteen patients continued to have erosions, and 6 new cases of Barrett esophagus were detected. [147] Likewise, a large retrospective European study with 6.5 years of follow-up found a high rate of complications (21%) including 13 esophageal ulcers, 15 with strictures, and 45 patients with Barrett epithelium. [148] However, these data must be contrasted with other studies in which no patients with erosive esophagitis developed Barrett esophagus in a 2-year trial in the United States, [149] and over a 12-year period, in 3800 French patients, development of stricture was reported in only 0.26%. [150]

COMPLICATIONS

Hemorrhage and Perforation

Hemorrhage and esophageal perforation are rare complications of reflux esophagitis and are usually associated with deep esophageal ulcers or severe diffuse esophagitis. Clinically important hemorrhage has been reported in 7% to 18% of patients with GERD. [151] Esophageal perforations are very rare in the PPI era, but they can result in mediastinitis and can be fatal if they are not rapidly recognized and treated.

Peptic Esophageal Strictures

Strictures occur in 7% to 23% of patients with untreated reflux esophagitis, especially in older men. [152] They usually evolve over many years and may be linked to the long-term use of nonsteroidal

antiinflammatory drugs. [153] The mechanism of stricture formation is complex, starting as a reversible inflammatory process with edema, cellular infiltration, and vascular congestion, progressing to deposition of connective tissue and collagen, and ending in irreversible fibrosis. With the onset of dysphagia, there is often less heartburn, reflecting the stricture's acting as a barrier to reflux. Dysphagia is usually limit to solids, but it may progress to liquids. Unlike malignant strictures, patients with peptic strictures have a good appetite, alter their diet, and lose little weight.

Radiographically, peptic strictures are smooth-walled, tapered, circumferential narrowings in the lower esophagus, which are usually less than 1 cm long, but occasionally they extend to 8 cm in length (Fig. 60-11). In these unusual cases, the clinician should suspect a predisposing condition, such as the Zollinger-Ellison syndrome, superimposed pill esophagitis, or prolonged nasogastric intubation. [152] A stricture in the middle to upper esophagus should raise the suspicion of Barrett esophagus or malignant disease. Although once controversial, most data today suggest that a Schatzki ring is a forme fruste of an early peptic stricture. [154] In all cases, the nature of a peptic stricture needs to be confirmed by endoscopy with biopsies because some patients may have Barrett esophagus or unsuspected cancer.

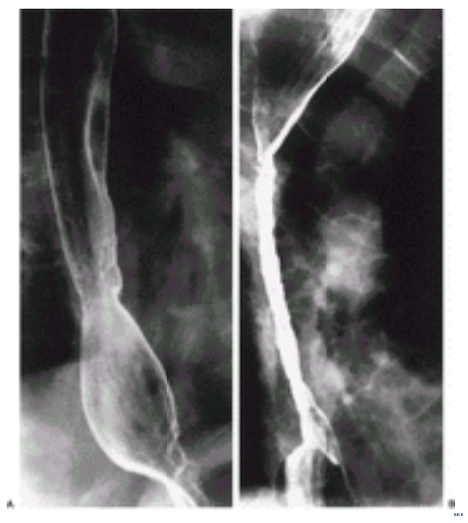

FIGURE 60-11. Barium esophagram of several forms of esophageal strictures. (**A**) Double-contrast study showing distal linear erosive esophagitis, an ulcer with a short stricture, and a large hiatus hernia. (**B**) A long, irregular distal stricture in a patient with Zollinger-Ellison syndrome. The stricture occurred after prolonged nasogastric intubation following head and neck surgery.

Barrett Esophagus

In some patients with GERD, the squamous epithelium of the distal esophagus is replaced by specialized columnar epithelium, resembling that of the intestine and containing goblet cells. Although Dr. Norman Barrett thought this lesion was a congenitally shortened esophagus, [155] studies consistently show that these patients have severe GERD with low LES pressures, poor esophageal motility, large hiatal hernias, and extensive acid and bile reflux. [156] Furthermore, most patients have had chronic reflux symptoms for at least 10 years. [157] Animal experiment show that, if the mucosal lining of the distal esophagus is excised in the setting of free acid reflux, columnar epithelium will regenerate in the area previously occupied by squamous epithelium. [158] If reflux is controlled, the mucosal lining will regenerate with squamous epithelium. Pluripotential stem cells derived from the stratified squamous epithelium are the origin of the specialized columnar epithelium. [159]

Barrett esophagus was once considered an uncommon condition, but estimates of its frequency at autopsy (1 in 57 to 1 in 105 cases), on general endoscopy survey (1 in 100 cases) and on endoscopic surveys of patients with GERD (10 in 100 to 15 in 100 cases), indicate that it is not uncommon, and it affects nearly 700,000 adults in the United States. [155] An autopsy series from Olmsted County, Minnesota found that most cases of Barrett esophagus go undetected during life and thus are not accessible for cancer surveillance programs. [160] Barrett esophagus is principally a disorder of white men; it is three times more frequent in men than in women and is rare in African Americans and Asians. [161] It is found predominantly in middle-aged and older adults; the mean age at diagnosis is approximately 55 years, but it has been reported in children older than 5 years of age. [162] The prevalence of Barrett esophagus increases with age, paralleling that of reflux esophagitis, but the length of the columnar-lined segment remains remarkably stable, even over years of endoscopic follow-up. [161] This finding suggests that it arises rapidly in the susceptible reflux damaged esophagus and early in the course of disease. Families have been reported with multiple members having Barrett esophagus, some with cancer affecting more than one generation. [10], [163] Although the columnar-lined esophagus in itself does not cause symptoms, most patients complain of heartburn and regurgitations. Approximately 25% of patients with Barrett esophagus discovered at endoscopy have no esophageal symptoms. [90]

Barrett esophagus is suspected at endoscopy and is confirmed by biopsy and histological examination. The columnar epithelium of the stomach is reddish pink, and the junction between the glossy white squamous mucosa and the columnar mucosa (Z-line) is normally found at the lower end of the tubular esophagus, just above the proximal folds of a hiatal hernia, if present. In Barrett esophagus, the distal esophagus is lined with columnar epithelium, extending upward for a variable distance, often 3 to 10 cm, but occasionally involving most of the esophagus (Fig. 60-12A, see also Color Fig. 60-12A). The proximal margin may be horizontal, or there may be irregular, often tongue-shaped upward extensions of columnar mucosa. Some patients have pale islands of regenerative or residual squamous epithelium, whereas others have punched-out benign ulcers in the columnar area. Strictures and esophagitis may be seen at the new squamocolumnar junction. The endoscopist should especially look for evidence of adenocarcinoma, such as nodularity or masses.

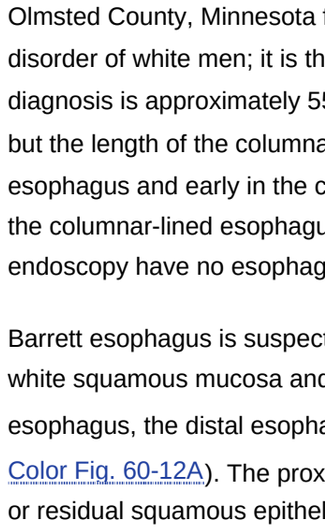

FIGURE 60-12. (See Color Fig. 60-12A.) Barrett esophagus. (**A**) Reddish pink columnar mucosa with tonguelike projections 3 to 4 cm into the tubular esophagus. (**B**) Specialized intestinal metaplasia with glandular epithelium and characteristic goblet cells.

The characteristic histological finding in Barrett esophagus is a distinctive specialized intestinal epithelium. This is a glandular epithelium with mucin-type cells and the distinguishing presence of goblet cells (see Fig. 60-12B). These are easily seen on hematoxylin and eosin–stained sections and can be demonstrated more prominently in sections stained with Alcian blue. It occupies most or all of the columnar-lined area and is the type of epithelium in which adenocarcinoma arises. [164] Other types of epithelia seen with Barrett esophagus include gastric fundic and cardia-type epithelia, but these alone do not make the diagnosis of Barrett esophagus, nor are they associated with adenocarcinoma.

Currently, there is some controversy over the classification of Barrett esophagus. [155] The classical or *long-segment Barrett esophagus* requires at least 3 cm of esophagus to be lined with columnar epithelium. This is the best-studied subset of Barrett esophagus, with traditional demographic features and a definite increased risk of becoming adenocarcinoma. *Short-segment Barrett esophagus* refers to shorter lengths or tongues of columnar epithelium, less than 3 cm, in the distal esophagus, with intestinal metaplasia on biopsy. This entity is three to five times more common than the long-segment variant, but, based on anecdotal reports, the risk of cancer appears to be lower. [165] *Intestinal metaplasia at the esophagogastric junction* refers to microscopic findings on biopsy but no visible columnar epithelium in the esophagus at endoscopy. [155] This finding has been reported in 10% to 32% of biopsies from unselected patients, many of whom have no reflux symptoms. [165] The percentage of woman and African Americans is also higher with this lesion than with either long- or short-segment Barrett esophagus. The cause is controversial; some investigators suggest that this is the earliest form of GERD, [166] whereas others believe these changes are secondary to *H pylori* infection. [167] Cancer risk is minimal, if it exists at all.

Patients with long-segment Barrett esophagus have an estimated 30 to 125 times increased risk of developing esophageal cancer compared with the general population. [168] Early studies suggested that the median cancer incidence was 1 per 100 patient-years of follow-up, [155] but more recent studies with longer follow-up suggest a lower cancer rate of 1 per 200 to 250 patient-years. [169], [170] This is an annual incidence of approximately 0.5%, with about 500 cases of adenocarcinoma diagnosed annually. However, since the early 1980s, the incidence of squamous cell carcinoma has stayed constant, whereas the incidence of adenocarcinoma of the esophagus and esophagogastric junction has risen fivefold—a growth rate exceeding that of any other cancer. [171] Currently, adenocarcinoma accounts for more than half of all esophageal cancers in the United States. Despite this cancer risk, most patients with Barrett esophagus die of unrelated causes. [170] More than 90% of patients who develop cancer present with symptoms caused by the tumor itself and are unaware of their antecedent Barrett esophagus. [172] Epidemiologic data suggest that the mean interval from developing Barrett esophagus to evolution to cancer may be 20 to 30 years. [161]

MEDICAL AND SURGICAL THERAPY

The rationale for GERD therapy depends on a careful definition of specific aims. In patients without esophagitis, the therapeutic goals are simply to relieve the acid-related symptoms and to prevent frequent symptomatic relapses. In patients with esophagitis, the goals are to relieve symptoms and to heal the esophagitis while attempting to prevent further relapses and the development of complications. These goals are set against a complex background: GERD is a chronic disease that may wax and wane in intensity, and relapses are common.

Nonprescription Therapy

Although GERD is common in the United States, very few persons seek medical care for their complaints, instead choosing to change their lifestyles and self-medicate with over-the-counter (OTC) antacids and low doses of H_2RAs. These observations have led to the “iceberg” model of the GERD population. Most heartburn suffers are invisible because they self-medicate and do not seek professional help; only those at the tip of the iceberg, typically patients with severe symptoms or reflux complications, are seen by physicians. [173]

Lifestyle Modifications Sensible *changes in lifestyle,* especially if their rationale is explained to the patient, should be part of the initial management of all subjects. These include head of the bed elevation, avoidance of tight-fitting clothes, weight loss, restriction of alcohol, elimination of smoking, dietary therapy, refraining from lying down after meals, and avoidance of evening snacks before bedtime. Physiological studies show that these maneuvers enhance esophageal acid clearance, minimize acid-reflux related events, or ease heartburn symptoms, but their therapeutic efficacy in controlled trials usually has not been evaluated. [174] The head of the bed can be elevated either by putting 6- to 8-inch blocks under the legs of the bed or by using a Styrofoam wedge under the mattress to elevate the upper torso. Eating several hours before retiring and avoiding evening snacks keep the stomach empty at bedtime, thereby decreasing the number of nocturnal reflux episodes. These three lifestyle changes are recommended for patients with nocturnal GERD symptoms or laryngeal complaints. One study found that head of the bed elevation was nearly as effective as ranitidine therapy in healing esophagitis. [175] Avoidance of tight-fitting clothes and weight loss are interventions aimed at reducing the incidence of reflux by the abdominal stress mechanism. The efficacy of weight reduction is especially controversial, [176] but it may be helpful when discrete periods of weight gain can be associated with exacerbation of reflux symptoms. Cessation of smoking and elimination of alcohol are valuable because both agents lower LES pressure, reduce acid clearance, and impair intrinsic squamous epithelial protective functions. [47], [177], [178] Dietary changes include reducing the size of the meal and intake of fats, carminatives, and chocolate, to reduce the frequency of reflux by decreasing gastric distention and by reducing the episodes of transient LESRs, and avoiding foods that lower basal LES pressure. [173], [176] Additionally, some patients complain of heartburn after consuming citrus drinks, spicy foods, tomato-based products, coffee, tea, or cola drinks. Stimulation of gastric acid or esophageal sensitivity to low pH or hyperosmolar liquid solutions may account for these symptoms. [179] However, the indiscriminate prohibition of food products should be avoided, but rather tailored to those foods that bring on individual symptoms, to promote dietary compliance. Finally, patients should avoid, if possible, drugs that lower LES pressure or can promote localized esophagitis.

Over-the-Counter Medications Over-the-counter antacids, Gaviscon, and H_2RAs (Table 60-14) are useful in treating mild and infrequent heartburn symptoms, especially when symptoms are brought on by lifestyle indiscretions. Antacids increase LES pressure but work primarily by buffering gastric acid in the esophagus and stomach, albeit for relative short periods. Heartburn symptoms are rapidly relieved, but patients need to take antacids frequently, usually 1 to 3 hours after meals and at bedtime, depending on symptom severity. Gaviscon, containing alginic acid and antacids, mixes with saliva to form a highly viscous solution that floats on the surface of the gastric pool and acts as a mechanical barrier. Both antacids [180] and Gaviscon [181] are more effective than placebo in relieving symptoms induced by a heartburn-promoting meal. However, these agents do not heal esophagitis, and long-term trials suggest effective symptom relief in only 20% of patients using antacids. [182], [183] Side effects of antacids and Gaviscon are minimal but include diarrhea from magnesium-containing antacids, constipation from aluminium-containing antacids, salt overload, magnesium or aluminium toxicity in patients with renal disease, and the milk-alkali syndrome (hypercalcemia, alkalosis, renal failure) from long-term and excessive use of calcium-containing antacids. H_2RAs are available in an OTC form at doses that are usually one half of the standard prescription dose. Although there are some differences in potency, duration, and rapidity of action, these drugs may be used interchangeably. Although their onset of relief is not as rapid as that of antacids, the OTC H_2RAs have a longer duration of action, up to 6 to 10 hours. [184] Therefore, they are particularly useful when taken before a potentially refluxogenic activity, such as a heavy meal or exercise. Like antacids, the OTC H_2RAs are ineffective in healing esophagitis and should not be used regularly for more than 2 weeks. [184]

Prescription Medication Therapy

Patients with frequent heartburn, esophagitis, or complications of GERD usually see a physician and receive prescription medications for their disease (see Table 60-4). Although prokinetic drugs attempt to correct the motility disorder associated with GERD, the most clinically effective medications for short- and long-term reflux treatment are the acid suppressive drugs.

TABLE 60-4 Drug Therapy for Gastroesophageal Reflux Disease

Prokinetic Drugs Until recently, three prokinetic drugs were available for the treatment of GERD: bethanechol, a cholinergic agonist; metoclopramide, a dopamine antagonist; and cisapride, a serotonin (5-HT_4) receptor agonist, which increases acetylcholine release in the myenteric plexus. These drugs improve reflux symptoms by increasing LES pressure, acid clearance, or gastric emptying. [185] However, none alter the frequency of transient LESRs, and their physiological activity decreases as the disease severity worsens. [186] Therefore, all the current prokinetics provide modest benefit in controlling heartburn, but they have little efficacy in healing esophagitis unless they are combined with an acid inhibiting drug. [185] The use of prokinetic drugs is limited by their side effect profiles. Bethanechol commonly causes flushing, blurred vision, headaches, abdominal cramps, and urinary frequency. Metoclopramide, which crosses the blood-brain barrier, has a 20% to 50%

incidence of fatigue, lethargy, anxiety, and restlessness and rarely causes tremor, parkinsonism, or tardive dyskinesia. It is possible to decrease the frequency of these side effects by dose reduction, by increasing the dosing regimen to twice a day, by taking a larger single dose before dinner or at bedtime, or by using a sustained-release tablet. Domperidone, another dopamine antagonist and one that does not cross the blood-brain barrier, has fewer side effects, but it is not available in the United States. Although the best prokinetic drug for treating GERD with an excellent safety profile, cisapride was withdrawn from the United States market because of increased reports of serious cardiac arrhythmias (ventricular tachycardia, ventricular fibrillation, torsades de pointes, and QT prolongation) with associated cardiac arrest and deaths related to possible drug interactions. [187] These drugs included common antibiotics (clarithromycin, erythromycin), antifungals (fluconazole, itraconazole, ketoconazole), and some antiviral agents. When taken with cisapride, these drugs inhibited the cytochrome P450 3A4 enzyme that metabolizes cisapride, thereby increasing cisapride blood levels to potentially dangerous values.

Histamine $_2$ Receptor Antagonists Cimetidine, ranitidine, famotidine, and nizatidine reduce acid secretion by competing with histamine receptors on the parietal cell. They are most effective in controlling nocturnal, as compared with meal-related acid, secretion because the parietal cell may also be stimulated postprandially by acetylcholine and gastrin. [188] All the H $_2$RAs are equally effective when used in proper doses, usually twice a day before meals. Clinical GERD trials show that heartburn, both day and night, can be significantly decreased by H $_2$RAs, when compared with placebo, although symptoms are rarely abolished (Fig. 60-13A). [189] Trials and a metaanalysis found that the overall esophagitis healing rates with H $_2$RAs rarely exceeded 60% after up to 12 weeks of treatment, even when higher than standard doses were used (see Fig. 60-13B). [189] , [190] Healing rates differ in individual trials, depending primarily on the degree of esophagitis being treated: grade I and II esophagitis heals in 60% to 90% of patients, whereas grade III and IV heals in 30% to 50% of patients despite high-dose regimens. [190]

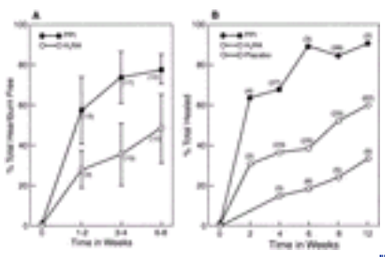

FIGURE 60-13. (**A**) Symptom relief-time curve expressed as the mean total heartburn relief for proton pump inhibitors (*PPIs*) or histamine $_2$ receptor antagonists (*H $_2$RA*) corrected for patients free of heartburn at baseline and over 8 weeks. By week 2, more patients treated with PPIs are asymptomatic compared with patients treated with H $_2$RAs even after a much longer duration of treatment (8 weeks). (**B**) Esophagitis healing-time curve as the mean total healing for PPIs, H $_2$RAs, and placebo over 12 weeks. By 4 weeks, PPIs healed esophagitis in more patients than the other two drug classes over 12 weeks, implying a substantial therapeutic gain. The number of studies is shown in *parentheses*. (From ref. [190] .)

Reflux symptoms associated with nocturnal gastric acid breakthrough during PPI therapy have been recognized. [191] At bedtime, H $_2$RAs successfully eliminated this problem, suggesting a new indication for H $_2$RAs in the PPI era. [192] However, this study used only a single evening dose and did not account for the tolerance that frequently develops to H $_2$RAs over weeks to months. [193] This may impair the ability of chronic long-term nocturnal dosing of H $_2$RAs to eliminate acid breakthrough symptoms, [194] but it suggests an important clinical role as medications used on an as-needed basis when lifestyle indiscretions may promote nocturnal symptoms. As a class of drugs, the H $_2$RAs are very safe, with a side effect rate (most of which are minor and reversible) of about 4%. [188] There have been some concerns about drug interactions with these agents. Serum concentrations of phenytoin, procainamide, theophylline, and warfarin are altered after the administration of cimetidine and, to a lesser degree, ranitidine, whereas this interaction is not reported with the other two H $_2$RAs. The former concern that these agents could alter blood ethanol levels has been discounted. [188]

Proton Pump Inhibitors This class of drugs markedly diminishes gastric acid secretion by inhibiting the final common pathway of acid secretion, the H $^+$, K $^+$-ATPase pump. PPIs inhibit daytime, nocturnal, and meal-stimulated acid secretion to a significantly greater degree than H $_2$RAs, [195] but they rarely make patients achlorhydric. Unlike H $_2$RAs, the degree of acid inhibition with PPIs does not correlate with plasma concentration, but it is related to the concentration and duration (area under the curve). After oral ingestion, acid inhibition is delayed because PPIs need to accumulate in the secretory canaliculus of the parietal cell to bind irreversibly to actively secreting proton pumps. [196] Therefore, the slower a PPI is cleared from the plasma, the more of it is available for delivery to the proton pumps. PPIs are best taken before the first meal of the day, when most proton pumps are active. Because not all pumps are active at any given time, a single PPI dose does not inhibit all pumps. A second dose, if needed, can be taken before the evening meal. The five available PPIs are omeprazole, lansoprazole, rabeprazole, pantoprazole, and esomeprazole, the S-isomer of the racemic omeprazole. Their superior efficacy compared with H $_2$RAs is based on their ability to maintain an intragastric pH high than 4 between 15 to 21 hours daily compared with approximately 8 hours daily with the H $_2$RAs. [197] Multiple studies show that the PPIs are superior to H $_2$RAs in completely relieving heartburn symptoms in most patients with severe GERD, usually within 1 to 2 weeks (see Fig. 60-13A). [190] Symptom relief is slightly better in patients with erosive as compared with nonerosive disease. [198] Controlled studies and a large metaanalysis report complete healing of even severe ulcerative esophagitis after 8 weeks in more than 80% of patients taking PPIs compared with 51% of patients taking H $_2$RAs and 28% receiving placebo (see Fig. 60-13B). [199] , [200] , [201] and [202] In those patients not healing initially, prolonged therapy with the same dose or an increased dose usually resulted in 100% healing. [203] Studies with PPIs consistently show that they are superior to both regular and high-dose H $_2$RA therapy, but therapeutic efficacy among PPIs is similar. However, a large study found that the newest PPI, esomeprazole, at a dose of 40 mg, was superior to the parent compound (omeprazole, 20 mg) for complete healing of esophagitis at 4 and 8 weeks. [204] This superiority over omeprazole is related to higher systemic bioavailability and less interpatient variability with esomeprazole. One PPI, pantoprazole, is available in the United States for intravenous use. [205] All the PPIs, are well tolerated with headaches and diarrhea described as the most common side effects in clinical trials. Although increased gastrin levels are reported with all the PPIs, the elevations generally do not exceed the normal range for gastrin and return to normal values within 1 week of stopping the drug. Omeprazole may decrease the clearance of diazepam and warfarin because of competition for the cytochrome P450 isoenzyme P2C19. [206] The four newer PPIs have minimal or no important drug-drug interactions.

Maintenance Therapy

GERD tends to be a chronic relapsing disease, especially in patients with low LES pressure, severe grades of esophagitis, and difficult to manage symptoms. [183] Although almost all patients with severe esophagitis can be healed with PPI treatment, recurrence can be anticipated in more than 80% of patients within 6 months of drug discontinuation. [145] The chronicity of less severe forms of GERD is less certain, but relapses probably occur in 15% to 30% of patients over 6 months. [207] Therefore, maintenance therapy is needed for many patients.

One-year maintenance studies always find the PPIs superior to H $_2$RAs or prokinetics, with remission rates higher than 75%. [146] , [208] , [209] H $_2$RAs and prokinetic drugs have lower overall remission rates (20% to 50%) and are most useful in patients with mild or no esophagitis. [146] , [210] The United States Food and Drug Administration (FDA) has approved all the PPIs, sometimes at half the short-term dose, for maintenance therapy (see Table 60-4), but only ranitidine, 150 mg twice daily, has maintenance indications for mild esophagitis. Conversely, clinicians are now placing their patients with severe erosive esophagitis on long-term PPI therapy indefinitely. The efficacy of this approach is supported by open, compassionate use data, primarily from the Netherlands and Australia. [211] , [212] In a study of 230 patients with severe esophagitis healed initially with 40 mg omeprazole, all subjects were kept in remission for up to 11 years. More than 60% were maintained on omeprazole, 20 mg a day, whereas higher doses of 60 mg or more were needed in only 12% of patients, suggesting a lack of tolerance to PPIs. Relapses were rare (1 per 9.4 years of follow-up), strictures did not occur, and Barrett esophagus did not progress. [212]

There was concern about the long-term safety of PPIs because of their profound acid suppression. Current evidence suggests this fear is unjustified because sufficient gastric acid is produced allowing for normal protein and carbohydrate digestion, iron and calcium absorption, and the prevention of bacterial overgrowth. The clinical effect of omeprazole on vitamin B $_{12}$ absorption is controversial. [213] Therefore, it may be prudent to monitor B $_{12}$ levels in patients receiving long-term PPI therapy, especially elderly patients or those with poor or unusual diets. The main concern with the long-term safety of PPIs stemmed from reports of omeprazole's producing hypergastrinemia and gastric carcinoid tumors in rats, changes also subsequently demonstrated with long-term ranitidine therapy and subtotal resection of the gastric fundus. [214] However, the rat has a high density of enterochromaffin-like cells and an exaggerated response to achlorhydria; long-term omeprazole therapy in species (mice, dogs, humans) with lower densities of enterochromaffin-like cells has not caused carcinoid tumors. Furthermore, other groups with massive hypergastrinemia (five to ten times the gastrin values on omeprazole), such as patients with pernicious anemia or Zollinger-Ellison syndrome, rarely develop carcinoid tumors. [214] Finally, one study suggested that patients taking long-term

omeprazole who are infected with *H pylori* develop atrophic gastritis, a precursor to gastric adenocarcinoma at a more rapid rate than noninfected patients. [215] Nevertheless, a subsequent FDA panel determined that the available data were insufficient for recommending screening and treatment of *H pylori* infection in patients receiving long-term PPI therapy. [216]

Treatment in Elderly or Pregnant Patients

Older patients often complain of less severe reflux symptoms than their younger cohorts, but because of prolonged acid exposure over years, the elderly may have more complicated disease. [5] Treatment of the older patient with GERD follows the same principles as in other adults, although they may require more aggressive acid suppression therapy. [217] Pill-induced esophagitis may complicate their treatment. Metoclopramide must be used with caution because of frequent side effects in the elderly. H_2RAs can be associated with mental changes in older patients, and doses need to be decreased in patients with renal insufficiency. Fewer drug interactions are seen with famotidine and nizatidine. Alternative methods of administering PPIs may be necessary in debilitated older patients who cannot swallow intact omeprazole or lansoprazole capsules. Both capsules can be opened and the granules taken with water, an HCO_3^--based suspension, or apple or orange juice, or the granules can be sprinkled on applesauce or yogurt. [218]

Teratogenicity or fetal harm from absorption of medications across the placenta is the foremost consideration in the treatment of GERD during pregnancy. [219] Lifestyle modifications and antacids or Gaviscon remain the cornerstones of treatment, providing adequate relief to the majority of women with mild symptoms. Although rarely used in adults, sucralfate is a nonabsorbable mucosal binder that has been found superior to lifestyle changes in a controlled study in pregnant women. [220] Metoclopramide, H_2RAs, and most PPIs (except omeprazole) have a Category B FDA safety profile for use during pregnancy, based on animal studies showing no risk, as well as on small case series and anecdotal human reports. Ranitidine is the only one of these drugs shown to be effective during pregnancy. [221] PPIs may be safe for aspiration prophylaxis before anesthesia for elective cesarean sections. Antacids, sucralfate, and most H_2RAs (except nizatidine) are safe to use during lactation, even though the latter group of drugs is excreted in breast milk. PPIs are not recommended during breast-feeding, based on safety concerns in animal studies. [219]

Treatment of Complications

Extraesophageal Presentations Acid reflux–related chest pain is easily treated by H_2RAs or PPIs, with efficacy substantiated by placebo-controlled studies. [222] The efficacy of acid suppression therapy in asthma, cough, and other pulmonary complications of GERD is more mixed. [98] Medical antireflux therapy improves asthma symptoms and reduces the need for asthma medications in more than 60% of patients, but objective improvement of peak expiratory flow rates is observed in only 25% of patients. Best results are found with higher doses of PPIs (usually twice-daily administration) given for 2 to 3 months. Potential positive predictors of PPI response include asthma that is difficult to control, associated acid regurgitation, proximal reflux on pH testing, and healing of esophagitis with antireflux therapy. Case studies report that 60% to 96% of patients with suspected acid-related ear, nose, and throat symptoms and signs improve with acid suppression. [104] Here again, PPIs are more effective than H_2RAs, and extended therapy for up to 3 months may be required. Predictors of response have not been identified, although patients with milder laryngeal signs show better symptom improvement. In all these possible extraesophageal presentations of GERD, failure to respond to aggressive PPI therapy, confirmed by adequate acid control by pH testing, suggests a cause of these complaints other than acid.

Esophageal Strictures Dysphagia in patients with esophageal strictures is related to stricture diameter and severity of esophagitis. [223] When the esophageal lumen diameter is less than 13 mm, dysphagia is a major complaint and esophageal dilation is required. Simple short strictures can be dilated by blind peroral passage of rubber Hurst (round ends) or Maloney (taper ends) mercury-filled dilators of increasing sizes (16 to 60 French, 3 French = 1 mm) to disrupt the fibrous bands producing the obstruction. Complicated longer, tighter, or more irregular strictures will require bougienage over a guidewire using hollow-centered Savary plastic-covered polyvinyl dilators or balloon (Gruentzig) dilators. The techniques of esophageal dilation are discussed in detail in Chapter 145. [224] Before and after dilation, medical therapy with PPI is indicated; it has been shown to be superior to H_2RAs in relieving symptoms and in reducing the frequency for repeat dilations. [225] Maintenance PPI therapy for patients with strictures has dramatically reduced the incidence of repeat esophageal dilations and the cost of treating these patients. [152] Recalcitrant strictures, requiring surgery, are now very uncommon and suggest another aggravating factor such as chronic pill injury.

Barrett Esophagus In general, esophagitis in the presence of Barrett esophagus can be easily healed with PPI therapy, but meaningful regression of Barrett epithelium, except for small squamous islands, is rarely reported even with high-dose PPI therapy. [226] Recent ex vivo studies [227] have suggested that intermittent “pulses” of acid result in enhanced Barrett epithelial cell proliferation, possibly increasing the risk of dysplasia and cancer. Some have suggested the need to eliminate all acid reflux in patients with Barrett esophagus, [228] but this would require high and frequent doses of expensive medications and serial pH monitoring to document efficacy of therapy. Pending further clinical studies, patients with Barrett esophagus should be treated like others with chronic GERD. Esophageal resection of Barrett esophagus can prevent the progression to cancer, but this requires a total esophagectomy with high mortality, except in selected surgical centers. Therefore, ablation of Barrett epithelium in the setting of strict PPI anacidity has been proposed. Photodynamic therapy, laser, multipolar electrocoagulation, argon plasma coagulation, and endoscopic mucosal resection have been used for this purpose. [229] In these studies, Barrett mucosa can be reversed completely in 70% to 80% of patients, but intestinal metaplasia underlying the new squamous mucosa is reported in almost all series, with the occasional residual foci of metaplasia developing adenocarcinoma. [230] Adverse effects of ablation therapy have ranged from mild chest pain, sore throat, or odynophagia to esophageal perforation and death. The incidence of adenocarcinoma in patients with Barrett esophagus without dysplasia is probably so low that endoscopic ablation cannot be advocated outside of study protocols. Endoscopic therapy for patients with high-grade dysplasia or early cancer holds more promise, especially in the older patients with comorbid illnesses. [231] Because there is currently no way of eliminating the malignant risk of Barrett esophagus, regular endoscopic surveillance is recommended. Biopsies should be taken from each quadrant every 2 cm axially within the metaplastic tissue. The rationale is that dysplasia within Barrett epithelium is often multifocal, and obtaining fewer tissue samples increases the risk of missing dysplastic areas. [232] Brush cytology can compliment endoscopic biopsies. [233] Biomarkers, [234] such as p53, and flow cytometry [235] may augment the yield of histological examination of biopsy specimens. Although prospective studies are not available, case series confirm that esophageal adenocarcinomas detected by endoscopic surveillance are at an earlier stage with a more favorable survival than cancers detected at the time of diagnosis of Barrett, typically when patients present with dysphagia. [236] The appropriate surveillance interval for patients with Barrett esophagus has not been studied prospectively. However, current programs, such as proposed by the American College of Gastroenterology, are based on the grade of dysplasia (Table 60-5). [237] In these recommendations, the management of high-grade dysplasia remains most controversial. Some groups suggest that high-grade dysplasia may regress to lesser grades, and an intensive biopsy protocol every 3 months will differentiate high-grade dysplasia from cancer. [238], [239] The surgical literature contrasts with this experience. Of 126 cases with high-grade dysplasia alone by endoscopic biopsies, 41% had cancer, although usually an early stage, at the time of esophagectomy. [237] Nevertheless, most patients with Barrett esophagus never progress to important degrees of dysplasia. Predictors of cancer progression at the initial diagnosis of Barrett esophagus are needed to define individual surveillance programs more appropriately. One study suggested that patients whose baseline biopsies are negative or show only low-grade dysplasia without increased 4N or aneuploidy on flow cytometry may have surveillance deferred for up to 5 years. [239] Another prospective multivariate analysis revealed that progression to high-grade dysplasia or cancer was significantly and independently associated with dysplasia at diagnosis or anytime during follow-up, hiatal hernia size greater than 2 cm, and Barrett esophagus length greater than 2 cm. [240] These patients may warrant more frequent surveillance programs.

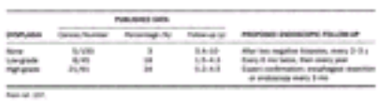

TABLE 60-5 Barrett Esophagus: Grade of Dysplasia and Proposed Follow-Up

Surgical Treatment

Antireflux surgery reduces GER by increasing basal LES pressure, decreasing episodes of transient LESRs, and inhibiting complete LESR. [241] This is accomplished by reducing the hiatal hernia back into the abdomen and thereby restoring an adequate length of intra-abdominal sphincter, reconstructing the diaphragmatic hiatus, and reinforcing the LES. [242] Before the explosion of laparoscopic surgery, the three most common operations were the Nissen fundoplication, the Belsey Mark IV repair, and the Hill posterior gastropexy repair. Since the advent of minimally invasive surgery, the two most popular procedures are the Nissen fundoplication and the Toupet partial fundoplication (Fig. 60-14). The former is a superior operation with more long-term durability, but it has a higher frequency of postoperative dysphagia and gas bloat symptoms. [243], [244] Both are now routinely performed laparoscopically through the abdomen. The hospital stay is 1 to 2 days, and many patients return to normal activity in 7 to 10 days. Patients with more severe disease and a short esophagus manifested by a large nonreducible hernia, a tight stricture, or a long-segment Barrett esophagus require a Collis lengthening procedure creating a 3- to 5-cm neoesophagus, so the fundoplication can be placed in the abdomen under minimal tension. [245]

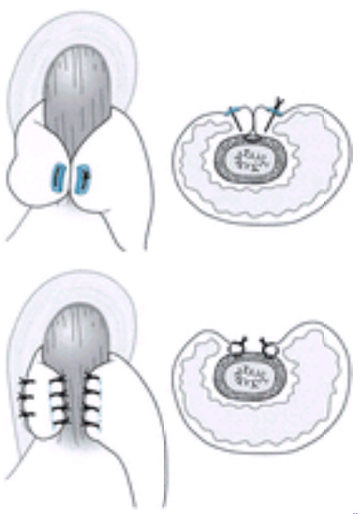

FIGURE 60-14. Appearance and cross-sectional views of the Nissen fundoplication, a full 360° wrap (**top**), and the Toupet partial fundoplication (**bottom**). These are the two most popular fundoplications used in laparoscopic antireflux surgery.

The common indications for antireflux surgery have evolved with the availability of more powerful medications for treating GERD. In the PPI era, the resolution of symptoms on treatment helps to predicate the success of antireflux surgery for both classical and atypical symptoms. [246] Antireflux surgery is a reasonable option in the following situations:

1. Healthy patients with GERD well controlled on PPIs who want alternative therapy because of drug expense, poor medication compliance, or a fear of unknown long-term side effects
2. Patients with atypical GERD symptoms responding to PPIs
3. Patients with volume regurgitation and aspiration symptoms not controlled on PPIs.

Patients recalcitrant to PPI therapy need to be approached cautiously with surgery because they may have another cause of their disorder (i.e., pill esophagitis, gastroparesis, functional heartburn).

Extensive physiological testing should be done before antireflux surgery is performed. All patients need endoscopy to exclude stricture, Barrett esophagus, and dysplasia. A barium esophagram can help define a nonreducible hernia, shortened esophagus, and poor esophageal motility. Esophageal manometry will identify ineffective esophageal peristalsis and previously misdiagnosed achalasia or scleroderma. Twenty-four hour pH testing is needed in all patients with nonerosive GERD or in those with esophagitis not responding to PPI therapy. Gastric analysis and gastric emptying studies may be indicated in selected patients. Careful testing will result in modification of the original operation or an alternative diagnosis in approximately 25% of patients. [131]

Antireflux surgery relieves reflux symptoms and reduces the need for stricture dilation in more than 90% of patients, [243] but Barrett esophagus rarely regresses, and the influence of surgery on the development of esophageal adenocarcinoma is controversial. [247] Comparison studies have found antireflux surgery superior to lifestyle changes, antacids, and H_2RA and prokinetic therapy, [149], [182] but not PPI therapy, especially when dose titration is permitted. [248] Mortality is rare (<1%) after antireflux surgery, but new postoperative complaints can occur in up to 25% of patients including dysphagia, gas bloat, diarrhea, and increased flatus. [249] Most symptoms improve over 1 year, but persistent complaints suggest too tight a wrap, a displaced fundoplication, or inadvertent damage to the vagus nerve. Successful antireflux surgery, however, does not guarantee a permanent cure. The best surgical results are obtained by experienced surgeons in high-volume centers who report long-term symptom recurrence in only 10% to 15% of patients. [243] However, most operations are performed by community or Veterans Affairs hospital surgeons. Here the results are not as good with studies finding relapse of esophagitis in 30% of patients [250] and return to regular use of antireflux medications in 62% of patients [251] 10 to 15 years after fundoplication. Potential factors contributing to these high relapse rates include inexperienced surgeons, low numbers of operations yearly per surgeon, and persistence of abdominal stressors (i.e., obesity, heavy isometric exercise or work) that gradually weaken the fundoplication. A suboptimal operation or severe symptom relapse may necessitate a second operation, which has less likelihood of a successful outcome. [242] Because optimal medical therapy is available to all patients with GERD, the risk and benefits of both long-term medical treatment and antireflux surgery must be carefully discussed with patients, so they can take part in this important decision.

New Treatments

The future medical treatment of GERD involves drugs that interfere with transient LESRs but do not cause dysphagia. Baclofen, a $GABA_B$ agonist, has been shown to decrease reflux symptoms and to improve pH studies in healthy persons and in patients with GERD. [252] Newer endoscopic treatments of GERD have been approved by the FDA. These techniques include an endoscopic suturing system, radiofrequency energy delivery to the gastroesophageal junction, and the injection of nonabsorbable polymers into the submucosa surrounding the LES. To date, these techniques have been applied only to patients with small (<2 cm) or no hernias and mild esophagitis. Results are encouraging, with improvement of symptoms and decreased medication requirements, but follow-up is short (<1 year), and a comparison with sham surgery has not been done. [253]

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CHAPTER 61

C. Mel Wilcox

ESOPHAGEAL INFECTIONS AND DISORDERS ASSOCIATED WITH ACQUIRED IMMUNODEFICIENCY SYNDROME

Remarkable progress has been made in the treatment of human immunodeficiency virus (HIV) infection as well as in the care of immunosuppressed patient. For patients with acquired immunodeficiency syndrome (AIDS), the efficacy and widespread adoption of highly active antiretroviral therapy (HAART) have resulted in a marked reduction in the incidence of opportunistic infections including those involving the esophagus. [1] Likewise, in the setting of organ transplantation, further advancements in immunosuppressive therapy and targeted antimicrobial prophylaxis for patients at high risk of developing infection, such as cytomegalovirus (CMV) infection, have also resulted in a reduction in esophageal infections. Nevertheless, the number of patients at risk of opportunistic infections remains high, especially as our population grows older, immunosuppressive therapy becomes more widely employed to treat other diseases, and the use of organ transplantation expands. Timely and accurate diagnosis of esophageal infections is critical because treatment is highly effective and usually results in rapid symptomatic improvement and often clinical cure. This chapter focuses on the specific causes of esophageal infections, in particular epidemiology, pathology, presentation, diagnosis, and therapy. Esophageal disorders associated with HIV infection and AIDS are also reviewed here.

EPIDEMIOLOGY AND PREDISPOSING FACTORS

Primary esophageal infection is rare in an otherwise healthy person in whom no permissive factor is present. In this setting, the most common pathogen is herpes simplex virus (HSV), [2] although candidiasis may be observed in elderly patients without other predisposing factors. [3] In general, immunocompetent patients who develop esophageal infection have conditions, either local or systemic, that weaken esophageal defense mechanisms. Normal oroesophageal flora may be altered by the use of antimicrobial agents, thus allowing overgrowth of *Candida.* Disorders of esophageal emptying including achalasia, progressive systemic sclerosis, benign or malignant esophageal strictures, and diverticula predispose to esophageal infection, usually with *Candida,* because of stasis of esophageal contents. Infection in adjacent structures may spread into and involve the esophagus secondarily.

More commonly, some form of humoral or cellular immunodeficiency leads to esophageal infection. Underlying conditions that predispose to infections include diabetes mellitus, alcoholism, malnutrition, malignant diseases, and advanced age. Hyperglycemia impairs granulocyte function and leads to candidal disease. Corticosteroid therapy predisposes to infection by suppressing lymphocyte and granulocyte function. Mucosal disruption commonly follows chemotherapy or radiation and provides a portal of entry for pathogens. Transplantation predisposes to infections through variable mechanisms, most of which affect both B-cell and T-cell number and function, including use of immunosuppressive agents, chemotherapy, and neutropenia. Episodes of rejection in recipients of solid organ transplants are also commonly complicated by infection, because these patients are given more immunosuppression including powerful agents such as antithymocyte globulin. Broad-spectrum antibiotics, antisecretory therapy, and surgical trauma further predispose to esophageal infections.

Infection after transplantation has a predictable time course. Bacterial and fungal infections are most common during the initial months after transplantation, because it is during this period that granulocyte number or function is most severely compromised. HSV infection also tends to occur early after transplantation because of reactivation of disease, whereas CMV typically presents 2 to 6 months after transplantation at a time when neutropenia is common and T-cell function is most severely impaired. The development of opportunistic infections in HIV-infected patients reflects severe immunodeficiency; esophageal infections usually become clinically manifest when the CD4 lymphocyte count is less than 200/mm ^{3}. [4], [5]

FUNGAL INFECTIONS

Candida Species

Epidemiology *Candida* species are the most common esophageal pathogens, primarily *C albicans,* but occasionally *C tropicalis, C parapsilosis, C glabrata,* and *C dublinensis.* These organisms are normal components of the oral flora, and their growth is kept in check by bacterial commensals. Conditions predisposing to *Candida* esophagitis in the "normal" host include antibiotic use, inhaled or ingested corticosteroids, antisecretory therapy (histamine H_2 receptor blockers, proton pump inhibitors or hypochlorhydric states, diabetes mellitus, alcoholism, malnutrition, old age, head and neck radiotherapy, and esophageal motility disturbances. [6] Alterations in cellular immunity lead to candidal colonization and superficial infection, whereas granulocytes function to prevent invasive disease and dissemination. The congenital immunodeficiency chronic mucocutaneous candidiasis may also be complicated by *Candida* esophagitis. Improvements in immunosuppressive regimens, targeted prophylactic antifungal therapy, and empiric use of fluconazole in symptomatic patients have dramatically reduced the incidence of candidal infections in recipients of solid organ and bone marrow transplants. [7] Candidiasis remains the most common esophageal infection in patients with AIDS; it represents approximately 50% of esophageal infections, [8], [9] and it frequently coexists with other esophageal diseases. [10]

Pathology The gross pathological appearance of esophageal candidiasis can range from a few white or yellow plaques on the mucosal surface to a dense, thick plaque that coats the esophageal mucosa (Fig. 61-1A; see also Color Fig. 61-1). This plaque material is composed of desquamated squamous epithelial cells, admixed with fungal organisms, inflammatory cells, and bacteria [11] (Fig. 61-1B,C; see also Color Fig. 61-1). True ulceration is infrequent and has been documented most commonly in immunosuppressed patients with granulocytopenia (Table 61-1). [12]

TABLE 61-1 Pathologic Findings of Esophageal Infections

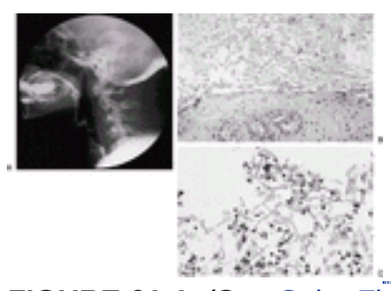

FIGURE 61-1. (See Color Fig. 61-1.) *Candida* esophagitis. (**A**) Endoscopic photograph shows multiple raised plaques involving the esophagus with normal intervening mucosa. (**B**) Desquamated squamous epithelial cells admixed with fungi and inflammatory cells adherent to the mucosa. The underlying squamous epithilium appears normal. (**C**). Close-up view of the plaque material demonstrates mycelia and spores typical of *Candida.*

Clinical Manifestations and Complications Although this infection is detected on occasion incidentally in an asymptomatic patient, the usual clinical presentation of esophageal candidiasis is dysphagia, with odynophagia less prominent (Table 61-2). Esophageal symptoms range from mild difficulty with swallowing to severe pain resulting in an inability to eat and secondary dehydration. When odynophagia is very severe, one must consider other causes or co-infections, particularly in patients with AIDS.

TABLE 61-2 Symptoms and Signs of Esophageal Infection

Physical examination may be helpful. Approximately two thirds of patients with AIDS and esophageal candidiasis have oral candidiasis (thrush). [13] Patients with chronic mucocutaneous candidiasis may have fungal involvement of various mucous membranes, hair, nails, and skin with a history of adrenal or parathyroid dysfunction. Complications of esophageal candidiasis are very rare. Esophageal hemorrhage may occur when the disease is severe (erosion or ulcer) and there is associated coagulopathy. Lumenal obstruction secondary to mycetoma, fibrosis and stricture formation, and fistulization into the bronchial tree have been described, [14] but these usually represent *Candida* colonization of these anatomic abnormalities caused by other underlying disorders.

Diagnosis Esophageal candidiasis should be suspected in any patient at risk of esophageal infection who complains of dysphagia or odynophagia. The presence of thrush further supports this diagnosis, but the absence of oral involvement does not exclude esophageal disease. Before the availability of endoscopy, radiographic examination (barium esophagram) was commonly used for initial evaluation. *Candida* esophagitis is characteristically manifested by multiple plaquelike lesions, often in a linear configuration; when severe, these lesions become confluent to cause a “shaggy” appearance of the esophagus resembling ulcerations (see Fig. 61-1). [15] Additional radiographic findings include pseudomembranes, cobblestoning, polypoid nodules, fungus balls, strictures, esophagopulmonary fistulae, mucosal bridges, or large, neoplastic-appearing esophageal ulcers and masses. [16] A large, well-circumscribed ulceration should not be attributed to *Candida.* Importantly, a normal barium esophagram does not exclude esophageal candidiasis. The presence of severe odynophagia may limit the ability of the patient to drink barium and thus hampers the utility of barium studies. Endoscopic examination of the esophagus is the most accurate method of diagnosing esophageal candidiasis. The gross endoscopic appearance of *Candida* esophagitis is usually diagnostic and may be graded according to published criteria. [11] During endoscopy, mucosal lesions can be brushed and submitted for cytologic evaluation or examined by biopsy for histological diagnosis. Multiple biopsies are essential to exclude coexisting disorders when ulceration is identified endoscopically. The use of periodic acid-Schiff or Gomori methenamine silver stain helps to highlight the organisms. Cytologic examination of esophageal brushings is more sensitive than histological examination of biopsy specimens because organisms may be washed off tissue surfaces in mild superficial candidiasis (i.e., grades 1 and 2) during processing of biopsy specimens. [3] Rarely, positive cytology but negative histology indicates colonization rather than infection. Skin testing and serologic tests for candidiasis are not useful in the diagnosis of *Candida* esophagitis. In patients with AIDS and thrush, the presence of dysphagia or odynophagia usually indicates *Candida* esophagitis. [5], [9] Given the prevalence of *Candida* esophagitis in AIDS, in the symptomatic patient with associated thrush, an empiric trial of antifungal therapy may be instituted, reserving endoscopy for those patients who fail to respond. However, this symptom complex does not exclude co-infection with other pathogens, and one third of patients with esophageal candidiasis do not have thrush. Further evaluation should not be delayed longer than 1 week in patients with severe persistent symptoms because the response to antifungal therapy is rapid, with clinical improvement occurring in most patients within days. [9], [17] When patients fail to improve with empiric antifungal therapy, endoscopy should be performed, given that disorders other than candidiasis are identified in most patients. [18], [19] The role of empiric antifungal therapy in symptomatic immunosuppressed patients who do not have AIDS has not been well studied, although this approach is commonly practiced.

Treatment Both oral and intravenous medications are available that are highly effective for the treatment of *Candida* esophagitis (Table 61-3). In general, oral therapies should be first initiated, reserving intravenous treatment for refractory disease or for patients who have contraindications to orally administered medication. Although candidal species other than *C albicans* may cause esophagitis, speciation is not widely employed because reliable culturing and sensitivity testing are lacking at most centers. For those patients with mild disease, minimal immunocompromise, or readily reversible immunodeficiency, an abbreviated course of therapy with an oral azole should be given. Immunocompromised transplant recipients and patients with AIDS with *Candida* esophagitis are best treated with systemically absorbed agents (azoles). In addition, patients with granulocytopenia are at significant risk of disseminated candidal infection, thus warranting the use of systematically acting intravenous agents such as the azoles or amphotericin B.

TABLE 61-3 Treatment Regimens for Common Esophageal Disease in AIDS

Orally administered systemically agents include ketoconazole (Nizoral), fluconazole (Diflucan), and itraconazole (Sporanox). All have efficacy for the treatment of *Candida* esophagitis. These agents, like other azoles, alter fungal cell membrane permeability by cytochrome P450–dependent interference with ergosterol biosynthesis resulting in fungal cell injury and death. The newer triazoles (itraconazole and fluconazole) have greater affinity than the imidazoles (miconazole and ketoconazole) for fungal cytochrome P450 enzymes. [20] Although other agents such as clotrimazole and nystatin may be effective for oral candidiasis and for prophylaxis against esophageal involvement, [21] these agents are less effective as primary therapy for esophageal candidiasis. Ketoconazole therapy (200 to 400 mg/d) is effective for the treatment of esophageal candidiasis. The dose may be increased, nausea permitting, to a maximum of 800 mg/d. Patients with AIDS usually require higher doses of ketoconazole, and a starting dose of 400 mg/d in divided doses has been recommended. Optimal absorption requires an acid milieu; thus, situations of gastric hypoacidity, such as pernicious anemia or iatrogenic hypochlorhydria, through the use of antisecretory agents, will reduce bioavailability. [22] Because patients with AIDS may have reduced gastric acid secretion that decreases absorption and thus impairs the efficacy of this agent, [23] ketoconazole is not generally recommended for initial therapy. The dose of itraconazole is 200 mg/d, and as with ketoconazole, further dose increases lengthen the drug’s half-life and improve efficacy. Itraconazole absorption is also reduced by increasing gastric pH. [24] Ketoconazole and itraconazole are extensively metabolized in the liver and are excreted in the bile. The half-life of these two agents are 7 to 10 hours and 24 to 42 hours, respectively. [20] Dose adjustments are not required in the patient with renal failure. Large randomized studies suggest that fluconazole (100 mg/d), the absorption of which does not depend on pH, has significantly greater efficacy for the treatment of *Candida* esophagitis in AIDS than ketoconazole (200 mg/d) [25] and itraconazole. [26] Fluconazole is available in oral and intravenous preparations. It is minimally metabolized and is excreted unchanged in the urine. Unlike ketoconazole and itraconazole, fluconazole is highly water soluble and is minimally protein bound. The half-life is approximately 30 hours if renal function is normal, and the presence of food or hypochlorhydria does not alter absorption. Both fluconazole and

itraconazole are also available in oral solutions. [27] The adverse effects of ketoconazole, fluconazole, and itraconazole are primarily dose dependent and include nausea, hepatotoxicity, and inhibition of steroid production and cyclosporine metabolism. [20] Minor increases in aminotransferases are common with these three agents but do not warrant drug discontinuation. Reversible inhibition of gonadal and adrenal steroid synthesis by ketoconazole may occur with doses of 400 mg/d or greater. [28] In recommended doses, fluconazole and itraconazole do not affect steroidogenesis. Finally, because of the effects on hepatic microsomal enzymes, all three azoles inhibit the metabolism of cyclosporine and result in an increase in cyclosporine blood levels, and this effect is most pronounced with ketoconazole. [20] Other important drug interactions with these agents have been noted, although these tend to be more common with ketoconazole. The other major family of antifungal agents consists of the polyene antibiotics, represented by amphotericin and nystatin (see Table 61-3). These agents bind irreversibly to sterols in fungal cell membranes, thereby altering the permeability characteristics of the membrane and causing cell death. Nystatin is effective for treating thrush but less so for esophageal disease. Furthermore, the efficacy, safety, and ease of administration of azole derivatives have made nystatin a second-line therapy. Although amphotericin B (Fungizone) is the most effective treatment for systemic mycoses, its severe side effects, coupled with the availability of azoles, have limited its use for the treatment of esophageal candidiasis. This agent is now available in an oral solution. Patients with esophageal candidiasis resistant to treatment with fluconazole or other azoles can be treated effectively with lower doses of intravenous amphotericin B (10 to 20 mg/d). Renal toxicity, which is usually reversible, is the most serious adverse effect of continued use of amphotericin B. The total dose for the treatment of esophageal candidiasis is approximately 100 to 200 mg. Combination therapy of flucytosine with amphotericin has been used in some patients with treatment-resistant disease.

Prophylaxis The use of ketoconazole or nystatin for the prevention of esophageal candidiasis in patients with cancer and in transplant recipients has yielded mixed results, and it can be problematic, especially in those receiving cyclosporine. [29] Primary prophylaxis has not been recommended in patients with AIDS, [30] although the long-term use of fluconazole in AIDS has been associated with a reduction in cryptococcal disease. [31]

Drug Resistance Because of widespread use, azole resistance has become an increasing problem in HIV-infected patients. Both the cumulative dose of azole and severe immunodeficiency have been shown to be highly associated with the development of resistance. [32] In patients with AIDS, clinical resistance correlates with in vitro resistance. When resistance occurs, increasing the dose of azole is often helpful. If higher doses fail, switching to another azole or use of oral solution of itraconazole [33] may be tried in higher doses because cross resistance is often present. Intravenous amphotericin B is usually required when high dose (>400 mg/d fluconazole) therapy fails. Resistance to amphotericin is rare. Improvement in immune function with HAART is also effective for the treatment of resistant candidiasis. [34]

Other Fungi

Epidemiology Esophageal involvement with other fungi is rare. Most instances of histoplasmosis and blastomycosis esophagitis represent secondary esophageal involvement from mediastinal lymph nodes rather than primary esophageal infection. [35], [36] Although no particular geographic distribution within the United States has been reported for aspergillosis or blastomycosis, histoplasmosis is endemic in the midwestern states and the Mississippi Valley. Mucormycosis has also been described in a patient with AIDS. [37]

Pathology, Clinical Manifestations, and Complications Other than the development of fistula, there are no unique pathological features of these fungal infections (see Table 61-1). Pulmonary symptoms may predominate when there is fistula formation to the tracheobronchial tree. Recognition depends on appropriate staining of biopsy specimen with the identification of the characteristic fungal elements. Histoplasmosis and blastomycosis are more likely to cause focal lesions as a consequence of extension from mediastinal lymph nodes, although mucosal infection occurring during the course of disseminated histoplasmosis has been noted in patients with AIDS. [38]

Diagnosis Although esophageal aspergillosis is rare, it should be considered in cases of apparent *Candida* esophagitis that are resistant to appropriate therapy. [39] Histoplasmosis should be considered in endemic areas and if extraesophageal manifestations such as hilar adenopathy, calcification or atelectasis of adjacent pulmonary tissue, or splenic calcification are present. Esophageal blastomycosis should be considered in patients with skin involvement and dysphagia. Endoscopy with biopsies and histological examination (with cytologic brushings) may establish the diagnosis if the pathologist is able to differentiate the septate hyphae of *Aspergillus* species from the pseudohyphae of *Candida* species. Culture of biopsy material using fungal media can be diagnostic. Barium esophagraphy or endoscopy may show a focal area of extrinsic compression of the esophagus, usually in the region of the carina, ulcer, or fistula. Because *Histoplasma capsulatum* does not generally invade the esophageal mucosa, endoscopic brushings or biopsies may not be diagnostic. Serologic tests are not useful because of the high prevalence of positive results in endemic areas. A urine antigen test has been developed that is highly specific for disseminated histoplasmosis.

Treatment Although histoplasmosis may resolve without antifungal therapy in the normal host, therapy should be administered. Ketoconazole, itraconazole, and amphotericin B are effective against histoplasmosis and blastomycosis. [20] Because of the toxicity associated with amphotericin, this agent should probably be reserved for severe infections or for cases of failure of ketoconazole therapy. Systemic aspergillosis should be treated with high-dose amphotericin B, although itraconazole has significant in vitro activity. Surgery may be required for drainage of abscesses or excision of fistulae.

VIRAL INFECTIONS

With the administration of *Candida* prophylaxis in selected patients undergoing transplantation coupled with the increasing use of oral antifungal therapies in AIDS, viral esophagitis is assuming more etiologic importance. Nevertheless, in the transplant setting, the incidence of clinically apparent viral esophageal disease has been decreasing as a result of targeted antiviral prophylaxis for herpesviruses in high-risk transplant recipients, the use of CMV-seronegative organs and blood products for seronegative recipients, and the use of leukocyte-depleted platelets for patients after bone marrow transplantation.

Herpes Simplex Virus

Epidemiology HSV type 1 (HSV-1) is one of three herpesviruses that affect the esophagus, the others being CMV and varicella-zoster virus (see later). HSV-2 rarely involves the esophagus. After *Candida* species, HSV-1 is the next most frequent agent that causes infectious esophagitis. Although well recognized as an esophageal pathogen in otherwise healthy people, HSV-1 esophagitis has been reported most often in patients with immunosuppression or other predisposing factors. After transplantation, HSV occurs as frequently as CMV as a cause of esophageal disease, [40] whereas in patients with AIDS, HSV esophagitis is relatively uncommon and is much less frequent than CMV infection. [41] In one study of 100 HIV-infected patients with esophageal ulcer, HSV was found in only nine patients, in four of whom it was a co-pathogen with CMV. [41]

Pathology HSV infection is generally limited to squamous mucosa, where the earliest manifestation is a vesicle. As these vesicles enlarge and ulcerate, they may coalesce to form larger lesions. Usually, the intervening mucosa between these lesions is normal. Microscopic examination of the squamous epithelial cells at the edge of the ulcers reveals multinucleation, ground-glass nuclei, and eosinophilic Cowdry type A inclusion bodies that may take up half of the nuclear volume. These inclusion bodies may be surrounded by haloes, and with time, become more basophilic, filling, enlarging, and deforming the nucleus.

Clinical Manifestations and Complications HSV-1 esophageal infection commonly presents with the sudden onset of severe odynophagia, often resulting in an inability to swallow liquids or solids. Herpes labialis (i.e., cold sores) and oropharyngeal ulcers may frequently coexist, antedate, or develop during the esophageal infection, whereas skin infection is rare. [40] In untreated immunocompetent persons, spontaneous resolution of HSV-1 esophageal infection occurs 1 to 2 weeks after the onset of symptoms. Complications are rare and include bleeding, tracheoesophageal fistula, [42] or dissemination. [43]

Diagnosis Esophageal disease caused by HSV-1 usually appears in radiographic studies as focal ulceration on a background of normal mucosa; vesicles are infrequently present (Fig. 61-2). These ulcers have been described as stellate or volcanic in appearance, with less propensity to form the longitudinal or linear lesions that are commonly seen in CMV infection. [44] Severe, diffuse herpetic esophagitis may result in a cobblestone or “shaggy” mucosal appearance resembling *Candida* esophagitis. [44] Although the radiographic appearance may be suggestive, definitive diagnosis of herpetic esophagitis requires endoscopic mucosal biopsies. The endoscopic appearance of herpetic esophagitis reflects the pathological changes (see Table 61-1) appearing as discrete, usually small (<2 cm) well-circumscribed ulcers, [45] diffuse erosive esophagitis, or, rarely, vesicles. Small scattered lesions covered with exudates can mimic esophageal candidiasis. Given the pathophysiological mechanism of disease, deep ulcers, as seen with CMV, are very rare. Cytologic or histological brushings or biopsies should be taken from the edge of an ulcer because the viral cytopathic effect is best identified here, rather than the granulation tissue in the ulcer bed. Immunohistochemistry on biopsy samples using specific monoclonal antibodies to HSV will help to confirm the diagnosis when the viral cytopathic effect is infrequent. Viral culture may also help to establish a definitive diagnosis. As with other causes of infectious esophagitis, serologic tests are unhelpful in establishing the diagnosis.

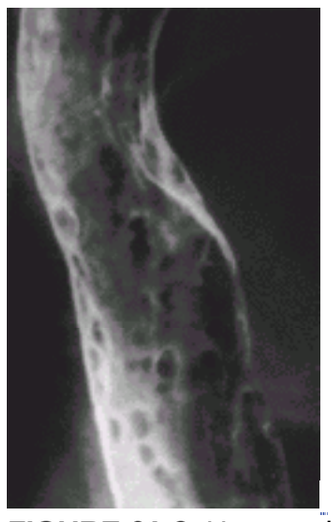

FIGURE 61-2. Herpes simplex virus esophagitis. Multiple vesicles and ulcers are present throughout the esophagus. (Courtesy of Robert Koehler, MD.)

Treatment Uncontrolled trials and vast clinical experience in both immunocompetent and immunodeficient patients suggest efficacy of acyclovir, a nucleoside analog, for the therapy of esophageal disease. In the largest study evaluating 34 patients with AIDS and HSV esophagitis, a clinical response was seen in essentially all treated patients. [46] Although spontaneous resolution of HSV-1 esophagitis is common in the immunologically normal host, because of its safety and efficacy, acyclovir therapy is commonly instituted in all patients regardless of immune status. When oral intake is hampered by severe odynophagia or when there is a question of drug absorption, intravenous administration is required. Side effects of intravenous acyclovir therapy are few and appear limited to irritation of veins used for drug infusion and rash. Although rare, drug resistance should suspected when there is clinical failure of acyclovir; in this setting, foscarnet (see Table 61-3) is the drug of choice and will lead to clinical cure in most patients. [47] Acyclovir is effective prophylaxis for HSV-antibody positive patients undergoing transplantation. Long-term secondary prophylaxis may be required when immunodeficiency persists because the relapse rate is high. More recently, valacyclovir, a prodrug of acyclovir, and famciclovir were released. Large studies of valacyclovir use in patients with genital HSV disease have shown equivalency to acyclovir and, as with acyclovir, minimal toxicity. [48] The advantage of this agent is that it can be administered three times per day, and the cost is equivalent to that of acyclovir.

Cytomegalovirus

Epidemiology Over the last several decades, CMV has become one of the most common opportunistic infections. Serologic studies from developed countries have shown seropositivity rates of 50% or greater; studies of homosexual men have found seropositivity up to 90% reflecting sexual transmission of the virus. [49] In transplant recipients who receive no antiviral prophylaxis, CMV and HSV are equally common esophageal pathogens. [40] In contrast, CMV is the most frequent cause of esophageal ulcer in patients with AIDS. [41]

Pathology The most prominent histological feature of CMV esophagitis is mucosal ulceration (see Table 61-1). Although there is variability, deep ulcers are very characteristic of disease in AIDS, whereas in immunosuppressed transplant recipients, lesions tend to remain more superficial. In contrast to HSV, the viral cytopathic effect of CMV is present in endothelial and mesenchymal cells in the granulation tissue of the ulcer base and not in the squamous epithelium. Inclusions are large (*cytomegalo*) and often have an eosinophilic appearance that may be present in the nucleus and cytoplasm (Fig. 61-3A; see also Color Fig. 61-3.). Because these inclusions may be atypical in appearance in patients with AIDS, [50] immunohistochemical stains are useful in confirming the presence of CMV and often highlight more infected cells than can be appreciated on routine hematoxylin and eosin staining. As with other esophageal infections, CMV may coexist with HSV or *Candida,* especially in patients with AIDS. [10] Whereas HSV leads to disease by a direct cytopathic effect on squamous epithelium, the pathogenesis of disease caused by CMV is not well understood. It has been speculated that CMV mucosal disease is the result of ischemia caused by endothelial cell involvement. More recently, inflammatory cytokines, such as tumor necrosis factor-a, have been suggested as playing an etiologic role. [51]

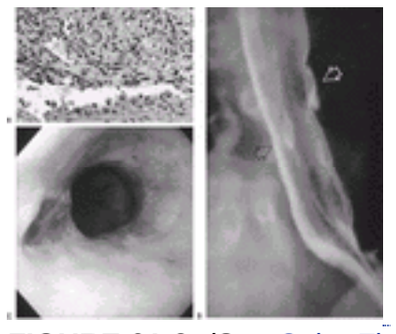

FIGURE 61-3. (See Color Fig. 61-3.) Cytomegalovirus esophagitis. (**A**) Biopsies from an ulcer base demonstrate granulation tissue with one large cell with intranuclear and intracytoplasmic inclusions diagnostic for cytomegalovirus. (**B**) Barium swallow demonstrates two well-circumscribed ulcers in the distal esophagus (*arrows*). (**C**) Endoscopic photograph shows the two ulcers, which are well circumscribed and have some depth, and the surrounding mucosa is normal.

Clinical Manifestations and Complications In contrast to HSV esophagitis, CMV esophagitis has very rarely been documented in immunocompetent persons. [52] Odynophagia is almost uniformly present and is typically severe (see Table 61-2). A prior or coexistent diagnosis of CMV infection in another organs (e.g., retinitis or colitis) is not infrequent. Although rare in transplant recipients, retinitis may be observed in approximately 15% of patients with AIDS at the time of diagnosis of gastrointestinal disease. [53] Complications include gastrointestinal bleeding (~5% of patients) and, rarely, strictures [54] or fistulae to the tracheobronchial tree. [55]

Diagnosis As with HSV-1, the radiologic appearance of CMV esophagitis is that of either focal or extensive ulceration. The radiographic characteristics of the ulcer depend in large part on the epidemiologic setting. Barium esophagraphy of CMV esophagitis may reveal thickening of mucosal folds or more typically ulcers; these ulcers may be vertical, linear with central umbilication, solitary, and deep, or occasionally they may manifest as diffuse superficial ulceration (see Fig. 61-3B). [44] In patients with AIDS, these ulcers are often large and deep and may exceed 2 cm in size. Rarely, the exuberant inflammatory response mimics a malignant disease. [56] The endoscopic appearance of CMV esophagitis is variable, ranging from multiple shallow ulcers, solitary giant ulcers, to diffuse superficial esophagitis (see Fig. 61-3C; see also Color Fig. 61-3). [57] Given the high rate of prior exposure to CMV, serologic testing is not helpful. In addition, some immunosuppressed transplant recipients fail to develop a brisk antibody response. Currently, identification of viral cytopathic effect in mucosal biopsies is the best diagnostic method. Multiple biopsies (up to ten) may be required to establish the diagnosis in patients with AIDS and should be taken from the ulcer base because the viral cytopathic effect is found here. [58] Viral culture of mucosal biopsies is less sensitive and specific than histology. [59] The identification of an isolated inclusion found by immunohistochemical staining should not be considered CMV disease but rather infection, and other causes of mucosal disease should be sought. [60] In contrast to *Candida* and HSV infections, cytologic specimens from esophageal lesion have very poor sensitivity. Because retinitis may coexist with gastrointestinal disease, and, when present, alters the duration of antiviral therapy, a diagnosis of CMV gastrointestinal disease in any patient with AIDS warrants ophthalmologic examination.

Treatment The therapies available for the treatment of CMV disease require intravenous administration and include ganciclovir, foscarnet, and, more recently, cidofovir. The most widely used therapy, ganciclovir, is an acyclovir derivative. A large prospective open-label trial of ganciclovir in 35 patients with AIDS documented clinical and endoscopic improvement in 77%. [53] The time course of the clinical response to ganciclovir is variable; a week of therapy may be required before there is symptomatic improvement. The total treatment duration should be based on the clinical and endoscopic response, and a 2- to 4-week treatment course is usually adequate. If retinitis is absent and there has been a complete response, the patient may be followed closely without maintenance therapy. Because of low bioavailability (<10%), oral ganciclovir is not effective for the treatment of active infections including those of the gastrointestinal tract. [61] After acute CMV disease in transplant recipients, treatment with ganciclovir should be given for approximately 1 to 2 months until the immunosuppressive regimen is either discontinued (bone marrow transplant) or significantly reduced (solid organ transplant). Ganciclovir is well tolerated, with its major side effect being myelosuppression, which may be severe when other bone marrow suppressive drugs, such as zidovudine (formerly known as azidothymidine or AZT), are coadministered. Clinical and virologic resistance has been documented, usually in patients receiving prolonged therapy. In this setting, foscarnet is often effective. [62] Ganciclovir has been used in conjunction with foscarnet to treat refractory disease or when side effects are limiting, because lower doses may be employed. [63] Foscarnet is a pyrophosphate analog that inhibits viral DNA polymerase and reverse transcriptase. A randomized trial comparing ganciclovir with foscarnet for patients with AIDS and gastrointestinal disease found clinical improvement in more than 80%, and there was no significant difference in efficacy between the two agents. [64] There are fewer data on its use in immunosuppressed patients who do not have AIDS. Foscarnet has been most frequently used when there is clinical resistance or intolerance to ganciclovir. The major side effect of foscarnet is reversible renal insufficiency. [65] This may be prevented by vigorous saline hydration before and during drug administration in combination

with dose adjustments based on creatinine clearance. Electrolyte disturbances, which include hypocalcemia and hypophosphatemia, are also common during or shortly after infusion, and slowing the rate of infusion may alleviate the mild cramps induces by these electrolyte shifts. All drugs for herpesviruses only inhibit viral replication; thus, relapse is frequent when therapy is discontinued. The relapse rate for transplant recipients also remains high until immunosuppressive therapy is reduced. Cidofovir is the newest systemic agent available for the therapy of CMV, but it has undergone evaluation only for the treatment of retinitis in AIDS. [66] Because of its long half-life, once-weekly administration is adequate. Like foscarnet, this drug is associated with renal insufficiency. In patients with AIDS, the relapse rate of CMV esophagitis is approximately 50% and is similar for HSV. [46], [53], [64] Initiation of HAART, when associated with significant improvement in CD4 count, may abrogate the need for long-term maintenance therapy. [67]

Prophylaxis High-dose acyclovir has been used with moderate success for the prophylaxis of CMV infection in transplant recipients, although ganciclovir has been shown to be superior. [68] Because of its cost, potential side effects, and intravenous route of administration, at many transplant centers, ganciclovir prophylaxis is limited to high-risk patients including CMV-seropositive patients, CMV-seronegative transplant recipients who receive CMV-seropositive organs or blood products, and patients receiving potent immunosuppression for episodes of transplant rejection. Oral ganciclovir is effective prophylaxis for CMV retinitis in patients with AIDS and a CD4 lymphocyte count less than 200/mm [3] but is untested for either primary or secondary prophylaxis of gastrointestinal CMV disease. There are also no data regarding the efficacy of oral ganciclovir prophylaxis in high-risk transplant recipients. Fortunately, despite long-term administration, resistance of CMV to ganciclovir and foscarnet is uncommon.

Other Viruses

The frequency of esophageal involvement caused by varicella-zoster during the course of chickenpox or herpes zoster infections is unknown but clinically uncommon. [69] Papillomavirus may infect the esophagus and may cause small polypoid lesions. [70] Esophageal ulcers have been reportedly caused by papovavirus and Epstein-Barr virus in patients with AIDS. [71], [72]

MYCOBACTERIAL INFECTIONS

Epidemiology

Mycobacterial involvement of the esophagus is very rare, even in immunosuppressed patients. Previously, tuberculous involvement of the esophagus was considered a rare autopsy finding seen in less than 0.15% of necropsies. [73] In developing countries, the rate of tuberculosis (TB) is much higher, and extrapulmonary manifestations, including esophageal disease, are more common. The combination of the AIDS epidemic with the upsurge in reported cases of systemic TB have increased the incidence of esophageal infection in developed countries. *Mycobacterium avium* complex (MAC) remains primarily a small bowel pathogen with only rare cases of esophageal involvement reported. [74], [75]

Pathology

Most commonly, TB affects the middle third of the esophagus at the level of the carina. Esophageal disease is caused by spread of infection from tuberculous-infected mediastinal lymph nodes by way of a draining fistula or obstructed lymphatics, often resulting in tracheoesophageal fistula. TB can also involve the upper third of the esophagus by direct extension from tuberculous pharyngitis or laryngitis. Primary esophageal TB in the absence of extraesophageal disease is exceedingly rare. [76] Granulomata are often present in ulcer tissue, with bacilli identifiable by mycobacterial staining.

Clinical Manifestations and Complications

The symptoms of esophageal TB depend on the degree and type of involvement. Pulmonary complaints often predominate because of the common occurrence of fistula to the trachea, bronchus, or pleural space. Rarely, formation of long strictures or traction diverticula resulting from the fibrotic response causes dysphagia. Upper gastrointestinal hemorrhage from tuberculous esophageal ulcers [77] and tuberculous arterioesophageal fistulae [78] has been reported.

Diagnosis

Esophageal TB should be suspected in patients with pulmonary or systemic TB who develop esophageal symptoms. Barium esophagram findings, including ulceration and stricture, are nonspecific. A sinus tract or fistulous connection to the bronchial tree or mediastinum is highly suggestive of TB but can also be seen with malignant disease or other infections; in this setting, the diagnosis may often be made by sputum staining and culture. [73], [75] An ulcerated tuberculous granulomatous mass may suggest an esophageal neoplasm on barium esophagraphy. [79] Although the gross appearance of ulcers or strictures is not diagnostic of TB, endoscopic biopsies from the edge of the lesions may reveal granulomata or acid-fast bacilli, and biopsy material may be cultured for further confirmation of the diagnosis and determination of sensitivities to antimycobacterial agents.

Treatment

Regardless of the presence of immunodeficiency, a 9-month course of multidrug therapy (in the absence of drug resistance) will cure esophageal TB and will often close fistulae. If fistulae do not close with medical therapy, surgical intervention will be required. The prevalence of multidrug-resistant TB is becoming an increasingly complex problem; thus, drug sensitivities to antituberculous therapy are essential to guide therapy. The most effective agents for the treatment of MAC are clarithromycin and ethambutol. [80] Although a clinical and bacteriologic response is common, long-term therapy for MAC is required in AIDS unless HAART is effective. [81]

BACTERIAL INFECTIONS

Epidemiology

Bacterial esophagitis is a rare cause of esophageal disease in immunocompromised patients. It has been described almost exclusively in patients with hematologic malignant disease complicated by severe granulocytopenia and occasionally after bone marrow transplantation and diabetic ketoacidosis. [82], [83], [84] and [85] For the most part, the infecting pathogens are oral flora, particularly gram-positive organisms, including viridans streptococci, staphylococci, and other bacilli. It is likely that these pathogens colonize and then invade mucosa damaged by reflux disease, radiation, or chemotherapy leading to local infection; dissemination may occur when granulocyte function is poor or there is absolute granulocytopenia. Reports in patients with AIDS have broadened the etiologic spectrum to include *Bartonella henselae,* the cause of catscratch disease, [86] actinomycoses, [87], [88] and *Nocardia*. [89]

Pathology

The gross pathological appearance of the esophagus in bacterial infection ranges from normal mucosa (colonization) to ulcers associated with erythema, plaques, pseudomembranes, or hemorrhage. [82] Microscopic examination reveals pseudomembranes and bacterial invasion that may be superficial and invade only the squamous epithelium or may be invasive and transmural with infiltration of blood vessels (i.e., phlegmonous esophagitis). Actinomycosis is characterized by ulcerative esophagitis and drainage of sulfur granules from sinuses leading from abscess cavities or with sulfur granules and filamentous gram-positive branching bacteria seen on tissue biopsies. [87] *Nocardia* has been reported to cause esophageal ulcer in AIDS. [89] *Bartonella henselae* esophagitis causes multiple nodules of the esophagus resulting from a lobulated proliferation of capillary vessels lined by plump endothelial cells. [86]

Clinical Manifestations and Complications

Bacterial esophagitis is usually found in a neutropenic patient with esophageal complaints who has undergone chemotherapy for a hematogenous malignant disease. Esophageal infection may serve as a focus for bacteremia and seeding of other organs. [82] No unique complications have been reported.

Diagnosis

The diagnosis of bacterial esophagitis should be considered in the clinical setting described earlier. Radiographic findings are nonspecific, and endoscopic biopsy and culture are required to establish this diagnosis. Additional stains including Gram stain are required to identify the etiologic bacteria. Positive blood cultures also pinpoint the bacterial pathogens and direct antimicrobial therapy.

Treatment

Broad-spectrum antibiotics that effectively treat both gram-positive and gram-negative oropharyngeal flora are required for treatment. Treatment of other bacterial infection found in these patients is similar to treatment of disease in other locations.

Treponema pallidum

Although esophageal involvement by *Treponema pallidum* was well recognized many years ago, this disease is now only of historical interest. Tertiary syphilis of the esophagus may present as a submucosal gumma or diffuse inflammatory reaction with fibrosis that often affects the upper third of the esophagus, and it may be associated with mucosal ulcers and structures. [90] Given the rarity of esophageal syphilis, most patients with infectious esophagitis and positive serologic tests for syphilis have another cause of esophagitis.

PROTOZOAL INFECTIONS

In developed countries, protozoal infections of the esophagus are very rare, occurring almost exclusively in patients with AIDS. In these patients, pathogens include *Pneumocystis carinii,* [91] *Cryptosporidium parvum,* [92] *Leishmania donovani,* [93] and *Trichomonas.* [94] In immunologically normal hosts from endemic areas in South America, *Trypanosoma cruzi* may involve the myenteric plexus of the esophagus resulting in Chagas disease. This disease is indistinguishable clinically, radiographically, endoscopically, and manometrically from idiopathic achalasia. The diagnosis may be established by antibody testing.

SPECIFIC HIV-RELATED ESOPHAGEAL DISORDERS

In addition to the infections described earlier, certain unique disorders cause esophageal disease in these patients.

Disorders Associated with Primary HIV Infection

Although primary HIV infection is largely asymptomatic, in some patients, a mononucleosis-like illness occurs around the time of infection consisting of fever, sore throat, and myalgias associated with a maculopapular rash. [95] Spontaneously resolving oropharyngeal and esophageal ulceration or candidal infection may also be observed during this seroconversion illness. [96], [97] Endoscopically, these esophageal ulcerations are multiple, small, and shallow. [96] In some of these patients, electron microscopic examination of biopsy specimens revealed enveloped viruslike particles with morphologic features compatible with retroviruses. [96] The diagnosis can be established at the time of presentation by the detection of HIV RNA in serum. [95] Antibody positivity to HIV occurs within 3 to 18 months after the illness.

Idiopathic Esophageal Ulcer

Epidemiology Early in the AIDS epidemic, large esophageal ulcerations were recognized in which no specific cause could be identified despite extensive histopathological examination of ulcer tissue. These ulcers are now termed *idiopathic esophageal ulcers* (IEU) or aphthous ulcers. These ulcers are very common and were found in 41% of HIV-infected patients with esophageal ulcer in a large prospective study. [41] They are seen in the later stages of immunodeficiency when the CD4 lymphocyte count is less than 100/mm [3]. [41]

Pathology These ulcers are variable in size, may be large, and are uniformly well circumscribed; diffuse superficial esophagitis has not been described. [98] Ulcer tissue resembles that seen in CMV and HSV infection, except viral cytopathic effect is absent. The presence of a superficial candidal infection overlying a large well-circumscribed lesion with histopathological findings of granulation tissue without viral cytopathic effect should still lead to the diagnosis of IEU. [10] Although HIV has been suggested as the direct cause of these lesions, there is little histopathological evidence to support a direct cytopathic role for HIV. [99], [100]

Clinical Manifestations and Complications IEU presents in a fashion indistinguishable from CMV esophagitis. Coexistent oropharyngeal aphthous ulcers are infrequent, [13] whereas thrush is common, especially if the patient has not been given empiric antifungal therapy. Complications include bleeding and fistula to the stomach but not to the tracheobronchial tree, [101] and strictures. [54]

Diagnosis The findings of IEU on barium esophagram are typically large, well-circumscribed, and often deep ulcers. [102] Because of the similarity to CMV infection, a definitive diagnosis cannot be based on the radiographic appearance alone. Because IEU is a diagnosis of exclusion, endoscopy and biopsy are the only definitive diagnostic tests. These ulcers are variable in size and appearance, and larger ulcers are endoscopically indistinguishable from CMV infection. [98] Distal esophageal ulcer may suggest gastroesophageal reflux disease; the histopathological features alone also cannot distinguish IEU from gastroesophageal reflux disease. Pill-induced esophagitis must be excluded by history because the pathological findings of esophageal biopsies are similar. These ulcers respond rapidly to either prednisone or thalidomide, with clinical and endoscopic cure seen in more than 90%. [103], [104]

Neoplasms Associated with AIDS

Kaposi Sarcoma With the advent of HAART, Kaposi sarcoma (KS) has been decreasing in frequency. Gastrointestinal KS is common in those with cutaneous disease. [105] Clinical experience suggests that gastric or duodenal involvement is more common than esophageal disease, and esophageal lesions, like other areas of the gastrointestinal tract, are most frequently an incidental finding. Findings on barium esophagram of KS have not been clearly characterized are well-circumscribed submucosal lesions that may ulcerate. [106] Small lesions are easily missed on barium studies. The endoscopic features of esophageal KS are characteristic and resemble their cutaneous appearance: violaceous macular or plaquelike lesions. These tumors typically involve the submucosa; therefore, mucosal biopsy must sample deeper tissue. Bleeding may occur when the tumor becomes large and ulcerated. Effective relatively nontoxic chemotherapy includes Doxil, a liposomal form of doxorubicin (Adriamycin). HAART therapy alone has been associated with lesion regression. Radiation therapy is effective to treat local lesions of the head and neck including the oropharynx.

Non-Hodgkin Lymphoma Unlike with other opportunistic processes, the advent of HAART has not altered the incidence of lymphoma in AIDS. Although gastrointestinal involvement is common, esophageal disease remains rare. Extraesophageal disease is common at the time of diagnosis. [107] In two studies comprising 23 patients with AIDS and gastrointestinal lymphoma, only 2 patients had esophageal involvement. [107], [108] The lesions appear radiographically as large ulcers or mass lesions typical of any carcinoma. [109] The endoscopic appearance of these lesions has been described as an ulcerated polypoid mass, often with a central ulceration, a submucosal lesion or extensive disease resulting in lumenal narrowing resembling an adenocarcinoma, [110] or a solitary ulcer. [111] Complications are rare and include bleeding. [109] Multidrug chemotherapeutic regimens are usually given for non-Hodgkin lymphoma in AIDS, and complete remission may be realized in approximately 50% of patients. Complications are frequent because of chemotherapy-induced neutropenia. The median survival in most series of AIDS-associated non-Hodgkin lymphoma is less than 8 months. Radiation therapy may be a potential option when the disease is localized.

Miscellaneous Neoplasms Despite the profound immunodeficiency seen in these patients, there does not appear to be an increase in the incidence of other esophageal neoplasms including squamous cell cancer and adenocarcinoma. Adenocarcinoma of the esophagus associated with Barrett esophagus has been reported in an HIV-infected patient. [112] Squamous cell esophageal carcinoma is uncommon in HIV-infected patients, and it has been reported in two patients with AIDS, [113] probably because of the relative young age of these patients.

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CHAPTER 62

Weijing Sun and Anil K. Rustgi

ESOPHAGEAL NEOPLASMS

SQUAMOUS CELL CARCINOMA

Epidemiology and Etiology

Esophageal *squamous cell carcinoma* (SCC) is the most common form of esophageal malignancy worldwide and is one of the leading causes of cancer mortality in men, especially African American men. The incidence of esophageal SCC varies according to geographic location, with a global range from 2.5 to 5.0 for men and 1.5 to 2.5 for women for 100,000 population. High-incidence regions, where rates may exceed 100 per 100,000 population, include northern China, India, northern Iran, areas north and east of the Caspian Sea, and the Transkei area of South Africa. [1], [2], [3], [4] and [5] Indeed, nearly 50% of all cancers are esophageal in southeastern South Africa. [1] In the United States, African Americans have a four- to fivefold increased risk compared whites, especially African American men, in whom the risk is 15.1 per 100,000 compared with 2.9 per 100,000 in white men. [6], [7]

The geographic variation in esophageal SCC strongly hints at the contribution of environmental factors, although its genetic basis is now being elucidated. Genetic predisposition to esophageal SCC is rare. The incidence of esophageal SCC is very low in persons less than 40 years of age, but it increases with each succeeding decade of life. [7] It is more common in men than in women, a two to three times increased risk, regardless of ethnicity and age. [7] However, there are some exceptions to this general finding. In northern Iran, the rate is 263 per 100,000 for women and 206 per 100,000 for men, [8] and upper esophageal cancers in association with the Plummer-Vinson syndrome are found in women.

Tobacco and Alcohol Together, tobacco and alcohol constitute the largest risk factors for the development of esophageal SCC in North America and Western Europe. [9], [10], [11] and [12] Because tobacco and alcohol are commonly consumed together, their individual contribution may be difficult to ascertain. However, a comprehensive study of alcohol consumption in nonsmokers and tobacco use in nondrinkers found that each acts independently as risk factors. [13] The relative risk was 2.0 for those who smoked fewer than 15 cigarettes per day, and 6.2 for patients who smoked more than 25 cigarettes per day. [13] Not surprisingly, there is a documented substantial increase in risk with patients who consume both tobacco and alcohol, with the amount and duration of use being pivotal factors. [14], [15], [16], [17] and [18] Thus, smokers who have a large intake of beer and whiskey harbor a ten- to 25-fold increased risk in esophageal SCC compared with smokers who do not drink alcohol. [18], [19] Additionally, the risk may be greater for pipe and cigar smokers than for cigarette smokers. [18] In a manner reminiscent of lung cancer, cessation of smoking does lead to a decreased risk of esophageal SCC after 10 years. [18], [20], [21] Although cigarette smoking is a key risk factor in North America and Western Europe, chewing tobacco, betel, or combinations of these with lime accounts for the high incidence of esophageal SCC in southern and Southeast Asia. [22], [23] and [24] Tobacco tars and cigarette smoke contain various chemical carcinogens, such as aromatic amines, lactones, peroxy compounds, halo ethers, *N*-nitroso compounds, polycyclic aromatic hydrocarbons, and peroxy compounds, [25], [26], [27] and [28] that individually or in combination likely lead to esophageal epithelial hyperproliferation and eventual malignant transformation. The type of alcohol (beer, whiskey) and the fashion in which it is distilled, but not the duration of ingestion, are risk factors for esophageal cancer. Groups of people who do not consume alcohol have a substantially decreased risk. There are several reasons underlying alcohol's contribution to the pathogenesis of esophageal cancer, as well as other cancers. [29] Alcohol may contain congeners or other ingredients, such as *N*-nitroso compounds, urethane, mycotoxins, tannins, pesticide residues, and asbestos products that are directly or indirectly carcinogenic. Ethanol itself, because it is a solvent for fat-soluble compounds, may facilitate the absorption of other carcinogens. Carcinogens that are oxidants may increase the risk of DNA damage. Acetaldehyde, a metabolite of

ethanol, can inhibit DNA methyltransferase activity. [30] Consequently, a state of methyl deficiency may ensue that may potentiate the carcinogenic effect of a methyl-deficient diet.

Diet and Nutrition Vitamins and deficiencies, especially of vitamins A and C, folic acid, vitamins E and B_{12}, and riboflavin, are crucial as risk factors. [31], [32], [33], [34] and [35] Diets containing high amounts of green and yellow vegetables that are rich in ß-carotene and citrus fruits that are high in vitamin C decrease the incidence of esophageal SCC. [35] Lower plasma levels of folic acid have been documented in patients with esophageal SCC. [33] Riboflavin deficiency manifested by cheilosis and glossitis is common in the Linxian Province of China, where esophageal cancer is endemic. [36] It is likely that vitamins A, C, and E exert antioxidant effects; the latter two vitamins are known to influence the formation of nitrosamines. The levels of certain trace elements, such as selenium, molybdenum, and zinc, show an inverse association with mortality from esophageal cancer in high-incidence areas in the world. [37], [38] and [39] Furthermore, in animal models, a significant increase in esophageal tumors is induced by *N*-nitrosomethylbenzylamine (NMBA) in rats with zinc-deficient diets. Mechanistically, zinc deficiency enhances the microsomal activation of NMBA to a methylating agent that yields *O*-6-methylguanine adduct in esophageal DNA. [40] As a component of superoxide dismutase, zinc also protects against oxygen-derived radical damage and free-radical formation. [33] Similarly, selenium has protective effects, in part mediated through inhibition of lipid peroxidation of cell membranes through the action of glutathione peroxidase, which is dependent on selenium. [32] The Linxian and adjoining provinces in China have foodstuffs low in molybdenum and documented reduced levels of this trace element in patients' serum and urine. It is conceivable that molybdenum deficiency leads to the accumulation of nitrates that are precursors of nitrosamines because this trace element is a key constituent of nitrate reductase. [24]

Achalasia Patients with long-standing achalasia have a prevalence of esophageal SCC of 5%. [41], [42], [43], [44], [45] and [46] The interval from dysphagia, weight loss, and chest pain ascribable to achalasia to the development of cancer is approximately 17 to 20 years. [46], [47] In a study of 195 consecutive patients with achalasia, the incidence of esophageal SCC was 3.4 per 1000 patients per year, representing a 33-fold increase when compared with age- and sex-matched controls. [48] A follow-up study of 146 of 147 patients with achalasia treated with myotomy revealed that 10 patients progressed to esophageal cancer, with a mean interval of 16.7 years; the predicted frequency was less than 1. [49] Thus, the condition of achalasia eventually may lead to cancer; however, the exact risk and mechanism remain to be determined. Although endoscopic surveillance of patients with achalasia has been advocated, the frequency of examinations remains the subject of debate.

Head and Neck Squamous Cell Carcinoma Reflecting use of both alcohol and tobacco, patients with head and neck SCC are at an increased risk of developing esophageal cancer. [50], [51] Either coexisting or sequential esophageal cancers may develop annually at a rate of 3% to 7%. [52], [53], [54] and [55] As a result, endoscopic surveillance of the esophagus has been advocated in this setting. Toluidine blue staining has been demonstrated to enhance the detection of indolent esophageal cancers in patients with head and neck cancers. [56] Not surprisingly, survival of patients with head and neck cancer is adversely affected by the coexistence of esophageal cancer. [57] A reduction in second primary tumors from 24% to 4% in patients with head and neck cancers has been observed with the use of isotretinoin, a chemopreventive agent that induces epithelial differentiation. [58]

Tylosis Although some studies from China have suggested familial aggregation of esophageal cancers, [59] it is difficult to discriminate between a common environmental exposure and an actual genetic basis for these observations. However, a genetic predisposition to esophageal cancer is clear in tylosis palmaris, an autosomal dominant disorder manifested by hyperkeratosis of palms and soles. [60], [61], [62], [63], [64], [65] and [66] Approximately 50% of patients with tylosis palmaris will develop esophageal cancer by the age of 45 years, and perhaps 95% by age 65 years. [61], [62] There is linkage between esophageal SCC and oropharyngeal leukoplakia. In endoscopic surveillance studies over a 5-year period, 4 of 29 patients were found to have dysplasia, and 1 patient had carcinoma in situ. [67] The gene responsible for tylosis has yet to be identified, but once achieved, it may provide insights into sporadic esophageal cancer as well.

Other Factors Lye ingestion, which results in stricture formation, has been postulated as a risk factor for esophageal cancer, mostly appearing at level of the bronchial bifurcation. [68], [69] and [70] Lye strictures constitute only a small proportion of esophageal cancer cases, and the progression to cancer after initial exposure requires 4 to 5 decades. Other factors implicated in the pathogenesis of esophageal cancer include ionizing radiation, [71], [72] celiac sprue, [73], [74] and [75] human papillomavirus (HPV), [76], [77] and [78] Plummer-Vinson syndrome, [79] esophageal diverticula, [80] and maté drinking. [81], [82] Experimentally, induction of radiation injury is associated with cancer formation. [83] In patients who have undergone high-dose radiation treatments, esophageal cancer is the most frequently observed malignant disease. [84], [85], [86], [87], [88], [89] and [90] DNA tumor viruses that have a tropism for squamous epithelial cells, such as HPV and Epstein-Barr virus, have been associated with esophageal SCC. Polymerase chain reaction techniques have demonstrated an association of the oncogenic variants of HPV, namely HPV-16 and HPV-18, as well as a novel HPV genotype with esophageal SCC. [77], [78] The same has been observed with Epstein-Barr virus. [91] Both viruses infect squamous epithelial cells, replicate, and produce oncogenic proteins that contribute to hyperproliferation and malignant transformation. Hot maté drinking is common in Uruguay and southern Brazil, [81], [82] and hot liquid ingestion is prevalent in China. [92] These findings suggest that thermal injury of the mucosa may increase the risk of esophageal cancer.

Biology and Genetics

Based on the colon cancer paradigm, it is thought that the development of cancer results from the accumulation of alterations in oncogenes, tumor suppressor genes, and DNA mismatch repair genes. This is probably true for esophageal cancer as well. However, a genetic basis for the pathogenesis of esophageal cancer by no means precludes the importance of environmental factors; rather, more than likely, there is cooperativity among these factors. For example, some of the environmental factors injure the esophageal epithelium, which, in turn, may act to signal changes in key genes through point mutations or augmented gene expression.

Perhaps the most critical oncogene in esophageal SCC pathogenesis is cyclin D_1. Mammalian cyclins represent a large family of cell cycle regulatory proteins that associate, in turn, with their catalytic subunit partners, designated cyclin-dependent kinases or CDK. (The cell cycle is reviewed in the literature. [93]) The activity of cyclin D_1 is restricted to the G_1 phase, and it preferentially associates with cyclin-dependent kinase 4 or 6 (CDK4 or CDK6). The complex of cyclin D_1 and CDK4 or CDK6 then phosphorylates the retinoblastoma tumor suppressor gene product (pRb). This relieves the negative regulation of pRb on the G_1 phase through release of key transcriptional factors, such as E_2F. It has been shown that cyclin D_1 is overexpressed in more than 50% of esophageal SCCs and is associated with a poor prognosis. [94], [95] and [96] A subset of esophageal cancers also show c- *myc* and epidermal growth factor receptor overexpression as a result of gene amplification. [97], [98] Knowledge of the association of cyclin D_1 and esophageal cancer has been used to develop a transgenic mouse model in which one of the Epstein-Barr virus promoters has been used to target cyclin D_1 specifically to the oral-esophageal squamous epithelia, resulting in dysplasia and cell cycle abnormalities. [99], [100]

The key tumor suppressor gene *p53* has many functions, including cell cycle regulation, response to environmental insults to induce apoptosis, transcriptional regulation of genes, and DNA replication. *p53* is mutated in up to 70% of esophageal cancers. [101], [102], [103] and [104] One of the functional consequences of *p53* mutation is the transcriptional downregulation of *p21,* a general CDK inhibitor. In addition, *p16,* also referred to as *INK4a* (designated $p16^{INK4a}$), is an inhibitor of CDK4. Located on chromosome 9p22-23, *p16* can be deleted, mutated, or hypermethylated, resulting in loss of its inhibitory function in esophageal cancer. [105], [106] A paradigm that emerges in esophageal SCC is that either cyclin D_1 is overexpressed or *p16* is altered to accelerate the cell through the G_1 phase. Loss of heterozygosity in other tumor suppressor gene loci, such as pRb (chromosome 11) and APC (Adenomatous Polyposis Coli) (chromosome 5q), does not appear to occur in esophageal cancer, [107], [108] suggesting that other genes in proximity to pRb and APC are the key targets.

Clinical Manifestations

Early esophageal SCC may be asymptomatic or may be associated with only mild specific symptoms. [109], [110], [111], [112] and [113] As the cancer grows, dysphagia occurs in nearly 90% and odynophagia in 50% of patients. [114], [115] and [116] This is usually a reflection of partial or total lumenal obstruction. Anorexia and weight loss from malnourishment may be observed in about 75% of patients. [116], [117] Retrosternal pain or radiation of pain to the back suggests mediastinal involvement by the cancer. Nausea, vomiting, and hematemesis may occur. Cough may reflect aspiration pneumonia or, rarely, tracheoesophageal fistula. Recurrent laryngeal nerve involvement may result in hoarseness. Skeletal metastases may cause bone pain. Physical examination may reveal cachexia, lymphadenopathy, hepatomegaly (if distant metastatic disease is present), and the presence of fecal occult blood.

Evaluation of dysphagia is imperative and is discussed in Chapter 31. The differential diagnosis may include peptic acid esophagitis with stricture, caustic ingestion resulting in stricture, malignant disease (either squamous or adenocarcinoma), and motility disorders such as achalasia or scleroderma.

Natural History and Complications

Esophageal cancer in Western countries usually presents in an advanced stage, and consequently, nearly 75% of untreated patients succumb to the disease within 1 year. [118] Biologically, esophageal SCC is not a rapidly growing tumor, and there is a prolonged period of time between dysplasia and cancer. [119], [120] Early stages of esophageal cancer have been shown to persist for more than 2 decades in China and Iran. [121], [122] Additionally, balloon cytologic examinations, used to monitor the

natural history of esophageal squamous dysplasia in China, indicate that dysplasia may progress to cancer (26.6% of cases), but it may regress to mild dysplasia or a normal state (40.5% of cases). Furthermore, of the mild dysplastic cases, nearly 15% progressed to severe dysplasia and about 45% regressed. [123] Other studies in China indicate that early superficial cancers remain in this state in 58% of cases for a follow-up period of 19 to 78 months. [120] Patients with untreated early esophageal cancer in China have a median survival time of 75 months.

There are probably biologic differences superimposed on environmental factors in the high-incidence areas versus Western countries that may account for the apparently lengthy period from dysplasia to cancer in China and the good prognosis of resected early esophageal cancer with a 5-year survival approaching 90%. [112] Reminiscent of the high prevalence of early gastric cancer in Japan relative to Western countries, it is not entirely clear whether early esophageal cancer is prevalent in North America and Western Europe. Several nonbiologic explanations could be evoked: use and accuracy of balloon cytology in China, definition of early esophageal cancer, and proper staging with aggressive therapy. [124], [125]

The average survival of untreated patients with advanced, symptomatic cancer is approximately 9.5 months. [118], [122] Patients may develop aspiration pneumonia complications of esophageal cancer from extension into the tracheobronchial tree or other mediastinal structures. [126] Fistulae have a 4.5% prevalence and most often communicate with the trachea, the main stem bronchi, and the lung parenchyma. Fistulae carry a poor prognosis. [127], [128] The fistula may cause presenting symptoms, such as cough, chest pain, dyspnea, hoarseness, and hemoptysis, or it may occur within 2 months of initial diagnosis in untreated patients. [127], [129] As a result of fistula formation, patients may develop pneumonia and sepsis. Direct extension, as opposed to fistula formation, of the esophageal cancer may involve the lung and tracheobronchial tree, aorta, pericardium, and upper abdomen. Hoarseness suggests recurrent laryngeal nerve involvement. Apart from direct extension or fistula formation, esophageal cancer may metastasize to lymph nodes, lung, and liver as frequent sites. [130], [131], [132], [133] and [134]

Diagnostic Evaluation

Implicit in a diagnostic evaluation is a complete history and physical examination. A laboratory profile may reveal anemia, hypoalbuminemia, elevated prothrombin time reflecting vitamin K deficiency, and possibly elevated liver function tests if metastatic disease is present. Serologic markers of cancer, such as carbohydrate antigen 72.4 in gastric cancer, carbohydrate antigen 19-9 in pancreatic cancer, carcinoembryonic antigen in colorectal cancer, and a-fetoprotein in hepatocellular carcinoma, have not proven to be of high sensitivity and specificity in esophageal cancer. Evaluation is driven by establishment of tissue diagnosis through biopsy and cytology, followed by staging for prognosis and therapeutic planning. Initial diagnostic tests usually entail barium swallow supplemented with a tablet and flexible fiberoptic endoscopy. Staging requires computed tomographic (CT) scan, endoscopic ultrasound (EUS), and radionuclide bone scan. Bronchoscopy should be considered if tracheal invasion is suspected.

Fiberoptic Flexible Endoscopy and Barium Esophagography The diagnosis of esophageal cancer requires endoscopy with biopsy and cytology. Multiple biopsies increase the diagnostic yield up to 96%; cytology identifies the remaining cases. For example, in a retrospective study of 155 patients with endoscopic biopsy and cytology of upper gastrointestinal lesions, 2 of 28 patients with malignant disease had the diagnosis through cytology alone. [135] Because endoscopy may prove too costly in some regions of the world, barium esophagrams are advocated by some experts, especially in patients with dysphagia. Intrinsic advantages include reduction in cost, noninvasiveness, and reduced complication rate. Additionally, a barium study allows information about motility as well as challenge with a solid bolus. Ultimately, endoscopy is a better diagnostic test for patients in whom one suspects esophageal cancer because of the availability to obtain tissue in one test and a greater positive predictive value of upper endoscopy: 100% for endoscopy and 33% for barium swallow. [136] The endoscopic appearance of early esophageal cancer includes superficial erosive ulcer, a raised plaque, a “congestive” lesion marked by red spots on the mucosa, and a small polypoid lesion. Vital staining dyes such as toluidine blue, aniline blue, or Lugol iodine may be used to enhance detection and simultaneously to discriminate early cancers from normal adjacent mucosa. In a study of 178 patients who underwent endoscopy and vital staining, 9 esophageal cancers were found, of which 7 were early cancers. [137] In contrast, endoscopy without vital staining only detected 4 of 9 cancers. Limiting the usefulness of vital staining is its inability or failure to distinguish inflammation from dysplasia. [138] Advanced esophageal SCCs typically are exophytic, ulcerated, and even circumferential. Submucosal invasion may produce friable nodules or strictures. Whenever possible, the endoscope should be advanced through the stricture to determine the proximal-distal extent of tumor and any distal abnormalities. The preponderance of SCCs is found in the proximal esophagus to the midesophagus. Radiologically, early esophageal cancer may have a granular mucosal appearance with single or multiple small ulcerations with tiny filling defects. Polypoid lesions are characterized by small intralumenal filling defects affecting one wall of the esophagus with ulcerations. [139] An infiltrative or an ulcerative appearance can be one of the defining features of advanced esophageal cancer, but the most common presentation of advanced esophageal carcinoma is that of a polypoid mass within the lumen produced by an exophytic cancer. [140] The mass may have the characteristic appearance of an “apple-core” lesion with circumferential involvement by the cancer (Fig. 62-1). Infiltrating carcinomas may result in diminished esophageal motility with or without stricture. Such a stricture may be symmetric or asymmetric and, in fact, can sometimes be difficult to distinguish from a benign stricture. The primary ulcerative type of esophageal cancer is rare, although ulceration can be a component of either polypoid or infiltrating cancer. Ulcerative cancers are characterized by a well-demarcated meniscoid ulcer with a thin rim of lucency representing neoplastic tissue. [141]

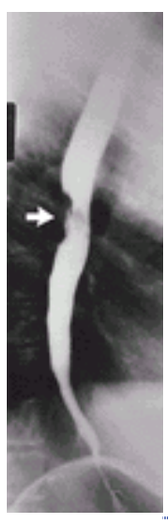

FIGURE 62-1. Advanced squamous cell carcinoma represented by an ulcerated mass in the midesophagus. (Courtesy of Deborah Hall, M.D.)

Screening High-incidence areas, such as in northern China, may merit endoscopic screening in the general population. In China, 5-year survival was nearly 90% after operative resection for cancer cases detected through screening. [142], [143] Surprisingly, nearly 80% of patients declined therapy because of noncompliance, unavailability of medical resources, and coexisting medical illnesses. [144] Although balloon cytology has achieved success in China, its applications in United States veterans did not prove very successful. [145] The reasons were the low prevalence of esophageal cancer and the low specificity of dysplasia because of a confounding factor such as inflammation resulting from reflux esophagitis. In this United States veterans study, follow-up measures included biannual endoscopy with vital staining and biopsy, balloon cytology, laryngoscopy, and chest radiography. [146] However, the results of cytology in more than 95% of screened patients were false-positive results. Thus, mass screening in the United States is probably not feasible for esophageal SCC, given the lack of cost effectiveness. However, institution of screening and surveillance endoscopic programs may be applied to certain clinical situations, such as patients with tylosis, achalasia, and head and neck cancer.

Staging and Staging Systems

After establishment of the diagnosis of cancer, staging is necessary to optimize treatment options and management. This should ideally commence with CT scanning of the chest and abdomen and should be supplemented with EUS for esophageal wall and local lymph node involvement.

The critical determinants of survival in nearly 4000 surgical specimens from Japanese patients were depth of invasion, the presence or absence of lymph node metastases, and the presence or absence of distant metastases in a retrospective study. [147] Five-year survival was 46% with submucosal invasion, 29.5% with invasion of the muscularis propria, 21.7% with invasion into the muscularis propria, and 7% with involvement of adjacent tissues and organs. [147] Worsening survival rates also correlate with involvement of distant lymph nodes and with metastasis to distant organs (3.0%). Although several staging systems have been recommended for esophageal cancer, an overriding consideration is that surgical pathological staging is more accurate for predicting survival than pretreatment clinical staging.

Thus, staging involves clinical evaluation before therapy and then pathological staging of the surgical resected esophagus and dissected lymph noes. A staging system that is widely used because of its accuracy is depicted in Table 62-1.[148]

Primary Tumor (T)			
TX	Primary tumor cannot be assessed		
T0	No evidence of primary tumor		
Tis	Carcinoma in situ		
T1	Tumor invades lamina propria or submucosa		
T2	Tumor invades muscularis propria		
T3	Tumor invades adventitia		
T4	Tumor invades adjacent structures		
Lymph Node (N)			
NX	Regional lymph nodes cannot be assessed		
N0	No regional lymph node metastasis		
N1	Regional lymph node metastasis		
Distant Metastasis (M)			
MX	Presence of distant metastasis cannot be assessed		
M0	No distant metastasis		
M1	Distant metastasis		
Stage Grouping			
0	Tis	N0	M0
I	T1	N0	M0
IIA	T2	N0	M0
	T3	N0	M0
IIB	T1	N1	M0
	T2	N1	M0
III	T3	N1	M0
	T4	N1	M0
IV	Any T	Any N	M1

TABLE 62-1 TNM Staging System for Cancer of the Esophagus (American Joint Committee on Cancer criteria)

Imaging Studies: Computed Tomography and Magnetic Resonance Imaging CT scanning has proven useful in the preoperative staging of esophageal cancer,[149],[150],[151],[152],[153],[154] and [155] especially in defining lymph node, pulmonary, and hepatic involvement. Accuracy of CT staging has a broad range, from 39% to 100%.[150],[151],[154],[156] Even documentation of aortic invasion, which precludes surgical resection, has a sensitivity of 80% to 100% and a specificity of 50% to 100% through CT scanning.[150],[151],[154] Tracheobronchial involvement is also discernible with CT scanning.[150],[151] and [152],[154] Comparisons between CT and magnetic resonance imaging (MRI) have shown little difference between the two modalities.[157],[158],[159] and [160] For example, sensitivity, specificity, and accuracy rates between CT scanning (100%, 80%, and 84%, respectively) and MRI (100%, 84%, and 87%, respectively) are comparable.[157] Early spread beyond the esophageal wall, regional lymph node involvement, and aortic invasion have led to problems with both false positivity and false negativity with both imaging modalities.[150],[151],[153],[154],[156],[158],[159] and [160] Thus, CT scanning remains the primary staging modality.

Endoscopic Ultrasound EUS has emerged as a key component of staging. It is particularly useful in gauging depth of esophageal invasion and local lymph node involvement(Fig. 62-2). EUS is superior to CT scanning in determination of T and N of the staging classification scheme and has been compared with surgical pathological and CT staging in several studies.[161],[162],[163] and [164] EUS accuracy in staging depth of tumor invasion varies from 87% to 92%, making it more accurate than CT. The overall accuracy of EUS in detecting regional lymph node status ranges from 80% to 88%. In these studies, EUS sensitivity varied from 92% to 95% and specificity varied from 50% to 54%, whereas CT accuracy ranged from 51% to 74%, with sensitivity of 34% and specificity of 88%.

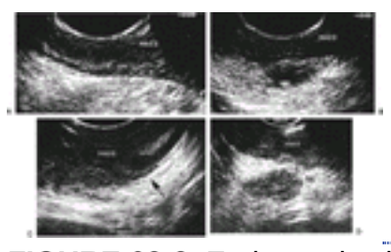

FIGURE 62-2. Endoscopic ultrasound images of different stages of esophageal cancer. (Courtesy of William Brugge, MD.) (**A**) T2N0 stage. Note the prominent muscularis propria (*m.p.*) (**B**) T2N1 stage. The mass is directly adjacent to the muscularis propria. (**C**) T3N0 stage. Note invasion into the muscularis propria (*arrow*). (**D**) T3N1 stage. A mass with a large lymph node in mediastinal fat.

EUS is useful in the assessment of resectability. It was able to predict resectability in 147 of 167 patients (89%), more accurately for adenocarcinoma (82%) than for SCC (64%).[165] Lumenal stenosis, which prevents passage of the scope, portends a higher stage, either III or IV (older staging system), in nearly 90% of cases; conversely, 58% of cancers without stenosis were not stage III or IV.[166] Overstaging (T1 to T3) or understaging (T1 or T2) may be a problem with EUS; 73% of T2 lesions are overstaged, and 71% of T3 lesions are understaged.[167] EUS is superior to CT scanning for T and N repeat staging after preoperative chemotherapy, although some difficulty in differentiating between residual tumor and fibrosis may ensue.[168] Cost-effectiveness studies of EUS staging are needed.

Laparoscopy Abdominal metastases are more common from distal esophageal tumors, and these subdiaphragmatic metastases may be confirmed by laparoscopy.[169] Nonetheless, laparoscopy is not common practice for staging of esophageal cancer.

ESOPHAGEAL ADENOCARCINOMA

Epidemiology

Barrett esophagus is the most important precursor to esophageal *adenocarcinoma*.[170],[171] Esophageal adenocarcinomas arising from esophageal glands [171],[172] or heterotopic gastric mucosa [173] are rare. Furthermore, many of the adenocarcinomas of the gastric cardia probably arise in short segments of Barrett metaplasia rather than from the gastric epithelium.[174],[175] and [176] Once considered rare,[177],[178],[179] and [180] the incidence of esophageal adenocarcinoma (both of esophageal and gastroesophageal junctional origin) has increased at a rate of 4% to 10% annually in different regions of the United States.[180] This rapid increase is greater than that of any other cancer.[9],[180] Although adenocarcinoma of the esophagus is among the top 15 malignant diseases and is more common than esophageal SCC in the United States, the latter remains more common worldwide than adenocarcinoma.

Esophageal adenocarcinoma affects mostly whites. The incidence typically begins to increase after the age of 40 years,[175],[177],[181],[182] and [183] and it rises with each succeeding decade.[9] It is a disease of men, with a male-to-female predominance of 3 to 5.5:1.[170],[180],[182] The annual age-adjusted incidence rates of adenocarcinoma of the esophagus and gastric cardia in white men is 1.3 and 2.8 per 100,000, respectively.[180]

Etiology

Barrett esophagus is a premalignant condition and is the most important risk factor for esophageal adenocarcinoma (see Chapter 60). Alcohol and cigarette smoking are not key risk factors in the development of Barrett esophagus, but they may be important once the condition is established.

Patients with Barrett esophagus are identified at an average age of 55 years. It typically affects whites, a feature shared with esophageal adenocarcinoma. Barrett esophagus may develop as a complication of chronic gastroesophageal reflux, and it is defined as the replacement of the normal squamous epithelium of the esophagus by metaplastic columnar epithelium.[183],[184],[185],[186],[187],[188],[189],[190] and [191] Patients with cystic fibrosis and scleroderma may be at increased risk of Barrett esophagus.

The incidence of adenocarcinoma arising in Barrett esophagus varies from 1 cancer per 55 to 441 patient years, or about 500 cases per 100,000.[192],[193],[194],[195],[196],[197],[198],[199],[200] and [201] This incidence rate is a figure greater than that for esophageal SCC in high-risk areas of the world, and it corresponds to nearly a 125-fold increased risk when compared with the general population in the United States. The prevalence of adenocarcinoma at the time of initial diagnosis of Barrett esophagus is approximately 8%.[177],[183],[191],[192],[193],[194],[195] and [196] Incidence estimates of esophageal adenocarcinoma arising in Barrett esophagus are highly

variable, probably because of problems with classification of Barrett esophagus as only affecting gastric cardia mucosa, with designating distal esophageal cancers as primary gastric cancers, and with whether the study was retrospective or prospective. [197] In any case, the incidence estimates for esophageal adenocarcinoma provide a compelling need for the detection of Barrett esophagus and associated dysplasia. [202], [203] Historically, investigators believed that long-segment Barrett esophagus (>3 cm) carries a higher risk of cancer than the more prevalent short-segment Barrett esophagus (<3 cm); however, one study found that the risk of cancer in short-segment Barrett esophagus is not much lower than in the long-segment variant. [204]

Apart from Barrett esophagus, symptomatic gastroesophageal reflux [205] and increased body mass index are reported to be associated with esophageal adenocarcinoma. [206] Additionally, there may be an association between medications that relax the lower esophageal sphincter and the risk of esophageal adenocarcinoma. [207]

Biology and Genetics

The transition of Barrett metaplasia to adenocarcinoma [208] involves progression through low-grade dysplasia, high-grade dysplasia, and carcinoma *in situ*. The molecular mechanisms underlying this progression are being elucidated. Abnormal DNA content, as reflected by aneuploidy measured through flow cytometry, is associated with dysplasia and adenocarcinoma, [209], [210], [211] and [212] as well as with metaplastic or dysplastic populations of cells or clones in close proximity to adenocarcinoma. [213], [214] Abnormalities in cell proliferation, as assessed by proliferating cell nuclear antigen (PCNA) and Ki-67 expression, are noted in Barrett metaplasia, dysplasia, and esophageal adenocarcinoma.

As with many cancers, esophageal adenocarcinoma involves the activation of oncogenes, inactivation of tumor suppressor genes, and mutations in DNA mismatch repair genes (Fig. 62-3). A key mechanism affecting tumor suppressor genes and putative tumor suppressor genes is loss of heterozygosity of chromosomal regions 17p (*p53*), 5q (*APC, MCC*), 18q (*DCC*), and 13q (*RB1*). [215], [216], [217], [218] and [219] Loss of heterozygosity of 17p (*p53*) typically occurs before 5q (*APC*) loss during neoplastic progression in Barrett esophagus, [308] rather than the reverse sequence, as in colon cancer. Furthermore, actual DNA mutational analysis indicate that *p53* gene alterations precede the onset of DNA aneuploidy and cancer. [220], [221] and [222] As in SCC, cyclin D_1 oncogene overexpression has been described. [223] Increased expression of epidermal growth factor and its receptor and expression of transforming growth factor-a are found in Barrett-associated adenocarcinoma. [224] Microsatellite instability, a reflection of changes in DNA mismatch repair genes, was found in 2 of 27 patients with Barrett metaplasia and in 8 of 36 patients with esophageal adenocarcinomas. [225] In fact, microsatellite instability can develop as an early event in metaplasia and in diploid tumor cells. *Ras* mutations are not present in esophageal adenocarcinoma.

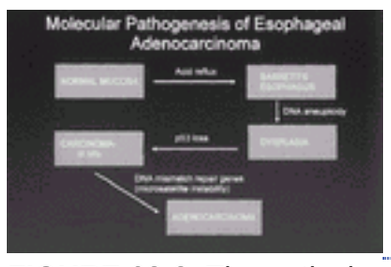

FIGURE 62-3. Theoretical genetic model of esophageal adenocarcinoma.

Clinical Manifestations

Early esophageal adenocarcinoma may be silent or may present with symptoms and signs of gastroesophageal reflux and its complications, including heartburn, regurgitation, benign strictures with dysphagia, and bleeding from ulcerations. [226], [227] and [228] Consequently, these manifestations merit meticulous endoscopic mucosal biopsies. [229], [230], [231] and [232] There are conflicting reports on the correlation between symptoms and the presence of esophageal dysplasia or early adenocarcinoma. An analysis of 741 patients in 15 published series indicated that 98% of patients had symptoms of gastroesophageal reflux or its complications. [233] An autopsy study, however, suggested that most cases of Barrett esophagus remain clinically unrecognized in the general population. [234] Symptoms referable to advanced esophageal adenocarcinoma are similar to those of advanced SCC: dysphagia, odynophagia, weight loss, chest pain, nausea, vomiting, cough, and usually occult gastrointestinal bleeding. Laboratory evaluation may reveal anemia (secondary to gastrointestinal bleeding) and signs of malnutrition (hypoalbuminemia or elevated prothrombin time secondary to vitamin K deficiency).

Natural History and Complications

Although the period of progression from Barrett metaplasia to adenocarcinoma remains undefined, and in fact may vary, [196], [201], [229], [230] it is clear that only a subset of patients with Barrett esophagus will develop adenocarcinoma. This is probably the result of the interplay of gastroesophageal reflux with genetic alterations. Metaplasia appears to migrate proximally with ongoing reflux but not with advancing age. [187], [188] and [189]

The events involved in the progression of Barrett esophagus to adenocarcinoma have been extrapolated from studying the high-grade dysplasia in mucosa surrounding the cancer. [182], [229], [230] Although it is clear that gastroesophageal reflux contributes to the development of dysplasia and adenocarcinoma, other factors, yet to be identified (e.g., bile reflux), are also important. This is underscored by the finding that pharmacological therapy or antireflux surgery does not convert the metaplastic epithelium back to normal squamous epithelium, nor does it prevent the development of adenocarcinoma.

Low-grade dysplasia may progress or regress at endoscopic follow-up. However, one third of patients with high-grade dysplasia who undergo esophagectomy have foci of adenocarcinoma in the resected specimens. [231], [235], [236], [237], [238], [239], [240], [241], [242], [243], [244], [245], [246], [247] and [248] Obviously, many cases of high-grade dysplasia do not progress to adenocarcinoma. [201], [229], [230] and [231] Given the absence of clinical parameters or biomarkers to stratify patients with high-grade dysplasia, esophagectomy is warranted whenever possible.

If undetected in its early stages, esophageal adenocarcinoma may invade the muscularis propria as well as the adventitia and metastasize to adjacent organs such as regional lymph nodes, lung, liver, peritoneum, bone, and brain. There is also an increased incidence of synchronous cancers in the esophagus.

Diagnostic Evaluation

Endoscopy The mainstay of diagnosis is endoscopy, with confirmation of Barrett esophagus by histopathology, and the establishment of metaplasia or dysplasia (Fig. 62-4, Fig. 62-5 and Fig. 62-6; see also Color Fig. 62-4 and Color Fig. 62-5). Suspicion of Barrett esophagus or adenocarcinoma warrants endoscopy with biopsy. In Barrett esophagus, the squamocolumnar junction is displaced proximal to the region of the lower esophageal sphincter and represents the junction between squamous epithelium and specialized metaplasia. [236] It is important to recognize hiatal hernias, so biopsies from the gastric hernia are not mistakenly classified as Barrett esophagus (see Chapter 60 and Chapter 138).

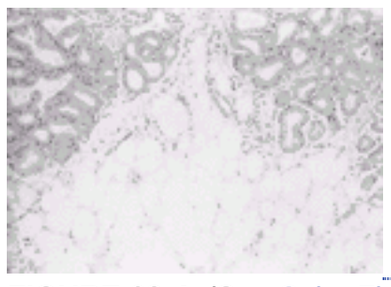

FIGURE 62-4. (See Color Fig. 62-4.) Specialized-type Barrett esophagus. The epithelium shows intestinal-type absorptive cells, goblet cells, and mucinous cells in a villiform pattern. (Courtesy of Robert Odze, MD.)

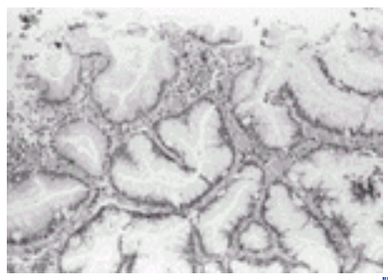

FIGURE 62-5. (See Color Fig. 62-5.) High-grade dysplasia in Barrett esophagus. The epithelium shows architectural complexity, atypia, pleomorphism, and nuclear stratification. (Courtesy of Robert Odze, MD.)

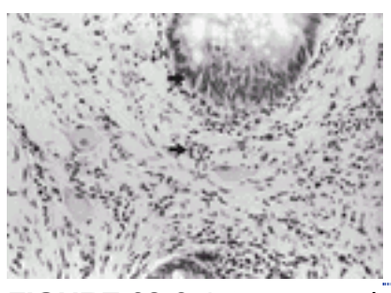

FIGURE 62-6. Intramucosal adenocarcinoma in Barrett esophagus. Tumor invasion beyond the basement membrane is present in the form of single cells, small glands, or sheets of cells. (Courtesy of Robert Odze, MD.)

Early cancers may be associated with superficial erosions, plaquelike elevations, or areas of nodularity. As the cancer becomes more advanced, it may appear as an intralumenal polypoid mass, a stricture (which may be asymmetric), or a deep ulceration. Esophageal adenocarcinomas may be detected in short-segment or long-segment Barrett esophagus. Furthermore, adenocarcinomas that appear to be in the gastric cardia may, in fact, have developed in short segments of Barrett metaplasia.

Endoscopic Biopsy and Cytology When either short-segment or long-segment Barrett esophagus is suspected, four-quadrant endoscopic biopsies should be performed at 2-cm intervals in a systematic fashion. Multiple biopsies also should be taken of any erosions, nodules, ulcers, or strictures, no matter how insignificant they appear. [230] In addition, biopsies should be obtained from “control” areas, namely, the gastric cardia and the normal-appearing squamous epithelium. This protocol has been shown consistently to detect high-grade dysplasia or early adenocarcinoma arising in Barrett esophagus. Endoscopic cytology has been advocated, but it has a variable high false-negative rate, [249], [250], [251] and [252] making it unacceptable as a primary diagnostic tool for clinical practice, although its role in screening has been resurrected. [253] Caution must be exercised in interpreting the histological diagnosis of dysplasia in Barrett esophagus. Although the diagnosis of high-grade dysplasia or carcinoma can be made with confidence, interobserver variability may exist for indefinite dysplasia or low-grade dysplasia. Such cases may require independent pathological confirmation. When biopsies reveal high-grade dysplasia from a mass, ulcer, or a malignant-appearing stricture, endoscopy and repeat biopsies should be performed to exclude malignancy. As outlined earlier, high-grade dysplasia may dictate esophagectomy anyway. Sometimes, high-grade dysplasia and well-differentiated adenocarcinoma may be difficult to distinguish histologically because the muscularis mucosae is not obtained with endoscopic mucosal biopsies, and its invasion is necessary to define adenocarcinoma. Flow cytometric detection of aneuploidy or increased G_2-tetraploid fractions may provide additional important information concerning patients at risk of esophageal adenocarcinoma. The prevalence of aneuploid cell populations increases with metaplasia to different grades of dysplasia. [254], [255] Moreover, some patients may have aneuploidy without histological evidence of high-grade dysplasia or adenocarcinoma. In 9 of 13 patients (cohort of 62 patients) whose biopsies initially showed aneuploid cell populations or increased G_2 fractions, but who later had high-grade dysplasia or adenocarcinoma, the detection of aneuploidy seems to have been an excellent prospective marker. This was not the case in the remaining 49 patients without aneuploidy or increased G_2 fractions who progressed ($P < 0.0001$). The value of increased G_2 fractions has been debated, but with proper processing of mucosal biopsies and appropriate extraction of nuclei, there does appear to be an association of high G_2 fractions with progression to dysplasia and adenocarcinoma. [256]

Barium Esophagography Barium swallow may raise the index of suspicion of Barrett esophagus in the presence of stricture or ulcer. Uncomplicated Barrett esophagus may be difficult to discern, except for mucosal irregularities. [257], [258], [259], [260] and [261] Advanced esophageal adenocarcinoma usually has the same radiographic signs as observed with SCC, except SCCs are more common in the proximal or midesophagus, whereas adenocarcinomas are typically in the distal esophagus (Fig. 62-7).

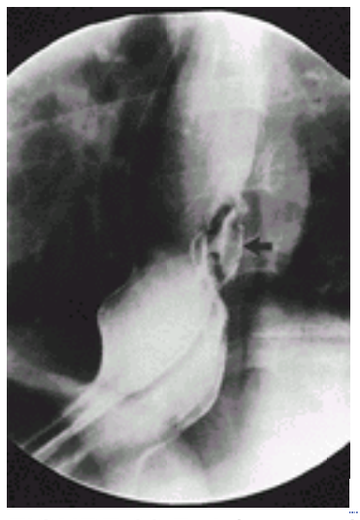

FIGURE 62-7. Adenocarcinoma represented by an ulcerated mass in the distal esophagus. (Courtesy of Deborah Hall, MD.)

Staging

Preoperative CT scans are useful in staging esophageal and gastroesophageal junction adenocarcinomas, and they carry the same advantages and limitations as in the staging of SCC. [262] EUS is also an important diagnostic tool, with the same technical issue as in SCC.

Surveillance of Barrett Metaplasia and Dysplasia

A decision model comparing no surveillance with surveillance every 1 to 5 years with surgical resection for high-grade dysplasia or cancer found two important variables for the selection of a particular surveillance strategy: the incidence of cancer arising in Barrett esophagus and the quality of life after surgical resection. [263] Notwithstanding these important considerations and the absence of definitive surveillance recommendations, certain guidelines from leading expert centers may help to guide clinical practice. Endoscopic biopsies taken systematically that are negative for histological dysplasia and are without flow cytometric and cytologic abnormalities should lead to repeat endoscopic evaluation at 2-year intervals. [264] This protocol can be modified on an individual basis if the clinical situation warrants it. If high-grade dysplasia is detected histologically without any endoscopic abnormalities, then early repeat endoscopy with multiple biopsies should be pursued to document the extent of high-grade dysplasia and to exclude coexisting cancer. Treatment recommendations for high-grade dysplasia need to be individualized. Surgical resection has to be balanced against the patient's performance status, coexisting illnesses, compliance with endoscopic surveillance, and institutional experience with surgical resection. For patients with high-grade dysplasia who do not opt for surgical resection but who would do so if cancer were diagnosed, then endoscopic surveillance biopsies should be repeated 3 months after the initial two endoscopies and at 6-month intervals thereafter. Adenocarcinoma can develop in small or large areas of high-grade dysplasia, and thus systematic biopsies must be taken.

In a provocative study of the use of balloon cytology in the surveillance of Barrett esophagus for dysplasia, adequate columnar epithelium was obtained in 52 of 63 patients with balloon cytology and in 59 of 61 patients with brush cytology. [253], [265] Balloon cytology obtained abnormal cells in 6 of 8 patients with adenocarcinoma, in 2 of 2 of patients with high-grade dysplasia, but in only 2 of 8 patients with low-grade dysplasia. Brush cytology was abnormal in all 11 patients with high-grade dysplasia but in only 2 of 9 patients with low-grade dysplasia. The authors concluded that balloon cytology may be adequate and cost effective for the detection of high-grade dysplasia or carcinoma, provided sampling is adequate and a more abrasive balloon could be pursued. Although these results are promising, more studies are needed.

The search for markers of intestinal metaplasia as a means of identifying patients who may develop high-grade dysplasia and adenocarcinoma has yielded promising results with the measurement of small intestinal brush border enzymes such as sucrose isomaltase and aminopeptidase N. [214] Additionally, markers of proliferation such as PCNA, Ki-67, tritiated thymidine uptake, and ornithine decarboxylase may be useful as well, but they have not been extended to wide-scale clinical application. [214]

THERAPY FOR ESOPHAGEAL SQUAMOUS CELL CARCINOMA AND ADENOCARCINOMA

The therapeutic approaches for esophageal SCC and esophageal adenocarcinoma are becoming increasingly indistinguishable, and therefore they are considered together. The presentation of esophageal cancer at advanced stages mitigates against curative surgical resection on a consistent basis. In fact, although nearly 60% of patients may undergo surgical exploration, only two thirds of such patients will undergo resection. Thus, more than 40% of patients will require palliative therapy. Overall, 1- and 5-year survival rates are approximately 18% and 5%, respectively. [266] The following discussion is divided into categories of potential curative modalities as well as palliative measures.

Surgery

Surgery remains the mainstay of treatment of esophageal cancer with curative intent for respectable, local, and locoregional disease. Advances in surgical therapy, staging techniques, patient selection, and supportive care in recent years have resulted in a marked improvement in surgery-related mortality and morbidity. Even if cure is not accomplished, palliative surgical bypass procedures may also have some impact on patients with advanced unresectable disease to relieve symptoms and to improve the quality of life.

There are various surgical approaches for esophageal cancer, including transthoracic, transhiatal, and radical en bloc resections. Despite the different advantages and disadvantages of each, and the controversies surrounding them, there is no evidence to suggest improved survival with any particular one. [267] Factors that may influence the decision of selecting a particular technique include tumor location, depth of tumor invasion, status of lymph node involvement, overall performance status and body habitus of the patient, previous treatment (e.g., radiation or chemoradiation therapy), and the preference and biases of the surgeon and institution.

In the 1940s, midesophageal tumors were approached through the left side of the chest. The benefits of a right-sided approach (Ivor-Lewis) have been advocated since then and include enhanced accessibility to the upper two thirds of the esophagus, location of the aorta as a barrier to the left pleural space, and visualization of the entire esophagus after division of the azygous vein. This approach has been applied as well for distal esophageal cancers, thus allowing for a longer segment of esophageal resection with a higher anastomosis. A cervical anastomosis has some inherent advantages, namely, decreased morbidity and mortality, reduced reflux, and a better opportunity for postoperative radiation if pursued. The left thoracoabdominal approach may be used for distal esophageal and gastroesophageal cancers.

The early complications of transthoracic esophagectomy include anastomotic leak (0%–13%), pulmonary problems such as pneumonia, retained secretions, and pulmonary emboli (6%–50%), recurrent laryngeal nerve injury (1%–13%), and cardiac problems such as myocardial infarction, arrhythmia, and congestive heart failure (2%–27%). The mortality ranges from 1% to 13%, with a 5-year survival of 1% to 35%.

Owing to high rates of morbidity and mortality associated with the thoracotomy associated with transthoracic esophagectomy, the transhiatal approach has also been advocated. Complications are the same as with transthoracic approach, but with a higher incidence of anastomotic leak (9%–36%). Mortality ranges between 2% and 13%, and 5-year survival ranges from 12% to 27%. [272], [273], [274], [275], [276], [277], [278], [279], [280] and [281]

The en bloc resection for cancers of the distal esophagus and the cardia involves removal of 10 cm of the esophagus on either side of the tumor along with contiguous tissues, vascular supply, and lymph nodes. Various series reported a 1.4% to 11% operative mortality, with a 5-year survival of 18% to 35%. [283], [284] En bloc resection as a curative procedure is controversial, and most surgeons in Western countries favor a transthoracic or transhiatal approach. [285], [286] and [287]

An important consideration during surgery is the choice of esophageal replacement. [288], [289], [290], [291], [292] and [293] The stomach, by virtue of its rich blood supply, length, and muscular wall, is preferred, whereas the left colon, because of its similarity to the esophagus in caliber, can be used to replace the esophagus when the stomach cannot be used. Jejunal interposition may be used for reconstitution of the hypopharynx and the cervical esophagus. Overall, complications of esophageal replacement include anastomotic leaks (2%–5%), anastomotic stricture (9%–29%), gastric dysfunction, and chylothorax.

Unresectable esophageal cancer may warrant a bypass procedure using a portion of stomach, jejunum, or colon as a conduit. However, bypass surgery is fraught with high morbidity and mortality rates, and it should be reserved for patients with a tracheoesophageal fistula or those in whom conservative medical measures have failed. Effective palliative resection of the cervical esophagus usually requires total laryngectomy and esophagectomy (transhiatal approach).

Radiation Therapy

Radiation (External Beam) Alone The uses of radiation as a single modality are limited to patients who are not candidates for surgical resection because of unresectable disease or patients who are medically unable to tolerate a surgical procedure and multimodality treatment. [294], [295] and [296] Limited experience with patients treated with radiation therapy alone indicate a 1-year survival of 18% and a 5-year survival of 6%, reminiscent of survival data after surgical resection. [297], [298] and [299] Favorable 5-year survival rates may be observed in women and in patients with cancers less than 5 cm in length (22.4% and 17.7%, respectively). [294], [295], [296], [297], [298] and [299] Radiation alone may lead to amelioration of dysphagia, with 50% of patients receiving relief for 2 months or more and 15% of patients for more than 1 year. In the acute setting, radiation-induced esophagitis may lead to dysphagia, and fibrotic strictures may form. Unusual complications of radiation therapy include pulmonary fibrosis, myelitis, and cutaneous burns.
Brachytherapy Intralumenal radiation (brachytherapy) can ameliorate symptoms resulting from obstructing cancers. Higher doses of radiation can be delivered with this technique without inducing injury to pulmonary, cardiac, and mediastinal structures. [300] A cobalt-60 source is commonly used, either alone or after external beam radiation therapy, and treatment is typically administered over 2 weeks. Overall and complete response rates with brachytherapy range between 74% and 85% and 9% and 53%, respectively. [301], [302] Most patients experience a prompt subjective response to brachytherapy. Complications of brachytherapy include ulcers, strictures, and tracheoesophageal fistula in up to 25% of patients. [301], [302]

Preoperative (Neoadjuvant) and Postoperative (Adjuvant) Radiation Therapy The potential advantage of neoadjuvant radiation therapy includes increased respectability, decreased tumor seeding at the time of surgery, and increased radiosensitivity as a result of more oxygenated cells. Neoadjuvant radiation has not been shown to improve overall survival of patients with potentially respectable disease. [303], [304], [305], [306] and [307] A metaanalysis of 1147 patients from 5 randomized neoadjuvant radiation therapy trials (median follow-up of 9 years) showed no statistically significant benefit in either the 2-year survival rate (34% versus 30%) or the 5-year survival rate (18% versus 15%). [307] There was benefit demonstrated in radiation treatment, with an overall reduction in the risk of death by 11%. Adjuvant radiation therapy provides the advantages of accurate pathological staging, and treatment is directed to areas with a high risk of disease recurrence. Postoperative radiation therapy is often given when the surgical margins are positive or the resection is incomplete. A slightly higher total dose range (4500 to 5500 cGy) than that administered in the neoadjuvant randomized trials has not yielded a survival advantage. [308]

Chemotherapy

Because approximately 30% of patients present initially with metastatic disease and at least 50% of others present with stage II and III disease, chemotherapy holds promise for local control of disease, increased survival, and prevention of distant metastasis. Cisplatin and 5-fluorouracil (5FU) have been used as a standard regimen, and they have produced response rates of 35% to 40% in advanced disease with a median survival of 33 weeks and a 1-year survival of 38%. Other active agents are mitomycin and bleomycin, as well as many newer agents such as taxans (paclitaxel and docetaxel), irinotecan, gemcitabine, and oxaliplatin.

Neoadjuvant (Preoperative) Chemotherapy The rationale of neoadjuvant chemotherapy is to eradicate occult metastases before surgery. No advantage of neoadjuvant therapy was demonstrated in the 1980s or 1990s. [311], [312], [313], [314] and [315] More recently, data from a large (802 patients), randomized, and well-controlled study of neoadjuvant chemotherapy in esophageal cancer conducted by the United Kingdom Medical Research Council upper gastrointestinal tract cancer group has been very encouraging. [316] Patients with esophageal cancer, 67% of whom had adenocarcinoma, were randomized to neoadjuvant chemotherapy before surgery or to surgery alone. Resectability was significantly higher in the treatment arm (78% versus 70%; $P < 0.001$), as was the overall survival ($P = 0.003$). Two-year survival rates were 43% in the neoadjuvant chemotherapy arm versus 34% in the surgery-alone arm. Postoperative complications were similar (40%–41%).
Adjuvant (Postoperative) Chemotherapy A trial to evaluate the efficacy of postoperative chemotherapy in resectable lesions showed an advantage of disease-free survival in the treatment arm ($P = 0.05$), but there was no overall survival benefit. [317] Several clinical trials are ongoing to evaluate this approach further, using newer chemotherapy agents and regimens.

Primary Chemoradiation Therapy

Cisplatin and 5FU combined with radiation therapy can substantially improve the 5-year survival in patients with esophageal cancer compared with radiation alone. Cisplatin and 5FU are radiosensitizers and improve the response to radiation therapy not only in the cancer cells, but unfortunately in normal tissues as well.

Definitive primary chemoradiation therapy for locoregional esophageal carcinoma is considered an alternative to surgical resection, although there are no large prospective randomized trials comparing it directly with surgery. The largest single-institution trial of primary chemoradiation was conducted by Coia [318] on 57 patients with stage T1 and T2 esophageal cancer. These patients were treated with radiation, 6,000 cGy, and two cycles of 5FU delivered as a continuous 4-day infusion during the first and fifth week of radiation therapy, and a single bolus of mitomycin C on day 2. The median survival was 18 months, and 3-year and 5-year survival rates were 29% and 18%, respectively.

A phase III multiple-institution randomized trial compared combined modality with radiation therapy alone. [319] Patients randomized to combination therapy received a total dose of 5000 cGy radiation and 5FU for 4 consecutive days and cisplatin on day 1 in weeks 1, 5, 8, and 11. Patients in the radiation-alone group were given a total dose of 6400 cGy. This trial was stopped early after an interim analysis demonstrated a survival advantage for patients in the combination therapy group. The 5-year survival rate for patients who had chemoradiation was 26% versus 0% for those receiving radiation only ($P < 0.001$), with median survival of 14.1 months versus 9.3 months, respectively. There was no statistical difference in survival related to histology of cancer (adenocarcinoma versus SCC).

Neoadjuvant Chemoradiation Therapy

Data from several nonrandomized studies suggest that neoadjuvant therapy with chemotherapy (5FU and cisplatin) plus radiation before surgery yielded complete response rates of 24% to 42%, with median survival of 12 to 23 months. [319], [320], [321] and [322] One hundred patients (25 with SCC and 75 with adenocarcinoma) were randomized to concurrent continuous infusion of 5FU, cisplatin, and vinblastine with radiation 45 Gy followed by surgery versus surgery alone in a University of Michigan study. Twenty-eight percent of patients in the neoadjuvant chemoradiation arm achieved pathological complete response. Three-year survival rates were 30% in the combination therapy arm versus 16% in the surgery alone arm, but this difference was not statistically significant ($P = 0.15$). Median survivals were similar in both arms of the trial. [323] A phase III trial had 113 patients with esophageal adenocarcinoma randomized to 5FU infusion and cisplatin with radiation (40 Gy) plus surgery or surgery alone, [324] and a trial of 297 patients with SCC (with surgery alone, and cisplatin with radiation followed by surgery) was inconclusive. [325] Large randomized trials are needed to address this approach further.

Adjuvant Chemoradiation Therapy

Medical and radiation oncologists have also administered combined modality chemoradiation therapy postoperatively to patients with esophageal cancer, based on the proven benefit observed in other gastrointestinal malignancies, such as rectal cancer. Several small or retrospective studies suggested a survival benefit of adjuvant chemoradiation for patients with esophageal and gastroesophageal junction cancer. [326], [327], [328] and [329] A large intergroup study from the United States demonstrated the survival benefit of adjuvant chemoradiation in such patients. [330] Six hundred and three patients with complete resections, negative surgical margins, and no evidence of residual disease were randomized to receive surgery alone or surgery plus postoperative chemotherapy with 5FU and leucovorin and concurrent radiation therapy. The median survival was 36 months for patients in the adjuvant chemoradiation arm versus 26 months for patients in the surgery-only arm ($P = 0.005$). The 3-year overall survival rates were 50% versus 40%, and disease-free survival rates were 48% versus 31%, for the treatment group versus the observation group. The median disease-free survival rates were 30 months for chemoradiation-treated patients and 19 months for surgery-only patients ($P < 0.001$). The toxicities of the chemoradiation were acceptable, with hematologic and gastrointestinal toxicities comprising the predominant adverse events. Three patients (1%) died as a result of toxicities. This study is the first large well-controlled study demonstrating an advantage to adjuvant therapy, with respect to both disease-free survival and overall survival. Unfortunately, the study was not designed specifically for answering the role of adjuvant therapy in esophageal cancer.

Summary

For patients with stage T1 and T2 lesions without any evidence of nodal involvement, surgery alone may be curative in more than 60%. For patients with locally advanced disease, the emerging recommendation entails perioperative chemotherapy and/or radiation therapy plus surgical resection. Patients should be encouraged to enter large randomized clinical trials whenever possible, because definitive survival benefits of either neoadjuvant or adjuvant multimodality therapy with surgery need further study.

Palliation Treatment

Local Disease Because fewer than 50% of patients are candidates for a curative approach after careful staging and preoperative assessment of comorbid disease, palliative approaches are mandatory. Because total esophagectomy is fraught with high mortality in palliative situations, bypass should be considered for those patients with expected survival of more than 6 months. External beam radiation therapy (with or without chemotherapy) may be suitable for annular or nearly obstructive lesions, but radiation-induced esophagitis may be problematic. Lower does may be an option. Endoscopic dilation can be employed to relieve the stricture and is associated with a low complication rate. [331], [332] However, repeated dilations may be necessary. Expandable metal stents are a good option to maintain patency; additionally, laser and bipolar electrocoagulation therapy may be helpful (see Chapter 145). It is worth emphasizing the exciting potential of photodynamic therapy. An intravenously administered photosensitizer, porfimer sodium (approved by the Food and Drug Administration for use in the palliation of esophageal cancer) is administered, and then light of a specific wavelength (630 nm) is shone on the tumor. Cell toxicity is believed to occur through release of singlet oxygen resulting in ischemia and, eventually, necrosis. Photodynamic therapy's depth of destruction is 2 to 5 mm, and it is preferred to laser in tumors longer than 8 cm (Fig. 62-8; see also Color Fig. 62-8).

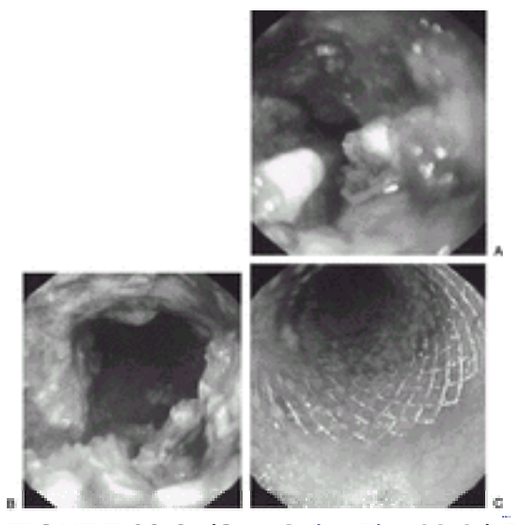

FIGURE 62-8. (See Color Fig. 62-8.) Photodynamic therapy of esophageal cancer with laser after administration of porfimer sodium, a photosensitizer. Light of 630 nm from a laser acts on cells that accumulate the photosensitizer (**A**). After 6 days, there is some decrease in the mass size (**B**). After 12 days, the mass is diminished in size significantly, and a metallic endoprosthesis is endoscopically inserted (**C**). (Courtesy of Norman Nishioka, MD.)

Metastatic Disease For distant metastatic disease, systemic chemotherapy with 5FU and cisplatin is the main modality for palliative care response, and survival rates are not impressive. With newer agents such as paclitaxel, docetaxel, gemcitabine, irinotecan, and oxaliplatin, response rates have improved to 50% to 60%, and median survival has increased to 10 to 14 months, with gains in quality of life. [333], [334] and [335]

Prognostic Factors

It would be helpful to be able to identify patients a priori who would benefit from certain types of therapeutic modalities. Performance status, weight loss than 10% of body weight, the tumor stage, and lymph node involvement from primary tumor are also very important. [336], [337]

The chances for curative surgical resection are optimized for lesions less than 5 cm, confined to the mucosa and without lymph node involvement. Poor prognosis is correlated with aneuploidy and histology. For those patients who achieve pathological complete response to neoadjuvant chemotherapy, long-term survival has been significantly improved. [338] Increased cyclin D_1 expression and reduced E-cadherin expression portend a less favorable outcome. [339] A high serum vascular endothelial growth factor (VEGF) was associated with tumor progression, poor treatment response, and poor survival in patients with SCC. [340] It is also suggested that high expression of *BAX* and *p16* ^{INK4a} are associated with a favorable prognosis. [341] Importantly, patients need to abstain from tobacco and alcohol during treatment.

The extent of disease is an important prognostic factor for esophageal adenocarcinoma. After resection, negative surgical margins are important for improving prognosis; conversely, the development of distant metastasis during treatment portends a poor prognosis. The type of operation is not an independent prognostic factor, nor is ultrasound assessment after neoadjuvant therapy. Identification of micrometastatic disease in lymph nodes of patients with esophageal cancer may permit stratification to better or worse survival. [342]

BENIGN EPITHELIAL TUMORS (SQUAMOUS CELL PAPILLOMA)

Squamous cell papillomas are usually discovered incidentally at endoscopy on radiography. Generally, they are small, sessile, and polypoid growths of normal or hyperplastic squamous epithelium covering a core of connective tissue. [343], [344] On removal, they typically do not recur. [345], [346] The average age of patients is 51.5 years, with a male-to-female ratio of 1.8:1. These tumors occur more frequently in Japan. Most are solitary and located in the distal third of the esophagus. Papillomas may result from chronic irritation from reflux esophagitis. Morphologic features suggest that papillomas result from HPV infection, and HPV DNA has been found in a subset of papillomas. Actual development of cancer has not been documented in these tumors. This may reflect that the nononcogenic genotypes of HPV may be associated with esophageal papillomas as in the skin. NMBA administered subcutaneously in rats leads to the development of esophageal squamous dysplasia and papillomas.

MALIGNANT EPITHELIAL TUMORS

Squamous Cell Carcinoma with a Spindle Cell Component

SCC with a spindle cell component is also referred to as carcinosarcoma, pseudosarcoma, spindle cell carcinoma, and polypoid carcinoma. [347], [348], [349], [350], [351], [352] and [353] Originally, carcinosarcoma was believed to be malignant, whereas pseudosarcoma was designated benign without metastatic potential. Local and distant metastasis of spindle cells, however, has been reported.

Spindle cell carcinomas are either solitary or multiple and are large and polypoid. Histologically, the squamous component at the surface is associated with dysplasia or carcinoma in situ. One may also see adenocarcinoma or undifferentiated epithelial components. The spindle cell component may range from mild proliferation to marked proliferation with pleomorphism, giant cells, and mitoses. Differentiation into smooth or skeletal muscle, bone, or cartilage may be rarely observed. Immunohistochemical and electron microscopic analyses indicate that the spindle cells stem from mesenchymal metaplasia of malignant squamous cells. Although there are reports that spindle cells contain epithelial cell markers such as cytokeratins, suggesting multiclonality, most pathologists believe that spindle cell carcinomas arise from SCCs that produce a spindle cell component.

Clinically, men are affected more than women, and most patients are middle aged or elderly. Located in the middle to distal esophagus, these cancers can produce obstruction and dysphagia by virtue of their large size. Most patients present with esophageal wall invasion and lymph node involvement.

Verrucous Squamous Cell Carcinoma

Verrucous SCC of the esophagus classically is a slow-growing malignant tumor that may be limited to local invasion but without distant metastasis. [354], [355] Some, however, do metastasize to other organs, with resulting poor prognosis.

Adenocanthoma and Adenosquamous Carcinomas

These rare tumors feature adenocarcinoma with squamous metaplasia (*adenocanthoma*) or with squamous carcinoma (*adenosquamous carcinoma*). [356], [357] These lesions can arise in the setting of Barrett esophagus or de novo. It is likely that these tumors arise in the squamous epithelium that has the ability to achieve squamous and glandular differentiation. Alternatively, squamous metaplasia or transformation within an adenocarcinoma may have occurred. Clinically, these tumors behave like adenocarcinomas.

Adenoid Cystic Carcinoma

This is a rare tumor that resembles salivary adenoid cystic carcinomas. Carrying a male predilection at an advanced age, these tumors are multilobulated or ulcerated and located in the midesophagus. [358], [359], [360], [361], [362] and [363] Histologically, *adenoid cystic carcinomas* contain many cystic spaces and are primarily submucosal.

Mucoepidermoid Carcinoma

This cancer consists of glandular and squamous elements; however, the mixture is more pronounced than in adenosquamous carcinoma. Found in the middle to distal

esophagus, *mucoepidermoid carcinomas* probably arise from submucosal glands or their ducts and carry a dismal prognosis.

Melanoma

Melanoma is a rare esophageal cancer that can be primary or metastatic, although the esophagus is less commonly a metastatic target than the stomach, small intestine, or colon. [364], [365], [366], [367], [368], [369] and [370] The mean age of patients is 60 years, and there are more cases in Japan than in other countries. Melanomas are polypoid and may be multiple, with a propensity for lateral spread. Primary esophageal melanomas arise from melanocytes in the basal layer. There must not be evidence of melanoma in the skin or internal organs to call an esophageal melanoma a primary tumor. Additionally, in situ or junctional melanoma must be evident in the mucosa adjacent to the tumor. Some cases are amelanotic and may require confirmation with immunohistochemical demonstration of S100 protein or homatropine methylbromide 45; alternatively, electron microscopy may reveal premelanosomes. Overall, the prognosis of malignant melanoma is dismal; most patients succumb to the disease within 2 years.

Neuroendocrine Tumors

Small cell carcinoma of the esophagus may include a small cell variant of SCC as well as oat cell carcinoma. [371], [372], [373] and [374] Oat cell carcinoma of the esophagus can be primary or secondary, but it is rare in either situation. A disease of the elderly who present with dysphagia, primary small cell carcinoma of the esophagus (typically distal and sometimes multiple) carries a dismal prognosis, with an average survival of 4.7 months. These tumors may occasionally secrete ectopic antidiuretic hormone or calcium. Microscopically, this tumor is an anaplastic small cell carcinoma resembling what is observed in the lung, with solid sheets of small anaplastic cells with mitoses. There can be evidence of squamous or glandular differentiation. Neuroendocrine markers employed are chromogranin and synaptophysin, as well as electron microscopic evidence of neurosecretory granules.

Esophageal carcinoid tumors are rare [375], [376], [377] and [378]; most occur at the gastroesophageal junction. Carcinoid syndrome has not been reported. There is little or no malignant potential, and, consequently, prognosis is very good.

Choriocarcinoma of the esophagus is an exceedingly rare tumor that may be primary or an adenocarcinoma with trophoblastic differentiation. [379], [380] Secretion of human chorionic gonadotropin has been reported.

BENIGN NONEPITHELIAL TUMORS

Leiomyoma

Leiomyomas are the most common benign esophageal tumors. [381], [382], [383], [384], [385] and [386] Men are affected more frequently than women, with a ratio of 2:1. These tumors typically are single, and most occur in the distal esophagus and in the inner circular layer of the muscularis propria. Typically, most leiomyomas are not associated with symptoms and are found incidentally. Symptoms, if present, include dysphagia and chest pain; gastrointestinal bleeding is rare.

The diagnosis is usually suggested by barium esophagography, which shows a smooth, round defect with sharp margins (Fig. 62-9), or endoscopy, in which there is a well-circumscribed, rounded mass with normal overlying mucosa. Biopsy is usually not necessary because of its nondiagnostic nature and also because of the potential for complication.

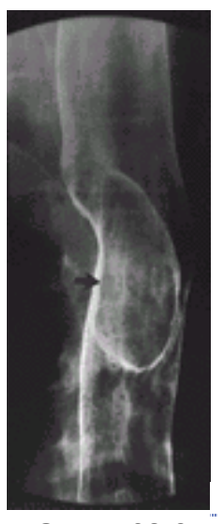

FIGURE 62-9. Leiomyoma of the esophagus represented by a smooth intramural mass outlined with air-contrast barium. (Courtesy of Deborah Hall, MD.)

Granular Cell Tumor

Granular cell tumors are derived from neural or Schwann cell elements, as evidenced by positive staining for S100 protein and neuron-specific enolase. Endoscopically, it is characterized as a smooth, sessile polyp with overlying normal mucosa. [387] Most tumors occur in the distal esophagus and may be single or sometimes multiple. [387] The tumors have an endoscopic appearance of being small, sessile, and intramural nodules or plaques. Patients are typically asymptomatic. Malignant transformation is rare. Microscopically, there are sheets of monomorphic histiocyte-like cells.

Fibrovascular Polyp

Intralumenal *fibrovascular polyps* of the esophagus are rare tumors that may grow and may prolapse into the larynx. Other symptoms include dysphagia, nausea, vomiting, and, occasionally, gastrointestinal bleeding from ulceration. The polyps are covered by a smooth mucosa and are composed of fibrous and vascular tissue, although adipose tissue may occasionally predominate, prompting the designation of a fibrolipoma or pedunculated lipoma. Fibrovascular polyps do not have any malignant potential.

Hemangioma

Hemangiomas are small, asymptomatic lesions found incidentally. [388]

Lymphangioma

Lymphangiomas are rare, with an appearance of a translucent, easily compressed mass on endoscopy. [389] Histologically, they contain dilated endothelial spaces.

Lipoma and Fibroma

Sessile submucosal nodules composed of adipose or fibrous tissue are called *lipomas* and *fibromas,* respectively. In aggregate, they constitute fewer than 5% of all benign esophageal tumors.

MALIGNANT NONEPITHELIAL TUMORS

Leiomyosarcoma and Other Sarcomas

Leiomyosarcoma is an uncommon tumor of the esophagus. [390], [391], [392] and [393] These tumors may be polypoid or infiltrative, and they can occur in all segments of the esophagus. Other esophageal sarcomas include soft tissue sarcoma, rhabdomyosarcoma, neurogenic sarcoma, and Kaposi sarcoma.

Metastatic Carcinoma

Metastatic carcinoma to the esophagus is unusual, representing 3% of esophageal malignancies in an autopsy series. [394] The most common metastatic tumor to the esophagus is melanoma, followed by breast cancer and, less commonly, gastric, renal, liver, prostate, testicular, bone, skin, lung, and head and neck cancer. Primary esophageal and head and neck squamous cancers may coexist or may develop sequentially. Metastatic carcinomas to the esophagus cause extrinsic compression. In this context, the esophageal stricture is extrinsic, without mucosal involvement.

Lymphoma

Primary esophageal *lymphoma* may be Hodgkin's or non-Hodgkin's type, [395], [396], [397] and [398] although, clearly, primary gastrointestinal lymphomas more commonly affect the stomach and small intestine. Symptomatic esophageal lymphoma may be present in immunocompromised patients, as in those with acquired immunodeficiency syndrome. Alternatively, lymphoma of the esophagus may be attributable to disseminated disease or infiltration from adjacent lymph nodes.

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CHAPTER 63

Douglas O. Faigel

MISCELLANEOUS DISEASES OF THE ESOPHAGUS: SYSTEMIC, DERMATOLOGIC DISEASE, FOREIGN BODIES AND PHYSICAL INJURY

FOREIGN BODIES

Risk Factors

The esophagus is the most common site of *foreign body impaction* in the gastrointestinal (GI) tract, accounting for 75% of all impactions (see Chapter 138). [1] Most cases occur in pediatric patients between the ages of 6 months and 3 years who ingest coins, toys, crayons, buttons, and batteries. [1], [3], [4], [5] and [6] In adults, those who wear dentures, prisoners (seeking secondary gain), those with mental retardation, alcohol and drug abusers, and psychiatric patients are at increased risk, and most often they present with meat and bone impactions. [1], [2], [3], [4], [5] and [6] Multiple foreign bodies may be present, and recurrence occurs in 3% to 10% of patients. [1] Typically, these items become lodged in areas of esophageal narrowing: the cervical esophagus (cricopharyngeus), aortic arch, distal esophagus just above the gastroesophageal junction, or any area of structural abnormality. Most patients with food impactions have an underlying esophageal disorder such as a Schatzki ring (Fig. 63-1; see Color Fig. 63-1.). [4] Esophageal bezoars have been described in elderly patients with esophageal dysmotility or in those receiving tube feedings and sucralfate. [7], [8] and [9]

FIGURE 63-1. (See Color Fig. 63-1.) Food impaction in the distal esophagus (**A**). When cleared by advancing the bolus into the stomach (**B**), a Schatzki ring is evident (**C**).

Diagnosis

A careful history usually provides the diagnosis. Most older children and fully conscious adults are able to recognize the ingestion at the time of the incident and convey this information to the physician. Patients usually complain of a sharp pain in the neck or chest, and they may be able to localize the level of obstruction, although this is not always reliable. [10] Dysphagia is the most common symptom, followed by odynophagia, choking, and drooling. [2] Respiratory symptoms (e.g., coughing, dyspnea, wheezing, stridor) occur in 5% to 15% and are primarily seen in children with foreign objects lodged proximally [3], [5], [11] A history of prior impactions is common. [1] Rarely, the diagnosis may be delayed for months or even years. [12]

The physical examination is often unremarkable. Neck swelling, erythema, tenderness, or crepitus are signs of oropharyngeal or proximal esophageal perforation. Radiographs of the neck and chest may reveal a radiopaque foreign object, whereas subcutaneous air or soft tissue swelling suggests the presence of a radiolucent object. [2], [13] A contrast examination should not be routinely performed when high-grade obstruction is suspected, owing to the risk of aspiration. Furthermore, opaque contrast agents such as barium coat the foreign body and esophagus and hamper subsequent endoscopy, whereas Gastrografin, a transparent contrast agent, causes severe pneumonitis if it is aspirated. [14] Metal detectors have been used to detect metallic foreign bodies and may be useful as a screening tool in the pediatric population. [15], [16]

Complications

Most ingested foreign bodies pass through the GI tract uneventfully. Complications depend on the size, shape, presence of sharp points, and location of the object. Esophageal perforation may cause mediastinitis and abscess formation. [17], [18] Hemorrhage, which may be life-threatening, occasionally occurs and, when massive, suggests the presence of damage or fistulization to surrounding vasculature such as the aorta or innominate artery. [19], [20] and [21] Long-standing foreign bodies in the esophagus may induce cricoid perichondritis, periesophagitis, esophageal diverticula, or esophageal stenosis. [22] Fistulization to the airway and pericardium have also been reported. [23], [24] Disc batteries are particularly noxious, because of direct pressure, leakage of alkali, and generation of electrical current, which causes liquefaction necrosis and perforation. [25], [26]

Management

Treatment of an esophageal foreign body depends on the object's physical characteristics, its location, and the length of time it has been in place. In all cases, the risk of pulmonary aspiration and airway obstruction must be minimized. Urgent intervention is indicated for sharp objects, disc batteries, and coins in the proximal esophagus, or when impactions result in high-grade obstruction causing the patient difficulties in handling secretions. [1], [2] Coins in the distal esophagus can be observed for 12 to 24 hours to see whether they will pass into the stomach. Once in the stomach, most objects pass through the remainder of the GI tract without problem. Food impactions or foreign objects should not be allowed to remain in the esophagus for more than 24 hours. [2] Flexible endoscopy performed by an experienced endoscopist forms the mainstay of therapy and has a success rate in foreign object removal of up to 98% and very low morbidity. [1], [14], [27] Nonendoscopic removal of blunt radiopaque esophageal foreign objects has been accomplished by use of a Foley catheter under fluoroscopic guidance, but it provides no control of the object as it is being removed, provides no airway protection, and does not allow for assessment of underlying esophageal disorders. [28] Blind bougienage carries a perforation risk and should be avoided. [2], [29]

Food usually impacts in the distal esophagus and is almost always associated with underlying esophageal disease (see Fig. 63-1). [1], [2] Glucagon, 1 mg intravenously, may relax the esophagus and allow spontaneous passage of the bolus, [30] but it does not obviate the need for subsequent esophageal examination. The use of effervescent agents is of limited efficacy, carries a perforation risk, and interferes with subsequent endoscopy. [31], [32] The use of enzymatic digestion of the meat with papain (Adolph's Meat Tenderizer) is especially discouraged because perforation and death have been associated with its use. [33]

The principles for the endoscopic management of impacted foreign objects and food boluses are similar. Successful removal requires the availability of a variety of endoscopic instruments employed by an experienced endoscopist. [34] An esophageal overtube is usually employed to protect the airway. An ex vivo trial with a similar foreign body helps to determine which endoscopic accessories would be most useful in the actual removal. Coins are most easily removed with grasping forceps, a polypectomy snare, or a retrieval basket. [35] Difficult to remove smooth, round objects, such as disc batteries, are most easily removed by capturing them with a polypectomy snare fitted with a net (Roth Retrieval Net, U.S. Endoscopy Group, Inc., Mentor, OH) [34] Advancing small (<2.0 cm), smooth objects under direct vision into the stomach can aid in their retrieval. [1] Endoscopic retrieval of sharp objects is accomplished with use of retrieval forceps or a polypectomy snare. [34] The risk of mucosal injury during sharp object retrieval can be minimized by orienting the object with point trailing during extraction, using an overtube, or fitting a protector hood to the end of the endoscope. [2], [36] In managing food bolus impactions, it is sometimes possible to distend the esophageal lumen with air and gently push the bolus into the stomach. [37], [38] Otherwise, by using a variety of snares, baskets, graspers, and forceps, the food bolus can usually be removed as a whole or in piecemeal fashion through an esophageal overtube. [2] A friction-fit adaptor fitted to the end of the endoscope can also be used as a direct-vision suction device to remove the impacted food. [39] The underlying esophageal stricture may be dilated at the time of the disimpaction, provided bolus removal has not been delayed and severe esophagitis is not present. [1], [37], [38]

Surgery is rarely needed to remove an impacted object or food bolus. [1] It is generally performed in patients with large objects in whom endoscopic management has failed, when the object has become embedded in the esophageal wall, or when there has been perforation. [1], [2], [21], [40] Objects in the high cervical esophagus or pharynx may require rigid laryngoscopy for removal. [4]

SYSTEMIC DISEASES

Sarcoidosis

Sarcoidosis is a systemic granulomatous condition that rarely involves the GI tract (2 of 1254 cases in one series). [41] Granulomata may be found in the submucosa of the GI tracts in 10% of patients with known sarcoidosis, but this is rarely associated with symptoms. [41] Granulomatous esophagitis usually presents in conjunction with generalized sarcoidosis.

Dysphagia, the most common clinical manifestation of esophageal sarcoidosis, may be caused by extrinsic esophageal compression from enlarged lymph nodes, an achalasia-like dysmotility syndrome, or stricture formation caused by granulomatous esophagitis, and it may resolve with steroid treatment. [42], [43] and [44] It may be difficult to differentiate esophageal sarcoidosis from Crohn's disease because both are characterized by the presence of granulomata. [45]

Therapy is aimed at the systemic disease and typically includes the use of corticosteroids. Esophageal strictures may require dilation, and rarely, esophagojejunal interposition. [44]

Crohn's Disease

Crohn's disease of the esophagus is rarely encountered, with one series reporting only one case of Crohn's esophagitis among 383 patients. [46] In a 4-year study of 500 patients with Crohn's disease, only nine had esophageal involvement. [47] The true prevalence may be higher, as suggested by one study of 41 consecutive children, 16 of whom were found to have esophagitis and 2 had esophageal ulcers. [48] Additionally, in 33% of patients with Crohn's disease, there is abnormal expression of HLA-DR in the esophageal mucosa associated with increased epithelial content of T cells, B cells, natural killer cells, and macrophages, even though clinical Crohn's esophagitis may be absent. [49]

Patients with Crohn's disease with esophagitis almost invariably have active disease elsewhere in the GI tract. Extraintestinal manifestations are common, and the degree of inflammation usually parallels the activity in the other involved segments. [50], [51] Dysphagia has been the presenting symptom of Crohn's disease, and isolated esophageal involvement without disease elsewhere has been reported. [51], [52] and [53] Esophageal epidermolysis bullosa acquisita, a rare autoimmune subepidermal bullous disorder, is associated with Crohn's disease, and in one case it responded to treatment of the intestinal disease with sulfasalazine. [54]

Patients with Crohn's esophagitis complain of dysphagia, odynophagia, pyrosis, or substernal chest pain. [47], [51] Perforation and fistulization to the bronchi, mediastinum, pleura or stomach may occur. [52], [55], [56] Barium radiography may reveal a variety of lesions. Aphthous ulceration is present early in the course of the disease but is nonspecific. Later in the course, cobblestoning and deep linear ulcerations may be present, along with decreased motility and distensibility (Fig. 63-2). There may be esophageal stricturing and the formation of sinus tracts, fistulae, or mucosal bridges. [50], [51], [57]

FIGURE 63-2. Crohn's esophagitis. Filiform polyps (*white arrowheads*) are associated with mucosal nodularity, deep ulcers, and intramural sinus tracts (*black arrowheads*). (From Eisenberg RL. Gastrointestinal radiology: a pattern approach. 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003).

Endoscopy with biopsy is helpful in delineating the degree of inflammation and in ruling out other causes of esophagitis. [50] Endoscopic biopsies are not able to demonstrate the transmural nature of the inflammation, however, and granulomata are rarely found. [47], [51] Endoscopic ultrasound may have a role in diagnosing esophageal Crohn's disease by demonstrating the transmural nature of the inflammatory process. [58]

Most patients respond to treatment with corticosteroids and have resolution of their symptoms with healing of the esophageal lesions in 2 to 4 weeks. [47], [50], [51] At least half of patients will then remain free of both esophageal lesions and upper GI symptoms, whereas the others will have either persistent lesions or recurrence after initial healing. [50] Progressive stricturing may respond to medical management and bougie dilation. Extensive mucosal bridges may be endoscopically excised with a papillotomy knife. [57] Medical therapy may be improved by the placement of a percutaneous endoscopic gastrostomy to provide nutrition and aid in healing. [99] Surgical resection may be required for the treatment of fistulae, extensive refractory strictures, and perforation, and for when malignancy cannot be excluded. [53]

Behçet Syndrome

Behçet syndrome is characterized by oral and genital aphthous ulcers and ocular inflammation, and it may have multiorgan involvement including the GI tract. [60] Rare in northern Europeans, Behçet syndrome is most common in the eastern Mediterranean, Middle East, and East Asian populations, primarily affecting young adults. [61]

Esophageal Behçet syndrome has been reported since 1973. [62], [63], [64] and [65] Patients may present with dysphagia, odynophagia, chest pain, and hematemesis. Lesions found include superficial erosions, diffuse esophagitis, perforated ulcers with mediastinal abscess, and esophageal strictures. Most of the lesions have been found in the middle and distal esophagus. All these patients had other concomitant manifestations of Behçet syndrome. Barium esophagrams and endoscopy in symptomatic patients may reveal the mucosal lesions, aphthous ulcerations, perforations, or strictures. [63] Biopsies show ulceration with nonspecific inflammation and neutrophilic infiltration. [60]

The clinical course parallels that of the multisystem disease with unpredictable exacerbations and remissions. Dysphagia caused by esophageal ulcers may resolve spontaneously. [60] Patients generally require systemic immunosuppression to prevent blindness from uveitis or death from cerebral vasculitis. [61] Corticosteroids may markedly improve the symptoms of esophageal involvement, and the intestinal and esophageal ulcers have been successfully treated with sulfasalazine and low-dose cyclosporine. [64], [65]

Graft-Versus-Host Disease

Graft-versus-host disease (GVHD) occurs after successful bone marrow implantation when immunologically competent cells from the grafted marrow target antigens in the recipient. It can occur in both acute and chronic forms. It also rarely occurs after a blood transfusion in an immunologically impaired patient. GVHD is primarily manifested by skin rash, liver test abnormalities, and GI involvement usually presenting as diarrhea. [66]

Esophageal involvement is relatively uncommon and is primarily seen in patients with chronic GVHD who have multisystemic disease. [67], [68] Upper GI involvement may occur in the absence of lower GI GVHD but is uncommon. [69], [70] Patients with GVHD esophagitis complain of odynophagia, dysphagia, pyrosis, and retrosternal chest pain.

GVHD esophagitis needs to be differentiated from opportunistic infection of the esophagus such as with herpesvirus, cytomegalovirus, and *Candida,* and endoscopy with biopsy should be performed. [70] In patients with confirmed GVHD esophagitis, the esophageal mucosa has appeared erythematous and friable, and strictures, webs, bullae, or an esophageal cast have been noted (Fig. 63-3). [68], [71], [72]

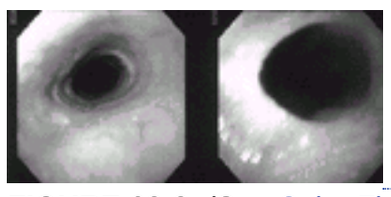

FIGURE 63-3. (See Color Fig. 63-3.) Graft-versus-host disease. Multiple fine mucosal webs are present in the esophagus.

Therapy consists of controlling the underlying immunologically mediated inflammatory process with the use of immunosuppressants such as prednisone, azathioprine, and cyclosporine. [66] Esophageal symptoms may respond to antireflux medications and to dilation of webs and strictures, although the risk of iatrogenic perforation may be increased in these patients. [67] Strictures and webs usually recur unless the underlying inflammatory process is controlled.

Eosinophilic Esophagitis

Idiopathic eosinophilic esophagitis (IEE) may occur alone or in conjunction with idiopathic eosinophilic gastritis. [73] In IEE, there is full-thickness infiltration of the esophagus by eosinophils. Most patients are male, with onset in childhood or early adulthood, and have an allergic disorder or peripheral blood eosinophilia, suggesting that the condition is a hypersensitivity reaction. [74] Patients complain of dysphagia or chest pain. [75], [76] Proximal esophageal stricture is the most common abnormality, followed by distal stricture and abnormal motility. Multiple esophageal rings have been noted. [77], [79] One case of esophageal rupture has been reported. [80] Skin testing may document an underlying food allergy, and patients may respond to elimination diets. [74], [76] IEE responds well to corticosteroids at a dosage of 1.5 mg/kg for 4 weeks, but topical treatment with inhaled steroids has also been described. [78], [79] Reflux esophagitis, which can also have eosinophilic infiltration noted on biopsy, may be differentiated from IEE by a more marked infiltration of eosinophils (5 to 20/high power field); on electron microscopy, the eosinophils demonstrate signs of activation, lack of adequate response to gastroesophageal reflux disease therapy, and normal esophageal pH testing. [74], [81], [82], [83] and [84] Additional causes of esophageal eosinophilic infiltration include parasitic and fungal infections, systemic connective tissue disorders, esophageal leiomyomatosis, inflammatory bowel disease, lymphomas and myeloproliferative disorders, vasculitis, and food allergy. [85]

Miscellaneous Autoimmune Diseases

Several other systemic autoimmune diseases rarely affect the esophagus. In *Sjögren syndrome,* more than half of affected patients complain of dysphagia, which does not appear to correlate with the degree of xerostomia. [86] Ten percent have upper esophageal webs. [87] Dysphagia may [88] or may not [89] correlate with the presence of peristaltic abnormalities on esophageal manometry.

Amyloidosis, either primary or secondary, may cause dysphagia. Amyloid deposition in the esophageal muscle or nerves produces motility abnormalities that may mimic achalasia and respond to pneumatic dilation. [90], [92] Amyloid deposition may predispose to esophageal rupture. [93]

The *anticardiolipin antibody syndrome* is associated with numerous thromboembolic phenomenon. Esophageal involvement includes ischemic necrosis from esophageal vascular thrombosis (which may perforate), and the formation of varices caused by portal vein thrombosis. [94], [95] and [96]

DERMATOLOGIC DISEASES

Pemphigus Vulgaris

Diseases that affect the squamous epithelium of the skin also affect the esophagus (Table 63-1). *Pemphigus vulgaris* is a chronic blistering autoimmune disease of older adults manifested by flaccid intraepidermal blisters and intraoral erosions caused by IgG autoantibodies directed against desmoglein 3 in keratinocyte membranes and keratinocyte acetylcholine receptors. [97], [98] and [99]

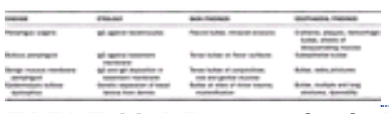

TABLE 63-1 Dermatologic Diseases with Esophageal Manifestations

Patients may be symptomatic, but at least 50% have esophageal involvement even if there are no symptoms. [100] Isolated esophageal involvement with no or minimal skin manifestations has rarely been reported. [101], [102] and [103] Endoscopy with biopsy is necessary to establish the diagnosis and to rule out other causes of esophageal symptoms (see Table 63-1). Complete sloughing of the esophageal mucosa (esophagitis dissecans superficialis) has rarely been reported. [104] Diagnosis is made by immunofluorescence microscopy, but this does not differentiate between idiopathic pemphigus vulgaris and drug-induced pemphigus. [99], [104], [105] Serologic testing for pemphigus antibodies and antibodies against desmoglein 3 support the diagnosis. [106]

Treatment of pemphigus vulgaris relies on the use of high-dose corticosteroids, but other immunosuppressive drugs (azathioprine, cyclophosphamide, cyclosporine, methotrexate) may be necessary to achieve a complete remission or as a steroid-sparing agent. [99] Plasmapheresis has been used successfully in severe cases. [99] A medication history should be sought for drugs associated with pemphigus, especially thiol compounds (e.g., penicillamine), angiotensin-converting enzyme inhibitors, and antibiotics (penicillins, rifampin, cephalosporins). [102]

Bullous Pemphigoid

Bullous pemphigoid is distinguished from pemphigus vulgaris by the presence of tense bullae with a predilection for the flexor surfaces. [99] Involvement of the oropharynx occurs in 20%. [107] The incidence of esophageal involvement in bullous pemphigoid is unknown and is likely underestimated because symptoms are infrequent, even in the presence of active disease. [107] The formation of bullae is the result of IgG autoantibodies directed against the basement membrane of the squamous epithelium and activation of the complement cascade. [99], [108] Immunofluorescence microscopy demonstrates linear deposits of IgG or C3 along the basement membrane of the involved epithelium. [108]

Esophageal involvement with bullous pemphigoid is usually asymptomatic, although rarely dysphagia and odynophagia occur. Massive upper GI hemorrhage has been reported. [109] Scarring, mucosal sloughing, and hemorrhage are rare. Treatment with prednisone is highly effective, but azathioprine, cyclophosphamide, cyclosporine, or methotrexate are sometimes required. [99]

Benign Mucous Membrane Pemphigoid

Benign mucous membrane pemphigoid (BMMP), also known as cicatricial pemphigoid, is a chronic blistering disease of the conjunctivae, oral mucosa, genital mucosa, and adjacent areas of skin. It occurs in late adulthood, usually after the sixth decade, producing tense blisters with scarring and strictures of the esophagus, trachea, anus, or vagina. [110] The pathogenesis of BMMP is idiopathic autoimmune deposition of IgG or IgA autoantibodies in the basement membrane zone, with specific antigens recently identified. [99], [111], [112]

Esophageal involvement in BMMP occurs in 2.3% to 13% of affected patients and can be seen initially or up to 10 years later in the course of the disease. Patients complain of dysphagia, odynophagia, and chronic cough resulting from aspiration. [113] Rarely, patients have been found with isolated esophageal BMMP with no other manifestations or with active esophageal disease without symptoms. [114] Esophagrams may show bullae as seen in bullous pemphigoid and epidermolysis bullosa dystrophica. More typical is the presence of webs and strictures, primarily of the cervical esophagus but also of the middle and lower esophagus (Fig. 63-4 and Fig. 63-5; see Color Fig. 63-5.). [115] Secondary manifestations include nasopharyngeal reflux, tracheal aspiration, and intramural pseudodiverticulosis. [115] Bullae are rarely seen endoscopically, but in one case endoscopy induced the formation of bullae. [114] Severe dysphagia may require dilation, which carries an increased risk of esophageal injury in these patients. [115] The medical management of severe BMMP requires prednisone, whereas milder cases respond to treatment with dapsone. [99] Surgical treatment has rarely been required. [116]

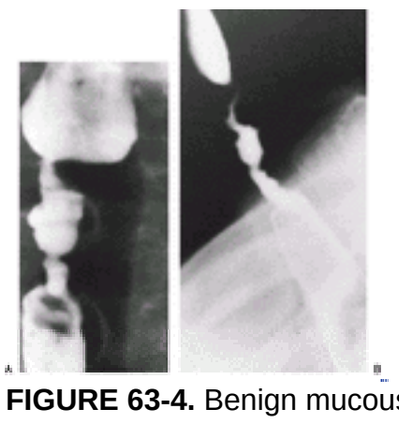

FIGURE 63-4. Benign mucous membrane pemphigoid. Postinflammatory scarring caused a long, irregular area of narrowing suggestive of a malignant process. (From Eisenberg RL. Gastrointestinal radiology: a pattern approach. 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003).

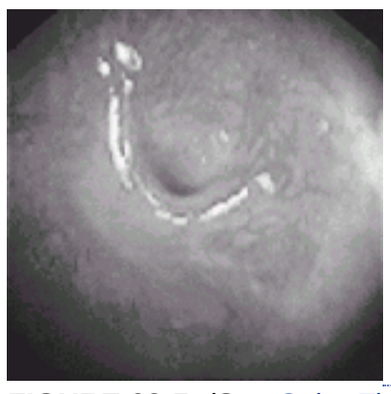

FIGURE 63-5. (See Color Fig. 63-5.) Benign mucous membrane pemphigoid. Also known as cicatricial pemphigoid, this endoscopic photograph depicts a tight esophageal stricture.

Epidermolysis Bullosa Dystrophica

Epidermolysis bullosa dystrophica is a rare inherited blistering condition primarily affecting squamous epithelium. Both dominant and recessive forms of the disease exist. Esophageal involvement is clinically important in the recessive form and is rarely seen in the dominantly inherited form. [117] The disease is produced by mutations in the collagen type VII gene (*COL7A1*). [118], [119] and [120]

Beginning in early childhood, cutaneous bullae are evident and form at sites of minor trauma. Histopathologically, there is a separation of the basal lamina from the underlying dermis. [121] This is caused by a marked reduction or absence of the anchoring fibrils that are normally present between the lamina densa and dermis. [121] The pathogenesis appears to be disordered transport or assembly of type VII collagen, which is absent from the anchoring fibrils in affected patients. [122] Healing is by scar formation. Over time, there are loss of hair and nails and formation of contractures and syndactyly by the scarring and healing process, a process termed *mummification.*

The same bullae formation and healing with scar formation occur in the esophagus as a result of the trauma of mastication and swallowing. The esophagus is involved primarily at the upper esophageal sphincter, carina, and distal esophagus. [123] Strictures, which are often multiple, are observed at all these sites, along with esophageal shortening and motility abnormalities. [123], [124] Dysphagia, pain with eating, and food impaction are the main symptoms in these patients and result from the strictures, bullae, or impacted food. [123], [124] Malnourishment is common, particularly in younger patients, and may necessitate the use of parenteral nutrition.

Owing to the striking cutaneous manifestations, the diagnosis of epidermolysis bullosa dystrophica is rarely difficult. Endoscopy should not be done in uncomplicated cases because of the risk of traumatic bullae formation. Barium esophagraphy is the test of choice and usually reveals typical strictures or webs (Fig. 63-6). [123], [124]

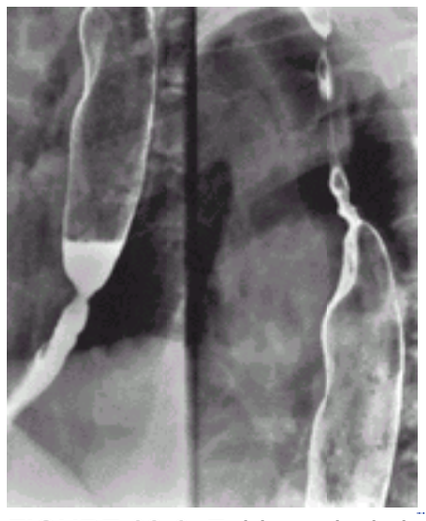

FIGURE 63-6. Epidermolysis bullosa dystrophica. Esophagram in patient with a recessive form of the disease demonstrating strictures of the proximal and distal esophagus.

The prognosis of esophageal involvement in epidermolysis bullosa dystrophica is poor. [123] Complications include esophageal perforation (which may be spontaneous or a result of therapeutic esophageal dilation) and complete occlusion. [123], [124] Patients should avoid coarse or hot foods. A pureed or nutritionally complete liquid diet may be necessary to avoid malnutrition. Medical management with corticosteroids or phenytoin is often used, but treatment failures are not uncommon. [123], [124], [126] Careful dilation of strictures with balloons or bougies may be successful, but there is an increased risk of esophageal trauma and perforation. [123], [127] In children, balloon dilation and intensive nutritional support may successfully relieve dysphagia and may lead to weight gain and overall clinical improvement. [128], [129] In patients with severe esophageal stenosis causing malnutrition or with esophageal perforation, esophagectomy and esophageal replacement, usually with colon interposition, should be performed, which improves dysphagia and nutritional status. [123], [126], [130]

An acquired form of epidermolysis bullosa (*epidermolysis bullosa acquisita*) may cause nonscarring bullous lesions of the esophagus. It is likely an autoimmune disease and has been seen in association with autoimmune thrombocytopenia and hypothyroidism and with Crohn's disease. [54], [131] In one case, circulating IgG anti-type VII collagen antibodies were identified. [132] Treatment is with steroids and dapsone. [131]

ESOPHAGEAL TRAUMA

Mallory-Weiss Syndrome

In 1929, Mallory and Weiss described patients with alcoholism who had retching and emesis followed by massive, in some cases fatal, upper GI hemorrhage caused by linear lacerations at the gastroesophageal junction. [133] Termed *Mallory-Weiss tears,* these lacerations account for 5% to 15% of upper GI bleeding episodes. [134], [135] and [136] The tears are nonpenetrating mucosal lacerations, 3 to 20 mm long and 2 to 3 mm wide, and are oriented along the long axis of the esophagus (Fig. 63-7; see Color Fig. 63-7). [137]

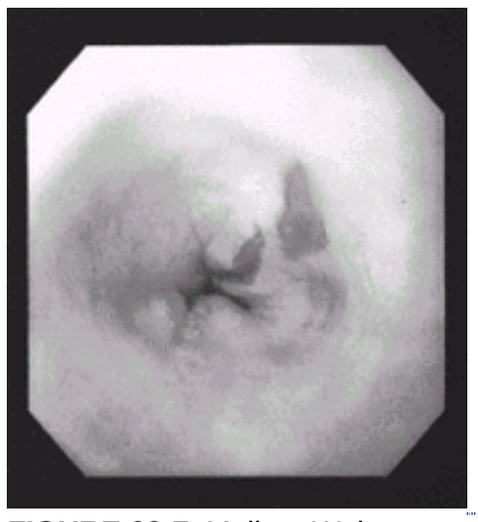

FIGURE 63-7. Mallory-Weiss tear. A nonpenetrating mucosal laceration occurred as a result of retching in a 53-year-old woman.

The pathogenesis of Mallory-Weiss tears involves the sudden rise of intra-abdominal pressure such as seen in vomiting, retching, and coughing. They are most commonly associated with alcoholic binges, but they may occur after retching or vomiting for any reason including the ingestion of emetogenic chemotherapeutic agents [135] or medical conditions such as diabetic ketoacidosis. [138] Mallory-Weiss tears may also occur as a result of seizures, hiccups under anesthesia, blunt trauma, straining at stool, asthma, heavy lifting, cardiopulmonary resuscitation, primal scream therapy, changes in air pressure, and complications of carbon monoxide poisoning. [140], [142] Mallory-Weiss tears rarely complicate upper endoscopy (0.5% of all endoscopies) and occur in patients with excessive retching or struggling during the procedure. [144] and [145] Nausea and vomiting from polyethylene glycol electrolyte lavage solutions used in preparation for colonoscopy have also been associated with Mallory-Weiss tears. [146], [147]

Hiatal hernias are found in a large proportion of patients with Mallory-Weiss tears and may be a risk factor. When the gastroesophageal junction is located above the diaphragm, a large pressure gradient between the intragastric and intrathoracic compartments may result in sudden dilation and laceration of the mucosa at the gastroesophageal junction. [137] An alternative hypothesis is that the tears are the result of prolapse of the stomach into the esophagus during retching. [137]

Patients with Mallory-Weiss tears present with upper GI bleeding. The classic presentation is of repeated, violent retching, vomiting, or coughing followed by hematemesis, although 40% of patients have no antecedent explanation for the tear, and 50% have hematemesis on first emesis. [135], [137] The syndrome can occur at any age, with a mean age at presentation in the 40s. [137], [139], [143] Male patients are affected more frequently (4:1) than female patients. [135], [137] Spontaneous cessation of bleeding is seen in 90%. [185], [186] However, a need for blood transfusion, endoscopic intervention, and surgery and up to a 12% mortality have been reported. [134], [135] Patients with portal hypertension and coagulation defects are at increased risk of bleeding from Mallory-Weiss tears and may have a poorer prognosis. [134], [148], [190] Twenty-seven percent of patients have multiple lacerations, and up to 77% have other associated upper GI lesions. [134], [135]

Endoscopy is the preferred modality for diagnosing and treating actively bleeding Mallory-Weiss tears. Endoscopic hemostasis has been achieved by the injection of anhydrous ethanol, polidocanol, or 1:10,000 epinephrine, cautery with multipolar coagulation or heater probe, or endoscopic band ligation. [135], [150], [151], [152], [153] and [154] Esophageal balloon tamponade may be successful but carries the considerable risk of extending the tear or rupturing the hiatal hernia. [136], [149] Surgery is occasionally required when bleeding cannot be controlled. [135] Vasopressin administered either systemically or by direct infusion into the celiac or left gastric artery has, in a few cases, been reported to control hemorrhage. [155], [156] Angiographic embolization has been used with success. [157] The long-term prognosis has not been well studied, but recurrences have been noted and may be related to continued alcohol abuse. [133], [135]

Esophageal Intramural Hematomas

Esophageal intramural hematoma (also termed esophageal apoplexy) is a rare but well-recognized clinical entity consisting of hematomas in the wall of the esophagus produced by local trauma. [158], [159], [160] and [161] Similar to Mallory-Weiss tears and Boerhaave syndrome, they typically occur in the setting of vomiting or retching, although spontaneous hematomas have occurred, particularly in patients with coagulopathies or undergoing treatment with anticoagulants or aspirin. [158], [159], [160], [161], [162], [163], [164] and [165] Iatrogenic hematomas have occurred after endoscopic sclerotherapy and cardioversion. [166], [167]

The pathogenesis is most likely mechanical injury to the esophageal wall from internal pressure (retching, coughing, or sneezing) or from blunt external trauma. [158], [159], [160] In several cases, there has been suspicion of esophageal damage from ingestion of a medication or foreign body. [168], [169] A history of a recent meal is also frequently noted. [170], [171] In most cases, an identifiable mucosal tear is present. [160]

Patients complain of the abrupt onset of substernal or epigastric pain followed by hematemesis. Dysphagia is present, and swallowing aggravates the pain. [158], [162], [168], [170] The amount of bleeding is usually not large, but up to 10% of patients require blood transfusions. [161], [170] In some cases, the degree of esophageal obstruction can be pronounced. [172]

Patients presenting with hematemesis and substernal chest pain should first undergo a chest radiograph to rule out perforation and then endoscopy. Typically, an esophageal mass with bluish or violet color protruding into the lumen is seen, associated with a mucosal tear. [160], [173], [174] Varying degrees of lumenal narrowing are present that can include complete esophageal obstruction. [172] An esophagram with a water-soluble contrast agent should be performed to rule out esophageal perforation before endoscopy. [172] The esophagram shows a filling defect in the esophagus as the most common abnormality, but leiomyoma, carcinoma, and metastatic disease can have similar radiographic appearances. [175] Computed tomography with contrast or magnetic resonance imaging may reveal a nonenhancing esophageal mass that has the density of blood. [163], [165], [168], [174], [175] and [176] Endoscopic ultrasound shows an intramural hypoechoic submucosal mass. [174]

Most patients respond well to conservative management and have complete resolution in 2 to 3 weeks with no long-term sequelae. [170] Aspiration precautions should be observed, but parenteral nutrition is rarely needed because most patients are able to swallow within a few days. Blood transfusion is sometimes required. Surgery is needed in 19% of cases to control bleeding or repair perforations. [170] Patients should be monitored for the development of fever or pleural effusions indicating an esophageal perforation not detected during the initial evaluation. [175]

Esophageal Injury and Rupture

Esophageal perforations and ruptures are life-threatening injuries that require prompt surgical intervention. Perforation is most commonly iatrogenic after instrumentation of the esophagus, but it may be secondary to external trauma or may occur spontaneously (Boerhaave syndrome). Esophageal rupture affects all age groups, with a mean age of 28 to 43 years, and it lacks a gender or race predominance. [177] Although diagnosis may sometimes be difficult, successful management depends on early diagnosis and intervention.

Medical instrumentation is the most common cause of esophageal perforation, accounting for at least half of all cases. [177], [178] and [179] Iatrogenic rupture is a well-recognized complication of rigid and flexible endoscopy and of therapeutic esophageal dilation. [177] It is most commonly seen after bougie or balloon dilation of benign and malignant strictures or for treatment of achalasia. [180], [181], [182], [183] and [184] Perforation has also been seen after placement of nasogastric tubes, erroneous inflation of a gastric balloon in the esophagus, sclerotherapy, and balloon tamponade for bleeding esophageal varices and as a complication of anesthesia. [177], [185], [186]

Both penetrating and blunt trauma to the neck, chest, and upper abdomen may cause esophageal perforation. Penetrating knife and gunshot wounds in these areas should raise the suspicion of esophageal injury. [187], [188] Blunt thoracoabdominal and cervical trauma may result in perforation of the midthoracic or cervical esophagus (less commonly the distal esophagus), most likely caused by rapid changes of the esophageal transmural pressure and rapid increases of intrathoracic pressure

against a closed glottis. [177], [189], [190], [191] and [192] The degree of associated trauma may be severe, as from blast and crush injuries, or more innocuous, as from assault, child abuse, or the Heimlich maneuver. [189], [190], [191], [192], [193], [194] and [195]

Spontaneous transmural rupture of the esophagus was first described in 1724 by Herman Boerhaave. His patient, Baron von Wassenauer, died suddenly after severe retching and was found at autopsy to have an esophageal rupture and food contaminating the thorax. [196] Boerhaave syndrome was largely a pathological curiosity diagnosed at autopsy until Barrett's first successful surgical repair in 1946. [197] As with Boerhaave's original patient, most patients have antecedent retching and vomiting, although any maneuver that transiently raises the intra-abdominal pressure may result in rupture. [198], [199] Esophageal rupture from vomiting caused by colonoscopy preparation has also been described. [200] Ninety percent of spontaneous ruptures occur in the distal esophagus, with more than two thirds on the left side, where studies have demonstrated an anatomic weakness of the left posterolateral aspect of the esophagus just above the diaphragm. [199], [201] The normal esophagus lacks a serosa, making it more prone to rupture than other parts of the GI tract. [198] In addition, some patients with spontaneous rupture have been found to lack a muscularis mucosae, which may make them anatomically predisposed to Boerhaave syndrome. [202] Recurrent spontaneous esophageal rupture has been reported. [203]

The clinical manifestations of esophageal perforation depend on the extent, acuity, and cause of the rupture. Iatrogenic perforations may remain asymptomatic for up to 8 hours in as many as 50% of patients. [177] Upper esophageal perforations may be accompanied by chest pain, dysphagia, odynophagia, nausea, vomiting, hoarseness, or aphonia. [177] Lower esophageal perforations may also cause abdominal pain, pneumothorax or hydropneumothorax (usually on the left side), and pneumomediastinum. Hematemesis (seldom severe) is noted in 55%. [198], [199], [200], [201], [202], [203] and [204] The classic Mackler triad of vomiting, chest pain, and subcutaneous emphysema in patients with spontaneous rupture should not be relied on, because it is found in only one third of patients with Boerhaave syndrome. [205], [206] and [207] Two patients with early spontaneous esophageal rupture had intense thirst for small sips of cold water, and this may be an early clue to the diagnosis. [208]

On physical examination, patients appear acutely ill with tachypnea and tachycardia. Fever may or may not be present. Sudden massive esophageal rupture may be dramatic, presenting with tension pneumothorax, hypotension, and shock. [205], [207], [209] Subcutaneous emphysema is present in 30% of patients. [210] Auscultation may reveal absence or reduced breath sounds on the side of the perforation if there is pleural involvement. Hamman sign (mediastinal crunch) may be noted in patients with mediastinal air. This mediastinal crackling occurs with each heartbeat and has been likened to the sound of boots walking through dry snow. It is best appreciated with the patient lying in the left lateral decubitus position.

The diagnosis of esophageal rupture requires a high index of suspicion. Early diagnosis and treatment are mandatory to avoid the high mortalities associated with patients presenting beyond 24 hours from the rupture. [204], [207] Misdiagnosis and treatment delays occur in more than 50% of patients with spontaneous rupture. [205], [211] Corticosteroid use may mask the symptoms of esophageal perforation, leading to delays in diagnosis. [212] The differential diagnosis includes pulmonary embolus, myocardial infarction, aortic dissection, Mallory-Weiss laceration, esophageal intramural hematoma, spontaneous pneumothorax, pneumonia, lung abscess, pericarditis, pancreatitis, strangulated diaphragmatic hernia, and perforated peptic ulcer. [198], [213] Chest and abdominal radiographs usually suggest the presence of a perforation by demonstrating subcutaneous emphysema, pneumomediastinum, mediastinal widening or air-fluid level, pleural effusion, pneumothorax, hydropneumothorax, or air in the peritoneal cavity. [198], [199] A completely normal chest radiograph is present in fewer than 5% of patients, although in fewer than one third of patients is the radiograph diagnostic of esophageal rupture. [214] Esophagrams employing water-soluble contrast (e.g., Gastrografin) with multidirectional views are diagnostic in most cases. [177], [198], [199] If these are negative, a barium esophagram should be obtained to provide better definition. If an esophagram cannot be obtained (e.g., uncooperative or severely ill patient), a noncontrast computed tomography scan may be useful in identifying minute amounts of air and oral contrast in the pleural space and in the evaluation of the mediastinum, pleura, and aorta. [198] However, computed tomography does not allow for localization of the exact site of perforation. [215] Flexible upper endoscopy is a highly accurate modality and has been advocated in patients with suspected blunt or penetrating esophageal trauma. [216], [217] Disadvantages of endoscopy include missing small defects and the risk of extending the perforation.

Complications of esophageal perforation include tension pneumothorax and infection. The diagnosis may be delayed for several days until the patient develops fever, mediastinitis, pleural effusion, empyema, or sepsis. Examination of the pleural fluid reveals an exudate with a pH as low as 6.0 owing to a combination of gastric acid refluxed through the defect and the metabolism of leukocytes. [218] The pleural fluid amylase is elevated and may raise the suspicion of pancreatitis with pancreaticopleural fistula. The pleural fluid amylase in esophageal rupture is of salivary origin, and obtaining isoenzyme levels may aid in the diagnosis. [219] Bacterial infection of the pleural fluid, empyema, is common, and both clostridial myonecrosis of the chest, and invasive candidiasis have been reported. [220], [221]

Treatment of both spontaneous rupture (Boerhaave syndrome) and traumatic perforation is primarily surgical. All patients should take nothing by mouth, and broad-spectrum antibiotics should be administered. Aggressive resuscitation and cardiopulmonary support may be needed. Specific therapy depends on the site of the rupture, the cause, the length of time between the perforation and the diagnosis, and the overall health status of the patient. Small cervical perforations can be treated with antibiotics and without surgery or with surgical drainage of the neck without esophageal repair. [222] Free intrathoracic perforations are treated aggressively with debridement and irrigation of the mediastinum, followed by primary closure of the defect. [223], [224] In patients operated on early, the mortality is less than 10%, but this rate triples in those treated more than 24 hours later. [225] Nonetheless, most authors recommend surgery with primary esophageal repair even in patients with a delayed diagnosis. [223], [224], [225], [226], [227] and [228] Children do particularly well, with less than a 5% mortality. [229] Esophagectomy is rarely required. It is primarily indicated when the diagnosis has been delayed and infectious complications have ensued, but it also may be indicated in patients with distal esophageal obstruction who have suffered iatrogenic perforation. [181], [230] Patients with perforation caused by endoscopy with dilation do well, with very low mortalities owing to prompt diagnosis and treatment, although recurrent dysphagia is common. [231] Nonoperative management of thoracic esophageal ruptures may be successful in selected cases. [198], [214], [232], [233] Minimally invasive interventions, such as thorascopic repair of the ruptured esophagus and endoscopic repair of a small dilation-induced perforation using metal clips, have been described. [234], [235] The use of covered expandable metal stents to treat esophageal perforation successfully has been reported. [236], [237], [238], [239] and [240] Although stents may be useful in patients with significant comorbidity (e.g., cancer), they are generally not removable and should not be used in patients who are otherwise operative candidates.

PILL ESOPHAGITIS

Epidemiology

The incidence of *pill esophagitis* is estimated to be 0.004% per year and ranges from 0.04% for alendronate to 20% or more for nonsteroidal antiinflammatory drugs (NSAIDs). [241], [242] and [243] More than 700 cases involving more than 70 different substances (Table 63-2) have been documented, although most cases have been caused by tetracyclines, potassium chloride, and quinidine. [244], [245]

Nonsteroidal antiinflammatory drugs (including aspirin)
Tetracyclines (including doxycycline)
Quinidine
Emepronium
Papain
Bisphosphonates (alendronate, pamidronate, etidronate)
Potassium chloride and citrate
Isotretinoin
Theophyllines
Oxybutynin
Captopril
Ascorbic acid
Iron preparations
Penicillins
Sodium valproate
Cromolyn sodium
Clindamycin
Chloral hydrate
Rifampin
Zidovudine, Zalcitabine
Alprenolol

TABLE 63-2 Medications Implicated in Pill Esophagitis

The risk of pill esophagitis appears to be related to patient, esophageal, and drug factors. Activities that increase the risk include lying down immediately after

ingesting the agent and the lack of adequate oral intake after medication ingestion to ensure prompt delivery of the drug into the stomach. Esophageal factors include dysmotility of the esophagus (e.g., achalasia and presbyesophagus) or mechanical narrowing of the esophagus (i.e., an esophageal stricture or extrinsic compression). Drug factors include intrinsic caustic characteristics of the medication and systemic effects, such as with NSAIDs.

Elderly patients are at increased risk of pill esophagitis resulting from more frequent use of high-risk medications (NSAIDs, potassium, quinidine, iron), an increased prevalence of esophageal dysmotility, and decreased salivation. However, esophageal medication-induced injury may occur at any age. Esophageal transit of standard-size medication tablets can be as long as 5 minutes in 50% or more of healthy persons (Fig. 63-8; see Color Fig. 63-8.). [246], [247] and [248] This was especially true with capsules or sticky tablets because of adherence to the esophageal wall, whereas nonadherent tablets ingested with adequate fluid while the patient is in an upright position usually pass through the esophagus without delay. [245] Transit is also delayed if pills are taken when the patient is supine or without sufficient (=15 mL) of fluid. Women have been injured twice as frequently as men in reported cases, probably because women are more likely to be treated with high-risk medications such as alendronate, antibiotics, NSAIDs and emepronium bromide (used for bladder irritability). [249]

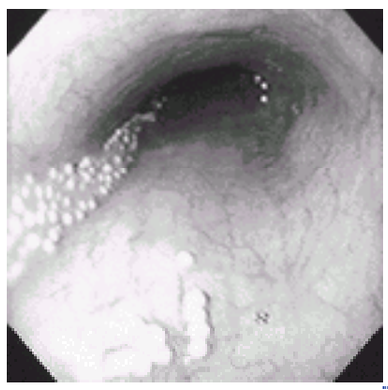

FIGURE 63-8. (See Color Fig. 63-8.) Dissolution of a medication in the midesophagus.

Pathogenesis

The pathogenesis of pill esophagitis generally involves prolonged, direct contact of the medication with esophageal mucosa. Some substances (quinidine, potassium) act directly as caustic substances, whereas others (tetracycline, ferrous sulfate) probably injure the esophageal epithelium as a result of their acidic pH, alkaline pH, or hyperosmolarity on dissolution. [250] Other agents (NSAIDs) appear to be absorbed by the esophageal mucosa and injure the mucosa from within. [243]

Sites within the esophagus at greatest risk are the middle and distal thirds. The middle third is at risk because of extrinsic compression by the left atrium and aortic arch that slow transit and increase contact time with the medication. However, the greatest incidence of pill esophagitis occurs in the distal, acid-exposed region of the esophagus related to concomitant acid reflux, as demonstrated by injury from NSAIDs and other drugs. [251] The most commonly encountered agents are detailed in Table 63-3.

AGENT (REFERENCE)	MECHANISM	ENDOSCOPIC FINDINGS
Bisphosphonates (254,257)	Direct chemical injury	Ulceration, exudate
Tetracyclines (250,258)	Acidic pH, inhibition of protein synthesis	Mild ulceration
Quinidine (250,259)	Direct chemical injury	Severe inflammation with stricture formation
Potassium (249,250)	Hyperosmolar injury	Severe inflammation
Nonsteroidal anti-inflammatory drugs (241,243, 251, 260)	Intracellular damage, prostaglandin inhibition	Distal erosive esophagitis

TABLE 63-3 Common Medications Associated with Pill Esophagitis

Symptoms

Patients with medication-induced esophageal injury usually complain of retrosternal chest pain, odynophagia, and dysphagia. [250] Pain and odynophagia are found in nearly all cases, and they may be severe enough to inhibit adequate oral intake of fluids leading to dehydration. Dysphagia is less common, usually is related to ulceration, and is rarely indicative of a stricture in this clinical situation. Patients may rarely present with hematemesis or other signs of upper GI hemorrhage. Even more unusual is a presentation of perforation with signs of sepsis, mediastinitis, or shock. [244]

Diagnosis

The diagnosis of pill esophagitis is based on the typical history of medication ingestion, risk factors, and symptoms. When confirmation the diagnosis is required, because of atypical or persistent symptoms, endoscopy is the most sensitive test. Endoscopy usually demonstrates erosions or ulcers, usually in the middle or distal third of the esophagus. Focal epithelial damage sparing the distal esophagus is nearly pathognomonic of pill esophagitis and effectively excludes gastroesophageal reflux as a cause of the esophagitis. Strictures or evidence of hemorrhage are rarely observed. If a stricture is found, it more likely is the cause of the esophagitis (pills lodging at that site) than a result of the esophagitis. Air contrast barium radiography demonstrating discrete ulcers in the midesophagus is suggestive of drug-induced esophagitis, although it is nonspecific. [252], [253], [260]

Prevention and Treatment

Adequate patient education is key to the prevention of medication-induced esophageal injury. Patients should be instructed to ingest medications in an upright position, to consume at least 100 mL of fluid at the same time, and to remain upright for 15 to 30 minutes after ingestion. Pills frequently implicated in esophageal injury should be avoided in bedridden patients and in those with esophageal compression, stricture, or dysmotility. [249]

Treatment of pill esophagitis is nonspecific and empiric. Most patients resolve their symptoms and heal mucosal lesions rapidly within days to weeks of withdrawal of the offending agent. [245] However, some patients have such severe symptoms that intravenous fluids or dietary modifications are needed to avoid dehydration and malnutrition. Although there are no data documenting the efficacy of antisecretory therapy (H_2-receptor antagonists, or proton pump inhibitors), the use of these agents can be supported to protect esophageal mucosa further from injury related to physiological or pathological reflux of gastric contents. Whether other mucosal “healing” agents, such as sucralfate suspension, are of benefit in treating pill esophagitis is unknown.

CORROSIVE ESOPHAGITIS

Epidemiology

There are more than 26,000 cases of *ingestion of corrosive substances* per year in the United States, and 17,000 or more of these cases occur in children. [261] Fifty percent of childhood ingestion cases occur in children less than 4 years of age. In children, corrosive esophageal injury usually represents accidental ingestion,

whereas most ingestions in adolescents and adults represent suicide attempts. Industrial accidents involving more concentrated agents continue to account for a small percentage of cases of corrosive esophagitis. Overall, ingestion of corrosive substances remains a leading cause of pediatric death in both developed and developing nations. Nearly one half of caustic ingestions injure the esophagus, frequently resulting in serious long-term sequelae. [261, 262]

Pathogenesis

In general, alkaline substances (lye, detergents) produce a greater depth of injury than acidic substances. [261, 263] A review of 115 Danish children hospitalized over an 18-year period revealed complications only in those ingesting strong alkali agents such as lye, whereas detergent ingestion (a weak alkali) caused a complication in only a single case. [264] Conversely, in Finland, where lye has not been available to the public for 30 years, 56 of 98 children treated over a 14-year period for caustic ingestion had ingested dishwashing detergent. [262] The severest location of caustic injury generally occurs in the narrowest portion of the esophagus, usually the midesophagus in the region of the aortic arch.

Alkaline substances tend to injure the esophageal mucosa more severely than acids because of the rapidity of the injury (seconds) and the nature of the injury (liquefaction necrosis). Previous studies have shown that the requisite pH for esophageal injury is 12.5 (0.4% sodium hydroxide has a pH of 13). [261] The severity of injury is dependent both on concentration and on mucosal contact time. Liquid lye is more injurious than the granular or nonalkaline corrosives because the granules often adhere to the mucous membranes of the mouth, thereby preventing further movement of lye into the esophagus. Nonalkalines or acidic corrosives produce coagulation necrosis and eschar formation, which protects the deeper esophageal epithelial layers from further injury. As with alkalis, the concentration of the acidic agent determines the degree of injury. Acidic substances are also cleared by esophageal peristalsis and are neutralized by swallowed salivary bicarbonate, further limiting their ability to injure the esophagus. [261] Pathologically, within 10 days of caustic esophageal injury, granulation tissue begins to replace the necrotic epithelium, and by 21 days fibroblasts are producing epithelial strictures.

Diagnosis

The diagnosis is usually based on history as well as physical findings. However, symptoms (vomiting, dysphagia, abdominal pain, oral mucosal injury) do not directly correlate with endoscopic findings, leading some to recommend that all patients with evidence of ingestion manifested by either symptoms of injury or physical findings involving the mouth should have endoscopy to stage the esophageal injury. [261, 263, 264] Asymptomatic patients are not at risk for further complications and do not require a staging endoscopy. [264]

Common findings on endoscopy, after caustic ingestion, include edema, hemorrhage, epithelial sloughing, and ulceration (Fig. 63-9; see Color Fig. 63-9.). Unfortunately, these endoscopic findings are not reliable in predicting the depth of injury. Nevertheless, some investigators have classified endoscopic esophageal injury as being first degree (superficial injury manifested by erythema), second degree (deeper injury manifested by ulceration), and third degree (penetration of injury through the esophageal wall and manifested by necrosis). Endoscopic ultrasound may provide better determination of the depth of injury and may prove to be adjunctive or even superior to endoscopy in staging caustic esophageal injury. [265] Plain abdominal or chest radiographs may reveal evidence of pleural fluid, mediastinitis, or frank perforation indicating severe esophageal injury. Barium swallow may demonstrate esophagitis, gastric abnormalities, and poor distention. [267]

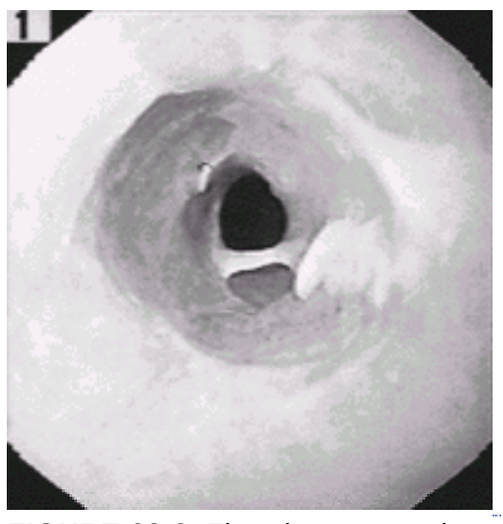

FIGURE 63-9. First-degree esophageal injury after alkali caustic ingestion with sloughing of the superficial mucosa without ulceration.

Long-term sequelae of caustic ingestion include formation of an esophageal stricture, which is usually long and rigid and is most commonly found at the region of the aortic arch. Distal esophageal peristalsis may be adversely affected, [266] and may result in decreased esophageal clearance of refluxate leading to esophagitis and a peptic stricture that would otherwise not occur. Stricture after caustic ingestion occurs only in patients with more severe injury (second and third degree) and is more likely in those with circumferential injury.

Symptoms

The esophageal symptoms of caustic ingestion include abdominal pain, retrosternal chest pain, dysphagia, and odynophagia. These symptoms usually last days to weeks and can be accompanied by other nonesophageal symptoms such as refusal to drink and increased salivation in children who are unable to describe their symptoms further. Late symptoms related to caustic ingestion include dysphagia and food impactions related to esophageal stricture.

Association with Cancer

Squamous cell esophageal cancer is estimated to be a thousandfold more common in patients having suffered caustic injury of the esophagus than in the general population. [261] The mean delay between the time of the caustic ingestion and the onset of the carcinomas is approximately 50 years. Because of the marked increased incidence of cancer in these patients, yearly endoscopic surveillance beginning 20 years after caustic esophageal injury has been recommended, although there is no outcome study showing the value of endoscopic and biopsy surveillance in this situation, and whether biopsy or cytology is useful in detecting early esophageal cancer is unproven. However, routine endoscopic biopsy of normal-appearing mucosa or biopsies guided by tissue staining with Lugol solution may increase the detection of dysplasia or early cancer and can be justified in this clinical situation.

Treatment

There is no value in attempting removal of the caustic substance by inducing vomiting, lavage, or other neutralizing attempts. In fact, those manipulations have the potential for inducing further mucosal damage. [261]

Steroids have been the most extensively investigated palliative modality. [268, 269] Steroid use remains controversial in the treatment of caustic esophageal injury because there have been no well-designed placebo-controlled trials showing an advantage of steroids over placebo treatment. Despite this, many authors continue to advocate the use of steroids in clinical practice. In a review of 13 publications of 361 patients, Howell and colleagues [269] calculated a 19% stricture rate in those treated with steroids versus a 41% rate in those not receiving treatment. Stricture never occurred in either group in patients with only first-degree esophageal burns, but it was significantly reduced in those with advanced injury who received steroids. A retrospective review of data suggested that dexamethasone (1mg/kg/d) is superior to prednisolone (2 mg/kg/d) in this setting. [268] Given that concomitant reflux may also contribute to stricture formation, antisecretory agents, such as proton pump inhibitors, should probably be given. [270]

Chronic strictures secondary to caustic esophagitis should be dilated, and approximately one third to one half of these patients will respond to this treatment. Placement of a gastrostomy has been advocated to facilitate so-called endless-loop bougienage in which a continuous string loop with plummets of progressively larger size is positioned to be passed through the mouth and esophagus to the gastrostomy. [271] Patients with severe strictures not responding to dilation require surgical therapy, most commonly with a colonic interposition. [270], [272], [273]

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CHAPTER 64

Jean-Pierre Raufman

STOMACH: ANATOMY AND STRUCTURAL ANOMALIES

ANATOMY OF THE STOMACH AND DUODENUM

As with most organs, the gross anatomy and microscopic anatomy of the stomach and duodenum are intrinsic to their function. Because in this case function relates primarily to digestion and nutrition, the structure and organization of these organs facilitate sequential physiological events that promote the fragmentation, enzymatic digestion, and absorption of nutrients. Moreover, appropriately timed autocrine, paracrine, and hormonal secretions of gastric and duodenal endocrine cells recruit other cells and organs to the digestive process. Our understanding of these complex integrated physiological events, which include secretory, peristaltic, and absorptive actions, remains incomplete. In this chapter, the anatomy and structural anomalies of the stomach and duodenum are considered together because these organs are structurally and functionally related.

Gross Anatomy

Anatomic Relationships and Divisions The stomach, a large, distensible bag with the largest diameter of any part of the gastrointestinal tract, is located in the mid-upper abdomen, just beneath the diaphragm (Fig. 64-1). The size and shape of the stomach vary greatly from person to person, depending on age, body habitus, posture, and interval since eating. The ability of the stomach to enlarge and to accommodate to meals is facilitated by its free mesentery and its distensibility, as well as location because its dilation is not constrained by other abdominal organs. Although fixed proximally to the esophagus and distally to the duodenum, and by the gastrocolic and gastrophrenic ligaments, the stomach has great latitude in distention and motion. When the stomach is nearly empty, gastric volume approximates a few hundred milliliters; when distended with food, the stomach can accommodate 2 L. The full stomach may reach from the diaphragm to the pelvic brim.

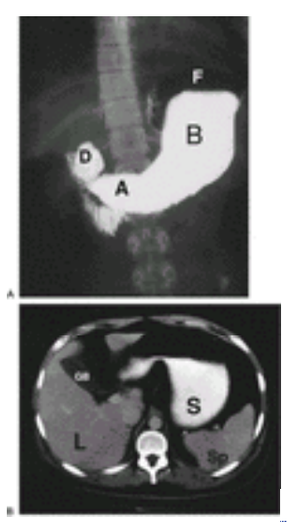

FIGURE 64-1. (**A**) Barium contrast radiograph of the stomach and duodenum (*F,* fundus; *B,* body; *A,* antrum; *D,* duodenal bulb). The pyloric channel can be seen connecting the antrum to the duodenal bulb. The second and third parts of the duodenum can be seen as well as the plicae circulares. (**B**) A computed tomogram of the abdomen shows the stomach in relation to adjacent organs. *S,* stomach; *GB,* gallbladder; *L,* liver; *Sp,* spleen. (Courtesy of Drs. Timothy Carter and Hemendra R. Shah, University of Arkansas for Medical Sciences, Little Rock, AR.)

In consequence of its size and central location in the abdomen, the stomach abuts many organs. These include the diaphragm superiorly, the liver and biliary tree to the right, the spleen to the left, and the pancreas and transverse colon posteriorly (see Fig. 64-1). The anterior surface of the stomach approximates the abdominal wall. The stomach is separated from these organs by the peritoneal lining. By convention, the somewhat J-shaped stomach is divided into the following: the cardia, a 1- to 2-cm segment adjacent to the esophagogastric junction; the fundus, the superior portion of the stomach lying above and slightly posterior to the rest of the stomach; the body or corpus, the voluminous portion of the stomach below the fundus; the antrum, the distal region of the stomach; and, finally, the pylorus or pyloric channel, a narrow (1- to 2-cm) channel that connects the stomach to the duodenum (Fig. 64-2). The shorter, right side of the stomach is the lesser curvature; the opposite, longer left side is the greater curvature. An intrusion, about two thirds of the way along the distal lesser curvature, near the junction of the body and antrum, is designated the angular notch or incisura angularis.

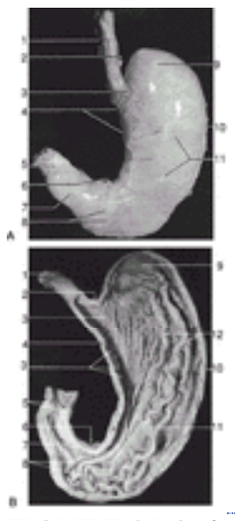

FIGURE 64-2. (**A**) Ventral aspect of the exterior of the stomach. (**B**) Ventral aspect of the posterior mucosa of the stomach. *1,* esophagus; *2,* cardiac notch; *3,* cardiac part of the stomach; *4,* lesser curvature; *5,* pyloric sphincter; *6,* angular notch; *7–8,* antrum; *9,* fundus; *10,* greater curvature; *11,* body (corpus); *12,* gastric folds (rugae). (From Rohen JW, Yokochi C. Color atlas of anatomy. New York: Igaku-Shoin, 1993.)

The tubular, C-shaped duodenum, nestling the head of the pancreas, starts at the pylorus and extends to the ligament of Treitz (Fig. 64-3). The word *duodenum* derives from its length, approximately the same as the breadth of 12 fingers (about 25 to 30 cm). The duodenum has no mesentery, and, in contrast to the stomach, it is largely retroperitoneal and fixed in position.

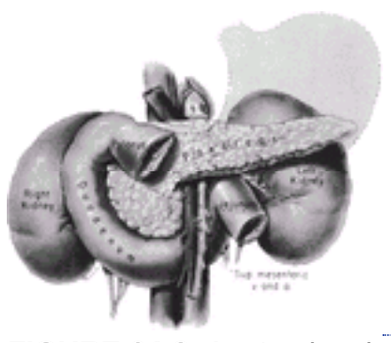

FIGURE 64-3. Anatomic relationships of the four parts of the duodenum to adjacent organs. The stomach is shown in outline. (From Rosse C, Gaddum-Rosse P. Hollinshead's textbook of anatomy. Philadelphia: Lippincott–Raven, 1997.)

The first, or superior, part of the duodenum is about 5 cm long, starts at the pylorus, and passes posteriorly, upward, and to the right. The initial 2 to 3 cm of the first part is the duodenal bulb. In contrast to the rest of the duodenum, which is lined by circular folds (plicae circulares), the lining of the bulb is relatively flat. The second, or descending, part of the duodenum takes a sharp curve and descends along the head of the pancreas for 7 to 10 cm. The common bile and pancreatic ducts, together or separately, enter the medial aspect of the second part of the duodenum at the posteromedial ampulla of Vater (Fig. 64-4). The accessory pancreatic duct (Santorini) enters the duodenum approximately 2 cm proximal to the ampulla (see Fig. 64-4). The third, or horizontal, part of the duodenum passes from right to left across the spine, inclining upward about 5 to 8 cm. The final, fourth, or ascending part of the duodenum starts left of the spine, ascends leftward, and terminates at the ligament of Treitz, where the intestine angles anteriorly and downward as the jejunum.

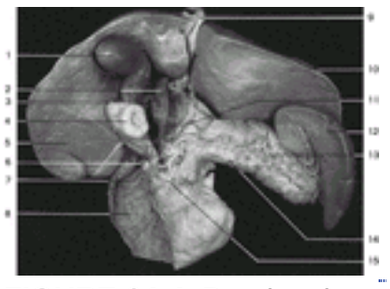

FIGURE 64-4. Duodenal papillae. The liver is slightly reflected upward. The duodenum (*8*) has been opened, and the pancreatic duct (*13*) has been dissected. A probe identifies the minor duodenal papilla (*6*), which is about 1.5 cm above the major papilla (probe, *7*). *1,* gallbladder; *2,* common hepatic duct; *3,* cystic duct; *4,* pylorus (cut); *5,* right lobe of liver; *9,* ligamentum teres; *10,* left lobe of liver; *11,* proper hepatic artery; *12,* spleen; *14,* pancreas; *15,* duct of Wirsung. (From Yokochi C, Rohgen JW, Weinreb EL. Photographic anatomy of the human body. New York: Igaku-Shoin, 1989.)

Circulation Gastric and duodenal blood supply derive from the celiac artery and the superior mesenteric artery (SMA). Both vessels arise from the abdominal aorta. The short celiac artery branches to form the splenic, left gastric, and common hepatic arteries. A dense anastomotic network that encircles the stomach is formed by vessels that branch from these major arteries. These vessels include the following: the left gastric artery, which feeds the anterior and superior portions of the stomach; the right gastric, gastroduodenal, and right gastroepiploic arteries, which arise from the hepatic artery and feed the lower right portion of the stomach and the lower greater curvature; and the short gastric and left gastroepiploic arteries, which arise from the splenic artery to feed the fundus and the upper greater curvature. The distal stomach, pylorus, and duodenum are supplied by the inferior pancreaticoduodenal branch of the SMA (see Fig. 64-3). The duodenum is also nourished by the right gastric and superior pancreaticoduodenal arteries that arise from the hepatic artery. Venous drainage from the stomach and duodenum leads directly or indirectly to the portal vein. From the stomach, the left and right gastric veins drain the lesser curvature, and the right and left gastroepiploic and short gastric veins drain the greater curvature. From the duodenum, the anterior inferior and superior pancreaticoduodenal veins drain into the superior mesenteric vein (see Fig. 64-3). The posterior superior pancreaticoduodenal vein drains directly into the portal vein.

Lymphatics The pattern of lymphatic drainage of the stomach is similar to that for its vasculature, with most lymph draining ultimately into celiac nodes. Submucosal, muscular coat, and serosal lymphatics join to drain into four major groups from the stomach and two from the duodenum. The first group of gastric lymphatics follows the left gastric artery, receives branches from the upper stomach, and ends in the superior gastric nodes surrounding the gastroesophageal junction. The second group drains the fundus and proximal stomach, follows the short gastric and left gastroepiploic arteries, and ends in the pancreaticolienal and splenic nodes, which drain into celiac nodes. The third group drains the distal greater curvature into inferior gastric nodes connected to subpyloric nodes. The final group of gastric lymphatics drains the pyloric area into superior gastric, hepatic, and subpyloric nodes. In the duodenum, anterior and posterior lymphatics drain into a series of small pancreaticoduodenal nodes near the boundary between the pancreas and duodenum. Efferents from these lymph nodes run superiorly and inferiorly to hepatic and pancreatic nodes, respectively, and to preaortic (superior mesenteric) nodes near the origin of the SMA.

Innervation The stomach and duodenum are innervated by sympathetic and parasympathetic neurons that comprise the autonomic nervous system. Sympathetic neuron cell bodies are located in the gray matter of the anterior columns of spinal thoracic segments T6 to T12; those innervating the stomach arise primarily from T7 and T8. Neural axons exit the spinal cord by the ventral roots and unite to form the greater and lesser splanchnic nerves that synapse in the celiac ganglia. Postganglionic fibers follow the hepatic, splenic, and left gastric arteries to the stomach, where they form perivascular intramural autonomic plexuses. Gastric afferent fibers follow the same course as efferent fibers; they pass through the celiac ganglia without synapsing, however, thereby reaching cell bodies in dorsal root ganglia of the spinal cord. Afferent fibers transmit visceral pain sensation from the stomach and duodenum. The vagus nerve, originating in the dorsal motor nucleus of the medulla, provides parasympathetic innervation to the stomach and duodenum. Vagal fibers course along the esophagus and enter the abdomen as the posterior and anterior vagal trunks. These neural trunks contain preganglionic efferent and visceral afferent fibers. Efferent fibers synapse with gastric cholinergic and peptidergic neurons in the wall of the stomach and duodenum that innervate various cells directly. Although afferent vagal fibers predominate, little is known about their function. After entering the abdomen at the esophageal hiatus of the diaphragm (Fig. 64-5), the anterior vagal trunk divides almost immediately into anterior gastric and hepatic branches. The anterior branch innervates the cardia and provides a branch running to the right of the lesser curvature as the anterior nerve of Latarjet. The hepatic branch innervates the liver, gallbladder, pylorus, and proximal duodenum. The posterior vagal trunk divides into celiac and posterior gastric branches. The former innervates the pancreas and other abdominal viscera. The posterior gastric branch innervates both surfaces of the stomach and forms the posterior nerve of Latarjet. The anterior and posterior nerves of Latarjet proceed along the lesser curvature, give off branches to the fundus and body, and terminate in a crow's foot neural distribution to the antrum and pylorus.

FIGURE 64-5. Distribution of the anterior (**left**) and posterior (**right**) vagal trunks. (**Left**) The anterior vagal trunk separates into (*a*) a principal branch along the lesser curvature of the stomach, (*b*) a branch that runs through the lesser omentum to reach the pyloric end of the stomach, and (*c*) a branch running higher in the lesser omentum to join the hepatic plexus. The unlabeled branch at the top innervates the liver, gallbladder, and pancreas. (**Right**) The posterior vagal trunk (seen through the stomach) separates into (*a*) a major branch along the posterior lesser curvature and (*b*) a branch to the celiac plexus. (From Rosse C, Gaddum-Rosse P. Hollinshead's textbook of anatomy. Philadelphia: Lippincott–Raven, 1997.)

Preganglionic vagal fibers synapse with ganglia in the intrinsic plexuses of the gastric wall: the myenteric (Auerbach) plexus, and the submucosal (Meissner) plexus. Postganglionic fibers from these plexuses innervate secretory and muscle cells.

Microscopic Anatomy

The stomach and duodenum have four tissue layers: mucosa, submucosa, muscularis propria, and serosa. The submucosa re- presents a connective tissue layer beneath the mucosa that is composed of a loose framework containing vasculature, lymphatics, and nerves. The muscularis propria of the stomach consists of a series of three muscle layers between the submucosa and the serosa (Fig. 64-6). The oblique muscle layer overlies the submucosa. The more substantial circular muscle layer, which thickens at the pylorus to form the pyloric sphincter, is covered by an outer longitudinal muscle layer that is a feature of the muscularis propria of the entire gastrointestinal tract. The thin serosa, the outer layer of the stomach that serves as the visceral peritoneum, is composed of areolar tissue covered by a monolayer of squamous mesothelial cells.

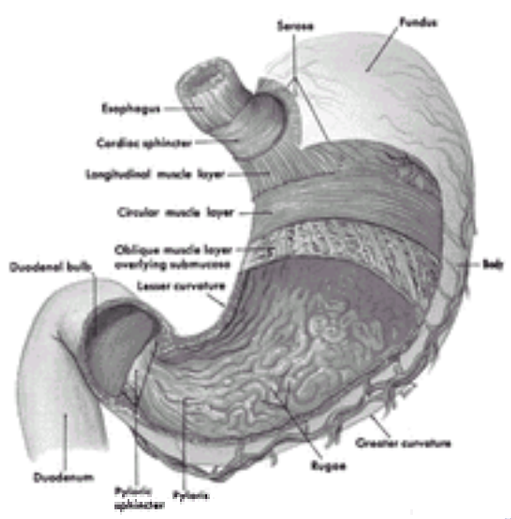

FIGURE 64-6. Muscle layers and rugal folds of the stomach. (From Thibodeau GA, Patton KT. Anthony's textbook of anatomy and physiology. St. Louis: Mosby–Year Book, 1996.)

Stomach The gastric mucosa is divided into epithelium, lamina propria, and muscularis mucosae. At the gastroesophageal junction, the mucosal boundary between the stratified squamous epithelium of the esophagus and the glandular columnar epithelium of the stomach can be seen as an irregular line (Z-line) encircling the lumen. When the stomach is empty, the mucosa and submucosa contract into thick folds called *rugae* (see Fig. 64-2, *right*). In the fundus and body, most rugae are aligned in a honeycomb pattern, whereas along the lesser curvature and in the antrum, they are arranged longitudinally (see Fig. 64-6). As the stomach distends, the rugae flatten and eventually disappear. Gastric pits (foveolae gastricae), which serve as exit channels for underlying gastric glands, stud the gastric mucosal surface. This surface epithelium is lined by a single layer of columnar epithelial cells that extends into the gastric pits and glands. Differences in the cellular makeup of glands permits histological division of the gastric mucosa into three types that differ in structure and function: cardiac (junctional), fundic, and antral (pyloric) (Fig. 64-7). The lamina propria is an area of loose connective tissue containing strands of collagen and smooth muscle, lymphatics, blood vessels, nerves, and a variety of cells, including lymphocytes, plasma cells, mast cells, fibroblasts, macrophages, and eosinophils. The muscularis mucosae, a smooth muscle layer, forms the inferior margin of the mucosa and separates it from the submucosa, which contains dense connective tissue, arterioles, venules, lymphatics, and neural plexuses.

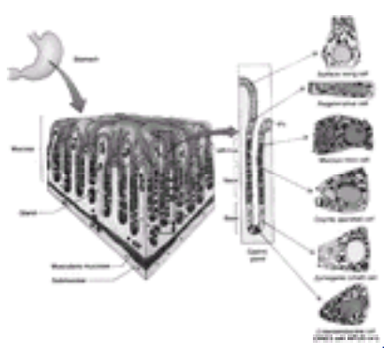

FIGURE 64-7. Drawing of gastric glands from fundus/body region of stomach showing approximate location within a representative gastric gland and typical histology of surface epithelial, mucous neck, parietal, chief, and enterochromaffin-like cells. (From Gartner LP, Hiatt JL. Color textbook of anatomy. Philadelphia: WB Saunders, 1997.)

Proceeding from the surface epithelium into the pits, the mucosal cells considered in detail in the following subsections include the following: mucous cells and mucous neck cells, which line the surface and extend into the pits, respectively; parietal cells, which secrete hydrochloric acid and intrinsic factor; endocrine cells, which secrete a variety of mediators; and, toward the base of the pits, chief cells, which secrete pepsinogens (see Fig. 64-7). [1], [2] and [3]

Mucous cells. The rectangular *mucous cells* are polarized (Fig. 64-8). Apically located mucous granules contain mucin, a neutral glycoprotein that can be stained with mucicarmine and periodic acid-Schiff. The mucous cell nucleus, Golgi complexes, and endoplasmic reticulum are located at the base of the cell. Like other columnar epithelial cells, mucous cells are attached at their apical margins by tight junction complexes and are connected by gap junctions and desmosomes at other cellular sites. Mucous neck cells are similar to the mucous cells just described, but they contain more rough endoplasmic reticulum and are found predominantly in the neck of glands and scattered deeper within glands. Because mucous neck cells contain acidic mucus (glycosaminoglycan), their granules are generally less dense than granules in surface mucous cells. Mucus is released by stimulated exocytosis; hence the degree of granule filling depends on the stage of digestion.

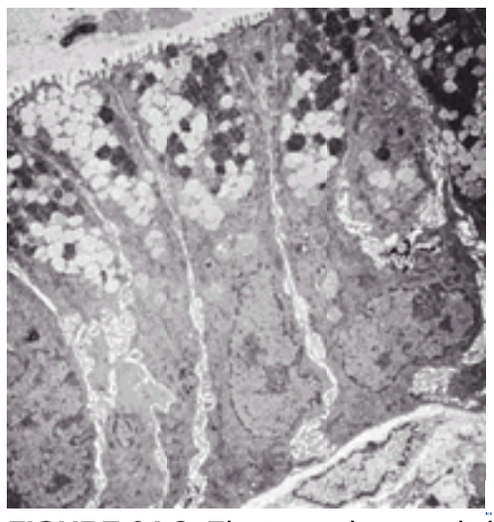

FIGURE 64-8. Electron micrograph (original magnification ×3200) of gastric mucous cells. Microvilli protrude from the apical surface, mucous granules fill the apical cytoplasm, and the nucleus lies near the base of the cell. (From Misiewicz JJ, Forbes A, Price A, et al. Atlas of clinical gastroenterology. London: Wolfe, 1994.)

Parietal (oxyntic) cells. *Parietal cells,* situated in the lower two thirds of fundic-type glands, secrete hydrochloric acid and intrinsic factor (see Fig. 64-7). In glands of the fundus and body, these cells have a concentric nucleus and appear pyramidal, with the tubular apical end abutting the glandular lumen. Numerous mitochondria and tubulovesicles occupy much of the cytoplasm (Fig. 64-9, *left*). When parietal cells are stimulated, tubulovesicles expand into microvillus-lined canaliculi that course through the cytoplasm and fuse with the apical membrane to connect with the lumen of the gastric gland (see Fig. 64-9, *right*). [4] In unstimulated cells, H^+,K^+-ATPase, which mediates the electroneutral exchange of cytoplasmic hydronium ion for potassium in the canalicular lumen, is found in the tubulovesicle membranes, whereas in stimulated cells, this enzyme is relocated to canalicular membranes. Translocation of the proton pump and expansion of the canaliculi are necessary for the acid-secreting function of parietal cells.

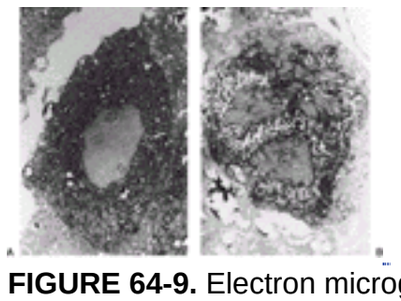

FIGURE 64-9. Electron micrographs of parietal cells. (**Left**) Resting parietal cell showing concentric nucleus and abundant mitochondria and tubulovesicles. (**Right**) Activated parietal cell with expanded canaliculus (approximate magnification ×4000). (From Prinz C, Kajimura M, Scott D, et al. Acid secretion and H,K-ATPase of stomach. In: Modlin IM, ed. Current clinical and scientific perspectives in gastroenterology. Yale J Biol Med 1992;65:170.)

Chief cells. *Chief cells,* found predominantly in the lower third of gastric glands (see Fig. 64-7), secrete proenzyme pepsinogens that are hydrolyzed in an acid environment to the acid protease pepsin. Chief cells are polar, with apically located dense pepsinogen-containing secretory granules. The basal pole contains granular endoplasmic reticulum, the nucleus, and prominent Golgi complexes (Fig. 64-10). When chief cells are stimulated, the zymogen granules migrate to the apical pole, fuse with the cell membrane, and release their contents into the lumen of the gastric gland.

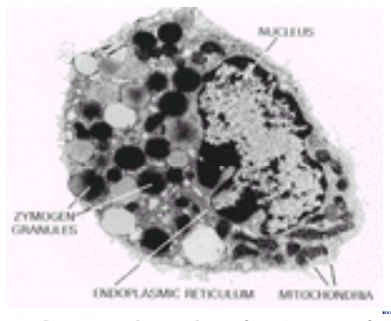

FIGURE 64-10. Electron micrograph of a dispersed chief cell from a guinea pig stomach (original magnification ×21,000). (From Raufman JP. Gastric chief cells: receptors and signal transduction mechanisms. Gastroenterology 1992;102:699.)

Endocrine cells. *Enterochromaffin-like (ECL) cells* are small, irregularly shaped cells scattered near parietal cells in fundic-type gastric glands. ECL cells, which do not connect with the lumen of the glands, are packed with histamine-containing granules. Histamine release from ECL cells has been shown to be a major regulator of parietal cell function. [5] *G cells,* located in the midportions of antral glands and in duodenal crypts, are characterized by a broad base resting against the basement membrane that runs beneath the epithelium. [6] Dense secretory granules containing gastrin cluster near the base. A narrow apex extends to the lumen of the gland. These cells secrete the hormone gastrin in response to stimuli associated with eating, such as vagal stimulation, antral distention, and the lumenal presence of aromatic amino acids. G-cell microvilli that protrude into the gland lumen sense secretory and, possibly, inhibitory lumenal stimuli. *D cells,* located in fundic and antral glands, contain the inhibitory peptide somatostatin. These cells have neuronlike extensions that terminate near other gastric mucosal cells, particularly G cells, ECL cells, parietal cells, and chief cells. Evidence indicates that inhibitory actions of D cells are mediated by paracrine actions of somatostatin released from these extensions. [7] Scattered gastric endocrine cells contain pancreatic polypeptide, glucagon, serotonin, and other secretory products whose actions are less well understood.

Duodenum In the pylorus, the mucosal pattern changes from the pits and glands (typical of gastric mucosa) to the villi and crypts (typical of intestinal mucosa). [8] From the proximal to distal duodenum, the villi become progressively taller and thinner until a ratio of villus to crypt of 4:1 or 5:1, characteristic of jejunal mucosa, is achieved. A monolayer of epithelial cells, consisting of absorptive, mucous, Paneth, and endocrine cells, covers the villi and lines the crypts. The lamina propria, composed of loose connective tissue, contains lymphatics (lacteals), blood vessels, nerves, and smooth muscle fibers, as well as varying numbers of plasma and mast cells, lymphocytes, fibroblasts, macrophages, and eosinophils. The muscularis mucosae lies below and perpendicular to the crypts. The submucosa of the duodenum provides a connective tissue framework for blood vessels, lymphatics, and nerves and contains Brunner glands that secrete alkaline mucus. Brunner glands extend through the muscularis mucosae, occupy part of the mucosa, and empty into the crypts. The muscularis propria consists of a layer of inner circular fibers surrounded by a layer of fibers arranged longitudinally. The myenteric plexus is located between these muscle layers.

Cellular Interactions Gastric and duodenal microscopic anatomy is central to the regulation of digestion; that is, the juxtaposition of secretory cells and neurons is crucial to their physiological interactions. For example, in the fundus and body, ECL cells, D cells, and cholinergic neurons must be sufficiently close to parietal cells to allow paracrine secretory products, such as histamine, somatostatin, and acetylcholine, respectively, to reach receptors on the acid-secreting cells. [5] Likewise, in the antrum, the interplay among neurons containing gastrin-releasing peptide and vasoactive intestinal polypeptide, histamine-containing mast cells, somatostatin-containing D cells, and gastrin-containing G cells requires physical proximity. [7]

Cellular Renewal Gastric and duodenal mucosal epithelial cells undergo continuous, rapid renewal, a process involving proliferation, migration, differentiation, senescence, and loss of epithelial cells. [9] To replace surface mucous cells, nascent cells migrate upward from the base of gastric pits or duodenal crypts to the mucosal surface or villous tip. In humans, migration of basal cells to the surface takes 2 to 6 days. [10] In contrast, parietal and chief cells are replaced by cells, possibly mucous neck cells, in the upper proliferative zone of the gastric glands, a process that takes weeks to months. [11] Senescent gastric or duodenal mucosal cells are sloughed into the lumen. Aspirin, indomethacin, and ethanol stimulate epithelial proliferation, perhaps by causing mild injury. [12], [13], [14], [15], [16] and [17] Epithelial proliferation is also increased by chronic inflammation, as in chronic superficial gastritis, gastric atrophy associated with pernicious anemia, and other hypergastrinemic states, such as Zollinger-Ellison syndrome. [18] In these conditions, the zone of proliferating cells is expanded. In contrast, corticosteroids and physiological stress, such as water immersion and restraint in rats, suppress epithelial proliferation. [15] Prostaglandins appear to delay senescence and loss of epithelial cells. [19] The result is mucosal thickening, which contributes to the cytoprotective effects of prostaglandins.

EMBRYOLOGY OF THE STOMACH AND DUODENUM

The primitive stomach and proximal duodenum derive from the foregut caudal to the developing lung, whereas the duodenum beyond the origin of the liver bud (the future ampullary papilla) derives from the cephalic end of the midgut. [20], [21] After the fourth week of intrauterine development, the primitive stomach rotates 90° clockwise around its longitudinal axis, ending with the left side facing anteriorly and the right side posteriorly. [22] This accounts for the course of the left vagus along the anterior wall and the right vagus along the posterior wall. The left wall of the stomach grows faster than the right, resulting in the size difference between the curvatures. The dorsal mesentery, which attaches the stomach to the posterior body wall, forms the omental bursa, or lesser peritoneal sac. The ventral mesentery attaches the stomach and duodenum to the liver.

As the stomach grows and rotates, the cephalic end moves leftward and downward, forming the fundus and cardia. The caudal end moves upward and to the right, forming the antrum and pylorus. Hence, the long axis of the stomach runs from above left to below right.

The duodenum also grows rapidly, forms a C-loop projecting ventrally, rotates to the right, and becomes retroperitoneal. Because of rapid epithelial proliferation during the fifth and sixth weeks, the duodenal lumen is temporarily obliterated. The duodenal lumen recanalizes over the ensuing weeks as some cells degenerate.

The epithelium and glands of the stomach and duodenum derive from the embryonic endoderm. The connective tissue, muscle, and serosa derive from the mesoderm.

CONGENITAL ABNORMALITIES OF THE STOMACH

Atresia

Gastric atresia results in a blind end in the antrum or pylorus. Atresia may involve only the mucosa and submucosa, with normal muscle and serosa, or it may involve the entire gastroduodenal wall. The cause is unknown, but complete atresia is familial, with autosomal recessive transmission. [23] In many cases, polyhydramnios complicates pregnancy. Complete pyloric atresia may be associated with junctional epidermolysis bullosa. Atresia probably results from failure of the antrum and pylorus to recanalize after transient occlusion by epithelium during embryogenesis.

In the newborn, gastric atresia is suggested by persistent, nonbilious vomiting, upper abdominal distention, dehydration, and hypochloremic, hypokalemic metabolic alkalosis. Gastric rupture may occur. Diagnostic abdominal radiographs demonstrate gaseous gastric distention without intestinal gas.

Initial therapy includes nasogastric suction and fluid resuscitation. Surgical treatment of a short atretic segment, involving only the mucosa, requires resection of the membrane and pyloroplasty. Extensive atretic segments require resection with gastroduodenostomy. Gastrojejunostomy should be avoided because, in the absence of vagotomy, peptic stomal ulceration may occur. Careful exploration must exclude unsuspected distal atresias.

Mucosal Membranes

Congenital mucosal membranes, which are probably related to atresias, occur in the antrum or pylorus and generally encircle, but do not occlude, the lumen. The membrane may contain either squamous or columnar epithelium. Peptic ulceration can result in acquired membranes. Vomiting may occur in infancy, but symptoms usually develop in late childhood or adulthood, depending on the diameter of the aperture.

Plain abdominal radiographs usually appear normal, but barium studies reveal a bandlike defect in the prepyloric antrum. Gastric emptying may be slowed, and the antrum between the mucosal membrane and the pylorus may simulate a second duodenal bulb. Previously, treatment was surgical, with excision of the membrane with or without pyloroplasty. More recently, endoscopic lysis of membranes has been reported. [24]

Gastric Duplication

Gastric duplications, containing mucosa, submucosa, and muscle, usually involve the greater curvature and share a common wall, but they are separate from the main part of the stomach. They sometimes connect with the gastrointestinal tract, usually the stomach or pancreas. Gastric duplications vary tremendously in size, occur more commonly in girls, and may coexist with other duplications of the digestive tract.

Patients usually present in the first year of life. Symptoms depend on size, location, and the presence or absence of communication with the gastrointestinal tract. Small children may present with vomiting, failure to thrive, and weight loss. Projectile vomiting may mimic that seen in pyloric stenosis. In older children, epigastric pain, abdominal fullness or mass, gastrointestinal bleeding, or symptoms of gastric obstruction may occur. Rarely, peritonitis results from cyst perforation, hemoptysis results from fistulation to the lung, or cancer develops in the duplication. [25] Gastrointestinal radiographs may show a mass that protrudes into the lumen, extrinsic compression of the stomach, or filling of the cyst with barium. Ultrasonography, computed tomography, or magnetic resonance imaging may demonstrate the abnormality. [25] Treatment usually consists of complete surgical excision, but this may not be feasible if a large part of the gastric wall is involved. Partial excision, partial gastrectomy, and cyst gastrostomy are other options.

Microgastria

Microgastria represents a rare failure of the stomach to enlarge after its development from the foregut. The structure becomes tubular, with a small capacity. Esophageal dilation may provide some storage area, but patients with microgastria generally present soon after birth with vomiting, diarrhea, aspiration pneumonia, malnutrition, and anemia. Microgastria is associated with developmental cardiac abnormalities, upper limb and spinal deformities, micrognathia, and asplenia. [26], [27] Many patients die within weeks to months. Supportive treatment requires frequent, small, high-caloric feedings and parenteral alimentation. If the child survives, surgical formation of a jejunal reservoir should be attempted, but anastomotic ulcers may occur. [28] Other anomalies should be sought and corrected at surgery.

Gastric Teratoma

These congenital tumors contain all three primary embryonic germ layers. They can occur anywhere, but they are rare in the stomach. *Gastric teratomas* may cause gastrointestinal bleeding, obstruction, or an upper abdominal mass. They occur almost exclusively in male patients. Although usually diagnosed in children, gastric teratomas may not be discovered until adulthood.

Plain abdominal radiographs may show calcified teeth or bone in the tumor, and barium contrast studies confirm a gastric mass. The large size of these teratomas may require total gastrectomy and formation of a jejunal pouch. [29] Gastric teratomas are not associated with other congenital anomalies, and the prognosis is good. [30]

Gastric Diverticula

Congenital *gastric diverticula* are rare, comprise all layers of the gastric wall, and arise primarily from the posterior gastric wall near the gastroesophageal junction. Gastric diverticula also may be found in the antrum and pylorus. Their cause is unknown, but they may be associated with ectopic pancreatic tissue, hiatus hernia, and diverticula in another part of the gastrointestinal tract. Ectopic pancreatic tissue, or pancreatic rests, may occur in the mucosa or submucosa of the duodenum or stomach and may mimic tumors, or they may ulcerate and bleed. Acquired diverticula may result from the scarring and dilation that accompany peptic ulceration, intestinal obstruction, cancer, or gastric surgery.

Gastric diverticula are commonly asymptomatic and are discovered as incidental findings on endoscopy or barium contrast studies. [31] These must be distinguished from acquired false diverticula, peptic ulcers, ulcerated neoplasms, and prominent folds. [32] Occasionally, epigastric or chest pain, heartburn, and vomiting may be reported. Severe symptoms or, rarely, bleeding and perforation require surgical excision and closure of the defect. Otherwise, no treatment is necessary.

Ectopic Gastric Mucosa

Rests of *ectopic gastric mucosa* can be found in any part of the gastrointestinal tract and may cause ulceration, obstruction, or bleeding. In the upper esophagus, this condition has been reported in as many as 5% of people. [33] In the small intestine, colon, and rectum, ectopic gastric mucosa may form bleeding masses resembling polyps. [34], [35] Meckel diverticula in the distal ileum may contain gastric mucosa with acid-secreting parietal cells that cause ulceration and bleeding in children and young adults. [36]

Hypertrophic Pyloric Stenosis

Neonates In *neonatal hypertrophic pyloric stenosis,* a congenital condition, muscular hypertrophy and mucosal edema of the pylorus result in gastric outlet obstruction. This disorder, the most common indication for surgery during the first 6 months of life, is more frequent in boys than in girls—1 per 150 births versus 1 per 750 births, respectively. [37] It has a genetic basis, clusters in families, and is more common in whites than in other races, but the mode of inheritance is unclear. Although the cause is unknown, lack of nitric oxide synthase in pyloric tissue may contribute to pylorospasm. [38], [39] Cases cluster in families and can be associated with other disorders, including maternal myasthenia gravis, fetal rubella, phenylketonuria, Hirschsprung disease, Turner syndrome, Smith-Lemli-Opitz syndrome, Cornelia deLange Amsterdam dwarf syndrome, and esophageal atresia. [37] Although the symptoms of pyloric stenosis, particularly regurgitation and nonbilious projectile vomiting, typically do not occur until 3 to 4 weeks after birth, up to 20% of infants may be symptomatic earlier. [37] Vomiting may occur immediately after eating or may be delayed until the stomach is full. Although the vomitus is classically nonbilious, it may contain blood. Despite vomiting, the infant remains hungry until malnutrition and weakness cause interest in feeding to wane. Decreased delivery of food and fluid to the intestines results in constipation, oliguria, and failure to thrive. The baby generally appears thin, weak, and dehydrated. Abdominal examination is likely to reveal gastric dilation, visible gastric peristalsis, and a palpable pyloric mass. Whereas gastric dilation and visible peristalsis are more evident during feeding, the olivelike pyloric mass is more likely to be palpable immediately after vomiting, when the abdominal wall is more flaccid. Typically, erect, plain abdominal radiography demonstrates a large gastric air bubble with little or no intestinal air. Studies indicate that, in most cases, the detection of an “olive” on physical examination in babies with typical symptoms is sufficient for diagnosis. [40] If further testing is needed, barium radiography appears to be more cost effective than ultrasonograpy. [41] When contrast radiography is performed, the stomach should be emptied with a nasogastric tube before barium is slowly instilled. Barium studies are likely to show a long, narrow pyloric channel, typically giving the appearance of a double channel, and a mass effect that indents the prepyloric antrum and duodenal bulb (Fig. 64-11).

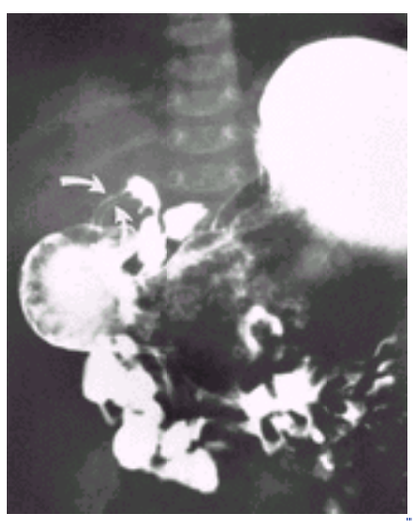

FIGURE 64-11. Barium contrast upper gastrointestinal series shows the long, narrow, double channel of the pylorus (*arrows*) in a patient with hypertrophic pyloric stenosis.

Initial therapy, before surgery is considered, requires replacement of fluid and electrolytes, as well as correction of the alkalosis resulting from repeated vomiting. Nevertheless, definitive therapy is surgical. The preferred operation is Ramstedt pyloromyotomy: longitudinal division of the anterior pyloric muscle from the serosa to the submucosa. Mild vomiting may persist, but symptoms usually disappear within several days. After surgical correction of the defect, growth and development are normal, and long-term results are excellent.

Adults Rarely, adults demonstrate signs and symptoms of congenital hypertrophic pyloric stenosis. In adults, however, most cases result from chronic peptic ulcer disease, severe gastritis, or cancer involving the pyloric area. In adults with congenital pyloric stenosis, common symptoms, usually dating from infancy, include early satiety, nausea, vomiting, epigastric pain, weight loss, and anorexia. The diagnosis is generally delayed, in part because physical findings observed in infants, such as a pyloric mass, are not present. Barium contrast radiography may reveal gastric dilation, delayed emptying, and a long, narrow pyloric channel. To exclude predisposing conditions, such as cancer or ulcer disease, upper endoscopy is helpful. Endoscopic dilation or surgical pyloromyotomy may be therapeutic, but, to exclude a small focus of cancer, pyloric resection with gastroduodenostomy may be preferable. [42]

CONGENITAL ABNORMALITIES OF THE DUODENUM

Atresias, Stenosis, and Membranes

The duodenal lumen is obliterated by epithelium during the fifth to sixth weeks of development. *Duodenal atresia, stenosis, or membranes* probably result from subsequent failure of the lumen to recanalize. This rare cause of duodenal obstruction occurs distal to the ampulla in 80% of cases, has no sexual or racial predilection, and is frequently associated with other congenital anomalies, including Down syndrome, intestinal malrotation, tracheoesophageal fistula, annular pancreas, cardiac and renal anomalies, and anorectal malformations.

In newborns with compete obstruction caused by a duodenal membrane or atresia, vomiting develops within hours to a few days after birth. The vomitus is nonbilious if obstruction is proximal to the ampulla and generally indistinguishable from that associated with pyloric stenosis. With partial obstruction, symptoms may be delayed and less severe. Rarely, duodenal membranes that persist or develop in adulthood cause upper abdominal distention, vomiting, and weight loss.

When obstruction is distal to the duodenal bulb, plain abdominal radiographs or ultrasonograms show a double bubble, caused by a distended stomach and duodenal bulb (Fig. 64-12). [43] The absence of distal intestinal gas suggests complete obstruction. Upper gastrointestinal contrast radiography may not be necessary but can confirm the site of obstruction.

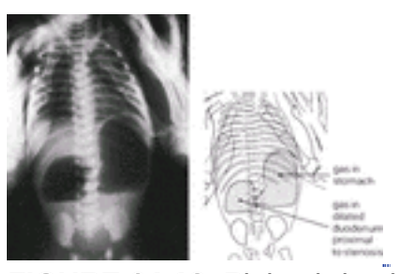

FIGURE 64-12. Plain abdominal radiograph in an infant with duodenal atresia shows the double bubble sign. The stomach and the proximal duodenum are distended with air. (From Misiewicz JJ, Forbes A, Price A, et al. Atlas of clinical gastroenterology. London: Wolfe, 1994.)

Immediate treatment requires nasogastric suction, with fluid and electrolyte resuscitation. Definitive treatment is surgical, with gastrojejunostomy or

duodenojejunostomy, depending on the level of obstruction. [44] At surgery, other correctable anomalies should be sought. The prognosis usually depends on the outcome of associated anomalies. In adults, endoscopic lysis of duodenal membranes may be successful. [45], [46]

Annular Pancreas

Annular pancreas is a rare condition that affects the sexes equally and may result in complete or partial obstruction of the second part of the duodenum. [47] The cause is unknown but probably involves incomplete migration of the ventral pancreatic bud during embryogenesis. This results in a pancreatic remnant within the duodenal wall or pancreatic encirclement of the duodenum. Annular pancreas is associated with intestinal malrotation, Down syndrome, facial cleft, imperforate anus, and cardiac and tracheoesophageal malformations. [48]

Neonates present with intractable vomiting. Children or young adults have abdominal pain and distention with less dramatic vomiting. The clinical presentation and plain abdominal radiographs are similar to those for duodenal obstruction caused by duodenal atresia, including a double bubble sign. Upper gastrointestinal contrast radiography or endoscopy confirms the level of duodenal obstruction but cannot distinguish among various causes of obstruction (Fig. 64-13). Computed tomography or ultrasonography may establish the diagnosis. In adults, endoscopic retrograde cholangiopancreatography can establish the diagnosis if the duct from the annular pancreas joins the main pancreatic duct. Impaired pancreatic ductal flow may result in acute or chronic pancreatitis.

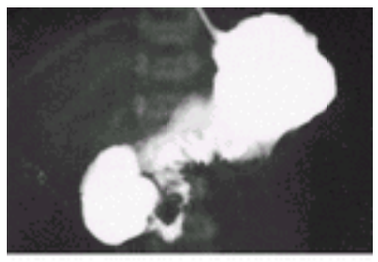

FIGURE 64-13. Barium contrast upper gastrointestinal series shows partial occlusion of the second part of the duodenum, consistent with annular pancreas, duodenal atresia, or other causes of duodenal obstruction. (From Misiewicz JJ, Forbes A, Price A, et al. Atlas of clinical gastroenterology. London: Wolfe, 1994.)

Treatment includes nasogastric decompression and correction of fluid and electrolyte disturbances. Resection or division of the annular pancreatic segment may cause pancreatitis or pancreatic fistula and should not be attempted. Definitive therapy requires duodenostomy or duodenojejunostomy. Surgical results are good, and long-term prognosis depends on the outcome of associated anomalies.

Duplication

Duplications are cystic or tubular malformations that contain all layers of the normal duodenum and may communicate with the main lumen. Intestinal obstruction may occur secondary to compression by a mass effect and may cause upper abdominal distention, early satiety, and vomiting. Duodenal duplications may ulcerate and bleed and, rarely, perforate into the head of the pancreas and cause pancreatitis. [49] Diagnosis can be established by ultrasonography, contrast radiography, or computed tomography. Treatment is surgical, and the approach depends on the relationship of the cyst with the biliary and pancreatic ducts, as well as the vasculature. If total resection is not possible, cystenterostomy may suffice.

Malrotation

In children, *intestinal malrotation* is the most common cause of duodenal obstruction. [50] This may occur in utero if the duodenojejunal loop does not rotate normally around the SMA and the duodenum fails to complete its rotation. Consequently, the ligament of Treitz may be situated to the right, rather than left, of midline, thereby kinking the duodenum. If the cecum fails to rotate completely into the right lower abdomen or when the hepatic flexure lies medial to the duodenum, mesenteric bands may compress the second or third parts of the duodenum. Other developmental anomalies, such as duodenal atresia, annular pancreas, or Hirschsprung disease, are present in 30% to 60% of patients with intestinal malrotation.

Patients with malrotation present within the few weeks of life with bilious vomiting and abdominal distention. Intestinal peristalsis may be visible on the abdominal wall. Occasionally, lax mesentery results in intestinal or cecal volvulus. If the vasculature is twisted, intestinal ischemia can result in bleeding, perforation, peritonitis, and sepsis. A few patients with malrotation present later in life with weight loss, hypoproteinemia, chylous ascites, increased susceptibility to infection, intermittent postprandial vomiting, and abdominal distention. Others, with minor malrotations, remain asymptomatic throughout life.

Plain abdominal radiographs demonstrate a large gastric air bubble and distended duodenum proximal to the obstruction. An upper gastrointestinal series can confirm the obstruction and, if barium is able to pass this area, may identify the type and extent of malrotation. Ultrasonography may reveal obstruction or volvulus caused by malrotation in utero.

Treatment is surgical: division of obstructing bands, resection of infarcted bowel, and fixation of the cecum to prevent further episodes of volvulus. When extensive bowel resection is required, mortality approaches 20%, and survivors may develop short bowel syndrome with profound malabsorption requiring long-term parenteral alimentation.

Superior Mesenteric Artery Syndrome

Rarely, the SMA obstructs the third portion of the duodenum by compressing it against fixed retroperitoneal structures, particularly the spine. Although *SMA syndrome* has been attributed to an acute angle between this vessel and the aorta, thereby trapping the duodenum, the exact cause is unknown. This syndrome also has been associated with rapid childhood growth, major weight loss in adults, immobilization in a cast that increases lordosis and accentuates the angle of the SMA with the aorta, abdominal surgery, and inflammatory diseases of the abdomen. [51], [52]

Symptoms are nonspecific, and documenting SMA compression of the duodenum as the cause is difficult. Symptoms, including episodic epigastric pain associated with vomiting, may be acute or chronic, sometimes lasting many years.

In adults, plain abdominal radiographs are usually unremarkable, but in children a double bubble sign may be seen (see Fig. 64-12). Upper gastrointestinal contrast studies may show dilation behind an abrupt cutoff in the third portion of the duodenum (see Fig. 64-13). On abdominal angiography, lateral views may show the narrowed angle of the SMA with the aorta. Because controversy exists about the clinical importance of SMA syndrome, it is important to exclude other causes of abdominal pain before making this diagnosis and initiating treatment.

Medical therapy includes small feedings or a liquid diet. Patients may benefit from lying prone or on their left side postprandially. In some cases, weight gain or removal of a body cast improves symptoms. Laparoscopic lysis of the ligament of Treitz has been reported beneficial in a few cases. [53] Duodenojejunostomy may be

required in refractory cases.

Preduodenal Portal Vein

A *preduodenal portal vein,* sometimes associated with a preduodenal common bile duct, can obstruct the duodenum. Only about 50 cases have been reported. In early embryonic life, both retroduodenal and preduodenal veins drain the primitive gut. The preduodenal vein usually atrophies, leaving a normal retroduodenal portal vein. When the retroduodenal branch atrophies, the preduodenal vein remains patent and may compress the duodenum. A preduodenal vein is associated with other congenital malformations, such as duodenal stenosis or atresia, annular pancreas, intestinal malrotation, and biliary tract abnormalities. [54]

Duodenal Diverticula

Congenital *duodenal diverticula* occur within a few centimeters of the ampulla of Vater (Fig. 64-14). [55] The ampulla may empty into a diverticulum. These diverticula are usually asymptomatic, incidental findings on barium contrast studies. Rarely, duodenal diverticula obstruct the common bile duct by external compression on the duct or distortion of its entry into the duodenum. Most cases of biliary obstruction associated with a duodenal diverticulum result from another cause, such as an obstructing common bile duct stone or neoplasm. Acquired diverticula in the duodenal bulb may result from peptic ulcer disease. When one is reasonably certain that diverticula are causing symptoms, the best procedure is choledochoduodenostomy or jejunostomy with Roux-en-Y anastomosis, not excision of the diverticulum.

FIGURE 64-14. Barium contrast upper gastrointestinal series shows two large diverticula arising from the medial border of the descending duodenum. (From Misiewicz JJ, Forbes A, Price A, et al. Atlas of clinical gastroenterology. London: Wolfe, 1994.)

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CHAPTER 65

Henry P. Parkman and Robert S. Fisher

DISORDERS OF GASTRIC EMPTYING

Gastric motility disorders include delayed gastric emptying (gastroparesis), rapid gastric emptying (as seen in dumping syndrome), and disorders with motor and sensory abnormalities (e.g., functional dyspepsia). Each disorder can present with a spectrum of symptoms that may be mild, leading to empiric treatment, or severe and incapacitating. Refractory symptoms often lead to a comprehensive evaluation including gastric emptying scintigraphy, antroduodenal manometry, electrogastrography (EGG), and possibly tests of gastric accommodation or compliance (satiety testing). Evaluation may guide treatments that target the underlying pathophysiology. Management of these patients requires an understanding of the pathophysiology, clinical tests, and new treatment options.

DELAYED GASTRIC EMPTYING (GASTROPARESIS)

Symptoms and Clinical Presentation

Gastroparesis is a chronic motility disorder of the stomach characterized by delayed gastric emptying in the absence of mechanical obstruction. The clinical presentation of gastroparesis may vary. Symptoms include early satiety, nausea, vomiting, bloating, and upper abdominal discomfort. Late postprandial vomiting of undigested food is typical. In 146 patients with gastroparesis, nausea was present in 92%, vomiting in 84%, abdominal bloating in 75%, and early satiety in 60%. [1] These symptoms, however, are nonspecific and may mimic structural disorders such as ulcer disease, partial gastric or small bowel obstruction, gastric cancer, and gallbladder or pancreatic disorders. [2] There is also an overlap of symptoms of gastroparesis and functional dyspepsia. At times, vomiting can be difficult to distinguish from regurgitation of gastroesophageal reflux disease (GERD) or rumination syndrome.

Abdominal discomfort or pain is present to varying degrees in gastroparesis, but it is not usually the predominant symptom. In the series of 146 patients with gastroparesis reported by Soykan and colleagues, [1] 46% had abdominal pain, whereas in another series, [3] 89% of 28 patients with gastroparesis had abdominal pain. Abdominal discomfort is generally described as burning, vague, or crampy. Visceral hypersensitivity to gastric distention has been suggested as a cause of pain in gastroparesis, analogous to that described in functional dyspepsia. [4], [5] In diabetic gastroparesis, abdominal pain may represent a visceral equivalent of autonomic neuropathy. Potential explanations for diabetic neuropathy include nerve injury from ischemic microangiopathy or abnormal metabolic function. [6] Abdominal pain from gastroparesis responds poorly to prokinetic agents and gastric electrical stimulation. [3]

Symptoms do not correlate well with delayed gastric emptying in diabetic gastropathy, idiopathic gastroparesis, and functional dyspepsia. [7], [8], [9] and [10] Early satiety, abdominal fullness or distention, and vomiting may, however, predict delayed emptying. [11], [12] Treatment with the prokinetic cisapride improved emptying and reduced symptoms of epigastric pressure and bloating with borderline improvement in early satiety, nausea, and vomiting. [13] In 343 patients with functional dyspepsia, 34% of whom had gastroparesis, the severity of postprandial fullness and vomiting correlated with delayed emptying. [12]

Most gastroparetic patients are women. In the series of Soykan and colleagues, [1] 82% of gastroparetic patients were female. Female gender and the later portion of the menstrual cycle (the luteal phase) have been associated with slower gastric emptying, [14], [15] thought to result from reduced gastric muscle contractility caused by female reproductive hormones, primarily progesterone. In the study by Stanghelleni and colleagues [12] of functional dyspepsia, female gender was an independent risk factor for delayed gastric emptying.

Evaluation of Patients with Suspected Gastroparesis

Gastroparesis is diagnosed by demonstrating delayed gastric emptying in a symptomatic patient after exclusion of mechanical obstruction by endoscopy or upper gastrointestinal (GI) series (Table 65-1). Although abdominal pain may be present in patients with gastroparesis, this is usually evaluated separately using other tests such as ultrasonography.

1. Initial investigation
   A. History and physical examination
   B. Blood tests
      Complete blood count
      Complete metabolic profile including glucose, potassium, creatinine, total protein, albumin, calcium
      Amylase, if abdominal pain is significant symptom
      Pregnancy test, if appropriate
   C. Abdominal obstruction series, if vomiting or pain is acute or severe
2. Evaluate for organic disorders
   A. Upper endoscopy to evaluate for mechanical obstruction or mucosal lesions (alternative: barium upper gastrointestinal series often with small bowel follow through)
   B. Biliary ultrasound, if abdominal pain is a significant symptom
3. Evaluate for delayed gastric emptying
   A. Solid phase gastric emptying test
   B. Screen for secondary causes of gastroparesis
      Thyroid function tests
      Antinuclear antibody
      Glycosolated hemoglobin
4. Treatment trial with prokinetic agent or antiemetic agent
5. If no clinical response, consider further investigation
   A. Electrogastrography
   B. Antroduodenal manometry
   C. Small bowel evaluation with enteroclysis

TABLE 65-1 Evaluation of Patients Suspected to Have Gastroparesis

The first step is taking a careful history. Understanding the patient's symptoms often leads to a clinical suspicion of gastroparesis. Typical symptoms in a young female patient may suggest idiopathic gastroparesis, just as typical symptoms in a patient with long-standing insulin-dependent diabetes should suggest diabetic gastroparesis. After abdominal surgery, patients can develop delayed gastric emptying, especially if the vagus nerve has been damaged. On physical examination, the presence of foul breath or a succussion splash may suggest gastroparesis. A succussion splash, detected by auscultation over the epigastrium while moving the

patient from side to side or by rapidly palpating the epigastrium, indicates excessive fluid in the stomach from gastroparesis or mechanical gastric outlet obstruction. [16] Dehydration and weight loss suggest chronic or severe symptoms.

The second step in evaluating gastroparesis is to exclude mechanical obstruction or ulcers by performing an upper endoscopy or upper GI series. Mechanical gastric outlet obstruction can be caused by pyloric stenosis, an active duodenal, pyloric channel, or prepyloric ulcer, scarring from prior ulcers, or neoplasia of the stomach. Gastroparesis is suggested on endoscopy by presence of food particles in the stomach after an overnight fast. In severe gastroparesis, bezoars may occur, especially if the gastric phase III migrating motor complex (MMC) is absent. Endoscopy is more sensitive for detection of mucosal lesions. [16]

Double-contrast techniques have increased the sensitivity of radiologic studies. When an upper GI series is ordered, a small bowel follow-through (SBFT) can look for small bowel lesions. The SBFT is accurate for detection of high-grade small bowel obstruction and usually provides an adequate assessment of the terminal ileum. However, it may fail to detect low-grade obstruction and smaller mucosal lesions. An SBFT may rarely suggest superior mesenteric artery syndrome as a possible cause or consequence of symptoms. Enteroclysis (small bowel enema) can be obtained after placement of a nasoduodenal or oroduodenal tube. Compared with SBFT, enteroclysis is more accurate in detecting small intestinal mucosal lesions, mild to intermediate grades of obstruction, and small bowel cancers. [17] Computed tomographic scanning with oral and intravenous contrast may also be useful for detection and localization of intestinal obstruction.

The third step is to obtain a gastric emptying scintigram using a radiolabeled solid meal. Patients should discontinue medications that may affect gastric emptying for 48 hours before this test (Table 65-2). An abnormal gastric emptying test suggests, but does not prove, that the symptoms are caused by gastroparesis. If gastric emptying is normal, other causes for symptoms should be sought. However, a gastric motility disorder should not be totally dismissed because localized motor dysfunction, such as impaired fundic relaxation or gastric dysrhythmias, may be associated with symptoms and normal gastric emptying. [18]

Delay Gastric Emptying
Opiate analgesics
Anticholinergic agents
Tricyclic antidepressants
Calcium channel blockers
Progesterone
Octreotide
Proton pump inhibitors
Histamine H_2 receptor antagonists
Interferon-α
L-Dopa
Fiber
Sucralfate
Aluminum hydroxide antacids
β-Adrenergic receptor agonists
Glucagon
Calcitonin
Dexfenfluramine
Diphenhydramine
Alcohol
Tobacco/nicotine
Tetrahydrocannabinol
Accelerate Gastric Emptying
Prokinetic agents
Metoclopramide
Erythromycin and related antibiotics
Cisapride
Domperidone
β-Adrenergic receptor antagonists

TABLE 65-2 Medications that Affect Gastric Emptying

Evaluation of a patient should not stop once delayed gastric emptying is demonstrated; an underlying cause should be sought. Thyroid function tests should be obtained to evaluate for hypothyroidism. A glycosylated hemoglobin level evaluates long-term glycemic control; poor glucose control can worsen gastric emptying. If a patient does not have diabetes mellitus and has not had gastric surgery, a search should be undertaken to evaluate some other causes of gastroparesis including smooth muscle, neurological, or metabolic or endocrinologic disorders. Idiopathic gastroparesis is diagnosed after other causes are excluded.

Evaluation of Gastric Emptying, Motor Function, and Myoelectric Activity

Radiographic contrast techniques: upper gastrointestinal series. The upper GI series is an insensitive method for measuring gastric emptying because it is difficult to quantitate, and barium is not a “physiological” meal (Table 65-3). [19] Gastric retention may be suggested by poor emptying of barium from the stomach, gastric dilation, and the presence of retained food or a gastric bezoar. Little or no emptying of barium at 30 minutes and minimal barium remaining at 6 hours are suggestive of delayed gastric emptying. [20] The upper GI series is primarily helpful in detecting mucosal lesions and mechanical outlet obstruction.

TABLE 65-3 Tests to Assess Gastric Motor and Myoelectric Function

Radionuclide gastric emptying scintigraphy. Gastric emptying scintigraphy remains the best current test for measuring gastric emptying because it is sensitive, quantitative, and physiological. It is used to confirm the presence of gastric stasis after excluding structural or mucosal disorders. The usefulness of emptying tests, however, in directing therapy and predicting response is debated. [13], [21] Measurement of gastric emptying of solids is more sensitive for detection of gastroparesis because normal emptying of liquids is often preserved until the disorder is advanced. In patients who have undergone gastric surgery, a dual solid and liquid emptying test may be indicated because symptoms may result from slow solid emptying or rapid liquid emptying. Most centers use a technetium-99m sulfur colloid–labeled egg sandwich as a test meal. [19] A meal using the processed food Egg Beaters has been proposed for standardization among centers. [22] Whatever meal is employed, the radiolabel needs to be cooked into it so the radioisotope binds to the solid phase, thus preventing elution of the radiotracer into the liquid phase with an erroneous measurement of the faster liquid phase gastric emptying. [23] Gastric emptying should be carried out to at least 2 hours after meal ingestion. For shorter durations, the test is less reliable because of large variations of normal gastric emptying. Extending scintigraphy to 4 hours increases the detection of delayed gastric emptying. [24], [25] The simplest approach for interpreting gastric emptying study is to report the percentage of retention at defined times after meal ingestion (usually 2 and 4 hours) (Fig. 65-1). Curve fitting techniques can calculate the half-emptying time, the time at which half of the stomach contents have emptied from the stomach. Extrapolation of the emptying curve to predict the half-emptying time is unreliable if the emptying had not reached 50% during the actual imaging. [26] Other parameters of potential use are the lag phase for solids, which represents the time required for trituration of solid food into 1- to 2-mm particles that can then empty through the pylorus. A prolonged lag phase suggests antral hypomotility. The slope of the gastric emptying curve after the lag phase represents the rate at which solids leave the stomach once emptying begins. A decreased slope suggests antral hypomotility or small intestinal dysmotility. [27]

FIGURE 65-1. Gastric emptying scintigraphy using a technetium-99m–labeled egg sandwich. The percentages of gastric retention are shown for 0, 30, 60, 120, 180, and 240 minutes after meal ingestion. (**A**) Normal gastric emptying with only 30% retention at 2 hours after meal ingestion (normal <50%) and complete emptying at 4 hours (normal <10%). (**B**) Markedly delayed gastric emptying with little emptying at 4 hours.

Advances in scintigraphy may provide information on fundic and antral abnormalities. Dynamic antral scintigraphy with frequent 1-second imaging can evaluate antral wall contractility. [19] Regional gastric emptying can assess intragastric meal distribution and transit from the proximal to distal portions of the stomach. Proximal

retention may be seen in GERD, distal retention in functional dyspepsia, and global retention in gastroparesis. The gastric emptying response to intravenous prokinetic agents was suggested to predict the response to long-term oral therapy. More recent studies have not supported this. [28] Most clinicians follow the symptomatic response to therapy. Glucose and gender are factors that affect gastric emptying. Premenopausal women empty the stomach more slowly than men. [14], [15] Some investigators have advocated separate reference values for premenopausal women. [29] Hyperglycemia delays gastric emptying. Checking blood glucose concentrations of diabetic patients before a gastric emptying test has been suggested, but it is not usually done.

Breath tests for gastric emptying. Breath testing can be used to measure gastric emptying with the nonradioactive isotope, ^{13}C, to label octanoate, a medium-chain triglyceride, which can be bound into a solid meal. [30], [31] and [32] Studies have also reported labeling ^{13}C to proteinaceous algae (*Spirulina*). [23] Octanoate, after ingestion and emptying from the stomach, is absorbed by the small intestine. The octanoate is metabolized to carbon dioxide in the liver that is excreted from the lungs during respiration. The rate-limiting step for excretion of ^{13}C is gastric emptying. By measuring ^{13}C in breath samples, gastric emptying can be indirectly determined. The octanoate breath test is reproducible and correlates with gastric emptying scintigraphy, and it offers promise as a clinical test for gastric emptying. The ^{13}C breath test does not use ionizing radiation; it can be used to study patients away from gamma camera facilities in community doctor's offices or at the patient's bedside, and breath samples can be shipped to a laboratory for analysis. The octanoate breath test has been used in clinical research and pharmaceutical studies, but not often for patient evaluation. Validation of this test in patients with emphysema, cirrhosis, celiac sprue, and pancreatic insufficiency is needed, because it is not clear whether substrate metabolism in these disorders may also be a rate limiting step for $^{13}CO_2$ excretion.

Ultrasonography. Transabdominal ultrasonography can measure several parameters of gastric motility. Serial changes in antral cross-sectional area are measured as an index of gastric emptying; emptying is considered complete when the antral area returns to the fasting baseline. [33] Duplex sonography may be used to evaluate transpyloric flow of liquid gastric contents. Ultrasonography has also been employed to measure accommodation in the proximal and distal stomach. [23] Unfortunately, ultrasonography for gastric emptying is operator dependent and generally measures liquid emptying only. The test is suboptimal in obese people.

Electrogastrography. EGG is the recording of gastric myoelectric activity, using cutaneous electrodes on the anterior abdomen overlying the stomach. [34] The dominant frequency of the EGG corresponds to the gastric electrical rhythm or frequency of the gastric slow wave. The normal gastric slow wave frequency is approximately 3 cycles/min. Meal ingestion increases the amplitude of the EGG signal by increases in gastric electrical activity and contractility or gastric distention from the meal. EGG measures the frequency and regularity of gastric myoelectric activity, detects abnormal rhythms of gastric myoelectric activity, and assesses the amplitude or power increase after a meal. [26] Gastric dysrhythmias (tachygastria, bradygastria) and decreased postprandial amplitude (or power) of the EGG have been described in idiopathic and diabetic gastroparesis [35] (Fig. 65-2). Gastric myoelectric abnormalities have also been described in patients with unexplained nausea and vomiting, motion sickness, and nausea and vomiting of pregnancy. [26] Studies have suggested a good correlation between delayed gastric emptying by scintigraphy and an abnormal EGG. [35] An abnormal EGG is present in 75% of patients with gastroparesis compared with only 25% of symptomatic patients with normal gastric emptying. Gastric dysrhythmias may be better predictors of symptoms than delayed gastric emptying and may correlate better with symptomatic response to medications. [8] In diabetic patients, hyperglycemia may itself provoke dysrhythmias, primarily tachygastrias. [36]

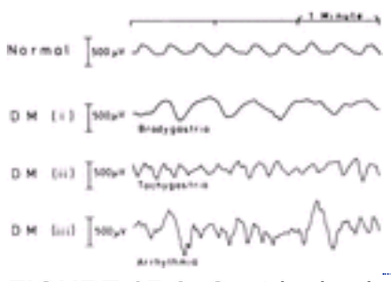

FIGURE 65-2. Gastric dysrhythmias recorded with electrogastrography from diabetic patients. The **top tracing** shows normal 3 cpm activity. The **next tracings** show examples of bradygastria, tachygastria, and arrhythmia (tachybradygastria). (From Hongo M, Okuno Y, Nishimura N, Toyota T, Okuyama S. Electrogastrography for prediction of gastric emptying state. In: Chen JZ, McCallum RW, eds. Electrogastrography: principles and applications. New York: Raven Press, 1994:257.)

EGG is used to demonstrate gastric myoelectric abnormalities in patients with unexplained nausea and vomiting or functional dyspepsia. The EGG is generally used as an adjunct to gastric emptying scintigraphy as part of a comprehensive evaluation of patients with refractory symptoms suggestive of an upper GI motility disorder. [26]

Antroduodenal manometry. Antroduodenal manometry, although somewhat invasive and lengthy (requiring at least 5 hours of recording), provides information about coordination of gastric and duodenal motor function in both fasting and postprandial periods. [26] Ambulatory studies, performed over 24 hours using solid-state transducers, allow correlation of symptoms with abnormal motility; catheter migration in the stomach prevents quantitation of antral contractility. The main indications for antroduodenal manometry are to evaluate the following:

- Unexplained nausea and vomiting
- The cause of gastric or small bowel stasis (e.g., neuropathic or myopathic disorders)
- Suspected chronic intestinal pseudoobstruction when the diagnosis is unclear. [26]

Decreased antral contractility and phase III MMCs originating in the small intestine rather than in the stomach can be seen in gastroparesis. Occasionally, pylorospasm or irregular bursts of small intestinal contractions, which increase outflow resistance, can be seen. Antroduodenal manometry can help to confirm or exclude a GI motility disorder if the gastric emptying test is normal or borderline. With an accurate stationary recording, a reduced postprandial distal antral motility index is correlated with impaired gastric emptying of solids. [27] An average of less than one antral contraction per minute postprandially has been suggested as a simple estimate of significant hypomotility. [37] A normal study with a normal transit test strongly suggests that motor dysfunction is not the cause of symptoms. [26] Antroduodenal manometry may differentiate between a neuropathic or myopathic motility disorder, and it may suggest unexpected small bowel obstruction or rumination syndrome. [26], [38] Myopathic disorders, such as scleroderma or amyloidosis, have low-amplitude contractions with normal propagation. Neuropathic disorders have normal amplitude but abnormal propagative contractions, seen readily in the phase III MMC, such as bursts and sustained uncoordinated pressure activity, and a failure of a meal to induce the fed-type pattern. Occult mechanical obstruction of the small intestine is suggested by two types of patterns: postprandial clustered contractions for more than 30 minutes' duration separated by quiescence and simultaneous prolonged (>8 seconds) or summated contractions suggesting a common cavity phenomenon from a dilated segment of intestine. [39] Antroduodenal manometry may demonstrate a characteristic pattern of rumination with an increase in intra-abdominal pressures at all levels of the upper gut (R waves), especially postprandially. [40] In pediatric studies, the absence of MMCs is an indicator of a poor response to prokinetic agents. [41] Some investigators perform the study with infusions of erythromycin or octreotide to predict the patient's response to long-term treatment of these agents. [42]

Gastroparesis and Disorders with Delayed Gastric Emptying

Gastroparesis occurs in many clinical settings (Table 65-4); the most common are idiopathic conditions, diabetes mellitus, and sequelae of gastric surgery. The relative prevalence depends on referral patterns of reporting investigators. In a series of 146 patients, gastroparesis was idiopathic in 36%, diabetic in 29%, and postsurgical in 13% of patients. [1]

Idiopathic
Endocrine or metabolic
Diabetes mellitus
Thyroid disease
Renal insufficiency
After gastric surgery
After resection
After vagotomy
Central nervous system disorders
Brainstem lesions
Parkinson disease
Peripheral neuromuscular disorders
Myotonia dystrophica
Duchenne muscular dystrophy
Connective tissue disorders
Scleroderma
Polymyositis/dermatomyositis
Infiltrative disorders
Lymphoma
Amyloidosis
Diffuse gastrointestinal motility disorder
Chronic intestinal pseudoobstruction
Medication-induced (see Table 65-2)
Electrolyte imbalance
Potassium, calcium, magnesium
Miscellaneous conditions
Infections (especially viral)
Paraneoplastic syndrome
Ischemic conditions
Gastric ulcer

Adapted from ref. 19.

TABLE 65-4 Etiology of Gastroparesis (Nonobstructive Delayed Gastric Emptying)

Diabetic Gastroparesis Gastroparesis is a well-known complication of *diabetes,* most commonly associated with long-standing, insulin-dependent diabetes mellitus with other complications of retinopathy, nephropathy, and peripheral neuropathy. Frequently, it occurs with autonomic neuropathy as identified by orthostatic hypotension. Longitudinal studies suggest that delayed gastric emptying is present in 30% to 50% of patients with long-standing type 1 or type 2 diabetes but is not necessarily associated with diabetic complications. [43] Diabetic gastroparesis is clinically important because it is associated with GI symptoms, alterations in glycemic control, and changes in oral drug absorption. [43] Symptoms from diabetic gastroparesis include primarily nausea and vomiting, but also early satiety, fullness, and abdominal discomfort. The presence of abdominal bloating or fullness in particular appears to be associated with diabetic gastroparesis. [44] Symptom severity, however, does not necessarily correlate well with the degree of gastric stasis. [7], [45] Patients with severe symptoms may have only a mild delay or even normal gastric emptying. One study, examining the effect of the prokinetic agent, domperidone, in diabetic gastroparesis suggested that symptom improvement did not correlate with improvement in gastric emptying, but rather with resolution of dysrhythmias. [8] Diabetic gastroparesis has been considered to be associated with a poor prognosis; more recent studies suggest this may be incorrect. [43] The motor dysfunction is typically confined to the stomach (gastroparesis), but sometimes a diffuse GI motility disorder may be present. Diabetic "gastropathy" has been used to include a range of gastric pathophysiological abnormalities observed in diabetic patients with dyspeptic symptoms. Gastric motor abnormalities include impaired fundic relaxation with antral dilation, gastric dysrhythmias, antroduodenal incoordination, antral hypomotility, and visceral hypersensitivity [4]; gastroparesis may be the end stage of gastric dysfunction. The term *gastropathy* has also been used because symptoms do not predict delayed gastric emptying, and the response to prokinetic treatment is not necessarily related to improvement in emptying. Diabetic gastroparesis likely results from impaired neural control of gastric motor function, possibly resulting from a vagal neuropathy. Morphologic abnormalities of the gastric myenteric plexus in diabetic patients have not been identified by conventional histology. [46] Abnormal neural innervation and neurotransmission of both inhibitory and excitatory neurotransmitters and underlying muscle dysfunction have been described in animal models of diabetes. Impairment of the inhibitory nitric oxide–containing nerves [47] and damage of the interstitial cells of Cajal, [48] the electrical pacemaker cells, have been described in animal models and in patients with diabetic gastroparesis. [49] Gastroparesis has also been described in type 2 diabetes mellitus. [7] However, it is also not uncommon in this disease for gastric emptying to be accelerated. [50] Usually, this change occurs early in the disease. This may be related to a lack of a fundic receptive relaxation response, a vagally mediated response. [51] Hyperglycemia itself may reversibly affect gastric motility. Hyperglycemia decreases antral contractility, decreases antral phase III MMC activity, increases pyloric contractions, causes gastric dysrhythmias (primarily tachygastria), and delays gastric emptying. [36], [52] Normalization of serum glucose in hyperglycemic patients has been shown to improve gastric myoelectric activity, to improve gastric emptying, and to restore antral phase III activity. [53] It is not clear how hyperglycemia adversely affects gastric motility. Hyperglycemia appears to cause a reversible impairment of vagal efferent function. [54] Glucose-responsive neurons have been identified in the central nervous system (CNS) that may modify vagal efferent activity. [55] Prostaglandins may also be involved, because indomethacin can reverse abnormal gastric electrical rhythms that occur during hyperglycemia. [56] Patients with insulin-dependent diabetes mellitus have an increased perception of gastric distention, with more nausea, fullness, and upper abdominal pain. [4] Hyperglycemia increases nausea and fullness during proximal gastric distention. [52], [57] Increased sensitivity of the proximal stomach may be responsible for dyspeptic symptoms postprandially when the stomach is distended by a meal. [4], [57] This is also reflected in studies showing that the prevalence of GI symptoms in insulin-dependent diabetes mellitus is greater in patients with poor glycemic control. [58] Changes of gastric motility may affect postprandial blood glucose concentrations. Delayed gastric emptying may contribute to poor glucose control because of unpredictable delivery of food into the duodenum. [52] Impaired gastric emptying with continued administration of exogenous insulin may produce hypoglycemia. Conversely, acceleration of emptying has been reported to cause hyperglycemia. [50] Problems with glucose control may be the first indication that a diabetic patient is developing gastroparesis. [52] Interestingly, gastroparetic symptoms may be mild or absent. Long-term oral administration of the prokinetic agents levosulpiride and erythromycin, which accelerate gastric emptying, were shown to decrease plasma glucose and glycosylated hemoglobin (HbA_{1c}) levels in diabetic patients. [59], [60]

Postsurgical Gastroparesis *Postsurgical gastroparesis* generally occurs after peptic ulcer surgery, [61] most commonly when a truncal vagotomy is performed. Vagal nerve stimulation causes both fundic relaxation and antral contractions; the effect of a total vagotomy is to accelerate gastric emptying of liquids and to delay emptying of solids. A gastric drainage procedure such as a pyloroplasty or gastroenterostomy is usually performed in an attempt to offset the gastroparetic effects of vagotomy. In most patients, the net result is that vagotomy combined with a drainage procedure produces little alteration in gastric emptying. The results of different gastric surgical procedures on gastric emptying are shown in Table 65-5.

TABLE 65-5 Gastric Emptying Alterations after Different Types of Gastric Surgery

About 5% of patients undergoing vagotomy with antral resection and gastrojejunostomy develop severe postsurgical gastroparesis; the antrum is not present to triturate solids, and the proximal stomach is unable to generate sufficient pressure to empty solids. [61] Patients with gastric outlet obstruction undergoing surgery may also develop postsurgical gastric stasis; the chronic gastric dilation may have led to irreversible gastric muscle dysfunction. The Roux-en-Y stasis syndrome is characterized by postprandial abdominal pain, bloating, nausea, and vomiting. [61] The combination of vagotomy, distal gastric resection, and Roux-en-Y gastrojejunostomy predisposes to gastric stasis. Gastric stasis is the esult of both slow emptying from the gastric remnant and delayed small bowel transit in the Roux efferent limb. [61] Ectopic jejunal pacemakers may develop in the transected and reattached jejunal segment. In addition, myoelectric and motor activity in the Roux limb is slower than normal and may be retrograde, causing reverse peristalsis toward the stomach and allowing the Roux limb to cause a functional obstruction. [62] Gastroparesis is unusual in patients with a highly selective parietal cell vagotomy. Transient myoelectric abnormalities may occur, but they usually disappear with time. [63] Development of delayed gastric emptying after highly selective vagotomy may suggest complete vagotomy. [64]

Post-Nissen fundoplication gastric motor dysfunction. Mild acceleration of gastric emptying can occur after Nissen fundoplication. [65], [66] Regional scintigraphy suggests rapid proximal gastric emptying after fundoplication. This has been attributed to an increase in intragastric pressure resulting from the fundic wrap preventing receptive relaxation. This may lead to symptoms of early satiety. [66] In patients with GERD and delayed gastric emptying, there can be normalization of emptying after fundoplication. [66] Emptying has been reported to improve after radiofrequency ablation of the LES. [67] *Gas bloat syndrome* refers to postoperative symptoms including inability to belch, pain, and bloating. It is more common in patients with preoperative delayed gastric emptying. [68] Early satiety, postprandial fullness, nausea, and vomiting may develop after fundoplication. [69] In some of these patients, postprandial antral hypomotility or a delay in gastric emptying can be seen. [70], [71] Delayed gastric emptying may have been present before surgery but not recognized because of the GERD symptoms. The development of postoperative gastroparesis after an open or laparoscopic Nissen fundoplication is uncommon but at times may result in significant morbidity. [71] Vagal nerve injury has been reported in 4% to as many as 40% of patients undergoing laparoscopic fundoplication. [72], [73]

After bariatric surgery. Surgical therapy for morbid obesity has increased since the late 1990s. Several procedures are designed to restrict stomach size. [74] The most commonly performed surgical procedure is the Roux-en-Y gastric bypass. The stomach is partitioned into a small proximal fundic pouch, and a bypassed distal stomach; a loop gastrojejunostomy through a small gastroenterostomy, drains the proximal pouch. [74] Ingestion of large meals is prevented by early satiety resulting from the small gastric pouch. The gastrojejunostomy configuration produces a relative malabsorptive state and can result in dumping if a high-carbohydrate liquid meal is ingested. Solid emptying is slower, and liquid emptying is faster after gastric bypass surgery. [75] Less commonly performed are gastric restrictive procedures

including vertical banding gastroplasty and adjustable gastric banding. [74] Vomiting can occur after vertical banding gastroplasty, reported in 21% of patients. [76] Another study found that emptying from the proximal pouch was normal and could not explain the early satiety. [77]

Idiopathic Gastroparesis *Idiopathic gastroparesis* refers to symptomatic gastroparesis in patients with no primary underlying abnormality. In most series, idiopathic gastroparesis is the most common form of gastroparesis. [1] Symptoms may fluctuate, with episodes of pronounced symptoms interspersed with relatively symptom-free intervals. Patients with idiopathic gastroparesis are typically young or middle-aged women. Even after adjusting for gender differences in gastric emptying, gastroparesis occurs more commonly in women. [12] A potential cause in some patients includes viral injury to the nerves or muscles of the stomach. Postviral gastroparesis was suspected in 23% of patients with idiopathic gastroparesis reported by Bityutskiy and associates. [78] This clinical diagnosis is suggested in previously healthy persons with an acute onset of viral illness with nausea, vomiting, diarrhea, fever, and cramps who have persistence of symptoms (nausea, vomiting, early satiety) for more than 3 months, with a delay in gastric emptying but no evidence of mechanical obstruction, metabolic disease, or systemic illnesses. Viruses suspected as potential causes are cytomegalovirus, (CMV) Epstein-Barr virus, and herpes varicella-zoster. Symptoms of idiopathic gastroparesis after a presumed viral illness tend to be less severe than in gastroparesis from other causes. Overall, these patients appear to have good prognosis, with many patients having a slow resolution of their symptoms. [78]

Other Disorders Associated With Delayed Gastric Emptying

Gastrointestinal Disorders

Gastroparesis associated with gastroesophageal reflux disease. Delayed gastric emptying can be seen in patients with GERD. Gastric stasis with distention may promote transient lower esophageal sphincter relaxation with subsequent gastroesophageal reflux. Some investigators have found delayed emptying in 40% of patients with GERD [79]; others have not shown this high prevalence. [80] Delayed emptying occurred equally in patients with and without reflux on esophageal pH monitoring, was independent of severity of reflux symptoms, and was independent of the presence of esophagitis. [80] Studies suggest that slow emptying from the proximal stomach, but not from the overall stomach, may correlate with esophageal acid exposure. [81] Ultrasound studies have shown that proximal gastric distention after a meal promotes GERD by increasing transient lower esophageal sphincter relaxations. [82]

Achalasia. Gastric emptying is difficult to study in achalasia because the ingested meal is often retained in the esophagus. Gastric emptying studies in these patients have been variable; several reported no abnormalities in gastric emptying. [83] Studies delivering liquid meals into the stomach through a nasogastric tube have shown rapid emptying of liquids. [84] After successful pneumatic dilation, gastric emptying of solid meals was delayed in 50% of patients with achalasia. [85] These emptying abnormalities may be related to abnormalities of the vagus nerve and dorsal motor nuclei of the vagus, which have been reported in some achalasia patients.

Gastric ulcer. Delayed gastric emptying has been reported in some patients with gastric ulcers. In some patients, the gastroparesis has resolved after ulcer healing. It may be difficult to determine whether the ulcer caused the gastric emptying delay or the gastric stasis caused the ulcer.

Atrophic gastritis. Atrophic gastritis is characterized by a decrease in gastric acid secretion usually leading to hypergastrinemia through acid-gastrin feedback regulation. Atrophic gastritis may result from pernicious anemia or may occur on a nonautoimmune basis. Delayed gastric emptying is present in two thirds of patients with achlorhydric atrophic gastritis and in one third of those with preserved acid secretion. Delayed gastric emptying in atrophic gastritis is associated with reduction of gastric acid output and hypergastrinemia, rather than severity of mucosal atrophy. [86]

Functional dyspepsia. See section “ Functional Dyspepsia” later in the chapter.

Hypertrophic pyloric stenosis. Infantile hypertrophic pyloric stenosis (IHPS), occurring in up to 0.3% of births, is the most common disorder during the first 6 months of life that requires abdominal surgery. A typical infant begins to vomit within the first 4 weeks of age. Hypertrophy of the inner circular muscle of the pylorus produces pylorospasm or functional gastric outlet obstruction resulting in nonbilious vomiting, dehydration, and metabolic alkalosis. An enlarged pylorus (pyloric olive) can be palpated or detected radiologically. Pylorospasm occurs from a defect in pyloric sphincter relaxation and not from a true obstruction. A lack of inhibitory nerves of the pylorus, primarily nitric oxide–containing nerves, and interstitial cells of Cajal has been shown in IHPS. [87], [88] Treatment with surgical pyloromyotomy is effective. Patients usually remain symptom free after surgical treatment; the muscle hypertrophy resolves. The nitric oxide and interstitial cells of Cajal abnormalities partially reverse, [89] although on manometry, the pyloric tone is higher and the phasic pyloric pressure waves are less frequent than in physiologically normal infants. [90] Medical treatment with atropine has been suggested by European groups. Hypertrophic pyloric stenosis with achalasia has been reported in the same patient. IHPS has been associated with the use of erythromycin for pertussis prophylaxis in neonates. [91] IHPS in adults is rare. [92] Diagnostic criteria for adults have been variable. Early reported cases were related to local inflammation and fibrosis from previous gastric or duodenal ulcer. In the uncommon idiopathic form, there is hypertrophy of the smooth muscle without any underlying disease. A certain proportion of these primary cases may represent persistence of the juvenile form, presenting later in life. Endoscopic dilation and laparoscopic pyloroplasty have been used for treatment. [93]

Celiac disease. Delayed gastric emptying is seen in 50% of patients with celiac disease, which normalizes with a gluten-free diet and recovery of small bowel mucosa. Delayed gastric emptying has been suggested to explain some of the dyspeptic symptoms in celiac disease. [94] Delayed emptying has been demonstrated by scintigraphy [95] and ultrasonography in assessing the antral area. [96] This was also reported with the ^{13}C-octanoate breath test for gastric emptying [94]; however, the small bowel malabsorption may “prolong” the gastric emptying values obtained, because excretion of ^{13}C is dependent on small bowel absorption.

Gallbladder disease. Delayed gastric emptying, in addition to delayed gallbladder emptying, may be present in cholelithiasis. [97] Studies have reported delayed solid phase gastric emptying in 20% to 42% of patients with gallstones, [98] whereas others have not found alterations in liquid phase emptying. [97] Interestingly, cholecystectomy has been shown to normalize gastric emptying. [98]

Chronic pancreatitis. Some patients with chronic pancreatitis have delayed gastric emptying. [99] Some of the abdominal pain, nausea, and vomiting seen in patients with presumed or documented chronic pancreatitis may result from gastroparesis; this may be related to the increased serum levels of endogenous cholecystokinin (CCK). [99] One study showed impaired gastric myoelectric activity in chronic pancreatitis [100]; this abnormality was restored toward normal with pancreatic enzyme replacement. Gastric emptying is variable in cystic fibrosis: early in the disease, it may be rapid; later in the disease, it is delayed. [101]

Liver disease. Abnormal scintigraphic gastric emptying has been reported in up to 78% of patients with cirrhosis of the liver. [102] The delay in gastric emptying is correlated with the severity of liver disease, as indicated by either Child-Pugh status, bilirubin level, prothrombin time, and presence of esophageal varices. [102] Secretin and endothelin have been suggested as playing a role in the gastric dysmotility with cirrhosis and portal hypertension. [103], [104] Delayed emptying has also been reported in patients with portal hypertension of either intrahepatic or extrahepatic pathology and does not correlate with corrected wedged hepatic venous pressure. [105] Other studies have not shown significant changes in gastric emptying [106] and have even reported acceleration of liquid phase gastric emptying. [107] Gastric emptying using the octanoate breath test may be prolonged in patients with cirrhosis, but normal with scintigraphy, because hepatic metabolism of octanoate may be abnormal. [108] Interferon-a treatment may delay gastric emptying and may cause dyspeptic symptoms; interferon treatment delayed gastric emptying in 12 of 14 patients and was associated with development of anorexia and abdominal discomfort. [109] Prokinetic therapy improved gastric emptying and GI symptoms during interferon-a therapy.

Constipation. Delayed gastric emptying is not uncommonly observed in patients with constipation. The finding of delayed gastric emptying suggests the possibility of a diffuse GI disorder. [110] This has implications for treatment; patients with chronic severe constipation and proximal GI tract dysmotility often have poor responses to colectomy. Volitional suppression of defecation can delay gastric emptying, suggesting a “cologastric brake” or the effects of stress. [111]

Diffuse gastrointestinal motor disorders. Gastroparesis can occur as a part of a diffuse GI motility disorder. *Chronic intestinal pseudoobstruction* is a syndrome with recurrent symptoms suggestive of intestinal obstruction in the absence of mechanical blockage. Symptoms are caused by ineffective peristalsis and include nausea, vomiting, and abdominal pain with abdominal distention. Radiologic findings consist of air-fluid levels within the small intestine. The term *diffuse GI motility disorder* rather than chronic intestinal pseudoobstruction, is often used if several portions of the GI tract have delayed transit but no air-fluid levels seen on abdominal radiographs. [19] Chronic intestinal pseudoobstruction can be caused by several systemic diseases including scleroderma, amyloidosis, myxedema, and long-standing diabetes mellitus. Often, it is idiopathic, with no known cause. The two main forms of idiopathic pseudoobstruction are myopathic and neuropathic. The bowel wall in the myopathic form (e.g., hollow visceral myopathy) shows thinning and degeneration of smooth muscle with replacement by fibrous tissue. The neuropathic form (e.g., visceral neuropathy) has normal smooth muscle histologically but abnormalities in neurons and glial cells within the splanchnic ganglia or myenteric plexus. Small bowel manometry may help to differentiate these two forms. In intestinal myopathy, low-amplitude contractions that propagate normally are seen. In intestinal neuropathy, individual contractions are of normal amplitude but disorganized: disruption of the phase III MMC, bursts of nonpropagating activity during fasting, and failure to convert from the fasting to the fed pattern with a meal are typical.

Nongastrointestinal Disorders Associated with Delayed Gastric Emptying

Eating disorders: anorexia, bulimia, rumination. *Anorexia nervosa* is a psychiatric disorder occurring primarily in adolescent and young adult women characterized by distorted body image and fear of obesity with compulsive dieting and self-imposed starvation to maintain a profoundly low body weight. GI symptoms are common and include lack of appetite, early satiety, epigastric fullness, abdominal bloating, nausea, and vomiting. [112], [113] Patients with anorexia nervosa often have delayed gastric emptying primarily for solids. [113] Interestingly, realimentation and maintenance of normal body weight improve gastric emptying and GI symptoms. [112], [114] Prokinetic therapy had been suggested to improve gastric emptying and to facilitate refeeding [113]; other studies have not shown an effect on weight gain. [115] Pathogenesis of gastric dysfunction in anorexia is probably multifactorial; starvation and malnutrition with marked endocrine disturbances may play a role; others have suggested a CNS disturbance related to the psychiatric abnormality. *Bulimia nervosa* is characterized by recurrent episodes of binge eating with a feeling of lack of control over the eating behavior during the binges, often followed by self-induced vomiting, the use of laxatives or diuretics, strict dieting or fasting, or vigorous exercise to prevent weight gain. [116] Self-induced vomiting allows bulimic patients to continue eating or to terminate the binge. In contrast to anorexia nervosa, concern about weight and body size does not lead to a fall in weight below normal. Symptoms of postprandial fullness, early satiety, bloating, nausea, and epigastric pain may

be occur. [117] Depression may be an important determinant of GI symptoms. [117] Gastric emptying studies in bulimia have conflicting results; some (37%) of bulimic patients appear to have delayed emptying. [116] *Rumination syndrome* is the effortless regurgitation of recently ingested food into the mouth with subsequent remastication and reswallowing or expectorating of food. Initially described in children and mentally retarded institutionalized persons, there is an increasing awareness of this disorder in adults of normal intelligence. Rumination can become a habit, often initiated by a belch, a swallow, or by stimulation of the palate with the tongue. Abdominal muscle contraction with LES relaxation in the early postprandial period is responsible for regurgitation. [118] Typically, the effortless repetitive regurgitation occurs within 15 minutes of starting a meal, in contrast to vomiting from gastroparesis, which occurs later in the postprandial period. [40] Symptoms often cease when regurgitated food becomes acidic to taste. Difficulty in diagnosis results from lack of awareness and the difficulty differentiating rumination from vomiting caused by gastroparesis or regurgitation from GERD. Gastric emptying and esophageal pH monitoring are usually normal. Gastroduodenal manometry shows characteristic brief simultaneous increases in gastric and small bowel pressure (R waves) seen in all abdominal recording ports during the postprandial period. These represent abdominal wall contractions, and, if performed with esophageal pH monitoring, they are associated with a decrease in distal esophageal pH. These characteristic findings are seen in only one third of patients with clinical symptoms of rumination. [40] Making the patient aware of this habit may be beneficial. Treatment is with behavioral modification and biofeedback therapy administered in a formal eating-regulation program. Concentration on diaphragmatic breathing after eating may help. Biofeedback techniques help to teach patients to relax abdominal muscle during and after eating. [119] Chewing gum postprandially has been suggested to be helpful.

Cyclic vomiting syndrome. *Cyclic vomiting syndrome* refers to recurrent episodes of intense nausea and vomiting lasting hours to days separated by symptom-free periods of variable lengths. Typically, each episode is similar. Vomiting often starts abruptly, although a prodrome of nausea and abdominal pain can occur. The vomiting reaches highest intensity during the first hours, diminishes thereafter, and ends rapidly. The duration of vomiting averages 3 to 4 days and can lead to marked dehydration. This disorder is well characterized in children, but it has also been described in adults. [120], [121] In adults, the vomiting episodes are longer (3 to 5 days), less frequent (every 3 to 4 months), and triggering events are less evident [121]; there is usually a long delay in diagnosis. Abnormal upper GI motility can be demonstrated by gastric emptying scintigraphy, antroduodenal manometry, and EGG even in the asymptomatic state; however, these tests are often normal. [122] Precipitating events, identified in a few patients, include onset of menstrual periods and stress. Cyclic vomiting syndrome, also referred to as *abdominal migraine or abdominal epilepsy,* is associated with migraine headaches in patients and other family members. The cause of this disorder is unknown: a mechanistic relationship to migraine, underlying hypothalamic-pituitary axis derangement, or an inappropriate activation of vomiting reflex has been suggested. [120] Mitochondrial disorders have been identified in some patients. [123] There is no standard management for this disorder. Sleep, a quiet environment, and the use of benzodiazepines such as lorazepam may be effective. [121] Tricyclic antidepressants (e.g., amitriptyline or doxepin) in low doses may prevent attacks [121]; others have suggested that ß-blockers such as propranolol may prevent attacks. Antiemetics such as ondansetron and prokinetic agents such as metoclopramide or erythromycin may provide temporary relief during vomiting episodes. Antimigraine therapies such as the 5-HT $_{1D}$ receptor agonist sumatriptan, given subcutaneously, may reduce the severity of attacks. The use of nonsteroidal antiinflammatory drugs, such as indomethacin or ketorolac, given intramuscularly, has been reported to prevent or reduce the intensity of attacks if these drugs are given during the prodromal period. [120] These agents inhibit prostaglandin formation and may correct gastric dysrhythmias or act centrally.

Neurological disorders. CNS causes of gastroparesis are rare. Any condition associated with increased intracranial pressure may produce emesis, with or without nausea, by activation of brainstem structures mediating vomiting. [16] Focal lesions including tumors, especially those involving the brainstem and posterior fossa, can cause nausea and vomiting and can even present with gastroparesis. Demyelinating diseases such as multiple sclerosis and other demyelinating disorders have been reported to cause gastroparesis. [124] An adult patient with vomiting from a central mass lesion usually has CNS symptoms or signs, such as headache, cranial nerve findings, long tract signs, or papilledema. [125] Rarely, objective neurological findings may be absent. [126] Magnetic resonance imaging is better than computed tomography for visualization of the posterior fossa. Gastroparesis is common in Parkinson disease, reported in 70% of patients. [127] The stomach can be affected by the underlying parkinsonian neurodegenerative pathology; Lewy bodies have been reported in the myenteric plexus of the stomach. [128] Medications used to treat Parkinson disease such as L-dopa can delay gastric emptying. Impaired gastric emptying with delayed levodopa absorption may be a reason for a poor response to medical therapy for Parkinson disease. [127] Domperidone may be a useful prokinetic agent in Parkinson disease; domperidone does not cross the blood-brain barrier and should not interfere with centrally acting antiparkinsonism medications. [129]

Collagen vascular disorders. *Scleroderma* (systemic sclerosis) is a systemic inflammatory disorder affecting the skin and other organs. GI manifestations usually follow cutaneous and other systemic manifestations. [130] The esophagus is involved in about 75% of patients, especially when Raynaud phenomenon is present. Myogenic abnormalities from smooth muscle fibrosis are seen: feeble middle and distal esophageal contractions and a low LES pressure. Gastric involvement is less frequent than that of the esophagus, small intestine, or colon. Gastric involvement has been reported in 10% to 75% of patients, commonly as delayed gastric emptying. Watermelon stomach is also reported associated with scleroderma. [131] Gastric emptying of radiopaque pellets was delayed in 50% of patients with systemic sclerosis. [131] Delayed gastric emptying is usually seen only in patients with esophageal transit abnormalities [132]; it can occur in some normal esophageal manometry. [131] Antroduodenal manometry classically reveals very low-amplitude antral and duodenal contractions with normal MMC propagation. [130] *Polymyositis and dermatomyositis* classically affect only the skeletal muscle, as recognized on esophageal manometry with dysfunction of the upper esophageal sphincter and upper esophagus. However, polymyositis and dermatomyositis may be associated with other GI motor dysfunction. Gastric emptying was found to be delayed in 8 of 13 patients and correlated with the severity of the peripheral (skeletal) muscle weakness. [133] Systemic lupus erythematosus rarely affects gastric motility. Delayed gastric emptying has been reported in a patient with systemic lupus erythematosus; functional gastric outlet obstruction with antral narrowing from thickened folds resolved with prednisone treatment. [134] *Amyloid-associated GI dysmotility* can be manifested as intestinal pseudoobstruction, diarrhea, or achalasia. [135] GI symptoms typically include anorexia, difficulty with eating from macroglossia, and altered bowel habits. Nausea may occur in up to 40% of patients with amyloidosis. [135] Clinical gastroparesis occurs rarely in patients with amyloidosis; one study reported only 3 cases in 769 patients. [136]

Endocrine and metabolic disorders: thyroid, parathyroid, renal failure. *Hypothyroidism* should be excluded in all patients with gastroparesis. Although it represents only a small percentage of patients with gastroparesis, it is easily treated with thyroid supplementation. There is poor correlation between the gastric emptying and the elevated thyroid-stimulating hormone level. [137] Intestinal pseudoobstruction has also been described in hypothyroidism. *Hyperthyroid* patients may be expected to have rapid gastric emptying; studies have reported normal, rapid, and delayed gastric emptying. [138], [139] Delayed gastric emptying with gastric dysrhythmias have been reported; these resolve with treatment of the hyperthyroidism. [139] The delayed gastric emptying has been suggested to result from associated autonomic dysfunction. [139] In both hyperthyroidism and hypoparathyroidism, small intestine pseudoobstruction has been reported. Parathyroid hormone has been shown to relax gastric and other GI smooth muscle directly. [140] Whether it influences gastric emptying is not known.

Chronic renal failure. GI symptoms, especially anorexia, nausea, and vomiting, are often present in renal failure. Symptoms may be from uremia, medications, gastric mucosal ulceration, or gastroparesis. Gastric emptying studies have yielded conflicting results. Delayed emptying has been seen in patients with symptomatic uremia. [141] Normal emptying is most commonly seen in patients undergoing hemodialysis. Delayed emptying has been reported to be associated with changes in nutritional status and autonomic neuropathy. [142], [143] Using the octanoate breath test, delayed gastric emptying was present in 62% of patients undergoing long-term hemodialysis; delayed emptying was present in 91% of patients with dyspeptic symptoms and in 40% of those without symptoms. [144] Chronic ambulatory peritoneal dialysis delays emptying, primarily when the abdomen contains the dialysate; emptying normalizes after the dialysate is emptied. [145]

Gastric infections. Delayed gastric emptying can be seen during a variety of acute *viral infections* including herpes zoster and infections with Epstein-Barr virus, CMV, rotavirus, and parvoviruses. [146] Nausea and vomiting have been associated with altered gastric motor function [147]; this has been shown during acute viral gastroenteritis with parvovirus-like agents (Norwalk and Hawaii viruses). Delay in gastric emptying is usually transient and returns to normal with recovery from the viral infection. In rare patients, chronic symptoms and delayed gastric emptying may result [148], [149]; this may be one cause of idiopathic gastroparesis. Gastric CMV occurs often in immunosuppressed persons, particularly organ transplant recipients. [150] Upper GI CMV infection has been reported to occur in one third of patients after liver transplantation, often associated with typical symptoms of gastroparesis including nausea and abdominal fullness. Endoscopy can show large antral folds, gastric inflammation including acute superficial gastritis, and duodenal erosions and ulcerations. Viral cultures of gastric biopsies and histological presence of CMV inclusions in the gastric mucosa are used for diagnosis. [150] The CMV infection can be treated with ganciclovir. GI infection with CMV is unusual in immunologically normal persons. A case report documents acute CMV infection causing GI dysmotility in an immunocompetent patient; the disturbance in GI motor function persisted for years after viral infection had been eradicated. [148] Gastric emptying may be delayed in one third of persons seropositive for human immunodeficiency virus (HIV), [151] particularly those with advanced disease evidenced by low CD4 counts, marked weight loss, and enteric infections. [152] In immunocompromised patients, the acute stage of gastritis and gastroparesis from viruses and other infectious agents, especially with CMV and *Mycobacterium avium-intracellulare,* may persist for prolonged times. Some investigators have found delayed gastric emptying of solids with diminished postprandial antral motility but rapid emptying of the liquids, suggesting an autonomic neuropathy that is common in HIV infection. [153] It had been speculated that *Helicobacter pylori* causes GI motor abnormalities. Although a few studies have suggested an association, [154] most show that *H pylori* does not cause delayed gastric emptying or functional dyspepsia. [155] One study reported that chronic antral *H pylori* gastritis was associated with a lower incidence of the phase III MMC starting in the antrum in dyspeptic patients. [156] One study suggests that dyspeptic patients with *H pylori* infection have a high prevalence of gastric dysrhythmias; eradication of *H pylori* normalized gastric myoelectric activity and improved symptoms. [157]

Chronic mesenteric ischemia (ischemic gastroparesis). Gastric ischemia may occur in chronic *atherosclerotic disease;* the gastric ischemia is insidious and often is not recognized. Ischemic gastropathy secondary to atherosclerosis may present as gastritis, ulceration, or gastroparesis. [158] Angiography demonstrates arterial occlusion. Cases of chronic *mesenteric ischemia* with chronic gastric ischemia causing gastroparesis and gastric dysrhythmias have been reported. [159] Surgical

revascularization may improves gastric emptying and corrects gastric dysrhythmias in select patients.

Gastroparesis after lung and heart lung transplantation. Gastroparesis has been reported after heart and lung transplantations. [160], [161] After lung transplantation, gastroparesis may predispose to gastroesophageal reflux with microaspiration and subsequent pulmonary infection, which can have deleterious effects on the transplanted lungs. Symptomatic delayed gastric emptying was present 25% of patients after single lung transplantation and in 50% of patients after combined heart and lung transplantation. [160] A more recent study reported delayed gastric emptying in eight of ten patients after combined heart and lung transplantation. [161] Several causes may explain gastroparesis after heart and lung transplantation: vagal nerve dysfunction, viral infection, and immunosuppressive medications. Vagal nerve dysfunction from thermal or ischemic injury or dissection of posterior mediastinum during surgery has been suggested as the most likely cause. [160], [161]

Gastroparesis secondary to malignant disease (tumor-associated gastroparesis). Tumor-associated gastroparesis has been described with esophageal, gastric, pancreatic, breast, and lung carcinoma. The pathophysiology is unknown, but frequently it is attributed to paraneoplastic effects, neural invasion of the tumor, or the side effects of chemotherapy. In *gastric cancers* that are nonobstructing, tumor infiltration into the stomach wall may disrupt smooth muscle or neural innervation. The symptoms and delay in gastric emptying can be responsive to prokinetic agents, including metoclopramide. [162] *Small bowel tumors* are rare and can be difficult to diagnose. Infrequently, patients initially diagnosed with gastroparesis are found, on further evaluation, to have a small bowel adenocarcinoma causing symptoms from partial obstruction. [163] These tumors can be missed on an SBFT examination. Enteroclysis provides a more detailed evaluation. In *pancreatic cancer,* delayed gastric emptying has been reported in 40% to 60% of patients without gastroduodenal invasion or obstruction. Usually, the gastroparesis is asymptomatic, but it may be responsible for the nausea, vomiting, and abdominal or back pain that occur. [164] Retroperitoneal nerve invasion of the tumor has been postulated as a cause of the gastroparesis, although gastroparesis can also occur with resectable cancers. *Pseudoobstruction with delayed gastric emptying* may represent a paraneoplastic syndrome that occurs primarily with small cell lung carcinoma. [165] Intestinal dysmotility is suspected to have an autoimmune basis, reacting with the myenteric plexus. Pathological studies have shown degeneration of the myenteric plexus with infiltration by plasma cells and lymphocytes and markedly reduced numbers of neurons. [166] The onset of GI dysmotility not infrequently precedes the diagnosis of cancer [165]; patients may have a normal chest radiograph at presentation with subsequent diagnosis of cancer by computed tomography, magnetic resonance imaging, or bronchoscopy. Antroduodenal manometry may show neuropathic abnormalities with incoordination of contractions. Type 1 antineuronal nuclear antibody (ANNA-1 or anti-Hu) can often be detected in paraneoplastic pseudoobstruction from small cell lung carcinoma. [165] Delayed gastric emptying with nausea and vomiting can occur during treatment of malignant disease. Gastroparesis can occur after radiation therapy for treatment of tumors, [167] during treatment with chemotherapeutic agents, [168] after bone marrow transplantation, [169] and after celiac plexus block for chronic pain from pancreatic cancer. [170]

Medication-induced gastroparesis. Nausea and vomiting can be side effects of medications. Nausea and vomiting in response to a drug are likely to present early in its use; medications usually cause acute rather than chronic nausea and vomiting. [16] Many drugs including anticholinergics, narcotic analgesics, tricyclic antidepressants, and calcium channel blockers are known to delay gastric emptying (see Table 65-2). Drug-induced gastroparesis may be asymptomatic, but it needs to be considered when delayed gastric emptying is found in a symptomatic patient; symptoms may not be from delayed gastric emptying. Gastric acid suppressing agents, such as histamine H_2 receptor antagonists and proton pump inhibitors, have been shown to delay gastric emptying. [171] Some studies, however, suggest that nizatidine, an H_2 receptor antagonist with cholinergic properties, may induce antral contractility and may promote gastric emptying. [172] Gastric emptying of solids is delayed in patients receiving total parenteral nutrition (TPN). [173] It is suggested that if assessment of gastric emptying and motility is performed in a patient who is receiving TPN, the TPN should be converted to saline for the test. [26] Slowing of gastric emptying during TPN is correlated with increase in blood glucose by the intravenous nutrients; hyperglycemia inhibits gastric emptying of solids. The inhibitory effect of hyperglycemia on gastric emptying may be mediated by a suppression of the vagal-cholinergic system. [173] Oral ingestion of fat is well known to delay gastric emptying through intestinal feedback and release of CCK. Intravenous infusion of fat emulsions also delays gastric emptying. [173], [174] The mechanism by which intravenous fat influences gastric motility is poorly understood; increased plasma CCK concentrations was reported in an initial study but was not confirmed in subsequent studies. [173], [174]

Treatment of Symptomatic Gastroparesis

The general principles for treatment of symptomatic gastroparesis are as follows: first, to correct fluid, electrolyte, and nutritional deficiencies; second, to identify and treat the underlying cause of gastroparesis; and third, to reduce symptoms. [16] The specific treatment depends on the severity of symptoms and the underlying disorder. For mild gastroparetic symptoms, dietary modifications and a low-dose prokinetic agent may be used. Medicines are reviewed to ensure that the patient is not taking an anticholinergic agent or narcotic analgesic that could delay gastric emptying or prevent the action of a prokinetic agent. For diabetic patients, blood glucose should be regulated carefully to prevent hyperglycemia. In severe cases of gastroparesis with vomiting, signs of dehydration, and poor glucose control, treatment may include hospitalization, intravenous hydration, nasogastric suction to decompress the stomach, regulation of blood glucose, and intravenous administration of antiemetic and prokinetic agents.

Dietary Treatment In gastroparesis, the diet should be modified to promote faster gastric emptying. Liquid nutrients in the diet should be increased, because emptying of liquids is usually less affected than solids. Because fat intake releases CCK, which can delay gastric emptying, low-fat meals with complex carbohydrates are suggested. Meals should be small to allow time for emptying to occur; gastric emptying of meals is regulated for a set number of calories to be emptied into the duodenum. [175] To compensate for small meals, patients may need to eat four or five times daily. A diet low in indigestible fiber and roughage will reduce the chance of bezoar formation. Alcohol and carbonated beverages should be avoided. Even low doses of alcohol can decrease antral contractility and impair gastric emptying. Carbonated beverages release carbon dioxide gas, which can aggravate gastric distention. Commercial or self-prepared liquid homogenized meals may be tolerated in small quantities. Larger volumes needed to cover daily caloric needs are often not. Sometimes enteral alimentation may be helpful. In some severe cases, TPN may be needed. This is not a long-term solution because of catheter-induced bacteremia, clot formation, and cost.

Glucose Control Diabetic patients with gastroparesis may have abnormally high glucose concentrations both after fasting and postprandially. Hyperglycemia itself has been shown to delay gastric emptying. [52], [53] This slowing is associated with reduced antral contractility and increased pyloric pressure. Hyperglycemia can inhibit the accelerating effects of prokinetic agents. [176] Improvement of glucose control can increase antral contractility, correct gastric dysrhythmias, and accelerate gastric emptying.

Antiemetic Agents Nausea and vomiting are treated with antiemetic agents. [16] Drug classes used to treat nausea and vomiting are phenothiazines, antihistamines, anticholinergics, dopamine receptor antagonists, and serotonin receptor antagonists. Phenothiazine compounds possess antiemetic properties. Their actions are mediated primarily through a central antidopaminergic mechanism at the chemoreceptor trigger zone in the area postrema of the medulla oblongata. [16] Commonly used agents include prochlorperazine (Compazine), trimethobenzamide (Tigan), and promethazine (Phenergan). These agents are commonly used for severe episodes of nausea and vomiting and are available as tablets, capsules, liquid suspensions, suppositories, and for injectable use. Sedation and extrapyramidal side effects may develop with these agents. [179] Antihistamines with H_1 receptor antagonistic properties have central antiemetic effects. [16] These include diphenhydramine (Benadryl), dimenhydrinate (Dramamine), and meclizine (Antivert). These agents exert antinausea effects by blocking H_1 receptors in the vestibular apparatus and the chemoreceptor trigger zone within the area postrema. In vection-induced motion sickness, dimenhydrinate reduced tachygastria, decreased symptoms, and caused drowsiness, suggesting that symptom improvement is by improvement of gastric myoelectric activity or depression of CNS activity. [178] Serotonin ($5\text{-}HT_3$) receptor antagonists, such as ondansetron (Zofran) and granisetron (Kytril), are useful in chemotherapy-induced nausea and vomiting. The primary site of action is likely the chemoreceptor trigger zone because of the high density of $5\text{-}HT_3$ receptors in the area postrema. [16] They may also act peripherally because $5\text{-}HT_3$ receptors are present in the GI tract. [16] If used, they are best given on an as needed basis because of the lack of evidence of prolonged efficacy and their expense. Benzodiazepines such as lorazepam and diazepam have been shown to be effective as adjunctive agents in the treatment of chemotherapy-related nausea and vomiting. [16]

Prokinetic Agents Prokinetic agents enhance contractility of the GI tract and accelerate the aboral movement of lumenal contents (Table 65-6). In general, prokinetic agents increase gastric antral contractility, correct gastric dysrhythmias, and improve antroduodenal coordination. Some of the prokinetic agents, metoclopramide and domperidone, also have antiemetic properties.

TABLE 65-6 Prokinetic Agents for Gastroparesis

Usually, prokinetic agents are administered 30 minutes before meals to maximize blood levels and gastric prokinetic effects at meal ingestion. In addition, a bedtime dose is often used so indigestible solids can be emptied from the stomach. Medications with anticholinergic activity may block the effects of prokinetics that work by facilitating acetylcholine release. In gastroparesis, pharmacological therapy with prokinetic agents is often needed on a prolonged basis. Because GI symptoms

correlate poorly with gastric emptying, the response to treatment is usually judged clinically; repeating gastric emptying tests is not necessary. Prokinetic therapeutic options are limited now that cisapride is not routinely available (see Table 65-6). Prokinetics available for clinical use are metoclopramide and erythromycin.

Metoclopramide. Metoclopramide (Reglan), a substituted benzamide structurally related to procainamide, has been used since the 1970s to treat gastroparesis. [179] Metoclopramide has both prokinetic and antiemetic effects. Metoclopramide releases acetylcholine from intrinsic cholinergic neurons and blocks peripheral dopamine receptors, where dopamine acts as an inhibitory agent of smooth muscle contraction. The prokinetic properties of metoclopramide are limited to the proximal GI tract. Metoclopramide increases amplitudes of esophageal contractions, resting lower esophageal sphincter pressure, gastric fundic and antral contractions and improves antropyloroduodenal coordination. Metoclopramide has antiemetic effects resulting both from dopamine antagonism in the chemoreceptor zone and from a central action at the vomiting center. Metoclopramide is approved for use in diabetic gastroparesis and for prevention of postoperative and chemotherapy-induced nausea and vomiting. Controlled trials have shown that metoclopramide may provide symptomatic relief while accelerating gastric emptying of solids and liquids in patients with idiopathic, diabetic, and postvagotomy gastroparesis and in patients with GERD. [180], [181] Metoclopramide has been reported to be effective in the short-term treatment of gastroparesis for up to several weeks. [180], [181] Long-term efficacy for metoclopramide has not been clearly demonstrated; its effect on gastric emptying may diminish during long-term treatment. [182] In diabetic gastroparesis, acute administration of metoclopramide accelerates emptying, but not after 1 month of treatment. [182] Metoclopramide may continue to relieve symptoms because of its antiemetic effects. The usual starting dose of metoclopramide in adults is 10 mg 30 minutes before meals and at bedtime. In patients not responding, the dose can be increased to 20 mg. In severe gastroparesis, oral metoclopramide may not be adequately absorbed because of vomiting or delayed gastric emptying; metoclopramide administered intravenously may improve gastric emptying. Metoclopramide may also be administered subcutaneously, by suppository, or even intraperitoneally in patients undergoing peritoneal dialysis. [183] Side effects of metoclopramide, resulting from its antidopaminergic properties, may occur in up to 30% of patients and are the major factor restricting its use. Acute dystonic reactions—facial spasm, oculogyric crisis, trismus, and torticollis—occur in 0.2% to 6% of patients. [184] These effects are usually observed during the first 48 hours of treatment and are more frequent with higher doses. Drowsiness, fatigue, and lassitude occur in 10% of patients. Mental depression may occur and may range from mild to severe. Metoclopramide can aggravate underlying depression. Other side effects may include restlessness, agitation, irritability, and akathisia. Increased prolactin release may result in breast engorgement, lactation, and menstrual irregularity. Side effects of long-term use of metoclopramide are of concern. Prolonged treatment with metoclopramide may result in parkinsonian-like symptoms, more commonly within the first 6 months after beginning treatment, but occasionally after longer periods. [184] These symptoms usually subside within 2 to 3 months after discontinuation of metoclopramide. Patients with Parkinson disease should be given metoclopramide cautiously, if at all. Tardive dyskinesia, consisting of involuntary movements of the face, tongue, or extremities, may occur with prolonged use and may not reverse after stopping the medication. The prevalence of tardive dyskinesia may range from 1% to 2% to as high as 15% when patients take metoclopramide for at least 3 months. [184] This effect is more common in elderly women.

Erythromycin and motilides. Erythromycin has prokinetic effects because it is a motilin receptor agonist. [185] Motilin, an endogenous GI peptide, plays a role initiating the gastric phase III of the MMC and is a potent stimulator of antral contractility. Erythromycin binds to motilin receptors located on smooth muscle and on cholinergic neurons; the latter appear to be important for actions in vivo. [185], [186] Erythromycin stimulates gastric emptying in diabetic gastroparesis, idiopathic gastroparesis, and postvagotomy gastroparesis. Erythromycin is the most potent prokinetic agent in accelerating gastric emptying. Erythromycin accelerates gastric emptying by increasing the amplitude and frequency of antral contractions. Interestingly, erythromycin has been reported to accelerate stomach emptying in postsurgical gastroparesis in which the antrum, the primary site of its motor effect, has been resected. [187] The prokinetic effect in this situation may result from a stimulatory effect on the fundus. Erythromycin is reported to be most effective when it is used intravenously during acute exacerbations of gastroparesis. [188] Intravenous infusion of erythromycin lactobionate (200 mg) accelerated emptying of solids in patients with diabetic gastroparesis. [189] In these same patients, after 4 weeks of oral erythromycin ethylsuccinate (250 mg orally three times daily, 30 minutes before meals), the magnitude of the acceleration was less than that observed in response to the single intravenous dose. Studies carried out over longer observation periods have reported a reduction of benefit over time with oral administration. Hyperglycemia attenuates the stimulation of antral contractility and gastric emptying by erythromycin. [176] The motilin receptor agonist ABT-229 was not efficacious in relief of postprandial dyspeptic symptoms in diabetic gastroparesis or functional dyspepsia. [190] This lack of efficacy has been attributed to motilin's propensity to increase fundic tone, impairing accommodation. Oral administration of erythromycin should be initiated at low doses, such as 125 to 250 mg three times daily. Many physicians prefer using erythromycin suspension; it takes less time to reach peak concentrations and is easier to adjust dosages. [191] One center reports use of intravenous erythromycin (100 mg every 8 hours) for severe refractory gastroparesis. [192] Side effects of erythromycin include nausea, vomiting, and abdominal pain.

Cisapride. Cisapride (Propulsid) is a serotonin (5-HT $_4$) agonist that facilitates release of acetylcholine from cholinergic nerves in the myenteric plexus throughout the GI tract. Cisapride stimulates antral and duodenal contractions, improves antroduodenal coordination, and accelerates gastric emptying. Cisapride accelerates gastric emptying and decreases symptoms in patients with gastroparesis, an effect that may last for a year. Cisapride also increases esophageal contractions and resting LES pressure. Cisapride was approved by the United States Food and Drug Administration (FDA) for nocturnal heartburn in patients with GERD. There have been rare instances of cardiac arrhythmias and sudden death. [16], [177] These were not due to cisapride's 5-HT $_4$ agonist properties, but rather they were an effect of the its molecular structure on cardiac potassium channels, prolonging the QT interval and predisposing to ventricular arrhythmias, particularly torsades de pointes. Cisapride is metabolized by the cytochrome P450 3A4 system; use of medications that interact with this enzyme system leads to increased cisapride blood levels and increased cardiac toxicity. Besides cardiac risks, side effects include diarrhea, lightheadedness, and abdominal cramps. In patients with a seizure focus, cisapride may rarely increase seizure activity. Cisapride is not available for routine clinical use in the United States; it can be obtained only through limited-access investigational study protocols.

Domperidone. Domperidone (Motilium) is a benzimidazole derivative and is a specific dopamine-2 receptor antagonist. The effects of domperidone in the upper GI tract are similar to those of metoclopramide. Domperidone stimulates upper GI motility by enhancing gastroduodenal contractions and coordination. Domperidone does not readily cross the blood-brain barrier; therefore, it is much less likely to cause extrapyramidal side effects than metoclopramide. [193] Domperidone has antiemetic properties by its actions at the chemoreceptor trigger zone on the blood side of the blood-brain barrier. Domperidone has been studied primarily in diabetic gastroparesis. It increases both solid and liquid emptying, especially in those patients with a severe delay in gastric emptying. [194] Symptomatic improvement may not correlate with improvement in emptying [8], [194]; symptom improvement may result from antiemetic effects. Symptomatic benefit of domperidone can occur in diabetic patients with normal gastric emptying. [195] The effect of long-term treatment with domperidone has been variable. At 6 weeks, the effect of domperidone on solid phase gastric emptying was lost, whereas that on liquid gastric emptying was maintained. [194] Another study found that nausea and vomiting were improved compared with baseline values after 6 weeks of treatment, but there was no change in solid phase gastric emptying. [195] Other studies have suggested that improvement in gastric emptying and symptoms was still present after 1 year of treatment. [129] Double-blind studies examining the effects of long-term treatment with domperidone on chronic nausea and vomiting have revealed symptomatic benefit; these observations, however, have not been uniform. [195] Domperidone has been suggested for therapy of nausea and vomiting in Parkinson disease, in which symptoms could be related to the dopaminergic agents (e.g., L-dopa) used for the gastroparesis associated with the disorder. [129] Domperidone would work peripherally for GI dysmotility and symptoms; L-dopa would still work centrally for the parkinsonian symptoms. The side effects of domperidone are usually secondary to increased prolactin levels, such as breast engorgement and galactorrhea. CNS side effects are minimal. Domperidone is not FDA approved in the United States, but it is available in Europe, Canada, Mexico, and Japan; in some countries it is an over-the-counter drug.

Bethanechol. Bethanechol (Urecholine) is a nonspecific cholinergic muscarinic receptor agonist. It enhances amplitude of contractions throughout the GI tract. Unfortunately, bethanechol does not coordinate contractions; gastric emptying and small bowel transit are not necessarily accelerated. Most gastroenterologists do not consider bethanechol to be a true prokinetic agent. [177] Occasionally, it may be helpful as adjuvant treatment in combination with prokinetic agents. The typical dose is 25 mg orally four times daily. The direct cholinergic action of bethanechol may also stimulate secretion of gastric acid and saliva. Potential side effects of bethanechol include increased salivation, blurred vision, abdominal cramps, and bladder spasm.

Octreotide. Somatostatin has variable effects on GI tract motor activity. Somatostatin inhibits antral motor contractions. In the intestine, somatostatin initiates ectopic myoelectric and motor fronts under basal conditions but inhibits fed motility. Octreotide is a cyclic analog of somatostatin administered by subcutaneous injection and induces small intestinal phase III–like contractile activity. It may improve small intestinal motor function in patients with scleroderma with small bowel involvement. [196] In patients with gastroparesis, octreotide initiates phase-III activity, which originates in the small intestine and inhibits postprandial antral contractility. [197] Thus, octreotide delays gastric emptying and may worsen symptoms in patients with gastroparesis. Clinically, it may be used for small bowel dysmotility. In this case, prokinetic agents are administered before meals and octreotide is given at bedtime. Prolonged usage may result in gallstones from inhibition of gallbladder emptying.

Refractory Gastroparesis There is no uniform way to handle patients with refractory gastroparesis—patients not responding to antiemetic and prokinetic agents or with side effects preventing their use. In one study, of 110 patients with "refractory" gastroparesis, [1] 74% responded to use of another prokinetic agent, and only 26% were refractory to all prokinetic agents. These truly refractory patients underwent enteral or parenteral feedings and gastric electrical stimulation, and some underwent gastrectomy. Poor responders to prokinetic agents include postgastrectomy patients, those with myopathic connective tissue disorders, juvenile-onset diabetic patients with severely delayed gastric emptying, and patients with idiopathic gastroparesis with abdominal pain. [1] An initial approach to refractory gastroparesis includes ensuring that gastroparesis is responsible for the patient's symptoms, optimizing the patient's current therapy, and changing prokinetic agents. [177] Why some patients respond to one prokinetic agent and not another is uncertain. Different prokinetic agents have different mechanisms of action (see Table 65-6). The efficacy of most prokinetic agents diminishes with prolonged use. A "drug holiday," as used for L-dopa in Parkinson disease, can be used for prokinetic agents. In patients who remain refractory to treatment, one may consider placement of a venting gastrostomy or a feeding jejunostomy. Newer therapies include gastric electric stimulation and injection of botulinum toxin into the pylorus. Double-blind studies are not available to demonstrate efficacy of these modalities. TPN, if used, should be temporary, owing to the risk of infection and deep venous thrombosis. Surgical subtotal gastric resection, usually accompanied by a Roux-en-Y diversion, should generally be

discouraged. Completion gastrectomy can be considered for selected patients with postsurgical gastroparesis.

Combination prokinetic therapy. Prokinetic agents have different mechanisms to enhance gastric emptying (see Table 65-6). Addition of a second prokinetic agent may augment the response of another. Combination prokinetic therapy with two different agents may be useful to accelerate emptying and reduce symptoms in some refractory gastroparetic patients. [198] The combination of the available prokinetic agents, metoclopramide and erythromycin, has not been studied.

Newer prokinetic agents. Newer prokinetic agents being tested for gastroparesis include 5-HT $_4$ receptor agonists (tegaserod and cilansetron), dopamine antagonists (levosulpiride), CCK antagonists (loxiglumide and dexloxiglumide), and several motilin agonists. Tegaserod, a partial 5-HT $_4$ receptor agonist, reduces symptoms in patients with constipation-predominant irritable bowel syndrome. Tegaserod has been shown to accelerate gastric emptying and small bowel transit. [199] It is being evaluated for gastroparesis. Levosulpiride, an antidopaminergic agent, has antiemetic and prokinetic effects. [200] In diabetic gastroparesis, levosulpiride has accelerated gastric emptying and improved symptoms; however, there was no correlation between the two. Loxiglumide, a CCK-A receptor antagonist, has been shown to increase antral contractility and to accelerate gastric emptying in normal subjects. [201] This also suggests that endogenous CCK may have a role in slowing gastric emptying. New studies are being conducted with its dextroisomer, dexloxiglumide. [202] Clonidine, an a $_2$-adrenergic agonist, has been reported to decrease symptoms and to accelerate gastric emptying in patients with diabetic gastroparesis. [203] Other studies have shown that clonidine reduces dyspeptic symptoms by improving gastric accommodation. [204] Some studies report clonidine delays gastric emptying. [205] A side effect is hypotension; clonidine should be used cautiously, especially in diabetic patients with autonomic dysfunction. Acetylcholinesterase inhibitors, such as physostigmine and neostigmine, may stimulate GI contractility by increasing acetylcholine levels with subsequent muscarinic receptor activation. Anticholinesterase agents may increase contractility, but they may not increase antroduodenal coordination; therefore, they may not increase emptying. [206] In animal studies, GI transit is affected in an inverted U-shaped dose response curve with acceleration at lower doses but inhibition at higher doses. [207] This dual effect may be related to activation of different muscarinic receptor subtypes. Some H $_2$ receptor antagonists, such as ranitidine and nizatidine, have been reported to have anticholinesterase activity. [171], [172]

Gastric electric stimulation. Gastric electric stimulation is an emerging treatment for refractory gastroparesis. There are several techniques for stimulating the stomach. First is gastric electrical pacing; the goal is to entrain and pace the gastric slow waves with high-energy, long-duration pulses. Pacing at 10% higher than the basal rate has been shown to accelerate gastric emptying and to improve dyspeptic symptoms. [208] Second is high-frequency, low-energy, short-pulse-duration stimulation. This has been shown to decrease symptoms with little effect on gastric emptying [209], [210]; it may activate sensory afferent nerves to reduce symptoms. Finally, sequential muscle stimulation using bursts of suprahigh-frequency stimulation to induce direct gastric muscle contractions in a peristaltic sequence has accelerated emptying in animal studies. [211] High-frequency gastric electric stimulation at 12 cpm has FDA humanitarian approval for treatment of chronic, refractory nausea and vomiting secondary to diabetic or idiopathic gastroparesis. Stimulating wires are placed into the gastric muscle at the greater curvature during laparoscopy or laparotomy. These leads are attached to an electric stimulator ("pacemaker"), which is placed in a subcutaneous abdominal pouch. An initial study demonstrated effectiveness in 20 of 26 patients with decrease in nausea and vomiting at 3 and 6 months; gastric emptying of liquids, but not for solids, was improved. [209] In long-term follow-up, 3 of 24 patients underwent total gastrectomy because of unsatisfactory results, and 3 had the stimulator removed because of erosion or infection. A subsequent study reported on 33 patients with chronic gastroparesis. [210] After implantation, the electrical stimulator was turned on or off in a randomized, double-blind, crossover design. Patients felt better when the stimulator was on, although the decrease in vomiting that occurred was not statistically significant. Long-term follow-up over 1 year showed a decrease in the mean vomiting frequency from 25 to 6 times per week. In addition, there was an improvement in the overall quality-of-life score and a mild improvement in gastric emptying. Complications were infection in 3 patients and electrode penetration of the stomach wall in 1 patient. An open-label study in 25 patients reported an overall improvement of 40% from "severe" to "moderate" in symptoms of nausea and vomiting. [212] There was an infection rate of 15%, requiring removal of the stimulator. Carefully designed studies are needed to determine the overall effectiveness of gastric electrical stimulation, which type of patient will likely respond, and optimal stimulation parameters.

Botulinum toxin injection into the pyloric sphincter. Gastric emptying is a highly regulated process reflecting the integration of propulsive forces of proximal fundic tone and distal antral contractions with inhibitory forces of pyloric sphincter resistance. Few studies have been performed to see whether inhibition of the pyloric sphincter muscle accelerates gastric emptying. Early studies with surgical pyloromyotomy did not show benefit; more recent studies have shown a beneficial response in diabetic gastroparesis. [212] Pyloromyotomy may be helpful in preventing gastric stasis after truncal vagotomy. Botulinum toxin is a potent inhibitor of neuromuscular transmission and has been used to treat spastic disorders of muscles by local injection into effected muscles. Injection of botulinum toxin directly into the LES reduces pressure and improves symptoms in patients with achalasia. [213] In several small series of four to eight patients with gastroparesis, botulinum toxin has been injected into the pyloric sphincter in an attempt to reduce gastric outlet resistance and to accelerate gastric emptying in patients with idiopathic and diabetic gastroparesis. [214], [215] Overall, these open-label studies have shown a mild improvement in gastric emptying and modest improvement in symptoms that may last for several months.

Alternative and unconventional medical therapy primarily for nausea and vomiting. Tricyclic antidepressants may be of some benefit in patients with functional vomiting. [216] Low doses may have an effect to reduce afferent sensory transmission. Some of these agents, especially the tertiary amines such as amitriptyline (Elavil), have anticholinergic properties and may delay gastric emptying. Stimulation of the PC6 acupuncture point on the wrist with conventional acupuncture, electroacupuncture, or acupressure with a wrist band (e.g., Sea-band) may help to control postoperative nausea, chemotherapy-induced nausea, and nausea associated with morning sickness. [217], [218] and [219] These studies also suggested acupressure had similar efficacy to standard antiemetic therapy for postoperative nausea. [217] Another acupuncture point that has been used for influencing gastric motility is the Zusanli point (ST36), located on the leg below the patella. [220] Electroacupuncture reduces the severity of symptoms of vection-induced motion sickness while decreasing gastric tachyarrhythmias. [221] Animal studies suggest that electroacupuncture enhances gastric emptying by vagal stimulation. Electroacupuncture has been postulated to modulate serotonin, substance P, and endogenous opiates along CNS pathways. [223] Naloxone, an opiate receptor antagonist, blocks the analgesic effect of acupuncture and worsens symptoms of motion sickness. [223] Ginger, a traditional Chinese herbal remedy, reduces nausea and associated tachygastria caused by circular vection, an experimental model of motion sickness. Ginger also has been shown to reduce hyperglycemia-induced gastric dysrhythmias and nausea. [224]

Venting gastrostomy and feeding jejunostomy tubes. In the treatment-refractory patient with severe nausea and vomiting, placement of a gastrostomy tube for intermittent decompression by venting or suctioning may provide relief to the patient who has prominent abdominal distention. The gastrostomy tube can be opened for drainage or aspirated to decompress the stomach; this can be employed when symptoms are distressing to alleviate nausea, pain, and bloating. Venting gastrostomy tubes may be placed by the endoscopist, surgeon, or interventional radiologist. In one series, six of eight patients were able to return to full-time work or school. [225] The percutaneous endoscopic gastrostomy tube can be converted to a button. Use of a gastrostomy tube for feeding may aggravate gastroparetic symptoms. A feeding jejunostomy is usually preferred. Although a J-tube extension can be placed through the gastrostomy, these often prolapse into the stomach. To prevent this, enteral feedings through a direct jejunostomy tube can be helpful. Jejunostomy tubes are effective for providing nutrition, fluids, and medications, provided the small intestine is functioning. [226], [227] Carefully regulated nutrient enteral infusion may allow better glucose control in diabetic patients whose glycemic control is otherwise poor because of vomiting and gastroparesis. [227] Jejunostomy tubes are usually inserted laparoscopically or at laparotomy [228]; in some centers they are placed endoscopically.

Surgical treatment. Surgical treatment of idiopathic and diabetic gastroparesis has generally been disappointing. [16] Surgery is performed only as a last resort. The procedure usually employed is a partial gastrectomy and Roux-en-Y gastrojejunostomy. For selected patients with postsurgical gastroparesis in whom medical therapy has failed, further resection of the stomach may occasionally be considered. Usually, these patients have had prior vagotomy. Postsurgical gastroparesis patients require radical surgery to eliminate hold-up in the atonic stomach and creation or revision of a Roux limb to prevent enterogastric reflux. The resection is performed as a completion of the gastrectomy or a near-total gastrectomy, rather than as a less extensive resection. [229], [230] In near-total gastrectomy, only a small rim of proximal stomach (<1 cm) is retained. Reconstruction is achieved by a Roux loop of at least 45 cm to the rim of the stomach. The extensive gastrectomy is to eliminate the gastroparetic stomach and to allow ingested nutrients to pass directly from the esophagus into the small intestine. The Roux-en-Y jejunal segment helps to prevent reflux of small intestinal contents into the remaining stomach and esophagus. This extensive surgery may lead to improvement in two thirds of patients in specialized centers. [229] In a series of 62 patients who underwent completion gastrectomy for severe postvagotomy gastric stasis, good symptomatic improvement in nausea, vomiting, and postprandial abdominal pain was obtained in only 43% of patients, without improvement in chronic pain, diarrhea, and dumping symptoms. [231] The combination of nausea, need for TPN and retained food at endoscopy predicted a poor outcome. In another series of 81 patients with severe postsurgical gastroparesis, near-complete gastrectomy with a 55-cm Roux-en-Y reconstruction was performed. Follow-up averaged 56 months for 52 patients; 78% reported improvement of their GI symptoms, 7% believed that there was no change, and 15% stated that their condition had worsened. [232] A subtotal (70%) gastrectomy with resection of antrum and pylorus, closure of the duodenum, and restoration of GI continuity with a 60-cm Roux-en-Y jejunal loop was reported in four patients with insulin-dependent diabetes with intractable vomiting from gastroparesis. [233] Three of the four patients did well, eliminating frequent hospital admissions.

Pancreatic transplantation for diabetic gastroparesis. Pancreatic transplantation and simultaneous kidney and pancreatic transplantation are being performed more often for insulin-dependent diabetes mellitus. With successful pancreatic transplantation, postprandial hyperglycemia resolves, and insulin therapy can be discontinued. [234] Simultaneous pancreatic and kidney transplantation helps to correct both uremia and hyperglycemia. Pancreatic transplantation has been shown to halt progression and slightly improve diabetic polyneuropathy. [235] Improvement during the first year after pancreatic transplantation may be primarily the result of correction of hyperglycemia, which affects nerve function. A relatively small degree of improvement in peripheral nerve function has been reported over time. [235] Gastric emptying may be improved in some patients after pancreatic transplantation, although the literature is sparse. One study reported, at 6 months after pancreatic transplantation, an improvement in liquid emptying in six of eight patients with previously delayed emptying, but no improvement in solid phase emptying. [234] Another study found emptying improved 1 year after pancreatic and kidney transplantation in 8 of 23 patients. [236] Despite limited improvement in gastric emptying,

symptoms and gastric dysrhythmias improved in diabetic patients after organ transplantation. [234], [236]

DISORDERS WITH RAPID GASTRIC EMPTYING

Postsurgical Dumping Syndrome

Description and Clinical Features Rapid gastric emptying and its consequences are responsible for *dumping syndrome,* symptoms of which include postprandial sweating, weakness, orthostasis, tachycardia, and diarrhea. Symptoms can be debilitating. Dumping syndrome, especially of liquids, occurs almost exclusively after gastric surgery and is the most common postgastrectomy syndrome, occurring in up to 20% of patients after gastrectomy (usually antrectomy) or truncal vagotomy with a drainage procedure (see Table 65-5). Postsurgical pathophysiological abnormalities causing the dumping syndrome include the following:

- Loss of proximal stomach receptive relaxation and accommodation secondary to vagotomy
- Decreased gastric capacity after gastrectomy
- Loss of controlled emptying from bypass or ablation of pyloric sphincter
- Loss of duodenal feedback inhibition resulting from a bypassed duodenum from gastrojejunostomy. [237], [238]

Dumping symptoms are often characterized as early or late relative to their time course after meal ingestion. Early dumping symptoms occur in the first 30 minutes and result from accelerated early gastric emptying of liquids with rapid filling of the intestine with hypertonic fluid, leading to bloating, crampy abdominal pain, and explosive diarrhea. Rapid filling of the intestine is associated with osmotic fluid shifts into the gut lumen and reduction of plasma volume resulting in secondary release of vasoactive substances that cause vasomotor symptoms such as lightheadedness, sweating, flushing, and palpitations with tachycardia. Late dumping symptoms occur 2 to 3 hours after a meal and are associated with reactive hypoglycemia (weakness, palpitations, and diaphoresis); these result from the hyperinsulinemic response to an overwhelming carbohydrate load. [239] Because of its relatively long half-life, increased serum concentrations of insulin and associated hypoglycemia persist for relatively long periods. Enteroglucagon is also released excessively in response to the high carbohydrate concentrations in the small bowel, and it sensitizes pancreatic ß-cells to hypersecrete insulin. [239]

Diagnosis Dumping syndrome is usually diagnosed clinically, based on the characteristic symptoms occurring in patients after gastric surgery. Another postsurgical disorder is postvagotomy diarrhea; in this disorder, the diarrhea is not related to meals, and postprandial hypotension and hypoglycemic are not seen. Scintigraphic studies of both solid and liquid phase can document rapid gastric emptying. The rapid emptying is most notable immediately after the meal, occurring in the first 15 minutes. [240] In patients with suspected dumping syndrome, gastric emptying of liquids may be the preferable measurement. Orocecal transit time may also be helpful. Responses to an oral hypertonic glucose load (50 g in 300 mL) have been suggested to be helpful in diagnosing dumping syndrome [241]; responses consistent with dumping include the following:

- A rise in hemoglobin in the early postprandial period suggesting hemoconcentration
- An increase in heart rate greater than or equal to 10 beats per minute
- Elevated plasma glucose early postprandially
- Low plasma glucose concentrations late postprandially.

Treatment Postsurgical dumping symptoms may resolve over time with conservative management. Specific treatment of the dumping syndrome is aimed at slowing delivery of nutrients into the small intestine and minimizing release of endogenous vasoactive substances. Initial dietary recommendations are to eat small, dry, frequent meals, to increase protein and fat content in the diet, to reduce the intake of carbohydrates by avoiding simple sugars, and to minimize liquid intake. [237] Lying down in a supine position after drinking liquids may delay liquid emptying. Increasing the viscosity of liquid meals by adding gel-forming carbohydrates (pectin or guar gum) may delay transit time and may slow carbohydrate absorption. Late dumping symptoms from hypoglycemia are usually relieved by ingestion of simple carbohydrates. Medical therapy has centered on delaying gastric emptying. Anticholinergic agents have been employed, but they are usually not beneficial. The somatostatin analog, octreotide, is the primary pharmaceutical treatment. Octreotide has a much longer half-life than somatostatin and is administered subcutaneously to prevent breakdown by gastric acid. Octreotide has effects on both GI transit and GI peptide release. Octreotide delays gastric emptying and prevents the accelerated emptying seen in dumping syndrome. [237] Octreotide also inhibits the release of enteric hormones and insulin secretion; this may actually be the primary mechanism of its symptomatic benefit. [239] Octreotide, 50 µg subcutaneously 30 minutes before meals, reduces symptoms (diarrhea, lightheadedness, and palpitations) and signs (orthostatic hypotension, tachygastria) and metabolic alterations (late hypoglycemia) with dumping syndrome. [239] This dose may be increased to 100 µg three times a day if needed. Side effects of octreotide when administered subcutaneously include injection site pain, iatrogenic diabetes, malabsorption with a worsening of diarrhea, and cholelithiasis. Less information is available regarding the long-term utility of octreotide and use of the newer long-acting depot form of octreotide. The fatty acid oleic acid, administered before a meal, may slow GI transit and may decrease diarrhea in patients with rapid GI transit including dumping syndrome. Oleic acid activates nutrient-triggered intestinal inhibitory feedback mechanisms (jejunal-ileal brake). [242] Revisional gastric surgery is performed in a few patients with severe dumping symptoms, with the aim to delay stomach emptying to alleviate symptoms. The results are variable and often disappointing. [237] Because of the risk of causing severe gastric stasis, surgery is recommended only when other measures have failed. Surgical procedures include takedown of a prior gastrojejunostomy with conversion to Billroth I gastroduodenostomy. A commonly used surgical procedure is creating a Roux-en-Y diversion, which has been reported to slow gastric emptying. [243] Insertion of an antiperistaltic jejunal segment between the stomach and the small bowel has been performed to produce a functional delay of nutrient outflow from the stomach. [243] Pyloric reconstruction has also been attempted.

Other Disorders with Rapid Gastric Emptying

Type II Diabetes Mellitus Accelerated gastric emptying has been reported early in *type 2 diabetes* and rarely in early type I diabetes. [50], [51] Although this condition primarily applies to liquids, rapid emptying of a high-carbohydrate solid meal may occur. Rapid emptying may result from impaired fundic accommodation from vagal neuropathy with reduction of inhibitory nitric oxide nerves. [47] Accelerated gastric emptying may result in faster intestinal absorption of glucose and increased postprandial glucose levels in part owing to a mismatch between absorbed food and insulin release. Excessive hepatic glucose release also contributes to postprandial hyperglycemia. [51] Slowing gastric emptying has been suggested to improve control of postprandial glucose in type 2 diabetes. [244] This may be the means by which the addition of dietary soluble fiber has been shown to improve glucose control. [245], [246] Agents that slow gastric emptying are being developed to improve glucose tolerance for diabetic patients; these include amylin and its analog pramlintide, CCK analogs, protease inhibitor II, and glucagon-like peptide 1. [244]

Pancreatic Exocrine Insufficiency In *pancreatic insufficiency,* accelerated gastric emptying of fatty meals has been reported. [247] Maldigestion and malabsorption of lipids may lead to the inability of unhydrolyzed fats to release CCK, which serves as a feedback inhibitor of gastric emptying. Pancreatic enzyme replacement slows the rapid gastric emptying. [247] Other studies report normal gastric emptying in pancreatic insufficiency. [248] Chronic pancreatitis has also been reported associated with delayed gastric emptying (see earlier). [99]

Duodenal Ulcer Some patients with duodenal ulcer have rapid gastric emptying of liquids with increased acid secretion. [249] Rapid gastric emptying of acid may exceed duodenal buffering capacity leading to mucosal injury. One study reported that enhanced gastric emptying was still present in patients with healed duodenal ulcers. Many of these studies were performed before the recognition that *H pylori* is an important factor in ulcer disease and acid secretion.

Zollinger-Ellison Syndrome Patients with *Zollinger-Ellison syndrome,* characterized by hypergastrinemia and increased gastric acid secretion, can have duodenal ulcers, diarrhea, or marked esophagitis. The diarrhea is usually attributed to massive gastric acid secretion leading to an increased amount of liquids propelled into the small intestine. These patients may have accelerated gastric emptying of liquids. However, gastric acid suppression may not normalize the rapid gastric emptying, suggesting the rapid emptying may not be related to gastric acid hypersecretion. [250] Pentagastrin administration also does not accelerate gastric emptying in physiologically normal subjects; instead, it seems to delay stomach emptying of solids. [250]

Hyperthyroidism Orocecal transit time is rapid in *hyperthyroidism* and may be a factor in hyperthyroid-associated diarrhea. [251] It returns to normal after reestablishment of the euthyroid state. Rapid gastric emptying and intestinal transit of barium have been reported. There are case reports of rapid gastric emptying using scintigraphy in hyperthyroidism [138]; most patients with hyperthyroidism appear to have normal gastric emptying [252]; some have delayed. [139]

Obesity *Morbidly obese patients* may have accelerated gastric emptying. [253] Dieting with weight loss has been shown to decrease this accelerated gastric emptying to a near-normal rate. [253] Gastric volumes are also larger than in normal-weight persons; this may cause satiety signals not to be triggered in response to meal ingestion and gastric distention. In addition, intestinal absorption of nutrients may be more efficient in obesity. [254]

FUNCTIONAL DYSPEPSIA

Definition and Subgroups

Dyspepsia refers to symptoms originating in the upper GI tract; the term is used to describe upper abdominal pain or discomfort often exacerbated with eating, early satiety, postprandial abdominal bloating or distention, and nausea. [255] Structural or organic disorders causing dyspepsia are gastroduodenal ulcer, gastritis, GERD, medication side effects, and gastric cancer. In most patients, no cause is apparent, and the dyspepsia is idiopathic, that is, functional or nonulcer dyspepsia. The 1999 Rome II criteria [256] for diagnosing functional dyspepsia include the following:

- Persistent or recurrent pain or discomfort centered in the upper abdomen
- Symptoms for at least 12 of the preceding 52 weeks
- No evidence of organic disease (including upper endoscopy) that is likely to explain the symptoms
- No evidence that dyspepsia is exclusively relieved by defecation or associated with the onset of a change in stool frequency or stool form (i.e., not irritable bowel syndrome). [256]

The Rome group [256] suggested that it may be useful to subcategorize functional dyspepsia into ulcer-like, dysmotility-like, and nonspecific dyspepsia based on the predominant or most bothersome symptom. In ulcer-like dyspepsia, pain in the upper abdomen is the predominant symptom. Dysmotility-like dyspepsia is characterized by symptoms of fullness, early satiety, bloating, and nausea. Nonspecific dyspepsia is used if there is no predominant symptom. In the 1991 Rome I criteria, reflux-like dyspepsia, predominantly heartburn or regurgitation accompanying dyspeptic symptoms, had been a subcategory of functional dyspepsia, but this symptom complex is now considered to be primarily GERD. The usefulness of these symptom-based subgroups has been questioned because there is overlap among them, and they are not helpful in predicting treatment responses. [257] The Rome II modifications of predominant symptoms for subcategorization may help to reduce this overlap. [258]

Pathogenesis with Emphasis on Gastric Dysmotility

Numerous mechanisms have been proposed for the pathogenesis of functional dyspepsia. [259] The *gastric acid or inflammation hypothesis* suggests that gastric acid or inflammation from acid, bile, or *H pylori* infection is responsible for symptoms. This acid hypothesis also includes occult GERD and postinfectious dyspepsia. [260] The *motor disorder hypothesis* suggests that gastric dysmotility disorders such as gastroparesis, impaired fundic accommodation, antral distention, or gastric dysrhythmias are important. The *visceral hypersensitivity hypothesis* suggests exaggerated symptoms in response to physiochemical stimuli such as distention, contraction, acid, and bile. The *psychological or psychiatric hypothesis* proposes that symptoms are related to depression, anxiety, or a somatization disorder.

Gastrointestinal Dysmotility Motility and sensory abnormalities of the stomach and upper small bowel have been found in many patients with unexplained dyspepsia. Delayed gastric emptying, impaired gastric accommodation to a meal, gastric dysrhythmias, and visceral hypersensitivity are important pathophysiological factors in functional dyspepsia. One or more of these factors may be detected in one half to two thirds of patients with functional dyspepsia. [261]

Delayed gastric emptying and antral hypomotility. Delayed gastric emptying is present in approximately 35% of patients with functional dyspepsia, with studies ranging between 25% and 80%. [12], [261] Differentiation between the dysmotility-like dyspepsia form of functional dyspepsia and idiopathic gastroparesis may be arbitrary. Specific dyspeptic symptoms and their severity have correlated poorly with the degree of gastric stasis. [10], [262] However, in a series of 343 patients with functional dyspepsia, the severity of postprandial fullness and vomiting correlated with delayed gastric emptying, which was present in 34% of the dyspeptic patients. [12] Detection of delayed gastric emptying in functional dyspepsia may have prognostic importance because these patients may respond better to prokinetic agents than patients with normal emptying. [13] However, this link of prokinetic agents in improving gastric emptying and symptoms is inconsistent; both delayed emptying and symptoms may be improved by prokinetic therapy; at other times, emptying or symptoms have improved, but not both. [13] Antroduodenal manometry has been used to study patients with functional dyspepsia. The most common abnormality, present in about half of patients, is postprandial antral hypomotility. [263] Antral hypomotility correlates with delayed gastric emptying. Several studies have suggested that small bowel motor dysfunction occurs in a large proportion of patients with nonulcer dyspepsia [263]; other investigators have reported small intestinal dysmotility in only a small percentage. [264] Small intestinal abnormalities may include retrograde, simultaneous, or reduced frequency of fasting MMC activity, as well as a reduced percentage of MMCs that start in the antrum. In the fed state, decreased or low-amplitude duodenal contractions may occur. In a study of 41 consecutive patients with severe chronic dyspepsia, a neuropathic disorder (contractions of normal amplitude but abnormal propagation) was present in 39%, myopathy (low-amplitude contractions with normal coordination) in 2%, and normal small bowel motility in 59%. [265]

Regional gastric dysfunction: impaired fundic accommodation and antral distention. Regional gastric function abnormalities may be present in many dyspeptic patients and appear to correlate with dyspeptic symptoms. Normally, the stomach accommodates a meal by the relaxation of the gastric fundus and corpus, providing the meal with a reservoir and enabling a volume increase without a rise in intragastric pressure. Reduced postprandial fundic relaxation and accommodation were found in 40% of patients with functional dyspepsia. [266] Impaired proximal gastric accommodation was associated with early satiety and subsequent weight loss and may be related to a vagal defect. Other studies have not found a clear relationship between postprandial symptoms and proximal stomach function. [267] Scintigraphy and ultrasound studies have shown that, in functional dyspepsia, the intragastric distribution of the meal is characterized by rapid transit of ingested food from the proximal stomach to the distal stomach (Fig. 65-3). [268] Rapid transit from the proximal stomach may result from poor receptive relaxation or accommodation of the proximal stomach. Rapid proximal gastric transit causes abnormally large quantities of food to enter the antrum immediately after ingestion. Early antral distention occurs in many dyspeptic patients. [268], [269] The degree of postprandial abdominal discomfort has been related to the degree of impaired accommodation of the proximal stomach and antral distention. [270]

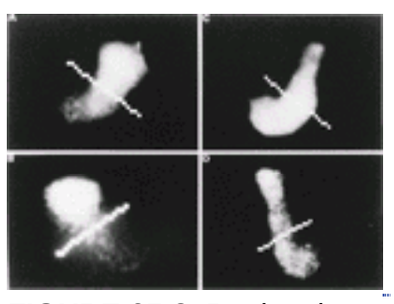

FIGURE 65-3. Regional gastric emptying abnormalities in functional dyspepsia. (**A** and **B**) Anterior and posterior views taken immediately after food ingestion from a physiologically normal subject. (**C** and **D**) Similar views from a dyspeptic patient. In normal subjects (**A** and **B**), food remained predominantly in the proximal half of the stomach after ingestion and then redistributed to the distal half over time. In dyspeptic patients (**C** and **D**), initial activity on the proximal half was lower because of rapid progression to the distal portion of the stomach. (From ref. [268].)

Radionuclide imaging of the gastric wall with single photon emission computed tomography (SPECT) to calculate gastric volumes has been used as a noninvasive measure of gastric accommodation. [271] SPECT imaging in 32 patients with functional dyspepsia suggested that impaired gastric accommodation (41%) may occur more commonly than delayed emptying (9%) in dyspeptic patients referred to tertiary centers. Most of the dyspeptic patients were found to have normal gastric emptying and accommodation. [271] Satiety testing with water or a nutrient drink has been suggested as a noninvasive technique to evaluate impaired accommodation and sensation. One example is the water load test, in which the subject drinks water until full. [272] Functional dyspeptic patients have reduced consumption. Another is a slow caloric drinking test in which Nutridrink (N. V. Nutricia: Zoetermeer, The Netherlands) is ingested at 15 mL/min. [273] This satiety test is correlated with gastric accommodation to a meal, as measured by the barostat. These satiety tests offer the potential to evaluate gastric accommodation in a noninvasive way. A reduced tolerated volume, however, may reflect either impaired accommodation or hypersensitivity of the stomach. [23]

Gastric myoelectric abnormalities. Gastric myoelectric abnormalities measured by EGG, such as tachygastria and bradygastria and decreased ratio of postprandial to fasting power, are found in about 40% of patients with functional dyspepsia. [11], [272], [274], [275] The prevalence of EGG abnormalities is similar to delayed gastric emptying in functional dyspepsia. EGG and gastric emptying tests appear to complement each other in correlating symptoms to gastric dysmotility. [11] How EGG results affect patient management is controversial. In two small studies, dyspeptic patients with an abnormal EGG had a more favorable response to treatment with prokinetic agent than patients with a normal EGG. [276], [277] Multichannel EGG recording using electrodes placed at several positions overlying the stomach has been suggested to assess slow wave propagation and coupling. Patients with functional dyspepsia were found to have inconsistencies in frequency and regularity of gastric slow waves suggesting impaired slow wave propagation and coupling. [278]

Visceral Hypersensitivity Augmentation of visceral afferent sensation (nociception) may be a major cause of symptoms in functional dyspepsia. Thirty five to 50% of the patients with functional dyspepsia have increased sensation to gastric and small intestinal distention. [5], [279] When the stomach is stimulated by increasing volumes or increasing intragastric pressures, patients with functional dyspepsia experience symptoms at volumes that physiologically normal subjects do not (allodynia), and they have greater amount of pain at levels when the physiologically normal subjects begin to have pain (hyperalgesia). The symptoms in functional dyspepsia may result from altered gastric perception with an exaggerated visceral sensory perception of normal physiological events. In addition to hypersensitivity to distention, some dyspeptic patients may be hypersensitive to intragastric or intraduodenal acid, bile salts, or even nutrients. [280] In other functional GI disorders, such as irritable bowel syndrome, different CNS regions are activated to process visceral pain when compared with physiologically normal subjects. Brain activation by visceral stimulation with gastric distention is being studied in functional dyspepsia using positron emission tomography and functional magnetic resonance imaging. In physiologically normal subjects, distal gastric distention produces symptoms of abdominal pain, nausea, and bloating, and it activates some of the same cortical and subcortical CNS regions implicated in processing somatic pain. [281] Thus, there are several pathophysiological alterations of gastric motility and sensation in functional dyspepsia. Distinct pathophysiological abnormalities may be present in subgroups of patients, perhaps causing different symptoms, and may predict different responses to treatment. One study found that 23% of patients with functional dyspepsia had delayed gastric emptying with associated symptoms of nausea, vomiting, and postprandial fullness, 35% of patients had hypersensitivity to gastric distention associated with symptoms of pain, belching, and weight loss, and 40% had impaired accommodation associated with symptoms of early satiety and weight loss. [261] Twenty three percent of patients had two or three of these abnormalities, whereas 26% had none, suggesting still another as yet unidentified mechanism of functional dyspepsia. Unfortunately, individual symptoms do not strongly predict underlying pathophysiology. [261] These disorders may be interrelated: reduced fundic accommodation may lead to increased wall tone in response to distention or meal ingestion and may lead to stimulation of visceral afferents and cerebral perception of gastric events/symptoms. [271]

Treatment: Focusing on Gastric Dysmotility and Nociception

There is much literature, although inconsistent, on treatment of functional dyspepsia. Methodological shortcomings are present in many studies including suboptimal study design, unclear inclusion criteria, unvalidated outcome measures, and a short duration of follow-up. [282] In addition, the placebo response in functional dyspepsia can be substantial, approximating 45%. [283] Treatment modalities that have been tested extensively include acid suppressive agents, promotility compounds, and *H pylori* eradication.

Promotility Agents The lack of available promotility compounds limits this treatment modality. Promotility agents including metoclopramide, cisapride, and domperidone have improved dyspeptic symptoms more effectively than placebo in the majority of studies [282], [284]; improvement in symptoms with promotility agents has been 40% to 45% greater than with placebo. [284] The few studies that compare the effects of acid suppressing and promotility agents on dyspeptic symptoms have favored the promotility agents. [285] Several metaanalyses of the prokinetic agent studies in functional dyspepsia have been published. [284], [286], [287] In a metaanalysis of 17 cisapride studies and 4 domperidone studies, both cisapride and domperidone seemed to be efficacious. [287] For cisapride, there were overall reductions in epigastric pain, early satiety, abdominal distention, and nausea. Another metaanalysis found a funnel plot of study results to suggest that publication bias may be skewing the data on the efficacy of prokinetic agents. [288] Erythromycin and other motilin receptor agonists are potent stimulants of gastric motor function. They have not yet demonstrated efficacy in functional dyspepsia. The motilin receptor agonist, ABT-229, was ineffective in dyspeptic patients with delayed gastric emptying. [289] It is speculated that because motilin receptor agonists increase proximal gastric tone, they may aggravate the impaired fundic accommodation in functional dyspepsia, and this could explain the worsening of postprandial symptoms with ABT-229. [289]

Psychotropic Medications and Antinociceptive Agents Strategies to inhibit pain or symptom perception (sensory modulators) are receiving attention because of the augmented visceral hypersensitivity in dyspepsia. Psychotropic medications including antidepressants and serotonin reuptake inhibitors are being studied. Anxiolytic agents, such as benzodiazepines, should be avoided because of their potential for addiction. Low-dose tricyclic antidepressants appear to have analgesic effects that are independent of psychological effects. The mechanism of these agents in functional bowel disorders is not known, but it may include reduction in visceral sensitivity. [290] Tricyclic antidepressants have been beneficial to treat abdominal pain syndromes, noncardiac chest pain, and irritable bowel syndrome. [291] Two studies using low-dose tricyclic antidepressants for functional dyspepsia [292], [293] reported a decrease in dyspeptic symptoms, primarily abdominal pain. In clinical practice, psychotropic medications have been suggested for patients with severe, intractable, refractory dyspeptic symptoms that interfere with daily activities. Dosages required for symptomatic improvement are usually lower than those used to treat psychiatric disorders. [290] Starting at low doses, with gradual dose increments, is the approach used. Typically, for amitriptyline, a starting dose is 10-25 mg at bedtime. If benefit is not observed in several weeks, doses are increased by 25-mg increments up to 75 to 100 mg. The positive effects are rapidly noticed by many patients, in contrast to the prolonged treatment needed to improve depression. Significant side effects of antidepressant agents are common, can interfere with management, and may lead to a change in medication in 25% of patients. [290] The sedative effects of many tricyclic compounds may be helpful for patients with difficulty in sleeping. Agents with strong anticholinergic properties, such as amitriptyline, may adversely affect gastric motility. The secondary amines, nortriptyline (Pamelor) and desipramine (Norpramin), have fewer side effects than amitriptyline (Elavil). There are no published studies demonstrating the utility of the selective serotonin reuptake inhibitors for functional dyspepsia. Some studies are reported for irritable bowel syndrome. [294] Interestingly, paroxetine appears to enhance gastric accommodation rather than improve the visceral hypersensitivity. [295]

Other Treatments Complementary and alternative medicines are used frequently by patients with functional dyspepsia. In a prospective German study of 3001 patients with functional dyspepsia, 40% of the patients used alternative medicine regimens including special diets, herbal teas, yoga, and acupuncture. [296] In Germany, commercially available herbal preparations such as Iberogast, with extracts from bitter candy tuft, [297] and Enteroplant, containing peppermint oil and caraway oil, [298] have been shown in placebo-controlled trials to improve symptoms. Psychodynamic interpersonal psychotherapy and hypnotherapy are being reported as effective in the treatment of functional dyspepsia. [299], [300] Interestingly, one preliminary study has shown that posthypnotic relaxation with gut-oriented suggestions is effective in accelerating gastric emptying in dyspeptic patients. [301]

Future Treatment

Prokinetic agents. Future treatments being evaluated for functional dyspepsia include newer prokinetic agents. These include 5-HT $_4$ agonists (tegaserod and cilansetron), dopamine receptor antagonists (levosulpiride), CCK antagonists (loxiglumide and its dextroisomer, dexloxiglumide), and several motilin agonists. The 5-HT $_4$ agonist tegaserod has been found to accelerate gastric emptying as well as to have an effect on visceral afferent sensation. [199]

Fundic relaxing agents. Agents that relax the fundus and improve the accommodation response may be helpful in some patients with functional dyspepsia, perhaps those with early satiety. 5-HT $_1$ agonists (sumatriptan, buspirone), a-adrenergic receptor agonists (clonidine), and nitric oxide donors (glyceryl trinitrate) have been tried in several small series. [204] The 5-HT $_1$ agonists, such as sumatriptan, induce relaxation of the gastric fundus through a nitric oxide–mediated pathway. Sumatriptan allows larger volumes to be accommodated before perception or discomfort is reached, and it improves meal-induced satiety in patients with functional dyspepsia. [302] It is given primarily by injection. Buspirone, an oral 5-HT $_1$ agonist, may have effects similar to those of sumatriptan. It also has anxiolytic properties. The a-adrenergic agonist clonidine reduces proximal gastric tone and pain perception during gastric distention in healthy subjects. [204] Thus, clonidine has the potential to reduce gastric sensation or increase gastric compliance without altering other physiological motor functions. Some studies have suggested that clonidine may delay gastric emptying. [205] Sildenafil (Viagra) has been shown to increase gastric accommodation. [303] In animal models, it also relaxes the pylorus and improves gastric emptying. [304] In humans, however, it slows gastric emptying. [303] Studies suggest that 5-HT $_3$ receptor agonists relax the gastric fundus without inhibiting antral motility. [305] These agents may be useful for impaired accommodation.

Sensory modulators. Strategies to inhibit pain or symptom perception include use of ?-opioid agonists and 5-HT $_3$ receptor antagonists. Fedotozine, a ?-opioid agonist, has visceral analgesic properties, blunting perceptions of gut distention. Fedotozine has been evaluated in Europe for functional dyspepsia and has demonstrated mild symptom benefit in two trials. [306], [307] Fedotozine provided relief of epigastric pain and nausea with a trend to reduced fullness when compared with placebo. [307] It is not available in the United States; concern has been expressed on the fairly modest symptom improvement and the exact mechanism of action. [308] Fedotozine may act through peripheral opiate and cholinergic pathways to stimulate antral and small intestinal contractility. [309] 5-HT $_3$ receptors are located on vagal afferents and dorsal root ganglion neurons. In physiologically normal subjects, the 5-HT $_3$ receptor antagonist, alosetron, reduced postprandial nausea and vomiting in response to a dyspepsia-inducing meal, without an effect on gastric accommodation or the amount of ingested food. [310] Preliminary studies suggest that alosetron may be mildly effective in decreasing symptoms of functional dyspepsia. [311] Alosetron was FDA approved for treating irritable bowel syndrome. It was sanctioned because of reports of cases of ischemic colitis and is currently not available for clinical use in the United States.

Gastric antidysrhythmics. This area has been receiving attention because of the increased availability of EGG. Many of the prokinetic agents such as metoclopramide, cisapride, [9] and domperidone [8] have been shown to correct gastric dysrhythmias. Prostaglandin inhibitors have been shown to resolve tachygastrias in vitro and in vivo during hyperglycemia. [56] In several patients, indomethacin has improved symptoms and gastric myoelectric abnormalities. [312] Unfortunately, indomethacin is also ulcerogenic.

OVERVIEW

This chapter reviews gastric motility disorders and focuses primarily on the pathophysiology, diagnosis, and treatment of gastroparesis, dumping syndrome, and the gastric dysmotility seen in functional dyspepsia. Refractory symptoms may lead to a comprehensive evaluation including gastric emptying scintigraphy, antroduodenal manometry, EGG, and possibly tests of gastric accommodation or compliance. This evaluation may guide treatments that target the underlying pathophysiology.

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CHAPTER 66

John Del Valle, William D. Chey, and James M. Scheiman

ACID PEPTIC DISORDERS

Sour eruptions, gas, and epigastric pain radiating to the back with occasional splashing noises and vomiting are familiar to most physicians who have treated patients with acid peptic disorders of the stomach. In fact, the early Greek Diocles of Carystos (350 BCE) described these as symptoms of a melancholic gassy illness originating in the stomach. [1] *Acid peptic disorders or peptic ulcer disease* (PUD) represent a common group of pathological states that affect as many as 4.5 million persons per year (new cases and recurrences) in the United States. [2], [3] The cost of health care for these common disorders has been substantial, with estimated annual direct (physician visit, diagnostic studies, treatment) and indirect (time lost from work) costs of more than $9 billion per year. [4], [5] The past several decades have witnessed dramatic advances in the diagnosis and therapy of acid-related disorders. The development of histamine (H_2)–receptor antagonists and H^+,K^+-ATPase inhibitors and an understanding of how the gastroduodenal protective equilibrium is altered by such culprits as nonsteroidal antiinflammatory drugs (NSAIDs) and *Helicobacter pylori* have changed how we think of peptic disorders. This chapter provides a comprehensive overview of the epidemiology, pathogenesis, diagnosis, and therapeutic approach to patients with PUD.

IMPACT OF PEPTIC ULCER DISEASE

Approximately 500,000 new cases and 4 million recurrences of gastric and duodenal ulcers in the United States led 4 million patients to visit physicians in 1995. [2], [3] Cost-analysis data published in 1998 revealed that there were more than 2.6 million PUD-related physician office visits, resulting in a total cost of $196.3 million. [5], [6] The inpatient setting was the second most common site of care, with 470,000 PUD-related visits. The total cost (facility and physician fees) was $1.8 billion, mostly for facility costs ($1.6 billion). PUD-related visits to hospital-based outpatient departments (250,000) and emergency departments (190,000) created a total cost of $370.2 million and $66.8 million, respectively. In the year 2000, the cost of drugs (prescription and over the counter) to treat ulcer disease was approximately $620.5 million. Taken together, the total direct cost of PUD is approximately $3.3 billion per year.

The indirect costs related to PUD, that is, the per person loss of productivity over a 3-month period, is estimated at $606. [7] Because more than 75% of patients with PUD are of working age (18 to 65 years old), the estimated productivity loss is estimated at $6.2 billion.

EPIDEMIOLOGY AND NATURAL HISTORY

Two major epidemiologic observations have markedly changed how we approach PUD: the association between *H pylori* and gastric duodenal mucosal ulceration, which leads one to consider peptic ulcer as an infectious disease; and the relationship between ingestion of NSAIDs and the production of mucosal damage, which has generated an acute awareness of preventive measures.

Incidence, Prevalence, Risk Factors, and Odds Ratios

Epidemiologic studies regarding acid peptic disorders are often derived from large databases in which the actual diagnostic criteria for establishing an ulcer are not available to the investigators. Despite this, the qualitative trends are valid because similar observations have been made by several investigative groups. [4], [8], [9] Ulcer complications, such as perforation and hemorrhage, usually require hospitalization and therefore can be assessed more accurately. The 1-year point prevalence of active gastric and duodenal ulcer in the United States is approximately 1.8%, [10] whereas lifetime prevalence ranges from approximately 11% to 14% for men and 8% to 11% for women. [11], [12], [13] and [14] In young Norwegians, the annual incidence of duodenal ulcer was approximately 2 in 1000 men and 0.9 in 1000 women, with similar annual rates for gastric ulcer in both sexes. [15] In Japan, the male-to-female ratio for peptic ulcer was 2:1, but the overall gastric ulcer rate was about 1.5 times greater than that for duodenal ulcer. [16] In 1998, it was estimated that there were 6.8 million cases of PUD in the United States, representing a prevalence rate of 2490 cases per 100,000 persons. [6] The male-to-female ratio for deaths from duodenal ulcer has decreased from 5:1 to about 1.3:1.2, in part because of a decrease in the rate of duodenal ulcer among men, with little change among women. It will be interesting to observe the impact of *H pylori* eradication on these statistics.

Temporal, Gender, and Mortality Trends

Before the 20th century, duodenal ulcer disease was distinctly unusual. [17] The major form of ulcer disease in the 1800s was gastric ulcer, particularly in women, and it was often associated with perforation. Duodenal ulcer disease increased progressively during the early 1900s, reaching a peak in about 1950. [18] Susser and Stein [19] introduced the key concept of birth cohorts (i.e., those born during the same time period) and attributed the changes in ulcer incidence to the urbanization of United States society. These investigators proposed that factors introduced to the birth cohort born between 1868 and 1908 predisposed this group to develop duodenal ulcers and those born about 10 years earlier to develop gastric ulcers. Those born before and after these dates had a lower lifetime prevalence of ulcer disease. This finding suggests that some environmental factor (e.g., *H pylori*) was introduced during the latter part of the 19th century and the early 20th century that increased the incidence of ulcers, and the risk factors were introduced early in life and decreased thereafter. [20] The birth cohort phenomenon applies to data collected in the United States, Britain, Europe, and Japan. [9]

The number of physician office visits for duodenal ulcer decreased between 1958 and 1993. [3] In 1998, there were almost 450,000 outpatient visit (office and emergency department) for PUD-related issues. Hospitalizations for duodenal and gastric ulcer disease decreased in the United States from 1965 through 1980, but there has been little change in the hospitalization rates for perforated duodenal ulcer and for gastrointestinal (GI) hemorrhage. [10] Similar statistics have been reported from Europe. [21]

The mortality rate for duodenal ulcer in 1962 was 3.1 per 100,000, and for gastric ulcer, it was 3.5 per 100,000; these rates had decreased to about 1 per 100,000 each by 1979. [22], [23] Data from the United States Centers for Disease Control and Prevention in 1998 estimate a crude mortality rate of ulcer disease of 1.74 deaths per 100,00 persons. [6] Gastric ulcer has a higher mortality rate than duodenal ulcer because of its prevalence in older patients. [24], [25] and [26]

Surveys in the United States and Europe have documented a substantial increase in hospitalizations of elderly patients for bleeding and perforation. [27], [28] Although the overall mortality rate remained stable throughout the 1970s, it rose significantly in patients older than 75 years of age, presumedly because of increased consumption of NSAIDs by an expanding geriatric population. In Australia, a twofold increase in the intake of nonaspirin NSAIDs from 1979 to 1988 by those older than 65 years of age was associated with a striking increase in peptic ulcer hospitalization rates. [29], [30] and [31] The availability of H_2-receptor antagonists, H^+,K^+-ATPase inhibitors, and prostaglandin analogs has not decreased the rate of ulcer complications.

Genetics of Ulcer Disease

Earlier studies that suggested a potential genetic predisposition to ulcer development [32] have been reconsidered in light of the role of *H pylori* in the pathogenesis of peptic disease. The concordance for peptic ulcer among identical (i.e., monozygotic) twins is approximately 50% and is increased among nonidentical twins, although not to the same degree. [33] The lifetime prevalence of developing an ulcer in first-degree relatives of patients with ulcers is about threefold greater than that in the general population. [34] Approximately 20% to 50% of patients with duodenal ulcer report a positive family history, contrasted with only about 10% of patients who do not have ulcers. [33], [35] Gastric ulcer also clusters in families, but the prevalence of gastric ulcer is not increased among families with duodenal ulcers or vice versa.

The increased incidence of ulcer disease in first-degree relatives is likely from clustering of *H pylori* within families, rather than secondary to genetic factors. Similarly, the theory that elevated levels of serum pepsinogen I may be a genetic marker for familial duodenal ulcer was explained by infection with *H pylori*. [36] In addition, reported genetic markers for PUD, such as blood group O antigen (1.3-fold increase is induced), [37] inheritance of nonsecreting blood group status, [38] and human leukocyte (HLA) subtypes (HLA-B5, HLA-B12, HLA-Bw35), were challenged. The lifetime prevalence of PUD was found to be equally high in persons with O and A phenotypes, but the Lewis phenotype Le (a-b-) and the ABH nonsecretor trait are relevant markers of peptic ulcer. [39] It is thought that these traits may confer an increased susceptibility to infection with *H pylori.* Rare genetic syndromes associated with duodenal ulcer include multiple endocrine neoplasia type I (MEN I) and systemic mastocytosis. [35]

PATHOPHYSIOLOGY

The development of gastroduodenal ulcerations is thought to result from an imbalance between endogenous noxious agents, including hydrochloric acid, proteolytic enzymes, and bile (aggressive factors), and the mucosal defense and repair mechanism that keep the surface intact. Geometric growth in basic science knowledge has facilitated our understanding of the aggressive and defensive factors involved in this equation.

Gastric Secretion

Two secretory products with the potential of producing mucosal damage include hydrochloric acid and pepsinogen. The mechanisms regulating secretion of acid and pepsinogen are reviewed in detail in Chapter 13.

Basal acid secretion follows a circadian pattern, with the lowest secretion in the morning and the greatest at night. [40] Cholinergic input through the vagus nerve and histaminergic input from locally released histamine are the main determinants of basal gastric acid output; food is the principal physiological stimulant of increased acid production. Meal-stimulated acid secretion occurs in three phases referable to origin of the stimulant: the cephalic, gastric, and intestinal phases. The principal determinants of the cephalic phase include sight, smell, and taste of food, which operate through cholinergic input through the vagus nerve. Once food enters the stomach, the gastric phase is activated by amino acids and amines that directly stimulate gastrin release, which, in turn, increases the secretory response. As nutrients enter the intestine, the principal stimulatory factors include distention and proteins and their breakdown products. Series of inhibitory pathways are activated during the different phases of gastric secretion and serve as a counterbalance for the secretory process. The GI hormone somatostatin seems to be an important element in this inhibitory process.

The parietal cell is found in the oxyntic gland in close proximity to the histamine-containing enterochromaffin-like (ECL) cell. The ECL cells express receptors for gastrin and acetylcholine, both of which stimulate histamine release. [41], [42]

The acid-secreting parietal cell contains receptors for histamine (H_2 class), gastrin, and acetylcholine (M_3) (Fig. 66-1). [43], [44] Histamine stimulates adenylate cyclase, whereas acetylcholine and gastrin activates the phosphoinositide–intracellular calcium cascade. The increase in intracellular cyclic adenosine monophosphate (cAMP) or cytosolic calcium leads to activation of various protein kinases, which, in turn, regulate the acid-secreting pump on the canalicular surface of the parietal cell: H^+,K^+-ATPase. The combination of these different signaling pathways leads to a significant increment in gastric secretion (*potentiation*). Potentiation explains why receptor blockade of one pathway with an antagonist also diminishes acid secretion derived from an alternate pathway. [45]

FIGURE 66-1. Gastric parietal cell in the resting and stimulated state. Parietal cells express receptors for gastrin, acetylcholine (*ACh*), and histamine (H_2 subtype). Ligand-mediated parietal cell stimulation leads to a marked increase in canalicular membranes containing H^+,K^+-ATPase. (Adapted from ref. [46].)

H^+,K^+-ATPase is a membrane-embedded protein consisting of two subunits: an a subunit (the active catalytic site) and a ß subunit, whose function is not fully elucidated. This enzyme uses the chemical energy of ATP to transport (pump) protons from the cytoplasm of the parietal cell into the secretory canaliculi in exchange for K^+. The pumps are located in the cytoplasmic membrane of the secretory canaliculus and in cytoplasmic tubulovesicles. [46], [47] They depend on the degree of parietal cell stimulation. The cytoplasmic vesicles are impermeable to K^+, thus rendering the proton pump inactive. In the resting state, only 5% of pumps are active and are located within the canaliculi. During stimulation, cytoplasmic tubulovesicles are rapidly transferred to the membrane of the secretory canaliculus, where 60% to 70% are activated. After stimulation of the parietal cell ceases, the proton pumps return to their inactive state in the cytoplasmic vesicles. [46], [47] The half-life of the proton pumps is 48 hours. [48]

Pepsinogen, the inactive precursor of the proteolytic enzyme pepsin, is produced by the chief cell, which is found primarily in the gastric fundus. The pathogenetic importance of derangements in pepsinogen secretion in PUD remains to be established (see Chapter 13).

Mucosal Defense and Repair

Defensive elements that prevent the gastroduodenal mucosa from damage can be divided into preepithelial, epithelial, and subepithelial factors (Table 66-1). This division is arbitrary, because the entire mucosa functions in concert to prevent injury. The term *cytoprotection* was coined by Andre Robert and colleagues [49], [50] to describe the ability of prostaglandins, independent of their ability to inhibit acid secretion, to protect rat gastric mucosa against a variety of noxious agents. They also observed that low doses of mild irritants released endogenous prostaglandins that could reduce or prevent deep epithelial injury produced by higher concentrations of these or other agents, [50] a phenomenon termed *adaptive cytoprotection.* The term cytoprotection itself is a misnomer, because histological studies of the cytoprotection model demonstrate superficial epithelial injury with preservation of deep mucosal integrity. [51] However, the cytoprotection phenomenon prompted an explosion of investigation into the role of endogenous prostaglandins, nitric oxide (NO), mucus and bicarbonate (HCO_3^-) secretion, maintenance of mucosal blood flow, stabilization of mucosal mast cells and lysosomes, and effects on gastric motility and cellular proliferation. No single mechanism entirely explains the cytoprotection phenomenon. [52]

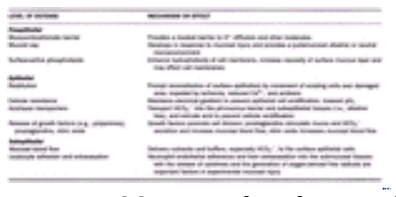

TABLE 66-1 Mechanisms of Gastroduodenal Mucosal Defense

Cyclooxygenase Isoenzymes The depletion of endogenous prostaglandins appears to play an essential role in the development of mucosal injury resulting from NSAIDs. Cyclooxygenase (COX) is the rate-limiting catalytic enzyme in the production of endogenous prostaglandins from membrane-esterified arachidonic acid released under physiological stimulation. Two isoforms of the enzyme have been identified (Fig. 66-2): COX-1, which is constitutively expressed and plays an important role in the maintenance of normal gastric (as well as other organ) function; and COX-2, which is an inducible form that is up-regulated in areas of inflammation. [53] COX-2 is also constitutively expressed in a variety of organ systems, including the brain, kidney, and reproductive tract. The normal stomach has very low levels of COX-2 expression, predominantly in parietal cells, endothelial cells, and macrophages. However, in the setting of *H pylori* gastritis, COX-2 is induced in the epithelial cells, and the degree of induction correlates with the extent of inflammation. [54] NSAIDs nonspecifically inhibit both COX isoforms, leading to both beneficial (antipyretic and antiinflammatory) and toxic (GI injury) effects. Differences in the selectivity of various NSAIDs for COX-2 versus COX 1 (Fig. 66-3) may partially explain differences in ulcer and complication risks associated with specific NSAIDs. [55] The reduced ulceration demonstrated in clinical trials with the COX-2–specific inhibitors, agents that provide similar efficacy to NSAIDs without affecting gastric prostaglandins, have further emphasized the critical role of COX-1 in gastric mucosal defense and repair. [56]

FIGURE 66-2. Tissue distribution and biologic actions of the cyclooxygenase (*COX*) isoenzymes. COX-1 is constitutively expressed in platelets, endothelium, stomach, and kidney, whereas COX-2 is inducible in sites of inflammation in macrophages, leukocytes, fibroblasts, and endothelial cells. COX-2 is also constitutively expressed in vascular endothelial cells and in the kidney, ovary, and brain. Nonsteroidal antiinflammatory drugs nonselectively inhibit both COX isoforms, whereas COX inhibitors inhibit COX-2 only.

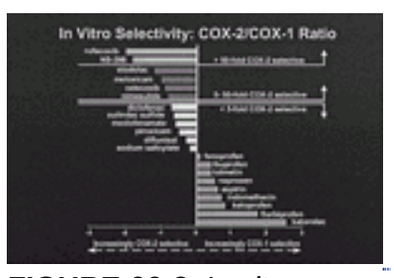

FIGURE 66-3. *In vitro* comparison of selectivity of various nonsteroidal antiinflammatory drugs and cyclooxygenase-2 (COX-2)–specific inhibitors. Drugs with selectivities toward the positive direction (**right**) are more COX-1 selective, and those toward the negative direction (**left**) are more COX-2 selective. These ratios vary depending on assay conditions, but the pattern is generally preserved. (Data from ref. [55].)

Preepithelial Defensive Factors The stomach and duodenum are covered by a mucous- HCO_3^- barrier that provides primary defense against the strongly acidic gastric lumenal contents (Fig. 66-4). The surface epithelial cells secrete mucus and form a gastric layer of mucus 100 µm thick. Gastric acid is secreted in a pulsatile manner through the mucous gel, allowing the formation of short-lived channels within the mucous gel that rapidly close to prevent lumenal acid back-diffusion. This maintains the pH at the mucosa near neutrality despite the low lumenal pH. [57] Gastroduodenal mucus is a viscous glycoprotein that serves as a modest barrier for H^+ diffusion (about fourfold compared with water), although it is an effective barrier to the diffusion of pepsin and other large molecules. Histamine, acetylcholine, and gastrin coordinately stimulate mucin production, whereas prostaglandin appears to increase mucin release by epithelial cells. [58] Dipalmitoylphosphatidylcholine, the predominant surface-active phospholipid found in pulmonary surfactant, is secreted to high levels by the gastric mucous cell. [59] Gastric surfactant, a complex of mucin glycoprotein with these surface-active phospholipids, appears to play an important physiological role in mucosal defense by forming a hydrophobic surface with nonpolar fatty acids that extend from the cell membrane into the lumen, increasing the viscosity of the surface mucous layer. [60] Topical administration of phospholipids to the gastric mucosa can prevent NSAID-induced mucosal damage in animals; efficacy in humans is under investigation. [61]

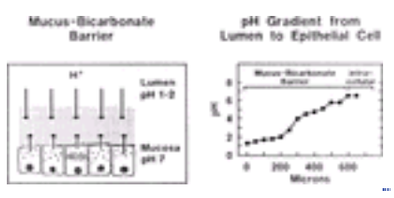

FIGURE 66-4. A layer of mucus overlies the gastroduodenal mucosa. This provides a barrier against the diffusion of acid (H^+) from the lumen to the mucosa. The surface epithelial cells secrete bicarbonate into the base of this barrier. The **right panel** depicts the pH gradient as measured by a pH microelectrode passed from the stomach gradually into the mucous-bicarbonate barrier. The lumenal pH is about 1.3, and the pH immediately adjacent to the surface cell is near 7.0. (Adapted from Flemstrom G, Kivilaakso E. Demonstration of a pH gradient at the luminal surface of rat duodenum in vivo and its dependence on mucosal alkaline secretion. Gastroenterology 1983;84:787.)

HCO_3^- secretion, the other preepithelial defensive factor, is secreted by the surface epithelial cells into the mucous gel to provide a protective pH gradient that maintains a neutral pH at the epithelial cell surface. Lumenal acidification and prostaglandins are potent stimulants of mucosal HCO_3^- secretion. [62] Because

epithelial HCO_3^- secretion requires the presence of HCO_3^- at the nutrient side of the basolateral membrane, states of acidosis, such as shock on decreased gastroduodenal blood flow, lead to marked suppression of surface epithelial HCO_3^- secretion.[63] This phenomenon is likely an important factor in stress-induced gastroduodenal mucosal injury.

Epithelial Defensive Factors The properties of the surface epithelial cells that provide the second line of gastroduodenal defensive factors include restitution, epithelial cell metabolism (e.g., transmembrane, transcellular resistances), acid-base transporters that maintain intracellular pH, and mucus secretion. The apical barrier of the gastric mucosal surface is maintained by tight junctional complexes. When superficial mucosal damage occurs, the surface epithelium rapidly repairs epithelial continuity through the migration of existing epithelial cells from the mucous neck cell area over the basal lamina. This process, known as *mucosal restitution,* can repair small epithelial defects.[64] Restitution occurs rapidly without cell division, depends on microfilaments, and is blocked in the presence of mucosal ischemia or low pH.[64],[65] Damage to the gastroduodenal mucosa results in a layer of gelatinous mucus, fibrin, and cellular debris, referred to as the *mucoid cap,*[66] that forms an alkaline microenvironment conducive to restitution and healing. When this first-line barrier of HCO_3^- and mucus is dissipated, however, the surface epithelial cells remain capable of maintaining intracellular pH, provided there is adequate HCO_3^- supplied through circulation and the Na^+/H^+ and Cl^-/HCO_3^- exchangers and a Na^+/HCO_3^- cotransporter on the cell membranes.[67],[68] When epithelial cells become acidified because of a failure of the overlying pH-mucous barrier, these mechanisms provide the capacity to extrude acid and maintain normal intracellular pH. The epithelial growth factor (EGF) family and trefoil peptides appear to play important roles in gastric physiology.[69] The two most important members are EGF and transforming growth factor-a (TGF-a). EGF is produced in saliva, by Brunner glands in the duodenum, by cells in chronically inflamed mucosa.[70] TGF-a is produced by gastric parietal cells, and it also binds to the EGF receptor. Activation of the EGF receptor leads to inhibition of gastric acid secretion, stimulation of mucin production, and epithelial cell migration and proliferation. Trefoil peptides, so named because of their unique three-leaf structure, are up-regulated at sites of GI injury and include human spasmolytic peptide, pS2, and intestinal trefoil factor.[71] Both human spasmolytic peptide and intestinal trefoil factor protect against experimental injury resulting from ethanol and indomethacin.[72],[73] The colocalization of trefoil factors with mucous cells and their close association with gastric mucin have led to the hypothesis that these peptides enhance the barrier function of mucus. *Regeneration,* the process by which larger epithelial defects that destroy the basal lamina (e.g., ulcers) heal, requires cellular proliferation. Regeneration is at least partly dependent on tissue prostaglandins and growth factors, including the EGF family,[69],[74] and by angiogenic factors such as fibroblast growth factor[75] and hepatocyte growth factor.[76] The regenerative and angiogenic ulcer healing response appears regulated, at least in part, by prostaglandins derived from COX-2.[77] This supports the clinical observation that NSAIDs may impair ulcer healing by their effects on both isoforms of COX, a concern that applies as well to the COX-2–specific inhibitors. The adverse impact on healing can be overcome when gastric acid is markedly reduced, such as with a proton pump inhibitor (PPI).

Subepithelial Defensive Factors The key subepithelial process that prevents mucosal injury is adequate mucosal blood flow. Diminished blood flow or acidosis markedly impairs preepithelial and epithelial defensive factors. The rich vascular anatomy within gastric mucosa is designed to carry the HCO_3^- released by actively secreting parietal cells at the base of the gland to the epithelial surface, where it can neutralize protons that breach the earlier lines of defense.[78] For each molecule of H^+ secreted by the parietal cells, one molecule of HCO_3^- is transported across the basolateral membrane, producing this *alkaline tide* in the submucosa. When gastric acid secretion is suppressed, by H_2-receptor antagonists or H^+,K^+-ATPase inhibitors, acidification of the gastric mucosa results in greater injury than in the acid-secreting mucosa because of the absence of this alkaline tide. Maintenance of endothelial integrity appears to define the cytoprotection phenomenon and, as discussed later, is a critical component of NSAID-induced GI injury.

Pathogenesis of Peptic Ulcer Disease

Pathophysiology of Duodenal Ulcers Before the role of *H pylori* was understood, many studies were published proposing a unifying, acid-driven hypothesis as fundamental to the genesis of peptic ulceration. Unfortunately, no single abnormality in the gastric secretory process has been found in all patients with ulcers. Despite this, acid is still an independent factor that negatively affects mucosal defense. The next section outlines some of the earlier work on the role of gastric secretion, motility, and mucosal abnormalities as primary pathogenic elements in ulcer development. This is followed by a review of *H pylori*–induced and NSAID-induced disease.

Acid dysregulation. The pathophysiology of duodenal ulcer disease has been examined in selected and often relatively small groups of patients and controls. Discrepancies exist among studies because of the heterogeneity of duodenal ulcer disease itself and the heterogeneity of study populations, especially the control groups. Despite these considerations, several abnormalities in gastric and duodenal secretion, gastrin release, and gastric emptying have been observed in patients with duodenal ulcer (Fig. 66-5).[79],[80] and[81] To what extent these abnormalities are associated with *H pylori* infection, which would have been present in almost 100% of patients with ulcers, but only in some control subjects, is unknown.

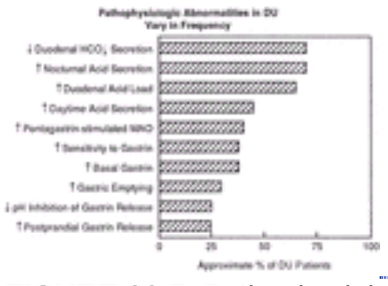

FIGURE 66-5. Pathophysiological abnormalities in patients with duodenal ulcer vary in frequency. Shown are the approximate percentages of patients with duodenal ulcers with some of the abnormalities that have been described. These data come from many separate studies with different experimental conditions and patient populations, and they represent approximations of these abnormalities. For example, approximately 70% of patients with duodenal ulcers have decreased proximal duodenal mucosal bicarbonate or increased nocturnal acid secretion.

Duodenal ulcer and duodenal pH. Attempts at establishing a role for altered duodenal pH as an important factor in ulcer formation has been fraught with technical difficulties; therefore, it is not surprising that the results of studies examining these parameters in patients with PUD are conflicting.[82],[83],[84] and[85]

Abnormalities in duodenal mucosal defense. Some patients with gastric or duodenal ulcer appear to have a mucous gel covering the mucosa that is somewhat structurally weaker than that of normal persons.[86] The mechanism for this is unknown, and these findings have not been confirmed. HCO_3^- production in the duodenal bulb, but not the distal duodenum, is markedly diminished in patients with active duodenal ulcer as compared with healthy persons (Fig. 66-6).[87] The mechanism of this defect is not established, but a cellular or subcellular defect in HCO_3^- secretion has been proposed.[87] Histological evaluation of the duodenum in patients with inactive duodenal ulcer fails to reveal any striking morphologic changes when compared with the mucosa of healthy persons.[88] Studies suggests that *H pylori* may alter duodenal mucosal HCO_3^- production.

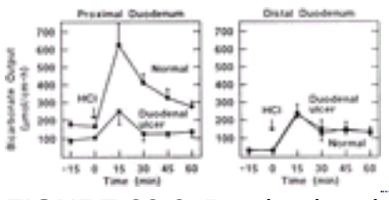

FIGURE 66-6. Proximal and distal duodenal mucosal bicarbonate secretion in patients with duodenal ulcers and in healthy persons. The proximal duodenal mucosal bicarbonate secretion is significantly impaired in patients with duodenal ulcers compared with healthy persons, and distal duodenal mucosal bicarbonate production is similar in both. Notice the gradient from the proximal to distal duodenum in the healthy persons. (Adapted from ref.[87].)

Pathophysiology of Gastric Ulcers Most (70%) of gastric ulcers are associated with NSAID use or *H pylori* infection. In an effort to develop a unified descriptive system, gastric ulcers have been classified into three groups. *Type I ulcers* occur in the body of the stomach and are not associated with other gastroduodenal disease. *Type II ulcers* are also located in the body of the stomach and are associated with a duodenal ulcer scar or active ulcer. *Type III ulcers* are located in the immediate prepyloric area. Acid secretion in types II and III is similar to that of duodenal ulcer, but ulcers located in the body of the stomach (type I) tend to be associated with normal or low levels of gastric acid secretion. Information about the *H pylori* status in these different types of gastric ulcer disease is not available. Widespread superficial gastritis often surrounds the ulcer and is present in the gastric antrum (i.e., chronic active gastritis or atrophic gastritis)[89] (see Chapter 68).

Motility abnormalities and duodenogastric reflux. Abnormalities in pyloric sphincter pressure and gastric motility have been reported in patients with gastric ulcers.

[90], [91] and [92] It seems unlikely that these effects are involved in ulcer pathogenesis because patients with gastroenterostomies and copious amounts of duodenogastric reflux often have superficial gastritis, particularly surrounding the anastomosis, but do not commonly develop gastric ulcers. Moreover, gastric ulcer healing is not enhanced by the bile salt–binding agent cholestyramine. [93]

Gastric secretion and gastric ulcers. Basal and stimulated gastric acid secretion is within normal limits in groups of patients with gastric ulcers. Unlike patients with duodenal ulcer, in whom there is a maximal acid output threshold of about 10 mmol/h for ulcers to develop, there is no minimal secretory response before gastric ulceration occurs, and gastric ulcers have been documented in the presence of achlorhydria. [94]

Mucosal blood flow. Gastric ulcers tend to occur near the gastric angulus, an area of prominent muscle bundles where the mucosa is directly supplied by the left gastric artery, rather than through a rich submucosal plexus, as observed throughout other portions of the stomach. [95]

Helicobacter pylori and Ulcer Disease

Background and history. Establishing the link between *H pylori* and the pathogenesis of PUD was one of the most exciting medical discoveries of the 20th century. We now know that *H pylori* causes most cases of histological gastritis, is the most important etiologic factor in most patients with PUD, has also been intimately linked to the pathogenesis of both low- and high-grade gastric mucosa-associated lymphoma (maltoma), and is a predisposing factor for the development of gastric adenocarcinoma. [96] Although some investigators have suggested that *H pylori* may be associated with nonulcer dyspepsia, eradication of this infection has been found to offer little, if any, benefit over placebo or a short course of PPI therapy [97], [98] (see Chapter 30 and Chapter 68). Although the recognition of *H pylori* colonization of the stomach has ushered in a new era in gastric microbiology, gastric urease activity [99] and spiral organisms have been observed in human stomachs since the turn of the 20th century. [100] Despite prior negative studies in 1975, [101] Steer and Colin-Jones [102] reported the presence of gram-negative bacteria in the gastric mucosa of approximately 80% of patients with gastric ulcers. In 1983, Warren and Marshall [103] published their landmark study characterizing the spiral-shaped *H pylori* bacterium and describing its correlation with histological gastritis. *H pylori* is a curved or S-shaped gram-negative rod approximately 0.5 by 3 µm in size, containing four to seven sheathed flagella at one pole. [104], [105], [106] and [107] Prolonged culture leads to coccoid forms of *H pylori,* which may represent a dormant state, allowing the survival of the bacterium in unfavorable environments. [108] Perhaps the most distinctive characteristic of *H pylori* is the ability to produce large amounts of highly active urease. [109]

Transmission. The natural habitat of *H pylori* is the human stomach, and perhaps the stomach of nonhuman primates, cats, and houseflies. [110], [111] and [112] It is probable that animals are secondary hosts and humans are the primary reservoir for the bacteria. Aside from colonization of gastric metaplasia or ectopic gastric mucosa in other parts of the GI tract, the organism is confined to the stomach. There are isolated reports of recovering the organism from the oral cavity, [113] but whether this represents true colonization or contamination by refluxed gastric contents remains unclear. Person-to-person transmission from a single environmental source is suggested by the increased prevalence among family members of patients with *H pylori* and among institutionalized patients. Moreover, DNA analysis reveals identical *H pylori* strains in infected members of the same family and in patients in a chronic care facility. [114], [115], [116], [117], [118], [119] and [120] *H pylori* can potentially be transmitted by three different modes. First, circumstantial evidence is most consistent with fecal-oral transmission. The organism has been isolated from feces, [121], [122] although several laboratories have not been able to duplicate these findings. Water has been suggested as a source, and, although polymerase chain reaction (PCR) assays have been positive, intact organisms have not been isolated from water. The age-related rise of *H pylori* infection, which follows a pattern similar to that of other diseases that are transmitted by the fecal-oral route, provides support for this mechanism of transmission. However, most believe that the age-related increase in *H pylori* prevalence is the result of the cohort effect (see section "Epidemiology" later). Oral-oral transmission of *H pylori* has also been postulated, based on isolation of *H pylori* from the oral cavity. This may be especially important among African infants whose mothers feed them premasticated food. [123] Moreover, it has been hypothesized that children who regurgitate or vomit infected gastric juice, especially during acute infection with *H pylori,* may transmit the infection to their contacts. [124] Finally, iatrogenic transmission has also been described in patients who have undergone acid secretory or endoscopic studies with a contaminated pH electrode, biopsy forceps, or endoscope. [125], [126] This is a rare occurrence.

Epidemiology. Gastritis, a common histological finding in the otherwise healthy population, closely parallels the presence of *H pylori* in a large percentage of persons in developing countries, with a high prevalence among children and young adults, indicating infection early in life. Indeed, in developing countries of Central and South America, Asia, and Africa, 80% of children are infected by 10 years of age. [127], [128] In contrast, the prevalence of *H pylori* and gastritis in the white population of the United States and other developed countries increases with age from approximately 10% at 20 years of age to about 50% at 60 years of age. [129], [130] These findings probably represent a cohort effect, in which most of the higher prevalence in older patients is the result of a high rate of infection earlier in life. Differences in hygiene and living conditions in the past may account for the higher rates of infection in older persons in the United States. [131] If this hypothesis proves true, the age-related *H pylori* prevalence in the United States will gradually decrease as younger persons with their lower *H pylori* prevalence advance in age. The prevalence of *H pylori* is influenced by environmental factors such as childhood socioeconomic status, domestic crowding, and the absence of a fixed hot-water supply during childhood. [131] Ethnicity may be risk factor, which may reflect childhood environmental factors or genetic susceptibility. In the United States, asymptomatic Latin Americans have approximately an 80% prevalence of *H pylori,* without significant variation in different age groups. [132] Asymptomatic African Americans have a rate of *H pylori* acquisition that parallels that of whites. However, the age-related prevalence is higher in African Americans than in whites. [130] More cogent evidence supporting genetic susceptibility comes from studies of monozygotic twins, in whom infection rates are higher than in matched dizygotic twins. [133]

Evidence that Helicobacter pylori causes peptic ulcer disease. There is no doubt regarding the central role of *H pylori* in both gastric and duodenal ulcers not otherwise associated with NSAIDs or acid hypersecretory states. Thus, nearly all non–NSAID-related ulcers should be considered manifestations of an infectious disease. This paradigm, however, needs to be understood within a changing milieu of the prevalence of *H pylori* infection. As the prevalence of *H pylori* infection declines to zero, the prevalence of ulcers will also decline, but an increasing percentage of ulcers that do occur will be unrelated to *H pylori.* [134] Three primary lines of evidence support the etiologic role of *H pylori* in PUD:

1. The natural history of *H pylori* infection. Follow-up of patients with gastritis (and therefore presumptively with *H pylori* infection) for up to 10 years revealed that 11% developed peptic ulcers (1% per year) compared with fewer than 1% of persons without gastritis. [135] The odds ratio for the association between peptic ulcer and *H pylori* infection was 3.4 (95% confidence interval = 1.8–6.3).
2. The consistent, age-independent worldwide association of *H pylori* and ulcer disease: approximately 90% of patients with duodenal ulcer and 70% to 90% of patients with gastric ulcers are infected with *H pylori.* [129], [136], [137]
3. The results of multiple prospective treatment trials indicate that eradication of *H pylori* significantly reduces the likelihood of gastric and duodenal ulcer recurrence. [138], [139], [140] and [141]

This is the best evidence supporting the concept that *H pylori* is an important contributing factor in PUD. Moreover, treatment of *H pylori* infection leads to more rapid and reliable ulcer healing than does treatment with antisecretory therapy alone. [141], [142] The beneficial effect of curing *H pylori* infection also extends to patients who present with bleeding ulcers; the rebleeding rate is significantly lower compared with patients in whom infection persists. [143] Data from the United States, however, suggest that the proportion of *H pylori*–negative ulcers is increasing. A multicenter treatment trial in the United States of patients with duodenal ulcer suggests that no more than 80% of patients had *H pylori* infection. [144] Moreover, despite cure of *H pylori* infection, the pooled recurrence rate of duodenal ulcers at 6 months after therapy in seven rigorously designed multicenter trials in the United States was 20%. [145] Similar results have been reported from Hong Kong. [146] Complicating matters further, in some populations with a very high prevalence (>80%) of *H pylori* infection; the occurrence of PUD may be mistakenly attributed to this infection. In addition, the prevalence of *H pylori* infection in complicated ulcers may be lower than that found in uncomplicated ulcer disease. Two centers found that approximately 70% of patients with bleeding duodenal ulcers were infected with *H pylori,* whereas another center observed that 50% of patients with perforated duodenal ulcers had *H pylori* infection. [146], [147], [148] and [149] In these reports, the prevalence of NSAID use could not be incriminated as an explanation for the low prevalence of *H pylori* infection. The swinging of the pendulum, from skepticism regarding the concept that PUD is an infectious disease to universal agreement regarding the importance of *H pylori* infection to a more circumspect qualified endorsement of a central etiologic role of *H pylori,* should not diminish the clinician's enthusiasm for diagnosing and treating *H pylori* infection in the setting of PUD. Nevertheless, the foregoing data lead to some uncertainty regarding the best management for patients with PUD. For patients (e.g., the elderly, those with severe comorbidity) in whom recurrence of an ulcer, especially a bleeding ulcer, may be associated with severe morbidity, full-dose antisecretory therapy may be appropriate, despite cure of *H pylori* infection. In most cases, maintenance antisecretory therapy is unnecessary in view of the low recurrence rate and morbidity of PUD after *H pylori* eradication.

Helicobacter pylori and nonsteroidal antiinflammatory drug interactions. Available data can be interpreted to support three potential interactions between NSAIDs and *H pylori*: no influence, increased adverse outcome, and beneficial effects. Most of the data suggest that NSAIDs and *H pylori* are independent risk factors for PUD. Circumstantial evidence favors the concept that NSAIDs increase the risk of developing complicated or otherwise symptomatic ulcers in patients with a prior history of ulcer disease. [150], [151] Although NSAIDs alone appear to cause more endoscopically detected simple ulcers in the stomach than in the duodenum, NSAID-associated ulcers occur with similar frequency in both these locations. [152], [153], [154], [155] and [156] Moreover, most NSAID-associated complications occur within 1 month after initiation of NSAID therapy. These findings suggest that NSAIDs induce complications in patients with preexistent clinically silent ulcers. More limited data suggest that in patients without a history of PUD, there is a higher incidence of peptic ulcers in association with NSAID use in patients infected with *H pylori* as compared with those without *H pylori* infection. This is based on a trial suggesting that cure of *H pylori* diminishes the incidence of peptic ulcers in patients beginning NSAIDs. [157], [158] In contrast to those studies, which focused on NSAID-naive subjects, two studies assessed the effect of *H pylori* eradication on ulcer healing and relapse in patients who were already taking long-term NSAIDs. [159] Eradication of *H pylori* did not influence the rate of ulcer healing, and there was only a trend

favoring *H pylori* eradication in reducing ulcer recurrence. [164] *H pylori*–positive long-term NSAID users who had ulcers at baseline (41 gastric and 46 duodenal ulcers) were randomly assigned to receive omeprazole with or without eradication therapy. Relapse was defined as recurrent ulcer and/or dyspepsia. Contrary to the findings in NSAID-naive subjects, curing *H pylori* infection did not reduce the relapse rate in long-term NSAID users. [160] How could *H pylori* affect NSAID-naive patients and long-term NSAID users in a different way? One explanation is that *H pylori* contributes to an excessive ulcer risk in NSAID-naive subjects, whereas NSAIDs cause most of the ulcers in long-term users of these agents irrespective of *H pylori* status. Epidemiologic studies have consistently shown that the risk of ulcer disease is substantially higher among patients who recently start taking NSAIDs than among long-term NSAID users. [161] This excessive ulcer risk may occur in a subgroup of *H pylori*–infected persons who are prone to develop ulcer on exposure to NSAIDs; that is, initiation of NSAID treatment aggravates or precipitates ulcer disease in these susceptible patients. Weeding out of susceptible patients will select a group of patients who can tolerate long-term NSAIDs irrespective of the *H pylori* status, and this explains why some studies did not find *H pylori* as a risk factor in long-term NSAID users. [162] One study raised concern that curing *H pylori* could impair ulcer healing. [163] In a subgroup of 163 patients with gastric ulcers, eradication of *H pylori* was associated with delayed ulcer healing; however, this finding was not confirmed by another larger randomized trial using ulcer healing as the predefined end point. [164] The lack of convincing evidence that curing *H pylori* infection has an adverse impact on the healing of NSAID ulcers supports the concept that antibiotic therapy should be offered to all patients. A randomized trial compared eradication of *H pylori* with maintenance omeprazole for secondary prevention of recurrent ulcer bleeding in patients with *H pylori* infection who were taking NSAIDs or low-dose aspirin. [165] Among long-term users of NSAIDs, eradication of *H pylori* alone was not sufficient to prevent recurrent bleeding. In contrast, among users of low-dose aspirin, eradication of *H pylori* alone was comparable to maintenance omeprazole in reducing the incidence of recurrent bleeding. Preliminary data have suggested that the combination of *H pylori* eradication and long-term treatment with a PPI may be superior to *H pylori* eradication alone for secondary prevention of ulcer bleeding associated with low-dose aspirin. [166] The divergent outcomes between low-dose aspirin and NSAIDs suggest that the increased ulcer risk with NSAIDs appears to overshadow the *H pylori*–attributable risk. Because low-dose aspirin has a much lower overall risk, *H pylori* may play a more important role in ulcer bleeding associated with this therapy than in bleeding associated with NSAIDs. Based on these observations, it appears that eradication of *H pylori* may be considered a cost-effective strategy to reduce ulcer risk in low-dose aspirin users and among NSAID users at high risk. [167] It remains clear that the residual increased risk associated with the aspirin and NSAIDs is not eliminated, and the use of cotherapy to prevent ulcers in these situations should be individualized based on clinical circumstances. In the case of the COX-2–specific inhibitors, the weight of current evidence suggests minimal interactions with *H pylori* infection. However, the elimination of this risk factor in patients taking these agents has the potential to reduce their overall risk of ulceration further.

Pathogenesis of Helicobacter pylori–induced peptic ulcer disease. The mechanism by which *H pylori* leads to gastric and duodenal ulceration has not been established. The organism colonizes many persons, yet the percentage of carriers who develop clinical sequelae is rather small. This discrepancy in carrier rates and development of disease results from multiple factors, including bacterial characteristics, variations in the host inflammatory response to the organism, and varying types of interactions between the host and the bacteria.

Gastric colonization. One of the most intriguing aspects of *H pylori* is its ability to inhabit and survive within the highly acidic conditions found within the stomach. Sachs and colleagues [168], [169] shed significant insight into *H pylori*–mediated acid resistance, a key feature of which is the expression of urease. Synthesis of this enzyme accounts for 15% of protein produced by the organism. The urease gene cluster consists of seven genes (*UreA* and *UreB* and *UreE* to *UreI*) that encode the structure and regulate portions of the enzyme. [170] *UreI* is a proton-gated urea transporter, which increases access of gastric juice urea to the intrabacterial urease by 300-fold. The steady entry of urea facilitates the rapid and continuous buffering of the bacterial periplasm to 6.0. Maintenance of a pH in this range (~6) permits acid resistance and growth of the organism in an acidic pH. [168], [169]

Virulence factors. Various *H pylori* factors contribute to the ability of the bacterium to colonize gastric mucosa, to evade host defenses, and to damage host tissue. These virulence factors are encoded by specific gene sequences located in a so-called *pathogenicity island. H pylori* is genetically a highly diverse bacterial organism, and some of these genes may [171], [172], [173] and [174] affect the ability of this organism to colonize and ultimately induce disease in its host. One such gene is the expression of the Cag island, a 35- to 40-kb genetic element, that is associated with human disease (see later) and is present in approximately 60% of strains in the United States. Persons infected with CagA $^{+}$ strains are at increased risk of peptic disorders as well as distal gastric cancer. *CagA,* which encodes CagA (cytotoxin-associated gene protein), and a gene (*picB*), which encodes a product that promotes induction of cytokines, are essential for induction of proinflammatory cytokine activation as well as for transporting proteins into host cells. [176], [177] and [178] Approximately 50% of *H pylori* strains produce a vacuolating cytotoxin (VacA), which is related to the pathogenicity island. The *vacA* gene has a very variable structure and is near, but not within, the pathogenicity island. Expression of the VacA cytotoxin usually is associated with the presence of *cagA* in the pathogenicity island. Thus, in general, those strains that are CagA $^{+}$ are VacA $^{+}$. The function of CagA is unknown, but once in the host cell, CagA is phosphorylated and is activated by a eukaryotic tyrosine kinase. [175], [179], [180] Once activated, CagA causes alteration in cell cycle and morphology, and it induces inflammation through regulation of interleukin-8 (IL-8) production by nuclear factor-?b and mitogen-activated protein kinases signaling pathways. The question remains whether these cellular changes indeed lead to peptic injury and malignant transformation. CagA expression was initially reported to represent an enhanced risk for the development of both gastric cancer and duodenal ulcer disease. However, the proportion of *H pylori* bacteria that possess the *cagA* gene varies in different geographic areas and populations infected with *H pylori.* In addition, other studies have not demonstrated an increased risk of severe gastroduodenal disease in association with CagA expression. Despite such data, the possibility that specific strains may be associated with specific diseases, especially duodenal ulcer disease, remains attractive. [181]

Mucosal immune responses. Paradoxically, *H pylori* induces host responses that promote inflammation and epithelial damage without conferring immunity against infection. Responses to *H pylori* infection include increased IL-1, IL-6, IL-8, and tumor necrosis factor-a expression (TNF-a), [182], [183] which may recruit and activate monocytes and neutrophils. Release of neutrophil mediators may, in turn, further disrupt epithelial cells and contribute to ulcer formation. *H pylori* may also induce B-cell–mediated autoimmune response. [184] *H pylori* lipopolysaccharide expresses antigens that cross-react with epithelial cell antigens. [185] Antibodies to *H pylori* may recognize these antigens and induce gastritis. *H pylori* infection is also associated with increased apoptosis. [186], [187]

Gastrin release and acid secretion. *H pylori*–positive patients have significantly higher basal, 24-hour, meal-stimulated, bombesin-stimulated, and gastrin-releasing peptide (GRP)– stimulated gastrin levels than do persons cured of *H pylori* infection. [188], [189], [190], [191], [192], [193], [194] and [195] Although ammonia produced by *H pylori* urease may raise the pH at the epithelial surface, increased gastrin is found when gastric contents are buffered to eliminate the pH effects, [188], [196] and acutely giving urea by mouth, by intragastric instillation, or by reducing urease activity does not alter serum gastrin level. [192], [197], [198] Hypergastrinemia in *H pylori*–infected patients may result from a decrease in acid secretion inhibiting antral somatostatin-producing D cells. Cure of *H pylori* reverses the decline in somatostatin mRNA. [199], [200] Decreased release of somatostatin is a plausible explanation for increased gastrin release. *H pylori* factors that inhibit somatostatin release have not been identified, although TNF-a has been implicated. [201] It is also possible that constituents of *H pylori* and proinflammatory cytokines may have direct stimulatory effects on gastrin release. [202] Basal and GRP-stimulated acid secretion is increased in patients with *H pylori*–associated duodenal ulcer disease as compared with asymptomatic infected volunteers, suggesting a greater acid response to gastrin in patients with duodenal ulcer disease. After cure of *H pylori* infection in patients with duodenal ulcer, acid secretion may either decrease by approximately 50%, into the range of uninfected healthy volunteers, or remain unchanged. [190], [191], [195], [203], [204], [205] and [206] Acid secretion is also decreased in response to GRP stimulation. In most, but not all studies, however, pentagastrin-stimulated acid output did not decline after cure of *H pylori* infection. A complete understanding of the complex effect of *H pylori* on acid secretion is hampered by the heterogeneity of duodenal ulcer disease. A summary of the potential impact of *H pylori* on gastric acid secretion is given in Figure 66-7.

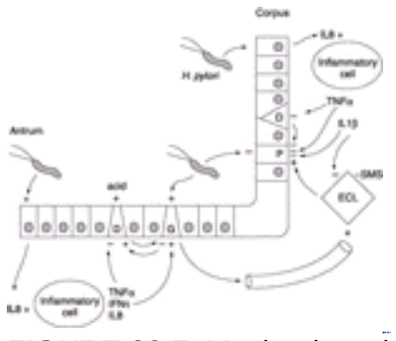

FIGURE 66-7. Mechanisms by which *Helicobacter pylori* may induce abnormalities in gastric acid secretion. *H pylori* may have both direct and indirect (through proinflammatory cytokines) effects on gastric parietal cell activity. *SMS,* somatostatin; *ECL,* enterochromaffin-like cell. (Adapted from Calam J, Gibbons A, Healey ZV, Bliss P, Arebi N. How does *Helicobacter pylori:* cause mucosal damage? Its effect on acid and gastric physiology. Gastroenterology 1997;113:S43.)

Mucosal bicarbonate secretion. Both basal and stimulated HCO_3^- production in the proximal duodenum is markedly diminished in patients with active duodenal ulcer disease, but distal duodenal HCO_3^- production is normal. Unlike other pathophysiological defects, there is less overlap (about one fourth) between those with and without ulcers. The mechanism of this defect, whether cause or effect, is under investigation. Prostaglandin E_2 (PGE_2), a potent agonist of gastroduodenal HCO_3^- production, stimulates significantly less HCO_3^- production in patients with quiescent duodenal ulcers, suggesting a cellular or subcellular defect in HCO_3^-. [87] Moreover, histological evaluation of the duodenum in patients with inactive duodenal ulcer fails to reveal any striking morphologic differences from findings in healthy

persons, and cure of *H pylori* infection normalized duodenal HCO_3^- secretion in patients with duodenal ulcers. In contrast, HCO_3^- secretion is not impaired in persons with *H pylori* infection who do not have ulcers, and in this group, HCO_3^- secretion does not change after *H pylori* cure. [207]

Gastric metaplasia in the duodenum. One hypothesis linking *H pylori* to duodenal ulcer disease is that *H pylori* organisms can only colonize areas of gastric metaplasia in the duodenal bulb, leading to active duodenitis and eventually to duodenal ulcer formation. Indeed, gastric metaplasia with active duodenitis is found in most, [208], [209] and [210] but not all, [211], [212] and [213] patients with duodenal ulcers. Moreover, gastric metaplasia in the duodenum is a patchy, multifocal process that is commonly found in healthy persons. [212], [213] and [214] There are conflicting data correlating the possible relationship among pH, gastric metaplasia in the duodenal bulb, and duodenal ulcer disease. [205], [206] and [207] In addition, others have found a similar prevalence of gastric metaplasia among patients with duodenal ulcers and nonulcer dyspepsia. [215] Furthermore, these latter investigators observed that, although pH values decrease in parallel with a greater extent of metaplasia in patients with nonulcer dyspepsia, the converse is true in patients with duodenal ulcer disease. Such data suggest that gastric acid does not determine the prevalence or extent of duodenal metaplasia. Overall, the association between gastric metaplasia and *H pylori* in the pathogenesis of duodenal ulcer, although conceptually attractive, is as yet unsubstantiated.

Nonsteroidal Antiinflammatory Drugs and Ulcer Disease

Epidemiology. It is estimated that 14 million persons in the United States take NSAIDs daily. In 1991, more than 70 million NSAID prescriptions in the United States were filled, representing 278 prescriptions per 1000 population, with ibuprofen and naproxen the most commonly prescribed agents. [216] In 2000, after the introduction of the COX-2–specific inhibitors, more than 111 million prescriptions were written in the United States at a cost of more than $4.8 billion. [217] Furthermore, the evolving interest in the ability of low doses of aspirin to reduce cardiovascular events and to protect against colorectal cancer has likely led to increased patient exposure to aspirin and its potential for GI injury. Currently, annual sales in the United States of over-the-counter agents of NSAIDs and aspirin together approach $2 billion. [218] NSAID-induced patient morbidity can range from symptoms such as nausea and dyspepsia (experienced by as many as 60% of users) to serious adverse GI events such as bleeding and perforation (estimated to affect 2% to 4% of users per year). In the United States, [219] it is estimated that more than 100,000 hospitalizations (at a cost of at least $2 billion) and as many as 10,000 to 20,000 deaths annually can be attributed to NSAID-related complications. [220] Although the probability of a drug-related complication in an individual patient is low, the vast numbers of patients exposed to NSAIDs translate into a considerable problem from the health care economic perspective. Estimated costs for excess hospitalizations, outpatient physician visits for symptoms, laboratory testing, and diagnostic studies such as endoscopy for NSAID toxicity seriously underestimate the true societal impact of this condition because indirect costs such as work loss and other lost productivity tend not to be included in these calculations. [221] In an effort to inform clinicians better, the United States Food and Drug Administration (FDA) and its counterpart in the United Kingdom, the Committee on Safety of Medicines, issued formal warnings regarding the risk of adverse GI events in patients using these medications.

Mechanisms. The mechanisms by which NSAIDs induce mucosal injury remain incompletely understood but appear to involve both direct topical injury and systemic effects mediated by prostaglandin depletion.

Topical damage. Aspirin and most NSAIDs are weak organic acids with ionization constants (pK_a) in the range of 3 to 5. [222] In the strongly acid environment of gastric juice (pH <2.5), these lipidic drugs are soluble and nonionized and freely diffuse across cell membranes into mucosal cells. Once inside the cell, the elevated intracellular pH favors dissociation of the H^+ ion and trapping of the negatively charged acid moiety. Because the nonionized form remains in equilibrium across the cell membrane, the total intracellular drug concentration (ionized and nonionized) is much higher than outside the cell, an effect known as *ion trapping* (Fig. 66-8). Ion trapping promotes direct toxic cellular injury and leads to abnormal ion flux across the epithelium, with increased H^+ back-diffusion. The exact mechanisms by which high cellular levels of NSAIDs damage the surface epithelial cells are not known. Changes in membrane permeability may be secondary to impairment of energy metabolism as a result of uncoupling of oxidative phosphorylation with secondary inhibition of ion transport, [223] inhibition of mucosal prostaglandin secretion, reduction of mucus secretion, and interference with cell turnover. [224] NSAIDs also directly attenuate the hydrophobic, or “nonwettable,” properties of the mucous barrier independent of prostaglandin-mediated actions. In short-term studies in rats, preassociation of NSAIDs with surface-active phospholipids was shown to prevent ulceration even in the presence of prostaglandin depletion. [61]

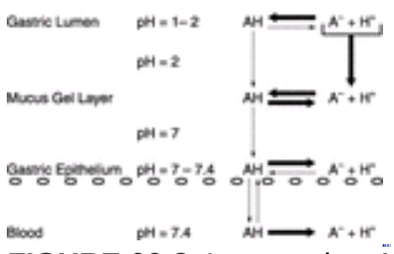

FIGURE 66-8. Ion trapping. Nonsteroidal antiinflammatory drugs (NSAIDs) are weak organic acids with ionization constants (pK_a) in the range of 3 to 5 and thus are soluble at the low pH in the stomach lumen. In the strongly acidic environment of gastric juice (pH of less than 2.5), these lipidic drugs are nonionized and freely diffuse across cell membranes into mucosal cells. Once inside the cell, the elevated intracellular pH favors dissociation of the H^- ion and trapping of the negatively charged acid moiety. Because the nonionized form remains in equilibrium across the cell membrane, the total intracellular drug concentration (ionized plus nonionized) will be much higher than outside the cell. Ion trapping allows direct cellular injury resulting from the toxicity of high intracellular levels of the NSAID. (Adapted from ref. [223].)

Systemic effects. The clinically important effects of NSAIDs–the production of ulcers with an increased risk of significant complications—appear to be largely caused by their systemic actions. [225] Inhibition of COX-1with a resultant decrease in endogenous prostaglandins, especially PGE_1, PGE_2, and PGI_2, is thought to be the most important mechanism of action. [225], [226] and [227] Traditional NSAIDs have been shown to inhibit plasma and mucosal prostaglandin production in humans, whereas COX-2 inhibitors lack this effect and are therefore associated with reduced rates of ulcers and complications. [228] Mucosal prostaglandins play an important role in normal mucosal defense, and, thus, NSAIDs may compromise mucosal mechanisms that prevent the development of gastroduodenal ulceration resulting from lumenal acid. [225] Platelet COX-1 is also inhibited irreversibly by aspirin and for as long as 18 hours by other NSAIDs. The resulting decreased platelet aggregation and prolonged bleeding times may potentiate GI bleeding from the upper and lower GI tract. [229] Three lines of evidence support the importance of mucosal prostaglandin depletion in the development of NSAID-induced ulcers. First, active or passive immunization against PGE_2, PGF_2a, or PGI_2 in animals leads to formation of ulcers that grossly and histologically resemble ulcers associated with long-term NSAID use. [230] Second, coadministration of the PGE_1 analog misoprostol with a variety of NSAIDs dramatically reduces the subsequent development of mucosal ulceration. [231], [232] Third, analgesic or antiinflammatory agents that do not inhibit COX or reduce prostaglandin production, such as acetaminophen or the COX-2–specific inhibitors, do not cause mucosal ulceration. [227], [228] Although inhibition of prostaglandin synthesis contributes significantly to NSAID-induced mucosal injury, there may be other important mechanisms of injury. Some studies have found a poor correlation between mucosal prostaglandin inhibition after NSAIDs and the degree of acute mucosal injury. [233], [234] Maximal mucosal prostaglandin and thromboxane inhibition occurs with doses of aspirin as small as 30 to 80 mg/d. However, clinical experience suggests that ulcer complications are less common at these low doses and appear to increase at higher aspirin doses. [234], [235] By inhibition of prostaglandin production, NSAIDs interfere with all three lines of mucosal defense: [236] mucous cell secretion of mucin and surface-active phospholipid, [237] basal bicarbonate secretion from gastric and duodenal mucosa (two defects that combine to form a mucous gel poorly suited to perform its role as a barrier to acid diffusion), and the mucosal proliferation critical to ulcer healing. [74] Prostaglandins also play an inhibitory role in the physiological regulation of acid secretion. [238] Feedback inhibition of gastric acid secretion is also dependent on endogenous prostaglandins; thus, NSAIDs may impair mucosal defense and increase acid secretion simultaneously. [239] Interruption of mucosal blood flow is an early event in experimental NSAID-induced mucosal injury, [240] which occurs, in part, by causing neutrophil adherence to the gastric vascular endothelium. This adherence is blocked by exogenous prostaglandin, neutrophil depletion, and antibodies to adhesion molecules, and it is characterized by up-regulation of intercellular adhesion molecule-1 (ICAM-1), an important endothelial adhesion molecule. [241], [242] Neutrophil adherence can also be blocked experimentally by administration of pentoxifylline, an agent that reduces TNF release and subsequent ICAM-1 expression. [243] Cytokines such as IL-1 reduce experimental NSAID injury, presumably by affecting neutrophil function. [244] The somatostatin analog octreotide also appears to have a similar capacity to block neutrophil-endothelial interaction and to reduce NSAID injury. [245], [246] Attention to the role of NO in the maintenance of epithelial integrity has focused on its role in mucosal blood flow. In animal models, inhibition of NO synthesis exacerbates NSAID injury, and NO donors reduce NSAID toxicity. [247] Data have demonstrated that induction of ulceration may be related to inhibition of COX-1 and COX-2 simultaneously. In an animal model, pure inhibition of either COX-1 or COX-2 did not cause damage, but only the combination led to ulceration. [248] Neutrophil adherence appears related to COX-2 inhibition, which also plays a role in regeneration. Furthermore, some studies have suggested that as a consequence of COX inhibition, arachidonic acid metabolism may be shunted toward the lipoxygenase pathway, resulting in increased synthesis of leukotriene B_4, a potent chemotactic agent for neutrophils. [243] Although the topical effects can be largely prevented by administering enteric-coated NSAID formulations or prodrugs, the failure of these agents to reduce the incidence of long-term NSAID-induced ulcers or complications implies that topical injury is not the most important component of NSAID-induced injury. [249] Parenteral administration of the NSAID ketorolac has been demonstrated to cause gastric ulceration, which proves that the systemic action of NSAIDs (in the absence of any direct topical

effects) is sufficient to cause mucosal ulceration. [250] Certain NSAIDs, such as indomethacin, piroxicam, oxaprozin, and ketorolac, may also have indirect topical effects because their entry into enterohepatic circulation results in repeated exposure to the GI mucosa. Increased intestinal concentrations of the active drug by this mechanism may contribute to small intestine and colonic ulcerations, strictures, and perforations, which are also noted with slow-release NSAID preparations such as diclofenac. [251], [252] Strategies to avoid local toxicity have included the use of enteric coating or prodrugs. although enteric-coated aspirin causes less gastroduodenal injury than plain or unbuffered aspirin, it can cause significant gastroduodenal ulceration. [253], [254] and [255] Prodrugs (such as sulindac) that avoid proximal GI absorption produce little acute superficial damage but appear to lack significant protective advantage against risk of upper GI bleeding compared with other NSAIDs. [256] Finally, acid may play an important secondary role for the development of NSAID-related ulceration of the proximal GI tract. The efficacy of high dose H_2 blockers and PPIs for the prevention of NSAID-induced ulcers has provided support for this concept. [257], [258] These agents primarily reduced ulcers, but, unlike misoprostol, they did not reduce the number of erosions, the presumed precursor lesion. This suggests that topical injury initiates the erosive disease induced by NSAIDs, and that acid and prostaglandin depletion are the essential components for the development of clinically important gastric and duodenal ulceration (Fig. 66-9).

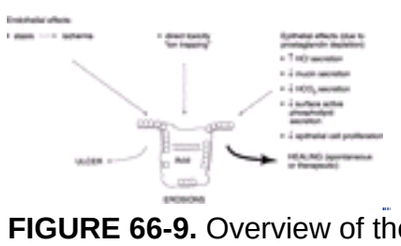

FIGURE 66-9. Overview of the pathogenesis of mucosal damage induced by nonsteroidal antiinflammatory drugs. (Adapted from Scheiman JM, Fendrick AM, Conaway DC. Optimizing outcomes for patients at risk for NSAID-induced ulcer disease. J Clin Outcomes Management 1996;3:23.)

Superficial mucosal lesions. Subepithelial petechiae or hemorrhages and erosions in the fundus may be seen with acute NSAID ingestion within 1 hour of ingesting a single dose of aspirin. Repeated doses of aspirin over a 24-hour period lead to gastric erosions, predominantly in the antrum. [233], [259], [260] If aspirin is continued for several days, the petechiae and erosions become less prominent, a process known as *gastric adaptation.* [261] Despite adaptation, gastric erosions are found at endoscopy in 30% to 50% of patients receiving long-term NSAIDs. [233], [259], [262] There is controversy with regard to the relationship between the presence or severity of superficial mucosal lesions and the subsequent development of clinically significant gastroduodenal ulceration, [263] although the presence of erosions at baseline endoscopic examination has predicted the development of larger endoscopic lesions in at least one study. [264] Scoring systems, devised to grade the severity of these superficial lesions, are useful in clinical investigation but have no utility in the clinical setting. [265] These superficial lesions appear to be caused largely by direct topical effects of the ingested agent and are of little clinical importance. No correlation exists between symptoms of dyspepsia and the number or severity of the lesions, [260], [262] and they are not associated with perforation or serious hemorrhage. [233] Although there may be considerable differences in incidence of superficial injury after short-term administration of enteric-coated or buffered NSAID formulations, these drug modifications have not significantly altered the incidence of chronic gastric ulceration or of NSAID-associated complications. [222], [224], [233] Studies of mucosal injury after acute NSAID administration do not provide uniformly reliable information for predicting the likelihood of clinically significant gastroduodenal injury after long-term administration of any given NSAID. [224]

Ulcers. Long-term NSAID ingestion may cause gastroduodenal ulcerations, but the relative risk appears to depend on the definition of an endoscopically confirmed ulcer, the dose and duration of NSAIDs used, and the characteristics of the patient population. Although the histological definition of an ulcer is a defect that penetrates the muscularis mucosa, endoscopic definitions typically are based on the size of the lesion. [233] The selection of a cutoff size, usually 3 to 5 mm, greatly influences the reported ulcer prevalence; studies that include 3-mm lesions probably include too many erosive lesions as ulcers. [266] Using a 5-mm criterion, the incidence of acute gastric ulcers was 6% to 12% and that of duodenal ulcers was 4% to 8%. [267], [268] It is unclear whether the endoscopically defined ulcers developing acutely during short-term NSAID therapy have the same natural history as do long-term NSAID-induced ulcers. In cross-sectional studies in which endoscopy is performed in medical patients receiving long-term NSAIDs, the prevalence of gastric ulcers ranged from 9% to 31%, and the prevalence of duodenal ulcers was 0% to 19%. [233], [266], [269], [270] The crude prevalence rate of gastric ulcer in long-term NSAID users is about 13%, and that of duodenal ulcer is about 11%; therefore, roughly 25% of patients taking long-term NSAIDs can be expected to have an endoscopically demonstrable gastric or duodenal ulcer. [233], [266] The calculated relative risk compared with that of a healthy population is a 46-fold increase for gastric ulcers and an 8-fold increase for duodenal ulcers. [266] In case control studies that have evaluated the risk of NSAID-induced ulceration relative to a matched control population, the relative risk of gastric ulceration was 4.0 and of duodenal ulceration was 1.7. [233], [271]

Associated symptoms. Dyspeptic symptoms, although extremely common in patients taking NSAIDs, [236], [262], [266], [272] do not reliably differentiate those who have ulceration. [235], [273] In the Aspirin Myocardial Infarction Study, severe dyspepsia was observed in 24% of patients taking aspirin (0.5 g, three times daily) and in 15% of patients given placebo. [274] Dyspepsia is more common in the first few weeks of therapy, and it tends to decline over time, regardless of treatment, [236] but it may be relieved by food, antacids, sucralfate, and antisecretory agents. [225], [275] Symptoms that are unrelieved by H_2 antagonists or lead to cessation of NSAID therapy may indicate underlying ulcers. [256] Patients with NSAID-induced ulcer complications appear less likely to have antecedent symptoms than do other patients with ulcers. [259] Thirty to 40% of patients with significant mucosal ulcerations are asymptomatic; [259], [262], [270], [276] and 60% who develop bleeding are asymptomatic, compared with 25% of asymptomatic bleeding ulcers occurring without NSAID use. [277] In a cross-sectional population study of long-term NSAID users, the positive predictive value of dyspepsia for the presence of an ulcer was only 26%, but the negative predictive value was 93%. [270] Thus, dyspepsia has little discriminatory value in identifying the subsets of long-term NSAID users most likely to have mucosal ulceration or ulcer complications. Dyspeptic symptoms may prompt a diagnostic evaluation or therapy, but the clinically silent ulcer usually remains undiagnosed, untreated, and at risk of complications.

Risk of developing complications. NSAID ingestion is associated with a significantly increased risk of GI complications, as determined by case control and cohort studies in which the frequency of the complication in NSAID users is compared with a matched control group of persons who do not use NSAIDs. [233] In most studies, the risks reported reflect GI bleeding, perforation, surgery, or death. [225] The differences in the reported relative risks may reflect differences in the study population, definition of outcome measures, type and dosage of NSAID, method of ascertaining use of NSAIDs, duration of treatment, and exclusion criteria. [225], [233], [276], [277], [278] and [279] The overall odds ratio of the risk of adverse GI events of all types related to NSAID use appears to be increased approximately threefold to fivefold. [161], [279] The estimation of relative risk may be assessed with prospective studies. The risk of developing an ulcer complication in a group of patients without a previous history of PUD is estimated variably as 5.5 for every 1000 patient-years, [274] or 22 for every 1000 patient-years, or 2% annually. This figure is in accord with observed complication rates of 1% to 3% per year in a Tennessee Medicaid population, of 1.6% per year from the Arthritis, Rheumatism and Aging Medical Information System (ARAMIS) study of patients with rheumatoid arthritis, of 2% to 4% per year from NSAID premarketing investigational new drug studies, and 1.5% in the 4000 subject, 3-year, Aspirin Myocardial Infarction Study. [276] It is estimated that approximately 30% of cases of upper GI bleeding and 29% of ulcer-related deaths are directly attributable to the effects of NSAIDs. [280], [281] and [282]

Risk factors for complications. Most NSAID users derive benefit from these agents and do not develop complications. To optimize clinical management, it would be helpful to predict which patients are at increased risk. Although the factors that predispose patients to complications are not well understood, clinical studies have identified several potentially important risk factors (Table 66-2).

Age > 60 years
Previous ulcer disease or bleeding
Concomitant corticosteroid therapy
High-dose, multiple-NSAID therapy
Chronic major organ impairment, e.g., cardiovascular disease
Concomitant anticoagulant use

*Possible: duration of NSAID therapy, Helicobacter pylori infection, dyspepsia on NSAID therapy, and extent of disability with rheumatoid arthritis. Unclear: cigarette smoking.
NSAID, nonsteroidal antiinflammatory drug.

TABLE 66-2 Risk Factors for Ulcers Induced by Nonsteroidal Antiinflammatory Drugs and Their Complications*

Prior history of peptic ulcer disease or gastrointestinal bleeding. The incidence of NSAID-induced ulcer complications, especially hemorrhages, is as much as 14-fold higher in those with a previous history of GI bleeding or ulcer disease. [161], [225], [276], [282], [283] The risk is highest in the first 1 to 3 months after initiation of NSAID therapy, suggesting that the NSAIDs exacerbate an underlying ulcer condition. [161], [280] Because most prospective, controlled NSAID trials have excluded patients with a known history of ulcer disease or GI bleeding, this high initial risk was not noticed. [225], [274] Extreme caution is advised in using NSAIDs in patients with a history of prior ulcers or ulcer complications, and prophylactic antiulcer therapy is recommended for this group of patients.

Increased risk in the elderly. Advancing age is a strong risk factor for NSAID-associated complications; [233], [276], [281] it is greater than 5 in NSAID users older than 60 years of age, but less than 2 in younger patients. [161] In the prospective Aspirin Myocardial Infarction Study, there was a linear relation in the aspirin-treated group

between ulcer complications and the age of the patient. [233], [274]

Relation to drug dose. The overall risk of NSAID-associated complications is dose related, [256], [279], [280] although complications occur even at low NSAID doses, particularly with aspirin. In the Physician's Health Study, healthy male physicians given aspirin (325 mg/d) had twice the rate of symptomatic duodenal ulcer and a 1.7 times higher rate of GI bleeding than placebo-treated persons, [284] whereas an aspirin dose three times that amount (the Aspirin Myocardial Infarction Study) was associated with an eightfold increased risk of ulcer complications compared with placebo. [274] The Dutch TIA Trial compared 30- and 283-mg doses of aspirin and found some major GI bleeding episodes even at 30 mg. [285] There may not be a truly safe aspirin dose for the GI tract, and the small antithrombotic effect may be offset by bleeding complications. [278] The doses of aspirin associated with hemorrhage are far lower than those generally associated with developing a gastric ulcer. An increased risk of developing GI bleeding is demonstrable within 30 days of initiating NSAID therapy, presumably before most NSAID-induced ulcers would develop de novo. [161] Although the incidence of de novo NSAID-induced gastric ulcers is greater than that of duodenal ulcers, the incidence of bleeding from these two sites is equivalent. [225], [233] These observations suggest that NSAIDs, even in low doses, can potentiate bleeding from preexisting, perhaps *H pylori*–associated, lesions, likely through their ability to reduce platelet function. In a study of consecutive patients admitted with upper or lower GI bleeding, evidence of current overt or covert NSAID ingestion (determined from platelet COX inhibition) was found in 80% of cases, compared with 24% of a matched control population. [229] Peptic ulcers accounted for 42% of upper GI bleeding; 80% of patients with bleeding duodenal ulcers and 100% of those with bleeding gastric ulcers had evidence of NSAID ingestion. NSAIDs were also implicated in 60% of the remaining cases of upper GI bleeding and in 85% of those with lower GI bleeding. [229]

Nonsteroidal antiinflammatory drugs and corticosteroids or anticoagulants. Despite a widespread impression among clinicians that corticosteroids cause peptic ulcers, the results of previous studies evaluating this association have been conflicting. [286], [287] The risk of developing an ulcer is not increased in corticosteroid users who are not taking NSAIDs, [288] and relative risk of complicated PUD in corticosteroid users is increased only in those concurrently taking NSAIDs (2X). [161] Thus, routine use of prophylactic antiulcer medications is not indicated for patients receiving steroid therapy, unless the patients are taking corticosteroids with NSAIDs. [289] The use of NSAIDs in combination with anticoagulants has been demonstrated to increase GI bleeding risk markedly, [290] presumably through exacerbation of bleeding from NSAID-induced lesions. NSAIDs should be used with extreme caution in patients receiving long-term anticoagulants.

Other possible risk factors. Several other factors have been implicated in the potentiation of NSAID-induced GI complications, including the use of tobacco and alcohol, as well as gender. [161] In the Misoprostol Ulcer Complications Outcomes Safety Assessment Trial (MUCOSA), cardiovascular disease was identified as a risk factor for complications. [291] Rheumatoid arthritis appears to be associated with increased risk in several studies. [291], [292] and [293] The observation that NSAID users in the ARAMIS database who had rheumatoid arthritis and who were taking antacids and H *2* blockers were at increased risk of GI complications requires further study, particularly given the observation that these agents have demonstrated efficacy to reduce ulcers. [294] The role of comorbid conditions, such as renal disease or other serious medical illness, requires further study.

Specific drugs and relative frequency of injury. NSAIDs are categorized in several chemical classes, which relate to their absorption, pharmacokinetics, and therapeutic performance, but there is little evidence of substantial differences in clinical efficacy or adverse GI side effects for most nonsalicylate NSAIDs. [256], [279], [295], [296] At equipotent antiinflammatory doses, there remains uncertainty with regard to differences in the frequency of upper GI complications among most users of NSAIDs. [259] However, some patterns have arisen with regard to difference among NSAIDs in clinical practice. In general, the potency and duration of COX-1 isoenzyme inhibition and subsequent depletion of upper GI prostaglandins correlate with development of ulcer disease and complication risk. [225], [295] This has been confirmed by metaanalysis [161] and by large, population-based, retrospective case cohort studies. [279] As shown in Table 66-3, there is considerable overlap in the risk ratios for complications among the various NSAIDs and their confidence intervals, demonstrating significant variations in individual NSAID risk. [296] It remains unclear whether there are truly substantial differences among NSAIDs when they are given at equivalent antiinflammatory doses.

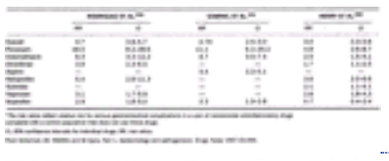

TABLE 66-3 Relative Risk Associated with Different Nonsteroidal Antiinflammatory Drugs*

The nonacetylated NSAIDs, as well as several newer NSAIDs, may be associated with a significantly decreased incidence of ulceration and serious GI side effects. The nonacetylated salicylates (salicylsalicylic acid) such as salsalate and the salicylate salts have minimal inhibition of COX and therefore do not diminish mucosal or systemic prostaglandin levels and do not affect platelet function. [227] In the absence of significant systemic prostaglandin inhibition, it is unclear how the antiinflammatory efficacy of salsalate and aspirin appear to be comparable in patients with rheumatoid arthritis. [297] Salsalate is insoluble in gastric acid, is not absorbed by the gastric mucosa, and produces minimal direct mucosal injury compared with other NSAIDs. [298] When compared with other commonly used NSAIDs, salsalate appeared to produce fewer adverse side effects. [255], [298], [299] Nabumetone and etodolac are purported to have a significantly decreased incidence of ulceration and serious GI side effects. [300], [301] Although these agents can inhibit systemic prostaglandin production, gastric and duodenal prostaglandin production appears to be relatively spared. [300], [302] The explanation for this has not been determined, but it may to be related to the relative selectivity of these agents for COX-2 compared with the COX-1 isoenzyme. These agents may have less inhibition of platelet aggregation, potentially decreasing the likelihood of bleeding complications. [301] In prospective endoscopic trials of patients with arthritis, the incidence of peptic ulceration after 3 months of nabumetone was 2% to 5%, compared with 14% to 25% in patients treated with naproxen or indomethacin. [303], [304] Other short-term endoscopic studies with etodolac also suggest a lower incidence of mucosal injury with these agents than with other NSAIDs. [300], [305] Postmarketing surveillance studies of large cohorts of patients who are taking these agents report an incidence of clinically evident ulcers of less than 1% and of major GI complications of less than 0.1%. [306] It appears from these limited data that these newer NSAIDs may result in a lower incidence of gastroduodenal ulceration and serious GI complications. However, differences in study design preclude a direct comparison with the results of studies of older NSAIDs. Thus, although these agents appear promising, a true determination of their safety will depend on large case control studies that assess ulcer complications in patients taking newer NSAIDs versus those taking older NSAIDs.

Gastrointestinal safety of COX-2–specific inhibitors. Pharmacologists took advantage of subtle differences in the active site of the two forms of COX to develop molecules (celecoxib, rofecoxib) that are highly selective inhibitors of COX-2. As predicted, these drugs, even at very high doses, spare GI prostaglandins. When studied with endoscopy, both celecoxib and rofecoxib produce rates of injury nearly equivalent to those of placebo (Fig. 66-10). [307] Rofecoxib also has been shown to be equivalent to placebo in fecal blood loss and intestinal permeability studies in humans. [308] The ulcer risk associated with celecoxib has been evaluated by endoscopy in patients with osteoarthritis and rheumatoid arthritis in studies lasting 3 to 6 months. In a 3-month study of patients with osteoarthritis and rheumatoid arthritis, celecoxib, 200 mg twice daily, caused fewer endoscopic ulcers than naproxen, 500 twice daily, and ibuprofen, 800 three times daily, but not fewer than diclofenac, 75 mg twice daily. [309] One report compared the incidence of endoscopic ulcers in patients with rheumatoid arthritis who were taking celecoxib, traditional NSAIDs, and placebo. [310] The incidence of upper GI tract ulcers was 4 of 99 (4%) in the placebo group, 9 of 148 (6%) on 100 mg twice daily, 6 of 145 (4%) on 200 mg twice daily, 8 of 130 (8%) on 400 mg twice daily, and 36 of 137 (26%) on naproxen 500 mg twice daily. The ulcer rates with celecoxib were not significantly different from those with placebo, but they were significantly less than those with naproxen.

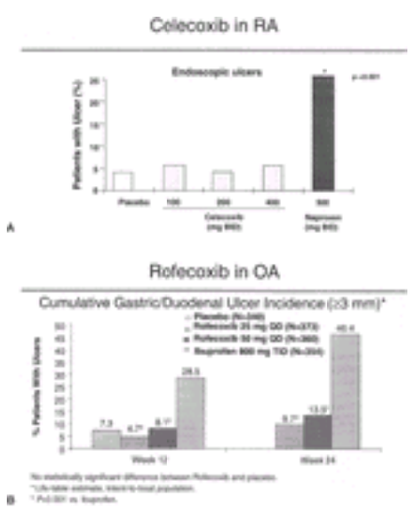

FIGURE 66-10. Incidence of peptic ulcers associated with selective cyclooxygenase-2 inhibitors. (**A**) Celecoxib leads to ulcer formation at a rate comparable to

placebo. (**B**) Rofecoxib is also associated with a rate of ulcer formation comparable to that seen with placebo. (*A,* Adapted from ref. [310]; *B,* adapted from ref. [311].)

Two 6-month, placebo-controlled, endoscopy studies were performed with rofecoxib, 25 and 50 mg, compared with ibuprofen, 800 mg three times daily, with a baseline endoscopy and endoscopies at 6, 12, and 24 weeks. In one study of 742 patients, at 12 weeks, 7.3% of patients receiving placebo developed an endoscopic ulcer compared with 4.7% taking 25 mg of rofecoxib, 8.1% taking 50 mg of rofecoxib, and 28.5% in the ibuprofen group. [56] The incidence of 5-mm lesions was similar in these two studies. In a second study (775 patients) of identical design, similar safety with rofecoxib was observed. When the two studies were combined, the incidence of endoscopic ulcers met predefined criteria for equivalence to placebo. [311] In an analysis of predictors of gastroduodenal ulcer development, the presence of erosions at baseline endoscopy, prior history of ulcer disease, age greater than 65 years, and the presence of *H pylori* were significant risk factors. The rofecoxib clinical trials examined the incidence of clinical ulcers (the so-called perforation, ulcer, and bleed or PUB rate) over 12 months, in more than 5000 patients. The drug was associated with a relative risk reduction of 0.51, [306] representing a PUB rate of 1.33 per 100 patient-years with rofecoxib compared with 2.60 with traditional NSAIDs. Most important, the incidence of clinically significant GI bleeding was also markedly reduced. In an analysis of the celecoxib clinical trials, the annualized incidence of upper GI complications with celecoxib was 0.20% compared with 1.68% with NSAIDs. [312] To confirm long-term safety and clinically important reduction in GI toxicity, both manufacturers performed long-term safety trials with the COX inhibitors. In a trial known by the acronym VIGOR, the safety of rofecoxib, 50 mg, was studied in comparison with naproxen, 500 mg twice daily, in more than 7000 patients treated worldwide over 6 to 12 months. [293] The primary and secondary end points of this study were the occurrence of symptomatic and complicated upper GI events. The results of the trial are shown in Table 66-4. The COX inhibitor was associated with a 50% to 60% reduction in ulcers, bleeding, or both.

TABLE 66-4 Event Rates in the Vioxx Gastrointestinal Outcomes Study (VIGOR)

In the celecoxib outcomes study, known by the acronym CLASS, 8000 patients worldwide with osteoarthritis (90%) and rheumatoid arthritis (10%) were randomized to celecoxib, 400 mg twice daily, ibuprofen, 800 mg three times daily, or diclofenac, 75 mg twice daily, in a study of 12 months' duration; an analysis of the first 6 months was published. [313] In this trial, the primary end point was also the development of complicated upper GI events. The secondary end point was the development of symptomatic plus complicated ulcers. Although the primary end point in all patients was not met in the CLASS trial (Table 66-5a), it was likely because of the inclusion of low-dose aspirin (Table 66-5b). A complete analysis of the entire 12 months of trial failed to show efficacy or safety of celecoxib compared with diclofenac, likely because of high dropout rates in the trial. [314] Despite the study design limitations, this work (see Table 66-5) suggests that the impressive GI safety of COX-2–specific inhibitors could be significantly impaired by the concomitant use of low-dose aspirin. Further studies to address this question are ongoing.

TABLE 66-5A Celecoxib Gastrointestinal Outcomes Study (CLASS): All Patients (Rates per 100 Patient-Years; 6-Month Data)

TABLE 66-5B Celecoxib Gastrointestinal Outcomes Study: Non-aspirin Users (Rates per 100 Patient-Years; 6-Month Data)

Extended scope of drug-induced complications. The GI complications of NSAIDs include a variety of lesions throughout the GI tract. [314], [315] and [316] Small bowel ulcerations have been identified at surgery, radiographically, and by enteroscopy. [316], [317] In a postmortem study, small bowel ulceration was found in 8% of NSAID users, compared with 0.6% of nonusers. [317] These ulcerations may be associated with occult GI blood loss and an increased risk of small bowel perforation. [317] NSAIDs also are associated with the development of weblike small bowel and colonic strictures that may produce clinically significant lumenal narrowing. In the colon, NSAIDs have been associated with the precipitation of relapses of established inflammatory bowel disease, and the development of acute colitis, strictures, and perforations. [318], [319] Whether COX-2–specific inhibitors have a reduced risk of intestinal complications is uncertain and requires further study. [314]

Summary of drug-induced complications. NSAIDs can cause acute and chronic gastroduodenal injury. The acute topical injury seen is dose dependent, varies with different NSAID formulations, declines with continued NSAID administration, is unrelated to symptoms, is not predictive of the development of chronic gastroduodenal ulceration or complications, and is of little clinical significance in most patients. Gastric or duodenal ulcers develop in approximately 25% of long-term NSAID users. In histologically normal mucosa or in mucosa inflamed because of infection with *H pylori,* NSAIDs may cause ulcers de novo or may exacerbate an underlying ulcer diathesis, but it remains unclear whether *H pylori* infection predisposes to NSAID-induced ulceration. The presence or absence of symptoms is of limited clinical utility in predicting which patients have ulceration or will have complications. Dyspepsia tends to decline with time and is relieved by antisecretory agents in most patients. Complications such as hemorrhage, perforation, and death occur in approximately 2% of long-term NSAID users per year. As many as 60% of complications may occur in asymptomatic patients, and these problems are more common in the elderly, in patients with prior peptic disease or ulcer complications, and in those receiving higher NSAID doses and concomitant corticosteroids. Complications arise from NSAID-induced ulcers and from exacerbations of preexisting (*H pylori*–related) ulcer disease. GI hemorrhage is potentiated by NSAID-induced platelet dysfunction. [320] Although COX-2 inhibitors have a favorable safety profile regarding GI toxicity, some reports have aroused concern regarding the potential prothrombotic and cardiovascular toxicity of these agents. [321]

Smoking and Peptic Ulcer Disease

Epidemiology and natural history. Although a large body of evidence demonstrates that cigarette smokers are at an increased (twofold) risk of developing ulcer disease with its related complications and, in addition, tend to have impaired ulcer healing rates. [322], [323], [324], [325], [326] and [327] This association was placed in doubt by Aldoori and associates. [328] In a prospective cohort of 47,806 men (40 to 75 years old), these investigators observed that neither current nor past smokers were at increased risk of development of a duodenal ulcer. The reason for this discrepancy may be the method of screening patients by questionnaire, which could be rather insensitive. Using metaanalysis of English-language papers, Kurata and Nogawa [329] examined the risk ratios for several factors, including cigarette smoking, and found that the attributable risk for cigarette smoking and peptic ulcer–related GI events was 23%. Cigarette smoking and *H pylori* are cofactors for peptic ulcer formation. [329] Svanes and colleagues [330] observed a tenfold increased risk of perforation in smokers, with a significant dose response relationship. An endoscopic study in patients with rheumatoid arthritis who were taking NSAIDs revealed that 63% of patients with gastric ulcers smoked, but only 33% without ulcers smoked. [330] This suggests that smoking and NSAID intake may be independent or additive risk factors for PUD.

Pathophysiological mechanisms. Despite the clear association between cigarette smoking and PUD, the pathophysiological basis for this association is not established. Smoking does not appear to have a consistent effect on gastric acid or pepsin secretion, [331] but it does appear to decrease the effectiveness of histamine H_2-receptor antagonists in duodenal ulcer healing. [332], [333] and [334] Cigarette smoking may increase the risk for infection with *H pylori,* especially in African Americans, [335], [336], [337] and [338] and it may potentiate the injurious effect of this pathogen on the gastric mucosa. [339] Additional factors that lead to mucosal injury and appear to be increased by smoking include free oxygen radicals, platelet-activating factor, pituitary vasopressin, gastric endothelin, and pituitary vasopressin. [331] Altered gastric motility has also been associated with smoking, with concomitant relaxation of the pylorus and reflux of duodenal contents into the stomach. [340], [341] Finally, smoking may also alter processes important in gastric and duodenal mucosal integrity or protection such as prostaglandin content, mucosal blood flow, or epidermal growth factor. [331]

Diseases Associated with Peptic Ulcer Disease Epidemiologic evidence demonstrates that duodenal ulcers occur more commonly in patients with certain chronic illnesses. However, studies reporting these associations did not consider *H pylori* status in the patients evaluated. Table 66-6 lists diseases associated with duodenal ulcer. The pathophysiological mechanisms by which some of these disorders increase peptic disorders are not clear.

Evidence Strongly Supports an Association
Zollinger-Ellison syndrome (gastrinoma)
Systemic mastocytosis
Multiple endocrine neoplasia type I
Chronic pulmonary diseases
Chronic renal failure
Hepatic cirrhosis
Kidney stones
α-antitrypsin deficiency
Evidence Only Suggests an Association
Crohn's disease
Hyperparathyroidism without multiple endocrine neoplasia type I
Coronary artery disease
Polycythemia vera
Chronic pancreatitis
Cystic fibrosis

Data from references: 361 to 373.

TABLE 66-6 Diseases Associated with Duodenal Ulcer

Role of Psychological Factors The role of psychological factors in the pathogenesis and natural history of PUD has been the subject of numerous studies, with conflicting conclusions. Early unblinded studies described certain personality characteristics thought to be markers for ulcer risk, including conflicts about dependency needs, hypochondriasis, and low ego strength. [342], [343], [344], [345] and [346] However, studies of patients in whom the diagnosis of duodenal ulcer disease was established endoscopically and with adequate controls (at least 50 persons in each study arm) concluded that several emotional factors (e.g., anger, anxiety, unhappiness, emotional control, hostility) were not different between patients with duodenal ulcer and well persons. [344], [345] Furthermore, even if personality traits are overrepresented in patients with PUD (e.g., neuroticism), it must be shown that neuroticism is a consequence of PUD and not a causal factor, and that these traits should be considered a manifestation of coping with the illness. Adami and colleagues [345] assessed the relationship between subject-perceived stress and the development of peptic ulcer in about 4500 adults in the United States. At baseline, 68% perceived themselves as stressed. Over the 13-year follow-up, 208 developed ulcers: 7% of those in the stressed group and 4% of the nonstressed group. The investigators observed that the relative risk increased from 1.4 to 2.9 with increasing degrees of stress. The pathophysiological factors underlying psychological stress and the development of ulcer disease warrant further investigation.

Other Factors Associated with Peptic Ulcer Disease

Alcohol. The data supporting a role for alcohol in the pathogenesis of PUD are quite limited. Alcohol is an agent commonly used to induce gastric mucosal damage in experimental animals, [347] but in these models, alcohol is often administered as absolute ethanol (i.e., pure ethanol, or 200 proof), which is hypertonic and lipid soluble and traverses the gastric mucosa promptly, resulting in frank acute mucosal damage. [347] Therefore, the practical implications of this model are nonexistent. In fact, mucosal injury does not occur at all in humans at ethanol concentrations of less than 10% (20 proof). [348] At concentrations of 20% (40 proof) and greater, the transmucosal gastric potential difference decreases, and endoscopic evidence of acute injury is observed. Because of prompt epithelial restitution, it is unlikely that these acute and transient mucosal changes result in chronic mucosal damage. Retrospective analysis of alcohol intake in patients with ulcer disease revealed that those who consumed modest amounts of alcohol had a decreased prevalence of ulcer disease compared with nondrinkers. [349]

Diet. Despite the observation that some types of foods and beverages are reported to cause dyspepsia, there are no convincing data indicating that any specific diet causes ulcers. Highly spiced foods are often reported to result in indigestion or dyspepsia by unknown mechanisms. Duodenal ulcer is more common in the south of India, an area of rice-eating, low-fiber diets, than in the northern part, a wheat-eating area; [350] again, the cause is unknown. Both decaffeinated and caffeinated beverages (e.g., tea, coffee) are potent stimulants of gastric acid secretion, [351], [352] as are cola-type beverages, beer, and milk. [351] However, epidemiologic studies have failed to reveal an increased risk of ulcer disease in persons who consume these beverages. Although bland meals and the frequent ingestion of milk were commonplace in ulcer treatment in the early 1900s, they neither expedite ulcer healing nor prevent recurrences or complications. In fact, they stimulate greater amounts of gastric acid secretion than three regular daily meals. Dietary alterations, other than instructions to avoid pain-causing foods, are unnecessary for patients with ulcers.

Miscellaneous factors. Peptic ulcer appears to have some seasonal variations; for example, duodenal ulcers occur more frequently in January and February than during the rest of the year. [353] Regional differences exist: ulcer perforation is greater in Scotland than in England, and the incidence of duodenal ulcer is significantly less in northern India than in southern India. [10] These geographic differences are not fully understood. PUD, especially gastric ulcer, seems to be somewhat more common in persons of lower economic status, [354] possibly related to a higher incidence of *H pylori* in this population. The mortality rate for ulcer disease is greater in nonwhites younger than 65 years of age. The differences, however, are minor in elderly persons, in whom the rates for races tend to equalize.

CLINICAL MANIFESTATIONS

Abdominal Pain

Abdominal pain is the most common symptom, leading a patient to seek medical attention for a perceived peptic ulcer. On careful questioning, most patients with PUD report abdominal pain, but epigastric discomfort is more commonly associated with diseases other than an ulcer. [353] Esophageal, cardiac, biliary, pancreatic, and intestinal disorders may cause epigastric distress and often produce pain that is localized to the epigastrium, mimicking ulcer pain. The sensitivity (i.e., symptom-positive and disease-positive) and the specificity (i.e., symptom-negative and disease-negative) of abdominal pain as a marker for ulcer disease are low. In a large survey of patients in a dyspepsia clinic, abdominal pain occurred in 94% of patients with ulcers, but it was also present in 82% of the larger group of patients without ulcer disease. [354] The diagnosis and management of nonulcer dyspepsia are discussed in Chapter 30.

Abdominal pain in ulcer disease is classically described as having a burning quality, localized to the epigastrium, nonradiating, occurring 2 to 3 hours after a meal or at night, and relieved by food or antacids. The most discriminating symptom is the presence of pain that awakens the patient from sleep (especially between 12 and 3 AM), [355] which is present in approximately two thirds of patients with duodenal ulcer and one third of patients with gastric ulcers. Unfortunately, it is also present in about one third of patients with nonulcer dyspepsia, which is at least twice as common as ulcer disease. [81] In the United States, fewer than 30% of patients with dyspepsia who undergo upper GI endoscopic examination have ulcer disease. [356] However, of those with classic ulcer dyspepsia, 40% have an ulcer crater, and 40% have endoscopic gastroduodenitis without an ulcer crater. This observation suggests that careful questioning may slightly improve diagnostic accuracy. [356] Although abdominal pain is the major presenting symptom of an ulcer, it is not possible to diagnose this entity with any significant degree of accuracy based on symptoms alone. However, some clinical clues may suggest ulcer disease: prior documented ulcer, nocturnal pain, family history of ulcer, and the intake of antacids or antiulcer drugs that result in pain relief. [354]

Approximately 10% of patients with PUD, particularly those with NSAID-associated ulcers, present with complications (e.g., hemorrhage, perforation) without an antecedent history of ulcer pain. Either they are insensitive to pain or ulcers can be present without producing pain.

Mechanisms of Abdominal Pain The genesis of abdominal pain in patients with ulcer disease is undefined. In a blinded, prospective study, no consistent association was found between pH of gastric infusate and production of pain in patients with active duodenal ulcer. [357] In an unblinded study, direct duodenal infusions of hydrochloric acid tended to induce pain in patients with duodenitis and duodenal ulcer. [358] Infusion of acid caused pain in only 40% of patients with active symptomatic duodenal ulcers, but a placebo-saline infusion resulted in ulcer-type pain in 10%. Other factors, such as pepsin, bile acids, and motility events, possibly could contribute to ulcer-type pain.

Pain Relief Pain of any type is very responsive to placebo therapy. In all clinical trials of therapy for peptic ulcer, abdominal pain resolved promptly in patients treated with placebo or an active drug. [359] Nonetheless, most studies suggest that active treatment with antacids, H_2-receptor antagonists, or omeprazole is somewhat superior to placebo. Furthermore, the proportion of patients (~30%) who have nonhealed ulcers and are symptom free equals the proportion who have healed ulcers but who continue to have ulcer symptoms. [356], [360]

Ulcer Complications

Complications of PUD include hemorrhage, perforation, penetration, and obstruction. Intractability, previously considered a complication of ulcer disease, has become exceedingly rare subsequent to the development of more effective antiulcer drugs (e.g., H^+,K^+-ATPase inhibitors).

Hemorrhage *Hemorrhage,* the most common complication of ulcer disease, occurs in approximately 15% of patients. [361] It tends to be more common in patients during the sixth decade of life and beyond, largely because of the increased use of NSAIDs in this age group. [362], [363] Approximately 10% to 20% of patients bleed from a gastric or duodenal ulcer without any antecedent symptoms. Approximately one third of patients treated with H_2-receptor antagonists have recurrent hemorrhage. [361] Studies suggest that eradication of *H pylori* prevents duodenal ulcer and non-NSAID gastric ulcer recurrences and may prevent recurrent hemorrhage. The diagnosis and management of GI hemorrhage are discussed in Chapter 33, Chapter 34, Chapter 70, and Chapter 148.

Perforation Ulcer-related *perforation* is less common than hemorrhage but more common than obstruction, occurring in approximately 7% of patients with PUD.

Duodenal ulcers tend to perforate anteriorly, and gastric ulcers tend to perforate along the anterior wall of the lesser curvature of the stomach. With the increased use of NSAIDs, the incidence of perforation is increasing, particularly in women more than 60 years of age.
Penetration *Penetration* is similar pathologically to perforation, except the ulcer crater burrows through the entire wall of the intestine, and instead of leaking digestive contents into the peritoneal cavity, the crater bores into an adjacent organ. Gastric ulcers most commonly penetrate into the left lobe of the liver, whereas duodenal ulcers penetrate posteriorly into the adjacent pancreas, sometimes leading to pancreatitis. Rarely, gastric ulcers may penetrate into the colon, resulting in a gastrocolic fistula.
Gastric Outlet Obstruction *Gastric outlet obstruction,* also referred to as gastric retention or pyloric stenosis, can result from functional impairment of antral motility caused by the effects of acute inflammation and edema or from mechanical obstruction caused by scarring near the gastroduodenal junction. The former tends to resolve with medical treatment, and the latter usually requires surgical or endoscopic (i.e., balloon dilation) intervention. The prevalence of gastric retention is approximately 2% in patients with PUD. The symptoms of gastric outlet obstruction tend to be insidious and can manifest as gastroesophageal reflux, early satiety, weight loss, abdominal pain, and vomiting. As the degree of retention increases, the quantity of vomitus also increases, often containing food ingested 12 or more hours previously.

Differential Diagnosis

Nonulcer Dyspepsia *Nonulcer dyspepsia,* which is abdominal pain or discomfort that occurs in the upper abdomen without the presence of an ulcer, is a major complaint that affects about 15% of the population annually, most of whom do not seek medical care. [364], [365], [366], [367], [368] and [369] In the United States, more than 2 million people seek outpatient evaluations each year for dyspepsia. For a detailed review of this entity, see Chapter 30.
Other Gastrointestinal Diseases GI diseases that mimic PUD include upper GI neoplasms (i.e., gastric and pancreatic cancer), mesenteric ischemia, which is sometimes referred to as abdominal angina, and pancreatitis or cholecystitis. Gastric or duodenal involvement with Crohn's disease or with typical and atypical strains of *Mycobacterium tuberculosis* can produce gastric retention and ulcer-type symptoms.

DIAGNOSTIC STUDIES

History and physical examination are not reliable tools for establishing a diagnosis of PUD; documentation of true ulcer disease depends on the use of radiographic or endoscopic methods. The high cost of endoscopic and radiographic procedures have led to attempts at developing a surrogate marker for ulcer diagnosis, such as the response to an empiric trial with antisecretory agents or the evaluation of *H pylori* status with serologic markers. The following section reviews the different diagnostic modalities that assist the clinician in assessing a patient with signs or symptoms of PUD.

Treat-First Strategies

A major tenet of the practice of medicine—establishing the precise diagnosis so specific therapy can be prescribed—has been questioned for patients with dyspepsia. [370], [371] Most physicians prescribe highly effective antiulcer drugs without further investigation, and it is unusual in today's environment to encounter a patient with untreated dyspepsia. This approach has been endorsed in a policy statement by the American College of Physicians. [370] This policy of "treat first" is appropriate for the patient younger than 45 to 50 years of age with mild and intermittent epigastric symptoms and no other systemic symptoms or evidence of ulcer complications. However, the therapeutic response should be evaluated to ensure prompt symptomatic relief and the absence of recurrence after therapy has been discontinued. For patients older than approximately 50 years of age, those in whom symptoms have been of long duration, those with systemic symptoms (e.g., anorexia, weight loss, back pain), or those in whom there is any suggestion of an occult malignant disease or another disease process (e.g., pancreatic or biliary tract disease), a specific diagnosis should be established.

Endoscopy and Radiography

The diagnostic procedure to be used depends, in large part, on the skill and experience of available personnel. In a center with superb GI radiography, this may be a reasonable first course to pursue. However, because of its superior sensitivity and specificity for significant organic disease and the ability to obtain biopsies, endoscopy is the preferred diagnostic test in most cases. There is no justification for the routine use of one procedure followed by the other. When a lesion such as a gastric ulcer has been observed radiographically, however, endoscopy with biopsy may be warranted. Conversely, there can be endoscopic lesions, such as extrinsic displacement of the stomach or duodenum, that require radiographic studies. Therefore, these two procedures may be complementary in specific circumstances.

Traditionally, endoscopy with biopsies was considered the standard of practice in patients with gastric ulcer. However, with the increased intake of NSAIDs, the finding of a clearly benign-appearing gastric ulcer on the radiograph in a 40-year-old patient with a history of recent NSAID intake does not necessarily warrant endoscopic examination with biopsies. However, all gastric ulcers should be observed until complete endoscopic healing because of the potential of an occult malignant disease. [372], [373]

Cost comparisons are complicated. Overall, the acquisition cost for routine upper GI endoscopy is approximately three to four times greater than the cost for upper GI radiography. Gastric biopsies for *H pylori* add further endoscopic and laboratory costs. Some endoscopists consider current professional fees to be excessive, and this has led to the proposal of a "screening endoscopy" at a cost similar to that of radiography. [374] Regrettably, this has not become generally accepted. It is likely that, with the changes in health care delivery, the costs for endoscopy will decrease substantially. Additional cost-benefit studies are needed in patients with acid peptic symptoms. The goals of any diagnostic test are to be sensitive and specific and to have high diagnostic accuracy. Although studies comparing air-contrast barium meals performed by expert GI radiologists with endoscopy reveal equal accuracy in the diagnosis of an ulcer, [375] extrapolation of these studies to the clinical practice is not warranted because the radiography and endoscopy studies were performed by experts. Such reports are often biased in favor of endoscopy because this test is commonly used as the standard. For example, lesions found on x-ray films but not observed endoscopically are assumed to be radiographic false-positive results, although they may represent endoscopic false-negative results (i.e., the ulcer was missed at endoscopy). With modern wide-angle endoscopic television chip and endoscopic instruments, visualization of the stomach and duodenum should be complete in almost all patients. Nonetheless, as many as 10% of duodenal ulcers have been missed by endoscopy. [376], [377] Postbulbar duodenal ulcer disease may be missed by the endoscopist, but these lesions can be seen with the use of longer endoscopes. Patient risk with endoscopy, although minimal, is greater than that associated with radiography.

Although there is generally good agreement between the diagnostic results of endoscopy and those of a double-contrast barium meal for gastric ulcers when each test is performed by skilled personnel, there are fewer radiologists skilled at contrast studies, and radiography does not permit biopsies or cytologic brushings of the lesion to determine whether a gastric ulcer represents an ulcerating carcinoma. [378], [379] Therefore, radiography has largely become obsolete in the evaluation of gastric ulcers. This modality is most useful for examining gastric and duodenal distensibility or for the presence of extrinsic compression.

Diagnosis of *Helicobacter pylori*

Endoscopic Tests From a pragmatic standpoint, the first decision regarding the choice of diagnostic testing for *H pylori* revolves around the need for endoscopy. It is also important to consider whether a test will be performed before (pretreatment test) or after the completion of treatment (posttreatment test). At the time of endoscopy, specific findings, such as a duodenal ulcer, are highly predictive of *H pylori* infection. Antral nodularity is also a specific, but insensitive, marker of *H pylori* infection. The critical role of endoscopy for the diagnosis of *H pylori* therefore is to obtain gastric mucosal biopsies that are used for both *direct tests* (culture and histology) and *indirect tests* (urease testing). The biopsy-based tests are dependent on the actual bacterial load and identify only patients with active *H pylori* infection. Medications that affect the density or viability of *H pylori* organisms within the stomach decrease the sensitivity of these tests by increasing the possibility of sampling error. For this reason, bismuth compounds and antibiotics should be withheld for 4 weeks, and PPIs should be withheld for 1 to 2 weeks before endoscopic *H pylori* testing.
Culture. At the current time, the importance of culture, a direct test, is more historic than practical. Although some laboratories achieve sensitivities of more than 95% for culture, other methods for the diagnosis of *H pylori* infection are less costly, simpler, and prone to less variability or error, and they provide results more rapidly. Culture, however, has the advantage of allowing for the determination of antibiotic sensitivity. Culture may also be of importance in research. Saline is a simple acceptable short-term (<6 hours) transport medium, whereas refrigerated Stuart medium or glycerol-containing media will be more appropriate if culture will be delayed. [380], [381] Glycerol-containing media are also suitable for long-term storage of biopsy specimens at -70°C. [381] Various selective and nonselective media are suitable for culture, such as brain-heart infusion agar plates with other supplements and media used for *Campylobacter.* Attempted culture on more than one medium may be helpful to increase sensitivity. In general, culture incubation is performed under microaerophilic conditions at 37°C, with positive cultures usually detected after 3 to 5 days. Identification of *H pylori* is made on the basis of colony morphology (approximately 3-mm translucent colonies) that contain gram-negative, curved rods that test positive for urease, catalase, and oxidase.

Histological assessment. The standard for the diagnosis of *H pylori* is the detection of the organism on gastric biopsies that have been processed using a variety of histological stains (see Chapter 68). The distribution of *H pylori* in the stomach is not uniform, nor are organisms usually found in areas of intestinal metaplasia. [382] Therefore, normal gastric mucosa should be sampled to minimize sampling error, and to help with interpretation of the specimen, jumbo biopsy forceps are recommended, although the effect of biopsy size on the sensitivity of histology has not been studied. Multiple biopsies should be obtained to achieve close to 100% sensitivity and specificity. The use of special stains is outlined in Chapter 68.

Rapid urease tests. Developed by Marshall, the CLOtest (Ballard Medical Products, Draper, UT) was the first of the commercially available biopsy urease tests designed specifically for *H pylori* detection. It is an indirect test for the presence of *H pylori. H pylori* urease hydrolyzes the urea contained in the agar gel of the test packet and leads to production of ammonia, a pH rise, and a color change of the phenol red indicator. The test is interpreted up to 24 hours after insertion of the gastric biopsy sample into the well containing the agar gel. The overall sensitivity of the test in the pretreatment setting is approximately 90%. However, sensitivity seems to vary from one study to another, even within the same institution, [383], [384] because of the use of different standards for the diagnosis of *H pylori,* the subjective nature of test interpretation, different biopsy sizes or number in different studies, or different protocols employed in the incubation of the biopsy. Two studies found that the sensitivity of the CLOtest at 24 hours was equivalent for different biopsy sizes, [384], [385] but in one of the studies, the CLOtest became positive more quickly with a larger biopsy size. Although warming also hastened the time to a positive result, no significant difference at 24 hours was noted between warmed and room temperature testing. [383] In addition, the sensitivity of the CLOtest is not affected by preimmersion of biopsy forceps in formalin. [386] Specificity of the CLOtest is uniformly excellent (95% to 100%), but when read at 24 hours, false-positive results may be infrequently encountered. Another gel test, Hpfast (GI Supply, Comphill, PA), is similar to the CLOtest, but it uses a different pH indicator. Hpfast also is interpreted up to 24 hours. PyloriTek (Horizons International Corp., Ponce, Puerto Rico) is a strip test. In the presence of urease, ammonia is produced from urea impregnated into a reaction strip. An overlying pH indicator detects the diffusion of ammonia through a membrane. A potential advantage of this test is that interpretation may be performed only 1 hour after tissue inoculation. Simultaneous studies of the three tests have been performed, and the results have been comparable. [387], [388] Another rapid urease test that provides an answer in 1 hour and requires no incubation or reagents (Pronto Dry [Endo-Surg Medical Inc., Columbia, TN]) is now available. In addition, it is anticipated that faster versions of the CLOtest and Hpfast will become available in the near future. As such, the choice of test should be based on factors such as cost, availability, or physician preferences. Rapid urease tests have been reported by some to be falsely negative in the setting of ulcer bleeding. [389], [390] and [391] Studies suggest that a mixture of blood and gastric and duodenal contents including bile may render the test falsely negative. [392] Conversely, false-positive results do not occur with increased frequency in the setting of acute ulcer bleeding. To maximize the yield of biopsy-based testing in patients with acute ulcer bleeding, the clinician should obtain tissue samples from the gastric body and antrum for rapid urease testing and histology. Some have advocated banking biopsies in formalin for histology and sending them for assessment only if the rapid urease test is negative. Unfortunately, this practice is difficult to implement in the clinical setting. An alternative strategy is to perform a rapid urease test and, if negative, to obtain a nonendoscopic test, such as serology, urea breath test, or stool antigen, to confirm the presence or absence of infection.

Polymerase chain reaction. PCR assays, which have been shown to be sensitive and specific, have been developed for the detection of *H pylori* in gastric mucosal biopsies. However, the diverse genetic organization of *H pylori* may affect the sensitivity of the assay. Uncertainty also exists about the specificity of some of the primer molecules used in PCR. Moreover, specimen preparation and conditions during PCR are not necessarily standardized among different laboratories, and successful detection of *H pylori* may require an uncertain minimum bacterial load. Currently, PCR assays should be restricted to the research setting for identification of different *H pylori* strains.

Nonendoscopic Tests

Antibody test. Many commercially available serologic tests that detect the presence of anti– *H pylori* IgG antibodies have received FDA approval, which ensures some likelihood of quality control and more uniform results across a spectrum of patients from different regions and demographic backgrounds of the United States. Despite FDA approval, however, the performance characteristics of the tests may vary widely. [393] Availability, cost, and ease of use may influence the choice of test. In general, in the United States, only FDA-approved tests detecting anti– *H pylori* IgG antibody should be used. Unfortunately, because there is no widely agreed on minimum standard for performance, even FDA-approved serologic tests are not necessarily consistently highly accurate. Individual reports of serologic antibody tests may suggest accuracies of approximately 95%, but these results may be misleading. Indeterminate results, the gray zone, may be excluded, or considered positive or negative at the discretion of investigators. [394] A metaanalysis of studies of 11 commercial enzyme-linked immunoadsorbent assay kits and one latex agglutination kit found an average sensitivity of 85% and specificity of 79%. [395] Thus, antibody tests both underdiagnose (false-negative results) and overdiagnose (false-positive results) *H pylori* infection with some frequency. In addition, the pretest probability of infection should be considered when using antibody tests as a screening tool. Patients with a high probability of having *H pylori* infection (e.g., patients with PUD) who have negative serologic tests deserve further evaluation. If the likelihood of infection is low, the predictive value of a positive test is low, and the test may need confirmation. Conversely, a negative test predicts a low probability of infection in patients who have a low likelihood of infection, and further evaluation for infection is unnecessary. Despite the foregoing commentary, antibody testing offers numerous advantages: it is noninvasive, relatively inexpensive, and avoids some of the pitfalls inherent to methods that identify patients with active infection such as the urea breath test or stool antigen test. The active tests depend on and reflect the current bacterial load and may be falsely negative because of temporary suppression of *H pylori* infection by ingested bismuth compounds, PPIs, or antibiotics. These medications do not cause false-negative serologic test results. However, there may be a small, transient fall in antibody titer even after failed antibiotic treatment of *H pylori,* presumably reflecting the decreased bacterial load. The fall in titer, however, is insufficient to lead to a negative result. In addition to the previously described limitations of antibody testing, patients with gastric atrophy, but no ulcers, may have false-positive results because of serologic evidence of prior *H pylori* infection that has spontaneously resolved. [396] Conversely, the serologic test may be more sensitive than the standards against which it is being evaluated, especially if the bacterial load is small. Although this may be true on occasion, [397] this seems unlikely in most discrepant cases, especially in studies including a large battery of different diagnostic tests. In addition, a few patients with *H pylori* infection who are treated with antibiotics for unrelated indications (e.g., bronchitis, pharyngitis, urinary tract infection) will experience successful cure of their infection. For example, clarithromycin used alone can lead to cure of *H pylori* infection in as many as 30% to 40%. Such patients will have a true-positive antibody test result (the test detects *H pylori* antibody) but no active *H pylori* infection. Given the widespread and often indiscriminate use of antibiotics, this phenomenon is likely to erode the utility of antibody testing further, particularly in countries where the prevalence of active *H pylori* infection is falling or is already low. [398] In addition to laboratory antibody tests, rapid qualitative antibody tests (in-office or near-patient tests) using either serum or fingerstick whole blood are commercially available. They are inexpensive (<$15), results are available in 5 to 15 minutes, and they are simple to use. Unfortunately, published reports suggest that the current generation of whole-blood tests may be less accurate than some laboratory-based tests. [399], [400] It is possible that newer whole-blood tests will offer greater accuracy than the currently available tests. [401] The office-based tests are only qualitative, precluding their use for early follow-up testing for assessment of cure after anti– *H pylori* treatment. Other antibody tests using saliva have not proven to be as accurate as those using serum or whole blood. [402] Numerous urine antibody tests for *H pylori* have been reported and may yield results comparable to those of traditional serologic testing. [403] At this time, saliva- and urine-based tests are not commercially available in the United States.

Breath and stool tests

Nonendoscopic urease tests. The ^{13}C and ^{14}C urea breath and blood tests identify only actively infected patients through *H pylori's* urease activity. Patients ingest ^{13}C- or ^{14}C-labeled urea. If *H pylori* is present in the stomach, urease hydrolyzes the labeled urea and releases labeled HCO_3^-, which is transported by the bloodstream to the lungs and is exhaled as labeled carbon dioxide. Breath or blood is collected, and either the radioactive ^{14}C isotope is detected using a scintillation counter or mass spectrometry or infrared spectroscopy is used to detect nonradioactive ^{13}C. [404], [405] Because oral organisms also hydrolyze urea, an early peak in labeled carbon dioxide may occur during the ^{13}C breath test because the urea is presented in liquid form. However, in practice, this is inconsequential, because breath samples are collected 15 to 30 minutes after urea ingestion. The ^{14}C test uses a capsule containing labeled urea. Because the solid form of the label minimizes any possibility of hydrolysis by oral bacteria, breath is collected after only 10 minutes. The normal background concentration of ^{13}C necessitates collection of a baseline breath sample to which subsequent samples are compared. In contrast, because there is almost no background concentration of ^{14}C, there is no need for a basal breath sample, and a very small dose of radioactive ^{14}C urea need be used. [406] Overall, the performance characteristics of both the ^{13}C and ^{14}C tests are similar. False-negative results can occur in patients taking certain drugs, such as PPIs, bismuth compounds, or antibiotics, that decrease the density of *H pylori* organisms or its metabolic activity. [407] There is also some evidence to suggest that H_2-receptor antagonists can rarely induce false-negative results of nonendoscopic urease tests. [408] For nonendoscopic urease tests to be reliable, acid-suppressive medications should be withheld for 1 to 2 weeks before testing. [409], [410] The minimum time interval required for a reliable breath test after the last dose of an incidentally prescribed bismuth compound or antibiotics has not been studied, but a period of 2 to 4 weeks appears to be adequate. [411] The choice of test (^{13}C versus ^{14}C) depends on availability, cost, and regulatory issues. The ^{13}C test is preferred for children and pregnant women. The ^{13}C and ^{14}C urea breath tests and ^{13}C urea blood test have been approved by the FDA.

Fecal antigen test. A test that identifies *H pylori* antigens in stool has become commercially available. The fecal antigen test uses polyclonal anti– *H pylori* capture antibody adsorbed to microwells. Diluted stool and a peroxidase conjugated polyclonal antibody are added, followed by substrate 1 hour later. In infected patients, enzyme-substrate binding leads to a color change, which can be detected visually or spectrophotometrically. After collection, stool samples can be stored at 2 to 8°C for 3 days and at -20°C indefinitely. Studies have reported sensitivity and specificity for the fecal antigen test of more than 90% in patients not previously treated for *H pylori* infection. [412], [413] Like the nonendoscopic urease tests, the stool antigen test identifies patients with active *H pylori* infection. Preliminary studies suggest that the stool antigen test may be useful as a means of establishing cure after antimicrobial therapy. [414] The sensitivity of the stool test is decreased by the recent use of antibiotics, bismuth, or PPIs, although to a lesser degree than the nonendoscopic urease tests. [415] The fecal antigen test is more accurate than antibody testing and is less costly than the nonendoscopic urease tests. Issues that have slowed the widespread use of this test include the inherent unpleasantness associated with the handling and storing of stool, limited availability, and highly variable state-to-state reimbursement. The fecal antigen test has been approved by the FDA.

Use of *Helicobacter pylori* Test for Initial Diagnosis

Should Helicobacter pylori status be determined in all patients with duodenal ulcer? Early reports indicated that *H pylori* could be identified in approximately 90% of patients with duodenal ulcers and in 70% of patients with gastric ulcers. However, studies in the United States suggest that the prevalence of *H pylori* infection in patients with ulcer disease may be 60%. [416], [417] Moreover, the prevalence of *H pylori* in bleeding ulcers is known to be lower than in uncomplicated ulcers. Because a substantial minority of patients (=40%) with ulcer disease do not have *H pylori* infection, testing to establish the presence of infection should be performed in all patients with an ulcer before the initiation of antibiotic therapy. The method of assessment of *H pylori* status depends on the clinical circumstances. If endoscopy is performed, biopsy-based methods can be used to establish the presence of *H pylori.* Because of a reduced sensitivity of rapid urease test in the setting of acute upper GI bleeding, [418] both rapid urease testing and histology should be performed in the setting of acute upper GI bleeding. If at the time of endoscopy, biopsies for *H pylori* were not obtained from a patient found to have a duodenal ulcer, or if diagnosis of a duodenal ulcer was made by radiographic studies or on the basis of a prior history of a documented duodenal ulcer, nonendoscopic tests are appropriate. Given the high pretest probability of *H pylori* infection in patients with ulcer disease, antibody testing is a reasonable diagnostic choice. The nonendoscopic urease tests and the fecal antigen test are also excellent options.

Should Helicobacter pylori be determined in patients taking nonsteroidal antiinflammatory drugs? If NSAIDs and *H pylori* together have additive deleterious effects on gastroduodenal injury, it would be appropriate to test and treat for *H pylori* before beginning NSAID treatment. Although some evidence favors this paradigm, [419] most of the data currently available do not support this approach. At this time, we do not recommend determining the *H pylori* status of patients who are taking NSAIDs unless these patients have a history or concurrent evidence of PUD. *H pylori*–infected patients in this latter category should be treated with anti– *H pylori* therapy.

Should Helicobacter pylori status be determined in all patients presenting with dyspepsia? Management options for patients with uncomplicated dyspepsia (in which patients do not have alarm features, such as vomiting, weight loss, bleeding, or anemia) include the "test-and-treat" strategy for *H pylori* and empiric antisecretory therapy. Several organizations, including the American Gastroenterological Association, [420] have supported the test-and-treat strategy, and large, randomized, controlled trials from Europe suggest that this strategy reduces endoscopic workload without compromising clinical outcomes in patients with uncomplicated, uninvestigated dyspepsia. [421], [422] Patients may be less satisfied with the test-and-treat strategy compared with early endoscopy, [421] a finding supporting the notion that patients are reassured by the more definitive diagnosis provided by endoscopy. In a study from the United States, clinical and economic outcomes of patients with a positive qualitative antibody test who were treated for *H pylori* compared with those with a negative antibody test who were treated with a PPI were similar. [423] Based on a large number of randomized trials, it appears that eradication of *H pylori* offers little, if any, benefit over placebo or a short course of PPI in patients with nonulcer dyspepsia [97], [98] (see Chapter 30). As such, the clinical benefits of the test-and-treat strategy are derived largely from improvement in patients with ulcer disease. Factors influencing the decision between the test-and-treat approach and empiric antisecretory therapy include the background prevalence of *H pylori* and PUD and the percentage of patients whose ulcers are caused by *H pylori.* [424] Populations with a low prevalence of *H pylori* or ulcer disease will derive less benefit from the test-and-treat strategy than populations where the opposite is true. The changing demographics of *H pylori* infection in the United States and many other Western countries will require ongoing reevaluation of the choice between the test-and-treat strategy and empiric antisecretory therapy in patients with uninvestigated dyspepsia.

Use of *Helicobacter pylori* Test for Posttreatment Diagnosis

Indications and timing. In patients with PUD, eradication of *H pylori* significantly reduces ulcer recurrence and rebleeding; therefore, all patients with *H pylori*–related ulcer bleeding, obstruction, or perforation should undergo testing to prove eradication of the pathogen after antibiotic therapy. Whether to pursue testing routinely after treatment of patients with an uncomplicated duodenal ulcer is less clear. Certainly, testing for *H pylori* infection should be pursued in patients who suffer with recurrent or persistent symptoms after antibiotic therapy. It is less clear whether patients with an uncomplicated ulcer who are rendered asymptomatic by *H pylori* therapy should be tested. One could argue that there is a risk of morbidity and even mortality associated with ulcer recurrence resulting from persistent *H pylori* infection; however, this risk is likely to be small, and the cost-effectiveness of posttherapy testing in patients with uncomplicated ulcer disease therefore remains controversial. Most patients with gastric ulcers undergo follow-up endoscopic evaluation, and at that time it is reasonable to test for cure of *H pylori* infection using biopsy methods. In general, evaluating for eradication of *H pylori* should be done with the same endoscopic and nonendoscopic tests used to identify active infection. Testing should be performed no less than 4 weeks after the completion of therapy, because up to 2 weeks after treatment that ultimately is shown to have failed, it may not be possible to detect *H pylori* as a result of temporary suppression or reduction of the bacterial load. This phenomenon, which has been termed *clearance,* can be appreciated only in retrospect, after recrudescence occurs. *Cure* (or eradication) is used to describe nondetection of organisms 4 or more weeks after treatment.

Endoscopic tests after treatment. Very limited data exist regarding the accuracy of biopsy-based methods for the detection of *H pylori* in the posttreatment setting. One study showed that two jumbo biopsies, one from the gastric angle and one from the greater curve of the gastric body, would suffice to detect bacteria in virtually all those patients who remained infected. [425] Interestingly, the least sensitive site was prepyloric, with a false-negative rate of approximately 10%. In the same study, rapid urease tests such as the CLOtest had a sensitivity of 95% based on a single antral biopsy. Because false-negative CLOtests may be associated with low *H pylori* density, [384] it is reasonable to anticipate that rapid urease tests may be less sensitive in the posttreatment setting, and this is supported by data from multicenter treatment trials in the United States. [426] In this study, it was found that biopsies from the antrum and body for histology obtained 8 weeks after therapy yielded a posttreatment sensitivity of 93%, whereas rapid urease testing alone yielded a sensitivity of only 78%. Combining histology and rapid urease testing yielded a posttreatment sensitivity of 98%. [427] Based on these results, we discourage the use of rapid urease testing as a sole means of establishing *H pylori* eradication. Rather, combining histology and rapid urease testing is appropriate when assessing for *H pylori* eradication using endoscopic methods.

Antibody test after treatment. After successful cure of *H pylori* infection, anti– *H pylori* antibody titers invariably fall, allowing quantitative serologic antibody methods to be used for follow-up evaluation of treated patients. A 50% decline in titer is reliably accurate in establishing cure. [428] Conventionally, convalescent and baseline serum samples are tested together, to minimize errors resulting from assay variation. Such variation, however, is minimal, and it is not likely to be consequential when evaluating for titer declines of approximately 50%. Therefore, provided the same assay is to be used on the convalescent sample as was used on the baseline sample, it is probably not essential to store the baseline sample. Of critical importance for determination of posttreatment cure is the timing of the convalescent assay. Distinguishing between treatment success and failure requires a 6-month posttreatment period before approximately 85% of patients will be correctly characterized. [428] Whether nonquantitative office-based tests may be useful for confirmation of cure is not well studied. However, most patients probably remain seropositive; in one report, only 36% of patients became seronegative during follow-up. [429] It is not known whether most patients eventually will become seronegative after cure (or spontaneous elimination) of *H pylori* infection, but this seems unlikely. Because the time frame is erratic for the antibody to decline sufficiently (usually 20% to 50% of the original titer) to assess cure, and it requires at least 6 to 12 months for acceptable sensitivities, antibody assays are impractical in the posttreatment clinical setting.

Nonendoscopic urease breath tests after treatment. In contrast to antibody tests, the nonendoscopic urease tests detect active infection and provide an indirect reflection of the *H pylori* bacterial load present at the time of the test. Intuitively, if the bacterial load has been reduced to zero by successful treatment, the nonendoscopic urease tests should accurately reflect this status and should be negative, as they are in patients who are uninfected in the first instance. Indeed, nonendoscopic urease tests have posttreatment sensitivity and specificity of greater than 90%. [430] As mentioned earlier regarding biopsy tests, it is currently recommended that nonendoscopic urease tests be performed no sooner than 4 weeks after completion of anti– *H pylori* therapy. There are some data to suggest that posttherapy testing can be performed as early as 2 weeks after therapy. [411] However, this finding requires confirmation in large clinical trials before being recommended for routine clinical care.

Fecal antigen test after treatment. The fecal antigen test can also be used to establish cure of *H pylori* after the completion of therapy. Original reports from Europe, using a standard of endoscopy-based methods and a urea breath test, suggested that the fecal antigen test had sensitivity and specificity that exceeded 90% as a test for eradication when done more than 4 weeks after the completion of therapy. [413] Unfortunately, more recent studies using less stringent standards have reported lower sensitivity and specificity of the fecal antigen test when it was done 1 month after the completion of therapy for *H pylori*. [413] Some investigators have suggested that it may be necessary to wait as long as 3 months to achieve acceptable accuracy in the posttherapy setting. At present, the optimal timing of stool testing to establish *H pylori* cure remains unclear.

Specialized Testing

Serum Gastrin Testing The incidence of gastrinoma (i.e., Zollinger-Ellison syndrome) in patients with ulcer disease is exceeding low, estimated at much less than 1% of all patients with ulcers (see Chapter 67). Clinical situations in which gastrin measurement should be considered include the following: patients with ulcer disease and endocrine tumors, especially hyperparathyroidism (possibility of MEN I); patients with ulcers located in the distal duodenum or jejunum; patients with diarrhea or weight loss (i.e., hyperacidity of the small intestine resulting in ulcer plus diarrhea and steatorrhea); patients with refractory ulcer disease; and patients with recurrent ulcer disease despite eradication of *H pylori* or satisfactory compliance with prophylactic therapy.

Gastric Secretory Testing Gastric secretory testing should be considered in patients with reproducible hypergastrinemia and in whom gastrinoma is suspected, in patients with gastrinoma who are receiving medical therapy and who should have gastric secretory testing to ensure adequate therapeutic suppression of gastric acid secretion, and patients with refractory ulcer disease.

THERAPY

History

Treatment of patients with acid peptic disorders before the 1970s relied on antacids, anticholinergics, a bland diet, and bed rest. In the latter part of the 1970s, Peterson and colleagues reported that a large dose of antacids (equivalent to 1000 mmol/d) resulted in healing of approximately 80% of duodenal ulcers after 4 weeks of therapy when compared with placebo. [401] As one would anticipate, large-dose antacid regimens were inconvenient and poorly tolerated by patients. In 1977, the first of four H_2-receptor antagonists, cimetidine, became available in the United States:. The availability of cimetidine, ranitidine, famotidine, and nizatidine, orally administered inhibitors of gastric acid secretion, revolutionized the treatment of acid peptic diseases. All the H_2-receptor antagonists produced ulcer healing rates of 80% to 95% after 6 to 8 weeks of therapy. [431] Their outstanding efficacy, ease of use, and excellent safety profile made them the drugs of choice for the treatment of acute ulcers throughout the 1980s.

More potent inhibition of acid secretion became possible with the advent of the H^+,K^+-ATPase inhibitors, which block the final step in H^+ secretion. [432] These potent PPIs appear to produce slightly more rapid ulcer healing than standard doses of H_2-receptor antagonists, an observation of uncertain clinical significance. Virtually all ulcers that are refractory to standard doses of H_2-receptor antagonists can be healed with PPIs, a dramatic confirmation of the importance of gastric acidity in ulcer pathogenesis. [432]

During this same period, other agents that did not affect acid secretion but appeared to expedite ulcer healing with an efficacy comparable to that of H_2-receptor antagonists were developed. These included sucralfate and bismuth (the latter not approved by the FDA for this purpose), which appear to enhance mucosal defense mechanisms. [433], [434] Despite their efficacy and safety, these agents did not initially achieve the widespread use for ulcer therapy enjoyed by H_2-receptor antagonists. Prostaglandin analogs of the E class, which stimulate gastroduodenal mucosal defense mechanisms and have a modest inhibitory effect on gastric acid secretion, were developed but have not been as effective as H_2-receptor antagonists in therapy of acute ulcers. However, the prostaglandin analog, misoprostol, has demonstrated dramatic efficacy in the prevention of NSAID-induced ulcers and is the only agent approved by the FDA for this purpose. [435]

Recognition of the importance of *H pylori* in the pathogenesis of acid peptic diseases precipitated another revolution in ulcer therapy. Historically, gastric and duodenal ulcer disease were chronic conditions with high recurrence rates. In the absence of continuous maintenance therapy, the endoscopically confirmed ulcer recurrence rates approach 60% to 90% at 1 year. [436] Long-term maintenance therapy with H_2-receptor antagonists or sucralfate reduces the recurrence rate to approximately 30% per year but does not alter the underlying predisposition to ulcer disease. Cessation of maintenance therapy is followed by rapid recurrence of symptomatic or asymptomatic ulcer disease in most patients. [437] Eradication of *H pylori* leads to a dramatic decrease in ulcer recurrence, thus affecting the natural history of this disorder.

Acid-Neutralizing and Acid-Inhibitory Drugs

Antacids Antacids were the mainstay of antiulcer treatment until the advent of H_2-receptor antagonists in the late 1970s, and these nonprescription agents continue to be used liberally by patients for the relief of dyspepsia of all causes. The clinical pharmacology and buffering effects of antacids have been reviewed extensively. [438] Antacids have their greatest benefit when administered 1 hour after meals.

Mechanisms of action. Until recently, antacids were believed to produce their therapeutic effect entirely through their acid-neutralizing action. The use of large doses (1000 mmol/d) of antacid administered in multiple doses in patients with duodenal ulcer was based on the principle of neutralizing secreted gastric acid. However, doses of aluminum-containing antacid as low as 120 mmol/d result in healing that is equivalent to larger doses. [439] The buffering effect of these doses of antacids is modest at best and cannot account for their therapeutic efficacy. Mucosal cytoprotective mechanisms have been postulated to explain the efficacy of low-dose antacids. Aluminum salts are thought to enhance mucosal prostaglandin levels, to bind EGF, to stimulate mucus and HCO_3^- secretion, to preserve microvascular flow, to bind bile acids, and to inhibit pepsin activity. [440], [441], [442] and [443]

Adverse effects. Antacids are well tolerated in patients with normal renal function. Sodium-containing antacids can result in significant sodium retention, which may be of concern in patients with hypertension or fluid overload. Large amounts of calcium carbonate can result in hypercalcemia, metabolic alkalosis, and renal insufficiency (i.e., milk-alkali syndrome). [444] Magnesium has a cathartic effect and can result in diarrhea. Conversely, calcium and aluminum may cause constipation. Many proprietary antacids contain a combination of magnesium and aluminum. In patients with renal insufficiency, magnesium-containing antacids can result in significant hypermagnesemia and should be avoided. Aluminum, like calcium carbonate, can bind with phosphate, leading to hypophosphatemia in some cases, and it is commonly used in chronic renal failure for this purpose. Plasma and urinary aluminum concentrations appear to increase in patients with chronic renal failure who take daily aluminum hydroxide. [445], [446] Aluminum absorption may be enhanced by concomitant citrate buffers (e.g., Shohl solution), which is also commonly used in chronic renal failure. Because of growing concerns that aluminum may cause chronic neurotoxicity in patients with chronic renal failure, aluminum-containing antacids are best avoided when possible. [446] Moreover, aluminum may inhibit the absorption of some drugs such as iron supplements and tetracyclines.

H_2-Receptor Antagonists The four H_2-receptor antagonists approved for clinical use by the FDA are cimetidine, ranitidine, famotidine, and nizatidine. These compounds share an aromatic ring system and a flexible side chain (Fig. 66-11). Cimetidine, like histamine, contains an imidazole ring; whereas ranitidine has a furan ring, and famotidine and nizatidine contain thiazole rings. [431]

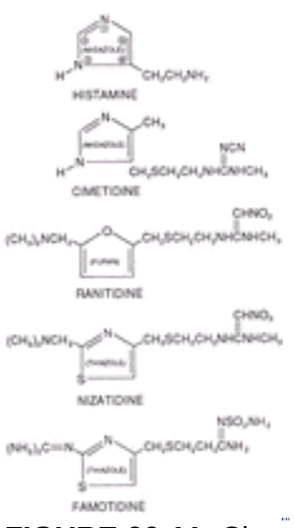

FIGURE 66-11. Chemical structures of histamine and H_2-receptor antagonists. All four antagonists share an aromatic ring system with a flexible side chain attached to a polar uncharged group.

H_2-receptor antagonists exhibit reversible competitive inhibition of histamine-stimulated acid secretion in vivo. [447], [448], [449], [450] and [451] Famotidine has a slower onset of dissociation from the receptor than other agents, suggesting some degree of noncompetitive inhibition; this may account for its longer half-life. [447], [448] H_2-receptor antagonists inhibit basal, histamine-, pentagastrin-, and meal-stimulated acid secretion in a linear, dose-dependent manner. [448], [449] and [450] As much as 90% inhibition of vagal- and gastrin-stimulated acid secretion occurs with H_2-receptor antagonists, reflecting the importance of histamine in potentiating or mediating the effects of cholinergic- and gastrin-stimulated acid secretion. [451] H_2-receptor antagonists lead to near-complete inhibition of nocturnal and basal acid secretion. On the basis of multiple pharmacological studies, famotidine is 20 to 50 times more potent, and ranitidine and nizatidine are 4 to 10 times more potent, than cimetidine on a molar basis (Table 66-7). [431] Increased potency does not necessarily confer a therapeutic advantage, provided the drugs can be administered in equipotent doses without toxicity. Once-daily bedtime dosing regimens of H_2-receptor antagonists are approved for the treatment of acute peptic ulcers. Once-daily morning administration of H_2-receptor antagonists is inferior to nighttime dosing in decreasing 24-hour intragastric acidity. [452]

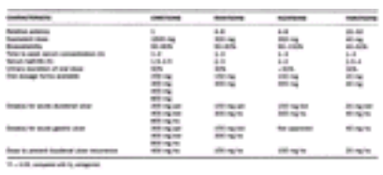

TABLE 66-7 Comparisons of H $_2$-Receptor Antagonists

Pharmacokinetics. All H $_2$ antagonists are absorbed rapidly from the small intestine (see Table 66-7). Absorption is not affected by food but may be decreased by antacids or sucralfate by as much as 30%. Peak blood levels occur within 1 to 3 hours after oral administration. Because of extensive first-pass hepatic metabolism, the bioavailability of oral cimetidine, ranitidine, and famotidine is between 40% and 65%. The bioavailability of nizatidine, which does not undergo first-pass metabolism, is almost 100%. The bioavailability of all intravenous H $_2$-receptor antagonist formulations is close to 100%. All H $_2$-receptor antagonists are well distributed throughout the body. [431], [448], [449], [450] and [451], [453] All agents cross the blood-brain barrier to a limited extent, yielding equivalent ratios of cerebrospinal fluid to serum of 0.07 to 0.20. [454] All H $_2$-receptor antagonists cross the placenta, and although they are believed to be safe, they are not recommended in the first trimester of pregnancy. [455] Cimetidine and ranitidine are concentrated 10- to 20-fold in breast milk; famotidine is not and appears in breast milk in clinically insignificant quantities. Nevertheless, H $_2$-receptor antagonists in general are not recommended in nursing mothers. H $_2$-receptor antagonists are eliminated by a combination of renal excretion (i.e., glomerular filtration and tubular secretion) and hepatic metabolism. With intravenous administration, 60% to 80% of drug is cleared largely unchanged by the kidney, and the remainder is cleared by the liver. After oral administration, 60% to 80% of cimetidine, famotidine, and ranitidine is metabolized by the liver, whereas oral nizatidine is excreted principally by the kidney. [431], [447], [448] and [449], [451] Plasma concentrations of H $_2$-receptor antagonists are affected by renal insufficiency. It usually is recommended that the doses be cut in half at a creatinine clearance of 15 to 30 mL/min for cimetidine and famotidine and less than 50 mL/min for nizatidine and ranitidine. [431] The percentage of H $_2$-receptor antagonist removed by dialysis is small, and replacement doses are unnecessary. Hepatic dysfunction has minimal impact on H $_2$-receptor antagonist pharmacokinetics, and dose adjustment is unnecessary in patients with compensated liver disease with normal renal function. It is unclear whether dose reductions are necessary in decompensated liver disease. [431], [456] The metabolism of H $_2$-receptor antagonists declines with age, and a dose reduction of as much as 50% may be indicated in the geriatric population. Unfortunately, the relation among serum drug levels, acid suppression, and ulcer healing has not been established in this population. [431]

Adverse effects. The H $_2$-receptor antagonists are well tolerated and have a remarkably low incidence of side effects (<3%). [448], [450], [457] In a metaanalysis of randomized clinical trials, the overall rate of untoward effects was no different between patients treated with H $_2$-receptor antagonists and those receiving placebo. [458] Risk factors for adverse effects include multiple medical illnesses, hepatic or renal disease, or advanced age. Much has been made in the clinical literature regarding the relative frequencies of adverse effects among the different H $_2$-receptor antagonists. Cimetidine, as the first H $_2$-receptor antagonist in clinical use, has undergone the most extensive postmarketing surveillance; the other H $_2$-receptor antagonists have had more limited surveillance. [459] With the exception of the increased potential for drug interactions associated with cimetidine, there is no evidence that any one H $_2$-receptor antagonist has a distinctly superior safety profile. In the only large study available, the adverse effects of cimetidine and those of ranitidine were found to be virtually identical. [460] Cimetidine exhibits weak antiandrogenic activity that may result in gynecomastia and impotence, but not the other H $_2$ antagonists effects. They occur in only 0.2% of patients receiving long-term cimetidine therapy at conventional doses, but they have been reported in as many as 44% of patients treated with high-dose therapy for hypersecretory conditions. [461], [462] The changes are readily reversed after discontinuing cimetidine or substituting another H $_2$-receptor antagonist. Various central nervous system (CNS) reactions have been reported with all H $_2$-receptor antagonists, including headache, lethargy, dizziness, depression, memory impairment, agitation, confusion, psychosis, and hallucinations. [463], [464] The incidence of CNS reactions in the outpatient setting is exceedingly low and is not clearly different from controls. [454], [458], [459] and [460] The prevalence of CNS side effects attributable to intravenous H $_2$-receptor antagonists is much more difficult to assess, [454] but trials suggest that the prevalence of CNS reactions definitely attributable to intravenous H $_2$-receptor antagonists is less than 1%. [454], [465] Toxicity does not correlate well with the H $_2$-receptor antagonist dose or serum concentration. [454] Although advanced age and severe hepatic or renal disease are commonly thought to be risk factors for CNS toxicity for these drugs, this has not been convincingly demonstrated. Although cimetidine has been most frequently associated with CNS reactions, there is no evidence that any one H $_2$-receptor antagonist has a higher rate of adverse reactions than another. Leukopenia, neutropenia, anemia, thrombocytopenia, and pancytopenia have been reported with H $_2$-receptor antagonists, with a prevalence ranging from 0.01% to 0.7%. [466] H $_2$-receptor antagonists have been associated with myelosuppression in as many as 5% of bone marrow transplant patients and should probably be avoided in this setting. [467] Reversible asymptomatic elevations of hepatic aminotransferase two to three times the normal level may occur, especially with intravenous therapy. [359], [456] Rare cases of acute hepatitis are generally mild and reversible, and no deaths have been reported. Case control studies and postmarketing surveillance studies do not detect an increased rate of gastric cancer or an increased mortality rate. [468], [469]

Drug interactions and metabolism. Drugs that require an acidic milieu for gastric absorption may have decreased uptake in the more alkaline milieu produced by H $_2$-receptor antagonists, but with the exception of ketoconazole, the impact of H $_2$-receptor antagonists on drug absorption is rarely clinically significant. [470] Cimetidine and, to a lesser extent, ranitidine bind to the hepatic cytochrome P450 microsomal mixed-function oxidase system, dose dependently inhibiting phase I oxidation and dealkylation. Phase II glucuronidation and sulfation reactions are not affected by these agents. [456], [470] Cimetidine is reported to interact with the metabolism of several medications that have a narrow therapeutic/toxic level ratio, including theophylline, phenytoin, lidocaine, quinidine, and warfarin. The metabolism of a variety of other P450-handled medications is also impaired by cimetidine. This may occasionally be significant for some ß-blockers (e.g., propranolol, metoprolol), calcium channel blockers (except diltiazem), tricyclic antidepressants, and benzodiazepines. [471], [472] It is more convenient and perhaps safer to use another H $_2$-receptor antagonist with less interactive potential. Ranitidine binds five- to tenfold less avidly to the P450 system than cimetidine and has far less potential for significant adverse drug interactions. Famotidine and nizatidine do not demonstrate significant avidity for the P450 system and are virtually devoid of significant related drug interactions. [471], [473] All H $_2$-receptor antagonists are secreted at the renal tubule, where they compete with the tubular secretion of creatinine and certain medications. With normal renal function, a clinically insignificant rise (15%) in serum creatinine may occur, but the actual glomerular filtration rate is unchanged. Cimetidine and ranitidine, but not famotidine, inhibit renal tubular secretion of procainamide by as much as 44%. [431]

H $^+$, K $^+$-ATPase Inhibitors

Structure and site of action. As reviewed in Chapter 13, the parietal cell H $^+$,K $^+$-ATPase is the key enzyme responsible for generating hydrochloric acid in the stomach. Its recognition as essential in the final step of acid secretion has made this enzyme a target for a category of drugs that block its activity, PPIs. Some PPIs with a common structural motif have been developed and include omeprazole, esomeprazole, lansoprazole, pantoprazole, and rabeprazole. [47], [474], [475], [476], [477], [478], [479], [480], [481] and [482] These compounds are substituted benzimidazoles, which vary in their substitutions (Fig. 66-12), but share common mechanisms for inhibiting acid secretion. PPIs are acid-labile lipophilic compounds that are absorbed in the proximal small intestine, circulate in the blood, and cross the basolateral surface of the parietal cell. Esomeprazole is the S-enantiomer of omeprazole, which is a racemic mixture of both S- and R-optical isomers. Through a series of in vitro studies, esomeprazole has been shown to undergo less hepatic metabolism through cytochrome P450 2C19 and decreased intrinsic clearance than the R-enantiomer of omeprazole. The difference in clearance results in slower plasma elimination of esomeprazole as compared with omeprazole. Clinical studies have demonstrated greater acid inhibition and diminished interindividual variability of esoprazole over omeprazole.

FIGURE 66-12. Chemical structure and reaction pathways of four of the proton pump inhibitors used in the United States. (Adapted with permission from ref. [482].)

PPIs are weak bases (pK $_a$ from 4.0 for omeprazole, lansoprazole, and pantoprazole to 5.0 for rabeprazole), and this facilitates their accumulation in acid compartments such as the parietal cell acid secreting space. Once in the acidic compartment, PPIs are converted into the reactive thiophilic sulfenamide through an acid-catalyzed reaction (see Fig. 66-12). The sulfenamide derivatives are permanent cations that are trapped within the parietal cell, where they react rapidly to form a covalent disulfide bond with a cysteine residue on the a-chain of the H $^+$,K $^+$-ATPase rendering it irreversibly inactive, with the possible exception of rabeprazole, which has been shown to have a shorter activation time and partial reversible binding in vitro. Acid secretion is blocked until a new enzyme unit is synthesized, which usually occurs within 96 hours. [481] The rate with which the different PPIs are converted to the corresponding active form varies and is determined by their structure

and pK $_a$. Relative rates of conversion are rabeprazole > omeprazole + lansoprazole > pantoprazole. [483], [484]

Effects on acid secretion. PPIs, the most potent inhibitors of acid secretion available, block both basal and stimulated acid formation. Although different doses of these agents are recommended for treatment of several acid-related disorders, when they are used at equivalent doses, PPIs are quite comparable in terms of their efficacy. [485] Through metaanalysis, Burget and colleagues [486] concluded that, to obtain healing of a duodenal ulcer at 4 weeks, the intragastric pH should be maintained at 3 or greater for 18 to 20 hours per day. Therefore, this gastric pH has often been used as a target when comparing the efficacy of the various PPIs. A detailed review of the many studies published examining the relative efficacy of each PPI is available. [474] It appears that, after reviewing all the studies, omeprazole, lansoprazole, and pantoprazole have equivalent potency when they are considered on a milligram-by-milligram basis. Rabeprazole (20 mg) has a significantly quicker antisecretory effect and longer duration of action than omeprazole (20 mg). [487] Whether the slight difference in onset and duration of acid inhibition of the various PPIs has any significant impact on clinical outcomes has not been established.

Pharmacokinetics. The onset of action for PPIs is approximately 1 hour, with maximal inhibition and duration of inhibition being dose dependent. They all have short plasma half-lives (~1 hour) and are unlikely to accumulate even when clearance is significantly diminished. [480] In contrast, the duration of the inhibitory effect on acid secretion is prolonged (48 to 72 hours) because of the irreversible modification of the H^+,K^+-ATPase. Rabeprazole dissociates from the acid secretory pump to a greater extent than other compounds; therefore, its duration of action is shorter. The inhibitory effect of omeprazole is increased with daily dosing, with steady state in action achieved within 3 to 4 days. Availability improves to 60% with repeated dosing, which may reflect impaired absorption as the gastric pH increases. [488], [489] In contrast, lansoprazole and pantoprazole have a higher and constant bioavailability, the latter achieving this availability independent of the dose used. [490], [491] and [492] Although ingestion of food has been shown to decrease absorption of lasoprazole in some studies, this has not been a consistent observation. [480] Concomitant use of antacids has been shown to have no effect on omeprazole availability, but conflicting results have been observed with the other PPIs. [480] PPIs are concentrated and activated only in acidified compartments; thus, they inactivate only those proton pumps that are actively secreting acid. In the fasting state, roughly 5% of proton pumps are actively secreting, compared with 60% to 70% in the meal-stimulated state. The acid-inhibitory potential of both omeprazole and lansoprazole are maximized by taking them immediately before a meal, providing the drug at the time of stimulated acid secretion. [493], [494] Unlike H $_2$-receptor antagonists, which are commonly administered at bedtime, it is best to administer the PPIs in the morning before breakfast. [495] Omeprazole is less effective when given to patients during prolonged periods of fasting, such as in an intensive care unit. Evidence of this is provided from pharmacodynamic studies of the intravenous form of omeprazole, which is not yet available in the United States. [496], [497] For these reasons, H $_2$-receptor antagonists are likely to remain the preferred agents for the inhibition of gastric secretion in the intensive care unit setting.

Formulation. PPIs are acid labile; therefore, each compound is prepared in a specialized formulation to minimize degradation and protonation within the stomach. Omeprazole and lansoprazole are administered as enteric-coated, pH-sensitive granules in a gelatin capsule. The capsules dissolve in the stomach, and the granules dissolve within the proximal small intestine, releasing the drug where a more favorable pH of 6 is encountered. Pantoprazole and rabeprazole are available as enteric-coated tablets. Pantoprazole is also available in a parenteral formulation for intravenous use.

Elimination. All PPIs are avidly bound to protein (>95%) and are rapidly metabolized in the liver with minimal clearance by the kidneys. They are metabolized in the liver in a first-pass effect by the cytochrome P450 enzyme system, and negligible amounts of unchanged drugs are found in the urine and feces. [480], [481], [498], [499] and [500] The absorption of omeprazole and lansoprazole is reduced in patients with cirrhosis, but the bioavailability is not affected. [475] The absorption of PPIs is not altered by renal failure, and renal function does not affect the elimination of PPIs.

Adverse effects. PPIs are well tolerated, related to the high level of drug selectivity and efficient clearance of these compounds, with adverse effects occurring to a similar degree in patients receiving an H $_2$ blocker or placebo. [501], [502] and [503] The most common side effects include headache, constipation, nausea, abdominal pain, and diarrhea. The frequency of these side effects is similar for all the PPIs (1% to 3%), except in the case of diarrhea and headache, which are reportedly less common with rabeprazole and pantoprazole. Minimal elevation in aspartate aminotransferase and alanine aminotransferase has also been observed. Withdrawal of drug because of side effects is less than 1% to 2%. Serious adverse reactions from PPIs are rare, but there are case reports of hepatitis with lansoprazole and omeprazole and of interstitial nephritis with omeprazole. [504], [505] and [506] As is the case with H $_2$-receptor antagonists, PPIs may inhibit the gastric absorption of drugs that require an acidic gastric pH for absorption such as ketoconazole, ampicillin esters, and digoxin. PPIs are extensively metabolized by the cytochrome P450 system. Increasing evidence suggests that omeprazole partly inhibits those drugs metabolized by the P450 enzyme subfamily IIC, including diazepam, phenytoin, and warfarin. [507], [508] The clinical significance of these interactions has been minimal in most studies. Nevertheless, caution is warranted when omeprazole is used in combination with these agents. [507], [508] Unlike cimetidine, omeprazole has little or no significant effect on the elimination of drugs metabolized by the P450 subfamily IA (e.g., theophylline), subfamily IID (e.g., metoprolol, propranolol), or subfamily IIIA (e.g., cyclosporine, lidocaine, quinidine, nifedipine). [508], [509] No significant interference in alcohol metabolism has been demonstrated. Lansoprazole does not interact with diazepam, warfarin, phenytoin, or prednisone. [510], [511] and [512] There is a small increase in theophylline clearance (10%) when it is administered consistently with lansoprazole. Rabeprazole, pantoprazole, and esomeprazole do not appear to interact significantly with drugs metabolized by the cytochrome P450 system.

Impact of Antisecretory Drugs on Serum Gastrin Gastrin secretion depends on gastric intralumenal pH, with inhibition of release occurring at a pH 3.0. or less. Conversely, elevating the postprandial intragastric pH to more than 6.0 results in an enhanced, sustained rise in the serum gastrin response to meals. The elevation can be abruptly suppressed by intragastric infusion of acid. [513] Twenty-four-hour intragastric acidity and simultaneous plasma gastrin levels are significantly increased after therapy with H $_2$-receptor antagonists and PPIs. [513], [514] and [515] The greater the degree of gastric acid suppression produced by a particular drug regimen, the greater is the integrated serum gastrin. [514] The mean rise in fasting and meal-stimulated gastrin release induced by short-term courses of “standard” doses of H $_2$-receptor antagonists is small but significant, with values remaining well within the range of normal in almost all cases. [513], [514] After discontinuation of H $_2$-receptor antagonist therapy, fasting gastrin levels may remain slightly elevated (although well within normal limits) for several days to weeks before returning to pretreatment levels. The impact of long-term H $_2$-receptor antagonist therapy on serum gastrin levels has not been well studied. An elevated fasting gastrin level occurred in 19% of a small group of patients receiving long-term nighttime maintenance H $_2$-receptor antagonist therapy; this value returned to normal in all cases within 6 weeks after discontinuing therapy. [516] Another study reported no difference in basal or meal-stimulated gastrin levels in patients receiving maintenance therapy. [517] As a consequence of their greater degree of acid suppression, courses of therapy with PPIs produce greater rises in serum gastrin levels than H $_2$-receptor antagonists. Although selected reports suggest there is some variation in the ability of different PPIs to induce hypergastrinemia (pantoprazole less than omeprazole, lansoprazole more than omeprazole, and rabeprazole more than omeprazole), these difference are not clinically significant. [480] Within 2 weeks of initiating PPI therapy, fasting gastrin concentrations are increased two- to fourfold in most patients. [513], [518], [519] and [520] Gastrin levels returned to pretreatment values within 7 to 14 days after discontinuation of therapy. The largest experience with PPI-induced hypergastrinemia has been observed with omeprazole. Large clinical trials confirm a statistically significant rise in fasting gastrin levels after 4 weeks of omeprazole therapy, with as many as 12% of values exceeding the upper limits of normal. [521] Studies of 24-hour gastrin profiles in patients taking omeprazole show that the meal-stimulated physiological fluctuations in serum gastrin are preserved, and gastrin levels approach baseline at the end of the dosing interval. The rise in integrated gastrin levels is proportional to the total omeprazole dose. [522] With long-term omeprazole (>1 year), gastrin levels appear to increase threefold to fourfold over the first 1 to 2 months of therapy and then generally remain stable. [523], [524] However, a continued gradual rise over 30 months has been reported. [524], [525] Gastrin levels are elevated in as many as 23% of patients receiving long-term omeprazole treatment, [525] and they return to normal within 2 to 4 weeks after discontinuing the drug. [432] The increase in 24-hour gastrin profiles in patients given omeprazole (20 mg/d) is slightly less than that after proximal gastric vagotomy, even though omeprazole inhibits a mean of 94% of 24-hour acidity compared with 78% for vagotomy. Vagotomized patients have not developed adverse clinical consequences after prolonged hypergastrinemia. [522], [526] The rise in the 24-hour serum gastrin level caused by omeprazole is an order of magnitude lower than observed in patients with pernicious anemia. [527] The rise in serum gastrin levels may be pronounced, exceeding 400 pg/mL in 3% to 6% of patients receiving long-term omeprazole therapy. [523], [524] and [525] This degree of hypergastrinemia is attributable in most instances to profound drug-induced acid suppression and may respond to lower doses of omeprazole or alternative acid-inhibitory agents. [518], [528]

Consequences of Prolonged Acid Suppression In addition to its acid-stimulating effects, gastrin is a trophic hormone that stimulates mucosal growth in the stomach. The most prominent trophic effect of gastrin in the mucosa is on the endocrine ECL cell in the oxyntic mucosa. [529], [530] In humans, prolonged, severe hypergastrinemia associated with such conditions as type A chronic atrophic gastritis, pernicious anemia, or Zollinger-Ellison syndrome (especially patients with MEN I) may result in hyperplasia of the ECL cells and, in 3% to 5% of cases, the development of gastric carcinoid tumors. [531], [532] and [533] The finding that female rats treated for 2 years with omeprazole developed ECL hyperplasia and as many as 40% developed gastric carcinoid tumors raised legitimate concerns about the risks of carcinoid development in patients receiving long-term PPI therapy. [534], [535] To put this theoretical concern into clinical perspective, several observations are relevant. PPIs have not been found to induce mutagenicity or teratogenicity in multiple genotoxicology studies (i.e., no direct carcinogenic potential). [536], [537] In rats, ECL cell hyperplasia and carcinoids may be induced after prolonged, marked hypergastrinemia from several causes: partial fundectomy, antral exclusion, prolonged infusions of H $_2$-receptor antagonists, and PPIs. [530], [538], [539] Increases in the ECL cell density occur as early as 9 days after initiation of hypergastrinemia, stabilize after 10 to 20 weeks of hypergastrinemia, and revert to normal within 20 weeks if gastrin levels are normalized. [540], [541] In these models, ECL cell hyperplasia and carcinoids are prevented by antrectomy, which eliminates secondary hypergastrinemia. [540], [541] In human patients receiving long-term omeprazole therapy for as long as 10 years, hyperplasia of ECL cells has been observed, but there has been no reported progression to dysplasia or carcinoid, other than for patients with Zollinger-Ellison syndrome. [522], [525], [542], [543] In two long-term studies of more than 500 patients receiving long-term omeprazole, the ECL cells have been assessed quantitatively and

qualitatively by repeat biopsies of the gastric corpus. In the largest study, the proportion of patients with ECL hyperplasia rose from 10% pretreatment to 16% after omeprazole treatment. This primarily resulted from an increase in the micronodular pattern of hyperplasia from 2.5% to 10%. [542] In a smaller study, there was a 100% increase in the mean ECL cell volume density over a 5-year period. This increase was linearly related to the mean serum gastrin concentration. The increase in endocrine cell number and size was paralleled by a decrease in the normal endocrine cell growth pattern from 64% to 33% of patients and an increase in micronodular hyperplasia from 9% to 17%. [525] The qualitative and quantitative increases in the ECL cell population were mostly restricted to the subset of patients with more severe hypergastrinemia (>240 pg/mL). [543] Long-term therapy (>2 years) with lansoprazole led to a fourfold increase in ECL volume, with patients developing hyperplasia without evidence of dysplasia or neoplasia. [544], [545] and [546] In both omeprazole studies, a significant proportion of patients treated had chronic corpus gastritis, primarily caused by *H pylori* infection, with varying degrees of active mucosal inflammation or atrophic gastritis. The changes in the endocrine cell population correlated with the severity of the corpus gastritis. ECL cell micronodular hyperplasia was detected in both studies in almost 50% of patients with chronic atrophic gastritis, compared with only 2% to 4% of those without evidence of gastritis. The increase in ECL cell hyperplasia observed over time in these studies paralleled an increase in the incidence of chronic atrophic gastritis. A similar progression in chronic gastritis and ECL cell micronodular hyperplasia has been shown in normogastrinemic patients with gastric ulcers who were followed without antisecretory therapy for years. [543] Kuipers and colleagues [547] noted the acceleration of atrophic gastritis in a cohort of *H pylori*–infected patients treated with a PPI compared with a cohort of historical controls who underwent fundoplication. Whether this translates into an increased risk of gastric cancer and whether *H pylori* eradication can reduce that risk has not been examined. The greatest potential for the development of ECL cell hyperplasia and carcinoids appears to be in patients with chronic fundic atrophic gastritis. The level of hypergastrinemia seen in most patients treated with omeprazole is mild and (in the absence of chronic gastritis) causes little change in the ECL cell population. However, the development of ECL cell hyperplasia in patients with chronic atrophic gastritis is clearly potentiated by hypergastrinemia, especially at gastrin levels greater than 240 pg/mL. Significant hypergastrinemia does occur in a small subset of treated patients, especially in those with chronic atrophic gastritis. At this time, there is no agreement regarding the need or utility of periodic monitoring of serum gastrin levels in patients receiving long-term PPI therapy. There has also been concern regarding the development of colonic dysplasia and neoplasia secondary to PPI-induced hypergastrinemia. No evidence of this was observed in rats in which colonic carcinomas were induced and animals were subsequently rendered hypergastrinemic. [545] In 1996, the FDA reviewed these data along with data from major pharmaceutical companies in the United States and concluded that there was insufficient evidence to support a clear association among the use of PPIs, *H pylori* infection, and the accelerated development of atrophic gastritis. No product labeling changes were proposed, and further investigations regarding the observation by Kuipers and colleagues are needed to explore this observation. Despite the lack of data, many practitioners have adopted an approach to diagnose and treat *H pylori* infection in this patient cohort.

Drugs that Promote Mucosal Protection

Sucralfate

Structure and pharmacology. Sucralfate is a complex salt of sucrose in which the eight hydroxyl groups have been replaced by sulfate and aluminum hydroxide. [548] Within this acidic milieu, there is gradual dissociation of aluminum hydroxide into solution, leaving highly polar sulfate anions that may bind electrostatically to positively charged tissue proteins and mucins. Sucralfate has little acid-neutralizing capacity. [433] Because of its poor solubility, only 3% to 5% is absorbed (and renally excreted), with the remainder excreted in the feces. [548] Aluminum, which accounts for 21% of sucralfate by weight, dissociates to some extent into solution, but less than 0.01% is absorbed. [445] This is similar to what occurs with antacids containing aluminum hydroxide, causing a small but significant rise in serum and urine aluminum levels within 2 days. [433] In patients with normal renal function, the minor amounts of aluminum absorption with short-term therapy are of no clinical significance. Sucralfate appears to be extremely safe for short-term and long-term use, but its impact on the disposition of aluminum in the body has not been adequately studied.

Mechanisms of action. Sucralfate has several actions that are postulated to be important in ulcer therapy. [433], [549], [550] and [551] As the sulfate residues bind to the tissue proteins, sucralfate forms a protective barrier that may retard diffusion of acid, bile salts, and pepsin from the lumen to the ulcer. Sucralfate may bind to and stabilize the gastroduodenal mucous layer, as well as stimulating mucus and HCO_3^- secretion and gastric mucosal prostaglandin E_2 synthesis, and other trophic effects, which may promote mucosal defense.

Adverse effects. Because of its lack of systemic absorption and its inert chemical structure, sucralfate is virtually devoid of systemic toxicity. Gastric bezoar formation has been reported. [433] Constipation occurs in 3% of patients. [433] In patients with chronic renal insufficiency, significant aluminum accumulation may occur with standard doses of long-term aluminum hydroxide or sucralfate. Rarely, aluminum neurotoxicity with these agents has occurred in patients undergoing hemodialysis, [446] and it is recommended that long-term therapy with aluminum-containing compounds be avoided in this patient population. [446] Aluminum salts effectively bind phosphates in the GI tract and result rarely in hypophosphatemia, especially in patients who are already phosphate depleted. [433], [549]

Drug interactions. Sucralfate may bind to and may limit their absorption of several drugs, notably quinolone antibiotics, phenytoin, and warfarin. However, significant interactions are rare, and these may be avoided by administration of sucralfate separately from other medications. [433]

Bismuth-Containing Agents

Formulations. Physicians and patients have used bismuth salts for hundreds of years to treat sundry GI conditions, including dyspepsia, diarrhea, abdominal pain, and amebiasis. [434] Beginning in the 1970s, bismuth salts were demonstrated to be effective as single agents in the healing of peptic ulcers, but they never achieved widespread usage because of the excellent patient acceptance of H_2-receptor antagonists. These compounds have been used alone and in combination with antibiotics for the eradication of *H pylori,* and they have undergone a major resurgence in the treatment of PUD. Two colloidal preparations of bismuth have been most widely used: colloidal bismuth subcitrate (CBS; De-Nol) and bismuth subsalicylate (BSS). CBS is not available in the United States but has been used widely in Europe for the treatment of PUD. BSS is available as an over-the-counter agent, Pepto-Bismol, in the United States for the nonspecific treatment of dyspepsia and diarrhea. It is a fixture in the medicine cabinet of an estimated 60% of households in the United States, [434] and it has been approved by the FDA for treatment of *H pylori* infection. BSS may be administered as a liquid (303 mg of bismuth and 260 mg of salicylate/30 mL) or as a tablet (151 mg bismuth and 102 mg salicylate). [434], [552]

Pharmacokinetics. More than 99% of bismuth is excreted in the feces. Colonic bacteria convert the bismuth salts to bismuth sulfide, which imparts a black color to the stools. [552] Minimal bismuth absorption occurs from the proximal GI tract and is greater with CBS than with BSS. [553] This has not resulted in significant toxicity with short-term use. [554] Mean serum levels after 6 weeks of CBS therapy are elevated (17 g/L), although much less than the level thought to be toxic (50 g/L). An estimated 0.2% of bismuth from CBS is absorbed. [434] Absorbed bismuth is thought to be sequestered in multiple tissue sites in the body and is slowly excreted in the urine for 3 months or longer at approximately 2.6% per day. [434] Concomitant H_2-receptor antagonists enhance bismuth absorption from CBS but not from other bismuth formulations. Mean salicylate levels in patients given BSS in conventional doses (2.1 g/d) for 3 weeks produce a steady-state plasma salicylate level of only 2.4 mg/dL. [434]

Mechanisms of action. The mechanisms of action of bismuth salts are largely unknown, but they may involve binding and protecting ulcer crater or enhancing mucosal defense. They have no effect on gastric acid secretion and only a minimal effect on peptic activity. In endoscopic studies of patients given short-term aspirin therapy, bismuth markedly reduced the severity of mucosal lesions. [555], [556] Bismuth agents may also retard peptic degradation of epidermal growth factor. [557] Bismuth agents inhibit the growth of *H pylori* by causing detachment of the organism from the mucosa and bacterial lysis. [434]

Adverse effects. There have been no documented cases of bismuth toxicity from acute courses of BSS or CBS in recommended doses. There have been only three reported cases of bismuth neurotoxicity related to CBS and BSS, all involving patients who had used these agents in large doses or for extended periods. [434] In view of the documented absorption that occurs with CBS, its use should be restricted to short-term treatment only. In view of its salicylate content, BSS should be used with caution in patients with salicylate sensitivity, bleeding disorders, and renal failure, in elderly patients or in combination with other salicylate-containing drugs. The safety of long-term BSS has not been ascertained.

Prostaglandin Analogs

Structure and formulations. Prostaglandins are derivatives of 20-carbon–chain unsaturated fatty acids, also referred to as eicosanoids. The human GI mucosa contains and synthesizes several prostaglandins, but the primary prostaglandins are PGE_2 and PGF_2, which are produced in a ratio of about 2:1. Although they have a multitude of biologic effects, naturally occurring prostaglandins have a short biologic half-life of only a few minutes, and this limits their potential use as therapeutic agents. The only prostaglandin analog approved by the FDA for clinical use in PUD is misoprostol, a derivative of naturally occurring PGE_1.

Pharmacokinetics. Misoprostol is readily absorbed from the GI tract, with a peak plasma concentration at approximately 30 minutes after oral administration. The mean serum half-life is approximately 1.5 hours. Misoprostol metabolites are primarily excreted in the urine, but dose reduction is unnecessary in patients with chronic renal failure. [558], [559] Misoprostol does not affect hepatic cytochrome P450 drug metabolism.

Mechanisms of action. The therapeutic actions of misoprostol may be attributed to inhibition of gastric acid secretion and to stimulation of mucosal defense mechanisms. [435] The relative importance of these actions on the therapeutic efficacy is unclear. Direct mucosal contact appears to be necessary for its therapeutic action. After binding to the parietal cell, where there is a receptor for PGE, misoprostol exhibits dose-related inhibition of gastric acid secretion that is mediated through inhibition of histamine-stimulated cAMP production. [560] Although not as potent as H_2-receptor antagonists, misoprostol results in a significant reduction in nocturnal, basal, and meal-stimulated acid secretion at a standard therapeutic dose of 200 µg given four times daily. [561], [562] Prostaglandin analogs also stimulate mucus and HCO_3^- secretion, which enhances the pH mucous barrier, [561], [563], [564] enhances mucosal blood flow, and inhibits mucosal cell turnover. [565]

Adverse effects. Dose-related diarrhea is reported in 10% to 30% of patients receiving prostaglandin analogs. [435] This tends to occur early in the course of therapy and may be self-limited. Diarrhea may be the result of prostaglandin stimulation of intestinal sodium, chloride, and water secretion, or it may be secondary to changes in intestinal postprandial motility, resulting in accelerated orocecal transit time. [566], [567] Administration of misoprostol with food may decrease the incidence of diarrhea. Prostaglandins stimulate uterine smooth muscle, resulting in an increase in uterine pressure. Uterine bleeding has been reported with prostaglandin analogs in as many as one third of women during the first trimester of pregnancy. The use of these agents in women who may be pregnant is contraindicated. All women of childbearing potential should be warned of this risk before initiating misoprostol therapy. Vaginal bleeding may also occur in postmenopausal women. [435]

Miscellaneous Drugs

Anticholinergics. Anticholinergic agents are ineffective inhibitors of gastric acid secretion compared with H_2-receptor antagonists or PPIs, and they have been less effective than these agents in ulcer healing. Moreover, anticholinergic side effects (dry mouth, blurred vision, urinary retention) make these agents less desirable in the treatment of peptic disorders.

Tricyclic antidepressants. Antidepressant agents cause a mild decrease in acid and pepsin secretion, presumably mediated by inhibition of cholinergic and histamine H_2 receptors. Because of multiple side effects and the ready availability of superior agents, however, they have no role in the routine therapy of PUD. [568]

Carbenoxolone. Carbenoxolone is a licorice extract that was used many years ago to treat PUD, but it has no utility in the treatment of this disease. Its mode of action is unknown, but many of its effects resemble those of prostaglandins, and it has been shown to decrease prostaglandin metabolism. Carbenoxolone has aldosterone-like effects that can result in significant fluid retention and hypokalemia.

Treatment of Active Ulcers

The discovery of *H pylori* and of its role in PUD has had a major impact on the approach to treatment of gastric and duodenal ulcers. Before this discovery, annual ulcer recurrence rates after using antisecretory agents alone were as high as 80%, [569], [570] often requiring long-term maintenance therapy for ulcer prevention. Treatment concepts today should focus primarily on *H pylori*– and NSAID-induced mucosal disease. Most peptic ulcer trials have examined a relatively healthy patient population. Patients who are elderly, who have complicated ulcer disease (e.g., bleeding, perforation), who have large or multiple ulcers, who have severe underlying medical illnesses, or who are pregnant or lactating were specifically excluded from these trials; therefore, these data must be interpreted in the context of the defined population. These limitations notwithstanding, results of the randomized, double-blind, controlled trials are the standards on which clinical practice should be based.

Most patients with gastric ulcers have normal or reduced levels of acid secretion. It may seem paradoxical to treat such patients with acid-inhibitory agents. Overall, it is the duration of treatment that appears to be one of the most important factors in gastric ulcer healing. With or without therapy, there is a strong correlation between gastric ulcer healing and the duration of treatment or observation. Placebo healing rates increase over time, approaching 50% to 60% at 8 weeks. [571] In contrast, duodenal ulcer healing rates with placebo do not increase significantly after 4 weeks. Therefore, the duration of therapy may be more important than the potency of the pharmacological agent.

The following section first outlines, in general terms, the efficacy of the previously described drugs in the therapy of duodenal and gastric ulcers. Sections focusing on the therapy of *H pylori*– and NSAID-related disease follow.

Therapy for Duodenal and Gastric Ulcers

Acid-Neutralizing and Acid-Inhibitory Drugs

Antacids. Antacids have been found in multiple trials to be comparable to H_2-receptor antagonists in efficacy over a wide dose range of 120 to 1008 mmol/d, [572], [573] and [574] and they are superior to placebo and equivalent to H_2-receptor antagonists in relieving ulcer symptoms. [572], [573] There appears to be no advantage to using antacid regimens of more than two or three Mylanta DS or Maalox TC tablets four times daily. [572] Because of the need for four-times-daily dosing and the moderate incidence of diarrhea (secondary to magnesium) and constipation (secondary to aluminum), antacid therapy therefore should not be used as first-line ulcer therapy.

Acid-inhibitory drugs. Burget and colleagues [575] developed an elegant model of the relation between duodenal ulcer healing and antisecretory therapy. The rate of duodenal ulcer healing was found to depend on one of three variables: the degree of acid inhibition, the duration of acid inhibition, and the total duration of therapy. Antisecretory regimens that increase intragastric pH to more than 3.0 for more than 18 hours daily result in ulcer healing rates of 90% within 3 to 4 weeks. Regimens that raise the pH to more than 3.0 for only 8 to 10 hours daily can achieve similar healing rates after 6 to 8 weeks of therapy. Recognizing the association between duodenal ulcer healing and these three factors, the differences in healing rates among different antisecretory agents, doses, and dosing regimens become easily understood.

H_2-receptor antagonists. All four available H_2-receptor antagonists are efficacious in the treatment of gastric and duodenal ulcers. [431], [576], [577] Several randomized, double-blind trials have confirmed that once-daily bedtime dosing of H_2-receptor antagonists yields overall ulcer healing rates that are similar to twice-daily regimens. [571], [578] All the H_2-receptor antagonists have been approved by the FDA for once-daily administration in the treatment of duodenal ulcers: cimetidine (800 mg), ranitidine (300 mg), famotidine (40 mg), and nizatidine (300 mg) at bedtime. Healing rates for these regimens at 4 and 8 weeks of therapy are approximately 80% and 90%, respectively. Once-daily dosing also appears efficacious for treatment of gastric ulcers. [431], [448], [449] Higher doses of H_2-receptor antagonists, such as 300 mg of ranitidine twice to four times daily, result in higher 2- and 4-week duodenal ulcer healing rates, [639] but with the availability of PPIs, high-dose H_2-receptor antagonists appear to offer little advantage in the treatment of PUD. In general, gastric ulcers heal more slowly than duodenal ulcers, with average healing rates of 63%, 75%, and 88% after 4, 6, and 8 weeks, respectively. [431] The optimal H_2-receptor antagonist dosing regimen for gastric ulcers is undetermined. Trial results indicate that once-daily nocturnal H_2-receptor antagonist regimens are superior to placebo, and these nocturnal regimens are approved by the FDA. [431], [579] In summary, H_2-receptor antagonists should be administered between dinner and bedtime for 6 and 8 weeks to patients with duodenal and gastric ulcers, respectively.

H^+,K^+-ATPase inhibitors. PPIs result in gastroduodenal ulcer healing rates of more than 90% after 4 weeks of therapy. [427], [470], [474] The ulcer healing rate with omeprazole at 60 mg/d exceeds that of 20 mg/d at 2 weeks of therapy; however, at 4 weeks, these doses result in comparable healing rates in uncomplicated duodenal ulcers. [432] Omeprazole (20 to 40 mg/d) and lansoprazole (30 to 60 mg/d) have demonstrated efficacy in the treatment of acute gastric ulcers, with more than 90% healing after 8 weeks of therapy. [437], [475], [477], [479] The optimal dose has not been established, but healing may be slightly faster using the 40-mg dose. In several comparative trials, omeprazole resulted in significantly higher healing rates than H_2-receptor antagonists after 4 weeks and, in some trials, after 8 weeks of treatment. [579], [580], [581], [582] and [583] In a multicenter trial, 40 mg of omeprazole each day had significantly higher healing rates at 4 and 8 weeks of treatment than 20 mg of omeprazole each day or 150 mg of ranitidine twice daily. A metaanalysis suggests a slight advantage to omeprazole compared with H_2-receptor antagonists after 4 and 8 weeks of gastric ulcer treatment. [489] Pantoprazole has also been shown to be superior to ranitidine in gastric ulcer healing. [584] In deciding between H_2-receptor antagonists and PPIs, the clinician has two acceptable alternatives for uncomplicated ulcers. Both classes of drugs are efficacious in more than 90% of patients. They are safe and well tolerated, have excellent patient acceptance, and may be administered once daily. Although PPIs are more expensive than H_2-receptor antagonists on a daily basis, the cost of a 4-week treatment course of omeprazole compares favorably to a 6- to 8-week course of nongeneric H_2-receptor antagonists. PPIs may also provide more rapid pain relief than H_2-receptor antagonists, and this may be important to the patient. Although there are sparse data regarding patients with complicated gastric or duodenal ulcers, the superior efficacy of PPIs to other agents makes these the drug of choice for initial ulcer treatment in these patients. This includes patients with ulcers complicated by GI bleeding, ulcers larger than 1.5 to 2.0 cm in diameter, and patients with severe underlying medical disease. The advantage of PPIs over H_2-receptor antagonists in ulcer healing is especially pronounced in large ulcers (>1 cm). [585], [586] and [587] Although smoking retards ulcer healing, its impact may not be as great in patients treated with PPI as with H_2-receptor antagonists. [431], [585], [586], [587], [588], [589] and [590] Omeprazole and lansoprazole healed ulcers faster and relieved pain faster than H_2-receptor antagonists in most studies. [586], [588] Data with pantoprazole and rabeprazole demonstrate similar efficacy to omeprazole and lansoprazole in regard to ulcer healing. [584], [589] In patients with complicated ulcers who require hospital admission and nasogastric suction, a continuous intravenous infusion of H_2-receptor antagonists should be administered, rather than oral omeprazole. High, frequent doses of omeprazole are required in the fasting patient to maintain adequate acid suppression, thus limiting its utility in this setting. Similarly, in patients with peptic disease and gastric outlet obstruction, the absorption and bioavailability of omeprazole may be unpredictable. Future studies will clarify the utilities of intravenous pantaprazole in this patient population

Drugs that promote mucosal protection

Sucralfate. Sucralfate (1 g, four times daily) results in healing of 70% to 80% of duodenal ulcers after 4 weeks and 85% to 99% after 8 weeks of therapy. [433] In multiple comparative trials with H_2-receptor antagonists, there was no discernible difference in healing rates at 4 or 8 weeks between these agents. [433], [590] Sucralfate has also been shown to be effective at a dose of 2 g given twice daily in limited trials. Sucralfate has not been approved by the FDA for treatment of gastric ulcers. [591], [592] The development of more effective and easier to administer agents such as H_2-receptor antagonist and PPIs has made sucralfate somewhat obsolete.

Bismuth compounds. CBS is effective as a single agent in the short-term treatment of duodenal ulcer but is not approved by the FDA for use in the United States. BSS has not been adequately studied as a single agent in the treatment of duodenal ulcer disease. Limited data available on gastric ulcer healing with CBS suggest

that its superior to placebo and comparable to H_2-receptor antagonists in inducing gastric ulcer healing. [555]

Prostaglandin analogs. Prostaglandin analogs have little role in the treatment of acute duodenal or gastric ulcers.

Treatment of *Helicobacter pylori* Infection

The list of antimicrobial agents used to treat *H pylori* has grown exponentially since the 1990s. Moreover, studies reveal an improved clinical outcome and cost savings when patients with duodenal ulcer are successfully treated for *H pylori*. [593] It is important to realize that efficacy of any regimen for the treatment of this organism must be defined by demonstrating the absence of the microbe at least 4 weeks after completing therapy (eradication). [594], [595] Many agents may appear to be effective in clearing the organism within earlier time points, but this usually represents a decrease in the number of bacteria sufficient to render the diagnostic test falsely negative. The following is an overview of the different agents used for therapy of *H pylori.* A summary of treatment recommendations is outlined later in this chapter.

Antimicrobial Sensitivity Antimicrobial agents most commonly used for treatment of *H pylori* include amoxicillin, bismuth compounds, clarithromycin, metronidazole, and tetracycline. PPIs are also frequently part of the regimens used. Unlike other bacterial infections, susceptibility testing to antibiotics is not routine before initiating anti– *H pylori* treatment. Because metronidazole and clarithromycin are the mainstays of anti– *H pylori* treatment, knowledge regarding resistance to these and the other antibiotics may help to guide therapy. Metronidazole resistance is most common among *H pylori* isolates, occurring in approximately 10% to 60% of patients in developed countries, [114] with an even higher prevalence (e.g., 95% in South Korea) of resistance in developing parts of the world. [116] In contrast, the prevalence of clarithromycin resistance is generally not more than 10% to 15%, [596] with a tendency to be lower in countries where clarithromycin has only recently become available. In countries where clarithromycin has been available for some time, it appears that resistance rates are increasing. [597] Although resistance to tetracycline is rare, it has been noted to occur in up to 6% of isolates. [596] Resistance to amoxicillin is also rare. [598] Certain combination therapies may minimize the clinical impact of metronidazole resistance. Quadruple therapy, consisting of a PPI, metronidazole, tetracycline, and bismuth subcitrate, has been reported to be highly efficacious even against metronidazole-resistant strains. [599], [600] In vitro, ranitidine-bismuth citrate has been reported partially to overcome the effects of resistance to clarithromycin. [601] Unless sensitivity determinations are performed before initiating treatment, however, even initial regimens should probably exclude metronidazole or clarithromycin in patients known or suspected of having been exposed to these antimicrobials previously.

Current Treatment Regimens

Bismuth-based traditional therapy. In the United States, traditional triple therapy consists of BSS, two tablets (262 mg), metronidazole, 250 mg, and tetracycline, 500 mg, each taken four times daily for 10 to 14 days, providing a cure rate of approximately 85%. Substitution of amoxicillin for tetracycline results in a lower eradication rate and is not recommended. In Europe, bismuth subcitrate is often used instead of BSS, and treatment for 7 days has been shown to be effective. In the setting of an active ulcer, an acid-suppressive agent is added to hasten pain relief. Thus, traditional triple therapy for active ulcer disease is really quadruple therapy. Quadruple therapy, that is, combining traditional triple therapy, together with PPIs, preceded by a loading treatment phase of 3 days with a PPI alone, has been shown to be more efficacious and associated with fewer side effects than routine triple therapy. [600] More recent data suggest that pretreatment with PPIs for 3 days followed by 4 days of quadruple therapy is also efficacious. [602], [603] There is ongoing evaluation of a combination capsule containing tetracycline, bismuth, and metronidazole, which would significantly reduce the number of capsules taken per day. Preliminary studies have suggested that therapy with a PPI twice daily and three combination capsules three times daily for 10 days yields an eradication rate comparable to traditional quadruple therapy. As discussed in the section "Antimicrobial Resistance," a potential advantage of quadruple therapy for 7 days is the high cure rate of infection even with *H pylori* strains that are considered resistant to metronidazole.

Dual therapy. Clarithromycin is the single most effective anti– *H pylori* agent, and studies combining this antibacterial agent with either PPIs or ranitidine bismuth citrate have shown efficacy rates of approximately 65% to 80%. Specific FDA-approved regimens include the combination of clarithromycin, 500 mg three times daily, together with either omeprazole, 40 mg every day, or ranitidine–bismuth citrate, 400 mg twice daily for 14 days. Dual therapies with amoxicillin and a PPI have yielded variable cure rates in the United States and are thus not recommended. However, lansoprazole, 30 mg three times daily, together with amoxicillin, 1 g three times daily for 14 days, has been approved by the FDA for patients intolerant or allergic to clarithromycin or who are known to have organisms resistant to clarithromycin. This regimen has an eradication rate of approximately 70%. Because of their marginal efficacy relative to PPI triple antibiotic regimens and quadruple therapy, dual regimens have been relegated to the realm of historical interest.

Proton pump inhibitor–based and other triple therapies. The combination of two antibacterial agents with PPIs is among the most effective (approximately 85% to 90%) anti– *H pylori* regimens. Together with PPIs, combinations of clarithromycin and either amoxicillin or metronidazole are more effective than amoxicillin plus metronidazole. A standard dose of a PPI is taken together with clarithromycin, 500 mg, and either amoxicillin, 1 g, or metronidazole, 500 mg, all twice daily. In Europe, these regimens are effective if used for only 7 days, but in the United States, results are less clear. Therefore, 10 to 14 days of treatment are still recommended.

Adverse Effects Side effects of treatment relate to the particular regimen chosen. Patients should be educated about the importance of compliance and forewarned regarding minor side effects. Clarithromycin may cause taste disturbances, but this does not usually affect compliance. Metronidazole may be associated with side effects, particularly so at a dose greater than 1 g/d. Traditional triple therapy may be associated with mild side effects in about 50% of patients, and this includes vaginal moniliasis in up to 10% of women. Pseudomembranous colitis, a serious complication, occurs infrequently. In general, therapy is discontinued because of side effects in fewer than 5% of treated patients.

Considerations in Patients Who Remain Infected after a Course of Therapy

Primary treatment failure. There are only limited data addressing the management of patients in whom an initial attempt at eradicating *H pylori* infection fails. Therefore, most recommendations for management come from expert opinion. Dual therapy should not be used in the setting of treatment failure. In addition, it is prudent for a second course of therapy to be given for 10 to 14 days. After unsuccessful treatment of *H pylori* infection using metronidazole, the rate of secondary resistance is much higher than the primary resistance rate. An overall estimate for secondary metronidazole resistance is 80%. Data suggest that secondary resistance to clarithromycin after failed therapy develops in a significant percentage of patients treated with clarithromycin containing triple therapies. [604] Related to the high likelihood of secondary antibiotic resistance in the setting of primary treatment failure, if metronidazole or clarithromycin was used in the initial regimen, a second course of therapy should avoid these antibiotics. If clarithromycin was not used initially and there is no history of penicillin allergy, the combination of a PPI with clarithromycin and amoxicillin or tetracycline twice daily for 14 days is a reasonable choice. Ranitidine bismuth citrate (RBC) can be used in place of a PPI and may even provide improved efficacy in the setting of salvage therapy. [605] If the initial therapy contained clarithromycin but not metronidazole, a good choice would be quadruple therapy (PPI, tetracycline, bismuth, metronidazole) for 10 to 14 days. If both metronidazole and clarithromycin were included in the initial therapy, we have had success using a PPI, tetracycline, bismuth, and furazolidone (100 mg four times daily) for 14 days. Furazolidone appears to be effective even for most metronidazole-resistant *H pylori* strains. When furazolidone is used, patients should be warned not to consume alcohol or take monamine oxidase inhibitors concomitantly. In a study from Italy, the combination of a PPI twice daily, amoxicillin, 1 g twice daily, and rifabutin, 300 mg once daily for 10 days, yielded an eradication rate of 87% in patients in whom primary *H pylori* therapy had failed. [606] The role of antibiotic sensitivity testing as a means of selecting appropriate therapy in the setting of treatment failure remains poorly defined. At present, *H pylori* culture and antibiotic sensitivity testing are relatively time consuming and expensive. As a consequence, these techniques are not widely available. A decision analytic model comparing upper endoscopy with biopsy and culture and antibiotic susceptibility testing for *H pylori* with upper endoscopy with biopsy but no culture and antibiotic susceptibility testing found that pretreatment susceptibility testing could be cost effective depending on the prevalence of antibiotic resistance, the cure rate for sensitive strains, the cure rate for resistant strains, and the cost of diagnosing and treating *H pylori*. [607] It seems clear that as resistant strains of *H pylori* become more prevalent and culture and antibiotic sensitivity testing become more practical and accessible, these techniques could take on an important role in the management of treatment failures.

Reinfection. *Reinfection* is defined as the recurrence of *H pylori* infection after successful cure, as determined at least 4 weeks after treatment. A distinction is made between reinfection, in which a new strain is identified, and recrudescence, in which the strain is identical to the initial pretreatment organism. A very high recurrence rate may mitigate the desire for treatment. Recurrence rates probably relate to two factors. First, there may be an incorrect assessment of initial cure because of premature timing of the assessment and because of the possibility of decreased sensitivity of diagnostic tests in the posttreatment setting associated with a decreased bacterial load. Second, the prevalence of infection in the community or the particular living circumstances of patients (e.g., close contact, as in institutions) probably influences the rate of reinfection. Recurrences within the first 6 months after treatment are more likely to represent recrudescences, whereas reinfection may be more common thereafter. In general, it seems reasonable to assume that recurrence rates would be similar to the incidence rate of primary infection, which in the developed world is less than 1%/year. [608] However, even in a developing country such as China, where the prevalence of *H pylori* is more than twice that of the United States, the rate of reinfection was found to be low (average annual recurrence rate of approximately 1%) and similar to that found in developed countries. [609]

Treatment recommendations. At the present, peptic ulcers are the best established indication for *H pylori* treatment. Treatment of patients with nonulcer dyspepsia who are infected with *H pylori* and therapy for the sake of preventing gastric carcinoma remain controversial. In 1994, the National Institutes of Health (NIH) Consensus Conference concluded that all patients infected with *H pylori* with documented gastric or duodenal ulcers should receive antimicrobial therapy independent of whether NSAIDs had been ingested. [610] This recommendation included patients with newly diagnosed or recurrent ulcers. A similar consensus recommendation was put forth by the American College of Gastroenterology in conjunction with the American Gastroenterological Association and the American Society for Gastrointestinal Endoscopy. [611]

Treatment of Gastric and Duodenal Injury Related to Nonsteroidal Antiinflammatory Drugs

Treatment of Active Ulcers For patients with NSAID-associated gastroduodenal ulcers, the inciting agent should be discontinued if possible. When NSAIDs are being used for analgesic purposes rather than for their antiinflammatory effects, substitution of NSAIDs with acetaminophen or narcotics may provide analgesia. If NSAIDs are discontinued, gastric and duodenal ulcers respond rapidly to conventional therapy with H_2-receptor antagonists, omeprazole, or sucralfate. [612], [613], [614] and [615] In some patients with severe inflammatory rheumatic conditions, discontinuation of NSAIDs may be undesirable despite active ulceration. The use of COX-2–specific inhibitors or lower-risk NSAIDs such as salsalate, nabumetone, or etodolac may be useful in this situation. The safety with regard to clinical outcomes is more soundly established for the COX-2–specific inhibitors. Unfortunately, the optimal method of treating ulcers in patients who continue to take NSAIDs is not well established by randomized, controlled clinical trials. [616]

Antisecretory agents. It appears that H_2-receptor antagonists in conventional doses are capable of healing small (<5 mm) NSAID-related gastric and duodenal ulcers within 8 weeks, but the healing of larger ulcers, especially gastric ulcers, is markedly retarded. [617] Several trials have documented that the healing of NSAID-induced gastric and probably duodenal ulcers is delayed with H_2-receptor antagonist therapy if NSAID treatment is continued. [618], [619] In view of these findings, PPIs appear to be the preferred agent for the treatment of NSAID-associated ulcers. In a multicenter, double-blind, randomized trial comparing omeprazole with ranitidine, gastric ulcers healed within 8 weeks in 95% of a subset of 56 patients who continued NSAID therapy and who were treated with omeprazole (40 mg/d), in 82% with omeprazole (20 mg/d), and in 53% with ranitidine. [618] In a direct comparison of two doses of omeprazole, 20 and 40 mg, and ranitidine, 150 mg twice daily, in 541 patients, omeprazole was significantly more effective than H_2-blocker therapy. No advantage of 40 mg over 20 mg was seen. [619] The ability to heal NSAID-related ulcers rapidly while the drug is continued may have important clinical implications, given the number of anecdotal reports of serious GI complications of NSAIDs in patients receiving full-dose H_2-blocker therapy.

Misoprostol. In a randomized, double-blind study of patients, misoprostol demonstrated a 67% rate of healing of gastric and duodenal ulcers at 8 weeks when NSAIDs were continued, compared with 26% of patients treated with placebo. [620] A comparative study of misoprostol, 200 µg four times daily, and omeprazole, 20 and 40 mg/d, in 935 patients with NSAID-related ulcers demonstrated therapeutic success rates that were equivalent for all agents, with no advantage seen with high-dose omeprazole. Omeprazole, at either dose, was more effective in relieving NSAID-associated abdominal pain and indigestion, and it caused less diarrhea than misoprostol, leading to a reduced patient withdrawal rate compared with misoprostol and improved remission rate overall.

Mucosal protectants. Sucralfate does not appear to be effective in the presence of continued NSAIDs. [621]

Recommendations For patients with endoscopically documented uncomplicated gastric or duodenal ulcers, it is recommended that NSAIDs be discontinued, if possible. For patients with large or complicated NSAID-induced ulcerations, more rapid ulcer healing probably can be achieved with omeprazole, 20 to 40 mg/d, or its equivalent. The NIH Consensus Development Panel on *H pylori* recommended antimicrobial treatment of *H pylori*–infected patients with NSAID-induced gastric ulceration. Eradication of *H pylori* will not eliminate the likelihood that such a patient will have an ulcer recurrence if NSAID therapy is reinitiated; thus, preventive therapy is recommended in this situation. If NSAID therapy must be continued, the optimal approach supports potent acid suppression. It appears that small gastric and duodenal ulcerations heal with H_2-receptor antagonists, but larger or complicated ulcerations (>5 mm) may be better treated with a PPI. Because large ulcers are associated with a higher risk of complications, it should not be concluded that continued NSAID or COX-2–specific inhibitor therapy in the presence of large ulcerations is safe or advisable; the risks are unknown.

Prophylaxis for Ulcers Related to Nonsteroidal Antiinflammatory Drugs Because of the clear association between NSAID use and gastroduodenal ulcers, it would appear desirable to use an antiinflammatory agent with improved safety profile such as a COX-2–specific inhibitor or to administer prophylactic antiulcer therapy to prevent these ulcers and their complications. To address the efficacy of prophylactic therapy, several prospective endoscopic trials were performed. Most of these trials studied a population of patients with osteoarthritis or rheumatoid arthritis who were receiving long-term NSAID therapy but who did not have endoscopic evidence of a peptic ulcer at the start of the trial. Patients were then randomized to therapy with a prophylactic agent or placebo and followed with endoscopy over a 1- to 3-month period to determine the incidence of peptic ulcers. There are no prospective outcome trials to compare cotherapy approaches, so clinical judgment relies on data largely from endoscopic end points.

Prostaglandin analogs. Misoprostol has been shown to be an effective agent in the prevention of NSAID-induced gastric and duodenal ulcers and their complications. It is the only agent approved by the FDA for this purpose. However, despite its efficacy, its lack of impact on dyspepsia and its side effect profile make it the least commonly used cotherapy in clinical practice. In two large, multicenter, placebo-controlled studies of patients taking long-term NSAIDs, the 3-month incidence of gastric ulcers (>0.5 cm) among patients taking misoprostol (200 µg, four times daily) was less than 2%, compared with 8% to 12% among placebo-treated patients. [622], [623] In the larger trial, duodenal ulcers were found in fewer than 1% of patients receiving misoprostol but in almost 5% of placebo-treated patients. [622] It is worth emphasizing that 32% of patients treated with misoprostol developed diarrhea, compared with 18% of placebo-treated patients, although fewer than 4% of patients dropped out of the study because of this symptom. It is also noteworthy that the incidence of dyspepsia was similar among patients treated with misoprostol and those treated with placebo. Misoprostol has been found to be superior to H_2-receptor antagonists and sucralfate for the prevention of NSAID-induced gastric ulcers in randomized, prospective trials. [621], [623] Doses of 200 µg twice and three times daily significantly reduce rates of gastric and duodenal ulceration, with the higher dose more effective for gastric protection. [624] These data are of clinical relevance because the occurrence of diarrhea in up to 30% of patients on full-dose therapy is reduced by lower misoprostol doses. Studies up to 1 year in duration have confirmed the continued efficacy of misoprostol for the prevention of endoscopic ulcers. [625] The MUCOSA trial confirmed the ability of misoprostol to prevent the evolution to the complicated NSAID ulcer. In this randomized, double-blind trial of 8849 patients with rheumatoid arthritis who were taking long-term NSAIDs, misoprostol, 200 µg four times daily, reduced GI complications by 40% from 42 of 4439 (0.95%) in the placebo group to 25 of 4404 (0.57%) compared with placebo. [626] This study provided insight into numerous controversial issues. It confirmed the value of endoscopic ulcers as a surrogate marker for the development of clinically significant lesions. However, the reduction in GI complications was far less than the reduction in endoscopic lesions, making problematic the extrapolation from endoscopic lesion rates to reduction in clinically relevant outcomes for other agents or reduced misoprostol doses. There remains considerable debate in the literature regarding the cost effectiveness of misoprostol therapy for the prevention of NSAID ulcers. Current cost-effectiveness analyses have used reduction of endoscopic ulcers as the outcome of interest, and a pharmacoeconomic analysis of the MUCOSA data has not been published to date. A cost-utility assessment of misoprostol therapy in patients with rheumatoid arthritis demonstrated increased cost without additional quality-of-life benefits. [627] A critical review of the literature supports the idea that misoprostol is most cost effective when it is targeted to high-risk patient subgroups, [628] such as these with a history of previous PUD (52% reduction), significant cardiovascular disease history (38% reduction), significant functional disability (87% reduction), and those who required concomitant antacid use (48% reduction).

Mucosal protectants. A comparative study with misoprostol demonstrated the PGE_1 analog to be far superior for the prevention of NSAID ulcers, and ulceration rates in the sucralfate group were equivalent to rates in the placebo group in studies of similar design. [621]

H_2 antagonists: conventional dose. Studies have examined the ability of H_2 blockers, in traditional ulcer healing doses, to prevent NSAID-associated ulcers in arthritis patients. The findings in these studies probably apply to other H_2 blockers as well, because, when they are given in equivalent acid-suppressive doses, all have similar effects on GI ulceration. In studies with endoscopic ulcer end points, acid suppression with traditional-dose H_2 blockers did not provide clear-cut control of NSAID-induced GI toxicity compared with placebo. [629], [630] This is in contrast to nonendoscopy-based trials, which demonstrate efficacy for cimetidine and antacids compared with placebo for the prevention of NSAID-related dyspepsia. [631] The widespread coprescription of H_2 blockers in NSAID users reflects their efficacy for symptom control in clinical practice, despite the evidence that the major site of injury caused by NSAIDs, the stomach, is not protected.

High-dose acid suppression with H_2 blockers. In a 6-month study from the United Kingdom, [257] the reduction in the incidence of gastric ulcer in NSAID-users was dose dependent. The reduction in gastric ulcer rate was significant with only high-dose famotidine. There were trends for symptom improvement, and adverse event withdrawal was uncommon. Additional work by Hudson and colleagues [252] demonstrated that patients with prior NSAID ulcers are a high-risk group, and there is currently no cotherapy with 100% protective efficacy.

Potent acid suppression with proton pump inhibitors. Given that high-dose acid suppression with an H_2 blocker reduced NSAID ulcers in the stomach and duodenum, it follows that other, more potent acid inhibitors, should be as or more effective. Early experience with high-dose omeprazole in the acute prevention of aspirin-induced injury demonstrated that use of this PPI caused a remarkable reduction in both gastric and duodenal injury rates. [632] Researchers examined the long-term efficacy of omeprazole, 20 mg daily, in 177 higher-risk long-term NSAID users over a 3-month period in a Scandinavian multicenter study. [258] The patients studied had a history of PUD or ongoing dyspepsia. The reduction in peptic ulceration rates with omeprazole prophylaxis for NSAID-related ulcers was statistically significant. The peptic ulcer incidence was 4.7% with omeprazole versus 16.7% with placebo ($P < 0.05$), and dyspeptic symptoms persisted in 8.2% of the patients taking omeprazole compared with 20.0% taking placebo. This reduction was seen both for gastric ulcer (2 versus 6) and duodenal ulcer (2 versus 9); however, given the relatively small number of patients, significance was achieved only for the combined ulcer end point. Moreover, omeprazole significantly reduced the ulceration rate both in patients with previous ulcer and in those without. As shown in other studies, prior ulcer history was associated with an extremely high recurrence rate on placebo (approximately 50%). In another large multicenter study conducted in Europe, primary prophylaxis with omeprazole for NSAID-induced ulcers was studied in 169 patients requiring continuous NSAID therapy. [633] After 6 months, 78% of the omeprazole-treated group remained in remission, compared with 53% in the placebo group ($P = 0.004$). Two large randomized, double-blind studies evaluated the efficacy of omeprazole compared with ranitidine and misoprostol for both the healing and the subsequent recurrence of ulcers in individuals using NSAIDs. [614], [634] Omeprazole prevented ulcers more effectively (72% remission) than ranitidine (59%

remission) in patients who used NSAIDs regularly with similar rates of adverse events. Maintenance therapy of NSAID-associated ulcers with omeprazole was associated with a lower rate of relapse (61% remission) than misoprostol (48% remission) and was better tolerated. This difference was maintained when relapse was based on the development of both symptoms and endoscopic lesions. Thirty-two percent of patients taking placebo had gastric ulcers at relapse compared with 10% taking misoprostol and 13% taking omeprazole. Duodenal ulcers recurred in 12% of the placebo group, 10% in the misoprostol group, and 3% in the omeprazole group (Fig. 66-13). In addition, omeprazole was effective in reducing the symptoms in addition to reducing NSAID-associated endoscopic lesions, whereas misoprostol was ineffective and was associated with reductions in quality of life.

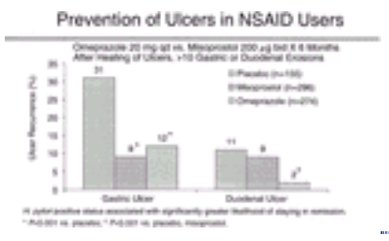

FIGURE 66-13. Prevention of ulcers in nonsteroidal antiinflammatory drug users with omeprazole or misoprostol. (Adapted from ref. 614.)

Studies that examined a pure NSAID-related ulcer group, namely, ulcers in NSAID users who are uninfected with *H pylori,* demonstrated that lansoprazole, 30 mg daily, was comparable to misoprostol,200 µg four times daily, with significantly fewer side effects, 635 supporting the widespread use of PPIs as an alternative strategy for prevention of NSAID-associated ulcers.

Recommendations. A prior history of peptic ulcer or ulcer complications increases the relative risk of NSAID therapy about 14-fold. It is axiomatic that NSAIDs should be avoided in such patients, and, if NSAIDs are to be used, extreme caution is urged. COX-2–specific inhibitors appear to be the drugs of choice in this situation. The use of the nonacetylated NSAIDs (e.g, salsalate) or the lower-risk NSAIDs (e.g., nabumetone, etodolac) may also be considered. Cotherapy with misoprostol or a PPI is recommended for all high risk patients taking NSAIDs. Because patients with major complications while taking NSAIDs were not studied in the outcome studies with the COX-2–specific inhibitors, cotherapy may also be considered for these patients. Patients who have a prior history of PUD or complications should be tested and treated for *H pylori* before initiation of NSAID therapy. It is unknown whether NSAID usage in such patients causes de novo ulcers, exacerbates the ulcer diathesis caused by *H pylori,* or both. It is unknown whether eradication of *H pylori* in these patients can lower the incidence of NSAID-induced ulcer complications. Pending further clinical study, ulcer prophylaxis is recommended for all patients with a history of symptomatic or complicated peptic ulcers, regardless of *H pylori* status. Because misoprostol has been shown to prevent gastric and duodenal ulcers and their complications, it is the preferred agent from the evidence-based medicine standpoint. In patients who are intolerant of misoprostol, alternatives should be considered. The use of lower doses of misoprostol is less expensive, is associated with less diarrhea, and may have significant efficacy in reducing ulceration. 623 In those situations, when misoprostol cannot be tolerated, potent acid suppression appears to be a reasonable alternative strategy to reduce ulcer risk. The benefits and cost effectiveness of prophylaxis in other situations are even more uncertain. Because outcome trials with the COX-2–specific inhibitors have established their superior GI safety compared with traditional agents, in the absence of head-to-head comparisons with cotherapy, either approach may be rational. The use of a single safer agent has obvious advantages for patient compliance. It is certain that the underlying patient risk and the likelihood of symptom-driven patient expenditures drive cost effectiveness among strategies to prevent symptomatic NSAID-related gastropathy. 636 Patients using concomitant corticosteroids with NSAIDs appear to be at greatly increased risk of developing NSAID-induced complications and should be considered for prophylactic or COX-2–specific inhibitor therapy. Similar considerations apply to those patients with serious underlying medical illness (marked disability, serious cardiac, pulmonary, renal diseases, rheumatoid arthritis, and patients receiving cotherapy with anticoagulants) 225 in whom an NSAID-induced complication would be poorly tolerated. Advancing age (>60 years) is also risk factor for NSAID-induced complications. However, it may not be cost effective to treat all elderly NSAID users prophylactically. 637

Prevention of Ulcer Recurrences and Complications

Natural History There is limited prospective information on the natural history of gastric and duodenal ulcers. Available data suggest that non–NSAID-related gastric ulcers have a relatively high recurrence rate of about 50% to 60% during the first 2 years after healing. The findings for duodenal ulcer suggest that about 70% to 80% of these patients have recurrences. Many of the reported duodenal ulcer recurrences represent asymptomatic ulcers found only at scheduled endoscopy in patients in ulcer trials, and the natural history of the asymptomatic duodenal ulcer requires further study.

Factors Associated with Ulcer Recurrence The major predictor of ulcer recurrence is the successful eradication of *H pylori,* which diminishes duodenal ulcer and gastric ulcer recurrences to 10% annually, in contrast to annual recurrence rates of 60% to 100% for those in whom *H pylori* is not eradicated. Because gastric and duodenal ulcer diseases are often associated with recurrences, it would be desirable to be able to predict and discriminately treat those who are destined for a recurrence. Several retrospective studies have attempted to relate various factors with ulcer recurrence rates. 638, 639 and 640 Many of these trials have been conducted in defined, sometimes small populations, such as teaching institutions or male veterans, and this makes the extraction of single predictive elements difficult and application of predictors to the general population questionable. Moreover, *H pylori* status was not determined in earlier studies. Despite these limitations, some tentative conclusions can be made about factors other than *H pylori* that can influence ulcer recurrence. Patients whose duodenal ulcers healed promptly, those who were younger, female, and nonsmokers, and those who had a more recent onset of ulcer disease, fewer pain episodes at entry, and a low basal or peak acid output had fewer ulcer recurrences than those without these features. 640 With follow-up periods as long as 6 years for 250 male veteran patients with duodenal ulcer with or without gastric ulcer, 11% of the total population developed ulcer complications, 639 and recurrences were threefold greater among those with prior ulcer complications requiring hospitalization than those without these criteria. Patient age, age at onset of ulcer disease, and family history were unrelated to the severity of disease. Van Deventer and colleagues 638 developed a risk index to predict the likelihood of ulcer recurrence. Cigarette smoking, alcohol intake, early disease onset (<40 years old), repeated duodenal ulcers, and endoscopic evidence of duodenal scarring or erosions were additive factors in the prediction of duodenal ulcer recurrence. 638 Each item has a value of 1, and patients with a risk index of less than 2 had about a 20% recurrence rate after 1 year, compared with a 100% recurrence rate for those with a score of 4. Although gastric ulcer disease is also associated with a high recurrence rate, there are no data, other than eradication of *H pylori,* that permit precise identification of specific factors associated with high or low ulcer recurrence rates. Continued NSAID intake, smoking, and possibly continued ethanol intake may contribute to gastric ulcer recurrences.

Role of Long-Term Maintenance Therapy Numerous clinical trials have confirmed the efficacy of continuous maintenance drug therapy in reducing the number of ulcer recurrences. 641, 642 and 643 It is our opinion, based on the available information, that long-term daily maintenance therapy with conventional medications has become obsolete. It has been estimated that the total cost of 2 weeks of triple therapy to eradicate *H pylori* is approximately $25. In the patient with recurrent ulcer disease, it is clearly cost effective to eradicate *H pylori* and to avoid the inherent costs of daily antiulcer drugs. Using a similar cost analysis, eradication therapy of *H pylori* is preferred to on-demand therapy. Based on the cost of approximately $3/d for a therapeutic dose of omeprazole, H_2-receptor antagonist, or sucralfate, and assuming that American patients with ulcers consumed doses comparable to those of Norwegian patients with ulcers (i.e., 10 weeks/y), the annual cost for antiulcer medications would exceed that of eradication of *H pylori.* These projections represent approximate estimates and do not include additional factors, such as the potential untoward effects of any medical program and indirect costs related to time lost from work. Careful cost analyses will be forthcoming.

Maintenance Therapy, *Helicobacter pylori* Eradication, and Ulcer Recurrences Eradication of *H pylori* and maintenance therapy with H_2-receptor antagonists change the proportion of symptomatic to asymptomatic ulcer recurrences. Although the true incidence of asymptomatic ulcers in the general populace is unknown, 30% of endoscopically documented ulcer recurrences are asymptomatic in patients receiving placebo treatment. 644, 645 In contrast, 50% to 70% of ulcer recurrences are asymptomatic in patients receiving maintenance therapy with H_2-receptor antagonists. 638, 646, 647 Some reports indicate that many patients with duodenal ulcer have asymptomatic ulcer recurrences after eradication of *H pylori.* The risk of serious complications in patients with chronic PUD is 2% to 3% per year, with a lifetime risk that approaches 20%. 642, 648 Complications are the major cause of ulcer mortality. Most maintenance studies have been limited to 12 months or less and have had insignificant sample size for addressing whether long-term maintenance treatment reduces this complication rate significantly. Reviews of placebo-controlled maintenance trials demonstrate a low rate of complications in active-treatment and placebo-treatment groups, although complications are numerically higher in the latter. 638, 649, 650 Patients with prior ulcer complications have an twofold increased risk of subsequent complications. Retrospective assessment of long-term maintenance trials suggests that such patients have an extremely low complication rate on maintenance therapy. 638, 649, 650 In a prospective study of patients with a history of bleeding peptic ulcers, recurrent bleeding occurred in 3.5% of 28 patients who were deemed compliant with their maintenance regimen. Recurrent bleeding occurred in 26% of 39 patients deemed noncompliant with the maintenance treatment. 651 Studies suggest that eradication of *H pylori* may diminish the frequency of ulcer bleeding. Thus, the eradication of *H pylori* or maintenance treatment may decrease the long-term ulcer complication rate. The low rate of complications observed in placebo-treatment groups in maintenance therapy suggests that close patient follow-up may be important in decreasing ulcer complication rates, but more data are necessary to answer this important clinical question. The increased proportion of asymptomatic ulcers compared with symptomatic ulcers does not appear to pose an increased risk of silent ulcer complications. Asymptomatic ulcer recurrences discovered incidentally in maintenance trials are likely to undergo spontaneous healing without an increase in H_2-antagonist therapy, and they may have little clinical relevance. 645, 646 and 647 Because of the very low incidence of ulcer complications among patients receiving maintenance treatment, it is difficult to ascertain whether patients who develop complications are more likely to be asymptomatic.

Recommendations for Treatment of Recurrent Ulcers The appropriate management of recurrent PUD depends on the numerous factors outlined previously. Only broad, tentative recommendations are given here. First, *H pylori* should be eradicated, patients should be admonished to give up smoking, and, if possible, NSAIDs should be discontinued. Eradication of *H pylori* should be documented by endoscopic biopsy or breath testing 1 month after therapy has been discontinued. If patients are still *H pylori* positive, they should receive an additional course of anti– *H pylori* and ulcer therapy. The few patients with recurrent ulcer who are *H pylori* negative in the absence of NSAIDs and other cofactors should receive maintenance therapy for 2 years. Fasting serum gastrin levels should be obtained in patients with frequent relapses to exclude Zollinger-Ellison syndrome. Because of the amelioration of ulcer symptoms that occurs in some patients over time, maintenance therapy may be electively discontinued in patients without prior ulcer complications or serious medical illnesses after 1 to 2 years, and patients are then observed closely for ulcer recurrence. Patients who continue to have frequent recurrences despite *H pylori* eradication should continue maintenance treatment indefinitely. Proximal gastric vagotomy or other ulcer surgery should be performed in the patient with recurrent ulcer complications that occur despite continuous maintenance treatment or because of medical noncompliance. Otherwise healthy patients with frequent ulcer recurrences in the absence of *H pylori* who require high-dose H_2-antagonist or omeprazole for maintenance therapy with an annual cost of about $600 should be considered for proximal gastric vagotomy.

Management of Refractory Ulcers A small subset (5% to 10%) of ulcers remains unhealed despite conventional therapy; these are called *refractory ulcers.* Although the definition of refractory ulcers varies among investigators, it is defined for the purposes of this discussion as a duodenal ulcer that fails to heal after 8 weeks or a gastric ulcer that fails to heal after 12 weeks of therapy. These ulcers pose a significant management problem, because they are more difficult to heal and because they generally have a high rate of early relapse. They are the subject of several reviews. [652], [653]

Factors contributing to refractory ulcer. Several factors may be important in the development of refractory ulcers: noncompliance, inadequate suppression of gastric acid, *H pylori* infection, and incorrect diagnosis. In clinical practice, an actual cause of refractoriness is seldom identified. [653] The most important causes of delayed healing are discussed in this section. Patients commonly stop taking their medications after their pain is improved. All patients with nonhealing ulcers should be asked about medical compliance before proceeding with further medical workup. In a series of 35 patients with refractory ulcers, 100% of patients had detectable serum salicylate levels, although more than 50% denied aspirin ingestion. [654] In a more recent study, NSAID use was found in 40% of patients with refractory ulcers (out of 60 patients), with 43.7% of the cases being surreptitious. [655] The history of NSAID ingestion should be elicited from all patients with refractory ulcers. Surreptitious aspirin ingestion may be uncovered by questioning family members or by measuring serum salicylate levels. Although platelet COX inhibition may be a more sensitive assay for detecting NSAID ingestion, this is not available in most clinical settings. [656] Smoking is an important risk factor for refractory ulcers. Patients should be strongly counseled to discontinue tobacco use. [657] Gastric acid hypersecretion has been detected by some investigators in patients with refractory duodenal ulcers. [652], [653] In a prospective study by Collen and associates [658] of 75 patients with duodenal ulcer, the mean basal acid output was 20.0 mEq/h among 20 patients with nonhealing ulcers at 8 weeks, compared with 6.6 mEq/h among the healed ulcer patients. It is likely that acid hypersecretion contributes to refractory duodenal ulcers in a subset of patients. The overlap of gastric acid secretion between groups with and without refractory ulcers is so great that measurement of gastric acid secretion is seldom useful in evaluating refractory duodenal ulcers or guiding therapy. Acid hypersecretion resulting from Zollinger-Ellison syndrome is a rare cause of refractory duodenal ulcers but should be excluded with a fasting serum gastrin level or a secretin stimulation test. In studies of H_2-receptor antagonist therapy lasting as long as 4 weeks, some degree of tolerance appeared to develop to these agents. [656], [659], [660] The antisecretory effect of H_2-receptor antagonists demonstrated on the first day of therapy diminishes with time. Tolerance is not observed with PPIs. [661] The mechanisms by which tolerance may occur are unclear but may include up-regulation of the histamine H_2-receptor and secondary hypergastrinemia, which could lead to increased basal and meal-stimulated acid secretion. [662] Because of the outstanding efficacy of H_2-receptor antagonists in treating acute peptic ulcers, tolerance to these drugs appears to be of little clinical significance for most patients with ulcers. [659], [661] The importance of tolerance in patients with ulcers that are refractory to standard doses of H_2-receptor antagonist therapy has not been studied. Most patients with refractory duodenal and gastric ulcers are infected with *H pylori.* For patients who do not have evidence of *H pylori*–associated gastritis, attention should be directed to other causes of peptic ulceration, including NSAIDs and acid hypersecretory conditions. An underlying gastric malignant disease may masquerade as a chronic nonhealing ulcer, and biopsies should be performed from the ulcer margin of all nonhealing gastric ulcers. There are several reports of malignant gastric ulcers that have undergone complete healing with initial H_2-receptor antagonist therapy. [662], [663] Although such events are rare, physicians should not assume that partial or even complete healing is proof of the benign nature of an ulcer. Unusual causes of chronic ulceration include Crohn's disease, amyloidosis, sarcoidosis, eosinophilic gastroenteritis, lymphoma, ischemia, and infections such as tuberculosis, syphilis, and cytomegalovirus. Biopsies should be obtained from duodenal and gastric nonhealing ulcers to exclude these conditions.

Evaluation and treatment of refractory ulcers. In managing the patient with a refractory peptic ulcer, it is important to verify patient compliance with the medication regimen. All patients should be admonished to discontinue smoking, and a careful inquiry should be made into possible NSAID ingestion. Upper endoscopy should be performed to confirm a nonhealing ulcer. Biopsies of the duodenal or gastric ulcer should be performed to exclude malignancy and other nonpeptic causes of ulceration. Biopsies of the gastric antrum or breath tests need to be obtained to assess *H pylori* status. A serum gastrin level should be measured to exclude Zollinger-Ellison syndrome. Gastric acid secretory studies are occasionally required. Regardless of the underlying pathophysiological mechanism, other than the patient's harboring *H pylori,* a marked reduction in gastric acid secretion usually results in healing of most refractory duodenal ulcers. Of the ulcers that are unhealed after 8 weeks of standard H_2-receptor antagonist therapy, approximately 50% to 60% heal with an additional 8 weeks of H_2-receptor antagonist therapy. [652], [653] The PPIs omeprazole (40 mg/d) and lansoprazole (30 to 60 mg/d) produce healing in more than 90% of refractory duodenal ulcers after 8 weeks of therapy. [653], [664] There have been multiple controlled, prospective trials using patients with refractory duodenal ulcers to compare continued or high-dose H_2-receptor antagonist therapy with omeprazole treatment. These trials demonstrated convincingly the superiority of 40 mg of omeprazole each day over H_2-receptor antagonists for the treatment of refractory duodenal ulcers. However, 20 mg of omeprazole daily appears to be an inadequate dose for the treatment of many patients with refractory ulcers. There is a dearth of information regarding the efficacy of antisecretory regimens for refractory gastric ulcers, because nonhealing gastric ulcers have generally been regarded as an indication for elective surgery to exclude occult malignant disease. [665] The few available studies report healing in more than 90% of refractory gastric ulcers after 8 weeks of treatment with PPIs. Potent antisecretory therapy has decreased the number of truly refractory ulcers that require surgery. Several centers have reported a higher ulcer recurrence rate after proximal gastric vagotomy in patients with refractory duodenal ulcers. The responsiveness of these postsurgical ulcers to medical therapy may be poor. The role of this surgical procedure in refractory duodenal ulcers compared with other surgical procedures that carry higher associated morbidity rate requires clarification. [653], [666]

Recommendations. Almost all refractory ulcers can be healed with omeprazole (40 mg/d). Because of the outstanding efficacy and short-term safety of this drug, it is recommended as the drug of choice for refractory ulcers. Similar results may also be achieved with all PPIs. Alternative therapies are less effective. A larger problem in the treatment of refractory ulcers is frequent, early ulcer recurrence. For patients who are *H pylori* positive, eradication therapy should be given in an attempt to prevent ulcer recurrences. Some patients with refractory ulcers may be *H pylori* negative, and eradication therapy may be unsuccessful for others. Maintenance therapy with full-dose or half-dose nocturnal H_2-receptor antagonists yields a 1-year symptomatic recurrence rate of 50% to 70%, approximately double the rate for other patients with ulcers. Omeprazole (40 mg/d) appears to be extremely effective in maintaining remission in limited trials, but a dose of 20 mg/d is less effective. [667] The risks and benefits of long-term omeprazole with surgery must be carefully weighed for these patients.

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CHAPTER 67

John Del Valle and James M. Scheiman

ZOLLINGER-ELLISON SYNDROME

“Fortunate indeed is the surgical service that does not have one or more problem cases of recurrent marginal ulceration despite the adequacy of the surgical attack,” noted the opening sentence of Zollinger and Ellison’s landmark description of the syndrome that today bears their names. [1] Benign ulceration of the upper jejunum associated with extremely high gastric acid secretion and the presence of a non–ß-cell adenoma of the pancreas was the triad of clinical findings described in two patients who eventually required total gastrectomy for relief of their severe ulcer disease. Although earlier reports [2], [3] described a similar clinical situation, it was Zollinger and Ellison who hypothesized that the components of this clinical triad were intimately related and were produced by a humoral agent that induced gastric acid hypersecretion and its sequelae. This hypothesis was confirmed by Gregory and colleagues, [4], [5] when they established that the tumor extract from a patient with Zollinger-Ellison syndrome contained a substance that was chemically and physiologically indistinguishable from the antral hormone gastrin.

Subsequent development of a sensitive and specific radioimmunoassay for gastrin [6], [7] facilitated the measurement of this peptide in the sera of patients with Zollinger-Ellison syndrome and confirmed that their circulating gastrin concentrations exceeded by severalfold the plasma levels found in a normal control population. This sensitive assay also permitted earlier tumor detection, a circumstance that has led to a steady increase in the number of patients described with gastrinoma. Potent inhibitors of gastric acid secretion and sophisticated preoperative and intraoperative diagnostic procedures have transformed Zollinger-Ellison syndrome from an illness for which total gastrectomy and tumor progression were the normal course to a condition for which a cure is feasible in many patients.

EPIDEMIOLOGY

The true incidence and prevalence of Zollinger-Ellison syndrome are unknown. The estimated incidence has ranged from 0.1% to 1% of patients who present with peptic ulcer disease in the United States. [8] The heterogeneous clinical manifestations, the widespread use of proton pump inhibitors (PPIs) for the treatment of acid peptic disease and its sequelae, [9] and the high incidence of silent pancreatic endocrine neoplasms [10] have all contributed to an underestimation of the incidence and prevalence of Zollinger-Ellison syndrome. The age range of affected persons has varied from 7 to 90 years, with the majority of patients diagnosed between the age of 30 and 50 years. Men with the disorder predominate over women by ratios varying from 2:1 to 3:2. [8], [11], [12]

Gastrinomas can be subdivided into tumors that are sporadic [13] and those that are genetically transmitted and associated with *multiple endocrine neoplasia type I* (MEN I). [14] Sporadic gastrinomas, the more common of the two, frequently behave as malignant tumors. Zollinger-Ellison tumors associated with MEN I occur at an earlier age than the sporadic tumors and have been characterized by some investigators to follow a more benign course. There are various distinguishing features between these two subtypes, as discussed later in this chapter.

PATHOPHYSIOLOGY

Gastric acid hypersecretion and severe peptic ulcer diathesis secondary to unbridled release of gastrin from a non–ß-cell endocrine neoplasm are the essential elements leading to Zollinger-Ellison syndrome. The neoplastic pancreatic cells secreting gastrin are thought to arise from the ductular epithelium and not from cells of the islets of Langerhans, despite the appellation of gastrinomas as “islet cell tumors.” [1] Normally, the adult pancreas does not secrete gastrin, but the fetal pancreas contains large quantities of this peptide. [15] After birth, the gastrin-secreting cells in the pancreas disappear and are not seen again, except as benign or malignant neoplasms in Zollinger-Ellison syndrome. The hypergastrinemia that results from the release of peptide from an endocrine neoplasm free of the usual regulatory restraints has two synergistic effects: overstimulation of gastric parietal cells to secrete acid and increased mass of parietal cells susceptible to overstimulation. The potentiated gastric acid hypersecretion that results is presumably the cause of the clinical manifestations (i.e., acid peptic disease and diarrhea) of the gastrinoma syndrome.

In contrast to many of the common malignant diseases, such as those originating in the pancreas, colon, or breast, gastrinomas are not associated with alterations in classic oncogenes such as *ras, myc, fos,* and *src* or with tumor suppressor genes such as *p53* or retinoblastoma susceptibility gene (*Rb*). [16] Despite this observation, studies reveal that 16% to 40% of sporadic gastrointestinal neuroendocrine tumors express mutations in the *MENI* gene, which is found to be defective in patients with MEN I. This is a 10-exon gene found in chromosome 11q13, [17], [18], [19] and [20] as further described later in this chapter.

Goebel and colleagues [21] attempted to correlate mutations of the *MENI* gene with clinical characteristics in 51 patients with sporadic gastrinoma. This study revealed that 31% of the tumors had a mutated *MENI* gene. Of interest, one mutation appeared to be clustered between a fairly narrow region (amino acids 61 to 166), a finding different from that seen in MEN I–related gastrinomas, in which the mutations occur throughout the gene. The presence of a mutation in the *MENI* gene did not appear to be useful as a marker for predicting patient outcome at an early stage of disease.

Additional studies have demonstrated a series of genetic alterations in gastrinomas including methylation and deletion of the *p16/MTS1* tumor suppressor gene, [22] amplification of the *HER-2/neu* protooncogene, [23] amplification of chromosome 9q, [24] and deletion of chromosome 1 [25], [26] or 3p. [24], [25], [26] and [27] More recently, Yu and colleagues [27] performed a detailed survey of genetic alterations in eight sporadic gastrinomas. Although the number of tumors was small, these investigators implied that important molecular features of gastrinomas include defects in chromosome 1, aneuploidy, and mismatch repair defects and deletions in the *MENI* gene. Serrano and colleagues [28] determined that alterations (hypermethylation of the promoter) in the tumor suppressor gene *p16INK4a/CDKN2A (p16INK4a)* was present in more than 50% of gastrinomas, independent of tumor extent. This genetic alteration is the most common one found in gastrinomas to date and suggests that changes in this gene may be an early event in its development. Although posttranslational processing variants of gastrin have been described in patients with gastrinoma, [29], [30], [31] and [32] the clinical relevance of this finding has not been established.

TUMOR DISTRIBUTION

More than 80% of gastrinomas have been localized in the anatomic area known as the *gastrinoma triangle.*[33] The boundaries of this triangle include the confluence of the cystic and common bile ducts superiorly, the junction of the second and third portions of the duodenum inferiorly, and the junction of the neck and body of the pancreas medially. The most common extrapancreatic site is the duodenum,[34] wherein up to 40% to 50% of gastrinomas arise.[35], [36] and [37] Other less common extrapancreatic sites include the stomach, bones, ovaries, liver, heart, and lymph nodes, which together account for fewer than 10% of gastrinomas.[38], [39] and [40] More than 50% of gastrinomas are considered to be malignant. Solitary pancreatic lesions were found in fewer than 30% of the earliest reported patients with gastrinoma.[11] With the trend toward earlier measurement of serum gastrin and exploratory surgery,[41] the incidence of metastatic disease at the time of operation has decreased. Nevertheless, multiple tumors or metastatic lesions are still observed in 30% to 55% of patients at the time of diagnosis.[35], [37]

The morphology of the neoplastic cells in gastrinomas is heterogeneous.[42], [43] The gastrin-producing cells are generally well differentiated and contain histological markers characteristic of endocrine neoplasms in general; that is, they contain chromogranin, neuron-specific enolase, and tyrosine hydroxylase. The degree of malignancy does not appear to correlate with a gastrinoma's histological appearance; however, this observation must be tempered by the knowledge that tumor aggressiveness is usually determined retrospectively.

As in the case with other endocrine neoplasms, gastrinomas have been found to express a variety of neuroendocrine peptides besides gastrin, including somatostatin, pancreatic polypeptide, adrenocorticotropic hormone, and vasoactive intestinal polypeptide. Although the clinical manifestations are generally associated with overproduction of one hormone, case reports illustrating combined syndromes have been described.[44], [45], [46], [47] and [48] Of these, Cushing syndrome in association with gastrinoma is the mixed syndrome most frequently described.[49], [50]

CLINICAL MANIFESTATIONS

More than 90% of patients with gastrinoma develop ulcers in the upper gastrointestinal tract at some point during the disease.[11], [12] and [13] Presenting ulcer symptoms are indistinguishable from those associated with benign peptic ulcer disease, but frequently they are less responsive to standard therapy. As in benign peptic ulcer disease, ulcers in patients with Zollinger-Ellison syndrome occur most frequently in the first portion of the duodenum (75%), and they are usually solitary. However, ulcers in patients with gastrinoma also may occur in the second, third, and fourth portions of the duodenum (14%), as well as in the jejunum (11%).[12] The ulcers are usually less than 1 cm in diameter but can occasionally present as giant lesions. Peptic ulcer disease refractory to standard medical therapy, recurrent ulcers after prior gastric surgery, diarrhea in patients with ulcers, or peptic disease presenting with complications such as obstruction, perforation, or bleeding suggest a possible diagnosis of Zollinger-Ellison syndrome. Esophageal symptoms caused by acid reflux are seen in as many as two thirds of patients with Zollinger-Ellison syndrome.[51], [52] The disease found in the esophagus may range from mild to severe esophagitis complicated by stricture formation and Barrett mucosa.

Diarrhea is the other common feature of Zollinger-Ellison syndrome; it can be present in more than half of patients, and it may precede the diagnosis by many years and occur in the absence of peptic ulcer symptoms.[11], [13], [53], [54] The pathogenesis of diarrhea is likely multifactorial, but its dependence on acid hypersecretion is demonstrated by amelioration of symptoms on continuous nasogastric suction or inhibition of acid secretion.[8], [55] Added features of the diarrhea are steatorrhea and maldigestion,[56], [57] which result from the inactivity of pancreatic enzymes at the low pH of the duodenum caused by the excessive acid load. The acidic pH of the small bowel also may lead to damage of the intestinal mucosa, resulting in a spruelike state with flattened intestinal villi and accompanying malabsorption.[58] Bile acids are poorly soluble in an acid milieu; thus, Zollinger-Ellison syndrome may result in decreased micelle formation and subsequent malabsorption of vitamin B 12 and other lipid-soluble nutrients and vitamins.[56] It is possible that in the presence of markedly high serum gastrin levels, gastrin itself may increase secretion of potassium and may reduce absorption of sodium and water by the small intestine,[59], [60] thus potentially leading to secretory diarrhea. Secretory diarrhea also may occur in Zollinger-Ellison syndrome in association with the secretion of another hormone besides gastrin, such as vasoactive intestinal polypeptide.

Twenty-five percent of patients with gastrinoma have MEN I syndrome.[14] The primary sites of organ involvement in this autosomal-dominant genetic disorder include the parathyroid, pancreas, pituitary, and, less commonly, adrenal cortex and thyroid. In a large series by Ballard and colleagues,[61] 87% of 85 patients with MEN I had involvement of the parathyroid glands, 81% had disorders of the endocrine pancreas, and 65% had pituitary involvement. Trump and colleagues[62] reported their findings in 220 patients with MEN I and observed different rates of organ system involvement, with 95%, 41%, and 30% of patients having parathyroid, pancreatic, and pituitary neoplasms, respectively. This later study also confirmed that parathyroid lesions were the first indication of this genetic disorder in 87% of patients. The clinical features of Zollinger-Ellison syndrome (ulcer complications, gastric or pancreatic surgery) account for the major morbidity and mortality related to MEN I.[63] The first clinical manifestation of MEN I may be Zollinger-Ellison syndrome in up to one third of patients, a finding suggesting that patients with the presumptive diagnosis of sporadic Zollinger-Ellison syndrome be monitored biochemically and, when feasible, genetically screened for evidence of MEN I.[64] Others recommend an oral calcium tolerance test to exclude early evidence of hyperparathyroidism and MEN I.[65]

The genetic defect in patients with MEN I has been mapped to the long arm of chromosome 11 (11q11-q13) by analysis of restriction fragment length polymorphism in affected families.[62], [63] As outlined earlier, the *MENI* gene consists of 10 exons and expresses the protein menin. Menin is a nuclear protein of 610 amino acids in length and is expressed widely throughout the body and functions as a tumor suppressor gene.[66] It interacts with Jun D an AP-1 transcription factor and with a protein involved in cell growth, SMAD-3.[67], [68]

Hyperparathyroidism and the associated hypercalcemia have direct bearing on the gastric acid hypersecretory state found in patients with Zollinger-Ellison syndrome and MEN I. Intravenous infusion of calcium in healthy human volunteers induces gastric acid hypersecretion, and calcium has been shown both in vivo[69] and in vitro[70] to stimulate gastrin release directly from gastrinomas. Moreover, gastrinomas express the calcium-sensing receptor,[71] thus explaining the mechanism by which calcium leads to gastrin release from these tumors. Indeed, previous reports[72], [73] have demonstrated that resolution of hypercalcemia by parathyroidectomy reduces basal acid output (BAO) and fasting serum gastrin concentrations in patients with gastrinoma with MEN I.

One consequence of prolonged hypergastrinemia in patients with Zollinger-Ellison syndrome with MEN I is the development of primary gastric carcinoid tumors. In a review of these data from the National Institutes of Health (NIH), Jensen[74] reported that 13% of patients with Zollinger-Ellison tumors with MEN I developed gastric carcinoids. Similar tumors have been noted previously in patients with achlorhydria and hypergastrinemia,[75] as well as in rats made hypochlorhydric and hypergastrinemic with potent antisecretory agents such as omeprazole. These findings presumably arise from the action of gastrin to induce cellular proliferation. However, gastrin may not be the only cause of the carcinoids, inasmuch as in Jensen's NIH series[74] only 0.6% of patients with sporadic Zollinger-Ellison tumors developed carcinoids. In support of the hypothesis that hypergastrinemia alone is not sufficient to promote enterochromaffin-like cell gastric carcinoid tumor development, it was observed that 75% (15 of 20 patients) of Zollinger-Ellison syndrome–related gastric carcinoid tumors had loss of heterozygosity at the *MENI* gene loci (11q13).[76] Therefore, it is postulated that the enterochromaffin-like cell carcinoid is an independent tumor type of MEN I, sharing a similar pathogenesis by inactivation of the *MENI* gene.

Once a secretory endocrine tumor is diagnosed, patients with MEN I should be evaluated carefully for the presence of other endocrinopathies. A thorough medical and family history must be obtained. Diagnostic studies should include evaluation of visual fields, fasting blood glucose, serum calcium, and prolactin levels. If there is suspicion of a pituitary lesion, a computed tomography (CT) or magnetic resonance imaging (MRI) scan of the head should be considered. MEN I is an autosomal-dominant disorder with a high degree of penetrance. Children of these patients have an approximately 50% risk of developing this disorder by 20 years of age[77]; thus, genetic counseling and screening of family members are imperative. In a study evaluating a large kindred (221 members) of patients with MEN I, elevated albumin-adjusted calcium, parathyroid hormone, gastrin, and prolactin (in girls and women) were the most frequently encountered laboratory abnormalities among presumed carriers of the *MENI* gene.[78] Trump and colleagues[62] evaluated 709 members of 62 families with MEN I and developed a screening program based on their observations (Fig. 67-1). The genetic defect in MEN I can be used to develop predictive testing for this syndrome[66], [79], [80] and can eventually serve as the basis for informed genetic counseling in families with MEN I.

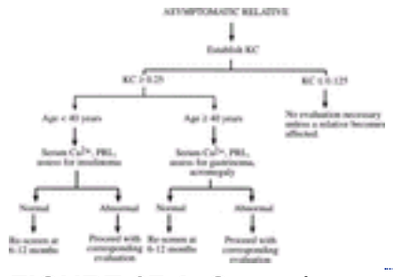

FIGURE 67-1. Screening protocol for relatives of patients with multiple endocrine neoplasia type I. *KC* refers to the coefficient of kinship, in which known affected individuals were assigned a KC of 1.0. Asymptomatic first-degree relatives (siblings, children) were assigned a KC of 0.5, and asymptomatic second-degree relatives (grandchildren, nephews, nieces) were assigned a KC of 0.25. Spouses were also screened and were assigned a KC of 0. (Adapted from ref. [62].)

A consensus statement for genetic screening and follow-up of patients with MEN I and their family members has been published. [81] These authors recommended that mutation analysis of the *MENI* gene be performed in patients meeting the following criteria: newly diagnosed neuroendocrine tumor typically found in MEN I (primary hyperparathyroidism, pancreatic endocrine tumor, or pituitary adenoma); age less than 40 years; positive family history; recurrent or multifocal neoplasia; or involvement of two or more organ systems. It is also recommended that genetic screening be performed in early adolescence of first-degree relatives of patients with MEN I. Persons carrying the *MENI* mutation should undergo annual biochemical screening, as outlined earlier.

DIFFERENTIAL DIAGNOSIS AND DIAGNOSTIC STUDIES

The various clinical circumstances in which a serum gastrin level should be obtained because of a suspected gastrinoma are outlined in Table 67-1. Fasting serum gastrin levels in healthy persons and in patients with routine peptic ulcer disease are usually less than 150 pg/mL. [82] The degree of hypergastrinemia in patients with Zollinger-Ellison syndrome varies greatly. Although occasional reports of normal levels, [83], [84] and values that vary from day to day, [85], [86] have been described, virtually all patients with gastrinoma have fasting levels greater than 150 pg/mL, with levels exceeding 100,000 pg/mL in some patients. A serum gastrin level greater than 1000 pg/mL in the right clinical setting is virtually diagnostic of Zollinger-Ellison syndrome; however, many patients do not have this level of hypergastrinemia. Elevated serum gastrin levels are seen in certain other clinical conditions as well (Table 67-2).

Multiple ulcers
Ulcers in unusual locations
Ulcers associated with severe esophagitis
Ulcers resistant to therapy, with frequent recurrences
Ulcer patients awaiting surgery
Extensive family history of peptic ulcer disease
Postoperative ulcer recurrence
Basal hyperchlorhydria
Unexplained diarrhea or steatorrhea
Hypercalcemia
Family history of pancreatic islet, pituitary, or parathyroid tumor
Prominent gastric or duodenal folds

TABLE 67-1 When to Obtain a Serum Gastrin Determination in Patients with Peptic Ulcer

Hypochlorhydria or achlorhydria with or without pernicious anemia
Retained gastric antrum
G-cell hyperplasia
Renal insufficiency
Massive small bowel resection
Gastric outlet obstruction
Other conditions (rheumatoid arthritis, vitiligo, diabetes, pheochromocytoma)

TABLE 67-2 Hypergastrinemia: Differential Diagnosis

Chromogranin A is a glycoprotein constituent of the secretory granules found in many neuroendocrine cells, [87] and as such it has been used as a histological and serologic marker for neuroendocrine tumors. [88], [89] and [90] Although serum gastrin and chromogranin A were found to be sensitive indicators for the presence of a gastrinoma, the latter had a relatively low specificity (67%). [90] Moreover, neither of the markers correlated with tumor growth or extent. However, it was observed in one patient that a rise in chromogranin A levels, which may reflect the extent of tumor and not enterochromaffin-like cell hyperplasia. This possibility requires further study.

With the ever-increasing use of potent antisecretory agents such as long-acting H_2 receptor antagonists and PPIs, drug-induced hypergastrinemia [91] must be excluded before proceeding with an extensive diagnostic evaluation for Zollinger-Ellison syndrome. The level of hypergastrinemia is reversible with drug cessation and is usually less than 1.5 to 2 times normal levels. Many patients with *Helicobacter pylori* infection and peptic disease develop hypergastrinemia and gastric acid hypersecretion, [92] which are reversible with eradication of the organism. The potential for *H pylori* infection to mimic the clinical and biochemical picture of Zollinger-Ellison syndrome was illustrated in a report [93]; thus, careful search and eradication of *H pylori* are mandatory in any patient with peptic ulcer disease before one considers a diagnosis of gastrinoma.

The most common cause of hypergastrinemia is gastric atrophy. [94] Gastric acid is the primary inhibitor of gastrin release; therefore, its absence leads to uninhibited secretion of the hormone with concomitant hyperplasia of antral G cells and hypergastrinemia as observed in pernicious anemia. [95], [96], [97], [98], [99], [100], [101] and [102] Up to 75% of patients with pernicious anemia have substantial hypergastrinemia. The gastrin levels in these patients can approximate those found in patients with gastrinoma, reaching values greater than 1000 pg/mL. Chronic atrophic gastritis and gastric carcinoma are two other conditions associated with hypochlorhydria or achlorhydria and hypergastrinemia. [98] Additional important causes of hypergastrinemia are outlined in Table 67-2.

To differentiate among the many causes of hypergastrinemia, various provocative tests [103], [104], [105], [106], [107], [108], [109], [110] and [111] have been developed (Table 67-3). The *secretin test,* initially developed by Isenberg and associates, [105] has proved to be the easiest and most reliable study to perform. Secretin can induce a significant early increase (albeit low) of serum gastrin levels in a few healthy persons and in patients with common duodenal ulcer disease, [110] but rarely in persons with achlorhydria. [111] The mechanism by which secretin stimulates release of gastrin from endocrine neoplasms is unclear. [70], [112], [113] and [114]

TABLE 67-3 Gastrin Provocative Testing

Unfortunately, naturally occurring purified porcine secretin is no longer available in the United States. Both porcine secretin and human synthetic secretin have been synthesized and are being considered by the United States Food and Drug Administration for clinical use. A randomized controlled crossover study of six patients with confirmed gastrinoma found that the synthetic secretin preparations could be used to confirm the presence of a gastrinoma. [115] Based on several studies composed of small numbers of patients, various criteria for establishing the positivity of a secretin provocative test have been proposed, including increases in serum gastrin of 100 pg/mL, 200 pg/mL, or 50% above basal levels. Frucht and colleagues [116] observed that the secretin test was found to be positive in 87% to 93% of all patients with

Zollinger-Ellison syndrome (80 patients) and proved to be more advantageous than the calcium infusion study (see later) because of its greater sensitivity, shorter sampling time, ease of performance, and lack of side effects. The parameter recommended by these authors for establishing a diagnosis of Zollinger-Ellison syndrome was an increase in serum gastrin of 200 pg/mL or greater. The false-positive secretin test results reported in other studies [116], [117] were not seen with these criteria. These same criteria may also apply to the synthetic secretin preparation, but this needs to be confirmed in a large-scale trial.

The less commonly employed provocative test is the *calcium infusion study.* More than 80% of patients with gastrinoma demonstrate an increase in gastrin levels of more than 400 pg/mL during the third hour of calcium infusion. As outlined earlier, lack of specificity, diminished sensitivity, and the potential adverse effects of administering intravenous calcium have made this provocative test less useful than the secretin test in diagnosing Zollinger-Ellison syndrome. It is usually reserved for patients with a negative secretin test in the presence of gastric acid hypersecretion and a strong clinical suspicion of gastrinoma. [116]

Standard meal studies have been employed to distinguish hypergastrinemia of gastric origin (as in G-cell hyperplasia or hyperfunction) from that of pancreatic origin (gastrinoma). Patients with Zollinger-Ellison syndrome may have less than a 50% increase in serum gastrin in response to a meal, and a postprandial increase in serum gastrin of more than 100% is characteristic of G-cell hyperplasia or hyperfunction. However, the studies of Frucht and associates [118] suggest that 50% of patients with Zollinger-Ellison syndrome have a greater than 50% postprandial increase in serum gastrin.

The net effect of hypergastrinemia is increased gastric acid secretion; thus, it is not surprising that a (BAO) of 15 mEq/h or greater is found in as many as 90% of patients with gastrinoma. [54], [86], [119], [120] and [121] However, 12% of patients with common duodenal ulcer disease can manifest similar levels of acid secretion. BAO of greater than 5 mEq/h is found in 55% of patients with Zollinger-Ellison syndrome, even after surgery. [119] The widespread use of potent anti secretory agents such as the PPIs further accentuates this problem. In fact, a study by Corleto and colleagues [9] suggests that the use of PPIs may mask Zollinger-Ellison syndrome, thus leading to delay in establishing this diagnosis. To enhance the sensitivity of the gastric secretory studies, *maximal acid output* (MAO) values may also be measured. Patients with gastrinoma are presumed to have near-maximal secretion of gastric acid even under basal conditions; thus, the acid secretory response to exogenously administered secretagogue (pentagastrin) is diminished. Accordingly, a ratio of BAO to MAO greater than 0.6 is highly suggestive of Zollinger-Ellison syndrome, [8] although a ratio of less than 0.6 does not exclude the diagnosis. [53], [86], [119] Unfortunately, pentagastrin is no longer available in the United States, making measurement of MAO virtually impossible. The other secretagogue used in gastric secretory studies was Histalog, which has been off the market for several years. Metz and Starr [122] observed that measurement of MAO had a small but significant benefit compared with measuring BAO alone.

There has been controversy over the utility of gastric analysis in view of the sensitivity and specificity of serum gastrin values with or without secretin stimulation. Investigators at the NIH performed the most exhaustive analysis to date of the role of gastric secretory studies in Zollinger-Ellison syndrome. [123] They prospectively examined 235 patients and reviewed 984 cases in the literature in an effort to delineate the potential role of gastric secretory studies in determining a diagnosis, clinical expression, and tumor extent. These authors observed that diarrhea, esophageal stricture, and pyloric scarring were associated with a higher BAO. It did not appear that the level of acid hypersecretion correlated with other clinical characteristics including tumor extent or size, primary tumor location, or other signs and symptoms in patients with gastrinoma. A summary of the recommendations made for measurement of gastric acid secretion in patients with Zollinger-Ellison syndrome is presented in Table 67-4. Under optimal circumstances, acid secretory studies should be part of the diagnostic evaluation in patients suspected of having gastrinomas. If the equipment needed for this test is not available, a simple basal gastric pH determination of greater than 3 essentially rules out the diagnosis of Zollinger-Ellison syndrome.

1. Secretory criteria for patients with no prior gastric acid–reducing surgery
   1.1. BAO ≥ 15 mEq/h
   1.2. BAO ≥ 18 mEq/h, in patients with known sporadic form
   1.3. No pH criterion useful for diagnosis
   1.4. BAC/MAC ratio ≥0.6
   1.5. Basal gastric volume >160 mL/h
   1.6. No MAO criterion useful
2. Patients with prior gastric acid–reducing surgery
   2.1 BAO ≥5 mEq/h
   2.2 No pH criteria useful for diagnosis
   2.3 BAC/MAC ratio ≥0.6
   2.4 BAO/MAO ratio >0.6
   2.5 Basal gastric volume >140 mL/h
   2.6 No MAO criterion alone useful
3. In hypergastrinemic patients, if pH >2, patient rarely has ZES
4. Acid measurements required only for 30 min

*All values refer to acid measurements made by nasogastric secretion with the patient not taking any gastric antisecretory medications (off proton pump inhibitors ≥1 wk; oral histamine H_2-receptor antagonists ≥48 h; intravenous histamine H_2-receptor antagonists ≥12 h).

BAC, basal acid H^+ concentration; BAO, basal acid output; MAC, maximal acid H^+ concentration; MAO, maximal acid output; ZES, Zollinger-Ellison syndrome.

Adapted from ref. 123.

TABLE 67-4 Recommendations for Measurement of Gastric Acid Secretion for the Diagnosis of Zollinger-Ellison Syndrome*

In summary, no single diagnostic test has a sensitivity or specificity of 100% in evaluating patients for Zollinger-Ellison syndrome. A fasting gastrin level greater than 1000 pg/mL and a BAO greater than 15 mEq/h in a patient with recalcitrant peptic ulcer is virtually diagnostic of Zollinger-Ellison syndrome. In the absence of this triad of findings, provocative testing may be required. Of these, the secretin stimulation test offers the greatest sensitivity and specificity.

TUMOR LOCALIZATION

Gastrinomas are notoriously difficult to localize. Case series of patients who eventually underwent surgery for Zollinger-Ellison syndrome before the 1990s noted the inability to identify the tumor in approximately 30% to 50% of patients, with more than 20% having metastatic disease at laparotomy. [35], [124], [125], [126], [127] and [128] More recent series, perhaps reflecting improvements in preoperative imaging modalities, have localized tumor in 85% of cases, although metastatic disease is frequently present (~50% of patients). Early intervention with resection of a localized tumor presents the only opportunity to cure this disease. [127], [128], [129] and [130] In addition, preoperative detection of metastatic disease will prevent unnecessary surgery. Accordingly, every effort should be made to localize the tumor preoperatively. A diagnostic plan for localizing gastrinomas in patients with Zollinger-Ellison syndrome is depicted in Figure 67-2.

FIGURE 67-2. Diagnostic plan for localizing gastrinomas in patients with Zollinger-Ellison syndrome. *CT,* computed tomography; *MRI,* magnetic resonance imaging;

EUS, endoscopic ultrasound; *EGD,* esophagogastroduodenoscopy; *R/O,* rule out; *SASI,* selective arterial secretin stimulation.

Many approaches have been used for the purpose of localizing gastrinomas. These include abdominal ultrasonography, [131], [132] CT scanning, [132], [133], [134], [135] and [136] abdominal arteriography, [137], [138], [139] and [140] selective portal venous sampling for gastrin, [139], [141], [142], [143] and [144] selective arterial stimulation of secretin (SASI), [145], [146], [147] and [148] endoscopic ultrasonography (EUS), [149], [150], [151], [152], [153], [154] and [155] and, most recently, somatostatin receptor scintigraphy (SRS) with the somatostatin analog octreotide. [156], [157], [158] and [159] The most recent additions to the imaging techniques, SRS and EUS, have emerged as the most useful in preoperative staging algorithms (see Fig. 67-2). The sensitivity of these modalities in localizing primary and metastatic gastrinomas is outlined in Table 67-5.

STUDY	SENSITIVITY (%)	
	Primary Gastrinoma	Metastatic Gastrinoma
Ultrasound	21–28	14
Computed tomography	35–59	35–72
Selective angiography	35–68	33–86
Portal venous sampling	70–90	N/A
Selective arterial stimulation of secretin	55–78	41
Magnetic resonance imaging	30–60	71
OctreoScan	67–86	80–100
Endoscopic ultrasound	80–100	N/A

TABLE 67-5 Sensitivity of Imaging Studies in Zollinger-Ellison Syndrome

Ultrasonography: Transcutaneous, Endoscopic, and Intraoperative

In general, transcutaneous abdominal ultrasonography is not considered useful for the diagnosis of gastrinomas. Most gastrinomas are small; therefore, sensitivity of this test is quite low, as demonstrated in a prospective study [160] in which transcutaneous ultrasonography was able to localize tumors in only 20% of patients later shown by surgery to have gastrinomas. Nevertheless, if a lesion is seen, it is likely to represent gastrinoma; thus, the specificity of ultrasonography was 90% to 100% in this prospective series.

EUS has facilitated high-resolution imaging of the pancreas, permitting delineation of structures smaller than 5 mm. Because the accuracy of this procedure in diagnosing small pancreatic carcinomas approaches 100%, [161], [162] it is not surprising that this diagnostic modality is useful in localizing small neuroendocrine tumors of the pancreas as well. [149], [150], [151], [152], [153], [154] and [155] Rösch and associates [152] examined 37 patients with surgically confirmed endocrine neoplasms of the pancreas (39 tumors), in whom preoperative ultrasonography and CT scan of the abdomen yielded negative findings. Thirty-two of the tumors were diagnosed by EUS (82% sensitivity), with a specificity of 95%. Tumors varied in diameter between 0.5 and 2.5 cm. However, all seven of the gastrinomas in this series were located in the pancreas, and none were located in extrapancreatic sites, such as the duodenum.

The University of Michigan group used EUS as a primary diagnostic tool to localize neuroendocrine tumors and evaluated more than 82 patients in a prospective study. [163] In their initial prospective study of 16 patients, Thompson and colleagues [154] observed that the sensitivity of a negative EUS result in predicting an extrapancreatic gastrinoma was 100%. Subsequently, their series was expanded to include 75 patients with neuroendocrine tumors with operative confirmation of the EUS findings. In 36 patients with suspected gastrinoma, these investigators confirmed a 100% accuracy to localize pancreatic lesions and to guide the surgeon to an extrapancreatic primary tumor, findings supporting the contention that EUS was invaluable in the preoperative evaluation of the patient with Zollinger-Ellison syndrome. [163] An early EUS localization strategy was found to be highly cost effective (and cost saving) when this group compared this approach with the one used before the introduction of EUS, which primarily used angiographic and venous sampling techniques. [164]

The sensitivity of EUS in detecting gastrinomas has been noted by other investigators. [155] In a study of 22 patients with Zollinger-Ellison syndrome, the sensitivity of EUS was 50% for duodenal wall tumors, 75% for pancreatic tumors, and 62.5% for tumor-bearing lymph nodes. These investigators observed that conventional endoscopy had a sensitivity of 40% for detecting duodenal wall lesions; thus, EUS has small incremental utility over endoscopy to localize duodenal wall lesions. Zimmer and colleagues [165] reported a similar experience, with EUS localizing 11 of 14 (79%) of all gastrinomas in 10 patients with Zollinger-Ellison syndrome. Of note, EUS localized 89% of the pancreatic lesions, and the results of conventional endoscopy are not reported. At the University of Michigan, a routine endoscopy (with a pediatric colonoscope) is performed to look for duodenal submucosal tumors, and EUS is primarily used to examine the pancreas.

Other centers without EUS experience have reported the utility of intraoperative ultrasonography, which appears to be useful in the localization of small gastrinomas. [166], [167] and [168] Tumors not detected by the surgeon through manual inspection may be imaged using this technique, with a sensitivity of 95% by experienced operators. [168]

Intraoperative Transillumination

Frucht and associates [169] prospectively examined the efficacy of intraoperative endoscopic transillumination for detecting duodenal gastrinomas in 26 patients. These investigators detected duodenal gastrinomas with 83% sensitivity, as compared with the sensitivity of the combined preoperative studies (25%) or intraoperative ultrasonography and surgical palpation (42%). This technique remains part of surgical protocols for the management of Zollinger-Ellison syndrome. [37]

Computed Tomography

The diagnostic accuracy of CT scanning reflects continued innovation in techniques and instrumentation. A prospective study [136] examined the value of CT in localizing tumors in 61 consecutive patients with gastrinoma. For extrahepatic gastrinoma, CT scanning with intravenous contrast had a specificity of 95%, a sensitivity of 59%, a positive predictive value of 96%, and a negative predictive value of 54%. For gastrinomas metastatic to liver, CT scanning had a specificity of 98%, a sensitivity of 72%, a positive predictive value of 93%, and a negative predictive value of 90%. Tumor size and location appear to be the key determinants in the successful diagnosis of gastrinoma by CT scanning. Approximately 80% of tumors in the pancreas and tumors larger than 3 cm are detected. Only 40% of extrapancreatic lesions and no tumors measuring less than 1 cm in diameter are detectable by CT scanning. The sensitivity and specificity of CT do not appear to be enhanced by performing dynamic studies. Whether helical techniques have led to improved sensitivity remains uncertain, although an initial report (requiring further comparative study) has reported 82% sensitivity for tumor localization. [170]

Angiography

Diagnostic innovations have limited angiography to selected patients without successful imaging by SRS or EUS. Maton and colleagues [140] prospectively examined the role of selective angiography in the diagnosis of 20 consecutive patients with gastrinoma. For hepatic gastrinoma, angiography had a specificity of 100% and a sensitivity of 86%, with a positive predictive value of 97% and a negative predictive value of 53%. In combination with CT scanning, 100% of the hepatic tumors were detected. Extrahepatic lesions were not as easily detected, inasmuch as CT scanning detected only 57%, angiography 70%, and the combination 73%, with a false-positive rate of 7%. This study suggests that after a CT scan, selective abdominal angiography will detect an additional 28% of patients with liver lesions and an incremental 16% of patients with extrahepatic tumors. Unfortunately, 24% of the extrahepatic lesions will be missed even when both tests are used. Nevertheless, angiography was found to be a useful adjunct to CT scanning in patients with Zollinger-Ellison syndrome who undergo surgery. This same group [171] subsequently reported less enthusiastic experience with CT (31% sensitivity for primary tumors, 42% for liver lesions) and angiography (28% and 62%, respectively). The reduced sensitivity likely reflects both increased numbers of patients referred with negative outside CT scans and angiograms and the ability of newer imaging modalities such as SRS to detect small lesions missed in the past.

Portal Venous Sampling

Before SRS and EUS, under the best circumstances the combination of ultrasonography, CT scanning, and angiography failed to localize tumors in up to 50% of the

patients with biochemical evidence of Zollinger-Ellison syndrome. Thus, percutaneous transhepatic selective portal venous sampling for gastrin was developed as a means to improve the diagnostic yield. [139], [141], [142], [143] and [144] There is general agreement with the relative sensitivity of this test in identifying the presence of gastrinomas (70% to 90%), but there is considerable debate over its specificity, with values ranging from 33% to 90%, depending on the series. Despite the potential usefulness of the selective venous sampling technique, its complexity and invasiveness limit its availability to only a few specialized centers, where it has largely been replaced by SASI; thus, it is not recommended for the evaluation of Zollinger-Ellison syndrome on a routine basis.

Selective Arterial Secretin Injection

SASI has been added to the growing list of modalities used for localizing gastrinomas. [145], [146], [147] and [148] For this study, initially described by Imamura and colleagues, [145] secretin (30 U) is injected selectively into the celiac, proximal splenic, gastroduodenal, proper hepatic, and superior mesenteric arteries, and blood samples for gastrin measurement are obtained simultaneously from the hepatic vein at short intervals (30, 60, 120, and 210 seconds). Given the result of emerging imaging modalities such as EUS and SRS, it appears that SASI will be employed infrequently and will be most helpful when imaging data are conflicting. [172]

Magnetic Resonance Imaging

MRI [173], [174] has been evaluated for the detection of both primary and metastatic gastrinoma, and initial results appear similar to those of CT scanning. In a prospective study, the NIH group found that MRI, using short-time inversion-inversion recovery sequences, was superior to CT for the detection of liver metastases (sensitivity, 71% versus 42%). MRI was equivalent to CT in detection of primary lesions, leading this group to suggest that it should replace CT as the cross-sectional imaging test of choice. The ability of SRS to localize occult liver metastases leaves the choice between CT and MRI a consideration of availability and local expertise.

Somatostatin Receptor Scintigraphy

Many gastrinomas express somatostatin receptors. Therefore, targeted nuclear scanning after injection of an isotopic analog of somatostatin—either iodine 123–Tyr3-octreotide or indium 111 (^{111}In)–diethylenetriamine pentaacetic acid–Dphe [1]-octreotide [156], [157], [158] and [159], [165], [171], [175]—has emerged as a readily available, noninvasive test to localize gastrinomas with a sensitivity and specificity of greater than 75%. The indium analog (^{111}In-pentetreotide) is marketed as OctreoScan 111 (Mallinckrodt, St. Louis, MO). In prospective studies, [174] the NIH group found that the scan was more sensitive than transcutaneous ultrasonography, CT, MRI, and angiography for localizing both primary and metastatic lesions, with sensitivities of 58% and 92%, respectively. The efficacy of EUS and SASI was not directly compared with SRS with regard to the ability to localize primary lesions. Consistent with these encouraging results, Cadiot and colleagues [175] observed that OctreoScan 111 scintigraphy was very useful in detecting small duodenal lesions (3 mm) and peripancreatic lymph nodes. In a review of 122 patients who underwent SRS to localize primary and metastatic gastrinoma lesions, the NIH group noted that SRS altered management in 47% of patients, with its greatest impact in the detection of liver metastases. [176] The investigators concluded that SRS should become the initial imaging modality of patients with gastrinoma. Among patients undergoing surgery for cure at the NIH, SRS was the most accurate preoperative imaging study, yet it still missed one third of all lesions found at surgery. [177] The ability of SRS to detect tumors was related to size, because only 30% of tumors 1.1 cm or smaller were imaged, compared with 64% of those 1.1 to 2.2 cm and 96% of those larger than 2 cm. Duodenal primary tumors were those most commonly missed by SRS. This same group also noted the limitations of SRS with regard to specificity; false positive results occurred in up to 10% of cases. [178] Knowledge of those clinical situations leading to false-positive results is essential for optimal interpretation of the OctreoScan 111, demonstrating the value of operator experience for all relevant diagnostic studies in the patient with Zollinger-Ellison syndrome.

In summary, surgical cure of Zollinger-Ellison syndrome requires accurate tumor localization. To avoid unnecessary laparotomy, OctreoScan 111 in combination with CT or MRI should exclude metastatic disease. If no metastatic disease is found, the approach to localize disease in the peripancreatic region is governed by local expertise and surgical experience. EUS (with upper gastrointestinal endoscopy to evaluate for duodenal tumors) appears to be the most cost-effective modality to evaluate the pancreas and duodenum, whereas SASI remains an alternative if EUS is not available. SASI also may detect occult metastasis and may assist in the interpretation of equivocal examinations. In other referral centers, the endocrine surgeon may go directly to exploration with intraoperative ultrasonography or transillumination. In the management of Zollinger-Ellison syndrome, it is clear that a team approach to localization and surgical exploration appears essential to ensure curative resection.

THERAPY

When considering the therapy of patients with Zollinger-Ellison syndrome, it is important to entertain two objectives: control of gastric acid hypersecretion and treatment of the malignant neoplasm. The emphasis placed on each of these objectives has shifted since the 1970s. Initially, total gastrectomy appeared to be the only alternative for effective treatment of the potentially lethal ulcer disease, and less attention was directed at tumor excision because many of these patients died of ulcer complications long before their tumors became problematic. The availability of highly effective antisecretory therapy such as histamine H_2 blockers and H^+, K^+-ATPase antagonists led to a significant reduction in mortality related to the complication of acid peptic disease. Under these circumstances, it has become increasingly apparent that the major cause of morbidity and mortality in Zollinger-Ellison syndrome is widespread metastatic disease [179]; thus, early tumor detection and excision have assumed primary importance.

Medical Therapy

The primary aim of medical therapy in Zollinger-Ellison syndrome is control of gastric acid hypersecretion. The development of histamine H_2-receptor antagonists was a major breakthrough toward this aim. Cimetidine, the first of these agents, proved very efficacious in controlling acid hypersecretion and prompted ulcer healing and symptom improvement in more than 80% of patients with Zollinger-Ellison syndrome treated on a short-term basis. [180] /SUP>, [181] and [182] However, over a longer term, these patients often required progressive increases in the frequency and dose of medication. Although H_2-receptor antagonists with greater potency and duration of action have been useful in treating patients with gastrinoma, [183], [184], [185], [186], [187], [188] and [189] an important newer class of drugs made available for the treatment of Zollinger-Ellison syndrome is the PPI, of which omeprazole was the first. [190], [191] The efficacy of these agents in inhibiting acid secretion, relieving dyspeptic symptoms, and promoting ulcer healing in patients with gastrinoma has been established. [192], [193], [194], [195] and [196]

Although earlier studies suggested that the doses of omeprazole required for effective inhibition of acid secretion in patients with gastrinoma were significantly higher than in patients with standard peptic ulcers (Table 67-6), a study by Metz and associates [197] suggests that the currently recommended maintenance dose of omeprazole (~60 mg/d) used in these patients is too high. Accordingly, a gradual reduction of omeprazole dose is recommended once the initial dose required for adequate control of gastric acid hypersecretion (as described earlier) has been achieved. More recently, the NIH group prospectively examined the effectiveness of low doses of omeprazole (20 mg/d) as initial therapy in patients with gastrinoma, [198] and based on their results, the investigators continue to recommend initiating omeprazole therapy at a dose of 60 mg/d. This dose allows rapid control of gastric acid secretion, thus minimizing complications related to peptic diathesis. Once an adequate maintenance dose is achieved, tapering the medication is then indicated while following symptoms and acid output. Lansoprazole has also been found to be safe and effective in the treatment of patients with gastrinoma for up to 10 years. [196] Tachyphylaxis is observed less frequently with PPIs than with H_2 blockers. One concern with long-term omeprazole therapy has been its potential for inducing enterochromaffin cell hyperplasia. A prospective study following 40 patients with Zollinger-Ellison syndrome who were treated with omeprazole for a mean of 29 months (6 to 51 months) reported no evidence of hematologic, biochemical, or gastric toxicity. [199], [200] Their marked potency, prolonged duration of action, and safety profile make PPIs the treatment of choice for peptic disease in patients with Zollinger-Ellison syndrome. The intravenous formulation of pantoprazole has become available and appears to be very effective in the treatment of patients with Zollinger-Ellison syndrome. [201]

AGENT	MEDIAN DOSES g/d (range)
H_2 Blockers	
Cimetidine	3.6 (1.2–12.6)
Ranitidine	1.2 (0.45–6)
Famotidine	0.25 (0.05–0.8)
H^+, K^+-ATPase Inhibitors	
Omeprazole	60 mg/d
Lansoprazole	60–90 mg/d

TABLE 67-6 Median Dose of Antisecretory Agents Used in Patients with Gastrinoma

Somatostatin is a known peptide inhibitor of gastric acid secretion [202] and gastrin release. [203] The biochemically stable analog of somatostatin, octreotide, has been used with varying success in patients with gastrinomas. [202], [203], [204], [205] and [206] At present, however, these compounds are available only in injectable form; thus, they are rarely used as first-line agents in the treatment of patients with Zollinger-Ellison syndrome. The long-acting depot formulation of octreotide has become available and shown in preliminary fashion to be beneficial in the treatment of neuroendocrine tumors of the gut. [207] Future studies will clarify the role of this compound in the therapy of patients with gastrinoma.

Surgical Therapy

Before the advent of potent antisecretory agents, total gastrectomy was the treatment of choice in patients with gastrinoma. Although initial operative mortality was very high (15% to 20%), [208] total gastrectomy offered better long-term survival than did more conservative approaches. [11] Increased experience and earlier diagnostic testing have reduced the operative mortality rate associated with total gastrectomy to less than 5%. [209] Nevertheless, the newer gastric antisecretory agents have obviated the need for total gastrectomy, and at present this procedure should be considered only in the rare patient with nonresectable gastrinoma in whom aggressive medical therapy has failed or in those patients who cannot take oral medications.

Another operation applied in the past as an adjunct to medical therapy in patients with Zollinger-Ellison syndrome is proximal gastric vagotomy. [210], [211] The rationale for this operation was the high failure rate with H_2 antagonists. The advent of PPIs may obviate the need for the routine use of this procedure.

As discussed earlier in this chapter, the hypercalcemia found in patients with MEN I may be associated with a gastric acid hypersecretory state that is refractory to medical therapy. A previous report [212] demonstrated that in addition to lowering both BAO and fasting hypergastrinemia in these patients, parathyroidectomy facilitates the management of peptic disease with H_2 antagonists. Although the availability of H^+,K^+-ATPase inhibitors may make this operation unnecessary, parathyroidectomy may be a surgical option for treatment of patients with gastrinoma with MEN I and hyperparathyroidism who are refractory to medical management.

Clearly, the appropriate surgical approach for therapy of Zollinger-Ellison syndrome today is curative resection of the neoplasm. Although initial series reported a cure rate of less than 10% after tumor resection, present data indicate that cure may be possible in as many as 30% of cases. Norton and colleagues [37] published the largest experience of surgical intervention in patients with gastrinoma. This group followed 151 consecutive patients (123 with sporadic cases, 28 with MEN I) who underwent surgery between 1981 and 1998. Thirty-four percent of patients with sporadic gastrinoma were free of disease at 10 years. Unfortunately, none of the patients with MEN I were disease free at the 10-year period. Moreover, the overall 10-year survival observed in these patients was 94%. These authors concluded that all patients with Zollinger-Ellison syndrome who do not have metastatic disease or MEN I should be offered surgical intervention for possible cure. Improved diagnostic capabilities have led not only to a reduction in unnecessary surgery on patients with metastatic disease, but also to the identification of extrapancreatic neoplasms with increasing frequency. [33], [34] and [35], [127], [128], [212], [213], [214] and [215] Studies have reported the incidence of extrapancreatic lesions to be as high as 66% and resection of these lesions to be associated with a high likelihood of long-term cure. [126], [216]

Patients with gastrinoma with MEN I represent a particularly difficult problem to the surgeon. Because of the multicentric nature and extrapancreatic location of tumors in MEN I syndrome, [217], [218], [219], [220], [221] and [222] curative resection is virtually impossible. [143], [187], [209], [223], [224] Some surgeons advocate an aggressive approach whereby all patients without evidence of hepatic metastasis are explored with removal of all duodenal lesions found by duodenotomy, enucleation of all tumors found in the head of the pancreas, and distal pancreatectomy. [219], [222] The rationale for this approach is to effect a potential cure and, failing that, to prevent the development of future pancreatic neoplasms. The value of such an aggressive approach remains to be established. If a single tumor is found during the preoperative evaluation, however, an attempt at surgical resection should be undertaken [225] because larger tumors may be associated with a higher likelihood of metastasis. The role of surgery in patients with gastrinoma and MEN I was explored in 81 consecutive patients. [226] All patients with limited disease (single tumor between 2.5 and 6 cm, $n = 17$) and advanced disease (two or more lesions >6 cm, $n = 31$) underwent exploratory laparotomy with tumor enucleation or resection. Patients with tumors smaller than 2.5 cm ($n = 17$) and those with liver metastases ($n = 8$) were observed. Approximately 40% of patients had advanced disease without metastases. Duodenal lesions were found in 80% of patients with Zollinger-Ellison syndrome and MEN I. The authors observed that tumors could be removed in patients with advanced disease with no mortality and a complication rate similar to that observed in patients with limited disease. An additional important observation was that patients with advanced tumor who underwent resection had the same survival rate as patients with limited disease and those without identifiable tumor. When looked at as a group (nonresected and resected), 15-year survival was between 89% and 100%, whereas for patients with metastatic disease, it was only 52%. The authors of this study concluded that surgical resection should be performed in patients with gastrinoma and MEN I with advanced localized disease. The ideal type of operation for this group of patients was not resolved by this study, but the authors did not recommend pancreaticoduodenectomy or distal pancreatectomy on a routine basis. Although a subgroup of patients with gastrinoma may have an aggressive course warranting a Whipple procedure, the tools to predict this subgroup of patients have not been developed (see later).

Operative management of Zollinger-Ellison syndrome requires careful and detailed mobilization and exploration of the entire pancreas duodenum (duodenotomy) and surrounding areas. [215], [219], [222] Any tumors that are found in the region of the pancreatic head should be enucleated, and tumors located elsewhere should be resected with great care. In the event that a tumor is not found at surgery, distal pancreatectomy should be avoided because more then 80% of the gastrinomas are located within the gastrinoma triangle. [33] If a lesion is found in the head of the pancreas, a Whipple procedure should be considered with great care because the mortality of this surgical procedure may outweigh its potential benefits. However, the decision may be facilitated if complete tumor excision is not possible by a less invasive procedure. [227]

Pisegna and colleagues [228] examined the effects of curative gastrinoma resection on gastric secretory function. They observed that the trophic effects of long-standing hypergastrinemia are largely reversible within a short period, with 50% and 75% decrease in MAO and BAO, respectively, within 3 to 6 months after curative resection. However, these observations also indicate that acid secretion must be monitored even after curative resection because as many as 55% of patients at 3 to 6 months and up to 67% at 4 years after gastrinoma resection remained mild hypersecretors requiring low doses of antisecretory drug.

Therapy for Metastatic Gastrinoma

Metastatic disease occurs in 25% to 40% of patients with gastrinoma and at present is the most common source of morbidity and mortality in this disease. One major difficulty in evaluating the effectiveness of the various therapeutic regimens is that most clinical trials have examined small numbers of patients. Antineoplastic chemotherapy has been of limited usefulness in Zollinger-Ellison syndrome. Combined treatment with streptozotocin, 5-fluorouracil (5FU), and doxorubicin (Adriamycin) has provided the most promising results. In one series of 42 patients with metastatic islet tumors of various types, [229] initial response rates of 63% and complete response rates of 33% were reported with this regimen. However, gastrinomas comprised a small percentage of the cases studied, and the response to chemotherapy may have varied according to the cell type of the tumor. In another study using a similar treatment regimen, Bonfils and colleagues [230] reported disappearance of the gastrinoma as assessed by imaging studies in 20% of their patients with Zollinger-Ellison syndrome with metastatic tumors.

A prospective study [231] by the investigators at the NIH provided a more sobering perspective on treatment of metastatic gastrinoma with chemotherapy. Ten consecutive patients with the disorder were treated with a regimen of streptozotocin, 5FU, and doxorubicin, and although a modest objective response was observed initially in four patients, the response lasted for less than 10 months. All the patients were considered as treatment failures thereafter, with no significant change in short-term survival, and many side effects of therapy, including nausea and vomiting (100%), alopecia (80%), proteinuria (40%), and leukemia (20%), were noted. At 10 months after initiation of therapy, all patients had evidence of tumor growth over the preceding 6 months. The question when to treat patients having metastatic gastrinoma with chemotherapy remains unanswered. Although an occasional patient with metastatic disease may survive up to 20 years, it appears that most will die within 5 years. Two approaches to treating these patients are to begin chemotherapy when the patient is symptomatic [229] or to reassess the patient 3 to 6 months after initial presentation and to use chemotherapy if the metastases are increasing in size. [232] Both approaches are based on studies including relatively small numbers of patients; thus, additional data are needed before firm recommendations can be given.

In view of the limited success of chemotherapy in the treatment of metastatic gastrinomas and neuroendocrine tumors of the pancreas in general (see Chapter 97), alternative therapeutic modalities

have been explored. As in the case of assessing the value of chemotherapy in patients with gastrinoma, the rarity of the disease coupled with the heterogenous group of patients studied and the variable natural history of these malignancies make the available data somewhat difficult to interpret.

Hormonal therapy with the stable somatostatin analog octreotide has been attempted in metastatic gastrinoma. [233], [234] Although early reports suggested a significant decrease in tumor size in response to octreotide, [235] subsequent studies have not been as encouraging. [236], [237] Tumor stabilization may be achievable, but this is unclear in the case of patients with gastrinoma. An interesting therapeutic alternative considered for treatment of neuroendocrine tumors has been octreotide radionucleotide therapy. [238] Although this option is in the very early stages of development, future studies may prove it beneficial for these patients.

Leukocyte interferon also has been used in the therapy of neuroendocrine tumors with reported success. [239], [240] and [241] Unfortunately, these encouraging results were not observed by Pisegna and associates [242] after examining the effect of human recombinant interferon-a in patients with Zollinger-Ellison syndrome. Smaller trials examining the efficacy of combining interferon with octreotide or 5FU have been published, [243], [244] but the limited number of patients evaluated makes these data difficult to interpret. In addition, there was a substantial increase in drug toxicity to interferon when this drug was combined with 5FU, making it a less desirable treatment option. [244]

Hepatic artery embolization is another treatment option considered for endocrine tumors that have metastasized to the liver. Ruszniewski and associates [245] examined the efficacy of chemoembolization with doxorubicin in 24 patients with metastatic tumors (5 patients with gastrinoma), and tumor stabilization was observed in 3 patients. More recently, Moertel and colleagues [246] treated 111 patients (with carcinoid and pancreatic endocrine tumors) with systemic chemotherapy in addition to chemoembolization and achieved an overall response rate of 80%. In view of the limited data available in patients with gastrinoma and the potential complications related to hepatic artery embolization, including abdominal pain, fever, nausea, and even death (<3%), [247], [248] this treatment should be reserved for treatment-refractory patients until further data are available.

Aggressive surgical resection (debulking) [249], [250], [251] and [252] has been performed successfully, with 5-year survival achieved in more than 75% of patients whose tumors were amenable to complete resection. [250] These data suggest a potential benefit of this surgical approach in patients who are experiencing severe refractory symptoms in which tumor distribution permits an attempt at resection (<10%).

Orthotopic liver transplantation also has been attempted as a therapeutic option in patients with hepatic metastases. The finding that these tumors are slow growing and are often unresponsive to aggressive medical management has served as the driving force behind this treatment modality. [253], [254] and [255] Unfortunately, the 4-year survival rate in patients with pancreatic neuroendocrine tumors was only 8% in the largest sample of patients examined. [255] Thus, orthotopic liver transplantation does not appear to provide an option for these unfortunate patients.

PROGNOSIS

Zollinger-Ellison tumors can be classified as either benign or malignant, [36], [256] based on a series of clinical characteristics. Of these characteristics, tumor extent and rate of tumor growth are the most important prognostic indicators for patients with gastrinoma. As reviewed by Maton and colleagues, [225] 5-year survival rates for all patients with gastrinoma vary between 62% and 75%, and 10-year survival rates vary between 47% and 53%. If one segregates the survival data according to extent of tumor present at the time of diagnosis, patients who have a negative laparotomy or who have all their tumor removed surgically have 5- and 10-year survival rates greater than 90%. In contrast, patients with tumors that were incompletely resected have 5- and 10-year survival rates of 43% and 25%, respectively. Weber and associates [36] critically evaluated 185 consecutive patients with Zollinger-Ellison syndrome and identified the criteria that determined metastatic rate and survival in these patients. These investigators confirmed that both benign and malignant categories of gastrinomas exist. The overall 10-year survival rates were 93% for patients with MEN I and 74% for patients without MEN I, somewhat better than rates reported previously, [225] but similar to results reported by Eriksson and colleagues. [257] It was also found by these investigators that there was no difference in survival for the following patients: those rendered disease free at surgery, those with tumor and persistent disease after surgery, and those with disease and negative exploratory laparotomy results. A poor clinical outcome is seen in female patients, in patients without MEN I, in patients with high serum gastrin levels, in patients with a large pancreatic tumor, and in patients with hepatic metastasis.

A detailed prospective analysis of the clinical course of 212 patients with Zollinger-Ellison syndrome [258] observed for a mean period of 15 years (range, 0.1 to 31 years) was published. Thirty-two percent of patients died (none of acid-related complications); 50% of them had gastrinoma-related causes of death. Gastrinoma-related causes of death are outlined in Table 67-7, with the most common being progressive cachexia associated with hepatic metastases (67%). Gastrinoma-related causes of death were more commonly observed in patients with the following characteristics: shorter disease duration; higher gastrin levels; pancreatic primary and large tumor; metastatic disease to lymph nodes, liver, or bones; and ectopic Cushing syndrome. When considering patients with hepatic metastases, tumor-related Cushing syndrome and development of metastases to bone were independent predictors of decreased survival time (Fig. 67-3). In addition to extent of hepatic metastases, the rate with which they developed was an independent indicator of poor prognosis. From this and other studies, [36] it appears that roughly 25% of patients with gastrinoma follow an aggressive pattern, with 10-year survival approaching 30%, whereas the remaining patients follow a more indolent course, with a 10-year survival of 95%.

CAUSE OF DEATH	NUMBER	PERCENTAGE (%)
Liver metastases with progressive inanition	22	67
Secondary PET syndrome	10	30
Liver metastases causing hepatic failure	4	12
Tumor-related embolism	2	6
Postoperative complications	1	3
Cardiac gastrinoma with arrhythmia	1	3

PET, pancreatic endocrine tumor.
Adapted from ref. 258.

TABLE 67-7 Related Causes of Death Related to Zollinger-Ellison Syndrome

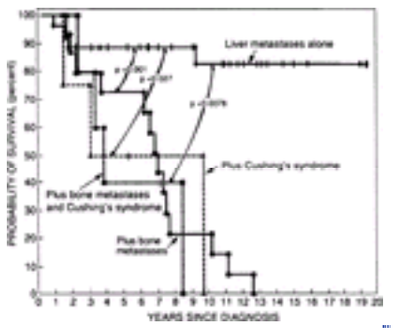

FIGURE 67-3. Impact of the presence of bone metastases, liver metastases, or ectopic Cushing syndrome, alone or in combination, on survival of patients with gastrinoma. (Adapted from ref. [258].)

Gibril and colleagues [259] performed a similar type of prospective study on the natural history of gastrinoma in 57 patients with MEN I. These patients were followed-up for a mean period of 8 years, with 23% having or developing liver metastases and 14% having a tumor with aggressive growth, defined as tumor growth of 25% volume increase or more per month or the appearance of any new lesion. In terms of survival, 5% of the study group, 23% of patients with hepatic metastases, and 38% of patients with aggressive tumor growth died during the study period. Factors predictive of aggressive gastrinoma growth were identified by the authors and are summarized in Table 67-8. Ultimately, further elucidation of the molecular determinants important in tumor development in MEN I coupled with recognition of clinical features predictive of poor outcome may allow earlier identification and intervention in patients prone to developing aggressive disease.

Clinical	
Age at MEN I diagnosis (y)	<35 ($P = 0.021$)
Age at ZES onset (y),	≤27 ($P = 0.043$)
Age at ZES diagnosis (y),	≤33 ($P = 0.032$)
Duration of ZES before diagnosis (y),	<2.1 ($P = 0.021$)
Laboratory	
Fasting gastrin levels,	≥10,000 pg/mL ($P < 0.0001$)
Tumor Characteristics	
Pancreatic tumor size,	>3 cm ($P < 0.0001$)*
Presence of liver metastases	($P < 0.00001$)
Presence of bone metastases	($P = 0.0019$)
Presence of gastric carcinoid	($P = 0.024$)

*Data from 41 patients who underwent abdominal exploration.

MEN I, multiple endocrine neoplasia type I; ZES, Zollinger-Ellison syndrome.

Adapted from ref. 259.

TABLE 67-8 Factors Predictive of Aggressive Growth of Gastrinomas in Patients with Multiple Endocrine Neoplasia Type I

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CHAPTER 68

Robert M. Genta

GASTRITIS AND GASTROPATHY

CLINICOPATHOLOGICAL APPROACH TO GASTRITIS

Gastritis, simply defined as the inflammation of the gastric mucosa, is a condition, not a disease. With rare exceptions (e.g., lymphocytic gastritis and the extremely rare phlegmonous gastritis), inflammation of the gastric mucosa per se does not produce signs or symptoms; its complications do. Thus, clinicians rarely search for gastritis. In dyspeptic patients with indications for endoscopy, biopsies from the stomach are often obtained to determine the patient’s *Helicobacter pylori* status. If an appropriate set of gastric mucosal specimens is collected and properly examined, valuable information in addition to the often implicit question “is there *H pylori*?” may be obtained: the type, severity, and distribution of gastritis and perhaps other causes of the gastric inflammation. This chapter is a discussion of useful strategies gastroenterologists and pathologists can use to optimize the diagnosis of gastric diseases.

Sydney System

The discovery of *H pylori* in 1982 coincided with a new trend in medicine: the birth of the expert group, whose task is to sift through the best available evidence (hence the term *evidence-based medicine*) and attempt to come to a consensus with regard to treatment strategies (*clinical guidelines*) or classifications of disease. Before the 1990 World Congress of Gastroenterology in Sydney, Australia, such a group of European gastroenterologists and pathologists set out to create a flexible matrix for the classification of gastritis. The resulting *Sydney System* had both endoscopic and histological divisions. [1] The former was met with general indifference and faded into oblivion. The latter unleashed passions that were distributed largely along geocultural lines: enthusiasm in Europe, where the system was conceived; indignation in the Americas and Asia, where investigators felt excluded both politically (none were asked to participate in the expert group) and nosologically (the types of gastritis commonly seen in Asia and South America received limited attention). In spite of these operational shortcomings, the Sydney System described a framework useful to generate diagnoses and flexible enough to incorporate new ideas as they emerged. Four years after its introduction, the Sydney System was reapprised by a group of pathologists including a wider geographic and disease representation. This group established a terminology of gastritis and identified, defined, and attempted to resolve some of the problems associated with the original Sydney System. The Houston workshop resulted in what is known as the Updated

Sydney System, which is currently the most widely used and cited method for the classification of gastritis. [2] Table 68-1 summarizes the different entities classified in the Updated Sydney System.

TYPE OF GASTRITIS	ETIOLOGIC FACTORS	GASTRITIS SYNONYMS
Nonatrophic	*Helicobacter pylori* ↑ Other factors	Superficial Diffuse antral gastritis Chronic antral gastritis Interstitial-follicular Hypersecretory Type B
Atrophic		
Autoimmune	Autoimmunity ↑ *H pylori*	Type A Diffuse corporal Pernicious anemia-associated
Multifocal atrophic	*H pylori* Environmental factors	Type B, type AB Environmental Metaplastic Atrophic pangastritis Progressive intestinalizing pangastritis
Special Forms		
Chemical	Chemical irritation Bile NSAIDs ↑ Other agents	Reactive Reflux
Radiation	Radiation injury	
Lymphocytic	↑ Idiopathic ↑ Autoimmune mechanisms ↑ Gluten Drugs (ticlopidine) ↑ *H pylori*	Varioliform Celiac disease-associated
Noninfectious granulomatous	Crohn's disease Sarcoidosis Wegener granulomatosis Foreign substances	
Eosinophilic	↑ Idiopathic Food sensitivity ↑ Other allergens	Isolated granulomatous Allergic
Other infectious gastritides	Bacteria (other than *H pylori*) Viruses Fungi Parasites	Phlegmonous, syphilitic, etc. Cytomegalovirus Mucormycosis

TABLE 68-1 Classification of Chronic Gastritis Based on Topography, Morphology, and Etiology

To create a report as suggested by the Sydney System, appropriate biopsy specimens should be methodically evaluated and the findings synthesized (Fig. 68-1). Whereas gastroenterologists in some institutions have made a habit of obtaining mapped specimens from each patient who undergoes gastroscopy, others continue to take few and often topographically unidentified specimens. These diagnostic guidelines can be best followed by those gastroenterologist-pathologist teams who work together and communicate effectively.

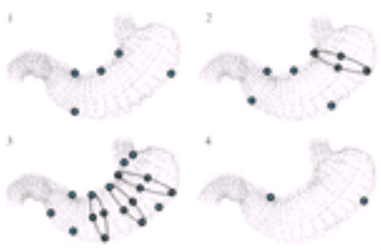

FIGURE 68-1. (**1**) The Updated Sydney System recommends a biopsy protocol that consists of five specimens, three on the lesser curvature and two on the greater curvature, from the antrum (*2*), the incisura angularis (*1*), and the corpus (*2*). (**2**) For protocols designed to assess the development of atrophy or endocrine cell hyperplasia, more extensive sampling from the corpus is required. (**3**) Extensive mapping protocols may be used for the detection of patchy conditions, such as the follow-up of mucosa-associated lymphoid tissue lymphomas. (**4**) In clinical practice, the most common sampling consists of one specimen from the antrum and one from the corpus. Although adequate to detect *Helicobacter pylori* gastritis, such a minimalist protocol does not permit the topographic definition of gastritis and is insufficient to establish the presence and extent of atrophy.

Biopsy Protocol

To obtain adequately representative samples for the classification of gastritis, the *biopsy protocol* depicted in Figure 68-1 is recommended. Biopsy specimens from the three compartments (antrum, incisura angularis, and corpus) should be separately identifiable when they are submitted to the laboratory. Proper orientation is indispensable for optimal histological evaluation; it may be accomplished either in the endoscopy suite when biopsy specimens are collected, or in the histopathology laboratory at the time of embedding. This latter option is generally preferable, unless endoscopy personnel are experienced and motivated to perform the precise and tedious work required to orientate minuscule fragments of fresh tissue properly.

To translate histopathological observations into well-defined topographic patterns, each feature is then graded using the standardized Visual Analogue Scale (Fig. 68-2). The final diagnosis issued should synthesize all individual evaluations, for example, " *H pylori* antrum-predominant gastritis" or "corpus-restricted atrophic gastritis without *H pylori* infection, suggestive of autoimmune gastritis."

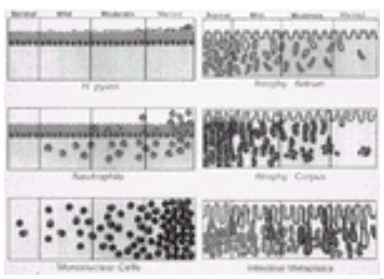

FIGURE 68-2. The Visual Analogue Scale is used to grade the five main components of gastritis (density of *Helicobacter pylori;* intensity of active and chronic inflammation; atrophy; and intestinal metaplasia).

The Updated Sydney System is also suitable for evaluating and diagnosing the special types of gastritis. A sample diagnosis might read "lymphocytic gastritis, corpus predominant, with *H pylori* infection." In the case of gastropathies, the Updated Sydney System is mainly useful for helping in the orderly assessment of the histopathological features of the mucosa. This applies even in the unlikely situation that the system's recommended set of five biopsy specimens is obtained from a patient with portal hypertension or watermelon stomach. However, in most such cases, attempting to grade each specimen individually is neither recommended nor necessary.

TOOLS TO DIAGNOSE AND CLASSIFY GASTRIC CONDITIONS

The different types of gastritides and gastropathies are characterized by various combinations of *histological changes,* many of which are expressions of immune, inflammatory, and adaptive responses common to several conditions. However, the presence, absence, and relative intensity of these responses provide important etiologic clues and are crucial in the categorization of the process. These histological changes can be viewed as the foundations for the terminology of gastritis, and some familiarity with them is indispensable to understand both the classification and the related manifestations of nonneoplastic gastric conditions.

Epithelial Degeneration

Surface epithelial degeneration is a nonspecific response to injury seen in all forms of gastritis. It is most conspicuous in chemical gastritis and *H pylori* gastritis. In *H pylori* gastritis, the intimate contact of bacteria with the surface cell membrane makes epithelial degeneration particularly prominent (Fig. 68-3). [3] Cell injury and necrosis can lead to erosions, which are seen endoscopically either as flat superficial lesions or as elevated lesions whose chronic nature is suggested by polypoid regenerative mucosa at the margins. The former are often the result of acute damage caused by drugs, bile reflux, or ischemia, whereas the latter are almost always associated with *H pylori* gastritis.

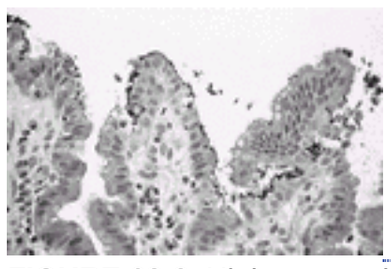

FIGURE 68-3. High-power view of antral epithelium with severe surface damage resulting from the adherence of large numbers of *Helicobacter pylori* organisms. Epithelial cells are depleted of mucin, flattened, and focally missing; the large hyperchromatic nuclei indicate increased regeneration.

Foveolar Hyperplasia

Elongation and increased tortuosity of gastric pits result from *hyperplasia of the foveolar cells,* a presumed adaptive response to increased cellular exfoliation from the surface epithelium. It can be viewed as a visual surrogate for increased epithelial cell turnover. Hyperplasia is accompanied by hyperchromatic nuclei and mitotic activity reaching an increased height of the pit and by other signs of cellular immaturity, such as mucin depletion and a high nucleocytoplasmic ratio. Marked foveolar hyperplasia is a prominent feature of chemical injury (Fig. 68-4A), [4] but lesser degrees are commonly seen in *H pylori* gastritis (Fig. 68-4B). [5]

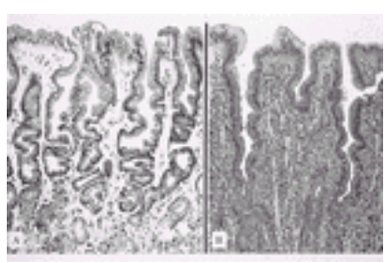

FIGURE 68-4. (**A**) Elongated, tortuous pits (foveolar hyperplasia) with minimal inflammation suggest chemical damage (bile or nonsteroidal antiinflammatory drug gastropathy). (**B**) In contrast, foveolar hyperplasia associated with inflammation of the lamina propria and surface epithelial damage is characteristic of *Helicobacter pylori* infection.

Hyperemia and Edema of the Lamina Propria

Mucosal hyperemia—often visible endoscopically—is considered to be an indicator of bile reflux gastritis, and a significant correlation has been found with the concentration of bilirubin in gastric juice. Histologically, marked edema of the lamina propria with minimal inflammation is a characteristic finding in bile gastritis. [4]

Neutrophilic Infiltration

The presence of *neutrophils* characterizes the “activity” in chronic gastritis (Fig. 68-5A). The cause is *H pylori* in most cases, but other infectious and inflammatory conditions (e.g., syphilis and Crohn’s disease [6]) may be responsible for the persistence of neutrophils, classically associated with “acute” inflammation. Neutrophils are found in virtually every patient with *H pylori* infection. [7], [8] The intensity of neutrophil infiltration may help to distinguish among the acute phase of infectious gastritis, *Helicobacter* gastritis with a particularly active component, and acute hemorrhagic gastritis resulting from chemical injuries (e.g., nonsteroidal antiinflammatory drugs [NSAIDs] or alcohol), in which inflammation is a minor component. Neutrophils disappear rapidly after successful eradication therapy; their persistence is a highly sensitive indicator of therapeutic failure. [7]

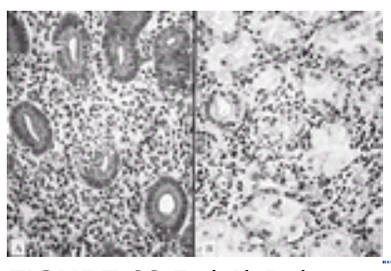

FIGURE 68-5. (**A**) Polymorphonuclear neutrophils in the lamina propria and infiltrating the foveolar epithelium are virtually pathognomonic of *Helicobacter pylori* gastritis. (**B**) A mixed infiltration of oxyntic glands may occur in *H pylori* gastritis, but also in autoimmune gastritis.

Eosinophilic Infiltration

Rare, scattered *eosinophils* may be present in the gastric lamina propria of healthy persons, particularly in underprivileged populations. Prominent eosinophilic infiltration of the gastric wall either may be part of the rare eosinophilic gastroenteritis or may represent a process confined to the stomach. In either case, the cause, suspected to have an allergic basis, is not known. Eosinophils are a major component in the responses to anisakiasis and may be a constituent of the granulomata that sometimes form around fragments of the helminths remaining in the gastric wall. [9] In adults with *H pylori* gastritis, there are usually small numbers of eosinophils. In contrast, children have been reported to have a greater eosinophilic component in the *H pylori*–infected gastric mucosa. [10] After eradication of the pathogen, eosinophils may increase for some time and then decline in parallel with mononuclear cells. [7]

Mononuclear Cell Inflammation

Infiltration of the lamina propria by lymphocytes, plasma cells, and small numbers of eosinophils and mast cells is a major feature of *H pylori* gastritis (Fig. 68-6A), except in areas of severe atrophy and metaplasia, in which the infiltrate tends to be sparse. When lymphocytes are seen infiltrating the surface or glandular epithelium, the possibility of lymphocytic gastritis should be considered. [11] In autoimmune gastritis, there is a diffuse infiltrate of mucosal plasma cells and

lymphocytes. [12] The latter are also present around and within oxyntic glands (Fig. 68-5B).

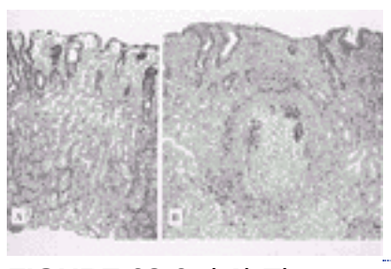

FIGURE 68-6. (**A**) The accumulation of mononuclear cells in the zone immediately subjacent to the surface epithelium pushes the oxyntic glands down, forming a thin band of inflammation limited to the upper portion of the mucosa, hence the term “superficial gastritis.” (**B**) Lymphoid follicles with germinal centers are found in almost all patients with *Helicobacter pylori* gastritis.

Lymphoid Follicles

Lymphoid follicles are rare in the stomach of healthy, *H pylori*–free adults. When an extensive biopsy protocol is used, lymphoid follicles or aggregates are found in virtually all patients with *H pylori* gastritis (see Fig. 68-6B). [13] In infected children and young adults, these entities may produce a distinctive nodularity in the gastric antrum, known endoscopically as *follicular gastritis.* [14] *H pylori* infection is the major determinant of gastric acquired mucosa-associated lymphoid tissue (MALT) and, therefore, a crucial factor in the origin of primary gastric B-cell lymphomas (MALT lymphoma). [15]

Atrophy

Gastric atrophy is defined as the loss of appropriate glands in a given gastric compartment [16]; that is, glands that are expected to be present in the portion of gastric mucosa under examination (e.g., oxyntic glands in the mucosa of the corpus) have been replaced by tissues not normally found there (Fig. 68-7). More recently, the narrower definition of *loss of specialized cells* has been proposed. [17] Whenever the gastric mucosa is damaged, irrespective of the mechanism or cause, it may either regenerate or return to normal (*restitutio ad integrum*), or it may undergo adaptive reparative processes leading to the replacement of the native mucosa with other structures. [18] Destroyed native glands may be replaced by fibroblasts and extracellular matrix, by glands of “pyloric” appearance (*pseudopyloric metaplasia*), or by an intestinal-type epithelium (*intestinal metaplasia*). During chronic *H pylori* infection, all these types of repair occur, the respective proportion of each probably modulated by environmental, genetic, and bacterial factors. [19] Widespread atrophy also occurs in autoimmune gastritis, as a consequence of the immune-mediated glandular destruction of the oxyntic mucosa. Atrophic foci found in stomachs with evidence of chemical gastropathy are probably the result of ulcer repair.

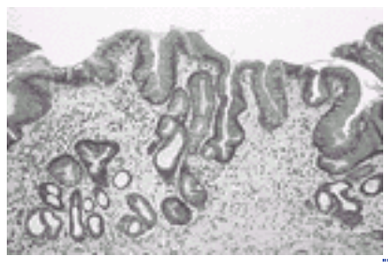

FIGURE 68-7. Biopsy specimen from the gastric corpus showing complete atrophy. No oxyntic glands are present, and the few foveae and mucous glands are widely separated by collagen.

Intestinal Metaplasia

Intestinal metaplasia is the replacement of the mucous cells that line the normal gastric mucosa with an epithelium similar to that of the small intestine (Fig. 68-8). The different types of intestinal metaplasia have been variously classified as follows: complete versus incomplete; types 1, 2a, and 2b; and types I, II, and III, based on their morphology and content in sulfomucins. [20] In general, the greater the extension of metaplasia in a stomach, the greater the proportion of the incomplete types. [21] Intestinal metaplasia is a virtually constant component of atrophic gastritis, and is found more frequently in the stomach of patients with *H pylori* gastritis. [22], [23] Small antral foci of intestinal metaplasia are also frequently found in bile reflux gastropathy, both in postoperative and in intact stomachs. [4], [24]

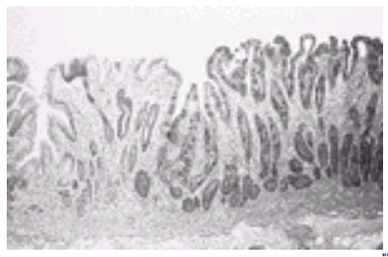

FIGURE 68-8. Corpus mucosa: the **left third** shows complete atrophy (i.e., absence of appropriate oxyntic glands) and moderate inflammation; the epithelium on the **right** consists of a small intestinal–type epithelium with goblet cells and a brush border (intestinal metaplasia).

Endocrine Cell Hyperplasia

Endocrine cell hyperplasia, a consequence of functional changes in chronic gastritis, is most prominent in autoimmune atrophic gastritis. In this condition, hypochlorhydria or achlorhydria lead to G-cell hyperplasia in the antral mucosa with an accompanying rise in serum gastrin levels. [25] This, in turn, induces the histamine-producing enterochromaffin-like cells in the oxyntic glands to undergo hyperplasia. [26] The type, number, and distribution of endocrine cells can now be established by using immunostaining techniques, which have replaced the less reliable classical argentaffin and argyrophil histochemical stains. Gastric endocrine cell proliferations are commonly classified according to the criteria proposed by Solcia and colleagues, [27] which distinguishes among hyperplasia, adenomatoid hyperplasia, dysplasia, and neoplasia.

Mild degrees of neuroendocrine proliferation may be seen, if they are searched for by immunoenzymatic methods, in many routine gastric biopsies: they represent an indirect and reversible effect of the widespread long-term use of proton pump inhibitors. [28] In this condition, hypertrophy occurs almost exclusively in enterochromaffin-like cells, the most common type of endocrine cell in the oxyntic mucosa, and is believed to depend on the trophic effect of the concomitant hypergastrinemia. [29]

Parietal Cell Alterations

Protrusions and pseudohypertrophy of oxyntic cells and glandular dilations are characteristic yet reversible responses to the long-term administration of proton pump inhibitors (Fig. 68-9). [29] These changes have also been described in patients with gastric ulcer disease, although the pathogenetic connection remains unclear. [30]

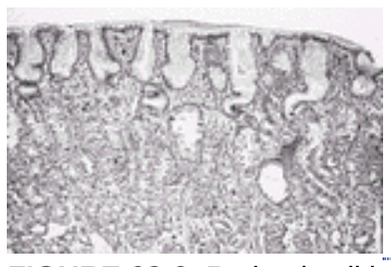

FIGURE 68-9. Parietal cell hyperplasia with protrusion into the lumen of dilated oxyntic glands is a distinctive effect of prolonged therapy with proton pump inhibitors.

HELICOBACTER PYLORI GASTRITIS

Chronic gastritis, one of the most common chronic conditions of humankind, is now known to be the result of specific and nonspecific responses mounted by the gastric mucosa against *H pylori* infection. [31], [32] *H pylori* infection is associated with most duodenal ulcer and gastric ulcers and with almost all primary gastric MALT lymphomas. In certain regions of the world, many infected persons develop metaplastic atrophic gastritis, a documented precursor of gastric carcinoma. Furthermore, certain extragastric conditions, including systemic autoimmune diseases, atherosclerosis, urticaria, and migraine, have been linked—albeit tenuously—to *H pylori* infection. [33] Thus, the importance of *H pylori* extends into the realm of numerous major diseases that may have gastritis as their common denominator.

Clinical Manifestations

The initial phases of *H pylori* infection elicit an acute mucosal inflammatory response whose clinical manifestations may include epigastric pain, nausea, and vomiting. Such symptoms are uncommon and are usually short-lived. Because patients rarely undergo endoscopic procedures in the early stages of *H pylori* infection, information regarding clinical aspects is limited. However, well-documented case reports allowed a glimpse into the early aspects of the infection [34] and of iatrogenic *H pylori* infection resulting from the use of inadequately disinfected endoscopes. [35] Most of these patients had multiple antral ulcers or erosions, whereas others had a predominance of hemorrhagic lesions; both their endoscopic and histopathological findings are virtually identical to those reported in subjects from human ingestion studies [36] and in patients with epidemic gastritis and achlorhydria, a condition described before *H pylori* was discovered and later found to be a manifestation of acute *H pylori* infection. [37]

In patients with uncomplicated chronic *H pylori* gastritis, the prevalence of dyspepsia is probably no greater than in uninfected persons, and among patients with nonulcer dyspepsia, the prevalence of infected and noninfected persons is similar. [38], [39] Furthermore, cure of *H pylori* in patients with nonulcer dyspepsia has not been shown conclusively to improve the dyspeptic symptoms, [40] although the reasons for this apparent lack of effect continue to be debated. [41] Even with its unclear relation to dyspepsia, *H pylori* infection has been estimated to be responsible for approximately 5% of gastrointestinal ailments in the community, [42] and patients with *H pylori* gastritis are at increased risk of duodenal and gastric ulcer, gastric cancer, and lymphoma.

Epidemiology

In many developing countries, the prevalence of *H pylori* in adults is close to 90%, with very high percentages of infected children, suggesting exposure to the bacteria early in life. In established industrialized countries (Western Europe, United States, Canada, and Australia), exposure occurs later, resulting in minimal percentages of infected children (<1% in Swedish and Danish schools in the year 2000) and low percentages of infected adults (~30% by age 50 years). Although the mechanisms of transmission remain poorly understood, improved socioeconomic conditions result in a decreased prevalence of *H pylori,* as vividly illustrated by historical data from Finland, [43] Sweden, [44] and Japan. [45] Furthermore, innumerable people receive amoxicillin, which, used alone, may cure approximately 10% of *H pylori* infections, [46] as well as other antibiotics for the treatment of respiratory and other infections. Among *H pylori*–infected patients undergoing such therapies, a small but cumulatively significant percentage is cured. Finally, it is likely that targeted anti–*H pylori* treatment in individual patients reduces transmission in the community and ultimately may contribute to the decreased prevalence of the infection in the population.

Endoscopic Appearance

Chronic *H pylori* gastritis has no distinct endoscopic pattern. Depending on the stage or distribution of gastritis, hyperemia, erosions, ulcerations, hypertrophy, and atrophy may coexist in various combinations in the same stomach, juxtaposed to one another and to apparently normal areas. Yet, none of these features is useful for predicting the presence or absence of chronic *H pylori.* Therefore, the diagnosis of *H pylori* gastritis requires histopathological analysis.

Histopathology

H pylori organisms are found in greatest numbers in the gastric mucous gel and attached to surface mucous cells (Fig. 68-10A). Bacteria are also present in the intercellular spaces and, particularly in patients receiving long-term antisecretory therapy with proton pump inhibitors, within the canaliculi of parietal cells. The difficulty of visualizing bacteria in these latter locations before the development of polyvalent staining techniques [47] may explain the misconception that *H pylori* is a strict gastric surface dweller.

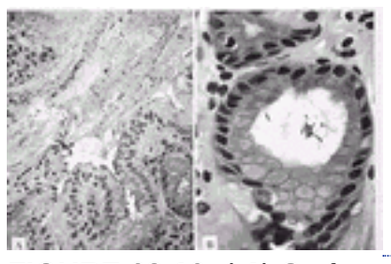

FIGURE 68-10. (**A**) Surface mucous layer containing innumerable *Helicobacter pylori* organisms. (**B**) *H heilmannii* at high magnification; these bacteria are more than twice as long as *H pylori,* and their spiral configuration is evident.

Antral, oxyntic, and cardiac mucosae are equally susceptible to infection and are colonized with similar frequencies. [8], [48] In patients with extensive antral intestinal metaplasia, a type of epithelium to which *H pylori* rarely adheres, [49] the infection is virtually confined to the nonmetaplastic areas of the corpus. This is also the case in many patients who regularly use proton pump inhibitors.

Infiltration of the gastric epithelium by polymorphonuclear neutrophils is the most distinctive feature of the gastric mucosa infected by *H pylori:* neutrophils are generally more abundant in the antrum and the cardia than in the corpus, where they may be rare or even completely absent in spite of visible bacterial colonization.

This distribution of inflammation characterizes *antrum-predominant gastritis,* the most common type of gastritis in Western populations. Neutrophils are usually the only inflammatory cells that infiltrate the gastric epithelium in *H pylori* infection, but in the lamina propria they are almost always mixed with lymphocytes, plasma cells, and variable amounts of eosinophils. Inflammation tends to be most intense in the superficial portions of the lamina propria (see Fig. 68-6A), hence the term *superficial gastritis,* still occasionally used as a synonym for nonatrophic gastritis.

Lymphoid follicles are virtually always found in infected stomachs (see Fig. 68-6B), and their presence is a reliable indication of active or recently treated *H pylori* gastritis. Their greatest density is in the region of the *incisura angularis,* and the lowest density is in the proximal greater curvature. [13]

"Disease-Specific" Virulence Factors

Since the discovery of the *H pylori* toxins VacA and CagA, which were linked to certain types of damage to the gastric mucosa, there have been continuous attempts to correlate them and other virulence factors with a specific manifestation or complication of *H pylori* gastritis. Although some of these factors can affect the intensity of inflammation and, perhaps through this mechanism, may ultimately influence the outcome of gastritis, [50] the virulence of *H pylori* seems to be largely host dependent, and none of these factors are disease specific. Thus, at present, there is no clinical application for tests that purport to determine the potential pathogenicity of an individual patient's *H pylori* strain. [51], [52] The best studied putative virulence factor is the cytotoxin-associated gene product (Cag) A, the product of one of the genes in the cag pathogenicity island. Subjects infected with *H pylori* with a functional cag pathogenicity island have elevated mucosal levels of interleukin-8, marked neutrophilic infiltration into the gastric mucosa, and an increased risk of developing a symptomatic outcome such as peptic ulcer or gastric cancer. However, the relationship between the presence of the cag pathogenicity island and outcome is not consistent in different geographic regions, especially in East Asia, where more than 90% of isolates possess the cag pathogenicity island. [53] In Western countries, where *H pylori* strains lacking the cag pathogenicity island are found in a higher percentage than in Asian countries, the increased likelihood of a symptomatic outcome can be seen. However, the presence of a functional cag pathogenicity island has no value in predicting current or future clinical presentation in individual patients. [54]

Other putative pathogenicity factors include the following: IceA (induced by contact with epithelium), a bacterial restriction enzyme for which no biologic or epidemiologic evidence as a virulence factor in *H pylori*–related disease has been confirmed; and VacA (vacuolating cytotoxin), which has been subtyped into an *s*1 genotype (presumably associated with duodenal ulcer disease) and an *s*2 genotype with reportedly low ulcerogenic potential. [55] A compilation of studies involving approximately 1500 isolates from Europe, the United States, and Asia has shown overwhelming that VacA genotyping is not useful to predict degree of inflammation, symptoms, presentation, or response to therapy.

The blood group antigen binding adhesin (BabA) is an outer membrane protein that appears to be involved in the adherence of *H pylori* to Lewis-b (Le^b) blood group antigens on gastric epithelial cells. [56] A small study suggested that infection with strains with the *babA*2 gene, *cagA* $^+$, and *vacA s*1 (triple-positive strains) may be correlated with duodenal ulcer, but a larger multinational study did not confirm the association. [52]

Diagnosis

The diagnosis of *H pylori* gastritis rests on the identification of *H pylori* in the gastric mucosa. When the search for *H pylori* relied exclusively on the availability of gastric biopsy specimens (for histopathology, rapid urease tests, or culture), only patients who required an endoscopic examination were tested. The development of increasingly accurate noninvasive tests allows the accurate diagnosis of *H pylori* gastritis based on indirect methods, that is, without necessarily visually identifying or culturing the organism. Furthermore, the availability and popularization of new, simple, and inexpensive tests have expanded the indications to a greater variety of patients and settings, including self-referring patients in the general practitioners' office.

Significant progress has been made also in our knowledge of factors that may influence the interpretation of the results and may interfere with the accuracy of each test. The prevalence of a condition in the population studied influences both the positive and negative predictive value of tests, even if a test's sensitivity and specificity are independent values inherent to the test itself. Therefore, clinicians should be familiar not only with the performance parameters of the tests they use, but also with their potential interpretative pitfalls as well as with the prevalence of *H pylori* in their patient population.

Invasive Tests

Histopathological examination of gastric biopsy specimens. *Helicobacter* spp. can be detected in histological preparations of gastric biopsy specimens stained with a variety of methods. Hematoxylin and eosin is a suboptimal choice of stains for the specific task of detecting *H pylori.* Reliable special stains include the Warthin-Starry and the Steiner silver stains, the Giemsa, Diff-Quick, and Gimenez stains, and a triple stain, [57] which, by combining modified Steiner staining, hematoxylin and eosin, and Alcian blue, allows the simultaneous visualization of the features of gastritis, including intestinal metaplasia, and the bacteria. Several modified versions of this stain have become available. [58] Anti– *H pylori* antibodies for the immunohistochemical detection of *H pylori* in paraffin-embedded biopsy specimens have high sensitivity and specificity; some laboratories use them for routine clinical diagnosis.

In situ hybridization and polymerase chain reaction. In situ hybridization may be used for the detection of *H pylori* in paraffin-embedded sections, but high cost and technical difficulties have relegated this procedure to the research laboratory. [59] Polymerase chain reaction for the detection of *H pylori* infection must also be considered a research tool because of its requirement for a sophisticated molecular biology laboratory, the availability of appropriate primers, and its high price.

Smear, brush, and touch preparations. Smears of gastric mucus and exfoliated epithelial cells, usually with Gram staining, may allow the detection of bacteria within minutes of the endoscopic procedure. Rapid urease tests have made cytologic assays obsolete.

Bacterial culture. *H pylori* is best cultured in a microaerophilic and humid atmosphere on culture media requiring fresh horse or sheep blood and antibiotics to suppress contaminants. [60] Cultures for *H pylori* are technically more complicated than those performed by the usual clinical microbiology laboratory. Because many clinical facilities are not equipped to perform the time-consuming procedures necessary to culture *H pylori,* several methods for transportation have been devised. [61]

Rapid urease tests. These assays exploit the high content of urease of *H pylori.* A fragment of gastric mucosa is placed into a broth or in agar containing various concentrations of urea. The urease produced by *H pylori* hydrolyzes the urea and releases ammonia, which raises the pH of the broth or agar, [62] and an appropriate indicator (e.g., phenol red) changes color as the pH increases. In the first commercially produced rapid urease test, the CLOtest, the original yellow gel capsule into which the specimen is placed becomes red within minutes to hours, depending on the quantity of bacteria present. Several rapid urease tests are now commercially available. Both their specificity and sensitivity, compared with histopathological examination, are extremely high, in most cases approaching 100%. [63], [64]

Noninvasive Tests

Serology. The development of new techniques has minimized the problems of cross-reactivity that plagued first-generation serologic tests. [65] Currently available tests are highly reliable. The large numbers of studies aimed at the discovery of an optimal diagnostic test for *H pylori* infection have also provided valuable information on the immune responses to this organism. For example, the selection of *H pylori* strains as sources of antigen is critical to the specificity and sensitivity of a test, and it is imperative to evaluate specific tests in the population to which it would be applied before selection of a test for use in specific settings. This population specificity highlights the importance of the many different geographic strains that infect different world populations.

Simplified "in-office" immunoenzymatic tests. Several in-office devices have been developed for the rapid detection of IgG anti– *H pylori* antibodies. Most of them consist of disposable kits that provide a yes/no answer within a few minutes of placing a drop of serum in a well that is preabsorbed with antigen and an immunoenzymatic detection system. Although some of these tests are accurate, results have been generally less than the minimum required sensitivity and specificity of 90%. [66], [67] Antibodies (mostly of the IgG class) against *H pylori* have been detected in the saliva and the urine of infected patients. The specificity and sensitivity of urine tests have been found to be satisfactory in several studies, particularly in Japan. [68]

Stool antigen assay. An enzymatic immunoassay (HpSA) that detects *H pylori* antigens in stools (thus providing information on the presence of current infection) has become available for the diagnosis of *H pylori* infection and for monitoring the response to therapy. The test is similar to an enzyme-linked immunosorbent assay, using polyclonal anti– *H pylori* antibody absorbed to microwells. A large European multicenter study has yielded encouraging results. [69], [70]

Urea breath tests. The urea breath tests are among the most important and innovative methods to detect *H pylori* infection. [71] These tests rest on the ability of *H pylori* to produce large quantities of urease. The ingestion of a solution containing urea is rapidly followed, in an infected patient, by the production of ammonia and carbon dioxide. The latter rapidly appears in the subject's breath. If the ingested urea is labeled either with the radioactive isotope ^{14}C or with the nonradioactive isotope ^{13}C, then the exhaled carbon dioxide will also be labeled and, therefore, measurable by an appropriate detection method. When ^{14}C-labeled urea is used, the general method consists of the ingestion of a solution or a capsule containing quantities between 0.5 and 10 µCi of the isotope-labeled urea. When ^{13}C-labeled urea is used, test subjects are given a solution of 125 g of 99.9% labeled urea followed by a meal aimed at increasing its permanence in the stomach. After a period of time, the subject inflates a balloon, which is immediately sealed and sent to a laboratory for the detection of the isotope-labeled carbon dioxide. Both types of tests are now well standardized and are approved by regulatory agencies in Europe and North America. The urea breath tests are extremely sensitive and specific and, in contrast to serologic tests, detect current active infection (not evidence of past infection). Their widespread use has made them more affordable, and they have

become the test of choice for a variety of populations, including children, pregnant women, and patients who cannot undergo an endoscopic procedure. [72], [73]

Helicobacter heilmannii Infection

More than 35 species of *Helicobacter* have been described, but only few other than *H pylori* have been shown to cause gastritis in humans: *H felis, H fennelliae, H cinaedi,* and *H heilmannii.* Among these, *H heilmanni*i (formerly known as *Gastrospirillum hominis*) is the most common, [74], [75] with an estimated prevalence of approximately 1% of all human *Helicobacter* infections. [75], [76] In some rural areas in Eastern Europe, this organism has been detected more commonly, leading to the hypothesis of zoonotic transmission. [75], [76] and [77] The bacterial morphology is characteristic: organisms measure 5 to 9 µm in length (twice as long as *H pylori*) and have five to seven spirals clearly visible with a silver stain. Gastritis caused by *H heilmanni*i is often milder and more patchily distributed than *H pylori* gastritis. The inflammation tends to be more circumscribed and to affect mostly the antrum, although cases with severe corpus active inflammation are seen. Concurrent erosions and ulcers have been reported to be less common than in *H pylori* gastritis. The diagnosis rests on the recognition of the bacterial morphology, although the distinction between *H heilmanni*i and *H felis* is not possible by light microscopy (see Fig. 68-10B).

Treatment of *Helicobacter pylori* Infection

At present, the only universally agreed on indications for treatment are *H pylori*–related duodenal and gastric ulcers and low-grade, primary B-cell MALT lymphoma. *H pylor*i should be eradicated in patients with documented ulcer disease, whether or not the ulcers are currently active, to reduce the likelihood of relapse. Most clinical trials do not provide convincing data in support of the benefits of eradication of infection in patients with nonulcer dyspepsia, and there are no controlled studies showing that eradicating *H pylor*i from a population will reduce the incidence of gastric cancer. For various logistical and ethical reasons, it is unlikely that such trials will be ever carried out to the satisfaction of those who demand unequivocal evidence. [78] The most pressing question then is whether we should hold back and wait for more data or act now based on current information. Today, there is ample epidemiologic and biologic evidence that whereas *H pylori* gastritis may not be the only cause of the development of atrophy and intestinal metaplasia, it almost always provides the necessary background on which these lesions arise. [79] By treating *H pylori* gastritis, we can prevent the development of atrophy and metaplasia, and most likely we would also arrest the progress of these lesions in infected persons who have already developed them. As a result, we should be able to prevent millions of gastric cancers. The incidence of non–NSAID-induced peptic ulcers would also be greatly reduced, and primary gastric lymphomas would all but disappear. Thus, we ought to put aside the teleological questions on the ultimate significance of our immemorial amphibiotic relationship with *H pylori* and its intriguing evolutionary and metaphysical implications [80] and proceed to cure infected patients.

In vitro, *H pylori* is susceptible to a wide range of antibiotics, but monotherapy has been disappointing in vivo, probably because of inadequate antibiotic delivery to the sites of colonization. Thus, several multidrug regimens have been developed, the most successful of which are triple and quadruple combinations that achieve *H pylori* eradication rates of more than 90% in many trials and more than 75% in clinical practice. The most commonly used 7- and 14-day drug regimens consisting of a proton pump inhibitor and two or three antimicrobial agents are listed in Table 68-2. The major determinants of therapeutic failures are inadequate patient compliance and drug resistance, particularly to metronidazole and clarithromycin. [81]

TABLE 68-2 Three Recommended Multidrug Regimens for the Eradication of *Helicobacter pylori* *

There are no established guidelines for posttreatment testing. When eradication therapy is given for gastric ulceration or MALT lymphoma, there is an opportunity to retest for *H pylori* at repeat endoscopy, which is performed to evaluate healing or regression. For duodenal ulceration, a urea breath test, a stool antigen test, or an endoscopy with gastric biopsy should be performed 4 to 6 weeks after treatment. When therapy is administered to treat asymptomatic infections, posttreatment testing is generally not deemed necessary.

Evolution and Associations of *Helicobacter pylori* Gastritis

H pylori gastritis is a life-long infection that can be viewed as a spectral disease. At the one end of the spectrum, inflammation remains mostly confined to the antrum and the cardia, the oxyntic mucosa is mildly affected, and atrophy is absent or minimal. At the other end, severe inflammatory changes involve the entire gastric mucosa and inflict progressive damage that results in loss of the normal gastric glands and their replacement by fibrous tissue and metaplastic epithelia. Although these are aspects of the same disease that can be placed on a continuous scale, the epidemiologic distribution and the associations of the two extremes are profoundly different. [82] Gastritis confined to the antrum and without significant atrophy does not impair acid secretion, is commonly present in patients with duodenal ulcer, and has not been linked to increased cancer risk. Conversely, generalized gastritis with atrophy reduces acid secretion and is strongly associated with gastric adenocarcinoma. [83] Thus, for the practical purposes of classification and prognostic evaluation, it has been expedient to divide gastritis into two phenotypes: nonatrophic antrum-predominant gastritis and multifocal atrophic gastritis.

Antrum-Predominant Gastritis This is the most common form of gastritis in the Western world. Its characteristics are as follows: a moderately to severely inflamed antrum; a mildly inflamed or normal corpus; minimal or absent atrophy or intestinal metaplasia, limited to the antrum; and normal or increased acid secretion. Most patients with this type of gastritis have neither symptoms nor complications. However, they have a risk of duodenal ulcer, estimated at 20% over their lifetime. [84], [85]
Atrophic Gastritis Also called *multifocal atrophic gastritis, metaplastic atrophic gastritis,* and *atrophic pangastritis,* this disorder is characterized by marked diffuse mucosal inflammation, often more severe in the oxyntic mucosa, by patches of atrophy and intestinal metaplasia in both antrum and corpus, and by variously reduced acid secretion. Atrophic gastritis is most prevalent in populations that are—or were, until a few decades ago—living in suboptimal sanitary conditions, including much of South and East Asia, Latin America, and parts of Central, Eastern, and Southern Europe. [86] Socioeconomic factors may be a surrogate for other unknown agents that modulate the evolution of gastritis, because there are notable epidemiologic exceptions to this association. Japan, a country with high levels of sanitation and personal hygiene, has one of the world's highest prevalences of atrophic gastritis and a high incidence of gastric adenocarcinoma. In contrast, Equatorial Africa, in spite of its precarious socioeconomic texture, inadequate sanitary standards, and a prevalence of *H pylori* close to 90%, appears to have a low prevalence of atrophic gastritis and a low incidence of gastric adenocarcinoma. [87] Several explanations have been proposed for this 'African enigma," ranging from diet, human and bacterial genetics, to unreliable statistics. [88] Atrophic gastritis is a risk factor for gastric epithelial dysplasia, a precursor of the intestinal-type adenocarcinoma of the stomach. [79] It also predisposes patients to gastric ulcer.
Peptic Ulcers, Carcinoma, and Lymphoma In addition to peptic ulcer disease (see Chapter 66) and gastric carcinoma (see Chapter 69), *H pylori* infection is also epidemiologically related to primary gastric MALT-lymphomas, which are also discussed in Chapter 69.
Extragastrointestinal Manifestations of *Helicobacter pylori* infection *H pylori* infection has been proposed to be associated with an ever-growing number of extragastric manifestations of *H pylori* infection, even if their causal relationship with *H pylori* is far from conclusively demonstrated. Most of these associations are founded on epidemiologic data; however, both *H pylori* infection and of some of the conditions allegedly associated with it (e.g., atherosclerosis) have a very high prevalence in many populations. Thus, biologic rather than epidemiologic data will be needed to prove causation. For one of these conditions, rosacea, there are several randomized trials that essentially disprove the association with *H pylori.* [89], [90] Conversely, several clinical studies based on treatment results lend some support to the possibility of a pathogenetic involvement of *H pylori* in iron deficiency anemia and autoimmune thrombocytopenic purpura. [91], [92] and [93] For most other conditions, data are insufficient to reach informed conclusions; however, the biologic plausibility in some of the proposed associations (e.g., with migraine) is so low that one must wonder whether attempts to prove causality are worth the effort.

INFECTIOUS GASTRITIS (EXCLUDING *HELICOBACTER* GASTRITIS)

Because of the high acid content of the normal stomach, the gastric environment is inhospitable to most infectious agents. However, in patients with atrophic gastritis and decreased acid secretion, in patients with impaired immune responses, or as part of systemic infections, numerous viruses, bacteria, and parasites can infect the stomach. Although rare, some of these infectious gastritides have characteristic clinical and pathological features.

Viruses

Enteric rotaviruses and *caliciviruses* probably infect the stomach during the course of gastroenteritis, but no pathological changes in the gastric mucosa have been documented in volunteer studies with these agents. Only *cytomegalovirus* (CMV) infection is known to have a distinct pathological appearance in the stomach. CMV gastritis is seen almost exclusively in young children and immunocompromised patients, and it is usually associated with concurrent CMV infection of other sites of the digestive tract. Endoscopically, the gastric mucosa may appear completely normal or may show erosions, shallow ulcers, [94] or hemorrhagic gastritis. [95] Rarely, the condition may present as grossly nodular mucosa that has been referred to as a pseudotumor. [96] CMV inclusions may be abundant and thus easily detected using the routine hematoxylin and eosin stain, or they may be rare and impossible to demonstrate without using immunohistochemistry or in situ hybridization techniques. A characteristic manifestation of CMV infection in the stomach of young children is massive foveolar hyperplasia accompanied by edema and mild inflammation of the lamina propria. The resulting endoscopic appearance is that of a giant-fold hypertrophic gastropathy indistinguishable from Ménétrier disease. [97]

The diagnosis of gastric CMV infection is made by demonstrating the characteristic nuclear or cytoplasmic viral inclusions (Fig. 68-11A). The only effective therapeutic agent is ganciclovir, a guanosine derivative that selectively inhibits CMV DNA polymerase. In patients with acquired immunodeficiency syndrome (AIDS) and CMV colitis, ganciclovir has a response rate of 70% to 90%. [98], [99]

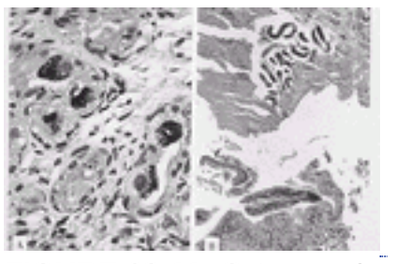

FIGURE 68-11. Cytomegalovirus cytoplasmic and nuclear inclusions (**A**) and *Strongyloides stercoralis* larvae (**B**) in gastric biopsy specimens from immunocompromised patients.

Bacteria

Bacterial overgrowth may occur in stomachs that have become achlorhydric as a result of atrophy, complete antrectomy, or vagotomy or as a result of long-term use of histamine H_2-receptor antagonists or proton pump inhibitors. [100] Patients with scleroderma and other severe motility impairments are also prone to bacterial overgrowth. [101] In contrast to *H pylori,* however, these bacteria colonize rather than infect the gastric mucosa, and they neither elicit inflammatory responses nor cause symptoms.

An extremely rare condition is *acute suppurative gastritis,* also known as *phlegmonous gastritis.* This life-threatening condition is caused by pyogenic bacteria (streptococci, staphylococci, *Escherichia coli, Proteus,* and *Haemophilus* spp.) and is characterized by large areas of purulent necrosis involving the full thickness of the gastric wall. When it is caused by gas-forming organisms, the term *emphysematous gastritis* has been used. Few cases have been reported, mostly in very young children, elderly persons, patients with alcoholism, and immunocompromised patients. Potential iatrogenic causes include polypectomy [102] and mucosal injection with India ink. [103] The diagnosis is made endoscopically or at surgery. Antibiotic treatment may have to be accompanied by surgical intervention. [104]

Primary gastric tuberculosis is rare, particularly in industrialized countries. [105], [106] However, in patients with *disseminated tuberculosis, necrotizing granulomata* may be found in the gastric mucosa. Another *Mycobacterium* that has gained prominence with the spread of AIDS is *Mycobacterium avium-intracellulare* complex, but the stomach is rarely involved; when it is, typical lesions consist of accumulations of foamy histiocytes in the lamina propria, sometimes with formation of ill-defined granulomata without necrosis.

In the 1980s and early 1990s, increasing numbers of cases of *gastric syphilis* were reported in patients infected with human immunodeficiency virus. [107] When associated with secondary syphilis, syphilitic gastritis is characterized by a prominent mixed inflammatory infiltrate consisting predominantly of plasma cells and with mucosal ulcerations. The infiltrate may be dense enough to cause the swelling of gastric folds, which may also undergo erosion and ulceration, sometimes mimicking the endoscopic appearance of lymphoma or infiltrating carcinoma. [108] Symptoms include severe dyspepsia, nausea, vomiting, and anorexia, with rapid weight loss. The diagnosis is often delayed by a low index of suspicion. Although spirochetes may be seen in sections stained with appropriate silver stains (Dieterle, Steiner, or Warthin-Starry), the search may be painstaking and is usually beyond the reach of the nonspecialist pathologist. Standard treatment for secondary syphilis is rapidly effective.

Fungi

Candida species, *Histoplasma capsulatum,* and Mucoraceae have been found in the stomach of immunocompromised patients, particularly those with AIDS, with disseminated infections, but none of these fungi have been reported as a primary cause of gastritis.

Parasites

The stomach is not a preferred site for human parasitic infections, but *Cryptosporidium* spp. [109] and *Giardia intestinalis* [110] have been identified in the gastric mucosa (see Fig. 68-11B). *Strongyloides stercoralis* has been found in the stomach of a few patients with widespread infections. [111]

The only nematodes that invade the human gastric wall are those of the family collectively known as Anisakidae, or “sushi worms.” *Anisakiasis* is an important cause of morbidity in countries such as Japan, where large quantities of raw fish are consumed. [9], [112] The muscle of many species of edible fishes contain larvae of Anisakidae. In a small proportion of persons who eat infected fish, larvae penetrate the gastric wall and cause a sudden onset of epigastric pain. Because the worms can be easily removed endoscopically by an experienced operator and the disease is self-limited in any case, patients presenting with epigastric pain in high-prevalence countries should be asked routinely about ingestion of raw or undercooked fish within 12 hours before the onset of symptoms. This practice would help to avoid unnecessary surgery.

AUTOIMMUNE GASTRITIS

Autoimmune gastritis is a corpus-restricted chronic atrophic gastritis associated with circulating serum anti–parietal cell and anti–intrinsic factor antibodies and intrinsic factor deficiency, with or without pernicious anemia. [12]

Clinical Manifestations

Most clinical manifestations of autoimmune gastritis become apparent only many years after the onset of the disease, when the parietal cell mass decreases beyond a critical point and the stomach becomes unable to produce sufficient amounts of acid, pepsinogens, pepsin, and intrinsic factor. *Achlorhydria,* the most direct result of the destruction of acid-producing oxyntic cells, typically occurs in the most advanced stages of the disease. *Hypochlorhydria,* however, may also occur in patients with moderate to large numbers of surviving parietal cells, suggesting that anti–proton pump antibodies or inhibitory lymphokines released by subsets of inflammatory cells may participate in the inhibition of acid secretion. Patients with corpus atrophy and achlorhydria have hypergastrinemia, which tends to correlate with the severity of the mucosal damage. [113] Injury to chief cells leads to a reduction of pepsin activity in gastric juice and of pepsinogens in blood. A low serum pepsinogen I level (<20

ng/mL) is a sensitive and specific indication of corpus atrophy. [113], [114]

Many patients with autoimmune gastritis develop either iron deficiency or pernicious anemia. Hypochromic anemia is associated with corpus-restricted chronic atrophic gastritis in approximately 15% of patients. [12] Achlorhydria seems to be the major contributor to the pathogenesis of iron deficiency anemia: gastric acid is important in the absorption of nonheme iron, which in Western diets supplies at least two thirds of nutritional iron needs. [115] Pernicious anemia is usually preceded by corpus-restricted chronic atrophic gastritis and reduced acid secretion by approximately a decade, and it is generally associated with a histological pattern of end-stage atrophic gastritis. [116] Pernicious anemia is a rarely diagnosed condition, with a reported prevalence of less than 1% even in elderly persons in high-incidence countries. [117], [118] Its true prevalence could be higher, but most patients are successfully treated by hematologists or general practitioners for their anemia and cobalamin (vitamin B_{12}) deficiency, and they are never referred to a gastroenterologist for the evaluation of atrophic gastritis, pepsinogens levels, or anti–parietal cell antibodies. Cobalamin deficiency affects the rapidly proliferating gastrointestinal epithelium, and patients with severe deficiency may complain of a sore tongue, which may be smooth and beefy red. Anorexia with moderate weight loss may also be evident, possibly accompanied by diarrhea and other gastrointestinal symptoms. Neurological manifestations include numbness and paresthesia in the extremities, weakness, and ataxia. There may be sphincter disturbances.

Autoimmune gastritis is a risk factor for hyperplastic and adenomatous polyps, carcinomas, and endocrine tumors. Polyps are found in 20% to 40% of patients with pernicious anemia, and they are mostly sessile, smaller than 2 cm in diameter, and often multiple. Most polyps are hyperplastic, but up to 10% contain dysplastic foci. [119] Gastric cancers associated with pernicious anemia are of the intestinal type and arise from intestinal metaplasia, a finding suggesting that the link between autoimmune gastritis and carcinoma may be intestinal metaplasia and its dysplastic transformation. [120]

Pathogenesis

Autoimmune gastritis is substantially more common in patients with other diseases thought to be of immunologic origin (Graves disease, myxedema, thyroiditis, idiopathic adrenocortical insufficiency, vitiligo, and hypoparathyroidism) than in the healthy population. The high prevalence of specific familial histocompatibility haplotypes such as human leukocyte antigen (HLA)-B8 and HLA-DR3 in patients with corpus-restricted atrophic gastritis is another strong indicator of its autoimmune origin. The precipitating factors, however, remain unknown. The hypothesis that autoimmune gastritis could be initiated by *H pylori* infection has received considerable attention. A high prevalence of antibodies with high specificity for gastric mucosa antigens has been reported in patients with *H pylori*–associated gastritis; preabsorption of the serum from these patients with *H pylori* removed most of these autoantibodies. [121] Furthermore, 20% of these patients had autoantibodies that reacted with the secretory canalicular structures of parietal cells, which are among the major targets in autoimmune gastritis. In other studies, *H pylori*–positive patients with and without previously demonstrated anticanalicular antibodies had a 30% to 50% prevalence of anti–H^+,K^+-ATPase antibodies, in contrast to a less than 3% prevalence in noninfected persons. [122] It has also been demonstrated that *H pylori* lipopolysaccharides express Lewis x and y blood group antigens, which are also expressed by either H^+,K^+-ATPase or gastric epithelial cells. [123], [124] Collectively, this information lends support to the concept that a cross-mimicking mechanism between *H pylori* and gastric mucosa antigens participates in the pathogenesis of autoimmune gastritis. This correlation, however, remains unproven, and much stronger biologic, clinical, and epidemiologic evidence is needed before *H pylori* can be viewed as the cause of autoimmune gastritis. [125]

Endoscopic Appearance

Atrophy causes a progressive thinning of the mucosa of the gastric corpus; this explains why few folds are left and why fine submucosal vessels are easily recognized on endoscopic examination, especially in advanced disease. The antral mucosa is endoscopically normal in the majority of cases. Polyps become common in the advanced stages of the disease; therefore, atrophic gastritis must be ruled out in patients in whom multiple hyperplastic gastric polyps are detected at endoscopy.

Histopathology

The main histopathological features of autoimmune gastritis are diffuse corpus-restricted chronic atrophic gastritis with mild to moderate intestinal metaplasia and, in the absence of concurrent *H pylori* infection, a normal gastric antrum. This pattern is characteristic of the advanced stage of the disease and is found in patients with pernicious anemia. Persons with parietal cell antibodies and no pernicious anemia show a broad spectrum of atrophic changes, from minimal oxyntic gland loss to severe and diffuse atrophy of the oxyntic mucosa. [12] In the early phases, one sees diffuse or multifocal, dense mononuclear cell infiltration of the lamina propria and lymphocytic infiltration of individual oxyntic glands. Later, marked atrophy of the oxyntic glands with diffuse mononuclear cell infiltration of the lamina propria develops. Pyloric metaplasia is extensive, whereas intestinal metaplasia tends to be still limited to few foci. The end stage is characterized by a great reduction in corpus mucosal thickness, foveolar hyperplasia, and replacement of oxyntic glands by pyloric, pseudopyloric, or intestinal metaplasia. The inflammatory infiltrate is minimal, although scattered lymphoid aggregates and follicles may be found.

In the majority of patients, the antral mucosa is either normal or shows only focal areas of mild chronic inflammation with intestinal metaplasia, similar in degree and extension to what is observed in the general asymptomatic population. Hyperplasia of gastrin cells secondary to achlorhydria is often seen. Enterochromaffin-like cell carcinoids may arise during the florid phase, but they are found more commonly in association with an end-stage histopathological pattern. [120]

Diagnosis

Autoimmune gastritis should be suspected in patients with megaloblastic anemia, with evidence of clinically significant cobalamin deficiency with values lower than 150 pg/mL, or with multiple gastric polyps. The diagnosis must be confirmed by the characteristic histopathological findings of corpus-restricted atrophic gastritis and by the presence of serum anti–intrinsic factor or anti–parietal cell antibodies.

Management

The management of patients with autoimmune gastritis should address two aspects of the condition: the gastric lesions and the manifestations related to the cobalamin deficiency. Gastric mucosal atrophy is irreversible. Patients with extensive intestinal metaplasia and those with multiple polyps may be at increased risk of gastric cancer. Although there are no accepted guidelines, a surveillance gastroscopy every 1 or 2 years may represent a sensible empiric approach.

With cobalamin replacement, most abnormalities resulting from cobalamin deficiency undergo complete and lifelong correction, except for the neurological manifestations. Their improvement depends on the extent that irreversible changes in the nervous system may have occurred before treatment.

LYMPHOCYTIC GASTRITIS

Lymphocytic gastritis is characterized by the presence of large numbers of mature lymphocytes infiltrating the surface and foveolar epithelium. [11]

Clinical Manifestations

In his original report of this condition, Haot suggested that lymphocytic gastritis corresponded to the endoscopic entity previously known as *varioliform gastritis.* The histological features of lymphocytic gastritis may also be found in endoscopically normal stomachs and in patients with celiac disease, and the varioliform aspect (numerous thickened folds capped by small nodules that contain a central erosion or ulceration) can be found in *H pylori*–infected stomachs. [126], [127]

Lymphocytic gastritis is rare, found in 1% to 3% of persons who undergo endoscopy with biopsy sampling. [128], [129] It is most commonly diagnosed in the sixth decade and appears to affect men and women equally. [130]

In contrast to patients with chronic active gastritis, patients with the varioliform type of lymphocytic gastritis are often symptomatic; rapid weight loss and anorexia are reported in about half of affected patients, whereas epigastric pain is less common. Hypoproteinemia, hypoalbuminemia, and peripheral edema suggesting protein-losing gastroenteropathy have been documented in approximately 20% of patients. [11] Lymphocytic gastritis is a chronic disease, but there have been few case reports of spontaneous resolution. When it is associated with gluten enteropathy, the signs, symptoms, and clinical course are those of celiac disease (see Chapter

76).

Pathogenesis

Both the etiology and the pathogenesis of most lymphocytic gastritides are unknown. Because virtually all intraepithelial lymphocytes are CD8 $^{+}$ suppressor T cells, the same cells that form the intraepithelial infiltrate in celiac disease, an allergic pathogenesis has been proposed. This hypothesis is supported by the increasingly apparent association between a subtype of lymphocytic gastritis and celiac disease. [129], [131], [132] The relationship with *H pylori,* initially enthusiastically embraced, then vigorously rejected, is now viewed with an agnostic attitude by most investigators.

Macroscopic and Endoscopic Appearance

Approximately 80% of patients in whom a histopathological diagnosis of lymphocytic gastritis is made have endoscopic lesions described as varioliform, aphthous, verrucous, or chronic erosive gastritis. [11], [133] The appearance is that of a complex pattern of enlarged folds, predominantly in the corpus, which are covered by thick mucus, are crossed by large erosions, and may be crested with clusters of elevated aphthoid nodules. Flat erosions may be found in the antrum. The remaining 20% of patients (including those with celiac disease and collagenous gastritis) have less dramatic lesions, with only scattered superficial erosions in the corpus or antrum. Some patients have an endoscopically normal stomach. This last group of patients with a normal-appearing stomach may have a pathogenetically distinct condition, even though the histopathological features are indistinguishable. Some authors have suspected a relationship between lymphocytic gastritis and Ménétrier disease. [134]

Histopathology and Diagnosis

Because the endoscopic appearance of varioliform gastritis may result from either *H pylori* infection or lymphocytic gastritis, the diagnosis can be suspected clinically, but it can be confirmed only by histopathologists. In biopsy specimens from these patients, a substantial increase in intraepithelial lymphocytes, particularly in the corpus, is associated with a histopathological spectrum ranging from marked chronic inflammatory cell infiltration of the lamina propria, activity, and focal erosions to a minor increase in chronic inflammatory cells with no activity (Fig. 68-12A). In most cases, the histological picture can be readily distinguished from that of chronic *H pylori* gastritis, in which few intraepithelial lymphocytes are present (rarely more that 5 or 6 per 100 epithelial cells). The diagnostic threshold for lymphocytic gastritis is generally accepted as more than 25 intraepithelial lymphocytes per 100 epithelial cells, but in most cases the counts are much greater, between 25 and 50 lymphocytes. [135] If *H pylori* infection is present, immunohistochemistry to detect CD8 $^{+}$ T cells may be helpful: in pure lymphocytic gastritis, most intraepithelial lymphocytes are CD8 $^{+}$, whereas a heterogeneous infiltrate characterizes *H pylori* gastritis.

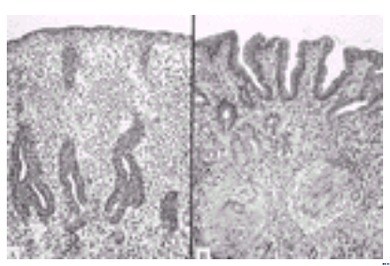

FIGURE 68-12. (**A**) Lymphocytic gastritis: a dense, mostly mononuclear infiltrate obliterates the lamina propria, and large numbers of CD8 $^{+}$ suppressor T cells infiltrate both the surface and the foveolar epithelium. (**B**) Two large, nonnecrotizing granulomata in the antral mucosa of a patient with no known infection or systemic granulomatous condition. In such a case, the provisional label of “idiopathic granulomatous gastritis” may be used.

Therapy

When concurrent *H pylori* infection is present, eradication of the infection may resolve the inflammation and is therefore recommended. [136], [137] Patients with gluten enteropathy will benefit from the standard dietary limitations recommended to restore the small intestinal integrity. For patients with “pure” lymphocytic gastritis and severe gastric mucosal lesions, there are no consistently effective therapies. The use of proton pump inhibitors has resulted in healing of the ulcers and erosions in some patients. Therapy with glucocorticosteroids or sodium cromoglycate, often successfully used for the treatment of eosinophilic gastroenteritis, [138], [139] has obtained unclear results.

GRANULOMATOUS GASTRITIS

Rather than a clinical entity, *granulomatous gastritis* can be viewed as a histopathological category to which stomachs bearing granulomata are temporarily assigned while the condition responsible for their development is identified. In most cases the morphologic appearance of granulomata does not provide useful clues as to their cause, except when foreign materials, acid-fast bacilli, or fungal forms are found. Thus, a specific diagnosis can be made only by integrating histopathological data with clinical and laboratory information.

Clinical Manifestations and Pathogenesis

Because granulomata can be found in the gastric mucosa of patients with infectious, inflammatory, and neoplastic diseases, as well as in otherwise healthy persons, neither clinical manifestations nor endoscopic appearances are useful to predict the presence of gastric granulomata. Thus, the stomach may be completely normal or may exhibit characteristics of the associated conditions.

Mycobacterium tuberculosis **and** ***Histoplasma capsulatum*** **Infection** *Tuberculosis* is the most common cause of granulomatous disease of the gastrointestinal tract, but the stomach is rarely affected, even in the presence of severe ileocecal involvement. Primary gastric tuberculosis has been reported mostly from developing countries with a high prevalence of *Mycobacterium tuberculosis* infection. [106], [140] In almost all such cases, gastric tuberculosis was diagnosed because of the presence of a large, nonhealing gastric ulcer. A few cases of primary infection of the stomach by the dimorphic fungus *Histoplasma capsulatum* have been reported. As in patients with gastric tuberculosis, these patients presented with signs and symptoms that led to the discovery of a large gastric ulcer initially interpreted clinically as malignant. [141], [142]

Helicobacter pylori **Infection** Unexplained granulomata have been found in patients with *H pylori* gastritis, but this finding is extremely rare, on the order of less than 1 in 1000 infected patients. Although a role for *H pylori* in the causation of granulomata has been postulated, there is no evidence to support this hypothesis. [143], [144]

Anisakiasis Early lesions of *anisakiasis* range from interstitial edema with a loose, eosinophilic inflammatory infiltrate to eosinophilic abscesses, in which well-preserved larvae are often detected. In later lesions, the most common finding is the presence of foreign body granulomata, sometimes associated with fragments of helminthic cuticles. [112], [145]

Foreign Bodies A common cause of gastric *foreign body granulomata* is suture material in patients who have undergone a partial gastrectomy. In patients with gastric ulcer, food particles may become engulfed in the ulcer crater and may cause a foreign body reaction. When such granulomata are found in biopsy specimens obtained from active ulcers, their origin is readily apparent; diagnostic difficulties may arise when granulomata are found in specimens from healed ulcers and the pathologist is unaware of the relevant clinical information.

Tumors Rarely, *adenocarcinomas* (particularly mucin-secreting tumors) may induce the formation of foreign body granulomata in the gastric mucosa or perigastric lymph nodes. Granulomata have also been noted in the gastric mucosa of few patients with gastric *non-MALT lymphoma.*

Other Unusual Causes In rare instances, granulomatous gastritis may be part of an immune-mediated vasculitis syndrome or Wegener granulomatosis. Sometimes, it may assume a form akin to xanthogranulomatous cholecystitis. [146]

Granulomatous Gastritis in Sarcoidosis and Crohn’s Disease

Granulomata detected in the gastric mucosa of a patient with established *sarcoidosis* or *Crohn's disease* can be assumed to be part of the systemic process, and no further investigation is required. Because the finding of gastric mucosal granulomata may precede the discovery of the disease in other organs, the careful interpretation of gastric biopsy findings together with appropriate suggestions for further tests may lead to the prompt diagnosis of a condition that could otherwise remain obscure for a long time. [143]

In sarcoidosis, involvement of the gastrointestinal tract is found occasionally at autopsy, but only rarely has it clinical importance. However, severe gastric disease with outlet obstruction or bleeding has been reported. Endoscopic findings may include nodularity, polypoid changes, erosions, ulcers, and segmental, usually distal, rigidity resembling linitis plastica. [147], [148] These gross changes reflect the presence of numerous mucosal granulomata and severe fibrosis.

Studies have challenged the perception that gastric involvement is rare in Crohn's disease, and they found a prevalence of gastric granulomata of approximately 10% to 15% and a prevalence of "focally enhanced" *H pylori*–negative active gastritis of between 30% and 70%. [6], [149], [150] In these patients, focal gastritis was almost twice as frequent in the corpus as in the antrum, and epithelioid granulomata were twice as prevalent in the antrum.

Isolated Granulomatous Gastritis

Isolated granulomatous gastritis is a "holding category," applied as a temporary diagnostic label to cases of granulomatous gastric inflammation for which no cause has yet been determined (see Fig. 68-12B). Support for this approach derives from studies showing that most gastric granulomata are an expression of either concurrent or incipient Crohn's disease or systemic sarcoidosis. [143], [151], [152] Even after careful evaluation, some gastric granulomata will remain unexplained. These lesions are usually asymptomatic, there is no information on their natural history or evolution, and, therefore, no treatment can be recommended.

CHEMICAL (REACTIVE) GASTROPATHY

An association between the presence of bile in the stomach and gastric mucosal damage (*duodenogastric reflux*) was first postulated by William Beaumont in 1859, and several subsequent clinical observations led to the development of surgical techniques to prevent or minimize the regurgitation of duodenal contents into the stomach. In 1983, Dewar, Dixon, and Johnston [24] systematically described the histopathological changes associated with bile reflux in the stomachs of patients who had undergone gastrectomy as well as patients with duodenal or gastric ulcer. The original term *bile reflux* [4], [24], [153] was later replaced by *chemical gastritis,* to include the recently discovered NSAID-induced changes in the gastric mucosa. The terms *reactive gastritis, type C gastritis,* and *chemical gastropathy* have also been used. Chemical gastropathy is now defined as the constellation of endoscopic and histological changes caused by chemical injury to the gastric mucosa. This is an implicit admission that this diagnosis can be made only when converging clinical and histopathological evidence is present. *Radiation gastritis*, which results from a physical agent rather than a chemical one, is described in Chapter 132.

Clinical Manifestations

Three categories of patients may exhibit the endoscopic and histological changes of chemical gastropathy: patients with alkaline reflux after partial gastrectomy, patients with duodenogastric bile reflux as part of a poorly understood dysmotility syndrome, and patients who take NSAIDs.

Postgastrectomy alkaline reflux may present with a syndrome characterized by burning midepigastric pain unresponsive to antacids and aggravated by eating and recumbency. [154], [155] Bilious vomiting, anemia, and weight loss may occur. Endoscopic confirmation of bile reflux and documentation of the characteristic histopathological findings support the diagnosis, and corrective surgery (e.g., creation of a 40- to 50-cm Roux-en-Y gastrojejunostomy) is successful in about half of all cases. [156]

Duodenogastric bile reflux secondary to gastroduodenal dysmotility or to cholecystectomy is a controversial condition, rarely considered in the differential diagnosis of dyspepsia in patients with an intact stomach. [157] Thus, the frequency of endoscopic or histological changes of chemical gastropathy in these patients is unknown.

Millions of persons take daily doses of *NSAIDs* for indefinite periods, in many cases for life, to control pain caused by osteoarthritis, rheumatoid arthritis, or other chronic conditions. Unknown numbers of these NSAID users experience epigastric pain; approximately 10% per year develop erosions or ulcers, and 1% to 2% per year have a major gastric bleeding episode. [158], [159] Reactive gastropathy has been documented histopathologically in 10% to 45% of long-term users of NSAIDs, but no relationship could be established between the appearance of the mucosa and dyspeptic symptoms.

Pathogenesis

Duodenogastric reflux (with alkaline pancreaticoduodenal secretions as well as acids, bile salts, and lysolecithin) disrupts the mucous barrier and directly damages the gastric surface epithelium. [160] This combined injury leads to accelerated exfoliation of surface epithelial cells and a histamine-mediated vascular response that manifests as edema and hyperemia. Persistent epithelial damage may lead to the release of other proinflammatory agents, such as platelet-derived growth factor, which among its many actions stimulates smooth muscle and, later on, fibroblastic proliferation. [161] Epithelial injury after exposure to NSAIDs appears to be mediated by reduced prostaglandin synthesis. Prostaglandins are important cytoprotective agents in the gastric mucosa and exert their effects by maintaining mucosal blood flow, by increasing secretion of mucus and bicarbonate ions, and by augmenting epithelial defense against cytotoxic injury. Thus, NSAID-inflicted injury can be partially prevented by simultaneous administration of prostaglandin analogs such as misoprostol [162] and by suppression of gastric acid production with proton pump inhibitors. [163] Newer selective cyclooxygenase 2 inhibitors (inhibitors, second-generation NSAIDs, or selective NSAIDs) are reportedly better tolerated by the gastric mucosa. [164] Whether the initial encouraging results will hold true when these drugs become widely used, remains to be evaluated.

Endoscopic Appearance

In patients who have undergone a Billroth II gastrectomy, the gastric mucosa at the anastomotic site may have a polypoid appearance with congestion, edema, and friability. [165] Superficial erosions may be present in more proximal areas of the gastric stump, but they are not specific, because they can be caused by a variety of injuries. In patients with nonoperated stomachs and possible duodenogastric bile reflux, the mucosa may exhibit congestion, edema, and erosions. In long-term NSAID users, the mucosa may be normal or may show congestion, erosions, or ulcers.

Histopathology

Although some histological features are more frequently found in regular NSAID users, the histopathological diagnosis of chemical gastropathy remains a challenging problem. The histopathological changes that have been associated with reactive gastropathy (see Fig. 68-4A) include evidence of epithelial regeneration, foveolar hyperplasia, edema of the lamina propria, and expansion of the smooth muscle fibers into the upper third of the mucosa, an area in which they are not normally found. Both the specificity and the predictive value of these features are low because of several potential confounding factors. Surreptitious use of NSAIDs, other substances in the diet (e.g., alcohol, spices, salt) that may cause similar mucosal changes, and clinically silent bile reflux [153], [157] can rarely be excluded; furthermore, the difficulty of obtaining a perfect control population cannot be overemphasized. *H pylori* infection induces some of the features traditionally considered characteristic of chemical gastropathy: mucosal hyperemia and edema, superficial erosions, foveolar hyperplasia, and regenerative changes. Therefore, the pathologist can suspect chemical gastropathy and communicate this suspicion to the clinician, but a firm histopathological diagnosis can be made only when supportive clinical data are available and *H pylori* infection is absent. [5]

Partial Gastrectomy and Carcinoma

The polypoid appearance of the distal portions of the gastric stump in patients who have undergone gastrectomy has been referred to as *gastritis cystica polyposa.* [166] Several studies, mostly conducted in Europe and Asia, have reported a high prevalence of low-grade dysplasia in these polypoid areas, as well as an increased incidence of gastric adenocarcinoma 2 to 3 decades after gastrectomy. [167], [168] The implications of these findings, potentially important because of the suggestion that patients who undergo partial gastrectomy may need endoscopic surveillance, seem less urgent in the present era, when partial gastrectomy for peptic ulcer disease

has become an exceedingly rare operation.

HEMORRHAGIC GASTROPATHY

Hemorrhagic gastropathy (hemorrhagic gastritis) refers to a group of conditions characterized by subepithelial hemorrhages and erosions. [169] These mucosal lesions do not cause major bleeding unless ulcers develop. Three major factors are involved in the pathogenesis of hemorrhagic gastropathy: use of NSAIDs, ingestion of large quantities of alcohol, and severe physical stress.

Aspirin and other NSAIDs may induce mucosal edema, hyperemia, and multiple erosions and ulcerations. Such lesions may occur suddenly, without warning symptoms such as pain or discomfort, in first-time NSAID users and in patients who have taken NSAIDs regularly for years. Except for generic risk factors such as older age, female sex, and previous episodes, there is no known trait to identify NSAID users prospectively who may be susceptible to severe gastric injury.

Similar mucosal lesions, although usually less severe and only rarely evolving to ulcerations, can be caused by ingestion of large quantities of alcohol. [169, 170] Because alcohol and aspirin may act synergistically to break down mucosal defenses, one wonders how many hemorrhagic gastropathies have been caused by attempts to prevent hangovers by taking aspirin after an alcoholic binge. [171]

The most severe degrees of hemorrhagic gastropathy are those induced by stress. Originally described in 1842 by Thomas Curling, [172] stress-induced gastroduodenal mucosal breakdowns (known as Curling ulcers) can be found in most patients admitted to an intensive care unit; approximately 20% are a source of overt bleeding, and 2% to 5% of these ulcers cause life-threatening hemorrhage. [173]

Pathogenesis

The pathogenesis of stress-induced hemorrhagic gastritis is not known, but lumenal acid seems to be essential. Acid exerts its deleterious effects when the mucosal defense mechanisms (e.g., the mucous-bicarbonate barrier and the epithelial layer) lose their integrity. [173] Vascular disturbances—in association with stasis, vasoconstriction, and increased vascular permeability—may further contribute to mucosal vulnerability. Aspirin and NSAIDs act by interfering with prostaglandin synthesis, as noted earlier. Alcohol causes direct damage to the gastric mucosa; at a concentration of 12.5% (that of table wine), it induces hyperemia and petechiae, and concentrations greater than 40% (“hard liquors”) cause necrosis of the surface epithelium and capillaries and subsequent interstitial hemorrhage. [169, 174]

Clinical Manifestations

Acute hemorrhagic gastritis is characterized by a hyperemic edematous mucosa with erosions and various degrees of active bleeding. The clinical history (e.g., shock, burns, recent alcohol binge, ingestion of aspirin), rather than the widely overlapping nature of the lesions, will help the endoscopist to determine the precipitating factors. Because the diagnosis is often clear from the clinical context, gastric biopsies are rarely obtained from critically ill patients who undergo endoscopy because of upper gastrointestinal hemorrhage.

Histopathology

Acute hemorrhagic gastritis, irrespective of its cause, is characterized by dilation and congestion of the capillaries, edema, and various degrees of interstitial hemorrhage in the lamina propria. Epithelial erosions are generally small; aggregates of fibrin and polymorphonuclear cells replace the eroded epithelium and may project above the surface to form small, elevated clumps of necrotic debris. In the absence of concurrent *H pylori* infection, there is no significant inflammation in the unaffected areas of the stomach. [169] Therefore, a diagnosis of erosive hemorrhagic gastritis can usually be made, and possible etiologic agents may be listed (e.g., alcohol or NSAID ingestion). If *H pylori* gastritis is present, widespread active inflammation often obscures or worsens the changes caused by other agents, and a separate diagnosis of the diverse causes of the changes is usually impossible.

Management

The suppression of acid with H_2-receptor antagonists or proton pump inhibitors helps to reduce the severity of the mucosal damage and facilitates healing. Sucralfate, although less commonly used, is also effective. [159, 169, 170]

VASCULAR GASTROPATHIES

Gastric vascular gastropathies [175] comprise a heterogeneous group of conditions characterized by alterations in the gastric circulation and their effects on the gastric mucosa. The best-defined conditions, from morphologic and pathogenetic viewpoints, are the *watermelon stomach,* which is described in Chapter 130, and *portal hypertensive gastropathy.*

Portal Hypertensive Gastropathy

Clinical Manifestations and Pathogenesis *Portal hypertensive gastropathy* is a dilation of the mucosal vessels, more prominent in the proximal stomach, that occurs in a proportion of patients with cirrhosis of the liver [176]; its prevalence parallels the severity of portal hypertension. Bleeding from this lesion is relatively uncommon and rarely severe; in general, patients with the most severe portal hypertension have diffuse lesions and higher rates of gastric bleeding. [177]
Endoscopic Appearance The endoscopic appearance of portal hypertensive gastropathy is nonspecific and does not correlate well with the degree of portal hypertension. The endoscopic patterns have been variously described as snake skin, scarlatina rash, cherry-red spots, and mosaic. [178] The mosaic pattern was found by a consensus conference [179] to be the most reliable indicator of mild portal hypertensive gastropathy with a low risk of hemorrhage. Red marks suggest a more severe degree of hypertension and a greater risk of hemorrhage.
Histopathology Dilation and tortuosity of small arteries and veins, with occasional thickening of the walls, are among the pathological changes of portal hypertensive gastropathy. The changes are more prominent in the corpus and are more apparent in submucosal vessels than in mucosal capillaries. Because of the location of these changes and the understandable reluctance of most gastroenterologists to obtain large and deep biopsy samples from patients who may have an increased risk of bleeding, the diagnosis of portal hypertensive gastropathy is not commonly reached by evaluating mucosal biopsies. In patients with concurrent *H pylori* gastritis, it is difficult, if not impossible, to separate the respective contribution of infection and congestion to the resulting complex of changes observed in the mucosa. [180, 181]
Management Because bleeding is relatively uncommon, most patients do no require specific therapy. Sclerotherapy of esophageal varices does not seem to influence the natural history of this condition. [177] In severe cases, portal decompression surgery, but not transjugular intrahepatic portosystemic shunting, is effective in reducing the risk of hemorrhage. [182]

HYPERTROPHIC GASTROPATHIES

In numerous inflammatory and noninflammatory conditions, the mucosal folds (*rugae*) that confer a rugged appearance to the normal gastric corpus may become extremely enlarged and may give the mucosa an appearance that has been compared to that of the cerebral cortex (cerebriform). Based on this macroscopic morphologic characteristic, shared in various degrees by all of them, certain etiologically heterogeneous conditions have been categorized as *hypertrophic gastritis, hypertrophic gastropathy,* or, more recently, *giant-fold gastropathies.* To complicate our understanding of these diverse conditions further, many authors have used indiscriminately the term *Ménétrier disease,* after the condition described in 1888 by the French gastroenterologist who initially called it *polyadenomes en nappe* (polyps in layers). [183]

In 1973, Ming [184] introduced the term *hyperplastic gastropathies* and proposed a classification that recognizes three main histological types:

1. Foveolar hyperplasia, with normal or atrophic oxyntic glands and corresponding to Ménétrier’s description
2. Hyperplasia of oxyntic glands, with a largely unaffected mucous component and corresponding to Zollinger-Ellison syndrome
3. A mixed type, in which both mucous and oxyntic glands show variable degrees of hyperplasia; it incorporates various conditions that may cause mixed glandular hyperplasia, including infections (*H pylori* infection, CMV infection in children, syphilis, and histoplasmosis) and other diseases of uncertain origin (lymphocytic

gastritis, eosinophilic gastroenteritis, sarcoidosis, and Cronkhite-Canada syndrome).

Zollinger-Ellison syndrome is covered in Chapter 67. Cronkhite-Canada syndrome is discussed in Chapter 90.

Ménétrier Disease

Ménétrier, in his original description, included patients with multiple hyperplastic polyps and patients with giant folds and focused his observations on the increased gastric cancer risk of these patients. Today, the condition associated with his eponym is defined as an idiopathic diffuse enlargement of the gastric folds in the antrum. Histologically, glands show massive foveolar cell hyperplasia with normal or slightly reduced numbers of parietal and chief cells. Neither significant inflammatory infiltrates in the lamina propria nor epithelial lesions (erosions, intestinal metaplasia, or cellular atypia) are present. If chronic active inflammation or lymphocytic infiltration of the epithelium is seen, the large-fold type of *H pylori* gastritis or lymphocytic gastritis should be considered. In children, giant folds are almost always associated with CMV infection.[185] This association has rarely been reported in adults. The hyperplastic foveolar cells secrete large amounts of mucus and fluid resulting in protein-losing enteropathy (found in almost all patients) and hypoacidity (in part from dilution of the acid produced by oxyntic cells).

Pure Ménétrier disease is a rare condition, with an estimated prevalence of no more than 300 or 400 persons worldwide. Patients are more often men in their fifth or sixth decade who present with dramatic weight loss, epigastric and abdominal pain, nausea, and vomiting. Later in the course of the disease, usually protracted for several years and even decades, virtually all patients develop hypoalbuminemia as a consequence of the protein-losing enteropathy. The pathogenesis is unknown, but an interesting hypothesis has been put forward. Transforming growth factor-a (TGF-a), a critical mediator of gastric mucosal homeostasis normally produced by the gastric mucosa, inhibits acid secretion, stimulates mucosal repair, cell migration, and proliferation, and augments gastric mucin levels. Overproduction of TGF-a could explain several of the disturbances occurring in Ménétrier disease.[186] Support for this hypothesis is further lent by the successful treatment of some patients by a monoclonal antibody against the TGF-a receptor (the epidermal growth factor receptor).[187] Other forms of treatment, including antacids, anticholinergic drugs, corticosteroids, H_2-receptor antagonists, and proton pump inhibitors, have been proven consistently ineffective. Eradication of *H pylori* has been reported to have successfully cured patients with Ménétrier disease.[188], [189] One of the possible pathological aspects of *H pylori* gastritis is the development of large, edematous folds,[190] and this variant may have been interpreted as a type of *H pylori*–induced Ménétrier disease. Not surprisingly, it regresses after the eradication of *H pylori* infection.

GASTRIC CARDIA

Since the 1970s, a rise in the incidence of *adenocarcinoma of the cardia* has occurred in the very populations in which the incidence of gastric cancer has been decreasing.[191], [192] Thus, there has been an emerging interest in exploring the pathology of this relatively ill-defined area of the stomach. Virtually all patients who have *H pylori* gastritis in other regions of the stomach also have bacteria and inflammation in the region immediately distal to the gastroesophageal junction.[193], [194] and [195] Some patients, including those without *H pylori* infection and patients with and without gastroesophageal reflux disease, have a localized inflammation of the cardia, commonly referred to as *carditis* (Fig. 68-13).[196] Intestinal metaplasia is found in the cardia of approximately 20% of patients with a variety of gastric disorders, even in the absence of *H pylori* infection, and apparently also in healthy persons. The relationship between intestinal metaplasia in this location and adenocarcinoma of the junction remains unclear,[197], [198] but it is important to differentiate the intestinal metaplasia of the cardia from Barrett specialized epithelium. Cytokeratin 7 and 20 immunoreactive patterns may help to distinguish between intestinal metaplasia in long-segment Barrett esophagus and junctional metaplasia[199]; conversely, the epithelium of short-segment Barrett esophagus may be impossible to separate from metaplasia originating in the cardia.[200] Until the implications of such findings are clarified, it is advisable to collect as much specific information as possible carefully from each patient with changes at the gastroesophageal junction. Lesions of uncertain prognostic value today could acquire clinical significance when new knowledge is acquired.

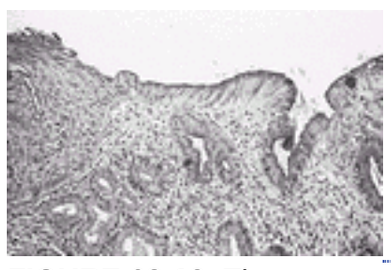

FIGURE 68-13. The squamocolumnar junction in a patient with gastroesophageal reflux and no *Helicobacter pylori* infection. The marked inflammation in the cardia contrasts with the lack of infiltrate in the squamous epithelium. The term “non- *H pylori* carditis” has been used for this entity of uncertain origin and significance.

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CHAPTER 69

Wai K. Leung, Enders K. W. Ng, and Joseph J. Y. Sung

TUMORS OF THE STOMACH

Gastric cancer is the second most common cause of cancer mortality, after lung cancer, worldwide. Although relatively uncommon in the United States, gastric cancer is the most common cancer in many Asian countries. With the changing global socioeconomic environments, and also possibly with the improvements in sanitary conditions, the incidence of gastric cancer is on a declining trend in many developed countries, a finding suggesting that this disease is preventable.

Because most gastric tumors are adenocarcinomas, that is the focus of this chapter. Other less common gastric malignant diseases such as lymphoma, stromal tumor, and gastric polyps are discussed briefly.

EPIDEMIOLOGY

Gastric cancer is the most common gastrointestinal malignant disease and is only second to lung cancer in global cancer mortality (Fig. 69-1). It is estimated that gastric cancer causes more than 620,000 deaths per year worldwide. [1] In the United States, approximately 21,500 new cases of gastric cancer were diagnosed in 2000, and 13,000 patients died of this disease the same year. [2]

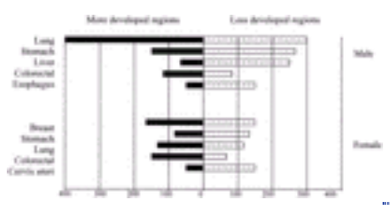

FIGURE 69-1. Estimated number of deaths (in thousands) from five leading cancers in men and women of more developed and less developed regions of the world.

In many developed countries, the incidence of gastric cancer is slowly declining. As shown in the time-trend analysis of the United States (Fig. 69-2), the incidence of gastric cancer decreases with time in both sexes. A similar pattern is observed in other parts of the world, including Japan, but the exact reason remains unknown. Nonetheless, because of the rapidly growing populations in developing countries with a high incidence of gastric cancer, the total number of gastric cancer cases in the world has actually remained the same as in the 1990s. [1] Murray and Lopez [3] projected that the annual number of new cases will continue to increase in the developing world over the next few decades as a result of population growth. Notably, the reduction in gastric cancer incidence largely reflects a decline in cancers in the distal stomach and mostly of the intestinal type. Conversely, there has been a steady rise in the incidence of adenocarcinoma of the proximal stomach and the gastroesophageal junction since the 1970s. [4], [5] From 1970 to 1995, hospitalization and death rates for gastric cancer in the United States Department of Veterans Affairs database fell, whereas the hospitalization rates for gastroesophageal reflux disease rose significantly. [6] According to the United States National Cancer Institute Surveillance, Epidemiology, and End Results database, an annual increase in proximal gastric lesions was estimated to be 3.6% to 5.6%. [4] This increase of gastric cancer in the proximal stomach is alarming because the increase on percentage basis is even greater than that of lung cancer or melanoma, and this disease is associated with poorer prognosis.

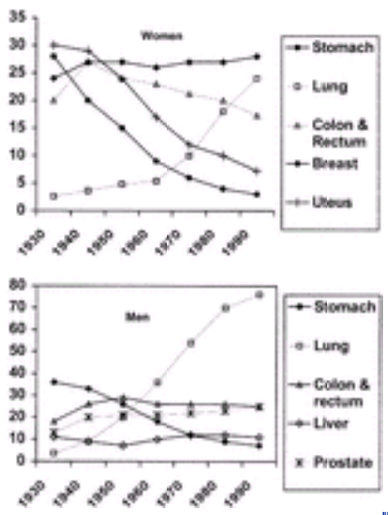

FIGURE 69-2. Time trend of the age-adjusted cancer death rates (per 100,000 population) for women and men in the United States from 1930 to 1996.

In all ethnic groups and countries, gastric cancer has a predilection for elderly patients, with incidence peaks at the seventh decade. Men are more commonly affected than women. Incidence rates in men are double those in women. Besides gender differences, the incidence of gastric cancer also appears to be inversely related to social class. Rates are higher in unskilled laborers (class V) and lower in professional groups (class I). [7] Conversely, there are considerable geographic variations in the incidence of gastric cancer (Table 69-1). The highest rates are observed in East Asia (the age-adjusted standardized rate of male patients is 44 years), where gastric cancer is still the most frequent cancer in men. The age-adjusted standardized rate of gastric cancer is 35 per 100,000 in Japan and China, [1] whereas the corresponding figure in the United States is less than 5 per 100,000. Even within the same country, variations in cancer incidence and mortality are also observed among different ethnic groups. In the United States, African Americans, Hispanic Americans, and Native Americans are 1.5 to 2.5 times more likely to have gastric carcinoma than are whites. [8] Similarly, in Singapore, the incidence of gastric cancer in Chinese persons is about twice that of Indians and is five times greater than that of Malays. [9] The reason of this discrepancy is unclear. It has been attributed to differences in socioeconomic factors and dietary and environmental conditions. In Japan and China, there appears to be a dichotomized distribution in which mortality and incidence of gastric cancer are higher in the northern parts than in the southern parts of those countries. [10], [11]

GEOGRAPHIC AREA	MALE	FEMALE
North America		
United States	4.4	2.0
Canada	6.2	3.0
Mexico	9.7	7.1
Central and South America		
Chile	32.2	11.7
Colombia	21.4	13.1
Asia Pacific		
Australia	6.6	2.7
China	26.9	12.7
Japan	30.2	12.3
New Zealand	6.0	3.2
Europe		
Finland	10.2	4.7
Germany	12	6.3
Hungary	18.8	8.7
Poland	18.9	6.8
Portugal	21.8	10.0
Russian Federation	36.9	15.3
Sweden	6.6	3.5
United Kingdom	9.5	3.9

Adapted from ref. 2.

TABLE 69-1 Age-Adjusted Gastric Cancer Death Rates per 100,000 Population from Different Countries, 1994–1997

ETIOLOGY

With the contrasting global epidemiology of gastric cancer, tremendous effort has been made to look into the causation of this malignant disease. It is now generally believed that gastric cancer has multiple etiologic factors including diet, exogenous chemicals, intragastric synthesis of carcinogens, genetic factors, and infectious agents (Table 69-2). Of these, the most extensive studied factors are dietary habits and *Helicobacter pylori* infection.

FACTOR	SOURCE
Diet	Consumption of salted, smoked, and poorly preserved foods
	Low consumption of fruits and vegetables
Infection	*Helicobacter pylori*
	Epstein-Barr virus
Genetic	Hereditary nonpolyposis colon cancer syndrome
Previous gastric resection	

TABLE 69-2 Possible Etiologic Factors in Gastric Cancer

Dietary Factors

Although dietary habits may influence the development of gastric cancer, it is extremely difficult to measure food consumption accurately. Most of these studies were based on retrospective data collection, which could be subject to recall bias. In spite of these caveats, many epidemiologic studies from regions with a high incidence of gastric cancer have identified several dietary factors to be related to the high cancer incidence and mortality. Apart from case control studies on dietary contents, migrant studies also provide invaluable information relating to the role of diet in gastric carcinogenesis. Migration is accompanied by changes in lifestyle, especially diet. The risk of developing gastric cancer changes slowly in populations moving from a high-risk community to a low-risk community. Japanese persons who migrated to the United States had a significant reduction in mortality from stomach cancer when compared with the native Japanese population. [12] These observations implicate the role of environmental factors in the pathogenesis of gastric cancer, with diet the most likely cause.

Most of the case control studies have shown a rather consistent inverse relationship between the consumption of fresh fruits and vegetables and the risk of gastric cancer. [13], [14], [15], [16] and [17] This observation may be partly related to the presence of vitamins present in fruit and vegetables, such as vitamins C and E, carotenoids and flavonoids. These vitamins may act through various mechanisms, such as antioxidant effects, the ability to inhibit nitrosamine formation, [18] and dilution or binding of carcinogens, thereby reducing the risk of cancer development. An epidemiologic study from China demonstrated that the risk of progression to gastric dysplasia and carcinoma was reduced by 80% (95% confidence interval, 30% to 90%) in study subjects with baseline ascorbic acid levels in the highest tertile when compared with those in the lowest tertile. [19]

Conversely, food substances rich in nitrates, nitrites, and secondary amines, which combine to form *N*-nitroso compounds, are known mutagens or carcinogens. Nitrates and nitrites were previously used in food preservation before the era of refrigeration. Consumption of preserved meat and vegetables has been consistently linked to an increased risk of developing gastric cancer. [14], [20] Humans are exposed not only to preformed *N*-nitroso compounds, but also to a wide range of precursors and nitrosating agents that can react in vivo to form potentially carcinogenic *N*-nitroso compounds. This process is facilitated in the presence of nitrate-reducing bacteria of the digestive tract. With the advent of refrigerators and the growth of the frozen food industry, consumption of preserved meat and vegetables declined in most developed countries. This has been postulated to be a crucial factor in the reduction of gastric cancer incidence in North America and Western Europe. [21]

High intake of carbohydrates and salt has also been suggested to increase the risk of stomach cancer. A high-salt diet in humans and experimental animals is associated with a higher risk of atrophic gastritis, a preneoplastic lesion. [22], [23] Salt also enhances the colonization of *H pylori* in mice and may therefore perpetuate the development of gastritis and glandular atrophy. [24] In Japan, a significant correlation was found between the amount of salt excreted in urine and stomach cancer mortality rates. [25] A study from 65 Chinese counties also recognized a significant positive association between stomach cancer mortality and consumption of salted vegetables. [26] Conversely, consumption of a carbohydrate-rich diet was also found to have a positive association with gastric cancer. A study from Shanghai showed a 1.5- to 1.9-fold increase in risk of gastric cancer for the highest quartile of carbohydrate intake. [17] Nonetheless, this association was not confirmed in other studies, [27] and the mechanisms of carbohydrate and gastric carcinogenesis remain poorly defined. Notably, the estimation of carbohydrate consumption is usually confounded by the amount of salt intake.

The polyphenols in green tea have been postulated to have an anticarcinogenic effect, including gastric cancer, through an antioxidant effect. This hypothesis is particularly attractive to Asian investigators. Several case control studies suggested the inverse association between consumption of green tea and risk of gastric

cancer. [28] However, the anticarcinogenic effect of green tea was refuted by a large-scale, population-based, prospective cohort study in Japan that involved more than 26,000 residents. [29] The role of green tea in the prevention of gastric cancer needs further evaluation.

Infectious Agents

Helicobacter pylori *H pylori* is a gram-negative, spiral-shaped organism found in the mucus layer of the human stomach. It was first successfully cultured by Marshall and Warren in 1984. [30] Since the discovery of this pathogen, numerous reports had been published to link this bacterium with various gastroduodenal diseases and even extraintestinal conditions. Based on several large-scale epidemiologic cohort studies published in the early 1990s, [31], [32], [33] and [34] the International Agency for Research on Cancer classified *H pylori* as a group 1 carcinogen in 1994. [35] Data from case control studies were summarized by a metaanalysis of 19 studies that included 2500 cases and 4000 controls. [36] The pooled data showed that the combined odds ratio for gastric cancer in *H pylori*–infected subjects is 1.92 (95% confidence interval, 1.32 to 2.78). Nonetheless, the attributable risk of *H pylori* infection to gastric cancer is still believed to be an underestimate. Because elderly patients with severe atrophic gastritis may have spontaneous remission of infection, the association of *H pylori* with gastric cancer is particularly strong in young persons with *H pylori* infection. In this regard, the odds ratio increased from 1.05 at age 70 years or older to 9.29 at age 29 years or younger (Fig. 69-3).

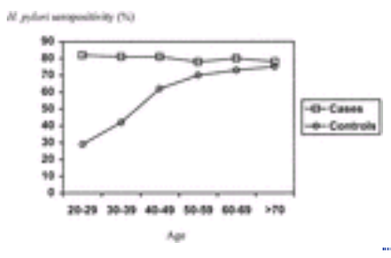

FIGURE 69-3. Age distribution of *Helicobacter pylori* seropositivity in gastric cancer cases and controls. (Modified from ref. [36].)

It is well known that *H pylori* infection is associated not only with gastric cancer but also with gastric and duodenal ulcers. Factors determining individual susceptibility to diverse disease outcomes remain poorly understood. In a large Swedish cohort study, the standardized incidence ratio for gastric cancer among patients with gastric ulcers was 1.8 (95% confidence interval, 1.6 to 2.0), whereas in patients with duodenal ulcer it was only 0.6 (95% confidence interval, 0.4 to 0.7). [37] This paradoxical relationship between duodenal ulcer and gastric cancer may be at least partially related to the distribution of gastritis. [38], [39] In some patients, chronic *H pylori* infection results in a corpus-predominant type of gastritis. The profound acid suppression predisposes them to the development of gastric ulcer and gastric carcinoma. Conversely, chronic *H pylori* can lead to antral-predominant gastritis in other patients that favors the development of duodenal ulcer (Fig. 69-4).

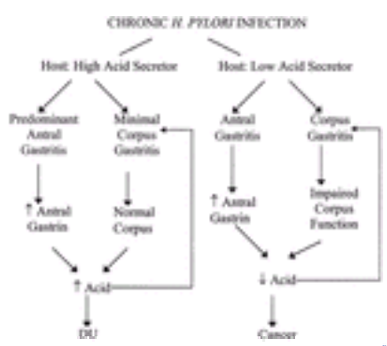

FIGURE 69-4. Divergent host response to *Helicobacter pylori* infection. (Modified from McColl KE, et al. *Helicobacter pylori* gastritis and gastric acid secretory function: an integrated approach. In: Hunt RH, Tytgat GN, ed. Basic mechanisms to clinical cure. Boston: Kluwer Academic Publishers, 1996.)

H pylori infection is linked to both intestinal and diffuse-type cancers. However, only noncardiac or distal gastric tumors are associated with this bacterium. Data from the National Cancer Institute even suggest an inverse relationship between *H pylori* infection and proximal gastric carcinoma [40]; that is, *H pylori*–infected persons had a reduced risk of gastroesophageal and cardia cancers. The logic of this observation may be related to the suppressed acid output in persons with *H pylori* infection. [41], [42] The presence of corpus gastritis protects them against gastroesophageal reflux and hence Barrett esophagus, the precursor of adenocarcinoma of distal esophagus and proximal stomach. Despite the availability of a wealth of epidemiologic data, some investigators are still skeptical about the causal relationship between *H pylori* infection and gastric cancer. In particular, gastric cancers still occur in persons without evidence of *H pylori* infection, and not all infected persons will develop cancer during their lifetime. An animal study from Japan may clear this skepticism. In the absence of exogenous mutagens, Japanese investigators demonstrated that Mongolian gerbils infected with *H pylori* developed severe active chronic gastritis, ulcers, and intestinal metaplasia within 6 months of infection. [43] After 1 year, adenocarcinoma of stomach was detected in one third of the infected gerbils. Although these data support a direct gastric carcinogenic effect of *H pylori,* most workers believe that gastric cancer is a multifactorial disease involving genetic and dietary factors. With the large number of *H pylori*–infected persons globally, it is difficult to explain why so few of them develop gastric cancer. In particular, the unexpectedly low incidence of gastric cancer in African countries with high prevalence of *H pylori* infection remains elusive. [44] It was found that concurrent helminth and *H pylori* infection modulates the type 1 T helper cell (Th1) responses, which normally promote cell-mediated immune responses and tissue injury, to a less damaging Th2 response. [45] Thus, the progression of *Helicobacter*-associated gastric atrophy, a premalignant lesion, is attenuated. This finding illustrates the potential interaction between *H pylori* infection and environmental factors that may partly explain the “African enigma.”

Epstein-Barr Virus The virus most extensively studied as an etiologic factor in gastric cancer is Epstein-Barr virus (EBV). EBV, an icosahedral herpevirus containing linear double-stranded DNA, is ubiquitous in all human populations. It was estimated that about 10% of gastric carcinomas throughout the world show monoclonal proliferation of EBV-infected cancer cells. [46] In contrast to Burkitt lymphoma and nasopharyngeal carcinoma, which are endemic in Africa and Southeast Asia, EBV-positive gastric carcinoma is a nonendemic disease distributed throughout the world. The highest rates are reported in the United States and Germany (16% to 18%), and the lowest rates are observed in China (3.1%). [46] Although EBV has been detected in all types of gastric cancer, it is closely linked to a form of undifferentiated gastric carcinoma called *lymphoepithelioma-like carcinoma.* Histologically, these tumors resemble nasopharyngeal carcinomas. They tend to be located proximally, they are of diffuse histological types, and they have a lower frequency of lymph node metastasis. [47] The prevalence of *H pylori* infection in this tumor is also found to be lower than those with cancer that is not lymphoepithelioma-like carcinoma. In EBV-associated gastric cancer, the EBV genome was detected in the adjacent preneoplastic lesions. [48]

Tobacco and Alcohol Although several studies have shown a 1.5- to 3.0-fold increase in the risk of gastric cancer among smokers, a clear dose-response relationship cannot be demonstrated. [49] Moreover, smoking may be related more to cancer in the cardia than to cancer in the distal stomach. Given the rapid rise in lung cancer incidences and the progressive fall in stomach cancer incidences worldwide (see Fig. 69-2), tobacco use is unlikely to be an important etiologic factor in gastric cancer. Reports of a positive association between smoking and stomach cancer may be confounded by other factors, such as lower socioeconomic class and dietary habits. Conversely, epidemiologic studies failed to show any significant correlation between alcohol use and stomach cancer. [15], [16]

Previous Gastric Resection

Many studies have suggested an increased incidence of gastric cancer in the remaining stump of the stomach after gastrectomy. An incidence of 3% was reported in a previous study. [50] Rates of gastric cancer appear to remain low for 15 to 20 years after resection of the distal stomach and increase substantially thereafter. [51], [52] Notably, Billroth II gastrectomy was associated with higher risk than Billroth I gastrectomy. This may be explained by the changes in gastric environment after acid-reducing surgery. As gastric surgery reduces acid production in the stomach and causes neutralization of the gastric milieu, the lumenal bacterial contents will increase. Of particular interest is the presence of anaerobic bacteria in the gastric remnant that could convert nitrates from the diet into nitrites, a potential gastric carcinogen. In addition, bile reflux after gastric surgery promotes gastric epithelial proliferation in the stomach remnant and possibly favors cancer development. [53]

Genetic Factors

Considerable evidence suggests the role of genetic factors in gastric carcinogenesis. Familial clustering of gastric cancer has been reported in the literature. The best-known example comes from the famous Bonaparte family of France. [54] Napoleon’s father and grandfather died of the disease, and so did several of his siblings.

Case control studies showed that first-degree relatives of patients with gastric cancer have a two- to threefold increase in the risk of gastric cancer. [55] Yet, a single gene defect to account for this disease clustering has not been found, except in family kindreds of hereditary nonpolyposis colorectal cancer syndrome. Conversely, the familial clustering of gastric cancer can be potentially explained by the socioeconomic status and the common exposure to various environmental factors, such as *H pylori* infection [56] and diet. It was shown that relatives of patients with gastric cancer had an increased prevalence of hypochlorhydria and gastric atrophy, [57] which were linked to the polymorphisms of the interleukin-1ß gene. [58] Interleukin-1ß is a proinflammatory cytokine as well as a potent inhibitor of gastric acid secretion. This finding may provide the missing link between genetic factors in the host and bacterial factors. However, further testing in different ethnic groups is still needed to confirm this observation.

In contrast to colorectal cancer, which has well-defined mechanistic pathways of carcinogenesis, the picture is far from clear for gastric cancer. Since the early 1980s, numerous genetic alterations have been implicated in the carcinogenesis of gastric cancer. To a certain extent, many of these alterations are akin to those found in colorectal cancer and were initially described as inactivation of tumor suppressor genes or activation of oncogenes. However, as our understanding of carcinogenesis broadens, other alterations, such as genes involved in apoptosis and proliferation control, cell cycle regulations, overexpression of growth factors, and genomic instability, have also been found to play a role. Mutations of tumor suppressor genes such as *p53* and *APC* have been reported in gastric cancer. *p53* is involved in the induction of apoptosis after DNA damage. Loss of heterozygosity in chromosome 17 or mutations of *p53* were observed in more than 60% of gastric cancer. [59], [60], [61], [62] and [63] Similarly, *APC* mutations were detected in more than 50% of well-differentiated, but not in any poorly differentiated, gastric cancers. [64] However, activation of protooncogenes, such as the *ras* oncogenes and c- *myc,* was infrequently detected in gastric cancer. [65] In addition, a broad spectrum of regulatory peptides is also incriminated in the gastric carcinogenesis pathways. This includes epidermal growth factor, transforming growth factor, platelet-derived growth factor, insulin-like growth factor II, and basic fibroblast growth factor.

Microsatellite instability (MSI), a form of genomic instability and a manifestation of cell defective in DNA mismatch repair, is present in 17% to 48% of gastric cancers. [66], [67], [68], [69], [70], [71] and [72] Gastric cancers with MSI phenotypes tend to have specific clinicopathological characteristics such as intestinal type, prominent lymphocyte infiltration, and location in the antrum, and they are associated with better prognosis. [67], [68], [69], [70] and [71] Interestingly, MSI can also be detected in gastric intestinal metaplasia, a preneoplastic lesion, in patients without gastric cancer. [72], [73] In contrast to hereditary forms of colorectal cancers with MSI phenotypes, germline mutations of DNA mismatch repair genes are infrequent in MSI-positive gastric cancer. It was found that hypermethylation of the promoter region of *hMLH1,* a DNA mismatch repair gene, that results in transcriptional silencing of the gene is the major mechanism underlying MSI in sporadic gastric cancers. [74], [75]

Cyclooxygenase-2 (COX-2) has also been implicated in the development of gastric cancer. COX-2 is an inducible form of prostaglandin synthase responsible for the conversion of arachidonic acid into prostanoids. Up-regulation of the *COX-2* gene and overexpression of COX-2 enzyme are frequently detected in cancer. In colorectal cancer, overexpression of COX-2 confers resistance to apoptosis, higher metastatic potential, and angiogenesis. [76], [77] and [78] Previous studies showed that approximately 70% of gastric cancers demonstrated COX-2 expression. [63], [79], [80], [81] and [82] Gastric carcinomas with COX-2 overexpression tend to have a higher frequency of lymphatic invasion, [80] and they are associated with a worse prognosis. [81], [82] Interestingly, COX-2 expression was also detected in *H pylori*–associated gastritis [83], [84] and [85] and intestinal metaplasia. [86] It thus appears that the persistence of COX-2 in *H pylori*–associated chronic gastritis may contribute to the pathogenesis of gastric cancer.

PATHOLOGICAL FEATURES

Adenocarcinoma accounts for 95% of the stomach cancers, whereas squamous cell carcinoma, carcinoid tumors, leiomyosarcoma, and lymphoma constitute the rest of the gastric tumors. Macroscopically, gastric cancers can be broadly categorized by their location and appearances. Categorization of tumors into proximal (cardia) versus distal (including body and antrum) cancers bears major etiologic significance. Distal cancers are more common in areas with a high gastric cancer incidence, whereas cardia cancers are more prevalent in whites from populations with a low background incidence of gastric cancer. The former may be closely related to chronic *H pylori* infection, whereas the latter is often the consequence of chronic gastroesophageal reflux disease and Barrett esophagus. Of interest, the fall in incidence of gastric cancer is more of the result of the decline in distal gastric cancer. By contrast, the incidence of gastroesophageal junction carcinomas is actually increasing in the United States [4] and in many other developed countries.

Gastric tumors can also be classified by their gross appearances. The macroscopic appearance has been classified by Borrmann into four types (Fig. 69-5). Type I represents polypoid lesions without necrosis or ulceration, type II encompasses fungating cancers that may contain superficial ulceration, type III represents ulcerated lesions infiltrating the gastric wall, and type IV tumors are diffusely infiltrating tumors or linitis plastica lesions. There is considerable overlap among the different subtypes, and this typing system does not appear to correlate with histological typing. Approximately 40% to 50% of tumors are fungating or polypoid in appearance, and similar numbers of tumors are ulcerative. *Linitis plastica* is an aggressive infiltrating tumor consisting of high-grade tumor cells accompanied by a marked desmoplastic reaction that results in a rigid stomach resembling a leather bottle. This form of tumor spreads through the submucosa and is associated with worse prognosis.

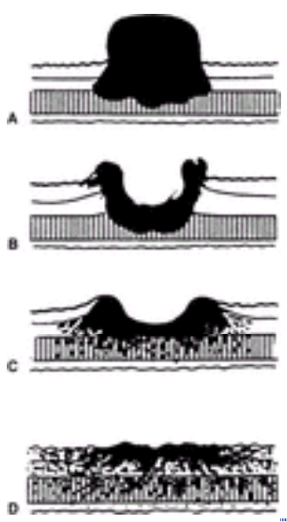

FIGURE 69-5. Macroscopic appearance of tumor as classified by Borrmann. (**A**) Polypoid. (**B**) Fungating with surface ulceration and hemorrhage. (**C**) Ulcerated without major lumen component. (**D**) Infiltrative. (From Borrmann R. Geshwulste des Magen und Duodenums. In: Henke F, Lunbarsch O, eds. Handbuch der speziellen pathologischen Anatomie und Histologie, vol 4. Berlin: Julius Springer, 1926:812.)

Microscopically, several histological classifications of gastric cancer are used. The most widely used classification, proposed by Lauren, [87] broadly divides gastric cancer into diffuse and intestinal types (Fig. 69-6; see also Color Fig. 69-6). Despite its simplicity, this classification categorizes tumors into two distinct types with contrasting epidemiology, etiology, and prognosis. The intestinal type is named by its histological morphology, and the diffuse type is characterized by its biologic behavior. Intestinal-type cancer is characterized by cohesive neoplastic cells forming glandular tubular structures, whereas diffuse-type cancer shows sheets of epithelial cells or cells scattered in a stromal matrix without evidence of gland formation. Diffuse cancers may contain signet-ring cells. Intestinal-type cancer is more common in the distal stomach and is often preceded by preneoplastic stages such as intestinal metaplasia. It is more prevalent in regions with a high background incidence of gastric cancer and is generally associated with a better surgical outcome than the diffuse type. Nonetheless, approximately 5% to 10% of tumors remain unclassified, and they are called mixed-type tumors. These unclassified tumors are usually regarded as diffuse-type tumors during clinicopathological evaluations.

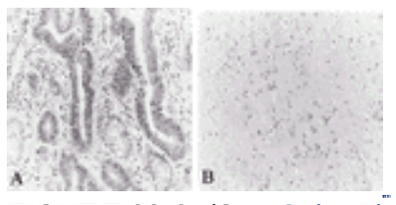

FIGURE 69-6. (See Color Fig. 69-6.) Lauren classification of gastric adenocarcinoma. (**A**) Well-differentiated intestinal-type gastric cancer. (**B**) Diffuse-type cancer with infiltrative and discohesive tumor cells. (Courtesy of Dr. K. F. To, Chinese University of Hong Kong, Prince of Wales Hospital, Hong Kong.)

Another classification system that is proposed by the World Health Organization [88] divided gastric tumors into ten types (Table 69-3). This classification is highly reproducible and is useful for routine pathological diagnosis of gastric cancer. Based on the degree of differentiation in architectural and cellular levels, gastric tumors can be divided into well-differentiated, moderately differentiated, or poorly differentiated types. Tubular adenocarcinoma, in which the glands form branching tubules, is the most common type of gastric adenocarcinoma. In papillary tumor, there are prominent intraglandular folding and projections with cuboidal to high columnar carcinoma cells lining the narrow bands of interstitial tissue in an arborizing pattern. Both types contain cells with absorptive type and tend to form a polypoid or fungating mass macroscopically. Signet-ring cell carcinomas are named because of the shape of the tumor cells that contain abundant intracytoplasmic mucus. There is minimal gland formation, and this type of tumor tends to infiltrate. Mucinous adenocarcinoma, also called mucoid or colloid carcinoma, is diagnosed when mucus occupies more than 50% of the tumor mass. [89]

Adenocarcinoma
Papillary
Tubular
Mucinous
Signet-ring cell
Adenosquamous
Squamous cell
Small cell
Undifferentiated
Others

Adapted from ref. 88.

TABLE 69-3 World Health Organization Histological Typing of Malignant Epithelial Tumors of the Stomach

Cascade of Gastric Carcinogenesis

Based on extensive observational studies in populations with high gastric cancer incidences, gastric cancer is generally believed to be a multistep progression from chronic gastritis, atrophy, and intestinal metaplasia ultimately to dysplasia and cancer. [90] This paradigm of gastric carcinogenesis has been coined the *Correa cascade* (Fig. 69-7), which was subsequently modified to incorporate *H pylori* in the initial stage of the sequence. [91] In this context, gastric atrophy and intestinal metaplasia are regarded as preneoplastic gastric lesions, particularly for intestinal-type cancer. These lesions are more prevalent in area with high gastric cancer incidences and are frequently linked to chronic *H pylori* infection. Both conditions are multifocal and are more prevalent in the antrum.

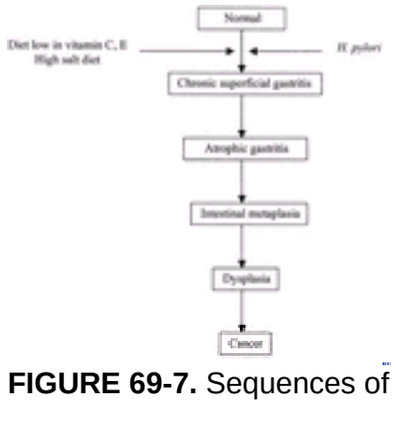

FIGURE 69-7. Sequences of gastric carcinogenesis.

Gastric atrophy usually predates the development of intestinal metaplasia and can be detected in 80% to 90% of patients with gastric cancer. It has been estimated that 10% of patients with chronic atrophic gastritis will develop gastric carcinoma in a 15-year follow-up period. [92] Because of the focal nature of gastric atrophy, the definitive diagnosis of gastric atrophy may be sometimes difficult, particularly with the sampling error of endoscopic biopsy. To evaluate the topographic distribution and extent of atrophy correctly, multiple representative samples are necessary. [93] By contrast, type A or autoimmune atrophic gastritis typically affects the corpus only, as compared with *H pylori*–associated multifocal atrophic gastritis, which involves both the antrum and the corpus. Patients with pernicious anemia are estimated to have a two- to threefold excess risk of stomach cancer. [94] However, the diagnostic yield from screening of these patients is generally considered to be low, and thus, the current recommendation by the American Society for Gastrointestinal Endoscopy involves only a single endoscopic study to identify the prevalent lesions such as carcinoid tumors or gastric cancer. [95] Currently, there are insufficient data to support the use of surveillance endoscopy in this group of patients.

The detection of small intestinal mucosa in the stomach can be dated back to the 19th century. [96] Intestinal metaplasia is recognized by the presence of goblet cells, absorptive cells, and cells resembling colonocytes or by its mucin content. It can be further divided into small intestinal and colonic types, or complete and incomplete types, on the basis of morphology and enzyme histochemistry. This classification of intestinal metaplasia into different subtypes is of clinical importance. By using mucin histochemistry, intestinal metaplasia can be divided into three types according to morphology and glycoprotein content. Type I (complete) refers to the presence of goblet cells containing sialomucins interspersed between nonsecretory absorptive cells with well-delineated brush borders. Type II (incomplete) indicates the presence of sialomucin-containing goblet cells scattered among gastric-type cells containing either neutral mucin or sialomucins. Type III (incomplete or colonic type) is diagnosed when there are tortuous and branched crypts lined by tall columnar cells containing abundant sulfomucins. Sulfomucins can be differentiated from sialomucins by using high iron diamine/Alcian blue staining (Fig. 69-8; see also Color Fig. 69-8). Complete or type I is generally believed to carry the lowest risk of gastric cancer, whereas type III (colonic) has the strongest association. In a large cohort study from Slovenia with 10 years of follow-up, patients with all types of intestinal metaplasia had at least a tenfold increased risk of cancer compared with those without intestinal metaplasia. [97] Furthermore, patients with type III intestinal metaplasia had a fourfold risk of cancer development compared with those with type I. In a cohort study from Shandong Province, China that involved more than 3000 subjects with a follow-up of 4 to 5 years, the odds ratio of progression from intestinal metaplasia to gastric cancer varied from 17 to 29. The risk was particularly high for those with more advanced baseline histological lesions such as the presence of dysplasia. [98] In spite of this finding, there are currently no data to support the use of screening endoscopy in these patients.

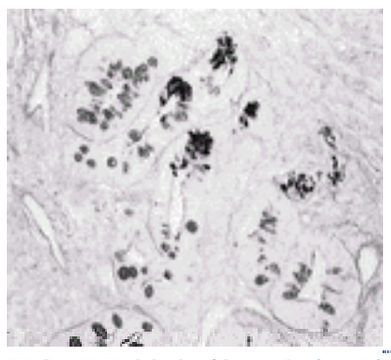

FIGURE 69-8. (See Color Fig. 69-8.) Type III intestinal metaplasia of stomach. The presence of sulfomucin in type III intestinal metaplasia is illustrated by the high iron diamine (*HID*)/Alcian blue staining.

Although it is universally accepted that dysplasia is a precursor of gastric neoplasia, the definition of dysplasia is controversial. The Japanese Research Society for Gastric Cancer (JRSGC) proposed a five-tiered group classification for interpretation of gastric biopsies, but this was not widely adopted by Western pathologists. Additionally, Japanese pathologists diagnose gastric carcinoma on nuclear and structural criteria, even in the absence of tumor invasion from a Western viewpoint. Thus, dysplasia according to Western pathologists is frequently interpreted as early carcinoma in Japan. [99], [100] Several consensus meetings have been held to arrive at a globally accepted histological classification of gastric dysplasia. [101], [102] However, the validity of these proposed classification systems is yet to be tested.

The question whether treatment of *H pylori* infection can actually prevent gastric cancer development remains unanswered. These studies are extremely difficult to perform because of the long cancer development process, which may take several decades. To look into the potential benefits of *H pylori* eradication, studies have been designed to intervene in the intermediate stages (i.e., intestinal metaplasia or atrophy) and to examine the regression of these changes as the surrogate end point of treatment success. An uncontrolled study from Japan demonstrated the regression of intestinal metaplasia at 12 months after *H pylori* eradication. [103] Gastric atrophy improved in 89% of patients, whereas intestinal metaplasia in the antrum improved in 61% of patients. Additionally, two large-scale, randomized, controlled studies confirmed that eradication of *H pylori* resulted in significant improvement of gastric histology, particularly intestinal metaplasia and gastric atrophy, when compared with patients with persistent infection. [104], [105] The study by Correa and colleagues [105] with a 6-year follow-up showed a significant regression of intestinal metaplasia and gastric atrophy. Interestingly, the supplementation of ß-carotene or ascorbic acid alone results in a similar degree of improvement in gastric histology. Whether it can translate into absolute reduction in cancer incidence awaits study with long-term follow-up.

Early Gastric Cancer

Early gastric cancer refers to tumor confined to the mucosa or submucosa, regardless of the presence or absence of metastasis. This disease entity carries an extremely good prognosis and is particularly prevalent in Japan, where mass screening of gastric cancer is performed. This entity is estimated to represent up to 65% of gastric cancers diagnosed in Japan. A 5-year survival rate of more than 90% was reported in Japan after gastrectomy and removal of primary and secondary lymph nodes. [106]

The JRSGC classified early gastric cancer into three types based on macroscopic appearances [107] (Fig. 69-9). JRSGC type I is an exophytic lesion, type II is flat (or superficial), and type III is an ulcerated or depressed lesion. Type II is further divided into three subtypes, depending on whether it is elevated (IIa), flat (IIb), or depressed (IIc). In addition, combined types have been defined to include complex configurations. The protruded-type (type I and type IIa), with a diameter of less than 25 mm, and the excavated type (type IIc), with a diameter of less than 20 mm, seldom produce lymph node metastasis. Moreover, the depth of tumor invasion also bears important prognostic significance. The incidence of nodal metastasis of intramucosal and submucosal cancer is reported to be 3% and 20%, respectively. [108] Other parameters that may determine the rate of lymph node metastases are the size of the tumors and the presence of ulcerations. Distant metastasis is uncommon in early gastric cancer.

FIGURE 69-9. Classification of early gastric cancer by the Japanese Gastroenterological Endoscopy Society. (From Japanese Research Society for Gastric Cancer. The general rules for the gastric cancer study in surgery. Jpn J Surg 1973;3:61.)

Symptoms and abnormal physical findings are unusual in early gastric cancer. Staging of early gastric cancer resembles that of non-early gastric cancer. Early gastric cancer is T1 carcinoma according to the TNM system. Treatment of early gastric cancer is dependent on the size, location, endoscopic appearance, and histological subtypes of the tumors, as discussed in the next section.

Many argue that early gastric cancer is not a genuine malignant condition, because of the diagnostic criteria for cancer adopted by Japanese pathologists. [109] Some are skeptical whether these so-called “early” cancers would eventually develop into advanced cancers. Tsukuma and colleagues reported their long-term follow-up of 56 patients with early gastric cancer diagnosed by endoscopy and confirmed by histology and in whom surgical resection was delayed or was not performed. [110] Over a period of 6 to 137 months, 36 (64%) patients progressed to the advanced stage. The cumulative risk of progressing to advanced cancer over 5 years was 63% in this series. Because of the inconsistency in the histological diagnosis of early gastric cancer, this result should be interpreted with caution. However, it appears that early gastric cancer progresses at a relatively slow pace.

CLINICAL FEATURES

Early gastric cancer typically produces no symptoms. Only patients with advanced disease will notice any discomfort. The most frequent symptoms and signs patients experience are weight loss and abdominal pain. [111] Persistent vomiting can occur in antral tumors obstructing the gastric outlet. Dysphagia may be present when tumors obstruct the gastroesophageal junction. Early satiety, although not a common presentation of gastric cancer, may suggest a diffusely infiltrative tumor resulting in loss of distensibility of the stomach. Gastrointestinal bleeding has been reported in about 10% to 15% of patients, but frank bleeding is rare. Overt massive upper gastrointestinal bleeding may be more common in gastric stromal tumors.

Physical signs are usually absent in patients with gastric carcinomas, except in metastatic disease. A palpable abdominal mass is rare before regional extension of the tumor. Gastric carcinomas spread by local extension to adjacent structures or metastasize by lymphatic, peritoneal, and distant routes. Tumor may penetrate through the gastric wall and may directly invade the omentum, pancreas, kidney, colon, spleen, and liver. Gastrocolic fistula is uncommon, but it may result in vomiting of fecolith material or passage of recently ingested food into stool. Lymphatic metastasis occurs early, and local lymph nodes are usually the first to be involved. Metastasis to the left supraclavicular lymph node produces the so-called *Virchow node.* Tumor spreading along the peritoneal surfaces may result in a periumbilical nodule (*Sister Mary Joseph node*), ovarian mass (*Krukenberg tumor*), or tumor mass in the cul-de-sac (*Blumer shelf*). Peritoneal seeding of tumor may produce malignant ascites. Hematogenous spread usually involves the liver and occasionally the lung, bone, or brain. Patients with gastric cancer occasionally present with paraneoplastic syndromes, such as acanthosis nigricans, membranous glomerulonephritis, microangiographic hemolytic anemia, arterial and venous thrombi (*Trousseau syndrome*), seborrheic dermatitis (*Leser-Trélat sign*), and dermatomyositis.

DIAGNOSIS

Because of the silent nature of the disease, timely diagnosis of gastric cancer is difficult. Conventionally, radiologic imaging played a major role in the initial diagnosis of gastric cancer. A barium meal was once the investigation of choice for all upper gastrointestinal symptoms. With the use of double-contrast techniques, better

visualization of mucosal detail can be obtained. However, false-negative rates of up to 25% have been reported in lesions between 5 and 10 mm in diameter. [112] Furthermore, it may not be possible to differentiate a benign from a malignant ulcer by barium studies. A malignant ulcer typically has an asymmetric ulcer crater eccentrically located in an irregular mass with distortion or obliteration of the normal mucosal fold surrounding the ulcer. Other features such as nodularity, clubbing, fusion, or amputation of radiating folds also suggested malignancy. The use of radiologic imaging has gradually been replaced by endoscopy.

Since its introduction in the 1960s, upper gastrointestinal endoscopy and biopsy have been the standards for the diagnosis of gastric cancer. However, the endoscopic appearance alone is insufficient for the diagnosis of stomach cancer (Fig. 69-10; see also Color Fig. 69-10). Diagnostic accuracy of 95% was reported, [112], [113] and [114] and this can be further improved by increasing the number of biopsies. [115] Biopsy should be taken from the edge and the base of a malignant ulcer. [116] The additional use of brush cytology may further increase the sensitivity of endoscopic diagnosis. The identification of two of the following cytologic features makes the diagnosis of gastric cancer more likely: single atypical cells with intact cytoplasm and with eccentric and atypical naked nuclei. [117] The diagnosis of early gastric cancer can be difficult because lesions may not be conspicuous. Endeavors to improve the diagnostic accuracy of endoscopy and to locate the proper site for the biopsy sample have led to the development of new technologies. Light-induced fluorescence endoscopy is a technique that detects the autofluorescence from tissues after irradiation with blue or violet light. Characteristically, gastric tumors emit a different fluorescence from that of normal tissues, and this can be used as an adjunctive technique for the evaluation of presumptive neoplastic tissue. [118], [119]

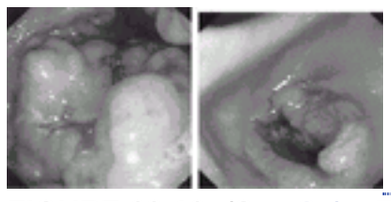

FIGURE 69-10. (See Color Fig. 69-10.) Endoscopic appearance of gastric cancer.

Screening of Gastric Cancer

Screening of gastric cancer in asymptomatic persons may increase the chance of detecting early cancer and hence may improve overall survival. In Japan, a screening program of gastric cancer used double-contrast radiology since the 1960s. [120] Seven-film photofluorography is the standard, which is then followed by endoscopic biopsies in suspicious cases. It was estimated that 40% to 60% of cancers diagnosed by the screening program were early cancer, with no lymphatic metastasis. [121] This program has been attributed as one of the reasons for the decline in stomach cancer mortality in Japan. However, a formal randomized study to evaluate the effectiveness of this screening strategy is lacking. Given the low incidence of gastric carcinoma in many developed countries, it is unlikely that a similar population-based screening program can be adopted. In areas with a low incidence of gastric cancer, screening may have to be targeted to high-risk persons only, such as those with family history of gastric cancer, chronic atrophic gastritis, previous gastric surgery, or pernicious anemia.

Apart from contrast study and endoscopy, other biochemical markers have been developed to identify those at risk of developing gastric carcinoma. Low serum pepsinogen I, which correlates with corpus atrophy, has been found to be significantly associated with gastric cancer. [122] In an evaluation, Kitahara and colleagues [123] showed that a pepsinogen I concentration of less than 70 ng/mL and a ratio of pepsinogen I to pepsinogen II of less than 3 are useful indicators, with a sensitivity of 84.6% and a specificity of 73.5% for detecting gastric cancer. In this context, an elevated serum gastrin level has also been used in the screening of severe atrophic gastritis. [124] Although an elevated gastrin level higher than 200 ng/L appears to be specific, the sensitivity is low (30%), thus precluding its use as a screening tool. The other potential serologic marker that may be used in screening of gastric cancer is *H pylori* antibody testing. However, because of the high prevalence of *H pylori* infection in most countries, a more discriminatory serologic test is necessary to identify which infected subgroups are at risk of developing gastric cancer. Antibody against CagA is the most extensively studied marker of *H pylori* infection for the diagnosis of gastric cancer. CagA is an immunodominant protein of *H pylori,* which is associated with the *cag* A gene or the cag pathogenicity island. Initial studies in countries with low *H pylori* prevalence suggested the link between gastric cancer and CagA antibody response. [125], [126] However, a similar association cannot be demonstrated in high-risk regions such as Japan and Colombia, probably because most *H pylori* strains circulating in these countries are CagA $^{+}$. [127] One study showed that no screening test (including *H pylori* and CagA serology, pepsinogen, and gastrin) was sensitive and specific enough to be used for the screening of chronic atrophic gastritis in populations with a high gastric cancer incidence. [128]

STAGING

The clinical and pathological staging of gastric carcinoma is of paramount importance in the management of the patient. The depth of the tumor invasion (T) (Fig. 69-11), the involvement of lymph nodes (N), and the presence of metastasis to other organs (M) are the major considerations. This TNM staging system has been extensively used in the evaluation of gastric cancer. However, there is considerable disparity between the Western and Japanese systems in the staging of gastric cancer.

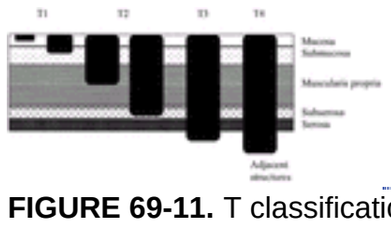

FIGURE 69-11. T classification of gastric carcinoma (TNM staging system).

In 1997, the TNM staging by the International Union against Cancer (UICC) and the American Joint Committee on Cancer (AJCC) underwent a substantial revision of the lymph node staging of gastric cancer. [129] The previous version of TNM staging had been based on the anatomic site or the distance of metastatic lymph nodes from the primary tumor, as in the Japanese classification system. [130] In the fifth edition of the TNM staging system, lymph node staging is determined by the number of regional lymph nodes involved (Table 69-4), which will ordinarily include sampling of 15 or more lymph nodes, instead of the sites of metastatic lymph nodes. If fewer than 15 lymph nodes are sampled, the N staging is defined as Nx. Regional lymph nodes are the perigastric nodes along the lesser and greater curvatures, the nodes located along the left gastric, common hepatic, splenic, and celiac arteries, and the hepatoduodenal nodes. Involvement of other intra-abdominal lymph nodes such as retropancreatic, mesenteric, and paraaortic are now classified as distant metastasis (M1). This change is supported by the observation that the absolute number of positive lymph nodes is of equal or even greater importance than the location of the lymph node. [131], [132] and [133] This new classification is easier to follow than the sophisticated localization of lymph nodes around the stomach and reduces the problems involved in the lymph node retrieval process. In this context, better reproducibility and standardization of histopathological examination can be achieved. Reports from various groups favor the use of this lymph node staging method over the previous version in determining the prognosis of patients. [134], [135], [136], [137] and [138] However, the possibility of unclassified cases (Nx) will increase by using this new system because of the inadequate number of lymph nodes sampled. In a Japanese study, 13% of patients were unclassified because fewer than 15 lymph nodes were examined. [137] This may be an even more serious problem in Western countries, where surgeons do not routinely perform extended dissection. If inadequate nodal dissection is performed, comparison of treatment results among different institutions will be difficult.

T: Primary Tumor	
Tis	Carcinoma in situ; intraepithelial tumor without invasion of lamina propria
T1	Tumor invades lamina propria or submucosa
T2	Tumor invades muscularis propria or subserosa
T3	Tumor penetrates serosa (visceral peritoneum) without invasion of adjacent structures
T4	Tumor invades adjacent structures
N: Regional Lymph Node	
N0	No regional lymph node metastasis
N1	Metastasis in 1 to 6 regional lymph nodes
N2	Metastasis in 7 to 15 lymph nodes
N3	Metastasis in more than 15 regional lymph nodes
M: Distant Metastasis	
M0	No distant metastasis
M1	Distant metastasis

II. STAGE GROUPING

Stage	T	N	M
0	Tis	N0	M0
IA	T1	N0	M0
IB	T1	N1	M0
	T2	N0	M0
II	T1	N2	M0
	T2	N1	M0
	T3	N0	M0
IIA	T2	N2	M0
	T3	N1	M0
	T4	N0	M0
IIB	T3	N2	M0
IV	T4	N1–3	M0
	T1–3	N3	M0
	Any T	Any N	M1

From ref. 129.

TABLE 69-4 Fifth Edition of the International Union Against Cancer (UICC) and American Joint Committee on Cancer (AJCC) TNM Classification of Gastric Cancer

Conversely, the JRSGC uses a TNM classification that is based on meticulous surgical and pathological findings. [107] Whereas the tumor staging resembles that of the UICC TNM system, lymph node metastases are divided into stations that fall into four N categories (Fig. 69-12). Additionally, the Japanese system has a P staging, which represents the presence of peritoneal metastases. Grouping of tumors into different stage also varies from the UICC system.

FIGURE 69-12. Regional lymph node stations as proposed by the Japanese Research Society for Gastric Cancer. (From Hermanek P, Huttter RVP, Sobin LH, et al, eds. TNM atlas, 4th ed. Berlin: Springer, 1997:82.)

Preoperative Staging

Traditionally, staging of gastric cancer is performed at the time of surgery. However, this approach includes many patients with unresectable tumors who may not benefit from surgery. In general, no single imaging technique is reliable as a sole diagnostic tool in the preoperative staging of gastric cancer. Multiple test modalities are required for accurate preoperative staging.

Endoscopy is widely used in the initial diagnosis of gastric cancer and has generally replaced contrast radiologic studies in many centers. However, its role in the staging of disease is not widely recognized, except in Japan, where emphasis has been put on macroscopic appearance of cancers. This approach may be more useful in the selection of early gastric cancer that is suitable for endoscopic resection.

Despite the widespread use of computed tomography (CT) scanning in the preoperative staging, its value remains disputable. Early studies in the 1980s reported encouraging results of CT imaging, [139] but these findings were not substantiated subsequently. [140] One of the limitations of CT is the inability to differentiate between different layers of the gastric wall and hence the inability to define the T stage. The role of CT is confined to its ability to demonstrate invasion outside the stomach wall and the presence of distant metastasis. However, the sensitivity of CT is generally considered to be low. Even with the use of spiral CT, the sensitivity of detecting tumor invasion into colon or mesocolon and pancreas was 76% and 50%, respectively. [141] Pancreatic invasion is extremely difficult to detect on CT because inflammation and neoplastic infiltration is difficult to differentiate. Moreover, although the presence of a clear fat plane between the stomach and the pancreas indicates noninvasion, the absence of such a tissue plane does not necessarily imply tumor invasion. For lymphatic metastasis, the sensitivity of CT varies widely, from 48% to 91%. [142], [143], [144], [145], [146] and [147] Nonetheless, CT appears to be quite specific (>90%) for the detection of lymph node metastasis. This in part may be related to the conventional cutoff size (10 mm) of the malignant lymph node used by most studies. By reducing the cutoff size to less than 10 mm, sensitivity will increase, but there is a compromise in specificity. Furthermore, the presence of a conglomeration of enlarged lymph nodes is more likely to be malignant than is a solitary enlarged lymph node. Although the two most common sites of metastasis of gastric cancer are the liver and the peritoneum, most liver metastases are small and peripheral and hence have a low rate of detection by CT. Sensitivity in detection of distant metastasis was reported as 57%, [141] but false-positive results were not uncommon. Finally, it is generally recognized that CT is poor in visualization of small peritoneal nodules, except in the presence of ascites. [148] Overall, CT tends to understage gastric cancer. It is more useful in identifying advanced-stage disease that may not be suitable for primary curative resection.

The role of magnetic resonance imaging (MRI) in the preoperative staging of gastric cancer is yet to be defined. MRI requires a longer scan time than CT and thus is easily subjected to artifact from respiration and bowel movement. The other limitation of MRI is the lack of a widely accepted oral contrast medium to distend the stomach. Nevertheless, MRI is superior to CT in delineating the layered structure of the stomach wall, and it may be used in predicting the depth of tumor invasion (T staging). In a small study involving 30 patients with gastric cancer, MRI was marginally superior to helical CT (73% versus 67%). [149] For N staging, the accuracy of MRI and helical CT is comparable. Little information is provided regarding the role of MRI in evaluating distant metastasis.

Whole-body [^{18}F]fluorodeoxyglucose (FDG) positron emission tomography (PET) is being increasingly used in the evaluation of gastrointestinal malignant disease. The preferential accumulation of positron-emitting FDG by tumor cells has been used in the detection of human cancers. Data on staging of gastric carcinoma are limited. Preliminary study showed that although PET appears to be sensitive in the detection of gastric tumors, it is of limited value in locoregional staging because of poor differentiation of primary tumor from metastatic lymph nodes. [150] Conversely, it may be more useful in the follow-up of treatment response. Changes in tumor FDG uptake were seen in gastric carcinomas after chemotherapy. [151]

Another imaging modality that has been gaining popularity in the preoperative staging is endoscopic ultrasonography (EUS). With the use of medium- to high-frequency ultrasound probes (7.5 to 30 MHz), the gastrointestinal tract is clearly delineated as a five-layered structure by EUS corresponding to the mucosa, submucosa, muscularis propria, and serosa of the gastric wall. EUS provides an accurate assessment of T staging and is superior to other imaging modalities for the local staging of gastric cancer (Fig. 69-13). The overall accuracy of EUS in T staging was estimated to be 78%. [152] The accuracy tends to be lower for T2 tumors because of the tendency to overstage these tumors as a result of poor differentiation between inflammation and infiltration. The overall accuracy of N staging was estimated to be 70%, [152] which is also superior to CT. It is more accurate for N0 status (85%) but least for N2 status (65%). EUS is generally unsuitable for the assessment of distant metastases because of the low penetration of high-frequency ultrasound. There are several caveats relating to the use of EUS in gastric cancer staging. The accuracy of EUS is highly operator dependent. Differentiation between malignant infiltration and fibrosis at the base of peptic ulcer can be difficult. Obstructing tumors, usually at the cardia, that prevent passage of echoendoscope into the stomach may limit its use. Moreover, the presence of metastases within a normal-size lymph node may be missed.

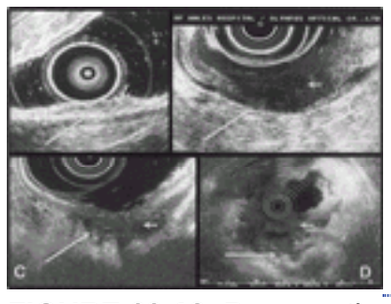

FIGURE 69-13. Preoperative staging of gastric carcinoma by endoscopic ultrasound. (**A**) Intramucosal early gastric cancer (T1is) with focal mucosal thickening (*short arrow*); the lamina propria (*long arrow*) is intact. (**B**) T2 gastric cancer (*short arrow*) invades into the muscularis layer, and the serosa (*long arrow*) is intact. (**C**) Small gastric cancer (*short arrow*) invades through the serosa (T3). (**D**) Large T4 gastric cancer (*short arrow*) invades into the neck of pancreas (*long arrow*). (Courtesy of Dr. Y.T. Lee, Prince of Wales Hospital, Hong Kong.)

Preoperative laparoscopy often detects unexpected peritoneal or liver metastasis that may not be identified by other imaging techniques (Fig. 69-14; see also Color Fig. 69-14). The role of laparoscopy in preoperative staging is difficult to evaluate because most published series involved heterogeneous patient populations and gave inconsistent results. The overall rate of detecting peritoneal and liver metastases by laparoscopy ranged from 3% to 37% and 2% to 25%, respectively. [153] Based on these findings, a corresponding treatment plan was altered in 6% to 42% of patients. The use of laparoscopic ultrasound has been advocated to enhance the ability to detect liver metastases. The use of laparoscopy may be particularly useful in a subgroup of patients with T4 disease who may benefit from multimodality treatment. [154]

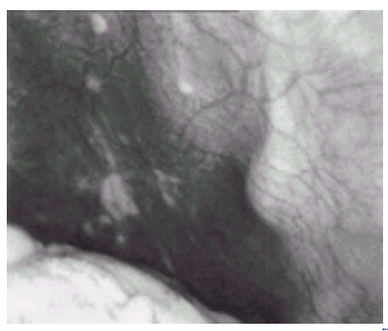

FIGURE 69-14. (See Color Fig. 69-14.) Laparoscopic view of a patient with gastric carcinoma with peritoneal metastasis (whitish nodule).

TREATMENT

To date, surgery remains the mainstay of treatment for gastric cancer. Unfortunately, two thirds of patients present with advanced disease and are thus incurable by surgical excision only. Even with the use of aforementioned advance staging techniques, 10% to 20% of patients who undergo surgical exploration for potentially curative resection are found to have peritoneal seeding at the time of laparotomy.

Surgery

Certain definitions need to be clarified before discussion of treatment options. Curative resection is defined according to the UICC/AJCC combined rules. [129] R0 resection indicates potentially curative removal of the tumor with no residual disease. R1 refers to resection that leaves behind microscopic tumors, whereas R2 resection indicates that gross residual tumors remain (i.e., palliative resection). Conversely, surgical resection is classified by the extent of lymph node dissection, as designated by D. D0 denotes no or incomplete dissection of the N1 nodes. D1 refers to dissection of tumor and adjacent nodes (N1) only, whereas D2 indicates dissection of nodes along the major celiac vessels (N1 and N2 nodes). D3 dissection removes the entire tumor mass together with N1, N2, and N3 lymph nodes.

Although few argue that surgery is the best treatment option, the extent of resection, particularly the extent of lymph node dissection, remains highly controversial. In practice, the extent of dissection is determined primarily by tumor location and staging and the premorbid conditions of the patient. Extensive resection has the benefit of removing the major tumor bulk, at the cost of higher risks of operation and postoperative complications. Even though total gastrectomy offers a larger surgical resection margin, studies failed to demonstrate any advantage of total gastrectomy over subtotal gastrectomy. [155], [156] The French multicenter study showed similar postoperative morbidity and mortality rates and 5-year survival rates for total and subtotal gastrectomy. The Hong Kong study, which compared subtotal D1 gastrectomy with total D3 gastrectomy, also showed a significant increase in morbidity from splenectomy and distal pancreatectomy in the latter group. The survival rate was actually better in the group with limited resection only. Conversely, total gastrectomy may be indicated for tumors involving the proximal stomach, particularly carcinoma of the gastric cardia, [157] because of the functional defect of proximal gastrectomy leading to reflux esophagitis. Yet, data suggested that total gastrectomy may not have any influence on the morbidity and survival of patients with proximal gastric carcinoma. [158]

There is considerable difference between Western and Japanese surgeons in the extent of lymphadenectomy for gastric cancer. Japanese surgeons systematically perform a more extended lymph node dissection (D2) than Western surgeons, based on the belief that the primary tumor will disseminate through lymphatic channels in an orderly manner from the nearest to the farthest lymph nodes. This practice is considered to be the major reason for the contrasting outcomes between Western and Japanese patients with gastric cancer. However, prospective randomized controlled trials from Western countries comparing D1 with D2 lymphadenectomy failed to demonstrate any survival benefit of the more aggressive approach with D2 dissection. [159], [160] and [161] Notably, both the British Medical Research Council trial and the Dutch Gastric Cancer Group trial showed a significantly higher short-term morbidity and hospital mortality among patients undergoing D2 dissection without improving the 5-year survival (Table 69-5). The high initial complication rate of D2 gastrectomy has been attributed to pancreaticosplenectomy and the unfamiliarly of the surgeons with this kind of operation. Conversely, the perioperative mortality of D2 resection, particularly with the spleen and pancreas preserved, by surgeons experienced with this procedure, is reported to be about 1% to 3%. [162], [163] and [164] Another controversial issue relating to these two trials is the high rates of noncompliance with either D1 or D2 gastrectomy, which may interfere with the final outcome. Patients randomized to D1 dissection may receive a more aggressive dissection, and patients randomized to D2 dissection may receive inadequate lymph node dissection. Thus, the extent of lymphadenectomy remains controversial despite the publications of these randomized controlled trials, and further studies are needed. [165]

TABLE 69-5 Short-Term and Long-Term Results of Major Randomized Trials Comparing D1 and D2 Lymph Node Dissection in Gastric Carcinoma

Patients with locally advanced disease may benefit from a more aggressive resection, such as splenic resection with left-sided pancreatectomy in cases with splenic hilar lymph node involvement. This en bloc procedure can remove all metastatic lymph nodes along the splenic artery. As shown by the British Medical Research Council and Dutch trials, [159], [160] and [161] the removal of extragastric organs, whether directly involved or not, was associated with higher morbidity and hospital stay. Both splenectomy and distal pancreatectomy are therefore not routinely recommended in D2 resection except in the presence of direct tumor invasion. Conversely, Japanese surgeons may favor a more aggressive approach in patients with T4 tumors in the absence of distant metastasis, to reduce the chance of subsequent tumor bed recurrence.

Because most cases of gastric cancer are diagnosed at an advanced stage, fewer than 50% of patients with gastric cancer can undergo potentially curative gastric resections. Nonetheless, palliative surgery may still be considered in certain circumstances such as tumor obstruction, perforation, and bleeding. There is also a reduction in the incidence of debilitating ascites after palliative gastrectomy. [166] If gastrectomy is performed, gross disease immediately adjacent to the stomach should be removed. Leaving behind a primary tumor mass in the stomach not only invites bleeding and perforation but also diminishes the likelihood of response to subsequent palliative chemotherapy. Alternatively, a bypass procedure or gastrojejunostomy can be performed, but it has been found to be associated with significantly shorter palliation period when compared with gastric resection. [167] There is also no significant difference in terms of operation-related morbidity and mortality. However, gastric resection may not be indicated in patients with a poor chance of survival, and the final decision is usually dictated by the overall clinical condition of the patient.

Endoscopic Therapy

In addition to its role in diagnosis, endoscopy has been developed as a curative as well as palliative treatment for gastric cancer. *Endoscopic mucosal resection* (EMR) or strip biopsy was first described by Tada and colleagues. [168] It is widely practiced in Japan as the treatment of choice for early gastric cancer after EUS confirmation of disease confined to the mucosa. Because of the low incidence of lymph node and distant metastasis, surgical resection of the stomach for early gastric cancer is considered unnecessary, and a limited resection is therefore more appealing. Various techniques of EMR have been described. [169] and [170] The procedure involves injection of saline into the submucosa under the lesion to lift the mucosal and submucosal layer from the muscle, to avoid perforation. Methylene blue may be added to the saline to demarcate the plane of separation. The endoscope is fitted with suction cap at its tip. The lesion is sucked into the chamber and is resected by an EMR snare. Resection of the mucosa may then be achieved by using a cap-fit device at the tip of the endoscope and a mucosectomy snare. The malignant tissue can be removed en bloc or by piecemeal resection. Muscle fibers can be seen at the base of the resected area. Re-epithelialization usually occurs within weeks. In contrast to ablation by laser or argon plasma, EMR can provide the original resected specimens for histological evaluation. The major complications of EMR are bleeding and perforation, which were estimated to be about 5%. [171] The indications for EMR as suggested by the Japanese Gastroenterological Endoscopy Society are as follows: elevated-type intramucosal cancer less than 20 mm in size; depressed-type mucosal cancer without ulceration, less than 10 mm in size; and intestinal-type adenocarcinoma. In a review of the 11-year results of EMR for early gastric cancer at the National Cancer Center Hospital of Japan, in which 479 cancer patients were treated by EMR, 69% of tumors were resected with clear margin, and 17 local recurrences were detected during subsequent follow-up. [171] There were no gastric cancer–related deaths during the follow-up period (3 to 120 months). In cases with residual tumor after EMR, patients can be offered surgery or local therapy such as laser ablation. However, to date, there is no randomized controlled trial comparing the efficacy of EMR with conventional modalities of treatment. [172] An alternative method in EMR is to employ an insulated-tip needle-knife, as described by Ohkuwa. [173] Although this technique allows resection of a much larger piece of mucosa, it is associated with significantly higher risks of the complications of bleeding and perforation. [173] Further refinement of this technique is necessary.

For advanced inoperable gastric cancer with obstruction, laser or argon plasma coagulation therapy and endoscopic stenting procedures can be considered. Laser ablation of tumors requires expensive equipment, and thus argon plasma coagulation is preferred by most gastroenterologists. The advent of self-expanding metallic stents made possible the palliation of malignant stricture with minimal complications. It is particularly useful in the palliation of proximal gastric cancer as well as gastric outlet obstruction. In addition, fistulae and perforations can be sealed by the covered stent. Endoscopic stenting can also be used to relieve obstruction at anastomosis after partial or total gastrectomy as a result of tumor recurrence (Fig. 69-15; see also Color Fig. 96-15). Tumor ingrowth, stent migration, and gastroesophageal reflux are complications of endoscopic stenting. Various newer covered stents are available to overcome the problem of tumor ingrowth, and antireflux mechanisms have been devised to avoid gastroesophageal reflux.

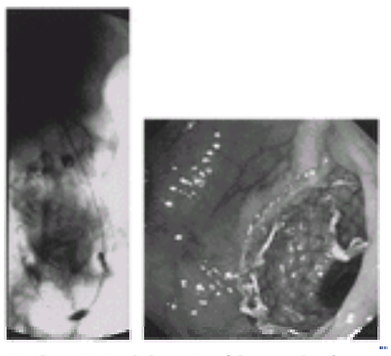

FIGURE 69-15. (See Color Fig. 69-15.) Self-expandable metal stent in patient with recurrent gastric carcinoma causing gastric outlet obstruction after subtotal gastrectomy.

Chemotherapy

Chemotherapy can be given preoperatively (neoadjuvant), intraoperatively, or postoperatively to reduce the chance of local or systemic tumor recurrence. Numerous chemotherapeutic agents, either given alone or in combination, have been used with variable success. The *response rate* refers to a 50% reduction of tumor mass. The activity of a single agent ranges from 5% to 31%. [174] 5-Fluorouracil (5FU) is the most frequently used agent, with a response ranging from 20% to 30%, depending on the route of administration. Mitomycin C, commonly used in Japan, has an overall objective response rate of 30%. Other agents with reported response rates of 19% to 26% include epirubicin, doxorubicin, cisplatin, and hydroxyurea.

Adjuvant therapy is generally considered as additional treatment for patients who have already undergone potentially curative surgical procedures (i.e., R0 resection). In patients who had gross residual disease remaining after surgery, treatment should be considered palliative therapy instead of adjuvant therapy. Numerous trials have been performed. Unfortunately, most of these have major methodological flaws or merely represent anecdotal reports. Two early studies conducted by the Veterans Administration Study Group with single-agent chemotherapy, using thiotepa or 5FU, after surgical resection failed to demonstrate any improvement in long-term survival. [175], [176] As a result of the poor response rate from single-drug chemotherapy, various combination therapies were devised. As yet, no consensus combinations are available for the treatment of gastric adenocarcinoma. Various combinations such as FAM (5FU, doxorubicin [Adriamycin], mitomycin C), FAMTX (5FU, doxorubicin [Adriamycin], methotrexate), FEP (5FU, etoposide, cisplatin), FAP (5FU, doxorubicin [Adriamycin], cisplatin), and ECF (epirubicin, cisplatin, infusional form of 5FU) have been used in various studies. [177] The median survival associated with multidrug chemotherapy ranges from 6 to 10 months, and the real survival benefit is debatable. Most of the published trials focused on response rate to chemotherapy without a formal evaluation of the patients' quality of life.

Overall, results from early trials were disappointing. In a review of adjuvant therapy for gastric cancer since the mid-1980s, investigators concluded that adjuvant therapy does not confer any survival benefits and thus cannot be recommended. [178] Nonetheless, the timing and mode of postoperative chemotherapy given in Western countries may undermine the effect of adjuvant chemotherapy. Whereas chemotherapy is usually given at 4 to 6 weeks after surgery, Japanese oncologists favor early institution of treatment during or immediately after the operation. Conversely, a multicenter study from Japan using adjuvant mitomycin and 5FU followed by oral uracil plus tegafur for 18 months in serosa-negative gastric cancer (T1 and T2) after curative gastrectomy failed to show any difference in survival between treatment and control groups. [179] The 5-year survival was 82.9% in the control group and 85.8% in the treatment group. Thus, it appears that, even in Japan, this group of patients with early-stage disease may not benefit from adjuvant treatment.

The rationale for neoadjuvant (preoperative) chemotherapy is to reduce tumor burdens before surgery with the hope of achieving a higher rate of R0 resection and better patient survival. Furthermore, treatment is targeted to minimize distant micrometastasis before operation and to limit potential cancer cell dissemination during the surgical procedure. Numerous trials have been conducted, but the only conclusion that can be drawn is that neoadjuvant chemotherapy does not increase postoperative morbidity and mortality. [180] In one small series, response to neoadjuvant chemotherapy was found to be the single most important predictor of overall survival. [181]

In addition to the administration of chemotherapeutic agents by the systemic route, antitumor drugs can be delivered intraperitoneally. The potential benefit of intraperitoneal chemotherapy is based on the ability to control locoregional tumor recurrence after curative resection. Hagiwara and colleagues [182] evaluated the use

of intraperitoneal mitomycin C adsorbed to activated charcoal in high-risk patients after curative resection. There was a highly significant difference in 2-year survival (68.6% versus 26.9%) favoring the use of this regimen. A prospective randomized trial of early postoperative intraperitoneal chemotherapy as adjuvant therapy in resectable gastric cancer also showed significant improvement in 5-year survival rate in a subset of patients with stage III disease. [183] The effects of intraperitoneal chemotherapy remain to be substantiated by further studies.

Radiotherapy

Gastric carcinoma is relatively resistant to radiotherapy. Radiotherapy includes both external beam and intraoperative radiotherapy. The British Stomach Cancer Group [184] found that postoperative radiotherapy in addition to surgery did not confer any benefit in survival. The 5-year survival in patients receiving surgery plus radiotherapy and surgery alone was 12% and 20%, respectively. Conversely, intraoperative radiotherapy is frequently given in Japan. [185] Although it appeared to prolong survival for patients with stage II and III disease, this therapy needs further evaluation.

PROGNOSIS

The mortality of gastric cancer in many parts of the world, with the exception of Japan, has not been changed over the past few decades. A review from the National Cancer Data Base revealed that the overall 5-year and 10-year survival rates of patients diagnosed with gastric cancer between 1985 and 1996 were 28% and 20%, respectively [186] (Fig. 69-16). The stage-stratified survival analysis reviews that men had a poorer prognosis than women, and proximal cancer was associated with worse prognosis than distal cancer. Difficulty in attaining an adequate proximal resection margin and the close proximity to paraaortic lymph nodes around the hiatus may be the reasons contributing to the worse outcome in proximal gastric tumors.

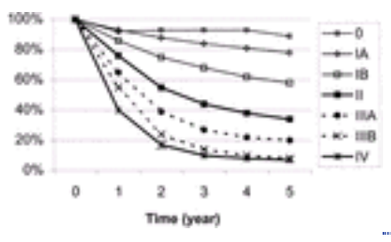

FIGURE 69-16. Five-year survival rates of gastric cancer patients treated by gastrectomy in the United States. (Modified from ref. [186].)

In contrast to the grave prognosis of gastric cancer in many developed countries, 50% of patients with gastric cancer can be cured of the disease in Japan. Several explanations have been put forward to account for this discrepancy. First, the adoption of screening policy for gastric cancer in Japan may lead to detection of early cancer and hence a higher resectability rate. Second, when compared with Western populations where proximal gastric cancers are prevalent, the Japanese population has predominantly distal gastric cancer. [186] Other factors that may contribute to this difference in prognosis include the more precise staging of the disease, the more aggressive surgical approach, and perhaps the use of adjuvant therapy by Japanese clinicians. [164]

GASTRIC TUMORS OTHER THAN ADENOCARCINOMAS

Gastric Lymphoma

Although *primary gastric lymphoma* is a rare malignant disease that accounts for less than 5% of all gastric malignancies, it is the most common form of gastrointestinal lymphoma. It is estimated that approximately 70% of primary extranodal lymphomas of the gastrointestinal tract arise from the stomach. [187] However, the diagnosis of primary gastrointestinal lymphoma is controversial. The definition proposed by Dawson and colleagues [188] requires that primary gastrointestinal lymphoma be limited to the stomach or intestine and its contiguous lymph nodes. This criterion has been criticized as being too restrictive. A more practical definition has been adopted that states that a tumor presenting with the bulk of disease in the gastrointestinal tract can be considered “primary” gastrointestinal lymphoma.

Most (80%) of gastric lymphomas are B cell in origin. The classification of gastric lymphoma, like that of many extranodal lymphomas, is confusing. Several systems have been proposed by various groups, which include the Rappaport classification, the Lukes-Collins classification, the Kiel classification, and the Working formulation (Table 69-6). Direct comparisons of past studies are difficult. Data suggest that the true incidence of primary gastric lymphoma is rising, particularly among elderly persons. [189], [190] This may in part be explained by improved diagnosis and by the increasing incidence of lymphoma in immunosuppressed patients such as in patients with acquired immune deficiency syndrome or those taking immunosuppressive agents. The origin of this disease remains elusive, but many of these tumors, particularly mucosa-associated lymphoid tissue (MALT) lymphoma, are related to chronic inflammation attributed to *H pylori* infection. [191] *H heilmannii* has also been linked to MALT lymphoma. [192]

B-cell	MALT-type
	Low-grade
	High-grade with or without low-grade component
	Mantle cell
	Burkitt
	Immunodeficiency-related
	Posttransplant
	Acquired immunodeficiency syndrome
	Congenital
	Others
T-cell	Enteropathy-associated
	Others

MALT, mucosa-associated lymphoid tissue.

TABLE 69-6 Primary Gastrointestinal Non-Hodgkin Lymphoma

Macroscopically, it may not be possible to differentiate gastric lymphoma from carcinoma. Gastric lymphoma can be polypoid, fungating, infiltrating, or even ulcerative in appearance. Most lymphoma spreads by submucosal infiltration, and the muscular layer is usually spared until a very late stage of the disease. Further spread to local and regional lymph nodes then follows. Microscopically, there is considerable heterogeneity among different classification systems.

Symptoms of gastric lymphoma are nonspecific; the most frequent symptoms are abdominal pain, weight loss, nausea, vomiting, and anorexia. Bleeding occurs in about 20% of cases. [193] Although upper gastrointestinal contrast studies are not the investigations of choice, certain radiologic features may suggest the presence of gastric lymphoma. These include diffuse mucosal hypertrophy with irregular thickening of folds, single or multiple ulcers associated with diffuse mucosal thickening, [194] and lesions larger than 15 cm. [190] Upper gastrointestinal endoscopy is widely used in the diagnosis of gastric lymphoma that can provide histological proof. However, multiple and deep gastric biopsies are usually required for histological diagnosis because tumors may develop in the submucosal lymphoid tissue covered by normal gastric mucosa.

The Musshoff-modified Ann Arbor staging system is widely used for staging of gastric lymphoma (Table 69-7). [195] This staging system was subsequently refined, with an emphasis on separate description of local penetration into neighboring structures. Stage IE was divided into IE_1 (infiltration of the mucosa and submucosa) and IE_2 (infiltration of the muscularis or (sub)serosa). Staging of disease can be done by CT scan and EUS. EUS is reliable in delineating the depth of tumor involvement and is superior to CT in distinguishing stage IE from stage IIE_1 disease. [196] The availability of these imaging techniques has reduced the prior reliance on surgery for staging of gastric lymphoma. To differentiate primary gastric lymphoma from systemic lymphoma with gastric involvement, other investigations such as peripheral

blood smear, bone marrow biopsy, and radiologic imaging are required.

STAGE	DESCRIPTION
IE	Localized involvement of one or more gastrointestinal sites on the same side of the diaphragm without lymph node involvement
IE_1	Confined to the mucosa and submucosa
IE_2	Extending beyond submucosa
IIE	Stage IE with lymph node involvement; penetration of lymphoma through the gut wall
IIE_1	Regional lymph node involvement
IIE_2	Lymph node infiltration beyond the regional wall
IIIE	Involvement of the gastrointestinal tract and/or lymph nodes on both sides of the diaphram
IVE	Diffuse or disseminated involvement of nongastrointestinal organs or tissues

Adapted from ref. 195.

TABLE 69-7 Musshoff's Modification of the Ann Arbor Staging System for Gastric Lymphoma

P>Until recently, gastrectomy was the only treatment available for gastric lymphoma. However, there is a current tendency toward stomach-conserving therapy, and the best treatment for primary gastric lymphoma has yet to be defined. Many of these controversies originate from the differences in classification systems, the small numbers of patients treated with a variety of protocols, and the lack of randomized controlled trials. Surgery is favored by some clinicians because it is potentially curative and can remove all tumor tissues for accurate typing and staging. Moreover, resection can prevent complications such as hemorrhage and perforation during radiotherapy and chemotherapy. It is particularly useful for localized diseases, such as stages IE and IIE, giving a favorable 5-year survival rate of 85% to 100%. [197], [198] and [199] However, the operative mortality rate ranges from 0% to 30%, and there is no consensus on the extent of surgical dissection. Chemotherapy or combination treatment may be more appropriate for patients with advanced-stage (stage IIIE and IVE) diseases. Traditionally, the combination of cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) is the standard regimen used in the treatment of gastric lymphoma. Radiotherapy, either used as primary treatment or as adjuvant therapy, has also been reported. For stage IE and IIE disease, comparable 5-year survival rates have been reported in studies comparing primary radiotherapy with surgery. [200] Although lymphoma is usually sensitive to radiation, there is always the concern of complications related to this treatment option, such as renal toxicity, perforation, and hemorrhage, although studies have shown that these complications are uncommon. [201] Based on experience in the treatment of nodal lymphoma, many investigators favor the use of stomach-conserving treatment, that is, primary systemic chemotherapy or radiotherapy. In the interim analysis of the prospective German multicenter study, the operative approach does not appear to be superior to the conservative management of primary gastric lymphoma. [202]

The grading and staging of gastric lymphoma are the two decisive prognostic factors. In a review by the German-Austrian Gastrointestinal Lymphoma Study Group, [199] the overall 2-year survival rates for low-grade lymphoma ranged from 89% to 96%. Conversely, patients with high-grade lymphoma fare much worse. When surgery was performed, patients with complete resection had significantly better survival than those with incomplete resection (2-year survival of 83% to 88% versus 53%).

Mucosa-Associated Lymphoid Tissue Lymphoma *MALT lymphoma* was first described in 1983 by Issacson and Wright [203] as a distinct pathological entity. They described that certain low-grade B-cell gastrointestinal lymphomas recapitulated the features of Peyer patches or MALT. MALT lymphoma is characterized by the presence of plasma cells, reactive follicles, and centrocyte-like cells that tend to invade mucosal epithelium and form characteristic lymphoepithelial lesions. Most are of B-cell origin, and the most frequent site of involvement is the stomach. These tumors tend to arise as polyclonal proliferation in chronic inflammatory tissues, either autoimmune or infectious. MALT lymphoma was the most common type of the marginal zone lymphomas according to the Revised European-American lymphoma (REAL) classification. [204] In the scheme proposed by Issacson and Wright, [205], [206] B-cell gastric lymphoma, which accounts for most of the gastric lymphoma, can be subdivided into low-grade and high grade MALT lymphomas. Low-grade MALT lymphoma is represented by small lymphocytic cells disrupting the gastric glands and forming characteristic lymphoepithelial lesions (Fig. 69-17; see also Color Fig. 69-17). Tumor cells infiltrate around and between reactive lymphoid follicles. In high-grade lymphoma, one sees confluent clusters or sheets of larger and transformed blastic cells that are usually thought to be centroblasts outside the colonized follicles. Notably, foci of high-grade components may be present in low-grade MALT lymphoma. [207]

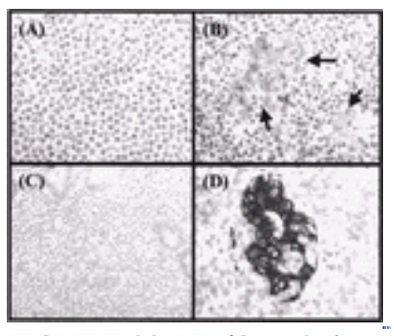

FIGURE 69-17. (See Color Fig. 69-17.) Gastric mucosa-associated lymphoid tissue (MALT) lymphoma: characteristics features of low-grade MALT lymphoma. (**A**) Centrocyte-like cells (hematoxylin and eosin, ×200). (**B**) Lymphoepithelial lesions (marked by *arrows*) (hematoxylin and eosin, ×200). (**C**) Plasmacytic differentiation (hematoxylin and eosin, ×100). (**D**) Lymphoepithelial lesions (immunostain for cytokeratin, ×200). (Courtesy of Dr. Wing Y. Chan, Chinese University of Hong Kong, Prince of Wales Hospital, Hong Kong.)

The endoscopic appearance of MALT lymphomas ranges from normal mucosa to erosions, ulcers, or protruding masses. These histological diagnoses of MALT lymphoma may sometimes be difficult because of the existence of a wide range of variations in the assessment of intermediate categories. The diagnostic yield for this tumor can be improved by the use of other techniques, such as polymerase chain reaction and immunohistochemistry. The detection of monoclonal rearrangements of the immunoglobulin heavy chain gene by polymerase chain reaction or Southern blotting suggests the presence of lymphoma. The immunophenotypes of low-grade MALT lymphoma overlap those of marginal B cells. Both demonstrate the presence of pan B-cell antigens (CD19, CD20, and CD79a) and the lack of CD5, CD10, CD23, and cyclin D_1 expression. [208] Conversely, the use of immunostaining for cytokeratin enhances the visibility of lymphoepithelial lesions, an important diagnostic criterion of gastric MALT lymphomas. Compelling epidemiologic and pathological evidence suggests the link between chronic *H pylori* infection and gastric MALT lymphoma. [191], [209] This disease is more prevalent in regions with a high prevalence of *H pylori* infection, particularly in northeastern Italy. [210] More than 90% of low-grade gastric MALT lymphomas showed evidence of *H pylori* infection. [209] In addition, clinical studies strongly support the potential for curing MALT lymphoma by eradicating *H pylori* infection. [211], [212], [213], [214] and [215] Cure of the infection results in apparent disappearance of the tumor and eventual decrease or disappearance of the monoclonal B-cell populations from the gastric mucosa. However, the time to complete tumor remission varies considerably and may not be apparent until several months after clearance of the organism. [215] Monoclonal B cells, as detected by polymerase chain reaction, may persist up to several years after cure of *H pylori* infection and complete histological and endoscopic remission. [216] The overall success of antibiotics in achieving complete remission ranges from 50% to 89%. [199], [215], [217] Staging by EUS may be able to predict the response of gastric MALT lymphoma to anti- *Helicobacter* treatment. Patients with stage IE_1 disease (confined to mucosa and submucosa) are more likely to have complete regression of MALT lymphoma after anti- *Helicobacter* therapy. [217], [218] Other factors that may identify nonresponders to antibacterial therapy include *H pylori*–negative status and the presence of high-grade features. Results from cytogenetic studies show that the presence of translocation of (11;18, q21;q21), which results in a chimeric transcript between the *API2* and *MLT* genes, may predict lymphoma resistance to antibiotic treatment. [219] When antibiotic treatment fails or in the presence of high-grade features or the absence of *H pylori* infection, other treatment modalities such as surgery, chemotherapy, and radiotherapy should be considered.

Gastrointestinal Stromal Cell Tumors

Gastrointestinal stromal tumors (GISTs) encompass a heterogenous group of mesenchymal tumors. There has been considerable controversy regarding the nomenclature, origin, cellular differentiation, and prognosis of this disease. Based on light microscopic appearance of primarily spindle-shaped cells, most gastrointestinal mesenchymal tumors were initially thought to be of smooth muscle origin. However, with the advent of immunohistochemistry and electron microscopy, it became apparent that there is considerable variability within these tumors, and further categorization is possible. GISTs can have predominantly myogenic, neural, or mixed features. The understanding of GISTs has evolved to account for most mesenchymal tumors arising within the gastrointestinal tract with an incomplete myogenic and neural or "uncommitted" phenotype. By contrast, gastric stromal tumors with predominant smooth muscle differentiation are termed *gastric smooth muscle tumors.* They were conventionally called leiomyomas and leiomyosarcomas. Gastric mesenchymal tumors of neural origin can be divided into four

main groups depending on the origin: peripheral nerve (schwannoma, neurofibroma, and neuroma); sympathetic or chromaffin system (neuroblastoma, ganglioneuroma, and paraganglioma); gastrointestinal plexus (gastrointestinal autonomic nerve tumor); and undetermined.

The precise cellular origin of GISTs has been proposed to be the interstitial cell of Cajal, an intestinal pacemaker cell. [220], [221] This is supported by the sharing of morphologic, immunohistochemical, and ultrastructural features between GISTs and the interstitial cell of Cajal. The interstitial cell of Cajal demonstrates an incomplete myogenic and neural differentiation on ultrastructural examination and exhibits features that can be identified in GISTs. In particular, immunoreactivity against CD34 and CD117, although not specific, are frequently detected in GISTs and interstitial cells of Cajal. CD34, a transmembrane glycoprotein, is expressed in 50% to 80% of GISTs. [222] However, it is also expressed in true smooth muscle cells. Conversely, CD117/c- *kit* is expressed in 80% to 100% of GISTs and may be more sensitive and specific than CD34. [223] The c- *kit* protooncogene encodes a type III tyrosine kinase growth factor receptor, which can be detected by immunohistochemistry for CD117. Mutations of c- *kit* that cause constitutive activation of the tyrosine kinase function of c- *kit* are detectable in most GISTs and appear to play a pivotal role in the pathogenesis of these tumors. [224] For smooth muscle tumor, immunoreactivity against desmin is frequently (50% to 70%) positive, and CD117 is usually negative.

GIST is an uncommon disease. The Finnish Cancer Registry [222] estimated an annual incidence of 4 cases per million, which may be an underestimation. In a review performed by the Memorial Sloan-Kettering Cancer Center, [225] there were only 200 patients with GIST treated during the previous 16 years. The most frequent site affected is the stomach, which accounts for about 70% of the cases. About 20% to 30% of GISTs are found in the small intestine, and fewer than 10% are found elsewhere in the gastrointestinal tract. [226] The peak incidence is in adults during the fifth and sixth decades, and the disease is uncommon in patients less than 40 years old. Both genders are affected. Etiologic agents have not been identified.

Histologically, the distinction between benign and malignant tumors is difficult, and there is no unequivocal criterion to differentiate the two. Conventionally, various parameters have been used to predict the biologic behavior including mitotic activity (>10/high-power filed), tumor size (>5cm), tumor necrosis, histological type or pattern, immunohistochemical profile, staining for proliferation antigens, and ploidy status. [222] Among these, mitotic activity and tumor size appear to be more reliable in categorizing tumors as benign, borderline, and malignant, but there is no agreed cutoff point for these values. The use of EUS may also help in differentiating benign from malignant tumors. Tumors less than 30 mm in diameter with regular margins and with a homogenous echo pattern are usually benign. [227]

Many of these tumors are asymptomatic and are discovered incidentally, but the clinical courses are highly variable. Patients can present with nausea, vomiting, abdominal pain, gastrointestinal bleeding, and even metastatic diseases. Bleeding is considered the most common presentation. Malignant tumors can metastasize to the liver and peritoneum and, rarely, to lymph nodes, bone, and lung. [225] Accurate preoperative diagnosis can be difficult, and percutaneous biopsy carries a theoretical risk of peritoneal seeding and tumor rupture.

Surgical resection of the tumor remains the only treatment option. Complete tumor resection can be accomplished in about 50% to 80% of cases. [225] For gastric tumors, limited dissection is known to achieve results comparable to those of extended resection. [228] Lymphadenectomy is not routinely practiced because of the low incidence of lymph node metastasis. In general, vital structures should be preserved if gross tumor clearance can be attained. The use of adjuvant therapy remains experimental, and there is no effective therapy for advanced metastatic disease. Promising results were achieved by the use of tyrosine kinase inhibitor in a single patient with metastatic GIST that expressed CD117. [229] This may be a new direction for treatment development, but results need to be confirmed in larger series.

Because of the low incidence of this disease, many of the previous studies were limited to small numbers of patients. The overall 5-year survival of these patients ranges from 28% to 65%. [225] Several features may predict higher chances of survival. These include female gender, tumor size less than 5 cm, and complete tumor resection. However, recurrence of disease is common and can occur years after surgery. A previous report estimated that only about 10% of patients were free of disease after long-term follow-up, [230] and long-term surveillance is necessary to detect tumor recurrence.

Gastric Carcinoid Tumors

Carcinoid tumors are traditionally divided into foregut, midgut, and hindgut in origin. Foregut carcinoids comprise bronchial, gastric, and duodenal carcinoids. By far, the most frequent sites of gastrointestinal carcinoids are the small bowel, appendix, and rectum. Gastric carcinoid tumors are rare, accounting for only about 3% of all carcinoids. [231]

Three distinct types of gastric carcinoid tumors have been proposed, with characteristic biologic behavior and prognosis. Enterochromaffin-like cells are the main endocrine cell types in type 1 and type 2 disease. Type 1 is the most frequent form (about 65%) of gastric neuroendocrine tumor and is localized in the oxyntic mucosa at the fundus and body. Patients usually have chronic atrophic gastritis, achlorhydria, and pernicious anemia. They often have elevated basal gastric levels and are highly sensitive to trophic stimulus of gastrin. Type 1 tumors tend to be small (<100 mm) and multiple. They are considered the most benign of the three forms of carcinoid tumors and have a relatively low frequency of metastasis, between 9% and 23%. [232], [233], [234] and [235] Type 2 tumor is associated with the gastrin-producing neoplasms in Zollinger-Ellison syndrome as part of the multiple endocrine neoplasia syndrome type I. This type of carcinoid has intermediate metastatic potential. By contrast, type 3 or sporadic carcinoid lesions are encountered less frequently. They are not associated with hypergastrinemia and are frequently solitary large lesions in nonatrophic mucosa. This type of tumor behaves more aggressively, with a high tendency of advanced local invasion and metastasis. [232]

The clinical presentations of gastric carcinoids are usually nonspecific and variable including pain, vomiting, anemia, and bleeding, or they can be discovered incidentally during endoscopy for other reasons. Presentation with the typical carcinoid syndrome of flushing, diarrhea, cutaneous edema, and bronchoconstriction is uncommon. [232], [234] This is more closely related to the secretion of histamine than to that of substance P, serotonin, or 5-hydroxyindoleacetic acid. In a review of the cases identified by the Surveillance, Epidemiology, and End Results program of the National Cancer Institute from 1973 to 1991, 52.9% of gastric carcinoids were localized, 10.3% were regionalized, and 20.6% with associated with distant metastasis. [231]

Upper gastrointestinal endoscopy is the most useful diagnostic tool, and typical lesions are small, rounded, submucosal masses that may often be yellow. [236] There is no definite histological criterion for establishing the degree of malignant potential, and the best indicator available is still the evidence of invasive growth and the presence of regional or distant metastases. [231] Because of the rarity of this disease, the best treatment remains controversial. For small type 1 gastric carcinoids, local removal such as by endoscopic polypectomy may be adequate. Surveillance endoscopy is recommend at 6-month intervals after initial removal. Antrectomy can be considered in patients with lesions larger than 1.0 cm, more than five lesions, and lesion recurrence after initial removal. [234] This operation can reduce serum gastrin levels and can thus result in regression of enterochromaffin-like cell carcinoids. [237] For type 3 gastric carcinoids, a more radical approach is necessary because of the high malignant potential and high risk of metastasis. Complete or partial gastrectomy with regional lymph node dissection is recommended. [238] Treatment with interferon-a or octreotide may be used to control symptoms associated with carcinoid syndrome, although the use of these agents may be more appropriate in patients with midgut carcinoid and pancreatic endocrine tumors. [233]

The overall 5-year survival in patients with all types is 49%. However, patients with type 3 gastric carcinoids have a significantly worse prognosis than do those with type 1 and 2 lesions. Five-year survival of patients with localized gastric carcinoid is 64.3%, whereas survival in patients with distant metastases drops to 10%. [231]

GASTRIC POLYPS

Gastric polyps are uncommon. The estimated incidence is less than 1% in autopsy or radiologic surveys. [239] However, up to 50% of gastric polyps are discovered incidentally during endoscopy for unrelated symptoms. [240] They may be solitary or multiple, and occasionally they are associated with other polyposis syndromes such as familial adenomatous polyposis, Peutz-Jeghers syndrome, Cowden syndrome, and Cronkhite-Canada syndrome. Different classification systems for gastric polyps have been proposed in the past, resulting in confusion. In essence, gastric polyps can be divided into neoplastic and nonneoplastic lesions. Neoplastic polyps are composed of adenomas similar to those present in the colon, whereas nonneoplastic polyps comprise hyperplastic, hamartomatous, inflammatory, and heterotopic types. One of the most important characteristics of gastric polyps is the potential for malignant transformation, particularly in adenomatous polyps. It is believed that the histological composition and size of the polyps may determine their malignant potential. The frequency of malignant transformation varies from 6% to 75% in the literature. [241]

In most reports that do not include fundic gland polyps, hyperplastic polyps are the most common (70% to 90%) gastric epithelial polyps. [239] Hyperplastic or

regenerative polyp frequently occurs in association with gastritis, particularly *H pylori*–associated gastritis. In general, the malignant potential of the hyperplastic polyp is minimal and has been reported to be between 1.5% and 3%. [242], [243]

Adenomatous polyps account for about 10% of gastric polyps, [244] and they are more prevalent in areas with a high incidence of gastric cancer. This type of polyp is associated with a higher risk of malignant transformation, and the risk is size dependent. [245] Coexistence of adenoma with carcinoma is not uncommon.

Fundic gland polyps are also known as fundic gland hyperplasia or glandular cysts. They are recognized as the most frequent type of gastric polyp, accounting for nearly half of all gastric polyps. [244] These lesions are composed of fundic glands lined with increasing numbers of normal-appearing parietal and chief cells. They are located exclusively in the acid-producing mucosa and thus are not found in the antrum. Notably, the polyp may regress and may have no malignant potential. Gastric polyps occurring as part of the familial adenomatous polyposis syndrome are usually fundic gland polyps.

Occasionally, misplaced gastric glands can present as a polypoid mass in the stomach. These heterotopic polyps are composed of tissue from the neighboring pancreas or duodenum. Heterotopic pancreas is frequently found in the prepyloric region and presents as a submucosal, nipplelike lesion, composed of mainly exocrine glands and ducts.

In a review by the American Society for Gastrointestinal Endoscopy, [95] all polyps causing symptoms, such as bleeding and obstruction, and those larger than 2 cm should be removed. Polyps smaller than 2 cm can be initially examined by biopsy and excised. If biopsies reveal adenomatous change, endoscopic excision should be considered. Most gastric polyps can be safely removed by snare polypectomy. Complications such as bleeding, perforation, and abdominal pain are rare. Surveillance endoscopy is recommended only in adenomatous gastric polyps 1 year after removal, to assess for any recurrence or the development of new polyps.

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CHAPTER 70

Neal E. Seymour and Dana K. Andersen

SURGERY FOR PEPTIC ULCER DISEASE AND POSTGASTRECTOMY SYNDROMES

The surgical treatment of peptic ulcer is most frequently required when complications of previously unappreciated ulcers occur. The elucidation of peptic ulcer pathogenesis and development of therapies that directly address the underlying causes of gastroduodenal mucosal injury have defined the management of uncomplicated ulcers. Eradication of *Helicobacter pylori,* withdrawal of nonsteroidal antiinflammatory drugs, and the use of potent antisecretory medications have essentially removed the surgeon from what can now be viewed as a historically important role in the elective management of peptic ulcer. However, old surgical therapies for peptic ulcer may continue to pose problems in clinical management, and it remains important to understand the impact of vagotomy and other surgical alterations of the foregut. It is also vitally important to understand when surgical care can contribute to the management of patients with complicated peptic ulcers.

ELECTIVE SURGERY FOR PEPTIC ULCER DISEASE

Duodenal Ulcers

Elective operation for peptic ulcer disease is now rarely offered in developed nations. Elective surgical treatment of duodenal ulcer implies that medical treatment has been ineffective or unfeasible. True failure of appropriate medical therapy is exceedingly rare, and it should prompt investigation for other disease such as gastrinoma. Antisecretory medications (generally proton pump inhibitors) given in the event of anti- *Helicobacter* treatment failure can represent a considerable expense, depending on duration of treatment. Cost analyses of elective surgical treatment versus maintenance antisecretory therapy, taking into account the impact of eventual treatment failures (ulcer recurrence) or of long-term morbidity, are not likely to be performed, and it is difficult to make rational arguments for one versus another treatment based on these factors. Patients who are noncompliant with reasonable medical regimens or who do not have reliable access to medication may be offered surgical treatment in the hope of definitively eradicating the disease.

Gastric Ulcers

The indications cited for elective operation for duodenal ulcer apply for gastric ulcer as well. However, it falls to the clinician caring for a given patient to decide when multiple cycles of evaluation and nonsurgical treatment may be excessively difficult, expensive, and burdensome to the patient. Nonhealing of any gastric ulcer raises the possibility of gastric carcinoma and warrants consideration of surgical treatment. Multiple endoscopic biopsies of the ulcer, if results are negative, reduce but do not eliminate the possibility of gastric carcinoma. [1] The clinical behavior of the ulcer as well as the results of endoscopic biopsy should influence the timing of operation, to avoid delaying treatment of a potentially early gastric cancer.

"Giant" Ulcers

The question whether ulcer size should influence the decision to offer early operative treatment has long been a source of controversy. Data suggesting that gastric and duodenal ulcers larger than 3 cm are more likely to bleed, perforate, or prove intractable were acquired before the availability of proton pump inhibitor and anti-*Helicobacter* therapy. [2], [3] Of 62 cases of giant gastric ulcers treated in Singapore between 1984 and 1989, 31% demonstrated penetration into adjacent structures (pancreas, liver, colon). [3] Malignant disease was found after histological examination in 13% of resected specimens, although it was not clear to what extent efforts were made preoperatively to obtain this information. Unexpected malignancy has not been a problem noted in more recent reports on the outcomes of medical therapy for large ulcers. These reports indicate that ulcer size has little bearing on response to appropriately aggressive treatment, [4], [5] and [6] although surgery may be necessary for ulcer complications in as many as 16%. [6]

SURGERY FOR DUODENAL ULCER

Vagotomy in the Treatment of Duodenal Ulcer

The reduction of acid secretion by gastric denervation serves as the physiological basis of all modern surgical treatments for duodenal ulcer. Truncal vagotomy reduces basal and maximal acid output by 85% and 50%, respectively. [7] This reduction in acid secretion results from the removal of the direct acetylcholine influences on the parietal cell, as well as reduction in sensitivity of the parietal cell to gastrin and histamine. [8], [9] Although pepsin may play a role in mucosal injury in patients with duodenal ulcer, the clinical benefit of vagotomy in reducing pepsinogen secretion and gastric proteolytic activity is less clear. [10] Gastric motor function is dramatically altered by truncal vagotomy. Receptive relaxation, or decrease in muscular tone of the proximal stomach with ingestion of food, and antral peristaltic and proximal gastric nonphasic motor activity are abolished by vagotomy. [11], [12] Loss of vagus-dependent gastric motor function and regulation of pyloric resistance require that truncal vagotomy be performed in conjunction with a procedure to facilitate gastric emptying. The combined effect of vagotomy and bypass or destruction of the pylorus appears to be differential emptying of solids and liquids. Emptying of liquids is accelerated, probably secondary to increased intragastric pressure with loss of receptive relaxation, [13], [14] whereas emptying of solids is more variable. [15], [16] Vagotomy-induced acceleration of emptying of liquids may contribute to dumping syndrome after gastric drainage or resection procedures.

Gastric denervation procedures have been devised that preserve a more normal neural relationship with the remainder of the gastrointestinal tract than is possible after truncal vagotomy (Fig. 70-1). Selective vagotomy preserves the celiac and hepatic branches of the vagus but still results in gastric atony and requires a concomitant drainage procedure to facilitate gastric emptying. The advantage of this procedure over truncal vagotomy in preventing postvagotomy diarrhea is not clear. [17] Highly selective vagotomy preserves the anterior and posterior nerves of Latarjet as well as their terminal branches to the pyloroantral region, while selectively denervating the acid-producing parietal cell mass. The previously described accelerated emptying of liquids from the stomach after vagotomy is least after

highly selective vagotomy. [14] Highly selective vagotomy has become the most widely accepted antiulcer procedure in the elective setting because it avoids the morbidity associated with loss of pyloroantral function.

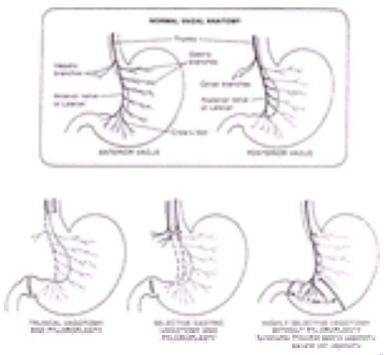

FIGURE 70-1. Normal vagal anatomy and types of vagotomy. Truncal and selective vagotomies are shown with a gastric drainage procedure (pyloroplasty). During truncal vagotomy, nerve trunks are divided above the celiac and hepatic branches. During selective vagotomy, the anterior and posterior nerves are divided *distal* to the celiac and hepatic branches, thus preserving extragastric innervation of the gastrointestinal tract and theoretically reducing the risk of gallstone formation and diarrhea. During highly selective vagotomy, individual branches of the anterior and posterior nerves of Latarjet to the body of the stomach are divided, sparing the terminal branches to the pylorus and antrum ("crow's foot"). Pyloroantral motor function is thus preserved, and gastric drainage is unnecessary. (From Debas HT, Orloff SL. Surgery for peptic ulcer disease and postgastrectomy syndromes. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995.)

Specific Operations

Vagotomy and Drainage If truncal or selective vagotomy is performed, the clinical effects of resulting gastric atony can be minimized by pyloroplasty, gastroduodenostomy, or gastrojejunostomy (Fig. 70-2). The Heineke-Mikulicz pyloroplasty has historically been the most widely used method of drainage. Vagotomy and drainage offer no real advantage over highly selective vagotomy in terms of recurrent ulcer risk, and they add the potential complications of diarrhea and dumping syndrome. The most common use of this procedure has been in the setting of bleeding duodenal ulcer in which a gastroduodenotomy is required to gain access to the bleeding site, and closure as a pyloroplasty is very convenient.

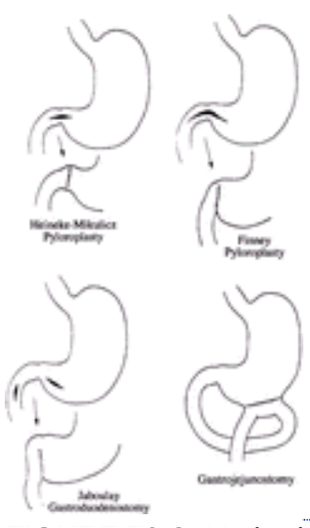

FIGURE 70-2. Methods of gastric drainage: The Heineke-Mikulicz pyloroplasty is most frequently used in cases of duodenal ulcer bleeding, because the transpyloric incision required to expose the bleeding ulcer can easily be closed in a transverse orientation. The Finney pyloroplasty and Jaboulay gastroduodenostomy were historically performed when the duodenal bulb was badly deformed by chronic ulcer scar. A gastrojejunostomy may be performed if access to the pyloroduodenal area is limited.

Highly Selective Vagotomy The ideal operation for peptic ulcer disease is one that is associated with a minimal risk of ulcer recurrence and is tolerated extremely well by patients to whom it is offered. In practice, although highly selective vagotomy is associated with better patient tolerance than other antiulcer procedures, there is considerable variability in reports pertaining to how well it achieves the goal of minimizing risk of ulcer recurrence. Parietal cell vagotomy, superselective vagotomy, and proximal gastric vagotomy are synonymous terms used to describe the procedure. Highly selective vagotomy is accomplished by dividing the terminal branches of the nerves of Latarjet to the lesser curvature of the stomach, from a point approximately 6 cm proximal to the pylorus to a point at least 5 to 6 cm proximal to the esophagogastric junction on the esophagus (Fig. 70-3). These nerves are found in the anterior and posterior peritoneal leaves of the lesser omentum (gastrohepatic ligament). The two to three terminal branches of the nerves of Latarjet ("crow's foot") to the antrum and pylorus are preserved. Great care must be taken to ensure sufficient periesophageal dissection, because high vagal branches can easily be missed.

FIGURE 70-3. Highly selective vagotomy. Selective denervation of the parietal cell mass is achieved by careful division of all proximal anterior and posterior vagal branches, distally to a point approximately 6 cm from the pylorus. All surrounding tissues are dissected from a 5- to 6-cm segment of esophagus proximally. Antropyloric innervation must be carefully preserved to prevent postoperative gastric stasis. The bare area on the lesser curvature can be closed with seromuscular sutures, but it is more typically left undisturbed.

Ulcer recurrence rates after highly selective vagotomy are variable and are thought to depend on operator skill as well as duration of follow-up. Ulcer recurrence rates between 9% and 17% have been reported at centers where the procedure has been done frequently. [18], [19], [20], [21], [22], [23], [24], [25], [26], [27], [28], [29], [30], [31], [32], [33], [34], [35], [36], [37], [38] and [39] A consistently described, time-dependent rise in the incidence of recurrent ulcer suggests that long periods of follow-up (>10 years) are required to examine the results of highly selective vagotomy reliably. [23], [25] Based on patterns of Congo red staining (a surface pH lower than 3.0 will stain black) during standard highly selective vagotomy, an "extended" highly selective vagotomy has been proposed that adds division of the right and left gastroepiploic pedicles to the other cardinal features of the operation. [26] Reports describing the use of extended highly selective vagotomy with intraoperative assessment of completeness of gastric

denervation suggest that very low rates of recurrent ulceration can be achieved by the skilled application of these procedures. [26], [27], [28] and [29] Highly selective vagotomy has been shown to result in an immediate decrease in both basal and maximal acid outputs. [20], [30] Afterward, there is a time-dependent recovery of basal and maximal acid outputs to approximately 30% and 50% of preoperative levels, respectively. [20] Patterns of acid secretion that have been associated with increased risk of ulcer recurrence after highly selective vagotomy include high preoperative or persistently elevated postoperative basal acid outputs. [31], [32] High postoperative maximal acid output has also been reported to correspond to an increase in the risk of ulcer recurrence. [33], [34] These patterns of postoperative acid secretion in patients evaluated on a long-term basis after highly selective vagotomy were correlated with untreated *H pylori* infection. [35] In addition to selected patterns of gastric acid hypersecretion, there are data indicating that the ulcer recurrence rate is higher in patients with prepyloric ulcer treated by highly selective vagotomy than in patients with ulcers of the first portion of the duodenum (Fig. 70-4). [36], [37] and [38] The role of *Helicobacter,* if any, and the potential impact of "extended" procedures in this pattern of disease are uncertain.

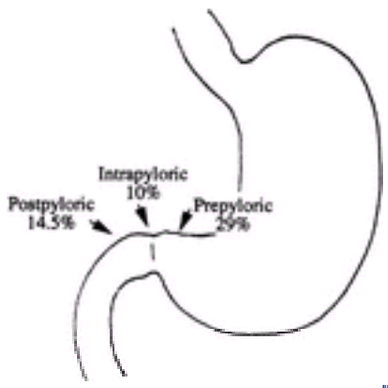

FIGURE 70-4. Ulcer recurrence rate after highly selective vagotomy according to preoperative site of ulcer. The data reported by Schafmayer and colleagues reflect 5-year follow-up of 392 patients. Prepyloric ulcer location appears to be associated with a higher risk of ulcer recurrence than duodenal or intrapyloric location. (Adapted from Schafmayer A, Börger WH, Köhler H, et al. Recurrent ulcers after proximal gastric vagotomy: special aspects of the prepyloric ulcer. Dig Surg 1989;6:4).

Even in the current era of peptic ulcer treatment, highly selective vagotomy will probably continue to play a role, albeit minor, in the management of duodenal ulcers that are *Helicobacter* negative or unresponsive to repeated cycles of antibiotic therapy. Because highly selective vagotomy is now performed so infrequently, referral of patients to tertiary care centers for this highly technical procedure is advisable. [39]

Vagotomy and Antrectomy Of the procedures historically performed for duodenal ulcer, antral resection coupled with truncal or selective vagotomy is associated with the lowest risk of ulcer recurrence (0% to 2%). [40], [41] However, the risk of postgastrectomy and postvagotomy problems associated with this procedure rendered it a less-favored option as compared with highly selective vagotomy even before the advent of anti- *Helicobacter* therapy. Removal of the gastrin-secreting tissues of the distal stomach retains a large gastric reservoir, which must be anastomosed to either the duodenum (Billroth I) or the proximal jejunum (Billroth II) (Fig. 70-5). After antrectomy for benign disease, gastroduodenostomy (Billroth I) is preferred to gastrojejunostomy, to avoid the problems associated with an afferent anastomotic jejunal limb and duodenal stump. To avoid retained antrum syndrome, it is advisable histologically to verify the presence of duodenal Brunner glands at the distal margin of resection by frozen section after antrectomy, particularly if scarring or active inflammation makes clear identification of the pylorus difficult.

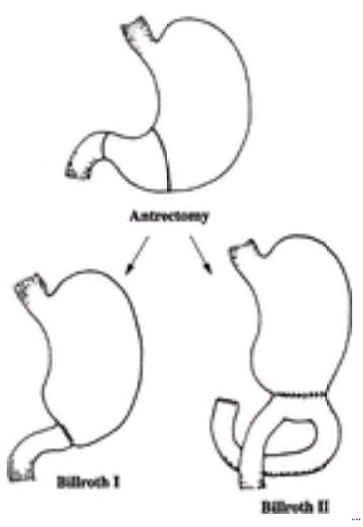

FIGURE 70-5. Reconstruction after gastrectomy: Billroth I (gastroduodenostomy) and Billroth II (loop gastrojejunostomy). Billroth I is the preferred reconstruction after gastrectomy for benign disease.

The decision to perform vagotomy and antrectomy for benign duodenal ulcer disease is an infrequent and difficult one. In the elective setting, highly selective vagotomy offers the advantage of greater patient tolerance, albeit with a higher risk of ulcer recurrence. In complex situations when *Helicobacter*-negative ulcers have failed to respond to antisecretory therapy and ulcer recurrence may be more likely after highly selective vagotomy (prepyloric ulcer or ulcers that are highly resistant to treatment with antisecretory agents), vagotomy and antrectomy may be more satisfactory operations despite their higher associated long-term morbidity.

Laparoscopic Procedures for Peptic Ulcer Disease Posterior truncal vagotomy and anterior seromyotomy, described by Taylor and colleagues, [42], [43] have been performed laparoscopically without substantially modifying the procedures [44], [45] and [46] (Fig. 70-6). Gastroparesis, dumping syndrome, and postvagotomy diarrhea are encountered no more frequently than after highly selective vagotomy, despite division of the posterior vagal trunk. The major concerns surrounding these operations relate to their unproven efficacy. No data examining extent of residual acid-secreting tissue either intraoperatively or postoperatively are available, nor has eventual gastric reinnervation after anterior seromyotomy been excluded. One report, which examined a modified stapled technique of anterior seromyotomy, described effectively reduced acid secretion and the absence of recurrent ulcer at a mean of 5 years after surgery in a small group of patients. [47] Similar results have been achieved with laparoscopic posterior truncal anterior highly selective vagotomy (Hill-Barker procedure). [48] The technical feasibility of laparoscopic highly selective vagotomy, [49], [50] and [51] as well as laparoscopic bilateral truncal vagotomy with pyloromyotomy for drainage, [52] has been reported, although the long-term results of these operations are unavailable.

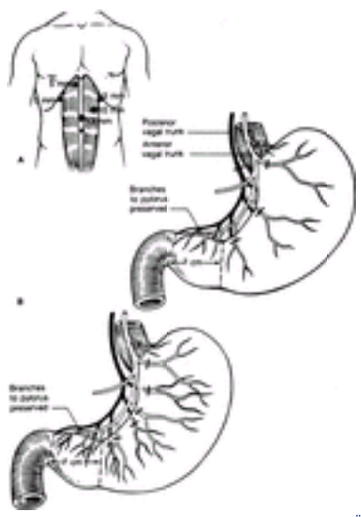

FIGURE 70-6. Laparoscopic operations for peptic ulcer. Procedures typically require five upper abdominal operative ports. Parietal cell denervation is accomplished by (**A**) anterior seromyotomy and posterior truncal vagotomy (Taylor procedure), interrupting the branches of the anterior nerve of Latarjet within the wall of the stomach, or (**B**) a more formal highly selective vagotomy, interrupting the branches of the posterior nerve of Latarjet as well. (From Debas HT, Orloff SL. Surgery for

peptic ulcer disease and postgastrectomy syndromes. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995.)

Laparoscopic truncal vagotomy and antrectomy have been performed at several centers worldwide, albeit in limited numbers. [53], [54], [55] and [56] These procedures are impressive technical accomplishments, and they offer the advantages conferred by laparoscopic access (decreased postoperative pain and early return to normal function). However, the long-term morbidity of such foregut alterations remains an active concern, irrespective of the method used for access to the abdomen.

SURGERY FOR GASTRIC ULCER

Like duodenal ulcers, benign gastric ulcers rarely require elective surgical management. However, the pathogenesis of gastric ulcers may be multifactorial, and nonhealing or recurrent ulceration is more frequent than after medical treatment of duodenal ulcers. Classification of gastric ulcer by location (Fig. 70-7) serves to define specific operations that could be applicable to management. [57], [58] Various surgical procedures may be used to treat benign ulcers at specific gastric sites (Fig. 70-8). Antrectomy inclusive of the ulcer is the standard operative approach for more distal (type I and II) ulcers. For more proximally situated ulcers on the lesser curvature, a narrow extension of the resection can be made to remove the ulcer en bloc with the antral specimen (Pauchet procedure). Reconstruction by Billroth I method is preferred to avoid a duodenal stump and afferent jejunal limb. Vagotomy provides no additional benefit over resection alone and is not routinely performed, except when a *Helicobacter*-negative patient has duodenal ulcers as well (type III). Local excision alone is technically feasible for small lesser curvature or gastric body ulcers, although it is associated with a high ulcer recurrence rate and has traditionally been reserved for situations in which more extensive procedures are contraindicated.

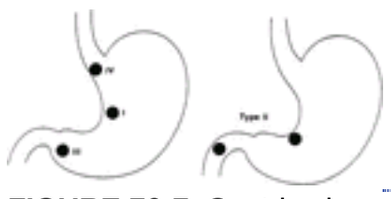

FIGURE 70-7. Gastric ulcer classification according to location. Type I describes an ulcer at the incisura angularis on the lesser curvature. A type II gastric ulcer is associated with a simultaneous duodenal ulcer. Type III describes a prepyloric ulcer, and type IV is a gastric cardia ulcer. (From Debas HT, Orloff SL. Surgery for peptic ulcer disease and postgastrectomy syndromes. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995.)

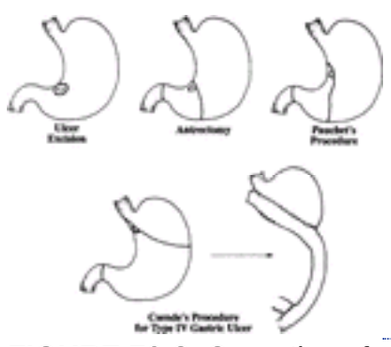

FIGURE 70-8. Operations for benign gastric ulcer.

Ulcers near the esophagogastric junction are more formidable technical challenges than distally situated ulcers. Csendes and colleagues [59] described resection of cardia ulcers (type IV) by subtotal gastrectomy that extends to the esophagogastric junction and is reconstructed with a large Roux-en-Y esophagogastrojejunostomy. These high gastric ulcers, although rare in the United States and Europe, accounted for 27% of the gastric ulcers reported by Csendes and associates in Chile.

SURGICAL TREATMENT OF PEPTIC ULCER COMPLICATIONS

Despite the availability of increasingly effective medical therapies, it has not been possible to demonstrate a clear reduction in the overall incidence of ulcer complications since the early 1980s. In contrast to the decrease in incidence of uncomplicated ulcers that preceded the availability of H_2-receptor antagonists, population-based studies suggest that the incidence of complicated ulcers did not diminish at all before 1990. [60], [61] and [62] Other data suggest that effective medication has been associated with a dramatic decrease in the incidence of such complications in children. [63] Mortality from complicated duodenal ulcers that require surgical treatment is high, particularly in elderly patients. [64] Expedient diagnosis and treatment offer the best chance of a favorable outcome, particularly in cases of massive hemorrhage or perforation. The immediate goals of surgical treatment are to control damage (e.g., suture of bleeding site, closure of perforation). Based on available data, which show a high incidence of *H pylori* infection in complicated ulcers, the role of definitive antiulcer procedures in the emergency setting has become questionable.

Hemorrhage

Life-threatening bleeding from duodenal and gastric ulcers requires prompt but carefully planned treatment, of which surgery can be an important component. Comprehensive management of upper gastrointestinal hemorrhage is beyond the scope of this chapter (see Chapter 33), but aggressive resuscitation and early identification of the bleeding source by endoscopy are the most important initial steps before direct treatment of the bleeding. Endoscopic bipolar cautery, heater probe, and chemical injection treatments have been established as expedient means of arresting hemorrhage. Avoiding operation in these often direly ill patients is thought ultimately to lessen mortality, although this benefit has been difficult to demonstrate clearly. A metaanalysis compiling data from 30 randomized trials of endoscopic therapy for nonvariceal bleeding showed a clear reduction in the need for surgery with all hemostatic methods employed. [65] However, a significant decrease in mortality was observed only for laser therapy and not for the more frequently used thermal contact methods. Despite the availability of advanced endoscopic methods, persistent or recurrent hemorrhage may occur in 20% to 33% of patients. [66] Mortality in the presence of this event may exceed 30%. [67]

Approximately 5% of patients with transfusion-requiring hemorrhage from peptic ulcers will require surgical treatment to control bleeding. [68] These patients can be separated from those not requiring surgery at decision points in a definable management algorithm (Fig. 70-9). Ongoing nonvariceal hemorrhage not amenable to endoscopic treatment requires immediate operation. The inability to control hemorrhage endoscopically may result from difficult ulcer location, high rate of bleeding, or insufficiently localizable bleeding point within the ulcer. Gastric ulcers are three times as likely to rebleed as duodenal ulcers. [69] Prompt surgical treatment after one or two unsuccessful endoscopic attempts to control bleeding is a highly definitive management strategy. High transfusion requirements have historically been associated with poor outcomes, and ongoing need for banked blood should be a general indicator of the need for surgical intervention. [68], [70] Several factors, in addition to rebleeding, have been associated with risk of mortality (Table 70-1). Immediate recognition of ongoing or recurrent hemorrhage, aggressive resuscitation, and prompt surgical management after failed endoscopic intervention are the best means of minimizing death rates from peptic ulcer bleeding.

Transfusion requirement >5 Units
Age >60 years
Shock on presentation
Concomitant medical illnesses

Adapted from ref. 68.

TABLE 70-1 Factors Predicting Mortality in Bleeding Duodenal Ulcer

FIGURE 70-9. Algorithm for management of bleeding peptic ulcer. (Adapted from Seymour NE. Surgery of the stomach and duodenum. In: Feldman M, ed. Gastroenterology and hepatology: the comprehensive visual reference. Philadelphia: Current Medicine, 1996.)

When operative management is undertaken, the most urgent goals are exposure and local control of the bleeding site. For bleeding ulcers of the first portion of the duodenum, this is achieved by a longitudinal incision across the pylorus and proximal duodenum. This permits exposure of the point of erosion into the gastroduodenal artery or branch vessel in the posterior wall of the duodenal bulb. Exposure through a duodenotomy alone with preservation of the pylorus has also been described. [71] Hemorrhage is controlled by direct suture, undersewing the vessel on either side of the bleeding (Fig. 70-10). The gastroduodenotomy can be closed as a pyloroplasty, and a truncal vagotomy can be performed. Alternatively, highly selective vagotomy with longitudinal closure of the gastroduodenotomy and precise reconstruction of the pylorus have been described. [72], [73] However, it is now unclear that the addition of vagotomy confers additional benefit once local control of bleeding is obtained. Reports of higher early ulcer rebleeding rates and associated mortality rates [74], [75] without vagotomy are subject to bias and are poorly controlled for perioperative medical therapy. Eradication of *H pylori* infection has been shown to prevent ulcer rebleeding in nonsurgical situations, [76] and it has become standard postsurgical treatment in conjunction with antisecretory therapy. The role of vagotomy for ulcer bleeding in the era of anti- *Helicobacter* therapy is very much in doubt, because the likelihood of ulcer recurrence after definitive postoperative medical therapy is probably less than the likelihood of significant postvagotomy symptoms.

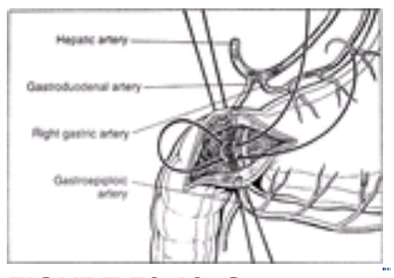

FIGURE 70-10. Suture control of bleeding posterior duodenal ulcer. Exposure is by longitudinal gastroduodenotomy. The bleeding point in the gastroduodenal artery is undersewn with nonabsorbable monofilament suture. Additional transfixing sutures along the course of the vessel above and below this point provide more secure hemostasis. (From Seymour NE. Surgery of the stomach and duodenum. In: Feldman M, ed. Gastroenterology and hepatology: the comprehensive visual reference. Philadelphia: Current Medicine, 1996.)

A bleeding gastric ulcer may be exposed by appropriate placement of a longitudinal gastrotomy, based on preoperative endoscopic data. If the gastric bleeding site was not visualized at endoscopy, both a pyloroduodenal incision and a longitudinal gastrotomy may be necessary to identify the lesion. Bleeding may be managed by antrectomy inclusive of the ulcer, ulcer excision alone, or simple oversewing of the bleeding site if the patient's condition is unstable.

Perforation

Perforation of a peptic ulcer occurs only after deep penetration of the ulcer through the full thickness of the stomach or duodenal wall to a free peritoneal surface. Perforation is generally followed by chemical peritonitis that, if untreated, can progress to bacterial peritonitis, sepsis, and death. Some perforations spontaneously seal, usually as the result of omental adhesion to the site.

Clinical suspicion of perforated duodenal ulcer may be prompted by abdominal pain, which is often very acute in onset. The differential diagnosis includes biliary tract disease and pancreatitis, although many other catastrophic abdominal problems can mimic a perforated ulcer, including acute appendicitis. The diagnosis of perforated gastric or duodenal ulcer must be considered in any patient with peritonitis of uncertain origin. Pneumoperitoneum may be observed on upright chest or left lateral decubitus radiographs in up to one half of patients.

Immediate abdominal exploration and repair of the perforation comprise the standard management, although this approach is probably not mandatory in all patients (Fig. 70-11). Factors predictive of mortality after ulcer perforation include major medical illness, preoperative shock, and duration of perforation for longer than 24 hours. [77] In a prospective evaluation of these risk factors in 259 patients with perforated ulcers, Boey and colleagues [77] reported a mortality rate of 100% in patients with all three risk factors. Other identified risk factors for mortality include advanced age, pulmonary disease, a perforation of more than 24 hours' duration, and glucocorticoid use. [78] An evaluation of patients treated by laparoscopic methods defined the APACHE II score as an important predictor of morbidity and mortality in perforated ulcer. [79]

FIGURE 70-11. Algorithm for management of perforated peptic ulcer. (Adapted from Seymour NE. Surgery of the stomach and duodenum. In: Feldman M, ed. Gastroenterology and hepatology: the comprehensive visual reference. Philadelphia: Current Medicine, 1996.)

Nonoperative management of perforated ulcer may be attempted in selected patients who present without diffuse peritonitis or sepsis. This requires demonstration of a contained perforation by water-soluble contrast study and should be attempted only in conjunction with nasogastric drainage and antibiotics. [80], [81] Reports advocating this approach indicate that abdominal exploration can be avoided in up to 70% of these highly selected patients. Increased mortality when necessary operative treatment is delayed by unsuccessful nonoperative management in patients who are more than 70 years old suggests that even careful observation may not be safe in this group. [82]

A perforated duodenal ulcer is repaired by applying a pedicle of greater omentum directly to the site of perforation and securing it there with absorbable sutures (Fig. 70-12). Direct suture closure of the inflamed edges of the perforation is at high risk of failure and is unnecessary if the omental patch is applied correctly. Although gastric ulcer perforations may also be repaired by omental patch, gastric resection inclusive of the ulcer is preferred, to lessen the risk of recurrent ulcer, provided the patient's clinical condition permits the more extensive procedure. Although highly selective vagotomy has been recommended in the setting of perforated duodenal ulcer, [83], [84] the addition of any antiulcer procedure to local closure is of questionable value. Current data indicate that more than 80% of duodenal ulcer perforations occur in the setting of *H pylori* infection, and postoperative treatment results in an expected low ulcer recurrence rate. [85]

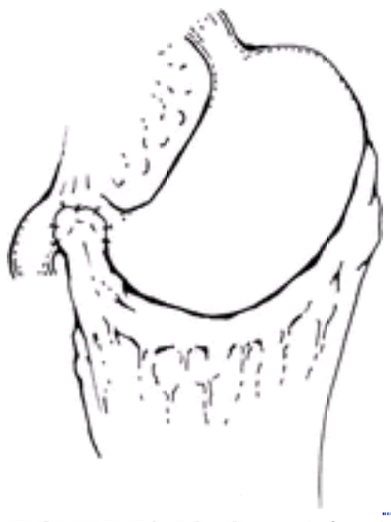

FIGURE 70-12. Omental patch of perforated duodenal ulcer. If no omentum is available, falciform ligament may be similarly used to patch the perforation.

Numerous reports indicate that peritoneal toilet and omental patch closure of peptic ulcer perforation can be effectively performed by laparoscopic method. [86], [87] and [88] The essential features of this procedure are the same as would be accomplished by open technique. Reported results of uncontrolled trials are excellent, [86], [87] with rapid recovery times and extremely low mortality rates. In a randomized, prospective evaluation of open versus laparoscopic management, although analgesic use in the laparoscopic group was less, there were no significant differences noted in duration of hospital stay, time to resume diet, and duration of nasogastric decompression. [89]

Obstruction

Gastric outlet obstruction is the peptic ulcer complication encountered least frequently by the surgeon. Obstruction generally results from a combination of chronic scarring, acute edema, and spasm in the pyloroduodenal area. Patients may present with significant weight loss and complaints of early satiety, bloating, nausea, and vomiting. If vomiting is significant, hypovolemia, alkalosis, and hypokalemia may be present. Early upper endoscopy is mandatory to rule out carcinoma and is generally reliable. After a 1-week course of total parenteral nutrition, nasogastric decompression, and antisecretory therapy, in the absence of improvement, surgical treatment should be undertaken. If Heineke-Mikulicz pyloroplasty is unfeasible because of pyloroduodenal deformation, then Finney pyloroplasty, Jaboulay gastroduodenostomy, or simple gastrojejunostomy may be performed. There are few data defining the role of *H pylori* in gastric outlet obstruction, and truncal vagotomy has typically be performed in conjunction with these decompressive procedure. Because of concerns regarding denervation of an enlarged, paretic stomach, highly selective vagotomy has been used in the management of pyloroduodenal obstruction, in combination with pyloric dilation, [90], [91] and [92] or with surgical decompression. [93], [94] and [95] Although the necessary bypass or destruction of the pyloric mechanism in this setting may seem to negate the advantages of highly selective vagotomy, dumping syndrome appears to occur less frequently than after similar drainage procedures performed in conjunction with truncal vagotomy. [95] A prospective comparison of procedures performed for gastric outlet obstruction suggested that highly selective vagotomy with gastrojejunostomy yields nutritional results superior to those with selective vagotomy with either antrectomy or pyloroplasty. [96]

COMPLICATIONS OF SURGERY FOR PEPTIC ULCER

Early Complications

The potential complications after gastric surgery are numerous (Fig. 70-13.). Bleeding, infection, and thromboembolism are potential complications after any abdominal procedure. Acute afferent limb obstruction with potential duodenal stump leak after Billroth II reconstructions remains a feared complication. A retrospective analysis of Billroth II resections for duodenal ulcer in 545 patients revealed a 7% incidence of major complications and a 1.5% mortality rate. [97] Multivariate analysis identified cirrhosis, leukocytosis greater than 10,000 white blood cells/mm [3], previous abdominal surgery, and ulcer involvement of contiguous structures as significant risk factors for complications and death.

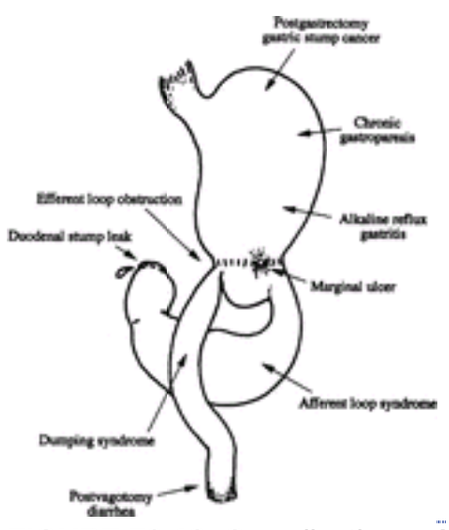

FIGURE 70-13. Complications of operations for peptic ulcer.

Recurrent Ulcer

Ulcer recurrence rates after operations for chronic peptic ulcer vary according to the nature of the procedure performed (Table 70-2). Although in the past, recurrent ulcer was attributed to a variety of factors, it now appears that *H pylori* infection accounts for most postoperative recurrent ulcers. [98], [99], [100] and [101] However, other specific issues associated with recurrent ulcer must be considered when examining specific cases. For example, Billroth II gastrojejunostomy (but not Billroth I or pyloroplasty) may result in retained antral mucosa in the closed duodenal stump that can be a source of hypergastrinemia. Recurrent ulcer after previous truncal vagotomy raises the possibility that the vagotomy may have been incomplete. Completeness of previous vagotomy can be determined by quantification of gastric acid with sham feeding. [102] Unsuspected gastrinoma must be considered in any patient with recurrent ulcer after what is believed to be an "adequate" operation, although

this situation is exceedingly rare.

OPERATION	RECURRENCE RATE (%)	REMARKS
Gastric resection	2–5	Varies according to amount resected; 75% optimal
Vagotomy and drainage	10–15	Pyloroplasty most frequent drainage procedure
Vagotomy and antrectomy	0–2	Lowest recurrence rates
Parietal cell vagotomy	10–17	Operator-dependent results

Adapted from ref. 103.

TABLE 70-2 Ulcer Recurrence Rates After Surgical Procedures

It is now standard therapy to treat *H pylori* infection in any patient with recurrent ulcer. In patients found on study to be infected in the absence of ulcer recurrence, specific treatment guidelines are more difficult to define. In *H pylori*–negative patients, effective antisecretory therapy is generally used on a maintenance basis to treat recurrent ulcers, to avoid further surgical treatment. Surgery for postoperative recurrent ulcers is a significant challenge not only because of the technical difficulties associated with reoperative upper abdominal surgery but also because a procedure more radical than the first is usually required to address the patient's disease. [103] For example, in a patient is known to have had an incomplete truncal vagotomy, repeat vagotomy is an appropriate option, as is antrectomy in a patient with recurrent ulcer after pyloroplasty.

Long-Term Complications of Surgery for Peptic Ulcer

Surgical alterations of gastric innervation, pyloric architecture, and gastric capacity may have profound effects on nutrition and quality of life. Even though elective operations for peptic ulcer are performed far less frequently than in the past, patients with chronic postoperative problems may continue to be encountered in clinical practice. Nutritional disturbances were well described and commonly encountered after extensive gastrectomy, but they occur rarely now that this procedure has been abandoned in the treatment of benign ulcer disease. Patients who continue to be seen after having had subtotal (three fourths) gastrectomy in the remote past may have disorders of calcium homeostasis or significant anemia secondary to iron and, less frequently, vitamin B_{12} deficiency, and they require appropriate nutritional supplementation. Extensive gastrectomy can also be associated with symptoms of a small gastric reservoir. Patients typically complain of early satiety and a sensation of epigastric postprandial discomfort that can be prevented by eating small, frequent meals. Although quality-of-life alterations may be significant, partial gastrectomy for benign ulcer disease does not appear to impart an increased risk of mortality beyond 1 year after the surgical procedure. [104]

Dumping Syndrome

Early dumping syndrome is thought to result from unregulated egress of gastric contents into the small intestine after gastroenteric anastomosis or pyloroplasty. It consists of vasomotor and gastrointestinal symptoms such as dizziness, flushing, and nausea, which typically occur 15 to 30 minutes after eating (Table 70-3). The precise pathophysiology of dumping is unclear, but current data suggest that rapid fluid shifts from the intravascular space to the gut lumen, [105] and [106] and release of vasoactive substances such as vasoactive intestinal polypeptide, neurotensin, and possibly motilin [107], [108] and [109] may be underlying causes. Symptoms are most marked after ingestion of high-carbohydrate, high-osmolarity substances and may be worsened if large volumes of liquids are also consumed. Up to 50% of patients will experience some degree of dumping symptoms after vagotomy and drainage. These symptoms are mostly mild and respond to minor dietary alterations such as avoidance of high-sugar foods and excessive fluid intake with meals. Most dumping symptoms improve with time, but severe dumping for extended periods can occur in 1% of cases. Octreotide acetate is generally effective in treating severe dumping symptoms that have not responded to appropriate dietary alterations. [110], [111]

VASOMOTOR	GASTROINTESTINAL
Tachycardia	Nausea
Palpitations	Vomiting
Sweating	Crampy abdominal pain
Flushing	Diarrhea
Dizziness	Belching
Syncope	

TABLE 70-3 Clinical Features of Early Dumping Syndrome

"Late" dumping syndrome consists of symptoms similar to those already described, but they occur 2 to 4 hours after eating. These are probably related to rapid delivery of sugars to the small intestine and an excessive serum insulin response to the resulting rapid rise in blood sugar. As in early dumping, dietary modification is generally effective in preventing late dumping symptoms. The addition of pectin and acarbose to the diet to delay carbohydrate absorption in the small bowel has been advocated for patients with continued symptoms after appropriate dietary changes. [112] Octreotide acetate is recommended in cases that are otherwise refractory to treatment.

Patients who which fail to respond significantly to the medical treatments described are extremely rare. Reversed jejunal segment Roux-en-Y gastrojejunostomy has been reported to achieve relief of dumping symptoms in 65% of the most severe cases. [113] However, most results of remedial operations for dumping predate the use of octreotide, and the role of these procedures is uncertain, given the demonstrated effectiveness of this agent.

Postvagotomy Diarrhea

Chronic diarrhea is a potential complication of truncal vagotomy and, to a far lesser extent, of selective and highly selective vagotomy. Although this clinical problem has been attributed to changes in intestinal motility after division of the posterior trunk proximal to the celiac branch, its actual pathogenesis is uncertain. Among the possible factors that may influence bowel habit changes after vagotomy are diminished intestinal transit time, decreased absorption, increased excretion of bile acids, and release of humoral factors that may stimulate episodic secretory diarrhea. Although clinically severe diarrhea is held to be a rare occurrence, the actual incidence of significant change in bowel habit after vagotomy is unclear, and reported rates range between 5% and 40%. [114], [115] Patients usually describe waxing and waning symptoms, although less frequently unrelenting diarrhea may occur. Kaolin-pectin may be helpful for very mild cases, but more typically loperamide or diphenoxylate/atropine are required for adequate relief.

Alkaline Reflux Gastritis

Excessive jejunogastric or duodenogastric reflux after gastric resection or drainage procedure for peptic ulcer has been associated with a syndrome of chronic abdominal pain and, less frequently, bilious vomiting. Although it is uncertain which components of the refluxate mediate gastric injury, bile acids were previously held to be likely candidates. An infrequently performed alkaline Bernstein test [116] has been described to assist in the diagnosis. Work characterizing histological change in the stomach after gastrectomy produced an association between *H pylori* and some of the changes that have generally been attributed to alkaline reflux. [117] However, at present it is uncertain whether *H pylori* plays a causative role in this clinical problem. The treatment of alkaline reflux symptoms can be quite difficult. Bile acid binding resins and sucralfate are not generally successful, [118], [119] and the most severe cases may require operative treatment to separate the gastric mucosa from pancreatobiliary secretions. This is accomplished by constructing a Roux-en-Y gastrojejunostomy with the Roux limb at least 45 to 60 cm in length to minimize the chances of ongoing reflux of alkaline small bowel contents into the stomach. [120] Although this approach is highly effective in controlling reflux, long-term follow-up suggests that alkaline reflux-like symptoms may continue to be reported by up to one half of patients. [121], [122] These clinical problems are seen less frequently as the number of operations for peptic ulcer has waned and the population of patients with prior surgical procedures for peptic ulcer grows older.

Delayed Gastric Emptying

Although symptoms of early satiety, nausea, and bloating are frequently reported in the early postoperative period after gastric surgery, chronic gastroparesis is an unusual occurrence. It most frequently observed after truncal vagotomy, but it may complicate any operation for peptic ulcer. Its incidence is smallest after highly selective vagotomy, which preserves antral motility. Risk factors for postoperative gastroparesis symptoms include diabetes mellitus and gastric outlet obstruction. [123] Treatment with metoclopramide or erythromycin can alleviate the symptoms of chronic postoperative gastroparesis. On the rare occasion when gastroparesis follows highly selective vagotomy, a drainage procedure may be necessary to facilitate gastric emptying. Although an extremely rare circumstance, some patients may require total gastrectomy to relieve unremitting symptoms. [124]

Gastric Cancer

Gastric adenocarcinoma in the proximal gastric stump after distal gastrectomy is a long-delayed event of uncertain pathogenesis. Hypochlorhydria, alkaline reflux, diminished gastrin production, uneradicated *H pylori* infection, and nitrosation have been suggested as possible causative factors. [125], [126] and [127] Gastric cancers diagnosed within 5 years of an initial gastrectomy are not generally regarded as true gastric remnant cancers, [128] although this is a somewhat arbitrary determination. Risk appears to be time dependent, reaching a four- to fivefold increase over the age- and gender-matched general population at 20 to 25 years after gastrectomy. [125] The precise incidence of gastric stump cancer is uncertain, and rates of 1% to 4% have been reported. [129], [130], [131] and [132] In a group of 285 patients operated on for benign ulcer disease in Finland between 1948 and 1954 and followed over 30 years, 2.1% developed cancer in the gastric remnant. [129], [130] Given this significant cancer risk, aggressive endoscopic screening has been recommended as a means of early detection. In 153 patients in the United States examined, including by biopsy, at least yearly over a 14-year period of surveillance, 8.4% developed severe dysplasia, and 54% of these were eventually proven to have adenocarcinoma. [132] All patients with cancer underwent total gastrectomy and were found to have limited disease. All had survived until the time of the report. Although it is a commonly held belief that gastric remnant cancers present at a more advanced stage and have a worse prognosis than gastric cancers as a whole, a comparison of stage of disease and survival for gastric remnant cancers and for proximal cancers of the previously unoperated stomach failed to show such a difference. [131] Surgical treatment for gastric remnant cancer usually requires completion of the gastrectomy or an esophagogastrectomy.

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CHAPTER 71

John C. Rabine and Timothy T. Nostrant

MISCELLANEOUS DISEASES OF THE STOMACH*

HIATAL HERNIAS

Under normal circumstances, the stomach is held in position by the gastrophrenic, gastrohepatic, gastroduodenal, and gastrosplenic ligaments. Furthermore, the phrenoesophageal membrane helps to prevent herniation through the diaphragmatic hiatus. This membrane attaches near the squamocolumnar junction and extends approximately 1 cm above this structure. During swallowing and longitudinal esophageal muscle contraction, there is “physiological” herniation of the gastric cardia through the diaphragmatic hiatus. The phrenoesophageal membrane has a recoil action to pull the squamocolumnar junction back into its normal anatomic position. When these mechanisms are defective, true herniation can develop.

Type I Hernias

The most common type of *hernia* is the *sliding, or type I, hernia,* with a prevalence of 10% to 80% depending on age (see Chapter 60). Predisposing factors include a widened hiatal tunnel and laxity of the phrenoesophageal membrane, resulting in passage of the gastric cardia into the posterior mediastinum. This herniation is often transient and asymptomatic, but larger hernias (typically defined as greater than 3 cm in length) are often nonreducing and predispose to gastroesophageal reflux.

The sliding hernia is an acquired condition, with a peak prevalence in the fifth decade of life. [1] Factors such as pregnancy, obesity, deep inspiration, vomiting, coughing, and the Valsalva maneuver that cause a positive peritoneopleural pressure gradient predispose to this type of hernia. Long-standing gastroesophageal reflux may also result in a hiatal hernia because of esophageal shortening from acid-induced tonic longitudinal muscle contraction. Age-related degeneration of the phrenoesophageal membrane likely increases vulnerability toward herniation as well. Endoscopically, a sliding hiatal hernia results in cephalad displacement of the squamocolumnar junction above the diaphragm and widening of the angle of His. [2] The cardia, identified by gastric folds, is seen extending above the diaphragmatic impression. In gastric retroflexion, one can see this herniation through the diaphragmatic hiatus because the lower esophageal sphincter has been displaced (Fig. 71-1; see Color Fig. 71-1A). As a result of disruption of the antireflux barrier (which is composed of the diaphragmatic crura, the phrenoesophageal membrane, and the lower esophageal sphincter in anatomic unity), the distal esophagus can also be clearly visualized while in gastric retroflexion.

FIGURE 71-1. (See Color Fig. 71-1.) (**A**) Endoscopic view of a type I hiatal hernia while in gastric retroflexion. (**B**) Appearance of a large hiatal hernia on a barium examination. (From Eisenberg RL. Diaphragmatic hernias. In: Gastrointestinal radiology: a pattern approach, 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003.)

Normally, the squamocolumnar junction is located 0.5 cm below the diaphragmatic hiatus, and the gastroesophageal junction high-pressure zone extends about 1 cm distal to this. [3] With a hiatal hernia, two high-pressure zones can be found manometrically as a result of disruption of the antireflux barrier, one corresponding to the lower esophageal sphincter and one corresponding to the diaphragmatic hiatus. The distal high-pressure zone (at the level of the diaphragm) is augmented by inspiration and abdominal compression. With regard to gastroesophageal reflux, patients with large hernias that are longer than 3 cm have weaker and shorter lower esophageal sphincters than do patients with smaller or no hernias; distal esophageal peristalsis and acid clearance are less effective as well. [4] Although esophagitis is more common in patients with a hiatal hernia of any size, it is often more severe in patients with large hernias. Not surprisingly, given the correlation between gastroesophageal reflux disease and Barrett esophagus, hiatal hernias are also associated with Barrett esophagus. [5] Gastroesophageal reflux disease and Barrett esophagus and their association with hiatal or gastric herniation are discussed in greater detail in Chapter 50 and Chapter 60.

Type II to IV Hernias

Approximately 5% of hiatal hernias are *types II to IV, the paraesophageal hernias.* [6] Unlike type I hernias, these hernias are associated with fundic herniation into the thoracic cavity (i.e., alongside the gastroesophageal junction), whereas the gastroesophageal junction may or may not be displaced as well. Although relatively uncommon, these hernias are associated with severe complications such as gastric volvulus and strangulation. These hernias frequently develop after surgical procedures in the region of the hiatus, such as fundoplication and splenectomy. In an era when laparoscopic Nissen fundoplications are becoming more prevalent, it is important to realize that postoperative paraesophageal hernias develop in 1% to 6% of cases. [7], [8], [9] and [10]

In type II hernias, there is a localized phrenoesophageal membrane defect allowing the gastric fundus to become the lead point of a herniation. The gastroesophageal junction is still in proper anatomic attachment to preaortic fascia and the phrenoesophageal membrane (Fig. 71-2 and Fig. 71-3; see also Color Fig. 71-2). As more fundus herniates into the thoracic cavity, in part because of gastrocolic and gastrosplenic ligament laxity, gastric rotation can develop along the longitudinal axis of the stomach. This organoaxial volvulus (see section “ Gastric Volvulus”) results in the greater curve lying anterior to the lesser curve. Type II hernias can also result in a mesenteroaxial volvulus in which rotation is along the transverse axis, but this is much less common. Type III hernias are a combination of types I and II; the fundus herniates through the hiatus, but stretching of the phrenoesophageal membrane results in displacement of the gastroesophageal junction above the diaphragm as well (Fig. 71-4). Type IV hernias are rare and require a massive hiatal defect that results in herniation of abdominal organs (colon, small bowel, spleen, and pancreas) into the thoracic cavity.

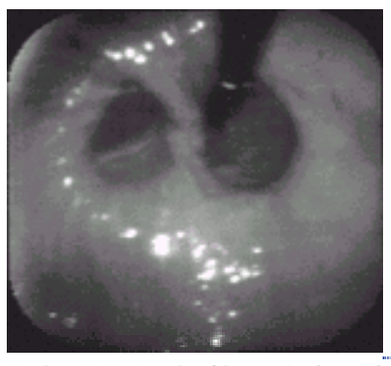

FIGURE 71-2. (See Color Fig. 71-2.) Endoscopic view of a paraesophageal hernia while in gastric retroflexion. The fundus is herniating into the thoracic cavity alongside a concomitant type I hernia.

FIGURE 71-3. Radiographic view of a type II paraesophageal hernia. The *arrow* denotes the gastroesophageal junction, which remains below the diaphragm. There is an associated gastric volvulus with a portion of the stomach within the hernia sac. (From Eisenberg RL. Diaphragmatic hernias. In: Gastrointestinal radiology: a pattern approach, 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003.)

FIGURE 71-4. A type III, or mixed, paraesophageal hernia in which the gastroesophageal junction is markedly elevated because of gastric herniation. The leading gastric edge has also herniated through the phrenoesophageal membrane and hence is the paraesophageal component of the hernia. (From ref. [11].)

Common symptoms of the paraesophageal hernias include epigastric and substernal pain, postprandial fullness, nausea, pyrosis, dysphagia, and nonproductive retching. [11] Acute bleeding from gastritis or ulceration is rare, but up to one third of these patients may have iron deficiency anemia resulting from erosions (Cameron ulcers) or venous dilation and engorgement from hiatal constriction and ischemia. [12], [13] Similar to type I hernias, these hernias are also associated with a hypotensive lower esophageal sphincter, shortened intra-abdominal lower esophageal sphincter length, distal esophageal dysmotility, and impaired acid clearance. [14], [15] Because of the vague nature of many complaints, the true prevalence of paraesophageal hernias is unclear. Nonetheless, up to 20% of such patients may present with acute gastric strangulation or perforation, with associated mortality rates of 50%. [16], [17]

As a general rule, surgical repair of paraesophageal hernias is indicated only in patients with symptoms. [14], [18] Perhaps one exception to this rule is the immediate repair of paraesophageal hernias that develop after routine fundoplication. Many authors recommend obtaining routine barium esophagrams in the first week after fundoplication to rule out the development of a hernia that is more easily repaired in the immediate postoperative period, even in asymptomatic patients. [8], [10] This approach is not unreasonable because these patients are considered to have a failed surgical procedure with persistent gastroesophageal junction derangements and a high likelihood of recurrent reflux symptoms at some point. The risks associated with paraesophageal hernia repair include bleeding, pneumothorax, vagal injury, early satiety and bloating, diarrhea, and nausea. Up to 20% of patients will have dysphagia initially, and 6% will have chronic dysphagia. [14], [19] In addition to hernia reduction and repair of the diaphragmatic defect, fundoplication is routinely performed, given the gastroesophageal junction abnormalities associated with paraesophageal hernias. Swanstrom and colleagues [19] followed-up 52 patients after surgery for paraesophageal hernias, most of whom had Nissen fundoplications rather than Toupet or Collis-Nissen procedures. Preoperatively, 59% of these patients had pyrosis, 50% had dysphagia, and 26% had esophageal body dysmotility. Postoperatively, pyrosis was reduced to 10%, dysphagia to 6%, and esophageal dysmotility to 13% after a mean follow-up of 18 months. In addition, more than 60% of patients had improved lower esophageal sphincter pressure after surgery, but 8% developed recurrent herniation. Laparoscopic surgery is feasible in most patients; conversion to an open procedure is necessary about 5% of the time. [20] Although there are no prospective trials with long-term efficacy data that compare open with laparoscopic repair of paraesophageal hernias, one study showed that the laparoscopic approach is associated with decreased operative blood loss, decreased rates of ileus, and decreased intensive care and total hospital stays. [21]

GASTRIC VOLVULUS

A *volvulus of the stomach* occurs when one portion of the stomach twists around another. If the twist occurs around an imaginary line between the pylorus and the gastroesophageal junction, it is an *organoaxial volvulus* (Fig. 71-5). Typically, the greater curve spins upward such that the stomach appears “upside-down,” with the true posterior wall lying anteriorly. The antrum rotates anteriorly and superiorly, whereas the fundus is displaced posteriorly and inferiorly. Alternatively, a *mesenteroaxial volvulus* develops when the distal stomach twists around an imaginary line between the center of the greater curve and the porta hepatis (Fig. 71-6). The antrum and the distal body twist to the right (anteriorly and superiorly) such that the posterior wall again becomes anterior in placement; occasionally, the antrum

rotates posteriorly in a mesenteroaxial volvulus, though this is rare. Approximately 60% of cases are organoaxial, 30% are mesenteroaxial, and the remainder are either mixed or unclassified. [22] There is no clear prognostic value in defining the type of volvulus, although defining the degree of the volvulus is important. [23] A volvulus may be partial, with no symptoms and spontaneous resolution, but a complete volvulus can result in vascular compromise, ischemia, and gastric infarction.

FIGURE 71-5. (**A**) Schematic representation of an organoaxial volvulus. (**B**) Large organoaxial volvulus with much of the stomach trapped within a hiatal hernia. (*A,* From Sandler RS, Todisco A. Miscellaneous diseases of the stomach. In: Yamada T, ed. Textbook of gastroenterology, 3rd ed. Philadelphia: Lippincott Williams & Wilkins, 1999; *B,* from Eisenberg RL. Diaphragmatic hernias. In: Gastrointestinal radiology: a pattern approach, 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003.)

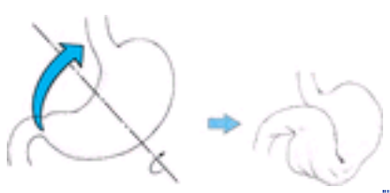

FIGURE 71-6. Schematic representation of a mesenteroaxial volvulus. (From Sandler RS, Todisco A. Miscellaneous diseases of the stomach. In: Yamada T, ed. Textbook of gastroenterology, 3rd ed. Philadelphia: Lippincott Williams & Wilkins, 1999.)

The peak incidence of gastric volvulus is in the fifth decade, with no clear gender predomination, but 20% of cases may occur in children, usually in the first year of life. [22], [24] Predisposing factors include paraesophageal hernias, abnormal ligament fixation (of the gastrophrenic, gastrohepatic, gastrosplenic, gastroduodenal, and gastrocolic ligaments), prior trauma, and phrenic nerve injury. [23], [25], [26] Abdominal surgery is another risk factor, in large part because of adhesions, but specific procedures such as Nissen fundoplications and gastrostomies are also frequently associated with gastric volvulus. [27], [28] and [29] In children, ligamentous laxity, splenic mobility, congenital diaphragmatic defects, and hiatal hernias are predisposing factors. [24]

Acute Gastric Volvulus

The acute presentation of gastric volvulus includes substernal or epigastric pain that frequently radiates to the neck and arms and may be confused with a myocardial infarction. Patients may have minimally productive emesis or dry heaves. Hematemesis is rare but suggests mucosal ischemia or tears. The upper abdomen may become tense and distended, whereas the lower abdomen is soft. Tympany is not uncommon, and abdominal tenderness may not be very impressive. Organoaxial rotations are much more likely than mesenteroaxial rotations to cause an acute volvulus. If the volvulus is complete, it may not be possible to place a nasogastric tube. An acute and complete volvulus may be accompanied by the Borchardt triad: abdominal pain, violent retching, and the inability to place a nasogastric tube. [30] In organoaxial volvulus, vascular compromise and gastric infarction occurs in 5% to 25% of cases, and there is a 15% to 20% overall mortality rate with acute volvulus. [27] In cases of intrathoracic volvulus resulting from a hiatal hernia, mortality rates from emergency surgery are 40% but approach 60% if there is gastric strangulation. [25], [31]

The diagnosis is suggested by the presence of a large, gas-filled viscus in the chest or upper abdomen on routine radiography with the appropriate clinical symptoms. Plain films, however, may be difficult to interpret in a patient with an organoaxial volvulus because the stomach still lies in a horizontal position with a single air-fluid level. A nasogastric tube should be placed if possible, although this is unlikely to untwist the volvulus. Although surgery is still considered standard therapy, some have proposed endoscopic reduction as a temporary maneuver in patients in whom vascular compromise is not suspected. There are case reports of volvulus reduction in which the endoscope tip is placed just beyond the torsion, the dials are locked, and the scope is rotated 180° to untwist the volvulus. [32], [33] and [34] Eventually, if not on an emergency basis, surgery should be pursued to untwist and repair the volvulus with either anterior gastropexy (in which the greater curve is fixed to the anterior abdominal wall) or tube gastrostomy. A subtotal or even complete gastrectomy may be necessary in cases of necrosis. Anatomic defects that may have predisposed to the volvulus may be repaired at the same time. Laparoscopic surgery may be a viable option in many patients, although this approach may be a better option in patients with chronic volvulus. [35], [36] Given the risk of ischemic strangulation, perforation, and death with acute volvulus, immediate surgical intervention should probably remain the initial treatment of choice, especially because endoscopic reduction does not prevent volvulus recurrence. [22]

Chronic Gastric Volvulus

Unlike acute volvulus, chronic volvulus is far more likely to involve a mesenteroaxial rotation. Because this volvulus is usually incomplete and intermittent, patients are more likely to present with chronic and vague symptoms. Abdominal discomfort, dysphagia, pyrosis, fullness, bloating, and postprandial borborygmi are the primary symptoms of chronic volvulus. Such symptoms are usually worsened after a large meal, and patients may report that changes in body position or self-induced vomiting help to alleviate the symptoms. [23] The diagnosis is often made incidentally on a chest radiograph or barium study. In a mesenteroaxial volvulus, plain films demonstrate a spherical stomach with two air-fluid levels in the upright position; the upper level reflects the antrum, whereas the lower level reflects the fundus. The mortality associated with chronic gastric volvulus is only 0% to 13%. [23], [37] Although surgery is again considered the mainstay of treatment for chronic and relapsing volvulus, the endoscopic reduction of chronic gastric volvulus and the placement of percutaneous gastrostomy tubes (in the body and antrum) have been reported in patients who are poor surgical candidates. [32], [33] and [34], [38], [39] The need to pursue surgical or endoscopic procedures is unclear in patients with an asymptomatic volvulus or in those with only mild symptoms attributable to the volvulus. In such cases, the risks of surgery or endoscopy must be weighed against the risk of an untreated volvulus.

GASTRIC RUPTURE

Nontraumatic tears of the gastric wall are relatively rare, but the true incidence is not well defined. In one autopsy study, 0.12% of more than 5100 patients were found to have a spontaneous *gastric rupture.* [40] The age group is generally the fourth decade of life, and women are more commonly affected. [41] Gastric ruptures may be as large as 15 cm, and the affected muscle may demonstrate edema and hemorrhage without an inflammatory infiltrate. [42] Initially, there is a mucosal tear, followed by dissection of the deeper muscle layers and serosa.

Numerous conditions may result in gastric rupture. Intragastric pressure and overdistention may play a role in many ruptures, especially if there are pyloric or

gastroesophageal junction obstructions to prevent gastric decompression. Normally, the gastric musculature will relax in response to distention, and it has been suggested that rupture does not occur from distention until the intragastric volume is about 4 L. [42], [43] Mechanical outlet obstruction, preventing decompression at either the gastroesophageal junction or the pylorus, may develop from tumors of the esophagus, pancreas, distal stomach, or duodenum, from complicated peptic ulcer disease, from congenital duodenal obstruction, or from fundoplication. [44], [45] and [46] There can also be functional obstruction at either gastric orifice. For example, increased intragastric pressure will normally result in a tightened lower esophageal sphincter, as well as an accentuated angulation of the esophagus against the right diaphragmatic crus, both helping to prevent gastroesophageal reflux and gastric decompression. [41], [45], [47] Similarly, a distended stomach may cause angulation and obstruction at the pylorus. [48] Other causes of gastric distention and rupture include gastric lavage in the setting of balloon tamponade for bleeding varices, mouth-to-mouth resuscitation with external cardiac compressions, aerophagia, oxygen administration from nasal cannulation, gastric insufflation during general anesthesia, and fermentation of retained gastric food with associated gas production. [45], [49], [50], [51], [52] and [53] Rapid decompression in diving accidents has also resulted in profound gastric distention and rupture. [54], [55] and [56] Finally, excessive use of sodium bicarbonate for indigestion in the setting of existing overdistention has been reported to cause rupture, although large volumes of sodium bicarbonate would be necessary to cause significant carbon dioxide gas production. [43], [57], [58]

Trauma and vomiting can also result in gastric rupture. Surprisingly, gastric rupture from blunt abdominal trauma occurs in fewer than 2% of trauma cases, in part because the stomach is somewhat protected by the ribs and liver. [59] Examples of trauma-induced ruptures include coughing, grand mal seizures, exertion during heavy lifting, the Heimlich maneuver, and motor vehicle accidents with seat-belt compression of the stomach on the vertebrae. [50], [60], [61], [62], [63], [64], [65] and [66] Endoscopic procedures, especially those involving cautery or laser therapies, can result in traumatic rupture. Violent vomiting that results in a gastric tear may be seen in peripartum and postpartum emesis, in retching from pyloric stenosis, and in vomiting induced by ipecac. [67], [68] and [69]

In cases of gastric distention, 60% of ruptures occur along the lesser curve near the cardia or at the fundus. [42] It is much less common for such ruptures to occur along the greater curve, on the anterior or posterior walls, or near the pylorus. Conversely, ruptures induced by vomiting typically occur on the greater curve and fundus because violent emesis results in herniation of the stomach into the chest. [70] Symptom onset is immediate, and patients frequently report a history of recent overeating and emesis. [42] Severe pain, abdominal distention, and dyspnea are typical. Shock may develop from peritonitis, blood loss, and tension pneumoperitoneum (causing inferior vena cava compression with reduced blood return to the heart and hypotension). [48] Subcutaneous emphysema, in which abdominal free air traverses the retroperitoneum and mediastinum to the neck, has also been reported. [47] These symptoms and findings can mimic other intra-abdominal catastrophes such as ulcer perforation, pancreatitis, appendicitis, neoplastic perforation, and splenic rupture. Plain radiographs typically demonstrate the pneumoperitoneum. Other imaging modalities are of questionable benefit because the clinical situation typically necessitates immediate surgical attention.

Gastric rupture carries a 100% mortality rate without intervention. [41] Death is caused by sepsis, bleeding, respiratory failure, and air embolism. [48] Mediastinitis may also develop with similar end results. [60] With surgical intervention, mortality rates still exceed 60%. Larger tears, reflective of increased peritoneal contamination, contribute to the high mortality. Perforated ulcers typically have a small tear of less than 2 cm and a better outcome, whereas gastric ruptures usually measure 5 cm or greater and have a poorer outcome. [57] If abdominal pressure is excessive and compromises the patient's cardiorespiratory status, a decompressive paracentesis may be necessary before surgical correction. [49] Surgery consists of resection of necrotic gastric tissue, correction of any predisposing obstructive lesions, and peritoneal lavage. Broad-spectrum antibiotics against aerobic and anaerobic organisms are warranted perioperatively.

FOREIGN BODIES

Foreign objects are frequently swallowed, either intentionally or accidentally, and approximately 1500 persons die within the United States each year from either swallowing or aspirating foreign bodies [71] (see Chapter 63). In children, small toys are common, but coins clearly account for most accidental ingestions. [72] In adults, items may be accidentally swallowed as a result carelessness, poor vision, or alcohol intoxication. [73], [74] Such items may be articles that are frequently placed in the mouth, such as toothbrushes, dental prostheses, or nails. Patients with dementia or psychological disease are most likely to ingest a foreign body intentionally, but patients with bulimia may inadvertently swallow objects while trying to induce emesis. [75], [76] and [77] Incarcerated criminals may swallow objects for secondary gain (hospitalization), whereas drug traffickers may be "body-packers" who intentionally ingest small packets of illicit drugs. [73], [78]

Most foreign bodies traverse the stomach without causing gastric symptoms; symptoms induced by a gastric foreign body suggest mucosal penetration or perforation, peritonitis, or obstruction. [73], [75] Mucosal tears, ulceration, perforation, abscess formation, hemorrhage, and fistula formation may all develop as a result of a retained foreign object. [73], [74] Sharp objects such as splinters or animal bones are more likely to induce a perforation. When to remove foreign bodies from the stomach is a clinical uncertainty. Objects more than 2 cm in diameter or 5 cm in length should strongly be considered for removal because such items may lead to duodenal obstruction. [79] Owing to the risk of perforation, sharp objects should also be removed promptly. If a sharp object is too large reasonably to retrieve endoscopically, a laparoscopic gastrostomy or other surgical method may be necessary. [80] Blunt objects that are less than 2 cm in diameter or 5 cm in length may be followed with serial radiographs. If the object remains in the stomach for 3 weeks or if the patient develops symptoms of mucosal irritation, endoscopic retrieval of the foreign body should be pursued.

If endoscopic removal is attempted, the patient should be placed in the left lateral decubitus position, and glucagon may be administered to limit motility (although this is more likely to be beneficial for objects in the duodenum rather than in the stomach). Although there is some risk of perforation, a protective overtube may be placed in the esophagus for sharp or difficult-to-grab objects, to prevent the possibility of aspiration or esophagopharyngeal trauma. [81], [82] Standard biopsy forceps or snares may be used, but larger forceps are available specifically for foreign body removal. Oval or round objects may be trapped within a basket or net. Surgery should be considered before endoscopy for large, jagged objects and for "body-packers," who have a significant risk of death from the endoscopic rupture of a packet containing illicit drugs.

The accidental ingestion of button batteries deserves special mention. Such batteries contain alkaline compounds such as sodium or potassium hydroxide. Damage to the stomach may occur as a result of low-voltage burns, pressure necrosis, or direct corrosion. [72] Batteries lodged within the esophagus should be removed immediately by endoscopy. If the plain radiograph demonstrates the battery within the stomach and the patient is asymptomatic, the patient can be followed with serial radiographs. Endoscopic removal is warranted if the battery remains in the stomach at 48 hours, if it is greater than 15 mm in diameter, if it is a mercury-based battery, or if the patient is symptomatic with localized abdominal pain, hematemesis, or melena. [83] Ipecac syrup should never be administered in cases of battery ingestion because it has not been shown to be effective and may result in esophageal impaction of the battery. [83] If there is battery disruption, heavy metal levels (depending on the type of battery) in the blood and urine should be measured.

CAUSTIC INGESTION INJURY

Often more dangerous and damaging than foreign bodies is the ingestion of *caustic materials* (see Chapter 63). Common household cleansers and chemicals may contain compounds such as sodium or potassium hydroxide, sodium carbonate, aluminum hydroxide, and bleaches such as hydrogen peroxide. Acids tend to cause immediate pain and are quickly expelled by the patient, even in the setting of an intentional ingestion. Alkali agents are often odorless and tasteless, leading to more significant ingestions and more damage from their solvent action on lipoproteins. [84] Acute manifestations of a caustic ingestion include persistent salivation, odynophagia or even a complete inability to swallow, and the development of palatal or pharyngeal ulcers and white plaques. [85], [86] Hoarseness, stridor, and hematemesis may also develop. The presence of peritonitis or mediastinitis suggests a perforation. Acute and chronic manifestations of caustic ingestion are more commonly seen in the oropharynx and esophagus, but gastric damage may result in late manifestations such as early satiety, weight loss, and vomiting (symptoms of a possible outlet obstruction). A reasonable approach to the patient with caustic ingestion should begin with abdominal and chest radiographs.

These radiographs are most useful to exclude the presence of free air or esophageal and gastric distention, which are commonly seen after caustic ingestions. If there is no free air but perforation is still suspected, barium studies or a computed tomography scan can be pursued. The patient may be sent home if there is no free air on plain films, if there are no symptoms, and if the ingestion was accidental. Obviously, admission and surgical consultation are warranted if there is evidence of perforation. If the patient has respiratory or gastrointestinal symptoms or if the ingestion was intentional, hospital admission is necessary. The patient should ingest nothing by mouth initially, and psychiatric consultation should be considered. If the patient has significant respiratory complaints, laryngoscopy may be justified to determine the need for intubation or tracheostomy. Endoscopy is warranted in all cases, but the timing of an endoscopic examination is unclear. Although some authors recommend immediate endoscopy, most recommend an examination at 24 to 72 hours to define the extent of injury better. [85], [86] An endoscopic grading system has been proposed: grade I, erythema and edema; grade II, hemorrhage, erosions, blisters, and superficial ulcers (A, linear, B, circumferential); grade III, multiple deep brown or black or gray ulcers; and grade IV, full perforation. For grade IA damage, liquids can be started with diet advancement as tolerated. Patients

with grade IIB or III damage should have a nasogastric tube placed for gastric decompression, and acid suppression should be employed. Oral liquids may be started approximately 48 hours after endoscopy if the patient is swallowing well. Immediate surgery is warranted in patients with grade IV damage.

Acid neutralizers have been used to limit caustic damage in the past, but these are no longer recommended because heat production from the neutralization process has been shown to worsen tissue injury. [87] Owing to the risk of esophageal strictures and antral stenosis, some investigators have proposed oral corticosteroids, based on animal or anecdotal data only. The only randomized controlled trial performed suggested that steroids did little to prevent or limit esophageal strictures, and routine steroid use is not recommended by all clinicians [88] (see Chapter 63).

In the long-term, caustic gastric injury may lead to antral stenosis within 3 to 6 weeks of the ingestion, or it may be delayed for many years. It is not unreasonable to perform an upper gastrointestinal series at 3 to 6 weeks to look for this complication. Some cases of antral or pyloric stenosis may respond well to endoscopic balloon dilation, but most patients will eventually require a surgical pyloroplasty or distal gastrectomy. In patients with prior caustic injury who are undergoing endoscopy for other reasons, antral hyalinization has been reported and may mimic gastric Crohn's disease or malignant disease. [89] Corrosive damage may increase the risk of subsequent gastric cancer, but such reports are isolated. Endoscopic surveillance is not warranted, based on the available literature. [90]

GASTRIC BEZOARS

Bezoars are concretions of foreign material that are retained most frequently within the stomach but have also been found in the esophagus and rectum. [86] Such matter may include plant and vegetable debris (*phytobezoar*), hair (t *richobezoar*), medications (*pharmacobezoar*), and persimmons (*disopyrobezoar*). The true prevalence of bezoars is uncertain because many patients are asymptomatic, but bezoars are uncommon in the intact stomach; that is, they have a prevalence at endoscopy of 0.4%. However, 10% to 25% of patients with prior antrectomies have been found to have symptomatic bezoars. [91], [92]

The formation of bezoars is likely multifactorial. Altered gastric motility and emptying are the primary causes, but size and digestibility of swallowed material are factors as well. Inadequate mastication, missing teeth, and poorly-fitting dentures may also contribute to bezoar formation. [93], [94] Prior gastric surgery, whether it be a pyloroplasty, antrectomy, or partial gastrectomy, clearly places patients at risk for phytobezoar or fungus ball formation. [95] Surgery can induce gastric atony and dysmotility, but outlet obstruction from stenosis and decreased secretion of acid and pepsin may also play a role in bezoar formation. Furthermore, many postsurgical patients develop chronic gastropathy that leads to increased mucus production, and this mucus may act like cement for developing concretions. [96] In addition to surgery, gastric stasis and bezoars have been linked to diabetic gastroparesis, mixed connective tissue disease, hypothyroidism, and myotonic dystrophy. [97], [98], [99] and [100]

Phytobezoars are commonly composed of apples, grapes, oranges, cherries, raisins, bran, oats, cabbage, potato peels, peanuts, and celery. [93], [101] Bezoars may also be composed of nonfood items such as plastic, paper, string, or Styrofoam. Disopyrobezoars develop when unripe persimmon material comes into contact with gastric acid; a tannin in the fruit forms a coagulum that acts as a base for bezoar formation. Trichobezoars may be quite large, depending on the amount of hair consumed. Medications have reportedly been trapped within hair fibers, and drug toxicity has been attributed to altered drug metabolism. [102] Trichobezoars may contain food material and fat as well. *Fungus balls,* also termed yeast bezoars, are associated with prior gastric surgery and frequently resolve without therapy. For symptomatic fungus balls, gastric lavage, antimycotics, and gastric outlet reconstruction have all been used. [103], [104]

Concretions are a type of bezoar that are typically very hard. Shellac, furniture polish, and concrete are classic components of such a bezoar, and surgery may be necessary to remove concretions because other therapies are generally ineffective. Pharmacobezoars may develop in patients with normal or abnormal gastric physiology. Aluminum hydroxide antacids have caused bezoars in patients with renal failure, and the cellulose-based coating of enteric-coated aspirin has been problematic in other patients. [105], [106] In patients with achlorhydria, calcium and magnesium carbonate preparations may not dissolve normally. Sucralfate will occasionally form concretions at normal doses and in patients with gastric outlet obstruction, so this agent should be avoided in patients with outlet obstruction or significant dysmotility. [107] Other drugs with capsules that do not digest (such as certain nifedipine or mesalamine preparations) should be avoided in patients with gastric outlet narrowing as well. [108]

Many patients with bezoars are asymptomatic, but epigastric pain, early satiety, nausea, and vomiting are very common manifestations of a bezoar. [86], [93] Bloating, malaise, and weight loss may also be reported. In some patients, these symptoms may persist despite bezoar resolution; in such cases, the nonspecific symptoms may be more reflective of the gastric dysmotility than of the actual presence of a bezoar. In patients at risk of bezoar formation, symptoms of small bowel obstruction may reflect the passage of a bezoar into the small bowel, although this is rare. [109] Gastric outlet obstruction and gastric bleeding (hematemesis or melena) can also be complications of bezoars. Given the nonspecific nature of the symptoms, the patient's history may suggest other causes such as peptic ulcer disease, gastric cancer, or many other intra-abdominal processes. On physical examination, potential findings include abdominal tenderness, a palpable mass, and a succussion splash. [86] Plain radiographs may demonstrate an air shadow or a mottled-appearing mass in the stomach. Barium studies are much more likely to demonstrate a bezoar; trichobezoars "absorb" barium and have a mottled appearance, whereas phytobezoars and concretions are typically impermeable to barium (Fig. 71-7). Radiographic studies may miss a bezoar, and endoscopy remains the test of choice. Although other studies such as ultrasound scanning or computed tomography may demonstrate a bezoar, endoscopy is the best for determining the type of bezoar, for excluding cancer, and for evaluating the mucosa for associated ulceration or outlet obstruction.

FIGURE 71-7. (**A**) Gastric phytobezoar on barium examination. (**B**) Glue concretion in a model airplane builder. (From Eisenberg RL. Filling defects in the gastric remnant. In: Gastrointestinal radiology: a pattern approach, 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003.)

Enzyme Therapy

Because many bezoars contain cellulose, protein, and mucus, enzymatic degradation of phytobezoars may be possible. For bezoars containing fibrous material, papain may be a useful proteolytic enzyme. Although this enzyme can be individually purchased, papain can be obtained more cheaply by using Adolph's Meat Tenderizer (1 teaspoon in 4 ounces of water before each meal). [86], [101] Some caution should be used with the latter salt-containing preparation because hypernatremia is a potential problem (1800 mg per tablespoon). Cellulase may be used for phytobezoars as well. The tablets are frequently crushed and given after each meal, or 2 L of cellulase solution may be consumed over the course of 2 days (20 g in 2 L of water). [101], [110] Biochemical supply companies can provide papain and cellulase preparations. Because many phytobezoars contain mucus as well, the mucolytic agent acetylcysteine may also be used. This agent has been reported to be effective at a dose of 15 mL in 50 mL of saline administered through a nasogastric tube two times per day for 2 days; although not specifically stated in this case report, it is presumed that a commercially available, 20% acetylcysteine preparation was used and diluted to a 5% solution as recommended by the manufacturer for other indications. [111] If there is ulceration associated with the bezoar, some investigators have suggested that mucolytic and cellulolytic agents may be safer than the proteolytic regimens provided.

Mechanical Therapy

Many bezoars contain insoluble agents that are also not amenable to enzyme therapy, and some patients are so symptomatic that more urgent therapy is warranted. Very small bezoars may be retrieved like other foreign bodies with endoscopic baskets, snares, or forceps. An esophageal overtube is frequently used to prevent inadvertent aspiration of the bezoar. [112] Similarly, many bezoars can be mechanically disrupted into small pieces with forceps such that the pieces are retrieved by suction or are allowed to pass through the pylorus. For soft and permeable bezoars, a large-bore orogastric lavage tube can be placed for aggressive water lavage

with subsequent aspiration of the fragmented bezoar. Frequently, a combination of endoscopic fragmentation and subsequent orogastric lavage is used, with immediate benefits to the patient.

Other modes of mechanical disruption have been proposed. Plastic tubing has been placed through an endoscope channel and attached externally to a Teledyne Water Pik system. Water or an enzymatic solution can be pulsated against the bezoar to allow fragmentation and subsequent gastric lavage. [101], [113] Neodymium:yttrium-aluminum-garnet laser may be useful for refractory bezoars, although multiple sessions may be necessary. [114] Extracorporeal shock wave lithotripsy and mechanical lithotripsy (with a lithotriptor for hard bezoars) have also been successful. [115], [116] In patients who have undergone gastroplasty and in whom the surgical stoma may be too small (predisposing to bezoar formation and not allowing passage of bezoar fragments), the stoma may require balloon dilation or even an endoscopic incision with a papillotome. [117], [118]

Endoscopic management is warranted only for symptomatic patients. If a bezoar is incidentally found or if symptoms are minimal, enzymatic and prokinetic therapies are likely adequate. Endoscopy is clearly indicated for more symptomatic bezoars, especially if complications such as obstruction develop.

Medical Therapy

Prokinetic agents such as metoclopramide may be used on a long-term basis to aid in the prevention and recurrence of bezoars. Metoclopramide has been used alone and with concomitant endoscopic therapy in the acute setting (40 mg intravenously over 24 hours) with some success, but patients should also be placed on a clear liquid diet under such circumstances. [119] Although liquid diets may have some benefit in bezoar dissolution, this is a slow process that is not practical as sole therapy. The prokinetic benefit of long-term metoclopramide use must be weighed against the severity of the patient's symptoms and the significant side effect profile. Domperidone, not available within the United States, may be another option, with fewer side effects. Although cisapride is currently available only on a restricted protocol basis for the treatment of diabetic gastroparesis, this agent may be an option for patients who qualify for study participation. A similar prokinetic, norcisapride, is currently under investigation and may be available in the near future without the cardiac risk and drug interactions that plague cisapride.

For patients with or at risk of the development of bezoars, prokinetic agents should be considered, based on the patient's symptom severity. Conversely, all such patients should follow basic dietary guidelines to limit bezoar formation. Specifically, smaller but more frequent meals are advisable to promote gastric emptying. Patients should chew their food adequately into small pieces. Foods that easily coalesce into a bezoar, such as persimmons, raw fruits, and stringy vegetables, should be avoided. [93]

Surgical Therapy

Surgery is rarely required for bezoars but may be necessary for complications such as perforation, gastric or small bowel obstruction, and uncontrollable hemorrhage. Symptomatic trichobezoars often require surgery more often than other bezoar types because they are very resistant to standard management techniques. Of course, other bezoars may also require surgery if they cannot be treated by the methods outlined earlier. Because surgery is unlikely to correct the underlying cause of bezoar formation and may worsen gastric motility and emptying, such a decision should not be made lightly.

HETEROTOPIC PANCREAS

Heterotopic pancreatic tissue, also termed a *pancreatic rest,* has been identified in 0.5% to 14% of persons at autopsy. [120] These rests are found in the stomach, duodenum, or jejunum in 75% of cases but may also be seen in the lung, gallbladder, common bile duct, ileum, Meckel diverticula, and numerous other locations. Typically, these rests are asymptomatic, but abdominal pain, nausea, vomiting, and rarely bleeding have been attributed to rests located in the stomach. In one series of 17 patients, pancreatic rests were the presumed cause of pain in 77% of patients, of abdominal fullness in 30%, and of melena in 24%. [121] After surgical resection of the rests, significant symptom improvement was reported, but whether rests truly do cause symptoms sufficient to warrant surgical excision is still somewhat controversial. These lesions typically appear as 2- to 4-cm submucosal masses in the prepyloric region, and central umbilication is not unusual (Fig. 71-8; see Color Fig. 71-8A). The nodules are firm and may be yellow. Because this typical appearance is not always appreciated, rests have been mistaken for malignant tumors in 67% of upper endoscopies and in 71% of barium studies. [121] If the endoscopic appearance is not diagnostic, endoscopic ultrasound may be helpful in evaluation. Surgery is indicated only in cases of symptomatic pancreatic rests.

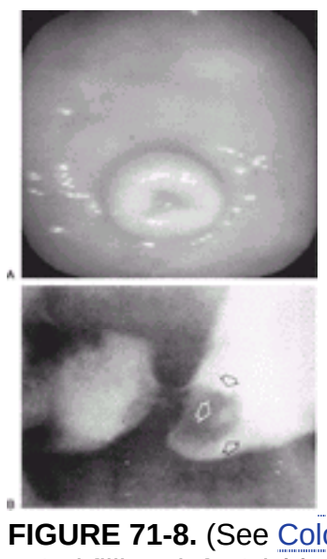

FIGURE 71-8. (See Color Fig. 71-8A.) (**A**) Pancreatic rest identified in the antrum during upper endoscopy. (**B**) Radiographic appearance of a pancreatic rest with an antral filling defect (*black arrows*); the *white arrow* denotes central filling with barium. (From Eisenberg RL. Filling defects in the stomach. In: Gastrointestinal radiology: a pattern approach, 4th ed. Philadelphia: Lippincott Williams & Wilkins, 2003.)

SQUAMOUS CARDIAC EPITHELIUM

Squamous epithelial extension into the proximal stomach has been reported in 16 patients. [122] All were male, and nearly all were white. Most patients had pyrosis (75%) and hiatal hernias (100%), and many had Barrett esophagus (38%). No patient had a history of caustic ingestion or prior gastric surgery. Most had solitary tongues of this squamous epithelium, but multiple tongues and islands were also identified. The true incidence and clinical significance of this finding are not known, but prior proximal gastric injury has been proposed as an explanation.

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* The views expressed in this article are those of the authors and do not reflect the official policy or position of the United States Air Force, Department of Defense, or the United States Government.

CHAPTER 72

Deborah C. Rubin

SMALL INTESTINE: ANATOMY AND STRUCTURAL ANOMALIES

GROSS ANATOMY

The small intestine begins at the pyloric sphincter of the stomach and extends to the cecum. Approximately 600 cm long, the small bowel is composed of three major segments: the duodenum, the jejunum, and the ileum. The junction between the duodenum and the jejunum is anatomically demarcated by the ligament of Treitz. No similar landmark distinguishes the jejunum from the ileum, but the jejunum is usually defined as the proximal two fifths of the small bowel, and the ileum as the distal three fifths.

After the appropriate rotation of the gut and its return to the abdominal cavity in fetal life, the normal position of the small bowel loops is attained. The jejunum is generally located in the mid upper and left upper quadrants, the proximal ileum in the middle abdominal region, and the distal ileum in the right lower quadrant. [1]

The first part of the duodenum, the bulb, is invested with mesentery. The descending, transverse, and ascending parts of the duodenum are retroperitoneal and partially encircle the pancreatic head (Fig. 72-1). The jejunum begins at the ligament of Treitz as the small intestine reenters the peritoneal cavity. The jejunum and the ileum are invested with mesentery. The very distal ileum may occasionally be retroperitoneally located. [2]

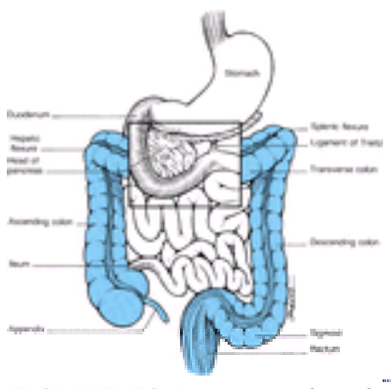

FIGURE 72-1. Anatomic relations of the stomach, small intestine, and large intestine. The duodenum encircles the head of the pancreas and is retroperitoneal. As the intestine reenters the peritoneal cavity at the ligament of Treitz, it becomes the jejunum. Generally, the jejunal bowel loops are located in the left and middle upper abdomen, the proximal ileum in the middle abdominal region, and the distal ileal loops in the right lower quadrant. **Inset:** Retroperitoneal duodenal loop and the ligament of Treitz, located behind the transverse colon.

The gross appearance of the small intestine changes from the jejunum to the ileum. The wall of the jejunum generally is thicker than that of the ileum. Also, the plicae circulares, or circular folds of mucosa and submucosa that invaginate the lumen and increase the gut surface area, are prominent in the duodenum and jejunum and disappear in the middle ileum. [1] Peyer patches usually are found on the antimesenteric border of the small intestine. They are particularly prominent in childhood and atrophy with aging. [1]

Extrinsic Arterial, Venous, and Lymphatic Supply

The arteries, veins, and lymphatics that supply the small bowel travel through the mesentery (see Chapter 131). The hepatic artery gives rise to the gastroduodenal artery, which branches into the anterior and posterior superior pancreaticoduodenal arteries. These anastomose around the duodenum and communicate with the inferior pancreaticoduodenal artery, which arises from the superior mesenteric artery. The superior mesenteric artery also supplies the jejunum and the ileum through a series of branches that form numerous arcades in the mesentery and then penetrate the intestine.

The superior mesenteric vein serves as the major venous conduit and joins the splenic vein to empty into the portal vein. Small lymph channels or villus lacteals drain into mesenteric lymph nodes located near the intestine and along the superior mesenteric and celiac arteries. These drain into the cisterna chyli and the thoracic duct.

Neural Supply

The intestine contains an abundant, complex intrinsic neural supply that coordinates motor activities and consists of myenteric (Auerbach) and submucosal (Meissner) plexuses (see Chapter 2). The extrinsic autonomic innervation of the small intestine consists of components from the parasympathetic and sympathetic systems. [3] Postganglionic fibers arising from the superior mesenteric ganglion supply sympathetic motor innervation. These synapse with preganglionic fibers from the spinal cord in the region of the tenth and eleventh thoracic roots and travel in the lesser splanchnic nerve. Adrenergic neurons innervate both the Auerbach and the Meissner plexuses. Sensory nerves arise from the dorsal root ganglia. The parasympathetic motor innervation consists of preganglionic nerve fibers that arise from the vagus nerve. Vagal fibers originate from the dorsal motor nucleus and then divide into esophageal, anterior, and posterior vagal trunks. Both the anterior and posterior trunks give rise to celiac branches that directly innervate the small bowel by communicating with the intrinsic nervous system.

The axons of a small number of intrinsic nerves project from the intestine to travel with extrinsic nerves and synapse with sympathetic postganglionic neurons at the sympathetic prevertebral ganglia.

Enhancement of Small Intestinal Surface Area: Gross and Microscopic Features

The surface area of the small intestine is enhanced by three features that are peculiar to the gut: the plicae circulares, the villi, and the microvilli (Fig. 72-2). [2] The plicae circulares, or circular folds, consist of visible mucosal/submucosal invaginations that are predominantly located in the duodenum and jejunum. The villi, fingerlike projections that protrude into the intestinal lumen, are approximately 0.5 to 1.5 mm long and cover the mucosal surface. They can be viewed by close inspection of the mucosa under low-power microscopy or as tiny mucosal protrusions at endoscopy. They consist of a layer of epithelial cells overlying the lamina propria. Their microscopic appearance varies; duodenal villi are characteristically broad and leaf-shaped, jejunal villi are tall and thin, and ileal villi are short and broad. The length and shape of the villi also vary with the geographic region; in some underdeveloped regions of the world, villi tend to be shorter than in the United States. [4], [5] At the base of the villi, the epithelium enters the lamina propria and forms the crypts of Lieberkühn (Fig. 72-3 and Fig. 72-4), which extend almost to the muscularis mucosae. The microvilli are sub–light microscopic tubular projections that are extensions of the apical cell membrane and compose the brush border. This complex membranous network contains the enzymes, receptors, and carriers required for terminal digestion and absorption.

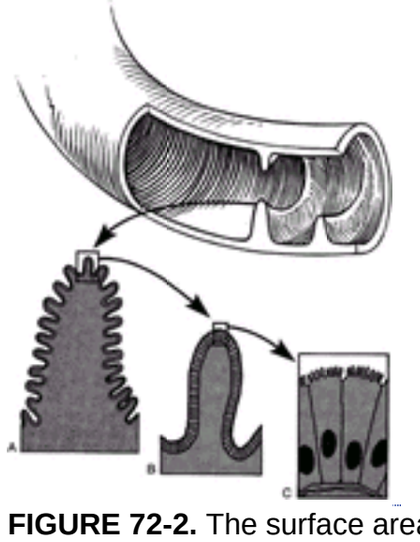

FIGURE 72-2. The surface area of the small intestine is enhanced by three mechanisms: the valvulae conniventes (**A**), the villi (**B**), and the microvilli (**C**). (Adapted from Weaver LT. Anatomy and embryology. In: Walker WA, ed. Pediatric gastrointestinal disease: pathophysiology, diagnosis and management. Philadelphia: BC Decker, 1991:195.)

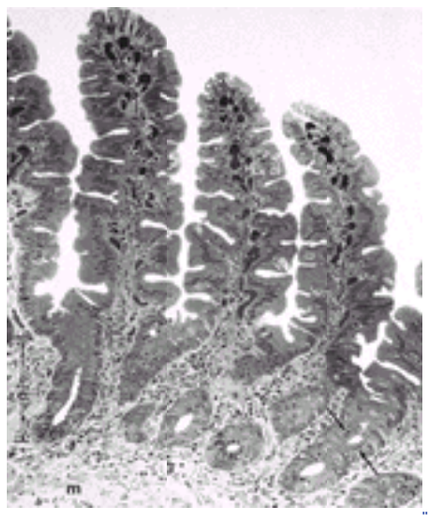

FIGURE 72-3. This semi-thin 1-µm epon section of human jejunum, stained with toluidine blue and viewed by light microscopy, depicts the mucosa. The villi are formed by a continuous sheet of columnar epithelial cells and underlying connective tissue, the lamina propria. The epithelial sheets are composed primarily of enterocytes; mucus-secreting goblet cells are also present but are much less abundant, as are rare enteroendocrine cells. At the base of the villi, the epithelial cells descend into the lamina propria to form the crypts, one of which is denoted at the left (*c*). The crypts are lined mainly by undifferentiated crypt cells, which can be observed in mitosis (*arrows*). Differentiation proceeds as these cells migrate up the villus. Paneth cells at the bases of the crypts are recognized by their dark granules. The muscularis mucosae (*m*), a thin layer of smooth muscle, separates the mucosa from the submucosa below. Original magnification ×150. (From Rubin W. The epithelial “membrane” of the small intestine. Am J Clin Nutr 1971;24:45.)

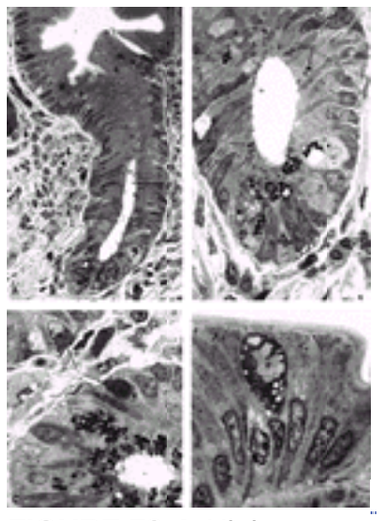

FIGURE 72-4. High-power views of the crypt. **A:** A crypt is observed extending to the base of a villus. Note the prominent brush border on the differentiated cells above the villus base and the inapparent brush border on the crypt cells below. The *arrow* shows a short and irregular brush border on an upper crypt cell. Large, dense Paneth cell granules are evident at the crypt base; the smaller apical granules higher in the crypt are in undifferentiated crypt cells. Original magnification ×300. **B:** The large granules in Paneth cells at the base of another crypt are located mainly in the apical cytoplasm. The scantier, lighter apical granules in cells above (*arrows*) are in undifferentiated crypt cells. Original magnification ×700. **C:** The large, dense apical granules in Paneth cells at another crypt base should be contrasted with the smaller, barely resolved basal infranuclear granules observed in two endocrine cells (*arrows*). Original magnification ×700. **D:** Differentiated villus epithelial cells and a goblet cell. Note the tall, prominent brush border. The narrow, light zone below the brush border represents the terminal web. The numerous light structures within the cells are mitochondria. The occasional dense apical structures are lysosome derivatives (*arrowheads*). The apical densities between adjoining cells at the level of the terminal web (*arrows*) are the terminal bars. They represent the junctional complexes of electron microscopy—the tight junctions, intermediate

junctions, and desmosomes; these are specialized structures that serve to bind adjoining epithelial cells at their apices. The accumulation of mucus granules within the apical cytoplasm of the goblet cell distends it into the shape of a brandy goblet. Original magnification ×1500. (From Rubin W. The epithelial “membrane” of the small intestine. Am J Clin Nutr 1971;24:45.)

MICROSCOPIC ANATOMY

The small intestine is composed of four concentric layers: the serosa, muscularis propria, submucosa, and mucosa.

Serosa and Muscularis Propria

The thin serosal lining of the small bowel consists of mesothelial cells overlying loose connective tissue. This outer layer becomes continuous with the mesentery as it joins the small bowel; many large blood vessels course through it. Those portions of the gut that are invested with mesentery are completely covered by serosa, whereas the retroperitoneal segments are invested only on the anterior surface. The muscularis propria consists of two muscle layers: an outer, longitudinally oriented layer and an inner, circumferential layer. Between the two layers lies the myenteric or Auerbach nerve plexus, composed of intrinsic ganglion cells and nerve fibers. The muscular layers of the gut are responsible for generating coordinated peristaltic movements.

Submucosa

The submucosa is a dense connective tissue layer containing numerous arteries as well as venous and lymphatic plexuses. Typical connective tissue cells are also present, but vascular structures predominate, consistent with the role of this portion of the gut as a conduit for absorptive and digestive products. In the duodenal bulb and descending duodenum are specialized Brunner glands that secrete mucus and bicarbonate. These are branched epithelial glandular structures that fill the submucosa and are thought to be important in neutralizing stomach acid. The submucosa also contains ganglion cells and nerve fibers, termed the Meissner plexus, a collection of autonomic nerves that communicate with the Auerbach plexus. A variety of neuroendocrine substances are produced in the Meissner plexus (see Chapter 2). These plexuses interact to produce regulated, coordinated gut peristalsis.

Mucosa

The mucosal layer of the gut consists of epithelial cells overlying the lamina propria or connective tissue core and resting on a narrow layer of smooth muscle, the muscularis mucosae (see Figure 72-3).

Lamina Propria The connective tissue core of the villus (lamina propria) contains a variety of cells and vascular structures (Fig. 72-5). The immune cellular component includes lymphocytes, macrophages, granulocytes, plasma cells, and mast cells (see Chapter 7). The majority of lamina propria T lymphocytes are T-helper/inducer cells that are surface antigen $CD4^+$, although smaller numbers of T-cytotoxic/suppressor cells ($CD8^+$) are also present. [6] The intraepithelial lymphocytes that reside between villus epithelial cells are primarily $CD8^+$ and, in mice, are ?/d T-cell receptor–positive. [7] The lamina propria plasma cells produce immunoglobulin, especially dimeric immunoglobulin A (IgA), which is taken up into the intestinal epithelium and secreted intralumenally after joining with secretory component. [8] A much smaller number of plasma cells produce IgM or IgG. The maturation of the B cell to an IgA-secreting plasma cell occurs during the course of a migratory journey throughout the body’s vascular system. Cells exit the Peyer patches through lymph vessels, enter the mesenteric lymph nodes, thoracic duct, and peripheral blood, and then finally “home” to the lamina propria of the gut (as well as to other organs, such as the lung, genitourinary tract, and breast). T lymphocytes similarly migrate from one lymphoid organ to another and eventually home to mucosal sites, which are determined by their interactions with specific proteins expressed on lymphoid cells as well as the high endothelial venules, specialized vessels of the lymph nodes, Peyer patches, and appendix. [9] These vascular structures can also proliferate in organs affected by chronic inflammation. Macrophages and eosinophils are present in the normal human intestinal lamina propria. However, neutrophils are primarily found in inflamed rather than normal gut.

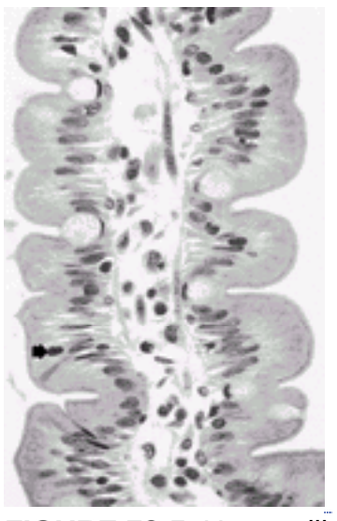

FIGURE 72-5. Upper villus region, with lamina propria, goblet cells, enterocytes, and intraepithelial lymphocytes (*arrow*). Hematoxylin and eosin stain; original magnification ×495. (From Phillips AD. The small intestinal mucosa. In: Whitehead R, ed. Gastrointestinal and oesophageal pathology. New York: Churchill Livingstone, 1989:29.)

Lymph follicles are small collections of lymphocytes in the mucosa and submucosa, scattered throughout the gastrointestinal tract. Peyer patches, found in the mucosa and submucosa, are localized collections of lymphoid follicles, may be as large as 30 cm in diameter, [10] and are most prominent in the ileum. The number of Peyer patches in the gut increases after birth. [10] The Peyer patch contains specialized epithelial cells, known as *M cells,* that overlie the lymphoid cells and allow lumenal antigens to enter the mucosa. [11] The dome area of the Peyer patch lies above the lymph follicles and just below the M cells and contains lymphocytes and macrophages. The follicular or central region contains large numbers of precursor B cells that undergo active cell division. The interfollicular zone is populated predominantly by T cells. Other cellular components of the lamina propria include fibroblasts and smooth muscle cells. Arterioles, venules, and a central lacteal function to deliver nutrients into the vascular system (see Figure 72-5).

Epithelium The villi and crypts contain a complex, rapidly proliferating, and perpetually differentiating epithelium, a continuous sheet of simple columnar cells that rests on a filamentous basal lamina or basement membrane (see Fig. 72-3 and Fig. 72-4). It constitutes the major barrier between the intestinal lumen and the lamina propria and regulates flux between these two compartments. The epithelial cells on the villi vectorially transport the products of digestion into the lamina propria, where they enter the venous capillaries or lymphatic system and are transported to other areas of the body. Tight junctions (zonula occludens) bind adjacent epithelial cells tightly together near their apices to restrict flux between adjoining cells (the paracellular route) to small ions, small molecules, and water. [12] The crypt and villus form the basic structural and functional unit of the small bowel (see Fig. 72-3 and Fig. 72-4; Fig. 72-6). Anchored stem cells located in the crypts of Lieberkühn are the source of the four major terminally differentiated cell types: absorptive enterocyte, mucus-secreting goblet cell, enteroendocrine cell, and Paneth cell. Other epithelial cell types include the tuft or caveolated cell, a rare constituent of the villus, and the M cell, a specialized epithelial cell that overlies the Peyer patch. Cellular differentiation proceeds during a complex, bidirectional migration process (see Fig. 72-6). [13] The slowly cycling gut epithelial stem cells are believed to reside in the lower crypt region, approximately four to five cell positions above the crypt base. [14], [15] They give rise to proliferating progenitor cells, which differentiate as they migrate up the crypt onto the villus to become enterocytes, goblet cells, and enteroendocrine cells. In contrast, the Paneth cells arise as their progenitors journey to the crypt base. [16] Several crypts contribute cells to a single villus. The number of crypts supplying each villus varies from the duodenum to the ileum. Epithelial cell migration and differentiation occur continuously, and the process of cellular renewal and migration to the villus tip takes approximately 5 days in humans. Apoptotic cell death occurs spontaneously near the stem cell region in the crypts, presumably to regulate stem cell numbers. Effete cells are removed from the villus tip by a process known as *anoikis,* in which altered cellular adhesion results in apoptosis. [16]

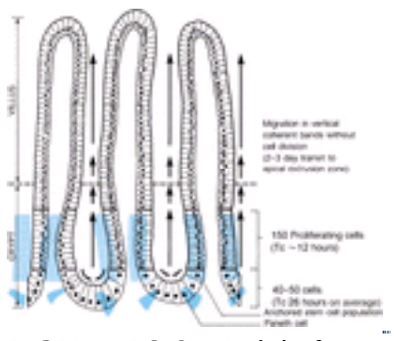

FIGURE 72-6. Model of organization of the crypt-villus axis in the adult mouse small intestine. The small intestinal crypt contains approximately 250 cells. The lower five cell positions contain 40 to 50 cells, which have an average cycle time (*Tc*) of 26 hours. This region includes Paneth cells and is postulated to include undifferentiated, anchored stem cells at the fifth cell position above the base. The undifferentiated cells divide asymmetrically to give rise to proliferating daughter cells (Tc of approximately 12 hours) that migrate up toward the villus and differentiate into enterocytes, goblet cells, and enteroendocrine cells. Paneth cells also arise from this stem cell during downward translocation to the crypt base. Senescent cells are extruded near the villus tips. (Adapted from Gordon JI. Intestinal epithelial differentiation: new insights from chimeric and transgenic mice. J Cell Biol 1989;108:1187.)

Cellular and topographic organization of the crypt. The present model of the organization of the crypt and its cellular kinetics is derived primarily from studies in the mouse. It is postulated that between 4 and 16 stem cells are present per crypt. [14], [15], [17] The number of active stem cells in any one particular crypt is unknown. [13] In the adult gut, these cells are monoclonal. [18], [19] The small intestinal epithelial stem cell presumably undergoes a process of asymmetric division to give rise to actively proliferating progenitor cells while the stem cell is maintained. [15] Proliferating progenitor cells or transit cells are located in the mid crypt region, above the presumptive stem cell region. In mouse gut, these cells enter the cell cycle every 12 hours. [20] The progenitor cells subsequently differentiate into the four major cell types.

Undifferentiated crypt cells. Undifferentiated crypt cells have many ribosomes and polysomes but scant endoplasmic reticulum, mitochondria, and microvilli and undeveloped terminal webs. They are basophilic because they contain large amounts of cytoplasmic RNA. Secretory granules are found near the apical surface (see Fig. 72-4B); their nature is unknown, but they stain with the periodic acid-Schiff (PAS) reaction and are not lysosomes. Undifferentiated crypt cells also synthesize secretory component (the receptor for IgA [21]), contain chloride channels, and are capable of secreting chloride in response to cholera toxin or other enterotoxins. [22], [23]

Absorptive enterocytes. Cellular differentiation commences during migration to the upper crypt and villus base. The cells that are fated to become the absorptive enterocytes begin to express a variety of specific gene products that enable the cells to digest and absorb many different nutrients. These include brush border enzymes (disaccharidases, peptidases, and alkaline phosphatase); proteins involved in lipid absorption (apolipoproteins and fatty acid–binding proteins); and many receptors, carriers, and transporters. [24], [25] Immunohistochemical and in situ hybridization analyses indicate that most major enterocyte genes, such as those encoding disaccharidases, apolipoproteins, fatty acid–binding proteins, and the sodium-dependent glucose transporter, are expressed as cells emerge above the crypt-villus junction, [26], [27], [28] and [29] but are not found in the crypts. This precise vertical differentiation continues as cells migrate up the villus. Microvilli become more prominent, and the capacity of the cell to absorb lipids, sugars, and amino acids increases. [30] Despite rapid cellular renewal, complex spatial differentiation in the gut is maintained from duodenum to colon. Many enterocyte genes are abundantly expressed in the proximal small bowel, but their messenger RNA (mRNA) levels decrease markedly in the distal gut, [27], [31], [32], [33] and [34] /SUP>whereas other genes are specifically expressed in the ileum. [35], [36] Cloning, sequencing, and promotor analysis of these genes have provided some insight into the molecular mechanisms underlying the regulation of regional differentiation in the gut. Transgenic mouse and cell culture transfection techniques have been used to map out regulatory promoter elements. [17], [28], [37], [38], [39] and [40] In addition, transgenic mice and mice in which embryonic stem cell mutations have been created to delete specific gene products (“knock-out” or null mice) have been used to determine the function of these novel regulatory genes, including the homeobox-containing Cdx family. [41], [42]

Ultrastructural features. As undifferentiated crypt cells travel up the villus, they acquire longer, more numerous microvilli, measuring 0.1 µm in width and about 1 µm in height, which produce a prominent striated (brush) border by light microscopy. The development of these long microvilli, fingerlike extensions of the apical cell membrane, markedly increases the absorbing surface of the small intestine (Fig. 72-7). The basolateral membrane is relatively smooth in comparison and contains the Na^{+}/K^{+} exchanger Na^{+},K^{+}-ATPase, which pumps sodium from the cell. These cells also develop a terminal web, an apical zone below the microvilli that contains many filaments and some vesicles. As cells migrate up the villus, they acquire more mitochondria and rough endoplasmic reticulum, lose their secretory granules, and acquire large apical dense bodies, which represent lysosomal derivatives (see Fig. 72-4D and Fig. 72-7). [43], [44] The functional polarity of the enterocyte, with its specific apical and basolateral membrane domains, is strictly maintained, thereby ensuring the vectorial transport of a variety of nutrients and ions from the apical to the basolateral surfaces of these cells.

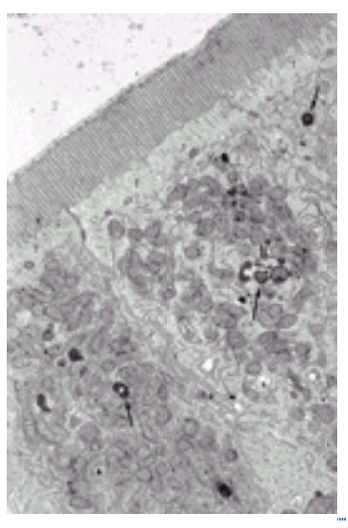

FIGURE 72-7. An electron microscopic picture of the apical halves of differentiated villus epithelial cells. Note the numerous tall microvilli covered with “fuzz” or glycocalyx and an organelle-free apical zone, the terminal web (*T*), just below the microvilli. There are abundant mitochondria and profiles of rough endoplasmic reticulum. The irregularly shaped dense bodies (*arrows*) are lysosome derivatives. Supranuclear Golgi profiles are denoted by *G*. The intercellular space is obliterated between the apices of adjoining cells just below the level of the microvilli by the tight junction (zonula occludens), an apparent fusion of adjoining cell membranes (best seen between the two cells on the left). Transport of substances from the lumen between cells (paracellular route) is restricted to water and small molecules and ions. Most products of digestion enter the cell by crossing the microvillar membrane. Original magnification ×15,000. (From Rubin W. The epithelial “membrane” of the small intestine. Am J Clin Nutr 1971;24:45.)

Filaments composed of actin extend from a dense plaque just beneath the cell membrane at the tip of the microvillus down the core of the microvillus into the terminal web. [43], [45], [46] The terminal web is a dense meshwork of filaments that contain actin, myosin, and other cytoskeletal proteins. The meshwork is oriented parallel to the surface and perpendicular to the microvillus filaments, and it is attached to the lateral cell membrane at the intermediate junction (zonula adherens). [45], [46] The numerous filaments in the microvilli and terminal web, known as the *cytoskeleton of the brush border,* probably confer a structural rigidity to the apices of the differentiated villus cells and may provide for the movement of the microvilli. [45], [47] This cytoskeleton probably also affects the uptake of certain nutrients and paracellular permeability. [45], [48] Short, thin, filamentous material extends from the outer leaflet of the cell membrane covering the microvilli into the intestinal lumen, producing a surface coat, the glycocalyx (or “fuzz”). The glycocalyx represents external extensions of proteins and glycoproteins in the microvillus membrane. [45], [49] The microvillus membrane of the differentiated villus cells contains many of the enzymes, receptors, and carriers necessary for terminal digestion and absorption, such as the disaccharidases, alkaline phosphatase, peptidases, the ileal cobalamin–intrinsic factor receptor, the bile acid receptor, and the Na^{+}-dependent glucose and amino acid transporters. [36], [45], [49], [50] and [51] The active portions of many of these are associated with the glycocalyx. The components of the microvillus membrane and glycocalyx turn over and are replenished by continuous synthesis and are transported from the rough endoplasmic reticulum via the Golgi apparatus to the microvillus membrane. [45], [49], [52], [53], [54], [55] and [56] The selective vesicular transport of microvillus constituents from the Golgi apparatus to the cell apex is dependent on microtubules. Disruption of microtubules with colchicine or vinblastine results in more random routing of these components, with the incorporation of microvillus enzymes into the basolateral surface and even the formation of basolateral microvilli. [52], [55] Some of the microvillus enzymes are finally processed in the glycocalyx by the proteolytic activity of pancreatic enzymes, especially elastase, that are adherent to the glycocalyx. [45], [49], [53]

Paneth cells. Paneth cells are pyramid-shaped cells in the crypt base that contain large apical eosinophilic secretory granules. They are much longer-lived than enterocytes, with a half-life of

approximately 20 days in the mouse. [14] A role in host defense and mucosal barrier function is suggested by their abundant expression of lysozyme, [57] cryptdins, [58] and other antibiotic proteins, and their ability to degranulate in response to live and heat-killed bacteria. [59] Several of the defensins, a family of small peptides that are abundant in human neutrophils, are highly expressed in Paneth cells. [60], [61] and [62] These 3- to 4-kd peptides are also known as *cryptdins,* for *crypt defensins,* [58] and exhibit microbicidal activity toward many different bacterial microorganisms in vitro. [58] mRNAs encoding tumor necrosis factor [63] and a $_1$-antitrypsin [64], [65] have been located in human Paneth cells. Studies in which these cells were specifically ablated in mice, with use of the cryptdin promoter to selectively express an attenuated diphtheria toxin A fragment, revealed no alteration in the differentiation of enterocytes or goblet or enteroendocrine cells and no obvious alteration in host-microbial interactions. [66] Paneth cells release defensins in response to bacteria, but not to fungi or protozoa, and the defensins have potent microbicidal activity. [67]

Enteroendocrine cells. The intestine contains a complex enteroendocrine cell population composed of a variety of paracrine-endocrine cell types. These cells were first characterized as argentaffin or argyrophil cells, depending on their reaction to silver staining in the presence or absence of a reducing agent. Electron microscopy has led to their classification by the appearance of granules. Immunohistochemistry has identified these cells based on their primary neuroendocrine product. The D cell produces somatostatin, the L cell produces glucagon-like immunoreactivity, and the enterochromaffin cell makes serotonin. This cellular population exhibits a very specific spatial distribution along the crypt-to-villus and duodenal-to-colonic axes of the gut. For example, serotonin cells are abundant throughout the gastrointestinal tract, whereas secretin and cholecystokinin cells are much more localized to the duodenum and proximal jejunum. [68], [69] and [70] In the mouse, secretin cells are predominantly located on the villi, whereas substance P–containing cells are found primarily in the crypts. Enteroendocrine cells may also simultaneously express more than one neuroendocrine product [68], [69], [70] and [71]; this coexpression is developmentally regulated in the rodent. [72] Although early data suggested that enteroendocrine cells were derived from the neural crest, later studies have indicated they arise from the undifferentiated crypt stem cell. [14], [15], [17], [73], [74] This differentiation seems to be hard-wired in the absence of lumenal contents, including pancreaticobiliary secretions and dietary factors. [71], [75] The patterns of coexpression of substance P, serotonin, and secretin along the crypt-to-villus axis of the small bowel suggests that these cells are representative of a specific differentiation pathway that arises independently of the other endocrine cell types. [69] Thus, there may be a branch point at which proliferating progenitor cells differentiate into the various endocrine cell lineages. [76] Targeted ablation of secretin cells in transgenic mice markedly decreased the numbers of L and secretin cells, also suggesting a common developmental relationship. [77] The secretions of these cells act in a paracrine manner and function locally, whereas others are true endocrine products. The peptides and amines may affect bowel motility, intestinal cellular secretion (see Chapter 4), and epithelial cell proliferation (e.g., glucagon-like peptide 2, a product of the intestinal L cell). [78], [79] In contrast to exocrine cells, which secrete apically into the lumen of an organ or the duct of a gland, endocrine cells are morphologically oriented toward the basement membrane. They exhibit an appreciable basal surface, and they narrow superiorly so that only narrow bands of apical cytoplasm reach the lumen. Their secretory granules are located predominantly in the basal cytoplasm below the nucleus, ready to be secreted by exocytosis through the basal membrane into the lamina propria (see Fig. 72-4C).

Goblet cells. The mucus-secreting goblet cells are present throughout the entire gastrointestinal tract but are more numerous in the ileum than the jejunum. These cells have the shape of a brandy goblet and apically located granules filled with mucin. Mucin is their primary secretory product and presumably serves a cytoprotective and lubricant function in the gastrointestinal tract. [80], [81] Biochemical differences among mucins are expressed in the stomach, small intestine, and colon, suggesting heterogeneity among goblet cell populations. [21], [82] The expression of specific mucins is also developmentally regulated Intestinal trefoil factor, one of a family of small peptides that includes pS2 and human spasmolysin, is abundantly expressed in goblet cells of the small intestine and colon. [83] Intestinal trefoil factor mRNA is expressed in the crypt and may therefore be a marker of early goblet cell differentiation. Embryonic stem cell gene ablation experiments demonstrate a cytoprotective role in the gut. [84] Goblet cells arise from the same committed stem cell as do the other principal cell types. [74] However, actively proliferating goblet cells have been noted in the crypt; these cells may also contribute to the villus-associated goblet cell population.

M cells. The specialized epithelial membranous, or M, cells are confined to the epithelium overlying the Peyer patch, known as the *follicle-associated epithelium*. Unlike other absorptive cells, they lack a typical brush border, demonstrating fewer, shorter microvilli and numerous apical endocytic vesicles. They serve as antigen-sampling cells and endocytose a variety of macromolecules, viruses, and bacteria from the lumen. These molecules are then rapidly transported across the epithelium and come into contact with immune cells, thereby initiating protective mucosal immune responses. [11], [85], [86] The invaginated basolateral membrane of the M cell forms an intraepithelial pocket and allows lymphocytes and macrophages to come into rapid contact with antigen. [85] Precise mechanisms involved in transepithelial transport of antigen across the M cell are still largely unknown. [86] The presumptive origin of this cell type is also the multipotent crypt stem cell. [85] M cells can be formed by culturing Peyer patch lymphocytes together with the intestinal Caco-2 cell line, or with native enterocytes. [87], [88] These and other data suggest that M cell differentiation may be immunoregulated and that lymphoepithelial cross talk may induce M-cell formation from follicle-associated epithelial cells. [89]

EMBRYOLOGY

In this section, a brief synopsis of the major events in midgut morphogenesis is presented as a basis for understanding the congenital anomalies discussed in subsequent sections (see also Chapter 23).

The primitive human gut forms when the dorsal part of the yolk sac is incorporated into the embryo at the fourth week of development, giving rise to the foregut, midgut, and hindgut. [90] The foregut is the progenitor of the esophagus, stomach, duodenum up to the biliary duct ampulla, pharynx, respiratory tract, liver, pancreas, and biliary tract. The midgut gives rise to the duodenum distal to the common bile duct, jejunum, ileum, cecum, appendix, ascending colon, and half to two thirds of the transverse colon. The rest of the colon and the superior anal canal are derived from hindgut.

The gut endoderm is the precursor of the gastrointestinal tract epithelium. Its endothelium arises from the ectoderm of the stomodeum and proctodeum as well as the endoderm. The splanchnic mesenchyme supplies the muscular and connective tissue components of the gastrointestinal tract. The midgut first freely communicates with the yolk sac and then narrows to be connected by the omphalomesenteric or vitelline duct. The primitive gut forms a U-shaped loop; this grows so rapidly in comparison with the embryo that it herniates into the umbilical cord at the sixth week of gestation. The proximal limb of the loop elongates into multiple intestinal loops, whereas the distal limb simply develops into the cecal diverticulum. The first stage of rotation is 90° counterclockwise around the superior mesenteric artery axis. At 10 weeks, the intestines return into the abdominal cavity and rotate a further 180° counterclockwise in the second stage. Finally, the cecum and appendix descend from the right upper quadrant to the right lower quadrant, and the proximal part of the colon elongates to form the hepatic flexure and ascending colon (third stage of rotation). Fixation occurs as the ascending colonic mesentery fuses with the parietal peritoneum and becomes fixed retroperitoneally. The small bowel mesentery attains a broad-based attachment to the posterior abdominal wall, extending from the duodenal-jejunal junction to the ileocecal region. The end result of this process is a normal location of the small and large intestine, diagrammed in Figure 72-1.

CONGENITAL ANOMALIES

Meckel Diverticulum

Description and Pathophysiology During early gestation, the omphalomesenteric or vitelline duct connects the fetal yolk sac to the primitive gut. By 7 to 8 weeks of gestation, this duct is normally completely obliterated. A Meckel diverticulum, the most common congenital anomaly of the gastrointestinal tract, results when this structure fails to resorb completely. The duct remnant most commonly persists as a diverticular sac; alternatively, the diverticulum may be connected to the mesentery or umbilicus by a fibrous band (Fig. 72-8). Occasionally, only a thick connective tissue band remains, attaching the gut to the umbilicus. These bands may lead to volvulus or the strangulation of bowel loops. Rarely, a fistula remains patent from the ileum to the umbilicus, leading to persistent external drainage of ileal contents. Other, more unusual duct remnants include umbilical polyps and vitelline cysts.

FIGURE 72-8. A Meckel diverticulum is attached to the umbilicus by the obliterated omphalomesenteric duct. A Meckel diverticulum may be unattached to the umbilicus or abdominal wall; alternatively, the vitelline duct may remain attached and patent, creating a fistula. Fibrous bands may predispose to volvulus and strangulation. (From Johns TNP, Wheeler JR, Johns FS. Meckel's diverticulum and Meckel's diverticular disease: a study of 154 cases. Ann Surg 1959;150:241.)

Large autopsy series indicate a 2% to 3% prevalence of Meckel diverticulum in the general population. [91] This anomaly is two to three times more common in males. [92] Meckel diverticula are true diverticula, containing all layers of the bowel from serosa to mucosa. Located on the antimesenteric border of the gut, they are most commonly found within 100 cm of the ileocecal valve but may also be in other regions of the small bowel. Most diverticula are between 1 and 10 cm in size; giant lesions may be as large as 100 cm in diameter. The two types of giant diverticula are the type I lesions, which are long but of the same caliber as the ileum, and the less common type II, or ovoid, lesions. [93] Heterotopic tissue is present in approximately 50% of all diverticula. [94] Most commonly found is gastric mucosa, pancreatic tissue, or a combination of the two. Diverticula containing colonic mucosa, Brunner glands, or jejunal or hepatobiliary tissue have also been described. The presence of heterotopic mucosa correlates with an increased risk for symptomatic, complicated Meckel diverticulum. Heterotopic gastric mucosa may lead to diverticular gastrointestinal bleeding in adults and children when ileal ulceration results from acid secretion [95], [96]; almost all bleeding diverticula contain gastric mucosa. [97], [98] Although the data are conflicting, some studies have indicated that *Helicobacter pylori* may rarely colonize the diverticular gastric mucosa and is associated with gastritis in this site. [99], [100] Heterotopic tissue is also associated with a modestly increased risk for other complications, including small bowel obstruction and diverticular inflammation. [95], [101] The risk for complications dramatically decreases with age, and most adults with a Meckel diverticulum remain asymptomatic. [102] Complications develop in approximately 2% of all adults with a Meckel diverticulum. [95], [96], [102] The complications of Meckel diverticulum include bleeding, intestinal obstruction, diverticulitis, perforation, and carcinoma. Obstruction is caused by intussusception of the diverticulum into adjacent bowel; volvulus around or herniation into a fibrous band; entrapment in inguinal, femoral, or umbilical hernia sacs (Littré hernia); or inflammation and scarring leading to blockage around the diverticular neck and adjacent ileum. The frequency of specific complications varies in adult and pediatric patients. The most common complications in children are gastrointestinal bleeding, most often occurring in infancy and early childhood (before 5 years of age), [97], [103] and intestinal obstruction. In adults, intestinal obstruction is by far the most frequent complication, and gastrointestinal bleeding is relatively uncommon. [95], [96] Whereas adults most often describe melena, children usually present with painless, multiple and rapid, bright or dark red bowel movements. More unusual complications in adults include the development of carcinomas such as carcinoids, sarcomas, and rarely adenocarcinomas. [104] Carcinoid tumors are typically small, with biologic characteristics resembling those of jejunoileal rather than appendiceal carcinoids (e.g., immunohistochemical staining and metastatic potential). [105] Patients with carcinoid of a Meckel diverticulum are generally older (in their sixth decade) and frequently asymptomatic. Tumors larger than 5 mm in diameter have considerable metastatic potential. [105] Other complications include enteroliths, which may become lodged in the diverticulum and cause abdominal pain, vomiting, bleeding, and obstruction.

Diagnosis The diagnosis of Meckel diverticulum remains a challenge. [106], [107] and [108] Sodium pertechnetate Tc 99m radionuclide scanning is particularly useful in children. The ^{99m}Tc isotope is taken up by normal stomach tissue and by ectopic gastric mucosa in the Meckel diverticulum. Surface mucus cells from the intestine as well as the stomach accumulate and secrete this anion. [109] To enhance the sensitivity of this test, cimetidine may be administered to decrease anion secretion from the gastric mucosa. [110] Pentagastrin may also be useful, enhancing anion uptake by a poorly understood mechanism. [111] The major drawback of this detection method is that heterotopic gastric mucosa must be present in the diverticulum if a true-positive result is to be obtained. In children with lower gastrointestinal bleeding from a Meckel diverticulum, this test is sensitive and specific because almost all bleeding diverticula contain gastric mucosa, but in adults, the false-positive and false-negative rates of the method are high, even in patients with bleeding. [106], [112] Crohn's disease and other inflammatory disorders lead to false-positive scans. In children, a scan for Meckel diverticulum is often followed by a technetium or stannous pyrophosphate red blood cell scan to localize the site of bleeding definitively. Small bowel follow-through is usually not useful because the diverticulum may not fill with barium and is also rapidly emptied. Enteroclysis examinations improve the sensitivity of barium studies because the contrast material under increased pressure better fills the diverticulum. Angiography can localize the source of hemorrhage and may also demonstrate the vitelline artery and its embryonic branches. This vessel arises from a distal branch of the superior mesenteric artery and ends in a characteristic blush of tortuous small vessels. Frequently, the vitelline artery involutes, and the diverticulum is directly supplied by branches from the superior mesenteric artery.

Management The treatment of Meckel diverticulum complicated by bleeding, obstruction, or perforation is surgical. Diverticulectomy is performed, possibly with concurrent ileal resection if the adjacent small bowel is ulcerated, inflamed, or obstructed. These operations can be complicated, and postoperative morbidity occurs in up to 6% of all cases. [95], [113], [114] Laparoscopic removal has been reported. [115], [116] and [117] The management of asymptomatic diverticula is controversial. [108] Because of the low risk for complications of Meckel diverticulum in adults, some have not recommended prophylactic removal. [95], [102], [118] However, others prefer to resect asymptomatic diverticula that are highly likely to cause complications, such as large (>2 cm) diverticula; lesions associated with an omphalomesenteric band, which are at risk for volvulus and obstruction; and lesions containing a palpable mass, which may represent tumor or ectopic mucosa. [98] Incidental diverticulectomy is safe and has been suggested for all patients. [113], [114], [119]

Duplications

Duplications of the gastrointestinal tract are rare, congenital cystic anomalies attached to the intestinal mesenteric border (Fig. 72-9). They may be spherical or tubular in shape. They are usually lined by gut mucosa, but like Meckel diverticula, they may contain heterotopic gastric mucosa or less commonly pancreatic, squamous, thyroid, or bronchial epithelium as well as lymphoid aggregates. [120] They share a blood supply with the associated native intestine and may also communicate lumen to lumen. Duplications may occur anywhere along the gastrointestinal tract from mouth to anus. Those of small bowel origin are most commonly found in the ileum. [120] The embryonic origin of gut duplications is unknown. Postulated mechanisms include aberrant recanalization of the gut lumen during morphogenesis, creating two attached yet distinct gut structures; abnormal notochord-midgut interactions; intrauterine ischemic events; and aborted (partial) twinning. [121], [122] Intestinal atresia may be associated with duplications, supporting a possible vascular etiology.

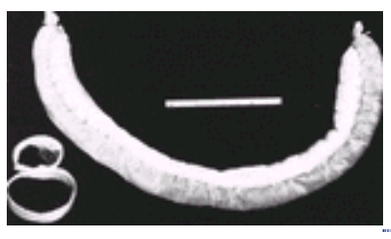

FIGURE 72-9. Tubular duplication of the terminal ileum in a 7-month-old boy. The duplication, lying on the mesenteric border of the bowel, was 29 cm long and communicated with the adjoining bowel near its distal end. The duplication, which is the smaller of the two cross sections, was lined largely by gastric mucosa, and an ulcer was within it, near its point of communication with the bowel. An island of aberrant pancreas was also in its wall. (From Arey JB, Valdes-Dapena M. Embryology and developmental disorders. In: Ming S, Goldman H, eds. Pathology of the gastrointestinal tract. Philadelphia: WB Saunders, 1992:113.)

Signs, Symptoms, and Complications In most patients, duplications are diagnosed in infancy or early childhood, but duplications are occasionally newly discovered in an adult. The frequency of symptoms varies inversely with age. Pediatric patients most commonly present with abdominal pain, obstructive symptoms (e.g., nausea, vomiting, pain), and hemorrhage. [121], [123] Obstruction may result from a mass effect, from volvulus produced by a large lesion, or more rarely from inflammation of the duplication. Much less frequently, these lesions are asymptomatic and are discovered

incidentally. In adults, the evaluation of relatively mild abdominal complaints by computed tomography (CT) or ultrasonography may lead to detection, [124] and ultrasonography has been used for prenatal detection. [125] Intussusception may be precipitated if a duplicated gut acts as a lead point and invaginates into normal intestine. Gastrointestinal hemorrhage may result from ulceration of the duplicated or surrounding mucosa; very frequently, these lesions contain ectopic gastric mucosa that secretes acid and causes ulceration. A rare complication in adults is the development of carcinoma in the duplication. [126] Carcinoid tumors, adenocarcinoma arising in ectopic gastric mucosa, and squamous cell carcinoma have been reported.

Diagnosis Small bowel duplications can be difficult to detect. Plain films may show a partially calcified wall of the duplication. Small bowel follow-through or enteroclysis barium examinations may reveal the duplication if it communicates with the gut lumen. Cysts that contain gastric mucosa may be detected by ^{99m}Tc abdominal scintigraphy, [127] which specifically delineates gastric surface mucus cells (see section " Meckel Diverticulum "). Ultrasonography (effective for prenatal diagnosis) or CT can provide clues to the diagnosis by revealing the presence of a cystic mass, and these tests are also valuable for detecting rare carcinomas, which appear as solid tissue within the cyst. [126]

Management Duplications are treated surgically. If small, the lesions are easily resected with the adjacent small bowel. If the duplicated bowel is quite extensive and resection would require the removal of too much normal bowel, the duplication is opened and the mucosa removed, with the serosa and muscular layers left intact. Alternatively, the common wall can be excised. It is important to remove all mucosa because residual ectopic gastric mucosa can lead to recurrent hemorrhage. [122]

Intestinal Atresia and Stenosis

In intestinal atresia, the lumen of a segment of the gut is totally occluded. Stenosis is a narrowing of the gut lumen that leads to partial obstruction. Atresia is one of the most common causes of bowel obstruction in neonates. [128] Duodenal atresia and stenosis are frequently associated with Down syndrome, midgut malrotation, esophageal atresia, annular pancreas, imperforate anus, and intrauterine growth retardation (see Chapter 64). [129] Jejunoileal atresia is much less frequently associated with other congenital anomalies. Atresias are most often single but may be multiple and are found from esophagus to rectum. The reported incidence of small bowel atresias varies but is approximately 1 in 3000 to 1 in 5000 live births. [130] Several types of small intestinal atresias have been described (Fig. 72-10). [131] In type I atresia, a membranous septum or diaphragm of mucosa and submucosa obstructs the lumen, but the bowel wall and mesentery are intact. In type II, two blind bowel ends are connected by a fibrous cord, with intact mesentery in between. In type IIIa lesions, two blind bowel ends are separated by a mesenteric gap, and type IIIb is the "apple peel" atresia, characterized by proximal small bowel atresia and absence of the distal superior mesenteric artery (<5% of all atresias). [132] In this case, the bowel distal to the atresia is foreshortened and coiled and receives a retrograde blood supply from the ileocolic, right colic, or inferior mesenteric artery. Finally, in type IV, multiple atresias are present throughout the small bowel, creating the appearance of a "string of sausages"; they may be of type I, II, or IIIa.

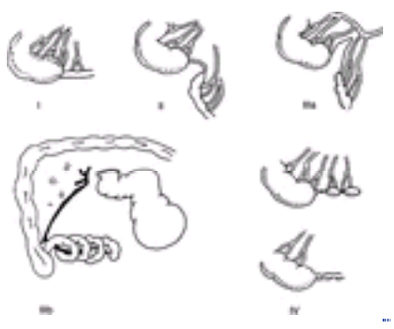

FIGURE 72-10. Classification of intestinal atresias. In *type I,* or membranous atresia, a membranous septum or diaphragm of mucosa and submucosa obstructs the lumen, but the bowel wall and mesentery are intact. In *type II,* two blind bowel ends are connected by a fibrous cord, with intact mesentery between them. In *type IIIa,* bowel lesions are separated by a mesenteric gap. *Type IIIb* is an "apple peel" atresia with features of proximal small bowel atresia, absence of the distal superior mesenteric artery, and coiled and foreshortened bowel. *Type IV* consists of multiple atresias of any type (I–IIIa). (From Smith GH, Glasson M. Intestinal atresia: factors affecting survival. Aust N Z J Surg 1989;59:151.)

Pathophysiology The pathogenesis of atresia and stenosis is unknown. Duodenal atresia is thought to be caused by a failure of recanalization of the intestinal lumen after the solid cord stage, which takes place at 4 to 8 weeks of gestation. [133] Mutations in the α_6- and β_4-integrin genes have been reported in families affected by junctional epidermolysis bullosa and duodenal or pyloric atresia. [134], [135] Ischemic intrauterine events such as midgut volvulus, arterial occlusion, intussusception, and localized volvulus secondary to meconium ileus or duplication have been implicated in the pathogenesis of small intestinal atresia, especially in single atresia and stenosis. [136] Atresia has been associated with intrauterine volvulus, gastroschisis, cardiac and other anomalies, [137] and cystic fibrosis. [137], [138] A retrospective review showed a more than 200-fold increased risk for cystic fibrosis in Caucasian infants with jejunoileal atresia. [139] Meconium ileus is thought to predispose the bowel to ischemia or cause volvulus as a consequence of hyperperistalsis. Pathological examination of atretic segments resected from neonates with multiple intestinal atresias suggests a role for either ischemia or a defect in recanalization of the gut lumen. [140] Rare familial cases of multiple intestinal atresias and apple peel (type IIIb) atresias have been reported. [128], [140], [141] An autosomal recessive inheritance pattern has been suggested, [141] but variability in the types of atresia found within families and discordance in a set of monozygotic twins suggest a more complex genetic transmission. [132] Other factors implicated in the pathogenesis of atresia and stenosis are maternal ergotamine use [142] and fetal exposure to methylene blue, which was formerly used as a marker during amniocentesis of twin pregnancies to ensure that both sacs had been sampled. [143] Multiple intestinal atresias have also been reported to be associated with neonatal immunodeficiency states. [144], [145]

Clinical Presentation Polyhydramnios is frequently detected in proximal gastrointestinal atresia, but the amniotic fluid may be normal in distal atresia. [136] Neonates most commonly present with signs and symptoms of obstruction within hours to 3 days after birth. Bilious vomiting early after birth is a characteristic symptom of proximal atresia, whereas abdominal distention occasionally associated with visible bowel loops, later vomiting, and a failure to pass meconium are frequent presenting complaints of distal lesions. High atresias of the jejunum and duodenum are also associated with jaundice. The course of infants who have intestinal stenosis without atresia may be much more indolent because the obstruction is usually partial. The diagnosis may therefore be delayed.

Diagnosis Polyhydramnios is an indication for ultrasonography, which may identify the obstructing defect. [125] Dilated loops of bowel are frequently detected; the presence of fetal ascites and extralumenal calcifications may indicate aseptic perforation of the bowel, with meconium peritonitis. The intralumenal calcifications found in type IV atresias may also be seen. After birth, routine abdominal radiography may demonstrate multiple air-fluid levels throughout the small bowel or dilated bowel proximal to the obstruction with an absence of gas distally. A double bubble sign on abdominal radiography or ultrasonography is classical for duodenal atresia. Barium enema shows an unused microcolon. The anatomic defect should always be definitively identified by plain films and cautious contrast radiography before surgical intervention is attempted. It is also important to identify all atretic sites, with the risk for multiple atresias kept in mind. Important in the differential diagnosis of neonatal bowel obstruction are Hirschsprung disease, malrotation, meconium ileus associated with cystic fibrosis, duplications, Meckel diverticulum, and mesenteric bands. [146] Barium enema is recommended to rule out Hirschsprung disease and malrotation with volvulus and to identify other atresias in the bowel distal to the primary obstruction.

Treatment and Prognosis Initial therapy consists of intravenous hydration, correction of electrolytes, and nasogastric suction to remove accumulated fluid and decompress the bowel. Surgical resection of atretic small intestine is the primary treatment, which may be challenging because bowel preservation is critical and dilation of the small bowel proximal to the obstruction makes anastomosis difficult. Several different surgical procedures have been developed, including tapering enteroplasty and the creation of an ileostomy. [137], [146] Patients with type IIIb and type IV atresias often require extensive resective surgery, which results in short bowel syndrome and a poor prognosis. All patients after surgery are placed on total parenteral nutrition and monitored for a return of normal bowel function as the diet is slowly advanced. A central venous catheter should be placed early in the course of the disease. Patients with short bowel syndrome frequently require prolonged parenteral nutrition. Isolated case reports of the use of growth hormone in ultrashort bowel syndrome suggest some beneficial effects. [137], [147] The morbidity and mortality rates in infants have dramatically decreased since the 1950s. Postoperative survival is approximately 90% to 95%. [133], [136] Enhanced survival is a result of improved management of the nutritional and pulmonary status. However, overall survival has remained at approximately 84% to 88%. [137] Factors adversely affecting the prognosis include the presence of other congenital anomalies (especially cardiac), chronic pulmonary disease, or pulmonary aspiration. [133], [135]

Malrotation

Description and Pathophysiology Alterations in the normal rotation and fixation of the gut during embryogenesis result in a series of anomalies with various clinical manifestations. Malrotation or incomplete rotation is the most common disorder of rotation and fixation; it occurs when the normal 270° bowel rotation is not completed. As a result, the cecum may lie in the subpyloric region or may be in a subhepatic location. The small bowel is not attached by a broad mesentery but remains suspended from a narrow vascular pedicle. Peritoneal bands (Ladd bands) may pass from the cecum across the duodenum to the right upper quadrant or may attach to the duodenum. The two major complications of malrotation are volvulus, caused by free rotation of the midgut around the vascular pedicle, and duodenal obstruction secondary to the Ladd bands. In nonrotation, the embryonic midgut loop does not rotate and the caudal limb of the loop reenters the abdominal cavity first.

Subsequently, the small intestine (jejunum and ileum) lies on the right side of the abdomen and the colon is entirely on the left. The cecum is usually located in the left iliac fossa. Frequently, the broad attachment of the small bowel mesentery does not develop, and the small bowel remains suspended by a narrow pedicle, at risk for volvulus. [129], [148] In reversed rotation, clockwise instead of counterclockwise rotation of the gut leads to reentry of the colon into the abdomen first. The position of the colon is then posterior to the superior mesenteric artery and the duodenum. The placement of the duodenum is anterior to the superior mesenteric artery. The small bowel mesentery passes in front of the transverse colon, so that a mesenteric tunnel forms that causes colonic compression and obstruction. Finally, failure of the cecum to descend into its normal position in the right lower quadrant may lead to mobile cecum; the cecum remains in the right upper quadrant and is unfixed posteriorly, predisposed to cecal volvulus. The ileum may similarly be unfixed and susceptible to volvulus.

Clinical Manifestations In approximately 50% to 80% of cases of midgut malrotation, symptoms develop in infancy. Related concurrent anomalies are biliary atresia and congenital heart disease. Intestinal atresia and stenosis may result from malrotation, and omphalocele and diaphragmatic hernia may lead to malrotation secondary to abnormal placement of the bowel during rotation. In adults, malrotation is sometimes associated with absence of the superior mesenteric vein and extrahepatic biliary anomalies. [149] The most severe complications of malrotation are small bowel obstruction and volvulus, which cause recurrent bilious vomiting, passage of bloody stools or acute onset of constipation, and bowel loop distention. In patients with Ladd bands, symptoms of duodenal obstruction may develop. Patients from 1 month of age through adolescence may have symptoms that are more varied and indolent, including intermittent vomiting, failure to thrive, and recurrent abdominal pain. [150] Adults may be asymptomatic for many years [151] and then present with midgut volvulus causing abdominal pain, bloody stools, and distention. [152] Alternatively, in adult patients with a long history of recurrent abdominal pain with nausea and vomiting since childhood, the obstruction or volvulus may never have been diagnosed because it was transient and resolved before presentation. [149]

Diagnosis The diagnosis may be challenging in the older child and adult. [153] Plain films may reveal evidence of duodenal or small bowel obstruction. A classic double bubble sign may be seen, indicating duodenal obstruction. However, early in the course of midgut volvulus, plain radiographs may be unrevealing, and children with bilious vomiting should undergo a contrast study to rule out volvulus. [154] Contrast radiographic studies may demonstrate anomalous small bowel, ligament of Treitz, and cecal locations. In adults with midgut volvulus, upper gastrointestinal contrast series may reveal a dilated stomach and duodenum with a typical “corkscrew” appearance of the barium in the twisted duodenum and proximal jejunum. [155] CT may show reversed positions of the superior mesenteric artery and vein. [156]

Treatment The surgical correction of malrotation (incomplete rotation) in symptomatic patients includes relieving the obstruction caused by Ladd peritoneal bands, freeing the duodenum, reducing midgut volvulus, and broadening the mesentery to prevent recurrent rotation of the bowel around a narrow mesentery. In the Ladd procedure, the bowel is placed in the position of nonrotation (small bowel on the right side of the abdomen and colon on the left). Appendectomy is performed because the cecum is incorrectly located. Variations of this operation are used to correct nonrotation and the very rare reversed rotation. In most patients, bowel function returns to normal. However, some children with recurrent and prolonged symptoms of nausea, vomiting, and pain preoperatively do not fare as well postoperatively, possibly because of persistent dysmotility syndromes associated with malrotation or damage to the small bowel resulting from long-standing, indolent obstruction. [157]

Gastroschisis and Omphalocele

Description and Embryonic Origin Although gastroschisis and omphalocele both result from abdominal wall defects, these rare disorders are distinct entities (see Chapter 121). In omphalocele, the abdominal viscera herniate through the umbilical ring and persist outside the body, covered by a membranous sac but not by skin. In gastroschisis, massive evisceration of the intestines occurs through a small defect in the abdominal wall, usually to the right of the closed umbilical ring. The bowel has no membranous covering, has been exposed to amniotic fluid in utero, and is often matted, thickened, foreshortened, and covered with adhesions. It has been hypothesized that omphalocele is caused by a failure of embryonic folding at the level of the lateral folds or persistence of the body stalk. [158], [159] Gastroschisis may be caused by vascular disruption in utero, leading to failure of differentiation of the somatopleural mesenchyme, [160] or later between the time of herniation into the umbilical cord and fixation, or by disruption of the right omphalomesenteric artery. The defect is thought to occur between the fifth and tenth weeks of gestation, but perinatal insults have also been suggested in patients in whom gastroschisis was not detected sonographically. [158], [161], [162] Other congenital anomalies that occur in association with omphalocele are chromosomal; about one third of cases of omphalocele with multiple anomalies have trisomy 13, trisomy 18, or the Beckwith-Wiedemann syndrome, which is characterized by macroglossia, large infant size, and visceromegaly. Gastroschisis is associated with intestinal anomalies, including atresia and malrotation, vascular disruptions, and renal and gall bladder agenesis. [158] Several mouse models of gastroschisis have been described. [163], [164] and [165] In mice that are null for the bone morphogenetic protein 1 (BMP-1) gene, the ALX-4 gene, or the aortic carboxypeptidase-like protein gene, gastroschisis develops with early mortality. Alx-4 is a homeodomain transcription factor. Both BMP-1 and the aortic carboxypeptidase-like protein appear to have important functions in the extracellular matrix. The relevance of these gene defects to human disease is unclear; a mutational analysis of the BMP-1 gene in human patients with gastroschisis was unrevealing. [166]

Prenatal Diagnosis Elevated levels of maternal serum a-fetoprotein are associated with ventral abdominal wall defects. Gastroschisis is detected with greater sensitivity than omphalocele. [159] Improvements in the accuracy of prenatal ultrasonography have led to more frequent detection. [167] One of the major advantages of prenatal diagnosis is that the obstetrician is alerted, so that at the time of delivery, the mother can be taken to a tertiary care center where appropriate surgical and neonatal intensive care support is available. [159] The role of preterm delivery to minimize intestinal damage is still unclear, and the factors that determine the need for this intervention are still being defined. [168] Recommendations for the appropriate mode of delivery (e.g., trial of labor versus elective cesarean section) are presently being debated, [169], [170] and [171] although it is clear that the rate of abdominal delivery is high because of peripartum complications. [172]

Treatment Surgical closure of omphalocele and gastroschisis is attempted shortly after birth. Primary closure may be possible with the use of retention sutures and stretching the abdominal wall. If this cannot be achieved, a silo or Silastic sac can be used with successive compression to reduce the herniation further. The decision to use a silo and a staged procedure may be based on intraoperative bladder or intragastric pressure measurements that reflect the intra-abdominal pressure. [168], [173] Careful regulation of the intra-abdominal pressure is critical for successful repair; high pressures that lead to respiratory distress and intestinal and renal ischemia, the major causes of morbidity and mortality, must be avoided. The material is placed over the herniated bowel or sac and reduced manually and serially sutured to reintroduce the bowel into the abdominal cavity. A comparison of primary emergent closure versus closure following reduction with a spring-loaded silo (for gastroschisis) suggested that silos improve the overall outcome. [174] Because almost all children have bowel hypomotility, total parenteral nutrition is recommended early in the course of the illness. Advances in surgical correction have greatly improved the prognosis of patients with gastroschisis, despite the fact that the bowel is usually damaged and slow to return to normal function. Survival rates now range from 70% to more than 90%. [169], [175], [176] The presence of necrotizing enterocolitis and other anomalies (affecting other organs) or associated intestinal conditions (e.g., atresias, perforations, necrotic segments, volvulus) affects mortality rates. [169], [177] In the absence of severe associated anomalies, the immediate survival and long-term outcome of patients with omphalocele have also improved in the past several years. [178], [179]

STRUCTURAL ANOMALIES

Volvulus

A volvulus is an abnormal twisting of the intestine around the axis of its own mesentery, resulting in obstruction of the more proximal bowel. Twisting of the mesentery may involve the mesenteric vessels and so make the involved loop particularly susceptible to strangulation and gangrene, with resulting perforation, peritonitis, and sepsis. In contrast to colonic volvulus, particularly of the cecum and sigmoid colon, small bowel volvulus is relatively rare in the United States and most of Europe. [180] It occurs more frequently in parts of Africa, the Middle East, and the Indian subcontinent. [180], [181] and [182] Volvulus in these countries occurs in the absence of anatomic abnormalities and is probably related to dietary factors. It has been postulated that ingesting large amounts of bulky foods after fasting may predispose to torsion of the small bowel loops. In the United States, small bowel volvulus is usually caused by a preexisting defect, such as an anomaly of rotation and fixation, postoperative adhesion, or congenital bands. [183]

Clinical Presentation and Diagnosis Patients present with symptoms of small bowel obstruction and an acute abdomen. Abdominal pain, nausea, and vomiting are almost always present [180], [184]; the severity of the pain may be out of proportion to the physical findings. Signs include abdominal distention, rebound, guarding and rigidity, and occasionally a palpable abdominal mass. Plain abdominal radiographs taken in supine and upright positions may demonstrate distended bowel with air-fluid levels, consistent with obstruction. Perforation may be indicated by the presence of free air. Barium studies can be useful. A typical corkscrew-like appearance of the barium in the distorted duodenum and jejunum is diagnostic. Angiography may reveal twisting of the branches of the superior mesenteric artery.

Therapy and Outcome The treatment of small bowel volvulus is surgical. Ischemic or gangrenous loops of bowel should be resected, although derotation of the bowel may be sufficient therapy in itself. Vascular compromise of the small bowel with subsequent gangrene is common and leads to increased morbidity and mortality postoperatively. Rapid recognition of volvulus and prompt surgical intervention are the keys to decreasing the fatality rate associated with this condition.

Intussusception

Description and Pathophysiology Intussusception occurs when a segment of bowel invaginates, or telescopes, into adjacent distal bowel, leading to obstruction and possible ischemic injury. The incidence, causes, clinical presentation, and therapy of intestinal intussusception are different in adult and pediatric patients. Intestinal intussusception is one of the most common causes of small bowel obstruction in children younger than 2 years of age, but it is an unusual cause of bowel obstruction in adults. Most pediatric cases occur in children younger than 5 years of age. [185] Pediatric

intussusception is most often idiopathic but may be associated with a pathological lead point in 8% to 12% of cases, [186], [187] including Meckel diverticulum; a variety of benign and malignant tumors, such as polyps, leiomyomas, and lymphomas; duplications; and Henoch-Schönlein purpura, in which an intramural hematoma acts as a lead point. An association between rotavirus vaccination and intussusception has been described, [188], [189] perhaps secondary to a lead point of vaccine-induced lymphoid hyperplasia. In idiopathic intussusception, an association with prominent Peyer patches and enlarged mesenteric lymph nodes has been observed. In children, ileocolic intussusceptions are most common, followed by ileoileocolic, cecocolic, and, much less frequently, ileoileal involvement. In adults in the Western world, small bowel intussusception occurs rarely, accounting for approximately 5% of all cases of intestinal obstruction. [190] A causative factor can be identified in more than 90% of adult patients, [191] whereas it cannot in children. Small intestinal intussusception may be precipitated by tumors, including leiomyomas, neurofibromas, lipomas, lymphomas, small bowel adenomatous polyps, and metastatic tumors with or without peritoneal carcinomatosis. Other causes include Meckel diverticulum or other diverticular disease and celiac disease with chronically dilated flaccid bowel. Postoperative intussusception may result from adhesions, or intussusception may follow trauma. [192] Jejunogastric intussusception may occur after Billroth II surgery, and bypassed bowel may become intussuscepted after jejunoileal bypass. [191] Patients with acquired immunodeficiency syndrome and Kaposi sarcoma or diffuse enteritis are also at risk. [193] Pregnancy and the use of long intestinal or cantor tubes are other predisposing factors.

Signs and Symptoms The typical pediatric patient is a well-nourished, previously healthy child between the ages of 5 months and 5 years. The peak presenting age is 3 to 11 months. [194] Classical signs and symptoms include the acute onset of intermittent abdominal pain, vomiting, and hematochezia. A palpable abdominal mass, diarrhea, and somnolence are other frequent findings. [186], [187] In older children, these diagnostic signs and symptoms are often not present. [185], [195] In adults, the clinical picture may be confusing. Abdominal pain is almost always present [196] but is often low-grade and chronic; patients may present after several episodes have spontaneously resolved. [190], [197] A partial or complete small bowel obstruction may be present, and an abdominal mass is often palpable. Nausea and vomiting are particularly associated with small bowel compared with large bowel intussusception. Weight loss may also occur in patients with chronic, indolent symptoms.

Diagnosis and Therapy

Pediatric patients. In children, an air or water-soluble contrast barium enema is performed because the ileocecal region is so frequently involved. [187] Contrast enemas demonstrate the location of the intussusception and are often successful in reducing it. Air enemas are also safe and effective in reducing intussusceptions. [154], [198], [199] and [200] The enema is usually administered with a surgeon in attendance in case of perforation or failure to reduce. Sonography may be used to diagnose intussusception and to monitor reduction. [200] In extremely ill patients with peritonitis, perforation, or shock, enema reduction is contraindicated. Radiologic reduction is successful in 50% to 90% of cases. [200] If it is not, the patient is taken to laparotomy for manual reduction. Resection is performed if ischemia or gangrene of the bowel is present. A careful search is made for a possible pathological lead point. Plain films may reveal a crescent of gas capping the intussusceptum, outlining its leading edge, or a target sign, consisting of two concentric radiolucent curvilinear lines outlining the intussusception. [201] A gasless area may also be identified, corresponding with the soft tissue mass of the intussusception. Ultrasonography may show a classical target or doughnut sign, characterized by multiple concentric rings of sonolucency alternating with one or two echogenic foci. The edematous outer and inner walls of the intussusception create two hypoechoic areas ringing the hyperechoic lumenal mucosa. [202] CT may similarly be useful, but it is usually avoided in children. Upper gastrointestinal contrast series may show proximal intestinal dilatation and a “bird’s beak” at the site of obstruction but is contraindicated if perforation or peritonitis is suspected.

Adult patients. Intussusception is diagnosed and treated differently in adult patients because its location and causes are different from those in children. Most intussusceptions in adults are in the small bowel, although they may also occur in the colon. [203] Therefore, a combination of plain film, upper gastrointestinal series, barium enema, and CT or ultrasonography is used in adult patients. Because of the often puzzling presentation of intussusception in this age group, CT is frequently performed. A mass of alternating high and low attenuation may be seen as a target, sausage-shaped, or bilobed lesion. [204] Mesenteric fat appears as areas of low attenuation, whereas the bowel wall itself appears as an area of high attenuation. Thickened bowel loops and an intralumenal soft tissue mass may be seen. However, this technique is not sensitive enough to determine the nature of the pathological lead point. Because a pathological lesion is found so frequently, treatment in adults consists of surgical intervention with bowel resection. Manual reduction alone may be pursued only if it is certain that no other lesions are present. [196] With present diagnostic techniques, however, it is unlikely that a tumor or another anatomic cause can be ruled out preoperatively. Therefore, manual reduction of intussusception in adults is usually not recommended because manipulation can lead to intralumenal or intravenous tumor seeding. [195], [205] Colonic intussusception is never treated with manual reduction because of the extremely high likelihood of malignancy. [197]

Lymphangiectasia

Description and Pathophysiology Intestinal lymphangiectasia is characterized by the obstruction of lymph drainage from the small intestine and the dilation of lacteals and other intestinal lymphatics, such as those in the serosa and mesentery, depending on the level of obstruction. As a result of obstruction and increased pressure in the intestinal lymphatics, the absorption of chylomicrons and fat-soluble vitamins such as vitamin D is impaired, the reentry of intestinal lymphocytes into the peripheral circulation is impeded, and excessive amounts of intestinal lymph “leak” into the intestinal lumen. Lymphenteric fistulae may form, and intestinal lymph containing chylomicrons, protein, and lymphocytes drains directly into the intestinal lumen. Chylomicrons are sequestered in the lamina propria as well as in the distended lymphatics. Blockage of serosal and mesenteric lymphatics may lead to chylous ascites, and blockage of the thoracic duct to chylous pleural effusions. Intestinal lymphangiectasia may occur as a primary congenital disorder or may be secondary to a disease that blocks the intestinal lymph drainage at some level. Causes of secondary lymphangiectasia include extensive abdominal or retroperitoneal carcinoma or lymphoma, retroperitoneal fibrosis, chronic pancreatitis, mesenteric tuberculosis or sarcoidosis, [206] Crohn’s disease, Whipple disease, scleroderma, [207] celiac disease, [208] constrictive pericarditis, [209] systemic lupus erythematosus, [210] and chronic congestive heart failure. Congenital intestinal lymphangiectasia (Milroy disease) results from a malformation of the lymphatics that often affects many areas of the body.

Clinical Presentation Patients present with varying degrees of steatorrhea and malabsorption, lymphocytopenia (especially of T lymphocytes), marked hypogammaglobulinemia with impaired cell-mediated immunity, and prominent manifestations of protein-losing enteropathy. They often have edema and low serum protein levels, the reduction in serum albumin usually being the most pronounced and the only one of clinical significance. Patients with congenital disease present at any time from birth to adulthood, often with asymmetric edema of an extremity caused by peripheral lymphatic obstruction. They may also present, as do the secondary cases, with more diffuse, symmetric edema, usually the result of marked hypoproteinemia. Despite lymphocytopenia and impaired delayed hypersensitivity reactions, opportunistic infections are not common. However, infections with atypical mycobacteria, warts, and cellulitis have been reported. [211] Gastrointestinal symptoms are usually not prominent, but some patients may have diarrhea, abdominal pain, distention, nausea and vomiting, and occasionally gastrointestinal bleeding.

Diagnosis Protein-losing enteropathy should be suspected in any patient with unexplained hypoalbuminemia, and intestinal lymphangiectasia should be considered if lymphocytopenia and steatorrhea are also present. Asymmetric lymphedema, especially dating from infancy or childhood, should suggest congenital (Milroy) disease. A diagnosis of lymphangiectasia rests on peroral jejunal biopsy demonstrating dilated lymphatic lacteals. Several specimens may be required to demonstrate the diagnostic findings because the lesions are often patchy and localized. [212] Endoscopy may reveal dilated lacteals that appear as white opaque spots or white-tipped villi, and nodular lesions and xanthomatous plaques have also been noted. [213], [214] These findings aid the endoscopist in selecting appropriate regions to sample. Other pathological findings include moderate villus blunting and mild to moderate inflammatory infiltration. [212] Other tests demonstrate protein-losing enteropathy by detecting excessive enteric loss of plasma proteins. The use of radiolabeled plasma proteins, such as ^{131}I-albumin, ^{51}Cr-albumin, and ^{51}Cr-chloride, has been generally replaced by the measurement of gastrointestinal clearance of a $_{1}$-antitrypsin. Double-contrast radiographs of the small bowel may reveal folds thickened by intestinal edema, nodular protrusions, and an absence of mucosal ulceration. [215] If secondary lymphangiectasia is suspected, appropriate tests such as CT of the abdomen should be performed to diagnose the underlying disease. The degree of malabsorption and nutritional deficiency may be assessed by quantifying stool fat and by measuring the prothrombin time, which is nonspecific, and the serum levels of calcium and carotene. In congenital cases, if necessary, the malformed, hypoplastic lymphatics can be demonstrated by lymphangiography. Pleural effusions and ascitic fluid may be tapped for conventional diagnostic studies and examined for chylomicrons.

Therapy The therapy for lymphangiectasia should be directed toward treating the pathophysiological consequences, and in the case of secondary lymphangiectasia, the underlying disease (e.g., lymphoma, tuberculosis, sarcoidosis, constrictive pericarditis) should be diagnosed and treated. Substituting medium-chain triglycerides for the usual long-chain triglycerides may reduce enteric protein loss, malabsorption, and diarrhea and may improve serum albumin levels. [216], [217] Medium-chain fatty acids are more water-soluble and may be more readily absorbed through portal venous channels than through the lymphatics. The concomitant reduction in dietary long-chain fat presumably reduces chylomicrons in obstructed lymphatics and thereby decreases the lymphatic pressure and rate of lymph loss. Anecdotal reports have demonstrated the efficacy of octreotide [218], [219] and [220] or antiplasmin therapy in a patient with increased plasma fibrinolytic activity, [221] yet it is probable that most patients do not respond to this treatment. [222] Peripheral edema can be minimized by postural drainage and the use of elastic stockings to reduce the risk for cellulitis and lymphangitis.

Celiac Artery Compression

The topic of celiac artery compression is covered in Chapter 131.

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CHAPTER 73

Michael Camilleri

DYSMOTILITY OF THE SMALL INTESTINE AND COLON

The functions of intestinal and colonic smooth muscle are controlled mainly by the intrinsic and extrinsic nerves of the gastrointestinal tract and, to a lesser degree, by gastrointestinal hormones (see Chapter 11). Therefore, any abnormality of smooth muscle, intrinsic or extrinsic nerves, or gastrointestinal hormones can theoretically cause intestinal or colonic dysmotility. Abnormal circulating levels of gastrointestinal hormones have not definitely resulted in dysmotility; however, diarrhea in patients with carcinoid syndrome and irritable bowel syndrome is associated with elevated circulating levels of serotonin, and acceleration of small bowel and proximal colonic transit in these patients may at least partly reflect abnormal motor function.

The human small bowel and colon display widely divergent patterns of absorption, motility, and transit. Propulsive, peristaltic motility can move contents over long distances of the small intestine very rapidly, with efficiency approaching that of the esophagus. The small intestine functions to facilitate emptying from the stomach, the mixing of chyme with digestive enzymes and bile, and ultimately delivery of residue to the colon. The ileum possesses a specialized type of contraction (giant migrating contraction) that sweeps unabsorbed residue to the colon and is analogous to the high-amplitude colonic contractions. Intermittent emptying ensures that enough time is available for salvage of the remaining nutrients in the small intestine. The small intestine and colon also set up feedback inhibitory reflexes (e.g., ileal brake) that retard proximal motor functions such as gastric emptying.

In contrast to the fundamental motor programs of other regions in the gut, those of the colon are under strong inhibitory influences, slowing colonic transit for fecal contents to solidify. The well-developed muscular wall of the colon exhibits basal tone, but at the same time, it has a pronounced propensity to relax. [1] As the organ responsible for the final stages of absorption and elimination, the colon is highly relevant to diarrhea and constipation. Colonic fermentation of dietary residues, mainly the “unavailable carbohydrates,” generates hydrogen, carbon dioxide, short-chain fatty acids, and methane through the anaerobic action of bacterial enzymes; [2] distention of the abdomen and flatulence can follow. Finally, for imagined, cultural, and psychosocial reasons, many populations focus on their habits of elimination. Constipation is thus a major concern of many persons, and disturbances of colonic motility figure prominently in the minds of patients and physicians.

Dysmotility of the small intestine and colon has a wide range of clinical manifestations, regardless of the underlying cause of the disorder. Patients at one end of the spectrum may be asymptomatic, and at the other, they may have chronic intestinal pseudoobstruction, a syndrome characterized by recurrent or chronic symptoms that suggest obstruction in the absence of structural occlusion of the lumen. Between the two extremes, patients may have dyspeptic symptoms, including intermittent postprandial epigastric or periumbilical abdominal pain, bloating, nausea, vomiting, and diarrhea.

In patients with small intestinal dysmotility, motor function in other parts of the gastrointestinal tract also is usually abnormal. In this chapter, we focus our initial discussion on the small intestine; motility disorders of the colon are manifested predominantly as constipation and megacolon. Motility disorders of other parts of the gastrointestinal tract are discussed in other chapters (see Chapter 59, Chapter 65, and Chapter 86).

EPIDEMIOLOGY

The prevalence of small intestinal dysmotility varies according to the underlying disease, and it seems to be less frequent than esophageal, gastric, or colonic dysmotility. The lack of sensitive techniques to evaluate small intestinal motility has made it difficult to estimate its prevalence. Since the early 1970s, several hereditary diseases with small intestinal dysmotility have been reported, including two groups of diseases associated with smooth muscle degeneration, termed *familial visceral myopathies (FVMs)* [3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15 /SUP>, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28 and 29 and *childhood visceral myopathies (CVMs),* [30, 31, 32, 33, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55 and 56 and a group of diseases associated with myenteric plexus degeneration, termed *familial visceral neuropathies (FVNs)*. [57, 58, 59, 60, 61, 62, 63, 64, 65, 66, 67 and 68 Only a small number of families have been reported, mostly Caucasian and less often African American [9] and Latino. [69] These rare familial disorders are discussed in the first part of this chapter.

Hirschsprung disease, the prototypical congenital colonic dysmotility, affects 1 in 5000 newborns. In contrast, constipation affects about 12% to 15% of the population, with epidemiologic data suggesting an equal split between functional constipation and evacuation disorders. [70, 71] A telephone survey of approximately 10,000 adults assessed the prevalence of constipation (defined as an inability to have spontaneous complete bowel movements). Consistent with the results of prior studies, the overall prevalence was approximately 15%; however, only 2% of the respondents were classified as having constipation-predominant irritable bowel syndrome (IBS). [72] In contrast to the results of prior studies, which suggested that constipation increases with advancing age, these data showed an inverse trend, with a slightly lower prevalence associated with increasing age. [72] Other studies of clinic patients have indicated an overlap in the diagnoses of constipation-predominant IBS and functional constipation despite the use of strict definitions. [73, 74]

NEURAL CONTROL OF SMALL INTESTINAL AND COLONIC MOTILITY

The enteric nervous system (ENS) is a vast network of ganglionated plexuses located in the wall of the gastrointestinal tract. [75, 76] Although several plexuses are identified anatomically, the most important, from a functional perspective, are the myenteric and submucosal plexuses. In association with the muscle layers, the networks of interstitial cells of Cajal are recognized as the likely pacemakers activating neuromuscular function. The ENS consists of approximately 100 million neurons in higher mammals, and this number is roughly equal to the number of neurons in the spinal cord (Fig. 73-1).

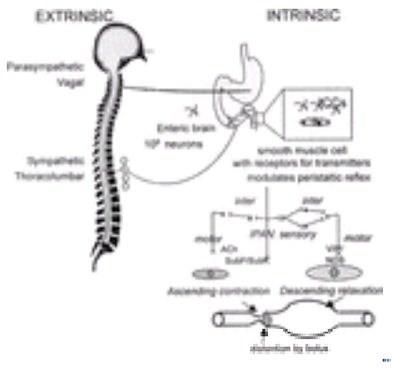

FIGURE 73-1. Extrinsic and enteric control of gut motility. The enteric nervous system controls stereotypical motor functions such as the migrating motor complex and the peristaltic reflex; enteric control is modulated by the extrinsic parasympathetic and sympathetic nerves, which respectively stimulate and inhibit nonsphincteric muscle. *ICC,* interstitial cells of Cajal; *IPAN,* intrinsic primary afferent neuron.

The ENS develops in utero as neural crest cells migrate to the developing alimentary canal. The migration of neural crest cells, and the sequence of innervation of different levels of the gut, are regulated by specific signaling molecules that include transcription factors (e.g., Mash-1), neurotrophic factors (e.g., the glial cell line–derived neurotrophic factor [GDNF] and its receptor subunits), and the neuregulin signaling system. These facilitate the growth, differentiation, and persistence of the migrating nerve cells once they arrive in the gut. Neuregulins are a large group of structurally related signaling proteins that are likely to have important roles in the development, maintenance, and repair of the nervous system and other selected tissues. Their receptors are the Erb-B protein tyrosine kinases, which are important in cell signaling.

Histological and electrophysiological studies [77], [78], [79] and [80] of the intestinal tract have characterized the properties of the neurons and transmitters mediating its functions, including the peristaltic reflex and the interactions between neurons and inflammatory cells. [81]

ONTOGENY OF THE ENTERIC NERVOUS SYSTEM

Migration

The ENS cells are derived from precursor cells from three axial levels of the neural crest. These include the vagal, [82] rostral-truncal, [83] and lumbosacral [82], [84] levels. The enteric neurons mainly arise from the vagal neural crest of the developing hindbrain and colonize the gut by migrating in a rostrocaudal direction. Vagal crest cells are not restricted to a particular intestinal region. Some enteric neurons arrive in the hindgut from the lumbosacral level via a caudorostral wave of colonization. Rarely, the migrating cells do not reach the entire gut; usually, the terminal portion of the bowel is affected, as in classical forms of Hirschsprung disease. Even more rarely, a zonal form of Hirschsprung disease may occur (Fig. 73-2 and Fig. 73-3), which is postulated to result from the incomplete caudorostral migration of neuroblasts during embryonal development. [85] The neural crest cells that migrate and colonize the gut become neuroblasts or neuronal support cells, glioblasts. However, *differentiation* into neurons and glial cells seems not to take place until they reach their final destinations in the gut. Movement through the gut mesenchyme, survival in the gut, and differentiation into mature cells are influenced by contacts of the precursor cells with the *microenvironment*.

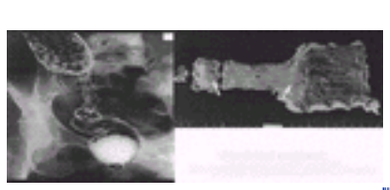

FIGURE 73-2. Zonal Hirschsprung disease in an adult: narrowed segment of the sigmoid colon on barium enema and gross pathological examination. (From ref. [85].)

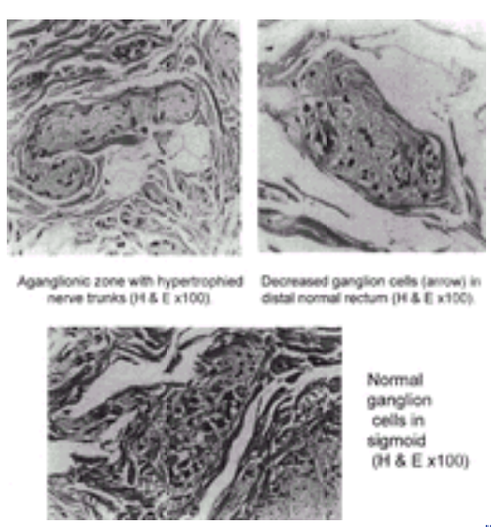

FIGURE 73-3. Zonal Hirschsprung disease in an adult: variation in the number of ganglion cells in different segments of the resected specimen. (From ref. [85].)

The microenvironment consists of other cells in the mesenchyme, neural crest–derived cells, and the extracellular matrix. The extracellular matrix components provide directional signals to migrating neural crest cells and, together with neighboring cells, provide signals for neural crest cell differentiation. For example, the appearance of neural crest cells in the gut is preceded by the expression of extracellular matrix molecules, [86] and other factors, such as GDNF, ensure the survival of committed neuroblasts. [87]

A subpopulation of sacral neural crest cells appears predetermined to function in the hindgut. These cells do not require the presence of vagus-derived enteric precursors to colonize the hindgut, nor are they capable of dramatically altering their proliferation or differentiation. [88] On the other hand, the environment at the sacral level allows neural crest cells from other levels of the axial region of the developing nervous system to enter the mesentery and gut mesenchyme. At least two environmental conditions at the sacral level enhance ventral migration of the sacral neural crest cells. First, sacral neural crest cells take a ventral rather than a medial-to-lateral path through the somites and arrive near the gut mesenchyme many hours *earlier* than their counterparts at the thoracic level. The window of opportunity to invade the mesenchyme of the mesentery and the gut is narrow, so their earlier arrival ensures that the sacral neural crest cells will gain access to the gut. Second, the gut endoderm is more dorsally situated (i.e., closer to the crest with its migrating cells) at the sacral level than at the thoracic level. As a result, sacral neural crest cells preferentially populate the colorectum. Moreover, a *barrier to migration* at the thoracic level prevents neural crest cells at that axial level from migrating to the gut. [89] The barrier also prevents lumbosacral crest neurons and glial cells from migrating to the nearby small intestinal tissue. [90]

Defects of the neural crest cells themselves or alterations of the microenvironment of the pathway through which the neural crest cells migrate may result in maldevelopment of the ENS. In humans, such disordered development results in congenital enteric neuromuscular diseases.

Differentiation of Neurons

Migrating crest cells are multipotent. [91] The gut wall is itself a critical site where *terminal differentiation of the enteric neurons and glia* occurs and determines what kind of nervous system arises within the bowel. [92] Enteric growth factor–receptor combinations influence differentiation. Combinations that enhance differentiation include the following: GDNF–GFR-1–RET, NT-3–Trk-C, and 5-HT–5-HT$_{2B}$. The RET protooncogene encodes a tyrosine kinase receptor necessary for the development of the ENS. A qualitatively different effect on nervous system development is shown by the peptide-receptor combination ET-3–ET-B, which *prevents the premature differentiation* of enteric neurons before colonization of the gastrointestinal tract has been completed.

The first molecule found to affect the development of enteric neurons and glia was neutrotrophin-3 (NT-3). [93] Crest-derived cells in the fetal bowel express tyrosine kinase C (Trk-C), the high-affinity receptor for NT-3. Overexpression of NT-3 in transgenic mice causes an increase in the size of developing ganglia and neurons in the myenteric plexus. [92] The ENS, however, is relatively normal in the bowel of mice following the knock-out of NT-3, [94] suggesting that NT-3 affects the development of only a subset of enteric neurons or glia. [91, 95]

Stimulation by GDNF is *absolutely* necessary for the survival of the vagal and sacral crest–derived cells that colonize the gut. If either GDNF [96, 97] or its signaling receptor RET [98] is knocked out in developing mice, the vagal and sacral domains of the bowel become totally aganglionic, and ganglia persist only in the small region of the gut that is colonized by cells from the truncal crest. GDNF potently promotes neuronal development in vitro [99, 100] and acts as a mitogen in early development, [99] greatly expanding the numbers of enteric crest–derived neural precursors. The initial GDNF-dependent crest-derived precursor that colonizes the bowel gives rise to multiple cell lineages that require particular growth or transcription factors. For example, the truncal crest depends on GDNF, not RET, [83] but it also requires Mash-1. From this GDNF- and Mash-1– *dependent* lineage, all the serotoninergic neurons (which develop early in ontogeny) and many excitatory and inhibitory motor neurons develop. Later, GDNF loses its ability to promote proliferation, and it acts only as a growth-differentiation factor for enteric neurons, not for glia. All enteric neurons that contain calcitonin gene–related peptide are derived from Mash-1– *independent* neurons and differentiate late in ontogeny, after the last serotoninergic neuron has become postmitotic (i.e., terminally differentiated).

Both crest-derived and non–crest-derived cells of the enteric mesenchyme also contain GFR-1, [99] a peripheral glycosylphosphoinositol-anchored molecule that binds GDNF and is necessary for the activation of RET. [101] Neural crest cells anchor GFR-1 to their plasma membranes (perhaps in a complex with RET), where GDNF can bind to it and ensure survival of the crest-derived cells that colonize most of the bowel. [92]

Enteric serotoninergic neurons appear so early that they coexist in primordial enteric ganglia with still-dividing neural precursors. Serotonin (5-HT) may be more than a neurotransmitter; the 5-HT$_{2B}$ receptor in the fetal bowel is regulated and optimal at specific times, so that 5-HT strongly promotes the development of neurons at specific times and affects the development of late-arising enteric neurons. [102]

These growth-differentiation factors affect virtually the entire bowel. The peptide endothelin-3 (ET-3) and its receptor, ET-B, play a critical *localized role* in ENS development. [103, 104] Examples of tyrosine kinase receptor mutations associated with specific genetic disorders, such as the multiple endocrine neoplasia (MEN) syndromes, are shown in Figure 73-4.

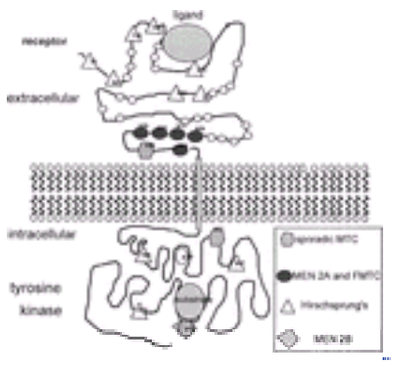

FIGURE 73-4. Tyrosine kinase receptor with examples of mutations associated with specific genetic disorders. *[F]MTC,* [familial] medullary carcinoma of the thyroid; *MEN,* multiple endocrine neoplasia. (Drawn from Edery P, Lyonnet S, Mulligan LM, et al. Mutations of the RET proto-oncogene in Hirschsprung's disease. Nature 1994;367:378.)

CHRONIC SMALL INTESTINAL PSEUDOOBSTRUCTION

For a review, the reader is directed to Di Lorenzo. [105]

Etiology

Table 73-1 lists the causes of small intestinal dysmotility. The mechanisms and manifestations of small intestinal dysmotility are shown in Figure 73-5. In general, small intestinal dysmotility may be secondary to neuropathy or myopathy. This is best distinguished by specialized histological and immunohistochemical study of the full thickness of the gastrointestinal wall, which is available at few centers. Smooth muscle is best studied by sectioning across the muscle fibers and using trichrome stain. [106] The best way to evaluate the myenteric plexus is still evolving. Silver staining of sections of the gastrointestinal wall taken parallel to its long axis, described by Smith [107] and by Schuffler and Jonak, [108] was state of the art but has been superseded by immunohistochemistry and confocal examination of neurons and a search for genetic markers (see below). Thus, a detailed description of phenotypes in familial cases is less pertinent than the more mechanistic or etiologic descriptions that appear below.

Primary Causes
Familial types
Familial visceral myopathies: types I, II (MINGE), III
Familial visceral neuropathies: types I, II
Childhood visceral myopathies: type I, type II (megacystis-microcolon-intestinal hypoperistalsis)
Nonfamilial or sporadic types
Visceral myopathies
Visceral neuropathies
Secondary Causes
Disease involving the intestinal smooth muscle
Collagen diseases (e.g., scleroderma, dermatomyositis, systemic lupus erythematosus, mixed connective tissue disease)
Muscular dystrophies (e.g., myotonic dystrophy, Duchenne muscular dystrophy)
Amyloidosis
Neurological diseases
Chagas disease, ganglioneuromatosis of the intestine, paraneoplastic neuropathy, Parkinson disease, spinal cord injury
Endocrine disorders
Diabetes mellitus, thyroid disease (i.e., hyperthyroidism, hypothyroidism), hypoparathyroidism
Pharmacological agents
Phenothiazines, tricyclic antidepressants, antiparkinsonian medications, ganglionic blockers, clonidine
Narcotics (morphine and meperidine)
Miscellaneous intestinal disorders
Celiac disease
Small intestinal diverticulosis
Radiation enteritis
Diffuse lymphoid infiltration of the small intestine
Jejunoileal bypass
After gastrointestinal viral infection

MNGIE, mitochondrial neurogastrointestinal encephalomyopathy.

TABLE 73-1 Causes of Small Intestinal Dysmotility

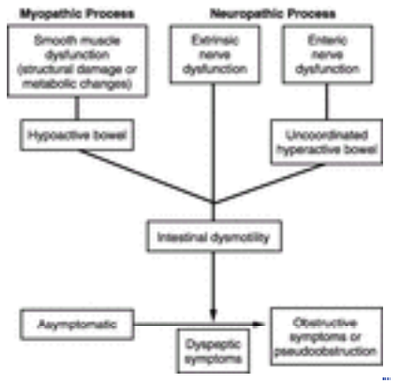

FIGURE 73-5. Mechanisms and manifestations of small intestinal dysmotility.

Primary Causes of Small Intestinal Dysmotility Primary dysmotilities are rare compared with secondary dysmotilities. They may be familial or sporadic.

Familial visceral myopathies. The FVMs are a group of genetic diseases characterized by the degeneration and fibrosis of smooth muscle of the gastrointestinal tract and, in a certain type, the urinary tract. At least three types of FVM have been reported based on gross lesions of the gastrointestinal tract and the pattern of inheritance (Table 73-2), and on documented mitochondrial DNA disorders in type II FVM, also called *oculogastrointestinal neuropathy*. On routine pathology, the histological features of these three types of FVM do not differ. [106] The involved areas show a characteristic change, consisting of degenerating muscle cells and fibrosis that may involve the full thickness of the muscularis propria but often is more prominent or limited to the external layer (Fig. 73-6). Degenerating muscle cells appear pale, poorly defined, and fragmented. As residual threadlike remnants become surrounded by collagen, the longitudinal and circular muscles take on a vacuolated appearance that is easily recognized microscopically (Fig. 73-7). Recognition of this change is greatly facilitated by the use of a trichrome stain. However, special histochemical stains may identify giant mitochondria as ragged red fibers in skeletal muscle or affected bowel; cytochemistry can also identify mitochondrial enzyme deficiencies in mitochondrial neurogastrointestinal encephalomyopathy (MNGIE), or type II FVM. Because several advances in our understanding of this condition have been made, a further detailed discussion of MNGIE is warranted.

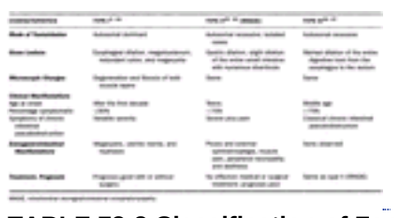

TABLE 73-2 Classification of Familial Visceral Myopathies

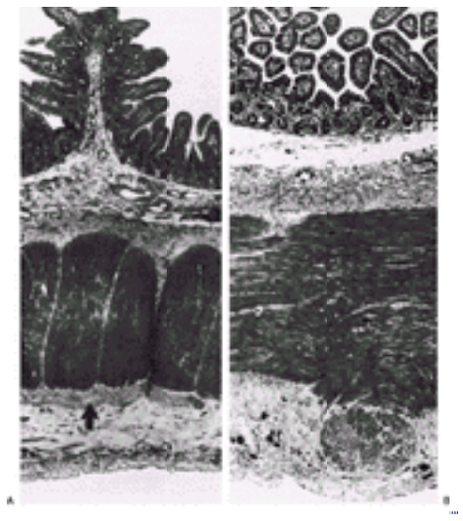

FIGURE 73-6. A: Longitudinal section of jejunum from a patient with familial visceral myopathy shows normal circular muscle layer and a prominent neural bundle (*arrow*). Scattered muscle fibers and collagen are observed in the longitudinal layer. Trichrome stain; original magnification ×40. **B:** Cross section of jejunum from area depicted in **A.** The outer muscle layer contains one large bundle of preserved smooth muscle fibers and scattered individual fibers throughout the collagen elsewhere. Trichrome stain; original magnification ×40. (From ref. [106].)

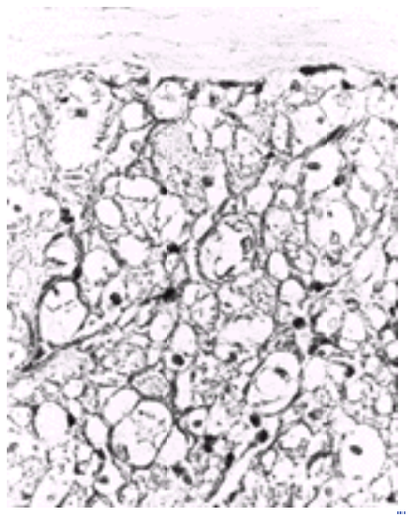

FIGURE 73-7. The characteristic vacuolar change, with collagen fibers encircling spaces filled by fragmented muscle cells, in a patient with familial visceral myopathy. Trichrome stain; original magnification ×470. (From ref. [106].)

Mitochondrial DNA and myopathies. These diverse diseases are characterized by mitochondrial abnormalities in skeletal muscle, which show “ragged red fibers” on a Gomori trichrome stain. The metabolic abnormality involves cells in other tissues: central and peripheral nervous system, gut, heart, kidney, liver, thyroid, pancreas, and bone marrow. The enzymes in the respiratory chain of mitochondria are encoded by nuclear and mitochondrial DNA. The disorders may be sporadic or inherited in an autosomal dominant, autosomal recessive, or X-linked pattern; maternal inheritance is more frequent. Mitochondria are involved in the production of energy (ATP), the generation of reactive oxygen species, and the initiation of apoptosis through activation of the mitochondrial

permeability transition pore. [109] Mitochondrial DNA is made up of two to ten double strands of circular DNA in the mitochondrial matrix. At least 37 genes have been discovered, some of which encode the respiratory chain enzymes. The transcription of mitochondrial DNA is not closely related to the cell cycle, and mitochondrial DNA is more susceptible to mutation than is nuclear DNA. Mitochondrial disorders manifest organ disturbances that reflect the importance of mitochondria to normal function, as in skeletal muscle and the nervous system; similarly, metabolic disorders result from deranged cellular mitochondrial functions. The locations of selected mutations in the mitochondrial 16,569-base pair genome that result in pathological disorders and syndromes have been documented. [109] The mitochondrial disorder affecting the gut is MNGIE, also referred to as *MEPOP, oculogastrointestinal muscular dystrophy,* and *FVM type II*. This is an autosomal recessive condition with gastrointestinal and hepatic manifestations that may present at any age, typically with hepatomegaly or hepatic failure in neonates, seizures or diarrhea in infants, and hepatic failure or chronic intestinal pseudoobstruction in children or adults. MNGIE is characterized clinically by severe gastrointestinal dysmotility, external ophthalmoplegia, ptosis, peripheral neuropathy, and leukoencephalopathy. The small intestine is dilated, or multiple diverticula are present (Fig. 73-8), and the amplitude of contractions is typical of a myopathic disorder. [110] Some patients have a combination of intestinal dysmotility and the Kearns-Sayre syndrome, or transfer dysphagia secondary to abnormal coordination and propagation of the swallow through the pharynx and skeletal muscle portion of the esophagus. Clearly, this becomes even more devastating when the smooth muscle portion of the esophagus is affected by the associated MNGIE.

FIGURE 73-8. Enteroclysis in a patient with type II familial visceral myopathy demonstrates numerous diverticula in the small intestine.

Apart from the obvious external ophthalmoplegia, these patients manifest skeletal muscle pain and cramps, and systemic (lactic) acidosis. Circulating levels of muscle enzymes (creatine phosphokinase [CPK], alanine aminotransferase [ALT], aldolase, others) are elevated, and muscle biopsy shows characteristic ragged red fibers on modified Gomori stain. This appearance results from the hypertrophy of mitochondria in the subsarcolemmal position in a few muscle fibers and the lack of mitochondria in other muscle fibers. Special stains for the respiratory muscle enzymes can identify the precise functional defect. For example, succinate dehydrogenase–positive fibers appear "ragged blue," and staining of the adjacent tissue section with cytochrome *c* oxidase demonstrates a deficiency of the latter enzyme and a gene defect in the control of the complex IV respiratory chain proteins (Fig. 73-9). [110] In the intestine, hypertrophy of the circular muscle layer, atrophy of the longitudinal muscle, and megamitochondria in myenteric neurons and muscle cells are seen (Fig. 73-10). [111]

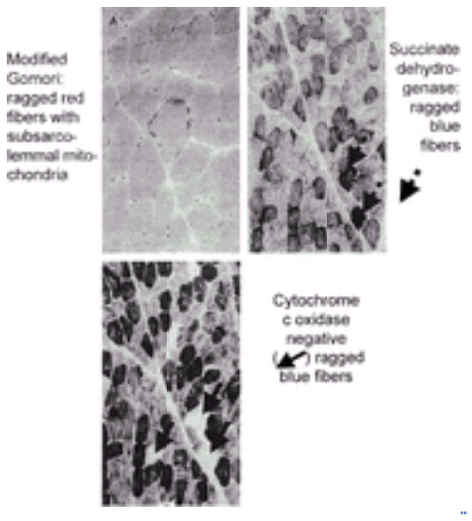

FIGURE 73-9. Histological and histochemical studies of a skeletal muscle biopsy specimen from a patient with mitochondrial myopathy; note the ragged red fibers, characterized by a subsarcolemmal location of giant mitochondria in a few fibers and a paucity of mitochondria in other fibers. On histochemical analysis, a few fibers are positive for succinate dehydrogenase (ragged blue appearance), but the same fibers do not express cytochrome *c* oxidase, suggesting a defect in the respiratory enzyme chain that results in mitochondrial dysfunction and systemic acidosis. (From ref. [110].)

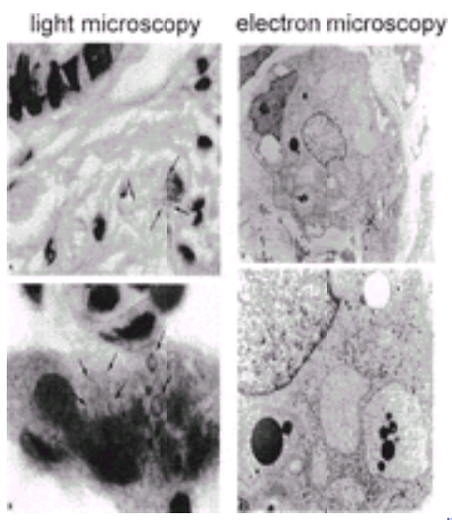

FIGURE 73-10. Submucosal plexus neurons showing megamitochondria in a rectal biopsy specimen from a patient with mitochondrial myopathy. (From ref. [111].)

The screening tests for MNGIE are measurement of serum lactic acid, muscle enzymes, and thymidine phosphorylase in circulating leukocytes. [112] The latter test is based on mutations identified in the gene for thymidine phosphorylase in 21 probands with MNGIE. [112]

Familial visceral neuropathies. The FVNs are a group of genetic diseases characterized by degeneration of the myenteric plexus. At least two distinct phenotypes, I and II, have been identified and are summarized in Table 73-3. A number of syndromic congenital neuropathies are related to disturbances in the ontogeny of the neural crest and the ENS.

TABLE 73-3 Classification of Familial Visceral Neuropathies

Congenital neuropathic motility disorders. These disorders can be broadly classified as follows: [113]

1. *Disorders of colonization* by migrating neural crest derived neurons, as in Hirschsprung disease, related to abnormalities in the RET gene and GDNF, or the disorder of ET-3 and its receptor, ET-B.
2. *Disorders of differentiation* of enteric nerves, as in intestinal ganglioneuromatosis, related to a specific germ-line point mutation in RET at codon 918 of exon 16 (M918T) or at codon 883 (A883F) in MEN type 2B syndrome.
3. *Disorders of survival or maintenance of enteric nerves,* as in hypoganglionosis and possibly congenital achalasia, which can result from one of several mechanistic derangements: GFR $_{a2}$/neurturin, GFR $_{a3}$/artemin, Trk-C/NT-3, and 5-HT $_{2B}$/5-HT.

Hirschsprung disease, a relatively common condition affecting 1 in 5000 live births and causing intestinal obstruction in neonates and megacolon in infants and adults, is often associated with mutations in genes encoding GDNF-RET [114], [115] or ET-3–ET-B. [104], [116] Mutations in RET and ET-B (i.e., the receptors) are much more frequently encountered than mutations in the two ligands. At least two different mechanisms can cause the terminal colon to become aganglionic. In the first, a deficiency of GDNF-RET is not so severe as to cause the entire bowel to become aganglionic. In the second, a deficiency of ET-3–ET-B, crest-derived cells differentiate prematurely and precursors cease dividing and migrating before the gut has been entirely colonized, leaving the last segment uncolonized. The roles of endothelins and of gene mutation M918T in the maldevelopment of the ENS and their role in Hirschsprung disease are discussed later. At least six separate genetic loci are involved in the control of four different intracellular mechanisms that result in models of congenital dysmotility and their phenotypic manifestations in the gut and other tissues (Table 73-4). These include abnormalities of c-RET, the gene that encodes the tyrosine kinase receptor; the ET-B system; the SOX-10 transcription factor; and c-kit, which is a marker for the interstitial cells of Cajal. Disturbances in these mechanisms result in syndromic dysmotilities such as Hirschsprung disease, Waardenburg-Shah syndrome (piebaldism, neural deafness, megacolon), and idiopathic hypertrophic pyloric stenosis. Figure 73-4 shows some of the mutations in the tyrosine kinase receptor that have been reported in dysmotilities associated with familial or sporadic medullary carcinoma of the thyroid, MEN types 2A and 2B, and Hirschsprung disease. Visceral neuropathy may result in bowel dilation (Fig. 73-11), although this is generally less frequent or less severe than in visceral myopathy. MEN type 2B is discussed in greater detail below because it is a prototypical illness that demonstrates the multiple effects of the mutations in the control of neural crest development.

TABLE 73-4 Genetic Models of Hirschsprung Disease

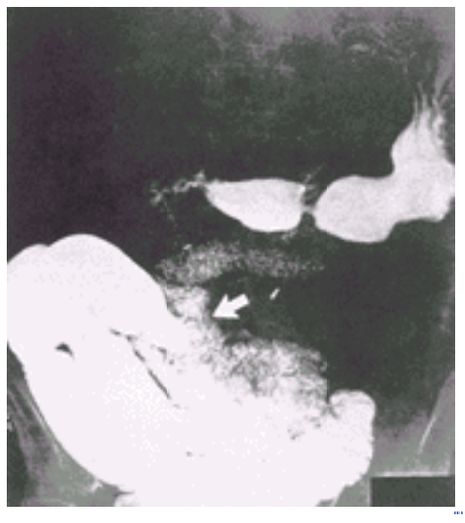

FIGURE 73-11. Small bowel radiograph from a patient with type I familial visceral neuropathy shows a normal stomach, duodenum, and proximal jejunum, but a dilated distal small bowel (*arrow*). (From ref. [58].)

Multiple endocrine neoplasia type 2B syndrome. MEN type 2B is a serious congenital neuropathic condition associated with tumors of the neuroendocrine system. It presents with severe constipation or megacolon (Fig. 73-12), diarrhea (when associated with enterocolitis), or obstruction, often in infancy. [117], [118] Other external stigmata of MEN 2B are a characteristic facies, “blubbery lips” resulting from mucosal neuromas, marfanoid habitus, medullated corneal nerve fibers, and even medullary thyroid carcinoma. [117], [119] The latter develops eventually in almost all patients.

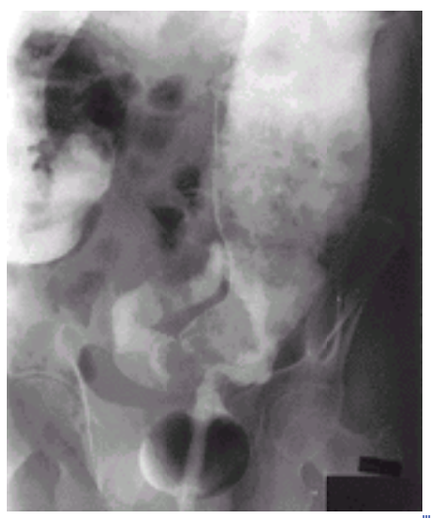

FIGURE 73-12. Megacolon in a patient with multiple endocrine neoplasia type 2B; note the marked distension of the colon with a relatively small rectal diameter (**A**), and the massive dilation of the transverse colon with scoliosis and deviation of the mediastinum (**B**).

In MEN type 2B, a feature of the intestinal pathology is transmural intestinal ganglioneuromatosis, in which massive proliferations of neural tissue (neurons, supporting cells, and nerve fibers) appear as thickened nerve trunks among mature nerve cells (Fig. 73-13).

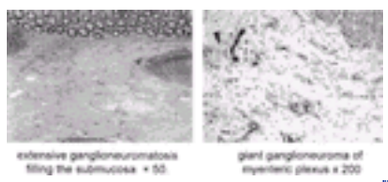

FIGURE 73-13. Histopathological features of ganglioneuromatosis in multiple endocrine neoplasia type 2B; note the extensive ganglioneuromatosis filling the submucosa and the giant ganglioneuroma of the myenteric plexus. (From ref. [125].)

MEN type 2B is a dominantly inherited disorder, but at least half of all patients present with a de novo mutation because only a few survive to reproductive age and are disabled by a variety of neurological symptoms that impair reproduction. The precise molecular abnormality is in the RET protooncogene (see Fig. 73-4), which encodes a tyrosine kinase receptor expressed particularly in neural crest–derived cells, including the enteric ganglia. A specific germ-line point mutation in RET in exon 16 at codon 918 (M918T) occurs in 95% of patients; the remainder have a point mutation at codon 883 (A883F). [120], [121] The M918T mutation alters RET substrate specificity and appears to act in a ligand-independent fashion (a gain-of-function mutation). [122], [123] and [124] Receptor tyrosine kinases with these mutations also bind to and phosphorylate substrates preferred by nonreceptor tyrosine kinases. A883F RET has not been directly tested, but given its location, it is thought that kinase specificity may also be altered in this mutant. [121] The receptor is expressed in the neural crest cell–derived ENS, adrenal medulla, parathyroid, and C cells of the thyroid. The gain-of-function mutations are susceptibility factors for endocrine tumors (medullary carcinoma of the thyroid, pheochromocytoma, and parathyroid tumors). Because surgery is the only curative procedure for medullary carcinoma of the thyroid, Smith and colleagues [125] recommend that patients who present with a clinical phenotype of Hirschsprung disease undergo adequate full-thickness biopsy of the bowel at the time of the pull-through operation, and that those with transmural intestinal ganglioneuromatosis undergo molecular diagnostic testing by RET mutation analysis. If germ-line M918T or A883F mutations are found, a prophylactic thyroidectomy is indicated; furthermore, adrenal gland surveillance with abdominal ultrasonography and urinary fractionated catecholamines and metanephrine or vanillylmandelic acid are indicated because a pheochromocytoma may develop subsequently in about 50%.

Other familial visceral neuropathies. A few reported families do not fit into the two types of FVN discussed previously. [20], [67], [68] Schuffler and colleagues [67] reported two siblings who had generalized neurological disease and intestinal pseudoobstruction. One had ataxic gait; small, irregular, poorly reactive pupils; dysarthria; absent deep tendon reflexes; and impaired vibratory and positional senses. Radiographs revealed hyperactive, nonpropulsive contractions in a dilated esophagus and small intestine, and extensive colonic diverticulosis. The postmortem examination revealed degeneration of the myenteric plexuses of the esophagus, small intestine, and colon with reduced number and round, eosinophilic, intranuclear inclusions. The neurons and glial cells of the brain and spinal cord and the dorsal root and celiac plexus ganglia contained identical intranuclear inclusions. Intestinal smooth muscle was normal histologically. In the family reported by Cockel and colleagues, [68] affected members had megaduodenum, generalized dilation of the small intestine, malabsorption, redundant colon, mental retardation, and calcification of the basal ganglia. Histological studies showed degeneration of the argyrophobe cells, which produce acetylcholine. The patients became symptomatic for intestinal pseudoobstruction during late childhood. Faber and colleagues [20] reported five patients with chronic intestinal pseudoobstruction in two families. Hypertrophy of nerve fibers and an absence of ganglion cells were noted. The disease was transmitted by an autosomal recessive gene. All patients had megaduodenum; one had jejunal diverticulosis, and another dilation of the entire small intestine. Three had gastric dilation. Most patients had external ophthalmoplegia, ptosis, distal weakness of all extremities, gait disturbances, and acroparesthesia. The symptoms developed during the teen years, and the prognosis was poor.

Childhood visceral myopathies. The CVMs have been recognized as distinctive diseases. There are two forms of CVM (Table 73-5), the second of which is identified by the phenotype of megacystis-microcolon-intestinal hypoperistalsis. These two diseases differ from the FVMs in their clinical manifestations and modes of inheritance. Degeneration and fibrosis of gastrointestinal tract and urinary tract smooth muscle can be detected in both types of CVM [28], [34], [45], [126] and result in bowel dilation (Fig. 73-14), ureteropelvic pyelocaliectasis (Fig. 73-15), or megacystis, which results from bladder degeneration (Fig. 73-16).

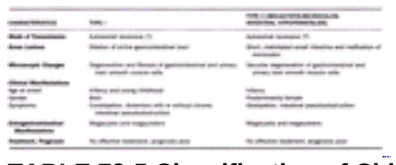

TABLE 73-5 Classification of Childhood Visceral Myopathies

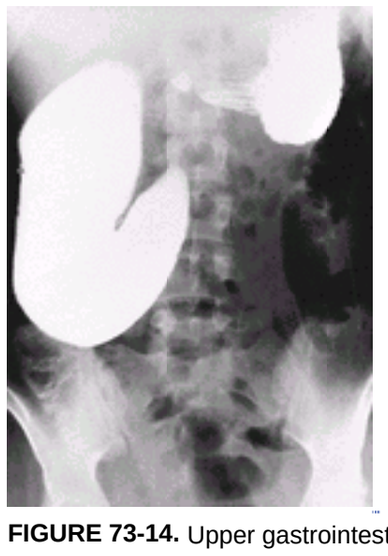

FIGURE 73-14. Upper gastrointestinal radiograph from a patient with type I familial visceral myopathy demonstrates severe megaduodenum.

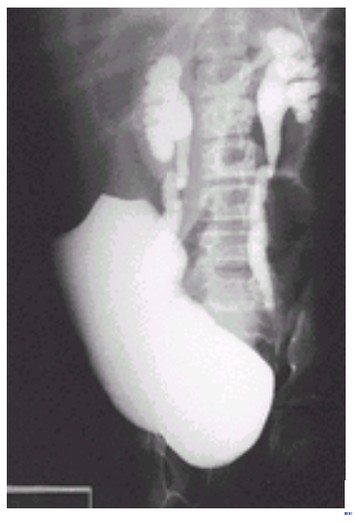

FIGURE 73-15. An intravenous pyelogram of a child with type I childhood visceral myopathy shows megacystis and bilateral ureteral pyelocaliectasis. (From ref. [126].)

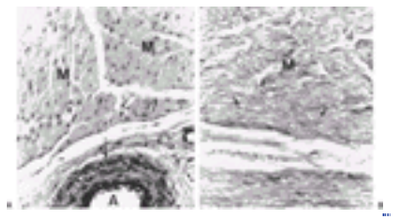

FIGURE 73-16. A: Bladder muscularis from a control specimen demonstrates elastic fibers (*arrow*) in the adventitia of a small artery (*A*). No elastic fibers are present within muscle bundles (*M*). **B:** Bladder muscularis from a patient with type I childhood visceral myopathy demonstrates numerous parallel, coarse, wavy, elastic fibers (*arrows*) within muscle bundles (*M*). Verhoeff-van Gieson stain; original magnification ×325. (From ref. 126.)

Nonfamilial visceral myopathies (sporadic hollow visceral myopathy). Nonfamilial visceral myopathies in adults have been reported a few times. 127, 128, 129, 130, 131, 132 and 133 The disease is heterogeneous, with some similarities to FVM types I and III. 130 Histologically, no difference is seen between the familial and nonfamilial types. Diffuse mixed inflammatory cell infiltration, including eosinophils, was observed in one patient. 127 In one reported case, 133 acetylcholinesterase stain of the small intestine showed a considerably increased number of positive fibers, but no degeneration of neurons in the myenteric plexus. The nonfamilial type of visceral myopathy is uncommon. A careful family history must be obtained in these patients to exclude a diagnosis of FVM.

Idiopathic nonfamilial visceral neuropathies (sporadic hollow visceral neuropathy or chronic idiopathic intestinal pseudoobstruction). Nonfamilial visceral neuropathy or chronic idiopathic intestinal pseudoobstruction typically results from damage to the myenteric plexus by chemicals, drugs, or viral infections. Delayed gastric emptying has been observed in patients after viral gastroenteritis, 134, 135 which may cause permanent damage to the myenteric plexus in some cases. In patients with nonfamilial visceral neuropathies, 136, 137, 138, 139, 140, 141, 142 and 143 the motility of the entire gastrointestinal tract is usually disturbed, but the urinary tract is generally not affected. The intestine is dilated but shows active, nonperistaltic contractions on barium examination. 142 In contrast, patients with familial visceral myopathies have an inactive, dilated intestine. 142 Histological examination of the myenteric plexus with the Smith silver staining technique in sporadic visceral neuropathy reveals a reduction in the number of neurons, and those that remain are markedly abnormal (Fig. 73-17). The neurons are enlarged, with thick, irregular, clubbed processes. The number of Schwann cells in the nerve trunks is increased, and the number of axons is reduced. Some of the remaining axons are beaded and fragmented. Hypertrophy of one or both muscle layers of the muscularis propria may also be observed. All reported cases have chronic intestinal pseudoobstruction. Infants and young children are also affected. 143

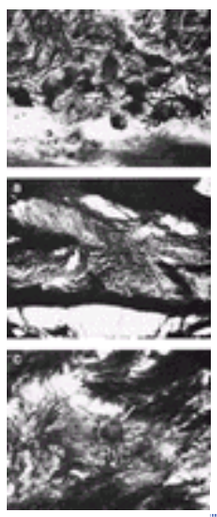

FIGURE 73-17. A: Ganglionic area from normal control ileum demonstrates numerous argyrophilic neurons and nerve fibers. Silver stain; original magnification ×230. **B:** Ganglionic area from a patient with type II familial visceral neuropathy demonstrates absence of argyrophilic neurons and few nerve fibers. There is a faint cluster of small nuclei of unknown cell type. Silver stain; original magnification ×230. **C:** Another ganglionic area from the same patient shows only one poorly staining, argyrophilic neuron (*arrow*). Silver stain; original magnification ×362. (From ref. 58.)

In a study of 40 patients with chronic idiopathic intestinal pseudoobstruction, the results of intestinal phasic pressure studies were abnormal, but the histologic features of the gastrointestinal tract were normal in those whose tissue was available for analysis. 144 Similar findings have been reported in children with chronic idiopathic intestinal pseudoobstruction. 145 In earlier reports, 146, 147 and 148 histological studies of smooth muscle or myenteric plexus were not generally available for evaluation. The precise mechanism and neurotransmitter deficiencies of this disorder are unclear. Differentiation from severe IBS may be difficult if the gut is not dilated. Manometric studies may also show similar features, such as bursts or clusters of phasic pressure activity; 149, 150 and 151 slow transit of chyme through the stomach or small bowel 152 provides an objective, noninvasive means by which dysmotility can be suspected.

Secondary Causes of Small Intestinal Dysmotility The secondary causes of small intestinal dysmotility are summarized in Table 73-6 and discussed in the next sections.

TABLE 73-6 Gastrointestinal Dysmotility Manifestations of Some Systemic Diseases

Scleroderma (progressive systemic sclerosis). The small bowel is the second most frequently involved gastrointestinal organ in scleroderma, after the esophagus. Radiographic evidence of gastric sclerosis, intestinal sclerosis, or both is found in 40% of patients with scleroderma. 153, 154, 155 and 156 Contrast studies reveal dilation of the duodenum and jejunum, delayed barium transit, and narrow valvulae conniventes, which remain tightly packed together despite the bowel dilation, producing an accordion appearance (Fig. 73-18). 10, 156, 157 Wide-necked diverticula may be seen but are more frequent in the colon. 158 An uncommon and potentially serious finding is pneumatosis cystoides intestinalis, which usually signifies a poor prognosis. 159

FIGURE 73-18. Upper gastrointestinal series in a patient with scleroderma shows dilation of the small bowel with normal appearance of the stomach. In some cases, the valvulae conniventes are not thickened, as is usually seen in disorders that produce dilation. In contrast, they remain narrow and crowded, producing the accordion appearance shown here. (From ref. [156].)

Degeneration of the smooth muscle and replacement with collagen is responsible for small bowel dysmotility in scleroderma. The circular muscle layer is involved more often than the longitudinal muscle layer. [10], [160], [161] The submucosal and myenteric plexuses appear normal by conventional staining. Small bowel dysmotility leads to bacterial overgrowth, resulting in steatorrhea, malabsorption, and weight loss. Intestinal pseudoobstruction is a well-described gastrointestinal complication of scleroderma. [10], [162], [163], [164] and [165] In patients with scleroderma and intestinal involvement, disturbances of small bowel motility [166], [167] and [168] and transit [169] have been documented in several studies. Many patients have a complete absence of the interdigestive migrating motor complex (MMC); clusters of propagated and nonpropagated contractions may be the only finding in others. An abnormally prolonged MMC cycle, diminished activity of phase III, and hypomotility of the fed pattern are other abnormal findings. [166], [168], [170] Antral hypomotility, when present, is characterized by low-amplitude contractions, typically below 40 mm Hg; intestinal involvement usually causes the contractile amplitude to fall below 10 mm Hg during fasting and postprandially. [171] In patients who have scleroderma without gastrointestinal involvement, the small bowel manometric pattern [171] and small bowel transit time are normal. [172] Treatment with cisapride, a prokinetic agent, improves gastric emptying and relieves upper abdominal symptoms in patients with scleroderma. [173] Oral erythromycin was effective in short-term studies that evaluated gastric emptying and symptoms in an open-label study. [174] Octreotide, a long-acting somatostatin analog, induces phase III activity in patients with a complete absence of MMCs and may reduce bacterial overgrowth and symptoms (Fig. 73-19). [167] More studies are required to confirm these observations and the long-term benefit of the treatments.

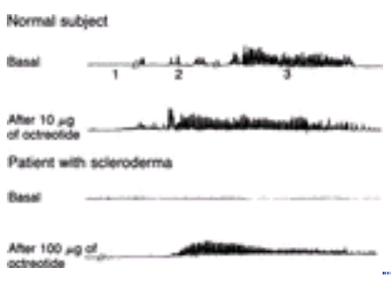

FIGURE 73-19. Effect of octreotide in a healthy person and in a patient with scleroderma. In the normal person, octreotide stimulates phase III activity. In the patient with scleroderma and pseudoobstruction, octreotide induces phase III activity even though no migrating motor complex activity was present in the basal state. (From ref. [167].)

Dermatomyositis and polymyositis. The inflammatory disorders dermatomyositis and polymyositis are characterized by weakness and atrophy of the skeletal muscles; an associated skin rash distinguishes the former from the latter. The gastrointestinal tract is involved in half of cases. Esophageal involvement causes dysphagia, which often is a presenting symptom. Studies of the small bowel in these patients are limited mainly to radiography and histology. Megaduodenum and delayed intestinal transit of barium are prominent features. [175], [176] and [177] Autopsy findings have shown atrophy and fibrosis of intestinal smooth muscle, suggesting a visceral myopathy. [176], [177] Acute colonic pseudoobstruction was reported in one patient with dermatomyositis, but it was limited to the colon. [178]

Systemic lupus erythematosus. Abdominal pain is the most common gastrointestinal symptom in systemic lupus erythematosus, occurring in 10% to 20% of patients. Autopsy studies disclose a 60% to 70% prevalence of previous peritoneal inflammation, but documented cases of serositis as the sole cause of abdominal pain are rare. [179] The term *lupus enteritis* is used to describe the bowel changes resulting from lupus-induced vasculitis of the small blood vessels. Ischemia leads to intestinal mucosal ulceration, edema, and hemorrhage. Smooth muscle dysfunction results in dilation of the small bowel and ileus. In severe cases, necrosis of the bowel wall develops, with perforation or infarction. Lupus enteritis is diagnosed radiographically. Small bowel changes include dilation, coarsened folds, and “thumbprinting” secondary to submucosal edema and hemorrhage. [180], [181] Diarrhea occurs in 5% to 10% of lupus patients. Unusual intestinal manifestations can pose diagnostic difficulties. Small bowel involvement in lupus may simulate Crohn’s ileitis in some cases. [182], [183] A severe protein-losing enteropathy was described in one patient with lupus, who demonstrated vasculitis and basement membrane thickening on full-thickness jejunal biopsy. [184] Transit or manometric studies of the small bowel have not been performed in patients with lupus.

Mixed connective tissue disease. Mixed connective tissue disease is a clinical entity characterized by overlapping features of scleroderma, polymyositis, and rheumatoid arthritis. It has a distinct serologic finding of high titers of antinuclear antibody against ribonucleoprotein. The extent of gastrointestinal involvement is unknown but appears to resemble that of scleroderma, with esophageal involvement being most common. Duodenal and jejunal dilation are seen radiographically, and pneumatosis cystoides intestinalis may complicate severe disease. [185]

Diabetes mellitus. Many diabetic patients have diarrhea, which has a variety of causes (e.g., bacterial overgrowth, pancreatic exocrine insufficiency, bile salt malabsorption, impaired absorption or secretion). Early radiographic studies of the small bowel demonstrated variable findings, including normal, delayed, or rapid transit. Similarly discrepant results are found when the hydrogen breath test is used, and the correlation of rapid transit, based on that test, with diarrhea is poor. [186], [187] and [188] The small bowel may be affected in patients presenting with the gastroparesis syndrome, and this can be detected by manometry [189] or prolonged transit. [169] Pathologically, demyelination of the proximal vagus nerve and sympathetic nerves supplying the bowel develops in diabetes. The intrinsic nervous system of the gut appears to be unaffected because no morphologic abnormalities of the myenteric or submucosal plexuses have been noted. [190], [191] Although thickening of the small bowel muscle layers and eosinophilic hyaline bodies in smooth muscle cells have been observed, [192], [193] and [194] most authorities do not believe that myopathy is a cause of gastrointestinal dysmotility in diabetic patients. The results of motility studies of the small bowel in patients with diabetes have been mixed. The MMCs are normal in many patients with documented gastroparesis. [195] Absence of intestinal phase III has been demonstrated in some patients. [186], [196], [197] Other abnormalities include MMCs originating in the distal duodenum or jejunum and uncoordinated (nonpropagated or abnormally propagated) bursts of powerful contractions lasting up to 2 minutes (Fig. 73-20). [186], [189] The clinical relevance of these abnormal findings is uncertain. In one study, abnormal transit time and manometric abnormalities could not be correlated. [186] Hyperglycemia, which can weakly delay gastric emptying, induces intestinal phase III–like activity and alters orocecal transit time in healthy subjects. [198] However, the relevance of these findings to diabetic bowel dysmotility is unclear. Among community diabetics, constipation and the use of laxatives are significantly more common than in age- and gender-matched controls. [199], [200] Constipation is probably multifactorial in diabetes, as it is in nondiabetic patients [201]; acute hyperglycemia does not significantly affect colonic motor function or tone. [202]

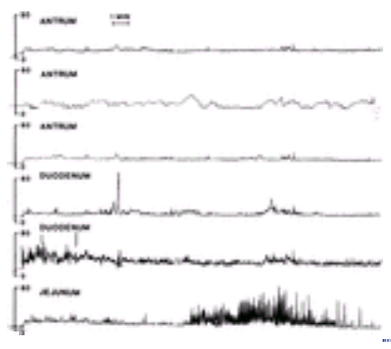

FIGURE 73-20. Small bowel dysmotility in diabetes. In this patient, phase III of the migrating motor complex begins in the jejunum, with no antecedent activity in the antrum or duodenum. Other patients may have a complete absence of phase III activity. (From ref. [186].)

Parkinson disease. Symptoms of gastrointestinal dysmotility are common in patients with Parkinson disease. High dysphagia, nausea, bloating, constipation, and difficulty with evacuation of stool occur frequently. [203] Dilation of the small bowel may be seen radiographically. [204] Small bowel dysmotility does occur; however, its frequency is not known. Manometric studies in patients with Parkinson disease reveal infrequent MMCs (which are even absent in some cases), hypomotility in the fed state, and an increased incidence of retrograde and tonic contractions in comparison with controls. [205] The contribution of these abnormalities to symptoms is not clear. The pathogenesis of small bowel dysmotility is unknown. In patients with dysphagia and megacolon, Lewy bodies, which are neurons containing cytoplasmic hyaline inclusions, have been found in the myenteric plexus of the esophagus and colon. [206], [207] Lewy bodies were originally identified in the brain and are highly characteristic of Parkinson disease. Lewy bodies have not yet been reported in the small bowel. Dopaminergic neurons are reported to be deficient in the colon of patients with parkinsonism and constipation. [208] Therapy for Parkinson disease includes drugs that adversely affect gut motility, such as dopamine agonists and anticholinergics. Symptoms are as common in untreated as in treated patients, however, suggesting that the disease itself is the primary factor producing dysmotility and resultant symptoms. [203]

Spinal cord injury. Extrinsic denervation of the small bowel by spinal cord injury usually produces just mild and probably insignificant dysmotility in the small intestine. The only change revealed in manometric studies performed in a group of patients with spinal cord injury was that a greater number of phase III contractions began in the duodenum rather than the antrum in those with high spinal cord lesions. [209] Patients with injury to the lower spinal cord demonstrated no abnormal findings, consistent with the preservation of innervation to the bowel from the vagus and third thoracic sympathetic levels. In one report, a woman with cervical spinal stenosis and paraplegia displayed normal MMCs but a twofold to threefold prolonged interval between MMCs. [210] Immediately after a spinal cord injury, a state of spinal shock develops. This is characterized by the complete loss of all sensory, motor, and reflex function below the level of injury. Paralytic ileus with abdominal distention follows and usually resolves in a few days. In the long term, postprandial abdominal distention and discomfort occur in more than 40% of patients with spinal cord injury. [211] These symptoms are likely caused by the more significant problem of constipation. In many stable patients with spinal cord injury, the amount of gas in the small and large intestine is increased on routine abdominal x-ray films (Fig. 73-21). [212] Colonic dysmotility is well recognized and is responsible for the common problems of constipation and difficulty of evacuation in these patients. [213]

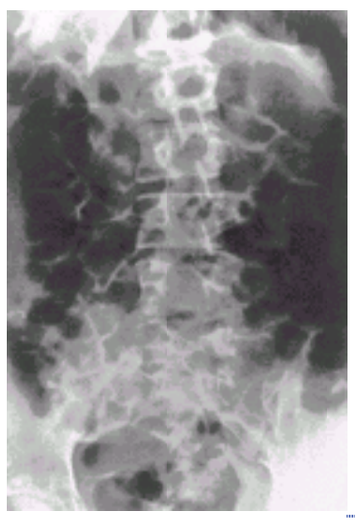

FIGURE 73-21. Abdominal radiograph from a stable, healthy patient with a spinal cord injury. A nonobstructive gassy abdomen is frequently observed, even in the absence of abdominal complaints. (From ref. [212].)

Neurofibromatosis (von Recklinghausen disease). Gastrointestinal neurofibromas are most commonly found in the small intestine. They are reported to occur in up to 10% of patients with neurofibromatosis; the true incidence of neurofibromas is probably underestimated, however, because they are generally asymptomatic. Small bowel neurofibromas may be single or multiple and are most frequently found in the ileum. These tumors are submucosal, and they may become quite large and extend to the serosa. Their presence is usually established by a small bowel series. [214], [215] Ulceration through the mucosa can cause acute or chronic bleeding. Mechanical small bowel obstruction or intussusception requires exploratory surgery. Chronic intestinal pseudoobstruction has been described in one patient with neurofibromatosis. The small bowel manometry findings were abnormal, and small bowel transit was markedly delayed. [152] Pathological studies of the nerves and muscles in the small bowel are lacking in this disease. Neurogenic changes consisting of nerve fiber proliferation in the myenteric and submucosal plexuses were found in the colon of a patient with megacolon. [216]

Paraneoplastic visceral neuropathy (pseudoobstruction). Chronic intestinal pseudoobstruction has been reported in association with small cell carcinoma of the lung [217], [218], [219], [220], [221], [222] and [223] and epidermoid carcinoma of the lip. [224] This phenomenon represents a paraneoplastic syndrome caused by visceral neuropathy. The small bowel, like the colon, shows widespread neuronal and axonal degeneration of the myenteric and submucosal plexuses, in addition to mononuclear cell infiltration and Schwann cell proliferation. [221] There are no metastases to the bowel. Symptoms of pseudoobstruction are the presenting features in patients with visceral neuropathy and small cell carcinoma. The lung cancer is usually occult, missed on chest radiography, and is often found at autopsy or by chest computed tomography (CT) and mediastinoscopy. The small bowel is usually not dilated on x-ray films, but barium passage through the small bowel is somewhat delayed. Small bowel manometry shows features suggestive of neuropathy, such as incoordinated bursts. [220], [221] In older patients with intestinal pseudoobstruction or feeding intolerance of unknown cause and recent onset, a malignancy, especially small cell carcinoma of lung, must be suspected. A screening blood test (antineuronal enteric antibody, or anti-Hu) is highly specific. [221] If the result is positive, CT of the chest and mediastinoscopy may be necessary to confirm the diagnosis.

Myotonic dystrophy. Myotonic muscular dystrophy is a slowly progressive disease characterized by myotonia, or difficulty in muscle relaxation. Besides muscle wasting, the patient often has a nasal voice as a result of pharyngeal and palatal weakness, an early onset of cataracts, and cardiac conduction defects. Dysphagia is the most common gastrointestinal symptom resulting from esophageal involvement. Diarrhea and abdominal cramping occur in up to one third of those affected. Malabsorption and steatorrhea have been reported in a few cases. [225], [226] and [227] Constipation also is frequent and can alternate with diarrhea. Intestinal pseudoobstruction is rare. [228], [229] The small bowel may demonstrate abnormal but nonspecific radiographic changes, including dilation, diminished motility, and delayed transit of barium. [230] Dysmotility of the small intestine may play a significant role in the production of intestinal symptoms. In a group of ten patients with symptoms of intestinal dysmotility but normal findings on small bowel contrast examinations, small bowel motility was abnormal in all. Manometric findings included low-amplitude contractions in the fasting and fed states, retrograde propagation of phase III, interruption of phase III, and an increased incidence of tonic contractions. [231] In one patient with chronic pseudoobstruction and dilated small bowel, manometry revealed low-amplitude duodenal contractions and an increased maximal rate of contractions in the duodenum. [229] Histologically, the small intestinal smooth muscle shows changes similar to those found in dystrophic skeletal muscle; it is swollen, partially destroyed, decreased in size, and replaced by fat. [232], [233] Silver stain revealed degenerative changes in the myenteric plexus of the colon in a patient with megacolon, [234] indicating that intestinal dysmotility may be caused by smooth muscle as well as enteric nerve dysfunction. In most patients, the predominant cause of dysmotility appears to be smooth muscle damage.

Duchenne muscular dystrophy. Duchenne muscular dystrophy is a severe, pseudohypertrophic muscular dystrophy that follows an X-linked recessive pattern, affecting young boys early in childhood. Skeletal and cardiac fibers are infiltrated and eventually replaced by connective and fatty tissue. Similarly, the smooth muscle of the gastrointestinal tract also is involved. Histological changes comprise degeneration and atrophy of smooth muscle fibers and separation of the fibers by connective tissue. [235], [236] and [237] The myenteric plexus is not involved. Despite typical dystrophic changes in the small bowel in most postmortem specimens, gastrointestinal symptoms are usually related to dysmotility of the esophagus and stomach, which are more severely affected than the small bowel. Dysphagia is the predominant symptom (36% in one series), followed less often by vomiting, diarrhea, and constipation. [237] The orocecal transit time is normal in asymptomatic subjects; [238] however, severe bowel dysmotility can occur. Episodes of acute intestinal pseudoobstruction, manifested by abdominal distention, persistent vomiting, and gastric or small bowel dilation, can be life-threatening. [239], [240] and [241] Chronic intestinal pseudoobstruction has also been reported in Duchenne dystrophy. [242]

Amyloidosis. Amyloid protein is deposited in the small bowel in all forms of amyloidosis: primary, secondary, myeloma-associated, and familial or hereditary. The degree of dysmotility is related to the amount and distribution of amyloid deposited. In all forms, the deposition of amyloid in blood vessel walls leads to ischemic ulcers, infarction, and occasionally perforation. In primary and myeloma-associated forms of amyloidosis, the muscle layers of the small bowel are more severely affected than the mucosa, resulting in dysmotility. In the secondary and hereditary forms, the mucosa is predominantly involved, resulting in malabsorption. Amyloid deposits are infrequently found in the myenteric plexus, although in hereditary forms, familial amyloidosis neuropathy may result from deposition in major nerve trunks supplying the bowel. [243], [244] Gastrointestinal symptoms include diarrhea, constipation (which is often present for years and then followed by diarrhea), malabsorption, and pseudoobstruction. Diarrhea can occur without malabsorption and is relieved by antibiotic therapy. [245] Radiographic features include coarsening of the small bowel mucosal pattern, dilation, and diminished motility with prolonged transit of barium. [246] These changes are nonspecific and can be mistaken for those of scleroderma. In a mixed group of patients with amyloidosis, the

orocecal transit time was prolonged to more than twice that of controls. [247] With heavy infiltration of the small bowel muscle layers, acute intestinal pseudoobstruction is a serious and often fatal complication (Fig. 73-22). [248] , [249] and [250]

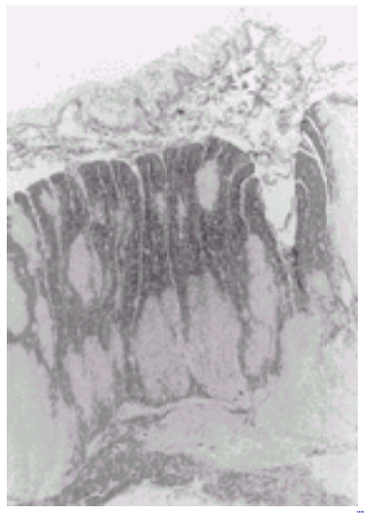

FIGURE 73-22. Small bowel myopathy in a patient with amyloidosis and pseudoobstruction. The large, pale areas in the muscle layer represent heavy amyloid deposition. The myenteric plexus is less frequently involved than the mucosa and muscle layers. (From ref. [250] .)

Chagas disease. Chronic infection with *Trypanosoma cruzi* leads to destruction of the submucosal and myenteric plexuses along the entire length of the gastrointestinal tract. Megacolon and megaesophagus are the most common presentations, although the small bowel may be similarly affected, with the development of megaduodenum and megajejunum. Small intestinal manometric studies in these patients demonstrate normal frequency of the MMCs, but the velocity of propagation of the activity front is significantly slowed to about half the normal value. [251] Patients with small bowel involvement may be entirely asymptomatic despite significant neuronal destruction. [252] Diarrhea, constipation, or intestinal pseudoobstruction can occur.

Thyroid disease. Thyroid dysfunction may present primarily as gastrointestinal disease. Hyperthyroid patients may have diarrhea and mild steatorrhea, whereas hypothyroid patients frequently are constipated. [253] , [254] and [255] Intestinal dysmotility resulting from the altered thyroid state has been historically recognized as the cause of symptoms. This is supported by abnormalities found in the intrinsic electrical control of gut motor activity. In the duodenum, slow wave frequencies are increased in hyperthyroid patients and decreased in hypothyroid patients. [256] , [257] These abnormalities return to normal after correction of the thyroid dysfunction.

Hyperthyroidism. Early radiographic studies, in which the crude barium meal test was used, reported rapid gastric emptying and small bowel transit. [258] , [259] Later studies, in which the hydrogen breath test was used, confirmed a rapid orocecal transit time; this is a consequence of accelerated small bowel transit because the rate of gastric emptying is actually unchanged. [253] , [260] , [261] and [262] Hyperthyroid patients may present with malabsorption; more than 25% of hyperthyroid patients excrete more than 7 g of fat in their stool daily. [263] , [264] This is apparently caused by decreased contact time of the lumenal contents with small bowel mucosa as a result of rapid transit; the abnormality regresses after correction of the hyperthyroid state.

Hypothyroidism. In contrast to small bowel transit in hyperthyroidism, which is accelerated, small bowel transit is significantly slowed in hypothyroidism. [253] With adequate hormone replacement, transit may be normalized. Constipation develops in many hypothyroid patients, probably a result of colonic dysmotility. A manometric study in one patient revealed decreased amplitude of the small bowel contractions and an overall decreased motility index. [265] In severe hypothyroidism (i.e., myxedema), paralytic ileus and intestinal pseudoobstruction can occur. [266] , [267] , [268] , [269] and [270]

Hypoparathyroidism. The mechanism by which parathyroid hormone affects gastrointestinal motility is not known. However, calcium is essential for smooth muscle contraction, and hypocalcemia may impair gut contractile activity. Intestinal pseudoobstruction and malabsorption have been reported in association with small bowel dysmotility in hypoparathyroid subjects. [271] Barium studies reveal dilated loops of small bowel and a prolonged transit time. The symptoms are relieved by calcium administration.

Drug-induced changes in small intestinal motility. Many drugs profoundly affect gastrointestinal motility. Although the colon is usually recognized as the organ most susceptible to drug-induced dysmotility, the small bowel also is frequently affected. Phenothiazines and some antiparkinsonian drugs decrease colonic and small bowel motility and can cause constipation, colonic pseudoobstruction, and adynamic ileus. [272] , [273] , [274] and [275] Tricyclic antidepressants in particular are noted for causing iatrogenic ileus. The anticholinergic agents atropine and scopolamine and related belladonna alkaloids decrease intestinal tone as well as the amplitude and frequency of peristaltic contractions. [276] Opiate analgesics are well known to suppress motility throughout the gastrointestinal tract. Morphine enhances the amplitude of nonpropulsive small bowel contractions and markedly decreases propulsive contractions by its effects on μ-opiate receptors on intestinal muscle cells. [277] , [278] The upper small bowel is more prone to these effects than the ileum. The net effect is delayed small bowel transit. Morphine and nalbuphine significantly delay small bowel transit in comparison with placebo. [279] Loperamide, a synthetic opiate used to treat diarrhea, also reduces small bowel motility and delays transit time. This effect is antagonized by the concomitant administration of naloxone, which by itself has no effect on transit time. [280] , [281] Ketorolac, a nonsteroidal antiinflammatory agent used for postoperative pain, has significantly less effect on small bowel transit than morphine. [282] Whether ketorolac slows motility in comparison with controls is not known. Calcium channel antagonists, especially verapamil, cause constipation in up to 20% of patients. The small bowel transit time in subjects taking verapamil was unchanged from pretreatment values; transit through the colon was slowed, however, and this effect likely accounts for the constipation associated with this drug. [283] Clonidine, an a_2-adrenergic agent used as an antihypertensive agent, prolongs the orocecal transit of liquids; [284] however, a comprehensive dose-response study of the transit of solids did not show any significant effects on gastric small bowel or colonic transit in healthy individuals. [285] Clonidine has been used successfully to treat diabetic diarrhea; however, its therapeutic benefit may also result from its known action of increasing fluid and electrolyte absorption from the gut. [286] Drugs that stimulate small bowel motility include cisapride, octreotide, erythromycin, and cholinergic agonists. Erythromycin binds to motilin receptors on the smooth muscle of the gastric antrum and duodenum. Motilin, in physiological doses, induces interdigestive motor complexes in the upper gut. Erythromycin has been used to treat diabetic gastroparesis; the long-term beneficial effects are not as pronounced as the short-term effects. [287] Cholinergic agonists, such as bethanechol, augment the amplitude of small bowel contractions and overall motility. [288] Octreotide is a long-acting synthetic analog of somatostatin. In normal individuals, it increases the frequency of MMCs by significantly shortening the duration of phase II. [289] , [290] It was shown to be useful in the short term in a small group of patients with scleroderma by inducing phase III contractions and possibly reducing small bowel bacterial overgrowth. [167] When given after a meal, intravenous somatostatin interrupts the fed pattern of motility and induces bursts of propagated activity similar to phase III in health [291] and disease. [292] , [293] Octreotide retards small bowel transit in health when given before meals. [294] Cisapride is a nondopaminergic prokinetic agent that works by stimulating 5-HT_4 receptors to release acetylcholine in the myenteric plexus. It accelerates the transit of both liquids and solids through the small intestine [295] and has been found useful in treating patients with chronic intestinal pseudoobstruction. [152] Cisapride induces phase II–type activity in the fasted state and also augments small bowel motility after a meal. [296] , [297] The latter effect is generated after a small meal but not after a larger meal.

Celiac disease. Intestinal pseudoobstruction occurs in association with nontropical sprue. [298] , [299] and [300] Dilated loops of small bowel in which the passage of barium is delayed are observed radiographically. In one patient who underwent exploratory laparotomy with full-thickness jejunal biopsy, the nerves and muscle coats appeared normal on both light and electron microscopy. [300] Parenteral nutrition may be required during prolonged periods of pseudoobstruction.

Jejunal diverticulosis. Diverticula can occur anywhere in the small intestine, but they are most common in the jejunum. Like their counterparts in the colon, they represent herniations through the mesenteric side of the bowel and are usually acquired. [250] Jejunal diverticulosis is associated with many diseases, including scleroderma, celiac disease, Fabry disease, type II FVM, and Cronkhite-Canada syndrome. [301] Patients present with diarrhea, steatorrhea, weight loss, and megaloblastic anemia. Chronic intestinal pseudoobstruction occurs in jejunal diverticulosis and can mimic mechanical obstruction. [165] , [302] The concept that dysmotility causes diverticula and not vice versa is supported by a case report in which resection of the small bowel segment containing diverticula failed to relieve the patient's symptoms of pseudoobstruction. [10] In one patient, small bowel manometry showed that contractions can be antegrade, retrograde, or both simultaneously. [301] Histology of the bowel reflects the heterogenous causes of dysmotility (e.g., visceral neuropathy, visceral myopathy). [303]

Irradiation. Ionizing radiation damages all structures of the small intestine, including the mucosa, blood vessels, connective tissue, enteric nerves, and smooth muscle. Radiation damage to the bowel can be classified as acute or chronic.

Acute injury. Above a certain threshold dose of radiation to the abdomen, symptoms of nausea, vomiting, abdominal pain, and diarrhea are common. These abate soon after exposure is discontinued. Reversible changes in small bowel absorptive function have been considered the main cause of diarrhea. Small bowel dysmotility may play a significant role in acute radiation enteropathy. In dogs, a single large dose of radiation abolishes the interdigestive MMCs. [304] Smaller, fractionated doses, which are used for radiation treatment in humans, also result in dysmotility in the dog model. In the fasting state, manometric changes during exposure include an increased frequency of giant migrating contractions and retrograde contractions, with only a mild decrease in MMC cycle length. [305] These features revert to a normal pattern within a few days after exposure. Other studies, however, show decreased fasting and fed intestinal motility, particularly in the distal small

bowel, persisting up to 1 month after the last exposure. [306], [307] Motility studies have not been performed in humans in the acute phase of radiation sickness. One study demonstrated accelerated small bowel transit in a group of patients undergoing abdominal and pelvic irradiation in comparison with pretreatment values. [308] More than 75% of the patients had diarrhea during treatment.

Chronic injury. Delayed gastrointestinal complications may occur months, years, or even decades after radiation exposure. Both neuronal and muscular structures of the small bowel are affected. The resulting dysmotility leads to bacterial overgrowth, diarrhea, and malabsorption. Edema, atrophy, and fibrosis of smooth muscle fibers are characteristic histological findings. [309], [310], [311] and [312] The myenteric plexus can appear normal on routine staining; however, proliferation of the submucosal neurons with extension into the circular muscle coat has been observed. [311], [312] Latent gastrointestinal symptoms resulting from previous radiation damage have been attributed to altered gut motility. Recurrent episodes of intestinal pseudoobstruction were described in two patients many years after they received abdominal radiation. [312], [313] Small bowel contrast studies show dilated loops of bowel with air-fluid levels and a thickened bowel wall. Manometric evaluation in one patient revealed normal MMCs in the proximal duodenum, decreased amplitude and frequency of contractions in the distal duodenum, and an absence of peristaltic contractions in the jejunum. Subsequent histological examination of the bowel suggested smooth muscle damage as the cause of dysmotility. [312]

Diffuse lymphoid infiltration. Four cases of chronic intestinal pseudoobstruction in association with diffuse lymphoid infiltration of the small bowel have been described. [314] All four patients were women who presented with diarrhea at an early age. Histology revealed lymphocytic infiltration of the lamina propria, muscularis propria, and myenteric plexus. The infiltrates were shown to be polyclonal by immunochemical stains, reflecting a pseudolymphoma rather than a true neoplasm. The myenteric plexus appeared normal with routine and silver stains. Smooth muscle cells were noted to be absent in the areas of lymphoid infiltration.

Jejunoileal bypass. Recurrent episodes of intestinal pseudoobstruction can occur in patients after jejunoileal bypass for morbid obesity. [165], [315] Symptoms begin as early as 1 week postoperatively and consist of abdominal distention and pain, diarrhea, vomiting, and fever. Radiography reveals massively dilated small bowel loops and occasionally pneumatosis cystoides intestinalis. Bacterial overgrowth of the bypassed small intestine causes this enteropathy; small bowel cultures typically grow fecal flora and anaerobes similar to those described in blind loop syndrome. Over time, either the symptoms disappear or the frequency and severity of pseudoobstruction decrease significantly. Intermittent antibiotic therapy may benefit patients with chronic symptoms. In contrast to intestinal pseudoobstruction, mechanical obstruction secondary to ileal volvulus at the jejunoileal anastomotic site can occur and presents a difficult diagnostic challenge. [316] Contrast radiography may aid in establishing the correct diagnosis. Recurrent acute colonic pseudoobstruction also has been reported after jejunoileal bypass. [317], [318] The effect on intestinal motility of bypassing a long segment of small bowel is unknown. In patients undergoing reoperation for various metabolic or hepatic complications, the bypassed segment appears normal, and normal small bowel function is restored after reversal of the bypass. [319]

Anorexia nervosa and bulimia. Both anorexic and bulimic patients frequently report bloating and constipation. Delayed gastric emptying of solids is a well-established abnormality in these disorders. The orocecal transit is also modestly delayed, [320], [321] as is the whole-gut transit time, measured by radiopaque markers, in comparison with controls. [321] Additional transit and manometric studies are required in patients with these disorders.

Clinical Manifestations

The small intestine is essential for the absorption of nutrients; therefore, individuals cannot survive without parenteral nutrition if a long segment of small intestine functions poorly. Fortunately, the small intestine is usually the last organ in the gastrointestinal tract to be affected by severe dysmotility. Severe dysmotility of the esophagus, colon, and even the stomach occur much more frequently than dysmotility of the small intestine. Isolated severe small intestinal dysmotility is very unusual; small intestinal dysmotility is generally associated with dysmotility in other parts of the digestive tract. In this chapter, the discussion is limited to the small intestine. Dysmotility in other organs of the digestive tract is discussed elsewhere in this book.

With a few exceptions, most patients with small intestinal dysmotility have similar clinical manifestations regardless of the underlying causes. The spectrum of clinical manifestations varies widely (see Fig. 73-5). At one end, the patient may be asymptomatic, and at the other, the patient may have recurrent symptoms and signs of small intestinal obstruction, termed *chronic small intestinal pseudoobstruction*. Between these two ends of the spectrum, the patient may have recurrent symptoms of postprandial cramping, periumbilical, and epigastric abdominal pain, in addition to abdominal bloating, easy satiety, anorexia, weight loss, nausea, and vomiting. The symptoms are usually related to eating. Diarrhea can occur in patients with bacterial overgrowth and malabsorption. In severe cases, the patients have episodes of chronic intestinal pseudoobstruction syndrome. Plain abdominal x-ray films of patients with intestinal pseudoobstruction during exacerbations show multiple air-fluid levels and dilation of the small intestine. The prevalence and severity of recurrent obstructive episodes vary from patient to patient and from episode to episode in the same patient.

The findings on physical examination vary according to the severity of the symptoms. Patients may be cachectic and malnourished because they are unable to take in adequate nutrients, or they may have malabsorption as a consequence of bacterial overgrowth in the small intestine. The abdomen may be distended and mildly tender. The bowel sounds are inactive and infrequent in patients with smooth muscle dysfunction, but they are hyperactive and high-pitched in those with myenteric plexus dysfunction. Borborygmi may be detected in some cases. In less symptomatic patients, the abdominal examination findings may be normal. In those with chronic intestinal pseudoobstruction, during an obstructive episode, the abdominal examination findings may be indistinguishable from those of true mechanical obstruction.

As mentioned previously, patients with small intestinal dysmotility usually also have symptoms of dysmotility in other parts of the gastrointestinal tract. Therefore, patients with chronic intestinal pseudoobstruction may have symptoms and signs of multiple-organ dysmotility.

Extragastrointestinal manifestations may be detected in some patients, depending on the underlying disease. Megacystis and megaureter are common in type I FVM and CVM and may be associated with urinary retention and infection. Mydriasis, ptosis, and external ophthalmoplegia may be seen in certain forms of FVM, and ataxia, dysautonomia, and neurological symptoms in some forms of visceral neuropathy.

In the secondary forms of small intestinal dysmotility, patients may also have systemic manifestations of the underlying disease (e.g., scleroderma, muscular dystrophies, autonomic neuropathy). These are important clinical clues in patients whose underlying disease has not yet been diagnosed.

Complications

Malnutrition Malnutrition develops in patients with severe dysmotility of the small intestine as the result of an inadequate intake of food and of vomiting. Postprandial abdominal pain and bloating limit oral intake. Anemia may develop secondary to iron, folate, and vitamin B_{12} deficiency. Serum cholesterol, calcium, and albumin levels may be low. Supplemental formulas are useful to improve nutrition. In severe cases, long-term parenteral nutrition may be the only method to provide adequate nutrients, particularly to patients with familial or sporadic forms of hollow visceral myopathy.

Bacterial Overgrowth in the Small Intestine Bacterial overgrowth in the small intestine may complicate severe intestinal dysmotility. [30], [129], [147], [322] The small intestine of these patients is usually dilated and atonic, and they have malabsorption and steatorrhea, which causes additional weight loss. The diagnosis can be made by culturing intestinal aspirates for both aerobic and anaerobic bacteria. A number of different species are found, and the total bacterial concentration generally exceeds 10^5 aerobic organisms per milliliter or 10^3 anaerobic organisms per milliliter. Another approach to diagnosing bacterial overgrowth is the timed analysis of breath excretion of volatile metabolites produced by intralumenal bacteria. The measurement of both expired ^{14}C-carbon dioxide after the oral administration of ^{14}C-labeled substrates (e.g., D-xylose) and expired hydrogen after the administration of nonlabeled substrates has been used. This method is less accurate than culture of duodenal aspirates. [323] The patients may have macrocytic anemia secondary to vitamin B_{12} deficiency. Tetracycline, metronidazole, ciprofloxacin, or ampicillin can be used intermittently to treat intestinal bacterial overgrowth for 7 to 10 days, depending on the recurrence of diarrhea. In some cases, an antibiotic may have to be given for 1 week of every 3 to 4 weeks. Abdominal bloating may be relieved by antibiotics in some cases.

Pneumatosis Cystoides Intestinalis Pneumatosis cystoides intestinalis is a rare condition characterized by multiple, gas-filled cysts in the wall of the small and large intestine. Pneumoperitoneum may occur. Pneumatosis cystoides intestinalis can develop in patients with small intestinal dysmotility and dilation. In most cases, the condition is found incidentally on radiographs. The pathogenesis is unknown, and no specific treatment is available. It has been recommended that surgery be avoided except in the cases of a complication such as an abscess or peritonitis because surgery usually causes the patient's condition to deteriorate. [164]

Diagnostic Studies

The diagnostic studies for patients with small intestinal dysmotility are outlined in Table 73-7.

Blood Tests
Complete blood cell count
Blood chemistries, including blood sugar
Antinuclear antibody
Thyroid functions: triiodothyronine, thyroxin, thyroid-stimulating hormone
Serum creatine phosphokinase and isoenzymes
Hemagglutination and complement fixation for Chagas disease
Antineuronal nuclear antibody or antititer for paraneoplastic process
Radiologic Studies
Plain abdominal radiography in patients with abdominal distention
Radiopaque marker transit
Upper gastrointestinal and small bowel radiography
Enteroclysis or small bowel radiography with fluoroscopy
Barium enema
Scintigraphic gastric, small bowel, and colonic transit
Intravenous pyelography
Computed tomography of chest and abdomen
Other Studies
Skeletal muscle biopsy
Cultures of small bowel aspirates
Small intestinal or colonic manometry
Anorectal manometry and balloon expulsion
Histological studies of full-thickness biopsy specimens to examine the smooth muscle and myenteric plexus by special silver stain, immunohistochemistry, confocal microscopy of enteric plexuses

TABLE 73-7 Diagnostic Studies for Patients with Small Intestinal or Colonic Dysmotility

Blood Tests A complete blood cell count may reveal anemia and macrocytosis secondary to malnutrition and bacterial overgrowth. Blood chemistries also reflect malnutrition and malabsorption. Diabetic patients have hyperglycemia, and hypoparathyroid patients may have hypocalcemia. Patients with connective tissue disease are positive for antinuclear antibody or SCL-70. Patients with thyroid disease have altered serum levels of triiodothyronine, thyroxine, and thyroid-stimulating hormone. Patients with muscular dystrophy may have elevated levels of CPK and isoenzymes. The results of hemagglutination testing and complement fixation for Chagas disease may be positive in patients who have lived in Central or South America. Antineuronal nuclear antibody testing is required in patients with a history of smoking to exclude paraneoplastic pseudoobstruction. Determinations of blood lactate, pyruvate (signs of acidosis), CPK, ALT (muscle damage), and leukocyte thymidine phosphorylase are used to screen for mitochondrial cytopathy.

Radiologic Studies

Plain abdominal x-ray films. These are very useful in patients with abdominal distention and bloating because they may show gaseous distention of the gastrointestinal tract. Upper gastrointestinal and small bowel x-ray films may show lesions typical of certain diseases. Areas affected by severe dysmotility usually are dilated.

Enteroclysis. Enteroclysis, or small bowel enema, [324] is very useful to detect lesions in the small intestine and rule out mechanical obstruction. Experienced radiologists can identify structural lesions in the small intestine in up to 98% of patients with mechanical obstruction. Barium enema and intravenous pyelography may be useful in detecting abnormalities of the colon and urinary tract, respectively.

Whole-gut transit with radiopaque markers. Discrete barium-impregnated, polythene markers that move with the colonic contents have become simple but robust measures of transit. Hinton and colleagues [325] measured gastrointestinal transit times using small solids (2- to 5-mm polythene pellets) containing barium sulfate. These inert, radiopaque, nonabsorbable markers, which have a specific gravity similar to that of gut contents, were completely recoverable in the stool. Followed by either abdominal or stool radiography, the movement of such markers was taken to represent the transit of meal residues through the gut (Fig. 73-23). Of 25 normal male subjects, all but one had passed 80% of the markers by the fifth day after ingestion, but none had passed 80% by the end of the first day.

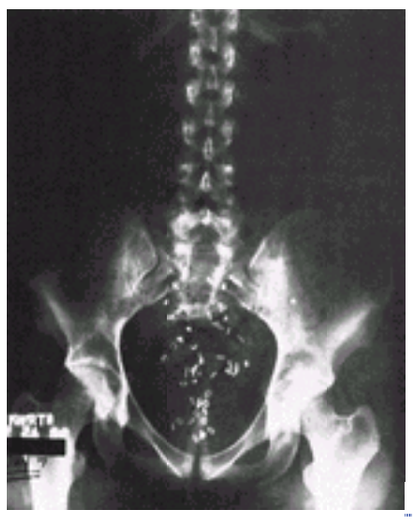

FIGURE 73-23. Radiograph of radiopaque markers as a measure of colon transit. Most of the markers are in the rectosigmoid colon of this patient.

Segmental colonic transit. The rate of disappearance of radiopaque markers from colonic segments was monitored with daily abdominal x-ray films obtained after the administration of a single dose of radiopaque markers. [326], [327] Right and left colonic, sigmoid, and rectal segments were defined simply by drawing lines between bony landmarks of the vertebral column and pelvis. Methods were then developed to calculate actual segmental transit times, rather than the rate of disappearance of markers from colonic segments. [328] Metcalf and colleagues [329] simplified this method so that only one x-ray film, obtained with a fast, high-kilovolt technique, was necessary; radiation exposure was therefore minimal. By this method, a fixed number of radiopaque markers were taken (24 per day) at the same time each day (9:00 AM arbitrarily) for 4 days. On day 5, again at the same time, an x-ray film was obtained. The method works on the assumption that a 24-hour sampling interval approximates continuous observation. In rapid transit, all the markers can be lost in the feces before radiography on day 4; conversely, in slow transit, all 96 markers may be present on the single radiograph. A film obtained on day 7 can then provide more information.

Radioactive isotope transit by scintigraphy. The transit of gamma ray–emitting radioactive isotopes through the colon can be quantified with a gamma camera linked to a computerized recording and processing system. [330] The isotope can be monitored for long periods without increasing radiation exposure; this is not possible with x-ray methods. The transit of solids and liquids can be measured simultaneously [331] if different isotopes are used to label liquid and solid bowel contents, and segmental regions of interest in the colon can be easily defined. A pH-sensitive polymer that dissolves at the pH of the ileum is used to coat capsules containing isotope-labeled beads. [330] With this approach, it has been possible to image the unprepared colon without the use of intubation; it is likely that such methods allow more “physiological” measurements of colonic transit. The net effects of fast or slow colonic transit are most clearly seen in the consistency of the stools. O’Donnell and colleagues [332] developed a 7-point scale for describing stool form and consistency, ranging from liquids to hard solids; they were able to relate stool consistency to the overall mouth-to-anus transit time. Faster transit was associated with looser stools. Stool form was then related specifically to colonic transit [331] after colonic transit times had been altered experimentally by rapid cecal infusions of fluid. When this noninvasive approach was used, [333] a good correlation was found between stool consistency and colonic transit; however, the correlation is clearly influenced by the extremes of transit and form, with little predictive value of stool that is not watery or extremely hard (Fig. 73-24).

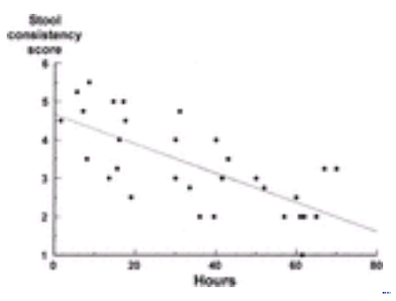

FIGURE 73-24. Correlation between mean colonic transit time (radiopaque markers) and physical appearance of stools (ranges from *1,* small and hard, to *6,* loose and semiliquid). (From ref. [333].)

Other Studies A biopsy of skeletal striated muscle may be required for patients suspected of having muscular dystrophy or mitochondrial cytopathy. Cultures of small bowel aspirates should be

obtained from patients with clinically suspected bacterial overgrowth (e.g., steatorrhea, bloating). Small intestinal manometry is indicated if no known underlying disease is present and the nature of the dysmotility (myopathy versus neuropathy) is unclear. If a full-thickness biopsy specimen of the dysfunctional part of the intestine is available, careful pathological examination for abnormalities in the smooth muscle or myenteric plexus with special trichrome and silver stains must be carried out. The most informative evaluation now includes assessment of the neurotransmitters by immunohistochemistry and morphologic assessment of the interstitial cells of Cajal with confocal microscopy.

Small Intestinal Manometry A low-compliance water infusion technique is the method most often used, but ambulatory studies with miniature transducers and radiotelemetry are also used. Impedance measurements have the potential to provide pressure profiles from very closely spaced sensors, although it is unclear whether these aid in the diagnosis. The small intestine has a unique pattern of motility. During fasting, a cyclic pattern of motility, the MMC, is seen. [334] The MMC is divided into three phases. Phase I is a quiescent period that lasts for about 15 to 30 minutes. Phase II, which lasts for about 60 minutes, is a period of intermittent contractions that increase in frequency with time until phase III is initiated. During phase III, intense contractions propagate aborally from the duodenum to the ileum. Phase III lasts for 4 to 10 minutes. After phase III, the small intestine become quiescent again (i.e., enters phase I) to start a new MMC cycle. This cyclic pattern continues until the subject eats. After eating, the MMC is replaced by frequent, intermittent, irregular contractions—the fed pattern. The fed pattern usually lasts for 2 to 6 hours, depending on the size of the meal; after it is complete, the fasting pattern, or MMC, returns. Small intestinal manometry has been used to study patients with various types of small intestinal dysmotility. [144, 145, 151, 167, 170, 186, 189, 195, 231, 322, 335, 336, 337, 338, 339, 340, 341] and [342] However, motility patterns are not diagnostic for any specific diseases. Many studies have been performed in patients with various clinical syndromes rather than specific diseases, which make the results difficult to interpret. Small intestinal motility studies have been performed in patients with small bowel bacterial overgrowth secondary to scleroderma, [322] chronic neuropathic intestinal pseudoobstruction, [144, 151] autonomic disorders, [343] FVM, [5, 17, 27, 337, 338] and [339] nonfamilial visceral myopathy, [30, 339] FVN, [58, 59] nonfamilial visceral (paraneoplastic) neuropathy, [219, 221] scleroderma, [167, 169] myotonic dystrophy, [231] and diabetes mellitus with gastroparesis. [187, 190, 341, 342] Three important patterns have been recognized from these studies.

Visceral myopathic pattern. In patients with smooth muscle dysfunction or degeneration, manometry demonstrates a decrease in the frequency and amplitude of contractions in the affected segment, generally during both the fast and fed periods. During fasting, the MMC is usually present but is diminished in amplitude. Myoelectric recordings may reveal abnormalities of the slow wave, such as variability in rate or rhythm, and abnormalities of propagation, such as loss of coupling or shortening of the length or even changes in the direction of propagation. These recordings are characteristic of diseases involving the smooth muscle, such as FVM, advanced scleroderma, dermatomyositis, the muscular dystrophies, and amyloidosis. [243, 246, 247, 343] Weston and associates [171] showed that in myopathic disorders, antral amplitudes are usually less than 40 mm Hg and duodenal amplitudes less than 10 mm Hg.

Visceral neuropathic pattern. Neurological disorders tend to produce disorganization and incoordination of motor activity. The MMC is often absent or abnormal. An abnormal rate of migration, as well as retrograde propagation of the activity front (phase III), also may be noted. Activity fronts may appear normal proximally and then become arrested or disappear in the more distal segments of the small intestine. In neurological disorders, the normal fed pattern may not replace the fasted pattern; MMC-like activity persists postprandially, and the frequency of antral contractions in the first hour is typically less than 1/min. [171] These abnormalities occur because the motility patterns are regulated by the ENS. The amplitude and frequency of contractions are usually normal when the inhibiting innervation and the coordinating function of the ENS are perturbed. Many of these abnormalities are seen best during sleep, when motor activity is quite stereotyped and normal phase II activity is suppressed. [344] The neurological disorders that have been described as producing manometric abnormalities include diabetes mellitus, Chagas disease, early scleroderma and amyloidosis, neurofibromatosis, paraneoplastic neuropathy, primary autonomic neuropathy, Parkinson disease, brain tumors, and multiple sclerosis. [58, 186, 189, 205, 251, 345, 346]

Mechanical obstruction pattern. Recurring episodes of intense contractions followed by periods of rest have been observed radiographically in the part of the intestine proximal to an obstruction. This phenomenon can be detected manometrically as clustered runs [151] or simultaneous prolonged contractions in conjunction with periods of intervening quiescence lasting longer than 1 minute (Fig. 73-25). [347, 348] Manometry is not the usual way to establish the diagnosis of mechanical obstruction, but these patterns should alert the physician to the possibility of mechanical obstruction and mandate careful small bowel barium radiography (e.g., enteroclysis).

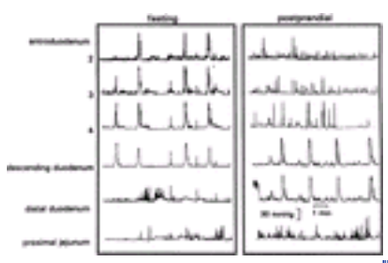

FIGURE 73-25. Gastroduodenal motility tracing demonstrating simultaneous prolonged contractions during fasting (**left**) and postprandial (**right**) phases. Contractions were present in the duodenum but not in the proximal jejunum (*bottom tracing*), a suggestion of mechanical obstruction in the distal duodenum or the proximal jejunum. (From Loftus EV, Farrugia G, Donohue JH, Camilleri M. Duodenal obstruction: diagnosis by gastroduodenal manometry. Mayo Clin Proc 1997;72:130.)

Overall, small intestinal manometry lacks the specificity to diagnose the underlying disease other than suggesting the pathophysiological process. The relative merits of a laboratory-based study [349] and ambulatory manometry [350] have been discussed in the literature. From a diagnostic standpoint, the latter does not significantly enhance the results of the shorter laboratory-based studies, which have the added advantage of examining antral motor function more accurately; however, the ambulatory study is more practical and probably less costly.

Colonic Manometry Pressure waves recorded manometrically are generally equated with contractile forces. Colonic contractions are often irregular, of varying frequency and amplitude, and subject to the emotional state of the subject, the type of meal eaten, and the composition of chyme entering the colon from the ileum. [351, 352, 353, 354] and [355] The colon is often “quiet” or exhibits isolated contractions (perhaps reflecting the dominant inhibitory control). At times, pressure waves are recorded in continuous bursts ranging in duration from 10 to 30 minutes, [353] with a dominant frequency of approximately six contractions per minute in the right side of the colon and approximately three to seven per minute in the distal colon. Phasic colonic contractions are stimulated by meals, beginning 20 to 30 minutes after a meal and lasting for up to 3 hours; sometimes, a second peak of activity occurs at approximately 70 to 90 minutes postprandially. [351, 352, 353, 354] and [355] This response to eating (gastrocolic reflex) remains after gastrectomy and after vagotomy. [356] If the stomach is intact, gastric distention and chemical stimulation by nutrients elicits a comparable response; lipids and a caloric content of more than 500 kcal are the most potent stimuli. [1, 357] Control of the colonic response to eating involves neural and possibly hormonal mechanisms. The part of the response mediated by gastric mechanoreceptors is very sensitive to blockade by atropine, but the part originating in the small intestine is only partially under muscarinic control. Cholecystokinin (CCK), which is normally secreted after meals, has been thought to play a role in the response because high blood concentrations of CCK stimulate contractile activity in both the small and large intestine. However, loxiglumide, a CCK A receptor antagonist, did not block the gastrocolic response to food. [358] Low-amplitude irregular phasic activity does not propagate and probably equates with segmentation; on the other hand, high-pressure, propagating waves are the equivalent of peristalsis. Isolated, high-amplitude (~200 mg Hg) peristaltic contractions (Fig. 73-26), propagating at about 1 cm/s over long distances, are thought to be important for transit or “mass movement” in the colon. These occur infrequently (mean of six per day in healthy humans) and are most frequent after waking and meals. [359] They can be reliably induced by instilling irritants like bisacodyl [360] or fatty acids [361] into the colon.

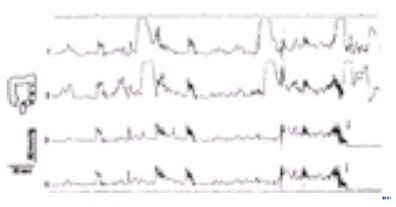

FIGURE 73-26. High-pressure waves recorded sequentially from four sites in the human colon. These contractions (giant migrating contractions) are thought to correspond to mass movements of colonic content. Their frequency is reduced in patients with constipation. (From ref. [359].)

An important variant of manometry uses the electromechanical barostat. [1, 362, 363] and [364] Originally developed to detect volume changes in the gastric fundus, the methodology is well suited for identifying relaxation in any hollow viscus. The 8- to 10-cm-long polyethylene bag used in humans is of large volume and “infinite compliance” until it reaches a volume of 500 to 600 mL. Air is introduced or withdrawn from the bag by a servomechanism to maintain a preset low pressure in the bag. Changes in volume are reflections of changes in wall tone. Basal tone in the proximal and distal colon exhibits rhythmic fluctuations, [1] and food induces an immediate and prolonged increase in tone. The postprandial response is inhibited by 5-HT $_3$ blockers [365] and modified by hyperglycemia. [366] The rectum also shows a tonic response to eating, [367] one that is impaired in patients with chronic slow transit constipation. [368] The rectum also demonstrates prominent relaxation during distention. [369, 370] and [371]

Differential Diagnosis between Chronic Intestinal Pseudoobstruction and Mechanical Obstruction

In patients with severe dysmotility of the small intestine, chronic intestinal pseudoobstruction may develop. Partial small bowel obstruction from adhesions, tumors, intussusception, or stricture can mimic chronic intestinal pseudoobstruction. The features listed in Table 73-8 can be used to differentiate between these two problems. Enteroclysis or careful small bowel radiography with fluoroscopy is probably the most helpful way to differentiate chronic intestinal pseudoobstruction from mechanical obstruction. In our experience, however, enteroclysis may not conclusively differentiate chronic intestinal pseudoobstruction from mechanical obstruction in some patients. Manometry proved useful in one series; [348] however, in most cases, exploratory laparoscopy may be necessary to rule out an obstructing lesion.

TABLE 73-8 Differentiation of Chronic Intestinal Pseudoobstruction from True Mechanical Obstruction

Treatment

Drug Therapy to Improve Small Intestinal Motility Theoretically, it is difficult to envision a drug that can either stimulate damaged muscle to contract effectively or normalize the coordinating functions of a damaged myenteric plexus. Drugs that stimulate intestinal motility in normal subjects (e.g., bethanechol, neostigmine, metoclopramide, erythromycin, tegaserod, prucalopride) have no beneficial effects in patients with small intestinal dysmotility. No information is available on the use of domperidone for small bowel dysmotility. Cisapride decreased the transit time of meal through the small bowel in normal subjects [295] and in patients with neuropathic forms of chronic intestinal dysmotility. [372], [373], [374] and [375] In a small, short-term study, octreotide, a somatostatin analog, stimulated intestinal motility, possibly reduced bacterial overgrowth, and relieved abdominal symptoms in patients with scleroderma. [167] Other open-label studies confirmed the long-term effectiveness of octreotide with erythromycin in the treatment of chronic intestinal pseudoobstruction. [293]

Symptomatic and Supportive Treatments Abdominal pain, bloating, nausea, and vomiting in patients with small intestinal dysmotility are often related to eating. Most of these symptoms can be minimized by manipulating the size, nature, and frequency of meals. The important point to keep in mind is to provide sufficient calories without overloading the inefficient bowel. A rule of thumb is to give approximately 25 cal/kg of the patient's ideal body weight per day. Adult patients should consume 1500 to 1800 cal/d divided into three or four equal feedings. At least half of the calories should come from supplemental formulas such as Ensure because a liquid meal empties faster from the stomach and probably progresses more easily through the small bowel than a solid meal. These formulas are lactose free and contain the daily requirements of vitamins and minerals. When patients still feel full several hours after the first meal, it is important that they not force themselves to eat subsequent meals to avoid aggravating their symptoms, and they that restrict their oral intake to fluids for the rest of the day. Patients with vagal dysfunction appear to respond less than those with myopathic dysfunction or no autonomic dysfunction. [374] In the long term, 60% appear to benefit from cisapride, and this benefit persists for up to 1 year. [375] In patients with chronic intestinal pseudoobstruction, recurrent symptoms and signs of intestinal pseudoobstruction may occur despite implementation of the previously described regimens. In these situations, nasogastric suction and intravenous fluids are needed when obstructive symptoms develop. When obstructive symptoms and pain persist or occur several times a week despite dietary manipulation, long-term parenteral nutrition is the only treatment that will relieve symptoms and improve nutrition. Abdominal pain unrelated to eating is uncommon in patients with small intestinal dysmotility, and if a patient has pain predominantly unrelated to meals, questions should be raised about the diagnosis. During episodes of obstruction, parenteral administration of narcotics such as morphine or meperidine may be required. Long-term narcotic use should be discouraged because addiction is possible, and narcotics can further disturb gastrointestinal motility. Constipation is common in patients who also have colonic involvement. It is important to make certain that the patient has a good bowel movement at least once every few days because constipation tends to increase the symptoms of intestinal dysmotility. We prescribe 30 to 60 mL (or two tablets three times daily) of Milk of Magnesia per day. Enemas may be used if the patient has no bowel movement for 3 days. Those with severe small intestinal dysmotility should avoid bulk-forming laxatives because they increase the load on an inefficient intestine and exacerbate symptoms.

Treatment of Secondary Causes A few types of secondary small intestinal dysmotility, such as those associated with myxedema, celiac sprue, and drug-induced dysmotility, can be treated with thyroid replacement, a gluten-free diet, and discontinuation of the offending drug, respectively. No specific treatments are available for most of the secondary types of small intestinal dysmotility.

Surgical Treatment Patients with dysmotility limited to short segments of the small intestine, such as those with megaduodenum, have a better prognosis than those with dysmotility throughout the length of the bowel because the dysfunctional segment can be resected or bypassed. [376] Megaduodenum, which is commonly seen in FVM type I and scleroderma, has been drained with a side-to-side duodenojejunostomy, which usually relieves symptoms. In some patients with a massively dilated duodenum, a side-to-side duodenojejunostomy may be inadequate to drain the duodenum. In these cases, resecting as much of the duodenum as possible with preservation of the papilla of Vater and anastomosing the opened jejunum to the cut edge of the duodenum to create a small conduit may be required as treatment. [13], [377] For patients with long segments of small intestinal dysmotility (i.e., >1.2 m), no effective surgical treatment is available. Any unnecessary surgery should be avoided in these cases because it can create adhesions and additional difficulties. Venting decompression by percutaneous jejunostomy or minilaparotomy or laparoscopy relieves symptoms and reduces the rate of hospitalization for recurrent exacerbations of pseudoobstruction. [378], [379]

Small bowel transplantation. Small bowel transplantation has become a reality with the introduction of effective immunosuppression (chiefly tacrolimus and mycophenolate); however, prognosis, survival, and complications seem quite different in children, in whom efficacy is greater than in adults. In a review of the world experience for the period from 1985 to 1997, the survival of patients and grafts was about 60%, and about three fourths of the survivors were free of parenteral nutritional support. [380] However, adults seem to fare less well; the actuarial survival rates of patients and grafts were 48% and 36%, respectively, at 3 years. [381] In reaching a decision about transplantation, it is important to recall that the 3-year survival of patients on home total parenteral nutrition (TPN) is 85%. [382] Several factors limit the success of small bowel transplantation and may result in death: infection (including infection with cytomegalovirus and Epstein-Barr virus, which accounts for approximately 50% of deaths); rejection (approximately 8% of deaths); technical problems; and lymphoproliferative disease (20%–40% of deaths in different programs). The latter is particularly linked to Epstein-Barr virus infection, the use of OKT3 immunosuppression, and steroids. [383], [384] In general, small bowel transplantation is restricted to life-saving situations, such as necrotizing enterocolitis, intestinal failure (short bowel syndrome) in patients who cannot be maintained on TPN, and rarely chronic intestinal pseudoobstruction in young patients who have TPN-related or other liver failure and lack intravenous access for TPN. In the future, gastroenterologists may be involved in the management of patients after small bowel transplantation. Recent observations that are likely to affect this practice include the following: (1) Zoom videoendoscopy is capable of showing the enlarged crypt areas and villus blunting or flattening that correlate with histological evidence of rejection. [385] (2) Severe rejection (mucosal necrosis with loss of villi) is of grave prognostic significance (11% graft, 18% patient survival) [386] and, pending newer immunosuppressive regimens, may be an indication for removal of the graft. (3) Living-related small bowel transplantation is technically feasible and has been reported to be followed by complete functional adaptation of the graft. [387] It is clear that survival after this formidable procedure depends on the experience of the entire transplant team and center, aggressive treatment of infection and rejection, and the rate of mortal complications, including lymphoproliferative disease, remains high. [388] The use of tacrolimus in immunosuppression is associated with a fivefold increase in lymphoproliferative disease relative to cyclosporine in children receiving liver transplants. [389] Molecular methods to screen for Epstein-Barr virus infection, improved immunosuppression, and possibly bone marrow transplantation to enhance chimerism may change the biology or prevalence of lymphoproliferative complications in the future.

COLONIC DYSMOTILITY (PSEUDOOBSTRUCTION) AND MEGACOLON

Interstitial Cells of Cajal in Maldevelopment and Acquired Diseases of the Colon

The significance of the interstitial cells of Cajal (ICCs) was established during the last 25 years through the careful histological and electrophysiological observations of Faussone-Pellegrini and colleagues [77], [78] and Thuneberg; [79] the latter established the candidacy of ICCs as the intestinal pacemaker cells. The protooncogene c-kit encodes a tyrosine kinase receptor that facilitates the development of the ICCs. [390] Furthermore, mice with mutations in the dominant white spotting (W) locus, which have cellular defects in hematopoiesis, melanogenesis, and gametogenesis as a result of mutations in the c-kit gene, also lack the network of ICCs and intestinal pacemaker activity. [391] In the last decade, the role of ICCs has become more clearly understood through studies of a mutant strain of mice (*w/w v*) that lack c-kit–positive cells and in which gastrointestinal dilation and failure of peristalsis develop. [392], [393], [394], [395] and [396]

ICCs are typically surrounded by collagen fibers and form close contacts (Fig. 73-27) with smooth muscle cells. [392] These gap junctions with smooth muscle cells allow transmission of the pacemaking activity that reflects the spontaneous oscillation in resting membrane potentials, which is unaffected by 10^{-6}-M verapamil, an L-type calcium channel blocker. [392] When studied in short-term primary culture, the cells assume a triangular shape with three to four branches that are in apposition with co-cultured smooth muscle cells (Fig. 73-28). Huizinga, Ward, Sanders, and their colleagues [393], [394] and [395] have characterized the expression profiles of pacemaker ICCs isolated from the murine small intestine and ICCs and smooth muscle cells involved in neurotransmission from the gastric fundus. All cell types express muscarinic receptor types M_2 and M_3, neurokinin receptors NK_1 and NK_3, and the inhibitor receptor vasoactive intestinal peptide 1 (VIP_1). [395]

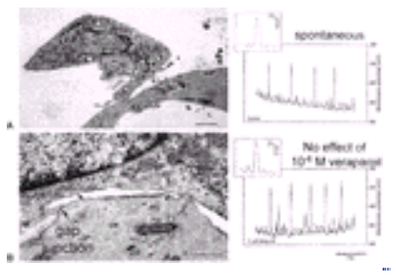

FIGURE 73-27. Gap junctions between interstitial cells of Cajal and smooth muscle cells; note the spontaneous electrical oscillations of the resting membrane potential of the interstitial cells of Cajal and the lack of inhibition by the L-type calcium channel blocker verapamil. (From ref. [392].)

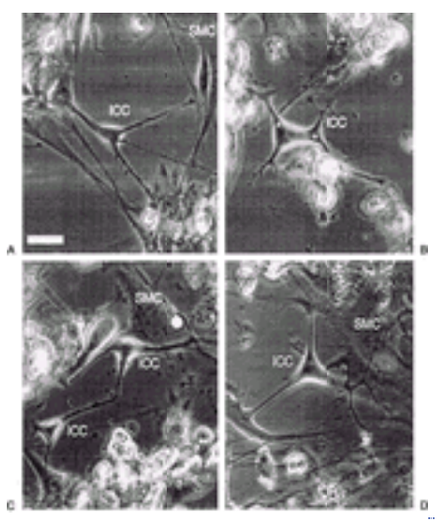

FIGURE 73-28. In short-term culture, interstitial cells of Cajal assume a triangular shape and have three to four branches that establish contact with cultured smooth muscle cells. (From ref. [392].)

In clinical studies, a relative deficiency of c-kit–positive cells has been reported in Hirschsprung disease, chronic intestinal pseudoobstruction, [396], [397] gastrointestinal stromal tumors, and multiple gastrointestinal autonomic tumors. [398], [399]

Delayed maturation or maldevelopment of ICCs is well documented (Fig. 73-29). [400] At 1 month of age, a child with chronic colonic pseudoobstruction underwent biopsy of the affected colon, which showed no peristaltic activity and no c-kit immunoreactivity within the circular muscle layer or the submuscular layer (see Fig. 73-29) but a normal ICC population in the myenteric plexus. At 6 months of age, the peristaltic activity was normal, and the ICCs were fully developed in all layers of a full-thickness biopsy specimen of the colon. This careful case study illustrates several points: (1) the concept of postnatal maturation of ICCs, (2) the importance of the conservative management of children with neonatal megacolon or pseudoobstruction, and (3) the importance of ICCs to the overall peristaltic function of the colon. Little is known about the transmitters expressed by ICCs in disease. However, immunoreactivity to NK1 has been detected in ICCs and may be used as a marker of the ICCs at the deep muscular plexus; these cells may participate in the actions exerted by tachykinins on muscle cells. [401]

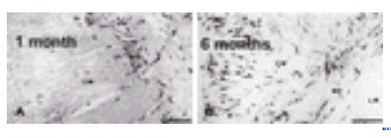

FIGURE 73-29. Full-thickness biopsy specimens from the colon of an infant with neonatal chronic colonic pseudoobstruction demonstrating delayed maturation of interstitial cells of Cajal at 1 month and subsequent expression of the cells at 6 months of age. (From ref. [400].)

Megacolon in an adult has also been associated with abnormal morphology and ultrastructure of ICCs. Faussone-Pellegrini and colleagues [402] demonstrated ICCs with several branches in the dilated transverse colon, but abnormal ICCs with a paucity of mitochondria, filaments, and caveolae in the nondilated descending colon.

In patients with acquired slow transit constipation not associated with colonic dilation, the number of ICCs in the different layers of the sigmoid colon is lower (Fig. 73-30) than in controls (specimens resected for other indications). Moreover, the confocal images show irregular surface markings and a paucity of branches of the cells (Fig. 73-31). [403] Staining of myenteric plexus cells with c-kit is reduced, consistent with a reduction of ICCs or their tyrosine kinase content.

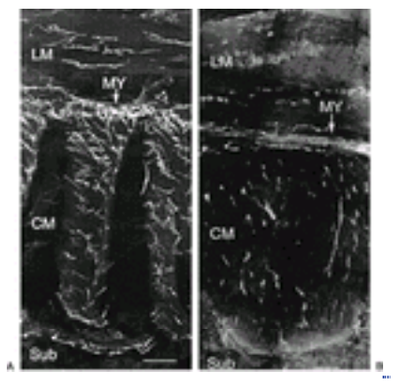

FIGURE 73-30. Distribution of interstitial cells of Cajal in whole transverse mounts of a normal-appearing disease control section of the sigmoid colon (**A**) and the sigmoid colon of a patient with slow transit constipation (**B**). (From ref. 403 .)

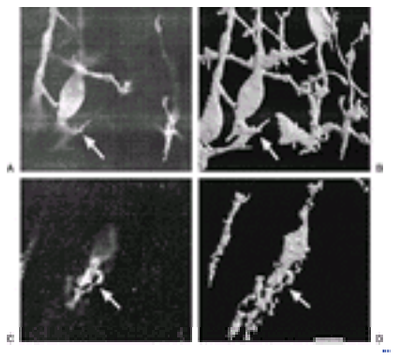

FIGURE 73-31. High-magnification confocal microscopy of the interstitial cells of Cajal in human sigmoid colon. **A** and **C** are single slices; **B** and **D** are reconstructions of 20 consecutive single slices. **A** and **B** are from a healthyappearing disease control colon; note the multiple fine processes and the network of interconnecting interstitial cells of Cajal. **C** and **D** are from a patient with slow transit constipation. Note the irregular markings and loss of fine processes. *Bar* = 10 µm. (From ref. 403 .)

Syndromes Generally Attributed to Disorders of Colonic Motility

Constipation Patients consider themselves to be constipated when they pass stools more infrequently, require more effort for passage (“straining”), or experience more pain or discomfort during passage than they think appropriate. Difficult evacuation of feces, especially when the consistency of stools is softer than normal, is more likely to be caused by disorders of the pelvic floor or anorectum than by slow colonic transit. Disorders of the pelvic floor are disturbances of evacuation (dyschezia, obstructed defecation) and are not be considered here (see Chapter 43 and Chapter 92). Slow transit through some or all segments of the colon leads to the formation of hard fecal masses that are passed infrequently and often with great difficulty. This condition, slow transit constipation (STC), should be considered as separate from IBS and should be diagnosed only if abdominal pain is not a prominent feature. On the other hand, STC certainly merges with IBS when pain, abdominal distention, excessive rectal mucus, and intermittent episodes of frequent, looser stools also occur (see Chapter 86). STC is considered in the next section.

Slow Transit Constipation Figure 73-23 shows radiopaque markers retained predominantly in the distal colon (rectosigmoid segment) 96 hours after ingestion. 325 This technique is simple, inexpensive, and of proven value in the documentation of STC. 325 , 326 , 327 , 328 and 329 Gamma scintigraphy 330 offers another means of testing, which can be made shorter (24 or 48 hours). Whatever method is used, the implications for therapy are potentially important. Most patients with STC respond to laxative programs, suppositories, or enemas. Resective surgery of the colon (usually subtotal colectomy with ileorectostomy) is occasionally necessary for severe STC. When tests of pelvic floor function identify a clinically relevant abnormality, colectomy is not indicated, even in the presence of slow transit. Retraining programs for the pelvic floor should first be used to correct the defect in expulsion, after which transit can be retested.

Manometry. In STC, the normal high-amplitude pressure (peristaltic) waves in the colon are reduced. Hirschsprung disease, or congenital megacolon (see section “ Hirschsprung Disease: Congenital Megacolon”), is a special case in which the pathophysiological abnormality is absence of relaxation of the internal anal sphincters, manifested by absence of the rectoanal inhibitor reflex.

Myoelectric patterns. Myoelectric patterns have confirmed the concept of “paradoxical colonic motility”: patients with diarrheal disease often have a colon that demonstrates relatively little overall myoelectric activity, whereas those with constipation often have very active motor patterns that favor “holding up” contents.

Histopathology. Many patients with STC severe enough to warrant subtotal colectomy display histological changes in the ENS of the resected colon on silver staining; Table 73-9 summarizes the information derived from a number of studies in the literature. 403 , 404 , 405 , 406 , 407 , 408 , 409 , 410 , 411 , 412 and 413 In general, it appears that reduced substance P and increased nitrergic neurons are associated with constipation that leads to surgical resection of the colon. In Parkinson disease, evidence is found of a reduction in the dopamine content of the myenteric plexus and a significant reduction in the dopamine-positive neurons of whole mounts of the plexus. 414

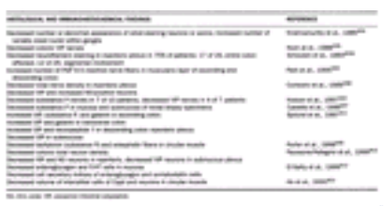

TABLE 73-9 Colonic Neuropathology in Slow Transit Constipation

Ultrastructural quantitative studies have demonstrated abnormalities of the morphology and numbers of ICCs. 403 It is assumed, therefore, that these patients have a morphologic abnormality of the neural control of colonic motility. These clinical and histopathological features appear to merge with the syndromes of intestinal pseudoobstruction (see section “ Colonic Pseudoobstruction and Megacolon”). The treatment of constipation is addressed in Chapter 43.

Colonic Pseudoobstruction and Megacolon

Megacolon (Table 73-10) and *megarectum* are descriptive terms, without etiologic or pathophysiological implications. *Megacolon* has been defined as a diameter of the rectosigmoid region or descending colon, on x-ray film, of more than 6.5 cm, of the ascending colon of more than 8 cm, or of the cecum of more than 12 cm. 415 Megacolon can be caused by aganglionosis (Hirschsprung disease), may be idiopathic (complicating chronic constipation of any cause), or may be a manifestation of diffuse gastrointestinal dysmotility (intestinal pseudoobstruction). *Acute megacolon* is often designated as *Ogilvie syndrome*. Rectal dilation, or *megarectum,* may be an isolated finding or, more often, associated with megacolon or simply prolonged chronic constipation, particularly in children with functional fecal retention. Note that megacolon does not include simple elongation of the colon (dolichocolon).

Congenital Megacolon (Hirschsprung Disease)
"Classical" type
Short segment
Ultrashort segment
Total colonic aganglionosis, zonal loss of ganglia, and other variants
Acquired Megacolon (Associated with Constipation)
Idiopathic
In children (associated with encopresis)
Acute form in adults (Ogilvie syndrome)
Secondary to neurological diseases
Chagas disease
Parkinson disease and central nervous system dysfunction; myotonic dystrophy
Diabetic neuropathy; others (ganglioneuromatosis, familial autonomic dysfunction)
Intestinal pseudoobstruction ("neurogenic" forms)
Secondary to diseases of intestinal smooth muscle
Scleroderma and other "collagen diseases"; amyloidosis
Intestinal pseudoobstruction ("myogenic" forms)
Secondary to metabolic diseases
Hypothyroidism, hypokalemia, porphyria, pheochromocytoma (with ganglioneuromatosis)
Drugs
Mechanical obstruction

TABLE 73-10 Clinical Classification of Megacolon

In *congenital megacolon* (Hirschsprung disease), colonic dilation results from functional obstruction (usually of the rectum) caused by a congenital absence of the intramural neural plexuses that mediate relaxation (aganglionosis). This leads to a narrow segment of the large intestine—that is, one that fails to relax.

Acquired megacolon can complicate any of the many causes of constipation, and it can be assumed that megacolon is acquired when it can be ascertained that colonic dilation was not present at some earlier examination. A common background for acquired megacolon is colonic inertia, which occurs frequently at both extremes of life. In children, this form can be confused with the congenital condition.

Worldwide, infection with *Trypanosoma cruzi* (Chagas disease) is the most common form of acquired megacolon. In this condition, the dilated segment of colon is abnormal because of destruction of the ENS by the neurotoxin of the organism. Although infection was originally confined to the South American continent, it is now estimated that there are 350,000 seropositive persons in the United States; among these, one third are thought to have chronic Chagas disease. [416] Some patients acquire megacolon as part of a generalized intestinal pseudoobstruction.

When colonic pseudoobstruction is acute and associated with another medical condition (abdominal or orthopedic surgery, spinal cord injury, serious cardiovascular or other medical problems), the term *Ogilvie syndrome* is often applied. Toxic megacolon is a life-threatening complication of inflammatory bowel disease and infectious colitides (see Chapter 83 and Chapter 88).

Hirschsprung Disease: Congenital Megacolon

Pathophysiology. Aganglionosis is caused by the arrested migration of cells caudad from the neural crest; these are the cells destined to develop as the intramural plexuses of the gut. In Hirschsprung disease, the aganglionic segment always extends from the internal anal sphincter for a variable distance proximally, but in most instances it remains *within the rectum and sigmoid colon*. The involvement of very short segments, affecting only the anal sphincters, has also been described. The aganglionic segment is permanently contracted, causing dilation proximally. Longer aganglionic segments occur in fewer than 20% of individuals; involvement of the entire colon is infrequent, and aganglionosis extending proximally into the small intestine is rare. Thus, the hallmark of diagnosis is the absence of ganglion cells from the myenteric and submucosal plexuses, as seen on a full-thickness or suction (mucosal-submucosal) biopsy specimen of the rectum. Proximal contents fail to enter the unrelaxed aganglionic segment. Morphologically, ganglion cells are absent from the narrowed segment and for a short distance (usually 1–5 cm) into the dilated segment. In contrast, the nerve fibers are hypertrophic, with abundant, thickened bundles. Specific stains for acetylcholinesterase highlight the abnormal morphology. Adrenergic denervation of the dilated segment is another inconsistent finding, as is a decreased supply of peptidergic nerves (containing VIP, substance P, enkephalins, and other peptides). The most characteristic functional abnormality of aganglionosis is a failure of the internal anal sphincters to relax following rectal distention. [417] Transient distention of a balloon in the rectum causes the intralumenal pressure at the level of the internal anal sphincter to drop; the drop is often accompanied by a reflex contraction of the external sphincter. Up to 20% of normal children may have a falsely absent reflex, especially if they are premature or of low birth weight, but *a positive response is strong evidence against Hirschsprung disease*.

Incidence and genetics. The defect occurs once in each 5000 live births and is in some cases familial, with an overall incidence of 3.6% among siblings of all index cases. [418] Because the disease was highly lethal until the introduction of curative surgery in the 1950s, accurate assessment of the incidence in the offspring of successfully treated patients is incomplete. Consanguinity of parents is exceptional, and the condition is reported to be discordant in dizygotic twins and concordant in monozygotes. The association of congenital aganglionosis of the colon with Down syndrome is ten times more frequent than would be expected by chance; [418] approximately 2% of patients with congenital megacolon have Down syndrome. A number of other congenital anomalies have been reported: hydrocephalus, ventricular septal defect, cystic deformities and agenesis of the kidney, cryptorchidism, diverticulum of the urinary bladder, imperforate anus, Meckel diverticulum, hypoplastic uterus, polyposis of the colon, ependymoma of the fourth ventricle, the Laurence-Moon-Bardet-Biedl syndrome, and congenital central hypoventilation syndrome (Ondine curse). The genetic disorders resulting in disordered development of the neural crest in Hirschsprung disease are discussed in detail above (see section "Ontogeny of the Enteric Nervous System").

Clinical features. Hirschsprung disease should be suspected shortly after birth when the infant passes little meconium and the abdomen is distended. Digital examination of the rectum, insertion of a rectal tube, or administration of a small enema causes retained fecal material to gush forth, with apparent relief of the symptoms. However, the respite is often short-lived; signs of partial intestinal obstruction return, with persistent vomiting and distention as the major features. In about 20% of patients, diarrhea persists; it is caused by pseudomembranous enterocolitis, which develops as a complication of the obstruction. Later in life, the presentation is often less dramatic and may not mimic an acute intestinal obstruction. Severe constipation and recurrent fecal impactions are more common. Children occasionally show evidence of anemia, malnutrition, and even hypoproteinemia resulting from protein-losing enteropathy; their resistance to infection can also be impaired. Although difficulties develop in most children before the second month of life, very-short-segment aganglionosis may not cause severe symptoms until after infancy.

Differential diagnosis. Hirschsprung disease must be distinguished in the neonate from other developmental causes of intestinal obstruction, such as atresias and imperforate anus. Later in life, acquired (secondary) megacolon is the other major consideration. The diagnosis of congenital megacolon is usually not difficult beyond the immediate neonatal period, and the better diagnostic methods now available allow a positive diagnosis in most cases. Obstipation, with infrequent spontaneous passage of stool, dates from infancy, and the rectal examination reveals an empty ampulla. Overflow incontinence is not a feature of Hirschsprung disease. A barium enema x-ray film (Fig. 73-32) confirms the diagnosis if the characteristic transition from the narrowed, distal rectum or rectosigmoid to the dilated proximal colon is seen. However, when the aganglionic segment is very short, a narrowed segment is not seen radiologically. In patients with acquired megacolon, encopresis is common, dilation extends all the way to the anus, and a narrowed zone is not seen.

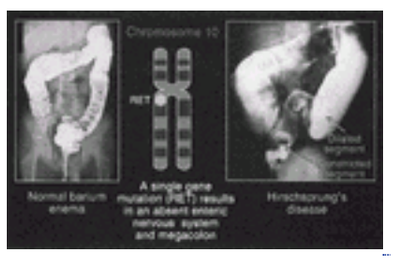

FIGURE 73-32. Barium enema in a normal child contrasted with a study showing megacolon and a narrow segment affected with Hirschsprung disease. The genetic defect involves a mutation of the RET protooncogene on chromosome 10q11.2.

Proctosigmoidoscopy reveals a normal but empty rectum. The dilated proximal bowel, if within range of the scope, is easily traversed except for abundant feces in the lumen; occasionally, stercoral ulcers are noted. The key findings are the empty lower segment and the absence of evidence of organic obstruction. The diagnosis is best substantiated by a full-thickness biopsy of the rectum. The presence of normal numbers of ganglion cells excludes the diagnosis. Mucosal suction biopsy, satisfactory in many instances, is the initial procedure of choice because it is performed easily and requires no anesthesia. If the depth of the examination is sufficient to show the presence of ganglia in the Meissner (submucosal) plexus, the classical form of Hirschsprung disease is excluded. However, the absence of ganglion cells does not establish the diagnosis, and a full-thickness biopsy specimen should be obtained at least 3 cm proximal to the pectinate line. A diminution or absence of ganglion cells distal to this point is difficult to interpret. Careful histology proximal to the internal sphincter reveals that myenteric ganglia may be absent from normal infants over a distance of 4 to 5 mm in this segment; ganglia may be absent from the deep and superficial submucosal layers for even longer distances. Immunohistochemical techniques can highlight the morphologic abnormalities,

showing an abundance of hyperplastic axons but an absence of ganglion cells. Several approaches, in which antibodies to acetylcholinesterase, neuron-specific enolase, neurofilament, and neuropeptides are used, have been described. [419], [420] Physiological tests complement the diagnosis in doubtful cases, and they may be crucial when the aganglionic segment is very short. Such cases are less easily detected on x-ray films and are also likely to be missed by biopsy. The most important pathophysiological test is the response of the anal sphincters to distention of the rectum. [417] In contrast to the internal sphincter in normal individuals and in patients with acquired megacolon, the internal sphincter in patients with congenital aganglionosis fails to relax (or contracts even further) after the rectum is distended. The most common cause of a false-positive test result is a capacious rectum in constipation or megarectum; under these circumstances, distension of the rectal balloon does not stimulate the reflex. Therefore, an enlarged rectum should be excluded before Hirschsprung disease is diagnosed.

Treatment of congenital megacolon. Definitive surgical cure is the treatment of choice. Preliminary decompression by colostomy is still sometimes necessary to relieve obstruction, or it may be necessary in some infants when it is decided to postpone definitive surgery. However, the goal should be early diagnosis and a one-stage surgical approach. The main goals are to establish regular and spontaneous defecation, to maintain normal continence, and not to interfere with sexual potency. The surgical procedure should cause essentially no mortality and minimal morbidity. A number of different operations have been used to remove successfully or to counterbalance the obstructing effect of the aganglionic segment. Long-term results are good in the great majority of patients, but 10% to 20% have residual problems, usually with fecal soiling. [421]

Variants of congenital megacolon. The spectrum of Hirschsprung disease has widened considerably. Patients with a compatible clinical picture may have an ultrashort segment of aganglionosis, involving only the internal anal sphincter. [422] Morphologic confirmation of the diagnosis may be difficult, and physiological testing becomes even more important. Patchy or zonal loss of ganglia (ladder pattern) and dysplastic neurons have also been described. Cases classified as acquired aganglionosis have been reported. In these, ganglia were seen in tissues removed at an initial operation, but when clinical failure led to further surgery, an aganglionic segment was clearly demonstrable. Rather than that these were cases of acquired disease, it seems more likely that a short aganglionic segment was missed on the initial evaluation. With greater awareness of the more subtle morphologic and physiological abnormalities, Hirschsprung disease is being detected more often in adults. [423] The clinical, physiological, and morphologic features in adulthood are usually similar to those of the milder form of the disease when recognized earlier in life. Thus, congenital megacolon (Hirschsprung disease) can be subdivided into the classical form, short-segment types, ultrashort-segment types, and other variants (see Table 73-10).

Acute Megacolon Acute toxic megacolon occurs in patients with severe, fulminant inflammatory bowel disease or infectious colitis (see Chapter 83 and Chapter 88); the colon also dilates in response to acute distal obstruction (e.g., volvulus or carcinoma). However, pseudoobstruction and acute megacolon can occur without evidence of intrinsic colonic disease or mechanical obstruction. In this latter instance, the patient has a form of acute colonic pseudoobstruction, named after Ogilvie. [424] In fact, the condition he described in two patients was longstanding and caused by malignant infiltration of mesenteric nerves. Regardless of the misnomer, the syndrome, now given Ogilvie's name, should be considered as a form of pseudoobstruction localized to the colon; the characteristic time course is acute or subacute. Moreover, most cases are clearly associated with an underlying disease; the most common predisposing conditions described in a metaanalysis of 400 cases were trauma, orthopedic surgery, obstetrical procedures, pelvic and abdominal surgery, metabolic imbalance, and neurological conditions. [425]

Pathophysiology. The precise cause of acute colonic pseudoobstruction is unknown. However, Ogilvie syndrome is the clearest example of the human colon dilating in response to nonmechanical factors. Ogilvie proposed that an imbalance of intrinsic and autonomic neural control is the basis for the syndrome. [424] This propensity of the human colon to relax and dilate has been studied experimentally with the barostat. [1] In these experiments, wall tone decreased markedly during sleep and in response to glucagon; a model of colonic dilation during acute distention of the ileum has been described in dogs. [426] This concept, that the wall of the colon is able to relax readily in response to physiological and pharmacological stimuli, may be an important basis for megacolon.

Clinical presentation. The typical patient is a middle-aged or older person who is recovering quite uneventfully from surgery performed a few days previously and who is already on a general diet. In one report, [427] more than half the patients had undergone recent surgery or manipulation of the spine or retroperitoneum. The abdomen becomes grossly distended and breathing becomes labored, but early in the course, no peritoneal signs are present and the white blood cell count is normal. An abdominal film shows massive gaseous distention of the colon with air distributed throughout, including the rectum. Usually, the small bowel is not seen. The diameter of the cecum at this point is often 9 to 10 cm.

Initial management. Oral feedings should be stopped, parenteral fluids started, and a nasogastric tube passed; all nonessential drugs should be discontinued. A Hypaque (water-soluble contrast) enema is administered; in this way, mechanical obstruction can be excluded and pseudoobstruction confirmed. As a side benefit, hyperosmolar Hypaque usually evacuates the colon during the diagnostic maneuvers. Once confirmed, acute colonic pseudoobstruction should be treated aggressively, with a rectal decompression tube and enemas. Any associated metabolic abnormalities or electrolyte disorders (e.g., hypokalemia) must be corrected; these are prominent in 20% to 30% of cases. [427] Efficacy has been reported for pharmacological blockade of ganglia with guanethidine followed by cholinergic stimulation with neostigmine, [428] although it must be emphasized that the condition resolves in many cases with simple measures. [429] Intravenous neostigmine (up to 2 mg) was effective in acute colonic pseudoobstruction in a randomized controlled trial. [430]

Subsequent management. Most of the patients who do not respond to enemas, nasogastric decompression, and drug therapy continue to have a normal white blood cell count, and fever or peritoneal signs do not develop. If the cecum measures more than 11 cm, the next step is colonoscopic decompression. The previous enema often empties the colon, facilitating endoscopy. Gas and liquid stool are aspirated while small amounts of additional air (or carbon dioxide) are insufflated. The mucosal detail is often obscured by the contents, but an obstructing lesion can usually be seen. It is not necessary to reach the cecum to accomplish adequate decompression; positioning the colonoscope at the hepatic flexure with aspiration of the distal contents usually collapses the right colon. An abdominal film should then be obtained; the collapsed cecum can be documented and the patient kept on enemas until stool and flatus pass spontaneously. Although colonoscopic decompression may have to be repeated, more than 80% of patients respond to colonoscopic decompression and require no further management. An operation is advisable for patients with a very large cecum (>14 cm) and for those whose condition remains intractable to medical and endoscopic management. The most useful and efficacious approach is tube cecostomy, now possible by laparoscopic techniques. Moreover, if at any time a patient manifests fever, leukocytosis, or peritoneal signs, abdominal exploration is mandatory. In these situations, the right side of the colon is frequently nonviable or is already perforated. For perforation, right hemicolectomy with ileostomy and a mucous fistula is the operation of choice. In patients with nonviable bowel but without perforation, a right hemicolectomy and primary anastomosis can be performed with little risk for serious complications. For patients with megacolon, a normal-sized rectum, and a normal mechanism of defecation as determined by the newer tests of pelvic floor function, ileorectostomy is the operation of choice; these patients usually establish a relatively normal bowel habit. Patients with megarectum, a normal-sized colon, and a defecation abnormality are candidates for the Duhamel operation or perhaps a coloanal anastomosis. The enlarged rectum is resected in its entirety; therefore, fecaliths in the rectal remnant, so common in patients after Duhamel procedures, are avoided.

Syndromes Partly Attributed to Disorders of Colonic Motility

Chronic Diarrhea Diarrhea is defined as more frequent defecation or the passage of stools that are less well formed than is thought normal by the individual. In patients with IBS and diarrhea, transit through the proximal colon is accelerated, and the rates at which the ascending and transverse portions of the colon empty are related to fecal weight. [431] Stools that are less solid (fragmented, semisolid, or liquid) result from more rapid transit through the colon; however, the most characteristic bowel pattern in IBS is one of wide fluctuations of stool frequency and consistency. Presumably, colonic transit changes dramatically in IBS, from slow to fast (or vice versa), to produce these stool patterns. The scintigraphic method has been used to demonstrate rapid small bowel and colonic transit in the diarrhea of carcinoid syndrome. [432] A fivefold increase in the emptying rate of the ascending and transverse parts of the colon was noted; this was associated with storage of a reduced volume in the ascending colon, as measured by the scintigraphic method. The rapid transit and reduced capacitance of the proximal colon were associated with increased postprandial tone as measured by the barostat. A serotoninergic mechanism has been implicated, and in carcinoid diarrhea, 5-HT$_3$ antagonists reversed the abnormalities. [433]

Irritable Bowel Syndrome: Motor Dysfunction IBS is now defined usually by the criteria of an international panel that evaluated the symptomatic features of this extremely common condition. The major features are altered stool frequency or consistency and abdominal pain that is directly related to a change in the pattern of bowel movements. Thus, it is not surprising that the colon has been incriminated as the origin of these symptoms. Colonic transit was evaluated in patients with IBS and diarrhea, [431], [434] and rapid transit of solids through the proximal colon was documented. It had earlier been reported that small bowel transit also was rapid in IBS with diarrhea. [434] The variable consistency of the stools that is so characteristic of IBS presumably is the result of slow transit of some of the fecal residue and rapid transit of other portions. The constipation experienced by some patients with IBS appears to blend imperceptibly into the syndrome of idiopathic constipation. Not surprisingly, then, the transit of radiopaque markers is prolonged in constipated patients with IBS. In a group of nearly 300 persons referred to a tertiary center with a major complaint of constipation, [435] the majority had only a modest slowing of colonic transit (it was sometimes normal) and were considered to be examples of IBS.

Inflammatory Disease of the Colon The diarrhea of inflammatory colitis is primarily caused by the exudation of fluid and mucus, bleeding, and impaired absorption; however, disordered motility and transit should also be considered. Connell [436] first proposed that the colon in advanced ulcerative colitis may behave as a semirigid tube, showing fewer phasic pressure waves than is normal; moreover, it is well recognized clinically that haustral contractions are absent in the late stage of colitis. These radiologic phenomena (haustra) are thought to be produced by repetitive, nonpropagated, phasic contractions. Reddy and colleagues [437] used combined manometric and scintigraphic techniques to study transit in ulcerative colitis. They confirmed the paucity of phasic pressure activity in the fasting state. Postprandially, however, low-amplitude pressure waves were more frequent in colitis, and when these were propagated, they always moved contents in an antegrade direction. Thus, movement of isotope from the splenic flexure to the sigmoid colon was usually rapid, perhaps reflecting the absence of retarding pressure waves. [437] Another important disorder of motility is seen in ulcerative proctitis, in which a heightened sensitivity of the rectum to distention has been reported. [438], [439] Clinically, such patients are often sensitive to very small amounts of feces or mucus in the rectum. Pressure waves of high amplitude in the rectum have been recorded in proctitis and implicated in the symptom of tenesmus.

Response of the Colon to Drugs

Laxatives The most complete study of laxative agents and drugs that cause diarrhea as a major side effect was that of Karaus and Sarna. [440] They studied the effects of hypertonic glucose, castor oil, neostigmine, and guanethidine in dogs with strain gauges sewn onto the colon. In all instances, when the agents induced diarrhea, powerful contractions migrated around most or all of the colon and led to an evacuation. On the other hand, a local stimulus to the rectum (distention) was able to provoke expulsion of the balloon without participation of the whole colon in the motor event. The motor effects of the local instillation of stimulant laxatives have been recorded from the human colon, [360] and the myoelectric responses of oral senna have been reported. [441] The effects of laxative doses of castor oil on the colon should be similar to those reported for oleic acid; [361] oleic acid and the active principle of castor oil (ricinoleic acid) are both C-18 aliphatic fatty acids. Lactulose has also been used experimentally as a stimulant of colonic transit; when it was administered in doses that induced a modest increase in stool frequency, transit through the right side of the colon was hastened. [442]

Antidiarrheal Agents The most effective antidiarrheal agents currently available are those related to opioids. Opiates are said to augment the mixing contractions that inhibit transit and to increase colonic tone, both of which might be expected to favor absorption. These actions should retard transit and help alleviate diarrhea. The effects of morphine on colonic tone have also been explored in dogs and humans with the electromechanical barostat. Under baseline conditions in the dog, the drug increased tone sharply [362] in association with an increase in phasic contractions. In the human colon, [443] the effects of morphine were more complex; it relaxed a segment showing high postprandial tone and caused a late (40 minutes after injection) decrease in spontaneous tone. Thus, the effects of opiates on the colon are complex, and perhaps species dependent.

Colonic Prokinetic Agents New 5-HT$_4$ agonists have been shown to accelerate colonic transit in health and in patients with functional constipation and constipation-predominant IBS. [444], [445], [446], [447], [448] and [449] This may represent a new generation of prokinetic agents for the gastrointestinal tract. However, confirmation of their clinical effectiveness awaits additional studies.

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CHAPTER 74

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SMALL INTESTINE: INFECTIONS WITH COMMON BACTERIAL AND VIRAL PATHOGENS

Each year, an estimated 4 million children worldwide younger than 5 years of age die as a consequence of diarrheal illness. [1], [2] More than a billion others become ill and are temporarily unable to attend to their normal daily activities. Infants and young children in developing countries with limited medical resources are challenged further by suboptimal sanitary conditions and frequent intestinal infections (10–20 episodes of diarrhea before the age of 3 years) that contribute to a poor nutritional state. [3] Diarrheal disease is also important in developed countries, where surveys indicate it ranks second only to respiratory disease as a cause of illness among the general population (average, 1.2–1.5 episodes per person per year). [4], [5] The incidence of diarrheal disease in industrialized countries has declined through the years, but outbreaks of acute gastroenteritis with a variety of pathogens reflect the continued the importance of this illness. New and emerging pathogens, as well as recognized pathogens, may cause severe, even fatal, illness and have far-reaching implications for the community and economy at large. In addition, international travel, fresh food products imported from developing countries, and changes in dietary habits expand the spectrum of microbial pathogens that an individual may encounter, even in developed countries. [6]

The economic impact of diarrheal illness is also quite substantial. In the United States in the 1980s, there were 99 million estimated cases of acute gastrointestinal illness annually, characterized by vomiting or diarrhea. [7] In 1985, 8.2 million affected individuals sought medical attention for their gastrointestinal illness, and of these, 250,000 required hospitalization. The medical costs of hospitalization were $560 million, whereas costs related to lost productivity were in the range of $200 million. The 7.9 million persons treated as outpatients incurred medical costs of $690 million and lost productivity costs of $2.06 billion. For the more than 90 million persons who did not seek medical attention, lost productivity costs alone were estimated to be $19.5 billion. Although these estimates are based on the 1980 population and 1985 economic statistics, they represent minimum costs and demonstrate the magnitude of the economic impact of diarrheal illness in the industrialized world. The Centers for Disease Control and Prevention (CDC) now estimate that 267 million episodes of diarrhea occur each year among adults in he United States, resulting in 612,000 hospitalizations and 3,000 deaths. [8]

This chapter focuses on the major bacterial and viral pathogens that infect the small intestine. Whether by toxin-mediated effects or direct destruction of intestinal epithelial cells, these microbial pathogens have devised ways to disrupt the normal fluid-handling capabilities of the intestinal tract and cause diarrhea. Normally, the small intestine is presented with approximately 9 L of endogenously secreted (7 L) and ingested (2 L) fluid each day, of which 7 to 7.5 L is absorbed in the small bowel. The remainder is presented to the colon, where all but approximately 200 mL is absorbed and excreted in stool. Diarrhea ensues when the absorptive capacity of either the small bowel or colon is exceeded and unable to accommodate larger fluid loads induced by infection. In general, the diarrhea caused by infection with a small bowel pathogen is characterized by high-volume, less frequent bowel movements, whereas lower-volume and more frequent bowel movements are associated with colonic diarrhea.

FOOD POISONING AND COMMON SOURCE OUTBREAKS

Foodborne disease is a widespread public health problem. [9] A recent estimate indicates that 76 million such illnesses occur annually in the United States, contributing to 325,000 hospitalizations and 5000 deaths. [10] Previous estimates ranged from 6 to 80 million illnesses per year, with the economic impact perhaps being $5 billion or more. [11], [12] Water and food (if improperly cooked or handled or left unrefrigerated before consumption) can easily serve as culture media and vehicles for the transmission of microbial agents.

Foodborne disease results from the ingestion of food or water contaminated with pathogenic microorganisms, microbial toxins, or chemicals. Although this chapter emphasizes gastroenteritis, other foodborne illnesses (e.g., hepatitis A, brucellosis, listeriosis, botulism, diphyllobothriasis) do not cause prominent gastrointestinal symptoms. Risk factors for foodborne illness include misidentification of unsafe foods, chemical contamination of unsafe foods, and faulty preparation practices (poor personal hygiene of food handlers, inadequate storage and cooking).

Although the individual with a foodborne illness may or may not recognize the symptoms as related to the ingestion of a particular contaminated food item, certain established guidelines, used by the CDC, aid in the diagnosis of this syndrome. Foodborne disease can in most cases be established if two or more people have a similar illness after consuming a common meal, if an epidemiologic survey implicates a particular food item(s) as the probable cause of the outbreak, or if microbiologic studies identify the same organism or epidemic strain from a common food, food source sample, or clinical specimens obtained in a suspected outbreak.

The CDC publishes annual summaries of foodborne illness in the United States. From 1993 to 1997, 2751 outbreaks, representing 86,058 cases, of foodborne illness were reported to the CDC. Causes were identified in 878 (32%) of the outbreaks, and of these, bacterial pathogens were the most frequent (75%), followed by chemicals (17%), viruses (6%), and parasites (2%). [13] A cause was not established in most (68%) of the reported outbreaks. The incubation time for nearly half of these outbreaks was 15 hours or longer, suggesting a viral cause, such as Norwalk-like viruses. [14] The incubation period and presenting complaint can be helpful in differentiating among the etiologic agents, as shown in Table 74-1.

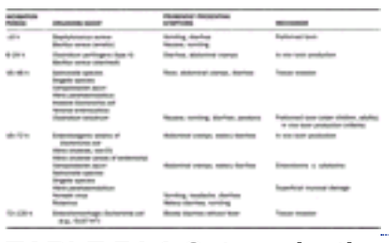

TABLE 74-1 Categorization of Intestinal Infection by Incubation Period and Major Presenting Complaints

Salmonella species caused 357 (55%) of the 655 foodborne diarrheal outbreaks reported. Seven percent of all outbreaks and 22% of outbreaks for which a cause was determined were attributed to *Salmonella enteritidis,* which also was associated with the greatest number of deaths (ten, four of which occurred in nursing home residents). [13]

Because the number of reported foodborne outbreaks is likely a fraction of all foodborne disease, [15] the CDC began an active surveillance program (Emerging Infections Program Foodborne Diseases Active Surveillance Network, or FoodNet) in 1996 to quantify and monitor foodborne illnesses from selected sites in the United States. [16] *Campylobacter, Escherichia coli* O157:H7, *Listeria monocytogenes, Salmonella, Shigella, Vibrio, Yersinia enterocolitica, Cryptosporidium,* and *Cyclospora cayetanensis* are monitored. Most frequently diagnosed during all 5 years of data collection was *Campylobacter* infection. *Salmonella, Shigella,* and *E coli* O157:H7 were commonly found also, but a majority of *Shigella* cases were not acquired through food.

Toxins Associated with Bacterial Food Poisoning

Staphylococcus aureus Staphylococcal food poisoning is one of the most common causes of foodborne illness, accounting for 1.0% to 2.0% of recognized bacterial food poisoning outbreaks reported to the CDC from 1993 to 1997. [13] Staphylococcal food poisoning outbreaks are most common in the summer, reflecting the association between *S aureus* outbreaks and large gatherings, such as picnics. *Bacillus cereus* produces a similar illness; both illnesses are characterized by profuse vomiting that occurs less than 12 hours after a contaminated food has been ingested. *S aureus* colonizes the skin and mucous membranes of 20% to 50% of healthy people, and these persons serve as the most frequent source of food contamination. *S aureus* produces at least seven antigenic types of classical enterotoxin and a delta-toxin, all of which are capable of causing net fluid secretion in the intestine. [17] *Staphylococcus* enterotoxins, purified as small 27- to 30-kd single-chain polypeptides, are resistant to heat, irradiation, pH extremes, and proteolysis. [18], [19] and [20] Assays to detect the enterotoxin in food and its production in vivo are important epidemiologic tools. [21] Phage typing of species can also be used for epidemiologic investigations to trace the source of infection, with phage type III being most often associated with outbreaks of foodborne disease. Foods high in salt, protein, and sugar content select for growth of *S aureus,* with dairy products, salads, and meats often implicated in outbreaks.

Bacillus cereus *B cereus* accounted for fewer than 1% of foodborne outbreaks reported to the CDC between 1993 to 1997. [13] *B cereus* is a ubiquitous aerobic, spore-forming, gram-positive rod organism that is found in soil and water throughout the world and in most raw foods. [22] These bacteria colonize 10% to 40% of humans. Two clinical syndromes are associated with the ingestion of this organism. One, an emetic syndrome characterized by a short incubation period (1–6 hours), presents with vomiting and abdominal cramping. The other, a diarrheal syndrome with a longer incubation period (6–24 hours), is characterized by watery diarrhea, abdominal cramping, and occasionally vomiting. [23], [24] The duration of illness is 2 to 10 hours for the emetic syndrome and 16 to 48 hours for the diarrheal syndrome. The organism can produce either an emetic or diarrheal toxin, depending on the food in which it grows. [25], [26] The emetic syndrome is associated with cooked rice and in 95% of cases is caused by the ingestion of a preformed toxin (5000–10,000 kd) that is resistant to heat. In contrast, the diarrheal syndrome is associated with vegetables, sauces, and puddings and appears to be caused by the ingestion of a heat-labile protein with a molecular weight of 38 to 46 kd. [27], [28] Outbreaks of foodborne illness caused by *Bacillus subtilis, Bacillus licheniformis,* and *Bacillus pumilus* have also been reported. [27]

Clostridium botulinum *C botulinum* is a ubiquitous, gram-positive, spore-forming, anaerobic bacterium that produces a neurotoxin capable of blocking the release of acetylcholine at the neuromuscular junction. The spores are heat-resistant and require anaerobic conditions plus a pH above 4 for production. Home-canned foods and fish products are the most common sources, although any food that meets the criteria for spore germination can become contaminated. Symptoms occur 12 to 36 hours after ingestion of the preformed toxin; the initial clinical features are nausea, vomiting, abdominal pain, and diarrhea. Neurological symptoms include diplopia, dysphagia, dysarthria, dysphonia, ophthalmoplegia, descending bilateral weakness, and respiratory muscle weakness. The case fatality rate remains at 15%. There are seven toxin types, A through G. Type A botulism is more common in the western United States, whereas type B is more common in the eastern parts of the country. Type F is more commonly associated with fish products. Wound botulism is similar, without the gastrointestinal complaints. Botulism is first recognized on the basis of clinical signs and symptoms, with the differential diagnosis including Guillain-Barré syndrome, myasthenia gravis, stroke, and chemical poisoning. Assaying the vomitus, stool, or suspected food for the toxin may lead to the diagnosis. Electromyography may be helpful in distinguishing the diagnosis from Guillain-Barré syndrome. Treatment is supportive, along with early administration of antitoxin. The development of infant botulism requires ingestion of the organism and is seen in children younger than 1 year of age. Antitoxin is given parenterally, but the role of antibiotics is controversial.

TRAVELER'S DIARRHEA

Diarrheal disease has been a well-recognized consequence of travel for many years. Despite advances in public sanitation and water treatment, travel to a less developed country from a developed country is still associated with a substantial risk (~40%) for diarrheal disease. [29], [30] Much has been learned, however, about the specific etiologic agents that cause diarrhea in the traveler and strategies to help reduce the risk for illness.

Pathogenesis and Etiologic Agents

Virtually any microbial pathogen that can cause acute diarrheal disease in the endemic setting is capable of causing traveler's diarrhea. To some degree, the specific agents that cause traveler's diarrhea vary by geographic region and season. For example, the parasitic pathogens *Giardia* and *Cryptosporidium* appear to be common causes of diarrheal disease in visitors to Russia, whereas *Aeromonas* species appear to be very common causes of traveler's diarrhea in visitors to Thailand. [31], [32] Studies of etiologic agents from individual regions have varied in the types of organisms detected and in the sensitivity and specificity of the assay systems employed. In addition to differences in the geographic distribution of various etiologic agents, seasonal variations in the causes of traveler's diarrhea have been noted. For example, toxigenic *E coli* is somewhat more frequently encountered during the warmer summer months, whereas *Campylobacter jejuni* appears to occur more frequently in the winter months. [33], [34]

Although bacteria, viruses, and parasitic organisms have all been linked to traveler's diarrhea, a large number of studies have demonstrated that bacterial pathogens are the most frequent etiologic agents of this syndrome. This conclusion has been reached from the results of studies that tried to identify all agents, and studies showing that antibiotic (antibacterial) treatment or prophylaxis is very efficient in reducing most episodes of traveler's diarrhea. Of the wide array of bacterial pathogens encountered, enterotoxigenic *E coli* (ETEC) appears to be the most common worldwide (Table 74-2). These organisms, which produce enterotoxins (heat-labile toxin [LT] or heat-stable toxin [ST]), account for between 5% and 50% of all episodes of traveler's diarrhea. Other forms of pathogenic *E coli,* including enteroadherent *E coli* (EAEC) and enteropathogenic *E coli* (EPEC), account for fewer illnesses. [35] *Shigella* species and *C jejuni* are perhaps the next most important groups of pathogens, accounting for between 10% and 15% of cases. *Shigella* infection is frequently associated with fever and a dysentery-like illness. Other bacteria, such as *Salmonella, Aeromonas* species, and *Plesiomonas shigelloides,* usually account for fewer than 10% of cases, as do the viral pathogens, such as Norwalk virus and other caliciviruses and rotavirus. Traveler's diarrhea resulting in a prolonged illness (>1 month) is most frequently associated with the parasitic pathogens *Giardia* and *Cryptosporidium*. Of note, an etiologic agent cannot be identified in about 20% to 30% of all cases of traveler's diarrhea. Chronic diarrhea of travelers without a detectable etiologic agent may be classified as Brainerd diarrhea. [36]

ETIOLOGIC PATHOGEN	UNITED STATES (%)	DEVELOPING COUNTRIES (%)	TRAVELERS (%)
Rotavirus	20–45	15–40	<10
Astroviruses, Norwalk-like viruses, enteric adenoviruses	5–20	5–10	0
Campylobacter jejuni	5–10	5–10	5–15
Enterotoxigenic *Escherichia coli*	<5	10–40	10–40
Enteropathogenic *Escherichia coli*	<5	<5	<5
Shigella species	5–10	5–10	10
Salmonella species	<5	<5	5–10
Aeromonas species, *Plesiomonas shigelloides*	<5	5–10	5–10
Giardia	<5	<5	<5
Unknown and other	40	30	20

TABLE 74-2 Etiology of Infectious Watery Diarrhea in Children in Various Geographic Locations and in U.S. Travelers to Developing Countries

The mechanisms by which each pathogen causes diarrhea are extremely varied and range from the secretion of a soluble enterotoxin, which stimulates intracellular cyclic adenosine monophosphate (cAMP) levels and subsequent chloride secretion, to destruction of mature villus tip cells and loss of absorptive epithelium.

Epidemiology

An increased incidence of diarrhea is associated with all forms of international travel, including travel from one developed country to another, travel from a less developed country to a more developed country, and, most commonly, travel from a developed country to a less developed country. [37] The third scenario is the most frequent cause of traveler's diarrhea (30%–50% of cases), whereas the first is the least common (<5% of cases). Other than microbial agents, traveler's diarrhea is most probably caused by, or exacerbated by, a variety of factors, such as stress, sleep deprivation, and excessive alcohol consumption.

The source of microbial contamination can vary. The commonly held notion that tap water is the most frequent culprit is inaccurate. Tap water in many large cities in less developed countries is safe, although contamination can occur during the rainy season and in more rural areas. [38] Food is the most common vehicle for the spread of microbes that cause diarrhea. [39] Fruit and vegetables may become contaminated by fertilization with human waste, preparation by food handlers who are ill, or inadequate washing or cooking; prepared food that remains at room temperature for long periods may also become contaminated.

Host factors also influence the incidence and severity of traveler's diarrhea. Travelers from developed countries tend to have lower levels of immunity to many enteric pathogens, especially ETEC, and are more susceptible to infection and illness. [40] Gastric acid is a general barrier to almost all enteric pathogens; people with lowered levels of stomach acid, either as a consequence of prior surgical therapy or of pharmacological treatment, are at increased risk. [41] Immunosuppressed individuals, irrespective of the cause, may be at some increased risk. Younger persons have higher rates of traveler's diarrhea than older travelers, presumably because of more adventurous eating habits. In some cases, known genetic characteristics, such as blood group O antigen, increase the risk for specific infections, such as cholera. [42]

Although the risk for traveler's diarrhea is highest when travelers from developed countries travel to less developed areas, a geographic spectrum of the relative risk for acquiring diarrhea during travel to less developed countries has been noted. The highest risk is associated with travel to most countries in Latin America and Africa and to many countries in Asia. Travel to the Mediterranean countries of Europe, eastern Europe, Russia, and China is associated with a somewhat lower risk.

Clinical Features

Traveler's diarrhea usually occurs within the first 14 days of travel. It can also occur within a week or more after return from a foreign country. The diarrhea is usually characterized as watery, with the passage of three to ten bowel movements a day for 2 to 5 days. [30] Abdominal cramps and pain often accompany the illness. Fewer patients experience fever, chills, vomiting, and dysentery-like symptoms. Fewer than 10% of patients have an illness that lasts longer than a week, and about 2% of patients have illness lasting a month or more. [43] Acute disease is severe enough to warrant bed rest in fewer than one fourth of patients. [44] Prolonged disease (>1 month) or dysentery-like symptoms help to narrow the potential etiologic agents (to *Giardia* and *Cryptosporidium,* or to *Shigella* and *Campylobacter,* respectively). It is not possible to determine the etiologic agent with any degree of certainty if the illness is characterized by watery diarrhea and cramps, however. Rarely is diagnosis required or available in the setting of acute watery traveler's diarrhea. For cases of dysentery, a diagnosis, if available, is more useful. Given that traveler's diarrhea occurs in the traveling patient, it is almost always reasonable to initiate treatment without an etiologic diagnosis. Stool examination for parasites is warranted only in cases of prolonged illness.

Prevention

One of the simplest and most effective means of preventing traveler's diarrhea is the utilization of safe eating practices. [45] Food should be thoroughly cooked. Peeled fruits and vegetables are safe, whereas salads and uncooked vegetables are high risk. Bottled water, carbonated drinks, and beer are low risk, whereas tap water and ice are associated with an increased risk for illness. Cooked foods that sit at room temperature or on a buffet table for long periods of time should be avoided. Food obtained from street vendors is frequently associated with traveler's diarrhea.

Traveler's diarrhea can also frequently be prevented by the prophylactic use of antibiotics or other medicines. Antibiotic prophylaxis is not without risk, however; side effects, including minor and major drug allergies, do occur. Prophylaxis should probably be recommended only for travelers engaged in very important activities, or for those with decreased gastric acid or immunosuppression that increases their risk for the development of illness. Antibiotics, including doxycycline, trimethoprim-sulfamethoxazole (TMP-SMX), and the fluoroquinolones (FQs), have been shown to be highly effective in preventing traveler's diarrhea. Because of widespread antibiotic resistance, both doxycycline and TMP-SMX are no longer reliably effective, and the FQs are generally the agents of choice (Table 74-3). [46], [47] Traveler's diarrhea can also be prevented with reasonable efficacy by prophylactic treatment with bismuth subsalicylate (BSS). [48], [49] A dose of two 262-mg tablets, chewed four times a day at mealtime and at bedtime, is more than 60% effective in preventing illness. Side effects include a black tongue and occasional tinnitus. *Lactobacillus* species have been shown to have some efficacy in preventing traveler's diarrhea. [50]

TABLE 74-3 Recommendations for Antimicrobial Treatment of Diarrhea Caused by Small Bowel Pathogens

Treatment

In most cases, the treatment of traveler's diarrhea is quite effective and preferable to prophylactic regimens. Several nonspecific therapies are available. Two tablets

of BSS every 30 minutes up to eight times per day has been shown to hasten recovery in mild to moderate illness. [51] This effect is probably mediated through both antisecretory and antibacterial activities. Antimotility agents, such as the synthetic opiates loperamide and Lomotil (diphenoxylate hydrochloride with atropine), are generally more effective than BSS and are most effective when taken shortly after the onset of symptoms. [52] Loperamide is preferable to Lomotil because it does not contain atropine. Loperamide is generally given in a dose of 4 mg (two capsules) initially, followed by 2 mg after each loose bowel movement; up to eight capsules can be taken per day. Lomotil or loperamide, although effective in reducing the frequency and persistence of diarrhea, may enhance disease by impairing intestinal motility, delaying clearance of the infectious agent, and facilitating infection, particularly when illness is caused by highly invasive organisms like *Shigella*. [53] Drugs such as Lomotil and loperamide should be avoided if infection by an invasive organism is suggested by the presence of bloody diarrhea, fever, fecal leukocytes, or substantial systemic symptoms.

Antibiotic therapy is the most effective intervention for traveler's diarrhea. Antibiotics substantially shorten the duration of illness as well as the number of bowel movements per day. [54], [55] An indications for antibiotic use is the passage of two or more unformed stools in 24 hours, diarrhea with abdominal distress or fever, or the passage of bloody diarrhea. Various antibiotics have proved effective, but given increasing drug resistance and the high incidence of illness caused by *C jejuni* in some areas, FQs are now the treatment of choice for adults (see Table 74-3). Because of the potential of FQs to damage articular cartilage, TMP-SMX is still recommended for children, although drug resistance is becoming more widespread. In areas where FQ-resistant *C jejuni* is prevalent, azithromycin is the treatment of choice. [56] Therapy for 3 days is probably sufficient in virtually all cases, and a single dose may be adequate in most. [57]

Vaccines for several of the bacterial and viral causes of traveler's diarrhea are under development (see later sections) and are likely to become available for travelers in the next decade.

BACTERIAL INFECTION

Pathogens such as *Shigella, Salmonella,* and *Campylobacter* are often spread through contaminated foods. *Shigella* and *Campylobacter* usually are colonic pathogens, and as such are discussed in Chapter 88. *Salmonella* and other organisms, including *Clostridium perfringens, L monocytogenes,* diarrheagenic *E coli, Yersinia* species, *Vibrio* species, *Aeromonas, Edwardsiella,* and *Plesiomonas,* are discussed in the following sections. As mentioned previously, the length of the incubation period and the presenting complaint help to distinguish among etiologic agents (see Table 74-1). A more detailed summary of the key features of specific agents is provided in Table 74-4.

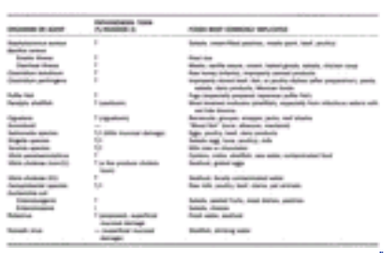

TABLE 74-4 Clinical and Epidemiologic Features of Agent-Specific Foodborne Diseases

Clostridium perfringens

Epidemiology *C perfringens* most commonly causes a mild foodborne illness. [22] Approximately 2% of all foodborne cases reported to the CDC are caused by *C perfringens* food poisoning. [13] This organism is an obligate anaerobe, but it can tolerate exposure to oxygen for up to 72 hours, and it is the only nonmotile species of *Clostridium*. Spores are heat-resistant and survive cooking to germinate. The organisms grow rapidly at temperatures of 15° to 50°C and can double in number in 10 minutes. Illness requires the ingestion of food heavily contaminated with more than 10^8 bacteria per gram. Most of the foodborne *C perfringens* cases are caused by type A strains, which are found in the soil throughout the world and in the intestinal tracts of all vertebrate animals. *Clostridium* types B, C, D, and E are found only in animals, not in soil. Most *C perfringens* outbreaks occur during the fall and winter months (unlike outbreaks caused by *Salmonella* species and *S aureus*), and typically involve the foods listed in Table 74-4. When implicated, these foods usually have been prepared in advance and allowed to cool without prompt refrigeration. In this fashion, heat-resistant spores that survive the cooking process germinate within the food during the cooling period (reduced redox potential within the food favors this process). On reheating before consumption, not all viable cells are killed, and heat-induced sporulation of the cells and enterotoxin production within the intestine cause diarrhea.

Pathogenesis The pathogenesis of *C perfringens* food poisoning is based on the ability of the organism to elaborate enterotoxins. Only 5% of the global *C perfringens* population possesses the *C perfringens* enterotoxin (CPE). CPE is a heat-labile, 35-kd protein toxin that is capable of causing fluid accumulation in animal ileal loop models. It is elaborated by *C perfringens* types A and, less commonly, C and D. CPE causing foodborne illness has been found to be the product of a chromosomal cpe gene, whereas CPE related to antibiotic-associated diarrhea is caused by clostridria isolates containing a plasmid cpe gene. [58] CPE appears to cause clinical symptoms by cytotoxic action in the intestinal epithelial cells. [59] Intralumenal CPE binds to mucosal cell surfaces, where it causes structural damage to these cells with subsequent loss of intracellular substances, primarily sodium, chloride, protein, and fluid. This effect probably leads to impaired macromolecular synthesis, further structural damage, and eventual cell death. [60] CPE also acts as a superantigen reactive with human T lymphocytes. [61]

Clinical Features *C perfringens* causes a watery diarrhea (two to four stools per day) that frequently is accompanied by pronounced abdominal cramping and pain; symptoms characteristically develop 8 to 24 hours (mean, 12 hours) after ingestion of the contaminated food. Attack rates during epidemics usually are high, and in the United States, the median percentage of people affected in an outbreak ranges between 23% and 35%. Constitutional symptoms (e.g., vomiting, fever, chills, malaise) are uncommon. The disease is self-limiting, and full recovery is expected within 24 hours. A fatal outcome is rare and usually associated with an older age and secondary dehydration. Although serum antibody to CPE has been demonstrated in asymptomatic adults and in some but not all volunteers challenged with CPE, the development of protective immunity has not been shown. Local production of anti-CPE antibody in the gut, however, may confer immunity in previously exposed people. [62] Several reports have implicated *C perfringens* as the cause of antibiotic-associated diarrhea and of prolonged and recurrent infectious diarrhea in the elderly and patients in long-term care settings. [63], [64] Although the incidence of carriage of *C perfringens* within the colon can be high, symptoms are associated with the presence of enterotoxin-producing strains. In addition, a significant association with sporadic cases of diarrhea has been reported in patients in whom symptoms of enterocolitis develop, and whose stools contain *C perfringens* and its enterotoxin but no other identifiable common pathogen. [65] The illness thus resembles that associated with *Clostridium difficile,* and although pseudomembranous enterocolitis per se has not been demonstrated in any of the patients in whom endoscopic examination has been performed, the presence of fecal blood and biopsy-proven mucosal hemorrhage and edema in some patients speaks in favor of a colitis. Although many experts do not recommend antimicrobial treatment for this enterotoxin-associated disorder, a favorable clinical and microbiologic response to oral metronidazole (400 mg three times a day for 7–10 days) has been demonstrated. [66] B-toxin is produced by types B and C strains and is associated with the disease pig-bel or, more aptly, necrotic enteritis, which is still reported from isolated parts of the world such as Papua New Guinea. Proteolytic enzymes in the intestine usually inactivate the B-toxin. Underlying diseases such as diabetes mellitus, [67] malnutrition, and a sudden change in the dietary intake of protein may place people at risk, with children and the elderly primarily affected. They may lack the necessary proteolytic enzymes to inactivate the toxin. [27] Bowel resection frequently has been required to treat this disease, but accumulated data indicate that passive immunization with *C perfringens* type C antiserum (beta-antitoxin) [68] and active immunization with pig-bel toxoid vaccine significantly reduce both morbidity and mortality from this illness. [69]

Diagnosis *C perfringens* food poisoning cannot be distinguished easily from other noninfectious, enterotoxigenic intoxications, such as those caused by *B cereus* and *S aureus*. Confirmation of an outbreak relies on the quantification of organisms in food (i.e., >10^5 organisms per gram) or the excreta (>10^6 spores per gram) of ill people. Simple-to-perform assays that are highly sensitive and specific for CPE (reverse passive latex agglutination and slide-latex agglutination) have proved very useful in accurately diagnosing illness caused by *C perfringens*.

Listeria monocytogenes

Epidemiology *L monocytogenes* is a relatively heat-resistant, non–spore-forming, gram-positive bacillus that has been isolated from the intestinal tracts of humans and animals, sewage, and well water. It also has been isolated from many different foods, including meats, fruits, vegetables, seafood, and dairy products. The organism inhabits the gastrointestinal tract of 1% of asymptomatic humans. [70] *L monocytogenes* is an uncommon cause of foodborne infection (0.1% of foodborne

outbreaks reported to the CDC between 1993 and 1997), [13] but several outbreaks have been reported from a variety of food sources. [71], [72] In 1985, Mexican-style soft cheese made from inadequately pasteurized milk was implicated in 142 cases of *Listeria* food poisoning and 45 deaths; 66% of these involved pregnant women and their infants. The remaining cases had a predisposing health problem. [73] *Listeria* produces a number of virulence factors that facilitate cell invasion and cell-to-cell spread. Cell-mediated immunity appears to play an important role in combating infection with *Listeria*. [74] A humoral response develops against several *Listeria* proteins, the clinical significance of which is not clear. [75] Both IgM, which is absent in neonates, and classical complement, which is low in neonates, appear to be important for the efficient opsonization of *L monocytogenes*. [76] Patients with altered cell-mediated immunity are clearly at risk for infections with this intracellular organism. Populations at risk include predominantly pregnant women and their infants, the latter acquiring *Listeria* through vertical transmission. Other populations at risk include immunocompromised persons, such as those who have hematologic malignancies, cirrhosis, diabetes mellitus, acquired immunodeficiency syndrome (AIDS), or renal failure, and those who are receiving corticosteroids. In addition, the elderly, veterinarians, and laboratory workers are at risk; the disease also occurs in persons with no recognizable predisposing or underlying conditions.

Clinical Features The gastrointestinal tract is the primary portal of entry in all cases except fetal and neonatal infections. The clinical syndromes caused by *L monocytogenes* can range from a mild febrile illness to frank bacteremia, meningitis, and sepsis. Pregnant women generally have a mild illness with flulike features of fever and myalgias. The incubation period may be as long as 2 to 3 weeks. [77] Cases of nonperinatal listeriosis are more likely to present with bacteremia, meningitis, or sepsis. Diarrhea and other gastrointestinal symptoms can precede the onset of systemic listeriosis.

Diagnosis The diagnosis of listeriosis depends on isolation of the organism from a normally sterile site or other site in the setting of an appropriate epidemiologic history. Successful therapy has been associated with early diagnosis and initiation of treatment; however, the optimal agent for therapy has not been defined by controlled clinical trials. Ampicillin is the most commonly recommended therapy, often in combination with an aminoglycoside. Other agents suggested are TMP-SMX, a macrolide, or a tetracycline, but cephalosporins should not be used because they have been associated with clinical failures. The optimal duration of therapy is also unknown, but at least 2 weeks seems to be prudent; longer courses, up to 6 weeks, are recommended for patients with endocarditis and brain abscess.

Escherichia coli Enteropathogens

E coli is a common inhabitant of the gastrointestinal tract in both humans and animals. Although most *E coli* strains do not possess known virulence characteristics, a subset exists that can be pathogenic to humans, specifically through the induction of gastroenteritis. The *E coli* organisms that induce intestinal derangement are named according to their pathogenic mechanisms. In the interest of simplicity, these pathogens are herein collectively referred to as *diarrheagenic E coli,* and the particular syndromes caused by each are discussed in the following sections.

Classification The members of the species *E coli* belong to the family Enterobacteriaceae and are characteristically facultative, aerobic, non–spore-forming, mostly motile gram-negative bacteria that grow well at 37°C in nonenriched media. *E coli* bacteria are distinguished from other members of the Enterobacteriaceae by the demonstration of characteristic biochemical reactions; however, different strains of the species of *E coli* are identified by serologic and virulence testing. Serologic markers are found on different parts of the organism. The H-antigen (H-Ag), or flagellar protein, is located on the peritrichous flagellum; the O-antigen (O-Ag), or lipopolysaccharide antigen, and the K-antigen are located on the cell wall and capsule or envelope (when present), respectively. Fimbria or pili and other attachment factors of these organisms also can be determinants of pathogenicity. Four syndromes of *E coli*–induced intestinal dysfunction have been well defined that show specific clinical and epidemiologic features: those caused by enterotoxigenic (ETEC), enteropathogenic (EPEC), enteroinvasive (EIEC), and enterohemorrhagic (EHEC) organisms. [78]

Enterotoxigenic Escherichia coli

Pathogenesis. ETEC produces disease by elaborating enterotoxins without invading or damaging intestinal epithelial cells. [79] One (or more) of at least three enterotoxins may be produced, which differ in their susceptibility to heat: LT (heat-labile toxin), STa, and STb (heat-stable toxins). The characteristics of LT are remarkably similar to those of cholera toxin (see section “ Vibrio Species”): a molecular weight of 84 kd, five identical binding B subunits, and an enzymatically active A subunit, which activates adenylate cyclase in a nicotinamide adenine dinucleotide (NAD)–dependent reaction. [80], [81] The homology between the A and B subunits of LTI and cholera toxin is approximately 80%. [82] In addition, the action of LTI is neutralized by cholera antitoxin. LTs that are not neutralized by cholera antitoxin are designated LTII. STa toxins are small, methanol-soluble peptides that activate the transmembrane guanylate cyclase system and lead to intestinal secretion. [83] STb toxins do not stimulate the guanylate cyclase system. [84] To be fully pathogenic, ETEC must express the virulence factors for adherence and enterotoxin elaboration. The bacteria first colonize the small intestine with the aid of specific CFA (colonization factor antigen) [coli surface (cs) antigens] attachment factors, namely CFA I, CFA II, CFA III (subdivided into CS 1–3), CFA IV (subdivided into CS 4–6), and additional higher-numbered CSs. [79] The plasmids that encode these attachment factors often, but not always, encode ST or LT. [85], [86] There are several major O serogroups. The serogrouping of ETEC has made possible the epidemiologic investigation of outbreaks. Most, but not all, traditional serogroups are associated with at least one CFA. [87] Prevalence studies indicate a relationship between CF type and the presence or absence of ST or LT. [88]

Epidemiology. ETEC has worldwide distribution and is a major etiologic agent of diarrhea affecting children in the developing world (10%–40%). Although ETEC is a rare cause of diarrhea in the United States, it accounts for most cases of illness in travelers to developing countries. The incidence of ETEC infection is equal to, or greater than, the incidence of rotavirus infection, and these two agents are the predominant cause of diarrheal disease throughout the developing world, although rotavirus more frequently causes severe dehydrating diarrhea. ETEC is acquired primarily through the ingestion of contaminated food or water. Person-to-person transmission is rare because a large inoculum (10^8 organisms) is required to cause illness in healthy people. [89], [90]

Clinical features. The illness is characterized by watery diarrhea, ranging from very mild to cholera-like. Other symptoms may include cramping abdominal pain, rarely low-grade fever, headache, arthralgias, myalgias, vomiting, and chills. The illness usually is self-limiting, lasting 3 to 5 days if untreated; those affected pass an average of 8 to 12 unformed stools per day. The stools are watery, yellow, and devoid of mucus, pus, and fecal leukocytes. Dehydration tends to be severe only in the very young and very old. Fatalities are rare, and treatment with antimicrobials (see “ Prevention” and “ Treatment” in section “Traveler’s Diarrhea”) shortens the duration of disease. Fluid and electrolyte replacement is emphasized, especially in the populations at risk.

Vaccine therapy. A number of approaches have been tried in an effort to develop effective vaccines against ETEC. The major problem is that ETEC organisms are antigenically diverse—that is, they are found in a number of O serogroups and possess one of several different CFAs. In addition, ST is not immunogenic because of its small size.

Enteropathogenic *Escherichia coli*

Pathogenesis. EPEC organisms lack *Shigella*-like invasive properties and fail to produce classical ETEC-type enterotoxins. [91] The mechanism of pathogenesis appears to reside primarily in the ability of the bacteria to attach intimately to and efface the microvilli of enterocytes, leading to the destruction of microvilli and cytoskeletal disruption beneath the adherent organisms. The linkage between the effects of EPEC on the epithelial cells and diarrhea remains unknown.

Epidemiology. EPEC is an endemic diarrheal pathogen among infants in developing countries, with person-to-person spread through the fecal-oral route the most likely mode of transmission. Our knowledge of the clinical aspects of EPEC infection remains incomplete, but infants (<2 years of age) are primarily infected, particularly those in day care centers and hospitals, where several outbreaks have been described. Older children and adults may serve as reservoirs for this organism; EPEC strains have been isolated from the oropharynx and the excreta of asymptomatic contacts of ill cases during several outbreaks.

Clinical features. Illness with EPEC is marked by watery diarrhea, which may become profuse and protracted. EPEC enteritis is one of the few identifiable causes of prolonged diarrhea in infancy. A variable percentage of patients display vomiting, fever, failure to thrive, and metabolic acidosis. Dehydration can be severe and life-threatening if the disease is allowed to progress without fluid and electrolyte replacement.

Diagnosis. The diagnosis of EPEC infection is usually based on slide agglutination of suspected colonies with the use of commercial polyvalent antisera for the O-Ag, which are considered to be more specific for EPEC. This method of screening is neither sensitive nor specific, but complete serotyping for all O-Ag and H-Ag is impractical. Stool culture, serotyping, the documentation of characteristic tissue culture adherence, or the detection of adherence factor by DNA probe if *E coli* is identified are the only ways to incriminate EPEC strains in a particular diarrhea episode.

Therapy. Treatment of EPEC infection usually is not required, given the self-limiting nature of the illness; however, attention should be paid to the replacement of fluids and electrolytes through oral or intravenous routes. In addition, consideration should be given to therapeutic measures to relieve diarrhea. The use of antimicrobial agents is advocated in severe illness and in cases of prolonged diarrhea, but which ones and for how long remain to be firmly established (see Table 74-3). [92] In vitro antimicrobial susceptibilities should be routinely determined in isolated strains because these organisms tend to have a surprisingly high rate of antimicrobial resistance. [93]

Enteroinvasive *Escherichia coli*

EIEC organisms are primarily colonic pathogens and are discussed only briefly here. Strains of *E coli* that display the ability to invade the intestinal mucosa are rare causes of diarrhea, although large outbreaks associated with the ingestion of soft cheeses and in the setting of a cruise ship have been reported. [94], [95] EIEC does not appear to be spread from person to person, and although food does appear to play a major role in transmission, the epidemiology of this illness is unclear. EIEC organisms have been associated with specific somatic antigens (O-Ag), and by definition, they are Sereny test positive (production of keratoconjunctivitis in guinea pigs). EIEC organisms possess the same 140-megadalton (Md) plasmid that is associated with invasiveness in *Shigella* strains. The Sereny test and tissue culture invasion were previously the only methods available to detect EIEC. Polymerase chain reaction (PCR) tests and enzyme-linked immunosorbent assays (ELISAs) developed for EIEC may prove useful in detecting these strains in the near future. The illness is indistinguishable from the dysentery-like disease caused by *Shigella*. Treatment with TMP-SMX is recommended for severe infection (see Table 74-3).

Enterohemorrhagic Escherichia coli

Infection with EHEC is characterized by hemorrhagic colitis and bloody diarrhea. Investigations of foodborne outbreaks of

diarrhea in 1982 in Oregon and Michigan incriminated *E coli* serotype O157:H7 as the etiologic agent in this syndrome. [96], [97] Since then, studies have shown that this particular organism is responsible for both sporadic and epidemic cases of hemorrhagic colitis. Epidemiologic studies point to transmission of this organism in food and water, and person-to-person transmission among household members has been documented. [96], [98] Bloody diarrhea caused by *E coli* O157:H7 can be complicated by the hemolytic uremic syndrome and thrombotic thrombocytopenic purpura, with a resultant high mortality rate. [96], [99] EHEC strains such as *E coli* O157:H7 do not invade the epithelium, do not produce STs or LTs, and do not display CFA I or CFA II (adherence factors). The primary virulence factor of these organisms appears to be the production of at least two toxins with properties similar to those of the toxin elaborated by *Shigella dysenteriae* type 1. These two toxins are called *Shiga-like toxin (SLT) I,* or *verotoxin I,* and *SLT II,* or *verotoxin II*. [100] It is known that SLT I has the same biologic properties as, and is immunologically and genetically indistinguishable from, Shiga toxin. EHEC organisms have two well-recognized virulence attributes—namely, the production of one or more SLTs and attachment and effacement. In addition, all isolates of *E coli* O157:H7 possess a 60-Md plasmid that appears to play an important role in adherence. [101] As has been well established, Shiga toxin and SLTs are lethal to certain cells in culture and are enterotoxic, mediating fluid accumulation in ligated ileal loops. They also can cause limb paralysis when injected parenterally into animals. [102], [103] Bacterial lipopolysaccharide and SLT may stimulate the production of interleukin-1 and tumor necrosis factor from macrophages, which may increase the sensitivity of endothelial cells to SLT by increasing the expression of globotriaosylceramide (Gb3) on the cell surface. [100], [104], [105] and [106] The Shiga toxin then inhibits protein synthesis and the endothelial cells may detach, exposing platelets to the subendothelium and initiating coagulation. [104] It is perhaps through this vascular endothelial damage that infection with *E coli* O157:H7 is capable of producing hematologic abnormalities (i.e., hemolysis and thrombosis). SLT production is not limited to the O157:H7 strain of *E coli,* and non-O157:H7 SLT-producing *E coli* have been isolated from both bloody and nonbloody diarrhea. In addition, culture-confirmed infections with *E coli* O157:H7 can produce a nonbloody diarrhea and can also cause asymptomatic infection. Infections associated with nonbloody diarrhea are less likely to be complicated by hemolytic uremic syndrome. It is likely that the amount, the site of production, and the chemical structure of the toxin, and perhaps the organism's ability to adhere to the intestinal mucosa, all play a role in the development of the particular clinical syndromes. The clinical illness caused by SLT-producing *E coli* is described in Chapter 88.

Salmonella Species

Classification Currently, all *Salmonella* isolates are classified in a single species, *Salmonella choleraesuis,* based on DNA sequence similarity. [107] The species *S choleraesuis* can be subclassified into seven subgroups, of which subgroup I contains almost all the serotypes and isolates pathogenic for humans. The correct taxonomic names for *Salmonella typhi* and *S choleraesuis* are *S choleraesuis* (group I), serotype *typhi,* and *S choleraesuis* (group I), serotype *choleraesuis,* respectively. There are more than 2000 *Salmonella* serotypes, all of which, with the exception of *S typhi* and the paratyphoid strains, have nonhuman reservoirs and are widely dispersed in nature. The classification scheme for the different species and particular biotypes/serotypes within each species is similar to that described for *E coli*. The somatic or polysaccharide O-Ag, the flagellar H-Ag, and the Vi-Ag are markers of *Salmonella* serotypes displaying varying degrees of host-specific adaptation and the ability to cause different clinical syndromes. *Salmonella* organisms are gram-negative, non–spore-forming, motile, facultatively anaerobic bacilli.

Nontyphoid Salmonellosis

Epidemiology. Nontyphoid *Salmonella* species have been, and continue to be, one of the most frequent causes of foodborne enteric infection in the industrialized world. *Salmonella* is estimated to cause more than 1.5 million cases of foodborne illness in the United States annually, [10] with most caused by *S enteritidis*. *Salmonella* also was the most frequently reported cause of foodborne disease outbreaks between 1993 and 1997, accounting for 13% of all outbreaks. [13] In addition, *Salmonella* caused more deaths (44.8%) than any other pathogen in these outbreaks reported to the CDC. The incidence of nontyphoid salmonellosis still continues to be a major problem in the United States. *S enteritidis* and *Salmonella typhimurium* are the most commonly isolated *Salmonella* serotypes. Nontyphoid *Salmonella* is widely distributed among different animal species and is particularly prevalent among animals raised for food. *Salmonella* serotypes, particularly those belonging to *S enteritidis,* have been isolated from a wide variety of animals, including poultry and all types of farm animals. Food and milk are commonly associated with diarrhea outbreaks, but many notable outbreaks have been traced to numerous other sources (e.g., pet turtles, marijuana). The major reservoirs for *Salmonella* organisms that cause human illness, however, are poultry and domestic livestock. One study found 50% of poultry in retail stores to be contaminated with viable *Salmonella* species. [108] An increase in the incidence of the nontyphoid *Salmonella* serotypes associated with reptiles and an overall decrease in the incidence of those associated with meat, poultry, and eggs have been noted. [109] This may in part be the result of programs that focus on reducing the risk for *Salmonella* exposure in food products to consumers. Eggs and egg products often have been identified as sources of epidemic salmonellosis. mall cracks may allow passage of the organism into the interior of an egg while it is being laid (contact with feces) or cleaned, or vertical contamination in utero before the shell calcifies. In the United States, an estimated 0.01% of intact eggshells contain *S enteritidis*. [110], [111] Person-to-person transmission of salmonellosis is thought to be minor, but it may be important in institutional settings (including day care centers), where fecal contamination of the environment and inadequate hygiene are common. About 85% of all outbreaks, however, occur through the consumption of contaminated food or drink, and only 10% through cross-infection. Both *S typhi* and nontyphoid *Salmonella* species show a seasonal variation, and the frequency of illness reaches a peak in the summer and fall, when picnics and barbecues are common and the cooking practices necessary to kill the *Salmonella* organisms (>150°F for 12 minutes) are often not followed. There is no sex preponderance, and attack rates are greatest in children younger than 5 years of age, followed by persons 20 to 30 years of age and older than 70 years. If one household member becomes infected, the probability that another will become infected is 50% to 60%. [112] Patients with malignancies, immunosuppression, alcoholism, sickle cell disease, an abnormal cardiovascular system, hemolytic anemia, schistosomiasis, or a major underlying disease are predisposed to progressive salmonellosis with bacteremia, as are postsurgical patients. A case fatality rate of 0.2% has been reported in epidemics, but the overall mortality rate averages 2% to 3% if any of the predisposing factors are present. The case fatality rates are highest in nursing home residents and infants in nurseries (7%–9%).

Pathogenesis. The development of salmonellosis hinges on the number of organisms ingested (10^3–10^8) and on predisposing host factors. The most important factor that allows a smaller dose to cause infection is a lack of gastric acidity (e.g., neonatal period, achlorhydria, gastric surgery, antacid use). [113] For diarrhea to develop, viable organisms must reach the small intestine and invade the intestinal mucosa. *Salmonella* organisms must traverse the acid barrier of the stomach and the mucous layer of the intestinal epithelium while evading intestinal, pancreatic, and gallbladder secretions. After crossing the small bowel mucous layer, *Salmonella* organisms interact with both enterocytes and microfold (M) cells. On contact with M cells, the organisms are rapidly internalized and transported into the submucosal lymphoid tissue, where they may enter the systemic circulation. Unlike *Shigella* species, *Salmonella* organisms usually do not cause epithelial damage by invasion. Salmonellae induce nonphagocytic cells to internalize them by means of fimbrial adhesins. [114] Salmonellae are internalized within membrane-bound vacuoles of epithelial cells. [115] After the organisms penetrate the epithelial barrier, they rapidly interact with macrophages and lymphocytes in Peyer patches in the small intestinal submucosa. [116] The ability of *Salmonella* organisms to delay lysosomal fusion and to attenuate the acidification of phagosomes likely contributes to their survival within macrophages. Rarely, nontyphoid *Salmonella* species can produce more extensive lesions, consisting of an intense colitis with small hemorrhages and ulcerations of the surface epithelium and multifocal microabscesses. Some nontyphoid *Salmonella* serotypes, especially *S choleraesuis,* show a much greater ability to invade. Invasion of the bloodstream by nontyphoid *Salmonella* species occurs in just under 10% of cases. The pathogenic mechanism of *Salmonella* gastroenteritis is poorly understood. Many of the genes that are important to the virulence of *S typhimurium* are regulatory proteins that control the synthesis of multiple proteins at the level of gene transcription. The *Salmonella* virulence plasmid appears to be important in pathogenesis, as do the major surface carbohydrate-containing molecules of *Salmonella,* such as the lipid A component of lipopolysaccharide. Host factors appear to be very important in contracting and warding off disease caused by *Salmonella* organisms. The increased incidence of nontyphoid bacteremia in AIDS patients and transplant hosts suggests that cell-mediated immunity is important in controlling these organisms. As mentioned previously, alterations in the gastric acid barrier predispose individuals to infection.

Clinical features. *Salmonella* infection in humans comprises four clinical syndromes: gastroenteritis, enteric fever, bacteremia, and asymptomatic carriage. The features of *Salmonella* gastroenteritis are indistinguishable from those of gastroenteritis caused by many gastrointestinal pathogens. Symptoms usually develop within 48 hours after the ingestion of contaminated food or water. At one end of the spectrum of disease is a very mild illness characterized by the passage of one to a few loose stools and only minimal symptoms; at the opposite end is a cholera-like illness with profuse diarrhea and significant dehydration. A dysentery-like syndrome is also occasionally seen. Fecal leukocytes are usually present. Fevers, abdominal cramping, nausea, vomiting, and chills are frequently reported. The diarrhea usually lasts 3 to 7 days, and the fever usually resolves within 2 to 3 days. Improvement is expected after the initial 36 hours. Bacteremia is uncommon with this type of illness (6%–8%), but it is seen more frequently in children and in people with underlying health defects (15%), in whom the mortality rate is also higher. Serious complications (e.g., osteomyelitis) can occur in association with underlying disorders such as sickle cell disease and other hemolytic diseases. Bacteremia and focal abscesses, which can result in septic shock, are complications seen in patients with AIDS and patients who have malignancies or are undergoing therapy for malignancies. Finally, infected aortic or iliac aneurysms in patients with preexisting atherosclerotic vascular disease can result in mortality rates higher than 60%. After the gastroenteritis resolves, the mean duration of carriage of nontyphoid *Salmonella* in the stool is 4 to 5 weeks and varies by *Salmonella* serotype. [117]

Therapy. Antimicrobials should not be used routinely in mild to moderately ill patients with gastroenteritis because their use may prolong the intestinal carriage of nontyphoid *Salmonella* species and may increase the incidence of bacteriologic relapse. [118], [119] Indications for antimicrobial therapy are predisposing factors that can complicate salmonellosis, including extremes of age, an immunodeficiency condition, signs and symptoms of sepsis, infection with organisms likely to produce bacteremia (e.g., *S typhi, Salmonella paratyphi, S choleraesuis*), focal infections (e.g., osteomyelitis, abscesses), and chronic typhoid carrier states. Drug resistance among *Salmonella* species (including *S typhi, S typhimurium,* and certain serotypes of *S enteritidis*) is increasing. A multidrug-resistant strain of *S typhimurium,* referred to as *definitive type 104 (DT104),* has emerged as a significant problem in United States and elsewhere. [120] DT104 and other multidrug-resistant *Salmonella* species demonstrate resistance to such antimicrobials as chloramphenicol, tetracycline, ampicillin, TMP-SMX, and the aminoglycosides. This resistance is mediated by large complex plasmids. [121], [122] In seriously ill persons with nontyphoid *Salmonella* infection, it is reasonable to administer two antimicrobial agents from different classes until the sensitivities are known. Agents that can be used (see Table 74-3) are ampicillin, amoxicillin, TMP-SMX, cefotaxime or ceftriaxone, chloramphenicol,

and FQs. Low-grade bacteremia, not involving the vascular structures, should be treated with 7 to 14 days of intravenous antimicrobial therapy, with 6 weeks of therapy reserved for documented or suspected intravascular infection. Persons with AIDS and a first episode of *Salmonella* bacteremia should be given 1 to 2 weeks of intravenous therapy followed by 4 weeks of an oral FQ to eradicate the organism and lessen the risk for recurrent bacteremia. Those with recurrent bacteremia may require suppressive therapy. Amoxicillin and TMP-SMX given for 6 weeks have a success rate of more than 80% in eradicating the chronic carriage state of *Salmonella*. [123], [124] FQs are also recommended in the treatment of the chronic carrier state. Antimicrobial agents, however, are not very effective in eradicating the carrier state in persons with anatomic abnormalities (e.g., biliary or renal stones).

Typhoid (Enteric) Fever

Clinical features. Enteric fever can occur with any *Salmonella* serotype, but most commonly with *S typhi* and *S paratyphi*. The hallmarks of enteric fever are prolonged fever and abdominal symptoms. The symptoms are often nonspecific and may be insidious in onset. The diagnosis should be considered strongly in a traveler with fever who has returned from a subtropical or tropical area. *S typhi* usually is transmitted through the ingestion of material contaminated by human feces, either from ill people or asymptomatic carriers. Because humans are the only reservoir of *S typhi,* the factors that directly relate to the prevalence of this organism in the environment are a lack of water sanitation and the number of carriers. In the United States, the rate of typhoid has dramatically decreased (in contrast to the rate of nontyphoid salmonellosis), but typhoid continues to be a prevalent endemic problem in developing nations. The incubation period of *S typhi* is approximately 1 week (range, 5–21 days). Following ingestion of the organism, enterocolitis may develop with diarrhea lasting several days, which usually resolves before the onset of fever. After this initial phase, the clinical features of typhoid are those of an acute febrile illness, lasting an average of 3 to 5 weeks and accompanied by specific and nonspecific signs and symptoms. The nonspecific symptoms appear early, are usually insidious, and include frontal headache, general malaise, prostration, mental confusion (at times delirium), anorexia, and abdominal discomfort and bloating. Upper respiratory tract symptoms, particularly dry cough, coryza, and sore throat, can be prominent presenting complaints and may lead the physician to misdiagnose the illness. Constipation is common initially but may give way to diarrhea in the late phase of the illness. During the initial phase of fever and nonspecific symptoms, blood cultures are positive in more than 90% of patients, and characteristic clinical signs develop. The liver and spleen frequently are enlarged, and abdominal tenderness can be observed, particularly over the lower quadrants (at times mimicking appendicitis) and over the liver. Thirty percent of patients have typical rose spots, a faint salmon-colored maculopapular rash on the trunk. These occur in the first 1 to 2 weeks and last only 3 to 4 days, but they are not specific for typhoid (i.e., they have been reported with psittacosis, shigellosis, brucellosis, and other illnesses). Relative bradycardia (pulse relatively slow for degree of fever) can be seen in fewer than 50% of patients. Symptoms usually begin to abate after the third week of untreated illness in the 90% who survive. In 3% to 13% of untreated patients (and about 20% of treated patients), relapse can occur, usually after a variable period of time (average, 2 weeks). Relapse tends to be a milder and shorter illness than the full-blown syndrome. Many of the complications of untreated enteric fever develop in the third and fourth weeks of infection. Complications include intestinal hemorrhage and perforation related to hyperplasia of the lymphoid tissue in the ileocecum, in addition to focal infections such as pericarditis, orchitis, and splenic or liver abscesses. Hematologic abnormalities include leukopenia and anemia. The chronic carrier state (excretion for 1 year or longer) is seen in 1% to 4% of patients with *S typhi* and *S paratyphi* infection and is commonly associated with an age younger than 1 year or older than 60 years, concomitant biliary tract disease, and schistosomiasis (important in urinary tract carriage of *Salmonella* species). Chronic carriage is also seen in 0.2% to 0.6% of patients with nontyphoid salmonellosis. The biliary tract commonly serves as a focus of *Salmonella* organism multiplication in typhoid carriers, especially if concomitant derangements are present (e.g., stones, obstruction).

Diagnosis. The diagnosis of enteric fever requires the isolation of *S typhi* or *S paratyphi* from the patient. Cultures of blood, stool, urine, rose spots, blood mononuclear cells, and bone marrow may be useful in making the diagnosis. The diagnosis is established in more than 90% of cases if blood, bone marrow, and intestinal secretions are all cultured. The sensitivity of blood culture alone is 50% to 70%, and of bone marrow alone is 90%.

Therapy. Chloramphenicol is a tried and true treatment for enteric fever, reducing mortality from 20% to 1% and reducing the duration of fever from between 14 and 28 days to between 3 and 5 days. Because of resistance, a high rate of relapse and chronic carriage, and potential bone marrow toxicity, however, chloramphenicol has been replaced as the intravenous antimicrobial of choice. Third-generation cephalosporins and FQs are effective in the treatment of typhoid fever.

Vaccine therapy. Enteric fever can be prevented by immunization. Parenteral whole-cell and purified capsular polysaccharide vaccines (Typhim Vi; Aventis Pasteur, Swiftwater, Pennsylvania) and an oral live attenuated vaccine (Ty21a, Vivotif Berna; Berna Products Corporation, Coral Gables, Florida) have been developed. Typhim Vi and Ty21a are available in the United States. Ty21a must be administered in four doses, given every other day for 4 days. Typhim Vi is given as a single injection. These vaccines confer only limited protection for a few years. [125] Efficacy studies indicate that the whole-cell vaccine has a significantly greater efficacy rate than Typhim Vi or Ty21a, but its toxicity profile is much greater. [126], [127] Boosters are recommended for the Ty21a vaccine every 5 years and for Typhim Vi every 2 years.

Yersinia species

Classification *Yersinia* species, members of the Enterobacteriaceae family, are gram-negative, non–lactose-fermenting coccobacilli that are motile at 25°C but not at 37°C. *Yersinia* species have been classified according to surface antigens, somatic (O-Ag) and flagellar (H-Ag), and on this basis, more than 50 serotypes of *Y enterocolitica* and six serotypes of *Yersinia pseudotuberculosis* have been recognized, with specific geographic distributions.

Epidemiology Most reported cases of gastrointestinal illness caused by *Yersinia* species have been in European children. However, *Yersinia* can infect people of all ages [128], [129] and [130] and is distributed widely throughout the world. The incidence of *Y enterocolitica* infection in the United States is unknown, but it accounts for approximately 0.1% of reported foodborne diarrheal illness. [13] *Y pseudotuberculosis* is uncommon in the United States but is more frequently isolated in Europe. Disease in humans presumably occurs through contact with animals, particularly swine. *Y enterocolitica* disease is mainly transmitted by the fecal-oral route, and foodborne outbreaks caused by this pathogen have been reported. Contact with dogs, ingestion of outbreak-associated foods (e.g., chocolate milk, ice cream, tofu), contact with contaminated surface water, and person-to-person transmission have been documented in *Y enterocolitica* infections.

Pathogenesis The virulence of *Yersinia* organisms depends on a heat-stable enterotoxin (Yst) and surface antigen (Myf). [131] Furthermore, V and W antigens confer dependency on calcium for growth at 37°C. Pathogenic strains are resistant to complement, penetrate epithelial cells, are lethal to mice, and demonstrate cytotoxicity. Some of these characteristics are mediated by plasmids. In particular, the 70-kb plasmid encodes virulence determinants that include a secreted protein kinase and an outer membrane protein with protein tyrosine phosphatase activity. [132], [133] *Y enterocolitica* grows better in the presence of iron because it does not have its own siderophore. Patients with iron overload syndromes may be at great risk for *Yersinia* infections. [134]

Clinical Features *Y enterocolitica* infection is usually a self-limiting febrile diarrheal illness. Vomiting occurs in 50% of cases, and abdominal pain is a very prominent symptom. The abdominal pain may be localized to the right lower quadrant, mimicking acute appendicitis to the extent that many unnecessary appendectomies are performed in patients with this illness. Localized right lower quadrant pain rarely is seen in children younger than 5 years of age. Most patients experience a *Shigella*-like illness, with the passage of a few watery stools per day, often blood-streaked, accompanied by abdominal pain, low-grade fever, and constitutional symptoms. The diarrhea and constitutional symptoms tend to diminish by the second or third day of illness, but in some cases the diarrhea may last up to 45 days. Other common symptoms, in order of decreasing frequency, include fever, dysentery, and arthritis, or the patient may have no symptoms. Pharyngitis can occur in 50% of children and 10% of adults. Chronic diarrhea persisting for months is observed in a small number of patients, usually children, but in the vast majority of cases, complete recovery is achieved by the second week of illness. Complications are rare but include appendicitis, intestinal perforation, ileocolic intussusception, peritonitis, toxic megacolon, and cholangitis. These features are seen almost exclusively in children and young adults. Septicemia caused by *Y enterocolitica* is uncommon but may develop in patients with a predisposing illness, particularly iron overload states such as hemochromatosis, cirrhosis, and hemolytic processes. Similarly, *Y enterocolitica* infections can lead to extraintestinal focal suppurative lesions that may involve any region of the body; infants younger than 6 months of age are particularly prone to the development of extraintestinal disease. Postinfectious complications can lead to thyroiditis, glomerulopathy, Reiter syndrome, carditis, reactive arthritis, erythema nodosum, skin rashes, ankylosing spondylitis, and inflammatory bowel disease. Except for patients with thyroiditis and glomerulopathy, these patients tend to be HLA-B27 antigen–positive, and their disease may be precipitated by an acute *Yersinia* infection. Molecular mimicry between HLA-B27 and *Yersinia* antigen has been postulated as a mechanism for reactive arthritis. Superantigenic activity has been found in cultures of *Y enterocolitica* and may be a mechanism for reactive arthritis. [135]

Diagnosis Examination of the stool reveals many white blood cells, and frequently red blood cells. The diagnosis can be made by using both bacteriologic and serologic techniques. The laboratory personnel should be alerted if *Yersinia* infection is suspected because standard processing of stool specimens may overlook this organism, and special media may be necessary.

Therapy No evidence has been found that antimicrobial therapy for *Yersinia* enteritis alters the course of this usually self-limiting illness, and therefore treatment is not commonly advocated. In sharp contrast, antibiotics should be administered for septicemic illness; however, mortality rates of 50% for *Y enterocolitica* and 75% for *Y pseudotuberculosis* infection are the rule despite antibiotic therapy. Antibiotics have no effect on the postinfectious complications. Aminoglycosides, tetracycline, chloramphenicol, TMP-SMX, piperacillin, and third-generation cephalosporins usually are effective, although resistance to ampicillin and other antibiotics has been reported (see Table 74-3). [136] A 3-week (or longer) course of antibiotic therapy is recommended for the treatment of extraintestinal disease. *Y pseudotuberculosis* usually is susceptible to ampicillin in addition to the antimicrobials already listed.

Vibrio Species

An estimated 5.5 million cases of cholera occur worldwide each year, with more than 100,000 deaths. [137] The illness is caused by *Vibrio cholerae* O-group 1, a member of the Vibrionaceae, which also includes *Aeromonas* species and other *Vibrio* species. Non-O1 strains, such as O139, have also been associated with epidemic cholera. [138] Cholera is endemic in southern Asia, Africa, and Latin America, where the disease results in high mortality. The risk for cholera acquired in the

United States is small. The ingestion of contaminated seafood from the Gulf Coast or contaminated imported food and travel to areas of endemicity have led to an increase in reported cases of both serogroup O1 and serogroup O139. More antimicrobial resistance is also being reported. [139]

Classification Gastroenteritis can be caused by seven of ten known pathogenic *Vibrio* species (*V cholerae, V parahaemolyticus, V fluvialis, V mimicus, V hollisae, V furnissii,* and perhaps *V vulnificus*), of which *V cholerae* and *V parahaemolyticus* are the most important. *V cholerae* is divided into 139 serotypes on the basis of the O-Ag of the cell surface lipopolysaccharide. The organism is a motile, monoflagellated, short, curved, gram-negative rod. It grows best in thiosulfate citrate bile salts (TCBS)–sucrose agar and is inhibited by the selective media commonly used in routine stool cultures. [140], [141] Strains associated with endemic or epidemic cholera are designated as O1, and the rest are commonly described in general as non-O1 *V cholerae*. *V cholerae* O1 is divided into two biotypes, classical and El Tor. Both the classical and the El Tor biotypes can be subdivided according to subspecificity of the O1 antigen as Ogawa, Inaba, or Hikojima.

Vibrio cholerae

Epidemiology. *V cholerae* O1 is a free-living inhabitant of brackish water and estuaries. Survival is prolonged in cold, saline conditions; the organism has the ability to change from a viable, culturable form to a nonculturable form under certain conditions. [142] Water, specifically fecally contaminated water, is the major vehicle of transmission for *V cholerae* O1, although food is increasingly implicated in disease transmission. The previous exposure of a population is an important factor in the risk for contracting cholera. Patients who recover from *V cholerae* O1 infection have long-lasting immunity. Person-to-person transmission is not thought to play a major role in the propagation of cholera, and chronic human carriage is rare. Persons with low levels of gastric acidity resulting from malnutrition, gastritis, surgery, or antacids are at increased risk for cholera infection. Bottle feeding may also be a risk factor for cholera in infants. Where endemic, cholera affects primarily children between the ages of 2 and 9 years, with a second peak occurring in women of childbearing age. In areas of endemicity, attack rates are higher in women than in men, perhaps because of cultural norms whereby women may have greater exposure to the organism through domestic chores, such as washing and caring for small children. All age groups and both sexes are affected equally when cholera is introduced into a previously uninfected area. For as yet undiscovered reasons, people with blood type O tend to experience clinically more severe cholera.

Pathogenesis. Pathogenesis requires colonization followed by the elaboration of numerous toxins, the best known being an enterotoxin. The cholera toxin of *V cholerae* O1 and its effect on the surface epithelial cells of the gut has been well studied and has contributed much to our understanding of the pathophysiology of diarrhea. [143] After ingestion, *V cholerae* O1 must pass through the gastric acid of the stomach to colonize the small intestine. The organism must then penetrate the mucous gel, adhere to the intestinal epithelial cells, and produce a number of extracellular proteins, such as cholera toxin, neuraminidase, and hemolysin. [144] Pili are important factors in the adherence of vibrios to the small intestine. The classical biotype of *V cholerae* O1 produces a pilus, the toxin-coregulated pilus, that is necessary for human colonization. [145], [146] Cholera toxin is a protein with a molecular weight of 84,000 d and six structural subunits: five binding B subunits and one active A subunit composed of two peptides (A1 and A2). The B-subunit pentamer binds holotoxin to the enterocyte surface receptor, the ganglioside GM_1. The A1 fragment catalyzes the ADP ribosylation of a GTP-binding protein and causes persistent activation of adenylate cyclase. The resultant increase of cAMP within the intestinal epithelial cells stimulates chloride secretion and decreases sodium reabsorption, leading to a loss of fluid and electrolytes and the production of watery diarrhea. Although the pathogenesis of the classical cholera syndrome has long been attributed to the effects of an elaborated enterotoxin, *Vibrio* can produce illness even if the bacteria do not possess the cholera toxin gene. Some strains of *V cholerae* appear to produce other enterotoxins, such as zonula occludens toxin and accessory cholera toxin, that may play a role in pathogenesis.

Clinical features. The clinical symptoms present a spectrum ranging from subclinical gastroenteritis to severe cholera (cholera gravis). The mild to moderate illness caused by cholera cannot be distinguished from other forms of enterotoxigenic diarrhea, and many affected persons do not seek medical attention. By contrast, severe, complicated cholera, now relatively rare if oral fluid therapy is made available, can lead to hypovolemic shock within 1 hour and death within 2 to 3 hours if left untreated. The key feature is that fluid and electrolyte loss leads to serious complications in children, including severe metabolic acidosis, hyponatremia, hypokalemia, lethargy, altered sensorium, and seizures. Clinical signs of such profound dehydration include wrinkled skin with minimal turgor, sunken eyes, cold extremities, restlessness, dry mucous membranes, depressed fontanels in infants, weakened voice, thready pulse, and systolic hypotension. Severe cholera starts abruptly with diarrhea, which becomes the classical rice water (watery stools with flecks of mucus) of cholera. The incubation period ranges from a few hours to 7 days, with some patients experiencing premonitory symptoms of abdominal cramping and anorexia. In most cases, however, the onset of the illness is sudden. The rate of diarrhea production, which can exceed 1 L/hour, usually peaks within the first 24 hours and slowly declines thereafter. Vomiting may complicate and exacerbate dehydration, but it usually abates when the patient is properly hydrated. Fever is rare, as is abdominal pain. Malnutrition in children places them at higher risk for the development of severe dehydration and prolonged diarrhea despite antibiotics. Full recovery without antibiotics can be expected in 1 to 6 days if rehydration is administered. If rehydration strategies are delayed, prolonged hypotension can lead to acute renal tubular necrosis. Hypoglycemia in children can result in grand mal seizures. Hypoglycemia and altered consciousness are risk factors for death. Paralytic ileus, muscle cramping, weakness, and cardiac arrhythmias may herald hypokalemia, the most common electrolyte abnormality of children in the tropics. Pulmonary edema has been reported in severely acidotic children rehydrated with normal saline solution not containing base, unless augmented with standard oral rehydration salts.

Diagnosis. The diagnosis can be made presumptively based on the clinical manifestations and the laboratory identification of toxigenic *V cholerae* O1 or *V cholerae* O139 Bengal. Stool Gram stain may help identify vibrio-like organisms. *V cholerae* organisms display a typical shooting-star motility that can be seen under dark-field or phase microscopy, but unless specific antisera (Ogawa or Inaba) are added to the stool preparation, a diagnosis of cholera cannot be made with certainty. When cholera is suspected, the laboratory should be alerted to use the proper media and identification techniques.

Therapy. The simple rule of thumb in the treatment of cholera is to replace lost elements and reduce the amount of purging. The first goal is easily accomplished in 92% to 95% of cases with the early institution of oral rehydration solutions endorsed by the World Health Organization (WHO; see section “ Oral Rehydration Therapy” and Chapter 53). [147] Severe dehydration (>10% body weight), incessant vomiting, or a change in mental status requires intravenous rehydration therapy, such as with lactated Ringer solution supplemented with potassium (Table 74-5). Hypoosmolar solutions, or those containing rice, may be more efficacious than standard oral solutions. [148], [149]

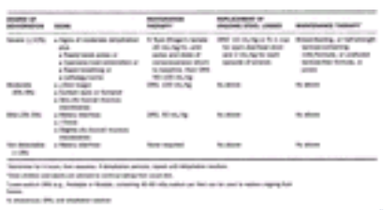

TABLE 74-5 Fluid Therapy for Dehydration

Although rehydration is the only treatment needed, antibiotics can shorten the period of excretion of *V cholerae* and reduce the volume and duration of diarrhea, thereby diminishing the cumulative fluid loss. Tetracycline and doxycycline have proved effective, as have alternatives (streptomycin, chloramphenicol, TMP-SMX, nalidixic acid, and ampicillin). A single 1-g dose of ciprofloxacin was more effective than a 300-mg dose of doxycycline in one study. [150]

Vaccine therapy. Currently, only a parenteral inactivated vaccine is available in the United States. An oral killed whole-cell and oral live attenuated vaccine are available outside the United States. Vaccines in general provide about a 50% rate of protection from disease for only 24 months, do not reduce the rate of asymptomatic infections, and do not prevent disease transmission (see section “ Enteric Vaccines”). [151], [152] Vaccines are not effective against non-O1 strains. Apart from *V cholerae* O1, a number of other *Vibrio* species can produce illness in humans. These pathogenic noncholera vibrios are encountered frequently in the marine environment or associated with seafood, and are known to cause or are suspected of causing gastroenteritis. [153]

Vibrio parahaemolyticus *V parahaemolyticus* is a halophilic (i.e., salt-loving) vibrio. It is commonly isolated from marine waters, their sediment, their inhabitants (fish, crustaceans, and shellfish), and, less commonly, from soft tissue infections of people who have come in contact with seawater. Isolation of the organism from asymptomatic people is rare. *V parahaemolyticus* is a major etiologic agent of bacterial diarrhea in Japan and has been implicated frequently as causative agent of outbreaks of food poisoning in the United States. Nearly all confirmed cases have been associated with either the consumption of raw or improperly stored (after cooking) seafood or the contamination of food with seawater. Disease presentation is variable, ranging from a mild, watery diarrhea to a dysentery-like syndrome. Typically, most cases present with an acute onset of diarrhea after an incubation period of less than 24 hours (range, 4–96 hours) resembling that of nontyphoid salmonellosis. Nausea, vomiting, headache, and low-grade fever are common, but severe dehydration is rare. Mortality rates are low, and the elderly and very young are at greatest risk, particularly if they have concomitant underlying illness. The diarrheal effluent is most commonly watery, but it may contain a few leukocytes and, less often, frank blood (<15%). The diagnosis is confirmed by culturing the organism from stool, a procedure that usually requires the use of selective media (i.e., TCBS agar). Treatment should be directed at the prompt replacement of lost fluid and electrolytes, usually orally but intravenously if the clinical situation so requires. The use of antibiotics has not been shown to shorten the natural course of the illness or duration of pathogen excretion. Some experts, however, suggest that tetracycline may be used in those rare instances when illness is severe (see Table 74-3). As with any organism involved in food poisoning, prevention lies in proper food preparation technique (i.e., appropriate cooking and storage temperatures and avoidance of raw or poorly cooked seafood, including oysters and clams).

Non-O1 Cholera Vibrios Non-O1 cholera vibrios are known as such because despite their biochemical resemblance to *V cholerae,* they do not agglutinate in antiserum against the O-group 1 (somatic) antigen. Non-O1 *V cholerae* have been found to cause gastroenteritis in many parts of the United States. They grow well in waters with increased salinity and are commonly found in the estuarine environment during the warm months and in marine animals, particularly filter feeders such as oysters but also other shellfish, in which they may be concentrated. Other foods, including grated eggs and potatoes, have been implicated in disease outbreaks. The

clinical manifestations of the illness range from a cholera-like syndrome to frank dysentery. Non-O1 *V cholerae* organisms have the capacity to produce cholera toxin or another LT that is active in biologic assays. Most organisms in the United States, however, are not enterotoxigenic, and illness tends to be less severe than typical cholera. Epidemiologic studies show that up to 25% of patients in the United States infected by non-O1 vibrios manifest bloody diarrhea. [154] The illness, which usually begins with diarrhea followed by abdominal cramping, fever, nausea, and vomiting after a 12-hour incubation period (range, 6 hours–3 days), lasts 1 to 6 days. The diagnosis is made by culturing the stool on TCBS agar. Antimicrobial treatment has not been shown to reduce the course or severity of the disease, but, as with *V parahaemolyticus,* tetracycline seems a logical choice in cases of severe diarrhea. The other pathogenic vibrios, including *V fluvialis, V furnissii, V hollisae,* and *V mimicus,* are associated with diarrheal illness more frequently than their counterparts, *V damsella* and *V vulnificus*. The epidemiology and features of the diarrheal illness caused by these organisms resemble those of *V parahaemolyticus* and non-O1 *V cholerae* infection; the organisms are found commonly in the marine environment, and illness is associated with seafood consumption.

Aeromonas, Plesiomonas, and Edwardsiella

Aeromonas Aeromonads are gram-negative, oxidase-positive bacteria that are members of the family Vibrionaceae. Aeromonads are commonly identified and isolated from fresh water fish and shrimp. [155] The role of *Aeromonas* as a cause of wound infection and septicemia is well recognized, but controversy still surrounds its role as a cause of gastroenteritis. *Aeromonas hydrophila, Aeromonas veronii* biotype *sobria,* and *Aeromonas caviae* account for more than 85% of all human infections caused by aeromonads. [156] During the acute phase of the diarrheal illness, the isolation of aeromonads in a diagnostic setting should not be difficult because moderate to large numbers of these bacteria are present in fecal samples. In cases of subacute or chronic diarrhea, however, *Aeromonas* may be overlooked if enrichment procedures are not included. *Aeromonas* infections at any site are more likely to occur during the warmer months of the year, but unlike outbreaks caused by other waterborne and foodborne pathogens, clearly defined outbreaks of diarrheal illness caused by *Aeromonas* have never been reported. Some studies have suggested that *Aeromonas* may simply be a nonpathogenic fellow traveler. In several studies, it has been isolated with almost equal frequency from stools of control patients without diarrhea. [157] Alternatively, a study of 1000 children with diarrhea in Australia demonstrated *Aeromonas* strains in 10% of children with diarrhea, as opposed to an incidence of only 1% in age- and sex-matched controls. [158] Furthermore, treatment of gastrointestinal symptoms with antibiotics directed against *Aeromonas* species has resulted in clinical improvement, and serologic conversions have been demonstrated after clinical infection. Infection with *A hydrophila* is commonly associated with exposure to water, and the organism has been demonstrated as a pathogen in both immunocompromised and otherwise healthy adults. The pathogenesis of *A hydrophila* infection is incompletely understood, but virulence probably relates to the ability of the organism to attach to epithelial cells and to the production of heat-labile enterotoxins. [159] Enteric illnesses caused by *Aeromonas* species range from acute watery diarrhea and dysentery to subacute or chronic diarrhea. Symptoms of the former include watery diarrhea that may contain blood and mucus in up to 22% of cases, mild fever, and vomiting. The clinical symptoms of illnesses caused by various *Aeromonas* species do not appear to differ. The symptoms usually last less than 1 week, but they may persist as long as 2 weeks or more in the case of short-term illness, and more than 1 year in the case of chronic illness. Most *Aeromonas* species are normally susceptible to tetracyclines, aminoglycosides, TMP-SMX, third-generation cephalosporins and their analogs, and FQs, but most are resistant to ampicillin. The treatment in *Aeromonas*-associated gastroenteritis is rehydration. Although most disease is self-limiting, it seems reasonable to treat those patients who have stool cultures positive for *Aeromonas* and who have had symptoms for at least 7 to 10 days (see Table 74-3). TMP-SMX is perhaps the drug of choice in cases of *Aeromonas* gastroenteritis. Other agents that may be used include doxycycline or tetracycline, FQs, and aminoglycosides in septic patients.

Plesiomonas The classification of *P shigelloides* is unresolved, with some studies suggesting that this organism be classified with the Enterobacteriaceae rather than the Vibrionaceae. [160] *P shigelloides* is a non–lactose-fermenting, facultatively anaerobic, gram-negative rod that grows on commonly used enteric media. Features associated with *Plesiomonas* enteric infection include travel to the tropics, abdominal pain, and gastrointestinal illness of more than 14 days' duration. The prevalence of *P shigelloides*–associated diarrhea appears to be increasing, probably as a result of migration and travel. [161] In a controlled study from Nigeria, *P shigelloides* accounted for 8% of cases of diarrheal illness in nonurban areas. [162] *P shigelloides* gastroenteritis affects both children and adults, and 70% of identifiable cases are associated with an underlying illness or risk factor, such as the consumption of contaminated food (e.g., seafood) or foreign travel. Virulence factors have not been clearly established. The features of this illness can vary; it may have an invasive character, similar to the illness caused by *Shigella* species, or manifest as an enterotoxigenic diarrhea. Severe abdominal pain and cramping are commonly seen. The untreated illness can last from 2 to 14 days; antimicrobial therapy has been associated with elimination of the organism from stool and concomitant improvement in clinical symptoms. [163], [164] Most strains are resistant to ampicillin in vitro but sensitive to tetracyclines, TMP-SMX, cephalosporins, chloramphenicol, and FQs. The susceptibility of the plesiomonads to the aminoglycosides varies. [165] FQs and TMP-SMX may be the best oral agents for the treatment of uncomplicated plesiomonad diarrhea.

Edwardsiella The genus *Edwardsiella* comprises gram-negative, oxidase-positive rods that are facultatively anaerobic. Three major species are included, but only *Edwardsiella tarda* is thought to cause human disease. *E tarda* is found primarily in water sources and aquatic animals. *E tarda* can cause wound infections, systemic illnesses, and gastroenteritis. [166] It most commonly causes a diarrheal illness that takes either of two forms—a benign secretory diarrhea or a more invasive process resembling dysentery. *Edwardsiella* strains penetrate nonphagocytic cells, produce hemolysins, and are cytotoxic. [167] The nature of the gene associated with this cytotoxicity has been described. [168] Culture remains the diagnostic method of choice. Based on antimicrobial susceptibility patterns but limited available patient data, ampicillin, TMP-SMX, and ciprofloxacin are all reasonable therapeutic choices. [169]

VIRAL PATHOGENS

Viruses from four distinct and diverse nonenveloped viral families are known to cause acute gastroenteritis in humans. Rotaviruses are the most important cause of severe diarrhea in young children worldwide and are members of the Reoviridae, with a segmented, double-stranded RNA genome. Enteric adenoviruses, by contrast, have a double-stranded DNA genome and belong to a unique subgroup of the Adenoviridae. Norwalk and Norwalk-like viruses, referred to in the older literature as *small round structured viruses* according to their morphologic features, and human caliciviruses, with classical calicivirus morphology, are members of the Caliciviridae, a family of positive-strand RNA viruses. Finally, human astroviruses, which are also positive-strand RNA viruses, belong to a separate and distinct viral family, the Astroviridae. These gastroenteritis viruses are shed fecally to varying degrees by symptomatically infected patients and can be identified by electron microscopy. Despite their genetic and morphologic diversity, the viruses cause diarrheal diseases that are virtually indistinguishable, and infection by these agents is generally limited to the gastrointestinal tract. The patient's history and the setting in which the illness has occurred may help to distinguish among these agents clinically. For example, a child with diarrhea who is younger than 5 years of age and from a developed country is most likely to be infected with rotavirus, but infection with enteric adenovirus, classical calicivirus, or astrovirus is also possible. By contrast, Norwalk and Norwalk-like caliciviruses are well-established causes of large outbreaks of foodborne or waterborne gastroenteritis among older children and adults.

Rotavirus

Pathogenesis Rotaviruses are triple-layered icosahedral viruses approximately 100 nm in diameter. [170] The outer protein layer of the triple-layered virion is composed of two viral proteins, VP4 and VP7. [171] These proteins are responsible for attachment and entry of the virus. Antibodies to both VP7 and VP4 neutralize the virus and are felt to be important mediators of protective immunity. A variety of serotypically distinct VP4s and VP7s have been identified, but most human isolates have one of four principal VP7 types and one of three principal VP4 types. [172] Although the precise role of rotaviral serotype in modulating protective immunity is not yet clear, it is generally felt that serotype-specific immunity is important for protection. [173] The second layer of the rotaviral virion is made up of VP6, which constitutes more than 50% of the mass of the virus. This protein is antigenically cross-reactive among various isolates and is the most immunogenic protein during natural infection or immunization. Antibodies to VP6 do not neutralize rotavirus in vitro but may play a role in protective immunity in vivo. [174] The innermost layer of the virion is composed of VP1, VP2, and VP3. These proteins are components of the viral transcription and replication machinery. [175] The three protein layers surround the genome of the virus, which is composed of 11 segments of double-stranded RNA. [176], [177] Most pathogenic human rotaviruses fall into a single genogroup, group A. The genomes of group A rotaviruses share substantial sequence similarity and are able to undergo gene reassortment with one another. Two other genogroups, groups B and C, also infect humans, although less commonly than group A strains. Group B strains have been primarily identified in China, where they have caused large waterborne epidemics of diarrheal disease in adults, whereas group C strains occur sporadically around the world. [172], [178] Rotavirus replication occurs exclusively in the cytoplasm of infected cells. The replication cycle has several interesting features, including a multiple-step binding and entry phase, a high frequency of gene reassortment during mixed infections, and intracellular budding through the endoplasmic reticulum during virion assembly. [179] Rotaviruses are common enteric pathogens in many mammalian and avian species. In most cases, host range barriers appear to restrict rotavirus strains to a single host. Genetic studies indicate that the basis of host range restriction is likely to be multigenic. [172], [180] Rotaviruses exhibit tropism for tissue as well as host. In immunologically intact animals, rotavirus replication appears to be restricted to the mature villus tip cells of the small intestine, [181] with infection occurring from the duodenum to the ileum. The basis for this tissue tropism is not known, but the restriction of infection to the intestine implies that local immune mechanisms are likely to be the primary determinants of protection. [182] In at least some animal models, rotavirus infection is lytic, and infected mature enterocytes are rapidly killed. It is generally felt that diarrhea is caused by the loss of mature villus absorptive cells and the resultant loss of absorptive capacity for water and sodium, in addition to the loss of brush border hydrolases such as lactase. [183] A rotavirus nonstructural protein, NSP4, has been shown to have enterotoxin-like activity in a variety of experimental circumstances [184], [185] and may

play a role in causing diarrhea. [184] Studies have also implicated activation of the enteric nervous system as a mechanism by which rotavirus causes diarrhea. [186] Because of the importance of rotaviruses, much attention has been devoted to the study of viral immunity. In animal model systems, it appears that both humoral and cellular immune mechanisms are responsible for the resolution of primary infection, [187] and children and animals with severe defects of both cellular and humoral immunity become chronically infected. Antibody appears to be the most important determinant of protection from reinfection, and a good correlation has been found in model systems between the level of antibody in the gastrointestinal tract and the degree of immunity. [188] As mentioned, it seems likely that both homotypic and heterotypic forms of humoral immunity exist, and both forms probably play a role in protection following primary infection. [189]

Epidemiology Rotaviruses are the single most important cause of severe dehydrating diarrhea in young children in virtually all areas of the world. Rotaviruses play a less critical role as a cause of milder diarrheal disease in young children. It is estimated that worldwide, rotavirus infections account for 125 million cases of diarrheal disease, 8 million of which are severe, and cause more than 600,000 deaths. [190] The high mortality rate in less developed countries is caused by a lack of readily available treatments, such as oral rehydration therapy. Even in highly developed countries such as the United States, however, rotavirus infection commonly leads to substantial morbidity. [191], [192] and [193] Studies indicate that approximately 1 million cases of rotavirus infection occur in the United States annually, leading to about 65,000 hospitalizations. The overall medical costs, both direct and indirect, have been estimated to exceed $1 billion annually. The annual death rate ascribed to rotavirus infection in the United States has fallen and is now less than 20 cases per year. [191], [192] and [193] Rotaviruses cause illness primarily in young children between the ages of 3 months and 3 years. [194] Although rotavirus infection has its greatest impact on the very young, illness can occur throughout life. Hence, immunity to reinfection is only partial and not long-lived. Rotavirus disease has also been described in elderly and immunocompromised populations. [195], [196] Studies of diarrhea among otherwise healthy adults indicate that rotavirus infection may be more common and more variable in severity than was appreciated previously. [197], [198] Although most patients are managed as outpatients, many require hospitalization. [198] Unlike rotavirus infection in childhood, infection among adults is not restricted to the winter season. [197] Rotavirus infection is generally spread by the fecal-oral route, although in some less developed areas it appears that waterborne transmission can occur. Transmission is highly efficient, even in developed countries, because of the very large load of viruses shed in the feces. Foodborne infection has also been described. [199], [200] In temperate climates, rotavirus infection generally occurs in the cooler, wetter winter months, whereas in tropical countries, infection tends to occur year round. In the United States, a wave of rotavirus illnesses spreads annually across the country, starting in the Southwest in the fall. [201] A similar sequential wave of spread also has been observed in Europe. [202]

Clinical Features Like many viral infections, most rotavirus infections are asymptomatic. This is especially true in children younger than 3 months of age, in whom maternal antibody may protect against illness, and older than 3 years of age, in whom prior exposure provides partial or complete protection. [203], [204] Symptomatic infection usually follows an incubation period of 1 to 3 days. Illness is characterized by the rapid onset of fever, malaise, vomiting, and watery diarrhea. Illness usually lasts between 3 and 8 days. [205], [206] In comparison with other acute enteric infections of children, rotavirus illness is characterized by significantly more vomiting and dehydration. Laboratory tests can show a mild elevation of the blood urea nitrogen and a mild metabolic acidosis. Stools are watery but do not contain red or white blood cells. If appropriate rehydration and other supportive therapy is instituted, recovery is uneventful in 3 to 10 days. In severely immunocompromised children, disease can be prolonged, and spread of infection to the liver has been reported. Rotavirus infection has been associated with a wide variety of other syndromes, including necrotizing enterocolitis and intussusception, but a causal relationship with these diseases has not been established. [172]

Diagnosis The clinical syndrome caused by rotavirus is relatively nonspecific. To make an accurate etiologic diagnosis, it is essential to use one of the numerous available diagnostic assays. However, it is debatable whether, in any individual case, it is necessary to determine the etiologic agent of an acute diarrheal illness in young child in a developed country. The original diagnostic assay for rotavirus detection was immune electron microscopy. This had the advantage of detecting a wide variety of etiologic viral agents in one test but the disadvantage of being labor-intensive, expensive, and relatively insensitive. Numerous solid phase immunoassays for rotavirus detection have been developed during the last 15 years, and these tests generally have sensitivities and specificities in excess of 90%. [207] Many are commercially available. Other tests include nucleic acid hybridization assays of one form or another, sometimes carried out in conjunction with reverse transcriptase-PCR (RT-PCR) assays. These tests can be even more sensitive than the solid phase immunoassays and can be used to determine the serotype of isolates as well. However, they are generally more difficult to perform and more expensive. [208] Rotavirus can also be readily detected by electrophoretic analysis of RNA isolated from stool specimens. [209] This assay is inexpensive and sensitive. Finally, rotavirus can now be cultivated with reasonable efficiency directly from fecal specimens. [210]

Treatment/Prevention The treatment of rotavirus infection is supportive. In most cases, oral rehydration therapy, as recommended by the WHO (see Table 74-5), is entirely sufficient. [211], [212] and [213] Only in the case of very severe vomiting or frank shock is parenteral hydration required. Current dietary recommendations are to begin refeeding with a complete diet as soon as rehydration is complete. Despite secondary lactase deficiency caused by rotavirus infection, restriction of milk intake is usually not required. Breast-feeding should be continued throughout the oral rehydration process. Currently, no effective antiviral medication is available for rotavirus infection. Several experimental studies have indicated that passive, orally administered immunotherapy has a beneficial role in treating acute or chronic infection and in preventing primary infection. [214], [215] Although nonspecific antidiarrheal treatment with opiates does not appear to be indicated, recent preliminary studies demonstrate a potentially useful therapeutic effect of an enkephalinase inhibitor in the treatment of rotavirus diarrhea. [216] The major approach to rotavirus disease is through vaccination, which is discussed in detail in the section “Enteric Vaccines.” Briefly, several live attenuated vaccine candidates currently are being studied and appear promising. [190], [217], [218] A reassortant rotavirus vaccine derived from a simian rotavirus parent was shown to be highly efficacious, but postlicensing analysis disclosed that this vaccine appeared to cause rare cases of intussusception. [219] This vaccine has been withdrawn.

Norwalk and Norwalk-like Caliciviruses

Norwalk virus is the prototype of a group of viruses that are the primary cause of epidemic gastroenteritis throughout the world. [8] In the United States, an estimated 23 million persons become ill with Norwalk-like virus infections each year. [10], [220] Most of the viruses in this group are named for the location of an outbreak (e.g., Hawaii, Snow Mountain, Taunton). Norwalk virus is named for a 1968 outbreak of epidemic gastroenteritis at an elementary school in Norwalk, Ohio, in which half of the teachers and pupils became ill. [221] The identification of Norwalk virus by immune electron microscopy in 1972 was the first proof of a viral cause of diarrhea. [222] Despite this early success, Norwalk virus proved difficult to study because it could not be propagated in cell culture, no animal models were available, and only small numbers of viral particles were shed in the feces of infected individuals. Newer methods for detecting Norwalk and related viruses became available in the early 1990s after cloning and sequencing of the viral genome [223], [224] and [225] and expression of the single capsid protein as viruslike particles (VLPs). [226]

Pathogenesis Human volunteer studies have helped to elucidate the pathogenesis of illness aused by Norwalk and Hawaii viruses. [227], [228], [229], [230] and [231] Jejunal biopsy specimens from symptomatically infected patients showed broad and blunted villi, crypt cell hyperplasia, cytoplasmic vacuoles, and infiltration of the lamina propria with polymorphonuclear and mononuclear cells. Specimens from the stomach and colon were unremarkable histologically. Jejunal specimens taken after resolution of the symptoms showed a return of the normal villus architecture. Viral particles were not evident in electron micrographs of intestinal biopsy specimens. [227] Functional studies showed a decrease in intestinal brush border enzyme activities [227] and delayed gastric emptying [232]; the latter may contribute to the nausea and vomiting experienced by infected persons. Immunity to infection with Norwalk virus is not well understood. [233], [234], [235] and [236] Various studies indicate that individuals who have been infected with Norwalk virus and in whom an immune response develops (measured by a rise in serum antibody level) are protected from reinfection for a few weeks to a few months (short-term immunity). However, the level of serum antibody does not predict an individual’s susceptibility to infection or illness in the long term. Issues such as the type of antibody (neutralizing versus nonneutralizing) and the role of genetically determined factors in long-term immunity require further study. Immune responses to purified recombinant Norwalk VLPs fed to human volunteers [237] or such VLPs expressed in plants fed to mice (edible vaccine) [238] and the possible utility of these reagents for vaccination are under investigation.

Epidemiology Outbreaks of gastroenteritis caused by Norwalk and related viral strains have occurred year round in a variety of settings, including homes, schools, hospitals, cruise ships, swimming pools, and military facilities. [239] Epidemiologic investigation has shown associations with the ingestion of contaminated food, particularly raw or poorly cooked shellfish, and uncooked or previously cooked foods, including frosting, salads, and sandwiches, that require manual preparation and handling by food service workers. Contaminated water and ice have also been implicated as causes of outbreaks. Primary attack rates of 50% or higher and secondary attack rates of 30% or higher are typical. Transmission is primarily through the fecal-oral route, but person-to-person, airborne, or fomite transmission has been suggested in certain outbreaks in which fecal-oral transmission was unlikely. [240], [241], [242] and [243] In most studies from developed countries, antibody acquisition curves (determined by older [244] and newer [245], [246] and [247] diagnostic methods) show a precipitous rise that begins in the teen years and reaches a plateau of 50% or higher by middle age. Most people in developing countries acquire antibody to Norwalk virus in childhood. It should be noted, however, that Norwalk-like viruses such as Toronto virus (previously called *minireovirus*) infect infants and produce a diarrheal illness of sufficient severity to warrant hospitalization in many cases. [248] With the refinement and broader use of molecular detection assays has come the recognition that Norwalk-like viruses are a significant cause of childhood diarrhea. [249], [250] Common features of Norwalk virus outbreaks that can be used to make a tentative diagnosis in outbreaks of viral gastroenteritis include the following [251]: (1) Bacterial or parasitic pathogens are not identified; (2) nausea, vomiting, diarrhea, and abdominal cramping with an acute onset are prominent symptoms, with fever and malaise in some cases; (3) illness lasts 12 to 60 hours; and (4) the incubation period is 12 to 36 hours. Newer methods for detecting viral antigen or nucleic acid or immune responses to the virus are used in research studies to identify the viral pathogen involved in the outbreak.

Clinical Features The full spectrum and distribution of symptoms in human volunteers reflect what is seen in natural infections. The incubation is 10 to 50 hours, and

symptoms last 12 to 72 hours with no long-term sequelae. Infected patients with symptoms most frequently reported nausea, abdominal cramps, or headache/body ache (96% each), whereas diarrhea was reported by 86%, vomiting by 57%, and temperature above 37.8°C by 32%. [235] Newer methods of viral antigen detection indicate that viruses may be shed for days to 2 weeks after the resolution of symptoms. [235], [252]

Diagnosis Toronto virus, a Norwalk-like virus that was originally found in diarrheal stool from children, was detected by direct electron microscopy (EM). By contrast, adults infected with Norwalk virus tend to shed more soluble antigen than viral particles, and their stool may not contain the concentration of 10^6 viral particles per gram required for visualization by direct EM. Immune EM can circumvent this problem, [222] but it is not available in all medical centers. Newer methods that can detect viral antigen (by ELISA with antibody to recombinant VLPs), the presence of viral RNA (by RT-PCR), or the serologic response to infection (by ELISA with VLPs as the antigen) are very promising but available only in certain research laboratories. [253] These tests are highly sensitive. With regard to specificity, the ELISA designed to detect viral antigen (based on antibody to recombinant VLPs) detects only Norwalk-like viruses that are closely related to the particular VLP to which the antibody was produced. [235], [254] By contrast, the ELISA that uses VLPs as the antigen can detect serum antibody to a broader range of Norwalk-like viruses. [255] For example, if recombinant Norwalk virus VLPs are used as antigen in the ELISA, antibodies to Snow Mountain virus, a Norwalk-like virus of a distinct genogroup, can also be detected. The success of RT-PCR depends on selecting the appropriate primer set(s) and removing substances in stool that may be inhibitory to RT or PCR. [253] Some strains of Norwalk-like virus may not be detectable with currently available primer sets. However, primers based on sequences from one of the more conserved regions of the genome encoding the viral RNA-dependent RNA polymerase have been helpful in obtaining initial sequences for several Norwalk-like viruses. RT-PCR is a powerful technique that has been applied to outbreak investigations and has helped to establish links between outbreaks and also links to contaminated vehicles of transmission.

Treatment/Prevention No specific therapy is available for this generally mild and self-limiting illness. In a study of young children in the community, acute gastroenteritis caused by Norwalk-like viruses was noted to be more severe than illness caused by Sapporo-like viruses (human caliciviruses with classical ultrastructural features). [256] Hospitalization and intravenous fluid resuscitation are rarely necessary. [249] Oral rehydration (see Table 74-5) and other supportive measures may be helpful during the acute phase of the illness. [257] BSS may decrease abdominal cramping. [258] Prevention starts with good hand washing and hygienic food preparation techniques. The public should be made aware of the risks of eating raw shellfish. The common practice of steaming clams until their shells just open (<1 minute) does not achieve the internal temperature of 100°C (4–6 minutes) required to inactivate viruses. [259] Public health efforts should be directed at properly chlorinating drinking water, developing methods to identify accurately shellfish and foods that are contaminated with enteric viruses, and identifying infected food handlers who may be asymptomatic.

"Classic" Human Calicivirus

The genome organization of the morphologically typical caliciviruses (or Sapporo-like viruses) more closely resembles that of animal caliciviruses (e.g., rabbit hemorrhagic disease virus) than that of the Norwalk-like viruses discussed previously. [260], [261] and [262] Differences in the nucleic acid and amino acid sequences of these caliciviruses have placed them in a distinct genogroup. [239]

Pathogenesis The virus is believed to replicate in the small bowel, but limited information is available.

Epidemiology Infection with caliciviruses was first described in infants and young children who had acute gastroenteritis, but it is not restricted to this age group. [263] Infection in adults, particularly elderly individuals, has been documented. [264] Outbreaks in a variety of institutional settings also occur, but less frequently than outbreaks caused by the Norwalk-like caliciviruses described previously. Seroepidemiologic studies of calicivirus strains isolated from infants in the United Kingdom indicate that this is a common infection among infants. [265], [266] and [267] Antibodies are acquired early in life, and high titers of antibodies can be seen in nearly all children (69%–100%) by the age of 12 years. In contrast to Norwalk virus antibodies, antibodies to the morphologically typical caliciviruses appear to be associated with long-term resistance to illness. [268] Infected children tend to excrete large numbers of viral particles that can be readily recognized by EM. Although the mode of transmission has not been clearly established, the fecal-oral route is likely. Oysters and cold foods have been identified as vectors of transmission. [269]

Clinical Features Vomiting and diarrhea are the most common symptoms associated with calicivirus infection. Patients may also experience abdominal pain, fever, and respiratory symptoms. Illness lasts 1 to 2 days and is preceded by an incubation period of 1 to 3 days. Calicivirus illness in infants and young children resembles a mild form of rotavirus illness. [270]

Diagnosis Because these viruses are shed in relatively large quantities in stool and have a distinctive appearance, direct EM is useful in diagnosing infection. An ELISA that detects all classic human caliciviruses has been developed but is not available commercially. [266], [271] Nucleic acid sequence data should hasten the development of molecular detection tests and VLPs. [262]

Treatment/Prevention The recommendations for treatment and prevention are similar to those enumerated for rotavirus and Norwalk virus in the previous sections.

Astrovirus

Pathogenesis Limited information is available on the pathogenesis of human astrovirus infection. The finding of astrovirus particles in intestinal epithelial cells of two patients who were shedding astrovirus in their stool suggests that astrovirus replicates in these cells. [272] The significance of this observation is not clear, however, because both children had severe underlying gastrointestinal pathology and chronic diarrhea. When immunocompetent lambs were infected with an outbreak strain of ovine astrovirus, diarrhea developed in the animals and astrovirus was found in mature enterocytes and subepithelial macrophages. [273], [274], [275] and [276] Viral aggregates were observed in some enterocytes, along with villus atrophy and crypt cell hyperplasia.

Epidemiology Astrovirus infection of humans occurs worldwide. Although astrovirus is primarily a pediatric pathogen, [277], [278] and [279] illness has occurred among institutionalized elderly patients [280], [281] and [282] and immunocompromised individuals. [283], [284], [285] and [286] Symptomatic illness tends not to develop in most otherwise healthy adults, except when they are exposed to contaminated food or water with high viral titers. Antibody acquisition begins in childhood, and antibody prevalence rates of 70% by school age have been reported. [286] Antibody to five of the eight serotypes of human astrovirus are detectable in gamma globulin pools from the United States [287] and Japan [277] (reagents to test for antibody to the three more recently described serotypes were not available at the time of these studies). Large-scale epidemiologic studies based on an ELISA capable of detecting all serotypes of human astrovirus have demonstrated the medical importance of astrovirus infection. [288], [289] Astrovirus was shown to be second only to rotavirus as a cause of viral gastroenteritis among children attending outpatient clinics in Thailand.

Clinical Features Gastroenteritis caused by astrovirus is usually characterized by a mild, watery diarrhea that lasts for 2 to 3 days and may be accompanied by vomiting, fever, and abdominal pain. [290] The incubation period varies from 1 to 4 days. In children, the symptoms of astrovirus gastroenteritis may be indistinguishable from those attributable to rotavirus infection. Although most cases of gastroenteritis that require hospitalization are caused by rotavirus, in a study from Australia, [291] hospitalization rates for gastroenteritis caused by astrovirus (4.2%) and adenovirus (3.7%) were comparable. Astrovirus was found to be a common pathogen among patients infected with human immunodeficiency virus (HIV) [283] and recipients of bone marrow transplants [284], [292] who had diarrhea. In some immunocompromised patients, prolonged infection and more severe illness may be seen. [285], [293], [294] In normal individuals, the production of serum antibody [295] and lamina propria $CD4^+$ T cells that recognize astrovirus antigens in an HLA-restricted manner [296] are important host defenses against repeated astrovirus infections. Protracted infection with astrovirus has been reported in patients with $CD4^+$ T cells depleted by chemotherapeutic agents, such as fludarabine. [285], [297], [298]

Diagnosis Shedding of up to 10^{14} astrovirus particles per gram of stool has been reported, [285] although shedding of 10^8 to 10^{10} particles per gram is more typical. [293] Since the identification of astrovirus in 1975, direct EM has played a prominent role in the diagnosis of astrovirus gastroenteritis. However, accurate diagnosis by EM may require an experienced microscopist because typically only about 10% of the particles display the characteristic surface star. [263] Astroviruses have been mistakenly identified as small round featureless viruses (e.g., parvovirus and picornavirus) and as Norwalk-like viruses. [299] An ELISA based on a monoclonal antibody that reacts with all eight types of human astrovirus [300], [301] is available as a commercial kit abroad, but not in the United States. This ELISA should facilitate the rapid diagnosis of human astrovirus infection.

Treatment/Prevention Issues regarding treatment and prevention are very similar to those discussed for Norwalk virus and rotavirus in the previous sections. In immunocompromised patients, the treatment of protracted astrovirus diarrhea with immunoglobulin administered intravenously, [285] orally, [292] or by a combination of both routes [302] has met with variable success. Experience with this treatment is limited, and it has not yet been studied systematically in controlled clinical trials.

Enteric Adenovirus

Adenoviruses are a large family of DNA viruses that have been associated with a variety of illnesses. Adenoviruses of subgroup F, specifically serotypes 40 and 41, are also known as *enteric adenoviruses* because of their consistent association with gastroenteritis in infants and young children. [303] Other types of adenovirus have also been identified in stool, but they do not fulfill Koch's postulates, and their role as gastrointestinal pathogens is not established.

Pathogenesis Adenoviruses replicate in the intestinal tract. Up to 10^{11} viral particles per gram of stool are excreted during infection. In one report of a fatal case of

gastroenteritis attributable to adenovirus, viral particles were observed in the nucleus of small intestinal mucosal cells. [304] Nonenteric types of adenovirus have been found in AIDS patients with acute and chronic diarrhea. [283], [305], [306] and [307] Their precise role in this disease entity is not well established. [308], [309]

Epidemiology Enteric adenovirus infections occur worldwide and primarily affect children younger than 2 years of age. [303] In developed countries, where the incidence of adenovirus infection is 4% to 10%, these viruses were formerly considered to be second to rotavirus in importance, but the application of newer assays for virus detection has led to the identification of astroviruses and Norwalk-like viruses more often than enteric adenoviruses. In most developing countries where studies have been conducted, an incidence of 2% to 3% typically has been found. A study from Guatemala, however, detected enteric adenovirus in 14% of patients with diarrhea, whereas rotavirus was identified in 4.7%. [310] With few exceptions, adenovirus infections appear to occur throughout the year and do not peak in any particular season. Infection is transmitted from person to person.

Clinical Features Infection with enteric adenovirus may cause an illness indistinguishable from that caused by other viral agents. The infection is generally mild, but the full spectrum of severity can be seen. An incubation period of about 7 days precedes the symptomatic phase, which is characterized primarily by watery diarrhea and vomiting. [311] Virus is shed in the stools for 10 to 14 days, significantly longer than in rotavirus infections. Respiratory symptoms and low-grade fever may accompany adenovirus gastroenteritis. Young children are most commonly affected. Protracted lactose intolerance and malabsorption have been observed following symptomatic infection with enteric adenovirus. [312], [313] It has also been speculated that adenovirus infection may be related to the development of celiac sprue. [314]

Diagnosis A commercially available, sensitive, and specific ELISA kit, based on monoclonal antibodies to each enteric type of adenovirus, is the preferred method for detecting enteric adenovirus antigen in stool. [315] Adenovirus particles are generally shed in large quantities in infected feces (=10^{11}/g) and can be readily identified by EM, but discerning the enteric serotypes requires immune EM. [316] Molecular methods, including PCR strategies, have been developed in research laboratories to detect minute quantities of enteric adenovirus DNA. [303]

Treatment/Prevention No specific antiviral agents are available. Mild to moderate dehydration may be treated with oral rehydration solutions or intravenous fluids, as indicated. The goal of therapy is to prevent severe dehydration and maintain nutritional intake.

THERAPEUTIC CONSIDERATIONS

Oral Rehydration Therapy

Of all the therapeutic modalities available to treat diarrheal disease, none has been more important than the discovery and development of oral rehydration solutions. [3], [317] These formulations are based on careful pathophysiological studies that characterized precisely the fluid and electrolyte losses in stools of cholera patients. The solutions take advantage of the glucose and sodium cotransport mechanism, in which sodium absorption is stimulated by the entry of glucose or other small nutrient molecules into intestinal epithelial cells. This mechanism of absorption is generally not affected by the infecting microbial agents.

The oral rehydration solution recommended by the WHO is slightly hypertonic and is prepared by mixing in 1 L of water sodium chloride (3.5 g), potassium chloride (1.5 g), sodium bicarbonate (2.5 g) or sodium citrate (2.9 g), and glucose (20 g). These components are available in packets that can be mixed with clean water as needed. Replacement of the glucose with other nutrients, such as amino acids and cooked cereals, does not interfere with the cotransport mechanism. [147] An alternative solution that can be prepared at home consists of 3/4 tsp of table salt, 1 tsp of baking soda, 1 cup of orange juice or two bananas, and 4 tbsp of sugar (or 50–60 g of cooked cereal flour from rice, maize, sorghum, millet, wheat, or potato) in 1 L of clean water. [318] Commonly recommended clear liquids, such as apple juice, carbonated beverages, and chicken soup, do not appropriately replace electrolyte losses and may precipitate osmotic diarrhea. [147], [319]

Despite its salutary features, treatment with an oral rehydration solution does not reduce stool volume. [3] Thus, the patient's hydration status should be monitored by measuring the heart rate, blood pressure, and urine output. It is imperative that the parents of infants with watery diarrhea understand that this life-saving treatment will not eliminate the diarrhea.

The fluid management of patients with diarrhea consists of (1) rehydration, (2) replacement of ongoing fluid losses in stool, and (3) maintenance therapy. A clinical assessment of the patient's degree of dehydration is used to guide management in each of the three areas, particularly the rehydration phase. [320] The approach is summarized in Table 74-5. It is important to note that these parameters were established for infants, but the utility of oral rehydration therapy extends beyond infant patients. Elderly adults are also quite susceptible to dehydration, but it may be more difficult to assess the level of volume depletion accurately, short of a catastrophic event such as circulatory collapse. [321] More reliable objective criteria, such as plasma specific gravity, [322] may help with this assessment.

Antimicrobial Therapy

Decisions regarding treatment with antimicrobial agents [323] frequently must be made at the time of the encounter with the patient on the basis of the history, physical examination, and epidemiologic setting, usually without the benefit of diagnostic test results. These decisions also must weigh the potential risks of treatment, which include side effects, the possible occurrence of superinfection when the normal bacterial flora is eradicated, the contribution to the development of infections that are resistant to common antimicrobials, and the possible induction of disease-producing phage with antimicrobials. Table 74-3 summarizes the indications for antimicrobial therapy and the recommended treatment of specific infections. Such treatment is clearly indicated for only two of the bacterial pathogens of the small intestine discussed in this chapter.

Antidiarrheal Drugs When the use of opiates or intralumenal agents is under consideration, it is important to remember that most cases of acute diarrhea are self-limiting and likely to resolve regardless of the use of these medications. The benefit of relieving the patient's symptoms must be weighed against the potential risks associated with medications that may adversely affect intestinal motility or interact with other drugs the patient is taking.

Opiates and opioids. Opiates and opioids [324] inhibit intestinal motility and thereby provide symptomatic relief of mild to moderate diarrhea. Diphenoxylate and loperamide are synthetic congeners of morphine that, in general, have little potential for abuse. Diphenoxylate (2.5 mg) is combined with atropine (0.025 mg) in a tablet (Lomotil); atropine causes discomforting anticholinergic effects if more than 10 tablets are taken in a day and is included to deter abuse of the diphenoxylate. Loperamide is poorly absorbed from the intestinal tract and poses no potential for abuse because it does not cross the blood-brain barrier. It is available over the counter and is safe for children. It may be as effective as BSS in curbing the symptoms of mild to moderate traveler's diarrhea.

Intralumenal agents. Bismuth, marketed as BSS, has antibacterial properties that are not fully characterized and stimulates fluid absorption. BSS is useful in preventing and treating traveler's diarrhea in adults. [325] It has also been shown to be effective in treating acute diarrhea in children. [326] Because salicylate intoxication is a potential problem, however, caution should be exercised in giving the drug to children with known salicylate sensitivity, patients taking other salicylate-containing medications, and patients with bleeding disorders. [327] Patients should be informed that their stools may turn black as the drug is converted to bismuth sulfide in the colon. Because the salicylate portion of the compound may interfere with the renal excretion of organic acids, the patient's medication list should be reviewed and the patient advised accordingly. A bismuth encephalopathy can develop in HIV-infected patients. [328] Adsorbents, such as kaolin-pectin compounds, alter stool consistency and have been touted as potential toxin binders, but controlled trials have failed to show an effect on stool weight [329] or efficacy comparable with that of loperamide. [52] Other antidiarrheal agents, including a-adrenergic agonists (e.g., clonidine), somatostatin and its analogs (e.g., octreotide), calcium channel blockers, nonsteroidal antiinflammatory drugs, and cholestyramine, have no role in the treatment of acute infectious diarrhea.

Enteric Vaccines Although many etiologic agents can cause diarrheal illness as a consequence of infection of the small bowel, the great majority of illnesses, especially in the young, are caused by a relatively limited array of pathogens. ETEC, EPEC, *Shigella, V cholerae* O1, and *S typhi* represent the critical bacterial pathogens, and rotaviruses are clearly the most important viral pathogens. Progress has been made toward developing vaccines for most of these agents.

Rotavirus. Animal studies have demonstrated that a variety of vaccine strategies, including parenteral immunization with inactivated virus, VLPs, and recombinant viral proteins, are all potentially efficacious. [330], [331] and [332] These vaccination approaches have not been tested in people, however. Other animal studies indicate that oral immunization with attenuated homologous virus, inactivated virus administered in microcapsules, and host range–restricted live viruses also effectively induces protective immunity. [333], [334] Human studies have focused on the effectiveness of animal viruses themselves or reassortant animal viruses containing human VP7 or VP4 as an immunogen. These approaches have been termed *jennerian* and *modified jennerian vaccination,* respectively. Other potential candidates that have been investigated in preliminary studies include homologous human strains repeatedly passed in cell culture or isolated from asymptomatic newborns. [335] In a variety of human trials of a tetravalent monoreassortant vaccine in which a simian virus was used as the genetic backbone, this modified jennerian vaccine was immunogenic and more than 80% effective in preventing severe disease. [336] The results of preliminary studies based on a similar approach involving a bovine/human reassortant vaccine are very encouraging. [218] A modified jennerian rotavirus vaccine, based on a simian rotavirus backbone, was licensed for use in the United States. Unfortunately, following licensure, the vaccine was associated with an excess rate of intussusception and was removed from the market by the manufacturer. [337], [338] Ongoing research is directed at clarifying the precise risk associated with the vaccine, which may be quite small. Future studies will be required to determine if the host range–restricted reassortant vaccines are highly effective in less developed countries and whether other approaches to vaccination, such as parenterally administered VLP or DNA immunization or homologous vaccination with attenuated human strains, are more effective or associated with less risk. Epidemiologic and

immunologic data are not yet sufficient that it can be determined whether vaccines for astroviruses and the human caliciviruses are either necessary or feasible. However, the Norwalk virus is serving as a very interesting model system to determine whether plant expression systems can be used to produce a low-cost “edible vaccine.” [339]

Enterotoxigenic Escherichia coli. Various approaches have been used to construct vaccines against ETEC. [340], [341] and [342] Because immunity to these agents appears to be mediated by local antibody against bacterial surface structures, LT, or ST, most vaccine strategies have focused on local immunization. Major problems in this area include the diversity of the O, H, and fimbrial antigens of the toxigenic strains and the difficulties involved in producing immune responses to the ST. Vaccination approaches that have been investigated to date include a *V cholerae* killed-vibrio/B-subunit vaccine, [340], [341], [343] the administration of purified *E coli* fimbriae or inactivated whole *E coli* in microcapsules, and the use of enteric bacterial expression vectors derived from *E coli, Shigella flexneri,* or *S typhi* to express ETEC LT/ST and colonization factors. [342], [344], [345] These various strategies have shown some success in experimental volunteer challenge systems but have not yet been tested extensively in the field.

Enteropathogenic Escherichia coli. Advances in our understanding of EPEC have accelerated progress toward vaccination in this area. [346], [347] Live bacterial vaccines, in which key virulence factors are deleted, are in the early stages of evaluation in human volunteers. It is too early to know if these approaches will be successful in the field, and it is not entirely clear that a vaccine against EPEC would have a substantial effect on infantile diarrhea.

Vibrio cholerae. Great progress has been made in the field of cholera vaccination since the early 1990s. The existing inactivated whole-cell parenteral vaccine is only modestly effective for just several months. A new approach involves the oral administration of a killed whole-cell preparation together with a purified preparation of the B subunit. This vaccine was shown to be moderately (85%) effective during a 6-month follow-up in Bangladesh [348] and demonstrated varying efficacy in a trial in Peru. [349], [350] Perhaps the most promising studies involved a genetically attenuated live vibrio called *vaccine candidate CVD 103-HgR*. The toxin gene has been removed from this strain. [351] In experiments in volunteers, the vaccine was highly effective in generating protection against challenge. [352] However, the efficacy of this strain in a very large field trial was minimal. [353]

Shigella. Progress toward an effective *Shigella* vaccine has been slow. Studies are under way to determine if conjugate O polysaccharide vaccines may be effective against *Shigella*. [354] The identification of two new enterotoxins [355], [356] offers hope that deletion of these virulence factors may lead to an attenuated but immunogenic live bacterial vaccine candidate. Several promising approaches to developing a live attenuated *Shigella* vaccine with recombinant techniques are being evaluated. [357], [358] and [359]

Salmonella typhi. Several approaches to vaccination against typhoid fever have been quite successful. Ty21a is a licensed orally administered *S typhi* vaccine produced by chemical mutagenesis of *S typhi*. It has been widely tested and has proved safe and 60% to 90% effective. [360], [361], [362] and [363] New live bacterial vaccine candidates, prepared by more precise genetic modification, offer the promise of increased immunogenicity and a reduced dosage requirement in comparison with Ty21a. [364], [365] and [366] In addition, parenteral immunization with *S typhi* Vi polysaccharide conjugated to carrier protein has been shown to provide considerable protective efficacy in human volunteer studies. [367] The Vi polysaccharide vaccine demonstrated more than 90% efficacy in a large field trial of young children. [368]

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CHAPTER 75

George T. Fantry, Lori E. Fantry, and Stephen P. James

CHRONIC INFECTIONS OF THE SMALL INTESTINE

WHIPPLE DISEASE

Whipple disease is an uncommon, chronic, systemic bacterial disease first described in a classical paper by George Hoyt Whipple in 1907. [1] Before 1985, it was estimated that 2000 patients had been afflicted since 1907. [2] Even with the advent of sensitive and specific diagnostic tests, the disease appears to be rare. Whipple disease may involve virtually any organ system in the body, with the predominant pathological changes occurring in the gastrointestinal tract and central nervous system. In most cases, the small intestine is involved, resulting in malabsorption. The clinical presentation is widely variable. Patients may present with classical symptoms of malabsorption; however, extraintestinal symptoms, such as chronic seronegative arthritis or fever of unknown origin, may predominate or predate the gastrointestinal symptoms by many years. Because Whipple disease is uncommon, with a varied presentation and insidious onset, it is often diagnosed late in its course. Before the availability of antibiotics, Whipple disease uniformly progressed to a fatal outcome.

In the early 1950s, further advances were made in our understanding of the histopathological findings associated with Whipple disease and the systemic nature of the disorder when periodic acid-Schiff (PAS)–positive macrophages were demonstrated in biopsy specimens of the small bowel [3], [4] and other organs. [5], [6] and [7] In 1952, the first report of antibiotic therapy resulting in a remission of Whipple disease was published. [8] Subsequently, patients exhibited a dramatic clinical response after treatment with a variety of antibiotic regimens. [9], [10], [11], [12], [13], [14], [15] and [16] In the early 1960s, the characteristic rod-shaped bacillus was found in the mucosa of untreated patients with Whipple disease, [17], [18] but the organism could not be reproducibly cultured. Eventually, these bacilli were identified in nonintestinal tissues. [12], [19], [20], [21], [22] and [23] In 1992, an exciting advance was made when a polymerase chain reaction (PCR)–based molecular genetic approach was used to identify the uncultured bacillus of Whipple disease, which was named *Tropheryma whippelii*. [24] In 2000, the bacterium was successfully subcultured, with establishment of a strain that can be used for further study. [25] These findings have provided a basis for more specific diagnostic testing for this organism. [26], [27]

Epidemiology

Although data are limited because of the low incidence of Whipple disease, a number of interesting features help define its epidemiology. The disease is most common between the fourth and sixth decades of life, the mean age of patients at onset being 50 years, [15], [16], [28], [29] but it can occur at nearly any age. Males are more frequently affected, with a male-to-female ratio of approximately 8:1. [15], [28], [29], [30] and [31] Whipple disease occurs predominantly in Caucasians and has rarely been described in persons of African descent and other ethnic groups. Although the disease occurs sporadically and no clearly defined geographic or environmental risk factors have been established, it appears to be more common in farmers and individuals involved in farm-related trades. [15] This may be an important trend because phylogenetic analysis reveals that *T whippelii* is closely related to aerobic soil organisms. However, *T whippelii* DNA has been found in samples of waste water [32] and in the gastric juice and saliva of persons with no clinical evidence of Whipple disease, [33], [34] and [35] suggesting that *T whippelii* may be an environmental or commensal organism that is ubiquitous. Direct person-to-person transmission has not been documented, although the disease was reported in brothers on two occasions [36], [37] and in a father-daughter pair. [38]

Etiology and Pathogenesis

For many years, Whipple disease was thought to be caused by bacteria. This notion was supported by the finding of gram-positive bacilli in involved tissues, and clinical improvement after treatment and disappearance of the organisms. In addition, the bacteria in infected individuals and involved tissues had a uniform electron microscopic appearance. Unfortunately, culture data were conflicting, and the Whipple bacillus could not be reproducibly cultured.

In the early 1990s, major advances were made when PCR was used to identify genes from the uncultured bacillus of Whipple disease. Bacteria have 16S ribosomal RNA genes that are highly conserved in nature but undergo random mutations with time. The sequences of ribosomal genes can be used to construct evolutionary trees of microorganisms, and the ribosomal gene sequences of new, unidentified microorganisms can be used to estimate their phylogenetic relatedness to known microorganisms. [39] Taking advantage of the fact that bacterial ribosomal genes are conserved and readily distinguished from human sequences, and the fact that human tissue should contain no bacterial genes other than those associated with infiltrating bacteria, Wilson and colleagues [40] were able to amplify a 645–base pair bacterial 16S ribosomal RNA sequence from the duodenal tissue of a patient with Whipple disease. Based on the sequence obtained, the authors suggested that the unidentified bacillus was most closely related to the nocardioforms. Subsequently, Relman and colleagues [24] used the same approach to amplify a unique 1321-base pair bacterial 16S ribosomal RNA sequence from the duodenal tissue of five patients with Whipple disease. This sequence was not detected in tissue from ten control patients without Whipple disease. Phylogenetic analysis of the sequence obtained suggested that the Whipple disease bacillus is a gram-positive bacterium more closely related to the actinobacilli than to the nocardioforms, and the bacterium was given the designation *T whippelii*. Actinobacilli, as well as nocardioforms, are actinomycetes; these are a diverse group of bacteria, most of which are aerobic soil organisms. Detailed analysis of sequences has more precisely placed the organism in the phylogenetic position of the actinomycetes. [41] Further independent evidence for the identity of the Whipple bacillus was obtained when RNA

sequences of *T whippelii* were identified by PCR in peripheral blood mononuclear cells and pleural effusion cells in a patient with Whipple disease, which confirmed the systemic nature of the disease. [42] In two subsequent cases, the Whipple bacillus was identified by PCR in peripheral blood. [43] Interestingly, in those patients who had undergone splenectomy, the bacillus was associated with erythrocytes. Finally, *T whippelii* was detected by electron microscopy and PCR in the vitreous aspirate of a patient with uveitis whose intestinal biopsy specimens had appeared normal by light microscopy. [44] These important discoveries illustrate the usefulness of molecular methods to identify uncultured organisms and the feasibility of developing specific diagnostic tests without having cultured the organism.

In 1997, the Whipple bacillus was isolated and grown in human macrophages inactivated with interleukin-4. [45] Subsequently, Raoult and colleagues [25] successfully propagated *T whippelii* in a human fibroblast cell line (HEL) using an isolate obtained from the aortic valve of a patient with endocarditis secondary to Whipple disease. In addition, these investigators were able to subculture the bacterium and establish a strain of the Whipple bacillus. Furthermore, serologic assays revealed that a significantly larger number of patients with Whipple disease had IgM antibody titers specifically against the bacterium of 1:50 or more than did controls. These critical advances will make it possible to undertake future investigations, such as developing serologic tests for Whipple disease and tests for antibiotic susceptibility, and to define the potential role of this pathogen in other human illnesses.

Because the distribution of *T whippelii* in the environment is unknown, it is uncertain to what extent the disease may be caused by exposure to an unusual pathogen. However, the finding of *T whippelii* DNA in the gastric juice and saliva [33], [34] and [35] of people without Whipple disease and the striking clinical features of the disease, with the persistence of intracellular bacteria in macrophages, strongly suggest that an abnormal host response defect may play a central role in pathogenesis. Dobbins, [46] collecting information on HLA phenotypes through literature searches and personal communications, found that the frequency of HLA-B27 (28%) was higher than would be expected in the ethnic groups affected by Whipple disease. This observation is of interest because it is known that the HLA-B27 phenotype is a host factor strongly predisposing to spondyloarthropathies that may be caused by abnormal interactions of the host with enteric bacterial flora. Despite this observation, the incidence of spondyloarthritis does not appear to be increased in patients with Whipple disease. One study even questioned whether the observed correlation is correct; in a study of 14 patients with Whipple disease in Argentina, the frequency of HLA-B27 (one patient, 7.7%) was no higher than that in a control population. [47] However, other observations suggest the presence of host defects. Patients generally exhibit lymphopenia, diminished skin test responses, and decreased mitogen-induced lymphocyte responses in vitro, and these changes may persist after treatment. [48] The abnormality of cellular skin test responses was confirmed and extended. [49] In a study of 20 patients with Whipple disease, none had skin test responses to streptococcal antigens, whether they were untreated or in remission. Because it is known that streptococcal antigens may cross-react with bacterial antigens of actinomycetes, the authors suggested that patients with Whipple disease may have selective defects in their ability to respond to certain types of bacterial antigens. It has been shown that the expression of CD11b, a complement receptor involved in bacterial phagocytosis on circulating lymphoid cells, [50] is decreased in patients with Whipple disease, suggesting that there may be a molecular basis for defective macrophage function in this disease. Subsequently, decreased monocyte interleukin-12 production leading to reduced T-cell production of interferon-? was demonstrated in patients with Whipple disease. [51] This finding provides confirmatory evidence that an underlying defect of monocyte/macrophage function may lead to the inability of the host response to eliminate the causal bacteria. In addition, it may have important clinical implications in resistant Whipple disease, in which immunomodulating therapy with interferon-? may be beneficial. [52]

Histopathology

The histopathological appearance of the small bowel mucosa in Whipple disease is distinct, unique, and usually diagnostic. The lamina propria is infiltrated by large foamy macrophages that grossly distort normal villus architecture, so that the villi take on a blunted, clublike appearance. The cytoplasm of these macrophages is filled with large glycoprotein granules that stain with PAS (Fig. 75-1). The lymphatic channels in the mucosa and submucosa are dilated, and fat droplets may be seen in the extracellular spaces within the lamina propria, reflecting lymphatic obstruction by enlarged mesenteric lymph nodes.

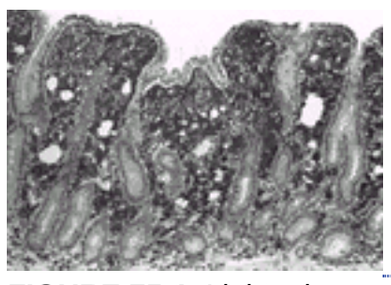

FIGURE 75-1. Light microscopic appearance of the intestinal mucosa in a patient with Whipple disease. The villi are clubbed, and the lamina propria is extensively infiltrated with periodic acid-Schiff (PAS)–positive macrophages. PAS and hematoxylin stain; original magnification ×250. (From ref. [15].)

With electron microscopy, many of the characteristic rod-shaped bacillary bodies can be seen in the lamina propria; they are most abundant just beneath the absorptive epithelium. The fine structure of the bacilli is identical in different patients. The bacilli are 0.25 µm wide and 1 to 2.5 µm long, with a characteristic cell wall and pale central nucleoid. [10], [17], [18], [53], [54] and [55] The bacilli are often seen within the PAS-positive macrophages (Fig. 75-2). Most of the PAS-positive glycoprotein within the macrophages likely represents remnants of the cell wall of the phagocytosed bacilli. Unlike the *Mycobacterium avium* complex, the Whipple bacillus is acid-fast–negative.

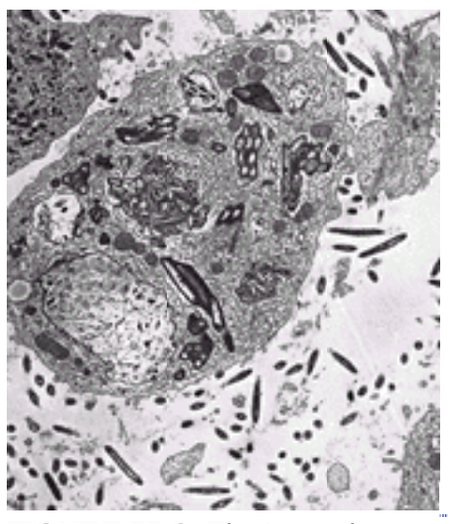

FIGURE 75-2. Electron micrograph showing numerous bacilli in the extracellular spaces and ingested bacilli in varying stages of breakdown within a macrophage. Original magnification ×11,000. (From ref. [15].)

PAS-positive macrophages and the characteristic bacilli have been identified in many nonintestinal tissues, including heart, [19], [56] lung, [20] liver, [12] and brain, [21] documenting the systemic nature of the disease.

The histology of intestinal Whipple disease was revisited in a study of 48 patients. [57] During treatment, several alterations in the histopathological findings were noted. In addition to the continuous decrease in PAS-positive macrophages, the pattern of mucosal infiltration changed from diffuse to patchy. The cytologic aspects of the PAS-positive macrophages changed substantially, leading to the proposal of four different subtypes. Finally, as had been previously shown, a few PAS-positive macrophages commonly persisted at long-term follow-up of treated patients. Because of this finding, it is recommended that during biopsy, undertaken either to

determine a response before treatment is discontinued or to confirm a clinical suspicion of relapse, sufficient samples be obtained so that electron microscopy can be performed to look for the characteristic bacillus, not just light microscopy alone.

Clinical Manifestations

The clinical features and presentation of Whipple disease are highly variable. Many patients present with gastrointestinal symptoms suggestive of malabsorption, such as steatorrhea, abdominal bloating, and weight loss. However, occasionally the diagnosis is made in patients with extraintestinal symptoms, such as arthritis and fever, but no significant gastrointestinal complaints. Symptoms are often present for several months to years before the diagnosis is made. Extraintestinal symptoms may precede gastrointestinal symptoms by many years.

The gastrointestinal symptoms associated with Whipple disease are nonspecific and may be seen in patients with other malabsorptive states (Table 75-1). Diarrhea is the most common presenting complaint. In a study of 29 patients with Whipple disease, diarrhea was the chief complaint in 39%. [16] In this group, 75% of the patients had diarrhea, a frequency similar to that found in other studies. [28], [29] and [30] It is noteworthy that 25% of the patients did not have diarrhea before the initial evaluation. This finding emphasizes that although diarrhea affects most patients with Whipple disease, it is not essential for the diagnosis of malabsorption or Whipple disease. The diarrhea is usually suggestive of steatorrhea; multiple large, watery, or semiformed stools are passed daily. Other intestinal symptoms that are often present include abdominal bloating, cramps, or distention, anorexia, and occasionally intestinal bleeding, which may be manifested by occult blood [30]; less common symptoms are melena and hematochezia. [7], [58], [59] Weight loss is the second most common presenting complaint in patients with Whipple disease and is present before the initial evaluation in 89% to 100% of them. [16], [28], [29] and [30] Severe cachexia may result from anorexia and malabsorption. Fatigue and generalized weakness are common. Continued malabsorption may result in specific nutritional deficiencies. With the loss of albumin into the intestinal lumen as a consequence of lymphatic obstruction, significant ascites and peripheral edema may develop.

GASTROINTESTINAL	EXTRAINTESTINAL	NEUROLOGICAL
Common	**Common**	**Common**
Diarrhea	Arthritis	Central nervous system
Weight loss	Arthralgias	Dementia
Anorexia	Fever	Myoclonus
Less Common	**Less Common**	Insomnia
Abdominal bloating	Pulmonary	Lethargy
Abdominal cramps	Chronic cough	Headache
Hematochezia	Pleuritic pain	Muscle weakness
	Cardiac	Ataxia
	Congestive heart failure	Cranial nerves
	Symptomatic pericarditis	Deafness
	Symptomatic valvular disease	Dysarthria
	Ocular	Facial numbness
	Decreased visual acuity	Diplopia

TABLE 75-1 Symptoms in Patients with Whipple Disease

Extraintestinal symptoms are very common in Whipple disease and may precede the gastrointestinal symptoms by many years (see Table 75-1). Arthritis is the most common extraintestinal symptom and affects the majority of patients. [16], [28], [29] and [30] It often develops several months to years before the initial diagnosis of Whipple disease and is typically an intermittent migratory arthritis of both the large and small joints. Some patients may have arthralgias only. Permanent joint deformity is rare. Fever is the second most common extraintestinal symptom of Whipple disease. [16] The fever is usually low-grade and intermittent. Because of the systemic nature of Whipple disease, numerous other extraintestinal symptoms develop that reflect involvement of other organ systems. Pulmonary involvement is often manifested by chronic cough or pleuritic chest pain. [1], [16], [20], [60] Cardiac involvement [61], [62], [63] and [64] may be manifested as congestive heart failure, valvular lesions, [61], [62] or pericarditis. [16], [19], [60], [64] Numerous reports of endocarditis secondary to *T whippelii* infection in the absence of overt gastrointestinal disease have been published. [65], [66], [67] and [68]

Although central nervous system (CNS) involvement is thought to be common, symptoms related to CNS Whipple disease may be present in only 10% to 20% of patients (see Table 75-1). [69], [70], [71], [72], [73] and [74] Neurological symptoms may occur in association with gastrointestinal symptoms or as isolated symptoms. [15], [21], [75], [76] The most common CNS symptoms are dementia, paralysis of gaze, and myoclonus. [15] Hypothalamic symptoms such as insomnia, hyperphagia, and polydipsia are also frequently present. [15]

The physical findings in patients with Whipple disease are variable, depending on the organ systems involved (Table 75-2). Frequently, the findings are related to severe malabsorption and the loss of fluid and electrolytes, such as emaciation, muscle wasting, peripheral edema, purpura, glossitis, muscular irritability, and peripheral neuropathy. Hyperpigmentation and peripheral lymphadenopathy are the most common physical finding, [16], [29], [30] seen in more than 50% of patients with Whipple disease. The palpable lymph nodes are usually firm, nontender, and mobile. Abdominal findings often include mild distention and tenderness. Abdominal fullness or mass may be caused by marked enlargement of the mesenteric lymph nodes. Ascites is uncommon but may be seen in association with severe hypoalbuminemia. Hepatomegaly and splenomegaly are uncommon. [16], [30] Additional well-recognized physical findings in patients with Whipple disease include fever, peripheral arthritis, heart murmurs, pleural or pericardial friction rubs, and ocular abnormalities. The neurological examination may reveal findings suggestive of CNS or cranial nerve involvement, such as dementia, ataxia, muscle weakness, sensory loss, and ophthalmoplegia.

COMMON	UNCOMMON
Weight loss	Ascites
Lymphadenopathy	Peripheral edema
Hyperpigmentation	Hepatomegaly
Abdominal distention	Splenomegaly
Fever	Pleural friction rub
Arthritis	Ocular findings
Heart murmurs	
Neurological findings	

TABLE 75-2 Physical Findings in Patients with Whipple Disease

Laboratory Findings Laboratory abnormalities, usually related to severe malabsorption, are very common in patients with Whipple disease. Most have steatorrhea, [16], [30] in addition to low serum carotene levels [16], [30] and hypoalbuminemia. [16], [29], [77] In patients with severe diarrhea and malabsorption, electrolyte disturbances such as hypokalemia, hypomagnesemia, and hypocalcemia may be seen. Anemia is a very common finding. [15], [16], [28], [29] and [30] The anemia is usually normochromic-normocytic, suggestive of chronic disease. Occasionally, it is hypochromic-microcytic, related to iron deficiency; however, macrocytosis secondary to folate or vitamin B_{12} deficiency is unusual. Many patients have an elevated erythrocyte sedimentation rate, and the prothrombin time is often prolonged secondary to malabsorption of vitamin K. [16]

Radiologic and Endoscopic Findings Radiologic evaluation can be helpful, particularly a barium contrast study of the small intestine, the results of which are abnormal in most patients with Whipple disease. [16], [30], [78] The most characteristic finding is marked thickening of the mucosal folds, suggestive of an infiltrative process. This finding is usually more prominent in the duodenum and proximal jejunum, with decreased involvement distally and sparing of the ileum. In addition to the marked thickening of the proximal small bowel, abdominal computed tomography (CT) often reveals massive paraaortic and retroperitoneal adenopathy. [16], [79] Gross endoscopic lesions in the duodenum have been described in Whipple disease. [80], [81] The characteristic finding is pale, shaggy, yellow mucosa in the postbulbar duodenum. Yellow plaques or erosions may also be seen. Although involvement of the esophagus and colon is unusual in Whipple disease, endoscopic lesions similar to those seen in the duodenum, consisting of pale, white, plaquelike nodules, have been described. [82]

Diagnostic Evaluation

Most important in the evaluation of Whipple disease is a high index of suspicion. The diagnosis should be considered in patients with diarrhea, weight loss, arthritis,

adenopathy, and fever. However, it is important to remember that the gastrointestinal symptoms related to malabsorption may not be the prominent complaint and, on occasion, may be absent. In addition, extraintestinal symptoms, particularly arthritis and fever, may be present years before gastrointestinal symptoms develop. Therefore, the diagnosis of Whipple disease should be considered in patients with seronegative arthritis in whom symptoms suggestive of malabsorption develop, such as diarrhea or unexplained weight loss, and in patients with culture-negative endocarditis.

Although the clinical presentation, laboratory findings, and x-ray studies may suggest Whipple disease as a diagnosis, small intestinal mucosal biopsy with histological examination is the diagnostic test of choice and is required for a definitive diagnosis. The histological finding of infiltration of the lamina propria of the small intestine by PAS-positive macrophages containing gram-positive, acid-fast–negative bacilli accompanied by lymphatic dilation is specific and diagnostic of Whipple disease (Fig. 75-3). Electron microscopy should also be routinely performed on the biopsy specimens to verify the presence of the characteristic bacillus. This is particularly important during the follow-up of patients treated for Whipple disease in which PAS positivity persists, often for many years; electron microscopy in successfully treated patients shows disappearance of the Whipple bacillus. [10], [16], [51], [83]

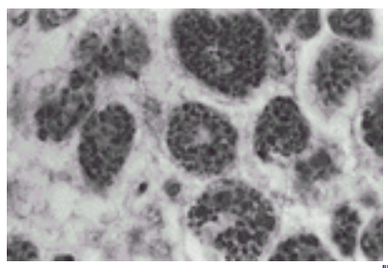

FIGURE 75-3. Light microscopic appearance of macrophages in the intestinal lamina propria in Whipple disease. Note that most of the periodic acid-Schiff (PAS)–stained inclusions are round or sickle-shaped. This appearance is virtually diagnostic, especially in the intestinal mucosa. PAS and hematoxylin stain; original magnification ×1000. (From ref. [15].)

Very rarely, the diagnosis of Whipple disease is established in the absence of intestinal involvement. The diagnosis in such cases is made by the identification of bacilli in involved tissues, including the brain and lymph nodes. Although the colonic mucosa may occasionally be involved in Whipple disease, caution is required in the interpretation of rectal biopsy specimens. PAS-positive macrophages may be seen in the rectal and colonic mucosa of healthy individuals. [84] Therefore, this finding, without demonstration of the bacilli, is not diagnostic of Whipple disease.

With the characterization of *T whippelii* by PCR and sequencing of the bacterial 16S ribosomal RNA gene in tissue from infected patients, [24], [40], [42], [43] and [44] a PCR-based diagnostic assay has become useful in confirming the diagnosis of Whipple disease and in monitoring the response during antibiotic treatment. A PCR assay for *T whippelii* was applied diagnostically to intestinal biopsy specimens in two retrospective studies that showed that PCR is a highly sensitive and specific test to confirm the diagnosis of Whipple disease. [26], [27] In the first study, *T whippelii* DNA was detected in all 30 patients with Whipple disease and in no controls. Despite negative findings of intestinal PCR studies after treatment, symptomatic cerebral Whipple disease appeared in three patients, suggesting that PCR assay may be less useful for monitoring the effects of treatment. [26] In the second study, none of the patients who had negative PCR findings after treatment experienced a clinical relapse, whereas most of the patients with positive PCR findings after treatment either never responded to treatment or experienced a clinical relapse during the follow-up period. [27] In contrast to the first study, [26] this study suggests that PCR may be useful to monitor therapy, with a negative PCR result predicting a low likelihood of clinical relapse. Additional studies have also demonstrated that PCR assay is a very useful diagnostic tool to confirm the diagnosis of Whipple disease and to monitor bacterial elimination during therapy. [85], [86] and [87] Furthermore, PCR assay has been shown to be a valuable aid in diagnosing extraintestinal Whipple disease and in differentiating Whipple disease from other diseases with similar histological features. [88], [89], [90] and [91] Although previous reports suggested that a PCR-based diagnosis of Whipple disease may be accomplished by the detection of *T whippelii* DNA in peripheral blood mononuclear cells or whole blood, [42], [43] this appears to be of limited diagnostic value. Multiple investigators have found no evidence of *T whippelii* 16S ribosomal DNA (rDNA) in peripheral blood monocytes in patients with active Whipple disease, suggesting that PCR of peripheral blood mononuclear cells is not a sensitive test for the diagnosis of Whipple disease and cannot be used as a substitute for the analysis of duodenal biopsy specimens. [85], [92]

Differential Diagnosis

Because the gastrointestinal symptoms in Whipple disease are nonspecific, other malabsorptive diseases with diffuse small intestinal involvement, such as celiac disease, may present in a similar manner. Infiltrative diseases of the small intestine, such as intra-abdominal lymphoma, can cause similar gastrointestinal symptoms and account for many of the x-ray findings associated with Whipple disease. These diseases can be easily and reliably differentiated by small intestinal mucosal biopsy.

Of particular importance, given the increasing incidence of acquired immunodeficiency syndrome (AIDS), Whipple disease must be distinguished from *Mycobacterium avium* complex (MAC) infection. Patients with AIDS are at risk for the development of intestinal infection with MAC, which can cause symptoms of diarrhea and weight loss. The histological lesion in MAC infection reveals infiltration of the lamina propria with PAS-positive macrophages, as in Whipple disease. [93], [94] These diseases can be differentiated by performing an acid-fast stain on the biopsy specimen (Fig. 75-4). MAC bacilli are acid-fast, whereas the Whipple bacillus is not. In addition, MAC organisms can be cultured and have an electron microscopic appearance quite different from that of Whipple bacilli. PAS-positive macrophages in the intestinal lamina propria can also be seen in systemic histoplasmosis and macroglobulinemia; however, each has histological features that should readily differentiate it from Whipple disease. In systemic histoplasmosis, large, PAS-positive, rounded, encapsulated *Histoplasma* organisms are easily seen in macrophages, and the faintly staining, homogeneously PAS-positive macrophages of macroglobulinemia should be readily recognized.

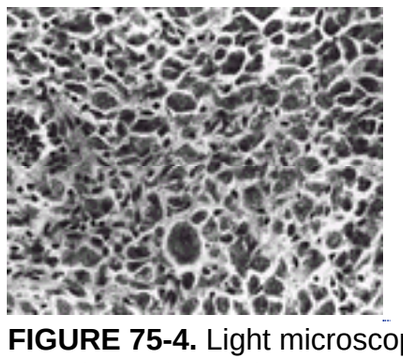

FIGURE 75-4. Light microscopic appearance of macrophages in the intestinal lamina propria of a patient with acquired immunodeficiency syndrome and *Mycobacterium avium* complex involvement of the gut. Note the easily identified bacilli within the macrophages. Acid-fast stain; original magnification ×500. (From ref. [15].)

Treatment and Prognosis

Whipple disease usually responds dramatically to antibiotic therapy. Numerous antibiotic regimens effective against gram-positive organisms and given for variable lengths of time have been used successfully, including penicillin, penicillin and streptomycin, erythromycin, ampicillin, chloramphenicol, tetracycline, and trimethoprim-sulfamethoxazole (TMP-SMX). [8], [9], [10], [11], [12], [13], [14], [15] and [16] Because Whipple disease is uncommon, no controlled trials have compared antibiotic regimens. Therefore, the best regimen and the optimal duration of treatment have not been established. Despite the initial response to antibiotic therapy, relapses are

common. Relapses can occur during treatment or months to years after completion of treatment. [10], [14], [15] and [16], [72] In a review of the long-term follow-up of 88 patients with Whipple disease who had initially responded successfully to antibiotic therapy, 31 patients eventually relapsed. [14] The mean time to relapse was 4.2 years. Of the 31 patients who relapsed, 21 had received initial therapy with tetracycline alone. Clinical relapses consisting of a recurrence of gastrointestinal symptoms and arthritis occurred both early and late and responded favorably to further antibiotic treatment, whereas CNS relapses tended to occur late and responded poorly to additional antibiotic therapy.

Given the concern about CNS relapses, [14], [64], [72], [75] it is reasonable to assume that all patients may have CNS involvement and to treat initially with an antibiotic that readily crosses the blood-brain barrier. One double-strength tablet of TMP-SMX (160 mg of TMP and 800 mg of SMX) given twice daily for 1 year is the best long-term option (Table 75-3). [14] TMP-SMX penetrates the blood-brain barrier and should effectively eradicate CNS involvement. Although shorter courses of antibiotic therapy may be effective, because of the concerns about CNS relapse and its poor response to additional therapy, it is appropriate to treat patients for 1 year. [14], [16], [95] Experts have suggested that initial therapy with parenteral penicillin G (1.2 million U/d) and streptomycin (1.0 g/d) for 10 to 14 days may be of additional benefit, [14], [16], [95] resulting in the lowest relapse rate. This should be considered in all patients with Whipple disease, particularly because compliance with the prescribed regimen is often limited (see Table 75-3). In patients who are allergic to or cannot tolerate TMP-SMX, parenteral penicillin and streptomycin for 10 to 14 days followed by oral penicillin VK or ampicillin for 1 year is reasonable. In these patients, if the development of CNS symptoms is a concern, chloramphenicol is an option. Oral chloramphenicol, like TMP-SMX, achieves relatively high CNS concentrations. In addition to antibiotic treatment, in patients with severe malabsorption and malnutrition, appropriate supplementation of specific nutrients, such as folic acid, vitamin B_{12}, fat-soluble vitamins, and iron, should be given to prevent deficiency.

	MEDICATION	DURATION
Treatment of Choice	Trimethoprim-sulfamethoxazole *or*	1 year
	Procaine penicillin G_1/ streptomycin (parenteral) *plus*	2 weeks
	Trimethoprim-sulfamethoxazole	1 year
Alternatives	Penicillin or ampicillin*	1 year
	Chloramphenicol†	1 year

*May be used in patients with sulfa allergy without central nervous system symptoms.

†May be used in patients with sulfa allergy with central nervous system symptoms.

TABLE 75-3 Treatment of Whipple Disease

At the conclusion of 1 year of antibiotic therapy, a small intestinal mucosal biopsy should be repeated to document the absence of residual bacilli before treatment is discontinued. Although PAS-positive macrophages may be present in the lamina propria for many years in patients treated for Whipple disease, the presence of bacilli on electron microscopy suggests inadequate treatment. PCR for *T whippelii* in intestinal biopsy specimens is not routinely recommended at this time; however, if available, it may provide useful information regarding the adequacy of therapy and the likelihood of relapse. PCR positivity suggests inadequate treatment, and PCR negativity predicts a low likelihood of clinical relapse but does not guarantee an absence of future CNS relapse. [26], [27]

The prognosis for patients with Whipple disease who receive appropriate effective antibiotic therapy is excellent. Gastrointestinal symptoms frequently resolve within 1 month. Extraintestinal symptoms such as fever and arthritis often disappear within a few days. Most patients are asymptomatic within a few months. After therapy has been discontinued, patients must be carefully observed for symptoms suggestive of relapse. They should be followed with clinical evaluation and laboratory studies. After endoscopy with intestinal biopsy at 1 year to confirm the response to treatment, routine invasive diagnostic studies, such as annual endoscopy with intestinal biopsy, are not warranted. Relapse may be signaled by a recurrence of gastrointestinal symptoms or by symptoms suggesting extraintestinal involvement. If relapse is suspected, small intestinal biopsy should be repeated and the specimen examined for the presence of free bacilli. The treatment of a relapse of Whipple disease is more complex but consists of a repetition of the initial therapy. Gastrointestinal relapse responds well to further antibiotic therapy. If the patient does not respond to TMP-SMX, then a trial of oral penicillin VK or tetracycline is reasonable. Patients with CNS relapse have a relatively poor prognosis. If the patient does not respond to a repeated course of the initial therapy, then a course of chloramphenicol is warranted. Combination therapy with antibiotics and interferon-? may be beneficial in patients with refractory Whipple disease. [52]

TROPICAL SPRUE

Tropical sprue, a syndrome occurring in individuals residing in certain tropical locations, is characterized by chronic diarrheal illness and malabsorption that gradually lead to the development of severe nutritional deficiencies. The typical clinical description of tropical sprue was first reported in 1759 in Barbados. [96] The cause is presumed to be bacterial overgrowth of the small intestine. The essential clinical features of the disease are chronic diarrhea and malabsorption, with resultant weight loss. Megaloblastic anemia secondary to folate and vitamin B_{12} deficiency is usually present. When clinically suspected, the diagnosis is made by small intestinal biopsy and confirmed by prompt clinical and laboratory response to treatment with folic acid and broad-spectrum antibiotics.

Epidemiology

Tropical sprue is endemic in many tropical locales. The disease is common in India, is relatively frequent among the indigenous populations of Cuba, the Dominican Republic, Haiti, and Puerto Rico, and may be seen in northern South America, Venezuela, and Colombia. Tropical sprue has also been described in a number of other countries, including the Philippines. Although the disease has been described in all age groups, it is primarily a disease of adults, occurring only rarely in children. Tropical sprue seems to occur mostly as sporadic isolated cases, but epidemics have been described among British forces serving in Burma during World War II, American military personnel serving in the Philippines, and villagers living in southern India. [97], [98] and [99] Several reports have indicated that in both sporadic cases and epidemics, a peak seasonal occurrence of tropical sprue is observed that is unrelated to the rainy season. [100]

Although tropical sprue can afflict visitors within weeks or months after their arrival in a region of endemicity, it rarely occurs in short-term travelers, usually developing in persons who have lived in a region for more than 6 months to 1 year. [101] The prevalence of tropical sprue among residents of developed countries visiting areas of endemicity for long periods of time appears to be decreasing, possibly related, in part, to the frequent use of self-medication with antibiotics for episodes of acute diarrheal disease. Tropical sprue also appears to occur more often among native and nonnative residents and those who live where sanitary conditions are poor. [102] In rare instances, tropical sprue does not become clinically apparent among native or nonnative residents until months or even years after they have moved to a temperate climate. [103]

Etiology and Pathogenesis

Tropical sprue is presumed to be an infectious disease that results from chronic bacterial overgrowth of the small intestine by toxigenic strains of coliform bacteria, but no unique organism has been identified. The epidemiology of the disease, with documented epidemics and seasonal variation, supports the possibility of an underlying infectious etiology. In addition, the disease often occurs after an acute diarrheal illness and usually responds to antibiotic treatment, lending further support for an infectious etiology for tropical sprue.

Although extensive studies have failed to identify a single specific infectious agent as the causative factor, most individuals with tropical sprue, including both native and nonnative residents, exhibit bacterial overgrowth of the jejunum with strains of *Klebsiella pneumoniae* or, less often, *Enterobacter cloacae* or *Escherichia coli*. [104], [105] and [106] Eradication of these bacteria with antimicrobial therapy results in cure of the intestinal abnormalities. [104], [106] In addition, among foreigners who have acquired tropical sprue after their return to a temperate climate, the bacterial overgrowth persists until they receive antibiotic treatment. [106]

The organisms that overgrow the small bowel of patients with tropical sprue are facultative anaerobes, typically consisting of only one or a few species, serotypes, or biotypes in each individual patient. [105] There are no anaerobes as classically seen in diseases manifested by small bowel stasis and resultant bacterial overgrowth, such as blind loop syndrome. These coliform bacteria have pathogenic properties that are not shared by similar isolates obtained from the small bowel of patients with blind loop syndrome. [107] The organisms isolated from patients with tropical sprue were found to have a secretory effect and to induce structural damage, presumably

by toxin production, in the small intestine of rabbits and rats. [107], [108] However, these studies were performed before current techniques and assays, such as DNA probes and ELISA, were available to identify and characterize any of these presumed toxins precisely.

In most nonnative residents, tropical sprue develops after an attack of acute watery diarrhea. Acute infectious diarrhea, caused by a variety of enteric pathogens, may be associated with transient jejunal contamination by coliform bacteria. [109] This event may be the precipitating factor in tropical sprue. Subsequently, the coliform bacteria are able to colonize the small intestine permanently. Eventually, prolonged exposure of the small bowel intestinal mucosa to the organisms and their toxins precipitates a series of events leading to progressively worsening abnormalities of intestinal structure and function.

The factors responsible for the persistent bacterial colonization in tropical sprue are unknown. Some evidence suggests that these coliforms may be unusually adherent to the intestinal mucosa [110]; however, light and electron microscopic studies of jejunal biopsy specimens obtained from patients with untreated tropical sprue have failed to show bacteria adherent to the intestinal lumenal surface. [111] No evidence has been found of intestinal immunologic dysfunction that might predispose to abnormal bacterial colonization in tropical sprue. [112], [113] and [114] It has been postulated that dietary fat, specifically long-chain fatty acids such as linoleic acid, may play a role in the development of tropical sprue. [100], [115], [116] In addition to altering intestinal motility and delaying intestinal transit time, long-chain fatty acids can alter intestinal physiology and contribute to the production of tropical sprue by a number of other mechanisms, [115], [117], [118] and [119] but it remains to be determined whether this is clinically relevant.

The unique role of folate deficiency in the pathogenesis of tropical sprue is not completely understood, but it is likely that whatever the initial insult to the small intestine may be in tropical sprue, the resulting folate malabsorption and deficiency contribute to ongoing pathogenesis. Folate deficiency leads to a decreased number of intestinal epithelial cells and atrophy of villi and is associated with a number of intestinal structural and functional alterations, including crypt hypertrophy, blunting of villi, and less efficient absorption of water, electrolytes, and carbohydrates. Treatment with pharmacological doses of folic acid results in structural and functional improvement in the small bowel of patients with tropical sprue. [101]

Although the available data suggest that tropical sprue is a syndrome of postinfectious malabsorption caused by chronic bacterial overgrowth of the small intestine with toxigenic coliform bacteria, it is important to emphasize that this is perhaps an oversimplification, and a number of questions remain. For example, coliform bacteria have been isolated from the proximal small intestine in healthy asymptomatic controls in tropical locations such as India and South Africa. [120], [121] Furthermore, these bacteria have not been found in all patients with tropical sprue. In addition, a single causative infectious agent has not been identified. The reasons for the rarity of this disease among children and its scattered geographic distribution are unclear.

Histopathology

The classical histopathological finding in patients with tropical sprue is partial atrophy of the villi in the small intestine. A completely flat mucosa, as is seen in untreated celiac sprue, is found in fewer than 10% of patients with tropical sprue. The histological changes consist of lengthening of the crypt area, broadening and shortening of the villi, chronic infiltration of the lamina propria by inflammatory cells, and variable degrees of abnormality of the surface epithelium (Fig. 75-5). [111], [122] The basement membrane usually appears thickened, staining as collagen on light microscopy. [122] Electron microscopy shows that the thickening represents an accumulation of dense material, of unknown composition, subjacent to a normal basal lamella. [111] Structural abnormalities are more prominent in the jejunum during the early phase of tropical sprue; however, eventually the entire small bowel is equally affected. These histological changes are nonspecific and may also be seen in a number of other clinical entities, including severe folate deficiency, bacterial overgrowth, and human immunodeficiency virus (HIV) enteropathy. Functional changes in the small intestine in tropical sprue reflect the morphologic changes and are most prominent in the jejunum and ileum, resulting in malabsorption and the associated clinical features.

FIGURE 75-5. A: Light microscopic appearance of the moderate villus abnormality of tropical sprue acquired by a North American during a 2-week visit to the Philippines. Note the broadened villi, infiltration of the lamina propria by plasma cells and lymphocytes, and lymphocytic infiltration of the epithelium. Periodic acid-Schiff and hematoxylin stain; original magnification ×80. **B:** Enlargement of a portion of **A** reveals the epithelial changes. Original magnification ×132.

Clinical Manifestations

Many individuals can identify the onset of their disease as an episode of acute diarrhea. After about a week, the symptoms become milder and chronic. Less commonly, the onset of tropical sprue is insidious. The chronic symptoms, including prolonged diarrhea, abdominal cramping, nausea, and anorexia with associated weight loss, are nonspecific and related to malabsorption. Fever may be present at the onset of the disease but rarely persists. Lactose intolerance often develops early in the course of tropical sprue, caused by a reduction in brush border disaccharidase enzyme activity. Within a few months, malabsorption results in folate deficiency. After a variable period of time, usually about 6 months but sometimes earlier in poorly nourished native residents in whom the disorder develops, severe depletion of folate coupled with the development of vitamin B_{12} deficiency causes symptoms associated with megaloblastic anemia, such as fatigue and weakness. Later in the course of the disease, peripheral neuropathy and confusion may occur. If the anemia is severe enough, symptoms of high-output heart failure may appear. These deficiencies and their manifestations can develop in about 10% of patients with tropical sprue who have never experienced any gastrointestinal symptoms.

The findings on physical examination are manifestations of nutritional deficiencies and therefore typically appear late. These include pallor, glossitis, and edema. [123], [124] Stomatitis, hyperpigmentation, and neurological manifestations of vitamin B_{12} deficiency may occur but are rare.

Laboratory Findings Megaloblastic anemia, caused by combined deficiencies of folate and vitamin B_{12}, is the hallmark of tropical sprue. Steatorrhea and reduced xylose absorption are frequently present. Malabsorption of fat-soluble vitamins often occurs, resulting in low serum concentrations of vitamin A (and ß-carotene), 25-hydroxyvitamin D, and calcium. Marked elevation of the prothrombin time is unusual. Hypoalbuminemia and hypocholesterolemia are also seen.
Radiologic and Endoscopic Findings A small bowel series may reveal findings suggestive of tropical sprue. These include prominent, thickened mucosal folds with a coarsened or irregular contour involving the jejunum and ileum. Lumenal dilation and flocculation of the barium may also be seen. [125] Although these findings are nonspecific, ileal involvement is characteristic of tropical sprue and relatively uncommon in celiac sprue, Whipple disease, and other small bowel enteropathies. Upper gastrointestinal endoscopy may reveal scalloping of the valvulae conniventes and a mosaic pattern of the mucosa, [126] findings frequently described in celiac sprue.

Diagnostic Evaluation

The clinical presentation of tropical sprue is nonspecific. The diagnosis should be considered in patients who present with chronic diarrhea, weight loss, and evidence of malabsorption. A history of recent travel to, residence in, or immigration from a tropical location is essential. Depending on the duration and severity of the disease, important physical findings may be detected that are related to anemia and nutritional deficiencies, and laboratory abnormalities, such as megaloblastic anemia caused by folate and vitamin B_{12} deficiency, can be important clinical clues. Stool culture is of no diagnostic value in tropical sprue, but the stool should be examined for ova and parasites to rule out other infectious causes of chronic diarrhea, such as *Giardia*.

When the diagnosis of tropical sprue is suspected, endoscopy with small bowel biopsy should be performed. The demonstration of characteristic changes in the architecture of the villi, including partial atrophy, supports the diagnosis. The diagnosis of tropical sprue is confirmed by a prompt clinical and laboratory response to the institution of appropriate treatment.

Differential Diagnosis

The differential diagnosis includes a number of other malabsorptive diseases and enteropathies. In temperate climates, the presentation of celiac sprue be similar to that of tropical sprue. In celiac sprue, the ileum tends to be minimally involved, and vitamin B_{12} deficiency is uncommon. In addition, more severe histological abnormalities are seen in celiac sprue, with flattening of the villi, and the clinical response to a gluten-free diet is dramatic. As in celiac sprue, significant involvement of the ileum and vitamin B_{12} deficiency is uncommon in Whipple disease. Whipple disease can easily be distinguished histologically by the presence of PAS-positive macrophages and the detection of organisms on electron microscopy. HIV enteropathy is manifested by chronic watery diarrhea in the absence of an identifiable infectious agent, and the intestinal histology may be very similar to that of tropical sprue. Therefore, it is important that a thorough history be taken, including questions relating to possible HIV exposure; for individuals at risk for AIDS, the appropriate diagnostic tests are indicated. Infection of the small intestine with parasites such as *Cryptosporidium, Giardia,* and *Strongyloides* can cause chronic diarrhea and malabsorption. However, the malabsorption is usually less severe, without significant malnutrition and weight loss. The diagnosis can often be made by stool examination for ova and parasites. Bacterial overgrowth caused by intestinal stasis may be seen in diseases that alter intestinal motility, such as amyloidosis, pseudoobstruction, scleroderma, and blind loop syndrome, resulting in chronic diarrhea. In addition to the presence of these underlying diseases, bacterial overgrowth can be distinguished from tropical sprue by the presence of anaerobes rather than coliforms in the small intestine and by the mild nonspecific findings on the small bowel biopsy specimen. Primary intestinal lymphoma may present with chronic diarrhea, often with significant associated abdominal pain. Intestinal TB may occur in some tropical countries but it is much less common than tropical sprue.

Finally, tropical sprue occurs in areas such as Haiti, where the frequency of an entity usually referred to as *tropical enteropathy* is high. [127] Tropical enteropathy is generally associated with milder abnormalities of intestinal structure and function, and clinically significant malabsorption or nutritional deficiencies do not develop in affected persons. Unlike the abnormalities of tropical sprue, the intestinal abnormalities of tropical enteropathy resolve within a year after return to a temperate climate, even without specific therapy. [128] It is thought that the intestinal abnormalities of tropical enteropathy are a manifestation of repeated, transient episodes of intestinal infection caused by various enteric pathogens.

Treatment and Prognosis

Although spontaneous remissions occurred rarely, tropical sprue was usually a fatal disease before the 1930s. This changed after treatment with liver extract was introduced in 1931, followed a decade later by folic acid treatment and subsequently by antimicrobial therapy in the 1960s.

Treatment with pharmacological doses of folic acid alone promptly relieves the megaloblastic anemia, and a return of appetite is accompanied by weight gain in the ensuing weeks. As intestinal morphologic abnormalities regress, gastrointestinal symptoms become less severe. Although this therapy alone may be curative for those in the early stage of the disease, treatment for up to 2 years may be required if intestinal structure and function are to return completely to normal. [101] In chronic disease, folic acid relieves the intestinal abnormalities and gastrointestinal symptoms; however, it does not completely reverse the intestinal abnormalities, and chronic symptoms often persist. [129]

Treatment with antimicrobials eradicates the contaminating coliform bacteria. [104], [106] Tetracycline and nonabsorbable sulfonamides are equally effective. [130], [131] Given alone, antimicrobial therapy results in a reduction of the gastrointestinal symptoms within weeks along with a slow, progressive regression of the intestinal abnormalities. Improved folate absorption results in a hematologic response and a return of appetite within several weeks.

Combination therapy with tetracycline and folate is the most logical approach to the treatment of tropical sprue. This consists of 250 mg of tetracycline four times a day and 5 mg of folic acid once daily. Although 1 month of therapy has been effective in travelers with tropical sprue, treatment for up to 6 months is often required for those with chronic disease to achieve complete remission of the malabsorptive syndrome and to prevent recurrence. [130], [131] Although not necessary for the acute phase of therapy, vitamin B_{12} (1000 µg), administered parenterally once a week for several weeks, should also be given to replete tissue stores. Despite treatment with combination therapy for 6 months, intestinal abnormalities may persist, and clinical relapse, requiring additional courses of therapy, may occur in 10% to 20% of patients who remain in tropical locations of endemicity. [132]

TUBERCULOSIS

Mycobacterium tuberculosis, the primary cause of tuberculosis (TB), infects a third of the world's population and causes more deaths per year than any other infectious agent. [133] It is found primarily in developing countries of the world, where overcrowding and poor sanitation contribute to its spread. Twelve countries in the world account for nearly three fourths of all cases. In the United States, the number of reported TB cases reached an all time low of 7.4 per 100,000 persons in 1997, and 39% of these were among foreign-born persons. Although pulmonary manifestations predominate in most cases of TB, gastrointestinal involvement can be part of a disease process affecting multiple organs; less commonly, gastrointestinal involvement in the absence of other identifiable sites of infection (so-called primary gastrointestinal TB) may be associated with symptoms and even be the primary cause of death. [134], [135] and [136] This diagnosis is rarely considered before death, and as a result the opportunity for effective therapy is often missed.

Epidemiology

Before the development of effective anti-TB chemotherapy, gastrointestinal TB was seen in about 70% of persons with advanced pulmonary disease. [137] Since the advent of effective chemotherapy, gastrointestinal TB has become much rarer, and in the United States and Canada, it is reported in fewer than 1% of all cases. [138], [139] In developed countries, it is most commonly found in association with immunosuppression, especially in patients with AIDS. [140] In developing countries, AIDS and immunosuppression, in combination with limited access to anti-TB therapy and inadequate sanitation, appear to have increased the incidence of gastrointestinal TB. In one series from South Africa, 46% of patients with smear-positive, cavitating pulmonary TB had gastrointestinal involvement. [141] In India, TB remains the most common granulomatous disease of the intestinal tract. [142]

Some data suggests that the risk for gastrointestinal disease is directly related to the severity of pulmonary disease, although not all persons with gastrointestinal TB have evidence of current or past pulmonary disease. [141], [142] Rare cases of congenital necrotizing enterocolitis have been reported in infants born to mothers with miliary TB. [143]

Etiology and Pathogenesis

Gastrointestinal TB is usually caused by *M tuberculosis,* but it may also be caused rarely by a closely related species of bacteria, *Mycobacterium bovis*. [133] In the United States, *M bovis,* an organism that can cause TB in cattle and in humans, is only of historical interest because the pasteurization of milk and the screening of cattle for infection have eliminated it as a cause of TB. It still may be rarely seen in other parts of the world, however, where these public health measures are not practiced. *M tuberculosis* is an acid-fast, curved bacillus that can infect certain primates, including humans, but humans are the only known reservoir. It spreads from human to human by aerosolization. It is thought that gastrointestinal disease arises when organisms are swallowed and survive destruction by gastric acid; the source of the organisms can be either dietary, such as infected milk in the rare cases of *M bovis* infection, or more commonly pulmonary secretions. Lesions may also occur as a result of disseminated miliary disease.

Infection of the upper gastrointestinal tract is very uncommon, and lesions are more often found in areas characterized by high lymphoid density and stasis, such as the distal small bowel, cecum, appendix, and, less commonly, colon and rectosigmoid. [144], [145] Gastrointestinal TB is thought to arise by the same pathophysiological sequence as pulmonary infection; initial infection of macrophages is followed by multiplication of the organisms in macrophages, caseation necrosis, and a secondary host immune response. The immune response is characterized by either cell-mediated immunity, with a marked influx of activated myeloid and lymphoid cells, or a delayed hypersensitivity reaction accompanied by cell death and caseation necrosis. [146] The defects of cell-mediated immunity seen in patients with AIDS and other immunocompromised hosts result in failure to contain the organism, so that these patients are predisposed to the development of clinical disease once infected.

Pathology

The pathological lesions of gastrointestinal TB have classically been divided into an acute ulcerative stage and a chronic hypertrophic stage of disease that is characterized by granulomata and extensive fibrosis. The ulcerations presumably develop as a result of infection of gastrointestinal macrophages, with subsequent caseation necrosis and intense inflammatory changes. The lesions are most often located in the distal small bowel and cecum. Multiple focal areas of involvement are typical. The ulcers may be deep; typically, they are circumferential, in contrast to the linear ulcerations of Crohn's disease. The ulcers are typically irregular with a necrotic base, and pseudopolyps may be found. Perforation may occur. Fistulae and anorectal lesions may form. Histologically, the ulcerative lesions are characterized by granulomata, usually with caseation and larger numbers of acid-fast organisms than in the later, fibrotic stages. Involvement of draining lymph nodes is common. In HIV-infected patients, inflammatory and ulcerating lesions are typical, with perforation reported. With more chronic inflammation, the bowel wall may become markedly fibrotic and stenotic, with the formation of mass lesions (tuberculomas). Strictures are typically short and may be multiple. The numbers of organisms may be markedly diminished in the fibrotic stages or absent in patients who have received treatment.

Clinical Manifestations

The symptoms of gastrointestinal TB are nonspecific, and in the absence of pulmonary TB, the diagnosis may be difficult. The most common symptom of gastrointestinal TB is abdominal pain. [142], [144], [147] Others include fever, anorexia, diarrhea, weight loss, constipation, bloating, and hemorrhage. [135], [137], [142], [144], [147] If disease is limited to the duodenum, the symptoms may be primarily those of peptic ulcer disease. [148] The physical findings may include cachexia and, in patients with an acute pulmonary process, pulmonary manifestations. [149], [150] and [151] The abdominal examination may reveal a palpable mass, which in various series is the abnormality most commonly found. [142], [144], [147] Other physical examination findings include evidence of complications such as perforation, obstruction, fistulae, massive hemorrhage, and malabsorption. [134], [145], [147], [148]

Diagnostic Evaluation

Barium contrast studies may demonstrate ulcerations, strictures, fistulae, and thickening, deformity, and incompetence of the ileocecal valve; evidence of mass lesions similar to those of Crohn's disease may be seen (Fig. 75-6). [152], [153] CT and ultrasonography both show mural thickening of the bowel at sites that correlate with the pathological findings at colonoscopy. [154] Mesenteric lymphadenitis, omental thickening, and ascites are frequently associated with tuberculous enterocolitis. [154] Peritonitis is found in about 20% of patients. [155] Lesions may also be found in liver, spleen, or pancreas. CT-guided or ultrasonographically guided fine-needle aspiration has been used successfully for diagnosis. [156]

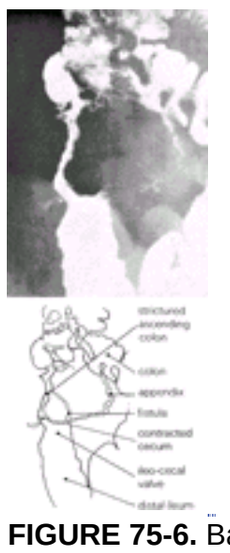

FIGURE 75-6. Barium study in a patient with hypertrophic tuberculosis shows a dilated distal ileum leading into a contracted cecum and strictured ascending colon. (From Misiewicz JJ, Forbes A, Price A, et al. Atlas of clinical gastroenterology, 2nd ed. London: Wolfe, 1994:5.4.)

Because tissue is required for histological confirmation and culture, and because the most common site of involvement is the ileocecal region, the diagnostic procedure of choice is usually colonoscopy with biopsy. [142], [143], [149] Upper gastrointestinal endoscopy with jejunal or duodenal biopsy is indicated less commonly, when disease at these sites is suspected. [135], [148] Visual inspection of the involved bowel most commonly reveals multiple areas of nodular mucosa with areas of ulceration. [144], [147]

Biopsy material, obtained either endoscopically or surgically, must be submitted for histological examination and the detection of mycobacteria. Histological studies showing caseating granulomata or Ziehl-Neelsen or Kinyoun stain showing acid-fast, beaded, slightly bent rods may provide immediate answers, but often the results of these tests are negative. In one series, the yield of acid-fast bacilli from colonic biopsy was zero. [147] In addition, typical caseating granulomata are found less frequently in biopsy material from lymph nodes than from the intestine and may be found in only half of specimens. [147] PCR has the potential to provide a rapid diagnosis with enhanced sensitivity and specificity, but studies of biopsy material from the gastrointestinal tract have shown sensitivities of only 40% to 75%. [142], [157] Culture remains the gold standard but requires 1 to 2 weeks for results with the Bactec radiometric system (Bactec Corporation, Houston, Texas) and 4 to 6 weeks when standard detection systems are used. However, even the results of culture are often negative, and a presumptive diagnosis is based on endoscopic findings in combination with a response to anti-TB therapy. [142], [147]

Differential Diagnosis

TB should always be considered in the differential diagnosis of patients with obscure abdominal symptoms or physical findings, especially those who are immunocompromised. Other entities that may present clinically and pathologically similarly to TB include Crohn's disease, lymphoma, carcinoma, diverticular disease, appendicitis, and other infections of the gastrointestinal tract. [137], [151], [154] The latter include histoplasmosis and, in AIDS patients, MAC enteritis and, less closely, cryptosporidiosis (Table 75-4). Cytomegalovirus infection is another diagnostic consideration when the colon is involved. *Yersinia* enteritis may also present with similar clinical manifestations and pathology in the terminal ileum, but its shorter course usually distinguishes it from TB. Despite the extensive tuberculous dissemination frequently associated with TB enteritis, the diagnosis is often delayed, and TB enteritis is most frequently discovered unexpectedly at exploratory laparotomy or autopsy. [134], [140]

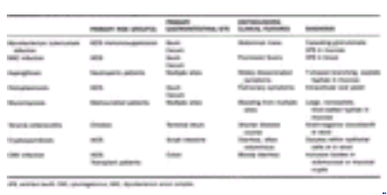

TABLE 75-4 Infectious Diseases Mimicking Tuberculous Enteritis

It seems paradoxical that Crohn's disease was recognized in the early part of the last century, when it was distinguished from gastrointestinal TB. The distinction remains a problem in countries in which TB is endemic. However, in most developed countries, Crohn's disease is now much more common, and intestinal TB is rare. The clinical challenge is to recognize TB in a patient thought to have Crohn's disease. Helpful distinguishing features are that Crohn's disease more commonly begins

earlier in life, ulcerations are more typically linear, and granulomata, when present, do not contain caseation necrosis or organisms. Severe active Crohn's disease is usually not found in patients with advanced HIV infection.

Treatment and Prognosis

The prognosis of a patient with gastrointestinal TB depends primarily on the immune status and the institution of the appropriate therapy in a timely manner. In untreated HIV-infected patients, the disease progresses rapidly and is almost always fatal. [140] However, the results of therapy are usually satisfactory, even in immunocompromised hosts, when it is started sufficiently early in the course of disease. The difficulty lies in making a timely diagnosis, and in many series, significant numbers of patients die before the disease is diagnosed. [136], [141]

If any indication of TB is noted, such as a compatible clinical picture in a high-risk patient, a positive acid-fast stain, or histological evidence of granulomata, therapy should be started immediately pending the results of culture and susceptibility testing. If the patient is immunocompetent and lives in a community where the rate of resistance is known to be less than 4%, the regimen of choice is 300 mg of isoniazid, 600 mg of rifampin, and 1.5 to 2.5 mg of pyrazinamide. [158] If the person is immunocompromised or lives in an area with high rates of drug resistance, then either 15 mg of ethambutol per kilogram or 15 mg of streptomycin per kilogram once daily should be added to the previously mentioned regimens. If the patient does not show evidence of malabsorption, we prefer ethambutol to streptomycin because it can be administered orally instead of by intramuscular injection.

If susceptibility testing reveals that the organism is sensitive to all drugs, then ethambutol or streptomycin can be stopped immediately and pyrazinamide continued for only 2 months. Isoniazid and rifampin must be continued for an additional 4 months, for a total of 6 months of therapy. In a randomized controlled trial, 6 months of therapy for gastrointestinal TB was equivalent to 12 months, the previous standard duration. [159] Alternative regimens for patients who cannot tolerate all three drugs include isoniazid and rifampin for 9 months and isoniazid and ethambutol for 18 months. [160]

If the organism is resistant to isoniazid (the most common form of resistance), then treatment can be with rifampin, pyrazinamide, and either ethambutol and streptomycin for 6 months or rifampin and ethambutol for 12 months. [160] In cases with a more complicated resistance pattern, alternative drugs are required for at least 1 year after cultures become negative.

Surgical intervention, although it was frequently used in the past for diagnosis, is usually not necessary and is reserved for complications such as obstruction, perforation, fistula, or a mass that does not resolve with medical therapy. [136], [145], [149] In most cases, a trial of medical therapy should be undertaken before surgical intervention. Complications can be fatal, even with surgical intervention, and may occur after the initiation of anti-TB medications. [134], [135] and [136], [161]

MYCOTIC INFECTIONS

Mycotic infections are most often a result of dissemination from a pulmonary source and usually occur in immunocompromised hosts, although they may also occur in normal hosts. The etiologic agents of aspergillosis, candidiasis, histoplasmosis, cryptococcosis, and mucormycosis have all been cultured from the small intestine. Although *Cryptococcus* has been cultured from the small intestine of patients with AIDS, it has not been associated with clinical disease and is not discussed further here. [162]

Histoplasmosis

Epidemiology Histoplasmosis is a very common infection in the midwestern and south central regions of the United States, where 80% of the inhabitants are infected. [163] Infection, however, rarely equates with disease. When symptoms do occur, they most commonly involve the lungs, the primary portal of entry. Gastrointestinal disease occurs usually, but not always, as a result of widespread dissemination in an immunocompromised host. Infants and children, the elderly, patients on immunosuppressive therapy, including steroids, and persons with immunosuppressive diseases (especially AIDS) are all at higher risk than the general population.
Etiology and Pathology Histoplasmosis is an infection caused by *Histoplasma capsulatum,* a fungus that exists in the mycelial form at ambient temperature (30°C) and in the yeast form at body temperature (37°C). [163] This dimorphic fungus produces both macroconidia and microconidia in clinical isolates. Visual inspection of the involved bowel may reveal entirely normal tissue or a variety of lesions. The most common lesions are single or multiple ulcers and tissue necrosis. [164], [165] Other lesions include flat elevations and polypoid protrusions. Microscopic examination of pathological specimens, after staining with methenamine silver, PAS, or Wright-Giemsa stain, reveals tiny (2–3 μm in diameter) oval yeast forms inside macrophages (Fig. 75-7). [163] Pathological specimens may also show granulomatous inflammation if the patient is not severely immunocompromised. [163] Culturing for the organism should be performed but is difficult, and up to 6 weeks may be required for a positive result. [163]

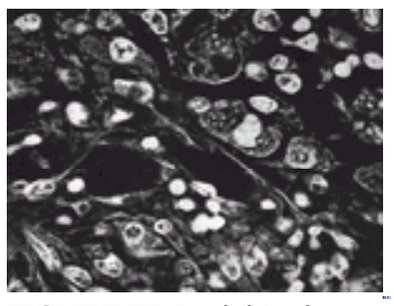

FIGURE 75-7. Light microscopic appearance of oval yeast forms of *Histoplasma capsulatum* within macrophages from a patient with disseminated histoplasmosis. High-power magnification. (From Shull HJ. Human histoplasmosis: a disease with protean manifestations, often with digestive system involvement. Gastroenterology 1953;25:389.)

Clinical Manifestations Although involvement of the small and large bowel is quite common in disseminated disease, symptomatic disease is much less common. In one series, only 50% of persons with gastrointestinal ulcerations had diarrhea or abdominal pain. [165] In addition, persons with disseminated disease who have little or no gastrointestinal pathology may have diarrhea or abdominal pain. Furthermore, even when gastrointestinal symptoms are present, they are often overshadowed by systemic or pulmonary complaints. [166] Occasionally, however, gastrointestinal complaints predominate. [166] The symptoms most commonly associated with small intestinal histoplasmosis include diarrhea, weight loss, fever, abdominal pain, nausea, vomiting, lethargy, and fatigue. [164] The stools are typically liquid or semiliquid and on occasion are bloody, especially if the colon is also involved. Weight loss of up to 30 lb has been reported. The abdominal pain is usually cramping and may be diffuse or localized, depending on the underlying pathology. Other reported presentations include edema of the legs and eyelids caused by protein-losing enteropathy [164] or malabsorption with steatorrhea. [166], [167] The most frequent complication and often a cause of death is perforation of the small bowel. [164], [168] In one series, this was the terminal event in 11% of persons with disseminated histoplasmosis. [164] Obstruction by ileocecal masses or enlarged retroperitoneal lymph nodes can also occur, as well as intussusception in association with pseudopolypoid or polypoid lesions. [164], [165]
Diagnostic Evaluation Involvement of the gastrointestinal tract should be suspected in the setting of blood cultures positive for *H capsulatum,* a fourfold rise in titer or a titer equal to 1:32 of *H capsulatum* serum complement fixation tests or higher, or a positive result of testing for *H capsulatum* in the blood or urine. For a definitive diagnosis of gastrointestinal involvement, upper gastrointestinal endoscopy with duodenal or jejunal biopsy or colonoscopy with terminal ileum biopsy is necessary. The choice of biopsy site should be guided by the symptoms and the knowledge that the ileocecal region is most commonly involved. [164], [165] If abdominal surgery is necessary, the specimens should be cultured to ascertain a diagnosis.
Differential Diagnosis Elongated constricting lesions, abnormally thickened loops of bowel, and lymphadenopathy can all be found on upper gastrointestinal contrast studies and abdominal CT and can mimic Crohn's disease of the terminal ileum, mycobacterial enteritis, other mycotic infections, and carcinoma. [164], [166]
Treatment and Prognosis The natural course of small intestinal histoplasmosis is difficult to determine because in many cases the disease is not diagnosed before death. Cases of disseminated disease, in which small intestinal involvement is frequent, have gone on for more than 10 years without specific therapy. [164] However, this situation is less likely today because of the increased association of the disease with immunosuppression—in particular, AIDS. Immunosuppressed persons often have more severe disease that progresses relatively rapidly, and death is inevitable without treatment. In the normal host with mild or moderate disease, the agent of choice is itraconazole; 200 to 400 mg is given per day, depending on the severity of illness. [169] Therapy should be continued for 6 to 18 months. If itraconazole cannot be taken, 400 mg of fluconazole per day is an alternative, but fluconazole is not as active against *H capsulatum.* Amphotericin B is usually reserved for patients who

have life-threatening disease or are severely immunocompromised. Adults with AIDS and those with life-threatening disease should be given 0.7 to 0.8 mg of amphotericin B deoxycholate per kilogram per day intravenously until clinical improvement occurs, usually after 1 to 2 weeks, and then they can be switched to itraconazole for the remaining course of therapy. Although only a few case reports of the successful use of lipid formulations of amphotericin to treat histoplasmosis have been published, [170], [171] experience in other disseminated fungal infections suggests that lipid formulations of amphotericin, given in higher doses than amphotericin deoxycholate, can be used for patients who have renal impairment or who fail to respond to amphotericin deoxycholate. [172] AIDS patients require maintenance therapy for life with 200 mg of itraconazole per day. [169]

Aspergillosis

Epidemiology Unlike histoplasmosis, which can develop in immunocompetent hosts, aspergillosis affects only severely immunocompromised patients. AIDS, organ transplantation, immunosuppressive chemotherapy, and the improved survival of premature infants have all contributed to an increase in the prevalence of aspergillosis. [173] In addition, the development of new oral antifungal agents, which have little or no activity against *Aspergillus* and are often used for prophylaxis and treatment in transplant, oncology, and AIDS patients, has resulted in the more frequent isolation of *Aspergillus*. [173] The small intestine is involved in about 5% of patients with disseminated aspergillosis. [173], [174]
Etiology and Pathology Aspergillosis is an infection caused by *Aspergillus,* a mold that grows by branching and longitudinal extension of wide (2- to 5-μm), Y-shaped, branching, septate hyphae. [175] The most common clinical isolate is *Aspergillus fumigatus,* followed by *Aspergillus flavus*. Clinical disease is produced by vascular invasion and necrosis. Lesions seen in pathological specimens or during endoscopy include linear ulcers, hemorrhagic infarcts, and necrotic mucous plaques. [173], [174], [176]
Clinical Manifestations The data on the clinical manifestations of aspergillosis of the gastrointestinal tract are sparse because much of the information has been derived from autopsy series. The most commonly reported symptom is gastrointestinal bleeding. [173], [174] Small intestinal perforation may also occur. [177]
Diagnostic Evaluation The diagnosis is especially difficult because *Aspergillus* is not easily cultured in the laboratory, and even when it is cultured, the culture may signify only contamination or colonization. [173], [175] Biopsy with histological evidence of tissue invasion is essential to verify that *Aspergillus* is the cause of the disease process (Fig. 75-8). *Candida* may also be seen on histological specimens and may contribute to the disease process (Fig. 75-9). [174], [176]

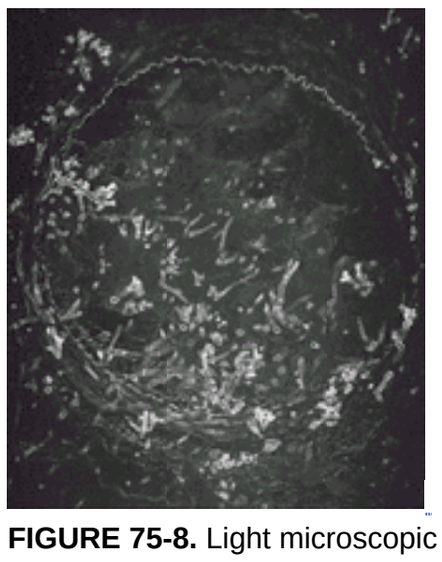

FIGURE 75-8. Light microscopic appearance of *Aspergillus* organisms showing vascular invasion. (From ref. [176].)

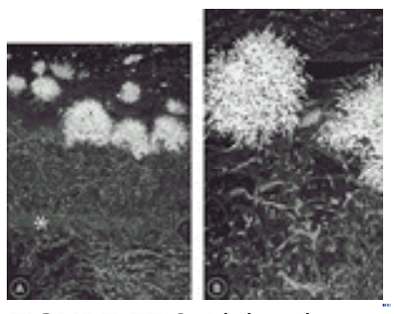

FIGURE 75-9. Light microscopic appearance of *Candida* organisms on the surface of infarcted bowel mucosa with *Aspergillus* hyphae in the underlying mucosa and submucosa. **A:** Low magnification. **B:** High magnification. Grocott-Gomori methenamine-silver stain. (From ref. [176].)

Differential Diagnosis The differential diagnosis includes mycobacterial enteritis, bacterial typhlitis, ischemic bowel, and other mycotic infections.
Treatment and Prognosis Disseminated aspergillosis, regardless of whether or not small intestinal involvement is present, carries a uniformly poor prognosis unless the underlying immunosuppression can be reversed. [175] It is critical for successful treatment that antibiotic therapy be started early in the course of disease. The recommended treatment for small intestinal aspergillosis is at least 1 mg of amphotericin B per kilogram per day for 2 weeks, continued until a clinical response occurs. If the patient is temporarily neutropenic, treatment should be continued until the marrow recovers. [175] Lipid formulations of amphotericin, at higher doses, can be used for patients who have renal failure or in whom renal failure develops during therapy or who do not respond to amphotericin deoxycholate. Salvage therapy for intolerance or failure to amphotericin B has been reported with itraconazole.

Candidiasis

Epidemiology *Candida* species are the fourth most common organisms isolated from the blood of hospitalized patients and are normal colonizers of the gastrointestinal tract. [178] The organisms are commonly associated with esophageal disease in immunocompromised patients but rarely cause disease of the small intestine. Persons at highest risk include those with AIDS, chronic mucocutaneous candidiasis, or malignancies, and those taking immunosuppressive agents.
Etiology and Pathology *Candida* is a yeastlike fungus that reproduces by budding small (4- to 6-μm), thin-walled, ovoid cells. Yeast forms, pseudohyphae, and hyphae can all be found in tissue specimens. *Candida albicans* is the species most often associated with disease of the small intestine. The most common lesions associated with *Candida* are single or multiple ulcerations. [176], [178] Superficial erosions, bloody masses, and pseudomembranes also occur, but with less frequency. White plaques and thickened mucosal folds may also be seen on endoscopy. [176]
Clinical Manifestations Candidiasis of the small bowel is rarely diagnosed antemortem, so which symptoms are attributable to fungal invasion is often unclear. The most common symptom associated with intestinal candidiasis is diarrhea. [176] Other reported symptoms include nausea, vomiting, flatulence, abdominal pain, abdominal distention, melena, and hematochezia. [176], [178] Complications include perforation and penetration of intestinal ulcers. [176]
Diagnostic Evaluation Because *Candida* is a normal colonizer of the entire gastrointestinal tract and even more abundant in persons at risk for disease because of decreased immune function and antibiotic treatment, a diagnosis of small intestinal candidiasis is not easily made. The mere presence of the organism on stain or culture of specimens from the small intestine does not establish the diagnosis. A biopsy specimen of the small intestine with histologic evidence of invasion is necessary. *Aspergillus* may also be found on histological sections and may be part of the disease process. [174], [176]
Differential Diagnosis The differential diagnosis includes diseases that involve the small bowel, particularly those seen in immunocompromised hosts, and includes other mycoses, TB enteritis, ischemic bowel, typhlitis, and MAC infection.
Treatment and Prognosis Therapy for *Candida* infection continues to evolve with the increased use of azoles. We now recommend, based on the results of both randomized trials in immunocompetent hosts and observational trials in immunocompromised hosts, [179] 400 mg of fluconazole per day for gastrointestinal candidiasis in stable patients who have not received recent azole therapy. For unstable patients or those who have previously been given therapy with azoles, 0.6 to 0.7 mg of amphotericin B per kilogram per day is recommended. As in other cases of disseminated fungal disease, lipid formulations of amphotericin B can be used if the patient does not respond to therapy or concerns about renal toxicity arise. Therapy should continue be continued for at least 2 weeks and until a clinical response occurs. In patients with temporary neutropenia caused by chemotherapy, treatment should not be stopped until the neutrophil count rises above 500/mm $_3$. Resection

of the diseased bowel or maintenance therapy may be required, especially in patients who are permanently immunosuppressed.

Mucormycosis

Epidemiology Like aspergillosis and candidiasis of the small intestine, small intestinal mucormycosis is most commonly found in persons with impaired defenses, such as patients on steroids or other immunosuppressive medications and patients with diabetes mellitus, malnutrition, chronic renal insufficiency, burns, aplastic anemia, leukemia, lymphoma, or AIDS. [180], [181] Premature infants may also have significant gastrointestinal mucormycosis, which is rapidly fatal within days to weeks after birth. [182], [183] Small intestinal infection is usually acquired through dissemination from a pulmonary source, although some authors have suggested that fungi can enter the body with food. [180]

Etiology and Pathogenesis The usual pathogens, *Rhizopus, Absidia,* and *Mucor,* are ubiquitous in nature and are inhaled as spores that produce infectious particles called *sporangiospores*. Organisms produce disease by invading blood vessel walls, causing infarction and necrosis. [181] They can be identified by their large, nonseptate, thick-walled hyphae with right-angle branching in tissue sections. [181] In culture, they grow as fluffy molds. Mucormycosis has a predilection for certain anatomic sites, including the sinuses, orbits, brain, and lungs. Isolated gastrointestinal lesions are uncommon. Involvement of the gastrointestinal tract may occur as a part of disseminated disease. Mucormycosis may cause mass lesions resembling carcinoma, cause infarction and perforation, or involve the ileocecal region in a pattern resembling perforated appendicitis. Small bowel infarction in neonates resembling necrotizing enterocolitis has also been reported. Mucormycosis can lead to superinfection of preexisting lesions of the gastrointestinal tract, including peptic ulcers, penetrating wounds, and TB. [184]

Clinical Manifestations Fever, abdominal pain, and distention are the most common symptoms. [180], [181] Nausea, vomiting, hematemesis, hematochezia, diarrhea, and melena are seen less commonly. Because multiple sites of the gastrointestinal tract are often involved, it is difficult to determine which symptoms are attributable to small intestinal involvement. In one reported case with only small intestinal involvement, the only symptom was abdominal distention. [180] The physical findings may include abdominal mass lesions or signs of bowel necrosis or perforation. Intestinal perforation is the most common complication. [180], [181]

Diagnostic Evaluation As in other small intestinal mycoses, disease is more commonly diagnosed postmortem than antemortem. [181] A definitive diagnosis requires biopsy for culture and histological examination for evidence of tissue invasion. [185] Endoscopy may reveal ulcerations, especially in the ileum. [180], [186] Culture is frequently unsuccessful, and even when it is successful, it may signify only colonization because the agents of mucormycosis are saprophytic and ubiquitous in nature.

Differential Diagnosis The differential diagnosis includes carcinoma, ischemic bowel, other intestinal mycoses, and TB enteritis.

Treatment and Prognosis The prognosis is usually poor, and the mortality rate is high regardless of therapy. [181] The treatment of choice is 1.0 to 1.5 mg of amphotericin B deoxycholate per kilogram per day with surgical resection. [187] Lipid formulations of amphotericin B can be used in higher doses to treat patients with renal insufficiency or those not responding to initial therapy. Once a clinical response to amphotericin B occurs, dosing can be decreased to every other day. The duration of therapy is unknown and may be lifelong. Fluconazole and itraconazole are not active, as they are in other mycotic infections.

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CHAPTER 76

Paul J. Ciclitira and H. Julia Ellis

CELIAC DISEASE

DEFINITION

Celiac disease is a condition in which an abnormal small intestinal mucosa reverts toward normal when patients are treated with a gluten-free diet and relapses when gluten is reintroduced. The disorder is sometimes called *celiac sprue* or *gluten-sensitive enteropathy*. It was previously termed *nontropical sprue, celiac syndrome, idiopathic steatorrhea,* or *primary malabsorption*.

Dermatitis herpetiformis is a related disorder characterized by an itchy, blistering skin eruption that frequently affects the knees, elbows, buttocks, and back. Patients with dermatitis herpetiformis invariably have some degree of related gluten-sensitive small intestinal enteropathy.

HISTORY

Celiac disease was mentioned by Aretaeus the Cappadocian in the second century [1]; the term denoted an illness associated with the abdominal cavity. “If diarrhoea does not proceed from a slight cause of only one or two days duration and if, in addition, the patient’s general system be debilitated by atrophy of the body, the celiac disease of a chronic nature is formed.” Samuel Gee, [2] who in 1888 described the condition, reported that the disorder affects all age groups. He recognized that “if the patient can be cured at all it must be by means of the diet.”

Dicke, during the early 1930s, became convinced that the consumption of bread and wheat flour had an adverse effect on patients with this condition. His first report appeared in *Het Nederlands Tijdschriftvoor Geneeskunde* in 1941, and his observations were expanded in his doctoral thesis, submitted to the University of Utrecht in 1950. During World War II in The Netherlands, food, particularly cereals used to make bread, was scarce. During this time, the condition of children with celiac disease improved, only to relapse following supply, by the Swedish Air Force, of bread at the end of the war. It was this serendipitous observation that led to the finding that wheat exacerbates celiac disease. [3]

EPIDEMIOLOGY

Celiac disease affects a large number of ethnic groups, but mostly people of European origin, including those living in North America and Australia. The prevalence throughout Europe was previously thought to be about 1 in 1000, with only 1 in 6000 affected in North America. However, serologic testing of blood donors indicates that the actual number in both Europe [4] and America [5] may be closer to 1 in 250. The condition may be even more common in Celtic populations. A report from Ireland suggests a prevalence of 1 in 120. [6]

Celiac disease occurs in non-Caucasians, although the incidence is probably lower. It has been reported in the wheat-consuming population of Bengal and the Punjab rather than the predominantly rice-consuming population of southern India. The condition has been reported in Arabs, Hispanics, Israeli Jews, and Sudanese of mixed Arab and Negro stock. The condition rarely, if ever, affects people from a purely Afro-Caribbean or Chinese background. [7]

The disorder has been reported to affect women more often than men; some authors have suggested a sex (female-to-male) ratio of 2:1, but others think that equal numbers of persons of both sexes are affected.

The survival of persons with untreated celiac disease before the description of a gluten-free diet was poor, with an estimated mortality rate of 20%; reports varied between 10% and 30%. The introduction of a therapeutic gluten-free diet greatly reduced mortality. However, the latest information suggests that the mortality rate for patients with celiac disease may still be twice that of the population without celiac disease, the excess risk being mainly a consequence of non-Hodgkin lymphoma. [8] The risks for neoplasm in patients with celiac disease who continue to ingest gluten were reported by Holmes and colleagues in 1989. [9] These authors reported an increased number of deaths from all cancers; the greatest increase in relative risk was for intestinal lymphoma when celiac patients ingesting gluten were compared

with both normal subjects and celiac patients on a strictly gluten-free diet.

CEREAL CHEMISTRY

The precise structure of the gluten proteins that exacerbate celiac disease remains unknown, although significant advances have been made in this area. Wheat grains comprise three major constituents, which are separated by milling: the outer husk, or bran; the germ; and the endosperm, or white flour, the last of which constitutes 70% to 72% of the whole grain by weight. Cereal storage proteins fall into four groups: the minor albumins, the globulins, the glutenins, and an ethanol-soluble fraction, the prolamins, termed *gliadins* in wheat. The prolamins from other cereals are termed *secalins* (from rye), *hordeins* (from barley), *avenins* (from oats), and *zeins* (from celiac nontoxic maize). The taxonomic relationship of the major cereal grains (*gramina*) is shown in Figure 76-1. Wheat glutenins give this cereal its characteristic baking quality by entrapping carbon dioxide in the dough, which then rises when it is left to prove. The gliadins provide the lubrication required for the glutenin proteins to slide over one another when the dough rises. The term *gluten* denotes the ethanol-soluble fraction and the glutenins.

FIGURE 76-1. Taxonomic relationships of the major cereal grains. (From Kasarda DD, Okita TW, Bernardin JE, et al. Nucleic acid [cDNA] and amino acid sequences of a-type gliadins from wheat [*Triticum aestivum*]. Proc Natl Acad Sci U S A 1984;87:4712.)

The initial separation of wheat proteins depends on their relative solubility characteristics: Gliadins are soluble in 40% to 90% ethanol, and glutenins are insoluble in neutral aqueous solution, saline solution, and ethanol. The gliadins can be subdivided further either according to their relative electrophoretic mobility into a, ß, ?, and ? subfractions or according to their N-terminal amino acid sequence into a, ß, and ? subfractions, the previously described ß subfraction being reclassified as a type of a-gliadin. [10] The molecular weights of these proteins increase from 32 kd for the a-gliadins to 58 kd for the ?-gliadins.

PATHOGENESIS

Enzyme Deficiency and Lectin Binding

Three main hypotheses for the pathogenesis of celiac disease have been proposed. According to the first, a small intestinal peptidase deficiency results in an inability to digest gluten, which in turn damages the small intestine. This theory has now been largely discounted, [11] although the possibility still exists that lack of an enterocyte intracellular enzyme may in some way affect gliadin peptide transport or processing. In the second, an enterocyte membrane defect allows lectin-like binding of gliadin to the small intestinal enterocytes, altering cell function and resulting in premature cell death. This hypothesis has fallen from favor. [12] It is now generally accepted that celiac disease is mediated by the cells of the immune system, with the normal oral tolerance to food antigens bypassed in a manner not yet understood. Considerable evidence is available to support this hypothesis.

Immune Hypersensitivity

A dense infiltration of the lamina propria with lymphocytes and plasma cells is found in the jejunal biopsy specimens of patients with untreated celiac disease. Furthermore, the ratio of intraepithelial lymphocytes to surface enterocytes is increased; most intraepithelial lymphocytes express the suppresser/cytotoxic phenotype and have the appearance of immunoblasts. [13] Splenic atrophy is known to occur, and a strong association with the histocompatibility antigen HLA-DQ2 has been noted, which is linked with autoimmune disorders. Patients with untreated celiac disease have high titers of circulating antibodies to gluten and antibodies to reticulin and endomysium. [14] The titers of these antibodies are known to decline when gluten is excluded from the diet, so that investigators have assumed that their presence is secondary to pathogenesis. However, the discovery that the antigen for antiendomysial antibody is tissue transglutaminase (tTG) [15] has opened a new chapter in our understanding of the condition. [16]

The action of this small intestinal enzyme on gliadin peptides has been shown to be crucial for T-cell activation in vitro. It appears that within antigenic gliadin peptides, certain glutamine residues are specifically targeted by tTG and deamidated to glutamic acid. This process greatly increases binding of the peptides to DQ2 and DQ8 molecules and facilitates T-cell activation. It has further been suggested that tTG may generate additional antigenic T-cell neoepitopes by cross-linking extracellular matrix proteins with gliadin (16).

The presence of a high proportion of T cells bearing the ?/d T-cell receptor in the epithelial compartment of the small intestinal mucosa of patients with untreated celiac disease has long been noted. As our understanding of the role of these primitive lymphocytes has increased, the concept has emerged that these cells may act to protect the mucosa from damage during chronic exposure to antigens such as gluten in those individuals in whom normal development of tolerance has failed. [16]

The finding of increased numbers of cells expressing the proinflammatory cytokines interferon-? (IFN-?) and tumor necrosis factor-a (TNF-a), but not interleukin-4 (IL-4), [17] supports the immune-mediated hypothesis. The isolation of gluten-sensitive T cells from both the peripheral blood and small intestinal biopsy specimens of patients with celiac disease, [18] , [19] the HLA-DQ2 restriction of most of these cells, and their production of proinflammatory cytokines when stimulated with wheat gliadin [20] provide further evidence, as does the phenomenon of “gliadin shock,” or “gluten shock.” This condition, which occurs in only a few treated patients with celiac disease after gluten challenge, is characterized by vomiting, tachycardia, and cardiovascular collapse; it responds to treatment with corticosteroids.

We previously proposed that gliadin is absorbed initially across and between the small intestinal enterocytes and reaches the dendritic cells in the lamina propria. These cells process the antigen and present it in conjunction with HLA-DQ2 to sensitized T cells (afferent limb). The sensitized cells generate immune products that damage the epithelial cells (efferent limb). The lymphocyte products, most notably IFN-?, induce HLA class II gene product expression in the small intestinal enterocytes, which can then present further antigen to the sensitized lymphocytes. This process is shown schematically in Figure 76-2. The reason for the breakdown in normal oral tolerance is not known.

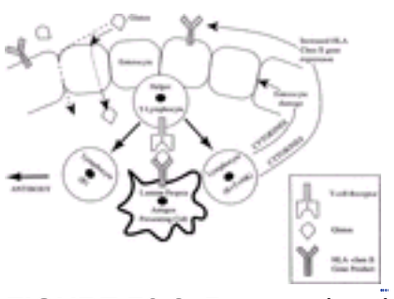

FIGURE 76-2. Proposed pathogenetic mechanisms of celiac disease. Gluten is absorbed into the lamina propria and presented in conjunction with human leukocyte antigen (HLA)–DQ2 or HLA-DQ8 cell surface antigens by dendritic cells to sensitized T lymphocytes expressing the a/ß T-cell receptor (afferent limb). These lymphocytes then activate other lymphocytes to generate immune products that damage the enterocytes (efferent limb). The induction of aberrant HLA class II cell surface antigens on the enterocytes permits additional antigen presentation by these cells to the sensitized lymphocytes.

Toxicity Studies

Initial work attempting to characterize the “fraction” within cereals that exacerbates celiac disease involved separation of the proteins according to the method described by Frazer and colleagues. [21] This method entailed the physiological digestion of wheat proteins with pepsin and trypsin, followed by separation according to their solubility properties. Six fractions were produced, all of which were toxic to celiac patients with the exception of fraction 6, which was studied in only one subject.

Hekkens and associates [22] purified a-gliadin to 60% purity and investigated the toxicity of this fraction in a single subject with celiac disease who was in remission. They concluded that a-gliadin was toxic to the subject, based on a decrease in small intestine brush border enzyme levels in several biopsy specimens taken over 55 hours. They did not report the results of assessments of the other gliadin fractions.

Both Howdle and colleagues [23] and Jos and associates [24] studied the celiac toxicity of gliadin fractions using a celiac in vitro jejunal biopsy organ-culture technique. They independently assessed the effects of the a, ß, ?, and ? subfractions of gliadin, and both groups concluded that all the gliadin subfractions are enterotoxic to celiac small intestinal mucosa. The subject remains controversial, but in vivo studies indicate that all four gliadin subfractions exacerbate celiac disease. [25]

Kagnoff and colleagues [26] suggested that environmental factors may initiate celiac disease. They conducted a computer analysis of a bank of protein sequences in search of potential sharing of amino acid sequence homologies with aggregated A-gliadin, a type of a-gliadin; the analysis revealed shared amino acid residues in a span of 12 amino acid residues and an identical pentapeptide with the 54-kd E1b coat protein of adenovirus 12, an adenovirus usually isolated from the gastrointestinal tract. Cross-reactivity of an antibody to the adenovirus with A-gliadin was seen, implying shared immunogenicity. These authors proposed that in susceptible persons, an encounter of the immune system with this adenovirus may be important in the pathogenesis of celiac disease. [26], [27]

The fraction of wheat that exacerbates celiac disease is a protein because defatted a-gliadin, which contains less than 0.007 mol of carbohydrate per mole of protein, exacerbates the condition. [28] All four gliadin subfractions share epitopes that also are found in rye secalins, as demonstrated by both polyclonal and monoclonal antibody studies. Detoxification of wheat gluten proteins by deamidation implies that glutamine, which contributes more than 35% of the amino acids within gliadin, may be involved, although the method used also may have altered the structure of other amino acid residues. [29]

It is possible to isolate and clone gliadin-sensitive T cells from both the peripheral blood [19], [30] and small intestinal mucosa [18] of patients with celiac disease. The first of these clones isolated from the peripheral blood [30] of a patient with celiac disease was restricted by HLA-DQ2 and responded to a peptide corresponding to amino acids 31 through 49 of A-gliadin. Removal of any residues from the C-terminal and more than two residues from the N-terminal abrogated the sensitivity of the cloned T cells to these smaller peptides. [30] More recently, a number of T-cell clones have been isolated from the small bowel of patients with celiac disease. The antigen specificity of some of these has been elucidated and may be summarized as follows [31]: three DQ2-restricted epitopes, of which one originates from ?-gliadin and two from a-gliadin, and two DQ8-restricted epitopes, of which one arises from a-gliadin and one from a glutenin protein. The two DQ2-restricted a-gliadin epitopes are of particular interest in that they comprise overlapping sequences, [18] and when they were tested with a panel of clones from DQ2-positive Norwegian patients, between them they stimulated all clones. A very similar peptide was found to stimulate interferon-? production in peripheral blood T cells of patients with celiac disease following gluten challenge. [19] It has been suggested [18], [19] that these peptides may represent the immunodominant epitopes in celiac disease. An in vivo challenge study has recently confirmed the toxicity of these peptides in four celiae patients. [32] however, this awaits confirmation. A preliminary celiac small bowel organ-culture study has suggested that a similar but not identical peptide does indeed cause in vitro damage to the celiac duodenal mucosa. [32] However, in this study, only one histological variable was measured. Further data are required to confirm this finding.

An earlier feeding study with peptides corresponding to amino acid residues 31 through 49 (peptide A), 202 through 220 (peptide B), and 3 through 21 (peptide C) of A-gliadin concluded that the peptide corresponding to amino acids 31 through 49 of A-gliadin is enterotoxic in celiac disease, whereas the other peptides are not. [33], [34] However, although one report of a peripheral blood clone that responds to peptide A has been published, [30] to date no small intestinal T-cell line or clone has been shown to respond to this peptide. It is possible that peptide A represents a minor epitope.

Several groups are independently studying the binding of sequenced gliadin proteins to the product of HLA-DQ2, the inheritance of the encoding alleles for which is closely linked to celiac disease. Two approaches are being used; one involves the binding of radioactive iodine–labeled peptides to affinity-purified HLA-DQ2 molecules, [35] and the other involves a cell-binding assay using biotinylated peptides and subsequent analysis by flow-activated cell-sorter analysis (FACS). [36] The initial results show binding of gliadin peptides, including a peptide corresponding to amino acid residues 31 through 49 of A-gliadin, to HLA-DQ2. This finding has permitted computer-aided modeling of the binding of gluten proteins to HLA-DQ2, an antigen-presenting molecule thought to be involved in the pathogenesis of celiac disease. [35], [36]

Genetics

The precise means of inheritance of both celiac disease and dermatitis herpetiformis is unknown, although 10% of first-degree relatives of probands may be similarly affected. The concordance rates are 70% to 100% for identical twins but only 30% for nonidentical twins, which supports a genetic etiology for celiac disease. Efforts to understand the mechanisms and genetics of polygenic human diseases have focused on identifying DNA or protein products and protein molecules that segregate with diseases in both populations and families.

The most significant observation is the increased frequency of specific serologically defined lymphoid cell surface proteins, termed *HLA class II molecules,* in persons with celiac disease. These are glycosylated transmembrane heterodimers consisting of both a and ß chains, the genes for which are organized into three related subregions: DR, DP, and DQ. These subregions are shown in Figure 76-3. [37] The genes are encoded within the HLA class II region of the major histocompatibility complex on the short arm of chromosome 6. The association of particular HLA-DR and -DQ types with both celiac disease and dermatitis herpetiformis is well established. [38], [39] Associations with the HLA-DP region and TNF-a genes have been reported but are thought to be secondary to linkage disequilibrium with HLA-DR and -DQ haplotypes. The genes most strongly associated with celiac disease are *DQA1*0501, DQB1*0201;* 98% of northern Europeans with celiac disease have these alleles in *cis* (DQ2), whereas in southern Europe, a third of the population with this disease express the same class II molecule from these alleles in *trans* (DR5, DR7). [38] Italian and Argentinian populations with celiac disease also are reported to have an increased frequency of the DR5, DR7 genotype. [40] In Israel, an association has been found between the HLA-DR4, -DQ8 genotype and celiac disease. This genotype encodes a class II molecule with considerable similarities in the peptide-binding groove configuration to that produced by the DQ2 genes, supporting a central role for the HLA class II molecule in an immune-mediated model of celiac disease. [41]

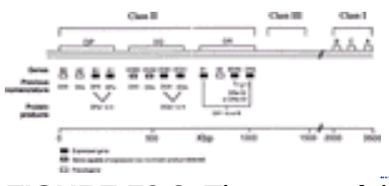

FIGURE 76-3. The gene of the human leukocyte antigen class II region. (From Trowsdale J, Campbell RD. Physical map of the human HLA region. Immunol Today 1988;9:2.)

It is estimated that the HLA associations account for only 30% of the genetic susceptibility to celiac disease, as evidenced by the prevalence of the susceptibility DR3, DQ2 haplotype in up to 25% of the general British and American populations. Segregation analyses propose an oligogenic model involving both the HLA-DQ alleles and the involvement of one or more other genes located elsewhere on the genome. [42], [43] To narrow the search for these non-HLA susceptibility loci, three genome-wide linkage studies using either sibling pairs or extended pedigrees from multiply affected celiac families have so far been published. These have produced largely conflicting results, and four follow-up studies of candidate regions suggested by the genome-wide studies have mostly failed to replicate linkage to celiac

disease in these regions. At present, no linkage to any single region has been consistently replicated between populations. The current state of our knowledge in this rapidly changing field has been reviewed by King and Ciclitira. [44]

PATHOLOGY

Celiac Disease

Celiac disease affects primarily the mucosa of the small intestine; the abnormalities are most marked proximally and decrease in severity with distal progression through the small intestine. In severe cases, however, the lesion can extend to the ileum and also may affect the stomach and colon.

The characteristic histological appearance of the jejunal mucosa in normal subjects is shown in Figure 76-4 and can be compared with that of a biopsy specimen from a patient with untreated celiac disease, shown in Figure 76-5. In the classical flat mucosa, no villi are seen; the normal architecture of the villi is lost, and the normal ratio of villus height to crypt depth (between 5:1 and 3:1) is reduced. The appearance of the mucosa can range from mild flattening through partial atrophy of the villi to a total absence of villi. The total thickness of the mucosa usually is increased because of crypt hyperplasia and cellular infiltration of the lamina propria with plasma cells and lymphocytes. The surface epithelial cells become pseudostratified, as opposed to their normal tall columnar shape, and surface enterocyte height is reduced. Crypt mitotic activity is no longer confined to the base, and although the histological appearance of the crypt is otherwise normal, crypt abscesses have been described. Cell migration time from the base of the crypt to the tip of the villus is reduced in untreated celiac disease to between 12 and 24 hours; the normal time is 3 to 5 days. The ratio of intraepithelial lymphocytes to surface enterocytes is increased in patients with active disease; the normal ratio of lymphocytes is 10 to 30 per 100 epithelial cells.

FIGURE 76-4. Histological appearance of the jejunal mucosa of a normal subject.

FIGURE 76-5. Histological appearance of the jejunal mucosa of a patient with untreated celiac disease. Note the absence of villi, expanded mononuclear cell population, and increased number of intraepithelial lymphocytes.

Isaacson [45] has suggested that the crypt abscesses and small ulcers represent underlying T-cell lymphoma. These ulcers can become troublesome and occasionally develop into a frank small intestinal lymphoma that was formerly thought to complicate 6% to 10% of cases of celiac disease. This figure may, however, be an overestimate, given the more recently described increased prevalence of celiac disease in various populations. Goblet cells are evident throughout the crypt and along the epithelial surface. Paneth cells in untreated celiac disease may discharge their contents into the crypts more readily, so that they are difficult to recognize and appear to be reduced in number. The numbers of endocrine-secreting cells in the small intestine of patients with untreated celiac disease may be both increased and decreased. In some cases, the basement membrane changes—that is, it develops the staining characteristics of collagen rather than reticulin, although this abnormality is frequently patchy.

Chronic inflammatory cells infiltrate the mucosa of the small intestine in untreated celiac disease. Also, the number of plasma cells in the lamina propria increases. Most intraepithelial lymphocytes express the common leukocyte antigen CD3, 70% express the suppressor/cytotoxic phenotype CD8, 5% express the helper/inducer CD4 phenotype, and 20% are $CD3^-$, $CD4^-$, and $CD8^-$. Most of these cells express the more primitive ?/d rather than the more usual a/ß T-cell receptor; as a result, the number of ?/d T-cell receptor–positive lymphocytes in the surface epithelium of the small intestine is significantly increased in both untreated and treated celiac disease.

Dermatitis Herpetiformis

In addition to dermatitis herpetiformis, several other skin disorders are associated with celiac disease: psoriasis, eczema, pustular dermatitis, cutaneous amyloid, cutaneous vasculitis, nodular prurigo, acquired ichthyosis, epidermal necrolysis, pityriasis rubra pilaris, and mycosis fungoides. Atopic eczema in celiac patients may respond to a gluten-free diet.

Dermatitis herpetiformis is the skin disorder most commonly associated with celiac disease, gluten-sensitive enteropathy being common to both conditions. Dermatitis herpetiformis is, however, relatively rare compared with celiac disease. Because figures for the prevalence of gluten sensitivity, as judged by serologic screening tests, have changed considerably in recent years, [4], [5] and [6] it is impossible to estimate the number of patients with celiac disease who also have dermatitis herpetiformis.

Dermatitis herpetiformis is characterized by an itchy papular vesicular eruption, usually symmetrically distributed on the elbows, knees, buttocks, sacrum, face, neck, and trunk and occasionally within the mouth. The predominant symptoms include itching and burning, which may become sufficiently severe to cause considerable pain. Rupture of the blisters results in a rapid relief of symptoms.

Only 10% of patients have symptoms attributable to malabsorption, although nearly 100% have some abnormality of the jejunal mucosa. Gastrointestinal symptoms, when present, are similar to those of celiac disease, although usually milder. The small intestinal lesion is patchy, and several biopsy specimens may be needed to demonstrate it. [46], [47] The risk for gastrointestinal lymphoma and malignancy is increased, as in celiac disease, [9] if a gluten-free diet is not adhered to strictly. Lowered circulating levels of IgM, raised levels of IgA, and variable changes in IgG occur in dermatitis herpetiformis, similar to the pattern observed in celiac disease.

The diagnosis of dermatitis herpetiformis requires the demonstration of granular IgA at the dermoepidermal junction in an area of skin not affected by blistering. This permits discrimination of the condition from another disorder, termed *linear IgA disease,* with a similar presentation but not associated with gluten-sensitive enteropathy.

The precise pathogenesis of dermatitis herpetiformis remains unknown, although it has been suggested that the formation of antibodies in the small intestinal mucosa is stimulated by the presence of gluten. The antibodies are carried through the circulation and bind at the dermoepidermal junction, causing the classical skin lesions.

Dermatitis herpetiformis is treated with dapsone, which should be given at a dosage of 1 to 2 mg/kg daily. All patients with dermatitis herpetiformis should be advised

to adhere to a gluten-free diet, although symptoms may not diminish significantly for 6 to 12 months. The dose of dapsone usually can be lowered, and in many cases it can be stopped. The problem is that it frequently takes months or even years for the full effect of the diet to be evident, by which time the patient may be unwilling to continue to maintain a gluten-free diet. [48]

Grades of Enteropathy

The classical celiac lesion of the small bowel has been described. However, it has become apparent that grades of enteropathy exist within the gluten-sensitive spectrum. Marsh [49] has classified them as follows: Grade 1 is an infiltrative lesion without any change in the architecture of villi but with an excess of lymphocytes in the epithelium. This lesion is typically found in patients with dermatitis herpetiformis and is not associated with symptoms. The hyperplastic, or grade 2, lesion is similar to the grade 1 lesion, but the crypts are hypertrophic, and the epithelium is also infiltrated with lymphocytes. The destructive, grade 3 lesion is the typical flat mucosa of untreated celiac disease. These changes are not exclusive to celiac disease; they may also occur in a number of other conditions, such as tropical sprue, giardiasis, and short-term food sensitivities in infants (see section " Differential Diagnosis "). Two further lesions occur, one at each end of the spectrum. The grade 0, or preinfiltrative, lesion is seen in some cases of dermatitis herpetiformis and is characteristic of latent celiac disease. The biopsy specimen appears totally normal but can be shown in the laboratory to be producing antibody to gluten and endomysium. At the time of this writing, patients with the grade 0 lesion are not treated with a gluten-free diet, but this may change. The grade 4 lesion, at the other end of the spectrum, is a hypoplastic lesion and is unresponsive to gluten withdrawal. It is seen in cases of intestinal ulceration, jejunoileitis, and enteropathy-associated T-cell lymphoma. It appears that the clonal expansion of malignant T cells renders the mucosa unresponsive to antigen withdrawal, resulting in hypoplasia and progressive atrophy of the entire lining of the intestinal wall.

Differential Diagnosis

A number of conditions may cause enteropathy similar to that of the celiac spectrum, so that it is essential to demonstrate that a lesion is gluten-dependent. Giardiasis may cause grades of enteropathy from infiltrative to destructive, as may tropical sprue, marasmus, and sensitivity to cows' milk protein. Other enteropathies associated with food proteins, such as those in egg, soy, chicken, or fish, tend to result in a destructive-type lesion. Other conditions causing some form of atrophy of the villi include acute bacterial or viral infection, bacterial overgrowth, human immunodeficiency virus (HIV) infection, histoplasmosis, and tuberculosis. Noninfectious causes are starvation (including anorexia nervosa), Whipple disease, nonsteroidal antiinflammatory agents, radiation, disorders of the immune response (e.g., hypogammmaglobulinemia), and Crohn's disease.

The grade 4 hypoplastic lesion is seen in unresponsive sprue and is associated with a number of interrelated conditions, including refractory sprue, collagenous sprue, ulcerative jejunitis, autoimmune enteropathy, and lymphoma. Refractory sprue is discussed in a later section.

Involvement of Other Parts of the Gut

It has become apparent that although primarily the small bowel is affected in celiac disease, other parts of the gastrointestinal tract are also sensitive to gluten. Local challenge of the buccal mucosa in patients with treated celiac disease leads to an increase in the numbers of $CD4^+$ cells in the lamina propria. [50] Results of studies suggest that gastric atrophy and achlorhydria are fairly common in patients with gluten sensitivity. [49] About 10% of patients with celiac disease may also have a lymphocytic gastritis, [51] defined by the presence of 25 or more intraepithelial lymphocytes per 100 gastric columnar cells. The intraepithelial lymphocytes are of T-cell lineage. In one series, the patients with celiac disease who did not have lymphocytic gastritis, as defined above, did nonetheless have significantly more intraepithelial lymphocytes than the patients in a control group.

Microscopic colitis is characterized by mucosal inflammation of an endoscopically normal colon. Depending on whether or not a thickened subepithelial collagen band is present, two separate terms are used to describe the condition: *collagenous colitis* and *lymphocytic colitis*. A feature of the colonic inflammatory reaction is intraepithelial lymphocytosis, suggesting that an immunostimulating agent may be presented to mucosa-associated lymphoid tissue. Potential agents include dietary antigens. [52] Microscopic colitis can be diagnosed only by rectal biopsy, although a full colonoscopy should then be undertaken. [53] Gluten challenge of the rectal mucosa of patients with treated celiac disease causes an increase in the number of intraepithelial lymphocytes, and this has been suggested as a possible means of diagnosis. [54], [55]

CLINICAL FEATURES

The classical description of Samuel Gee, [2] with its evocative account, was concerned largely with gross manifestations of the disorder. This florid presentation is unusual in the Western world, and although some patients have severe illness, many others have few or no symptoms at diagnosis. Such cases can be found by screening the relatives of patients during research studies or screening patients with associated disorders, such as diabetes mellitus or Down syndrome. Abnormalities encountered in routine hematologic or biochemical profiles or knowledge of HLA class II profiles may provide diagnostic indicators of celiac disease.

Infancy and Childhood

Classically, celiac disease in infancy appears shortly after weaning, when cereals are first introduced into the diet. Failure to thrive is associated with apathy, pallor, anorexia, and abdominal distention. Developmental delay, with particular retardation of motor skills, sometimes occurs in more extreme cases, in which muscle wasting and hypotonia may be features. Because of numerous other symptoms, the patient may be brought to medical attention. Presenting complaints can include behavioral problems and rectal prolapse, which is much more common in cystic fibrosis.

The child with untreated celiac disease typically passes soft, bulky, clay-colored stools with an offensive odor, but watery diarrhea or constipation is occasionally reported. Young children may present with effortless vomiting, which can be of large volume and usually is associated with abdominal distention but little or no diarrhea. Abdominal pain is sometimes so severe that the child undergoes laparotomy because of a mistaken diagnosis of intestinal obstruction. Older children tend to have more varied symptoms and present with anemia, rickets, or failure to thrive, so that they fall below the third percentile for weight. Height also may be affected, although usually less than weight. Unexplained short stature should be an indication for jejunal biopsy, even when gastrointestinal symptoms are mild or absent. Catch-up growth, once a gluten-free diet is commenced, is well documented. [56] Biochemical vitamin D deficiency or even clinical rickets is not uncommon, especially among Asian children with untreated celiac disease. This situation improves with the introduction of a gluten-free diet, although supplements may be required in the short term.

Considerable debate continues about why celiac disease now tends to be diagnosed later and with milder signs and symptoms than previously. The impact of changes in infant feeding practices remains unclear, and the total amount of gluten presented to the immature gastrointestinal system, in addition to the age of the child at introduction, may be important factors. [57]

Adulthood

It is not uncommon for adults and adolescents with celiac disease to have had illness for many years, but with symptoms insufficiently severe for them to seek medical advice. An individual may come to accept chronic ill health as normal. Sometimes, an additional illness or surgery precipitates symptomatic diarrhea and a clinical presentation. The presentation that leads to a diagnosis may be with varied symptoms and to almost any hospital department. [58] The patient usually, but not always, has diarrhea at diagnosis, and constitutional symptoms (e.g., unexplained lassitude, weight loss, glossitis, angular stomatitis, cheilosis) and symptoms pertaining to anemia are common. Aphthous stomatitis may be the only presenting symptom, so that it is important to exclude celiac disease in all cases of severe recurrent, unexplained aphthous mouth ulceration. Nausea and vomiting usually are associated only with severe bouts of diarrhea, and although anorexia can occur, some younger adults and pediatric patients have an increased appetite. Failure to gain weight despite the ingestion of 7000 to 8000 calories a day has been reported.

Typically, the patient with untreated celiac disease has diarrhea, passing three to four times a day loose movements that are pale, often foul-smelling, occasionally frothy, and sometimes difficult to flush away; some patients report that they have to keep a bucket of water near the toilet to flush the stools. Specific questioning about stool color and frequency is essential because some patients may regard this type of bowel habit as normal, particularly when the diagnosis has been delayed. Normally formed and colored stools do not preclude a diagnosis of celiac disease, and the bowel habit fluctuates depending on gluten intake.

Abdominal pain with borborygmi, visible peristalsis, and symptoms of increased flatulence are reported, but severe pain is uncommon in celiac disease and should

alert the clinician to other forms of intra-abdominal pathology, such as volvulus, intussusception, cholelithiasis, peptic ulcer, and pancreatitis.

Bleeding into the skin may reflect vitamin K deficiency or scurvy complicating celiac disease, which was common at one time but is rarely seen today. Purpura was reported in l0% of patients and was the presenting feature in 20% of cases before the therapeutic effect of a gluten-free diet was discovered.

Amenorrhea and spontaneous abortions occur more commonly in untreated patients with celiac disease than in normal persons or patients with celiac disease who are on a gluten-free diet. [59] Female infertility or male impotence is the mode of presentation for some adults with celiac disease. In some women, the condition may be unmasked by the metabolic demands of pregnancy. Untreated celiac disease can be associated with an abnormal psychological state or frank psychiatric symptoms, including schizophrenia and depression, which usually diminish when the patient is on a gluten-free diet.

It is important to remember that frequently the symptoms are nonspecific, and the index of suspicion should be high when significantly abnormal immunologic, hematologic, or biochemical profiles are encountered. In particular, the finding of an unexplained mild macrocytic anemia with a persistently low serum or red cell folate level should be an indication for further investigations, including an investigation of possible celiac disease. A common problem is a previous diagnosis of celiac disease in childhood that has been forgotten by both patient and doctor, so that the patient incorrectly resumes a normal diet during adolescence but does not immediately experience symptoms. Careful inquiry should therefore be made regarding previous medical problems, such as childhood diarrhea, short stature, recurrent anemia, and rickets. It should not be forgotten that the prevalence of the condition among first-degree relatives of affected subjects is high. It is in such relatives that latent celiac disease is most likely to appear—that is, persons who have normal jejunal biopsy findings while on a gluten-containing diet but who at some time have had or will have a gluten-sensitive enteropathy. [60], [61] These individuals may be detected by screening tests that reveal the presence of serum antibodies to gliadin and endomysium, but on biopsy, their architecture is normal (Marsh grade 0). [49]

Osteomalacia, rickets, and bone pain, although common in children, may lead to a diagnosis of celiac disease, reflecting poor calcium and vitamin D absorption. Symptoms pertaining to low levels of calcium and vitamin D are frequently insidious and may become severe before they are recognized. Vitamin D deficiency or osteopenia may be the presenting problem when celiac disease is diagnosed in adults.

The ages at presentation of a group of adults who were found to have celiac disease are shown in Figure 76-6.

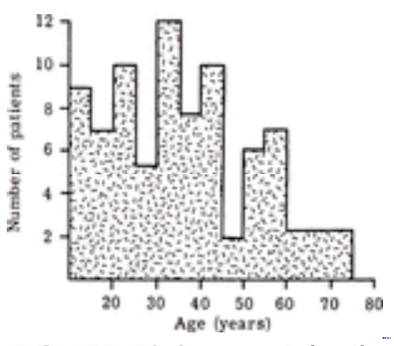

FIGURE 76-6. Age at the time of diagnosis of celiac disease in 84 adults. (From Ferguson A. Coeliac disease [gluten hypersensitivity]. J Hum Nutr 1976;30:193.)

Physical Signs

Psychological abnormalities are not uncommon in newly presenting patients. Affected children are usually irritable and unhappy; these signs, combined with a history of diarrhea, should alert the clinician to the diagnosis. Adults may present with depression, Korsakov syndrome, neurasthenia, or frank symptoms of schizophrenia. Although most patients with celiac disease do not appear psychologically abnormal in comparison with their peers, many affected subjects report a striking improvement in mood shortly after commencing a gluten-free diet. [62], [63]

Patients with celiac disease frequently are of short stature. Growth retardation is common in children, but when they start a gluten-free diet before puberty, a compensatory growth spurt usually occurs, so that the effect on adult height is minimized. Persons with celiac disease are on average 8 cm shorter than their peers. Tall patients are seen, however, and a height of more than 1.8 m does not preclude the diagnosis. Weight loss is common in celiac disease in both children and adults. Sixty percent of children with newly diagnosed disease fall below the third percentile for weight. The average loss of weight in one series of adults was 124.5 kg, and 10% had lost more than 12 kg since the onset of symptoms. It is common for adults with celiac disease to experience a weight gain of more than 6 kg following institution of a gluten-free diet. Edema and ascites are rare but can occur secondary to hypoproteinemia. Abdominal distention and general dilation of the intestinal tract are common in celiac disease, so that the abdomen has a doughy feel, which makes the detection of ascites difficult.

Clubbing of the fingers may occur. Koilonychia may develop and is associated with long-standing iron deficiency anemia. Tooth eruption may be delayed in children, and hypoplasia of dental enamel has been described. [64]

Gynecologic and obstetrical problems are common in women with untreated celiac disease. [59] Delayed menarche (typically by a year) is frequent in untreated subjects. Amenorrhea may occur, and menstrual loss is frequently less than normal. The average age at menopause is 53 years in women with celiac disease who are on a gluten-free diet. Women with untreated celiac disease may present with infertility, [65] and the mean time required for female patients with celiac disease to conceive on a normal diet is 19 months, compared with 12 months for those on a gluten-free diet. It is common for infertile women with celiac disease to become pregnant shortly after commencing a gluten-free diet. Spontaneous abortion or miscarriage is common in female patients with untreated celiac disease. [66] Reduced potency and an abnormally low sperm count are common in adult men with untreated celiac disease. Levels of plasma testosterone, free testosterone, and plasma 5-dihydrotestosterone are low; the level of plasma luteinizing hormone is often raised, which may explain the frequently observed delay in puberty and late development of secondary sex characteristics in untreated male patients with celiac disease. All these abnormalities disappear following prolonged treatment by exclusion of gluten from the diet.

Some untreated persons have a low-grade fever associated with anemia; this finding may indicate a concurrent complication, such as malignancy and, in particular, lymphoma. Peripheral lymphadenopathy is uncommon in celiac disease, and if significant lymphadenopathy is found, the clinician should suspect complicating lymphoma. Rarely, lymphadenitis is seen that disappears during treatment with a gluten-free diet. Cavitating abdominal mesenteric lymph nodes that resolve with a gluten-free diet also have been reported. Computed tomography (CT) sometimes reveals low-grade lymphadenopathy that frequently is associated with hyposplenism. [67]

ANTIBODY TESTS FOR SCREENING

If a diagnosis of celiac disease is being considered, serologic antibody screening tests can be informative, but they do not replace the need for small intestinal biopsy. In patients with celiac disease, levels of antibody to gliadin of both the IgG and IgA classes are raised. The sensitivity and specificity of IgA antigliadin antibody were recently reported as 83% and 82%, respectively, whereas those of IgG antigliadin antibody were 86% and 76%. [68] It has been our observation that when these tests are performed in laboratories not experienced in this area, the sensitivity can fall to as low as 50%. Raised titers alone should not be used to make a definitive diagnosis because titers of antibodies to gliadin can be elevated in other disorders, such as inflammatory bowel disease and hepatic disease. These antibodies were previously used routinely to screen for celiac disease, but although they are still considered to be of some value, they have been largely superseded by other, more specific tests.

Patients with untreated celiac disease have serum IgA antibodies directed against the connective tissue endomysium. These can be detected on sections of monkey esophagus or, more recently, human umbilical chord. The method requires use of a fluorescence microscope and considerable expertise in interpreting the results. However, a sensitivity of 90% and a specificity of 99% have made antibody measurement the test of choice. [68]

The discovery that tTG is the antigen for antiendomysial antibody [63] has made it possible to develop a cheap and simple enzyme-linked immunosorbent assay (ELISA) for this antibody. IgA anti-tTG has a reported sensitivity of 93% and a specificity of 95%. [68] Concerns have been raised that the quality of the enzyme used as the ELISA coating antigen may vary between centers. The use of human recombinant tTG may help to overcome this problem.

It must be remembered that about 2.5% of patients with celiac disease have selective IgA deficiency, and in such cases, the results of tests for either antigliadin, antiendomysial, or anti-tTG antibody will be falsely negative. To attempt to overcome this problem, some authors have investigated the use of IgG anti-tTG and IgG1 antiendomysial antibody. [69], [70] Measurement of these antibodies appears useful in persons with selective IgA deficiency.

The European Society of Paediatric Gastroenterology and Nutrition (ESPGAN) considers elevated titers of these antibodies to be important both in follow-up and in support of the diagnosis. [71] Estimates of the antibody titers can be useful in detecting intentional or inadvertent ingestion of gluten by patients with celiac disease, particularly children, when they are unwell.

Despite reports of the high sensitivity rates of antiendomysial antibodies in patients with celiac disease, some authors have reported disappointing results of antibody measurement in clinical practice, especially in patients with a minor degree of villus atrophy. [72] When malabsorption is suspected, the absence of celiac antibodies should not be a reason to delay biopsy. Equally, the presence of celiac antibodies should be followed by a small bowel biopsy because serologic tests are not 100% specific.

DIAGNOSIS

Laboratory Tests

A bone marrow evaluation in adults usually reveals megaloblastic erythropoiesis. Forty-five percent of bone marrow aspirates from adults with untreated celiac disease have a pure megaloblastic appearance, 55% exhibit megaloblastosis with a lack of iron, and a small number have a pure iron deficiency.

The characteristic hematologic profile is one of a mild hypochromic macrocytic anemia. Patients with mild disease may present only with anemia secondary to iron or folate deficiency and without gastrointestinal symptoms. [73], [74] Iron deficiency is common in celiac disease as a consequence of impaired absorption and increased losses into the gastrointestinal tract, either because of rapid epithelial cell turnover or associated blood loss. Treatment with a gluten-free diet or corticosteroids increases iron absorption. Severe anemia occurs uncommonly, usually in patients with extensive disease, and should arouse suspicion of a superimposed complication, such as malignancy. The exception is during pregnancy, when patients often present with a combined iron and folic acid deficiency anemia and hemoglobin levels as low as 4 to 6 g/dL. Persistent iron deficiency anemia in treated patients should alert the clinician to the possibility of continued long-term inadvertent gluten intake.

The peripheral blood film may reveal target cells, Howell-Jolly bodies, siderocytes, irregular and crenated red cells, Heinz bodies, microspherocytes, acanthocytes, occasional erythroblasts, and thrombocytosis. [75], [76] An erythroblastic appearance with circulating nucleated red cells, pale ring cells, and Howell-Jolly bodies is suggestive of splenic atrophy, which invariably occurs in celiac disease. [77] When both folate and iron deficiency patterns of anemia are superimposed, variations in the size and shape of the erythrocytes and the degree of hemoglobin may give rise to the characteristic appearance of dimorphic anemia. Serum iron, ferritin, and folic acid levels are invariably low. A macrocytic megaloblastic anemia may occur in both children and adults, although in children, a hypochromic macrocytic anemia that slowly responds to oral iron therapy is more typical. Sometimes, the patient has a remote history of a previous requirement for parenteral iron injections.

The serum folate level is low in most untreated patients with celiac disease. Red cell folate is a better indicator of total body stores, and the level is reduced in 85% of patients with untreated celiac disease. The mean serum levels of vitamin B_{12} of untreated patients with celiac disease are lower than those of controls, although only 14% have levels lower than normal. Moreover, the low values are usually close to the normal range, suggesting that some low levels of vitamin B_{12} are secondary to folate deficiency. Rarely do untreated patients exhibit an anemia secondary to a combined deficiency of vitamin B_{12} and folic acid, which requires combined therapeutic replacement. Normally, vitamin B_{12} levels rise spontaneously when patients are treated with a gluten-free diet. Rarely, pernicious anemia may be present, which is linked to the DR3, DQ2 haplotype.

Patients with persistent, frequent loose stools are at risk for the development of sodium and potassium depletion, which results in fatigue and a loss of muscle power and may present with symptoms resembling those of a peripheral neuropathy. Potassium depletion can simulate a renal disorder, but oral potassium or sodium replacement will reverse this problem. Untreated patients with celiac disease frequently have hypocalcemia and hypomagnesemia, although the latter rarely occurs independently. Symptoms are unlikely to develop unless the serum calcium level falls below 1.5 mEq/L. These include personality changes, limb tremors, uncoordinated movements, convulsions, increased limb reflexes, nausea, vomiting, abdominal pain, and paralytic ileus. Malabsorption and insensitivity to vitamin D can occur, the latter causing resistant osteomalacia and elevated serum levels of parathyroid hormone. Rickets, bone pain, or pathological fractures may be presenting features; symptoms pertaining to low levels of calcium and vitamin D are frequently insidious and may become severe before they are recognized. Cramps and tetany also can be associated with low serum calcium and magnesium levels, but calcium and magnesium replacement therapy usually is effective treatment for all the symptoms of deficiency. [78]

Albumin levels are low in up to one third of subjects with untreated celiac disease. Unexplained hypoalbuminemia should be considered an indication for investigating for celiac disease. Severe hypoalbuminemia in a patient receiving active treatment suggests a complication, such as gastrointestinal lymphoma. Zinc deficiency is likely to occur in celiac disease, and if it does, it should be treated by replacement. Rarely, zinc therapy has been reported to induce anemia, apparently as a result of the induction of concomitant copper deficiency. [79] Early reports of elevated circulating immune complex levels in patients with celiac disease have not been confirmed, so their presence is not of diagnostic significance. [80]

Most patients with untreated celiac disease have steatorrhea at some time, but fat excretion is normal in up to 30% at diagnosis. A customary Western diet usually contains 60 to 100 g of fat per day, but patients with untreated celiac disease often discover that they can reduce their diarrhea by decreasing their fat intake. It is therefore recommended that subjects ingest a diet that contains at least 100 g of fat for 2 days before and during the study. All stools should be collected during a 3-day period to eliminate day-to-day variations. The accepted upper limit of normal for fecal fat excretion is 5 or 6 g/d. [81] The estimate is a relatively crude test that is liable to errors of collection or measurement and not specific to celiac disease. Fecal fat estimation no longer has a place in routine investigations for celiac disease. Results of hydrogen and ^{14}C-bile acid breath tests may be mildly abnormal, reflecting a degree of bacterial overgrowth in the small intestine, which usually regresses during treatment of the celiac disease.

Patients with celiac disease characteristically have a flat glucose tolerance test result, although this finding is not of diagnostic value because a significant proportion of normal persons exhibit a similar pattern. Measurement of the excretion of L-rhamnose and lactose after the ingestion of a hypertonic solution containing both these sugars may prove a valuable diagnostic adjunct when obtaining a small intestinal biopsy is difficult. The result is invariably abnormal. One study reported the lactulose–to–L-rhamnose urinary excretion ratio to be sevenfold higher in patients with untreated celiac disease than in controls; no overlap between the two groups was found. [82] These tests can be useful for diagnostic management when other tests are contraindicated, such as during pregnancy, and can be used for research purposes. [83] They should not, however, replace biopsy of the small intestine, which remains the diagnostic investigation of choice.

The xylose absorption test is not useful because urinary excretion values can be normal in up to 20% of patients with untreated celiac disease. This test is now rarely used.

Most patients with celiac disease have a degree of lactose and sucrose intolerance; this usually disappears during treatment with a gluten-free diet. A small percentage of patients, however, exhibit concomitant lactase or sucrase deficiency that does not normalize during treatment with a gluten-free diet, and they usually have persistent diarrhea despite treatment with a gluten-free diet. It is important to identify these patients because appropriate dietary restriction usually causes their symptoms to resolve.

Pancreatic function testing frequently reveals low intralumenal trypsin and lipase concentrations in resting samples. Following stimulation with secretin and pancreozymin, the pancreas is able to respond normally in patients with untreated celiac disease. Similarly, results of the pancreolauryl test are invariably normal in

this condition. Pancreatic function should be assessed only to exclude underlying concurrent pancreatic insufficiency if the patient does not respond to a strict gluten-free diet. Reduced cholecystokinin levels following a fatty meal have been recorded in celiac disease, which may explain the observed gallbladder inaction and diminished pancreatic response to oral challenge observed in patients with untreated celiac disease. [84] A generalized reduction in many gut hormones may be present; levels tend to normalize following the introduction of a gluten-free diet. The exception is a poor rise in plasma gastric inhibitory peptides following an oral stimulus. Grossly elevated levels of enteroglucagon and abnormal responses to oral glucose have been reported to be unique to celiac disease. [85], [86]

Small Bowel Biopsy

The diagnosis may be suspected on clinical grounds or on the basis of various screening tests results. For confirmation, however, it is mandatory to proceed to a small intestinal biopsy. A biopsy of the small intestine should be undertaken in anyone with a history suggestive of celiac disease, including patients with mild symptoms, particularly relatives of probands, and all patients with dermatitis herpetiformis. Even when multiple specimens are taken, some symptomatic relatives may have a normal mucosa even though their serum contains antiendomysial antibodies. [60], [61] The most common indications in childhood are diarrhea, failure to thrive, anemia, and short stature.

Biopsy of the small intestine can be undertaken either endoscopically or with a small intestinal suction (Watson or Crosby-Krugler) biopsy capsule. The hemoglobin level, platelet count, and prothrombin time should be checked before the biopsy is performed because the prothrombin time is increased in a significant number of patients with untreated celiac disease; this can be corrected with parenteral vitamin K.

Endoscopic biopsy specimens should be taken from the second part of the duodenum with the largest forceps, frequently termed *jumbo forceps*. At least three specimens should be taken to avoid difficulties in interpretation resulting from the presence of mucosa without villi that normally overlies Brunner glands. The macroscopic appearance of the duodenum at endoscopy should be noted because duodenal ulcer or patchy duodenitis can produce a pattern of inflammation similar to that found in celiac disease. In severe cases, a loss of the normal circular folds in the second part of the duodenum is seen. This loss, and the loss of normal villi, can be visualized more easily if india ink is injected into the duodenal lumen and the pattern that forms is observed. [87] Typically, in mucosa without villi, a mosaic rather than the usual velvety appearance is seen. The specimen should be retrieved quickly from the capsule or endoscopic forceps and oriented before fixing so that on sectioning the villi are cut in a plane parallel to their longitudinal axis. The cut surface of the specimen should be placed on a small square of absorbent paper (e.g., Millipore filters) with a dissecting microscope and the whole specimen immediately placed in fixative.

It is important that the specimens be correctly oriented before embedding because tangentially cut biopsy specimens make subsequent histological interpretation difficult. [88] Placing the sample mucosa side up on a piece of cellulose nitrate/acetate filter paper provides the correct orientation for cutting because the filter can be included in the paraffin block and processed with the tissue. It is now appreciated that the classical appearance of mucosa without villi is not seen in all cases of gluten-sensitive enteropathy. Marsh [49] has identified a spectrum of gluten-sensitive lesions that may merely represent an increase in the number of intraepithelial lymphocytes of a patient on a gluten-containing diet. A decrease in their number toward normal while the patient is on a gluten-free diet can be taken as evidence of celiac disease.

If larger and better samples are needed, and certainly if the interpretation of an endoscopic biopsy specimen is doubtful, a proximal jejunal suction biopsy should be undertaken with a Crosby-Krugler or Watson biopsy capsule. [89] A Quinton or Rubin biopsy capsule can be used to obtain multiple specimens from the esophagus, stomach, small intestine, or colon [90] and has been used in research studies. [33] The patient fasts overnight. Sedation and local anesthesia to the throat may make the procedure easier for the patient to tolerate but are not mandatory. The patient swallows the capsule, which is attached to a flexible radiopaque tube within a semisoft polypropylene outer tube measuring 4 mm in diameter; a brass ring at the end facilitates manipulation of the capsule into the pyloric antrum. Metoclopramide can be given intravenously to increase intestinal motility. The patient is asked to turn onto the right side, and use of the outer tube permits the capsule to be passed into the duodenum. Radiographic screening can be used to visualize passage of the capsule into the proximal jejunum, where it is fired. In the case of the Watson or Crosby-Krugler capsule, rapid suction is applied through the peroral connecting tube with either a 50- or 20-mL syringe. Inexperienced operators can check the position of the capsule before firing by injecting 15 to 20 mL of water-soluble contrast solution through the connecting tube to outline the jejunum. If this is done, however, the oral tube should be washed through with 20 mL of tap water followed by 30 mL of air to ensure removal of fluid from the capsule, so that it can fire.

Radiology

Dilation of the colon was noted to be an early radiologic sign, although the most common feature is dilation of the small intestine. The upper limit for the diameter of the normal proximal small bowel on barium follow-through is 30 mm in adults, 24 mm in children 2 years of age, and 14 mm in children up to the age of 6 months. The ileum does not exhibit much distention and measures up to 24 mm in normal adults. In normal adults, the diameter of the small intestine may increase to 50 mm during enteroclysis or small bowel enema.

Barium follow-through examination of the small intestine reveals a loss of the fine, feathery mucosal pattern with thin mucosal folds in 85% of subjects (Fig. 76-7). The normal appearance of an enteroclysis examination does not include this feathery pattern. Usually, some straightening of the valvulae conniventes, thickening of the mucosal folds, and an increase in their separation are seen. Superimposed on dilation and thickening of the mucosal folds are varying degrees of flocculation, segmentation, and clumping, which are more relevant when they occur early in the examination. These changes are, however, not specific to celiac disease.

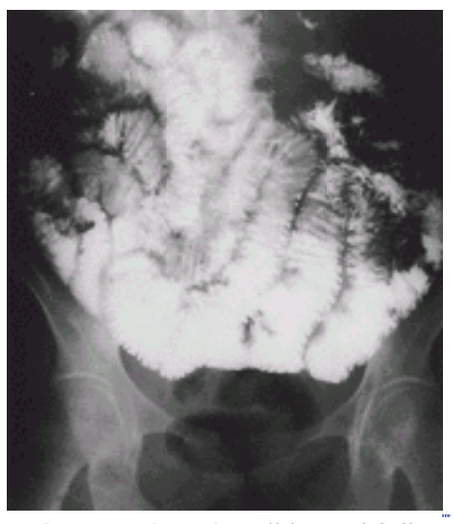

FIGURE 76-7. Small bowel follow-through showing dilated loops of jejunum with the characteristic appearance of untreated celiac disease.

The radiologic findings of most patients with untreated celiac disease suggest the condition, but in other cases, the findings resemble those of Crohn's disease or scleroderma, the latter of which should readily be differentiated on screening by the presence of decreased motility. The findings may also be suggestive of tropical sprue and occasionally chronic pancreatitis. A dilated and often redundant colon is a feature that renders the untreated patient susceptible to the development of volvulus, with resultant abdominal pain. Intussusception can complicate celiac disease and may be diagnosed by the classical radiologic appearance.

Although 25% of patients with untreated celiac disease do not have any radiologic abnormality, radiology remains an important tool in cases with abdominal pain to exclude complicating lesions, such as jejunal ulceration, stricture, lymphoma, and carcinoma. Abdominal CT and nuclear magnetic resonance imaging of the abdomen may provide a diagnostic clue to the presence of celiac disease by revealing hyposplenism, which is invariably associated with the disorder, and abdominal lymphadenopathy.

DISEASE ASSOCIATIONS

The numerous diseases reported to be associated with celiac disease are listed in Table 76-1. [91] In particular, an association with autoimmune disorders has been noted, in which the prevalence of the genes encoding HLA-DR3 and HLA-DQ2 is also high. The relationship to dermatitis herpetiformis has been described.

FACTOR/DISEASE	APPROXIMATE REPORTED INCIDENCE (%)
First-degree relative of celiac patients	10
Dermatitis herpetiformis	U
IgA deficiency[132]	2.5
Hyposplenism[133–136]	100
Aphthous ulceration	5
Down syndrome[100–101]	U
Small intestinal T-cell lymphoma[10]	10
Thyroid disease[137]	6
Diabetes mellitus[92–95]	6
Cutaneous vasculitis[138–140]	U
Liver disease[103–105]	U
Fibrosing alveolitis[106,107]	U
Sjögren syndrome[141]	U
Polymyositis[142]	U
Addison disease	U
Systemic lupus erythematosus[143]	U
Ulcerative colitis/Crohn's disease[108–111]	U
Rheumatoid arthritis	U
Idiopathic pulmonary hemosiderosis[144]	U
Glomerulonephritis[145]	U
Schizophrenia	U
Sarcoidosis	U
Histocompatibility antigens HLA-DQ2 or HLA-DQ8[37–40]	99

U, incidence unknown; HLA, human leukocyte antigen.

TABLE 76-1 Factors and Diseases Associated with Celiac Disease

Diabetes Mellitus

Celiac disease is associated with diabetes mellitus in both children and adults, and the prevalence of diabetes mellitus in patients with celiac disease is between 4% and 6%. [92], [93], [94] and [95] Diarrhea is a symptom of both disorders. The presentation of celiac disease in a diabetic patient often is preceded by poor diabetic control. Anemia, finger clubbing, glossitis, recurrent oral ulceration, abdominal discomfort or distention, and a poor nutritional state or unexpected episodes of hypoglycemia with exacerbations of diarrhea should alert the clinician to the diagnosis. The dietary treatment of concomitant celiac disease improves diabetic control and usually makes possible a reduction in insulin dosage. It has become apparent that individuals in whom celiac disease is diagnosed early in life, particularly before the age of 2 years, are at no greater risk for the development of autoimmune disease, such as insulin-dependent diabetes mellitus, than the general population. This protection is partially abrogated if a gluten challenge is used as part of the diagnostic process. [96]

Neurological Disorders

Patients with celiac disease may have a number of neurological disorders. [97], [98] A strong association has been found between celiac disease and epilepsy, which may or may not involve cerebral calcification. [99] In one series, 5% of adults with celiac disease had epilepsy. Although the seizures of some patients can be controlled by gluten restriction and folate supplementation, drug-resistant seizures develop in others despite these measures (see ref. [97] for review).

The incidence of celiac disease is greatly increased in patients with Down syndrome. [100], [101] This finding prompted investigators to examine a possible association between celiac disease and chromosome 21; however, none was found. [102]

Liver Disease

The incidence of chronic hepatitis, primary biliary cirrhosis, and sclerosing cholangitis is higher than expected in patients with celiac disease. [103], [104] and [105]

Lung Disease

Chronic fibrosing alveolitis and other interstitial lung diseases, including idiopathic pulmonary hemosiderosis, have been reported in association with celiac disease. [106], [107]

Inflammatory Bowel Disease

Celiac disease is associated with inflammatory bowel disease, particularly ulcerative proctitis, which affects up to 20% of patients. [108], [109], [110] and [111] The proctocolitis is usually relieved by a gluten-free diet; therapy with sulfasalazine (Salazopyrine), mesalamine, or corticosteroid enemas is also effective. The demonstration of an inflammatory infiltration of the rectal mucosa following a rectal gluten challenge suggests that the cause of proctocolitis may be similar to that of small intestinal enteropathy. [54], [55] Microscopic colitis, another chronic diarrheal syndrome, is characterized by mucosal inflammation of an endoscopically normal colon. Depending on whether or not a thickened subepithelial collagen band is present, two separate terms are used to describe the condition: *collagenous colitis* and *lymphocytic colitis*. A feature of the colonic inflammatory reaction is intraepithelial lymphocytosis, suggesting that an immunostimulating agent may be presented to mucosa-associated lymphoid tissue. Potential agents include dietary antigens. [52] Microscopic colitis can be diagnosed only by rectal biopsy, although a full colonoscopy should then be undertaken. [53] The therapeutic alternatives have been discussed by Zins and colleagues. [112]

Intestinal Pseudoobstruction

Symptoms of intestinal obstruction without any evidence of an organic lesion obstructing the lumen of the intestine rarely can be associated with celiac disease; if so, the symptoms are relieved by treatment with a gluten-free diet. [113]

Nonceliac Gluten Intolerance

This condition has been described in infants and children, usually in association with intolerance to cows' milk. [71], [114] The ingestion of gluten-containing cereals causes diarrhea, vomiting, and occasionally rhinitis and bronchitis. Other symptoms include vomiting, headaches, and recurrent oral aphthous ulceration. The disorder also affects adults, whose symptoms are usually abdominal pain and diarrhea. Investigation reveals a normal jejunal mucosa with a normal intraepithelial lymphocyte count and disaccharidase activity. Results of immunologic tests, including measurement of immunoglobulin and gliadin antibody levels, are normal. The cause of the intolerance is unknown, and treatment with sodium cromoglycate does not help. All patients experience dramatic relief with a gluten-free diet and relapse on challenge, and it is possible that they have a functional bowel disorder exacerbated by wheat starch. [115] A diet that omits cereals, particularly wheat, and replaces dietary fiber with ispaghula husks or ground psyllium seeds usually is effective.

TREATMENT

Celiac disease is treated with a gluten-free diet; products containing wheat, rye, barley, and traditionally oats are avoided. [3] The toxicity of oats is a matter of disagreement. [116] Several studies have indicated that moderate amounts (50 g/d) of oats are not harmful to adult patients with celiac disease, and one study has suggested that children with celiac disease can consume oats, although this must be confirmed. [117] Many commercially available sources of oat flour are contaminated with wheat flour because the mills are not scrupulously cleaned after previous milling of wheat. Oats should therefore be purchased from a dedicated miller.

Holmes and colleagues [9] showed that a gluten-free diet significantly decreases the incidence of T-cell lymphoma of the small intestine in patients with celiac disease.

For this reason, and the need to protect patients with celiac disease from dietary deficiency, even those with relatively mild symptoms should be advised to follow a strict gluten-free diet. The advice must be emphasized because some patients improve significantly on a partially gluten-free diet and may be tempted to take a rather relaxed approach to the gluten-free diet. A small percentage of patients become severely systemically ill with untreated or unresponsive celiac disease, in which case a short course of oral or parental steroids should be given.

Gluten-Free Diet

The gluten-free diet is based on the complete avoidance of wheat, rye, and barley, and products containing these grains. Triticale, which is a hybrid of wheat and rye, must be avoided, as must spelt or “ancient” or “wild” wheat. Attempts have been made to market spelt as suitable for individuals with gluten intolerance. Claims that this cereal is in some way different from wheat are totally spurious. Malted barley products, malt extract or flavoring, and beers and lagers of all types contain residual gluten peptides and must be excluded from the diet.

The distant cousins of wheat, such as maize, rice, and sorghum, contain storage proteins that bear little similarity to gluten, and these cereals can be safely consumed by patients with celiac disease. Buckwheat is a source of confusion; actually, it is a legume rather than a cereal and can thus be safely consumed by individuals with gluten sensitivity. Carbohydrate sources such as chick peas and potato flour are also free of gluten by nature. Thus, a number of flour substitutes that are naturally free of gluten have been used as substitutes for wheat. They do not, however, have the baking qualities of wheat. It is possible to produce a palatable cake or biscuit with, for example, maize flour. However, bread made without wheat is a good deal less successful. This has led manufacturers to use wheat starch, which has a better texture and taste. Despite improvements in manufacturing techniques, small amounts of gluten remain to contaminate the wheat starch, the significance of which is disputed. A study in Scandinavia [118] suggested that the regular use of wheat starch made according to the stringent standards of modern regulations is compatible with good health in patients with celiac disease. Some groups of patients, however, feel that any foods labeled as gluten-free should contain levels of gluten lower than those found in wheat starch. Some patients are exquisitely sensitive to tiny amounts of gluten and must follow a gluten-free, wheat-free diet that excludes wheat starch.

In addition to wheat starch, many other sources can contaminate foods that appear not to contain the protein gluten. Errors during the manufacturing, storage, and handling of foodstuffs can and do lead to the contamination of foods with gluten. [119] Additionally, incorrect or ambiguous food labeling creates further hazards. A number of organizations provide regularly updated information about the contents of manufactured foods. Useful contacts include the Celiac Sprue Association of the United States of America (csaceliacs.org), the Celiac Disease Foundations (celiac.org), the U.K. Coeliac Society (coeliac.co.uk), and a commercial database (www.brandbeach.com/celiac/upc/index.html).

A gluten-free diet contains little fiber, which may cause some constipation. Patients should be encouraged to eat good quantities of fruit and fibrous vegetables, possibly with the addition of regular dietary rice bran and ispaghula. Commercially available gluten-free products, which are available by prescription in many countries, may be useful supplements to a gluten-free diet, particularly for children. Any specific dietary deficiencies that occur, including deficiencies of iron, folic acid, calcium, and rarely vitamin B_{12}, should be corrected.

After 3 or 4 months of treatment with a gluten-free diet, biopsy of the small intestine should be repeated to demonstrate improvement in the appearance of the jejunal mucosal morphology. If improvement has not occurred, other possible causes of atrophy of the small intestinal villi, such as giardiasis, intolerance to cows’ milk, and hypogammaglobulinemia, should be considered and another biopsy performed after an additional 6 to 9 months.

If the patient’s symptoms persist or the jejunal specimen morphology remains grossly abnormal while the patient is on a gluten-free diet, commercial gluten-free products based on wheat starch should be discontinued and the initial diagnosis questioned. The completely wheat-free diet should be organized by a dietitian who is competent and experienced in the field. It should not be forgotten, however, that the most common reason for the lack of a response is either poor patient compliance or the inadvertent ingestion of gluten.

If a clear improvement in the jejunal specimen morphology with complete resolution of the histological abnormalities and symptoms does not occur, further confirmation can be made by a demonstrating a deterioration in the small intestinal morphology after a gluten challenge. Such confirmation is essential in children, in whom conditions such as infectious diarrhea and cows’ milk intolerance may cause similar abnormalities of small intestinal morphology. The most convenient method of gluten challenge is to ask the patient to ingest at least 10 g of gluten in the form of four slices of normal gluten bread each day for 4 to 6 weeks. If severe symptoms result, the date of the biopsy should be brought forward. The ESPGAN suggests that it is not mandatory to repeat the jejunal biopsy following a gluten challenge if a gluten-free diet has been associated with good relief of symptoms and significant improvement in the morphology of a repeated jejunal biopsy specimen. [71] Nearly half of all adult patients have an incomplete histological resolution despite marked symptomatic improvement. Many physicians do not routinely use gluten challenge in patients with celiac disease, reserving it for cases of diagnostic uncertainty.

Patients who do not adhere strictly to a gluten-free diet invariably experience chronic ill health and recurring symptoms, which usually can be traced to dietary lapses that are either deliberate or inadvertent. Difficulty may arise when a given food is thought to be gluten-free but in fact contains gluten, including those with low levels of contamination by wheat flour. An experienced dietitian may have to analyze the diet in detail. Frequent problem foods include malted breakfast cereals, snacks, sauces, and beer.

Rarely, failure to respond to a gluten-free diet is caused by the development of a small intestinal lymphoma or ulcerative jejunitis (see section “ Refractory Sprue”) or the presence of another concurrent disorder, such as chronic pancreatitis. Occasionally, the condition of a patient on treatment will deteriorate, and the patient may die unaccountably.

Pink and Creamer [120] reported that 70% of patients with celiac disease on a gluten-free diet quickly return to normal health, with improvement noted within 2 weeks. The remaining 30% of patients fall into three groups. Patients in the first group experience progressive deterioration, which is halted in some cases by corticosteroids but leads to death in others. Patients in the second group have an associated pancreatic lesion. Those in the third group do not adhere strictly to the diet, but even when this problem is addressed, their minor abdominal symptoms, including diarrhea, persist. [71] It is important to note that dietary failure is not always the cause of persistent symptoms.

REFRACTORY SPRUE

Lymphoma, Jejunal Ulceration, and Autoimmune Enteropathy

Enteropathy-associated T-cell lymphoma (EATL) may occur in patients with a long history of celiac disease, but it is often seen in adults with a short history of celiac disease, which suggests that it is a consequence of long-term ntreated disease. T-cell lymphoma often presents as a gluten-resistant enteropathy also known as *refractory sprue*. [121] Not all cases of refractory sprue progress to EATL.

A related complication of celiac disease is known as *ulcerative jejunitis,* which has clinical features similar to those of lymphoma. Jejunitis is characterized by multiple chronic, apparently benign ulcers that are found most frequently in the jejunum. Both jejunitis and EATL tend to affect persons in the fifth and sixth decades of life and present with fever, lassitude, anorexia, weight loss, abdominal pain, and diarrhea. [122] Anemia, both microcytic and macrocytic, is common, as is steatorrhea and a protein-losing enteropathy. The jejunal ulcers may be complicated by hemorrhage, perforation, or stricture formation, and emergency laparotomy is often required. [45] A flat jejunal mucosa on small intestinal histology is similar to that found in celiac disease. Patients may benefit from a gluten-free diet, but the response to such a diet is frequently suboptimal. The HLA genotype in the few reported cases is identical to that found in celiac disease and supports an association of these disorders. [123] The prognosis is poor, with a third of patients dying of complications, although it is considerably improved if the ulcerated or strictured segment is resected. Some patients with jejunitis progress to EATL, whereas multiple inflammatory mucosal ulcers frequently accompany EATL itself.

Refractory sprue—that is, the loss or lack of response to a gluten-free diet—may also progress to EATL. Both refractory sprue and ulcerative jejunitis are characterized by a nonlymphomatous monoclonal T-cell population in the affected mucosa. In EATL, a similar population that demonstrates clonal identity with the lymphoma itself is seen. Bagdi and colleagues [121] demonstrated that in all three cases, the monoclonal T-cell population consists of cytologically normal noninvasive intraepithelial T lymphocytes that share an identical aberrant immunophenotype with EATL. The authors concluded that patients with refractory sprue or ulcerative jejunitis have a neoplastic T-cell disorder. Others [124] found that a subgroup (16%) of individuals with refractory sprue who did not have aberrant clonal intraepithelial

lymphocytes made a complete recovery when treated with steroids in addition to a gluten-free diet.

Antienterocyte antibodies have been detected in a subgroup of patients with refractory sprue. [125] These antibodies are found in autoimmune enteropathy, a condition originally described in children and diagnosed by histological evidence of enteropathy in the presence of antienterocyte antibodies and the absence of an identified dietary protein trigger. Other organ-specific antibodies are common, whereas antiendomysial antibodies are not usually found. It is suggested that celiac-like enteropathy may be a manifestation of a multisystem autoimmune disorder. Routine searching for the presence of antienterocyte antibodies in all cases of refractory sprue is suggested because they may respond better to immunosuppressive therapy. [125]

Steroids

Celiac disease can be treated with systemic steroids, which induce a rapid cessation of diarrhea, weight gain, and improvement in fecal fat excretion and absorption, although these effects rarely persist once treatment has been stopped. Corticosteroids are not indicated in the routine management of celiac disease but are appropriate in the treatment of acute celiac crisis, manifested by severe diarrhea, dehydration, weight loss, acidosis, hypocalcemia, and hypoproteinemia. Steroids also can be used in the rare condition of gliadin or gluten shock that occurs in occasional treated patients who are subjected to gluten challenge. [126]

Azathioprine or 6-mercaptopurine can be used as a steroid-sparing agent if a dose of 10 mg of prednisolone or more per day is required to keep the condition under control. [127] The role of cyclosporine remains unproved. [128], [129]

The dose of steroids depends on the individual case. If intravenous fluid replacement is required because of vomiting, diarrhea, or surgery, 100 mg of hydrocortisone should be given intravenously every 6 hours. A patient who is eating normally but exhibiting a crisis should be given 40 to 60 mg of prednisolone daily. It should be possible within a few weeks to reduce the dose to that given for celiac disease that has not responded adequately to a gluten-free diet (i.e., 5–10 mg/d). Failure to be able to do so should alert the clinician to the possibility of either a failure to adhere to the diet or a complication, such as lymphoma or ulcerative jejunitis.

COMPLICATIONS

All patients with celiac disease exhibit lactose and sucrose intolerance at the time of diagnosis. Only a small percentage have a persistent disaccharidase deficiency following treatment with a gluten-free diet, and they experience abdominal pain and diarrhea with intake of lactose or sucrose. These conditions can be diagnosed either by enzyme assays of a small intestinal biopsy specimen or, more simply, by an appropriate sugar tolerance test. If concomitant disaccharidase deficiency is present, the relevant disaccharide should be excluded from the diet.

Some patients with treated celiac disease have bacterial overgrowth in the small intestine, which can be diagnosed by an abnormal result of a hydrogen or bile acid breath test or by an elevated urinary or small intestine bacterial count (see Chapter 78). Treatment is with an antibiotic, such as 250 mg of oxytetracycline four times daily, which can be rotated every 2 weeks with another antibiotic, such as ciprofloxacin.

Neurological Complications

Neurological complications, reviewed by Willis, [97] may occur in 10% of patients with celiac disease. [62], [63] These include disorders of the central nervous system, such as epilepsy, myoclonus, ataxia, and dementia, which frequently respond poorly to the exclusion of gluten from the diet. However, peripheral neuropathies of the axonal and demyelinating types may respond to a gluten-free diet. The neurological complications may be related to vitamin deficiencies or may be of immune origin. The suggestion that occult celiac disease may underlie a proportion of cases of neurological dysfunction of unknown cause has not been universally accepted. However, workers in the United Kingdom identified patients with “gluten ataxia,” characterized by the presence of antigliadin antibodies with or without small bowel histological abnormalities. [130] All had gait ataxia, and some had limb ataxia. A large number of the patients had some form of peripheral neuropathy. Some of them responded symptomatically to a gluten-free diet. Two patients who died showed evidence of lymphocytic infiltration of the cerebellum, damage to the posterior columns of the spinal chord, and sparse infiltration of the peripheral nerves. The literature contains a number of case reports. [97] The neuropathological findings have been heterogeneous, some suggesting an immune etiology and others showing only degenerative change.

Osteoporosis

Good evidence has been found of reduced bone mineral density in celiac disease. [131] In persons in whom the disease is not diagnosed until after bone mineral density peaks in the third decade, full mineralization may never be achieved, so that the risk for osteoporosis later in life is very high. Conversely, studies have shown that those who have been on a gluten-free diet since childhood have a normal bone mineral density. Osteoporosis in celiac disease is probably related to calcium malabsorption leading to parathyroid hormone secretion and resultant increases in bone turnover. Vitamin D malabsorption may coexist, requiring vitamin D therapy.

Some sources favor investigating the bone mineral density at diagnosis, although it is recognized that this policy might put undue strain on the densitometry services. [131] In this case, a compromise might be to test the bone mineral density at menopause in woman, with the test repeated 2 years later in those with a normal value at menopause. In men, testing at the age of 55 years is suggested.

The British Society of Gastroenterology has suggested a strategy for the prevention and treatment of this common complication of celiac disease. Patients should be counseled about the importance of a strict gluten-free diet. Adequate dietary calcium should be supplemented, if necessary, to ensure a daily intake of 1500 mg. The importance of exercise and moderate alcohol consumption should be stressed. Smoking is prohibited. Vitamin D deficiency should be sought and treated. Those with osteoporosis should be treated with bisphosphonate or calcitonin and hormone replacement therapy in postmenopausal women.

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CHAPTER 77

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DISORDERS OF EPITHELIAL TRANSPORT IN THE SMALL INTESTINE

The intestinal mucosa represents a semipermeable barrier that controls the flux of solutes and fluid into the systemic circulation. It is highly differentiated at the cellular level, and in particular, enterocytes acquire increasing functional capacity as they migrate along the crypt-villus axis. Maximal absorptive activity thus is found along the villus, characterized by the presence of an enzyme-rich brush border membrane [1], [2], [3] and [4] and the expression of mRNA for specific transporters. [5] Secretory function is concentrated at the base of the crypt. [6] Most defects of intestinal transport affect the absorption of a specific nutrient or a group of nutrients. Accordingly, the phenotypic expression varies significantly from none to life-threatening as the result of a wide range of intestinal and extraintestinal effects. Intestinal effects are usually secondary to the presence of excessive amounts of unabsorbed solutes in the lumen, which leads to acute fluid and ionic imbalances; in general, extraintestinal effects reflect the systemic deficiency of a specific nutrient and therefore may be more insidious and progressive. Transport is closely integrated with digestion [7]; therefore, this chapter focuses on specific mucosal transport defects and common hydrolytic enzyme deficiencies (primary or secondary) that may be phenotypically indistinguishable.

DISORDERS OF LACTOSE AND SUCROSE DIGESTION

The clinical symptoms associated with lactose or sucrose malabsorption customarily are ascribed to low intestinal levels of the microvillus membrane disaccharidases required for their hydrolysis: lactase–phlorizin hydrolase and sucrase-isomaltase, respectively. Such reduced levels may be a consequence of genetic alterations in expression of the enzymes or of reductions in enzyme activity as a result of intestinal injury. Depending on the severity of injury, nutritional complications may occur. The average adult ingests about 300 g of carbohydrate daily, 50% as calories, 17% as starch (mainly as cereals and potatoes), 25% as sucrose, 5% as lactose (mainly in milk), and 3% as fructose (in fruit and honey). Glycogen, glucose, and maltose are minor constituents, and cellulose accounts for about 4 g of carbohydrates per day. In contrast, lactose contributes 35% to 55% of the calories ingested daily by infants. As weaning foods are introduced, lactose intake decreases toward quantities commonly ingested by adults.

Lactose Intolerance

Biology and Genetics The human lactase gene, located on chromosome 2q22, comprises 17 exons and covers about 70 kilobases, giving rise to an mRNA of slightly more than 6 kilobases. From the initiation codon to the stop codon, human lactase mRNA encodes 1927 amino acids forming the complete translation product. [8] The nascent protein is heavily glycosylated, so that the final translation product is about 220 kd. [9] This high-molecular-mass glycoprotein undergoes two cleavage events and is then is inserted into the microvillus membrane of the enterocyte as a mature enzyme of approximately 160 kd. [10], [11] The cleaved intracellular portion of the precursor protein is degraded. [12] Analysis of the human lactase gene from subjects with high and low levels of lactase demonstrates interesting single-nucleotide polymorphisms, but their function in lactase expression remains to be delineated. [13], [14] The developmental pattern of expression of lactase in the human fetus is unique. Before week 24 of gestation, the level of intestinal lactase activity is low. It then begins to increase, and during the third trimester, lactase activity increases markedly until levels in term neonates are at or above those of infants ages 2 to 11 months. [15] Unlike the response of sucrase activity to sucrose feeding, [16] lactase activity cannot be increased by lactose feeding, and it is not reduced during periods of lactose elimination in humans. [17] Lactase exhibits a characteristic proximal-to-distal pattern of expression; enzyme activity is greatest in the midjejunum, with decreased activity proximally, especially in the duodenum, and distally, especially in the terminal ileum. [18] Lactase is expressed more distally on the villus, which is said to account for its sensitivity to villus flattening. [19] Within the enterocyte, lactase–phlorizin hydrolase mRNA is colocalized with its encoded protein in the apical enterocyte. [20] In most of the human population, lactase activity decreases during middle childhood (~5 years of age), and levels are low in adulthood. In contrast, in a minority of the human population, especially persons of northern European extraction and a few other racial groups, high levels of activity are retained throughout adult life (Table 77-1). [21] It is important to note that in all placental mammals, intestinal lactase activity is reduced to about 10% of the suckling level around the time of weaning. It is now clear that in all mammals, including humans, the primary mechanism of this decline in lactase activity is regulated by gene transcription, and lactase activity is strongly correlated with the intestinal content of lactase mRNA. There is some evidence that posttranscriptional factors may also be involved in the intestinal lactase decline. [22] A small number of Caucasian persons with a racial/genetic lactase decline have an abnormality of the intracellular processing of newly synthesized lactase protein. [17], [23]

TABLE 77-1 Distribution of Lactose Phenotypes in Selected Populations

Pathophysiology Lactose digestion is slower than that of sucrose, and hydrolysis has been considered the rate-limiting step for the overall process of lactose absorption. [24] Lactose is hydrolyzed to glucose and galactose on the microvillus membrane of the intestinal absorptive cells. Uptake of these monosaccharides is accomplished by the sodium-dependent glucose carrier SGLT1 (see below). When lactose is not absorbed by the small bowel, it passes rapidly into the colon as a consequence of the osmolality of the intralumenal disaccharide. In the colon, lactose is converted to short-chain fatty acids and gas (hydrogen, methane, carbon dioxide) by the bacterial flora, forming acetate, butyrate, and propionate. The short-chain fatty acids are absorbed by the colonic mucosa, and this route salvages malabsorbed lactose for energy utilization. This is the mechanism by which the newborn colon also salvages lactose, and the adult with low intestinal lactase activity may adapt to persistent lactose ingestion. [21] This fermentative process, which conserves nutritionally important carbohydrate, serves as the basis of the lactose breath hydrogen test (see below). Clinical symptoms in persons with lactose intolerance are quite variable. Important factors include the osmolality and fat content of the food

in which the sugar is ingested, the rate of gastric emptying, the individual sensitivity to intestinal distention produced by the osmotic load of unhydrolyzed lactose in the upper small bowel, the rate of intestinal transit, and the response of the colon to the carbohydrate load. The higher the osmolality of the gastric contents and the higher the fat content of the food containing the lactose, the slower the gastric emptying and the fewer the symptoms induced by the sugar.

Clinical Features When clinical symptoms are induced by the ingestion of lactose, the term *lactose intolerance* is applied. [21], [23], [24] Lactose intolerance may be caused by a primary reduction in lactase expression or by the secondary effects of a variety of mucosal diseases. Three clinical phenotypes are associated with primary reductions in lactase activity. Developmental lactase deficiency occurs in preterm infants as a result of relatively low intestinal lactase activity before 32 weeks' gestation. A second phenotype, human congenital lactase deficiency, is an extraordinarily rare condition. It is inherited in an autosomal recessive fashion and results in the complete absence of active lactase enzyme. This condition has been recognized in isolated populations, as in Finland, where most cases have been reported. The locus for congenital lactase deficiency has been linked to a 350-kilobase interval more than 2 megabases away from the lactase–phlorizin hydrolase gene. [25] The most common form of lactose intolerance appears in the majority of the world's population, in which lactase activity declines at about the age of 5 years; thereafter, only low lactase levels persist (see Table 77-1). In Western countries, where patients may be immigrants from developing areas of the world, physicians should be aware of the prevalence and racial distribution of genetically controlled low levels of lactase activity. The occurrence of low lactase levels in patients older than 5 years is most prominent in Asian, African, and all indigenous populations. Most infants and children up to the age of 5 years in every racial and genetic group have preserved lactase levels. Population genetic analysis indicates that a persistently high level of lactase activity in adult life is inherited as a single autosomal dominant gene, whereas a low level of lactase activity is inherited as a recessive trait. Secondary lactase deficiency is the result of mucosal injury. Diseases that can cause mucosal damage or villus flattening include infectious gastroenteritis (rotavirus is the most common cause), parasitic infections (giardiasis), celiac disease, tropical sprue, radiation enteritis, drug-induced enteritis, and Crohn's disease involving the proximal small intestine. The literature is unclear about the prevalence of lactase deficiency in Crohn's disease involving other parts of the small intestine. In one study, the intestinal lactase activity of patients with Crohn's disease reflected their genetic background. [26] In contrast, in another study, patients with Crohn's disease had a higher prevalence of lactose malabsorption than would have been expected for their ethnic group and in comparison to patients with ulcerative colitis. [27] These dichotomous findings remain to reconciled. In many secondary disorders, mucosal damage may be focal or patchy, so that a diagnosis of lactase deficiency by lactase assay in biopsy samples is difficult because lactase levels may vary in different samples. Lactose intolerance is characterized by abdominal pain, cramps or distention, nausea, flatulence, diarrhea, or vomiting. The abdominal pain may be cramping and may develop in the periumbilical region or the lower quadrant. Borborygmi may be audible on physical examination and to the patient. Lactose intolerance generally causes abnormal stools, which are usually bulky, frothy, and watery. In severe cases, mostly in infants, acidosis and dehydration may be a problem. Vomiting after lactose ingestion often is seen in adolescent patients. Different persons appear to be more or less sensitive to the ingestion of lactose and exhibit variable degrees of abdominal distention and variable complaints when ingested lactose stimulates an influx of water into the lumen of the small intestine or when the production of gas distends the colon. Persons with greater tolerance report fewer symptoms. These subjective responses are difficult to quantify. On careful testing, most lactose-intolerant adults who are offered alternatively lactose- and glucose-containing milk experience little difference in clinical symptoms despite differences in gas production. [28], [29] In fact, women proven to be lactose maldigesters were able to tolerate a lactose-containing diet that provided 1300 mg of calcium per day without experiencing significant symptoms, such as bloating, abdominal pain, or diarrhea. [30] Thus, subjective contributions to the clinical symptoms of lactose intolerance are important. Patients with irritable bowel syndrome who are also lactose intolerant may experience increased pain after lactose ingestion. Intestinal transit also is influenced by the quality of the diet and individual motility patterns. Accordingly, some lactose-intolerant persons have a rapid movement of sugar to the cecum, whereas others have slower motility. Fecal flora adapt to ingested carbohydrates. In many intolerant people, therefore, if lactose is provided slowly over a long period, the flora may adapt to the load and the symptoms caused by gas and acid in the colon may be reduced or eliminated. The breath hydrogen excretion and symptoms of African American adolescents who were lactose maldigesters significantly decreased when they were fed a lactose-containing diet over 21 days, presumably because of colonic flora adaptation. [31] This mechanism of lactose tolerance in persons with low lactase levels accounts for the discrepancy between lactase malabsorption, defined by lactose breath hydrogen testing, and clinical symptoms of lactose intolerance. Human adult-onset lactase decline has been correlated with osteoporosis, which can lead to bone fragility and an increased risk for bone fracture. [23] Women with osteoporosis had a significantly higher prevalence of lactose malabsorption than age-matched controls of similar ethnic origin. [32] Lactose avoidance can also lead to bone demineralization in children. In an evaluation of children whose intake of lactose was restricted because of various disorders (genetic lactose intolerance, short bowel syndrome, mild protein allergy, hypercholesterolemia), 50% of the patients had signs of osteoporosis or osteopenia based on bone mineral density measurement. [33] Adequate supplementation of calcium intake is necessary in people with lactose intolerance for the long-term maintenance of bone health.

Diagnosis The confirmation of lactose intolerance depends on reproducing symptoms when lactose is ingested. Alternatively, when the diagnosis of lactose intolerance is strongly suspected, resolution of symptoms after the temporary elimination of lactose from the diet serves as a diagnostic test. The diagnosis of lactose malabsorption is based on the results of appropriate tests that reflect or confirm the inability to absorb lactose. A low fecal pH or the presence of reducing substances in stools indicates lactose malabsorption, but the results of these tests are valid only when lactose has been ingested, intestinal transit time is rapid, stools are collected fresh and assays performed immediately, and bacterial metabolism of colonic lactose is incomplete. In general, lactose malabsorption is best confirmed with a more specific test. The capacity for lactose absorption can be measured with a lactose absorption test. In adults, it has a sensitivity of 75% and a specificity of 96%. In children, however, it is cumbersome, invasive, and time-consuming and has been replaced largely by the lactose breath hydrogen test. [24], [28], [29] The breath hydrogen test really measures lactose nonabsorption rather than lactose hydrolysis and monosaccharide uptake. Its sensitivity and specificity are superior to those of the absorption test, and it is simple and noninvasive. The lactose hydrogen breath test has become the most popular and reliable method for the diagnosis of lactose malabsorption. The test can be performed in patients of all ages. The administered dose is customarily 2 g of lactose in water per kilogram of body weight, not to exceed 50 g. Breath is sampled before lactose is ingested and at 30-minute intervals thereafter for 3 hours and is analyzed for hydrogen content with readily available desktop or portable devices. It is customary to consider a hydrogen value of 10 parts per million as normal, with samples obtained after lactose ingestion compared to the baseline value. Hydrogen values between 10 and 20 parts per million may be indeterminate unless accompanied by symptoms, but hydrogen values higher than 20 parts per million represent lactose malabsorption. False-positive results are caused by inadequate pretest fasting or recent smoking. False-negative results are obtained when patients have recently used antibiotics or are not hydrogen producers (~1% of the population). In children younger than 5 years of age, an abnormal lactose breath hydrogen test result signifies intestinal mucosal injury or bacterial overgrowth, both of which require further definition by appropriate diagnostic tests. A normal breath hydrogen test result does not rule out an intestinal mucosal lesion, and it cannot be used to avoid an intestinal biopsy. The assay of disaccharidase activity in small bowel biopsy samples establishes the presence of disaccharidase deficiency and has been used to define populations at risk for low lactase levels. [21] Although a low level of lactase activity accompanies intestinal injury, the lesion may be focal or patchy; consequently, intestinal biopsy samples may not yield an abnormal result. Clinical and biochemical data always must be compared to obtain the correct diagnosis.

Treatment The treatment of lactose intolerance is based on four general principles (Table 77-2 and Table 77-3): reduction or restriction of dietary lactose, substitution of alternative nutrient sources to avoid reductions in energy and protein intake, regulation of calcium intake, and the use of a commercially available enzyme substitute. When lactose restriction is necessary, patients must be instructed to read labels of commercially prepared foods to identify hidden lactose (see Table 77-2). Complete restriction of lactose-containing foods should be necessary for a limited period only to ascertain the specificity of the diagnosis. Because many patients can tolerate graded increases in lactose intake, small quantities of lactose should be reintroduced subsequently into the diet, with careful attention paid to associated symptoms. Because of its high sugar and fat content, ice cream may be a good way to introduce lactose into the diet. The diet should be reviewed with the patient to ensure that protein, fat, and other nutrients are supplied at appropriate levels. Calcium is supplemented in the form of calcium carbonate; Tums is a popular and effective product. Standard preparations contain 500 mg of calcium carbonate, equivalent to 200 mg of elemental calcium, which is 20% of the U.S. Recommended Dietary Allowance for adults. For infants and young children, liquid calcium gluconate is readily tolerated and available. When complete lactose restriction is recommended, the U.S. Dietary Reference Intake (DRI) for calcium should be provided as a supplement. Commercially available lactase preparations (see Table 77-3) are actually bacterial or yeast ß-galactosidases. When added to lactose-containing foods, or when ingested with meals containing lactose, they effectively reduce symptoms and breath hydrogen values in many lactose-intolerant subjects. These products cannot completely hydrolyze all dietary lactose, however, and the results achieved in individual patients are variable. The commercial lactase preparations can be added to foods, ingested with foods, or sprinkled on food products. Commercially available Lactaid milk comes in 70% and 100% hydrolyzed forms. It should be noted that acidophilus milk is not sufficiently depleted of lactose. Live-culture yogurt, which contains endogenous ß-galactosidase, is a useful alternative source of calcium and calories and should be well tolerated by a number of lactose-intolerant patients. [34] Types of yogurt in which milk or milk products are added back after fermentation may cause symptoms. Whereas the consumption of yogurt alone by persons with low levels of lactose tolerance reduces symptoms, the consumption of yogurt together with additional lactose does not reduce symptoms. The capacity of probiotics to provide lactose digestion in individuals with low lactase levels appears to vary. For example, milk treated with *Lactobacillus bulgaricus* has been shown to decrease both breath hydrogen excretion and clinical symptoms of lactose intolerance in lactose maldigesters. [35] However, *Lactobacillus acidophilus* has not affected these markers significantly. [35], [36]

PRODUCT	UNIT	LACTOSE (APPROXIMATE g/UNIT)
Milk	1 cup (244 g)	11
Low-fat milk, 2% fat	1 cup (244 g)	9–13
Skim milk	1 cup (244 g)	12–14
Nonfat dry milk, instant	1⅓ cup (91 g)	46
Whipped cream topping	1 tbs (3 g)	0.4
Light cream	1 tbs (15 g)	0.6
Cheese		
Cheddar	1 oz (28 g)	0.4–0.6
Cream	1 oz (28 g)	0.8
Parmesan, grated	1 oz (28 g)	0.8
American	1 oz (28 g)	0.5
Swiss	1 oz (28 g)	0.4–0.6
Cottage	1 cup (210 g)	5–6
Ice Cream		
Vanilla, regular	1 cup (133 g)	9
Sherbert, orange	1 cup (193 g)	4
Ice, orange	100 g	0

From Bayless T, ed. Current therapy in gastroenterology and liver disease, 4th ed. St Louis: Mosby-Year Book, 1994:303.

TABLE 77-2 Lactose Content of Selected Foods

NAME	DOSE FORM	SUPPLIER
Lactaid	Liquid/tablets	Lactaid
Lactrase	Capsules	Kremers Urban
LactAce	Capsules	Nature's Way Products
DairyEase	Tablets	Glenbrook Laboratories
Lactrol	Caplets	Advanced Nutritional Technology

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TABLE 77-3 Some Commercial "Lactase" Substitutes

Sucrase-Isomaltase Deficiency

This rather rare disorder is the consequence of faulty synthesis or processing of the intestinal enzyme sucrase-isomaltase. [17] Typically, patients present in infancy, although sometimes the diagnosis is not made until childhood or, occasionally, adolescence or adulthood. [37]

Biology and Genetics The sucrase-isomaltase gene is located on chromosome 3, and 12.2 kilobases of its promoter have been cloned. The polypeptide sequence deduced from the human cDNA consists of 1827 amino acids, forming the complete translation product. The final form of the protein inserted into the microvillus membrane is heavily glycosylated, such that the molecular mass is about 270 kd. Once it is inserted into the microvillus membrane, the 270-kd form is cleaved by pancreatic proteases into its mature form, an enzyme complex in which sucrase (~120 kd) is noncovalently linked to isomaltase (~140 kd) inserted into the microvillus membrane by a hydrophobic anchor sequence. The development of expression of sucrase-isomaltase in the human fetus differs from that of lactase. Even in the youngest fetuses studied (10 weeks), the activity of sucrase-isomaltase is similar to that in the adult human intestine. Sucrase-isomaltase activity remains unchanged throughout development. [15] The activity of sucrase-isomaltase, however, is responsive to dietary carbohydrate and increases after prolonged fructose and sucrose feeding. [16] Sucrase exhibits a proximal-to-distal expression pattern similar to that of lactase, but with relatively more activity in the duodenum and ileum than is found for lactase. On the vertical axis, sucrase is expressed at the lower villus; in contrast, lactase is expressed at the upper villus. [19] Within the enterocyte, sucrase mRNA is colocalized with encoded protein in the apical side of the cell. [20] Sucrase-isomaltase deficiency is an autosomal recessive disorder characterized by undetectable levels of intestinal sucrase activity and reduced isomaltase activity. Although intestinal histology may show minor degrees of villus atrophy with associated malnutrition, [38] most patients with sucrase-isomaltase deficiency have normal villus architecture. The prevalence of sucrase-isomaltase deficiency is about 0.8% in North Americans and about 10% in the Eskimos of Greenland. The biochemical defects identified indicate that the deficiency state arises from varying allelic mutations of the sucrase-isomaltase gene. [17], [39] At least six different defects have been identified: arrest of prosucrase-isomaltase in the rough endoplasmic reticulum; arrest and degradation of prosucrase-isomaltase in the Golgi apparatus; catalytically altered enzyme in the microvillus membrane and partial missorting to the basolateral membrane; missorting to the basolateral membrane and accumulation in the rough endoplasmic reticulum; intracellular loss of the sucrase subunit and expression of the isomaltase subunit at the microvillus membrane; cleavage and secretion of the mutant sucrase and absent immunoreactive protein. [17], [40] Thus, most genetic defects seem to reflect mutations in the gene that lead to changes in the primary amino acid sequence of sucrase-isomaltase. [39] The resulting mutant proteins display a broad variety of mechanisms of aberrant intracellular sorting or premature degradation.

Clinical Features Sucrase-isomaltase deficiency presents at different ages. [38], [41], [42] In infancy, symptoms may occur when sucrose is introduced into the diet: diarrhea, secondary malabsorption, and failure to thrive. In young children, chronic diarrhea may occur immediately or months after the introduction of sucrose. Because sucrose is not the primary carbohydrate in most infant formulas, malnutrition is less commonly associated with sucrase-isomaltase deficiency. Occasionally, the first manifestations appear in adolescence or adulthood after a gastroenteritis with symptoms of diarrhea, abdominal cramps, and flatulence that are intermittent in nature and mimic the irritable bowel syndrome. The stools of patients with sucrase-isomaltase deficiency often contain nonhydrolyzed sucrose; however, a special technique is needed to hydrolyze the disaccharide before testing for reducing substances because sucrose is not a reducing sugar. The diagnosis of sucrase-isomaltase deficiency is achieved most easily by means of the sucrose breath hydrogen test, which is performed in exactly the same manner as the lactose breath hydrogen test. However, a false-positive rate of 20% to 30% in some studies may suggest the need for confirmation of the diagnosis by small intestinal biopsy with disaccharidase determination. [38]

Treatment The primary approach to the treatment of sucrase-isomaltase deficiency is the elimination or restriction of dietary sucrose, which leads rapidly to resolution of the clinical symptoms. Over time, small quantities of sucrose can be introduced into the diet because colonic fermentation of ingested sucrose does occur (see section " Lactose Intolerance"). The induction of sucrase-isomaltase activity by means of dietary carbohydrate was reported in one patient, whose enzyme activity increased after a diet high in fructose. [15] This approach has not been used widely. Furthermore, depending on the underlying basic defect, substrate induction of enzyme activity would not be expected to occur. A new approach to the treatment of sucrase-isomaltase deficiency involves the use of enzyme supplements. [43] Exogenous enzyme can compensate to produce adequate sucrose hydrolysis. A randomized, double-blinded, placebo-controlled trial demonstrated lower sucrose breath hydrogen values, decreased stools, and resolution of symptoms after affected patients with sucrase-isomaltase deficiency were treated with sacrosidase (a liquid produced from *Saccharomyces cerevisiae*). [44]

DEFECTS IN INTESTINAL CARBOHYDRATE TRANSPORT

D-Glucose, D-galactose, and D-fructose are the end products of lumenal and membrane digestion of ingested carbohydrates. These two processes occur through the activity of various enzymes that are either secreted into the lumen or inserted into the apical membrane of villus enterocytes as integral proteins. In humans, the major route of entry of hexoses into enterocytes is by brush border membrane carrier systems (Fig. 77-1). Whereas fructose is transported by facilitated diffusion by GLUT5, [45] glucose and galactose influx occurs through an apical Na^+-coupled glucose cotransporter (SGLT1), which is the only Na^+-coupled glucose transporter expressed in enterocytes. Differential expression of the *SGLT1* gene may occur along the crypt-villus axis. [46] This mechanism of coupled transport is an example of secondary active transport. [47] At the basolateral membrane, the two hexoses are transferred from the enterocyte into the circulation down their concentration gradient through a facilitated glucose transporter (GLUT2) that bears no homology with the SGLT family. [48] This basolateral carrier is a member of the gene family (GLUT) encoding the facultative glucose transporters present in erythrocytes, hepatocytes, myocytes, and adipocytes. [49], [50]

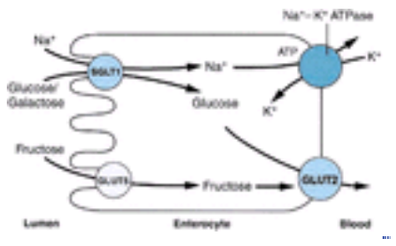

FIGURE 77-1. A model of sugar transport across the enterocyte showing the apical SGLT1 and GLUT5 transporters and the basolateral Na^+/K^+ pumps and sugar transporter GLUT2. (Adapted from ref. [64].)

SGLT1 is a Na^+-dependent cotransporter with a coding sequence of 15 exons distributed along 72 kilobases of DNA localized to chromosome 22q13.1. [51], [52], [53] and [54] The primary protein carrier is a glycosylated integral brush border membrane protein (84 kd) with 14 transmembrane-spanning domains. [55] Specific antibodies raised against extramembranous domains of the rabbit *SGLT1* sequence as well as cDNA probes have been used to identify this protein in a large number of species, indicating that the *SGLT1* DNA and protein sequence are highly conserved throughout evolution. [56], [57] and [58] SGLT1 has been shown to function in the cotransport of 2 Na^+, 1 glucose or galactose, and 200 to 400 water molecules along with urea and glycerol. [59] Through mutational analysis, it has been determined that hexose binding and translocation occur on the C-terminal domain of the protein, whereas Na^+ binding and translocation occur on the N-terminal domain. Expression of *SGLT1* is regulated by both transcriptional and posttranscriptional events. [57], [60]

Glucose-Galactose Malabsorption

The phenotype associated with glucose-galactose malabsorption was first described in the late 1960s. [61] The authors noted an association with consanguineous mating, although 30% of patients with glucose-galactose malabsorption are the result of nonconsanguineous relationships. The disorder is inherited as a autosomal recessive trait and remains rare, with only 200 patients reported worldwide. Glucose-galactose malabsorption is caused by an inability of SGLT1 to absorb hexoses, whereas the digestion of lactose, sucrose, and maltose and the absorption of fructose and xylose occur normally. [46]

Genetics A single missense mutation in the *SGLT1* gene (at position 92) was identified in two sisters carrying the glucose-galactose malabsorption phenotype. [62] Both parents were heterozygotes for the base substitution, pointing to the autosomal recessive mode of inheritance. The protein product revealed an asparagine residue at position 28 instead of aspartate. This report was the first of a genetic disorder correlated with a membrane transport protein mutation, and it definitively established SGLT1 as the principal mediator involved in intestinal glucose transport. More than 20 different types of missense, nonsense, and splice site mutations of *SGLT1* have subsequently been identified in patients with glucose-galactose malabsorption. Most mutations cause either truncated proteins or defects in trafficking between the endoplasmic reticulum to the Golgi complex to the plasma membrane of the enterocyte. [54], [55], [63], [64] and [65] The geographic distribution of reported cases includes regions as varied as Europe (Sweden), North America, Japan, Iraq, Lebanon, Morocco, Syria, Saudi Arabia, and Bangladesh.
Clinical Features Typically, infants carrying the glucose-galactose malabsorption genotype present during the first week of life with profuse watery diarrhea and hyperosmolar dehydration, failure to thrive, abdominal distention, and vomiting. Stool analysis reveals the presence of reducing substances and a low pH. Renal glycosuria has been reported in the absence of hyperglycemia. Symptoms resolve as soon as dietary glucose and galactose are withdrawn and a fructose-based formula is instituted. Cases of glucose-galactose malabsorption diagnosed in adulthood have been reported. [66], [67] The growth and development of children will be normal if symptoms are controlled, demonstrating a direct response to appropriate dietary management and individual tolerance. The most reliable diagnostic test for glucose-galactose malabsorption is the hydrogen breath test. [64] The presence of reducing substances in neonatal diarrheal stools without another cause strongly suggests glucose-galactose malabsorption. If the elimination of dietary glucose and galactose is not accompanied by resolution of symptoms and normal weight gain, however, other causes of carbohydrate malabsorption must be ruled out.
Pathophysiology Glucose-galactose malabsorption is characterized by gastrointestinal losses of electrolytes and fluids. Unabsorbed carbohydrates contribute to an increase in osmotic pressure in the intestinal lumen, with secondary fluid secretion. [68] The transit time is reduced as a result of the increased flow rate. Diarrhea develops because the osmotic load and increased flow rate exceed the colonic capacity for water reabsorption. [69] The fecal osmolality may be reduced in the colon as the colonic flora converts malabsorbed carbohydrates to short-chain fatty acids. [70], [71] These are in turn oxidized by bacteria or absorbed, allowing for some caloric salvage and decrease in the osmotic load as transit progresses distally. [68], [72] In contrast to what occurs in secretory diarrhea (see below), the contribution of electrolytes to stool osmolality is minimal, as revealed by a high osmolal gap. Also, the adaptive changes observed in the colon may explain the variable degree of carbohydrate tolerance observed in adults with glucose-galactose malabsorption. The absorption patterns of these patients are consistent with the selective malabsorption of glucose and galactose in the small intestine secondary to the genetically defective transporter SGLT1.
Treatment As outlined above, the therapeutic response to a fructose-containing formula establishes the diagnosis and subsequent management. It is important to note that oral rehydration solutions, such as Pedialyte, must be avoided during the treatment of acute dehydration because they contain glucose. Galactomine 19 (Nutricia, Zoetermeer, Holland), which is free of glucose and galactose, may be used in the first few months of life; alternatives are the carbohydrate-free formula RCF (Ross Laboratories, Columbus, Ohio) and Product 3232A (Mead Johnson, Evansville, Indiana). Fructose must be added to the formula to increase the calories by 3 kcal/oz per day to an end point of 20 kcal/oz to ensure gastrointestinal tolerance. [73] Compliance usually decreases with age, but gastrointestinal tolerance increases with the introduction of meats, eggs, and fats into the diet. The dietary load of glucose and galactose tolerated symptomatically varies from individual to individual, but malabsorption of both sugars is lifelong. Patients should avoid foods in which the disaccharide or glucose content represents more than 45% of the total carbohydrate, and alternative sweeteners (i.e., honey) should be used. [73] New foods should be introduced every 5 to 7 days to ensure tolerance in children. In most cases, growth and development are normal during childhood, [74] as is adulthood. [66], [67]

DISORDERS OF AMINO ACID ABSORPTION

Amino acid transport defects were described as early as 1889. Epidemiologic studies based on the screening of newborns have shown that the prevalence varies greatly, depending on the populations studied. Cystinuria, in particular, is one of the most commonly inherited disorders; its overall prevalence has been estimated at 1 in 7000 (ranging from 1 in 100,000 in Sweden to 1 in 2000 in England). [75] Most of these defects affect both the renal and intestinal epithelia, but concomitant phenotypic expression may not be seen.

L-Isomers of amino acids, like glucose, are normally absorbed at the brush border membrane through carrier-mediated mechanisms. Accordingly, the influx of peptides into enterocytes is a Na^+-dependent secondary active transport mechanism that carries either single amino acids or oligopeptides (dipeptides, tripeptides). Peptide efflux into the systemic circulation occurs through distinct carrier systems at the basolateral membrane. The hallmark of intestinal and renal amino acid transporters is their multiplicity and overlapping specificities. Certain L-amino acids compete for one specific carrier system. Conversely, neutral amino acids are transported by four different processes with variable specificity.

The clinical relevance of the complexity and interaction of these carrier systems becomes apparent in disease states. It should be emphasized that the final phenotypic expression of each of these defects is largely variable (Table 77-4). The disparity between genotype (identified by urinary excretion patterns and intestinal malabsorption) and clinical phenotype (overt symptoms) has been noted often, particularly since the introduction of newborn screening. Clinical protein malnutrition and essential amino acid deficiency are rare in most of these defects.

DISORDER	SUBSTRATES	CLINICAL ASSOCIATIONS
Hartnup disease	Neutral amino acids	Pellagra-like rash Neuropsychiatric symptoms
Cystinuria	Dibasic amino acids Cystine	Renal calculi Chronic pancreatitis
Lysinuric protein intolerance	Dibasic amino acids	Failure to thrive Growth retardation Hepatosplenomegaly Hyperammonemia
Blue diaper syndrome	Tryptophan	Bluish, discolored diapers Hypercalcemia Nephrocalcinosis
Oasthouse urine disease	Methionine	Mental retardation Seizures
Lowe syndrome	Lysine Arginine	Mental retardation Cataracts Renal failure
Joseph syndrome (iminoglycinuria)	Glycine Proline Hydroxyproline	Aminoaciduria

Data from Freeman HJ, Sleisinger MH, Kim YS. Human protein digestion and absorption: normal mechanisms and protein-energy malnutrition. Clin Gastroenterol 1983;12:357, and Silk DBA. Disorders of nitrogen absorption. Clin Gastroenterol 1982;11:47.

TABLE 77-4 Disorders of Intestinal Amino Acid Transport

Cystinuria

In classical cystinuria (dibasic aminoaciduria), the renal and intestinal lumenal reabsorption of cystine and cationic amino acids (lysine, arginine, ornithine) is decreased. Cystinuria is inherited in an autosomal recessive fashion. The three types of cystinuria are classified according to the amount of cystine present in the urine and plasma after oral loading. [76] In 1993, the gene encoding the transporter for cystine and dibasic amino acids was identified on chromosome 2p21 and was designated *SLC3A1.* Patients with type I cystinuria had various mutations in this gene sequence. [75] The type III cystinuria gene defect has been linked to chromosome 19q13.1. [77] The gene *SLCA9* encodes a b $^{o,+}$ amino acid transporter that is expressed in the kidney, liver, and small intestine. utations have been found in this gene in patients with non–type I cystinuria. [76] The diagnosis is made in patients who excrete more than 250 mg of cystine per gram of creatinine in 24 hours. Urolithiasis is the only clinical manifestation resulting from the excretion of poorly soluble cystine and accounts for 10% of cases of nephrolithiasis in children. The treatment of cystinuria consists of managing stone disease, preventing stone formation with hydration, ingesting sodium bicarbonate to maintain the urinary pH at about 7.5, limiting dietary sodium and methionine, and using sulfhydryl reagents to solubilize cystine. The recognition of several genetic subtypes led to the conclusion that cystinuria is caused by allelic mutations with a complex mode of inheritance. [76]

Lysinuric Protein Intolerance

Lysinuric protein intolerance (familial protein intolerance or hyperdibasic aminoaciduria type II) results from a transport defect at the basolateral membrane of the enterocyte, renal tubular cell, fibroblast, and probably also the hepatocyte. The mode of inheritance is autosomal recessive, and the prevalence is higher in Finland and southern Italy. [78], [79] The influx of peptides into enterocytes is normal, but the transcellular flux is impaired at the level of amino acid efflux into the systemic circulation. The defect affects cationic amino acids (lysine, arginine, ornithine). The diagnosis is established by the detection of high rates of urinary excretion and low plasma concentrations of these amino acids, which accumulate within the enterocyte. The gene causing lysinuric protein intolerance was assigned to chromosome 14q11.2 after linkage analysis of affected patients. [80] Mutational analysis, along with functional oocyte expression models, revealed mutations in the gene *SLC7A7,* which encodes a y+L amino acid transporter-1. [81] These studies have made possible molecular diagnosis, genetic counseling, and prenatal diagnosis. The clinical signs of lysinuric protein intolerance consist of failure to thrive, vomiting, growth retardation, and muscular hypotonia. Hyperammonemia following a protein-containing meal contributes to postprandial nausea and vomiting; secondary protein aversion further contributes to systemic protein deficiency. The pathophysiology of hyperammonemia is incompletely understood, but data point to a defect in the intramitochondrial transport of ornithine. [82] Because patients may limit their protein intake spontaneously, they may remain phenotypically silent. It is worth noting that breast-fed infants typically present later, during weaning or the introduction of other foods, because human milk is relatively low in protein in comparison with commercial formulas. Progressive hepatosplenomegaly and osteoporosis are other prominent features. Late complications, which at times may be the only manifestation of this defect, include alveolar proteinosis and glomerulotubular disease leading to renal failure. Treatment includes a low-protein diet to prevent hyperammonemia (children, 1–1.5 g/kg daily; adults, 0.5–0.7 g/kg daily) but may not always ensure adequate caloric intake, particularly in children. Citrulline supplementation (2.5–8.5 g) normalizes the urea cycle by restoring its pool of intermediates.

Hartnup Disease

Hartnup disease (neutral aminoaciduria and indolic aciduria) is an autosomal recessive disorder. The phenotypic expression results from intestinal and renal wasting of tryptophan. [83] Most subjects in whom neutral hyperaminoaciduria is identified are phenotypically normal; however, clinical symptoms such as a pellagra-like rash, cerebellar ataxia, mental retardation, and psychotic behavior have been reported. [84] The symptoms are caused by an absence of nicotinamide, the metabolized product of tryptophan. The dissociation between genotype and phenotype most likely is a consequence of superimposed polygenic and environmental factors. The diagnosis is established by the detection of an increased amount of urinary neutral amino acids and indican in the absence of intestinal bacterial overgrowth. Treatment with nicotinamide (50–150 mg daily) relieves the skin lesions and any neurological manifestations that are present. A protein-enriched diet also is recommended. [85]

DISORDERS OF ELECTROLYTE AND MINERAL TRANSPORT

See also Chapter 14.

Specific intestinal defects affecting ionic transport have been described for copper (Menkes disease), chloride (congenital chloride diarrhea), and sodium (congenital sodium diarrhea). The two latter present invariably with significant gastrointestinal symptoms. Other selective deficiencies of minerals such as magnesium (familial hypomagnesemia) [86] and zinc (acrodermatitis enteropathica) [87] have been reported as recessively inherited disorders. Sandstrom and associates [88] suggested a cellular defect of zinc metabolism rather than impaired absorption in a patient with acrodermatitis enteropathica. Gastrointestinal symptoms may be present, although extraintestinal signs usually prevail. With the exception of Menkes disease, these deficiencies can be corrected with oral supplementation, which suggests that distinct uptake processes may occur in the intestine.

The pathophysiology of cystic fibrosis is linked to abnormal cyclic AMP–dependent chloride conductance in the apical chloride channel, which leads to disturbances of mucus and electrolyte secretion in the pulmonary and gastrointestinal tract. The electrolyte defect and the clinical features are discussed in Chapter 98.

Menkes (Steely Hair) Disease

Copper is an essential nutrient for eukaryotes and prokaryotes, [89], [90] being an integral component of a multitude of key enzymes. [91] It is thus not surprising that a primary defect of copper transport, such as occurs in Menkes disease, leads to a fatal clinical picture involving a multitude of organs if copper is not provided. [92] As the molecular genetics of Menkes disease have been increasingly refined, however, it has become clear that the same gene is involved in clinically milder forms, such as occipital horn syndrome.

Genetics Menkes disease is a genetically transmitted, X-linked disorder that is the result of defective copper homeostasis. The incidence of Menkes disease has been estimated at between 1 in 100,000 and 1 in 250,000, with an estimated 15 to 35 patients affected in the United States per year. [93] The gene for Menkes disease was isolated in 1993 by positional cloning within Xq13 in an affected female subject bearing a balanced t(X;2) translocation. [94], [95] and [96] The gene product was found to be a transmembrane P-type adenosine triphosphatase (ATPase7A). [95] More than 150 different mutations that lead to Menkes disease have been identified in the ATPase7A gene, and it appears that distinct splice sites underlie the various phenotypes, which range from the lethal form to a milder form called *occipital horn syndrome*. [97], [98]

Clinical Features The clinical features of Menkes disease are summarized in Table 77-5. Affected patients are usually in good health for the first 8 to 10 weeks of life. At 2 to 3 months of age, typical signs include loss of previously acquired milestones, hypotonia, and seizures, along with the scalp hair characteristics. Of note, affected infants may present with vomiting, diarrhea, and failure to thrive, with or without a protein-losing enteropathy. The long-term prognosis is poor because

cerebral degeneration persists despite copper supplementation; death usually occurs during childhood.

	NEONATAL PERIOD	INFANCY TO EARLY CHILDHOOD
Prematurity	+	
Hypothermia	+	+/-
Hyperbilirubinemia	+	
Growth retardation		+
Abnormal facies	+/-	
Depigmentation (skin, hair)		+
Trichorrhexis, monilethrix	+/-	+
Pili torti		+
Developmental delay/neurodegeneration (seizures, ataxia)		+
Vascular complications		+
Osteoporosis		+
Death		+

TABLE 77-5 Clinical Features of Menkes Disease According to Age

The diagnosis is clinical, although confirmation can be sought by measuring serum levels of copper and ceruloplasmin. During the newborn period, a plasma catecholamine analysis and measurement of the placental copper concentration are more reliable tests to make the diagnosis. [93] Copper in the intestinal mucosa and liver and in cell cultures should be measured if the diagnosis remains uncertain (particularly in the milder forms). Cell cultures reveal intracellular copper accumulation resulting from defective transport and cellular copper toxicity. [99], [100]

Pathophysiology Dietary copper is normally absorbed in the stomach and small intestine. Within the enterocyte, high-affinity copper transporters (hCTR1) transport copper into the cell. Copper is then transported to the trans-Golgi network and delivered by ATPase7A into the secretory pathway. The secretory pathway involves the production of copper-dependent enzymes such as lysyl oxidase, dopamine ß-hydroxylase, and tyrosinase. [101] When cellular copper concentrations increase, ATPase7A relocalizes to the plasma membrane and functions to transport copper out of the cell by the efflux pathway. [101] In Menkes disease, abnormality or absence of ATPase7A prevents the function of both these pathways. Copper is trapped at the intestinal level, resulting in severe deficiency at the tissue level. Impaired function of key enzymes ensues, leading to the observed clinical features (see Table 77-5).

Treatment Therapy includes parenteral copper histidine supplementation (up to 600 ng/kg weekly), although it does not appear to arrest cerebral degeneration. [102] Copper adducts are needed that cross the blood-brain barrier and might restore brain copper levels toward normal; these are not currently available. It has been suggested that the neuropathology is of early onset, linked to prenatal copper deficiency during the critical period of central nervous system development. [103] These findings are corroborated by the fact that copper deficiency secondary to malnutrition [104] is not usually associated with neurological complications.

Congenital Chloride Diarrhea

In 1945, two infants presented with severe diarrhea during the newborn period. Their feces was characterized by a high chloride content, hence the term *congenital chloride diarrhea*. Since then, nearly 100 cases have been described. [105], [106] A high proportion of cases have been reported from Finland, Poland, Saudi Arabia, and Kuwait; however, the distribution is worldwide. [107]

Genetics Congenital chloride diarrhea is inherited as an autosomal recessive trait, with a high prevalence found in the children of consanguineous relationships. By genetic linkage in Finnish families, the gene was localized to chromosome 7q31, proximal to the cystic fibrosis transmembrane regulator (CFTR). [108], [109] A candidate gene known as *DRA* ("down-regulated in adenoma") because of its reduced expression in colonic adenoma was identified in this position. The product of this gene is expressed in the brush border of normal ileum and colonic epithelial cells. [108] In a *Xenopus* expression model, *DRA* encodes a transporter with properties of Cl^-/OH^- exchange. [110] Twenty different mutations of the *DRA* gene have been identified in patients with congenital chloride diarrhea. [108]

Clinical Features One of the pathognomonic features of congenital chloride diarrhea is the fetal onset of diarrhea, evidenced by the development of maternal polyhydramnios. [106], [111] Other clinical features may include prematurity, absence of meconium, hyperbilirubinemia, abdominal distention, and failure to thrive. Radiographic studies of newborns reveal fluid-filled intestinal loops with or without ascites, again pointing to an intrauterine onset of the disease. If these early signs are absent, the diagnosis should be strongly suspected in any neonate in whom profuse watery diarrhea develops that is unresponsive to fasting and is often mistaken for urine. Without treatment, life-threatening dehydration and electrolyte imbalance may ensue as a result of persistent osmotic diarrhea. Fecal electrolyte analysis reveals an elevated chloride content (>90 mmol/L), a negative anion gap ($Na^+ + K^+ < Cl^-$), and a low fecal pH. The serum electrolyte levels and acid-base status may reveal hyponatremia and hypochloremia in the newborn period, although isosmolal dehydration is common; metabolic acidosis also has been reported. Metabolic alkalosis and hypokalemia develop progressively as a result of both intestinal and renal abnormalities. [111], [112] Not uncommonly, the condition is diagnosed in early infancy or early childhood if affected children survive the newborn period. They often carry another diagnosis related to their prevailing symptoms. [106], [111], [113] If congenital chloride diarrhea is diagnosed early and treated appropriately, adult patients are healthy but require lifelong supplements because of continued diarrhea (1.5–2 L/24 h). [112] The development of renal disease (i.e., hypertensive arteriopathy, juxtaglomerular hyperplasia, glomerular hyalinosis) has been described in a group of patients whose diagnosis and treatment were delayed or whose therapy consisted solely of potassium chloride supplements without sodium replacement. [114], [115]

Pathophysiology Intestinal perfusion studies [116], [117] have revealed a single defect in ileal and colonic sodium/hydrogen exchange. In subjects with congenital chloride diarrhea, the active exchange of anions (Cl^-/HCO_3^-) is absent, whereas cation exchange (Na^+/H^+) occurs normally. From the dietary NaCl delivered to the small intestine, Na^+ is absorbed in exchange for H^+ (equivalent of $Na^+HCO_3^-$ reabsorption) and excreted in the urine either as $Na^+HCO_3^-$ or with an organic anion (high urinary anion gap and pH). [106], [118] Chloride influx, however, in exchange for HCO_3^- does not occur; chloride-rich, acidic fecal fluid is excreted, the volume of which correlates with the osmotic load. [112] It is important to note that chloride influx takes place in patients with congenital chloride diarrhea by distinct pathways, most likely passive as revealed by all perfusion studies. [116], [117], [118] and [119] Strictly speaking, chloride is not actively secreted. Overall, affected subjects are unable to maintain a positive chloride balance in view of the daily obligate losses (intestinal and extraintestinal). Metabolic alkalosis is maintained by a combination of renal mechanisms; hypovolemia-induced aldosterone secretion promotes renal reabsorption of $NaHCO_3$ in the proximal tubule [120] and K^+ excretion in the cortical collecting duct, particularly in light of bicarbonaturia, [121], [122] leading to excessive K^+ losses. More importantly, hypokalemia itself stimulates renal bicarbonate sparing, as shown in micropuncture studies of euvolemic rats, thus contributing to persistent alkalosis. [120]

Treatment The mainstay of therapy is adequate replacement of Na^+, K^+, and Cl^-. [106] This implies that other mechanisms of intestinal chloride reabsorption (possibly passive) must be effective to render the defect compatible with life, [116] further allowing enteral electrolyte supplementation. As noted, NaCl replacement alone is not appropriate because hypokalemia contributes to the maintenance of metabolic alkalosis. Multiple drugs have been tried without success. [118] A report described a beneficial effect of omeprazole, [118] with reduction of stool frequency and volume. The authors proposed that inhibition of gastric chloride secretion (i.e., HCl) allowed a reduction of distally unabsorbed chloride. Indeed, fecal Na^+ and Cl^- losses decreased significantly, as did renal K^+ excretion in light of a drop in the bicarbonaturia, the latter most likely reflecting the decrease in gastric HCl loss; however, KCl supplementation had to be maintained to avoid hypokalemia. Of note, fecal K^+ losses decreased proportionally less than Na^+ and Cl^- losses during omeprazole therapy.

Differential Diagnosis The hallmarks of secretory diarrhea are twofold. First, Na^+, K^+, and their accompanying anions account for most osmoles excreted in the feces; the osmotic gap is thus small. Second, stool volume generally does not diminish with fasting. The differential diagnosis of secretory diarrhea is extensive, and the reader is referred to Chapter 42. One defect that is indistinguishable on clinical grounds from congenital chloride diarrhea is congenital sodium diarrhea.

Congenital Sodium Diarrhea

In four reports, neonatal watery diarrhea was ascribed to a defect of Na^+ reabsorption in the small intestine. [123], [124], [125] and [126] The clinical presentation was similar to that of congenital chloride diarrhea. In all three cases, the prenatal onset of watery diarrhea led to polyhydramnios, neonatal abdominal distention, and lack of meconium. A number of patients had choanal atresia, which has been associated with congenital secretory diarrhea. [126] In contrast to the infants with congenital chloride diarrhea, these infants had hyponatremia and metabolic acidosis. The fecal chloride was significantly lower than in congenital chloride diarrhea, the fecal Na^+ was high (100–200 mmol/L), the anion gap was positive (fecal $Na^+ + K^+ > Cl^-$), and the fecal pH was high. In brush border membrane vesicles obtained from their patient, Fell and colleagues [125] demonstrated a defect in the jejunal Na^+/H^+ exchange mechanism. Of interest, several Na^+/H^+ exchanger isoforms have been cloned to date, [127], [128] in particular NHE-3, which appears to be an epithelial brush border exchanger confined to the intestine and kidney. However, multifocal linkage analysis of patients excludes NHE-1, -2, -3, and -5 as potential genes for congenital sodium diarrhea based on their chromosomal location. [126] Therapy includes Na^+

and K^+ supplementation in the form of bicarbonate or citrate salts.

PRIMARY BILE ACID MALABSORPTION

Thaysen and Pedersen [129], [130] were the first to coin the term *idiopathic bile acid malabsorption* in their report of patients with chronic diarrhea of unknown cause who had concomitant fecal bile acid wasting. Several reports confirmed abnormal bile acid absorption in a number of patients with long-standing idiopathic diarrhea, [131], [132] and [133] as assessed by the elimination of 23-seleno-25-homotaurocholic acid (SeHCAT), a gamma-emitting synthetic analog of taurocholic acid. [134], [135] Bile acid malabsorption in general is associated with increases in intestinal fluid loss as the spillover of dihydroxy bile acids (i.e., chenodeoxycholic acid and deoxycholic acid) from the ileum into the colon leads to cyclic AMP–mediated sodium secretion. [136], [137] and [138] Interestingly, in some studies, numerous affected patients carried the diagnosis of functional diarrhea or irritable bowel syndrome before their bile acid absorption was evaluated, and their symptoms resolved after the administration of cholestyramine. The fact that some patients do not respond to cholestyramine [133] would indicate that another pathophysiological process is involved in the development of diarrhea with secondary bile acid malabsorption. Accordingly, some reported cases did have altered intestinal transit times, [131] although it is unclear whether these times were a cause or an effect of bile acid malabsorption. Analysis of the bile acid composition revealed normal concentrations of dihydroxycholic acids in the duodenal fluid of patients with bile acid malabsorption in comparison with controls. [139] Bile acid malabsorption has been classified into three distinct types. [138], [140] Type I is secondary to ileopathy or ileal resection; type II is an idiopathic entity with normal histological findings; type III occurs in association with a variety of other gastrointestinal disorders, such as chronic pancreatitis, cholecystectomy, and diabetes mellitus, and after peptic ulcer surgery. [131], [138], [141]

Pathophysiology

The absorption of bile acids in the small intestine is a highly efficient process whereby more than 95% of the bile acid pool secreted into the gastrointestinal tract is conserved. The major site of the intestinal conservation of bile acids is the terminal ileum. [142] The mechanisms participating in the enterohepatic recirculation of bile acids include both active and passive processes, the latter involving simple diffusion and possibly a facilitated, protein-mediated, Na^+-independent uptake process. [143] A specific Na^+-dependent transporter has been characterized in rabbit ileal enterocytes, [144] and membrane bile acid–binding polypeptides have been identified in rats. [145], [146] and [147] When transport rates of both conjugated and unconjugated bile acids in the jejunum and ileum were compared in intestinal perfusion studies performed in rabbits as well as in brush border membrane vesicles, the active Na^+-dependent uptake of conjugated bile acids was confined to the ileum. [148] Passive diffusion, although it occurs in both jejunum and ileum, appeared to be greater in the ileum in light of decreased membrane fluidity and a higher cholesterol-to-phospholipid ratio at that level. These membrane properties would contribute to the prevention of Na^+ backward flux in a segment containing multiple other Na^+-dependent transport mechanisms, which emphasizes the highly specialized functional character of the ileum in intestinal absorption processes. Data indicate that homeostasis of the circulating bile acid pool is achieved not only at the level of bile acid synthesis in the liver but also at the level of intestinal conservation. Following expression cloning of the sodium-dependent bile acid transporter of the rat liver basolateral membrane [149] and the hamster ileum brush border, [150] a 48-kd glycoprotein was demonstrated and expressed at the brush border membrane of the ileal villus enterocyte [151] that was highly homologous to the ileal hamster transporter. [150] The developmental regulation of this transporter appears to be mostly transcriptional at the time of weaning, with marked increases in mRNA and protein levels, similar to those of other brush border enzymes. [152] These data corroborate the results of earlier studies of the ontogeny of ileal bile acid transport in brush border membrane vesicles whereby carrier-mediated bile acid transport was absent at birth, only to appear abruptly in the early weaning period. [153] The expression of ileal bile acid transport appears to be initiated in part by the presence of lumenal substrate because expression of ileal bile acid transport can be induced with bile acid feedings during the suckling period. [151]

Considering the growing body of molecular data on ileal bile acid uptake, the hypothesis of a defect in the ileal sodium-dependent transporter as a cause of chronic diarrhea becomes more plausible, particularly if the onset is early in life. The human ileal Na^+/bile acid cotransporter gene (*SLC10A2*) was cloned and used for single-stranded conformational polymorphism analysis to find possible causative mutations in a family with primary bile acid malabsorption. Mutations were identified and shown to inhibit bile acid transporter function in transfected COS cells. [154] This report linked a mutation in a bile acid transporter to primary bile acid malabsorption. However, it did not explain adult-onset idiopathic bile acid malabsorption.

Clinical Features

Typically, patients present with a history of long-standing, at times intermittent, watery diarrhea and increased stool volume. Abnormal secretion of mucus, as well as increased colonic motility with urgency, may be caused by the presence of lumenal bile acids. Abdominal cramping usually resolves with defecation, and occasionally irritable bowel syndrome is diagnosed. As mentioned, bile acid malabsorption can be documented by using ^{75}SeHCAT to measure either fecal radioactivity or retained abdominal radioactivity. Increased levels of serum 7-a-hydroxy-4-cholesten-3-one were positively correlated with an abnormal ^{75}SeHCAT pattern in 19 of 23 patients studied. [155] The diagnosis of primary bile acid malabsorption remains one of exclusion; an increase in fecal bile acids in itself, even in the absence of other findings, does not imply a decrease in ileal uptake. [140] Similarly, a positive response to cholestyramine does not confirm that bile acid malabsorption is the primary defect; at most, it points to the fact that increased intralumenal bile acids may be contributing to the patient's symptoms. However, symptoms commonly recur without therapy.

DISORDERS OF LIPID MALABSORPTION

The assimilation of dietary fat is a complex process in which the enterocyte plays a pivotal role. Following digestion, solubilized fatty acids and monoglycerides are taken up by the enterocyte and rearranged into apolipoprotein-containing particles necessary for their transfer into the systemic circulation. A defect in the formation of these particles impairs the assimilation of dietary fats and leads to severe deficiencies, as is the case in abetalipoproteinemia.

Abetalipoproteinemia

Abetalipoproteinemia is an autosomal recessive disease characterized by the virtual absence of apolipoprotein B (apo B) and apo B–containing lipoproteins in the plasma of affected subjects. Triglycerides accumulate in the cytoplasm of hepatocytes and enterocytes secondary to abolished secretion.

Genetics Since abetalipoproteinemia was first reported in 1950, fewer than 100 cases have been reported with the abetalipoproteinemia phenotype. The pedigrees described show a recessive mode of inheritance, and some reports have suggested that consanguinity is as high as 50% in affected families. The phenotype is linked to a defect in the microsomal triglyceride transfer protein (MTTP), a neutral lipid transfer heterodimeric enzyme that includes a protein disulfide-isomerase (PDI) and a 97-kd subunit. The relationship between apo B and MTTP is discussed below. The human gene encoding the 97-kd subunit of MTTP has been cloned and sequenced [156], [157], [165], [166] and localized to chromosome 4q22-24. [158] Genetic studies were undertaken in subjects carrying the abetalipoproteinemia phenotype. [159], [160] All were carriers of a homozygous or compound heterozygous mutation of both alleles. Most subjects demonstrated a truncated form of the MTTP large subunit (97 kd) that impeded its interaction with PDI and rendered the complex nonfunctional in its role of shuttling lipids between membranes of the endoplasmic reticulum. [161]
Pathophysiology The major lipids found in tissues and cells, triglycerides and cholesteryl esters, are secreted into the extracellular fluid in particles in association with the large hydrophobic protein apo B, which is the product of a single gene and occurs in two distinct forms, apo B-48 (264 kd) and apo B-100 (550 kd). The former is essential for chylomicron formation in enterocytes, in which dietary free fatty acids and cholesterol are esterified to triglycerides and cholesteryl esters, respectively. These esters are incorporated into apo B–containing particles (chylomicrons) before they are transferred into the extracellular space. A similar process occurs in hepatocytes, in which apo B-100 is secreted in association with triglycerides to form very-low-density lipoproteins (VLDLs), lipid (triglyceride)–rich particles exporting long-chain fatty acids synthesized in the liver. Apo B is also the major protein component of low-density lipoprotein (LDL) particles carrying cholesteryl esters to peripheral tissues by means of LDL receptors. [162] Apo B, which contains insufficient quantities of lipids that enhance its stability within these particle structures, undergoes intracellular degradation by binding to heat shock protein 70, which disposes of misfolded proteins by the ubiquitin-proteosome pathway, in keeping with the findings that its secretion is regulated posttranscriptionally. [163] The binding of heat shock protein 70 to apo B is increased in cells depleted of lipids to regulate the amount of lipoprotein formed. [163] The discovery that abetalipoproteinemia is not the result of defective apo B synthesis, [164], [165] and [166] as initially contended, and that no genetic linkage exists between the disorder and the apo B gene [167] has not only allowed a basic understanding of the defect but also has shed new light on the mechanisms of triglyceride-rich particle assembly. Wetterau and colleagues [156], [168], [169] first isolated and characterized MTTP, a protein complex, from microsomal membranes of bovine liver. MTTP is a heterodimeric enzyme composed of PDI, a ubiquitous multifunctional resident endoplasmic reticulum protein, and a unique 97-kd subunit that confers lipid transfer activity to the complex. Studies in subjects with abetalipoproteinemia have shown that the assembly of apo B–containing lipoproteins is defective as a result of a nonfunctional MTTP. [156], [170], [171] The process whereby lipids are transferred to apo B, allowing apo B to form a

secretion competent particle, is thus abolished. Accordingly, the concomitant expression of MTTP and apo B in CO8-1 cells and HeLa cells is sufficient to mediate the secretion of triglyceride-rich lipoproteins. [172], [173] Furthermore, mRNAs for apo B and apo B protein are present in the intestinal mucosa and liver of affected subjects. [174], [175] The exact relationship between MTTP structure and its function in apo B lipidation has been only partly elucidated. Data extracted from studies of lipid-binding sites on MTTP [176] and from studies of the regulation of expression by dietary fat content [177] point to distinct mechanisms leading to particle assembly. MTTP functions in the initial transfer of lipids from the endoplasmic reticulum during apolipoprotein translation and translocation into the rough endoplasmic reticulum. After translocation, apo B is initially folded into a compact globular structure of a-helices supported by disulfide bonds. The subsequent amphipathic ß-sheet domain of apo B binds lipid that is transferred by MTTP. [163] This forms a framework for the lipoprotein molecule that would otherwise misfold and be degraded though interaction with heat shock protein 70. The particle then fuses to triglyceride droplets at the junction of the rough and smooth endoplasmic reticulum to form mature chylomicrons and VLDL. [163] Further involvement of MTTP in the formation of chylomicrons and VLDL remains to be elucidated. Abetalipoproteinemia may not be the only phenotype caused by a defect in the MTTP gene.

Clinical Features The clinical expression of this disorder depends on the age at onset. Symptoms of malabsorption and failure to thrive secondary to steatorrhea are the hallmarks of early onset in infancy and childhood. [178] Poor growth may precede overt gastrointestinal symptoms, and affected children are frequently thought to have celiac disease. The steatorrhea may diminish with time if dietary fat is avoided. [178] In addition, polyunsaturated fats appear to be better tolerated than saturated fats. If the disease is unrecognized as patients enter their second and third decades of life, neurological and ophthalmic symptoms become apparent, along with their progressively debilitating effects. [178] Spinocerebellar axonal degeneration and pigmented retinopathy result from a chronic deficiency of fat-soluble vitamins E and A, respectively, mimicking Friedreich ataxia. Clinically, early signs include the loss of deep tendon reflexes, followed by a decrease in proprioceptive and vibratory senses in the lower extremities. Cerebellar signs, such as sensory ataxia (Romberg sign), dysmetria, and spastic gait, ensue. Typically, these central nervous system abnormalities are devastating and lethal. A sensory peripheral neuropathy also has been described that is a result of the degeneration and demyelination f large nerve fibers. Ophthalmic symptoms of retinitis pigmentosa (night and color blindness) develop first. Nystagmus and ophthalmoplegia are frequent. Progression to macular involvement in untreated patients leads to blindness, usually in the fourth decade of life. The diagnosis of abetalipoproteinemia is based on hematologic, biochemical, and histopathological findings, the last of which depend on which organs are involved. Hematologically, mild to moderate anemia results from chronic hemolysis. Severe vitamin E deficiency causes autohemolysis. The presence of predominantly spiculated acanthocytes within the red blood cell population (50%–70%) further contributes to the anemia. Their abnormal membrane lipid composition shortens the life span of erythrocytes. Vitamin K deficiency may increase the prothrombin time. The biochemical hallmark in affected patients reflects the lack of lipoprotein secretion by enterocytes. Plasma lipid and lipoprotein profiles include hypocholesterolemia (0.5–1.30 mmol/mL), low triglyceride levels, and virtually undetectable levels of chylomicrons (including postprandially), VLDLs, LDLs, and apo B. Plasma levels of vitamin A and both plasma and tissue levels of vitamin E are significantly decreased. [178] Histopathology of the gastrointestinal tract reveals normal villi, distinguishing the condition from celiac disease, but enterocytes are lipid laden. Their cytoplasm contains free lipid droplets, whereas the Golgi apparatus is devoid of lipoprotein-containing particles. Elevation of hepatic enzymes and hepatic steatosis have been observed but are extremely rare. Other clinical abnormalities have been described in isolated cases, such as aminoaciduria, hypogammaglobulinemia, and physical anomalies of digits, but it is unclear whether these findings are related to abetalipoproteinemia.

Treatment Restriction of fats, in particular long-chain fatty acids, alleviates the gastrointestinal symptoms. Medium-chain fatty acids may be used temporarily if severe malnutrition is present, but their routine use should be avoided because they can worsen hepatic steatosis. [179] Appropriate vitamin supplementation must be the basis of therapy because the inability to form chylomicrons leads to a deficiency of fat-soluble vitamins. [179] Vitamin E supplementation is beneficial in both preventing and potentially reversing the neurological and ophthalmic features of the disease. [180] Because it is one of the most nonpolar lipids known, the assimilation of vitamin E is affected not only by decreased intestinal absorption but also by the absence of LDL and VLDL particles involved in its hepatic secretion and systemic transport, respectively. [181] These negative effects can be overcome by administering large doses of vitamin E (100–200 mg/kg daily) polyethylene glycol 1000 succinate (Liqui-E). [180] Monitoring the plasma levels of vitamin E alone is unreliable, but quantification of the vitamin E levels in adipose tissue does reflect the vitamin E status of these patients; however, this method is not practical for routine clinical use. The hydrogen peroxide hemolysis test does serve as a good screening test for increased vitamin E stores. [180] When the total serum lipids are reduced, the ratio of vitamin E to total lipids is not a reliable measure of the vitamin E status. [180] Clinically, improvement in tissue vitamin E levels may be reflected in improved visual or somatosensory evoked responses. Vitamin A (200–400 IU/kg per day or 25,000 IU daily or every other day) and vitamin K (5–10 mg/d) are given in their water-miscible forms (Aquasol A and Mephyton, respectively) to normalize the levels of vitamin A in plasma as well as the prothrombin time. Disorders of lipid metabolism that may clinically mimic abetalipoproteinemia include chylomicron retention disease (Anderson disease), caused by the inability of enterocytes to secrete chylomicrons, and familial hypobetalipoproteinemia, caused by truncations of apo B. The homozygous form of familial codominant hypobetalipoproteinemia is clinically and biochemically indistinguishable from homozygous recessive abetalipoproteinemia. Anderson disease also may present in early childhood with severe steatorrhea and growth failure [182]; in contrast to the fasting triglyceride levels in the two former disorders, however, the fasting triglycerides in Anderson disease are normal. The exact defect is not known; apo B-48 is present in enterocytes, which are filled with fat, but not in plasma, suggesting that a defect may be present at several steps involving lipidation, translocation, or secretion of apo B-48 (see Chapter 18).

DEFECTS IN INTESTINAL COBALAMIN ABSORPTION

The intestinal absorption of cobalamin (vitamin B_{12}) occurs in the ileum and is receptor mediated. [183] Dietary cobalamin undergoes proteolytic release from food sources and then is complexed with haptocorrin in the presence of a low gastric pH. Exposure of this complex to pancreatic proteases permits the release of cobalamin and its binding to intrinsic factor (IF), which is synthesized in the gastric parietal cells. These initial steps require intact gastric function for acid secretion, proteolytic activity, and IF secretion. Only IF-bound cobalamin binds to the ileal receptor, allowing endocytosis of the complex. Other ligands for cobalamin include transcobalamins I, II, and III (TCI, TCII, and TCIII). TCII, which is synthesized in most tissues as well as in the ileal mucosa, is required for the uptake of cobalamin into tissues. It is thought that after internalization of the IF-cobalamin complex bound to its ileal receptor, cobalamin is transferred intracellularly from IF to TCII. This secondary complex then enters the systemic circulation. Cobalamin also may be transported systemically bound to TCI and TCIII, which are circulating haptocorrins in the plasma. Their precise role in the homeostasis of cobalamin is not known, but they are cleared by the liver, and the cobalamin is secreted in the bile as one arm of an enterohepatic circulation.

Congenital defects affecting intestinal vitamin B_{12} uptake include congenital IF deficiency (congenital pernicious anemia) and congenital vitamin B_{12} malabsorption resulting from an ileal defect (Imerslünd-Grasbeck syndrome). TCII deficiency affects cellular uptake and is unique in that serum levels of cobalamin are normal. TCI (R binder) deficiency also has been described. [184], [185] Clinical signs are absent despite low levels of serum cobalamin, and affected subjects do not require vitamin B_{12} supplementation. Addisonian pernicious anemia, in contrast to congenital pernicious anemia, presents in adulthood and is characterized by the presence of anti-IF antibodies (types I and II), antiparietal cell antibodies, atrophic gastritis, and achlorhydria (Table 77-6).

TABLE 77-6 Vitamin B_{12} Malabsorption Syndromes

Pathophysiology

The biochemical abnormalities associated with the disorders of cobalamin deficiency reflect dysfunction of the cobalamin coenzymes adenosylcobalamin and methylcobalamin, which are involved in the synthesis of succinyl-CoA from L-methylmalonyl-CoA and methionine from homocysteine, respectively. Hence, methylmalonic aciduria, homocystinuria, or both have been observed in cobalamin-deficient patients. The clinical manifestations, of which megaloblastic anemia is the hallmark, result from disruption of the folate cycle, which depends on the conversion of homocysteine to methionine by methylation; this, in turn, allows the regeneration of tetrahydrofolate. The accumulation of methyltetrahydrofolate ensues, leading to functional folate deficiency. It has been suggested that the neurological effects of cobalamin deficiency may be secondary to demethylation of myelin basic protein. [186]

Congenital Pernicious Anemia

A juvenile form of pernicious anemia has been reported in a number of children presenting with megaloblastic anemia and developmental delay. [187] The age at onset

is typically between 1 and 5 years. The delay in the clinical expression of vitamin B_{12} deficiency compared with TCII deficiency generally is explained by replete body stores in early infancy. These depend on maternal vitamin B_{12} stores. Because the transplacental transfer of cobalamin to the fetus occurs in the third trimester, infants who received inadequate vitamin B_{12} in utero express clinical symptoms earlier. In humans, IF has been detected in fetal gastric extracts from 11 weeks onward [188] in the corpus and pylorus. Solubilized IF receptor (IFR) activity has been found in both small intestine and colon between 10 and 25 weeks' gestation but only in ileum thereafter. The exact mechanism of this proximal-to-distal gradient of IFR expression requires further elucidation. From a functional point of view, the early expression of all components of the vitamin B_{12} entry pathway demonstrates the potential for uptake of the receptor-IF-B_{12} complex during fetal life. [188]

Congenital pernicious anemia differs from the ileal defect syndrome (see below) by the absence of functional IF. IF may be immunologically undetectable because of decreased synthesis, [184] or it may be present but nonfunctional. [189] Reports of nonfunctional IF have included a low affinity of IF for either cobalamin or the ileal receptor. In addition, Levine and colleagues [190] described three siblings who presented in their second year of life with megaloblastic anemia and abnormal Schilling test results (<1%), corrected with exogenous IF. The native IF demonstrated increased susceptibility to gastric acid and proteolysis, with loss of its normal structure and function. A cDNA for IF in both rats and humans has been cloned, [191], [192] and the IF gene has been localized to chromosome 11. [192] Nevertheless, the genetic defect, which is inherited in an autosomal recessive manner, remains unidentified to date. Southern blot analysis of native DNA from probands did not reveal a major gene mutation, suggesting altered gene expression or a possible point mutation. Therapy consists of the parenteral administration of cobalamin (OH-cobalamin, CN-cobalamin) to bypass gastric and ileal factors [193] and to achieve resolution of all clinical abnormalities. [183]

Ileal Defect Syndrome (Imerslünd-Grasbeck Syndrome)

Grasbeck and associates [194] and Imerslünd [195] independently described a syndrome consistent with an autosomal recessive selective malabsorption of vitamin B_{12}. Children present after the first year of life with hematologic abnormalities, infection, and failure to thrive, as in congenital pernicious anemia, with the exception that the Schilling test result cannot be corrected by the administration of IF. The activity of the urinary receptor for the IF-cobalamin complex is more than 600 times lower in affected patients than in controls, and this has been suggested as a diagnostic test for Imerslünd-Grasbeck syndrome. [196] Proteinuria that is not associated with progressive renal failure and does not correct with treatment is also part of the phenotype specific to this syndrome. [197] The defect occurs in the transport mechanism of the IF-cobalamin complex at the level of the ileal enterocyte. The study of various probands has shown that the defect affects either receptor binding [198] or postbinding processing in the cell. Deficient cobalamin-IFR activity has been demonstrated in the ileal biopsy specimens of patients with Imerslünd-Grasbeck syndrome. [199] However, in one report of two sisters with Imerslünd-Grasbeck syndrome, [200] the affected patients showed significantly greater receptor activity than controls, with undetectable protein on immunoblotting of total biopsy membrane preparations. The investigators hypothesized that the patients overexpressed an unstable receptor. An explanation of these seemingly contradictory results requires further investigation of individuals affected with the disorder. [201] Linkage studies have been used to assign the location of the disorder to chromosome 10. Within this locus, the human cubilin gene encodes a protein that has IF-cobalamin activity with megalin, another protein. [202], [203] However, the nature of this interaction has not been discovered. As in IF deficiency, the exact genetic defect is not known, and the parenteral administration of cobalamin leads to resolution of the symptoms.

Transcobalamin II Deficiency

Three forms of TCII deficiency have been described with the following characteristics: (1) absence of TCII from the plasma (the most common phenotype); (2) presence of immunoreactive TCII that is unable to bind cobalamin 1; and (3) immunoreactive TCII that is unable to promote the uptake of cobalamin 1 into cells. An absence or abnormality of TCII impairs the TCII-dependent tissue uptake of cobalamin. Intestinal absorption is usually normal, as are serum levels of cobalamin bound to TCI and other haptocorrins. [204] Hakami and colleagues [205] reported low intestinal absorption despite exogenous IF in two siblings with neonatal megaloblastic anemia and TCII deficiency. In contrast to patients with congenital IF deficiency, patients are normal at birth but present shortly thereafter. The genetic defect that causes TCII deficiency remains to be fully described. Patients with TCII deficiency have been shown to have various mutations within the coding region of the gene for TCII. [206] The 5' promoter for TCII contains regulatory elements that may also play a role in its expression in vivo. [207] Symptoms of TCII deficiency include vomiting, diarrhea, and hypotonia in the first few weeks or months of life associated with megaloblastic anemia and failure to thrive; subsequently, immunologic and neurological abnormalities develop. [208] The administration of large doses of parenteral vitamin B_{12} is the treatment of choice to achieve passive diffusion of the vitamin into cells.

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CHAPTER 78

Ellen Li

BACTERIAL OVERGROWTH

Bacterial overgrowth of the small bowel is a syndrome in which nutrient malabsorption is associated with excessive numbers of bacteria in the small intestine. [1], [2] Other terms that have been used to describe the syndrome of bacterial overgrowth are *blind loop syndrome, contaminated small bowel, small bowel stasis,* and *stagnant loop syndrome*. Recognition dates back to 1897, when Faber [3] reported an association between pernicious anemia and strictures of the small intestine. In 1939, Barker and Hummel [4] first postulated that the association of macrocytic anemia with intestinal strictures or anastomoses is the result of bacterial overgrowth, or "putrefaction." Patients with a variety of structural or functional disorders of the gastrointestinal tract are predisposed to the development of bacterial overgrowth. [1], [2] Evidence suggests that bacterial overgrowth may develop in the absence of an anatomic lesion or a severe motility disorder in the elderly and in children. [5], [6] Bacterial overgrowth should be considered as a possible cause of diarrhea, malabsorption, or abdominal pain in these clinical settings. Because the diagnosis of bacterial overgrowth presents several difficulties and limitations, empiric treatment often is used in clinical practice.

CONDITIONS FAVORING BACTERIAL OVERGROWTH

The conditions favoring bacterial overgrowth can be classified according to the mechanisms involved in the normal control of the enteric flora (Table 78-1). Furthermore, the presence of a single condition may not result in clinically significant small intestinal overgrowth until another condition favoring bacterial overgrowth develops.

Intestinal Stasis
Anatomic
Strictures (e.g., Crohn's disease, radiation enteritis)
Diverticulosis of the small intestine
Surgical operations
End-to-side enteroenteric anastomoses
Billroth II anastomoses
Jejunoileal bypass
Koch distal ileal pouches
Small intestinal motility disorders
Scleroderma
Idiopathic intestinal pseudoobstruction
Diabetic autonomic neuropathy
Abnormal Connection between Proximal and Distal Bowel
Fistulae (secondary to peptic ulcer disease, carcinoma)
Gastrocolic
Gastrojejunocolic
Resection of the ileocecal valve
Hypochlorhydria
Chronic atrophic gastritis
Hypochlorhydric medications
Surgical therapy for peptic ulcer disease
Immunodeficiency
Primary immunodeficiency states
Acquired immunodeficiency syndrome
Malnutrition
Age

TABLE 78-1 Conditions Favoring Bacterial Overgrowth

Stasis of the Small Intestine

Small intestinal peristalsis plays a major role in preventing bacterial overgrowth. Anatomic or structural disorders that interfere with intestinal peristalsis frequently lead to small bowel bacterial overgrowth. These include chronic small intestinal obstruction secondary to intestinal strictures, such as those caused by Crohn's disease, lymphoma, or radiation injury. [7], [8] and [9] Surgical alterations of the intestinal anatomy that create blind pouches or divert long segments small bowel, such as end-to-side enteroenteric anastomosis, the Billroth II anastomosis, jejunoileal bypass, and the Koch distal ileal pouch (for continent ileostomies), have been associated with clinically significant bacterial overgrowth [10], [11] and [12] in stagnant pouches and loops.

Diverticula of the small intestine can serve as pockets of stagnation that become clinically significant, particularly in the setting of hypochlorhydria or achlorhydria. [10] Generalized impairment of small intestinal motor function (see Chapter 11) also may lead to small bowel bacterial overgrowth. Malabsorption resulting from small intestine bacterial overgrowth has been documented in patients with intestinal hypomotility in association with scleroderma, idiopathic intestinal pseudoobstruction, or diabetic autonomic neuropathy. [13], [14] and [15] The absent or disordered migrating motor complex has been associated with bacterial overgrowth [16]; in one patient, the return of phase III of the migrating motor complex preceded the resolution of bacterial overgrowth. [17]

Abnormal Connections between Proximal and Distal Portions of the Bowel

Bacterial overgrowth of the small intestine may arise secondary to abnormal connections between the proximal bowel and the colon that lead to seeding of the proximal bowel with colonic flora. Resection of the ileocecal valve may cause retrograde seeding of the small intestine with colonic flora and contribute to the development of small intestinal bacterial overgrowth. [18], [19] Evidence also indicates that resection of the ileocecal valve (as well as a portion of the terminal ileum and cecum) results in alterations of motility that may play an important role in the development of bacterial overgrowth. [20] Gastrocolic or gastrojejunocolic fistulae may arise in patients with gastric ulcers and ulcerating carcinomas of the stomach or colon. In this situation, malabsorption results from reflux of the colonic contents into the proximal bowel. [21]

Gastric acid is responsible for decreasing the bacterial inoculum reaching the proximal small intestine. The proximal gastrointestinal bacterial concentration is significantly increased in patients with achlorhydria secondary to chronic gastritis in comparison with control subjects. [22] Gastric acid secretion also is impaired as a result of medical and surgical therapy of peptic ulcer disease. Increased bacterial counts and overgrowth have been reported in association with cimetidine therapy [23]; these may become even more significant clinically when more potent hypochlorhydric medications, such as omeprazole, are used. Gastric surgery affects bacterial

proliferation by impairing gastric acid secretion and by altering intestinal motility. The results of studies of animal models and humans after surgery suggest that impaired gastric acid secretion alone is not sufficient to cause clinically significant bacterial overgrowth. [11], [24] Hypochlorhydria, however, may promote intestinal overgrowth in patients who also have intestinal disorders associated with stasis (e.g., diverticula, delayed intestinal transit).

Immunodeficiency

Chronic diarrhea, malabsorption, and weight loss have been observed in a number of immunodeficiency states (see Chapter 51). For example, 60% of patients with common variable immunodeficiency have chronic diarrhea. Frequently, malabsorption is associated with *Giardia lamblia* infestation and responds to treatment of the giardiasis. Overgrowth of bacteria is a common finding in these patients; however, the bacterial counts (particularly those of anaerobic organisms) are lower than those in cases associated with intestinal stasis. [25] Bacterial overgrowth also may be related to decreased acid secretion in these patients. [25] Chronic diarrhea is a prominent symptom in patients with the acquired immunodeficiency syndrome (AIDS), who may have multiple opportunistic infections (see Chapter 124); however, the cause of their diarrhea and malabsorption frequently cannot be identified. Bacterial colonization of the proximal bowel has been observed in patients with AIDS, but the role it plays in the pathogenesis of AIDS-related diarrhea remains speculative because the concentration of anaerobic organisms is far lower than that usually observed in bacterial overgrowth associated with stasis. [26]

Age

The prevalence of small bowel bacterial overgrowth increases with age (15%–56%), and overgrowth can occur in the absence of an anatomic defect of the small bowel. [5], [27], [28], [29] and [30] Possible explanations are reduced gastric acidity, impaired intestinal motility, or both. Because bacterial overgrowth is not associated with baseline anthropomorphic measurements or bowel habits, the clinical significance of bacterial overgrowth in the elderly is unclear, and it should not be routinely treated. However, in patients with evidence of malabsorption and bacterial overgrowth, treatment with antibiotics can be useful. [30], [31]

PATHOGENESIS

The pathogenesis of small bowel bacterial overgrowth can be discussed in terms of the consequences of misplacing the colonic flora into the small intestine. The enteric flora, which normally is located principally within the colon, plays an important role in the metabolism of intralumenal substances (Table 78-2). The amount of intralumenal substances that the bacteria normally encounter, however, is limited by small intestinal absorption. In cases of small bowel bacterial overgrowth, the bacteria have much greater access to nutrients and other intralumenal substances. In addition to intralumenal bacterial metabolism, evidence suggests that mucosal injury also results in the malabsorption of fats, carbohydrates, and proteins [32], [33]; however, bacterial invasion does not appear to be involved. [34]

Bacterial Metabolism of Intralumenal Substances
Impaired deconjugation of bile acids
Impaired vitamin B_{12} metabolism
Impaired metabolism of carbohydrates
Malabsorption of fats and vitamins A, D, and E; generation of secretagogues
Malabsorption of vitamin B_{12}
Intestinal gas (H_2, CO_2), osmotic diarrhea
Mucosal Injury
Decreased brush border enzyme activity
Lactase, enterokinase, peptidase
Impaired uptake of monosaccharides
Impaired uptake of peptides
Impaired uptake of lipids
Lactose intolerance
Hypoproteinemia
Carbohydrate intolerance
Fat malabsorption

TABLE 78-2 Pathogenesis of Bacterial Overgrowth

The surgical creation of a self-filling blind loop (Fig. 78-1) in a variety of animal models results in a marked proliferation of both aerobic and anaerobic bacteria and reproduces many of the manifestations of overgrowth observed in humans. [35], [36] Much of the experimental work defining the pathogenesis of small bowel bacterial overgrowth is based on these models.

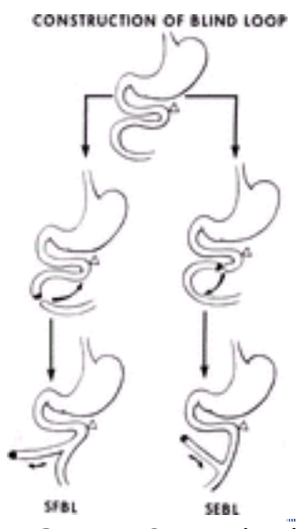

FIGURE 78-1. Animal model of small bowel bacterial overgrowth. Diversion of an isoperistaltic segment of jejunum results in the formation of a self-filling blind loop (*SFBL*). The SFBL is often used to characterize the pathophysiology of bacterial overgrowth in the small bowel because it results in the proliferation of colonic-type bacterial flora and sequelae that mimic those observed in human disease. A short, patent, antiperistaltic segment of jejunum, referred to as a self-emptying blind loop *(SEBL),* serves as an operated control because bacterial overgrowth and its sequelae do not develop postoperatively. (From ref. [45].)

Malabsorption of Fats and Fat-Soluble Vitamins

The bacterial deconjugation of bile acids is the primary mechanism for the malabsorption of fats and fat-soluble vitamins. [37], [38] Normal fat absorption requires a critical concentration of conjugated bile acids for the assembly of mixed micelles. The deconjugation of bile acids by lumenal bacteria, particularly anaerobic organisms, [39] reduces the level of conjugated bile acids below the critical micellar concentration. Steatorrhea has been relieved in patients [37] and experimental animals [38] with bacterial overgrowth by the administration of conjugated bile acids; however, the observation that complete biliary diversion in animals does not produce the steatorrhea seen in bacterial overgrowth suggests that other factors may play a role. [19] Deficiencies of vitamins D, A, and E have been reported as complications. [40], [41] The synthesis of vitamin K by lumenal bacteria, however, accounts for the absence of coagulopathy in patients with bacterial overgrowth. It has been suggested that bacterial metabolites (e.g., hydroxylated fatty acids and unconjugated bile acids) may have a toxic effect on the intestinal mucosa, resulting in the malabsorption of fats, carbohydrates, and proteins. Many of these metabolites serve as secretagogues that contribute to the development of diarrhea.

Carbohydrate Intolerance

Bacterial overgrowth results in carbohydrate intolerance secondary to a reduction of brush border disaccharidases and a decreased uptake of monosaccharide. [42], [43], [44] and [45] Lactase activity is the first to be reduced and is the last disaccharidase activity to recover after antibiotic therapy has been administered. [43], [45] Evidence

suggests that anaerobic organisms produce proteases and glycosidases that release or destroy hydrolases on the brush border. [42], [43], [46] Carbohydrate intolerance results in an increased delivery to the small intestine of osmotically active carbohydrate fragments, which contribute to the development of the diarrhea associated with bacterial overgrowth. Bacterial metabolism of carbohydrates to hydrogen and carbon dioxide may play a role in the development of abdominal pain and is the basis of various breath tests used to diagnose bacterial overgrowth.

Hypoproteinemia

Hypoproteinemia is common in bacterial overgrowth, although severe protein nutrition is rarely seen. [47] Normal protein assimilation is disrupted by multiple factors. Bacteria compete with the host for protein substrates [44]; decreased amino acid and peptide uptake has been demonstrated in experimental models, perhaps reflecting mucosal injury [47]; decreased levels of enterokinase have been noted in patients with bacterial overgrowth and may impair the activation of pancreatic proteases [48]; and finally, a protein-losing enteropathy has been described in patients and experimental animals with bacterial overgrowth. [49]

Malabsorption of Water-Soluble Vitamins

The association of macrocytic anemia with bacterial overgrowth is the result of direct competition between the intestinal flora and the host for vitamin B_{12}. The anaerobic organisms are primarily responsible because only anaerobicidal therapy reversed vitamin B_{12} deficiency in an experimental model of stasis. [19] Anaerobic organisms, in contrast to aerobic organisms, can use vitamin B_{12} either in its free form or when it is complexed with intrinsic factor. [50], [51] When bacteria take up the vitamin, not only does it become unavailable to the host, but inactive metabolites are produced that compete with normal vitamin B_{12} binding and absorption. [52] In contrast, folate levels are normal or elevated in bacterial overgrowth because bacteria synthesize folate that subsequently is absorbed by the host. Levels of thiamin [53] and nicotinamide, [54] two other water-soluble vitamins, have also been reported to be low in bacterial overgrowth.

Although iron deficiency anemia has not been clearly associated with bacterial overgrowth in humans, increased intestinal losses of iron and blood were documented in animal models. [55] Mineral and trace element deficiencies have not been reported in patients with bacterial overgrowth.

Liver Disease

Hepatobiliary injury has been observed in experimental models of bacterial overgrowth in susceptible rat strains. [56], [57] The injury can be prevented by antibiotic treatment. [58] It has been suggested that bacterial cell wall polymers or other bacterial toxins from the blind loop cause hepatic lesions in genetically susceptible hosts. [56] A high prevalence of bacterial overgrowth and elevated levels of tumor necrosis factor-a have been detected in patients with nonalcoholic steatohepatitis (NASH), and it has been proposed that bacterial overgrowth plays a role in the pathogenesis of NASH by triggering cytokine release. [59], [60]

Small bowel bacterial overgrowth occurs with increased frequency in patients with cirrhosis. [61], [62] The mechanisms are likely to be multifactorial and include gastric hypoacidity and delayed small bowel transit. The consequences of this change are not fully understood. It has been suggested that patients with cirrhosis and small bowel bacterial overgrowth are predisposed to the spontaneous development of bacterial peritonitis. [61], [62] and [63]

PATHOLOGY

Lumenal bacteria clearly influence intestinal morphology. Many of the normal morphologic characteristics of the intestinal epithelium are seen only in association with a resident bacterial flora (see Chapter 25). In the germ-free state, the villi are thinner and longer, and the crypts are shallower. The mucosal cells are cuboidal rather than columnar. The lamina propria consists of a sparse stroma infiltrated with a few lymphocytes and macrophages, plasma cells are absent, and the Peyer patches are smaller, with fewer germinal centers. On light microscopy, the histological appearance of the small intestine with bacterial overgrowth usually is not significantly different from that of the intestine with a normal enteric flora. The main purpose of obtaining a mucosal biopsy specimen in the evaluation of these patients is therefore to rule out mucosal diseases such as celiac sprue. Histological evidence of mucosal damage, however, has been found in some cases of bacterial overgrowth, including subtotal villus atrophy and increased inflammatory cells within the lamina propria. [33] Focal areas of ulceration and erosion have been observed in some instances and may be important in understanding the symptoms of “pouchitis” associated with bacterial overgrowth after the surgical creation of a continent ileostomy (Koch pouch) or ileoanal anastomosis. [64], [65] Ultrastructural studies of experimental models of bacterial overgrowth have revealed vacuolization of microvillus membranes and swelling of the mitochondria (Fig. 78-2), suggesting that bacterial overgrowth does result in damage to the enterocyte. [32]

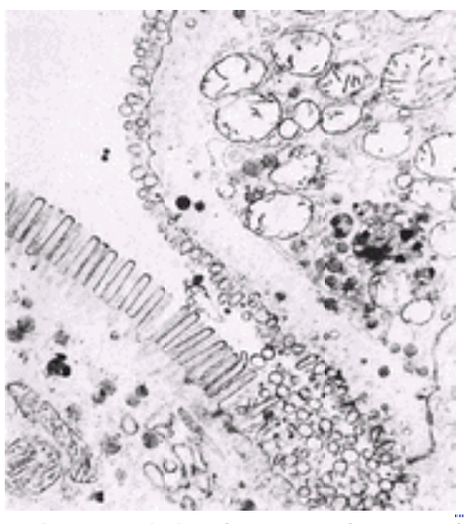

FIGURE 78-2. Electron micrograph of columnar cells in the midvillus area of a self-filling blind loop. In the cell at the lower left, the microvilli are regular, and the mitochondria and endoplasmic reticulum are unaltered. In the cell at the upper right, the microvilli are altered, and the mitochondria and endoplasmic reticulum are swollen. Alterations of the microvilli include blunting, swelling, budding, and ballooning. Original magnification ×22,000. (From ref. [32].)

CLINICAL MANIFESTATIONS

Many of the clinical features of bacterial overgrowth can be predicted based on the pathogenesis of the disorder (Table 78-3). Diarrhea and weight loss are prominent symptoms and are largely a consequence of the malabsorption and maldigestion of fats, carbohydrates, and proteins. In addition, many patients with bacterial overgrowth curtail their intake to reduce symptoms. [2] The association of macrocytic anemia with intestinal strictures is a manifestation of vitamin B_{12} deficiency. Patients also may present with the neurological changes associated with vitamin B_{12} deficiency. Recurrent abdominal pain may be reported, particularly among young children, as a presenting symptom. [6] Other presentations include night blindness, osteomalacia, tetany, peripheral neuropathy, and edema, [66], [67] reflecting malabsorption of micronutrients such as vitamins A, D, and E, as well as thiamin and nicotinamide.

Diarrhea
Steatorrhea
Malnutrition
Macrocytic anemia
Abdominal pain
Peripheral neuropathy
Tetany
Osteomalacia
Night blindness
Dermatitis, hepatic injury, nephrotoxicity, and arthritis (observed in jejunoileal bypass)

TABLE 78-3 Clinical Manifestations of Bacterial Overgrowth

Extraintestinal manifestations of bacterial overgrowth have been observed, especially in patients who have undergone jejunoileal bypass. These include dermatitis, hepatic injury, nephrotoxicity, and arthritis. [68], [69] and [70]

DIAGNOSIS

The diagnosis of bacterial overgrowth should be considered in patients with chronic diarrhea who have the predisposing conditions discussed previously in this chapter. The differential diagnostic considerations include other causes of malabsorption and maldigestion: intestinal mucosal diseases (e.g., celiac sprue), infections (particularly parasitic diseases such as giardiasis), and pancreatic insufficiency. In patients with underlying diseases that favor bacterial proliferation, such as Crohn's disease or scleroderma, it may be difficult to distinguish whether clinical deterioration is caused by the development of bacterial overgrowth or by worsening of the primary intestinal disease. It is possible that the diagnosis should also be considered in patients with symptoms of irritable bowel syndrome. A high prevalence of bacterial overgrowth has been observed in patients in whom irritable bowel syndrome has been diagnosed. Furthermore, eradication of bacterial overgrowth appears to reduce their intestinal complaints. [71]

Microbiologic culture of small bowel aspirates is considered the most direct method for diagnosing bacterial overgrowth (Table 78-4). [1], [2] In general, the diagnosis of bacterial overgrowth is considered to be confirmed if the count exceeds 10^5 colonies per milliliter after duodenal intubation. [2] A variety of techniques have been reported: the use of sterile and nonsterile tubes, [72] the capsule method, [73] direct needle aspiration of the gut contents, [74] and the string test. [75] The feasibility of collecting aspirates from the proximal small bowel under direct visualization at the time of endoscopy has been demonstrated and can facilitate the collection of samples during routine endoscopy. [76] Small intestinal aspirates are obtained by placing a sterile suction catheter inside a sterile overtube that is passed through the suction channel of the endoscope after the desired collection site is reached. The aspirates should be transferred immediately to an anaerobic transport vial and the contents plated for both aerobic and anaerobic organisms. [76] Significant bacterial overgrowth usually is associated with the detection of anaerobic organisms. Because of the technical difficulties of culturing fastidious anaerobic organisms, the presence of more than 10^5 colony-forming units per milliliter in the duodenum is considered diagnostic for bacterial overgrowth. [2] Analyses of unconjugated bile acids [77] and short-chain fatty acids [78] in the fluid also have been performed but are largely investigational tools. In addition to the technical difficulties involved in collecting the specimen properly, without contamination by mouth flora, and in transporting and culturing the fluid, an aspirate of the proximal bowel may not reflect distal bacterial overgrowth. [79]

TESTS	LIMITATIONS
Culture of small intestinal aspirate	Invasive, possible contamination with oral flora, difficulty in transport of anaerobic cultures, distal intestinal overgrowth can be missed
^{14}C-xylose breath test	Radioactivity, decreased sensitivity
H_2 breath tests	
Glucose-H_2	20% of population unable to produce H_2, decreased sensitivity
Lactulose-H_2	Distal intestinal overgrowth can be missed because of overlap between early and late peak

TABLE 78-4 Diagnostic Tests for Bacterial Overgrowth

Breath tests were devised as a less invasive alternative to intubation of the proximal bowel. These tests measure the breath excretion of carbon dioxide or hydrogen produced during the intralumenal bacterial metabolism of an administered substrate (see Chapter 42). The bile acid or ^{14}C-cholylglycine breath test was one of the first breath tests developed, but it cannot distinguish bacterial overgrowth from ileal malabsorption. [80] Because of its poor sensitivity and specificity, this test largely has been abandoned. A radioactively labeled pentose, ^{14}C-D-xylose, is a more ideal substrate for breath testing because it is minimally metabolized by the host after absorption. Xylose is absorbed in the proximal bowel, whereas bile acids are absorbed in the ileum, so the chance of obtaining a false-positive measurement as a result of colonic metabolism is reduced. [19] The sensitivity of this test in comparison with that of microbiologic culture ranges from 30% to 100%. [31], [81] The specificity of the test appears to be high (89%–100%), and it is well tolerated. [31], [81] The use of stable isotopes in combination with mass spectrometry, such as the ^{13}C-xylose breath test, may become more widespread in the future, in an effort to avoid the administration of radioactive substrates. [82]

The hydrogen breath tests also have the advantage of not involving radioactive substrates. A major problem with these tests is that 15% to 20% of the human population harbor flora that does not produce hydrogen. [83] Lactulose and glucose are two substrates often used for the purpose of diagnosing bacterial overgrowth. Patients with bacterial overgrowth who are administered lactulose exhibit an early breath hydrogen peak from small intestinal bacterial fermentation, followed by a prolonged peak from colonic bacterial metabolism. [83] In a case of distal intestinal overgrowth, it may be difficult to resolve the two peaks. In a study from Burma comparing the 10-g lactulose-hydrogen breath test with the Enterotest string test, only 2 of 15 children with more than 10^5 organisms per milliliter had a positive breath test result. [84] A comparison of the glucose-hydrogen and lactulose-hydrogen tests showed sensitivities of 62% and 68%, respectively, and specificities of 83% and 44%, respectively, in comparison with jejunal culture. [85] An elevated level of fasting breath hydrogen has been observed in some patients with bacterial overgrowth, but this is a relatively insensitive indicator of bacterial overgrowth. [86] Analysis of unconjugated bile acids in the serum has been performed in patients with bacterial overgrowth and may provide a noninvasive test for bacterial overgrowth. [87]

In summary, the available diagnostic tests all have their limitations. Many of them are used primarily as investigational tools and are not widely available in institutions. Barium studies may be useful for documenting an anatomic defect or suggesting the presence of a severe motility disorder. In certain clinical situations, such as a need to document fat malabsorption or vitamin B_{12} malabsorption in the setting of an anatomic defect or severe motility disorder, it may be reasonable to consider a course of empiric antibiotic therapy without further confirmation of the diagnosis. In the absence of a documented predisposing abnormality of the gastrointestinal tract, however, it is recommended that further diagnostic testing be performed. Routine upper gastrointestinal endoscopy with small bowel biopsy and culture of an aspirate obtained by means of a sterile overtube [69] may be the most efficient means of confirming the diagnosis and excluding other causes of malabsorption, such as celiac sprue and giardiasis.

TREATMENT

Initial management consists of fluid and nutritional support, including the correction of vitamin deficiencies (Table 78-5). After bacterial overgrowth has been diagnosed, an attempt should be made to identify an underlying cause. Surgical correction of an anatomic cause of intestinal stasis should be considered. Severe motility disorders are more difficult to manage. The administration of low doses of octreotide to patients with scleroderma for 3 weeks cleared bacterial overgrowth, as demonstrated by a reversal of abnormal breath hydrogen excretion. [88] Octreotide stimulated intestinal motor activity in these patients. At higher doses, octreotide may cause steatorrhea secondary to hypomotility and bacterial overgrowth. [89] The prokinetic agent cisapride has been shown to normalize intestinal motility in patients with motor disorders [90] and has been efficacious in the treatment of bacterial overgrowth in patients with cirrhosis. [63], [91] The efficacy of motilin receptor agonists in the treatment of dysmotility-associated bacterial overgrowth remains to be tested. [92] Often, the underlying lesion cannot be corrected, and the primary treatment is directed at suppressing the bacterial overgrowth with antibiotics. Although numerous antibiotics have been reported to be effective, very few controlled trials have examined the selection of antimicrobial agents. In a crossover randomized trial, both norfloxacin and amoxicillin-clavulanic acid given for 7 days were effective in treating bacterial overgrowth–related diarrhea and in reducing the mean volume of breath-excreted hydrogen. [93] The mean daily number of stools decreased within 1 to 3 days, but diarrhea often recurred 6 days after antibiotics had been discontinued. Pouchitis in ileal reservoirs, in patients with ulcerative colitis, is associated with

overgrowth of anaerobes and responds to metronidazole. [94]

Nutritional Supplementation
Increase calorie intake
Correct micronutrient deficiencies (e.g., vitamin B_{12})
Correction of Intestinal Stasis
Surgery (e.g., resection of strictures)
Prokinetic agents (e.g., octreotide in scleroderma)
Antibiotic Treatment

TABLE 78-5 Treatment of Bacterial Overgrowth

In a number of patients, symptoms recur after cessation of therapy. For these patients, it may be necessary to rotate antibiotic regimens. Probiotic therapy may be useful in prolonging remission after antibiotic therapy. Probiotics may also exhibit some efficacy as primary therapy in the treatment of small bowel bacterial overgrowth, but the data are less convincing. Treatment with *Saccharomyces boulardii,* a probiotic agent, for 7 days showed no efficacy in one trial. [93] Probiotic therapy with *Lactobacillus plantarum* 299V and *Lactobacillus* GG has been reported helpful in treating children who have small bacterial overgrowth associated with short bowel syndrome. [95] The administration of a high dose of a probiotic preparation (5×10^{11} viable lyophilized bacteria per gram: four strains of lactobacilli, three strains of bifidobacteria, and one strain of *Streptococcus salivarius* subspecies *thermophilus*) to patients in remission from chronic pouchitis significantly reduced the rate of relapse. [96]

Because some patients with chronic underlying gastrointestinal disease respond dramatically to a brief course of antibiotic treatment, bacterial overgrowth is an important entity to consider in their management.

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CHAPTER 79

Richard N. Fedorak

SHORT BOWEL SYNDROME

The term *short bowel syndrome* covers the symptoms and pathophysiological disorders associated with a malabsorptive state resulting from the removal of a large portion of the small or large intestine. The extent of intestinal resection that will produce this syndrome varies from one person to another. In children, survival without enteral supplements or total parenteral nutrition is generally possible if more than 40 cm of the small intestine (i.e., 20% of the normal length), measured distally from the ligament of Treitz, remains. In adults, survival without enteral or parenteral nutrition is generally possible if the residual length of the small intestine, measured distally from the ligament of Treitz, is more than 150 cm (i.e., 25% of the normal length). In general, if the ileocecal valve is removed, longer lengths of residual bowel may not prevent short bowel syndrome.

Short bowel syndrome refers only to a well-organized clinical pattern that is sometimes seen in patients with intestinal resection and is not necessarily related to the length of intestine removed. The clinical consequences of removing a portion of the small intestine are extremely variable and depend on a number of factors, including the extent of resection, the site of resection, and subsequent adaptive processes. Thus, it is possible that the removal of identical lengths of small intestine may cause short bowel syndrome to develop in one person but not in another.

Significant confusion exists regarding the actual length of the small intestine. Autopsy studies report the length of the small intestine as the distance from the pylorus to the ileocecal valve. Because it is difficult to measure the length of the duodenum, however, measurements made during operations often report the length of the small intestine as the distance from the ligament of Treitz to the ileocecal valve. The mean length of the small intestine at autopsy (from pylorus to ileocecal valve) has been reported as 630 cm in men and 592 cm in women; the mean colonic length is 150 cm in both sexes. These lengths are likely to be greater than the lengths measured in vivo because tone is lost in the postmortem state. Furthermore, small intestinal lengths probably differ according to racial and genetic factors. Full-term newborns have about 250 cm of small intestine and 50 cm of colon. The small intestine develops to its adult length by about 9 years of age.

Although clear functional distinctions exist between the proximal jejunum and the distal ileum, it is difficult to determine the exact normal ratio of jejunal length to ileal length because no clear demarcation exists between the two. In general, in North American adults, the proximal two fifths (~240 cm) of the small bowel are referred to as the *jejunum,* and the distal three fifths (~360 cm) are called the *ileum.* [1], [2]

ETIOLOGY

Adults

Several conditions are associated with short bowel syndrome in adults (Table 79-1). The most common of these are intestinal resection following a vascular insult to the small intestine (thrombosis or embolism of the superior mesenteric artery, thrombosis of the superior mesenteric vein, volvulus, strangulation) and Crohn's disease, with or without multiple intestinal resections. Less common causes of short bowel syndrome are jejunoileal bypass for obesity, abdominal trauma with resultant intestinal resection, inadvertent gastrocolic fistulization at the time of operation, radiation enteropathy, and neoplasms involving the gastrointestinal tract.

Vascular Insults
Thrombosis or embolism of the superior mesenteric artery
Thrombosis of the superior mesenteric vein
Volvulus
Strangulation
Postoperative
Jejunoileal bypass for obesity
Abdominal trauma necessitating bowel resection
Inadvertent gastrocolonic/ileal anastomosis
Miscellaneous
Crohn's disease, with or without surgical resection
Radiation enteropathy
Primary or secondary neoplasms involving the gastrointestinal tract

TABLE 79-1 Causes of Short Bowel Syndrome in Adults

Infants and Children

Most of the underlying causes of short bowel syndrome in children develop during intrauterine life (Table 79-2). Prenatal vascular accidents presenting as intestinal atresia or volvulus that result from malrotation are a particularly common cause of short bowel syndrome in this age group. Less common postnatal causes include vascular insults, surgery for necrotizing enterocolitis or Crohn's disease, and trauma.

Prenatal
Vascular insults
Intestinal atresia
Volvulus
Abdominal wall defects
Postnatal
Arterial thrombosis or embolism
Venous thrombosis
Necrotizing enterocolitis
Trauma
Crohn's disease
Volvulus
Hirschsprung disease
Radiation enteropathy

TABLE 79-2 Causes of Short Bowel Syndrome in Infants and Children

FACTORS INFLUENCING SHORT BOWEL SYNDROME[3], [4], [5] and [6]

Extent of Intestine Removed

Generally, the more extensive the resection of the small intestine, the greater the loss of absorptive surface area that transports nutrients, water, and electrolytes. It is clear, however, that the length of residual intestine and its function are more important prognostically than the extent of intestine removed. Short bowel syndrome can result from either a single massive intestinal resection or repeated lesser resections. Mesenteric vascular disease is more prevalent among patients who have undergone massive resection, whereas Crohn's disease is more prevalent in patients who have undergone repeated surgical resections leading to short bowel syndrome.[7], [8] Although the 30-day mortality rate is higher in patients who have had a massive resection, those surviving longer than 30 days have similar survival rates at 5 years.[8]

The amount of small intestine remaining after resection determines the surface area available for absorption, as well as the transit time and subsequently the time during which nutrients and nonnutrient fluids and electrolytes are in contact with the absorptive surface of the intestine.

About 50% (~300 cm) of the small intestine usually can be resected without the development of the significant nutrient or electrolyte losses that lead to short bowel syndrome. The removal of more than 75% (~450 cm) of the small intestine almost invariably results in fluid or electrolyte malabsorption that requires enteral or parenteral replacement therapy.

Site of Intestine Removed

Jejunum The jejunum, in its intact state, absorbs a significant fraction of the nutrients and water required by the body, but removal of the jejunum alone generally causes only a limited defect in the absorption of macronutrients, electrolytes, and free water. This limited effect is a consequence of the fact that the ileum is the part of the intestine with the greatest capacity for adaptation, and it is able to compensate for, and take over, almost all the absorptive functions of the jejunum.[9] Thus, jejunal resections are often reasonably well tolerated. Removal of the jejunum reduces the secretion of cholecystokinin and secretin, which may lead to subsequent decreases in biliary and exocrine pancreatic secretions; in turn, these decreases compromise nutrient absorptive capacity. In addition, cholecystokinin, vasoactive intestinal peptide, gastric inhibitory peptide, and serotonin, all of which are distributed mainly in the jejunum, inhibit gastric secretion. Thus, gastric hypersecretion is more likely to occur after resection of the jejunum.[10], [11] Increased serum gastrin levels and gastric hypersecretion also are seen more frequently after jejunal resection; these increases probably are also related to the length of bowel resected.[12], [13]

Ileum In the ileum, which is relatively impermeable in comparison with the jejunum, the solvent drag of water and electrolytes is less than in the jejunum; however, the ileum remains critically important in the active transport of both nutrients and nonnutrients. If the ileum is resected, the remaining colon is often incapable of handling fluids or adapting to prevent extensive fluid losses as the jejunum continues to pour large amounts of chyme into the impermeable residual ileum and nonabsorptive colon. For this reason, patients usually poorly tolerate extensive ileal resections. The ileum is the only site of active bile acid absorption from the intestine, although small amounts of bile acids are absorbed by passive diffusion in the jejunum and colon. Therefore, a loss of more than 100 cm of ileum usually precludes the active intestinal absorption of bile acids; thereafter, the passive absorptive capacity of the rest of the small intestine is rapidly overwhelmed, and bile acids are left in the lumen and spill into the colon.[5] These bile acids are deconjugated by colonic bacteria. The deconjugated bile acids directly stimulate the colon to secrete fluid and electrolytes, causing a secretory watery diarrhea and thereby aggravating the short bowel syndrome.[9], [10] Bile acids stimulate active anion secretion in the colon through several mechanisms: increasing cytosolic calcium levels within colonocytes through a calcium ionophore effect; increasing prostaglandin synthesis in lamina propria cells; stimulating excitatory enteric neurons, which then stimulate active anion secretion and propulsive motor contractions; and increasing tight junction permeability to fluid and electrolytes. During intestinal adaptation, the body compensates for bile acid loss into the colon by maintaining the size of the bile acid pool through an up to eightfold increase in hepatic bile acid synthesis. The more extensive losses of ileum, however, result in severe bile acid malabsorption wherein loss exceeds synthesis. A reduction in the circulating pool of bile salts follows, leading to impaired intestinal micelle formation and the spillage of fat (steatorrhea) into the colon. Fat within the colon is hydroxylated by bacteria into hydroxy fatty acids. Hydroxystearic acid is the hydroxy acid most often associated with steatorrhea, and it has stimulatory effects on colonic fluid and electrolyte absorption. Hydroxy fatty acids stimulate net secretion through several mechanisms: changing net absorption to net secretion, increasing mucosal permeability, and increasing propulsive motor activity. These effects are similar to those of bile acids and may be related to the detergent properties of both hydroxy fatty acids and bile acids. The ileum is the primary site of vitamin B_{12} absorption. Malabsorption of vitamin B_{12} occurs after the resection of more than 60 cm of ileum. The intestine appears to be largely unable to recruit new vitamin B_{12} receptors into residual ileum or jejunum. Resection of the distal ileum also affects nutrient absorption in the jejunum. A 50% distal ileal resection in one rat model decreased transmucosal jejunal calcium absorption by unknown mechanisms that did not appear to involve the brush border membrane.[14] Whereas the mucosal morphologic and functional absorptive response to distal ileal resection has been well documented, little is known about the motor response. In dogs, initial motor responses to resections of the distal small intestine of various lengths were dominated by the development of abnormal motility patterns, with prominent cluster activity noted in the intestinal remnant.[15] The intact ileum markedly slows intestinal transit[16]; thus, ileal resection decreases the intestinal transit time significantly, impairing the ileal brake phenomenon thought to be controlled by peptide YY.[17]

Colon Throughout the intestine, motility is slowest and absorptive capacity is highest in the colon, and absence of the colon significantly increases the rate of transit and the liquidity of stools. The primary function of the colon is to absorb the 1 to 2 L of fluid it receives from the ileum. The liquidity of the feces and the extent of fluid and electrolyte loss in the colon depend on the total volume of fluid and its rate of arrival at the cecum, and also on the presence of unabsorbed solutes such as bile salts. The removal of both ileum and colon obviously drastically increases fluid loss, dehydration, volume depletion, hypocalcemia, and hypomagnesemia. Preservation of the colon in cases of major small intestinal resection appears to lessen the severity of short bowel syndrome because the colon not only provides an absorptive surface area but also exerts a braking effect on early gastric emptying.[17] Preservation of even 50% of the colon is associated with significantly lower morbidity and mortality rates.[18] Although retention of the colon improves fluid and electrolyte absorption, colonic conservation also can have maladaptive consequences. Colonic bacteria deconjugate and hydroxylate unabsorbed bile salts and fats, respectively, creating solutes (bile acids and fatty acids) that limit water absorption in the colon through direct stimulation of the secretion of colonic fluid and electrolytes and through osmotic effects. In addition, the combined effects of massive small bowel resection, fat malabsorption, and an intact colon lead to the formation of calcium oxalate renal stones as free fatty acids in the colonic lumen preferentially bind calcium, leaving oxalate unbound and free to be absorbed systemically through the colonic mucosa.

Presence of an Ileocecal Valve

The ileocecal valve functions to prolong the intestinal transit time, thereby increasing the duration of contact of the lumenal contents with the small intestinal mucosal surface and enhancing the absorption of nutrients, fluids, and electrolytes.[5], [6] If the ileocecal valve is removed during an intestinal resection, the likelihood of the development of short bowel syndrome, often in a severe form, is increased.[18], [19] In an outcome study of neonatal short bowel syndrome, the presence of an ileocecal valve was not significantly correlated with successful weaning from parenteral nutrition, but it did reduce the mean duration of parenteral nutritional therapy from 20 to

7 months. [20] In addition, the ileocecal valve serves as a barrier that prevents the migration of lumenal colonic microorganisms into the distal small bowel. [10], [21] Removal of the ileocecal valve may result in bacterial overgrowth within the small intestine. These bacteria deconjugate bile salts within the small intestinal lumen; impaired micelle formation follows, so that the absorption of fat and fat-soluble vitamins is reduced. Furthermore, deconjugated bile salts spill into the colon and directly stimulate the colonic secretion of fluid and electrolytes, further aggravating the short bowel syndrome. Bacterial overgrowth associated with enteritis in children with short bowel syndrome prolongs the need for parenteral nutrition. [22] Finally, the bacteria use intralumenal vitamin B_{12} for their own metabolic purposes, limiting the availability of vitamin B_{12} for absorption by the host and exacerbating vitamin B_{12} deficiencies.

Intestinal Adaptation

Jejunum and Ileum Following resection of a major portion of the small bowel in experimental animal models, adaptive changes affect all layers of the remaining intestine. Lumenal dilation with thickening and lengthening of the gastrointestinal tract develop in association with hyperplasia of the crypt-villus axis, and the mucosal surface area increases significantly. [23], [24], [25], [26], [27] and [28] These effects are more noticeable in residual ileum (i.e., after jejunal resection) than in residual jejunum (i.e., after ileal resection). The number of cells in the proliferative zone of the crypts increases, and these cells rapidly migrate up the villi, causing villus hyperplasia and a parallel increase in the production of brush border membrane enzymes. This increase in mucosal mass is confirmed by increases in mucosal wet and dry weight, protein, and RNA and DNA contents. [29] An attempt to identify the molecular mechanism behind the observation of ileal hyperplasia in the rodent model of short bowel syndrome demonstrated the expression of a variety of nutrient absorptive and heat shock proteins in the residual intestine early in the adaptive process. [30] In addition to morphologic changes, an up-regulation of specific functional absorptive activity occurs in these animal models. [31], [32] The absorption of glucose, sucrose, bile acids, vitamin B_{12}, and calcium is increased in animal models of short bowel syndrome. [33] This increase is related to an increase in the number of absorptive cells rather than to an increase in the capacity of individual cells. [28], [34] In contrast to the plethora of findings in animal models of short bowel syndrome, few data are available regarding functional absorptive adaptation following the resection of human intestine. Dowling and Booth [35] reported an increase in jejunal glucose absorption in patients with short bowel syndrome that was proportional to the amount resected; furthermore, intestinal adaptive responses in humans most often required more than 1 year to reach maximum development. [10], [12], [13], [35]

Colon The colon also undergoes adaptive dilation, lengthening, and mucosal proliferation. These forms of adaptation result in an increased capacity to absorb fluid and electrolytes. [36] Several animal studies suggest that the colon also may be able to up-regulate the absorption of glucose and amino acids, if only to a limited extent. [12], [29]

Mechanisms of Intestinal Adaptation

Direct Effect on Epithelial Cells Enteral nutrients appear to stimulate intestinal adaptation through at least three major mechanisms: direct contact with epithelial cells, stimulation of trophic gastrointestinal hormone secretion, and stimulation of pancreatic and biliary secretions trophic to the small intestine. [12] Again, most experiments on potential stimuli and mechanisms of intestinal adaptation focus on animal, rather than human, models. Evidence that intralumenal nutrients have a direct effect on intestinal adaptation comes from animal studies in which segments of ileum were transposed into the proximal jejunum. The mass of ileal mucosa in this position increased markedly, and transposed villi became larger than those in the adjacent jejunum. In these experiments, the concentration of intralumenal nutrients, rather than simple osmolality, stimulated lengthening of the relatively short ileal villi. [5], [6] Without exposure to intralumenal nutrients, intestinal adaptation did not occur; rather, hypoplasia developed. [5], [6] In addition, nonnutritive agents did not stimulate mucosal growth, implying that the absorption or metabolism of lumenal nutrients is necessary for the process of adaptation to take place. [5], [37] The extent to which enteral nutrients stimulate intestinal adaptation also depends on the type and complexity of the nutrient administered. [37], [38] Disaccharides are more potent stimulants of adaptation than monosaccharides. [38] In addition, a nutrient need not always be metabolized for it to induce adaptation: 3- *O*-methyl-D-glucose, a nonmetabolizable glucose analog transported by the sodium/glucose cotransport system, stimulates intestinal adaptation in a rat model of short bowel syndrome. [39], [40] Fats that are highly unsaturated appear to be more effective in inducing intestinal adaptation than fats that are less unsaturated. [41] In animal models of short bowel syndrome, morphologic and functional intestinal adaptation following intestinal resection generally does not occur in the absence of enteral nutrition or oral feeding; in fact, a significant degree of mucosal atrophy results if all nutrients are provided parenterally. [12], [29], [42] Whether it is appropriate to extrapolate findings in rodent models to human short bowel syndrome has been questioned (see below). [40], [43] Jejunal biopsy specimens taken from eight normal volunteers before and after 14 days of parenteral nutrition therapy demonstrated decreased villus height, a decreased villus cell count, and increased intestinal permeability during parenteral nutrition, but no changes in DNA, RNA, and protein content in comparison with controls, suggesting that the total enterocyte number did not change. [44], [45] In a study of similar design, parenteral nutrition did not affect immunoglobulin-producing lymphocytes in the lamina propria, intestinal permeability, or bacterial translocation. [46] These results suggest that, in contrast to the findings in animal models, depriving the human intestinal lumen of enteral feeding does not lead to intestinal hypoplasia or immune dysfunction. Additional human studies of longer duration and with functional absorptive measurements must be conducted before the direct effects of enteral feeding on intestinal adaptation, identified in the rodent models of short bowel syndrome, can be confirmed. [47]

Stimulation of Trophic Gastrointestinal Hormones The second mechanism by which enteral nutrients stimulate intestinal adaptation is stimulation of the secretion of trophic gastrointestinal hormones. Thiry-Vella fistula models have been used to demonstrate this mechanism of action. Thiry-Vella fistulae are segments of bowel in which the blood supply is intact; however, because of the creation of proximal and distal ostomies, they are excluded from the flow of enteric contents. Animals fed orally after the creation of such a fistula exhibit greater adaptation in both intact intestine and in the Thiry-Vella fistula than do animals fed intravenously. [48], [49] and [50] Several different hormones are under consideration as candidate trophic hormones. Gastrin appears to have a trophic effect only in the duodenum. Secretin and cholecystokinin stimulate intestinal mucosal growth; however, their effects seem to be exerted through their actions on biliary and pancreatic secretions rather than through direct trophism. Other hormones, including corticosteroids, prostaglandins, epidermal growth factor, enteroglucagon, and growth hormone–releasing factor, appear to stimulate epithelial cell proliferation in the small intestine. [28], [51], [52], [53] and [54] The role of these hormones in the intestinal adaptation that occurs following intestinal resection remains to be clarified.

Gastrin. Gastrin produces mucosal hyperplasia in the rat stomach and duodenum. When the major source of endogenous gastrin is removed by antrectomy, the resulting gastric and duodenal hypoplasia can be prevented through pentagastrin treatment. Gastrin does not promote adaptive changes in the jejunum or ileum. Furthermore, the relationship between gastrin levels and gastroduodenal adaptation is variable. [55]

Cholecystokinin. Cholecystokinin exerts a consistent trophic action on the small intestine. When infused into dogs or rats maintained on total parenteral nutrition, cholecystokinin completely prevented villus hypoplasia in one study, [56] but other investigators have been unable to substantiate this effect. Cholecystokinin probably exerts no direct trophic effect on the intestinal mucosa; instead, it likely influences the adaptive process by stimulating pancreatic or biliary secretions. [56]

Enteroglucagon and polyamines. Enteroglucagon-producing cells are found primarily in the terminal ileum. Polyamines (putrescine, spermidine, spermine) are found in all tissues of the body and directly stimulate tissue growth. Both hormones increase RNA and DNA synthesis, which results in epithelial hyperplasia. [50], [57], [58] Blocking polyamine synthesis negatively affects the adaptation process, whereas blocking polyamine degradation increases intestinal adaptation. [58], [59] Enteroglucagon-producing cells are stimulated by carbohydrates and long-chain triglycerides to release preformed enteroglucagon, which stimulates ornithine decarboxylase, increasing polyamine production and, thus, cellular hyperplasia and intestinal adaptation. Changes in enteroglucagon levels correspond closely to changes in the crypt cell production rate, and this peptide remains a favorite candidate for identification as the humorally mediated trophic influence on the small bowel. [55], [60] Evidence suggests that precursors of enteroglucagon, such as proglucagon-derived peptides, may be responsible for some of the activities previously attributed to enteroglucagon. Ileal proglucagon messenger RNA levels increase rapidly after intestinal resection. [61] Glucagon-like peptide 2, a product of proglucagon, is capable of inducing marked villus hyperplasia within 4 days after administration, and this change provides compelling evidence that proglucagon plays a significant role in the growth and adaptation of intestinal epithelium. [62], [63] and [64] The mechanisms responsible for the up-regulation of proglucagon in short bowel syndrome are not yet known; however, the fact that dietary fiber fermentation modulates intestinal proglucagon expression may explain why intestinal adaptation requires oral intake. [65] One clinical study demonstrated elevated fasting and meal-stimulated plasma levels of glucagon-like peptides 1 and 2 in patients with a resected ileum and a preserved colon who had short bowel syndrome. [66] In a separate study, the meal-stimulated increase in plasma glucagon-like peptide 2 in patients with short bowel syndrome was smaller than that in control patients. [67] Although these findings in patients may simply represent an epiphenomenon of glucagon-like peptides, the results of previous animal studies suggest that these peptides do indeed play a significant role in the intestinal adaptation of short bowel syndrome (see section “Medical Management: Glucagon-like Peptide 2 Therapy,” Page 1636).

Somatostatin. Somatostatin reduces the crypt cell production rate and hyperplasia in the small intestine after resection; it also inhibits the rise in enteroglucagon levels that normally follows jejunal resection. [68] Therefore, somatostatin may modulate mucosal cell turnover by influencing glucagon release. Somatostatin causes a number of competing effects on the intestine. It increases intestinal transit time and inhibits gastric secretions, both of which would benefit the patient with short bowel syndrome; in contrast, somatostatin-induced reductions in pancreatic and biliary secretions can lead to frank fat malabsorption and steatorrhea.

Neurotensin. In rats with 80% of their intestine resected, the administration of exogenous neurotensin increased villus length in both jejunum and ileum. Increases in circulating enteroglucagon found after the administration of neurotensin suggest that the trophic effect of neurotensin may be mediated, at least in part, by enteroglucagon. [69]

Growth factors. Epidermal growth factor (EGF), a potent stimulant of gastrointestinal epithelial proliferation, stimulates DNA synthesis and polyamine synthesis in the small intestine of experimental animals. [70], [71] In animals with 70% to 80% of their intestine resected, EGF receptor activity was increased, and EGF augmented not only intestinal size but also expression of the sodium/glucose cotransport carrier. [71], [72], [73], [74] and [75] Insulin-like growth factors also stimulated intestinal adaptation after small intestinal resection in the rat. [76], [77] Insulin-like growth factor type I transgenic mouse models have been used to demonstrate that insulin-like growth factor regulates intestinal mass and augments colonic fluid and electrolyte absorption. [64], [78], [79] Nevertheless, no evidence is available to support any beneficial effects of the administration of EGF at the time of resection, or to support the use of EGF in well-adapted short bowel syndrome, [73] which suggests the existence of an optimal window for growth factor augmentation of adaptation.

Prostaglandins. Prostaglandins of the E_2 class stimulate mucosal hyperplasia in the proximal small bowel of rats. [74], [80] Hollworth and colleagues, [75] however, found that prostaglandins have a strong trophic effect not only in the small bowel but also in the cecum and colon.

Glutamine. Glutamine is an important fuel for enterocytes, colonocytes, and other rapidly dividing cells. [81] In humans who have undergone intestinal resection, however, the turnover of glutamine is markedly diminished. [82] Glutamine appears to play an important role in maintaining the structural integrity of the small intestine. In animal studies, glutamine-enriched parenteral nutrition prevented the deterioration of gut permeability and preserved mucosal structure. [83], [84] In contrast, rat enteral diets supplemented with 2% glutamine had no beneficial effect on intestinal adaptation following massive small bowel resection. [85] In three human subjects with short bowel syndrome, intestinal use of glutamine was increased during adaptive hyperplasia following intestinal resection. [86], [87] Glutamine is unstable, however, and it is not generally available in parenteral form except as alanylglutamine. Furthermore, to be effective, glutamine must be administered in pharmacological doses of at least 0.5 g/kg daily. The requirements for large doses of glutamine and the variable results have raised the question of whether the role of glutamine alone, as a fuel for the enterocyte, has been exaggerated. In initial studies, the intestinal metabolism and oxidation of lumenal glutamate and glutamine were identical, with 60% of the carbon atoms of both precursors oxidized to carbon dioxide, which led to the hypothesis that perhaps both glutamate and glutamine are important amino acids in intestinal adaptation. [88] In an infant swine model in vivo, glutamate rather than glutamine appeared to be the preferred source for mucosal glutathione synthesis [89], [90] and a preferred substrate for intestinal proline synthesis. [91] Taken together, these results highlight the relative importance of enteral and systemic glutamate and glutamine as key precursors for the intestinal synthesis of amino acids, which in turn may become essential during conditions of short bowel syndrome. However, no controlled human clinical trials have thus far demonstrated that glutamate or glutamine supplementation directly enhances morphologic or functional improvement in the small intestine of patients with short bowel syndrome. [92]

Arginine. The intestine is an important site for the synthesis of citrulline, which then enters the circulation to act as the primary precursor for renal arginine synthesis. Dietary arginine thus becomes nutritionally indispensable in rats with massive small intestinal resection. [93] The results of studies in pigs suggest that intestinal enterocytes are capable of turning on the synthesis of arginine, which in turn induces intestinal polyamine synthesis for maintenance and growth of the intestinal mucosa. [94], [95], [96], [97] and [98] Thus, dietary and synthesized amino acids may play an important role in intestinal adaptation following intestinal resection.

Short-chain fatty acids. Acetate, propionate, and butyrate are the principal short-chain fatty acids produced by bacterial fermentation of the carbohydrates and fibrous polysaccharides that spill into the colon. [99] These short-chain fatty acids are absorbed by the colon and metabolized in the colonic epithelial cells as a source of fuel; carbon atoms are salvaged that would otherwise be lost. It has been estimated that up to 3 kJ of energy can be absorbed each day by the human colon in the form of short-chain fatty acids. [100] Supplementation of an elemental diet with pectin, a fiber completely fermented to short-chain fatty acids in the colon, improved adaptation of the ileum, jejunum, and colon after massive small bowel resection. [101] Subsequent studies on the supplementation of parenteral nutrition with short-chain fatty acids [102] and the intracecal infusion of short-chain fatty acids [103] confirmed that supplementation reduces the mucosal atrophy and intestinal immune dysfunction [104] associated with total parenteral nutrition following massive small bowel resection in animal models. [105] In addition to their local effects, systemic short-chain fatty acids can affect the motility of both the stomach [106] and the ileum [107] through neuroendocrine mechanisms, likely through the expression of proglucagon and peptide YY. [108], [109] and [110] Furthermore, both systemic and enteral short-chain fatty acids exert a trophic effect on the jejunum [111] by increasing mucosal mass, DNA, and villus height. These morphometric effects were associated with an increase in amino acid absorption in a pig model of small bowel resection. [105] The effects are yet to be confirmed in human short bowel syndrome. It is important to remember that excess amounts of intralumenal short-chain fatty acids, amounts higher than the concentrations that can be absorbed by the colon, aggravate diarrhea by directly stimulating fluid and electrolyte secretion and by exerting an osmotic effect that limits water absorption from the colon.

Stimulation of Pancreatic and Biliary Secretions The third mechanism by which lumenal nutrients stimulate intestinal adaptation is the production and stimulation of pancreatic and biliary secretions. Nutrients that stimulate biliary and exocrine pancreatic secretions produce mucosal hyperplasia, especially when such nutrients are present in concentrations higher than usual in the residual ileum. Ileal mucosal growth is markedly enhanced when biliary and pancreatic ducts are transplanted to the residual ileum of otherwise normal rats. [28] The presence of food is likely essential for these gastrointestinal secretions to exert their trophic effect. [12]

CLINICAL MANIFESTATIONS

As a result of gradual intestinal adaptation, the clinical course of a patient with short bowel syndrome passes through a number stages. [112], [113] and [114]

The first stage usually lasts 1 to 2 weeks and is characterized by a period of substantial fluid and electrolyte loss resulting from overwhelming diarrhea. During this stage, fluid, electrolytes, and nutrition are usually supplied by the parenteral route. The intensity of the watery diarrhea generally decreases during the next several months. Oral intake and the subsequent gastric hypersecretion can aggravate fluid and electrolyte loss. Parietal cell hyperplasia, hypergastrinemia, and hypersecretion of gastric acid also can alter the duodenal pH and inactivate pancreatic lipase, contributing to steatorrhea.

The second stage is the period of intestinal adaptation, during which oral intake is initiated and gradually increased. Full or partial enteral or parenteral supplementation often is required to maintain optimal nutritive and fluid and electrolyte status. This stage may last from several months to longer than a year.

During the third stage, maximum intestinal adaptation is achieved, and a relatively normal oral intake is usually possible. Whereas some patients never reach the stage of full oral nutrition, life at home can continue with the patient supported by a combination of enteral or parenteral nutrients and electrolytes.

Diarrhea

Diarrhea is inevitable in patients who have undergone extensive small intestinal resection as a consequence of multiple factors: reduction of the absorptive surface area; decreases in intestinal transit time; humorally mediated gastric, small intestinal, and colonic hypersecretion; increases in the osmolality of colonic contents, followed by osmotic diarrhea secondary to malabsorption of fat and carbohydrates; bacterial overgrowth in the small intestine and associated structural damage to enterocytes (increased permeability, decreased brush border membrane function); and colonic hypersecretion, which results from increased levels of dehydroxylated bile acids and hydroxylated fatty acids associated with the spillage of bile acids and fat, respectively, into the colon, where bacteria metabolize them into their secretory solutes. Because the diarrhea in short bowel syndrome is complex, it is critical to evaluate its possible causes so that management is approached correctly.

Gastric Hypersecretion

Gastric hypersecretion secondary to hypergastrinemia occurs within 24 hours after intestinal resection and can result in excessive fluid and electrolyte loss, peptic ulcer disease, and compromised intestinal absorption. [115], [116] and [117] Absorptive compromise occurs as a consequence of reduced lipase activity (as a result of spillage of gastric acid into the duodenum and subsequent lowering of the duodenal pH), which leads to impaired micelle formation and reduced intralumenal lipid absorption. Gastric hypersecretion responds well to histamine receptor antagonists or proton pump inhibitors; furthermore, it almost always abates with time. The cause of gastric hypersecretion remains unclear because serum gastrin levels have been found to be both elevated [115] and unchanged [118] in patients with gastric hypersecretion following intestinal resection. Normal acid secretion also has been noted in patients with hypergastrinemia, and in some patients, hypochloriduria develops after intestinal resection.

Steatorrhea

Several mechanisms are responsible for the steatorrhea of short bowel syndrome. Following surgical resection, gastric hypersecretion can alter the pH in the duodenum and inhibit the action of pancreatic enzymes, particularly lipase (see section “ Gastric Hypersecretion”). In the case of ileal resection and loss of the bile salt–reabsorbing region of the intestine, depletion of the bile salt pool can lead to insufficient micelle formation and steatorrhea (see section “ Site of Intestine Removed”).

Carbohydrate Malabsorption

After extensive resection, carbohydrates pass from the small intestine into the colon, where they are metabolized by bacteria into short-chain fatty acids. These short-chain fatty acids (butyric acid, propionic acid, acidic acid) cause diarrhea through two mechanisms (see section “ Mechanisms of Intestinal Adaptation”). First they cause an osmotic diarrhea, and then they directly stimulate the colon to secrete fluid and electrolytes. Changes in the osmotic load and malabsorption of carbohydrates are probably the most important causes of diarrhea in short bowel syndrome. Increases in intestinal permeability and further carbohydrate and fluid malabsorption have been demonstrated in patients with short bowel syndrome who have episodes of sepsis [119] and may help explain the intermittent and rapid deterioration that sometimes occurs in patients with short bowel syndrome with seemingly minor infections.

In children with short bowel syndrome, the development of clinically significant D-lactic acidemia and D-lactic aciduria is well described and leads to increased serum levels of lactate, acidosis, and compensatory hyperventilation. [120] The D-lactic acid is derived predominantly from malabsorbed fermentable carbohydrates that are delivered to the colon, where lactobacilli abundantly metabolize them to D-lactic acid.

Protein Malabsorption

Protein is usually well tolerated as a caloric source in short bowel syndrome and contributes relatively little to the osmotic load. However, protein is metabolized by colonic bacteria in the same way as malabsorbed carbohydrates and can produce a similar osmotic type of diarrhea, although it is consumed in much smaller quantities than carbohydrates and so usually causes much less severe diarrhea.

Fluid and Electrolyte Malabsorption

Fluid and electrolyte losses can be enormous and are common during the first few weeks after intestinal resection. Daily fecal effluents larger than 5 L can be seen. It is during this time that prompt replacement therapy is important to prevent severe hypovolemia, hyponatremia, and hypocalcemia.

Vitamin and Micronutrient Deficiencies

Vitamin B_{12} deficiency is likely after ileal resection because the recruitment of vitamin B_{12}–intrinsic factor receptors into the jejunum is extremely limited. In addition, colonic or small intestinal bacteria can metabolize vitamin B_{12}, further aggravating vitamin B_{12} deficiency. Vitamin B_{12} deficiency (as well as folate deficiency) leads to hyperhomocysteinemia and has been associated with venous thrombosis in patients with short bowel syndrome. [121] Other water-soluble vitamins are well absorbed throughout the entire small intestinal tract and are generally well preserved except in the shortest intestines. [122], [123]

Mineral deficiencies, including those of calcium and magnesium, are common. They occur secondary to the malabsorption of fatty acids, which form soap complexes with these divalent cations. [122] The administration of vitamin D and calcium supplements is usually helpful in correcting calcium deficiency, but magnesium deficiency may be more difficult to manage because the enteral administration of most magnesium salts results in osmotic diarrhea. Magnesium gluconate is less likely to cause diarrhea than is magnesium oxide or magnesium sulfate. Calcium depletion can be exacerbated by malabsorption of vitamin D. Although calcium deficiency normally results in an increased secretion of parathyroid hormone, in patients with concomitant calcium and magnesium depletion, the release of this hormone is decreased because of the magnesium deficiency. [122]

Steatorrhea also is associated with the decreased absorption of fat-soluble vitamins, particularly vitamin D, but also vitamins A, K, and (rarely) E.

The absorption of trace elements, like that of water-soluble vitamins, is usually sufficient after intestinal resection. [122], [123] Clinical zinc deficiency resulting from diarrhea can develop fairly rapidly, however, and has been described to impair intestinal epithelialization and adaptation.

Systemic Manifestations

Renal dysfunction, both permanent and intermittent, can occur as a consequence of hypovolemia. This complication often develops soon after intestinal resection, but the fluid balance of some patients with short bowel syndrome is sufficiently precarious that short episodes of viral gastroenteritis can “tip” them into renal failure.

An increased incidence of renal calculi in association with ileal resection or ileal disease is well documented. [124], [125], [126] and [127] Hyperoxaluria is frequent in these patients, and the calculi are predominantly formed from calcium oxalate. [127] The hyperoxaluria appears to be secondary to an increased absorption of dietary oxalate, particularly in the colon. The increase involves two mechanisms. First, steatorrhea contributes to the increased absorption of oxalate through an increase in lumenal fatty acids, which preferentially bind to calcium, leaving free oxalate in the lumen that can readily be absorbed. Second, bile salts that enter the colon of these patients appear to increase the colonic mucosal permeability, thereby allowing increased oxalate absorption in the colon. [126], [127] In addition to hyperoxaluria, other mechanisms may be responsible for increases in renal calcium oxalate and calculi. [128] These include a decreased urinary concentration of calcium-binding anions, such as phosphate and citrate, and diminished urine flow.

Patients with short bowel syndrome are at significant risk for cholelithiasis if the length of the intestinal remnant is less than 120 cm, if total parenteral nutrition is required, or if the terminal ileum is resected. The incidence of gallstones increases threefold after ileal resection. [129], [130] Interruption of the enterohepatic circulation of bile and the subsequent bile salt malabsorption lead to an increase in the hepatic synthesis of cholesterol; bile becomes supersaturated with cholesterol, and gallstones develop. A significant number of patients with ileostomies have calcium-containing radiopaque gallstones rather than radiolucent cholesterol gallstones. [129] Some other mechanism, therefore, must be involved. It also appears that interruption of the intrahepatic circulation predisposes humans to an increase in pigment stones. [131], [132] and [133] With this understanding of the pathophysiology of gallstone formation, prophylactic cholecystectomy should be considered in all patients who have undergone extensive intestinal resection. [134]

In addition to the increased risk for cholelithiasis, the incidence of intrahepatic cholestasis and hepatic dysfunction is increased (65%) in patients with short bowel syndrome; these conditions are usually associated with parenteral nutrition therapy and sepsis. [135] Hepatic dysfunction, particularly in children with short bowel syndrome, results in liver failure, cirrhosis, and the need for liver transplantation.

An increased risk for infectious complications, mainly caused by the egress of lumenal intestinal bacteria to distant organs, a process termed *bacterial translocation,* has been described in pediatric, but not adult, patients with short bowel syndrome. [40], [136] In animal models of short bowel syndrome, rates of bacterial translocation were increased as a consequence of intestinal bacterial overgrowth in the residual bowel, and bacterial translocation was positively correlated with removal of the ileocecal valve. [137]

MEDICAL MANAGEMENT

Nutritional Management

The nutritional management of short bowel syndrome is a dynamic process that follows the evolution of the clinical and adapted state of the bowel. Depending on the length of resection and the postsurgical adaptation, the process will result in one of four nutritional outcomes: maintenance of a balanced nutritional status on a normal or modified oral diet, maintenance through the use of defined enteral formula diets, maintenance through enteral intake with parenteral electrolyte and fluid supplementation, or maintenance through total or partial parenteral nutritional intake supplemented by variable amounts of enteral intake. Whatever the source of nutrition, the caloric intake should be increased slowly and progressively until a target is reached of about 32 kcal/kg of ideal body weight per day. This targeted caloric intake, in general, will compensate for the increased losses that result from inefficient and inadequate absorption.

Parenteral Therapy In the immediate postoperative period, all patients who have undergone extensive small intestinal resection require total parenteral nutrition. Sodium losses in early secretions are often high (80–120 mmol/L), and electrolyte losses must be monitored carefully. As the amount of enteral nutrition is gradually increased, the duration and intensity of parenteral nutrition infusion can be reduced. Initially, parenteral nutrition can be stopped for one or two nights each week.

Eventually, administration every other night or every third night is possible. If more than 25% of the patient's intestine remains, it should be possible to stop total parenteral nutrition completely. Some patients, however, require parenteral therapy, particularly fluids and electrolytes, for extended periods. In a study of 135 patients with short bowel syndrome, small intestinal length (>35 cm with an ileojejunal anastomosis), preservation of the colon, and an ileocolonic anastomosis were the features most predictive of the ability to wean from parenteral nutrition. [138]

Oral Therapy A balanced solution containing carbohydrates and electrolytes can be given orally once stool output is less than 2 L daily. As previously noted, early oral feeding is important to intestinal adaptation. Clear fluid diets are not useful because their nutritional value is inadequate; they are also severely hyperosmolar and likely to provoke osmotic diarrhea (Table 79-3). Full fluid diets are also poorly tolerated because they contain lactose; most patients with short bowel syndrome cannot tolerate lactose. Because the tight junctions of the jejunum are extremely permeable, supplements high in glucose and low in sodium often promote sodium and water losses and aggravate short bowel syndrome. To optimize oral fluid and electrolyte absorption, a balanced oral replacement solution is necessary. A solution that contains an isosmotic sodium and glucose mixture takes advantage of the small intestinal sodium/glucose cotransport carrier to enhance salt and water absorption. These solutions can be purchased ready-made (Table 79-4) or mixed at home (Table 79-5). The patient should sip the solution at frequent intervals throughout the day to prevent nausea and to avoid passing a large bolus of fluid rapidly through the short intestine. [139] The oral replacement solution also can be used instead of parenteral electrolytes at a later stage to reduce parenteral requirements. Patients with short bowel syndrome, in general, require an oral intake with a higher sodium concentration. Those with less than 100 cm of residual jejunum are usually net sodium secretors [140] and can achieve an optimal sodium balance with an intake of 120 to 160 mmol/L per day, [139] whereas oral replacement solutions with lower sodium concentrations of 90 mmol/L do not maximize sodium balance. [141] Sports drinks like Gatorade are too low in sodium to be of use as oral replacement solutions in patients with short bowel syndrome. They are designed to replace fluid losses and prevent dehydration from perspiration, not intestinal efflux.

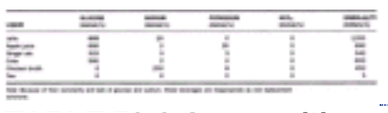

TABLE 79-3 Composition of Some Common Household Beverages

TABLE 79-4 Commercially Available Oral Replacement Solutions

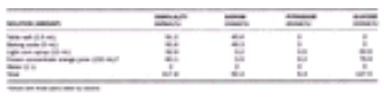

TABLE 79-5 An Isosmotic Oral Replacement Solution That Can Be Mixed at Home

For patients with more than 60 to 80 cm of small bowel, oral feeding should be reintroduced slowly until a normal or modified oral diet is reached. Because gastric emptying is slower for solids, they should consume dry solids at meals and take only isotonic fluids 1 hour later; this regimen improves the absorption of nutrients. Diarrhea that occurs as a consequence of oral feeding usually can be managed with an antidiarrheal agent, which should be taken on a regular basis 1 hour before meals and snacks. Waiting to take an antidiarrheal agent until after a meal has been started and diarrhea has occurred is not effective (see section " Management of Diarrhea"). Care must be taken to limit the amount of lactose in the diet because the initial disease, gastric hypersecretion, or surgical therapy can cause lactose intolerance. It may be useful to test the lactose tolerance of patients to provide them with specific guidelines about lactose intake. The lactose tolerance of patients with short bowel syndrome and a jejunocolonic anastomosis is greater than that of patients with a terminal jejunostomy; the lactose provided in yogurt is generally better tolerated than the lactose in milk. [142] Patients with significant ileal resection and an intact colon in whom steatorrhea is expected to develop may require monitoring of their oxalate secretions (see section " Systemic Manifestations"). A low-oxalate diet is difficult to follow, but guidance can be provided regarding which foods are high in oxalate. Oral calcium decreases oxalate absorption because insoluble salts form. A 4-g dose of cholestyramine given three times daily reduces bile salt–induced diarrhea and also binds intralumenal oxalate, further decreasing oxalate absorption. Attention to vitamin and mineral supplementation is critical. In general, a multiple-vitamin preparation with minerals will suffice (see section " Vitamin and Mineral Supplementation"). For patients with ileal resection, vitamin B_{12} should be administered once monthly. Traditionally, low-fat diets (often with medium-chain triglycerides) and high-carbohydrate diets have been sed to treat patients with short bowel syndrome. This practice was based on an understanding that steatorrhea has many adverse effects on fluid absorption, that dietary fat induces the secretion of gastrointestinal hormones, and that carbohydrates malabsorbed in the small bowel undergo fermentation in the colon to short-chain fatty acids. These short-chain fatty acids are easily absorbed across the colonic mucosa, so that carbohydrate energy is salvaged that would have otherwise been lost in the feces. In one study, a high-fat diet had the same effect on ileostomy volume output as a low-fat diet. [143] In this study, although steatorrhea from the ostomy increased with the high-fat diet, the proportion of fat absorbed was equivalent to that absorbed from a low-fat diet, suggesting that net absorption is more important than whether the regimen is high in fat. [144], [145], [146] and [147] In contrast, in patients without an ileostomy and with an intact colon, oral fat intake often is limited by the amount of fat that reaches the colon and is subsequently metabolized to hydroxy fatty acids, which in turn lead to secretory and osmotic diarrhea. Divalent cations, including calcium, magnesium, zinc, and copper, may bind to fatty acids in the stool, and excessive fecal losses of these minerals have been documented in patients with steatorrhea. [143], [146] Steatorrhea also exacerbates malabsorption of the fat-soluble vitamins A, D, E, and K. Increased fecal losses of fat enhance dietary oxalate absorption, oxaluria, and renal stone formation. Recommendations regarding dietary fat intake therefore must be based on the individual patient's symptoms following bowel resection. In addition, it is important to balance the beneficial effects of fat restriction against the reduced food palatability and caloric intake associated with low-fat diets. A low-fat and low-oxalate diet, for instance, is likely to be completely unpalatable. Medium-chain triglycerides can be used as caloric supplements for patients with short bowel syndrome. Medium-chain triglycerides are hydrolyzed in the intestinal lumen to water-soluble components, which are absorbed in the absence of bile salts. [148] These medium-chain fatty acids do not require resynthesis in the enterocyte and can be absorbed directly into the portal circulation. Thus, medium-chain triglycerides may serve as a nutritional source for patients with significant long-chain triglyceride malabsorption. However, medium-chain triglycerides have an unpleasant taste, their osmotic load in the proximal small intestine sometimes causes diarrhea (in doses of more than 35 g/d), and they do not provide essential fatty acids. Furthermore, they have a slightly lower caloric density than long-chain fatty acids (8.3 kcal/g compared with 9.0 kcal/g). Low-fiber diets remain controversial. Their use is based on observations that some types of dietary fiber, such as wheat bran, increase intestinal transit times and fecal weight. [149] It is now clear, however, that dietary fiber has variable effects on the intestine. [150] Soluble fibers such as pectin do delay gastric emptying and increase intestinal transit time. [151], [152] and [153] They also bind bile salts and have a significant water-retaining capacity. In addition, bacterial fermentation of fiber in the colon can produce short-chain fatty acids. In high concentrations, the short-chain fatty acids cause moderate diarrhea and colonic secretion, but in low concentrations, they actually may provide fuel for colonocytes and help to heal and prevent inflammation.

Enteral Therapy Patients who cannot tolerate a normal oral diet and those with a short bowel (<60–80 cm) often benefit from an enteral formula. If a chemically defined formula is used, it is important to control the rate of infusion to match osmotic inflow with osmolar absorption. Rates of full-strength infusion usually begin at 25 mL/h and gradually increase to 125 mL/h. The rate is modulated according to the tolerance displayed by the small intestine. If a polymeric formula is used, it should be lactose free because most patients with a massive small intestinal resection do not have an adequate reserve of lactase. Elemental or semi-elemental diets require a minimal absorptive surface. [112], [154] These diets may be useful for persons with a short gut during the early adaptive stage [155]; they also decrease the secretion of digestive juices [156] and gastric hypersecretion. Nevertheless, significant evidence suggests that elemental diets are not absorbed more readily than polymeric diets. [144], [157], [158] Elemental or semi-elemental formulas contain amino acids in a free form as well as dipeptides and tripeptides. Because separate carrier mechanisms exist for free amino acids and dipeptides, a mixture of amino acids and dipeptides results in increased protein absorption. [159], [160] and [161] Many enteral feeding formulations have a high content of carbohydrates in the form of sucrose or glucose polymers. These high levels lead to carbohydrate malabsorption and spillage of carbohydrates into the colon, with resultant osmotic diarrhea. Elemental formulas are low in fat, often providing the minimal amounts to meet essential fatty acid requirements. The formulas are effectively absorbed within the first 100 cm of jejunum [162] and stimulate adaptation of the proximal small intestine. [163] The unpleasant taste of elemental or semi-elemental formulas, however, often limits oral intake, and because they are hyperosmolar, they may considerably worsen diarrhea. The formulas therefore should be provided by slow, constant infusion. Polymeric diet supplements generally provide about 30% of calories as fat and contain intact protein sources. They are more palatable than elemental diets; they are also less hyperosmolar and considerably less expensive. Patients with short bowel syndrome absorb less than 100% of oral calories and protein. Many of them therefore need to consume calories and protein in excess of the normal estimated requirement. [164] Nevertheless, overfeeding

must be avoided carefully because it can induce a loss of endogenous fluid and electrolytes into the gut. Excess consumption of fats, fiber, concentrated sweets and sugar, milk and milk products, caffeine, and alcohol may increase normal outputs. [165], [166], [167] and [168] High enteral loads of carbohydrate should be avoided. Alterations of the bacterial flora in the colon, particularly overgrowth of *Lactobacillus acidophilus,* may result in the excessive production of D-lactate from unabsorbed carbohydrates, and severe metabolic acidosis and encephalopathy have been described. [169], [170] and [171]

Vitamin and Mineral Supplementation

In general, vitamin and mineral supplements are included in enteral and parenteral solutions. As patients are weaned from these solutions, and once they have stopped them completely, vitamin and mineral deficiencies may slowly develop because of inadequate intake (as patients try to prevent diarrhea), excess nutrient losses (from the short bowel), or a combination of both. Table 79-6 presents an example of a liquid vitamin and mineral oral supplementation regimen that could be considered for some patients as they are weaned from parenterally supplied vitamins and minerals. Liquid supplements are preferable to solid pills because hard outside matrices often do not dissolve during rapid transit through a short bowel.

ADEKs multiple vitamin Four tablets per day, chewed or crushed throughly; each tablet contains 4000 IU of vitamin A, 3 mg of β-carotene, 400 IU of vitamin D, 150 IU of vitamin E, 0.15 mg of vitamin K, 60 mg of vitamin C, 0.2 mg of folic acid, 1.2 mg of thiamin, 1.3 mg of riboflavin, 10 mg of niacin, 1.5 mg of vitamin B_6, 12 μg of vitamin B_{12}, 50 μg of biotin, 10 mg of pantothenic acid, and 7.5 mg of zinc.
Calcium gluconate liquid 400 mg PO twice daily = 448 mg of elemental calcium twice daily
Ferrous sulfate liquid 300 mg PO twice daily = 60 mg of elemental iron twice daily
Vitamin D_2 capsule 50,000 IU once per week
Phosphate-Novartis (tablet dissolved) 500 mg PO twice daily = 500 mg of elemental phosphorus twice daily
Potassium liquid 60 mL PO twice daily = 40 mEq of potassium chloride twice daily
Magnesium glucoheptonate liquid 30 mL PO twice daily = 150 mg of magnesium twice daily
Follow-up blood work (every 4 to 6 months): vitamins A, B_{12}, D, and E; prothrombin time and International Normalized Ratio; zinc, magnesium, phosphorus, calcium, albumin, red blood cell folate, iron, total iron-binding capacity, ferritin; complete blood cell count; electrolytes.

Note: Exact doses will vary depending on the requirements of individual patients.

TABLE 79-6 An Oral Vitamin and Mineral Supplement for Use in Selected Patients with Short Bowel Syndrome

Bile Supplementation

Bile acid replacement with oral conjugated bile acids (2 g per meal in the form of ox bile or synthetic cholylsarcosine supplementation) did not worsen diarrhea in a patient with an ileostomy, and it did cause fat absorption to increase by 40 g/d and eventually led to a significant weight gain. [172] This carefully conducted balance study suggests that conjugated bile acid replacement therapy can be considered for some patients with short bowel syndrome.

Management of Diarrhea

In general, the cause of the diarrhea must be identified before appropriate treatment can begin (e.g., bile acid–induced diarrhea, steatorrhea, osmotic diarrhea, bacterial overgrowth; see Chapter 42 and Chapter 78). Nevertheless, in the absence of sufficient intestinal adaptation, debilitating diarrhea develops simply because the intestinal surface area is inadequate for absorption to take place, and antidiarrheal therapy becomes an integral part of the management of short bowel syndrome.

Opiates are the most widely used products in the treatment of diarrhea associated with short bowel syndrome. Clinically available opiates include natural substances, such as paregoric and opium alkaloids, and synthetic preparations, such as codeine, diphenoxylate (Lomotil), and loperamide (Imodium). These agents relieve diarrhea by reducing intestinal secretion or by promoting intestinal absorption and reducing intestinal motility. For the diarrhea of short bowel syndrome, these agents should be taken on a regular basis 1 hour before meals, as the liquid formulation when available: 30 to 150 mg of codeine daily, 7.5 to 20 mg of diphenoxylate daily, or 6 to 20 mg of loperamide daily. As-needed dosing, rather than regular dosing, often results in unnecessary diarrhea and loss of the nutrient value of a meal before the opiate has had time to exert a therapeutic effect.

a $_2$-Adrenergic agonists are potent stimulators of intestinal absorption. These agents stimulate sodium and chloride absorption and inhibit bicarbonate and chloride secretion in the colon and small intestine. In addition, a $_2$-adrenergic agonists reduce intestinal motility. A 0.1- to 0.2-mg dose of clonidine, taken with each meal, can act as an adjunct to opiates, helping to prevent the difficult-to-control diarrhea of short bowel syndrome. [173], [174] Despite the fact that clonidine has profound antidiarrheal effects in experimental conditions, its centrally mediated hypotensive and sedative effects limit its use as an antidiarrheal agent.

Octreotide, a long-acting somatostatin analog, has been used to treat diarrhea of both neuroendocrine and nonneuroendocrine origin. The response in cases of short bowel syndrome–associated diarrhea has been variable, and although the diarrhea may respond initially, tachyphylaxis has been known to occur. [167] A therapeutic trial of octreotide when diarrhea cannot be controlled with conservative measures is warranted. In animal models of massive intestinal resection, physiological and pharmacological doses of octreotide reduced intestinal secretion and, importantly, did not alter the development of intestinal adaptation. [168] The standard initial therapy is 50 to 100 μg given subcutaneously over an 8- to 12-hour period; titration is based on the clinical and biochemical effects, with the dose escalated by 50 μg per week up to a maximal dose of 200 μg. The dose can be increased in 100-μg increments from 200 to 500 μg every 8 hours. Once the effectiveness of octreotide has been demonstrated in an individual patient, the patient can be switched to a comparable dose of the long-acting release depot octreotide preparation, which has similar beneficial effects of decreasing fecal output. [175]

The adverse effects of octreotide are usually mild and depend on the duration of therapy. Short-term side effects include pain at the injection site and transient nausea and bloating. Lanreotide, a new somatostatin analog, significantly reduces duodenal-to-cecal transit time in healthy male patients; however, it concomitantly decreases carbohydrate and fat absorption and thus may worsen diarrhea. It is quite possible therefore that somatostatin analogs have mixed effects in the management of diarrhea associated with short bowel syndrome. [176]

Recombinant Growth Hormone Therapy

The effects of low-dose recombinant human growth hormone (0.024 mg/kg daily) on body composition and absorptive capacity in patients with short bowel syndrome resulting from Crohn's disease were examined in a placebo-controlled crossover clinical trial. [177] Body weight and lean body mass increased, but no effect on nutrient absorption was noted. In an uncontrolled study, 8 weeks of recombinant human growth hormone treatment led to increases in body weight and in lean and fat-free body mass; the changes correlated positively with increases in serum levels of insulin-like growth factor-I. The combination of growth hormones and oral glutamine supplementation with a high-carbohydrate, low-fat diet appeared superior to either agent alone. [178] In this uncontrolled study of 47 patients, protein absorption increased by 40% and stool output decreased by 30%; of the patients, 40% were weaned from total parenteral nutrition within 1 year and another 40% were able to decrease their total parenteral nutrition requirements when they followed the diet. [178] In contrast, in a randomized, double-blinded, controlled trial, no benefit of growth hormone and glutamine on intestinal morphology, stool losses, or macronutrient absorption could be demonstrated in patients with short bowel syndrome. [179] Similarly, in a randomized, double-blinded, placebo-controlled crossover study, a combination of growth hormone (0.12 mg/kg per day) with oral (28 g/d) and parenteral (5.2 g/d) glutamine for 28 days did not improve intestinal absorption in patients with short bowel syndrome on their usual diet, [180] nor did it affect body weight or composition. [181] An analysis of published clinical studies of the use of recombinant growth hormone, glutamine, and low-fat, high-carbohydrate diets did not demonstrate benefit. [182]

The importance of a multidisciplinary approach involving a dedicated nutritional support team to optimize diet is demonstrated in the review by Wilmore [183] encompassing more than 300 patients with short bowel syndrome who received glutamine, growth hormone, and a dietary regimen optimized to enhance absorption.

After 2 years of long-term follow-up, 40% of the group could be weaned completely from parenteral nutrition, and 40% were able to reduce their parenteral nutrition requirements.

Whether growth hormone treatment will yield better outcomes in pediatric than in adult patients remains to be determined. Indeed, case reports have been published describing particularly impressive results in children with a combination of surgical lengthening and growth hormone administration. [184] These findings may imply different intestinal responses in children and adults to trophic growth hormones. Further studies are required to evaluate this promising therapy more fully.

Glucagon-like Peptide 2 Therapy

In a human clinical trial, 400 µg of glucagon-like peptide 2 was administered subcutaneously twice a day for 35 days to eight patients with short bowel syndrome. Treatment with glucagon-like peptide 2 improved the intestinal absorption of energy and nitrogen, increased body weight, and slowed the gastric, but not the small intestinal, emptying time. [185] Although the clinical effects of glucagon-like peptide 2 in this pilot study were modest, they are encouraging and will provide a basis for additional studies to determine the optimal dose and duration of therapy.

SURGICAL MANAGEMENT

Many of the surgical approaches require technical perfection and careful patient selection, and they are yet to be tested in clinical trials; however, along with intestinal transplantation, they will likely provide a foundation of therapy into the twenty-first century.

Techniques Designed to Slow the Transit Time

Antiperistaltic segments act like physiological valves by slowing intestinal transit and altering myoelectric activity. [186], [187] These reversed segments increase the absorption of water, nitrogen, and fat. [188], [189] and [190] The treatment of more than 30 patients with short bowel syndrome by this method has been reported thus far, with good results in about 70% of them. The ideal length for a reversal segment is 10 to 15 cm of distal small bowel in an adult and about 3 cm in an infant. [191], [192], [193], [194], [195], [196] and [197] Reversal may provide temporary relief while intestinal adaptation is taking place. [198] Patients with an extremely short intestine may not have the 10 cm of intestine required to fashion the reversal segment. Furthermore, small bowel obstruction at the antiperistaltic site is always a risk and may necessitate additional intestinal resection.

Peristaltic activity in the colon differs from that in the small intestine; it is slower and segmental and progresses by means of infrequent peristaltic rushes. [199] In dogs and rats, isoperistaltic interposition of a segment of colon between jejunal or ileal segments prolongs the intestinal transit time without causing obstruction. [199], [200] The isoperistaltic colon is placed proximally and functions by allowing the slow delivery of chyme to the distal intestine. Despite slowed transit, however, the interposed colonic segment does not show evidence of small bowel intestinalization [201] and appears to have no significant effect on the absorption of nutrients. [202] So far, about 30 cases of colonic interposition in short bowel syndrome have been reported. In these case reports, the length of the interposed colon, which does not appear to be critical, varied from 8 to 24 cm.

The colon also can be interposed in an antiperistaltic fashion. The antiperistaltic colon is placed distally. In one study, this procedure significantly improved the absorption of fat and xylose in monkeys. [203] Other authors, however, did not identify significant benefits to intestinal absorptive function. [204], [205] and [206]

Various techniques are used in the construction of valves to replace lost ileocecal valves. Nipple valves can be inserted at an ileostomy site. [207] In general, excision of the outer longitudinal muscle layer allows the inner circular muscular layer to contract without opposition, so that submucosal tunneling and the development of a nipple-like valve are possible. [208], [209] Distal bowel also can be everted and telescoped into the proximal bowel. [210] These techniques of constructing an artificial valve do not modify the intestinal motor response to intestinal resection but simply create a low-grade mechanical obstruction. [211] In both experimental animal models and in humans with short bowel syndrome, such valves have reduced the number of evacuations. [212], [213] However, despite a significantly prolonged transit time, the operation has not been shown to result in increases in absorptive surface area, functional absorptive capacity, or body weight. [214]

In principle, recirculating loops should prolong the transit time by promoting repeated exposure of the intestinal mucosal surface area to lumenal contents. This procedure is associated with high morbidity and mortality rates in experimental animals, however, [215] which appear to be related to bacterial overgrowth and the absorption of toxic metabolites.

Retrograde electrical pacing is designed to slow and reverse the flow of intestinal contents, so that chyme pools and absorption is increased through prolonged mucosal contact. In dog jejunum, retrograde electrical pacing increased water, sucrose, and sodium absorption. [216] The technique has not been applied successfully in humans.

In animal studies, intrinsic myenteric denervation of the ileum, in an attempt to slow the forward flow of intestinal contents, led to dilation of the residual small and large intestine in rats that had been subjected to massive intestinal resection and resulted in an increase in absorptive surface area and postoperative weight gain. [217]

Tapering and Bowel-Lengthening Procedures

Adaptive dilation of the proximal small bowel causes stasis and ineffective peristalsis. The simplest way to deal with this problem is excision of the dilated segment, provided the length of small intestine is sufficient. In cases in which bowel length is insufficient, the diameter of the dilated bowel can be reduced through intestinal plication or tapering enteroplasty. These simple and safe techniques allow all the mucosal surface to be preserved. [218] Recently, Ramanujam, [219] using mongrel puppies, demonstrated that plication is a remodeling technique that is superior to tapering enteroplasty.

Bianchi [220] described a technique for lengthening the bowel of pigs by using the dilated segment. The gut is tapered, and the redundant small bowel is used for lengthening purposes. Since the first clinical application, this has been the surgical technique most frequently used in attempts to reverse short bowel syndrome in children. [221], [222] Although the bowel length is increased, mucosal function may be compromised. In one study, sucrose transport was found to be decreased across the segment involved in the bowel lengthening. [31] In dogs with a 75% distal ileal resection, tapering and lengthening impaired nutritional status and intestinal absorption and adaptation. The changes in absorption and transit appeared to be related to altered intestinal transit rates, and the impaired adaptation was related to decreased enteroglucagon levels and increased somatostatin levels. [223] These findings are not surprising because intestinal lengthening does not provide new mucosa. Any benefit is likely to be attributable to the mechanical effect of slowed intestinal transit.

In a series involving five human subjects, isoperistaltic tapering and lengthening (Bianchi procedure) resulted in improvement of intestinal function. At 1 and 6 months postoperatively, stool output was decreased, D-xylose absorption was increased, and fecal fat was decreased in comparison with preoperative measurements. [224] Isoperistaltic tapering and lengthening resulted in survival off parenteral nutrition in 45% of 20 pediatric patients. Successful weaning from parenteral nutrition was related to an initial bowel length of more than 40 cm. [225] Similar results have been reported by a German pediatric surgical group in 18 children. [226]

Georgeson and associates [227] combined several different techniques. If the bowel was not sufficiently dilated, a nipple valve was constructed distally to provide partial small bowel obstruction and so induce dilation. Following this, the Bianchi technique was used to lengthen the bowel. In a small series of case reports, length was increased on average by a factor of 2.5 during 1 year; enteral intake averaged 66% of the total requirement, in comparison with less than 10% before the operation.

Intestinal regeneration takes place in full-thickness intestinal defects patched with a variety of substances, including colonic serosa and prosthetic materials. [228], [229] The new intestinal tissue, which develops by lateral growth, is functionally similar to normal intestinal mucosa. [230] In dogs, however, intestinal interposition of a patch of colon denuded of its mucosa resulted in the regeneration of colonic, rather than small intestinal, mucosa. Growth of neomucosa has never been attempted clinically.

INTESTINAL TRANSPLANTATION

Indications

Intestinal transplantation has become a life-saving treatment that can be considered for patients with irreversible intestinal failure who cannot be maintained on parenteral nutrition. Life-threatening complications of parenteral nutrition that warrant a consideration of intestinal transplantation include parenteral nutrition–associated liver disease, recurrent sepsis, and threatened loss of central venous access. The indications are similar for children and adults. Contraindications to transplantation include profound neurological difficulties, life-threatening illness, multiple-system immune diseases, and lack of venous access. The fact that intestinal failure itself can lead to the development of profound neuropathological abnormalities in the brain may be a reason to consider earlier transplantation. [231]

Because the shortage of donor organs is critical, waiting times for intestinal transplantation are prolonged. Therefore, it is important that patients with life-threatening complications of intestinal failure and parenteral nutrition therapy be identified and referred for transplantation early. [232], [233] Because the survival rate of isolated intestinal transplants is higher than that of combined intestinal-liver transplants, patients should be considered for transplantation before bridging fibrosis and cirrhosis develop. Finally, an appreciation of the unique and specific nutritional needs of the recipient and the donor is important in maximizing survival. [234]

Natural History of Short Bowel Syndrome Treated with Parenteral Nutrition

The long-term rates of survival and dependence on parenteral nutrition of adult patients with short bowel syndrome have been reviewed. [235] Rates of 5-year survival and dependence on parenteral nutrition are 55% and 45%, respectively. Survival is negatively related to end-enterostomy, a small bowel length of less than 50 cm, advanced age, and arterial infarction as a cause of the short bowel syndrome. [235], [236] Dependence on parenteral nutrition is negatively related to a small bowel length of less than 100 cm and to the absence of a terminal ileum or colon. After 2 years of parenteral nutrition, the probability of a permanent need for parenteral nutrition is 95%. Similar survival rates have been reported for children with short bowel syndrome treated with parenteral nutrition. [237] Long-term home parenteral nutrition, although it can maintain normal nutritional parameters in 80% of patients, results in frequent hospitalizations, predominantly for catheter-related problems, a 30% rate of regular opiate use, and an inability to achieve complete rehabilitation. [238]

Results

Intestinal transplantation is an evolving procedure with survival rates now comparable, at some larger centers, to those of parenteral nutrition. [239] The intestinal transplant data registry (Web site, http://www.intestinaltransplant.org/) provides updated figures for graft and patient survival from intestinal transplant centers across the world. Two thirds of the recipients are children or adolescents. Short bowel syndrome with a failure of maintenance on parenteral nutrition is the most common indication for transplantation. [240] Types of transplantation include small bowel with or without colon (40%), intestine and liver (45%), and multiple-visceral grafts (15%). As of the last major review from the transplant registry in 2001 (updated reviews are periodically posted on the Web site), the 1-year graft/patient survival rates at major centers doing more than 10 transplants since 1999 are 60%/75% for intestinal grafts, 60%/66% for intestinal and liver grafts, and 65%/70% for multiple-visceral grafts. [241] In a retrospective review from a single center performing a large number of intestinal transplants, the 1-year graft/patient survival was reported as 93%/71%. [242]

The single most important factor in both graft and patient survival is the number of transplants a center completes each year. Donor type, previous treatment of the donor, diagnosis, recipient age, and organs transplanted do not affect survival. Approximately 80% of survivors of transplantation have been able to discontinue parenteral nutrition and resume oral nutrition, 10% have a partial or nonfunctioning transplant in place, and 10% have had their transplant removed. Survival results during the last several years have improved as consequence of refinements in operative techniques, a reduced requirement for immunomodulation, and improvements in postoperative care. Rejection remains the single most important factor (60%) leading to graft failure and removal. Advances in immunosuppression have significantly reduced rejection rates. Part of the challenge in managing these patients is maintaining the delicate balance between immunosuppression, which is necessary to prevent graft versus host disease, and rejection. At the same time, the required degree of immunosuppression is associated with a high risk for lymphoproliferative disorders and intestine-derived sepsis. The incidence of lymphoproliferative disease after intestinal transplantation is 15%; it appears to be linked to Epstein-Barr virus infection, OKT3, and corticosteroids. [243] Additional reasons for graft failure are thrombosis, ischemia, and bleeding (25%), multiple-organ failure (10%), sepsis (6%), and lymphoma (1%).

Sepsis (60%) is the most frequent cause of death in this transplant population. [243] Reduced rates of sepsis and improved survival rates have been described following living-related transplantation. [244], [245] and [246] Investigations of bacterial translocation following intestinal transplantation have suggested that cold ischemia time, not rejection or the associated transplantation of the colon, influences bacterial translocation. [247] Additional causes of death are nontransplant organ failure (14%), lymphoma (14%), thrombosis, ischemia, and bleeding (13%), and graft rejection (12%).

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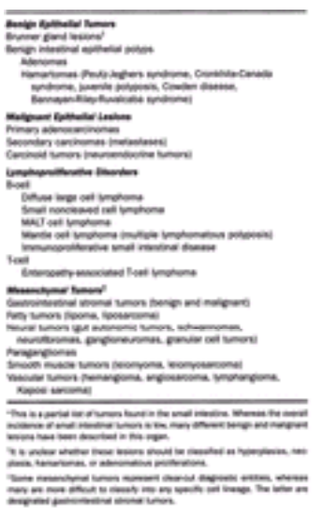
CHAPTER 80

Robert S. Bresalier and Tamir Ben-Menachem

TUMORS OF THE SMALL INTESTINE

Small intestinal tumors (tumors of the duodenum, jejunum, and ileum) are uncommon by comparison with those occurring elsewhere in the gastrointestinal tract, and fewer than 2% of malignant gastrointestinal tumors are derived from this organ. [1], [2], [3], [4], [5] and [6] Primary small intestinal tumors are derived from both epithelial and mesenchymal components of the small bowel (Table 80-1). Primary malignant tumors include adenocarcinomas, carcinoids, lymphomas, and sarcomas (now classified as gastrointestinal stromal tumors). Approximately 5300 new cases of primary small intestinal cancer occurred in the United States in 2002 (equally distributed between men and women), with 1100 estimated cancer deaths. [1] The diverse nature of small intestinal tumors makes it difficult to make generalizations about the group as a whole. Some tumors represent distinct entities, whereas others share overlapping features.



Benign Epithelial Tumors
Brunner gland lesions[†]
Benign intestinal epithelial polyps
Adenomas
Hamartomas (Peutz-Jeghers syndrome, Cronkhite-Canada syndrome, juvenile polyposis, Cowden disease, Bannayan-Riley-Ruvalcaba syndrome)

Malignant Epithelial Lesions
Primary adenocarcinomas
Secondary carcinomas (metastases)
Carcinoid tumors (neuroendocrine tumors)

Lymphoproliferative Disorders
B-cell
Diffuse large cell lymphoma
Small noncleaved cell lymphoma
MALT cell lymphoma
Mantle cell lymphoma (multiple lymphomatous polyposis)
Immunoproliferative small intestinal disease
T-cell
Enteropathy-associated T-cell lymphoma

Mesenchymal Tumors[‡]
Gastrointestinal stromal tumors (benign and malignant)
Fatty tumors (lipoma, liposarcoma)
Neural tumors (gut autonomic tumors, schwannomas, neurofibromas, ganglioneuromas, granular cell tumors)
Paragangliomas
Smooth muscle tumors (leiomyoma, leiomyosarcoma)
Vascular tumors (hemangioma, angiosarcoma, lymphangioma, Kaposi sarcoma)

*This is a partial list of tumors found in the small intestine. Whereas the overall incidence of small intestinal tumors is low, many different benign and malignant lesions have been described in this organ.
†It is unclear whether these lesions should be classified as hyperplasias, neoplasia, hamartomas, or adenomatous proliferations.
‡Some mesenchymal tumors represent clear-cut diagnostic entities, whereas many are more difficult to classify into any specific cell lineage. The latter are designated gastrointestinal stromal tumors.

TABLE 80-1 Classification of Small Intestinal Tumors*

DESCRIPTIVE EPIDEMIOLOGY

The frequency of small bowel tumors varies among different populations. [7] Reviews of the incidence and natural history of small bowel tumors are composed of small series of patients, and some may include both primary or secondary (e.g., lymphomas) tumors. Some series include periampullary tumors and tumors of the ampulla of Vater, whereas others do not. [2] The relative frequency of primary tumors ranges in the literature, with adenocarcinoma (24% to 52%), malignant carcinoid (17% to 41%), lymphoma (12% to 29%), and sarcomas (11% to 20%) the most frequent. [2], [3], [4], [5] and [6], [8], [9], [10], [11], [12], [13] and [14] The pattern of distribution of tumors in the small intestine is dependent on histological type. In Western countries, adenocarcinomas are most commonly found in the duodenum. In the setting of Crohn's disease, adenocarcinomas may occur distally in the ileum. Carcinoids and primary lymphomas occur predominantly in the ileum and jejunum. Sarcomas are found more evenly distributed. Metastases to the small bowel from adenocarcinomas and sarcomas account for approximately 50% of all small bowel tumors.

An overall age-adjusted incidence of primary small bowel cancers of 1.4 per 100,000 was reported in a United States population-based study. [5] Small but significant increases in the incidence of adenocarcinomas and carcinoids have been noted since the 1970s, [15] with a slight male predominance. The incidence of small bowel cancers rises with age, beginning at 30 years, with a mean age of diagnosis of approximately 60 years. [17] Incidence rates for adenocarcinomas and malignant

carcinoids are higher in African Americans than in whites (6.4% and 1.4% increase per year, respectively). [15]

ETIOLOGY AND PATHOGENESIS

Why Are Small Bowel Tumors Rare?

Several hypotheses have been proposed to explain this phenomenon, but objective data are mostly lacking. [17]

1. Rapid transit time and liquid lumenal contents reduce mucosal contact with putative carcinogens as well as physical damage, whereas mucosal enzymes such as benzpyrene hydroxylase detoxify them.
2. Bacterial enzymes have been postulated to play a role in colonic carcinogenesis. When bacterial flora is altered, as in bacterial overgrowth syndromes, small intestinal carcinomas develop with a higher frequency than expected. The small intestine, conversely, has low bacterial counts, with an absence of anaerobes under normal conditions.
3. Stem cells located at the crypt base lie deeper in the small intestine than the colon, perhaps reducing contact with lumenal carcinogens. The rapid differentiation of stem cells into mature, nonproliferating enterocytes and goblet cells may also protect against factors that deregulate cell growth and promote cancer development.
4. Based on preclinical data from murine models, [18] apoptosis involving damaged cells and potentially tumorigenic clones differs between the small and large intestine.
5. Lymphoid tissue in the lamina propria and Peyer patches of the ileum may provide immune surveillance against neoplastic cells through IgA-rich secretions.

Diet

The limited descriptive epidemiology of small intestinal tumors has not provided strong evidence for environmental influences on tumor formation similar to that of the large intestine. Weekly or more frequent consumption of red meat or salt-cured foods has been associated with a two- to threefold increase in risk of small intestinal cancer. [19] Another study [20] reported odds ratios (OR) for cigarette smoking of 4.6 for adenocarcinomas and 4.2 for carcinoids and an OR for alcohol consumption of 4.0 for adenocarcinomas and 3.1 for carcinoids. A population-based case control study [21] observed a significant threefold increased risk of adenocarcinoma of the small bowel in heavy drinkers (=80 g/d of ethanol) relative to moderate drinkers or nondrinkers. Frequent intake of foods rich in heterocyclic amines (fried bacon, ham, barbecued or smoked meat and fish) was associated with a significant four- to fivefold increased risk of adenocarcinoma in men, but not in women. There was also a consistent and significant trend in risk with increasing sugar intake, with an OR of 3.8 in the highest intake category. In a hospital-based case control study, [22] alcohol and tobacco did not increase the risk of adenocarcinoma of the small intestine. Risk was positively related to intake of bread, pasta, or rice (OR = 3.8), sugar (OR = 2.9), and red meat (OR = 4.6), and it was inversely related to coffee (OR = 0.4), fish (OR = 0.3), vegetables (OR = 0.3), and fruit (OR = 0.6).

Molecular Biology and Biochemical Changes

In comparison with colorectal cancer, relatively little is known about the molecular genetic events associated with the evolution of small bowel tumors. Genetic changes that lead to development of gastrointestinal cancer can generally be categorized into alterations in protooncogenes, loss of tumor suppressor gene activity, and abnormalities in genes involved in DNA mismatch repair.

Cellular protooncogenes are evolutionarily conserved human genes that play a role in signal transduction and normal regulation of cell growth. Human *ras* genes encode guanine nucleotide binding proteins that regulate intracellular signaling pathways. Point mutations in K-*ras,* especially at codon 12, have been reported in 14% to 53% of primary small intestinal adenocarcinomas, [23], [24] but not in carcinoids. K-*ras* mutations were common in sporadic and Crohn's disease–associated adenocarcinomas of the small intestine. [25] K-*ras* mutations were present in all four sporadic carcinomas with contiguous adenomas, in 2 of 11 (18%) without adenomas, and in 4 of 7 (43%) of Crohn's disease–associated carcinomas.

Allelic losses, particularly involving tumor suppressor genes at chromosome locations 5q, 17p, and 18q, play major roles in the genesis of large bowel tumors. The *APC* gene on the long arm of chromosome 5 (5q21) is mutated in the germ line of patients with familial adenomatous polyposis (FAP), and somatic mutations of *APC* occur in 60% to 80% of sporadic colorectal carcinomas and adenomas (see Chapter 89, Chapter 90, and Chapter 91). Mutation of the mouse homolog of *APC* (*Apc*) by chemical carcinogenesis or by genetic manipulation [26] results in development of large and small intestinal polyps. Small intestinal and periampullary adenomas and carcinomas are common in patients with FAP. *APC* mutations have also been reported in sporadic small intestinal adenocarcinoma, [24] but they appear to occur at a much lower frequency than in sporadic colorectal cancer.

Deletions of chromosome 17p involve the *p53* tumor suppressor gene whose product normally prevents cells with damaged DNA from progressing from the G_1 phase to the S phase in the cell cycle and regulates apoptosis. Deletions within chromosome 17p are present in approximately 75% of colorectal cancers. Mutations in *p53* were found in 27% of small bowel adenocarcinomas in one series, [24] whereas alterations in the p53 gene product and allelic loss of chromosome 17p were present in 47% of sporadic small bowel adenocarcinomas, 33% of contiguous adenomas, and 71% of Crohn's disease–associated carcinomas in a second series. [25]

Candidate tumor suppressor genes on chromosome 18q include the *DCC* or deleted in colon cancer gene and *DPC4* (*SMAD4*). *DPC4* belongs to the SMAD gene family involved in signal transduction pathways activated through the transforming growth factor ß family receptors. Loss of 18q has been reported in intestinal tumors from the Apc 1638N mouse model, [26] but rarely in human small intestinal adenocarcinomas. [25] Chromosome 18 deletions appear to be common events, however, in classical midgut carcinoids of the small intestine. [27], [28]

Alterations in genes that help to maintain DNA fidelity during replication (mismatch repair genes) are characteristic of patients with hereditary nonpolyposis colorectal cancer (HNPCC), but they may be found in approximately 15% of sporadic colorectal cancers. Mismatch repair gene mutations in *hMLH1* and *hMSH2* were present in 15 of 42 (36%) of HNPCC-associated small bowel carcinomas in one large series. [29] DNA replication errors characterized by microsatellite instability were also reported in 13% of sporadic small bowel carcinomas. [25]

RISK FACTORS AND ASSOCIATED CONDITIONS

Numerous risk factors and associated conditions have been described with relation to tumors of the small intestine (Table 80-2).

Adenocarcinoma
Familial adenomatous polyposis
Hereditary nonpolyposis colorectal cancer
Peutz-Jeghers syndrome
Juvenile polyposis syndrome
Crohn's disease
Previous history of cholecystectomy
Long-standing ileostomy (especially with Crohn's disease)
Ileal loop conduits, ileal pouches, ileal cystoplasty
Duplication cysts and Meckel diverticula
Gluten-sensitive enteropathy
Non-Hodgkin B-Cell Lymphoma
Immunoproliferative small intestinal disease
Gluten-sensitive enteropathy
Nodular lymphoid hyperplasia
Acquired immunodeficiency syndrome
Crohn's disease

TABLE 80-2 Conditions Associated with an Increased Risk of Primary Small Intestinal Neoplasia

Familial Adenomatous Polyposis

Periampullary duodenal carcinoma is the most common extracolonic malignant tumor in patients with FAP. There were 11 cases of duodenal and ampullary

adenocarcinoma reported among 1391 patients with FAP, [30] a relative risk (RR) of duodenal adenocarcinoma of 331, and an RR of 124 for ampullary carcinoma compared with the general population. Mean ages of diagnosis were 53 years for duodenal carcinoma and 49 years for ampullary carcinoma. Duodenal adenomas are common in patients with FAP, and they have been reported in up to 90% of such patients.

Hereditary Nonpolyposis Colorectal Cancer

HNPCC is an inherited disease in which colon cancers arise in discrete adenomas, but polyposis (i.e., hundreds of polyps) does not occur (see Chapter 90). Forty-two members of 40 families with HNPCC developed 42 primary and 7 metachronous small bowel tumors. [29] There were 46 adenocarcinomas and 3 carcinoid tumors. The median age at diagnosis of the index small bowel tumor was 49 years. The small bowel was the first site of carcinoma in 24 (57%) patients.

Hamartomatous Polyposis Syndromes

Hamartomatous polyposis syndromes are a group of disorders marked by the finding of multiple hamartomatous polyps in the gastrointestinal tract (see Chapter 90). The *Peutz-Jeghers syndrome* is an autosomal dominant disease, which in most families has been mapped to chromosome 19p13.3 and the *STK11* (serine threonine kinase 11) gene. Adenomatous changes have been reported in 3% to 6% of hamartomas. Extracolonic cancers are common, occurring in 50% to 90% of patients (small intestine, stomach, pancreas, esophagus, ovary, lung, uterus, breast). The RR for all cancers is 15.2 compared with the general population. [31] The small intestine represents the most frequent site of cancer development, with an RR of development of adenocarcinoma of 520 compared with the general population. Patients with Peutz-Jeghers syndrome have characteristic mucocutaneous pigment spots, which are frequent in the perioral region and across the vermillion border. Polyps are distinctive, with an arborizing pattern of growth of the muscularis mucosa extending into the branching fronds of the polyp. Patients often present with obstructive symptoms and may develop intussusception of the small bowel.

Juvenile polyposis syndrome is an autosomal dominant disease in which hamartomatous polyps may be limited to the colon, limited to the stomach, or occur throughout the gastrointestinal tract. Mixed juvenile and adenomatous polyps have been reported. The RR for small intestinal cancer is unclear, but cancers of the duodenum have been reported. Patients may present with bleeding, intussusception, and obstruction. A life-threatening protein-losing enteropathy has been reported in some patients. Germ-line mutations in the *SMAD4* (chromosome 18q21.1) and *PTEN* (chromosome 10q23) genes have been reported in separate kindreds with juvenile polyposis syndrome. [32]

Crohn's Disease

An increased incidence of both small intestinal adenocarcinoma and lymphoma has been reported to occur in *Crohn's disease,* but the magnitude of the risk is unclear. The RR of small bowel carcinoma in Crohn's disease (mostly ileal) has been estimated to be as much as 100 times that of the general population. [33] A retrospective cohort analysis of 373 patients with Crohn's disease reported 2 cases of ileal adenocarcinoma compared with an expected 0.04 cases of expected small bowel cancer (RR = 50). [34] A dysplasia-carcinoma sequence occurs in the small intestine in Crohn's disease similar to that of the colon and rectum. [25], [35] Crohn's disease–associated small bowel carcinomas and dysplasias are accompanied by frequent genetic alterations in K-*ras* and *p53*.

Diversion of Bile (Previous Cholecystectomy)

Diversion of bile to the lower small intestine either surgically or by feeding cholestyramine increases the yield of intestinal carcinomas in carcinogen-treated animals. Thus, cholecystectomy, which increases delivery of bile to the proximal bowel, could theoretically lead to an increased risk of small and large bowel cancer. Indeed, patients who underwent cholecystectomy had an increased risk of proximal intestinal adenocarcinoma that declined with increasing distance from the common bile duct. [36] The risk was significantly increased for adenocarcinoma of the small bowel (RR, 1.71) and the right-sided colon.

Gluten-Sensitive Enteropathy

An increased risk of both small intestinal adenocarcinoma and non-Hodgkin lymphoma has been reported to be associated with *celiac sprue* (gluten-sensitive enteropathy). [37], [38] (see Chapter 76). The overall incidence of malignant disease in this condition has been reported to be 11% to 13%, with an incidence of small bowel lymphomas of 4% to 7%. In some reported cases, it is not clear whether the primary diagnosis is actually celiac sprue or whether the histological changes are secondary to lymphoma per se. Lymphomas in the setting of celiac disease are usually of T-cell origin (enteropathy-associated T-cell lymphomas or EATLs). Adenocarcinomas may arise in focal areas of dysplasia. [39]

Other conditions may carry an increased RR for small bowel neoplasia (see Table 80-2), but they are relatively rare. Patients with urinary diversion to the small bowel (ileal loop conduits) appear to have an increased risk of carcinoma. Adenocarcinomas arise in the mucosa of ileostomies and ileal pouches in patients with inflammatory bowel diseases and FAP, [40] although this event is probably rare. [41]

CLINICAL PICTURE

The clinical signs and symptoms of small bowel neoplasms may have some common features, but for the most part they depend on the individual tumor type (Table 80-3). Abdominal pain may result from partial or complete small bowel obstruction, intussusception, or ischemia. Obstruction may be the result of the physical bulk of the tumor, infiltration, or annular constriction. An intense dermoplastic reaction by carcinoids causes intestinal buckling or kinking. Intussusception may occur with benign polypoid lesions, accounting for half of adult intussusception, and is common in patients with gastrointestinal stromal cell tumors (GISTs). Occult gastrointestinal blood loss is another common feature of benign and malignant small bowel tumors. Bleeding may be acute and brisk, however, in the case of ulcerated GISTs. Weight loss is especially common and pronounced in patients with small bowel lymphoma.

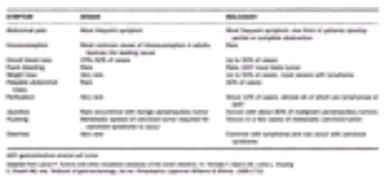


TABLE 80-3 Clinical Features of Small Bowel Tumors

ADENOCARCINOMA

Epidemiology and Risk Factors

Adenocarcinoma represents the most common primary malignant disease of the small intestine. The incidence of primary small bowel adenocarcinoma ranges between 3.0 and 6.5 per million, representing approximately 40% of primary small bowel tumors in most population-based studies (range, 24% to 42%). [17] Small bowel adenocarcinoma accounts for approximately 2% of gastrointestinal tumors and 1% of gastrointestinal cancer deaths. Fifty-five percent of these tumors occur in the duodenum, 18% in the jejunum, 13% in the ileum, and 14% in nonspecified sites. [2] The overall 5-year survival is 30.5%, with a median survival of 19.7 months. A slight male predominance is usual. The incidence of small bowel adenocarcinoma is slightly higher in blacks compared with whites. [15]

Adenocarcinomas arise from adenomas or dysplastic changes of the small intestine (see section " Pathology, Natural History, and Staging "). Adenomas and adenocarcinomas of the small intestine, and especially of the duodenum and ampulla of Vater, are most frequently encountered in the setting of FAP. Untreated duodenal and ampullary adenomas in patients with FAP were followed in the context of an endoscopic surveillance program. [42] Duodenal polyps progressed in size in 26% (25 of 95), in number in 32% (34 of 106), and in histology in 11% (5 of 45). Morphology and histology of ampullary adenomas progressed in 14% (15 of 110) and

11% (12 of 105) of patients, respectively. One patient developed a periampullary carcinoma.

Pathology, Natural History, and Staging

Adenocarcinomas of the small intestine arise in discrete adenomatous polyps, in adenomatous changes involving the ampulla of Vater, or in dysplasia associated with inflammatory bowel disease. In hamartomatous polyposis syndromes, carcinomas may arise from discrete adenomas or mixed polyps containing adenomatous components. [32] Dysplasia is associated with familial juvenile polyps, but not with solitary, sporadic juvenile polyps. Dysplastic polyps contain markers suggesting a loss of proliferative control within the epithelium and mutations in *APC*. [43]

Adenomas in the small intestine display the same gross and microscopic features as those in the large intestine. They may be pedunculated or sessile. Tubular adenomas tend to be small, whereas those with villous architecture tend to be larger. The presence of multiple duodenal adenomas or adenomatous changes in the ampulla of Vater suggests the diagnosis of FAP. Tubular adenomas contain tall, columnar, pseudostratified epithelial cells. Villous adenomas contain fingerlike villous or papillary processes similar to those in the colon. Small intestinal adenomas exhibit a spectrum of dysplasia ranging from mild dysplasia to intramucosal carcinoma and invasive cancer. Glandular crowding ("back to back" glands), loss of epithelial polarity, an increased nuclear-cytoplasmic ratio, and increased mitoses characterize high-grade dysplasia. These changes, accompanied by invasion into the lamina propria, define *intramucosal carcinoma.*

Invasive carcinoma may be found in one fourth to one half of resected villous adenomas in the setting of FAP, [43] but most duodenal adenomas appear to progress slowly to carcinoma, [42], [44] and the actual risk of malignant degeneration is probably considerably lower. Adenomatous polyps of the small intestine do occur outside the setting of FAP, but even less is known about their natural history.

Small intestinal adenocarcinomas may appear grossly as flat, stenosing, ulcerative, infiltrating, or polypoid lesions. Most are moderately differentiated tumors with gland formation and variable degrees of mucin secretion. Poorly differentiated tumors occur in approximately 20% of cases. The presence of *signet cells,* in which a large vacuole of mucin displaces the nucleus to one side, is a feature of some of these tumors. Rare adenosquamous carcinomas of the duodenum have been reported.

Adenocarcinomas are staged depending on the extent of bowel wall invasion, the presence or absence of invasion into adjacent structures, and the presence or absence of lymph node or distant metastases (Table 80-4). [45] The prognosis of patients with adenocarcinomas of the small bowel is poor. In one report, the overall 5-year disease specific survival in the United States was 30.5%, with a median survival of 19.7 months. [2] Factors significantly correlated with survival included age (poorer prognosis for patients >75 years), tumor site (duodenum worse than jejunum or ileum), disease stage, and whether cancer-directed surgery was performed. Five-year survival rates were 65% for stage I, 48% for stage II, 35% for stage III, and 4% for stage IV tumors. Similar trends have been reported from Europe. [6], [46], [47] and [48] Adenocarcinomas arising in the setting of HNPCC have a better prognosis than those occurring in FAP or in the general population. [29] Patients with tumors arising at or near the ampulla of Vater may do better than those with more distal tumors, perhaps because they become symptomatic earlier. Five-year survival rates approaching 50% have been reported for patients with node-negative cancers of the ampulla after radical resection.

Stage 0
Carcinoma in situ: TIS N0 M0
Stage I
Tumor invades lamina propria or submucosa: T1 M0 N0
Tumor invades muscularis propria: T2 N0 M0
Stage II
Tumor invades muscularis propria into the subserosa or into the nonperitonealized perimuscular tissue (mesentery or retroperitoneum*) with extension 2 cm: T3 N0 M0
Tumor perforates the visceral peritoneum or directly invades other organs or structures includes other loops of small intestine, mesentery or retroperitoneum 2 cm, and the abdominal wall by way of the serosa; for the duodenum only includes invasion of the pancreas: T4 N0 M0
Stage III
Any degree of bowel wall perforation with regional lymph node metastasis; any T N1 M0
Stage IV
Any degree of bowel wall perforation with or without regional lymph node metastasis, with distant metastasis

*The non-peritonealized perimuscular tissue is, for the jejunum and ileum, part of the mesentery, and for the duodenum, in areas where serosa is lacking, it is part of the retroperitoneum.

Adapted from ref. 45.

TABLE 80-4 Staging of Small Intestinal Adenocarcinoma: American Joint Committee on Cancer (TNM Classification)

Clinical Picture

The mean age of presentation for adenocarcinomas is approximately 65 years, with a wide range of age at presentation. [2] Fewer than 1% of tumors occur before age 30 years, and approximately 85% occur after age 50 years. Symptoms relate to tumor size, location, and blood supply. Small tumors are asymptomatic or may present with anemia secondary to chronic blood loss, but for the most part they are indolent and difficult to diagnose. Abdominal pain and other obstructive symptoms such as nausea and vomiting are common late symptoms as tumors obstruct from infiltration with lumenal narrowing or mass effect. Anorexia and weight loss are also common symptoms. Ileal tumors may present uncommonly with intussusception. Tumors located in the periampullary duodenum or ampulla of Vater may cause obstruction of the common bile duct, presenting with jaundice and other signs of biliary obstruction.

Diagnosis

Endoscopy (with forward and side viewing) may be used to examine the duodenum where a lesion is suspected or in families with FAP. Screening for duodenal adenomas and lesions of the ampulla of Vater, and surveillance after adenomas are removed endoscopically, should be performed in persons exhibiting the phenotype of FAP or in family members in whom genetic testing is positive for mutations in the *APC* gene. Individual polyps may be examined by biopsy or, if possible, removed by snare cautery and the ampulla examined by biopsy as necessary. There is a high incidence of adenomatous change in the papilla, even if the appearance is normal. It has been suggested that the slow evolution of duodenal and papillary lesions may justify an endoscopic surveillance interval of 3 years for most untreated patients with FAP. [40] Upper endoscopic surveillance should begin at age 20 years, and biopsies should be taken of any suspicious lesions and of the papilla, even if normal appearing. Push enteroscopy is of use in identifying and biopsying small bowel lesions seen on small bowel barium studies or computed tomography (CT), in evaluating occult gastrointestinal bleeding, and in surveillance of patients with Peutz-Jeghers syndrome, in whom polyps occur throughout the gastrointestinal tract. [49] Colonoscopy with intubation of the terminal ileum may be helpful in examining the terminal ileum in patients with Crohn's disease or in biopsy of suspicious lesions suggested by radiographic procedures.

Small bowel follow-through is 70% to 80% accurate for detection of duodenal lesions. [50] This can be improved by the use of hypotonic duodenography. Barium studies, however, are not as accurate in detecting more distal tumors. The tumor detection rate of enteroclysis (95%) is greater than that for small bowel follow-through (61%). [51] The radiologic appearances of small bowel adenocarcinomas are similar to those of the colon, including annular narrowing or stricture formation (Fig. 80-1) and filling defects comprising polypoid or ulcerated masses. Seventy percent of tumors in the duodenum are polypoid. Large duodenal lesions are ulcerative in 20% of cases. Adenocarcinomas of the jejunum are primary annular-constricting lesions (75%) and may be partially ulcerated or fungating. Differentiation of the infiltrating form of adenocarcinoma from stenosing Crohn's disease may be difficult.

FIGURE 80-1. Annular constricting adenocarcinoma of the jejunum. This upper gastrointestinal barium study demonstrates an “apple-core” lesion (*arrow*).

CT is increasingly used in demonstrating small bowel tumors and their complications and is often preferable to small bowel follow-through. [52] Occasionally, intussusception and obstruction may be associated with polypoid adenocarcinomas that may be detected on CT examination (Fig. 80-2). CT can be helpful in guiding the surgeon to the source of obstruction. Endoscopic ultrasonography can also be used for diagnosing and staging small bowel tumors.

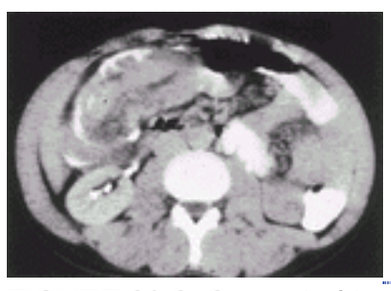


FIGURE 80-2. Computed tomography scan demonstrating intussusception in the terminal ileum resulting from a small intestinal cancer (*asterisk*). Intussusception gives the impression of a soft tissue “mass” or “bowel within bowel” surrounded by contrast. Layers of serosal fat are seen in the periphery and centrally.

Therapy

Pedunculated adenomas of the duodenum may be removed by endoscopic snare polypectomy. It is often difficult to remove villous adenomas of the small intestine in this manner. These lesions are often sessile and contain foci of invasive carcinoma. Local mucosal resection can be performed surgically for some lesions, but wide excision is indicated for villous adenomas containing invasive cancer.

Surgical resection is the treatment of choice for adenocarcinoma of the small intestine. Resection of the tumor with adjacent mesentery and lymph nodes with a wide margin (=10 cm) of normal bowel on either side of it is practical for adenocarcinomas of the jejunum. Malignant tumors of the terminal ileum are treated as cecal tumors with terminal ileal resection and right hemicolectomy. Villous adenomas of the duodenum containing invasive carcinoma and carcinomas of the ampulla of Vater usually require pancreatoduodenectomy (Whipple procedure) with curative intent. In some cases, local resection of pedunculated polyps may be adequate, but a clear margin of resection is mandatory in this situation. Disease-specific survival for primary small bowel adenocarcinomas confined to the small intestine (local disease) was 47.6, for regional disease it was 33%, and in distant disease it was 3.9%. [2]

The role of postoperative chemotherapy, either in the adjuvant setting or for unresectable lesions, has not been defined in patients with adenocarcinoma of the small intestine. Small series have claimed some benefit for 5-fluorouracil [53] and the nitrosoureas, but objective evidence of an impact on survival is lacking. Palliative radiation may have some role in controlling chronic blood loss associated with duodenal tumors, but it is difficult in more distal small bowel tumors because of a mobile mesentery and difficulty in localizing the target field. Chemoprevention with aspirin and other nonsteroidal antiinflammatory drugs and selective cyclooxygenase-2 inhibitors may be useful in the care of patients with FAP. Whereas several short-term studies demonstrated the ability of these agents to reduce the size and number of colonic polyps in FAP, data concerning small intestinal tumors are scant. These agents may reduce the size and number of small intestinal polyps in murine models of FAP. [26] Sulindac therapy was associated with a decrease in mucosal proliferation, but a trend toward duodenal polyp regression was not significant in one 6-month human trial. [54]

CARCINOID TUMORS

Epidemiology

Carcinoid tumors—or argentaffinomas—belong to a family of rare neuroendocrine neoplasms known also as amine precursor uptake and decarboxylation tumors. This entire family of neoplasms has in common the ability to secrete amines and polypeptides, which produce the characteristic clinical syndromes with which they sometimes present. Seventy-four percent of carcinoid tumors occur in the gastrointestinal tract. [55] Most appear in the appendix, followed in frequency by the small bowel and rectum. Most of the clinically significant carcinoid tumors are located in the small bowel. Most (87%) small bowel tumors are in the ileum, especially within 2 feet of the ileocecal valve. The incidence rate of small bowel carcinoids is estimated to be 1.2 to 6.5 cases per million persons. [5], [16], [17], [55], [56], [57], [58], [59] and [60] Autopsy studies cite a tenfold higher rate of carcinoids that were not evident clinically. [61] The median age at diagnosis is 60 years, but carcinoids have been detected in a wide range of ages (22 to 84 years). In the United States, the incidence of small bowel carcinoids is somewhat higher in men and in African Americans. [55]

No specific risk factors for carcinoids have been proven. However, cigarette smoking and alcohol consumption have been associated with an increased risk of developing small bowel carcinoids. [20] The incidence rates of carcinoids in first-degree relatives of patients with carcinoid tumors was higher than the expected population incidence rates. [62] The calculated cumulative probability that a first-degree relative will develop a carcinoid tumor by age 80 years was 1.5%. Persons with carcinoid tumors, particularly tumors of the small bowel, may have an increased incidence of coexisting noncarcinoid malignant diseases, [55], [61], [63] although a lack of association [60], [62] has also been reported. The noncarcinoid neoplasms are mostly adenocarcinomas of the gastrointestinal tract (50%), followed in frequency by lung, cervix, and prostate carcinomas. [61]

Pathology, Natural History, and Staging

Carcinoid tumors are malignant, despite their sometimes indolent course. Small bowel carcinoids, in particular, are associated with locoregional spread or metastases at the time of diagnosis. The tumors are usually intramucosal, and they rarely ulcerate to the lumen of the bowel. Small bowel carcinoids spread locally. When serosal breach has occurred, an intense local fibroblastic reaction is commonly seen. [64], [65] The desmoplastic reaction is responsible for many of the clinical findings in patients with small bowel carcinoid. Another characteristic feature of small bowel carcinoids is multicentricity. At the time of diagnosis, 35% of patients will have more than one tumor in the small intestine. [61], [66] Tumor size varies in several series, but most are 2 cm or less in diameter. Only 6% of small bowel carcinoids smaller than 1 cm have been associated with metastases, whereas 80% of patients with tumors larger than 2 cm will present with metastases. [67], [68] and [69] Microscopically, sheets of uniform cells with hyperchromatic nuclei are arranged in characteristic clumps (Fig. 80-3). Light microscopy of metastases to the liver cannot differentiate a carcinoid from an islet cell tumor.

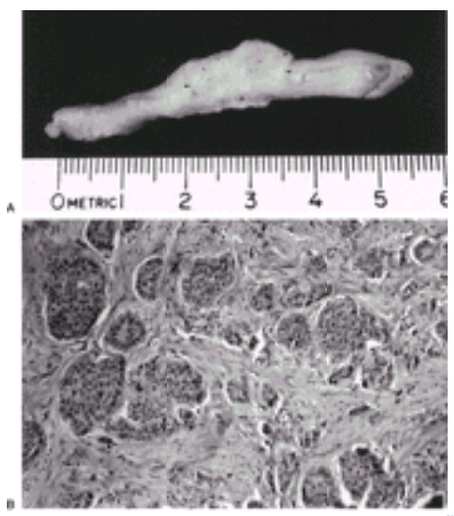


FIGURE 80-3. Carcinoid tumors of the small intestine. (**A**) A gross specimen of a nodular small intestinal carcinoid is demarcated by *arrows.* Carcinoid tumors develop deep in the mucosa and grow slowly, extending into the underlying submucosa and the overlying mucosa. (**B**) Histological section shows a small intestinal carcinoid tumor characterized by closely packed, round, regular and monomorphous cell masses, buds, and islands. Lumina and rosette-like structures are also present. Desmoplastic reaction is responsible for many of the clinical findings of small intestinal carcinoid tumors.

Carcinoid tumors can be classified in terms of the substances they secrete. [70] "Typical" small bowel (midgut) carcinoids secrete serotonin (5-HT), and they contain the enzyme dopa decarboxylase, which converts 5-hydroxytryptophan (5-HPT) to 5-HT. 5-HT is further metabolized and is excreted in the urine as the metabolite 5-hydroxyindoleacetic acid (5-HIAA). "Atypical" carcinoids (mostly foregut and hindgut) lack dopa decarboxylase and do not secrete 5-HT. [71] 5-HPT is released into the blood and can be measured in platelets and urine. Some 5-HPT is decarboxylated in the kidney before excretion, and 5-HT can thus be detected in the urine, but not 5-HIAA. [72]

Carcinoid tumors do not have a specific staging system. Most authors describe small bowel carcinoids in terms of size, degree of bowel wall invasion, presence or absence of lymph node involvement, and presence or absence of distant metastases.

The natural history of small bowel carcinoids is more favorable than that of adenocarcinomas of comparable stage. Five-year survival rates are 65% for local disease, 64% for regional disease, 36% for metastatic disease, and 55% overall for all patients with small bowel carcinoids. [55] However, the 25-year survival of patients with regional spread is reported to be less than 20%. [61] Patients with liver metastases and the carcinoid syndrome have a prognosis similar to that of patients with metastases who do not have the syndrome, but survival is lower for patients with carcinoid heart disease who develop valvular dysfunction. [61], [72]

Clinical Picture

Patients with small bowel carcinoids can present with nonspecific gastrointestinal symptoms, small bowel obstruction, intestinal ischemia, intussusception, gastrointestinal hemorrhage, hepatomegaly, or symptoms of the carcinoid syndrome. [73] Many of the signs and symptoms of small bowel carcinoids result from the intense desmoplastic reaction of the mesentery in proximity to the tumor, leading to partial small bowel obstructions and nonspecific gastrointestinal symptoms. [74] Complete bowel obstruction was the presenting symptom in 13% of patients in one series. [75] Nodal involvement causes compression of the main mesenteric arteries, leading to small bowel infarction. [76] Regional vascular thickening of medium-size arteries has been described with carcinoid, and this may act synergistically toward bowel ischemia formation. [77], [78] Gastrointestinal hemorrhage and intussusception are uncommon presentations of small bowel carcinoids. [79] Asymptomatic hepatomegaly from metastases is more common with carcinoids and islet cell tumors than with neoplasms of epithelial origin, so transaminases may not be abnormally elevated despite hepatomegaly and a large tumor burden. [80]

The most common clinical situation is a prolonged history of nonspecific abdominal pain that did not much alter the patient's lifestyle. The median time from onset of symptoms to diagnosis is reported to be 2 years, and it may be as long as 20 years. [61], [81] Investigation is not always sought by the patient, or has a low yield, thus prolonging the interval to diagnosis and treatment.

Patients with liver metastases from carcinoids may exhibit symptoms and signs of the *carcinoid syndrome,* caused by a variety of mediators. Overall, 10% to 18% of patients with small bowel carcinoid tumors will present initially with the carcinoid syndrome. [82], [83] and [84] Flushing of the face and neck occurs in most patients, and it may be episodic or permanent. Flushing is most commonly transient and simulates a mild sunburn involving the face, neck, and upper trunk (the *midgut flush* associated with ileal carcinoids). The so-called *foregut flush* is more intense, frequently associated with conjunctival injection and edema, and has a greater tendency to involve the body more diffusely. [61] Diarrhea is seen in 75% of patients and manifests as intermittent episodes of explosive, watery diarrhea resulting from intestinal hypermotility. Steatorrhea is rare. Abdominal cramping may accompany the diarrhea episodes about 50% of the time. Dyspnea can result from advanced carcinoid heart disease or, less frequently, from bronchoconstriction and asthma. Two thirds of patients with carcinoid syndrome will present with a large liver or an abdominal mass; 40% will have heart valve abnormalities that can be auscultated at presentation. Other less common signs include cyanosis (25%), peripheral edema, arthritis, and pellagra (2% to 7%). [82], [84]

Diagnosis

Carcinoid tumors of the small intestine frequently do not manifest clinically until regional spread has occurred or metastatic disease is present. Small, localized tumors are most often diagnosed incidentally during endoscopic or radiologic examinations performed for unrelated reasons.

Biochemical Markers Urinary 5-HIAA is the most common test used to screen for carcinoid tumors. Most small bowel carcinoids secrete 5-HT, and urinary 5-HIAA is subsequently high. [85] Unfortunately, it is not highly specific or sensitive for localized tumors because other neuroendocrine tumors can secrete 5-HT, and small carcinoids may secrete very small amounts of 5-HT. In contrast to localized tumors, urinary 5-HIAA is very sensitive and specific for the carcinoid syndrome because of the large tumor burden necessary to produce the features of the syndrome. [86] Elevated levels of more than 30 mg of 5-HIAA in 24 hours with provocative testing are diagnostic of the syndrome. False-positive 5-HIAA measurements can occur in diseases such as celiac disease, Whipple disease, and tropical sprue and when 5-HT–rich foods such as walnuts, bananas, and avocado are ingested, [87] but these levels are lower than those associated with carcinoid tumors. False-negative 5-HIAA determinations have been reported with the use of aspirin, levodopa, and phenothiazines. 5-HT overproduction can also be assessed by measuring platelet and urinary 5-HT levels, which are not affected by food or medications. [26] Urinary 5-HIAA, platelet 5-HT, and urinary 5-HT levels are elevated in all patients with small bowel carcinoids. [71] Chromogranin A is another useful biochemical marker of carcinoid tumors found in the serum. Chromogranin A is produced by most neuroendocrine tumors, and it has a very high sensitivity for all types of neuroendocrine tumors. The sensitivity of this substance for carcinoid tumors approaches 80%. [88] It can be used to monitor treatment response and recurrence or for diagnostic purposes in patients who do not have elevated 5-HIAA urine measurements. Neuron-specific enolase is less specific than chromogranin A, and this value is elevated in only 40% of patients with carcinoid tumors. [89]

Radiologic Imaging

Conventional barium studies of the small intestine may identify the primary lesion as a smooth, semilunar filling defect in the lumen or, more often, as wall thickening, lumen angulation, and nonspecific mass effect. Enteroclysis is generally more successful than conventional small bowel barium studies. [52], [90] CT scans of the abdomen are more sensitive for detecting regional and distal spread, rather than a localized primary tumor. Seventy percent of patients with regional spread will have some degree of mesenteric calcification. [91] One third of patients with small bowel carcinoid may have a characteristic bulky, conglomerate calcification of a mesenteric mass. [92] The mass may also be associated with linear soft tissue strands radiating into the surrounding mesentery in a stellate pattern and displacing adjacent loops

of bowel. CT scans are also useful to detect hepatic metastases and are somewhat less sensitive than magnetic resonance imaging examinations. [93]

Scintigraphy has long been used for detecting neuroendocrine tumors. Radioiodine (^{131}I or ^{123}I)-labeled metaiodobenzylguanidine (I-MIBG) has a sensitivity of 50% to 60% for diagnosing carcinoid tumors and their metastases. [94], [95] Somatostatin receptors are expressed by 87% of carcinoid tumors and are therefore ideal for detection using radiolabeled somatostatin analogs. Indium-labeled [^{111}In]pentetreotide has a sensitivity of 67% to 80% for detection of carcinoid tumors and their metastases. [96], [97] and [98] [^{111}In]pentetreotide is more sensitive than [^{123}I]MIBG and conventional CT scanning. [99] Octreotide-labeled scintigraphy is useful for diagnosis, to predict who will respond to octreotide therapy and to locate tumors before surgical debulking. [98], [99] Positron emission tomography with octreotide labeled with copper tetraazacyclotetradecane is as sensitive as indium-labeled pentetreotide scintigraphy for detecting metastatic carcinoids. [100]

Endoscopy Small bowel carcinoids have been detected endoscopically in the duodenum, proximal jejunum, and terminal ileum. The tumors generally appear as nodular, submucosal protuberances that are yellowish and shiny. Ulcerated lesions and pedunculated lesions have been reported as well. [101] Endoscopic biopsies of lesions are often unrevealing, because the tumors are frequently subepithelial. Snare polypectomy or endoscopic mucosal resection of the lesion is much more successful for obtaining a histological diagnosis. [102] Duodenal carcinoids can also be diagnosed by endoscopic ultrasound. Small, noninvasive lesions usually arise from the deep mucosa or submucosa, and at endosonography they appear as moderately hypoechoic masses in the second or third echo layers. [103], [104], [105] and [106] Determination of invasion with endoscopic ultrasound can be useful for staging purposes, [103] as well as to gauge the appropriateness of endoscopic resection as a therapeutic measure. [105], [107], [108]

Treatment

Localized Tumors Localized small bowel tumors should be resected and removed completely. [67], [70], [80], [109] Duodenal carcinoids are less likely to metastasize, and they can often be removed endoscopically, preferably after endosonographic verification that there is no deep tumor invasion. Pedunculated carcinoids can be removed by snare polypectomy, but subepithelial, sessile growths will require endoscopic mucosal resection after submucosal injection of saline. [102], [107], [108], [110], [111] and [112] Duodenal carcinoids that show spread beyond the submucosa and carcinoids of the jejunum or ileum should undergo surgical resection. The 5-year survival of patients with local disease who undergo wide excision and removal of regional lymph nodes approaches 75%. [113]

Tumors with Regional Spread Small bowel carcinoids with regional spread should also undergo wide surgical resection. [67], [70], [80], [109] In some case series, the 5-year survival of patients for whom careful excision of all peritoneal and nodal metastases was possible was 65% to 71%, [114], [115] compared with a 5-year survival of 38% for patients in whom regional tumor spread could not be completely excised. [115]

Distant Metastases The approach to patients with metastatic small bowel carcinoids is multimodal. [116] The goals of therapy are to suppress the symptoms of carcinoid syndrome, to improve quality of life, and, if possible, to prolong survival. Patients with symptomatic bowel obstruction can undergo segmental bowel resection or enteroenteric bypass and, if possible, surgical debulking of tumor deposits to decrease symptoms related to tumor burden. [61], [72] Targeted scintigraphic therapies may be more successful when the tumor burden is small. [117], [118] and [119] Wedge resection of solitary liver metastases and hepatic lobectomy have both been performed successfully in patients with metastatic carcinoid tumors. [120] Hepatic artery embolization results in ischemia of liver metastases, [121] decreases urinary 5-HIAA levels by 40%, and provides partial improvement of the symptoms of carcinoid syndrome in up to 80% of patients, [116], [122] with a survival benefit of a median increase of 2 years. [119], [123] Combination intraarterial chemotherapy followed by hepatic artery embolization may be more effective than embolization alone. Complete resolution of carcinoid syndrome symptoms was reported in 70% of patients for an average of 29 months using doxorubicin (Adriamycin) and iodized oil. [124] Similar success has also been reported with a combination of 5-fluorouracil, doxorubicin, cisplatin, mitomycin C, and polyvinyl alcohol, although with a higher rate of complications. [125] The mortality from hepatic artery embolization or chemoembolization is 0% to 2%. Common complications include fever, abdominal pain, nausea and vomiting, and transient transaminase elevations. Less commonly, myelosuppression, arterial thrombosis, liver abscess, and cardiac dysrhythmias may occur. [121], [126], [127] Receptor-targeted therapy with [^{131}I]MIBG and [^{111}In] pentetreotide is variably successful for controlling the symptoms of metastatic carcinoid syndrome. [70], [128], [129] and [130] One series reported complete response in 50% of 20 patients treated with [^{131}I]MIBG, [131] but a review of 229 patients reported a mean symptomatic response of only 23%. [132] [^{131}I]MIBG therapy has relatively few side effects, and it decreases the octreotide requirements for patients with metastatic carcinoid syndrome. [131] [^{111}In]pentetreotide and yttrium-labeled octreotide show promise for symptom control and reducing tumor burden. [128], [133] Symptom control is best achieved with octreotide, a somatostatin analog that inhibits 5-HT release. Therapy with octreotide reduces flushing in more than 70% of patients and diarrhea in more than 60%, [134], [135] and [136] it decreases urinary 5-HIAA levels in about 70% of patients, and it may prevent further tumor growth in patients who respond symptomatically to octreotide. [137] The newer long-acting somatostatin analog, lanreotide, can be injected once every 2 to 4 weeks, with comparable efficacy to that of daily administered octreotide. [137], [138] Complications of prolonged high-dose octreotide therapy include steatorrhea, gallstone formation, and development of resistance to the medication. The role of systemic cytotoxic chemotherapy in the management of metastatic carcinoid is debated. Response rates range between 10% and 40%, and toxicity is significant. [139], [140], [141], [142], [143], [144], [145], [146] and [147] At present, chemotherapy is reserved for patients who do not respond to the other treatment modalities described earlier. Patients treated with interferon had a median survival of 80 months, compared with a median survival of 8 months in patients treated with streptozocin and 5-fluorouracil. [148] The addition of interferon to octreotide monotherapy may improve symptom control, [149] or it may lead to stabilization of tumor progression. [150] Interferon in combination with 5-fluorouracil decreased 5-HIAA levels in 50% of patients, but tumor regression was seen in only 10% to 20%. [151] Combination interferon therapy and hepatic chemoembolization has been shown to improve the median survival of patients treated with interferon alone. [152]

MESENCHYMAL TUMORS

Of the *mesenchymal small intestinal tumors,* the most common variant is the *mesenchymal spindle cell tumor* (see Table 80-1). Some mesenchymal tumors represent clear-cut diagnostic entities (e.g., lipoma, ganglioneuroma), whereas most are more difficult to classify into any specific cell lineage. Tumors may share overlapping features of several diagnostic entities, or they may be histologically heterogeneous. Thus, the general term *gastrointestinal stromal cell tumors* (GISTs) was coined to describe the group. Most GISTs have a spindle cell appearance, but electron microscopy and immunostains indicate that most GISTs are not true smooth muscle tumors. [153], [154] It has been suggested that up to 94% of "smooth muscle" tumors in the older literature are actually not of muscle origin and would be classified today as GISTs. [153], [155]

Epidemiology

Approximately 60% of GISTs are found in the stomach, 20% to 30% occur in the small bowel, and the remaining tumors are scattered in the esophagus, colon, and mesentery. [154] Benign GISTs are three to four times more common than malignant GISTs. [153], [156], [157] Malignant GISTs are gut-specific sarcomas and represent 11% to 12.7% of all small bowel malignant tumors, with an incidence of 1.2 to 1.5 cases per million persons. [5], [16], [59], [158] Malignant GISTs are somewhat more common in male than in female patients, and they can be found in all ages, but the most common age range at the time of diagnosis is 50 to 59 years. [17], [159]

Kaposi sarcoma is thought to result from a herpes-type virus. [160], [161] Small bowel schwannomas can be seen in patients with neurofibromatosis. [162] Small bowel ganglioneuromas have been reported in association with the multiple endocrine neoplasia type 2 syndrome. [163]

Pathology

GISTs of the small bowel occur most frequently in the jejunum, followed by the ileum, and then the duodenum. Benign GISTs are generally smaller than 5 cm in diameter, but tumors larger than 20 cm have been reported. Malignant GISTs are greater than 5 cm at time of diagnosis in 80% of patients. [159], [164], [165] GISTs mostly arise from the muscularis propria and generally grow extramurally, but benign GISTs of the duodenum are more likely to grow intralumenally or intramurally. [159]

GISTs have a slightly different histological appearance than true leiomyomas, leiomyosarcomas, and schwannomas, with more cellularity and less cytoplasmic eosinophilia. Small bowel GISTs generally have a spindle cell–like appearance, but infrequently they can appear epithelioid. [153] GISTs can be classified by ultrastructural characteristics as myoid, neural, or ganglionic phenotypes, but the frequent presence of several different ultrastructural phenotypes suggests a common origin to all subtypes. [155]

Ninety-four percent of GISTs express CD117, the c-kit protooncogene protein that is a transmembrane receptor for the stem cell growth factor, [166], [167] and 70% to 80%

of GISTs express CD34, the human progenitor cell antigen. [168], [169] and [170] Less frequently, GISTs stain positive for actin and desmin (implying myogenic differentiation), but seldom are they S100 positive (a protein found in neuron-differentiated cells). [153] True leiomyomas and leiomyosarcomas are positive for actin and desmin, but do not express CD117 or CD34; and schwannomas express S100, but not CD117 or CD34. When spindle cell tumors are excised, immunohistochemical markers should be used to determine whether the neoplasm is a GIST or not. [154]

Observations that normal intestinal Cajal cells also express CD117 and CD34 pose the possibility that GISTs are of the same embryonic origin as Cajal cells. This would explain the similar morphology and intramural location of GISTs and true smooth muscle tumors. [153] The c- *kit* gene on chromosome 4 shows mutations of exon 11, especially in malignant tumors. [171] Some malignant GISTs also display loss of 14q and chromosome 22 on comparative genomic hybridization. [172] These genetic changes are not found on true smooth-muscle tumors.

Natural History and Prognosis

GISTs of the small bowel tend to invade locally, and they frequently present with peritoneal seeding or direct invasion to adjacent organs. [159], [165] Lymph node metastases are uncommon, but 31% to 41% of patients with small bowel GISTs will present with liver metastases.

Overall, 5-year survival rates are variable, ranging between 7% and 56%. [159], [165], [173], [174] and [175] In general, survival is favorable for patients without extralumenal tumor invasion or metastases and for tumors smaller than 2 to 5 cm.

Many factors have been suggested as prognostic indicators for GISTs (Table 80-5). [165], [173], [174], [176], [177] Unfortunately, except for tumor size and the number of mitotic figures, most of these factors are not found consistently. The most useful indicators of survival and the risk of metastases are the size of the tumor at presentation, the mitotic index (the number of mitotic figures per 50 high-power fields), and histological evidence of tumor invasion into the lamina propria. [154] None of the other factors in Table 80-5 have consistently been shown to predict prognosis accurately. GISTs should not be classified as “benign” or “malignant,” but rather by the risk for malignant behavior.

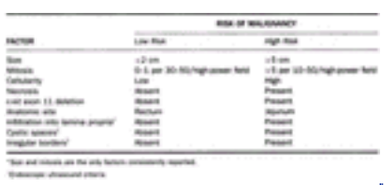


TABLE 80-5 Gastrointestinal Stromal Cell Tumors: Prognostic Factors*

Clinical Picture and Diagnosis

Most GISTs less than 5 cm in diameter are discovered incidentally at endoscopy, by barium x-ray studies, or in gastrointestinal specimens resected for other reasons. The small tumors are generally asymptomatic. In contrast, at least 80% of patients with larger tumors will have symptoms related to the GIST. [159], [165], [173], [174] and [175], [177] More than 50% of patients with tumors greater than 5 cm will have either a palpable abdominal mass or gastrointestinal hemorrhage. Bleeding from ulcerated small bowel GISTs is usually acute, and it may be brisk. [178] Thirty to 40% of patients with small bowel GISTs will present with abdominal pain, nausea and vomiting, or weight loss, and 40% of patients with ileal tumors may present with intussusception. [159]

Small bowel barium studies can detect GISTs 53% to 72% of the time. [179] CT has been reported to detect 89% to 98% of large GISTs. [180] Angiography may also be useful because of the characteristic “tumor blush” noted in more than 85% of tumors.

Endoscopic diagnosis of small bowel GISTs is difficult because most tumors are submucosal. If ulceration is present, biopsies can identify the tumor as a GIST. GISTs appear by endoscopic ultrasound as hypoechoic masses arising from the fourth echo layer (muscularis propria). Putative endosonographic criteria to differentiate benign from malignant GISTs include tumor size greater than 4 cm, irregular extralumenal margins, and cystic spaces. [181], [182] and [183] The positive predictive value for malignancy approaches 100% when any two of these findings are present.

Immunohistochemical staining should be performed on resected spindle cell tumors and biopsies of tumors suspected to be GISTs. CD117 positivity confirms that the tumor is a GIST.

Treatment

Segmental bowel resection, and not simple enucleation, is the treatment of choice for GISTs. [155], [156], [157], [158] and [159] Given the biology of these tumors, wide margins and extensive lymph node dissection are not necessary. Aggressive resection of organ segments that have been invaded with tumor and of hepatic metastases also appears to confer some improvement in survival. [175], [177], [184], [185], [186] and [187] Nonetheless, despite complete resection with negative margins, 44% to 80% of patients will suffer local or peritoneal recurrence. Most recurrences will occur within 2 years, but lag times of 10 years have also been reported. [188], [189]

Chemotherapy and radiation therapy have not been beneficial for patients with unresectable metastases and for patients with tumor recurrence. [190], [191] The tyrosine kinase inhibitor STI571, specific for the unregulated mutant c- *kit* (CD117) tyrosine kinase, was given to a patient with metastatic GIST for 11 months. The volume of metastases decreased fivefold, and no new lesions were detected. [192] Clinical trials with STI571 are eagerly awaited.

Although the accepted treatment of small bowel GIST larger than 4 or 5 cm is surgical resection, it is unclear what to do with smaller tumors. This gap in our knowledge is important because as endosonography use becomes more prevalent, more small GISTs will likely be diagnosed.

LYMPHOMAS

A *primary small bowel lymphoma* (PSBL) is characterized by the following:

1. The absence of palpable peripheral lymphadenopathy
2. A normal peripheral leukocyte count and differential
3. No mediastinal lymphadenopathy on a chest radiograph
4. Involvement of only the organs of the gastrointestinal tract and proximal regional lymph nodes
5. No involvement of the liver or spleen, unless by direct extension from the primary gastrointestinal tumor. [193], [194] and [195]

Extranodal lymphomas of the intestine can be classified according to the cell lineage (see Table 80-1) or according to their clinical characteristics. The latter method categorizes small bowel lymphomas into either PSBLs, which tend to be focal, or immunoproliferative small intestinal disease (IPSID), which is diffuse.

Epidemiology

Small bowel lymphomas represent 1% to 10% of all extranodal lymphomas [193] and 7% to 25% of all small bowel tumors. [5], [16], [59] Rates of small bowel lymphomas ranges from 0.9 to 3.6 cases per million. [5], [16], [58], [59], [193], [196], [197] The median age at diagnosis of PSBL is 67 years, whereas IPSID is generally diagnosed in the second or third decade of life. PSBL is more common in men than in women, at a ratio of 1.5:1. In the United States, small bowel lymphomas are more common in

whites than in African Americans. Population registries from the Middle East suggest a higher incidence than in Europe or the United States, [193] perhaps because of the high preponderance of IPSID. Studies from the United Kingdom, [196] Germany, [58] Denmark, [197] Ireland, [198] and the United States [17] show an increase of 30% to 50% since the early 1970s.

Risk Factors

Several diseases and immuno deficiency states are associated with an increased risk of developing PSBL. Patients with long-standing celiac disease have a 4% to 7% incidence of small bowel lymphomas. [199], [200] These are usually T-cell lymphomas and are frequently termed EATLs. Nodular lymphoid hyperplasia is usually an incidental finding in the terminal ileum. Diffuse nodular lymphoid hyperplasia is seen in primary immunodeficiencies, and it is associated with intestinal lymphomas. [201] Rarely, diffuse nodular hyperplasia and intestinal lymphoma can occur in the absence of immunodeficiency. [202] Some authors report an increased incidence in patients with inflammatory bowel disease using immunosuppressive medications, [203] some report an increased risk only in male patients with Crohn's disease, [204] and others report no increased risk at all. [205], [206] Small bowel lymphoma has been associated with congenital and acquired immunodeficiency states. [207] Patients with acquired immunodeficiency syndrome have a 30% risk of developing lymphomas, of which 10% to 25% will be PSBLs. [208] Immunosuppression after solid organ transplantation has been associated with an increased risk of lymphoma. Most extranodal lymphomas are PSBLs. There appears to be a correlation with the nature and dose of immunosuppression. For example, 30% of small bowel, heart, or lung recipients will develop lymphoma, whereas only 5% of kidney recipients will acquire the disease. [209]

IPSID occurs almost exclusively in the Middle East, Africa, Southeast Asia, and South America, regions in which poor hygiene and malnutrition are endemic. Malnutrition can impair the bowel's normally vigorous defenses in a variety of ways. [210] In response to the abnormal antigenic presentation, specific clonal lines of IgA-secreting B cells proliferate in the lamina propria. [156] The a-heavy chains can be detected in the blood of up to 69% of patients with early-stage IPSID. [211] IPSID is reversible when treated with antibiotics in the early, prelymphomatous phase. [212], [213]

Pathology

PSBLs occur most often in the ileum, followed by the jejunum, and then the duodenum. [5], [16], [58], [59] They are generally localized to one segment of the bowel, except in the case of *mantle cell lymphoma,* otherwise known as *multiple lymphoid polyposis* (MLP). The tumors have many different appearances. They may be large exophytic masses (Fig. 80-4 *A*), polyplike (Fig. 80-4 *B*), ulcerlike, or nodular. [214] Tumor growth and extension are frequently intramural for a prolonged period. Involvement of regional lymph nodes is present in approximately 50% of patients with most types of PSBL, except low-grade *mucosa-associated lymphoid tissue (MALT) lymphomas,* in which lymph nodes are involved in only 30% of patients. [214], [215], [216] and [217] IPSID tends to be a diffuse jejunal disease. Gross findings can range from thickened folds to discrete masses, [212], [218], [219] affecting a significant portion of the intestine in a contiguous fashion. [220] This feature probably explains the different clinical presentation of IPSID from PSBL, which usually has normal mural architecture in bowel segments not affected.

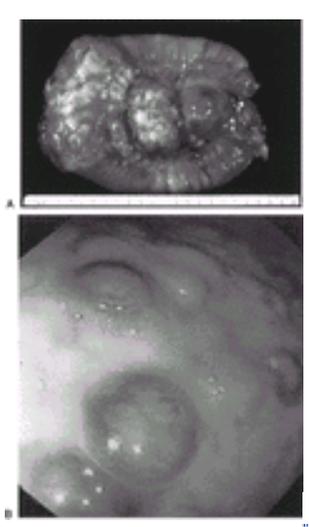


FIGURE 80-4. Primary small intestinal lymphomas. (**A**) A gross specimen of a primary lymphoma extensively involving the small intestine. (**B**) Mantle cell lymphomatous polyposis of the intestine seen endoscopically. The mucosa is studded with elevated polypoid nodules.

Most small bowel lymphomas are B-cell derived. The different histological patterns of small bowel lymphomas are summarized in Table 80-6. The diffuse large cell type—the most common intestinal lymphoma—and the small noncleaved cell type are similar histologically to their nongut counterparts. [199]

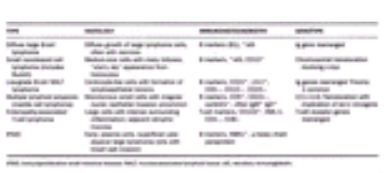


TABLE 80-6 Features of the Major Gastrointestinal Lymphomas

MALT lymphomas occur in nongut sites, but they are most common in the gastrointestinal tract. The characteristic cells are centrocyte-like cells that resemble the small cleaved centrocyte cells of nodal follicular cell lymphomas. [221] Mantle cell lymphomas (MLP) show monotonous small cells with irregular nuclei arranged in a pattern similar to the mantle zone of lymph node follicles. The mucosa and submucosa are most often involved. Despite the lack of deep invasion, MLP is a very aggressive lymphoma that presents frequently with distant lymph node and bone marrow involvement. [217], [222] EATL is a high-grade lymphoma composed of large cells resembling histiocytes. The adjacent mucosa displays the characteristic findings of celiac disease: villous atrophy, crypt hyperplasia, and abundant intraepithelial lymphocytes. Epithelial invasion is common, and the subsequent mucosal ulceration displays an intense inflammatory infiltrate. [223]

IPSID has a distinctive histological appearance that is characterized by a dense mucosal and submucosal cellular infiltrate throughout the length of the affected intestine. In the early (possibly benign) phase, plasma cells and small lymphoid cells predominate. In the late phase, dysplastic changes in the small cells are noted, with an increase in the proportion of large cells evolving into a diffuse large cell lymphoma. [212], [218], [219]

Staging and Prognosis

MLP, EATL, and IPSID tend to have the worst prognosis. [224], [225], [226], [227] and [228] The staging system for PSBL has evolved from the Ann Arbor classification originally devised for Hodgkin lymphoma (Table 80-7). Patients presenting with stage I PSBLs or low-grade MALT PSBLs have median 5-year survivals of 50% to 60% and 44% to 65%, respectively, and they have the best chance of cure. [229] The median survival of patients with stage II disease is only 20%. [224], [225], [226], [227] and [228]

Stage I Tumor confined to the gastrointestinal Single primary lesion or multiple noncontiguous lesions
Stage II Tumor extending in the abdomen from a primary gastrointestinal site Nodal involvement II1 local (paragastric or paraintestinal) II2 distant (mesenteric, paraaortic, paracaval, pelvic, inguinal)
Stage IIIE Penetration of serosa to involve adjacent organs or tissues
Stage IV Disseminated extranodal involvement or a gastrointestinal tract lesion with supradiaphragmatic nodal involvement

TABLE 80-7 Staging System of Primary Gastrointestinal Lymphoma

Two different staging systems have been devised for IPSID. One stages IPSID by the histological findings of prelymphomatous and lymphomatous stages, [219] and the other stages the disease by the bulk of tumor and extent of distant nodal and organ involvement. [220] Neither system has been validated in large survival cohorts.

In addition to the stage at presentation and the cell type, many criteria have been reported as being adverse prognostic factors for PSBL. These include a tumor larger than 7 cm, age greater than 70 years, the presence of B symptoms (fever, night sweats, and weight loss), mural invasion beyond the muscularis propria, distant lymph node involvement, unresectability of the tumor, elevated serum lactate dehydrogenase, elevated serum ß $_2$-microgobulin, and the presence of *BCL-2* genomic alterations. [224], [225], [226], [227] and [228], [230], [231], [232], [233], [234] and [235]

Clinical Picture and Diagnosis

The symptoms of PSBL are usually nonspecific and may continue for 4 to 18 months before a diagnosis is rendered. [224], [226] Abdominal pain, often described as crampy, is reported in 65% to 87% of patients with PSBL. Weight loss is seen in approximately 50% of patients with PSBL. The remaining symptoms are present in fewer than 30% of patients and include gastrointestinal hemorrhage, malaise, night sweats and fatigue, or acute abdomen resulting from bleeding or perforation. [220], [224], [225], [227], [228], [232] Small bowel obstruction is the presenting symptom in 5% to 12% of patients. Diarrhea and malabsorption are common in EATL. In fact, patients with celiac disease who become unresponsive to a gluten-free diet should be investigated aggressively for small bowel lymphoma. [223] The most common physical finding is a palpable abdominal mass, present in 30% to 50% of patients.

The symptoms of IPSID differ from those of PSBL. Nearly all patients with IPSID have diarrhea, weight loss, anorexia, and abdominal pain. Emesis and fever occur in 50% of patients. The physical examination in IPSID reveals clubbing of the fingers and ankle edema in 50% to 75% of patients. Later in the disease, ascites, hepatomegaly, and a palpable mass may be present. [212], [218], [219] and [220] Up to 70% of patients will have the a-heavy chain paraprotein in the serum.

Small intestine barium studies, especially enteroclysis, are quite accurate for detecting small bowel neoplasms. [179] CT is less sensitive for small lesions, but it detects most lesions larger than 5 cm. Small bowel lymphomas are categorized by CT as follows: primary, divided into circumferential and cavitary; diffuse, usually EATL; and mesenteric. [180] Because of their location, most small bowel lymphomas require laparotomy for tissue diagnosis. Those located within the proximal 60 cm of the jejunum can usually be identified and sampled for biopsy at enteroscopy. [101] The accuracy of endoscopic biopsy for diagnosing lymphomas is 82% on the first attempt, with an additional 10% on the second attempt. [162] Tissue should be sent both in formalin for standard studies and in saline for flow cytometry analysis. Characteristic findings at endosonography of an irregular hypoechoic mass disrupting the normal echo architecture may suggest the presence of a PSBL despite normal mucosal biopsies. [104] Unlike primary gastric lymphomas (especially MALT lymphoma), the role of endoscopic ultrasound in staging of duodenal lymphomas is not clear. Laparotomy for a diagnosis of lymphoma should be performed if the suspicion for IPSID is high despite normal endoscopic biopsies.

Patients with a confirmed diagnosis of small bowel lymphoma should undergo several studies for staging purposes, including a physical examination, indirect laryngoscopy to exclude involvement of Waldeyer ring, CT scan of the abdomen and chest to assess distant lymph node sites, bone marrow aspiration and biopsy, and lactate dehydrogenase and ß $_2$-microgobulin determinations. [209]

Treatment

The mainstay of treatment for PSBL is surgical resection. Complete resection with lymph node sampling is recommended to avoid a further operation. [209] The ability to achieve complete resection is associated with an improved survival compared with retained tumor after the surgical procedure [236], [238] and may improve response to chemotherapy and radiotherapy. [237], [238] and [239]

PSBL that cannot be completely resected is usually treated by chemotherapy and sometimes with the addition of radiotherapy. The chemotherapeutic regimen usually includes anthracycline, but there is no consensus on the best combination of drugs. [240], [241], [242] and [243] Aggressive chemotherapy causes perforation of the small bowel in 5% to 15% of patients. For some patients with diffuse MLP and EATL, chemotherapy is the only option, but response to chemotherapy is usually poor. The 5-year survival for patients with stage I and II MLP and EATL is approximately 20% to 25%. [214], [226], [227], [244]

IPSID is less amenable to surgical resection than PSBL because of the diffuse nature of the tumor and the low performance status of patients at the time of presentation. Nutritional support with total parenteral nutrition is essential, because malnutrition is severe and likely will not improve unless significant tumor regression is possible. Patients with IPSID in the prelymphomatous stage can be offered a trial of tetracycline or metronidazole for 6 to 12 months. [213], [218] Patients who do not respond to antibiotics or patients already in the lymphomatous stage may benefit from anthracycline-based chemotherapy. [245] A few patients with resectable stage I and II disease have a 5-year survival of 40% to 47%, whereas most other patients whose tumors are unresectable have a 5-year survival of only 0% to 25%. [246] A complete response rate of 62% to 69% and a 3.5-year survival rate of 58% to 70% have been reported in small groups of patients receiving anthracycline-based combination chemotherapy with or without tetracycline. [247], [248] and [249]

SECONDARY TUMORS

Metastatic tumors represent the most common tumors involving the small intestine in many series. Grossly, secondary tumors often present as submucosal nodules or plaques, and they may grow to form intramural masses that cause obstruction, intussusception, or perforation. Often, tumors present as stenotic lesions or infiltrative lesions simulating Crohn's disease. Metastases from melanoma and from carcinomas of the lung, testes, adrenal, ovary, stomach, large intestine, uterus, cervix, liver, and kidney to the small intestine have all been reported. Metastatic melanoma accounts for one third of small bowel metastases. [250] Lesions are typically multiple and present as polypoid lesions that may cause obstruction or intussusception. Tumors of the pancreas, stomach, colon, or mesentery may also involve the small bowel by contiguous spread.

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CHAPTER 81

Marc S. Levin

MISCELLANEOUS DISEASES OF THE SMALL INTESTINE

The diseases discussed in this chapter should be considered in patients presenting with gastrointestinal hemorrhage, abdominal pain, diarrhea, intestinal obstruction, or perforation that eludes diagnosis by routine gastroduodenal endoscopy and radiographic studies. Topics include ulceration of the small intestine, small intestine complications of drug therapy, necrotizing enterocolitis (NEC), and protein-losing gastroenteropathy (PLGE). Although most of these entities are infrequently encountered in clinical practice, differentiating them from diseases that are more common can yield important therapeutic benefits.

ULCERS OF THE SMALL INTESTINE

Many causes of *small intestine ulcerations* distal to the duodenum have been identified (Table 81-1). Most of these produce solitary ulcers with sharp borders and normal surrounding mucosa. With surgical resection or removal of offending agents, these ulcers seldom recur and are associated with a benign clinical course. The mortality from multiple ulcers occurring in an abnormal mucosa, as is occasionally seen with gluten-sensitive enteropathy (GSE) or unclassified sprue, chronic ulcerative (nongranulomatous) jejunoileitis (CUJ), or lymphoma, can be as high as 75%. [1] It is essential to attempt to define the underlying cause of small intestine ulcers if possible.

Infectious: tuberculosis, typhoid, cytomegalovirus, syphilis, parasites, strongyloidosis hyperinfection, *Campylobacter*, yersiniosis
Toxic: acute jejunitis (*jt*-toxin-producing *Clostridium perfringens*), arsenic
Inflammatory: Crohn's disease, systemic lupus erythematosus with high serum antiphospholipid levels, diverticulitis
Mucosal: gluten-sensitive enteropathy (jejunoileitis)
Neoplastic: malignant histiocytosis, lymphoma (primary), adenocarcinoma, melanoma, Kaposi sarcoma (secondary)
Vascular: mesenteric insufficiency, giant cell arteritis, vasculitis, vascular abnormality, amyloidosis (ischemic lesion)
Hyperacidic: Zollinger-Ellison syndrome, Meckel diverticulum, stomal ulceration
Metabolic: uremia
Drug-related: potassium chloride, nonsteroidal antiinflammatory drugs, antimetabolites
Radiation-induced: therapeutic, accidental
Idiopathic: primary ulcer, Behçet syndrome

TABLE 81-1 Causes of Small Intestine Ulceration

Primary (Idiopathic) Small Bowel Ulcers

Epidemiology and Etiology The diagnosis of primary small bowel ulcers is made after identifiable causes of small bowel ulcers have been eliminated and thus is unlikely to represent a distinct etiologic entity. With increasing recognition of secondary causes of small intestine ulceration, fewer patients fall into this diagnostic category. The incidence of idiopathic small bowel ulceration is unknown. A review based on 59 patients (6 were taking enteric potassium preparations) found an incidence of 4 small bowel ulcers per 100,000 new patients. [2] Men and women were affected equally, and all age groups were affected (Table 81-2). [2], [3], [4], [5], [6] and [7] These data suggest that primary small bowel ulceration is an uncommon disorder. However, because asymptomatic and uncomplicated intestinal ulcers are rarely detected, the true incidence of this entity is certainly higher than estimates, based on clinical experience.

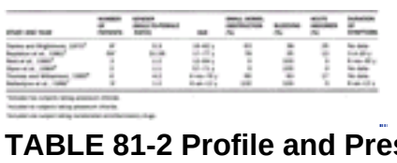

TABLE 81-2 Profile and Presenting Symptoms of Primary Small Bowel Ulcers

Clinical Features Symptomatic complications of small bowel ulcers include bleeding, perforation, and obstruction. The incidence of these complications is not known. Intestinal obstruction appears to occur more frequently than with duodenal ulcers. Intermittent crampy abdominal pain resulting from partial small bowel obstruction is the most frequent presenting complaint (63% to 100% of patients; see Table 81-2). Symptoms related to intermittent partial small bowel obstruction have been reported for 3 days to 20 years preceding presentation. [2] Patients with small intestine obstruction or perforation also can present with an acute abdomen. Although ulcers are more commonly seen in the ileum, perforated ulcers are more common in the jejunum (78% versus 11%). [2] Because ileal ulcers, which are less likely to perforate, are more prevalent, the overall incidence of perforation is only 22%. Gastrointestinal bleeding is also common, especially in younger patients, who present with intestinal hemorrhage or with occult blood loss. Patients presenting with gastrointestinal bleeding often have symptoms consistent with a history of intermittent intestinal obstruction as well. The diagnosis of small intestine ulceration should be considered if intestinal bleeding and evidence of small intestine obstruction coexist.

Pathology Most primary small intestine ulcers (75%) are located in the middle to distal ileum. [1] In one report, a solitary ulcer was found in 70%, two or three ulcers in 20%, and more than three in 10% of patients. [2] Ulcers typically have sharply demarcated borders and vary in diameter from 0.3 to 5 cm. [2] The pathological features are nonspecific and are identical to those of peptic ulcerations. [8] A marked eosinophilic infiltrate is sometimes present. [9] The surrounding mucosa is usually normal. Granulomata, sinus tracts, and pathological vascular changes are not seen. Granulation tissue and fibrosis that extend deep into the bowel wall may produce intestinal stenosis. The absence of granulomata and the normal surrounding mucosa are features that help to distinguish idiopathic ulcers from those seen in Crohn's disease and in CUJ.

Diagnosis Both detection of intestinal ulceration and exclusion of known etiologic agents are required to support a diagnosis of primary small bowel ulcers. A careful history is necessary to identify exposure to potentially ulcerogenic substances such as nonsteroidal antiinflammatory drugs (NSAIDs) and enteric-coated potassium preparations. Prior abdominal surgery and other risk factors that predispose to intestinal ischemia should be identified. Assessment of immune status also may be

useful. It is important to exclude cytomegalovirus infection in immunocompromised patients because of the propensity for cytomegalovirus ulcers to bleed or perforate. Other infectious causes, including *Salmonella typhi,* need to be excluded. [10], [11] There are no pathognomonic biochemical abnormalities, and the only laboratory tests that are useful for diagnostic purposes are those that help to establish an alternative diagnosis. Small intestine ulcers can be detected radiographically, endoscopically, or at laparotomy. Although small intestine ulcers cannot be detected by plain abdominal radiographs, small bowel dilation suggestive of obstruction and pneumoperitoneum indicative of perforation may be detected. Conventional contrast radiography of the small intestine rarely identifies primary ulcers of the small intestine. [2] Enteroclysis is a superior technique for demonstrating small intestine mucosal lesions (see Chapter 152). It should be considered the radiographic procedure of choice in the evaluation of suspected ulcers of the small intestine. Although intestinal strictures may be readily detected, small intestine ulcers are seldom demonstrated radiographically. Ileal ulcers can, on rare occasions, be detected if contrast material is refluxed into the terminal ileum; however, the yield from barium enemas does not justify their use for detecting small bowel ulcers. Other radiographic techniques, such as technetium 99m (^{99m}Tc)–labeled red cell scanning and mesenteric angiography used for the evaluation of gastrointestinal bleeding, or ^{99m}Tc pertechnetate for the diagnosis of a Meckel diverticulum, may lead indirectly to the detection of small intestine ulcerations. Small intestine enteroscopy is a useful adjunct for the diagnosis of small intestine lesions because of improvements in equipment and technique. [12], [13], [14], [15] and [16] In the setting of obscure gastrointestinal bleeding, for example, a diagnosis was made in 50% of 258 patients, all of whom had negative results of enteroclysis. [17] These data suggest that enteroscopy is useful in the evaluation of small bowel ulcers, although it has not been specifically evaluated for this purpose. Intraoperative enteroscopy with laparotomy or laparoscopy also can be a valuable adjunct in difficult cases. [18], [19] and [20] Based on animal studies and clinical experience in detecting small bowel angiodysplasias, wireless capsule endoscopy may prove superior to push enteroscopy for the detection of small intestinal ulcers. [21], [22] and [23] Despite the refinement of enteroclysis and enteroscopy, many symptomatic idiopathic small intestine ulcers are not detected without surgical intervention. In the patient with persistent gastrointestinal blood loss or abdominal pain with evidence of small bowel obstruction, surgical exploration with or without intraoperative enteroscopy should be given serious consideration (see Chapter 161).

Therapy The therapy for primary small intestine ulcers is dictated by the severity of complications. Emergency surgical intervention is required if an acute abdomen or pneumoperitoneum indicative of intestinal perforation is present. Life-threatening intestinal bleeding is an indication for diagnostic and therapeutic mesenteric angiography or surgical exploration (see Chapter 33). The best therapy for ulcers that present with less serious complications is also surgical resection. Detection of small bowel ulcers is usually readily accomplished at laparotomy. Intraoperative enteroscopy may be a useful adjunct. Most ulcers can be managed by surgical resection with end-to-end anastomosis, providing adequate material for pathological analysis with a low incidence of complications and ulcer recurrences. Simple oversewing of primary small bowel ulcers is not recommended. With such an approach, the amount of material available for pathological analysis is limited, and the risk of ulcer recurrence is high. Medical management with antiinflammatory or immunosuppressive agents is not effective therapy for intestinal ulcers or strictures. Furthermore, unsuspected perforation is often found at surgery. [2]

Drug-Induced Small Bowel Ulcers

The entrapment of a pill or capsule before dissolution or the release of large concentrations of a drug in a segment of intestine can result in localized ulceration because of physical pressure or specific cytotoxic effect. NSAIDs remain the most common cause of drug-induced small intestine ulcers. In the 1960s, enteric-coated potassium chloride (KCl) was introduced and was soon recognized to be an intestinal ulcerogen. Ferrous salts, digoxin, corticosteroids, zirconium, and clofazimine have all been implicated as ulcerogens as well. [24]

Nonsteroidal Antiinflammatory Drugs These drugs are a heterogeneous group of organic acids that inhibit prostaglandin synthesis and have analgesic antiinflammatory and antipyretic properties. [25] Although they have been used for decades in the treatment of inflammatory arthropathies, their use now includes many nonrheumatologic problems. [25] Many generic NSAIDs are available without prescriptions. As a result, NSAIDs are among the most commonly used drugs worldwide. Numerous publications have addressed the magnitude and cost of NSAID-associated gastrointestinal complications, which have been described as an emerging epidemic. [26], [27], [28], [29], [30], [31], [32], [33], [34], [35] and [36] Because of the increased use of NSAIDs, the introduction of more potent NSAIDs, and improvements in small intestine diagnostic methods, small bowel enteropathy is a more frequently diagnosed complication of NSAID use. Multiple ulcerations, mucosal diaphragms, ileal strictures, and perforations have all been associated with NSAID use. [30], [37], [38], [39], [40] and [41] Although the rate of small bowel ulcers with cyclooxygenase-2–selective inhibitors is unknown, the risk is likely to be less than for generic NSAIDs. The epidemiology of NSAID-induced enteropathy is not well characterized. Up to 70% of patients who take NSAIDs have evidence of increased intestinal permeability or inflammation, [30], [42], [43], [44], [45] and [46] but most are asymptomatic. An autopsy study showed the presence of nonspecific small intestine ulcers in 8.4% of 249 patients who had NSAIDs prescribed during the 6 months before death, compared with an incidence of 0.6% in 464 patients who had not used NSAIDs. [47] Three of the long-term users of NSAIDs were found to have died of perforated nonspecific small intestine ulcers. Small intestine erosions or ulcerations were detected by small bowel enteroscopy in 7 of 15 iron-deficient patients receiving long-term NSAID therapy for rheumatoid arthritis. [48] All the patients had negative results of upper and lower endoscopy. These studies indicate a high prevalence of unsuspected gastrointestinal lesions in patients taking NSAIDs. A metaanalysis of studies examining the association between NSAIDs and serious adverse gastrointestinal events indicated that NSAID users are at three times greater risk than are nonusers. [47], [49] Independent risk factors seem to be age exceeding 60 years, previous NSAID complications, and concomitant steroid use. This study did not analyze small bowel effects independently, so it is not certain whether this risk profile is applicable to NSAID-induced small intestine enteropathy.

Pathophysiology. Increased intestinal permeability, which enhances susceptibility to lumenal macromolecules and toxins, has been implicated in the pathogenesis of NSAID-induced enteropathy. [50] This defect in permeability is evident within hours after NSAID administration, [30], [51] and in rats it may be ameliorated by pretreatment or simultaneous treatment with glucocorticoids, sulfasalazine, or tetracycline. [52] The mechanisms by which NSAIDs injure the small intestine mucosa are unknown but are thought to include a role for enteric bacteria, the enterohepatic circulation, neutrophil infiltration, and suppression of prostaglandin synthesis. [53], [54] Because cyclooxygenase-1–mediated synthesis of prostaglandins appears to be less important as a cytoprotective mechanism in the small intestine, NSAID-mediated cyclooxygenase inhibition is not thought to be the major pathogenic mechanism in NSAID-induced enteropathy. [55], [56] Furthermore, cyclooxygenase-1 knock-out mice had no apparent intestinal disorders. [57] Although nitric oxide derivatives of NSAIDs are equally potent suppressers of prostaglandin synthesis as their parent compounds, the magnitude of small intestinal injury is markedly attenuated with these agents. [58], [59], [60] and [61] In rats, diclofenac, but not nitrofenac (a nitric oxide derivative that does not undergo extensive enterohepatic circulation), caused increases in epithelial permeability, gram-negative bacterial colonization, and intestinal ulceration. [62] /SUP>In these rats, the increased bacterial load, which occurred after the changes in intestinal permeability, may have exacerbated the NSAID injury. [53] These studies suggest that NSAIDs primarily alter intestinal permeability through local rather than systemic effects and suggest a pathogenic role for enteric bacteria. Additional studies indicate that NSAID-induced microvascular leakage also contributes to NSAID-induced enteropathy. [63] In rats, NSAID-induced plasma leakage requires induction of calcium-independent nitric oxide synthase and occurs only in the presence of gut bacteria. [64] The relative importance of local and systemic effects in the pathogenesis of NSAID-induced enteropathy was also addressed in human volunteers who received either indomethacin or the prodrug nabumetone. Because only indomethacin increased intestinal permeability, local effects of NSAIDs appear to be more important. [65] Another prodrug, sulindac, was compared with indomethacin, with similar results. [66] To date, clinical trials comparing cyclooxygenase-2–selective inhibitors with nonselective cyclooxygenase inhibitors have not had the power to address the comparative magnitudes of small intestinal enteropathy. A proposed cyclooxygenase-independent direct mechanism for NSAID damage is inhibition of glycolysis and the tricarboxylic acid cycle, leading to reduced cellular adenosine triphosphate production and uncoupling of oxidative phosphorylation. [55], [67], [68] Coadministration of glucose and citrate with indomethacin to humans was shown to prevent the increased permeability seen with indomethacin alone, [67] a finding compatible with this hypothesized mechanism. NSAID-associated intestinal injury primarily affects the distal small intestine. Intestinal ulcers induced by NSAIDs are pathologically identical to primary ulcers and to other drug-induced ulcers. Strictures are seen in up to 5% of patients receiving long-term NSAID therapy for arthritis. [30] In addition to strictures that are indistinguishable from those seen in Crohn's disease and other disorders, apparently unique, diaphragmlike strictures have been described (Fig. 81-1). [37], [39], [69], [70] and [71] These strictures encroach on and narrow the lumen to an opening as small as 1 mm in diameter. [69] They usually occur as multiple strictures separated by only a few centimeters of normal bowel. [69] Submucosal fibrosis replacing or merging with the muscularis mucosae is characteristic. [37], [69] Mild inflammation of the overlying mucosa is invariably present, with or without shallow ulceration. [37] The adjacent mucosa is usually completely normal. Patients with this "diaphragm disease" present with weight loss, hypoalbuminemia, anemia, and intermittent vague abdominal pain. [69] The diaphragm-like strictures are difficult to detect radiologically and can be missed at laparotomy unless an enterotomy is performed. [69], [70] and [71] Small bowel enteroscopy, which is useful for detecting intestinal ulcers, [48] may also be useful for detecting NSAID-related intestinal abnormalities. [72]

FIGURE 81-1. Nonsteroidal antiinflammatory drug–induced small intestine strictures. (**A**) Diaphragm strictures of the small intestine are apparent in resected intestine. (**B**) Enteroclysis study in a

patient taking nonsteroidal antiinflammatory drugs. Diaphragm strictures (*arrows*), which were found at surgery, are difficult to appreciate and resemble exaggerated plica circularis. (From ref. [55].)

Morphologically, the presentation of NSAID-related enteropathy is often similar to that of Crohn's disease, with transmural injury that can cause stricture formation. [55, 68] In addition, although most patients with NSAID-associated small intestine enteropathy are asymptomatic, some patients receiving long-term treatment present with clinical features of Crohn's disease. [25] In a prospective study, 10% of patients with colitis not secondary to Crohn's disease were thought to have NSAID-induced colitis, [73] but NSAIDs may exacerbate Crohn's disease and ulcerative colitis. [28, 74, 75 and 76] These reports emphasize the importance of obtaining an accurate drug history for all patients presenting with an illness resembling Crohn's disease. NSAIDs have been reported to be associated with a variety of other intestinal disorders, including collagenous colitis, [77] nonceliac flattening of small intestinal mucosa, [78, 79 and 80] celiac sprue, intestinal malignant diseases, [80] and perhaps diverticulitis. [55] Nevertheless, the available data are not sufficient to establish a true association between NSAID use and any of these conditions. [68]

Therapeutic Considerations. There are no practical means to diagnose NSAID-associated enteropathy routinely. Noninvasive techniques to assess NSAID effects on the small intestine are being studied as alternatives to endoscopy or surgery. [81] Most patients who take NSAIDs appear to have intestinal inflammation by indium 111 leukocyte scans and fecal excretion, [43] and increased intestinal permeability by chromium 51 (^{51}Cr)-EDTA absorption. These abnormalities usually resolve after the medications are discontinued, but inflammation may persist for up to 16 months. [30] Discontinuation of NSAIDs should be sufficient for treatment of uncomplicated ulcers. Surgical intervention is required for therapy of intestinal perforation or symptomatic strictures. The optimal regimen for patients for whom there is no good alternative to continued NSAID use has not been established. Based on the observation that nabumetone and sulindac did not increase intestinal permeation to ^{51}Cr-EDTA, switching to these or other prodrugs may be beneficial. Observations in rats indicate that switching to NSAIDs that are less potent inhibitors of cyclooxygenase also may be helpful. [82, 83] The hypothesis that cyclooxygenase-2–selective NSAIDs, by preserving the cytoprotective effects of prostaglandins synthesized by cyclooxygenase-1, may be less enteropathic than nonselective NSAIDs has not been directly addressed by the clinical trials completed to date. A double blind crossover study demonstrated that unlike indomethacin, cyclooxygenase-2 inhibition with rofecoxib did not increase intestinal permeability in healthy subjects. [84] In another double-blind, crossover study, nimesulide, another cyclooxygenase inhibitor, did not increase intestinal permeability or excretion of calprotectin, a marker of intestinal inflammation. [85] Delivery of NSAIDs encapsulated in liposomes was protective in a rat study. [86] The degree of ulceration in rats was related to the amount of active drug excreted in bile. [82, 83, 87] Human studies have not established that the incidence of small intestine enteropathy or upper gastrointestinal bleeding can be reduced by using prodrugs or NSAIDs with decreased biliary excretion of active drug or enteric-coated or slow-release formulations. [81] Germ-free rats are resistant to indomethacin-induced intestinal lesions, [88] and antibiotics can reduce the incidence of indomethacin-induced small intestine ulcers in rats. [89] In humans, metronidazole [66] and sulfasalazine [90] reduce intestinal inflammation, a response not yet proven to correspond to prevention of or recovery from NSAID-induced complications. In studies using rats, pretreatment or concurrent treatment with glucocorticoids, sulfasalazine, tetracycline, sucralfate, [91] pentagastrin, [92] naloxone (tested because morphine potentiates the ulcerogenic effect of indomethacin), [93] clonidine, [94] or thromboxane synthetase inhibitors [52] reduced indomethacin-induced permeability and inflammatory changes in rats. Drugs that exacerbated the intestinal toxicity of indomethacin in rats include cyclosporine [95] and morphine. Although prostaglandin E analogs such as misoprostol and enprostil prevent NSAID-associated injury of the stomach and duodenum, [96, 97, 98, 99 and 100] their effects in the remaining small intestine are not known. Protection was demonstrated in animal models. [101, 102 and 103] In human subjects, high-dose misoprostol prevented NSAID-induced permeability changes in one study, [104] but therapeutic doses did not. [65]

Potassium Chloride The introduction of enteric-coated KCl tablets in the 1960s was associated with an unacceptably high incidence of small intestine ulceration, attributed to high lumenal concentrations of KCl resulting from the rapid dissolution of the tablets in the small intestine. Slow-release formulations using a wax-polymer matrix (dissolution in approximately 4 hours) or microencapsulation (dissolution in 8 to 10 hours) were developed in an attempt to eliminate this adverse effect; the incidence of gastrointestinal ulceration was reduced but not eliminated. Two cases of small intestinal ulceration occurred from 1970 through 1983 (3 in 100,000 patient-years of slow-release wax matrix tablet use), compared with 56 cases from 1960 through 1965 (65 in 100,000 patient-years of enteric preparations). [105] Regardless of the preparation used, the risk of ulceration is increased if intestinal transit is delayed by concurrent medications, general debility, or advanced age. [106] The clinical presentation, diagnosis, and treatment of patients with KCl-induced small intestine ulcers or strictures are the same as those for patients with primary or NSAID-induced ulcers. Although the ulcers can occur anywhere along the gastrointestinal tract, they are most common in the distal ileum. [107]

Other Drugs Parenteral gold therapy also has been associated with enterocolitis, with edema and ulceration limited to the ileum. [108, 109 and 110] This rare complication is most common in middle-aged women and can occur shortly after the initiation of therapy. This disorder has a high mortality rate; thus, prompt cessation of gold therapy is essential. Therapy includes bowel decompression and antibiotics. Steroids are frequently used, although efficacy has not been proven. Octreotide was used successfully in one case. [111] Other drugs for which there are reports of small intestine ulcers or perforation include corticosteroids, [112] cytarabine [113] and other chemotherapeutic agents, digoxin, [114] and ferrous sulfate preparations. [14]

Small Bowel Ulcers Associated with Systemic Disorders (See Chapter 128)

Behçet Syndrome *Behçet syndrome* is a systemic disease that affects the skin, joints, vascular system, central nervous system, and intestinal tract. Oral and genital ulcers are a common feature of the syndrome; intestinal ulceration has been described in fewer than 1% of all patients with Behçet syndrome. [115] The ileocecal region is most frequently involved. [116] Multiple deep ulcers in a background of minimally inflamed mucosa are characteristic. The lesions predominate at sites coinciding with intramucosal lymphoid tissue. [117] The ulcers bleed easily and are often penetrating. The abnormalities are readily differentiated from those of Crohn's disease or ulcerative colitis. [116, 118] The optimal treatment of Behçet syndrome has yet to be established. No medical treatment significantly alters the natural course of this syndrome. Immunomodulation with corticosteroids, methotrexate, colchicine, thalidomide, [119, 120 and 121] cytotoxic agents, cyclosporine, and tacrolimus (FK506) have been used for treatment of multisystem Behçet syndrome. [122] Recombinant interferon-a may benefit patients with systemic involvement including intestinal ulcers. [123, 124, 125 and 126] Surgery with wide resection margins should not be delayed in patients with complicated intestinal cases, although ulcer recurrence is common postoperatively. [116, 127, 128] Most recurrences occur within 2 years of surgery and consisted of one or two deep ulcers at or near the anastomotic site. [119]

Collagen Vascular Diseases Patients with *systemic lupus erythematosus* (SLE) can have intestinal ulcerations. [129] Microthrombosis and vasculitis with intestinal ischemia are the likely mechanisms for this uncommon complication of SLE. Mesenteric vasculitis often presents with vomiting, fever, and hematochezia. It can progress to bowel infarction, which is associated with high mortality. Mesenteric vasculitis presenting with small bowel ischemia and stricture has also been reported in patients with other inflammatory disorders, [130] including rheumatoid arthritis, scleroderma, polyarteritis nodosa, Henoch-Schönlein purpura, Wegener granulomatosis, giant cell arteritis, Churg-Strauss syndrome, and Sézary syndrome. [131] Spondylarthropathies have also been associated with an increased incidence of ileal ulceration, although this is most often caused by concurrent inflammatory bowel disease. [132] Patients with acquired antiphospholipid antibodies and SLE or related autoimmune diseases are at increased risk of developing intestinal ischemia and hemorrhage related to mesenteric arterial thrombosis. [133, 134] *Cryptogenic multifocal ulcerous stenosing enteritis* is an idiopathic syndrome characterized by intermittent bouts of intestinal obstruction and ulcerative stenosis and steroid responsiveness. [135] Patients present with multiple jejunal and proximal ileal stenoses associated with superficial ulcers but share no other characteristics of Crohn's disease. The presence of systemic symptoms (weight loss, fever, malaise) in most patients, the responsiveness to steroids, and the demonstration of C2 deficiency in one reported case suggest that this syndrome is a variant of polyarteritis nodosa or an unclassified independent vasculitis. [135]

Diffuse Small Bowel Ulceration and Concurrent Malabsorption

A rare clinical syndrome consisting of malabsorption, abdominal pain, and multiple nonmalignant ulcers of the small intestine has been described. [136] Typically, these patients are in the sixth or seventh decade of life and have long-standing GSE. In such patients, the disorder is characterized by extensive villus atrophy and small bowel ulcerations that fail to improve on gluten withdrawal. There is also a distinct subgroup of patients who do not have GSE. The term *chronic ulcerative (nongranulomatous) jejunitis* (CUJ) and synonyms such as idiopathic mucosal enteropathy, idiopathic chronic ulcerative enteritis, unclassified sprue, malignant histiocytosis, and enteropathy-associated T-cell lymphoma (EATL) have been applied to this latter group of patients. The nomenclature reflects the poor understanding of the etiology and pathophysiology of CUJ. [136, 137] For the purposes of this chapter, *CUJ* is used to describe this disease in patients with and without evidence of GSE.

The typical histological features of CUJ are partial or total villus atrophy, mucosal ulceration, crypt hyperplasia, and an inflammatory cell infiltrate. [138, 139] The loss of intestinal villi results in a marked decrease in the absorptive area of the intestine, and the aborted maturation of the enterocytes results in lower levels of digestive enzymes on the intestinal brush border. The effects of these disturbances are malabsorption of all dietary components and a variable degree of PLGE. Although the mechanism is poorly understood, villus atrophy appears to be related to infiltration by activated T cells. [140]

Etiology CUJ that occurs without a prior history of GSE is usually more extensive in distribution and fails to respond to gluten withdrawal. CUJ and GSE are both associated with splenic atrophy of unknown origin, and most cases of CUJ are preceded by a variable period of suspected or proven GSE. [1, 141, 142 and 143] Nevertheless, it seems clear that CUJ can develop without GSE. [144, 145] Although small bowel lymphoma can resemble GSE clinically, [146] there is strong evidence that lymphomas arise in chronic GSE. [147, 148, 149, 150 and 151] The incidence of malignant degeneration may be decreased by a strict gluten-free diet. [152] In contrast to primary small intestine lymphomas, the typical lymphoma associated with GSE is derived from T cells. [153, 154 and 155] These lymphomas, which are referred to as *enteropathy-associated T-cell lymphomas* (EATLs), are difficult to differentiate from those of CUJ. Histologically, both EATLs and CUJ exhibit villus atrophy, ulceration, and mucosal infiltration by activated T cells. Both disorders usually present with splenic atrophy. The major diagnostic difficulty results from the observations that EATL may be indolent in its early stages,

and patients with CUJ can develop lymphoma several years after diagnosis. In some cases, retrospective examination of biopsy samples obtained at the time of diagnosis of CUJ have revealed typical malignant cells. [138] , [141] , [156] Using the polymerase chain reaction, clonal rearrangements of the T-cell receptor were found in tumor specimens and in enteropathic bowel without evidence of tumor involvement from the same patients. [157] , [158] Analysis of intestinal biopsies from patients with CUJ and refractory GSE revealed that the monoclonal cytologically normal, noninvasive intraepithelial T lymphocytes share an aberrant immunophenotype with EATL. [159] In one series, all the patients with CUJ had metastatic disease at the time of diagnosis, [138] but in a different retrospective study, no evidence of preexisting lymphoma was detected. [142] These data indicate that CUJ is a significant risk factor for the development of lymphoma and suggest that this development may be invariable. Non–T-cell lymphomas, such as immunoproliferative small intestinal disease and Mediterranean (B-cell) lymphoma, can also be accompanied by mucosal atrophy and ulceration and may have all the features of CUJ (see Chapter 80). This group may have a dramatic response to antibiotics or cytotoxic agents. [160] , [161] , [162] and [163] These lymphomas cannot be differentiated solely based on morphology; thus, it is important to determine cell surface markers for proper diagnosis.

Clinical Features The age at diagnosis of CUJ tends to be older than it is for typical GSE. Childhood cases have not been reported. The onset of CUJ is indolent. Patients with CUJ with and without known GSE were symptomatic for a mean of 13 and 4.5 years, respectively. Most patients present with chronic symptoms that are typical of malabsorption syndromes. Midepigastric pain and weight loss associated with steatorrhea and diarrhea are common. [142] Shortly after presentation, 20% to 30% of patients have ulcer complications such as small bowel obstruction (30%), melena (25%), and perforation (22%). Along with symptoms caused by these complications, patients with CUJ may have low-grade fever and signs of malnutrition.

Pathology Multiple superficial or deep ulcers are invariably present in the stomach, small bowel, and colon. [142] , [164] Splenic atrophy is often present. Lymphomatous infiltration of lymph nodes, liver, spleen, and bone marrow is common with concurrent lymphoma. [138] According to Isaacson, [139] there are “variable degrees (usually severe) of villus atrophy, crypt hyperplasia, intraepithelial lymphocytic infiltration, irregularity of surface enterocytes, and a dense lamina propria infiltrate consisting almost solely of plasma cells with a variable number of eosinophils and occasional neutrophils.” Invasion of the epithelial layer may cause ulceration (Fig. 81-2). Subepithelial collagen deposition, which is also seen in GSE, and marked fibrosis of the ulcer base also may be present. Crypt hyperplasia and villus atrophy are seen in the adjacent mucosa, and the remaining mucosa is usually normal. It is imperative that multiple sections be exhaustively examined for malignant cells to avoid missing the diagnosis of lymphoma. Neoplastic cells are derived from T cells. [153] , [154] and [155] , [165] They resemble histiocytes, with minimal or no atypia, and the presence of intracellular erythrocytes, platelets, and cell debris is a common feature (Fig. 81-3). [138] Confusion with Hodgkin disease may occur because of similarities with Reed-Sternberg cells. [146] , [147] , [166]

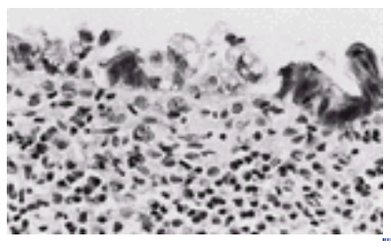

FIGURE 81-2. The malignant chronic ulcerative nongranulomatous jejunoileitis cells have invaded the surface epithelium and produced an ulcer. An exuberant plasma cell infiltrate is seen in the subepithelial tissue (original magnification ×400). (From ref. [139] .)

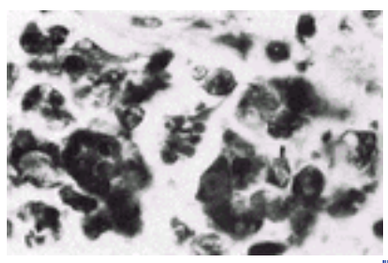

FIGURE 81-3. Chronic ulcerative nongranulomatous jejunoileitis tumor cells show typical erythrophagocytosis (original magnification ×1000). (From ref. [138] .)

Diagnosis The diagnosis of CUJ should be considered in patients with established or apparent GSE who present with worsening malabsorption despite continued compliance with gluten-free diets. CUJ is essentially a diagnosis of exclusion, whether or not a history of celiac disease is present, unless lymphoma is detected. Disorders presenting with malabsorption and intestinal ulceration that must be excluded include Crohn’s disease, ischemic enteritis, radiation enteritis, lymphoma, drug-induced ulceration, vasculitis, Zollinger-Ellison syndrome, and Whipple disease. The patient with suspected CUJ should be assessed for malnutrition, and a careful evaluation of the extent of disease should be undertaken. Malabsorption and PLGE account for hypoalbuminemia, and hypoglobulinemia and deficiencies of divalent cations, iron, and folate are common. Hyposplenism is often present. [143] Intestinal perforation should be ruled out if leukocytosis, with or without fever and peritoneal signs, is present. The diagnosis of CUJ is rarely made radiographically. Evidence of bowel thickening and of complications such as small bowel perforation or obstruction may be present on plain films. Demonstration of small bowel strictures by contrast studies (Fig. 81-4) or by computed tomography helps to distinguish CUJ from uncomplicated GSE: small bowel dilation is typical of the latter. [167] , [168] and [169] Bowel thickening, mesenteric lymphadenopathy, and splenic atrophy are consistent with CUJ. However, lymphadenopathy in patients with GSE is not diagnostic of lymphoma.

FIGURE 81-4. Chronic ulcerative nongranulomatous jejunoileitis (CUJ). Thirty-minute small bowel follow-through shows prominent intestinal folds, thickening of the bowel wall, diffuse lumenal narrowing, and mucosal ulceration (*arrows*). Dilation, which is typical of gluten-sensitive enteropathy, is not a feature of CUJ. (From ref. [169] .)

Small bowel biopsies are essential for establishing the presence of CUJ. Although the yield from blind biopsies is poor, improvements in the ability to obtain endoscopic biopsy samples should markedly reduce the need for surgery to establish the diagnosis. [169] Biopsy samples obtained from ulcerated regions are the most desirable. As discussed earlier, it is essential that adequate tissue is obtained, sectioned extensively, and examined by an experienced pathologist before excluding the diagnosis of EATL. [139] A careful search for disseminated lymphoma at the time of presentation is essential. [138] It is also important to assess for the presence of B-cell surface markers because B-cell lymphomas and immunoproliferative small intestinal disease may be associated with villus atrophy. Polymerase chain reaction has been used to detect diagnostic T-cell receptor gene rearrangements in biopsies. [160]

Therapy No specific therapy has been shown to modulate the course of CUJ. Most patients have unresponsive malabsorption, and life expectancy at diagnosis is less than 2 years. Surgical resection of severely affected bowel has been of benefit in some cases. [142] , [156] Although patients with CUJ are at increased risk of intestinal perforation, prednisone therapy should be considered in all symptomatic patients with no evidence of lymphoma. The dose should be reduced to the minimum necessary to maintain a symptomatic response with tolerable side effects. Although CUJ does not respond to removal of dietary gluten, it is possible that this measure may impede the development of lymphoma. [152] If a localized lymphomatous infiltrate is found, local excision is mandatory. Although some patients have been cured by resection alone, [170] most remain symptomatic after the surgical procedure. Mortality in these patients is very high, presumably because of progression of the lymphoma. The impact of adjuvant radiation and of chemotherapy has not been evaluated. Disseminated T-cell lymphoma should be treated with aggressive chemotherapy. No trials have been conducted, but the principles are probably similar to those used in the treatment of other lymphomas, using combinations such as cyclophosphamide, hydroxydoxorubicin, vincristine, and prednisone. [171]

DRUG-INDUCED SMALL BOWEL DISEASE

Many of the drugs used today mediate various mechanisms of injury, including the following: erosive damage by NSAIDs and KCl; ischemic damage by cardiac drugs, cocaine, ergotamine alkaloids, and oral contraceptives; hematomas from anticoagulants; motility disorders from opiates; malabsorption; and inhibition of epithelial cell turnover by chemotherapeutic agents. These adverse reactions may be reversible if the drug is discontinued, but some medications produce permanent effects. This section focuses on drugs that have a profound or permanent effect on the small intestine, except for those causing erosive damage, as discussed earlier. The pathological and radiologic features of drug-related lesions of the intestinal tract have been reviewed. [172], [173]

Drugs Causing Ischemia

Mechanisms by which drugs and toxins can produce occlusive or nonocclusive intestinal ischemia include induction of arterial vasoconstriction, systemic or splanchnic hypotension, and thrombosis of mesenteric vessels (see Chapter 131). Regardless of cause, the mortality rate from small bowel ischemia progressing to bowel infarction ranges from 70% to 90%. [174], [175] Delays in presentation and diagnosis and coexistent health problems account for this high mortality. Patients with drug-induced mesenteric ischemia usually present with severe, poorly localized abdominal pain, low-grade fever, and hematochezia. Signs of peritonitis or abdominal distention secondary to ileus may be present. Evidence of intestinal perforation, obstruction, or ileus can be obtained by radiographic studies. The demonstration of bowel wall edema and thumbprinting, pneumatosis intestinalis, or portal venous gas are consistent with mesenteric infarction (Fig. 81-5). [174] Computed tomography is a reliable diagnostic method. [176], [177] Radiographic studies may be normal in patients with nonocclusive ischemia, such as occurs with digitalis-induced mesenteric spasm and antihypertensive medications. [178], [179] Mesenteric angiography may be needed to document digitalis-induced ischemia. [180]

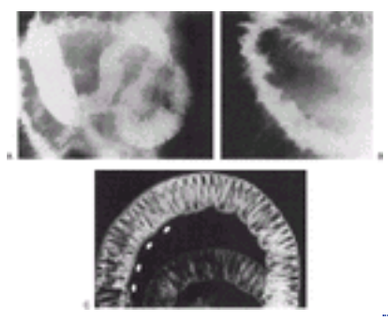

FIGURE 81-5. Acute spontaneous mesenteric venous thrombosis. (**A**) Involved jejunum shows separation of loops and thumbprinting. (**B**) Blurring of mucosal folds is seen in a distal zone of involvement. (**C**) Barium study of a surgical specimen shows thickening of mucosal wall and folds, thumbprinting, and pseudomembrane formation (*arrows*). (From Pringot J, Bodart P. Inflammatory diseases. In: Margulis AR, Burhenne HJ, eds. Alimentary tract radiology. St. Louis: CV Mosby, 1989:804.)

Besides measures specific to the offending agent, general supportive measures include administration of fluids and oxygen, correction of electrolyte and acid-base disorders, and antibiotic therapy. [174] Surgical resection of infarcted small bowel is necessary, although most patients are very high-risk surgical candidates.

Cardiac Drugs Intestinal ischemia or infarction may result from drugs that produce or potentiate arterial vasoconstriction or hypotension. [181] The incidence of these side effects is greatest when the splanchnic circulation is already compromised, such as in congestive heart failure. Antihypertensive agents and diuretics can induce intestinal ischemia secondary to hypotension and hypovolemia. Drugs with direct vasoconstrictive effects, such as norepinephrine, dopamine, and intraarterial or intravenous vasopressin, reduce intestinal blood flow and can cause mesenteric infarction. [182], [183] Digitalis glycosides can further reduce splanchnic flow in patients with congestive heart failure. Intestinal ischemia resulting from digoxin use may be reversed with nitroprusside and calcium channel blockers. [180], [184]

Cocaine Cocaine is a lipophilic drug that is readily absorbed by mucosal surfaces, including the intestinal mucosa. It has potent stimulatory effects on the central nervous system because of inhibition of presynaptic reuptake of norepinephrine, dopamine, and serotonin (5-HT) as well as increased synthesis of norepinephrine and dopamine. [185], [186] Excessive sympathetic stimulation produces peripheral vasoconstriction and tachycardia, which can cause hypertension and nonocclusive intestinal ischemia. [187] Rebound vasodilation after intense vasoconstriction can further compromise intestinal perfusion. Gastrointestinal manifestations of cocaine use include appetite suppression, nausea, vomiting and hyperperistalsis, perforated gastroduodenal ulcers (most common in crack users), and hepatotoxicity. [188] Intestinal ischemia is a serious but rare complication of cocaine and crack abuse. [189], [190], [191], [192], [193] and [194] Life-threatening gastrointestinal problems are more likely to be encountered by “body packers.” Such persons transport illicit drugs by ingesting drug-containing packets or condoms or by inserting them in the rectum. [187], [195], [196] Complications include mechanical intestinal obstruction and rupture of a packet, which can release a lethal quantity of cocaine. [196], [197] Signs of drug toxicity, mechanical intestinal obstruction, or marked leukocytosis, which may indicate the presence of gangrenous bowel, are indications for laparotomy. [182] Asymptomatic patients should be treated with activated charcoal to bind any leached drug and with mild cathartics to stimulate gastrointestinal transit. [182], [195] Abdominal radiographs may reveal evidence of intestinal obstruction or ileus as well the pathognomonic “double-condom sign.” [198]

Ergotamines Ergot alkaloids such as ergotamine, dihydroergotamine, methylergonovine, and methysergide are used in the symptomatic treatment or prevention of migraine headaches. [199] Common intestinal complications of ergots include abdominal pain, nausea, and vomiting. Symptomatic vasospasm affecting the extremities is a common complication of ergot toxicity, whereas severe splanchnic vasoconstriction is rare. [200], [201] There are several reports of bowel infarction after administration of large doses of ergots. [200], [201] Ergots have high affinity for a-adrenergic and 5-HT receptor subtypes. [202] Ergot-induced vasospasm is thought to occur as a result of peripheral adrenergic stimulation. [200], [202] The diagnosis of ergot-induced intestinal ischemia is based on the presence of abdominal symptoms in a patient taking ergotamines. Arteriographic demonstration of mesenteric vascular spasm and collateralization supports the diagnosis. [201] Discontinuation of ergots and supportive care with fluids are usually sufficient therapy. Intravenous sodium nitroprusside may be beneficial in severe cases. [201] Emergency laparotomy is essential if there is evidence of gangrenous bowel.

Oral Contraceptives Oral contraceptives have been reported to cause small intestine infarction and, more commonly segmental, colitis. [203] The mechanisms are not known, although mesenteric arterial occlusion [204], [205], [206] and [207] or mesenteric venous thrombosis [208], [209], [210] and [211] is often present. Progestins cause arterial occlusion, whereas estrogens can produce both arterial and venous occlusion. [212], [213] Mechanisms that have been implicated include hypercoagulability, reduced mesenteric venous blood flow, and endothelial proliferation. [206], [214] The risk is increased by ancillary factors such as collagen-vascular disease, hypercoagulable states, smoking, and hypertension. [215] The duration of contraceptive use is not an independent risk factor and does not correlate with the severity of symptoms. [206] The diagnosis should be considered in contraceptive users presenting with poorly localized colicky abdominal pain, bloody diarrhea, or hematochezia. Fever is often present, and presenting symptoms also may include nausea and vomiting. [207] Although abdominal pain may be present for several weeks or months, pain serious enough to warrant hospitalization is usually of recent onset. Results of laboratory studies, including platelet count, prothrombin time, and partial thromboplastin time, are usually normal. [207] The diagnosis depends on the demonstration of intestinal ulceration or ischemia in contraceptive users. Abdominal radiographs may demonstrate bowel wall thickening or ileus. Ulceration, which may be detected with contrast studies, is usually continuous in involved segments. [207] Oral contraceptives should be discontinued and supportive therapy initiated. Although mesenteric ischemia may be reversible, the mortality rate in severe cases is almost 50%. [207] The risk of thromboembolic complications decreases to normal levels within 1 month of discontinuation of oral contraceptives. [214]

Anticoagulants Gastrointestinal hemorrhage is a common complication of anticoagulant therapy with heparin or warfarin. Antiplatelet therapy with ticlopidine has been associated with chronic severe diarrhea that is related to altered motility but not to hemorrhagic complications. [226], [227] The incidence of bleeding depends on the intensity of therapy, the adequacy of monitoring and dosage adjustment, the route of administration, concurrent drug therapy, and the gender, age, and underlying condition of the patient. The incidence of gastrointestinal bleeding requiring transfusion for medical inpatients without predisposing illness in the Boston Collaborative Drug Surveillance Program was 1.2% and 0.2% for heparin and warfarin, respectively. [216] Minor gastrointestinal bleeding occurred in 8.3% and 3.3%, respectively. Gastrointestinal bleeding should be thoroughly investigated, even if it is a minor symptom, and regardless of the prothrombin time. [217], [218], [219], [220] and [221] Most episodes of gastrointestinal bleeding in patients who are taking anticoagulants are attributable to a demonstrable gastric or intestinal lesion. [220], [221] The most common hemorrhagic small intestine complication is spontaneous or trauma-induced intramural hematoma, [222] usually in the jejunum in the absence of abdominal trauma. The relative fixation of the descending duodenum is responsible for a higher susceptibility to intramural hematoma in response to blunt abdominal trauma. [222] Hematomas may cause colicky abdominal pain and symptoms secondary to intestinal obstruction. Biliary symptoms or pancreatitis may result from obstruction of the biliary or pancreatic ducts. Minor intestinal bleeding is detected in 25%, and major bleeding occurs in 3.5%. [223] Patients may present with an acute abdomen that can be differentiated from that owing to other causes by the more gradual onset of symptoms and the acute decrease in hematocrit. [224] Abdominal

tenderness, low-grade fever, and a palpable abdominal mass are common physical findings. Evidence of proximal small bowel obstruction may be present on plain abdominal radiographs. The most useful diagnostic test is an upper gastrointestinal series demonstrating the classical coiled-spring, picket-fence, or stack-of-coins sign (Fig. 81-6), caused by extravasation of blood into the valvulae conniventes, narrowing the lumen with spikelike projections of barium outlining the normal caliber of the lumen. [217, 222] Serial ultrasound examinations can be useful for monitoring the behavior of the hematoma. [225]

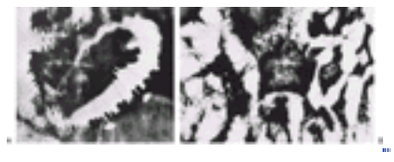

FIGURE 81-6. Small intestine ischemia. (**A**) Segmental ischemia in a picket-fence pattern of regular thickening of small bowel folds (*arrows*). (**B**) Complete resolution of the ischemic process is achieved after conservative therapy. (From Eisenberg R. Regular thickening of small bowel folds. In: Eisenberg R, ed. Gastrointestinal radiology: a pattern approach, 3rd ed. Philadelphia: Lippincott–Raven, 1996:471.)

Conservative medical management is usually sufficient because most hematomas undergo spontaneous reabsorption, with resolution of symptoms within 3 weeks. Reversal of anticoagulation with fresh-frozen plasma and vitamin K, intravenous hydration, and continuous nasogastric suction are sufficient for management of most patients. Surgical intervention may be required if there is complete intestinal obstruction or if conservative medical management fails to resolve intestinal bleeding, obstructive symptoms, or fever. [219, 223]

Drugs Causing Motility Disorders

Drug-induced intestinal pseudoobstruction can occur as follows: with use of anticholinergic drugs; with use of drugs with anticholinergic effects such as phenothiazines and tricyclic antidepressants, opioids, verapamil, and clonidine; and, occasionally, with initiation of cyclosporine therapy. Neurotoxicity induced by such drugs as vincristine can also produce intestinal pseudoobstruction, [228] which frequently appears within 3 days of the initiation of therapy and resolves within 2 weeks after the cessation of therapy. Direct toxicity to the enteric nervous system is probable because evidence of peripheral nerve dysfunction is not always present in patients with this complication.

Persistent use of narcotic analgesics can produce the *narcotic bowel syndrome,* characterized by abdominal pain, intermittent vomiting, weight loss, and other symptoms suggestive of intermittent pseudoobstruction. [229] Poorly localized chronic abdominal pain that is colicky, with acute exacerbations, is typically the major symptom. The pain is initially responsive to narcotic analgesics, but, with continued narcotic use, progressively larger doses are required for pain relief. Although the differential diagnosis includes biliary colic, pancreatitis, peptic ulcer disease, and renal calculi, the history, physical examination, and laboratory tests are usually not consistent with these entities. Abdominal radiographs typically are consistent with ileus and, on rare occasions, with mechanical small bowel obstruction. [229] The diagnosis of narcotic bowel syndrome is readily made with an intake history of more than 2 weeks of moderate to heavy doses of narcotics and a workup that excludes more serious disorders. [229]

Narcotic withdrawal is essential for treatment of narcotic bowel syndrome. On withdrawal of these drugs, the patient may experience vomiting, diarrhea, and cramping resulting from increased intestinal motility. [229] These symptoms can be markedly reduced or abolished by the a_2-adrenergic receptor agonist, clonidine. [230, 231] Opiates are discontinued, and clonidine is gradually increased from 0.1 mg twice daily to 0.1 mg four times daily. [230] After 1 week of therapy, the dose is tapered over 3 days. Narcotic bowel syndrome and withdrawal symptoms can be successfully treated in 90% to 95% of patients with this regimen. Metoclopramide therapy has been of benefit in patients with cancer. [232]

Drugs Causing Malabsorption

Drug-induced malabsorption of nutrients, electrolytes, and concurrently used medications occurs by several different mechanisms. [178, 207, 233] These include the following: intralumenal interactions that interfere with the solubilization, digestion, or transport of nutrients; increased rate of intestinal transit; mucosal injury; direct inhibition of absorptive processes; and inhibition of gastric, biliary, or pancreatic secretions. Drugs that impede nutrient assimilation by direct interaction include the following: tetracycline, which chelates calcium ions; cholestyramine, which binds to iron and vitamin B_{12}; mineral oil, which reduces the solubilization of ß-carotene and fat-soluble vitamins; and aluminum and magnesium hydroxide, which precipitate calcium and phosphate ions. Prokinetic agents and cathartics can impair fat absorption by increasing intestinal transit. [234] Mucosal injury resulting in diminished nutrient absorption has been reported with drugs such as colchicine, neomycin, and methotrexate. Colchicine inhibits mitosis and brush border disaccharidases and impairs absorption of fat, vitamin B_{12}, ß-carotene, D-xylose, lactose, bile salts, and steroids. [235] Neomycin is thought to cause brush border damage by inhibiting enterocyte protein synthesis, [236, 237] as well as to impair micellar solubilization of bile salts, cholesterol, fatty acids, and fat-soluble vitamins by directly binding to bile salts. Azotorrhea and decreased D-xylose absorption also can occur with neomycin. The degree of malabsorption with neomycin is dose related and can be seen with as little as 3 g/d. [236] Discontinuation of the drug typically reverses the malabsorption and diarrhea.

Methotrexate decreases the height of intestinal microvilli and brush border membrane protein and lipid content. Other drugs that produce histological changes in the jejunal mucosa associated with fat malabsorption include methyldopa, [238] allopurinol, [239] and mefenamic acid. [240] The histological features of mefenamic acid enteropathy closely resemble those associated with celiac disease. Sodium aminosalicylate [241] and thiazide diuretics, which impair ileal vitamin B_{12} and sodium transport, respectively, are examples of drugs that directly inhibit nutrient transport.

Clofazimine, when used in high doses for the treatment of leprosy, can cause a distinctive enteropathy characterized by red brown birefringent crystals in the small bowel mucosa and submucosa as well as in the mesenteric lymph nodes. [242] Clofazimine enteropathy is associated with nonspecific clinical and radiologic signs including diarrhea abdominal pain, anorexia, vomiting, diarrhea, and weight loss but not clinically significant malabsorption. [243]

The clinical significance of drug-induced malabsorption is influenced by such variables as baseline nutritional status and dietary intake, underlying disorders that interfere with intestinal function, and pharmacological considerations such as drug dosage and schedule and duration of therapy. Awareness of the nutritional consequences of drug therapy facilitates the prevention and early detection of these disorders. For some drugs, prophylactic nutritional supplementation is reasonable. For example, patients treated on a long-term basis with sulfasalazine, phenytoin, or colestipol should receive folic acid, and patients receiving the latter two drugs should also receive vitamin D supplementation.

Chemotherapeutic Agents

Most chemotherapeutic agents produce cytotoxic effects in normal cells because they have low therapeutic indices. Cells that have a high turnover rate, such as those of the small intestine crypt epithelium, are particularly vulnerable to drugs that inhibit cell proliferation. The magnitude of gastrointestinal toxicity is highly variable among patients and is affected by dose, duration of treatment, specific agents used, concurrent radiotherapy, and nutritional status of the patient. Nausea and vomiting are common acute side effects of antineoplastic drugs and are mediated in part by the chemoreceptor trigger zone in the brain. These symptoms predominate in the early stages of therapy. After a single course of therapy, intestinal mucosal damage is usually evident within the first 3 days. Regeneration and repair occur rapidly after cessation of therapy, with grossly normal mucosa present by 14 days, although inflammatory changes may be present for several weeks. [244]

Erosive enteritis, with or without stomatitis, presents with abdominal pain, bleeding, vomiting, ileus, or diarrhea. It is most common with methotrexate therapy but is also seen with 5-fluorouracil, actinomycin D, doxorubicin, cytarabine (cytosine arabinoside; Ara-C), bleomycin, and vincristine. These chemotherapeutic agents are also associated with other complications. Methotrexate is also associated with malabsorption (discussed in the previous section). The erosive enteritis associated with cytarabine when it is used in combination regimens occurs within 8 to 11 days from the start of therapy and is associated with hypokalemia, hypocalcemia, PLGE, and a syndrome characterized by telangiectasis and intramural hematomas. [245] Recombinant interleukin-2, which is used for treatment of advanced renal cell carcinoma, has been associated with intestinal ischemia, necrosis, perforation, and diffuse bowel ulceration requiring surgery. [246] These complications have occurred with interleukin-2 alone or in combination with interferon-? or lymphokine-activated killer cells. Cyclosporine-induced vasoconstriction can contribute to the development of nonocclusive mesenteric ischemia in renal transplant recipients. [247] Vincristine can induce acute intestinal pseudoobstruction (as already discussed). Preconditioning for bone marrow transplantation with cyclophosphamide and total-body irradiation produces diffuse intestinal injury. Symptoms, including crampy abdominal pain, watery diarrhea, and anorexia, are common within the first 3 weeks after transplantation. Symptoms that persist beyond 3 weeks should be investigated to rule out enteric infection or other intestinal disorders. Neutropenic enterocolitis or typhlitis is most frequently encountered after chemotherapy for leukemia or lymphoma. Diffuse patchy mucosal necrosis involving the ileocecal region is associated with nonspecific symptoms and fever, nausea, vomiting, right lower quadrant

pain, and hematochezia. Colon complications resulting from these drugs are discussed in Chapter 85.

NECROTIZING ENTEROCOLITIS

NEC is also referred to as pigbel, enteritis necroticans, darmbrand, nonspecific jejunitis, epidemic regional jejunitis, Pasini regional jejunitis, and necrotizing jejunitis. Although there were numerous European outbreaks during and after World War II, [248] acute jejunitis is largely a problem in nonindustrialized nations. [249], [250] and [251] Outbreaks are most frequent in communities in which protein deprivation and poor food hygiene are prevalent. [252] The disease was a major cause of illness and death among children in the highlands of Papua New Guinea until immunization against the ß-toxin of the causative organism, *Clostridium perfringens,* was begun in 1980. [253] Isolated cases continue to be reported from industrialized nations, although detection of the causative organism is not always reported. [254], [255], [256] and [257] A well-documented case occurred in Atlanta, Georgia in a 12-year-old boy with poorly controlled diabetes who had eaten pig intestines (chitterlings). [257]

C perfringens type C (initially identified as *C welchii* type F) is a heat-resistant bacterium that can often be isolated from the tissues, stool, and food of affected patients. [258], [259], [260] and [261] One sixth of healthy persons were shown to harbor a less pathogenic strain. A similar illness attributed to pathogenic strains of this organism has been described in certain animal species. [262], [263] Immunization against the ß-toxin resulted in a reduction in hospital admissions for acute jejunitis to less than one fifth of previous figures. [253] These observations have firmly established that this organism is a causative agent of acute jejunitis. The agent has also been isolated from children in Bangladesh with nonnecrotizing diarrheal illnesses. [264] Nevertheless, there are patients who have an identical syndrome without evidence of *C perfringens* type C infection or the ß-toxin. [265] A newer cytotoxin isolated from *Bacillus cereus* has been identified as another putative cause of acute jejunitis. [266] Additional factors associated with development of acute jejunitis include ingestion of foods containing trypsin inhibitors (e.g., sweet potatoes), reduced gastric acidity, and reduced gastrointestinal motility. [257]

Acute jejunitis occurs sporadically and in epidemics. Illness is characterized by bloody diarrhea, fever, and abdominal pain, with a high mortality rate (58%). [252] Nonocclusive small intestinal ischemia resulting in necrosis of varying severity, with areas of full-thickness necrosis, is usually found. Early recognition, treatment with antibiotics, and surgical resection are essential for successful treatment.

Neonatal NEC is a disease of focal or diffuse small intestine ulceration and necrosis that is seen almost exclusively in premature infants and low-birth-weight neonates who have been fed enterally. Similar pathophysiological mechanisms may be operative in adult necrotizing enteropathies. The putative causes, clinical presentation, differential diagnosis, pathology, and management of NEC are discussed in several excellent reviews. [267], [268], [269] and [270]

The cause of NEC is unknown, but several factors appear to be implicated, including prematurity, intestinal ischemia, infectious agents and bacterial colonization, and the initiation of enteral nutrition. [271], [272] However, a randomized trial and subsequent reviews [273], [274] are at odds with earlier case control studies suggesting that stressed infants are at greater risk of developing NEC if enteral feedings are advanced rapidly or if excessive volumes are used. [275], [276], [277] and [278] The pathogenesis of NEC is clearly multifactorial, and susceptibility of individual patients to NEC is probably also dependent on unidentified host factors. [271], [272] Successful delineation of these factors and other pathogenic mechanisms should improve the ability to prevent and treat NEC and should provide insight into the mechanisms of similar disorders seen in adults.

PROTEIN-LOSING GASTROENTEROPATHY

Definition

PLGE is a syndrome characterized by enteric loss of plasma proteins in abnormal amounts. Many intestinal and extraintestinal diseases may be associated with PLGE, and treatment is aimed almost exclusively at the underlying disorder. The recognition of PLGE mandates a careful search for an associated illness.

The defining characteristic of PLGE is hypoproteinemia resulting from gastric or intestinal losses. Studies of iodine 131–labeled albumin secretion identified the stomach as the site of excessive protein loss in patients with hypertrophic gastritis and atrophic gastritis and the intestine as a site of loss in patients with inflammatory bowel disease. [280], [281] and [282]

Under physiological conditions, sloughed enterocytes and pancreatic and biliary secretions account for almost all the endogenous protein that is found in the intestine. Most of these proteins are digested, and the constituent amino acids are reabsorbed. [283], [284] Gastrointestinal loss of serum proteins, as indicated by measuring serum albumin kinetics, accounts for less than 10% of daily protein catabolism. [285], [286], [287], [288] and [289] This may increase four- to fivefold in patients with PLGE. Hypoproteinemia results if the capacity to increase protein synthesis is not sufficient to compensate for increased intestinal protein losses. For example, albumin synthesis is increased no greater than twofold in patients with PLGE. [290]

Etiology

Most gastrointestinal disorders associated with PLGE cause exudative protein losses secondary to ulcerative or nonulcerative enteropathies or secondary to lymphatic obstruction or increased lymphatic hydrostatic pressure (Table 81-3). Enteropathies resulting in PLGE include ulcerating gastrointestinal carcinomas and inflammatory disorders such as Crohn's disease and ulcerative colitis. Increased enteric protein loss and increased catabolism are the primary contributors to the hypoalbuminemia and reduced protein pools that are seen in these disorders. In the case of inflammatory bowel disease, the magnitude of enteric protein loss is most significant, and it is directly related to the extent and activity of mucosal inflammation. [291], [292]

Increased Interstitial Pressure
Congenital intestinal lymphangiectasia, mesenteric lymphatic obstruction, tuberculosis, sarcoidosis, lymphoma, retroperitoneal fibrosis, increased central venous pressure, constrictive pericarditis, congestive heart failure
Ulcerative Diseases
Erosive gastritis or enteritis, neoplasia-carcinoma or lymphoma, Crohn's disease, pseudomembranous enterocolitis, acute graft-versus-host disease
Nonulcerative Diseases
Giant hypertrophic gastropathy (Ménétrier disease), viral enteritides, bacterial overgrowth, parasitic diseases (malaria, giardiasis, schistosomiasis, helminth infections), Whipple disease (see also lymphatic obstruction), allergic enteritis, eosinophilic gastroenteritis, gluten-sensitive enteropathy, tropical sprue, systemic lupus erythematosus

TABLE 81-3 Causes of Protein-Losing Enteropathy

Nonulcerative diseases such as Ménétrier disease, [280] atrophic gastritis, tropical sprue, celiac disease, allergic gastroenteritis, eosinophilic gastroenteritis, collagenous colitis, and polyposis syndromes including Cronkhite-Canada syndrome, juvenile polyposis, and Peutz-Jeghers syndrome have also been associated with PLGE. [280], [293], [294], [295], [296], [297], [298], [299], [300] and [301] In addition to neurological symptoms, defects in glycoprotein biosynthesis (the carbohydrate-deficient glycoprotein syndromes) can also present with severe PLGE. [302]

Acute and chronic intestinal infections can result in PLGE, including acute staphylococcal, *Salmonella,* [283] and *Shigella* [303] infections. The incidence of PLGE is species and strain dependent; for example, *Shigella dysenteriae* type 1 infections cause greater loss of enteric protein than do other strains. [303] Both pseudomembranous colitis and asymptomatic colonization by *C difficile* can result in enteric loss of protein. [304], [305] In a study of elderly nursing home patients, PLGE was documented in all 12 patients with pseudomembranous colitis, in 6 of 14 with *C difficile* diarrhea, and in 6 of 12 colonized with *C difficile* without toxin or diarrhea. [305] Parasitic diseases with documented PLGE include *Strongyloides stercoralis* infection, [306] schistosomiasis, [307], [308] and giardiasis. [309], [310] and [311] Acute viral illnesses including cytomegalovirus, measles, and varicella also have been linked to PLGE. [312], [313], [314], [315] and [316] Small bowel bacterial overgrowth can also be associated with PLGE. [317]

PLGE has been well documented in patients with SLE. [318], [319], [320], [321], [322] and [323] It occurs most commonly in young women, and it may be the initial clinical manifestation of SLE. Fifty percent of these patients present with steroid-responsive diarrhea without steatorrhea. [324] Mixed connective tissue disease, [325], [326] Henoch-Schönlein purpura, [327] and other collagen-vascular diseases also can be complicated by PLGE. [328] Another cause of PLGE is secondary amyloidosis complicated by small intestinal tract ulcers, resulting from arteriole and lymphatic infiltration with amyloid deposits. [329],

[330] PLGE also has been seen in association with mesenteric vascular changes resulting from neurofibromatosis [331] and idiopathic mesenteric thrombophlebitis. [332]

Diseases that produce disruption of intestinal lymphatic vessels or obstruction of lymph flow are an additional cause of PLGE. Tortuous, dilated mucosal and submucosal lymphatic vessels are the hallmark of primary intestinal lymphangiectasia (see Chapter 72). Most patients with this disease present by 30 years of age with edema, diarrhea, hypoproteinemia, and lymphocytopenia resulting from epithelial leakage and lymphatic rupture. [333] A similar lesion has been implicated as the mechanism of enteric protein loss in some patients with systemic sclerosis and SLE. [334], [335]

Inflammatory granulomatous diseases such as sarcoidosis and mesenteric tuberculosis cause PLGE. [341], [342] By producing retroperitoneal lymph node enlargement or fibrosis, Whipple disease, [336], [337] lymphoma, [338] endometriosis of the small intestine, [339] and Crohn's disease [291], [340] impair lymphatic egress by causing mechanical obstruction or increased hydrodynamic pressure. Additional retroperitoneal processes, including pancreatitis, pancreatic cancer, and other retroperitoneal tumors, also can present with PLGE. Right-sided congestive heart failure, constrictive pericarditis, superior vena caval obstruction, and other causes of elevated central venous pressure [343], [344] commonly produce PLGE.

Clinical Features

The major clinical manifestation of PLGE is dependent edema resulting from decreased plasma oncotic pressure. Although all plasma proteins are usually lost, selective loss of low-molecular-weight proteins may occur without lymphatic obstruction. Proteins such as albumin, immunoglobulins (IgM, IgG, and IgA but not IgE), fibrinogen, ceruloplasmin, and a_1-antitrypsin that have long circulating half-lives and a limited capacity for increased synthesis are most likely to exhibit depressed levels. Decreased levels of proteins other than albumin are rarely symptomatic, although in the setting of lymphatic obstruction, symptomatic lymphopenia and steatorrhea may be present.

Diagnosis

Edema and hypoproteinemia resulting from enteric protein loss are presenting manifestations of disease, but they are often minor aspects of the underlying disease. The diagnosis should always be considered in hypoalbuminemic patients if other causes such as malnutrition or protein loss from other sites are not apparent. If it is necessary to document intestinal protein losses, the preferred method is to quantitate fecal a_1-antitrypsin concentration or clearance. Alternative methods employing radiolabeled substrates for quantitating enteric protein loss remain useful research tools, but they are seldom used for clinical purposes. Clinical studies indicate that PLGE can be diagnosed and the sites of albumin loss can occasionally be determined scintigraphically using ^{99m}Tc-labeled albumin or dextran. [345], [346], [347], [348], [349] and [350]

a_1-Antitrypsin is particularly valuable because it constitutes approximately 4% of serum proteins, has a molecular weight similar to that of albumin, is resistant to proteolysis, is not actively absorbed or secreted, is normally present in low quantities in stool, and is easy to assay. The test cannot be used for analyzing gastric protein losses or losses in patients with Zollinger-Ellison syndrome because a_1-antitrypsin is degraded at a pH below 3. [351] The a_1-antitrypsin concentration in stool and the plasma clearance of a_1-antitrypsin were compared in healthy persons and in consecutive patients with chronic diarrhea, malabsorption, or unexplained hypoalbuminemia. [352] The plasma clearance is the product of daily stool volume and stool a_1-antitrypsin concentration divided by the serum a_1-antitrypsin concentration. [353] In contrast to earlier studies, [354], [355] and [356] there is ample evidence that analysis of fecal a_1-antitrypsin concentration is not a reliable substitute for measurement of a_1-antitrypsin clearance. [352] There is a highly significant correlation between a_1-antitrypsin clearance and serum albumin concentration and fecal loss of radiolabeled albumin. When the a_1-antitrypsin clearance was elevated more than threefold, serum albumin levels were less than 3.0 g/dL. a_1-Antitrypsin clearance levels may be increased in otherwise healthy persons with diarrhea secondary to ingestion of lactulose, sorbitol, sodium sulfate, or phenolphthalein. [352] Clearance levels also may be overestimated if hematochezia or meconium is present.

Therapy

There is no specific therapy for PLGE. Optimal therapy of the primary illness is the only effective remedy. In the case of Ménétrier disease, octreotide and eradication of *Helicobacter pylori* may reduce enteral protein losses. [357], [358] Prophylactic measures to avoid complications resulting from peripheral edema should be instituted. In the setting of lymphangiectasia, reduction of lymphatic transport may reduce enteric protein loss. Low-fat diets supplemented with medium-chain triglycerides may be beneficial in this setting. [359], [360] Surgical drainage of dilated lymphatic channels (lymphovenous anastomosis) is occasionally helpful. [340], [361]

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CHAPTER 82

Steven M. Cohn and Elisa H. Birnbaum

COLON: ANATOMY AND STRUCTURAL ANOMALIES

ANATOMY OF THE COLON

The colon is a tubular organ that extends from the ileocecal valve to the proximal rectum. It is approximately 3 to 5 feet in length. Its principal functions are absorption of water and electrolytes as well as storage of intralumenal contents to permit controlled elimination of the feces. The terminal ileum enters the cecum on its posteromedial border at the ileocecal valve. The cecum is a large, blind pouch, approximately 7.5 to 8.5 cm in diameter, which projects from the antimesenteric side of the ascending colon. The colon progressively diminishes in size toward the sigmoid colon, which is approximately 2.5 cm in diameter and is the narrowest portion of the colon. This size discrepancy accounts for the fact that cecal tumors often grow to be large and bulky before the onset of symptoms but sigmoid tumors are symptomatic at smaller sizes. In addition, tension on the bowel wall is directly proportional to the diameter of the bowel. This is explained by the law of LaPlace: T = PR, where *T* is tension in the wall of the bowel, *P* is internal pressure, and *R* is the radius of the bowel. Because the cecum has the largest diameter, it is usually the first part of the bowel to rupture as a result of distal obstruction.

The omentum is attached to the transverse colon on its anterior superior edge. The ascending colon, descending colon, rectum, and posterior surface of the hepatic and splenic flexures are fixed retroperitoneal structures. The cecum, transverse colon, and sigmoid colon are intraperitoneal and are prone to volvulus because of their location and relative lack of fixation.

The longitudinal muscle is an incomplete layer and is seen as three bands of muscle, called teniae coli, located 120 degrees apart around the circumference of the colon. The teniae converge proximally at the appendix as distinct bands and disappear at the level of the sacral promontory. Haustra coli are sacculations between the teniae and are separated by crescent-shaped folds called plicae semilunares. Appendices epiploicae are fatty appendages covered by peritoneum and have no anatomic or pathological significance.

P>The rectal wall consists of mucosal, submucosal, inner circular, and outer longitudinal muscular layers (Fig. 82-1). There is no serosal layer in the rectum. The rectum is approximately 12 to 15 cm in length and extends from the sigmoid colon to the anal canal, following the curve of the sacrum. The proximal rectum begins at the sacral promontory (third sacral vertebra), at which point the teniae fan out to form a complete layer of longitudinal muscle.

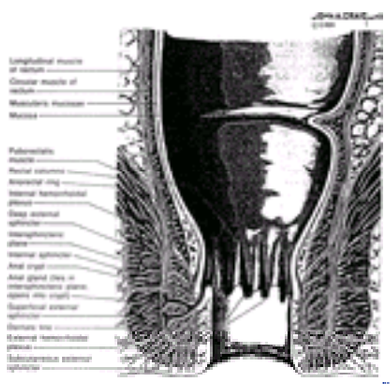

FIGURE 82-1. Anorectal anatomy. Copyright 1985. Novartis. Reprinted with permission from Clinical Symposia 37/6, illustrated by John A. Craig, M.D. All rights reserved.

The anal canal is approximately 4 cm long and appears as a collapsed anteroposterior slit in the normal patient (see Fig. 82-1). The anal verge is the junction between anal and perianal skin. Anal epithelium (anoderm) lacks hair follicles, sebaceous glands, and sweat glands. The dentate line is a true mucocutaneous junction located 1 to 1.5 cm above the anal verge. A 6- to 12-mm transitional zone exists above the dentate line, in which the squamous epithelium of the anoderm becomes cuboidal and then columnar epithelium. The columns of Morgagni are 8 to 14 mucosal folds located just above the dentate line that are surrounded by anal crypts. Small, rudimentary glands open into some of the crypts. These glands go through the internal sphincter into the intersphincteric groove but do not penetrate the external sphincter. The anorectal ring is 1 to 1.5 cm above the dentate line and is the palpable upper border of the anal sphincter complex. The anatomic anal canal extends from the anal verge to the dentate line. The surgical anal canal extends from the anal verge to the anorectal ring.

Arterial Supply

The superior mesenteric artery arises from the ventral surface of the aorta below the celiac axis, passes behind the pancreas, and crosses in front of the third portion of the duodenum (Fig. 82-2). The cecum, ascending colon, and transverse colon are supplied through its ileocolic and middle colic branches. Cadaver studies have shown that the right colic artery frequently arises from the ileocolic artery in the majority of patients and arises from the superior mesenteric artery in less than 15% of patients. [1]The inferior mesenteric artery arises from the infrarenal aorta and supplies the descending colon, sigmoid colon, and upper rectum through its left colic, sigmoidal, and superior rectal branches.

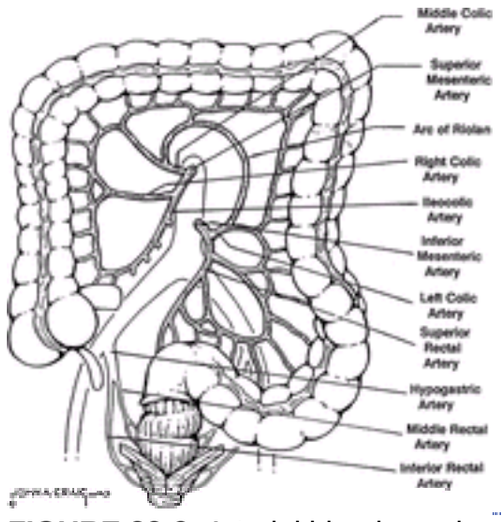

FIGURE 82-2. Arterial blood supply of colon and rectum. (From Kodner IJ, Fry RD, Fleshman JW, Birnbaum EH. Colon, rectum and anus. In: Schwartz SI, ed. Principles of surgery, 6th ed. New York: McGraw-Hill, 1993.)

Collaterals exist between the superior and inferior mesenteric arteries in the region of the splenic flexure. [2] The arcades of the ileocolic, right, middle, and left colic arteries are peripherally connected by the marginal artery of Drummond, which runs along the mesenteric border of the colon and provides the vasa recta to the colon. The arc of Riolan is a tortuous, inconstant vessel that exists between the left colic branch of the inferior mesenteric artery and the middle colic branch of the superior mesenteric artery. It is frequently referred to as the meandering mesenteric artery and is best visualized if there is an occlusion of either the inferior or superior mesenteric artery.

The terminal branch of the inferior mesenteric artery becomes the superior rectal artery, which descends in the sigmoid mesocolon and bifurcates at the level of the third sacral vertebra (Fig. 82-3A). Branches of the superior rectal artery supply the upper and middle rectum. The middle rectal arteries arise from the internal iliac arteries, and the inferior rectal arteries are branches of the internal pudendal arteries. These arteries supply the lower two thirds of the rectum. [3] The middle sacral artery arises just before the aortic bifurcation and provides very little blood supply to the rectum. Anastomosis exists between the middle and superior rectal arteries; there are no anastomoses with the inferior rectal arteries. Preservation of the middle rectal arteries is necessary to maintain viability of the rectal stump after high ligation of the inferior mesenteric artery. [3]

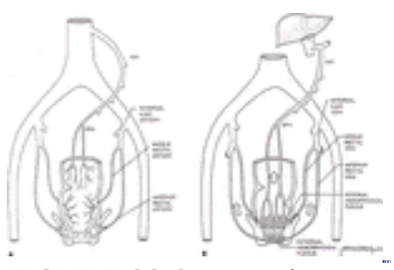

FIGURE 82-3. Vascular supply of the anus and rectum. **A**: Arterial supply. **B**: Venous drainage. *IMA*, inferior mesenteric artery; *IMV*, inferior mesenteric vein; *SRA*, superior rectal artery; *SRV*, superior rectal vein. (From Kodner IJ, Fleshman JW, Fry RD. Anal and rectal cancer principles of management. In: Schwartz SI, Ellis H, eds. Maingot's abdominal operations, 9th ed. Norwalk, CT: Appleton & Lange, 1989.)

Venous Drainage

The veins draining the colon follow the same course as the corresponding arteries except for the inferior mesenteric vein. The inferior mesenteric vein drains the descending colon, sigmoid colon, and proximal rectum and runs in a retroperitoneal location to the left of the ligament of Treitz, at which point it enters the splenic vein. The superior mesenteric vein drains the cecum, ascending colon, and transverse colon and joins the splenic vein to form the portal vein.

The venous drainage of the rectum enters the portal or systemic (caval) system. The upper and middle rectum are drained by the superior rectal vein, which enters the portal system through the inferior mesenteric vein (Fig. 82-3B). The lower rectum and upper anal canal are drained by the middle rectal vein, which empties into the internal iliac veins and then into the caval system. The inferior rectal veins drain the lower anal canal and empty into the pudendal veins, which empty into the caval system through the internal iliac veins. Rectal tumors can metastasize through venous channels into either the portal or systemic venous systems.

Three submucosal internal hemorrhoidal complexes are located above the dentate line. Hemorrhoidal tissue receives its blood supply from the superior, middle, and inferior rectal arteries. The left lateral, right posterolateral, and right anterolateral internal hemorrhoids drain into the superior rectal vein (Fig. 82-3B). The external hemorrhoids are located below the dentate line and drain into the pudendal veins. There is communication between the internal and external plexi, and mixed internal-external hemorrhoids result if these communications become engorged (see Chapter 92).

Lymphatic Drainage

Colonic mucosa has rich vascular plexi but no lymphatics. Lymphatic capillaries encircle the colon in the submucosal and muscularis mucosae layers. This segmental architecture limits longitudinal intramural extension of tumors, and circumferential extension results in annular lesions. Lymphatic vessels follow the blood supply of the colon.

Lymph nodes are located on the bowel wall (epicolic), along the inner margin of the bowel (paracolic), around the named mesenteric arteries (intermediate), and along the origin of the superior and inferior arteries (main) (Fig. 82-4). Lymph from the upper and middle rectum drains into the inferior mesenteric nodes. The lower rectal lymphatics follow the superior rectal artery and enter the inferior mesenteric nodes. Lymph from the lower rectum can also flow laterally along the middle and inferior rectal arteries, posteriorly along the middle sacral artery, or anteriorly through channels in the rectovesical or rectovaginal septum. [4] These channels drain to the iliac nodes and subsequently to periaortic lymph nodes.

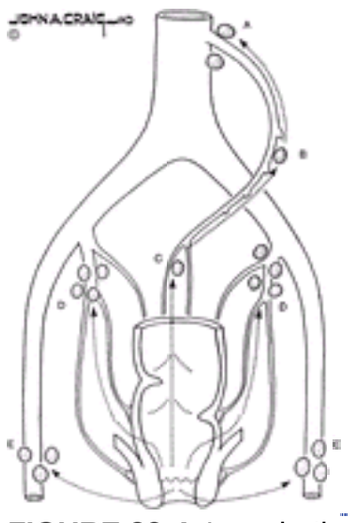

FIGURE 82-4. Lymphatic drainage of the rectum and anus. *A*, nodes at origin of inferior mesenteric artery; *B*, nodes at origin of sigmoid branches; *C*, sacral nodes; *D*, internal iliac nodes; *E*, inguinal nodes. (From Kodner IJ, Fleshman JW, Fry RD. Anal and rectal cancer: principles of management. In: Schwartz SI, Ellis H, eds. Maingot's abdominal operations, 9th ed. Norwalk, CT: Appleton & Lange, 1989.)

Lymphatics from the anal canal above the dentate line drain through the superior rectal lymphatics to the inferior mesenteric lymph nodes or laterally to the internal iliac lymph nodes. Below the dentate line, the lymphatics drain primarily to the inguinal nodes but they can drain to the inferior or superior rectal lymph nodes as well. [4]

Nerve Supply

The sympathetic and parasympathetic nerves to the colon follow the course of the blood vessels. Sympathetic nerves inhibit and parasympathetic nerves stimulate peristalsis. Sympathetic fibers to the right colon originate in the lower six thoracic segments and travel in the thoracic splanchnic nerves to the celiac and then to the superior mesenteric plexus. Sympathetic supply to the left colon and rectum originates in the first three lumbar segments. These nerves join the preaortic plexus and become the inferior mesenteric plexus below the bifurcation of the aorta. The parasympathetic supply to the right colon is presumed to come from the right vagus. The parasympathetic nerves to the left colon ascend from the pelvis, pass through the sigmoid mesocolon, and spread out toward the sigmoid and the descending colon.

The rectum is innervated by both sympathetic and parasympathetic nerves. [5] Sympathetic nerves from thoracolumbar segments unite below the inferior mesenteric artery, forming the inferior mesenteric plexus, and descend to the superior hypogastric plexus below the aortic bifurcation. The nerves bifurcate and descend in the pelvis as the hypogastric nerves, supplying sympathetic innervation to the lower rectum, bladder, and sexual organs, in both males and females. Injury to the inferior mesenteric plexus can result during ligation of the inferior mesenteric artery at its origin.

Parasympathetic fibers from the second, third, and fourth sacral roots (the nervi erigentes) unite with the hypogastric nerves anterior and lateral to the rectum, forming the pelvic plexus. [5] The periprostatic plexus arises from the pelvic plexus. Mixed fibers from these plexi innervate the rectum, internal anal sphincter, prostate, bladder, and penis. The pudendal nerve (originating from the second, third, and fourth sacral nerves) mediates sensory stimuli from the penis or clitoris through the dorsal nerve. Damage to the periprostatic plexus may occur during dissection of the rectum. Injury to the pelvic autonomic nerves may result in bladder dysfunction, impotence, or both.

Below the dentate line, afferent fibers of the inferior rectal and perineal branches of the pudendal nerve convey cutaneous sensations of temperature, pain, and touch. Numerous free nerve endings make this area very sensitive to sensations. [6] Above the dentate line, a poorly defined dull sensation, experienced if the mucosa is pinched or if internal hemorrhoids are ligated, is probably mediated by parasympathetic fibers.

HISTOLOGY

The colonic wall consists of mucosa, submucosa, inner circular muscle, outer longitudinal muscle, and serosa (Fig. 82-5). The colonic mucosa is similar in organization to the small intestine mucosa except that it lacks villi. The mucosa is lined by a simple columnar epithelium that forms straight tubular crypts that are about 0.5 mm in length (Fig. 82-6). The lamina propria extends from the simple columnar epithelium to the muscularis mucosae and contains many cells involved in immunologic functions of the gut (see Chapter 7). Numerous immunoglobulin-secreting plasma cells, macrophages, and lymphocytes are present, in addition to abundant lymphoid nodules that often extend through the muscularis mucosae into the underlying submucosa (Fig. 82-7). Subepithelial fibroblasts surround each crypt and produce the collagen table as well as many of the extracellular matrix components that underlie the basal lamina of the epithelium (Fig. 82-8). These fibroblasts and the extracellular matrix that they secrete appear to have an important role in regulating cell proliferation and differentiation events within the overlying epithelium. [7], [8], [9], [10] and [11] The submucosa contains numerous small veins, arteries, and lymphatic channels surrounded by loose connective tissue. The inner circular muscle fibers form a continuous layer around the colon. The outer longitudinal smooth muscle fibers are condensed into three bands (teniae coli) equidistant around the circumference of the colon. Haustra are the bulging sacculations that form between adjacent teniae coli. The serosa is a mesothelial-derived cell layer that covers the peritoneal surface of the colonic wall. Therefore, regions of the ascending colon, the descending colon, and the rectum that lie outside the peritoneal cavity have no outer serosal layer (see section " Anatomy").

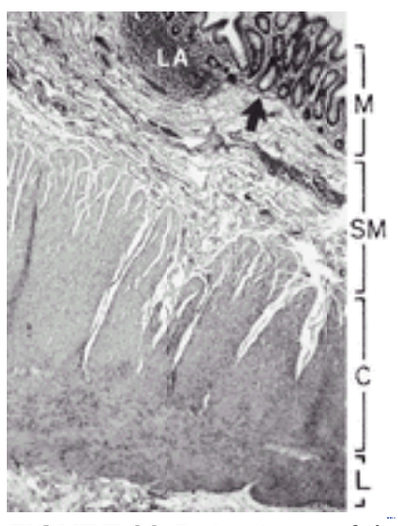

FIGURE 82-5. Anatomy of the colonic wall. The muscularis mucosae is indicated (*arrow*). (*C*, circular smooth muscle layer; *L*, longitudinal smooth muscle layer; *LA*, lymphoid aggregate; *M*, mucosa; *SM*, submucosa. Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×40.)

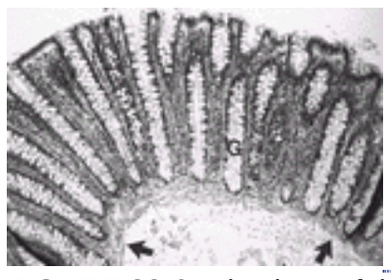

FIGURE 82-6. Histology of the colonic mucosa. Straight, simple colonic glands (*G*) extend from the muscularis mucosae (*arrows*) to the lumenal surface of the colon. Note the numerous small arteries, veins, and lymphatics coursing through the loose connective tissue stroma in the submucosa. (Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×100.)

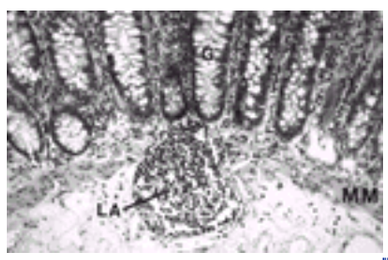

FIGURE 82-7. A lymphoid aggregate (*LA*) arises in the lamina propria and extends through the muscularis mucosae (*MM*) into the submucosa. (Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×200.)

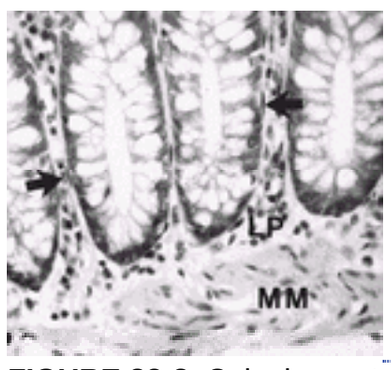

FIGURE 82-8. Colonic crypts. Note the subepithelial fibroblasts (*arrows*). Numerous plasma cells, macrophages, and lymphocytes are seen in the lamina propria (*LP*). (*MM*, muscularis mucosae. Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×400.)

The colonic epithelium undergoes continuous and rapid renewal; epithelial cells are sloughed from the flat lumenal surface of the colon and are replaced by replication of the descendants of epithelial stem cells located in the bottom one third of each crypt. There are three principal, differentiated epithelial cell types present in the adult colonic epithelium: absorptive colonocytes, goblet cells, and enteroendocrine cells. All these cell lineages appear to be derived from a common epithelial cell stem cell precursor. Undifferentiated cells, replicating cells, and enteroendocrine cells predominate near the base of each colonic gland (crypt). [12] Cells belonging to each of the principal cell lineages differentiate during a highly ordered migration away from the zone of proliferation. The average life span of goblet cells and absorptive cells, from their birth deep in the crypt to the point where they are sloughed into the lumen, is approximately 6 days. [13] Some enteroendocrine cell subtypes appear to have a much longer life span, suggesting that their migration up the crypt to the surface epithelium must be uncoupled from that of adjacent epithelial cells. [13] Animal studies using X-linked enzyme markers, chimeric mice, and transgenic mice have suggested that the epithelial cell population of each small intestine or colonic crypt is derived from a single stem cell or, at most, a very small number of stem cells. [14], [15], [16], [17] and [18] If normal homeostasis of the epithelial cell populations within the colonic mucosa is to be maintained, the rate of epithelial cell production within each crypt must be closely matched to the rate of cell loss from the surface epithelium. Although the mechanisms that regulate epithelial cell renewal have yet to be fully elucidated, a number of peptide growth factors are emerging as important potential regulators of the dynamic balance between cell proliferation and cell loss in the intestinal and colonic epithelium (see Chapter 23). [19]

One of the principal functions of absorptive columnar cells is the regulation of water and electrolyte content of the feces. The mature adult absorptive columnar cell in the colon absorbs sodium, chloride, and water; potassium and bicarbonate are secreted (see Chapter 14). [20] In contrast to the enterocytes of the small intestine, however, colonocytes are not able to absorb significant amounts of glucose or amino acids. Under normal conditions, lipid absorption is complete in the small intestine, but under pathological conditions in which lipid absorption in the small intestine is incomplete, the surface columnar cells of the colon have a limited capacity for uptake of fatty acids and subsequent packaging of them into chylomicrons. [21] As these cells differentiate during their migration up the crypt, they develop short microvilli and clear, apically oriented vesicles containing a fibrillar, glycoprotein-rich secretory product that may contribute to the glycocalyx. [21] These apical vesicles are lost, and microvilli elongate and increase in number as the maturing absorptive cells emerge onto the surface epithelium (Fig. 82-9). At this point, alkaline phosphatase activity appears on the brush border, and the basolateral membranes have acquired a considerable amount of Na^+,K^+-ATPase activity, reflecting their function in water and electrolyte transport.

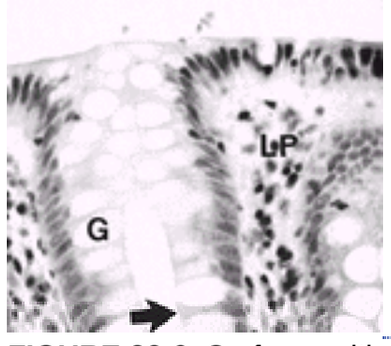

FIGURE 82-9. Surface epithelium of the colon. The mature absorptive columnar epithelial cells (*arrow*) lose their apical vesicles as they migrate into the upper regions of the colonic glands and emerge onto the lumenal surface. Goblet cells (*G*) elongate as they emerge onto the surface epithelium because of accelerated secretion of their mucus granules. (Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×400.)

Goblet cells are flask-shaped cells with large apical vesicles that store and secrete mucus. [22] Mucus secreted by the goblet cell forms a viscous gel that functions as a lubricant and also protects the surface epithelium against adherence of invading pathogens. Mucins are a family of large, sulfated glycoproteins that are synthesized in the rough endoplasmic reticulum (see Fig. 82-9). Glycosylation and sulfation of these proteins occur posttranslationally in the Golgi apparatus. The composition of mucus shows regional variability along the cephalocaudal axis of the gut. [22] The membrane-bound vesicles bud off the Golgi apparatus and are slowly transported to the apical cell surface, where they secrete their contents into the lumen by exocytosis. Mucus granules accumulate during the migration of goblet cells toward the

lumenal surface of the colon, distending the apical aspect of the cell and giving it its broad, flasklike appearance. As goblet cells emerge onto the surface epithelium, the secretion of mucus occurs more rapidly, depleting the apically stored granules and causing the goblet cells to elongate (see Fig. 82-9). Increased cholinergic stimulation can cause accelerated secretion of mucus from crypt-associated goblet cells, leading to deep invaginations of the apical surface. Certain components of mucus may become depleted in pathological conditions such as ulcerative colitis (see Chapter 83).

Originally thought to be of neuroectodermal origin, the enteroendocrine cells found in the intestinal and colonic epithelium are now recognized to arise from the same common stem cell precursor as the other principal cell lineages found in the gut epithelium. A number of different enteroendocrine cell types are found within the colonic epithelium, including L cells, which contain both enteroglucagon and peptide YY (PYY); cells that secrete only PYY; EC1 cells, which secrete serotonin, substance P, and leuenkephalin; pancreatic polypeptide-secreting cells; and rare somatostatin-secreting cells. [23], [24] These enteroendocrine cells are characterized by their polygonal shape, their broad base, and the numerous membrane-bound secretory granules present in their basilar portions (Fig. 82-10). Enteroendocrine cells are identified in pathological specimens using argentaffin or agyrophilic silver staining techniques or through immunohistochemical methods using antibodies to the particular neuroendocrine products stored in each enteroendocrine cell subtype. The narrow apical process of the enteroendocrine cell communicates with the lumen of the crypt and may serve to sense changes in the lumenal environment. Each enteroendocrine cell is joined to other adjacent cells in the epithelium through junctional complexes located near the apical pole. [13] Despite these intracellular attachments, each enteroendocrine cell appears to migrate within the glandular colonic epithelium independent of its neighbors (see previous discussion of migration). The regulatory peptides or bioamine products stored in the basally located secretory granules are secreted through the basolateral membrane and act through paracrine or endocrine mechanisms as mediators of gastrointestinal secretion, absorptive function, and motility in response to lumenally or basolaterally derived signals (see Chapter 8). Enteroendocrine cells are more numerous in the appendix and the rectum than in the rest of the colon. This may explain, in part, the more frequent occurrence of certain subtypes of neuroendocrine tumors at these sites.

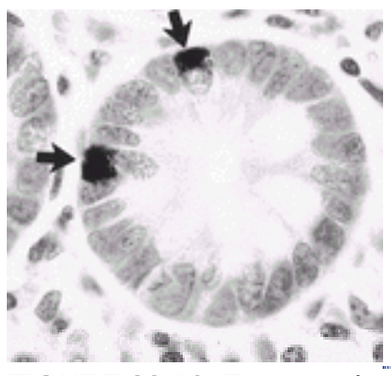

FIGURE 82-10. Enteroendocrine cells (*arrows*) seen in a cross-section of a colonic crypt have a narrow apical process and a broad base filled with membrane-bound granules containing the neuroendocrine secretory product. (Paraffin-embedded 4-µm section, argentaffin stain, Fontana-Masson technique, original magnification ×1000.)

The appendix is similar in histological organization to the rest of the colon. The mucosa of the appendix consists of deep folds lined by a columnar epithelium forming simple tubular or forked glands. [21] This epithelium contains abundant goblet cells and enteroendocrine cells. Numerous lymphoid nodules are found in the lamina propria. The normal histological architecture of the adult appendix is often replaced by fibrous scar tissue as a result of subclinical bouts of appendicitis.

COLONIC DEVELOPMENT

The primitive foregut, midgut, and hindgut develop during the fourth week in utero. The midgut lengthens, herniates out of the abdominal cavity, and rotates 90 degrees counterclockwise around the superior mesenteric artery during the sixth week in utero. By the tenth week, the midgut rotates an additional 180 degrees as it returns to the abdominal cavity. The cecum descends to the right iliac fossa, where it becomes fixed. The base of the small bowel mesentery then runs from the ligament of Treitz in the left upper quadrant to the ileocecal valve in the right lower quadrant. [25], [26]

The small intestine, distal to the entrance of the common bile duct, cecum, appendix, ascending colon, and proximal transverse colon arise from the midgut and are supplied by the superior mesenteric artery. The distal transverse colon, descending colon, sigmoid colon, rectum, upper portion of the anal canal, and part of the lower urogenital tract are derivatives of the hindgut and are supplied by the inferior mesenteric artery. The lower third of the anal canal is ectodermal in origin and is supplied by branches of the internal pudendal artery.

The cloaca is located at the distal end of the hindgut and by the end of the sixth week is separated by the anorectal septum into a ventral urogenital sinus and a dorsal rectum. [27] The anal membrane covering the cloaca ruptures at the end of the eighth week, forming the anal canal. The dentate line is the approximate location of the transition from endoderm to ectoderm, at the junction of the upper two thirds of the anal canal with the lower one third.

The enteric nervous system forms during early fetal life. Neural crest cells, precursors of ganglion cells, migrate from cephalad to caudad during the first 12 weeks in utero. [28] The migration from the midtransverse colon to the anus takes 4 weeks to complete. Smooth muscle cells begin to appear within the mesenchymal layer around the tenth week of gestation. The developing circular muscle layer spreads cranially. The outer longitudinal bands of smooth muscle (teniae coli) and haustra begin to appear between the tenth and eleventh weeks of gestation. [29] By the twelfth week of gestation, Auerbach and Meissner plexi and the circular and longitudinal muscle layers of the colon are well formed, and colonic motility may be observed in isolated fetal colon as early as this developmental stage. [29], [30]

Failure of the neural crest cells to complete their cephalocaudal migration results in a congenital absence of the myenteric plexus in a distal segment of bowel and is thought to be the cause of Hirschsprung disease (see section “Structural and Congenital Abnormalities”). In vitro studies have suggested that components of the extracellular matrix, including laminin and type IV collagen, promote neuronal outgrowth, migration, and survival. Qualitative differences in laminin and type IV collagen have been found in some patients with Hirschsprung disease, suggesting a potential role for abnormalities in fetal extracellular matrix components in the pathogenesis of this disease. [31] Studies using the lethal spotted (ls) mouse, a model for Hirschsprung disease, confirm the importance of nonneural crest–derived tissue components in directing appropriate migration of ganglion cell precursors during normal embryogenesis. [32], [33] Mice that are homozygous for the ls mutation are born without ganglion cells in the distal bowel. Experiments with aggregation chimeras formed from homozygous ls and wild-type mice demonstrate that the aganglionic phenotype results from a defect in mesenchymal cells rather than an intrinsic abnormality of the neural crest–derived neuroblasts. [33]

A number of other genes involved in the susceptibility and development of Hirschsprung disease have also been identified. A recessive susceptibility locus for Hirschsprung disease that encodes the endothelin-B receptor has been mapped to human chromosome 13q22. [34] Targeted and naturally occurring mutations of the mouse endothelin-B receptor gene or in the mouse endothelin-3 ligand gene also result in aganglionosis and a megacolon phenotype as well as defects in development or migration of other neural crest–derived cell lineages, including epidermal melanocytes. [35], [36] Together, these studies suggest that defects in the endothelin-B receptor or its ligand can cause a hereditary form of Hirschsprung disease. Other studies have identified mutations in the RET receptor tyrosine kinase gene located on human chromosome 10q11.2 in an autosomal-dominant form of Hirschsprung disease. [37], [38], [39], [40], [41] and [42] The ligands for this receptor are members of the glial cell line–derived neurotrophic factor family. Over 30 distinct missense and deletion mutations distributed along the entire length of the receptor have been identified in cases of Hirschsprung disease. Most RET mutations associated with Hirschsprung disease result in loss of receptor function. [39] In contrast, mutations in this gene which result in a constitutively active receptor have been found associated with multiple endocrine neoplasia type 2 syndromes (see Chapter 128). [39], [42] RET mutations are found in approximately 10% to 20% of sporadic cases and 50% of patients with a familial history of Hirschsprung disease. [39], [40] and [41] Segregation analysis in a Mennonite family has indicated the existence of other major loci for susceptibility to Hirschsprung disease. [34] These findings confirm the multigenic nature of this disease.

In many respects, the development of the colonic epithelium parallels that of the small intestine epithelium (Fig. 82-11; see Chapter 72). At 8 to 9 weeks of gestation, the developing colon consists of a pseudostratified columnar epithelium several cell layers deep, with a slitlike lumen surrounded by mesenchymal tissue forming a simple tubular structure. [43], [44] and [45] Epithelial ridges form between 10 and 11 weeks of gestation, giving the colonic lumen a stellate appearance. Small secondary

lumens form at the base of the pseudostratified epithelium and subsequently extend to the primary lumen, resulting in the formation of broad primary villi. Between the thirteenth and fifteenth weeks of gestation, the formation of cystlike spaces within these primary villi and the accompanying upward growth of mesenchymal tissue result in the division of primary villi into numerous secondary villi. During this developmental stage, epithelial cells near the bases of the primary villi undergo rapid proliferation and bud into the surrounding mesenchyme to form the developing colonic crypts. Numerous goblet cells, columnar epithelial cells with well-developed microvilli, and enteroendocrine cells can be observed in the epithelium by this time. [23], [46] The fetal enteroendocrine cell population has been found to secrete a variety of neuroendocrine mediators, some of which may have important trophic effects on the developing epithelium. The mesenchymal tissue underlying the developing colonic epithelium appears to play a crucial role in the induction of appropriate regional differentiation in the gut epithelium. [47]

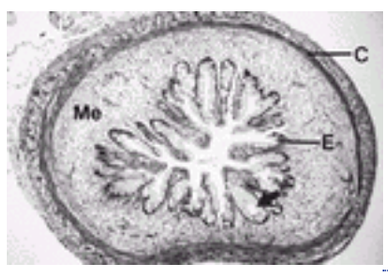

FIGURE 82-11. Human fetal colon at 14 weeks of gestation. Note the villus architecture of the developing epithelium. The circular smooth muscle layer is already well formed. *Arrowhead*, secondary lumen; *E*, columnar epithelium; *C*, circular smooth muscle layer; *Me*, mesenchymal tissue. (Paraffin-embedded 4-µm section stained with hematoxylin and eosin, original magnification ×100.)

Some studies suggest that homeotic genes of the CDX family may regulate regional specificity of the stem cells in the developing gut. [48], [49] and [50] Prominent supranuclear inclusion bodies can be observed in the principal epithelial cells lining the fetal colon. These inclusion bodies were originally termed meconium corpuscles because it was thought that they arose from uptake of lumenal contents present in the developing colon. However, more recent studies have found that these membrane-bound inclusion bodies are a result of apoptosis, emphasizing the role of programmed cell death in the morphologic restructuring of the epithelium that occurs during fetal life. [51] Colonic villi are present through at least week 29 of gestation but are not found in the colon of full-term infants. [46], [52] During fetal life, the colonic epithelium is also similar to the developing small intestine in its biochemical characteristics. For instance, the initial appearance and the activity of many brush border hydrolases found in the fetal colonic epithelium, including sucrase-isomaltase complex and at least five dipeptidases, are similar to those found in the developing small intestine. [43], [52], [53], [54] and [55] By full term, however, the activities of these brush border hydrolases have declined to levels similar to those found in the adult colon.

STRUCTURAL AND CONGENITAL ABNORMALITIES

Congenital abnormalities of the intestine are common; most arise from errors in rotation or fusion of the mesenteries or from failure of neural crest cells to complete their migration during fetal colonic development. Imperforate anus, duplication, and malrotation are usually identified in the neonatal period. Hirschsprung disease may present in the neonatal period, in childhood, or, more rarely, in the young adult.

Omphaloceles and Gastroschisis

Abdominal wall defects are present at birth. Omphaloceles are identified by a mass of bowel and viscera in the central abdomen covered by a translucent membrane. The size of the defect varies, and it can be associated with other congenital anomalies. Gastroschisis is usually a small defect that occurs at the junction of the umbilicus and normal skin. Although no membrane covers the bowel, there are usually no associated anomalies. Both omphaloceles and gastroschisis require immediate surgery. Only in gastroschisis are reduction of the bowel and primary closure of the defect usually possible. If the bowel is edematous and thickened or the defect is too large to close primarily, a Silastic silo can be constructed to achieve gradual reduction of the bowel (see Chapter 72 for a more complete discussion).

Hirschsprung Disease and Congenital Megacolon

Hirschsprung disease is a result of the failure of neural crest cells (precursors of ganglion cells) to complete their caudal migration during normal colonic development (see section “ Colonic Development”). The aganglionic segment does not relax and causes a functional obstruction. The normal proximal bowel hypertrophies and eventually dilates. The rectosigmoid is involved in approximately 75% to 80% of cases. [56] The entire colon and various lengths of small bowel are aganglionic in 5% to 10% of cases. [57] A very short aganglionic segment is the typical finding in adult Hirschsprung disease.

Delayed passage of meconium and abdominal distention are often the presenting symptoms in the newborn. [58] Chronic constipation, abdominal distention, volvulus, or perforation may be symptoms of the disease in a child. Hirschsprung disease in the adult is rare but must be considered in the evaluation of patients with chronic constipation dating back to childhood. [59], [60], [61] and [62] Patients with total colonic aganglionosis may present with less dramatic intermittent symptoms of constipation. [57]

Digital rectal examination aids in the diagnosis. The rectal vault is empty in patients with Hirschsprung disease and is full of stool in patients with idiopathic megacolon. Abdominal radiographs usually show colonic dilation and a paucity of gas in the rectum. Barium enema may suggest the presence of a short, narrow segment or transition zone (Fig. 82-12). Patients with total aganglionosis or very short segment Hirschsprung disease may have a normal-appearing barium enema. Newborns may have a normal-appearing barium enema because the proximal bowel has not had time to dilate.

FIGURE 82-12. Radiologic study from a patient with Hirschsprung disease. The short, collapsed, aganglionic segment (*arrows*) is evident on this radiograph of a barium enema performed on a 28-month-old infant with constipation. The colon proximal to the aganglionic segment is often dilated and filled with stool.

Anorectal manometry may be useful in the evaluation of patients with suspected Hirschsprung disease but is difficult to perform in the newborn and is reserved for older infants and children. Typically a normal sphincter profile (resting pressure of 40–80 mm Hg and squeeze pressure of 80–160 mm Hg) and an abnormal rectoanal inhibitory reflex are seen in patients with Hirschsprung disease. The rectoanal inhibitory reflex is the normal relaxation of the internal sphincter in response to rectal

distention. The internal sphincter does not relax in response to rectal distention in patients with Hirschsprung disease, and an abnormal reflex during manometry aids in the diagnosis. [63], [64] This reflex is easily identified using anal manometry with a manometric catheter and a balloon to distend the rectum. When the rectal balloon is distended, the manometry tracing will reveal a normal external sphincteric contraction but no compensatory relaxation of the internal anal sphincter. The rectoanal inhibitory reflex may be normal in patients with short-segment Hirschsprung disease. These patients may have a short segment as their primary disease or the short segment may be residual disease after surgery. High anal resting pressures and impaired rectal emptying may be seen.

Histological evaluation is necessary to make the diagnosis of Hirschsprung disease. Hyperplasia or hypertrophy of nerve fibers can be seen, but ganglion cells are absent in the submucosa and myenteric plexus of these patients. Mucosal pinch biopsy specimens are not adequate for diagnosis because sufficient submucosa must be present in the biopsy specimen in order to conclude that ganglion cells are absent. Suction biopsies are simple to perform, but the mucosa and submucosa must be identifiable to have an adequate specimen. Full-thickness rectal biopsy samples may be necessary for confirmation. Routine hematoxylin and eosin staining misses ganglion cells in almost 40% of normal patients. [65] Acetylcholinesterase staining has been found to be more accurate in identifying patients with Hirschsprung disease. Acetylcholinesterase-positive nerve fibers are increased in number in the lamina propria and muscularis propria in these patients. A positive diagnosis may be obtained even by evaluation of superficial suction biopsy samples.

Colostomies are usually performed after the diagnosis is made in newborns. This allows a period of stabilization and growth before definitive repair. A colostomy may be used in the older child or adult to allow hypertrophied bowel to resume a more normal caliber. Pull-through operations (Swenson technique, Duhamel procedure, or Soave procedure) have been devised to anastomose normally innervated bowel to the anus with or without resection of the abnormal bowel. [58], [59], [60] and [61] Intraoperative biopsy results must demonstrate the presence of ganglion cells at the level of the colostomy and the proximal margin of the colonic resection. Rectal myectomy may be curative in patients with short-segment Hirschsprung disease.

Intestinal Duplications

Intestinal duplications are rare congenital anomalies that can occur anywhere along the gastrointestinal tract. There are many theories regarding the cause of intestinal duplication, including failure of the bowel to recanalize in utero, caudal twinning, and incomplete separation of the notochord from the entoderm. The resultant structures may either be tubular, communicating with the intestinal lumen, or, more commonly, closed cystic duplications. These cysts are lined with gastrointestinal mucosa and contain a layer of smooth muscle within their walls. Duplications are usually found on the mesenteric side of the intestine. They frequently share a common wall and blood supply with the involved bowel. All regions of the gastrointestinal tract may be involved; the esophagus and ileum are the most commonly involved sites. The colon and rectum account for 5% to 10% of all gastrointestinal duplications. [66]

Duplications are usually symptomatic during childhood. They can produce symptoms of obstruction, volvulus, or hemorrhage. Duplications in adults are often asymptomatic and may be discovered incidentally. They can be complicated by infection, bleeding, or malignant degeneration. Plain abdominal radiographs, barium enemas, and computed tomography scans may reveal soft tissue masses or the point of communication with normal bowel. Evaluation of the genitourinary tract should be performed to rule out associated anomalies.

Resection is indicated for rectal duplications because of the risk of neoplasia, even for asymptomatic duplications. Cystic duplications can often be excised. Dividing the common wall is the treatment for tubular duplications. Mucosal excision may be necessary if heterotopic gastric mucosa is present. [66]

Malrotation

Malrotation occurs if the midgut fails to complete the 270-degree counterclockwise rotation as it returns from herniation (10–12 weeks of gestation). [26], [67] The base of the small bowel mesentery normally stretches from the ligament of Treitz in the left upper quadrant to the ileocecal valve in the right lower quadrant. In malrotation, the entire midgut and its vascular pedicle have a narrow-based mesenteric attachment in the right upper quadrant. The duodenojejunal junction (ligament of Treitz) comes to lie to the right of the spine. The cecum and right colon overlie the duodenum in the right upper quadrant, and Ladd bands form over the duodenum in an attempt to peritonealize the cecum. Associated anomalies, such as small bowel atresias, intussusception, Hirschsprung disease, and abdominal wall defects, have been reported in 30% to 60% of patients. [67], [68]

Most infants present with evidence of malrotation within one month of birth. [67], [68] The clinical picture is one of a high small bowel obstruction, volvulus, or ischemia. One should suspect the diagnosis in an infant with bilious vomiting and a flat abdomen. Bloody stools indicate vascular compromise. Less severe forms of malrotation can present in childhood or into adulthood.

Plain abdominal radiographs may show evidence of a complete gastric or duodenal obstruction with a paucity of gas in the distal small bowel. A barium upper gastrointestinal series shows the ligament of Treitz to the right of midline. A barium enema is rarely needed, but it may demonstrate the cecum in the right upper quadrant.

The treatment of malrotation is operative. Most neonates with an obstruction from malrotation have a midgut volvulus. [68] The volvulus is reduced by untwisting it in a counterclockwise rotation. [69] Ladd bands overlying the right colon and duodenum are lysed. The duodenum is placed in the right upper quadrant and the cecum in the left upper quadrant. An appendectomy is performed because the alteration in anatomy would make future diagnosis of appendicitis difficult. [68]

Imperforate Anus

Imperforate anus occurs in 1 of 20,000 live births and is associated with other congenital anomalies in 50% of cases. Genitourinary anomalies and sacral abnormalities are frequently seen. Cardiac and gastrointestinal anomalies (primarily esophageal atresia) are also associated with imperforate anus.

Imperforate anus is classified according to the relationship of the rectum to the levator ani muscles. [70] High lesions are those in which bowel of normal or greater caliber terminates above the levator muscle complex. The rectum often ends in the prostatic urethra in males. In low lesions, the rectum terminates distal to the levators, often ending in a fistula. Low lesions are more common in females. Fistula tracts may connect the distal rectum with the perineum, vagina, or other hollow organs. Intermediate lesions are found within the levator complex and may be associated with a partial anal canal or perineal fistula.

The diagnosis is usually made by physical examination at birth. Most infants with imperforate anus fail to pass meconium at birth unless a fistula is present. Radiography can be used to determine the type of defect. Air invertograms can be used to identify the location of the rectum in relation to the perineum but are difficult to interpret. Fistulography may be performed, and computed tomography and magnetic resonance imaging are useful in evaluating high lesions. The urinary tract should be evaluated with an intravenous pyelogram and voiding cystourethrogram because of the associated urogenital anomalies.

Treatment is dependent on the type of lesion. [70] Low lesions can be treated by dilation of fistula tracts with or without anoplasty. Most healthy infants with low lesions do not require colostomies. Intermediate and high lesions are treated with an initial diverting colostomy to allow a period of stabilization and growth. A number of pull-through operations have been advocated. The posterior sagittal approach developed by Peña and deVries [71] identifies and divides the levator muscle complex. The rectal segment is then identified and brought through the center of the divided muscles. A coloanal anastomosis is performed in the location of the anal dimple. Most series have claimed a 70% to 82% success rate with intermediate and high lesions. [72] A greater success rate can be expected with low lesions.

Volvulus

Volvulus occurs whenever an air-filled segment of bowel twists about its mesentery and almost never occurs if the colon is filled with solid stool. A dilated, redundant colon with a narrow-based mesocolon is a prerequisite for colonic volvulus. In the United States, fewer than 10% of colonic obstructions are caused by colonic volvulus. In countries with high-fiber diets, volvulus has been reported as the most common cause of intestinal obstruction. [73] Volvulus is classified and treated according to its location in the colon.

Sigmoid volvulus accounts for approximately 60% of all volvulus seen in the United States. [74] It usually occurs in older adult or institutionalized patients or in patients with various neuropsychiatric disorders. Chronic constipation, laxative abuse, and colonic atony have been implicated as the causes of the dilated, redundant sigmoid

colon.

Patients usually present with abdominal pain, distention, and obstipation. Evidence of peritoneal irritation on physical examination, fever, or an elevated leukocyte count indicates gangrenous bowel. Plain abdominal radiographs showing an inverted, U-shaped, sausagelike loop with a dense line running toward the point of torsion are diagnostic for sigmoid volvulus. In questionable cases, radiologic enema examination with barium or with water-soluble contrast may be diagnostic. The "bird's beak" deformity forms where the contrast tapers to a point at the site of obstruction (Fig. 82-13). Reduction of the volvulus can occur during these examinations. If the patient has signs of peritoneal irritation or gangrene is suspected, barium or Gastrografin enema (Bracco Diagnostics, Inc., Princeton, NJ) should not be attempted, and the patient should undergo emergency exploration.

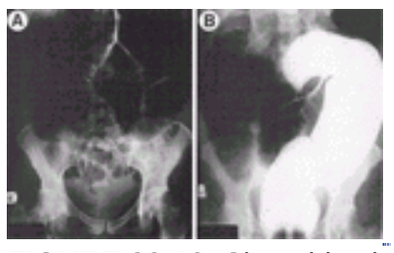

FIGURE 82-13. Sigmoid volvulus. **A**: The gas-filled, dilated colon is evident on a plain film of the abdomen. Note the paucity of gas in the rectum. **B**: Hypaque enema shows tapering of the column of contrast at the site of torsion ("bird's beak" sign) of the sigmoid colon.

If peritonitis is not present, sigmoidoscopy should be performed. Inserting a soft rectal tube past the point of obstruction can often reduce the volvulus. This results in dramatic decompression, and the rectal tube is left in place to assist with further decompression. Volvulus beyond the limits of the rigid sigmoidoscope may be reduced by flexible sigmoidoscopy or colonoscopy. [75] A 43% recurrence rate after nonoperative reduction can be expected. [76] Therefore, after reduction, medically stable patients should undergo mechanical bowel preparation and an elective resection. Evidence of mucosal ischemia, bloody discharge, or unsuccessful detorsion indicates strangulation and possibly gangrene. If this occurs, sigmoidoscopy should be terminated, and the patient should undergo an emergency exploration. Mortality is related to the presence of gangrenous bowel. [76]

Cecal volvulus accounts for fewer than 20% of all cases of colonic volvulus and is generally seen in younger patients. [75] It is thought to be caused by anomalous fixation of the right colon, leading to a freely mobile cecum. [77] Other precipitating factors include adhesions from previous surgeries, pregnancy, and obstructing lesions of the left colon. Ninety percent of patients with cecal volvulus have a full axial volvulus twisting the associated mesentery and blood vessels. [73] In the remaining patients, the cecum folds in an anterior cephalad direction (cecal bascule). The cecal bascule is not a true volvulus around a mesentery, but gangrene can result from tension on the bowel wall.

Abdominal pain, nausea, vomiting, and obstipation are common symptoms of cecal volvulus. Patients with cecal volvulus clinically appear to have a small bowel obstruction. Many patients give a history of similar, chronic, intermittent symptoms. [73] A plain abdominal radiograph may show the cecum as a kidney-shaped, air-filled structure in the left upper quadrant. Multiple air-filled levels may be seen and indicate a distal small bowel obstruction. Radiologic examination with barium or with a water-soluble contrast enema may show obstruction of the column of contrast at the level of the volvulus (Fig. 82-14). The tapered edge of the contrast points toward the site of torsion. Barium studies should not be routinely performed if the diagnosis is clear. Colonoscopy has been used to treat cecal volvulus with limited success and should rarely be attempted. [75], [78]

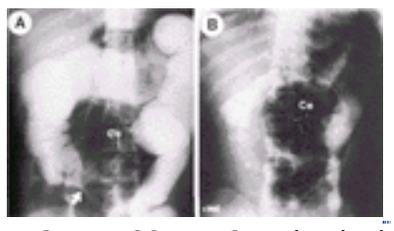

FIGURE 82-14. Cecal volvulus. **A**: Hypaque enema shows tapering of the column of contrast at the site of torsion (*arrow*) and displacement of the cecum toward the epigastrium. **B**: The gas-filled, dilated cecum (*Ce*) is evident in the epigastrium in this postevacuation radiograph. Note the paucity of gas in the normal location of the cecum in the right lower quadrant of the abdomen.

Operative detorsion alone, cecopexy, and cecostomy are associated with variable rates of recurrence. [73], [79] Right hemicolectomy with primary anastomosis or cecopexy and cecostomy is recommended if there is no evidence of gangrenous bowel. Resection, ileostomy, and mucous fistula are indicated if there is evidence of gangrene. Mortality is increased in the presence of gangrenous bowel.

Volvulus of the transverse colon is rare because the mesentery is short and broad-based. Failure of the mesentery to fuse normally or narrowing of the mesenteric attachments may predispose the transverse colon to volvulus. Patients present with a clinical picture of large bowel obstruction. Barium enema is diagnostic and reveals the point of obstruction. Although rare successful attempts at colonoscopic detorsion have been reported, most patients require operative detorsion and resection of the redundant transverse colon. [80], [81]

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CHAPTER 83

William F. Stenson and Joshua Korzenik

INFLAMMATORY BOWEL DISEASE

Ulcerative colitis and Crohn's disease, chronic inflammatory diseases of the gastrointestinal tract, are identified and diagnosed by the appearance of a set of clinical, endoscopic, and histological characteristics. [1], [2] and [3] The inflammatory response in ulcerative colitis is largely confined to the mucosa and submucosa, but in Crohn's disease inflammation extends through the intestinal wall from mucosa to serosa. Ulcerative colitis is confined to the colon, and colectomy is a curative procedure. Crohn's disease, in contrast, has the potential to involve the patient's entire gastrointestinal tract, even though only a small segment is involved initially. Resection of the inflamed segment is not curative in Crohn's disease, and inflammation is likely to recur in the gastrointestinal tract sometime after resection. Despite these differences in distribution, no single finding is absolutely diagnostic of one disease or the other. Moreover, a group of patients, accounting for 5% to 15% of those with inflammatory bowel disease (IBD), have characteristics of both diseases and cannot be cleanly classified; these patients are considered to have indeterminate colitis.

The genetic basis of these diseases is being elucidated. Both appear to be multigenic diseases. Patterns of family aggregation suggest that the genetic influences at least partially overlap. Relatives of patients with each disease have an increased risk of that particular disease as well as the other form of IBD. [4] Consequently, not only is there an increased incidence of Crohn's disease in relatives of those with Crohn's disease but their relatives also have an increased incidence of ulcerative colitis. This pattern holds for patients with ulcerative colitis as well.

In view of the similarities in clinical and histological presentation and the shared genetic background, these two diseases are presented in a single chapter. In the absence of established etiologic agents or definitive markers, it is possible that what is designated as ulcerative colitis or Crohn's disease may in fact be a mixture of

diseases of diverse causes but common clinical presentation. Actually, more than two diseases may be under the name of inflammatory bowel disease. Presentation in a single chapter emphasizes the common characteristics of these diseases and minimizes splitting into groups that may not be valid. This combined presentation allows the areas of similarity to be detailed without repetition and allows comparisons in areas of dissimilarity.

EPIDEMIOLOGY

The incidence and prevalence of Crohn's disease (1–6 and 10–100/100,000, respectively) and ulcerative colitis (2–10 and 35–100/100,000, respectively) vary greatly with geographic location. These rates are for Caucasian populations in northern Europe and North America. The countries of greatest incidence and prevalence encompass the industrialized world. Rates in central and southern Europe are somewhat lower. In South America, Asia, and Africa these diseases remain uncommon but appear to be increasing. Within geographic areas, ethnic and racial variations exist in the incidence of IBD. [5] Crohn's disease is three to eight times more common in Jewish than in non-Jewish persons. Similarly, the incidence of ulcerative colitis is two to four times higher among Jews. The incidence among Israeli Jews, however, is much lower than among American and European Jews. Furthermore, in Israel the incidence is lower among Sephardic or Oriental Jews of Asian or African origin than among Ashkenazi Jews of European or American origin. [5] In the United States, the incidence of both ulcerative colitis and Crohn's disease in the African-American population has been one-fifth to one-half that in the Caucasian population, but in recent years that gap appears to be narrowing. [6]

Some of the apparent increases in incidence of IBD over time may reflect greater awareness of these diseases, better diagnostic studies, and improved reporting. Moreover, some patients now diagnosed as having Crohn's colitis may have been diagnosed as having ulcerative colitis in the past. Despite these ambiguities, certain trends are apparent. [7], [8] In geographic areas where the incidence of these diseases has been slight, it is now increasing. In northern Europe and North America, where the incidence of both diseases has been substantial, the incidence of ulcerative colitis has leveled, but that of Crohn's disease is still increasing.

The peak age of onset for both diseases is between 15 and 25 years. In some but not all series, a second, lesser peak of incidence occurs between 55 and 65 years. [6] Both diseases occur in childhood, although the incidence is much lower before the age of 15 years than after. Ulcerative colitis is more common than Crohn's disease in children younger than 10 years. Most series show an approximately equal incidence of both diseases in males and females, but other studies show an incidence of Crohn's disease that is greater in females by up to 30% and an incidence of ulcerative colitis that may be greater among males. [6] These gender differences of disease distribution may represent confounding environmental factors such as use of tobacco. The similarities between ulcerative colitis and Crohn's disease in geographic distribution, racial and ethnic distribution, distribution by sex, and age of onset support the contention that the two diseases are related.

One fascinating but unexplained difference between patients with these two diseases is the incidence of smoking. Among patients with ulcerative colitis, the incidence of smoking is less than that in the general population. In several studies, the risk of developing ulcerative colitis was increased among nonsmokers compared with current smokers and increased even further among former smokers compared with lifetime nonsmokers. [9], [10], [11] and [12] Quitting smoking can provoke the emergence of ulcerative colitis, though still much less of a risk than smoking on overall health. [13], [14] In contrast, the incidence of smoking among patients with Crohn's disease is higher than in the general population. [10] Smokers have a more severe course of Crohn's disease, require more immunosuppressive medications, and demonstrate a faster recurrence after surgery. [15], [16] Cessation of smoking may moderate the clinical course although the greatest benefit of cessation is seen with the heaviest smokers. [17] The active ingredients in cigarette smoke that increase the risk of Crohn's disease or decrease the risk ulcerative colitis have not been defined, nor have mechanisms of action. On the assumption that nicotine is the active ingredient in cigarette smoke, trials of transdermal nicotine and nicotine gum for the treatment of ulcerative colitis have been performed; in two trials transdermal nicotine was somewhat effective. [18], [19]

Appendectomy strongly protects against the development of ulcerative colitis, while not having any influence on Crohn's disease. [20] This effect in ulcerative colitis is noted particularly when appendectomy is performed for an inflammatory problem, such as lymphadenitis or appendicitis and if done before the age of 21. [21] Poor sanitation in childhood is a protective factor against the development of Crohn's disease, though not against ulcerative colitis. [22], [23]

ETIOLOGY AND PATHOGENESIS

Genetics

Any theory of the pathogenesis of IBD must deal with the evidence for a genetic basis of the disease. The most firmly established and quantitatively greatest risk factor for developing IBD is a positive family history. [24] Approximately 15% of patients with IBD have first-degree relatives who also have IBD. [25] Lifetime risk of developing IBD among first-degree relatives of affected individuals is 8.9% for offspring, 8.8% for siblings, and 3.5% for parents. [26] The incidence of disease among first-degree relatives is higher in Crohn's disease than in ulcerative colitis. The incidence of IBD among first-degree relatives of IBD patients is 30 to 100 times that of the general population. Moreover, although the relatives of patients with Crohn's disease are more likely to have Crohn's disease than ulcerative colitis, the incidence of ulcerative colitis in this group is also higher than in the general population. [4] Similarly, relatives of patients with ulcerative colitis have a higher incidence of both ulcerative colitis and Crohn's disease than the general population. These data support the contention that ulcerative colitis and Crohn's disease are related diseases. The incidence of IBD is not increased in spouses of patients.

Studies of twins also support the presence of a genetic basis for these diseases. A study of unselected twins from a Swedish twin registry demonstrated that dizygotic twins have the same rate of concordance as would be expected for siblings. There was concordance for Crohn's disease in 8 of 18 monozygotic twin pairs; whereas there was concordance for ulcerative colitis in only 1 of 16 monozygotic twin pairs. [27] No case has been reported of monozygotic twins in which one twin had Crohn's disease and the other ulcerative colitis, suggesting that these diseases have a similar but not identical genetic background.

No clear-cut mendelian pattern of inheritance in IBD has been established. There are, however, human leukocyte antigen (HLA) class II genes that have been associated with Crohn's disease and ulcerative colitis. The *DR1/DQw5* and *DRB3*0301* haplotypes have been associated with Crohn's disease, [28] and HLA-DR2 has been associated with ulcerative colitis. [29] Linkage analysis has identified two susceptibility loci; IBD1 on chromosome 16 [30] and IBD2 on chromosome 12. [31] IBD1 has been associated primarily with Crohn's disease, whereas, IBD2 has been associated with both Crohn's disease and ulcerative colitis. Work by two different groups has led to the identification of NOD2 as the gene associated with IBD1. [32], [33] NOD2, a cytosolic protein expressed in monocytes, functions as an intracellular receptor for lipopolysaccharide. Crohn's disease is associated with frameshift mutations in NOD2 resulting in a truncated and nonfunctioning protein. Although NOD2 mutations are seen much more commonly in Crohn's disease than in the general population, only a small portion (~15%) of all Crohn's disease patients have a mutation in NOD2. This demonstrates that Crohn's disease is genetically heterogeneous and suggests that other mutations in other genes may contribute to Crohn's disease in other populations.

It has become increasingly clear over the last few years that Crohn's disease is clinically heterogeneous and that there is a genetic basis for the clinical heterogeneity. Not only are patients with Crohn's disease more likely to have relatives with Crohn's disease, but those relatives are likely to have Crohn's disease that is similar to that of the proband in terms of anatomic location, age of onset, and disease behavior (inflammatory vs. fistulizing vs. stenotic). [34], [35], [36] and [37] In affected parent-child pairs, the age of onset for the parents was significantly higher. In contrast, affected siblings were highly concordant for age of onset. [35] It is not clear whether these genetically based clinical patterns reflect differences in the genes associated with Crohn's disease or the presence of another set of genes that affect the timing and location of the appearance of Crohn's disease in patients who are genetically disposed. Genetic markers have been implicated as predictive of disease severity in ulcerative colitis. [38] HLA *DRB1*0103* and allele 2 of the interleukin 1 (IL-1) receptor antagonist are associated with extensive disease.

Another approach to understanding the genetic basis of IBD is the search for subclinical markers, that is, parameters used to detect the abnormal genotype in the absence of the full clinical phenotype. The two subclinical markers that have received the most attention are increased intestinal epithelial permeability in Crohn's disease and antineutrophil cytoplasmic antibodies (ANCA) in ulcerative colitis. There is a report of increased intestinal permeability to PEG-400 in Crohn's disease patients and their first-degree relatives. [39] A genetic defect of intestinal epithelial cells allowing permeation of lumenal antigens across the epithelial monolayer could result in nonspecific activation of the intestinal immune system; however, subsequent studies have yielded inconclusive data. [40], [41] The other subclinical marker that has received considerable attention is the presence of ANCA in the sera of patients with ulcerative colitis and Crohn's disease. [42] Two groups of ANCA-positive patients have been defined based on this intracellular staining pattern: pANCA positivity reflects a perinuclear pattern, whereas cANCA reflects a diffuse cytoplasmic pattern. About 70% of patients with ulcerative colitis are pANCA positive, with no correlation with disease extent or activity. Fewer than half of patients with Crohn's disease are ANCA-positive and these patients are about evenly divided between cANCA-positive and pANCA-positive. Many patients with Crohn's disease who are pANCA-positive have left-sided colitis and a clinical picture similar to ulcerative colitis. [43] The role, if any, of ANCA in the pathogenesis of IBD is unknown; however,

clinically healthy relatives of patients with ulcerative colitis have an increased incidence of ANCA positivity compared with the general population, suggesting that ANCA is more than just a marker for colonic inflammation. [44]

H4>Potential Etiologic Agents and Antigenic Triggers

Clear evidence exists for the activation of the immune response in IBD. The lamina propria is infiltrated with lymphocytes, macrophages, and other cells of the immune system. A gene chip analysis of gene expression in ulcerative colitis mucosa demonstrated up-regulation of many genes associated with immune activation. [45] In any immune response, a specific antigen serves as a trigger for the response and as a target for the effector arm of the response. Over the past 30 years, an intensive search has been conducted for the antigens that trigger the immune response in IBD. Immune activation in IBD is largely confined to the gastrointestinal tract; therefore, the search for the antigenic trigger has focused on the intestinal lumen, where most of the antigens are of microbial or dietary origin. Three major hypotheses as to the antigenic triggers in IBD have been postulated.

1. One hypothesis is that the antigenic triggers are microbial pathogens that have not yet been identified because of fastidious culture requirements. According to this hypothesis, the immune response in IBD is an appropriate but ineffective response to these pathogens. Various viruses and bacteria have been proposed as candidate organisms, but little evidence has been found to support any of these organisms as having a causative role in IBD. One organism that created some interest is a species of mycobacteria similar to or identical with *Mycobacterium paratuberculosis*; however, there is little data to support this organism as a causal agent in Crohn's disease. [46], [47] and [48]
2. The second (and best supported) hypothesis as to the antigenic trigger in IBD is that it is some common dietary antigen or usually nonpathogenic microbial agent against which the patient mounts an abnormal immune response. In healthy persons, a finely tuned, low-grade chronic inflammation is present in the intestinal lamina propria. Presumably, this chronic inflammation is a product of chronic exposure of the lamina propria to lumenal antigens. Failure to suppress this inflammatory response could result in the uncontrolled immune activation seen in IBD. As a result of failure of normal suppressor mechanisms, immune activation in IBD may be an inappropriately vigorous and prolonged response to some normal lumenal antigen. The genetic basis of IBD may relate to a genetically determined ability to mount an immune response to a specific lumenal antigen. It may be that patients with IBD are genetically programmed to mount an intense immune response to some common lumenal antigen—either dietary or microbial—to which most people do not respond. In genetically based mouse models of colitis, defects in T-cell function or cytokine production result in uncontrolled immune responses to normal colonic bacteria. [49] In IL-10(-/-), IL-2(-/-), and T-cell receptor (TCR-/-) mice inflammation only develops in the presence of normal colonic flora. If these mice are grown under germ-free conditions they do not develop inflammation. These studies demonstrate that normal colonic flora can stimulate a pathological inflammatory response in the presence of a defect in immune regulation. Moreover, a number of quite different defects in immune regulation result in a mucosal inflammatory response to normal colonic flora. This has two potential implications for IBD. The first is that IBD may result from the initiation of an immune response to normal flora as a result of a defect in immune regulation. The second is that ulcerative colitis and Crohn's disease may be a group of diseases resulting from a number of defects in immune regulation but all manifesting themselves as a mucosal inflammatory response to normal flora. The demonstration that a fraction of patients with Crohn's disease have mutations in NOD2, a gene that codes for a protein involved in the response to bacterial products, supports both the suggestion that Crohn's disease involves a dysregulated immune response to bacteria and the suggestion that Crohn's disease is genetically heterogeneous. [32], [33]
3. The third hypothesis (autoimmunity) as to the antigenic trigger in IBD is that it is one expressed on the patient's own cells, particularly intestinal epithelial cells. In this theory, the patient mounts an appropriate immune response against some lumenal antigen, either dietary or microbial; however, because of similarities between proteins on the epithelial cells and the lumenal antigen, the patient's immune system also attacks the epithelial cells. Under the autoimmune theory, the immune response is directed specifically toward the epithelial cell, and the epithelial cell is destroyed by one of two immune effector mechanisms, either antibody-dependent cellular cytotoxicity, which requires an antibody directed against an epithelial cell surface antigen, or direct cell-mediated cytotoxicity. Although little evidence exists to support a role for autoimmunity in Crohn's disease, several investigators have reported the presence of anticolon antibodies in the sera of ulcerative colitis patients; [50], [51] however, similar antibodies have been found in the sera of healthy persons and in patients with systemic lupus erythematosus in whom there was no intestinal inflammation. [52] Das and colleagues [53], [54] demonstrated the presence of a tissue-bound antibody that could be eluted from ulcerative colitis surgical resections; this antibody is directed against tropomyosin. Whether this antibody participates in the pathogenesis of ulcerative colitis or is just a marker for immune activation is not clear. The presence of an anticolon antibody does not necessarily mean that the antibody plays a pathogenetic role. A more convincing case for a pathogenetic role could be made by the demonstration that this antibody participates in antibody-dependent cellular cytotoxicity.

Immune Response

The first step in the immune response to an antigen is the uptake and processing of the antigen by macrophages or other antigen-presenting cells (Fig. 83-1). CD4 (helper/inducer) T cells recognize soluble antigens in conjunction with HLA class II molecules (HLA-DP, -DQ, and -DR) present on macrophages. Class II antigens on intestinal epithelial cells also may play a role in antigen processing within the intestinal immune compartment. Class II molecules are present on epithelial cells of the normal human small intestine but not on those of the colon. Class II antigen expression on small intestine epithelial cells is retained in patients with Crohn's disease. HLA-DR-like molecules on human intestinal epithelial cells may be involved in antigen-induced triggering of intraepithelial lymphocytes. Marked enhancement of colon epithelial cell HLA-DR staining occurs in active ulcerative colitis and Crohn's colitis, and intestinal epithelial cells from patients with IBD are capable of processing and presenting antigens in vitro. [55]

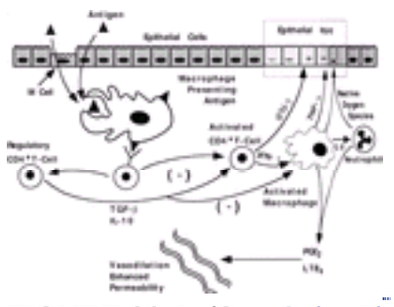

FIGURE 83-1. (See Color Fig. 83-1.) An immunologic sequence that has been postulated to account for the inflammatory response observed in inflammatory bowel disease starts with an exogenous sensitization to lumenal antigens, presumably bacterial, possibly facilitated by undefined genetic influences. The first cell population involved is the intestinal macrophage, which phagocytoses and processes antigen and presents it to a $CD4^+$ T cell. Two populations of $CD4^+$ T cells are produced: one an activated population and the second a population of CD45RBlo $CD4^+$ regulatory T cells. The activated T cells produce interferon-? (*IFN*-?), which activates macrophages and participates in epithelial injury. The CD45RBlo $CD4^+$ regulatory T cells produce transforming growth factor-ß (*IFN*-ß) and interleukin-10 (IL-10), which down-regulate both the activated $CD4^+$ cells and the activated macrophages. Activated macrophages participate in epithelial injury by producing tumor necrosis factor-a (*TFN*-a) and reactive oxygen species and by recruiting neutrophils, which also produce reactive oxygen species. Macrophages and neutrophils also produce prostaglandin E_2 (PGE_2) and leukotriene B_4 (LTB_4), which contribute to the vasodilation and enhanced vascular permeability that is characteristic of inflammatory bowel disease.

The activated macrophage, in addition to presenting antigen to the T cell, releases IL-1, which activates the T cell. The activated T cell releases IL-2, which promotes the clonal expansion of cytotoxic T cells and increases the function of helper T cells and B cells. Cell-mediated immune responses may be involved in the pathogenesis of IBD. There is a finely balanced chronic low-grade immune response in the normal intestine and colon. The immune response is a product of $CD4^+$ T-cell activation by bacteria in the lumen, as the response is largely absent in germ-free animals. This activation is promoted by interferon-? (IFN-?) and is down-regulated by IL-10 and transforming growth factor-ß (TGB-ß) produced by regulatory T cells (see Fig. 83-1). The current data suggest that IBD is a product of genetically determined immune dysregulation to lumenal antigens. Whether the immune dysregulation produces an overly active up-regulation of the immune response or an inadequate down-regulation is not clear. It is also not clear whether this dysregulation is specific to certain antigens.

Antibody Secretion

In view of the increased access of lumenal antigens to the immune cells of the lamina propria, it is not surprising that there is increased antibody secretion by

intestinal mononuclear cells in IBD. Not only is the number of antibody-secreting cells increased, but the distribution of immunoglobulin (Ig) classes is changed. In healthy persons, the vast majority of the immunoglobulin secreted by intestinal mononuclear cells is IgA. In IBD, there is markedly increased production of IgM and IgG. IgA does not fix complement, whereas IgM and some IgG subclasses do. There are subclasses of IgG that are defined by the structure of their constant regions. There is some subclass specificity in the increase in IgG production seen in IBD. Ulcerative colitis is associated with increased production of IgG1 and IgG3, whereas Crohn's disease is associated with increased production of IgG2. IgG1 and IgG3 antibodies account for the predominant IgG response to proteins and T cell–dependent antigens. IgG2 provides the predominant IgG response to carbohydrates and many bacterial antigens. Delineation of the stimuli and antigens that induce the increased secretion of IgG subclasses may provide insight into the etiology and pathogenesis of IBD.

Cytokines

Cytokines are glycosylated proteins that mediate potent biologic function at low concentrations in a hormonelike fashion (see section " Cytokines", Chapter 7). Like hormones, they act through receptors, but unlike hormones, which act on cells at a distance from their cell of origin, cytokines act on neighboring cells. Originally, cytokines were described as being produced by immune cells and acting on immune cells, but it is clear that they are both produced by and act on nonimmune cells. Cytokines almost certainly play important regulatory roles in IBD, as they do in other chronic inflammatory states. [56], [57] The best developed data are those demonstrating increased production of most of the major proinflammatory cytokines (IL-1, IL-6, IL-8) and tumor necrosis factor-a (TNF-a). The proinflammatory cytokines contribute to many of the characteristic attributes of IBD: neutrophil infiltration (IL-8), T-cell activation (IL-1), expression of adhesion molecules (IL-1 and TNF-a), and enhanced epithelial permeability (IFN-?). TNF-a and IFN-? may contribute directly to epithelial injury (see Fig. 83-1). The natural inhibitor of IL-1, IL-1 receptor antagonist (IL-1ra), is a protein produced by macrophages in parallel with IL-1. The ratio of IL-1ra to IL-1 is lower in IBD mucosa than in normal mucosa. Other cytokines, including IL-10 and TGF-ß serve to down-regulate the immune response (see Fig. 83-1). Genetic mouse models in which IL-10 or TGF-ß is disrupted have colitis as their phenotype. These models demonstrate that failure of the regulatory processes that down-regulate the immune response can lead to chronic inflammation.

$CD4^+$ T cells have been divided into subsets based on the profile of the cytokines produced. IL-12, which is produced by macrophages, causes naive lymphocytes to differentiate to Th1 lymphocytes. The cytokines produced by Th1 lymphocytes, IL-2 and IFN-? are associated with cellular immune responses and increased IgG2 production. IL-4, which is produced by macrophages, causes naive lymphocytes to differentiate to Th2 lymphocytes. The cytokines produced by Th2 lymphocytes, IL-4, IL-5, IL-6, and IL-10, are associated with hypersensitivity reactions and increased IgG1 production. Crohn's disease appears to be associated with activation of Th1 lymphocytes as demonstrated by increased levels of IL-2, IL-18, IFN-?, and IgG2. [58], [59], [60] and [61] Ulcerative colitis is associated with activation of Th2 lymphocytes as demonstrated by increased IL-5, IL-10, and IgG1. [58] These findings indicate that Crohn's disease is characterized by a dysregulated Th1 response and ulcerative colitis by a dysregulated Th2 response but they do not reveal the mechanisms involved in the induction of these dysregulated responses. Genetically based animal models of colitis are also characterized by dysregulated Th1 and Th2 responses. In these animal models it is clear that an abnormal immune response is mounted against normal bacterial flora raising the possibility that the antigenic trigger for the dysregulated immune response in IBD is the normal flora. The evidence for dysregulation of the Th1 and Th2 systems in IBD is quite strong. A number of therapeutic strategies that target these pathways are sure to be tested. [62]

The major cell source of the proinflammatory cytokines is almost certainly activated macrophages in the lamina propria, although other cell types may make important contributions. Cytokines, particularly IL-1, TNF-a, and IFN-?, stimulate epithelial, endothelial, and mesenchymal cells and activate immune cells. [63] They also are involved in the regulation of wound healing and fibrosis. [64] Cytokines may be useful as markers of disease activity, especially tissue IL-1 levels in ulcerative colitis and serum IL-2 receptor levels in Crohn's disease. [65], [66]

The demonstrated efficacy of infliximab, a chimeric antibody to TNF-a, in Crohn's disease supports the key role of cytokines in IBD pathogenesis. [67] TNF-a is produced in Th1-mediated immune responses and the effectiveness of infliximab in Crohn's disease supports the suggestion that Th1 lymphocytes are important in mediating immune activation in Crohn's disease. The success of infliximab will certainly lead to other attempts to treat Crohn's disease by modulating Th1 cytokines.

Genetically Based Animal Models of Colitis

A number of genetically based animal models of IBD have been developed. [49], [68], [69] In most cases, these models were developed to study the effects of specific genetic perturbations of the immune system rather than as models of colitis. A surprising product of these manipulations of the immune system was the discovery that many genetic abnormalities in the systemic immune system have as their only phenotype the development of colitis. Furthermore, the development of colitis in these genetic models requires the presence of intestinal bacteria. The genetic defects that have colitis as their phenotype in animals with conventional bacterial flora have no phenotype in germ-free animals.

These genetically based models of colitis fall into three groups. One group of models includes three cytokine knockouts. Mice with disruption of the genes for IL-2, IL-10, and TGF-ß $_1$ all develop colitis. [70], [71] and [72] The second group of models includes mice or rats with T-cell defects. Among these are mice with defects in the T-cell receptor and transgenic rats into which the human *HLA-B27* gene has been introduced. [73], [74] In the *HLA-B27* transgenic rats, colitis is not the only phenotype; these animals also have gastritis and arthritis. An especially interesting model is severe combined immunodeficiency disease (SCID) mice repleted with CD45RBhi $CD4^+$ T cells. [49] SCID mice lack all $CD4^+$ cells yet develop no colitis; however, when these mice are repleted with a subset of $CD4^+$ cells, they develop colitis. The third group of models includes mice lacking Ga_{12}, a subunit of a G-protein. [75] Whether T cells are the important target in this genetic model is not clear.

It is possible that all these models relate to T cells and their regulation by cytokines. One could imagine a model in which immune activation in the intestinal lamina propria in response to lumenal bacteria is kept in check by a population of regulatory T cells through the secretion of down-regulatory cytokines, such as IL-10 or TGF-ß (see Fig. 83-1). Defects in that T-cell population or in its ability to produce down-regulatory cytokines would remove the brake on immune activation induced by bacterial stimulation of other T-cell populations. The interaction of T cells and cytokines was demonstrated further in studies with the CD45RBhi $CD4^+$ T- cell repleted SCID mice. Administration of a monoclonal antibody against IFN-? to these animals completely eliminated the colonic inflammation, and administration of a monoclonal antibody against TNF-a reduced it significantly. These studies raise the possibility that the intestinal damage caused by unrestrained T-cell activation is mediated by IFN-? and TNF-a.

In general the animal models that most resemble Crohn's disease are those associated with abnormalities in Th1 cytokines. Colitis induced by rectal administration of trinitrobenzene sulfonic acid has a histological picture consistent with Crohn's disease. There are high levels of IFN-? in the colons of mice receiving trinitrobenzene sulfonic acid and the colitis can be abrogated with antibodies to IL-12. [76] Both these findings are consistent with inflammation mediated by Th1 lymphocytes. There is an especially intriguing spontaneous mouse model, called the SAMP1/Yit mouse, that closely resembles Crohn's disease. [77] Adoptive transfer studies demonstrate that $CD4^+$ T cells that produce a Th1-like profile of cytokines mediate intestinal inflammation in this model. In contrast animal models that most resemble ulcerative colitis are those associated with elevated levels of Th2 cytokines. A histological picture similar to ulcerative colitis develops in mice with a mutant T-cell receptor alpha gene. [73] Analysis of spontaneous cytokine expression by mesenteric lymph node cells showed a decrease in IL-2 and an increase in IL-4 in the TCRa(-/-) mice with colitis. The decrease in IL-2 and increase in IL-4 is consistent with a Th2 mediated response.

Two of the major themes in the pathogenesis of IBD have been the importance of the lumenal bacteria driving the immune response and the importance of Th1 and Th2 T cells in mediating the immune response. These two themes were combined in a study of a spontaneously occurring model of colitis in the C3H-HeJBir mice. [78] Adoptive transfer of bacterial antigen–activated $CD4^+$ T cells from colitic C3H/HeJBir mice to SCID mice induced colitis. This study demonstrated that T cells reactive with conventional antigens of the enteric flora can mediate IBD.

Although most animal models of colitis involve defects in lymphocyte regulation, enhanced Th1 activity and chronic colitis occur in mice with a cell type–specific disruption of the *Stat-3* gene in neutrophils and macrophages. [79] *Stat-3* codes for a protein involved in the signal transduction of the receptor binding of a number of cytokines and growth factors including IL-6, IL-10, and granulocyte colony-stimulating factor. There are other genetic models of intestinal inflammation based on defects in epithelial cells. N-cadherin is a protein involved in the attachment of adjacent epithelial cells. Mice expressing a dominant-negative N-cadherin along their crypt-villus axis develop IBD. [80] Keratin 8 is a cytoskeletal molecule in epithelial cells. Mice in which the keratin 8 gene is disrupted develop crypt abnormalities and inflammation of the lamina propria and submucosa. [81]

The finding that a wide variety of defects in the systemic immune system and epithelial cells results in colitis is informative in defining the regulation of the mucosal immune system. It is less informative in defining the defects in immune regulation or epithelial cells that may be involved in the mediation of human IBD. The

pathogenesis of IBD may involve one or more of these regulatory systems or, more likely, may involve other regulatory systems.

Inflammation

IBD is considered a disorder of chronic inflammation, a view reinforced by the large numbers of lymphocytes and histiocytes in the diseased mucosa and submucosa. Ulcerative colitis and Crohn's disease also have histological characteristics of acute inflammation, including an intense infiltration of the mucosa and submucosa with neutrophils. Large numbers of neutrophils leave the bloodstream and enter the inflamed mucosa and submucosa of the bowel. [82] Some neutrophils migrate across the epithelium into the lumen and are passed in the stool, and others are destroyed in the inflamed tissue before they have a chance to migrate into the lumen. This constant flux of neutrophils is mediated by the expression of adhesion molecules on circulating neutrophils and vascular endothelial cells, which allow the neutrophils to bind to the endothelium before migrating into the tissue. Both the expression of adhesion molecules and the migration of neutrophils are regulated by inflammatory cytokines (IL-8 and TNF) and lipid mediators of inflammation, including platelet activating factor and leukotriene B_4 (LTB_4). Neutrophil activation in the inflamed gut results in the release of granule-bound proteases and the production of superoxide and other reactive oxygen species (see Fig. 83-1). [83] These products of activated neutrophils probably play a role in the destruction of epithelial cells in IBD.

Some of the functional and macroscopic changes seen in IBD, including mucosal hyperemia and edema, are typical of changes seen in any inflammatory state, no matter which organ system is involved. These changes are the products of soluble mediators released in the process of inflammation. Mediators cause tissue edema by increasing vascular (postcapillary venule) permeability to albumin and other macromolecules; hyperemia results from mediators that induce vasodilation. Progress has been made in characterizing the soluble mediators of inflammation and their role in the amplification of the immune response in IBD. There are two reasons for defining their pathogenic role. First, the soluble mediators appear to be largely responsible for the clinical and histological changes seen in the disease. Second, the drugs that have proved to be beneficial for ulcerative colitis and Crohn's disease appear to exert their therapeutic effects by blocking the synthesis of those mediators. Corticosteroids down-regulate the expression of proinflammatory cytokines and block the migration of neutrophils out of the bloodstream. 5-aminosalicylate (5-ASA) appears to act farther downstream; it is an antioxidant and thus may act to block the tissue damage induced by reactive oxygen species. Corticosteroids and sulfasalazine block the synthesis of prostaglandins and leukotrienes in vivo and in vitro. [84], [85] Until the etiologic agents of ulcerative colitis and Crohn's disease are identified, it is likely that advances in medical therapy will be in the area of regulation of the synthesis of soluble mediators of inflammation.

Almost all mammalian cells, including intestinal epithelium and cells associated with inflammatory events (e.g., mast cells, macrophages, platelets), produce prostaglandins. Prostaglandins, particularly those of the E series, have biologic properties that are proinflammatory, including enhanced vascular permeability, vasodilation, and production of pain. Prostaglandin levels are elevated in mucosa and serum in IBD and correlate with disease activity. [86] There are two cyclooxygenases that synthesize prostaglandins. Cyclooxygenase-1 (COX-1) is a constitutive enzyme, whereas COX-2 is induced by proinflammatory cytokines. COX-2 is not expressed in colonic epithelial cells in the absence of inflammation but is expressed in epithelial cells from areas of inflammation in ulcerative colitis. [87] Nonsteroidal antiinflammatory drugs (NSAIDs), which are potent inhibitors of prostaglandin production through both pathways, reduce inflammation in chronic inflammatory diseases, such as rheumatoid arthritis, but are not effective in IBD. Small trials of indomethacin administered orally and rectally revealed no improvement in ulcerative colitis; there was even some suggestion that indomethacin causes clinical deterioration despite decreasing prostaglandin production. [88], [89] Prostaglandin E_2 (PGE_2) produced through COX-2 appears to be important in gastrointestinal wound healing. [90] The negative effects of NSAIDs in IBD may be related to impaired wound healing.

IBD mucosa contains large amounts of LTB_4, [91] a neutrophil chemotactic factor. [92] The concentrations of LTB_4 are markedly higher in rectal dialysates from patients with ulcerative colitis than from controls. [93] In vitro studies indicate that LTB_4 is the major neutrophil chemotactic agent in IBD mucosa. [92] Despite the strong evidence that LTB_4 is the major neutrophil chemotactic factor in IBD, clinical trials of inhibitors of LTB_4 synthesis in ulcerative colitis have failed to demonstrate clinical efficacy. [94] Dietary supplementation with fish oil containing eicosapentaenoic acid, which reduces LTB_4 production by acting as a competitive substrate for 5-lipoxygenase, results in significant but modest improvements in ulcerative colitis symptoms. [95]

Nitric oxide is found in large amounts in IBD mucosa, and inducible nitric oxide synthase is expressed in epithelial cells in patients with IBD but not in epithelial cells from healthy persons. [96] Nitric oxide may contribute to vasodilation and mucosal edema. Whether the net effect of nitric oxide in IBD is to promote tissue injury or tissue repair is not clear.

The inflammatory response can be viewed as a component of the larger process of wound healing. The inflamed mucosa in IBD is characterized by the activation not only of typical inflammation-associated genes, such as COX-2 and IL-8, but also by the activation of genes associated with wound healing including growth factors and metalloproteinases. [97], [98]

ULCERATIVE COLITIS: CLINICAL FINDINGS AND NATURAL HISTORY

Symptoms and Physical Findings

The dominant symptom in ulcerative colitis is diarrhea, which is usually, but not always, associated with blood in the stool. [99] Bowel movements are frequent but small in volume as a result of irritability of the inflamed rectum. [100] If inflammation is confined to the rectum, blood is seen only on the surface of the stool, but if inflammation is more extensive, blood is mixed in with the stool. Other symptoms include fever and pain, which may be in either the lower quadrant or the rectum. Many patients also experience weight loss. The patient's symptoms are a function, in part, of the extent of the disease. Systemic symptoms—fever, malaise, and weight loss—are more common if all or most of the colon is involved. Fatigue and malaise are major problems and may have a greater effect than diarrhea on the patient's ability to function. If disease is confined to the rectum, the patient may complain only of urgency and tenesmus with or without blood and diarrhea. Although bloody diarrhea is considered the dominant symptom in ulcerative colitis, urgency and fecal incontinence bother many patients more. Few patients will volunteer complaints of incontinence but, if pressed, will reveal that incontinence and the fear of incontinence are the most limiting aspects of their disease. The inflamed rectum loses its ability to relax and distend when stool enters; as a result, the entry of small amounts of stool into the rectum results in the urge to defecate immediately. Some patients, especially older adults, complain of constipation rather than diarrhea. In these patients, rectal spasm may prevent the passage of stool.

In ulcerative colitis of mild to moderate severity, the physical findings are minimal. There may be tenderness over the affected area of the colon, and rectal examination may reveal tenderness or blood on the glove. In severe disease, the patient is more likely to be febrile and tachycardic. Prolonged episodes of severe disease lead to muscle wasting, edema, and other signs of malnutrition. Chronic blood loss results in pallor. Children with severe disease may have retarded growth and development.

Classification by Severity

Assessment of disease severity in ulcerative colitis is important for clinical management and in the design of drug trials. Truelove and Witts [101] devised a system for dividing ulcerative colitis patients into those with mild, moderately severe, and severe disease based on their symptoms, physical findings, and laboratory values (Table 83-1). This classification is useful and helps the clinician to predict both the anatomic extent of the patient's disease and the range of likely outcomes based on the presentation. This classification also helps the clinician to judge how vigorously the patient needs to be treated.

Severe
Diarrhea: six or more bowel movements per day, with blood
Fever: mean evening temperature higher than 37.5° C., or higher than 37.7° C on at least 2 of 4 days at any time of day
Tachycardia: mean pulse rate higher than 90 beats/min
Anemia: hemoglobin of 75 mg/dl, or less compared with normal values, allowing for recent transfusions
Sedimentation rate: more than 30 mm/h
Mild
Mild diarrhea: fewer than four bowel movements per day, with only small amounts of blood
No fever
No tachycardia
Mild anemia
Sedimentation rate: below 30 mm/h
Moderately severe
Intermediate between mild and severe

Adapted from Truelove SS, Witts LJ. Cortisone in ulcerative colitis: final report on a therapeutic trial. BMJ 1955;2:1041.

TABLE 83-1 Truelove and Witts' Classification of Ulcerative Colitis

Edwards and Truelove [102] found that in 54% of their ulcerative colitis patients, the first attack was mild. Mild disease usually is associated with inflammation confined to the rectum and sigmoid, but it can occur even in the presence of pancolonic involvement. Diarrhea and rectal bleeding are usually the only complaints in mild disease. Patients with diarrhea but without rectal bleeding may be misdiagnosed as having irritable bowel syndrome, and those with rectal bleeding but without diarrhea may be misdiagnosed as bleeding from hemorrhoids. Mild disease, especially if confined to the rectum, frequently is associated with a normal physical examination.

Most patients with ulcerative proctitis have disease of mild severity. Ulcerative proctitis should not be viewed as a separate entity but rather as part of the spectrum of ulcerative colitis. Although most patients who present with proctitis continue to have disease confined to the rectum, there is extension to involve the more proximal colon in 15% of cases with follow-up of 10 years, [103] with extension to the hepatic flexure in 7%.

In 27% of patients with ulcerative colitis, the first attack is of moderate severity. These patients typically present with five or six bloody bowel movements per day. Abdominal pain, which is uncommon in mild disease, may be present. Patients are fatigued and may have a low-grade fever. In moderate disease, there is often some degree of abdominal tenderness, but bowel sounds are usually normal. Initial attacks of moderate severity may be misdiagnosed as infectious colitis.

Edwards and Truelove [102] found that 19% of their patients with ulcerative colitis presented with severe disease. In addition to having frequent episodes of bloody diarrhea, a patient with severe colitis is likely to be weak and may even be bedridden. If the attack is sustained, the patient develops weight loss, marked anemia, and hypoalbuminemia. Fever, tachycardia, and postural hypotension indicate severe disease. Some abdominal tenderness is common even in mild disease, but significant abdominal tenderness, particularly with rebound over the colon, suggests severe disease. Markedly hypoactive or absent bowel sounds, indicating that the colon has lost all motility, is also a sign of severe disease. Protuberance of the abdomen, reflecting atony and dilation of the colon, raises the possibility of toxic megacolon.

Initial Attack

The initial attack of ulcerative colitis may be fulminant, with bloody diarrhea present from the beginning; more commonly, the disease begins indolently, with nonbloody diarrhea progressing to bloody diarrhea. [104] Symptoms often gradually worsen over the course of a few weeks. Ulcerative colitis can present initially with any extent of anatomic involvement, from disease confined to the rectum to pancolitis, and with any degree of activity, from mild diarrhea to toxic megacolon. At initial presentation, colitis extending to the cecum is seen in only about 20% of patients. Limited colitis is more common, with 75% of patients having no disease proximal to the sigmoid. [105] The greater the anatomic extent of disease at the first presentation, the more severe the symptoms are likely to be. [104] Extraintestinal manifestations, most commonly arthralgias and mild arthritis, are present in fewer than 10% of patients at initial presentation. In mild ulcerative colitis, the rectal mucosa is erythematous and edematous on endoscopic examination. In more severe disease, edema is more marked, the mucosa bleeds spontaneously, and there is an extensive purulent exudate. In the most severe cases, frank ulceration is seen.

The outcome of the first attack of ulcerative colitis can be predicted on the basis of the extent of the disease and the severity of the symptoms. [105] More than 90% of patients with mild disease go into remission after the first attack. Among patients with more severe disease, a significant number require colectomy, and a few worsen and die; death occurs primarily in patients who present with toxic megacolon.

Clinical Evaluation

Management of ulcerative colitis involves repeated evaluation of the patient's clinical condition over time. These evaluations are required at the initial presentation, at the beginning of each subsequent attack to define the severity of the attack, and at multiple points during each attack to determine whether the clinical picture is worsening or improving. The extent of the clinical evaluation is guided by the presentation—the milder the presentation, the less extensive the evaluation that is required. The most important parts of the clinical evaluation are the history and physical examination. The frequency and severity of diarrhea is a good first guide to the severity of the disease. More than six bowel movements per day usually is associated with severe disease; however, comparison of the number of bowel movements with the patient's normal bowel habits may be more informative than the absolute number. Moreover, the number of bowel movements is a more useful indication of severity in acute rather than in chronic disease. Systemic signs and symptoms (fever, hypotension, tachycardia) are markers for the presence of severe disease and demand a more extensive evaluation. Coexistence of irritable bowel syndrome with ulcerative colitis makes evaluation of the patient's symptoms more difficult.

Endoscopic examination is required at the initial presentation to help establish the diagnosis and to define the extent of disease. [106] The initial endoscopic examination is usually a colonoscopy unless severe disease is present; in patients with severe disease, proctoscopy or sigmoidoscopy carries less risk. Endoscopic examination may be useful at presentation of subsequent attacks if there is any question as to whether the attack is a recrudescence of ulcerative colitis. In patients with a past history of ulcerative colitis who develop diarrhea, proctoscopy is a useful method for distinguishing recurrences of ulcerative colitis from infectious diarrhea. Endoscopic examination also may be indicated if a recurrence of ulcerative colitis is more severe than earlier occurrences, suggesting anatomic extension of the disease. For patients with long-standing disease, colonoscopic examination is used for surveillance for dysplasia. [107], [108] Some clinicians routinely follow the course of ulcerative colitis exacerbations by repeated proctoscopy or sigmoidoscopy, with the thought that the endoscopic picture provides important information about the response to drug therapy; however, there is no definitive evidence that the information yielded is any more useful than that gained by asking the patient about the severity of symptoms.

One of the most important functions of clinical evaluation in exacerbations of ulcerative colitis is the identification of patients who are progressing to toxic megacolon. [109], [110] Fever, leukocytosis, and marked abdominal tenderness all indicate severe disease. Plain films of the abdomen demonstrate whether there is colonic dilation consistent with toxic megacolon. Colonic dilation usually occurs without obvious abdominal distention.

Laboratory studies also play a role in the clinical evaluation of ulcerative colitis. If the history and physical examination indicate mild disease activity, laboratory studies are likely to add little to the evaluation. They are most useful in confirming a clinical impression of severe disease and in following the clinical course of a severe exacerbation. The laboratory studies most likely to reflect disease activity are hemoglobin level, leukocyte count, and erythrocyte sedimentation rate. An elevated leukocyte count, however, may be difficult to interpret in patients treated with corticosteroids. Other biochemical abnormalities may be present. Hypoalbuminemia is a sign of severe disease, usually of long duration. Electrolyte disorders, particularly hypokalemia, are seen with severe diarrhea.

The role of microbial agents in inducing exacerbations of IBD has been a somewhat controversial area. Intercurrent intestinal infections, either bacterial (*Clostridium difficile*, *Campylobacter* species, *Yersinia* species) or viral (cytomegalovirus), have been implicated in inducing exacerbations. Although one study reported high levels of recovery of *C. difficile* toxin from patients with symptomatic relapses, [111] other studies have reported low rates. [112] Cytomegalovirus causes gastrointestinal infection and is fairly common in the immunosuppressed host (e.g., patients taking immunomodulators). The typical presentation is abdominal pain and watery diarrhea. Endoscopic biopsies show cytomegalovirus vasculitis within the lamina propria. Typical intranuclear inclusions are seen in endothelial cells. If cytomegalovirus is found, corticosteroids and immunomodulators should be tapered and stopped if possible.

Natural History

Most commonly, ulcerative colitis follows a chronic intermittent course, marked by long periods of quiescence interspersed with acute attacks lasting weeks to months

(Table 83-2). [102] In a few patients diagnosed as having ulcerative colitis, there is a single acute attack of colitis with no recurrences. It seems likely, however, that in some of these patients the diagnosis is not correct. A small but significant percentage of patients suffer a chronic, continuous course with persistent symptoms and no complete remission. Patients who require continuous treatment with steroids to maintain remission are also part of this group. In drug studies in acute ulcerative colitis, the placebo remission rates average 10% and the placebo improvement rates average 30%. [113]

CLINICAL COURSE	NUMBER OF PATIENTS	PERCENTAGE
Acute fulminating	20	8.0
Chronic intermittent	161	64.4
Chronic continuous	18	7.2
One attack only	45	18.0
Total colectomy in first attack	2	0.8
Died in first attack of other causes	1	0.4
Unknown	2	0.8
Total	249	100.0

Adapted from ref. 102.

TABLE 83-2 Clinical Course of Ulcerative Colitis

The risk of relapse after the first attack of ulcerative colitis is a function of the patient's age at the time of the first attack. [105] Older patients are more likely than younger ones to go long periods without relapse. For patients younger than 50 years, the median time for relapse after the first attack is 2 to 3 years. Neither the severity of the first attack nor the extent of colonic involvement at the time of diagnosis has any effect on the frequency of recurrence; however, the severity and extent of the disease at initial presentation affect the likelihood and timing of subsequent colectomy. [105] For those with severe disease at first presentation, the rate of colectomy reaches 50% by 2 years after the initial attack. For patients with pancolitis, the rate of colectomy reaches 50% after 5 years. In contrast, fewer than 10% of patients who present with mild disease or with proctitis alone have undergone colectomy after 10 years. As with the risk of relapse, the cumulative resection rate among patients with ulcerative colitis is inversely proportional to age. Those in the oldest age group are the least likely to suffer relapse and the least likely to undergo resection.

No systematic studies of triggering events for relapses in ulcerative colitis have been undertaken. Among the potential triggering events that have been suggested are:

noncompliance with maintenance medications
NSAIDs
antibiotics given for problems unrelated to ulcerative colitis (e.g., sinusitis, pneumonia)
intercurrent colonic infections (e.g., *Shigella*, *Salmonella*, *C. difficile*)
cessation of smoking.

Antibiotics and intercurrent infections are thought to induce relapses by altering the colonic flora and thus altering the relationship between the mucosal immune system and the antigens in the lumen. For patients with ulcerative colitis with enteric infections, it is important to determine if the patient has only an enteric infection or an enteric infection plus a flare of ulcerative colitis.

Those who present initially with proctitis have a more benign course than those who present initially with more extensive disease. These patients are likely to respond to local therapy and thus are spared the side effects of systemic therapy. However, those who present with proctitis are at risk for extension of their disease; they have a 50% chance of extension and a 12% chance of colectomy within 25 years. Those with disease proximal to the sigmoid have a 9% chance of progression to pancolitis and 23% chance of colectomy. In contrast those who start with pancolitis have a 40% chance of colectomy. [114]

Mortality in patients with ulcerative colitis has decreased dramatically and now life expectancy is similar to that of the general population. Ulcerative colitis has a significant lifelong impact on a patient's quality of life. Most patients lead productive lives with 90% employed after treatment of the initial attack. [115] More patients have impairment of social and emotional functioning as manifested by worry about the need for surgery, the social problems associated with an ostomy, side effects of medication, and the uncertain course of the disease. These concerns are likely to be reduced if the physician is actively involved in teaching the patient about the natural history of the disease.

CROHN'S DISEASE: CLINICAL FINDINGS AND NATURAL HISTORY

Disease Location

Crohn's disease is a more complex and difficult clinical entity than ulcerative colitis, partially because of the diversity of anatomic locations in which it is detected and the effects of this diversity on presentation, clinical course, and therapeutic options. There are three major patterns of disease distribution: disease present in the ileum and cecum, a pattern seen in 40% of patients at presentation; disease confined to the small intestine, a pattern seen in 30% of patients at presentation; and disease confined to the colon, a pattern seen in 25% of patients at presentation. [116] Among those with colonic disease, most have pancolitis, but about a third have segmental disease. Much less commonly, Crohn's disease involves more proximal parts of the gastrointestinal tract—the mouth, the tongue, the esophagus, the stomach, and the duodenum.

Symptoms

The predominant symptoms in Crohn's disease are diarrhea, abdominal pain, and weight loss. [116] , [117] Any of these three symptoms may be most prominent, in contrast to ulcerative colitis, in which diarrhea is almost universally the most prominent complaint. The initial presentation of Crohn's disease may not be dramatic. Patients may complain for months or years of vague abdominal pain and intermittent diarrhea before the diagnosis of Crohn's disease is considered.

Diarrhea occurs in almost all persons with Crohn's disease, but the pattern varies with the anatomic location of the disease (Table 83-3). [116] In patients with colonic disease, especially with rectal involvement, diarrhea may be of small volume and associated with urgency and tenesmus. Inflammation in the rectum causes a loss of distensibility. The entry of even a small amount of stool into a nondistensible rectum causes an immediate and urgent need to defecate. Prolonged rectal inflammation and scarring in the rectum can leave it so rigid and nondistensible that the patient is incontinent. In disease confined to the small intestine, stools are of larger volume and are not associated with urgency or tenesmus. Patients with severe involvement of the terminal ileum and those who have had surgical resections of the terminal ileum may have elements of bile salt diarrhea or, in more severe cases, frank steatorrhea. [118] Strictures in the small intestine may lead to bacterial overgrowth with deconjugation of bile salts and fat malabsorption. If diarrhea is a product of fat malabsorption, the timing and severity of the diarrhea are functions of the pattern of fat ingestion. Finally, internal fistulae are common in Crohn's disease and can lead to diarrhea either by colonization of the small bowel with bacteria, as in enterocolonic fistulae, or through bypass of large segments of absorptive epithelium, as in enteroenteric or enterocolonic fistulae.

	DISEASE LOCATION (%)		
CLINICAL FEATURE	Ileitis (%)	Ileocolitis (%)	Colitis (%)
Diarrhea	~100	~100	~100
Abdominal pain	65	62	55
Bleeding	22	10	46
Weight loss	12	19	22
Perianal disease	14	38	36
Internal fistulae	17	34	16
Intestinal obstruction	35	44	17
Megacolon	0	2	11
Arthritis	4	4	16
Spondylitis	1	2	5

Adapted from ref. 116.

TABLE 83-3 Frequency of Clinical Features in Crohn's Disease

The location and pattern of pain in Crohn's disease often correlate with disease location. One common pain pattern is cramping right lower quadrant pain in patients with ileocolonic disease. [119] This pain usually occurs after eating and probably is related to partial intermittent obstruction of a narrowed intestinal lumen. Pain is caused by stretching of the wall in the dilated segment proximal to the obstruction and by powerful contractions of the small bowel musculature attempting to push intestinal contents through the obstructed segment. Abdominal distention, nausea, and vomiting may accompany pain in this circumstance. A second common pain pattern in Crohn's disease is visceral pain resulting from inflammation of the serosa, as seen in transmural Crohn's disease. The pathophysiologic basis of abdominal pain in nonobstructed or stable patients with Crohn's disease is less clear. Crohn's disease is associated with an increased risk of depression; pain management is more difficult in patients with depression.

Weight loss of some degree occurs in most patients with Crohn's disease, irrespective of anatomic location. Loss of more than 20% of body weight is less common, occurring in 10% to 20% of affected persons. Some weight loss is a product of malabsorption, but weight loss usually is a product of diminished intake. Patients, especially those with small bowel disease, may avoid food because eating brings on pain or diarrhea or, more commonly, because they are anorectic. Colonic disease is associated with a high incidence of rectal bleeding and perianal involvement but a low incidence of internal fistulae and obstruction (see Table 83-3). In contrast, disease confined to the small intestine is associated with a lower incidence of bleeding and perianal involvement but a higher incidence of obstruction.

Constitutional symptoms contribute significantly to the overall morbidity in Crohn's disease. Fever and chills often accompany Crohn's disease activity; a low-grade fever may be the patient's first warning sign of a flare of activity. Fatigue and malaise can reduce the ability to function markedly and may impact work performance negatively. Although they are more likely to complain of pain and diarrhea, patients may be more bothered by fatigue. In patients in whom fatigue is the dominant symptom, coexisting depression should be considered. Induction of remission by drugs or surgery invariably is associated with increased energy and a sense of well being.

The initial presentation of Crohn's disease can usually be characterized as either obstructing or fistulizing. Obstructive disease is the result of inflammation narrowing the intestinal lumen and obstructing the flow of intestinal contents. Over time, fibrosis and thickening of the intestinal wall also contribute to obstruction. Crampy abdominal pain, nausea, vomiting, and diarrhea are the major symptoms associated with obstructing disease. Fistulizing disease occurs when the inflammatory process extends completely through the intestinal wall. The escape of bacteria through these defects in the wall can result in abscesses. Extension of the inflammatory process into adjacent organs results in fistulae (enteroenteric, enterocutaneous, enterovesicular, etc.). Fistulizing disease can present with fever, leukocytosis, and evidence of fistulization (e.g., enterocutaneous fistulae). Obstruction and fistulization are not mutually exclusive, in fact the enteric end of a fistula is often found immediately upstream from a stricture.

Distinguishing the obstructive and fistulizing forms of Crohn's disease is important because the distinction affects therapy. For example, steroids are likely to be effective in obstructing disease but not in fistulizing disease. The distinction between obstructing and fistulizing disease also affects prognosis. Relapses in patients with a past history of fistulae are more likely to be characterized by fistulization and relapses in those with a past history of obstruction are more likely to be characterized by obstruction. Obstruction and fistulization are discussed in detail later in the chapter.

Physical and Laboratory Findings

Physical findings in Crohn's disease also vary with the distribution and severity of the disease. [120] When the disease is active, the patient looks pale, weak, and chronically ill. Aphthous ulcers in the mouth are common in active Crohn's disease. These are shallow ulcers covered by a grayish exudate and surrounded by an erythematous margin. These painful lesions can be found on the lips, gingiva, or buccal mucosa. Occasionally, oral lesions are the presenting problem (see Chapter 50). [121] Long-standing severe disease with malnutrition results in temporal and interosseous wasting. The abdomen may be tender, typically over the area of disease activity. Thickened bowel loops, thickened mesentery, or an abscess may cause a sense of fullness or a mass, often in the right lower quadrant. Rebound tenderness (most often referred) is often present. Fistulous openings, induration, redness, or tenderness near the anus suggest the presence of perianal Crohn's disease. The mucosa at the anal verge may appear purplish because of vascular engorgement. Fissures in the anal canal can occur; bleeding from these fissures can be confused with active colitis.

Laboratory findings in Crohn's disease are largely nonspecific. [122] The peripheral blood count may reveal anemia resulting from chronic disease, blood loss, or nutritional deficiencies (iron, folate, or vitamin B_{12}). A modestly elevated leukocyte count is indicative of active Crohn's disease, but a marked elevation suggests the presence of an abscess or other suppurative complication. Thrombocytosis may occur with active disease. Bleeding or thrombotic complications may occur secondary to thrombocytosis. The erythrocyte sedimentation rate has been used to monitor disease activity in Crohn's disease, and it tends to be higher in colonic disease than ileal disease. Hypoalbuminemia is a good indication of disease severity and malnutrition. Ileal disease or resection of more than 100 cm of ileum results in a diminished serum vitamin B_{12} level because of malabsorption.

Crohn's disease may rarely involve the stomach and duodenum. In these cases, more distal involvement, particularly of the ileum, is usually also present. Gastroduodenal Crohn's disease may present with epigastric pain similar to that of duodenal ulcer. Upper gastrointestinal series may show ulceration and narrowing of the antrum and duodenum (Fig. 83-2). Aphthous ulcers and linear ulcers in the gastric antrum may be seen on endoscopy. [123] Therapy is the same as for ileal Crohn's disease, although a trial of H_2-receptor antagonists is useful for those with ulcers. Duodenal Crohn's disease often leads to stenosis, obstruction, and postprandial vomiting. [124] Gastrojejunostomy without resection may be required to bypass the obstruction but should be avoided if possible.

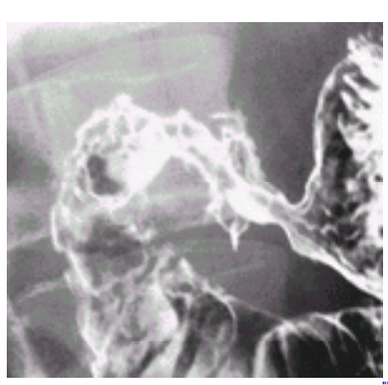

FIGURE 83-2. (See Color Fig. 83-2.) Crohn's disease involving the antrum and duodenum. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

Disease Activity Indices

It is often difficult to assess the course of disease activity in Crohn's disease, especially when fatigue or pain are prominent symptoms. Usually, a number of different disease manifestations (increased number of bowel movements, diminished general sense of well being, fistula formation, weight loss, decreased hematocrit, endoscopic appearance, histology, increased sedimentation rate, increased orosomucoid levels) occur and do not necessarily improve or worsen in parallel. This problem becomes particularly obvious in comparing medical therapies or in assessing the efficacy of new medical therapies. Many markers of disease activity can be monitored, and it is difficult to interpret studies in which some markers improve but others do not. Several attempts have been made to establish numerical systems for evaluating disease severity and response to therapy in Crohn's disease. [125], [126], [127], [128] and [129] The National Cooperative Crohn's Disease Study devised the Crohn's Disease Activity Index (CDAI) for evaluating response to therapy in that study. [124] The CDAI was calculated by assigning numerical scores to the number of diarrheal stools, abdominal pain, general well being, systemic manifestations, use of antidiarrheal agents, presence of abdominal mass, hematocrit, and body weight. Other groups have proposed and used alternative numerical indices to quantify Crohn's activity, but these indices, although promoted as simplified, are not especially easy to use, nor are they necessarily more valid than the CDAI. [122], [127], [128] and [129] Thus, the use of indices is not suggested for following disease activity in patients except in research studies.

Natural History

Crohn's disease, like ulcerative colitis, is a relapsing and remitting disease. About 30% of placebo-treated patients with Crohn's disease of mild to moderate activity go into remission within 4 months. [130], [131] Conversely, in a group of patients with Crohn's disease in remission and not receiving any type of therapy, about 70% remained in remission at 1 year and 50% at 2 years. Within 10 years of diagnosis, 60% of patients with Crohn's disease have surgery. Postoperatively, endoscopic signs of recurrence are seen in 70% of patients at 1 year and recurrence of symptoms is seen in 40% to 50% within 4 years. [132], [133] Of those patients who have one surgical resection for Crohn's disease, 45% eventually require reoperation. The younger the age at onset, the more likely the patient will have a complicated course and the more likely surgery will be required. [134]

Life expectancy in Crohn's disease patients is near normal but there is some increased mortality, especially among those who develop the disease before age 20. The increase in mortality is most prominent in the early years of the disease. [135], [136] Despite the low mortality, Crohn's disease can have a large negative impact on quality of life. Patients have problems with employment, social activities, and interpersonal relationships. The quality of life in Crohn's disease is, on average, less good than in ulcerative colitis. The majority of patients are employable; however half of them need to modify their employment as a result of their disease. These modifications include decreased hours, leaves of absence, and career changes. [137] Patients with Crohn's disease have concerns about the uncertain nature of their disease, decreased energy, medication side effects, and surgery, especially fear of an ostomy. [138] The high incidence of depression as a co-morbid condition also diminishes the quality of life for patients with Crohn's disease.

EXTRAINTESTINAL MANIFESTATIONS

Although ulcerative colitis and Crohn's disease primarily involve the bowel, both are associated with manifestations in other organ systems. For some patients, especially those with sclerosing cholangitis or ankylosing spondylitis, the extraintestinal manifestations may be more troublesome than the bowel disease. The extraintestinal manifestations can be divided into two major groups: those in which the clinical activity follows the activity of the bowel disease and those in which the clinical activity is unrelated to the clinical activity of the bowel disease. Most extraintestinal manifestations occur more commonly with ulcerative colitis or Crohn's colitis than with Crohn's disease confined to the small intestine.

Peripheral Arthritis

The most common extraintestinal manifestation of IBD is arthritis (Table 83-4). [139], [140] and [141] The two arthritic complications of IBD are colitic or peripheral arthritis and ankylosing spondylitis or axial arthritis. Colitic arthritis is a migratory arthritis or arthralgia that affects the large joints of the knees, hips, ankles, wrists, and elbows, in that frequency. Usually, fewer than six joints are involved and small joints of the hands are typically not involved. [139] Colitic arthritis is more common with Crohn's colitis than ulcerative colitis and is uncommon with Crohn's disease confined to the small intestine. Most arthritic flares last only a few weeks, though recurrences are common with renewed disease flares. Deformity with radiologic changes occurs in fewer than 25% of cases. The joint pain, swelling, and stiffness parallel the course of the bowel disease. Serology for rheumatoid factor is negative in these patients. Colitic arthritis, uveitis, and erythema nodosum often are seen together. Successful treatment of the intestinal inflammation results in improvement in the arthritis. Acetaminophen can be used for joint discomfort. NSAIDs are generally avoided as they often may induce a flare or worsening of the disease. Often, treatment of bowel disease with corticosteroids results in a dramatic improvement in the associated arthritis.

ORGAN INVOLVED	NUMBER OF PATIENTS	PERCENTAGE OF TOTAL
Joint	53	26
Polyarthralgia	27	13
Spine	8	4
Extremities	18	9
Skin	39	19
Erythema nodosum	9	4
Pyoderma gangrenosum	10	5
Other	20	10
Mouth	8	4
Eye	9	13
Total patients	202	

Adapted from Greenstein AJ, Janowitz HD, Sachar DB. Extra-intestinal complications of Crohn's disease and ulcerative colitis: Study of 700 patients. Medicine (Baltimore) 1979;55:401.

TABLE 83-4 Extraintestinal Manifestations in 202 Patients With Ulcerative Colitis

Axial Arthritis

Ankylosing spondylitis is seen in patients with IBD, but its activity does not follow that of the bowel disease, and treatment of the bowel disease does not affect the spondylitis. [140], [141] Ankylosing spondylitis presents with morning stiffness, low back pain, and stooped posture. Radiography of the spine in ankylosing spondylitis shows squaring of the vertebrae and straightening of the spine. Syndesmophytes appear along the lateral and anterior portions of the intervertebral discs, giving the picture of "bamboo spine" (Fig. 83-3).

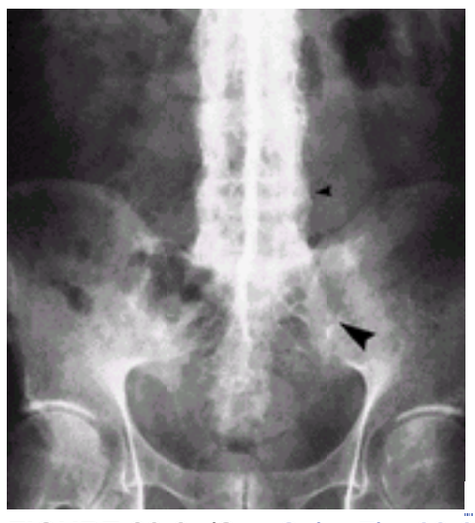

FIGURE 83-3. (See Color Fig. 83-3.) Sacroiliitis and ankylosing spondylitis demonstrating obliteration of the sacroiliac joints (*large arrow*) and bamboo spine with syndesmophytes (*small arrow*). (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

Patients with ulcerative colitis have a 30-fold increase in the incidence of ankylosing spondylitis compared with the general population. Even though there is no increased incidence of HLA-B27 in ulcerative colitis, 80% of those with both ulcerative colitis and ankylosing spondylitis are B27-positive. [142] IBD-associated ankylosing spondylitis occurs frequently in females (perhaps up to 40%), whereas, ankylosing spondylitis unassociated with intestinal disease occurs at a male-to-female ratio of nearly 9:1. In contrast to colitic arthritis, which is episodic and usually nondeforming, ankylosing spondylitis can be relentlessly progressive and crippling. The results of medical management of ankylosing spondylitis in IBD are poor. Physical therapy plays a major role in maintaining function. Pain management is a problem, and narcotic addiction is common. NSAIDs, though they reduce inflammation and pain in spondylitis and in colitic arthritis, do not halt the progression of the disease and may exacerbate intestinal inflammation. Medical treatment of IBD and colectomy are not helpful in managing the ankylosing

spondylitis. A report of successful treatment with infliximab suggests a hopeful, possibly effective option. [143]

Sacroiliitis, inflammation of the joint between the sacrum and the ileum, can be seen in conjunction with ankylosing spondylitis but more often is seen alone. Over 20% of IBD patients, most of whom have no back pain, have radiographs consistent with sacroiliitis. [144], [149] Some will present with low back pain which can be slowly progressive, not related to intestinal inflammation. Radiographs of the pelvis reveal blurring of the margins of the sacroiliac joints, with patchy sclerosis.

Osteoporosis

Osteoporosis or osteopenia occurs in as many as half of patients with IBD. [145] In Crohn's disease osteopenia may be present at diagnosis. Numerous factors may contribute including malnutrition, malabsorption, and smoking. Persistent inflammation and steroid use may be the most important factors. Tumor necrosis factor enhances bone resorption. Although osteopenia may occur independent of steroid use, steroids can still worsen the problem. In ulcerative colitis, the loss of bone density occurs more directly as a result of steroid use. Patients with primary sclerosing cholangitis, particularly with longer standing disease, can have more severe osteoporosis. The therapeutic options of estrogen replacement therapy, raloxifene, and calcitonin have expanded considerably, with the addition of bisphosphonates-alendronate (Fosamax) and risedronate (Actonel) which can augment bone density considerably. Screening patients with dual photon absorptiometry is recommended. [146] Guidelines for the management of osteoporosis in IBD have been developed by the British Society of Gastroenterology. [147]

Renal Complications

The incidence of kidney stones in IBD patients, with estimates ranging between 1% to 25%, is several times that in the general population. [148] Calcium oxalate stones are seen in patients with small intestine Crohn's disease. Ileal resections or ileal disease can lead to fat malabsorption, and unabsorbed fatty acids bind calcium in the lumen. In healthy persons, oxalate in the lumen is bound to calcium. Calcium oxalate is poorly soluble and poorly absorbed. If calcium is bound to malabsorbed fatty acids, however, oxalate combines with sodium to form sodium oxalate, which is soluble and is absorbed in the colon. [149] The development of calcium oxalate stones in Crohn's disease requires an intact colon to absorb oxalate; patients with ileostomies do not develop calcium oxalate stones. However, the presence of an ileostomy predisposes to other kinds of kidney stones, particularly urate stones, because of the diminished urine volumes in patients with significant ileostomy volume losses. [150] Measurement of 24-hour urinary oxalate, uric acid, and citrate may be useful to guide therapy. Dietary restriction of oxalate and citrate supplementation can help prevent recurrence of oxalate stones.

Other urinary tract complications are associated specifically with Crohn's disease. Inflammation from the bowel extending into the retroperitoneal space can occlude the ureters, leading to obstruction and hydronephrosis. [151] This process most commonly extends from the area of the cecum and terminal ileum and involves the right ureter. Also in Crohn's disease, fistulae can form between an inflamed bowel and the urinary bladder, leading to urinary tract infections. [152] Amyloidosis is seen in patients with Crohn's disease and has caused death from renal insufficiency. [153] Renal amyloidosis does not respond to conventional medical therapy for Crohn's disease and, for the most part, does not improve after the inflamed section of intestine is resected.

Dermatological Manifestations (see Chapter 49)

The two common dermal complications of IBD are pyoderma gangrenosum and erythema nodosum. Pyoderma gangrenosum is seen with colitis and ileocolitis and is associated with extensive long-standing disease. The incidence of pyoderma in ulcerative colitis is low (1%–5%), and it is even lower in Crohn's disease; however, 36% to 50% of all patients with pyoderma gangrenosum have IBD. [154], [155] The typical lesion is a discrete ulcer with a necrotic base, usually found on the lower extremities (Fig. 83-4) but may occur elsewhere, particularly the face. Lesions often occur at the site of previous injury (pathergy). The ulcers may drain purulent material, but the drainage is sterile on culture. These ulcers can become large and deep, with destruction of surrounding soft tissues. Activity of pyoderma gangrenosum may or may not follow the activity of the bowel disease, but these lesions almost always develop during a bout of acute colitis. Usually, the lesions resolve with control of the colitis by use of oral corticosteroids. Alternatively, corticosteroids can be injected directly into the skin lesions. Therapies with Periactin, dapsone, and azathioprine have been reported, with cyclosporine increasingly recommended. [156], [157] and [158] Infliximab has also been reported to be dramatically effective in some cases. [159] Topical tacrolimus has also been effective. [160]

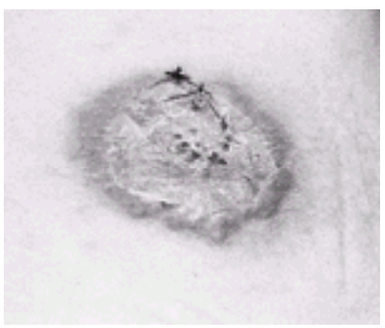

FIGURE 83-4. (See Color Fig. 83-4.) Pyoderma gangrenosum (with stitches after biopsy). (Courtesy of Ira Kodner, M.D., St. Louis, MO.)

Erythema nodosum is seen particularly in association with Crohn's disease in children. [161] The lesions are raised, tender nodules, usually found over the anterior surface of the tibia. Activity of erythema nodosum follows the activity of the bowel disease and responds to treatment of the bowel disease. Most patients have only a single episode of erythema nodosum. Sweet syndrome (acute febrile neutrophilic dermatosis), which has some similarity to erythema nodosum, is another dermatologic condition associated with IBD. [162]

Eye Complications

The ocular complications of IBD are uveitis and episcleritis. [163] Uveitis (iritis) is an inflammatory lesion of the anterior chamber, presenting with blurred vision, headache, eye pain, photophobia, and conjunctival injection. Diagnosis is made by slit-lamp examination, which demonstrates perilimbic edema and cells in the anterior chamber. Scarring and blindness can be prevented with local therapy of corticosteroids and atropine or other agents that dilate the pupil. Episcleritis is a less serious problem, presenting with burning eyes and scleral injection. Treatment with topical steroids is effective.

Thromboembolic Complications

Thromboembolic complications, with arterial as well as venous events, occur in about 1% to 2% of patients with ulcerative colitis and Crohn's disease. [164] Activation of clotting factors and thrombocytosis are common. [165] In patients with active disease, procoagulant factors tend to be increased and fibrolytic factors are decreased, potentially generating a hypercoaguable state. In both Crohn's disease and ulcerative colitis increased levels of factor V, factor VIII, and fibrinogen and decreased levels of antithrombin III have been demonstrated. [166], [167] Deep vein thrombosis and pulmonary emboli affect patients with active disease. Thromboembolic events in the eye and in intracranial vessels also have been described. [166], [168]

Hepatobiliary Complications

The hepatic complications of IBD include fatty liver, pericholangitis, chronic active hepatitis, and cirrhosis. The biliary tract complications are sclerosing cholangitis and gallstones. Cholesterol gallstones occur in patients with ileal disease or ileal resections owing to malabsorption of bile salts and the resultant decrease in the size of the bile salt pool. Pericholangitis is the most common hepatic complication of IBD, with prevalences as high as 50% to 80% reported. [169], [170] Patients with pericholangitis are usually asymptomatic. Elevations of alkaline phosphatase are seen frequently; elevations of bilirubin are less common. Histologically, there is inflammation of the portal tracts, with lymphocyte and eosinophil infiltrates and degenerative changes of the bile ductules. [171] In more advanced cases, progressive

fibrosis can develop. The relationship between pericholangitis and primary sclerosing cholangitis is uncertain. Some studies have suggested that pericholangitis may be a part of the spectrum of sclerosing cholangitis, and the histological manifestations of sclerosing cholangitis and pericholangitis on liver biopsy may be indistinguishable. [172] Patients diagnosed on liver biopsy as having pericholangitis may need to undergo endoscopic retrograde cholangiopancreatography (ERCP) to rule out sclerosing cholangitis. There is no effective therapy for pericholangitis.

Sclerosing cholangitis is a chronic cholestatic liver disease marked by fibrosing inflammation of the intrahepatic and extrahepatic bile ducts; it occurs in 1% to 4% of patients with ulcerative colitis and with lower frequency in Crohn's disease. [173], [174] Most patients with sclerosing cholangitis have IBD. The prevalence of IBD is so high in patients with sclerosing cholangitis that colonoscopy should be performed even on patients who are without intestinal symptoms. Endoscopic and histological evidence of IBD are commonly found in patients with sclerosing cholangitis without intestinal symptoms. Patients with ulcerative colitis and sclerosing cholangitis have a higher risk of colon cancer than those with ulcerative colitis alone and should undergo more intensive screening. [175] Sclerosing cholangitis can affect intrahepatic bile ducts, extrahepatic bile ducts, or both. In patients with IBD and sclerosing cholangitis, usually both intrahepatic and extrahepatic ducts are affected. Early in the disease, liver biopsy shows enlargement of the portal tracts, with edema and bile duct proliferation. Later, biopsies show extension of fibrosis out of the portal space, eventually leading to cirrhosis.

Bile duct strictures are the major clinical problem in sclerosing cholangitis; strictures can occur in both intrahepatic and extrahepatic bile ducts (Fig. 83-5). Patients may be asymptomatic until the disease is far advanced and then present with fever, right upper quadrant pain, and jaundice. More often, sclerosing cholangitis is first recognized in the evaluation of abnormal laboratory studies: elevated alkaline phosphatase, bilirubin, and transaminases. The differential diagnosis of sclerosing cholangitis includes biliary tumors and common duct gallstones; diagnosis is made by ERCP or transhepatic cholangiography. Sclerosing cholangitis progresses to cirrhosis, hepatic failure, and death in 5 to 10 years. [176] Clinical features at presentation associated with a poorer prognosis include hepatomegaly, splenomegaly, elevated serum alkaline phosphatase, advanced histological stage, and age. [177] Endoscopic balloon dilation or stenting of extrahepatic strictures may be palliative. Cholestyramine, a bile salt–binding resin, may help to relieve pruritus. Colectomy and medical therapy of the bowel disease are without benefit in the management of sclerosing cholangitis. Ursodeoxycholic acid can decrease alkaline phosphatase and transaminases but does not alter histological progression or influence the occurrence of clinically important events such as time to transplant or death. [178] Liver transplantation has been used with success, and sclerosing cholangitis is now one of the most common indications for liver transplantation in adults. Cholangiocarcinoma develops in 10% to 15% of IBD patients with long-standing sclerosing cholangitis. Differentiating cholangiocarcinoma from the benign strictures of sclerosing cholangitis is difficult. ERCP with brushing or cholangioscopy with biopsy may help to make this distinction.

FIGURE 83-5. (See Color Fig. 83-5.) Sclerosing cholangitis with segmental strictures (*arrows*) in common bile duct. There are also strictures in the intrahepatic bile ducts. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

ENDOSCOPY AND RADIOLOGY

Endoscopy

Endoscopy and radiology serve complementary roles in the diagnosis and management of patients with IBD. Endoscopy serves to diagnose IBD, to distinguish between ulcerative colitis and Crohn's disease, to determine the extent and severity of disease, to assess response to treatment, and to screen for dysplasia. Due to its ability to visualize the mucosa directly and obtain biopsies, endoscopy has become established as the primary diagnostic tool. Differentiating between acute infectious colitis and an initial flare of ulcerative colitis or Crohn's disease by endoscopic findings may be difficult. The endoscopic findings in several varieties of infectious colitis are indistinguishable from those seen in IBD. [106], [179], [180] *Yersinia, Campylobacter, Shigella*, and cytomegalovirus infections all can have endoscopic pictures that do not differ from that of ulcerative colitis. They show mucosal edema, granularity, erythema, and easy friability. [179], [180] Pseudomembranous colitis in its later stages may produce diffuse mucosal changes like those of ulcerative colitis, but the characteristic raised, yellow, plaquelike pseudomembranes should be diagnostic. Amebiasis is the infectious disease most likely to result in discrete, punched-out ulcers with normal intervening mucosa, as seen in Crohn's colitis. [181], [182]

Over time, endoscopic diagnosis of IBD is more certain, and the distinction between ulcerative colitis and Crohn's disease can be made in most cases. The important distinguishing features of Crohn's disease include rectal sparing; areas of active disease interspersed with normal mucosa; sharp, punched-out ulcerations surrounded by normal mucosa; and "cobblestoning," or nodular mucosa often intersected by crossing linear ulcerations. The characteristic findings of ulcerative colitis are rectal involvement with a confluent distribution, friable mucosa, a blurred vascular pattern, and ulcerations in areas of active inflammation.

Endoscopy is more sensitive than radiographic contrast studies in defining the margins of inflammation. Contrast studies may be unable to detect endoscopically evident disease in up to 20% of cases. [183] Comparisons of double-contrast radiography with colonoscopy show that radiography underestimates the extent of colonic involvement in two thirds of cases. [184] However, as many as 65% of patients will have active disease in the ileum beyond the reach of the colonoscope, with 13% having a normal terminal ileum. [185] Colonoscopic assessment of the extent of inflammation is frequently part of preoperative evaluations, especially for Crohn's disease, to guide the surgeon on the extent of resection so that all the severely involved tissue is removed.

Endoscopy may be useful in following the course of disease. Arbitrary values of 0 to 4+ can be assigned to erythema, granularity, edema, and friability to yield an endoscopic severity score. These scores correlate reasonably well with the patient's clinical condition as assessed by symptoms in ulcerative colitis. In many cases, however, the colonoscopic picture is either much better or much worse than would be predicted by the patient's symptoms. In these cases, the usual approach is to base management decisions primarily on the patient's clinical presentation rather than on the endoscopic findings.

Similarly, in Crohn's disease, there can be a discrepancy between clinical symptoms and endoscopic findings. A randomized trial was performed to determine whether colonoscopy is useful in deciding how long to continue corticosteroid therapy in patients with Crohn's colitis. In this study, all patients were placed on steroids. In one group, steroid tapering was begun as soon as the patients achieved clinical remission; in the other group, endoscopy was used to guide the timing of the start of steroid withdrawal. [186] The two groups did equally well; suggesting that colonoscopy was not required to time the start of tapering a steroid. Endoscopy can be useful in guiding therapy; colonoscopy in the year after an ileal resection may help identify patients with progressive disease that might benefit from more aggressive therapy. Among patients free of lesions or with mild disease at 1 year, 80% remained quiescent for 3 years. In contrast, 92% of patients with more severe endoscopic findings worsened clinically during that time period. [187]

Strictures and mass lesions in the colon in patients with IBD are suspicious for carcinoma, especially if the disease has been present for longer than 10 years and particularly in ulcerative colitis. These lesions should be investigated colonoscopically and biopsied. [188] Carcinoma causing a stricture usually appears as an eccentric, friable mass. Malignant strictures have these endoscopic features: rigidity, nodules within the stricture or at its margins, an eccentric lumen, and a shelflike

margin. Sometimes in ulcerative colitis the bulk of the tumor may be submucosal, even though the tumor starts in the epithelium. In these cases, epithelial biopsies may be negative. Strictures that appear malignant by endoscopy probably should be resected regardless of the histological appearance on biopsy. Colonoscopy for dysplasia surveillance is discussed later in this chapter.

Colonoscopy also has therapeutic potential in IBD. Polypectomy is approached much the same way in patients with IBD as in the general population; however, pseudopolyps are not premalignant and do not need to be resected. However, adenomatous polyps removed endoscopically in patients with IBD raise the issue of dysplasia-associated lesions or masses (DALM). These lesions may be less of a herald of carcinoma than previously thought and some patients may be followed with conservative colonic screening after endoscopic removal of the lesion. [189] Biopsies around the polyp base and tattooing the area for later surveillance may be helpful. Pseudopolyps can be resected colonoscopically if they are bleeding or causing obstruction. Colonoscopic dilation of strictures is sometimes possible; dilation is performed with a balloon passed through the colonoscope. The best results have been achieved with short, un-inflamed strictures. Longer strictures are less likely to be dilated successfully and more likely to perforate during attempted dilation.

For the most part, the precautions to be taken in performing colonoscopy in patients with IBD are similar to those for the general population; however, some considerations are peculiar to IBD. [190] Perforation during colonoscopy is more likely; the bowel wall is inflamed, and its integrity may be weakened by penetrating ulcers and fistulae, particularly in Crohn's disease. Known or suspected severe inflammation with deep ulceration is a relative contraindication to colonoscopy. Similarly, toxic megacolon is an absolute contraindication to colonoscopy because of the paper-thin colonic wall. Scarring and fibrosis, often present in IBD, make the colon less elastic and distensible and more susceptible to perforation from pressure. Great care should be taken not to use excessive force *or air* in advancing the scope. The risk of perforation in severe ulcerative colitis makes it unwise to attempt a complete colonoscopy. Examination of the rectum with a flexible sigmoidoscope should yield enough information to confirm the diagnosis.

The earliest colonoscopic manifestation of ulcerative colitis is loss of the fine vascular pattern seen in the normal rectal mucosa and the development of diffuse erythema (Color Fig. 83-1). [106], [190] Erythema usually is accompanied by mucosal edema, which is manifested endoscopically by blunting of the rectal valves, loss of normal vasculature, and development of granular-appearing mucosa. In the normal colon, finely branching vessels are seen easily through the mucosa. In mild colitis, the vessels still are seen but may be tortuous *and less sharply defined*. As colitis becomes more severe, edema blunts the haustral pattern, and vessels cannot be seen at all (Color Fig. 83-2). Normal rectal mucosa is flat and smooth and reflects light in large patches. If the mucosa becomes edematous, small mounds of swollen tissue surround the crypts, creating an uneven surface (Color Fig. 83-3). Light reflects off the uneven surface as numerous small spots rather than large patches. This pattern of reflection is termed granularity (Table 83-5). Inflammation is associated with the presence of collections of yellowish exudate on the mucosa. The exudate is called mucopus, and the more active the disease, the larger the area of mucosa it covers. The inflamed mucosa bleeds easily if it is touched with the endoscope or a cotton swab. This easy bleeding is termed friability. In more severe disease, the mucosa bleeds spontaneously and small ulcerations appear. In the presence of inflammation and spasm, the rectum does not distend easily when insufflated with air. This is the equivalent of the loss of distensibility seen on radiography. An important aspect of the endoscopic findings in ulcerative colitis is their distribution. Inflammation begins in the rectum, extends proximally a certain distance, and then stops; all the mucosa proximal to that point is normal, and all the mucosa distal to it is abnormal (see Fig. 83-6). However, a modest amount of cecal inflammation may be noted in ulcerative colitis, particularly around the appendiceal orifice. Cecal inflammation does not indicate a skip lesion or suggest Crohn's disease as a more appropriate diagnosis. [191] As ulcerative colitis heals, residual mucosal edema, absent vasculature, and postinflammatory polyps in a tubular ahaustral lumen occur (Color Fig. 83-4). The ileum is uninvolved in ulcerative colitis but in 10% to 20% of patients with pancolitis, mild ileal inflammation with edema may be seen. This finding in ulcerative colitis is termed "backwash ileitis" but ulcers are not usually present and the endoscopic appearance is not particularly characteristic of Crohn's disease.

FIGURE 83-6. (See Color Fig. 83-6.) Colectomy specimen from patient with ulcerative colitis demonstrates sharp demarcation in the midtransverse colon between involved and uninvolved mucosa. (Courtesy of Ira Kodner, M.D., St. Louis, MO.)

LESION	ULCERATIVE COLITIS	CROHN'S DISEASE
Inflammation		
Distribution		
Colon		
Contiguous	+++	+
Symmetric	+++	+
Rectum	+++	+
Friability	+++	+
Topography		
Granularity	+++	+
Cobblestoned	+	+++
Ulceration		
Location		
Overt colitis	+++	+
Ileum	0	++++
Discrete lesion	+	+++
Features		
Size>1 cm	+	+++
Deep	+	++
Linear	+	+++
Aphthoid	0	++++
Bridging	+	++

[a] Specificity index range: 0 (not seen) to ++++ (diagnostic).
From ref. 214.

TABLE 83-5 Colonoscopic Mucosal Features and Their Diagnostic Specificity in Inflammatory Bowel Disease [a]

The earliest endoscopic manifestation of Crohn's disease is the aphthous ulcer (Color Fig. 83-5), a small discrete ulcer a few millimeters in diameter surrounded by a thin red halo of edematous tissue (see Table 83-5). [192], [193] Aphthous ulcers are usually multiple, and the intervening mucosa is normal. They can grow to form large stellate or linear ulcers. Ulcers may be rounded (Color Fig. 83-6) or long and serpiginous. Longitudinal and transverse ulcers may intersect to form a grid with intervening cobblestone-like areas of non- ulcerated mucosa. In Crohn's disease, large, deep penetrating ulcers can be surrounded by areas of normal-appearing mucosa. The diffuse mucosal irregularities of erythema, edema, and granularity, which are prominent in ulcerative colitis, occur less commonly and later in the course of Crohn's disease. The rectum may or may not be involved in Crohn's disease. Areas of involvement typically are interspersed with normal "skip areas." In a prospective endoscopic study of patients with Crohn's disease who were about to start a course of prednisone, the incidence of endoscopic findings was as follows: superficial erosions, 93%; deep erosions, 74%; mucosal edema, 48%; erythema, 44%; pseudopolyps, 41%; aphthous ulcers, 10%; ulcerated stenosis, 8%; and non-ulcerated stenosis, 2%. [194]

Radiology

Contrast radiology and computed tomography (CT) are both important in the management of IBD. They are more central to the management of Crohn's disease. In ulcerative colitis endoscopy is the preeminent diagnostic tool. The decision as to which radiologic modality to use is made on the basis of the question being addressed as they are complementary techniques with their own strengths and weaknesses. Air-contrast radiography is better for assessing mucosal detail, colonic distensibility, and the presence of strictures. In an acutely ill patient with Crohn's disease, a CT scan is often the preferred initial diagnostic test to answer urgent

clinical questions concerning the presence of abscesses or possible obstruction. Both techniques may be useful to detect the presence of fistulae. Radiography without contrast serves two functions in IBD: the assessment of colonic dilation in suspected toxic megacolon (Fig. 83-7) and the initial approach to the definition of the location and completeness of intestinal obstruction.

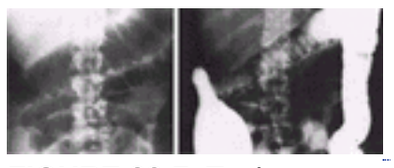

FIGURE 83-7. Toxic megacolon. **A:** Plain film radiograph shows colonic dilation. **B:** Contrast radiography reveals large ulcerations. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

For the most part, air-contrast or double-contrast studies are preferred to single-contrast studies because they give much better definition of fine mucosal detail and even can assess the depth of ulcerations. [195] Single-contrast studies are more useful, however, for the definition of fistulae, strictures, and tumors. [196] Although contrast studies are usually well tolerated, some precautions should be observed. Barium enema is contraindicated in patients with severe colitis (either ulcerative colitis or Crohn's colitis) because injection of air and barium into the inflamed colon may precipitate toxic megacolon. Even patients with significant diarrhea usually need laxatives to clean the bowel in preparation for barium enema. Harsh, irritative laxatives should be avoided, but osmotic laxatives such as citrate of magnesia usually are tolerated. Oral contrast agents should be used with caution when obstruction may be present.

In both ulcerative colitis and Crohn's disease, radiographic findings may not correlate well with disease activity. [197] In particular, asymptomatic patients may have markedly abnormal radiographic studies because of fibrosis of the bowel wall with resultant loss of distensibility. A peroral pneumocolon, a regular air-contrast, small bowel follow-through in which air is insufflated in the colon, can assist in determining distensibility of the terminal ileum and can improve the visualization of the ileum. Air-contrast studies are frequently unhelpful in distinguishing whether a narrowed segment is due to fibrosis or inflammation. Air-contrast studies are also often inadequate to determine disease activity or to monitor the patient's response to medical management over relatively short periods. The patient's clinical response or endoscopic findings are more useful for these purposes.

In early ulcerative colitis, the barium enema may be normal. The most sensitive radiologic finding in ulcerative colitis is limited distensibility of the involved segment, resulting in a narrowed, shortened, tubular form of the lumen. The haustral markings disappear, and the normally tortuous appearance of the colon is straightened. Air-contrast examination reveals a fine granular appearance to the mucosa, with a slightly irregular surface (Fig. 83-8). In more severe disease, the granularity becomes coarser and eventually nodular. These changes may be more obvious on evacuation films. In addition to the granularity, small discrete ulcers surrounded by mounds of inflamed tissue may be seen. In more severe disease, ulcers penetrate through the mucosa and can be seen in profile as small, collar-button collections of barium extending beyond the colonic lumen (Fig. 83-9). Inflammatory polyps, either nodular or filiform, may be seen in either active or quiescent disease (Fig. 83-10). Inflammation and fibrosis in the rectum increase the presacral space, which is the distance from the sacrum to the rectal lumen, on lateral views. An enlarged presacral space (>2 cm) is consistent with severe rectal inflammation. In chronic ulcerative colitis, the colon becomes shortened and loses its haustral markings (Fig. 83-11 and Fig. 83-12). In cases of burnt-out ulcerative colitis, markedly abnormal radiographs may be seen in patients who are asymptomatic.

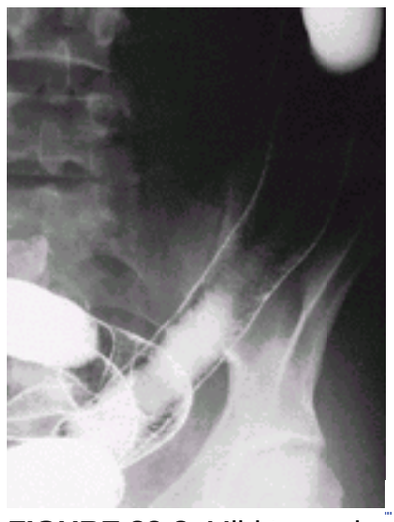

FIGURE 83-8. Mild to moderate, left-sided, ulcerative colitis shows granularity of mucosa without ulceration. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

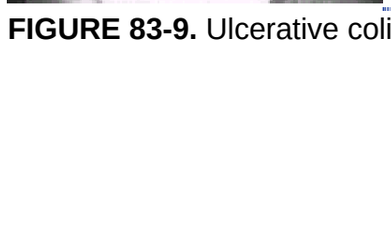

FIGURE 83-9. Ulcerative colitis with collar-button ulcerations seen in profile (*arrowhead*). (Courtesy of Dennis Balfe, M.D., St. Louis, MO.

FIGURE 83-10. Inflammatory polyps are present in an otherwise featureless colon. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

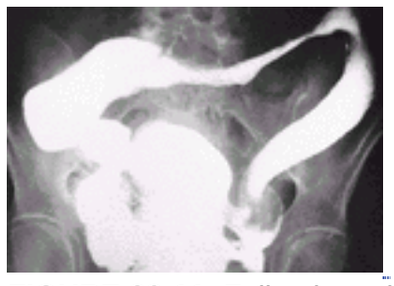

FIGURE 83-11. Full-column barium enema in a patient with ulcerative colitis shows shortening of the colon and loss of haustral markings. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

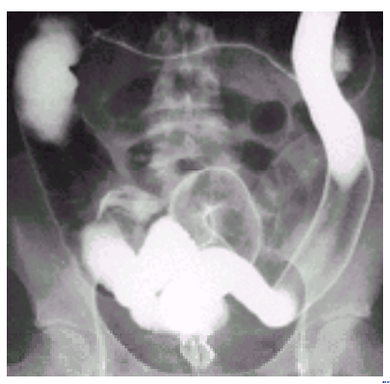

FIGURE 83-12. Air-contrast study of chronic ulcerative colitis with ahaustral colon. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

In 15% to 20% of ulcerative colitis patients with pancolitis, the terminal ileum appears abnormal radiographically. The ileocecal valve is deformed and open, the terminal ileum is dilated, and the mucosa is irregular, but there are no ulcerations. This process, termed backwash ileitis, should be differentiated from terminal ileal involvement in Crohn's disease, which is marked by lumenal narrowing, wall thickening, ulceration, and fistula formation. [198]

Carcinoma in ulcerative colitis can appear as a mass protruding into the lumen, as it may in those without ulcerative colitis. However, ulcerative colitis carcinoma is more often an infiltrating process that appears on radiography either as a flattened, rigid area of bowel wall or, in more advanced cases, as a stricture. Radiologic methods fail to detect cancer in patients with ulcerative colitis about 15% of the time. [196], [199] Benign strictures may be indistinguishable from malignancy radiographically (Fig. 83-13). The presence of a flattened, rigid area or a stricture necessitates colonoscopy with biopsy. Because of the infiltrating nature of carcinoma in ulcerative colitis, colonic biopsies may be negative even if malignancy is present. If the radiograph is suggestive of carcinoma, surgery may be necessary despite a negative endoscopic biopsy.

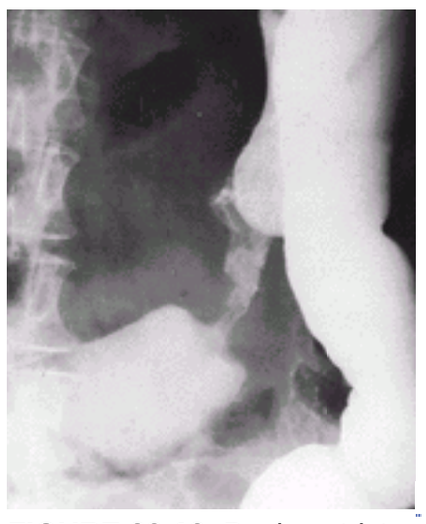

FIGURE 83-13. Benign stricture in transverse colon of patient with chronic ulcerative colitis. This lesion is radiographically indistinguishable from a malignancy. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

The earliest form of Crohn's disease detectable by air-contrast barium enema is marked by the presence of aphthous ulcers, which appear as small discrete collections of barium surrounded by radiolucent halos of inflammatory infiltrate (Fig. 83-14). These small ulcers are usually multiple, and the intervening mucosa is normal. Aphthous ulcers are not, however, unique to Crohn's disease; they are seen in amebiasis, Behçet syndrome, shigellosis, and other conditions. As Crohn's disease becomes more severe, the aphthous ulcers enlarge, deepen, and connect with one another to form stellate and linear ulcers; the intervening mucosa develops a nodular appearance on radiograph, a process termed cobblestoning.

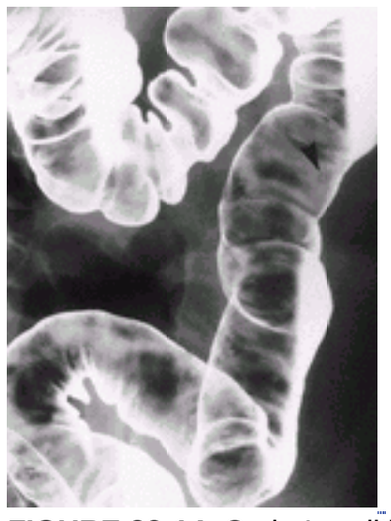

FIGURE 83-14. Crohn’s colitis with numerous aphthous ulcers (*arrowhead*). (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

Progressive deepening of ulcers can lead to formation of abscesses or fistulization. Contrast studies are far more likely than endoscopic studies to identify fistulae. Transmural inflammation and fibrosis lead to limited distensibility, with decreased lumenal diameter. If transmural inflammation and fibrosis are circumferential, the result is stricture formation and possible obstruction. Like fistulae, strictures are appreciated more easily on radiographic studies than by endoscopy. Long areas of circumferential inflammation and fibrosis result in long areas of lumenal narrowing: if this process occurs in the ileum, the result is the “string sign.” Transmural inflammation and fibrosis result in thickening of the bowel wall, and as a result there are wide gaps between the barium-filled lumens of loops of inflamed small bowel (Fig. 83-15 and Fig. 83-16). The distribution of radiographic abnormalities can support the diagnosis of Crohn’s disease. Findings typical of diffuse colitis may be consistent with either Crohn’s disease or ulcerative colitis, but distal sparing is more consistent with Crohn’s disease. Thickening of the bowel wall and retraction and thickening of the mesentery make the bowel rigid, stiff, and immobile on fluoroscopy. Small bowel Crohn’s disease can be evaluated by small bowel follow-through or by enteroclysis. Comparison of the two techniques in patients with known small bowel Crohn’s disease suggested that small bowel follow-through gives better mucosal detail and identifies more fistulae and, thus, is the procedure of choice. [200]

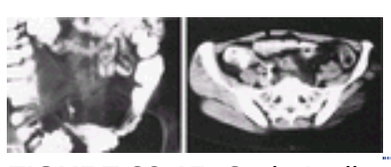

FIGURE 83-15. Crohn’s disease of the terminal ileum (*arrows*) on small bowel follow-through (**left**) and computed tomography (CT) scan (**right**) in the same patient. Thickening of the intestinal wall is easily appreciated on the CT scan. On small bowel follow-through, wall thickening is indicated by the separation of the columns of barium. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

FIGURE 83-16. In Crohn’s ileitis with a long segment of involved ileum, the barium columns are widely separated as a result of wall thickening. (Courtesy of Dennis Balfe, M.D., St. Louis, MO.)

The CT scan, with its ability to assess wall thickening, inflammatory reaction in adjacent fat, and extralumenal complications allows for a rapid assessment of a patient with Crohn’s disease and helps guide therapy. While these issues may be considered for ulcerative colitis as well, CT is less central to the management of patients with ulcerative colitis. A CT scan may be the diagnostic test that first suggests the diagnosis of IBD in a patient who presents with abdominal pain and has signs and symptoms suggesting a structural lesion. Thickened bowel wall, which is appreciated on barium studies as separation of the columns of barium, is easily seen on CT scan (see Fig. 83-15). The presence of isolated right colon involvement, mesenteric fibrofatty proliferation, or intra-abdominal abscess are all significantly more common in Crohn’s disease, whereas submucosal fat is more common in ulcerative colitis. [201], [202] Fat surrounding bowel loops, seen in surgery as “creeping” fat, can be suggestive of Crohn’s disease, particularly of more chronic disease. Stranding in the adjacent fat indicates areas of active inflammatory disease.

CT is particularly critical in evaluating a patient for intestinal complications of Crohn’s disease, most notably fistulae, abscesses, and obstruction. While barium studies may be superior in demonstrating most fistulae, CT is better in visualizing the associated inflammatory response and thus may allow for the detection of a fistula only intermittently open and not apparent on barium study. The iodinated contrast used in CT scans is safer in the presence of an intestinal obstruction than barium. In addition, extraintestinal manifestations of Crohn’s disease (including hydronephrosis, sacroiliitis, gallstones, and renal calculi) can be identified by CT. CT also can be used to direct percutaneous drainage of abscesses.

Other imaging modalities occasionally may have a role in IBD, including radionuclide studies, ultrasonography, and magnetic resonance imaging (MRI). In IBD, there is a markedly increased migration of neutrophils into the inflamed tissue and then out into the intestinal lumen. This process can be followed by harvesting a patient’s peripheral blood neutrophils, labeling them in vitro with indium 111 (^{111}In) or technetium 99 (^{99}Tc), and then injecting them intravenously into the patient. [82], [203] The radiolabeled neutrophils are carried by the bloodstream to the bowel, where they migrate into the inflamed tissue. A gamma camera then can be used to identify the sites of inflammation. This method not only allows the definition of the anatomic extent of inflammation but also allows an assessment of the severity of the inflammatory response by defining the number of neutrophils leaving the circulation and entering the bowel wall. MRI may also have a role in defining perianal disease and associated fistulae.

PATHOLOGY

Ulcerative colitis and Crohn’s disease each have a characteristic pathological appearance, but in any given case the pathological picture may not be specific enough to distinguish between them or to differentiate them from other diseases such as infectious colitis or ischemic colitis. In both ulcerative colitis and Crohn’s disease, the pathological picture is influenced by the degree of disease activity; the pathological assessment of disease activity may or may not correlate with the clinical and endoscopic assessments.

Macroscopic features of pathological specimens in ulcerative colitis depend on the severity and duration of the disease. In severe disease, the mucosa is edematous and deep purple owing to congestion with blood (see Fig. 83-6). The distribution of the involved mucosa is also characteristic of ulcerative colitis. The inflammation begins in the rectum, extends proximally a certain distance, and then abruptly stops, with a clear demarcation between involved and uninvolved mucosa (see Fig. 83-6). In mild disease there are superficial erosions or only mild erythema, with blurring of the vascular pattern as the initial, subtle findings. In severe disease there may be large ulcers or areas where the mucosa is denuded completely. In chronic disease, the mucosa loses its normal folds and becomes flat. Inflammatory polyps or pseudopolyps may be present; rarely, these polyps may be long (filiform polyps) and occur in groups (Fig. 83-17).

FIGURE 83-17. Filiform pseudopolyps. (Courtesy of Ira Kodner, M.D., St. Louis, MO.)

Most of the pathological findings in ulcerative colitis are limited to the mucosa and submucosa (Fig. 83-18); the muscularis propria is affected only in fulminant disease. Active ulcerative colitis is marked by an intense infiltrate, with neutrophils in the mucosa and submucosa and clumps of neutrophils in crypt lumens (crypt abscesses) (Fig. 83-19). Although characteristic of active ulcerative colitis, the presence of a neutrophil infiltrate with crypt abscesses also may be seen in Crohn's disease and infectious colitis. In active ulcerative colitis, mucus depletion and mucosal edema are seen; vascular congestion with focal hemorrhage is seen in more severe cases. Ulcers, if present, are superficial, only penetrating the muscularis mucosae in severe disease. In addition to signs of acute activity, there are usually also signs of chronicity, with lymphoid aggregates, plasma cells, mast cells, and eosinophils in the lamina propria. In quiescent ulcerative colitis, the inflammatory infiltrate is less intense than in active disease, but the mucosa is usually architecturally abnormal. The number of crypts is reduced and often branched. Shortening and branching of the crypts are typical of ulcerative colitis, although they also may be seen in Crohn's disease. In quiescent disease, goblet cell mucin content is restored to normal.

FIGURE 83-18. Ulcerative colitis in the appendix showing mucosal inflammation with crypt abscesses. Note the absence of inflammation in the muscularis propria. (Courtesy of David Lacey, M.D., St. Louis, MO.)

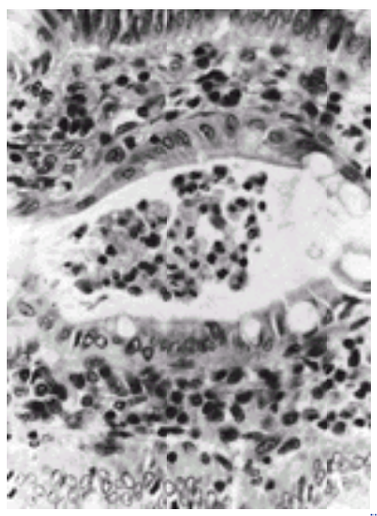

FIGURE 83-19. Crypt abscess from the specimen shown in Figure 83-18. (Courtesy of David Lacey, M.D., St. Louis, MO.)

Histological evaluation of rectal biopsy may be helpful in differentiating ulcerative colitis from acute self-limited colitis and from Crohn's disease (Table 83-6). [204] Distorted crypt architecture, mixed acute and chronic inflammation in the lamina propria, a villous mucosa, and crypt atrophy are all far more common in ulcerative colitis than in acute self-limited colitis. The best histological distinction between Crohn's colitis and ulcerative colitis is the presence of granulomas, which, in one series, was seen in 16 of 26 patients with Crohn's colitis but in only 3 of 56 patients with ulcerative colitis. Endoscopic skip lesions, or uninvolved areas, should be biopsied and may be histologically normal in Crohn's disease but histologically active in ulcerative colitis. Crypt atrophy, polymorphonuclear leukocytes in the epithelium, and surface erosions are each more common in ulcerative colitis than Crohn's colitis. Despite these distinctions, ulcerative colitis and Crohn's disease cannot be distinguished histologically in 15% to 25% of cases. [205]

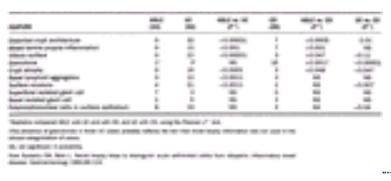

TABLE 83-6 Histological Features That Allow the Physician to Distinguish Among Acute Self-Limited Colitis (ASLC), Ulcerative Colitis (UC), and Crohn's Disease (CD)*

Gross examination of the intestine in Crohn's disease shows the bowel wall to be thickened and stiff. The mesentery is thickened and edematous and may be contracted, which fixes the intestine in one position. Adipose tissue extends from the mesentery and "creeps" over the serosal surface of the intestine. Transmural inflammation may cause loops of intestine to be matted together. All layers of the intestine are thickened, and the lumen is narrowed (Fig. 83-20). Deep linear ulcers with intervening normal mucosa give the mucosal surface the appearance of having been clawed (Fig. 83-21). The distribution of inflammation in the gross specimen may suggest the diagnosis of Crohn's disease. Colonic inflammation with rectal sparing is more consistent with Crohn's disease than with ulcerative colitis.

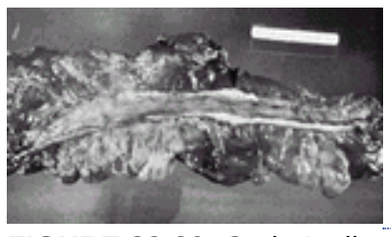

FIGURE 83-20. Crohn's disease of the small intestine showing lumenal narrowing and thickening of the intestinal wall (*arrow*). There is also a large amount of fat on the serosa. (Courtesy of Ira Kodner, M.D., St. Louis, MO.)

FIGURE 83-21. Crohn's colitis colectomy specimen shows deep, clawed linear ulcers in the transverse and descending colon. (Courtesy of Ira Kodner, M.D., St. Louis, MO.)

The earliest lesion of Crohn's disease is the aphthous ulcer. In the small intestine, aphthous ulcers typically occur over Peyer patches and in the colon over lymphoid aggregates. Aphthous ulcers can occur anywhere in the epithelium, however, even where there is no lymphoid tissue. As the disease progresses, aphthous ulcers enlarge and become stellate. Eventually, the stellate ulcers coalesce to form longitudinal and transverse linear ulcers. The remaining islands of non-ulcerated mucosa give a cobblestone appearance. Fissures develop from the base of aphthous ulcers, extending down through the muscularis to the serosa. Free perforation is uncommon because serositis induces the adherence of other bowel loops into which the fissure extends.

Transmural inflammation is the histological hallmark of Crohn's disease (Fig. 83-22). [206] Lymphoid aggregates are found in the submucosa and external to the muscularis propria. Occasional lymphoid aggregates in the muscularis propria complete the transmural pattern. Granulomas are found in most surgical resections for Crohn's disease (Fig. 83-23), but the exact percentage of positive specimens is not clear. The likelihood of finding granulomas in biopsies is a function of the number of biopsies taken, the number of sections examined, and the definition of a granuloma. [207] Granulomas occur more commonly in the submucosa than in the mucosa and therefore are found more easily in surgical specimens than in mucosal biopsies. Granulomas also can be found in lymph nodes, mesentery, peritoneum, and liver. In most cases, clinical, endoscopic, and pathological characteristics allow the diagnosis of Crohn's disease, even if no granulomas are found. Intestinal granulomas can be found in a number of infectious diseases, including tuberculosis, fungal infections, yersiniosis, and chlamydial infections. They also are seen in sarcoidosis and foreign body reactions. Granulomas in Crohn's disease resemble those in sarcoidosis and lack the caseating necrosis seen in tuberculosis. Granulomas are less commonly part of the pathological picture in ulcerative colitis. If pathological findings are used to differentiate ulcerative colitis from Crohn's colitis, the presence of granulomas suggests Crohn's colitis, but the absence of granulomas is not helpful. One pathological finding thought to be characteristic of Crohn's disease is the presence of axonal necrosis of autonomic nerves. [208]

FIGURE 83-22. Fissure in a patient with Crohn's disease. A fissure and a chronic inflammatory infiltrate are present, both of which extend into the muscularis propria. (Courtesy of David Lacey, M.D., St. Louis, MO.)

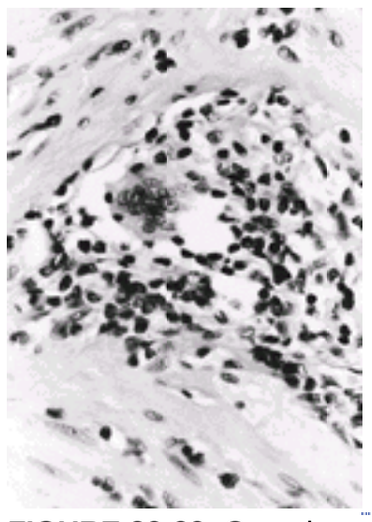

FIGURE 83-23. Granuloma in a surgical resection specimen from a patient with Crohn's disease. (Courtesy of David Lacey, M.D., St. Louis, MO.)

DIFFERENTIAL DIAGNOSIS

Crohn's Disease and Ulcerative Colitis

For many therapeutic decisions, it is not critical to know whether the patient has ulcerative colitis or Crohn's disease. In some circumstances, however, the distinction is important. For example, an ileoanal anastomosis could be recommended with much more enthusiasm if the physician were confident that the patient had ulcerative colitis rather than Crohn's colitis. Similarly, colectomy could be recommended more confidently as a curative procedure if it were certain that the patient had ulcerative colitis. As unequivocal genetic or other serologic markers do not yet exist for either disease, the diagnoses rest on clinical evaluation. If a patient appears in all other ways to have ulcerative colitis, but is found to have a feature usually seen in Crohn's disease, such as a fistula or a granuloma, it is a diagnostic dilemma to determine whether the patient should be considered to have ulcerative colitis, Crohn's disease, or indeterminate colitis. In as many as 5% of cases when ulcerative colitis is confidently diagnosed and a colectomy with an ileoanal anastomosis performed, Crohn's disease is eventually diagnosed, though at times many years later.

The clinical presentation of ulcerative colitis resembles that of Crohn's disease, especially Crohn's colitis. [209], [210] Diarrhea may be the most prominent symptom in both diseases, although it is more likely to be bloody in ulcerative colitis. The presence of pain as a prominent symptom is more suggestive of Crohn's disease, although patients with severe ulcerative colitis also may have significant pain, usually in the rectum. Systemic features such as malaise, fever, and weight loss are more common with Crohn's disease. The presence of a right lower quadrant mass and tenderness directs the diagnosis toward Crohn's disease. Certain complications are far more common in Crohn's disease than in ulcerative colitis; among these are fistulae (enteroenteric, enterocutaneous, enterovesicular, enterovaginal), intraabdominal abscesses, strictures, and malabsorption. Extensive perianal involvement with fistulae and abscesses point to Crohn's disease as the diagnosis. The patient who smokes cigarettes is more likely to have Crohn's disease and a nonsmoker, or a former smoker, is more likely to have ulcerative colitis.

The serologic markers being advanced to diagnose and differentiate ulcerative colitis and Crohn's disease may have an adjunctive role in diagnosis but are not yet proven in their utility. The marker associated with approximately 60% of patients with ulcerative colitis, perinuclear antineutrophil cytoplasmic antibodies (pANCA), is also found in approximately 10% of patients with Crohn's disease. The pANCA-positive patients with Crohn's disease appear to be a subtype with an ulcerative colitis-like left-sided, often hemorrhagic, colitis. Anti- *Saccharomyces cerevisiae* antibody (ASCA) found in approximately 60% of patients with Crohn's disease and about 10% of patients with ulcerative colitis appears highly specific for Crohn's disease if IgA and IgG are both positive. Further studies with these markers will determine their use in diagnosing and differentiating ulcerative colitis and Crohn's disease. [211], [212] and [213]

Usually, ulcerative colitis and Crohn's disease can be differentiated endoscopically (see Table 83-5). [106], [214] The anatomic distribution of the inflammatory response is suggestive of the diagnosis. In ulcerative colitis, inflammation is seen in the rectum and extends proximally for some distance; in extensive disease, inflammation extends to the cecum. Although ulcerative colitis does not involve the small intestine, a few centimeters of inflamed mucosa without ulceration may be present in the terminal ileum. If the rectum is spared or if there are areas of un-inflamed mucosa (skip areas) between areas of inflamed mucosa, Crohn's colitis is more likely. Ulcerative colitis is not only continuous along the longitudinal axis of the colon, but the degree of inflammation is also consistent and circumferentially symmetric at any level. In contrast, in Crohn's colitis, areas of normal mucosa may separate deep linear ulcers. Involvement of more than a few centimeters of the terminal ileum or ulceration in the terminal ileum is also indicative of Crohn's disease. In addition to the presence of skip areas and asymmetric involvement, other endoscopic findings also suggest Crohn's colitis. Ulcers are more likely to be large and deep and to have discrete margins in Crohn's colitis than in ulcerative colitis. Aphthous ulcers occur commonly in Crohn's disease but infrequently, if at all, in ulcerative colitis.

The pathological changes seen in Crohn's disease and ulcerative colitis are discussed elsewhere in this chapter (see Table 83-6). A major distinguishing mark in favor of Crohn's disease is the presence of transmural inflammatory changes; in ulcerative colitis, inflammation is confined to the mucosa and submucosa. The presence of noncaseating granulomas suggests Crohn's disease, but even in Crohn's disease, most patients have no granulomas on biopsy.

Despite all these differences, a small but significant number of patients with IBD cannot be assigned with confidence to one disease category or the other. [215] These patients are considered to have indeterminate colitis. As for patients who seem to fall definitively into one category or the other, it is important to keep an open mind about the diagnosis. Later developments may require that it be reconsidered. Usually these cases present as ulcerative colitis but ultimately develop signs of Crohn's disease rather than the reverse.

Inflammatory Bowel Disease Versus Other Diseases

Much has been written about the necessity for differentiating the initial episode of ileocolonic Crohn's disease from acute appendicitis; however, in practice, this distinction is seldom a major problem. [216] Patients with Crohn's disease usually give a history of diarrhea of significant duration before the episode of severe pain. More severe pain, guarding, and a more rapid course is indicative of appendicitis. A CT scan is likely to help sort out this dilemma preoperatively. If the surgeon operates and finds ileocolic Crohn's disease rather than appendicitis, a decision must be made as to whether to resect the inflamed intestine. If there is no compelling reason for resection, medical management of the condition should be attempted. Another consideration in this differential diagnosis is acute ileitis caused by *Yersinia* infection. [217] *Yersinia* infection may present with a clinical picture resembling appendicitis or the initial onset of Crohn's disease. At laparotomy, the acute *Yersinia* ileitis may be indistinguishable from Crohn's ileitis. The diagnosis of this infection is made by stool culture, and antibiotic therapy is curative. The possibility of *Yersinia* infection is another reason to avoid hasty resections for what is thought to be the acute onset of Crohn's ileitis.

The most common problem in the differential diagnosis of the recent onset of IBD is ruling out infectious colitis (see Chapter 88 and Chapter 125). [218] Infections with *Shigella,* amoeba, *Giardia, Escherichia coli* O157:H7, and *Campylobacter* organisms all cause symptoms similar to those of ulcerative colitis. Moreover, the endoscopic picture of infections with *Shigella* or *Campylobacter* organisms can be identical to that seen in ulcerative colitis. The endoscopic picture of amebiasis also may resemble that of ulcerative colitis, but in amebiasis there are usually scattered ulcers, 5 to 15 mm in diameter, covered with a yellow exudate that contains organisms. These ulcers may occur anywhere in the colon but are most common in the cecum and ascending colon. An important distinction between these infectious diseases (excepting amebiasis) and IBD is that the diarrhea in the infectious diseases tends to be limited to a period of days to a few weeks, whereas the diarrhea of IBD is typically of longer duration. Stool cultures for bacterial pathogens and serologic tests for amebiasis help to distinguish infectious diarrhea from IBD. In patients who present with prolonged diarrhea, the diagnosis of other protozoal diseases, such as giardiasis, must be considered. Giardiasis, like Crohn's disease, can present with cramping abdominal pain, weight loss, and lactose intolerance.

Sexual transmission of enteric pathogens by anal intercourse in homosexual men can lead to proctitis resembling ulcerative proctitis. [219] *Neisseria gonorrhoeae,* chlamydia organisms, herpes simplex, and *Treponema pallidum* can all cause proctitis. Chlamydial infections of the lymphogranuloma venereum immunotype can result in diffuse rectal ulceration that is difficult to distinguish from Crohn's colitis. The increased prevalence of the human immunodeficiency virus (HIV) has broadened the number of potential enteric pathogens to include *Cryptosporidium* and *Isospora* species, *Mycobacterium avium* complex, and cytomegalovirus. In patients without risk factors for HIV infection, the only diagnostic studies needed to rule out infectious colitis are routine stool cultures, examination of stool for ova and parasites (on three separate days), and sigmoidoscopy. Patients who are at risk for HIV infection require a more elaborate evaluation. Viral culture and

histological examination of biopsy specimens will identify cytomegalovirus and herpesvirus. Cytomegalovirus can complicate IBD, particularly when immunosupressive medication is being used. Serology and culture are diagnostic for chlamydial infections. Intestinal involvement in HIV infection is discussed more fully in Chapter 124.

Intestinal tuberculosis resembles Crohn's disease both clinically and pathologically (see Chapter 75). [220] The areas most commonly involved with intestinal tuberculosis are the cecum and the ileum. If pulmonary tuberculosis is also present, the diagnosis of intestinal tuberculosis is not difficult, but intestinal tuberculosis can occur without pulmonary involvement. It is important not to mistake tuberculosis for IBD because steroid treatment exacerbates tuberculosis. Although Crohn's disease may be increasing in the developing world, intestinal tuberculosis remains a higher diagnostic probability among recent emigrants from Third World countries and should be ruled out among this population before a presumptive diagnosis of Crohn's disease is made.

Intestinal lymphoma can produce symptoms similar to those of Crohn's disease—fever, weight loss, diarrhea, and abdominal pain. Small bowel radiographs in lymphoma may show diffuse involvement with masses in the bowel wall, but in Crohn's disease there is usually more localized involvement of the ileum, with ulceration and narrowing of the lumen.

Collagenous or lymphocytic colitis is a chronic inflammatory disease marked pathologically by the presence of a thick collagen deposition in the subepithelial layer of the colonic mucosa, lymphocyte infiltration, or both. [221] The typical clinical presentation is chronic watery diarrhea in a middle-aged woman who smokes and uses NSAIDs. Endoscopically, the mucosa appears mildly inflamed or, more commonly, absolutely normal. The histological picture provides the diagnosis. Collagenous colitis is discussed more fully in Chapter 85.

Ischemic colitis is part of the differential diagnosis of the initial bout of IBD and should be considered in older adults or others at particular risk for ischemic disease. [222] Bloody diarrhea and abdominal pain are common with ischemic colitis. Colonoscopy reveals mucosal edema and erythema, and there may be discrete ulcers similar to those seen in Crohn's colitis. Usually, ischemic colitis spares the rectum because of its extensive collateral circulation. Differentiation of ischemic colitis from IBD, especially Crohn's colitis, may be impossible on clinical, histological, or endoscopic grounds. In contrast to IBD, ischemic colitis usually resolves spontaneously over a few weeks. However, as many as 10% of patients with acute ischemic disease can continue into a chronic picture resembling Crohn's disease, distinguished only in its failure to respond to treatment. If the mesenteric vasculature is involved, systemic vasculitis can have small intestinal manifestations similar to Crohn's disease. Small intestinal ulcerations, perforation, and hemorrhage are seen in systemic lupus erythematosus, polyarteritis nodosa, rheumatoid arthritis, and dermatomyositis. [223]

Henoch-Schónlein purpura presents with rectal bleeding and abdominal pain before the development of the characteristic rash on the legs and buttocks. Behçet syndrome is marked by oral and genital ulcers with uveitis. There is an associated inflammation of the ileocecal area with aphthous ulcers and a lymphocytic infiltrate; these lesions can resemble Crohn's disease. Moreover, Behçet syndrome and Crohn's disease share a number of extraintestinal manifestations, including arthritis, erythema nodosum, and iridocyclitis.

Diverticulitis may be difficult to separate from acute Crohn's colitis, especially in older adult patients. [224] Both diseases can present with abdominal pain, fever, diarrhea, and rectal bleeding. Endoscopically, the presence of inflammation over a long segment of colon or in the rectum, where there are no diverticula, suggests Crohn's disease, as does the presence of perianal disease or extracolonic disease. Involvement of the pericolonic fat, which can be detected by CT is more consistent with Crohn's disease. Diverticulitis, like ischemic colitis, tends to be an acute problem and does not lead to a chronic inflammatory state as does IBD. However, patients with focal Crohn-like changes complicating diverticulitis have been described. [225] Diverticulosis can be associated with rectal bleeding, but the bouts of bleeding are abrupt in onset and blood loss is great, whereas in ulcerative colitis or Crohn's colitis, blood loss is usually chronic and small in volume.

Pseudomembranous colitis presents with profuse watery diarrhea and may last from a few days to 2 months (see Chapter 88). [226] The presence of small membranous plaques adherent to the mucosa on sigmoidoscopy is pathognomonic. Not all patients with pseudomembranous colitis however have the characteristic sigmoidoscopic picture. As part of the initial evaluation of patients with acute presentation of IBD, it is appropriate to check the stool for *C. difficile* toxin, especially if there has been recent antibiotic exposure.

Mild ulcerative colitis or proctitis in which rectal bleeding is the primary manifestation can be confused with hemorrhoids or anal fissures. The presence of urgency or diarrhea is more consistent with ulcerative colitis or proctitis. Sigmoidoscopy should easily differentiate ulcerative colitis from these perianal problems.

If IBD, especially Crohn's disease of the small intestine, has a long, indolent course with relatively mild symptoms, it may be difficult to differentiate from irritable bowel syndrome. [227] It should be easy to distinguish between irritable bowel syndrome and ulcerative colitis by proctoscopy, which is normal in irritable bowel syndrome. Chronic, cramping abdominal pain and diarrhea are typical of both irritable bowel syndrome and Crohn's disease. The presence of fever, abdominal tenderness, and weight loss all suggest Crohn's disease. This differential diagnosis may be particularly troublesome in that some patients with Crohn's disease also have irritable bowel syndrome. Making the diagnosis of irritable bowel syndrome in a patient with coexisting IBD is difficult because of the overlapping symptoms and the absence of any definitive histological or biochemical marker for either IBD or irritable bowel syndrome. For many patients with IBD, there are symptoms, typically pain and diarrhea, that appear not to correlate with disease activity as assessed endoscopically or histologically. Whether these symptoms are products of undetected disease activity, "functional" symptoms of IBD, or signs of irritable bowel syndrome is difficult to establish. A confounding issue is the high incidence of depression in both irritable bowel syndrome and Crohn's disease and the influence of depression on the patient's symptoms. The coexistence of these diseases is important not only in making the diagnosis but also in following therapy. Patients who have both Crohn's disease and irritable bowel syndrome have been treated aggressively (and unsuccessfully) with corticosteroids for abdominal pain and diarrhea that were, in fact, products of irritable bowel syndrome.

Diversion colitis is a nonspecific inflammation of segments of colon excluded from the fecal stream. It sometimes occurs in patients who have had surgery for Crohn's colitis with creation of a Hartmann pouch. In these cases, diversion colitis may be difficult to differentiate from recurrence of Crohn's disease in the excluded segment. Many patients with diversion colitis have severe endoscopic findings with minimal symptoms. A clinical response to antibiotics or to enemas of short-chain fatty acids should differentiate diversion colitis from active Crohn's disease. [228]

NUTRITIONAL MANAGEMENT

Malnutrition is an important clinical problem in IBD, particularly in Crohn's disease. At diagnosis, most patients with Crohn's disease have lost weight. Hypoalbuminemia, anemia, and vitamin D deficiency all can result from malnutrition in Crohn's disease.

Five factors lead to malnutrition in Crohn's disease. The first, diminished oral intake primarily as a result of loss of appetite, is the most important cause of malnutrition in patients with Crohn's disease. [229] Anorexia may result from inflammation, depression, or the side effects of medication. Even in the absence of anorexia, many patients diminish their food intake because eating causes pain, cramps, and diarrhea; this problem is seen particularly in patients with small bowel obstruction. The second factor is the increase in caloric requirements because of the catabolism associated with fever and inflammation; however, this factor makes a relatively minor contribution to malnutrition in Crohn's disease. [230] The third factor is malabsorption of nutrients as a result of a reduction in the absorptive area by extensive surgical resections or involvement of large areas of small bowel with inflammation. Rarely, fistulae can result in malabsorption as a result of nutrients bypassing absorptive epithelium. The nutrients most likely to be affected are calcium, magnesium, fats and fat-soluble vitamins, and vitamin B_{12}. Bile salts and vitamin B_{12} are selectively absorbed in the distal ileum, the area of the small intestine most commonly involved in Crohn's disease and the area most commonly resected. Malabsorption of bile salts leads to a decrease in the total bile salt pool and thus to fat malabsorption. Bacterial overgrowth in the small intestine proximal to strictures leads to deconjugation of bile salts by bacteria. The deconjugated bile salts are absorbed passively and thus are not retained in the lumen to participate in the absorption of fat and fat-soluble vitamins. The fourth factor is the loss of proteins and electrolytes in the intestinal lumen in areas of inflammation. Bleeding into the intestinal lumen results in iron deficiency. Magnesium and zinc can be depleted by losses into the intestine. The fifth and last factor is drug therapy. Sulfasalazine impairs folate absorption but uncommonly leads to anemia. Corticosteroids induce negative nitrogen balance and decrease intestinal calcium absorption.

For most patients with IBD, the only nutritional therapy required is the instruction to eat adequate amounts of a well-balanced diet. Patients look for a nutritional basis to their disease and for nutritionally based therapy. These expectations lead many to decide that exacerbations of pain or diarrhea were caused by the foods they ate before the exacerbation. They exclude from their diets a lengthy list of foods that were associated temporally with an exacerbation of disease. As a result, these patients place themselves on a diet that has limited variety and is likely to be nutritionally unbalanced. Foods should be eliminated from the diet only if they consistently and reproducibly result in symptoms. Some patients have lactose intolerance and should avoid dairy products or use lactase-containing products, such as Lactaid. Patients with strictures should avoid fibrous, high-residue foods such as nuts, popcorn, bean sprouts, and celery.

More involved nutritional therapy in Crohn's disease can be directed toward one of two goals: treating nutritional deficiencies or reducing inflammation. Deficiencies of specific nutrients usually can be managed by supplementation. Oral or parenteral iron supplementation may be necessary in patients with significant intestinal bleeding. Specific supplementation with calcium, magnesium, zinc, vitamin B_{12}, vitamin D, and vitamin K may be required if clinical or biochemical manifestations of deficiency occur (see Chapter 53). These nutrients may be malabsorbed if there is extensive involvement of the small bowel mucosa. Bone densitometry will reveal evidence of calcium loss from bone caused by calcium malabsorption or steroid therapy. Even in the absence of clinical or biochemical evidence of micronutrient deficiency, it is reasonable to give multivitamin supplements to patients with small bowel Crohn's disease. Extensive (>100 cm) resections of the terminal ileum or lesser resections combined with active disease or scarring in the remaining ileum lead to vitamin B_{12} malabsorption. Some clinicians evaluate patients with serum vitamin B_{12} levels, but others empirically treat all patients with significant ileal resections with parenteral vitamin B_{12} (1000 µg once a month). Ileal resection and ileal disease also lead to malabsorption of bile salt and fat. Diarrhea induced by bile salts can be managed with cholestyramine, a resin that binds bile salts and prevents them from inducing water and electrolyte secretion from the colonic mucosa. Cholestyramine therapy, however, further depletes the bile salt pool and can worsen fat malabsorption. Unabsorbed fatty acids entering the colon are hydroxylated by bacteria. This leads to fat malabsorption that induces diarrhea because the hydroxylated fatty acids induce colonic water and electrolyte secretion. Fatty acid–induced diarrhea is managed by reducing the fat content of the diet. Caloric deficiencies are made up by increasing the intake of complex carbohydrates. Alternatively, medium-chain triglycerides, which do not require bile salts for absorption, can be substituted for part of the conventional long-chain triglycerides in the diet. The dose of medium-chain triglycerides is limited to about 3 tablespoons per day (300 kcal) because larger amounts cause diarrhea.

Anorexia leads to inadequate nutritional intake and malnutrition in many patients with IBD, even if absorptive function is normal. Supplementation of the patient's oral intake with defined-formula diets can prevent or reverse malnutrition. Defined formula diets can be taken orally or administered by nasogastric tube, gastrostomy tube, or jejunostomy tube (see Chapter 53). Most IBD patients who require nutritional supplementation can be managed with enteral therapy. In some cases, however, parenteral nutrition is required: in the presence of chronic small intestine obstruction that is not amenable to surgical resection; after massive small bowel resection, especially if the amount of remaining small intestine is insufficient for absorption of daily caloric needs; or if enteral nutrition results in severe, uncontrollable diarrhea.

The use of total parenteral nutrition (TPN) or the enteral administration of an elemental diet as specific therapy in IBD is controversial (see Chapter 53). Elemental diets consist of amino acids, monosaccharides, vitamins, minerals, and essential fatty acids. The results of controlled trials comparing elemental diets with drug therapy have been inconsistent. [231], [232], [233] and [234] In Crohn's disease, elemental diets have been used more extensively in children than adults. Providing adequate nutrients can reverse growth failure in Crohn's disease. Elemental diets taken orally or by nasogastric tube enhance linear growth in children with Crohn's disease. [235] In these studies, enteral nutrition was superior to steroids in promoting growth.

Malnutrition is less commonly troublesome in ulcerative colitis than in Crohn's disease, in part because malabsorption of nutrients is not a problem in ulcerative colitis. Diminished food intake, however, can be a problem in ulcerative colitis. Because of loss of appetite and because eating worsens their diarrhea, patients with ulcerative colitis reduce their calorie intake. Nutritional requirements are increased in severe ulcerative colitis because of the catabolism associated with fever and inflammation. TPN has been used to improve the nutritional status and reduce the symptoms of patients with severe ulcerative colitis. [236] This approach has been especially successful in improving the nutritional status of patients before colectomy is undertaken. Improved nutritional status decreases the likelihood of complications for surgical procedures in general and, presumably, for IBD surgery in particular. No evidence has been found that TPN reduces the necessity for colectomy or that it is effective primary therapy for the disease. Two studies compared the rates of colectomy in ulcerative colitis patients receiving TPN with those on regular hospital food. In both studies, the rates for colectomy were the same in the two groups. [237], [238]

Dietary supplementation with fish oil may have antiinflammatory activity because the n-3 fatty acids in fish oil compete in the substrate pool for the enzymes that synthesize prostaglandins and leukotrienes. Supplementation with high-dose fish oil over a period of months reduced steroid requirements in ulcerative colitis. [239] Fish oil supplementation was tested in placebo-controlled trials as maintenance therapy in Crohn's disease. In one trial it was effective, [240] but in the other it was not. [241]

DRUGS USED IN INFLAMMATORY BOWEL DISEASE

General Supportive Therapy

Drugs (antidiarrheal agents, antispasmodics, analgesics) that reduce the patient's symptoms without affecting the level of disease activity have a role in the management of IBD. The use of these drugs should not be a matter of routine for all patients; rather, their use should be individualized. Moreover, they should supplement antiinflammatory drugs, not substitute for them.

Antidiarrheal agents, usually loperamide or diphenoxylate, are useful in patients with mild disease activity to reduce the number of bowel movements and to relieve rectal urgency. In addition to controlling disease-related diarrhea, these agents can control diarrhea caused by bile salt and fat malabsorption, as seen in patients with surgical resections of the terminal ileum. Antidiarrheal agents can be given on a regular schedule or on an as-needed basis. Antidiarrheal agents may allow reduction of steroid dosage in patients with chronic steroid requirements. In patients with severe colitis, antidiarrheal agents are contraindicated because they predispose to the development of toxic megacolon. For patients who have undergone ileocolic resections and do not have fatty acid–induced diarrhea, treatment with cholestyramine (up to six scoops per day) may be particularly useful. Cholestyramine is a resin that binds bile salts in the lumen and prevents bile salt–induced colonic secretion. If the gallbladder is present, most of the dose should be given in the morning to take advantage of the concentrated bile emptied into the lumen with breakfast. Much more commonly, fatty acid–induced diarrhea will be present in patients with ileal resections; these patients will respond better to a low fat diet without the addition of cholestyramine.

Abdominal cramping is often a prominent complaint in IBD. Anticholinergics (tincture of belladonna, clidinium, propantheline bromide, dicyclomine hydrochloride) may reduce cramps, pain, and rectal urgency. They are best given before meals to depress peristalsis brought on by eating. An especially effective combination of an antidiarrheal and an antispasmodic is powdered opium (25 mg) and belladonna (15 mg). This formulation is not widely available and must be specially prepared. Opium and belladonna can be associated with drowsiness and confusion. Diarrhea that is not controlled with loperamide or diphenoxylate often responds to opium and belladonna. As with antidiarrheal agents, antispasmodics are contraindicated in severe colitis because of the risk of precipitating toxic megacolon. Antispasmodics also should be avoided if obstruction is present. Analgesia can be a difficult problem in IBD. In general, the best approach to pain management is control of disease activity. Sometimes tricyclic antidepressants can be helpful, even in the absence of clinical depression, as an adjunct for chronic pain management. Narcotics can cause dependency and, by reducing bowel motility, induce toxic megacolon. They should be avoided, especially as IBD patients with pain, but without obstruction or acute inflammation, seem especially susceptible to opiate dependency.

Anemia associated with IBD usually is due to iron deficiency or, less commonly, vitamin B_{12} or folate deficiency. Oral iron and folate or parental vitamin B_{12} should be given as indicated by laboratory studies. Parenteral iron is indicated if oral iron is not absorbed or causes gastrointestinal upset. Recombinant erythropoietin may be indicated in IBD for patients in whom anemia does not respond to iron and vitamins. [242] Folate supplementation has been suggested for patients with ulcerative colitis or extensive Crohn's colitis as a possible means for decreasing the risk of colon cancer. [243]

5-Aminosalicylic Acid Compounds

Sulfasalazine consists of 5-aminosalicylic acid (5-ASA) connected to sulfapyridine by a diazo bond. In the colon the diazo bond is cleaved by bacteria, yielding free 5-ASA and sulfapyridine. Free 5-ASA is the active agent in the treatment of ulcerative colitis; it appears to act from the lumen since little 5-ASA is absorbed into the systemic circulation from the colon. Free 5-ASA given orally is rapidly absorbed from the small intestine into the systemic circulation and has no therapeutic efficacy in ulcerative colitis. Sulfapyridine is absorbed after release in the colon and is responsible for most of the side effects associated with sulfasalazine including headache, nausea, and abdominal discomfort.

A variety of approaches have been taken to deliver 5-ASA to the colonic lumen without exposing the patient to sulfapyridine (Table 83-7). One approach was to formulate free 5-ASA into suppositories and enemas for treatment of colitis confined to the rectum and the left colon, respectively. Another approach has been to formulate oral 5-ASA to yield high colonic luminal concentrations with little systemic absorption. In olsalazine, one 5-ASA is bound covalently through an azo linkage to another 5-ASA. Like sulfasalazine this compound releases its 5-ASA in the colon when bacteria cleave the azo bond. Olsalazine causes chloride secretion in the small intestine, resulting in watery diarrhea in 5% to 10% of treated patients. [244] Formulation of 5-ASA in coated granules results in pH-dependent release (Pentasa, Asacol). Asacol, 5-ASA coated with Eudragit-S, dissolves a pH 7 or above and releases 5-ASA primarily in the colon and terminal ileum. Pentasa, 5-ASA in a semipermeable membrane, releases 5-ASA at pH6 and above. Pentasa releases 35% of its 5-ASA in the small intestine and may be useful for small intestine Crohn's

disease. Although Pentasa is widely thought of as a treatment for Crohn's disease, the FDA has not approved its use for this purpose. A controlled study of Asacol suggests it may be useful in Crohn's disease. [245] In addition, as with Pentasa, the pharmacokinetics of Asacol suggest that it may be effective in small intestinal Crohn's disease, especially when disease is confined to the distal ileum. Release in the small intestine results in increased absorption, however, and therefore increased risk of toxicity. High doses of oral 5-ASA are nephrotoxic in rats, but this has not been a major problem in human clinical studies with 5-ASA preparations. [246] The newest oral 5-ASA formulation is balsalazide (Colazide), in which 5-ASA is linked via a diazo bond to 4-aminobenzoyl-ß-alanine, an inert carrier. [247] Balsalazide releases 5-ASA in the colon.

TABLE 83-7 5-Aminosalicylate (5-ASA Preparations)

The exact role of the newer 5-ASA preparations, both rectal and oral, in the management of IBD remains to be seen. The incidence of side effects is lower with these compounds than with sulfasalazine. The increased level of tolerability allows patients to achieve higher colonic luminal concentrations of 5-ASA with these preparations than with sulfasalazine and thus increases the likelihood of a clinical response. These compounds are useful as substitutes for sulfasalazine in patients with known adverse reactions to sulfapyridine. In addition, they may have a broader therapeutic role in circumstances in which increased local concentrations of 5-ASA or an increased area of distribution are important. Pentasa and Asacol may have a place in the management of small intestinal Crohn's disease where sulfasalazine does not.

A large number of pharmacological effects have been described for sulfasalazine and 5-ASA; however, which, if any, of these pharmacological effects accounts for its efficacy in IBD is unknown. Sulfasalazine and 5-ASA inhibit 5-lipoxygenase and thus the synthesis of leukotrienes in both peripheral blood neutrophils and in IBD mucosa. [248], [249] 5-ASA acts as a free-radical scavenger and thus inactivates the products of the neutrophil respiratory burst, which are important in the microbicidal properties of neutrophils and may also be important in neutrophil-induced epithelial damage in IBD. [250] Nuclear factor-?N (NF-?B) is a transcription factor critical to the expression of multiple genes involved in the inflammatory response including cytokines and adhesion molecules. Sulfasalazine inhibits NF-?B activation by a number of activating agents including TNF-a and lipopolysaccharide. In contrast, 5-ASA does not block NF-?B activation. [251]

Two groups of side effects are associated with sulfasalazine: dose-related toxic effects thought to be related to serum sulfapyridine levels, and hypersensitivity reactions not related to serum sulfapyridine levels. The dose-related toxic effects include nausea, vomiting, headache, abdominal discomfort, and low-grade hemolysis. Sulfapyridine is metabolized by acetylation; slow acetylators have high serum levels of free sulfapyridine and low levels of acetylated sulfapyridine; whereas, fast acetylators have low levels of free sulfapyridine and high levels of acetylated sulfapyridine. The dose-related side effects are more common in slow acetylators. Most of the dose-related side effects of sulfasalazine develop in the first few weeks of therapy and resolve with discontinuation of the drug. The incidence of dose-related side effects increases at doses of 4 g/day or more. Most patients can avoid these side effects if given a dose that is sufficiently low. For patients whose only side effect is dyspepsia, relief may be obtained by taking the drug with meals or by using the enteric-coated preparation. [252] The second group of side effects, hypersensitivity reactions, includes fever, aplastic anemia, pancreatitis, lupuslike rash, nephrotoxicity, hepatitis, agranulocytosis, and autoimmune hemolysis. These effects are not related to the dose of sulfasalazine, acetylator status, or serum sulfapyridine concentrations. Nephrotoxicity can be seen with any 5-ASA compound. Caution should be exercised when using these drugs in patients with known renal dysfunction or a history of kidney disease. Hypersensitivity reactions occur less commonly than toxic reactions, but tend to occur with greater severity. Myeloblastic anemia, presumably a result of folate deficiency, has been reported with sulfasalazine but is much less common than a reduction in the serum folate concentration. [253] Malabsorption of folic acid and reduced serum folate levels are seen with sulfasalazine therapy; some clinicians recommend folic acid supplementation of 1 to 2 mg/day for patients treated with sulfasalazine. This is not necessary for the other mesalamine preparations. Although uncommon, agranulocytosis with a mortality rate of 35% reported in one series, is probably the most common fatal side effect. A small number of patients can develop a hypersensitivity reaction that can worsen colitis. It is particularly important to recognize this complication and withdraw the medication rather than treat the patient for refractory disease.

Sulfasalazine commonly causes changes in sperm morphology and decreases sperm count, leading to infertility. [254], [255] The incidence of abnormalities in sperm morphology and number appears to be higher in slow acetylators. These effects are reversible with 3 months of sulfasalazine withdrawal. The effects of sulfasalazine on sperm may relate to its antifolate activity.

5-ASA compounds have been used successfully as single agents in mild to moderate acute attacks of ulcerative colitis with favorable responses of 64% to 80% in patients treated with sulfasalazine compared to 35% to 40% favorable response in placebo-treated controls. [256], [257] and [258] In these series, the patients were treated with sulfasalazine at doses of 4 to 6 g/day. Success rates with sulfasalazine and acute ulcerative colitis are dose-related. Most clinicians do not use dosages above 4 g/day; however, several studies demonstrate better success rates with 6 g or more per day. The incidence of side effects increases with the dosage; however, if a patient tolerates 4 g/day and has not fully responded clinically, there is no reason not to push the dose higher. To lessen the side effects, sulfasalazine is given in gradually increasing doses, beginning at 500 mg twice a day and increasing to 1 g four times a day over the course of about a week. Taking sulfasalazine with food in four divided doses lessens gastric symptoms in most patients, but if the patient tolerates the drug the schedule can be changed to 2 g twice a day. Patients who respond to sulfasalazine usually do so in 2 to 3 weeks although some take 4 weeks or longer.

5-ASA preparations are as effective as sulfasalazine, although not superior to sulfasalazine, in the management of acute exacerbations in ulcerative colitis. [259], [260] and [261] Steroids induce remission in severe ulcerative colitis in a higher percentage of the patients than 5-ASA compounds and induce them more quickly. [262], [263] For this reason corticosteroids, either alone or in combination with 5-ASA compounds, are the treatment of choice in moderate to severe attacks. There is no evidence to suggest that the combination is better than corticosteroids alone. One advantage of combined therapy is that when the time comes to taper steroids the patient is already on a 5-ASA compound. If patients on combined therapy go into remission the usual approach is to taper and stop steroids and continue the 5-ASA compound.

The efficacy of 5-ASA compounds in preventing relapse in ulcerative colitis is well established. [264] In maintenance therapy, as in acute therapy, the higher the dose of sulfasalazine the greater the likelihood of success. When maintenance doses of 1, 2, and 4 g of sulfasalazine per day were compared the recurrence rates were 33%, 14%, and 9%, respectively (Fig. 83-24). These findings indicate a dose-response effect to the prevention of relapse by sulfasalazine. Some clinicians use sulfasalazine at a dose of 2 g per day for maintenance therapy because more patients tolerate 2 g/day than 4 g/day. However, if patients tolerate higher doses of sulfasalazine the higher doses should be used for maintenance therapy. The oral formulations of 5-ASA are also effective in the maintenance of remission in both ulcerative colitis and Crohn's disease. Both olsalazine and Pentasa are as effective as sulfasalazine in the maintenance of remission in ulcerative colitis. [265], [266] The risks of long-term maintenance therapy with sulfasalazine and 5-ASA compounds are modest. There are no definitive guidelines for the duration of maintenance therapy; the usual approach has been to continue therapy for several years in patients who have had a single attack of mild to moderate severity and to continue maintenance therapy indefinitely in patients with multiple relapses.

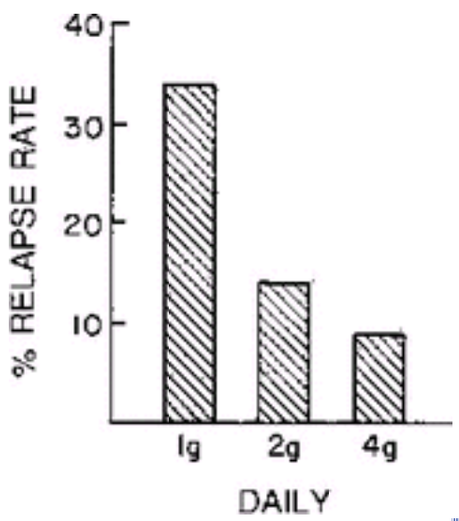

FIGURE 83-24. Relapse rate with maintenance sulfasalazine. (From ref. [352].)

5-ASA compounds are also useful in acute attacks of Crohn's colitis. In the National Cooperative Crohn's Disease Study, sulfasalazine at a dose of 1 g per 15 kg of body weight per day for 17 weeks was superior to placebo, inducing symptomatic improvement for patients with colonic involvement whether or not the small bowel was involved. [267] Sulfasalazine was not effective against disease confined to the small bowel, however, and patients were less likely to respond to sulfasalazine if they had been on prednisone before being randomized to sulfasalazine. The European Cooperative Crohn's Disease Study also found sulfasalazine therapy to be beneficial for patients with active colonic disease. [268] In that study, treatment with sulfasalazine (3 g/day) resulted in a modest but statistically significant improvement in the CDAI. Asacol is as effective as corticosteroids in the treatment of active Crohn's ileitis. [269]

Several studies investigated the usefulness of 5-ASA compounds in preventing relapse in patients with Crohn's disease. The National Cooperative Crohn's Disease Study examined the effects of sulfasalazine in patients who achieved remission during the initial study. [267] Most patients in both the sulfasalazine- and placebo-treated groups remained in remission, with no significant differences between the groups. Similarly, the European Cooperative Crohn's Disease Study showed no benefit of sulfasalazine treatment for patients with quiescent disease at entry into the study. [268] Despite the lack of support in the literature, many physicians use sulfasalazine as maintenance therapy in Crohn's disease, especially for patients with Crohn's colitis in whom acute disease responded to sulfasalazine. One study did suggest a modest benefit of Pentasa at 4 g/day in Crohn's disease. [245] Asacol was also superior to placebo in the maintenance of remission in patients with Crohn's disease, particularly in patients with ileitis. [270], [271]

A number of studies have investigated the effects of 5-ASA compounds on infants in utero and on nursing children. No evidence of harmful effects has been found, and the usual recommendation is that the drug can be used safely either in acute or maintenance therapy in pregnant and lactating women. [272], [273] Folate supplementation in early pregnancy reduces the risk of neural tube defects. The antifolate properties of sulfasalazine suggest that dietary folate supplements may be especially important in women taking sulfasalazine who are pregnant or contemplating pregnancy. Children can be treated safely with sulfasalazine. For acute disease in children older than 2 years, the initial dosage is 25 to 40 mg/kg daily in three to six doses; this dosage can be increased gradually to 75 mg/kg daily. [274] For maintenance therapy in children older than 2 years, the dosage is 20 mg/kg daily in divided doses. An oral suspension is available.

Corticosteroids

Steroids are highly effective in bringing about remission in patients with ulcerative colitis and Crohn's disease. The clinical benefits are counterbalanced by their predictable and considerable side effects. In addition, although clinically beneficial, corticosteroids do not often bring about mucosal healing in Crohn's disease. The hazards of steroids as well as their questionable benefit in mucosal healing and a high rate of steroid dependence has brought about a reevaluation of their role in IBD. Attempts to substitute other medications for steroids at an earlier time point or to avoid steroids all together, particularly in Crohn's disease, are increasingly advocated by some gastroenterologists.

Corticosteroids prevent or suppress inflammation induced by chemical, mechanical, infectious, and immunologic agents. Corticosteroids block the early manifestations of inflammation: enhanced vascular permeability, vasodilation, and neutrophil infiltration. They also block the later stages of the inflammatory process: vascular proliferation, fibroblast activation, and collagen deposition. Corticosteroids have in vitro effects on immunologic events, such as T-cell response to antigens, as well as on inflammatory events, such as neutrophil migration and mediator production.

In 1955, Truelove [101] compared patients with ulcerative colitis treated with cortisone (100 mg/day) with subjects treated with placebo for 6 weeks. Cortisone was superior to placebo in patients with disease ranging in severity from mild to severe; the difference was less striking in the more severely ill patients. Corticosteroids are effective in ulcerative colitis given either orally or parenterally; oral steroid therapy is effective in most patients with mild to moderate disease. [262] Few studies have assessed the relative value of various dosage regimens of oral corticosteroids. In one study (Fig. 83-25), two thirds of outpatients treated with 40 to 60 mg of prednisolone per day improved over a period of 3 weeks compared with just one third of those receiving 20 mg/day. [275] Most responders improved within 2 weeks. As a result of this study, 40 mg/day has been widely recommended as an initial dosage of prednisone for patients with moderately severe ulcerative colitis (prednisone and prednisolone are effectively equal in antiinflammatory potency). Some practitioners give an inadequate dose of prednisone because of the fear of side effects. This approach markedly reduces the likelihood of a positive response. For most patients, administration of oral prednisone in a single morning dose is as effective as divided doses; [276] however, use of divided doses may be appropriate in selected patients. Administration of prednisone as a single dose in the early morning may cause less suppression of endogenous steroid production.

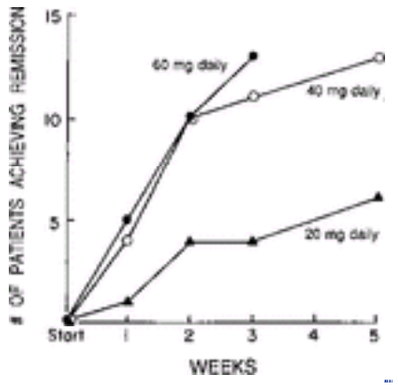

FIGURE 83-25. The number of patients with active ulcerative colitis achieving remission over a 5-week period with increasing doses of prednisolone. (From ref. [275].)

Corticosteroid enemas are useful in left-sided colitis. [277], [278] In most patients, a 100-mL corticosteroid enema reaches the mid-descending colon, although in some the enema reaches the midtransverse colon. About 80% of patients with proctitis obtain a remission or significant improvement with one enema per night for 2 to 3 weeks. The extent of systemic absorption varies with the corticosteroid preparation and the severity of inflammation. In general, systemic absorption is much less than that with orally administered steroids; however, in some cases, as much as 75% of the dose administered rectally is absorbed. [279], [280] In addition to enema preparations, corticosteroids are also available in foam suspension. Because proximal penetration with foam is not as great as with enemas, foam can be used only for disease confined to the rectum; however, foam preparations are less bothersome for the patient than enemas.

The National Cooperative Crohn's Disease Study found that a 17-week course of prednisone (0.25–0.75 mg/kg daily) was more effective than placebo in Crohn's

disease of the small bowel. [267] Sixty percent of prednisone-treated patients achieved remission compared with 30% of those receiving placebo. The European Cooperative Crohn's Disease Study found similar benefits with oral corticosteroids in Crohn's disease. [268] In a large series of 142 patients with active Crohn's disease, treatment with prednisolone (1 mg/kg daily) resulted in clinical remission in 92% of patients within 7 weeks, although the rate of endoscopic remission was much lower. [194] Considering the prominent role that corticosteroids have played in the management of Crohn's disease, few studies have addressed the issues of optimal doses, duration of therapy, and parameters for adjusting dosages. The physician must ensure that there is no abscess present if the use of corticosteroids is being considered. An abscess requires surgical drainage and antibiotics, and the use of corticosteroids in a patient with an undrained abscess may lead to uncontrolled sepsis. In addition, the physician also must be certain that the patient's symptoms relate to inflammation and not to a stricture or other fibrotic process that will not respond to steroids. The usual starting oral dosage is 40 mg of prednisone per day; starting with lower doses diminishes the likelihood of a positive response. These dosage recommendations should be adjusted if the patient's response to steroids is known from past experience. Corticosteroids can be given parenterally if the patient cannot tolerate oral medication. The patient is left on high doses of corticosteroids until symptoms begin to diminish, after which the dose is reduced gradually.

Maintenance therapy with corticosteroids for the prevention of recurrences in patients in clinical remission has been demonstrated to be ineffective in both ulcerative colitis and Crohn's disease. [267] In both diseases, however, some patients have continuously active disease that responds to corticosteroids but flares up if the dose of steroids is reduced.

The many side effects of corticosteroids are the major factor limiting their use in IBD. [268], [281] Although the side effects of corticosteroids are dose-dependent, there is a wide range of patient susceptibility to these side effects. Some patients tolerate prolonged courses of high-dose corticosteroid therapy without ill effects, but others develop devastating side effects with short courses of low-dose therapy. The range of patient susceptibility to developing corticosteroid side effects is a function of differences in plasma protein binding (with hypoalbuminemic patients being at risk) and of variations in rates of metabolism and clearance of synthetic steroids. The major differences between the manifestations of iatrogenic steroid administration and the manifestations of spontaneous Cushing syndrome are related to the increased levels of androgens and mineralocorticoids in spontaneous Cushing syndrome. [282] Features more common in spontaneous Cushing syndrome include hypertension, virilism, striae, and purpura. Features more common in iatrogenic steroid administration include posterior subcapsular cataracts, avascular necrosis of bone, osteoporosis, and glaucoma. Although there is much individual variability in response, patients taking 10 mg or more of prednisone daily for longer than 3 weeks should be viewed as having a suppressed hypothalamic-pituitary-adrenal axis for 1 year after cessation of therapy and should receive supplemental glucocorticoids for surgery or severe illness. Patients should be instructed about adrenal suppression and the need for glucocorticoid supplementation for surgery and severe illness even after cessation of therapy.

Patients also need to be educated about the benefits and toxicities of glucocorticoids. In particular, they should understand the more predictable side effects of osteoporosis and cataract formation and the necessity of observing for their occurrence. Common predictable side effects include increased appetite, centripetal obesity, moon facies, acne, insomnia, increased risk of infection, and, in children, growth arrest. Less common problems for which the patient should be monitored include hypertension and glucose intolerance. Steroid-induced glucose intolerance may require treatment with insulin or oral hypoglycemic agents. In patients with diabetes, steroids may make blood-sugar control more difficult. Psychiatric problems occur, including depression and psychosis. Avascular bone necrosis, particularly of the femoral heads, occurs and can result in permanent disability.

Osteoporosis is a common and potentially devastating side effect of corticosteroid therapy. Every patient treated with glucocorticoids develops negative calcium balance. The effects of corticosteroids on bone include both enhanced bone resorption and diminished bone formation. [282] Steroid-induced bone loss is a particular problem in small intestine Crohn's disease, in which malabsorption of vitamin D and calcium also can accelerate calcium loss from bone. Bone loss characteristically involves trabecular bone in ribs, vertebrae, and distal radius. Rib fractures and vertebral compression fractures are common. The incidence of symptomatic bone loss is higher in children and postmenopausal women. Data are conflicting on the effects of low-dose corticosteroids on bone loss. Dykman and colleagues [283] reported significant bone loss with dosages as low as 8 to 10 mg of prednisone per day, but Sambrook and associates [284] reported no increase in bone loss over a period of 7 years using an equivalent dosage of prednisolone. To minimize bone loss, the management of patients on corticosteroids should include the following [147]:

use of the lowest possible dose of corticosteroids
smoking cessation, limitation of alcohol consumption, and 30 to 60 minutes of weight-bearing exercise per day
history, physical examination, and laboratory evaluation, including bone mineral densitometry
dietary or supplemental calcium (1500 mg/day)
vitamin D (50,000 IU 1–3 times/week) or calcitriol (0.5 µg/day)
hormone (estrogen) replacement therapy where appropriate.

Posterior subcapsular cataracts occur frequently with corticosteroid therapy, and their incidence correlates with dose and duration of therapy. [285] Of patients receiving 15 mg of prednisone for 1 year, 25% developed lenticular changes. Glucocorticoids also increase intraocular pressure in up to 40% of patients. Irreversible glaucoma and blindness can occur in susceptible persons. Referral to an ophthalmologist for baseline measurement of intraocular pressure and slit-lamp examination should be considered if long-term corticosteroid management is expected. Periodic examinations should be performed if therapy is continued.

The high level of efficacy of systemic corticosteroids in IBD combined with a high level of undesirable side effects has led to attempts to develop new corticosteroids with the same level of efficacy but diminished toxicity compared with currently available drugs. Budesonide is a potent corticosteroid whose systemic toxicity is diminished by rapid first-pass metabolism in the liver. It was formulated with a Eudragit-S coating to direct its delivery to the terminal ileum and proximal colon, areas likely to affected by Crohn's disease. In theory, this formulation would allow the absorption of budesonide in the area of active inflammation in Crohn's disease. The absorbed budesonide then would travel through the portal circulation to the liver, where it would be metabolized; relatively little unmetabolized budesonide would escape from the liver to cause the systemic side effects associated with conventional corticosteroids. This formulation was used in an 8-week, placebo-controlled Crohn's disease trial; budesonide was effective in inducing remission, but the doses of budesonide that induced remission also suppressed plasma cortisol levels. [286] Budesonide is more effective than mesalamine [287] and as effective, or almost as effective, as prednisone in inducing remission in Crohn's disease. [288], [289] The suppression of serum cortisol with budesonide is appreciable but less than that seen with prednisone. Oral budesonide at 3 or 6 mg daily was compared with placebo as maintenance therapy in Crohn's disease. [290] Mean time to relapse was 124 days and 168 days in those receiving 3 and 6 mg of budesonide, respectively. In comparison, the mean time to relapse was 39 days in those receiving placebo. However, at 1 year the rate of relapse was similar in all three groups. In a study of the prevention of recurrence after surgery in Crohn's disease, budesonide was found to decrease the rate of recurrence in those who had undergone resection for disease activity but not in those who had resections for fibrostenotic disease. [291] All available studies are of relatively short duration; a comparison of the side effects of budesonide and prednisone in a long-term study is needed to confirm any relative clinical advantage for budesonide.

Immunomodulators

Immunomodulators are a loosely defined group of drugs that act by blocking lymphocyte proliferation, activation, or effector mechanisms. There is fairly extensive experience in the use of two immunomodulators, azathioprine and its metabolite, 6-mercaptopurine (6-MP), in the treatment of IBD and lesser experience with other immunomodulators, including cyclosporine and methotrexate (reviewed by Sandborn). [292] The mechanisms of action for these compounds in IBD are not completely understood.

Azathioprine is a prodrug that is non-enzymatically converted to 6-MP. The role of these immunomodulators in Crohn's disease therapy has been controversial. The National Cooperative Crohn's Disease Study found that azathioprine was no more effective than placebo in a 17-week trial. [267] The therapeutic effects of azathioprine and 6-MP are not seen until after 3, 4, or more months of therapy, and the 17-week treatment period in this study was too short to demonstrate the maximal effect. The use of azathioprine and 6-MP in the treatment of Crohn's disease is no longer a source of controversy because several studies have shown these drugs to be efficacious. [293], [294], [295], [296] and [297] In one placebo-controlled study, 67% of the 6-MP-treated patients improved, compared with 8% of those receiving placebo. [293] In another study, patients with active Crohn's disease were randomized to prednisone or prednisone plus azathioprine; 38% of those receiving prednisone alone achieved remission, compared with 70% of those receiving both drugs. [298] A metaanalysis of the use of azathioprine and 6-MP in Crohn's disease led to the conclusion that these drugs are effective both in treating active disease and in maintaining remission. [299] In earlier studies, azathioprine and 6-MP were used at relatively low doses to minimize toxicity. More recently somewhat higher doses have been used, and typical initial doses are 1 to 1.5 mg/kg for 6-MP and 2.0 to 2.5 mg/kg for azathioprine. Many patients who fail to respond to lower doses will respond to higher doses; if there are no adverse effects it is reasonable to push the dose higher.

Azathioprine and 6-MP induce leukopenia in a dose-dependent manner; some investigators suggest that leukopenia and efficacy are related. In one retrospective

study, patients who became leukopenic were more likely to go into remission and to do so sooner. [300] One problem with the use of 6-MP and azathioprine is the long delay between the initiation of therapy and the clinical response. This delay is typically 3 or 4 months. In one open-labeled study, the administration of an intravenous loading dose of azathioprine reduced the time to response; [301] however, a subsequent placebo-controlled trial could not confirm this effect. [302]

Both the therapeutic efficacy and the adverse events associated with azathioprine and 6-MP are related to their metabolism and the serum levels of their active metabolites. [303], [304] 6-MP can be metabolized intracellularly by thiopurine methyltransferase (TPMT) to an inactive metabolite, 6-methylmercaptopurine (6-MMP), or anabolized to the active metabolites, 6-thioguanine (6-TG) nucleotides and 6-methylmercaptopurine ribonucleotides (6-MMPRs) (Fig. 83-26). Both the therapeutic efficacy and the hematologic toxicity of 6-MP are positively related to the serum levels of 6-TG. There is an established genetic polymorphism for TPMT, which results in significant interindividual variation in its activity. One in 300 (0.3%) individuals have low to absent TMPT activity, whereas 11% have intermediate activity and 89% have normal to high levels of activity. Low levels of TMPT activity have been associated with high levels of 6-TG and, as a consequence, with high levels of both therapeutic efficacy and bone marrow suppression. Either TPMT genotyping or measurement of 6-TG levels may assist clinicians in optimizing the therapeutic response to azathioprine and 6-MP and in identifying individuals at increased risk for drug-induced toxicity. [303], [304] Allopurinol reduces the metabolism of azathioprine and 6-MP; coadministration of allopurinol requires a reduction in dose.

FIGURE 83-26. Metabolism of azathioprine and 6-mercaptopurine.

Although most investigators and clinicians agree that these drugs have an important role in the management of Crohn's disease, opinions differ as to what that role should be. Their use is widely accepted in patients who have active disease that is unresponsive to steroids (steroid-refractory patients) and by those with active disease that is controlled by steroids but in whom steroids cannot be withdrawn (steroid-dependent patients). In these patients, 6-MP or azathioprine is added to steroid therapy; after 3 or 4 months, when the 6-MP and azathioprine are likely to have taken effect, the dose of steroids is gradually tapered. The patient is considered to be in remission induced by 6-MP or azathioprine if the activity of the disease does not flare as steroids are withdrawn. If the activity flares as steroids are withdrawn, the patient is considered to have failed to respond to 6-MP or azathioprine and the drug is withdrawn. Some clinicians use 6-MP and azathioprine as first-line therapy in Crohn's disease, although most reserve these drugs for steroid-refractory or steroid-dependent patients. These drugs also have been promoted as primary therapy for fistulae in Crohn's disease. [293] Although many fistulae heal, the fistulae tend to recur after the drugs are stopped. Azathioprine and 6-MP also are useful as maintenance therapy in Crohn's disease. Three placebo-controlled trials of 6-MP or azathioprine in the maintenance of remission have been done in patients who were brought into remission by these drugs. [305], [306] and [307] In these trials, the relapse rate of patients on placebo was 2.5 to 8 times the relapse rate on active drugs. The duration of maintenance therapy with 6-MP and azathioprine has evolved over time. Previously clinicians would discontinue these drugs after a few years of maintenance therapy; however, patients relapsed within a few months after these drugs were withdrawn. [308] Now most clinicians continue these drugs indefinitely after remission is achieved.

The major limiting factor in the use of 6-MP and azathioprine is their toxicity. [309] They commonly cause leukopenia, and the leukocyte count of patients receiving them must be monitored carefully. [310] There is no universally accepted timetable for monitoring blood counts. Monitoring every week or two at the start of therapy and then every 1 to 3 months is reasonable. These drugs should be tried only in patients who are likely to be compliant with the required laboratory monitoring. Pancreatitis is a common side effect, but withdrawal of the drug usually leads to prompt resolution. The largest review of the toxicity of 6-MP in IBD cataloged experience with 396 patients with an average follow-up of 60 months. [311] Significant but reversible morbidity included pancreatitis (3.3%), bone marrow depression (2%), and allergic reactions (2%). Infectious complications were seen in 7%, including one case of herpes zoster encephalitis. There were 12 neoplasms, but probably only one, a diffuse histiocytic lymphoma of the brain, was associated with the drug. Another retrospective study reviewed the incidence of cancer in a group of 755 patients who had received azathioprine for IBD. [312] The average dose was 2 mg/kg and the median follow-up was 9 years. No increase occurred in the incidence of lymphoma or in total cancer cases in the azathioprine-treated patients. An analysis of the effect of azathioprine on life expectancy in Crohn's disease revealed that the enhanced life expectancy due to successful treatment of Crohn's disease exceeded any decrease in life expectancy due to the increased risk of lymphoma. [313]

The roles of 6-MP and azathioprine in ulcerative colitis are less clear than in Crohn's disease. Controlled trials demonstrated the efficacy of azathioprine (1.5–2.5 mg/kg daily) as a steroid-sparing agent in ulcerative colitis. [314], [315] As in Crohn's disease, 6-MP and azathioprine are effective in maintaining remission in ulcerative colitis in patients brought into remission with these drugs. [316]

Methotrexate is another immunomodulator that has been used in IBD, [317] although experience with this drug is much less than with 6-MP and azathioprine. It is a folate antagonist that has been widely used in other chronic inflammatory diseases, including psoriasis and rheumatoid arthritis. Three placebo-controlled trials of methotrexate in Crohn's disease have been reported. [318], [319] and [320] In one study, methotrexate was given intramuscularly (25 mg once a week) for 16 weeks. The remission rate in the placebo group was 19% and in the methotrexate group 39%. [318] In the second study, methotrexate was given orally (5 mg three times a week) for a year to steroid-dependent patients. Patients receiving methotrexate were more likely to be able to withdraw from steroids. [319] In the third study, methotrexate was given orally 15 mg/week. There was a trend for fewer flares in the Crohn's disease group but this did not reach statistical significance. [320] Patients who enter remission on methotrexate are likely to remain in remission if a low dose (25 mg IM once a week) of methotrexate is continued. [321]

Methotrexate has a high incidence of side effects, including nausea, bone marrow suppression, and hepatic toxicity. Elevated transaminases are common in patients with IBD treated with methotrexate. The incidence of methotrexate-associated fibrosis/cirrhosis is high in psoriasis and low in rheumatoid arthritis, although the reasons for this distinction are not clear. There is not enough experience with methotrexate in IBD to predict the incidence of fibrosis/cirrhosis. Monitoring for both hepatic injury and bone marrow suppression is required with methotrexate therapy. Methotrexate induces abortions and should not be used in pregnant women or those considering pregnancy. Folic acid (1 mg/day) reduces the bone marrow suppression seen with methotrexate. The risk of bone marrow suppression with methotrexate is increased in renal disease and with simultaneous use of trimethoprim/sulfamethoxazole.

The exact place of methotrexate in the management of Crohn's disease is not clear. In general, the same steroid-dependent and steroid-refractory patients who are candidates for 6-MP or azathioprine could be candidates for methotrexate. The larger experience with 6-MP and azathioprine would make them the more likely choice. There is no basis in the literature for treating ulcerative colitis with methotrexate. The only placebo-controlled trial found no benefit for methotrexate in steroid-dependent ulcerative colitis. [322]

Cyclosporine is a powerful immunomodulator that is widely used to prevent rejection in transplant patients; there is increasing experience with the use of cyclosporine (parenteral, oral, enema formulations) in IBD. [323] One clear advantage of cyclosporine over azathioprine and 6-MP is that the onset of action is seen in a few days rather than several months. There is some experience with the use of intravenous cyclosporine in ulcerative colitis. Here, the major indication is an attempt to avoid colectomy in severe disease that is unresponsive to intravenous corticosteroids. In one series 36 of 42 patients with severe ulcerative colitis, 62%, avoided colectomy. [324] Those who received 6-MP or azathioprine in addition to cyclosporine did better. In another series, patients who failed to respond to 7 to 10 days of intravenous corticosteroids were given placebo or cyclosporine at a daily dose of 4 mg/kg by continuous infusion. Nine of 11 patients who received cyclosporine responded, but none of the nine placebo-treated patients responded. [325] The approach to the use of cyclosporine in ulcerative colitis is to control disease activity with 1 to 2 weeks of parenteral cyclosporine and then switch to oral cyclosporine for 3 months. During that time, the patient is begun on 6-MP with the hope that after 3 months the cyclosporine can be stopped and the 6-MP continued. How many cyclosporine-treated patients remain under control after being switched to oral cyclosporine is not known, and the long-term success rate with this approach is not established. Anecdotal reports suggest that many patients treated this way come to colectomy within a few months despite cyclosporine therapy. Nonetheless, if there is a role for cyclosporine in IBD, it is in patients with severe ulcerative colitis for whom colectomy is being considered. Cyclosporine has been tested in four double-blind studies in chronic Crohn's disease. In one study, patients were treated for 3 months with a high dose (5–7.5 mg/kg daily) of oral cyclosporine. [326] Cyclosporine was superior to placebo in achieving treatment goals designated for each patient but was not superior

to placebo in reducing the CDAI. Moreover, the benefits did not persist when cyclosporine was withdrawn. In three other placebo-controlled trials of lower dose oral cyclosporine in Crohn's disease, no benefit was seen with the drug. [327], [328] and [329] All three of the negative studies had much larger numbers of patients than the single positive study.

Side effects are a major issue with cyclosporine therapy. Renal toxicity is the greatest concern. Cyclosporine causes a 20% decrease in glomerular filtration rate. This decrease was thought to be entirely reversible, but it is now clear that many patients suffer irreversible renal damage with cyclosporine. Risk factors include increased initial creatinine, increased age, and high cyclosporine blood levels. Other side effects with cyclosporine include hypertension, paresthesias, tremor, and headache.

Antibiotics

Except in cases of overt sepsis, antibiotics have been thought to have only a limited role in the management of ulcerative colitis. Antibiotics do not affect the remission rate, [330] and a trial of metronidazole in ulcerative proctitis showed it to be ineffective. [331] Moreover, the risk of inducing antibiotic-associated pseudomembranous colitis must be considered. However, one blinded study demonstrated that long-term (6 months) treatment with ciprofloxacin improved the results of conventional therapy with mesalamine and prednisone in the induction and maintenance of remission. [332]

Antibiotics have a larger role in the management of Crohn's disease where they are important in the treatment of the suppurative complications, especially abscess formation and perianal disease, although surgical drainage is the primary therapy for abscesses. In addition, antibiotics are used to treat the diarrhea caused by overgrowth of bacteria in the small bowel. This problem usually occurs because of stasis behind a stricture. If the stricture is not amenable to surgical or endoscopic management, the symptoms of pain, gaseousness, and diarrhea can be managed with courses of broad-spectrum antibiotics, such as tetracycline.

Metronidazole has been the most extensively studied antibiotic in the management of Crohn's disease. The efficacy of metronidazole has been demonstrated in perianal Crohn's disease and in colonic Crohn's disease. A Swedish trial compared metronidazole (800 mg/day) and sulfasalazine (3 g/day) in patients with Crohn's disease. [333] The two drugs were equally effective, and both were more effective in disease confined to the colon. A second study compared metronidazole at 10 mg/kg and 20 mg/kg with placebo in Crohn's disease and found dose-related improvements in disease activity, but the incidence of side effects was high, and only half the patients completed the study. [334] The mechanism of action of metronidazole in Crohn's colitis is not known. An important side effect of metronidazole is neurotoxicity resulting in numbness and burning in the feet.

Ciprofloxacin is another antibiotic that is used with some frequency in Crohn's disease. A typical dosing schedule is 500 mg twice a day for a few weeks. In a randomized comparison of ciprofloxacin 500 mg bid and Pentasa 2 g bid for 6 weeks in patients with mildly to moderately active Crohn's disease, the rates of remission and the rates of improvement were the same in the two groups. A controlled trial of steroids versus combined treatment with ciprofloxacin and metronidazole demonstrated an equivalency between the two treatment arms. [335] Antibiotics are generally more useful in Crohn's colitis than in ileal disease.

Infliximab

Infliximab, a mouse-human chimeric monoclonal IgG1 antibody directed against TNF, is approved by the FDA for treatment of Crohn's disease and rheumatoid arthritis. The mechanism of action of infliximab is not clear. One possibility is that it neutralizes free TNF; another possibility is that, as an IgG1 antibody, it lyses lymphocytes that express TNF on their surface through complement fixation or antibody-dependent cell-mediated cytotoxicity.

After a promising open label study in Crohn's disease, [336] a randomized placebo-controlled trial of infliximab was performed in patients with moderate to severely active disease. [67] Single infusions of infliximab (5, 10, or 20 mg/kg) resulted in a decrease of 70 points or more in the CDAI in 65% of treated patients at 4 weeks. In comparison, 17% of patients treated with placebo responded. The response rate was somewhat better in patients receiving 5 mg/kg than in those receiving 10 or 20 mg/kg. Patients who continued to respond were re-randomized at week 12 to receive either placebo or infliximab at 10 mg/kg every 8 weeks for an additional four infusions. Although there were no statistically significant differences in the response rate after repeated dosing there was a trend toward a higher response rate in patients treated with infliximab at week 44, 8 weeks after the last infusion. There was a significant difference in the rate of remission at week 44 with 52.9% of patients treated with infliximab in remission compared with 20% of patients given placebo.

Infliximab is also useful in closing fistulae in Crohn's disease. [337] In a randomized controlled trial, patients with enterocutaneous fistulae were randomized to infusions of placebo or infliximab at 5 or 10 mg/kg at weeks 0, 2, and 6. Success was defined as closure of 50% or more of the fistulae at two visits one month apart. The success rate was 68% in patients receiving 5 mg/kg of infliximab and 56% for those receiving 10 mg/kg compared with 26% in the placebo-treated group. These results were statistically significant. In the infliximab-treated group the medium duration of response was 3 months.

The short-term toxicity after a single dose of infliximab appears to be modest but toxicity increases with repeated dosing. It is a chimeric antibody, part human and part mouse, and is associated with both acute and delayed hypersensitivity reactions. [338] Some patients develop high titers of human antichimeric antibodies. Retreatment is associated with delayed hypersensitivity reactions marked by rash, fevers, arthralgias, and myalgias; these typically occur 3 to 10 days after retreatment. Infliximab has been associated with the development of a number of infections including varicella zoster, *Candida* esophagitis, and tuberculosis. TNF is an important regulator of the immune response to infectious agents; thus, it is not surprising that an antibody to TNF would be associated with infectious complications. At least six cases of lymphoma have been seen in patients receiving infliximab; four of these occurred in patients with rheumatoid arthritis, one in a patient with Crohn's disease, and one in a patient with acquired immunodeficiency syndrome (AIDS). [339] Both rheumatoid arthritis and Crohn's disease are themselves associated with an increased incidence of lymphoma, thus it is difficult to establish a causal relationship for infliximab and lymphoma in these cases. An interesting and unexpected complication associated with infliximab therapy has been the development of strictures in the gastrointestinal tract associated with the rapid healing of ulcers. Some of these strictures have required dilation.

The fast-track approval of infliximab by the FDA resulted in its release at a time when there was little experience with either repeated dosing or combination therapy with other agents. Moreover, the rapid approval resulted in its release at a time when little safety data were available. This has created problems for clinicians in deciding on which patients to treat with this drug. Although there is some evidence for the efficacy of repeated dosing [340] the general impression has been that both the strength and duration of the response declines with each additional infusion. Moreover, the incidence of adverse events increases with subsequent infusions. Therefore, it would seem reasonable to combine infliximab with an immunomodulator such as azathioprine or 6-MP with the hope that a short course of infliximab would control disease activity until the efficacy of azathioprine or 6-MP was established 3 or 4 months after the initiation of therapy. There are, as yet, no large-scale controlled studies investigating this approach.

Future Therapy

A large number of new therapeutic approaches are under investigation for the management of IBD, most of them directed at Crohn's disease. Several of these therapeutic approaches involve either treatment with recombinant cytokines or treatment with antibodies to cytokines.

Recombinant human IL-10 has been tested in the treatment of Crohn's disease. The rationale for IL-10 therapy is relatively straightforward: Crohn's disease is mediated by Th1 lymphocytes and IL-10 suppresses production of IL-2 and IFN-? by Th1 lymphocytes. There have been three controlled trials of IL-10 in Crohn's disease. [341] On the whole, the results of these studies have been disappointing and it is not clear that IL-10 as monotherapy will have a role to play in the management of Crohn's disease. In a single study 37 adults with moderate to severely active Crohn's disease were randomized to 4 months of self-administered injections of growth hormone (loading dose 5 mg/day subcutaneously for one week, followed by a maintenance dose of 1.5 mg/day) or placebo. [342] At 4 months the decrease in the CDAI in the treated group was significantly greater than in the control group.

CDP571 is a humanized IgG4 antibody against TNF-a. In a randomized placebo-controlled trial of patients with mild to moderately active Crohn's disease CDP571 was significantly better than placebo. [343] CDP571 has two characteristics that distinguish it from infliximab. One distinction is that CDP571 is a humanized antibody, whereas, infliximab is a mouse-human chimeric antibody; this distinction may account for the superior safety profile of CDP571 compared with infliximab. In particular, CDP571 was associated with fewer infusion reactions. The second distinction is that CDP571 is an IgG4 antibody, which does not bind complement and cannot mediate antibody-dependent cell-mediated cytotoxicity; whereas, infliximab is an IgG1 antibody which can bind complement and which can mediate antibody-dependent cell-mediated cytotoxicity. It is possible that the improved efficacy of infliximab in comparison with CDP571 may be due to its ability to bind

complement and thus to lyse lymphocytes that express TNF-a on their surface.

A central role of the intestinal flora has been proposed for both Crohn's disease and ulcerative colitis. One approach to altering the intestinal floral involves the use of probiotics, the administration of live bacteria as a therapeutic agent. A nonpathogenic strain of *E. coli*, Nissle 1917, has been evaluated in ulcerative colitis for induction of remission as well as maintenance of remission. In a two-part study, initial treatment for active colitis and then maintenance, the *E. coli* strain appeared as effective as mesalamine for attaining remission (75% patients in the mesalamine group attained remission compared with 68% in the *E. coli* group). [344] The successful use of probiotics was demonstrated in a placebo-controlled trial for the maintenance of remission in chronic pouchitis. [345] The probiotic used in this study, VSL#3, is composed of eight strains of bacteria including lactobacilli, *Bifidobacterium*, and *Streptococcus* at a dose of 1.8 × 10 [12], a much higher dose than studied in other probiotic studies. Only 2 of 20 patients relapsed after 9 months of treatment with probiotics in contrast to 100% of the 20 patients receiving the placebo. These products are being studied for use in ulcerative colitis and Crohn's disease. A variation of these approaches is the use of a parasite, *Trichuris suis*. Infection with helminths modulates the Th1 response and it was thought that this modulation might be therapeutic in Crohn's disease. After positive pilot studies, placebo-controlled trials are testing *T. suis* in both Crohn's disease and ulcerative colitis. [346]

An alternate approach of immune stimulation in Crohn's disease relies on genetic immune deficiency syndromes, such as chronic granulomatous disease and glycogen storage disease type Ib, as models to understand the pathophysiology of Crohn's disease. Crohn's disease or a Crohn-like intestinal disease is highly associated with these genetic diseases which have a defect of innate immunity. These immunodeficiency syndromes suggest that the characteristic inflammatory response of Crohn's disease may be a secondary compensatory response to a primary deficiency of innate immunity. Focused immune stimulation has been proposed as an alternate therapeutic paradigm. [347] Immune stimulation with G-CSF and GM-CSF was effective in pilot trials and further studies are anticipated.

Heparin emerged as a potential therapeutic agent from serendipitous observations of improvement of ulcerative colitis for patients being treated with heparin for thromboembolic disease. [348] Heparin has been proposed to be beneficial as an anticoagulant, helping reverse microvascular thrombotic disease responsible for an element of the pathophysiology of IBD. However, heparin also acts as an antiinflammatory agent as well as a potentiator of cellular repair. Controlled trials have demonstrated some conflicting data using standard heparin in ulcerative colitis. Lidocaine enemas appear beneficial in distal ulcerative colitis, studied in large open-labeled trials which have not yet been validated with placebo-controlled trials. [349] This approach suggests that manipulation of neural input may influence the inflammatory process and enable resolution of colitis. Rosiglitazone emerged as a potential therapy in ulcerative colitis through another serendipitous observation. Pilot studies suggested a positive therapeutic effect, presumably acting through the PPAR? pathway. [350]

MEDICAL MANAGEMENT OF ULCERATIVE COLITIS

Active Proctitis

Several reasonable approaches to the initial treatment of active ulcerative proctitis can be undertaken. [351] One relatively effective and rapidly acting approach is the nightly administration of 5-ASA (Rowasa) retention enemas or suppositories. If the patient responds, the frequency of the enemas or suppositories can be reduced from nightly to every other night after 2 to 3 weeks. 5-ASA enemas often are supplemented with oral sulfasalazine or an oral 5-ASA preparation. An acceptable alternative is the use of local corticosteroids in the form of retention enemas (Cortenema) or, for more limited disease rectal foam (Cortifoam or Proctofoam-HC) administered at bedtime for 2 to 3 weeks. If there is a good clinical response, this therapy is continued for another few weeks every other night. Either 5-ASA suppositories or corticosteroid foam is appropriate for disease involving up to 20 cm of distal colon; 5-ASA or corticosteroid retention enemas can be used for active disease involving up to 60 cm of distal colon. A third acceptable initial therapeutic approach to proctitis or distal colitis is an oral 5-ASA preparation. The response to sulfasalazine or other oral 5-ASA compounds may not be seen for 3 to 4 weeks, which is slower than the response to corticosteroid or 5-ASA enemas. Sulfasalazine is begun at a dose of 500 mg twice daily and is increased gradually every few days until a daily dose of 3 to 4 g is reached. Mesalamine (Asacol) and olsalazine (Dipentum) can be used instead of sulfasalazine, although Dipentum is not approved by the FDA for acute disease. The initial dose of Asacol is 800 mg three times a day; this dose can be increased up to 4.8 g/day if necessary. A metaanalysis suggests that mesalamine enemas are superior to steroid enemas or oral 5-ASA products in the management of left-sided colitis. The same metaanalysis found that mesalamine suppositories were superior to topical steroids in ulcerative proctitis. [351]

The patient with proctitis who does not respond to corticosteroid enemas can be given 5-ASA enemas, oral sulfasalazine, or oral 5-ASA. Conversely, patients who are not responsive to these therapies can be given corticosteroid enemas. Patients not responding to any of these modalities should be given oral corticosteroids; treatment with prednisone can be started with a dose of 40 mg/day. If there is a response, the dose is gradually tapered over 3 months. Patients who do not respond to oral corticosteroids (steroid-refractory) or who cannot be weaned off corticosteroids (steroid-dependent) can be given azathioprine or 6-MP, although there is usually a delay of several months before a clinical response is seen with either of these drugs.

For the patient who does not respond to corticosteroids or immunomodulators, surgery becomes a consideration. Loop ileostomies and temporary colostomies have been tried to give the colon a chance to heal. Whether a diverting ileostomy or colostomy has the potential to affect the course of the disease is not clear; however, diverting procedures do eliminate the diarrhea, urgency, and incontinence associated with proctitis. Some patients eventually require colectomy and an ileoanal anastomosis for proctitis, although this step is unusual. The creation of a diverting ostomy prior to a proctectomy reduces inflammation and thus reduces the technical difficulty of the proctectomy.

For patients with mild disease activity, whether with proctitis or more extensive disease, there is likely to be a role for symptomatic antidiarrheal therapy, usually with loperamide or diphenoxylate or opium and belladonna. They can be used to reduce the number of bowel movements or to reduce rectal urgency. Most patients use them on an as-needed basis after diarrheal stools, but some find it useful to take them with meals to reduce food-stimulated diarrhea.

After the proctitis is in remission, maintenance therapy should be considered. There is no compelling evidence that patients with a single, easily managed bout of proctitis need maintenance therapy; however, patients who have disease that is difficult to manage or with frequent or early relapses should have chronic maintenance therapy. The greatest experience with maintenance therapy is with sulfasalazine. The usual maintenance dose of sulfasalazine is 2 g/day; however, there is convincing evidence that maintenance with 4 g/day is more effective. [352] For patients who have undesirable side effects with sulfasalazine, maintenance therapy with olsalazine (1 g/day) or other oral 5-ASA formulations should be considered. If active disease is controlled with 5-ASA enemas, maintenance therapy with the same enemas may be appropriate. [353] Typically, maintenance therapy with 5-ASA enemas is given every other day or every third day. Because of the expense and inconvenience, maintenance therapy with 5-ASA enemas is reasonable only in patients who have failed maintenance therapy with sulfasalazine and other oral 5-ASA compounds.

Active Extensive Colitis

For patients with colitis of mild to moderate activity and extension proximal to the sigmoid colon, the initial drug of choice is either sulfasalazine or an oral 5-ASA compound. Typical doses are sulfasalazine 4 g/day, Asacol 2.4 g/day, and Pentasa 3 g/day. The efficacy of 5-ASA therapy increases with the dose, and it is reasonable to try doses higher than those typically used. Many patients tolerate gradual increases in sulfasalazine to a dose of 8 g/day or Asacol to a dose of 4.8 g/day. Not every patient responds to sulfasalazine or other 5-ASA formulations, however, and the response, if it occurs, may take 3 or 4 weeks. For patients with more active disease (more than five or six bowel movements per day) or in whom a therapeutic response is desired in less than 3 to 4 weeks, the initial treatment of choice is oral prednisone. This is also the drug of choice for those who have not respon- ded to 3 to 4 weeks of therapy with oral 5-ASA preparations. The typical starting dose of prednisone is 40 mg/day. Because of the side effects associated with high-dose prednisone there is a temptation to start therapy at a lower dose. This should be avoided as lower doses are associated with lower response rates. Most patients respond to oral corticosteroids within a few days to 3 weeks. After the symptoms are controlled, the dose of prednisone can be reduced gradually. One standard approach is to cut the daily dose by 5 to 10 mg every 1 to 2 weeks. It is usually relatively easy to reduce the corticosteroid dose to 20 mg/day, but the disease is more likely to flare as the dose is reduced further. It may be necessary to taper the dose more slowly below 20 mg/day or to use alternate-day dosing as part of the tapering protocol. Patients who respond to oral prednisone and who can be fully withdrawn from prednisone should be maintained on sulfasalazine or an oral 5-ASA preparation, these maintenance drugs should be started at least 3 to 4 weeks before prednisone is discontinued.

Severely Active Colitis

Some patients develop incapacitating disease that requires hospitalization. [101] The most common reason for hospitalization is intractable diarrhea, although blood loss is also a common problem. Incapacitating symptoms may develop acutely over a period of a few days or insidiously over weeks to months. Management with oral

corticosteroids usually has been tried before admission. Patients with severe active ulcerative colitis should be evaluated for toxic megacolon, the management of which is discussed in a subsequent section of this chapter. Anticholinergics and antidiarrheal agents are contraindicated in severe ulcerative colitis because of the risk of precipitating toxic megacolon. The mainstays of therapy for severe ulcerative colitis are bed rest, rehydration with intravenous fluids, and intravenous steroids (hydrocortisone 300 mg/day, prednisolone 60–80 mg/day, or methylprednisolone 48–60 mg/day). Patients with significant anemia should be transfused, and those with peritoneal signs or signs of systemic infection should be treated with parenteral antibiotics. Because active ulcerative colitis often is associated with a moderate leukocytosis and a low-grade fever, it may be difficult to identify those who have bacterial infection in addition to ulcerative colitis. If there is any doubt, cultures should be obtained, and the patient should be given broad-spectrum antibiotics such as ampicillin, metronidazole, and an aminoglycoside. Dehydration with or without hypokalemia occurs in those with severe diarrhea. Fluids should be administered aggressively to replenish water and electrolytes. The patient is given nothing by mouth, and nasogastric suction is used if there is an ileus or colonic dilation.

Maintaining adequate nutrition is often a major problem in severe active ulcerative colitis. Patients whose disease has been active for a considerable time may enter the hospital malnourished; these patients and those who are likely to be unable to take adequate oral nutrition for an extended period should be placed on TPN. If the bout of acute colitis has been of short duration, the patient's current nutritional status is good, and the history of previous exacerbations suggests that this one will be of short duration, then TPN is probably unnecessary. Patients with severe active ulcerative colitis should be given a trial of bed rest, TPN, and intravenous steroids for 7 to 10 days. Patients who do not improve during this period should be considered for either colectomy or a trial of intravenous cyclosporine, which is given at a dose of 4 mg/kg daily by continuous infusion, aiming for therapeutic serum levels. [325] For patients who do respond to cyclosporine, the average time to response is 7 days. If therapeutic success is achieved with intravenous cyclosporine, the patient is switched to oral cyclosporine (8 mg/kg daily).

Steroid-Refractory and Steroid-Dependent Disease

A small but troublesome group of ulcerative colitis patients includes those who do not respond to corticosteroids (steroid-refractory patients) and those who do respond but whose disease flares whenever the corticosteroids are withdrawn (steroid-dependent patients). In these cases, the clinician is faced with three potential courses of action: a trial of an immunomodulator; colectomy; and, if the patient has responded but cannot be withdrawn from corticosteroids, indefinite continuation of corticosteroids. The option of continued chronic corticosteroid therapy is frequently the least appealing of the three options because of the long-term side effects, including osteoporosis and cataract formation. Continuation of high-dose corticosteroid therapy for too long is the most common serious error in the management of ulcerative colitis. If the patient is on a substantial dose (>15 mg of prednisone per day) for longer than 6 months, a trial of an immunomodulator or surgery should be given serious consideration. The use of immunomodulators, usually 6-MP or azathioprine, is worth considering in many cases. For patients who have been on high-dose steroid therapy, the steroids are continued, and 6-MP (1–1.5 mg/kg daily) or azathioprine (2–2.5 mg/kg daily) is added. The combination of an immunomodulator plus steroids is continued for 3 to 4 months, and then the steroids are tapered gradually. If the steroids cannot be tapered after 6 months on immunomodulator therapy, the immunomodulator should be stopped. For patients who respond to an immunomodulator in this circumstance, the next question is how long they should be maintained on the drug and what happens if it is withdrawn. Although some clinicians attempt to withdraw the patient from immunomodulators after 1 to 3 years of therapy, most patients will have their disease flare within a few months of withdrawal. For this reason it is probably best to continue the immunomodulator indefinitely. The other option is colectomy. Given the risks of malignancy associated with long-standing ulcerative colitis and the risks associated with long-term immunomodulator therapy, the prospect of colectomy, especially if the creation of an ileoanal anastomosis is possible, should be offered as a realistic alternative rather than as an enforced consequence of failed medical management.

Maintenance Therapy

There is compelling evidence that oral 5-ASA preparations can reduce the incidence of recurrences in patients with ulcerative colitis. Most patients with ulcerative colitis should be viewed as candidates for chronic maintenance therapy. For the patient with a single episode of easily managed ulcerative colitis, it may be difficult to justify long-term maintenance therapy. In this case, the usual course is to have the patient on maintenance therapy for 1 year and then discontinue it. For patients who have difficulty controlling the disease or who have multiple recurrences, it is reasonable to recommend lifetime maintenance therapy. Sulfasalazine and other oral 5-ASA agents are probably equally effective. The efficacy of sulfasalazine as maintenance therapy in ulcerative colitis is a function of the dose; the efficacy of 3 to 4 g/day is greater than the efficacy of 2 g daily, even though 2 g daily is the usual recommended maintenance dose. [352] The dose of sulfasalazine should be pushed to the higher levels in patients who tolerate higher doses. Other oral 5-ASA agents can achieve higher lumenal concentrations of 5-ASA than usually are achieved with sulfasalazine and, on that basis, may be more effective as maintenance therapy. Asacol at 2.4 g/day is commonly used for maintenance therapy but higher doses may be more effective. Corticosteroids are not effective as maintenance therapy and should not be used. Most of the experience with 6-MP as maintenance therapy in ulcerative colitis is in patients whose acute disease has been brought under control with 6-MP; withdrawal of 6-MP from these patients has resulted in a high incidence of exacerbation.

COMPLICATIONS OF ULCERATIVE COLITIS

Perforation

Free perforation of the colon commonly complicates toxic megacolon but can occur in severe ulcerative colitis even without toxic megacolon. Perforation occurs more often during first episodes of colitis, perhaps because of lack of fibrosis and scarring from previous attacks. Most perforations occur in the left colon, particularly the sigmoid. Steroid therapy has been suggested to be a risk factor for colonic perforation, but some investigators have not found this to be the case.

Stricture

Clinically important strictures are relatively uncommon in ulcerative colitis, but some degree of narrowing may be seen in as many as 12% of surgical specimens. [354] Careful histological examination of strictures demonstrates hypertrophy and thickening of the muscularis mucosae without fibrosis. Strictures most often occur in patients with extensive disease and continuous symptoms without remission. They tend to appear later in the course of the disease, typically between 5 and 25 years after onset, most commonly in the sigmoid and rectum. Most strictures are short, typically 2 to 3 cm long, but they may be much longer. Symptoms associated with strictures include an increase in diarrhea and fecal incontinence. Strictures often are associated with malignancy in ulcerative colitis and should be viewed as potentially malignant lesions. Unfortunately, biopsy of strictures containing malignancy does not always yield malignant tissue. Development of a stricture in a patient with long-standing ulcerative colitis should be a source of concern and may necessitate resection because of the risk of malignancy.

Toxic Megacolon

If the inflammatory process extends beyond the submucosa into the muscularis, the colon loses its ability to contract and as a result becomes dilated. Dilation of the colon is associated with a worsening of the patient's clinical condition and the development of fever and prostration. A colonic diameter larger than 6 cm is sufficient to raise the possibility of toxic megacolon. Criteria for the clinical diagnosis of toxic megacolon, [109] include radiographic evidence of colonic distention (see Fig. 83-7) in addition to at least three of the four following conditions: fever higher than 38.6°C; heart rate greater than 120 beats per minute; neutrophil leukocytosis greater than 10,500 cells/mm ^{3}; or anemia. At least one sign of toxicity—dehydration, mental changes, electrolyte disturbance, or hypotension—also must be present. Physical examination of the patient with toxic megacolon reveals fever and postural hypotension. The abdomen is tender over the distribution of the colon, and there may be rebound tenderness. Colonic dilation causes abdominal distention. Bowel sounds are markedly hypoactive or absent because of loss of colonic motility.

Toxic megacolon usually occurs in patients with pancolitis but has been reported in patients with more limited disease. Patients are at greatest risk for developing toxic megacolon early in the course of their disease, and it can even occur as the initial presentation. In one series of 55 patients who developed toxic megacolon, 23 developed it within 3 months of the initial presentation of ulcerative colitis. [109]

Pharmacological agents that impair colonic motility are likely to initiate or exacerbate toxic megacolon. [355] Anticholinergic drugs diminish muscular tone in the colon and inhibit motility. Narcotic analgesics and antidiarrheals also inhibit the propulsive activity of the colonic musculature. Many cases of toxic megacolon are temporally associated with the initiation of therapy with anticholinergics or opiates or with increases in the doses of those drugs. [355], [356] For this reason, anticholinergic agents and antidiarrheals are contraindicated in patients with severe ulcerative colitis. Even in disease of moderate activity, these drugs should be used with caution, and the patient should be instructed to discontinue them if symptoms worsen. Toxic megacolon also has been temporally associated with barium enema examination and with colonoscopy. [357] Barium enema and colonoscopic examination are contraindicated in patients with severe ulcerative colitis; however, the patient can be examined safely by rigid proctoscopy or by limited flexible sigmoidoscopy as long as the physician takes care not to insufflate air into the already dilated colon.

Radiographic examinations are useful both for establishing the diagnosis and for following the course of toxic megacolon. X-ray films of the abdomen reveal colonic dilation, usually maximal in the transverse colon, which exceeds 6 cm in diameter. Although the transverse colon may not be the site of the most severe disease, it is usually the most dilated segment, because colonic gas tends to accumulate in the highest portion of the colon, and the transverse colon is the highest portion when the patient is supine. Serial flat-plate films of the abdomen taken at 12- to 24-hour intervals are useful in following the clinical course.

Although laboratory findings are not diagnostic in toxic megacolon, a pattern of laboratory abnormalities is commonly seen. Patients with toxic megacolon may be anemic because of blood loss, and there is usually a leukocytosis with predominance of neutrophils. Hypokalemia and hypoalbuminemia as a result of losses from diarrhea also are seen.

Medical therapy is designed to reduce the likelihood of perforation and to return the colon to normal motor activity as rapidly as possible. The patient is given nothing by mouth, and a nasogastric tube is placed in the stomach. Some clinicians use a longer tube, such as a Miller-Abbott or Cantor tube, but there is no evidence to suggest that these tubes are more efficacious than a nasogastric tube. Intravenous fluids should be administered to replace water and electrolytes. [358] Fluid and electrolyte disorders impair the normal motility of the colon and increase the risk of perforation from toxic megacolon. Frequently, large deficits of water, sodium, chloride, and, especially, potassium occur. Patients with toxic megacolon often receive broad-spectrum antibiotics in anticipation of peritonitis resulting from perforation. The value of corticosteroids in the treatment of toxic megacolon has not been established in controlled studies; however, most clinicians treat patients who have the condition with parenteral corticosteroids at a dose equivalent to more than 40 mg of prednisone per day. [359], [360] Medical therapy for toxic megacolon is most likely to be successful if the condition is recognized early and appropriate steps are taken. The patient should be monitored carefully. Signs of improvement include a decrease in abdominal girth and the return of bowel sounds. Deterioration is marked by the development of rebound tenderness, increasing abdominal girth, and cardiovascular collapse.

If medical therapy is successful during the first 24 to 48 hours, both an improvement in the signs of clinical toxicity and a decrease in the diameter of the dilated colon on radiographic examination should be seen. Persistence of fever after 48 hours on high-dose steroids suggests perforation or abscess. If the patient does not begin to show signs of clinical improvement during the first 24 to 48 hours of medical therapy, the risk of perforation increases markedly, and surgical intervention is indicated. [361] The most common procedure is colonic resection with creation of an ileostomy. The rectum may be left in place and removed at a later date; however, single-stage proctocolectomy is done more commonly. The key steps in management are early recognition of the condition and identification of patients who do not respond to medical therapy. Surgical therapy after perforation is likely to have a poor outcome. [362] Mortality in that group is 44%, compared with 2% in those who were operated on before perforation. Among patients with toxic megacolon, many who are successfully treated medically are likely to undergo colonic resection for intractable disease within a year. Toxic megacolon is a sign of severe disease that may not respond to medical therapy over a long period. One study followed the clinical course of 38 patients who had been successfully treated medically for toxic megacolon; 18 eventually underwent colectomy. [363]

MEDICAL MANAGEMENT OF CROHN'S DISEASE

General Approach

It is more difficult to develop generally applicable guidelines for the management of Crohn's disease compared with ulcerative colitis. Crohn's disease involves a greater variety of anatomic locations and has more varied clinical presentations, more gastrointestinal complications, such as fistulae, abscesses, strictures, and perforations, more insidious onset and recovery from attacks; and frequently lacks definable therapeutic end points. The guidelines given here are for active disease without complications; management of specific complications is discussed separately. Adequate rest is essential, and a few days of enforced rest (or a few hours of additional rest each day) may speed up the resolution of a flare. Patients with Crohn's disease who smoke should be strongly encouraged to stop. Many practitioners believe continued smoking reduces the likelihood of response to medical management.

Symptomatic therapy plays an important role in the management of mild Crohn's disease. Antidiarrheal agents can reduce the number of bowel movements in patients with mild disease activity or in those with diarrhea secondary to surgical resections, bile salt malabsorption, or fat malabsorption. Loperamide and diphenoxylate are the antidiarrheal agents most commonly prescribed; however, opium and belladonna should be considered if loperamide and diphenoxylate are ineffective. Antispasmodics can reduce the pain associated with Crohn's disease but are contraindicated in patients with obstruction or near obstruction. Appropriate symptomatic therapy can reduce steroid requirements; however, antispasmodics and antidiarrheal agents should be used with caution in patients with severe disease because of the risk of precipitating toxic megacolon. Psychotropic drugs, particularly tricyclic antidepressants, can be useful adjunctive therapy in Crohn's disease. The incidence of depression is high among patients with Crohn's disease, and the manifestations of depression can make management of Crohn's disease more difficult. A trial of a tricyclic antidepressant or a selective serotonin reuptake inhibitor (SSRI) is indicated in patients who have clinical manifestations of significant depression.

A common problem in the management of Crohn's disease is a marked discrepancy between the severity of the patient's symptoms and the objective signs of disease. Patients with severe pain and diarrhea may have minimal findings on endoscopy or radiographic studies. In these cases, several possible explanations may need to be considered. Patients who have undergone ileal resections may have significant diarrhea on the basis of their surgery alone. Removal of the ileocecal valve speeds transit and can increase the number of bowel movements. Surgical removal of the distal ileum can result in failure to reabsorb bile acids, which induce colonic chloride secretion, leading to diarrhea. More extensive ileal resections may result in enough bile salt malabsorption to decrease the size of the bile salt pool, resulting in fat malabsorption. Malabsorbed fatty acids can be hydroxylated by colonic bacteria; hydroxyfatty acids also induce colonic chloride secretion and contribute to diarrhea. Crohn's disease patients with a history of ileal resection who present with diarrhea should have a trial of a low-fat diet (75 g/day or less) to determine how much of their diarrhea is a product of their surgery rather than a product of active Crohn's disease. A few patients with bile salt–induced diarrhea also may need cholestyramine.

For medical therapy, response is monitored by empiric clinical assessment directed at the problem that is most troublesome for the patient. If the major complaint is pain, the success of therapy is assessed by the severity of pain; if diarrhea is the major problem, success is measured by the frequency of diarrhea. In assessing the patient's response to drug therapy, it is important to understand the basis of the chief complaint. If abdominal pain is a result of obstruction behind a fibrotic stricture, it is not reasonable to expect corticosteroids to relieve the pain. Similarly, if diarrhea is caused by small bowel overgrowth behind a stricture, it is unlikely that corticosteroids will improve the diarrhea.

Mildly to Moderately Active Disease

For colonic or ileocolic Crohn's disease with mild to moderate activity, an oral 5-ASA formulation is a reasonable first approach to therapy. The dosage and treatment schedules are the same as those described for ulcerative proctitis or ulcerative colitis of mild to moderate activity (see Table 83-7). Pentasa, an oral 5-ASA preparation with greater availability of 5-ASA in the ileum than sulfasalazine, may be a better choice for patients with ileitis or ileocolitis. In a controlled trial, Asacol was as effective as methylprednisolone in the treatment of active Crohn's ileitis. [364] An alternative to 5-ASA preparations is treatment with antibiotics. Ciprofloxacin (500 mg bid) and/or metronidazole (10–20 mg/kg day) can be used a first-line therapy. A disadvantage of the 5-ASA preparations and antibiotics is that a clinical response may not be seen for 3 to 4 weeks.

Oral prednisone may be used as first-line therapy for patients with ileal disease or for patients with colonic or ileocolic disease with higher levels of disease activity. Prednisone is also the drug of choice for patients who have failed to respond to 5-ASA preparations or antibiotics. The response to prednisone is usually seen in a few days, which is more rapid than that to 5-ASA formulations or antibiotics. In a patient with Crohn's disease with abdominal pain, fever, and a high leukocyte count, an abdominal CT scan should be obtained to rule out the presence of an abscess before corticosteroids are given. Corticosteroid therapy in a patient with Crohn's disease with an abscess, especially in the absence of antibiotic therapy, can lead to serious septic complications. The most common clinical indication for the use of prednisone in Crohn's disease is the presence of an ileal narrowing caused by mucosal inflammation. If the patient's ileum is only partially obstructed, he or she is managed as an outpatient with bed rest, a low-residue diet, and oral prednisone at dose of 40 mg/day. If the symptoms come under control, the dose of prednisone is tapered as described in previous sections. If disease activity flares as the dose of prednisone is tapered, the possibility of surgery or a trial of immunosuppressives should be considered.

A frequently troublesome area is management of the patient who has been brought into clinical remission on steroids. The general goal is to taper and discontinue the steroids without inducing a flare of activity. The rate at which the steroid dose is tapered is arbitrary, and no studies have attempted to define optimal tapering schedules. The rate of taper is influenced by the patient's response to steroid tapers in the past and by the side effects of the steroids. Those who have tolerated rapid tapers in the past are likely to tolerate rapid tapers again. Many patients have side effects with high doses (40 mg/day) of prednisone. These side effects, which include insomnia, agitation, and blurred vision, may necessitate more rapid tapering. Usually the prednisone dose can be tapered from 40 mg daily to 20 mg daily

relatively rapidly (a 5–10 mg reduction every 1 to 2 weeks) without inducing a flare of disease activity. Among patients with Crohn's disease receiving their first course of steroids, 20% do not respond (steroid-resistant), 36% respond but cannot be withdrawn from therapy (steroid-dependent), and 44% respond and remain in a prolonged response after steroids are withdrawn. [365] There is a clinical pattern to patients likely to become steroid-dependent. They are more likely to have a younger age at diagnosis, to be smokers, to have colonic disease, and to have inflammatory rather than fibrostenotic disease. [366]

If the patient has not been on a 5-ASA preparation, one should be added to increase the likelihood of a successful steroid withdrawal. The 5-ASA preparation should be added by the time the prednisone dose is reduced to 20 mg/day. Pentasa (3 g/day) for ileal disease or Asacol (2.4 g/day) for colonic or ileocolic disease is appropriate for maintenance therapy. Metronidazole or ciprofloxacin can be added to enhance the likelihood of a successful steroid taper, especially if the patient has a history of responding to antibiotics in the past.

Once the dose of prednisone has reached 20 mg per day, the taper is slowed to 5 mg every 10 to 14 days. Symptoms often flare as the prednisone dose is tapered; these flares usually are due to a continued high level of intestinal inflammation that persists despite apparent clinical remission. When symptoms flare, the dose of prednisone is increased. At this point, several approaches are possible. Continuing steroid therapy indefinitely is the least desirable approach; the long-term side effects of even modest doses of steroids are likely to be significant. For a few patients, surgery is a reasonable approach at this point. It is a more attractive option if the area of involvement is small or if there are features (e.g., strictures) that increase the appropriateness of a surgical approach. For most patients, the best approach at this point is a trial of an immunomodulator, either 6-MP (1–1.5 mg/kg daily) or azathioprine (2–2.5 mg/kg daily). Treatment with the immunomodulator is initiated and steroid therapy continued for 3 to 4 months, and then the dose of steroids is tapered gradually. About 60% of steroid-dependent patients will be able to withdraw from steroids using this approach. If steroid withdrawal is tolerated, the patient is continued on the immunomodulator indefinitely. If the clinical activity flares when the dose of steroids is tapered in patients receiving 6-MP or azathioprine, the dose of the immunomodulator should be increased. Blood counts need to be monitored carefully as the risk of leukopenia increases with increasing doses of 6-MP or azathioprine. An alternative to increasing the dose of 6-MP or azathioprine would be to substitute methotrexate (25 mg parenterally once a week). Although there are data to support the use of methotrexate as a steroid-sparing agent in Crohn's disease, there is much less experience than with 6-MP or azathioprine. For patients who fail to respond to immunomodulators, the options are surgery or long-term steroids. A significant number of patients have allergic or idiosyncratic reactions to immunomodulators. Those who have had an idiosyncratic reaction, such as pancreatitis, or an allergic reaction to either 6-MP or azathioprine should not be tried again on either drug but could be given methotrexate.

Another therapeutic approach is the use of infliximab although there is considerable uncertainty as to the circumstances in which it is best used. In patients who are steroid-resistant or steroid-dependent it may be reasonable to use infliximab and start therapy with 6-MP or azathioprine at the same time. The expectation would be that the response to one or two infusions of infliximab would keep the disease under control until the response to 6-MP or azathioprine was established at 3 or 4 months. Infliximab can also be used in patients who are steroid-resistant or steroid-dependent and who have had an adverse reaction to 6-MP or azathioprine. If these patients respond to infliximab, the next question is how many times can infliximab be infused before either the efficacy is lost or adverse reactions develop? Although there is little experience with the long-term use of infliximab some patients have received five infusions over 36 weeks and stayed in remission. [340] The safety and efficacy of repeated infusions of infliximab will need to be addressed in future long-term studies. Similarly the safety and efficacy of the combination of infliximab with 6-MP and azathioprine or with corticosteroids will need to be addressed.

Severely Active Disease

Severe disease refers to patients who have persisting symptoms despite the introduction of oral corticosteroids or patients presenting with abscess, obstruction, high fever, vomiting, rebound tenderness, or cachexia. The approach to severe Crohn's disease is similar to the approach to severe ulcerative colitis. The patient is hospitalized, given nothing by mouth, rehydrated with intravenous fluids, and given parenteral steroids in the dosages listed in the previous section. Patients who respond to parenteral corticosteroids are switched to oral corticosteroids (prednisone 40 mg/day), and the dose is gradually reduced. For patients who require moderate to high doses of corticosteroids for a long time (15 mg of prednisone per day for 3 to 6 months) to control disease activity, a trial of immunosuppressives may be indicated. Patients with severe Crohn's disease who do not respond to parenteral steroids within a week should be considered for surgery. Particular efforts should be made to avoid surgery if possible in patients with previous extensive small bowel resections to avoid the risk of short bowel syndrome. A course of TPN may be useful as adjunctive therapy.

Maintenance Therapy

Therapy in IBD traditionally has been divided into therapy for active disease and maintenance therapy. The suggestion is that patients on maintenance therapy are in remission and do not have active disease. For Crohn's disease and, to a lesser extent, ulcerative colitis, the distinction between therapy for active disease and maintenance therapy is false. It is more reasonable to consider that all patients with Crohn's disease have active disease; that is, patients in clinical remission have activity even though it is not clinically apparent.

Patients with Crohn's disease who are in clinical remission tend to relapse over time. There have been at least a dozen trials of 5-ASA preparations for the maintenance of remission of Crohn's disease. [367], [368], [369], [370] and [371] Most of these trials have demonstrated positive results and metaanalysis has demonstrated efficacy. However, one large (318 patients) prospective randomized double-blind trial failed to find any benefit of 4 g of mesalamine compared with placebo in preventing relapse after surgical treatment of Crohn's disease. [372] In the studies that found 5-ASA compounds to be effective in maintaining remission, there were approximately 50% reductions in the rate of relapse over the course of the studies, which typically lasted 1 or 2 years. The efficacy of 5-ASA compounds for maintaining remission in Crohn's disease over longer times has not been addressed. Data are not available to make firm recommendations about the choice of preparation and doses for maintenance. The dose needed presumably will be proportional to the degree of continued inflammation present in patients in clinical remission. The trials suggested that 5-ASA maintenance therapy was more effective in patients with ileitis alone and more effective in patients whose remission was achieved surgically rather than medically. As in the treatment of active disease with 5-ASA compounds, higher doses of 5-ASA appeared to be more effective in maintaining remission. Pentasa (3 g/day) for patients with ileitis and Asacol (2.4 g/day) for patients with colonic or ileocolic disease are reasonable choices for maintenance therapy. There are some practical problems in using 5-ASA preparations for maintenance in Crohn's disease. The 5-ASA preparations, as a group, are expensive and require multiple dosing; moreover, the benefits of maintenance therapy in Crohn's disease are modest. For these reasons, some patients may not be motivated to participate in a 5-ASA maintenance program.

Although it is commonly stated that there is no role for corticosteroids in maintenance therapy in Crohn's disease, patients we categorize as steroid-dependent are being maintained in clinical remission with steroids. It may be more useful to state that there is no role for long-term, high-dose steroids in the management of Crohn's disease and that patients who require long-term steroids to keep their disease clinically inactive should be offered other therapies.

Many patients who are brought into remission with immunomodulatory agents, particularly azathioprine and 6-MP, are maintained on these drugs for years. For patients who have been brought into remission with azathioprine or 6-MP, and who tolerate those drugs, maintenance therapy is justified.

There is limited experience with antibiotics or fish oil as maintenance therapy in Crohn's disease. One study demonstrated a benefit of 3 months of metronidazole immediately after a surgical resection in preventing recurrence. [373] Two studies investigated the use of fish oil supplementation for maintenance of remission in Crohn's disease. One study demonstrated efficacy, [240] and the other did not. [241]

In brief, maintenance with 5-ASA preparations is recommended for those brought into remission on steroids or with surgery, though the benefit is limited. Maintenance with 6-MP or azathioprine is recommended for patients brought into remission on those drugs or who were steroid-dependent and could be converted to those drugs. Maintenance therapy may have increased importance in patients who have a history of multiple surgical resections or particularly short periods of remission.

COMPLICATIONS OF CROHN'S DISEASE

Abscesses and Fistulae

Abscesses and fistulae are both products of the extension of a mucosal fissure or ulcer through the intestinal wall and into extraintestinal tissue. Leakage of intestinal contents through a fissure into the peritoneal cavity results in an abscess. Extension of the inflammatory process through the wall of adjacent viscera or through the abdominal wall to the exterior results in a fistula.

Abscesses occur in 15% to 20% of patients with Crohn's disease. [374] They can arise from any affected area, but the terminal ileum is an especially likely point of

origin. If confined to the abdominal cavity, abscesses form between loops of intestine within the mesentery or between the intestine and the parietal peritoneum. Abscesses also extend into the iliopsoas and retroperitoneal regions. One pattern is extension of an abscess from the terminal ileum to the right iliopsoas; such a lesion presents as pain on flexion of the right hip, which can be mistaken for sciatica. [375] Hepatic and splenic abscesses also occur. Abscesses may develop postoperatively at the site of an anastomosis after resection. Development of an abscess soon after resection usually results from an anastomotic leak, whereas development of an abscess several months after surgical resection is a result of disease recurrence. [376] The typical clinical presentation of intra-abdominal abscess associated with Crohn's disease is fever and abdominal pain. The location and quality of the pain are determined by the location of the abscess. Tenderness and abdominal mass may accompany the pain and fever. Leukocytosis is the most common laboratory abnormality. Spontaneous rupture of an abscess through the abdominal wall can occur and results in an enterocutaneous fistula.

Abdominal abscess is most often diagnosed by CT scan, but barium enema, ultrasonography, and radionucleotide scanning with gallium 67 (^{67}Ga) are also useful. [377] , [378] and [379] The frequently encountered organisms in Crohn's abdominal abscesses are *E. coli, Bacteroides fragilis,* enterococcus, and hemolytic *Streptococcus* species. [378] Many different organisms may be found in a single abscess. Broad-spectrum antibiotic therapy, including anaerobic coverage, is indicated. Antibiotic coverage should be adjusted on the basis of results of culture of blood and abscess contents. Corticosteroids are not effective in the treatment of these abscesses and may impair the host response to infection. If the patient has not been taking steroids, they should not be started until the infection has been brought under control with drainage and antibiotics. If the patient has been taking steroids and the dose is being tapered, higher doses may be required to deal with the stress of infection.

Simple drainage of abscesses in patients with Crohn's disease may not provide adequate therapy because of persistent communication between the abscess cavity and the intestinal lumen. In fact, drainage in such circumstances commonly results in the formation of an enterocutaneous fistula. Resection of the portion of involved intestine containing the communication usually is required for definitive therapy. First, the abscess is drained percutaneously under guidance by ultrasound or CT scan. After the abscess is adequately drained and inflammation is reduced, the involved segment of bowel is resected. The site of communication is not always obvious and radiographic examination after oral administration of contrast material or injection of contrast into the abscess cavity may help in its identification.

The prevalence of fistulae is 20% to 40% in Crohn's disease. [374] Most fistulae are enteroenteric or enterocutaneous, with smaller numbers that are enterovesical or enterovaginal. Irrespective of the location, the mechanism of fistula formation appears to be the same. A deep abscess penetrates through the intestinal wall and into an adjacent organ or the skin. Enteroenteric fistulae develop either between a loop of involved bowel and a loop of uninvolved bowel or between two loops of involved bowel. The terminal ileum is the segment most commonly involved; fistulae from the terminal ileum may extend to other loops of small intestine or to the sigmoid. Fistulae develop when the disease is active but frequently persist even after the disease is no longer active. Enteroenteric fistulae by themselves seldom cause significant symptoms and often are found incidentally by barium contrast studies. [380] Most enteroenteric fistulae are of small diameter and do not have significant flow rates; however, those of larger diameter may allow a flow great enough to cause malabsorption, diarrhea, and weight loss.

Pain and diarrhea in patients with enteroenteric fistulae usually are caused by active Crohn's inflammation and not by the mere presence of the fistula. The treatment of fistulae in Crohn's disease has been reviewed. [381] Patients should be treated as they would be in the absence of a fistula; asymptomatic fistulae require no treatment. If an enteroenteric fistula is thought to be responsible for significant symptoms, administration of TPN or immunomodulator therapy may induce fistula closure; [382] , [383] however, fistulae often recur after the TPN or immunomodulator is stopped. Korelitz and Present [382] performed a double-blind, placebo-controlled study using 6-MP at a dose of 1.5 mg/kg/day to close fistulae. Of the 29 patients who received 6-MP, 9 completely healed their fistulae, compared with 1 of 17 in the placebo group. The mean time required for healing with 6-MP was 3 months; the fistulae reopened after 6-MP was stopped. Corticosteroids are not effective in closing fistulae. Infliximab is effective in closing enterocutaneous fistulae. Three doses (0, 2, and 6 weeks) of infliximab at 5 mg/kg resulted in the closing of most fistulae in 68% of patients and of all fistulae in 55%. [337] The median duration of closure was 3 months. Whether fistulae closed on treatment with infliximab would stay closed if the patient were treated indefinitely with azathioprine or 6-MP has not been tested. Although nutritional and medical therapy may play a role in the management of some fistulae in Crohn's disease, the problem of recurrence with the discontinuation of these treatments leaves surgery as the major therapeutic modality. [384] , [385] and [386] Surgical therapy includes resection of the segment involved with active disease. If both loops are involved, both need to be resected; if only one is involved, it should be resected and the other oversewn. Fistulae commonly develop in the high-pressure zone proximal to a stricture. Successful management of the fistula and prevention of recurrence are more likely if the stricture is resected with elimination of the high-pressure zone.

Enterocutaneous fistulae are more often troublesome than enteroenteric fistulae. They commonly occur as a result of anastomotic leaks after resections for active disease. [387] In these cases, the cutaneous end of the fistula is in the scar, and the enteric end is at the anastomosis. Enterocutaneous fistulae also occur spontaneously in the absence of previous surgery. Although the presence of an enterocutaneous fistula does not constitute an absolute indication for surgery, most patients find significant persistent drainage through the abdominal wall to be intolerable. The decision in favor of surgical therapy depends on the amount of drainage, the extent of disease activity, and the patient's nutritional status. More than one cutaneous opening may be present, but usually the enteric ends of the fistulae are close together, and the amount of bowel requiring resection may be no more than if only one cutaneous opening were present.

Rectovaginal fistulae are seen most commonly in active rectal Crohn's disease. [388] Smaller fistulae cause only a foul vaginal discharge, but larger fistulae result in the passage of gas and stool through the vagina. Rectovaginal fistulae usually can be identified by proctoscopy or by speculum examination of the vagina; however, some fistulae are apparent only on barium enema. If the symptoms are minimal, no therapy is necessary. Metronidazole leads to healing in some patients; [389] , [390] however, definitive therapy usually requires surgical intervention. [391] Primary closure has been successful in some patients, whereas diverting colostomy or abdominal perineal resection has been necessary in others. Enterovesicular fistulae usually involve diseased segments of ileum or sigmoid. [152] Signs include gas in the urine (pneumaturia) and recurrent urinary tract infections. Diagnosis may be made by barium enema, upper gastrointestinal series with small bowel follow-through, cystoscopy, or intravenous pyelogram. Definitive surgical management is usually recommended, especially if there are recurrent urinary tract infections, because of the risk of irreversible kidney damage. Surgical therapy includes resection of the involved segment of bowel. The bladder defect may be closed primarily or, more rarely, resected.

Obstruction

Obstruction is a common complication of Crohn's disease, particularly in the small intestine, and is a leading indication for surgery. [116] , [392] Small bowel obstruction in Crohn's disease may be caused by mucosal thickening from acute inflammation, by muscular hyperplasia and scarring as a result of previous inflammation, or by adhesions. Obstruction also may occur because of impaction of a bolus of particularly fibrous material in a stable, long-standing stricture. Obstruction presents with cramping abdominal pain and diarrhea that worsen after meals and resolve with fasting. Symptoms worsen as the obstruction tightens. A common history is that of intermittent episodes of mild cramps and diarrhea, followed by prolonged episodes of severe pain and diarrhea accompanied by nausea and vomiting. The prolonged episodes of symptoms may be brought on by fibrous meals. It is important to distinguish obstruction caused primarily by mucosal inflammation and edema from obstruction caused primarily by scarring because medical therapy may benefit inflammation but will have no effect on scarring. Such a distinction is often difficult, as both processes are usually present, and response to therapy will be determined by whichever effect is predominant. Patients with obstruction caused by inflammation usually have a prolonged course of gradually worsening symptoms: although the symptoms may vary in severity over time, they do not resolve entirely. In contrast, the patient with obstruction caused by scarring may feel well except when totally obstructed. Depending on the anatomic location, strictures may be evaluated by oral contrast studies, barium enema, or colonoscopy. Evaluation of strictures includes assessment of location, length, and lumenal diameter. Dilation of the intestine proximal to the stricture suggests chronicity. There is often an apparent discrepancy between the tightness of the stricture as assessed radiographically and the severity of the patient's complaints. Some patients have minimal complaints with near-obstructing strictures, but others have severe complaints with only modest narrowing. Lesions that block the passage of food particles may allow liquid contrast agents to pass freely; as a result functionally significant strictures and adhesions may be completely missed by radiographic studies.

Initial therapy is to give nothing by mouth, to apply nasogastric suction, and to give intravenous fluids. Anticholinergics should not be used in patients with obstruction. If acute inflammation is an important component of the obstructive process, parenteral corticosteroids may help; however, corticosteroids are not useful in the management of fibrotic strictures. A common error in the management of Crohn's disease is to treat patients with obstructive symptoms from fixed anatomic lesions with long courses of corticosteroids. If the obstruction does not resolve with nasogastric suction and corticosteroids, dilation or surgery is necessary. If the fibrotic stricture is short and accessible, endoscopic balloon dilation can be attempted; more likely, surgical intervention, either resection or stricturoplasty, will be required. [393]

Perianal Disease

Perianal disease is an especially difficult complication of Crohn's disease. [394] , [395] Patients with IBD are subject to the same perianal problems as the general

population, primarily hemorrhoids and anal fissures. Patients with Crohn's disease also may have perianal disease that is specifically associated with Crohn's disease. Although perianal disease can be seen with Crohn's disease anywhere in the gastrointestinal tract, it is seen more commonly in association with colonic disease. Patients with Crohn's disease also develop a complex of problems marked by ulcers in the anal canal which result in perirectal abscesses or fistulae that extend from the ulcers. The fistulous openings are most commonly in the perianal skin but can be in the groin, the vulva, or the scrotum. A single rectal ulcer can give rise to a fistulous tract with multiple openings. The mucosal ends of fistulae often arise from anorectal glands. There are 10 to 12 anorectal glands at the level of the dentate line (see Chapter 92) that extend through the internal sphincter to the intermuscular plane. Two of these glands, one in the anterior midline and one in the posterior midline, are the source of most fistulae and rectal abscesses. These fistulae extend into the intersphincteric space and from there can extend in a variety of directions following tissue planes (Fig. 83-27).

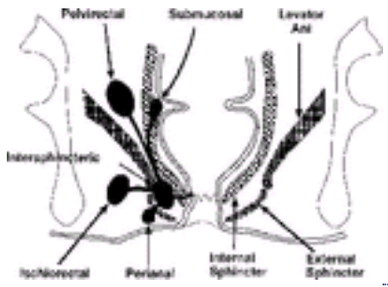

FIGURE 83-27. In Crohn's disease perirectal abscesses develop from fistulous tracts that begin in rectal ulcers of anorectal glands. These fistulae extend into the intersphincteric space and then follow tissue planes to a variety of pelvic locations where abscesses may form.

Fistulae present with drainage of serous or mucous material. If the fistula does not drain freely, there is local accumulation of pus and abscess formation with redness, pain, and induration. Perianal abscesses present with pain that is exacerbated by defecation, sitting, or walking. The typical presentation of abscess is redness and pain in the perianal region with tenderness on digital examination; however, supralevator and pelvirectal abscesses may present with rectal pain or have no local manifestations and may present with fever alone. Usually, perirectal fistulae and abscesses are seen in conjunction with disease activity, but they may occur in the absence of other symptoms and may be the initial presentation of the disease.

Adequate assessment of perianal disease usually requires proctoscopic examination under anesthesia. Barium enema may reveal the course of fistulae. CT scans are useful in defining the presence and extent of perianal abscesses. Endoscopic ultrasound [396] and MRI [397] may be useful in characterizing fistulae. Defining the extent of perianal disease is important for therapy and prognosis. Simple fistulae are low lying and have one external opening. Simple fistulae frequently heal with minimal surgical intervention. Typically, the only surgery required with simple fistulae is a fistulotomy. More complex fistulae and rectovaginal fistulae may require more extensive surgical intervention and have a worse prognosis. Persistent severe perianal Crohn's disease can result in destruction of the anal sphincter and fecal incontinence.

The wide variety of presentations and locations of perianal disease requires individualized therapy. For complex disease, surgical and medical therapy must be combined and coordinated. The goals of therapy in perianal disease are relief of local symptoms and preservation of the sphincter. Abscesses must be surgically drained. The mere presence of a fistula does not require therapy; if it is not causing problems, it does not need to be treated. The aggressiveness of therapy is determined by the severity of the perianal disease. Careful local cleansing with sitz baths is an important first step in management. Limited disease can be approached with sitz baths and antibiotics, but in most cases, adequate external drainage is also required. Setons or drains in the fistulous tracts allow for continuing drainage. Optimal management of perianal complications includes control of disease activity, which reduces the amount of diarrhea passing through the perianal area. Control of disease activity is a more important issue when both perianal disease and rectal disease are present. When active rectal disease is present, effort should be made to minimize disease activity with medical management before any consideration is given to further surgical intervention for perianal disease.

Metronidazole plays a role in the management of perianal disease. Bernstein and colleagues [389], [390] treated perianal Crohn's disease with metronidazole at a dosage of 20 mg/kg/day in divided doses; complete healing occurred in 45% of patients and improvement without complete healing in another 23%. Other investigators also have had success with metronidazole but with a lower response rate. [398] Prolonged therapy at this dosage is associated with a high rate of side effects, of which peripheral neuropathy is the most troublesome. Numbness and tingling in the feet are seen in as many as 75% of patients after 3 months of therapy. [398], [399] Paresthesias usually resolve if the dose is reduced or the drug discontinued. To minimize side effects, particularly paresthesias, initiate therapy at a lower dosage (250 mg four times daily). Discontinuation of metronidazole therapy results in recurrence of perianal disease in most patients, even if complete healing has been achieved. Despite these problems, a trial of metronidazole is recommended if the patient still has significant symptoms after adequate surgical drainage of perianal disease. The experience with ciprofloxacin is less than with metronidazole, but one small series reported success with 1000 to 1500 mg/day for 3 to 12 months. [400] If the response to antibiotics is inadequate, azathioprine or 6-MP can be tried. There is a high rate of success in closing fistulae with these immunomodulators [401] although the time to response is long. Fistulae can also be closed with infliximab. [337] Fistulae recur when the drugs are stopped, and the results of long-term therapy are unknown.

If simple drainage is unsuccessful, there are a number of possible surgical approaches to perianal disease. Many patients respond well to careful surgical drainage and placement of setons and mushroom catheters, which can be left in place for months as the perianal disease slowly heals. Partial internal anal sphincterotomy can be used to remove the cryptoglandular epithelium from which the fistulae arise. [402] An alternative approach is to marsupialize all the fistulous tracts and lay open the crypt of origin. Fecal diversion by colostomy has also been used; however, the general experience is that fecal diversion does not lead to healing of perianal disease. [403], [404]

SURGICAL MANAGEMENT (see also Chapter 84)

Ulcerative Colitis

About 20% to 25% of patients with extensive ulcerative colitis eventually undergo colectomy, usually because their disease has not responded adequately to medical therapy. [405], [406] For some, the indications for colectomy are urgent and compelling, but more commonly the decision for or against colectomy is made at a time when the disease is active but stable. The decision between surgery and continued medical therapy is often not clear, and in many cases arguments can be made for either course. It is important in these cases to attempt to balance the risks and benefits of the two approaches. Excessive enthusiasm for or avoidance of operative therapy are both inconsistent with the patient's best interests. At the time of first diagnosis, the patient may reject even the consideration of eventual colectomy. However, a single prolonged episode of severe colitis or multiple recurrent episodes of more moderate severity increase the patient's desire to eliminate this debilitating disease with its restrictions on activity, and the prospect of colectomy becomes less intolerable.

In ulcerative colitis, colectomy is a curative procedure, in contrast to the situation in Crohn's disease, in which there is a significant likelihood of recurrence sometime after colectomy. [407] The assurance of a permanent cure increases the appeal of colectomy in ulcerative colitis. The development of the ileoanal anastomosis, eliminating the need for an ostomy, has made the idea of colectomy more tolerable for many patients. The patient's age, social circumstances, and duration of disease all influence the decision for or against colectomy. The morbidity of severe ulcerative colitis in childhood and the morbidity of corticosteroids in childhood combine to lead to the early consideration of colectomy as definitive therapy. For young adults with moderately severe disease, colectomy may be more acceptable for those in stable marriages than for those who are single. The risk of developing malignancy enters into the equation when considering colectomy in those with long-standing ulcerative colitis; if the other indications are equivocal, the risk of malignancy may tilt the balance in favor of colectomy.

Emergency colectomy may be required in toxic megacolon or in a severe fulminating attack without toxic megacolon. [408] Although colectomy for acute attacks of ulcerative colitis is more commonly associated with toxic megacolon, it is clear that acute fulminating ulcerative colitis in the absence of colonic dilation also can proceed to perforation. The goal is to recognize the necessity for surgical intervention before perforation occurs. If perforation does occur, it is an absolute indication for surgery. Perforation in this circumstance is a medical disaster, with a 40% mortality rate. [409] For fulminant colitis, in the absence of toxic megacolon, the general indications for operation are similar. Before the patient undergoes colectomy for intractable ulcerative colitis, it is important to rule out complicating conditions that can

be treated. The possibilities of infection with *Giardia* organisms, amoebae, *C. difficile,* and cytomegalovirus should be considered and, if indicated, testing for these organisms should be done. The presence of worsening colitis and especially worsening systemic signs (e.g., fever, prostration, tachycardia, hypotension) in the face of aggressive medical therapy is a sign that surgical intervention may be indicated. Severe hemorrhage is another indication for emergency colectomy.

The failure of medical therapy to control the activity of ulcerative colitis, resulting in an unacceptable lifestyle, is the most common indication for colectomy in ulcerative colitis. There are several varieties of intractability. In one variety, the disease enters a chronic continuous phase in which the patient no longer achieves remission even with maximal medical therapy, and chronic diarrhea is severe enough that the quality of life is severely compromised. Frequent bowel movements prevent the patient from moving far from a bathroom and do not allow normal travel or employment. In some cases, the patient's or the physician's aversion to colectomy results in months or years of functional disability far worse than would have existed if a colectomy had been performed. Another type of intractability occurs when adequate control of the patient's symptoms requires high doses of corticosteroids over a long period. It is appropriate to attempt to reduce steroid requirements by administering azathioprine or 6-MP. The side effects of corticosteroids are dose- and time-dependent and vary considerably from patient to patient. Unacceptable side effects (e.g., psychosis, accelerated hypertension) occur in some patients even with modest doses given over a short period. In these cases, the decision for surgery is clear.

A more difficult and more common problem is the patient who requires moderate to high doses of corticosteroids to control disease activity but who has had no specific problem with the corticosteroid treatment. Although this is a decision in which factors specific to the individual patient play a large role, some general guidelines may be useful. In patients who have taken large doses of prednisone (>15 mg/day) for more than 6 months, even if no specific corticosteroid side effects are present, the morbidity of continued steroid therapy must be weighed against the morbidity of surgery. Ophthalmologic examination for cataract and bone densitometry may help to identify patients with clinically silent corticosteroid side effects. The identification of asymptomatic but clinically important corticosteroid side effects may make the decision for colectomy easier. Growth failure in children is another form of intractability. Adequate nutritional support helps in most young patients, but some require colectomy. Surgical intervention should be strongly considered if growth retardation persists despite maximal nutritional and medical therapy. [410] In such a case, it is important to initiate therapy before puberty and the closing of epiphyses.

Surgery plays a relatively small role in the management of extraintestinal manifestation in ulcerative colitis. Uveitis, pyoderma gangrenosum, and arthritis usually resolve with colectomy. Some other extraintestinal manifestations, including ankylosing spondylitis and sclerosing cholangitis, do not. The extraintestinal manifestations that respond to colectomy usually respond to medical management as well. Extraintestinal manifestations are an infrequent indication for colectomy.

The standard operation for ulcerative colitis is a proctocolectomy and Brooke ileostomy. The use of newer procedures is measured against this standard. Proctocolectomy and ileostomy can be performed in one or two stages. Both have a number of advantages: the procedure is definitive and curative, and additional surgery is seldom required; the patient's functional status with the operation can be predicted with confidence; most patients make a remarkably good adjustment to the ileostomy and lead full, useful lives; many surgeons can perform this procedure successfully.

The major disadvantage of the proctocolectomy and ileostomy is that the patient has an ostomy and is totally incontinent of gas and stool; an appliance must be worn at all times. The most popular alternative operation is ileoanal anastomosis. [411], [412] In this procedure, the colon is removed completely, but the mucosa and submucosa of the rectum are dissected from the muscularis; the mucosa and submucosa are removed and the muscularis, including the internal and external sphincters, is left in place. A pouch is constructed from the terminal 30 cm of ileum. The distal end of the pouch is pulled through the anal canal, and the ileal mucosa is sewed to the dentate line. The advantage of the ileoanal anastomosis is that the patient has no ostomy and no appliance; however, a proximal temporary diverting loop ileostomy commonly is used to protect the ileoanal anastomosis until it heals. For several months after the creation of an ileoanal anastomosis, the patient has numerous bowel movements. [413] The number gradually declines so that, at 12 months, most patients are having five or six bowel movements per day. Twelve months after operation, 75% of patients have complete daytime continence, 23% have daytime seepage (minor staining of underclothes), and 2% have daytime incontinence. [414] Twelve months after the operation, 48% of patients are completely continent at night, 47% have nighttime seepage, and 5% have nighttime incontinence. Results are better for patients younger than 50 years than for older patients. An in-depth discussion of these surgical alternatives is presented in Chapter 84.

Crohn's Disease

Within 10 years of diagnosis, about 60% of patients with Crohn's disease undergo surgery for their disease. [415] The surgical approach to Crohn's disease differs markedly from the approach to ulcerative colitis. Surgical resection is not curative in Crohn's disease as colectomy is in ulcerative colitis. As a consequence, there has been a more conservative approach to the amount of tissue removed, in the knowledge that recurrences are likely and additional surgical resections in the future may be necessary. A controlled trial comparing limited resection (narrow margins) and extended resection (wide margins) found no difference in the incidence of recurrence. [416] Intractability, or failure of medical management, is a common cause for resection in Crohn's disease, as it is in ulcerative colitis, but complications (e.g., obstruction, fistula, abscess) are often indications for resection in Crohn's disease. The line between failure of medical management and a more defined complication, such as obstruction, is often not distinct. If a patient with a radiographically identifiable stricture has symptoms consistent with intermittent partial obstruction (i.e., diarrhea and pain brought on by eating) and does not respond to medical management, it is difficult to say whether the indication for surgery is obstruction or failure of medical management. Indications for surgery have been used for defining two subsets of patients with Crohn's disease, those with perforating indications (perforation or fistula) and those with nonperforating indications (obstruction or stricture). [417] If a patient's initial surgery was for a perforating indication, it is likely that subsequent surgeries also will be for perforating indications; similar results are seen with surgery for nonperforating indications.

Defining the point at which medical management has failed in Crohn's disease is somewhat more involved than in ulcerative colitis, because in Crohn's disease symptoms may not be caused by disease activity. Diarrhea and weight loss may be results of fatty-acid malabsorption, bile salt–induced colonic secretion, or small bowel bacterial overgrowth. Pain may not be due to obstruction. Optimal medical management requires recognition and treatment of these disorders. If these problems are not identified, patients may be subjected to unnecessary surgery.

As in ulcerative colitis, surgery for failure of medical management in Crohn's disease falls into two groups. In the first group, surgery is required because the progression of the disease has led the patient to a condition of unacceptable disability and invalidism despite optimal medical management. Whereas in ulcerative colitis the major source of disability is diarrhea, in Crohn's disease there is more commonly a combination of diarrhea, pain, weight loss, and malnutrition. In evaluating the possibility of surgical intervention in ulcerative colitis, the patient's current morbidity from disease activity is balanced against the relatively predictable morbidity of a colectomy. In Crohn's disease, however, the current morbidity must be balanced against the less well-defined morbidity of more limited surgery and also against the projected long-term morbidity as the disease progresses. Another distinction between surgical therapy in ulcerative colitis and Crohn's disease is that surgery for ulcerative colitis is almost always performed to remove actively inflamed tissue, but surgery for Crohn's disease may be required to remove actively inflamed tissue or to remove the sequelae of inflammation. For example, strictures requiring surgical intervention may occur in Crohn's disease at the site of a surgical anastomosis where there is considerable scarring with only a modest amount of active inflammation. This distinction is important because there is no point in treating a stricture with a therapeutic trial of steroids if scarring and fibrosis has caused the stricture. In addition, if the problem is known to be largely scar tissue and fibrosis, a stricturoplasty can be done or the extent of the surgical resection can be limited to the area of narrowing without worry about maintaining adequate uninflamed margins.

The second group of patients who undergo surgery for failure of medical management in Crohn's disease are those suffering side effects from drugs, usually corticosteroids. The guidelines for surgical intervention in patients with Crohn's disease with complications of medical therapy are exactly the same as for patients with ulcerative colitis. A problem frequently faced by physicians treating Crohn's disease is the patient whose disease is controlled with corticosteroids but flares if the dose of corticosteroids is reduced below a certain level. The level varies from patient to patient, but one study put the average at 0.15 mg of prednisone per kilogram per day. [418] Several options are available in this circumstance. One approach is to maintain the patient at a steroid dose above the critical level and to monitor the patient carefully for manifestations of steroid toxicity. This follow-up includes bone densitometry and ophthalmologic examinations. A second approach, not exclusive of the first, is the use of immunomodulators to achieve a steroid-sparing effect. This approach trades potential complications of corticosteroids for those of immunomodulators. A third approach is to resect the involved segment, knowing that the chance of eventual recurrence is high. Each of these approaches has advantages and disadvantages, all of which should be explained to the patient before a decision is made.

The incidence of recurrence after ileal resection for ileitis or ileocolic resection for ileocolic disease is about 50% after 10 years and 75% after 15 years as assessed by rates of reoperation. [419] If the presence of recurrence is assessed endoscopically, it is clear that recurrence after resection occurs more rapidly than previously appreciated. [132] Routine colonoscopic surveillance 1 year after ileocolic resections revealed a 72% incidence of recurrence, as manifested by the presence of aphthous ulcers on the ileal side of the ileocolic anastomosis. [419]

A surgical alternative to resection for small intestine strictures, termed stricturoplasty, involves longitudinal incision of the strictured segment, followed by transverse closure. [420], [421] This method preserves bowel and has a low rate of complications. [422], [423]

Surgical approaches to Crohn's colitis include segmental resection, subtotal colectomy with ileoproctostomy, and total colectomy with ileostomy. Segmental resection of Crohn's colitis is appropriate if a relatively short segment of colon is involved. Recurrence rates are high, approximating those described for ileal and ileocolic disease. Subtotal colectomy with ileoproctostomy should be considered only in a patient with an absolutely normal rectum. [424] Even in selected patients, the recurrence rate after this procedure is high, about 75% at 10 years. [425] For patients with extensive disease including the rectum, the procedure of choice is total proctocolectomy with a Brooke ileostomy. Total colectomy with ileoanal anastomosis is not appropriate in Crohn's colitis because recurrence of Crohn's disease in the ileal segment forming the new pouch would require a repeat operation and loss of a long segment of ileum.

COLON CANCER, DYSPLASIA, AND COLONOSCOPIC SURVEILLANCE

Patients with extensive ulcerative colitis have a markedly increased risk for colon cancer compared with the general population. The magnitude of this risk is uncertain. Early reports of extremely high incidences of colon cancer among patients with ulcerative colitis (50% cumulative risk at 30 years) were biased by the use of referral-based populations that were not representative of the general population. [426] A report of the experience of a private gastroenterology practice in New York probably comes closer to the experience in the general population. In this practice, the probability that colon cancer would develop in patients with extensive colitis was 11.7% at 26 years. [427] Retrospective studies reveal cumulative risks as high as 34% at 25 years in Sweden and as low as 1.4% at 18 years in Denmark. Several series have put the lifetime risk of cancer in ulcerative colitis patients in the 3% to 5% range. [428], [429] and [430] The identified risk factors for colon cancer in patients with ulcerative colitis include duration and extent of disease, primary sclerosing cholangitis, and the presence of backwash ileitis. A family history of colorectal cancer is an independent risk factor as well. The risk of developing cancer becomes appreciable 8 to 10 years after diagnosis and increases with time. Age of onset of ulcerative colitis seems to have little impact on the annual incidence of colon cancer. The risk of malignancy, however, is a larger issue for those who develop ulcerative colitis earlier in life because they have a longer period in which they are at risk and thus a higher cumulative incidence of cancer. Recent studies have suggested that among patients developing ulcerative colitis after the age of 45, the risk of cancer might increase in less than 8 to 10 years. Colon cancer in the general population is a disease of late middle and older age, but in ulcerative colitis, colon cancer occurs earlier and is commonly seen in the fourth decade of life. The risk of malignancy is also a function of the anatomic extent of the disease; the risk is much greater with pancolitis than with left-sided disease. Those with pancolitis and backwash ileitis are at greater risk than those with pancolitis alone. [431] Patients with long-standing ulcerative colitis are at risk for developing cancer even if their symptoms have been relatively mild. [426], [432] Patients are seen with colon cancer whose ulcerative colitis has been quiescent for 10 to 15 years.

The response of the medical community to the risk of colon cancer in ulcerative colitis has been that the increased risk is too high to ignore but not high enough to recommend prophylactic colectomy at some arbitrary time after diagnosis. These circumstances led to a desire to be able to identify patients at special risk for colon cancer so that they can be offered a colectomy before the development of malignancy or after malignancy has developed but curative resection is still possible. Examination of stool for occult blood is a widely used screening test for colon cancer in the general population, but it is useless in ulcerative colitis because of colitis-induced bleeding. Even overt symptoms are of less use in identifying colon cancer in ulcerative colitis than in the general population. Rectal bleeding, a symptom associated with left-sided colon cancers in the general population, is a common feature of ulcerative colitis and thus is not a reliable marker for the presence of malignancy.

Surveillance colonoscopy is used in patients with long-standing ulcerative colitis to search for malignancies and to obtain random mucosal biopsies to be examined for dysplasia. In patients with ulcerative colitis, colon cancers are often submucosal and may be easily missed at colonoscopy. Dysplasia, if present, does not occur universally throughout the colon but only in certain areas, and cannot be identified by visual inspection but only by microscopic examination of biopsies.

The pathology of colon cancers in ulcerative colitis also differs from that in the general population. Multicentric tumors account for only 2% to 3% of all colon cancers in the general population, but for up to 26% of colon cancers in patients with ulcerative colitis. [428], [433], [434] Cancers in ulcerative colitis are more likely to be largely submucosal. The tumor may be indistinguishable from the surrounding mucosa; it may appear as a flat, plaquelike lesion, or it may present as a stricture. The large exophytic masses with sharp margins seen in colon cancer in the general population are less common in ulcerative colitis.

Dysplasia is an unequivocal neoplastic transformation in the epithelium without penetration to the lamina propria. It is marked by nuclear stratification, loss of nuclear polarity, and nuclear and cellular pleomorphism (Fig. 83-28). In 1967, Morson and Pang [435] reported a group of nine patients with ulcerative colitis who were found to have dysplasia on rectal biopsy and underwent colectomy. Of these nine patients, five were found to have malignancies elsewhere in the colon. The investigators then reviewed 23 colon specimens removed for cancer in patients with ulcerative colitis and found that all of them had areas of dysplasia. Other groups confirmed these observations, and it is now clear that about 88% of colons resected for malignancy in ulcerative colitis have dysplasia somewhere in the specimen. These findings suggest that dysplasia may precede the development of carcinoma and that dysplasia may be a marker to identify patients with ulcerative colitis who have developed or are at risk of developing carcinoma. [436] Dysplasia should therefore be viewed as not a marker for future cancer but indicative of a high risk of concurrent cancer in the colon at that point in time. This finding, in turn, led to the suggestion that patients with long-standing ulcerative colitis undergo periodic colonoscopy with biopsies and, if the biopsies show dysplasia, prophylactic colectomy.

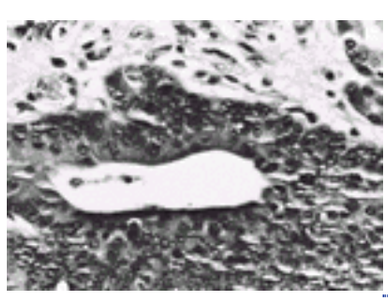

FIGURE 83-28. High-grade dysplasia with nuclear stratification, nuclear and cellular pleomorphism, and loss of nuclear polarity. Compare with Figure 83-19, in which nuclear polarity is clearly preserved despite the presence of inflammation. (Courtesy of David Lacey, M.D., St. Louis, MO.)

This plan of action, however, has several problems. The first is that dysplasia is not easy to read on colonic biopsy, and interobserver variation is considerable. A group of pathologists developed a classification for dysplasia that should help to resolve this problem. [437] This classification divides biopsies into negative, indefinite, and positive groups; the positive group is further divided into those of high-grade and low-grade dysplasia. Even in major academic centers, considerable interobserver variation occurs in diagnosing low-grade dysplasia. A second problem is that the presence of inflammation often makes it effectively impossible to decide whether dysplasia is present. A third problem is that, although dysplasia may be extensive, it is more often patchy and can be missed entirely if blind biopsies are taken at 10-cm intervals through the colon. In some cases, dysplasia exists only in small patches in the immediate vicinity of the tumor. Although 88% of carcinomas are associated with dysplasia elsewhere in the specimen, this does not mean that colonoscopy with biopsies will always find the dysplastic area.

In one group of patients the finding of dysplasia appears to have special meaning: those in whom dysplasia is found in the vicinity of a suspicious lesion or mass. [438], [439] This condition is called DALM (dysplasia-associated lesions or masses). Preliminary figures suggest that if an endoscopically visible lesion and low-grade dysplasia are present together on a first colonoscopy, there is a 40% chance the lesion will be carcinoma; if high-grade dysplasia and an endoscopically visible lesion are present, the incidence of colon cancer increases to 60%. However, recent studies have suggested that not every polyp needs to be considered a DALM. [440] Some patients with ulcerative colitis can be followed safely after polypectomy without colectomy, though with conservative surveillance. [441] Polypectomy with biopsy around the base is recommended to determine if the adenoma arose from a fat lesion. This approach to polypectomies is recommended for patients with ulcerative colitis, particularly for those in whom an adenoma developed in mucosa proximal to the extent of colitis or in those whose polyp did not arise from a dysplastic, flat lesion.

The association of dysplasia and colon cancer in ulcerative colitis is firmly established, but the benefits of surveillance for dysplasia are less clear. [442], [443], [444] and [445]

The usual approach to surveillance has been to perform a screening colonoscopy with biopsy in patients with pancolitis of 7 to 10 years' duration. The value of this screening examination is well established. What is less clear is the value of surveillance colonoscopy with multiple biopsies done every 1 to 2 years later.

After the pathological diagnosis of either high-grade or low-grade dysplasia has been made, the next step is to decide on management. A wide range of approaches to management can be found in the literature. If the diagnosis of high-grade dysplasia is made and confirmed by an experienced pathologist, the patient should have a colectomy. The approach to low-grade dysplasia probably should be the same as the approach to high-grade dysplasia. Some authors have recommended that if the diagnosis of low-grade dysplasia is made, the colonoscopy and biopsy should be repeated and the patient should have a colectomy if the diagnosis is confirmed. Low-grade dysplasia is a neoplastic lesion, however, and is usually patchy so that repeat endoscopy with biopsy may miss the dysplastic area. In one series of patients undergoing colectomy for low-grade dysplasia, 19% were found to have colorectal cancer. [446], [447] In addition, surveillance of patients with low-grade dysplasia eventually found high-grade dysplasia in a high percentage of cases. [448] On this basis, some recommend colectomy on a single firm diagnosis of low- grade dysplasia. Repeat biopsies should be reserved for a reading of indefinite dysplasia.

Table 83-8 shows the results of four large studies of surveillance in ulcerative colitis. [438], [448], [449], [450], [451], [452] and [453] These results are difficult to interpret. The most obvious success is identification of seven patients with Dukes stage A carcinoma. How does one interpret the 46 patients who, as a result of surveillance studies, had colectomies that contained dysplasia but no carcinoma? Some would declare all these to be surveillance successes, but we do not know how many of these patients would have developed carcinoma if untreated. We know that there is a high incidence of dysplasia in patients with ulcerative colitis whose colons were resected for carcinoma. However, both the *sensitivity* (how many patients with ulcerative colitis with early cancer will have biopsies positive for dysplasia) and the *specificity* (how many people who test positive for dysplasia will go on to develop cancer) for dysplasia surveillance are unknown. Thus, despite huge amounts of effort over a long period, we do not have the information to make clear recommendations about the usefulness or the timing of surveillance. Developing guidelines for surveillance would be much easier if we had a clearer idea of the magnitude of the risk of developing carcinoma in ulcerative colitis. It would be easier to be enthusiastic about a program that called for prophylactic colectomy in patients with ulcerative colitis with dysplasia if the cumulative lifetime risk of patients with ulcerative colitis developing colon cancer were 25% than if it were 2%.

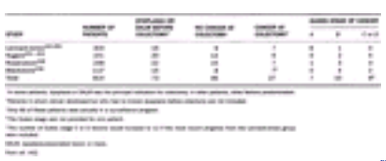

TABLE 83-8 Summary of Four Surveillance Studies in Patients With Ulcerative Colitis

The present system of surveillance is not ideal, but no more definitive or more useful alternative markers of malignancy in ulcerative colitis have been identified. There is also no clear evidence that surveillance improves survival. How should the practitioner deal with the issue of surveillance in the face of inadequate data? The first step is to inform the patient of the risks of malignancy. Patients with long-standing quiescent disease need this information as much as those with active disease because they too are at increased risk. Similarly, patients with subtotal colectomies and retained rectums need to know that they are at risk. The second step is to explain the purpose of surveillance. If the patient would refuse colectomy even if high-grade dysplasia were found, there is no purpose in initiating surveillance. Conversely, knowledge of the increased risk of malignancy may drive the patient to seek a prophylactic colectomy, especially if he or she has already suffered severe problems from disease activity. Another consideration is the availability of a pathologist competent to evaluate dysplasia. Many pathologists have little or no experience in identifying and grading dysplasia in ulcerative colitis.

If the patient is willing to undergo surveillance and competent pathology support is available, the next question is the design of the surveillance protocol. Most protocols involve taking three or four biopsies every 10 cm throughout the colon. It is important that the entire colon be examined. Biopsies should be taken from uninflamed areas. [437] The colon also should be examined for masses, strictures, and plaquelike lesions, and biopsies be performed on any that are found. Additionally the colon should be examined for polyps, which should be removed. Pseudopolyps are not premalignant lesions and need not be removed. Surveillance should begin about 8 to 10 years after the start of pancolitis and can be delayed longer than that for left-sided disease. Some protocols call for colonoscopy every year, but that is probably excessive. One group calculated the optimal screening interval on a cost-benefit basis and suggested surveillance every 3 years initially and then more frequently as the duration of the disease and the risk of malignancy increase. [454] The algorithms for most surveillance programs are similar, and the specifics of one are given in Figure 83-29. High-grade dysplasia associated with a mass or lesion is a strong indication for colectomy. High-grade dysplasia, even without an associated lesion, is also reason for colectomy. Low-grade dysplasia is viewed by some as an adequate indication for colectomy. Despite the absence of evidence for increased survival, most clinicians use a surveillance program for patients with long-standing extensive disease. Although most gastroenterologists use some surveillance protocol, it would be difficult to fault one who, because of the absence of a well-documented benefit to surveillance, did not engage in any surveillance program. In addition, administration of folic acid at 1 mg daily has been advocated to reduce the risk of colorectal cancer. Although hardly conclusive, the data suggest that low red cell folate levels correlate with a greater risk of cancer. [243] But as controlled trials are likely never to be done, given the benign nature of folic acid supplementation and the supportive evidence for a role in decreasing the risk of colon cancer in individuals without ulcerative colitis, many recommend daily folate supplementation for their patients.

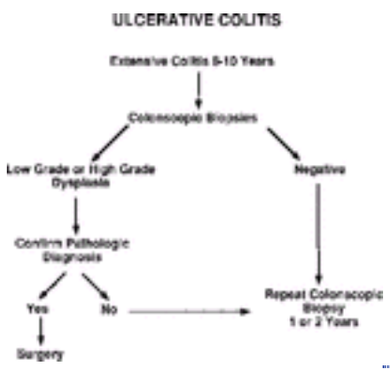

FIGURE 83-29. A proposed system of surveillance for cancer in patients with ulcerative colitis using colonoscopy and biopsy.

Much less information is available on cancer in Crohn's disease than in ulcerative colitis. [455] For those with Crohn's colitis, the incidence of colon cancer is higher than in the general population; the risk of colorectal cancer in extensive Crohn's colitis appears to be similar to that of ulcerative colitis. [441], [456], [457] As with ulcerative colitis, the incidence of colon cancer in Crohn's colitis increases with the duration of the disease and is associated with dysplasia. Also, as in ulcerative colitis, colon cancers occur in patients with Crohn's disease at a younger age than in the general population. A similar surveillance strategy has been suggested with extensive Crohn's colitis as with ulcerative colitis. [457]

An association between small intestine Crohn's disease and adenocarcinoma of the small intestine has been found. [458] Most adenocarcinomas occur in areas of the small intestine actively involved with Crohn's disease. Adenocarcinoma of the small intestine occurs at an earlier age in patients with Crohn's disease than in the general population. There is also an association of small intestine adenocarcinoma with surgically bypassed loops of small bowel. It is not clear whether the high incidence of cancer in bypassed bowel is a product of the bypass procedure or merely reflects the long duration of the disease.

PREGNANCY AND INFLAMMATORY BOWEL DISEASE

Because IBD affects many women in early adulthood, the effect of IBD on pregnancy is an important clinical issue. Fertility in women with IBD is normal or only

minimally impaired. [459], [460] Fertility in men taking sulfasalazine is diminished but returns to normal a few months after cessation of the drug. [255], [461]

Most pregnancies in women with IBD produce healthy babies. The incidences of prematurity, stillbirth, and developmental defects are similar to those of the general population. [273] The incidence of spontaneous abortion is slightly higher in women with IBD (12.2%) than in the general population (9.9%). There is some suggestion that the incidence of fetal complications is somewhat higher in cases in which the mother's disease is clinically active, irrespective of drug therapy. [273] The incidences of prematurity and spontaneous abortion are both higher in patients with more active disease. Previous proctocolectomy or the presence of an ileostomy is not an impediment to the successful completion of a pregnancy. [462]

Many women with IBD take medication on a chronic basis, and the possibility of the fetus suffering undesirable effects from the drugs is a matter of concern. [463], [464] Many women have taken sulfasalazine throughout the course of pregnancy, and there is no evidence for its causing harm to the fetus or newborn. In one series of 174 patients, sulfasalazine had no effect on spontaneous abortion, prematurity, or fetal weight. [272] Pregnant women have an increased requirement for folic acid, and sulfasalazine interferes with folate absorption. Women taking sulfasalazine who are pregnant or considering pregnancy should receive folate supplementation (1 mg twice daily) to ensure that the fetus receives amounts adequate for normal development. Up to 1 mg daily of folate supplementation is now recommended for all pregnant women to prevent congenital neural tube malformations. It seems reasonable, therefore, that all women taking sulfasalazine who are pregnant or contemplating pregnancy should take 2 mg of folate daily to ensure absorption of adequate amounts, as the period surrounding conception is the most critical for neural tube development. The use of corticosteroids by pregnant women with IBD is not associated with an increased rate of fetal complications. [272] In general, it appears that the risks to the pregnancy of treatment with sulfasalazine or corticosteroids are less than the risks of allowing disease activity to go untreated. Nonetheless, it is advisable for pregnant women to minimize their exposure to drugs, and sulfasalazine and corticosteroids should be withdrawn if possible. Even better, the patient should delay becoming pregnant, if possible, until her disease is quiescent and drugs can be withdrawn.

Most data on azathioprine and 6-MP in pregnancy come from the transplant literature and involve higher doses than are commonly used in IBD. Reported fetal effects in the transplant population include congenital malformations, immunosuppression, prematurity, and growth retardation. Risks in the IBD population are not known. It may be reasonable for a patient to continue maintenance therapy through pregnancy if the potential risks to the fetus are explained. Breast-feeding is not recommended for women taking these drugs. Methotrexate induces abortions and should not be used in women at risk for pregnancy. Metronidazole, in the short courses used to treat trichomoniasis, appears to be safe in pregnancy, although there are no data with the longer courses commonly used in IBD.

A number of studies have assessed the effects of pregnancy on disease activity in IBD. If the patient's disease is inactive at the time of conception, it is likely that it will remain inactive during the course of the pregnancy. [465], [466] If the disease is active at the time of conception, the course is harder to predict. Ulcerative colitis that is active at the time of conception tends to worsen. In two thirds of Crohn's disease cases that are active at conception, the degree of activity remains the same; among the other third, some improve clinically and others deteriorate.

INFLAMMATORY BOWEL DISEASE IN CHILDHOOD AND ADOLESCENCE

Although the peak incidence of IBD is in young adulthood, 15% of patients with ulcerative colitis and 25% to 33% of patients with Crohn's disease present before 20 years of age. For the most part, IBD presenting in childhood is similar to the disease in adults. The clinical presentation of ulcerative colitis in childhood, as in adulthood, is marked by diarrhea and rectal bleeding. The clinical presentation of Crohn's disease, however, is somewhat different in childhood. [467] Abdominal pain, weight loss, and diarrhea are common in both adults and children, but extraintestinal manifestations (e.g., arthritis, iridocyclitis, clubbing, erythema nodosum) are more likely to be major components of the initial clinical presentation in children than in adults. Abdominal pain in Crohn's disease tends to be periumbilical and colicky and is confused with the pain of functional disease.

Growth failure is a major presenting complaint in 30% of children with Crohn's disease. [467], [468] and [469] In mild disease of short duration, growth failure manifests itself as reduced weight for height; but, in long-standing disease, linear growth may be markedly retarded. Retarded bone development and delayed sexual maturation also may be manifestations of growth failure. In one third of children with impaired growth and Crohn's disease, the failure of linear growth antedates the onset of intestinal symptoms. Malnutrition appears to be the major cause of growth retardation. Poor oral intake and malabsorption both contribute to malnutrition. Poor oral intake results from anorexia and food avoidance as a result of pain and diarrhea associated with eating. Hypoalbuminemia is common, as is anemia, which may result from blood loss or from diminished intake of iron and folate. In addition to malnutrition, the presence of inflammation may contribute to growth failure. Extensive inflammation increases caloric requirements. Whether cytokines and other factors produced during inflammation contribute to growth retardation is unknown. Corticosteroid therapy also may contribute to growth failure. Growth retardation is less common in children with ulcerative colitis than in those with Crohn's disease.

Diagnosis of Crohn's disease in children is commonly delayed. The prominence of extraintestinal manifestations and growth retardation and the lesser role of intestinal symptoms contributes to the delay in the consideration of Crohn's disease as a diagnosis. Recurrent abdominal pain with or without diarrhea is a common complaint in childhood and usually reflects no significant pathology. In Crohn's disease, however, pain and diarrhea are seldom the only presenting symptoms. [467] Urgency, rectal bleeding, fever, and weight loss are signs that the child's abdominal pain is not on a functional basis and suggest that evaluation for IBD is indicated. Findings of perianal disease, uveitis, clubbing, or arthritis should suggest the possibility of Crohn's disease.

Nutritional supplementation plays a major role in the management of IBD, particularly Crohn's disease, in childhood. Children have smaller nutritional reserves and higher nutritional requirements per kilogram than adults. Moreover, nutritional deficiencies are a major source of growth retardation in childhood IBD. Oral supplementation with liquid formulas, [470] continuous supplementation by nasogastric tube, [471] and TPN all have been successful in reducing symptoms and reversing growth retardation. Irrespective of the method of delivering nutrition, all programs attempt to administer a number of calories in excess of the number recommended for healthy children, because the children with Crohn's disease have lost weight. Calorie intake on the various regimens ranges from 50 to 93 kcal/kg daily and protein intake ranges from 1.6 to 3.5 g/kg daily.

Medical management of IBD in childhood (reviewed by Winter and Grand [472]) is largely the same as in adulthood. All the major controlled studies of therapy in IBD purposefully excluded children. Treatment in children is extrapolated from those studies. The indications for sulfasalazine therapy in children are the same as in adults. In children, sulfasalazine is begun at 25 to 40 mg/kg daily and increased to 75 mg/kg daily. The indications for corticosteroid therapy are also the same as for adults with IBD. Typical starting doses for moderate to severe disease would be 1 to 2 mg of prednisone per kilogram per day. Continuous therapy with prednisone in doses as low as 10 mg/day can inhibit normal growth. After acute symptoms are under control, an attempt should be made to convert the patient to an every-other-day prednisone regimen. [473] If prednisone is given as a single dose every other day, the side effects are markedly reduced. In particular, prednisone at doses as high as 40 to 50 mg every other morning allows normal growth in some children, although lower doses (10–20 mg every other day) are more likely to be compatible with normal growth. [474] Not all patients can be converted to an alternate-day regimen, however. In some patients, the disease flares on the day off prednisone.

Immunomodulators, specifically 6-MP and azathioprine, have been used in childhood Crohn's disease much as they have been used in the adult disease. [474], [475] and [476] Reservations about side effects are, if anything, greater if these drugs are used in children. The possibility of malignancy is particularly worrisome in children, who have the potential for longer exposure. Nonetheless, the problems with steroid therapy in children are so great that a trial of 6-MP or azathioprine is probably justified in steroid-dependent children.

Indications for colectomy in ulcerative colitis in childhood are the same as for adults, with the addition of growth failure as an indication. Growth failure may be the result of either disease activity or corticosteroid administration. Surgical resection of localized areas of disease activity may be useful in children with Crohn's disease; however, the success of surgical resections in reversing growth retardation has been disappointing. [477]

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CHAPTER 84

Alessandro Fichera and Fabrizio Michelassi

SURGICAL TREATMENT OF INFLAMMATORY BOWEL DISEASE

Crohn's disease, ulcerative colitis, and the rare forms of true indeterminate colitis represent a disease spectrum with protean manifestation and complications. Although as many as half of the patients with inflammatory bowel disease require at least one surgical procedure during their lifetime, the decision to operate is rarely an easy one. Such a decision should be the result of a thorough collaboration between the gastroenterologist and the surgeon, assisted by the radiologist and the pathologist. Factors to take into consideration are the age and the general conditions of the patient; the extent of the disease; the duration of the disease and the treatment offered to that point; and the specific complication in regard to the available treatment options. Once the need for surgical intervention has been established, other factors, specific to each individual presentation, must be considered in order to choose the most appropriate surgical procedure. This chapter addresses surgical options and their results in view of the different indications for surgical treatment of inflammatory bowel disease. Emphasis is given to new options offered by advances in laparoscopic surgery.

CROHN'S DISEASE

Crohn's disease is a heterogeneous entity, which requires individual approaches for management due to the variety of intestinal segments affected and the different clinical manifestations of the disease. The first attempt to classify patients with Crohn's disease was based on the anatomic locations of the disease. [1] Subsequently Greenstein and Greenstein [2] proposed that perforating and nonperforating presentations might separate patients into two different clinical forms of Crohn's disease. Indeed recurrent postoperative disease usually follows the pattern of the initial presentation. [2], [3] The more recent Vienna Classification divides patients with Crohn's disease into groups based on age at diagnosis, location of involved segment, and clinical behavior (nonstricturing and nonpenetrating, stricturing and penetrating). [4] These distinctions have proven implications with respect to medical therapy, indication for surgery, and risk of postoperative recurrence.

The majority of patients presenting with uncomplicated active disease are initially treated with medical therapy. Although medical therapy has become more sophisticated with the adoption of new biologic agents, it still fails to significantly change the natural history of the disease. [5], [6] Patients still tend to require surgery as time progresses: after 20 and 30 years of symptoms, 78% and 90% of patients, respectively, require surgery. [7]

Indications for Surgery in Crohn's Disease

The chronic, unrelenting, and recurrent nature of Crohn's disease brings these patients to the attention of the gastroenterologist during the early phases of the disease. [1] In the absence of complications, the initial management is medical, until the treatment fails or a complication ensues. Table 84-1 lists the indications to surgical treatment.

Sepsis
Inflammatory masses or abscesses
Fistulae
Perforation
Failure of medical therapy
Obstruction
Hemorrhage
Cancer
Fulminant colitis with or without toxic megacolon

TABLE 84-1 Indications for Surgery in Crohn's Disease

A significant number of patients with perforating Crohn's disease present with septic complications as the first presentation or recurrence. [2] Not all the septic complications are an absolute indication for surgery, rather they are a marker of a severe and aggressive form of the disease. Fistulae are identified in one third of patients with Crohn's disease, [8] but they are rarely the primary indication for surgery. Specific indications for surgical treatment include enterocutaneous and enterovaginal fistulae, where the enteric drainage becomes a matter of personal embarrassment for the patient; [9] enterovesical or colovesical fistulae, where the connection of the intestine to the genitourinary system causes repeated urinary tract infections and eventually impairment of renal function; [10] and those enteroenteric fistulae that produce functional and anatomic bypass of a major segment of intestine with consequent malabsorption and/or profuse diarrhea. Inflammatory masses and abscesses occur in as many as 20% of patients. [8], [11] These patients should be treated with antibiotics, the abscesses drained percutaneously, if feasible, and surgery should be postponed until a definitive procedure can be safely performed. Primary free perforation is a very unusual complication of Crohn's disease: usually the severity of the transmural inflammation leads to the formation of adhesions, which wall-off the perforation resulting in an abscess. The secondary rupture of an abscess into the abdominal cavity requires prompt surgical intervention.

Medical treatment has failed and surgery is necessary (a) when maximal medical therapy proves inadequate; (b) for those patients who may be asymptomatic while on maximal medical therapy, but develop recurrence of symptoms with tapering of the medications; (c) when the disease progresses with worsening symptoms or a complication arises while the patient is receiving maximal medical therapy; and (d) in the presence of significant treatment-related complications, including steroid-induced Cushingoid features, cataracts, glaucoma, hypertension, aseptic necrosis of the head of the femur, myopathy, vertebral body fractures, myelosuppression, anaphylactic reaction, or growth retardation in children. [12] In our series, failure of medical therapy was the primary indication for surgery in 33.6% of patients. [8]

As many as 22% of patients present to the surgeon with worsening obstipation. [8] Symptoms are caused by a single or multiple strictures or a lengthy diseased segment and differ depending on the location of the disease in the gastrointestinal tract: delayed gastric emptying in gastroduodenal disease; postprandial cramps in

jejunoileitis; distention, pain, and diarrhea in colonic disease; laborious defecation in perianal Crohn's disease. Intestinal obstruction, even when complete, is rarely an indication for urgent surgery. In general, it is advisable to let the obstruction, even if complete, resolve with nasogastric decompression, intravenous hydration, and medical therapy and to postpone surgery after resolution of the obstruction to allow for a definitive procedure (i.e., strictureplasty or resection).

Other less frequent complications requiring surgical treatment include gastrointestinal hemorrhage, cancer, and toxic megacolon. Massive hemorrhage is an extremely rare complication occurring in less than 1% of patients. [1], [13] Preoperative localization studies are mandatory to minimize the length of intestine resected. Cancer has been described in the small bowel [14] and the colon [15] and in a previous strictureplasty site. [16] The risk of cancer correlates with duration of the disease and may be higher in bypassed loops. Surgery for cancer in patients with Crohn's disease follows the same oncologic principles as for sporadic cancers. Fulminant colitis with or without toxic megacolon is a known entity in inflammatory bowel disease. [17], [18] The surgical treatment of this complication will be discussed in more detail in the ulcerative colitis section of this chapter.

Crohn's Diseases of the Stomach and Duodenum

Crohn's disease may involve any portion of the gastrointestinal tract, from the mouth to the anus. About 2% to 4% of patients with Crohn's disease present with involvement of the stomach or duodenum. [19] The most common indications for surgical treatment include hemorrhage and obstruction [20] (Fig. 84-1). Indeed, in a recent review of 108 patients, 83% underwent surgery for this complication. [21] Multiple surgical procedures have been advocated for the treatment of gastroduodenal Crohn's disease. Resectional antiulcer procedures have been performed mostly in situations in which an ulcer was misdiagnosed, and they have been associated with high morbidity and mortality rates. [19] Bypass procedures, such as gastrojejunostomy with or without vagotomy, have been the procedures most often advocated. Based on the location of the disease, gastroduodenostomy or duodenojejunostomy have also been advocated. These procedures are associated with good short-term outcome and acceptable morbidity rates, although delayed gastric emptying occurs in up to 24% of patients, [22] occasionally requiring reoperation. [23] Strictureplasty has been advocated as an alternative to bypass procedures in selected patients. Only two retrospective reports are available that compare strictureplasty with bypass surgery for treatment of gastroduodenal Crohn's disease. [22], [23] With a follow-up between 42 and 192 months the strictureplasty-treated group had a lesser rate of delayed gastric emptying (15% vs. 24%), but suffered more anastomotic dehiscences (12% vs. 6%) and had a higher rate of repeated surgical interventions: 11 of 26 patients (42%) required a second operation after strictureplasty, 9 of them due to recurrent Crohn's disease, compared with 24% in the bypass-treated group. [22], [23] Based upon these limited data, both bypass and strictureplasty procedures appear to offer merit in appropriately selected patients.

FIGURE 84-1. Upper gastrointestinal barium study showing Crohn's disease of the third and fourth portions of the duodenum. (From Block GE, Michelassi F, Tanaka M, et al. Crohn's disease. Curr Probl Surg 1993;30:173.)

Crohn's Disease of the Small Bowel

The jejunum and ileum, not including the terminal ileum, are affected by Crohn's disease in 3% to 10% of patients. [8], [24] The two most common indications for surgical treatment are obstruction and sepsis; massive hemorrhage and carcinoma are much less common. Chronic, high-grade small bowel obstruction may be caused by single or multiple, short or long strictures. These patients present with postprandial abdominal pain, nausea, and vomiting and often progress to a high-grade obstruction. When multiple tight strictures are present, the small bowel is transformed into a sequence of dilated saccular segments separated by tight, ringlike strictures (Fig. 84-2). The dilated segments, which contain partially digested food particles, become the ideal environment for bacterial overgrowth. Because of bacterial overgrowth and stagnation, patients report symptoms of occasional diarrhea, malabsorption, and vitamin B_{12} deficiency.

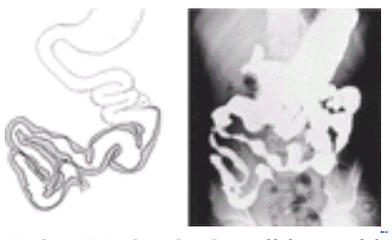

FIGURE 84-2. Small bowel barium study showing extensive jejunoileal disease with strictures and saccular dilations. (From Block GE, Michelassi F, Tanaka M, et al. Crohn's disease. Curr Probl Surg 1993;30:173.)

With this clinical picture, patients have traditionally undergone resection of long segments of small bowel. Considering that Crohn's disease is a recurrent disease and up to 30% of patients require a second operation, [25] short bowel syndrome may eventually be precipitated in 1.5% to 12.6% of cases. [26], [27] In an attempt to preserve bowel length, Lee and Papaioannou in 1982 [28] and subsequently Alexander-Williams and Haynes in 1985 [29] described the use of strictureplasty, which had been previously described in India to correct tubercular stricture of the terminal ileum and cecum. [30]

Many different strictureplasty techniques have been described. The most popular are the Heineke-Mikulicz, the Finney, and the side-to-side isoperistaltic strictureplasties. [31], [32], [33], [34], [35], [36], [37], [38] and [39] Generally for short strictures, less than 10 to 12 cm in length, a Heineke-Mikulicz strictureplasty is performed (Fig. 84-3); for longer strictures either a Finney type (Fig. 84-4) or a side-to-side isoperistaltic strictureplasty (Fig. 84-5) is indicated. Prior to performing a strictureplasty, it is advisable to perform biopsy on all suspicious areas to rule out the presence of malignancy. Strictureplasty is contraindicated in the presence of active sepsis (peritonitis, abscess, or acute fistula), when the bowel wall is thickened and unyielding, when the stricture has undergone malignant degeneration, or in patients with severe weight loss and marked hypoalbuminemia. [40]

FIGURE 84-3. Heineke-Mikulicz strictureplasty for short stenosis (up to 7 cm). (From Fazio VW, Galandiuk S, Jagelman DG, et al. Strictureplasty in Crohn's disease. Ann Surg 1989;210:621.)

FIGURE 84-4. Finney strictureplasty for single stenosis longer than 7 cm. (From Shahrif H, Alexander-Williams J. The role of strictureplasty in Crohn's disease. Int Surg 1992;77:15.)

FIGURE 84-5. Isoperistaltic side-to-side strictureplasty for multiple sequential strictures and extensive jejunoileitis. **A:** The mesentery of the diseased loop is divided at its midpoint, and the small bowel is severed between atraumatic intestinal clamps. The proximal intestinal loop is moved over the distal one in a side-to-side fashion. **B:** The two loops are approximated by a layer of interrupted seromuscular Cushing stitches using nonabsorbable sutures. **C:** A longitudinal enterotomy is performed on both loops and the intestinal ends are spatulated to avoid blind stumps (*inset*) **D:** Both outer and inner suture lines are continued and finished anteriorly. The completed side-to-side isoperistaltic strictureplasty is shown in the *inset*. (From Michelassi F, Hurst RD, Melis M, et al. Side-to-side isoperistaltic strictureplasty in extensive Crohn's disease. Ann Surg 2000;232:401.)

The Cleveland Clinic group reviewed their experience of 1124 strictureplasties in 314 patients with a median follow-up of 7.5 years. [40] The overall morbidity rate was 18% including 2% anastomotic dehiscence rate and 7% postoperative bleeding from the suture line requiring transfusion. [41] These investigators reported 34% surgical recurrence rate, which compares favorably with the results of resective surgery. [42] The authors identified age and weight loss as the only factors associated with postoperative morbidity and younger age at diagnosis as the primary factor associated with early recurrence. [40] These results confirm the safety and efficacy of strictureplasty in selected patients. Yet, as a word of caution, one of their patients developed adenocarcinoma at the strictureplasty site 7 years after the index operation. Negative results of biopsies of the stricture had been obtained at the time of the strictureplasty. [43]

Septic complications, such as an inflammatory mass, an abscess and, occasionally, a fistula, are also considered indications for surgical treatment of small bowel Crohn's disease. As already discussed, fistulae usually reflect severe disease rather than an absolute indication for surgical treatment. Septic complications will be discussed in further detail in the section on Crohn's disease of the terminal ileum.

The first case of a carcinoma of the small bowel in Crohn's disease was described by Leon Ginzburg in 1956 in a case of stricturing small bowel Crohn's disease. [44] Since then there have been more than 100 cases described in the literature. [16], [43], [44], [45], [46], [47], [48], [49], [50] and [51] Most cancers are adenocarcinomas, [46] but they differ from de novo cancers in many respects: they present at a younger age, are more common in the distal small bowel, and may be multifocal or diffuse. [47]

Cancer may occur in association with chronic perineal fistulae and excluded loops of small bowel. Several cases of small bowel adenocarcinoma in bypassed loops have been reported. [47], [48], [49] and [50] The increased risk of cancer development in a bypassed loop and the inability to image such a loop has deterred surgeons from performing intestinal bypass or allowed them to consider leaving excluded intestinal segments in situ for a long time.

The surgical treatment of small bowel carcinomas in Crohn's disease is segmental radical resection, when feasible. The prognosis is poor, with survival rates reported between 23% at 3 years [49] and 5% at 5 years. [51] The patients with cancers in a bypassed loop usually do not survive longer than 18 months. [49]

Crohn's Disease of the Terminal Ileum

The terminal ileum is the most common gastrointestinal location requiring surgery in Crohn's disease and accounts for approximately 40% of patients with Crohn's disease referred to the surgeon. [1] Classically, patients present with obstructive symptoms or with septic features suggesting either a contained perforation or an abscess with or without a fistula. Cancer and hemorrhage are infrequent indications to surgical resection.

For obstructive disease of the terminal ileum secondary to strictures (Fig. 84-6) or an inflammatory mass (Fig. 84-7), resection is the treatment of choice. An ileocecectomy usually suffices; if the disease extends in the colon, an ileocolectomy may be necessary.

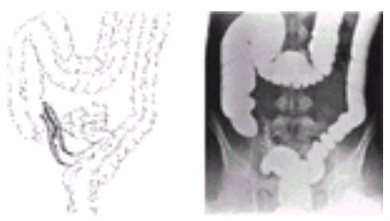

FIGURE 84-6. Barium enema with reflux in the terminal ileum showing long stenosis, referred to as the "string sign." (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

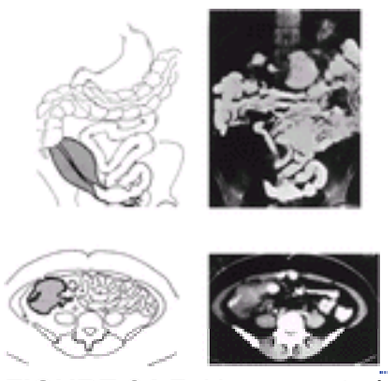

FIGURE 84-7. Upper gastrointestinal series and computed axial tomography showing a large right lower quadrant inflammatory mass in a patient with disease confined to the terminal ileum. Note the displacement of several loops of small bowel in the right lower quadrant. (From Michelassi F, Balestracci T, Chappell R, et al. Primary and recurrent Crohn's disease. Experience with 1379 patients. Ann Surg 1991;214:230.)

Abscesses (Fig. 84-8) and fistulae are two relatively common complications of Crohn's disease of the terminal ileum. Their occurrence may influence the timing of surgery and significantly increase its complexity. When feasible, these patients should be managed with image-guided drainage and antibiotics with elective resection at a later time. This approach is feasible in approximately 50% of patients. [52] If percutaneous drainage is not successful or in the presence of a secondary free rupture of the abscess in the peritoneal cavity, open surgical exploration is necessary. The procedure should aim at draining the abscess, clearing the sepsis, and resecting the diseased intestinal segment. A primary anastomosis can then be fashioned if the patient's hemodynamic and nutritional conditions allow it and if the final suture line can be placed away from the abscess cavity. It is also advisable to lay an omental flap on the residual wall of the abscess cavity to quarantine any residual infection from the abdominal cavity and the anastomosis. If a primary anastomosis is not advisable, a stoma needs to be brought out as a temporary end ileostomy. When the extent of the inflammatory reaction is massive and involves several adjacent loops of bowel, the procedure should be limited to drain the abscess and place a proximal diverting stoma. In due time, after most of the acute inflammatory reaction has subsided, a limited resection of the diseased bowel can be performed, saving a significant amount of bowel that otherwise would have been resected at the time of the first exploration.

FIGURE 84-8. Computed axial tomography showing a right lower quadrant abscess originating from Crohn's disease of the terminal ileum and perforated through the abdominal wall into the subcut. Air fluid level is evident. (From Michelassi F, Balestracci T, Chappell R, et al. Primary and recurrent Crohn's disease. Experience with 1379 patients. Ann Surg 1991;214:230.)

Psoas abscess occurs as a result of a retroperitoneal perforation of the ileocecal region. Clinical manifestations vary from mild sepsis to severe psoas spasm with hip pain, flexion, and external rotation of the thigh associated with an abdominal mass. The retroperitoneal process can compress the ureter and cause right hydronephrosis (Fig. 84-9). Resection of the inflammatory mass and drainage of the abscess usually relieves the compression on the ureter and cures the hydronephrosis.

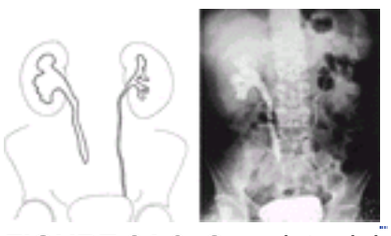

FIGURE 84-9. Complete right ureteral obstruction with hydronephrosis. (From Michelassi F, Balestracci T, Chappell R, et al. Primary and recurrent Crohn's disease. Experience with 1379 patients. Ann Surg 1991;214:230.)

Fistulae are common in Crohn's disease of the terminal ileum, but they rarely represent the only indication for surgical treatment. In our series of 639 patients undergoing surgery for Crohn's disease, 331 patients had disease in the terminal ileum. Of these, 217 patients (65.6%) were found to harbor 285 intra-abdominal fistulae; yet these fistulae represented the primary indication for surgical treatment in only 6.3% of our patients. [53] Enteroduodenal, enteroenteric, and enterocolic fistulae usually are asymptomatic and often discovered only during a careful abdominal exploration or at inspection of the resected specimen. [54] We consider these fistulae the indication to surgical treatment only if they cause massive diarrhea because of the bypass of a sizable length of intestine.

Enterocutaneous fistulae usually drain through a previous abdominal scar or through the umbilicus. [55] At times, they result from surgical incision and drainage of a subcutaneous abscess complicating severe intra-abdominal disease or from percutaneous drainage of an abdominal abscess. [52] The presence of an enterocutaneous fistula does not necessarily dictate the need for immediate surgical intervention. [9], [56] Patients may be reluctant to undergo surgical treatment when the enterocutaneous fistula has a minimal output and the underlying disease is under satisfactory control. However, in most cases the difficulty in maintaining personal hygiene, the fear of social embarrassment, the bothersome symptoms associated with the severely diseased segment that led to the formation of the fistula, and the skin excoriation, which invariably forms around the cutaneous opening of the fistula, become factors in indicating the need for surgical treatment.

Enterovesical fistulae occur in 2% to 5% of patients with Crohn's disease. [57], [58] Some controversy exists regarding the timing of surgical intervention in the presence of these fistulae. Nevertheless, most surgeons and gastroenterologists agree that the consequences of chronic urinary tract infections on renal function, in addition to the symptoms affecting the intestinal tract itself, represent an indication for operation. Enterovaginal fistulae are rare complications of Crohn's disease and most often occur in women who have undergone a previous hysterectomy. The vaginal discharge is cause for discomfort, social and sexual embarrassment, and difficulty in maintaining personal hygiene. Most patients readily accept surgical intervention.

Massive intestinal hemorrhage is a rare complication of terminal ileitis. [59] Because the hemorrhage originates from active disease, patients with Crohn's disease are likely to have repeated episodes ultimately requiring surgical intervention. If the extent of the disease is limited, as assessed by contrast radiography, an elective resection should be considered after two episodes of self-limited hemorrhage. In the presence of an ongoing, unrelenting hemorrhage, an angiogram should be obtained to localize the source of bleeding, especially in the presence of multiple sites of disease involvement. [13] The possibility of a life-threatening hemorrhage in patients with Crohn's disease should not be underestimated. Five cases of exsanguinating gastrointestinal hemorrhage have been reported in the literature. [60]

Malignant transformation of the terminal ileum is fortunately rare. Its treatment is based on oncologic principles and it has been described in the previous section.

Controversies still persist about the timing of the operation. In view of the fact that 70% to 90% of these patients require a resection in their lifetime, [61], [62] some authors have advocated early resection to avoid possible septic complication associated with recurrent attacks and the side effects of aggressive medical treatment. [63], [64] On the other hand, in general, most physicians and surgeons recommend a resection only after medical treatment has failed or a complication of the disease has arisen.

Overall the results of resection are excellent. Large series have reported a surgical recurrence rate between 31% and 36% at 10 years with 69% of patients requiring only one resection. [61], [63] Several disease-related and patient-related factors associated with high risk of recurrence have been identified. The presence of perianal disease, extensive ileal involvement, and cigarette smoking seem to be significantly associated with high recurrence rate after ileocolic resection. [61], [65], [66] When Crohn's disease recurs, it usually involves the ileum just proximal to the anastomosis; less frequently the colon just distal to the anastomosis or the anastomosis exclusively are involved. Strictureplasty has been used for the treatment of anastomotic stricture secondary to recurrent Crohn's disease after ileocolic resection with excellent results. [67], [68]

Crohn's Colitis

The colon is affected by Crohn's disease in up to 30% of patients. [1], [8], [69] The involvement can be limited to a segment or extend to the entire colon and rectum; furthermore, patients may present with associated anorectal or small bowel disease. In the presence of pancolitis without perineal or small bowel manifestations of the disease, the differential diagnosis between Crohn's disease and ulcerative colitis may be difficult and many patients end up carrying a diagnosis of indeterminate colitis as a result of diagnostic uncertainty [70] or with the wrong diagnosis. [71], [72], [73] and [74] With the advent of ileoanal pouch procedures this differentiation has become crucial since pouch reconstruction in patients with Crohn's disease is contraindicated, with a pouch failure rate up to 40%. [71], [72], [73] and [74]

The most common indication for surgery in patients with disease localized primarily to the colon is failure of medical therapy. [8], [75] These patients present with persistent and often bloody diarrhea and abdominal pain not responding to medical therapy. With worsening conditions, toxic colitis with or without megacolon may develop. This complication is less frequently associated with Crohn's disease than ulcerative colitis, but it carries similar mortality rates (14%–16%) in both conditions. Factors affecting mortality include age (30% for patients older than 40 years old, vs. 5% for those younger than 40), gender (21% in women vs. 13% in men), and the occurrence of colonic perforation (44% for cases with perforation vs. only 2% in those without perforation). [18] In the absence of overt perforation, initial therapy consists of high-dose steroids, bowel rest, and antibiotics. Lack of improvement over a short period of time or any signs of worsening condition are indications for an urgent operation to avoid colonic perforation.

The risk of colorectal cancer is increased in Crohn's colitis between 4-fold and 20-fold, [76], [77] with an incidence between 1.4% and 1.8%. [15], [44], [45] and [46], [78], [79] Carcinoma can arise in a long-standing benign stricture, probably due to chronic inflammation. [80]

Routine surveillance colonoscopy should be considered after 7 to 10 years of disease duration, with the recommendation to proceed with resection in the presence of dysplasia. Prophylactic colectomies should be performed for tight strictures not allowing passage of the endoscope and, therefore, interfering with a complete colonoscopy; a stricture difficult to survey; multiple pseudopolyps rendering surveillance difficult; and, as mentioned earlier, the presence of an excluded intestinal segment (retained rectal stump). Surgery for adenocarcinoma in Crohn's colitis should be based on oncologic principles and on the extent of the lumenal disease.

Septic complications occur in the form of inflammatory masses or, more infrequently, fistulae or abscesses. Inflammatory masses can be found in any segment of the colon and rectum with equal frequency. Transverse colon disease occasionally fistulizes into the stomach. A common location for abscesses to occur is the descending colon at the junction with the sigmoid colon. These abscesses, which form in the left parietocolic gutter and lay over the psoas muscle, may be confused with a complication of diverticular disease. Usually, the age of the patient and other manifestations of Crohn's disease help in arriving at the correct diagnosis.

Several different surgical approaches are available for the treatment of patients with Crohn's colitis. The surgical plan depends on the location of the disease, the urgency of intervention, the presence of complications, and the general condition of the patient. In Crohn's colitis the need to preserve bowel length is less pressing than in small bowel Crohn's disease, but still the extent of surgery should be carefully planned by the surgeon in conjunction with the gastroenterologist, the pathologist, the radiologist and, foremost, the patient.

Surgery for Crohn's disease of the colon must encompass the entire gross disease. In patients with disease limited to the right colon, a right hemicolectomy will suffice (Fig. 84-10); if the disease extends beyond the midtransverse colon, an extended right hemicolectomy may be necessary; for patients with disease involving the entire abdominal colon (Fig. 84-11), an abdominal colectomy will be needed. All these procedures can be performed in one stage. In the past, a diverting ileostomy was widely used as the sole initial step in patients too ill to tolerate definitive surgery. [81], [82], [83], [84] and [85] A more current approach consists of the appropriate colon resection with an end ileostomy and delayed reconstruction of the gastrointestinal continuity. This procedure is also indicated in patients with a diagnosis of indeterminate colitis to preserve the option of an ileal pouch procedure if the diagnosis of ulcerative colitis is later confirmed or established.

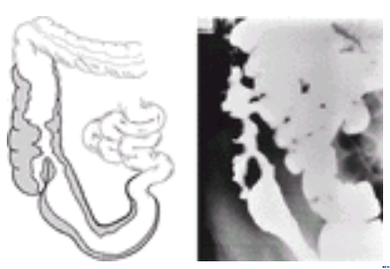

FIGURE 84-10. Small bowel follow-through series showing Crohn's disease of the ileal cecal valve and right colon. (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

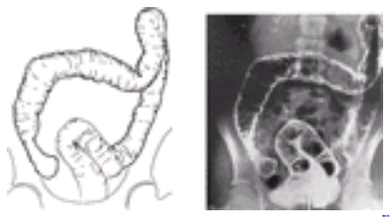

FIGURE 84-11. Barium enema showing Crohn's colitis with sparing of the rectum and rectosigmoid. (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

Abdominal colectomy and ileorectal anastomosis, in one or two stages, is ideally suited for young patients with no sigmoid or rectal involvement and normal anal sphincter function. [86] In older adult patients, the decision in favor of an abdominal colectomy with ileorectal anastomosis is a difficult one and needs to carefully consider the degree of anal sphincter function and the possible debilitating high fecal frequency.

Total proctocolectomy and end ileostomy, in one or two stages, has been and still is the surgical therapy of choice in patients with Crohn's pancolitis or in Crohn's colitis with extensive anorectal involvement or anal incontinence. [87] Recurrence of Crohn's disease in the small bowel after total proctocolectomy has been reported in 3% to 46% of patients [88], [89] and it involves the distal 25 cm of ileum in up to 90% of patients. [90] If Crohn's disease is limited to the rectum (Fig. 84-12) or the patient is affected by severe anorectal disease or incontinence, an abdominoperineal resection with a left-end colostomy is the procedure of choice.

FIGURE 84-12. Barium enema showing Crohn's proctitis with loss of the rectal ampulla, several rectosigmoid fistulae, and an intervening perirectal abscess. (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

In the presence of severe perineal sepsis, patients undergoing a proctocolectomy or an abdominoperineal resection are at risk of developing perineal wound problems. Large series have reported up to 34.4% of nonhealing perineal wounds after proctectomy in Crohn's disease. [90], [91] and [92] A subtotal colectomy or an abdominal proctectomy with colostomy may be indicated as the first step to allow for appropriate drainage and healing of the perineal sepsis. A perineal proctectomy at a later time is still necessary to avoid the risks of a retained rectal stump. The use of myocutaneous flaps may further decrease the incidence of nonhealing perineal wounds. [93]

For left-sided Crohn's colitis (Fig. 84-13), segmental resection is a valuable alternative. It allows for restoration of bowel continuity and maintenance of the absorptive mucosa of the right colon, but patients are at risk of recurrence and additional surgery. [94] Small series have reported a recurrence rate of 62% at 5.5 years, requiring further surgery, [95] but up to 86% of patients maintaining bowel continuity at 14 years. [96]

FIGURE 84-13. Barium enema showing Crohn's colitis limited to the proximal portion of the descending colon. (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

Ileal pouch surgery is contraindicated for patients with Crohn's colitis. [72], [74] Despite the effort to correctly diagnose patients before surgery, some patients undergo surgery with a preoperative diagnosis of indeterminate colitis or ulcerative colitis and are found to have Crohn's disease on final pathological evaluation of the specimen. [97] Sagar and colleagues [98] and Deustch and associates, [71] in two separate unselected series, reported a pouch failure rate of 45% at 10 years in a total of 46 surgical patients with a preoperative diagnosis of mucosal ulcerative colitis who were subsequently proven to have Crohn's disease. Hyman and colleagues [73] analyzed the outcome of this procedure on 25 patients: 16 patients, at a mean follow-up of 38 months, had a functioning pouch, 7 had required pouch excision, 1 had a diverting stoma, and 1 had died. Only 1 of 9 patients with preoperative clinical features suggestive of Crohn's disease had a functioning pouch, with complications uniformly occurring within months of ileostomy closure. In contrast, 15 of 16 patients with out preoperative features of Crohn's disease had maintained their pouch, generally with good results. These studies suggest that pelvic pouch procedure should not be performed in patients with clinical features of Crohn's disease because of the high associated complication rate. Yet, there may be a subgroup of patients with Crohn's colitis who may ultimately be recognized as possible candidates for an ileal pouch procedure. To this extent Panis and colleagues [99] reported a series of 31 patients with Crohn's disease with no evidence of perineal or small bowel disease. These patients were specifically selected for ileoanal pouch as an alternative to ileostomy. Their results were compared with those obtained from patients with ulcerative colitis undergoing the same procedure. Of the 31 patients with Crohn's disease, six (19%) experienced specific complications 9 months to 6 years after surgery: three had pouch-perineal fistulae, which required pouch excision in two cases. At the 5-year follow-up, there was no significant difference between patients with Crohn's disease and patients with ulcerative colitis with a functioning pouch in stool frequency, continence, gas/stool discrimination, leak or need for protective pads, and sexual activity. It is probable that in the future more sophisticated diagnostic tests may allow selection of a subgroup of patients with Crohn's disease appropriate for ileal pouch procedures.

Perianal Crohn's Disease

Anorectal Crohn's disease may manifest with edematous skin tags, fissures, ulcers, abscesses, fistulae, strictures and, as a complication of chronic, long-standing inflammation, anal cancer. The reported incidence of perianal Crohn's disease requiring surgery varies between 25% and 34%. [93], [100] Crohn's colitis is much more frequently associated with anal lesions than Crohn's disease of the small bowel (52 % vs. 14 %). When an anal lesion is the manifesting sign, Crohn's disease will soon develop elsewhere in the intestine. [100] Since these lesions frequently herald the onset of intestinal Crohn's disease, the physician must always be aware of the possibility of inflammatory bowel disease when dealing with a suspicious anal lesion.

Skin tags, fissures, and ulcers rarely require surgical treatment. The most common indications for surgery in perianal Crohn's disease are septic in nature. For these complications the treatment plan should be based on the patient's general condition, anal continence, and degree of colorectal involvement. In patients with a grossly normal rectal mucosa, the anal stenosis can be dilated, ischiorectal abscesses can be drained, appropriate fistulectomies can be performed, and repair of complex fistulae or low rectovaginal fistulae can be attempted with every expectation of successful outcome. In patients with mild to moderate involvement of the rectal mucosa, treatment of the rectal disease with topical antiinflammatory agents may occasionally effect dramatic improvement. If the improvement persists, surgical repair of even complicated defects may result in complete healing and normal function. In patients with severe perineal disease or incontinence, only a proctectomy with a permanent stoma may be expected to bring relief. Extensive perineal disease, while not a sole indication for proctectomy, usually is associated with destruction of the sphincter mechanism and incontinence, often dictating a resection with a permanent stoma.

Patients presenting with an ischiorectal abscess require prompt incision and drainage. In the majority of patients, the incision will heal by secondary intention without any need for further intervention. In 35% of patients, a fistula-in-ano will develop. [93]

Many surgeons mistakenly believe that perineal wounds do not heal in patients with Crohn's disease and therefore have adopted a philosophy of therapeutic nihilism even in the presence of simple fistulae-in-ano. [101], [102], [103], [104] and [105] This attitude may result in the patient's continued suffering and may lead to the development of new abscesses and fistulae, progression to a "watering-pot" perineum, and destruction of the sphincter mechanism. Patients with Crohn's disease with symptomatic low anal fistula involving minimum sphincter musculature can be treated safely with fistulotomy [106] with healing rates up to 85%. [107], [108] In patients with horseshoe abscesses and high fistulae, aggressive local surgical intervention aimed at drainage of sepsis and appropriate usage of setons permit preservation of the sphincter and good postoperative function. [106], [107], [109], [110]

Rectovaginal fistulae occur as a complication of anorectal Crohn's disease in about 10% of patients [111], [112] and require special mention. Most fistulae are truly anovaginal, with the internal opening located at the dentate line. These fistulae result from deep anterior anal ulceration and less commonly from a crypto-glandular source. These patients may be treated with a mucosal advancement flap. [113], [114] and [115] This procedure entails performing a semicircular incision at the dentate line, with the internal opening of the fistula at its center (Fig. 84-14). A 3- to 4-cm flap of mucosa, submucosa, and smooth muscle is elevated in a proximal direction. The tract is curetted and closed and the flap is advanced to the anoderm and sutured without tension. Several variations have been proposed when the rectal mucosa is significantly diseased, including performing the repair from the vaginal side [116], [117] and using an anocutaneous flap from the perianal skin. [118] Success rates with this approach have been reported around 70% to 75% in relatively small series. [113], [114] and [115] In the event of a failure, the advancement can be repeated as reported by Joo and colleagues. [115] In their series, four of five patients who did not heal with the first procedure were successfully closed after a second attempt.

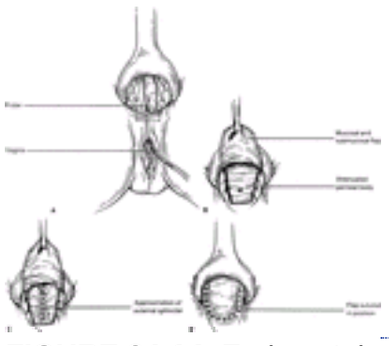

FIGURE 84-14. Endorectal advancement flap for low rectovaginal fistula. **A:** The patient is in the jack-knife prone position and the probe identifies the rectovaginal fistula. **B:** The rectal wall is elevated from the external sphincter, starting at the dentate line. **C:** Plication of the external sphincter with closure of fistula. **D:** The rectal wall flap is advanced over the plicated external sphincter; the tip of the flap with the fistula opening is excised; the flap is then sutured in position with interrupted sutures. (From Michelassi F. Crohn's disease. In Bell RH Jr, Rikkers LF, Mulholland MW, eds. Digestive tract surgery: a text and atlas. Philadelphia: Lippincott-Raven, 1996:1201.)

Anorectal complications also include stenosis and cancer. In the absence of severe rectal disease, anal stenosis can be dilated to achieve prolonged symptomatic relief. Cancer may develop from the diseased anorectal mucosa, a chronic fistula-in-ano, or a retained rectal stump. In all cases, an oncologic resection needs to be carried out. In the presence of a resulting large soft tissue and cutaneous perianal defect, the use of myocutaneous flaps may be the only way to achieve primary closure of the perineal wound (Fig. 84-15).

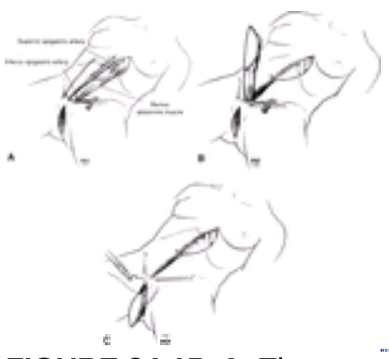

FIGURE 84-15. A: The superior epigastric vessels supply the rectus abdominis myocutaneous flap. **B:** The flap includes skin, anterior fascia, and the rectus muscle. **C:** The flap is delivered through the pelvis to the perineal defect. (From Hurst RD, Gottlieb LJ, Crucitti P et al. Primary closure of complicated perineal wounds with myocutaneous and fascia cutaneous flaps after proctectomy for Crohn's disease. Primary myocutaneous flap closure following resection of locally advanced pelvic malignancies. Surgery 2001;130:767; originally from Radice E, Nelson H, Mercill S, et al, Br J Surg 1999;86:349, with permission of the Mayo Foundation.)

All told, an aggressive surgical approach should allow healing of the Crohn's disease perineal septic complication with sphincter preservation in 62% to 86% of patients. [112], [119] In our series of 224 consecutive patients, the reasons for proctectomy in 85 patients included aggressive disease nonresponding to conservative measures in 66 patients, extensive fistular disease in 15 patients, fecal incontinence in 2 patients, and tight anal stenosis in 2 patients. We found that patients with rectal disease had a significantly higher rate of proctectomy than patients with rectal sparing (77.6% vs. 13.6%), and that in the absence of rectal involvement, patients with multiple complications had a significantly higher rate of proctectomy than patients with single complications (23% vs. 10%). [119]

Laparoscopic Surgery for Crohn's Disease

Although laparoscopic techniques have become standard approaches to the treatment of several upper abdominal pathologies (i.e., gallbladder disease and reflux esophagitis) and benign colorectal conditions (i.e., diverticulitis, limited colectomies for benign polyps, endometriosis involving the intestine), the use of laparoscopy in Crohn's disease is still debated. On the one hand, surgery for Crohn's disease can be quite challenging even with a conventional approach. Many of the unique features of Crohn's disease, such as the intense inflammation, thickened mesentery, enteric fistulae, inflammatory masses or abscesses, and the multiplicity of areas of intestinal involvement, have deterred many surgeons from even considering a laparoscopic approach. On the other hand, the natural history of the disease may lead to the need for repeated operations and a less "invasive" approach would indeed be appealing. Additionally, patients are generally young and interested in undergoing procedures that involve minimal scarring and prompt recovery.

Although a number of clinical reports have described that laparoscopic-assisted ileocolic resections are feasible and safe in the treatment of Crohn's disease, these series have been uncontrolled and nonrandomized. [120], [121], [122], [123] and [124] Additionally, several reports question whether or not any advantages at all support the use of laparoscopic methods compared to conventional methods in Crohn's disease. [124], [125]

Two prospective and controlled studies have shown several advantages of the laparoscopic-assisted approach over the conventional open approach. Bemelman and associates [126] compared 48 open ileocolic resections with 30 laparoscopic-assisted resections. They showed that laparoscopic ileocolic resection for Crohn's disease is associated with similar morbidity rates, a shorter hospital stay, and improved cosmetic results. Alabaz and colleagues [127] compared 48 open ileocolic resections with 26 laparoscopic-assisted resections. Patients in the laparoscopic-assisted group returned to work more quickly, had better cosmetic results, and were more likely to have improved social and sexual lives.

A prospective randomized trial comparing open and laparoscopic-assisted ileocolic resections in 60 patients [128] showed a faster postoperative recovery of respiratory function (measured as recovery of 80% of forced expiratory volume and forced vital capacity), a lower incidence of minor complications, and the need for a shorter abdominal incision in the laparoscopic-limited group. With a follow-up of 20 months there were no recurrences in either group. This limited single-surgeon experience needs to be validated by others and needs to be extended to a longer follow-up to see whether the purported advantages for patients who may need repeated surgical procedures are indeed validated.

The indication for laparoscopic surgery in Crohn's disease should not differ from the indications for conventional open surgery. Contraindications to a laparoscopic approach include patients who are critically ill and unable to tolerate the pneumoperitoneum due to hypotension or hypercarbia, patients with dense adhesions or extensive intra-abdominal sepsis (abscess, free perforation, complex fistulae), and the inability to clearly identify the anatomy (previous surgery, obesity, adhesions). [129]

Laparoscopic assisted ileocolic resection (Fig. 84-16) is currently the most commonly performed laparoscopic procedure for Crohn's disease. [120], [130] Laparoscopic fecal diversion and closure of stomas may obviate a laparotomy and allow for a full evaluation of the gastrointestinal tract. Laparoscopic-assisted small bowel resection or strictureplasty may be performed as the index procedure or when associated with another bowel resection, usually an ileocolic resection. The diseased area or areas can be marked with intracorporeal stitches, exteriorized through a small abdominal incision needed for removal of the resected specimen, and the strictureplasty performed extracorporeally in a standard fashion.

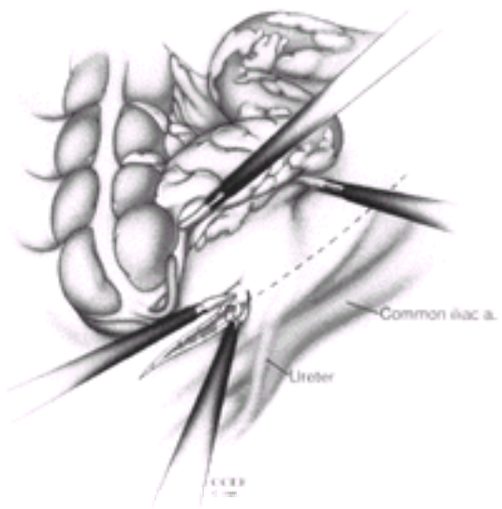

FIGURE 84-16. Laparoscopic ileocecectomy (From Kim SH, Milsom JW. Laparoscopy in inflammatory bowel disease. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:326.)

Segmental or total colectomies with or without anastomosis are can be performed by the laparoscopic-assisted method. Due to their complexity these procedures are still controversial. [131] A few series have reported a small number of cases; [124], [130], [132], [133] and [134] some authors do not consider laparoscopic colon resection for Crohn's disease advantageous. [135]

In summary, laparoscopy in Crohn's disease may allow for faster recovery and reduced surgical trauma; morbidity, in experienced hands, compares favorably with the conventional open approach. With the continuous improvement of laparoscopic instruments and the refinement of laparoscopic surgical techniques, this approach in the near future may also be offered to patients with more challenging and extensive complications of Crohn's disease.

ULCERATIVE COLITIS

There is a major difference between the management of ulcerative colitis and Crohn's disease. In Crohn's disease the surgical approach is palliative, with the more limited objective of relieving the complications of the disease to improve the patient's clinical status and preserving bowel length. In ulcerative colitis, removal of the colon and rectum eliminates and cures the disease.

Ulcerative colitis is a disease confined to the colon and rectum and the presence of intestinal involvement outside these boundaries must be ruled out to confirm the diagnosis. Anamnestic evidence of recurrent episodes of perianal sepsis or bowel obstruction should suggest the possibility of Crohn's disease. On physical examination, the presence of active perianal sepsis and fistulae, healed scars, or anal stenosis should raise doubts about the diagnosis of ulcerative colitis. A complete preoperative evaluation should include: stool cultures to rule out an infectious etiology; small bowel x-ray series to exclude involvement of the small intestine; colonoscopy to evaluate the extension of the disease, rule out the presence of malignancy, and obtain histological confirmation.

The majority of patients with ulcerative colitis present with bloody diarrhea, urgency, and fatigue; during an acute flare, they may present with fever and, possibly, abdominal distention. In absence of absolute indications for immediate surgery, such as perforation or carcinoma, the patient is treated initially with medical therapy. [136], [137] and [138] However, a definitive treatment plan should be formulated after complete diagnostic evaluation, taking into account the extent and duration of the disease, the presence of dysplasia or cancer, medical therapy used, presence of side effects due to the medical treatment, and the patient's general condition and expectations. As in Crohn's disease an optimal treatment plan requires input from the patient as well as the surgeon and the gastroenterologist, assisted by the radiologist and pathologist.

Indications for Surgery

The most common indication for surgical treatment of ulcerative colitis in our experience is the failure of medical therapy. [139] This constitutes the following scenarios: when maximum medical therapy proves inadequate; for those patients who may be asymptomatic while on maximum medical therapy, but develop recurrence of symptoms with tapering of the medications; when the disease progresses with worsening symptoms while the patient is receiving maximum medical therapy; and when the patient develops significant treatment-related complications.

Other indications for surgical treatment include the presence of a mass at colonoscopy, dysplasia on colonic biopsy, and dysplasia-associated lesions or masses (DALM). Colonoscopic detection of mucosal dysplasia is considered the best available surveillance tool for the detection of cancer. The presence of dysplasia has been reported in a much higher percentage of specimens of ulcerative colitis complicated by cancer than in ulcerative colitis without malignant transformation. Because the correlation between dysplasia and cancer is not constant, when dysplasia is found on surveillance colonoscopy, some suggest immediate colectomy, whereas others opt for continued surveillance. Gorfine and colleagues [140] reviewed the pathology reports of 590 patients who underwent total proctocolectomy or restorative proctocolectomy for ulcerative colitis and found that cancers were significantly more common among specimens with dysplastic changes. Specimens with dysplasia of any grade were 36 times more likely to harbor invasive carcinoma. Stage III disease was found in association with indefinite or low-grade dysplasia in 19.2% of cases. More important, tumor stage did not correlate with dysplasia grade. These investigators concluded that even though dysplasia is an unreliable marker for the detection of synchronous carcinoma, when dysplasia of any grade is discovered at colonoscopy, the probability of a coexistent carcinoma is high. We believe that a histopathological diagnosis of dysplasia is an indication for colectomy.

Carcinoma was the indication for surgery in 2% of our series. [139] Patients with ulcerative colitis are known to have an increased risk of colorectal cancer. [141], [142] The risk of cancer increases with the extent and the duration of the colitis. Gyde and associates [143] reported an overall 8-fold increase in the risk of cancer in these patients, with a 4-fold increase in the left-sided colitis and proctitis group and a 19-fold increase in the pancolitis group. In addition, the pancolitis group had a cumulative risk of 7.2% at 20 years and 16.5% at 30 years from diagnosis.

Acute fulminant colitis with or without an acute abdomen develops in about 13% of patients. [139] About 60% of these patients fail to respond to medical therapy [136], [144] or develop persistent abdominal pain, distention, diffuse abdominal tenderness, rebound, tachycardia, and fever, suggesting an acute abdomen. The acute abdomen may be due to the development of a toxic megacolon or a walled-off or free perforation. These patients should undergo emergency surgical intervention after appropriate volume resuscitation.

Massive hemorrhage is also another indication for urgent surgery. Persistent hemorrhage despite maximal medical treatment is a manifestation of severe disease. The hemorrhage originates from extensive mucosal ulcerations, rather than a definite arterial source. As such, an abdominal colectomy is necessary to remove the majority of the bleeding surface. A primary anastomosis should be avoided in favor of an ileostomy and closure of the rectal stump. With fecal diversion obtained by placing the ileostomy, postoperative rectal bleeding is rare. When it happens it usually responds to increased doses of systemic steroids, or administration of topical steroids or intrarectal tamponade with gauzes soaked in a diluted epinephrine solution. Retention of the rectal stump maintains the option of a subsequent restorative procedure.

Proctocolectomy With Brooke Ileostomy

Proctocolectomy with Brooke ileostomy was the gold standard for the treatment of ulcerative colitis until the early 1980s when Utsonomiya popularized the ileo pouch–anal anastomosis. [145] Since then its role in the surgical treatment of ulcerative colitis has become more limited, although it is still valuable in selected patients. [146] By removing all diseased epithelium, a proctocolectomy cures patients of the disease, eradicates the risk of malignancy associated with it, and eliminates the need for costly medications and time-consuming lifelong follow-up. The disadvantages of this operation include the presence of a permanent ileostomy, the potential for nerve injury during pelvic dissection, and the risk of perineal wound healing problems, even if significantly less than in Crohn's disease.

A proctocolectomy is indicated in patients who are not candidates for an ileal pouch–anal anastomosis or a Kock pouch, either because of older age, the presence of

poor anal sphincter function, or obesity. The operation may also be indicated if other medical problems make a more complex, longer operation too risky [147] or in the presence of a low rectal cancer in need of resection of the anal sphincter or postoperative adjuvant radiotherapy. Finally, proctocolectomy should be considered in patients who desire a single operation for cure or whose work makes it easier to handle an appliance rather than frequent bowel movements. There are no absolute contraindications to this procedure, although, in the emergent setting, it is advisable to stage the procedure with an initial abdominal colectomy. This strategy avoids the morbidity associated with the rectal dissection, which can be potentially difficult and time consuming in an unstable patient, and maintains the option of an ileal pouch–anal anastomosis as a second stage.

Preoperatively, the surgeon is well advised to have a thorough discussion with the patient for a complete understanding of this procedure and its implications on body image and self-perception. In addition, a stoma therapist, in charge of the postoperative care of the stoma, should mark the abdominal site for the ileostomy (Fig. 84-17). The appropriate site is the least inconvenient for the patient and usually it is away from bony prominences, previous scars, and does not interfere with the patient's clothing. The site is usually just lateral and inferior to the umbilicus midway between the midline and the right anterior iliac spine.

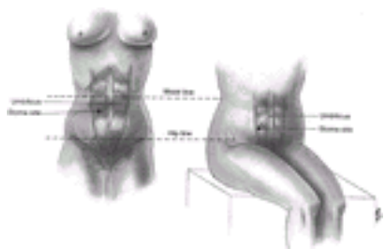

FIGURE 84-17. The stoma site is marked prior to surgery. The ileostomy should be located over the right rectus abdominus muscle on a flat area away from skin folds and bony prominences. (From Hurst R. Proctocolectomy with ileostomy, abdominal colectomy with ileostomy, and abdominal colectomy with ileoproctostomy. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:157.)

The proctocolectomy is associated with very low postoperative morbidity. Delayed healing or nonhealing of the perineal wound is rare if the perineal dissection is performed in the intersphincteric plane, between the internal and external anal sphincter. Primary closure is associated with an 80% to 90% success rate. [148], [149] and [150] In the event of perineal wound infection, drainage of the wound abscess and debridement usually are sufficient for secondary healing. Rarely, myocutaneous flaps or skin grafts are necessary to achieve closure of the perineal wound. Impotence is reported in 2% to 3% of male patients after proctectomy for benign disease. [150], [151] Impotence is secondary to injury of the autonomic nerves during pelvic dissection and its occurrence is minimized by keeping the dissection close to the rectum and by choosing an intersphincteric plane for the most distal anorectal resection. In women, dyspareunia may occur due to the posterior displacement of the vagina. Fortunately, it is usually temporary. [150], [151]

Late complications are largely related to the stoma and include retraction, prolapse, parastomal hernia, bleeding, stenosis, and varices. They usually require surgical revision. Other late complications are intestinal obstruction, urinary stone formation, and gallstone formation.

Restorative Proctocolectomy With Ileal Pouch–Anal Anastomosis

Most patients with ulcerative colitis who are candidates for elective operation are suitable for proctocolectomy with ileal pouch–anal anastomosis. Exceptions include patients with low rectal cancers, patients with fecal incontinence, or patients who, for work-related issues or personal reasons, prefer not to have a pouch. [147]

Obesity is considered a relative contraindication. A fatty ileal mesentery may cause the pouch to be rather bulky and ill fitting in a narrow pelvis; furthermore, the ileal pouch may not reach the upper anal canal despite operative techniques designed to lengthen the ileal mesentery. [152] A stapled ileoanal anastomosis, as described later in the chapter, offers advantages over a conventional hand-sewn anastomosis in these challenging patients. Alternatively, patients may need to undergo an initial colectomy with ileostomy to allow for cessation of steroid therapy and a successful weight reduction program.

The presence of cancer of the colon or proximal rectum does not rule out the possibility of an ileal pouch–anal anastomosis, provided that the cancer can be completely resected during the operation. [153], [154], [155] and [156] In the presence of metastatic cancer and a short life expectancy, less extensive procedures, such as colectomy and ileorectal anastomosis or Brooke ileostomy should be considered. The Mayo Clinic group presented their experience with 77 patients with carcinoma complicating ulcerative colitis. Although pouch failure was more common in patients with cancer than in the overall registry (16% vs. 7%), there were no differences between cancer and noncancer groups in operative complications, median stool frequency, incontinence, pad usage, or pouchitis. [153] A study from Toronto that reviewed function and quality of life after ileal pouch–anal anastomosis for colitis-associated neoplasia confirmed the functional results reported by the Mayo Group and showed that quality of life of these patients is excellent and equal to that of patients who have a pouch for failure of medical therapy. [154]

Patients with sclerosing cholangitis and cirrhosis are at increased risk of chronic pouchitis and pouch loss after an ileal pouch–anal anastomosis, but the operation usually can be performed successfully and with morbidity rates similar to that in patients without hepatobiliary complications of ulcerative colitis.

In the past, if a patient was older than 50 years, this was considered a contraindication to the procedure. However, this is no longer true. The clinical results in fit patients over the age of 50 are similar to those in younger patients. [157], [158], [159], [160] and [161] Takao and colleagues [160] have shown that there are no significant differences among age groups relative to manometric results, frequency of bowel movements, incontinence scores, or overall patient satisfaction. The same results were confirmed by others. [157], [159] Jorge and associates [158] showed that the effect of ileoanal reservoir on anal sphincter function in patients older than 50 years is similar to that in younger patients. Even though impairment of internal anal sphincter function is more pronounced after ileoanal reservoir in older patients, this appears to be transient, as it is in younger patients.

Although there are small series showing that an ileal pouch–anal anastomosis is feasible in fulminant ulcerative colitis, [162] most patients with emergent indications to surgical treatment should undergo an initial colectomy with ileostomy followed by ileal pouch–anal anastomosis [163] several months later.

The ileal pouch–anal anastomosis is most commonly performed in two stages. The colon and the rectum are excised to the pelvic floor, an ileal pouch is constructed with the terminal ileum (Fig. 84-18), an ileal pouch–anal anastomosis is constructed, and a diverting loop ileostomy is fashioned. The loop ileostomy is closed at a second operation about 3 months later. The operation, or portions of it, can be accomplished by using laparoscopic techniques.

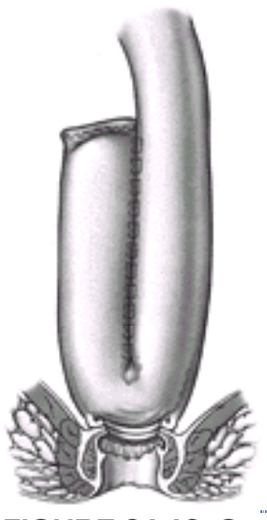

FIGURE 84-18. Stapled J-pouch ileoanal anastomosis. (From Michelassi F, Takanishi D, McLeod RS, et al. Ileal reservoirs. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:186.)

Occasionally a diverting ileostomy can be omitted after construction of the ileal pouch. Omitting this transforms an elective ileal pouch–anal anastomosis into a single-stage procedure and avoids the need for a third stage in those patients who underwent an initial colectomy and ileostomy in the emergent setting or in elective circumstances (indeterminate colitis, obesity).

Several large series examining complications and long-term outcome have been published. [164], [165], [166] and [167] The Cleveland Clinic group reported their experience with 1005 ileal pouch–anal anastomoses, 858 of which had been performed for ulcerative colitis. [165] One death (0.1%) related to pouch necrosis and sepsis was reported. The overall morbidity rate was high, with 630 patients (62.7%) suffering 1218 complications. Septic complications and reoperation occurred in 6.8% and 24%, respectively. The ileal pouch was removed in 34 patients (3.4%) overall, but in only 15 patients (1.8%) with ulcerative colitis. Approximately 25% of patients developed pouchitis. Functional results and quality of life were good to excellent in 93% of patients. The Mayo Clinic series [164], [167] of more than 1300 patients showed similar results. Three patients died as a consequence of the procedure. Postoperative pelvic sepsis rates decreased from 7% in the 1981 to 1985 period to 3% in the 1991 to 1994 period. After mean follow-up of 6.5 years, the mean number of stools was five during daytime and one at nighttime. Frequent daytime and nighttime incontinence occurred in 7% and 12% of patients, respectively, and did not change over a 10-year period. The cumulative probability of suffering at least one episode of clinical pouchitis was 18% and 48% at 1 and 10 years, respectively, and the cumulative probability of pouch failure at 1 and 10 years was 2% and 9%, respectively. [167] The most common early postoperative complications were bowel obstructions (13%), pelvic (5%) and wound sepsis (3%), transient urinary dysfunction (7%), and large enteric losses from the temporary loop ileostomy. Postoperative impotence and retrograde ejaculation was noted in up to 3% of men; transient dyspareunia occurred in 11% of patients after operation. [164] Pregnancy and delivery are well tolerated by women with a pouch. [164]

Late complications of the procedure include: anal fistulae and abscesses, which may need drainage and possibly temporary fecal diversion; stricture of the anastomosis, which can be repeatedly dilated or treated with pouch advancement in presence of dense scar tissue or inflammation of the mucosal cuff; and pouchitis, which is the most frequent late complication. Patients with pouchitis present with abdominal cramps, frequent watery stools, urgency, incontinence, malaise, and fever. Of 120 patients who underwent ileal pouch–anal anastomosis for ulcerative colitis at the University of Chicago, 50 patients suffered at least one episode of pouchitis. Two thirds of patients had multiple episodes. Chronic pouchitis occurred in 6 patients, necessitating pouch removal in 2. [168] In a prospective evaluation of 149 patients in Sweden, [169] the risk of pouchitis was highest during the initial 6 postoperative months. The cumulative risk leveled off after 2 years but was substantial (51%) at 4 years. Similar to our data and that of the Mayo Clinic, fewer than 10% of the Swedish patients had severe chronic pouchitis, and only 2 patients (1.3%) had their pouch removed because of pouchitis.

The etiology of pouchitis remains unknown. It may be due to bacterial overgrowth secondary to poor pouch emptying, or to an immunologic reaction to bacterial products or a chemical injury; ischemia and reperfusion injury has also been postulated; or it may be a novel manifestation of inflammatory bowel disease. Patients who are positive for antineutrophil cytoplasmic antibody (ANCA) are more likely to develop pouchitis than those who are not; [170] also, patients with primary sclerosing cholangitis and extraintestinal manifestation of ulcerative colitis are at a higher risk to develop pouchitis, especially of the chronic type. [171] Backwash ileitis, on the other hand, does not predispose to pouchitis. Pouchitis is rare in patients who have undergone ileal pouch–anal anastomosis for familial polyposis, an observation which seems to suggest that whatever causes colitis may predispose to pouchitis.

The activity of pouchitis can be scored using a Pouchitis Activity Index, [172] which uses clinical, histological, and endoscopic criteria. The activity index may help with the medical treatment of pouchitis, which is based on oral antibiotics (metronidazole and ciprofloxacin) in the majority of cases. Interestingly, smoking seems to prevent pouchitis. [173]

The risk of cancer in the ileal pouch is, to date, unknown. Although, no reports have appeared describing the development of carcinoma in the mucosa of a pelvic pouch, the pouch mucosa undergoes structural transformation: initially the mucosa appears normal or with mild villous atrophy and, possibly, mild inflammation. Then the mucosa shows transient atrophy with temporary moderate inflammation followed by normalization of architecture. Finally it develops a pattern of persistent atrophy with severe inflammation. [174] These changes can progress to dysplasia. [175], [176] The continual contact with a noxious medium and changes in bacterial flora are hypothesized as causes of these phenomena. The large number of proliferating cells and the frequent presence of inflammation provide fertile ground for carcinogenesis. Pouch surveillance is probably indicated, as the natural history of the pouch mucosal change is not known at this point.

Controversies in Ileal Reservoir Surgery

The technique of restorative proctocolectomy and ileal pouch–anal anastomosis for adult patients was originally described by Parks and Nicholls [177] and Utsunomiya and colleagues. [145] Since these initial descriptions several modifications have been introduced into clinical practice: they mainly involve the configuration of the pouch and the method of anastomosing the pouch to the anal canal (Table 84-2).

Pouch configuration
J
W
S
The anal transition zone
Mucosectomy and hand-sewn pouch–anal canal anastomosis
Stapled pouch–distal rectal anastomosis
Diverting loop ileostomy
One-stage procedure
Multiple-stage procedure

TABLE 84-2 Controversies in Pouch Surgery

In the initial description by Parks and Nicholls [177], the ileal reservoir was fashioned as a triple loop S-pouch (Fig. 84-19), with a 5-cm-long exit conduit, which created problems with evacuation. Subsequently the limb was shortened to less than 2 cm with significant functional improvement. [178] The two other pouch designs that are currently in use are the double loop J-pouch [145] and the quadruple loop W-pouch. [179], [180] The lateral isoperistaltic H-pouch (Fig. 84-20) is no longer used and it is now of historical interest only. [181] Each pouch design has its own advantages and currently there is not a single configuration ideal for every patient. The S-pouch with the long exit limb allows for further reach in tall male patients with short mesentery. [182] The W-pouch (Fig. 84-21) has larger capacity and better compliance [179] and is ideal when part of the terminal ileum has been or needs to be resected. The J-pouch is technically easier and faster to perform. [183], [184] When comparing the W-pouch with either the S-pouch [182] or the J-pouch, [180], [185], [186] patients with the W-pouch have significantly fewer bowel movements compared to the S-pouch or J-pouch group during the first 6 to 12 months after closure of the ileostomy. However, Keighley and colleagues, [187] comparing the J-pouch to the W-pouch in a randomized

prospective trial, did not confirm those findings and Johnston and colleagues [188] showed that both types of pouches conferred the same functional results at one year.

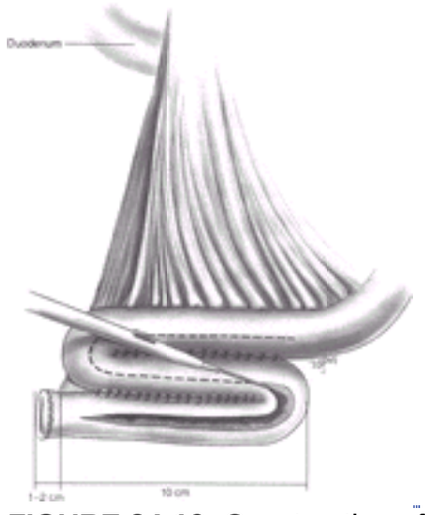

FIGURE 84-19. Construction of a triple loop S-pouch. (From Michelassi F, Takanishi D, McLeod RS, et al. Ileal reservoirs. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:186.)

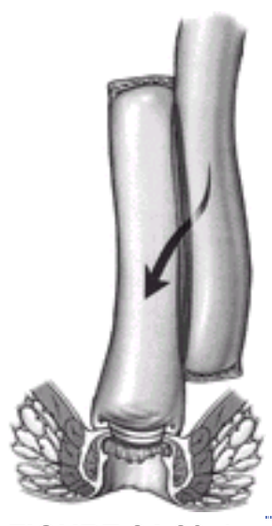

FIGURE 84-20. Isoperistaltic H-pouch. (From Michelassi F, Takanishi D, McLeod RS, et al. Ileal reservoirs. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:186.)

FIGURE 84-21. Quadruple loop W-pouch. (From Michelassi F, Takanishi D, McLeod RS, et al. Ileal reservoirs. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:186.)

We elect to perform a J-pouch for the vast majority of our patients because of the lower complication rate and the excellent functional results experienced. [139], [168], [183], [184], [189] We reserve the S-pouch for patients in whom we suspect a difficult “reach” to the anal canal and the W-pouch for patients in whom a substantial portion of terminal ileum has been or needs to be sacrificed.

Controversy still exists on whether the ileoanal anastomosis should be stapled or hand sewn. The initial descriptions of ileal pouch–anal anastomosis included a mucosectomy to the dentate line. [145], [177], [190] The anal dilation necessary for a complete mucosectomy [191] or the eversion of the anorectum advocated by some authors [192], [193] caused significant decrease in the maximum resting pressure [191] and increase of the threshold sensation, which correlated with increased number of episodes of incontinence. [192], [193] Furthermore removal of the most distal lower rectal–upper canal epithelium, the anal transitional zone (ATZ), results in the loss of the anoderm sensory capacity, which together with the rectoanal inhibitory reflex allows for sampling of rectal contents and contributes to continence. [194] The technique of stapled ileal–pouch anal anastomosis was introduced to address some of the shortcomings of the hand-sewn technique. The potential advantages of the stapled technique include less tension on the anastomosis, ease of construction, preservation of the lower rectal–upper anal canal and less trauma to the sphincter mechanism. These advantages translate to a shorter operation with fewer anastomotic complications and, potentially, better long-term functional results.

Several comparative long-term studies have reported better functional results for the stapled ileal pouch–anal anastomosis. [195], [196], [197], [198] and [199] Gemlo and associates [195] evaluated 235 patients with a mean follow-up of 70 months and found that elimination of a mucosectomy dramatically reduced nocturnal minor and major incontinence, daytime minor incontinence, and daytime protective pad use. Sagar and colleagues [199] studied anal physiological results in 20 patients up to 12 months after stapled ileal pouch–anal anastomosis. Resting anal pressure, which decreased immediately after the surgical procedure, was almost back to normal 12 months later, the rectoanal inhibitory reflex was present in 19 patients and sampling was observed in 17 patients. Ability to discriminate flatus from feces was associated with return of the rectoanal reflex and sampling. The functional results obtained following a stapled or hand-sewn ileal pouch anal anastomosis were evaluated by several prospective randomized studies. [200], [201] and [202] No statistically significant differences were detected, although the validity of the results of these studies is compromised by their small size and by the short follow-up.

Several studies have looked at the rate of anastomotic complications after mucosectomy and hand-sewn ileal pouch–anal anastomosis. Preserving a short rectal cuff with the stapled technique lessens the tension on the anastomosis and reduces anastomotic complications. In a retrospective study, the Cleveland Clinic group

evaluated 692 patients, 238 with hand-sewn and 454 with stapled ileal pouch–anal anastomosis. In the hand-sewn ileal pouch–anal anastomosis group, 25 patients (10.5%) had 32 septic complications, and 24 required 89 reoperations. In 7 patients, the pouch was excised. In the stapled ileal pouch anal anastomosis group, 21 patients (4.6%) had 23 septic complications, and 14 required 40 reoperations. One patient needed pouch excision. Patients with hand-sewn anastomosis experienced more early septic complications and more pouch excisions related to these complications than patients with stapled ileal pouch anal anastomosis. [203] Yet, when these complications were evaluated in prospective randomized studies, no difference was noted. [200], [201] and [202]

The potential disadvantages associated with preserving the ATZ include persistence of ulcerative proctitis symptoms and risk of malignant degeneration. The Cleveland Clinic [204], [205] noticed that symptomatic inflammation of the retained mucosa occurred in 14.7% of their patients; 4.1% of patients had inflammation of the anal canal alone, and 10.6% had pouchitis in addition. The need for surgical treatment occurred in 12.9% of the total patients with anal canal inflammation and 10.6% with anal canal inflammation plus pouchitis. [204], [205] These patients are usually treated with topical steroids or 5-aminosalicylate (5-ASA), and when that fails and surgical treatment is needed, a transanal mucosectomy with ileal pouch advancement is performed with excellent results. [205]

Advocates of the mucosectomy argue that the entire diseased anorectal mucosa, including the ATZ, must be removed to eliminate the risk for future dysplasia and cancer. To determine the long-term risk of dysplasia and cancer in the retained mucosal cuff after stapled ileal pouch–anal anastomosis the Cleveland Clinic group published their series of 210 patients with at least 5 years (median 77 months) of follow-up. [206] Dysplasia developed in 7 patients (3.3%) at a median of 11 months postoperatively. Patients with history of cancer or dysplasia in the colon or rectum were at a higher risk of developing dysplasia. Two patients, each with low-grade dysplasia detected on three separate occasions, underwent mucosectomy 29 and 38 months after detection of low-grade dysplasia and, fortunately no cancer was found. The 5 other patients with dysplasia on one or two occasions were treated expectantly and were apparently dysplasia-free for a median of 72 months. More important, preservation of the ATZ did not lead to the development of cancer after 5 to 10 years of follow-up. The authors recommend long-term surveillance to monitor dysplasia and, if repeat biopsy confirms persistent dysplasia, mucosectomy with pouch advancement. [206]

The actual extent of the ATZ is highly variable and mucosectomy does not assure its complete removal. Fenger, [207] using Alcian blue staining, found that the mean span of the ATZ was 8.9 mm (range 0 to 20 mm), starting up to 6 mm below the dentate line. On the other hand, Thompson-Fawcett and colleagues [208] measured the ATZ by two techniques: whole-mount Alcian blue staining and a computer map of the histological findings based on longitudinal sections taken every 3 mm. They found that the Alcian blue technique overestimates the length of the ATZ, which usually commences just above the dentate line. The median length of the ATZ measured from computer maps of the histology was only 4.5 mm. Due to this variability, mucosectomy does not reliably remove completely the entire rectal mucosa. [209] Small islets of residual rectal mucosa have been identified in up to 14% of patients; in 7% at the ileo anal anastomosis. [209]

This probably explains why, currently in the literature, the reported cases of cancer after a pouch procedure are all in patients who had a mucosectomy, [210], [211] and [212] except in one case, where the cancer was detected 16 months after stapled ileal pouch–anal anastomosis following long-standing ulcerative colitis complicated by a cancer in the upper rectum. [213] It is likely that this cancer represents a local recurrence rather than a metachronous cancer originated in the ATZ.

No hard data exist to support the superiority of one technique over the other and we believe that there is a role for both procedures in clinical practice. [139], [183] We preserve the ATZ in older patients with borderline sphincter function and in tall or obese patients to decrease tension on the anastomosis. Mucosectomy is otherwise advised in presence of dysplasia or cancer in the colon or rectum in the pediatric population [214] and in patients with primary sclerosis cholangitis, known to have a high risk of dysplasia and cancer. [215] If the ATZ is preserved, surveillance anoscopies with biopsies are performed on an annual basis to detect dysplasia and indicate the need for a completion mucosectomy.

With the perception that anastomotic complications are reduced by the stapled ileal pouch–anal anastomosis technique, some authors have questioned the need for a diverting ileostomy. [216] The Medical College of Virginia group started to perform stapled ileal pouch–anal anastomosis without diverting ileostomy in the early 1980s. [217], [218] Their series of 193 patients who underwent a stapled ileal pouch–anal anastomosis as a one-stage procedure without ileostomy, was reported with a mean follow-up of 5.1 years. [218] They noticed an anastomotic leak rate of 12% (23 patients), with 5% (9 patients) requiring a diverting ileostomy; ileal pouch–anal anastomosis function was excellent in 19 of these patients, although 2 required permanent ileostomies. Daytime stool control was 95% and nighttime control was 90%. In patients taking steroids, there was no significant difference in complication rate.

Similar results were reported by Mowschenson and colleagues. [219] In 102 patients they reported a 9.8% anastomotic leak and subsequent diverting ileostomy rate. Overall 74.8% of their patients reported total satisfaction, and 84.7% regarded themselves as being in perfect health. Once again patients on steroids did not experience a higher complication rate. These results are in contrast with several studies where patients on steroids either had a higher incidence of anastomotic complication, [220], [221] and [222] or were not even considered candidates for a one-stage procedure. [223]

The Mayo Clinic group [164], [220] and the Cleveland Clinic group [165], [221] are strong advocates of a two- or three-stage operation, especially in patients on more than 20 mg of prednisone. When comparing the two procedures these studies reported septic complications in up to 22% of the nondiverted group [220] and it was noted that half of the nondiverted patients with septic complications required relaparotomy for diversion and drainage, whereas none of the patients in the diverted group with septic complications required surgery of any kind. [221] We believe that a very select group of patients may benefit from the avoidance of a temporary stoma and its associated complications. [223] Healthy, young patients, not on immunosuppressants or steroids, whose surgery was uneventful and who show minimal tension on the anastomosis, are better served by a one-stage procedure. We usually leave a rectal tube in these patients for few days to avoid pouch distention and perianal skin irritation due to the initial diarrhea.

Proctocolectomy With Continent Ileostomy

The proctocolectomy with continent ileostomy, or Kock pouch, offers an alternative to an ileal pouch–anal anastomosis for few and selected patients with ulcerative colitis. Specifically, this operation should be offered in specialized centers to patients with a locally advanced low rectal cancer that will need adjuvant therapy postoperatively. Patients who already have a Brooke ileostomy, after proctocolectomy, and wish to have a continent ileostomy; patients not candidate to an ileal pouch–anal anastomosis because of poor sphincter function; patients who prefer a continent ileostomy to an ileal pouch–anal anastomosis as a personal choice; and patients who have failed an ileal pouch–anal anastomosis but prefer a continence-preserving procedure to a Brooke ileostomy are all candidates. [224], [225] Contraindications to this procedure include older age, Crohn's disease, obesity, patients who are critically ill, and those who are deemed psychologically unfit. This procedure has also been performed in the pediatric population with satisfactory results. [226], [227]

The distal 30 cm of the terminal ileum are used to form the internal Kock pouch. Intussuscepting the terminal ileum backward into the pouch and anchoring the intussusceptum in place with staples creates a valve that prevents distal outflow from the pouch. The end of the terminal ileum is brought to the skin surface as a flash stoma (Fig. 84-22). The patient empties the pouch intermittently by passing a catheter through the stoma and valve into the pouch. Between intubations, the pouch is continent to both gas and stool.

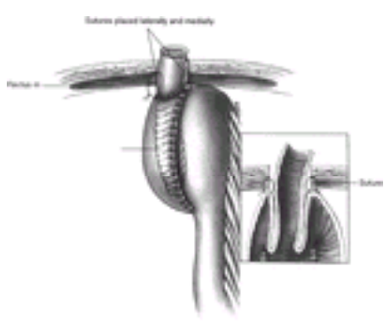

FIGURE 84-22. Kock pouch. (From Cohen Z, McLeod RS. Proctocolectomy with Kock pouch. In: Michelassi F, Milsom JW, eds. Operative strategies in inflammatory bowel disease. New York: Springer-Verlag, 1999:215.)

Kock pouch procedures have fallen out of favor in recent times, as the majority of patients with ulcerative colitis undergo an ileal pouch–anal anastomosis. The largest series reporting on the Kock pouch were published in the late 1970s. [224], [228], [229] Overall long-term follow-up showed excellent results, between 70% and 89% continence for gas and stool, with 95% of these patients free from wearing an appliance. [224], [228], [229] The two main long-term problems with continent ileostomy are malfunction of the valve and pouchitis. Malfunction of the valve causes incontinence and difficult intubation of the pouch, resulting in reoperation. Between 11% and 20% of patients developed this problem, [224], [228], [229] precipitating the need for valve revision. The incidence of pouchitis is nearly identical to that after ileal pouch anal anastomosis, and management is similar.

Kohler and colleagues [230] evaluated quality of life in 406 patients with Brooke ileostomy, 313 with Kock pouches, and 298 with ileal pouch–anal anastomosis. They found that patients with ileal pouch–anal anastomosis had fewer restrictions in sports and sexual activity than the Koch pouch group, which in turn had fewer restrictions in these activities, but more travel restrictions than patients with Brooke ileostomy. Overall as far as social life, recreation, work, and family activities there was no difference between those groups and more than 90% of patients in each group were satisfied with the results. [230]

Total Abdominal Colectomy With Brooke Ileostomy

In patients who are too ill to withstand a definitive procedure for ulcerative colitis, a total abdominal colectomy with end ileostomy is warranted. [231], [232], [233] and [234] Acute toxic colitis nonresponsive to medical therapy [232] with or without megacolon or perforation or hemorrhage is the primary indication for a total abdominal colectomy, end ileostomy, and either a rectal or rectosigmoid Hartmann stump or mucous fistula. This procedure eradicates the disease, requires no bowel anastomosis or pelvic dissection, and allows the patients to be weaned off medications.

Patients who are potential candidates for a total abdominal colectomy are those who carry a diagnosis of indeterminate colitis. This preliminary step allows the pathologist to obtain a final diagnosis after reviewing the resected colon, thus avoiding performance of an ileal pouch–anal anastomosis in a patient with Crohn's disease. As mentioned earlier, this procedure may also be indicated in the obese patient as a first step toward an ileal pouch–anal anastomosis. In these patients, a total abdominal colectomy with Brooke ileostomy eradicates most of the active disease, allows for decrease and eventual cessation of any steroid therapy, and for the possibility of successful weight reduction.

The abdominal colectomy with ileostomy entails resection of the entire colon with exteriorization of the terminal ileum as an end ileostomy. Distally the colon is usually transected at the level of the distal sigmoid or upper rectum, based on the surgeon preference. Controversy persists regarding the management of the rectal stump. Some authors prefer to place it as a closed rectal stump into the subcutaneous tissue of the midline abdominal incision, others leave it as an open mucous fistula through a separate abdominal wound; [233] the majority prefer closure of the intestinal stump and intrapelvic placement (Hartmann pouch). [232], [233] Only small comparative studies have examined this issue. Carter and associates [231] suggested that exteriorization of the closed rectal stump following total abdominal colectomy is associated with fewer pelvic septic complications and minimal local morbidity, facilitates subsequent pelvic dissection, and is not associated with increased disease activity in the retained rectum. Yet, the increased incidence of wound infections mitigates against this strategy as routine. A surgeon will be well served by choosing appropriately, using the different options after having evaluated the patient's general condition, the presence or absence of generalized peritonitis, and the local status of the rectosigmoid.

The removal of the vast majority of the diseased intestine and the diversion of the fecal stream from the residual disease allow these patients to recover from the debilitating effects of high-dose medications, [232] malnutrition, and sepsis. Delaying the ileal pouch–anal anastomosis does not affect postoperative functional results. [234]

Colectomy With Ileorectal Anastomosis

Colectomy with ileorectal anastomosis is only performed in few selected patients. The obvious advantages of this procedure are its relative simplicity, the preservation of the anal route of defecation, and the maintenance of fecal continence in all but the most debilitated patient. In addition, the risk of pelvic nerve dysfunction and perineal wound problems is virtually eliminated. Yet, this operation does not excise the diseased rectum, which maintains a risk of neoplastic transformation and may continue to cause symptoms. Indications for this conservative approach include patients with relative rectal sparing at high medical risk or older patients who are not good candidates for ileal pouch–anal anastomosis; in addition, patients with metastatic colon carcinoma in ulcerative colitis are good candidates due to their limited life expectancy. [86]

Contraindications include severely diseased rectum, dysplasia or nonmetastatic rectal carcinoma, and poor sphincter function; noncompliant patients are subject to lifelong surveillance. [147]

Based on the patient's medical condition and presentation, colectomy and ileorectal anastomosis may be performed as a one-stage procedure or as a two-stage procedure in those requiring emergency surgery. This operation can also be done by laparoscopic techniques.

The presence of pelvic anastomosis puts these patients at risk of septic complications due to an anastomotic leak, which occurs in about 4% of patients. [86] Dehiscences can manifest themselves in the immediate postoperative period as diffuse peritonitis or later, as abscess or fistula. When diffuse peritonitis occurs, patients need to be emergently reexplored, drained, and diverted; if the dehiscence manifests itself as a pelvic abscess, this can be treated by percutaneous, transrectal, transvaginal, transabdominal, or transperineal drainage, depending on its location, and, possibly, proximal fecal diversion. Patients who have suffered an anastomotic dehiscence have worse functional results with more than eight stools per day, continued need for steroids, and incontinence. Eventually, between 12% and 56% of these patients require proctectomy within a few years. [86], [235], [236]

Long-term poor function can also be due to recurrent or worsening proctitis. Endoscopy and biopsy can guide in the diagnosis and treatment. Persistent disease requires either topical or systemic therapy and, ultimately, proctectomy may be necessary. [147]

The risk of cancer in the rectal remnant has been reported to be 15.3% at 12 years, [86] but it varies considerably between series [235], [236] due to the difference in follow-up rates. The risk, however, is major, considering that most of these patients are relatively young.

More than 90% of patients with a functioning ileorectal anastomosis considered their health status to be better than before surgery. [86] Comparative studies, however, have shown no major benefit of colectomy and ileorectal anastomosis over ileal pouch–anal anastomosis in terms of postoperative morbidity, mortality, and long-term function. [237], [238]

Laparoscopic Surgery for Ulcerative Colitis

Skepticism still exists regarding laparoscopic management of patients with ulcerative colitis. Several authors believe that laparoscopic total colectomy is associated with higher morbidity than the open approach [132] and that restorative proctocolectomy does not have the same short-term benefits as seen in other laparoscopic colorectal procedures. [135] Critics of laparoscopic colorectal surgery cite a few early, small studies which suggest that laparoscopy offers no advantages compared with open surgery in terms of return of bowel function or length of hospital stay in ulcerative colitis. [132], [239], [240], [241] and [242] They furthermore claim that laparoscopy increases operative time and cost, and may even have a higher complication rate.

However, as experience in laparoscopy has increased, both technique and instrumentation have improved. Laparoscopic-assisted restorative proctocolectomy has been shown to be technically safe and feasible by various authors. [243], [244] In a case-matched series comparing laparoscopic and open restorative proctocolectomy procedures, Marcello and colleagues [244] have shown that return of intestinal function and length of hospital stay are reduced in the laparoscopic group without increase of morbidity or mortality.

Different techniques have been proposed for laparoscopic-assisted restorative proctocolectomy. [242], [244] Usually the entire colon is mobilized intracorporeally; the vessels are divided either intracorporeally or extracorporeally through a Pfannenstiel incision, based on surgeon preference and skills. The rectal dissection can be

accomplished intracorporeally, but some authors argue that, since a Pfannenstiel incision is needed to extract the specimen, the dissection should be safely and expeditiously performed through that approach. [243] Once the specimen has been delivered through the incision, the rectum is divided at the top of the anal canal using a linear stapler and an ileal pouch is constructed. A double-stapled anastomosis is then fashioned in a standard procedure. The use of a diverting loop has been advocated after laparoscopic-assisted ileal pouch–anal anastomosis. [239], [242], [244] Given that techniques in laparoscopic large bowel surgery are evolving rapidly, the role of this operation in the surgical treatment of patients with ulcerative colitis is likely to expand in the future.

INDETERMINATE COLITIS

A definitive pathological diagnosis is not feasible in 5% to 20% of patients with pancolitis as the only manifestation of inflammatory bowel disease, even after the entire proctocolectomy specimen has been analyzed. [70], [245], [246], [247] and [248] This group of patients has been classified as having indeterminate colitis. The fear that many of these patients with indeterminate colitis may actually harbor Crohn's disease [249] has created uneasiness in the ultimate treatment recommendation. [70] The largest series of patients with indeterminate colitis with a long-term follow-up after ileal pouch–anal anastomosis is from the Mayo Clinic. [248] The authors evaluated 82 patients with indeterminate colitis with a median follow-up of 83 months. They noticed that 15% of patients with indeterminate colitis and only 2% of patients with ulcerative colitis, as a control group, had their original diagnosis changed to Crohn's disease. At 10 years, patients with indeterminate colitis had significantly more episodes of pelvic sepsis, pouch fistula, and pouch failure (27%) than patients with ulcerative colitis. [248] Marcello and colleagues [245] also reported a 13% incidence of Crohn's disease in patients with an initial diagnosis of indeterminate colitis and a 37% pouch failure rate in this group.

Considering that approximately 15% of patients with indeterminate colitis will develop Crohn's disease over time, Bodzin and colleagues [250] advocated ileorectal over ileal pouch–anal anastomosis in patients with indeterminate colitis. Yet, as approximately 85% of patients with indeterminate colitis will remain free of Crohn's disease, we believe that, unless there are preoperative signs suggestive of Crohn's disease, [97] ileal pouch–anal anastomosis, rather than ileorectal anastomosis, should be considered as an option in the treatment of these patients. Patients should be informed preoperatively that they face a higher risk of development of Crohn's disease, but should also be reassured that, after ileal pouch–anal anastomosis, they will experience long-term outcomes identical to those of patients with ulcerative colitis, [248] in the absence of development of Crohn's features.

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CHAPTER 85

R. Balfour Sartor, Mark E. Murphy, and Edward Rydzak

MISCELLANEOUS INFLAMMATORY AND STRUCTURAL DISORDERS OF THE COLON

A spectrum of colonic inflammatory conditions can be distinguished from ulcerative colitis and Crohn's disease by clinical, endoscopic, and histological characteristics, although there is some overlap due to the limited repertoire of pathological and physiological responses to injury (Table 85-1). This chapter discusses conditions distinct from idiopathic inflammatory bowel diseases (IBD), explores radiation, ischemic, and infectious enterocolitis and provides distinguishing characteristics to aid in their differential diagnosis and treatment, and emphasizes pathophysiology and etiologic theories.

TABLE 85-1 Clinical and Pathological Characterization of Inflammatory Disorders of the Colon*

COLLAGENOUS AND LYMPHOCYTIC COLITIS

Clinical Features

Collagenous and lymphocytic colitis are characterized by chronic watery diarrhea with normal endoscopic and radiographic evaluations but histological evidence of chronic mucosal inflammation. These disorders represent two extremes of the spectrum of “microscopic colitis.” Collagenous colitis is diagnosed by increased subepithelial collagen deposition and chronic mucosal inflammation while the only histological abnormality of microscopic colitis is increased numbers of mucosal mononuclear cells. [1], [2], [3] and [4] The term lymphocytic colitis was proposed by Lazenby and colleagues [6] to distinguish microscopic colitis from mild IBD and infectious colitis. Although these disorders share certain epidemiologic features (Table 85-2) and 35% to 40% have associated arthritis, autoimmune disorders, and celiac disease, [4], [5], [7] collagenous colitis tends to have a slightly higher age of onset, female predominance, and influence of smoking. [7], [8] and [9] Collagenous and lymphocytic colitis can occur in children [10] and [11] and at least 25% of cases are diagnosed before the age of 45. The predominant symptom is chronic nonbloody diarrhea, 300 to 1700 g/24 hours, [12] with 50% of patients having more than five stools per day. Nocturnal stools are common, and patients report occasional fecal incontinence and decreased stool volume with fasting. Associated symptoms include mild crampy abdominal pain, nausea, mild weight loss, and abdominal distention. [7] Fever and visible rectal bleeding are unusual. The diarrhea usually has an insidious onset and is persistent, between 2 months and 20 years in duration, with a fluctuating clinical course marked by spontaneous relapses and remissions in 90% of patients. [7], [8] Despite increased use of colonoscopic biopsies to evaluate patients with chronic diarrhea, these disorders remain relatively unusual with an incidence of 1 to 3 per 100,000 population. Routine laboratory tests are usually normal, although patients may occasionally exhibit mild anemia, hypoalbuminemia, increased sedimentation rate and, rarely, steatorrhea. Fecal leukocytes are present in approximately half of patients with collagenous colitis [13] although stool lactoferrin is rarely increased.

	COLLAGENOUS COLITIS	LYMPHOCYTIC COLITIS
Incidence (per 100,000 population)	1.0–2.3	3.1
Gender: male:female	1:3–1:10	1:1–1:2
Mean age of onset	55–65 y	51–63 y
Smoking: active	25%	14%
former	8%	23%

TABLE 85-2 Epidemiologic Features of Collagenous and Lymphocytic Colitis

Pathology

Collagenous and lymphocytic colitis are both characterized by moderate numbers of mononuclear cells (lymphocytes, plasma cells, and macrophages) infiltrating the mucosa, with epithelial cell damage and preservation of the crypt architecture (Fig. 85-1). Intraepithelial lymphocytes are increased five- to tenfold (20–25 lymphocytes/100 epithelial cells). [6] Flattened surface epithelial cells, occasionally with a syncytial appearance, decreased numbers of goblet cells, Paneth cell hyperplasia, and increased epithelial mitoses are common histological features. [6], [13] In some studies, eosinophils and mast cells are increased [14], [15] with eosinophilic cationic protein in the lumen, [16] although neutrophils, cryptitis, and crypt distortion are rarely present. Collagenous colitis is histologically differentiated from lymphocytic colitis by a linear subepithelial collagen layer (Fig. 85-1A) which is greater than 10 µm thick and can extend to 100 µm. [1], [17] The normal basement membrane is less than 4 µm thick. The thickened collagen layer is most prominent beneath the surface epithelium between crypts and is frequently penetrated by capillaries. Linearly arranged type IV collagen constitutes the basement membrane in normal subjects. The composition of the thickened band in collagenous colitis is controversial; in one study types I and III collagen and fibronectin predominated [18] but in another report type IV collagen and tenascin were more prevalent. [19] Although cellular infiltration in both disorders is pancolonic, distribution of the thickened collagen table is spotty, being most prevalent in the cecum (82% of cases) and transverse colon (83% of cases) and least likely to be detected in the rectum (72% of cases). [17] Marked variability in simultaneous samples from the same patient and regional distribution of lesions suggest that reports of resolution and development of collagenous colitis over time with serial biopsies [20], [21] could reflect sampling error.

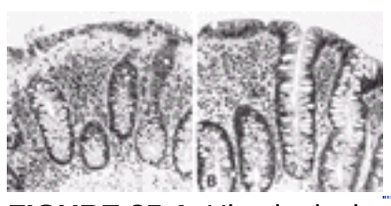

FIGURE 85-1. Histological comparison of lymphocytic and collagenous colitis. **A:** Collagenous colitis is characterized by a thickened subepithelial collagen layer as well as the inflammatory features of increased plasma cells in the lamina propria and a damaged surface epithelium with increased epithelial lymphocytes. **B:** Lymphocytic colitis shows all the inflammatory features of collagenous colitis but lacks the subepithelial collagen layer. (Hematoxylin and eosin stains; original magnification ×225. Courtesy of Audrey Lazenby, M.D., Baltimore, MD.)

Etiology and Pathogenesis

These disorders remain idiopathic, but seem to have an immune-mediated basis by virtue of infiltrating $CD8^+$ T-cell receptor a///ß intraepithelial lymphocytes and $CD4^+$ lamina propria T lymphocytes. The antigens stimulating these T cells are unknown, but are presumably luminal in origin, since diverting the fecal stream decreases inflammation. [22] Inflammation is diffuse and superficial, and 60% of patients respond to antibiotics. [7] Infections with *Clostridium difficile* toxin or *Campylobacter jejuni* have been postulated as environmental triggers initiating this process, possibly accounting for the abrupt onset seen in a minority of cases, and direct toxic effects of bacterial products must be considered to explain therapeutic responses to bismuth subsalicylate. [23] The usual benign prognosis with eventual resolution of symptoms [7], [8] raises the possibility of a protracted postinfectious injury.

The relationship of these disorders to celiac disease remains unresolved. Patients with celiac disease whose diarrhea does not respond to a gluten-free diet have a high incidence (approximately 75%) of lymphocytic colitis. [24], [25] However mild colonic inflammation is less common in untreated, unselected patients with celiac disease (20%), [24], [25] the majority of patients with microscopic colitis do not exhibit malabsorption, and no clinical improvement is noted with a gluten-free diet in the majority of patients with microscopic colitis. However, patients with collagenous/microscopic colitis have similar human leukocyte antigen (HLA) class II haplotypes as celiac disease and have a relatively high frequency of small bowel inflammation and celiac disease serology. [26], [27]

An autoimmune etiology is suggested by the frequent association of arthritis, thyroid abnormalities, asthma and diabetes, [7] perinuclear antineutrophil cytoplasmic antibodies (pANCA) in 20% of cases, [28] and a slightly increased frequency of antinuclear antibodies. [29] Patients with diabetes seem to be at particular risk, with thickening of the subepithelial collagen plate even in the absence of diarrhea. [30] A minority of patients appear to have extracolonic abnormalities of fibrogenesis, with reports of collagenous gastritis [31] and fibrosis in salivary glands leading to the sicca syndrome. [32] In situ hybridization and enzymatic studies show no increased collagen production by subepithelial myofibroblasts, decreased metalloproteinases, and increased inhibitors of metalloproteinases, suggesting defective collagen remodeling rather than enhanced synthesis. [19], [33]

The strong association of arthritis with these disorders raises the possibility that nonsteroidal antiinflammatory drugs (NSAIDs) may play an etiologic role. In a well-controlled study, significantly more patients with collagenous colitis (61%) had used NSAIDs for more than 6 months than age- and gender-matched controls (13%; $P < 0.02$). [34] In uncontrolled studies, up to one third of patients in two collagenous colitis registries had used NSAIDs. [7], [35] NSAIDs can cause gastric, small bowel, and colonic inflammation in humans and experimental animals. [36] Given the high frequency of NSAID use in a 60-year-old population and the relative rarity of collagenous colitis and lymphocytic colitis, other factors must be important. The possibility of genetically determined host susceptibility factors was raised by reports of familial associations of microscopic colitis [37], [38] and of differential susceptibility of inbred rat strains to indomethacin-induced enterocolitis. [39] Of interest, collagenous and lymphocytic colitis can occur in the same family. [37], [38]

The possibility that luminal bile acids injure colonic epithelia in these disorders is raised by clinical responses to cholestyramine and the demonstration of bile salt malabsorption in 60% of patients with lymphocytic colitis and 27% to 42% of those with collagenous colitis. [40], [41] Finally, the ability of ingested substances to injure the colon is illustrated by clinical and experimental inflammation induced by L-tryptophan. [42] Future studies must concentrate on the role of environmental agents, especially NSAIDs, and host genetic factors in the etiology of these disorders.

Diarrhea in microscopic colitis is caused by defective active and passive absorption of sodium and chloride and reduced chloride-bicarbonate exchange in the colon with coexisting abnormal small intestine fluid and electrolyte absorption in one-third of patients. [2] Active colonic chloride secretion associated with increased luminal prostaglandin E_2 and nitric oxide concentrations [43], [44] and prevention of diarrhea by an H_1 antagonist in a patient with increased numbers of mast cells [14] suggest that soluble mediators produced by activated immune and mesenchymal cells mediate epithelial cell absorptive dysfunction. This hypothesis is supported by correlation of diarrhea with increased numbers of mucosal inflammatory cells rather than thickness of the collagen table. [45] The latter observation refutes the theory that subepithelial collagen blocks electrolyte and water absorption.

Diagnosis and Differential Diagnosis

In patients with chronic watery diarrhea whose endoscopic, radiographic, microbial, and endocrine evaluations are normal, lymphocytic and collagenous colitis are diagnosed by characteristic histological features. A minority (29%) of patients with collagenous colitis have nonspecific endoscopic abnormalities including erythema, edema, or abnormal vasculature. [7] Multiple biopsies must be taken from different colonic segments to reliably diagnose collagenous colitis. [13], [17] Biopsy of multiple segments demonstrated a single positive site in 5 of 17 patients and a greater than 50% variability of collagen thickness in multiple samples taken simultaneously. [17]

Biopsies on flexible sigmoidoscopy are usually sufficient to make the diagnosis, but random biopsies from the right and transverse colons will increase the yield by 5% to 10%. Proper orientation of the specimen is essential to prevent tangential sectioning leading to artifactual thickening of the subepithelial basement membrane; the presence of associated lamina propria inflammation is an important co-diagnostic feature of collagenous colitis. [46]

Ulcerative colitis and Crohn's disease can be differentiated from microscopic colitis by diagnostic colonoscopy, radiography, and histological features. Infectious agents must be excluded by stool ova and parasite examinations, standard fecal bacterial cultures, and *C. difficile* toxin assays. In addition, *Giardia* infestation and small bowel bacterial overgrowth can be evaluated by jejunal aspirates which may be taken at the time of small bowel biopsies performed to rule out celiac disease and Whipple disease. Although not a characteristic feature of lymphocytic colitis and collagenous colitis, malabsorption may be present. Hormone-producing tumors, laxative abuse, ischemic colitis, amyloidosis, and hyperthyroidism must be considered in the differential diagnosis. Irritable bowel syndrome can be differentiated by normal stool volume and colonic biopsies.

Several disorders resembling lymphocytic colitis and collagenous colitis can be differentiated by careful histological analysis. [46] Acute infectious colitis is identified by mucosal edema, neutrophilic infiltration, and a paucity of intraepithelial lymphocytes. Ulcerative colitis and Crohn's disease biopsies have crypt abscesses, neutrophils, and possible granulomas with few intraepithelial lymphocytes. Pericrypt eosinophilic enterocolitis shares the characteristics of chronic watery diarrhea and normal endoscopy but lacks increased intraepithelial and superficial lamina propria lymphocytes and is associated with eosinophilic infiltration, crypt foreshortening, and inflammation in the deep lamina propria and muscularis mucosa. [47] Ischemic colitis, radiation colitis, and solitary rectal ulcer syndrome (SRUS) have diffuse fibrosis in the lamina propria, and colonic collagen deposition in progressive systemic sclerosis occurs along all basement membranes and may be transmural. Similarly, amyloid deposits, which have a characteristic histochemical staining pattern, occur diffusely along the basement membrane of the crypts, blood vessels, and surface epithelium.

Treatment

A variety of medical therapies have been used in collagenous and lymphocytic colitis with reported benefit, [7], [48] although only prednisone [49] and bismuth subsalicylate [23], [50] have been subjected to placebo-controlled trials due to the small number of cases at any single institution. Because the course of these disorders is usually benign, it is prudent to start with the least toxic interventions prior to sequentially progressing to more toxic drugs such as prednisone and immunosuppressive agents. Bohr and colleagues [7] have demonstrated that prednisolone had a somewhat higher response rate in patients with collagenous colitis than 5-aminosalicylate (5-ASA) products and antibiotics (82% vs. 50%–67%), but most patients responding to steroids had recurrence of symptoms immediately after stopping treatment. Some studies have suggested excellent responses to relatively nontoxic bismuth subsalicylate [23], [50] and oral budesonide, [7], [51], [52] and [53] as well as to 6-mercaptopurine and azathioprine, [54] although larger trials must be conducted before widespread use of these agents. Ung and associates [41] demonstrated an excellent clinical response to cholestyramine in unselected patients, and a response rate of 92% in those patients with documented bile acid malabsorption. Given the chronic, slowly progressive nature of these disorders, a progressive approach beginning with the least toxic agents is suggested (Table 85-3).

TABLE 85-3 Sequential Approach to Treating Microscopic Colitis

Prognosis

Although chronic, symptoms tend to spontaneously wax and wane. The prognosis is excellent, with the majority of patients responding to treatment, although it appears that collagenous colitis is somewhat more resistant to therapy than is lymphocytic colitis. [8], [48], [55], [56] and [57] The presence of Paneth cell hyperplasia has been postulated as a marker of refractory disease. In contrast to ulcerative colitis and Crohn's disease, Chan and colleagues [58] reported that microscopic colitis is not associated with colonic adenocarcinoma, although the mean follow-up of 7 years may not be adequate for full expression of cancer risk. However, there is no rationale for periodic colonoscopic surveillance for dysplasia in lymphocytic or collagenous colitis.

Overlap or Distinct Diseases?

It is unclear whether lymphocytic and collagenous colitis are two separate but similar diseases or different presentations of a single disorder. [45] These syndromes share similar clinical, demographic, and histological characteristics and differ primarily by the presence or absence of a thickened subepithelial collagen plate. Serial biopsies show resolution or development of this collagen band with time [20], [21]; however, the spotty nature and segmental distribution of the collagen plate [17], [45] make it doubtful that lymphocytic colitis can develop into collagenous colitis. The thickness of the collagen table follows a multimodal distribution and does not increase with age or with duration of symptoms [45] suggesting that collagen deposition is not merely a consequence of active inflammation and the natural progression of long-standing lymphocytic colitis. Moreover, in small studies, the HLA associations of lymphocytic colitis (HLA-A1) and collagenous colitis (HLA-A2) are distinct, [59] and a higher female:male gender ratio and smoking history is present in collagenous colitis. [7], [8] This controversy can only be resolved by more complete understanding of the origin, pathogenesis, and natural history of these disorders.

DIVERSION COLITIS

Clinical Features and Natural History

Inflammation insidiously develops in the distal bypassed colon within Hartmann pouches or mucus fistulae after exclusion of the fecal stream. Most patients with proximal colostomies or ileostomies created for treatment of cancer, diverticulitis, Crohn's disease, ulcerative colitis, Hirschsprung's disease, or trauma have no symptoms from their bypassed colonic segment. However, approximately one third of patients develop frequent mucoid discharges which may progress to rectal bleeding and pain 1 to 9 months after fecal diversion. [60], [61] Symptoms are more prevalent in patients operated on for IBD than for carcinoma or other indications. [62] Histological abnormalities are apparent in almost all bypassed segments. [60], [61], [62], [63], [64] and [65] In a prospective study, endoscopic evidence of inflammation was found in 91% of patients. [63] Colitis was mild in 52% of cases, moderate in 44%, and severe in only 4%, although symptoms were present in only 6% of patients. Endoscopic features include diffuse erythema, granularity, friability, aphthous ulcers, and exudates, with nodularity and diffuse ulceration in more advanced cases. Clinical, endoscopic, and histological abnormalities appear to be progressive, with mild changes at 3 months [64] and more severe findings by 6 to 9 months after proximal fecal diversion. [60] Most patients, however, particularly those without preoperative Crohn's disease, do not develop debilitating symptoms. Anal sphincter function remains normal, although rectal volume diminishes by 3 months postoperatively. [64] Usually the inflammatory process completely resolves soon after restoration of the fecal stream.

Pathology

A histological spectrum of inflammation is present, ranging from mild follicular hyperplasia and lymphoplasmacytic infiltrates to severe inflammation. Diffuse follicular lymphoid hyperplasia with frequent germinal centers is a characteristic feature of all stages of diversion colitis. [63], [65], [66] and [67] Early inflammatory changes consist of mucosal infiltration by lymphocytes, plasma cells and neutrophils, aphthous ulcers overlying lymphoid aggregates, and reactive epithelial cells. Immunohistochemical studies show increased numbers of B and T lymphocytes within lymphoid follicles and in the lamina propria, but no increase of macrophages and plasma cells. [66] More advanced lesions consist of crypt abscesses, relatively mild crypt architectural changes, and Paneth cell metaplasia. Mucin granulomas are present in a minority of cases. In severely involved resected tissues, gross abnormalities include diffuse nodularity and minute ulcerations with exudate. [66] Large ulcers and transmural changes are not typically present. These features are recapitulated in distal colons in rats with fecal diversion. [68]

Pathogenesis

Considerable clinical and experimental evidence incriminates luminal nutrient deficiency as the cause of diversion colitis. Roediger and colleagues [69], [70] have demonstrated that lumenal short-chain fatty acids (SCFAs), metabolic products of carbohydrate and peptide fermentation by anaerobic bacteria, are the principal fuels for the distal colonocyte. Butyrate provides the majority of oxidative energy for the rectal epithelial cell; acetate, ketone bodies, and glutamine are alternative sources. Harig and colleagues [71] demonstrated that excluded colonic segments contained negligible concentrations of SCFAs and that infused carbohydrates were not metabolized to SCFAs. Moreover, instillation of SCFA enemas twice daily induced clinical, endoscopic, and histological remissions in four patients with diversion colitis. Butyrate has multiple effects on cultured colonic epithelial cell lines, including accelerated differentiation, increased constitutive expression of secretory component and HLA class I antigens, and down-regulation of tumor necrosis factor (TNF) and interleukin-1 (IL-1)-induced cytokine expression. [72] Although this theory has achieved widespread acceptance [73] and is supported by the observations that colonic epithelia in ulcerative colitis exhibit aberrant SCFA metabolism and respond to SCFA enemas, [74] it should be noted that SCFA therapy of diversion colitis is not always successful. [75] In addition, factors other than SCFAs may be involved in the pathogenesis of diversion colitis. Mucosal atrophy, rather than inflammation, occurs in germ-free rodents and rats receiving long-term parenteral nutrition or elemental diets [76] and urinary colon conduits (ureterosigmoidostomies) that are not in continuity with the fecal stream, [77] conditions in which lumenal SCFAs would be predicted to be quite low. The fact that diversion colitis rapidly resolves after reanastomosis to restore bowel continuity suggests that lumenal factors provide protection from inflammation. Alternatives to the SCFA hypothesis are that lumenal growth factors, dietary constituents, or other bacterial metabolic products are necessary to maintain optimal colonocyte function, or that post-exclusion alterations in bacterial profiles (dysbiosis) induce damage. In support of dysbiosis, normal anaerobic bacterial concentrations are significantly diminished, certain enterobacterial strains proliferate in the excluded colon, [78] and aerobic nitrate-reducing bacteria increase in frequency. [79] Colonic diversion in rats resulted in crypt atrophy, epithelial erosions, and mucosal inflammation associated with qualitative and quantitative changes of enteric bacteria and alterations in distribution and expression of sulfomucins and sialomucins [68] These observations raise the possibility that mucosal barrier function is diminished, leading to enhanced uptake of lumenal bacterial antigens which activate T and B lymphocytes and innate immune cells. [66]

Diagnosis and Differential Diagnosis

Evaluation of inflammation in the diverted colon is relatively straightforward in the patient who had no preoperative intestinal inflammation. Flexible sigmoidoscopy with biopsies and examination of luminal aspirates for ova and parasites, bacterial culture, and *C. difficile* toxin are usually adequate to establish the diagnosis and evaluate activity of inflammation. Radiographic contrast studies can be performed but add little additional information unless a fistula or abscess is suspected, in which case a computed tomography (CT) scan may be helpful. Visualization of the bypassed colonic segment must be included as part of periodic screening after resection of colonic adenocarcinomas, because recurrent cancers and polyps can cause bleeding in the excluded bowel. [62] Radiation or ischemic colitis must be considered in appropriate clinical situations.

The major diagnostic challenge is to differentiate recurrent Crohn's disease from severe diversion colitis, which is an important distinction if reanastomosis is being considered. Advanced diversion colitis has focal ulceration and a nodular appearance because of lymphoid follicular hyperplasia, which can mimic Crohn's disease. [65] Endoscopic features favoring recurrent Crohn's disease are longitudinal ulcers and possibly strictures, because mucosal ulcers are usually quite small, even in advanced diversion colitis. [66] Aphthous ulcers occur in both entities. CT scanning and endoscopic ultrasonography to measure rectal wall thickness has not yet been investigated in this setting. Histological features suggesting Crohn's disease are transmural inflammation, marked crypt architectural changes, and epithelioid granulomas. Granulomas can occur in diversion colitis but are usually mucinous. Lymphoid hyperplasia occurs in both disorders but is particularly prominent in diversion colitis. [80] The absence of preoperative rectal involvement in the patient with Crohn's disease can also be useful information; all four patients in the study by Korelitz and colleagues [81] regained their normal rectal appearance after reanastomosis, which is safe with mild to moderately active diversion colitis. [81], [82]

Treatment

Because most patients are asymptomatic or have only mild mucus discharge, therapy is rarely indicated. Although SCFA enemas (60 mmol/L acetate, 30 mmol/L propionate, and 40 mmol/L butyrate) induce a remission in some patients, [71], [83] SCFA enemas have not been beneficial in other studies, [75] are not yet commercially available in the United States, and have an unpleasant, odor-limiting compliance in some patients. Hydrocortisone enemas are usually not successful. [81], [75] 5-ASA enemas have induced an endoscopic and histological remission in one patient [84] and have been successful in our practices.

ENDOMETRIOSIS

Endometriosis, defined as the presence of endometrial glands and stroma outside the uterine cavity and musculature (myometrium), is a common disorder, occurring usually without symptoms in approximately 15% of menstruating women. [85] Approximately one third of patients who undergo surgical exploration for endometriosis have intestinal implants, usually in the rectosigmoid (95%), appendix (10%), and ileum (5%), with proximal colon locations uncommon. [86] Meyer [87] described the first case of rectal endometriosis in 1909.

Clinical Features

Most patients with serosal endometrial implants in the rectosigmoid have no specific intestinal symptoms apart from those symptoms typically experienced with pelvic endometriosis. Most patients with intestinal endometriosis have associated implants on their ovaries, anterior and posterior cul-de-sacs, uterus, and uterine ligaments. [85] Pelvic symptoms of dysmenorrhea, dyspareunia, infertility, and dysfunctional uterine bleeding almost always accompany and frequently overshadow intestinal symptoms. Patients with rectal serosal implants may have localized tenderness and palpable nodules on rectovaginal examination. Penetration of endometriomas into the bowel wall and adhesions can lead to symptoms of partial obstruction, including intermittent abdominal pain and constipation. [88], [89] and [90] Partial obstruction caused by compression of the lumen by submucosal endometriomas, fibrosis, and contraction of the mesentery usually occurs in the rectosigmoid area but may involve the ileum. Diarrhea and rectal bleeding have variable incidences, with grossly evident hematochezia present in 3% to 33% of patients who undergo surgical bowel resection. [88], [89] and [90] Bleeding is a rare event in nonoperative series. In a recent surgical series, presenting complaints in order of frequency included abdominal pain, mass, obstruction, rectal bleeding, infertility, diarrhea, and increasing urinary frequency. [90] Cyclic episodes of intestinal symptoms corresponding with menses occur in less than half of patients, although the vast majority (80%) of patients with intestinal endometriosis have associated gynecological symptoms. [91] There have been reports of acute appendicitis caused by intussusception or obstruction by an endometrioma. [92]

The average age of women undergoing surgical resection for intestinal endometriosis is 32 to 44 years, but ages range from 16 to 60 years. [88], [89] and [90] Endometriosis is found almost exclusively in women of reproductive age, with mean age of diagnosis at 25 to 29 years. [85] The somewhat older age of patients undergoing bowel resection probably is related to the chronic nature of the inflammation, which progresses to obstructive complications. Although it is unusual, postmenopausal women can develop symptomatic colonic obstruction, especially if they are treated with estrogen replacement or even tamoxifen. [93] Intestinal symptoms can persist after hysterectomy and oophorectomy if partially obstructing lesions are not resected at the time of initial surgical treatment. [92], [94]

Pathology

Serosal implants are common but usually do not produce specific symptoms unless they invade the intestinal wall. Peritoneal or serosal implants are classically described as bluish-gray nodules ranging from 2 mm to 2 cm in diameter, [85] although mural masses on resected tissue are larger, averaging 2.6 cm. [90] Secretory glands lined by cuboidal endometrial epithelial cells are surrounded by endometrial stroma embedded in fibrotic tissue. These glands can become cystic or be filled with blood. Endometrial deposits invading the bowel wall usually do not involve the mucosa but are associated with localized muscular hypertrophy and fibrosis (Fig. 85-2) [95] which may lead to strictures or asymmetric kinking of the bowel. Histological mucosal abnormalities are confined to the region of invading endometrial nodules and range from mild crypt distortion to active inflammation with crypt abscesses. [95] Mucosal ulceration and polypoid masses, which resemble primary intestinal inflammation or neoplasms, occur in 23% of resected tissues. [90]

FIGURE 85-2. A: Air-contrast barium enema shows an intramural filling defect in the sigmoid colon (*arrows*) which produced stenosis of the lumen. **B:** Operative specimen of the sigmoid colon demonstrates an intramural endometrium. The mucosa is intact but thinned, and the muscle layer is dramatically thickened. (From Croom RD, Donovan ML, Schweisinger WH. Intestinal endometriosis. Am J Surg 1984;148:660.)

Intestinal neoplasms can arise from endometrial implants, even after hysterectomy, particularly in the setting of unopposed estrogen therapy. [96], [97] At least 70% of these tumors involve the rectosigmoid region, but some arise from the ileum, paralleling locations of endometrial serosal implants. The most common type of malignancy is endometrial adenocarcinoma, followed by müllerian adenosarcoma and endometrial stromal sarcomas. [96], [97] These tumors arise from the serosal implants, but may extend into the mucosa.

Pathophysiology

Three theories of the genesis of endometriosis have clinical and experimental support. [98], [99] The *transplantation theory* suggests that dissemination of viable uterine epithelial and stromal cells by retrograde menstruation, vascular, lymphatic, or iatrogenic spread leads to implantation of cells in ectopic locations. Epithelial and stromal cells then proliferate under hormonal stimulation to form invasive nodules. Retrograde menstruation is an almost universal occurrence and the frequency of endometrial implants in the ovaries and dependent areas of the pelvis support dissemination by this route. The *coelomic metaplasia theory* suggests that peritoneal cells undergo metaplastic transformation to endometrial epithelial cells under estrogen stimulation. Although endometrial and peritoneal cells are derived from the coelomic-wall epithelium, the rarity of endometriosis in anovulatory women with persistently elevated estrogen levels and the low frequency of endometrial nodules within the thoracic cavity, which has a similar embryonic derivation, make this theory unlikely. The *induction theory* a combination of the first two theories, states that unknown secreted factors within the shed endometrium induce endometrial metaplasia. Evidence of endometrial-like glands adjacent to micropore filter chambers containing endometrial cells in rodents supports this theory, although the bulk of evidence supports the transplantation theory.

Regardless of which theory is correct, proliferation of endometrial glands and stroma is under hormonal influence. Estrogen stimulates growth, and progesterone inhibits it. A large variety of growth factors such as hepatocyte growth factor and transforming growth factor ß modulate hormonal influences, providing a mechanism by which local inflammatory and stromal cells can regulate growth of implanted tissues. [98], [100] Progressive invasion of intestinal muscle and submucosal layers leads to muscular hypertrophy and fibrosis resulting in segmental narrowing of the adjacent intestinal lumen and secondary obstructive symptoms. Smooth muscle hyperplasia, hypertrophy, and collagen deposition are almost certainly the result of stimulation by the same growth factors that regulate endometrial proliferation. Increased expression of IL-1a, IL-6, and IL-10 by ectopic endometrial tissue and the presence of microscopic inflammation [90], [101] support the concept of activated macrophages in the pathogenesis of this disorder. [98] Integrins such as the $a_V ß_3$ vitronectin receptor appear to be important in implantation. [102] Genetic predisposition is strongly suggested by increased familial incidence and dramatically increased concordance of endometriosis in monozygotic siblings compared with dizygotic sisters. [103]

Diagnosis

Clinical diagnosis of intestinal endometriosis can be difficult because of nonspecific symptoms. The classic presentation is partial obstruction of the colon in an infertile woman of reproductive age with progressive dysmenorrhea and dyspareunia. Cyclical symptoms associated with menses are suggestive but are present in less than half of patients. Most patients have symptoms of pelvic endometriosis. Tender nodules palpable in the rectovaginal septum (posterior cul-de-sac) on rectovaginal examination are highly suggestive of endometriosis, but fixation of the rectum is nonspecific. Radiographic and endoscopic examination are normal in the presence of noninvasive serosal implants. Most patients with obstructing endometriosis have extrinsic compression or smooth strictures with normal mucosa [104] (see Fig. 85-2A) and normal biopsy specimens. If rectal bleeding is present, endoscopic evaluation with biopsies has been reported to be diagnostic in four of six patients with histological confirmation in five. [105] CT and magnetic resonance imaging scans are usually nonspecific because of the small size of implants and cannot exclude carcinoma or lymphoma. [96] Endorectal ultrasonography can detect intestinal wall invasion by endometrial implants and distinguish these lesions from mucosal processes. [105], [106]

Definitive diagnosis is made by surgical exploration, either by laparoscopy or by celiotomy, and by tissue histological diagnosis. [88], [89] and [90] The vast majority of patients with intestinal endometriosis have multiple associated serosal implants on pelvic organs and the intestine.

Differential Diagnosis

Intestinal endometriosis with altered bowel habits is difficult to differentiate from irritable bowel syndrome, Crohn's disease, and invasive adenocarcinoma. [89], [90] The distinction can be suggested by associated infertility, dysmenorrhea and dyspareunia, presence of tender nodules on rectovaginal examination, and evidence of extrinsic rectosigmoid compression on flexible sigmoidoscopy or barium-contrast examinations. Laparoscopy or even surgical resection is usually necessary to establish the diagnosis. Strictures secondary to Crohn's colitis usually have associated mucosal ulceration and histological evidence of inflammation; these findings are rare in endometriosis. Of interest, ileal endometriosis has been reported to complicate established Crohn's disease. [107] Primary colonic carcinoma or lymphoma must be considered in all cases of bowel stricture regardless of age. Endoscopic and radiologic features favoring adenocarcinoma include mucosal ulceration and sharp margins; endometriosis is usually submucosal. [90] Additional clinical features suggesting carcinoma are advanced age (postmenopausal), hematochezia, multiparity, and lack of dysmenorrhea and tenderness. However, none of these features is absolute, particularly since carcinoma can arise from endometriosis even in postmenopausal with estrogen replacement, [96], [97] and resection with transmural histological examination is the only way to definitively rule out carcinoma. Rarely, mucosal involvement by endometriosis can simulate adenomatous polyps. [97] Long, smooth ischemic strictures are usually in the descending colon, whereas shorter eccentric endometrial strictures occur more distally in the rectosigmoid region. Diverticular masses and radiation-induced strictures occur in older age groups and can usually be differentiated on clinical grounds.

Treatment

Hormonal therapy or nonresective surgery can be used to treat superficial serosal implants without stricture. Current medical therapeutic options diminish estrogen stimulation by inducing pseudopregnancy, pseudomenopause, or chronic anovulation. Danazol, a 17a-ethinyltestosterone derivative, medroxy-progesterone, and gonadotropin-releasing hormone analogs are clinically comparable and superior to placebo in diminishing pain and size of pelvic endometrial nodules. [108], [109] Although Fedele and colleagues [110] reported excellent results in intestinal endometriosis with a 6-month course of luprolide, a gonadal-releasing hormone, all patients relapsed and these investigators suggested definitive surgical treatment. Ablation of infiltrating endometrial implants on the anterior rectum and rectovaginal septum can be safely accomplished with a laparoscopic carbon dioxide laser, [111] but most authors advocate segmental resection of partially obstructed colons because of poor results with medical therapy or superficial ablation and the inability to exclude carcinomas associated with endometriosis by clinical assessment. [91], [94], [97], [112], [113] Laparoscopic versus open explorative approaches to resection depend on the individual case and experience of the surgeon. [112], [114], [115] In postmenopausal women and those who do not desire pregnancy, hysterectomy and bilateral oophorectomy should be performed at the time of intestinal resection to treat associated pelvic endometriosis and to minimize the risk of recurrent disease. Patients who desire to preserve fertility should have excision or laser ablation of associated endometriosis in the pelvic organs in conjunction with bowel resection. Reports of successful pregnancies after surgical treatment vary, but results seem to be superior to those achieved with medical therapy. [85] However, recurrence of symptoms of endometriosis with preservation of ovarian function is substantial, and patients may subsequently develop obstructive intestinal symptoms despite medical therapy. [116]

DRUG- AND CHEMICAL-INDUCED COLONIC INJURY

Toxic colitis has been associated with numerous orally and rectally administered agents; in addition, certain drugs cause ischemic colitis, melanosis coli, and cathartic colon (Table 85-4). This section reviews the various types of chemical- and drug-induced colonic injury, with an emphasis on clinical findings and pathogenesis.

Enemas: soap, water-soluble contrast media (e.g., Gastrografin), hydrogen peroxide
Laxatives: melanosis coli, cathartic colon
Agents inducing ischemia: oral contraceptives, vasopressin, ergotamine, alosetron, cocaine, dextroamphetamine, neuroleptics, digitalis
Miscellaneous: nonsteroidal antiinflammatory drugs, selective cyclooxygenase X 2 inhibitors, penicillamine, gold, isotretinoin, antibiotics, chemotherapy, methyldopa, flucytosine, glutaraldehyde

TABLE 85-4 Colonic Injury Induced by Therapeutic Agents

Syndromes Associated With Enema Use

Colitis with a broad spectrum of clinical and endoscopic findings may result from rectal administration of various chemicals. High-risk groups for toxic colitis include patients with a history of self-mutilation or of self-medication for constipation, members of certain African tribes who ritualistically use herbal enemas, and those receiving soap enemas or water-soluble contrast media. Individuals with chemical colitis usually present with abdominal pain, bloody or nonbloody diarrhea, tenesmus, fever, and leukocytosis; rectal and abdominal tenderness, with or without peritoneal signs, may be seen on physical examination. [117], [118] and [119] Nonspecific endoscopic findings range from friable, granular mucosa to frank ulceration, with or without pseudomembrane formation. One endoscopic clue to toxin-induced colitis is the association of perianal excoriation with a predominantly distal colitis. [117] Treatment is primarily supportive, with intravenous fluids, bowel rest, and, if clinically indicated, broad-spectrum antibiotics. However, surgery may be necessary in some cases. [117], [119], [120]

Soap Colitis Syndromes of soap-induced colitis range from mild inflammation with increased stool frequency to severe acute co-litis with bloody diarrhea resembling idiopathic ulcerative colitis. [118] Acute colitis may heal with scarring and colonic cicatrization [120] or may progress to transmural necrosis and perforation. [121], [122] Endoscopic findings range from mucosal edema (with loss of normal vascular pattern) to mucosal sloughing and ulceration. [118] The pathophysiology of soap-induced colonic injury centers on its detergent effects on the colonic mucosa. Detergent enemas in animals caused acute liquefaction necrosis which resembled alkaline-induced corrosive esophagitis, with an acute necrotic phase (days 1–4), an ulceration/granulation phase (days 3–5), and a cicatrization phase (beginning in weeks 3–4). [120] The severity of damage in soap-induced colitis is related to both the soap concentration and the duration of mucosal contact. [118] Based on these well-described toxicities, there is no clinical role for soap enemas.

Water-Soluble Contrast Media Mild focal inflammation ranging to severe colitis with necrosis and perforation may be associated with several hyperosmolar water-soluble contrast media, including Gastrografin, Hypaque, and Renografin. [123] These agents are used to opacify partially obstructed colons and avoid the potential complications of barium with extraluminal leakage. Gastrografin is also used to treat meconium ileus in neonates with cystic fibrosis and severe fecal impactions in adults; its hypertonicity and the presence of the detergent Tween 80 are thought to facilitate stool passage by drawing fluid into the gut lumen. Gastrografin had a greater propensity for inducing colitis in animals than Hypaque, Renografin-76, or barium. [123] Tween 80 may damage the gut mucosa in a manner similar to soap enemas. Enemas containing Tween 80 in addition to Hypaque sodium 25%—which by itself did not cause substantial inflammation—increased the incidence and severity of colonic inflammation in rats. [123] These results, however, were not borne out in later animal studies. Animals without lumenal distention suffered no ill effects regardless of the medium used to fill the colon, causing Wood and colleagues [124] to postulate the pathogenetic importance of compromised mucosal blood flow in the distended gut. In humans, damage almost always occurs proximal to obstructing lesions, mainly in the cecum and ascending colon, [119], [125] raising the possibility that prolonged mucosal exposure and preexisting mucosal injury may be contributing factors.

Hydrogen Peroxide Hydrogen peroxide enemas used in the disintegration of impacted feces, treatment of meconium ileus, and removal of intestinal gas in preparation for radiologic study have been associated with bloody diarrhea, pneumatosis coli, colonic perforation, sepsis, and death. [126] The colitis may be caused by ischemia as a result of the entrance of gas into the loose connective tissues of the mucosa and submucosa and, eventually, into the vasculature, resulting in abolition of mucosal blood flow. In animal models hydrogen peroxide enemas resulted in the rapid formation of gas cysts in the mucosa, submucosa, and serosa, with subsequent absorption of the gas into the local circulation and eventual mucosal ischemia. [127], [128] Oxygen free radicals released by hydrogen peroxide may also contribute to mucosal damage.

Other Miscellaneous Substances Inadvertent ethanol enema administration has been reported to cause an acute hemorrhagic colitis with associated lower abdominal pain. [129] Self-administration of a hydrofluoric acid enema resulted in rectal pain and bloody diarrhea necessitating segmental resection of a necrotic, ulcerated sigmoid colon. [117] The ritual use of enemas by South African tribesmen —traditionally involving roots, herbs, tree bark, and other natural constituents—has been adapted to more modern agents, including vinegar, potassium dichromate, potassium permanganate, copper sulfate, unknown herbal medicines, and chloroxylenol (Dettol), which induced colitis ranging from mild rectal inflammation to transmural coagulative necrosis. [130] The vinegar enema induced extensive bowel infarction with destruction of the rectovaginal septum.

Syndromes Associated With Laxatives

Melanosis Coli Melanosis coli is a dark pigmentation of the colonic mucosa seen in 73% of patients who use anthraquinone laxatives (cascara sagrada, aloe, rhubarb, senna, and frangula), although the relationship is not exclusive. [131] Its gross appearance is a reticulated pattern of striations and spots that resembles alligator skin (Fig. 85-3; see Color Fig. 85-3). [132], [133] Melanosis can appear as rapidly as 4 months after the initiation of anthraquinone laxatives, with an average time to appearance of 9 months. Withdrawal of anthraquinones results in resolution of pigment changes in an average of 9 months. [132] Melanosis has also been described in patients with IBD. [134] The incidence of melanosis coli in endoscopy and autopsy series ranges from 0.25% to 23.6%, with an increased frequency in women and among those older than 40 years of age. [133] Melanosis coli is found mainly in the cecum and the rectum, although it may involve the entire colon, and may be detected in biopsy specimens even when not grossly visible. [114]

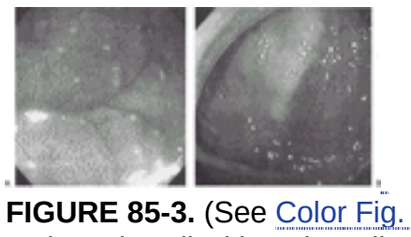

FIGURE 85-3. (See Color Fig. 85-3.) Endoscopic photographs of melanosis coli. **A:** The reticulated, alligator-skin appearance is characteristic of this condition. **B:** Melanosis coli with a sharp line of demarcation at the small bowel–colonic anastomosis.

Histologically, melanosis coli is associated with the deposition of a brown, granular pigment within lamina propria macrophages (Fig. 85-4). The number and size of mucosal macrophages are increased, and they are situated between the colonic crypts. Two distinct groups of macrophages are evident; those nearest the lumen have the least pigment, and those furthest from the lumen have the most pigment. [135] The colonic epithelial cells are normal by light microscopic examination. However, electron microscopy has revealed decreased numbers of microvilli, vacuolization of the apical cellular surface, lipid droplets near the basal membrane, and small vesicles near the lateral cell membrane. [136] The source of the pigment in melanosis coli is unclear. Its biochemical features of lipofuscin, ceroids, and melanin suggest a derivation from apoptotic epithelial cells. [137] Other suggested origins include degenerating mitochondria or lysosomes [135] and absorbed pigments derived from anthraquinone laxatives. [132] Guinea pigs fed danthron, an anthraquinone laxative, developed transient large-scale apoptosis of colonic surface epithelial cells. [138] Cellular debris phagocytosed by macrophages and carried into the lamina propria was transformed into the typical lipofuscin-like pigment seen in melanosis coli. These experimental results agree with dramatically increased apoptosis of colonic epithelial cells in patients with melanosis coli. [137], [138] and [139] The clinical significance of melanosis coli, apart from its association with anthracene laxative abuse, is uncertain; however, there is universal agreement that it is a benign condition. Colonic adenomas in patients with melanosis coli are not pigmented, and neoplastic tissues lack pigment-laden macrophages. [140] Biopsy of islands of unpigmented tissue in patients with background melanosis is therefore advised. Deposition of pigment proximal to carcinoma of the colon has led to speculation that

colonic stasis may be a contributing factor. [133] However, no correlation is found between the presence of melanosis and colorectal transit. [131] The risk of colonic neoplasia with melanosis coli is the subject of controversy. Siegers and colleagues [141] reported a relative risk of 3 for colorectal cancer with chronic anthranoid use, but Nusko and colleagues [142] found no increased risk of cancer with melanosis coli.

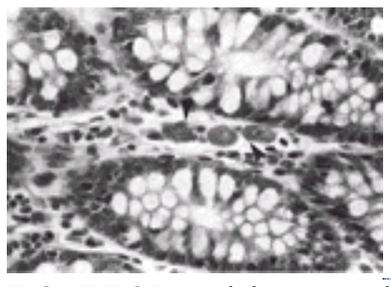

FIGURE 85-4. High-power photomicrograph demonstrates lipofuscin-like pigment within macrophages (*arrows*) in the lamina propria of a rectal biopsy specimen from a patient with melanosis coli.

Cathartic Colon The incidence of cathartic colon has dramatically decreased since its initial description in 1943. [143], [144] Most patients are women who have abused laxatives, either overtly or surreptitiously, for 15 or more years who develop bloating, abdominal fullness, and vague lower abdominal pain. [145] Symptoms of incomplete evacuation without laxatives lead to escalating doses of laxatives until these patients cannot defecate normally without cathartic use. [146] Ninety percent of surreptitious laxative abusers are women; many are emotionally disturbed. [146] These patients frequently undergo multiple diagnostic evaluations to evaluate complaints of chronic diarrhea, vague abdominal discomfort, thirst, and weakness. [147] Electrolyte and fluid abnormalities, particularly hypokalemia and hypovolemia, and steatorrhea have been reported. Heizer and colleagues [148] detailed two cases of protein-losing enteropathy with hypoalbuminemia which resolved after cessation of cathartic abuse. The most striking abnormalities in cathartic colon are seen on contrast radiographs (Fig. 85-5). Mild cases have findings limited to a foreshortened, conical cecum and loss of the typical beaklike appearance of the ileocecal valve. More severe cases are characterized by a dilated, tubular colon distal to the cecum, with loss of haustral markings; segments of bowel may appear narrowed, but these pseudostrictures are transient in nature. [145] The radiographic appearance of cathartic colon resembles that of long-standing ulcerative colitis but can be differentiated by the distensibility of the bowel in cathartic colon, the lack of mucosal inflammation and sparing of the rectosigmoid region. [143], [145] Complete resolution of these abnormalities within 4 months of cessation of laxative use has been reported. [149]

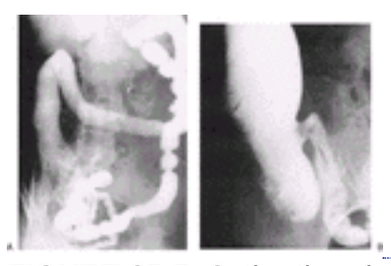

FIGURE 85-5. Cathartic colon. **A:** Barium enema study reveals that the right and transverse colon are devoid of haustrations. No ulcerations are seen. **B:** Detail of the cecum and proximal ascending colon reveals a gaping ileocecal valve. The lack of haustration in the right colon is striking. (From Campbell WL. Cathartic colon: reversibility of roentgen changes. Dis Colon Rectum 1983;26:445–448.)

Cathartic colon is related to chronic use of irritant laxatives (primarily anthraquinones); however, the precise mechanism is unknown. Anthraquinones are potent cellular toxins; for example, intravenous or oral administration of syrup of senna to mice causes damage to the myenteric plexus. [150] Resected human specimens show hypertrophy of the muscularis mucosae, thinning of the muscularis propria, excessive submucosal fat deposition, loss of myenteric plexus neurons, and replacement of ganglia by Schwann cells. [150] Reversible abnormalities in colonic nerve plexi exist in chronic laxative abusers who have not yet developed typical radiographic changes. [151] Toxic effects of anthraquinones on the colonic myenteric plexus nerves are the likely cause of the cathartic colon syndrome, although it is possible that a subgroup of patients with chronic idiopathic constipation has a developmental anomaly of the myenteric plexus which leads to chronic laxative ingestion. [152] Treatment of cathartic colon centers on withdrawal of irritant laxatives. Bulking agents, a high-fiber diet, and bowel retraining are the most effective measures. In the early phases of bowel retraining, use of an osmotic laxative or Fleet enemas may be helpful. Patient cooperation is critical; lack of compliance is a common problem. [146] In refractory cases, surgical approaches have been used, the most effective being total or subtotal colectomy, [153] but resection should be considered only after failure of maximal medical therapy in light of the reversibility of some cases. [130]

Drugs Resulting in Ischemic Colonic Injury

Several substances compromise mucosal blood flow through vasospasm, thrombosis, or both, resulting in ischemic injury (see Chapter 81). Although ischemic colitis is not uncommon in older adult patients with mesenteric atherosclerotic disease, its presence in a younger patient should prompt a search for a predisposing cause, such as medications. [153] The symptom complex manifested by these patients is fairly uniform, consisting of the abrupt onset of severe abdominal pain, bloody or nonbloody diarrhea, tenesmus, nausea, vomiting, and fever. Rebound tenderness and guarding, with hypoactive or absent bowel sounds, may be noted, although less striking physical findings may be found. Leukocytosis is variably present, and plain or contrasted abdominal radiographs may reveal “thumb printing.” Endoscopy may show mucosal friability, edema, erythema, granularity, ulceration with or without pseudomembranes, or necrosis. [155], [156] and [157] As in atherosclerotic mesenteric vascular disease, the junction of the distributions of the superior and inferior mesenteric vessels (from the distal end of the transverse colon to the proximal descending colon) is a common site of injury in drug-induced ischemic colitis.

Oral Contraceptives Transient ischemic colitis in otherwise healthy young women taking oral contraceptives was first described in 1968. [155] Mesenteric thrombosis, both arterial and venous, has been extensively described in association with oral contraceptives; arterial thrombosis carries twice the mortality of venous thrombosis but is half as common. [156] Although the typical presentation is that of classic ischemic colitis, symptoms can be indistinguishable from those of Crohn's disease. [157] Estrogens can cause a hypercoagulable state, vasospasm in the mesenteric vessels, and endothelial proliferation with subendothelial fibrosis, all of which may play a role in ischemic colitis. [156] High-dose estrogen preparations, high-dose progesterone combinations, depot progesterone dosages, and sequential estrogen/progesterone therapy have all been implicated in mesenteric thrombosis. [156], [157] The duration of oral contraceptive use in these patients ranges from 10 days to 11 years. The dose of the estrogenic fraction correlates with thrombogenicity. [119], [155], [156]

Vasopressin Vasopressin, a vasoconstrictor used to treat bleeding esophageal varices, decreases colonic blood flow in a dose-dependent fashion. A case of reversible ischemic colitis associated with intravenous vasopressin infusion has been reported. [158]

Ergotamine The association between ergot preparations and ischemic colitis is presumably related to vasospasm. Stillman and associates [159] described a 50-year-old woman with no risk factors for mesenteric ischemia who developed acute bloody diarrhea and acute colitis in the splenic flexure area while taking ergotamine tartrate; her symptoms resolved after discontinuation of ergotamine. Rectal ulceration, with obliteration of small blood vessels, endothelial proliferation, and vascular wall thickening, can develop in patients using ergotamine suppositories.

Alosetron Alosetron, which had been approved for treatment of diarrhea-predominant irritable bowel syndrome, was withdrawn from the market because of reports of ischemic colitis, but it has been reintroduced under controlled usage for severely symptomatic patients. [160]

Cocaine Several reports of severe, ischemic colitis in patients using high doses of cocaine have been published. [161], [162] Cocaine-induced ischemic colitis should be strongly suspected in any young or middle-aged person with abdominal pain and bloody diarrhea, particularly if there are no potential thromboembolic sources and if there is no history of estrogen use. [161] Inflammation can involve the rectum, which is an unusual location for ischemic colitis. The putative mechanism of cocaine-related ischemic colitis is catecholamine-induced mesenteric vasospasm by this powerful sympathomimetic agent. [161]

Dextroamphetamine A case of reversible ischemic colitis attributed to intense mesenteric vasoconstriction has been described in a 47-year-old man taking dextroamphetamine as his only medication. [154] His acute presentation of abdominal pain and bloody diarrhea resolved after cessation of dextroamphetamine therapy. Mesenteric angiography was normal.

Methamphetamine Methamphetamine, a drug frequently sold illegally as “ice” or “crystal meth,” has been implicated in several cases of ischemic colitis. [163]

Neuroleptics Ischemic colitis has been linked to the use of neuroleptics, especially tricyclic antidepressants, in some case reports. [164], [165]
Digitalis Preparations Digitalis preparations have been associated with ischemia in the small or large intestine; isolated colonic ischemia has not been reported. It is likely that these reports represent a manifestation of the clinical setting of digitalis use (i.e., systemic hypoperfusion), although digitalis preparations have been shown to cause mesenteric vasoconstriction in laboratory animals. [166]

Miscellaneous Drug-Induced Colitides

Nonsteroidal Antiinflammatory Drugs Several inflammatory syndromes have been associated with the use of NSAIDs. Colitis in previously asymptomatic individuals has been associated with numerous NSAIDs, including mefenamic and flufenamic acid, diclofenac, indomethacin, enteric-coated aspirin, ibuprofen, phenylbutazone, naproxen, and piroxicam. [34], [167], [168] and [169] It has been estimated that 10% of newly diagnosed colitis cases may be related to NSAID use. [170] Typical patients are older (average age, 63 years) with no gender predilection and present with diarrhea and either occult or gross lower gastrointestinal bleeding. [168] Some have weight loss or fever. The majority are anemic; many have an elevated erythrocyte sedimentation rate and leukocytosis. Endoscopic findings were variable and range from mild proctitis to ulcerative pancolitis. Ulcerations or other inflammatory changes can occur at any point in the colon, and can be focal or diffuse. Treatment is primarily withdrawal of the offending NSAID, although treatment with steroids, sulfasalazine, or even surgery for complications such as bleeding and perforation may be required. Medical treatment with steroids and sulfasalazine has been used on several occasions. In a 1992 review of NSAID colitis by Gibson and colleagues, [168] all 12 patients who were rechallenged with NSAIDs had relapses. Relapse of quiescent IBD was described in 19 patients; these patients were significantly younger than those with de novo NSAID-induced colitis, the median latency period between NSAID exposure and colitis symptoms was less than 1 week and relapse occurred in all eight patients rechallenged with NSAIDs. [168] A similar experience was described by Gleeson and colleagues [171] who reported an average age of 53 years. Most cases were self-limited, improving rapidly upon withdrawal of the NSAID and with 4- to 8-week courses of 5-ASA products; only one of the 23 patients in this study required surgery (for toxic megacolon). An increased rate of spontaneous colonic perforation or hemorrhage is associated with NSAID use. [34], [168] NSAIDs have been associated with perforated colonic diverticula, proctitis related to NSAID suppository use, eosinophilic colitis, and diaphragm disease of the colon. [34], [172], [173] NSAIDs have also been implicated in the development of collagenous colitis. [34], [173] The pathogenesis of NSAID-induced colonic injury is uncertain but probably is mediated by the effects of cyclooxygenase inhibition leading to loss of the mucosal cytoprotection and immunosuppression afforded by inducible prostaglandins. Studies using selective gene deletion and pharmacological inhibitors in rodents implicate both cyclooxygenase 1 and 2 in the pathogenesis of intestinal inflammation. [174], [175] Loss of vasodilating prostaglandins and production of vasoconstricting leukotrienes and cytotoxic oxyradicals associated with lipoxygenase activation could impair mesenteric blood flow. Epithelial injury and enhancement of colonic mucosal permeability by NSAIDs, with resultant increased susceptibility to luminal bacterial toxins, may also play a role. [176] One report of apparent NSAID-induced colitis occurring in a patient taking misoprostol raised some doubt about the role of prostaglandins in the pathogenesis of NSAID colitis; uncoupling of mitochondrial oxidative phosphorylation was offered as an alternative hypothesis. [177]
Gold Gold salt–induced colitis in patients with rheumatoid arthritis is probably caused by local toxicity of the drug. Up to 95% of orally administered gold is excreted in the feces, and stainable gold is present in mucosal biopsy specimens. [178] Most patients are women; clinical manifestations may include bloody diarrhea, abdominal pain, tenesmus, fever, leukocytosis, and even fulminant colitis. [179] Onset of colitis can be delayed for several weeks after cessation of gold therapy. [180] Diagnosis is usually made by the endoscopic appearance of ulceration and friability, often with hemorrhage; lesions are most often found in the rectosigmoid, although colitis may be diffuse. [178] Biopsies may reveal nonspecific acute and chronic inflammation, occasionally with eosinophils; some have histological findings that might cause confusion with Crohn's disease. [178], [181] Treatment is primarily withdrawal of gold therapy and supportive care with an average recovery time of 2 weeks, although surgery, steroids, British anti-Lewisite (BAL), and sulfasalazine have been used with success. [178] One case of eosinophilic gold-induced colitis responded to oral cromolyn therapy. [182] The older literature reports a 40% mortality with this complication; however, a review of 14 cases reported between 1980 and 1986 reveals no deaths. [178]
Isotretinoin Isotretinoin, a synthetic analog of vitamin A used in treatment of severe cystic acne, has been linked to acute colitis and to the reactivation of quiescent IBD. Martin and colleagues [183] reported that cessation of therapy resulted in prompt colitis resolution, whereas rechallenge caused an almost identical recurrence.
Antibiotics Hemorrhagic colitis associated with ampicillin, amoxicillin, and erythromycin differs from typical *C. difficile* colitis in that patients present with bloody diarrhea, generally do not have pseudomembranes, have predominantly right-sided colitis, rapidly improve with discontinuation of antibiotics, and test negative for *C. difficile* toxin. [184], [185] The pathogenesis is unknown, although this may represent a variant of *C. difficile* disease or an overgrowth of an unrecognized pathogen.
Chemotherapeutic Agents Rapidly dividing intestinal epithelial cells are particularly susceptible to the toxic effects of cancer chemotherapeutic agents with almost complete blockade of DNA synthesis by cytotoxic doses of antimetabolites. [186] Multiple chemotherapeutic agents cause colonic damage, including cytosine arabinoside, methotrexate, cyclophosphamide, and 5-fluorouracil. Clinical manifestations range from mild segmental colitis to fulminant ulcerative colitis with toxic megacolon. [186], [187], [188] and [189] The pathogenesis is probably multifactorial and includes direct epithelial injury by chemotherapeutic agents, invasion of the mucosa by intestinal pathogens, and ischemic injury. [186], [187] and [188] Cytosine arabinoside induces a three-stage pattern of damage consisting of initial injury—replacement of normal mucosal cells with atypical undifferentiated cells, with conspicuous absence of mitotic figures; progressive injury—cellular necrosis and glandular dilation, with persistent lack of mitosis; and regeneration—resumption of normal mitotic activity, with mature goblet cells in the bases of crypts. [187] These changes can probably be extrapolated to most chemotherapeutic agents. [188]
Methyldopa Colitis associated with methyldopa therapy presents as bloody or nonbloody diarrhea with prompt resolution of symptoms after drug withdrawal. Three patients who were rechallenged with methyldopa demonstrated symptom recurrence. [190]
Flucytosine Flucytosine, an antifungal agent, been associated with the development of colonic inflammation resembling idiopathic ulcerative colitis. [191]
Sodium Phosphate Bowel Preparation Solution Focal colitis caused by oral sodium phosphate bowel preparation solution has been described. In 687 consecutive patients who underwent sodium phosphate bowel preparation, unexplained aphthous ulceration was present in 18 patients (2.6%), with resolution within 1 to 8 weeks when reexamined. [192]

COLONIC ULCERS

A number of conditions other than medication use are associated with colonic ulceration (Table 85-5). Common presenting symptoms include hematochezia, abdominal pain, and a mucopurulent rectal discharge. Diarrhea correlates with the surface area affected and is uncommon with isolated ulceration. Careful endoscopic and histological examinations can help distinguish these conditions using the criteria established in Table 85-5.

TABLE 85-5 Clinical Characteristics of Colonic Ulcers

Isolated Nonspecific Colonic Ulcer

In a review of 127 patients with colonic ulcers, the average age was 45 years, with a range of 8 to 84 years and a 55% female predominance. [193] Sixty-seven percent of patients had cecal ulcers, another 18% had ascending or transverse colonic lesions, and 15% had descending or sigmoid ulcers. Virtually all were solitary ulcers located on the antimesenteric side of the lumen, ranging from 0.5 to 6.5 cm in diameter. [193], [194] Cecal ulcerations are most often found near the ileocecal valve, and are round or oval in configuration, with sharply demarcated margins and relatively normal surrounding mucosa. [193], [195], [196] Histology is nonspecific, with acute and chronic inflammation. [193], [196]

Pathogenesis The cause of nonspecific colonic ulceration remains unknown; therefore the diagnosis is one of exclusion. Several hypotheses for the origin of nonspecific colonic ulceration include ischemia, cecal diverticulosis, acid, and miscellaneous causes. Microvascular thrombosis and vascular abnormalities, including thickening of vascular walls, thrombosis, thrombus organization, and recanalization within local vessels, in areas underlying and adjacent to nonspecific colon ulcers suggest a localized form of ischemic colitis, perhaps promoted by fecal stasis. [194], [195], [196] and [197] However, ischemic injury usually occurs in the watershed area of the descending colon and splenic flexure, not the cecum, and right-sided fecal impaction is relatively rare. [198] Finally, thrombosis may be secondary to the adjacent inflammation, making determination of cause and effect difficult. Based on the observation that 50% of cecal ulcerations had histological evidence of cecal diverticula, it has been reported that cecal diverticulitis could result in ulcer formation. [184] However, less than 15% of colonic ulcers occur in the sigmoid colon, the site of highest

incidence of diverticulosis, and cecal nonspecific ulcers are almost invariably located on the antimesenteric border of the lumen, in contrast to the mesenteric location of cecal diverticula. [194], [198] It has been postulated that because nonspecific ulcers frequently occur in the cecum just distal to the ileocecal valve (where colonic contents are usually neutral to slightly acidic in pH), an altered acid-base status of this region could lead to cecal ulceration. Correlations have been noted between the use of corticosteroids, NSAIDs, oxyphenbutazone, and oral contraceptives and the occurrence of colonic ulcers. [193], [195], [198] However, causation has not been established for any of these substances, and they cannot be implicated in a substantial majority of colonic ulcers. [199] Other potential causative explanations include digestive enzymes, foreign body trauma, neurological stress, lipomas, bacterial or viral infections, and unknown toxins. [194], [200] The cause of nonspecific colon ulceration at this point remains unclear and is almost certainly multifactorial.

Clinical Features Most of the historical and physical examination findings are nonspecific. In a 1982 review by Ona and colleagues, [193] the signs or symptoms most frequently cited were acute or chronic right lower quadrant abdominal pain (50% of cases) with symptoms similar to those of appendicitis; lower gastrointestinal bleeding (33% of cases) most commonly presenting as hematochezia; perforation, with acute surgical abdomen (19%); and abdominal mass (16%), which is most common in left-sided or transverse colonic ulcers. Other reported findings include obstruction, fever, and leukocytosis. Hemoccult-positive stool is occasionally seen in the absence of any other demonstrable source of gastrointestinal blood loss.

Diagnosis Colonoscopy is the diagnostic test of choice and has shifted management of this condition from the surgical to the medical realm. The findings on air-contrast barium enema are abnormal in 69% to 75% of cases but are nonspecific, consisting of luminal narrowing, filling defects, mucosal irregularities, localized colonic spasm, or mass effects. [193], [200] Sigmoidoscopy misses most colonic ulcers by virtue of their predilection for cecal location. [193] In the setting of brisk bleeding from a colonic ulcer, mesenteric angiography is sensitive (89%) but nonspecific. [193] A CT scan is useful primarily in cases with associated perforation or abscess.

Differential Diagnosis Because colonic ulcers are histologically nonspecific, diagnosis is based on exclusion of other potential entities. Solitary rectal ulcers occur distally and have a characteristic histological appearance (discussed later in the chapter). Ulcers associated with Crohn's disease are usually multiple, may be longitudinal, and are often associated with inflammation elsewhere in the gastrointestinal tract. [195] Infectious causes of colonic ulceration include tuberculosis, *Entamoeba histolytica* cytomegalovirus (especially in immunocompromised patients), and *Salmonella typhi* and are usually multiple in number. Diagnosis can often be made by biopsy. [176] Tuberculosis of the colon usually results in multiple transverse cecal ulcers with associated nodularity and deformity of the ileocecal valve and biopsies diagnostic of tuberculosis, either by culture or histology. [201] Ischemic ulcers occur in the splenic flexure and the rectosigmoid junction of older patients; frequently, these are multiple lesions with a serpiginous or linear configuration, irregular margins, and abnormal surrounding mucosa. [195] Biopsy is nonspecific. Stercoral ulcers usually occur in the sigmoid area in association with fecal impactions (discussed later in the chapter). Malignancy (including carcinoma and lymphoma) must be ruled out histologically, because gross appearance is not diagnostic. [195] Other miscellaneous causes of colonic ulceration which have been reported include systemic lupus erythematosus, Behçet disease, Wegener granulomatosis, essential mixed cryoglobulinemia, and Churg-Strauss syndrome. [193], [202], [203] and [204] These can usually be differentiated by the setting of their underlying disease.

Prognosis and Therapy The earliest reports of nonspecific colonic ulceration were of ulcers discovered during surgery for complications (perforation, abscess, or bleeding) or during postmortem examination. As recently as 1974, surgery was the recommended treatment for all nonspecific colon ulcers. However, in 1980, Blundell and Earnest [198] described four patients who had nonspecific ulcers discovered at colonoscopy. Three of these patients were managed conservatively, with satisfactory clinical outcomes. Currently, it is recommended that uncomplicated colonic ulcers be followed endoscopically, with multiple biopsies taken during each endoscopy to document healing and to rule out malignancy and infection. The optimal interval between colonoscopies is uncertain; 4 to 6 weeks seems a reasonable period based on healing rates. [193], [194], [198] Surgery including oversewing of the ulcer, local ulcer excision, and right hemicolectomy is recommended for ulcers complicated by perforation, substantial bleeding, or associated intra-abdominal abscess, or for those that fail to heal with observation. [193], [196]

Dieulafoy-type Colonic Ulceration

In 1898, Georges Dieulafoy described three cases of previously asymptomatic patients exsanguinating from minute gastric ulcerations less than 2 mm in diameter, with rupture of a relatively large submucosal artery into the gut lumen. Several reports have been published detailing bleeding from similar colonic lesions found in the rectum, cecum, anal verge, and elsewhere in the colon. [205], [206], [207], [208] and [209] The colonic Dieulafoy ulcer is a rare lesion; Dy and colleagues, [206] in the largest published series, reported an incidence of five patients with Dieulafoy colonic ulceration among 3059 patients presenting with gastrointestinal bleeding. Gender distribution is roughly equal among males and females, with ages ranging from 20 to 94 years. Histologically identical to Dieulafoy-type lesions elsewhere in the gastrointestinal tract, colonic lesions are typically solitary mucosal defects, extending no deeper than the upper submucosal layer, with erosion of an underlying submucosal large-caliber artery (Fig. 85-6). The artery is tortuous and curved toward the mucosa, with erosion at the apex of its curve. There is no associated arterial aneurysm, vasculitis, or other vascular anomaly with little or no inflammation in the ulcerated area. [205] The cause is unknown, although some have postulated that the pulsations of a large submucosal artery may cause mechanical damage to the overlying mucosa, allowing an inflammatory reaction to the luminal contents to further erode the exposed arterial wall. [205] Clinically, patients present with the acute onset of massive bleeding; they are otherwise asymptomatic. The mucosal lesions are notoriously difficult to visualize on endoscopy; selective mesenteric angiography has been useful in localization. Systemic vasopressin infusion has not been successful in managing bleeding from these lesions. The definitive therapy has traditionally been segmental surgical resection. [205] Endoscopic therapy of Dieulafoy lesions has been increasingly used, and now may be considered the intervention of choice. [206], [210] Endoscopic methods that have been successfully employed have included vascular clipping, band ligation, epinephrine injection, absolute ethanol injection, sclerotherapy followed by electrocoagulation, and injection followed by heater probe thermocoagulation. [210], [211] and [212]

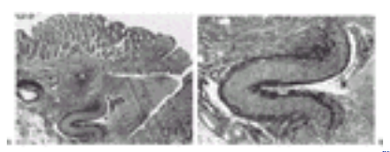

FIGURE 85-6. Dieulafoy ulcer of the colon. **A:** Low magnification shows a thick-walled, tortuous muscular artery just below the muscularis mucosa, with fresh ulceration overlying the artery at the site of bleeding. **B:** Higher magnification reveals details of the site of bleeding, at the apex of the artery's curve toward the mucosa. (From Barbier P, Luder P, Triller J, et al. Colonic hemorrhage from a solitary minute ulcer. Gastroenterology 1985;88:1065.)

Stercoral Ulcer (Huntley Syndrome)

Colonic ulceration caused by ischemic pressure necrosis from a stercoraceous mass may result in colonic perforation, with resultant peritonitis. [213] In Serpell and Nicholls' review [214] of stercoral perforation, the median age of patients was 60 years, with a range of 16 to 89 years. There was a slight female predominance. Risk factors included chronic constipation (61%) and conditions associated with constipation, such as confinement to a nursing home or other chronic care facility (23%), renal failure, or transplantation (13%); use of constipating medications (aluminum hydroxide-containing antacids, narcotics, phenothiazines); hypothyroidism; and colonic strictures. Foreign bodies have also been implicated. [213], [214] The actual incidence is unknown. In an early review of 175 unselected autopsies, 4.6% were found to have stercoral ulcerations; half of these had perforations. [215] However, later autopsy studies revealed a rate between 0.04% and 0.3%. [216], [217] A later review found only 64 reported cases of colonic perforation as a consequence of stercoral ulcer. [214]

Patients who have perforated ulcers usually present with complaints of abdominal pain and signs of peritoneal irritation (80%–100%). [214] An abdominal mass (23%) or a fecal mass on rectal examination (6%) may be palpable. Plain radiographic findings include pneumoperitoneum (53%), marked fecal loading (30%), and calcified fecaliths within the colon (21%). Contrast radiography is rarely helpful. [214] The correct preoperative diagnosis is rarely made, although survival is improved if the diagnosis is established. [218] Nonperforating stercoral ulcers are rarely diagnosed but must be considered in a constipated patient presenting with abdominal pain, rectal bleeding, and leukocytosis.

Perforating stercoral ulcers are usually located on the antimesenteric border of the sigmoid or rectosigmoid colon (77%), with 47% of these within the sigmoid alone. [214] Other reported sites include the cecum (9%), the descending colon (5%), and the splenic flexure (2%). Resected specimens reveal sharply demarcated, irregular ulcers that conform to the contours of the fecal masses present. Histologically, the colonic mucosa is necrotic and denuded, with acute and chronic inflammatory changes. [215]

The pathophysiology of stercoral ulceration is related directly to the presence of a hard mass of feces (scybalum), which may be present for months to years. [214] The

pressure of the scybalum on the colonic wall exceeds the capillary perfusion pressure, which causes ischemic necrosis, with resultant ulceration and eventual perforation. [214], [218] The sigmoid predominance is probably caused by progressive desiccation of the fecal mass in the distal colon, the narrower luminal diameter of the sigmoid, and the relatively poor colonic blood supply in this area. [214]

The differential diagnosis of stercoral ulceration is limited and includes idiopathic (spontaneous) colonic perforation and perforation from trauma, malignancy, infection, or other cause. Spontaneous colonic perforation, in particular, may be confused with stercoral perforation. It often occurs during a difficult bowel movement related to an acute tear in the mucosa with no inflammation (as opposed to the round or ovoid perforations seen in the bases of stercoral ulcerations).

Surgical resection or proximal diversion is required for definitive therapy of both perforated and nonperforated stercoral ulcers. Intravenous antibiotics are indicated as adjunctive therapy; the majority of deaths occur as the result of uncontrolled sepsis. [214] Resection of the perforation with end colostomy and Hartmann pouch or mucous fistula has been associated with a lower mortality (23%) than other procedures (53%) and has been recommended as the surgical procedure of choice. [218] Lavage of the peritoneal cavity is required to reduce fecal contamination and intraoperative colonic lavage to remove scybala proximal to the site of the perforation may prevent recurrence of ulceration. [219]

Solitary Rectal Ulcer Syndrome (SRUS)

Although first described by Cruveilheir in 1829, the classic 1969 review by Madigan and Morson [220] established the diagnostic criteria for what is now known as the SRUS. This syndrome, with more than 400 cases reported in the literature, primarily affects young adults; the average age is in the third and fourth decades, with a range from 10 to more than 80 years. There is a slight female predominance. [221]

Patients with SRUS typically present with symptoms related to bowel disturbance; 70% to 90% present with constipation, tenesmus, incomplete evacuation, straining at stool, or lower abdominal pain with rectal bleeding in 56% to 89%. [221] Passage of mucus per rectum, overt rectal prolapse, and fecal incontinence are less frequent. One large review found that 26% of patients with endoscopic discovery of a solitary rectal ulcer were asymptomatic. [222] Systemic symptoms are uncommon, and bleeding is rarely significant enough to warrant transfusion.

Gross endoscopic appearance of SRUS is variable; the lesion may appear as a single, well-demarcated ulcer up to 5 cm in size, as multiple ulcers, as a polypoid lesion, or as a flat region of erythematous mucosa. Typically, the lesion appears on the anterior rectal wall (68%–95% of cases) approximately 6 to 10 cm from the anal verge. [221], [222] Specific histological criteria are used to establish the diagnosis, as defined by Madigan and Morson. [220] The lamina propria is replaced by fibroblasts, smooth muscle, and collagen, with associated hypertrophy and disorganization of the muscularis mucosa termed *fibromuscular obliteration of the lamina propria* (Fig. 85-7). Displacement of the mucosal glands into the submucosa and erosion of the mucosal surface may also be seen; the occasional presence of submucosal cystic glands has led to speculation that SRUS and colitis cystica profunda are identical or closely related syndromes with a common cause. [222] Endoscopic ultrasonographic findings in colitis cystica profunda and SRUS are nonspecific; it has been suggested that endoscopic ultrasonography be used to rule out malignancy and to assess the extent and severity of disease. [223] Levine and colleagues [224] noted that diffuse collagen infiltration of the lamina propria is seen almost exclusively in SRUS and can be used to differentiate this syndrome from Crohn's disease, ulcerative colitis, and chronic ischemic colitis. Malignancy, amebiasis, lymphogranuloma venereum, secondary syphilis, and endometriosis [225] must also be considered in the differential diagnosis.

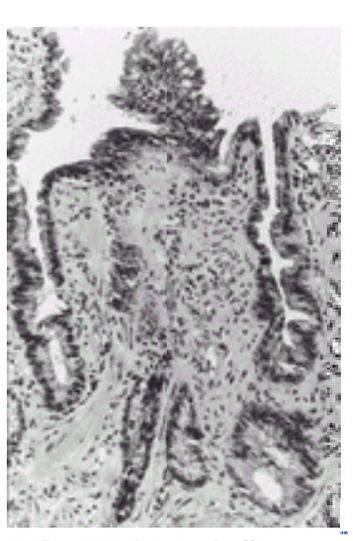

FIGURE 85-7. Solitary rectal ulcer syndrome is characterized by fibromuscular obliteration of the lamina propria with crypt distortion. (Hematoxylin and eosin stain; original magnification ×200. Courtesy of John Woosley, M.D., Chapel Hill, NC.)

The pathophysiology of SRUS is uncertain. Self-digitation (seen in up to 50% of patients with SRUS) has been suggested as a cause, as have a congenital anomaly of the anterior rectal wall, a localized form of IBD, and infectious agents, although convincing support of these hypotheses is lacking. Prolapse-induced rectal mucosal trauma or ischemia has been strongly implicated as a possible cause of SRUS. [223] The incidence of mucosal or full-thickness rectal prolapse ranges from 13% to 100% in published series of patients with SRUS. [221] A combination of rectal prolapse and a high fecal voiding pressure (caused by overactivity of the external anal sphincter or failure of puborectalis relaxation during defecation) has been theorized to cause local ischemic injury and trauma to the rectal mucosa, resulting in ulceration. [226], [227] However, other studies have failed to find electromyographic evidence of a hyperactive anal sphincter in a majority of patients. [221]

Treatment of SRUS depends on the gross morphology of the lesion and on the presence or absence of rectal prolapse. Patients with polypoid lesions usually are younger and are more likely to be asymptomatic, tend to have a better prognosis, and respond to conservative medical therapy. [221] Such therapy includes bowel retraining, the use of bulk laxatives, and reassurance. Antiinflammatory drugs (i.e., corticosteroids, salicylates, sucralfate enemas) have been used with varying degrees of success, but controlled trials have been performed. Surgery is advised for patients with rectal prolapse because of their poor response to medical therapy. A 65% overall response rate to surgical intervention was noted in Tjandra's review, [221] with best results in patients with polypoid lesions. A more recent review of 81 patients with SRUS treated surgically revealed a 55% to 60% long-term response to surgical intervention with a minimum follow-up interval of 12 months (median 90 months). [228] The procedure of choice in patients with rectal prolapse is abdominal rectopexy. [229] Anteroposterior rectopexy, local excision, colonic resection, and, rarely, diverting colostomies have been successfully employed in nonprolapsing patients with intractable symptoms refractory to medical therapy. [221], [229] Prolonged evacuation time is predictive of poor prognosis. [214] Although not typically associated with carcinoma, there have been cases of unexpected colorectal malignancy found in resected solitary rectal ulcers. [230]

TYPHLITIS

Typhlitis (from the Greek “typhlos,” meaning blind sac), an acute necrotizing colitis principally involving the cecum, was originally described in 1970 by Wagner and colleagues in terminally ill leukemic children with severe neutropenia. [231] Synonymous terms include neutropenic enterocolitis, necrotizing enterocolitis, and ileocecal syndrome because inflammation may involve the terminal ileum, ascending colon, and appen- dix. [232], [233] and [234] Typhlitis occurs primarily in severely immunosuppressed patients with leukemia, but has been described with combination immunosuppression for other malignancies or renal transplant and autologous bone marrow transplant, autologous blood stem cell transplantation, drug-induced granulocytopenia, aplastic anemia, cyclic neutropenia, and the acquired immunodeficiency syndrome (AIDS). [234], [235], [236] and [237] The rate of typhlitis in autopsy series of children with acute leukemia ranges from 10% to 24%. [232], [234] There is a suggestion of increasing frequency, attributed to more aggressive chemotherapeutic regimens for both hematologic and solid tumors. [238], [239] No race or gender predilection or specific regimen of chemotherapy is apparent.

The clinical spectrum of disease varies from mild, self-limited cecal inflammation to fulminant necrosis and perforation. Symptoms and signs usually develop at the

nadir of neutropenia 1 to 2 weeks after completion of induction chemotherapy and is less common during consolidation. Virtually all patients are neutropenic and febrile. Most patients have abdominal pain (localized to the right lower quadrant in approximately 40% to 60% of cases) and many have nausea, vomiting, abdominal distention, and diarrhea (20%–45% bloody), with associated stomatitis and necrotizing pharyngitis, indicative of diffuse mucositis. Peritoneal signs or shock suggest the possibility of perforation or sepsis. Positive blood cultures usually grow enteric bacteria, especially *Pseudomonas* or *Candida* species organisms.

Typhlitis is difficult to diagnose because of nonspecific symptoms, signs, and radiographic findings. [232] In the appropriate clinical setting, helpful radiographic patterns include abdominal plain films showing a fluid-filled, distended cecum with dilated small bowel loops, diminished or absent colonic gas, “thumb printing” of the ascending colon, evidence of a right lower quadrant soft tissue mass with small bowel displacement, or cecal pneumatosis. CT scan may reveal symmetrical cecal wall thickening (average about 10 mm) with pericecal fluid or a soft tissue mass if an abscess is present (Fig. 85-8). Ultrasound examination of the right lower quadrant may show the target or halo sign of a solid mass with echogenic center (collapsed mucosa and intestinal contents) and hypoechoic periphery (thickened bowel wall). [238] This finding is nonspecific and also has been seen with intestinal hemorrhage, ischemic colitis, intussusception, and lymphoma. Barium enema may be useful in diagnosing typhlitis but is hazardous due to a risk of perforating the necrotic bowel. [239] Instillation of air by endoscopy may be similarly hazardous; however, a gentle, flexible sigmoidoscopy may be useful to exclude IBD or pseudomembranous or ischemic colitis.

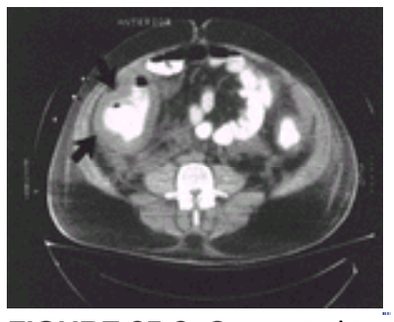

FIGURE 85-8. Computed tomographic scan reveals a massively dilated cecum with thickened walls, spiculation of pericolic fat, and inflammatory changes in the soft tissues of the right flank of a patient with typhlitis. (Courtesy of David Warshauer, M.D., Chapel Hill, NC.)

Typhlitis must be considered in any neutropenic patient with fever, abdominal pain, and diarrhea. The differential diagnosis includes pseudomembranous colitis secondary to *C. difficile* drug-induced or infectious colitis, appendicitis, diverticulitis, ischemic colitis, Crohn's disease, intussusception, intramural hemorrhage, or leukemic infiltrate.

Inflammation almost always involves the cecum but may additionally affect the terminal ileum or ascending colon, or the patient may have sporadic ulcers throughout the large and small intestine. The involved segments are dilated, edematous, and often hemorrhagic on external appearance. The cecal wall is thickened and hemorrhagic; necrotic material covers the ulcerated mucosa. Superficial ulcers can coalesce to form larger lesions. The serosal surface usually remains intact despite severe mucosal damage. Microscopically, there is marked submucosal edema with hemorrhagic necrosis of the mucosa but without a significant inflammatory reaction or leukemic infiltrate. Fungal organisms may be seen, and bacteria (usually gram-negative rods) may surround and infiltrate small blood vessels. [232], [233], [240]

The pathogenesis is unknown, but proposed causes include leukemic infiltration of the bowel, intramural hemorrhage and necrosis, local ischemia, toxic effects of chemotherapy, gastrointestinal stasis with bowel erosion, bacterial translocation, or a combination of these factors. The following sequence of events may occur: failure to maintain an intact epithelial barrier of the right colon (from the listed factors); bacterial invasion of bowel wall; intramural proliferation of bacteria secondary to decreased phagocytosis and killing (neutropenia); production of bacterial endotoxins with subsequent necrosis, hemorrhage, and perforation; and sepsis. In the spectrum of mucosal barrier injury, typhlitis is the most severe gastrointestinal manifestation. [241] It is unclear why the cecum is always affected. [232], [233]

Survival in this illness depends on prompt recognition of the condition, appropriate management, and rapid return of normal levels of neutrophils. Management is primarily medical, with selective surgical intervention. Medical therapy includes nasogastric suction for bowel rest, intravenous hydration, broad-spectrum antimicrobial therapy with *Pseudomonas* coverage, and avoidance of antimotility agents. If fever persists for more than 72 hours after antibiotics are begun, antifungal therapy with amphotericin B should be considered and a CT scan performed to identify an abscess. Granulocyte transfusions have been used with some success, but their benefit has not been proven. Surgery has been effective in patients failing medical therapy, and its use should depend on prognosis of underlying primary disease. Indications for surgery include evidence of free intraperitoneal perforation, persistent or life-threatening gastrointestinal hemorrhage, clinical deterioration suggesting uncontrolled sepsis, and abscess. The preferred procedure is a right hemicolectomy with ileostomy and mucous fistula formation. Total excision of the necrotic focus is imperative because incomplete removal uniformly results in death. [242], [243] Typhlitis has a high mortality rate (averaging 40%–50%) whether treated medically or surgically. Patients who develop typhlitis during induction and are medically treated are likely to relapse during subsequent chemotherapy (up to 67%). [244] Allowance of complete healing is advocated before resumption of chemotherapy to minimize risk of relapse.

COLITIS CYSTICA PROFUNDA

Colitis cystica profunda is a rare, benign disease most often involving the rectum. It is characterized by the presence of submucosal, mucus-filled cysts. This lesion was first reported by Stark in 1766 and was termed colitis cystica polyposa by Virchow in 1863. In 1957, the term colitis cystica profunda was used to differentiate this condition which is located below the muscularis mucosae from colitis cystica superficialis, in which multiple mucus-filled cysts are limited to the mucosa in association with pellagra and celiac sprue. [245], [246]

Colitis cystica profunda can be solitary or multiple in a localized, segmental, or diffuse distribution. In a review of 144 cases, 85% were localized lesions, typically involving the rectum; there was a slight female preponderance and a median age of 30 years. [247] Symptoms most often include blood or mucus discharge from the rectum with variable diarrhea; abdominal, rectal, or sacral pain; and tenesmus. Bulky or fibrotic lesions may rarely produce either partial or complete obstruction. [248] The localized form is often found on digital rectal examination, which reveals one or more smooth, firm masses not adherent to the underlying muscle layer, a thickened rectal wall, or stenosis.

Lesions are usually found within 12 cm of the anal verge and are commonly located on the anterior rectal wall. Endoscopic findings range from normal overlying mucosa to mucosal edema, erythema, friability, or even ulceration overlying a polypoid submucosal mass with or without umbilication. Obvious cysts may be present and when multiple, can form a mass up to 3 cm in size. Rectal stricture or prolapse may be seen. Barium enema may demonstrate no abnormalities or may show thickened valves of Houston, increased presacral space, various mucosal abnormalities, filling defects, circumferential stricture, or visible prolapse. [249] Transrectal ultrasound may be helpful by localizing cysts in rectal submucosa and confirming absence of lymph node involvement or invasion of the muscular layer. [223], [250] The differential diagnosis of these symptoms and findings is extensive and includes broad categories of inflammatory, infectious, and neoplastic diseases. [245] Definitive diagnosis of colitis cystica profunda is based on typical histological findings and absence of malignant features. Microscopically, the submucosal layer is enlarged by cysts which occasionally involve the muscularis propria and serosa (Fig. 85-9). Submucosal cysts are found in a variety of conditions [249] and must be differentiated from a well-differentiated mucinous adenocarcinoma. The cysts may be bordered by a well-differentiated colonic epithelial lining ranging from tall columnar to squamous cells, or they may have no lining at all. The lamina propria stroma may be replaced with collagen and misoriented smooth muscle cells. Adjacent mucosal edema, superficial ulceration, acute and chronic inflammation, pseudomembranes, and distorted crypt architecture may be present.

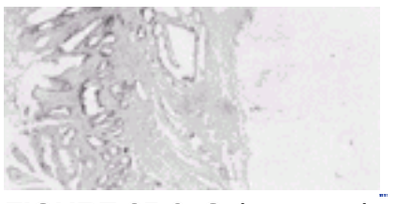

FIGURE 85-9. Submucosal mucin-filled cysts are characteristic of colitis cystica profunda.

The pathogenesis is not known; however, three mechanisms have been proposed. [244], [249] A congenital origin and herniation of epithelium owing to a weakened muscularis mucosa are two unsubstantiated theories. An acquired origin is supported by the absence of colitis cystica profunda in large pediatric autopsy series, an association with conditions causing trauma of the bowel wall, regression after diverting colostomy, and experimental studies implicating an inflammatory cause. Colitis cystica profunda may be one manifestation of a spectrum of disease states that includes SRUS and internal rectal prolapse. Rectal prolapse occurs in up to 54% of patients with colitis cystica profunda, and the anterior location and lamina propria fibrosis resemble SRUS. [220], [221], [249] Localized colitis cystica profunda is probably caused by ischemia, secondary to trauma induced by rectal prolapse. Defecography showing prolapse and abnormal pelvic electromyography are frequently seen, [247], [251] but these findings are not always present, suggesting that inflammation and mucosal trauma also contribute to the pathogenesis of this lesion.

The diagnosis of colitis cystica profunda must be considered to avoid inappropriate or radical surgery for suspected cancer. Because shallow mucosal biopsies are usually inadequate, definitive diagnosis may require a surgical excision. The usual course of this disease is a chronic, stable appearance of the rectal mucosa; however, cysts, ulcers, and symptoms may come and go. The lesions are not premalignant, however, adenocarcinoma may be located adjacent to colitis cystica profunda. [252] The severity and duration of symptoms should guide choice of treatment options. Most patients can tolerate symptoms with treatment consisting of reassurance, dietary fiber to soften bowel movements, and education to avoid straining during defecation and digital manipulation of the rectum. Only a small preliminary experience has reported the benefits of rectal prolapse repair. Local surgical procedures to excise cysts and ulcers, such as transanal excision, posterior proctectomy, electrocautery, or injection sclerosis, are usually adequate, although lesions may recur. [245], [248], [253], [254] Resectional surgery is usually unnecessary. Diverting colostomy, performed in the most disabled patients, alleviates the symptoms. Complications such as hemorrhage, obstruction, severe pain, or rectal stenosis need surgical intervention.

PNEUMATOSIS CYSTOIDES INTESTINALIS

This uncommon condition is characterized by multiple, thin-walled, noncommunicating, gas-filled cysts with no epithelial lining in the wall of the small or large intestine. Less frequently, other areas can be involved, including the stomach, duodenum, and extraluminal structures (e.g., mesentery, lymph nodes, omentum, gastrohepatic/falciform ligaments, peritoneum). [255] Usually pneumatosis cystoides intestinalis is discovered unexpectedly in an asymptomatic or minimally symptomatic patient and follows a benign course. Occasionally, it is associated with more fulminant illness such as bowel infarction, pseudomembranous enterocolitis, or necrotizing enterocolitis. After serious illness is excluded, pneumatosis intestinalis can be classified as either primary (idiopathic) or secondary, the latter being associated with a variety of conditions. Pneumatosis is most commonly associated with chronic obstructive pulmonary disease, intestinal obstruction, collagen vascular diseases (scleroderma; dermatomyositis, mixed connective tissue disease), systemic amyloidosis, and iatrogenic conditions (after surgery or endoscopy). Pneumatosis has occurred in patients with late-stage AIDS and characteristically involves the cecum and right colon. There is an association with cryptosporidial diarrhea, [256], [257] *C. difficile* colitis, [258] and Crohn's disease, especially with steroid use. [259] In Koss's review in 1952, [255] only 15% of cases were classified as idiopathic. He also reported an increased male prevalence of 3.5:1, but more recent reports suggest an equal male-to-female ratio. Pneumatosis intestinalis may be seen at any age, but in adults it usually occurs between 30 and 50 years of age. [260]

If symptoms are present, the most common complaints are diarrhea, vague abdominal discomfort, and abdominal distention with occasional hematochezia, mucus from the rectum, and weight loss. Typically, the condition is discovered incidentally during radiographic or endoscopic evaluation or at laparotomy. The abdomen may be tender, and occasionally an abdominal mass is palpable; however, fever and peritoneal signs are unusual. [261] Secondary cases of pneumatosis intestinalis typically involve the small bowel and ascending colon, whereas idiopathic cases tend to involve the left colon. The rectum is infrequently involved. [260], [262] The cysts range in size from a few millimeters to several centimeters and are typically found in clusters. A segmental distribution with skip areas is common, and from a few centimeters to a meter of bowel can be involved. An association with sigmoid colon redundancy has been suggested. [262] Colonic transit has been shown to be normal or even slow. Complications occur in about 3% of cases and include volvulus, pneumoperitoneum, intestinal obstruction, intussusception, tension pneumoperitoneum, hemorrhage, intestinal perforation, and recurrent asymptomatic pneumoperitoneum. [260]

Diagnosis is most commonly made on plain radiograph of the abdomen, on which linear, curvilinear, or cystic lucencies are seen in the bowel wall (Fig. 85-10). Additionally, pneumoperitoneum or retroperitoneal air may be found. Barium studies may help confirm location within the bowel wall. However, abdominal CT scan is more sensitive than conventional radiography in detecting intramural gas, which parallels the bowel wall. [262] CT scan can also detect portal or mesenteric venous gas, which, when associated with pneumatosis, is highly suggestive of bowel infarction. Endoscopically, pneumatosis appears as multiple, pale to bluish, rounded, soft polyploid masses protruding into the lumen. Because the cysts contain air under pressure, they deflate with a pop or hiss sound if punctured. [261] Submucosal pneumatosis of the colon can mimic polyposis, pseudopolyposis, or intramural hematomas on barium study; endoscopy can help differentiate causes. Enterogenous intestinal cysts are rare and occur in children and young adults. They are usually single, intramural, and most often involve the terminal ileum; their normal intestinal mucosal lining helps to differentiate them from pneumatosis. Lymphangiomas of the peritoneum differ in that lymph and not gas is present within cysts and no giant cells line these areas. Diffuse emphysema caused by infection with gas-producing organisms is distributed in all tissue spaces. In fat necrosis and sclerosing lipogranulomatosis, a fat stain is positive.

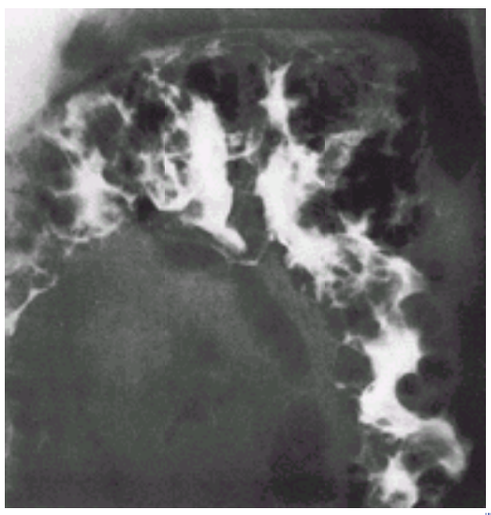

FIGURE 85-10. Pneumatosis cystoides intestinalis in an asymptomatic person. The intramural collections of gas simulate multiple colonic filling defects. Note that the filling defects in this condition appear to be more radiolucent than the soft-tissue masses in the intestinal polyposis syndromes.

The cysts in pneumatosis cystoides intestinalis have a submucosal or subserosal location and contain a high concentration of hydrogen (up to 50%). [263] The serosal cysts usually occur near the mesenteric border. [260] They may be partially lined by endothelial cells and are surrounded by foreign body multinucleated giant cells. Inflammation with neutrophils, eosinophils, plasma cells, lymphocytes, and epithelioid granulomas can be observed. Koss [255] postulated that fibrosis progresses until cysts disappear. The cause of pneumatosis is unknown, but several theories exist. The mechanical theory suggests that gas is forced into the bowel by one of several routes: pulmonary, trauma, mucosal breaks, anastomoses, obstruction, increased pressure, or increased peristalsis. The bacterial theory proposes that gas-producing bacteria invade the bowel wall. Finally, the biochemical theory postulates that excessive hydrogen produced by bacterial fermentation of carbohydrate in the intestinal lumen is absorbed and trapped within the intestinal wall by a process called counterperfusion supersaturation. [264] Pneumatosis patients seem to produce excess

hydrogen. Exposure to alkyl halides (chloral hydrate, trichloroethylene, chloroform) can enhance hydrogen production in patients with pneumatosis cystoides coli by inhibiting bacterial hydrogen consumption. Indeed, associations between pneumatosis coli and chloral hydrate use (rapidly metabolized to its active metabolite trichloroethanol) have been reported and an epidemic of pneumatosis in Japan has been linked to trichloroethylene which is used in the watch-making and camera industries. [265], [266]

In asymptomatic patients, treatment is indicated only for associated illnesses, because the cysts usually resolve spontaneously. Medical therapy may be tried for symptomatic lesions, but recurrences may ensue. [267] High-flow oxygen breathing using 55% to 75% oxygen to achieve PO_2 values between 200 and 350 mm Hg for 4 to 10 days may result in cyst resolution. [260], [261] To avoid pulmonary and central nervous system toxicity, hyperbaric oxygen at 2.5 atm for 2.5 hours on 2 or 3 consecutive days has also been successful. [268] Antibiotic treatment with metronidazole or ampicillin and elemental diets have been used to resolve cysts. [260], [269], [270] Surgical resection should be reserved for severe, refractory symptoms or to manage complications. Unfortunately, surgery is not always successful, and pneumatosis may become more extensive after resection. [271] Surgery is indicated in fulminant cases of pneumatosis in which delay may lead to extensive necrosis of bowel, sepsis, and death. History, physical examination, CT scan of the abdomen, and evaluation for the presence of metabolic acidosis (especially lactic acidosis) and marked hyperamylasemia may help differentiate the patient with intestinal infarction. Mortality is high in these cases, even with surgery. [260], [261], [271] Finally, pneumatosis occurs in organ transplant patients; if present without an underlying systemic illness, it is usually a self-limited and benign illness but is a poor prognostic sign in the setting of systemic complications. [272]

MALAKOPLAKIA

This rare chronic, granulomatous, inflammatory disorder was described in 1902 by Michaelis and Gutmann and named by von Hansemann in 1903 (from the Greek: malakos, “soft”; plakos, “plaque”). It most commonly affects the urinary tract, but it can involve many other organs, including the male or female genital tract, skin, lung, bone, brain, and gastrointestinal tract. [273] The most common site of gastrointestinal malakoplakia is the distal colon and rectum, although all regions of the gastrointestinal tract may be involved. [275] There is a bimodal distribution of age incidence, with a small cluster of children younger than 13 years of age and a later peak among middle-aged adults. The average age of the adults was 57 years (range, 18–88 years), with a slight male preponderance; [275] most were Caucasian. The patient may be asymptomatic or may have diarrhea, abdominal pain, rectal bleeding, or symptoms of intestinal obstruction. Physical findings may include abdominal tenderness, rectal or abdominal mass, or weight loss.

The lesions are yellowish, soft plaques or nodules 1 to 20 mm in size or larger, usually elevated and sometimes with a central depression. [274], [275] and [276] They can be single, multiple, or involve adjacent structures (i.e., lymph nodes, mesentery, or retroperitoneum). Malakoplakia can present as an intestinal mass, stricture, or fistula, thereby mimicking other inflammatory or neoplastic processes.

Diagnosis is usually made on histological examination of biopsy material. Diagnosis by fine-needle aspiration has been reported. [277] Histologically, there is a diffuse histiocytic infiltrate with a strongly periodic acid-Schiff (PAS)–positive eosinophilic granular cytoplasm (von Hansemann cells) containing the characteristic basophilic, laminated cytoplasmic calculospherules called Michaelis-Gutmann bodies.

Predisposing conditions include chronic infections by coliform bacteria (especially *Escherichia coli*) [278], [279] and granulomatous processes such as sarcoidosis and tuberculosis. [273] Pulmonary malakoplakia has been associated with *Rhodococcus equi* infection in immunocompromised patients, especially those with AIDS. [280], [281] In addition, malakoplakia has been identified in patients who are immunosuppressed by steroids, immunosuppressant agents (transplant patients), hypogammaglobulinemia, or AIDS. [282] McClure [273], [274] reported that about one third of patients with gastrointestinal malakoplakia had associated colorectal carcinoma, and up to one half had a coexistent malignancy. Of interest, carcinoma-associated colonic malakoplakia is usually adjacent to the tumor. [283]

Although the pathogenesis is unclear, some theories exist. Many authors believe that there is a defect in the phagocytic or digestive activity of the macrophages. [284], [285] One patient had low levels of intracellular cyclic GMP with poor release of a-glucuronidase during phagocytosis. More recently, two kidney transplant patients with malakoplakia improved significantly after immunosuppressive therapy was tapered or discontinued, lending support to an immune origin. [282]

Because this condition is rare, no systematic evaluation of treatment has been undertaken. The possibility of associated malignancy or infection mandates a thorough history and physical examination. Excisional biopsy or fulguration can be used to treat localized intestinal malakoplakia. If involvement is more diffuse or extensive, empiric trials of antituberculosis medications, antibiotics (e.g., trimethoprim-sulfamethoxazole, ciprofloxacin), or cholinergic agonists (bethanechol, especially in patients with hypogammaglobulinemia) and stopping immunosuppressive agents have been used with varying degrees of success. [275], [285], [286] and [287]

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CHAPTER 86

William L. Hasler and Chung Owyang

IRRITABLE BOWEL SYNDROME

DEFINITION

Irritable bowel syndrome (IBS) is characterized by altered bowel habits and abdominal discomfort in the absence of organic disease. No clear diagnostic markers exist for IBS; thus, all definitions are based on the clinical presentation. The Rome II criteria for the diagnosis of IBS include at least 3 months of continuous or recurrent abdominal pain or discomfort that has two of the following features: relief by defecation, association with a change in the frequency, and a change in stool form (Table 86-1). [1]

At least 3 months of the following continuous recurrent symptoms:
Abdominal pain or discomfort that is relieved by defecation or is associated with change in frequency or consistency of stool
and
Disturbed defecation involving two or more of the following characteristics at least 25% of the time:
Altered stool frequency
Altered stool form (e.g., lumpy or hard, or loose or watery)
Altered stool passage (e.g., straining, urgency, or feeling of incomplete evacuation)
Passage of mucus
Bloating or feeling of abdominal distention

TABLE 86-1 Criteria for Diagnosing Irritable Bowel Syndrome

SOCIETAL IMPACT

IBS is the most prevalent digestive disease, accounting for 12% of visits to primary care physicians and 28% of referrals to gastroenterologists. [2] IBS is the seventh most common outpatient diagnosis with a rate of 10.6 visits per 1000 population per year. [3] Over 2 million prescriptions are written for IBS. [4] Although usually an outpatient disorder, IBS was the major discharge diagnosis in 96,000 inpatients as recently as 1976. [5] As awareness of the disorder has increased and hospital admission criteria have become stricter, admissions for IBS have markedly decreased. [6] Nonetheless, health care costs for the management of IBS are high. In a survey of health service utilization, the odds of a patient with IBS incurring charges were 1.6-fold greater than for a similar asymptomatic individual, which translated into overall median yearly charges of $742 in 1992 dollars for each patient with IBS compared to $429 for controls. [7]

IBS produces health consequences in addition to its economic costs. One survey [8] reported greater impairments in health-related quality of life in patients with IBS than the U.S. general population Decrements are most pronounced in energy/fatigue, role limitations caused by physical health problems, bodily pain, and general health perceptions. Another study noted that persons with the most intense IBS symptoms had the lowest quality of life measures. [9] IBS produces disability rates equal to or greater than severe organic gastrointestinal disease and is the second leading cause of absenteeism behind the common cold, with patients with IBS missing three times as many work days yearly as persons without IBS. [10], [11] and [12] The impact of IBS is emphasized by a British study reporting that 8% of patients with IBS retire early due to their symptoms. [13]

EPIDEMIOLOGY

IBS is a disorder of young people, with most new cases presenting before age 45. [3] However, some reports suggest that older adults are troubled by IBS symptoms up to 92% as often as middle-aged persons. [12], [14], [15], [16] and [17] Indeed, patients over 65 years of age make 500,000 physician visits per year for IBS. [4] Many of the diagnoses of “painful diverticular disease” given to older adult patients may represent IBS. [18] Functional gastrointestinal disorders also are prevalent in pediatric populations. IBS symptoms were reported by 14% of high school students and 6% of middle school students in one study and in 16% of students between 11 to 17 years of age in another. [19], [20] and [21] Women are diagnosed with IBS two to three times as often as men (Fig. 86-1). [3], [22], [23] Moreover, women make up 80% of the population with severe IBS. [24] Studies indicate that a history of abdominal pain or irregular bowel habits in first-degree relatives is significantly associated with IBS and dyspepsia. [25] Whether the familial associations represent similar exposures in a shared environment, heightened familial awareness of gastrointestinal symptoms, or genetic factors remains to be determined. Fewer Asians and Hispanics appear to develop the syndrome relative to other ethnic groups. [24], [26] The epidemiology of IBS outside the Western world is poorly characterized.

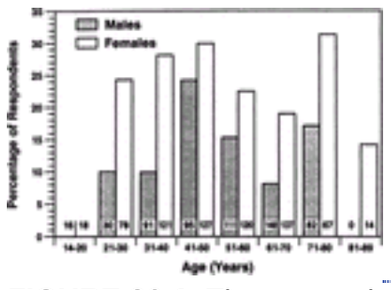

FIGURE 86-1. The age and gender distributions of individuals with symptoms of irritable bowel syndrome (IBS) are shown. The peak age range of symptom reports is in the forties, although both younger and older persons experience symptoms. Women report IBS-like symptoms significantly more than men across all ages. (From Longstreth GF, Wolde-Tsadik G. Irritable bowel–type symptoms in HMO examinees. Prevalence, demographics, and clinical correlates. Dig Dis Sci 1993;38:1581.)

The number of patients with IBS who seek medical care is considerably less than the number of individuals who experience symptoms. Using strict criteria, 9% to 22% of the population experience symptoms diagnostic for IBS, yet only a small fraction (9%–33%) seek medical attention, suggesting that other factors are important in the decision to obtain medical care. [14], [15], [27], [28]

CLINICAL FEATURES

Gastrointestinal Symptoms

IBS is a heterogeneous disorder with distinct symptom presentations. Different subsets of IBS can be defined based on the dominant symptom.

Abdominal Pain According to the new Rome II criteria, abdominal pain or discomfort is a prerequisite clinical feature of IBS. [29] The intensity and location of abdominal pain in IBS are highly variable, even within an individual patient. Abdominal pain in IBS is localized to the hypogastrium in 25%, the right side in 20%, the left side in 20%, and the epigastrium in 10% of patients. [30] The pain is generally crampy or achy, although sharp, dull, gaslike, or nondescript pains are also common. [31] It may be mild enough to be ignored or it may interfere with daily activities. Despite this, malnutrition due to inadequate caloric intake is exceedingly rare with IBS. Sleep deprivation also is unusual because abdominal pain is usually present only during waking hours. However, one study indicated that patients with severe IBS frequently wake repeatedly during the night and more than half of these patients reported that they were awakened by their abdominal pain. [32] Hence nocturnal pain is a poor discriminating factor between organic and functional bowel disease. Several factors exacerbate or reduce pain in IBS. Many patients report increased symptoms during periods of stress or emotional turmoil such as that associated with job or marital difficulties. Defecation may provide temporary relief from the pain of IBS, while meal ingestion may exacerbate the discomfort usually 60 to 90 minutes postprandially. This is especially true after consuming foods rich in carbohydrates and fat. [33] In addition female patients with IBS commonly report worsening symptoms during the premenstrual and menstrual phases. [34] Pain that is progressive, prevents sleep, leads to anorexia or inability to eat, or is associated with weight loss warrants exclusion of organic disease.

Alteration in Bowel Habits Patients with IBS present with constipation, diarrhea, or constipation alternating with diarrhea. With constipation, stools usually are hard and may be scybalous or pelletlike. Long periods of straining may be required for fecal evacuation both in constipation- and diarrhea-predominant patients. [35] Constipation can persist for weeks to months, interrupted by brief periods of diarrhea. Feelings of incomplete fecal evacuation may lead to multiple attempts at stool passage daily. In diarrhea-prone patients, stools characteristically are loose and frequent but of normal daily volume. Generally, diarrheal stools occur only during waking hours, often early in the day, and an urgent desire to defecate after a meal is reported by 36% of patients. [36] Mucus discharge has been reported in up to 50% of patients with IBS. [36] As with abdominal pain, fecal urgency and loose stools may develop during stress. Early descriptions of IBS observed patient populations who presented with painless diarrhea, representing 17% in one report. [31] However, disorders such as lactase deficiency were less readily appreciated and others such as microscopic and collagenous colitis were undefined; thus, it is conceivable many patients with functional diarrhea had one of these entities. In fact, painless diarrhea does not strictly fulfill the Rome II criteria to be classified as IBS. A recent survey revealed that women with IBS report mucus discharge, a sensation of incomplete fecal evacuation, pelletlike stools, and distention more commonly than men. [37] It is important to note that symptoms of IBS typically wax and wane over time. Patients may have some days with symptoms and some days without. In a 12-week trial study of 59 patients with IBS, Hahn and colleagues [38] found that the population reported pain or discomfort on 50% of days. On 33% of days bloating was reported, while altered stool formation and passage were found, respectively, on 25% and 18% of days. For more than half of the study period 50% of the patients had at least one symptom. The symptoms lasted on average for 5 days or less, although pain/discomfort and bloating tended to last the longest. Symptoms not associated with IBS that warrant exclusion of organic disease include nocturnal diarrhea, rectal bleeding, malabsorption, and weight loss.

Other Gastrointestinal Symptoms Upper gut symptoms are common in IBS, with 25% to 50% of patients reporting heartburn, early satiety, nausea, and vomiting; up to 87% note intermittent dyspepsia. [39] It appears that there is a great deal of overlap between dyspepsia and IBS. A community-based study in Spain [40] reported that the prevalence of IBS was higher among subjects with dyspepsia (31.7%) than among those who reported no symptoms of dyspepsia (7.9%). Conversely among the subjects with IBS, 55.6% reported symptoms of dyspepsia. Other studies [41] indicated that functional abdominal symptoms can change over time. Those with predominant dyspepsia or IBS can flux between the two. In contrast, those with predominant reflux symptoms seldom changed to dyspepsia or IBS. Thus it is conceivable that functional dyspepsia and IBS are two manifestations of a single, more extensive digestive system disorder. IBS symptoms are prevalent in patients experiencing noncardiac chest pain, but not in those experiencing cardiac chest pain, suggesting overlap with other functional gut disorders. [42] Patients with IBS may report increased gas production and abdominal distention, although most do not produce excess gas. [43] Another study indicated that the majority of patients with IBS have impaired transit and tolerance of intestinal gas loads. [44] This may be responsible for gas retention and the symptoms of gas and bloating in patients with IBS. Other patients with IBS may demonstrate a maximum rate of gas excretion greater than healthy subjects. [45] Although their total gas production was not increased, hydrogen production was higher. In these patients an exclusion diet reduced symptoms and produced a fall in maximum gas excretion. [45] In a recent study it was demonstrated that the perception of intestinal gas accumulation depends on the mechanism of retention. [46] Obstructed evacuation increased symptom perception whereas gas retention in a hypotonic gut was virtually unperceived. Computed tomography has shown increased lateral profiles in patients with IBS which was postulated to result from altered gastrointestinal motility or tone. [47] Another study has reported that those patients with significant bloating are most likely to have gained weight and more commonly had weak abdominal muscles. [48] This observation, however, was not confirmed by a subsequent study which showed no anterior abdominal muscle weakness in patients with IBS with gas and bloating. [49]

Extraintestinal Symptoms

Although gastrointestinal symptoms predominate, extraintestinal complaints are common in IBS. Patients with functional gut disorders visit primary care physicians three times as often for nongastrointestinal problems as do healthy persons and undergo more appendectomies and hysterectomies. [22], [50], [51] and [52] Chronic pelvic pain is much more commonly reported by patients with IBS than patients with inflammatory bowel disease. [53] Genitourinary dysfunction, including dysmenorrhea, dyspareunia, impotence, urinary frequency, nocturia, and a sensation of incomplete bladder emptying, is prevalent. [31] Impaired sexual function is reported by 83% of patients with IBS versus 30% of patients with inflammatory bowel disease and 16% of patients with peptic ulcer. [54] An association between IBS and primary fibromyalgia has been noted, with two thirds of patients with IBS reporting rheumatologic symptoms. Conversely, IBS symptoms are found in 42% of patients with fibromyalgia. [55] Patients with both IBS and fibromyalgia have worse scores on health-related quality of life indices than patients with either disorder alone. [56] The presence of fibromyalgia in patients with IBS seems to be associated with severity of bowel symptoms. [57] At the same time co-morbidity with fibromyalgia results in somatic hyperalgesia in patients with IBS. [58] Similarly, 63% of patients with chronic fatigue syndrome report symptoms of IBS. [59] Patients with functional bowel disorders have higher incidences of peptic ulcer disease, hypertension, low back pain, headaches, and rashes than the general population and more commonly report fatigue, loss of concentration, insomnia, palpitations, and unpleasant tastes in the mouth. [30], [60], [61]

PATHOPHYSIOLOGY

The pathogenesis of IBS is poorly understood, although roles for abnormal gut motor and sensory activity, central neural dysfunction, psychological disturbances, stress, and lumenal factors have been proposed. Unfortunately, there is no single pathophysiological abnormality that clearly separates IBS from organic gastrointestinal disease or from normalcy.

Gastrointestinal Motor Abnormalities

Abnormal Colonic Motor Activity The myoelectrical activity of the colon is comprised of slow waves, which regulate the frequencies of some phasic contractions, and spike potentials, which elevate the membrane potential above a contractile threshold. [62] Spike potentials may occur in the form of short-spike bursts (SSBs) of 5 to 15 seconds' duration which are temporally in phase with the slow-wave activity. Long-spike bursts (LSBs) of 15 to 60 seconds' duration are unrelated to the slow wave. [23], [63], [64] SSBs are responsible for generation of short-duration contractions, which are nonpropagative and segmenting in character leading to fecal mixing and resistance to stool passage. [62] LSBs produce long-duration contractions, some of which are nonpropagative and some of which may propel feces in either an oral or aboral direction. Finally, high-amplitude propagated contractions (HAPCs) are intensely propulsive, beginning in the right colon and occurring once or twice daily, producing mass movements of feces and defecation. [62] Studies of colonic myoelectrical and motor activity under unstimulated conditions have not shown consistent abnormalities in IBS. A predominant slow-wave frequency of 3 cycles per minute (cpm) with increased 3 cpm motor activity was observed in both diarrhea- and constipation-predominant patients with IBS in an older study that has not been confirmed by more recent studies. [65], [66] Furthermore, similar slow-wave abnormalities are demonstrable in psychoneurotic patients without bowel symptoms suggesting that the myoelectric disturbance is neither diagnostic for IBS nor pathogenic of symptoms. [67] Constipation-predominant patients were reported to exhibit increased numbers of SSBs while diarrhea-prone patients had reduced SSB activity. [63] Most investigators have observed no colonic motor abnormalities under basal conditions although others have reported increases in basal motility. [65], [66], [67], [68] and [69] Increased numbers of HAPCs have been noted in diarrhea-predominant patients with IBS compared to healthy volunteers. [70] Using scintigraphic assessment of colonic transit, selective acceleration in right colon transit has been shown in some patients with IBS with diarrhea. [71] It is unknown if these are pathogenic motor abnormalities in IBS or if all diarrhea-prone patients exhibit these findings. Similar studies have not been reported in constipation-predominant IBS, although patients with idiopathic constipation exhibit delayed right colon transit. [72] In contrast to the unstimulated colon, colonic motor abnormalities are more prominent under stimulated conditions in IBS. In normal volunteers, meal ingestion induces a rapid increase in spike potential and contractile activity that peak 20 to 30 minutes after eating, decrease to a basal level after 50 to 60 minutes, and may exhibit a second increase at 80 minutes. [63] Conversely, patients with IBS may exhibit increased rectosigmoid motor activity for up to 3 hours after eating. [64], [73] Although this response is most prominent in diarrhea-prone patients, constipated patients with IBS also show an exaggerated gastrocolonic response albeit to a lesser degree. [74] In contrast to healthy volunteers, sham feeding increases rectosigmoid motility in IBS suggestive of a cephalic phase of the gastrocolonic response in affected patients. [75], [76] and [77] Provocative stimuli induce exaggerated colonic motor responses in patients with IBS compared to healthy volunteers. Inflation of rectal balloons both in diarrhea- and constipation-predominant patients with IBS leads to marked distention-evoked contractile activity that may be prolonged. [64], [78] Patients with painful IBS exhibit greater increases in rectosigmoid motility after cholinergic stimulation than patients with painless diarrhea. [79] Intravenous cholecystokinin induces painful rectosigmoid contractions that reproduce the presenting symptoms in patients with IBS with postprandial pain. [80] An exaggerated motor response is demonstrable in patients with IBS after colonic perfusion of deoxycholic acid. [81] In contrast, one study reported rectal motor characteristics in response to isobaric distentions were not different between patients with constipation-predominant IBS, patients with slow transit constipation, and healthy controls. [82] On the other hand, sensations of urge were reduced in slow transit constipation and sensations of pain were increased in IBS. Until recently most of the colonic motility studies in IBS were performed in the rectosigmoid colon. Recording from the transverse, descending, and sigmoid colon, Chey and colleagues [83] reported that the motility index and peak amplitude of HAPCs in diarrhea-prone patients with IBS were greatly increased compared to healthy subjects. These were associated with rapid colonic transit and accompanied by abdominal pain. Similar findings were made by another group who reported increased numbers of HAPCs and reduced duration of increased colonic tone postprandially in a similar group of patients. [84]

Abnormal Small Bowel Motor Activity Small intestinal motor patterns under unstimulated conditions are organized and well characterized. During fasting, the migrating motor complex (MMC) cycles every 90 to 120 minutes to clear the proximal gut of undigested debris. Prolonged manometric studies suggest subtle abnormalities in fasting small intestinal motility in IBS. Decreases in MMC contractile amplitudes in constipation-predominant patients with IBS and reductions in MMC cycle length in diarrhea-predominant individuals have been characterized, although these abnormalities have not been universally described. [85], [86] These abnormalities have not been observed during sleep. [85], [86] and [87] Studies involving nonlinear analysis of 24-hour jejunal motility revealed that patients with IBS had increased irregularities for the intercontractile intervals during phase II of MMC. [88] Other physiological motor patterns include discrete clustered contractions (DCCs), which are bursts of phasic contractions occurring every minute in the duodenum or jejunum, and prolonged propagated contractions (PPCs), which are intense ileal complexes that evacuate the ileum and prevent coloileal reflux. In patients with IBS, DCCs and PPCs have been reported to make up a larger fraction of fasting recording time; however, this has not been confirmed by investigations (Fig. 86-2). [35], [89], [90], [91] and [92] It is unlikely that these complexes have pathogenic significance as their occurrences are similar in patients with constipation and with diarrhea.

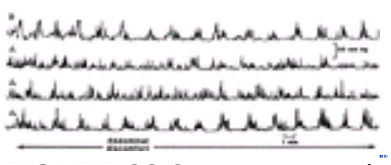

FIGURE 86-2. A manometric recording of small intestinal motility from a patient with irritable bowel syndrome shows 30 minutes of discrete clustered contractions in the duodenum and jejunum, which are associated with discomfort. (*D* duodenal pressure recording; $J_{1,2,3}$ jejunal pressure recordings). (From Kellow JE, Phillips SF. Altered small bowel motility in irritable bowel syndrome is correlated with symptoms. Gastroenterology 1987;92:1885.)

As in the colon, small intestinal motor abnormalities under stimulated conditions have been described in IBS. The fed-motor pattern is a period of intense irregular phasic contractions that occurs for 2 to 3 hours after eating. In both constipation- and diarrhea-predominant IBS, the duration of the fed pattern is shorter than in healthy volunteers. [35] Increases in the postprandial intestinal contraction frequency have been observed in diarrhea- and constipation-predominant IBS [92] although the contractile amplitudes were lower in constipation-predominant patients. [93] Some studies [94] further demonstrated that increased frequency of retrograde propagation of duodenal pressure waves occurred postprandially in both constipation-prone and diarrhea-prone patients with IBS. This abnormal propagation pattern of duodenal pressure waves was associated with symptoms in this group of patients. Intravenous cholecystokinin evokes exaggerated high-amplitude ileal contractions in diarrhea-predominant patients with IBS, while the anticholinesterase neostigmine induces frequent DCCs in both diarrhea- and constipation-prone patients. [95] Small intestinal transit of a meal is reportedly delayed in patients with IBS with constipation or distention plus abdominal pain whereas transit is characteristically accelerated in diarrhea-predominant patients. [18] Ileocecal function in IBS is variable with one study demonstrating delayed ileal emptying and decreased ileocecal clearance compared to healthy individuals and a second study reporting accelerated ileocecal transit both in constipation- and diarrhea-predominant patients. [96], [97]

Other Motor Abnormalities In addition to colonic and small intestinal motility disturbances, IBS is associated with motor abnormalities in other smooth muscle sites. Decreases in lower esophageal sphincter pressures and abnormalities of esophageal peristalsis have been found in IBS including increased spontaneous motor activity, repetitive contractions, and simultaneous waves in the esophageal body. [64], [98] In contrast, the upper esophageal sphincter, which is striated muscle, exhibits no manometric abnormalities in IBS. Gastric slow-wave dysrhythmias (tachygastria, bradygastria) have been reported in dyspeptic patients with IBS symptoms. [64] Fasting gallbladder volumes and residual volumes after contraction are greater in patients with IBS, whereas gallbladder emptying is impaired, although there is no increase in gallbladder disease with IBS. [99], [100] However, another study has reported that constipation-predominant patients exhibit enhanced gallbladder contraction, whereas patients with diarrhea exhibit impaired contraction compared to controls. [101] Sphincter of Oddi dysfunction has been described in association with IBS with demonstration of impairment of the sphincter relaxation in response to exogenous cholecystokinin. [102] Detrusor instability of the bladder has been demonstrated in 10 of 30 patients with IBS compared to 1 of 10 controls. [103] Similarly, patients with IBS exhibit hyperreactive airway function after administration of the smooth muscle stimulant methacholine. [104]

Visceral Sensory Abnormalities

A major focus of investigation is to define the visceral sensory abnormalities, which may be responsible for sensations of pain, gas, or bloating in IBS. Perception of abdominal symptoms is mediated by afferent neural pathways which are activated by visceral stimuli acting on chemoreceptors (which sense osmolarity, temperature, pH, etc.), mechanoreceptors (which are located in the gut wall and discharge in response to changes in tension), and receptors in the mesentery which may play a role in painful stimulation of the gut. [105] Information from these activated receptors is carried in spinal afferent nerves which synapse in the dorsal horn of the spinal cord. From there, the impulses are transmitted to the brain where conscious perception occurs. It is postulated that IBS results from sensitization of afferent pathways such that normal physiological gut stimuli not perceived by healthy individuals induce pain in the patient with IBS. The sensitizing event responsible for induction of symptoms in IBS is unknown.

Disturbed Sensation Under Basal Conditions The temporal association of symptoms with gut myoelectric and motor events provides evidence for disturbed visceral

sensation under unstimulated conditions. Myoelectric recordings show correlations between the appearance of intense, fused colonic SSBs and abdominal pain in constipation-predominant patients with IBS. [64] Irregular jejunal motor activity has been described coincident with the development of abdominal pain although other investigators have detected no abnormal motor patterns. [86], [106], [107] In one study, 61% of PPCs were associated with symptoms in patients with IBS compared to 17% in controls. [89] Similarly, DCCs were correlated with abdominal pain reports in 4 of 12 patients with IBS but in no healthy volunteers. [89] Another investigation has shown that nearly half of patients with IBS perceive different phases of the MMC as discomfort, a tugging sensation, or frank pain. [35] Thus, in contrast to healthy individuals, patients with IBS possess the ability to sense physiological motor events in the gastrointestinal tract. These findings may provide an explanation for older studies postulating a role for intestinal gas in the generation of abdominal symptoms in patients with functional bowel disorders. It was once believed that affected patients had abnormal amounts or distributions of intestinal gas. Early investigators described an entity called the splenic flexure syndrome, which consisted of severe left upper quadrant abdominal pain associated with apparent gas collections in the splenic flexure on abdominal radiographs. [108] Relief could be obtained by defecation, passage of flatus, or administration of an enema, and symptoms were reproducible by inflation of a balloon in the splenic flexure. Subsequent studies have shown that the absolute volume of gas in patients who complain of gas and bloating is normal. Using a washout technique, total volumes and transit times of intestinal gas were similar in asymptomatic individuals and in subjects complaining of excess gas. [43] However, patients with complaints of excess gas refluxed more gas from the intestine to the stomach and complained of distention and pain at low volumes of gas infusion that did not affect the controls. Thus, in addition to showing abnormal propulsion of intestinal gas, abnormal sensitivity to physiological amounts of intestinal gas is demonstrable in patients with functional symptoms.

Disturbed Sensation Under Stimulated Conditions As with studies of motor activity, patients with IBS frequently exhibit exaggerated sensory responses to visceral stimulation. Postprandial symptoms in IBS are associated with characteristic motor patterns. Postprandial pain has been temporally related to entry of the food bolus into the cecum in 74% of patients. [18] Some patients with concurrent nonulcer dyspepsia and IBS experience postprandial pain in association with increased rectosigmoid pressure. [109] Similarly, food has been shown to induce intestinal bursts that correlate temporally with pain. [110] A recent study showed that lipids lowered the thresholds for the first sensation of gas, discomfort, and pain in patients with IBS. [88] Furthermore, patients with IBS had an increased area of referred pain after lipids which was not observed in healthy subjects. Hence postprandial symptoms in patients with IBS may be explained in part by a nutrient-dependent exaggerated sensory component of the gastrocolonic response. Exaggerated symptoms can be induced by visceral distention in patients with IBS. Rectal balloon inflation produces nonpainful and painful sensations at lower volumes in patients with IBS than in healthy controls without altering rectal tension, suggestive of visceral afferent dysfunction in IBS (Fig. 86-3). [111] This effect persists even after exclusion of psychological factors such as neuroticism. However more recent studies [93], [94] suggested that psychological factors influenced pain thresholds in patients with IBS as stress altered sensory thresholds. When perception tests that minimize psychological influences were used, many patients with IBS showed normal sensory threshold. On the other hand, sexual abuse was not associated with lower pain threshold in patients with IBS. The visceral hyperalgesia of IBS appears to be selective for mechanoreceptor-activated stimuli as perception of intestinal mucosal electrical stimulation is normal in IBS. [112] In patients with IBS visceral hypersensitivity is not limited to the colon and rectum. Enhanced sensitivity to distension of the small bowel has been demonstrated. Jejunal sensitivity is increased after feeding in patients with IBS. [113] Furthermore nonperceived rectal stimulation appears to modify the intensity of jejunal perception to a greater extent in IBS than in healthy individuals. Similar studies showed gastric and esophageal hypersensitivity in patients with nonulcer dyspepsia and noncardiac chest pain, respectively. [111], [114] Furthermore, patients with IBS, nonulcer dyspepsia, or both all exhibited similar hypersensitivity to small intestinal distention, raising the possibility that these conditions have a similar pathophysiological basis. [115]

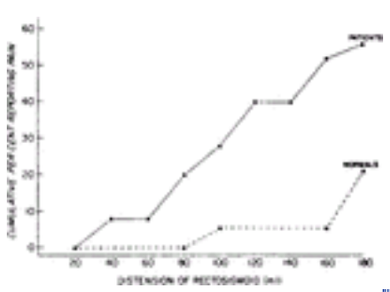

FIGURE 86-3. Perception of rectal balloon inflation is compared in healthy volunteers (*dashed line*) and patients with irritable bowel syndrome (IBS) (*solid line*). More patients with IBS reported pain with rectal distention. Furthermore, they experienced pain at much lower volumes than the healthy volunteers. (From Whitehead WE, Engel BT, Schuster MM. Irritable bowel syndrome: physiological and psychological differences between diarrhea-predominant and constipation-predominant patients. Dig Dis Sci 1980;25:404.)

Other studies of patients with IBS and nonulcer dyspepsia report enhanced sensitivity to esophageal distention, suggestive of a generalized visceral hyperalgesia in the functional bowel disorders. [116], [117] One group has categorized patients with IBS into three groups based on the response to rectal distention, including (a) those with a sensitive rectum, with a low sensory threshold to distention and normal to low intrarectal pressures, (b) those with a stiff rectum, with normal to low sensory thresholds and high pressures, and (c) those with an insensitive rectum, with a high sensory threshold and normal or high pressures. [118] The sensitive-rectum subtype was found in 57% of diarrhea-predominant patients versus 7% of patients with constipation. Seventy-five percent of diarrhea-predominant patients exhibited abnormal balloon distention responses versus 30% of constipated patients, suggesting that visceral hypersensitivity is not universal in IBS. This has been questioned in a study involving both rectal and sigmoid distention in patients with IBS. [119] In an investigation of patients with IBS with normal perception of rectal distention, all exhibited abnormal responses to sigmoid distention including alterations in colonic compliance and reflex relaxation. The duration of symptoms does not seem to be a factor in the degree of visceral hypersensitivity. No differences in threshold pressures for sensations of urgency and discomfort in patients with fewer than 2 years of symptoms have been observed compared to those with more than 5 years of complaints. [120] Finally, some investigators have postulated that IBS symptoms may result from interaction of visceral sensory dysfunction and abnormal gut motor patterns. In a study of women with IBS, those who exhibited enhanced perception of small intestinal distention had abnormal postprandial intestinal motor patterns. [121] In addition to visceral hyperalgesia, patients with IBS exhibit abnormal pain referral patterns with balloon inflation of the stomach, small intestine, or colon. [62], [122], [123] In contrast to healthy controls who perceive discomfort in only one quadrant with colonic distention, patients with IBS experience pain diffusely in the right upper and lower quadrants, right flank, hypochondrium, and epigastrium as well as in the back, shoulders, thighs, and chest (Fig. 86-4). [122] One group has proposed that, when alterations in viscerosomatic referral patterns are coupled with hypersensitive perception of rectal distention, visceral sensory abnormalities can be detected in nearly all patients with IBS and thus represent objective markers for the disorder. [124] These investigators further showed that repetitive sigmoid colonic distentions could induce development of rectal sensory abnormalities in 100% of patients with IBS, although the number of patients studied was small. [125]

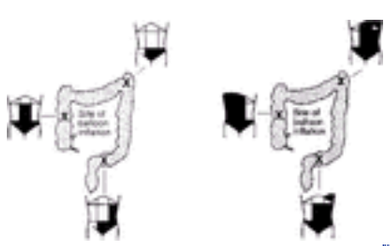

FIGURE 86-4. Pain referral patterns upon inflation of colonic balloons is compared in healthy volunteers (**left**) and patients with irritable bowel syndrome (IBS) (**right**). Colonic distention in healthy volunteers produced discomfort generally localized to a single abdominal quadrant while inflation in patients with IBS produced diffuse abdominal pain, providing evidence for altered viscerosomatic pain referral in IBS. (Adapted from Mayer EA, Gebhart GF. Basic and clinical aspects of visceral hyperalgesia. Gastroenterology 1994;107:271.)

In contrast to enhanced gut sensitivity, patients with IBS do not exhibit heightened sensitivity elsewhere in the body. Patients with IBS and Crohn's disease exhibit higher touch and pain thresholds with electrical stimulation of the skin than volunteers without these conditions. [126] Furthermore, patients with IBS, lactose intolerance, and healthy volunteers exhibit similar perception of ice water immersion of the hand. [127] Thus, the afferent pathway disturbances in IBS appear to be selective for visceral innervation with sparing of somatic pathways.

Central Nervous System Factors

The role of central nervous system factors in the pathogenesis of IBS is strongly suggested by the clinical association of emotional disorders and stress with symptom exacerbation and the therapeutic response to therapies that act on cerebral cortical sites.

Central Neural Dysfunction It is unknown whether IBS represents a primary gut disturbance with inappropriate input from the central nervous system or a central nervous system disorder with centrally directed changes in gut motor and sensory activity. [128] It is further undefined if the peripheral afferent nerve pathways that project from the gut to the brain exhibit abnormalities. New technologies are being developed to study this issue. Using cerebral evoked potential recordings, investigators reported that afferent pathways stimulated by esophageal distention are not abnormal in noncardiac chest pain and concluded that perceptual disturbances result from abnormalities in the central nervous system. [129] Conversely, evoked potential recordings in patients with nonulcer dyspepsia and patients with IBS show abnormal waveform latencies and amplitudes suggestive of visceral afferent dysfunction. [130], [131] Thus, the presence of peripheral afferent dysfunction in IBS is uncertain. Recent studies indicated that visceral hypersensitivity in IBS appears to be related to alterations in the nervous system rather than biochemical parameters such as the tension and strain of the gut wall. [132] Cerebral functioning in IBS is poorly understood; however, methodologies to assess neurological disease are being used to address this issue. Both mental stress and administration of the cholinesterase inhibitor neostigmine evoke increases in colonic motility and changes in electroencephalographic waveforms which are exaggerated in patients with IBS compared to healthy volunteers, suggesting that both the gut and brain are hypersensitive in IBS. [133] More recently, positron emission tomography has been employed to quantify regional cerebral blood flow in IBS. [134] In healthy individuals, rectal distention increased blood flow in the anterior cingulate cortex, a region that has been postulated to mediate the affective responses associated with pain. In contrast, patients with IBS exhibited no increased blood flow in this region but showed altered prefrontal cortical blood flow during sham rectal distention. This observation has been refuted by a functional magnetic resonance imaging (MRI) study, which reported that patients with IBS activated the anterior cingulate cortex to a greater extent than healthy controls in response to a painful rectal stimulus. [135] This suggests heightened pain sensitivity of the brain-gut axis in IBS, with a normal pattern of activation. The implications of these findings are unclear but they raise the possibility that central neural disturbances may play primary pathogenic roles in the induction of symptoms in IBS.

Abnormal Psychological Features Abnormal psychological features are recorded in up to 80% of patients with IBS, especially in referral centers; however, no single psychiatric diagnosis predominates. [3], [111], [136], [137] The lifetime incidences for major depression, somatization disorder, generalized anxiety disorder, panic disorder, and phobias are higher in patients with IBS than in healthy controls. [138], [139] An early investigation observed that 80% of patients with IBS had underlying depression or anxiety, whereas another study reported hysteria or depression in 72% of patients with IBS compared to 18% in normal volunteers. [31], [137], [140] Conversely when depressed patients are examined, 25% meet criteria for IBS compared to 2.5% of nondepressed controls. [141] Similarly IBS is found in 59% of dysthymic patients. [142] A significant overlap of panic disorder and IBS has been observed, with similar demographic and clinical characteristics. [143] In one study, 46% of patients with panic disorder met criteria for IBS versus 2.5% of controls. [144] Increases in neuroticism and decreases in extroversion are reported in IBS. [140] Additional abnormalities associated with IBS include hostility, hypochondriasis, and increased interpersonal sensitivity. [111] In an illness attitudes assessment, three abnormalities specific to IBS were bodily preoccupation, hypochondriacal beliefs, and disease phobia. [145] Psychological abnormalities predate or occur simultaneously with the onset of bowel symptoms in 67% to 85% of patients, suggesting that it is not the symptoms of IBS that induce psychiatric disease. [111], [146], [147] and [148] An association between prior sexual or physical abuse and development of IBS has been reported. [10], [149] Rates of severe lifetime sexual trauma, severe childhood sexual abuse, and any lifetime sexual victimization are significantly greater in patients with IBS than individuals with inflammatory bowel disease. [150] Forms of sexual abuse associated with IBS include verbal aggression, exhibitionism, sexual harassment, sexual touching, and rape. [151] Sexually traumatized patients experience significantly higher rates of lifetime development of depression, panic disorder, phobia, somatization, ethanol abuse, and sexual dysfunction. Furthermore, they exhibit higher rates of physician visits and more gastrointestinal and nongastrointestinal symptoms, and they undergo more surgeries. [149] Another study has reported histories of sexual abuse in 40% of patients with functional lower gastrointestinal disease both in tertiary referral centers and in private gastroenterology practices (Fig. 86-5). [152] Prior sexual abuse is more common in constipated patients and is not prevalent in patients with functional upper gut symptoms. In a complementary study, patients with prior abuse were more likely to report symptoms of IBS than those with no history of abuse. [153] From a pathophysiological standpoint, sexual abuse was not associated with lower pain threshold in patients with IBS. [154]

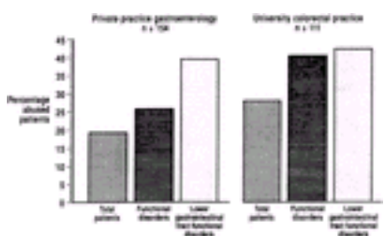

FIGURE 86-5. The prevalence of prior sexual abuse is shown for all patients seen in a private gastroenterology practice and a university practice as well as for patients with functional gut disorders in general and those localized to the lower gastrointestinal tract. Previous sexual abuse was reported significantly more often in those patients with functional lower gut disorders (such as irritable bowel syndrome) in both practice environments. (From Leroi AM, Berier C, Watier A, et al. Prevalence of sexual abuse among patients with functional disorders of the lower gastrointestinal tract. Int J Colorectal Dis 1995;10:200.)

Only a small fraction of individuals with symptoms of IBS present to a physician. Investigators have attempted to characterize differences between symptomatic individuals who seek medical attention and those who do not, known as nonreporters. Patients with IBS have been shown to exhibit more personality abnormalities and greater illness behavior than nonreporters. [50] Similarly, patients with IBS possess psychiatric disturbances greatly exceeding the general population; however, nonreporters are not different from asymptomatic individuals. [146] How a person responds to his or her symptoms determines whether he or she will present to a physician. Patients with IBS have histories of more frequent and serious childhood illnesses resulting in school absences and pediatrician visits and report that gifts were provided during childhood illness, suggesting that their expectations for illness behavior involve a reward. [60], [155]

Role of Stress on Visceral Function in Health and IBS The role of the central nervous system in modulating gastrointestinal function has intrigued researchers for decades. Early investigations of the effects of stress reinforce the importance of the brain-gut axis in the regulation of colonic activities. Painful somatic stimuli, such as ice water immersion of the hand or tightening of a circumferential band around the head, were shown to produce rectal spasm and mucosal engorgement. [156] Psychological stressors have been shown to have similar effects. One hoax involved telling a medical student during proctoscopy that a rectal cancer had been found, after which he was shown a piece of potato that he was told was a biopsy of the alleged malignancy. [157] As with the painful stimuli, the rectum exhibited intense contractions and mucosal reddening, which disappeared when the hoax was revealed. Similarly, in seven constipated subjects and eight with diarrhea-prone IBS, changes in rectosigmoid motility were inducible with stressful interviews. [158] Hostile or aggressive reactions provoked increased motor activity, whereas passive reactions or dejection led to motor quiescence. Other stressors such as anger, ball sorting, and stimulus differentiation testing increase colonic motility or spike potential activity in patients with IBS and controls. [68], [69], [159] The central nervous system also modifies visceral perception that suppresses or enhances afferent impulses from the gastrointestinal tract. [105] Hypnosis reduces perception of rectal distention in diarrhea-predominant patients with a lesser effect on constipated patients. [160] The lack of effect of hypnotherapy on rectal compliance or motor activity is indicative of active suppression of visceral afferent function by central nervous system activity. Stressful stimuli also induce physiological responses elsewhere in the gastrointestinal tract. Vertigo induced by cold caloric stimulation of the external ear delays gastric emptying and converts the fed-motor pattern to fasting complexes. [161] Barium radiography of the small intestine performed on subjects during stressful interviews showed segmentation, spasm, widening of the mucosal folds, and areas of distention. [162], [163] Psychological stress, as with intense participation in a video game or driving in rush hour traffic, decreased cycling of the intestinal MMC in healthy volunteers. [164] The relative effects of stress on small intestinal motility in patients with IBS compared to healthy individuals are controversial. Using radiotelemetry capsules in the small intestine, long periods of mental stress induced motor abnormalities in 19 of 22 patients with IBS compared to 1 of 10 controls. [107] In 7 patients, MMC cycling was eliminated and, in 18 patients, there were episodes of irregular motor activity lasting 1 to 6 hours. However, another study showed equivalent suppression of MMC activity in patients with IBS and controls as well as inhibition, rather than induction, of DCCs in both subject groups, suggesting that the intestine in patients with IBS may not be more susceptible to stress than asymptomatic controls. [165] Current stressful events may exacerbate symptoms in a large subset of patients with IBS, although they are not likely to be the cause of IBS. A strongly positive relationship has been reported between daily stress and daily symptoms in women with IBS. [166] Disturbances of sleep have been correlated with symptom development in IBS, especially in the morning. [167] In addition to current stressors, a history of severe emotional upheaval in the distant past often is elicited upon careful questioning. Loss of a parent, either through death or marital dissolution, has been reported to be an important stressor in one third of patients. [168], [169] and [170] A strong association between IBS and posttraumatic stress disorder has also been noted. [171] Furthermore, military veterans returning from the war in the Persian Gulf presented with gastrointestinal complaints and exhibited visceral hyperalgesic properties similar to those of patients with IBS, suggesting that a subset of persons with Gulf War syndrome may in fact have IBS. [172]

Other Studies on the Pathogenesis of IBS

Induction of IBS by Gastrointestinal Infection Some recent studies correlate development of IBS with a prior gastrointestinal infectious illness. In an investigation of 544 patients with confirmed bacterial gastroenteritis, one fourth reported persistently abnormal bowel habits 6 months later. [173] Seven percent developed IBS with a

3.4-fold increased risk in women compared to men. A similar study of 75 patients with acute gastroenteritis requiring hospital admission observed that 20 had persistent symptoms for more than 6 months compatible with the diagnosis of IBS. [174] Similarly, another group [175] reported that about a third of patients with IBS experienced an acute "gastroenteritis-like" illness at the onset of their chronic IBS symptomatology. The previously cited studies have shown that IBS following an enteric infection occurred more commonly in females, and affected younger rather than older patients and those who had a protracted acute diarrheal illness. Psychometric assessment at the time of acute infection found that those who developed IBS had higher scores for anxiety, depression, somatization, and neuroticism compared to those who recovered. Increases in ileal mast cell density are reported in IBS, raising the possibility that subtle inflammation is pathogenic. [176] The microbes involved in the initial infection were *Campylobacter Salmonella* and *Shigella*. [173], [174] and [175] Those patients with *Campylobacter* infection who were toxin-positive were more likely to develop postinfectious IBS. [177] Increased rectal mucosal enteroendocrine cells, T lymphocytes, and increased gut permeability were acute changes following *Campylobacter enteritis* which could persist for more than a year and may contribute to postinfectious IBS. [178] In a provocative study, Pimentel and associates [179] reported that of 202 patients with IBS 78% had small intestinal bacterial overgrowth. Forty-eight percent of the subjects who had eradication of bacterial overgrowth experienced symptomatic improvement. This interesting finding has far-reaching clinical implications but requires confirmation.

Role of Malabsorption or Intolerance to Specific Foods The role of carbohydrate malabsorption as a cause of bloating, pain, and diarrhea in IBS has been extensively evaluated. Lactase deficiency is a well-described syndrome where ingestion of milk products causes symptoms similar to those in IBS. Using hydrogen breath testing, 25.8% of one group of patients with presumed IBS exhibited lactase deficiency but 52% of these individuals did not relate their symptoms to lactose ingestion. [180] Another study of lactase-intolerant patients with IBS reported that symptoms were independent of lactase supplementation when given in placebo-controlled fashion, suggesting that lactase deficiency was not pathogenic of IBS in these patients. [181] Conversely, a recent hydrogen breath test analysis of patients with presumed IBS identified a subset of patients with lactase deficiency unsuspected by the clinical history who responded to dietary lactose exclusion. [182] This observation was confirmed in another study which reported that the majority of patients with IBS with lactose malabsorption (which was previously clinically unrecognized) placed on a lactose-restricted diet noticed markedly reduced symptoms both in the short term and the long term. [183] These studies suggest that lactose malabsorption should be excluded before diagnosing IBS. Malabsorption of other sugars has been proposed as a cause of symptoms in some patients with IBS. In one study, fructose and sorbitol, which are found in fruits as well as soft drinks and candy, produced prominent symptoms in most subjects; however, dietary exclusion of these sugars was not performed. [184] A more recent investigation reported increased symptoms in patients with functional gastrointestinal disease after lactose or fructose plus sorbitol compared to healthy controls. [185] In this study, 40% of patients experienced symptom reduction after restriction of the offending carbohydrate. A study in Israel indicated that combined carbohydrate malabsorption was common in IBS and may contribute to symptomatology in many of the patients investigated. [186] However, the weight of available evidence suggests that carbohydrate malabsorption is not the cause of symptoms in most patients with IBS in the United States, although intolerance to certain sugars probably plays a role in some individuals. Food hypersensitivity likely plays only a minor role in most patients, although some individuals with IBS may have a true allergic component to their symptoms. In a study of 14 patients with diarrhea-predominant IBS who noted symptoms after eating wheat, dairy products, coffee, tea, or citrus fruits, a blinded challenge with the offending food led to worsening of the symptoms in six patients; this was associated with elevations in rectal prostaglandin E_2. [187] However, subsequent studies [188], [189] have reported that only a small fraction of diarrhea-prone patients with IBS improve on an exclusion diet, an effect that may not be greater than with placebo. Even those individuals with positive skin test responses to particular foods do not show symptom resolution upon dietary exclusion of those foods. [190]

Disturbed Neurohumoral Activity Systemic disturbances in neural and hormonal activity have been explored as pathogenic factors in IBS. Efferent vagal dysfunction is suggested by demonstrations of reductions in vagally mediated increases in lower esophageal sphincter pressure after abdominal compression, impaired pulse variability with inspiration, and reduced ratios of insulin-induced peak gastric acid output to pentagastrin-stimulated maximal acid output in subsets of patients with IBS. [191] Decreases in digital temperatures and enhanced peripheral electromyographic activity have led investigators to postulate a diffuse autonomic neural disturbance in IBS. [192] A quantitative assessment of sympathetic adrenergic and cholinergic and cardiovagal cholinergic function revealed abnormalities in 9 of 33 patients with IBS. [193] Another investigation suggested that constipation-predominant patients with IBS exhibit abnormalities of cholinergic function, while diarrhea-prone individuals have evidence of adrenergic neural dysfunction. [194] Parasympathetic tone was significantly lower in the constipation-predominant patients with IBS compared with the diarrhea-predominant patients with IBS. [195] Prolongation of rapid eye movement (REM) sleep with episodes of sleep apnea has been reported in IBS, raising the possibility that the central nervous system exhibits abnormalities outside of the brain-gut axis. [196], [197] Blunting of a-adrenergic pathways in the central nervous system is suggested by the observation of impaired growth hormone release in patients with IBS, but not patients with ulcer or healthy controls, in response to a2-adrenoceptor stimulation. [198] Patients with IBS appear to have an exaggerated response to corticotropin-releasing hormone (CRH). [199] Not only did exogenous CRH induce a higher plasma corticotropin level in patients with IBS, it also produced abdominal symptoms accompanied by a greater motility index in the colon as well as duodenal dysmotility in this group of patients. Diarrhea-predominant patients with IBS showed a significant postprandial increase in cortisol which was not evident in healthy subjects or constipation-predominant patients with IBS. [200] Hormonal disturbances have been postulated to be pathogenically important in patients with IBS who experience symptoms during menses. [52], [201] Abnormal circulating pancreatic polypeptide, neurotensin, insulin, motilin, cholecystokinin, and gastrin levels are reported in some patients with IBS. [202], [203] and [204] In women with IBS, higher urinary norepinephrine, epinephrine, and cortisol levels have been observed. [205] The roles of these neurohumoral disturbances in the pathogenesis of IBS are unclear but are likely to be epiphenomenal. Serotonin (5-HT) is present in abundance in the gastrointestinal tract. It may play an important role in the regulation of gastrointestinal motility and visceral perception. One study indicated that 5-HT–containing enterochromaffin cells in the colon were increased in diarrhea-predominant patients with IBS compared to healthy subjects or patients with ulcerative colitis. [206] Furthermore, postprandial plasma 5-HT plasma levels were significantly higher in diarrhea-predominant patients with IBS compared to healthy controls. [207] This suggests a possible role for 5-HT in the postprandial symptoms of these patients and provides a rationale for the use of 5-HT antagonists in the treatment of this disorder.

Abnormal Stool Characteristics Abnormal characteristics of the fecal material have been postulated as symptom inducers in IBS. As bile acids induce colonic contractions, investigators have proposed that diarrhea-predominant patients with IBS have bile acid malabsorption. [63] Bile acid binders such as cholestyramine are used in IBS with anecdotal reports of success; however, this may be due to the general constipating effects of the resin. However, one study reported that about one third of patients with IBS had bile acid malabsorption and 70% of these patients responded to bile acid sequestrants whereas only 15% responded to conventional therapy. [208] Others have proposed that elevated levels of fecal short-chain fatty acids may cause diarrhea in IBS; however, symptoms do not correlate with fecal levels of these compounds. [209] An investigation revealed that diarrhea-prone patients have lower fecal concentrations of total short-chain fatty acids, acetate, and propionate, and higher levels of *N*-butyrate than controls. [210] Gut flora disturbances have been suggested as causes of symptoms in some patients, and trials to modify the dominant organisms have met with some success in reducing symptoms. [211], [212] The demonstration of increased hydrogen production in patients with functional gut disease after lactulose is supportive of qualitative and/or quantitative alterations in the fecal flora of some patients with IBS. [185] Regardless, to date there is little convincing evidence that stool characteristics are abnormal in IBS.

DIAGNOSTIC APPROACH TO THE PATIENT WITH PRESUMED IBS

History and Physical Examination

Confident diagnosis of IBS relies on recognition of characteristic symptom profiles as well as the detection of alarm findings (weight loss, bleeding, fever, or palpable masses) which are more suggestive of organic disease. The symptoms that define IBS also are prominent complaints of many other conditions including malignancies, inflammatory diseases, infections, and ischemic diseases of the gastrointestinal tract as well as some nongastrointestinal conditions. Several investigators have attempted to ascertain if specific symptom patterns can distinguish IBS from more severe illnesses. In a questionnaire survey, Manning and colleagues [213] defined four symptoms that were significantly more common in IBS than organic disease including relief of abdominal pain upon defecation, looser stools with onset of pain, more frequent stools with the onset of pain, and abdominal distention. Ninety-one percent of patients with IBS had two or more of these symptoms versus 30% of those with organic disease. Other symptoms which were common but which did not distinguish IBS from organic disease included passage of mucus, a sensation of incomplete evacuation, fecal urgency, harder or less frequent stools at pain onset, pain eased by passage of flatus, and defecation before breakfast, at night, or between meals. In a study by Kruis and associates, [214] an absence of organic disease was predicted by (a) the combination of pain, flatulence, and irregular defecation, (b) symptoms persisting for more than 2 years, (c) pain described as burning, cutting, very strong, or terrible, (d) a defecation pattern of alternating defecation and constipation, (e) pencil-like or pellet-like stools, and (f) the passage of mucus in the stools. These initial investigations have been followed by international assemblies that have devised the commonly employed Rome I, and more recently Rome II, criteria. [29]

Studies have quantified the reliability of symptom-based criteria in the diagnosis of IBS. An early evaluation observed that the Manning criteria could distinguish IBS from organic disease with a sensitivity of only 58% and a specificity of 74%. [215] In a second study, the Manning criteria were found to discriminate IBS from organic disease with a sensitivity and specificity of 90% and 87%, respectively, while the Kruis system exhibited a sensitivity of 81% and a specificity of 97%. When employed together, the two sets of criteria exhibited 80% sensitivity and 97% specificity for the diagnosis of IBS. [216] A retrospective analysis [217] determined that, in the absence of alarm symptoms or physical findings, the Rome I criteria had a sensitivity of 65%, a specificity of 100%, and a positive predictive value of 100% for the diagnosis of IBS. Furthermore, no patient required revision of the diagnosis in a 2-year follow-up. When evaluated prospectively, the positive predictive value of the Rome I criteria

was 98%. [217]

The physical examination of the patient with IBS usually is normal. Abdominal compression may elicit tenderness that is vague and poorly localized. Tender bowel loops are commonly palpable. [22] Masses, adenopathy, hepatosplenomegaly, ascites, blood in the stool, or autonomic or peripheral neuropathy are suggestive of organic disease and are not consistent with IBS.

Laboratory and Structural Findings

A minimal laboratory and structural evaluation has been advocated as a screen for organic disease in the patient with presumed IBS. Basic testing includes a complete blood count to assess for anemia, leukocytosis, or leukopenia, a sedimentation rate to screen for inflammation, thyroid chemistries to exclude hormonal causes of altered bowel function, and serum electrolytes and stool studies for ova and parasites or *Giardia* antigen in patients with diarrhea. For diarrhea-predominant individuals, hydrogen breath testing to exclude lactose intolerance should be considered. [218] Empiric dietary lactose restriction is an alternative when breath testing is not available. For pain-predominant patients, plain abdominal radiography during an episode of pain can exclude intermittent bowel obstruction. [218] Sigmoidoscopy is recommended for most young individuals (<50 years old) to rule out distal obstruction in patients with constipation and inflammatory diseases in those with diarrhea. In diarrhea-predominant patients, random biopsies are obtained to search for microscopic colitis. Sigmoidoscopy should reveal normal anatomy in IBS, although increased mucus secretion, mucosal engorgement, and pain with air insufflation are commonly noted. [22] In patients over the age of 50, colonoscopy or air-contrast barium enema radiography are indicated because of the prevalence of colonic neoplasm in this age group. A technical review for practice guideline development by the American Gastroenterological Association [218] delineated factors to be considered when determining the aggressiveness of the diagnostic evaluation. These include the duration of symptoms, the change in symptoms over time, the age and sex of the patient, the referral status of the patient, prior diagnostic studies, a family history of colorectal malignancy, and the degree of psychosocial dysfunction. Thus a younger individual with mild symptoms requires a minimal (or in select cases no) diagnostic evaluation while an older person or an individual with rapidly progressive symptoms should undergo a more thorough exclusion of organic disease.

The validity of this approach has been subject to scrutiny. In the absence of alarm findings, a normal complete blood count and sedimentation rate provide 83% sensitivity and 97% specificity for the diagnosis of IBS. [30] However, a study of 196 patients satisfying the Rome I criteria found no diagnostic yield from sedimentation rates, thyroid profiles, and stool examinations in the absence of specific historical or physical examination findings. [180] Furthermore, barium enema radiography or lower endoscopy revealed only one case with colon cancer, nine with polyps, and one with colitis out of a sample of 196 patients. [180] In data from two other multicenter trials, lactose intolerance was diagnosed in 23% of patients, colonic structural abnormalities were found in 2%, abnormal thyroid chemistries were observed in 6%, and positive fecal ova and parasites were detected in 2%. [219] Nevertheless, a thorough initial evaluation of the patient with presumed IBS precludes the need for further diagnostic testing years later. In 112 patients with IBS followed for a median of 29 years, organic disease developed in only 10 patients a median of 15 years after the original diagnosis of IBS. [220]

TREATMENT

The treatment of IBS ranges from patient education and dietary modifications to medications or even psychological intervention. Some patients, especially those with new onset symptoms, will express relief that they do not have a serious condition such as cancer. Whenever possible, the physician should resist the temptation to rely on drugs which might provide only limited efficacy and which could provoke unacceptable side effects. Nonetheless, medications are prescribed at 75% of outpatient visits for IBS. [3] High placebo response rates ranging from 30% to 70% have diminished the ability of controlled studies to detect therapeutic benefits of drug therapies of IBS. [221] Reviews have judged that few studies offer convincing evidence of drug efficacy, although some benefit for antispasmodics in reducing pain and for loperamide in controlling diarrhea has been observed. [222], [223] Critiques of published IBS drug trials raised methodological concerns about the peer-reviewed literature including studies too short in duration to detect efficacy, poorly designed crossover protocols in which washout periods between active drug and placebo were inadequate, inappropriate statistical methods, and large placebo effects which would mandate the enrollment of hundreds of patients to detect a statistically significant drug effect. [221] In the design of new studies, these critiques recommended careful description of the randomization method, use of international diagnostic criteria, use of placebo control and double-blinding, and defining clear outcome measures.

Dietary Measures and Fiber Supplements

Dietary modifications are commonly recommended in IBS, although little controlled investigation has been performed. It is reasonable to limit fat intake as lipids most potently activate exaggerated motor reflexes such as the gastrocolonic response. Patients with bloating and diarrhea should restrict their intake of poorly digestible sugars such as fructose and sorbitol. A subset of patients with excess gas may derive benefit from additional dietary modification. Certain foods promote intestinal gas production. Pork and bean meals increase colonic gas passage from 15 to 176 mL/h as a consequence of maldigestion of oligosaccharides such as stachyose and raffinose. [224] Fiber content in the diet also can contribute to gas generation. A high-fiber diet with beans produces 49.4 mL of gas/h vs. 10.9 mL/h on a liquid low-fiber diet. [225] However, most patients with IBS who complain of excess flatus in fact produce normal amounts of gas. Exclusion of foods associated with increased flatulence including beans, onions, celery, carrots, raisins, bananas, apricots, prunes, brussels sprouts, wheat germ, pretzels, and bagels has been reported to reduce flatus expulsions in uncontrolled reports of patients with excess gas (Table 86-2). [226]

Normoflatulogenic Foods
Meat, fowl, and fish
Vegetables (e.g., lettuce, cucumber, broccoli, pepper, avocado, cauliflower, tomato, asparagus, zucchini, okra, olives)
Fruits (e.g., cantaloupe, grapes, berries)
Carbohydrates (e.g., rice, corn chips, potato chips, popcorn, graham crackers)
Nuts
Miscellaneous (e.g., eggs, non-milk chocolate, flavored gelatin, fruit ice)
Moderately Flatulogenic Foods
Pastries
Potatoes
Eggplant
Citrus fruit
Apple bread
Extremely Flatulogenic Foods
Milk and milk products
Vegetables (e.g., onions, beans, celery, carrots, brussels sprouts)
Fruit (e.g., raisins, bananas, apricots, prune juice)
Miscellaneous (e.g., pretzels, bagels, wheat germ)

Adapted from VanNess MM, Cattau EL. Flatulence: pathophysiology and treatment. Am Fam Physician 1985;31:108.

TABLE 86-2 Foods and Flatus Production

Fiber supplements are widely recommended for treating IBS and exhibit several properties that theoretically should be beneficial in IBS including enhanced water retention and bulking of the stool, formation of gels to provide stool lubrication, and binding bile acids which could cause symptoms in some cases. [227] Of note, patients with IBS do not consume less dietary fiber than healthy individuals. [228] Studies in animal models have observed induction of propagative colonic motor activity with augmentation of dietary fiber content. [229] In a review of human investigations, bran increased fecal weight in 18 of 20 studies and accelerated fecal transit in 16. [230] Bran accelerated colonic transit in healthy volunteers with an initial colonic transit time of 3 or more days, whereas it retarded propulsion in those with a transit time of 1 day suggesting that dietary fiber promotes a more regular bowel pattern in normal individuals. [231] Psyllium reduces perception of rectal distention, indicating that fiber may have positive effects on visceral afferent function as well. [232]

Fiber supplements that have been studied in IBS in placebo-controlled fashion include bran, psyllium, process flea seed husk (ispaghula), and calcium polycarbophil. Because of high placebo response rates and enrollment of small numbers of patients, studies have failed to document a therapeutic benefit of fiber supplementation in the IBS population as a whole. [221] Most investigations report increases in stool weight, decreases in colonic transit times, and improvement in constipation. [233], [234], [235] and [236] Others have noted benefits in patients with alternating diarrhea and constipation, pain, and bloating, however most studies observe no responses in patients

with diarrhea- or pain-predominant IBS. [237] It is possible that different fiber preparations may have dissimilar effects on selected symptoms in IBS. One study observed symptom exacerbation in 55% of patients with IBS with bran, while only 10% experienced symptom improvement. [238] A crossover comparison of different fiber preparations found that psyllium produced greater improvements in stool pattern and abdominal pain than bran. [239] Furthermore, bran exacerbated complaints of abdominal distention while psyllium supplements reduced such complaints. A third study reported increases in flatulence with bran therapy. [240] Additionally, patients with IBS were noted to benefit from placement on a fiber-restricted diet but with use of commercial bulking agents as needed suggesting that low-fiber diets may be advisable in some patients. [241]

Medication Treatments

Osmotic Laxatives For patients with constipation-predominant IBS who do not respond to fiber, osmotic laxatives are often recommended to effect defecation. Available agents include hypertonic salt solutions such as milk of magnesia, poorly absorbable sugars such as lactulose and sorbitol, and isotonic electrolyte solutions containing polyethylene glycol. There is little controlled literature to support the use of osmotic laxatives specifically in IBS, although several studies do report benefits for polyethylene glycol solutions in accelerating colonic transit and increasing stool frequency in patients with idiopathic slow transit constipation. [242]
Antidiarrheal Agents Peripherally acting, opiate-based agents are the initial therapy of choice for diarrhea-predominant IBS. Physiological studies demonstrate increases in segmenting colonic contractions, delays in fecal transit, increases in anal pressures, and reductions in rectal perception with these drugs. Placebo-controlled investigations of loperamide report decreases in stool frequency and urgency, improved consistency, reduced borborygmi, and benefits in well being in patients with diarrhea or alternating diarrhea and constipation. [243] Other agents have been proposed for use in patients with IBS with diarrhea. An uncontrolled study of diarrhea-predominant patients with IBS noted a 70% response to bile acid–sequestering drugs. [208] Acid suppressing drugs in the H_2 receptor antagonist and proton pump inhibitor classes have been reported in an uncontrolled trial to reduce diarrhea in IBS. [244]
Antispasmodic Agents Antispasmodic agents are the most commonly prescribed agents for IBS and are the initial recommended therapy for pain-predominant IBS. [218] Included in this category are agents that block cholinergic nerve function, drugs that prevent calcium flux, direct gut smooth muscle relaxants, and agents that act via unknown pathways. From a physiological standpoint, anticholinergic drugs and calcium channel antagonists are potent inhibitors of the gastrocolonic response. [245], [246] A metaanalysis of 26 double-blind clinical trials of antispasmodic agents in IBS reported better global improvement (62%) and abdominal pain reductions (64%) compared to placebo (35% and 45%, respectively), although none of the medications analyzed (cimetropium, pinaverium, trimebutine, octylonium, and mebeverine) are approved in the United States. [247] A second metaanalysis of controlled trials of cimetropium, hyoscine, mebeverine, octylonium, pinaverium, and trimebutine quantified improvements in global well being in 56% versus 38% of patients on placebo and reductions in pain in 53% compared to 41% of patients on placebo. [248] Anticholinergic agents are the major class of antispasmodics prescribed in the United States. In small trials, dicyclomine, prifinium, and cimetropium have been reported to reduce symptoms including fecal urgency and pain in IBS compared to placebo. [249], [250] However, issues of blinding, study duration, and statistical analysis raise questions as to the validity of these conclusions. [221], [249] The gut selective anticholinergic M_3 muscarinic receptor antagonist zamifenacin has been demonstrated to blunt the colonic motor response to meal ingestion and shows promise as therapy for pain-predominant illness. [251] In animal models, M_3 receptor antagonists inhibit diarrheal responses to stress, serotonin, prostaglandins, and castor oil. [252] Calcium channel blockers have been proposed for use in IBS because of their smooth muscle relaxant properties and their inhibitory effects on the gastrocolonic response. A double- blind trial of diltiazem did not show global improvement, but did observe trends to reduced diarrhea and abdominal pain. [253] In a randomized, placebo-controlled trial, pinaverium reduced the duration of abdominal pain in IBS. [254] Octylonium produced significant improvements in the number of pain episodes, the severity of distention, and global assessments in a placebo-controlled trial in 325 patients. [255] Peppermint oil relaxes gastrointestinal smooth muscle via reduction of calcium influx. [256] In a recent large trial, 79% of subjects experienced pain reduction, 83% reported less abdominal distention, 83% had reduced stool frequency, and 79% passed less flatus, values that were all superior to placebo. [257] A metaanalysis of eight randomized, controlled trials reported a trend toward therapeutic benefit of peppermint oil in IBS, however this could not be definitively concluded due to methodological concerns. [258] Peppermint oil may also be of benefit in children with IBS, as the agent has been shown to reduce abdominal pain severity in 75% of cases. [259] Other antispasmodics unavailable in the U.S. that do not act via inhibition of cholinergic activity or calcium flux have been used for IBS. Mebeverine is a smooth muscle relaxant similar to papaverine which inhibits ileal and colonic motility in patients with IBS. [260] Two small studies have reported benefits for mebeverine over placebo while a third investigation observed no improvement. [260], [261] In a nonplacebo-controlled trial [262] both the short- and long-acting forms of mebeverine have been reported to be “effective” or “very effective” by more than 80% of patients with IBS. Trimebutine is an antispasmodic which stimulates small intestinal motility by acting on peripheral opiate receptors but inhibits colonic motility via a aloxone-insensitive pathway. [263] In the largest placebo-controlled trial involving 60 patients with IBS, [264] there were no differences in pain relief with trimebutine versus placebo.
Antidepressant Agents Antidepressant agents in the tricyclic class have demonstrated clear efficacy in selected IBS subsets, but the mechanisms of their beneficial effects are uncertain. In diarrhea-predominant patients with IBS, imipramine slows propagation of jejunal pressure waves and delays orocecal and whole gut transit indicative of a motor inhibitory effect. [265], [266] Some investigations have reported reduced perception of luminal distention suggestive of inhibition of visceral afferent transmission, although other studies have shown no effects on visceral perception. [267], [268] The effects of tricyclic antidepressants on central neural processing of painful visceral stimulation are unknown. A controlled trial of amitriptyline reported benefits in global well being, abdominal pain, and bowel pattern and identified young age and increased extroversion as predictors of drug response. [269] Studies of trimipramine have reported improvements in vomiting, depression, sleeplessness, and fecal mucus with variable reductions in pain compared to placebo. [270] Placebo-controlled trials of desipramine observed decreases in pain, stool frequency, and depression which were confined to diarrhea-predominant patients. [271] Nortriptyline given in combination with fluphenazine was reported to reduce pain and diarrhea compared to placebo. [272] The efficacy of antidepressant agents in other classes in treating IBS is less clear. In contrast to tricyclic agents, the selective serotonin reuptake inhibitor (SSRI) paroxetine accelerates orocecal transit raising the possibility that this drug class may be useful in constipation-predominant patients. [266] The SSRI citalopram blunts perception of rectal distention and reduces the magnitude of the gastrocolonic response in healthy volunteers. [273] A small placebo-controlled study of citalopram in patients with IBS reported reductions in pain. [274] An investigation of mianserin, with serotonin 5-HT $_2$ and 5-HT $_3$ receptor antagonist and a $_2$-adrenoceptor antagonist effects, reported reductions in pain, distress, and functional disability compared to placebo. [275] A metaanalysis of tricyclic and nontricyclic antidepressants reported an odds ratio for clinical improvement of 4.2 (confidence interval 2.3–7.9) (Fig. 86-6). [276] On average, 3.2 patients needed to be treated to produce symptom improvement in 1 patient. A second metaanalysis of all antidepressant trials in IBS reported an odds ratio for improvements in pain of 8.0 and for global improvement of 4.4. [277]

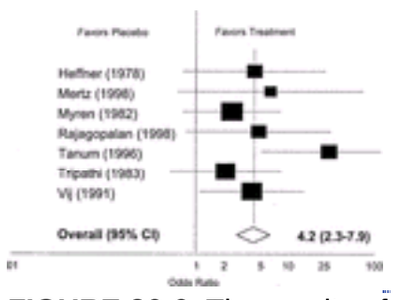

FIGURE 86-6. The results of metaanalysis of antidepressant medications on the overall improvement in gastrointestinal symptoms are shown. The *boxes* represent the odds ratios favoring active drug or placebo while the *horizontal lines* show the 95% confidence intervals. The box sizes are proportional to each study’s weight in the analysis. When the data are pooled, the odds ratio for a benefit of antidepressant treatment is 4.2 with a 95% confidence interval ranging from 2.3 to 7.9. (From Clouse RE, Prakash C, Anderson RJ, Lustman PJ. Antidepressants for functional gastrointestinal symptoms and syndromes: a meta-analysis. Gastroenterology 2001;120:3252[abst].)

Serotonin Receptor Antagonists Serotonin receptor antagonists have been evaluated as therapies for diarrhea-predominant IBS. Serotonin acting on 5-HT $_3$ receptors enhances the sensitivity of afferent neurons projecting from the gut. [278] In humans, the 5-HT $_3$ receptor antagonists ondansetron, granisetron, and alosetron reduce perception of painful visceral stimulation in IBS. [279], [280] 5-HT $_3$ receptor antagonists also induce rectal relaxation, increase rectal compliance, and delay colonic transit. [281], [282], [283] and [284] In small, placebo-controlled trials, ondansetron and granisetron provided relief of loose, frequent stools and flatulence. [279], [280] Large, 12-week placebo-controlled trials of alosetron reported reductions in discomfort and improvements in stool frequency, consistency, and urgency in nonconstipated patients with IBS. [285], [286], [287] and [288] A follow-up 48-week study confirmed the long-term efficacy of alosetron. [289] For unclear reasons, women with IBS derived greater benefit than men. An additional trial reported superior responses to alosetron than the antispasmodic agent mebeverine. [290] Subsequent investigations reported improvements in quality of life, improving physical, social, and mental parameters. [291] However in postrelease surveillance, 70 cases of severe constipation or

ischemic colitis were observed including 10 cases that required surgery and 3 deaths that could potentially be attributed to the drug. [292] As a consequence, the medication was voluntarily withdrawn by the manufacturer, but has since been reapproved for limited use. Preliminary studies in nonconstipated patients with IBS of a newer 5-HT $_3$ receptor antagonist cilansetron have shown improvements similar to alosetron in reduction in abdominal pain and diarrhea. [293] Follow-up investigations will determine if side effects might undermine the use of this agent. In addition to its effects on the 5-HT $_3$ receptor, serotonin modulates gut perception via action on 5-HT $_4$ receptors. In a controlled trial in 18 patients with diarrhea-predominant IBS, the 5-HT $_4$ receptor antagonist SB-207266-A slowed orocecal transit and tended to decrease rectal sensitivity. [294] These effects were associated with symptomatic improvement.

Other Medication Treatments Prokinetic medications, which stimulate gastrointestinal motility, have been proposed for constipation-predominant IBS. In placebo-controlled trials, the 5-HT $_4$ receptor agonist cisapride produced modestly increased stool frequency, improved stool consistency, reduced straining, and acceleration of whole gut transit. [295], [296] This drug was withdrawn from the general market in 2000 because of the risk of cardiac dysrhythmias and is currently available only to restricted patient populations. Recent pharmaceutical research has focused on novel 5-HT $_4$ receptor agonists which exhibit prokinetic activity by stimulating peristalsis (Fig. 86-7). In patients with IBS with constipation, tegaserod accelerated small intestinal and ascending colon transit. [297] In animal studies, tegaserod stimulates peristalsis, accelerates transit, and blunts afferent responses to distention. [298] Clinical trials have reported reductions in discomfort and improvements in constipation and bloating compared to placebo. [298], [299] A second investigational 5-HT $_4$ receptor agonist, prucalopride, was shown to accelerate gastric, small intestinal, and colonic transit and improve bowel function in patients with idiopathic slow transit constipation. [300] Studies are ongoing to assess the side-effect profiles of these agents. The peripheral dopamine receptor antagonist domperidone enhances gastric emptying and small intestinal transit but has little prokinetic activity in the colon and may blunt the postprandial gastrocolonic response. [301] One placebo-controlled trial reported improvements in flatulence, abdominal pain, and bowel dysfunction on domperidone while two other studies observed no benefits. [302]

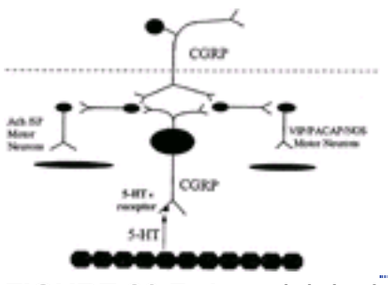

FIGURE 86-7. A model depicting the role of serotonin (5-HT) pathways in the mediation of peristalsis is shown. An extrinsic pathway activated by muscle stretch consists of calcitonin gene-related peptide (CGRP) neurons projecting to the spinal cord while an intrinsic pathway activated by mucosal stimulation consists of CGRP neurons within the enteric nervous system. Serotonin is released by mucosal enterochromaffin cells and acts on 5-HT $_4$ receptors in humans. The orad contractile arm of peristalsis is mediated by acetylcholine (*Ach*) and substance P (*SP*), whereas caudad relaxations are mediated by vasoactive intestinal polypeptide (*VIP*), pituitary adenylate cyclase activating peptide (*PACAP*), and nitric oxide synthase (*NOS*). (From Grider JR, Foxx-Orenstein AE, Jin JG. 5-Hydroxytryptamine $_4$ receptor agonists initiate the peristaltic reflex in human, rat, and guinea pig intestine. Gastroenterology. 1998;115:370.)

Because symptoms are often associated with menstrual cycling, investigators have searched for hormonal treatments for IBS. Continuous gonadotropin-releasing hormone (GnRH) administration reduces synthesis of the gonadotropins, follicle-stimulating hormone, and luteinizing hormone, with inhibition of cyclic gonadal hormone variations. Small trials of the GnRH analog leuprolide in patients with functional gastrointestinal symptoms reported improvements in nausea, abdominal pain, early satiety, bloating, and altered bowel habits for up to one year. [303], [304] A large multicenter trial of leuprolide reported improvements in abdominal pain, nausea, altered bowel habits, and quality of life in functional bowel disease patients. [305] The mechanisms of action of leuprolide are uncertain. In an immunosuppressed patient with intestinal pseudoobstruction, leuprolide evoked intense, propagative upper gut motor activity. [306] It is unlikely that this action is responsible for the clinical benefits of the drug in IBS as most patients with IBS do not exhibit pronounced dysmotility. As it induces amenorrhea and osteoporosis, leuprolide is unlikely to achieve widespread long-term use in IBS. Several agents have been promoted to reduce gas and bloating. Anticholinergics blunt the gastrocolonic response to a meal and reduce the increase in flatus caused by ingestion of beans. [226] Uncontrolled studies using silicone agents such as simethicone show reduction in excess bloating and flatus. [307] Activated charcoal has a large surface area to mass ratio (450–1800 m ^{2}/g) which allows it to be an excellent adsorbent of gas. [308] Small doses of activated charcoal decrease the number of flatus passages after ingestion of beans in healthy volunteers. The microbial a-galactosidase agent Beano reduces hydrogen production after black bean ingestion by healthy volunteers. In a controlled trial in healthy volunteers, pancreatic enzymes reduced bloating, gas, and fullness after eating high-fat, high-calorie meals compared to placebo. [309] No trials of Beano or pancreatic enzymes have been reported in IBS. In one study, the nonabsorbable antibiotic rifamaxin reduced hydrogen production, flatus emissions, and abdominal distention to greater degrees than activated charcoal in healthy volunteers and patients with functional bowel disease. [310] Mast cell degranulation inhibitors have been proposed as treatments for diarrhea-predominant patients by investigators who postulate a role for food allergy in IBS. One study evaluated 101 diarrhea-predominant patients with IBS with evidence of food hypersensitivity on the basis of exclusion diets. [311] Of those with positive skin prick tests for food allergy, 67% reported improved symptoms on sodium cromoglycate while only 41% with negative skin tests experienced relief. In a second uncontrolled report, sodium cromoglycate produced improvement in 67% of diarrhea-predominant patients with IBS when given for 1 month, especially in those with food allergy. [312] The response to sodium cromoglycate may be more impressive in pediatric populations with IBS symptoms where true food intolerance may be more prevalent. A multicenter trial reported that 97% of children (mean age 4 years) exhibited a clinical response to the drug over a one-month period. [313] These results, however, require confirmation in placebo-controlled fashion. Tranquilizers have been used for decades to treat IBS. Older studies reported benefits for phenaglycodol, meprobamate, heteronium plus amobarbital, propantheline plus phenobarbital, chlordiazepoxide, diazepam, medazepam, and alprazolam; however, many of these studies were small or poorly designed. [314], [315] and [316] Buspirone increases colonic compliance, however, its efficacy in reducing symptoms has not been convincingly demonstrated in IBS. [317] Coupled with their abuse potential and development of tolerance, the use of sedatives is not indicated in most patients with IBS.

Potential Future Therapies Investigations have been focusing on medications that blunt visceral hyperalgesia in IBS. Such “anti-afferent” agents might act though one or more mechanisms including (a) modification of release of pain-inducing mediators in the gut wall, (b) blockade or activation of peripheral afferent nerve receptors, (c) inhibition of afferent nerve transmission, or (d) modification of afferent activity in the central nervous system. [105] Some currently available drugs including calcium channel antagonists and opiate agents reduce visceral perception in association with reducing gut motor function or altering central nervous system processing of pain. [318] Kappa-opioid compounds inhibit somatic pain via action on peripheral sensory nerve endings. [319] In patients with IBS, the ?-opioid analog fedotozine increases the thresholds to colonic distention for first perception and pain without affecting compliance. [320] It also modifies neural reflexes in experimental gut inflammation, chemically induced peritonitis, and postoperative ileus. [321] In a double-blind, placebo-controlled, multicenter trial in 238 patients with IBS, fedotozine reduced pain and overall disease severity over a 6-week period. [322] Alpha $_2$-adrenoceptor agonists also have been proposed to reduce perception of visceral pain. In healthy volunteers, the a $_2$-adrenoceptor agonist clonidine increased colonic and rectal compliance and decreased tone, urgency, and pain during balloon distention. [323] This suggests possible use for this class of drugs in pain-predominant IBS. Conversely, the a $_2$-adrenoceptor antagonist yohimbine increased tone and perception of colorectal distention. Another a $_2$-agonist lidamidine reduced perception of rectal distention compared to placebo in 50 patients with IBS. [324] This drug has been reported to have benefit in treating patients with IBS. [325] Substance P has been postulated to participate in the mediation of visceral pain via activation of neurokinin receptors. In animal models, neurokinin-1 (NK $_1$) receptor antagonists have decreased stress-induced defecation as well as reduced the colonic response to the central nervous system stress mediator CRH. [326] Preliminary studies of the NK $_1$ receptor antagonist CJ-11,974 have reported benefits in reducing symptoms in patients with IBS with trends to reductions in visceral sensitivity. [327] Physiological studies suggest that other agents may have visceral analgesic effects. The somatostatin analog octreotide reduces perception of rectal distention via inhibition of visceral afferent pathways in healthy volunteers. [328] Patients with IBS with fecal urgency were shown to exhibit a similar response to octreotide. [329] Additionally, octreotide reduced the increased pressures that resulted from rectal distention in these patients. In a second study, octreotide blunted perception of rectal distention without altering luminal compliance in an IBS population with more constipation-predominant patients. [330] Subsequent work using cortical and spinal evoked potentials showed that octreotide specifically inhibits spinal afferent transmission from the rectum, documenting a peripheral site of action for the somatostatin analog. [331] Finally, intravenous oxytocin increases the thresholds for perception of colonic distention via opiate-independent pathways without affecting luminal compliance. [332] To date, no placebo-controlled trials of the clinical efficacy of these agents have been performed, thus their use in IBS is unproved.

Psychological Therapy

Psychological therapy has been most commonly employed for patients with IBS with psychosocial features that do not respond to standard therapies. A systematic

review of controlled psychological treatments in the literature from 1966 to 1994 uncovered only 14 studies that exhibited appropriate study design. [333] Eight studies reported that psychological treatment was superior to control treatment while six observed no benefits. Because of methodological inadequacies, it was believed that the efficacy of psychological therapy of IBS was not established and that further trials were needed. Nonetheless, many investigators have proposed a range of treatments for IBS including psychotherapy, cognitive therapy, biofeedback, and hypnosis.

Psychotherapy has shown promise in patients who do not respond to medications. In 101 patients with IBS, 3 months of dynamically oriented psychotherapy plus medications provided greater improvements in somatic symptoms than medical treatment alone. [334] A second psychotherapy study of medically refractory patients reported reductions in abdominal pain and diarrhea, but not constipation, as well as improvement in depression. [335] A long-term follow-up of patients undergoing psychotherapy showed maintenance of symptom control at one year, while individuals who declined psychotherapy had relapsed. Group psychotherapy, including techniques such as psychodrama, in which the patient involves other group members in his or her bowel symptoms, does not reduce abdominal pain or improve bowel habits although anxiety levels may decrease. [336]

Cognitive therapy produces improvements in bowel symptoms in some patients with IBS. When compared with daily symptom monitoring, cognitive therapy produced clinical improvement in 80% of patients versus 10% of the symptom-monitoring group. [337] Similarly, patients with IBS undergoing cognitive therapy experienced greater reductions in bowel symptoms than did patients assigned to a self-help support group or to a symptom-monitoring wait list. [338] The benefits of cognitive therapy have been reported to persist in long-term follow-up after completion of the treatment. [339] A recent trial compared therapeutic responses on standard medical therapy to standard medical therapy plus a multicomponent program consisting of IBS education, muscle relaxation, cognitive coping strategies, and assertiveness training. [340] The combination therapy group reported superior reductions in symptoms and improved quality of life, although perception of rectal distention was unaffected by either therapy. Another study suggested that listening to a therapeutic audiotape on IBS provided results nearly as good as those achieved with hypnotherapy. [341]

Biofeedback and stress reduction techniques have been employed by some clinicians. Muscle relaxation training and stress-reduction education reduced bowel symptoms in an uncontrolled study, especially in patients who exhibited improved depression and anxiety. [342] When followed up at 4 years, 50% reported sustained improvements in pain, diarrhea, nausea, and flatulence. Similarly in another study, the frequency and severity of painful IBS attacks were decreased compared to antispasmodic drugs. [343] Additionally, relaxation treatment was shown to reduce primary gastrointestinal symptoms to a greater degree than symptom monitoring. [344] However, other studies have shown no improvement in bowel symptoms. A 3-month relaxation-response meditation program produced significant improvements in flatulence, belching, bloating, and diarrhea. [345] In a novel study, patients were trained to reduce symptoms by listening to their bowel sound pattern through a stethoscope and attempting to modify their borborygmus. [346] This bowel sound biofeedback led to improved symptomatology in three of five patients enrolled, with long-term success in two of the five. In uncontrolled studies computerized biofeedback video games have been found to reduce stress and improve relaxation in significant fractions of patients with IBS. [347]

Hypnosis has been proposed for refractory cases of IBS. In a study of 30 patients with IBS, hypnosis provided better improvements in abdominal pain, distention, and well being but not bowel pattern after 3 months of treatment compared to psychotherapy. [348] When followed for 18 months after being instructed in autohypnosis, sustained remission was reported by most individuals although patients over the age of 50 or with underlying psychopathology were less likely to respond. Subsequently the authors reported a success rate of 85% in more than 200 patients with IBS treated with hypnosis. Another controlled trial of hypnosis also reported improvements in abdominal pain, constipation, and flatulence. [349] No correlation was observed between initial susceptibility to hypnosis and treatment gain. In addition to improving symptoms of pain, bloating, bowel habit, nausea, and flatulence, a hypnosis trial reported beneficial effects on quality of life measures including physical and psychic well being, mood, and work attitude. [350] Furthermore, patients treated with hypnosis missed less work and visited their physicians less frequently than control patients. An anorectal manometric study of 15 patients with IBS demonstrated reduced perception of rectal distention with hypnosis suggesting that hypnosis may inhibit afferent pathways mediating perception of visceral discomfort. [160]

Alternative Medicine

Because of the lack of universally effective, well-tolerated prescription therapies for IBS, many individuals turn to alternative medicine for symptom relief. Recent investigations have begun to explore the use of such measures in scientifically rigorous fashion. An open trial in a small number of diarrhea-predominant patients with IBS reported therapeutic benefits from arrowroot. [351] Similarly, artichoke leaf extract reduced symptom severity in an open study of patients with IBS. [352] In a placebo-controlled, double-blind trial in 24 patients with IBS with predominant bloating, *Lactobacillus* had no significant effect. [353] Conversely in a study in which freeze dried cultures of *Streptococcus faecalis* were administered in placebo-controlled fashion, 81% of patients with IBS noted an improved sense of well being versus 41% with placebo. [212] In another placebo-controlled study, administration of *Lactobacillus plantarum* produced superior reductions in flatulence in 60 patients with IBS. [354] In a large placebo-controlled, double-blind, multi-armed trial of a number of herbal remedies, abdominal pain, stool pattern disturbances, and quality of life were not better with *Fumaria officinalis Curcuma xanthorrhiza* Ayurvedic, or spagyric remedies. [355] In contrast, a randomized, controlled 16-week trial of traditional Chinese herbal medicine reported benefits in bowel symptoms as well as global well being as rated both by the patients and their gastroenterologists. [356] A small uncontrolled study demonstrated improvements in bloating and overall well being after acupuncture. [357] Although inconclusive, these studies suggest there is a role for further investigation into the usefulness of alternative approaches to treating IBS.

APPROACH TO DIFFERENT SUBSETS OF PATIENTS WITH IBS

The management of the patient with IBS is dependent on the predominant symptom. In the technical review for practice guideline development published by the American Gastroenterological Association, specific therapeutic trials of 3 to 6 weeks' duration were proposed for each symptom subtype prior to embarking on a more extensive diagnostic evaluation (Fig. 86-8). [218]

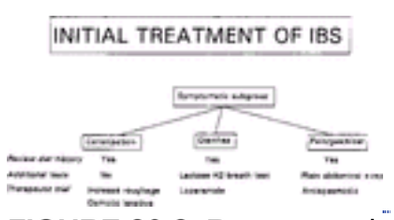

FIGURE 86-8. Recommendations of a technical review for practice guideline development for the initial management of irritable bowel syndrome are shown based on the dominant symptom subtype. (From Hahn B, Watson M, Yan S, et al. Irritable bowel syndrome symptom patterns; frequency, duration and severity. Dig Dis Sci 1998;43:2715.)

Constipation-Predominant Patient

The management of the constipation-predominant patient with IBS involves measures to increase stool water and bulk and reduce the effort of defecation. The most widely used and efficacious compounds for this purpose are bulking agents in concert with a high-fiber diet. Patients should be cautioned that fiber supplements may take several weeks to produce satisfactory results and should be introduced gradually to prevent excess gas. Additionally, any medications that slow colonic transit should be discontinued. Patients with an inadequate response should be given an osmotic agent such as milk of magnesia or an isotonic solution containing polyethylene glycol in addition to the fiber program.

In patients with refractory constipation, further diagnostic testing may be needed to rule out other functional or structural diseases. Using radiopaque markers or colonic scintigraphy, generalized delays in colonic transit indicate the presence of slow transit constipation (also known as colonic inertia) while localized delays suggest functional outlet obstruction in the distal colon. Defecography (cinefluoroscopy of the defecation process) can identify puborectalis muscle dysfunction, rectocele, or rectal prolapse as independent causes of constipation. In IBS, defecography studies have shown an inability to widen the rectoanal angle as well as impaired perineal descent during simulated defecation indicative of diffuse alterations in pelvic floor mobility. [358] Anorectal manometry, in addition to demonstrating sensory abnormalities, can screen for Hirschsprung's disease. These techniques are useful for detecting patients who might be better treated surgically or with a

biofeedback program of bowel retraining. Laboratory studies can screen for porphyria or lead toxicity.

Diarrhea-Predominant Patient

Controlling diarrhea in IBS centers on reducing defecation frequency and urgency and improving stool consistency. Opiate agents such as loperamide are most effective as initial therapy of diarrhea in IBS. Individuals with prominent meal-related symptoms may achieve benefit from antispasmodic agents in the anticholinergic and calcium channel antagonist classes. Tricyclic antidepressants and bile acid binding agents may also be useful in some diarrhea-prone patients. A careful dietary history should screen for foods containing lactose, sorbitol, or fructose. Other dietary manipulations usually are without benefit and are not encouraged in most patients. Use of sodium cromoglycate for food hypersensitivity is unsubstantiated in most diarrhea-prone patients.

Patients not responsive to these therapies may be considered for additional testing. Duodenal aspiration is rarely required to exclude giardiasis. Endomysial antibodies can screen for celiac sprue. Small intestinal biopsy and barium radiography may be performed to rule out Crohn's disease or celiac sprue. Stool volume and fat quantitation can assist in the diagnosis of secretory processes or malabsorption. Laxative screens are indicated for the evaluation of refractory or atypical symptoms.

Painful IBS

Controlling abdominal pain in IBS can be challenging. Narcotics should be avoided because of the risks of tolerance and dependence. Antispasmodic drugs are the preferred initial agents for pain-predominant IBS. Antidepressant medications may benefit an additional subset of patients with more severe symptoms. The GnRH analog leuprolide can be considered for rare patients with refractory symptoms, especially those prominently associated with menstrual cycling.

Further evaluation to investigate the cause of pain in medication-refractory cases should be directed at objective historical and examination findings. Upper endoscopy may detect acid-peptic injury in cases of severe dyspepsia or heartburn. Liver and pancreatic chemistries and imaging tests, such as abdominal ultrasonography or computed tomography, are obtained if hepatobiliary or pancreatic disease is suspected. Small intestinal barium radiography can detect atypical presentations of Crohn's disease, while gastric scintigraphy or gastrointestinal manometry may be obtained in the patient with associated nausea and vomiting. Exclusion of intestinal bacterial overgrowth may be obtained with hydrogen breath testing. Rare patients will need laboratory testing to exclude lead toxicity or porphyria.

PATIENT OUTCOME

Most long-term studies of IBS report that symptoms persist for more than 5 years in greater than 75% of patients despite appropriate therapy. Only 34 of 103 patients were symptom-free on long-term follow-up in an older study. [359] A second investigation followed 50 patients with IBS at 2-month intervals for 12 to 31 months. [360] In 44 patients, the qualitative nature of the symptoms did not change with long-term follow-up although symptom severity was variable. A follow-up of 4581 individuals with IBS reported that only 5% were symptom-free after 5 years. [361] This lack of significant improvement in patients with IBS may not hold true for all populations with IBS symptoms. In a study of elderly Danes, 50% to 79% of subjects suffering from IBS on initial inquiry no longer experienced symptoms on 5-year follow-up. [362]

Even though symptoms persist in many patients with IBS, their quality of life can be enhanced by appropriate physician intervention. In one study, 18 of 44 patients noted reduced symptom intensity after 12 to 31 months of continued medical follow-up. [360] Of 34 fully employed patients, 20 lost no work in the first year after diagnosis. Only 7 patients lost more than 2 weeks because of their symptoms, as a consequence of improved coping skills. A second investigation of 43 patients with IBS reported that abdominal pain and the overall sense of well being were reduced after 5 years of follow-up while other symptoms were unchanged. [363] Those who improved exhibited reduced anxiety compared to those who did not. Symptom resolution was observed in 20% of 97 patients with IBS who were studied for 5 to 7 years on a treatment program of antispasmodic drugs, bulking agents, and high-fiber diets and significant symptom reduction was reported in an additional 42% of individuals. [359] Patients who were male, had a short history of symptoms, had a history of recent acute onset of symptoms, exhibited predominant constipation, and noted a good initial response to treatment were most likely to achieve good long-term improvement. Other studies have found that reductions in anxiety and fear of cancer correlate with improvement over a 6-month period. [364] In the longest follow-up reported to date (median 29 years), fewer return visits to physicians were reported for patients who had undergone inquiry about psychosocial issues and precipitating factors and who had been given detailed discussions about their diagnosis and treatment (Fig. 86-9). [220] These studies suggest that counseling, reassurance, and education provided by the physician, judicious use of medications, and continued interest in a patient's well being may promote successful outcomes in many patients with IBS.

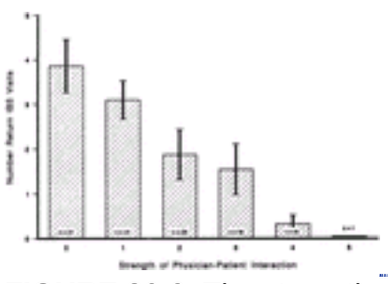

FIGURE 86-9. The strength of the physician-patient interaction is correlated with the number of return visits noted for patients with irritable bowel syndrome (IBS) in follow-up. In general, those patients who experienced a better interaction with their physicians required fewer return visits for care of IBS. Those physicians who documented that they took careful histories and provided reassurance and discussion about IBS did not need to see these patients as frequently in follow-up. (From Caballero-Plasencia AM, Sofos-Kontoyannic S, Valenzuela-Barranco M, Martin-Ruiz JL, Casado-Cabillero FJ, Lopez-Manac JG. Irritable bowel syndrome in patients with dyspepsia: a community-based study in southern Europe. Eur J Gastroenterol Hepatol 1999;11:517.)

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CHAPTER 87

Tonia M. Young-Fadok and Michael G. Sarr

DIVERTICULAR DISEASE OF THE COLON

A diverticulum (plural: diverticul *a*) is a saclike protrusion of the colonic wall. The spectrum of diverticular disease encompasses diverticulosis, diverticulitis, and diverticular bleeding. *Diverticulosis* is merely the presence of diverticula. *Diverticulitis* refers to symptomatic inflammation of a diverticulum. “Complicated” diverticulitis refers to a diverticular abscess, fistula, small bowel or colonic obstruction, or free intraperitoneal perforation; “simple” diverticulitis is the absence of these complications.

This chapter initially discusses the common epidemiology, etiopathogenesis, pathophysiology, and natural history of diverticular diseases of the colon. A detailed discussion of the clinical presentation, diagnosis, and treatment follows, concentrating on the specific diverticular diseases and addressing special situations dictating a more individualized approach. Attempts have been made to concentrate on recent literature and technology whenever appropriate and to identify and discuss controversies.

EPIDEMIOLOGY

The epidemiology of diverticular disease of the colon has changed markedly during the twentieth century. Diverticular disease was rarely encountered at the beginning of the 20th century. Indeed, the first reported surgical resection for complicated diverticulitis was performed by W. J. Mayo and colleagues in 1907. [1] The apparent prevalence of diverticulosis has increased from 5% to 10% in 1918 [2] to 35% to 50% in an autopsy series from 1969; [3] there are no good recent, population-based studies. Prevalence of colonic diverticula is well known to increase with age from about 5% at 40 years of age, to 30% at age 60, and to 65% by age 85. [4], [5]

The predominance of males reported in the early series has changed more recently to either equal distribution or to a slight female preponderance. [4] The gender distribution appears also to vary with age. In one series, 60% of symptomatic patients were women, [6] but, in patients under 50 years, men predominated with a male-to-female ratio of 2.2:1. In several other reports, male preponderance has also been noted in patients less than 40 years of age. [7], [8]

Geographic variations also exist both in the prevalence and anatomic pattern of diverticulosis. Westernized countries have prevalence rates of 5% to 45%, depending on the method of diagnosis and age of the population. [3], [9] Diverticular disease in these countries is predominantly left-sided, with right-sided diverticulitis present in only about 2% of symptomatic patients. [10] This distribution differs markedly in Africa and Asia, [2], [11] where the prevalence of diverticulosis is less than 0.2%, and diverticulitis is usually right-sided. With Japan and Singapore adopting a Western lifestyle, these cultures have experienced an increase in the prevalence of diverticulosis. [12], [13] A review of 13,947 barium enema examinations performed over a 15-year period in Tokyo revealed a steady increase in the incidence of diverticulosis of the right colon. [14] Somewhat surprisingly it remains right-sided; [3] moreover, 70% of symptomatic patients are less than 40 years old. [13] In Hong Kong, diverticulosis involves the right colon in 76% of patients, [15] and right-sided diverticulitis accounts for 17% of patients with acute diverticulitis, [16] a much greater percentage than in Western cultures.

ETIOPATHOGENESIS

Painter and Burkitt [5], [17] first speculated that low dietary fiber was a major factor predisposing to development of diverticulosis. They recognized that where those who were native to Africa had a diet high in dietary fiber and produced a large volume of daily stool output, diverticulitis was distinctly unusual. This concept, however, remains controversial. [11], [18], [19] One study of a cohort of 47,000 men provided strong evidence for a primary role of lack of fiber in the etiopathogenesis of left-sided diverticulosis; [20] a diet low in fiber content increased the risk of symptomatic diverticular disease. Diverticular disease is also less common in vegetarians than nonvegetarians. [21] Of related interest, a low-fiber diet may be a common etiologic factor in the development of both diverticulosis and colon cancer. In one report of 7000 patients, an excess number of colon and rectal cancers was noted in the first 2 years after diagnosis of diverticular disease, [22] an incidence not explained solely by investigations performed for the evaluation of diverticular disease.

Other dietary factors have been examined. There is no substantially increased risk associated with smoking or with caffeine or alcohol intake. [23] The recognized association of acute diverticulitis with obesity in men under 40 years of age agrees with observations that a high intake of total fat or red meat plus a diet low in fiber particularly augments the risk of symptomatic diverticular disease. [20] Whether fat and red meat are independent risk factors or merely markers of a lower fiber diet remains speculative.

PATHOPHYSIOLOGY

Pathology

The typical colonic diverticulum is a false or pulsion diverticulum; that is, it does not contain all layers of the wall as in a true (congenital) diverticulum. Mucosa and submucosa herniate through the muscle layer of the colon, separated from the intraperitoneal space only by the serosa. Diverticula develop at the four constant points of the colonic circumference where the vasa recta penetrate the circular muscle layer. [24] These vessels enter the wall of the colon on each side of the mesenteric tenia and on the mesenteric border of the two antimesenteric teniae (Fig. 87-1). Of special note, diverticula do not develop in the rectum, presumably related both to the coalescence of teniae into a continuous longitudinal muscle layer and the absence of associated potentially weakened areas in the rectum.

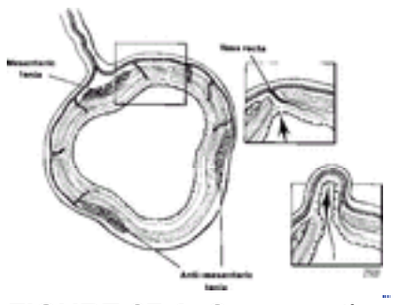

FIGURE 87-1. Cross-section of the sigmoid colon. The main illustration indicates the points of penetration of the vasa recta around the bowel circumference. **Inset:** The development of a diverticulum at one such point of weakness. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: epidemiology and pathophysiology. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Diverticula are distributed unevenly throughout the colon. In Western cultures, 95% of patients with diverticula have sigmoid diverticula. Diverticula are limited to the sigmoid colon in 65% of patients, whereas, in 24%, diverticula involve predominantly the sigmoid but with other segments of the colon involved to a lesser degree; in 7% of patients the distribution is equally divided throughout the colon, while in only 4% diverticula are limited to a segment proximal to the sigmoid. [25] Similarly, 95% of all operations for diverticular disease are directed at the sigmoid colon. [6]

Pathophysiology of Diverticular Disease

Diverticulosis The points at which diverticula develop, that is, where the vasa recta penetrate the colon wall, [26] are considered to be areas of potential weakness within the wall of the colon. Most patients with sigmoid diverticula exhibit grossly evident myochosis, that is, thickening of the circular muscle layer, shortening of the teniae, and lumenal narrowing. While there is no apparent hypertrophy or hyperplasia of the muscular layer of the colonic wall, there is increased deposition of elastin in the teniae. [27] Structural changes in collagen content of the wall of the sigmoid colon are similar to those resulting from aging but are greater in magnitude [28] and may decrease resistance of susceptible segments of the wall to abnormal increases in intralumenal pressure. Similar changes in patients with connective tissue disorders, such as the Ehlers-Danlos and Marfan syndromes, may explain the appearance of diverticula at an early age. Application of the principles of the law of LaPlace may explain, in part, the development of colonic diverticula. The law states that pressure (*P*) is proportional to wall tension (*T*) and inversely proportional to the radius (*R*) of the colon—that is, $P = k\ T/R$ (where k is a conversion factor). Normally intralumenal pressure is the same throughout the colon. However, segmentation of the colon (a motility process in which proximal and distal segmental muscular contractions separate the lumen into isolated chambers) is believed to be exaggerated in diverticulosis. Strong occlusive contractions at both ends of the chamber lead to marked segmental increases in intralumenal pressures (Fig. 87-2), [29] which may predispose to herniation of mucosal diverticula at the sites where the blood vessels penetrate the muscular layers of the colon. As the sigmoid colon is the segment of the colon with the smallest diameter, elevation of intralumenal pressure will result in even greater increases in wall tension compared to larger segments of the colon. Whether the thickened teniae of the sigmoid colon are a result of the environmental milieu such as a lower fiber diet and less bulky stools (and a resultant less wide colonic diameter), a response to increased intralumenal pressure, a primary genetic variation in muscle structure and function, or an acquired abnormality from some other as yet undefined cause remains unknown.

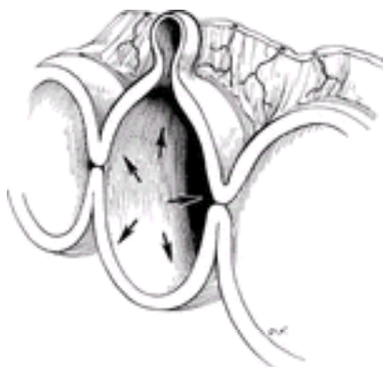

FIGURE 87-2. Painter and Burkitt's concept of segmentation causing formation of pulsion diverticula. (From Pemberton JH, Armstrong DN, Dietzen CD. Diverticulitis. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995:1870.)

Diverticulitis Inflammation of the wall of the diverticulum and the surrounding peridiverticular tissue has a range of clinical manifestations from subclinical inflammation to generalized peritonitis. The underlying cause of the inflammation must involve a micro- or macroscopic perforation of this thin-walled diverticulum. It was previously believed that obstruction of the ostia of the diverticula, for example, by fecaliths or lumenal material, led to increased intradiverticular pressure and subsequent perforation; this hypothesis is now thought to be rare. [11] Increased intralumenal pressure in the involved segment of the colon secondary to abnormal motor activity and/or local trauma of inspissated food particles may erode the wall of the diverticulum. Inflammation and focal necrosis result in perforation. Frequently, a small perforation becomes walled off and contained by pericolic fat and mesentery, with a localized abscess resulting, or with resolution without further consequence. In contrast, with a more extensive inflammatory reaction, surrounding hollow organs adhere to the inflamed segment of colon and intestinal obstruction or an internal fistula may develop. Poor containment, as may occur in antimesenteric diverticula, results in free intraperitoneal perforation and peritonitis. In summary, diverticula occur at weakened points in the intra-abdominal, teniae-bearing colon where intramural vessels penetrate the muscular layer. Development of diverticula appears to be multifactorial. Localized increases in intralumenal pressure may be caused by abnormal motor patterns that predispose to exaggerated segmentation and are likely exacerbated by a low-fiber diet. Morphologic changes in the muscle and collagen content of the bowel wall lead to decreased tensile strength of the wall; these changes are similar to, but more pronounced than those that occur with age. Local trauma to the wall of the diverticulum may produce inflammation, resulting in either localized perforation with peridiverticulitis or free intraperitoneal perforation with generalized peritonitis. Similarly, local trauma to the vasa recta within the diverticula can predispose to hemorrhage.

NATURAL HISTORY

Understanding the natural history of diverticular disease is critical in determining which patients will develop complications if left untreated (and who are therefore appropriate candidates for operative intervention) and those who are best served by a more conservative approach.

Diverticulosis

Of patients with significant diverticulosis, 70% remain asymptomatic and never develop complications related to their diverticulosis. With long-term follow-up, however, as many as 15% to 25% of patients with diverticulosis will develop acute diverticulitis, and 5% to 15% will exhibit diverticular bleeding (Fig. 87-3). As the majority of patients remain asymptomatic, the mere presence of multiple diverticula does not indicate the need for intervention other than possibly recommending a high-fiber diet in an attempt to decrease the incidence of complications.

FIGURE 87-3. The natural history of diverticulosis. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: natural history, clinical features and diagnosis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Diverticulitis

Diverticulitis may present with manifestations ranging from mild abdominal discomfort to perforated diverticulitis with fecal peritonitis, sepsis, and even death. Diverticulitis can be divided into simple and complicated presentations (Fig. 87-4). Complicated diverticulitis describes localized or free intraperitoneal perforation, obstruction, abscess, or fistula, which arise in 25% of patients with their first episode of acute diverticulitis; nearly all of these patients will eventually require operative treatment. Simple diverticulitis accounts for 75% of episodes and refers to the absence of complications; the majority of these patients respond rapidly to medical therapy.

FIGURE 87-4. The natural history of diverticulitis. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: natural history, clinical features and diagnosis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Simple Diverticulitis Most patients with simple diverticulitis are initially treated conservatively (see Fig. 87-4); 85% respond rapidly, but 15% will require operative intervention. [30] Long-term follow-up of patients after their first episode of acute diverticulitis suggests a readmission rate of 2% per patient year; [30] thus, elective colectomy is not necessary for all patients who respond to medical therapy. Indeed, after successful conservative therapy of the first attack of diverticulitis, 30% to 40% of patients remain asymptomatic, 30% to 40% have episodic abdominal cramps without frank diverticulitis, and about one third proceed to a second attack of acute diverticulitis. [4] Older data suggest that the prognosis is worse with a second attack because the rate of complicated diverticulitis approached 60% and the mortality rate doubled. [4], [31] Only 10% of these patients remained asymptomatic after nonoperative resolution of a second attack of acute diverticulitis. These older studies form the basis for the current recommendation that operation should be recommended after two confirmed attacks of diverticulitis, with one episode of sufficient severity to merit hospitalization. In 2001 Chautems and colleagues [32] reported on 110 patients treated conservatively for a first episode of diverticulitis. Of 83 in whom complete follow-up was achieved, 15 patients (18%) recurred once and only 6 (7%) recurred twice or more. Nine of these patients required inpatient therapy. Two patients underwent operation, and no emergency procedure was required. The authors concluded that recurrent diverticulitis may be treated in a similar fashion to a first episode and that medical therapy remains a valuable alternative to surgery. This study was useful in having a long median follow-up of 10.5 years, but the conclusions were hampered by a high death rate of 34% from unrelated causes during follow-up.

Complicated Diverticulitis Most patients with complicated diverticulitis require operative intervention (see Fig. 87-4). With the first attack of acute diverticulitis, 15% to 29% of patients [30], [33] require operative treatment, most of whom have complicated diverticulitis. The mortality rate is 1% to 5%. [30] Indications for emergency or early operative intervention are generalized peritonitis, nonresolving intestinal obstruction, abscess not amenable to radiologically guided percutaneous drainage, clinical deterioration, or failure to improve with appropriate conservative management (Table 87-1). Indications for elective operative intervention are recurrent or intractable symptoms, persistent mass, inability to exclude carcinoma, functional colonic obstruction, presence of a fistula, and previous percutaneous drainage of an abscess. Several unique situations are discussed below.

ABSOLUTE	RELATIVE
Complications of diverticulitis	
Peritonitis	**Symptomatic stricture**
Abscess (failed percutaneous drainage)	**Immunosuppression**
Fistula	**Right-sided diverticulitis**
Obstruction	**? Young patient**
Clinical deterioration/failure to improve	
Recurrent episodes	
Intractable symptoms	
Inability to exclude carcinoma	

TABLE 87-1 Indications for Operative Treatment of Acute Diverticulitis

Those who are treated operatively are generally believed cured, even though the standard extent of resection may leave diverticula in the remaining proximal colon. Diverticulosis progresses in the remaining colon in about 15% of patients, [34] and future operative treatment is required in as many as 2% to 11%. [34], [35] and [36] Up to 27% of patients after a sigmoid colectomy may describe abdominal pain postoperatively in the same location; these persistent symptoms, however, may be secondary to coexistent irritable bowel syndrome rather than to recurrent diverticulitis.

CLINICAL PRESENTATION AND MANAGEMENT OF DIVERTICULAR DISEASES

Diverticulosis

Diverticulosis in the majority of patients is asymptomatic. Some patients with uncomplicated diverticulosis have symptoms of cramping, bloating, flatulence, and irregular defecation; it is unclear if these symptoms are related directly to the underlying diverticulosis or to the coexistence of irritable bowel disease. Because older patients with this symptom complex are investigated with a contrast enema and the prevalence of diverticulosis in the elderly is so common, this ostensible association of symptoms and diverticulosis has often been made, but a true causal relationship is unproven.

Diverticulitis

Presenting Symptoms Constant, noncolicky, left lower quadrant pain is the most common complaint occurring in 70% of patients. The pain has often been present for several days prior to presentation, assisting differentiation from other causes of an acute abdomen; only 17% of patients have symptoms of less than 24 hours. [6] When carefully questioned, almost half the patients describe previous episodes of similar, though less intense, pain. Nausea and vomiting occur in 20% to 60%, diarrhea coexists in one fourth to one third of patients, [37] and recent obstipation has been present in half. Urinary symptoms, such as dysuria, urgency, and frequency, coexist in 10% to 15%.

Presenting Signs Physical examination reveals abdominal tenderness, usually in the left lower quadrant. A tender mass is palpable in 20%. [4] Right lower quadrant tenderness may result from a redundant, inflamed sigmoid colon lying to the right of the midline or, less frequently, from right colonic diverticulitis. Generalized peritoneal signs suggest free perforation and peritonitis. Abdominal distention occurs in two thirds of patients. Low-grade fever and mild leukocytosis are common but not always present; 14% of patients have no fever, and 45% have a normal white count. [38] Urinalysis may reveal a sterile pyuria apparently secondary to adjacent inflammation; mixed colonic flora, fecaluria, or pneumaturia strongly suggest a colovesical fistula. Similarly, fecal discharge from the vagina is pathognomonic of a colovaginal fistula.

Diagnosis The diagnosis of diverticulitis should be suspected and made primarily on the basis of the history and physical examination. In the acute stage, when complicated diverticulitis is suspected, imaging studies can confirm the diagnosis and exclude other sources of acute abdominal signs; in contrast, elective evaluation

after resolution of an acute episode of diverticulitis should evaluate and visualize the whole colon.

Diagnosis in the acute setting. Abdominal radiographs are useful and appropriate in excluding other causes of acute abdominal pain, rather than in making the diagnosis of diverticulitis. In addition, the presence of free air on the abdominal radiograph signifies an abdominal emergency and obligates further evaluation. The diagnosis of acute diverticulitis can frequently be made on the basis of the history and physical examination alone. Published guidelines have recommended that when the clinical picture is clear additional tests are not necessary to make a diagnosis. [39], [40] and [41] This may, however, occasionally, be a disservice to the patient. A clinical diagnosis alone may be incorrect in up to one third of patients. [42] In addition, should there be additional unconfirmed attacks, the patient may potentially be put at risk when surgical management is refused, whereas had there been evidence of an earlier attack, a surgical approach might have been prudent, and the decision to proceed with resection would have been possible. Thus in the patient who has symptoms of sufficient severity to merit hospitalization, our preference is to obtain radiographic confirmation with computed tomography (CT). [43] After an episode of uncomplicated acute diverticulitis, complete evaluation of the colon can be performed electively. The immediate use of CT is not controversial if the diagnosis is not secure or if complicated acute diverticulitis is suspected. CT has become the investigation of choice for diagnosis, recognition of complicated disease (abscess, fistula, obstruction), potential therapeutic intervention, and quantifying resolution of disease. Ambrosetti and colleagues [44] performed a prospective comparison of CT and water-soluble contrast enema in 420 patients over an 11-year period. Among 136 patients that subsequently underwent colonic resection, 132 had histologically proven diverticulitis. CT was more sensitive than contrast enema (98% v. 92%, respectively) and identified all patients who had an associated abscess, while contrast enema only provided indirect evidence of complicated diverticulitis in 29% of these patients. Helical CT with rectal contrast only has a sensitivity of 97% and specificity of 100%, similar to studies using oral contrast. [45] This technique may avoid the risks, discomfort, and costs of oral and intravenous (IV) contrast and allows for immediate scanning. The CT features of acute diverticulitis (Fig. 87-5) include increased soft tissue density in pericolic fat secondary to inflammation in 98% of patients, air-filled diverticula in 84%, bowel wall thickening in 70%, soft tissue masses representing a pericolic phlegmon and pericolic (usually intramesenteric) fluid collections representing abscesses in 35%. [46], [47] In 10% of patients, acute diverticulitis cannot be distinguished from carcinoma, as both may show focal thickening of the bowel wall; features more suggestive of acute diverticulitis are fluid at the base of the mesentery, edematous thickening of the mesentery, and mesenteric vascular engorgement. [48] CT can identify complicated diverticulitis—that is, peritonitis (diffuse inflammatory changes and scattered, loculated fluid collections), fistula formation (usually inferred from air or rectally administered contrast in bladder, vagina, or abdominal wall, rather than direct visualization of the fistulous tract), and obstruction. CT more clearly stages the extent of extralumenal inflammation, which is underestimated by contrast enema. [44], [47] Findings on CT can be classified as mild (localized colonic wall thickening and inflammation of pericolic fat [see Fig. 87-5]) or severe (abscess, extralumenal air- or water-soluble contrast [Fig. 87-6]); the latter findings have been employed as criteria for elective resection in patients responding initially to medical therapy with resolution of acute clinical symptoms. [49] In addition, CT functions as a therapeutic modality when combined with percutaneous drainage of localized abscesses in selected patients; this approach avoids the need for emergent operative intervention and permits an elective, single-stage resection in the future. [50] Contrast radiography or colonoscopy generally should be avoided in patients with suspected acute diverticulitis due to concerns that overzealous infusion of even water-soluble contrast or insufflation of air may lead to rupture of previously contained diverticulitis. However, a single-contrast enema using water-soluble contrast is safe in the acute phase if performed carefully. Pneumoperitoneum or evidence of generalized peritonitis should be absolute contraindications to the use of barium as the contrast agent. [51] Some physicians prefer a water-soluble contrast enema, even in the absence of these findings, in any patient suspected of having acute diverticulitis in the event there is an unsuspected leak (Fig. 87-7). Contrast enema has the advantage of evaluating the colonic lumen, is less expensive and more readily available than CT, and in some practices is the initial procedure of choice; in the patient in whom the diagnosis of uncomplicated acute diverticulitis is secure, however, contrast enema examination adds little therapeutically useful information.

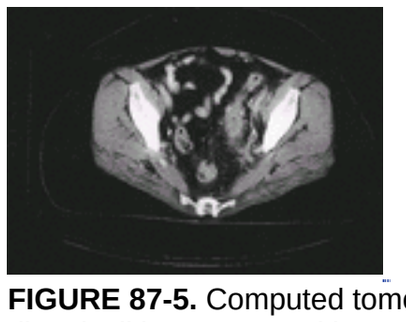

FIGURE 87-5. Computed tomography with oral and intravenous contrast in a 59-year-old woman with increasing abdominal pain for 3 weeks. Mild changes of diverticulitis are seen with narrowing of colonic lumen, bowel wall thickening in the midsigmoid, tissue stranding in pericolic pelvic fat, and diverticula. There is no pericolic fluid collection. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: natural history, clinical features and diagnosis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

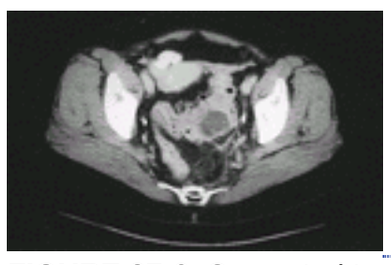

FIGURE 87-6. Computed tomography with oral and intravenous contrast in a 60-year-old woman with a 2-week history of crampy lower abdominal pain, fever, and chills showing a low-density mass adjacent to the sigmoid in the left pelvis. The central, low-density region within the mass, the pockets of surrounding gas, and inflammatory changes in pericolic tissues are consistent with diverticular disease.

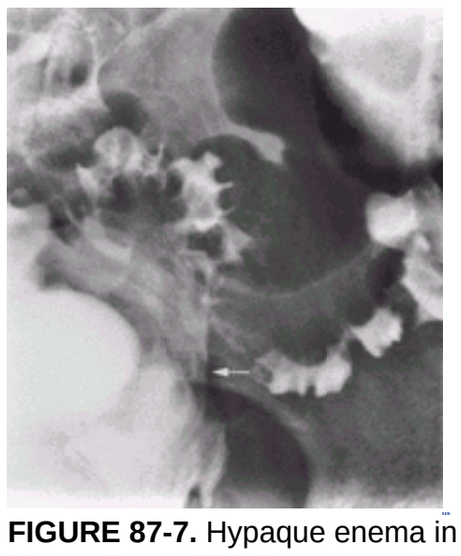

FIGURE 87-7. Hypaque enema in a 79-year-old woman shows nonanatomic distribution of contrast (*arrow*) around the rectum, demonstrating perforation. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: natural history, clinical features and diagnosis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

High-resolution, graded-compression ultrasonography may offer an innovative method to evaluate acute diverticulitis in experienced hands. Visualization of an abnormal colonic segment, as evidenced by mural thickening greater than 4 mm and involving a segment 5 cm or longer at the point of maximal tenderness, is found in 85% of patients and represents the most specific sonographic feature. [52] In cross-section, the thickened colon has the appearance of a target (Fig. 87-8). Inflamed diverticula, mural abscess, extralumenal gas bubbles, peridiverticular abscess, and inflammation may be visualized. Reported sensitivities of ultrasonography range from 85% to 98%, with specificities of 80% to 98%, [52], [53] but undoubtedly are highly operator-dependent. Although ultrasonography is less widely accepted and

validated in the evaluation of diverticulitis than CT, it may be a useful additional tool—for instance in following resolution of an abscess, or in allowing percutaneous, transrectal, or transvaginal drainage of an abscess.

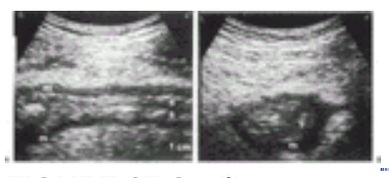

FIGURE 87-8. Ultrasonographic findings of sigmoid diverticulitis. Longitudinal and transverse sections showing thickening of the entire wall and the hypoechoic muscularis propria (*m*), with lumenal narrowing. *s* mucosa/submucosa. (From Schwerk WB, Schwarz S, Rothmund M. Sonography in acute colonic diverticulitis. Dis Colon Rectum 1992;35:1077.)

Diagnosis in the elective setting. After resolution of acute diverticulitis, evaluation requires imaging of the entire colon to determine the extent of disease and to rule out coexistent lesions, such as polyps or carcinoma. Pancolonic imaging may be achieved with colonoscopy, or flexible sigmoidoscopy plus barium enema. An investigational method currently being developed, virtual colonoscopy (also colography or colonography) (Fig. 87-9), may prove to be useful in the elective setting. This technique employs air insufflation of the colon and rapidly acquired CT images, which allow computer generation of a three-dimensional image of the colonic lumen. On occasion, diverticulitis may lead to stricture formation with the characteristic appearances of carcinoma; this finding requires further intervention as for suspected colon cancer.

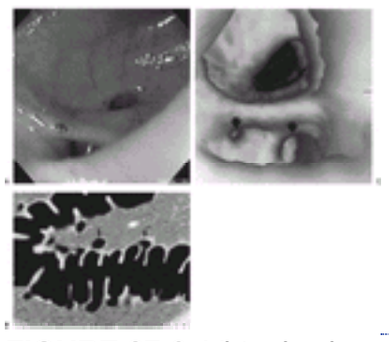

FIGURE 87-9. Virtual colonoscopy. **A:** View of diverticula as seen on routine colonoscopy. **B:** Two-dimensional image of diverticula (*arrows*) obtained by spiral computed tomography scan. **C:** Three-dimensional reconstruction (virtual colonoscopy) illustrating diverticula (*arrows*). (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: natural history, clinical features and diagnosis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Treatment

Decision to observe or operate. Treatment must be individualized for each patient; correct initial therapy depends on clinical acumen (see Table 87-1). The spectrum of disease can be divided into mild symptoms, moderate to severe symptoms, and diffuse peritonitis (Fig. 87-10). The first and last groups require little decision making regarding operative intervention; the second group is the more challenging.

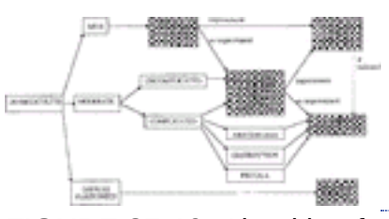

FIGURE 87-10. Algorithm for treatment of acute diverticulitis. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: acute diverticulitis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Mild symptoms. Patients with mild diverticulitis are characterized by left lower quadrant pain, low-grade fever, and minimal physical findings without peritonitis. These patients are best treated as outpatients with a liquid diet and broad-spectrum oral antibiotics such as ciprofloxacin and/or metronidazole. Exceptions include the very elderly or immunosuppressed patients and other high-risk groups, such as diabetics. Patients can be followed for increasing fever, pain, or inability to tolerate oral intake. Hospitalization is only necessary if the patient fails to improve. Diet is reintroduced as symptoms resolve. Once the episode has resolved, the colon should be evaluated 2 to 4 weeks later.

Moderate to severe symptoms. This patient group can be distinguished by the presence of more severe, left-sided abdominal pain, fever, chills, obstipation, and left lower quadrant peritoneal irritation. Classic symptoms do not initially require advanced diagnostic imaging, and medical therapy is instituted on the basis of a clinical diagnosis. [54], [55] When the diagnosis or extent of disease is in doubt, CT or water-soluble contrast enema should then be considered. In stable, immunocompetent patients, initial treatment is conservative with bowel rest, intravenous fluids, and broad-spectrum parenteral antibiotics. Antibiotics need to be active against gram-negative rods and anaerobes; a single antibiotic, active against both aerobes and anaerobes, appears to be as effective as combination therapy (i.e., a second or third generation cephalosporin, or an aminoglycoside combined with an anaerobically active agent such as metronidazole or clindamycin). [41], [56], [57] The aim is to obtain a clinical response to avoid the need for early, nonelective operative intervention. Except for patients with generalized peritonitis, this approach is usually successful. CT may help to identify patients likely to respond to medical therapy. [50], [58] Three outcomes are possible: improvement, failure to improve, or deterioration. Clear improvement should occur within 24 to 48 hours, with a decrease in pain, fever, tenderness, and leukocytosis. [6], [11], [59] Oral intake can be resumed once abdominal tenderness has resolved. Workup is completed 2 to 6 weeks after the acute phase has settled, either with colonoscopy or flexible sigmoidoscopy combined with barium enema. Fiber supplementation of the diet helps to prevent recurrence in 70% of patients. [59] If there is no objective improvement, the diagnosis may be incorrect or complicated diverticulitis is present. CT should be obtained. Abscesses amenable to treatment should be drained percutaneously at the time of the CT. Operative intervention is necessary if there is no reversible condition, if abscesses cannot be drained, or if drainage does not result in improvement. If a patient's condition deteriorates and there is no treatable condition on CT, operative exploration should proceed without waiting.

Peritonitis. Diffuse peritonitis mandates initial resuscitation, broad-spectrum parenteral antibiotics, and operative intervention; diagnostic studies are rarely necessary. Perforated diverticulitis is a serious disease carrying a mortality rate of 6% for purulent peritonitis and 35% for fecal peritonitis. [60]

Which operation—surgical principles. The surgeon's aims are to remove the septic focus by resecting the involved segment of colon, to treat the associated obstruction or fistula, and, if possible, to restore bowel continuity, while minimizing morbidity and mortality.

Preoperative preparation. In nonemergency situations, mechanical bowel preparation with or without oral antibiotics is usually possible and helps to decrease the rate of wound infection. Patients should be warned of the possibility of a stoma and the abdomen marked for optimal stoma placement preoperatively, taking into consideration body habitus, belt line, and so forth. Preoperatively placed ureteral stents are usually unnecessary but may be helpful if there is intense pericolonic inflammation. Positioning the patient in a modified lithotomy position allows a circular stapler to be passed transanally if anastomosis is possible; this positioning also allows intraoperative proctoscopy to assess and empty the rectum if necessary.

Intraoperative assessment. Under emergency conditions, the abdomen is explored through a midline incision; a laparoscopic approach may be possible electively at least 6 weeks after the episode of diverticulitis has resolved. Assessment of peritoneal contamination, proximal obstruction, and fecal load determine the advisability of a primary anastomosis (Hinchey classification; Table 87-2). [61] For instance, class I and II disease may still allow a primary anastomosis, depending on whether the abscess is removed en bloc with the resected segment (as with an intramesenteric abscess), or whether the abscess has been well controlled by the peritoneum, can be safely drained without local contamination, and is distant from the anastomotic site.

STAGE	CHARACTERISTICS
I	Pericolic or mesenteric abscess
II	Walled-off pelvic abscess
III	Generalized purulent peritonitis
IV	Generalized fecal peritonitis

Adapted from ref. 61.

TABLE 87-2 Hinchey Classification of Diverticulitis

In elective or semi-elective situations, resection and primary anastomosis are usually possible, as the inflammatory condition is well localized or has resolved significantly. After resection, the ends of the bowel for anastomosis must be well vascularized, nonedematous, and tension-free. The distal margin of resection should be in the upper third of the rectum, since there are no diverticula distal to this point. The proximal resection should extend to where colon becomes soft and nonedematous. While it is not absolutely necessary to remove all diverticula-bearing colon proximal to the intended anastomosis, [36] attempts should be made to remove the majority of diverticula on the left side of the colon. Absolute contraindications to primary anastomosis are fecal or purulent peritonitis; relative contraindications include severe co-morbid conditions that might jeopardize blood flow, poor nutrition, and immunosuppression. [54] In emergency situations with peritoneal contamination, a two-stage procedure is appropriate (Fig. 87-11). The most common two-stage approach involves resection of the diseased colon, oversewing of the rectal stump, and an end colostomy–a Hartmann procedure. In patients with fecal or purulent peritonitis, this approach is the preferred method. Creation of a mucous fistula from the rectal stump is not possible after resection of the entire sigmoid. Many surgeons mark the rectal stump with a long, nonabsorbable suture and tack it to the anterior abdominal wall or sacral promontory for ease of location in the future. The colostomy can be closed 3 months later.

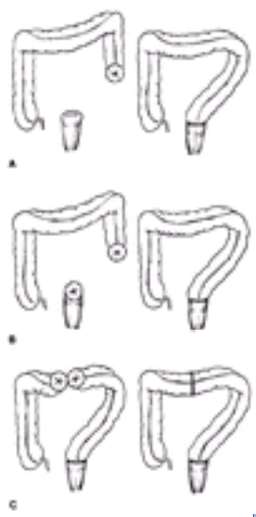

FIGURE 87-11. Two-stage approaches to resection of diverticular disease. Common among the approaches is that the offending segment of diverticular disease is resected at the first operation. **A:** Stage 1: The Hartmann procedure. Diseased sigmoid colon is removed, the fecal stream is diverted, and the rectum is oversewn. Stage 2: Intestinal continuity is reestablished by a descending colorectostomy. **B:** Stage 1: Diseased sigmoid is removed and both ends of bowel are brought to the surface, one as an end colostomy and the other as a mucous fistula. Stage 2: Intestinal continuity is reestablished as in (**A**). **C:** Stage 1: Diseased sigmoid colon is resected and a colorectostomy is constructed and protected by a diverting transverse colostomy or diverting loop ileostomy. Stage 2: Closure of the transverse colostomy is achieved. (From Pemberton JH, Armstrong DN, Dietzen CD. Diverticulitis. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995:1870.)

The Hartmann procedure removes the source of sepsis, but reversal of the colostomy requires a formal celiotomy and is often difficult because of adhesions and difficulty identifying the rectal stump. [6], [11] Up to one third of patients are left with a permanent stoma. [62] Relative contraindications to an unprotected anastomosis include presence of a chronic abscess cavity and mild systemic illness, in which case the patient may be better served by creation of a protected primary anastomosis. This increasingly popular two-stage approach entails resection, primary anastomosis, and proximal diverting stoma; the second stage then involves a simple stoma closure. This approach is employed when there are relative contraindications to primary anastomosis, but no purulent or fecal peritonitis, and the bowel is nonedematous. This option has advantages because the second-stage stoma closure may often be accomplished through the stoma site alone, especially if a diverting loop ileostomy (our preference) is used, thereby not mandating a full celiotomy. Another factor in the emergent setting is that bowel preparation is not possible, and a large fecal load precludes a primary anastomosis. Consideration may be given to intraoperative lavage in selected patients to permit primary resection and anastomosis. In addition to mobilization of the affected sigmoid, mobilization of the splenic and sometimes the hepatic flexure may help. [43] Bowel proximal to the sigmoid is occluded with tape, and immediately above this, the lumen is cannulated with large bore corrugated plastic tubing (anesthesia ventilator tubing). The end of the tube is passed off the field. The cecum, appendix base, or distal ileum is cannulated with a Foley catheter through which warm saline containing 50 mL of povidone iodine solution per liter is infused until the efflux is clear. Lavage was performed in 33 of 62 patients undergoing non-elective operation for diverticulitis: 18 with stage I Hinchey disease, 10 with stage II, and 5 with stage III. [63] Only one anastomotic complication occurred. The outdated classic three-stage procedure (Fig. 87-12) is no longer indicated [55] as it has a mortality rate of 26% versus 7% after a two-stage resection. [60] In rare and exceptional cases, if mesenteric inflammation precludes a safe dissection of the involved segment from the iliac vessels and ureters, or if the patient is unstable, it may be necessary simply to drain the area and create a proximal stoma. In rare circumstances (i.e., areas lacking a well-qualified abdominal surgeon), drainage and diversion are the optimal patient care. This allows for stabilization and early transfer to a large center for definitive therapy.

FIGURE 87-12. Three-stage resection of diverticular disease. **A:** Stage 1: Diverticular abscess is drained and the fecal stream diverted by a double-barreled colostomy. **B:** Stage 2: Sigmoid diverticular disease is resected and a colorectostomy constructed. **C:** Stage 3: The colostomy is closed to restore intestinal continuity. (From Pemberton JH, Armstrong DN, Dietzen CD. Diverticulitis. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995:1870.)

Laparoscopic versus open approaches. Laparoscopic techniques may be valuable in several facets of diverticular disease. [64] These include diagnosis, diversion, resection of the affected segment of colon, and restoration of colonic continuity (either as a staged procedure or primarily). Absolute contraindications to the laparoscopic approach are free perforation with fecal peritonitis or extensive purulent peritonitis because of the limitations on complete exploration and clearance of contaminated material. Both single institution [65], [66] and multicenter studies [67] have demonstrated benefits of the approach in terms of reduced ileus and hospital stay, with comparable complication rates to laparotomy. Randomized trials confirming superiority of the approach, however, have not been performed. The inflammation that may accompany diverticulitis is often challenging, even in open procedures. Resolution of the inflammatory component permits a successful laparoscopic approach in most cases. [64] The presence of a colovesical, colovaginal, or coloenteric fistula reduces the chances of completing the procedure laparoscopically, as does prior use of a drain, but neither are absolute contraindications to this approach. Even in open cases, the bladder side of a colovesical fistula frequently requires nothing more than pinching off the fibrous fistula track and leaving the bladder decompressed with transurethral catheter drainage, and the same principles apply to the laparoscopic approach. Ureteral stents may be helpful, and the firm tubular structure of the stent in the retroperitoneum can usually be detected with the laparoscopic instruments, making lighted stents unnecessary. As with open procedures, it is helpful to approach the mass in the sigmoid from both cephalad and caudad directions, having identified normal tissue planes away from the phlegmon. Significant experience is required to identify these planes. A low threshold for conversion to laparotomy should be maintained to avoid damage to vessels and ureter. The same extent of resection should be accomplished laparoscopically as for an open procedure, namely proximal resection of the sigmoid back to soft, pliable tissue, and distal resection to a point where the tenia have coalesced, usually to a point below the sacral promontory. This thus precludes an extracorporeal anastomosis and mandates an intracorporeal stapled anastomosis.

Complications

Obstruction. Diverticular obstruction of the colon is rarely a true or complete mechanical obstruction. However, obstipation due to functional obstruction is a common presenting complaint. Evaluation often shows a ratty stricture of the involved segment, with a picket fence appearance (Fig. 87-13). The primary concern is differentiation from carcinoma (Fig. 87-14); even with negative biopsies, resection is usually unavoidable for this suspicious appearance. Resection with primary anastomosis is often possible, with colostomy if the preoperative bowel preparation is inadequate. In rare situations, acute diverticulitis leads to complete mechanical large bowel obstruction; this requires an emergent exploration. The inflammatory reaction may also involve secondarily the small bowel, leading to a small bowel obstruction; if this fails to respond to nonoperative treatment, operative intervention may be necessary.

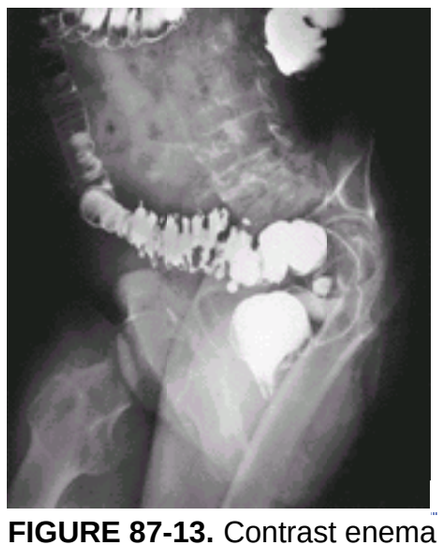

FIGURE 87-13. Contrast enema revealing picket fence appearance of the sigmoid colon, associated with symptoms of obstruction.

FIGURE 87-14. Barium enema demonstrating constricting lesion in sigmoid colon in a region of diverticulosis. There is also a 3-cm localized collection of barium (*arrow*) off the lateral colonic wall consistent with an abscess cavity. The most likely radiologic diagnosis is diverticulitis but carcinoma cannot be excluded.

Perforation. Free intraperitoneal rupture of diverticulitis is unusual but is associated with high mortality rates of 20% to 30% [60], [68] and appears to be more common in severely ill or immunocompromised patients. Treatment involves an emergent two-stage procedure with resection of the involved segment and proximal diversion.

Abscess. Abscesses occur in 15% of patients with acute diverticulitis presenting without peritonitis [49] and in 30% to 56% of patients requiring early operative intervention for diverticulitis. [6] Prior to the introduction of radiologically guided, percutaneous techniques of drainage, abscesses mandated operative intervention, often requiring a two-stage procedure. Percutaneous drainage now permits a future, elective, single-stage operative approach in 60% to 80% of patients; [69], [70] and [71] indeed, in selected patients with severe co-morbid conditions, which contraindicate operative therapy, catheter drainage alone may relieve symptoms and provide definitive treatment. [70] The catheter should remain in place until drainage is less than 10 mL in 24 hours; this approach may take up to 30 days. [70] Contrast radiographs through the drainage catheter (sinograms) can show persistent communication between the colon and the abscess cavity and may be one way to follow resolution of the abscess. The route of drainage is preferably through the anterior abdominal wall; abscesses deep in the pelvis or obscured by other organs may be accessed transgluteally or through the rectum or vagina, [72], [73] but these approaches are less optimal for patient comfort and catheter care. Operative intervention is indicated if improvement does not occur.

Fistula

Incidence. Fistulas account for up to 20% of all indications for operative intervention in complicated diverticular disease. [74] Because 90% of diverticular inflammation involves the sigmoid colon, fistulization most commonly arises from this segment. Colovesical fistulae (Fig. 87-15) comprise 65% of diverticular fistula, 25% are colovaginal fistulae, and less common are coloenteric and colouterine fistulae, [74] but fistulae can form with any hollow organ and have been described in many unexpected sites (Fig. 87-16).

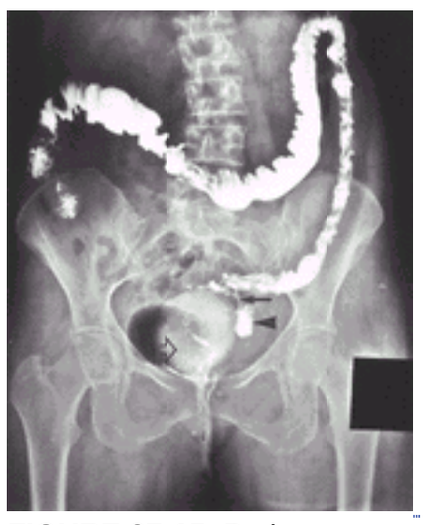

FIGURE 87-15. Barium enema in a 54-year-old woman with pneumaturia and left lower quadrant pain. A fistulous tract (*closed arrow*) arises in the midsigmoid passing into an abscess cavity (*arrowhead*) and then into bladder, where contrast is also seen (*open arrow*). (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: hemorrhage. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

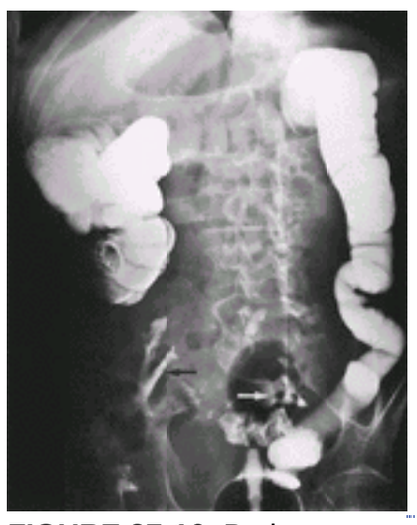

FIGURE 87-16. Barium enema in a 64-year-old man with 3 weeks of right hip pain. There is extravasation of barium from a perforated sigmoid diverticulum with collection of contrast material in an abscess cavity (*white arrow*) adjacent to the diverticulum. A fistulous communication extends from the abscess cavity to the right

lower quadrant and extends to the region of the right hip (*black arrow*). (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: acute diverticulitis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Colovesical fistulae are the most common form of diverticular fistula and, conversely, diverticulitis is the most common cause of colovesical fistula, accounting for 40% to 89% of cases. [75] Although acute diverticulitis has an overall slight female predominance, males comprise the majority (66%–78%) of patients with colovesical fistulae secondary to diverticulitis. [74], [76] The uterus appears to protect the bladder from the inflamed sigmoid; indeed, 50% to 68% of women with colovesical fistulae and 83% of patients with colovaginal fistulae have had a previous hysterectomy. [74], [75], [77]

Clinical features and diagnosis. Colovesical fistulae are illustrative of most forms of diverticular fistulae. Only about 50% of patients will give a history of diverticulitis; in the remainder, diverticulosis is diagnosed when the fistula manifests clinically. [76] Patients describe passage of feculent matter and/or gas during urination; this manifests as pneumaturia in 67% to 75% of patients, dysuria in 56% to 94%, or fecaluria in 50% to 76%. [74], [75] and [76] Crampy abdominal pain, diarrhea, hematuria, and passage of urine through the rectum occur in a much lower percentage of patients. [74], [76] Physical examination is characteristically unremarkable. Urinalysis is invariably abnormal with pyuria or fecal particulate matter; cultures reveal a polymicrobial growth. Such findings suggest a fistula but not the etiology, which may be diverticulitis, Crohn's colitis, or carcinoma. CT, barium enema, colonoscopy, cystoscopy, cystogram, and intravenous urography have all been used for the diagnosis of colovesical fistula. The reported diagnostic yield of each modality varies widely in different reports. The diagnostic yield from contrast enema and colonoscopy tends to be low (20%–26% and 0%–3%, respectively) [75], [76] but these studies are valuable in assessing the proximal colon. Although the actual fistula is rarely demonstrated, abnormalities suggestive of the diagnosis are present in up to 100% of barium enemas and 25% of colonoscopies. If the diagnosis remains questionable after these studies, or if further anatomic delineation is required, CT is the most accurate and sensitive test. Occasionally the fistula itself is opacified, but more commonly colonic thickening adjacent to an area of thickened bladder, associated diverticula, and the presence of rectally administered contrast material or more commonly gas in the bladder (prior to instrumentation of the urinary tract) are present and are highly sensitive and specific for colovesical fistulae. [78] Gas bubbles within organs other than the bowel are highly suggestive of fistulization. [46] Although CT may not actually delineate the fistula tract, associated findings predict the location of the involved segment of colon and provide important information regarding extralumenal inflammation and its relationship to the ureters. Cystoscopy, lauded by some authors, directly identifies only 20% of fistulae but strongly suggests the diagnosis by localizing inflammation and bullous edema of the bladder mucosa in the vast majority (96%) of patients. [75] Others consider such changes too nonspecific and find cystoscopy useful in only 37% to 46% of patients. [74], [76] Similarly, retrograde cystography and intravenous urography objectively demonstrate the fistula in less than 30% of patients and generally are of little clinical benefit.

Treatment. In general, diverticular fistulae do not close spontaneously, but also the presence of a fistula is rarely an indication in itself for urgent operative intervention; indeed, the very rare instance of an acute initial presentation of a fistula may be an indication to postpone operative correction until the acute inflammation inducing the fistula resolves. As most patients present electively and can tolerate a bowel preparation, a one-stage procedure with resection and primary anastomosis is usually feasible and is associated with decreased morbidity and hospital stay. [75] If extensive inflammation is present and operative intervention is required to treat nonresolving acute diverticulitis, a two-stage procedure may be necessary. The portion of the colon from which the fistula arises should be resected as previously described (see " Treatment"). Management of the fistula site at the bladder depends on the size of the fistula, the degree of local inflammation, and the anatomic site. In most patients, the fistula can be pinched off, and either no visible defect in the bladder wall or a very small defect will be evident. Merely leaving an indwelling transurethral bladder catheter for 7 to 10 days with or without closing the bladder defect with a few absorbable sutures over the site or leaving a closed suction drain in the region of the bladder defect is suitable management. With a larger defect, simple closure is almost always preferred to formal resection, which is almost never necessary, as the cause of the fistula originates in the colon, and the bladder itself is normal; any local induration usually settles rapidly once the inciting cause (the diseased segment of colon) has been removed. [54] These simple principles of minimal intervention apply to the distal aspect of most types of diverticular fistula, be they in small bowel, vagina, or uterus. Although operative management is usually recommended, there may be a rationale for conservative nonoperative management in highly selected patients, especially with severe co-morbid conditions, but no good follow-up studies are available to support this approach in most patients. In one series, six patients were managed nonoperatively for 3 to 14 years, treated only with prophylactic antibiotics; there were no complications and specifically no impairment of renal function and no urinary sepsis. [79] In the absence of urinary tract obstruction, observation appears safe in patients with significant contraindications to operative management; this nonoperative approach may, however, be unacceptable if the symptoms are bothersome.

Special Situations

Diverticulitis in the younger patient. Management of patients with their first episode of acute diverticulitis at less than 40 years of age is controversial. The current standard of therapy is to recommend resection after one episode, based on two premises: first, these patients suffer a more virulent disease; and, second, patients responding to medical therapy are much more likely to have recurrent diverticulitis and ultimately require surgical intervention. However, controversy exists as to the true virulence and natural history in this age group. Young patients comprise 12% to 29% of the larger series of patients with acute diverticulitis requiring operative treatment. [7], [8], [33], [54], [80] Common findings are a male predominance of from 2:1 to 4:1 [7], [8], [33], [54] and the coexistence of obesity in 84% to 96% of these patients. [8], [54] The concept that diverticulitis is more virulent in the young arises from follow-up studies of younger patients with their first episode of acute diverticulitis; 48% to 88% of this group will require operative treatment either acutely or in the future. [8], [54], [80] Others describe much lower rates of operative intervention (15%–41%) and a more successful nonoperative management approach. [7], [33] These inconsistencies may arise because the younger patient may undergo operation because of an incorrect diagnosis; [81] some series have quite high rates of incorrect diagnoses approaching 50%. [8], [54] This inordinately high rate is probably because the diagnosis of diverticulitis is not entertained in this young population, and the presence of localized peritonitis is interpreted as an abdominal catastrophe requiring emergent operative exploration. If CT or contrast studies are used more widely, the operative rate in young patients is actually only 15% versus as high as 33% in older patients; [33] however, the diagnosis of acute diverticulitis must first be entertained. The natural history of acute diverticulitis in this young population after initially successful medical management is also disputed. Some studies report readmission rates of 55% [82] and subsequent operative rates of 20% to 41%; [81], [82] and [83] others report no need for operation during a mean follow-up of more than 4 years. [84] When the total operative rates are examined with long-term follow-up, data are more consistent with an ultimate operative risk of about 50%. Some series have very high initial operative rates, [8], [54], [80] while others with a lower rate of acute operation (15%–25%) have higher rates at later times (29%–32%). [33], [83] Although there are no reliable convincing arguments that necessitate elective resection in this younger population after resolution of uncomplicated diverticulitis, in patients with no co-morbid conditions, early elective colectomy after one episode of acute diverticulitis seems the most reasonable recommendation. However, the issue may become one of individual patient evaluation of their risk and lifestyle concerns.

Immunocompromised patient. Pharmacological or functional immunosuppression is associated with an increased risk of perforated diverticulitis. This patient group includes those patients treated with chemotherapy, corticosteroids, and medications such as azathioprine, cyclosporine, and so forth, but the phenomenon is also apparent in diabetics and those with chronic renal failure. These patients may present with minimal symptoms and unimpressive abdominal findings even when they have advanced peritonitis; thus, the diagnosis is frequently delayed. While aggressive medical therapy is successful in 75% of normal patients with acute diverticulitis, operative intervention is necessary in most immunocompromised patients; [85] early operative intervention must be entertained in these patients, especially if they fail to show an objective response early in the course of aggressive medical treatment. Patients with connective tissue disorders, such as lupus, appear to have an increased risk of complicated diverticulitis over and above that related to immunosuppression from use of steroids. [86]

Right-sided diverticulitis. Right-sided diverticula comprise 5% of diverticulosis reported in Western countries [3] but up to 20% in Asian countries. [12], [13] Most of these cases are wide-mouthed, false diverticula. [15] Patients tend to be younger than those with left-sided disease. [12], [13] Because right-sided acute diverticulitis presents clinically as acute appendicitis, the correct preoperative diagnosis is made in only 4% to 16% of patients. If an objective diagnosis can be secured preoperatively, aggressive medical management should be undertaken. More commonly, the diagnosis is made or suspected only intraoperatively. If there is no peritoneal contamination, an obviously normal or absent appendix, and no worry of colonic malignancy, appropriate management involves parenteral antibiotics and an appendectomy if the appendix is present. [12], [13] If the diverticulitis is limited, consideration can be given to simple diverticulectomy [13] (and appendectomy). However, if there is diverticular perforation, extensive inflammation, or a colonic malignancy that cannot be confidently excluded, some form of right colectomy is justified. Primary anastomosis is usually possible, but one should not hesitate to construct an ileostomy and distal mucous fistula in the presence of severe peritonitis or in the compromised host. [12], [16]

Diverticular Bleeding

Incidence Diverticular bleeding is the most common cause of acute massive colonic blood loss, accounting for 30% to 50% of massive bleeding; [87], [88] although angiodysplasia makes up an additional 20% to 30%, [88] it is a more common cause of chronic or intermittent low-grade, lower gastrointestinal bleeding in patients over 65 years old. [89], [90]

Pathophysiology One explanation of the pathophysiology of diverticular bleeding is the hypothesis that as a diverticulum herniates, the vessel responsible for the weakness in the wall of the colon at that point becomes draped over the dome of the diverticulum, separated from the bowel lumen only by mucosa (see Fig. 87-1). With time, the artery is exposed to repeated local injury along its lumenal aspect secondary to inflammation and/or increased intralumenal pressure. Eccentric intimal thickening and thinning of the media occur in the vasa recta along the side adjacent to the lumen. [26] These localized changes in vessel wall structure may result in segmental weakness of the artery and rupture into the lumen. Right-sided diverticula tend to have wider ostia and domes than those in the left colon, possibly exposing vasa recta to injury over a greater length of the vessel wall and hence accounting for the higher incidence of hemorrhage recognized from right-sided colonic diverticula. [26] Others believe that the thinner wall of the right colon predisposes to diverticular bleeding. [31]

Natural History Bleeding will develop in 15% of patients with significant diverticulosis; in a third of these patients (5%) it is massive and can potentially lead to cardiovascular instability. [31] Many patients are older adults (mean age, 68–77 years) [87], [88] with co-morbid conditions that contribute to morbidity and mortality rates of 10% to 20%. [88], [91] Several studies have suggested that management with a specialized bleeding team may reduce mortality. [87] Bleeding stops spontaneously during 75% of episodes; a requirement for blood transfusion of less than 4 units in 24 hours is associated with spontaneous cessation of bleeding in 99% of patients. [92] The risk of rebleeding, however, is significant, ranging from 14% to 38%; [87], [92] with a second episode, the risk of further hemorrhage approaches 50%. [92] The right colon is the source of diverticular bleeding in 48% to 90% of these patients [26], [87], [93] despite the fact that 75% of all diverticula are left-sided, and when right-sided diverticula occur, they are usually associated with left-sided diverticula. [11]

Clinical Presentation The hallmark of diverticular bleeding is painless, bright red rectal bleeding. Most episodes are minor (<1 unit of blood), and up to 50% of patients [93] give a history of intermittent passage of maroon or bright red blood. Abdominal pain is usually absent; indeed, it is extremely rare for bleeding to coexist with acute diverticulitis. [26] Physical examination is usually unremarkable, without abdominal tenderness or mass. Less than 5% of patients with bleeding diverticulosis present with massive hemorrhage requiring transfusion: this bleeding is usually not of an acute exsanguinating form but rather is a persistent bleed.

Management Management of massive colonic bleeding has three components: resuscitation; diagnosis and localization of the site of bleeding; and subsequent management. A coordinated approach by gastroenterologists, radiologists, and surgeons, as members of a specialized bleeding team, has been shown to reduce mortality. [94] The American College of Gastroenterology has published practice guidelines addressing lower gastrointestinal bleeding. [95]

Resuscitation and initial assessment. After aggressive fluid resuscitation and cross-matching of blood, blood loss must be estimated, an assessment made of hemodynamic stability, and the source sought. The patient's age is not a great help in diagnosis nor is the physical examination. Rectal examination, anoscopy, and proctoscopy must be part of the initial assessment; massive hemorrhoidal bleeding, though rare, can be easily excluded. If initial assessment cannot determine whether bleeding is proximal, the appropriate diagnostic workup is initiated. If assessment does not indicate if bleeding is proximal or distal to the ligament of Treitz, an extended upper endoscopy should be performed, as massive hemorrhage from the upper gut is statistically more likely than from the lower gastrointestinal tract (82% vs. 18%); [87], [96] if negative, evaluation of the lower gastrointestinal tract should begin.

Diagnostic modalities. The aim of the diagnostic evaluation is twofold: to localize the site of bleeding should operative treatment become necessary; and to determine the cause of bleeding, because treatment of diverticular bleeding differs from that of other causes. Radionuclide "bleeding scan," mesenteric angiography, and colonoscopy have all been employed; the appropriate order in which they are used depends on the rate of bleeding and on available expertise. A combination of diagnostic modalities can successfully identify the source preoperatively in 90% of patients with massive bleeding. [97]

Colonoscopy. The role of colonoscopy in acute diverticular bleeding is evolving, but in some centers it has become the preferred initial approach. The first report of endoscopic management of a bleeding diverticulum, with irrigation of 1:1000 epinephrine, was in 1985. [98] Success has since been reported with the heater probe, [99] bicap probe, [100] injection therapies, [101], [102] and fibrin sealant. [103] Endoscopic "stigmata" with prognostic value have been reported. [104] Cumulative results of small studies suggest a 95% hemostasis rate. [105] Some proceed with emergent colonoscopy in unprepared bowel as blood is cathartic. [106] Others use enemas or rapid preparation with balanced electrolyte solutions given orally or through nasogastric tube. [107], [108] When performed successfully (i.e., satisfactory visualization of the mucosal surface), colonoscopy has a diagnostic accuracy of 72% to 86%. [106], [107] and [108] Advantages include precise localization and the potential for therapeutic intervention. Only a select few patients, however, are realistically amenable to colonoscopy, and include those with low volume, intermittent colonic bleeding.

Radionuclide imaging. Two types of radionuclide bleeding scans have been employed. The first uses a sulfur colloid labeled with technetium 99m (^{99m}Tc). Scans obtained shortly after intravenous injection can image the area of bleeding, even if the rate of bleeding is as low as 0.1 mL/min. This method is exceedingly sensitive. The short half-life of the marker within the intravascular space (the sulfur colloid is rapidly cleared by the spleen and liver) limits the usefulness of this imaging modality to patients who are actively bleeding during the short duration that the label remains in the vascular space. Because of this exceedingly short functional half-life of efficacy, this imaging modality has been replaced with the so-called tagged red cell scan, which uses ^{99m}Tc pertechnetate to label or "tag" autologous red blood cells removed from the patient prior to reinjection into the patient (Fig. 87-17). Abdominal images are obtained frequently during the first 30 minutes, and then every few hours for up to 24 hours. The major advantage of this label is that patients may be scanned several times over 24 hours thereby imaging the site of intermittent bleeding or localizing the appropriate quadrant of the abdomen.

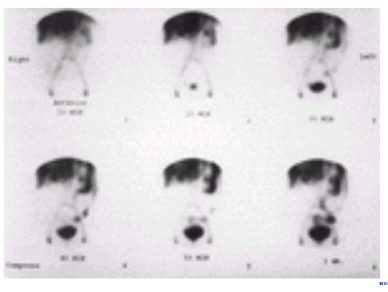

FIGURE 87-17. Technetium 99 (^{99m}Tc)–labeled red blood cell scan for evaluation of lower gastrointestinal bleeding in a 63-year-old woman with a 24-hour history of bright red blood from the rectum. Increased activity in the distal transverse colon near the splenic flexure progresses toward the descending colon and sigmoid colon by termination of the examination at 1 hour. Findings are consistent with a bleeding source in the distal transverse colon near the splenic flexure. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: fistula. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Radionuclide bleeding scans have advantages and disadvantages. Advantages are their noninvasiveness and sensitivity. One series demonstrated a sensitivity of 97%, specificity of 83%, and positive predictive value of 94%. [109] The disadvantages of radionuclide scans are that they localize bleeding to a general area of the abdomen without necessarily localizing the bleeding source to a specific organ, for example, colon versus small bowel, and thus accuracy rates range from 24% to 91%. [31] Poor organ localization occurs because the extravasated blood may move within the hollow organ in either a peristaltic or antiperistaltic direction, and, in addition, localization to an area of the abdomen cannot objectively define a site in the colon; for example, bleeding from a redundant sigmoid colon may appear as extravasation in the right lower quadrant suggesting bleeding either from the right colon or the distal small bowel. Hence, radionuclide imaging is often used prior to angiography to confirm bleeding rates sufficient to justify the use of the more invasive selective angiographic studies.

Angiography. In the absence of a likely source in the distal colon or rectum, the first mesenteric vessel examined is the superior mesenteric artery, because 50% to 80% of diverticular bleeding, and virtually all bleeding from angiodysplasia, occur in segments of the colon and bowel supplied by this artery. [110] If no source is found, the inferior mesenteric and celiac vessels are then studied. Successful localization of the anatomic site of bleeding varies from 14% to 72%, [88], [91], [111] and depends on the rate of bleeding (sensitivities are as low as 0.5 mL/min), timing (the patient must be actively bleeding at the time of the intravenous contrast administration), and the expertise of the angiographer. Angiographic localization is 100% specific, but sensitivity varies with the pattern of bleeding; with acute bleeding, sensitivity approaches 50% but decreases to 30% with a pattern of recurrent hemorrhage. [112] A positive angiogram is associated with an 86% likelihood of operative treatment. [112] The use of radionuclide imaging to screen for active bleeding may reduce the frequency of negative arteriograms, [96] but some groups maintain that the incidence of negative arteriograms is increased by the delay inherent in performing radionuclide scans. [88] Complications of urgent arteriography occur in about 9% of patients and include arterial thrombosis, mesenteric or peripheral embolization of atherosclerotic debris from the aorta (occurring during catheter manipulation), and renal failure. The advantages of angiography are that bowel preparation is not required, and the anatomic localization provided is organ-accurate, especially in the colon. Moreover, angiographic techniques may allow therapeutic intervention with selective infusion of vasoconstrictor agents or vascular embolization. Vasopressin can be infused directly into the artery supplying the segment of colon bleeding by a subselectively positioned intraarterial catheter (Fig. 87-18). Up to 91% of patients receiving intraarterial vasopressin will stop bleeding; however, as many as 50% will rebleed on cessation of vasopressin. [88] In these patients, however, vasopressin may permit resuscitation, stabilization, and bowel preparation with the goal of resection and primary reanastomosis. [88] Transcatheter embolization is a more definitive means of controlling hemorrhage, but it is associated with an incidence of colonic infarction in up to 20% of patients embolized. Attempts to reduce this risk have included use of temporary occluding agents or have employed a "superselective" catheterization technique. [113]

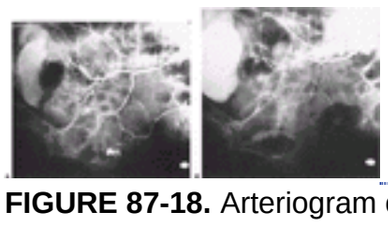

FIGURE 87-18. Arteriogram of the inferior mesenteric artery in a patient with diverticular bleeding. **A:** Extravasation of contrast medium into the lumen of the descending colon. **B:** Arteriogram of the same patient after infusion of vasopressin. Note the markedly reduced flow in the left colonic and sigmoidal branches. (From Pemberton JH, Armstrong DN, Dietzen CD. Diverticulitis. In: Yamada T, ed. Textbook of gastroenterology, 2nd ed. Philadelphia: JB Lippincott, 1995:1870.)

Management approach. During resuscitation, the management rationale is determined by categorizing patients into one of three groups: hemodynamically unstable with active bleeding; continuing bleeding but hemodynamically stable; and hemodynamically stable with apparent cessation of bleeding (Fig. 87-19).

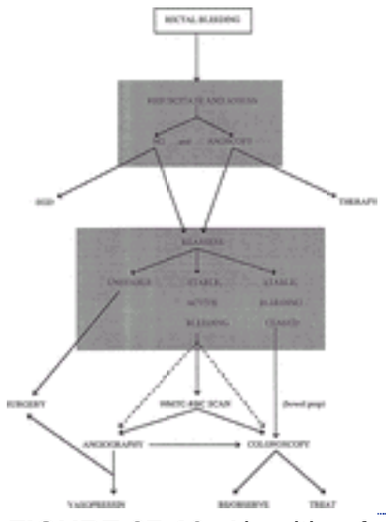

FIGURE 87-19. Algorithm for management of diverticular bleeding. (From Young-Fadok TM, Pemberton JH. Colonic diverticular disease: acute diverticulitis. In: Rose BD, ed. UpToDate in medicine [CD-ROM]. Wellesley, MA: UpToDate, 1997.)

Hemodynamic instability. Persistent instability despite aggressive resuscitation requires emergency operative intervention in 18% to 25% of patients with diverticular bleeding who require blood transfusion. [92], [114] Every attempt should have been made to exclude an upper gastrointestinal source of bleeding (e.g., nasogastric intubation or even emergency gastroscopy), because the operative approach is markedly different for the surgeon. At operation, a careful exploration is undertaken looking for an obvious source or a mass. Because diverticular bleeding is not associated with acute diverticulitis, the involved diverticulum is not immediately evident externally. Indeed, most patients are elderly and would be expected to have colonic diverticula. Without preoperative localization, the decision of what or where to resect becomes problematic. Multiple techniques have been described, including dividing the colon into sections by applying occlusive clamps across the bowel and determining which section fills with blood; this technique, which requires the presence of ongoing massive bleeding, is generally unsatisfactory. A much better approach involves intraoperative colonoscopy, [97] possibly after a rapid on-the-table intraoperative bowel preparation using an infusion catheter placed into the cecum, through which a warmed electrolyte solution is rapidly infused and a transanal catheter to control the efflux of infusate from below. This approach allows the possibility of a localized segmental colectomy involving only the site of bleeding. If, however, no obvious colonic site can be identified, and the bleeding is massive and life threatening, the surgeon may need to perform an abdominal colectomy involving the entire colon susceptible to diverticula and angiodysplasia. Thus, all possible attempts at specific localization preoperatively and intraoperatively are important to limit the extent of colectomy required. A source of bleeding is ultimately identified in 78% of patients undergoing emergent operation for lower gut bleeding. [97]

Hemodynamically stable with continued active bleeding. The most straightforward approach is to start with a tagged red blood cell scan. If positive, it can serve as a useful screen before mesenteric arteriography, which may permit infusion of vasopressin to stop the bleeding and allow bowel preparation for future operative treatment should the bleeding recur. Infusion begins at 0.2 units/min, with repeat angiography after 20 minutes; if bleeding persists the dose of vasopressin is increased to 0.4 to 0.6 units/min. [115] If the bleeding is controlled, the infusion should be continued in an intensive care setting for 24 to 36 hours and then tapered over 24 hours. [115] In the high-risk patient who has failed the initial vasopressin trial but has significant contraindications to operative intervention, embolization may be used followed by close observation for ischemic complications. If angiography is negative, the next step in the diagnostic algorithm (see Fig. 87-19) is colonoscopy, which may identify a source and/or localize the site of active bleeding. Diverticular bleeding is rarely observed at colonoscopy and is often a diagnosis of exclusion. If the radionuclide scan does not demonstrate bleeding, angiography is unlikely to be helpful. Colonoscopy is then performed after bowel preparation. Some institutions proceed directly to angiography, especially for massive bleeding, whereas others use a rapid bowel preparation prior to colonoscopy. Of note is that barium enema (versus water-soluble contrast) has no role in the management of acute colonic bleeding; it prevents angiography because of retained barium.

Hemodynamically stable, bleeding stopped. The patient who has stopped bleeding provides a diagnostic challenge, but also allows a semi-elective evaluation, which involves bowel preparation and colonoscopy in preference to contrast enema.

Operative Treatment The need for operative intervention varies with the degree of bleeding. With severe or massive rectal bleeding, 24% to 78% of patients will require operative intervention. [88], [92], [97], [107] Preoperative localization of the site of bleeding and the use of vasopressin as a temporizing measure have reduced operative morbidity from 37% after emergency resection to 9% after segmental colectomy. The operative mortality of 9% to 11% generally reflects co-morbid conditions. [88], [97], [114] When the source of bleeding has been localized, a segmental colectomy is performed, and the rate of rebleeding is 0% to 14%. [88], [92], [116] Blind segmental resection of the most involved diverticula-bearing segment carries a rebleeding rate as high as 42% [116] and an ultimate morbidity and mortality of 83% and 57%, respectively, and thus is contraindicated. [116] Even in patients with extensive diverticular disease, if the site of bleeding has been localized objectively, a segmental resection eradicating the bleeding site is adequate. [88] Abdominal colectomy is reserved for the patient who continues to bleed without a documented site of bleeding but is associated with a relatively high morbidity and mortality (37% and 11%–33%, respectively). [88], [92], [117] Although the rebleeding rate after abdominal colectomy approaches 0%, [116] abdominal colectomy is associated with relatively disabling diarrhea in the elderly (in about 15%) and socially unsatisfactory diarrhea in a larger percentage. Thus, this extent of operation should not be undertaken lightly.

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CHAPTER 88

Gail Hecht

BACTERIAL INFECTIONS OF THE COLON

Some clinical features of infectious colitis differ from those associated with infection of the small bowel. In general, colonic infections manifest as small volumes of diarrhea, usually less than 1 L/day, whereas small bowel infections induce larger volumes. Lower abdominal pain and tenesmus typify bacterial colitis, whereas central, crampy, abdominal pain suggests small intestinal infection. The presence of gross blood or mucus in the stool is indicative of bacterial or amebic co- litis, findings rarely associated with viral or bacterial infections of the small bowel. Acute inflammatory changes (e.g., erythematous, friable, ulcerated mucosa) associated with co- litis can be visualized by sigmoidoscopy or colonoscopy, but such changes are difficult to see with infection of the small bowel.

Individuals with bacterial colitis who seek medical attention tend to have more severe symptoms, such as bloody diarrhea, fever, and abdominal pain. Pathogens most typically associated with such clinical manifestations include *Campylobacter, Shigella* and *Salmonella* species, *Escherichia coli* O157:H7, and *Entamoeba histolytica.* In the face of this symptom complex, ulcerative colitis and, in older patients, acute ischemic colitis, must also be considered. The nonspecific nature of symptoms associated with infectious colitis requires that stool cultures and examination for parasites be performed to identify a specific pathogen. Colonic biopsies may be necessary to exclude the diagnosis of ulcerative or ischemic colitis.

Considered in this chapter are the five major bacterial pathogens that infect the colon: *Shigella* and *Campylobacter* species, *Clostridium difficile* enteroinvasive and enterohemorrhagic *E coli* and the bacterial and viral pathogens that cause anorectal disease, *Chlamydia trachomatis, Neisseria gonorrhoeae, Treponema pallidum* and herpes simplex virus (HSV) and human papillomavirus (HPV). This group of infections most typically occurs in homosexual men. Additional information regarding infections of the gastrointestinal tract can be found in Chapter 74 and Chapter 75.

SHIGELLA

Classification

The genus *Shigella* originally isolated and described by Shiga in 1898 [1], consists of four species or groups (A through D) of gram-negative, aerobic rods: *S dysenteriae* (group A), *S flexneri* (group B), *S boydii* (group C) and *S sonnei* (group D). Each species, with the exception of *S sonnei*, is divided into several serotypes, defined by the cell wall O antigen. Only *S flexneri* is further divided into subtypes. The distribution of *Shigella* species worldwide correlates with the level of development and hygiene, the basis of which is not understood. *S dysenteriae* a particularly virulent strain associated with significant morbidity and mortality, occurs in less developed countries and has pandemic potential. *S flexneri* is also most commonly isolated in developing countries, especially those with tropical climates. *S sonnei* which causes mild cases of watery diarrhea, accounts for most of the cases of *Shigella* infection in industrialized nations, including the United States and countries of western Europe. *S boydii* is rarely isolated.

Epidemiological Aspects

Shigella whose only reservoir is humans, typically causes bacillary dysentery. Although having a single reservoir should offer an advantage for control of infection, *Shigella* remains one of the most common causes of bloody diarrhea. This dilemma is likely explicable by its exquisitely low infecting dose. Transmission is person to person through contaminated hands, fomites, food, and water. [2] Contributing to its survival is the emergence of antibiotic-resistant strains. About 165 million cases of *Shigella* species infection are reported annually worldwide, [3] 163 million of which occur in developing countries. In the United States, about 15,000 cases are reported annually, [4] although the actual incidence is likely twice this number. An estimated 1.3 million deaths occur each year from *Shigella* species infection. [3] Most episodes of shigellosis occur in children younger than 5 years of age (69%), accounting for 61% of the deaths.

In industrialized countries, shigellosis is primarily a problem in subpopulations exposed to substandard hygienic practices (e.g., day-care centers, custodial institutions, and nursing homes). An outbreak of *S sonnei* infection was reported, however, in association with playing in a neighborhood park recreational spray fountain. [5] *Shigella* is a cause of traveler's diarrhea in 4% of adults returning from foreign travel with diarrhea. [6] Ingestion of contaminated food or water is the most common mode of transmission in developing countries.

Shigella is highly contagious, requiring a very small innoculum to establish infection. Ingestion of as few as 180 organisms induced diarrhea in 22% of healthy volunteers, [2] but ingestion of 10 [7] *Salmonella* species organisms resulted in an infection rate of only 50%. [7] *Shigella* persists as a pathogen because fecal organisms in patients suffering acute shigellosis are abundant (about 10 [9] per gram of stool), and they survive harsh environmental conditions for long periods.

Of greatest concern epidemiologically is *S dysenteriae* type 1, because of the severity of the associated clinical symptoms and potential for serious complications and the propensity of this strain to cause pandemics. In the face of emerging antibiotic resistance, these traits present a major health concern, [8] as highlighted by epidemics of resistant dysentery in Asia and Africa.

Pathogenesis

The pathogenesis of *Shigella* is multifactorial and includes the production of enterotoxins (important but not essential) and the organism's ability to invade and destroy host tissues, resulting in large part in the extensive inflammatory response associated with this infection. *Shigella* lipopolysaccharide may play a role in initiating inflammation by activating the inflammatory-associated transcription factor nuclear factor-?B (NF-?B). [9] About 30 gene products are involved in *Shigella* invasion and its intercellular spread, [10], [11] encoded on large virulence plasmids present in all *Shigella* species. [12] Some of these encoded proteins comprise a type III secretion apparatus, a multiprotein complex that allows direct delivery of bacterial proteins into host cells. Recognition of host cell components by bacterial sensors may trigger the secretion of bacterial factors through this apparatus. A complex series of host cell signals then ensues, ultimately resulting in bacterial invasion and spread. [10], [11] The ability of *Shigella* to invade and colonize the intestinal epithelium is crucial to the establishment of disease and associated morbidity.

Shigella species also elaborate toxins that play a role in disease, including enterotoxins, cytotoxins, and neurotoxins. The classic *Shigella* toxin, Shiga toxin (Stx), is produced by *S dysenteriae* type 1, although other strains likely produce similar toxins. The 70-kd Stx consists of one catalytic A subunit and five B subunits involved in binding the toxin to host receptors, a structure common to many bacterial toxins. The Stx receptor, globotriaosylceramide (Gb_3), is internalized by receptor-mediated endocytosis after toxin binding. [13] A role for this receptor-ligand interaction in *Shigella*-associated disease is suggested by the correlation between the number of receptors per cell and fluid secretion after Stx administration. [14] Once internalized, the catalytic A subunit is released, leading to cytotoxicity through inhibition of protein synthesis. The absorptive capacity of the intestinal epithelium is impaired, thus contributing to diarrhea. Stx does not activate adenylate or guanylate cyclase, as do other bacterial enterotoxins, but it is biologically similar to the Shiga-like toxins secreted by enterohemorrhagic *E coli.* Like those toxins, it can affect renal endothelial cells, resulting in hemolytic uremic syndrome (HUS).

Other *Shigella* enterotoxins have been identified. *Shigella* enterotoxin 1 (ShET1), a 55-kd holotoxin also of the A and B subunit structure, is unique to the *S flexneri* 2a serotype. As for Stx, the gene encoding this protein is also located within the chromosome. The presence of such a toxin was predicted by studies in monkeys showing that small intestinal secretion accounted for the watery diarrhea seen early in shigellosis. [15] ShET2, a 64-kd holotoxin expressed by 80% of diverse *Shigella* serotypes, also contributes to the watery diarrhea associated with infection. Unlike Stx and ShET1, the gene for ShET2 is carried on the large virulence plasmid that confers invasion. [16] Another *Shigella*-secreted protein, designated SigA, with both cytopathic and enterotoxic activity, has been reported. [17] SigA, a 103-kd product encoded within the chromosomal *she* pathogenicity island of *Shigella* has serine protease activity likely accounting for its toxic effects.

Invasion is more important than toxin production in shigellosis. [18] Infection of macaques with isogenic mutant strains of *S dysenteriae* that lacked either the invasion plasmid or the Stx gene showed that the invasive but nontoxigenic strain induced disease undistinguishable from wild-type *Shigella* except that the diarrhea in response to Stx-expressing strains was bloodier. A correlation between the level of Stx production and bleeding has been suggested in humans. [19]

The cardinal feature of *Shigella* is its ability to invade the host intestinal epithelium. This is initially achieved by invasion of M cells, specialized epithelial cells strategically positioned over lymphoid follicles. After entering M cells, the internalized organisms traverse these cells and emerge into the gut-associated lymphoid tissue of the terminal ileum and colon, where they are engulfed by macrophages. In contrast to the usual scenario whereby engulfed organisms are destroyed, *Shigella* organisms induce macrophage apoptosis, [20] releasing interleukin-1ß (IL-1ß), [21] thus recruiting neutrophils to and across the intestinal epithelial layer. Intercellular tight junctions are breached, and lumenally situated *Shigella* directly access the subepithelial space and basolateral pole of the host intestinal epithelial cell, the only membrane domain through which these microbes can invade. [22], [23] *Shigella* organisms disseminate intercellularly throughout the intestinal tissue and destroy the epithelium. This marked inflammatory cell infiltrate and tissue destruction characterize shigellosis.

Clinical Features

Most individuals infected with *Shigella* species have a benign, self-limited course lasting 5 to 7 days in adults and 2 to 3 days in children. Some adults, however, experience a subacute presentation characterized by 2 to 3 weeks of waxing and waning diarrhea. Such a presentation may mimic new-onset ulcerative colitis, especially because the endoscopic appearance of the mucosa is similar. In such cases, it is prudent to perform stool cultures; if *Shigella* is isolated, treatment with appropriate antibiotics should rapidly resolve the symptoms. A long-term carrier state for *Shigella* has been described, defined as fecal shedding after the resolution of symptoms from acute dysentery caused by *S sonnei* or *S flexneri* but this state appears to be rare. [24], [25]

Shigella dysentery is characterized by acute diarrhea containing blood and mucus and accompanied by fever and abdominal pain. The presentation may occur in two phases: the initial one characterized by the abrupt onset of frequent (every 20 to 30 minutes), watery diarrhea, followed by the second, or dysenteric, phase 12 to 72 hours later. In this phase, the passage of small amounts of blood-tinged mucus or blood clots occurs. About half of patients infected with *Shigella* experience bloody diarrhea, but the bleeding may be occult. Onset of fever occurs 1 to 3 days after infection, followed by abdominal cramps and diarrhea within 24 hours. In most patients, the diarrhea resolves after 7 days, although symptoms can persist for up to 30 days. A much more severe clinical course may be seen in children that includes fever, abdominal pain, malaise, vomiting, watery or bloody diarrhea, and tenesmus. [26] Patients experiencing more severe symptoms usually are malnourished or debilitated and without ready access to medical treatment.

The physical examination often reveals only lower abdominal tenderness, and bowel sounds are normal or increased. More alarming physical findings, such as rebound tenderness and ileus, are not typical of *Shigella* infection and should suggest an alternate diagnosis. Dehydration may occur in young children, especially with prolonged (48 hours) diarrhea and vomiting. Leukocytosis is unusual but is more likely to be seen in patients, both adults and children, with severe rectal histology. [27]

Confirmation of *Shigella* infection requires isolation and identification by stool culture. Stool cultures were positive as early as 3 days after oral challenge in volunteer studies and, if untreated, remained positive for an average of 27 days. [28], [29] Interestingly, not all patients with positive stool cultures suffered symptoms. Conversely, some volunteers with classic symptoms of *Shigella* dysentery had negative cultures; thus, a negative stool culture does not exclude the diagnosis. To optimize the isolation of *Shigella*, liquid stool specimens should be collected in a sterile container and inoculated promptly on media selective for *Shigella* and *Salmonella.* The presence of fecal leukocytes and erythrocytes greatly enhances the percentage of stool specimens that are positive for bacterial pathogens. Only 20% of stool cultures from adults with acute diarrhea were positive if performed nonselectively; however, the yield increased to 75% or greater if the specimen contained leukocytes. [30] A report of bloody diarrhea should also prompt a search for *E coli* O157:H7 and an examination of fresh stool to exclude *E histolytica* especially in patients with a history of foreign travel.

Complications

Shigella bacteremia is an uncommon but severe complication of *Shigella* dysentery, occurring most frequently in malnourished children, immunocompromised patients, and patients older than 65 years of age. *S flexneri* caused bacteremia in a middle-aged, immunocompetent women, [31] leading to the speculation that *Shigella* bacteremia may be underestimated because of failure to obtain blood cultures or prior administration of antibiotics. The mortality rate associated with *Shigella* bacteremia is high, about 20%, resulting from renal failure, hemolytic anemia, thrombocytopenia, gastrointestinal bleeding, and shock. [32]

Reiter syndrome, a triad of arthritis, urethritis, and conjunctivitis, most often complicates bacillary dysentery in men aged 20 to 40 years expressing the human leukocyte antigen (HLA)-B27 phenotype. About 1% to 2% of infected individuals develop Reiter syndrome, [25], [33], [34] 80% of whom are HLA-B27 positive. [34] Of the HLA-B27–positive individuals who suffer bacillary dysentery, about 20% develop Reiter syndrome.

The clinical presentation of Reiter syndrome can be quite variable, with mild forms being common [35] and genitourinary symptoms absent or minimal. Patients often present with asymmetric lower extremity arthritis or periarthritis 2 to 4 weeks after shigellosis or other bacterial dysentery. By this time, the intestinal symptoms have usually resolved, and antibiotic treatment is not indicated. The arthritic symptoms tend to be chronic and relapsing, even in the absence of dysentery. Reiter syndrome is therefore treated symptomatically with nonsteroidal antiinflammatory drugs.

HUS, most often associated with infection by enterohemorrhagic *E coli* that produces Shiga-like toxins (Stx1 and Stx2), is also a recognized complication of *S dysenteriae* infection. Shiga toxin and Stx1 of *E coli* are nearly identical structurally. HUS (microangiopathic hemolytic anemia, thrombocytopenia, and renal failure) is a direct result of Stx production. Stx gains access to the circulation and binds to tissues that express the Gb_3 receptor. In humans, renal tissue and endothelial cells

of the microvasculature express the highest concentration of Gb $_3$, accounting for the targeting of these tissues. Pathologically, the kidneys reveal microangiopathic damage and cortical necrosis secondary to thrombosed interlobular arteries. Since the 1960s, the mortality rate associated with HUS has dropped significantly, from 50% to less than 10%. [36] Still, about 30% of HUS survivors are left with permanent disabilities, including chronic renal insufficiency, hypertension, and neurological deficits. [37] Although other Stx-producing organisms cause HUS, infection with *S dysenteriae* type 1 is the most common cause in parts of Africa and Asia. [38], [39]

Treatment

Antibiotic treatment for shigellosis shortens the clinical course of illness and reduces the duration of fecal excretion of the organism. In otherwise healthy individuals, the diarrheal illness is often self-limited, lasting only 5 to 7 days, and therefore does not require antibiotic treatment. Antimicrobial therapy is recommended, particularly for elderly patients, malnourished children, patients with co-morbidities, those infected with the human immunodeficiency virus, health care workers, food handlers, and healthy children who attend day-care centers. In all cases, treatment of dehydration with either oral rehydration solution or intravenous fluids for mild to severe diarrhea is important. Antidiarrheal medications, on the other hand, have been shown to prolong symptoms and delay clearance of the organism and thus are not recommended. [40]

The antimicrobial treatment recommendations for shigellosis are rapidly changing as a result of the emergence of multidrug-resistant strains. Ampicillin had been an acceptable treatment option for shigellosis, but by 1990, [41] 32% of *Shigella* strains were resistant. Surveillance data from the United States in 1985 to 1986 found that only 7% of *Shigella* isolates were resistant to trimethoprim-sulfamethoxazole (TMP-SMX). [41] Therefore, TMP-SMX has been broadly endorsed for the treatment of shigellosis in the United States. [42], [43] and [44] Reexamination of TMP-SMX resistance in *Shigella* isolates in the United States showed that of 369 isolates collected between July 1995 and June 1998, [45] 59% were resistant to TMP-SMX, 63% were ampicillin resistant, 1% were cefixime resistant, and 0.3% were resistant to nalidixic acid. Multidrug resistance was identified in 13% of the isolates, but none of the isolates was resistant to ciprofloxacin. The increase in resistant *Shigella* isolates likely stems from imported cases or the induction of resistance after the introduction of new antibiotics. Based on these findings, the recommen-ded treatment options for shigellosis should be reconsidered (Table 88-1).

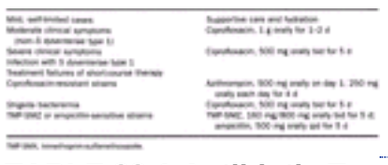

TABLE 88-1 Antibiotic Treatment Regimens for Adults with *Shigella* Infection

Many cases of shigellosis are self-limited and are easily managed with supportive care and hydration. Antimicrobial therapy should be reserved for the most severe cases and those warranting elimination of carriage as a public health priority. Ideally, selection of antibiotic therapy should be dictated by culture and antibiotic susceptibility data, but such testing requires at least 2 days. Therefore, antimicrobial choices are usually made empirically. TMP-SMX and ampicillin can no longer be regarded as appropriate empirical therapy for shigellosis, even in developed countries (see Table 88-1). For adults, quinolones provide effective therapy, [32] and ciprofloxacin, 500 mg orally twice each day for 5 days, led to the resolution or significant symptomatic improvement in 95% of patients with severe disease. [46] For mild infections, shorter courses of therapy may be adequate. Two consecutive 1-g oral doses are effective for strains other than *S dysenteriae* 1. [47] For *S dysenteriae* type 1 infections, however, treatment failures did occur with one- and two-dose regimens but not with the standard course of 500 mg twice per day for 5 days. [47] Azithromycin is an effective alternative to ciprofloxacin for resistant strains. The study from Bangladesh [46] demonstrated that azithromycin, 500 mg orally on day 1 followed by 250 mg per day for 4 days, was as effective (94%) as ciprofloxacin, 500 mg twice a day for 5 days (100%). Although concentrations of both of these drugs was 200 times greater in the stool than the minimum inhibitory concentration (MIC) for *Shigella* only ciprofloxacin reached a sufficient MIC in serum. Thus, ciprofloxacin is recommended in *Shigella* bacteremia. If sensitivity to TMP-SMX or ampicillin is demonstrated, the recommended doses are 160 mg/800 mg orally twice per day for 5 days and 500 mg orally 4 times per day for 5 days, respectively.

In children, the use of quinolones has been limited to compassionate use because of reports of arthropathic effects in the weight-bearing joints of immature animals. [48], [49] Data accumulated from hundreds of children who have received quinolones, however, failed to disclose arthropathies. [50], [51] and [52] A direct comparison of oral ciprofloxacin and intramuscular ceftriaxone as empiric therapy for acute diarrhea in children [53] showed ciprofloxacin to be safe and effective when administered as 10 mg/kg every 12 hours for 3 days. Nalidixic acid is also effective against shigellosis and is approved for use in children older than 3 months of age in the United States, but resistance develops rapidly. [54], [55]

CAMPYLOBACTER

Campylobacter species were not recognized as important human pathogens until the late 1970s. [56] *Campylobacter jejuni* and *Campylobacter coli* have now emerged as the most common bacterial enteropathogens in developed countries, [57] including the United States. [58] The Centers for Disease Control and Prevention (CDC) estimated that *C jejuni* was responsible for nearly 2 million illnesses in the United States in 1999, composing 14.2% of all food-borne infections, making *C jejuni* the leading cause of bacterial diarrhea in the United States, [58], [59] exceeding *Salmonella* species. The actual number of cases is estimated to be 30 to 40 times the reported number. [58]

Of the 18 species and subspecies of *Campylobacter, C jejuni, C coli, C lari, C upsaliensis, C hyointestinalis, C concisus, C mucosalis* and *C fetus* are confirmed pathogens of the human gastrointestinal tract. Of these, *C jejuni, C coli* and *C fetus* are most commonly associated with human disease, although newer diagnostic techniques have identified *C hyointestinalis, C concisus* and *C upsaliensis* at an increasing rate. [59], [60] In clinical laboratories, *Campylobacter* organisms are not routinely identified at the species level.

Campylobacter organisms are long (0.5 to 5.0 µM), gram-negative rods that are curved, S shaped, or spiraled. Isolation of this organism from stool is enhanced by immediate transfer into special transport medium and inoculation onto plating medium. If necessary, samples can be held at 4°C in transport medium for up to 24 hours.

Epidemiology

Foods of animal origin are the most common source of pathogenic *Campylobacter* species infection because the organisms colonize the gastrointestinal tracts of many animals, including chicken, lamb, pig, ox liver, [61] and cows. [62] Meats from retail outlets in the United Kingdom that were screened for the presence of *Campylobacter* revealed that various species were isolated from 73% of samples examined. [61] The highest contamination rate (14% to 98%) was found in chicken. [63], [64] Infection may also be acquired from contaminated water, unpasteurized milk, or direct contact with infected humans or pets. [57] Most cases of *Campylobacter* infection are sporadic and unrelated to outbreaks, making identification of the source difficult. The ingestion of undercooked poultry was responsible for 10% to 50% of *Campylobacter* species infections in case-controlled studies. Freezing diminishes the organism's viability. *Campylobacter* is also a cause of traveler's diarrhea. Seasonal variation of infection occurs in the United States and other temperate countries, with few infections occurring between January and April, a marked increase in May and June, and peak incidence in July and August, followed by a sustained plateau. [57]

Pathogenesis

The pathogenic mechanisms for the symptoms associated with *Campylobacter* infection remain undefined. Four major virulence properties of *C jejuni* and *C coli* have been recognized: motility, adherence, invasion, and toxin production. [65] Motility moves the organism near host cells so that it can attach and invade. Although the role of flagella in invasion is not defined, they are equired for this property. [65] Adherence of *Campylobacter* to host intestinal epithelial cells permits colonization, enhancing the concentration of secreted factors encountered by these cells. Adherence is presumed to be a prerequisite for invasion. The rate of invasion by

Campylobacter species varies considerably [66] but may be as low as 1% of the initial inoculum, [67] and intracellular killing of organisms occurs. [68] Thus, toxin production may be the most important pathogenic factor.

Campylobacter species produce several different toxins, including cytotoxins and possibly an enterotoxin. Controversy surrounds the production of a cholera toxin–like enterotoxin by *Campylobacter* species and its potential role in pathogenesis. [65] Although functional studies suggest that *C jejuni* and *C coli* may express a cholera toxin–like enterotoxin, no genes with cholera toxin similarity were identified on sequencing of the *C jejuni* genome. [69] *Campylobacter* species can produce several distinct cytotoxins, [65] and the best defined is the multisubunit cytolethal distending toxin (CDT). This toxin, clearly an important virulence factor, causes cell cycle arrest leading to a characteristic elongation and swelling of susceptible cells. Although intoxicated cells remain viable for prolonged periods of time (up to 72 hours), they eventually die. One of the subunits, CdtB, possesses DNase activity and induces DNA damage, ultimately leading to cell death. [70], [71] The role of CDT in *Campylobacter* pathogenesis is unclear. Other organisms, including pathogenic *E coli, Shigella* species, and certain *Helicobacter* species also express CDT, but the contribution of CDT to intestinal fluid secretion associated with infection by these strains is variable. *E coli* CDT failed to stimulate intestinal secretion in rabbit ileal loops, [72] but CDT from *S dysenteriae* induced intestinal fluid secretion and tissue damage in the suckling mouse. [73]

Clinical Aspects

The infective dose of *C jejuni* is as little as 500 organisms, [74], [75] suggesting that this pathogen, like *Shigella* species and *E coli* O157:H7, possesses mechanisms of protection against gastric acid. However, suppression of gastric acid by inhibiting the proton pump doubles the risk for *Campylobacter* species infection. [76] The typical course of infection is depicted in Figure 88-1. The incubation period is 18 hours to 8 days (average, 3.2 days). [77] Onset of symptoms may be abrupt, with abdominal cramps followed by diarrhea or a prodrome of fever, headache, malaise, and myalgia. Febrile-associated rigors occur in up to 22% of cases. When this influenza-like prodrome persists for 2 to 3 days in the absence of gastrointestinal symptoms, it predicts a more severe form of the infection. *Campylobacter*-associated diarrhea (up to 10 stools per day) is watery initially and progresses to bloody diarrhea after 1 to 2 days in about 15% of patients. [77] This may indicate extension of infection, with the endoscopic appearance in the colon ranging from mild hyperemia to severe mucosal destruction with friable, edematous, and even ulcerated epithelia. Histologically, acute inflammation predominated by neutrophils and crypt abscesses is seen. The absence of distorted crypts and a normal mucous cell population may aid in differentiation from inflammatory bowel disease (IBD) (Table 88-2). With chronic infection, however, the distinction becomes less clear. Further blurring of the boundary between *Campylobacter* species infection and IBD is the potential for relapse of the *Campylobacter* infection, reported in 15% to 25% of patients, and the possibility that IBD patients may be infected with *C jejuni*.

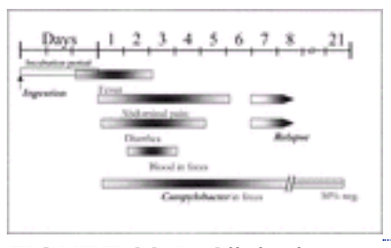

FIGURE 88-1. Clinical course of *Campylobacter* species infection. (Redrawn by V. K. Viswanathan; from Greenwood D, Slack R, Peutherer J, eds. Medical microbiology. 15th ed. 1997, Edinburgh: Churchill Livingstone, 1997.)

TABLE 88-2 Comparison of Histological Features that Help to Differentiate *Campylobacter* and Other Infectious Causes of Proctocolitis from Acute Infammatory Bowel Disease

The abdominal pain associated with *Campylobacter* infection may be sufficiently severe and localized to mimic appendicitis. This is usually caused by terminal ileitis and mesenteric adenitis, but true appendicitis may be present, and *Campylobacter* has been isolated from inflamed appendices. [78] Other symptoms include nausea and vomiting (15% of patients) and significant weight loss of up to 5 kg. Although most patients begin to improve after 3 to 4 days, they may excrete organisms for an extended period (mean, 37.6 days; maximum, 69 days). [79]

Antibodies against *Campylobacter* antigens can be detected in the serum at about day 5, with peak levels appearing within 2 to 4 weeks. [74] Short-term immunity to homologous strains is provided, but the duration and breadth of protection are undefined. The incidence and severity of *Campylobacter* infection decrease in at-risk groups over time, suggesting some level of long-term protection. Anti- *Campylobacter* antibodies have been reported in intestinal secretions. [79]

Complications

Intestinal and extraintestinal complications of *Campylobacter* infection have been reported. Intestinal complications include hemorrhage, [80] ulceration of ileostomy stomas, [81], [82] and toxic megacolon. Extraintestinal complications include erythema nodosum, [83] urticaria, [84] bacteremia, [85] cholecystitis, [86] pancreatitis, [87] abortion, [88] HUS, [89] and nephritis. [90] More commonly occurring complications, however, are reactive arthritis, Reiter syndrome (for which HLA-B27 tissue antigen gives a strong predisposition), and Guillain-Barré syndrome (GBS).

GBS is defined by flaccid paralysis, areflexia, and albuminocytological dissociation in the spinal fluid. It is caused by autoimmune-induced inflammation of peripheral nerves that disrupts their function. GBS is the leading cause of acute paralysis, although its incidence is only 1 to 2 per 100,000. [91] One third of GBS patients have either positive stool cultures or antibodies to *C jejuni* antigens. [92] Serum antibodies to *C jejuni* lipopolysaccharide share antigenic determinants with peripheral and central nervous system gangliosides. Because antiglycoconjugate antibodies are present in GBS patients, molecular mimicry of neural oligosaccharides by microbial pathogens may underlie GBS pathogenesis.

Diagnosis and Treatment

Definitive diagnosis of *Campylobacter* infection can only be made bacteriologically. Because diagnosis is delayed using this approach, rapid detection methods are being developed, including enzyme immunoassays of stool specimens, [93], [94] DNA probes, and polymerase chain reaction (PCR).

Besides oral rehydration, no specific therapy is required for most patients because symptoms are usually resolving by the time a confirmed diagnosis is made. Patients with severe dysentery, relapsing symptoms, systemic infection, or immunosuppression and those who are institutionalized, however, may benefit from antibiotic treatment. *C jejuni* and *C coli* are widely resistant to penicillins, cephalosporins, trimethoprim, sulfamethoxazole, rifampin, and vancomycin. [95] Erythromycin is usually the drug of choice for *Campylobacter* infection when antibiotic therapy is indicated. Resistance rates of *C jejuni* to erythromycin range from 0% to 11% but trend higher (0% to 68%) for *C coli.* In Thailand and Sweden, however, resistance to erythromycin is on the rise. [96], [97] The recommended dose of erythromycin is 500 mg twice daily or 250 mg four times per day for 5 days. Erythromycin stearate, which is acid resistant, stable, and incompletely absorbed, may offer a treatment advantage, but clinical evidence of superiority is lacking. Erythromycin ethyl succinate, 40 mg/kg/day in divided doses for 5 days, is the recommended treatment for children. When resistance or other reasons preclude the use of erythromycin, tetracyclines and chloramphenicol are reasonable alternatives, although the prevalence of tetracycline resistance is about 60%. Resistance rates of *Campylobacter* to fluoroquinolones are in the range of 41% to 88% in Europe and Asia. [98], [99] Although

quinolone resistance in the United States has increased more slowly, it is also on the rise, increasing in Minnesota from 1.3% in 1992 to 10.2% in 1998. [100] Similar rates of resistance have been reported in Canada. [101] This trend likely reflects acquisition of resistant strains from extensive antibiotic use, overseas travel, or poultry treated with fluoroquinolones. Azithromycin, 500 mg daily for 3 days, is an effective alternative in areas where fluoroquinolone resistance is high. [102] For systemic infection, an aminoglycoside, such as gentamicin or imipenem, may be warranted; simultaneous administration of oral erythromycin may aid in eradicating the intestinal infection.

CLOSTRIDIUM DIFFICILE

Although *Clostridium difficile* initially called *Bacillus difficile* because of its difficulty in culturing, was discovered in 1935, [103] it was not until 40 years later that Bartlett and colleagues identified that the cytotoxins of this organism were the cause of antibiotic-associated pseudomembranous colitis. [104] *C difficile* is a gram-positive, anaerobic rod that is fastidious in its vegetative state but sporulates in conditions that fail to support its growth. The ability of *C difficile* to sporulate allows it to persist for long periods in harsh environments, including dry surfaces and soil. This capacity for survival contributes to making *C difficile* the number-one cause of health care–associated diarrhea. [105] In addition, hospitals and long-term health care facilities are often reservoirs of this pathogen because establishment of infection is usually dependent on disruption of the indigenous intestinal microflora by antibiotic therapy. Although this organism is isolated from the stool of less than 3% of healthy individuals, it is found in about 20% of those hospitalized for longer than 1 week. [105], [106] *C difficile* can be cultured from the stool of up to 80% of healthy neonates and infants but fails to produce symptoms, possibly owing to lack of expression of receptors for the toxins (A and B) responsible for tissue damage.

Epidemiology

C difficile is the most common cause of nosocomial diarrhea, with an incidence ranging from 20 to 60 cases per 100,000 patient days. [107], [106] The incidence of hospitalization and death due to diarrhea caused by this organism may be increasing. [108] In contrast, the incidence of community-acquired *C difficile*–associated diarrhea is only 7.7 cases per 100,000 person-years of observation. [109] Although *C difficile*–associated diarrhea is rare in outpatients, this may in part reflect inadequate diagnostic efforts in patients receiving antimicrobials. An educational program that encouraged testing for *C difficile* in outpatients with diarrhea increased the rate of detection from 2.6% to 10.7%. [110]

There are three major risk factors associated with *C difficile* diarrhea. The first is exposure to antimicrobials. Antibiotics disrupt the normal colonic flora, allowing colonization after the patient is exposed to the organism or its spores. Clindamycin is highly associated with *C difficile* disease, and its removal from hospital formularies has controlled outbreaks. [111] Other antibiotics, especially ampicillin and cephalosporins, often predispose to *C difficile* infection, whereas others, such as parenteral aminoglycosides, vancomycin, and metronidazole, are less frequently implicated. For unclear reasons, third-generation cephalosporins are more highly associated with *C difficile* infection than are other broad-spectrum agents. [112]

The second risk factor is hospitalization. There is a direct correlation between length of hospital stay and percentage of patients infected. Among patients hospitalized for longer than 4 weeks, 50% acquired *C difficile.* [113] *C difficile* has been isolated from numerous sites within hospital rooms, including telephones, call buttons, bed rails, toilets, and bathroom floors, and from stethoscopes and the hands of health care workers. [106], [114] Patient-to-patient spread also occurs, particularly if a room is shared with an infected patient. [114]

Although antibiotic administration and hospitalization are essentially required for *C difficile* infection, most patients exposed to these two risk factors do not manifest symptoms. A third, but ill-defined, factor is needed. This may be host immunity because patients with *C difficile* diarrhea, versus colonization, generate lower levels of antibodies against *C difficile* and its toxins. [115] Other possible factors include host susceptibility and strain virulence.

Outbreaks of *C difficile*–associated diarrhea within hospitals are often due to a single strain or related group of strains. Three large outbreaks in geographically diverse areas were attributed to a single strain. [116] Some strains may have disseminated across different countries and continents, as determined by the collaborative international typing study. [117]

Pathogenesis

The pathogenesis of *C difficile* colitis is largely the result of the production and secretion of two potent toxins, A and B. [118], [119] These toxins are high-molecular-weight proteins, 308 and 270 kd, respectively, that enter the intestinal epithelial cells through specific, but as yet unidentified, receptors expressed on the apical surface. Once inside the host cell, these distinct yet highly homologous (49% amino acid identity) toxins exert similar biochemical effects, by inactivating Rho proteins (a family of small guanosine triphosphate–binding proteins) that regulate actin polymerization. Rho inactivation is accomplished by toxin-induced glucosylation of a specific threonine residue of the Rho proteins, such as Rho $_{ABC}$, Rac, and Cdc 42. [120], [121] This modification leads to the depolymerization of actin filaments, cytoskeletal collapse, and the characteristic-cell rounding that is the basis of the diagnostic tissue culture assay for *C difficile* toxin. The physiological effects on the intestinal epithelial cell are an increase in tight junction permeability [122], [123] and fluid secretion. [124], [125] In addition to the direct effects of toxins A and B on intestinal epithelial cells, a profound inflammatory response is elicited, contributing further to disruption of the tight junction barrier and increased lumenal fluid. Although most investigations have focused on toxin A, a study using human tissues demonstrated colonic damage in response to both. [125]

The marked inflammation associated with *C difficile* infection is a multifactorial response. One early event is activation of the inflammation-associated transcription factor NF-?B, which increases expression of many proinflammatory cytokines, including IL-8, [126] tumor necrosis factor-a, prostaglandin E $_2$, and leukotriene B $_4$. [127] Inhibitors of cytokine synthesis attenuate toxin A effects. [127] Toxin A can also cause monocytes to release IL-1ß and IL-6 [128] and neutrophils to be recruited to the intestine. [129] Inhibition of neutrophil infiltration of the intestine by specific antibodies [129], [130] or induction of neutropenia significantly reduced fluid secretion. [131] Inhibition or depletion of mast cells attenuated neutrophil activation and fluid secretion. [132], [133] These effects may follow an initial disruption of the intestinal epithelial barrier, allowing direct interactions between toxin A and the inflammatory cells residing within the lamina propia.

Enteric nerves and neuropeptides are involved in *C difficile*–associated disease. Substance P expression by nerve cells in spinal dorsal root ganglia is immediately up-regulated after exposure of the intestine to toxin A. Substance P binds to neurokinin-1 receptors that are expressed by macrophages, releasing tumor necrosis factor-a, a potent cytokine. [134] Neurokinin-1–deficient mice show a diminished response to intestinal toxin A. [135] Neurotensin (NT) is also involved in *C difficile* colitis. [136] Both NT and substance P receptors, whose expression is NF-?B dependent, are markedly up-regulated within 15 to 30 minutes of toxin A exposure. An NT receptor antagonist reduced the intestinal effects of toxin A, including mucosal mast cell activation. [136]

Immunity

Hospitalized patients receiving antibiotics but not colonized with *C difficile* showed no immune protection against *C difficile* colonization. [115] After colonization, high serum immunoglobulin G (IgG) levels against toxin A strongly correlated with asymptomatic carriage of *C difficile* whereas those with low serum levels of toxin A IgG antibody had a much greater risk for *C difficile* diarrhea. There was an eight-fold increase in the risk for *C difficile* diarrhea in patients with severe disease, defined using a modified Horn's index. [137] These data support the development of passive or active immunization strategies to control nosocomial *C difficile* diarrhea.

Clinical Features

The clinical manifestations of *C difficile* infection are broad, encompassing a carrier state devoid of symptoms, colitis without or with pseudomembranes, and fulminant colitis with megacolon or perforation. Antibiotics commonly cause diarrhea independent of *C difficile* infection. The mechanisms underlying the broad range of symptoms associated with *C difficile* are unclear but are likely attributable to host rather than bacterial factors and include toxin receptor density, status of the resident intestinal flora, and concentration of antibodies against bacterial or toxin antigens. Symptoms resulting from *C difficile* infection typically begin 4 to 9 days after initiation of antibiotic therapy, but diarrhea can develop even after antibiotic treatment has been halted in up to 30% of patients. This delay complicates the diagnosis of *C difficile*–associated diarrhea unless the clinician is aware of this scenario.

Antibiotic-associated, Non– *Clostridium difficile* Diarrhea The occurrence of mild diarrhea during antibiotic therapy is not uncommon, and only a minority (about

20%) [138] of these cases is due to *C difficile* infection. In general, antibiotics interfere with the digestion of carbohydrates by eliminating colonic bacteria that metabolize this energy source. Decreased carbohydrate metabolism can then lead to osmotic diarrhea, diminished production of short-chain fatty acids, and disruption of water and electrolyte absorption. [139] In these cases, evidence of *C difficile* infection is absent, and the colonic mucosa appears normal or mildly hyperemic. Discontinuation of antibiotics resolves the symptoms.

Asymptomatic Carriage of *Clostridium difficile* Although hospitalization is a risk factor for infection with *C difficile* most of these patients are free of symptoms, [106], [114] that is, are asymptomatic carriers. This subset of *C difficile*–infected individuals serves as a reservoir of this organism and thus contaminates the hospital environment. Asymptomatic carriers should not, however, be treated with antibiotics because this may prolong the carrier state.

Non-pseudomembranous *Clostridium difficile* Colitis About half of patients with diarrhea who are confirmed as positive for the *C difficile* organism or toxin have clinical symptoms similar to but less severe than those associated with pseudomembranous colitis. Typical manifestations include abdominal pain, cramps, watery diarrhea, malaise, nausea, and possibly dehydration and fever. Laboratory tests commonly reveal a neutrophilic leukocytosis. In fact, the sudden onset of leukocytosis in hospitalized patients with a history of, or currently receiving, antibiotics should immediately trigger testing for *C difficile* infection. [140] Sigmoidoscopy may show only friable and erythematous mucosa but no pseudomembranes.

Pseudomembranous Colitis Pseudomembranous colitis is the prototypic entity associated with *C difficile* infection. Symptoms, including diarrhea and abdominal pain, usually begin within 1 week of antibiotic administration but may not appear until even 6 weeks after antibiotics have been discontinued. [141], [142] Fever, chills, nausea, vomiting, dehydration, and tenesmus may accompany the diarrhea and lower abdominal cramping. Physical findings are nonspecific and include diffuse abdominal tenderness and distention. Occult blood may be present in the stool, but hematochezia is rare. Fecal leukocytes are detected in about half of patients, but peripheral leukocytosis is common and may serve as a surrogate marker of this infection. [140] Three patterns of leukocytosis have been reported: a sudden increase in the white blood cell (WBC) count correlating with symptoms of *C difficile* co- litis; leukocytosis preceding *C difficile*–associated symptoms; or exacerbation of preexisting leukocytosis plus nonspecific symptoms of *C difficile* disease. [140] In the last two scenarios, the diagnosis of *C difficile* colitis is generally delayed. Consideration of *C difficile* infection as a cause of unexplained leukocytosis is therefore recommended. Although not usually necessary, the diagnosis of pseudomembranous colitis can be confirmed rapidly by sigmoidoscopy. Characteristic yellow-white, raised plaques about 2 to 10 mm in diameter are seen (Fig. 88-2). These small plaques may coalesce and cover the entire mucosa. Microscopic analysis of biopsy samples from pseudomembranes shows the typical ummit or "volcano" lesion in which inflammatory cells, fibrin, and mucus exude from microulcerations in the surface epithelium (see Fig. 88-2). Although sigmoidoscopy usually reveals such findings in up to 30% of patients, pseudomembranes can be restricted to the more proximal colon. [143] Colonoscopy is not recommended, however, unless other diagnostic measures have failed.

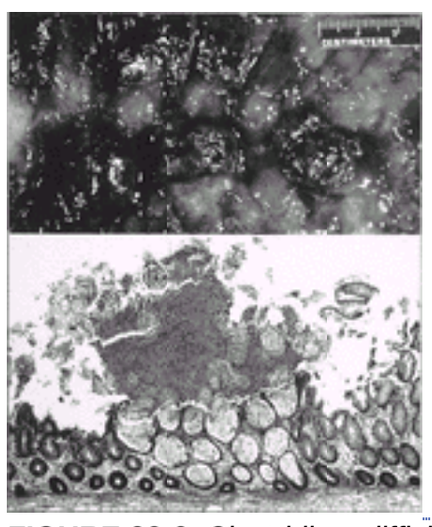

FIGURE 88-2. *Clostridium difficile* pseudomembranous colitis. The gross photo (*upper panel*) shows the severe, nearly confluent light-colored pseudomembranes that are typical of severe *C. difficile* pseudomembranous colitis. The pseudomembranes are composed of purulent debris and contrast sharply against the edematous surrounding dark-colored mucosa. The photomicrograph (*lower panel*) shows a classic type II volcano-like eruption with a mushroom-shaped cloud of adherent of inflammatory exudate. (Courtesy of Dr. Jerrold R. Turner, M.D., Ph.D., The University of Chicago, Chicago, IL).

Fulminant Colitis In fulminant *C difficile* colitis, transmural inflammation can lead to the most serious complications associated with this infection. These include ileus, perforation, megacolon, and even death [144] and occur in about 3% of patients. Fulminant colitis may present acutely or develop over the course of a milder infection. The symptoms are typically more severe with signs of toxemia, constant and localized abdominal pain with guarding, and decreased bowel sounds. Fever, tachycardia, and ascites may also be present. Severe leukocytosis (up to 40,000 WBCs/µL) can occur. Hypoalbuminemia may result from severe protein-losing enteropathy. [138] Diarrhea may actually decrease in the absence of clinical improvement, as a result of acute colonic dilation and paralytic ileus. The diagnosis of toxic megacolon is defined as dilation of the colon to more than 7 cm and severe toxemia, manifested as fever, chills, leukocytosis, and dehydration. A kidney-ureter-bladder radiogram can reveal a dilated colon (Fig. 88-3), and thickening of the colonic wall can be seen by computed tomography. Dilation of the small bowel with air-fluid levels may accompany the large bowel findings, thus mimicking intestinal obstruction or ischemia. [138] In the presence of perforation, abdominal guarding, rebound tenderness, rigidity, and diminished bowel sounds are typically seen. The presentation of acute abdomen in a patient with a history of current or recent antibiotic therapy mandates the urgent exclusion of *C difficile* colitis to prevent unnecessary surgical intervention. [145] In such cases, a limited and cautious endoscopy with minimal air introduction should be considered. The finding of pseudomembranes will establish the diagnosis of *C difficile* colitis. If, however, more severe signs, such as extreme leukocytosis (more than 30,000 to 80,000 WBCs/µL), hypotension, or metabolic acidosis are present, laparotomy and colectomy may be indicated as treatment for impending or existing colonic perforation. [145], [146]

FIGURE 88-3. Abdominal radiogram of a patient with fulminant pseudomembranous colitis and toxic dilation of the transverse colon.

Diagnosis

Unlike the diagnostic strategy for other enteric pathogens, stool culture for *C difficile* is not used because both toxigenic and nontoxigenic strains are detected by culture and high rates of isolation require strict anaerobic precautions. Isolates must then be tested for toxin production, delaying the diagnosis even further. Instead, the gold standard for diagnosing *C difficile* infection is a tissue culture assay for the presence of toxins A and B in the stool. [147], [148] Diarrheal stool is filtered and applied to cultured human fibroblasts that are examined 24 to 48 hours later for cytotoxic effects (cell rounding or detachment). Incubating the stool sample with a neutralizing antitoxin antibody and assessing for protection against cytotoxic changes confers specificity. Test results are reported as positive or negative because titers have no correlation with disease activity. [148] Although this test is highly sensitive (94% to 100%) and specific (99%), [142] it is expensive and is often not performed on site; thus, the incubation period is prolonged. Because both toxins A and B possess cytotoxic activity, this assay detects both. On a molar basis, however, toxin B

is much more potent than toxin A; thus, the stool cytotoxic effect is primarily due to toxin B.

Several enzyme-linked immunosorbent assays are now available for detecting *C difficile* toxins.[149] Although these tests are less sensitive than the cell culture assay, they are equally specific (99%), provide same-day results, and are currently used by most hospital laboratories. Sigmoidoscopy and colonoscopy are not required to diagnose *C difficile* colitis. If a rapid diagnosis is required, such as in an acutely ill patient, or if the diagnosis is in doubt, these tests can be performed, and the presence of pseudomembranes (see Fig. 88-2) essentially establishes the diagnosis of *C difficile* colitis.

Treatment

C difficile–associated colitis is a consequence of antimicrobial therapy; therefore, the first step in treatment is discontinuation of such drugs. This is sufficient to resolve symptoms in 15% to 23% of patients,[150] but those who do not respond require specific antibiotic treatment. Metronidazole and vancomycin are the mainstays of therapy for *C difficile* colitis. Metronidazole is the initial drug of choice because of its lower cost, excellent clinical efficacy, and rising concern regarding glycopeptide resistance in other organisms such as enterococci. The recommended dosage is 250 mg four times per day or 500 mg three times per day for 10 days. The efficacy of this therapy is greater than 95%.[151] Symptomatic improvement is typically seen within 2 to 3 days and resolution by 7 to 10 days.[152] Although metronidazole is absorbed, it accesses the colonic lumen by diffusing from the blood across the inflamed intestinal mucosa where permeability is enhanced. Excretion into bile also contributes to the intestinal concentration.[153] Metronidazole causes a metallic taste, induces alcohol intolerance, and may cause nausea.

Oral vancomycin, 125 mg four times daily for 10 days, was the initial drug of choice for treating *C difficile* colitis and has equal or better efficacy than metronidazole.[154] It is not well absorbed, accounting for high fecal levels and rare side effects. Expense and the emergence of resistant organisms have led to vancomycin being recommended for patients who have failed metronidazole treatment or are intolerant of this drug.[155] For severe or fulminant pseudomembranous colitis, however, initial treatment with vancomycin is recommended. The rationale is the superb delivery of vancomycin to the colon and a slightly, but potentially important, better efficacy in the treatment of pseudomembranous colitis.[152] In the case of ileus or toxic megacolon, however, intravenous metronidazole, 500 mg every 6 hours, should be given either alone or possibly in combination with vancomycin administered by nasogastric tube or rectally. Intravenous vancomycin has no role in the treatment of *C difficile* colitis.

Relapsing *Clostridium difficile* Colitis A minority (5% to 30%) of patients develop recurrent symptoms 1 to 2 weeks after the discontinuation of antibiotics for *C difficile* treatment[151] due either to relapse from the original infecting organism or reinfection by a different strain, especially if the patient remains in the hospital environment. A screen of 93 hospitalized patients with recurrent *C difficile* diarrhea revealed that nearly half (48%) of these cases were due to reinfection with a different strain.[156] Because recurrence of *C difficile*–associated diarrhea is not a result of antimicrobial resistance,[150] the first approach should be to administer another 10- to 14-day course of the antibiotic used initially. If only mild diarrhea is experienced, consideration should be given to symptomatic treatment without antibiotics. For multiple relapses, various treatment options exist (Table 88-3). The first is a 4-week course of either metronidazole or vancomycin or a tapering schedule of vancomycin over a several-week period (see Table 88-3). This regimen was curative in all patients tested, with no relapses seen during a 2- to 12-month follow-up.[157] An anion-binding resin, such as cholestyramine, 4 g given twice daily, which theoretically binds the toxins of *C difficile* may be added to this regimen. Patients must not take cholestyramine and vancomycin simultaneously, but should separate the doses by 3 hours, because the resin can bind and neutralize the antibiotic. The combination of vancomycin, 125 mg four times daily, plus rifampin, 600 mg twice daily, for 1 week has also proved effective for relapses.[158]

First relapse
- Symptomatic treatment only if diarrhea is mild
- 10- to 14-day course of metronidazole or vancomycin if symptoms are more severe or persistent

Second relapse:
- Vancomycin taper
  - 125 mg/6 h for 7 d
  - 125 mg/12 h for 7 d
  - 125 mg/d for 7 d
  - 125 mg every other day for 6 d (i.e., 3 doses)
  - 125 mg every 3 d for 9 d (i.e., 3 doses)

Other reported treatments for multiple relapses:
- Vancomycin in tapering dose as above plus cholestyramine 4 g bid
- Oral *Saccharomyces boulardii* in combination with metronidazole or vancomycin
- Vancomycin, 125 mg qid, and rifampicin, 600 mg bid for 7 d
- Intravenous immunoglobulin.

From ref. 138, with permission.

TABLE 88-3 Suggested Approach to Recurrent *Clostridium diffcile* Diarrhea and Colitis

More novel strategies for the treatment of *C difficile* infection are being developed.[159] *Saccharomyces boulardii* a nonpathogenic yeast, has been used to prevent antibiotic-associated diarrhea.[160] *S boulardii* also decreased significantly the number of relapses in patients treated with high-dose vancomycin (2 g/day), but not 500 mg/day or metronidazole (1 g/day) for 10 days.[161] Other strategies to reestablish normal flora include the administration of *Lactobacillus* species and rectal infusions of feces preceded, or not, by bowel irrigation.[162] Intravenous immunoglobulin and passive oral immunotherapies, such as bovine colostrum from cows immunized with *C difficile*, have produced increased serum antitoxin levels or resolution of symptoms.[163], [164] and [165]

Prevention and Control

Guidelines for the prevention and control of *C difficile* infection have been published by the American College of Gas- troenterology[155] and the Society for Healthcare Epidemiology of America[151] (Table 88-4). Because the most likely method of transfer of *C difficile* to patients is by means of the hands of health care workers, good hand-washing technique is the most important step in decreasing infection. Wearing gloves when handling *C difficile*–infected patients or working in their environment may further reduce transmission. The use of chlorine-containing disinfectants (1:100 dilutions or unbuffered hypochlorite or phosphate-buffered hypochlorite [1600 ppm]) decreases significantly environmental contamination.

TABLE 88-4 Practice Guidelines for the Prevention and Control of *Clostridium difficile* Infection

PATHOGENIC *ESCHERICHIA COLI* THAT CAUSES COLITIS

Of the five major classes of pathogenic *E coli* enterotoxigenic, enteropathogenic, enteroaggregative, enteroinvasive, and enterohemorrhagic,[166] only the last two groups primarily affect the colon. The symptoms associated with infection by these pathogens are similar to those seen in shigellosis. Enteroinvasive *E coli* (EIEC) carries the same large virulence plasmid that confers the invasive phenotype to *Shigella* strains.[167] Enterohemorrhagic *E coli* (EHEC) expresses Shiga-like toxins that are structurally similar to Shiga toxin and whose role in the development of HUS has been clearly established.

Enteroinvasive Escherichia coli

The first report of an invasive strain of *E coli* causing disease was associated with a food-borne outbreak.[168] EIEC is now recognized as a rare cause of traveler's diarrhea, usually transmitted through contaminated food.[169] The primary virulence factor of EIEC is its ability to invade. The genes required for this event are encoded on a large 140-megadaltons (MDa) virulence plasmid, identical to that possessed by *Shigella.*[167] Hence, the stages of EIEC entry into cells are essentially the same as for *Shigella:* attachment to cells, invasion of cells, intracellular multiplication, intercellular spread, and killing of host cells. Because invasion is the major virulence

factor of *Shigella* and EIEC, the associated clinical features closely resemble each other, that is, typical signs and symptoms of dysentery.

Laboratory identification of EIEC, however, is much more difficult than for *Shigella* because biochemical reactions used to distinguish many enteric pathogens, such as inability to ferment lactose and classic serotypes, are either variable or lacking. Although molecular approaches such as DNA probes and PCR can identify EIEC, [169] these methods are not widely used or available in the clinical setting. This limitation is of little consequence clinically because EIEC generally causes a milder illness than *Shigella* and treatment is guided by clinical symptoms.

As for most enteric infections, the first step in management is fluid and electrolyte replacement. Although antibiotics are usually not needed for EIEC infection, no clinical trials comparing antibiotic regimens have been performed. If clinically indicated, antimicrobial recommendations for the treatment of shigellosis can be applied to EIEC infection. It is not known whether antibiotic resistance data for *Shigella* species can be extrapolated to EIEC. In the absence of clinical trials, a reasonable empirical approach is treatment with either TMP-SMX 160 mg/800 mg twice per day, ampicillin, 500 mg four times per day, or cipro-floxacin, 500 mg twice per day for 5 days.

Enterohemorrhagic Escherichia coli

Infection with enterohemorrhagic *E coli* (EHEC) O157:H7 is a major health concern in the United States. EHEC strains, most notably O157:H7, cause severe morbidity and mortality in humans. Since its identification in 1982, more than 100 outbreaks have been reported. [170]

Epidemiology Before 1982, only one O157:H7 strain had been isolated by the CDC from a patient with bloody diarrhea. [171] Soon after, two major outbreaks were reported in those who had consumed hamburgers obtained from a single fast-food restaurant. [171] In 1999, the CDC estimated that *E coli* O157:H7 caused 73,000 illnesses and 60 deaths in the United States alone. Non-O157:H7, Stx-expressing *E coli* added another 37,000 estimated cases. [172] EHEC most commonly infects those of extreme age, although the median age is 14 years. [173] The highest incidence of infection occurs in children younger than 5 years of age. Elderly patients, particularly those who are bedridden, are more susceptible to infection and have a greater likelihood of developing HUS with subsequent mortality. [174] Cattle are the primary reservoir of EHEC O157:H7. [175] Over half the reported outbreaks are due to ingestion of beef products. [175] The prevalence of EHEC O157 in the feces (28%) and on carcasses (45%) of cattle is high. [175] Cattle are tolerant of intestinal colonization by *E coli* O157:H7 and do not develop systemic vascular damage [176] because of the lack of expression of the Gb_3 receptor for Stx in both the gastrointestinal tract and blood vessels within the gastrointestinal tract and kidney. [176] Other sources of EHEC infection include apple cider, vegetables, salads, cantaloupe, raw milk, mayonnaise, and water. [173] Cases of person-to-person transmission have been reported in day-care centers and health care facilities. [177], [178] A seasonal increase in the incidence of EHEC infection occurs in North America in the summer and early fall. Despite the increasing incidence of EHEC-associated illnesses, many hospital and microbiology laboratories do not routinely screen diarrheal or bloody stools for *E coli* O157:H7. When sought, this organism is identified in 15% to 39% of such stool specimens. [173]

Pathogenesis EHEC strains derive their virulence from a number of different factors, including infectivity at a dose of less than 100 organisms, [179] a consequence of resistance to the low pH of the stomach. Other virulence factors include expression of adhesins for initial attachment to host cells and gene products within the chromosomal pathogenicity island termed the *locus of enterocyte effacement* (LEE). LEE-encoded virulence proteins include intimin, an outer membrane protein that serves as a ligand for the LEE-encoded receptor that is translocated and inserted into the host cell membrane (*t*ranslocated *i*ntimin *r*eceptor, Tir), and other proteins that are secreted into host cells. [180] *E coli* O157:H7 also expresses a plasmid-encoded hemolysin and two Shiga-like toxins, Stx1 and Stx2, encoded on phages. These toxins, structurally similar to each other and to *S dysenteriae* toxin, are composed of an enzymatically active A subunit and five B subunits, and induce cytotoxicity by inhibiting host cell translation. Binding of these toxins to host Gb_3 receptors and internalization of the enzymatically active A subunit causes HUS. The susceptibility of cells to Stx directly correlates with the number of Gb_3 receptors expressed. [181] In humans, the highest concentration of receptors is found in renal tissue and on endothelial cells of the intestinal, renal, and central nervous system vasculature. [182] Toxin-induced damage to the endothelium causes fibrin thrombi deposition, intimal hyperplasia, and perivascular inflammation. The resulting thrombus formation and vasculitis lead to intestinal ischemia, vasculitis and hemorrhage, and hence hemorrhagic colitis. *E coli* O157:H7–induced colitis can microscopically resemble ischemic colitis; therefore, infection with this pathogen should be considered in patients presenting with colonic ischemia. [183] Histology of kidneys from HUS patients shows similar ischemic injury. Shiga and Shiga-like toxins are very potent; therefore, the concentration required to induce HUS is exceedingly small. [181]

Clinical Features The spectrum of symptoms associated with EHEC infection ranges from asymptomatic colonization to severe hemorrhagic colitis. The incubation period ranges from 3 to 9 days, with symptoms persisting for an average of 9 days in children and 6.5 days in adults. A minority (10%) of those infected with EHEC have nonbloody diarrhea or lesser clinical manifest-ations. [184] Patients with hemorrhagic colitis suffer severe abdominal cramps and diarrhea that becomes bloody after 2 to 5 days of illness. Blood loss may be minimal or up to 4 cups/day. [185] Vomiting occurs in up to 75% of patients. Ileus accompanies hemorrhagic colitis in 24% of patients. [186] Fever is absent in most cases, but a correlation between the degree of fever and elevation in the WBC count (more than 20,000 WBCs/µL) with the development of HUS and central nervous system injury has been reported. [187], [188] The physical examination is typically unremarkable, although right lower quadrant tenderness and abdominal distention may be present in children. Although colonoscopy is not necessarily indicated, mucosal edema, erosions, and hemorrhage, more severe in the proximal colon, may be seen. [185] Barium enema may show thumbprinting, indicative of mucosal edema. In addition to HUS, other complications associated with EHEC infection include intestinal hematoma causing intestinal obstruction, rhabdomyolysis, and pancreatic necrosis with resultant diabetes mellitus. [184] Rectal prolapse has been reported in 8% of children with HUS. [189]

Diagnosis The diagnosis of EHEC infection is important because its identification may not only improve patient care but also prevent person-to-person transmission and assist in identifying the source of the outbreak. Laboratory tests show most commonly an elevation in the WBC count (7,600 to 20,000 WBCs/µL) and a normal erythrocyte sedimentation rate. [171] Stool leukocytes are typically absent. EHEC O157:H7 is most often identified from diarrheal stool as clear colonies on sorbitol-enriched MacConkey agar (SMA) because this strain does not ferment sorbitol. Because up to 15% of diarrheal stools contain non-O157:H7 organisms that fail to ferment sorbitol, final identification of the O157 serotype requires agglutination tests with O157 antiserum. Presence of the H7 flagellar antigen is similarly identified using H7 antiserum. EHEC strains other than O157:H7 have been implicated in human disease including HUS, but many of these strains ferment sorbitol, thus complicating identification. The recovery of EHEC is greatest when stools are cultured early in the course of disease. [190] Recovery rates approach 100% if cultures are performed within the first 2 days of illness and remain high (91%) for up to 6 days. After 7 days of diarrhea, EHEC is recovered in only one third of stool samples. Often, EHEC cannot be cultured from stool after the onset of HUS because frequently the organism has been cleared from the intestinal tract by this time. Delay in the processing of samples and antibiotic therapy diminishes recovery rates. Although other methods of detection are being tested, including DNA probes and PCR for Stx, the gold standard for O157:H7 identification continues to be SMA culturing followed by O157 and H7 serotyping. This approach detects 75% to 88% of Stx-producing *E coli* that are of clinical concern. [191]

Treatment The most important treatment for those infected with EHEC is fluid and electrolyte replacement. Patients should also be closely monitored for blood loss and for associated complications, most importantly HUS. Antimicrobial therapy has no role in the treatment of this infection. It does not diminish the duration of symptoms or prevent complications, including HUS. A prospective study of children younger than 10 years of age with *E coli* O157:H7 infection found antibiotic therapy to be a risk factor for the development of HUS. [188] Exposure of EHEC to antibiotics increases the release of Stx, [192], [193] resulting in increased concentrations of Stx in the bloodstream and enhancing the likelihood of HUS. Although children who have not received antibiotic therapy may also develop HUS, it is recommended that children with acute diarrhea not be treated with antibiotics until infection with *E coli* O157:H7 has been ruled out. Although identical studies have not been conducted in adults, similar logic should be followed. As for infection with most enteric pathogens, neither antimotility agents nor opioid narcotics should be used in severe cases. These drugs depress colonic motility and increase the likelihood of distention that can further compromise the integrity of the colonic mucosa. Efforts to develop new nonantibiotic strategies for treating *E coli* O157:H7 infection are promising. One such approach has been to design a material that displays the Stx receptor, Gb_3, in the hope of adsorbing the harmful toxin and preventing its interaction with host cell receptors. One reagent, called Synsorb-Pk, is diatomaceous earth to which synthetic Gb_3 receptors have been attached. Oral administration of this compound to mice infected with Shiga toxin–producing *E coli* was not protective but did delay death by 1 day. [194] Another approach used a nonpathogenic *E coli* strain transformed to express an Stx receptor mimic on its surface. Mice infected with Stx-producing *E coli* were completely protected against an otherwise fatal dose of this pathogen. [195] The discrepant results for Synsorb-Pk and the Gb_3 mimic–transformed *E coli* may reside in their differential binding capacities because the transformed organisms have the potential to bind 10,000 times more toxin then Synsorb-Pk. [195] Gb_3 mimic–expressing *E coli* lacks the lipopolysaccharide O antigen and therefore does not colonize the gut.

Hemolytic Uremic Syndrome The association of infection by Stx-producing EHEC strains and HUS was first recognized and reported in 1983. [196] HUS develops in 10% to 15% of patients infected with *E coli* O157:H7, that is, 1.5 per 100,000 persons in the United States. [197] HUS is defined as microangiopathic hemolytic anemia, acute renal failure, and thrombocytopenia. Typically, this triad develops acutely after about 1 week of diarrhea. Because the Stx receptor Gb_3 is expressed by central nervous system vasculature, complications, including seizures (17% to 24%), cerebral edema, and coma (7% to 40%) are common. [198] More than half of HUS patients experience diminished sensorium. Renal sequelae range from mild proteinuria to severe nephropathy. HUS is the leading reason for kidney transplantation in children in the United States. [197] Most (75% to 100%) HUS patients have evidence of EHEC infection, but only 5% to 10% of those infected with EHEC progress to HUS. [179]

The median age of those who develop HUS is 2.7 years, with the peak ages ranging from 6 months to 5 years. [199] An initial high WBC count, performance of stool cultures early in the course of illness, and administration of antibiotics are associated with the development of HUS in children with *E coli* O157:H7 infection. [188] The administration of antimotility agents is an additional risk factor for HUS. [200] Histology of kidneys from HUS patients reveals severe endothelial cell injury, including separation of cells from the basement membrane and thrombotic microangiopathy. Cortical necrosis may also be seen. Injury to these cells stimulates the release of endothelin, a potent vasoactive peptide, which is likely responsible for the associated hypertension. [201] The mainstay of therapy for HUS is supportive measures. [173] Heightened awareness of this syndrome and its association with EHEC infection, coupled with improved fluid and electrolyte balance and dialysis, has decreased the associated mortality from 50% in the 1960s to about 5%.

Prevention The best treatment for infections with Stx-producing *E coli* is prevention. Because infection is usually a result of ingestion of contaminated food products, especially beef, elimination of EHEC from the food supply is the first step. [175] Beef should be adequately cooked so that pathogens are eliminated. An internal temperature of 160°F is effective. Good hand-washing habits and hygiene reduce secondary transmission. The size of outbreaks can be curtailed by prompt identification of the contaminating source.

SEXUALLY TRANSMITTED INFECTIONS OF ANUS AND RECTUM

The populations at greatest risk for acquiring anorectal infections are homosexual men and heterosexual women who engage in anorectal intercourse. The most commonly occurring infections in women are *C trachomatis, N gonorrhoeae* and *T pallidum.* In addition, infections with *E histolytica* HSV, and HPV are endemic in homosexual men. Infection with multiple pathogens occurs in 22% of this population who are symptomatic [202] and is associated with high-risk behaviors.

Bacterial Infections

Chlamydia trachomatis *C trachomatis*, a gram-negative, obligate, intracellular bacterium, is the leading bacterial etiology of sexually transmitted diseases in the United States. *C trachomatis* is represented by three biovars, two of which cause human disease: trachoma and lymphogranuloma venereum (LGV). The replication of trachoma is restricted to columnar epithelial cells, whereas LGV can also proliferate in macrophages, explaining the difference in symptoms. The clinical features associated with infection by *C trachomatis* range from asymptomatic proctitis, due to non-LGV biovars, to severe granulomatous proctitis, usually caused by LGV biovars. Sigmoidoscopic examination of individuals infected with non-LGV biovars may reveal normal mucosa or erythema, friability, and erosions. Histological examination of these tissues shows neutrophilic infiltration of the lamina propia. [203] In contrast, LGV biovars induce severe proctitis or proctocolitis manifested as pruritus, purulent rectal discharge, hematochezia, and diarrhea or constipation. Physical examination may reveal lymphadenopathy and lower abdominal tenderness. The sigmoidoscopic findings are more marked, revealing friable or bloody mucosa with multiple ulcerations. [203] The infiltrate is varied, showing eosinophils, mononuclear cells, and plasma cells in addition to neutrophils. Crypt abscesses and granulomas with giant cells may also be present and can lead to the mistaken diagnosis of Crohn's disease. Anorectal LGV may also cause strictures, fistula formation, and perirectal abscesses if left untreated, further confusing the diagnosis with Crohn's disease. *Chlamydia* species infection is diagnosed by culturing samples obtained by rectal swab in McCoy cells. Infected cells are identified by immunofluorescent staining, using a monoclonal antibody against *Chlamydia*-specific antigens, achieving a sensitivity and specificity of 90% and 100%, respectively. [204] The treatment of choice for chlamydial infection is tetracycline, 500 mg four times daily, or doxycycline, 100 mg twice daily, for 21 days. Less severe infections with non-LGV biovars may be treated with doxycycline, 100 mg twice daily for 7 to 10 days or a single 1-g oral dose of azithromycin. Surgical resection may be required to alleviate strictures.

Neisseria gonorrhoeae *N gonorrhoeae* is a gram-negative diplococcus that typically infects the epithelial lining of the endocervix and urethra but may infect other sites as well, including the rectum. A robust inflammatory response including submucosal abscesses occurs after attachment, internalization, and subepithelial delivery of the organism. Up to half of homosexual men assessed in sexually transmitted disease clinics are infected with this organism, with 40% manifesting only rectal involvement. Of infected women, only 20% had rectal infection alone. [205] Most gonorrheal anorectal infections are asymptomatic but when present include purulent, creamy, and sometimes bloody discharge accompanied by pruritus ani, tenesmus, and even constipation. Symptoms typically appear 5 to 7 days after exposure. Rare complications include stricture formation, abscesses, and fistulae. The sigmoidoscopic findings are nonspecific, including friable, erythematous mucosa with superficial erosions most prominent at the anorectal junction. Culture of material obtained by rectal swab plated on selective Thayer-Martin media is the recommended diagnostic test. Gram stain of this same material is also suggested but is only positive in about half of those infected. The treatment of choice for anorectal gonorrhea is ceftriaxone, 250 mg given intramuscularly, which cures more 98% of such infections. [206] Because of a high co-infection rate with *Chlamydia* species, it is recommended that all *N gonorrhoeae*–infected individuals also receive doxycycline, 100 mg twice a day for 7 days. [206] Alternative regimens include a single oral dose administration of 500 mg ciprofloxacin or 800 mg norfloxacin or a single intramuscular injection of spectinomycin (2 g), cefotaxime (1 g), or ceftizoxime (500 mg). [207] A follow-up culture is recommended for those receiving an alternate treatment regimen.

Treponema pallidum *T pallidum* is a spirochete capable of infecting damaged skin or mucosa. Anorectal syphilis is primarily diagnosed in homosexual men. Primary anorectal syphilis presents as a chancre of the squamous epithelial lining of the anal canal or in the rectum. These lesions, which develop 2 to 6 weeks after exposure, appear as 1- to 2-cm indurated lesions with a raised border. "Mirror" or "kissing" lesions may be present. [208] Anorectal chancres are often asymptomatic unless superinfected. The anorectal presentation of secondary anorectal syphilis is condyloma lata. This stage develops usually 6 weeks to 6 months from the time of the initial infection. Condyloma lata are smooth, wartlike lesions near or in the rectum. Colitis associated with secondary syphilis may also occur in the distal 15 to 20 cm of the colon. This lesion spontaneously resolves after 3 to 4 weeks. Intestinal lesions are rarely associated with tertiary syphilis, but rectal gummas have been reported. Anorectal syphilitic lesions show a chronic inflammatory infiltrate of lymphocytes, histiocytes, and plasma cells. [209] Dark-field microscopic examination reveals the presence of spirochetal organisms, but nonpathogenic spirochetes may also be present in the rectum. [210] Anorectal syphilis should be treated with a single intramuscular injection of 2.4 million units of benzathine penicillin. If intolerant of penicillin, doxycycline, 100 mg twice daily, or tetracycline, 500 mg four times daily, can be given orally for 2 or 4 weeks for early and latent syphilis, respectively. Proof of eradication should be sought 3 and 6 months after treatment.

Viral Infections

Herpes Simplex Virus HSV type 2 accounts for 90% of the cases of anorectal herpetic infections. The virus, for which humans are the sole reservoir, is transmitted by direct contact with an infected individual who is actively shedding viral particles. Symptoms of the primary infection begin 4 to 12 days after contact and spontaneously resolve after 3 weeks but recur. Symptoms associated with subsequent bouts range from none to severe and painful ulcerative proctitis. Other symptoms include tenesmus, pruritus ani, constipation, inguinal adenopathy, sacral and posterior thigh paresthesias, and urinary difficulty. The symptomatic triad of constipation, urinary retention, and anorectal pain is strongly suggestive of anorectal HSV infection. Anoscopic examination typically reveals vesicles, pustules, or shallow ulcers depending on the stage of the lesions. Biopsies of these lesions reveal acute and chronic inflammatory change, superficial ulcerations, and microabscesses. [211] Perirectal lesions are relatively uncommon, occurring in 19% to 27% of women and men, respectively. [212] Viral cultures should be performed to confirm the diagnosis. PCR can also be used to detect HSV DNA in clinical samples. [213] Depending on the severity of the symptoms, one may choose to treat mildly symptomatic individuals with only stool softeners, sitz baths, and analgesics or to employ antiviral therapy with acyclovir, 400 to 800 mg/day for 10 days, for painful infections. This approach has been shown to diminish both viral shedding and the duration of symptoms. [206]

Human Papillomavirus HPV enters basal germinal epithelial cells and replicates in the nucleus, resulting in increased cell proliferation. This response accounts for the skin and mucosal lesions that appear as irregular, verrucous lesions, called *anal warts* or *condyloma acuminata.* These lesions must be distinguished from the condyloma lata in syphilis that are more moist and smooth. Histological examination of condyloma acuminata shows an irregular, thickened epithelium containing areas of koilocytotic cells with peri- nuclear cavitation and nuclear atypia. Some HPV strains cause neoplastic lesions, including cervical and anal cancer. [214] The virus is transmitted through anal intercourse or auto- inoculation to the perianal area and anus. The latency period ranges from 1 to 6 months. If condyloma acuminata are present in the perianal area, anoscopic examination must be performed to determine whether lesions also exist inside the anal canal. Individuals with anal warts may be asymptomatic or complain of rectal discharge, bleeding, or pruritus ani. Diagnosis is by clinical observation, and anorectal syphilis and squamous cell carcinoma should always be excluded. This is accomplished by histological examination that distinguishes benign and malignant lesions and dark-field microscopy to exclude syphilis. Although curative treatment is still lacking, lesions can be controlled with either cryotherapy, which is 63% to 91% effective, [215], [216] or a 20% solution of podophyllin in tincture of benzoin, which removes 77% of lesions. [217] This solution is applied for up to 12 hours and may be repeated one or two times weekly until all lesions are gone. Its caustic nature precludes intranal use. Other therapeutic options include electrocautery, electrodessication, CO_2 laser, and surgery. Recurrence rates vary depending on the treatment modality used: 29% after surgery and 65% after podophyllin treatment. [218]

Sexual Transmission of Intestinal Pathogens In addition to the pathogens discussed previously, intestinal parasites, including *Entamoeba histolytic* and *Giardia lamblia* and other bacterial pathogens, such as *Shigella* and *Campylobacter* species, can be transmitted sexually. The signs and symptoms associated with infection by these pathogens in homosexual men are the same as those experienced by individuals who acquire these organisms by ingestion.

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CHAPTER 89

Graeme P. Young and Finlay A. Macrae

NEOPLASTIC AND NONNEOPLASTIC POLYPS OF THE COLON AND RECTUM

A *polyp* is any protrusion above the mucosal surface. Polyps may take various sizes and shapes, including sessile or pedunculated. They can also be described according to texture of the mucosal surface, position, color, ulceration, bleeding, and histological type. Polypoid lesions of the large bowel are normally classified into three main subgroups (Table 89-1): epithelial polyps, subclassified into neoplastic polyps (consisting of adenomas or carcinomas) and nonneoplastic polyps; and submucosal lesions (which produce a polypoid appearance).

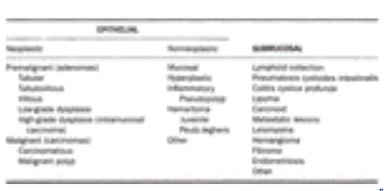

TABLE 89-1 Classification of Colorectal Polyps

Colorectal polyps are important for several clinically relevant reasons. First, they are easily detected and removed with modern flexible endoscopes (see Chapter 140). Largely because of this, our understanding of their role as precursors of colorectal carcinoma has increased (see Chapter 91). Ready access to polyps has led to the elucidation of the genetic basis of the major inherited polyposis syndromes. Furthermore, mutations in the adenomatous polyposis coli (APC) gene responsible for the major inherited syndrome, familial adenomatous polyposis (FAP), also occur in sporadic colorectal neoplasms—but in this case being acquired and developing only from a single clone of cells somewhere in the colon. Finally, interventional trials aimed at removal of adenomas or reduction of recurrence have demonstrated

polypectomy to be valuable for controlling colorectal cancer (see Chapter 34 and Chapter 91).

Neoplastic polyps are of greatest clinical significance because of their implication in the process of colorectal tumorigenesis. The natural history of the adenomatous polyp is variable, but it appears that when progression to carcinoma occurs, it generally takes 3 to 7 years. Based on the relationship of adenoma size to histological features as assessed at polyp resection, it also appears that as adenomas grow, they tend to become more villous and to progress from mild to severe dysplasia, before invasion through the muscularis mucosae (a key feature of cancer). The progression of adenomas in patients with hereditary nonpolyposis colorectal cancer (HNPCC) appears to be faster. With respect to future risk for cancer after polypectomy, the patient with an advanced adenoma (larger than 1 cm, with a substantial degree of villous histological change, or with severe dysplasia) or with multiple adenomas has a significant future risk for colorectal cancer that is not confined to the site of that adenoma.

Colorectal polyps do not usually cause symptoms and are frequently found during diagnostic, screening or surveillance procedures. When clinically suspected, the definitive diagnostic procedure is complete colonoscopy. Endoscopic polypectomy can adequately treat most adenomatous polyps, including some with focal malignancy. Malignant polyps with poor prognostic features may require surgical resection, however.

Nonneoplastic epithelial polyps have little or no malignant potential (see Chapter 90 and Chapter 91), except in the setting of inherited polyposis syndromes, in which they may be associated with cancers of the gastrointestinal tract and other organs. The malignant potential of submucosal lesions depends on their pathological nature.

Management of the patient with a colorectal polyp requires an understanding of the pathology, epidemiology, etiology, clinical manifestations, and course as well as the appropriate diagnostic studies and therapeutic options. [1], [2] and [3]

EPIDEMIOLOGY

Prevalence

Information on the prevalence of polyps comes from autopsy studies and from small-scale colonoscopic surveys. Autopsy studies generally show a higher prevalence rate of polyps and adenomas than endoscopic studies. In the United States, autopsy studies suggest an overall polyp prevalence of up to 50%, [4], [5], [6] and [7] but colonoscopic surveys suggest no more than 40%. [8], [9], [10], [11], [12], [13], [14] and [15] In autopsy studies, fixation of the colon and careful use of a magnifying glass allow detection of small polyps that are more readily missed during colonoscopy. Furthermore, subjects in autopsy studies are older. On the other hand, colonoscopic surveys may include patients with positive occult blood, which would bias them to a higher prevalence. [13]

More than 80% to 90% of polyps detected at colonoscopy are adenomas or hyperplastic polyps, and up to 75% are adenomas. [8], [9], [10], [11], [12] and [13] In the case of diminutive polyps (less than 5 mm), about half are hyperplastic. [16], [17]

Autopsy studies show that the prevalence of adenomas varies widely among countries and parallels the frequency of colorectal cancer in that country (Table 89-2), confirming the close association of adenomas with colorectal carcinomas. [4], [5], [6] and [7], [18], [19], [20], [21], [22], [23], [24], [25], [26], [27], [28], [29], [30], [31] and [32] Most studies show prevalence rates that are 30% higher in men than in women.

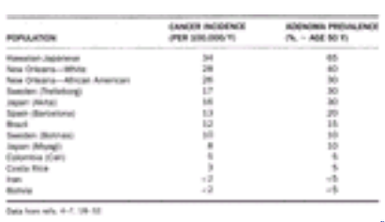

TABLE 89-2 Prevalence of Colorectal Neoplasia in Men

Age is the major determinant of adenoma prevalence. [4], [7], [25] Size at detection also increases with age. [24] Colorectal adenomas are uncommon in patients younger than 30 years of age (see Chapter 90). The prevalence of polyps other than adenoma or hyperplastic is unknown.

Anatomic Distribution

Colonoscopic studies and some autopsy studies show that colorectal adenomas are more common in the distal colon and rectum, similar to the distribution of colorectal cancer (Table 89-3). Distribution, however, relates to size. Small adenomas are more uniformly distributed throughout the entire colon, whereas large adenomas (greater than 1 cm) show a distal predominance. [7], [8] and [9], [33], [34], [35] and [36]

TABLE 89-3 Anatomic Distribution of Colorectal Adenomas

Hereditary Factors and Adenomas

Hereditary factors obviously play the key role in FAP and HNPCC; adenoma biology is characteristically disturbed in each and now well characterized at the molecular level (see Chapter 90 and Chapter 91). However, hereditary factors also play a role in the more common, sporadic adenoma, contributing to as much as 30% of the pathogenesis. [37], [38] and [39]

Analysis of family pedigrees in the state of Utah has identified an autosomal-dominant pattern of inheritance of adenomas. About 200 members of a large pedigree with familial clustering of colon cancer, without evidence of the known inherited syndromes of colorectal cancer, were systematically examined by flexible sigmoidoscopy. Twenty-one percent of family members, but only 9% of spouses, were found to have one or more adenomas. In comparison, hyperplastic polyps were found in 24% of family members and 29% of spouses. [37], [38] and [39]

Subsequent analyses of additional pedigrees continue to demonstrate a twofold to threefold increase in adenomas in family members compared with spouse controls. [40], [41], [42] and [43] Pedigree analysis using likelihood methods was used to estimate which of the possible mechanisms could account for familial clustering of adenomas. It was found that an autosomal-dominant inheritance of susceptibility to adenoma (and colorectal carcinoma) best explained the pattern of adenoma and carcinoma occurrence in the pedigrees. [37] In addition, the gene frequency was estimated to be 19% in that population. A case-control study addressing risk for cancer when a first-degree relative has had one or more adenomas showed risk to be increased, although these studies are few and difficult to interpret because adenomas usually go undiagnosed and are common. [44]

Considered together, these results suggest that familial or hereditary factors play at least a 20% role in the causation of adenomas. [37], [38], [39], [40], [41], [42], [43], [44] and [45] If it is the adenoma rather than cancer that signals the inherited risk, and if surveillance programs are based on family history of colorectal adenoma or cancer rather

than cancer alone, then the complexity, cost, and magnitude of these programs will increase dramatically. More information is needed on the risk to other family members when an adenoma is detected.

Association of Adenomas with Other Diseases

Associations between colonic polyps and other diseases are summarized in Table 89-4.[46,47,48,49,50,51,52,53,54,55,56,57,58,59,60,61,62,63,64,65,66,67,68,69,70,71,72,73,74,75,76,77,78,79,80,81,82,83,84,85,86,87,88,89,90,91,92,93,94,95,96,97] and [98] None of the conditions with a strong association is common. In a prospective study of acromegalic patients, nearly 50% had adenomas, especially when older.[59]

DISEASE	POLYP TYPE	STRENGTH OF ASSOCIATION
Ureterosigmoidostomy	Adenoma/carcinoma[46–51] Juvenile[52,53] Inflammatory[48]	Strong
Acromegaly	Adenoma/carcinoma[54–59]	Strong
Streptococcus bovis infections	Adenoma/carcinoma[60–64]	Strong
Breast cancer	Adenoma[65–68]	Weak
Atherosclerosis	Adenoma[69–76]	Weak
Cirrhosis	Adenoma[77–80]	Weak
Skin tags	Adenoma[81–88]	Very weak
Cholecystectomy	Adenoma[89–96]	Very weak
Diverticula (colon)	Adenoma[97]	Very weak
Lymphoid follicles	Adenoma[98]	Very weak

TABLE 89-4 Disease Associations with Colorectal Polyps and Their Strengths

Nonneoplastic Polyps

The epidemiology of nonneoplastic polyps is not well studied except for the hyperplastic polyp, the second most common colon polyp.[16,99] Hyperplastic polyps usually are smaller than 5 mm in diameter and usually are found in the rectosigmoid area. Some studies have found an association between distal hyperplastic polyps and more proximal adenomas and colon cancer,[100,101,102,103,104,105] and [106] but others have not.[10,107,108,109,110] and [111] The predominant evidence from the larger and better controlled studies would suggest that hyperplastic polyps are not premalignant and do not identify a risk for colorectal neoplasia. In the U.S. National Polyp Study, individuals with distal hyperplastic polyps were no more likely than those without to have more proximal colorectal neoplastic polyps.[107] Unless other risk factors are present, asymptomatic patients found to have diminutive distal hyperplastic polyps at sigmoidoscopy do not need complete colonoscopy.

A possible exception to the rule about the benign and inconsequential nature of hyperplastic polyps is those individuals with multiple hyperplastic polyposis.[112] Despite the epidemiologic information indicating the lack of association between adenomas and hyperplastic polyps, molecular studies indicate that some hyperplastic polyps show genetic change that overlaps with those that accumulate in the adenoma-carcinoma sequence.[112]

Risk for malignant disease should be considered when hyperplastic polyps are numerous (more than 20, i.e., there is polyposis), large (more than 1 cm) or severely dysplastic, and proximally located (especially if more than five are proximal to sigmoid colon) and especially when there are serrated adenomas (adenomatous and hyperplastic change intermingled in a single polyp). Family history of high-risk hyperplastic polyposis or colorectal cancer also points to an increased risk.[113]

Hyperplastic polyps also take on significance in the context of their relationship to serrated adenomas. Serrated adenomas have the same sawtooth architecture as hyperplastic polyps, but cytologically, they have the features of neoplastic nuclei (dysplasia; see later discussion). A third pathway to colorectal cancer involving transition of a small proportion of hyperplastic polyps to serrated adenomas and passing through to cancer with low levels of microsatellite instability (MSI) has been proposed. Methylation is an important mechanism of inactivation of genes in this pathway, which also has the unusual coexistence of both chromosomal instability and MSI (see later). In this context, the finding of serrated adenomas with multiple hyperplastic polyps also signals a need for careful surveillance.[114]

ETIOLOGY AND PATHOGENESIS

The etiology of colonic polyps is identified by their histological type. For instance, inflammatory polyps result from the inflammatory and regenerative response to injury. Because of their clinical significance and prevalence, the remainder of this section focuses largely on neoplastic polyps.

Adenomas are benign neoplasms; all show dysplasia (by definition). Dysplasia is characterized by cells with enlarged, hyperchromatic, elongated, and less polar nuclei arranged in a picket-fence pattern (i.e., stratified), with variably decreased cytoplasmic volume, stroma, apoptosis, and cellular mucin and increased numbers of mitotic cells. These changes result in a more basophilic appearance to the adenomatous epithelium in hematoxylin- and eosin-stained sections.[115,116] Dysplasia ranges from mild to severe (Fig. 89-1). The term *severe dysplasia* has been used synonymously with, and is preferred over, the terms *carcinoma in situ* and *intramucosal carcinoma* when the severely dysplastic cells are confined within the basement membrane.[117,118] Severe dysplasia is characterized by marked cytologic change with severely disordered crypt architecture. Adenomas may be classified by predominant histology; tubular adenomas are the most common, constituting 80% to 85%.[119,120] Tubulovillous adenomas constitute about 10%, with villous adenomas making up the remaining 5%.[120]

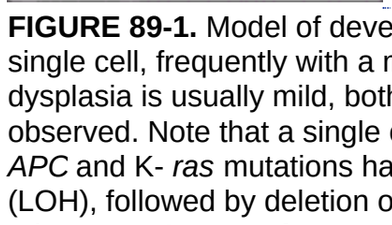

FIGURE 89-1. Model of development of a malignant colorectal carcinoma by a series of phenotypic changes and related genetic alterations. **Normal epithelium:** A single cell, frequently with a mutation in the APC gene (on chromosome 5), will give rise to the small adenoma in the next stage. **Small adenoma:** At this stage, when dysplasia is usually mild, both alleles of APC are affected by deletion or truncation (chromosome 5q). Changes in the level of methylation of DNA also may be observed. Note that a single cell in the clone has acquired an additional mutation in its K- *ras* gene (chromosome 12P). **Intermediate adenoma:** The cell with the *APC* and K- *ras* mutations has given rise to a new clone, forming an intermediate adenoma. An additional mutation in one of the cells, such as loss of heterozygosity (LOH), followed by deletion of a tumor suppressor gene (such as *DCC* [deleted in colorectal cancer] on chromosome 18q), gives this cell an additional survival advantage. **Large adenoma:** The cell with mutations and deletions in the *APC* and K- *ras* genes and LOH on chromosome 18q has clonally expanded into a large adenomatous polyp that has not invaded underlying mucosal layers. One cell of this clone acquires an additional mutation in the *p53* gene (chromosome 17p). **Carcinoma:** The cell that has accumulated mutations in the *APC* K- *ras* and *p53* genes and developed LOH on chromosome 18q has grown into a clone of malignant cells that penetrates through the lamina propria and invades underlying tissues. Note how cells are capable of lysing capillary walls and entering the vessels to be transported to distant sites where they form metastatic tumors. (Additional changes to proteolytic enzymes such as urokinase or telomerase may be required for metastatic behavior.) *M* mucosa; *LP* lamina propria; *MM* muscularis mucosae; *SM* submucosa; *MP* muscularis propria.

Cell Kinetics in Normal Epithelium and Polyps

The healthy colonic epithelium is characterized by constant cell proliferation and differentiation, resulting in total renewal of the surface epithelial cells about every 3 to 6 days. The proliferative compartment is situated in the basal third of the crypt. Daughter cells migrate up the crypt and out onto the surface. As the cells migrate, proliferation ceases, and maturation begins–goblet cells become common.[121,122,123] and [124] The process of cell proliferation and differentiation is highly regulated,

resulting in a homeostatic balance among cell production, maturation, and death. [121]

In adenomas, the normal homeostatic process is disordered. Cell proliferation extends up the crypt. Differentiation is delayed (with fewer goblet cells), and senescence is reduced. [115], [123], [125] The entire colonic mucosa of patients with adenomas may show increased epithelial cell proliferation. [124]

Proliferation and differentiation are not grossly disordered in hyperplastic or inflammatory polyps; cell proliferation is increased but remains confined to the basal third of the crypt. With increased cell production, there are increased epithelial cell elements and some infolding, leading to the characteristic polypoid appearance. However, cell maturation proceeds normally, and goblet cells are plentiful. [116]

Physical Formation of a Polyp

This enhanced proliferation and retarded cell maturation results in an increased number of cells. These undergo downward infolding of the crypts that interpose and branch between the normal crypt elements. [115], [122], [126], [127] Mesenchymal proliferation is also thought to play a role in the formation of adenomas, especially when villous change occurs. [122], [126], [128]

Environmental Factors

Epidemiologic studies suggest that environmental factors play an important role because worldwide variation in incidence is not fully explained by genetic variations. [129] Diet is probably the most important environmental factor. Smoking is also a risk factor for adenomas, and recent evidence also shows an association between tobacco use and colorectal cancer: 15 of 16 studies conducted after 1970 in men and, in the 1990s, women, showed this association. [130], [131], [132] and [133] The dietary factors that predispose to adenoma formation are similar to those that have been associated with colon cancer (see Chapter 91), although the evidence is less complete for adenomas. Although many diet-related factors have been proposed as potential causative agents, the strongest evidence points to fat, bile acids, and fecal bacteria. [134], [135], [136], [137] and [138] Fiber and dietary carcinogens also seem important, although recent studies have been less supportive of a preventive role for dietary fiber. [139], [140] and [141]

Dietary Fat The prevalence of colorectal adenomas, as with cancers, has been correlated with high dietary fat intake (animal fat intake greater than 40% of total caloric intake) [142] (Table 89-5). Conversely, dietary fat intake of less than 15% of total caloric intake has been correlated with a low frequency of colorectal adenomas and cancers. [78], [143], [144], [145], [146], [147], [148] and [149]

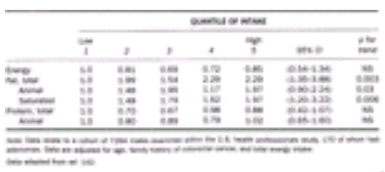

TABLE 89-5 Relative Risk for Colorectal Adenoma in Relation to Dietary Intake

Increased dietary fat has been shown in experimental animals to lead to increased hepatic synthesis of cholesterol and bile acids as well as their increased presence in colonic contents. Bacterial flora in the colon might convert these sterols into cholesterol metabolites and oxidized bile acids, which have tumor-promoting activity. [135] Higher fecal concentrations of bile acids and oxidized bile acids have been found in populations at higher risk for colorectal adenomas and cancers, compared with those at lower risk. Subjects living in areas with high prevalence of colorectal adenomas have higher concentrations of fecal bile acids. Similarly, subjects in these geographic areas have been found to have higher numbers of fecal anaerobic bacteria. There is also an association among high numbers of fecal anaerobic bacteria, increased amounts of metabolized cholesterol and oxidized bile acids in the feces, and the high prevalence of colorectal neoplasms in different populations. [134] and [135]

A likely exception to the relationship between dietary fat and colorectal cancer risk concerns fish oil. Fish and fish oil intake correlate inversely with colorectal cancer incidence. [145], [150] Fish oil consumption has been shown to reduce rectal epithelial proliferation. [145], [151] No studies seem to have related fish intake to adenoma prevalence. Obesity and lack of physical exercise are consistent relationships with colorectal neoplastic risk [152] and may induce adenoma or cancer growth by trophic effects of insulin induced in a state of obesity-related insulin resistance. [153]

Dietary Fiber Dietary fiber is a heterogeneous mix of plant-derived components, specifically nonstarch polysaccharides as well as undigested starch polysaccharides and noncarbohydrates that are resistant to digestion in the upper digestive tract; this complicates interpretation of epidemiologic data. Nevertheless, subjects consuming diets high in fiber that lead to large, bulky stools tend to have a lower frequency of colorectal adenomas and cancers. [78], [138], [142], [144], [147] Case-control studies of dietary fiber and colorectal neoplasia have shown more consistent protection from vegetable than cereal fiber sources, although with a notable recent exception. [137], [138], [147], [148] and [149], [151], [154], [155] However, recent animal studies focus on the relative solubility of different cereal fibers [156] and support the hypothesis that poorly soluble fibers, such as wheat bran, continue to be fermented throughout the colon, releasing metabolic products of fermentation along its length, including the distal colon, which is the region of highest risk for colorectal cancer in humans. [138] Among these products are the short-chain fatty acids: acetate, propionate, and butyrate. Apart from altering the pH to a more favorable acidic state, short-chain fatty acids are an important fuel for the colonocyte. Attention has focused particularly on butyrate, which is capable of inducing a more differentiated phenotype, reducing cell proliferation, inhibiting tumorigenesis, and regulating gene expression by inhibition of histone deacetylase. [156], [157] In carefully controlled animal experiments, tumorigenesis is inhibited, with one third of the protective effect accounted for by the measured stool parameters of fecal pH and butyrate production. [158] Butyrate also induces apoptosis in vitro in colon cancer cell lines by activating the caspase cascade [159] and blocks the cell cycle. The role of fiber in prevention of colorectal adenomas has been tested in several randomized controlled trials and is described later. Resistant starch, by definition, also escapes digestion in the small bowel and may benefit colonic physiology in similar fashion to the nonstarch polysaccharides. [160], [161] Resistant starch is being tested at present in a randomized controlled trial of high-risk individuals in HNPCC families (the CAPP-2 study).

Dietary Carcinogens Studies have demonstrated increased fecal mutagenic activity in the stools of patients at risk for development of colon adenomas and cancers. [135], [162], [163] Whether the increased fecal mutagenic activity is related to dietary fat intake, fecal bile acids, or bacteria is still unclear. One interesting line of investigation relates to metabolic activation of dietary procarcinogens such as heterocyclic amines formed in high-temperature cooking; acetylator status has been shown to correlate with risk for colorectal cancers and adenomas. [163], [164] Recent attention has also focused on fecal *N*-nitroso compounds, which are formed in the colon after meat ingestion and which may cause DNA adducts and be mutagenic. [165] Dietary carcinogens may be detoxified by hepatic enzymes (NAT 1 and 2), which are themselves polymorphic and have variable activity, thereby providing another explanation of familial risk. [166] Glutathione *S*-transferase *M1* and *T1* genes are also involved in the metabolism of polycyclic aromatic hydrocarbons, carcinogens found in tobacco smoke and the diet. [167]

Micronutrients Studies using antioxidants, such as vitamins A, C, and E and beta carotene, have attempted to decrease the production of mutagens in the stool as a way of decreasing colon adenomas and cancers. Randomized controlled trials of these antioxidants in subjects with previous adenomas have universally failed to show a reduction in the risk for metachronous adenomas or cancer. [168], [169], [170] and [171] However, beta carotene does reduce cell proliferation in the colon. [172], [173] This dichotomy highlights the multistage nature of colorectal carcinogenesis, with the opportunity for preventive agents to intercept the process at different stages. Because the actions of dietary agents may be stage specific, the final marker of benefit should be cancer, if this can be demonstrated. There is also evidence that calcium, perhaps by binding fecal fat, bile acids, and cholesterol, may confer a protective effect. [174] Calcium carbonate (3 g, i.e., 1200 mg of elemental calcium) reduced adenoma occurrence in a randomized controlled trial and is one of only a few interventions to do this. [175] Calcium (such as in calcium-fortified low-fat dairy products) also reduces cell proliferation in randomized controlled trials and inhibits tumorigenesis in animals. [176] Selenium also shows promise and is under active investigation. [177] and [178]

Molecular Genetics of Adenomas

Multiple alterations in DNA have been noted in adenomas. Aneuploidy, tetraploidy, allelic deletions, and hypomethylation have been observed in sporadic adenomas (chromosomal instability pathway) and in FAP. [179], [180] and [181] There is also altered patterns of expression of, or mutations in, oncogenes, such as *fos myc* and *ras* and deletions of the *APC* gene [182], [183] and [184] (see Fig. 89-1). Increasing data from studies of very small (2 mm) adenomas highlights the importance of *APC* mutation as the initiating (gate-keeping) event. [185], [186] In the alternate MSI pathway characteristic of HNPCC, replication errors occur during mitosis and mutations occur in other genes, including *TGFßRII MSH6* and insulin-like growth factor receptor type II. Inactivation of promoter regions by methylation (the CpG island methylator phenotype) of tumor suppressor and other genes (*p16, THBSI* E cadherin, and *HPP1*) or mismatch repair genes (*hMLH1* and *MGMT*) is another important molecular mechanism

that may be evident in adenomas. [114], [187]

A third pathway is initiated by silencing of the *HPP1* gene (due to methylation), loss of expression (also by methylation) of the DNA repair gene O^6-methylguanine DNA methyltransferase (causing loss of its expression), and progress through the serrated adenoma pathway to low-level MSI cancers. [187], [188]

These low-level MSI cancers have a higher frequency of K-*ras* mutations than microsatellite stable (MSS) cancers. [189] *hMLH-1* expression may also be lost in serrated adenomas and even in nondysplastic crypts within such polyps, although this usually leads to high-level MSI cancers. *HPP1* may also be the initiating event for this high-level MSI pathway. [187]

Molecular Changes in Hyperplastic Polyps

A range of genetic abnormalities has been described in sporadic hyperplastic polyps, including oncogenes and tumor suppressor genes, especially K-*ras* and *TGFßR11* and also loss of chromosome 1p. *APC* mutations, however, are very rare. These changes suggest a need to reevaluate the premalignant importance of hyperplastic polyps. Correlation of the clinical outcomes and molecular pathology of hyperplastic polyps may teach us further lessons about the pathogenicity of certain accumulated mutations.

ADENOMA-CARCINOMA SEQUENCE

It is generally accepted that most colorectal carcinomas arise from colorectal adenomas; probably all arise one way or another from dysplastic epithelia. The frequency, inevitability, and time course of this neoplastic progression is not clearly defined, and prospective observational studies of adenomas left in patients are unethical. The evidence for the adenoma-carcinoma sequence is thus derived from epidemiologic (see earlier and Table 89-2), morphologic, biologic, anecdotal clinical [190] and therapeutic data. Studies in FAP patients also support this sequence.

Morphologic Evidence

If the sequence is correct, adenomas and carcinomas should be closely associated. Thirty to 50% of those with adenomas have at least one other (synchronous) adenoma. [26], [27], [28] and [29], [190] Furthermore, having once had an adenoma, the likelihood of a future (metachronous) adenoma is increased. [28], [29], [30] and [31] In those with two or more synchronous cancers, 50% to 85% have a synchronous adenoma. Adenomas found in those with colorectal carcinoma are more advanced. [26], [30], [31] and [32]

If the adenoma-carcinoma sequence is correct, one would expect to find small adenocarcinomas arising within adenomas but few or no small adenocarcinomas arising de novo. Systematic morphologic studies of resected specimens show that when minute adenocarcinomas occur, they are frequently found within adenomas; de novo minute carcinoma is rare. [190], [191], [192] and [193]

The relative frequency of de novo versus adenoma pathway of cancer development is, however, now under debate. Japanese investigators claim that a sizable minority of cancers do develop without recognizable adenomatous co-pathology. [194] Japanese investigators studying Western patients have identified similar phenomena, indicating a need to review this question further in Western societies with high colorectal cancer incidence. [195], [196] and [197]

A plethora of reports of "flat adenomas" in Western populations are now appearing, implying a change in nomenclature rather than epidemiology or pathogenesis. [195], [196] and [197] Nevertheless, careful reports using magnifying colonoscopy and dye spray clearly show small flat cancers (with malignant invasion), especially where the central area is depressed. [198] As the resolution of colonoscopes improves and the Japanese skills at endoscopy are spread around the world, more may be found and the debate resolved. It should be noted, however, that the de novo process does not exclude dysplasia as the precursor—it simply establishes that some precancer lesions do not take on the polypoid shape.

If the adenoma-carcinoma sequence is correct, then carcinomas arising within adenomas should gradually increase in size and ultimately replace all the adenomatous elements with carcinoma. Indeed, the frequency of finding adenomatous tissue in a carcinoma varies inversely with the size and extent of invasion of the carcinoma. Associated adenomatous tissue is found in more than 50% of carcinomas with some mucosal invasion, in less than 20% of carcinomas with invasion of the muscularis propria, and in less than 10% of carcinomas with serosal invasion. Furthermore, the proportion of adenomas showing severe dysplasia, which is cytologically tantamount to cancer, is proportional to the size of the adenoma. These observations are founded on extensive experience with large series of adenomatous polyps from many centers, but they were initiated by the studies of Morson. [190]

Evidence from Progressive Genetic Changes

Molecular analysis of colorectal neoplasia at all putative stages of development reveals an apparent progressive accumulation of genetic abnormalities [185], [186], [199] (see Fig. 89-1). The most prevalent molecular genetic finding in the earliest neoplastic pathology identifiable (sporadic tumors developing through the chromosomal instability pathway)—aberrant crypt foci or microadenomas—is that of mutations in the *ras* oncogene. By the time these microadenomas have progressed to a size of 1 cm, about half have K-*ras* gene mutations, primarily in the codon 12 position. [182], [183], [184], [185], [186], [187], [188], [189], [190], [191], [192], [193], [194], [195], [196], [197], [198], [199] and [200] There is no further accumulation through to colorectal cancers. Mutations in the tumor suppressor *APC* gene on chromosome 5, which leads to a variety of neoplastic pressures in the cell, appear to be another early molecular event associated with small adenoma development (somatic mutations or deletions), but they are not present in aberrant crypt foci (except in FAP patients). [199], [200] and [201]

Larger adenomas frequently show an allelic loss of certain chromosomes, especially 5, 17, and 18. About one third of larger adenomas and carcinomas have allelic loss of chromosome 5, and half have allelic loss of chromosome 18. [186] Loss of function of the tumor suppressor gene *p53* on chromosome 17 occurs late in the adenoma stage and seems pivotal to the cancer phenotype. [202], [203] See Chapter 24 and Chapter 91 for a more detailed discussion.

Evidence from Familial Disorders

Familial Adenomatous Polyposis The increasing risk for occurrence of colorectal carcinoma with increasing numbers of adenomas is well established in the polyposis syndromes. Typical FAP is characterized by the development of more than 100 adenomas; the risk for colorectal carcinomas is virtually 100% if the colon is not removed. In addition, adenomas precede the appearance of carcinoma by about 5 to 7 years. Mutations in the *APC* gene occur constitutionally in FAP, whereas the same gene has mutations in the tumor cells (only) in sporadic colorectal cancers. [204], [205], [206] and [207]

Hereditary Nonpolyposis Colorectal Cancer In the HNPCC (Lynch) syndromes, carcinomas may arise through small precursor adenomas and flat adenomas. [45], [208], [209] This progression may occur quite rapidly, owing to an accumulation of oncogene mutations or suppressor gene deletions. Thus, the morphologic sequence of dysplasia to cancer is retained in the HNPCC pathway, but the molecular pathway is different. There are also subtle differences in pathology: more mucinous tumors at a more proximal location and a more prominent lymphoid reaction with tumor-infiltrating lymphocytes. MSI due to DNA mismatch repair is the hallmark feature of this sequence; frequency of MSI in adenomas is not yet clear. In HNPCC, the MSI pathway does not usually involve K-*ras DCC* or *p53.* [209] Molecular alterations in adenomas of patients with HNPCC involve a different pathway, including *TGFßRII* and *MSH6* itself. [188] Up to 15% of cancers show MSI, although only about 1% to 5% are due to a germ-line mutation of one of the mismatch repair enzyme loci *(MLH2, MSH1, MSH6, PMS2).*

Evidence from Clinical Observations

The average age at which adenomas develop (50 years) appears to precede the age of development of colon carcinomas (57 years). [26] Although it is considered unethical to follow an untreated adenoma for the possibility of progression, available evidence suggests that progression may occur. Adenomas have occasionally been left untreated because of patient noncompliance or technical difficulties; invasive carcinomas have been found to develop at the site of these adenomas. [190], [210], [211] Follow-up studies of patients who have had polyps smaller than 1 cm in diameter, demonstrated on barium enema in the precolonoscopic era when such lesions

were not surgically removed, have shown that cancers subsequently developed at this site (15%) or elsewhere in the colon (25%). [210]

The frequency of finding carcinoma within an adenoma also increases proportionally with increasing size, more severe dysplasia, and greater villous component of the adenoma. [190], [211], [212] Increasing numbers of adenomas is associated with increasing risk for future carcinoma. [190]

Evidence from Therapy

In a large, uncontrolled study, patients underwent periodic sigmoidoscopies, and all polyps that were found were removed. Most polyps removed were found to be adenomatous. The frequency of carcinomas occurring in a subsequent 7- to 14-year period was found to be about one third of the expected rate in an untreated population. [213] This has been confirmed in a retrospective study [214] and in the prospective National Polyp Study. [215] In the Murakami Study, [214] risk for subsequent cancer was highest in the patients who had biopsy alone and lowest when polypectomy was performed. In the National Polyp Study, patients underwent colonoscopy and polypectomy on enrollment with repeat colonoscopy at 1 year (in half) and at 3 and 6 years. No symptomatic cancers and five asymptomatic malignant polyps were found after almost 6000 patient-years of follow-up over more than 4 years. [215] This represents a 58% to 85% reduction in mortality rate from colorectal cancer, depending on whether the reference population is of average risk or includes adenomatous patients. In a study in patients with FAP who had colectomies and ileorectal anastomoses, it was found that periodic removal of adenomas from the rectal stump prevented the subsequent development of rectal carcinoma. [26]

Exceptions in the Evidence

There are exceptions to this adenomatous polyp–carcinoma sequence. There are rare reports of very small carcinomas developing de novo in apparently nonadenomatous epithelia. [216], [217] It is still possible, however, that the carcinoma may have arisen in a focus of the adenomatous epithelium that had not assumed a polypoid appearance (an event that has been observed in FAP), and that the carcinoma obliterated the dysplastic epithelium. The frequency with which this nonpolypoid progression occurs is important because effectiveness of strategies to prevent colorectal cancer through detection and removal of adenomatous polyps will be limited to the proportion that evolve through this pathway. [208], [218]

Although there is no precursor polyp, colorectal carcinoma in the setting of longstanding ulcerative colitis is believed to develop from a dysplastic focus. The progression from inflammation-injury-regeneration to dysplasia and then to carcinoma is believed to be analogous to the adenoma and dysplasia–carcinoma sequence. In addition, the dysplastic lesion in longstanding ulcerative colitis often assumes a raised plaquelike appearance. [219]

Overall, it would appear from these lines of evidence that most carcinomas arise from adenomas or an analogous dysplastic change in the colonic epithelium, even though different molecular biologic pathways may be responsible.

PRIMARY PREVENTION

A 20-fold change in risk for colorectal cancer has been observed in individuals at the extremes of the dietary spectrum. [78] If populations can be convinced to recognize unhealthy eating habits and change them, substantial control of colorectal cancer incidence could follow. The feasibility of this has already been explored in one of the first controlled trials of dietary intervention, the Australian Polyp Prevention Project. [169] In that study, adenoma recurrence after polypectomy was used as the surrogate for effect of diet on cancer. Except in the dominantly inherited colorectal cancer syndromes, the extreme of diet-related risk is greater than the degree of familial risk; hence, attention to dietary issues could have substantial impact. Unfortunately, the prospect for primary prevention by dietary means has suffered setbacks with the publication of the negative findings in the Arizona wheat bran trial and the National Cancer Institute study of low-fat plus fresh fruit and vegetables diet. [140], [141] The option of chemoprevention, especially by nonsteroidal antiinflammatory drugs, may have benefit. [220], [221], [222], [223], [224], [225], [226], [227], [228] and [229]

Dietary Intervention Studies

A study in New York of patients with FAP showed that wheat bran reduced adenoma formation in compliers, although not when analyzed on an intention-to-treat basis. [168] The Toronto Polyp Study [230] was a randomized controlled trial of low fat and high-fiber wheat biscuits versus normal fat and placebo biscuits in a population of adenoma patients. This study showed no beneficial effect of the intervention. The results, however, were not analyzed with respect to the size of adenomas developing during intervention, and the intervention biscuits necessarily involved some processing, which may have detracted from the fermentability of the fiber source.

The Australian Polyp Prevention Project [169] was a factorial randomized controlled trial in 424 subjects with recently removed adenomas, testing a low-fat diet (less than 25% energy as fat, red meat retained), a 25-g fine-particle unprocessed wheat bran supplement, and a 20-mg capsule of beta carotene. This trial proceeded over 4 years. None of the subjects randomized to the combination of low fat and wheat bran developed adenomas more than 9 mm in size, a result that was significant ($p < 0.03$) at both 2 and 4 years (Table 89-6). Trends for less dysplasia with wheat bran and fewer large adenomas with low fat were also seen. Although the proportion of patients with large adenomas was not the primary outcome parameter used to establish the sample size in the Australian Polyp Prevention Project, adenoma size was always considered an important outcome variable. Furthermore, pathological, [119] epidemiologic, anatomic, [33], [34] and [35] and molecular biologic [179] considerations all point to the importance of large adenomas in the pathogenesis of colorectal cancer. The Australian Polyp Prevention Project demonstrated the feasibility of compliance with dietary change in a high-risk group of patients (those with a history of adenomas). [169]

TABLE 89-6 Recurrence of Adenomas of Larger than 9 mm at 2 and 4 Years of Follow-up in the Australian Polyp Prevention Project

A larger randomized controlled trial of 13.5 g of processed wheat bran fiber versus placebo bran, not associated with fat reduction, showed no effect on adenoma recurrence of any size. [141] The National Cancer Institute Polyp Study also yielded a null result with a low-fat diet (less than 20% calories as fat) in combination with 3.5 servings per 1000 calories per day when compared with a no-change diet. [140] Neither of those latter studies combined a low-fat diet with wheat bran supplementation as in the Australian Polyp Prevention Project, but they have been influential in reducing enthusiasm for the preventative effects of dietary fiber.

SIGNS AND SYMPTOMS OF POLYPS

Colorectal polyps are usually asymptomatic. [231], [232] and [233] Most often, the polyps are discovered as an incidental finding during examination for intestinal or nonspecific abdominal complaints or other disorders or during screening endoscopy. Even in the case of FAP, in which adenomas number in the hundreds to thousands, symptoms occur late or result from development of colorectal carcinomas.

In a report of more than 800 hospitalized patients with colorectal polyps, 32% were found to be asymptomatic. [232] When symptoms occur, they tend to be in patients who have colorectal polyps of 1 cm in size or larger. The most common symptoms are rectal bleeding, both overt and occult, change in bowel habit, abdominal pain, and rectal prolapse. Of course, the association of these with a polyp may be coincidental. Furthermore, symptoms have little correlation with histological status, pathological condition, or location. [234]

Histological studies have shown that compared with carcinomas, adenomas tend to have minimal ulceration and an intact lumenal surface. Rectal bleeding tends to be intermittent and small in amount, only rarely resulting in anemia, with the exception of FAP. [235] Studies have demonstrated that only patients with polyps of 1.5 cm

in size or larger have consistently higher amounts of (usually occult) fecal blood loss than healthy control subjects. [236]

Many screening studies using fecal occult blood tests (FOBTs) demonstrate that 20% to 40% of asymptomatic patients with positive FOBT results have colonic or rectal adenomas, and 5% to 10% have cancer. Conversely, 15% to 40% of patients with adenomas have positive FOBT results, depending principally on the size of the adenoma. Newer fecal occult blood technology (immunochemical tests) and fecal DNA testing might increase both the sensitivity and specificity of fecal tests. [237], [238]

Less than 20% of patients presenting with frank rectal bleeding are found to have colorectal adenomas as the only obvious pathology. Rectal bleeding, especially of recent onset and in patients older than 40 years of age or those with other risk factors such as family history, should not be ascribed to coexisting hemorrhoids without a thorough evaluation of the anus, colon, and rectum. [239], [240]

A syndrome of profuse, watery diarrhea, resulting in massive fluid and electrolyte depletion, has been described for large villous adenomas, especially those located in the distal colon and rectum. [233], [241] Colorectal polyps, especially juvenile polyps, may occasionally undergo autoamputation, resulting in rectal bleeding. The presence of recognizable polyp tissue in the feces is extremely rare. [242], [243] Colocolic intussusception due to large polyps has been found in children. [244] Diffuse, nonspecific abdominal pain has been at times attributed to intussusception, particularly of larger polyps. Rectal prolapse of larger polyps also has been reported. [244]

Physical examination rarely reveals signs of colorectal polyps. A digital examination by an experienced person is important as part of all anorectal and colonic evaluations, especially because flat villous lesions close to the dentate line are easily missed at endoscopy, and the digital examination frequently reveals the diagnosis most easily.

MORPHOLOGIC AND CLINICAL FEATURES OF NEOPLASTIC POLYPS

The initial diagnosis is usually made by direct endoscopic visualization of the polyp, which should then undergo biopsy or be removed by polypectomy to determine its histological type. At the time of endoscopic examination, certain morphologic features may help classify the polyp, although these features can be misleading. [245], [246]

Because the chance of malignancy in an adenoma is proportional to its size, its villous component, and the degree of dysplasia, these parameters should be described by the pathologist. [247], [248], [249], [250] and [251] This alerts the clinician to the need to look especially carefully for malignancy in multiple sections in, for example, the large villous adenoma with severe dysplasia. It also partly determines the risk for future advanced adenomas in that individual, thereby guiding decisions on surveillance.

Tubular Adenomas

Tubular adenomas (Fig. 89-2 and Fig. 89-3; Color Fig. 89-2 and Color Fig. 89-3) usually are darker red than the surrounding mucosa, have a smooth surface, and tend to be smaller than villous adenomas. Histology shows the presence of infolding tubules. These adenomas may display the entire spectrum of dysplasia, ranging from mild to severe [26], [115], [120], [190] (Fig. 89-4 and Fig. 89-5; also see Color Fig. 89-4 and Color Fig. 89-5 and Fig. 89-9 and Fig. 89-10).

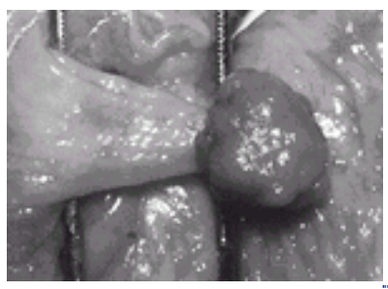

FIGURE 89-2. (See Color Fig. 89-2.) Macroscopic appearance of a pedunculated tubular adenoma. The forceps mark the base and the neck of the stalk.

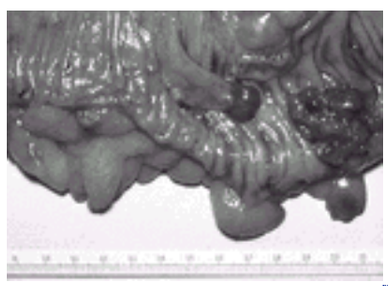

FIGURE 89-3. (See Color Fig. 89-3.) A synchronous, pedunculated, tubular adenoma adjacent to a sessile, tubulovillous adenoma. Synchronous adenomas not infrequently are found adjacent to each other. In this instance, a 1-cm, pedunculated, tubular adenoma is found about 5 cm from a 3-cm, sessile, tubulovillous adenoma. Note the smooth surface of the tubular adenoma and the smooth, lobulated surface of the tubulovillous, primarily tubular adenoma.

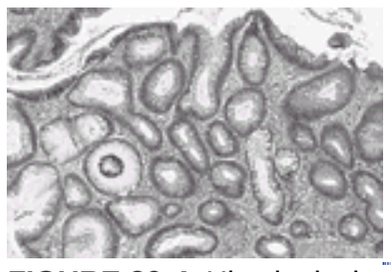

FIGURE 89-4. Histological appearance of a tubular adenoma with low-grade dysplasia. Architectural organization includes orderly, closely packed epithelial tubules. Cells have a normal nuclear-to-cytoplasmic ratio, nuclei have a predominantly basal orientation, and goblet cells are present.

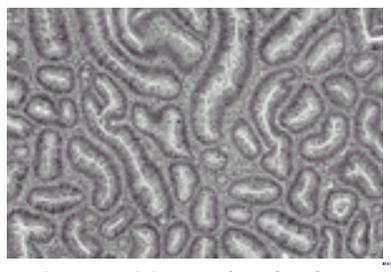

FIGURE 89-5. Histologic appearance of a tubular adenoma with low-grade dysplasia but slightly more architectural distortion than in Figure 89-4. There is also more cellular crowding, variation in nuclei, and occasional loss of basal polarity with an increased nuclear-to-cytoplasmic ratio. No goblet cells are seen. In the three-tier system of grading dysplasia, this could be classed as moderate in grade.

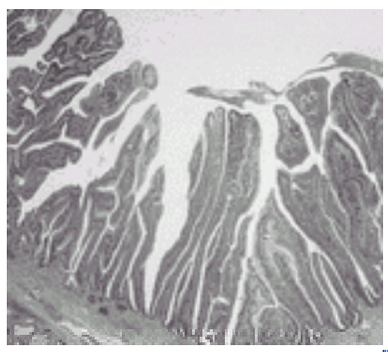

FIGURE 89-9. Histological section of a villous adenoma with typical frondlike appearance. Note the more marked architectural disturbance than seen with tubular adenomas. The nuclear-to-cytoplasmic ratio is increased, and nuclei are not always basally situated.

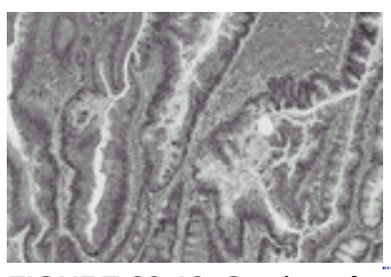

FIGURE 89-10. Section of a tubulovillous adenoma showing high-grade dysplasia. Note the substantial architectural disturbance and pallisading of nuclei.

Villous Adenomas

Villous adenomas constitute about 5% of all adenomas and tend to occur more often in patients older than 60 years of age [191], [192], [247], [248], [249] and [250] (Fig. 89-6, Fig. 89-7, Fig. 89-8, Fig. 89-9; Color Fig. 89-6, Color Fig. 89-7, and Color Fig. 89-9). When seen during endoscopy, they usually are larger and typically sessile compared with tubular adenomas. The surface has a shaggy, cauliflower-like or frondlike appearance, may be soft and velvety, and is often friable. A finely granular surface pattern and a reddish color with white spots may be seen, and spraying of dye may show a characteristic pattern. [248] Villous adenomas tend to be distal in location, large, and bulky. [248], [250]

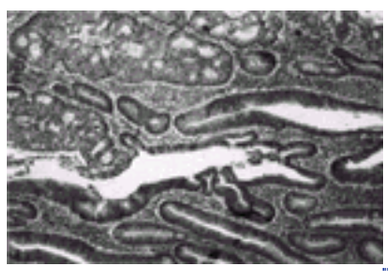

FIGURE 89-6. Histological appearance of a tubulovillous adenoma with severe dysplasia and contiguous carcinoma. Note the hyperchromatic elongated nuclei with loss of basal polarity and the increased nuclear-to-cytoplasmic ratio. There is a sharp and drastic transition between these adenomatous features and carcinoma.

FIGURE 89-7. (See Color Fig. 89-7.) This cecal villous adenoma is virtually circumferential, with submucosal carcinomatous invasion. The lesion is sessile in nature and has a cauliflower appearance with a shaggy, frondlike, friable surface that contrasts with the smoother, lobulated surface seen in Figure 89-2.

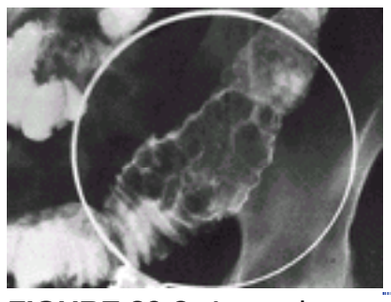

FIGURE 89-8. A very large, multilobulated, polypoid mass is demonstrated in the colon by radiography. Note the shaggy-appearing mucosal surface. The mass was

subsequently found to be a large, sessile, villous adenoma that was virtually circumferential, similar to the lesion in Figure 89-7.

Tubulovillous Adenomas

Tubulovillous adenomas contain at least a 25% villous component and at least a 25% tubular component, without either architectural type predominating. Their clinical behavior tends to be intermediate between that of tubular and villous adenomas [119] (Fig. 89-6 and Fig. 89-10; Color Fig. 89-6 and Color Fig. 89-10).

Flat Adenomas

Nonpolypoid adenomas, or flat adenomas (localized dysplasia without macroscopic protrusion), have been described, particularly in some cancer-prone families. These tend to be minimally raised if at all, small (usually less than 1 cm) discoid plaques, and they are often erythematous. Because they are not polypoid, they are often missed during colonoscopy. [45], [193], [194], [195], [196] and [197], [216], [217] and [218], [252], [253] and [254] Judicious use of air insufflation during colonoscopy is needed to detect these lesions because overinflation makes them very difficult to detect at colonoscopy.

A familial syndrome with flat adenomas has been described by Lynch and colleagues. [218], [255] Most of these "hereditary flat adenoma syndromes" are atypical (or "attenuated") FAP, with mutations in the 5'-proximal end of the gene producing a very short, truncated protein. Flat adenomas might be an early phase of adenomas in HNPCC, but this is less certain.

Despite their small size, more than one third of these flat adenomas have foci of high-grade dysplasia, especially if centrally depressed. [198], [255], [256] and [257] In addition, flat adenomas are often multiple and associated with small adenocarcinomas. [252], [253], [254], [255], [256], [257] and [258]

Serrated Adenomas and Mixed Hyperplastic-Adenomatous Polyps

Serrated adenomas have the architectural features of hyperplastic polyps but include intermingled glands with the cytologic features either of adenomas (dysplasia) or of hyperplastic glands [259], [260], [261] and [262] (Fig. 89-11; Color Fig. 89-11). They account for less than 1% of colorectal polyps. [260] Serrated adenomas are associated with a high frequency of high-grade dysplasia (up to 40%). Mixed hyperplastic-adenomatous polyps show the two types of histology in distinctly separate regions; they should also be treated with caution.

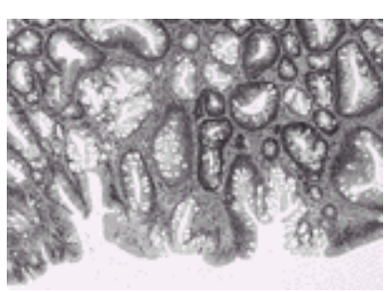

FIGURE 89-11. Histological section of a serrated adenoma showing intermingling of dysplastic and hyperplastic cytology, the latter being responsible for the serrated appearance at a lower power of magnification. Goblet cell density is highly variable. Typically, the hyperplastic cytology is more prominent toward the surface. Note the substantial architectural disturbance and the great variation in nuclear-to-cytoplasmic ratio between the different types.

MORPHOLOGIC AND CLINICAL FEATURES OF NONNEOPLASTIC POLYPS

The nonneoplastic polyps consist of distinct and unrelated groups: simple mucosal, hyperplastic, inflammatory (including pseudopolyps), hamartomatous (including juvenile polyps), and other submucosal polypoid lesions. Dysplasia does not occur by definition.

Simple Mucosal Polyps

Simple mucosal polyps are small, pearl-like excrescences with coloration and appearance similar to adjacent normal mucosa. They are usually smaller than 5 mm, consist of normal colonic mucosa, and have no known clinical significance. [119]

Hyperplastic Polyps

Hyperplastic polyps are usually small (less than 5 mm) and sessile, are found predominantly in the distal colon, and are pale in color. The frequency of hyperplastic polyps appears to increase with age. [19] They are characterized by elongated glands with papillary infolding and increased mucus (Fig. 89-12 and Fig. 89-13; Color Fig. 89-12 and Color Fig. 89-13), which often gives a serrated appearance to the surface of the polyp. [116], [126], [263], [264] The cribriform crypt pattern of adenomas is not usually seen. Such changes may be more easily discerned by using chromoendoscopy.

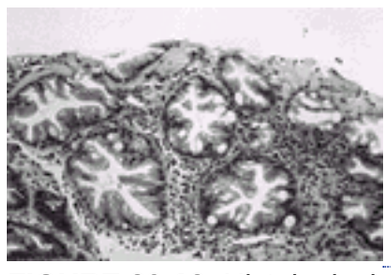

FIGURE 89-12. Histological appearance of a hyperplastic polyp, characterized by the typical elongated glands with papillary infolding, which produces a serrated surface appearance. The epithelial cells are well differentiated, with abundant cytoplasm, a normal nuclear-to-cytoplasmic ratio, and abundant goblet cells. The nuclei are not hyperchromatic, retain their basal polarity, and show no dysplasia. The starfish appearance of the hyperplastic crypts seen in cross section is characteristic.

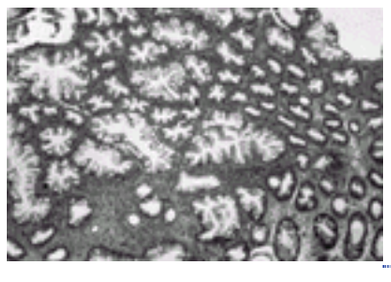

FIGURE 89-13. Histological appearance of adenomatous epithelium in a predominantly hyperplastic polyp. This polyp should be managed as if it were an adenoma.

Hyperplastic polyps have been associated with the presence of adenomas and even carcinomas in the same colon, [212], [265], [266] although larger studies report this association as serendipitous. Therefore, they are not considered a risk factor for colorectal cancer, unless in the context of hyperplastic polyposis or mixed polyps (see earlier). Despite their lack of malignant potential, hyperplastic polyps are often removed because it is not possible to be certain at endoscopy that they are not adenomas. [245], [246]

Inflammatory Polyps

Inflammatory polyps are usually found in the setting of severe inflammatory diseases of the colonic mucosa: ulcerative colitis, [267], [268] and [269] Crohn's disease, [270], [271] amebiasis, [272] strongyloidiasis, [273] tuberculosis, [274] solitary rectal ulcer syndrome, [275] ischemic colitis, [276] severe diverticular disease, [277] or schistosomiasis. When a polyp consists of residual islands of intact colonic epithelium in an area denuded of mucosa, the term *pseudopolyp* is often applied. Inflammatory polyps can also consist of exuberant masses of granulation tissue. Inflammatory polyps may be large and solitary, with marked inflammation and ulceration, and can thus appear to be malignant at endoscopy. [267], [269], [270] In schistosomiasis, the inflammatory polyp may contain schistosome eggs or adult worms. Dysplasia in schistosomiasis-associated polyps may be premalignant. [278]

Inflammatory polyps may produce symptoms of pain and obstruction [278] and may become pedunculated [278], [279] or filiform. [274], [280] Occasionally, they may occur in settings of minimal inflammation, such as ulcerative colitis in remission, [267], [281] or in a familial setting. [282] Inflammatory polyps must be carefully distinguished from dysplastic and neoplastic mass lesions [268] (Fig. 89-14).

FIGURE 89-14. Multiple inflammatory polyps are present throughout the colon in a patient with ulcerative colitis. Colonoscopy and biopsy are necessary for proper identification of the lesions.

The inflammatory "cap" polyp, found in the rectal area, is thought to be a result of mucosal prolapse. It is covered by a cap of inflammatory, hemorrhagic, or granulation tissue. Associated symptoms include mucous diarrhea, tenesmus, and rectal bleeding. Symptoms resolve after polypectomy. [283], [284] and [285] An unusual inflammatory polyp arising in the anorectal mucosa, and often presenting with rectal bleeding, is the inflammatory cloacogenic polyp. [275], [286]

Another inflammatory polyp has been termed the *inflammatory myoglandular polyp* because of the presence of inflammatory and granulation tissue in the lamina propria with abundant smooth muscle proliferation and hyperplastic glands with occasional cystic dilation. When symptoms are present, they consist of overt or occult rectal bleeding. [287]

Carcinoma can occur in inflammatory polyps; unusual inflammatory polyps in complex formations should be sampled. [268]

Hamartomatous Polyps

Juvenile Polyps Juvenile polyps, also known as *retention polyps* because they contain mucin cysts, are so termed because three fourths of them are found in children younger than 10 years of age. [288], [289] They tend to be single, pedunculated, cherry-red, and round and possess a smooth surface. They are often friable and ulcerated, bleed frequently, and tend to prolapse because of their pedunculated nature. [242], [289], [290], [291], [292] and [293] They are also liable to volvulus with resultant ulceration, hemorrhage, autoinfarction, and autoamputation. Histologically, juvenile polyps are hamartomas with distended, mucus-filled glands, often with cystic dilation and edematous lamina propria containing abundant vasculature. Juvenile polyps are rare in the first year of life and are thus presumed to be acquired and not congenital. They occur rarely in the second decade of life but become common again in adulthood. The true prevalence is difficult to determine. In one study, a frequency of about 2% was found in asymptomatic children. [294] In series of children seen for rectal bleeding, 30% have juvenile polyps, with up to half of these being multiple. [291], [292], [295] Of all polyps found in children younger than 15 years of age, up to 97% can be juvenile polyps. [242] Isolated pure juvenile polyps usually are not dysplastic and have no intrinsic malignant potential. Juvenile polyps also may occur in a familial setting, [296], [297] with a pattern of autosomal-dominant inheritance of polyposis (see Chapter 90). In familial juvenile polyposis, adenomatous and carcinomatous changes are frequent throughout the gastrointestinal tract and in family members. [296], [297], [298], [299], [300], [301], [302], [303], [304] and [305] Jass has provided guidelines for the diagnosis of juvenile polyposis as follows: [306]

- More than five juvenile polyps of the large bowel
- Juvenile polyps throughout the gastrointestinal tract
- Any number of juvenile polyps with a family history of juvenile polyposis.

Because solitary juvenile polyps occasionally have associated adenomatous changes, all juvenile polyps should be excised in toto. After removal, juvenile polyps recur only infrequently, with a rate of about 3% in 3 years. [294] If extensive adenomatous changes or severe dysplasia is seen, the patient may be considered for surveillance. [302], [303], [304], [305] and [306] Two candidate genes responsible for familial juvenile polyposis have been reported: the *PTEN* gene [307] and the *SMAD4*/ *DPC4* gene [308] on chromosomes 10 and 21, respectively. Recently, another locus, the bone morphogenetic receptor 1A gene *(BMPR1A)* has also been described in juvenile polyposis. [309]

Peutz-Jeghers Polyps Multiple hamartomatous polyps are found throughout the gastrointestinal tract in Peutz-Jeghers syndrome [310] and usually show prominent branching of smooth muscle. This is a rare autosomal-dominant syndrome of gastrointestinal hamartomatous polyposis associated with melanin spots of the lips, buccal mucosa, and extremities [311] (see Chapter 90). Peutz-Jeghers polyps are not considered premalignant, although the syndrome has occasionally been associated with an increase in carcinomas, mostly extracolonic. [312], [313], [314] and [315] The genetic locus for Peutz-Jeghers syndrome is the serine-threonine protein kinase gene on chromosome 19p. [316] Unusual hamartomas with abundant adipose tissue have been described. [317], [318]

Submucosal Polyps

Submucosal polyps represent elevations of the mucosal surface by submucosal lesions. They are diverse in origin.

Lymphoid Hyperplasia When a focus of diffuse lamina propria lymphocytes becomes hyperplastic, diffuse nodular lymphoid hyperplasia may result. [319], [320], [321] and

[322] Typically, the colonic (or more usually, small intestinal) mucosa is covered with polypoid nodules, generally 5 mm in size or smaller. [320], [321], [323], [324] Symptoms are unusual, and clinically significant nodular lymphoid hyperplasia is usually a condition of the small bowel in children, without any clinical significance. [320], [323], [325] In a series of 1000 consecutive autopsies, incidental nodular lymphoid hyperplasia was found in 3%. [326] Discrete lymphoid polyps, which appear pale yellow or white, may also occur but these do not produce symptoms. [327], [328] Diffuse nodular lymphoid dysplasia occurring in the large bowel needs to be distinguished from familial adenomatous polyposis; biopsy with histopathological diagnosis is important. [329], [330] The latter requires surgery; the former is usually coincidental and requires no intervention. Distinction between malignant lymphoma and multiple polyps can be difficult [331], [332], [333] and [334]; lymphoma has developed in a few patients with apparent nodular lymphoid hyperplasia. [321], [335], [336] It is unclear whether the malignant lymphoma represents a progression from the lymphoid hyperplasia or a de novo development of lymphoma.

Pneumatosis Cystoides Intestinalis Pneumatosis cystoides intestinalis refers to multiple air-filled cysts found within the submucosa of colon or small intestine. The air-filled cysts are easily recognizable by radiographic examination as well as endoscopy (Fig. 89-15). At endoscopy, they are seen as multiple polypoid lesions in the colonic mucosa, which on closer examination appear to be translucent polyps that may look like air blebs just underneath the mucosa. On biopsy, there is often a release of the submucosal air and deflation of the polypoid lesion. [337], [338]

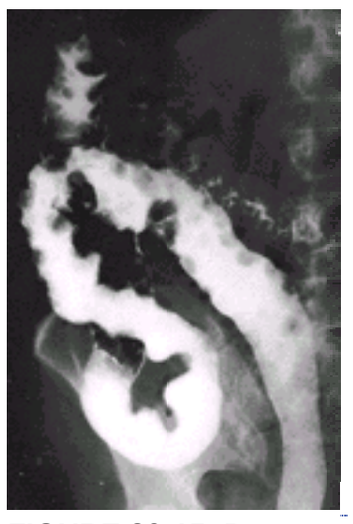

FIGURE 89-15. Pneumatosis cystoides intestinalis seen on barium-enhanced radiography. The intramural gas-filled cysts could have been identifiable on a plain abdominal radiograph.

Pneumatosis intestinalis is generally asymptomatic [337], [338] but can also be associated with pneumoperitoneum. [339], [340] Occasional cases associated with scleroderma or chronic obstructive pulmonary disease have been seen. [338] In these cases, the air-filled cysts may resolve after treatment with oxygen. [340] Pneumatosis intestinalis may also complicate ischemia, necrosis, or necrotizing enterocolitis, when it is often fatal. [336]

Colitis Cystica Profunda Colitis cystica profunda is usually a manifestation of rectal prolapse. The radiographic appearance of colitis cystica profunda is virtually indistinguishable from that of solitary or multiple colonic polyps. The endoscopic appearance can be alarming, mimicking a polypoid cancer. The polyps are sessile and consist of dilated, epithelium-lined mucous cysts within the muscularis mucosae. They are found most often in the rectum in association with inflammatory colitis, or after surgery. [341], [342], [343], [344] and [345] When seen within an adenoma, colitis cystica profunda has been referred to as *pseudoinvasion, epithelial displacement* or *misplaced epithelium*. [346], [347]

Lipomas Lipomas within the gastrointestinal tract are found most commonly within the colon, usually in the right colon on or near the ileocecal valve. These lesions tend to be solitary but may be multiple in 20% or more of cases. Radiographically, they often show up as slightly radiolucent submucosal lesions. At colonoscopy, they appear yellow with a normal mucosa overlying the polyp continuous with surrounding nonpolypoid mucosa. They are easily identifiable on probing with the biopsy forceps. Polyps with these characteristics need neither biopsy nor polypectomy. [348], [349]

Other Submucosal Lesions A number of other rare submucosal lesions also may present radiographically or endoscopically as polypoid lesions. Biopsies of the surface would reveal normal colonic epithelium. Deeper biopsies, or biopsy after submucosal saline injection to elevate the mass, may be required to reach the submucosal mass. Endorectal ultrasonography is particularly useful in diagnosing the origin of a submucosal tumor and whether invasion is present. Malignant submucosal lesions include carcinoids [350], [351]; metastatic carcinomas, most commonly malignant melanoma [352] and lymphoma [332], [333]; gastric cancer [353]; Kaposi sarcoma [349]; and plasma cell leukemia. [354] Benign lesions include lymphangiomas, fibromas, neurofibromas, [355] leiomyomas, myoblastomas, hemangiomas (including the blue rubber bleb syndrome), and endometriosis. [356] Other colorectal lesions may occasionally masquerade as polyps. Such lesions include hemorrhoids, condylomata, [357] enlarged anal papillae, fibrous anal polyps, inverted diverticula, [358] enteroenteric and ureteroenteric anastomoses, [359] and the residual stalk of a pedunculated polyp after polypectomy. [360] Inverted diverticula are important to recognize and not misdiagnose because “polypectomy” can result in perforation. They are rather flat and a little distensible, often with a slight central umbilication.

SCREENING FOR ADENOMAS

Detection of asymptomatic colorectal adenomatous polyps is achieved in the settings of colorectal cancer screening and postcancer and postadenoma surveillance, and as an incidental finding during colonoscopy for other reasons of early detection of colorectal cancer. [96], [361], [362], [363], [364], [365], [366], [367], [368] and [369] These issues are discussed in detail in Chapter 34 and Chapter 91.

Fecal Occult Blood Testing

FOBT is relatively ineffective at detecting polyps, especially small ones. On the other hand, polyps detected as a result of this type of screening are usually large, [236] and large polyps are known to bleed.

Sigmoidoscopy

Ten percent of colorectal neoplasms are located in the rectum, and an additional 35% to 45% occur in the sigmoid colon (see Table 89-3). The standard rigid sigmoidoscope has a potential depth of insertion of 25 cm, although on average, a 17- to 20-cm depth of insertion is achieved. A 60-cm flexible sigmoidoscope could achieve an average length of insertion of 48 to 55 cm and thus should detect between 50% to 70% of all colorectal neoplasms. [370], [371] and [372] In general, flexible sigmoidoscopy provides a two to three times higher yield of adenomas than rigid sigmoidoscopy. Flexible sigmoidoscopy, however, is more expensive, requires more skill, and takes longer to perform than rigid sigmoidoscopy. Nevertheless, it is preferred over rigid sigmoidoscopy because of its higher yield and greater patient comfort and acceptance. [373], [374]

Only one controlled trial of screening sigmoidoscopy has been reported; this demonstrated a mortality benefit for sigmoidoscopy when combined with other cancer-protective interventions, but the contribution from sigmoidoscopy is difficult to determine. [375] Two case-control studies have shown that patients who develop fatal distal colorectal cancer are 60% to 70% less likely to have had a rigid or flexible sigmoidoscopy in the previous 10 years compared with controls without cancer. [376], [377] Those studies are also somewhat difficult to interpret because it was not clear that the procedure led to more polypectomies, necessary for a direct impact on colorectal cancer. A large randomized controlled trial of flexible sigmoidoscopic screening for colorectal cancer has commenced in the United Kingdom, but results are not yet available. [378]

Flexible sigmoidoscopy alone will not detect all colorectal cancers. Even allowing for the fact that some proximal cancers will have distal adenomas signaling their presence, almost 80% of proximal cancers and significant adenomas will not be suspected at flexible sigmoidoscopy. [379] In later studies by Lieberman and associates, [369] nearly two thirds of asymptomatic patients with advanced proximal neoplasia had no adenomas in the rectum and sigmoid colon, and over half of the same patients had no adenomas distal to the splenic flexure. In a parallel study reported by Imperiale and colleagues, [370] 46% of patients with advanced proximal neoplasia had no distal polyps; however, there was a gradient of risk for proximal advanced neoplasia in patients with distal hyperplastic polyps (relative risk [RR], 2.6), distal adenomas (RR, 4.0) and distal advanced neoplasia (RR, 6.7) as well as with older age (RR, 1.3 per 5 years) and male sex (RR, 3.3).

Radiography

Use of barium enema for the screening of asymptomatic, average-risk people in large populations has not been reported. On the other hand, a place for virtual colonoscopy (spiral or multislice computed tomography scanning after bowel preparation) is emerging in screening. Accuracy is still less than with colonoscopy, but the procedure is safe, and costs are comparable with colonoscopy. [380], [381] and [382]

Colonoscopy

Although colonoscopy is highly sensitive and specific (when combined with histopathology) for colorectal neoplasia, its expense, discomfort, and potential complications can be prohibitive. In the absence of proof from randomized clinical trials, it is recommended by few national authorities as a screening tool for average-risk, asymptomatic people, although such trials are now in pilot phase. [383] In a study of 3121 veterans (mean age, 62.9 years) neoplastic lesions were found in 37.5%, with advanced lesions (more than 9-mm adenomas, villous lesions, high-grade dysplasia, or invasive cancers) in 10.5%. There were 30 cancers. Distal adenomas were a significant although nonspecific marker of proximal advanced disease. [371] Colonoscopy is considered an appropriate screening tool for selected high-risk groups: individuals who have, on the same side of the family, multiple first-degree relatives or a first- and second-degree relative with colorectal cancer, a single first-degree relative diagnosed before reaching 55 years of age, [41], [44], [231], [384], [385], [386] and [387] family members at risk for HNPCC, patients with longstanding ulcerative colitis, and those with a previous history of adenoma or cancer.

Several studies show only a modest increase (twofold) in lifetime risk for colorectal cancer in first-degree relatives of single-case families in which the index case is older than 55 years of age at diagnosis. [40], [388] Colonoscopic surveillance studies addressing family risk have rarely stratified their results with respect to the age of the index case. One has shown only a nonsignificant trend to higher yields when the index case was younger than 60 years of age. [231] Another showed a significant increase in adenoma incidence compared with controls, only when the index case was younger than 65 years of age. [389] Other than in HNPCC and FAP, relatively low yields of advanced neoplastic lesions in those undergoing colonoscopic surveillance and younger than 50 years of age regardless of the risk category have been consistently observed, because age is the dominant determinant of risk (yield). [231], [386], [387], [390], [391] Many authorities advise commencing screening at 50 years, or 10 years younger than the earliest age of onset in the family, whichever is earliest. A case-control study of endoscopy or barium enema experience before diagnosis of cancer, however, indicated a 50% reduction of risk with use of sigmoidoscopy, colonoscopy, or barium enema, a protection that extended for 6 years. [392]

DIAGNOSIS OF ADENOMAS

Digital rectal examination, FOBT, and sigmoidoscopy were discussed earlier and are also discussed in Chapter 34 and Chapter 91. In the symptomatic patient, their use is limited.

Radiography

Single-contrast barium enema has been shown to have about 75% sensitivity and specificity for detection of polypoid lesions [393], [394]; it is better with double-contrast (air-contrast) barium enema. [395], [396] Even so, most studies have found that double-contrast barium enemas miss a proportion of polypoid lesions subsequently detected at colonoscopy. [395], [397], [398] and [399]

Colonoscopy

When a patient is clinically suspected of having a colorectal neoplasm, colonoscopy is the procedure of choice (see earlier and Chapter 140). Colonoscopy allows detection, diagnosis, and removal of adenomas in most instances. Colonoscopy itself is safe, even in young children. [398], [400], [401], [402] and [403] Most complications are due to polypectomy (see “Polypectomy as Therapy”).

Colonoscopy is highly accurate for polyp detection but not infallible. [404], [405] and [406] Miss rates for adenomas of any size have been estimated at 17% from studies of paired colonoscopies performed within 120 days. [407] Three of 46 adenomas larger than 9 mm were missed in this study. This may be a biased estimate because the reason for performing the second colonoscopy may have been the suspicion of inadequate colonoscopy the first time. In a back-to-back tandem colonoscopy study, there was a 24% miss rate overall, with 6% of the adenomas larger than 1 cm being missed. [14], [408] In a study at St. Mark’s Hospital in London, colonoscopy and double-contrast barium enema were performed in tandem; 92% of polyps larger than 7 mm were detected at colonoscopy, compared with 74% by double-contrast barium enema. [409]

Small rectal tubular adenomas are not considered to represent a risk factor for future colorectal cancer [410] (Table 89-7) and are weak but significant predictors of proximal advanced lesions [367], [368], [411]; thus, one could question whether full colonoscopy is needed when these are detected. The question is somewhat academic because polyp removal requires an adequate bowel preparation to ensure safety (eliminating risk for explosion from electrocautery), and thus the patient is already prepared for total colonoscopy. The evidence that the future risk is low comes from sigmoidoscopic series in which the polyps were initially completely removed. [410] Complete removal is critically important. Indeed, most of the cancers found in an earlier series from the same institution occurred in incompletely removed adenomas, albeit mostly villous in histology.

	RELATIVE RISK*	95% CI
Single tubular adenoma <1 cm	0.5	0.4–1.3
Single TVA, VA, or TA >1 cm	3.4	2.3–4.8
Multiple adenomas	7.7	3.7–14.2

*Risk assessed from 1618 patients at St. Mark's Hospital, diagnosed in the pre-colonoscopy era.

TVA, tubulovillous adenoma; VA, villous adenoma; TA, tubular adenoma.

Data adapted from ref. 410.

TABLE 89-7 Risk for Colorectal Cancer After Polypectomy from Rectum or Sigmoid Colon

Colonoscopy or Radiography?

In general, colonoscopy is 10% to 12% more accurate than air-contrast barium enema and gives fewer false-positive results. [398], [399], [406], [412], [413] In addition, it allows for polypectomy and biopsy of all suspect lesions. Diverticular disease, redundant loops of colon, and poor coating or residual feces compromise the interpretation of barium enema examinations, particularly in patients older than 50 years of age, who are the ones most likely to have a neoplasm. [409]

When directly compared in a double-blind study, colonoscopy detected 95% of large adenomas, whereas barium enema detected only two thirds. [409] Comparing the diagnostic experience in more than 2000 patients with colorectal cancer, barium enema was 85.2% and consecutive colonoscopy 95% accurate. [413] Patient discomfort with barium enema is about the same as with colonoscopy.

The accuracy of both procedures, however, depends on the expertise of the examiner, and quality assurance programs should be maintained for both. Also, there are patients in whom colonoscopy cannot be completed to the cecum for anatomic or clinical reasons. In these patients, the air-contrast barium enema or virtual colonoscopy is required to detect proximal lesions (Fig. 89-16). Radiography should, therefore, be considered complementary to colonoscopy.

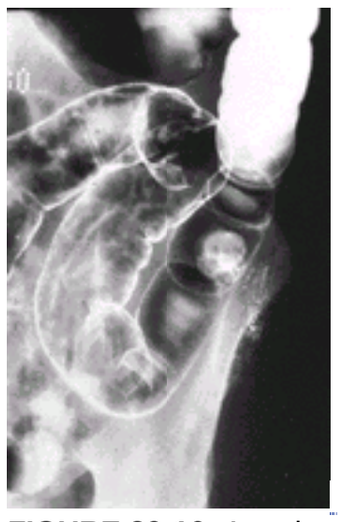

FIGURE 89-16. A pedunculated polyp demonstrated by double-contrast radiography. Note the lobulated appearance of the polyp surface. The mass was subsequently found at polypectomy to be a tubulovillous adenoma.

Concerning virtual colonoscopy, the ideal technique is still being developed; bowel preparation and technological refinements need further exploration before conclusions can be reached about how it compares with colonoscopy. Virtual colonoscopy has not been compared head to head with barium enema.

Biopsy and Histopathological Examination

Biopsy or polypectomy of a polypoid lesion is necessary because histopathological analysis is the definitive diagnostic test. Pathology review with the pathologist [414] is necessary to optimize biopsy and polypectomy procedures as well as handling of specimens. Separate specimen containers should be used when specimens are obtained from different sites; otherwise, lesions may be interchanged, and it may be difficult or impossible to determine which of several anatomically distinct lesions is the one showing adenomatous changes or malignancy on histological examination. Marking the site of a polyp with sterile India ink may help subsequent localization, especially if surgery is needed for malignancy within a polyp. Alternatively, a metal clip can be attached to the mucosa at the site.

NATURAL HISTORY /H3>

Nonneoplastic Polyps and Submucosal Lesions

The natural history depends on the type of lesion. Most nonneoplastic polyps are removed by endoscopic polypectomy for definitive diagnosis. Metastatic lesions, such as malignant melanomas and lymphomas, are aggressive tumors. Other lesions, such as lipomas and fibromas, usually pose no clinical concern.

Adenoma Growth and Malignant Conversion

Because of their potential premalignant nature, understanding the natural history of adenomatous polyps is of major clinical importance. Unfortunately, the natural history, growth rate, and likelihood of progression to malignancy of adenomas are still not completely understood.

Not all adenomas are destined to grow. A study using serial sigmoidoscopy to follow asymptomatic tattooed rectal polyps smaller than 1 cm (not all of which would have been adenomas) found that over the course of 3 to 5 years, only 4% of the polyps increased in size, 70% remained unchanged, 8% were smaller, and 18% disappeared spontaneously. [415], [416] Studies using serial barium enemas also suggest that most polyps (undefined type) grow very slowly, if at all. [417], [418]

A retrospective study examined the risk for colorectal carcinoma at the site of an adenoma 1 cm or larger left in situ. Altogether, 226 patients were followed for an average of more than 5 years with an average of five barium enemas per patient. During the course of follow-up, 37% of the polyps increased in size, colorectal carcinomas developed at the site of the index polyp in 10% of the patients, and colorectal carcinoma developed at sites at a distance from the index polyp in another 5%. The cumulative risk for carcinoma at the site of a polyp 1 cm or larger was estimated to be 3% at 5 years, 8% at 10 years, and 24% at 20 years. [210]

A prospective follow-up of the natural history of 215 unresected polyps less than 5 mm in diameter has been reported. At polypectomy 2 years later, about 50% were found to be hyperplastic polyps, 25% were mucosal polyps, and only 25% were adenomatous polyps. The course was variable. Usually, mucosal polyps regressed, and the adenomas and hyperplastic polyps enlarged, with a doubling in size in the 2-year period; however, no polyp was significantly larger than 5 mm, and no high-grade dysplasia or cancer was found. [419]

Using the available data on the growth rate of adenomas and carcinomas and including information on average age of diagnosis of adenomas by size and level of dysplasia and cancer, [190], [415] a mathematical model has suggested that for those that grow, it takes 2 to 3 years for a small adenoma (less than 0.5 cm) to grow to 1 cm in size and another 2 to 5 years for the 1-cm adenoma to progress to carcinoma. [419], [420] Invasion and metastasis probably takes another 2 years, and on average, death occurs 2 years after this. These results, when taken together, suggest that most adenomatous polyps enlarge only slowly if at all and that those most likely to progress to malignancy are those that have reached 1 cm in size.

If one compares adenoma prevalence data in autopsy studies (see earlier, 40% to 50% prevalence) with lifetime cumulative incidence of colorectal cancer (of about 4% to 5% in most Western countries), after allowing for multiplicity of adenomas, it can be deduced that only about 1 of 20 adenomas is destined for malignancy.

Adenoma Recurrence after Removal

In the National Polyp Study conducted in the United States, the rate of occurrence of new adenomas in patients followed by periodic colonoscopy and polypectomies was about 27.5% (a significant proportion were probably missed synchronous polyps) at 1 year and 32% at 3 years. These results suggest a new adenoma rate of about 5% per year on average in those who have had an adenoma, not allowing for missed adenomas. Those with more than two adenomas or severe dysplasia were more likely to have advanced neoplasia at follow-up, as were men older than 60 years of age with a family history of colorectal neoplasms. [399], [421] In this study, neither size nor histology was predictive of advanced lesions. All studies on risk for metachronous adenomas identify multiplicity as a risk factor for metachronous adenomas, whereas many show size as an independent predictor as well. [422], [423], [424] and [425]

Malignant Polyps

Severe dysplasia or “intramucosal carcinoma” within a polyp is not considered to be cancer because it does not breach the muscularis mucosae. Lymphatics in the colon do not extend beyond the muscularis mucosae into the lamina propria, and hence the possibility of spread is negligible. There have not been any confirmed reports of lymphatic invasion resulting from severe dysplasia or intramucosal carcinoma. [212], [426], [427] and [428]

Evaluation of invasiveness comes from pathological examination of a completely removed polyp and serial sectioning of it; otherwise, malignancy may be missed. [212] If the neoplastic lesion invades through the muscularis mucosae (with exposure to the lymphatics), the adenoma is then termed a *carcinomatous* or *malignant polyp*. [428] These may metastasize.

Misplaced epithelium (pseudoinvasion) may be difficult to distinguish from invasive carcinoma. In general, the displaced epithelial cells are surrounded by lamina propria and not by desmoplastic reaction and are similar to the overlying adenomatous epithelium. [346], [429] Hemosiderin deposition in the polyp indicates torsion.

THERAPY

Chemoprevention of Adenomas

Therapy may be directed at prevention of adenoma recurrence in the case of sporadic adenomas or FAP, or at regression of existing adenomas in FAP when removal or resection is to be avoided, such as small duodenal polyps and polyps in retained rectum after colectomy.

Dietary factors have been discussed under “Primary Prevention”—in reality, the interventional evidence comes from prevention of new adenoma formation, and it is assumed that the effect would be the same in those who have never had an adenoma.

At this stage, the most promising agents for chemoprevention are the nonsteroidal antiinflammatory drugs.

Nonsteroidal Antiinflammatory Drugs

Most evidence now indicates that use of nonsteroidal antiinflammatory drugs, specifically aspirin, is associated with reduced mortality from colorectal cancer. [220], [221], [222], [223], [224], [225], [226], [227], [228] and [229], [430] This effect extends to adenomas as seen in the Nottingham screening study [224] and, as a nonsignificant trend, in the Australian Polyp Prevention Project. Sulindac has been shown to be effective in reversing rectal adenoma growth in patients with FAP after ileorectal anastomosis. [225], [226] and [227] The European CAPP-1 and CAPP-2 trials are evaluating aspirin in randomized controlled trials in FAP and HNPCC, respectively. Celecoxib is effective in FAP. [431]

Evidence to date does not support chemoprevention with aspirin or sulindac in patients with sporadic (common) adenomas. [228], [229], [432] Use of celecoxib for sporadic adenomas, if proved of value, may achieve prevention without side effects that would be associated with aspirin or other cyclooxygenase-1–inhibiting agents.

polypectomy as Therapy

Endoscopy and snare polypectomy provide a simple and cost-effective means of managing colorectal polyps. [8], [433], [434] and [435] Cold snare polypectomy or garroting of small polyps is effective. [436] In practice, all polyps should be considered for removal because it is not possible to determine the histological type by visual appearance. The number of polyps that can be safely removed by polypectomy depends on their location and size.

Rectal polyps that are smaller than 5 mm, also referred to as *diminutive polyps* usually are benign and rarely produce bleeding. They may be left in place if they are multiple and appear flat and hyperplastic. At least three of them should be removed for confirmatory histological examination. [437], [438] They should all be subsequently removed if any is found to be an adenoma. Although hot biopsy [439], [440], [441] and [442] and bipolar electrocoagulation [440] have been advocated for eradication of diminutive polyps, these techniques can leave behind residual polyp (adenomatous) tissue [441], [442] and are more dangerous in the thin-walled cecum.

Polyps larger than 1 cm are usually adenomatous and should be removed in toto if pedunculated. If sessile, they may need to be removed piecemeal. [443], [444] Histological evaluation of the presence or degree of invasion of any carcinomatous foci could then be difficult or impossible, especially because the chance of malignancy increases with increasing size. [190] If there is any concern that residual polyp tissue may be left, the polypectomy site should be tattooed with sterilized India ink to facilitate follow-up evaluation. [445] This should also be done if there is a high index of suspicion that a polyp may be malignant. If polyps are inflammatory, hyperplastic, or otherwise nonadenomatous by histological examination, complete removal is unnecessary, but a sampling of several polyps from each cluster is required.

Multiple polyps require particular care. In patients with newly diagnosed FAP, colectomy, not polypectomy, is indicated. For multiple adenomatous polyps not due to FAP, when there is the possibility of removing them endoscopically, the colon should be cleared of polyps a section at a time to reduce the chance of complications. Note that the thinner wall of the cecal area and right colon requires care when removing polyps through cautery. [444], [446], [447]

Polypectomy is safe in adults [400], [401] and children [402] when performed by experienced physicians. Complications are generally related to polypectomy and the major complication rate is less than 2%. The requirement for hospitalization or surgical intervention (due usually to hemorrhage or perforation) is less than 0.3%, and death is rare. [400], [401] Polypectomy should be deferred in patients on anticoagulation and in those with severe bleeding diatheses, unstable cardiac arrhythmias, recent myocardial infarction, acute colitis, pregnancy (second or third trimester), recent colonic surgery, or abdominal abscess or perforation. [447] Some consider that aspirin should be ceased 1 week before colonoscopy in case polypectomy is required, but others do not; although the issue is unresolved, the risk would appear to be very low.

Removal of sessile polyps can be assisted by submucosal injection of saline (2 to 10 ml) to lift the polyp from the muscular wall and minimize the risk for perforation. Kits to facilitate this are commercially available.

The procedural aspects of colonoscopy and polypectomy, including patient preparation, premedication, antibiotic prophylaxis, polypectomy, and polyp retrieval techniques, are discussed in Chapter 140.

Laser Ablation and Argon Plasma Coagulation

Ablation with the neodymium:yttrium-aluminum garnet (Nd:YAG) laser or argon plasma coagulation are alternative methods for the rapid removal of large numbers of small polyps from segments of the colon. [46], [448], [449], [450], [451] and [452] In experienced hands, it is efficacious, rapid, and relatively safe; complications occur in about 5% of cases. [452] Perforation is rare, but extra care is needed in the thin-walled cecal area. [451] Such ablation should be restricted to the removal of multiple polyps after the histological diagnosis has been confirmed by previous biopsies or polypectomies. Laser ablation is also useful in the rectal stump of patients with FAP and in patients with cancer or large adenomas who are not surgical candidates, in whom it often results in rapid symptomatic relief. [452] Argon plasma coagulation has appeal because it is a nonlaser technique and does not require the rigorous training associated with the use of clinical lasers. It can also be applied tangentially, which is helpful in the colon and rectum. The depth of ablation is naturally limited to 3 to 5 mm, adding a potential increment of safety over lasers. The disadvantages, of course, are that laser and argon plasma ablation are destructive and do not yield specimens for pathologic evaluation. It is also possible that a focus of invasive malignancy in the polyp may be missed.

Surgical Resection

In cases in which a polyp is large and sessile or otherwise cannot be removed by endoscopic polypectomy, surgical resection may be necessary. Surgical resection is also indicated for some adenomas with invasive carcinoma (malignant polyps; see later). [453], [454] and [455] Large rectal adenomas may be extirpated by transanal excision using an operating sigmoidoscope and rigid snare under anesthesia or with an operating microscope in association with a sigmoidoscope. Other large polypoid lesions and malignant polyps need to be removed by laparotomy or laparoscopy. [456] Once laparotomy is indicated, many surgeons advocate the use of a formal segmental colectomy, including lymph node dissection. The extra resection adds little anesthesia time or perioperative mortality and morbidity. [455]

Severe Dysplasia and Intramucosal Carcinoma

Severe dysplasia and intramucosal carcinoma are nonmalignant adenomas and can be cured by adequate polypectomy.

The progression of severe dysplasia to invasive malignancy demands careful management and complete removal. Evaluation of a removed polyp by fragmental biopsy or piecemeal polypectomy is difficult. [118], [190], [453], [457] The presence of cancerous cells on a biopsy or cytology specimen may not reveal the malignant nature of a polyp without the architectural information provided by polypectomy.

Malignant Polyps

Definition and Prognosis The term *malignant polyp* refers to that situation in which severely dysplastic cells have penetrated the muscularis mucosae. [457] Care must

be taken to distinguish this from misplaced epithelium (pseudoinvasion; see earlier). [347], [429] The management of malignant polyps remains controversial. [435] There is general agreement that poor prognostic features include incomplete polypectomy, poorly differentiated carcinoma, and malignant cells within 2 mm of the polypectomy margin. [458], [459] and [460] The presence of venous or lymphatic invasion is an independent poor prognostic factor in some series. [121], [461], [462] Lymphatic invasion is, however, subject to observer variability. [463] Malignant polyps with venous (or lymphatic) invasion are almost always associated with higher-grade cancers, and when they occur in isolation from the other prognostic variables, they need to be taken into account with respect to the overall context of the patient's co-morbidity. [464] Controversial prognostic factors are the replacement of the bulk of the adenoma by carcinoma, sessile nature, and large polyp size. Sessile malignant polyps are generally considered to have a worse prognosis, and most usually have other poor-risk markers. [465] Of those with pedunculated polyps, most have invasion limited to the head, neck, or stalk.

Results of Removal Of 62 evaluable patients with malignant polyps who had endoscopic polypectomy, 4 (7%) had recurrence. [462] In other studies, patients who underwent endoscopic polypectomy for pedunculated malignant polyps did better. [466] In a retrospective review of 43 patients with pedunculated malignant polyps who were followed for almost 5 years, 19 patients initially underwent colonic resection, but none from either treatment group had a recurrence. [467] Most data indicate that successfully removed polypoid carcinomas do not spread and therefore do not require segmental resection unless poor prognostic features are present. [468] These guidelines, however, are derived from incomplete studies, and it is difficult to be rigid in their application. For the individual patient, it is necessary to weigh the estimated risks of surgery against the risks of leaving residual malignancy in situ after endoscopic polypectomy of a malignant polyp. The presence of poor prognostic features is generally accepted as a reason for colectomy (see Table 89-7), [118], [212], [410], [458], [459] and [460] but age of the patient and any coexistent morbidity should always be considered. The decision depends on an estimate of the risk that residual but localized cancer is present compared with the risk for perioperative mortality and morbidity in the individual patient. [469] Operative mortality rates range from 2% to 10%, increasing with the age of the patient. [455], [470] Because most patients with malignant polyps are in their seventh and eighth decades of life, the risk of colectomy is significant. Based on current evidence, [118], [190], [200], [457], [458], [459], [460], [461], [462], [463], [464], [465], [466], [467], [468], [469], [470] and [471] the following recommendations for management of malignant polyps can be made (Fig. 89-17):

1. A pedunculated malignant polyp with invasion of the head, neck, or stalk is adequately treated by polypectomy alone if the polypectomy was not piecemeal, the cancer was not poorly differentiated, and the resection margin was free of cancer for 2 mm.
2. Malignant polyps with involvement within 2 mm of the margin of resection or with nonassessable margins, polyps with invasion of the submucosa of the bowel wall (especially if there is less than 2 mm margin of clearance), and polyps containing poorly differentiated invasive carcinoma should be treated by colectomy unless the surgical risk is prohibitive. For sessile malignant polyps, invasion of the submucosa is highly likely, and surgery should usually be undertaken. Blood vessel and lymphatic involvement may influence a decision to perform colectomy but not strongly.
3. Malignant polyps treated by polypectomy alone should be followed by repeat colonoscopy within 3 to 12 months to assess for recurrence at the polypectomy site.

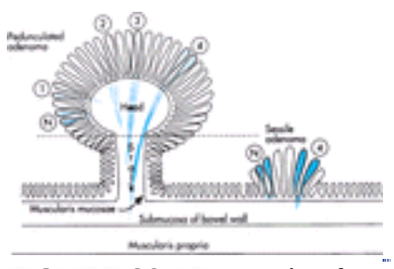

FIGURE 89-17. Levels of carcinomatous extension in malignant polyps—pedunculated versus sessile adenomas. When carcinomatous cells are contained within the mucosa, without breaching the muscularis mucosae and so extending into the "head" of the stalk, the lesions are known as noninvasive carcinomas, otherwise referred to as severe focal atypia or intramucosal carcinoma (*N*). Once cancer has invaded through the muscularis mucosae, the polyp is then termed a malignant polyp. Because lymphatics are present in the submucosa, metastatic spread is then possible. Increasing levels of invasion of the carcinoma, to the head (*1*), neck (*2*), stalk (*3*), and base of the stalk or submucosa (*4*), may carry worsening prognostic implications. A malignant sessile adenoma has, by definition, invaded the submucosa of the bowel wall (*4*). Noninvasive carcinomas (*N*) require no additional therapy after polypectomy. Usually, malignant polyps with invasions only of the head, neck, and stalk (*1, 2, 3*) and with clear resection margins (at least 2 mm) and no poor prognostic features (see Table 89-7) are adequately treated by polypectomy alone. In malignant polyps with invasion of the submucosa of the bowel wall (*4*) or with poor prognostic features, colectomy should be seriously considered for potential cure.

In all cases, patients must be informed of the rationale behind the physician's therapeutic decision and should participate in that decision. Patients should be aware that evidence-based guidelines are not available for the treatment of malignant polyps. [435]

FOLLOW-UP FOR METACHRONOUS ADENOMAS AND CANCERS

Current recommendations for following patients with adenomatous polyps vary, [435] and several prospective studies are addressing this issue. The current St. Mark's Neoplastic Follow-up Study is reexamining patients after previous polypectomies for adenomatous polyps at three different time intervals (1, 3, and 5 years) and randomized after stratification for risk. [472]

Predicting Recurrence of Adenomas

In the initial St. Mark's Neoplastic Follow-up Study, age, multiplicity of previous adenomas, and size of largest adenoma were independent risk factors for further development of clinically significant (metachronous) adenomas or cancer. [423]

These prognostic risk factors for adenoma recurrence have been confirmed in colonoscopic follow-up studies such as the U.S. National Polyp Study. In addition to multiplicity (more than two), the U.S. study also identified family history in association with older age (more than 60 years) at diagnosis of adenomas as a risk factor for future advanced neoplasms. This study critically found that an interval of 3 years after initial, complete polypectomy for adenomas was as effective as follow-up colonoscopy after 1 year. [215]

Rectal epithelial proliferation has been shown in some studies to predict future adenomas. [473]

Longer intervals might be equally effective, especially in those patients with small tubular adenomas in the rectosigmoid region. Although definitive data are not available, follow-up intervals extending to 10 years are being recommended for low-risk adenomas. [410], [421], [425], [435] The yield in such patients appears to be less rewarding than that in subjects without prior adenoma who are older than 60 years of age. [376], [377], [421]

The role of FOBT in adenoma surveillance remains unclear. FOBT is better than no follow-up at all and could be used in intervening years to maintain annual contact with patients and aid detection of rapidly developing interval cancers or large adenomas.

Predicting Future Occurrence of Cancer

Factors predicting future cancer are similar. In 1618 patients followed for a mean period of 14 years after initial sigmoidoscopy and polypectomy for rectal adenoma, the risk for subsequent rectal cancer depended on size, histological type, and multiplicity, with a size greater than 1 cm and villous architecture being the strongest risk factors (see Table 89-7). Conversely, patients with small tubular rectal adenomas without severe dysplasia had no increased risk for colon cancer. [410]

Patients who have undergone surgical resection of a primary colorectal cancer are not only at risk for recurrence of their first cancer but also at a threefold to fourfold increased risk for metachronous adenomas or cancer. [29], [30], [474] Patients with cancers, either sporadic or in HNPCC, with MIS are at particularly high risk. Therefore, follow-up should seek not only evidence of metastatic disease and recurrent disease at the surgical anastomosis but also development of a second primary cancer or an adenoma. If the entire length of the colon was not completely visualized before surgery (owing to tumor obstruction or poor preparation), a complete colonoscopy to look for synchronous lesions is indicated after 3 to 6 months. After the colon has been documented to be free from neoplasms, initial follow-up may be at 3 years,

as in the follow-up after polypectomy for adenomatous polyps. [475] Examining the anastomosis for local recurrence in the first 2 years after left-sided cancer is also advisable.

Surveillance Recommendations

When an adenoma is found, surveillance colonoscopy after 3 years suffices provided that the colonoscopist has reasonable confidence that polypectomy is complete and that the entire colon has been cleared of polyps. [399], [421], [425] If not, repeat within 3 to 6 months is indicated. If the repeat surveillance colonoscopy is clear of adenomas after 3 years, some consider that the interval may be lengthened to 5 years, although there is no good evidence to support this. For patients at higher risk for colorectal cancers, such as those at risk because of hereditary nonpolyposis colorectal cancer, more frequent examinations may be in order. On the other hand, patients with only small tubular adenomas in the rectum are not at increased risk for colorectal cancer and can be dismissed from follow-up or offered repeat examination at 5 years. [410]

The appropriate surveillance recommendations for patients with acromegaly are uncertain. If surveillance is to be undertaken, it should commence within a few years of diagnosis and be repeated at about 3-year intervals. [59]

IMPACT OF POLYPECTOMY ON CANCER INCIDENCE AND MORTALITY

The expectation is that introduction of polypectomy in the community should reduce the incidence and mortality from colorectal cancer. This has been, to date, difficult to document in national statistics. Trends in colorectal cancer incidence vary around the world, with a general trend upward in countries that follow or are adopting a Western lifestyle. [129] Yet despite the availability of screening and colonoscopic surveillance, the trend continues upward in some countries. One such country is Australia; the age-adjusted upward trend is 2% per annum in men and 1% per annum in women since the 1970s. [476] The continuing increase may be due to environmental factors that drive up the incidence despite polypectomy or may be due to an increase in early diagnosis of cancers ahead of an anticipated later decrease in cancer diagnoses. Fortunately, mortality has remained stable (age adjusted).

In the United States, however, improvements in national mortality statistics and apparently incidence are evident. [477] This improvement is multifactorial, including earlier diagnosis; better surgery, anesthesia, and postoperative care; and the impact of adjuvant therapy.

Four large-scale adenoma follow-up studies have examined the impact of colonoscopic surveillance on subsequent cancer development [215], [478], [479] and [480] (Table 89-8). The London, Danish, and Australian studies each showed that the incidence of cancer during follow-up was not significantly different from the incidence in age- and sex-matched population controls, implying a reduction from a higher risk if surveillance was not undertaken. Unfortunately, without control groups, the real significance is unclear. The U.S. National Polyp Study demonstrated a statistically significant reduction in risk in participants within the study compared with either population controls or an independent (but historical) population of adenoma patients who did not have colonoscopic follow-up (see earlier). This is the best study demonstrating effectiveness.

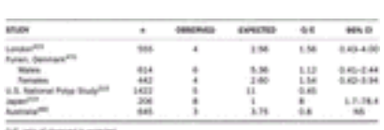

TABLE 89-8 Relative Risk for Colorectal Cancer in Subjects Undergoing Surveillance for Adenomas, Compared with Age- and Sex-Matched Control Groups

Acknowledgments

We wish to acknowledge assistance from Dr. Tony Thomas with provision of Figure 89-4 , Figure 89-5 , Figure 89-9 , Figure 89-10 , and Figure 89-11 , and the original contribution made to this chapter by Dr. Gordon Luk, the author of the first edition. Some of the structure and materials, including some figures, from the original chapter have been retained.

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CHAPTER 90

Randall W. Burt and Russell F. Jacoby

POLYPOSIS SYNDROMES

The polyposis syndromes are a group of conditions in which multiple gastrointestinal (GI) polyps occur in the lumen of the gut (Table 90-1). Most of these syndromes are inherited, and most exhibit an increased risk for colon cancer. Many benign and malignant extraintestinal manifestations are also observed. The polyposis syndromes have traditionally been categorized according to polyp histology. Recent elucidation of the responsible genes has allowed refinement of the categorization as well as a more precise understanding of the phenotypes and cancer risks. The clinical importance of the syndromes relates to their inheritance as well as to their benign and malignant manifestations. Cancer prevention is often possible with proper management of patients and their families. The syndromes are also of great importance to biologic investigation. Recent elucidation of their genetic and molecular etiologies has contributed immensely to the understanding of cancer pathogenesis and cell biology.

Familial adenomatous polyposis
Gardner syndrome
Two thirds of Turcot syndrome families
Attenuated adenomatous polyposis coli
Syndromes with hamartomatous polyps
Peutz-Jeghers syndrome
Juvenile polyposis
Cowden syndrome
Bannayan-Riley-Ruvalcaba syndrome
Hereditary mixed polyposis syndrome
Gorlin syndrome
Polyposis syndromes with neural polyp histology
Neurofibromatosis type I
Multiple endocrine neoplasia type IIB
Isolated intestinal neurofibromatosis of the colon
Polyposis syndromes of uncertain etiology
Cronkhite-Canada syndrome
Hyperplastic polyposis syndrome
Polyposis syndromes with inflammatory polyps
Inflammatory bowel disease
Devon polyposis
Cap polyposis
Polyposis conditions arising from lymphoid tissue
Nodular lymphoid hyperplasia
Multiple lymphomatous polyposis
Immunoproliferative small intestinal disease
Miscellaneous noninherited polyposis syndromes
Leiomyomatosis
Lipomatous polyposis
Multiple lymphangiomas
Pneumatosis cystoides intestinalis

TABLE 90-1 Polyposis Syndromes

FAMILIAL ADENOMATOUS POLYPOSIS

Familial adenomatous polyposis (FAP) is the first described, the most common, and the best characterized of the polyposis syndromes. The first reports of FAP appeared in the medical literature in 1861 and 1873. [1], [2] The hereditary nature of this disease was recognized by 1882, and cases of associated malignancy were published in 1887 and 1890. The autosomal-dominant inheritance pattern of the disease was clearly established by 1934. [3]

FAP is now known to be an inherited condition characterized by hundreds to thousands of colonic adenomatous polyps that most often begin to emerge in the second and third decades of life. Colon cancer is inevitable if the colon is not removed. The condition includes Gardner syndrome, about two thirds of Turcot syndrome families, and attenuated adenomatous polyposis coli (AAPC), also called attenuated FAP. FAP and its variants arise from mutations of the adenomatous polyposis coli (APC) gene. Inheritance is autosomal dominant with nearly complete penetrance. Synonymous terms for FAP include *adenomatous polyposis coli* and *familial polyposis coli*.

Epidemiology

Estimates of the prevalence of FAP vary from 1 in 6850 to 1 in 31,250 people (2.29 to 3.2 cases per 100,000 population). [1], [4], [5] The frequency is fairly constant throughout the world, with men and women being affected equally. Up to one third of newly diagnosed cases, those not belonging to previously identified families, appear to represent new mutations. [1] About 0.5% of all colon cancer cases historically have arisen in the setting of FAP, [4] although the figure may now be as low as 0.07%, possibly owing to screening in identified FAP families. [5]

Genetics

***APC* Gene** FAP arises from germ-line mutations of the *APC* gene on the long arm of chromosome 5 (5q21–q22). The *APC* locus was identified in 1987. [6], [7] Four years later, the gene itself was identified and characterized. [8], [9], [10] and [11] The gene has been shown to be alternatively spliced in multiple coding and noncoding regions. The main transcript has 15 exons with 8532 base pairs (bp) that code for 2844 amino acids and result in a 311.8-kd protein. Exon 15 is large and constitutes more than three fourths of the coding region of the gene. The APC protein localizes to the nucleus and to the membrane and cytoskeleton in human epithelial cells. [12] It also homodimerizes [13] and binds to other proteins, including glycogen synthase kinase (GSK)-3ß, ß-catenin, ?-catenin, tubulin, EB1, and hDLG. [14] Mutational inactivation of the *APC* gene is not only responsible for FAP but also appears to be an early, if not the first, step in the pathogenesis of more than 80% of all colonic adenomas and colon cancers [14] (see Chapter 91). In FAP, one allele is inherited in a mutated form, and the mutation is present in every cell of the colon (and body). Adenoma formation is initiated when the second allele is damaged or lost by a somatic event. In sporadic adenomas, both alleles must be damaged or lost by somatic events for adenoma formation to begin. Thus, patients with FAP form large numbers of adenomas at a young age, whereas only one or several sporadic adenomas occur in a patient with sporadic polyps and at a much later age. Genes that require both alleles to be lost for disease to occur are called *tumor suppressor genes* of which *APC* is a prime example. The progression of adenoma to carcinoma after *APC* inactivation is similar in FAP and the sporadic setting, with accumulation of mutations in additional relevant genes, including K- *ras* and *p53* a gene or genes on chromosome 18 and possibly others [14], [15] (see Chapter 91).

***APC* Gene Function** An important function of the APC protein is control of cell growth through the Wingless and Wnt signaling pathway. [16] Activation of the pathway favors cell growth and suppression of apoptosis. In the nonstimulated state of this pathway, GSK-3ß forms a complex with APC, ß-catenin, and axin and phosphorylates these proteins. The phosphorylation of ß-catenin targets it for ubiquitin-mediated proteasomal degradation. A stimulus to the Wnt pathway causes uncoupling of ß-catenin from the complex, loss of phosphorylation of ß-catenin, and a resultant increase in cytoplasmic concentrations of this protein. ß-Catenin then translocates to the nucleus, where it interacts with the transcription factor, T-cell factor and lymphoid-enhancing factor (TCF-Lef). This transcriptional activation causes an up-regulation of a number of genes, including the oncogenes c- *Myc* and cyclin D1, and also matrilysin, c- *jun*, *fra-1*, urokinase-type plasminogen activator receptor, and the peroxisome proliferator-activated receptor-d (PPAR-d). These genes and possibly others presumably mediate the cell growth and antiapoptotic effects of the Wnt signaling pathway. Mutation of the *APC* gene results in a defective complex of GSK-3ß, APC, and axin. The complex is unable to bind or phosphorylate ß-catenin. This in turn gives rise to a persistent increase in cytoplasmic ß-catenin concentration and therefore a constitutive or constant activation of the entire Wnt signaling pathway. The APC protein also binds to microtubules and can promote microtubule assembly in vitro. [17], [18] It has been postulated that this association is part of the normal migration mechanism of the colonocyte up the colonic crypt. [19], [20] Mutated *APC* would then cause disruption of normal cellular positioning in the crypt, consistent with present models of carcinogenesis. A recent finding of great interest is that the APC protein has been shown to accumulate at the kinetochore during mitosis, contribute to kinetochore-microtubule attachment, and play a role in chromosome segregation in mouse embryonic stem cells. [21], [22] The APC protein itself may thus play a role in the chromosomal instability, seen as chromosomal loss of heterozygosity, that is often observed when APC function is lost.

***APC* Mutations in Familial Adenomatous Polyposis** More than 90% of APC germ-line mutations that cause FAP give rise to a truncated protein product. The causative mutations are insertions, deletions, and nonsense mutations that lead to either frame shifts and downstream premature stop codons or de novo premature stop codons. [23], [24] More than 825 germ-line mutations have been found to date in families with FAP. [25] More than 80% of these mutations are different and distinct for each family. The most common germ-line *APC* mutation observed in multiple unrelated families is a 5-bp deletion that results in a frameshift mutation at codon 1309. Although mutations have been found scattered throughout the gene, they are predominantly located in the 5' or proximal end of the gene. The number of colonic adenomas in FAP correlates to some extent with the location of the APC germ-line mutation (Fig. 90-1). AAPC is associated with mutations in the 5' part of the gene (5' to codon 158), [26], [27] and [28] in exon 9, [29], [30] and [31] and in the distal 3' end of the gene (3' to codon 1596). [30], [32], [33], [34], [35], [36] and [37] Profuse or dense polyposis has been reported with mutations in codons 1250 to 1464, with less dense polyposis proximal and distal to this location. [38], [39] As stated previously, the most frequent FAP mutation is at codon 1309. One study found that patients with mutations at that codon presented with symptoms at an average age of 20 years, whereas patient with mutations between codons 168 and 1580, excluding 1309, presented with symptoms an average of 10 years later. [40] Patients with mutations proximal to codon 168 and distal to codon 1580 presented at an average age of 52 years.

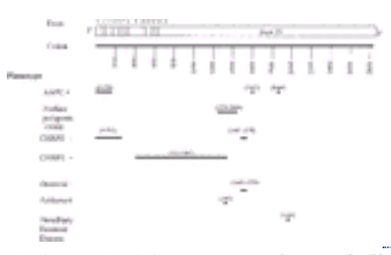


FIGURE 90-1. Location of disease causing mutations in the *APC* gene correlated with phenotypic manifestations. Courtesy of Cindy Solomon, M.S.

Correlations have also been found between the locations of the mutations in the *APC* gene and extracolonic manifestations of FAP (see Fig. 90-1). Congenital hypertrophy of the retinal pigment epithelium (CHRPE) is observed with mutations between codons 463 and 1387, [41] although mutations between codons 1444 and 1578 lack CHRPE expression. [42], [43] Thyroid cancer associates with *APC* mutations in the 5' end of exon 15. [44] A higher incidence of desmoid tumors is found with mutations between codons 1444 and 1578. [42], [43] A study of 269 patients with information on the presence or absence of desmoids and identified *APC* mutations found a wide but uneven distribution of mutations in patients with desmoids. [40] Desmoids occurred in 20% of patients with mutations proximal to codon 1444 but in 49% of patients with mutations distal to this codon. Sixty-one per cent of patients with mutations between codons 1445 and 1580 exhibited desmoid tumors, whereas only 18% had desmoids when the mutation was distal to that region. A number of studies have indicated that desmoid tumors occur more frequently with mutations in the 3' end of the *APC* gene and specifically distal to codon 1444. [43], [45], [46] and [47] Another study found that individuals with mutations between codons 1395 and 1493 had significantly higher rates of desmoids, osteomas, and epidermoid cysts compared with those with mutations between codons 177 and 452 and significantly more desmoids and osteomas compared with those with mutations between codons 457 and 1309. [48] None of the 79 patients with mutations between codons 177 and 452 developed osteomas, hepatoblastomas, periampullary region tumors, or brain cancers, whereas hepatoblastomas and brain cancers were seen only in patients with mutations between codons 457 and 1309. The correlation of germ-line mutation location with polyp density or extracolonic manifestation is not consistent, however, and identical mutations can be associated with differing phenotypes. [49], [50] Modifying genes and additional mutations of the *APC* gene are two hypotheses that might explain these observations. Finally, a polymorphism of the *APC* gene unique to people of Ashkenazi Jewish descent has also been identified. [51] This polymorphism, I1307K, is present in 6% of Ashkenazi people but in 12% with colon cancer and 29% with colon cancer and a family history of colon cancer. It confers an approximate 1.5- to 2-fold increased risk for colon cancer compared with the general population and is believed to cause disease by increasing the mutability of the *APC* allele on which it resides. [51], [52], [53] and [54]

Laboratory Genetic Testing Methods The laboratory methods used for genetic testing in FAP include protein truncation testing (PTT), gene sequencing, and linkage testing. [55] PTT exploits the observation that most *APC* mutations in FAP cause premature termination of the APC protein. The method examines for truncation of the APC protein in vitro and thus also ensures that a mutation is disease causing. [56] It must be understood, therefore, that PTT infers, but does not specifically identify, the disease causing mutation. PTT is successful in identifying evidence for a disease causing mutation in 80% to 90% of families with clinically defined FAP. Once a disease-causing mutation is inferred by PTT in an index case (a family member known to have FAP), other family members can be tested for the presence or absence of this mutation with near 100% accuracy. The cost is $750 to $1000 to examine for an unknown mutation and about $500 per person in relatives once a mutation is known to be present. DNA sequencing is more accurate than PTT, with the specific disease causing mutation being identified in more than 90% of families with known FAP. Once the mutation has been identified in an index case in the family, other family members can be tested for the presence or absence of that specific mutation by one of a number of highly accurate polymerase chain reaction–based methods. The accuracy for mutation finding in relatives at this stage is near 100%. A drawback with sequencing is that it is sometimes difficult to know whether a certain mutation is disease causing or a harmless polymorphism. Cost for sequencing is $800 if the mutation is not known and about $200 once the mutation is known. Additional laboratory genetic methods are often used in initial mutation finding, including denaturing gradient gel electrophoresis and single-strand conformation polymorphism testing. These methods can often narrow the area in which a mutation is found. They are thus frequently used in combination with PTT and sequencing to limit the area of DNA for specific mutation finding. Linkage testing employs DNA markers to track the segregation of a purported mutation through a family. The sensitivity of linkage in identifying genetically affected individuals approaches 99% because of the highly informative markers now available. Linkage is limited, however, in that two or three known affected people involving two generations must be available for sampling to apply the method successfully. Linkage is thus usually applied only when PTT or sequencing fail. The cost is about $245 to $260 per person tested.

Clinical Process of Genetic Testing Genetic testing is usually done on DNA obtained from the white blood cells of peripheral blood. A number of genetic testing laboratories now perform genetic testing for FAP and other inherited conditions. These laboratories can be identified at a website (http://www.genetests.org/). Genetic counseling is an essential prelude to genetic testing. [57] A number of important medical and psychosocial issues surround asymptomatic genetic testing. Children's rights are of particular concern when testing in children is being considered. All of these issues must be thoroughly understood by the physician and patient when genetic testing is anticipated. Informed consent for testing is then required before testing is done. [58] The physician must either be familiar with the issues or use trained genetic counselors. Details of genetic counseling are given in Chapter 54. There are three settings in which to consider genetic testing for FAP [59]: (1) testing

a person with some features of FAP, but where the clinical diagnosis is not certain; (2) testing a person with clinically defined FAP, but the mutation is not known in the family; and (3) testing relatives in an FAP family once the mutation is known. The first setting is usually defined as a person with at least 20 cumulative adenomas but no known family history of FAP. Finding a mutation confirms the diagnosis of FAP and allows relatives to be tested with high accuracy. Failure to identify a mutation does not rule out FAP, however, because initial mutation finding is 90% successful at best. Genetic testing in the second situation is similar, but the patient has more than 100 adenomas. Failure to find a mutation means that all close relatives must still be screened as if they have the condition because it was already clinically defined in one case. In the third setting, relatives of a person with a known APC disease mutation are tested for the presence or absence of that mutation. A positive test indicates the diagnosis of FAP, whereas a negative test (absence of mutation) essentially rules it out. Such individuals then need only average risk screening for colon cancer.

Clinical Manifestations

Colonic Polyposis The clinical diagnosis of FAP is made with the presence of at least 100 colonic adenomatous polyps or fewer polyps and an immediate relative with FAP [1] (Fig. 90-2 and Fig. 90-3; Color Fig. 90-2). The average number of adenomatous polyps in a person with fully expressed FAP is 1000, with some people exhibiting more than 5000 polyps. Fewer adenomas are seen early in the development of the disease and in AAPC (see later), with further subdivisions of expected polyp number now known to correlate with APC mutation location. [39]

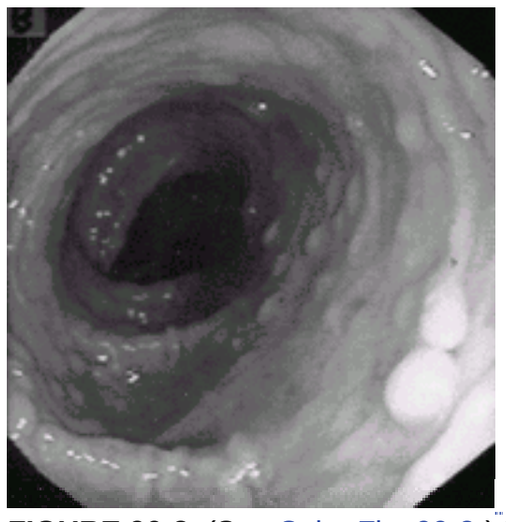


FIGURE 90-2. (See Color Fig. 90-2.) Early colonic polyposis in familial adenomatous polyposis.

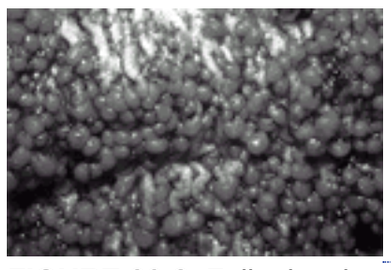


FIGURE 90-3. Fully developed colonic polyposis in familial adenomatous polyposis. Courtesy of Karen Kim, M.D.

Polyps most often begin to appear in the second or third decade of life. The mean age of polyp occurrence assessed by prospective rigid sigmoidoscopy is 15.9 years (range, 8 to 34 years), [60] although occurrence of polyps at younger and older ages has been reported. The average age of FAP diagnosis in patients presenting with symptoms is 35.8 years (range, 4 to 72 years). [1] The age of symptom presentation correlates with gene mutation location, which, in turn, correlates with colonic polyp density. [39], [40] Adenomatous polyps are usually distributed evenly throughout the colon, with a slight distal colonic excess. The size of the polyps depends on the stage at which the patient is examined. Even in fully developed cases, however, 90% of adenomas are less than 0.5 cm in diameter, and less than 1% of polyps are larger than 1 cm. Polyps may either carpet the colon with myriad small lesions or occur as more distinct and somewhat larger lesions. These patterns appear to have a genetic origin (see earlier), although striking heterogeneity of polyp number and growth rate also has been observed. [49], [61], [62] Histopathology demonstrates tubular adenomas, indistinguishable from common or sporadic adenomas. Villous and tubulovillous histologies are also seen, but much less frequently and usually in larger polyps. A histologic feature of FAP not observed in the general population is dysplastic or adenomatous epithelial cells in single crypts or even portions of single crypts. These are called *microadenomas* and are often seen in biopsy specimens of normal-appearing flat mucosa. [1] Budding of dysplastic epithelium from normal crypts is also observed. Aberrant crypt foci also have been reported to occur with increased frequency in FAP. [63] These lesions are similar to microadenomas but are identified with methylene blue staining of the colonic mucosal surface.

Colon Cancer Adenocarcinoma is the inevitable consequence of FAP unless the colon is removed. In the St. Mark's series, the average age at cancer diagnosis was 39 years. By 45 years of age, 87% had developed cancer, and by 50 years, 93%. Multiple colonic malignancies were present in about 48% of those with cancer (41% synchronous, 7% metachronous). Eighty-four percent of malignancies were at or distal to the splenic flexure, a fraction almost identical to that found in their series of random colorectal malignancies at that time. Average life expectancy after diagnosis of cancer was 2.6 years. Colon cancer has been reported as early as 9 years of age, although the occurrence of malignancy before adolescence is very unusual. [64] Polyp number correlates with cancer risk. A recent study found a 2.3-fold increase in cancer risk at diagnosis for those with more than 1000 colonic polyps compared with those with fewer than 1000 polyps. [65] Each 10-year older age group also exhibited a 2.4-fold increase in cancer risk. There remains a 25% incidence of colon cancer in newly diagnosed FAP patients, most likely due to the high rate of new mutations. [66]

Upper Gastrointestinal Polyps and Cancer

Stomach. Gastric polyps occur in 23% to 100% of FAP patients [67], [68], [69] and [70] (Fig. 90-4). In the gastric fundus and body, the polyps are most often nonneoplastic fundic gland polyps, considered hamartomas. These polyps are histologically seen to consist of simple hyperplasia of the fundic glands with microcysts. Endoscopically, the polyps are multiple sessile lesions, 1 to 5 mm in diameter, that are the same color as surrounding mucosa. Considerable variation in size and number is observed. The polyps are sometimes so numerous that they coalesce, forming areas of irregular, matted surface mucosa. Fundic gland polyps rarely cause symptoms. They have been observed to occur as early as 8 years of age. Size and number may gradually increase, remain static, or even decrease. [71] Fundic gland polyps may rarely progress to cancer. [72], [73] and [74] Similar polyps, although fewer in number, also are sometimes observed in the general population and possibly with chronic use of proton pump inhibitors. [75], [76]

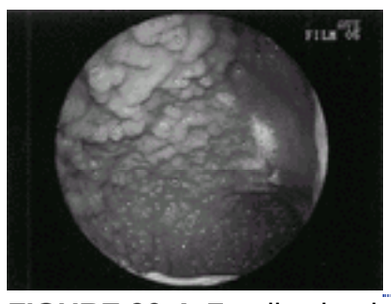


FIGURE 90-4. Fundic gland polyposis in familial adenomatous polyposis.

Adenomatous polyps occur in the stomach of about 10% of patients with FAP. They are most often confined to the antrum but are occasionally found in the body and fundus. [68], [77] The lifetime risk for gastric cancer in FAP is about 0.6%, believed in large part to arise from adenomatous polyps.

Duodenum and small bowel. Endoscopically visible duodenal adenomas are found in 50% to greater than 90% of FAP patients, and microadenomas are frequently observed. [70] The polyps are most commonly 1 to 5 mm in diameter and multiple. Most patients have numerous small adenomas scattered throughout the duodenum, particularly in the second and third areas, whereas in others, the polyps are restricted to the periampullary area. Adenomas may progress, often slowly, and there is evidence of an adenoma-carcinoma sequence similar both histologically and genetically to that observed in the colon. [70] Adenomas may sometimes grow large, exhibit villous histology and increasing degrees of dysplasia, and may cause symptoms. A scoring system has been developed to evaluate the severity of duodenal polyposis and is now widely applied as the *Spigelman staging system* [78] (Table 90-2).

POLYPS	1 POINT	2 POINTS	3 POINTS
Number	<4	5–20	>20
Size	0–4 mm	5–10 mm	>10
Histology	Tubular	Tubulovillous	Villous
Dysplasia	Mild	Moderate	Severe

Stage I, 4 or fewer points; stage II, 5–6 points; stage III, 7–8 points; stage IV, 9–12 points.
Adapted from ref. 78.

TABLE 90-2 Duodenal Adenomatosis Staging System

The lifetime risk for duodenal cancer, which most often occurs in the periampullary area, is 3% to 5%. [1], [70] The age of duodenal cancer diagnosis ranges from 17 to 81 years, with a mean between 45 and 52 years. Duodenal cancer is one of the leading causes of death in FAP patients who have had prophylactic colectomy. No consistent association between mutation location and the presence or severity of duodenal polyposis has been made, [70] although mutations downstream from codon 1051 have recently shown association with severe periampullary lesions. [79] One study also found that the occurrence and severity of lesions at the papilla segregated in certain FAP families. [80] The frequent clustering of adenomas and cancer in the periampullary area suggest that bile or pancreatic secretions may play a role in carcinogenesis, and a number of mechanisms have been suggested. [70], [77] Adenomas may occur throughout the small bowel but are concentrated for the most part in the proximal jejunum (50% of cases) and distal ileum (20% of cases). [81], [82] Malignancy is unusual, but small bowel cancer has been reported. Adenomatous polyps are also known to occur, possibly with increased frequency postoperatively, in the distal ileum after subtotal colectomy, colectomy with ileostomy, and colectomy with ileoanal pull-through. Distal ileal cancer also has been reported in these settings, but it is very unusual. Prominent lymphoid polyps may occur frequently in the terminal ileum of younger patients with FAP and should be differentiated from adenomas by biopsy.

Gallbladder, bile ducts, and pancreas. Both adenomatous change and cancer have been reported in the gallbladder, bile ducts, and pancreas. [67], [81], [83], [84] Lesions in these locations appear to be unusual, but their exact frequency is unknown. Biliary and pancreatic duct obstruction have arisen from both benign and malignant lesions.

Benign Extraintestinal Growths

Osteomas and dental abnormalities. Multiple osteomas were the first extracolonic lesion to be associated with colonic polyposis. [1], [85], [86] These lesions are benign bone growths found most commonly on the skull and mandible but may occur on any bone of the body. The size ranges from imperceptible to several centimeters in diameter, and they number from one to dozens. Osteomas may occur in children who are at risk for FAP before the onset of colonic polyposis, [87] but they can continue to occur throughout life. [88] Osteomas have no malignant potential and are not a clinical problem except for occasional cosmetic concern. Subtle radiopaque jaw lesions are often evident by panoramic dental radiographs in patients with FAP and no other apparent extraintestinal lesions of Gardner syndrome. [89], [90] Various dental abnormalities also have been described in FAP, including unerupted teeth, supernumerary teeth, dentigerous cysts, and odontomas. [86], [87], [91] These all may precede the development of colonic polyposis, and the prevalence is estimated at 17%, compared with 1% to 2% in the general population.

Cutaneous lesions. Epidermoid cysts, sebaceous cysts, fibromas, and lipomas have all been described as a part of FAP. [87], [92], [93], [94], [95] and [96] Epidermoid cysts occur most commonly on the legs, face, scalp, and arms, in that order, but they may appear anywhere on the body surface. [94] They range in size from millimeters to several centimeters. Although common in the general population, in FAP, they often occur before puberty, may precede the onset of polyposis, and occur at an average age of 13 years. Fibromas, ranging in size from millimeters to centimeters, are found most commonly on the cutaneous surface of the scalp, shoulders, arms, and back. The cutaneous lesions may cause cosmetic problems, and cysts occasionally become infected.

Desmoid tumors. Desmoid tumors (also called *desmoid fibromatosis*) are benign fibrous growths that occur rarely in the general population (5 to 6 per 10 [6] per year [97]) but frequently in FAP, affecting between 3.6% and 20% of patients. [98], [99] and [100] About 2% of all desmoids arise in patients with FAP, although the relative risk for the tumor in patients with FAP compared with the general population is 825. [101] The peak incidence of desmoids in FAP is between 28 and 31 years, although they may occur at any age. Independent predictors for desmoids include an *APC* mutation 3' of codon 1444 (see earlier section " Genetics"), family history of desmoids, female gender, and the presence of osteomas. [45] Desmoid tumors are one of the most common causes of mortality in FAP patients who have had prophylactic colectomy, with a 10% to 50% mortality rate from the tumor in those with desmoids and a 10-year survival rate of 63%. Desmoids may arise in musculoaponeurotic structures throughout the body, although they are most common in the abdomen. [100] They do not metastasize but may infiltrate adjacent structures, extend along facial planes, attach to and erode bones, and may engulf and compress blood vessels, nerves, ureters, and other hollow organs of the abdomen. Severe and even fatal clinical problems are sometimes caused by these tumors, especially if mesenteric vessels or other abdominal organs are obstructed. In the abdomen of FAP patients, mesenteric plaquelike lesions occur that may progress to mesenteric fibromatosis and finally desmoid tumors, often stimulated by surgery. [102] About half of abdominal desmoids are intra-abdominal, whereas the other half occur in the tissues of the abdominal wall. [98] Desmoid tumors usually enlarge very gradually and sometimes stop growing altogether. Only about one third of abdominal desmoids cause pain, although the most common symptom is abdominal pain. Intra-abdominal desmoids sometimes become massive, occupying much of the abdominal cavity and encasing many segments of viscera. These tumors may be the first manifestation of FAP in some patients and families. Furthermore, some families with APC mutations exhibit desmoids as their only disease manifestation. [47], [103] A distinctive histopathology has been demonstrated in desmoid tumors of young FAP patients, whose first manifestation of the condition was often this lesion. [104] Desmoids in FAP are monoclonal growths of hyperproliferative fibroblastic cells. [105] Excess cell growth appears to occur on the basis of *APC* gene inactivation and accumulation of ß-catenin in the cells. [106] The relationship of desmoids in FAP to sporadic cases is uncertain in view of the distinctive early histopathology [104] and the observation that *APC* gene mutations are uncommon in sporadic desmoids. [107]

Congenital hypertrophy of the retinal pigment epithelium. Multiple and bilateral patches of CHRPE, also called *pigmented ocular fundus lesions*, have been described as a common manifestation of FAP. [108] The lesions are discrete, darkly pigmented, round, oval, or kidney shaped and range in size from 0.1 to 1.0 disc diameters. The presence of bilateral or multiple (more than four) lesions is highly specific (94% to 100%) but only moderately sensitive (58% to 84%) for FAP. The lesion appears to be congenital, has been detected in a 3-month-old at-risk infant, and may be the earliest clinically detectable lesion of FAP. Careful slit-lamp examination is often required for detection. The presence of CHRPE correlates closely with mutations in specific areas of the *APC* gene (see earlier section " Genetics"). [41], [42] and [43], [109], [110] and [111]

Adrenal adenomas. Adrenal adenomas have been reported in FAP. [112], [113] Two studies found their occurrence to be significantly higher than in the general population, 7% and 13%, respectively, versus 3%. [114], [115] Several cases of functioning adrenal adenomas and adrenal carcinomas have also been noted in these same series, but any association is uncertain. Most adrenal masses in FAP are found incidentally on computed tomography (CT) done for other reasons. It has been recommended that adrenal lesions be managed similarly to those in the general population, which is generally hormonal investigation if relevant symptoms are present and needle biopsy if a simple adenoma is not indicated by CT or the lesion is 5 cm or larger. [114], [115]

Nasal angiofibroma. Nasal angiofibromas have been correlated with FAP in a number of patients. [116]

Extracolonic Malignancies A number of malignancies outside the colon are known to exhibit an increased lifetime risk in patients with FAP compared with the general population. These include duodenal and periampullary (3% to 5%), thyroid (2%), pancreatic (2%), gastric (0.6%), central nervous system (CNS; less than 1%), hepatoblastoma (1.6%), and possibly small bowel distal to the duodenum and adrenal. The most important are duodenal and periampullary because of their frequency. These and gastric cancer were discussed earlier under "Upper Gastrointestinal Polyps and Cancer." Thyroid cancers affect about 2% of individuals with FAP, have been reported in 112 FAP patients, [117] and exhibit a relative risk compared with the general population of 7.6 (95% confidence interval [CI], 2.5 to 17.7). [118] The mean age of diagnosis is 28 years, ranging from 12 to 62 years. A female preponderance is apparent, and the histology is predominantly papillary, commonly with a cribriform pattern. Familial aggregation has been observed, and mutation analysis of 24 patients revealed that most mutations identified were 5' to codon 1220. The relative risk for pancreatic adenocarcinoma in the John's Hopkins FAP registry compared with the general population was 4.46 (95% CI, 1.2 to 11.4). [118] The same study found no cases of biliary or adrenal cancer and no increased risk for lung or breast cancer. Hepatoblastoma affects 0.75% to 1.6% of children with FAP, exhibits a male predominance, and associates somewhat with mutations in the 5' end of the *APC* gene. This malignancy most often occurs in the first 5 years of life, with some risk up to 15 years of age. The risk for hepatoblastoma in children with FAP is 800-fold greater than in the general population. [119], [120]

Syndromes Related to Familial Adenomatous Polyposis

Gardner Syndrome In the early 1950s, Gardner and colleagues described a large kindred that exhibited typical FAP, but also very prominent extracolonic growths, especially osteomas, epidermoid cysts, and fibromas. [86] All of the other extracolonic lesions noted previously were eventually added to the phenotype of Gardner

syndrome. Discovery of the *APC* gene led to the knowledge that FAP and Gardner syndrome arise from mutations of that gene. Further, the expression of extracolonic manifestations correlates to some extent with location of the mutation in the gene, [121] although gene modifiers may still play a role. [122] The term *Gardner syndrome* is thus mainly of historical interest but will likely continue to be used for families with frequent and obvious extraintestinal manifestations of the condition.

Turcot Syndrome Turcot syndrome is characterized by colonic polyposis and CNS tumors. [123] About two thirds of Turcot syndrome cases arise from mutations of the *APC* gene and exhibit medulloblastoma-type CNS tumors as well as some anaplastic astrocytomas and ependymomas. [124], [125] Such cases are thus a variant of FAP and usually exhibit typical colonic polyposis. The other third of cases come from mutations of the mismatch repair genes and are thus a variant of hereditary nonpolyposis colorectal cancer (HNPCC). The CNS malignancies in HNPCC-related Turcot syndrome are glioblastoma multiforme tumors, and affected patients also exhibit fewer adenomatous polyps, typical of HNPCC. [125] The risk for cerebellar medulloblastoma in FAP is calculated to be 92 times that of the general population. [124] The occurrence of CNS malignancies is not a completely random event in FAP, however, because 40% of families with brain tumors include more than one affected person. *APC* mutation location correlations have not been made with CNS tumors.

Attenuated Adenomatous Polyposis Coli AAPC, also called attenuated FAP, arises from *APC* mutations at either the extreme proximal (5') parts of the *APC* gene, [26], [27] and [28] at the extreme distal (3') portion of the gene, [30], [32], [33], [34], [35], [36] and [37] or in certain parts of exon 9. [29], [30] and [31] Patients with AAPC have many fewer adenomas than those with typical FAP, often averaging about 30 and exhibiting a proximal colonic preponderance. Equally notable, however, is that the polyp number is extremely variable in many kindreds, with some members of the same kindred having a small number of adenomas whereas others have numbers approaching that of typical FAP. [26], [30], [31], [37] The lifetime colon cancer risk has been estimated to be about 80%, with the age at diagnosis about 50 years [37] and an average age at symptomatic presentation of 52 years. [40] The emergence of adenomas is also delayed about 10 years in my experience. The expression of upper GI polyps, however, both gastric and duodenal, does not appear to be attenuated in number, age at emergence, or cancer risk. [30], [36], [72], [74] AAPC may be difficult to diagnose without genetic testing because some patients exhibit very few adenomas, often proximal colonic, even at advanced ages.

Differential Diagnosis

All of the polyposis syndromes reviewed in this chapter are included in the differential diagnosis of a patient found to have GI polyposis. HNPCC must occasionally also be distinguished, particularly from AAPC. Histology is paramount to specific diagnosis because most of the syndromes have histologically characteristic polyps. The colonoscopic appearance of myriad 1- to 5-mm polyps is characteristic of FAP, although lymphoid hyperplasia and hyperplastic polyposis (HP) may mimic this appearance, emphasizing the importance of histology. Additional parameters to be considered in differentiating the various polyposis conditions include polyp distribution throughout the GI tract; polyp number and size; the presence of extraintestinal growths, lesions, or malignancy; and the family history of any of these features. Finally, genetic testing is a powerful tool that can now be used to make a definitive syndrome diagnosis in most cases, further allowing genetic testing in family members when the relevant mutation is found. Each of the polyposis syndromes in this chapter is characterized in terms of these parameters. The features that differentiate the polyposis syndromes are summarized in Table 90-3 and Table 90-4.

TABLE 90-3 Distinguishing Features of Familial Adenomatous Polyposis and the Hamartomatous Polyposis Syndromes

TABLE 90-4 Additional Conditions that Exhibit Gastrointestinal Polyposis

Clinical Course and Complications

Most individuals with FAP remain asymptomatic until colon cancer occurs. Adenomatous polyp development is slow and insidious. There are usually only a few, minute polyps when adenomas are first detected. Polyp number and size increase gradually over ensuing years. Symptoms from the disease are seldom experienced if a timely colectomy is performed. Left untreated, nonspecific symptoms eventually develop from the polyposis or the inevitable colon cancer. The most common symptoms include rectal bleeding (79%), diarrhea (70%), and abdominal pain (40%). [1] The average age at onset of symptoms is 32.8 years. Sixty-six percent of individuals who present with symptoms have already developed cancer. Age of symptom development can now be predicted somewhat by *APC* mutation location (see earlier), although these observations have little clinical applicability. Presymptomatic screening and appropriate prophylaxis are the favored approach in FAP.

Upper GI polyps are usually asymptomatic until cancer occurs, with the exception of polyps at the duodenal papilla or in the bile or pancreatic ducts, which may cause obstructive jaundice or pancreatitis. Symptoms from polyps and cancer are nonspecific and include abdominal pain, weight loss, GI blood loss, palpable mass, and lumenal obstruction.

The average life expectancy of patients with untreated FAP has been estimated to be 42 years but approaches that of the general population up to 18 years after colectomy. [126] The major causes of death after colectomy are duodenal cancer (particularly periampullary), desmoid tumors, and rectal cancer if a subtotal colectomy was done. [127], [128] and [129] Other cancers are occasionally observed, such as pancreatic, biliary, adrenal, and small bowel. The major causes of death in postcolectomy patients from one study were desmoid tumors in 11 of 36 patients (31%); periampullary cancer in 8 (22%); rectal cancer in 3 (8%); adrenal cancer in 1 (3%); and carcinomatosis in 1 (3%). [127] The remainder died of causes not directly related to FAP. Those with desmoid tumors died an average of 6.6 years after colectomy, whereas those with periampullary cancer died an average of 23.1 years after surgery. The average age at death for the entire group was 41.6 years. This must not be equated with overall life expectancy because only patients who had died were studied. The cumulative probability of an FAP patient developing extracolonic cancer of any type after colectomy in the St. Mark's Registry was found to be 11% by 50 years of age and 52% by 75 years, although most of the malignancies, like in other series, were in the periampullary region. [130] This overall risk was significantly higher than the compared risk in the general population of about 4% and 22% at ages 50 and 75 years, respectively.

Management

Diagnosis The diagnosis of FAP is made by finding characteristic clinical manifestations, specifically more than 100 colonic adenomas at any age; finding adenomas in a younger first-degree relative of a person with known FAP; and finding a disease-causing mutation in the *APC* gene in a person suspected of having FAP. A constellation of other findings, such as typical upper GI polyps and osteoma, can lead to a high suspicion of FAP. Thus, diagnosis virtually always involves colonoscopic examination with biopsy of the colon and frequently genetic testing. The indications and clinical process of genetic testing were outlined in the earlier section "Genetics."

Screening and Surveillance The primary goal of FAP management is to prevent cancer. In view of the extreme cancer risk in the colon and emerging knowledge of other cancer risks, empiric screening guidelines have been suggested on the basis of these risks and the likely ages of cancer development (Table 90-5).

TABLE 90-5 Cancer Risks and Screening Recommendations in Familial Adenomatous Polyposis

Colon screening. Colon screening with subsequent surgery has been definitively shown to decrease mortality from large bowel malignancy. A study from the Swedish National Polyposis Registry that included 431 FAP patients from 145 families found a mortality rate of 44% in self-presenting FAP patients compared with 1.9% in relatives who were called up for screening. [131] The Finnish polyposis registry found survival in postcolectomy FAP patients who had undergone screening because of a relative with known FAP to be equivalent to that in the general population and statistically much lower than that in FAP patients presenting with symptoms up to 18 years after colectomy. [126] Colon screening should be performed in those with a genetic diagnosis of FAP or in first-degree relatives of those with FAP if genetic testing is uninformative or has not been done. Generally accepted colon screening guidelines for typical FAP are annual sigmoidoscopy beginning at 10 to 12 years of age. [55], [132], [133] Screening intervals can be increased each decade and can change to average-risk screening at 50 years of age if polyposis is not present. [55] Those initially screened at an older age should probably have colonoscopy for the first examination. If surgery is delayed longer than a year after polyps emerge, annual colonoscopy should be considered for surveillance. For AAPC, colonoscopy should always be used for screening in view of polyp distribution but can reasonably be delayed until the late teens to mid-20s.

Upper gastrointestinal screening. Upper GI screening has not been demonstrated to improve prognosis but is nonetheless recommended in view of the cancer risk and expectation that mortality can be improved. The most commonly given recommendation is to begin screening at the age of 20 to 25 years, although some recommend an initial examination at the time of diagnosis and earlier screening if other family members have exhibited advanced duodenal disease at an earlier age. [70] Standard upper endoscopy should be supplemented with a side-viewing instrument to visualize the duodenal papilla properly. Endoscopic ultrasound or endoscopic retrograde cholangiopancreatography may also be indicated, depending on endoscopic findings or symptoms. A 1- to 3-year interval for reexamination is given as follows: (1) every 3 years for Spigelman stages I and II disease; (2) every 2 years for stage III disease; and (3) every 6 to 12 months for stage IV disease [70] (see Table 90-2). Another approach is every-3-year endoscopy if adenomas are not found and annually if they are. It would certainly seem reasonable to examine the stomach during endoscopy and to biopsy any polyps of concern because of size, color, or gross appearance. One study found that most patients with advanced duodenal and periampullary findings exhibited *APC* mutations downstream from codon 1051, [79] although more work is needed before this observation can be applied in the clinical setting.

Screening for other malignancies and desmoids. Screening for the other associated cancers and for desmoids is entirely empiric. The thyroid should be examined annually, starting at age 10 to 12 years, in view of risk and ease of examination. Some argue that other sites should be evaluated only for symptoms or if these cancers have occurred in relatives. Screening is not done for desmoids, but evaluation is done only for palpable masses or symptoms. At my institution, in addition to that described earlier, patients are presented with the option of abdominal CT with oral contrast every 3 years to examine the pancreas, small bowel, and adrenal glands, starting at age 20 to 25 years. Careful questioning is done for CNS symptoms and periodic head magnetic resonance imaging is recommended if any family member has had CNS malignancy. Pediatric gastroenterologists do a careful liver palpation annually during the first decade. [132] Specific biliary evaluation is done only for symptoms or issues raised on surveillance endoscopy. The small bowel is specifically examined periodically by x-ray or push enteroscopy if upper endoscopy demonstrates severe duodenal polyposis.

Ileal and rectal screening after colectomy. Adenomas may develop in the ileal pouch after colectomy with ileoanal pull-through surgery, or they may develop from rectal epithelium left at the time of such surgery. [82], [134] Whether there is risk for cancer in ileal pouch adenomas is uncertain, although cases have been reported. [135] Screening of the pouch should be done within 2 years after surgery and then every 6 months to 3 years, depending on the presence and number of adenomas. When subtotal colectomy with ileorectal anastomosis is performed, rectal adenomas often regress, [136] possibly because of changes in bile acid concentrations. [137] They almost always recur, however, and are associated with a substantial cancer risk. The most recent evaluation of cancer risk in the rectum after subtotal colectomy indicates a 40-year risk of 0.32. [138] Chronologic age was the only independent risk factor, but cancer was more common when *APC* mutations occurred in areas that give rise to dense polyposis. Sigmoidoscopy with ablation of recurrent adenomas is believed to minimize and delay cancer occurrence and is suggested every 3 to 6 months, depending on the rate of adenoma recurrence. [82] Ablation techniques include electrocautery, laser, heater probe, and argon beam coagulation.

Treatment

Colon and rectum. An appropriately timed colectomy remains the keystone of cancer prevention in FAP. It should be considered once adenomas emerge. Most centers prefer to wait until after high school if colonic adenomatosis remains minimal, although delay has been associated with some cancer occurrence, particularly in FAP patients older than 20 years of age. [139] Surgical options include: (1) subtotal colectomy with ileorectal anastomosis, and (2) total colectomy with mucosal proctectomy, ileal pouch construction, and ileoanal pull-through. Colectomy with ileostomy is rarely needed. Subtotal colectomy is a single-stage procedure with slightly less morbidity than the pull-through surgery, but in view of rectal cancer risk, it remains an option only for patients and families with few rectal adenomas, particularly when AAPC is the diagnosis. [66], [138] Mutation location may help in determining choice of procedures, but adenoma expression is quite variable, even with identical mutations, making clinical expression of polyps the determining factor in surgical decisions. [40] Despite rectal screening, the cumulative risk for secondary proctectomy to control polyposis or treat cancer in those with a remaining rectum was 0.70 at 40 years of follow-up in one study, although it was never necessary in patients with AAPC. [138] In my experience, some patients with AAPC can be managed for many years with colonoscopic polypectomy of millimeter-sized polyps and may possibly never need colectomy. In general, this approach should only be considered with AAPC patients who exhibit fewer than 5 to 10 1-mm sized adenomas at each examination and probably fewer than 30 cumulatively. Even in such patients, however, caution is important in view of cancer occurrence when colectomy is delayed. [139] Nonsteroidal antiinflammatory drugs (NSAIDs) have not been studied in this situation, and although they would be expected to reduce adenoma number, their effect on cancer risk is unknown. A number of studies have demonstrated that NSAIDs induce colonic adenoma regression in FAP. [140], [141], [142] and [143] It is presumed but not proven that cancer risk is also diminished. Equally important, polyp ablation is considerably easier in patients with a remaining rectum because NSAID therapy often minimizes the number of new adenomas encountered during follow-up examinations. A selective cyclooxygenase-2 inhibitor, Celebrex, has, in fact, been approved by the U.S. Food and Drug Administration for polyp regression therapy in FAP patients who have had a subtotal colectomy with a remaining rectum. [144] The published studies would suggest, however, that the effect of Celebrex is modest compared with sulindac. Developmental agents that would minimize the upper GI side effects of NSAIDs in different ways are now under study, including an agent that is the sulfone derivative of sulindac, exisulind. [145] This agent has no antiprostaglandin activity but appears to retain the polyp regression effect of the parent compound. The use of NSAIDs to prevent the emergence of adenomas in patients with a genetic diagnosis of FAP or to delay colectomy in those with polyposis is not yet recommended, although investigations addressing these issues are underway.

Upper gastrointestinal tract. Intervention for gastric polyps is unusual. Adenomas should be endoscopically resected if possible. The development of high-grade dysplasia in multiple fundic gland polyps can lead to the consideration of partial gastrectomy. Duodenal adenomas, particularly ampullary and periampullary ones, more often need therapy. [70] Possible therapies include endoscopic ablation or polypectomy, surgical polypectomy, endoscopic and surgical papillectomy, surgical duodenectomy, and the Whipple procedure. In view of their malignancy potential, removal of duodenal adenomas should be considered if they become large (probably at least 2 cm) or exhibit villous histology or advanced dysplasia. Single, large, pedunculated adenomas can usually be removed endoscopically, whereas larger ones, especially if they are sessile with advanced dysplasia, may require surgical duodenotomy for resection. Severe diffuse duodenal adenomatosis may require duodenectomy or the Whipple procedure, particularly if severe dysplasia is present. Unexpected cancers are often found in the surgical specimen, although radical surgery is still debated for benign lesions in view of possible complications and morbidity. Endoscopic papillectomy is indicated for ampullary adenomas that become symptomatic (including laboratory cholestasis) or develop villous change or advanced dysplasia. [70], [79], [146], [147] and [148] The recurrence rate is high, however, and multiple treatment sessions or surgical therapy may also be needed in selected cases. The Heidelberg Polyposis Register study is particularly demonstrative of the various options. [70] In that registry, endoscopic follow-up of the duodenum of FAP patients was accomplished in 135 patients with an average follow-up of 55.6 months (range, 6 to 172 months). One hundred and thirteen of the patients (83.7%) needed no therapy of the duodenum during the follow-up period, whereas 22 (16.3%) did. Endoscopic polypectomies were accomplished in four cases, whereas surgical duodenotomy was needed because of polyp size (large and sessile) in five. Endoscopic papillectomy was done in six cases because of symptomatic adenomas or severe dysplasia. Whipple procedures were needed in seven patients for severe diffuse duodenal adenomatosis, with cancer being unexpectedly found in four of these surgical specimens. Two of these involved the papilla, whereas two were in the distal duodenum. Other therapies are under investigation for duodenal adenomas, including photodynamic therapy and various NSAIDs. Trials of both have been inconclusive thus far. Studies with higher doses and longer duration NSAID therapy are now underway, however, planned because of the known affect of these agents on colonic adenomas and the need for effective chemoprevention in the duodenum.

Desmoid tumors. CT and magnetic resonance imaging are equally effective in following desmoid tumors. [149] Treatment is undertaken for symptoms, cosmetic issues, or functional involvement of or imminent risk to adjacent structures. Extra-abdominal and abdominal wall desmoids are best treated by surgery with adequate resection of margins. [98], [99] and [100] Less than half of tumors so treated recur, and further therapy with radiation and repeat excision is usually successful. Surgery is difficult, and sometimes impossible, in cases of intra-abdominal desmoids but remains as an important option in selected cases. [98], [150], [151] Because intra-abdominal desmoids often involve the mesentery or encase vessels or organs, medical therapies are often first attempted. The most consistent initial therapies for abdominal desmoids appear to be the NSAID sulindac and the antiestrogen tamoxifen. Both give a better than 50% response after 3 to 6 months of therapy for FAP. [152], [153] The response may be even better with a combination of the two. Chemotherapy can be offered for unresectable cases that fail to respond to this therapy. The protocol of doxorubicin and dacarbazine, followed by carboplatin and doxorubicin, has resulted in either complete remission or partial remission with stabilization of the tumor in

a high percentage of cases. [154] Other therapies have also been used with desmoids with variable success. A combination of low-dose methotrexate and vinblastine demonstrated stabilization of tumor growth or shrinkage in 60% of patients with sporadic desmoids at various stages. [97] This therapy has not yet been reported in FAP desmoid cases. Prednisolone was successful in one case [155] and hyperthermia in another. [156] Intestinal transplantation has now been used successfully in a number of cases of unresectable intra-abdominal desmoids. [157]

SYNDROMES WITH HAMARTOMATOUS POLYPS

Peutz-Jeghers Syndrome

Epidemiology and Clinical Characteristics Peutz-Jeghers syndrome (PJS) is an autosomal-dominant inherited syndrome of histologically distinctive hamartomatous polyps of the GI tract and characteristic mucocutaneous pigmentation. [158] Its incidence is estimated at 1 in 120,000 births. A high risk for both GI and non-GI cancers is now known to be part of this condition. [159] The mucocutaneous melanin pigment spots are seen in more than 95% of cases (Fig. 90-5). They are 1 to 5 mm in diameter and most commonly occur in the perioral and buccal areas (94%). Pigment spots on the lips are distinctive in that they cross the vermilion border and are often much darker and more densely clustered than common freckles. These spots also occur on the face, forearms, digits, palms, soles, perianal area, and rarely the intestinal mucosa. The pigment appears in infancy and may fade with age, but less so on the buccal mucosa.

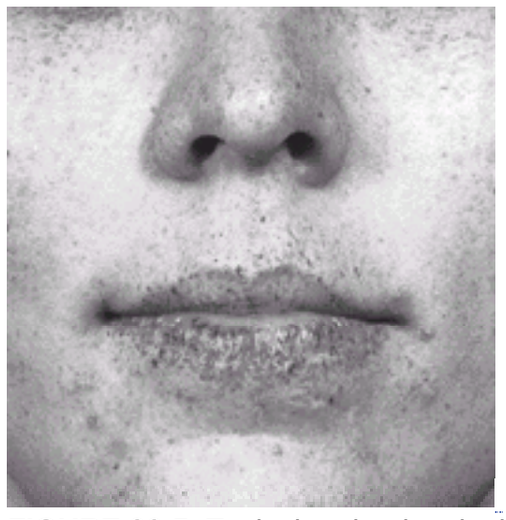


FIGURE 90-5. Typical perioral melanin pigmentation in Peutz-Jeghers syndrome.

The GI polyps occur in 88% to 100% of patients. Their frequency by segment is as follows: stomach, 24%; small bowel, 96%; colon, 27%; and rectum, 24%. [160] Between 1 and 20 polyps are usually found per GI segment. Polyps range from 0.1 to 3 cm in diameter and have a coarsely lobulated surface. Larger polyps are pedunculated, but smaller polyps are sessile, except in the stomach, where they are most often sessile regardless of size. Polyp growth begins in the first decade of life, but patients typically do not develop symptoms until the second or third decade. Symptoms arise from larger polyps, which may infarct, ulcerate, bleed, and cause intestinal obstruction and intussusception. Peutz-Jeghers polyps are histologically distinct. They are nondysplastic, have normal overlying epithelium specific to the GI site in which they are found, and exhibit an arborizing pattern of growth with muscularis mucosae extending into branching fronds of the polyp. Epithelial infolding may result in what is termed *pseudoinvasion* which can lead to an incorrect diagnosis of cancer. Adenoma and cancer may occur in Peutz- Jeghers polyps, exhibit monoclonality, and are thought to be the cause of intestinal cancer in this syndrome. [160], [161], [162] and [163] This is sometimes referred to as the hamartoma-adenoma-carcinoma sequence. Benign complications from the polyps predominate in the first three decades of life. The average age at diagnosis is 22 years in men and 26 years in women. [160] One third of patients experience symptoms in the first decade of life, and 50% to 60% occur before the age of 20 years. The clinical course of PJS is characterized in many patients by repeated surgeries for complications of small bowel polyps. This, together with related malignancies, leads to substantial morbidity and a decreased overall survival rate. [164] Malignant complications become the dominant cause of clinical problems after 30 years of age, [160] with an average age of 43 years for cancer diagnosis of any type. Both GI and non-GI cancers are common in PJS disease, with a combined frequency of all cancers being 93% by age 65 years. [159], [160], [162], [165] Cancer risks and average age of cancer diagnoses are shown in Table 90-6 and Table 90-7.

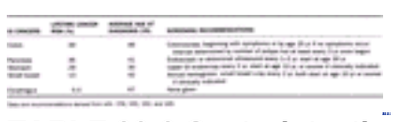


TABLE 90-6 Gastrointestinal Cancer Risks and Screening Recommendations in Peutz-Jeghers Syndrome

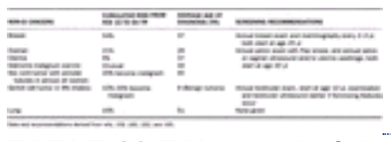


TABLE 90-7 Nongastrointestinal Cancer Risks and Screening Recommendations in Peutz-Jeghers Syndrome

Genetics and Pathophysiology PJS arises from mutations of the *STK11* gene (also called *LKB1*) on chromosome 19p. [166], [167], [168], [169] and [170] About 50% of patients with PJS belong to affected families, whereas 50% are isolated cases and may represent new mutations. [171] Germ-line, disease-causing mutations of the *STK11* gene are found in 60% of those with inherited and 50% of those with isolated PJS, [160] suggesting the possibility of other pathogenic PJS genes. Families with no *STK11* gene mutations exhibit a high incidence of biliary adenocarcinoma compared with families with mutations of this gene. [172] Loss of the wild-type allele of *STK11* by mutation, loss of heterozygosity, or promoter hypermethylation has been found in PJS polyps and cancers, verifying the central role of *STK11* as the disease-causing gene of PJS. [173], [174] Furthermore, the gene codes for a serine-threonine kinase that associates with the *p53* gene and regulates specific *p53*-dependent apoptosis pathways. [175] PJS polyps are devoid of STK11 protein staining and exhibit reduced apoptosis. Several types of germ-line *STK11* mutations have been found to be disease causing. [176] They include frameshift mutations, a 4–amino acid deletion, amino acid substitutions, and splicing errors but have in common the loss of kinase activity. Mutations of ß-catenin and *p53* but not *APC* appear to play a role in neoplasm development after STK11 inactivation. Genetic testing is now commercially available and is performed similar to testing in FAP. A person known to have the clinical syndrome is first tested. If a relevant mutation can be found in the *STK11* gene, other family members can be tested with near 100% accuracy.

Diagnosis, Screening, and Treatment Diagnosis is based on the finding of typical melanin pigment spots and characteristic GI polyps. Management involves screening as outlined in Table 90-6 and Table 90-7 and removal of all polyps larger than 0.5 or 1 cm in diameter to prevent both benign and malignant complications. Screening guidelines are empiric and based on the risk for GI complications and cancer. Colectomy is sometimes necessary to control colonic polyps and should be considered, especially if neoplastic change is found in the colon. When small bowel polyps become large or symptomatic, surgery is often necessary and includes careful examination of the entire small bowel to eliminate all significant polyps. Intraoperative endoscopy may be a helpful adjunct in this regard, and this is also an appropriate time to examine and remove gastric and duodenal polyps of significant size. [177]

Juvenile Polyposis

Epidemiology and Clinical Characteristics Juvenile polyposis (JP) is an autosomal-dominant inherited condition, with 20% to 50% of cases having a family history of the disease. [178] It is characterized by multiple juvenile polyps of the colon, although polyps may occur throughout the GI tract. [171], [178], [179], [180] and [181] There is also a very high risk for colon cancer; an increased risk for gastric, duodenal, and pancreatic cancers; and the frequent occurrence of certain benign extraintestinal features. The incidence of JP is about 1 in 100,000 individuals. The generally accepted clinical criteria for JP include the following: (1) at least five juvenile polyps in the

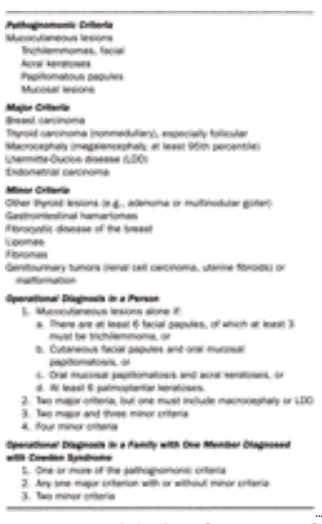
colorectum; (2) juvenile polyps throughout the GI tract; or (3) any number of juvenile polyps in a person with a known family history of juvenile polyps. [182] Inherited syndromes that exhibit the GI phenotype of JP must also be ruled out, including Cowden syndrome (CS), Bannayan-Riley-Ruvalcaba (BRR) syndrome, and Gorlin syndrome (see later). The polyps in JP vary in size from small sessile nodules to pedunculated lesions that are 3 cm or larger in diameter. Most large polyps are pedunculated, but small polyps, especially those in the stomach, are sessile. Grossly, most polyps exhibit a surface that is smooth, rounded, reddish-colored, and without fissures or lobulations, except for large polyps, which may appear to be multilobulated. A white exudate is often seen on the polyp surface. On cut section, there are cystic spaces filled with mucin. Microscopically, there is abundant lamina propria with benign but often elongated and cystically dilated glands and lack of a smooth muscle core. Excess chronic inflammatory cells are sometimes present. The epithelial lining of the surface and cysts is nondysplastic and reflects the area of the GI tract where the polyp is located. [183] The polyps of JP are most commonly found in the colon but may occur throughout the GI tract. [178] Polyps begin to appear in the first decade of life, and dozens to many hundreds of polyps are present in the fully developed syndrome. Most patients develop symptoms in the first two decades of life. The average age at diagnosis is 18.5 years. [184] Rectal bleeding with anemia is the most common presenting symptom, followed by abdominal pain, passage of tissue per rectum, and intussusception. A rare and often lethal form of the disease can be observed in infancy. It includes large numbers of juvenile polyps, GI bleeding, protein-losing enteropathy, and significant morbidity and even mortality. [185] Estimates of the risk for colon cancer vary from 9% to 68%. [171], [178], [182], [186], [187] More recent studies indicate the higher figures, and the latter figure is likely the most accurate because it represents the projected incidence of colorectal cancer by 60 years of age considering all JP cases in the St. Mark's Registry in London. The mean age of colon cancer is 34 years, with a range of 15 to 68 years. [181], [184], [187], [188] Although occasional isolated adenomas are observed in JP, the cancer risk is believed to arise from the occurrence of adenomatous tissue within the juvenile polyp. Up to 50% of juvenile polyps in JP contain areas of adenomatous change. Villous and malignant foci may be seen in larger polyps. [182], [186] Cancers of the stomach, duodenum, pancreas, and biliary tree have been reported in JP. [189] One study found that 6 (21%) of 29 JP patients from one large kindred had developed cancer of the stomach or duodenum. [187] The average age of upper GI carcinoma is 58 years, with a range of 21 to 73 years. [189] The precise phenotype of JP is somewhat in question because it was previously difficult to distinguish CS and BRR syndrome from JP, each exhibiting multiple intestinal juvenile polyps. Now that these syndromes can be defined genetically, a much more accurate clinical picture of each is emerging. Past reports, for example, found that congenital anomalies were present in 11% to 15% of JP cases. [189] Abnormalities included congenital deformities of the heart, CNS, soft tissues, GI tract, and genitourinary system and were primarily found in sporadic JP cases. A more recent evaluation found a higher incidence of congenital anomalies in JP, but mostly macrocephaly and hypertelorism once the other syndromes were separated. [190] Similarly, there has been an effort in the past to separate cases of JP involving just the colon from those involving the entire GI tract. A reexamination of each of these issues is clearly needed in the context of genetic diagnosis.

Genetics and Pathophysiology At least half of the families clinically affected with JP are found to have a disease-causing mutation of either the *SMAD4* gene (also called the *DPC4* gene) on chromosome 18, or the bone morphogenetic protein receptor 1A (*BMPR1A*) gene on chromosome 10, both tumor suppressor genes. [191], [192], [193] and [194] It was first estimated that *SMAD4* mutations would account for 35% to 60% of JP cases [192]; however, subsequent studies have suggested a smaller fraction, [195], [196] and a recent survey from Europe indicated that only 15% of JP families arise from germ-line mutations of the *SMAD4* gene, whereas 38% are from germ-line mutations of the *BMPR1A* gene. [194] Only a few families with JP appear to arise from mutations of the *PTEN* gene, [197], [198] although some maintain that these families are actually CS families. The *SMAD4* gene encodes a cytoplasmic mediator involved in the transforming growth factor-ß (TGF-ß) signal transduction pathway. Activation of the pathway leads to cell growth inhibition by way of regulating the transcription of genes involved in cell cycle and transcriptional regulation. [199] There are a number of additional *SMAD* genes in the pathway, but JP-associated mutations have not been found in them. [199], [200] and [201] The *BMPR1A* is a serine-threonine kinase type 1 receptor that also belongs to the superfamily of TGF-ß. It activates *SMAD4* independently of TGF-ß signaling, but the effector pathway, nuclear control, and transcriptional regulation are similar. The genetic etiology of JP in up to half of cases remains elusive, however, indicating that other genes may be involved in the etiology of the syndrome. It has been suggested that genetic events that give rise to JP affect stromal cells primarily and that epithelial cells acquire dysplasia only as a secondary event. [193] A recent investigation, however, demonstrated biallelic inactivation of the *SMAD4* gene in both the epithelium and stromal cells of JP polyps. [202] This observation would indicate that inactivation of *SMAD4* is a primary event in dysplastic transformation of epithelial cells in the polyps of JP.

Diagnosis, Screening, and Treatment Diagnosis is based on recognition of the disease characteristics and on genetic testing, especially in families. Commercial genetic testing has recently become available for JP, and clinical testing is approached as it is in FAP and PJS. Genetic testing is particularly important in JP, both to confirm the diagnosis in a proband and to test relatives. Testing is also important to separate JP from CS in view of the high risk for colon cancer in JP but not CS. Genetic testing has also been shown to increase markedly the screening compliance in relatives of those with JP. [203] It has been suggested that upper endoscopy and colonoscopy examinations begin in the late teens, or earlier if symptoms occur in those with a clinical or genetic diagnosis, or in those at risk if a genetic diagnosis is not possible. [181] Studies should be repeated every 3 years if no polyps are present but annually for endoscopic therapy if polyps are found. Colectomy is indicated if polyps are difficult to control endoscopically or if advanced neoplastic change or cancer is observed in polyps. Complete or partial gastrectomy may be necessary for cancer or advanced lesions that cannot be effectively controlled endoscopically. Screening recommendations have not been suggested for pancreatic cancer.

Cowden Syndrome

Epidemiology and Clinical Characteristics CS, also called *multiple hamartoma syndrome* is an autosomal-dominant inherited condition characterized by multiple hamartomas of the skin, mucous membranes, GI tract, and other organs and a high risk for cancer of the breast, thyroid, and perhaps other sites. [181], [204], [205], [206], [207] and [208] The disease occurs in about 1 in 200,000 individuals but is believed to be underdiagnosed. An international consortium has developed diagnostic criteria for CS [207] (Table 90-8). The hallmark of the syndrome is the presence of multiple facial trichilemmomas. Verrucous skin lesions of the face and limbs and cobblestone-like hyperkeratotic papules of the gingiva and buccal mucosa are common. [206] Biopsy examination has demonstrated facial lesions to most often be trichilemmomas, oral mucosal lesions to be fibromas, and all hand and foot lesions to be hyperkeratoses. [209] By the third decade of life, 99% of affected patients have developed mucocutaneous stigmata of the disease. [205]



Pathognomonic Criteria
Mucocutaneous lesions
- Trichilemmomas, facial
- Acral keratoses
- Papillomatous papules
- Mucosal lesions

Major Criteria
Breast carcinoma
Thyroid carcinoma (nonmedullary), especially follicular
Macrocephaly (megalencephaly, at least 95th percentile)
Lhermitte-Duclos disease (LDD)
Endometrial carcinoma

Minor Criteria
Other thyroid lesions (e.g., adenoma or multinodular goiter)
Gastrointestinal hamartomas
Fibrocystic disease of the breast
Lipomas
Fibromas
Genitourinary tumors (renal cell carcinoma, uterine fibroids) or malformation

Operational Diagnosis in a Person
1. Mucocutaneous lesions alone if:
   a. There are at least 6 facial papules, of which at least 3 must be trichilemmoma, or
   b. Cutaneous facial papules and oral mucosal papillomatosis, or
   c. Oral mucosal papillomatosis and acral keratoses, or
   d. At least 6 palmoplantar keratoses
2. Two major criteria, but one must include macrocephaly or LDD
3. Two major and three minor criteria
4. Four minor criteria

Operational Diagnosis in a Family with One Member Diagnosed with Cowden Syndrome
1. One or more of the pathognomonic criteria
2. Any one major criterion with or without minor criteria
3. Two minor criteria

TABLE 90-8 Diagnostic Criteria for Cowden Syndrome

Other common manifestations of the disease include multinodular goiter (50%), follicular (sometimes papillary) thyroid carcinoma (10%), [205] thyroid adenoma, fibrocystic breast disease and breast fibroadenomas (up to 75%), breast cancer (25% to 50%; median age at diagnosis, 41 years; range, 14 to 65 years), [209], [210] GI hamartomas (about 60%, although systematic study has not been undertaken), [206] multiple, early-onset uterine leiomyomas, macrocephaly (specifically, megencephaly), and mental retardation. Possibly related cancers include those of the endometrium (lifetime risk, 2% to 5%), kidney, colon, ovary, melanoma, lung, and retinal glioma. [206], [207] and [208] Male breast cancer has been reported in patients with CS. [208], [211] Additional benign soft tissue and visceral tumors that have been reported include hemangiomas, lipomas, lymphangiomas, neurofibromas, and meningiomas. Developmental or congenital abnormalities also occur and include hypoplastic mandible, a prominent forehead, and a high-arched palate. The GI hamartomas occur throughout the GI tract. A review of the reported cases in Japan found polyp distribution among 93 cases to be as follows: esophagus, 66%; stomach, 75%; duodenum, 37%; and colon, 66%. [212] Polyps of the esophagus are actually glycogenic acanthosis, which appear as whitish, flat elevations in the esophagus. [213] In the stomach, small bowel, and colon, a number of different types of hamartomas are observed, including juvenile polyps (by far the most common), lipomas, inflammatory polyps, ganglioneuromas, and lymphoid hyperplasia. Juvenile-like polyps that contain some neural elements are considered characteristic of this disease. GI cancer risk has generally been considered not elevated, although the study from Japan quoted previously found 9.6% of 93 patients to have colon cancer.

Related Syndromes

Lhermitte-Duclos disease. The rare occurrence of a benign hamartomatous overgrowth of ganglion cells in the cerebellar cortex (called *dysplastic gangliocytoma of*

the cerebellum) is called Lhermitte-Duclos disease. This same tumor is a known manifestation of CS. Recently, it has been found that virtually all cases of Lhermitte-Duclos occur as a part of CS. [214], [215] and [216]

Bannayan-Riley-Ruvalcaba syndrome. BRR syndrome, also previously called *Ruvalcaba-Myre-Smith syndrome* and *Bannayan-Zonanna syndrome* is characterized by the features of CS together with macrocephaly, delayed psychomotor development, lipomatosis, hemangiomatosis, and pigmented macules of the glans penis. [217], [218] Similar to CS, intestinal polyps are primarily juvenile polyps, and some contain ectopic ganglion cells. [219] It is now known that BRR syndrome and CS arise from mutations of the *PTEN* gene [220], [221] and that the identical mutation can lead to either phenotype. [221], [222] and [223] It has been suggested that CS and BRR syndrome be referred together as the *PTEN* hamartoma tumor syndromes.

Genetics and Management CS was recently found to arise from mutations of the *PTEN* gene. [224], [225], [226] and [227] *PTEN* is a ubiquitously expressed dual-specificity phosphatase, which acts as a tumor suppressor gene. [228] It regulates cellular processes, such as cell cycling, translation, and apoptosis by blocking activation of Akt, a serine-threonine kinase involved in cell growth and survival. [229], [230], [231] and [232] Up to 80% of patients who meet the diagnosis criteria of CS are found to have a *PTEN* mutation, [226], [233] whereas only 10% to 50% of people will have a *PTEN* mutation when the criteria are not fulfilled. [233], [234] and [235] Fifty percent to 60% of BRR syndrome patients exhibit a PTEN mutation. [221] Mutations are found in more than 90% of families that exhibit features of both CS and BRR. [207] One family with a CS-like phenotype was found to have a disease causing germ-line mutation of the *BMPR1A* gene. [194] Commercial genetic testing is now available for CS and BRR.

Surveillance Management of CS involves prevention of the associated cancers. [207] The specific risks and screening recommendations are given in Table 90-9. No recommendations have been given for the surveillance of GI areas, mainly because GI cancer risk is not increased in CS. Intervention is obviously needed whenever GI symptoms occur, especially rectal bleeding. It is now believed that most cases of CS are diagnosed because of either skin or GI findings. [206] Surveillance of the GI tract should probably be synthesized on the basis of polyp location, number, size and histology, once they are delineated. Finally, the finding of an increased risk for colon cancer in CS patients in Japan [212] indicates that better definition of the GI phenotype is needed in other populations.

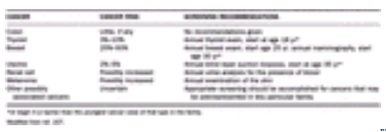


TABLE 90-9 Cancer Risks and Screening in Cowden Syndrome

Hereditary Mixed Polyposis Syndrome

Three families were described in which the polyposis phenotype links to a locus on chromosome 6. [236], [237] and [238] Affected patients exhibited primarily juvenile polyps but also adenomatous polyps and hyperplastic polyps and sometimes polyps of these histologies mixed.

Gorlin Syndrome

Gorlin syndrome, also called *nevoid basal cell carcinoma syndrome* is an autosomal dominant disorder with an occurrence of 1:55,600. [206] It accounts for about 0.5% of patients with basal cell cancer and occurs secondary to mutations of the *PTCH* gene on chromosome 9q. The primary clinical findings include multiple basal cell carcinomas, mandibular bone cysts, characteristic pits in the skin of the palms and soles, intracranial calcification, large head circumference, and congenital skeletal anomalies. Gastric hamartomatous polyps have been reported as a part of the disease.

POLYPOSIS SYNDROMES WITH NEURAL POLYP HISTOLOGY

Neural elements can be observed in the juvenile polyps of CS and BSS, and when found, are somewhat distinctive for those diagnoses. Multiple GI polyps with histologic neural elements can also be found in neurofibromatosis type I, in multiple endocrine neoplasia type IIB (MEN IIB) and independently in families without any known syndromes.

Neurofibromatosis Type I

Neurofibromatosis type I, also called *von Recklinghausen disease* is defined by the presence of more than five cutaneous café-au-lait spots together with frequent neurofibromas of the skin. [206] The incidence is 1 in 3000, and one third to one half of cases are new mutations. The condition is autosomal dominantly inherited and arises from mutations of the *NF1* gene on chromosome 17q. About 25% of affected patients exhibit multiple intestinal polypoid neurofibromas and less commonly ganglioneuromas. [239], [240] The small bowel is affected most commonly, followed by the stomach and then the colon. GI symptoms are unusual but may include GI bleeding, intussusception, or obstruction. [241], [242] Neurofibrosarcoma has only rarely been reported.

Multiple Endocrine Neoplasia Type IIB

Also called *multiple neuroma syndrome* MEN IIB is characterized by the presence of multiple tumors, including medullary thyroid carcinoma, pheochromocytoma, parathyroid adenomas (less common than in MEN IIA), enlarged and nodular lips (from ganglioneuromas), a marfanoid habitus, and ganglioneuromatosis of the GI tract. GI involvement in MEN II is common, with ganglioneuromatous polyps occurring throughout the GI tract, from lip to anus, but most commonly in the colon and rectum. [240], [243], [244] and [245] GI symptoms, including diarrhea and constipation, are common (more than 90% of cases) and arise from dysmotility of the GI tract secondary to the underlying ganglioneuromatoses because the lesions are often transmural. [240], [246], [247] Symptoms often begin in the first months of life and may present as megacolon. A single mutation at codon 918 in the tyrosine kinase domain of the RET receptor (of the RET protooncogene on chromosome 10q) has been associated with the MEN IIB phenotype. [244] About half of cases discovered are considered to be new mutations. Genetic testing is now available for this condition. [248]

Isolated Intestinal Neurofibromatosis of the Colon

Multiple colonic ganglioneuromas have been observed in families and isolated cases unrelated to von Recklinghausen disease or to MEN IIB. [243], [244], [249], [250] and [251] The observed ganglioneuromas are both microscopic and polypoid. There may be dozens of visible colonic polyps ranging up to 0.5 cm in diameter. The distribution of affected individuals in the one family described was typical of autosomal-dominant inheritance.

POLYPOSIS SYNDROMES OF UNCERTAIN ETIOLOGY

Cronkhite-Canada Syndrome

Cronkhite-Canada syndrome (CCS) is a noninherited condition characterized by generalized GI polyposis, cutaneous hyperpigmentation, hair loss, and nail atrophy. [252], [253] and [254] The syndrome has a worldwide distribution. [255], [256] Sixty percent of reported cases occur in men, and the average age at symptom onset is 59 years, with a range of 31 to 86 years. No familial occurrences have been observed. Nutritional, infectious, and immunologic associations have been discussed, but the cause of CCS remains elusive [257], [258]

The polyposis of CCS is present throughout the GI tract except for the esophagus. [253] Hundreds of polyps are observed, with polyp size ranging from a few millimeters to 3 cm in diameter. Colonic polyps are almost always sessile, even when large, which can often differentiate them from juvenile polyps. This is not, however, a reliable feature of the gastric polyps, which often become pedunculated. [257] Ulceration and friability are surface features of some of the polyps. Microscopically, the polyps are almost identical to juvenile polyps. In contrast to JP, however, the mucosa between polyps is also histologically abnormal, with edema, congestion, and inflammation of the lamina propria and focal glandular ectasia. Polyps may appear over several years or several months. They usually resolve after remission of symptoms, but they may persist for years.

Adenomatous change and colon cancer have been reported in the hamartomatous polyps of this syndrome. [255], [256], [259], [260] The overall incidence of colon cancer

appears to be about 12% to 15%. [253], [256], [260] Carcinoma of the stomach is also reported as a consequence of CCS. [261] It remains uncertain whether cancers of the GI tract arise from adenomatous change in the hamartomatous polyps, arise directly from the hamartomatous polyps, or both. [261]

A number of extraintestinal, ectodermal manifestations are observed in almost all patients with CCS. [253] Nails of the fingers and toes exhibit various degrees of dystrophy, described as thinning, splitting, and partial separation from the nail bed (i.e., onycholysis). Hair loss occurs over a few weeks on the scalp, eyebrows, face, axillae, pubic area, and extremities. Hyperpigmentation is described as dark, brownish macules, ranging from a few millimeters to 10 cm in diameter, and it occurs over the upper extremities, followed by the lower extremities, face, palms, soles, neck, back, chest, and scalp, in that order. The nail, hair, and pigmentation abnormalities are all reversible on remission of the disease.

Hypogeusia is the dominant initial symptom in most patients, which is soon followed by diarrhea, abdominal discomfort, anorexia, and weight loss. [253], [254] The diarrhea results from protein loss caused by excess mucous secretion by crypt cells. [257] Variable degrees of fat and disaccharide malabsorption also occur because of a somewhat decreased absorptive surface, but not from damage to the absorptive epithelium. Other common symptoms include fatigue, weakness, edema, nausea, and vomiting.

The disease often exhibits a fairly acute onset and a rapidly progressive course. [253] Weight loss and edema ensue, and ectodermal changes occur in a few weeks to a few months. GI bleeding also may occur and may be severe in some cases. Intussusception from small bowel polyps has been reported but is unusual. The diarrhea and protein-losing enteropathy may be extremely severe, resulting in profound malnutrition. Complications resulting from malnutrition are a major cause of morbidity and mortality in this syndrome. They include severe cachexia, anemia, congestive heart failure, and impaired immunity, resulting variously in pneumonia, sepsis, and septic shock. The disease may be fatal within a few months, although a more protracted course is also possible, especially if the patient responds to therapy or remits spontaneously.

A number of spontaneous remissions have been reported in CCS, and partial or complete remissions have resulted from several different interventions. [253], [256], [262], [263], [264], [265], [266] and [267] Therapies have included corticosteroids, antibiotics, colectomy, parenteral nutrition, and combinations of these. Each therapy has been successful in some cases but unsuccessful in others. Present recommendations are for aggressive supportive therapy, including enteral or intravenous alimentation. Corticosteroid therapy is undertaken if deterioration continues, and antibiotic therapy may be attempted, although its usefulness is questionable. Surgery is most often used to treat complications, including bleeding, malignancy, intussusception, and sometimes protein-losing enteropathy. Colonoscopy should be performed, if feasible, to consider the possibility of malignancy. Periodic examination of the colon and stomach seems to be indicated in long-term survivors with persistent polyps to screen for adenomatous change and colon cancer.

Hyperplastic Polyposis Syndrome

Rare patients are described with large numbers of hyperplastic polyps distributed throughout the colon. [268], [269] and [270] In such patients, polyps are most often sessile, 1 to 7 mm in diameter, but larger, sessile, or pedunculated hyperplastic polyps may be observed. [271], [272] Smaller polyps are distributed equally throughout the colon, whereas larger lesions often have a proximal colonic distribution. [269], [270] Colon cancer has often been reported with HP, but most of the cases were ascertained because of the cancer, so that the actual malignant risk remains unknown. In favor of a cancer association is the frequent finding of adenomatous polyps in patients with HP and even a description of serrated adenomatous polyposis. [273] Familial cases have been reported, but inheritance remains poorly defined. [270], [271] and [272], [274]

The most difficult issues with HP are to distinguish it from cases with multiple sporadic hyperplastic polyps and to determine appropriate management in view of the probable cancer risk, the frequent accompanying adenomas, and the large numbers of polyps with which the endoscopist must deal. Working criteria have been suggested to assist with diagnosis: (1) at least five hyperplastic polyps proximal to the sigmoid colon with two or more of them larger than 10 mm in diameter, (2) any number of hyperplastic polyps proximal to the sigmoid colon in an individual who has a first-degree relative with HP, or (3) more than 30 hyperplastic polyps of any size, but distributed throughout the colon. [269] A definition of more than 20 hyperplastic polyps has also been used. [270] Germ-line mutations for HP have not been identified, but chromosome 1p allelic loss has been observed in large polyps of HP patients. [270]

Management includes interval colonoscopic polyp removal, ablation, or both. I perform colonoscopy every 1 to 3 years, depending on the number of both hyperplastic and adenomatous polyps at each examination. Subtotal colectomy should probably be considered if HP with large numbers of polyps is found simultaneous to a colon cancer diagnosis.

POLYPOSIS SYNDROMES WITH INFLAMMATORY POLYPS

Inflammatory Bowel Disease

Multiple inflammatory polyps are frequently found in both ulcerative colitis and Crohn's disease. The polyps represent remaining normal tissue, with inflammatory elements that persist during the healing phases of the diseases. The polyps themselves have no malignant potential, although both ulcerative colitis and Crohn's disease have a colon malignancy risk that parallels the extent of colonic involvement and the duration of the disease.

Devon Polyposis

Devon polyposis, also known as *Devon family syndrome* was described in a family in which three generations of females were found to have multiple inflammatory fibroid polyps of the ileum. [275] The polyps varied in size from 0.5 to 8 cm, and each affected person experienced intussusception or small bowel obstruction. Similar polyps were found in the gastric antrum of one patient. Histologically, the polyps were benign proliferations of histiocytes.

Cap Polyposis

Cap polyposis is a rare syndrome of multiple inflammatory polyps in the rectosigmoid. Histologically, the polyps are inflammatory, with elongated tortuous crypts covered by a "cap" of granulation tissue. Clinically, patients may have diarrhea, rectal bleeding, and protein-losing enteropathy. [276] The authors of one case hypothesized mucosal prolapse as an etiology because they observed histological similarities to solitary rectal ulcer syndrome, in which multiple 1- to 3-mm–sized polypoid lesions in the distal rectum can also be observed. [276]

POLYPOSIS CONDITIONS ARISING FROM LYMPHOID TISSUE

Nodular Lymphoid Hyperplasia

Nodular lymphoid hyperplasia (NLH) is a rare polyclonal lymphoproliferative disorder of unknown cause that, in most cases, is not related to a distinct disease. [277] It is found in the terminal ileum of some patients with Gardner syndrome, in about 20% of patients with common variable immunodeficiency syndrome, [277], [278], [279], [280] and [281] as a rare association with intestinal lymphoma, [282], [283], [284] and [285] and in some otherwise healthy children. [286], [287] NLH also has been described in the small bowel of adults without immunodeficiency. [288], [289] It has been suggested that hyperplasia of the lymphoid nodules occurs as a local immune response to antigens in the gut lumen. It has been suggested that cellular immune dysfunction, as well as immunoglobulin deficiency, must be present for NLH to be present. [290]

The hyperplastic lymphoid nodules of NLH are most often found in the small bowel but also may occur in the stomach and colon. They are described as numerous, 3 to 6 mm in diameter, and exhibiting the same color as surrounding mucosa. Occasionally, nodules may reach 10 mm in diameter and even larger. The enlarged lymphoid follicles are morphologically indistinguishable from lymphoid follicles that occur normally in the GI tract. The nodules themselves do not usually cause symptoms, which are more likely related to the underlying conditions.

The primary clinical issue is to distinguish NLH from normal lymphoid tissue, where follicles range in size from 0.6 to 3 mm in diameter throughout the gut. Barium enema may reveal lymphoid follicles without evidence of disease in up to 50% of patients younger than 30 years of age and in up to 17% of those older. [291] Normal lymphoid nodules are less frequently detected by colonoscopy but become very apparent in melanosis coli. More recent endoscopic characterization of lymphoid

follicles, however, may show these normal structures to be more common. [292] Lymphoid hyperplasia of the terminal ileum is already found in almost half of patients examined by colonoscopy. [293] Follicles range in size from 1 to 10 mm or more and are larger, more numerous, and more frequent in younger individuals. Immunohistochemistry staining and tissue genotyping can now be used to distinguish malignant changes of lymphoid tissue and NLH. [294]

NLH as an entity does not require therapy.

Multiple Lymphomatous Polyposis

About 30% of primary extranodal lymphomas occur in the GI tract. Most of these present as a single lesion, although about 10% exhibit GI polyposis, including multiple lymphomatous polyposis (MLP) and immunoproliferative small intestinal disease (see later). [295], [296]

MLP is a rare intestinal malignancy characterized by the presence of numerous GI polypoid lesions of malignant lymphoma. It is a non-Hodgkin's B-cell lymphoma that is the GI counterpart of mantle cell lymphoma. [297], [298] and [299] MLP arises from a malignant transformation of lymphocytes that exhibit homing receptors for lymphoid tissue of the GI tract, thus giving rise to a diffuse polyposis presentation. [300] The polyps range from a few millimeters to several centimeters in size, involve the small and large bowel in 80% to 90% of cases and the stomach or duodenum in 50% of cases. [301] This condition occurs predominantly in men (88%), with reported cases ranging in age from 43 to 82 years at the time of diagnosis. [302] Symptoms are nonspecific, including weight loss, fatigue, diarrhea, abdominal pain, anemia, and occult GI bleeding. The condition often exhibits extra-abdominal dissemination, especially to peripheral lymph nodes. Chemotherapy is the treatment of choice, although the overall prognosis is poor. [299]

Mucosa-associated lymphoid tissue lymphomas, follicular lymphomas, and primary T-cell lymphomas have now also been described that are morphologically similar to MLP in that they exhibit a multiple polypoid appearance in the GI tract and are thus considered types of MLP. [303], [304], [305], [306], [307] and [308]

Immunoproliferative Small Intestinal Disease

Immunoproliferative small intestinal disease (IPSID), previously called both *Mediterranean-type lymphoma* and *a-heavy chain disease* can also exhibit multiple nodular lesions of the small bowel. [309], [310] IPSID most commonly occurs in people living in the Mediterranean region in the second or third decade of life. Patients present with a malabsorption syndrome lasting months to years that includes chronic diarrhea, weight loss, and abdominal pain. Small intestinal endoscopy may variously show thickened mucosal folds, diffuse nodularity, ulcers, a mosaic pattern, and nondistensibility from infiltration.

Histologically, IPSID begins as an intense proliferation of plasma cells in the lamina propria. It eventually proceeds to an overt malignant lymphoma with extension beyond the lamina propria and sometimes the occurrence of multiple nodular lesions. The process may remain in deeper intestinal layers, however, requiring full-thickness biopsy for diagnosis. The lymphoma is a plasma cell tumor or an immunoblastic sarcoma. An abnormal paraprotein is frequently resent, which represents the a-chain fragment of immunoglobulin A. IPSID is now thought in most, if not all, cases to be the late stages of a-heavy chain disease. [309] Unlike MLP, it is confined to the gut and is most often present in the small bowel.

MISCELLANEOUS NONINHERITED POLYPOSIS SYNDROMES

A number of reports have been made of leiomyomatosis of the colon and other segments of the GI tract. [311], [312] and [313] A multinodular submucosal tumor appearance is observed lumenally, which reflects single or multiple benign growths of smooth muscle of the bowel wall.

Lipomatous polyposis is also rarely observed. [314], [315] In one case, between 700 and 1000 lipomatous polyps were observed, but about 60 adenomatous polyps were also present. Polyps were 2 to 50 mm in diameter, were found throughout the colon, but were more dense in the left colon. [315] In another case, dense colonic lipomatous polyposis was observed together with lipomas of the peritoneum. [316] A third patient was found to have multiple lipomas throughout both the small and large intestine and a presentation of intussusception. [317]

Lymphangiomas are unusual solitary lesions in the colon, representing dilation or overgrowth of lymphatic channels. Clusters of elevated but apparent submucosal colonic polypoid lesions were observed colonoscopically in a recent case in which the lesions were found to be multiple lymphangiomas. [318]

Finally, pneumatosis cystoides intestinalis is characterized by multiple air-filled cysts of the wall of the GI tract that can have the appearance of polyposis by barium enema and colonoscopy. [319] The diagnosis is made by abdominal radiography or colonoscopic biopsy.

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CHAPTER 91

C. Richard Boland

MALIGNANT TUMORS OF THE COLON

Colorectal cancer has been the focus of clinical and basic research since the early 1980s because of its importance in public health, and it has become the cancer most responsible for the explosive increase in our understanding of tumor biology. We know more about the biology and clinical characteristics of this tumor than any other digestive tract cancer and, possibly, any other tumor. This knowledge provides the gastroenterologist with a powerful array of diagnostic and prognostic tools for our patients and permits us to provide advice for patients who are concerned about their risk for this disease.

It is now clear that the development of colorectal cancer represents an interaction between the genome of the colorectal epithelial cell and its environment; both factors are essential for the development of a tumor. The likelihood of developing tumors is related to diet, which accounts for the striking international epidemiology of this disease. In certain instances, the genetic predisposition in a person is overwhelming, and the patient faces an extremely high risk for cancer. Several of the genes associated with these predispositions have been identified. In most instances, however, the risk factors are more subtle, and it is possible that the risk for cancer can be modified. Primary prevention, that is, dietary modification or the introduction of a chemopreventive drug, would appear to be the most appealing approach to this disease, but insufficient data are available to bring a quantitative estimate of the impact of any changes in diet. No chemopreventive drug is known yet to be sufficiently safe and effective that we can offer it to our patients. Secondary prevention, that is, colonoscopic surveillance and the removal of precursor lesions, is highly effective, and the physician is faced with implementing the most appropriate regimen for each patient. Progress in these areas has created a challenge to keep current with this disease.

Colorectal cancer is one of the most common potentially lethal gastrointestinal diseases encountered in clinical practice and is readily cured with proper management. Most of the neoplastic lesions in this organ are adenomas and adenocarcinomas, and the comments made in this chapter are directed toward these tumors, unless specifically stated otherwise.

Colorectal neoplasia is a multifaceted problem. The basic biology of gastrointestinal neoplasia has been discussed in Chapter 24 and includes discussion of concepts such as tumor genetics, multistep carcinogenesis, oncogene activation, inactivation of tumor suppressor genes, clonal expansion of neoplastic cells, and the generation of cellular diversity in tumor progression as a background for understanding colonic neoplasia. Additional details regarding the performance and interpretation of tests for occult fecal bleeding are found in Chapter 34. Small intestine tumors share some of the characteristics of colonic tumors, but this type of tumor is addressed specifically in Chapter 80. Benign colonic polyps and gastrointestinal polyposis syndromes are discussed here in the context of cancer but are described in more detail in Chapter 89 and Chapter 90, respectively. Management of malignant polyps is discussed in the context of adenomatous polyps in Chapter 149 of this edition. Carcinoma of the anus is not described in this chapter, but it is covered in Chapter 92. The use of colonoscopy in the diagnosis of colonic neoplasia is discussed here, but additional details regarding the mechanics of this procedure may be obtained from Chapter 140 and Chapter 142. Colonic lymphoma and carcinoid tumors of the colon are distinct from adenocarcinomas and are discussed separately at the end of this chapter, together with rarer pathologic forms of colon cancer.

Epidemiology

Cancer is considered an acquired genetic disease produced by exposure to environmental carcinogens; the damage caused by these carcinogens accrues over many years. Exposure to environmental carcinogens is a constant and cumulative process. Because colonic carcinogenesis is a multistep process, considerable time is required for these chance events to accumulate. Gut epithelia are dynamic tissues that constantly undergo proliferation and renewal. No single gene is so critical to the process that a mutation in it will result in cancer. Each mutation or genetic alteration involved in carcinogenesis produces a new cell that has a slight survival advantage over the previous ones. This growth advantage permits clonal expansion of the cells. Additional genetic events occur to cells within the clone, which then give rise to new clones that, in turn, overgrow their progenitors. A critical but currently undefined nuclear event eventually occurs that results in genomic instability, giving rise to the genetic diversity observed in tumor cell populations. After the onset of genomic instability, additional mutational changes continue spontaneously without the necessity for additional exogenous carcinogens. Eventually, clones emerge that are capable of forming metastatic colonies.

The likelihood of producing a neoplastic clone, that is, one that can free itself from homeostatic growth controls, increases with time and total carcinogen exposure (which is constant and perhaps inevitable), and the clinical result is that cancer incidence rises as an exponential function of age. [1] Deaths from colorectal cancers

typically begin to increase slowly in the fifth decade of life, rising steeply with advancing age. [2] In the Western world, incidence of cancers of the colon and rectum increases between 50 and 80 years of age as a function of the sixth power of age. [3] Any consideration of cancer epidemiology must therefore be corrected for differences of age in the study populations.

In the United States, in 2001 there were an estimated 135,400 new cases of colorectal cancer and 56,700 deaths from this disease (42% of the incidence rate). [4] In 2000, there were about 553,400 cancer deaths in the United States, and just more than 10% were caused by colorectal cancer. [4] Several countries around the world have incidence and mortality rates higher than those in the United States. Attack rates for colon cancer are considerably lower in Japan and in most developing nations.

Rectal cancer is a distinct disease from colon cancer in its epidemiology, in its pathogenesis, and in the way we treat it. Interestingly, the incidence of rectal cancer in Japan is similar to that in the United States. Mortality from rectal cancer in the United States appears to be slowly declining, even though incidence is slowly increasing. [5], [6] The divergence between incidence and mortality is associated with a shift to cancers with a more favorable stage, suggesting the impact of early detection of tumors and removal of premalignant lesions from the distal colon and rectum.

GEOGRAPHIC PATTERNS

An essential feature of colorectal cancer is the wide geographic variation in its incidence at the international level. As illustrated in Table 91-1, the age-adjusted incidence of colorectal cancer varies by as much as a factor of 4 between different population groups. [3] Industrialized nations are at greatest risk. These differences appear not to be caused principally by genetic factors because populations migrating from low- to high-incidence regions quickly experience an increase in their rate of colorectal cancer. [7] Also, there is a marked difference in risk between African Americans and black African populations. [8], [9] This is strong evidence for the impact of environmental factors on the incidence of colon cancer and is a source of optimism that the high rates seen in the Western world might be reduced by identifying and removing critical causative factors from the diet or environment. Conversely, changes in dietary practices are thought to be responsible for adverse changes in this disease in Japan. The age-adjusted incidences of cancer of both the colon and rectum have risen more than threefold from 1974 to 1991 in Japan, and these rates now approach those seen in the Western world. [10]

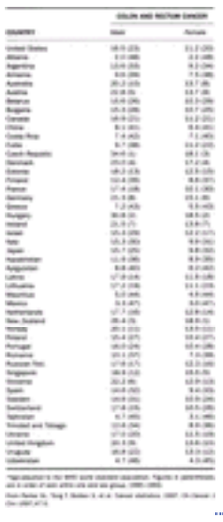


TABLE 91-1 Age-Adjusted International Colorectal Cancer Death Rates Per 10 [5] Population*

The estimated incidence rates of cancer vary widely, depending on the means of ascertainment used by the epidemiologist. It is often not possible to compare incidence rates estimated by different studies. In addition to considerations of accuracy in diagnosis and completeness of data retrieval, some studies do not correct the raw incidence figures for differences in age between populations. Only differences in disease incidence that have been determined using the same methodologies can be interpreted reliably. Accepting the inherent difficulties in collecting accurate data, it has been estimated that gastrointestinal cancers (i.e., colonic, gastric, esophageal, oral) account for four of the seven most frequently occurring cancer deaths observed worldwide and that lethal tumors of the colorectum rank just below cancers of the lung and stomach. [11] The rates are skewed in the United States, where colorectal cancer plays a more important role in cancer incidence and mortality, and stomach cancer is conspicuously low.

Whereas the incidence of colorectal cancers slowly drifted upward in the United States from 1950 to 1990, a slight downward drift in mortality occurred, suggesting that some introduced factor (perhaps the use of screening and colonoscopic polypectomy) resulted in better patient outcomes from this disease. [12], [13] One should keep in mind that incidence and mortality rates have not been constant over time.

No consistent observations of high-risk industries or occupations have been made, and smoking, for example, does not seem to play a role, despite the fact that tobacco use is associated with twofold to threefold increases in adenoma prevalence generally. Alcohol use, specifically beer drinking, is associated with a twofold to threefold increase in risk for colorectal adenomas and a significant increase in risk for colorectal cancer. [14], [15], [16], [17] and [18] The role of alcohol may be exacerbated by concomitant low intakes of methionine and folate. [19] There is a 2- to 3.6-fold elevated risk for colorectal cancers among women who have been irradiated for gynecologic cancer. [20] Asbestos exposure (either inhaled or ingested in the water supply) does not appear to increase the risk for colorectal cancer. [21], [22]

Physically active women have a relative risk for colorectal cancer of 0.54, [23] and virtually identical reductions in the risk for colorectal adenomas or carcinomas has been found in physically active men. [24] Increased total energy intake, which may complicate issues of physical activity, however, may reduce this benefit, [25] as discussed later.

DIETARY PATTERNS

Macronutrient Considerations

Investigators are most suspicious of the role of diet in the causation of colorectal cancer. [26] Dietary composition has been compared among populations with divergent incidences of colorectal cancer. The estimated risk for colon cancer increases by 2.3% for each 100 calories per day, and the total energy intake may be powerful enough to overwhelm other individual components of the diet. [27] Compelling epidemiologic leads have come from studies of the dietary intakes of fat, meat, and fiber, [28] and there is a strong dose-related association between per capita intake of total dietary fat or meat and the incidence of colorectal cancer (Fig. 91-1). However, dietary intervention studies over periods of 3 to 4 years have failed to reduce significantly the incidence of recurrent adenomas. [29], [30]

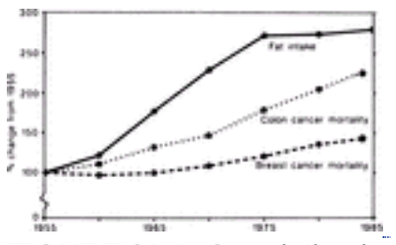


FIGURE 91-1. Correlation between changing fat intake and age-adjusted mortality from cancer of the colon and breast from 1955 to 1985 in Japan, during which time an increase in fat intake of 180% occurred. (From ref. [28].)

Meat and Fat Large epidemiologic studies have examined the role of diet in cancer. [31] In a study of more than 88,000 women, the intake of animal fat was positively associated with colon cancer incidence, with a relative risk of 1.89 for the highest quintile compared with the lowest, which is statistically significant but modest. The relative risk among daily eaters of beef, pork, or lamb compared with those reporting consumption less often than once a month was 2.49. Animal fat from dairy sources was not an independent risk factor because the relative risk for consumption of more than 18 g daily, compared with less than 7 g daily, was 0.91. There was no apparent risk for ingestion of vegetable fat or linoleic acid. Fat intake was not independent of total energy intake as a risk factor in this study. Eating fish or skinned chicken was associated with lower risk as well. Lower intake of fiber appeared to be a contributing dietary factor but was not independent of increased meat consumption. [32] In U.S. men, relative risks for colorectal cancer of 3.57 have been reported for those who ingest beef, pork, or lamb, [33] although other studies have not confirmed this finding. [34], [35] These nutrients may be part of the general phenomenon of a large total caloric intake, and it is not obvious whether these issues are separable. [36]

Serum Cholesterol Serum cholesterol is, in part, a reflection of total dietary intake of fat, and two large studies showed a direct relation between cholesterol levels and colon cancer. After excluding patients with colon cancer at the time of blood collection, a prospective study of 8000 Hawaiian Japanese men indicated that higher cholesterol values were significantly associated with later tumor development, particularly in the proximal colon. [37] A prospective study of more than 92,000 Swedish subjects revealed a positive association between serum cholesterol level and the risk for rectal cancer in men, with a relative risk of 1.65 for levels greater than 276 mg/dL. Similar trends were observed in women, although they were not statistically significant. [38] Decreased levels of high-density lipoprotein cholesterol and increased levels of low-density cholesterol have been reported in patients with colorectal adenomas. [39] Obesity in middle age is associated with increased colon cancer risk in men (relative risk for heaviest compared with lightest quintiles, 2.40); however, increased physical activity appears to eliminate the risk associated with obesity. [40] Women were not examined in this study. Curiously, studies from several countries have reported an inverse relation between serum cholesterol and colorectal cancer risk, [41], [42] and [43] and the Framingham Study reported a fourfold increase in risk for colon cancer among men who were both obese and had low serum cholesterol. [44] This finding may be explained, at least in part, by a case-control study that demonstrated an average decline of 13% in the serum cholesterol of colon cancer patients over a 10-year period compared with controls, who experienced a 2% rise during the same time frame. [45] Serum cholesterol may therefore reflect the increased risk associated with a high-fat diet but may fall by an unknown mechanism during the development of colorectal neoplasia.

How Does Dietary Fat Promote Cancer? The mechanism by which a high-fat diet enhances tumor production appears to be related to the role of bile acids on colonic epithelial proliferation. Increasing the intake of animal fat from 62 to 152 g/day produced a significant increase in total fecal bile acid and fatty acid excretion in humans, without affecting fecal weight, number of stools, transit time, fecal ß-glucuronidase, or fecal steroid degradation. [46] In rats, the intracolonic instillation of deoxycholic acid (DOC) increased cellular proliferation as measured by ornithine decarboxylase activity and new DNA synthesis. The proliferative response to DOC may be abolished by agents that destroy superoxide (such as superoxide dismutase) or inhibit lipoxygenase activity. DOC produces reactive oxygen radicals in the colon, and the generation of such molecular species can independently stimulate mucosal proliferation. [47] Because the oxidation of unsaturated fatty acids produces compounds that may stimulate cell proliferation, the generation of reactive oxygen may be the mechanism by which oxidized fatty acid residues are produced and colonic cell proliferation is stimulated, [48], [49] which could explain why unsaturated fatty acids are more effective in supporting tumor production in animal models and the coordinate role of bile acids and fat in the pathogenesis of colorectal cancer. It has been suggested that fish oil contains fats that protect against colorectal neoplasia. Marine fish oils are rich in unsaturated fatty acids, including n-3 or ?-3 fatty acids (which refers to the location of the double bonds being 3 positions from the end of the chain), in contrast to the n-6 or ?-6 fatty acids that predominate in unsaturated plant lipids. Geographic regions where people have large amounts of marine fish in the diet tend to have lower incidences of colorectal cancer. Furthermore, supplemental marine oils do not promote cancer in animal models. After treatment for 12 weeks with 4.1 g of eicosapentaenoic acid daily and 3.6 g of docosahexaenoic acid daily, subjects with adenomatous polyps showed a rapid contraction of the zone of proliferation into the lower portion of the colonic crypt, although no change in the total rate of proliferation was seen. [50] The mechanism by which ?-3 fatty acids suppress proliferation in the superficial portion of the crypt is not known, and the predictive value of this observation is not proven. Increased ingestion of dietary fat results in an elevation of fecal bile acids. [46] A significant elevation in fecal bile acid concentration has been reported in 82% of patients with large bowel cancer compared with 17% of patients with other diseases. [51] This observation has been confirmed in a wide range of population groups. [52] Patients with colorectal cancer have relatively high levels of unconjugated primary bile acids and neutral animal sterols and relatively low levels of esterified neutral sterols and saponifiable bile acids in their feces compared with a group of nonvegetarian patients without cancer. These differences are even more profound if cancer patients are compared with vegetarians, who have a lower risk for colorectal cancer than omnivores. [53]

Dietary Fiber The Western diet is relatively deficient in fiber compared with the diet of non-Western populations, and this may be important in the pathogenesis of colon cancer. [54] Countries with high fiber intake also tend to have lower intakes of fat and often a lower life expectancy, which introduces confounding variables. Although data obtained in laboratory animals have been somewhat confusing, the epidemiologic observations are more consistent. [55] Some case-control studies have demonstrated a protective effect of dietary fiber, [56] but other studies have failed to confirm this finding. [57] If the entire body of data accumulated on the subject is considered, most papers show a consistent protective effect of fiber, a small number of studies show no effect, and no study shows a deleterious effect of fiber on the incidence of colon cancer. [58] In a prospective questionnaire study of more than 760,000 people, the ingestion of a diet rich in vegetables and high-fiber grains was found to be significantly protective against fatal colorectal cancer, and the highest quintile of intake had relative risks of 0.62 (for women) and 0.76 (for men) compared with the lowest quintile. [59] A metaanalysis of 13 case-control studies that examined the impact of dietary fiber intake on colorectal cancer incidence revealed relative risks of 0.53, 0.63, 0.69, and 0.79 after comparing the four highest quintiles of fiber intake with the lowest ($P = 0.0001$). Although statistically significant, this is a relatively weak effect and not as powerful as the effect of family history, for example. After adjustment for fiber intake, relatively weak independent protective effects also were seen for vitamin C and beta carotene, which underscores the complexity of the role of diet in cancer causation. [60] The intake of nonstarch polysaccharides (i.e., fiber) correlates directly with fecal weight in studies from diverse international populations, and the incidence of colorectal cancer shows a significant inverse correlation with fecal weight (Fig. 91-2). Fecal weights in Great Britain were found to be about 106 g/day (corresponding to a fiber intake of 12.8 g/day). Based on the observation that high-risk Western nations have fecal weights in the range of 80 to 120 g/day, these investigators suggested that an increase in fiber intake to more than 18 g/day would increase fecal weights to more than 150 g/day and might reduce cancer incidence. [61] One trial demonstrated that the addition of 13.5 g of bran fiber to the diet (equal to a 1.5-ounce bowl of a commercially available all-bran cereal) resulted in a significant reduction in rectal epithelial proliferation in a group of high-risk patients with a past history of resected colorectal cancers. [62] Nonetheless, the projected impact on the incidence of colorectal cancer as a solitary intervention is not likely to be dramatic.

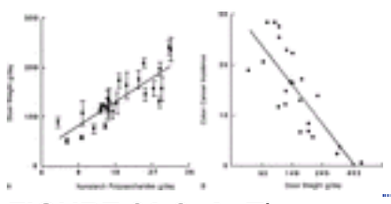


FIGURE 91-2. A: The mean stool weights ± standard error of the mean (*SEM*) in 11 groups of healthy subjects ($n = 206$) on controlled diets containing different amounts of dietary fiber. **B:** The relation between colon cancer incidence (age-adjusted cases per 100,000 population) from 23 population groups in 12 countries, plotted against stool weight in grams per day ($r = 0.78$). (From ref. [61].)

A number of mechanisms have been proposed for the protective effect of fiber against colorectal cancer. Fiber decreases fecal transit time and, by virtue of its sheer presence in stool, tends to dilute the concentration of other colonic constituents per g of stool. Both of these would tend to minimize contact between carcinogens and colonic epithelium. Second, fiber polymers may bind toxic substances and remove them from contact with epithelium. Third, fiber is neither digested nor absorbed in the small intestine but undergoes fermentation in the presence of the colonic flora, which reduces fecal pH and generates short-chain (volatile) fatty acids. [63] Certain short-chain organic acids derived from the fermentation of fiber can protect isolated colonic epithelial cells from DOC-induced injury in culture. [64] One of these, butyric acid, present in high concentrations in the colonic lumen, is thought to be an important energy source for colonic epithelium and has the ability to induce cellular differentiation in certain cultured cell lines.

Micronutrient Considerations

Calcium Cell proliferation normally takes place in the lower half of the colonic crypt. Early colorectal neoplasia has been associated with a shift of the proliferative zone from the lower to the upper portion of the crypt. [65] A generalized disturbance in the normal regulation of cell proliferation has been reported throughout the colons of patients with adenomatous polyps or cancers. [66] Groups at low risk for colorectal cancer, such as Seventh-Day Adventist vegetarians, [67] have low levels of colonic epithelial cell proliferation, and this has been used as an intermediate end-point biomarker for interventions in high-risk groups. Dietary calcium appears to participate in the regulation of colonic epithelial cell proliferation, although the mechanism of this control, and its interaction with bile acids, is not entirely clear. For example, colonic cell damage and epithelial proliferation may be stimulated by the addition of a bile acid, and both of these may be minimized by the supplementation of the diet with calcium. [68], [69] It has been hypothesized that the damaging and mitogenic effects of free fatty acids and bile acids could be reduced in the presence of supplemental calcium by precipitating free fatty acids as calcium soaps. [70] Correction of the hyperproliferative indices by supplemental calcium can also be achieved using epithelial explants in vitro, [71] which indicates that calcium may play an additional systemic role that is not explained by modification of the lumenal concentrations of fatty acids. Proliferative indices in the colons of a small group of patients at risk for familial colonic cancer have been studied with the patients on a conventional diet (assumed to contain about 700 mg of calcium per day) and then studied again after adding 1.25 g of calcium carbonate to the daily diet. Repeat study after 2 to 3 months indicated a reduction of the abnormal colonic proli-ferative indices to almost normal levels. [72] Although this result has been confirmed in another high-risk population, [73] contradictory results have been reported in other controlled trials. [74] The dose of calcium administered and the underlying proliferative activity of the epithelium may be important issues. Supplements of 1200 mg/day of elemental calcium did not reduce colonic epithelial cell labeling index in patients with adenomatous polyps, [75] but a daily dose of 2000 mg of calcium (i.e., 5 g of calcium carbonate) significantly suppressed rectal proliferation. [76] The mechanism by which supplemental calcium corrects the hyperproliferative states seen in patients at high risk for colon cancer has been questioned because of the ability to inhibit DNA synthesis with supplemental calcium in vitro. [71], [77] Although proliferation may be inhibited by supplemental calcium in normal tissues in vitro, growth inhibition is not observed in neoplastic tissues (i.e., adenomas and carcinomas). [77] No clinical trial has suggested more than a modest reduction in neoplasia with calcium supplementation, and large doses can be dangerous to some people, and may contain contaminants.

Selenium Low levels of selenium have been associated with an increased risk for colorectal neoplasia. [78] Selenium is a constituent of glutathione peroxidase, which prevents free radical damage to tissue and, by an independent mechanism, antagonizes toxicity caused by certain heavy metals. Attempts to assess interventions with supplemental selenium are in progress in geographic locations where the selenium content of soil and foods is poor. Excessive doses of selenium can be toxic to humans. Blood samples were obtained from a group of healthy subjects who were monitored prospectively for the development of cancer. This approach demonstrated significantly lower levels of serum selenium in patients who later developed gastrointestinal cancers, but no excess risk was found for patients with lower levels of serum retinoids, carotenoids, or vitamin E, all of which are potent antioxidants and were hypothesized to play a protective role against cancer. [79] Threshold limits for lower levels of these micronutrients are not known, and their roles in cancer prevention remain open questions. Selenium intake has been estimated in 27 countries and has been correlated with cancer incidence at various body sites. Significant inverse correlations were found between selenium intake and cancer of the colon and rectum. [78], [80]

Vitamins A, C, and E A number of vitamins may have anticancer effects. Vitamin A, beta carotene, and vitamin E are known to have powerful antioxidant activities, and retinoids function in certain laboratory models as differentiating agents. Also, a metaanalysis of five cohort studies suggested that those in the highest quartile for serum a-tocopherol (vitamin E) had a matched odds ratio of 0.6 for colorectal cancer compared with the lowest quartile (95% confidence interval [CI], 0.4 to 1). [81] Although it has long been suspected that people who have low serum levels of antioxidants would be at greater risk for certain cancers, prospective studies have been unable to confirm significant negative relationships between serum levels and subsequent risk for gastrointestinal cancer. [79] A 6-month chemopreventive interventional program using daily doses of 30,000 IU of vitamin A, 1 g of vitamin C, and 70 mg of vitamin E (D,L-a-tocopherol acetate) in patients with colorectal adenomas demonstrated a reduction in certain proliferation indices, but prevention of recurrent neoplasia has not yet been proved. [82] A small study of the effects of 400 mg of ascorbic acid and 400 mg of a-tocopherol per day has suggested a small reduction in the rate of colonic polyp recurrence over a period of 2 years. The minor benefit suggests that large numbers of people must be examined over a long period to detect significant benefits from this type of intervention. [83] In another study, a group of patients with familial adenomatous polyposis (FAP) was treated with 4 g of vitamin C plus 400 mg of a-tocopherol per day with or without 22.5 g/day of supplemental fiber from bran cereal. A modest regression of rectal polyps occurred among patients treated with vitamin C, vitamin E, and the high-fiber diet compared with patients given vitamins and a low-fiber diet or only placebo; however, this benefit did not quite reach statistical significance using rigorous analysis. [84] Unfortunately, a fairly large prospective, placebo-controlled clinical trial of antioxidant vitamins (including beta carotene, vitamin C, and vitamin E) failed to reduce the recurrence of adenomatous colorectal polyps. [85]

Dietary Folate and Hypomethylation of DNA Abnormal methylation of DNA has been proposed to play a role in carcinogenesis, and dietary folate and methionine are required for normal methylation. The actual role of this process in carcinogenesis is likely to be complex because some genes may be inappropriately expressed as a result of hypomethylation of its promoter site, whereas other tumor suppressor genes are silenced by hypermethylation of these regulatory sequences. Higher levels of dietary folic acid have been associated with lower risk for colorectal adenomas. [86], [87] A high level of alcohol intake may reduce levels of S-adenosylmethionine, which is required for methylation, and, as mentioned, alcohol intake has been correlated with risk for colorectal neoplasia. It remains to be seen how important dietary folate is in this process.

Other Micronutrient Anticarcinogens A growing list of dietary micronutrients and synthetic compounds have anticancer activity in animal models of cancer. [88] Many of these are blocking agents that prevent carcinogenesis by a variety of mechanisms. For example, benzyl isothiocyanate, a naturally occurring constituent of cruciferous vegetables, prevents the activation of certain procarcinogens. Organosulfur compounds from *Allium* species (i.e., garlic and onion families) also prevent procarcinogen activation, as do monoterpenes found in citrus fruits. A second class of blocking agents enhances the detoxification of carcinogens through phase 1 or 2 enzymatic pathways and are referred to as type A and B inhibitors, respectively. A third class of blocking agents is effective by inhibiting the increased cell proliferation that plays a role in promoting tumor formation, and they are effective inhibitors after carcinogen exposure. Many compounds share this activity, including phenols, flavones, tannins, curcumin, glycyrrhetinic acid, and glucarates. Other agents, referred to as suppressing agents, also act to suppress tumor development after carcinogen exposure but through poorly understood mechanisms distinct from those of the third class of blocking agents. This group includes protease inhibitors, terpenes, and other compounds. Some compounds have activities that both block carcinogens and suppress tumor formation. Many of these agents have been proposed as potential chemopreventive substances for human studies.

Role of Aspirin in Colorectal Cancer It has been appreciated by epidemiologists for some time that aspirin takers suffer fewer cancers than the rest of the population. In animal models of colorectal cancer, aspirin and non-steroidal antiinflammatory drugs (NSAIDs) inhibit the development of tumors. [89] Furthermore, patients with FAP experience a regression of their adenomas after treatment with the NSAID sulindac. [90], [91] and [92] Sulindac is a prodrug that is absorbed and reduced in the liver to the sulfide metabolite, which then is excreted in the bile and delivered to the colon. In 1991, a survey of more than 660,000 people sponsored by the American Cancer Society revealed a significant reduction in colon cancer deaths among aspirin users. Among those who used aspirin more than 16 times per month, the relative risk was 0.60 for men and 0.58 for women. [93] Risk reduction was seen even in people who reported aspirin use as rarely as once per month, raising interesting speculation regarding the mechanism and dose effects of this intervention. Subsequent studies of nearly 48,000 male health professionals confirmed a relative risk of 0.51 among those who used aspirin more than twice weekly; [94] however, a similar study of more than 500,000 nurses using this dose of aspirin incurred no measurable benefit unless they had used the drug consistently for 20 years or more. [95] A smaller, hospital-based case-control study of colorectal cancer patients estimated that the relative risk of cancer was 0.5 (95% CI, 0.4 to 0.8) among aspirin users, again suggesting that a longer duration of use is more protective, and indicated no protection for nonregular users or for subjects who had discontinued use more than 1 year before the study. [96] A significant reduction in adenoma recurrence (odds ratio, 0.52; 95% CI, 0.31 to 0.89) was found at the 1-year interval after initial colonoscopy among consistent aspirin users but not in the group of intermittent users; this result was independent of the number of initial polyps. [97] Significant reductions of about 40% for deaths from cancer of the rectum, esophagus, and stomach were reported recently from the large American Cancer Society survey. [98] The protection was greatest among long-time users (i.e., longer than 10 years) and was not found for tumors of nondigestive organs. The concept that NSAIDs may be of benefit in colorectal neoplasia came from the unexpected impact of sulindac on existing polyps in FAP (see Chapter 90). Unfortunately, reports on the use of this drug in patients with sporadic colonic polyps have provided divergent results, [99], [100] and no pharmacologic regimen can yet be recommended as an effective cancer-preventing intervention. The mechanism of action by which aspirin and NSAIDs protect against tumor formation in the digestive tract remains an issue of speculation and includes many possibilities. [101] It cannot be assumed that the antitumor effect is necessarily related to modifications in the production of prostaglandins in the gastrointestinal tract. In fact, an oxidized derivative of sulindac (sulindac sulfone) has the ability to inhibit polyp formation in the animal model even though it has no inhibitory effect on prostaglandin formation. There is growing evidence that NSAIDs may induce programmed cell death (apoptosis) in some early adenomatous cells but not in carcinoma cells. [102] One concept that initially challenged the prostaglandin-related hypothesis was the demonstration that NSAIDs increase the arachidonic acid in cells (due to inhibition of its conversion to prostaglandins), which stimulates the conversion of sphingomyelin to ceramide, a mediator of apoptosis. [103] Additional work has shown that NSAIDs may also inhibit some of the downstream effects that occur after disruption of the *APC* gene, which is an early event, mediating the appearance of the adenoma. [104] Also, combinations of NSAIDs with other agents have been found to be potent inhibitors of adenoma formation in the experimental model of FAP, raising novel therapeutic options for the future. [105]

Recommendations Although there are wide ranges in the international incidences of colorectal cancer, there is a fairly uniform, high rate of this disease in North

America, Western Europe, and much of the industrialized world. Groups migrating from low-risk into high-risk regions rapidly experience an increase in the incidence of colorectal cancer. It is a reasonable assumption that diet plays the greatest role in determining the incidence of colorectal cancer in the population. High fat intake and low fiber intake have been the most consistent associations with colorectal cancer, but the degree of risk attributable to these nutrients is modest. Unfortunately, interventions in which dietary fat has been reduced, fiber increased, and fruits and vegetables increased have not been able to reduce significantly the rate of recurrent adenomas over periods of 3 or 4 years among patients who have had adenomatous polyps removed. [29], [30] Several different interpretations are possible. First, it may be that 4 years of a new diet is not sufficient to alter the emergence of adenomas. It is possible that the intended intervention was not adequate and that insufficient changes were achieved in the diet. It is considered a reasonable recommendation that a healthy diet consist of less than 25% fat, include perhaps 25 g of dietary fiber, and be enriched with fresh fruits and vegetables, [106] but there is not yet any evidence that this will reduce the incidence of or mortality due to colorectal neoplasia.

ETIOLOGY

Colorectal Cancer Development through Multistep Carcinogenesis

Carcinogenesis in the colon and rectum initially was described in terms of the classic initiation-promotion model formulated more than four decades ago to explain carcinogenesis of the skin (see Chapter 24). According to this conceptual framework, the first step involved factors that directly damaged DNA, mutated the genome, and initiated the cells. The process was driven to completion by promotional factors that were not themselves mutagenic. It was thought that the two components of the process were strictly sequential in that promotional factors were effective only after the administration of initiating agent.

This model was replaced by a new model of multistep carcinogenesis driven by mutations at multiple loci. Colorectal neoplasia is best characterized as a process that begins with genetic mutations in critical growth-regulating genes that either enhance proliferation or prevent death, which provides growth and survival advantages for affected cells. As the cells expand their numbers, they accumulate additional mutations, some of which may be lethal but certain ones of which further enhance the ability of cells to proliferate. As the most advantageous genetic changes occur, these cells overgrow neighboring cells. Through successive waves of clonal expansion, the mutations appear to accumulate in the expanding clones. After the accumulation of sufficient genetic damage, and given a sufficient amount of time, the neoplastic phenotype is likely to occur, and the ability to invade and metastasize eventually emerges.

Caretaker and Gatekeeper Genes

The genetic events involved in carcinogenesis are typically inactivations of genes that have been characterized either as caretakers or gatekeepers, which helps to illustrate the functional nature of the genes involved. [107] This concept acknowledges two essential concepts of neoplasia: first, the genome of the neoplastic cell is unstable; second, mutations have occurred at critical growth-controlling genes. Loss of caretaker genes results in genomic instability, which increases the likelihood that mutations at other genes (including gatekeeper genes) will accumulate. Mutations tend to occur randomly, however, and most will have no effect on the growth potential of a cell; considering the demands of evolution, most mutations are likely to be deleterious. As illustrated by the fact that an entire metazoan organism may be created by cloning a somatic cell, it should be apparent that every cell has the theoretical capability of unlimited growth. A substantial number of our genes are expressed for the purpose of restraining this potential for growth, and these are called *tumor suppressor genes*. The inactivation of critical genes permits the cell to express normally forbidden behaviors, which is the basis for the gatekeeper concept. Each cell type appears to be controlled by a limited number of cell-specific gatekeepers. In a multistep process as occurs in the colon, the incremental changes in behavior gradually lead first to the formation of an adenoma and later to the evolution of carcinoma. Furthermore, some carcinomas may grow large without metastasizing, whereas others spread early in their course. The time required for these changes to occur depends on the degree of genomic instability and on chance. These issues are addressed in more detail in Chapter 24; the genes involved in colorectal carcinogenesis are discussed in the next section.

The Adenomatous Polyposis Coli Gene Is a Gatekeeper for the Colorectal Adenoma Much of what we know about colorectal carcinogenesis has been learned from studies of the familial syndromes FAP and hereditary nonpolyposis colorectal cancer (HNPCC). [108] FAP is caused by a germ-line mutation in the adenomatous polyposis coli (APC) gene, which is located on chromosome 5q. [109], [110], [111] and [112] Patients with FAP begin to develop adenomatous polyps in great number by early adolescence, and evidence has emerged that *APC* is the gatekeeper for the formation of adenomas. Furthermore, mutations in the *APC* gene are found in the smallest adenomas [113], [114] and do not appear to be responsible for later events in tumor progression. [114], [115] and [116] The alterations in the *APC* gene are almost always nonsense mutations, that is, those that result in a premature stop codon and consequently the production of a truncated *APC* gene product. Characteristically, adenomatous polyps also show loss of the nonmutated (or wild-type) *APC* alleles, called loss of heterozygosity (LOH, see later), as one would expect from the requirement for a two-hit mechanism at this tumor suppressor gene locus, in which both parental alleles must undergo inactivation. [114], [115] The APC protein appears to be involved in regulating cell death, which is discussed in Chapter 90.

K-*ras* Gene Mutations Are Associated with the Progressive Growth of Adenomas Most adenomas remain small and never progress beyond a benign stage. Growth and progression require additional disruption of growth controls. Mutations have been identified in the K-*ras* cellular protooncogene, and this has been demonstrated in larger adenomas, leading to the interpretation that this mutation may permit them to grow larger. [115], [116] and [117] The *ras* genes are highly conserved in nature; they encode for proteins located on the inner leaflet of the plasma membrane, bind guanine nucleotides, and are involved in signal transduction from the cell membrane to the nucleus. [118] Mutations located at critical positions in the gene alter the Ras protein in such a way that the signal transduction is constitutively activated, which leads to additional cell growth. A mutation at just one of three codons—12, 13, and 61 of the K-*ras* gene—is the mechanism by which *ras* is activated in many colorectal neoplasms. [119] For example, in one study of human tumors, mutations of K-*ras* were present in 10 of 27 colorectal cancers, and in 9 of these instances, the mutation occurred at codon 12 of the gene. [120] Mutation at codon 12 was found in 40% of 66 primary human colon cancers. The transforming mutations all occurred in the first or second of the three nucleotide positions of the codon but were not found elsewhere in the gene. [121] Not all colorectal cancers have K-*ras* mutations, and it is likely that mutations in genes encoding for other members of the signal transduction pathway occupied by K-*ras* may account for the constitutive activation of this signaling pathway in the absence of mutations in the K-*ras* gene itself. K-*ras* mutations appear to mediate a specific growth pattern and are associated with exophytic adenomas and carcinomas. [122]

The *p53* Gene Is a Gatekeeper for the Transition from Adenoma to Carcinoma Large adenomas are benign neoplasms, and despite the fact that they occasionally grow to a size of 6 cm or more in diameter, adenoma cannot invade the muscularis mucosae. The gene most responsible for malignant conversion appears to be the *p53* gene, which is activated by the two-hit mechanism characteristic of tumor suppressor genes. [115], [116] In the stepwise process, *p53* is the gatekeeper gene for the adenoma-to-carcinoma transition. [114] The *p53* gene serves as a cell-cycle checkpoint regulator and prevents nuclear replication after injuries that are likely to damage the DNA. [123] In the presence of damaged DNA, the p53 protein level rises in the cell, and progression into the cell cycle is prevented by G_1/S arrest. The cell either repairs the damage, or apoptosis (i.e., programmed cell death) ensues. Inactivation of the *p53* gene permits mutated DNA to be replicated and removes a level of restraint on abnormal cell behavior. Unlike what occurs in the *APC* gene, mutations in *p53* are usually missense; that is, they encode for a different amino acid somewhere in the protein chain. Truncating mutations do not occur commonly; rather, the aberrations tend to stabilize a nonfunctional p53 protein. The *p53* mutations tend to occur in hot-spot locations in colon cancers (codons 175, 248, and 273), and virtually all the mutations occur in one of four evolutionarily conserved regions of the gene located between codons 117 and 286. [124] This region of the p53 protein is responsible for DNA binding, and oncogenic forms of the protein lose the ability to bind DNA and activate gene expression. [125] Mutations in *p53* occur in a wide range of human tumors, [126] and this is perhaps the most common and most powerful tumor-causing genetic lesion. The mutational hot-spot region is where viral oncoproteins and at least one normally expressed regulatory protein bind and inactivate the function of *p53.* [123], [124], [127] In normal cells, *p53* is expressed at very low levels, has a short half-life, and is not readily detectable. Certain mutations in the *p53* gene result in the expression of a more stable but dysfunctional p53 protein that may be detected using standard immunohistochemistry. Overexpression of *p53*, as evidenced by observing the protein histochemically, may be seen in some colorectal adenomas, [128] but the biologic significance of a single mutated *p53* gene is uncertain if there is still a second, normal allele present in the nucleus because this is sufficient for normal cell growth. A second event occurs in which the normal or wild-type *p53* allele is lost, leaving the cell in the hemizygous, mutated state. As mentioned, the combination of a mutation in one allele, together with loss of the other allele, is the genetic combination responsible for the conversion of an adenoma to a carcinoma. [114], [129], [130] and [131] This conversion must occur in the context of an adenoma, which, as described, already has lost both copies of the *APC* tumor suppressor gene. The order of events appears to be critical in this instance because patients with a germ-line mutation in the *p53* gene (i.e., those with Li-Fraumeni syndrome) have only a modest increase in colorectal cancer. [132] Furthermore, mice who have had both copies of *p53* deleted from their genome have no tendency to develop gastrointestinal tract tumors but are at risk for lymphomas. [133] Overexpression of the p53 protein in colorectal adenomas is more prevalent in more highly dysplastic polyps and correlates with the proliferative activity of the lesion. [128] Increased expression of p53 and loss of the wild-type allele are associated with worse prognosis in colorectal cancer independently of other pathological features. [134], [135] There is evidence that loss of the wild-type allele is first seen in high-grade dysplasia (i.e., carcinoma in situ) and is associated with the onset of chaotic genomic instability, in which genetic material is randomly and rapidly deleted from the nucleus, resulting in LOH. [114]

Other Genetic Alterations in Colorectal Cancer Other genetic abnormalities occurring in colorectal neoplasia may participate in multistep carcinogenesis. For

example, although mutation in the *myc* oncogene has not been reported, this oncogene is amplified in some colon cancers. [136], [137] Although amplified expression of the *myc* RNA has been reported in more than two thirds of primary adenocarcinomas of the colon, levels of expression do not correlate with adverse patient prognosis. [138] Hypomethylation of DNA may lead to the altered expression of genes that are neither mutated nor amplified at the genomic level. This genetic modification has been reported in colorectal neoplasms and is found to a similar degree in benign adenomatous polyps. It may be one of the earlier genetic modifications in colorectal neoplasia. [139] The cellular protooncogene *src* is activated by mutation, resulting in increased tyrosine kinase activity, which is associated with increased cell proliferation, similar to that described for mutated K- *ras* genes. Increased protein kinase activity of activated *src* is an early event in the progressive neoplastic sequence and is seen in premalignant lesions such as adenomatous polyps. The activity of the enzyme is associated with the malignant risk for the polyp. [140] Several other oncogenes are mutated or otherwise abnormally regulated in colorectal cancer, but their roles in the overall scheme of colorectal carcinogenesis remain to be elucidated.

Genomic Instability in Colorectal Cancer

Colorectal cancers evolve by the accumulation of genetic alterations at loci involved in the control of cell growth. There is not one single human gene that, by itself, can cause cancer by virtue of mutation because of the redundancies that have evolved in the human genome. The fact that cancer occurs at all is best explained in the context of genomic instability and the progressive accumulation of mutations. First, a genetic caretaker function is lost that destabilizes the normally reliable process of DNA replication, resulting in genomic instability. Genomic instability, a genome-wide disorder, accelerates the accumulation of mutations throughout the nucleus. Many mutations are either neutral or deleterious and may not be relevant to the neoplastic process. A mutation eventually will occur that creates a growth advantage that is censored in normal cells, and this cell will overgrow its neighbors. Over time, additional mutations will accumulate that add to the growth advantage of the unstable clone. There are two types of genomic instability: one that results in loss of heterozygosity, and another that leads to microsatellite instability (MSI).

Chromosomal Instability Leads to Loss of Heterozygosity The type of genomic instability found in most colorectal cancers is characterized by aneuploidy, the loss of chromosomal segments throughout the genome, and LOH. [141], [142] In this poorly understood process, the nucleus no longer can symmetrically divide its chromosomes during mitosis. A variety of chromosomal rearrangements occur. In the case of LOH, one daughter cell receives more copies of a genetic segment, and the sister cell gets fewer. Increased numbers of lost genetic loci (expressed as the “fractional allelic loss”) are significantly associated with worse prognosis in colorectal cancer [141] because this increases the chance that LOH will occur at specific chromosomal locations such as 17p (the locus of the *p53* gene) and 18q (the locus of the several potential metastasis suppressor genes). [142], [143] Many of the lost genetic segments are random and may not be important for the malignant behavior of the cell, but in certain instances, LOH results in the loss of copies of tumor suppressor genes. If critical growth-restraining genes are lost, the altered cell gains an advantage in growth and survival over other cells, and undergoes clonal expansion. Widespread LOH events are characteristic of colorectal and other cancers, and the changing genetic background results in the generation of cellular diversity and, eventually, the metastatic phenotype with a primary tumor mass. Candidate genes have been proposed for various biologic characteristics of the metastatic phenotype, but the metastasis genes remain a complex and incompletely understood area.

Loss of DNA Mismatch Repair Activity Causes Microsatellite Instability At least two distinct pathways to colorectal neoplasia involve different mechanisms of genomic instability. In the first instance, which involves most colorectal cancers, there is the loss or gain of whole chromosomes or fragments thereof. [144] This has been called *chromosomal instability*. The existence of a second genetic pathway for colorectal cancer was discovered during the search for the gene for HNPCC or Lynch syndrome. This familial colon cancer gene was found using linkage analysis to map the phenotype to a specific genetic locus. This search required the use of genetic polymorphisms called *microsatellites*, which are short, repeated DNA sequences. Although the length of these sequences varies from person to person, they are stable and uniform throughout the cells of any individual. These sequences, which are interspersed more than 100,000 times throughout the genome, were found to be highly susceptible to mutation in HNPCC tumors, the phenomenon of MSI. [145], [146] This type of hypermutability also was found in about 12% of sporadic, nonfamilial colorectal cancers, particularly those in the proximal colon. [145], [147], [148] Tumors with MSI did not demonstrate the chaotic nuclear disorganization produced by LOH, and the two pathways with different types of genomic instability appear to be somewhat mutually exclusive. Furthermore, tumors with MSI are more likely to be diploid and are associated with a significantly better prognosis. [147], [149] Most of the tumors with MSI are not HNPCC but rather represent an acquired epigenetic lesion that mimics HNPCC but that tends to occur in older patients. [150]

Clinical Implications of Specific Genetic Lesions It is likely that many of the differences in clinical behavior of tumors are based on variations in the appearance and accumulation of specific genetic alterations. It has long been known that patients with HNPCC have a significantly better prognosis, when matched stage for stage against sporadic cancers. [151] Furthermore, when young patients (50 years of age or younger) have tumors with MSI, their survival is significantly better. [152] Also, HNPCC cancers are significantly more likely to be mucinous, to look poorly differentiated, and to have a Crohn’s disease–like lymphocytic infiltration, [153] features that are typically overrepresented in all tumors with MSI. In fact, the acquired form of MSI is almost always found in the proximal colon, where the majority of HNPCC tumors are. [154] As mentioned previously, tumors with MSI do not have chromosomal instability and, as a result, are usually diploid. [155] K- *ras* mutations are found rarely in tiny adenomatous polyps but are found increasingly with the larger sized polyps. K- *ras* mutations are found in about 50% of colorectal cancers and are significantly associated with exophytic adenomas compared with flat ones, and with polyploid cancers compared with cancers that arise de novo in flat mucosa. [122], [156]

Genetic Alterations in Cancers Complicating Ulcerative Colitis In specialized clinical situations, such as cancers complicating ulcerative colitis, different mechanisms may be operating. These tumors are significantly less likely to have K- *ras* gene mutations, [157], [158] but they show a high frequency of LOH, including the *p53* locus. [159], [160]

Aneuploidy

Because of the inability to identify histologic features of prognostic significance in most colorectal cancers, diagnostic approaches have been attempted to select patients at greatest risk for metastasis. Flow cytometry can be used to measure the DNA content of cell populations and to estimate the percentage of cells in different stages of the cell cycle. Resting cells are normally diploid. After the stage of new DNA synthesis is complete, the cells are briefly tetraploid, and after mitosis, they return to the diploid state. During the phase of DNA synthesis (S phase), the DNA content of a cell is transiently between the diploid and the tetraploid state. Aneuploidy is a state in which a population of cells has a stable but irregular DNA content, which appears as a spike of nondiploid, nontetraploid cells on flow cytometric analysis. About 80% of solid tumors contain aneuploid populations; several studies of colon cancers agree that about 65% are aneuploid. [161], [162] Furthermore, a smaller percentage (6% to 19%) of adenomatous polyps are aneuploid. [162], [163] and [164] Correlations between aneuploidy and state of differentiation or patient survival have been reported [165] but are on the whole inconsistent, and the studies in colorectal polyps add little to the current histopathologic classification of these lesions. Aneuploidy appears to occur early in the natural history of neoplasia (i.e., it is apparent in larger adenomas); however, as many as 40% of malignant tumors may never become aneuploid despite their ability to metastasize. [166] Although this remains an area of research interest, no clear clinical benefit is gained by measuring ploidy in colorectal tumors.

Mutagenesis

It is apparent that colorectal cancer begins with mutations in the nuclei of colorectal epithelial cells. The human diet contains a large number of naturally occurring mutagens and substances that may be metabolized into mutagens. Most of these mutagens are toxic chemicals synthesized by plants as a primary defense against attack by bacteria, fungi, insects, and other animals. The presence of these toxic compounds is so ubiquitous that an efficient and redundant network of protective mechanisms is present throughout the gut mucosa to detoxify these compounds. In fact, the ability of the enterocyte to metabolize certain xenobiotics is amplified by their presence in the diet. [167] The metabolic fate of procarcinogens is complex and may involve absorption from the gut, metabolism in the liver, secretion into the bile, and additional oxidative activation in the immediate vicinity of the target organ. [168], [169]

Animal Models of Colon Cancer

Much has been learned from rodent models of colonic carcinogenesis. The most commonly used model involves the administration of a member of the dimethylhydrazine family of carcinogens to rats or mice, which results in adenocarcinomas of the colon, small intestine, and other sites, depending on the carcinogen used, method of administration, and species of rodent. In the rat, adenocarcinomas occur throughout the proximal and distal large intestine, [170] although this may vary among rat strains. In certain inbred mouse strains, the tumors occur predominantly in the distal colon, and the characteristic adenoma-carcinoma sequence is seen. [171], [172], [173], [174] and [175]

Despite the conceptual limitations of the initiation-promotion model, these terms are referred to frequently in descriptions of the animal model. In this context, *initiation* refers to the period during which the carcinogen or mutagen is administered, and *promotion* refers to any manipulation that takes place in the postinitiation period. Tumorigenesis can be enhanced or reduced in experimental animals either by maneuvers that modify the generation of mutagen or by events that occur long after

administration of carcinogen, which either stimulate or inhibit the growth of dormant clones of mutant cells.

Role of Diet in Animal Carcinogenesis

The rodent model of colon carcinogenesis has been widely used for introducing dietary manipulations and observing their effects on tumor production. Increasing the total dietary fat from 5% to 30% of caloric intake produces a significant increase in the number of animals that develop cancers and the number of tumors per animal.[176],[177],[178] and [179] If rats are fed a high-fat diet more than 1 month after the last injection of carcinogen, significantly more tumors are produced than if the high-fat diet is given only during the period of administration of carcinogen.[176] This finding suggests that a high-fat diet mediates some aspect of neoplastic progression in the post-initiation period.

Polyunsaturated fats initially were thought to have greater tumor-enhancing effects than saturated fats for colonic as well as other tumors, such as mammary tumors. Differences between types of fat disappeared at the 20% level, however, at which a higher overall incidence of tumors was produced in all animals.[179] A diet supplemented with 4% to 22.5% menhaden oil, a lipid mixture derived from fish and seals that contains high amounts of polyunsaturated fatty acids of the n-3 or ?-3 series, had no tumor-enhancing effects compared with corn oil.

Bile acids also promote the development of tumors in the rodent model. The addition of cholic acid to the diet increased the number of rodents that developed tumors, the number of tumors per animal, and the fecal excretion of secondary bile acids.[180] Enhanced tumor production may result from other maneuvers designed to increase colonic exposure to bile acids, including ileal resection,[181] cholecystectomy,[182] and distal repositioning of the papilla of Vater.[183]

Fiber is a generic term and includes a wide variety of nondigestible soluble and insoluble forms of carbohydrate (e.g., cellulose, hemicelluloses, gums, mucilages, pectins) and other substances, such as lignins.[63] Bran is a heterogeneous preparation from the seed coats of cereal grains composed of fibers and other contaminating substances, some of which may have effects on epithelial proliferation. Cellulose is a ß-linked polymer of glucose found in virtually all plant tissues and is a common component of many forms of fiber.[184] Purified cellulose, in doses ranging from 4.5% to 15% of the diet, reduces tumor production in rats.[185],[186] and [187] Dietary cellulose decreases mitotic indices in the colon,[188] whereas virtually opposite effects are seen with pectin, which provides no protection against tumor production.[186] Dietary histories regarding fiber intake are difficult to estimate in humans, and the data are correspondingly confusing. The experimental model has permitted a dissection of the issues and has clarified the fact that certain components of fiber may be protective while others are not.

Administration of a 10% wheat bran diet inhibits tumor production and, in fact, can prevent the tumor-enhancing properties of bile salts.[189] Administration of a 20% wheat bran diet enhances cellular proliferation in the colon,[190] however, and either has no effect[191] or actually increases tumor production in the rat.[192] The tumor-enhancing effect is seen only if 20% wheat bran is administered during the initiation period, whereas a reduction in benign tumors is seen if this diet is administered after the administration of the initiating agent.[192] The administration of a 14% corn bran diet increases fecal bile acid excretion and enhances tumor production, reinforcing the conclusion that not all types or doses of fiber are equally protective against colon cancer.[193] The proliferative response elicited by 20% wheat bran[188] also can be produced by a diet of 10% guar, another fiber.[194] Two other fibers, 20% oat bran and 10% pectin, have no effect on colonic proliferation.[194]

The animal model also has permitted exploration of factors that may modify large bowel carcinogenesis, as summarized in Table 91-2[183],[192],[193],[195],[196],[197],[198],[199],[200] and [201] and Table 91-3.[186],[189],[193],[202],[203],[204],[205],[206],[207],[208],[209],[210],[211],[212],[213],[214],[215],[216],[217] and [218] Manipulations that increase the delivery of lipid and bile acids to the colon, such as the administration of cholestyramine[195] or neomycin,[200] small intestine resection,[201] and biliary diversion to the colon,[183] all increase the number of tumors occurring in experimental animals.

Cholestyramine
Suture material
Experimental colitis
Vitamin A deficiency
Neomycin
Small intestine resection
Diversion of bile to colon
Wheat bran, 20% of diet during initiation
Corn bran, 15% of diet

TABLE 91-2 Factors that Increase Experimental Large Bowel Cancers in Rodents

Indomethacin, aspirin, piroxicam, sulindac, most NSAIDs
Cyclooxygenase-2 inhibitors
Dietary fiber
Wheat bran, 10% of diet
Cellulose, 4.5%-9% of diet
Lignin, 7.5% of diet
Selenium
Vitamin C
Vitamin E
Retinoic acid
β-carotene
ε-Aminocaproic acid
Antibiotics
Dioctylsulfosuccinate
2-Difluoromethylornithine
β-Sitosterol
Butylated hydroxyanisole
Cabbage
Diallyl sulfide (garlic)
Disulfiram

TABLE 91-3 Factors that Decrease Experimental Large Bowel Cancers in Rodents

A large number of factors have been identified that inhibit colonic carcinogenesis, and in some instances, inhibition is specific to either the induction phase or the promotion phase. Antioxidants, such as vitamin C, vitamin E, vitamin A, butylated hydroxyaminisol (BHA), and butylated hydroxytoluene (BHT), do not prevent DNA damage and seem to exert their protective effects during the postinduction promotional period. Much of the current laboratory research of colon cancer has been performed on newer models in rodents of FAP or HNPCC, which develop through different types of genetic alterations and may require different types of preventive approaches.

Clinical Manifestations and Risk Factors for Colorectal Neoplasia

Clinical Presentation

Tumors of the colon and rectum grow at a slow, somewhat unpredictable rate. It is difficult to make any estimates of growth rates because colorectal tumors are rarely left in place for observation. Some information from the precolonoscopy era suggests that about 11.4 years may elapse before polyps with mild atypia become malignant, although polyps containing severe dysplasia become malignant in 3.6 years.[219] Colorectal neoplasms develop cellular heterogeneity with time because of genomic instability. As a result, some tumors grow more rapidly than others, and the rapidity of progression is not uniform. Colorectal neoplasms begin as adenomas and progress through stages of advancing degrees of dysplasia as they grow. *Hypothetical* modeling suggests that a tiny polyp requires about 2 to 3 years to reach a size of 1 cm.[220] The period from the mean appearance of adenomas to the mean diagnosis of cancer in FAP is 23 years.[221],[222] These estimates cannot be generalized to all neoplasms because they are based on selected lesions that have become malignant, and they fail to take into account the important consideration that many adenomas may never grow. These estimates serve to underscore the fact that most colonic tumors grow relatively slowly and tend not to give rise to dramatic or characteristic symptoms. Therefore, changes in bowel habits may be a sign of distal colorectal cancer, even if the symptoms have developed over a period as long as 1 year. The presence of symptoms for up to 1 year before diagnosis seems to have no adverse effect on survival.[223] Unsurprisingly, comparing colorectal cancer patients with controls in a retrospective case-control study, no significant differences were elicited in the historical frequency of bowel movements,

the presence of constipation, or the use of laxatives. [224]

As a colon cancer grows, it may give rise to one or more of four different groups of symptoms. First, the colonic lumen may be obstructed, relatively or completely, and produce corresponding symptoms. Obstruction may produce abdominal distention, pain, or, in its most extreme degree, nausea and vomiting. Most colon cancers grow as an expanding circular lesion; the traditional apple-core lesion occurs after the diameter of the tumor approaches the circumference of the bowel wall and obstructs the lumen. The colonic diameter is greatest in the cecum and ascending colon, where obstruction is much less likely to occur. The diameter is less in the transverse, descending, and sigmoid colons, which are more likely sites for obstruction. Gastrointestinal obstruction suggests the presence of a large tumor and is an ominous symptom; it is associated with a significant adverse effect on survival. [225]

Second, as colonic tumors expand into the bowel lumen, they tend to bleed, both because of the presence of abnormal vasculature and because of trauma from the fecal stream. Bleeding is likely to occur regardless of the location of the tumor, but it is usually not brisk. In fact, colon cancers typically lose considerably less than 6 mL of blood per day. [226] If a tumor is located near the anus, the blood may be deposited on the surface of the stool and may be grossly visible to the patient. More typically, the blood is mixed in with the stool and evades detection. Tumors in the proximal colon tend to grow larger without producing obstructive symptoms; thus, they may bleed longer and present with iron deficiency anemia. Tumors in the sigmoid colon or rectum are more likely to produce hematochezia or give rise to a positive fecal occult bleeding test (FOBT).

Obstruction and bleeding reflect complications of tumor growth into the lumen; however, an invasive tumor eventually penetrates the muscularis propria and invades adjacent tissues, which can lead to the third symptom complex of pain and other specific symptoms, depending on the organ invaded. Local invasion may produce tenesmus in the rectum, urinary symptoms (including pneumaturia) in the bladder, nonspecific symptoms in the pelvic organs, or an acute abdomen from colonic perforation. Invasion by rectal cancers into the perirectal fat may be associated with rapid extension and can produce ureteral obstruction. Invasion of other adjacent organs is less common, but the tumor may extend through the mesentery and compromise a vascular structure or, rarely, create a fistula between the colon and the small intestine or stomach. Manifestations of local extension all indicate invasion through the muscularis propria and are associated with an adverse prognostic implication. A metastatic lesion also may produce local symptoms because of its expansive or penetrating qualities. In these instances, the clinician may be drawn to the liver or bony site because of pain and later find a primary tumor in the colon that has produced a minimum of symptoms.

Finally, some tumors produce a wasting syndrome that is out of proportion to the size of the tumor, and normal resting energy expenditure, despite excessive metabolic activity of the tumor itself. Cancer cachexia is characterized clinically by a loss of appetite, weight, and strength. [227] Cancer cachexia is common in patients with any gastrointestinal malignancy, and affected patients may experience a more profound loss of subcutaneous fat than that caused by an equivalent degree of benign inflammatory disease, accounting for the characteristic general appearance of a cancer patient. [228]

Three of the four symptom complexes usually are indications of advanced stages of disease. The only exception is occult gastrointestinal bleeding. Of patients with colorectal cancer who present with symptoms at the time of diagnosis, most will have advanced disease and probably will die of their cancer. Therefore, strategies developed to reduce the morbidity and mortality of colorectal cancer have attempted to detect the excess bleeding caused by the neoplasm. Early forms of colorectal cancer lose blood at rates that are only minimally greater than normal rates of blood loss, however. Thus, it has been difficult to develop accurate and sensitive tests for early diagnosis. [226] This difficulty has placed additional emphasis on the improvement of tests for occult fecal blood and for the identification of subgroups of patients at greater risk for this disease. No substance has been detected in the serum that correlates with the presence of early cancer. Novel attempts are underway to look for altered genes shed from the tumor that can be measured in the stool. [229] The role of this type of testing in a clinical screening program is being evaluated.

Familiality and Colorectal Cancer

The principal feature used to identify higher-risk patients for co-lorectal cancer is the family history. This is not a straightforward issue because 5% of the U.S. population develops the disease, and depending on the number of relatives known, at least 10% of people report one first-degree relative who has developed colorectal cancer. Therefore, additional information is needed to determine increased risk based on family history. Key issues are the number of affected people in a family, the age at the time of tumor development, the number of first-degree relatives who *did not* develop cancer (a point often overlooked), and associated syndromic features. This issue is best understood first by considering syndromic familial cancer syndromes in which the genetic basis is understood, with the realization that much of the weaker forms of familiality may represent variations on these themes.

Familial Adenomatous Polyposis and Gardner Syndrome

The premier candidates to develop colorectal cancer are people who inherit a germ-line mutation (not to be confused with the somatic mutations that occur in sporadic tumors) at the *APC* locus and therefore have FAP or Gardner syndrome. These two conditions are variations on a single genetic disease and are described in detail in Chapter 90. FAP and Gardner syndrome patients develop adenomatous polyps at a median age of 16 years, [221] and virtually all of them progress to cancer, with a median age of 39 years for symptomatic malignancy. [222] This group of patients represents the extreme end of the spectrum and serves as a paradigm of the adenoma-carcinoma sequence. The sequence is terminated after the colon is removed or the patient dies of a complication of the disease; it is therefore not possible to know whether every adenoma would enlarge or undergo malignant degeneration with time. In older patients with this disease, it appears that many or most adenomas simply remain small. Genetic testing and counseling have now become a part of standard care for patients with these conditions. [231], [232]

It has now become apparent that the location of germline *APC* mutations correlates with FAP disease manifestations, thus giving clues to the functional domains of this gene. A mild form of FAP, called attenuated FAP, has now been described and is associated with mutations at the extreme proximal or distal end of the *APC* gene. [233], [234] and [235] Extremely dense polyposis correlates with mutations in the central portion of the gene, [236] whereas a trivial retinal lesion occurs when mutations are found distal to exon 8 through the midportion of exon 15. [237] A condition previously called *flat adenomas syndrome* is now known to be *attenuated FAP*, based on genetic testing. [238], [239], [240] and [241] Finally, an *APC* mutation that causes hypermutability of the allele on which it resides gives rise to a mild colon cancer susceptibility in about 6% of Ashkenazi Jewish people. [242]

Hereditary Nonpolyposis Colorectal Cancer or Lynch Syndrome

HNPCC is an autosomal-dominant disorder characterized by the occurrence, within a family, of multiple cases of colorectal cancer in the absence of gastrointestinal polyposis. [151], [243], [244] The population prevalence of this syndrome is not yet clear, but it accounts for 2% to 3% of all colorectal cancers. [245] Before the identification of the genetic basis of this syndrome, the disease could be recognized only by obtaining a history of the familial aggregation of colorectal cancers with an early age of onset, an excess of proximally located tumors, multiple primary tumors, and an excess occurrence of cancers in certain other organs, particularly the endometrium. The recognition of MSI in the tumor tissue, which is present in almost all cancers from HN PCC patients (and in about 12% of sporadic cases), led to a series of discoveries that linked this hypermutability to a defect in the DNA mismatch repair system. Independent investigators identified a small number of HNPCC-causing genes from a larger number of DNA mismatch repair genes, not all of which can cause HNPCC (Table 91-4). HNPCC can be linked to germ-line mutations in *hMSH2* and *hMSH6* (which are homologs of the prokaryotic DNA mismatch repair gene *mutS*) in *hMLH1*, and possibly in *hMLH3* and *hPMS2*. The last three of these are homologs of the prokaryotic DNA mismatch repair gene *mutL*. The *hMSH2* and *hMLH1* genes account for most classic HNPCC-causing mutations, but germ-line abnormalities in the other genes have been found in HNPCC. This has been better characterized for *hMSH6*, [246] which causes an attenuated form of the disease (see later), than for *hMLH3*, which is possibly associated with HNPCC, or for *hPMS2*, which may require homozygous mutations to create an increased risk for familial cancer. Given this complex situation, a major target for research in this area is the development of clinically practical screening tests for the genetic carrier state of HNPCC.

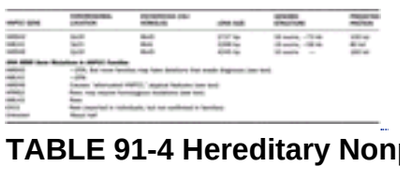


TABLE 91-4 Hereditary Nonpolyposis Colorectal Cancer (HNPCC) Genes

In HNPCC, there is a high risk for cancer but with features distinct from the polyposis syndromes. [247], [248] and [249] This syndrome is inherited as an autosomal dominant disease, and it produces colon cancers two to three decades earlier than in sporadic tumors. The syndrome was historically divided into two variants: [248] Lynch syndrome I, which included the early-onset colorectal cancers, and Lynch syndrome II, also called *cancer family syndrome*, [250] which included early onset of carcinoma at other sites, such as the endometrium, ovaries, upper urinary tract, small intestine, and stomach [251], [252] and [253] (Table 91-5). Identification of the genes responsible for this disease revealed that the variable expression of the tumor phenotype may be seen with mutations in any of the HNPCC genes, which obviates the necessity to subdivide the disease into these variants. Common tumors such as those of the lung, breast, and prostate are not more common in these HNPCC families. [251], [252] The extracolonic tumors are not uniformly distributed, and they tend to cluster in certain families, underscoring the necessity to obtain detailed information on tumor development. [249], [251] Most HN PCC families have some extracolonic cancers, most commonly in the endometrium. [253]

Autosomal dominant inheritance; high penetrance
Early-onset colon cancers (mean age ~40 y); risk begins in the 20s
Absence of multiple adenomatous polyposis
Multiple primary cancers
Multiple synchronous primaries in 18%
Metachronous tumors accrue at 3%–5% per year
Increased proximal tumors (60%–80% proximal to the splenic flexure)
Increased mucinous adenocarcinomas (35%–39%)
Linked to germ-line mutations in DNA mismatch repair genes
Increased number and early occurrence of other adenocarcinomas
Endometrium (20%–40% risk)
Stomach
Ovaries
Small intestine
Renal (pelvis and ureters)
Hepatobiliary system
Other sites (see text)
Muir-Torre syndrome
All the features of hereditary nonpolyposis colorectal cancer (most cases linked to germ-line mutations in hMSH2)
Unique skin tumors: multiple keratoacanthomas and sebaceous neoplasms

TABLE 91-5 Hereditary Nonpolyposis Colorectal Cancer

HNPCC was first recognized in 1895 by a University of Michigan pathologist named Aldred S. Warthin, and the index family was followed up in 1971 by Henry T. Lynch, who obtained data on 650 relatives over six generations. [250] This carefully studied family argued strongly for a genetic rather than an environmental cause of this disease because the progeny of each of the affected members of the initial kindred continued to show this characteristic, whereas the offspring of unaffected individuals did not. HNPCC has been reported from many countries throughout North America and Europe. It has been estimated that HNPCC is one of the most common forms of familial cancer and in some settings may account for up to 5% of cases of colon cancer. [254] Using rigorous criteria for the diagnosis, however, others estimate the incidence to be on the order of 2% to 3% of all colorectal cancer. [255], [256] Given the small family size typical in industrialized countries, it is difficult to establish familiality for common illnesses with variable presentations. A set of guidelines has been developed to assist in the use of molecular diagnostics for HNPCC. [257]

The principal difficulty in making this diagnosis is that the phenotype is not necessarily distinctive, and no definitive premorbid markers have been identified. For this reason, criteria for a working definition of HNPCC for clinical studies, developed by an international collaborative group, have been referred to as the Amsterdam criteria. The diagnosis is assumed if a family has all of the following: three or more relatives have a verified colorectal cancer, with one person being a first-degree relative of the other two; colorectal cancer involves at least two generations, and one or more cancers is diagnosed in family members younger than 50 years of age. Using databases collected in the United States—where the likelihood of colorectal cancer by age 74 years is 0.049 for men and 0.038 for women and 7% of the diagnoses are made in people younger than 50 years of age—the Amsterdam criteria would be met by chance alone in 0.08% of families of eight people, if all have lived to 74 years of age and if colorectal cancer had no familial clustering. [249] Because colorectal cancers are not randomly distributed among families, however, the number of such familial clusters is much higher. It is not necessary to meet all these criteria to make the diagnosis in clinical practice, however, because they are stringent and may exclude small affected families. The original Amsterdam criteria were revised in 1999 to account for the occurrence of noncolonic cancers, and it is clinically appropriate to consider endometrial cancer, and other cancers of the HNPCC spectrum, among the three cancers “required” by the initial Amsterdam criteria. [258]

One study of 19 European families with documented HNPCC gene mutations reported an estimated lifetime risk for co-lorectal cancer of 80% and a differential risk for endometrial cancer, depending on whether the germ-line mutation was in *hMSH2* (61%) or *hMLH1* (42%). Elevated risks for small intestinal cancer were found for both genes, and increased risks for cancers of the urinary tract, stomach, and ovaries were found among those with *hMSH2* mutations. [259] In a separate study using a population-based strategy and 67 known gene carriers whose disease outcome to age 70 years was known, an estimate has been made that the risk for any cancer in men was 91%, whereas in women it was 69%. Colorectal cancer occurred in 74% of men and in 30% of women, but women carried an additional 42% risk for uterine cancer. [260]

The penetrance of the HNPCC gene is high, but the age at presentation varies. Occasionally, patients develop tumors at very young ages (i.e., in their teens) and may have malignant disease before it develops in the parent, creating the erroneous initial impression of a skipped generation. Therefore, in considering this extreme degree of familiality, it is important to take family histories that include grandparents, aunts and uncles, and children.

In the general population, most colorectal cancers occur in the distal colon, and only 23% to 32% occur proximal to the splenic flexure; in HNPCC, 65% to 88% of the tumors occur in the proximal colon. [249] Significantly more mucinous carcinomas are seen in HNPCC than in control cases (35% to 39%). [261] A higher proportion of poorly differentiated tumors has been reported in HNPCC, but this may be a reflection of a large number of tumors with atypical cytologic features that have been overinterpreted. In fact, patients with these tumors are diagnosed at lower Dukes stages and have a better survival rate than controls. [249]

Although diffuse polyposis does not occur in these syndromes, these cancers develop from a discrete, small number of preexisting adenomas, and there is indirect evidence that they develop through an aggressive adenoma-carcinoma sequence in this disease. [262] In a colonoscopic screening program, 30% had polyps, and these were more likely to be multiple, large, and villous compared with those in the general population. [263] The prevailing opinion is that the HNPCC gene may predispose for rapid growth and malignant conversion in benign adenomas. In only 40% of these patients was a positive family history available before the diagnosis of the cancer, which is unfortunate because the level of suspicion of cancer is usually low in young patients. These patients have an extremely high likelihood of developing metachronous colorectal tumors, with mean annual cumulative rates reported to be 3% to 5%. [244] The mean age for developing the first tumor is about 40 years, and cancers occurring in patients in their 20s are not unusual. [239], [248] Because at initial diagnosis 18% have multiple synchronous tumors, it is inappropriate to treat this syndrome with a limited segmental colonic resection.

Muir-Torre syndrome is a rare variant of HNPCC characterized by multiple sebaceous gland neoplasms, including sebaceous adenomas, sebaceous carcinomas, and keratoacanthomas, in addition to all the features of HNPCC. MSI has been found in the skin and colorectal tumors in this disease, and nearly all of the families with this syndrome have been linked to mutations in the *hMSH2* DNA mismatch repair gene. [264], [265] The key clinical point is that a patient with a history of multiple sebaceous neoplasms (or keratoacanthomas) must raise the suspicion of HNPCC and the attendant risks for colorectal and other cancers.

Perhaps the most unique aspect of HNPCC is that it represents a separate pathway of tumor development in which the loss of DNA mismatch repair activity results in a cascade of inactivation of other tumor suppressor genes, including one of the tumor growth factor-ß receptors (TGF-ß1 RII), the apoptosis gene *BAX*, the insulin-like growth factor-2 receptor, and two of the DNA mismatch repair genes, *hMSH3* and *hMSH6*. [266], [267] and [268]

The diagnosis of HNPCC is first suspected on clinical grounds, but the management is sufficiently demanding for affected patients that it is clinically important to be more certain about the diagnosis. The first generation of diagnostic tests for HNPCC made use of the truncated protein test for the *hMSH2* and *hMLH1* genes, which is about 50% sensitive for families that met the Amsterdam criteria. [269] However, it is now possible to make the diagnosis by direct DNA sequencing from lymphocyte DNA, and databases are available, which can help distinguish common polymorphisms from disease-causing mutations. [270] An Internet database may be consulted for this purpose at the URL address: www.nfdht.nl/database/mdbchoice.htm. If a DNA sequence variation is found in a patient, one can determine whether this has

previously been seen in an HNPCC family or whether this sequence occurs at some frequency in the population without increasing the risk for cancer.

Germ-line mutations in *hMSH2* and *hMLH1*, which might be called the “major” DNA mismatch repair genes, account for most of diagnosed HNPCC; however, genetic testing is conclusive in fewer than 50% of cases, depending on how the families are ascertained and what genetic testing is used. [271] One explanation for this is that some of the mutations in the *hMSH2* genes are large genomic deletions that are not detected by either direct sequencing strategies or strategies based on the truncated protein test. [272]

Germ-line mutations in the “minor” DNA mismatch repair gene *hMSH6* accounts for a smaller proportion of the disease, where it produces an attenuated form of the disease. Mutations in this gene have been found in 8% of familial colon cancer families in which the phenotypes *did not* meet the Amsterdam criteria for HNPCC. The colon cancers occur somewhat later in life than in the *hMSH2* or *hMLH1* forms of the syndrome (at a median age of 61 years), the penetrance is reduced (i.e., not everyone with the mutation necessarily develops a cancer), and these families may have more endometrial cancer. [273], [274]

It is not entirely clear to what degree germ-line mutations in certain other minor DNA mismatch repair genes, that is, *hMLH3*, *hPMS2* and *hPMS1*, cause HNPCC, despite the fact that sequence variations have been found in some patients suspected of having HNPCC. After the initial reports that HNPCC could be linked to germ-line mutations in *hPMS1* and *hPMS2*, it was subsequently determined that there are no well-documented HNPCC families associated with *hPMS1* germ-line mutations and probably none with *hPMS2*. However, in the case of *hPMS2*, it is has been speculated that compound bi-allelic germ-line mutations at the *hPMS2* locus may rarely be associated with Turcot syndrome. [275] Likewise, germ-line sequence variations in the *hMLH3* gene have been found in a small number of patients with colon cancer, but large families with this have not been reported, and the most of the sequence variations are missense mutations, which are not necessarily disease causing. Until convincing, large families are found that confirm the inheritance of a colon cancer predisposition with these germ-line sequence variations, any conclusions remain subject to alternative interpretations. [276]

The diagnosis of familial colon cancer is a complex area. An Internet-based information site has been assembled and maintained by GeneTest at the Web site http://www.genetest.org/. Additional assistance can be obtained in interpreting mutations in a review of genetic testing for hereditary colon cancer. [277] Not all patients who are young with colon cancer or with modest degrees of familial involvement are candidates for undergoing genetic testing. One means of determining which patients should be tested for HNPCC is to perform microsatellite analysis on the excised tumor tissue, which can be done on an archival specimen embedded in paraffin. Tumors showing MSI suggest that one is dealing with HNPCC, and the absence of this makes the diagnosis quite unlikely. The guidelines for interpreting microsatellite analyses can be found in a workshop consensus statement published by the National Cancer Institute. [278]

Nonsyndromic Familiality for Colorectal Cancer

The risks conferred by a family history of colorectal cancer have been estimated in many studies by obtaining family medical data from patients who have developed a colorectal neoplasm; however, patients usually present with the family history and ask for an estimate of their risk for disease. [279] Nonetheless, the data provide reasonable guidelines for clinical decision making. This is not a trivial issue because about 10% of patients report a positive family history for colorectal cancer, and the increment of added risk is not uniform from patient to patient.

A prospective study of nearly 120,000 U.S. health care professionals led to the diagnosis of colorectal cancer in 463 individuals. The age-adjusted relative risk for colorectal cancer in patients with a first-degree relative with the disease was 1.72 (95% CI, 1.34 to 2.19). When there was more than one first-degree relative with colon cancer, the risk rose to 2.75 (95% CI, 1.34 to 5.63). If the affected individual was aged 30 to 45 years, the relative risk was 5.37 (95% CI, 1.98 to 14.6), and the risk to the family decreased significantly with increasing age of the proband. [280] Interestingly, the relative risk was close to 1.00 when the cancer occurred in a person older than 60 years of age. The implications are clearly that the general nonsyndromic familiality appears to be similar to the principles predicted from an understanding of FAP and HNPCC, in which the familial risk is associated with earlier age of disease. The genetic factors may represent milder variations of the syndromic diseases or other genetic diseases yet to be found. The key clinical issue is that when familiality is identified, the suspicion of disease and screening interventions must begin earlier than recommended for sporadic disease. Additional support for the observation that familial risk is proportional to the number of affected relatives came from a smaller Australian case-control study that found an odds ratio for colorectal cancer of 1.8 for one affected first-degree relative and 5.7 for two. [281]

The National Polyp Study, which enrolled nearly 1200 patients for familial analysis, found that the relative risk for colorectal cancer was 1.78 (95% CI, 1.18 to 2.67) for the parents and siblings of patients with adenomatous polyps. The relative risk rose to 2.59 (95% CI, 1.46 to 4.58) when the polyp-bearing patient was younger than 60 years old, and the risk increased significantly with an earlier age for diagnosis of the adenoma. Furthermore, the risk increased to 3.25 (95% CI, 1.92 to 5.52) for siblings of a polyp-bearing patient of whom a parent also had colorectal cancer. [282] This study underscores the fact that adenomatous polyps are important issues in the family history and that the age at onset and multiplicity of family member involvement should be taken into account while taking family histories.

A study from Utah screened a large number of individuals, their spouses, and first-degree relatives using a 60-cm flexible proctosigmoidoscope. This study demonstrated a significant impact of family history on the prevalence of distal colorectal neoplasia. [283] The investigators used pedigree analysis to identify whether any form of mendelian genetics would describe the inheritance seen. Their data supported dominant inheritance of a susceptibility to colorectal neoplasms (mostly adenomatous polyps) in the distal colon with a gene frequency of 19% in the general population. The expression of the phenotype (i.e., penetrance) among gene carriers increased with age, reaching 63% by age 80 years. Conversely, the penetrance for the nonsusceptible genotype was estimated to be zero (i.e., nonsusceptible families would develop no polyps).

This genetic model implies that a sizable segment of the population (19%) is susceptible to distal colonic neoplasia on a familial basis and that a large proportion of these people (i.e., 63%) will develop benign neoplasms by 80 years of age. Because about 5% to 6% of the population as a whole develop colon cancer, it could be further speculated that additional factors, presently unknown but possibly environmental, promote the development of malignant from benign neoplasms.

A very large twin study in Scandinavia indicated that 35% of colon cancer cases arose from inherited susceptibility, whereas 8% occurred on the basis of shared environmental factors in families. [284] The authors concluded that additional susceptibility genes for colon cancer should be sought. A number of such genes have been implicated and together with others are now under study as to their importance to the pathogenesis of familial colon cancer. [285]

Prior Polyps and Cancers Autopsy studies provide estimates that about 30% to 40% of colons contain adenomatous polyps, most of which are less than 1 cm in diameter. [286], [287] and [288] Patients with adenomatous polyps constitute the principal group identified as being at elevated risk for the development of cancer. Patients with adenomas have a 50% risk for developing a metachronous adenoma after 15 years of observation, with a risk of 1 in 12 (for men) or 1 in 20 (for women) for developing a cancer in that time frame. [289] Thus, individuals who develop one adenoma are at risk for developing more over time. Although the prevalence of adenomas is high, not all adenomas carry the same risk for recurrence to their host. A pair of retrospective studies reported that patients with small adenomas removed (i.e., those smaller than 1 cm) had a diminished risk for recurrent neoplasia, and their survival rate was not reduced compared with general risk estimates established for this community. [290], [291] On the other hand, those with adenomas larger than 1 cm had an increased risk for cancer of 2.7 times that estimated for the general population. [291] The relative risk for rectal cancer was found to be 0.6 for patients who had small (less than 1 cm) rectal adenomas removed with an average follow-up period of almost 14 years. In contrast, the removal of large adenomas was associated with a relative risk for rectal cancer 2.1- and 2.6-fold greater for lesions 1 to 2 cm in diameter and lesions larger than 2 cm, respectively. [292] Excess risk for rectal cancer was 3.8-fold after removal of villous adenomas and 5.1-fold after removal of adenomas containing severe dysplasia (i.e., carcinoma in situ). No increased risk followed the removal of multiple lesions. Risk for colon cancer (above the reach of the rigid sigmoidoscope) was increased 3.3-, 5-, and 5.9-fold after the removal of severely dysplastic, villous, and large (more than 2 cm) lesions, respectively, and a 4.8-fold increase was seen after the removal of two or more rectal adenomas. [292] Therefore, risk estimation depends on the size, number, and histology of the index lesions. Single, small (less than 1 cm) adenomas probably do not predict a significantly increased risk for recurrent neoplasia. It is more difficult to estimate the actual incidence of metachronous cancers from the literature, and the results are influenced by the amount of colon removed during the first operation and the duration of follow-up. The best estimate is that about 5% of patients will develop a second cancer after the removal of a primary colorectal cancer. [293] Because the average age to develop colorectal cancer is about 70 years, the risk for metachronous tumor development is likely to be higher in patients who are younger at the time of the first tumor. It may not be possible to refine this estimate because of the practice of removing all adenomas during surveillance colonoscopy of colon cancer patients.

Inflammatory Bowel Disease Patients with chronic inflammatory bowel disease are at increased risk for gastrointestinal cancer. The degree of risk, however, has been a subject of debate. A British study of 624 patients at a referral center indicated that 3.5% of their patients developed colorectal cancer, about seven times as many as expected in the general population. [294] The diagnosis of cancer in this condition was made at an average age of 41 years (range, 20 to 74 years). Relatively

fewer of these cancers occurred in the rectum (22%) compared with the expected rate in the general population (38%). Thus, there are important differences in clinical presentation in the setting of inflammatory bowel disease, the most important being age. The duration of inflammatory bowel disease is a critical factor in predicting the likelihood of developing adenocarcinoma of the colon. A retrospective study of 267 referral patients with ulcerative colitis revealed a 10% incidence of colon cancer. Cancer was more likely to occur among patients with pancolitis (13%) than among those with only left-sided disease (5%), and the latter group tended to develop cancer a decade later than the former. The incidence of colorectal cancer was less than 1% during the first decade of disease but progressively rose to 7% in the second decade, 16% in the third decade, and 53% in the fourth. [295] The high incidence of cancer complicating ulcerative colitis may be a reflection of bias in patient selection from referral centers. A review of the clinical courses of 258 patients treated for ulcerative colitis in a private practice revealed a cancer incidence of 7% after 26 years and 11% after 32 years of disease. [296] Most of these patients had less than universal colitis, and patients who were referred with known colon cancer were excluded from the analysis. Thus, the inclusion of a relatively small number of patients referred because of cancer could produce a large impact in the apparent risk for this disease. Geographic factors also may be important in influencing the risk for neoplasia in the setting of colitis. The incidence of cancer among children referred to a large referral center in the United States [297] was similar to that reported in Great Britain. [294] Cancer developed in 3% of the patients during the first 10 years after the onset of colitis, but the incidence increased by 20% in each of the next two decades and was estimated to be 43% 35 years after the onset of disease. In contrast, a center receiving referrals throughout Czechoslovakia reported a cumulative cancer risk of zero at 10 years, 5% at 20 years, 15% at 30 years, and 20% at 35 years, [298] even though the incidence of colorectal cancer is quite high in that country. [11] Although this was a large study consisting of 959 patients, only 32% of the patients had colitis of the entire colon, and most patients in this group (60%) had follow-up of less than 10 years. Thus, it is not clear whether the differences reported in the literature reflect geographic differences in the incidence of this complication or whether all of the increased risk is the result of greater extent and duration of disease. Patients with Crohn's disease are also at increased risk for colorectal cancer; however, the incidence is lower than that reported with ulcerative colitis. [299] It is not yet possible to estimate accurately the risk for cancer in this setting; however, an excess of carcinomas of the colon, small intestine, stomach, and anus, as well as an excess of lymphomas, has been reported. [299], [300] and [301] The survival rate of patients who develop cancers in the setting of ulcerative colitis is similar to that seen for noncolitic patients, [302] despite routine surveillance for cancer. However, a complicating carcinoma is unlikely to produce unique symptoms early in its natural history, and these tumors may be difficult to diagnose even if viewed colonoscopically in the setting of inflammation and mucosal distortion. In addition, the natural biology of cancers in ulcerative colitis may be different from that of sporadic ones. To improve the early detection and survival from colon cancer in colitic patients, attempts have been made to identify early neoplastic lesions. Dysplasia is currently the best marker for early cancer in this setting. Developing a standardized classification for dysplasia has been a major undertaking because inflammation and attendant repair can be easily confused for early neoplasia. [303] True dysplasia is an early benign neoplastic lesion, and biologically, it is analogous to adenomatous tissue in colonic polyps. Just as it is difficult to predict the clinical behavior of a small adenoma, low-grade dysplasia does not necessarily indicate the need for colectomy, and the predictive value for all grades of neoplasia has been controversial. [304] Low-grade dysplasia in a biopsy may reflect inflammation and be a transient change, it may reflect the presence of a higher-grade lesion (such as cancer) immediately adjacent to the biopsy site, or it may be a marker of a generalized problem in the colon, with additional neoplastic lesions elsewhere. Dysplasia in a plaque or elevated mass is especially ominous, and some have advocated total colectomy for this. [305] If low grades of dysplasia are found in random biopsies of flat mucosa, this is less likely to be associated with a synchronous cancer, and continued surveillance is the best option. High-grade dysplasia is a substantially more worrisome finding; if it is found in a colonic mass, total colectomy should be given serious consideration. After examination of the surgical specimens, most of these colons are found to have either confirmed high-grade dysplasia (which is the equivalent of carcinoma in situ) or a frank, invasive cancer.

Colon Cancer in Patients with Nonadenomatous Polyposis Syndromes

Juvenile Polyposis Syndromes and Peutz-Jeghers Syndrome Inherited syndromes that exhibit juvenile polyposis as a major phenotypic characteristic include the juvenile polyposis syndrome, Cowden syndrome, and Bannayan-Riley-Ruvalcaba syndrome. [306] They are all rare autosomal dominantly inherited conditions that give important clues to colon cancer pathogenesis. See Chapter 90 for a complete discussion of each of these. Juvenile polyposis syndrome gives rise to multiple juvenile polyps that exhibit benign complications in patients younger than 30 years of age but exhibit a high risk for adenomatous change in polyps and even cancer after that age. Cowden syndrome exhibits juvenile polyps and carries little colon cancer risk but a high risk for breast cancer and other hamartomatous growths. Bannayan-Riley-Ruvalcaba syndrome is similar to Cowden syndrome and adds the phenotypic manifestations of macrocephaly, delayed psychomotor development, and pigmentation of the glans penis to those of Cowden syndrome. Possibly explaining the phenotypic and cancer differences of these juvenile polyposis conditions is the recent elucidation of their genetic etiologies. Juvenile polyposis arises from germ-line mutations of the *SMAD4* [307], [308] and *BMPRA1A* tumor suppressor genes, [309], [310] both involved in the TGF-ß signaling pathway, whereas Cowden and Bannayan-Riley-Ruvalcaba syndromes are allelic and arise from germ-line mutations of the *PTEN* tumor suppressor gene. [311], [312] Peutz-Jeghers syndrome is characterized by perioral melanin pigment spots and histologically characteristic gastrointestinal polyps that are most common in the small bowel. [306] Similar to the juvenile polyp syndromes, benign complications of polyps predominate in the first three decades of life, whereas both gastrointestinal and extraintestinal malignancies become extremely common thereafter. [313] Peutz-Jeghers syndrome arises from germline mutations of the *STK11* gene, which appears to be a tumor suppressor gene because both alleles are found to be inactivated in neoplastic tissues. [314], [315]

Other Clinical Associations

Dietary History As mentioned in the initial section on epidemiology, more cancers would be expected in patients who ingest a high-fat, low-fiber, meat-rich diet, and obese, hypercholesterolemic patients would seem to be a target group for the disease. Nonetheless, the diet is relatively uniform within high-risk populations, and the individual variations in fat intake are too minor compared with the general characteristics of the diet to make dietary history valuable for selective screening. Other features have therefore been sought to identify high-risk patients, generally without success.

Colon Cancer and Cholecystectomy It has been suggested that patients who have undergone cholecystectomy may be at a greater risk for colon cancer, but the data on this issue have been inconsistent. Cholecystectomy has been implicated as a cause for an excess of cancers among women, [316] for an increased incidence of proximal colon cancers, [317] and for an increased incidence of adenomas. [318] It has been difficult to confirm the association between cholecystectomy and adenomatous polyps, [319] and other groups have suggested that the association with colon cancer is weak or absent. [320], [321] Several recent studies and one metaanalysis have demonstrated no excess in the risk for either adenomas or carcinomas after cholecystectomy. It is reasonable to disregard neoplastic risk when making decisions regarding gallbladder surgery, and there is no need for extra surveillance in patients who have had this operation. [322], [323] and [324]

Endocrine Abnormalities Acromegalic patients have a threefold excess of colon cancers. [325] The role of growth hormone has not been explored. Elevated serum gastrin levels are seen in patients with colorectal cancer, [326] and a postoperative reduction has been seen after surgery. [327] There is evidence that gastrin undergoes alternate processing in normal and neoplastic colon [328] and may function as an autocrine hormone to support the growth of tumor cells. [329] The total number of patients with these disorders is too small to permit an evidence-based recommendation for screening.

Skin Tags The presence of acrochordons (skin tags) suggests the simultaneous presence of colonic polyps. In a study of 94 men referred for colonoscopy, 48 had skin tags, usually located in the axilla, upper chest, or neck, and 46 did not. Among the group with skin tags, 77% had colonic polyps; in the subgroup without skin tags, 20% had colonic polyps. [330] In an attempt to confirm this finding in an unselected group of 492 men in a primary care clinic, skin tags were found in 46% of patients. Flexible sigmoidoscopy revealed polyps in 10% of individuals with skin tags, but 8% of those without skin tags also had polyps. [331] A review of the collected studies of this issue concluded that skin tags have prognostic significance only in symptomatic patients; therefore, this finding is of little practical significance. [332] Skin tags also do not correlate with carriage of the gene for FAP. [333]

Colon Cancer and *Streptococcus bovis* Patients with carcinoma of the colon occasionally present with septicemia or endocarditis caused by *S bovis,* [334], [335] which may be cultured from the feces of 10% to 16% of healthy subjects, but it is significantly more prevalent in the stools of cancer patients. In a study of 29 patients with *S bovis* septicemia, 16 had gastrointestinal neoplasms, and nearly half of the group did not undergo a complete diagnostic evaluation. [335] *S bovis* endocarditis also has been reported as the presenting symptom in one patient with FAP. [336] The presence of *S bovis* in the blood should prompt a complete gastrointestinal evaluation, beginning with the colon, and if this evaluation is negative, an evaluation of the esophagus and the stomach should be done. Septicemia produced by other members of the group D streptococcus family (*Streptococcus equinus*) also has been associated with colon cancer. [337]

Breath Methane Several laboratories reported that colon cancer patients are more likely to be breath methane excreters than the general population, presumably reflecting a difference in the anaerobic flora. [338] The elevated methane production falls to normal after the cancer is resected, suggesting that the presence of the tumor influences methane production and that increased methane production may be a result of colon cancer rather than a reflection of a fixed abnormality in the colonic flora. [339] Increased methane production also was reported in patients with familial polyposis and extensive ulcerative colitis in this study. Because there is a strong correlation between the methane excreter status of an individual and the status of other family members, these observations potentially have broad implications; however, breath methane measurements in more than 1000 people from South Africa from four population groups with wide differences in colon cancer risk demonstrated marked interethnic differences but no relation to cancer risk. [340] Furthermore, methane excretion is strongly affected by the use of laxatives or antibiotics and must be interpreted with caution.

Other Features Additional risk factors for colorectal cancers have been identified, but these generally are derived from exceptional circumstances. As previously mentioned, women who have been irradiated for gynecologic cancer have a 2- to 3.6-fold increased risk for colorectal cancer. [2C] Populations infected with

Schistosoma hematobium may develop colon cancers in the immediate vicinity of the polyps produced by the parasite eggs. [341] Patients who have undergone an implantation of the ureter into the sigmoid colon are at risk for carcinomas, adenomas, or severe dysplasia in the vicinity of the ureterosigmoidostomy. [342] In a study of 34 such patients, 29% had developed one of these neoplastic lesions at a mean interval of 22 years after their urinary diversion. Bloom syndrome is an autosomal recessive form of congenital dwarfism associated with unusual facies and hypersensitivity to sunlight. [343] This syndrome has been anecdotally associated with colorectal cancer; however, its rarity makes it difficult to document the actual incidence. The Bloom syndrome gene, *BLM,* is a helicase and is required for the orderly segregation of chromosomes during mitosis. Its inactivation leads to a type of genomic instability and predisposes to cancer. [343] Of interest, at least one individual with Bloom syndrome has been reported with multiple colonic polyps. [344] This genetic disorder is exceedingly rare, but one should keep in mind the possibility of a predisposition to colorectal neoplasia in this setting. It has been suggested that patients with Barrett esophagus may be at increased risk for colon cancer, [345], [346] and one small, prospective, controlled, colonoscopic study of 36 patients with Barrett esophagus revealed three cancers and nine adenomas; 2 of these patients had severe dysplasia. The authors suggested that such patients might benefit more from colonoscopic surveillance than screening of the esophagus. [347] This concept has not been confirmed in other studies. Patients with irritable bowel syndrome and diverticulosis [348] have no increased risk for colorectal cancer. Patients with these disorders have symptoms attracting attention to their lower gastrointestinal tracts; however, there is no need for extra surveillance for cancer among these patients.

Pathology

Most colorectal cancers are adenocarcinomas, and most cancers represent malignant conversion occurring in a preexisting adenomatous lesion. As a result, a spectrum of lesions may be found, ranging from the small adenomatous neoplasm containing no more than low-grade dysplasia and no immediate ability to invade or metastasize at one end of the spectrum, to the poorly differentiated adenocarcinoma with an unlimited capacity for local and distant spread at the other.

Gross Pathology

Colon cancers develop within preexisting foci of adenomatous tissue. This occurs usually, but not always, in a polypoid lesion. Adenomatous change may occur in flat mucosa, and a cancer may develop in this setting. Therefore, tiny cancers in flat mucosa are reported, but not commonly. [349], [350] and [351] These cases represent instances in which malignant conversion occurred early in the natural history of the neoplastic lesion, and they should be considered the exception to the rule.

Colon cancers characteristically begin as round mass lesions, but deviations from the ideal shape occur as a result of the asymmetric sloughing of cells and the emergence of clones with rapid growth capacity. If the resected colon is opened, a cancer typically has an elevated advancing edge, and the lumenal aspect usually is ulcerated and irregular. After the diameter of a cancer approaches the circumference of the colon, the opposite edges of the tumor converge, creating the characteristic apple-core lesion described by radiologists. This most often occurs in the sigmoid colon, which has the smallest circumference.

The older literature states that 75% of cancers are present in the rectum and sigmoid colon, making them accessible to detection by sigmoidoscopy. During the past several decades, a larger percentage of lesions have been found in the proximal colon. [352], [353], [354], [355], [356] and [357] This appears to be related, in part, to the fact that proximal colonic cancers are more common among people older than 65 years of age; as the population ages, a greater number of proximal colon cancers would be expected. Some have reported a decrease in rectal carcinomas, with corresponding increases in sigmoid cancers and proximal lesions, whereas others have observed an absolute increase in proximal cancers. [357] Even after correcting for age, however, investigators from several countries have reported a shift from a predominance of rectal tumors to those located more proximally, [354], [355] and [356] and in some registries, the decrease in the incidence of rectal cancer has been particularly dramatic. [357]

It has been estimated that about one fourth of cancers may be detected with a rigid sigmoidoscope, and this may be increased to about two thirds by the use of the 65-cm flexible sigmoidoscope, depending on the depth of the examination (Fig. 91-3). A substantial proportion of lesions (at least one third) is missed with limited examinations, and about one fourth of colorectal cancers are in the cecum and ascending colon, which is the most challenging part of the colon for imaging and visualization. [358] The changing location of colon cancers has obvious implications for clinical management. Although it has not been proved, environmental factors may be increasing the risk for colorectal cancer and that screening measures and polypectomy—which are more effective for the distal colon—have simultaneously decreased cancer incidence in the sigmoid colon and rectum.

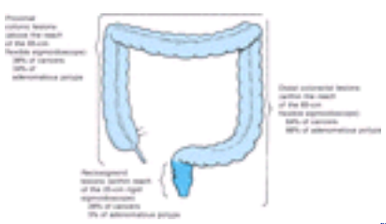


FIGURE 91-3. Distribution of neoplasms in the colon and rectum.

It is not rare to find multiple primary colorectal cancers. Multiple lesions generally are divided into those that occur simultaneously (synchronous lesions) and those that occur in different time frames (metachronous lesions). Synchronous colorectal cancers occur in 3% to 6% of de novo colon cancer diagnoses. [359], [360] and [361] The lesions may be either near one another or located in different portions of the colon. The incidence of multiple synchronous lesions is significantly higher in the setting of FAP or ulcerative colitis, in which it occurs in 21% and 18% of patients, respectively. [362] Synchronous adenomatous polyps are found in 36% of patients. [361] Multiple primary neoplastic lesions occur often enough that total colonoscopy is an essential part of the workup if a neoplastic lesion is found at a more limited examination. This permits the removal of synchronous polyps and the detection of synchronous cancers, which will modify the surgical approach.

Microscopic Pathology and Tumor Staging

Although most colorectal cancers are adenocarcinomas (Fig. 91-4), squamous cell carcinomas, adenosquamous carcinomas, lymphomas, and endocrine tumors such as carcinoids also occur in the colon. Most colonic adenocarcinomas are moderately or well-differentiated tumors, and there are few morphologic features of prognostic significance among them. About 20% of adenocarcinomas are poorly differentiated or undifferentiated tumors, and these two types are well known to be associated with a poorer outcome. Most adenocarcinomas secrete a small or moderate amount of mucin. About 10% to 20% of tumors may be described as mucinous or colloid carcinomas on the basis of a more prodigious production of mucin. These tumors are associated with a poorer 5-year survival rate than nonmucinous tumors. [363], [364]

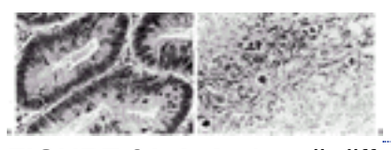


FIGURE 91-4. A: A well-differentiated adenocarcinoma of the colon demonstrates the characteristic glandlike formation. In each of these glands, the nuclei are large, the chromatin is poorly condensed, and the nuclei tend to drift away from the basal lamina. Mitoses are common. **B:** In a mucin-producing or colloid carcinoma, the cancer cells (*dark nuclei*) are suspended in pools of mucin, which is unstained in this section. The typical glandlike configuration is not seen. This microscopic appearance is associated with a more aggressive tumor.

The classification of tumor invasion was first undertaken by Dukes for rectal carcinoma. [365] The Dukes staging has undergone many modifications, and the stages

must be defined precisely for purposes of discussion because there is no standard classification. A system that is perhaps a consensus of most common use is as follows (Fig. 91-5):

Carcinoma in situ (also called high-grade dysplasia) is intramucosal carcinoma that does not penetrate the muscularis mucosae.
Stage A tumors invade through the muscularis mucosae into the submucosa but do not penetrate the next layer, the muscularis propria. (Note: Dukes was inconsistent here, as are other systems that include invasion of the muscularis propria in stage A, as illustrated in the figure.)
Stage B1 tumors invade into the muscularis propria, and B2 lesions completely penetrate the smooth muscle layer to the serosa but go no further. Some use B3 to describe a lesion that invades an adjacent organ.
Stage C lesions encompass any degree of apparent invasion but are defined by regional lymph node involvement. Some studies subdivide Stage C lesions based on the number of lymph nodes involved, with C1 lesions having one to three (or four) involved, and C2 having more positive nodes. The presence and number of involved lymph nodes, however, depend entirely on the number of nodes resected and examined pathologically, which may explain some of the inconsistencies in the literature.
Stage D lesions include all those with distant metastases.

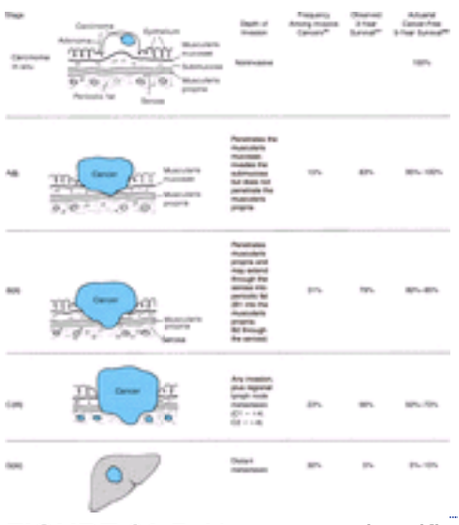
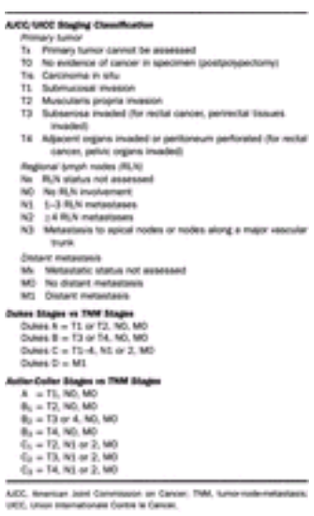


FIGURE 91-5. Numerous classifications have been proposed for colorectal cancer. Demonstrated here is the Dukes-Turnbull classification A through D (accompanied by the TNM stage in parentheses), but it is not the only classification currently in use. Of potential importance are subclassifications in classes B and C. Subclass B1 has been proposed to include invasion to the muscularis propria but not through the serosa. Subclass B2 includes tumors that penetrate the serosa to the pericolic fat but do not have regional lymph node or distant metastases. Patients with B1 lesions have a better prognosis and 5-year survival rate than patients with subtype B2 lesions. Class C lesions have been subdivided into C1, with four or fewer involved regional lymph node metastases, and C2, with more than four involved regional lymph nodes. Patients with subtype C1 lesions have a better outcome than patients with C2 lesions. The observed 3-year survival and actuarial cancer-free 5-year survivals are estimated for each stage. The actuarial survival estimates reflect excessive disease-related mortality from the cancer, and the crude survival rates are considerably lower because of co-morbidity.

The relation between advancing stage and cancer mortality has been repeatedly demonstrated. [365], [366], [367] and [368] There is no important relation between the tumor size in the colonic lumen and clinical outcome. The aggressiveness of a colorectal tumor is reflected by its ability to invade, and growth into the colonic lumen is irrelevant to this biologic characteristic, except to the degree that it reflects the increasing probability that an invasive clone has had the opportunity to arise in a larger tumor.

In an attempt to create more uniform pathological categories for clinical studies, the American Joint Commission on Cancer and the Union Internationale Contre le Cancer have classified many tumors by a tumor-node-metastasis (TNM) system (Table 91-6). This classification is outlined as follows:

Tis refers to carcinoma in situ.
T1 indicates submucosal invasion (i.e., Dukes stage A), T2 indicates invasion of the muscularis propria (Dukes B1), T3 indicates invasion through the muscularis propria into the subserosa or perirectal tissues (Dukes B2), and T4 indicates invasion into adjacent organs or tissues (B3).
N0 indicates no involved lymph nodes; N1, one to three regional lymph node metastases (as in Dukes C1); N2, more than three regional lymph node metastases (Dukes C2); and N3, a nodal metastasis along the course of a major blood vessel.
Metastatic status is divided into M0 (no distant metastasis) and M1 (metastasis present). [369], [370]



AJCC/UICC Staging Classification

Primary tumor
- Tx Primary tumor cannot be assessed
- T0 No evidence of cancer in specimen (postpolypectomy)
- Tis Carcinoma in situ
- T1 Submucosal invasion
- T2 Muscularis propria invasion
- T3 Subserosa invaded (for rectal cancer, perirectal tissues invaded)
- T4 Adjacent organs invaded or peritoneum perforated (for rectal cancer, pelvic organs invaded)

Regional lymph nodes (R/LN)
- Nx R/LN status not assessed
- N0 No R/LN involvement
- N1 1–3 R/LN metastases
- N2 ≥4 R/LN metastases
- N3 Metastasis to apical nodes or nodes along a major vascular trunk

Distant metastasis
- Mx Metastatic status not assessed
- M0 No distant metastasis
- M1 Distant metastasis

Dukes Stages vs TNM Stages
- Dukes A = T1 or T2, N0, M0
- Dukes B = T3 or T4, N0, M0
- Dukes C = T1–4, N1 or 2, M0
- Dukes D = M1

Astler-Coller Stages vs TNM Stages
- A = T1, N0, M0
- B_1 = T2, N0, M0
- B_2 = T3 or 4, N0, M0
- B_3 = T4, N0, M0
- C_1 = T2, N1 or 2, M0
- C_2 = T3, N1 or 2, M0
- C_3 = T4, N1 or 2, M0

AJCC, American Joint Commission on Cancer; TNM, tumor-node-metastasis; UICC, Union Internationale Contre le Cancer.

TABLE 91-6 Colorectal Cancer Pathological Classifications

Figure 91-6 provides estimated 10-year survival rates for colon cancers according to the TNM staging system. [371]

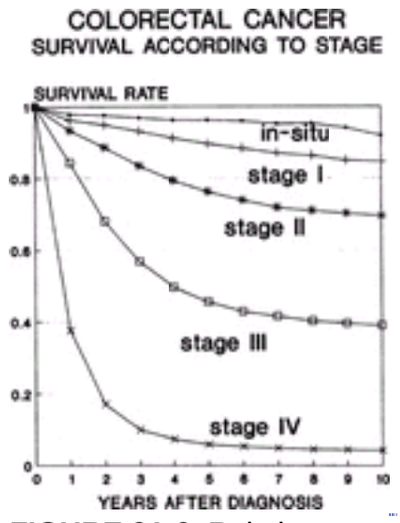


FIGURE 91-6. Relative rates of survival for patients with colon cancer, by stage of disease from a database of more than 110,000 patients using the tumor node metastasis (*TNM*) staging system. (*n* = 4841 stage 0; *n* = 19,623 stage I; *n* = 33,798 stage II; *n* = 29,615 stage III; *n* = 23,233 stage IV.) (From ref. [371].)

Additional microscopic features of rectal cancers defined by Jass and colleagues have been found to correlate with survival. This classification considers the observation that the following three factors correlated best with more aggressive tumors and reduced 5-year survival: spread through the bowel wall and lymph node metastasis (as noted by all other systems); absence of lymphocytic infiltration; and an assessment of whether the invasive margin of the tumor is infiltrating (i.e., more aggressive) or expanding (i.e., less aggressive). Numeric scores were assigned to each of these features, and a high degree of correlation with outcome was found by linear regression analysis. [372]

Several groups have used flow cytometry to measure the relative DNA content, or ploidy, of colorectal cancers. [373], [374], [375] and [376] Tumors with normal DNA content are diploid, and those containing cells with abnormal DNA content are aneuploid. There has been consensus agreement that diploid and near-diploid tumors have significantly better prognoses than aneuploid tumors and that this parameter is prognostic independent of Dukes or other stage (up to the point of distant metastases, after which all patients do poorly). The use of flow cytometry is not indicated in routine clinical practice because no prospective study has demonstrated that it can be used to direct therapeutic decision making.

Several investigators from Japan have recognized that some patients develop early cancers of the colon that appear to arise de novo in that they are lesions smaller than 1 cm and there is no evidence that they arose in a preexisting adenoma. [377], [378] It is not clear how commonly carcinoma arises directly from normal colonic tissue without an early adenomatous stage or whether this variant represents the early conversion of a microadenoma to carcinoma that fortuitously was detected as a small lesion.

One pathological variant bears mention because of the possibility for misinterpretation. Colitis cystica profunda represents the displacement of mucus-secreting cells beneath the normal epithelium, usually as a result of surgery, other trauma, or inflammation. This confers an abnormal, masslike configuration to the overlying mucosa. A biopsy reveals pools of mucus in the submucosa, and these pools may be mistaken for invasive colloid carcinoma. If this condition is seen in the head of an adenomatous polyp, it is referred to as *pseudoinvasion*. [379] This lesion also may occur at the site of a prior surgical anastomosis, and it can be difficult to identify correctly. In general, anastomotic recurrences are relatively uncommon, except at the site of a resection in the rectum or at the rectosigmoid junction, where the surgeon may have taken an inadequate margin of mucosa adjacent to the tumor in an attempt to preserve the rectum. Furthermore, recurrences are likely to be histologically similar to the initial tumor. Therefore, the pathologist should assess carefully the cytologic characteristics of the cells in the biopsy if this entity is being considered.

The colonic epithelium immediately adjacent to a colon cancer (transitional mucosa) is thicker than normal, is characterized by a distortion of the normal architecture, and shows an increase in sialomucin compared with normal tissues, in which sulfomucins normally predominate. [380], [381] Initially, it was suggested that this mucosa was a field of premalignant tissue from which tumors arise. Similar morphologic and histochemical changes are found adjacent to nonneoplastic colonic lesions [382] and lesions metastatic to the colon, however. The accumulated data suggest that transitional mucosa is hyperplastic epithelium that has developed in response to an adjacent tumor or other pathologic condition. [383] The presence of sialomucins in the mucosa at the margin of surgical resection predicts an anastomotic recurrence and is an adverse prognostic variable. [384]

Differential Diagnosis

Symptomatic colon cancers may present in several ways: as a partial or complete lower gastrointestinal obstruction; as overt gastrointestinal bleeding (i.e., hematochezia); as a locally invasive or expanding tumor with invasion of the perirectal tissue, bladder, or other pelvic organs; as a fistulous connection with another portion of the gastrointestinal tract; as a locally expanding metastatic lesion in the liver, bone, or other site; and, less commonly, as a systemic wasting disease (i.e., cancer cachexia).

All these symptoms carry a substantial list of possibilities in the differential diagnosis. If a patient presents with gastrointestinal obstruction, the first step is to determine the level of the obstruction, most typically with supine and upright abdominal radiographs. A tumor in the cecum may obstruct the ileocecal valve and produce a selective small bowel obstruction. Alternatively, an obstructing tumor may occur at any point throughout the colon. Diseases that most commonly produce this clinical picture are tumors, diverticulitis, inflammatory masses (either in the setting of inflammatory bowel disease or an acute infectious process), postinflammatory or ischemic strictures, and volvulus. A more complete differential diagnosis of lesions that may present as a colonic mass or obstruction and thereby mimic colon cancer is given in Table 91-7.

Mass Lesions
Benign tumors
Diverticulitis
Inflammatory masses
Ulcerative colitis
Crohn's colitis
Tuberculosis
Amebiasis
Fungal masses (e.g., mucormycosis)
Schistosomiasis
Viral lesions (CMV, LGV)
Cronkhite-Canada syndrome
Fatty ileocecal valve
Colitis cystica profunda
Surgical anastomoses
Feces (x-ray diagnosis)
Solitary rectal ulcer
Endometriosis
Appendectomy stump
Lymphoid polyps, lymphoma
Carcinoid tumors
Metastatic lesions
Kaposi sarcoma
Extrinsic compression
Submucosal mass
Submucosal hematoma
Extrinsic mass
Obstructing Lesions
Strictures
Late sequelae of inflammation
Radiation
Ischemic colitis
Volvulus
Extrinsic compression
Endometriosis
Pancreatitis

CMV, cytomegalovirus; LGV, lymphogranuloma venereum.

TABLE 91-7 Lesions that May Mimic Colon Cancer

Colorectal neoplasms begin to lose blood early in their natural history, but most patients are unaware of this blood loss. Most colon cancers lose less than 10 mL of blood per day, and patients are not likely to notice this bleeding unless it is deposited on the surface of a formed stool from a lesion in the distal colon or rectum. If a patient observes bright rectal bleeding, it may represent bleeding from hemorrhoids or an anal fissure. Patients with inflammatory bowel disease commonly experience rectal bleeding, and so it is not a reliable sign of early neoplasia. Bleeding from ischemic bowel disease or a diverticulum is usually more brisk than that

seen from a neoplasm.

Sometimes, a locally invasive carcinoma is difficult to differentiate from an inflammatory process, even if visualized endoscopically or surgically. A mass produced by a perforated diverticulum can be bulky and indurated, suggesting a neoplastic process, but biopsy specimens from an inflammatory mass may not be diagnostic. Furthermore, certain entities may give rise to an ambiguous biopsy, including colitis cystica profunda, which may occur at a surgical anastomosis, and endometriosis.

Diagnostic Approaches to Colon Cancer

Several distinct issues associated with the diagnosis of colorectal neoplasia overlap to some degree and are confused frequently. The first of these involves the workup of the patient with symptoms of colonic obstruction, bleeding, or a locally invasive disease. Closely related to this is the workup of a positive test for occult fecal bleeding in a patient without other symptoms. Finally, there is the controversial issue of how to screen the asymptomatic patient.

Symptomatic Patients

In the symptomatic patient, colonic disease may be strongly suspected, but cancer is only one item in the differential diagnosis. The presence of a guaiac-positive stool increases the suspicion of colorectal neoplasia, but a negative test does not exclude it. [385] If the patient is not obstructed and there is no suspicion of perforation, the patient's colon should be cleansed, and colonoscopy is the diagnostic procedure of choice. Colonoscopy is the most accurate and sensitive diagnostic modality available, and it permits the biopsy of suspicious mucosal lesions. The complication rate (including perforation and hemorrhage) is less than five cases per 1000 patients. The barium enema is a less sensitive diagnostic procedure, and removal of the administered barium is required for colonoscopic verification of any suspected lesions. Nonetheless, a barium enema may provide important complementary information not available using colonoscopy, especially if there is a colonic stricture or obstruction or when an extrinsic lesion is involved. In evaluating a patient for specific colonic symptoms, the absence of blood in the stool should not exclude evaluation by colonoscopy or barium enema.

Estimating Risk for Colorectal Cancer In determining the appropriate evaluation in a patient with symptoms that may be referable to the colon, it is helpful to focus on risk factors that make the presence of colorectal cancer more or less likely. Patients younger than 40 years of age are relatively unlikely to have cancer unless they have ulcerative colitis, have an FAP variant (i.e., attenuated FAP), or come from an HNPCC kindred. The incidence of colorectal cancer begins to rise after the age of 40 years, but it remains relatively low until 50 years of age and older, when the risk rapidly accelerates. The risk for colorectal cancer continues to rise relentlessly with each decade thereafter, and age is the greatest single risk factor for colorectal cancer. [386] Genetic factors play a special role in co-lorectal cancer, but most patients do not have any genetic risk factors that are currently identifiable. A more important issue to consider is a history of adenomatous polyps because these are the precursor lesions for cancers. Particular attention should be given to patients with multiple adenomas, large adenomas, or adenomas containing villous features or carcinoma in situ. Genetic testing should be reserved for young patients with colorectal neoplasia in the setting of multiple family members with cancer.
Diagnostic Workups It is important not to overlook the visual inspection and digital examination of the anus and distal rectum, even if additional studies are planned. This permits the diagnosis of anal neoplasms and the occasional adenoma or carcinoma present on the rectal side of the anal verge that can be missed on an incomplete endoscopic examination. Furthermore, stool is obtained during this procedure to test for the presence of occult bleeding. Anoscopy and proctoscopy using a rigid instrument may be used to examine the rectum and anus, and therapeutic procedures may be performed using these instruments. The rigid sigmoidoscope is not used in the routine diagnosis of cancer except for special instances to evaluate the rectum. It is painful to advance this instrument into the distal sigmoid colon, and even in the best of hands, this is an insensitive test compared with flexible instruments. Flexible fiberoptic proctosigmoidoscopy may be performed using instruments of virtually any length. Instruments measuring 60 cm are widely available, and rectosigmoid examinations also may be performed with the standard colonoscope. A rigorous preparation is not required because examination of the sigmoid colon and rectum is usually undertaken after cleansing enemas. No sedation is required, and patient acceptance of the procedure is much higher than for the rigid instrumentation. The use of the 60-cm instrument may be mastered by general internists and endoscopic technicians with appropriate training. The barium enema is largely of historical interest in the diagnosis of polyps and cancers of the colon. The single-contrast barium examination is inadequately sensitive to exclude polyps and cancers and should be replaced by the double-contrast study. Preparation for a barium enema varies from center to center; most frequently, a clear liquid diet, a combination of saline cathartics and stimulant cathartics, and cleansing enemas are requested by the radiologist. Sedating medication is not required. About 0.03 Gy is delivered to the abdomen during this study, and patient acceptance is lower than it is for flexible sigmoidoscopy or colonoscopy; however, the entire colon may be examined by this technique. Overlapping loops of bowel in the sigmoid colon and the flexures are areas of particular difficulty in interpretation. The presence of extensive diverticulosis or residual fecal material can present additional diagnostic difficulties. There is difficulty in excluding lesions in the rectum because of the presence of the obstructing balloon required for this procedure. Thus, proctosigmoidoscopy is suggested as an adjunctive study to improve the sensitivity of a barium enema. The sensitivity of the barium enema is directly related to the patience and diligence of the radiologist. One retrospective analysis suggested that barium enemas detected 83% of colon cancers, whereas colonoscopy performed by a gastroenterologist detected 97% of colorectal cancers. Cancers detected using colonoscopy were substantially more likely to be Dukes class A (25% versus 10%). [387] One small, uncontrolled study of a series of patients who underwent single-contrast barium enema surveillance after the removal of an index large (greater than 1 cm) adenomatous polyp reported that half of the patients who developed cancers (median size, 4.5 cm) had had a negative barium enema within the past 3 years. [388] As an adjunct to the National Polyp Study, the use of barium enema was found to be inadequate for surveillance after an initial polypectomy. Only 32% of diminutive (5 mm or smaller) colonic adenomas were detected by barium enema—which is understandable and not necessarily a problem, but the radiologic approach missed 53% of adenomas 6 to 10 mm in size and 48% of those larger than 1 cm. [389] It may be most accurate to refer to the barium enema as a screening rather than as a diagnostic test because the results are not diagnostic, and abnormal findings require colonoscopy and biopsy to diagnose a neoplasm. The 180-cm colonoscope is the most widely used diagnostic instrument to study the colon. The entire colon can be examined in at least 90% to 95% of studies, and this approach has the highest diagnostic sensitivity of all available tests. Preparation of the colon is achieved using a nonabsorbable gastrointestinal lavage solution. Intravenous sedation and analgesia are administered during the examination; therefore, patient acceptance is high. However, this is the most expensive and invasive of examinations and carries with it a low (0.1% to 0.3%) incidence of severe complications, such as hemorrhage and perforation, which can require surgical intervention. A valuable aspect of this procedure is the fact that mucosal biopsy and endoscopic polypectomy may be undertaken. Thus, definitive diagnosis and even treatment may be accomplished. The performance of tandem, back-to-back colonoscopy on the same day has provided data on the minimal rate of false-negative examinations. The "miss" rate for adenomatous polyps was inversely related to size and was 6% for polyps larger than 1 cm, 13% for adenomas measuring 6 to 9 mm, and 27% for adenomas smaller than 5 mm. [390]
What Is the Definitive Diagnostic Test for Colorectal Neoplasia? If the initial diagnostic procedure is a barium enema, a negative result may create a diagnostic dilemma. In a study of 97 patients who had persistent large bowel symptoms and a negative air-contrast barium enema, colonoscopy revealed 4 carcinomas and 24 adenomatous polyps not detected by the radiologic approach. [391] In a similar study of 76 patients with symptoms of colonic disease, the diagnostic evaluation began with rigid sigmoidoscopy, after which all the patients underwent flexible sigmoidoscopy, double-contrast barium enema, and then colonoscopy. The double-contrast barium enema alone reached a final diagnosis in 67% of patients, whereas colonoscopy was successful in 91%. The addition of flexible sigmoidoscopy to double-contrast barium enema improved the diagnostic yield to 76%, [392] suggesting that colonoscopy is the preferred diagnostic procedure for the initial workup of a patient with symptoms of colonic disease and, moreover, indicates that colonoscopy should be used to evaluate colonic symptoms or bleeding even if the air-contrast barium enema is negative. A review of 31 colon cancers overlooked on double-contrast barium enema revealed that half of the lesions were missed as a result of perceptive errors; that is, the lesions were visualized on the x-ray film and recognized in retrospect, but they could not be identified as neoplasms on the initial reading. [393] Another third of the lesions were missed because of perceptive error complicated by technical factors obscuring the lesion; the tumor was not visible even on respective viewing in 10%, and in 6%, the lesion was seen but misinterpreted. In addition, almost one third of barium enemas in patients older than 65 years of age are technically inadequate because of inability of the patient to obtain a suitable preparation or to cooperate with the procedure. [394] Clearly, the figures obtained are highly dependent on the skill and persistence of the radiologist and endoscopist, and it must be emphasized that some neoplastic lesions may be missed by the colonoscopist. A second prospective study of tandem colonoscopy on 90 patients who had a total of 221 lesions demonstrated that an experienced colonoscopist missed about 15% of neoplastic lesions smaller than 10 mm but no lesions larger than 10 mm. [395] Colonoscopy is the current gold standard for the diagnosis of colorectal neoplasia, and it would appear unlikely that any indirect approach, such as radiography or ultrasonography, can approach this technique for sensitivity.

Screening Asymptomatic Individuals for Colorectal Cancer

Because colorectal cancer produces few symptoms while the tumors are small and most readily curable, screening of asymptomatic patients has been advocated (see Chapter 34). In this context, *screening* refers to testing patients in the absence of specific symptoms. Screening for colorectal cancer can reduce the mortality from this disease. There is still disagreement regarding which screening approach is optimal, the costs of the program, and how much screening can be afforded.

A good screening test must improve the lives of those screened, either by prolonging life or by improving its quality. To be effective, the test must be sensitive (i.e., it should detect all diseased individuals), specific (i.e., it should not detect nonmalignant lesions), and acceptable (i.e., it should not subject nondiseased individuals to excessive anxiety or extra testing) and affordable by those tested. Two modalities have been evaluated for efficacy as screening tests for colorectal cancer: testing of

feces for occult blood and endoscopic examination of the bowel. Both modalities are effective in reducing cancer mortality, but each has limitations.

Fecal Occult Bleeding Tests: Technical Considerations In a typical stool (150 g), the following rule of thumb may be used: Each 1 mL of blood results in about 1 mg of hemoglobin per gram of stool. The detection of tiny blood losses into the gastrointestinal tract seems like a simple task, but, in fact, this test is fraught with complexities. The normal gastrointestinal losses of blood may be estimated by the intravenous administration of ^{51}Cr-tagged erythrocytes, followed by measurement of the excretion of radioactivity in the stools. This has been done by several laboratories, and there is agreement that normal losses are about 0.5 to 1 mL/day. [396], [397] During its transit in the gastrointestinal tract, the blood is dispersed throughout the stool and undergoes degradation. Moreover, there are natural inhibitors of peroxidase activity in feces. It had previously been noted that the guaiac test was less sensitive if the stool had dried out on the filter paper; therefore, a drop of water often was added to the back of the Hemoccult II card (Beckman Coulter, Fullerton, CA) to rehydrate the stool before development of the test. In one study, 156 normal subjects were given a diet restricted in red meat, fruits, and vegetables; none of 310 of the guaiac tests done on subjects given the low-peroxidase diet was positive, and only 2 (0.6%) of 310 were positive after rehydration of the slides. [398] If the slides were rehydrated, as many as 6.6% of the tests became positive. Dietary peroxidase was principally a problem for the guaiac test after the ingestion of beef, but not with poultry, fish, or pork. [399] The standardized guaiac test is therefore reliably negative in control subjects on restricted diets, and less than 1% of tests are falsely positive on low-peroxidase diets. The test becomes unreliable if performed with rehydration unless the diet is strictly regulated, diminishing the value of this maneuver. The next critical issue in the development of a screening test was to understand how much colorectal neoplasms bleed. Mean blood loss (fecal ^{51}Cr-rbc) from a cohort of symptomatic tumors of the cecum and ascending colon was 9.3 mL/day (range, 2 to 28 mL/day) but was much less for lesions located distal to the hepatic flexure, from which the mean blood loss was less than 2 mL/day. [226] Despite the sensitivity of the guaiac test, a false-negative rate of 31% was encountered using this test, which was reduced to 9% if the guaiac slides were rehydrated. As previously mentioned, however, rehydration is a problem if the diet has not been strictly regulated with regard to foods containing peroxidase. A single guaiac test was falsely negative 50% of the time in patients with cancer. The false-negative rate fell to 31% with 3 consecutive days of testing and then to 13% after 10 days of continuous testing. The sensitivity improved with rehydration, and the false-negative rate was only 5% after 5 days, but again, rehydration may be accompanied by sharp loss in specificity depending on the diet. The proportion of positive guaiac tests is closely related to the amount of blood in the stools. The tests are usually negative if the stool hemoglobin concentration is less than 2 mL/g of stool, and they are more likely to be positive with increasing fecal hemoglobin. [226] Colonic polyps also may be detected by the tests for occult bleeding, but benign lesions lose less blood, and the sensitivity of the test is much lower. [226], [400] The mean blood loss from an adenomatous polyp is about 1.3 mL of blood per day, regardless of its location. Polyps in the distal colon (descending colon, sigmoid colon, and rectum) produce 54% positive tests, whereas those in the proximal colon produce positive tests only 17% of the time. [226] Gastrointestinal bleeding from polyps is usually detected only when polyp size exceeds 20 mm. [226] A positive guaiac test is more likely if a polyp is located in the distal rather than proximal colon. [398] Numerous attempts have been made to develop a better FOBT. The manufacturers of Hemoccult II developed a more sensitive slide test, called Hemoccult SENSA (Beckman Coulter, Fullerton, CA), which seems to provide sensitivity similar to that seen with rehydrated Hemoccult slides. The same manufacturer also developed an immunochemical test for fecal hemoglobin called HemeSelect (Smith Kline Diagnostics, Palo Alto, CA), which uses a specific antibody for human hemoglobin but is performed in the laboratory, and a result is not available immediately in the clinic, as are the guaiac-based slide tests. The antibody-based test has the theoretical advantage of not cross-reacting with nonhuman hemoglobin, and it also should not detect bleeding from an upper gastrointestinal site because of degradation of the intact molecule. Both Hemoccult SENSA and HemeSelect are much more sensitive than Hemoccult II, detecting 94% and 97%, respectively, of the symptomatic colorectal cancers, compared with 89% for Hemoccult II. The sensitivity for silent cancers, which are obviously more difficult to detect and are the target lesions, is uncertain. For adenomatous polyps larger than 1 cm, Hemoccult SENSA was positive in 76%, HemeSelect in 60%, and Hemoccult II in 42%. The Hemoccult SENSA and HemeSelect were positive in 5% and 3% of screened subjects, respectively, and most of these positive tests were not associated with colorectal neoplasia. [401] Therefore, the price for increased sensitivity is the need to perform a large number of definitive workups (i.e., colonoscopy) because of false-positive tests. A combination of these tests may be more economical and a clinically reasonable compromise. When a highly sensitive screen (i.e., the Hemoccult SENSA test) was positive and then the positive stools retested with a more specific (and expensive) test (i.e., HemeSelect), a sensitivity of 65.6% for cancers and a specificity for predicting carcinoma of 97.3% was found. [402] The combined approach had the most favorable positive predictive value of the strategies analyzed. The HemoQuant test (Mayo Medical Laboratories, Mayo Clinic, Rochester, MN) provides a quantitative measure of fecal heme and can detect the tiny increments of bleeding that occur in the setting of colorectal neoplasia. [403] The test is not influenced by dietary peroxidase and should be superior to Hemoccult as a diagnostic test. [404] Unfavorable performance characteristics and high cost considerations are such that this test has not gained popular use.

Fecal Occult Bleeding Tests: Factors Modifying Results The Hemoccult card test has been developed so that patients can take it home, modify their diets, collect two samples per day from a stool on each of 3 consecutive days, and return the cards for developing. Thus, some or all the specimens will have undergone changes because of storage. Using early-generation Hemoccult cards, less sensitive than Hemoccult II, about 8 mg of hemoglobin per gram of stool was required to produce a positive test. After stool samples had been stored on Hemoccult cards for 4 days, the intensity of the reaction diminished, and it became negative in 8 days. Therefore, low levels of fecal hemoglobin become undetectable, with a longer delay between the collection of the stool and the development of the test. [405] An inexperienced processor of Hemoccult cards can overread the result, thus seriously affecting the outcome of a screening project. [406] The effect of iron preparations on the guaiac test has been somewhat controversial. A study of 1700 guaiac tests concluded that oral iron does not result in positive guaiac reactions. [407] A small study suggested that ferrous sulfate and ferrous gluconate produced false-positive reactions in more than half of 10 patients tested, and that an aqueous solution of iron could produce a positive guaiac test. [408] However, the black pigment in stools of patients on iron supplements may have been misinterpreted as the blue color characteristic of a positive guaiac test, confirming that oral iron supplements do not produce true-positive guaiac tests. [409] Although ferrous sulfate dissolved in water produces a positive Hemoccult II test, at the pH of the stool (pH > 6.o), the iron precipitates out of solution and does not produce a positive test. Ferrous (Fe^{2+}) iron produces a positive reaction only after the addition of hydrogen peroxide, which oxidizes Fe^{2+} to Fe^{3+}. The black iron pigment in stool is insoluble ferrous sulfide, and no Fe^{2+} or Fe^{3+} is elutable into water from the black stools. [409] It is unlikely that oral iron therapy contributes appreciably to false-positive guaiac reactions, and, furthermore, it does not appear to cause occult gastrointestinal bleeding. [410] Because the basis of the guaiac test is the oxidation of an indicator substance (guaiac), the presence of strong antioxidants (e.g., 1 to 2 g/day of ascorbic acid) produces a spuriously negative guaiac test. [411]

Screening Asymptomatic Populations for Occult Fecal Bleeding Several studies illustrate the benefits and limitations of screening for colon cancer. More than 54,000 FOBT kits were sent on request to asymptomatic people [412]; only 26% of the tests were completed, of which 4.4% were positive. More than one third of those with positive tests failed to respond to repeated inquiries for follow-up, and another 20% had an incomplete diagnostic workup after referral to a physician. Among all those with positive tests, 5% had cancers detected, nearly two thirds of which were Dukes stage A or B lesions. Another 30% of the patients had abnormalities other than cancer, including adenomatous polyps. Thus, compliance is an issue in a community-screening program. In a large controlled trial, a cohort of patients aged 40 years or older was screened on a meat-free, high-bulk diet. The slides were not rehydrated, and there was a 4-day storage interval between slide preparation and testing. [413], [414] The Hemoccult tests were positive in 1.7% of the group, and results were strongly age dependent. The first-generation tests were positive in 1%, compared with 3.7% using the more sensitive version (Hemoccult II). The first-generation Hemoccult slides were predictive for neoplastic lesions in 50% of patients, including 12% cancers and 38% adenomas. The more sensitive Hemoccult II slides had a predictive value of 44% for any neoplastic lesion. Modifications intended to make the test more sensitive gave a higher proportion of positive tests, found more neoplasms, and generated many more false-positive results. Although the entire group of patients was offered at least one rigid 25-cm sigmoidoscopic examination, a 43% reduction in cancer mortality was reported in the half of the group randomized to receive the FOBTs. [414] A large (20,000 patients) controlled prospective trial studied the impact of screening for colon cancer on the detection of colon cancer subsequent to symptoms. Half were encouraged to have FOBT, and half were advised to consult their physicians if they developed colonic symptoms. [415], [416] Only 38.5% of patients who were offered screening returned their slides. Of the group returning slides, 4.1% had a positive test, and 36% of these had neoplastic disease on air-contrast barium enema and flexible sigmoidoscopy. Of the group with a positive test, 8.5% had cancer, and 28% had one or more adenomas, 30% of which were larger than 2 cm in diameter. Among the 17 invasive carcinomas detected, 13 were Dukes stage A, 2 stage B, and 2 stage C. One patient among the 4716 patients with an initial negative screen developed a stage C carcinoma of the ascending colon during the year after screening, suggesting that there had been a false-negative test. Among the subjects randomized to the screening group in the initial study, there were 384 who either refused the test or failed to reply; in this group, 11 developed symptomatic neoplastic disease within the following year, including 8 with cancer (5 stage B and 3 stage D), and 3 patients presented with large adenomatous polyps. Among the initial 10,000 patients randomized to the screening arm, a total of 24 had cancers diagnosed. Among the 10,000 control subjects, 10 patients presented with symptomatic carcinomas of the colon, 4 of which were stage B, 4 stage C, and 2 stage D. This study clearly indicated that a substantial number of asymptomatic cancers could be detected using the FOBT, and early-stage lesions were often diagnosed. [415], [416] The initial group of compliant subjects were offered rescreening; 85% were compliant, and 2.8% of those patients had positive tests. [416] Of the 80 patients with positive tests, 4 (5%) had cancers, 3 of which were stage A. In the original control group of more than 10,000 unscreened patients, 7 patients presented with symptomatic colorectal cancer during the second follow-up year (in addition to the 10 detected during the first year). No tumor was stage A; 47% (8) were stage B, 35% (6) stage C, and 18% (3) stage D. This group has updated their study after accruing more than 100,000 subjects, and the results are essentially unchanged. [417] A summary of the five published controlled studies of screening for colon cancer is provided in Table 91-8. Asymptomatic populations have about 1% to 2.4% positive tests. If more than 1% to 2% of tests are positive, it suggests that dietary restrictions have not been adequately followed, that the clinicians have used rehydration of the guaiac test, or that a more sensitive version of the test has been used. It is recommended that beef be eliminated from the diet during the lead-in period. In addition, antiinflammatory agents and antioxidants such as vitamin C should be avoided. Furthermore, the physician or technician should develop the slide as soon as possible, should not rehydrate the slides, and should be aware of the potential for misinterpretation of the slides in patients taking iron supplements. These measures minimize the number of false-positive FOBTs, which would increase the cost of a surveillance program. By limiting the sensitivity of the test, at least one third of colorectal cancers are missed.

TABLE 91-8 Controlled Trials of Fecal Occult Blood Testing in Screening for Colorectal Neoplasms

Early-stage cancers can be detected using FOBTs, and co-lorectal cancer mortality is reduced by finding these tumors. A case-control study demonstrated an odds ratio of 0.69 for colorectal cancer mortality in patients who had had their stools screened for occult fecal blood within the previous 5 years. [418] The odds ratio was lowest for the first year after screening but increased to 1 (i.e., no benefit) 3 years after screening. The impact of FOBT screening, therefore, has a time limit. The most important study of FOBTs was reported from the University of Minnesota and involved the use of Hemoccult cards, 83% of them rehydrated, on more than 46,000 patients studied prospectively for an average of 13 years. [419] In this three-armed study, annual FOBTs resulted in a 33% reduction of colorectal cancer mortality. This reduction was achieved largely through stage shift, in which there was a 50% reduction in the number of Dukes stage D tumors in the screened group, which in turn had a profound effect on survival. A substantial reduction in mortality was demonstrated in this large, credible, carefully controlled study, and the magnitude was similar to that predicted by the previous studies. [414], [418] The use of the rehydrated Hemoccult cards raised the positivity rate among screened individuals from 2.4% (their rate without rehydration) to 9.8%, necessitating diagnostic evaluations in a large proportion of the study population. In fact, 38% of the annually screened group underwent at least one colonoscopy during the study. As a result, only 2.2% of those with a positive test were found to have colorectal cancer, compared with 5.6% when the nonrehydrated tests were used briefly in the middle of the study. Although the number of adenomatous polyps removed from this group was not reported, this intervention may have added to the large and highly significant reduction in mortality seen only in the annually screened group. A follow-up analysis of the Minnesota trial demonstrated that the biennially screened group had a 21% reduction in mortality, suggesting a narrow window of opportunity for annual FOBT screening. [420] Two more prospective randomized clinical trials of the effects of FOBT screening on colorectal cancer mortality were published from Denmark and Britain. [421] and [422] Both used nonrehydrated Hemoccult cards, a practice representative of common standards, and biennial screening. The Danish study of 140,000 people aged 45 to 75 years had a 67% completion rate for screening, and the group offered biennial screening had a colorectal cancer-specific mortality ratio of 0.82 ($P = 0.03$) compared with the group offered standard care without screening. [421] The British study involved more than 152,000 people aged 45 to 75 years, about 60% of whom completed at least one screening, and a 15% reduction in cumulative colorectal cancer mortality was observed ($P = 0.026$). In the Minnesota and Danish studies, similar numbers of cancers were found in the screened and unscreened groups, whereas 4.3% more cancers were found in the screened group from Britain. In the British study, the reduction in colorectal cancer mortality among patients found in the first screen was 39% of that in the control group. It appears that the benefits from FOBT screening occur primarily by virtue of diagnosing early-stage tumors, and there is an important benefit of offering annual rather than biennial screening. Using a rehydrated Hemoccult card results in many more positive tests but substantially affects the overall costs of the program because of the need for a definitive diagnostic workup for each positive FOBT.

Role of Sigmoidoscopy in Screening for Colon Cancer Screening with FOBTs is insensitive and often nonspecific, and about two thirds of colorectal neoplasms are within reach of the flexible sigmoidoscope. A large, uncontrolled observation of the effects of screening by rigid sigmoidoscopy suggested that frequent inspection of the rectum and removal of all premalignant lesions would reduce the incidence of invasive rectal cancers to a low number and even could eliminate the occurrence of advanced-stage disease. [423] The development of flexible instruments that could reach from the anus to the cecum raised the obvious issue of how far to take the examinations and how frequently they should be repeated. Two studies have suggested a significant reduction in mortality from screening sigmoidoscopy. A case-control study examined the use of screening sigmoidoscopy among 261 patients who died of cancer of the rectum or distal colon, compared with case-matched controls. Only 8.8% of the cancer patients had undergone screening sigmoidoscopy, compared with 24.2% of the controls. The authors estimated that screened subjects had only 30% of the risk for fatal cancers of the rectum and distal colon, compared with the unscreened cohort. Furthermore, the data suggested that screening sigmoidoscopy might provide risk reduction for as long as 10 years. A similar number of fatal colon cancers were above the reach of a sigmoidoscopic examination in both the screened and unscreened groups, suggesting that the two groups were evenly matched for colorectal cancer risk. [424] This observation was confirmed by another case-control study. [425] A history of at least a single screening sigmoidoscopy over a 10-year period of study was present in 10% of those who died of colorectal cancer but in 30% of case controls. The estimated risk for death from colorectal cancer was reduced by 79% after a single examination, and the adjusted odds ratio for death from cancer in the rectum or distal colon was 0.05. Case-control studies have features similar to both prospective and retrospective studies, but they do not represent the most convincing evidence of clinical effectiveness. No large, controlled, prospective trial has been published that tests the efficacy of endoscopic screening with either sigmoidoscopy or colonoscopy. Nonetheless, there is strong and compelling evidence for a protective effect of sigmoidoscopy and, presumably, the removal of premalignant lesions. Endoscopic screening appears to bring with it a protective effect that lasts substantially longer than the 1–2 year protection of FOBTs. The magnitude of protection is substantially greater than that derived from FOBTs, presumably because it is possible not only to detect early-stage lesions but also to remove premalignant lesions and interrupt neoplastic progression early in its long natural history. Unfortunately, the costs of endoscopic screening will be considerable, depending on the protocol chosen, and the demand for this testing will compete for other health care resources.

Role of Colonoscopy in Screening for Colon Cancer The case-control studies predicted a powerful impact for screening sigmoidoscopy on mortality rates for distal colorectal cancer. Several groups have investigated the diagnostic yield of screening colonoscopy in asymptomatic adults. To date, there have been no controlled prospective trials of mortality reduction due to colonoscopic screening; hence, the actual impact of this modality still remains speculative. In a study by the Veterans Affairs Cooperative Study Group on the diagnostic yield at screening colonoscopy, more than 3000 patients underwent colonoscopy, and these were nearly all men with a median age of just under 63 years. Of the individuals who underwent colonoscopy, 37.5% had one or more neoplastic lesions, and 1% had an asymptomatic, invasive cancer. Of the individuals who had no polyps in the colon distal to the splenic flexure, 2.7% had a neoplasm in the proximal colon that was either large (10 mm or larger) or had an advanced histology. More than half of patients with advanced proximal neoplasms had no distal adenomas that may have been detected at sigmoidoscopy and triggered additional workup. [426] The major limitation of the study is that it involved virtually all men, exclusively U.S. veterans. However, a similar study undertaken in 1994 consecutive asymptomatic adults aged 50 years or older, including both men and women, with a median age of 60 years, reported similar findings to the Veterans Administration study. Twelve cases of cancer were detected (0.6% of the total); 3.1% of subjects had advanced lesions in the distal colon, and 2.5% had advanced lesions in the proximal colon. The risk for an advanced proximal lesion in the absence of distal polyps (i.e., those that would be missed with a complete sigmoidoscopy) was 1.5% of the total and represented 46% of all the patients with advanced proximal neoplasms. The relative risk for an advanced proximal neoplasm increased with the severity of lesions found in the distal colon and sequentially rose as the distal colon showed a hyperplastic polyp (relative risk, 2.6), tubular adenoma, or an advanced neoplasm (relative risk, 6.7). [427] A follow-up to the Veterans Administration study demonstrated that performing one-time screening with a rehydrated FOBT and sigmoidoscopy would fail to detect advanced colonic neoplasia in 24% of those patients. [428] These findings indicate that screening only the distal colon will miss some proportion of advanced proximal neoplasms and that using an index lesion in the distal colon to determine the need for full colonoscopy will miss some advanced lesions; however, that proportion is relatively small and represents about 1.5% to 2.7% of the entire group. Additional strategies are being explored to detect cancers and other advanced neoplasms in the proximal colon without the risks and expenses involved with total colonoscopy. The term *virtual colonoscopy* refers to the use of an imaging procedure such as a computed tomography (CT) or magnetic resonance imaging of the colon. These techniques are still in development. One study of 100 patients documented that a complete exam could be performed in 87% by virtual colonoscopy compared with 89% using conventional colonoscopy. However, virtual colonoscopy fails to detect 9% of lesions 10 mm or larger, 18% of lesions 6 to 9 mm, and 45% of diminutive polyps. There was also a 19% false-positive report of polyps, all of which will lead to a second diagnostic procedure. [429] The use of noninvasive imaging studies is promising; however, they will require additional technical improvements before they will be instituted widely.

Screening Recommendations for Asymptomatic Populations The published data indicate that mortality from colorectal cancer can be reduced by annual FOBT programs by as much as a 33% but that biennial FOBTs (which may more accurately reflect actual practice) reduce mortality by only 15% to 21%. A greater degree of prevention is achieved with screening sigmoidoscopy, which may provide a reduction in cancer mortality of about 80% for that portion of the bowel screened. Programs that use FOBTs are complicated by the variables involved in the techniques used, the need for annual testing, and the need for follow-up colonoscopy in all patients with a positive screening test. If FOBTs are used in a screening program, they should be done annually, and all positive tests must be pursued vigorously. Screening sigmoidoscopy is substantially more effective in preventing cancer mortality, but only the distal bowel is examined. The American Cancer Society, the National Cancer Institute, and several other advisory panels have recommended that screening for colorectal cancer in average-risk individuals begin at 50 years of age and consist of annual FOBTs plus flexible sigmoidoscopy every 10 years. Positive screening examinations should be followed by colonoscopy or, alternatively, double-contrast barium enema, preferably in combination with flexible sigmoidoscopy. Consensus statements have been developed for screening asymptomatic patients and surveillance of high-risk populations. [430] Although conclusive empiric data are not available to support it, colonoscopy has been recommended as a screening test every 10 years, [431] and U.S. Medicare began paying for this in 2001.

Surveillance for Colorectal Cancer among High-Risk Groups

Surveillance is not the same as screening. It refers to the periodic follow-up testing—usually colonoscopic—of individuals who are at high risk for colorectal cancer and who have previously had negative screening examinations. Each clinical situation should be considered individually.

Familial Adenomatous Polyposis All first-degree relatives of FAP patients are at 50% risk for the disease and should have annual flexible sigmoidoscopy beginning

at age 10 to 12 years because adenomas begin to appear at an average age of 16 years (see Chapter 90). [221], [277] Genetic testing can now be used to guide screening in families and is informative in more than 80% of families. [277] If a family has attenuated FAP, annual colonoscopy should be employed, probably beginning in the late teens. [277]

Hereditary Nonpolyposis Colorectal Cancer HNPCC is more complex than FAP because there is no distinctive premalignant phenotype, and the diagnosis is often suspected only because of a strong family history. Patients at risk for HNPCC develop a small number of adenomas at an early age, and these have a high propensity to become malignant. Most polyps and cancers occur above the reach of the sigmoidoscope; hence, the only acceptable screening technique is periodic total colonoscopy. The time required for the progression of adenomas to carcinomas is unknown for this disease, but it is thought to be more rapid than for sporadic tumors, and a cautious approach is necessary. Colonoscopy is recommended, beginning at about 25 years of age or 5 years before the earliest colorectal cancer developed in that family, and it should be repeated every 2 years if negative. A trial of colonoscopy every 3 years has demonstrated a 62% reduction in cancer incidence, and no cancer deaths occurred in 133 examined individuals over 15 years, compared with 9 deaths in 119 unscreened controls. [432] If adenomatous polyps are found, it is not unreasonable to repeat the examination in 1 year. If a cancer is found, subtotal colectomy is the appropriate surgery, followed by annual sigmoidoscopic examination of the residual bowel. [151] The availability of diagnostic genetic testing has a great impact on the management of HNPCC because it will permit the patient to consider prophylactic subtotal colectomy on the basis of a firm diagnosis and can free many first-degree relatives of affected patients from the anxieties of uncertainty about this disease.

Positive Family History of Colorectal Cancer After the autosomal dominantly inherited familial cancer syndromes (i.e., FAP and HNPCC) have been ruled out, many patients remain with one or more relatives who have had co-lorectal cancer (Fig. 91-7). The recognition of attenuated phenotypes adds to the possibilities of genetic factors in older patients with limited family histories. Given the possibility that this disease is susceptible to environmental influences, it follows that not all of these people are necessarily at an increased risk for cancer.

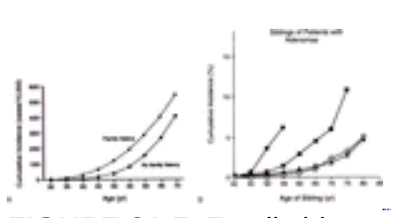


FIGURE 91-7. Family history and colorectal neoplasia. **A:** The influence of any family history of colorectal cancer on cumulative incidence of the disease. **B:** The influence of a history of colorectal cancer in siblings and the age at diagnosis on the cumulative incidence of colorectal adenomas. Filled circles: <60, parent with colorectal cancer; filled squares: =60, parent with colorectal cancer; filled triangles: <60, no parent with cancer; open squares: spouse controls. (**A** from ref. [280]; **B** from ref. [282].)

The following information may be used to target surveillance measures, but this area remains controversial. A family history of colorectal cancer in a first-degree relative (i.e., parents, siblings, children) increases the risk by about 75% to 80% over those with no family history, and variables such as multiple affected relatives or young affected relatives affect this risk sharply, as described earlier. [279], [281] Colonoscopy in first-degree relatives of those with colon cancer are found to exhibit adenomatous polyps twice as often as controls. [431], [432] Individuals with a single, elderly affected relative are not necessarily at increased risk for colorectal cancer, but the risk increases if there are two or more affected relatives or if the tumors have occurred in young people (i.e., younger than 55 years of age). Patients with any family history of cancer should enter a routine surveillance program that involves annual FOBTs and periodic sigmoidoscopy, perhaps every 3 to 5 years. Patients with multiple affected family members or with first-degree relatives who developed colorectal cancer before the age of 55 years need more surveillance, but it is not clear how much more is justified. One should perform a colonoscopic examination perhaps 5 years earlier than the earliest cancer in the family because of the tendency for familial cancers to occur above the range of the sigmoidoscope. If this is negative for neoplasia, follow-up surveillance should consist of annual FOBTs and sigmoidoscopies every 3 to 5 years unless the family history is more worrisome and suggests attenuated FAP or HNPCC, in which case colonoscopy every 2 to 3 years may be necessary. Genetic screening is valuable for the expanding group of syndromes related to FAP and is in evolution for HNPCC; referral to a geneticist is an important option for confusing histories or worried patients.

Prior History of Adenomatous Polyps Many patients have adenomas discovered through screening examinations, workups of colorectal symptoms, or other evaluations. The recommended surveillance of such patients has undergone an important change in light of new data, and a degree of restraint is appropriate for certain patients. Most adenomatous polyps (or flat adenomas) are small, do not produce symptoms, and have been discovered serendipitously by colonoscopy. The presence of solitary adenomas with diameters less than 1 cm does not predict an increased likelihood of recurrent neoplasia. [290], [291] and [292] Therefore, the detection of a solitary, small, low-grade adenomatous polyp, especially in an elderly patient, does not mandate colonoscopic follow-up. Patients who have had more advanced lesions, such as larger adenomas (more than 1 cm), villoglandular or villous adenomas, multiple adenomas, or adenomas containing high-grade dysplasia, require follow-up colonoscopy. For this group, there is no need for a repeat examination at the 1-year time interval after the colon has been cleared of lesions by a technically optimal examination. The National Polyp Study Group indicated that a 3-year interval is adequate for follow-up colonoscopy. [433] The detection of adenomas was common at the 1- and 3-year points, with polyps found in 32% of the patients examined once and 41.7% of those examined twice during the 3 years of follow-up; however, the incidence of advanced lesions (large or higher pathologic grade) was only 3.3% in both groups, and cancers were rare and discovered at an early stage. [433] Thus, patients who have had small, solitary adenomas removed do not need colonoscopic follow-up, and those with larger lesions can wait 3 years after the colon has been cleared of lesions for follow-up, if not longer, given the relatively small number of advanced lesions at 3 years. Subsequent studies suggest that for many average-risk patients, a 5-year surveillance interval may be suitable. [434] These intervals might require modification for issues such as a incomplete removal of neoplasia at first colonoscopy, a personal history of rapidly growing neoplasms, or the presence of multiple neoplasms.

Prior History of Colorectal Cancer Follow-up colonoscopy after the resection of a cancer is frequently overdone, and the objectives of such examinations should be clear to the examiner. Patients who have had sporadic cancers removed are not necessarily different from those who have had highly dysplastic adenomas removed; the principal difference between a cancer patient and a high-grade adenoma patient is the timing of discovery of the lesion. Cancer patients need an examination of the anastomotic site within the first 3 to 6 months after surgery, and perhaps again at 1 year, but the effectiveness of this approach has never been rigorously tested. In the absence of an anastomotic recurrence, the rationale for continued colonoscopic surveillance is to remove recurrent adenomas and to detect metachronous cancers. New lesions develop through the adenoma-carcinoma sequence, which typically evolves over a period of a decade or more. Colonoscopic examination every 3 years is recommended, just as for patients with advanced adenomas. If no lesions are found in a surveillance exam, the interval may be expanded to 5 years. [434]

Inflammatory Bowel Disease Patients with ulcerative colitis are at increased risk for developing cancer. The risk is lowest in the first decade of disease, but 9% develop cancer each decade thereafter. [435] Patients with colitis limited to the distal colon are at much lower risk. It is recommended that patients undergo screening colonoscopy after 8 to 10 years of disease, during which time biopsies should be done of all suspicious lesions and from multiple regions throughout the colon. If high-grade dysplasia is found in the colon, the patient should undergo repeat colonoscopy immediately, with careful inspection of the site of dysplasia and collection of multiple biopsies to exclude an adjacent cancer. Although the optimal intervals for periodic examination have not been determined, colonoscopic surveillance appears to reduce significantly colorectal cancer mortality in this setting by detecting cancers at earlier stages. [436] Barium enemas play no role in the screening for cancer in these patients because of the abnormalities produced by inflammation and scarring. Alternatively, if the patient's disease is sufficiently severe, a colectomy should be considered at this time. Such a decision should take into account the adverse effects of chronic disease as well as the risk for cancer. Patients who have no dysplasia on multiple biopsies are at relatively low risk for neoplasia during the next 1 to 2 years, and these patients should undergo surveillance colonoscopy about every 2 years. If no dysplasia is found on follow-up colonoscopy, a longer interval between surveillance studies may be appropriate. Patients with milder disease and disease limited to the distal colon are also candidates for increased intervals between surveillance studies because of the relatively lower incidence of new dysplasia among patients initially free of this abnormality. Low-grade dysplasia is more difficult to interpret, especially in the setting of acute inflammation. It is not clear whether the diagnosis of low-grade dysplasia should increase the threshold for a consideration of colectomy; however, these patients should be studied with colonoscopy at annual intervals to detect the evolution of higher grades of neoplasia. Low-grade dysplasia is not sufficiently predictive to suggest the need for colectomy. Patients with Crohn's disease are at increased risk for gastrointestinal cancer, including carcinoma of the colon. There is inadequate information to recommend how frequently patients with Crohn's disease of the colon should undergo routine screening colonoscopy. Crohn's colitis involving the entire colon should be considered in the same risk category as ulcerative colitis and should be screened the same way.

Clinical Course and Complications

The clinical course of colorectal neoplasia includes two major considerations: First, what determines the behavior of a colon cancer? Second, if a primary tumor is removed, what determines the behavior of the rest of the colon?

Natural History of Colorectal Neoplasia

The pathological stage of colorectal cancer is the best clinical predictor of outcome. Pathological stage is principally a reflection of depth of invasion. Tumor diameter

and total tumor mass are not strong independent predictors of clinical outcome. It is not unusual to find a large, bulky tumor that neither penetrates the muscularis propria nor metastasizes, and it is also not unusual to find a small, 2- to 3-cm tumor that invades and metastasizes to distant sites. This latter situation presumably represents the temporal compression of tumor progression events resulting in the early appearance of a biologically aggressive tumor. In fact, as a group, Dukes stage B tumors may be larger than stage C lesions. The relative distribution of cancers by stage is listed in Table 91-9. Actuarial age-adjusted 5-year survival rates for colorectal cancers are about 99%, 85%, 67%, and 14% for stage A, B, C, and D tumors, respectively. [365] The best prognostic indicator is the depth of invasion; crude 5-year survival rates for modified Dukes stage A tumors range from 81% to 84%; for stage B, 62% to 65%; for stage C, 36% to 40%; and for stage D, 0% to 3%. [436], [437] Five-year survival data corrected for anticipated age-related mortality are higher. Of patients presenting with large bowel cancer, about 70% will appear resectable for cure, but one third of that group (45% of the original sample) will experience recurrent disease. Only 11% of those presenting with advanced disease (30% of the group) or those with postoperative recurrence (25%) will be cured by additional surgery. [438]

STAGE*	ESTIMATED DISTRIBUTION (%)†	DETECTION BY SCREENING (%)‡
A	10	60
B	50 (± 10)	20
B1	15	
B2	35	
C	25	10
C1	13	
C2	13	
D	15	10

*Pathological stage varies by the investigator.

† These are estimates drawn from several reported studies that used differing methods to recruit and exclude patients.

‡ From Boland CR. Diagnosis and management of primary and metastatic colorectal cancers. Semin Gastrointest Dis 1992;3:33; in several large studies, 65%–90% of cancers were stage A or B when detected by screening.

TABLE 91-9 Distribution of Colorectal Cancer Patients by Pathological Stage

Metastatic Disease

It is a tenet of tumor biology that cancers are relentlessly progressive. Rarely, a patient can survive for 20 or 30 years despite an unresected primary tumor. The median survival period for patients who have hepatic metastases diagnosed at the time of surgery is only 4.5 months. [439] The clinical objective is to detect colorectal tumors early in their natural histories and to intervene with appropriate surgical therapy. Five-year corrected survival rates have been reported to range from 40% for all colorectal cancers [440] to 67% for patients who are candidates for potentially curative surgery for rectosigmoid tumors. [441] About 25% of all these patients are diagnosed with stage D tumors. In general, metastatic disease is disseminated when first detected and is only infrequently localized. One group of patients, perhaps 15% to 40%, do not have grossly detectable metastatic disease at the time of surgery but eventually succumb to recurrent disease within 5 years. It has been difficult to identify these patients prospectively to assess the impact of adjuvant treatment on their outcome.

It has long been appreciated that circulating tumor cells may be found in the mesenteric and systemic circulation of patients with colorectal cancer, and curiously, this is of no prognostic significance. [442] It is therefore not surprising that K-*ras* mutations are found in the plasma of patients with colorectal cancer, [443] but it is not clear whether any prognostic significance can be attached to this finding. More than 80% of co-lorectal adenocarcinomas are well or moderately differentiated tumors, and few prognostic features can be found to predict tumor recurrence in this group. Poorly differentiated and colloid (or mucinous) tumors make up the remainder of the adenocarcinomas, and the 5-year survival rate in these instances is somewhat worse than for their better differentiated counterparts. The primary adverse prognostic indicators are the depth of invasion and the presence of metastasis at the time of surgery. Tumor size is not an independent predictor of outcome. [444]

About three fourths of patients presenting with colorectal cancer undergo a resection that appears to be curative. The expected location of metastatic disease is related, to some extent, to the location of the primary tumor (Table 91-10). One third of these patients develop recurrent disease. About 60% of this group die with recurrences in multiple sites, one fourth develop a local recurrence at the site of the primary tumor, and a small residual number develop isolated metastases in a single organ. [438] Among patients who underwent surgical exploration for their primary tumor, 6 (14%) of 43 had overt hepatic metastases, and the other 37 (86%) had follow-ups at 3-month intervals for the development of recurrent disease. During 2 years of follow-up, 11 (30%) of the 37 developed overt hepatic metastases. Among these 11 patients with occult metastases, CT was most sensitive in detecting silent recurrences. [445] Only 1 of these 11 patients with an occult hepatic metastasis survived 3 years. [446] Colorectal cancers may metastasize to sites other than the liver or regional lymph nodes, but these tend to occur with symptoms of colonic or metastatic disease.

METASTATIC SITE	PRIMARY TUMOR SITE (%)		
	Right Colon	*Sigmoid Colon*	*Rectum*
Liver	67	68	48
Lung	31	43	52
Pelvis	9	27	41
Regional lymph nodes	49	57	59
Peritoneum	31	41	25
Adrenals	13	23	18
Bone	7	7	16
Brain	7	4	5

Summarized and adapted from Welch JP, Donaldson GA. The clinical correlation of an autopsy study of recurrent colorectal cancer. Ann Surg 1979;189:496. From Boland CR. Diagnosis and management of primary and metastatic colorectal cancer. Semin Gastrointest Dis 1992;3:33.

TABLE 91-10 Distribution of Metastases from Colorectal Cancer at Autopsy

In a study of 302 consecutive autopsies on patients with early metastatic cancer from an unknown primary site at the time of presentation, only 3.6% came from the colon or rectum. [447] Similarly, only of 3% of 62 patients who presented with cerebral metastases had a colorectal primary tumor. [448] Therefore, a colonoscopy or barium enema is a low-yield procedure in the absence of colorectal symptoms in the workup of a metastasis of unknown origin.

Treatment

Colon Cancer

Primary Surgical Treatment for Cure of Colon Cancer The preoperative workup in a patient ound to have colonic cancer requires an inspection of the entire colon, preferably with colonoscopy, because of the 5% risk for a second primary cancer, which modifies the surgical approach. An air-contrast barium enema may suffice if there is difficulty advancing the colonoscope proximal to the index lesion. Additional preoperative workup includes a CT scan of the abdomen to search for metastatic neoplastic disease, particularly in the liver. If a CT scan is performed, there is no need to perform an additional radionuclide or ultrasonographic imaging procedure of the liver. Radionuclide scanning with a monoclonal antibody directed against co-lorectal tumor antigens is available [449] but does not routinely add to the information gained from a CT scan. This modality is more valuable in localizing pelvic or abdominal recurrence, which may be difficult to find on CT. A chest radiograph is part of the routine preoperative evaluation, but a CT scan of the thorax is not recommended as a routine preoperative assessment. An imaging procedure of the central nervous system is also a low-yield procedure, and the cost is not commensurate with the probable benefit. A complete blood count is needed because of the possibility of iron deficiency anemia and its correction before surgery. In the absence of detectable metastatic disease, a measurement of serum carcinoembryonic antigen (CEA) should be done. The CEA should be drawn before removal of the primary tumor because not all cancers produce this glycoprotein, and if the preoperative value is not elevated, the test is not informative in the postoperative period. If the CEA is elevated, and if the operative assessment is that all tumor has been removed, the CEA measurement should be repeated about 1 month after surgery, assuming the physician and patient are prepared to undertake repeat surgery for any evidence of tumor recurrence. The upper limit of normal for plasma CEA is 5 ng/mL. Because CEA is excreted in bile, levels are difficult to interpret in the face of biliary obstruction or hepatic dysfunction. CEA measurement is not useful as a primary screening test because of the large number of patients with nonneoplastic conditions who have minor elevations, and it is not useful in locating the source of metastatic lesion. Serial measurements of CEA may be of value in detecting early recurrences in the postoperative patient; however, this test should not be undertaken unless the patient is prepared to accept exploratory laparotomy for an elevated

result. If this course of action is unacceptable to the patient, it may be best not to measure it. The primary modality for treatment for colonic cancer is surgical resection. The surgical approach is directed by the location of the tumor and the desire to remove not only wide margins of resection (a minimum of 5 cm proximal and distal to the edges of the tumor) but also regional lymph nodes. A cancer in the cecum, ascending colon, hepatic flexure, or transverse colon is treated with a right hemicolectomy, including a dissection of the mesenteric lymph nodes. Examination of the liver for gross metastatic disease helps predict the postoperative outcome. Tumors from the splenic flexure to the sigmoid colon are treated with a left hemicolectomy and primary colonic anastomosis preserving the rectum. Lesions in the sigmoid colon are treated with a segmental (low anterior) resection with an end-to-end primary colonic anastomosis. Every attempt is made to treat cancers in the rectosigmoid region with a low anterior resection, which permits preservation of the rectum and obviates the need for colostomy. Rectosigmoid lesions, however, require the dissection of hypogastric lymph nodes in addition to the standard mesenteric dissection. [450] The skill of the surgeon is an important prognostic factor that should be considered because the operative mortality and complication rates vary significantly among surgeons. [451] Every patient with colon cancer who is an acceptable surgical candidate should undergo resection of the primary lesion, if possible. Colonic cancers have a tendency to produce intestinal obstruction late in the clinical course, when the patient may no longer be a surgical candidate. Patients who present with obstruction are best managed with a primary resection of the tumor rather than a multistage procedure leading to delayed resection of the tumor; however, technical considerations may require tumor resection and diverting colostomy, with a delayed anastomosis. The operative mortality rate for colon cancer surgery is about 5%, [437], [451] but it may be as high as 17% for emergency operations [437] and as low as less than 3%. [452] In planning treatment, it is important to realize that this disease tends to occur in older patients with comorbid conditions.

Cancer in an Adenomatous Polyp A situation in which a segmental colonic resection may be avoided is that in which in situ or invasive carcinoma is found in a pedunculated adenomatous polyp. This subject is discussed in more detail in Chapter 149. If the endoscopist and pathologist agree that the cancer has been removed, for example, by snare cautery, additional surgery is not necessary. [453] A margin of 2 mm between the deepest extension of cancer, and the resection line is predictive of a favorable outcome with regard to tumor recurrence. [454], [455] An exception to this policy is the case in which poorly differentiated cancer is encountered.

Adjuvant Chemotherapy of Colon Cancer After Surgery for Apparent Cure The postoperative prognosis for patients who undergo primary resection of colonic cancer is based on the surgical stage of disease; about 70% appear to have received curative surgery (i.e., all tumors Dukes stage A, B, and C), and yet about one third of this group eventually develop recurrent disease. [438] The administration of chemotherapy to patients who have undergone a complete surgical resection with curative intent is referred to adjuvant therapy, which is distinct from treating known metastatic disease. Numerous chemotherapeutic regimens have been used in an attempt to reduce the recurrence rate of metastatic disease. [456] Before 1989, no regimen had been shown to improve survival in this disease. The first step forward was demonstration of a significant survival benefit with levamisole plus 5-fluorouracil (5FU) as adjuvant therapy in patients with Dukes stage C colon cancer. [457] This study included 401 patients with Dukes stage B or C colorectal cancer, excluding those with stage A cancer (in which the predicted outcome is too good to justify adjuvant therapy) and those with stage D cancer (who, by definition, have advanced disease). The patients were randomized to receive either no additional therapy or 1 year of treatment with levamisole (50 mg every 8 hours for 3 days, repeated every 2 weeks) plus 5FU (450 mg/m ^{2} daily intravenously for 5 days, followed on day 28 with single weekly injections of 450 mg/m ^{2}). Separate arms were randomized to receive only levamisole or to receive no adjuvant treatment. Stage B patients were those with tumor that had invaded the serosa, pericolonic or perirectal fat, or adjacent organs by direct extension, without lymph node metastasis (stage B2 according to some classifications). Stage C patients had involvement of regional lymph nodes but no distant metastases. The median length of follow-up was longer than 7 years, and the minimum was longer than 4 years. A significant reduction (31%) in tumor recurrence rate was found for levamisole plus 5FU, and a reduction of borderline significance (27%) was found for levamisole alone. Combination therapy also produced a delay in the time to tumor recurrence. Considering patients of both stages eligible for analysis, no improvement in survival was seen for any treatment regimen compared with controls. The patients in this study were analyzed separately by tumor stage. No improvement in the recurrence interval or survival was found for patients with stage B tumors, but a significant improvement in recurrence-free interval and survival was found in stage C tumors for the combination of levamisole plus 5FU compared with control patients. No benefit was found for levamisole alone. The authors concluded that substantial benefits would be achieved if stage C patients were treated with adjuvant therapy. These results were confirmed in a larger study of 1296 patients with either locally invasive (stage B2) or stage C colon cancers; rectal tumors were excluded, and median follow-up time was 3 years. [458] No benefit for tumor recurrence or survival was found among the patients with stage B2 disease. A 41% reduction in tumor recurrence was found in patients with stage C disease treated with levamisole plus 5FU ($P > 0.0001$), and the death rate was reduced by 33% in this group (Fig. 91-8). No benefit was found with levamisole alone. A final report on this study confirmed the durability of the initial findings, [459] and a cost analysis suggests that adjuvant therapy for stage C (III) colon cancer is very cost-effective. [460] Although patients with stage B2 cancers are considered for adjuvant therapy by some oncologists, their relatively good 5-year prognosis (77% 5-year survival rate) suggests that it may be difficult to provide significant improvement with acceptable toxicities. [461]

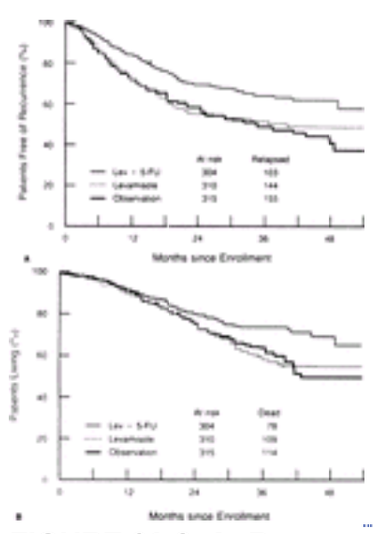


FIGURE 91-8. A: Recurrence-free interval after adjuvant treatment of stage C colon cancer with levamisole (*Lev*) and 5-fluorouracil (*5-FU*). There was a significant improvement in recur-rence-free status from 47% to 63% among treated patients at 42 months ($p < 0.0001$; **top**). The reduction in recurrence was greatest for sites outside the abdomen. **B:** Survival after adjuvant treatment of stage C colon cancer with Lev and 5-FU. There was a reduction in the death rate by about 33% in the treated group compared with controls ($p = 0.006$; **top**). (From ref. [458].)

Patients should be advised that considerable toxicity probably will be encountered with chemotherapy, but that significant improvement in disease-free survival may be expected for patients with stage C cancer of the colon. No benefit has been demonstrated for any adjuvant regimen applied to patients with stage A or B colorectal cancer. More than half of patients experienced nausea, 47% experienced diarrhea, and 17% experienced vomiting during the maintenance phase of treatment, although the side effects have been described as mild by the authors. [458] Stomatitis, dermatitis, and alopecia each were observed in about one fourth of the patients. Mild leukopenia (2000 to 4000 leukocytes) was encountered in 38% of patients. Numerous neurologic symptoms were experienced by 18% of patients; these resolved after therapy was discontinued. Fully 30% of patients discontinued their therapy prematurely because of toxicity, most commonly because of nausea. The current standard of practice in the community for stage III colorectal cancer consists of 5FU weekly and levamisole given 3 days every 2 weeks for a year. Leucovorin plus 5FU for a year is equally efficacious, and leucovorin has largely replaced levamisole in many communities. [462] 5FU, levamisole, and leucovorin given together for only 6 months also reduces mortality, whereas 5FU plus levamisole for only 6 months is not as efficacious.

Follow-up of Patients After Surgical Resection Although about 70% of patients appear to have complete excision of all malignant disease, more than one third of this group suffer later recurrences. In a small proportion of these patients, timely therapy may prolong survival. Several approaches have been developed to detect early recurrences and to treat them effectively. Colonoscopy should be performed once between postoperative months 3 and 6 and again 12 months postoperatively to detect recurrences of the primary tumor and the development of metachronous or missed synchronous tumors. Recurrences of primary tumor occur at the anastomotic site and may or may not be detectable using colonoscopic surveillance because some occur on the serosal aspect of the bowel. Anastomotic recurrences most often develop after rectal or rectosigmoid resections, and the likelihood is strongly dependent on the Dukes stage of the resected cancer. Most appear within 2 years of the initial operation, and 90% are heralded by persistent symptoms. It is important to diagnose local recurrences and to be aware that some of these are submucosal because there is a 49% 5-year survival rate after resection of such disease. [463] Measurements of serial CEA levels at 2-month intervals and CT scans at 3- to 6-month intervals have been evaluated as means for detecting early metastatic disease. There is little sense in measuring serial CEA levels unless the patient and physician are prepared to undertake additional surgery for an elevated level. CEA measurements vary among laboratories; therefore, a reference laboratory with careful quality control is essential. Recurrent tumor was found at a second-look operation in 56 (93%) of 60 patients operated on solely because of serial elevations of CEA. Recurrent tumor was later identified in 3 of the remaining 4 patients, and 1 had a spurious elevation of CEA as a result of liver disease. About half of these patients had resectable tumor, and more than one third have had long-term disease-free survival times. [464] Another group reported recurrent disease in 33 (89%) of 37 laparotomies performed for elevations of CEA in asymptomatic patients, with a 43% resectability rate, evenly split between local recurrences and liver metastases. [465] Others have reported that the use of serial CEA values to direct laparotomy for recurrent disease may result in long-term, disease-free survival in only 3% of a group of patients who had follow-up after colorectal surgery. [465], [466] Furthermore, the cost of follow-up per resectable tumor has been estimated at about $25,000, exclusive of the cost of CT scanning; the added cost of the radiologic procedure, if it is used on all patients, could triple this cost. [467] Therefore, the willingness of the patient to

undergo additional procedures and operations as well as the impact of total cost must be considered in planning an appropriate follow-up schedule in light of the minor impact on patient outcome provided by this surveillance.

Radiation Therapy for Colon Cancer Radiation therapy has been evaluated as an adjuvant therapy for colon cancer and as treatment for metastatic disease. There is no benefit with the use of radiation therapy as an adjunctive agent for colon cancer outside the rectum. Adenocarcinomas of the colon tend to be relatively radioresistant, and the toxicities from abdominal radiation are considerable.

Rectal Cancer

Surgical Therapy The approach to the treatment of rectal carcinoma is dictated by the fact that most of this organ is located beneath the peritoneal reflection and is surrounded by perirectal fat. Any degree of invasion greater than Dukes stage A requires a wide excision, usually consisting of a combined abdominoperineal resection and subsequent colostomy, which is not required for resections of carcinoma in the colon. Lesions located high in the rectum (more than 6 cm from the anus) may be treated with a low anterior resection without total proctectomy, but this approach brings with it a higher likelihood of local recurrence. Invasive cancers in the distal 6 cm of rectum are best treated with an abdominoperineal resection and hypogastric lymph node dissection. If the degree of invasion is not clear on physical examination, endorectal ultrasound may help to provide important staging information. [468] A patient with a small rectal neoplasm, even in the distal 6 cm, may refuse a proctectomy and colostomy because of the possible complications and changes in lifestyle that accompany this operation, or the patient may not be a suitable operative candidate for this procedure. In this instance, a local excision of the tumor may be considered. The preferred lesions for this approach should be small (less than one third the circumference of the rectum); they should be readily mobile, indicating only early or localized invasion of the submucosal tissues; and they should not be mucinous or poorly differentiated carcinomas, which carry a more ominous prognosis. Abdominoperineal resection was once standard treatment for many rectal cancers, but surgeons with interest and expertise in this tumor are able to perform successful local resections with a sphincter-sparing operation on a growing proportion of these lesions. A study of 57 patients with distal rectal cancer treated with full-thickness local excision demonstrated an overall 5-year survival rate of 83%, and the cancer-specific mortality rate was 11%. [469]

Rectal Polyps Pedunculated polypoid lesions in the rectum can be removed using snare polypectomy. Sessile polyps, which may create a therapeutic dilemma elsewhere in the colon, often can be treated more aggressively in the rectum, because of the additional margin of safety provided by the subperitoneal location of the rectum. Transanal local excision may be elected for mobile cancers that do not penetrate the bowel wall and are not poorly differentiated adenocarcinomas, especially for the patient who is not a surgical candidate or who refuses abdominoperineal resection and colostomy. Current standards of care would suggest that the resectability of a questionable rectal lesion should be confirmed with endorectal ultrasound.

Other Therapies Because of the relatively encouraging outcome after local excision in patients with small rectal cancers, other nonsurgical modalities have been evaluated. Neodymium:yttrium-aluminum garnet (Nd:YAG) laser photocoagulation has been safely applied for the removal of adenomatous polyps that cannot be removed by standard snare polypectomy. [470] Laser ablation also may be used as palliative treatment for symptomatic rectal cancers in patients who are not candidates for surgical resection. [471], [472]

Radiation Therapy to Improve Operability in Rectal Carcinoma The role of preoperative radiation therapy is often overlooked in the management of rectal cancer. A retrospective review indicated that 20 (80%) of 25 patients with initially unresectable rectal cancer were converted into surgical candidates by preoperative radiation therapy, and 16 (80%) of 20 patients underwent resection for cure, with 2- and 5-year survival rates of 56% and 43%, respectively. [473] Other groups confirmed the value of preoperative radiation for rectal cancer. [474] As many as 50% of patients with a fixed rectal cancer can be operated on with apparent long-term benefit after radiation therapy. [475] A subsequent prospective study indicated that postoperative radiation therapy (or radiation therapy plus adjuvant chemotherapy) for rectal cancer prolonged survival in treated patients compared with untreated controls. [476] Additional experience in a controlled study suggests that some of the benefits achieved by preoperative radiation therapy also may be achieved using postoperative radiation. [477] Most surgeons prefer not to delay surgery for preoperative radiation; therefore, postoperative radiation is more commonly used. Nonetheless, a prospective trial of preoperative short-term radiotherapy (25 Gy delivered in five fractions over 1 week) for resectable rectal cancers demonstrated that the therapy did not adversely affect postoperative mortality and was associated with a 58% 5-year survival rate and 74% cancer-specific survival at 9 years. Outcome measures such as recurrence rates and survival significantly improved for most of the subgroups and were diminished in none. [478] The use of preoperative radiation may be of benefit and is probably an underused therapeutic option.

Adjuvant Therapy for Rectal Carcinoma The incidence of local recurrence in rectal cancer is dependent on the pathological stage; local recurrence is found in 8% of Dukes stage A, 31% of Dukes stage B, and 50% of Dukes stage C patients, and the unadjusted 5-year survival rates are 77%, 44%, and 23%, respectively. [479] Effective adjuvant therapy must take into account both local and systemic disease. A regimen of intravenous 5FU in combination with external-beam radiotherapy, significantly prolonged the time to tumor recurrence, but not overall survival, in a multiinstitutional study of patients with Dukes stage B and C tumors of the rectum. [480] In another large study, patients with stage B and C rectal cancers showed some benefit from chemotherapy with 5FU, semustine, and vincristine, but improvements in survival or disease-free intervals with radiation alone were not seen. [481] Newer regimens involving radiation and combination chemotherapy are used for pre- and postoperative regimens.

Combined Radiation and Chemotherapy for Rectal Cancer Combined postoperative adjuvant treatment consisting of chemotherapy and radiation is of benefit in patients with locally invasive (Dukes stage B2) or Dukes stage C rectal cancer. [482] Combined therapy consisted of an oral dose of semustine plus 5FU, followed by external-beam radiation. The radiation therapy consisted of a total of 4500 cGy and was delivered in divided doses 5 days a week for a period of 5 weeks. This form of therapy reduced pelvic recurrences by 46%, distant tumor spread by 37%, and patient deaths by 27%. Estimated 5-year recurrence rates in patients receiving radiation only was about 63% and was reduced to 42% in patients receiving the combination therapy (Fig. 91-09). Toxicity of this treatment may be considerable. The acute side effects include bone marrow suppression and diarrhea, and late complications of radiation include proctitis, strictures, and bowel obstruction (occurring in 6.5% of all patients). Combined radiation and chemotherapy constitutes an appropriate adjuvant therapy to offer patients who have undergone apparently successful resections of Dukes stage B2 and stage C rectal cancers. A randomized study of 660 patients with TNM stage II or III rectal cancer demonstrated that the use of semustine did not add to the increased survival time provided by 5FU alone, and dropping semustine avoids the potential problem of introducing secondary neoplastic complications. Protracted infusions of 5FU during pelvic irradiation produces delays in relapse times and results in improved survival. [483]

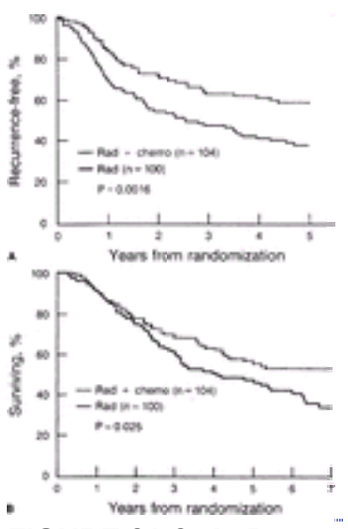


FIGURE 91-9. A: Recurrence-free intervals in patients with stage B2 or C rectal cancer treated by postoperative radiation therapy with or without chemotherapy. Patients receiving chemotherapy had 34% fewer recurrences. **B:** Survival rates in patients with stage B2 or C rectal cancer treated by postoperative radiation therapy with or without chemotherapy indicated a significant improvement in survival with adjuvant chemotherapy. (From ref. [482].)

Patients with rectal cancer who appear to be good operative candidates at the time of presentation should undergo immediate operative therapy, either abdominoperineal resection or local excision with mesorectal resection. Patients with Dukes stage A and B1 lesions have a relatively favorable 5-year prognosis and need no further therapy. Patients with stage B2 and C lesions should undergo resection and postoperative combined adjuvant radiation and chemotherapy. Patients who appear to be inoperable at the time of presentation should undergo radiation therapy first and then should be evaluated for the possibility that the tumor has been rendered operable. Newer combinations of chemotherapeutic agents are continuously being investigated.

Metastatic Colorectal Cancer

Resection of Metastatic Disease Anastomotic and other local recurrences of cancer may occur after resections in the colon but are more likely to occur in the pelvis after a rectal or rectosigmoid resection. Most patients with recurrences present because of symptoms, and it is reasonable to consider surgical resection of the metastatic disease. The use of CEA measurement and CT scanning has resulted in the identification of an increasing number of patients with isolated metastatic

disease in the liver. If a single hepatic metastasis or a cluster of lesions in a single lobe is discovered without other evidence of extrahepatic disease, surgical resection should be considered the primary approach because it is the only treatment that may provide a long-term, disease-free clinical course. The median survival time for an untreated solitary hepatic metastasis is about 10.6 months. [484] The operative mortality rate for this procedure is less than 5% in experienced hands, and 25% to 35% of operated patients are reported to experience a 5-year, disease-free survival. [485] Tumors recur in the liver in about one third of these patients, but a larger number of recurrences develop in extrahepatic organs. Solitary resected lesions that are smaller, that have wide margins of resection, and that are unassociated with extrahepatic disease are more likely to be resected for cure. [486] Up to three resectable metastases and a preoperative CEA level of less than 5 ng/mL are also factors that are predictive of a better outcome. [487] The limiting factor in successful resection of metastatic disease is the identification of patients who are suitable for resection because they represent fewer than 10% to 15% of all patients with follow-up after initial colonic surgery with curative intent. A useful application of positron emission tomography (PET) is the detection of foci of metastatic disease. PET can detect intrahepatic foci of tumor that are not apparent by conventional imaging, but it can be particularly helpful in detecting extrahepatic foci. [488] The outlook is less promising for resection of isolated pulmonary metastases. A series of 62 patients reported 5-year survival rates of 42% and 10-year rates of 22% after thoracotomy, with a mean overall survival time of 24 months. A solitary pulmonary metastasis less than 3 cm in diameter should be considered for a surgical resection, but the impact of this on survival and cure rates is undetermined. [489], [490]

Chemotherapy for Metastatic Colorectal Cancer Fewer than half of all colorectal cancer patients eventually develop metastatic disease, either at a local site or in a distant organ. Only a small proportion of metastatic disease is amenable to surgical resection. Hence, many forms of medical therapy have been tested in an attempt to manage this complication effectively. Several systematically administered cytotoxic therapies can shrink measurable metastatic disease. A wide range of objective responses have been reported in the literature, but a partial response rate of 15% to 20% is a reasonable estimate of efficacy. Although patients whose tumors shrink in response to 5FU live longer than patients whose tumors are resistant to treatment, there is no evidence that a group of patients treated with chemotherapy experiences prolonged survival, nor is it possible to know whether patients whose tumors respond to treatment live longer than if they had not been treated at all. Attempts have been made to enhance the effectiveness of 5FU for metastatic (stage D) colorectal cancer by adding leucovorin or methotrexate. Trials suggest that combined therapy may increase the number of responses, although the improvement is modest. [491], [492] and [493] The added agent often enhances toxicity more than the response rate because of the inherent limitations of the antimetabolites used to treat colorectal cancer. Any claims of treatment efficacy must be documented in the setting of a prospective, controlled clinical trial. The drug irinotecan (Camptosar), a topoisomerase I inhibitor, has been tested in a clinical trial to treat metastatic co-lorectal cancer in combination with 5FU and leucovorin. The use of three drugs resulted in a significantly longer progression-free survival, and a median survival time of 7.0 months was observed, compared with 4.3 months in patients treated with 5FU and leucovorin alone. In addition, a higher rate of confirmed response (39% versus. 21%) and a longer overall survival (14.8 versus 12.6 months) was observed with the three-drug combination. However, although statistically significant differences in outcome were confirmed in these trials, the benefits collapse in the period 12 to 24 months after treatment, and the clinician must discuss with the patient whether 1 week of infusion therapy every 4 weeks for 6 months is an acceptable strategy to gain a few months of disease-free survival. [494] Some patients will find this to be a reasonable form of therapy; however, others will refuse.

Chemotherapy for Localized Hepatic Metastases More than 80% of patients who die with metastatic colorectal cancer suffer hepatic involvement, a fact that has prompted interest in developing effective therapies to control the growth of tumor in this organ. Surgical resection should be considered for isolated hepatic metastases; unfortunately, these are uncommon. Because 95% of the blood flow to metastases is delivered by the hepatic artery, attempts have been made to intensify cytotoxic therapy while limiting systemic toxicity by administering the drug through the hepatic artery. The initial experiences with the intraarterial hepatic artery infusion chemotherapy used a percutaneous access port and reported complete disappearance of tumor in 15% of patients, 50% reduction in tumor sizes in 39%, and 25% shrinkage of tumors in another 21%. [495] Systemic drug toxicity was minimized, but a number of complications occurred, including displacement of the catheter tip and arterial thrombosis. A totally implantable intraarterial infusion pump was introduced in 1979 in an attempt to improve the uniformity of drug delivery. A group of 93 patients were treated with hepatic arterial infusion of 5-fluorodeoxyuridine (FUDR), of whom 45% had failed prior systemic chemotherapy. Significant reductions in tumor size were seen in 83% of patients, with a median response of 13 months and a median survival of 25 months after diagnosis of the liver metastasis. Extrahepatic tumor did not respond to the therapy. [496] Unfortunately, the study included no control group. In a similar study, 65 patients were treated in a prospective randomized trial of surgery (if possible) plus continuous hepatic artery chemotherapy, and no significant improvement in survival was seen with FUDR. [497] In a follow-up study, toxicity was reported in more than 80% of the patients, including mucosal damage in the stomach and duodenum, cholestatic liver disease, and strictures of the bile ducts. [498] These investigators subsequently reported the results on 162 patients after gaining additional experience with the technique and limiting the complications. Intrahepatic arterial FUDR produced a significant improvement in response compared with systemic FUDR, with response rates greater than 50%. [499] A randomized study of 100 patients with liver metastases treated with continuous hepatic arterial floxuridine reported a prolongation of survival with improved quality-of-life measures among the treated patients. [500] A metaanalysis of this approach suggested that the overall benefit is modest. [501] Unfortunately, this therapy requires an operation, it still has a considerable number of serious complications, and a significant prolongation of survival has never been demonstrated. [493] A growing number of patients will undergo resection of hepatic metastases from colorectal cancer, and there is a modest body of evidence suggesting that the actuarial survival may be improved if these patients are treated with hepatic arterial infusions of 5FU and FUDR, with or without leucovorin in six cycles. This treatment provided a modest but statistically significant increase in survival free of hepatic progression compared with surgery and systemic therapy alone. [493]

Emerging Therapies for Colon Cancer Several groups have attempted to manipulate the immune system to improve outcomes in patients with locally advanced or metastatic colorectal cancer. To date, neither vaccine-based approaches nor the use of tumor-infiltrating lymphocytes have been shown to have an impact on colon cancer. The treatment of colon cancer with 5FU produces modest benefits but with relatively modest toxicity compared to other forms of cytotoxic chemotherapy. One major drawback is the need for infusion therapy. Patients who are dealing with what is likely to be a lethal disease may be reluctant to sacrifice high quality of life to the infusion center receiving treatment when they are feeling well. Thus, oral fluoropyrimidine therapy is now being tested for colorectal cancer, including tegafur (UFT), capecitabine, eniluracil, and others. [462] The clinical benefit in terms of tumor regression is likely to be modest; however, the new approaches to giving the drug may improve the quality of life for patients with metastatic colorectal cancer who wish to take some therapy for their disease.

Other Tumors of the Large Intestine

A variety of cancers other than adenocarcinoma also occur in the large intestine and enter into the differential diagnosis of a colonic mass discovered because of symptoms or an abnormal screening test. Lymphomas and carcinoid tumors are neoplasms that arise from cells intrinsic to the colon. Much less commonly, sarcomas may arise from the smooth muscle or adipose tissue of the submucosa. Adenoacanthomas, squamous cell carcinomas, and undifferentiated carcinomas appear to represent variant types of differentiation that may have originated from the colonic epithelium. Additionally, primary tumors from other sites occasionally may metastasize to the colon. These tumors are so rare that it is difficult to generalize about their clinical characteristics.

Colonic Lymphomas

Primary colonic lymphomas constitute a relatively small proportion of all colonic malignancies (less than 0.5%) but are important because of the substantial differences in treatment and prognosis. [502], [503] A study of 117 patients with gastrointestinal lymphoma indicated that 48 (41%) occurred in the stomach, 37 (32%) in the small intestine, 13 (11%) in the ileocecal region, 2 (2%) in the appendix, and 11 (9%) in the colon. [502] Using Rappaport's classification, 60% were diffuse histiocytic lymphomas, and the remainder were divided among multiple categories. The clinical features of ileocecal and large intestinal lymphomas are presented in Table 91-11. The symptoms are nonspecific and include abdominal pain, gastrointestinal bleeding, and constipation. Ileocecal lymphoma tends to occur in a younger group of patients than does lymphoma in the rest of the large intestine. About half of gastrointestinal lymphomas have regional lymph node involvement or widespread dissemination at the time of diagnosis. The 2-year actuarial survival rate was about 40% but was related to the stage of disease; patients with stage IE or IIE lymphomas had 2-year actuarial survival rates of 82% and 71%, respectively, whereas no patients with stage IV disease survived 2 years.

	LOCATION OF LYMPHOMA	
	Large Intestine	*Ileocecal*
Number of patients	8	13
Average age (range) in years	58 (52–77)	37 (7–70)
Gender (M:F)	0.6:1	3.3:1
Presenting features (%)		
Abdominal pain	50	100
Anorexia	12	23
Diarrhea	50	38
Constipation	0	7
Weight loss	12	31
Gastrointestinal bleeding	62	29
Abdominal mass	25	29
Two-year actuarial survival	38%	41%

From ref. 502.

TABLE 91-11 Colonic and Ileocecal Lymphomas: Clinical Features

An increased likelihood of gastrointestinal lymphoma is seen in a variety of autoimmune diseases, including rheumatoid arthritis, Sjögren syndrome, systemic lupus erythematosus, Wegener granulomatosis, congenital immune deficiency syndromes, and acquired immunodeficiency syndrome (AIDS). Patients who receive organ transplants and are treated with immunosuppressive drugs have a greatly increased likelihood of developing secondary malignancies, including gastrointestinal lymphomas. The gross and microscopic pathology of these lesions is highly variable, but they are more likely to produce single or multiple discrete lesions than diffuse colonic involvement, which may be found in about 10% of cases. Although rectal lymphoma has been reported in the setting of chronic ulcerative colitis, this complication of inflammatory bowel disease is rare. [504]

Isolated colonic non-Hodgkin lymphoma is rare, occurring mostly in the cecum, and immunosuppression is a common risk factor. [505] A report of eight rectal lymphomas also showed few unifying characteristics. Patients presented with a variety of symptoms, including a change in bowel habits, lower abdominal pain, and rectal bleeding, and the lesions ranged from small submucosal nodules to diffuse friable abnormalities. [506]

Treatment of colonic lymphomas has included the use of surgery, radiation, and chemotherapy. Most treatment regimens include chemotherapy; however, the optimal form of treatment and its impact on survival remain to be established.

Kaposi Sarcoma in the Colon

Before the emergence of the AIDS epidemic, Kaposi sarcoma was a rare neoplasm largely confined to the skin, with infrequent systemic involvement. It appears, however, that Kaposi sarcoma as a complication of AIDS includes gastrointestinal involvement in 75% of patients. [507]

The stomach and duodenum are involved more frequently, but about one third of these patients also have colonic involvement, and subcutaneous nodules and infiltrative lesions may be demonstrable using radiographic studies. [508] /SUP>In a study of 50 patients with Kaposi sarcoma, characteristic lesions were visualized using a 60-cm flexible sigmoidoscope in 28% of patients. Gastrointestinal Kaposi sarcoma lesions are dark-red macules, plaques, or nodules with the appearance of a submucosal hemorrhage, and they are recognizable to those familiar with the cutaneous lesion. These lesions are submucosal, and the biopsy confirms the visual impression in only 36% of cases. The lesions tend to be multiple but are rarely symptomatic, and no adverse clinical sequelae of these lesions have been identified. Visceral involvement of Kaposi sarcoma in AIDS patients is associated with a significantly poorer prognosis, and only one of nine patients with follow-up for 2 years after the diagnosis of gastrointestinal Kaposi sarcoma was still alive. [509]

Carcinoid Tumors of the Colon and Rectum

Carcinoid tumors are neoplasms derived from cells capable of synthesizing a wide variety of hormones and autacoids. Carcinoid tumors begin as small submucosal nodules and tend to be asymptomatic until the overproduction of a functionally active peptide produces a paraneoplastic syndrome such as flushing, wheezing, or diarrhea. Carcinoid tumors may occur anywhere throughout the gastrointestinal tract, but they are most common in the appendix, ileum, and rectum (Table 91-12). More than half of these tumors occur in the appendix or ileum, and 17% may be found in the rectum. Whereas carcinoid tumors originating from the stomach or small intestine can give rise to a variety of systemic syndromes, rectal carcinoids do not produce the carcinoid syndrome. Most are asymptomatic, and about one fourth present with rectal bleeding. In a series of rectal carcinoid tumors, 23 (61%) of 38 were less than 2 cm in diameter, only one of these was malignant, and none had distant metastases. Therefore, most carcinoid tumors are amenable to local excision. Among the 15 (39%) of 38 tumors 2 cm or more in diameter, 14 were malignant, but only one gave rise to distant metastases. [510] These larger lesions should be treated aggressively, like adenocarcinomas. Carcinoid tumors of the distal colon often are diagnosed incidentally during the biopsy of a polypoid lesion, and the key to their management is to ensure their complete removal and to consider the possibility of multiple lesions. Carcinoid tumors in the proximal colon, appendix, and ileum have a somewhat different behavior: they may be locally invasive; stimulate desmoplasia in the retroperitoneal or mesentery, giving rise to a variety of mechanical complications such as intestinal obstruction; and may metastasize to the liver with symptoms such as abdominal pain, cramps, diarrhea, flushing, fixed cutaneous changes, and wheezing. A carcinoid tumor confined to the intestinal tract does not produce the characteristic features of carcinoid syndrome, which is actually a manifestation of metastatic disease. [511] and [512]

ORGAN	PERCENTAGE OF TOTAL	PERCENTAGE WITH METASTASIS
Stomach	3	18
Duodenum	1	16
Jejunum	2	35
Ileum	28	35
Appendix	47	3
Colon	2	60
Rectum	17	12

Adapted from ref. 510.

TABLE 91-12 Distribution of 3000 Gastrointestinal Carcinoid Tumors

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CHAPTER 92

Jeffrey L. Barnett

ANORECTAL DISEASES

ANORECTAL EXAMINATION

The anorectal examination is an important part of the gastrointestinal evaluation and not merely a means of obtaining stool for occult blood testing. It must be performed with particular care and thoroughness in anyone with anorectal complaints (Table 92-1). The anorectal evaluation is usually reserved for the terminal portion of the examination after the patient-physician relationship has at least been partially established. Step-by-step explanation, reassurance, and gentle technique help to minimize patient embarrassment and discomfort. The patient should be placed in the left lateral decubitus position with the buttocks protruding just beyond the edge of the examining table. The hips may be slightly raised with a sandbag or a folded sheet. The prone jackknife position is ideal for examination of the anorectum, although it requires a specialized examining table and places the patient in a somewhat unfamiliar position. After the patient is adequately draped and the instruments are at hand but out of direct patient view, examination begins with inspection of the perineum. The buttocks, sacral region, and thighs are observed for signs of pilonidal disease, dermatologic conditions, and infections. The buttocks are then firmly retracted with both hands to permit inspection of the perianal region. Fistulous openings, fecal or mucus soiling, and excoriations or chronic skin changes give clues to underlying disease processes. Anal pathology such as tumors, skin tags, anal fissures, and prolapsing hemorrhoids are often best identified at this time. Lesions should be described with regard to their anatomic location.

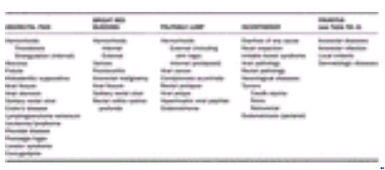

TABLE 92-1 Differential Diagnosis of Primary Presenting Symptoms of Anorectal Diseases

Clock-face descriptions are confusing unless patient position and orientation are specified. Digital examination begins with palpation of the perianal area. Induration, tenderness, or the cord of a fistulous tract may be appreciated. The examining finger is well lubricated and slowly, gently inserted into the anus. Palpation begins away from the area of suspected pathology. Anesthetic lubricant may be necessary in those with painful lesions. Occasionally, adequate examination is impossible except under anesthesia. External hemorrhoids, thrombosed internal hemorrhoids, fissures, and fistulous tracts should be sought. Sphincter tone and the posterior puborectalis impression should be appreciated with the patient at rest, during squeeze, and when bearing down. The presacral area, the prostate, and even the cul-de-sac region may be palpated. Anteriorly, the impression of the cervix or an indwelling tampon should not be confused with pathology. During the examination of the rectal mucosa, the presence of polyps, tumors, feces, and foreign bodies is ascertained.

Anoscopy is the best means of examining the anal canal. It should be performed only if the patient has specific anal complaints. Fistulous openings and anal canal lesions are noted. Internal hemorrhoids may be seen arising just above the dentate line. Their true magnitude is best appreciated if the patient bears down and the hemorrhoids are seen to bulge into the lumen. Fissures and perianal lesions may be appreciated as the instrument is withdrawn to below the dentate line. Rigid proctosigmoidoscopy is performed for full evaluation of the distal colon and rectum. The rigid instrument is particularly useful for preoperative documentation of the position of a rectal cancer, large-bite rectal biopsy specimens for diagnosis of Hirschsprung disease, removal of foreign bodies, and evaluation of the mucosa if fiberoptic or video equipment is unavailable. For most other indications, the flexible instrument is preferred.

HEMORRHOIDS

Hemorrhoids result from dilation of the superior and inferior hemorrhoidal veins. These veins form a hemorrhoidal plexus, or cushion, in the submucosal layer of the lower rectum. Because the hemorrhoidal cushion is a normal anatomic structure, all adults are candidates for the development of symptomatic hemorrhoids. In the United States, estimates of prevalence range from 4.4% to as high as 50% of the adult population. [1], [2] Although it is widely believed that constipation is an important

risk factor for hemorrhoids, recent studies suggest that diarrheal disorders are more frequently associated with hemorrhoidal disease. [3], [4]

Anatomic Considerations

Hemorrhoids may be either external or internal, and often both types are present in the same individual. Internal hemorrhoids arise from the superior hemorrhoidal cushion above the mucocutaneous junction of the anorectum, or dentate line. Internal hemorrhoids are lined with rectal mucosa and occur in three primary locations: right anterior, right posterior, and left lateral, although anatomic variability is common. The end branches of the superior and middle hemorrhoidal arteries terminate in the submucosa above the dentate line with an anterior and posterior branch on the right and a single lateral branch on the left, corresponding to the three primary hemorrhoid locations. The right posterior and left branches give off two end branches to potentially form secondary hemorrhoids. External hemorrhoids arise from the inferior hemorrhoidal venous plexus below the mucocutaneous junction and are lined by perianal squamous epithelium. The perianal squamous epithelium of the anus contains numerous pain receptors, so that thrombosis of external hemorrhoids causes significant pain. Internal and external hemorrhoidal plexuses freely communicate to drain the lower rectum and anus. Internal and external hemorrhoids drain into the inferior vena cava through the internal pudendal veins.

Pathogenesis

Elegant histological studies have shown that hemorrhoids are normal features of the human anatomy. They have three important parts: the lining (rectal mucosa or anoderm), the stroma (blood vessels, smooth muscle, supporting connecting tissue), and the anchoring connective tissue (which secures the hemorrhoids to the sphincter mechanism). Hemorrhoidal tissue has anatomic similarities to erectile tissue such as the corpora cavernosa. [5] With age or other aggravating factors, the anchoring and supporting connective tissue deteriorates, causing the hemorrhoids to bulge and descend, leading eventually to symptoms. [6] This theory is supported by the increased incidence of hemorrhoids in patients with chronic constipation, diarrhea, pregnancy, or pelvic tumors—conditions that increase pelvic venous pressure. In certain individuals, the internal sphincter becomes hypertrophic and the anal outlet becomes functionally narrowed. At straining, the fecal bolus acts as an obturator forcing the hemorrhoidal cushions to descend through the hypertrophic sphincter, enlarge, and become symptomatic.

Definitions

External skin tags are redundant folds of skin that arise from the anal verge. External hemorrhoids arise from the inferior hemorrhoidal plexus below the dentate line and are covered by anal squamous epithelium. Internal hemorrhoids arise from the superior hemorrhoidal plexus above the dentate line and are covered by columnar epithelium of the rectum. They may be classified according to their degree of protrusion or prolapse. First-degree hemorrhoids bulge into the lumen of the anorectal canal on anoscopy but do not protrude out of the anus. Second-degree hemorrhoids prolapse out of the anus with defecation or straining but reduce to a normal anatomic position spontaneously. Third-degree hemorrhoids prolapse out of the anus with defecation or straining and require digital reduction. Fourth-degree hemorrhoids are irreducible and are at risk for strangulation. The clinical presentation and recommended therapy for the various degrees of hemorrhoids are different and are discussed later. Anorectal varices are not hemorrhoids; they occur as a consequence of portal hypertension and are discussed separately.

External Skin Tags After thrombosis of an external hemorrhoid, the overlying skin becomes redundant, and this excess skin remains long after the underlying clot resolves. External skin tags may also arise after formal hemorrhoidectomy or de novo in cases of inflammatory bowel disease. Primary symptoms, if present, are complaints of a palpable growth near the anus and difficulty with anal hygiene. Skin tags are relatively easy to distinguish from more serious pathology such as anal cancer or condyloma acuminata by their gross appearance as normal skin and their soft, fleshy texture on palpation. Treatment is conservative whenever possible, and surgical excision is only necessary in cases of poor hygiene or patient anxiety.
External Hemorrhoids Thrombosis of an external hemorrhoid can be an extremely painful event. Distention of overlying perianal skin and inflammation associated with the process of thrombosis may cause severe patient discomfort. Bleeding usually occurs late in the course of thrombosed external hemorrhoids after the overlying perianal skin ulcerates and the resolving, liquefied hematoma necessitates. External hemorrhoids should be distinguished from strangulated internal hemorrhoids and anorectal varices. Strangulated internal hemorrhoids tend to be larger and more circumferential, encompassing the entire anus. Anorectal varices should be considered in any patient with a history of cirrhosis or portal hypertension. Many thrombosed external hemorrhoids can be treated with warm sitz baths two to three times per day. Stool softening agents such as psyllium seed preparations, synthetic mucilloids, and the sodium or calcium salts of dioctyl sulfosuccinate can minimize straining at stool and prevent aggravation of the pain and thrombosis. Topical therapy with anesthetic ointments and witch hazel–impregnated pads may provide additional relief. One must temper the attribution of clinical improvements to medical therapy with the knowledge that the natural history of thrombosed external hemorrhoids is resolution after 48 to 72 hours. If the pain is severe and the patient is seen within 48 hours of symptom onset, surgical evacuation or excision of the thrombosed external hemorrhoid should be performed. After the thrombus has organized, it cannot be evacuated. This can usually be done in the clinic setting and provides prompt relief.
Internal Hemorrhoids Internal hemorrhoids may be asymptomatic or associated with discomfort, pruritus ani, fecal soiling, or prolapse. Bleeding, however, is the typical complaint that brings the patient to the physician and hemorrhoids are the most common cause of rectal bleeding. Such bleeding is described as bright red spotting on the toilet tissue or dripping into the toilet bowl. It most often occurs at the end of defecation and is separate from the stool. It does not usually occur apart from defecation. Rarely, acute severe bleeding requires transfusion and, occasionally, ongoing chronic losses cause iron-deficiency anemia. [7] However, hemorrhoids should not be considered the source of hematochezia until other potential bleeding sources in the colon and rectum have been investigated. With the possible exception of a young patient with a bleeding pattern typical of hemorrhoids, a flexible sigmoidoscopy or, if clinically appropriate, a full colonoscopy should be performed. Occult bleeding should not be attributed to hemorrhoids. Occult blood in the stool deserves a complete evaluation regardless of the presence of hemorrhoids. Prolapse of the hemorrhoidal tissue is another common complaint. Prolapse may manifest itself anywhere along a continuum of symptoms from difficulty with anal hygiene to painful strangulation. Prolapsed tissue may also be a presenting symptom of rectal prolapse, rectal polyps, or rectal cancer. Sigmoidoscopic evaluation with biopsy of suspicious lesions should be carried out if appropriate. Anal condyloma and anal cancer are easily differentiated from prolapsing hemorrhoids, and thrombosed external hemorrhoids tend to cause pain as a presenting symptom. All degrees of internal hemorrhoids may be associated with mild discomfort, but only strangulated hemorrhoids cause significant pain. Strangulated hemorrhoids usually possess an external and internal component and occur secondary to prolapse with subsequent lack of blood supply. Progression to gangrene with resultant infection is life threatening and will occur if immediate surgical therapy is not instituted. In contrast to complete rectal prolapse, strangulated hemorrhoids lack concentric mucosal folds and are irreducible.

Treatment

Conservative Therapy Dietary counseling, behavior adjustment, and topical agents are effective for most first-and second-degree hemorrhoids. A high-fiber diet and adequate fluid intake should be prescribed to promote passage of soft, bulky stools to prevent straining. Some patients may require the addition of hydrophilic bulk-forming agents such as psyllium extracts or mucilloids. Excessive and prolonged defecatory straining should be discouraged. Many patients benefit from warm sitz baths twice daily and attention to proper anal hygiene. A number of proprietary agents such as suppositories, ointments, and medicated pads (witch hazel and glycerin) may provide topical astringent relief. Hydrocortisone or anesthetic-containing preparations (benzocaine, lidocaine, pramoxine, dibucaine) offer short-term relief of pruritus, burning, and soreness, although firm efficacy data are lacking. If conservative therapy does not suffice, a definitive procedure is necessary (Table 92-2). The available treatment modalities can be broadly classified into one of two categories: those that involve loss of redundant mucosa and those that do not. In general, first- and second-degree hemorrhoids can be treated without removal of redundant hemorrhoidal tissue. Third- and fourth-degree hemorrhoids usually require a form of therapy that results in both thrombosis of the hemorrhoid and removal of redundant tissue.

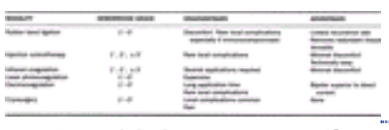

TABLE 92-2 Nonoperative Procedures to Treat Internal Hemorrhoids

Rubber Band Ligation Barron [8] first described a rapid, simple, effective device for the treatment of internal hemorrhoids in the office or outpatient setting. In a large office-based practice, 21,000 patients were treated and 44.8% required rubber band ligation. [9] Band ligation is associated with both thrombosis of the hemorrhoid and removal of redundant mucosal tissue. By inducing submucosal scarring, development of new hemorrhoidal tissue is prevented. Rubber band ligation is a good treatment for refractory first-degree hemorrhoids as well as all second- and selected third-degree hemorrhoids. Fourth-degree hemorrhoids are not well treated by this method. Because it is an outpatient procedure, no special preparation is needed for the majority of patients. After anorectal evaluation is performed, the anoscope is inserted and a hemorrhoidal cushion is selected for band application. No anesthetic is required if care is taken to place the band at least 0.5 cm above the dentate line. Some authors recommend banding of only one column per session to minimize tissue necrosis, but up to four ligations have been performed with acceptable morbidity. [10], [11] Rubber band ligation allows a controlled removal of tissue rivaled only by formal hemorrhoidectomy. Complications are rare but serious. In a series of

512 patients treated with rubber band ligation, 2.5% required hospitalization. [12] Migration of the band onto the anoderm is associated with excruciating pain requiring immediate removal of the band. Persistent severe pain, fever, urinary retention, or foul-smelling rectal drainage may herald the presence of a rectal infection. A number of case reports of necrotizing infection and death as a result of band ligation have appeared in the literature. [13], [14] Significant discomfort lasting for days occurs in some patients and mucosal slough with potential for bleeding occurs 5 to 7 days after band application. Results of rubber band ligation are generally excellent. Long-term patient satisfaction is about 90%. [15] No requirement for anesthesia and the ability to perform band ligation in the clinic setting continue to make this a popular treatment option for patients with hemorrhoids. Novel methods to perform banding have also been employed. A band ligator cap device attached to the tip of a flexible endoscope allows excellent visualization of band placement. [16] Inexpensive plastic single-handed ligators have been developed that employ suction to capture the hemorrhoidal tissue for band placement. [17], [18]

Injection Sclerotherapy The use of a sclerosant solution such as sodium morrhuate or 5% phenol is an accepted treatment for first- and second-degree hemorrhoids. The sclerosant is injected with a special hemorrhoidal needle into the submucosa around, but not into, symptomatic hemorrhoids. An intense inflammatory reaction results in fixation of the mucosa to the underlying muscle thus obliterating the submucosal layer where hemorrhoids form. Injection of sclerosants is a less controlled and therefore less popular technique than rubber band ligation. Known complications are mucosal slough, prostatic infection, contact hypersensitivity, and rectal infection. A refinement of injection sclerotherapy for hemorrhoids uses a flexible endoscope to inject 23.4% saline through a sclerotherapy needle into the hemorrhoidal cushion. In a small preliminary series of patients with grade I, II, or III hemorrhoids, success rates were similar to those seen with other, more conventional therapies. [19]

Cryosurgery Special cryoprobes activated by liquid nitrogen, carbon dioxide, or nitrous oxide have been developed. Local tissue destruction is caused by freezing and subsequent necrosis. All symptomatic hemorrhoids are treated in one session. If deep freezing of the submucosal hemorrhoidal cushions does not occur, symptoms persist. Tissue damage is uncontrolled, and wound healing is accompanied by prolonged anal drainage, late bleeding, and pain. [20] Compared with rubber band ligation, patient satisfaction is less and local complications more frequent with the cryosurgical technique. [21], [22]

Electrocoagulation Electrosurgical units have been adapted for use in the outpatient therapy of internal hemorrhoids. [23] Application of direct electrical current is less precise than with band ligation or formal hemorrhoidectomy and requires several minutes of contact time. Excess electrical current is grounded through the patient's body, and occasionally discharges occur that cause injury at sites distant from the area of intended tissue destruction. Bipolar electrocoagulation is as efficacious as the direct current technique and is performed more rapidly. [24], [25] It may be easier to perform and less painful than rubber band ligation. [26]

Photocoagulation Both infrared light and lasers have been used to treat symptomatic hemorrhoids. [27] Photocoagulation stimulates fibrosis of the submucosal layer by first causing tissue coagulation and necrosis. Advocates of photocoagulation point to the lack of electrical contact with the patient's body and a more controlled application of destructive force compared with electrofulgeration. [28] Infrared devices are hand-held and much less expensive than laser devices. Equipment for both forms of photocoagulation is more expensive than equipment for rubber band ligation or injection sclerotherapy. The incidence of discomfort and complications compare favorably with those of rubber band ligation and injection sclerotherapy. Rubber band ligation therapy produces more posttreatment pain than injection sclerotherapy or infrared photocoagulation, but fewer patients are likely to need re-treatment because of symptomatic recurrences. [29] Laser photocoagulation is an additional, albeit expensive, alternative. Using a carbon dioxide laser, 1816 consecutive patients were treated with success rates approaching those of more established therapies. [30]

Hemorrhoidectomy Fewer than 10% of symptomatic hemorrhoids require surgical hemorrhoidectomy. [9] It is the treatment of choice for most third-degree hemorrhoids, all fourth-degree hemorrhoids, strangulated hemorrhoids, and hemorrhoids that have persisted despite other forms of therapy. Advantages of the surgical approach include precise removal of all internal and external hemorrhoids, control of bleeding, and rapid wound healing. Disadvantages include the need for regional anesthesia, postoperative pain, risk of postoperative urinary retention, and expenses incurred from both hospitalization and lost time from work. A number of procedures have been developed, but all incorporate three basic principles: removal of all symptomatic diseased tissue, preservation of wide intervening skin bridges to prevent anal stenosis, and avoidance of damage to the anal sphincter mechanism. Some surgeons advocate primary closure of the entire hemorrhoidectomy wound, but others close the rectal mucosal wound only to the dentate line and leave the anoderm open to allow drainage. Using primary closure of the entire wound, Ferguson and colleagues [31] reported excellent results 5 years after hemorrhoidectomy in 95% of patients and a wound infection rate of 0.20%. In a more recent trial [32] however, the open technique led to faster and more reliable wound healing with a similar rate of pain and complications. Even with careful attention to perioperative fluid management, acute urinary retention requiring Foley catheterization may occur in 10% of patients. [20] The incidence of transient bacteremia may approach 8% even in uncomplicated cases. [33]

Ablation of the Internal Anal Sphincter Two strategies have been employed to decrease the abnormally high resting anal pressure found in some patients with hemorrhoids. Lord [34] advocated forceful anal dilation under general anesthesia, but the procedure causes variable and uncontrolled damage to the anal sphincters with resulting fecal incontinence. Good results occur in 80% of patients, but some of the efficacy of this procedure may reflect submucosal hemorrhage and subsequent scar formation. A more controlled disruption of the internal sphincter is achieved with lateral internal sphincterotomy (see section " Anal Fissure"). A metaanalysis of randomized controlled trials assessing at least two treatment modalities for symptomatic hemorrhoids found that rubber band ligation was superior to sclerotherapy and patients undergoing ligation were less likely to require future therapy than those treated with sclerotherapy or infrared coagulation. [35] Hemorrhoidectomy resulted in a better response than rubber band ligation, but the surgical group had significantly more pain and a higher incidence of complications. Based on these findings, it seems reasonable to suggest band ligation or a similar treatment as first-line therapy for grades 1 to 3 hemorrhoids, reserving hemorrhoidectomy for large grade 3 and grade 4 hemorrhoids and for those failing other techniques.

Anorectal Varices Meticulous histological study has shown that anorectal varices and hemorrhoids are unrelated. [5] Hemorrhoids are vascular cushions of ectatic venular-arteriolar connections of the hemorrhoidal plexus and they have no direct connection to the portal system. They occur independently of anorectal varices and the presence or degree of portal hypertension. [36], [37] Alternatively, rectal varices represent enlarged portal-systemic collaterals which correlate with the presence of portal hypertension. They develop as a result of hepatofugal portal venous flow through the inferior mesenteric vein to the superior hemorrhoidal veins. The varices represent the communication between these superior hemorrhoidal veins (portal circulation) and the middle and inferior hemorrhoidal veins (systemic circulation) which, in turn, flow into the femoral vein and then to the inferior vena cava. The distinction of anorectal varices from external hemorrhoids may be difficult. Often patients with anorectal varices have other known manifestations of portal hypertension such as esophageal varices and ascites. Anorectal varices are usually discrete, serpentine, submucosal veins. In contrast to external hemorrhoids, varices are compressible and refill rapidly. They extend from the squamous portion of the anal canal across the dentate line and into the rectum proper. Nearly 45% of patients with cirrhosis will be found to have anorectal varices by careful endoscopic examination and the prevalence increases to 75% using rectal endosonography. [36], [37] and [38] In one series, 5% of patients with bleeding as a manifestation of portal hypertension bled from anorectal varices. Bleeding may occur from either the anal or rectal portion of the varix and can be massive and life threatening. [39] The optimal management of anorectal varices is not known. Injection sclerotherapy, cryotherapy, rubber band ligation, and hemorrhoidectomy have all been associated with torrential, occasionally fatal bleeding. [40], [41] Treatment by underrunning the variceal columns with absorbable suture achieves primary control in a majority of cases and has a very low rate of morbidity. [39] Rubber band ligation has also been advocated but must be done in a controlled environment with full resuscitation capabilities. Inferior mesenteric vein embolization or ligation has been reported. [42] Ultimately, surgical or transjugular intrahepatic portosystemic shunting may be required. [43]

ANORECTAL ABSCESS AND FISTULA

Suppurative anorectal infection can be divided into two categories—anorectal abscess and anorectal fistula. Anorectal abscess may be defined as an undrained collection of perianal pus. Anorectal fistula is an abnormal communication between the anorectal canal and the perianal skin. Abscess is the acute manifestation and fistula the chronic manifestation of suppurative anorectal infection. In most cases, the underlying pathophysiology is thought to be the same and the treatment for each is essentially surgical.

Epidemiology, Etiology, and Differential Diagnosis

In a series of 1023 patients treated for anorectal abscess or fistula, the male-to-female ratio was 2:1. The age distribution was from 10 to 82 years, with the majority in the third and fourth decades of life. The most common associated medical diseases were hypertension, diabetes, heart disease, and inflammatory bowel disease. [44] The incidence of perirectal infection in patients with acute leukemia is approximately 8%. [45]

Current evidence suggests that infection of the anal glands is the most common cause of anorectal abscess and anorectal fistula. The most common bacterial isolates are *Escherichia coli, Enterococcus* species, and *Bacteroides fragilis*. [46] Histological specimens from patients with anorectal fistula revealed infected anal glands 70% to 90% of the time. Anal glands arise from the anal canal at the level of the crypts of Morgagni. At least half of the glands are observed to penetrate into the intersphincteric space. [47] Obstruction of the anal glands may occur in the presence of trauma, anal eroticism, diarrhea, hard stools, or foreign bodies, with resultant stasis and secondary infection. This cryptoglandular origin of anorectal abscess and fistula is further supported by the fact that the primary internal orifice is found at the level of the dentate line.

Specific diseases may cause anorectal abscess and anorectal fistula in the absence of primary cryptoglandular infection. These include Crohn's disease, anorectal

malignancy, tuberculosis, actinomycosis, lymphogranuloma venereum, radiation-induced proctitis, leukemia, and lymphoma. Other disease states that may cause a similar clinical picture and should be included in the differential diagnosis of suppurative anorectal conditions are infected presacral epidermal inclusion cysts, hidradenitis suppurativa, diverticulitis, pilonidal disease, and Bartholin abscesses.

Anatomic Considerations

Anorectal abscesses may be classified by anatomic site of origin and potential pathways of extension. Abscesses that are inferior to the puborectalis and levator ani muscles are classified as low intermuscular abscesses, and those that extend above these muscles are classified as high intermuscular abscesses. Low intermuscular abscesses are subclassified as perianal, submucosal, intersphincteric, or ischiorectal. High intermuscular abscesses are described as pelvirectal, retrorectal, or rectovesical. Proper surgical management is guided by correct anatomic identification of the type of anorectal abscess.

Anorectal fistulae are described based on pathogenesis of the disease (e.g., Crohn's disease, hidradenitis suppurativa) and classified according to the normal muscular anatomy of the pelvic floor. All anorectal fistulae are anatomically divided into one of four groups (Fig. 92-1). The most common type of anorectal fistula is the intersphincteric fistula, in which the fistula ramifies only in the areolar tissue between the internal and external anal sphincters. Transsphincteric fistulae pass from the intersphincteric plane through the external sphincter and into the ischiorectal fossa. Suprasphincteric fistulae pass upward in the intersphincteric plane, over the puborectalis muscle, and into the ischiorectal fossa. Extrasphincteric fistulae pass from the perianal skin through the ischiorectal fat and levator muscles into the rectum. Division of the entire tract of either suprasphincteric or extrasphincteric fistulae results in total division of the muscles of continence. [48] Anal endosonography and magnetic resonance imaging (MRI) have emerged as useful modalities for determining the position of an anorectal abscess or fistula relative to the anal sphincter complex. [49], [50] MRI has been shown to predict clinical outcome based on the initial severity of fistulous disease better than surgical exploration. [51]

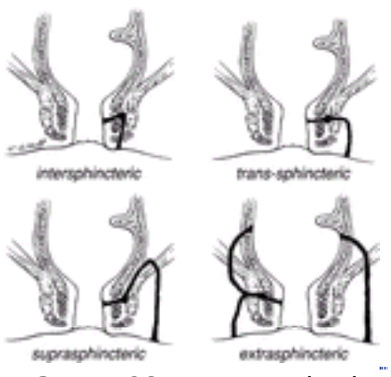

FIGURE 92-1. Anatomic classification of anorectal fistulae.

Clinical Manifestations

Acute pain and swelling are the most common complaints in the patient with anorectal abscess. Pain may occur in the absence of swelling, especially with small intersphincteric or pelvirectal abscesses. Sitting, movement, and defecation exacerbate pain. Antecedent history may reveal a bout of constipation, diarrhea, or minor trauma. Constitutional symptoms include malaise and fever. The presence of foul-smelling drainage means that the abscess has necessitated, or is discharging through the primary anal orifice.

Inspection of the perineum of a patient with perirectal abscess reveals the cardinal signs of inflammation: redness, heat, swelling, and tenderness. Drainage may be observed from the infected crypt orifice. An intersphincteric abscess may manifest only as localized tenderness. Rectal examination is often difficult because of pain, so evaluation under anesthesia is indicated if a complete examination is otherwise impossible. Delay in making the diagnosis of anorectal abscess leads to extension of the infection into previously uninfected spaces and also increases the subsequent risk of overwhelming sepsis.

Chronic, purulent drainage is the chief complaint of patients with anorectal fistula. A history of prior anorectal abscess is frequently elicited. Discomfort or pain often occurs with defecation but is not as severe as that associated with anal fissure or anorectal abscess. The perianal skin may be pruritic or excoriated. Bleeding is usually minor and caused by granulation tissue at the orifice of the primary or secondary anal orifice.

Anorectal fistulae can usually be diagnosed by the presence of a red, granular papule from which pus is expressed. The primary orifice is the fistulous opening at the level of the dentate line, which is thought to be the original site of the infected anal gland. The secondary orifice is the fistulous opening anywhere else on the perineum. Multiple secondary openings should alert one to the possibility of either Crohn's disease or hidradenitis suppurativa. It is often possible to palpate a fistulous tract as a firm cord just beneath the perianal skin. Attempts to pass metal probes are best made in the operating room. Anoscopy and sigmoidoscopy are performed to identify the primary orifice and to determine the presence of proctocolitis.

Patients who are neutropenic as a result of hematologic malignancy are particularly susceptible to serious anorectal infection. [52] Mortality rates from anorectal infection in patients with acute leukemia, if untreated, may be more than 45%. [46] Early diagnosis and aggressive surgical drainage may be life-saving in these individuals, with improvement in mortality rates to below 10%. [45] Point tenderness and poorly demarcated induration are the most frequent findings. Frequent reexamination may allow detection of an abscess if initial findings are equivocal. Because of the profound granulocytopenia, fluctuant masses are not usually seen. Necrosis and tissue breakdown may proceed quite rapidly, with extension into the genitalia and pelvis. If spontaneous drainage has already occurred, pain rapidly subsides and further surgical drainage may be postponed. *Pseudomonas aeruginosa* is a common wound isolate, and appropriate perioperative intravenous antibiotics should be administered if it is found. [53]

Treatment

Because of the risk of extension of pus into adjacent spaces and the potential for the development of necrotizing anorectal infection, the treatment of anorectal abscess is a surgical emergency. In one series, the time interval from onset of primary anorectal abscess to necrotizing anorectal infection was from 0.5 to 5 days, with a mortality in excess of 50%. [54] If one chooses to temporize waiting for an abscess to point or become ripe, it must be with the realization that the risks of necrotizing infection and extension into previously uninfected spaces rise dramatically.

In healthy patients with superficial abscesses in the perineal or ischiorectal location, drainage can be performed in the outpatient setting with local anesthesia. All other abscesses should be drained in the operating room with adequate anesthesia, lighting, and surgical instrumentation. If a primary fistula tract is identified and is not thought to encompass a large proportion of the sphincter mechanism, fistulotomy may be done in selected cases. In general, the patient should be counseled as to the possibility of persistent drainage from a retained or unidentified fistula which may require a second operation. [55] An abscess may also recur if the underlying fistula has not been definitively treated.

Antibiotics are usually not necessary, and they may temporarily mask the underlying suppurative infection and delay surgical therapy. Only in occasional cases does a perianal cellulitis resolve with antibiotic therapy alone. In otherwise healthy individuals with minimal infection of the surrounding tissues, incision and drainage of the abscess is all that is required. Patients with significant underlying disease, such as diabetes, acute leukemia, valvular heart disease, or extensive soft tissue infection, benefit from perioperative antibiotics. The choice of antibiotics should be directed by the clinical situation and culture results but, in general, both gram-negative aerobic and anaerobic bacteria should be covered. Ticarcillin/clavulanate, piperacillin/tazobactam, or a broad-spectrum cephalosporin are recommended for empiric coverage, but a particular clinical situation may mandate antibiotics with specific anaerobic, enterococcal, or pseudomonal coverage. The most common wound isolates are polymicrobial with *Escherichia coli, Proteus vulgaris, Bacteroides* species, streptococci, and staphylococci predominating. [56] A high proportion of necrotizing anorectal infections contain *Clostridium* species. [54] Immunosuppressed patients often demonstrate *Pseudomonas aeruginosa*. [45]

Postoperative management consists of frequent wound inspection, warm sitz baths, attention to stool consistency, and judicious analgesia. The wound should be

observed to heal from the base up so that skin bridges do not form, allowing the abscess to recur. If persistent drainage occurs, a second operation for fistulotomy is necessary. Warm baths improve hygiene and offer some symptomatic relief. Narcotic analgesics and perirectal pain both predispose to constipation, so a high-fiber diet, bulk-forming agents, or laxatives should be prescribed.

The presence of an anorectal fistula is an indication for operation. The operative approach depends on the location of the fistulous tract in relation to the sphincteric mechanism. Anorectal manometry has been used to improve the clinical and functional results of surgery for fistula-in-ano. [57] The basic prerequisites for successful therapy of a fistula include removal of the primary orifice, identification and opening of the entire extent of the fistula, and conservation of as much external sphincter as possible. Postoperative care is similar to that for anorectal abscess. In a series of 624 patients undergoing anal fistula surgery the fistula recurred in 8%, and 45% complained of some degree of postoperative incontinence. [58] A novel fibrin sealant has been touted as a less invasive method to heal fistulae. Further refinement is needed to improve the healing rates and lessen the chance of recurrences. [59]

Anorectal disease as a manifestation of Crohn's disease requires special consideration. In addition to standard therapy with 5-aminosalicylate derivatives and immunosuppressives, the use of metronidazole or ciprofloxacin is modestly beneficial in the healing of perineal Crohn's disease. Unfortunately, the required long-term therapy with metronidazole is associated with several side effects, most notably paresthesias. Also, discontinuation of therapy is often associated with flaring of disease. [60] Conservative surgical techniques are usually adequate to drain abscesses, reduce inflammation, and provide relief of symptoms. [61] In one study, proctectomy—once widely practiced—was necessary in only 12% of patients with complicated perianal Crohn's disease. [63] Surgical diversion of the fecal stream, usually in conjunction with resection of diseased intestine or a local anorectal procedure, may be necessary for perianal disease. [62] Infliximab, a chimeric monoclonal antibody to tumor necrosis factor, has emerged as an agent with impressive effectiveness for persistent Crohn's perianal fistulous disease. A controlled trial using 5 mg/kg at 0, 2, and 6 weeks improved (68%) and healed (55%) of patients compared to those receiving placebo (26% and 13%, respectively). [63]

RECTAL PROLAPSE

Rectal prolapse is simply protrusion of the rectum through the anal orifice. Complete rectal prolapse, or procidentia, is the classic situation in which all layers of the rectum visibly descend through the anus. Occult rectal prolapse refers to internal intussusception of rectal tissue without visible protrusion at the anus. Mucosal prolapse is a common condition in which only distal rectal tissues and not the entire rectal circumference protrude through the anus.

Rectal prolapse in children is an uncommon problem usually seen in infancy. It may be idiopathic, associated with congenital defects, such as spina bifida or myelomeningocele, or associated with cystic fibrosis. Prolapse occurs with defecation and usually reduces spontaneously. Treatment is conservative, and the condition is often self-limited.

Rectal prolapse in adults occurs at least 3 to 10 times more often in women than men and is not associated with multiparity. Men may develop prolapse at any age, but in women, the peak incidence is in the sixth and seventh decades. It is associated with poor tone of the pelvic musculature, chronic straining at stool, fecal incontinence, and, sometimes, neurological disease or traumatic damage to the pelvis.

Pathogenesis

Anatomic defects that have been described with rectal prolapse include a weakened endopelvic fascia and diastasis of the levator ani, loss of the normal horizontal rectal position in the sacrum, an abnormally deep pouch of Douglas, a redundant rectosigmoid colon, and a weak anal sphincter. [64] Most authors support the view that prolapse is caused by the intussuscepting rectum and that most of the anatomic defects described occur secondarily. Weakening of the fascial attachments of the rectum to the presacral fascia allows lengthening of the rectosigmoid and its mesentery. The normal positioning of the rectum in the sacral hollow is lost. The subsequent vertical orientation of the rectum enhances the ability of the rectum to intussuscept. Chronic straining in a misguided attempt to evacuate the internally prolapsing rectal tissue only serves to exacerbate the problem. Signs of pelvic neuropathy and anal sphincter dysfunction are common in patients with rectal prolapse. Many patients complain of partial or major fecal incontinence. On manometric evaluation, incontinent patients have low basal and voluntary contraction pressures. [65] Denervation of striated musculature on electromyogram, perineal descent, and absence of the anocutaneous reflex are also common findings. The presence of disturbed sphincter function and pelvic denervation may explain the disappointingly high incidence of persistent incontinence after surgical correction of the prolapse. Manometric findings associated with a higher risk of postoperative fecal incontinence include a resting anal pressure of less than 10 mm Hg and a maximal voluntary contraction pressure of less than 50 mm Hg. [66]

Clinical Manifestations

Patients complain of prolapse of tissue with defecation. As the condition progresses, rectal prolapse may occur with straining or even upright posture alone. Common accompanying symptoms include straining at stool, the sensation of incomplete evacuation, tenesmus, and fecal incontinence. Protrusion of the rectum through the anus is a striking clinical sign. Complete rectal prolapse is signaled by the presence of red concentric mucosal folds with a palpable double thickness to the rectal wall tissue. The protruding rectum may extend many centimeters, and usually the lumen tip points slightly posteriorly. The patient is asked to sit and strain to produce prolapse if it is not immediately obvious. Endoscopic or barium examination is performed on all patients with rectal prolapse to exclude tumors and mucosal lesions. Sigmoidoscopy may reveal changes consistent with solitary rectal ulcer. A voiding defecogram is the best way to identify occult prolapse (internal intussusception).

Complete rectal prolapse must be differentiated from mucosal prolapse, prolapsing internal hemorrhoids, anorectal varices, anal polyps, benign and malignant anorectal tumors, and hypertrophic anal papillae. Mucosal prolapse is characterized by a short segment of mucosa with disordered or radially arranged (not concentric) mucosal folds. Internal hemorrhoids usually have a varicose appearance and are separated into discrete cushions.

Every attempt should be made to manually reduce a persistently prolapsed rectum to avoid potential complications such as strangulation, ulceration, bleeding, and perforation. Some form of intravenous sedation may facilitate manual reduction. Placing granulated sugar on the prolapsed mucosa often eliminates edema and allows reduction. [67] Gangrene of the anterior rectal wall, a surgical emergency, is occasionally associated with evisceration of small bowel onto the perineum.

Treatment

Mucosal prolapse may be treated with procedures designed to remove redundant tissue and induce local fibrosis (see previous discussion of treatment of internal hemorrhoids). There is controversy over the appropriate treatment for occult rectal prolapse. To oversimplify, occult prolapse may be best treated surgically if fecal incontinence or chronic solitary rectal ulcer is present and conservatively if defecation difficulties or lesser symptoms predominate. [68] One study [69] suggests that sclerotherapy is successful if there is only a small associated rectocele and short perineal descent but that transanal excision of prolapsing mucosa is necessary if the rectocele and perineal descent are more prominent.

To avoid complications and ongoing damage to the pelvic floor and sphincter muscles, complete rectal prolapse should be surgically corrected. Perineal muscle exercises and buttock strapping offer palliation in the patient who refuses or is unable to undergo surgery. There are many surgical options advocated for the treatment of rectal prolapse, but they can be simply summarized as follows. Management in healthy patients involves replacement of the rectum into the sacral hollow with or without resection of redundant rectosigmoid colon. The two intra-abdominal operations that have been popularized in the United States are the anterior sling rectopexy (Ripstein procedure) and abdominal proctopexy with or without sigmoid resection.

Ripstein operation involves mobilization of the rectum to the tip of the coccyx and attachment of the rectum to the presacral fascia by means of a band of nonabsorbable plastic such as Teflon or Marlex mesh. Abdominal proctopexy as a sole procedure can be performed with very acceptable recurrence rates and function. Constipation is common in patients who are continent, and continence is not assured in others. Internal anal sphincter pressures and continence usually improve after rectopexy but not to normal values. [70] A laparoscopic approach has been used to further decrease operative morbidity. [71]

Abdominal proctopexy and sigmoid resection as a combined procedure eliminates two of the theorized causes of rectal prolapse by fixing the rectum directly to the sacral hollow by means of nonabsorbable sutures and removing the redundant sigmoid colon. Prosthetic materials, such as Marlex, are generally not placed in the peritoneal cavity if sigmoid resection is performed because of the risk of contamination of the mesh and resultant sepsis. In a series of 102 patients with abdominal proctopexy and sigmoid resection, 80% had good to excellent results, no mortality, and improved morbidity, compared with those treated with the Ripstein procedure. [72] Abdominal proctopexy and sigmoid resection is physiologically the most demanding procedure and should be used only in patients in good general physical

condition. These procedures have continued fecal incontinence as the major postoperative complaint. Patients who continue to have incontinence 6 to 12 months after definitive correction of the rectal prolapse often benefit from a Parks postanal repair or a plication sphincteroplasty. [73], [74] A systematic review suggested that residual fecal incontinence was less common after abdominal (vs. perineal) approaches and that division (vs. preservation) of the lateral ligaments during rectopexy was associated with less recurrent prolapse but at the price of more constipation. [75]

In the elderly or debilitated patient, a perineal or extra-abdominal approach is associated with acceptable morbidity and mortality rates. An elegant study of perineal rectosigmoidectomy for complete prolapse in 114 elderly patients demonstrated a recurrence rate of 10% with no mortality and good functional results. [76] A diverting colostomy may be also be an appropriate alternative for this group of high-risk patients.

ANAL FISSURE

Anal fissure is a painful linear ulcer in the anal canal. Primary anal fissures are usually found in young and middle-aged adults and occur equally in males and females. Primary fissures are located in the posterior midline more than 90% of the time. The remainder are found in the anterior midline. Fissures may occur secondary to an underlying disease such as inflammatory bowel disease (especially Crohn's disease), proctitis, leukemia, carcinoma, and, rarely, syphilis or tuberculosis. These lesions, in contrast to primary fissures, are usually found in a more lateral position.

Etiology

The elliptical arrangement of the anal sphincter fibers offers less muscular support to the anal canal posteriorly. This deficient support predisposes the posterior anal canal to traumatic tears during passage of a large, hard stool. Fissures may become chronic because of high resting anal sphincter tone and repeated trauma during passage of fecal boluses. Rectal distention normally causes a transient internal anal sphincter relaxation. Patients with anal fissure have an abnormal overshoot contraction following the normal relaxation. The overshoot contraction may explain the reflex spasm and pain seen after defecation. This phenomenon disappears after successful treatment of the fissure. [77] Preoperative maximal resting pressure and maximal contraction pressure are also elevated in patients with fissure. [78], [79] A histopathological study documented the presence of fibrosis throughout the anal sphincter in patients with anal fissure. [80] Recent studies have emphasized the potential role for ischemia in fissure disease. Vascular perfusion of the anoderm is lower in the posterior midline than in other locations and measurements performed are particularly low in patients with anal fissure. Measurements before and after lateral sphincterotomy in patients with chronic anal fissure demonstrated the inverse relationship between anal sphincter pressures and posterior anoderm perfusion pressures. [81], [82]

Clinical Manifestations

Severe pain associated with scant, bright red rectal bleeding is the hallmark of anal fissure. The pain occurs during and after defecation ("like passing a piece of glass") and usually seems out of proportion to the clinical findings. Severe pain may make anoscopy impossible, and even digital examination is difficult without topical anesthesia. The fissure is best identified by simple inspection after spreading the buttocks. Acute fissures are small, linear tears oriented perpendicular to the dentate line in the posterior midline. Fissures located in a lateral position should prompt a search for a secondary etiology. If an acute anal fissure does not heal promptly, certain characteristic secondary features develop. The classic triad of chronic anal fissure includes the fissure, a proximal hypertrophic papilla, and a sentinel pile or fibrotic nubbin of skin found at the anal verge (Fig. 92-2).

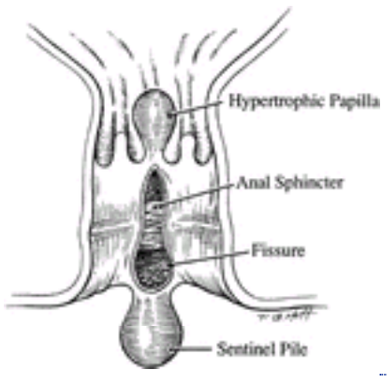

FIGURE 92-2. The classic triad of chronic anal fissure: hypertrophic papilla, anal fissure, and sentinel pile.

Treatment

Prescribing a high-fiber diet and adequate fluid intake should soften stools. Hydrophilic bulk agents or salts of dioctyl sulfosuccinate are important aspects of therapy because many fissures are precipitated by the traumatic passage of hard, dry stools. The temporary use of topical anesthetic preparations containing agents such as benzocaine or pramoxine hydrochloride provides symptomatic relief. The use of these medications, in addition to warm sitz baths two to three times daily, acts to decrease sphincter spasm to provide additional relief. [83] A combination of bran supplements and sitz baths was shown to be superior to a topical anesthetic or hydrocortisone cream with respect to symptoms and healing. [84] With such a conservative regimen, most acute fissures heal in 4 to 6 weeks. Occasionally, patient anxiety, severe pain, or other considerations may rule out such a prolonged trial of conservative therapy.

Chronic fissure usually requires some form of surgical therapy to reduce internal sphincter tone. Reduced sphincter tone permits easier passage of the fecal bolus through the anal canal. Repetitive injury is avoided so that the traumatic tear may finally heal. The beneficial effects of sphincterotomy on internal sphincter spasm and healing of anal fissure have been well documented, and surgical cure rates on the order of 95% can be expected. [77] Midline sphincterotomy with fissurectomy also offers definitive therapy for chronic anal fissure. However, in a retrospective study of 300 patients, a higher rate of postoperative complications was seen compared with the lateral sphincterotomy. [85] Also, an unfortunate complication of posterior midline sphincterotomy and fissurectomy is development of a residual keyhole deformity of the anus, which predisposes to long-term leakage of feces and mucus. Another approach to treating sphincter spasm is manual dilation. The anus is dilated to six fingers under regional anesthesia. A metaanalysis of operative techniques concluded that internal anal sphincterotomy was superior to manual dilation for both fissure healing and the occurrence of incontinence to flatus. [86]

Chemical methods may also be used to lower sphincter pressures to allow healing. Topically applied nitroglycerin heals fissures by reducing maximum resting pressure and by increasing anodermal blood flow. [87] Several open and controlled trials [88], [89] have demonstrated the efficacy of 0.2% nitroglycerin ointment although a large, controlled trial [90] failed to confirm this. Headaches are a frequent adverse effect of even a pea-sized application of nitroglycerin. For this reason, other topical smooth muscle relaxants have been investigated. A controlled trial of nearly 300 patients found that nifedipine gel healed 90% of fissures at 3 weeks and more recently, diltiazem ointment healed 75% of patients after 2 to 3 months of therapy. [91], [92] Headaches were not seen. Botulinum toxin injected into the sphincter muscles directly through the perianal skin heals the majority of fissures (>80%), a finding confirmed in a controlled trial (73% vs. 13% healing at 2 months). [93] Temporary mild fecal incontinence appears to be rare. In a direct comparison, botulinum toxin healed 96% of fissures versus 60% using 0.2% nitroglycerin ointment. [94] A randomized trial found surgical sphincterotomy to be more effective than nitroglycerin. [95] However, sphincterotomy may cause fecal incontinence in approximately 8% of patients. [96] A reasonable, but not yet evaluated, strategy to treat chronic anal fissure disease is to use a topical muscle relaxant or botulinum toxin initially and reserve surgical sphincterotomy for those who fail to respond.

ANAL STENOSIS

Anal stenosis is an abnormal narrowing of the anal canal associated with a variety of underlying diseases. Patients may complain of narrow stools, painful or resistant defecation, and bleeding from recurrent anal tears. The clinician is typically unable to perform a digital rectal examination. Malignant causes of anal stenosis include anal and rectal cancer and, less commonly, transmural invasion of the anorectum by a urogenital malignancy. Common causes of benign anal stenosis are prior rectal

surgery, trauma, and inflammatory bowel disease. Postsurgical stenosis of the anal canal is most often a result of injudicious excision of anal skin during hemorrhoidectomy. [97] Laxative abuse and chronic diarrhea are also associated with anal stenosis because lack of a solid fecal bolus to intermittently dilate the anus causes hypertrophy and narrowing of the anal sphincters. [98] Other causes of anal stenosis include radiation injury, tuberculosis, actinomycosis, lymphogranuloma, and congenital abnormalities.

Treatment is based on the severity and location of the anal stenosis. Mild strictures respond to periodic dilation and a high-fiber diet. Severe stenosis usually requires some form of anoplasty to increase the amount of perianal skin. [99] Lateral internal sphincterotomy is often used in conjunction with anoplasty. Cure rates in excess of 90% are usually seen. [97]

SOLITARY RECTAL ULCER

Solitary rectal ulcer is a chronic benign disorder related to abnormal defecation. The usual presenting symptoms are the passage of mucus and blood, altered bowel habits, anorectal pain, a feeling of incomplete evacuation, and straining at defecation. Classically at sigmoidoscopy, a shallow, discrete, 1-cm punched-out ulcer with a hyperemic margin is seen 7 to 10 cm from the anal verge on the anterior wall. However, ulcers range from 0.5 to 4 cm in diameter, are located a few centimeters to 13 cm from the anus, and are occasionally found on the posterior wall. Furthermore, there may be multiple ulcers or no ulcer at all. Sometimes only a localized erythematous or nodular area of mucosa is noted. Initial misdiagnosis is common and the lesion may be mistaken for inflammatory bowel disease or neoplasia. [100] Characteristic histological changes seen on biopsies taken from abnormal mucosa or ulcer margins include fibrous obliteration of the lamina propria, disorientation and thickening of the muscularis mucosae and regenerative changes in the crypt epithelium. [101]

Pathogenesis

The pathogenesis of solitary rectal ulcer is related to prolonged straining at stool and difficulty initiating defecation. Digital evacuation is frequently practiced in these patients, but trauma caused by self-digitation probably plays a minor role. Although clinically apparent rectal prolapse through the anal orifice is only occasionally seen with solitary rectal ulcer, careful observation during sigmoidoscopy or defecography demonstrates subtle forms of mucosal prolapse and delayed evacuation during defecation in 90% of these patients. [102], [103] However, patients with solitary rectal ulcer syndrome have higher sphincter pressures and thicker rectal walls (primarily muscularis propria) than patients with complete rectal prolapse. [104], [105] Womack and colleagues [102] have shown that rectal prolapse coupled with high transmural pressures during defecation is probably responsible for the mucosal trauma that causes ulceration. The anterior wall mucosa, 7 to 10 cm proximal to the anal margin, is often the first and largest part of the intussusceptum as it descends downward into the anal canal. This corresponds to the usual location of ulceration seen clinically.

Treatment

Complications of massive bleeding, rectal stricture, and even ulcer penetration into the prostate gland have been reported but are so rare that the patient should be reassured as to the benignity of the syndrome. [106], [107] Because solitary rectal ulcer is caused by repeated mechanical trauma of the prolapsing mucosa during defecation, it is appropriate to begin treatment by eliminating straining and improving bowel habits. Beneficial results should not be expected for months, and relapses are common. [108] Sulfasalazine, antibiotics, and enemas of steroids or 5-aminosalicylic acid are of no use, but sucralfate retention enemas may be beneficial. [109] Symptoms refractory to conservative management may respond to surgical rectopexy. [110], [111] Biofeedback therapy may be helpful as a primary therapy and may also decrease postsurgical symptomatic recurrences. [100], [112]

FECAL INCONTINENCE

Fecal incontinence is the release of rectal contents against one's wishes. It is perhaps the most embarrassing and socially disabling of all gastrointestinal problems. Prevalence rates vary, but it is clear that the elderly and institutionalized persons are particularly victimized. More than 16% of elderly persons living in residential homes are incontinent of stool at least twice a month, and 10% are incontinent on at least a weekly basis. [113] Risk factors in the elderly include a history of urinary incontinence, neurological disease, poor mobility, cognitive decline, and age older than 70 years. [114] Random surveys of the general population identify 18% who admit to at least one episode of fecal incontinence and 4% with symptoms more than once a week. [115], [116] The complaint of fecal incontinence is not readily volunteered because of the social stigma attached to it, and instead the initial complaint may be "diarrhea." [117] Clinically, incontinence must be placed in the context of surrounding circumstances. The frequency of fecal soiling in an individual with urgency may simply reflect availability of a toilet. Fecal soiling in a mentally disabled or demented person might relate directly to a communication or recognition problem.

Pathophysiology

Anal Canal and Sphincters The anal canal is 3 to 4 cm long and represents the distal outlet of the gut. It normally remains closed as an anteroposterior slit by tonic contractions of the surrounding musculature. Spongy vascular tissues within the canal are distensible and play a minor, fine-tuning role in fecal continence by assisting with anal closure. Minor degrees of seepage after hemorrhoidectomy may be caused by partial excision of this tissue. The circular smooth muscle layer of the rectum terminates in the thickened, rounded internal sphincter muscle. The internal sphincter is under autonomic control and is responsible for the majority of resting anal canal tone. The external sphincter is a surrounding sleeve of striated muscle that blends into the puborectalis and levator ani muscles proximally and extends to just below the internal sphincter margin distally. It is a voluntary muscle with somatic innervation from the pudendal nerve (sacral branches S2, S3, and S4). Although it is a striated muscle, the external sphincter maintains a degree of tonic neural activation and thus contributes to resting anal tone (approximately 30%). However, it assumes a major role in maintaining continence by contracting in a reflex manner during times of sudden rectal distention or increased abdominal pressure (i.e., coughing, lifting). Voluntary contraction of the external sphincter approximately doubles the normal resting tone of the anal canal. Traumatic obstetrical and surgical injuries, prolapsing rectal and hemorrhoidal tissues, and various neuropathic diseases are examples of processes involving the anal canal and sphincters that may cause incontinence.
Puborectalis Muscle The puborectalis muscle attaches to the symphysis pubis and wraps around the posterior aspect of the anorectal junction effectively forming a sling. Tonic contraction of this muscle produces an acute angle of approximately 80 degrees between the axes of the rectum and anal canal, forming an important mechanical kink or barrier to the passage of stool. The action of the puborectalis may also contribute to an as yet unproven flap-valve effect that produces an occlusion of the upper anal canal by the anterior rectal mucosa. [118] Sudden increases in intra-abdominal pressure accentuate the anorectal angle to reduce transmission of pressure into the anal canal so that, in concert with the external sphincter, continence is maintained. The anorectal angle formed by the puborectalis muscle is arguably the most important structure in the preservation of gross fecal continence. [119], [120] Any traumatic or neuropathic injury to this muscle causing persistent straightening of the anorectal angle leads to major fecal incontinence.
Anal Sensation Although the rectum itself is sensitive only to stretch, the anal canal epithelium is endowed with a rich network of nerve endings sensitive to touch, pain, and temperature. Together, these sensory inputs make up an important part of so-called anal sampling, which allows differentiation between solid, liquid, and gas contents in the rectum. After material enters and distends the rectum, a transient reflex relaxation of the internal sphincter occurs (the rectoanal inhibitory reflex) to permit the rectal contents to come into contact with the sensitive anal canal mucosa. Using tactile and perhaps thermal properties, anal sampling differentiates flatus from solid or liquid stool to allow selective passage of these materials. [121] Simultaneously, the external sphincter reflexively contracts to maintain continence until voluntary contractions consciously take over. Within a short time, rectal accommodation to the new volume deactivates stretch receptors, and the sensation of urgency dissipates.
Rectum The motility, compliance, and sensation of the rectum are important features of fecal continence. Baseline rectal contractions are of higher frequency and amplitude than are those in the sigmoid colon. This reverse peristalsis acts as a subtle barrier to fecal flow. The rectum has the ability to accommodate different fecal volumes presented to it. This reservoir function maintains continence and allows for appropriate timing of defecation. In a similar manner, the sampling reflex and a normal call to stool depend on intact rectal sensation. Neuropathies caused by diabetes and other diseases impair rectal sensation and therefore interfere with continence. [122] Poor distensibility of the rectum in ulcerative colitis, radiation proctitis, and chronic rectal ischemia may lead to fecal urgency and incontinence. [123] Similar symptoms in those with irritable bowel syndrome are probably caused by hypersensitivity to distention and abnormal motility patterns in the rectum.
Stool Volume and Consistency Nearly everyone has experienced fecal urgency and perhaps incontinence during an acute diarrheal illness. Large volumes of liquid stool pose a special challenge to even healthy mechanisms of continence. Some persons are incontinent of stool only if stressed with a liquid bolus; others are continent of stool but cannot retain flatus. Any disease that causes diarrhea, especially if it is associated with large volumes and rapid transit, may produce fecal incontinence.
Defecation The process of normal defecation depends on intact mechanisms of continence. The sequence of events leading to defecation is initiated when material from the sigmoid colon enters the rectum. Rectal distention causes reflex relaxation of the internal anal sphincter, exposing the rectal contents to the anal mucosa so

that sampling may occur. External sphincter contraction maintains continence until the rectum can accommodate to the bolus. After feces fills the rectum to a sufficient degree, the urge to defecate is experienced. If a conscious decision to initiate defecation is made, a squatting or sitting position is assumed. This position straightens the anorectal angle in preparation for passage of stool. If the urge is strong and the individual is exerting conscious resistance to passage of stool, simply relaxing voluntary control allows defecation to proceed. Otherwise, a Valsalva maneuver is performed to increase intra-abdominal pressure and push the feces toward the anal canal. The pelvic floor descends, the rectum contracts, external sphincter activity is inhibited, and the fecal bolus is expelled out of the rectum. Stool is passed piecemeal with episodic straining or, alternatively, propulsive contractions may empty portions of the left colon in a single motion.

Clinical Manifestations

It is clinically useful to categorize fecal incontinence as either partial or major. [115] Partial incontinence is defined as minor soiling of loose stools and poor control of flatus. The elderly and those with internal sphincter deficiencies and prolapsing hemorrhoidal or rectal tissues are particularly prone to minor soiling. Fecal impaction also predisposes to partial incontinence. A rectum chronically distended with feces continuously induces internal sphincter relaxation, allowing leakage of stool in those with weakened sphincter mechanisms. Major incontinence refers to frequent loss of control of even solid stools. Neurological diseases, traumatic injuries, and surgical damage to the puborectalis or sphincter muscles may cause major incontinence. One may also consider the category of “leakers” characterized by the loss of small amounts of liquid or solid stool smears. These patients typically remain continent of flatus and stool and have near normal sphincter pressure and innervation. Many, but not all, have identifiable pathology such as hemorrhoids or fistulae on physical examination. [124] A comprehensive list of the causes of fecal incontinence is provided in Table 92-3. Many incontinent patients have no obvious predisposing history of major trauma, anorectal disease, or neurological disease. Careful studies using electromyography and other neurophysiological techniques have identified striated muscle denervation damage in most of these patients with idiopathic incontinence. [125] Perineal descent associated with chronic straining at stool may cause stretch injury and subsequent denervation of the pudendal nerve. [126], [127] Pudendal nerve injury and occult sphincter disruption commonly occur during vaginal delivery of childbirth. [128] Approximately 30% of women will have occult sphincter defects detectable by endosonography after their first vaginal delivery and more than 10% will complain of urgency or incontinence. [129] Multiparity, high birth weight, forceps delivery, and third-degree perineal tears are important risk factors. It is not surprising that more than one mechanism of continence may be disturbed in those with neuropathic incontinence. [130] Careful testing has found that motor neuropathy of the external sphincter and pelvic floor is associated with damage to the internal sphincter and a sensory deficit of the anal canal. [131]

Diarrhea
Fecal impaction
Irritable bowel syndrome
Anal pathology
Anal carcinoma
Congenital abnormalities
Protruding internal hemorrhoids
Rectal prolapse
Perianal infections
Fistula
Injury
Surgical (hemorrhoidectomy, fistulotomy, etc.)
Obstetrical
Accidental trauma
Rectal pathology
Rectal carcinoma
Rectal ischemia
Proctitis
Inflammatory bowel disease
Radiation induced
Infectious
Neurological diseases
Central nervous system
Stroke, dementia
Toxic/metabolic
Cord injury, tumors
Multiple sclerosis, tabes dorsalis
Peripheral nervous system
Diabetes, others
Cauda equina lesions
“Idiopathic” (primary neurogenic)
Childbirth injury
Chronic constipation
Descending perineum
Old age

TABLE 92-3 Causes of Fecal Incontinence

An important cause of fecal incontinence not associated with pathology of the pelvic nerves or sphincter muscles is irritable bowel syndrome. The incontinence of irritable bowel syndrome commonly occurs in those with diarrhea-predominant symptoms associated with fecal urgency and cramping relieved by defecation. Physiological or emotional stressors lead to rapid delivery of loose stools and flatus into a hypersensitive rectum. This produces occasional soiling even in the individual with an intact sphincter mechanism.

Clinical Evaluation After the existence of incontinence has been established, the clinical evaluation begins with an assessment as to the frequency, severity, and circumstances surrounding the incontinent episodes. Does the patient suffer from partial or major incontinence? How acute are the symptoms? Are the episodes associated with urgency, or is there no prior warning? Does incontinence occur on a background of constipation or diarrhea? Does the patient require assistance to reach the toilet? After incontinence has been characterized, detailed questions concerning childbirth, prior anorectal surgery, rectal prolapse, hemorrhoids, and straining at stool should be addressed. A history of central or peripheral nervous system diseases, diabetes mellitus, prior pelvic irradiation, or diarrheal disease should be elicited. Perianal inspection and digital examination provide clues to the cause of incontinence. Anal deformities, tumors, perianal infections, fistulae, and prolapsing hemorrhoids may be visually apparent. Absence of the anal wink (contraction of the subcutaneous portion of the external sphincter) in response to a perineal pin stroke suggests a neuropathic process. Digital examination allows a crude assessment of anal resting and voluntary squeeze tone. A rough estimate of the anorectal angle and puborectalis function can be made by palpation of this muscle in the posterior midline at the proximal edge of the external sphincter during rest and voluntary squeeze. The presence of fecal impaction, rectal prolapse, and perineal descent can also be determined.

Clinical Testing Several tools are available to evaluate the mechanisms of continence. The extent of investigation depends on local expertise and the patient’s clinical presentation.

Anoscopy and sigmoidoscopy. Inspection of the distal colon is a minimum requirement in the evaluation of incontinence. Discovery of tumors or mucosal inflammation (infectious, ulcerative, ischemic, or radiation proctitis) directs treatment and probably eliminates the need for further evaluation.

Manometry. Sphincter function is determined by measuring resting and maximal squeeze pressures of the anal canal using balloons, miniaturized transducers, or perfused catheters. Rectal balloon inflation is performed to measure the reflex relaxation of the internal sphincter and contraction of the external sphincter muscles. Sophisticated techniques can determine spatial sphincter defects by cross-sectional mapping of the whole sphincter. [132] The techniques of dynamic manometry and prolonged anal manometry using a sleeve sensor may offer more information than standard static anal manometry. [133], [134]

Tests of rectal compliance and sensation. Estimates of rectal sensation are made by inflating a balloon in the rectum and measuring various subjective thresholds such as first volume sensed, volume producing urgency, maximum tolerated volume, and the delay in sensation. [135], [136] Rectal compliance is determined by obtaining concurrent balloon volume and pressure measurements so that pressure-volume curves can be generated.

Neurophysiological tests. External sphincter and puborectalis muscle activity can be measured using conventional concentric needle electromyogram (EMG) or more sophisticated single-fiber EMG. These techniques measure muscular denervation and reinnervation characteristic of neuropathy and allow sphincter mapping but require considerable expertise and are uncomfortable for the patient. Nerve dysfunction can be more directly characterized by measuring pudendal nerve terminal motor latencies and pudendal evoked potentials. [125], [137] Pudendal nerve terminal motor latency can be easily measured using a simple digitally placed electrode. Abnormal nerve conduction, especially bilaterally, may predict a poor surgical outcome. [138]

Imaging modalities. Muscle thickness and defects in both the internal and external sphincter can be mapped in patients with traumatic or idiopathic incontinence without radiation or the discomfort of EMG needle electrodes. Anal endosonography offers an exciting new way to image the anorectal angle and sphincter musculature with high resolution. [139] Endosonography after vaginal delivery may detect clinically inapparent sphincter tears that may be associated with subsequent incontinence. [140] Endosonography may also predict the response to nonoperative treatment in men with incontinence. [141] Using surgical anatomy at the time of repair as a gold standard, endoanal MRI may be even more accurate than sonography. [142]

Anal sensory tests. Miniature probes can now measure thermal and electrical sensitivity of the anal canal. [121], [143] Information from these novel tools may offer insights into the anal sampling reflex and anal sensory abnormalities identified in incontinent patients. [144]

Defecography. Defecography radiographically evaluates defecation by video fluoroscopic imaging during actual voiding of a simulated barium stool. [145] It is well tolerated and easy to perform. Static and dynamic measurements of the anorectal angle, pelvic floor, and puborectalis function are made during rest, squeeze, and attempted defecation. Pathology best recognized during defecation, such as perineal descent, rectal prolapse, enterocele, and rectocele, can be diagnosed, although symptoms may not correlate well with these defects. [146]

Continence testing. Several tests have been devised to challenge the integrated mechanisms of continence. These tests quantitate continence by measuring

leakage of rectally infused saline or resistance to passage of a solid object. [147] They offer only an approximation of the clinical symptom and cannot tease out specific pathologies. However, continence tests standardize the measurement of incontinence and require no expensive equipment or particular expertise. They may be useful for comparing groups of patients or assessing an individual's response to therapy over time. The strategy employed to evaluate fecal incontinence depends on the patient's clinical presentation, the availability of investigative tools, and the likelihood of altering future therapy. Soiling associated with an acute diarrheal illness probably requires no specific anorectal evaluation other than evaluation of the diarrhea itself. In a similar vein, the discovery of ulcerative proctitis by proctosigmoidoscopy requires no further anorectal studies, and treatment may be instituted immediately. For the majority of cases that are less clear-cut, anorectal manometry, combined with a balloon inflation estimate of rectal sensation, is an important functional study. It also allows for future treatment in the form of anal biofeedback if necessary. Endosonography, or perhaps MRI, offer the best anatomic testing. Defecography, rectal compliance tests, and nerve conduction measurements are useful adjunctive tools depending on local expertise.

Treatment

Fecal incontinence often responds well to a combination of relatively simple interventions. Therapy is at least partially dependent on the underlying cause. As a first priority, diarrheal stools, if present, must be treated. Solid stools are much easier to retain than liquid stools, so this simple measure alone may eliminate all symptoms. If possible, the underlying cause of diarrhea should be corrected, but, if not, empiric therapy with fiber agents or antidiarrheals is appropriate. Loperamide and diphenoxylate are effective in producing solid stools and reducing stool frequency in those with chronic diarrhea and incontinence. [148], [149] However, loperamide does this more effectively than diphenoxylate and also appears to improve continence to a standard saline load test. [149], [150] Seepage of liquid stools around a fecal impaction should, of course, not be treated as diarrheal stools. Instead, the impaction must be cleared with enemas and a treatment plan for constipation instituted. Incontinent debilitated patients or those with nervous system injuries usually respond best to a bowel program of regular defecation. Such a program should take advantage of the gastrocolonic response and judiciously use constipating agents or enemas and suppositories as appropriate. Fiber is useful for the frequent, low-volume, loose stools of irritable bowel syndrome but may need to be restricted for diseases with poor rectal reservoir capacity in order to decrease fecal volume. Anticholinergics should be tried to blunt the gastrocolonic response in patients with irritable bowel syndrome with urgency-associated incontinence after meals. Practice of sphincter exercises several times daily to improve both the tone and the awareness of the pelvic diaphragm and sphincter muscle is a useful adjunct to treatment in the incontinent patient.

If incontinence persists despite the measures outlined and the patient is motivated, sensitive to rectal distention, and able to contract the external sphincter, anal biofeedback should be attempted. [151], [152] This technique applies simple operant conditioning to the anal sphincter. Similar techniques can be used to gradually lower sensory thresholds to rectal distention in those with impaired rectal sensation caused by diabetes or other neuropathies. [122], [135] A report of biofeedback using a simple anal plug to record external sphincter contractions argues against the need for a rectal balloon, although a recent controlled trial suggests that enhancement of rectal sensitivity is important. [153], [154] Large well-designed studies are needed to confirm the approximate 70% success rates of biofeedback therapy. [155], [156] Conditions that may predict a poor response to biofeedback therapy include severe neuropathy causing poor rectal sensation and near absent external sphincter contraction during squeezing, irritable bowel syndrome, anterior resection of the rectum, and the surgical keyhole deformity of the anal canal caused by previous posterior sphincterotomy. [152], [153], [157] Urgency incontinence responds better than passive leakage. [158]

Patients with major fecal incontinence unresponsive to medical management are the most appropriate candidates for surgery. Surgical correction of rectal prolapse or third-degree hemorrhoids should be performed to improve the fecal soiling associated with these conditions. Several operations have been advocated for intractable symptoms. Incontinence caused by trauma, anal surgery, or obstetrical injury may respond to simple external anal sphincter apposition or overlapping repair. [159] Postanal repair is the procedure of choice for complex sphincter injuries or for neuropathic damage to the pelvic floor (often idiopathic) and loss of the normal anorectal angle. [74], [119] More than two thirds of patients can expect good results, although reduction of the anorectal angle cannot be demonstrated. [120], [160] Gracilis muscle transposition (graciloplasty) is a technically demanding procedure that may benefit patients with a destroyed sphincter or congenitally poor pelvic floor development. [161] The addition of an implantable device to electrically stimulate the neoanal sphincter (dynamic graciloplasty) provides additional benefit. [162], [163] More than 60% have satisfactory results but complications requiring reoperation are common. [164] Implantation of an artificial sphincter may be considered for those with nonreconstructable sphincters or severe neurogenic incontinence. Modifications of the device have reduced mechanical complications, but infectious complications still occur and long-term results are adequate in only 50% of patients. [165] Finally, as a last resort, placement of a colostomy should be considered. Substituting management of a stoma for unremitting fecal incontinence may significantly improve the quality of life for selected patients.

PRURITUS ANI

Pruritus ani is an annoying itchy sensation of the anus and perianal skin. It may be temporary or chronic, unrelenting, and difficult to treat. Pruritus is often associated with burning and soreness. Periods of intense itching vary in frequency but are characteristically most disturbing at night after daytime distractions are removed. It affects 1% to 5% of the population with a male-to-female predominance of 4:1. [166]

Etiology

Pruritus ani may occur as a symptom of a specific disease affecting the perianal skin or, more commonly, as an idiopathic condition possibly related to residual fecal material (Table 92-4). A number of anorectal diseases, including fissures, fistulae, and hemorrhoids, produce pruritus ani because of fecal soiling and contamination of the perianal region. These lesions make local hygiene especially difficult, and irritated, macerated skin may develop. Pruritus, discomfort, and mucus drainage are more common in those suffering from hemorrhoids than in a control population and often resolve with treatment of the hemorrhoids. [167] Anal malignancies, including Bowen disease, epidermoid cancers, and extramammary Paget disease, may rarely present as pruritus ani and should not be discounted without careful anal inspection, even if symptoms have existed for years. [168]

Anorectal diseases and fecal contamination
Diarrhea
Anal incontinence
Hemorrhoids
Fissures
Fistulae
Rectal prolapse
Malignancy: Bowen disease, epidermoid cancer, perianal Paget disease
Infections
Fungal: candidiasis, dermatophytes
Parasitic: pinworms, scabies
Bacterial (nonvenereal): Staphylococcus aureus, erythrasma
Venereal: herpes, gonococcal, syphilis, condylomata acuminata
Local irritants
Moisture: obesity, excessive perspiration
Soaps: hygiene products
Toilet paper: perfumed, dyed
Underwear: irritating fabrics, detergents
Anal creams, suppositories
Dietary: coffee, beer, acidic foods
Drugs: mineral oil, ascorbic acid, hydrocortisone sodium succinate, quinidine, colchicine
Dermatologic diseases
Psoriasis
Atopic dermatitis
Seborrheic dermatitis

TABLE 92-4 Causes of Pruritus Ani

Candida albicans and dermatophyte infections appear as characteristic localized erythematous rashes but may also be cultured from the perianal area even if skin lesions are not present. *C albicans* is more likely to be identified if random cultures are obtained, but dermatophyte infections, if present, are more likely to be associated with pruritus ani. [169] Pinworm (*Enterobius vermicularis*) causes pruritus ani that is characteristically nocturnal. Children are most commonly affected, but adults, especially those exposed to children, may occasionally become infected. Diagnosis is best made by applying adhesive cellophane tape to the perianal skin early in the morning in an effort to detect eggs. Scabies (*Sarcoptes scabiei*) and pubic lice (*Pthirus pubis*) can also cause pruritus ani. Itching in the genital region and identification of organisms or eggs on pubic hair lead to the proper diagnosis.

Perianal bacterial infections have been implicated in pruritus ani. Separate reports have identified *Staphylococcus aureus* and erythrasma (*Corynebacterium minutissimum*) as pathogens in patients with pruritus ani. [170], [171] However, a controlled study has failed to provide a bacteriologic basis for this condition. [172] A number of sexually transmitted diseases have been associated with pruritus ani. These include herpes simplex, gonorrhea, syphilis, condyloma acuminatum, and molluscum contagiosum. [166] Dermatologic conditions, such as psoriasis and eczema, must also be considered. One series of patients referred to a combined colorectal and dermatologic clinic had a dermatosis, particularly psoriasis, as the most common cause of pruritus ani. [173] A variety of local irritants, allergens, and chemicals that contact the perianal skin may cause pruritus. Perfumes and dyes present in toilet paper and soaps, personal hygiene products, irritating fabrics, laundry detergents, and even well-intentioned medications in the form of topical anal creams or suppositories are examples. Even intravenous steroids and chemotherapies have been implicated. [174], [175] Clinical experience and elimination diets implicate certain dietary products such as coffee, cola, beer, tomatoes, chocolate, tea, and citrus fruits in the production of anal pruritus, although the mechanism is unknown. [176], [177] Any food product that causes diarrhea in a patient may lead to pruritus simply because of frequent fecal contamination of the perianal region.

Idiopathic pruritus ani is usually caused by a combination of perianal skin fecal contamination and trauma. Ambulatory monitoring has shown exaggerated transient internal sphincter relaxation which likely predisposes to subtle fecal leakage in idio-pathic pruritus ani. [178] Not surprisingly, feces and colonic mucous secretions are irritating to perianal skin. [179], [180] Acute diarrhea in an otherwise healthy person may lead to irritation of the perineum because of frequent wiping. Tiny amounts of feces repeatedly contaminate the sore perianal skin, leading to further irritation. Soon the itch-scratch cycle develops, serving to perpetuate skin trauma. Patients with chronic pruritus ani often suffer from frequent loose stools or some degree of fecal incontinence or seepage. This hostile environment is repeatedly traumatized from vigorous wiping or scratching, and adequate healing can never occur. Pruritus is further exacerbated by inadequate perianal hygiene or excessive moisture caused by perspiration, airtight clothing, severe obesity, or a particularly deep-set anus.

Treatment

With patience and time, most causes of pruritus ani are successfully treated. Dermatologic diseases and infectious causes of pruritus ani are uncommon but, if identified, should be treated in the usual manner. Anorectal disorders that confound attempts at good perianal hygiene, such as prolapsing internal hemorrhoids or painful anal fissures, should be corrected. Fecal leakage or incontinence must be aggressively managed to avoid soiling of perianal skin. For the same reason, frequent loose stools should be minimized with antidiarrheals and fiber agents if appropriate. Foods or beverages that produce diarrhea or pruritus symptoms in a particular individual may be discontinued. Blanket recommendations to discontinue or curtail coffee, beer, citrus fruits, and other foods reported to cause pruritus ani lack firm support, but elimination diets may be appropriate in particular cases. The key to management of pruritus ani is to keep the anal area clean and dry while minimizing trauma caused by wiping and scratching. [181] The patient should be instructed to gently cleanse the anal skin with a premoistened pad or tissue after defecation. Witch hazel preparations or soothing lanolin-containing lotions are useful for this purpose. This always removes more residual feces than dry wiping alone. The area should be dried with a blow dryer or a soft tissue using a gentle dabbing, not rubbing, motion. Those with unpredictable small amounts of fecal discharge are instructed to wear a thin cotton pledget. This is barely perceptible when applied to the anus and should be changed frequently throughout the day. Excessive perspiration is corrected with application of baby powder and avoidance of tight, nonporous clothing. Irritating fabrics, greasy salves, perfumed toilet paper, hygiene products, and soaps should be avoided. A 1% hydrocortisone cream may be applied sparingly twice daily during the acute phase of pruritus ani but should not be used for longer than 2 weeks to avoid skin atrophy. Applying a protective ointment (zinc oxide) over the antiinflammatory agent may facilitate healing. For severe nocturnal symptoms, a nighttime dose of a systemic antipruritic such as diphenhydramine is appropriate. Long-standing, intractable pruritus ani has responded very well to intracutaneous injections of methylene blue and other agents. [182], [183]

RECTAL FOREIGN BODIES AND TRAUMA

An astonishing hodgepodge of foreign bodies can become incarcerated in the rectum. [184] These include thermometers, enema tips, tools, bottles, light bulbs, pieces of food, and sexual devices inserted for purposes of medical treatment, concealment, assault, or, most commonly, eroticism. In an attempt to cope with an embarrassing situation, patients may claim the object was inserted to obtain relief from a particular discomfort or symptom. Frequently, the patient may complain of pain or bleeding without admitting to a foreign body insertion. [185] A directed interview and careful abdominal and rectal examination of an often sheepish patient are necessary to identify the foreign body and the potential risk of rectal trauma. Anteroposterior and lateral radiographs should be obtained to determine the object's outline and location and to detect pneumoperitoneum if present. Alternatively, a patient may unknowingly swallow a toothpick or piece of bone that later impacts in the anal canal and may mimic an abscess or fissure. [186]

Nearly all objects can be removed transanally without resorting to surgery. Even if the object is easily palpable, removal under direct vision is the best way to avoid iatrogenic injury. [187] Foreign bodies may be classified by position as low-lying if in the rectal ampulla or high-lying if at or proximal to the rectosigmoid junction. [187] Small, low-lying objects are retrieved transanally using an operative anoscope. Removal of larger objects, such as vibrators or rubber phalluses, may require regional anesthesia, anal dilation, and a grasping forceps. Large, bulky objects, such as glass bottles and light bulbs, require special care. After adequate anesthesia, several Foley catheters are inserted past the object. Air insufflation through the catheters relieves the proximal vacuum effect that may occur during attempted withdrawal. Gentle manipulation coupled with slow traction using the inflated Foley balloons successfully extricates the bulbous object in most cases. An obstetrical vacuum extractor has also been used to remove a glass foreign body. [188] If a glass object is broken accidentally during attempted removal, laparotomy is required. High-lying foreign bodies are managed using spinal anesthesia and the lithotomy position. By palpating the abdomen, the object is pushed distally and then simultaneously grasped with forceps through the sigmoidoscope to coax it into the rectal vault, from which it may be removed as a low-lying object. If the object cannot be delivered to within reach of the rigid sigmoidoscope manually or after 12 hours of observation, or if abdominal distress develops, laparotomy is indicated. Proctosigmoidoscopy should be performed after all foreign body retrievals to rule out retained objects, lacerations, hematomas, and perforations.

Other causes of rectal trauma include penetrating injuries (usually gunshot wounds), blunt trauma (motor vehicle accidents), impalement injuries (criminal assaults), homosexual activities (fist fornication), and iatrogenic injuries (endoscopy, enemas, surgical procedures). [189] Surgical management of major rectal trauma usually includes a diverting colostomy, presacral drain placement, distal rectal irrigation, and maximum preservation of sphincter musculature. [190]

ANAL CARCINOMA

Anal carcinomas are relatively rare, comprising only 1% to 2% of all colonic cancers. A variety of histological types have been described, including squamous cell (70%–80%); basaloid, also known as transitional cell or cloacogenic (20%–30%); mucoepidermoid (1%–5%); and small cell anaplastic type (<5%). [191] All can be broadly classified as epidermoid carcinomas for purposes of clinical discussion. The mean age at presentation of patients with anal cancer is approximately 60 years. [192] Perianal and rectal carcinoma are more common in men, but the occurrence of anal canal tumors is nearly twice as common in women. [192]

Etiology

Although the cause of anal cancer is unknown, several risk factors have been identified. These tumors are strongly associated with homosexual behavior and receptive anal intercourse in men and with a history of sexually transmitted diseases, especially genital warts, in both sexes. [193] These data, immunohistological studies, and polymerase chain reaction techniques suggest a causal role for human papillomavirus infection, particularly type 16. [194], [195] Smoking, human immunodeficiency virus (HIV) infection, multiple sexual partners, cervical neoplasia, and immunosuppression after organ transplantation are other important risk factors. [196] Anal cytology screening may be cost-effective in homosexual and bisexual men. [197]

Clinical Manifestations

Bleeding, pain, and sometimes pruritus are presenting symptoms of anal cancer. However 25% of patients are symptom-free, and it is not unusual for the lesion to be discovered incidentally at routine examination. Almost 2% of surgical specimens for benign anorectal disease are reported to contain previously unsuspected tumor, although a survey of hemorrhoidectomy specimens suggests a much lower incidence. [198], [199] Benign anal lesions are temporally associated with, but do not appear to cause anal cancer. [200] High-grade dysplasia identified by screening cytology must be confirmed by biopsy and may be present even before visible lesions exist. [201] Rarely, a palpable metastatic inguinal lymph node is the sole presenting manifestation. Because symptoms are usually mild and nonspecific and easily confused with those of common benign anorectal lesions, the tumor is discovered late in more than 60% of patients. At presentation, 15% to 30% of patients have metastases to

pelvic or inguinal lymph nodes, and 10% have distant spread to the liver or lungs. [191]

Diagnosis is made by biopsy (under anesthesia, if necessary), and the local extent of disease is determined by palpation, anoscopy, and sigmoidoscopy. Lesions originating from the anal canal tend to be undifferentiated and more aggressive, whereas those arising from the anal margin are more differentiated and less malignant. [202] A poor prognosis is portended by squamous cell tumors greater than 2 cm in size (especially those >5 cm), basaloid or anaplastic carcinomas, sphincter muscle invasion, or spread to regional pelvic or inguinal lymph nodes. [191]

Treatment

The standard therapy for small, noninfiltrating anal cancer has been wide local excision in an attempt to preserve normal anal function. [191], [192] Split-thickness skin grafts may be required to cover large denuded surfaces. Large or infiltrating lesions require an abdominoperineal resection for radical excision of structures, including the sphincters, levator muscles, and rectum. Prophylactic inguinal lymph node resection is not indicated.

The advent of supervoltage radiotherapy alone or combined with surgery or chemotherapy has changed the approach to this neoplasm. Studies by Nigro and others [203], [204] and [205] suggest that radiotherapy (external beam or interstitial) plus chemotherapy with 5-fluorouracil and mitomycin causes complete tumor regression in most cases and is superior to radiotherapy alone. Cisplatin-based regimens may prove to be superior to those with mitomycin. [206] Subsequent abdominoperineal resection is unnecessary if follow-up biopsies document eradication of tumor. This combined approach minimizes local tumor recurrence while retaining normal anal function. The 5-year survival rate exceeds 70%. [1] Abdominoperineal resection for combined chemotherapy radiation therapy failures has a poor prognosis. [207]

Nonepidermoid Anal Malignancies

A variety of uncommon neoplasms of the anus and perianal skin make up the nonepidermoid tumors of the anal region. Adenocarcinoma of the anal canal is a rare tumor often arising in anorectal fistulae. [192] This lesion may also arise from other anal glands or may be confused with distal extension of a primary rectal carcinoma. Despite radical abdominoperineal resection, recurrences are typical. Extramammary Paget disease is a perianal glandular tumor that tends to spread along the epidermis and to eventually metastasize. [192] It typically appears in the seventh decade of life as an erythematous, well-demarcated eczemoid plaque with ulcerations. Perianal Paget disease is frequently associated with an underlying colonic malignancy. [208] Wide total excision is indicated for localized disease. [208] More advanced lesions require a radical abdominoperineal resection with ipsilateral groin dissection if inguinal node involvement is seen. Anal melanoma accounts for 1% of all anal tumors and 1.6% of all melanomas. Typical lesions are 4 cm in diameter, nonpigmented in one third, and tend to metastasize early with 5-year survival rates of only 15% to 20%. [209] Radical resection offers no clear advantage over wide local excision. [210], [211] Basal cell carcinoma of the perianal skin is a very rare lesion that produces rolled skin edges with central ulceration. The prognosis is quite good after adequate local excision or radiotherapy. [212] Bowen disease is a slow-growing, cutaneous squamous cell carcinoma in situ. Lesions occur as reddish-brown scaly or crusted plaques that may resemble a patch of psoriasis or dermatitis. Treatment ranges from simple observation alone in lesions found incidentally in hemorrhoidectomy specimens to wide local excision. [213]

PROCTALGIA FUGAX AND THE LEVATOR SYNDROME

Proctalgia fugax and levator ani syndrome are types of functional anorectal pain that differ based on the duration, frequency, and quality of discomfort. Limited evidence suggests that the former may be caused by smooth muscle spasm and the latter by striated muscle tension. [214] Proctalgia fugax (literally “fleeting rectal pain”) is an obscure condition characterized by sudden, brief episodes of severe rectal pain. Rome diagnostic criteria require at least 3 months of the recurrent episodes of pain localized to the anus or lower rectum lasting from seconds to minutes and no pain between episodes. [214] The true incidence of the disorder is unknown, although surveys of healthy people reveal attacks of rectal pain in approximately 15%. [215] Most people do not seek medical advice, probably because the pain typically lasts less than 1 minute (84%) and occurs fewer than 6 times per year (72%). [216] Modern surveys identify no consistent gender predominance. [215], [216]

Attacks are described as an intense stabbing or aching midline pain above the anus, lasting several seconds to many minutes. Pain may be variably associated with an urge to expel flatus, a desire to lie on one side with hips flexed, and, rarely, cold sweats, syncope, and priapism. Occasionally, attacks are exclusively nocturnal. Stress, fatigue, heat, cold, and sexual activity have all been reported to induce pain, but usually no clear precipitant is identified.

Not surprisingly, proctalgia fugax has been associated with irritable bowel syndrome and a variety of psychogenic disorders such as anxiety and hypochondriasis. [217], [218] The cause of proctalgia fugax is unknown. Unsubstantiated theories include rectosigmoid intussusception, vascular migraine equivalent, accumulation of rectal gas, or spasm of the anal sphincter, rectosigmoid colon, or pelvic musculature. The diagnosis is made on the basis of a characteristic history alone. Anorectal examination and sigmoidoscopy are normal. A familial internal anal sphincter myopathy causing proctalgia fugax and difficulty with rectal evacuation has been reported. [219]

Of primary importance in the treatment of proctalgia fugax is reassurance as to the benign nature of the disorder. This advice in conjunction with treatment of irritable bowel syndrome or psychogenic disorders, if present, often suffices. A number of specific remedies have been proposed for more refractory symptoms. Their efficacy is difficult to determine given the transient nature of symptoms and the lack of even uncontrolled trials. Local therapies include rectal massage, firm manual pressure to the perineum, and application of warm soaks or baths. One author [220] claims good results if the patient spreads the buttocks while in a knee-chest position to allow expulsion of rectal gas. Pharmacological intervention should be reserved for those with particularly frequent, severe, or disabling symptoms. Anecdotal reports claim success with a variety of drugs to reduce spasm, including amyl nitrate, sublingual nitroglycerine, salbutamol, clonidine, and diltiazem. [221], [222] and [223] One controlled trial [224] found that salbutamol aerosol significantly shortened painful attacks.

The levator syndrome refers to an aching rectal pain caused by tenderness and spasm of the levator ani muscle group (ileococcygeus, pubococcygeus, and puborectalis). It is perhaps best thought of as a variant of proctalgia fugax, although the two disorders are frequently confused in the literature. The pain is more chronic, aching, and pressure-like than that of proctalgia fugax and is most commonly seen in middle-aged women. [225] Rome diagnostic criteria require at least 3 months of chronic or recurrent rectal pain or aching, episodes lasting at least 20 minutes, and absence of other causes of rectal pain. [214] Defecation, prolonged sitting, and precipitants of proctalgia fugax have been described as precipitants of levator syndrome pain. The key diagnostic finding is palpable tenderness and spasm of the levator muscles as the examining finger sweeps 180 degrees from the coccyx to the pubis posteriorly to anteriorly. [225] For unknown reasons, findings are more frequently localized to the left side. Treatment consists of variable combinations of reassurance, local heat, vigorous digital rectal massage of the levator musculature, and muscle relaxants. In those with persistent symptoms, several groups have reported beneficial results using biofeedback training or electrogalvanic stimulation. [226], [227], [228] and [229]

MISCELLANEOUS CONDITIONS

Coccygodynia refers to pain in the coccyx. The discomfort is sharp or aching in quality and may radiate into the rectal region or to the buttocks. Traumatic arthritis, dislocation or fracture as a result of injury, or difficult childbirth can all cause organic coccygodynia. [1] Prolonged sitting has been suggested to cause functional coccygodynia. In both cases, manipulation of the coccyx reproduces pain and muscular spasm. Many patients respond to symptomatic treatment with warm soaks and analgesics. Local injection of steroid with or without coccygeal manipulation is usually beneficial in those with persistent symptoms. [230] Rarely, coccygectomy is required.

Unusual causes of rectal pain include cauda equina tumors, pelvic tumors, perianal endometriosis, intermittent enteroceles, a variety of rare retrorectal tumors and cysts, and rare genetic syndromes. [1], [231], [232] Other idiopathic, presumably functional, perineal and rectal pain syndromes remain poorly understood. [233]

Pilonidal (“nest of hair”) disease is a common skin lesion of the gluteal cleft seen most frequently in young men. Family history, obesity, and local trauma are risk factors. [234] Patients complain of recurrent drainage, swelling, or pain. Once thought to be a congenital lesion, pilonidal abscess is now recognized to be an acquired condition of the midline coccygeal skin region, often secondarily invaded by hair. Definitive treatment is usually surgical. [235], [236]

Hidradenitis suppurativa is a chronic suppurative condition of the apocrine glands most often involving the axillary and inguinoperineal skin regions. Apocrine glands

are activated at puberty, so the disease is not seen before adolescence and rarely occurs initially after middle age. Axillary hidradenitis suppurativa is most common, but perineal involvement is seen in nearly one third of those with the disease, especially in men. [237] The best recognized risk factors are obesity and acne, although perspiration and mechanical trauma to the skin and hair of the anogenital region may also predispose to perianal hidradenitis. Warm wet compresses and antibiotics are appropriate for early disease but surgical treatment is usually necessary. [238]

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CHAPTER 93

Diane M. Simeone and Michael W. Mulholland

PANCREAS: ANATOMY AND STRUCTURAL ANOMALIES

Cross-sectional imaging techniques have greatly increased the understanding of pancreatic anatomy, and the ability to manipulate the pancreas surgically, endoscopically, and percutaneously has made it necessary for a wide variety of physicians to be familiar with pancreatic anatomic variations. The anatomy of the pancreas has been described in many standard texts, but the functional and therapeutic implications of its anatomic relations often are underemphasized.

Diseases of the pancreas are often more difficult to manage medically or surgically than those of other abdominal viscera. The pancreas lies hidden in the deeper recesses of the retroperitoneal space of the upper abdomen, is largely protected by the lower rib cage anteriorly, and is almost completely covered by the stomach, transverse colon, and transverse mesocolon. At laparotomy, the pancreas cannot be properly visualized or palpated without extensive dissection, mobilization, and retraction.

The central position of the pancreas provides for lymphatic drainage along several major routes, namely, the splenic, hepatic, and superior mesenteric nodal systems as well as the aortocaval and other posterior abdominal wall lymphatics. Moreover, the intimate anatomic association of the pancreas with vital major vessels of the epigastrium at once limits the extent of any surgical procedure and also dictates what must be removed. Thus, when a tumor spreads a short distance to involve the superior mesenteric vein, the portal vein, or the celiac axis, it usually becomes incurable. Similarly, if the gland is removed in radical fashion, the need to excise the vessels and lymph nodes associated with it frequently makes necessary the removal of the duodenum, gallbladder, distal bile duct, spleen, upper jejunum, and part of the stomach. Finally, the vascular nature of the pancreas and the adjacent organs makes it easy to understand why the most common intraoperative and postoperative complication of pancreatic resection is hemorrhage.

EMBRYOLOGICAL DEVELOPMENT

The pancreas develops from two primordial outpouchings of the duodenum and is first apparent at 4 weeks' gestation. The dorsal pancreatic bud grows more rapidly than the ventral pancreas and by 6 weeks extends into the dorsal mesentery. The ventral pancreas remains smaller and is carried away from the duodenum by the development of the hepatic rudiment into the biliary system. Differential growth of the duodenum and axial rotation of the gut result in the dorsal pancreas being carried to the left and the ventral pancreas being carried to the right of the duodenum (Fig. 93-1). Migration of the distal common bile duct behind and to the left of the duodenum causes the ventral pancreas to lie below the dorsal pancreas, forming the uncinate process of the pancreas. The common bile duct lies posterior to the dorsal pancreatic duct. Fusion of the two parts of the pancreas occurs during the seventh week of gestation. Fusion of the ventral duct with the dorsal duct results in the formation of the main pancreatic duct. The proximal end of the dorsal pancreatic duct usually does not communicate with the main duct and forms the accessory pancreatic duct.

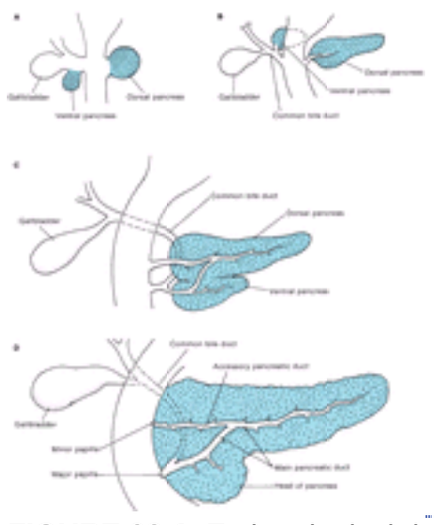

FIGURE 93-1. Embryological development of the pancreas. The various stages of intrauterine pancreatic growth are shown at 4 weeks' gestation (**A**), 6 weeks (**B**), 7 weeks (**C**), and birth (**D**). (Adapted from Arey LB. Developmental anatomy, 7th ed. Philadelphia: WB Saunders, 1974:259.)

The pancreatic acini and the first zymogen granules appear at 12 weeks' gestation. Groups of endocrine cells develop from multipotential stem cells in the ductular epithelium (nesidioblasts) at 9 weeks' gestation. Discrete islets of Langerhans' cells can be identified at 12 weeks. Most of the islet cells develop within the tail of the pancreas and the dorsal pancreas. The first cells to produce granules are the alpha cells, soon followed by the beta and delta cells. Complete maturation of the pancreatic gland does not occur until sometime after the end of gestation. The smooth muscle of the sphincter of Oddi develops independently of the duodenal musculature, and only later does it become incorporated into the duodenal wall.

Most recent evidence indicates that islet cells arise from stem cells that appear in pancreatic ducts during the third month of gestation. Insulin-containing granules may be demonstrated immunocytochemically by the end of the third month. Islet cells migrate away from the ducts in which they arise and move into the interlobular connective tissue. Mature islet morphology is established before birth.

GROSS ANATOMY

The pancreas is a soft, flattened, elongated gland, 12 to 20 cm long in the adult, that lies behind the peritoneum of the posterior abdominal wall and is obliquely rather than transversely oriented (Fig. 93-2; see also Fig. 93-1). The pancreas has a lobular structure and, although totally invested in fine connective tissue, does not have a true fibrous capsule. The adult gland weighs 85 to 95 g. Because of its oblique orientation, a transverse section or computed tomography scan normally does not pass through the entire length of the gland. The head of the pancreas is on the right side and lies within the C-shaped concavity of the duodenum at the level of the

body of L2. The tail of the gland is to the left; the tail lies between the two layers of peritoneum that form the lienorenal ligament and is located at the level of the body of L1. The first portion of the duodenum is suspended in front of the head of the gland. The lesser curvature of the second part of the duodenum and the upper aspect of the third part of the duodenum intimately invest the head of the pancreas. Superiorly, the head is related to the gastroepiploic foramen and the structures that form the contents of the free border of the lesser omentum. Anteriorly, the first portion of the duodenum covers the superior part of the pancreatic head, and below this the right side of the transverse mesocolon is attached transversely. Posteriorly, the head of the pancreas contacts the right renal hilum, both renal veins, the inferior vena cava, and the termination of the right gonadal vein as well as the right side of the aorta. [1]

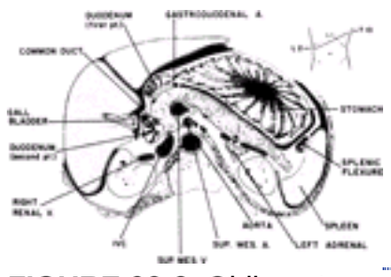

FIGURE 93-2. Oblique transverse cross-section of the upper abdomen viewed from below. The section passes through the long axis of the pancreas at approximately the level indicated in the inset. The disposition and relations of structures shown approximate those seen on oblique transverse ultrasonic scanning.

The distal end of the common bile duct passes behind the upper border of the head of the pancreas. The bile duct grooves the posterior aspect of the head of the gland before passing through the substance of the head to reach the duodenal papilla of Vater. From the posterior aspect of the head, a tongue of pancreatic tissue of variable size, the uncinate process, extends to the left to occupy the concavity formed by the third and fourth part of the duodenum. The uncinate process lies anterior to the inferior vena cava and aorta and is covered superiorly and anteriorly by the superior mesenteric vessels as they emerge below the neck of the pancreas.

From the head of the pancreas, a constricted part of the gland, the neck of the gland, extends toward the left. It is 3 to 4 cm wide and joins the head of the gland to the body of the gland on the left. The pancreatic neck lies behind the posterior peritoneum of the lesser sac, and attachments of the transverse mesocolon and the root of the small bowel mesentery cover its inferior border. The neck of the pancreas lies anterior to and is closely related to the confluence of the superior mesenteric and splenic veins. These form the lowest part of the portal vein.

In front of the aorta in the midline, the body of the pancreas continues its retroperitoneal course toward the left side, held securely against the aorta and the posterior parietes by the posterior peritoneum of the lesser sac. The antrum and the body of the stomach contact the body of the pancreas anteriorly. The left renal vein, passing between the aorta and the pancreas, is separated posteriorly by the latter from the first part of the superior mesenteric artery. At a slightly higher level, elements of the celiac and superior mesenteric plexus ramify between the pancreas and the aorta. The midline part of the body of the pancreas is pushed anteriorly by the bodies of L1 and L2 and, therefore, lies closest to the anterior abdominal wall. Because of its prominence and fixity in this region, this area of the pancreas is most vulnerable to blunt abdominal injuries. In addition, a tumor in this region may be palpated as a mass that does not move with respiration and that strongly transmits the aortic pulsation. The celiac axis, surrounded by the celiac plexus, divides into its major branches between the superior border of the body of the pancreas and the crura of the diaphragm.

The body of the pancreas passes laterally, posteriorly, and slightly cephalad behind the posterior peritoneum of the lesser sac and merges imperceptibly with the tail of the gland. The exact junction of the body and tail is not discernible anatomically. The posterior relationships of the body and tail of the pancreas include the posterior attachment of the left crus of the diaphragm, the left suprarenal gland, the upper pole of the left kidney, and hilum of the spleen. The splenic vein above and the left renal vein below lie close to one another behind the body and tail of the gland.

At the upper border of the body and tail of the pancreas, the splenic artery courses to the left from its origin at the celiac axis. The transverse mesocolon is attached to the anterior part of the lower border of the gland. The splenocolic ligament attaches the splenic flexure of the colon to the hilum of the spleen and brings it adjacent to the tail of the pancreas.

The shape and disposition of the pancreas varies, as demonstrated by pancreatography. In 57% of people, the bulk of the head of the gland is to the right of the spinal column. In about 38% of cases, it lies directly over the spine, but only rarely (5%) does the head of the pancreas lie to the left of the spine. There is a tendency in older adults for the pancreas to be ptotic. The duodenal opening of the main pancreatic duct lies at the level of L2 to L4 in 92% of cases, but, rarely, it has been described at a lower level.

SURGICAL EXPOSURE

Although the pancreas is located retroperitoneally, it must be approached by way of an anterior laparotomy incision. Little of the gland can be seen or palpated at laparotomy without dissection. In a thin patient, small areas of the head may be seen directly behind the peritoneum of the supracolic and right infracolic compartments, and the inferior border of the body and tail may be seen from the left infracolic compartment at the root of the transverse mesocolon. These limited views, however, usually are obscured by omental, mesocolic, and retroperitoneal fat. By passing a finger through the gastroepiploic foramen of Winslow, the neck of the pancreas may be felt from above.

The head of the pancreas may be inspected and palpated more closely by performing two maneuvers. [2] First, the hepatic flexure of the colon is mobilized downward and medially, dividing the attachments of the transverse colon to the front of the duodenum and pancreatic head as far as the origin of the middle colic vessels. In addition, the attachments of the right side of the greater omentum to the transverse colon are divided. Thus, the lesser sac can be widely exposed on the right side of the middle colic vessels. Second, the peritoneum lateral to the second part of the duodenum is incised, and the duodenum and pancreatic head may be elevated and swept to the left by blunt dissection, exposing the right renal vein, the inferior vena cava, the right gonadal vein, the origin of the left renal vein, and the retroduodenal and retropancreatic portions of the distal common bile duct. After this mobilization (Kocher maneuver), the head of the pancreas may be palpated anteroposteriorly between the thumb and fingers. The mesenteric vessels are obscured from view by the uncinate process.

Limited visualization of the superior part of the body of the pancreas may be obtained by opening an avascular part of the lesser (gastrohepatic) omentum and retracting the lesser curvature of the stomach inferiorly. This maneuver also brings the celiac axis into view. A much more adequate visualization of the body of the pancreas may be obtained by widely opening the lesser sac, which can be achieved by dividing the gastrocolic omentum at its attachment to the transverse colon. Extending this opening to the right into the subpyloric region allows visualization of the anterior aspect of the head and neck of the pancreas, especially if the right gastroepiploic vessels are ligated and divided. Extending the opening to the left and dividing the short gastric (gastrosplenic) vessels in the gastrosplenic ligament superiorly and the relatively vascular splenocolic ligament inferiorly will give complete visualization of the anterior surface of the body and tail of the pancreas. The spleen, splenic vessels, and tail of the pancreas may be mobilized medially and anteriorly en bloc, allowing inspection of the posterior aspect of the tail and body of the gland. All these surgical maneuvers may be carried out quickly and safely with little risk of damage to vital structures or troublesome bleeding. By this means, all except the region of the neck and uncinate process of the pancreas may be evaluated fully.

ARTERIAL BLOOD SUPPLY

The pancreas derives its blood supply from the celiac axis and the superior mesenteric artery (Fig. 93-3; see also Color Fig. 93-3). These major vessels and their branches also provide the blood supply to other vital organs adjacent to the pancreas.

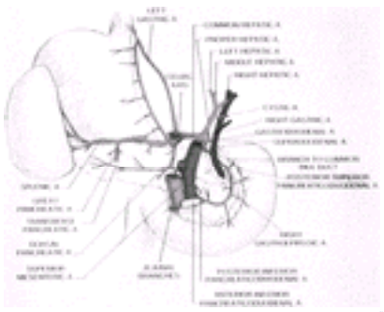

FIGURE 93-3. (See Color Figure 93-3.) Blood supply of the pancreas. The pancreas, duodenum, stomach, and spleen are viewed from their posterior aspects.

Typically (in 90% of people), the celiac axis divides into the common hepatic, splenic, and left gastric arteries. The common hepatic artery passes to the right, anterior to the portal vein, to reach the free border of the lesser omentum and usually lies to the left of the common bile duct. After giving off the gastroduodenal artery, the hepatic artery turns upward toward the porta hepatis and divides into the left and right hepatic arteries. The middle hepatic artery, supplying the caudate lobe of the liver, is usually a branch of one of these two vessels. This arrangement of the hepatic arterial blood supply, however, occurs in only 55% of people. Replaced or accessory hepatic arteries are frequent, occurring in as many as 30% of people. The most frequent variation from normal is the right hepatic artery branching off from the superior mesenteric artery and coursing behind the uncinate process and the head of the pancreas to reach the free border of the lesser omentum. [3]

The gastroduodenal artery usually originates from the hepatic artery. The gastroduodenal artery courses from its origin behind the first part of the duodenum and, after giving off the posterosuperior pancreaticoduodenal artery, lies on the anterior surface of the head of the pancreas. At the lower border of the first part of the duodenum, it branches into the right gastroepiploic artery and the anterior superior pancreaticoduodenal artery.

For the most part, the head of the pancreas and the duodenum have a common blood supply: the anterior and posterior pancreaticoduodenal arcades. The anterior arcade gives off the only arteries that enter the head of the gland from its anterior aspect. This arcade is formed by the anastomosis of the anterosuperior pancreaticoduodenal artery, which is a terminal branch of the gastroduodenal artery, and the anteroinferior pancreaticoduodenal artery, which is usually a branch of a common inferior pancreaticoduodenal artery, arising from the superior mesenteric artery.

The posterior pancreaticoduodenal arcade is formed by the anastomosis of the posterosuperior pancreaticoduodenal artery, usually a branch of the gastroduodenal artery, and the posteroinferior pancreaticoduodenal artery, which arises in a way analogous to the anterior pancreaticoduodenal artery. Of surgical importance is the relation of the posterosuperior pancreaticoduodenal artery to the common bile duct. Typically, this vessel arises from the gastroduodenal artery and passes to the right, anterior to the common bile duct, contributing the major source of the structure's blood supply. The supraduodenal portion of the common bile duct also may be crossed anteriorly by the right hepatic artery, the right gastric artery, and the supraduodenal artery, making mobilization of the lower end of the common bile duct more difficult.

Because of the shared blood supply of the duodenum and pancreatic head, extensive interference with the pancreaticoduodenal arcades in the course of a 95% pancreatectomy may compromise the blood supply of the duodenum. [4] In addition, because the duodenojejunal flexure and the first part of the jejunum may derive their blood supply from branches of the inferior pancreaticoduodenal artery or the pancreaticoduodenal arcades, ligation of these vessels in the course of a resection may render the proximal jejunum ischemic.

The body and tail of the gland derive their blood supply chiefly from branches of the splenic artery. This vessel, the largest branch of the celiac axis (5–11 mm in diameter), courses laterally at the upper border of the pancreas; its characteristic marked tortuosity appears to be related to age: it is often absent in infants and is most marked in the elderly. The splenic artery gives multiple side branches to supply the neck, body, and tail of the pancreas. The termination of the splenic artery passes between the layers of the lienorenal ligament and, at the hilum of the spleen, four or five branches enter the splenic hilum separately.

VENOUS DRAINAGE

The general pattern of veins draining the pancreas is the same as that of the arterial blood supply (Fig. 93-4). Blood from the pancreas ultimately drains into the portal vein, which is formed by the junction of the superior mesenteric vein and splenic vein behind the neck of the gland. The close relationship of the portal and superior mesenteric veins to the head, neck, and uncinate process of the pancreas is of vital surgical importance; in determining the resectability of a pancreatic lesion, a key step is assessment of involvement of these veins. The portal vein originates from behind the neck of the pancreas as a continuation of the superior mesenteric vein after it becomes confluent with the splenic vein. The portal vein passes upward to the right to gain access to the free border of the lesser omentum, where it lies posterior to the hepatic artery and the common bile duct. At the porta hepatis, it divides into a short, broad right branch and a longer, narrower left branch. In adults, the portal vein is about 8 to 10 cm long and 8 to 14 mm wide. Its average length is about 8.4 cm. [5]

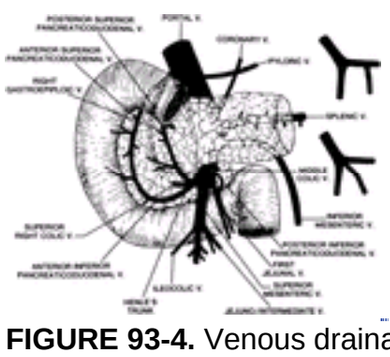

FIGURE 93-4. Venous drainage of the pancreas. The pancreas and duodenum are viewed from their anterior aspects. **Insets:** Normal variations in the termination of the inferior mesenteric vein.

The splenic vein originates at the hilum of the spleen by the confluence of five or six tributaries draining the spleen. It receives the basal brevia (short gastric veins) and left gastroepiploic veins at the hilum. It passes through the lienorenal ligament behind the tail of the pancreas and below the splenic artery and is the large, straight vein that courses to the right in contact with the posterior surface of the pancreas. It receives many tributaries from the tail, body, and neck of the gland.

Below the pancreas, the ileocolic vein passes directly, and usually without tributaries, from the ileocecal region to join the right side of the superior mesenteric vein in the midline just above the inferior border of the third part of the duodenum. At about the same level on the left side, the jejunointermediate vein joins the superior mesenteric vein. The confluence of these three veins forms the lower limit of what has been termed the surgical trunk of the superior mesenteric vein. Above this point, about two thirds of the superior mesenteric vein lies below the inferior border of the neck of the pancreas, with the remaining one third being retropancreatic. The superior jejunal vein joins the left side of the superior mesenteric vein. The middle colic vein usually joins the superior mesenteric vein immediately below the inferior border of the pancreatic neck.

The inferior mesenteric vein typically joins the splenic vein vertically behind the body of the pancreas. The latter passes horizontally and combines with the superior mesenteric vein at a right angle to form the portal vein, which turns obliquely to the right to pass into the free edge of the lesser omentum. Small veins draining the head of the pancreas pass directly into the portal vein. Similarly, small veins draining the uncinate process terminate in the right and posterior aspects of the retropancreatic superior mesenteric vein. The anterior aspects of the major veins are usually free from tributaries. From the left side, the coronary vein joins the retropancreatic portal vein, and the pyloric vein joins the suprapancreatic portal vein. The superior and posterior pancreaticoduodenal veins drain into the portal vein from the right side, deep to the first part of the duodenum and opposite the pyloric vein. The coronary vein (also known as the left gastric vein) usually (in 60% of cases) drains into the left side of the retropancreatic portal vein, but it may have a high termination well above the neck of the pancreas.

The close anatomic relationship of the splenic vein with the pancreas often leads to splenic vein occlusion in inflammatory or neoplastic diseases involving the body

and tail of the gland. Retrograde venous drainage toward the splenic hilum and then by way of the short gastric veins and the left gastroepiploic vein creates the syndrome of left-sided portal hypertension and gastric varices.

LYMPHATIC DRAINAGE

The lymphatic vessels of the pancreas conform to the general pattern of deep lymphatic drainage and accompany the arterial supply (Fig. 93-5; see also Color Fig. 93-5). The duodenum and head of the pancreas have a common lymphatic drainage; lymph from the foregut and midgut structures, including the pancreas, liver, stomach, spleen, small bowel, and proximal large bowel, eventually flows into the celiac and superior mesenteric groups of paraaortic nodes and into the cisterna chyli. [6] The lymphatics of the tail of the pancreas pass to the splenic nodes at the hilum of the spleen, and those of the body of the pancreas pass upward to the pancreaticosplenic nodes lying along the superior border of the gland. These nodes, along with the retropancreatic nodes, drain into the celiac nodes. Lymphatics of the upper anterior part of the head of the pancreas pass through the subpyloric nodes lying behind the first part of the duodenum. Obstruction of these pancreatic lymphatic drainage pathways by tumor may result in the shunting of lymph through local collateral channels, resulting in the involvement of nodes primarily concerned with hepatic or gastric drainage. Inferiorly, the retropancreatic and antepancreatic group of nodes drain into the superior mesenteric nodes, and lymph from the latter also may pass into nodes in the root of the transverse mesocolon. The absence of fascial or retroperitoneal coverings on the posterior aspect of the pancreas allows easy communication between lymphatics of the pancreas and those of neighboring retroperitoneal tissues and organs. The lymphatic network is so rich that a thorough lymphadenectomy en bloc with a pancreatic resection frequently produces a profuse postoperative chylous leakage. [7]

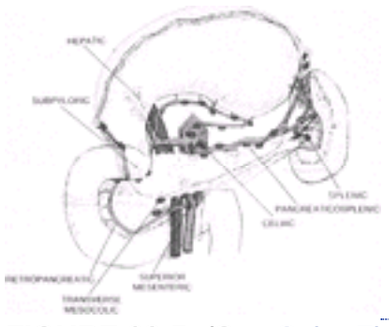

FIGURE 93-5. (See Color Figure 93-5.) Lymphatic drainage of the pancreas. The pancreas is viewed from its anterior aspect. The gastrocolic ligament has been divided along the greater curvature of the stomach, which has been retracted anterosuperiorly. The transverse mesocolon has been detached from the peritoneum of the posterior abdominal wall. Labels indicate representative lymph nodes in the major regional nodal groups.

NERVE SUPPLY

The sympathetic efferent innervation of the pancreas is derived from the greater, lesser, and least splanchnic nerves. The bodies of the preganglionic sympathetic neurons originate in the lateral gray matter of thoracic spinal segments 5 through 10. After transversing the sympathetic trunks, presynaptic nerve fibers synapse with postganglionic sympathetic neurons within the celiac plexus, although there is some minor distribution to the pancreas through the hepatic and superior mesenteric plexuses. The celiac ganglion consists of two masses that lie on either side of the aorta, anterior to the crura of the diaphragm and close to the adrenal glands. The right celiac ganglion is partly covered by the inferior vena cava, and the left celiac ganglion is covered by the peritoneum of the lesser sac close to the upper border of the pancreas. The cell bodies of afferent sympathetic neurons are located in dorsal root ganglia. Afferent fibers often cross the midline in celiac ganglia before projecting centrally so that sympathetic afferent innervation is bilateral.

The parasympathetic innervation of the pancreas is derived from the vagal nerves. The cell bodies for efferent vagal fibers are located in the medulla in the dorsal motor nucleus. Efferent fibers pass to the pancreas by way of the celiac division of the posterior vagal trunk. No synaptic connections are made within the celiac ganglia; postsynaptic neurons are located within the pancreatic parenchyma. Afferent vagal fibers also pass through the celiac ganglia; afferent cell bodies are located within the nucleus ambiguus. [8]

The ultrastructure of pancreatic innervation has been studied extensively in several animal species in addition to humans. Myelinated fibers usually are not found within the parenchyma. Intrapancreatic ganglion cells are seen within the interlobular tissues, with nonmyelinated fibers passing to both exocrine and endocrine portions of the gland. The sites of nerve termination may be generally grouped as blood vessels, pancreatic acinar cells, ductal cells, and pancreatic islets. Functional correlates suggest that pancreatic nerves may modulate the function of each of these pancreatic elements. In addition to the classic cholinergic and adrenergic neurons, evidence suggests that a large number of peptide-containing neurons also exist within the pancreas. Immunocytochemical methods have demonstrated the presence of fibers or cell bodies containing vasoactive intestinal polypeptide, substance P, cholecystokinin-8, gastrin-releasing peptide, enkephalin, galanin, neuropeptide Y, and calcitonin gene-related peptide. [9], [10] The physiological actions of many of these peptidergic nerves within the pancreas remain to be defined, but anatomic studies suggest that modulation of release of acetylcholine and catecholamines from autonomic nerve terminals within the pancreas may be important. Regulation of pancreatic secretion and blood flow is also an important function of intrapancreatic neurons.

Pain fibers from the pancreas travel through the celiac ganglia and by way of the sympathetic splanchnic nerves and the thoracic sympathetic chain to reach the spinal root ganglia. Pain from the head of the pancreas tends to be broadly localized in the midepigastrium, and pain from the body and tail tends to be localized in the left upper quadrant. Visceral pain from the pancreas usually is sensed as a severe, constant discomfort in the epigastrium. Because the pancreas does not contact the somatically innervated parietal peritoneum, sharply localized pain usually does not occur. Radiation of pancreatic pain to the back in the area of the lower thoracic vertebrae is common.

Recent studies suggest that pancreatic innervation may be altered in disease states such as chronic pancreatitis. In patients with chronic pancreatitis, the number and diameter of intralobular and interlobular nerve bundles have been reported to be increased relative to normal numbers. Pancreatic nerves in these patients demonstrated increased immunostaining for substance P and calcitonin gene-related peptide. [11] Because these peptides are known to be expressed in afferent neurons, the changes have been postulated to be responsible for the pain syndrome associated with chronic pancreatitis. Alterations in peptidergic pancreatic innervation also have been reported in animal models of chronic pancreatitis. [12] The cellular mechanisms responsible for these abnormalities are unknown; interactions of nerves with pancreatic inflammatory cells have been proposed as a cause. [13]

DUCTAL SYSTEM

The main pancreatic duct of Wirsung extends from the tail of the pancreas to the major duodenal papilla or ampulla of Vater (Fig. 93-6 and Fig. 93-7). The average diameter of the duct in the adult tapers from 4 to 2 mm, and it is widest in the head of the gland. [14] The main duct is close and almost parallel to the distal common bile duct for 2 to 3 mm before combining to form a common duct channel before opening into the duodenum. The accessory pancreatic duct of Santorini, which is present in 40% to 70% of people, usually communicates with the main duct and passes transversely to the right in the upper part of the head of the pancreas. The duct of Santorini lies anterior to the intrapancreatic common bile duct and usually opens into the proximal portion of the second part of the duodenum at the minor papilla, proximal to the ampulla of Vater.

FIGURE 93-6. Terminology variously applied to describe the pancreatic duct system. One system indicates an understanding of ductal embryology, particularly with

regard to the development of the more unusual variations (**left**). For clarity and practicality, however, the terms given on the right are preferred.

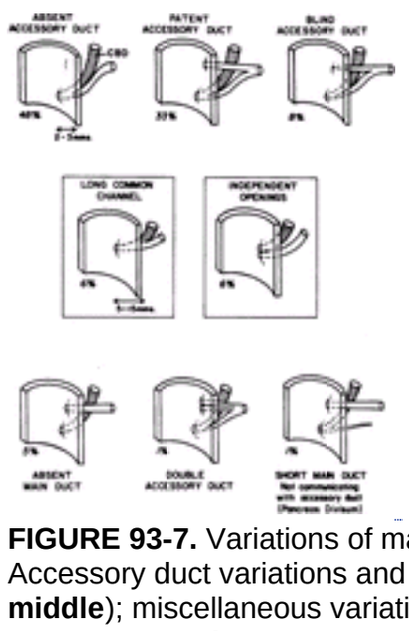

FIGURE 93-7. Variations of main and accessory pancreatic ducts, and their relation to the common bile duct (CBD) from a series of 143 postmortem preparations. Accessory duct variations and usual short common channel for CBD and main pancreatic duct (**top**); variations of CBD and main pancreatic duct terminations (**middle**); miscellaneous variations (**bottom**). (Adapted from Berman LG, Prior JT, Abramow SM, et al. A study of the pancreatic duct system in man by use of vinyl acetate casts of post mortem preparations. Surg Gynecol Obstet 1969;110:391.)

This “typical” ductal anatomy actually may be present in as few as 33% of people. [15] Important variations include nonpatency of the accessory duct (8%), independent openings of the common bile duct and the main pancreatic duct (6%), an absent main duct (5%), and patent double accessory ducts (1%). Noncommunication between the main duct and the accessory duct, with the body and tail of the gland draining exclusively by way of the duct of Santorini, results in the anatomic variant known as *pancreas divisum,* which occurs in 2% to 6% of otherwise healthy people.

The sphincter of Oddi consists of circular smooth muscle that surrounds the common channel of the common bile duct and the main pancreatic duct at the ampulla of Vater. The muscle fibers of the sphincter of Oddi extend around the common bile duct just distal to the latter’s oblique entry into the wall of the duodenum to form the choledochal sphincter. A short region of circular smooth muscle also surrounds the pancreatic duct just before its termination to form the pancreatic duct sphincter.

ULTRASTRUCTURE

The pancreas is a mixed endocrine and exocrine gland. The glandular constituents by weight are 80% exocrine tissue, 18% ductular system, and 2% endocrine tissue. The pancreas is a lobulated organ with lobular subunits composed of acini. The acini are rounded or have a short tubular form and consist of single rows of epithelial cells lying on a basal lamina. Lining the lumen of the acinus are pyramidal acinar cells and pale-staining centroacinar cells, which are unique to the pancreas. The acinar lumen connects with the intralobular ducts to form the interlobular ducts, which, in turn, coalesce to form the main pancreatic duct. Lining the ductules are columnar cells, goblet cells, and occasional argentaffin cells. Thick layers of connective tissue and elastic fibers surround the large ducts.

In the resting state, the basal portion of the acinar cell contains a centrally located spherical nucleus lying within a highly basophilic cytoplasm secondary to the large number of ribosomes in the rough endoplasmic reticulum. The abundant ribosomes attest to the high protein-synthesizing capacity of the acinar cells. A clear region containing the Golgi complex separates the nucleus from numerous eosinophilic zymogen granules, each about 1 µm in diameter, lying at the apex. The acinar cell has short microvilli, averaging 0.2 µm in length, which protrude into the acinar lumen. At the apical portion, the cells are held together by tight junctions, which prevent reflux of lumenal contents. Laterally, the cells are connected by gap junctions, which permit intercellular communication.

The thin basal lamina on which the acini rest is supported by collagen fibers. A rich capillary plexus surrounds the acinus in this connective tissue and is penetrated by numerous nerve fibers, which reach the acinar cells. [16]

The endocrine portion of the pancreas consists of about one million islets of Langerhans, which are distributed throughout the gland but are relatively concentrated in the tail of the pancreas. The islets are about 200 µm in diameter, and each is associated with a prominent capillary plexus. The islets contain several cell types. About 75% to 80% of islet cells are beta or B cells, which secrete insulin, and 10% to 20% are alpha or A cells, which contain glucagon. Delta or D cells constitute about 5% of the islets and contain somatostatin. [17] The B cells occupy the center of the islets, whereas the perimeter is lined by A cells. D cells are dispersed between these groups. Other peptide-secreting cells that may be associated with the islets include enterochromaffin (EC) cells containing 5-hydroxytryptamine and pancreatic polypeptide (PP) cells containing pancreatic polypeptide.

CONGENITAL ANOMALIES

Agenesis or Hypoplasia of the Pancreas

Agenesis of the pancreas is a rare and, at one time, universally fatal condition. [18] Failure of the pancreas to develop may occur as an isolated anomaly, or it may be associated with other congenital defects, such as absence of the gallbladder. [19] Although the cause is not known, pancreatic agenesis has been associated with retarded intrauterine growth, presumably resulting from failure of the endocrine pancreas to produce insulin. [20] Partial agenesis of the pancreas results from incomplete formation of either the dorsal or ventral pancreas and has a more favorable outcome than complete agenesis. Involvement of the dorsal segment appears to be more common than involvement of the ventral pancreas. These glands possess normal exocrine and endocrine function.

Hypoplasia of the pancreas is a congenital disease involving exclusively the exocrine pancreas and has been referred to as *lipomatous pseudohypertrophy of the pancreas*. [21] Pathologically, the gland appears enlarged and of normal shape but of a fatty consistency. The major pancreatic ducts are developed, and islets of Langerhans are present; however, secondary pancreatic ducts and acinar lobules are absent or underdeveloped, and fatty tissue replaces normal acinar cells. [22] The finding that the gland is of normal shape, but missing differentiated cellular components, led investigators to postulate that embryological development is normal in this condition. It is proposed that an intrauterine insult occurs, such as infection, causing hypoplasia of the exocrine gland. [23] Although this disease usually is diagnosed in infants, it has been reported to occur in adults. [21], [22] Manifestations result from severe pancreatic exocrine insufficiency.

Annular Pancreas

Annular pancreas is an unusual complication of disturbed embryological development in which the head of the pancreas surrounds the duodenum, often resulting in duodenal obstruction. Annular pancreas frequently is associated with other congenital defects, including Down syndrome, Meckel diverticulum, malrotation of the intestine, duodenal atresia and bands, intestinal webs, tracheoesophageal fistulae, imperforate anus, absence of the gallbladder, and certain types of cardiac defects. [24], [25], [26], [27] and [28] Annular pancreas has been described in association with pancreas divisum (see section “ Pancreas Divisum”). [29] Men appear to be affected more commonly than women. The occurrence of annular pancreas is usually sporadic, although several reports describe a familial association and apparent autosomal dominant transmission. [30], [31] and [32]

Typically, the annulus is a band of pancreatic tissue completely encircling the second portion of the duodenum (Fig. 93-8). The ring usually lies proximal to the major papilla, and in a few cases the annulus involves the first or third portion of the duodenum. Histologically, pancreatic tissue frequently invades the muscularis layer of the duodenum. [33]

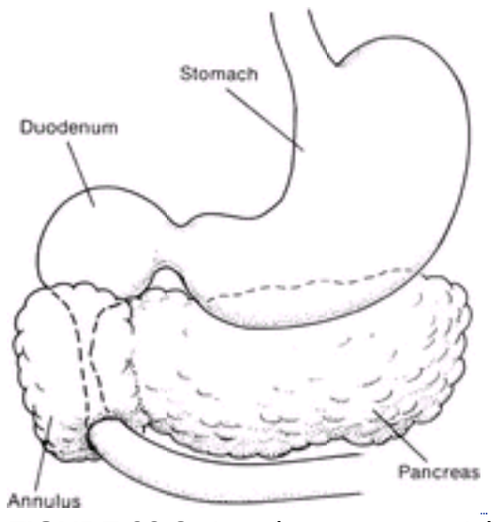

FIGURE 93-8. Annular pancreas. A band of pancreatic tissue surrounds the midportion of the duodenum.

The most popular etiologic theory suggests that the ventral pancreatic bud becomes fixed; as the pancreas and duodenum rotate, a band of pancreatic tissue is left encircling the duodenum. [34] Supporting this theory is the report that a high concentration of pancreatic polypeptide cells, which is characteristic of the ventral pancreas, was found in annular tissue. [35] Other theories propose that hypertrophy of the dorsal and ventral ducts or ectopic pancreatic tissue causes the annulus. [36]

More than half of all cases are diagnosed in the first year of life. Severe duodenal stenosis is usually apparent within the first few days of birth. Because pancreatic tissue encircles the duodenum, causing obstruction, newborns and infants are intolerant of oral feedings and have vomiting, often of bilious material. Upper abdominal distention and visible peristalsis are often present on physical examination. [37] Although vomiting is the usual presenting symptom in children, adults often complain of postprandial colicky abdominal pain, bloating, and fullness that also may be associated with nausea and vomiting. [38], [39], [40] and [41] Upper gastrointestinal bleeding and duodenal ulcer disease occur in one third of adults, and acute pancreatitis is a common associated finding in this disorder.

Radiographic signs of annular pancreas are those of duodenal obstruction. Plain radiographs of the abdomen often show a “double-bubble” sign resulting from dilation of the duodenum and stomach. With high degrees of obstruction, a duodenal cutoff sign also may be seen. In infants, these findings are the only necessary radiographic tests. Radiographic findings are not specific in older children and adults, and it is usually necessary to document duodenal obstruction by upper gastrointestinal contrast studies. Constricting bands, 0.8 to 5 cm long, around the duodenum have been detected radiographically. [42] Other signs include dilation and reverse peristalsis of the proximal duodenum. Endoscopic evaluation of annular pancreas usually is not helpful. The mucosa usually is normal, and it is difficult to appreciate mild narrowing of the duodenum. In severe cases, concentric narrowing of the duodenum or stenosis and associated peptic ulcer disease are suggestive of the diagnosis. Endoscopic retrograde cholangiopancreatography (ERCP) has been used to visualize the ductal system in annular pancreas and is diagnostic of the disorder. [43], [44], [45] and [46] Magnetic resonance (MR) pancreatography is being used with increasing frequency as a noninvasive alternative to diagnostic ERCP in the evaluation of pathological conditions of the pancreas such as annular pancreas and pancreas divisum. [47] Characteristic features can also be determined by computed tomography. [48] In symptomatic patients, treatment of annular pancreas is surgical alleviation of the obstruction. Division of the annulus is not recommended because of the high incidence of pancreatitis and pancreatic fistulae complicating that procedure, and bypass of the obstructed intestinal segment is preferred. [49]

Heterotopic Pancreas

Aberrant localization of pancreatic tissue, also known as heterotopic pancreas or pancreatic rests, refers to segments of pancreatic tissue not in continuity with the main body of the pancreas. The condition is usually asymptomatic, and therefore its true incidence is difficult to determine. Heterotopic pancreas has been identified in one of every 500 laparotomies, and autopsy studies have reported frequencies ranging from 0.6% to 15%. [50], [51] Seventy percent of such rests are found along the upper gastrointestinal tract, with the stomach (25.5%), duodenum (27.7%), and jejunum (15.9%) representing the most frequent locations. Other intra-abdominal locations include the gallbladder, liver, small intestine, colon, appendix, omentum, and Meckel diverticulum. [52], [53] and [54] In the stomach, pancreatic rests are found primarily in the prepyloric region along the greater curvature. Extra-abdominal sites include the lung and umbilicus. [55], [56] and [57] Histologically, heterotopic pancreas can contain acinar tissue, islets, ducts, or any combination thereof. Seventy-five percent of rests are located in the submucosa of the stomach or intestine and appear as firm, yellow nodules 2 to 4 cm in diameter underlying the mucosa. These often have a central mucosal depression, thought to represent the presence of a vestigial duct, which may be recognized endoscopically or by radiographic contrast studies. The origin of heterotopic pancreas is not known, and it is likely that several abnormalities of embryological development account for the locations in various sites. As pluripotential cells, pancreatic rests may represent abnormal differentiation of endodermal stem cells. Conversely, disturbances in migration of the pancreas as it rotates around the gut may account for some heterotopic locations. [58]

Heterotopic pancreas is usually an asymptomatic condition that is an incidental finding at surgery or autopsy. Symptoms attributed to this disorder include abdominal pain, nausea, vomiting, and gastric outlet obstruction. [59], [60] Peptic ulcer disease and upper gastrointestinal bleeding also have been described. Because of the varied locations of pancreatic rests, involvement of other organs can occur. Pancreatitis, biliary obstruction with jaundice, intestinal obstruction, and intussusception all have been associated with heterotopic pancreas. [61], [62], [63], [64], [65], [66] and [67] As a result of the combined exocrine and endocrine function of the pancreas, any pathological change in the normal pancreas also can occur in ectopic tissue. [68] Therefore, malignant degeneration, cyst formation, and islet cell tumors also may be found in pancreatic rests. [69], [70], [71], [72] and [73]

Pancreatic rests involving the upper gastrointestinal tract often are detected initially by contrast radiography or endoscopy by the presence of a submucosal bulge with central umbilication. [74], [75] These lesions are covered by normal mucosa, making routine endoscopic biopsy impossible. Heterotopic pancreas may be difficult to differentiate from leiomyomas, fibromas, carcinoid tumors, or other malignant tumors. Over the last several years, endoscopic ultrasound (EUS) has become an increasingly useful modality in the evaluation of these submucosal gastrointestinal masses. EUS not only can distinguish between an intramural lesion and compression from an extralumenal mass, but it also allows identification of the wall layer from which the mass originates. For example, pancreatic rests, located in the submucosal layer, can be differentiated from leiomyomas, the most commonly encountered submucosal masses, which are located in the layer of the muscularis propria. [76] In other cases in which a definitive diagnosis cannot be made based on EUS appearance, EUS-guided fine-needle aspiration may be used to obtain a histological diagnosis. [77]

Treatment of ectopic pancreatic tissue is indicated only for those who have significant symptoms or complications, such as recurrent upper gastrointestinal bleeding, biliary or intestinal obstruction, or malignant degeneration. Definitive treatment is surgical removal of the ectopic tissue. Asymptomatic people with incidental discovery of heterotopic pancreas do not require further evaluation or treatment. [68] If unsuspected pancreatic rests are found at surgery, excision prevents the possible complication of malignant degeneration and eliminates confusion if subsequent symptoms develop. [68], [78]

Pancreas Divisum

Pancreas divisum, the most common congenital anomaly of the pancreas, is caused by failure of the ducts of the dorsal and ventral anlagen to fuse during the fifth and sixth weeks of gestation. Normally, the proximal one third of the dorsal pancreatic duct regresses as it fuses with the ventral duct, forming the main pancreatic duct. In pancreas divisum, the ventral duct of Wirsung empties into the duodenum through the major papilla but drains only a small portion of the pancreas. Secretions from the tail, body, neck, and remainder of the head of the pancreas drain into the duodenum through the minor papilla by way of a persistent duct of Santorini. In 2% to 3% of cases in which the ducts do not fuse, the ventral duct may not be demonstrable. Normally, drainage of the pancreas can occur by either duct, depending on fusion of the two ductal systems and the degree of patency of each. Most pancreatic drainage is through the duct of Wirsung, and although some drainage occurs through the duct of Santorini, this volume is relatively small. Occasionally, the duct of Santorini ends blindly in the duodenal wall.

Although pancreas divisum is a long-recognized entity, only with the advent of ERCP has its clinical significance become apparent. In autopsy series, pancreas divisum has an incidence of 5% to 10%; [79] as an ERCP finding, its incidence is 4%. [80], [81] and [82] In a series of patients with pancreatitis, however, there was a 16% incidence of pancreas divisum, and the incidence of the abnormality increased to 25% in idiopathic pancreatitis. [80] Therefore, it appears that pancreas divisum is associated with pancreatitis. It has been postulated that pancreatitis may result from a combination of pancreas divisum and stenosis at the level of the accessory papilla, with impediment of pancreatic secretory flow. As a congenital defect, clinical manifestations of pancreas divisum may occur at any age but are uncommon in childhood. Symptoms may be mild with only occasional epigastric pain occurring postprandially, but more often episodes of severe acute pancreatitis occur. Chronic pancreatitis also may develop, with all of its associated sequelae. Changes in the pancreatic ducts characteristic of chronic pancreatitis may be detected on ERCP.

Pancreas divisum is diagnosed by pancreatography (Fig. 93-9). On ERCP, the major papilla is often difficult to cannulate, but on injection with contrast fluid, the duct of Wirsung appears shortened and of small diameter. There is rapid filling of small accessory ducts, and too much contrast can be injected before it is realized that the duct of Wirsung is not in communication with the main pancreatic duct. As a result, the patient may experience sudden pain, and pancreatitis of the ventral pancreas can develop. What appears as an abrupt cutoff of the duct of Wirsung must not be confused with a mass lesion such as a malignancy or pancreatic pseudocyst. The delicate nature on the pancreatogram of the accessory ducts is a helpful indicator of pancreas divisum. If possible, the accessory papilla should be cannulated, which should reveal a duct of Santorini running the entire length of the pancreas that is not in communication with the duct of Wirsung. While ERCP remains the gold standard for diagnosis of pancreas divisum, MR pancreatography is an emerging, noninvasive radiographic technique that is being used to define ductal anatomy (see " Annular Pancreas"). If stenosis of the papilla is not demonstrated radiographically, pancreatic manometry and secretin ultrasound scanning may be useful in identifying patients who may benefit from endoscopic or surgical intervention. [83], [84]

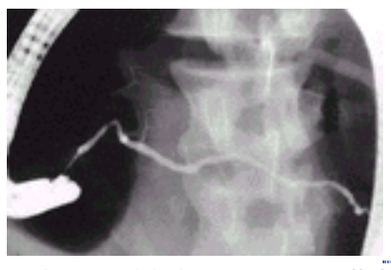

FIGURE 93-9. Pancreas divisum. An endoscopic retrograde pancreatogram performed through the accessory papilla shows the dorsal duct in pancreas divisum. (Courtesy of Peter B. Cotton, M.D., Durham, NC.)

Patients with mild symptoms can be managed conservatively. Patients with recurrent episodes of acute pancreatitis or chronic pain require intervention to alleviate accessory ductal stenosis. Surgical accessory duct sphincteroplasty has been successful in treating patients, with the best results observed in patients with recurrent episodes of acute pancreatitis. [85], [86] and [87] Endoscopic therapy with a combination of minor duct sphincterotomy and stenting has achieved similar rates of success; however, the follow-up period in these patients has been less than in the surgical groups, [88], [89] and [90] and long-term stenting of the pancreatic duct has been reported to result in chronic ductal changes. [91] Determination of which patients with acute recurrent pancreatitis would be served best by surgical or endoscopic treatment awaits further studies. Patients with chronic pancreatitis have a poor response to both surgical and endoscopic treatment modalities. In cases in which sphincterotomy has failed or the ductal involvement is more extensive, direct ductal drainage or resection of the involved pancreas is necessary. [92], [93]

Congenital Cysts

Cysts of the pancreas are distinguished from more common pseudocysts by the presence of an epithelial lining. Pseudocysts often follow bouts of acute pancreatitis, with trauma being a major precipitant. Identification of true columnar or cuboidal epithelium in congenital cysts may be difficult, and it is not always possible to distinguish a congenital cyst from a pancreatic pseudocyst. In cysts located in the peripancreatic space, identification of pancreatic enzymes in the cyst fluid is helpful in determining that it is of pancreatic origin; however, not all pancreatic cysts contain fluid rich in pancreatic enzymes. Solitary congenital cysts are rare. [94] They are more commonly diagnosed in early childhood but may remain asymptomatic and undetected into adulthood. Although other congenital anomalies have been reported with pancreatic cysts, these are more often isolated defects. [95]

The most common presentation of a solitary pancreatic cyst is as an abdominal mass, which may be associated with abdominal pain or complications resulting from expanding size, such as gastroduodenal obstruction. Encroachment on the intrapancreatic or extrapancreatic common bile duct may cause biliary obstruction. [96] Subcutaneous fat necrosis and osteolytic lesions secondary to pancreatic enzymes leaking into the circulation from a developmental pancreatic cyst have been described. [97]

A plain radiograph of the abdomen or an upper gastrointestinal radiographic series demonstrating a mass displacing a portion of the stomach, duodenum, or colon may suggest the presence of a pancreatic cyst. The best diagnostic test is computed tomography, which demonstrates a fluid-filled cystic lesion in the pancreas and its relationship to surrounding structures. In considering the differential diagnosis, it is important to consider other possible cystic lesions of the pancreas, including pancreatic pseudocysts, cystadenomas, and cystadenocarcinoma.

Cystic neoplasms of the pancreas are rare, representing about 10% of pancreatic cysts and 1% of pancreatic carcinomas. [98] Abdominal or back pain, the most common symptom, is present in about half of patients. One third of patients have an abdominal mass or weight loss. One third of patients are asymptomatic, with the cyst discovered during evaluation of other complaints. There is a clear predilection for women, with a female-to-male ratio of 8:1. Computed tomography is the most useful diagnostic modality and typically reveals a multiloculated lesion, 3 to 6 cm large, located in any region of the pancreas. Two major histological variants exist: serous cystadenomas and mucinous cystic neoplasms. Mucinous cystic neoplasms of the pancreas have clear malignant potential. Serous cystadenomas, in contrast, are almost invariably benign. Preliminary experience with biochemical and cytologic analysis of cyst fluid to distinguish serous and mucinous neoplasms has been reported. [99] The differentiation of congenital cysts, acquired pseudocysts, and cystic neoplasms is crucial. The operative therapy for chronic pseudocysts is nonresectional, whereas therapy for cystic neoplasms requires resection. [100]

Symptomatic congenital cysts should be removed whenever possible, which can be done by enucleation of the cyst if it is located in the body or head of the pancreas or by resection of the cyst with adjacent pancreatic tissue if it is confined to the distal pancreas. If the location of the cyst precludes either of these approaches, such as a cyst in the head of the pancreas, drainage into the stomach or jejunum can be performed. With surgical drainage of the cyst, pathological examination should be done to exclude malignancy.

Multiple Cysts

Multiple congenital pancreatic cysts are rare and usually are associated with other congenital anomalies. In particular, polycystic kidney disease and cystic fibrosis are frequently associated with pancreatic cysts. Other clinical syndromes involving multiple pancreatic cysts include von Hippel-Lindau syndrome, Ivemark syndrome, and Bruber syndrome, in which cysts of the lung, liver, and central nervous system also are found. [101], [102] Many of the associated anomalies are lethal, although patients may have no symptoms referable to the pancreatic cysts. Because of the numerous cysts and the involvement of several organ systems, no treatment is necessary unless specific symptoms or a particular complication, such as infection of a cyst, dictates surgical excision or drainage.

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CHAPTER 94

Mark Topazian and Fred S. Gorelick

ACUTE PANCREATITIS

Acute pancreatitis is a clinical syndrome defined by a discrete episode of abdominal pain and elevations in serum enzyme levels. There is inflammation of the pancreas with varying amounts of injury to adjacent and distant organs. In the United States, more than 80% of the cases are related to biliary stones or alcohol use. Although there are many factors that may precipitate acute pancreatitis, it is rare for any but alcohol, hyperlipidemia, or genetic disorders to lead to chronic pancreatitis.

The first reports of pancreatitis may date to the 1700s, but the first systematic analysis of pancreatitis was published by Reginald Fitz in 1889. [1] In this landmark study, the clinical and pathological findings in 53 patients were presented. Fitz's analysis of the patterns of abdominal pain, fever, and jaundice, along with physical findings, established the criteria for diagnosing pancreatitis. Etiologic factors such as gallstones, alcohol, penetrating ulcers, and trauma were mentioned. Pancreatic pseudocyst, splenic vein thrombosis, disseminated fat necrosis, and abscess were also described. Fitz's report established the framework for studies and treatments of pancreatitis that have spanned more than a hundred years.

INCIDENCE

The incidence of acute pancreatitis ranges from 1 to 5 per 10,000 per year. This value is approximate and may vary widely among populations for several reasons. First, histological confirmation of pancreatitis is unavailable in most patients and the diagnosis must rely on clinical assessment, biochemical tests, and radiologic evaluations, which are limited in their sensitivity and specificity. Second, the incidence of precipitating factors such as alcoholism and gallstones varies among populations. Finally, some patient groups may be systematically excluded; for example, those with mild disease may not come to a hospital.

CLASSIFICATION

Pathology, etiology, and clinical presentation and course may all be used to classify acute pancreatitis. The existence of various groupings reflects the different goals of such classifications. Some are useful in establishing a differential diagnosis or understanding disease mechanisms, and others may provide prognostic information or guide intervention.

A sensible classification system based on a number of clinical parameters has been described by Bradley. [2] The most important feature of this classification is that it defines a common nomenclature for describing acute pancreatitis. It separates pancreatitis into mild and severe disease based on physiological findings, laboratory values, and diagnostic imaging tests, which are discussed in this chapter. Mild pancreatitis is not associated with organ dysfunction or complications, and recovery is usually uneventful. Severe pancreatitis is associated with impaired pancreatic function, local and systemic complications, a complicated recovery, and significant mortality rates. Bradley's classification makes several important distinctions by defining pancreatic fluid collections, pseudocysts, and necrosis, and separating infected pancreatic necrosis from pancreatic abscesses. This classification does not provide the criteria for establishing prognosis early in the patient's course, as discussed later in this chapter (see section " Clinical and Physiological Assessment "), but does provide a clear framework for discussing acute pancreatitis using a

defined set of terms.

PATHOLOGY

Detailed histological studies of pancreatic tissue are available from a limited number of cases of human acute pancreatitis. A histological spectrum of acute pancreatitis is recognized ranging from mild, interstitial disease to coagulation necrosis. [3] Interstitial pancreatitis may lead to local and systemic complications but is rarely fatal; necrotizing pancreatitis may be fatal in up to 30% of cases.

Interstitial

In interstitial pancreatitis the gland is edematous, but its gross architecture is preserved. Parenchymal inflammatory cells are present together with interstitial edema. Disruption of the normal acinar cell architecture is common and may contribute to the reduced enzyme secretion characteristic of acute pancreatitis. Zymogen granules are displaced from their fusion site in the apical domain of the cell and become dispersed throughout the cell, and the apical membrane appears contracted and microvilli disappear. [4] Zymogen granules fuse with each other instead of the apical membrane. Similar to animal models of pancreatitis, a distinct form of cell necrosis is observed in which the apical domain of the acinar cell is shed into the lumen, resulting in intact zymogen granules within the lumen. This pattern of partial cell necrosis may allow the acinus to regenerate rapidly after injury.

Necrotizing

Macroscopically, marked tissue necrosis and hemorrhage are apparent. Surrounding areas of fat necrosis are also prominent. These chalky areas of dead adipose tissue are found within the peripancreatic tissue and throughout the abdomen. Large hematomas often are located in the retroperitoneal space. The microscopic appearance of the pancreas parallels the gross changes, with marked fat and pancreatic necrosis. Vascular inflammation and thrombosis are common.

PATHOPHYSIOLOGY

Pancreatitis is triggered by a variety of mechanisms, including pancreatic duct obstruction, pancreatic ischemia, and toxic, immunologic, or metabolic factors. These various insults activate three major pathological processes within the acinar cell that initiate pancreatic injury (Fig. 94-1). First, inactive digestive zymogens are converted into active enzymes. Second, pancreatic exocrine secretion is inhibited and third, the pancreas generates proinflammatory mediators. The relative importance and sequence of these processes may vary with the clinical setting. Once injury has been initiated, it is perpetuated and amplified by other processes, including the inflammatory cascade and vascular damage. The importance of thinking about the steps in acute pancreatitis, especially the latter ones such as the release of damaging cytokines, neutrophil recruitment, platelet activation, and formation of free radicals, lies in the potential to target future therapeutic interventions.

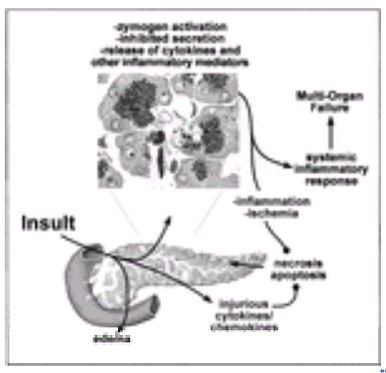

FIGURE 94-1. Initiation of acute pancreatitis. Various forms of insults initiate a cascade of events that result in acute pancreatitis. The pancreatic acinar cell responds to a variety of insults with activation of zymogens, inhibition of secretion, and release of proinflammatory substances. Inflammatory cells are activated. Expression of adhesion molecules promotes retention of inflammatory cells within the pancreas. Pancreatic blood flow may decrease because of capillary leaks, compression by edema, and vascular damage and occlusion. Toxic factors produced by pancreatic cells and inflammatory cells cause pancreatic cell death. Generalized activation of inflammatory cells may lead to the systemic inflammatory response syndrome and multisystem organ failure.

Activation of Pancreatic Zymogens

Active pancreatic enzymes have been detected within the pancreas in experimental and clinical pancreatitis. Further, a marker of trypsinogen activation, the trypsinogen activation peptide (TAP) increases within 1 hour of the onset of experimental pancreatitis. [5] A mutation in the trypsinogen molecule that is predicted to amplify trypsin activity causes at least one form of familial pancreatitis. [6] Further, mutations in the pancreatic trypsin inhibitor (SPINK 1) appear to predispose to developing pancreatitis. [7], [8] Finally, pretreatment with the protease inhibitor gabexate may reduce endoscopic retrograde cholangiopancreatography (ERCP)-induced pancreatitis. [9] Premature activation of zymogens may be the first step in a process that leads to pancreatic autodigestion and induces other pathways that lead to pancreatic injury.

Although premature zymogen activation may take place within pancreatic ducts or the interstitial space, the earliest activation site is within the acinar cell. Treatment of isolated pancreatic acini with concentrations of cholecystokinin (CCK) that generate pancreatitis in vivo leads to intracellular zymogen activation. [10] Two mechanisms have been proposed to account for intracellular activation of trypsinogen and the zymogen cascade: trypsinogen autoactivation, and trypsinogen activation by the lysosomal enzyme cathepsin B. The contribution of each potential pathway to zymogen activation remains unclear. However, the finding that genetically deleting cathepsin B from mice dramatically reduces trypsin activation in a model of pancreatitis suggests that cathepsin B plays a major role. [11] Although the site and cellular mechanism of the activation remain unclear, pathological increases in intracellular calcium may be required for zymogen activation. Alcohol, a common cause of pancreatitis, has been shown to sensitize the acinar cell to the enzyme-activating effects of CCK and to stimulate the release of CCK from the small intestine.

The potential consequences of zymogen activation are manifold, and include damaging local effects, attack on other tissues, and promotion or activation of additional pathways leading to tissue injury. Once released from the acinar cell, active digestive enzymes undoubtedly attack normal tissue structures; however, potent enzyme inhibitors are present in circulation and play an important role in limiting tissue damage. The major serum protease inhibitors include a2-macroglobulin, a1-antitrypsin, antichymotrypsin, serum trypsin inhibitor, and C1 esterase inhibitor. After these inhibitors bind to activated enzymes, the resulting complex is cleared by the reticuloendothelial system. Decreased amounts of circulating protease inhibitors and delayed clearance of serum protease-inhibitor complexes by the reticuloendothelial system have been associated with more severe forms of pancreatitis. [12]

Inhibition of Secretion

A feature common to several models of pancreatitis is the inhibition of enzyme secretion from the pancreas. Although increased paracellular permeability may contribute to this decrease, reduced acinar cell secretion from its apical membrane is likely to play a major role. Disruption of the acinar cell cytoskeleton may play a role in blocking secretion. [13] This inhibition leads to the retention of active enzymes within the acinar cell instead of their secretion into the pancreatic duct. The inhibition of secretion is likely to play a central role in the initiation of disease.

Generation of Inflammatory Mediators

Several major classes of inflammatory mediators are generated and released during acute pancreatitis. [14] These agents act by stimulating recruitment of inflammatory cells, enhancing the activation and adherence of inflammatory cells to the vascular wall, or cause direct cell injury. Soluble inflammatory mediators are known as

cytokines. Some, such as tumor necrosis factor-alpha (TNF-a) and platelet-activating factor (PAF), are generated by both the pancreatic acinar cells and inflammatory cells. Chemokines are a family of low-molecular-weight cytokines that attract particular subsets of inflammatory cells. For example, interleukin-8 (IL-8) attracts neutrophils and monocyte chemotactic protein-1 (MCP-1) attracts monocytes. Substance P is released from nerve endings and appears to mediate inflammation and particularly lung injury in experimental pancreatitis.

Substances that either directly injure acinar cells and endothelium or mediate the inflammatory response play an important role in both the early and late phases of acute pancreatitis. Higher circulating levels of proinflammatory cytokines are associated with more severe disease. Not all cytokines exhibit a harmful effect. Indeed, in one study but not another, inteleukin-10 (IL-10) has been shown to prevent ERCP-induced pancreatitis. [15], [16] The concept of treating clinical acute pancreatitis with cytokine inhibitors has promise, but has not yet succeeded in clinical trials.

Perpetuation of Disease

Inflammation Recruitment of inflammatory cells into pancreatic blood vessels and later into the pancreatic parenchyma is an early feature of acute pancreatitis. Increased expression of intercellular adhesion molecule-1 (ICAM-1) during pancreatitis leads to enhanced adhesion of activated neutrophils to the vascular endothelium. Trypsin activation of the alternate complement pathway may play a role in neutrophil recruitment. The influx of inflammatory cells may be so great as to form white cell emboli and plug small blood vessels. Activation and the influx of lymphocytes may also be an important component of injury in some forms of pancreatitis. Fibrinolysis and complement activation correlates with disease severity. Prominent generation of proinflammatory mediators can lead to the systemic inflammatory response syndrome (SIRS). Marked leukocyte activation can lead to distant organ injury and the development of multisystem organ failure. SIRS and associated multisystem organ failure is one of the two major causes of death from acute pancreatitis (the other being pancreatic infection).
Vascular Damage, Edema, and Ischemia Vascular damage and ischemia probably play an important role in many forms of acute pancreatitis. Kinins released by inflammatory cells make capillary walls permeable and promote tissue edema and fluid losses. Pancreatic edema may indirectly compromise pancreatic blood flow and increase tissue ischemia. Increased capillary permeability in other organs may contribute to organ failure. The pancreas and inflammatory cells release TNF-a, which mediates cell death, inflammation, and plays a major role in the development of shock. Oxygen free radicals also promote pancreatic injury and may be generated by a number of mechanisms. They are formed soon after acute ethanol ingestion by its oxidative metabolism. Free radicals may also be generated during vascular reperfusion and are released by neutrophils. Levels of free radicals correlate with both decreased pancreatic protein and nonprotein sulfhydryl groups and the severity of pancreatic injury. [17] Free radical damage may involve both abnormal peroxidation and depletion of thiol donors such as glutathione. Attempts to prevent pancreatic injury using free radical scavengers have been only partially successful in experimental models, and are largely untested clinically.

Protective Mechanisms

The acinar cell has a number of cellular mechanisms to limit the effects of zymogens activated within the acinar cell. Potentially active enzymes are packaged within a secretory granule membrane that is impermeable to proteins. Breakdown of the zymogen granule membrane may allow enzymes to be released into the cell. The pancreatic trypsin inhibitor is packaged together with trypsinogen in the secretory granule. Although the inhibitor is efficient, the zymogen granule contains quantities sufficient to inhibit only a small portion (10%–20%) of activated zymogen. Thus, large-scale zymogen activation might easily overwhelm the cellular protease inhibitors. Relative deficiencies of the pancreatic trypsin inhibitor have been found in some forms of recurrent pancreatitis. [18] In addition to trypsin inhibitor, proteases are also present in the zymogen granule that can degrade activated enzymes. Finally, active enzymes can be secreted from cells.

Experimental Models

Animal models of acute pancreatitis have been developed to aid in the understanding of the disease and evaluate potential therapy. Many models induce an injury that closely resembles interstitial pancreatitis, but models of severe necrotizing pancreatitis have also been established. A common shortcoming of animal studies that evaluate therapy for acute pancreatitis is timing of intervention; most treat at or before the onset of pancreatitis in a time frame that would not be clinically feasible.

Hyperstimulation One hundred years ago, Mouret [19] observed that excessive cholinergic stimulation was associated with pancreatic injury. Mouret speculated that the activation of trypsin might play an important role in this injury. In subsequent experimental models, infusions of the CCK analog, cerulein, were shown to generate pancreatic injury in a time- and dose-dependent manner. [4] Pathological changes within the first hour of hyperstimulation include inhibition of secretion into the pancreatic duct, pancreatic edema, and margination of neutrophils. Within 3 to 6 hours, inflammatory cells invade the interstitium and vacuoles form within acinar cells. Isolated pancreatic acini are often used to examine organ function at the cellular level. Recent studies have demonstrated that similar to in vivo models of pancreatitis, hyperstimulation of isolated acini by CCK causes intracellular zymogen activation. [10] Moreover, this treatment results in inhibition of secretion from the acinar cells and generation of proinflammatory mediators such as TNF. [20] The advantage of using this system is that it can be easily manipulated and examined without the confounding factors of hypoxemia and inflammation found in vivo. Thus, isolated acini appear to be a useful model for examining some of the earlier pathological events in acute pancreatitis.
Other Models A number of other models of experimental pancreatitis have been developed. It has been demonstrated that obstruction of the opossum pancreatic duct alone results in pancreatitis; [21] this model may be useful for examining the mechanism of gallstone pancreatitis. The diet-induced model is based on the observation by Lombardi and associates [22] that a choline-deficient diet supplemented with the synthetic amino acid analog of methionine, DL-ethionine, induced a fatal hemorrhagic pancreatitis in young female mice. One possibility raised by the diet model is that abnormal metabolites may contribute to pancreatitis under certain conditions such as refeeding.

SPECIFIC ETIOLOGIES

It is important to review the differential diagnosis of acute pancreatitis in each patient. Many patients have an easily identifiable and treatable cause such as hyperlipidemia or gallstones. Less common causes should be considered in those with no obvious precipitant, especially in patients with recurrent, unexplained bouts of pancreatitis. An overview of the prevalence of various etiologies is shown in Fig. 94-2.

FIGURE 94-2. Etiologies of acute pancreatitis.

Obstructive

Many forms of pancreatic duct obstruction have been associated with pancreatitis, including gallstones, pancreas divisum, sphincter of Oddi dysfunction or stenosis, ampullary or pancreatic tumors, duodenal diverticula, and parasites, clots, or foreign bodies lodged in the ampulla. Gallstones are the commonest cause of acute pancreatitis in many parts of the world.

Gallstones Although gallstones are etiologically linked to pancreatitis, this condition develops in only a small percentage of patients with gallstones. For example, in a cohort from Minnesota with gallstones, the incidence of acute pancreatitis was 0.17% per year. [23] The presence of gallstones, however, increases the relative risk for acute pancreatitis 14- to 35-fold in men and 12- to 25-fold in women. [24] Small gallstones, less than 5 mm in size, increase the risk of acute pancreatitis. A contributing factor that places men at greater risk may be larger cystic ducts favoring the migration of stones into the common duct. Anomalous union of the pancreatic and biliary ducts or a long common channel in the ampulla may also predispose to pancreatitis, possibly by promoting reflux of bile into the pancreatic duct. [25] Metabolic factors also may increase the risk for development of pancreatitis from gallstones. For example, underlying hyperlipidemia manifest by delayed chylomicron clearance may

make patients more susceptible to gallstone pancreatitis. [26] Many models have attempted to reproduce the pancreatitis generated by stones in the ampulla. A century ago, on the basis of autopsy findings, Opie proposed that a gallstone lodged in the papilla would obstruct the ampullary orifice and allow reflux of bile into the pancreatic duct via the common channel. Several observations have challenged that theory: patients without a common channel can have gallstone pancreatitis, [27] and retrograde flow of bile into the pancreatic duct at physiological pressures does not induce pancreatitis. [28] Although addition of increased intraductal pressure to bile infusion results in severe pancreatitis, pressures of such magnitude are not thought to occur under physiological conditions. Opie also proposed that pancreatic duct obstruction alone might be the mechanism by which stones cause pancreatitis. In general, simple obstruction of the pancreatic duct in animals has not generated pancreatitis. In the American opossum, however, pancreatic duct obstruction alone generates acute pancreatitis. [29] This supports the "ductal obstruction theory," as does the occurrence of human pancreatitis in other forms of pancreatic duct obstruction. It seems likely that most biliary pancreatitis is precipitated by the transient or persistent obstruction of the papilla by a stone. When patients are selected based on hyperbilirubinemia and pancreatitis, gallstone recovery from the stool has been reported in 30% to 85%. [30] Thus, pancreatitis is usually accompanied by passage of a stone into the lumen of the intestinal tract. Stone passage in most patients with gallstone pancreatitis probably occurs on the day of the attack, and in many patients it may already have occurred by the time the patient reaches the emergency department. Persistence of stones in the bile duct or ampulla is associated with more severe disease. [31], [32] If gallstones predispose to acute pancreatitis, removal of the gallbladder should decrease this risk. In Minnesota, cholecystectomy in patients without a history of acute pancreatitis reduced the risk for the subsequent development of pancreatitis at least 10- to 20-fold. [23] Despite the increased risk for development of acute pancreatitis in those with gallstones and the success of cholecystectomy in decreasing the risk, the incidence of the disease is too low to warrant prophylactic cholecystectomy in the patient with asymptomatic gallstones.

Microlithiasis Studies of patients with idiopathic recurrent pancreatitis have provided compelling evidence that crystals or aggregates of crystals in bile, referred to as microlithiasis, may cause recurrent bouts of pancreatitis. [33], [34] Microlithiasis is a leading cause of otherwise unexplained acute pancreatitis. Microlithiasis is identified during ultrasonography by observing amorphous, layering, nonshadowing material (sludge) or brightly echogenic cholesterol crystals within the gallbladder (Fig. 94-3). Sonographically apparent sludge is especially common after prolonged fasting, rapid weight loss, total parenteral nutrition, gastric surgery, and adminstration of some drugs such as octreotide or ceftriaxone.

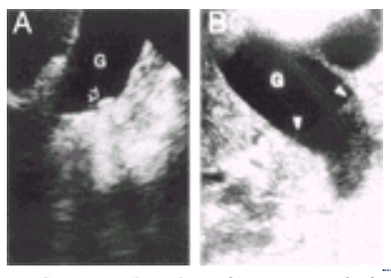

FIGURE 94-3. Ultrasound demonstrates cholelithiasis. **A**: Gallbladder (*G*) with single gallstone (*arrow*). **B**: Microlithiasis: the lower third of this gallbladder is filled with layering, nonshadowing sludge (*arrowheads*). (Courtesy of Caroline Taylor, M.D., Yale University.)

Microlithiasis can also be diagnosed in the absence of sonographically detectable sludge by examining bile for birefringent cholesterol monohydrate crystals or reddish brown, calcium bilirubinate granules. Bile can be obtained from the bile duct during ERCP or aspirated from the duodenum after slowly administering CCK octapeptide intravenously; dark gallbladder bile should be collected and examined. Endoscopic ultrasound (EUS) may be more sensitive than bile microscopy for detection of cholesterol crystals in the gallbladder. [35] Crystals, like stones, usually cause pancreatitis that begins abruptly, with an initial rise in serum levels of both pancreatic enzymes and transaminases. A minority of patients whose pancreatitis is ultimately attributed to microlithiasis have normal serum liver tests with their pancreatitis. Several forms of treatment have been used. Patients with cholesterol crystals have received ursodeoxycholic acid. Others have undergone cholecystectomy or endoscopic papillotomy alone. Each form of intervention has been reported to reduce the frequency of attacks by at least two thirds. These observations suggest that microlithiasis should be considered in patients with recurrent bouts of idiopathic pancreatitis.

Stenosis and Spasm of the Sphincter of Oddi The sphincter of Oddi is a complex muscular structure that regulates the flow of pancreatic secretions and bile into the duodenum. The sphincter acts as a variable resistor. Abnormalities of the sphincter are best defined in patients with recurrent abdominal pain after cholecystectomy, but have been associated with recurrent acute pancreatitis as well. The biliary and pancreatic portions of the sphincter of Oddi are partly independent anatomically and functionally. Acting indirectly through inhibitory neural pathways, CCK causes both components of the sphincter to relax, facilitating the flow of bile and pancreatic juice into the duodenum after a meal. CCK may also act directly on the sphincter and cause contraction. [36] This paradoxical dysfunction of the sphincter may occur after biliary tract surgery (perhaps because the innervation of the sphincter is interrupted), vagotomy, or gastric resection, and has been demonstrated to occur in both components of the sphincter after intravenous administration of CCK octapeptide to patients during sphincter of Oddi manometry. Other secretagogues and transmitters also appear to mediate relaxation of the sphincter, including vasoactive intestinal polypeptide (VIP), glucagon, and nitric oxide. [37] Secretin has been reported to induce an initial increase in phasic contractions of the sphincter in normal subjects, followed by relaxation. [38] This may account for the transient dilation of the pancreatic duct that can be observed after secretin is administered to normal subjects. The role these substances play in the development of sphincter dysfunction is unclear. Obstruction to flow through the ampulla may be caused by dysfunction of the sphincter or by scarring and stenosis of the sphincter and the ampullary outlet. Stenosis of the sphincter is most commonly the result of previous trauma from stone passage, surgical manipulation, or endoscopic sphincterotomy, but may also be due to a primary inflammatory or fibrosing process in the ampulla of Vater. Stenosing papillitis may be caused by chronic pancreatitis, sclerosing cholangitis, or by opportunistic infections in the acquired immunodeficiency syndrome (AIDS). Criteria for clinical diagnosis of sphincter of Oddi stenosis or dysfunction are well established for biliary sphincter disease. Geenan and colleagues have described three abnormalities which suggest a diagnosis of biliary sphincter dysfunction or stenosis: dilation of the bile duct, elevation of the serum liver tests, and slow drainage of contrast from the bile duct (as seen on biliary scintigraphy or after ERCP). When all three of these criteria are present (type 1), a presumptive diagnosis of sphincter disease can be made. [39] When the obstruction is intermittent or less severe, some or none of these criteria may be evident (type 2 and type 3', respectively), and biliary manometry may be useful for making the diagnosis of sphincter dysfunction. A blinded, controlled trial showed that type 2 patients with postcholecystectomy pain who had abnormal biliary manometry were more likely to benefit from endoscopic sphincterotomy. [40] Sphincter dysfunction is also described in patients with unexplained pancreatitis. Although a dilated pancreatic duct and slow pancreatic drainage may be seen in association with pancreatic sphincter dysfunction, the use of a clinical classification incorporating these findings is unproven. Diagnosis depends on pancreatic sphincter manometry performed during ERCP. Less invasive tests for diagnosis have been proposed. The secretin-ultrasound test was developed as a noninvasive method of demonstrating pancreatic outflow obstruction, but investigators disagree on the nature of normal and abnormal responses. Some have found that dilation of the pancreatic duct after secretin administration implies obstruction of pancreatic drainage, but others report that transient ductal dilation is normal after secretin, and that failure of the duct to dilate suggests a diagnosis of chronic pancreatitis. [41] While prolonged ductal dilation after secretin administration may be abnormal, this finding has been reported in patients with previous alcoholic or biliary pancreatitis, suggesting a nonspecific response. [42] A preliminary report suggests that the rate of secretin-stimulated pancreatic secretion into the duodenum assessed by magnetic resonance (MR) can also be used to assess pancreatic ductal obstruction. Manometric abnormalities of the pancreatic sphincter have been reported in a high percentage of patients with idiopathic recurrent pancreatitis. [43] The presence of manometric abnormalities does not exclude other causes of pancreatitis that might lead to secondary sphincter disease. Uncontrolled surgical and endoscopic experience suggests that treatment of a hypertensive pancreatic sphincter prevents recurrent pancreatitis in some patients. [44] Manometry of the sphincter of Oddi is a specialized endoscopic technique. The use of a manometry catheter permitting simultaneous perfusion and aspiration and the addition of meperidine to benzodiazepines for patient sedation [45], [46] improves the safety and success of the procedure. The most accepted bnormality of the sphincter of Oddi is an elevated basal sphincter pressure of at least 40 mm Hg above duodenal baseline. Tachyoddia (frequent phasic contractions of the sphincter) or retrograde phasic contractions may also cause obstruction of flow. Because manometry is done in the fasting state and for a brief time, it may miss functional abnormalities of the sphincter induced by eating. Some patients may respond to nitrates or calcium channel blockers, and endoscopic sphincterotomy often provides symptom relief. The risks of sphincterotomy are increased in patients with suspected sphincter of Oddi dysfunction. In patients with recurrent pancreatitis and sphincter dysfunction, it is unclear if the best initial therapy is biliary sphincterotomy alone or combined biliary and pancreatic sphincterotomy. Some patients improve after biliary sphincterotomy alone because of underlying biliary microlithiasis, by disruption of a hypertensive sphincter zone located in the common channel, or because the pancreatic sphincter (which partially encircles the biliary sphincter) functions differently when the biliary sphincter is severed. Endoscopic pancreatic sphincterotomy opens the pancreatic sphincter incompletely in most patients, and those with recurrent pancreatitis or persistent symptoms after pancreatic sphincterotomy may have a persistent zone of sphincter hypertension. Recurrent pancreatitis in such patients may be due to primary sphincter hypertension or to another etiology with secondary sphincter disease. Repeated endoscopic sphincterotomy or surgical sphincteroplasty may be performed, benefiting some patients. Symptomatic stenosis of a pancreatic sphincterotomy or sphincteroplasty may occur in up to 25% of patients. Stenosis may recur after treatment and can be difficult to treat effectively. Alternatives to sphincter ablation are of unclear value in pancreatic sphincter hypertension. Medical therapy with calcium channel blockers or intra-ampullary botulinum toxin injections has been used in patients with biliary sphincter dysfunction and may be effective in pancreatic sphincter dysfunction as well. Pancreatic stenting has questionable use as a therapeutic trial, since stents can cause pain in some patients, and may induce changes of chronic pancreatitis when left in place for more than a short time. [47]

Pancreas Divisum During fetal development, the dorsal and ventral buds of the pancreas each form ducts that enter the duodenum. When the pancreatic buds fuse, the ducts ordinarily join as well. The ventral duct drains the pancreas through the major papilla (duct of Wirsung) and the remnant opening of the dorsal duct forms the

minor papilla (duct of Santorini). When fusion between the two ducts is partial or absent, the dorsal duct drains most of the pancreas through the minor papilla. This anatomic variant is known as pancreas divisum, and may cause pancreatitis. The relationship of pancreas divisum to pancreatitis has been debated. Pancreas divisum is a common abnormality; autopsy series report an incidence of about 7%. The incidence assessed by ERCP ranges from 0.3% to 8%. This variability is likely the result of differences in the patient populations and definition of the anomaly. For example, some studies include only patients with a complete separation between the dorsal and ventral ducts. Although earlier studies estimated that pancreas divisum can be found in about 25% of patients with unexplained recurrent pancreatitis, many subsequent studies have not confirmed this association. [48] Proponents of the pathological association have suggested that resistance to the flow of secretions through the duct of Santorini causes ductal hypertension and pancreatitis. Studies of experimental pancreatitis have shown that obstruction of the pancreatic duct alone may result in pancreatitis. [29] It is likely that a small number of people with pancreas divisum experience recurrent pancreatitis related to obstruction at the duct of Santorini. These may be individuals with a particularly diminutive orifice of the minor papilla. Treatment studies provide the best evidence that obstruction to flow through the minor papilla can cause pancreatitis. A small, blinded, randomized trial showed that endoscopic treatment of the minor papilla prevented recurrent pancreatitis when compared to sham therapy in patients with pancreas divisum. [49] A larger body of uncontrolled surgical and endoscopic experience supports this conclusion and suggests that improved dorsal duct drainage is most likely to help patients with otherwise unexplained recurrent acute pancreatitis. About 70% of such patients will improve substantially after minor papilla sphincterotomy. Patients with chronic pancreatitis are less likely to respond, and those with chronic abdominal pain but no history of pancreatitis will have symptomatic improvement only about 30% of the time. [50] Endoscopic treatment of pancreas divisum is possible because the narrowest part of the minor papilla orifice is usually at the level of the duodenal mucosa. An experienced therapeutic endoscopist best accomplishes deep cannulation of the dorsal duct and minor papilla sphincterotomy. Most experts leave a stent or nasopancreatic drain after minor papilla sphincterotomy to prevent edema from temporarily obstructing the pancreatic duct orifice. Botulinum toxin injections into the minor papilla have been reported to predict response to endoscopic sphincterotomy in patients with divisum and recurrent pancreatitis. [51]

Toxic

Alcohol Alcoholic pancreatitis may present as an acute bout of pancreatitis. Although experience has suggested that these episodes are invariably associated with established, chronic, underlying pancreatic damage, this may not always be the case. In one series of patients who died of acute alcoholic pancreatitis, almost 40% demonstrated little histological evidence of chronic pancreatitis. [52] The incidence of pancreatitis is surprisingly low in alcoholic patients. Dreiling and Koller [53] reported that for 100 alcoholic patients, clinical pancreatitis develops in only 5%, cirrhosis in 15%, both in 1%, and neither in 80%. While these data may underestimate the true prevalence of disease, they indicate that in addition to the amount of alcohol ingested, unknown factors affect a person's susceptibility to disease. Several major physiological mechanisms may contribute to the development of alcoholic pancreatitis, including abnormal sphincter of Oddi spasm or relaxation, obstruction of small ductules by proteinaceous plugs, and direct toxic and metabolic effects. [54] Chronic ethanol feeding may also sensitize the pancreas to other insults. Stimulation of proinflammatory cytokines by ethanol may have a central role in this sensitization. Alcohol has been reported to have variable effects on the pressure at the sphincter of Oddi. Alcohol has also been found to stimulate pancreatic secretion through cholinergic pathways. Some have proposed that the combination of alcohol-induced sphincter of Oddi spasm and stimulated secretion is responsible for pancreatitis. The pharmacological doses of ethanol used in many of these studies, however, leave the importance of these effects in question. Ethanol may change the concentration and composition of proteins secreted by the pancreas, resulting in the formation of protein plugs within small pancreatic ductules. These plugs are indistinguishable from those found early in the course of cystic fibrosis, which raises the possibility that these two diseases somehow may be linked. The protein plugs may obstruct small pancreatic ducts and promote pancreatitis. Increased viscosity and elevated protein concentrations found in pancreatic secretions from alcoholic patients are likely to contribute to plug formation. By forming a rigid, insoluble protein structure, specific secretory proteins such as lithostathine and glycoprotein-2 may contribute to the formation of these protein plugs and to stone formation. [55] Of note, glycoprotein-2 is closely related to the principal constituent of renal tubular casts, Tamm-Horsfall protein. The failure to alkalinize the acinar lumen in alcoholics may also contribute to plug and stone formation. Ethanol also may change the amounts of potentially damaging proteases in pancreatic secretion. For example, increased amounts of lysosomal enzymes and an increased trypsinogen:pancreatic trypsin inhibitor ratio has been reported in pancreatic juice from alcoholic patients. Alcohol's metabolic by-products may generate some its toxic effects. The pancreatic acinar cell has the ability to metabolize ethanol through oxidative and nonoxidative pathways. The oxidative metabolites include acetaldehyde and acetate. The nonoxidative metabolites of alcohol are nonesterified fatty acids. Both types of products have been identified in the pancreas and may contribute to toxicity. Alcohol may affect systemic and pancreatic lipid metabolism. One of the first pancreatic effects of ethanol in animals is the accumulation of lipid droplets within the acinar cell. Acute ethanol ingestion has been found to enhance de novo pancreatic triglyceride synthesis and alter membrane fluidity and integrity. [56] Fragile pancreatic lysosomes develop in ethanol-fed rats. [57] Pancreatic stimulation by supraphysiological doses of CCK can cause pancreatitis. Further, by augmenting pancreatic injury, CCK may play a role in many forms of pancreatitis. [58] Ethanol dramatically sensitizes the acinar cell to CCK-stimulated intracellular zymogen proteolysis. [59] Ethanol also may stimulate CCK release from the small intestine. [60] Thus, a combination of moderately elevated CCK levels and ethanol may result in pancreatic "hyperstimulation," zymogen activation, and autodigestion. In addition, ethanol may have systemic effects that promote the development of pancreatitis. Ethanol can exacerbate hyperlipidemia, for example. Diets, especially those rich in fat, may contribute to the deleterious effects of alcohol. Chronic alcoholics have diminished reticuloendothelial function and delayed clearance of protease-inhibitor complexes. Finally, inherited factors, such as deficiencies in protease inhibitors, may predispose one to alcoholic pancreatitis.

Drugs A variety of drugs have been associated with pancreatitis. [61] With few exceptions, the mechanisms of drug-induced disease are unknown and most are idiosyncratic reactions. Drug-induced pancreatitis may be classified by the certainty of its relationship to disease. Rechallenge is taken as the gold standard of proof, but such information is often unavailable. A partial listing of medications grouped according to probability that they cause pancreatitis is presented in Table 94-1.

PROVEN	PROBABLE	POSSIBLE/ QUESTIONABLE
L-Asparaginase	Protease inhibitors	Carbamazepine
Azathioprine	Acetominophen	Corticosteroids
Didanosine	5-Aminosalycilic acid	Cimetidine
Estrogens	Ergotamine	Furadantin
ACE inhibitors	Furosemide	Metronidazole
6-Mecaptopurine	Isoniazid	Minocycline
Pentamidine	Procainamide	Piroxicam
Sulfasalazine	Rifampicin	Ranitidine
Valproate	Thiazides	Tetracycline

ACE, angiotensin-converting enzyme.

TABLE 94-1 Examples of Drug-Induced Pancreatitis

Immunosuppressants are one family of drugs that can cause pancreatitis. Within this group, azathioprine and its major metabolite, 6-mercaptopurine are the major offenders; in one series of 396 patients taking these medications, pancreatitis was induced in 3.3%. [62] Symptoms developed in all patients within the first month of therapy. Rechallenge with either azathioprine or 6-mercaptopurine was invariably associated with a rapid recurrence of pancreatitis. Furthermore, attempts to desensitize with slowly increasing doses of 6-mercaptopurine were not successful. Both cyclosporine and tacrolimus have been rarely associated with acute pancreatitis. The rare (approximately 2%) but severe (approximately 50% mortality) pancreatitis associated with renal or liver transplantation may be linked to drug therapy or to infection. Some drugs used to treat AIDS can cause severe pancreatitis. Trimethoprim-sulfamethoxazole can precipitate pancreatitis, albeit uncommonly. Intravenous or aerosolized pentamidine cause pancreatitis depending on the cumulative dose of the drug. Another cause of pancreatitis in patients with AIDS is the antiviral agent, 2',3'-dideoxyinosine (DDI). The onset of DDI-associated pancreatitis is typically delayed for several months after the initiation of therapy. Although the mechanism of DDI-induced pancreatitis is unknown, patients with a previous history of pancreatitis and those with advanced infection seem to be at the highest risk for this complication. Decreasing the dose of DDI reduces the incidence of this serious complication. Human immunodeficiency virus (HIV) protease inhibitors may cause pancreatitis indirectly by elevating serum triglyceride levels. Antiinflammatory agents cause pancreatitis. Sulfasalazine and, in a small number of patients, oral 5-aminosalicylic acid have both been reported as causes of pancreatitis. [63] Early reports of corticosteroids and corticotropin causing pancreatitis are difficult to interpret because most patients had other possible causes of disease. Corticosteroids may directly precipitate pancreatitis as demonstrated by rechallenge, but this appears to be a rare event. [64] Tetracycline has been linked to pancreatitis in patients with fatty degeneration of the liver, particularly during pregnancy. Although symptoms usually develop within weeks, they have been reported after 2 years of medication. Rarely, tetracycline may precipitate pancreatitis in the absence of underlying liver disease. [65] Antihypertensives and diuretics are widely used and though the incidence of pancreatitis is low with these drugs, they constitute an important family of disease-producing agents. Included on the list are furosemide, thiazide diuretics, and angiotensin-converting enzyme (ACE) inhibitors. A few agents induce metabolic abnormalities that predispose to pancreatitis. For example, therapy with estrogens or the retinoid derivative, isotretinoin, is associated with a dose-dependent increase in triglycerides and pancreatitis. [66] Agents that overstimulate neural pathways may cause acute pancreatitis. In animal models, neural stimulation can initiate pancreatitis or augment that caused by other agents. Parathion, an alkylphosphate cholinesterase inhibitor, is a widely applied agricultural insecticide. Ingestion or absorption of parathion through the skin may induce injury. Almost 50% of patients with toxic exposure to this agent will exhibit biochemical evidence of pancreatitis. Although parathion-induced pancreatitis is usually mild, fatal hemorrhagic pancreatitis has been reported. [67] Acting through neural pathways, ergotamine poisoning decreases blood flow to cause ischemic pancreatitis and hepatitis. [68]

Scorpion Venom The venom of the scorpions *Tityus trinitatis* and *Tityus serrulatus,* found in Trinidad and Brazil, respectively, may induce a form of hyperstimulation pancreatitis. The toxic polypeptides in tityustoxin have been characterized; acting through cholinergic pathways, they are potent stimulants of pancreatic secretion in vitro and induce pancreatitis in animals. [69] In addition to causing pathological enzyme activation, tityustoxin may stimulate pancreatic secretion, induce sphincter of

Oddi spasm, and change blood flow.

Metabolic

Hyperlipidemia Although hyperlipidemia may result from a bout of pancreatitis, hyperlipidemia may also precipitate acute or chronic pancreatitis. [70] The breakdown products of triglycerides are probably responsible for inducing pancreatitis. When lipase in the pancreatic capillary bed acts on the high levels of triglycerides in the serum, toxic free fatty acids are generated. The endothelial lining of small pancreatic blood vessels is the first site of injury. Damage of small blood vessels leads to recruitment of inflammatory cells and thrombosis. This predicted mechanism is consistent with the clinical observation that the earliest injury in hyperlipidemia-associated pancreatitis is located in small pancreatic blood vessels. Although triglyceride levels greater than 2000 to 3000 mg/ dL usually are required for development of pancreatitis, a few patients seem to manifest this condition when serum levels are lower. In general, a level of greater than 1000 mg/dL should prompt suspicion of an underlying hyperlipidemic cause for pancreatitis. Striking increases in serum triglycerides after a fatty meal, with lower values while fasting, have been observed in some forms of hypertriglyceridemia. It is important to measure serum triglycerides on the first serum sample obtained from a patient with otherwise unexplained pancreatitis. Measuring postprandial triglyceride levels after recovery may also be required to detect markedly elevated values. In addition to patients with primary hyperlipidemia, elevated triglycerides may result from therapy with estrogen, retinoid derivatives, and HIV-protease inhibitors. Typically, estrogen-related pancreatitis occurs within the first months on medication. Patients who are obese, and have underlying glucose intolerance or hypertriglyceridemia, are at greater risk. Treatment of acute hyperlipidemic pancreatitis includes standard supportive care. If parenteral nutrition is given, serum triglyceride levels should be monitored. Occasionally, patients with acute pancreatitis and severe hyperlipidemia (serum levels of over 10,000 mg/dL) may benefit from plasmapheresis. Once the patient recovers from their acute symptoms, the cornerstone of therapy for hyperlipidemic pancreatitis is a low-fat diet. Attention must also be given to underlying diabetes or thyroid disease, as well as alcohol use, all of which can elevate serum triglycerides. In some cases, treatment with lipid-lowering drugs may be indicated.
Hypercalcemia The relationship of hypercalcemia to pancreatitis has been a topic of discussion. Experimentally, acute calcium infusion causes increased pancreatic duct permeability that may lead to a nonspecific increase in serum enzymes. [71] In animals, acute hypercalcemia may induce a very mild form of pancreatic injury. [72] Virtually all causes of hypercalcemia, including hyperalimentation, infusions associated with cardiac bypass, immobilization, multiple myeloma, and hyperparathyroidism have been linked to hyperamylasemia. [73] These studies indicate that hypercalcemia is associated with an increase in serum enzymes, but probably causes pancreatitis infrequently.

Infectious

Viral Including AIDS The most common viral infections that involve the pancreas are mumps and coxsackie B virus. These are self-limited and may account for lone bouts of pancreatitis. Viral hepatitis and fulminant hepatitis, especially hepatitis B, have been associated with pancreatitis. Influenza A and B, Enterovirus, Cytomegalovirus, and rubella may cause acute pancreatitis. The high incidence of pancreatic abnormalities in patients with AIDS deserves comment. Hyperamylasemia has been reported in up to 40% of patients with AIDS. [74] Clinical pancreatitis, however, probably occurs in less than 10% of all patients with AIDS and even fewer of those not receiving toxic medications. The diagnosis of pancreatitis should be based on bedside findings and imaging tests in combination with elevated enzyme levels, and not on the basis of elevated pancreatic enzymes alone. Unless pancreatitis is associated with medications, it usually is mild. In addition to drugs (see section “ Drugs”) and the other common causes of pancreatitis, at least three pathological processes contribute to pancreatitis in patients with AIDS:

1. HIV may infect the pancreas. This involvement may be responsible for the elevated serum amylase that sometimes precedes or accompanies the clinical onset of AIDS, and may account for the changes of otherwise unexplained chronic pancreatitis that have be seen at autopsy of patients with AIDS. However, HIV infection alone seems rarely to lead to clinical pancreatitis.
2. Opportunistic infections may involve the pancreas. Most infections are caused by Cytomegalovirus or *Mycobacterium avium.* In the remainder, *Cryptococcus, Toxoplasma gondii, Mycobacterium tuberculosis,* and *Candida* species have been reported. Usually, opportunistic infections in the pancreas involve other, more accessible organs. While histological evidence of pancreatic opportunistic infection is common at autopsy, it is unusual for these opportunistic infections to produce clinical pancreatitis during life.
3. Pancreatic neoplasms develop in up to 5% of patients with AIDS. Kaposi sarcoma and lymphoma are the most common and usually involve the pancreas in the setting of widely disseminated disease. Pancreatitis, however, is not a common manifestation of this tumor involvement.

Bacterial and Parasitic The list of bacteria associated with acute pancreatitis includes *Salmonella* species, *Shigella* species, *Campylobacter* species, hemorrhagic *Escherichia coli, Legionella* species, *Leptospira* species, and even *Brucella* species. [75], [76] Pancreatitis associated with these infections is most likely secondary to released toxins. Usually, acute pancreatitis is not the primary manifestation of these infections. Direct involvement of the pancreas or ampulla with parasites is a common cause of pancreatitis in some populations. Almost 15% of cases of biliary pancreatitis in a series from Hong Kong could be ascribed to *Ascaris lumbricoides* infection. [31]

Autoimmune

Sclerosing Pancreatitis Sclerosing pancreatitis, also called “autoimmune pancreatitis,” has distinctive clinical and laboratory features and responds to corticosteroid therapy. Patients present with subacute symptoms of pancreatitis and frequently have obstructive jaundice. Some patients also have other autoimmune illnesses. Cross-sectional imaging shows a diffusely enlarged pancreas, often with a focal pancreatic mass, and without marked peripancreatic inflammatory changes. The gland may appear to have a capsule-like edge on computed tomography (CT) or MR, and there may be delayed enhancement with intravenous contrast. [77] ERCP shows irregular narrowing of the main pancreatic duct, with either focal or diffuse changes (Fig. 94-4). Histology shows a prominent lymphocytic infiltrate especially in the region of the ducts, with little or no pancreatic calcification.

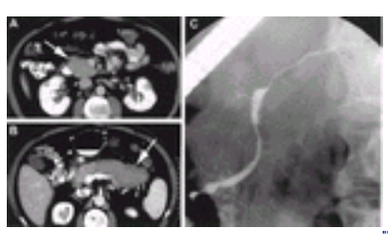

FIGURE 94-4. Sclerosing pancreatitis. **A** and **B**: Computed tomography demonstrates a diffusely enlarged pancreas with the appearance of a mass in both the head and tail of the gland (*arrows*). **C**: Endoscopic retrograde cholangiopancreatography shows diffuse irregularity of the main pancreatic duct and side branches. This patient improved with oral steroid therapy. (Courtesy of John Dobbins, M.D., New Haven, CT.)

A variety of laboratory abnormalities have been described, including elevated autoantibody levels and serum immunoglobulin E (IgE) concentrations, but the most characteristic laboratory findings appear to be an elevated serum IgG4 level. [78] The differential diagnosis often includes pancreatic neoplasm. Patients may have involvement of adjacent vessels or new onset hyperglycemia, further increasing the clinical suspicion of malignancy. Intraductal brushings obtained during ERCP or EUS-guided needle aspiration of a mass may be appropriate in some cases. A key feature of the disease is the good response to oral glucocorticoids with resolution of pancreatic swelling and a decrease in IgG4 and circulating immune complex levels. [78] Impaired pancreatic endocrine function may also improve. [79] Recurrence after a slow taper of steroids appears to be uncommon.
Inflammatory Bowel Disease Pancreatitis encountered in patients with inflammatory bowel disease may be related to drugs, structural and inflammatory changes, or may be idiopathic in nature. Most medications used for the treatment of inflammatory bowel disease have been reported to precipitate pancreatitis. This list includes azathioprine, 6-mercaptopurine, sulfasalazine, and 5-aminosalicylic acid, as discussed. Reports linking corticosteroids and metronidazole to pancreatitis are rare. Patients with pancreatitis and Crohn’s disease often have inflammatory bowel disease involving the duodenum. The inflammatory process may be associated with stenosis of the ampulla or a fistula into the pancreatic duct. The increased incidence of gallstones in patients with Crohn’s disease should always direct one to look for biliary causes of pancreatitis. In a small number of patients, however, no etiology is identified. [80] Sclerosing cholangitis is occasionally associated with clinical pancreatitis. This may be due to passage of small pigment stones through the ampulla, or to a sclerosing process affecting the pancreatic ducts as well as the bile ducts. [81]
Systemic Autoimmune Diseases Pancreatitis may occur in the setting of a systemic autoimmune disorder. Vasculitis affecting the pancreas has been described in systemic lupus erythematosus, [82] periarteritis nodosa, Wegner disease, [83] and Behçet disease. [84] Pancreatitis may respond to successful treatment of the underlying vasculitis.

Genetic

The best-described form of familial pancreatitis is the autosomal dominant disorder known as hereditary pancreatitis. Penetrance is probably less than 80%. Affected family members have bouts of pancreatitis that often begin in childhood and typically progress to chronic pancreatitis (see Chapter 95). The course of the disease appears to be accelerated by alcohol. These patients have been found to have mutations in the trypsinogen gene. The most common mutation (at arginine 117) may make trypsin resistant to proteolytic degradation. [6] Patients with hereditary pancreatitis have a dramatically increased risk of pancreatic cancer. [85] Mutations in the pancreatic trypsin inhibitor may also predispose to disease. [7], [8]

Mutations in the cystic fibrosis transmembrane regulator (CFTR) cause two patterns of pancreatic disease. First, classic cystic fibrosis results in pulmonary disease and pancreatic insufficiency, but rarely pancreatitis. These patients have elevated sweat chloride levels and diminished CFTR function in the nasal mucosa. Second, a pancreatitic phenotype is recognized that is not associated with pulmonary disease but can likely cause both acute and chronic pancreatitis. [86], [87] These patients have normal sweat chloride values, but the CFTR function in the nasal mucosa is diminished. Although the pancreatitic patients exhibit mutations in CFTR, they are not homozygous for the mutations that result in classic cystic fibrosis. The link between pancreatic disease and mutations in the CFTR gene is not understood. The prevalence of CFTR-related disease in patients with idiopathic pancreatitis appears to be increased compared to the general population. Its contribution to alcoholic pancreatitis remains unclear.

Iatrogenic

ERCP Acute pancreatitis is the commonest complication of ERCP. Retrospective studies report an incidence of 1%, but subsequent prospective studies with careful patient follow-up document an incidence of about 5%. [88] In most cases the pancreatitis is mild and resolves within several days; complications requiring surgery are infrequent. Diabetes can rarely result from an episode of severe post-ERCP pancreatitis, and deaths due to pancreatitis have been reported in 0.02% of patients undergoing the procedure.

Risk factors. The chance of developing post-ERCP pancreatitis varies depending on both patient-related and procedure-related factors. The skill and experience of the endoscopist also affect the chance of pancreatitis, with low case volume increasing the risk for the patient. [89], [90] The risk of post-ERCP pancreatitis is substantially higher when the pancreatic duct is visualized than when it is not. [91] The number and hydrostatic pressure of pancreatic duct injections are probably also important, as well as the volume of fluid injected. In one study, more than four pancreatic duct injections was associated with an increased risk. Acinarization (visualization of the pancreatic parenchyma with injected contrast) is associated with subsequent pancreatitis about a third of the time. In addition to filling and overfilling of the pancreatic duct, pancreatic outflow obstruction due to ampullary edema or spasm is a likely contributor to post-ERCP pancreatitis. Repeated attempts at bile duct cannulation may in this way cause postprocedure pancreatitis, even in the absence of pancreatic duct cannulation. [89] Pancreatitis has been reported after percutaneous biliary drains are advanced through the ampulla into the duodenum, with no injection or manipulation of the pancreatic duct. Endoscopic sphincterotomy further increases the risk of pancreatitis, and may do so both by thermal injury to the pancreas and by provoking ampullary edema. Precut sphincterotomy appears to increase the risk in some centers, but not in others where this technique is used routinely. Sphincter of Oddi manometry has been thought to increase the risk, perhaps because of the volume of fluid perfused into the pancreatic duct. The development of a manometry catheter that permits aspiration of fluid from the duct during manometry has decreased the incidence of postmanometry pancreatitis, and a large study of postsphincterotomy complications found that the performance of manometry did not affect the incidence of pancreatitis in patients with suspected sphincter of Oddi dysfunction. [89] Patient factors also affect the chances of ERCP-induced pancreatitis. Patients with suspected sphincter of Oddi dysfunction are at higher risk. About 20% of such patients undergoing sphincterotomy without prophylactic pancreatic duct stenting develop pancreatitis. The risk is especially high in patients with small bile ducts. [89] Younger age and obesity probably also predispose patients to pancreatitis after ERCP. Conversely, patients with a chronically diseased or obstructed pancreatic duct appear to be at decreased risk for pancreatitis from ERCP. The presence of acute biliary pancreatitis does not seem to increase the risk of the procedure. A history of contrast allergy has been reported not to increase the risk of post-ERCP pancreatitis, but many endoscopists favor pretreatment with steroids or use of nonionic contrast media in such patients. [92] Pretreatment may be especially important in patients with a history of serious allergic reactions to topical or ingested iodine.

Diagnosis. Symptoms of post-ERCP pancreatitis typically develop within 2 to 6 hours of the procedure, although occasionally patients will first develop symptoms the following day. The presence of abdominal symptoms shortly after the procedure is a poor predictor of subsequent pancreatitis. Consensus criteria for diagnosis of pancreatitis after ERCP require elevation of serum amylase and/or lipase to three times the upper limit of normal on the day after the procedure, together with compatible clinical symptoms. [92] Early predictors of post-ERCP pancreatitis have been identified. Although elevation of the serum amylase is a nonspecific finding in the hours following ERCP, the height of amylase elevation may be useful. In one prospective study, a serum amylase less than 276 U/L (upper limit of normal 115 U/L) was a strong negative predictor of pancreatitis when measured 2 hours after ERCP. [93] Post-ERCP pancreatitis may occur even if the procedure is easily accomplished and all identified risk factors are avoided. Patients sent home after ERCP should be instructed to seek help promptly if pain or vomiting develop. It is our practice to use a clear liquid diet until the morning following ERCP in many patients, in order to limit pancreatic stimulation in the postprocedure period.

Prevention. Both pharmacological and endoscopic strategies for the prevention of post-ERCP pancreatitis have been proposed. Some, such as use of low-osmolarity contrast media or pretreatment with calcium channel blockers, octreotide, or aprotinin (a protease inhibitor) have yielded disappointing results. Other prophylactic treatments, however, may be useful. A protease inhibitor, an antisecretory drug, and a cytokine that inhibits inflammation each appear to have some efficacy. Gabexate mesilate is a protease inhibitor that inhibits proteolytic activity of pancreatic enzymes. The few individual trials of gabexate for prevention of post-ERCP pancreatitis have all shown a benefit, although not statistically significant in most cases. A metaanalysis demonstrated a statistically significant benefit with gabexate pretreatment, decreasing the incidence of pancreatitis from 6.5% to 1.6%. [94] Pretreatment of 27 patients would prevent one episode of pancreatitis. This agent is not licensed for use in the United States, and enthusiasm has been tempered by the drug's cost and its adminstration as a prolonged intravenous infusion (beginning one hour before ERCP and continuing for 12 hours afterward). Somatostatin also appears to decrease the incidence of post-ERCP pancreatitis. Numerous controlled trials of somatostatin have shown a benefit, often not statistically significant; however, in metaanalysis a significant benefit was found. [94] Taken together, the incidence of pancreatitis was decreased from 13.5% to 5.6% in these studies. Pretreatment of 13 patients would prevent one episode of pancreatitis. Somatostatin appears to be effective when given either as a 12-hour intravenous infusion or as a bolus intravenous injection immediately before cannulation of the papilla. [95] Somatostatin is not licensed for use in the United States. The lack of benefit of octreotide, a somatostatin analog, may be due to its effects on sphincter of Oddi function and pancreatic blood flow. A recent study of IL-10, administered as a single intravenous injection 30 minutes before therapeutic ERCP, demonstrated a significant reduction in postprocedure pancreatitis from 24% in the placebo group to 9% in the treatment groups. [15] However, another study did not find benefit from IL-10. [16] Since gabexate, somatostatin, and IL-10 have different mechanisms of action, combination therapy could theoretically increase the benefit of pharmacological prophylaxis. Temporary pancreatic duct stenting dramatically decreases the risk of postsphincterotomy pancreatitis in patients with sphincter of Oddi dysfunction affecting the pancreatic sphincter. [96] This finding highlights the usefulness of sphincter of Oddi manometry in patients with suspected sphincter dysfunction. The benefit of prophylactic stenting may not apply to patients without sphincter dysfunction. A controlled trial of pancreatic duct stenting immediately after biliary sphincterotomy in a mixed group of patients did not show a reduction in postprocedure pancreatitis. [97]

Postoperative In the postoperative period there is an increase in the incidence of hyperamylasemia, even after extra-abdominal procedures. [98] It appears that in many cases these increases are nonspecific and unrelated to clinically overt pancreatitis, although severe pancreatitis may ensue and can be related to hypotension, medication, or pancreatic trauma. Postoperative pancreatitis is described after renal lithotripsy and percutaneous pancreatic biopsy. [99], [100]

Coronary Artery Bypass In a prospective study, Rattner and colleagues reported that 32% of patients had hyperamylasemia after cardiac bypass, but only 2.7% had overt pancreatitis. [73] In a retrospective study of 4473 patients who underwent cardiopulmonary bypass, Huddy and associates [101] reported acute pancreatitis in only 0.1% of patients, all of whom died. In a similar study by Lefor and colleagues, [98] acute pancreatitis was observed in 0.44% of those undergoing bypass, with a mortality of 44%. Finally, Fernandez-del Castillo and colleagues [73] demonstrated a direct correlation between the elevation of pancreatic enzymes and the amount of calcium administered to patients during cardiac bypass. Together, these studies indicate that although elevations in pancreatic enzymes are common after cardiac bypass surgery and may relate to a nonspecific effect of calcium on pancreatic duct permeability, overt pancreatitis is uncommon. A severe form of pancreatitis sometimes does occur after cardiac bypass, however, and may be related to pancreatic ischemia.

Neoplastic

Pancreatitis is associated with both primary pancreatic tumors and metastases. In one series, 3% of patients with pancreatitis admitted to a university hospital were found to have pancreatic malignancy. [102] Pancreatitis has been reported in up to 10% of patients with pancreatic cancer, but it is usually mild. [103] Cancer can mimic chronic pancreatitis, presenting with a dilated pancreatic duct, or a pseudocyst that forms because of downstream ductal obstruction by the tumor. It can also mimic acute pancreatitis, especially when bleeding into the pancreatic duct (hemosuccus pancreaticus) temporarily obstructs the duct. As clots pass through the papilla the serum liver enzymes may rise together with the amylase. Frequently, symptoms of pancreatitis are associated with an early-stage pancreatic neoplasm and may precede the diagnosis of pancreatic cancer by months. Older patients with unexplained pancreatitis should undergo a careful evaluation for occult malignancy looking

for a minute pancreatic cancer amenable to resection. [104]

Vascular

Ischemia is an uncommon cause of acute pancreatitis. Pancreatic infarcts may occur in patients with underlying atherosclerotic vascular disease, but are unusual because the pancreas is richly perfused from several different arterial sources. Cholesterol emboli may cause pancreatitis, cholecystitis, or bowel ulceration or infarction, and should be suspected when acute pancreatitis occurs after vascular interventions such as cardiac catheterization. [105] Patients may have associated evidence of renal, gut, or peripheral cholesterol emboli. Ischemic pancreatic and hepatic injury may be associated with malignant hypertension, low flow states due to severe heart failure, or administration of potent vasoconstrictors. Vasculitis may cause pancreatitis associated with systemic autoimmune diseases, as noted previously.

Other Etiologies

Trauma Traumatic pancreatitis from penetrating or blunt injury is an important cause of disease in both adults and children. The symptoms of pancreatic trauma may be difficult to elicit in the severely ill patient and suspected only by the location of the injury. On occasion, recurrent bouts of pancreatitis may develop months to years after injury. The midpancreatic duct as it crosses the vertebral column is particularly susceptible to fracture from blunt trauma. In the acute setting, CT is useful in detecting a disrupted pancreatic duct, focal pancreatitis, and pancreatic necrosis (Fig. 94-5). Similarly, ERCP has been used to demonstrate persistent duct disruption, ductal stricture, and focal pancreatitis. Traumatic pancreatitis has been treated with stenting of the disrupted duct, pancreatic resection, or drainage. [106]

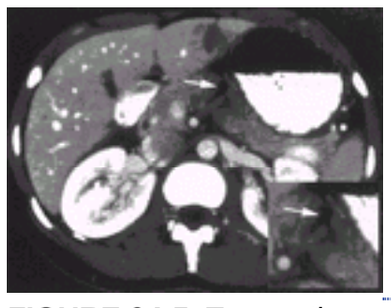

FIGURE 94-5. Traumatic pancreatitis. There is disruption of the pancreatic body anterior to the spine (*arrow*). The left lobe of the liver is also injured. (Courtesy of Jeffrey Neitlich, M.D., Yale University.)

Peptic Ulcer Ulcers located at the posterior duodenal bulb may burrow into the head of the pancreas. Pancreatitis associated with duodenal ulcers is rarely severe and is usually not associated with dramatic increases in the serum amylase. This diagnosis should be considered in patients with ulcer disease when the typical epigastric burning pain of an ulcer begins to radiate to the back and is less responsive to ulcer medications.
Childhood Pancreatitis may first present in childhood. Many cases of childhood pancreatitis are associated with multisystem disease such as sepsis, viral infections, hemolytic-uremic syndrome, and Reye syndrome. Blunt trauma caused by accidents or child-abuse is the single most common cause of acute childhood pancreatitis, followed in decreasing incidence by congenital defects (including biliary cysts, anomalous pancreatobiliary junctions, and pancreas divisum), metabolic diseases, and drugs. Children with hemolytic disorders such as sickle cell anemia commonly form pigment stones in the gallbladder or bile ducts. Rarely, annular pancreas, duodenal diverticulum, ectopic pancreatic tissue, and duodenal duplication cysts may cause pancreatitis.
Miscellaneous An increased incidence of pancreatitis has been reported during pregnancy and the early postpartum period. The increased incidence of hypertriglyceridemia and biliary stone formation during pregnancy may contribute to this predisposition. [107] An increased risk of acute pancreatitis is reported in patients undergoing renal dialysis. The risk may be limited to those treated with chronic peritoneal dialysis and not hemodialysis. [108] Refeeding after long periods of fasting is said to precipitate pancreatitis, but this has not been well characterized. A few patients with possible food allergy-associated pancreatitis have been reported. [109]
Idiopathic The number of patients having unexplained bouts of pancreatitis has diminished in large part owing to improvements in imaging tests and increased insight into the genetic abnormalities in unexplained pancreatitis. The compelling evidence that biliary microlithiasis causes pancreatitis, as well as the wider use of sphincter of Oddi manometry, has further decreased the size of the idiopathic group.

DIAGNOSIS

The diagnosis of acute pancreatitis is almost always based on the presence of severe abdominal pain together with biochemical evidence of pancreatic injury. Rarely, patients with severe acute pancreatitis present without complaints of abdominal pain, but with respiratory failure or changes in mental status. The diagnosis of pancreatitis may be missed entirely when organ failure dominates the clinical picture. For example, in series published during the 1980s, fatal pancreatitis went undetected until postmortem 22% to 42% of the time. [110]

Symptoms

Pain is the most common symptom of acute pancreatitis and occurs in 95% of patients. It is often located in the epigastric and umbilical region and may radiate to the low thoracic region of the back. The pain usually does not reach its peak for 30 minutes to several hours and may last for hours to days. This deep, visceral pain is among the most severe and often leaves even the medicated patient in discomfort. In some, pain will decrease when sitting and leaning forward or curling up on the left side, compared to laying flat. This symptom is due to the retroperitoneal position of the pancreas and may be produced by other retroperitoneal processes. Nausea and vomiting are present in 85% of patients and may occur without an ileus or gastric outlet obstruction. Unlike patients with ulcer disease, vomiting does not relieve pain.

Signs

Low-grade fevers are reported in 60% of patients, and high-grade fevers may indicate the presence of cholangitis or either sterile or infected necrosis. Tachycardia and hypotension are found in up to 40% of patients and may result from intravascular volume depletion, enhanced vascular permeability, vasodilation, and hemorrhage. Abdominal tenderness and guarding is common, and bowel sounds are often decreased or absent. Pleural effusions are most often found on the left, but may be bilateral. Mild jaundice is not unusual, but bilirubin levels greater than 4 mg/dL suggest extrahepatic obstruction or underlying liver disease. Dark discoloration in the back, flank, or the paraumbilical region may arise from any retroperitoneal hemorrhage, including hemorrhage from pancreatitis.

Laboratory Tests

Markers of Pancreatic Injury Although the gold standard for diagnosing acute pancreatitis is a direct histological examination of the pancreas, this is rarely available. The goals of diagnostic tests for pancreatitis are to establish the diagnosis and cause noninvasively in patients with abdominal pain and to determine the prognosis. Clinicians should be mindful that a number of factors influence the level of serum markers of pancreatitis. First, serum levels of pancreatic enzymes are the sum of tissue production, release into the blood, and clearance. Thus, in patients with renal failure, the serum amylase may increase because the kidney does not clear the enzyme. Second, the measured enzyme activities may be influenced by a number of “serum factors” and may not reflect absolute enzyme levels. For example, hyperlipidemia is known erroneously to reduce routine serum amylase determinations. Third, enzymes may be produced from nonpancreatic tissues, and this may limit the specificity of serum enzyme assays. In general, the higher the enzyme levels the better the chances that one is dealing with pancreatitis. Finally, standard enzyme assays, such as amylase and lipase, provide no information on the severity of the pancreatitis.
Amylase. The serum amylase level has been the biochemical standard for diagnosing acute pancreatitis. Many organs generate amylase, an activity that hydrolyzes the internal 1–4 a-linkages of starch. Because there are no circulating inhibitors of amylase activity, measurements of amylase activity in serum are reliable. Lactescent sera may inhibit amylase, but this effect can be eliminated by sample dilution. Although many tissues synthesize amylase, most of the serum activity originates from the pancreas (approximately 40%) and the salivary glands (approximately 60%). P-isoamylase arises from the pancreas and S-isoamylase from the salivary glands as well as other tissues such as the fallopian tubes and lungs. Pancreatic amylase enters the blood through an unknown pathway and has a serum half-life of about 2 hours. Although the major portion of serum amylase and other pancreatic enzymes is probably cleared by the reticuloendothelial system, about one fourth of serum amylase is excreted in its intact form by the kidney. Increased serum amylase during acute pancreatitis results from both enhanced release into the blood and decreased renal clearance. Serum amylase levels increase within the first hours after the onset of pancreatitis and parallel serum lipase levels. After an

attack, however, the serum amylase falls much more rapidly than lipase, and may return to normal within 24 hours. Lipase levels are more reliable indicators of pancreatitis in patients who are first seen several days after the onset of their illness (Fig. 94-6). In most situations, the degree of amylase elevation does not reflect the cause or severity of pancreatitis. Amylase levels tend to be lower in patients with alcoholic pancreatitis, pancreatic cancer, or penetrating ulcers, compared to patients with gallstone disease.

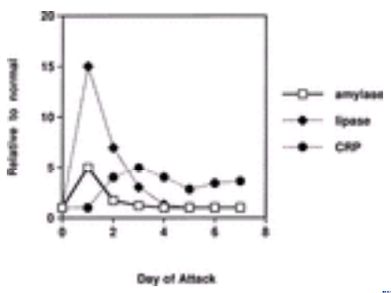

FIGURE 94-6. Serum markers of pancreatic injury.

Defining abnormal values influences the sensitivity and specificity of the total serum amylase. Cutoff values of just above the normal range have been estimated to yield sensitivities greater than 90%, but specificities less than 70% for acute pancreatitis. If the cutoff value is increased to three times the upper limit of normal, the specificity is near 100%, but the sensitivity may fall below 60%. Nonspecific increases of one- to twofold in the serum amylase have been reported after cigarette smoking. [111] In an attempt to enhance the specificity of amylase measurements, amylase isoforms may be measured. P-isoamylase is probably best used to confirm that an elevated total amylase level arises from the pancreas. [112] The clinical value of measuring P-isoamylase may be compromised by the inability of the assay to accurately measure small amounts of this isoform. Furthermore, P-isoamylase levels may increase in the absence of pancreatitis in patients with biliary tract disease, intestinal perforation, intestinal obstruction or ischemia, and ruptured abdominal aortic aneurysms. Diseases of the salivary glands may lead to an elevated S-isoamylase. S-amylase levels, however, may increase in the absence of salivary disease in chronic alcoholics, postoperative states, lactic acidosis, in patients with anorexia nervosa and bulimia, after esophageal perforation, and in neoplasms such as pulmonary, gastric, and breast carcinoma, and myeloma.

Macroamylasemia. On occasion, a persistently elevated serum amylase is encountered in the absence of symptoms referable to the pancreas. A benign and uncommon condition that can lead to such an elevated serum amylase is macroamylasemia. Amylase is normally found in the bloodstream as a molecule with a molecular weight of 45 kd, but macroamylase often has a molecular weight of greater than 200 kd. The normal ratio of pancreatic to salivary amylase is maintained in macroamylasemia. The high-molecular-weight macroamylase may result from antibodies complexed to amylase or from amylase polymers. [113] Unlike normal amylase, the macroamylase complex cannot enter the renal tubule for excretion and accumulates in the serum. Several methods can be used to diagnose macroamylasemia. First, amylase-to-creatinine clearance ratios of less than 1% strongly support the diagnosis. To confirm the diagnosis in patients with a low clearance, the clinical laboratory may determine the molecular weight of the serum amylase. Alternatively, immunologic assays also may be used. Acquired macroamylasemia may occasionally signal the presence of a lymphoproliferative disorder.

Lipase. Rapid and reproducible methods for measuring serum lipase are available. A number of gastrointestinal tissues, including the pancreas, intestine, liver, and biliary tract generate lipase activity, as do lingual and gastric sources. In some laboratory assays, the serum lipoprotein lipase activity is also detected. [114] The contribution of these various sources of lipase to serum lipase activity is not clear. Furthermore, the different substrates used in the various commercial assays may affect the specificity of the lipase measurement for pancreatic lipase. The least specific seems to be the Ektachem test, which has been found to have a specificity as low as 25%. [115] For many clinicians, the serum lipase has supplanted the amylase as the single test of choice for the diagnosis of pancreatitis. In most series, it is as sensitive and more specific than the total serum amylase, and promises to improve further when the L2 pancreatic lipase is measured. This enhanced specificity of the lipase compared to the amylase level also has been reported in alcoholics. [116] Because lipase levels increase in parallel to serum amylase levels, lipase is a useful early marker of pancreatitis. The lipase level remains elevated longer than the amylase level, and thus may help to diagnose pancreatitis after an acute attack has passed (see Fig. 94-6). Another advantage of lipase is that levels are normal in diabetic ketoacidosis and macroamylasemia.

Other enzymes. Presumably all pancreatic enzymes are released into the blood during acute pancreatitis. Serum trypsin, chymotrypsin, elastase, ribonuclease, and phospholipase A_2 have all been reported to be elevated in acute pancreatitis, but the specificity of these tests has not been sufficiently defined.

Urinary enzymes. One of the most promising tests for acute pancreatitis measures urinary trypsinogen 2 using a rapid dipstick assay. In a series of 500 patients seen in the emergency department with abdominal pain, the test was found to have a sensitivity of 94% and a specificity of 95% for pancreatitis. [117] These values are superior to other enzymatic tests and this assay deserves broader use.

Trypsinogen activation peptide. Premature activation of trypsinogen within the pancreas is thought to be an early signal of pancreatitis. The small peptide removed during activation of trypsinogen to trypsin is known as trypsinogen activation peptide (TAP). Measurement of TAP levels in the blood or urine reflects trypsinogen conversion to trypsin. Serum levels of TAP have been reported to separate those with severe from those with mild pancreatitis when measured within the first 36 hours of disease. [118]

Inflammatory Markers The inflammatory response is a prominent component of pancreatitis and increases in parallel with its severity. Several serum markers have been used to quantitate the inflammatory response. Inflammatory cells release neutrophil-specific elastase, and elevated serum levels are detected within the first 12 to 24 hours of pancreatitis. Interleukin-6 (IL-6), released by macrophages, also increases during the acute phases of pancreatitis. Acute-phase reactants such as C-reactive protein are induced by IL-6 and increase by the second day after the onset of acute pancreatitis. [14] Inflammatory markers may increase during any inflammatory process, and would not be specific for pancreatitis. In contrast to amylase and lipase measurements, however, some of these markers have prognostic value (discussed subsequently).

Markers of Biliary Tract Involvement Differentiation between biliary and nonbiliary forms of pancreatitis has important implications for treatment. Both biochemical and radiologic tests are useful (Table 94-2), but in some patients identification of a biliary cause may require EUS, bile microscopy, or empiric cholecystectomy. The timing of laboratory tests and ultrasound also affects their interpretation, especially when biliary tract obstruction is early in its course or when it is intermittent.

DIAGNOSTIC TEST	BILIARY	ALCOHOLIC
Laboratory		
Amylase	Values >1000 IU	Values <500 IU
AST, ALT	Increased levels that fluctuate	Not increased or mild increases with liver disease that do not fluctuate
Alkaline phosphatase and direct hyperbilirubinemia	Increased with persistent obstruction	Not increased unless there is obstruction of the intrapancreatic portion of the common bile duct
Imaging		
X-ray	Gallstones	Pancreatic calcifications
Ultrasonography	Gallstones, dilation of biliary tract	Changes of chronic pancreatitis
CT/Spiral CT	Gallstones, dilation of biliary tract, common duct stones	Pancreatic calcification, pancreatic duct stones, dilated pancreatic duct

ALT, alanine aminotransferase; AST, aspartate aminotransferase; CT, computed tomography.

TABLE 94-2 Laboratory Findings and Imaging in Biliary Versus Alcoholic Pancreatitis

Most patients with gallstone pancreatitis have elevations of the serum liver tests, and the alanine aminotransferase (ALT) is the single most sensitive liver test for acute biliary obstruction by a stone. A rapid rise and fall in the serum liver tests is characteristically seen in gallstone pancreatitis in association with transient biliary obstruction at the papilla. Fluctuation is especially helpful in patients with chronic liver disease who have stable baseline elevations in aspartate aminotransferase (AST) and ALT levels. In addition, marked elevations of the amylase of greater than 2000 IU/L also favor a biliary source. [119] The ratio of lipase to amylase may be greater in alcoholic than in biliary pancreatitis, but has not been a reproducible finding. Noninvasive indicators of persistent bile duct stones are needed for gallstone pancreatitis. Elevation of the serum bilirubin level over 3, or persistent elevation of the liver tests over time, have been advocated as useful screening tests. Laboratory scoring systems, such as the modified Glasgow criteria or the urea-glucose scale, can identify patients whose biliary pancreatitis is predicted to be severe; these patients, in turn, are more likely to harbor persistent ductal or ampullary stones. Early trends in the serum liver tests and pancreatic enzyme levels are also useful. Patients in whom all these values remain constant or decrease over the first 48 hours of hospitalization have a low (8%) risk of a persistent stone, while those in whom any of these laboratory values increase have a moderate (30%) risk of a persistent bile duct stone. [120] None of these predictors is accurate enough to replace direct bile duct imaging, but may be useful in selecting patients for EUS, MR cholangiopancreatography (MRCP), or ERCP rather than elective intraoperative

cholangiography.
Systemic Markers of Acute Pancreatitis A low-grade to moderate fever is not unusual in acute pancreatitis. Persistent high fevers suggest infection. Leukocytosis is common, and severe pancreatitis has been associated with leukemoid reactions even in the absence of infection. Inflammatory markers, such as the serum levels of neutrophil elastase, C-reactive protein, and IL-6, are good markers of severity, but elevations are not specific for pancreatitis.

Imaging

Abdominal Radiographs To exclude nonpancreatic diseases, such as intestinal perforation, standard and upright chest and abdominal radiographs are recommended for every patient with severe abdominal pain. Plain radiographs may be omitted in some cases if CT is available immediately. A variety of radiographic findings have been associated with pancreatitis. Pleural effusions are most common on the left, but may be bilateral and rarely are limited to the right pleural space.[121] Intestinal gas patterns may demonstrate an ileus pattern or an isolated dilated loop of small bowel overlying the pancreas, known as the sentinel loop. Colonic obstruction may result in air in the right and transverse colon that abruptly terminates at the splenic flexure, the so-called colon cutoff sign. Loss of the psoas margins and increased separation between the stomach and colon suggest pancreatic inflammation. Pancreatic calcification or calcified gallstones may suggest an alcohol or biliary etiology, respectively.
Sonography When the pancreas is inflamed it appears hypoechoic on sonography because of increased water in the parenchyma (Fig. 94-7). Ultrasound is thus a relatively sensitive test for the diagnosis of acute pancreatitis, when the pancreas is seen. Although ultrasonography can be performed rapidly and at the patient's beside, intestinal gas or adipose tissue limits pancreatic visualization in 30% to 40% of patients. This is especially common in patients with ileus due to their pancreatitis. Ultrasonography may miss focal masses or fluid collections, especially in and around the pancreatic tail. Its greatest value is in evaluating the biliary tract for gallstones, sludge, or ductal dilation. [33], [34] Ultrasound is the single best noninvasive test for the presence of gallbladder stones, with a sensitivity of about 95%. It is much less reliable for direct visualization of a bile duct stone because overlying bowel gas often obscures the common bile duct.

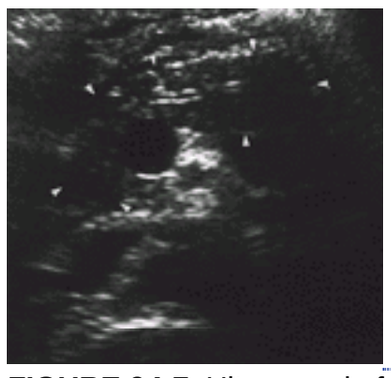

FIGURE 94-7. Ultrasound of acute pancreatitis. The pancreas (*arrowheads*) is hypoechoic; the walls of the pancreatic duct are prominent because of the surrounding parenchymal edema. (Courtesy of Jeffrey Neitlich, M.D., Yale University.)

Radionuclide Imaging Radionuclide imaging of the biliary tract has been used to detect obstruction of the biliary tract and gallbladder disease. Since pancreatitis of any etiology can be associated with obstruction of the biliary tract, radionuclide imaging does not reliably distinguish a biliary cause of disease.
Computed Tomography CT is a valuable tool for pancreatic imaging. In contrast to ultrasound, which demonstrates changing echogenicity in the parenchyma of the gland, the CT diagnosis of pancreatic inflammation is made based on pancreatic enlargement, effacement of the usual lobulated contour of the pancreas, inhomogeneity of the pancreatic parenchyma, or fluid infiltrating the peripancreatic fat (Fig. 94-8). While CT and ultrasound are equally sensitive for diagnosis of pancreatitis, missing the diagnosis in some mild cases, CT is better than ultrasound for detection of focal pancreatic masses or fluid collections because it reliably visualizes the entire gland.

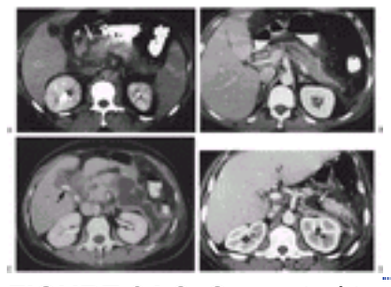

FIGURE 94-8. Computed tomography of acute pancreatitis. **A**: Mild pancreatitis. Fluid is seen between the antrum and the pancreatic head (*arrow)*. **B**: Moderate pancreatitis, with peripancreatic fluid infiltrating the retroperitoneum. The pancreas shows uniform contrast enhancement. **C**: Pancreatic necrosis, with no perfusion of the tail and part of the body. Stones are present in the gallbladder (*arrow*). **D**: Gas within an area of pancreatic necrosis (*arrow*), suggesting infection. (Courtesy of Jeffrey Neitlich, M.D., Yale University.)

Dynamic CT performed during bolus administration of intravenous contrast can demonstrate areas of poor perfusion or nonperfusion in the pancreas. [122] This finding is typically interpreted as necrosis of the poorly perfused segment; estimates of pancreatic necrosis by dynamic CT correlate with observations made at surgery. The CT finding of necrosis probably identifies patients who are at higher risk for pancreatic infection and death. Investigators have debated whether early administration of bolus intravenous contrast worsens pancreatitis. Studies in rats have shown decreased pancreatic capillary flow rates and decreased pancreatic oxygen saturation after intravenous contrast administration. [123] These effects persisted at 60 minutes after contrast was given. However, there is no compelling evidence of this phenomenon in humans. A retrospective study at a university hospital found that patients who underwent contrast-enhanced CT had longer hospitalizations than those who did not, despite similar Acute Physiology and Chronic Health Evaluation II (APACHE II) scores on admission. [124] A subsequent prospective study randomized patients with severe pancreatitis to CT on the first hospital day with or without bolus intravenous contrast. There was no difference in systemic markers of inflammation, length of hospital stay, or mortality. [125] In practice a contrast-enhanced CT is usually unnecessary in the first 24 hours of hospitalization, but early CT may be performed if it will have an important effect on early management of the individual patient. Helical CT technique has improved CT sensitivity for gallstones in the bile duct to 85% [126] (Fig. 94-9). CT detection of ductal stones is still limited by stone composition; calcium must be present in sufficient amounts to yield a CT density different than that of the surrounding bile or soft tissue. When looking for biliary stones, helical CT should be done without contrast so that a bright rim at the periphery of a stone is not mistaken for contrast enhancement of the bile duct wall.

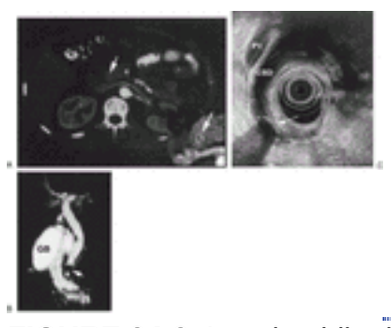

FIGURE 94-9. Imaging bile duct stones (*arrows)*. **A**: Computed tomography shows the calcified rim of a stone. **B**: Magnetic resonance cholangiopancreatography (MRCP) shows a filling defect in the distal bile duct. The gallbladder (*GB*) and duodenum (*Duod*) are seen because nonflowing fluid is visualized with this technique. **C**: Endoscopic ultrasound shows an oblong stone with an acoustic shadow. The bile duct (*CBD*) is thick walled due to cholangitis. A segment of portal vein (*PV*) is seen. (Images A and B courtesy of Jeffrey Neitlich, M.D., Yale University.)

Magnetic Resonance MR imaging has not been widely applied to acute pancreatitis, but is probably similar to CT when imaging the acutely inflamed pancreas. When intravenous gadolinium is given, MR demonstrates areas of poor pancreatic perfusion. Animal studies have shown no deleterious effect of intravenous gadolinium on the inflamed pancreas. Although MR is more cumbersome and expensive than CT and more difficult to obtain quickly, it does offer clinical advantages

in pregnant patients and those with contrast allergy. MR (like EUS) is more accurate than CT for demonstration of solid debris in a pancreatic fluid collection; complex collections often have a homogenous appearance on CT. [127] Secretin-stimulated MR may have a role in the diagnosis of functional pancreatic duct outlet obstruction. MR angiography (MRA) and MRCP are additional techniques that offer noninvasive diagnosis of vascular complications and persistent bile duct stones in acute pancreatitis. MRCP (see Fig. 94-9) has a sensitivity of over 90% for bile duct stones, although its yield in patients with normal caliber ducts and small ampullary stones is not as well defined.

Endoscopic Cholangiopancreatography ERCP does not play a role in establishing the diagnosis of acute pancreatitis. Most patients with acute pancreatitis have normal pancreatograms, although some may have pancreatic duct strictures or ductal disruptions. ERCP is useful in the differential diagnosis of otherwise unexplained, recurrent pancreatitis, especially for diagnosis of mild chronic pancreatitis, pancreas divisum, or sphincter of Oddi disease. ERCP has been used to diagnose and treat persistent bile duct stones in acute pancreatitis, as discussed elsewhere in this chapter.

Endoscopic Ultrasound EUS is useful in some patients with acute pancreatitis. EUS is a sensitive test for detection of persistent bile duct or ampullary stones (see Fig. 94-9) and can be used to identify patients likely to benefit from ERCP. [128] EUS is also useful for differential diagnosis of unexplained acute pancreatitis since it demonstrates biliary sludge and stones missed with other imaging modalities, as well as small pancreatic or ampullary tumors, pancreas divisum, and chronic pancreatitis. [129] Finally, EUS can demonstrate complex material within a pancreatic fluid collection and can guide endoscopic pseudocyst drainage.

Use of Cross-Sectional Imaging Studies in Acute Pancreatitis The choice of imaging modality for patients with acute pancreatitis depends on the clinical question that needs to be answered. Imaging studies can establish the diagnosis of pancreatitis, aid in differential diagnosis, provide prognostic information, or assess possible complications. When the diagnosis of pancreatitis is uncertain, both ultrasound and CT may be useful. A patient with rapidly resolving biliary pancreatitis may have near-normal serum enzymes but persistent pancreatic edema seen on ultrasound or peripancreatic inflammatory changes on CT. CT can show changes of chronic pancreatitis or focal pancreatic masses. For differential diagnosis, the choice of imaging test depends on the diagnosis suspected. When biliary pancreatitis seems likely, ultrasound is the standard investigation because it detects almost all gallbladder stones. When chronic pancreatitis or an underlying pancreatic malignancy is suspected, CT is more useful. When a metabolic or drug-induced pancreatitis is likely, imaging tests may still be indicated to exclude other common causes such as stones. In patients with biliary pancreatitis, imaging studies such as EUS or MRCP may usefully identify patients with persistent bile duct or ampullary stones. Early bile duct imaging is particularly appropriate when the pancreatitis is severe or laboratory trends suggest an increased risk of a persistent stone. Contrast-enhanced CT or MR can identify patients with necrosis, and these patients benefit from prophylactic antibiotics. CT, however, is not necessary in every patient with pancreatitis. Necrosis is uncommon in patients with clinically mild disease who improve rapidly in the hospital. Serial hematocrits and the C-reactive protein level on the second hospital day are useful predictors of the presence or absence of necrosis, as described in the section on “Local Complications” and following. When necrosis is suspected or the patient is not improving over the first 48 hours of hospitalization, contrast-enhanced CT is usually worthwhile. CT is also valuable in the diagnosis and management of local complications of acute pancreatitis. Jaundice or a marked drop in hematocrit may also be indications for CT. When thrombosis of the splenic or superior mesenteric veins is suspected, MRA or CT may be useful.

Evaluation of Severity

There are several important reasons to establish the severity in acute pancreatitis. Since severity correlates with prognosis, such evaluation may allow the clinician to predict the patient’s course and anticipate complications or the need for closer monitoring in an intensive care unit. Prognostic information should guide the use of prophylactic antibiotics, urgent bile duct imaging, and early ERCP. Finally, prognostic markers allow the clinical investigator to measure the success or failure of therapeutic interventions. The tools used to measure prognosis fall into three categories: specific laboratory tests, clinical and physiological assessment, and CT.

Laboratory Tests The standard laboratory tests used to diagnose pancreatitis provide little prognostic information, but other tests appear to separate patients with mild disease from those with severe disease. The first group of tests measures the severity of the inflammatory response. These include the neutrophil elastase and the monokine IL-6 tests. Both are elevated within 24 hours and correlate with the severity of pancreatitis. C-reactive protein is induced by IL-6 and is a later marker of severity, distinguishing patients with severe disease on the second hospital day (Fig. 94-10). Since the C-reactive protein is easily and inexpensively measured and appears to be a good prognostic marker, it should be used more widely to assess prognosis.

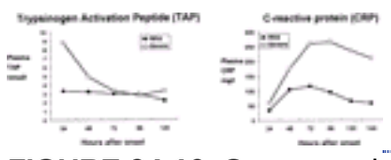

FIGURE 94-10. Serum markers of severity. Serum values of the trypsinogen activation peptide (TAP) and C-reactive protein (CRP) as a function of the duration and severity of acute pancreatitis. (Derived from ref. [118] and Wilson C, Heads A, Shenkin A, Imrie CW. C-reactive protein, antiproteases and complement factors as objective markers of severity in acute pancreatitis. Br J Surg 1989;76[2]:177.)

The TAP assay is an indirect measure of the amount of active trypsin. From a pathophysiological standpoint, this is an elegant measure of severity; the more trypsin activation, the more pancreatic damage. In a large prospective study, TAP levels discriminated between mild and severe disease. [118] Unfortunately, TAP assays are not yet widely available. With the exception of the TAP assay, these tests measure inflammatory activity only, and are not specific for pancreatitis.

Computed Tomography Both noncontrast CT and contrast-enhanced CT yield useful prognostic information. Noncontrast CT is the basis for a prognostic scoring system called the CT Severity Index (Table 94-3) that has been shown to correlate with the Ranson criteria for assessing severity. [122] In one study, patients with CT Severity Index scores up to 2 had only 4% morbidity and no mortality, but those with scores from 7 to 10 experienced 92% morbidity and 17% mortality [100] (Fig. 94-11). Contrast-enhanced CT demonstrates the presence and extent of pancreatic necrosis, and is therefore an important predictor of complications and death, as discussed in the section on “Necrosis and Infected Necrosis.”

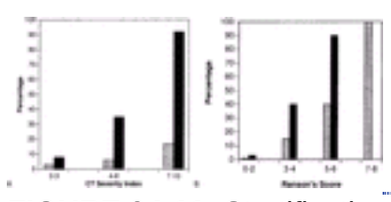

FIGURE 94-11. Stratification of severity according to Computed Tomography (CT) Severity Index (Table 94-3) and Ranson criteria (Table 94-4). The *light bar* represents mortality. Morbidity (*dark bar*) was defined as greater than 7 days in the intensive care unit. (Derived from refs. [122], [130].)

	POINTS
Grade of Acute Pancreatitis	
A: Normal pancreas	0
B: Pancreatic enlargement	1
C: Inflammation confined to pancreas and peripancreatic fat	2
D: One peripancreatic fluid collection	3
E: Two or more fluid collections	4
Degree of Necrosis	
No necrosis	0
Necrosis of one third of pancreas	2
Necrosis of one half of pancreas	4
Necrosis of more than one half	6

Computed tomography severity index = grade points + degree of necrosis. See the text and Figure 94-11 for correlation between the severity score and outcome.

Adapted from Balthazar EJ, Robinson DL, Megibow AJ, Ranson JH. Acute pancreatitis: value of CT in establishing prognosis. Radiology 1990;174:331.

TABLE 94-3 Computed Tomography Severity Index

RANSON	SIMPLIFIED GLASGOW
On Admission	**Within 48 h**
Age >55 y	Age >55 y
WBC >16,000 mm^3	WBC >15,000 mm^3
LDH >350 IU/L	LDH >600 IU/L
Glucose >200 mg/dL	Glucose >180 mg/dL
AST >250 IU/L	Albumin <3.2 g/dL
Within 48 h	Calcium <8 mg/dL
Hematocrit decrease by >10%	Arterial PO_2 <60 mm Hg
Urea nitrogen increase by >5 mg/dL	Urea >45 mg/dL
Serum calcium <8 mg/dL	
Arterial PO_2 <60 mm Hg	
Base Deficit >4 MEq/L	
Estimated fluid sequestration >6 L	

See the text and Figure 94-11 for the correlation between scores and severity. AST, aspartate aminotransferase; LDH, lactate dehydrogenase; WBC, white blood cells.

Adapted from Agarwal N, Pitchumoni CS. Assessment of severity in acute pancreatitis. Am J Gastroenterol 1991;86:1704; and Marshall JB. Acute pancreatitis: a review with an emphasis on new developments. Arch Intern Med 1990;150:1185.

TABLE 94-4 Ranson and Simplified Glasgow Prognostic Scoring Criteria

Clinical and Physiological Assessment

The Ranson criteria are still the most widely applied measure for the assessment of acute pancreatitis. Ranson criteria (Table 94-4) distinguish between mild and severe pancreatitis about 80% of the time. As shown in Figure 94-11, the Ranson score correlates with morbidity and mortality. [130] The disadvantage of Ranson criteria is that they include 11 values that must be monitored over 48 hours. Blamey and colleagues [131] developed the Glasgow classification, which can be calculated any time within the first 48 hours of hospitalization and measures only eight parameters (Table 94-4). The simplified Glasgow criteria appear to have prognostic accuracy similar to that of Ranson criteria. The Glasgow criteria were developed in a population of predominately biliary pancreatitis, while the Ranson criteria were developed in patients with more alcoholic pancreatitis.

Although some prognostic classifications have dropped the base deficit and fluid requirements from Ranson criteria, others have identified *obesity* as an independent risk factor. [132] The higher risk in the obese patient may result from respiratory compromise from physical restraints, or from the provision of toxic substrates such as free fatty acids from triglycerides and harmful cytokines from peritoneal fat stores.

The APACHE II has been used to establish the prognosis in patients with pancreatitis. [133] This system uses 14 routinely measured parameters of physiological activity and biochemical function, and generates a numerical score that depends on their deviation from the normal range. The patient's age, organ insufficiency, neurological state, and postoperative state are also weighted. The potential advantage of the APACHE II is that it can be calculated instantaneously from routine measurements. The initial value distinguishes patients at high risk for complications and death, and trends in the patient's APACHE II score over time add to its predictive value. A major shortcoming of this classification is its complexity. It is difficult to apply the APACHE system routinely outside of an intensive care unit.

Are these scoring systems of use to the clinician? A group of experienced pancreatic surgeons compared bedside assessment at admission and after 48 hours with the APACHE II, Ranson, and Glasgow [133] scores for predicting the severity of acute pancreatitis. Bedside assessment alone missed over half of the cases that ultimately proved to have a complicated course. Thus, a high score on a standardized scoring system alerts the clinician to the possibility of complications. This in turn should prompt frequent reevaluation of the patient and the adequacy of supportive treatment, perhaps in an intensive care unit.

Approach to the Patient with Unexplained Pancreatitis

In over 10% of patients, pancreatitis is idiopathic despite results of history and physical examination, routine laboratory tests, transabdominal ultrasound, and CT. An etiologic diagnosis can be made in most of these patients with further evaluation. A thorough diagnostic approach should be taken, including use of the medical history, laboratory tests, and additional imaging studies.

The patient's history should be reviewed, particularly with regard to alcohol and medication use, family history of pancreatic disease, and prior abdominal trauma. Metabolic disease should also be considered. Serum triglycerides decrease with fasting, and a hyperlipidemic cause of pancreatitis may be missed if triglycerides were not measured soon after the patient's symptoms began. Hypercalcemia may not be apparent until after the patient has recovered from pancreatitis, because acute pancreatitis often lowers serum calcium levels.

When there is evidence of chronic pancreatitis in the absence of alcohol use or familial pancreatitis, further evaluation can include laboratory tests for a-1-antitrypsin deficiency and hemochromatosis. Trypsinogen gene mutations may be present without a family history of pancreatitis due to occurrence of a new mutation or low penetrance, and patients with CFTR mutations and pancreatitis usually do not have a family history of disease. Genetic testing for these disorders can be performed; however, enthusiasm for this testing must be tempered. The absence of effective specific therapy, the lack of effective pancreatic cancer screening for patients with familial disease, and the possible psychological and insurability problems that may arise from the diagnosis of a genetic disorder are all arguments against genetic testing. Genetic diagnosis may nevertheless be desirable, particularly for patients who wish to have children. In general it is wise to refer the patient for genetic counseling before obtaining these tests.

Biliary microlithiasis is a leading cause of recurrent acute pancreatitis that should be actively pursued in patients with unexplained disease. EUS and bile microscopy appear to play a complementary role in diagnosis, and microlithiasis is unlikely if both of these investigations are negative. Nevertheless, empiric cholecystectomy may be a reasonable option in some patients, especially if the clinical features of their acute attacks suggest a biliary source and ampullary disease has been excluded.

Abnormalities of pancreatic duct drainage, including pancreas divisum, ampullary lesions, and sphincter of Oddi dysfunction, should be excluded in patients with recurrent acute attacks and no evidence of microlithiasis. While EUS and MRCP can diagnose ampullary masses and divisum, definitive diagnosis of these conditions is often made during ERCP. ERCP is particularly useful for diagnosis of sphincter dysfunction and "incomplete divisum," in which the dorsal and ventral ducts communicate but dominant drainage is via the minor papilla. Such patients may present with recurrent mild pancreatitis. [134] ERCP is also the most definitive diagnostic test for intraductal papillary mucinous tumors of the pancreas, which often present with pancreatic symptoms and serum enzyme elevations.

It is generally our practice to use EUS instead of ERCP as the first endoscopic test in unexplained pancreatitis. EUS is particularly useful in patients with intact gallbladders, who may have microlithiasis, and in those over 40 years old, in whom the possibility of an early pancreatic malignancy should be considered. Missed chronic pancreatitis, pancreas divisum, and ampullary lesions can be diagnosed during EUS as well. EUS makes a diagnosis in about two thirds of patients. [129] When disease remains idiopathic and is recurrent, ERCP (with the capability to perform sphincter of Oddi manometry) can be pursued.

LOCAL COMPLICATIONS

The commonest complication of acute pancreatitis is the development of a pancreatic or peripancreatic collection. A useful classification of pancreatic collections was adopted at a consensus conference on acute pancreatitis held in Atlanta in 1992 and has since been widely accepted, replacing older terminology such as "phlegmon." [2] The Atlanta classification distinguishes acute fluid collections, necrosis, pseudocysts, and abscesses. Each of these entities has a different pathology, natural history, and treatment.

Acute Fluid Collections

Acute fluid collections form in or near the pancreas, and are common among patients with pancreatitis. They occur early in the course of the acute illness, and do not have a defined wall. Fluid collections may be simple and exhibit a uniform CT density that approximates that of water, or complex and show multiple CT densities and poorly defined margins (Fig. 94-12).

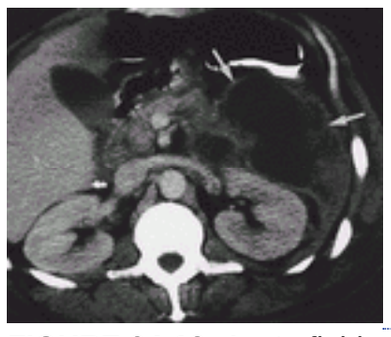

FIGURE 94-12. Acute fluid collection complicating acute pancreatitis. Computed tomography of pancreatitis after coronary artery bypass graft demonstrates a mixed-attenuation fluid collection (*arrow*) contiguous with the tail of the pancreas (not shown). (Courtesy of Caroline Taylor, M.D., West Haven, CT.)

Most acute fluid collections resolve spontaneously. The factors determining resolution of an acute collection versus persistence of the collection and progression to a pseudocyst are incompletely understood. An underlying pancreatic duct disruption that empties into a fluid collection certainly contributes to their persistence and enlargement in some patients. Nevertheless, prospective studies show that ductal disruptions often resolve with conservative management alone, without surgical intervention. [135]

Although some have advocated drainage of acute fluid collections, evidence demonstrating a benefit for such intervention is lacking. Acute collections may be difficult to drain percutaneously, and CT scans often underestimate the complexity of these collections. Unless an acute collection is rapidly expanding or associated with clinical evidence of infection, it should be observed. Infection is an uncommon complication. Infected collections have to be drained and have been successfully treated using percutaneous catheters. Because fluid collections are frequently associated with acute pancreatitis and most resolve spontaneously, a fluid collection should be defined as a pseudocyst only if it persists for more than 4 weeks.

Necrosis and Infected Necrosis

Any form of acute pancreatitis may progress to severe disease and extensive pancreatic necrosis. Pancreatic necrosis may result from ischemia or marked inflammation and represents a severe form of disease. Necrosis may affect the pancreas diffusely or focally, and necrotic areas may be separated by segments of viable tissue. Histological examination may show devitalized pancreatic tissue with vessel damage and necrosis of parenchyma and ducts, as well as hemorrhage and fat necrosis.

Pancreatic necrosis is diagnosed clinically with dynamic contrast-enhanced CT, which demonstrates well-marginated zones of non-enhanced pancreatic tissue (see Fig. 94-8). Although some non-enhancing parenchyma may be viable, there is generally good correlation between CT, operative findings, and histology in patients with necrosis undergoing surgery. CT is less useful for diagnosis of peripancreatic fat necrosis.

Necrosis is uncommon in patients with clinically mild disease. Laboratory predictors of necrosis have been described. A serum C-reactive protein level greater than 150 mg/L is seen in 95% of patients with necrosis by the second hospital day [136] (see Fig. 94-10). Two useful and readily available laboratory predictors are:

- an admission hematocrit equal to or greater than 44%, and
- failure of the hematocrit to decrease 24 hours after admission.

When either of these criteria are present, necrosis can be found in about 50% of patients, while if both criteria are absent necrosis has been demonstrated in only 4%. [137]

Necrosis is associated with other major complications and death from acute pancreatitis. Major complications of necrosis include distant organ failure and infected pancreatic necrosis. Early in the course of necrotizing pancreatitis there may be a marked systemic inflammatory response with increased capillary permeability, a sepsis-like syndrome, and distant organ failure most often affecting the lungs. Organ failure is more likely in patients with necrosis of over 50% of the pancreas. [138] Somewhat later in the course, infection of the necrotic pancreatic tissue may develop. Since there is no fibrotic wall or capsule to contain the infection, adjacent inflamed tissues become involved and overwhelming sepsis is likely unless adequate debridement of the infected tissue is achieved.

Infected pancreatic necrosis is a leading risk factor for death from acute pancreatitis. Infection of pancreatic necrosis occurs in 30% to 70% of patients not receiving prophylactic antibiotics. [139] This high frequency is in large part due to increased bacterial translocation from the gut seeding the necrotic tissue. Alterations in mucosal villus architecture and bowel microvasculature occur in animal models of acute pancreatitis, with bacterial translocation from the intestine to the pancreas, liver, and spleen. [140] In patients, the same bacterial species that infect the pancreas can be previously cultured from the colon. [141] The prevention and treatment of infected necrosis is described in greater detail later in this chapter.

Infected necrosis may be heralded by a sudden increase in abdominal tenderness, high fever, marked leukocytosis, and bacteremia. The presentation may also be subtle, particularly in patients receiving prophylactic antibiotics, with persistent or worsening organ failure despite ongoing supportive care. Negative blood cultures do not exclude the diagnosis. Although infection of pancreatic necrosis was thought to occur mainly after the first 10 to 14 days of illness, recent reports have stressed its occurrence earlier and later in the course of necrotizing pancreatitis.

The main diagnostic modality for infected necrosis is CT-guided, fine-needle aspiration of the necrotic pancreatic tissue. Samples should be immediately examined by Gram stain, which has a high applicability in this setting. Fungal stains are advisable as well, and cultures are also mandatory. Polymicrobial, bacterial, gram-negative enteric and anaerobic organisms are most common, but with the widespread use of prophylactic antibiotics bacterial gram-positive and fungal infections are increasingly frequent. [142]

Laboratory predictors of infected pancreatic necrosis have been sought. The serum procalcitonin level has been useful in some studies, but not in others. [143], [144] Further clinical experience with this and other markers of infected necrosis is desirable.

Sterile pancreatic necrosis has a variable clinical course. The necrotic tissue may gradually disappear, leaving residual inflammation and scar. In other cases the necrotic region gradually evolves into a pseudocyst or a region of organized pancreatic necrosis (Fig. 94-13). In a minority of patients, a persistent sequestrum of sterile pancreatic necrosis is responsible for ongoing symptoms or persistent organ failure despite prolonged supportive care. Drainage of sterile necrosis may be considered in patients with persistent symptoms or organ failure, as described later in this chapter.

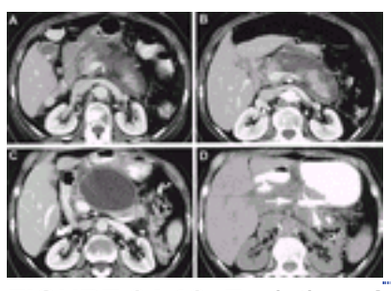

FIGURE 94-13. Evolution of central necrosis. **A**: Necrosis of the pancreatic body in a patient with gallstone pancreatitis. **B**: One week after onset of pancreatitis. **C**: Seven weeks after onset. **D**: The collection has resolved after endoscopic drainage with placement of multiple internal stents (*arrows*).

The term “organized pancreatic necrosis” has been used to describe a region of necrosis that is at least 4 weeks old and has developed a wall, but still contains necrotic material. [145] The distinction between organized pancreatic necrosis and pseudocyst has importance when nonsurgical drainage is considered. Aggressive endoscopic or percutaneous techniques are needed to adequately remove the thick necrotic material. Attempts at drainage may otherwise result in a poorly drained, infected collection. CT often underestimates the complexity of an organized area of pancreatic necrosis, and both EUS and MR have been used to identify solid material in these collections.

Pseudocysts

Pseudocysts are collections of pancreatic fluid contained by a wall of fibrosis or granulation tissue. The presence of a well-defined wall distinguishes pseudocysts from acute fluid collections and pancreatic necrosis. An epithelial lining is absent (in contrast to true pancreatic cysts). Formation of a well-defined wall generally requires at least 4 weeks. Pseudocysts usually contain fluid rich in pancreatic enzymes, and often contain particulate matter.

The advent of pancreatic imaging by ultrasonography and CT led to the realization that pancreatic pseudocysts are common and appear in up to 10% of patients with acute pancreatitis. [146] Although they may form most frequently in patients with alcoholic pancreatitis, up to 30% of cases arise in gallstone pancreatitis. The most common presenting symptom of a pseudocyst is upper abdominal pain that is either constant or postprandial. Other symptoms referable to a pseudocyst include early satiety and nausea and vomiting secondary to gastric outlet obstruction. Pseudocysts may also enlarge rapidly and rupture, hemorrhage, leak pancreatic fluid, obstruct the biliary tract, compress adjacent vessels, erode into surrounding structures, extend into the mediastinum, and become infected.

The risk of complications varies with the size of a pseudocyst and its age. Symptomatic pseudocysts may also be more prone to complications. Early reports based on ultrasound detection of pseudocysts in persistently symptomatic patients found a complication rate of 30% to 50% in pseudocysts that persisted for more than 6 weeks, and were the basis for recommendations to drain all persistent pseudocysts. More recent studies of asymptomatic pseudocysts detected with liberal use of CT document a low incidence of complications, even in large pseudocysts, and showed that most pseudocysts less than 6 cm in diameter resolved over time. A third of lesions more than 10 cm in diameter also resolved with observation alone. [146], [147]

Indications for early drainage include infection or progressive enlargement of the pseudocyst. Symptomatic pseudocysts typically require drainage. In the absence of symptoms, pseudocysts can be followed conservatively, although large lesions that fail to shrink with time should probably be drained. Pseudocysts can be drained endoscopically, percutaneously, or surgically. If there is a question of true pancreatic cyst or pancreatic cystic neoplasm rather than pseudocyst, surgical drainage provides the opportunity to obtain a large piece of the cavity’s wall for histological examination. This is especially important when a pancreatic “pseudocyst” is found in the absence of a clinical bout of pancreatitis, or in recent pancreatitis of unclear etiology (Fig. 94-14).

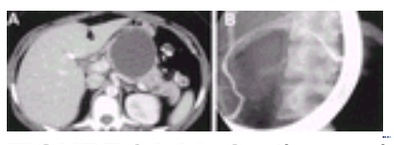

FIGURE 94-14. Cystic neoplasm simulating a pseudocyst. The patient developed epigastric pain without a discrete episode of pancreatitis. **A**: Computed tomography demonstrates a round fluid collection of the pancreatic body with a well-defined wall. **B**: Endoscopic retrograde cholangiopancreatography (ERCP) shows bowing of the pancreatic duct around the lesion, without communication. Pathology examination of the resected lesion showed cystadenocarcinoma.

The location of a pseudocyst and its relationship to the pancreatic duct can affect the approach taken for cyst drainage. Pancreatography may be helpful in delineating obstruction of the pancreatic duct and ductal communication with a pseudocyst. Those that do not communicate are amenable to either internal or external drainage. Pseudocysts that communicate with the duct, especially when the duct is strictured or disrupted, often resolve slowly when drained externally, and are best considered for internal drainage. Pseudocysts of the pancreatic head may be best treated with transpapillary stenting, especially if the cyst communicates with the pancreatic duct and there is an associated ductal stricture. [148] Pancreatography may be irrelevant if endoscopic internal drainage is planned, especially for pseudocysts of the pancreatic body and tail.

Endoscopic or percutaneous therapies are attractive nonsurgical options for pseudocyst drainage in many cases. Endoscopic techniques include transmural drainage into the stomach or duodenum with placement of an internal stent between the pseudocyst and the gut lumen. A high incidence of major hemorrhage was initially reported with endoscopic drainage, but use of an over-the-wire Seldinger technique, rather than electrocautery, may lower the risk of major complications such as bleeding or perforation to less than 5%. [149] EUS can facilitate pseudocyst drainage, particularly when an obvious bulge is not visible endoscopically, and may further decrease the risk of major hemorrhage by allowing the endoscopist to identify and avoid larger vessels between the stomach and pseudocyst lumens. [150] Endoscopic drainage of infected pseudocysts has been described.

Percutaneous drainage is also a well-established method for draining sterile or infected pseudocysts. When technically possible, a transgastric approach is favored, as internal drainage into the stomach through a mature catheter tract can result. External drainage may otherwise result in persistent drainage of pancreatic juice through the external fistula created by the catheter.

Incomplete pseudocyst drainage by either endoscopic or percutaneous means may result in infection of the pseudocyst contents. This is particularly common in patients with “organized pancreatic necrosis,” in whom the organized collection may have the CT appearance of a fluid-filled pseudocyst. A complex collection should be suspected in patients with necrosis that has evolved into a pseudocyst-like structure on serial CT scans. EUS or MR may be used to assess for solid material in the cavity prior to drainage. If solid matter is present in the collection more aggressive endoscopic or percutaneous techniques are necessary for adequate treatment, such as placement of multiple stents and a nasocystic tube to allow lavage of the pseudocyst cavity.

Fluid contents of a pseudocyst should be sampled during drainage for several reasons. First, a high amylase concentration within a cyst probably reflects its persistent communication with the pancreatic duct and bodes a poor outcome for external drainage. Second, symptoms of pseudocyst infection may be indolent and suspected only after bacterial and fungal culture. Third, true cystic lesions of the pancreas, including cystic neoplasms, may be mistaken for pseudocysts. Thus, amylase, viscosity, cytology, and tumor markers (including carcino embryonic antigen [CEA]) should be obtained from cyst aspirates when possible.

Pancreatic Abscesses

Pancreatic abscesses are circumscribed intra-abdominal collections of pus, usually within or in close proximity to the pancreas. In contrast to infected pancreatic necrosis, a pancreatic abscess contains little or no necrosis and typically presents later in the course of the illness, often more than 4 weeks after the onset of acute pancreatitis. Clinical and laboratory findings of acute pancreatitis have usually resolved before pancreatic abscess is detected. Because the infected collection is walled off and the surrounding tissue is viable, spread of infection to adjacent tissues is uncommon. Percutaneous drainage of pancreatic abscess is usually successful, although a minority of patients will require surgery. [151] Endoscopic transpapillary drainage is also feasible when the abscess cavity communicates with the pancreatic duct. [152]

Ascites and Fistulae

Several processes may contribute to the formation of ascites in acute pancreatitis. Peritoneal inflammation resulting from chemical peritonitis usually generates a small amount of fluid. Attendant chronic liver disease may contribute varying amounts of ascites. Pancreatic ascites refers to the presence of large amounts of fluid rich in pancreatic secretions and results from a communication between the pancreatic duct or a pseudocyst and the peritoneal cavity. Pancreatic ascites produces a gradual increase in abdominal girth, and is sometimes associated with subcutaneous fat necrosis. Since the pancreatic enzymes usually remain inactive, peritonitis is uncommon. The diagnosis should be suspected when ascites fails to respond to diuretic therapy and the serum amylase level is persistently elevated. Marked elevations of the ascitic fluid amylase and ascites protein concentrations above 3 g/dL are typical of pancreatic ascites. The site of ductal disruption can sometimes be

identified by CT, but ERCP is often required. [106]

Three forms of treatment have been used to treat massive pancreatic ascites. Endoscopically placed stents can be used to bridge the ductal disruption. Continuous infusion of somatostatin or octreotide with or without prior drainage of the ascites has also been used. However, some patients will require surgical drainage or pancreatic resection.

Pancreatic fistulae usually arise after a bout of alcoholic pancreatitis or from pancreatic pseudocysts, but also may form after pancreatic trauma, acute pancreatic duct disruptions, or pancreatic surgery. Anterior pancreatic ductal discontinuities can generate ascites, whereas posterior disruptions may result in fistulae that burrow through to the skin or track through the retroperitoneum. Fistulae sometimes extend through the esophageal or aortic hiatus into the chest, producing pleural effusions or mediastinal collections rich in amylase. Internal fistulae may communicate with the colon, gallbladder, and small bowel. Fistulae to vascular structures, such as the portal vein, have rarely been reported, and are often associated with subcutaneous fat necrosis.

The fluid from a pancreatic fistula has high protein and amylase content. The anatomy of an external fistula may be revealed by a fistulogram. Although ERCP is the most useful test for identifying an internal fistula, CT scanning may be very useful for fistulae that communicate with the thoracic cavity. Spontaneous healing is more common for cutaneous fistulae than internal fistulae. Drainage of fluid collections by thoracentesis or paracentesis may help to heal serosal discontinuities. Decreasing pancreatic secretion with somatostatin analogs has been reported to promote the resolution of fistulae, particularly those with cutaneous communications. Stenting of the pancreatic duct or surgical resection, however, are often required for internal fistulae.

Vascular and Splenic Complications

Pancreatitis infrequently results in bleeding or thrombosis of peripancreatic vessels. The commonest venous complication of pancreatitis is occlusion of the splenic vein, which is adjacent to the posterior superior margin of the pancreas throughout most of its course. Splenic vein thrombosis results in splenomegaly, gastric varices, and rarely colonic varices. Occlusion of the superior mesenteric vein can also occur, causing mesenteric venous congestion and small bowel edema and ischemia. Severe pancreatic inflammation or a large pseudocyst may thrombose or compress these vessels. When acute thrombosis of these veins is detected thrombolytic therapy is sometimes considered, but there are little data regarding the safety of such therapy and the risk of retroperitoneal hemorrhage in patients with ongoing pancreatitis.

Arterial hemorrhage may also occur as a result of pancreatitis, and is a ife-threatening event. This typically occurs when a pseudocyst erodes into a pancreatic artery, transforming the pseudocyst cavity into a pseudoaneurysm whose walls are partially formed by the artery and partially by the pseudocyst. (Fig. 94-15). The splenic artery may be involved if the pseudocyst is in the pancreatic body or tail, while the gastroduodenal artery or branches of the pancreaticoduodenal arcade are typically affected in the head of the pancreas. If the pseudocyst communicates with the pancreatic duct, hemosuccus pancreaticus (or bleeding into the duodenum via the pancreatic duct and ampulla) ensues. CT scanning and angiography are used to make the diagnosis. The bleeding artery often can be treated by embolization, but sometimes requires surgical ligation and, occasionally, broad surgical resection. [153] If hemorrhage is controlled angiographically, subsequent endoscopic treatment of the pseudocyst has been described. [154]

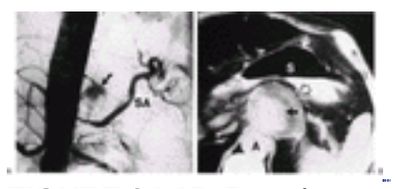

FIGURE 94-15. Pseudoaneurysm complicating acute pancreatitis. **A**: Angiogram demonstrates pseudoaneurysm arising from the splenic artery (*SA*) and a jet of contrast leaking from the pseudoaneurysm (*arrow*). **B**: Dynamic contrast-enhanced computed tomography of the same patient demonstrates the pseudoaneurysm (*open arrow*) with contrast enhancement (*black arrow*) anterior to the aorta (*A*) and posterior to the stomach (*S*). (Courtesy of Caroline Taylor, M.D., West Haven, CT.)

Extension of a pseudocyst, or occasionally inflammation alone, from the pancreas into the spleen may lead to a splenic hematoma and rupture. This complication is most often associated with chronic disease and may present with abdominal pain that is referred to the left shoulder, fever, and an abdominal mass. Ultrasonography or CT suggests the diagnosis. Arteriography may demonstrate splenic artery pseudoaneurysms. Emergency splenectomy is required for splenic rupture, and prophylactic splenectomy has been advocated by some authors even when splenic pseudocysts are asymptomatic. [155]

Gastrointestinal Obstruction

Severe acute pancreatitis may result in compression of the duodenum, with gastric outlet obstruction. Bile duct obstruction may also occur. These forms of gastric or biliary obstruction will usually resolve unless there is underlying chronic pancreatitis or pseudocyst.

It is common for pancreatic inflammation to extend to the transverse colon and splenic flexure. Obstruction secondary to spasm and edema may cause the classic colon cutoff sign, with gas present in the right colon but absent distal to the region of obstruction. This finding is seen in 1% to 10% of patients with acute disease, and cecostomy is occasionally required to prevent colonic perforation in patients with colonic obstruction progressive dilation of the proximal colon. [156] Extension of pancreatic inflammation may lead to persistent colonic strictures, fistulae, or even perforation. During surgery for acute pancreatitis, especially when performed for debridement, the colon is particularly susceptible to devitalization and subsequent perforation. Rarely, erosion into colonic blood vessels may result in acute lower gastrointestinal hemorrhage or inferior mesenteric vein thrombosis and colonic infarction.

SYSTEMIC COMPLICATIONS

Systemic complications of acute pancreatitis may be due to the direct toxicity of circulating activated pancreatic enzymes, the systemic inflammatory response to pancreatic injury and necrosis, circulating endotoxins due to bacterial translocation from the gut, or combinations of these factors. Organ failure can result, and may be manifested as respiratory insufficiency, shock, a sepsis-like syndrome, renal failure, gastrointestinal bleeding, and confusion or coma. [2] Coagulopathy and disseminated intravascular coagulation may also occur. The syndrome of multiple organ failure (MOF) may develop. Organ failure is seen uncommonly in interstitial pancreatitis, develops in about 50% of patients with pancreatic necrosis, and is an independent predictor of mortality. [157] Early mortality in acute pancreatitis is often due to multiple organ failure, while late mortality is more often related to infection.

Pulmonary

The most common form of organ failure in acute pancreatitis is pulmonary insufficiency due to the adult respiratory distress syndrome (ARDS). This hypoxemic state is associated with a normal pulmonary venous wedge pressure, decreased pulmonary compliance, and pulmonary capillary injury. It is often delayed in onset. Patients with hypertriglyceridemia are at increased risk for developing the complication. In patients with severe ARDS, pulmonary artery pressure monitoring and respiratory support with positive end expiratory pressure are generally required.

Pulmonary function may be further affected by pleural effusions and decreased diaphragmatic movement due to ascites or retroperitoneal inflammation. The contribution of decreased diaphragmatic movement is unclear, but may be a factor in the increased risk of a pulmonary death in those with acute pancreatitis and obesity. Effusions are reported in 4% to 17% of patients with acute pancreatitis; up to 50% may be associated with pseudocysts or pancreatic fistula. Most effusions are found on the left side or are bilateral, and most resolve spontaneously. [121] Early appearance of effusion has been associated with more severe disease. Thoracentesis is suggested when needed for diagnosis, to exclude infection, or when the size of the effusion results in respiratory compromise. Marked elevation of the pleural fluid amylase level may suggest an underlying pancreatic fistula, which may respond to antisecretory or endoscopic therapy.

Fat Necrosis

Clinically evident fat necrosis in regions distant from the pancreas is seen in less than 1% of patients with acute pancreatitis. Pancreatic enzymes have been detected in areas of fat necrosis and are presumably responsible for initiating this injury. The initial injury may arise from the cleavage of adipocyte plasma membrane glycerosphingolipids by phospholipase A. Adipocyte-associated or circulating lipase activities may then enter the cell and convert triglycerides to monoglycerides and toxic free fatty acids. Several clinical syndromes are associated with fat necrosis. Cutaneous necrosis generates widely disseminated, raised, erythematous, and tender nodules of less than 2 cm in diameter. The process is frequently associated with fever and eosinophilia. [158] The nodules typically resolve in days to weeks without residual; however, some may rupture and heal with hyperpigmentation. Fat necrosis may affect the marrow cavity and lead to vascular damage and bone infarction. The typical bone lesions are frequently painless, osteolytic, develop 3 to 6 weeks after the onset of pancreatitis, and involve the ends of long bones such as the femur. Synovial pad fat necrosis may be associated with arthritis. The increased amounts of free fatty acids recovered from such joints may initiate this inflammatory process. Fat necrosis may rarely affect the middle ear.

Other Complications

Renal insufficiency is not uncommon in acute pancreatitis. Confusion and coma can occur and may be due to cerebral edema. Gastrointestinal hemorrhage may be due to mucosal erosions and ulcerations, gastric varices, colonic varices, or a pancreatic pseudoaneurysm, as described earlier in the section on “Vascular and Splenic Complications.” Temporary blindness has been reported in pancreatitis and may be attributed to the retinal ischemia induced by leukocyte emboli.

ACUTE TREATMENT

The treatment of acute pancreatitis is largely supportive. Pancreatitis patients are often in critical need of such care, and appropriate treatment is frequently lifesaving. Aggressive fluid replacement, management of pain and nausea, prompt identification and treatment of complications, nutritional support, and prophylaxis and treatment of infection are important elements of supportive care.

Some treatments have lost favor due to lack of convincing efficacy. These include peritoneal lavage and nasogastric suction. The use of nasogastric suction in the past was based on the unproved concept that removing the acid-dependent stimulation of the pancreas would allow the inflamed organ to rest and promote recovery. However, clinical trials showed no benefit to nasogastric suction for treatment of underlying pancreatitis. Nasogastric suction should be reserved for patients with persistent emesis and the occasional patient with evidence of colonic obstruction complicating acute pancreatitis.

Supportive care includes careful observation of the patient. Regular assessment of vital signs and oxygen saturation should be a routine part of care. Decreased urine output, hemodynamic instability, or worsening oxygen saturation should lead to prompt evaluation and treatment, perhaps in an intensive care unit.

Parenteral narcotics are often required for treatment of pain in acute pancreatitis, and meperidine has traditionally been the narcotic of choice because it does not cause pancreatitis or significantly alter sphincter of Oddi function. [45] However meperidine is a less effective analgesic than many other narcotics, and accumulation of meperidine metabolites may cause paranoia, psychosis, or seizures. Morphine administration is discouraged in acute pancreatitis because it can cause sphincter of Oddi spasm and, rarely, pancreatitis. The effect of other narcotics on the sphincter of Oddi is less well studied, but hydromorphone or other agents may be appropriate in some cases, particularly if meperidine is ineffective, large doses are required, or prolonged use of narcotics is anticipated.

Fluid and Electrolyte Replacement

The cornerstone of supportive therapy is maintenance of an adequate intravascular volume. The analogy between the injury of severe pancreatitis and a burn is a useful one: pancreatitis often leads to “third spacing” of fluid and intravascular volume depletion. This may in turn worsen pancreatic perfusion and exacerbate the pancreatitis. Hemoconcentration is associated with the development of pancreatic necrosis. [137] The clinician should regularly reassess intravascular volume and aggressively replace fluids and electrolytes to avoid volume depletion. Often such assessment is made on the basis of urine output and vital signs. An elevated hematocrit or renal insufficiency may be signs of inadequate volume replacement. In severely ill patients, central venous pressure monitoring may be useful. Although dextrans have been used to expand intravascular volume and enhance pancreatic blood flow, their use has not been widely accepted.

Metabolic Abnormalities

Hyperglycemia is present in some patients with acute pancreatitis. It is wise to err on the side of maintaining the blood glucose level in the high range for several reasons. Limited glucagon and glycogen reserves, especially in alcoholics, diminish the ability of patients to respond to hypoglycemia. Furthermore, hyperglycemia is often transient, improving when the pancreatitis has resolved.

Serum calcium levels may be decreased during acute pancreatitis and levels below 7.5 mg/dL are associated with a poor prognosis. Factors that account for this decrease are sequestration of calcium by free fatty acids that have been generated by peritoneal fat necrosis and dilution by intravenous administration of calcium-poor fluids. In addition, hypoalbuminemia may result in low total serum calcium with normal serum ionized calcium. [159] Humoral factors, such as parathyroid hormone resistance associated with hypomagnesemia and hypersecretion of calcitonin and glucagon, have been implicated in hypocalcemia, but are unlikely to play a major role. Calcium should be slowly administered to patients with decreased ionized serum calcium.

Hyperlipidemia may lead to pancreatitis, accompany alcoholic pancreatitis, or be a consequence of acute pancreatitis. Therapy should generally not be instituted for hyperlipidemia encountered during acute pancreatitis, but values should be reassayed after recovery. One exception is patients with severe pancreatitis and respiratory failure. In this subgroup, and in the occasional patient with extreme hyperlipidemia, lowering serum triglycerides with plasmapheresis may be useful. [160]

Inhibition of Enzymes, Secretion, and Inflammation

Pharmacological therapy could theoretically be used to limit the severity of pancreatitis at several levels: by inhibiting active pancreatic enzymes, inhibiting pancreatic secretion, or interrupting the inflammatory cascade. The first agents to be tested extensively in humans were drugs that inhibit activated pancreatic enzymes. Of these “protease inhibitors” the best studied is gabexate mesilate, a drug that is not currently available in the United States.

Most studies of gabexate have shown a benefit in acute pancreatitis that did not reach statistical significance. Two meta-analyses have evaluated controlled clinical trials of gabexate in acute pancreatitis, and these metaanalyses reached similar conclusions. Gabexate does not reduce mortality from acute pancreatitis, but does appear to reduce the incidence of complications by about 30%. [161], [162] While a recent additional trial showed decreased early mortality in patients with severe disease, it is unlikely to alter the basic conclusions of the metaanalyses, which drew on studies enrolling over 800 patients with acute pancreatitis. [163] The lack of a more decisive clinical effect is consistent with the fact that activated pancreatic enzymes play their most important role early in the course of pancreatitis, before the patient presents for treatment.

Somatostatin and octreotide, two agents that inhibit pancreatic secretion, have also been extensively studied in clinical trials. When only randomized controlled trials were considered, metaanalysis showed a statistically significant reduction in mortality with somatostatin therapy, and a lesser benefit with octreotide that did not reach statistical significance. [161] Curiously, despite their effect on mortality, neither agent was shown to decrease complication rates. The lack of demonstrated benefit with octreotide therapy may reflect its effects on sphincter of Oddi function and pancreatic blood flow. While somatostatin appears to have a significant impact on mortality, it is not currently available in the United States.

Severity of the systemic immune response, as measured by blood levels of circulating proinflammatory cytokines, correlates with the development of local and systemic complications in pancreatitis. The inflammatory response often continues to increase after the patient is hospitalized, suggesting that pharmacological blockade of proinflammatory cytokines soon after hospital admission could ameliorate the consequences of pancreatitis. Although animal studies support this hypothesis, the one cytokine inhibitor tested in a large clinical trial did not have a significant effect on complication rates or mortality. [164] Experimental studies suggest that specific cytokines may promote injury in distinct tissues. Therefore, a different cytokine inhibitor or a combination of inhibitors might be needed to be fully

beneficial.

Enteral and Parenteral Nutrition

One long-standing tenet of supportive treatment has been to avoid pancreatic stimulation by giving the patient nothing to eat during the acute phase of illness. Patients with mild disease who rapidly improve and resume oral intake require no special form of nutritional support, but those with more protracted disease are generally given nutritional therapy.

Controlled trials of total parenteral nutrition (TPN) have shown little or no benefit in patients awaiting surgery or undergoing chemotherapy. While there are little controlled data in pancreatitis, some retrospective studies suggest that pancreatitis patients given early parenteral hyperalimentation are at increased risk for complications such as sepsis and pancreatic infection. While intravenous lipid infusion does not stimulate pancreatic secretion, it can produce hyperlipidemia that can then trigger further pancreatitis. Since TPN-induced elevations of the triglyceride level have been reported to cause pancreatitis, it seems prudent to monitor triglyceride levels and attempt to keep them below 500 mg/dL in patients with pancreatitis receiving TPN. [165]

A growing body of evidence suggests that enteral feeding is safe in pancreatitis, and preferable to parenteral nutrition. Oral feeding stimulates pancreatic secretion, but feeding directly into the distal small bowel may result in negligible pancreatic stimulation. [166] Theoretically, enteral feeding could limit the changes in gut mucosal integrity seen in pancreatitis, with a resultant decrease in bacterial translocation, systemic immune response, and pancreatic infection. These considerations have led to several controlled trials of enteral versus parenteral nutrition in pancreatitis.

In one randomized study enteral feeding via a nasojejunal tube was compared to TPN in patients with mild acute pancreatitis mostly attributed to alcohol. [167] Enteral feeding was begun within 48 hours of admission and was well tolerated in all cases. There was no significant difference between the two groups in major end points, including time to normalization of serum pancreatic enzyme levels, resolution of pain, time to oral feeding, or incidence of infection. Several patients tolerated enteral feeding well despite failing subsequent trials of oral intake, or had recurrent pancreatic symptoms when the tip of the enteric feeding tube fell back into the stomach.

Two other studies have randomized patients with severe disease to TPN or early enteric feeding. In both studies enteric feeding was safe and less costly than TPN, and was associated with improved outcomes in either laboratory markers of inflammation or in the incidence of infections and other complications. [166], [168] Patients with clinical evidence of ileus generally tolerated enteric feeding well. An additional study showed no benefit of enteral feeding on markers of immune response or intestinal permeability, but tube dislodgement was common and patients received only 20% of their predicted caloric requirements. [169]

Enteral feeding can be recommended in acute pancreatitis, especially when it is unlikely that the patient will quickly resume oral intake. Early initiation of nasojejunal feeding (within 48 hours of admission) is safe and may improve the course of the illness in patients with severe disease. The major impediment to this therapy is the logistical difficulties of placing a nasojejunal tube. Methods for simple enteric feeding tube placement have been described, including both fluoroscopic techniques and bedside transnasal endoscopy. [170] Isocaloric full-strength formula can be used, beginning at 25 mL/h, and increasing the rate every 4 hours until the patient's target rate is met. Feeding is generally successful even in the presence of ileus, but high gastric residuals or symptoms of nausea and abdominal fullness may prompt a decrease in the infusion rate.

When the patient is ready for oral feeding, tradition holds that they should be started on a low-fat diet, but there is little evidence that this restriction affects recovery. Indeed, lumenal amino acids are also potent stimulants of pancreatic enzyme secretion. Probably more important than fat restriction is the subjective response of the patient to feeding. The most reliable marker for beginning oral feeding is hunger. Steady improvement in pain and tenderness allowing discontinuation of parenteral pain medication is also a useful indicator. Persistently abnormal serum enzymes or CT scans in the absence of symptoms or complications should not dissuade the clinician from feeding a hungry patient.

Management of Necrosis

Prophylaxis of Infected Pancreatic Necrosis Infection of pancreatic necrosis is a life-threatening complication and a major risk factor for death from pancreatitis. [171] Strategies for the prevention of infected necrosis have been evaluated, including systemic antibiotic prophylaxis and selective decontamination of the gut. Older studies of antibiotics to prevent pancreatic infection did not find a benefit. These studies probably enrolled many patients at low risk for infection, and did not select antibiotics based on their ability to penetrate pancreatic tissue. Subsequent investigations demonstrated that penetration of pancreatic tissue by ampicillin and aminoglycosides is poor, whereas imipenem-cilastatin, mezlocillin, third generation cephalosporins, metronidazole, and quinolones exhibit moderate to excellent penetration. [172] More recent prospective studies have demonstrated benefits to prophylactic intravenous antibiotic therapy in patients with pancreatic necrosis. Prophylactic imipenem-cilastatin, cefuroxime, and a combination of ceftazidime, amikacin, and metronidazole have been tested against placebo, and each regimen resulted in a significant reduction in the incidence of infectious complications. [173], [174] and [175] Only the cefuroxime study showed a significant reduction in mortality, but this study has been criticized for a high rate of nonpancreatic and staphylococcal infections, failure to reduce the incidence of pancreatic infections significantly, and changes in the antibiotic regimens given to most patients in the course of the study. Two metaanalyses of antibiotic prophylaxis concluded that prophylaxis decreases sepsis and mortality in patients with necrosis. [176], [177] In a recent comparative study, pefloxacin was less effective than imipenem-cilastatin for prevention of infectious complications. [178] Taken together, these data support the prophylactic administration of antibiotics. Patients with necrosis documented by CT or, in the absence of early contrast-enhanced CT, patients with severe pancreatitis (who are at highest risk for necrosis) should be treated. Treatment should generally be started within 48 hours of admission and continued for 2 to 3 weeks, and imipenem-cilastatin (500 mg every 8 hours) is probably the most effective agent. Unfortunately, prophylactic antibiotics do not eliminate the risk of infected necrosis. Recent large series of patients treated with prophylactic antibiotics report an incidence of infected necrosis as high as 32%, with a shift toward gram-positive and fungal infections. [142] Fluconazole penetrates pancreatic tissue [179] but its role in prophylaxis of pancreatic fungal infections is unproven. An alternative to systemic antibiotic prophylaxis is "selective decontamination" of the gastrointestinal tract with lumenal antibiotics. The rationale for this approach is the finding that bacterial translocation from the gut is the likely source of infection in pancreatic necrosis. [140] Studies in animal models of severe pancreatitis show a benefit to intralumenal antibiotics and intestinal lavage. One large controlled clinical trial has tested this prophylactic strategy in patients with severe pancreatitis, who were randomized to receive no antibiotic prophylaxis or colistin sulfate (200 mg), amphotericin (500 mg), and norfloxacin (50 mg) orally every 6 hours, and in a rectal enema daily. Intravenous cefotaxime was also given until gram-negative bacteria were eliminated from the mouth and rectum. The incidence of pancreatic infection was halved by selective decontamination, and mortality was significantly decreased as well. [141]

Treatment of Infected Pancreatic Necrosis As discussed earlier in this chapter, the diagnosis of infected pancreatic necrosis should be entertained in patients with systemic signs of infection, or in patients with persistent or worsening organ failure despite supportive care. Diagnosis requires a high level of suspicion. Currently available noninvasive laboratory and imaging tests do not reliably exclude infection, and CT-guided aspiration of necrotic areas for Gram stain, fungal stain, and cultures are the most important diagnostic modalities. Anecdotal reports suggest that an occasional patient with infected necrosis will survive with antibiotic treatment alone. [180] However antibiotic treatment alone is generally inadequate for treatment of documented infected necrosis, and the infected necrotic material should be removed. Prompt surgical debridement has been the standard therapy, and repeated reexploration for further debridement is often necessary. This treatment reduces mortality to between 20% and 60%. [181] Better surgical outcomes are in part due to changes in technique, including open packing of the wound and prolonged postoperative lavage. Percutaneous drainage of infected necrosis was long thought to be an inadequate therapy, since the solid necrotic material would not drain out through small caliber percutaneous catheters, and the drainage tube would only serve to introduce other organisms into the necrotic debris. However, recent reports demonstrate that percutaneous catheter drainage may be effective. [182], [183] In contrast to percutaneous treatment of pseudocysts or pancreatic abscesses, repeated interventions and aggressive techniques are required to adequately remove the infected material. Techniques may include placement of multiple large bore (28 French) drainage tubes, frequent bedside catheter irrigation, periodic vigorous manual irrigation under fluoroscopic or CT guidance, and endoscopy of the percutaneous catheter tract for removal of debris using jet irrigation, forceps, and snares. Reported mortality rates of 15% to 20% from expert centers compare favorably with surgical series. Complications include hemorrhage from retroperitoneal vessels. Patients with persistent pancreatic sepsis requiring surgery had a high mortality rate.

Drainage of Sterile Necrosis The role of surgery in sterile necrosis has been debated. Although routine surgical debridement of sterile pancreatic necrosis was advocated in the past, the weight of available evidence does not show a benefit to surgery over supportive medical care. [184] Nevertheless, intervention may be warranted in a small subset of patients in whom sterile necrosis is associated with either a deteriorating course despite maximal supportive care, or prolonged organ failure. [185] In such patients the percutaneous techniques described previously may be appropriate as well. Some patients with sterile necrosis require intervention not because of organ failure or an ongoing systemic inflammatory response, but because of recurrent pancreatic symptoms and an enlarging collection with attempts at refeeding. This is particularly common in patients with "central necrosis" (extensive necrosis of the pancreatic body) and is usually associated with a persistent disruption of the pancreatic duct, resulting in an isolated pancreatic tail and ongoing leakage of pancreatic juice from the disruption. Internal drainage of the collection may suffice, but resection of the pancreatic tail is required in some cases. [182], [186] Endoscopic transmural or transpapillary drainage of regions of organized pancreatic

necrosis has also been described. [145] In contrast to patients drained surgically or percutaneously, patients treated endoscopically have generally been less ill, without organ failure, and were treated later in the course of their illness, on average 6 weeks after onset of pancreatitis. The drained collections had liquefied to some extent and developed a wall. Endoscopic drainage of “organized pancreatic necrosis” usually requires placement of multiple stents and a transnasal or transgastrostomy catheter for lavage of the collection. The endoscopist should be prepared to perform repeated endoscopic interventions over time as necessary to ensure adequate drainage.

ERCP and Sphincterotomy in Biliary Pancreatitis

Urgent ERCP in Gallstone Pancreatitis Persistent bile duct stones are associated with more severe pancreatitis, and both surgeons and endoscopists have investigated the theory that prompt relief of ampullary obstruction by removal of an impacted stone would decrease the severity of gallstone pancreatitis. While studies of urgent surgery have yielded disappointing results, [187] endoscopic studies have identified a subgroup of patients who benefit from urgent ERCP. Several randomized studies have assessed the value of urgent ERCP in patients with gallstone pancreatitis. The first, performed in Great Britain, found that ERCP within 2 days of presentation decreased the complication rate of gallstone pancreatitis. [188] Sphincterotomy was done only if stones were present in the ampulla or bile duct. The benefit was limited to the subset of patients who were predicted to have severe pancreatitis on initial assessment using the modified Glasgow criteria (Table 94-4). These results suggested that urgent ERCP might alter the course of an episode of gallstone pancreatitis. A similar study subsequently reported from Hong Kong also found a decreased complication rate when urgent ERCP was performed and sphincterotomy was done for ampullary or ductal stones. [31] This second study also showed that benefit was limited to patients predicted on admission to have severe pancreatitis, using the simpler urea-glucose criteria. The Hong Kong study included some patients with other causes of pancreatitis, and probably included more patients with primary pigment stones of the bile duct. Complications were reported in greater detail, and the benefit of ERCP appeared to be largely due to prevention of bacterial cholangitis rather than amelioration of pancreatitis. Both of these studies found a benefit in patients predicted to have severe pancreatitis and showed that ERCP can be safely performed in this setting. It remains unclear if the benefit was due to a decrease in the severity of pancreatitis or to treatment and prevention of cholangitis. CT of the pancreas was not used in these studies to direct image the severity of pancreatitis. ERCP was not shown to prevent a predicted mild episode of pancreatitis from progressing in severity, and there were no statistically significant changes in survival. A third large study showed a significant improvement in mortality with early ERCP and sphincterotomy, but was only published in abstract form. [189] A fourth study with a similar design from Germany showed a trend toward higher mortality and more severe complications in patients undergoing early ERCP. [190] This study excluded patients with a serum bilirubin above 5 mg/dL, probably excluding many patients with persistently impacted stones while including patients with nonobstructing ductal stones. Since nonobstructing stones theoretically have little effect on either the development of cholangitis or the severity of pancreatitis, this study probably excluded those patients most likely to benefit from urgent ERCP. Unfortunately, none of these studies reported the location of calculi or the degree of obstruction these stones caused. Further studies using noninvasive methods for visualization of ampullary stones and pancreatic inflammatory changes may shed more light on the benefit achieved by ERCP and sphincterotomy. When should the clinician consider urgent ERCP? The clearest indication is the presence or suspicion of biliary sepsis. Anicteric patients with mild pancreatitis and no signs of cholangitis are unlikely to require urgent ERCP. Jaundiced patients and those with predicted severe pancreatitis, especially those who fail to improve over the first 12 to 24 hours of observation, may benefit from prompt bile duct evaluation. In these patients, newer and less invasive methods of demonstrating ductal or ampullary stones (such as MRCP, helical CT, and EUS) will probably be helpful in choosing patients to undergo urgent ERCP [128] (see Fig. 94-9). Traditionally, cannulation of the pancreatic duct has been avoided when possible in patients with acute biliary pancreatitis undergoing ERCP. Some experts have advocated intentional visualization of the pancreatic duct early in the course of severe acute pancreatitis for identification and endoscopic treatment of pancreatic duct disruptions. However some disruptions resolve spontaneously, [135] and instrumentation may introduce infection into a previously sterile collection. Controlled data evaluating this strategy are needed before it can be generally recommended. Similarly, a strategy of early ERCP and sphincterotomy in all patients with biliary pancreatitis cannot be generally recommended.

Preoperative ERCP in Gallstone Pancreatitis Most patients with gallstone pancreatitis rapidly improve with supportive management, and by the time of their cholecystectomy only a minority have bile duct stones. The bile duct should be imaged in such patients, and the choice of imaging technique depends on the likelihood of finding a bile duct stone, the surgeon’s ability to extract bile duct stones laparoscopically, and the confidence of the biliary endoscopist. Patients with gallstone pancreatitis whose serum liver tests and pancreatic enzyme levels steadily fall after admission have a low (8%) incidence of bile duct stones, [120] and are best evaluated with intraoperative cholangiography. Patients in whom any of these chemistries increase in the first 48 hours of admission, and patients with a dilated bile duct on ultrasound, have an intermediate (30%) incidence of ductal stones. These patients may also best be evaluated with intraoperative cholangiography, depending on the comfort and skill of the available surgeon and endoscopist. Preoperative MRCP or EUS may also be appropriate in this patient group. Direct visualization of a ductal stone on ultrasound, CT, MR, or EUS establishes a high risk (60% or greater) of a bile duct stone in gallstone pancreatitis, and such patients, as well as those with persistent jaundice, may reasonably undergo preoperative ERCP.

Sphincterotomy as Sole Therapy for Gallstone Pancreatitis Endoscopic sphincterotomy usually eliminates the common channel between the bile duct and pancreatic duct, which afterward enter the duodenum at slightly different locations. Sphincterotomy appears to prevent recurrence of gallstone pancreatitis in patients whose gallbladder remains in place. At least 15% of patients treated in this fashion will subsequently develop other complications of gallstones, however, including biliary obstruction, cholangitis, and acute cholecystitis. [191] These complications are most common within 6 months of sphincterotomy but may occur years later. In addition, a small percentage will develop stenosis of the sphincterotomy site. Cholecystectomy thus remains preferable to sphincterotomy in patients with uncomplicated pancreatitis who are fit for surgery. When the risks of cholecystectomy are high (as in patients with unstable cardiovascular disease or advanced cirrhosis), endoscopic sphincterotomy is a reasonable therapeutic alternative.

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CHAPTER 95

Chung Owyang

CHRONIC PANCREATITIS

The terminology surrounding the classification of inflammatory disease of the pancreas is confusing and based on both clinical and morphologic criteria. Since the 1970s, multiple classifications have been proposed. Differentiation by etiology allows for the prediction of responsible factors for pancreatitis but gives little information on potential overlapping pathophysiologic mechanisms. Clinical and pathological classifications by themselves have prognostic implications, with fulminant or hemorrhagic pancreatitis having a poor prognosis and edematous pancreatitis being associated with a good prognosis; only in a minority of patients is histological information ever obtained, however, and clinical classifications are not helpful pathophysiologically. In 1963, a symposium on pancreatitis was held in Marseilles, France, in which a clinical classification of pancreatitis was proposed, and this classification has since been widely used. [1] According to the Marseilles classification, pancreatitis can be divided into acute and chronic, with both types having relapsing and nonrelapsing varieties. The essential difference between acute and chronic pancreatitis is the presence of permanent and progressive morphologic or functional damage in the latter. Both subtypes are further classified into relapsing and nonrelapsing varieties depending on their clinical presentation. In acute pancreatitis, if the primary causes or factors are eliminated, clinical and morphologic restitution of the pancreas occurs. On the other hand, in the chronic form, anatomic or functional pancreatic damage remains even after the primary causes are eliminated.

The major drawback of the Marseilles classification is the frequent inability to distinguish acute and chronic forms of the disease based on clinical presentation. This prompted two subsequent international symposia in 1983 [2] and 1984 [3] to discuss the classification of pancreatitis. Some minor modifications of the original Marseilles classification were proposed. The classification of pancreatitis should be reduced to include only acute and chronic forms because it is frequently difficult to distinguish between recurrent acute pancreatitis and recurrent attacks of chronic pancreatitis, and several years' follow-up may be needed. Accordingly, chronic pancreatitis is defined as an inflammatory disease of the pancreas characterized by persistent and often progressive lesions, whereas lesions of acute pancreatitis regress when the cause is suppressed or removed. In an addendum, it was recognized that alcohol-induced pancreatitis may present acutely, and it is not inevitably progressive.

The second symposium of Marseilles [2] also proposed to distinguish obstructive chronic pancreatitis from other forms of chronic pancreatitis. Obstructive chronic pancreatitis is characterized by dilation of the ductal system, diffuse atrophy of the acinar parenchyma, and uniform fibrosis. In contrast to other forms of chronic pancreatitis, intraductal plugs or stones usually are rare or absent, and both structural and functional changes may improve when the obstruction is relieved. Based on morphology, biochemistry, molecular biology, and epidemiology, Sarles [4] reclassified chronic pancreatitis into four groups. The first and largest group, lithogenic pancreatitis (chronic calcifying pancreatitis), consists of five subgroups of stones: hereditary pancreatitis, transparent stones, nutritional pancreatitis, hypercalcemia pancreatitis, and pure calcium stones. The other three groups include obstructive pancreatitis caused by obstruction of pancreatic ducts preceding the onset of pancreatitis, with uniform distribution of the lesions caudal to the obstruction; inflammatory pancreatitis, which is characterized by diffuse fibrosis and the destruction of exocrine parenchyma with infiltration of mononuclear cells; and pancreatic fibrosis, characterized by silent, diffuse perilobular fibrosis.

INCIDENCE AND PREVALENCE

The prevalence of chronic pancreatitis in autopsy materials ranges from 0.04% [5] to 5%. [6] Epidemiologic studies based on clinical data are few. Most epidemiologic data are obtained either from retrospective studies [7], [8] and [9] or by calculation from data given in clinical material. [10], [11] The only prospective study on the incidence and prevalence of chronic pancreatitis was performed in Copenhagen, Denmark, in 1978 and 1979. [12] It showed an incidence of 8.2 new cases per 100,000 inhabitants per year, and a prevalence of 26.4 cases per 100,000 inhabitants. The epidemiologic studies clearly demonstrate that there is enormous difference in incidence rates between different areas. Alcohol consumption is considered to be the major factor in the development of chronic pancreatitis. This may well explain the low incidence (rate) of chronic pancreatitis in Japan, which has traditionally a very low alcohol intake. On the other hand, Switzerland has a substantially higher alcohol consumption than observed in Denmark, [13] but the incidence of chronic pancreatitis is lower in Switzerland than in Denmark. The reason for this discrepancy is unknown.

Environmental or hereditary factors may influence susceptibility to alcohol-induced pancreatitis. Alternatively, this may be related to regional differences in diagnostic criteria for chronic pancreatitis. Thus, figures for frequency of chronic pancreatitis differ markedly from one center to another. Most likely, this does not reflect a real

difference in frequency but points strongly to regional differences in patient selection and diagnostic criteria.

ETIOLOGY

Alcohol in Western societies (70% to 80%) and malnutrition worldwide represent the major etiologies of chronic pancreatitis. [14], [15] In addition, metabolic and mechanical disturbances, as well as hereditary disposition, have been implicated (Table 95-1).

Alcohol, 70%
Idiopathic (including tropical), 20%
Other, 10%
Hereditary
Hyperparathyroidism
Hypertriglyceridemia
Autoimmune pancreatitis
Obstruction
Trauma
Pancreatic divisum

TABLE 95-1 Etiology of Chronic Pancreatitis

Alcohol

Alcohol consumption is the major cause of chronic pancreatitis. In Western societies, about 70% of cases are caused by alcohol. [16] The incidence of pancreatitis found at autopsy in alcoholics is as high as 45%—50 times the rate in nondrinking controls. [17] There is a linear relationship between the logarithm of the risk for development of chronic pancreatitis and the mean daily consumption of alcohol. [18] Furthermore, it has been demonstrated that the risk for abstainers is lower than the risk for people drinking a low quantity of alcohol (i.e., 1 to 20 g/day). Therefore, it appears that there is no statistical threshold for alcohol toxicity but a continuous spectrum of individual thresholds. This makes it difficult to distinguish between low-threshold alcohol-induced pancreatitis and idiopathic pancreatitis. The duration of alcohol consumption is also important. For the same daily intake, the risk increased with duration, [19] but the type of alcohol beverages and the pattern of drinking (weekend versus daily users) have no influence on the risk for development of chronic pancreatitis. [20] This concept, however, has been challenged. [21]

In general, prolonged alcohol intake is required to produce symptomatic chronic pancreatitis (6 to 12 years), although the length of time required for histological damage in humans is unknown. [22] Clinically, alcoholic chronic pancreatitis is characterized by an initial period of recurrent acute attacks lasting 5 to 6 years, followed by the development of chronic pain, exocrine and endocrine insufficiency, or both. A natural history study [23] demonstrated a median time to exocrine insufficiency of 13 years in patients with alcoholic chronic pancreatitis and a median time to endocrine insufficiency of 20 years. Interestingly, not all patients with an acute episode of alcohol-induced pancreatitis progress to clinical chronic pancreatitis. It is generally accepted that at the time of the initial attack of pancreatitis, histological, chronic pancreatitis is already present. Two retrospective studies [24], [25] reported that 15% to 20% of patients, followed for up to 30 years after an initial attack of alcoholic chronic pancreatitis, did not develop pancreatic calcification or exocrine or endocrine insufficiency. The factors responsible for this variability in progression of disease are not clear but do not appear to be caused by differences in avoiding further alcohol. Patients with progressive disease were more likely to have more severe and more frequent acute clinical attacks of pancreatitis, and these patients frequently developed significant abnormalities affecting the main pancreatic duct. Kondo and associates [26] emphasized that alcohol requires other factors to induce chronic pancreatitis. They reported that the aberrant pancreas might not be susceptible to alcoholic pancreatitis. Anatomic differences in the aberrant pancreas, such as innervation and different ductal drainage, may explain the lack of damage after chronic alcohol ingestion. In addition, dietary factors may play a significant role. Both experimental and epidemiologic studies indicate the risk for alcohol-induced pancreatitis is higher in people with high-fat, high-protein diets. Experimentally, focal lesions of chronic pancreatitis developed in more than 50% of rats that ingested alcohol for 20 to 30 months. [27] The pancreatic juice of these rats contained higher protein concentrations than controls, a condition that appears similar to chronic pancreatitis in humans. In separate studies, Tsukamoto and colleagues [28] showed that in rats with low dietary intake of corn oil fat, chronic alcohol intoxication produced only mild pancreatic injury. Severe focal lesions of chronic pancreatitis, however, were observed in rats fed higher amounts of fat, which resulted in striking potentiation of alcohol-induced pancreatic injury. Observations from these experimental studies were supported by clinical and epidemiologic studies. It has been observed in many European countries that alcoholics consuming a high-fat, high-protein diet are predisposed to chronic pancreatitis. [5], [29], [30] and [31] Durbec and Sarles [18] noted that the risk was less with average consumption of fat but was increased by both high (more than 100 g) and low (less than 85 g) daily fat intake. Furthermore, the effects of alcohol, protein, and fat consumption appear to be additive. This may explain why chronic pancreatitis is more commonly observed in patients who drink more alcohol and eat more protein and fat than controls. [19] Observations made in Europe, however, are not confirmed by studies reported from the United States and Australia. [32], [33] and [34] It appears that whereas a high-fat or high-protein diet may predispose to pancreatitis, such a diet is not a prerequisite for development of pancreatitis in chronic alcoholics. Differences in genetic predisposition, quantity of alcohol, and type of fat consumed by various populations may explain some of these conflicting observations.

The pathogenic mechanisms of alcohol-induced chronic pancreatitis are uncertain. Results from a prospective clinicopathological study of alcohol-induced recurrent attacks of acute pancreatitis evolving to chronic pancreatitis [35] support the hypothesis of the *necrosis-fibrosis* sequence, originally proposed in 1946 by Comfort and associates. [36] In this study of resected pancreatic tissue or autopsy specimens in 73 patients and in 10 patients with both types of specimens, it was found that after a mean of 4.1 years from the onset, focal necrosis and mild perilobular fibrosis were the predominant lesions. In contrast, the major lesions in the autopsy specimens, as compared with previous resected tissues (mean interval between biopsy and autopsy, 8 years), were severe perilobular and intralobular fibrosis and calcification but not necrosis. This suggests that the initial lesion in alcohol-induced pancreatic injury is cellular necrosis and, over time, that fibrosis will occur in those patients developing chronic pancreatitis. The cellular mechanisms by which alcohol produces chronic pancreatitis are not known. Inferences about the pathophysiologic basis of alcohol-induced progressive pancreatic inflammation can be made from the alcohol's effects on pancreatic secretion and the known pathological changes described by Sarles and others. Chronic alcoholism causes an increase in basal secretion of proteases, amylase, and lipase, and a decrease in trypsin inhibitor in rats. [37] Furthermore, an increased responsiveness of the pancreas to cholecystokinin (CCK) stimulation has been reported in dogs [38] and humans. [39] In addition to the perturbation in the content and secretion of pancreatic enzymes, chronic alcohol consumption also causes perturbation in the lysosomal enzymes; acid phosphatase was increased in cell fractions, and cathepsin B was increased in the mitochondrial-lysosomal fraction and the zymogen granule fractions of the pancreas from alcohol-fed rats. [40] Therefore, it is conceivable that alcohol produces pancreatitis by interfering with the intracellular transport and discharge of digestive enzyme, causing colocalization of digestive and lysosomal hydrolases, a condition conducive to the initiation of autodigestion. The augmented responsiveness of the exocrine pancreas may account for the high-protein, low-bicarbonate, and low-volume pancreatic secretory output after chronic alcohol consumption. [41] Theoretically, protein precipitates would be formed, particularly in low-flow-rate areas such as the secondary pancreatic ducts. This may explain the frequent involvement of the secondary duct, with relative sparing of the main pancreatic duct, in early stages of chronic pancreatitis.

The discovery of a pancreatic stone protein (lithostatin) found in pancreatic juice and calculi has generated considerable interest because this protein is capable of inhibiting the formation of insoluble calcium salts in a supersaturated milieu. [42], [43] Lithostatin has been found to be decreased in the pancreatic juice of patients with alcoholic pancreatitis and in some patients with nonalcoholic chronic pancreatitis. [44] Hence, it is conceivable that a deficiency of lithostatin may play an important role in the development of pancreatic calcification. [42] The importance of this protein remains unclear, however, because not all studies have shown a reduction in lithostatin in pancreatic juice from patients with chronic pancreatitis. [45]

One report [46] showed that GP2, a protein found on the zymogen membrane that is secreted into the pancreatic duct by exocytosis, is a reproducible integral component of protein plugs found in patients with noncalcific, chronic pancreatitis. On the other hand, lithostatin was found less frequently and in variable amounts. Interestingly, GP2 has significant homology with the urinary protein uromodulin (Tamm-Horsfall protein), which is involved in the formation of protein precipitates in urine. It has been suggested that GP2 may be involved in the formation of protein plugs in the pancreatic duct in patients with chronic pancreatitis.

In addition to hypersecretion of protein, viscosity of pancreatic juice is enhanced in chronic pancreatitis because of a higher concentration of hexosamine. [47] There is a correlation between the concentrations of protein, hydrolytic enzymes, hexosamine, and viscosity. [47] Hyperviscosity of pancreatic juice may result in decreased flow and contribute to protein precipitation in chronic pancreatitis. Therefore, it appears that chronic alcoholism results in a number of changes in the pancreatic juice that create a conducive environment for the formation of intraductal protein plugs that block small ductules in a random fashion. Ductal blockade produces progressive structural abnormalities in the ducts and acinar tissue. In time, calcium is complexed to protein plugs, initially in small ductules and, eventually, in the main pancreatic

duct. Progressive blockade results in injury and destruction of pancreatic tissue. [48]

The biochemical mechanisms responsible for fibrosis to occur in chronic pancreatitis are also unclear. Recent evidence suggests that pancreatic stellate cells are responsible for pancreatic fibrosis. [49], [50] Pancreatic stellate cells are perivascular [51] and derived from vitamin A–containing cells that can acquire a myofibroblast phenotype. [49], [52] These cells may be activated by transforming growth factor-ß $_1$ (TGB-ß $_1$) and basic fibroblast growth factor [49] during fibrosis in chronic pancreatitis. Transgenic mice that overexpress TGF-a develop interstitial pancreatic fibrosis. One study [53] demonstrated high levels of TGF-a and its messenger RNA in pancreatic tissue from patients with chronic pancreatitis, coupled with high levels of epidermal growth factor receptor, the receptor through which TGF-a acts. Another study [54] demonstrated overexpression of acidic and basic fibroblast growth factors in tissue from patients with chronic pancreatitis. These mitogenic peptides are involved in cellular proliferation and differentiation. Although these observations are intriguing, it remains to be determined whether these abnormalities are contributing to the progression of disease or are merely a result of the disease process.

Cigarette Smoking

Recent studies indicate that there may be a strong association between cigarette smoking and chronic pancreatitis. [55] Lin and associates [56] reported that the odd ratios (95% CI) for chronic pancreatitis for smokers compared with nonsmokers, after adjustment for body mass index, education level, and alcohol consumption, was 7.8 (2.2 to 27.3) for all smokers, and greater risk was observed for those who had smoked for 25 years or more. The risk for chronic pancreatitis significantly increased with increasing cumulative amount of smoking. Other studies showed that cigarette smoking increases the risk for pancreatic calcification of late-onset but not of early-onset idiopathic chronic pancreatitis. [57]

Based on these and other studies, the role of smoking in the causation of chronic pancreatitis in the absence of alcoholism is strongly suspected but not conclusively proved. However, it appears that smoking enhances the progression of chronic pancreatitis, especially of calcification. In hereditary pancreatitis, smoking increases the risk for pancreatic cancer and lowers the age at onset by about 20 years. [58] Hence, patients with chronic pancreatitis, regardless of the cause, should stop smoking.

Tropical (Nutritional) Pancreatitis

Tropical chronic pancreatitis, one of the major nutritional forms of chronic pancreatitis, is an important disease among juveniles and young adults in some Afro-Asian countries (Indonesia, southern India, and tropical Africa). [59], [60] The natural history of tropical chronic pancreatitis is succinctly summarized by Gee Varghese [60] as “recurrent abdominal pain in childhood, diabetes around puberty and death at the prime of life.” Abdominal pain characterizes the onset of tropical pancreatitis. The onset of diabetes typically occurs a few years after the onset of abdominal pain. Diabetes is characteristically brittle, with marked fluctuations of blood glucose. At this time, abdominal radiographs invariably show diffuse pancreatic calculi. Microscopically, dilation of the ducts, pancreatic lithiasis, chronic inflammatory cell infiltration, and atrophy of the pancreatic parenchyma are seen.

The etiology of tropical pancreatitis is not clearly understood, although the common denominator appears to be malnutrition in most cases. [61] Evidence suggests that other factors, such as toxic products in certain nutritional components (cassava), may be more important. [62] Cassava is consumed in large quantities by most poor people in many Afro-Asian countries. It contains 65 mg of toxic glycoside per 100 g. When glycosides react with gastric hydrochloric acid, hydrocyanic acid is liberated. The enzyme rhodanase acts on hydrocyanic acid, leading to thiocyanate production in the presence of adequate amounts of methionine and cysteine. Cyanogens impair a number of enzymes, including superoxide dismutase, an important scavenger of free radicals, which are proposed to cause cell injury. [63], [64] Associated nutritional deficiencies, such as deficiencies of zinc, copper, and selenium, which are common in malnutrition, interfere with detoxification of cyanogens. Thus, the pathogenesis of tropical pancreatitis may be partly explained by micronutrient-antioxidant deficiencies and unopposed free radical injury secondary to dietary cyanogens.

Hereditary Pancreatitis

The clinical picture of hereditary pancreatitis was well described in 1952 by Comfort and Steinberg. [65] Since then, many similar cases have been reported from different areas of the world (United States, Ireland, France, and New Zealand). It is an autosomal-dominant disorder, with 80% penetrance and variable expression, that begins in childhood. The incidence is about equal in both sexes. [66] Lebodic and colleagues [67] from France, and Whitcomb and associates [68] from the United States, concurrently mapped the gene to the long arm of chromosome 7 (7q55). Subsequently, by positional cloning, Whitcomb and colleagues [69], [70] identified two genetic mutations in the cationic trypsinogen gene in hereditary pancreatitis—a missense mutation R117H on exon 3 [69] and a mutation in the exon 2 of the gene (N21I) [70]—in kindred without the R117H mutation. It was hypothesized that cleavage at these sites of the trypsinogen molecule is part of a failsafe mechanism to inactivate trypsin within the pancreas. Gene mutations result in failure of cleavage and persistent tryptic activity, causing autodigestion of the pancreas and pancreatitis. These mutations have since been described in families with pancreatitis around the world. [71], [72], [73], [74] and [75] They represent the causative mutation in about two thirds of the families with classic hereditary pancreatitis and currently are the only mutations for which genetic testing is recommended.

In addition to the typical hereditary form described by Comfort and Steinberg, which begins at a young age, familial aggregations of two or three cases, with attacks beginning in the third or fourth decade, [76] have been reported. This suggests that there may be different forms of hereditary transmission. Mutations at codons 16, [77] 22, [78] and 23 [79] in the trypsinogen gene, although significantly associated with chronic pancreatitis, have a low penetrance. These mutations may be present in patients with a diagnosis of sporadic idiopathic pancreatitis. However, trypsinogen gene mutations are not the cause of tropical pancreatitis, even if many family members are affected. [80]

The clinical picture of hereditary pancreatitis differs little from that of nonhereditary pancreatitis [66] and carries a high risk for pancreatic cancer. [81] Patients frequently experience recurrent attacks of severe upper abdominal pain. Overt diabetes develops 8 to 10 years after the onset of pain in 20% of cases, and gross steatorrhea in 15% to 20%. The diagnosis of hereditary pancreatitis should be suspected if several family members have pancreatitis in the absence of alcohol consumption or other causes of chronic pancreatitis.

Hyperparathyroidism

Calcified chronic pancreatitis occurs in untreated hyperparathyroidism. Surveys suggest that the incidence is decreasing and is no greater than 1% to 2%. [82] A likely explanation is that serum calcium is measured routinely in virtually all patients who undergo a medical checkup, and, as a result, hyperparathyroidism does not remain undiscovered and untreated for many years. The pathogenesis of pancreatitis in hyperparathyroidism is presumed to be related to the effect of hypercalcemia. Acute hypercalcemia is a potent stimulus of human pancreatic enzyme secretion. [83], [84] Furthermore, chronic hypercalcemia causes a significant increase in pancreatic calcium secretion in patients with hyperparathyroidism and in experimental animals. This results in precipitation of intraductal calcium in pancreatic juices. Moreover, chronic hypercalcemia causes a decrease in the diffusion barrier between the pancreatic interstitial compartment and the ductular system, resulting in excessive diffusion of calcium into the pancreatic juices. These events may damage the pancreas and promote the development of calcified chronic pancreatitis.

Hyperlipidemia

As discussed in Chapter 94, it is well known that hyperlipidemia can cause acute pancreatitis. After repeated episodes of acute pancreatitis, hyperlipidemia may, on rare occasions, cause chronic pancreatitis. This unusual cause is important to consider because effective therapy is available. Glueck and associates [85] highlighted the risk for estrogen replacement in postmenopausal women with underlying hypertriglyceridemia. Twelve women receiving estrogen replacement were studied; 10 of these had fasting triglycerides greater than 1200 mg/dL while receiving estrogens, and 6 developed pancreatitis. Stopping estrogen replacement therapy and treatment of the underlying hypertriglyceridemia returned triglycerides to normal levels. Fasting triglyceride levels of less than 300 mg/dL pose no risk for estrogen-induced pancreatitis, whereas levels greater than 750 mg/dL are associated with a high risk. Therefore, it is important to measure fasting serum triglycerides before initiating estrogen replacement in postmenopausal women to prevent acute pancreatitis and avoid the potential for chronic pancreatitis.

Autoimmune Pancreatitis

Chronic pancreatitis may be caused by autoimmunity. [85], [86] In the Japanese literature, more than 100 cases of autoimmune pancreatitis have been reported. [86], [87] and [88] Several autoantibodies, such as antinuclear antibody, antilactoferrin antibody, anticarbonic anydrase II–antibody rheumatoid factor, and anti–smooth muscle antibody, are frequently present in patients with autoimmune pancreatitis. [89] This form of chronic pancreatitis is characterized by increased serum ß-globulin or immunoglobulin G levels, presence of diffuse enlargement of the pancreas, irregular narrowing of the main pancreatic duct, an extrinsic stenosis of the pancreatic portion of the common bile duct on endoscopic retrograde pancreatography, and cholestatic liver dysfunction. [87] Most patients have mild symptoms without attacks of acute pancreatitis. Obstructive jaundice caused by stenosis of the common bile duct is characteristic of autoimmune pancreatitis and is rare in other types of pancreatitis. [87] All these patients showed dramatic improvement with steroid treatment. Computed tomography (CT) and ultrasonography usually show a diffusely enlarged pancreas, with a sausage-like appearance and a capsule-like rim that is of low density on CT. Pancreatic calcification or pseudocyst is seldom observed. Endoscopic retrograde cholangiopancreatography (ERCP) usually shows segmental or diffuse narrowing of the main pancreatic duct and stenosis of the common bile duct, mainly in the intrapancreatic area, resulting in dilation of the proximal biliary tract. [87] Steroid therapy is usually effective in alleviating the stenosis of the common bile duct as well as that of the pancreatic duct. [90], [91] The differential diagnosis of diffuse pancreatic enlargement includes malignant lymphoma, plasmacytoma, metastasis and diffuse infiltrative pancreatic carcinoma. Definitive diagnosis can be made by histological findings that show fibrotic changes with infiltration of lymphocytes and plasma cells and sometimes, eosinophils around the pancreatic duct. The lymphocytes are usually $CD4^+$ and $HLA\text{-}DR^+$ T cells. [92], [93] and [94] HLA-DR is also expressed on pancreatic duct cells. [92], [95] Because pancreatic specimens are usually not available, the diagnosis of autoimmune pancreatitis is made from a combination of clinical and laboratory findings and pancreatic imaging studies.

Obstruction

Obstruction of the main pancreatic duct by tumors, benign vaterian stenosis, [96] scars (e.g., from traumatic pancreatitis), and pseudocysts [97] can lead to a distinct form of chronic pancreatitis known as *obstructive chronic pancreatitis*. This is characterized by acinar atrophy and fibrosis and dilation of the ductal system. In contrast to alcohol-induced chronic pancreatitis, intraductal plugs or stones are very rare in obstructive chronic pancreatitis, and both structural and functional changes may improve when obstruction is relieved. [96] The regression of fibrotic lesions may be explained by the finding that fibrosis in experimental obstructive pancreatitis consists of fractions of collagen with short half-lives (fibronectin, laminin, collagen III, and procollagen III) rather than the long-lived fraction collagen I. [98]

Trauma

Trauma to the abdomen or back may be clinically insignificant and still produce significant pancreatic injury, leading to chronic pancreatitis. The pathogenesis may follow that of the obstructive type, but inflammation and pseudocysts frequently develop. Recognition of trauma as the cause for chronic pancreatitis is important because most cases are associated with severe ductal disruption, which responds poorly to medical treatment, yet results of surgical correction (particularly partial pancreatectomy) have been excellent. [99], [100]

Pancreas Divisum

Pancreas divisum is the most common congenital abnormality of the pancreas (4% to 11%). [101], [102] Fusion of the dorsal and ventral pancreatic ducts is absent or incomplete, and drainage of the major portion of the pancreas occurs through the minor papilla. The clinical significance of pancreas divisum is unknown, although considerable controversy surrounds this issue. Several reports suggest that the incidence of pancreatitis is increased in subjects with pancreas divisum. Cotton reported that the incidence of pancreas divisum in patients with idiopathic pancreatitis undergoing ERCP was 25.6%, whereas only 3.6% of patients with biliary disease undergoing ERCP had pancreas divisum. [103] Similarly, Sahel and colleagues [104] noted that the incidence of pancreas divisum among patients with acute pancreatitis was 21%. This was much higher than a 5% incidence of pancreas divisum among all patients undergoing ERCP. Richter [105] also reported a 19% incidence of pancreas divisum in patients with idiopathic pancreatitis, compared with an overall 5% incidence of pancreas divisum in patients undergoing ERCP. These observations suggest that pancreas divisum might be an important cause of disease in patients with idiopathic pancreatitis. It has been hypothesized that the opening of the lesser papilla might be too small to permit free flow of pancreatic juice into the duodenum, and, as a result, pancreas divisum might cause a form of obstructive pancreatitis.

The clinical significance of pancreas divisum has been disputed. Delhaye and associates [106] reported the results of a study in Belgium involving 6324 patients undergoing ERCP for biliopancreatic complaints and noted that the incidence of pancreas divisum was similar in patients with chronic pancreatitis (6.4%), acute pancreatitis (7.5%), and nonpancreatic disease (5.5%). Similar results have been reported by Sugawa and colleagues, [107] who noted a 2.7% incidence of pancreas divisum among 1529 patients undergoing ERCP. There was no increase in incidence of pancreas divisum among patients with idiopathic pancreatitis or people with unexplained upper abdominal pain. Burtin and colleagues [108] reported a 5.9% prevalence of pancreatic divisum in more than 1000 patients undergoing ERCP. The proportion of patients with pancreas divisum was similar among patients with and without pancreatitis. Thus, the results reported by Burtin and colleagues, [108] Delhaye and associates, [106] and Sugawa and colleagues [107] contradict the earlier studies by Cotton, [103] Sahel and colleagues, [104] and Richter, [105] which indicated an increased incidence of pancreas divisum among patients with pancreatitis. It is possible that the results from earlier studies [103], [104] and [105] might be biased by patient selection. Patients with pancreas divisum and patients with idiopathic pancreatitis are likely to be referred to centers with exceptional endoscopic skills, and, as a result, the incidence of pancreas divisum may appear to be increased, whereas such an increase is not present in a more random sampling of patients.

Idiopathic Pancreatitis

The major form of nonalcoholic chronic pancreatitis in North America and Europe is the idiopathic type (10% to 40%). Epidemiology and clinical studies suggest that idiopathic chronic pancreatitis may be divided into two distinct groups: a juvenile type, with onset of symptomatic disease at a median age of 18 years; and a senile type, with an incidence peak at 60 years of age. In juvenile chronic pancreatitis, abdominal pain dominates the clinical picture at initial presentation, whereas in senile groups, painless disease is frequent, and most patients present with exocrine insufficiency, diabetes, and pancreatic calcification. [109]

Recent genetic studies indicate that cystic fibrosis transmembrane conductance regulator (*CFTR*) gene mutations are linked to some patients with chronic pancreatitis. Sharer and colleagues [110] searched for 22 *CFTR* mutations in 134 patients who had idiopathic chronic pancreatitis or chronic pancreatitis due to alcoholism, hyperparathyroidism, or hypertriglyceridemia. In these patients, *CFTR* mutations were seen at 2.5 times the expected frequency, and the frequency of the 5T allele (which is associated with a reduced proportion of functional *CFTR*) was twice the expected frequency. In a separate study, Cohn and associates [111] found that 10 of 27 patients with idiopathic chronic pancreatitis had at least one of 17 *CFTR* mutations; both alleles were affected in three patients. The 5T allele was present in 5 of 27 patients; this was twice the expected frequency. Hence, it is intriguing that at least a subset of patients who would be classified as having idiopathic chronic pancreatitis has *CFTR* mutations. The functional significance and clinical relevance of these mutations on the pancreas are unclear, but most patients in the above studies did not have classic cystic fibrosis. They all had normal sweat chloride test results and nasal potential difference. Cohn and associates, in a review, stated, “At present, the role of CFTR mutation testing in idiopathic chronic pancreatitis is uncertain because no guidelines exist for genetic counseling or altered clinical management of idiopathic chronic pancreatitis based on the results of such testing.” [112]

CLINICAL PRESENTATION

Pain

The presenting symptom of most patients with chronic pancreatitis is abdominal pain. The pain usually is epigastric, dull rather than sharp, and constant rather than colicky. Although the pain may radiate to both upper quadrants and, occasionally, to the lower quadrants, a characteristic feature is radiation directly through to the back. The pain is partially relieved by sitting with the trunk bent forward or lying prone; the supine posture aggravates the discomfort. The almost instantaneous aggravation of pain by food ingestion is characteristic of chronic pancreatitis or carcinoma of the pancreas, and this symptom always should raise the suspicion of pancreatic disease. Although food ingestion may aggravate the pain of other abdominal conditions (i.e., irritable bowel syndrome), there is usually a much greater interval between the meal and the discomfort than is the case with pancreatic disease. Ingestion of alcohol also may aggravate the pain, although a sizable fraction of patients claim that the pain develops after 12 to 24 hours of abstinence. [113]

Pain patterns may differ depending on the type of chronic pancreatitis. [23] At the beginning of disease, pain is present in 75% of patients with alcoholic chronic

pancreatitis, 50% of patients with late-onset idiopathic chronic pancreatitis, and nearly all patients with early-onset idiopathic chronic pancreatitis. Pain tends to be less severe in the late-onset idiopathic group. In a prospective longitudinal study of alcoholic chronic pancreatitis, Ammann and colleagues [114] identified two pain patterns: 44% of the patients had short episodes of pain, usually less than 10 days' duration, and were separated by pain-free intervals of months to years. None of these patients underwent surgery for pain relief. By contrast, 56% of patients had episodes of constant daily pain, occurring 2 or more days per week for at least 2 months. All these patients underwent surgery, and most had a complication such as pancreatic pseudocysts or cholestasis that was amenable to surgical correction. It is important to emphasize that the above study only evaluated alcoholic chronic pancreatitis patients not addicted to narcotics. Therefore, these findings are not applicable to patients who are addicted to narcotics and who have any form of chronic pancreatitis.

Pain in chronic pancreatitis may continue, diminish, or disappear completely. Several reports have suggested that pain may disappear as the severity of pancreatitis increases. [115], [116] and [117] According to Ammann, [117] pain disappears coincident with the appearance of calcifications, steatorrhea, and diabetes, which occurs 5 to 18 years after the onset of chronic pancreatitis. Chronic pancreatitis is painless or relatively painless (i.e., insufficient pain to require medical evaluation) in about 15% of patients. [118] Idiopathic pancreatitis is more likely to be painless than the alcoholic variety.

Weight Loss

Nausea, vomiting, anorexia, and weight loss are common in chronic pancreatitis. The major cause of weight loss is decreased caloric intake owing to fear of aggravation of the abdominal pain, although malabsorption or uncontrolled diabetes also may play a role. Marked weight loss is relatively unusual in other painful abdominal conditions, such as peptic ulcer or irritable bowel syndrome, and the combination of chronic upper abdominal pain and rapid loss of weight always should suggest the possibility of pancreatic disease.

Malabsorption

Diarrhea, steatorrhea, and azotorrhea occur when exocrine secretion of pancreatic enzymes is insufficient to maintain normal digestion. Although most attention has been directed to the malabsorption of fat and protein in this condition, studies using breath H_2 excretion also have demonstrated malabsorption of starch. [119]

Because malabsorption does not occur until enzyme secretion is reduced to less than 10% of normal, [120], [121] diarrhea and steatorrhea occur relatively late in the course of chronic pancreatitis. As a rule, fecal weight is less in pancreatic malabsorption than in other conditions with comparable steatorrhea. This relatively low fecal weight reflects a lesser quantity of fecal water, and patients may pass bulky, formed stool, as opposed to the frank watery diarrhea observed in other conditions. The relatively low fecal water in pancreatic insufficiency probably results from the better absorption of some carbohydrates, such as disaccharides, that occurs in conditions with mucosal pathology, such as celiac sprue. Occasionally, the patient observes that gross oil leaks from the rectum or floats as an "oil slick" on the surface of water in the toilet bowel. If ingestion of mineral oil is excluded, this finding usually is indicative of pancreatic steatorrhea. For a given degree of steatorrhea, the absorption of fat-soluble vitamins (A, D, E, and K) is much better in pancreatic insufficiency than in celiac sprue. [122] Marked deficiency of these vitamins seldom is observed in pancreatic insufficiency, and presumably this reflects the relative unimportance of lipolysis for the absorption of these compounds. The body stores of these vitamins, however, have been shown to be reduced in a large fraction of patients with chronic pancreatitis, although clinical manifestations of the deficiencies are quite rare. [123]

Pancreatic Diabetes

Although glucose intolerance is common early in the course of chronic pancreatitis, clinically evident diabetes occurs relatively late in the disease. Endocrine insufficiency eventually occurs in most patients with chronic pancreatitis. In a follow-up study of 500 patients with chronic pancreatitis, of whom 85% were alcoholic, it was reported that diabetes developed in 83% within 25 years after the clinical onset of chronic pancreatitis, and more than half required insulin treatment. [124] The prevalence of diabetes was similar in patients who underwent surgery and those who received medical treatment. However, for those who had pancreatic surgery, the prevalence of diabetes was significantly higher 5 years after distal pancreatectomy (57%) than after pancreaticoduodenectomy, pancreatic drainage, or cystic drainage (24% to 36%). Pancreatic drainage did not prevent the onset of diabetes. Distal pancreatectomy and early onset of pancreatic calcifications were the only independent risk factors for diabetes.

In most patients, the diagnosis of chronic pancreatitis is established long before the development of symptomatic hyperglycemia; however, the occasional patient with relatively painless pancreatitis may present initially with diabetes. Ketoacidosis and diabetic nephropathy are relatively uncommon in this form of diabetes. For a given duration of diabetes, however, retinopathy and neuropathy are thought to occur with a frequency similar to that in idiopathic diabetes mellitus. [125]

Other Clinical Features

Other clinical presentations of chronic pancreatitis include jaundice, secondary to common bile duct compression by the pancreas; ascites or pleural effusion, caused by leak of pancreatic secretions from a ruptured duct or pseudocyst; painful nodules, usually over the lower extremities, resulting from fat necrosis; and, rarely, a polyarthritis of the small joints of the hands. In the United States, at least 70% of chronic pancreatitis is attributed to prolonged, heavy ingestion of ethanol, whereas the bulk of the remaining 30% is idiopathic. Thus, historical information concerning length and duration of ethanol ingestion is critical in all suspected cases of chronic pancreatitis. In contrast, gallstone disease virtually never is the cause of chronic pancreatitis. A family history of pancreatitis or chronic abdominal pain of unknown origin may suggest the diagnosis of familial pancreatitis.

The physical examination usually is of limited assistance in the diagnosis of chronic pancreatitis. There is epigastric tenderness during the painful episodes (and sometimes during periods of remission). Tenderness ranges from mild to marked, but involuntary guarding and rebound are unusual. The physical signs characteristically are trivial relative to the intensity of the patient's complaints. Complications of chronic pancreatitis, such as pseudocysts, ascites, or pleural effusions, may be detected on physical examination.

PATHOMECHANISM OF SYMPTOMS

Abdominal pain and malabsorption are the major symptoms of chronic pancreatitis. The pathophysiologic processes responsible for the development of these symptoms are discussed in the following sections.

Pain

The mechanisms of pain in chronic pancreatitis are unclear. Possible causes include inflammation of the pancreas, increased intrapancreatic pressure, neural inflammation, and extrapancreatic causes, such as common bile duct stenosis and duodenal stenosis.

Acute inflammation of the pancreas, during a relapsing attack of chronic pancreatitis or peripancreatic inflammation involving the duodenum and retroperitoneum, may cause pain. This may be mediated by inflammation or fibrosis. Cytokines such as interleukin-6 (IL-6) and IL-8 [126] and chemokines such as MOB-1 and tumor necrosis factor-a may be involved. Whether fibrosis by itself produces pain is unclear, but in chronic pancreatitis the link between fibrosis and pain likely is related to pancreatic stellate cells, which are responsible for pancreatic fibrosis. [127] Because of their contractile potential and perivascular location, these cells could cause microvascular ischemia and pain [127] when stimulated by growth factors. [128]

Clinical and experimental evidence suggests that pain also may be related to increased intraductal pressure secondary to continued pancreatic secretion in the face of ductal obstruction caused by strictures or intraductal stones. [129] The increased pancreatic ductal and parenchymal pressure also may produce a compartment syndrome that induces ischemia and pain. This thesis is supported by experimental studies, which showed increased interstitial and perfusion pressures and decreased blood flow in feline chronic pancreatitis. [130], [131] These abnormalities were significantly reversed by surgical incision of the gland and draining the pancreatic duct. [131]

Several clinical studies showed that pain relief correlates with the development of pancreatic insufficiency in patients with alcohol-induced chronic pancreatitis. For example, Ammann and colleagues [115] noted that most patients with chronic calcific pancreatitis eventually became pain free, and the onset of relief was clearly

associated with decreased pancreatic secretion. A similar observation was made by Girdwood, [116] who reported that 31% of patients with painless pancreatitis had severe pancreatic insufficiency, compared with 3% who had painful pancreatitis. These observations suggest that decreased secretion may reduce intraductal pressure and relieve pain. Measurements of intraductal pressure have been made in patients without pancreatic disease and in those with ductal abnormalities. Among patients without pancreatic disease, intraductal pressure has been found to be 7 mm Hg by direct puncture at surgery in one patient and 10 to 16 mm Hg by ERCP among 33 patients without pancreatic disease. [132], [133], [134] and [135] By comparison, in 59 patients with a dilated duct, intraductal pressure measured by direct puncture at surgery ranged from 18 to 48 mm Hg. [135] In one study, [132] all 19 patients with a dilated main pancreatic duct with elevated intraductal pressure had prompt relief of pain after decompression surgery. Pancreatic tissue pressure also appears to be elevated among patients with chronic pancreatitis with a dilated pancreatic duct and those with pseudocyst. [129], [136] The pancreatic tissue pressure normalized after decompression of the duct or cyst, and this resulted in pain relief in 12 of the 14 patients. Hence, these observations support the thesis that pain in chronic pancreatitis is related to increased intraductal and pancreatic tissue pressure. This provides the rationale for treatments that may reduce pancreatic secretion, such as octreotide, specific diets, and pancreatic enzyme therapy.

Intrapancreatic neural inflammation is another factor that may play an important role in the genesis of pain in chronic pancreatitis. Morphologic studies indicate that pancreatic nerves appear to be larger and more numerous in chronic pancreatitis. [137] The organization of intraneural organelles, such as microtubules, is disrupted. Most significantly, however, there is an alteration in the perineurial sheath that ordinarily shields nerves from surrounding connective tissue. [137] The damaged perineurium allows penetration of biologically active materials from the surrounding extracellular matrix, and pain may result from the continual stimulation of the sensory fibers by noxious substances. In this regard, immunohistological studies have shown that the amount of neurotransmitters, such as substance P and calcitonin gene-related peptide, are increased in afferent pancreatic nerves in chronic pancreatitis. [138], [139] Immune cell infiltration and growth-associated protein 43 also correlate with pain intensity. [140] There is also evidence that eosinophils are increased in the perineurial space among patients with recent alcohol consumption. [141] It is conceivable that degranulation of these eosinophils might be a factor in the generation of pain.

Peripancreatic inflammation, involving the duodenum and retroperitoneum, may cause pain. Extension of active inflammation of the pancreatic tissue within the wall of the duodenum may result in extensive fibrosis and stenosis of the descending duodenum. [142] Peripancreatic inflammation may cause stenosis of the distal common bile duct, and this has been noted to be associated with severe abdominal pain. Pain may also result from continued inflammation of the head of the pancreas. [142] Thus, it is likely that pain in chronic pancreatitis is multifactorial. Depending on the etiologic factors, structure, alteration, severity of the disease, and degree of pancreatic insufficiency, different factors may predominate in the genesis of pain. A study correlating pancreatic pathological condition and symptoms found that intermittent attacks of pain resulted from recurrent tissue necrosis, whereas chronic pain appeared to be secondary to segmental distention behind obstructed ducts. [143]

Malabsorption

There is a 10-fold reserve for exocrine pancreatic enzyme secretion. Malabsorption occurs only after the capacity for enzyme secretion is reduced by more than 90% [120] (Fig. 95-1). In chronic pancreatitis secondary to alcoholism, it usually takes 10 to 20 years for severe pancreatic insufficiency to develop, but lipase secretion decreases more rapidly than secretion of proteolytic enzymes [121] (Fig. 95-2). Hence, steatorrhea is often an earlier and more severe problem than azotorrhea. The importance of colipase in fat malabsorption in adult patients is unknown, although colipase appears to be an important factor in children because steatorrhea and colipase correlate better than lipase and steatorrhea. [144]

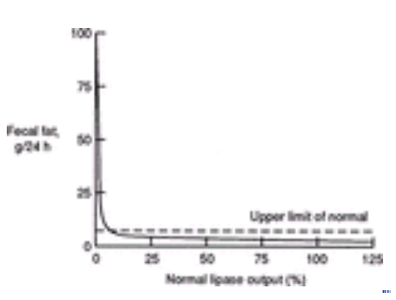

FIGURE 95-1. Relation between steatorrhea and lipase output. Steatorrhea does not occur until lipase output is reduced below 10% of normal. (Adapted from ref. [120].)

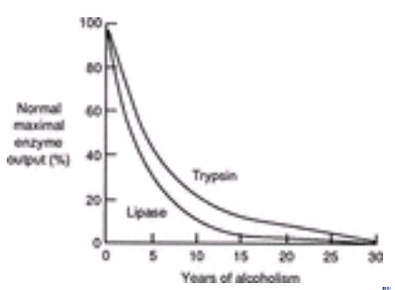

FIGURE 95-2. Relation between reductions in enzyme output and years of alcoholism in patients with alcoholic pancreatitis. (Adapted from ref. [121].)

Concurrent with the reduction of pancreatic enzyme secretion, there is also decreased bicarbonate secretion in patients with severe chronic pancreatitis. Not infrequently, the duodenal pH may fall to 4.0 or less [145], [146] 90 minutes after eating. The abnormally low duodenal pH (less than 4.0) reduces lipid digestion by inactivating pancreatic enzymes and precipitating bile acids. These factors are important to consider in the treatment of pancreatic steatorrhea.

DIAGNOSIS

Overview of Diagnostic Approaches

The diagnosis of chronic pancreatitis usually is suggested by historical information and then confirmed by radiography or laboratory tests. Over the years, an enormous number and variety of tests have been developed for the diagnosis of pancreatitis (Table 95-2). Many of these tests have not been adequately evaluated, and their true sensitivity and specificity are unknown. This section reviews many of the proposed tests for chronic pancreatitis; most are of limited clinical value. An understanding of the rationale underlying the test, however, often provides an insight into the physiologic alterations that occur in chronic pancreatitis. The diagnosis of chronic pancreatitis often can be made on the basis of history and relatively simple radiographic tests. Only the rare patient requires extensive and complicated testing. Thus, the workup should be tailored to the individual patient.

I. Measurement of pancreatic products in blood
A. Enzymes
B. Pancreatic polypeptide
II. Measurement of pancreatic exocrine secretions
A. Direct measurement
1. Enzymes
2. Bicarbonates
B. Indirect measurement
1. Bentiromide test
2. Dual Schilling test
3. Fecal chymotrypsin concentration
4. [^{14}C] olein absorption
III. Imaging techniques
A. Plain film radiography of abdomen
B. Ultrasonography
C. Computed tomography
D. Endoscopic retrograde cholangiopancreatography
E. Magnetic resonance cholangiopancreatography
F. Endoscopic ultrasonography

TABLE 95-2 Tests for Chronic Pancreatitis

Laboratory Evaluation

Routine Laboratory Findings Routine blood studies usually are not helpful in making the diagnosis of chronic pancreatitis. Leukocytosis may be observed during acute exacerbations. The anemia frequently associated with malabsorption secondary to celiac disease seldom is present in patients with pancreatic malabsorption. Although patients with chronic pancreatitis commonly have an abnormal Schilling test result owing to the inability to digest R protein normally, megaloblastic anemia is rare. [147] Fat-soluble vitamin deficiency states (hypocalcemia, hypoprothrombinemia) seldom are observed in association with the steatorrhea of chronic pancreatitis, in contrast to their frequent occurrence in celiac sprue. Compression of the common duct by a fibrotic process in the pancreas not infrequently leads to varying degrees of cholestasis. [148] Alkaline phosphatase elevation in the absence of jaundice or other symptoms is the most common manifestation. Clinical jaundice results from more severe compression. Although liver disease is extremely common in alcoholics, this condition usually does not take the form of relatively pure cholestasis. The finding of cholestatic liver function in patients with pancreatitis requires extensive radiologic testing, usually culminating in ERCP, to establish that the biliary obstruction results from chronic pancreatitis.

Specific Tests for the Diagnosis of Chronic Pancreatitis

The multiple tests available for the diagnosis of chronic pancreatitis can be separated into chemical measurements of pancreatic function and radiologic procedures that provide information on pancreatic structure.

Several factors make it difficult to determine accurately the true sensitivity and specificity of these tests. First, biopsies of the pancreas usually are not obtained in patients with possible chronic pancreatitis, and most series report that patients have not been followed for a period sufficient to establish the presence or absence of this condition with certainty. Thus, in most early or questionable cases, there is no histological gold standard that can be used to determine the accuracy of the more indirect chemical or radiologic tests. Second, the sensitivity of the various diagnostic tests depends on the extent of the pancreatic damage. In long-standing disease with marked functional and anatomic alterations, virtually all proposed tests are positive. The sensitivity of the tests falls dramatically in early or mild chronic pancreatitis, however. The apparent sensitivity and specificity of the various tests also depend on the composition of subjects used to establish the normal range. For example, patients with a variety of disease states such as diabetes mellitus, [149] celiac sprue, [150] and cirrhosis [151] may have subnormal pancreatic secretion of enzymes. If such patients are used to establish the normal range, the lower limits of normal are reduced, and the sensitivity of the test for chronic pancreatitis drops accordingly. If the normal range is established with healthy controls, the specificity of a low secretion is reduced. Thus, although claims are made with regard to the sensitivity and specificity of tests for chronic pancreatitis, these claims have to be interpreted in light of the types of patients composing the pancreatitis groups as well as the subjects used to establish the normal range.

Serum Pancreatic Enzymes In contrast to attacks of acute pancreatitis, in which the serum level of pancreatic enzymes is almost always elevated, serum enzyme levels may be elevated, normal, or low in chronic pancreatitis. In an acute exacerbation of the chronic process, the levels of these enzymes (amylase, lipase, trypsin) may be elevated; however, their serum concentrations seldom reach the levels observed in acute pancreatitis. A very high serum concentration of pancreatic enzymes during a painful episode in a patient with very advanced chronic pancreatitis should raise the possibility that some other abdominal process, such as gut perforation or infarction, is responsible for the pain.

Tests of Pancreatic Exocrine Function Tests of pancreatic exocrine function can be divided into those that directly assess pancreatic exocrine function (e.g., measurements of enzyme or bicarbonate secretion) and those that test some secondary effect of impaired enzyme secretion (e.g., malabsorption of a compound that requires pancreatic digestion for normal absorption).

Direct tests of pancreatic exocrine secretion. Because the basal secretory rate of the pancreas is highly variable, meaningful direct measurements of pancreatic secretion must be carried out in the stimulated state. Varying doses of secretin or CCK have been used for this purpose as well as a standard meal (the Lundh test). Early studies used simple drainage of the intestine at the ligament of Treitz to obtain pancreatic secretions. Because this technique does not recover all lumenal fluid passing the collection site, studies have used a constant perfusion of a nonabsorbable marker, such as polyethylene glycol, into the duodenal bulb. Measurements of the concentration of this marker passing the distal collection site permit assessment of the rate at which fluid passes this site and make it possible to quantitate the rate at which pancreatic secretory components pass this site. [120] A wide variety of pancreatic juice components, including the concentration or output of bicarbonate, amylase, lipase, and trypsin, have been assessed to find the most sensitive and specific indicator of chronic pancreatitis. Although claims have been made for the superiority of one of these measurements over another, comparative studies suggest that all of them tend to be equally depressed in chronic pancreatitis and that there is no clear-cut advantage of one measurement over another. Similarly, none of these measurements appears to be able to differentiate chronic pancreatitis from carcinoma of the pancreas. Collection of pure pancreatic juice by way of cannulation of the ampulla of Vater during ERCP apparently adds little to the accuracy of the test. [152] Stimulation of pancreatic secretion has been produced over a wide range of doses and dosing schedules using secretin, [153] CCK, [154] cerulein, [155] and bombesin, [156] singly or in combination. Although it is difficult to compare the results of different studies, there appears to be no clear-cut advantage of one stimulatory technique over another. The relative value of direct stimulation of the pancreas with secretin-CCK versus indirect stimulation by Lundh meal also is somewhat controversial, although most comparative studies suggest that direct stimulation is superior. [157], [158] For many years, the finding of a subnormal pancreatic secretion of enzymes or bicarbonate in aspirated duodenal contents was considered the most sensitive test for chronic pancreatitis, and, if pancreatic carcinoma could be excluded, a low secretion was assumed to be diagnostic of chronic pancreatitis. Recently, natural porcine secretin has been unavailable because of the unproven use of the agent for the treatment of autism. Synthetic porcine secretin has been produced and compared favorably to the natural porcine secretin in the diagnosis of chronic pancreatitis. [159] The sensitivity of the CCK or secretin pancreatic function tests was examined in 48 patients with suspected chronic pancreatitis. [160] All patients were evaluated with ultrasound, CT, and endoscopic retrograde pancreatography (ERP). All 10 patients with normal ERP results demonstrated normal function by direct hormonal stimulatory testing, and all 16 patients with advanced changes on ERP had grossly abnormal function testing. In 12 patients with mild to moderate changes on ERP, all demonstrated some abnormality of function, and of 10 patients with minimal or equivocal changes on ERP, all had some degree of functional impairment. CT and ultrasound were accurate in advanced chronic pancreatitis (94% to 100%) but performed poorly in patients with less advanced changes on ERP (40% to 50% accurate in patients with equivocal changes). This study is in agreement with other previous reports, which indicated direct hormonal stimulation testing may be the most sensitive test (70% to 95%) [161], [162], [163], [164] and [165] to diagnose chronic pancreatitis in all stages. However, it should be noted that patients with a variety of conditions, including diabetes mellitus, [149] hepatic cirrhosis, [151] Billroth II gastrectomy, [166] and celiac sprue, [150] may have diminished pancreatic secretory output with no clinical or radiologic evidence of chronic pancreatitis. Another major consideration is that direct hormonal stimulation testing is only available at a few centers; therefore, the search continues for a convenient and readily available diagnostic test.

Indirect tests of pancreatic exocrine secretion. The complexity and patient discomfort involved in the direct measurement of pancreatic secretory capacity have led to a variety of simpler tests that indirectly assess the secretion of pancreatic enzymes. Most of these tests measure the absorption of some compound that first requires digestion by pancreatic enzymes. Because clinically detectable malabsorption of nutrients does not occur until pancreatic enzyme secretion has diminished to less than 10% of normal, it is axiomatic that indirect tests of pancreatic function will not be able to detect early chronic pancreatitis and, thus, will have very poor sensitivity. One of the more commonly used of the indirect tests is the bentiromide test, which involves ingestion of *N*-benzoyl-L-tyrosyl- *p*-aminobenzoic acid (NBT-PABA), a tripeptide that is digested by chymotrypsin with the release of paraaminobenzoic acid (PABA). [167], [168] Free PABA is absorbed in the small bowel and excreted by the kidney; the quantity excreted in urine is used as a measure of pancreatic exocrine function. As appears to be the case with virtually every pancreatic function study, innumerable variations of the basic test (i.e., dosage alterations, length of urine collection, various forms of stimulation of pancreatic secretion) have been evaluated in an attempt to improve specificity, without appreciable success. The sensitivity of the bentiromide test varies with the extent of damage to the exocrine pancreas, with sensitivity as high as 100% [169] reported with very severe disease and as low as 40% to 50% with minor damage. [170], [171] The specificity of this test is far from perfect. Reduced PABA excretion has been reported to occur commonly in diabetes mellitus, [172] renal insufficiency, [173] liver diseases, [173] and malabsorption states other than pancreatic insufficiency, including celiac sprue, [174] Crohn's disease, [175] and postgastrectomy states. [173] The low excretion in malabsorptive states other than pancreatic insufficiency apparently results from failure to absorb the free PABA that has been split from the parent compound. These conditions can be separated from pancreatic insufficiency by testing the patient's ability to absorb free PABA in a subsequent test [169] or by simultaneously

administering free [^{14}C]PABA along with NBT-PABA and measuring the urinary excretion of the labeled and unlabeled PABA. [176] Because sensitivity of the NBT-PABA test is limited, the results of this measurement are helpful primarily when the test is positive (and other forms of malabsorption have been excluded). Thus, it appears that the major role of this test is not in diagnosis of chronic pancreatitis but rather in determining the extent of the pancreatic insufficiency present in patients with known chronic pancreatitis. The demonstration that vitamin B_{12} is malabsorbed in patients with pancreatic insufficiency, owing to their inability to degrade R protein, [177] provided the rationale for a test for chronic pancreatitis. In this test, the subject ingests both [^{57}Co]cobalamin bound to intrinsic factor and [^{58}Co]cobalamin bound to R protein, plus excess intrinsic factor and a cobalamin analog that saturates all empty R protein–binding sites. [178] Theoretically, the patient with pancreatic insufficiency should absorb the cobalamin bound to intrinsic factor much more efficiently than that bound to R protein. All other subjects should absorb both forms of cobalamin equally. Although the initial paper describing this technique reported excellent sensitivity and specificity for chronic pancreatitis, a subsequent study found very poor sensitivity. [179] The diagnosis of pancreatic insufficiency by way of measurements of fecal chymotrypsin was first described 40 years ago, [180] and a number of subsequent reports have evaluated the sensitivity and specificity of various modifications of the initial technique. The major advantage of this method is that it is rapid and simple. The far more cumbersome measurement of total fecal chymotrypsin output in timed fecal collections appears to offer little advantage over the much simpler measurement of chymotrypsin concentration in a random fecal sample. [180] Reports of sensitivity of the test range from 45% [181] to 100%, [182] depending on the extent of pancreatic damage in the study population. Specificity ranges from 49% [183] to 90%, [184] with false-positive results reported for malabsorptive conditions such as celiac sprue, damage secondary to Crohn's disease, and postgastrectomy states. [184], [185] Patients with appreciable chronic pancreatitis malabsorb fat when ingested in the form of triglyceride, and a number of indirect tests have been developed to assess such malabsorption. Most of these tests involve the feeding of [^{14}C]olein. Hydrolysis of the triglyceride and absorption of [^{14}C]oleate lead to the production and pulmonary excretion of $^{14}CO_2$. Measurement of breath $^{14}CO_2$ excretion can be used as a simple measure of the normality of the hydrolysis of the labeled triglyceride. [186] Unfortunately, the test is negative in a high percentage of patients with early chronic pancreatitis, and false-positive results are observed in many malabsorption states other than chronic pancreatitis. [187]

Imaging Studies

The demonstration of diffuse, speckled calcification of the pancreas on a plain film of the abdomen is diagnostic of chronic pancreatitis. Although the sensitivity of this finding is limited (perhaps, 30% to 40%), a plain film of the abdomen should be the first diagnostic test used when attempting to establish the diagnosis of chronic pancreatitis because a positive finding obviates the need for additional testing.

The development of ultrasound, CT, and ERCP has made it possible to assess, routinely, the gross structure of the pancreas. These tests all have excellent specificity and reasonably good sensitivity. Of major importance is the ability of these imaging procedures to differentiate between carcinoma of the pancreas and chronic pancreatitis. As a result, imaging techniques have largely supplanted tests of pancreatic function in the workup of the patient with possible chronic pancreatitis.

Ultrasound is the simplest and least expensive of the three imaging techniques. Findings on ultrasound that correlate with marked pancreatic changes on ERCP include dilation of the main pancreatic duct to greater than 4 mm, large (more than 1 cm) cavities, and calcifications. [188] Findings associated with less severe changes on ERCP include dilation of the duct up to but not exceeding 4 mm, small cavities, reduction in echogenicity or echogenic foci in the parenchyma, and an irregular contour to the gland. [188] When a satisfactory ultrasound examination is obtained, the reported sensitivity of this test for chronic pancreatitis is of the order of 70%, and the specificity is quite high, roughly 90%. [189], [190], [191] and [192] The finding of chronic pancreatitis on ultrasound usually requires no additional confirmatory testing.

CT is 10% to 20% more sensitive than ultrasound for the diagnosis of chronic pancreatitis [191], [192] and [193]; these two techniques have roughly comparable specificity. CT is more expensive than ultrasonography and involves exposure to ionizing radiation. Therefore, in the workup for chronic pancreatitis, CT usually should be limited to patients who have negative or unsatisfactory ultrasound examinations. The most common diagnostic findings of chronic pancreatitis on CT include duct dilation, calcifications, and cystic lesions [194], [195] and [196] (Fig. 95-3). Less common diagnostic findings include enlargement or atrophy of the pancreas and heterogeneous density of the parenchyma. This imaging technique is particularly helpful in the differentiation of chronic pancreatitis from carcinoma of the pancreas. The dilated duct system resulting from carcinoma tends to be smooth or beaded, in contrast to the irregular, calcified ducts that are typical of chronic pancreatitis. The sensitivity of CT for the diagnosis of chronic pancreatitis is in the neighborhood of 80%, whereas the specificity is roughly 90%. [191], [197], [198] ERCP commonly is considered to be the most sensitive and specific test available for the diagnosis of chronic pancreatitis, and this technique has become the gold standard against which all other tests are evaluated. In the absence of histological confirmation of disease in most patients, it is difficult to speak with complete certainty about the diagnostic accuracy of ERCP (because ERCP is the gold standard). Most studies indicate that the sensitivity and specificity of ERCP are 70% to 90% and 80% to 100%, respectively. [198], [199], [200], [201] and [202]

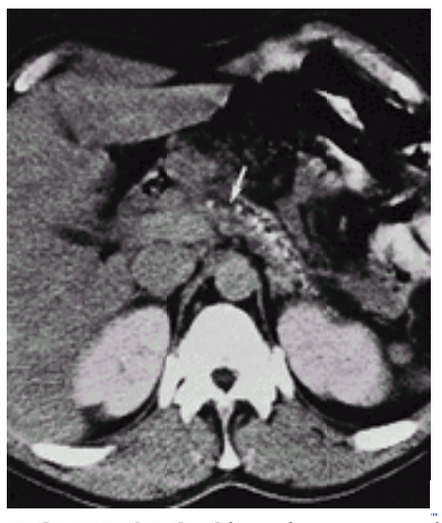

FIGURE 95-3. Chronic pancreatitis. Computed tomography scan shows pancreatic atrophy along with multiple intraductal calculi and dilation of the pancreatic duct (*arrow*).

The most commonly used classification of ERCP changes in chronic pancreatitis was developed at an international symposium. The Cambridge classification of chronic pancreatitis is based on abnormalities seen in the main pancreatic duct and in the side branches [203], [204] (Table 95-3). In minimal pancreatitis, the changes are limited to the branches and fine ducts (less than three), which show dilation and irregularity (Fig. 95-4). There is considerable observer variation in the detection of these minor changes, and differentiation of normal from abnormal is not always clear. Because minor ductal dilation and intraductal calculi may be observed in healthy elderly people, it may be difficult to differentiate early chronic pancreatitis from normal senile changes. [205] Moderate pancreatitis is characterized by abnormalities in at least three or more side branches and the additional finding of dilation, tortuosity, and stenosis of the main pancreatic duct (Fig. 95-5). Advanced pancreatitis has the additional findings of cyst formation, duct obstruction, severe ductal dilation, or irregularity (Fig. 95-6). Pancreatic carcinoma usually can be distinguished from chronic pancreatitis in that there is a single strictured region in malignant obstruction, in contrast to the multiple stenoses, irregular branching ducts, and intraductular calculi observed in chronic pancreatitis.

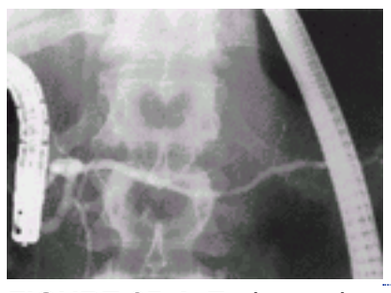

FIGURE 95-4. Endoscopic retrograde cholangiopancreatography (ERCP) study shows minimal changes consistent with chronic pancreatitis. The main duct is slightly irregular, and the branches are slightly dilated. These findings are not diagnostic.

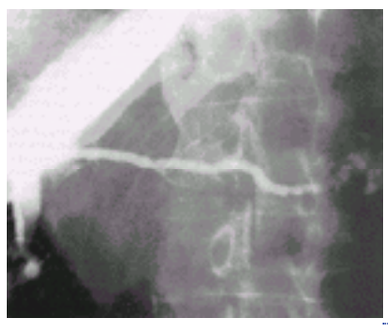

FIGURE 95-5. Endoscopic retrograde cholangiopancreatography (ERCP) study shows changes of moderate pancreatitis. The main duct is irregular and dilated, and there is a pseudocyst in the tail of the pancreas.

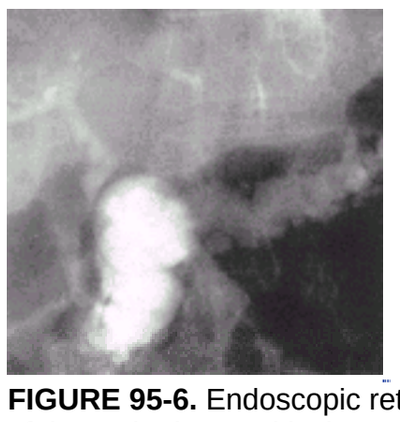

FIGURE 95-6. Endoscopic retrograde cholangiopancreatography (ERCP) study shows changes of advanced chronic pancreatitis. There are massive cystic dilations of the main duct and its branches.

GRADE	MAIN PANCREATIC DUCT	SIDE BRANCHES
Normal	Normal	Normal
Suggestive	Normal	<3 Abnormal
Mild	Normal	≥3 Abnormal
Moderate	Abnormal	>3 Abnormal
Severe	Abnormal plus at least one of the following: Large cavity Duct obstruction Dilation or irregularity Intraductal filling defects	

Modified from ref. 203.

TABLE 95-3 Cambridge Classification of Chronic Pancreatitis by Endoscopic Retrograde Pancreatography

In general, there is a good correlation between the changes observed on ERCP and measurements of pancreatic secretory capacity. An occasional patient, however, may have markedly diminished secretory function with seemingly minor changes on ERCP. Patients with minor abnormalities on ERCP often have normal secretory function.

It should be noted that pancreatitis occurs in about 5% of patients undergoing ERCP. [206] This risk for pancreatitis increases significantly in female patients with normal serum bilirubin, patients suspected of sphincter of Oddi dysfunction, and those with a previous history of post-ERCP pancreatitis. [206] In fact, patients with the least probability of harboring truly obstructive pathology are at highest risk for pancreatitis after ERCP, even if the procedure is for diagnosis alone. Because ERCP is an expensive procedure and has a low but not insignificant rate of associated complications, this test ordinarily should be reserved for the rare patient with chronic pancreatitis in whom the diagnosis cannot be clearly established by way of other imaging techniques.

Although chronic pancreatitis and carcinoma of the pancreas usually can be reliably distinguished based on imaging studies, in an occasional patient, this differentiation may be difficult. [207] Attempts to use serum or pancreatic juice tumor markers, such as CA 19-9, to differentiate benign from malignant pancreatic processes have not been impressive, [207] and it is usually necessary to make a tissue diagnosis. Although laparotomy with open biopsy was required in the past, percutaneous fine-needle aspiration biopsy now makes it possible to obtain a histological diagnosis in a very high percentage of patients with pancreatic carcinoma. Under CT or ultrasonographic guidance, a small-gauge needle is inserted into the pancreatic mass, and aspirated material is examined for the presence of malignant cells. The reported specificity of this test approaches 100%, and the sensitivity is 80% to 90%. [208]

In recent years, advances in magnetic resonance (MR) imaging have led to the development of MR cholangiopancreatography (MRCP) for the evaluation of the biliary and pancreatic ducts (Fig. 95-7). Similar to ERCP, MRCP allows direct visualization of the pancreatic duct but without radiation exposure or the use of oral or intravenous contrast. Hence, it can be used in patients who are pregnant or have contrast allergies. In severe chronic pancreatitis with marked changes of the main pancreatic duct, agreement between MRCP and ERCP is excellent. [209], [210], [211] and [212] However, MRCP is relatively insensitive in mild chronic pancreatitis, in which changes are limited to the secondary ducts. False-negative results may also be seen in patients with distal focal pancreatitis involving the tail of the pancreas. [209] Recently, secretin-enhanced MRCP has been proposed as a reliable test for assessment of pancreatic exocrine function. [213] The pancreatic T2 signal intensity and the diameter of the duodenum showed a significant increase after secretin administration in both healthy subjects and patients with chronic pancreatitis, but the increases were much smaller in patients with varying degrees of pancreatic insufficiency. [213] However, the validity of these findings requires further evaluation. It appears that the current generation of MR imaging is quite sensitive in the diagnosis of severe chronic pancreatitis, but it is rather ineffective in detecting mild to moderate pancreatitis, in which major ductal abnormalities are seen in the secondary branches. Currently, MRCP is used mainly as a backup when ERCP fails.

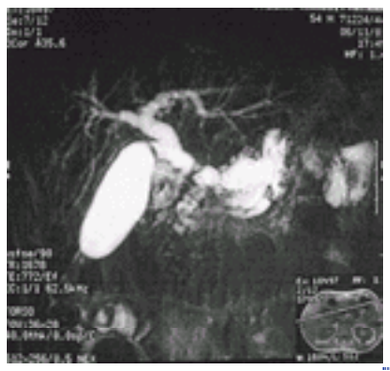

FIGURE 95-7. Magnetic resonance cholangiopancreatography shows calcification in the head of the pancreas and obstructions to both the pancreatic duct and common bile duct. (Courtesy of Dr. Theesa Rangsitwatana.)

Endoscopic ultrasonography (EUS) [214] is a major advance in imaging the pancreas because it may provide more detailed structural information of the pancreas than routine ultrasonography and CT scan. EUS features of chronic pancreatitis may be used to establish the diagnosis and evaluate the severity of the disease. [215] These include ductal changes and parenchymal changes, such as echo texture of the gland, calcifications, lobulations, and bands of fibrosis (Fig. 95-8 and Fig. 95-9; see Table 95-4). For EUS to be considered an accurate test for chronic pancreatitis, there must be good interobserver agreement. To address this issue, EUS was performed on 33 patients with suspected chronic pancreatitis based on typical symptoms as well as on 12 control patients. [216] Eleven experienced endosonographers blinded to clinical information independently evaluated all videotaped examinations for the presence of chronic pancreatitis based on nine validated features of chronic pancreatitis: echogenic foci, strands, lobularity, cysts, stones, duct dilation, duct irregularity, hyperechoic duct margins, and visible side branches. There was moderately good overall agreement for the final diagnosis of chronic pancreatitis (? = 0.45). Agreement was good for individual features of duct dilation (? = 0.6) and lobularity (? = 0.51) but poor for the other seven features (? < 0.4). The presence of stones was considered the most predictive feature of chronic pancreatitis, followed by visible side branches, cysts, lobularity, irregular main pancreatic duct, hyperechoic foci, hyperechoic strands, main pancreatic duct dilation, and main duct hyperechoic margins. Hence, EUS appears to be a reliable method for the diagnosis of chronic pancreatitis, with good interobserver agreement among experienced endosonographers. A prospective evaluation of EUS compared with ERP and secretin stimulation test in the diagnosis of chronic pancreatitis was conducted by Catalano and colleagues. [215] Eighty consecutive patients with recurrent pancreatitis participated in the study. EUS criteria for chronic pancreatitis included mild (one to two features), moderate (three to five features), and severe (more than five features; see Table 95-3). Abnormal EUS, ERP, and secretin test were observed in 63, 36, and 25 patients, respectively. Secretin test had 100% agreement with normal and severe chronic pancreatitis by EUS criteria, but agreement was poor for mild (13%) and moderate (50%) disease. On the other hand, the agreement between ERCP- and EUS-specific criteria was excellent for normal (100%), moderate (92%), and severe (100%) chronic pancreatitis and poor for mild (17%) disease. The main source of disagreement in this and other studies involves patients with abnormal results of EUS and normal ERCP or pancreatic function tests. Therefore, it appears that with the exception of mild changes noted on EUS, there is excellent agreement between EUS and ERCP in the diagnosis of chronic pancreatitis. Similar findings were reported in another prospective study to determine the ability of EUS to diagnose, exclude, or establish the severity of chronic pancreatitis found by ERP. [217] In general, histology is the most obvious candidate for a diagnostic gold standard, but very few studies have compared EUS with pancreatic histology. In a preliminary study comparing EUS with the histological features of chronic pancreatitis in 34 patients who underwent EUS and later pancreatic surgery (21 with chronic pancreatitis and 13 with pancreatic carcinoma), the sensitivity of EUS for diagnosis of chronic pancreatitis was 78%, specificity was 73%, and the threshold for diagnosis was four or more echo features. [218] However, these findings are only applicable to mostly severe pancreatitis. The specificity of EUS changes in mild or early chronic pancreatitis is unknown but probably very low. Reliance on a few EUS criteria, such as inhomogeneity, hyperechoic strands, and locularity, probably would give rise to many false diagnoses of chronic pancreatitis. Long-term follow-up of patients with mild EUS changes is needed to determine the usefulness of EUS in diagnosing mild chronic pancreatitis. In a recent study, 130 patients with a history of chronic use of alcohol and recurrent abdominal pain underwent ERCP and EUS. [219] All patients with chronic pancreatitis confirmed by ERCP (70.8%) had ductal or parenchymal changes detected with EUS. Among 38 patients (29.2%) with normal ERCP, 32 (84.2%) presented with morphologic features consistent with chronic pancreatitis by EUS. During follow-up (median, 18 months), chronic pancreatitis was confirmed by repeat ERCP in 22 of these 32 patients (68.8%). Hence, it appears that EUS is more sensitive than ERCP in the detection of early chronic pancreatitis in patients with abdominal pain and a history of chronic and continued ingestion of alcohol. More long-term follow-up studies are needed to determine the usefulness of EUS in diagnosing mild nonalcoholic chronic pancreatitis.

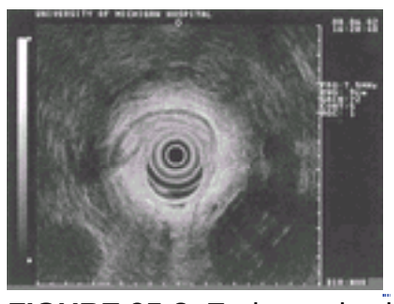

FIGURE 95-8. Endoscopic ultrasound showing mild chronic pancreatitis. Characteristic features include hyperechoic pancreatic duct margins and irregular contour of the pancreatic duct (*arrow*).

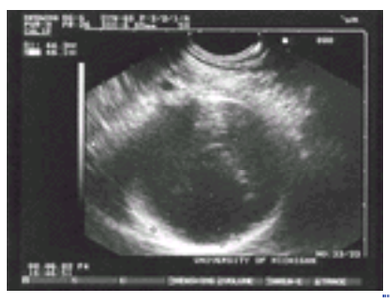

FIGURE 95-9. Endoscopic ultrasound showing a large pseudocyst (4.4 × 4.6 cm) with a calcified rim from a patient with chronic pancreatic pancreatitis.

ENDOSCOPIC ULTRASOUND FEATURE	IMPLICATION
Ductal changes	
Duct size >3 mm	Ductal dilation
Tortuous pancreatic duct	Ductal irregularity
Intraductal echogenic foci	Stones or calcification
Echogenic duct wall	Ductal fibrosis
Side-branch ectasia	Periductal fibrosis
Parenchymal changes	
Inhomogeneous echo pattern	Edema
Reduced echogenic foci (1–3 mm)	Edema
Enhanced echogenic foci	Calcifications
Prominent interlobular septae	Fibrosis
Lobular outer gland margin	Fibrosis, glandular atrophy
Large, echo-poor cavities (>5 mm)	Pseudocyst

Adapted from ref. 215.

TABLE 95-4 Endoscopic Ultrasound Features of Chronic Pancreatitis

Rational Use of Tests in the Diagnosis of Chronic Pancreatitis

The evaluation should begin with a plain film of the abdomen. The finding of pancreatic calcification is virtually diagnostic of chronic pancreatitis, and no further testing is required. If calcifications are not present, the next test should be ultrasound. Once again, if this test demonstrates findings indicative of chronic pancreatitis, further procedures are not necessary. If the ultrasound examination is nondiagnostic, CT is indicated only if there is a high degree of clinical suspicion of chronic pancreatitis. Only a small percentage of patients with chronic pancreatitis have negative ultrasound and CT findings. Thus, ERCP should be reserved for those patients in whom the diagnosis of chronic pancreatitis is thought to be very likely on clinical grounds or for patients in whom ultrasound, CT, and fine-needle biopsy have not been able to differentiate between carcinoma of the pancreas and chronic pancreatitis. If ultrasound, CT scan, and ERCP are normal, a secretin test may disclose evidence of mild chronic pancreatitis. Unfortunately, the secretin test is not widely available. Studies suggest that EUS may be useful to establish the diagnosis and evaluate the severity of the disease. Additional studies are needed to define the sensitivity of EUS for the diagnosis of early disease. MRCP may be used as a backup alternative if ERCP fails and EUS is unavailable or technically not feasible, such as in patients with Roux-en-Y-gastrojejunostomy.

COMPLICATIONS

Pseudocyst

A pseudocyst is a collection of pancreatic juice, outside the normal boundaries of the ductal system, that is enclosed by a fibrous tissue membrane. It is the most common complication of chronic pancreatitis, occurring in up to 25% of cases in some series. [220] Most chronic pseudocysts occur in patients with chronic alcoholic pancreatitis, [221] and they are found more commonly in the body of the pancreas than in the head or tail. [221]

Little is known about the mechanism responsible for the initial formation of pancreatic pseudocysts. They probably result from rupture of a pancreatic duct, activation of interstitial pancreatic juices, necrosis of the surrounding parenchyma, escape of the pancreatic juice into the lesser sac, and reaction of local mesothelial cells to wall off the collection by a fibrous membrane. Once initiated, expansion of the embryonic pseudocyst continues until the surrounding tissue pressure equals pancreatic secretory pressure, which is usually elevated in chronic obstructive pancreatitis. Although not conclusively demonstrated, some evidence exists to suggest that pancreatic juice contained within a pseudocyst can exchange with plasma. If so, this may represent a mechanism responsible for resorption of immature pseudocysts and spontaneous resolution of chronic pseudocysts.

Pain is the major symptom of pancreatic pseudocyst. Pseudocyst should be suspected in a patient with stable chronic pancreatitis who experiences worsening of abdominal pain. Abdominal ultrasonography is most frequently used in the diagnosis and management of pseudocysts. It is best used as a serial monitor of pseudocyst size in those patients in whom definitive drainage is delayed. CT has emerged as the single most accurate method of diagnosing pancreatic pseudocyst. [222] In addition to its high accuracy rate, it is capable of providing structural details, such as the size of the common duct or pancreatic duct. This has important bearing on the choice of an operative approach. In the presence of dilation of either or both the common bile duct and pancreatic duct, drainage of the pseudocyst alone is rarely sufficient. [223] Although some surgeons have advocated ERCP for all patients with pseudocysts, [224], [225] others have not found routine preoperative pancreatography desirable for fear of secondary infection, which may occur in up to 25% of cases. [225] It is generally agreed, however, that patients with suspected concomitant common duct obstruction should undergo ERCP before surgical drainage.

In contrast to acute pseudocysts, chronic pseudocysts, especially those larger than 6 cm, almost never resolve spontaneously. [226], [227] On the other hand, they also do not carry the same risk for serious complications as acute pseudocysts. In a study involving 75 patients with chronic pancreatitis and pseudocyst, about half of the patients could be managed conservatively. [228] Only one pseudocyst-related complication occurred in this group, and there was no mortality. According to this study, indications for operative management include persistent abdominal pain, enlargement, or complications of pseudocyst.

Effective treatment of pseudocyst includes excision and internal or external drainage. Surgical excision provides definitive treatment and is feasible when the cyst is localized to the tail of the pancreas. In most cases, however, internal drainage is the treatment of choice. Pseudocysts that are more than 6 weeks old usually have a mature cyst wall that permits internal drainage, where the cyst is anastomosed to the stomach (cystogastrostomy), duodenum (cystoduodenostomy), or jejunum (cystojejunostomy). The fluid in the pseudocyst drains into the gastrointestinal tract, and the cyst cavity collapses and becomes obliterated, usually within 1 week. There also have been reports of endoscopic drainage for the treatment of pseudocyst in chronic pancreatitis. [229], [230] A pseudocyst that indents the stomach or duodenum can be punctured transendoscopically using electrocautery or lasers to create a fistulous tract between the pseudocyst and the stomach or duodenum. Cremer and colleagues [229] reported a success rate of 96% for endoscopic cystoduodenostomy and 100% for endoscopic cystogastrostomy in 33 patients with chronic pancreatitis. No significant complications were observed after successful endoscopic cystoduodenostomy, and the two complications of endoscopic cystogastrostomy were gastric hemorrhage and iatrogenic pseudocyst infection. To decrease the risks for bleeding and perforation, the use of endosonography-guided drainage of pancreatic pseudocyst has been reported. [231] Potentially, the precise ultrasonographic definition of the cyst and local anatomy at the time of endoscopic drainage may improve the safety of endoscopic drainage, but the results are too preliminary for adequate evaluation.

External drainage usually is used for patients with infected pseudocysts and for those in whom the wall of the pseudocysts is not mature enough to hold sutures. Drainage is achieved by surgical placement of a large catheter in the pseudocyst, and the fluid drains externally through the abdominal wall. This form of treatment usually is safe but is associated with a higher recurrence rate than internal drainage. [232]

CT-guided, percutaneous, transabdominal catheter drainage of chronic pseudocysts has gained popularity and is considered by some a preferred alternative to surgical treatment in the management of pancreatic pseudocysts. Van Sonnenberg and colleagues [233] reported a 90% success rate in the percutaneous drainage of 101 pancreatic pseudocysts. The mean duration of drainage was 20 days. There was no mortality, and the only major complication was superinfection in four cases. Adams and Anderson, [234] in a retrospective study, also found that percutaneous catheter drainage was as effective as internal drainage in the management of pseudocyst, although drain tract infection was as high as 48%. In a long-term retrospective study of 42 patients undergoing percutaneous catheter drainage, however, Criado and associates [235] reported that in nine patients, the pseudocysts failed to resolve, and seven recurred after initial resolution. Hence, the effectiveness of percutaneous drainage in the management of pseudocysts remains to be established.

Somatostatin is a potent inhibitor of pancreatic secretion and therefore may be useful in the treatment of pancreatic pseudocyst. Gullo and Barbara [236] reported that 2-week treatment with octreotide, a long-acting somatostatin analog, resulted in a 42% reduction in size of the pseudocysts in four of seven patients. In addition, octreotide also appears to be effective in decreasing persistent catheter drainage from chronic pseudocyst. [237], [238] Therefore, patients with pancreatic pseudocyst resistant to drainage should be offered a course of octreotide before surgery is contemplated.

Pancreatic Ascites

The incidence of pancreatic ascites in chronic pancreatitis is usually considered to be less than 1%. It occurs as a consequence of persistent leakage of pancreatic juice from a pseudocyst or the pancreatic duct. Fifteen percent of patients with a pseudocyst may have pancreatic ascites. [239] On the other hand, 60% of patients with pancreatic ascites have pseudocysts. [240], [241] Pancreatic ascites occurs typically in alcoholic patients with cirrhosis, who may complain of mild to moderate abdominal pain because of distention. The diagnosis is often erroneously attributed to decompensated cirrhosis; however, paracentesis reveals elevation of pancreatic enzyme levels and a high total protein or albumin level. These findings are diagnostic.

Most patients need surgery. A period of 2 to 3 weeks of observation and medical treatment is warranted. During this period, the patients are given nothing orally and are placed on parenteral nutrition to improve their nutritional status. Suppression of pancreatic secretion by agents such as octreotide may be tried (100 to 250 µg subcutaneously every 4 hours). With this regimen, the ascites may resolve spontaneously in up to one third of patients, and surgery can be avoided. For patients who require an operation, ERCP should be performed to identify the precise site of the leak.

Pancreatic Fistula

External pancreatic fistula as a complication of chronic pancreatitis is rare. It occurs most commonly after operative or percutaneous drainage of a pseudocyst. It also may follow pancreatic biopsy or arise from a leaky pancreatic anastomosis. Clinically, pancreatic fistula should be suspected when clear fluid drains from a cutaneous

orifice, and the diagnosis is confirmed when the amylase content of the fluid is found to be elevated. Treatment is conservative and consists of fluid and electrolyte replacement, nutritional support, and eradication of infection if present. Somatostatin, which suppresses pancreatic secretion and decreases the volume of fistula drainage, may be used. During the course of therapy, a fistulogram should be obtained when the tract is well formed. If there is ductal obstruction between the site of the leak and the duodenum, surgical repair may be necessary. Most of the fistulas, however, close spontaneously.

Splenic Vein Thrombosis

Splenic vein thrombosis with extrahepatic (sinistral) portal hypertension is a well-known complication of chronic pancreatitis. [242], [243] Because the splenic vein courses along the posterior surface of the pancreas, thrombosis may occur as a result of peripancreatic inflammation associated with acute attacks or with pseudocyst formation and extension. Rosch and Herfort [244] found occlusion of the splenic vein in about 4% of cases of chronic pancreatitis studied with splenoportography. Almost all patients had some impingement on the splenic vein. Varices involving the esophagus, stomach, duodenum, and colon have been described with resultant hemorrhage. A more recent study [243] involving 484 consecutive patients with chronic pancreatitis reported that splenic vein thrombosis occurred in 7% (34 patients), of whom 35% (12 patients) had gastroesophageal varices. Overall, 18% of them bled either before first surgery or on follow-up. The 22 patients who underwent splenectomy never bled after surgery. These findings support the recommendation that patients who have symptomatic sinistral portal hypertension who are undergoing surgery should have concomitant splenectomy. Splenectomy also should be strongly considered in patients undergoing surgery for chronic pancreatitis who have varices secondary to splenic vein thrombosis regardless of presence or absence of prior history of variceal bleeding.

TREATMENT

Treatment of chronic pancreatitis is aimed mainly at the control of pain and the correction of malabsorption with adequate pancreatic enzyme replacement. Clinical diabetes occurs relatively late in the disease, and insulin is usually needed for its management.

Control of Pain

The management of pain in chronic pancreatitis can be a challenging problem. It is the symptom that most commonly prompts the patient to seek medical attention and is the one that is most difficult to manage. Pain patterns are highly variable, and this may reflect different causative mechanisms. The problem is further exacerbated by a lack of good controlled trials. As highlighted in an American Gastroenterological Association technical review on treatment of pain in chronic pancreatitis, [245], [246] "a reliable database for making sound judgements and recommendations is lacking in most areas."

Avoidance of Alcohol A long-held clinical aphorism is that avoiding alcohol ingestion decreases the frequency and severity of abdominal pain in chronic alcoholic pancreatitis. Trapnell [247] reported that 75% of his patients with chronic alcoholic pancreatitis experienced pain relief when they stopped drinking. This observation may be explained by the stimulatory effect of alcohol on pancreatic secretion. [248] The relationship between alcohol and pain in chronic pancreatitis, however, is controversial. Bornman and colleagues [248] noted that half of their patients with painful pancreatitis and half of their patients with painless pancreatitis continued to drink. Similar observations were made by Marks and associates. [249] These findings call into question whether continued alcohol ingestion is related to pain. It is conceivable that pancreatic pain is related to exocrine pancreatic secretion. In patients who maintain significant exocrine secretory function, pain may be provoked by alcohol, which acts as a secretagogue. In patients whose exocrine secretion is drastically reduced, alcohol no longer plays a role in the mechanism of pain. Further studies are needed to clarify the role of alcohol in pain production.
Analgesics Analgesics remain the main method for pain control in chronic pancreatitis. Initially, nonnarcotic analgesics, such as salicylates or acetaminophen, should be used. Preferably, these analgesics should be given before meals to prevent postprandial exacerbation of pain. Drug doses should be individualized, and the lowest effective dose should be used. As the severity of pain increases, the dose strength or frequency of these simpler analgesics should be increased before switching to narcotics. In severe cases, however, opiate analgesics are required, and they should not be withheld for fear of narcotic addiction.
Celiac Plexus Block Percutaneous, radiologically or ultrasonographically guided, celiac ganglion alcohol injection has been used for control of pancreatic pain. This technique has been helpful in patients who have severe pancreatic pain secondary to pancreatic cancer. [250] In small, uncontrolled series of patients with chronic pancreatitis and debilitating pain, this procedure has produced mixed results. The occasional benefits almost never last for more than a few months, [251] and repeated treatment may not be as effective. The risk for procedure-induced transient hypotension, nerve root pain, and focal neuropathic damage, combined with the need for repeated blocking, even if successful, make this an unattractive technique for most patients. [251] An open trial of celiac plexus block using botulinum toxin A instead of alcohol was unsuccessful. [252] The advances in EUS have allowed the performance of celiac plexus block under the guidance of EUS. This has become a relatively simple and short outpatient procedure. Because of the anterior transgastric approach taken during the procedure, it has mostly eliminated the risk for nerve or spinal cord injury. Recently, a prospective study with EUS-guided celiac plexus block was performed in 90 patients with painful chronic pancreatitis. [253] A significant improvement in pain score occurred in 55% of patients. Unfortunately, the benefit persisted beyond 24 weeks in only 10%. Hence, although EUS-guided celiac plexus block is safe and well tolerated, it is not very effective to treat pain in chronic pancreatitis because it offers only temporary relief.
Enzyme Therapy Much interest has been generated by a series of observations suggesting that the intralumenal action of pancreatic proteases plays an important role in regulating pancreatic enzyme secretion. The underlying concept of feedback regulation of the pancreas is based primarily on studies in rats that showed that diversion of pancreatic juice from the duodenum stimulates CCK release and pancreatic enzyme secretion. [254], [255] On the other hand, intraduodenal administration of trypsin or chymotrypsin inhibits the release of CCK and pancreatic enzyme secretion. The increased plasma CCK levels and pancreatic secretion after diversion of pancreatic juice appears to be mediated by a trypsin-sensitive substance secreted by the proximal small intestine that has been designated CCK-releasing factor (CCK-RF). [256] When trypsin is present, this peptide is cleaved and inactivated. CCK-RF may act as a mediator of pancreatic enzyme secretion in response to dietary protein intake in rats. Dietary protein in the intestine competes for the trypsin that would otherwise inactivate CCK-RF. [257] The resulting increase of CCK-RF in the intestinal lumen enhances CCK release, stimulating pancreatic enzyme secretion. A number of CCK-releasing peptides have been identified. These include a peptide extracted from porcine intestinal extracts with a structure identical to the diazepam-binding protein [258] and a lumenal CCK-releasing peptide isolated from the duodenal secretion of rats. [259] The physiologic significance of these releasing peptides is yet to be defined. The protease-sensitive feedback mechanism was first reported to occur in humans in 1977. [260] Subsequently, several groups of investigators confirmed that intestinal administration of trypsin or chymotrypsin inhibits pancreatic enzyme secretion. [260], [261], [262], [263] and [264] Thus, it is conceivable that in patients with chronic pancreatitis, decreased enzyme secretion may result in the hyperstimulation of the pancreas by elevated plasma CCK levels, with resultant pain. Some, [265], [266] and [267] but not all, [268], [269] studies reported elevated plasma CCK levels in patients with chronic pancreatitis. It has been proposed that effective enzyme replacement therapy should reduce pancreatic stimulation, decrease intraductal pressure, and diminish pain. This hypothesis was tested in several clinical studies. In a double-blind, crossover study, Isaksson and Ihse [270] reported pain relief in 15 of 19 patients with chronic pancreatitis. There was a 30% reduction in pain intensity and a significant decrease in the frequency of pain attacks in patients on active enzymes. Slaff and colleagues [261] studied 20 patients with painful chronic pancreatitis in a double-blind, crossover study. Pain reduction was observed in 9 patients with mild to moderate pancreatic insufficiency, whereas only 2 of 11 with severe insufficiency responded to the treatment with enzyme replacement. Most of the good responders were women with idiopathic chronic pancreatitis, whereas most of the poor responders were men with alcohol-induced chronic pancreatitis. Favorable response was also reported by Ramo and associates, [271] who showed significant pain relief after self-administration (ad libitum) of an enzyme preparation, compared with administration of the regular dose in 10 patients with painful chronic pancreatitis. These three double-blind studies [261], [270], [271] demonstrated good results by enzyme treatment in 73% (36 of 49) of the patients evaluated. A double-blind, 4-week crossover study from Denmark, however, showed that oral administration of two capsules of Pancrease with each meal did not significantly control pain compared with placebo. [272] Another multicenter 2-week controlled study in more than 40 patients using an enteric-coated supplement also showed no benefit. [273] Feedback inhibition of pancreatic enzyme secretion depends on delivery of high concentrations of protease to the duodenal lumen. Enteric-coated preparations preferentially release most of their enzymes distal to the duodenum and may therefore be less effective at suppressing CCK levels and producing pain relief. [274] Finally, a 4-month double-blind, crossover study used an enteric-coated preparation with microspheres that release their contents at a lower pH (pH > 5.0) than most commonly used microspheres (pH > 6.0). Despite a high delivery rate of trypsin into the duodenum, this study showed no benefit in pain scores in patients receiving pancreatic enzymes compared with placebo. [275] However, this study contained a small number of women and evaluated patients with recurrent rather than chronic pain. Long-term trials comparing nonenteric and enteric-coated preparations in patients stratified according to etiology, severity, and pattern of disease should help to clarify these issues. Until then, it seems reasonable to initiate a trial of high-dose, non–enteric-coated oral pancreatic enzymes taken with meals for several weeks in any patient with painful chronic pancreatitis. The best results may be seen in pancreatitis of nonalcoholic etiology, with symptoms of constant, rather than recurrent, pain and only mild to moderate pancreatic insufficiency. [276]
Treatment with Octreotide Somatostatin is a naturally occurring hormone that has been shown to inhibit pancreatic secretion. [277] Theoretically, it may also have cytoprotective action and beneficial effects on the reticuloendothelial system in the setting of pancreatic inflammation. [278], [279] and [280] In addition, clinical studies have shown that somatostatin has antinociceptive activity in human [281], [282] and animal [283], [284] models of pain, suggesting that this peptide may be helpful for pain control in chronic pancreatitis. Somatostatin also appears to gain its beneficial effects in inflammatory conditions through its inhibition of cytokine release. In a recent study,

somatostatin was shown to reduce markedly the secretion of IL-8 and IL-1 from intestinal epithelial cells. [285] In another study, somatostatin significantly inhibited IL-8 and interferon-? production from activated peritoneal macrophages. [286] Octreotide is a synthetic, long-acting analog of somatostatin that has been shown to inhibit CCK release and both basally and neurally stimulated pancreatic secretion. [287], [288] and [289] Octreotide's long duration of action allows subcutaneous dosing and makes it a more practical alternative for the treatment of chronic pancreatitis than the short-acting native somatostatin. The drug is well tolerated, although long-term therapy predisposes to cholesterol gallstones. Unfortunately, controlled trials using octreotide for chronic pancreatitis pain are limited. Short 3-day studies, using 100 and 150 µg three times a day, showed no benefit. [290] A multicenter randomized 4-week study of 91 patients with advanced chronic pancreatitis and severe pain was performed. [291] Octreotide, at doses ranging from 40 to 100 µg three times daily, was compared with placebo. Although results in this pilot study did not reach statistical significance, 200 µg three times a day of octreotide produced the greatest pain relief (65% versus 35% of patients with placebo), especially in patients with constant as opposed to intermittent pain. Additional studies are needed to clarify the role of octreotide in the management of chronic pancreatitis pain. Recently, a long-acting formulation of octreotide acetate (Sandostatin LAR Depot) was developed for patients requiring long-term octreotide therapy. It consists of octreotide acetate microencapsulated by a biodegradable polymer. Drug release occurs slowly as cleavage of the polymer takes place primarily through tissue fluid hydrolysis. After single intramuscular injection, the serum octreotide level reaches plateau concentrations at about day 14 and remains relatively constant during the following 3 to 4 weeks. This long acting form of octreotide may provide significant benefit over octreotide because it requires only monthly injection. A double-blind, randomized, multicenter trial of the long-acting octreotide in the treatment of painful chronic pancreatitis is ongoing.

Endoscopic Therapy Endoscopic therapy has been used for control of pain in chronic pancreatitis, with the aim of alleviating obstruction of flow caused by ductal strictures, stones, or papillary stenosis. Ductal strictures are sometimes treated by balloons or dilating catheters, but in most cases, dilation is followed by stent placement across the stricture. Technical success in reported studies ranges from 82% to 100%, [292], [293], [294], [295], [296], [297] and [298] and pain improvement occurs in 55% to 100% of patients during a follow-up of 8 to 39 months. True success rates are difficult to decipher, however, because stent placement frequently was performed with other procedures. Ponchon and colleagues [298] used a standardized protocol to evaluate endoscopic stenting for pain relief in a highly selected subgroup of patients with chronic pancreatitis. Patients with multiple strictures, pancreatic calculi, pancreatic divisum, common duct narrowing, any duodenal impingement, or a pseudocyst larger than 1 cm were excluded. They successfully placed 10-French, multi–side-hole stents after biliary and pancreatic sphincterotomy and balloon dilation of strictures in 28% of 33 patients (85%) with a distal pancreatic duct stricture and upstream dilation. The stents were exchanged at 2-month intervals, for a total stenting duration of 6 months. The report was based on 23 patients observed for at least 1 year after removal of the stent. During the stenting period, 21 patients (91%) had resolution or reduction in pain, and 17 discontinued analgesic medications. Twelve patients (52%) had a persistent beneficial outcome for at least 1 year after stent removal. Disappearance of the stenosis on pancreatography, at stent removal, and 1 year later and reduction in the diameter of the pancreatic duct were significantly associated with relief of pain. Therefore, it appears that in patients with isolated stricture, endoscopic stenting may be useful to relieve pain. Based on reports in the literature, the incidence of occlusion of pancreatic stents appears to be similar to that for biliary stents. [299] Sherman and Lehman [300] reported that 50% of pancreatic stents (5 to 7 French) were occluded within 8 weeks of placement. Another complication is stent migration, which may be upstream (into the duct) or downstream (into the duodenum). Migration in either direction may be heralded by return of pain or pancreatitis. Johanson and associates [301] reported inward migration in 5.2% of patients and duodenal migration in 7.5%. Modifications in the design of pancreatic stents may reduce the frequency of such occurrences. [302] Long-term studies of stent placement also indicated that morphologic changes occurred in most patients. These usually are mild and reversible in most patients. [303], [304] Endoscopic techniques also have been used for the removal of pancreatic stones in chronic pancreatitis. In different series, pancreatic duct stones were removed after endoscopic sphincterotomy in 27% to 100% of patients. [305], [306] In some cases, stone removal was facilitated by fragmentation with extracorporeal shock wave lithotripsy, with clinical improvement observed in 50% to 80% of cases in which the stones were successfully removed. [306] Inherent in these various therapies is the belief that stones are themselves causing symptoms by intensifying obstruction and that their removal will improve pancreatic flow and thereby reduce symptoms. On the other hand, it is also possible that pancreatic stones form as a result of pancreatic stricture and abnormal pancreatic juice protein and have little to do with pain. None of the studies on endoscopic treatment of chronic pancreatitis is controlled, and no valid comparisons have been made among medical, surgical, and endoscopic treatments. These procedures should therefore be considered experimental and performed only in patients entered into prospective, randomized trials. In one study, minor papilla endoscopic sphincterotomy was performed in 52 patients with pancreas divisum who had either chronic abdominal pain, acute recurrent pancreatitis, or chronic pancreatitis. [307] Among the three groups of patients, only those with acute recurrent pancreatitis benefited from the endoscopic procedure. This suggests that minor papilla sphincterotomy should be avoided in patients with pancreatic divisum and chronic pancreatitis.

Surgical Treatment After all medical measures have failed to relieve pain, surgery should be considered. The type of surgery is selected according to the perceived mechanism for the pain. When the main pancreatic duct is dilated, it is assumed that there is pancreatic duct obstruction and that pancreatic duct hypertension is the cause of pain. On the other hand, if there is no ductal dilation, pain may result from diffuse parenchymal disease or blockage of small side ducts by stones, protein plugs, or scars. Relief of major duct obstruction should not affect this type of pain. Therefore, the type of surgery should be chosen according to the severity of pain, ductal morphology, and the extent of parenchymal disease. Patients who have ductal dilation have a 70% to 80% chance of obtaining pain relief with either a partial resection with pancreaticojejunostomy or lateral pancreaticojejunostomy (modified Puestow procedure). The modified Puestow pancreaticojejunostomy is particularly suitable for patients who have ductal obstruction and dilation. It is a safe and effective operation, with a morbidity rate of less than 5%, mortality rate of less than 2%, and effective pain relief rate on the order of 80%. [308], [309] On the other hand, for patients with moderate to severe parenchymal disease and no ductal dilation, partial pancreatic resection should be considered. Ninety-five percent distal resection is recommended for patients with diffuse parenchymal disease, [249] whereas local resection of major site of involvement may be sufficient for those with regional parenchymal disease. [249] Overall, 50% of these patients have had satisfactory results. Near-total pancreatectomy, or Whipple procedure, includes sacrificing portions of normal stomach and duodenum with resulting metabolic and digestive complications. To correct these problems, the original Whipple procedure underwent two subsequent procedural modifications. In 1978, Traverso and Longmire [310] described pylorus-preserving pancreatic duodenectomy, thereby avoiding the digestive complications of partial gastric resection. Several years later, Beger and associates [311] described a more conservative procedure that resected the diseased pancreatic head and preserved all extrapancreatic organs, including the duodenum (Fig. 95-10). A randomized trial comparing these two procedures in patients with painful chronic pancreatitis was performed. [312] There was no postoperative mortality, and morbidity was comparable in both groups (15% to 20%). After 6 months of follow-up, patients who underwent duodenum-preserving pancreatic head resection (the Beger procedure) experienced less pain, better preservation of insulin secretion and glucose tolerance, and more stable weight than those who underwent pylorus-preserving Whipple resection. This suggests clinical superiority of the Beger procedure as compared with the pylorus-preserving Whipple resection. However, the limited sample size and lack of long-term follow-up preclude any definitive conclusions at this time.

FIGURE 95-10. Duodenum-preserving resection of the pancreatic head as described by Beger. A metal probe is inserted through the common bile duct into the duodenum to prevent inadvertent injury during resection of the pancreatic head. **B:** Reconstruction includes an end-to-end pancreaticojejunostomy and a side-to-side anastomosis between the jejunum and the rim of the cavity remaining after resection of the pancreatic head. *D,* duodenum; *HA,* hepatic artery; *PD,* pancreatic duct; *RC,* resection cavity of the pancreatic head; *SMV,* superior mesenteric vein; *ST,* stomach; *T,* tube; *TC,* transverse colon. (From ref. [315].)

In 1987, Frey and Smith [313] described another operation for painful chronic pancreatitis that consisted of local resection of the pancreatic head combined with longitudinal pancreaticojejunostomy (Fig. 95-11). This procedure was designed to improve decompression of the head of the pancreas, which often was not drained well by conventional pancreaticojejunostomy. Frey and Amikura [314] reported their results in 50 patients with chronic pancreatitis in a 7-year follow-up study. There were no operative deaths, and 22% of patients developed postoperative complications. Pain relief was complete or improved in 87% of patients. Twenty percent of patients developed new-onset postoperative diabetes, and 11% experienced worsening of their diabetes.

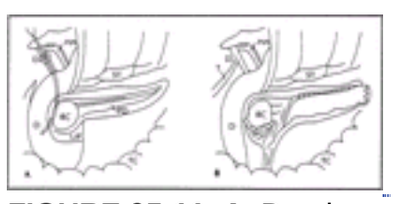

FIGURE 95-11. A: Duodenum-preserving resection of the pancreatic head as described by Frey. A metal probe is inserted through a choledochotomy into the duodenum before resecting the pancreatic head mass to minimize the risk for bile duct injury. **B:** Reconstruction consists of a side-to-side pancreaticojejunostomy including the resection cavity of the pancreatic head. *D,* duodenum; *HA,* hepatic artery; *PD,* pancreatic duct; *RC,* resection cavity of the pancreatic head; *ST,*

stomach; *T,* tube; *TC,* transverse colon; *UP,* uncinate process. (From ref. [315].)

The two techniques of duodenum-preserving resection of the pancreatic head (the Beger and Frey procedures) were compared in a prospective study. [315] There was no operative mortality in both groups, and postoperative morbidity was significantly lower in patients who had undergone the Frey procedure, which is technically less demanding. Both groups of patients reported similar postoperative decreases in pain and improved quality of life index scores. Postoperative pancreatic endocrine and exocrine function was also similar in both groups. These results are encouraging, but the mean follow-up of 1.5 years is relatively short. Long-term studies are needed to determine whether either or both procedures will suffer from attrition of the initial good results. It should also be noted that the duodenum-preserving resections are not appropriate for all patients with symptomatic chronic pancreatitis but seem especially useful for patients with predominant involvement of the pancreatic head. Recently, Nealon and Matin [316] evaluated the success rates of different surgical procedures in relieving both the chronic abdominal pain and the recurrent exacerbations associated with chronic pancreatitis. Patients were divided into three groups based on the characteristics of their pain: severe unrelenting pain alone, severe pain with intermittent acute exacerbations, and intermittent acute exacerbations only. Postoperative pain relief was achieved in 83% of patients overall, with 86% relief from lateral pancreaticojejunostomy, 67% from distal pancreatectomy, and 91% from pancreatic head resection. It appears that pancreatic head resection was most effective for relief of chronic pain (91%), whereas distal pancreatectomy was least effective (67%). On the other hand, lateral pancreaticojejunostomy gave the best relief of acute pain exacerbations (91%), whereas distal pancreatectomy was rather ineffective (33%). Pancreatic head resection was much more effective in relieving chronic pain (91%) than pain related to acute exacerbations (58%). These observations suggest that stratification of patients according to their pain patterns may be helpful to select the most effective surgical procedure for pain relief. In patients undergoing a near-total pancreatectomy, islet cell autotransplantation by infusion of islet cell preparations into the portal system may prevent diabetes. [317], [318] and [319] Islets from the resected pancreas can be isolated by mechanical disruption and collagenase digestion of the dispersed tissue and injected into the portal vein. Using this method, Farney and colleagues [319] reported that 9 of 22 patients with intraportal islet autografts were insulin independent for at least several months after surgery. Wahoff and associates [320] reported long-term insulin independence in 33% of 48 patients who underwent pancreatic resection combined with islet autotransplantation and followed up for 2 to 10 years. Insulin independence after islet autotransplantation correlated most closely with the number of islets transplanted and therefore relates to the available mass of pancreatic tissue and the degree of pancreatic fibrosis. Of those patients who received less than 200,000 islets, only 22% achieved long-term insulin independence. By comparison, nearly 60% of patients who received more than 300,000 islets achieved long-term independence. With recent advances in islet isolation techniques, there appears to be an overall improvement of the results of islet autotransplantation. Recent studies indicate that about 50% of patients undergoing total or subtotal pancreatectomy can become insulin independent after intraportal autologous islet transplantation. [321], [322] In the largest published series of 43 patients undergoing total or near-total pancreatectomy for chronic pancreatitis, insulin independence persisted in 34% of patients 2 to 10 years after surgery, and no graft failure occurred after 2 years. [323] Patient selection is important to determine the success of islet cell autotransplantation on diabetes after total pancreatectomy for chronic pancreatitis. Patients with end-stage fibrotic glands and those who are narcotic dependent are unlikely to do well postoperatively. [322]

Summary of Treatment Approach for Control of Pain We recommend the following approach to treat pain of chronic pancreatitis: When a patient first presents with pain or shows a different pain pattern, we determine whether there is an anatomic reason for the occurrence of or a change in pain. Pseudocyst, phlegmon, obstruction, and peptic ulcer are ruled out by appropriate tests. Patients are advised to stop all alcohol ingestion and to use salicylate or acetaminophen as needed. We also recommend the use of enzyme (e.g., 8 tablets of Viokase with each meal), especially in patients with non–alcohol-induced chronic pancreatitis. If pain continues, a trial of octreotide may be considered. After all medical measures have failed, we recommend surgery; some centers, however, may consider celiac plexus block before surgery (Fig. 95-12).

FIGURE 95-12. Algorithm for management of the patient with painful chronic pancreatitis.

Management of Pancreatic Insufficiency

At first glance, treating malabsorption secondary to pancreatic insufficiency might appear to be relatively easy. It would seem that simple oral replacement of pancreatic enzymes would be efficacious. Unfortunately, complete correction of steatorrhea is rarely accomplished. This is related to insufficient amounts of enzyme in oral supplements and acid pepsin inactivation of pancreatic enzymes.

The maximal postprandial delivery of pancreatic lipase is about 140,000 IU/hour for 4 hours after meals. [120] It has been shown that malabsorption does not occur if more than 5% of the normal maximal enzyme output is delivered to the duodenum. To meet this requirement, 28,000 IU of lipase should be delivered during a 4-hour postprandial period. Pancreatic supplements are highly variable in enzyme activity, with lipase content ranging from 0 to 8000 IU per tablet. [145], [324], [325] Therefore, if the clinician uses a commercially available preparation containing 3500 IU per tablet of lipase, then under the best of circumstances, at least 8 tablets or capsules must be taken per meal to abolish malabsorption. Smaller amounts of enzymes may reduce steatorrhea but not abolish it. Therefore, the clinician should choose those commercial enzyme preparations that have been shown to contain higher lipase activity, and ensure that sufficient amounts have been used (Table 95-5).

		CONTENT (UNITS)		
PREPARATION	TYPE	*Lipase*	*Amylase*	*Protease*
Cotazyme	C	8000	30,000	30,000
Cotazyme-S	ECMS	5000	20,000	20,000
Creon 10	ECMS	10,000	33,200	37,500
Creon 25	ECMS	25,000	74,700	62,500
Ilozyme	UCT	11,000	30,000	30,000
Ku-zyme HP	ECMS	8000	30,000	30,000
Pancrease	ECMS	4000	20,000	25,000
Pancrease MT-4	ECMT	4000	12,000	12,000
Pancrease MT-10	ECMT	10,000	30,000	30,000
Pancrease MT-16	ECMT	16,000	48,000	48,000
Protilase	ECMS	4000	20,000	25,000
Viokase	UCT	8000	30,000	30,000
Viokase	P	16,800	70,000	70,000
Zymase	ECMS	12,000	24,000	24,000

C, capsule; ECMS, enteric-coated microspheres encased in a cellulose capsule; UCT, uncoated tablet; ECMT, enteric-coated microtablets encased in a cellulose capsule; P, powder.

Adapted from Berardi RR, Dunn-Kucharski VA. Pancreatitis and cholelithiasis. In: DiPiro JT, Talbert RL, Hayes PE, et al, eds. Pharmacotherapy: a pathophysiologic approach. Norwalk, CT: Appleton & Lange, 1992:614.

TABLE 95-5 Commercial Pancreatic Enzyme Preparations

Another important factor to consider is acid-peptic inactivation of pancreatic enzymes. When duodenal samples were examined for lipase and trypsin activity, less than 8% of ingested lipase and less than 22% of ingested trypsin was recovered, regardless of the dosing schedule used (either prandial or hourly dosing). To

eliminate steatorrhea in pancreatic insufficiency, a large number of tablets are necessary. Hyperuricosuria [326] and kidney stones have been associated with such high dosing of pancreatic supplements secondary to their high purine content, and therefore, this high dosing schedule may not be clinically justified. The relatively low delivery of lipase into the proximal small bowel compared with the delivery of trypsin with routine treatment schedules may explain the relative impossibility of completely correcting steatorrhea in pancreatic insufficiency as well as the usual correction of azotorrhea.

Preventing acid-peptic neutralization of enzyme supplements by coating capsules with acid-resistant and alkali-sensitive materials or by using antacids has produced mixed results. [327], [328] In most studies, there has been little success with oral antacids or enteric coating with regard to clinical improvement or alleviating steatorrhea. Antacids increase gastric secretion, and dilution of enzyme concentrations below critical levels may explain the relative ineffectiveness of antacids. [328] Enteric coating is effective only if pancreatic enzymes are delivered into the duodenum with ingested food from the stomach and adequate intraduodenal dissolution occurs. Because emptying of large tablets from the stomach may be different from that of typical antral contents, and duodenal and gastric pH have been shown to be low (pH < 4.0) for prolonged times in many patients with pancreatic insufficiency, [329], [330] enteric coating may not be an effective improvement over oral pancreatic supplements alone.

Pancrease (pancreatin coated with a pH-dependent polymer) is another improvement in pancreatic enzyme supplements. [331] Because this supplement is stable at an acidic pH (pH < 4.0) and dissolves at a pH of greater than 5.0, it is most effective (i.e., better than standard tablets alone) in patients who maintain acidic pH levels in the stomach postprandially. This maneuver delivers pancreatic enzymes to the upper small bowel intact, whereas standard supplements are irreversibly inactivated. In several studies, the mean reduction of steatorrhea with the use of Pancrease is not better than that with Viokase given with meals. [328], [332] Normally, in chronic pancreatitis, the gastric pH increases to above 5.0 in the early postprandial period and then decreases to less than 4.0. It is likely that, under these circumstances, pancreatin initially will be liberated from its enteric coat, but its enzyme activity will be irreversibly inactivated when the intragastric pH falls to less than 4.0. Conversely, if gastric and duodenal pH remain low (pH < 4.0) throughout the postprandial period, [333] the enteric coat of Pancrease remains intact as it traverses the upper gastrointestinal tract, and pancreatin is liberated en masse only as it reaches the jejunum, where intralumenal pH is greater than 5.0. This may explain the effectiveness of Pancrease in abolishing steatorrhea in some patients who secrete more acid and presumably can maintain an acidified upper small intestine for a prolonged period of time.

Until recently, the major criterion for determining the efficacy of enteric-coated enzyme preparations was the size of the microspheres, which influences the timing of enzyme delivery to the intestine. It has been shown that microspheres with a diameter of about 1.4 mm appear to mix with chyme most thoroughly and are emptied from the stomach at the same rate as food; large spheres remain in the stomach after food has been emptied into the small bowel and are less effective in correcting malabsorption in chronic pancreatitis. [334] Some commercially marketed microspheres (more than 2 mm) of pancreatin empty too slowly to be effective in digestion of food. This may explain why some enteric-coated preparations are not effective in correcting pancreatic steatorrhea. However, a recent prospective, randomized, double-blind, multicenter, crossover trial in chronic pancreatitis patients with a stool fat excretion of 7.5 g/day showed that there was no difference between Creon given as microspheres with a diameter of 1 to 2 mm and as mini-microspheres with a diameter of 0.7 to 1.6 mm. [335] The mean stool fat excretion per day was about 16 gm in both groups. Thus, reducing microsphere size does not improve the efficacy of the enzyme supplements. An explanation for this observation may be that in vivo microspheres aggregate into larger particles in the presence of free oil. [336] It should be noted that high doses of pancreatic enzyme supplements in children with cystic fibrosis have been associated with recurrent bowel obstruction due to fibrosing colonopathy characterized by dense submucosal fibrosis in the large bowel. In recent years, two cases of fibrosing colonopathy have been reported in adult patients, one with and one without cystic fibrosis. [337], [338] The cause of fibrosing colonopathy associated with high doses of pancreatins remains speculative. [339]

Histamine H_2 receptor antagonists decrease acid production, pepsin activity, and gastric secretory volume simultaneously and should optimize pancreatic enzyme concentrations. [328], [340] Duodenal lipase and trypsin activities are consistently increased with concomitant Viokase and histamine H_2 receptor antagonist treatment. Cimetidine should be considered only in patients who do not respond to standard oral pancreatic supplements because of lifelong increased cost and potential unknown long-term side effects. It has been shown that omeprazole further reduces fecal fat excretion in patients with cystic fibrosis receiving enzyme replacement for pancreatic insufficiency. [341] Therefore, in patients in whom histamine H_2 receptor antagonist fails to abolish steatorrhea, use of omeprazole should be considered.

We recommend the following approach to treat patients with pancreatic insufficiency. It is critical that sufficient amounts of enzyme tablets be given (e.g., 8 tablets of Viokase with each meal) to abolish azotorrhea and significantly reduce steatorrhea. Most patients on this regimen achieve satisfactory nutritional status and become relatively asymptomatic. In some of the symptomatic patients, the number of tablets given prandially can be increased or the amount of dietary fat reduced. These measures usually are effective in alleviating symptoms. In occasional patients, the use of Pancrease may be necessary. The addition of histamine H_2 receptor antagonist or omeprazole should be reserved for those patients who are resistant to these maneuvers and who have documented acidic duodenal pH levels, because of cost considerations as well as potential long-term side effects. If all of these measures are ineffective, documentation of diagnosis (by pancreatic function testing) and exclusion of other contributing causes (celiac sprue, terminal ileal disease, or bacterial overgrowth) must be done.

Management of Pancreatic Diabetes

Clinical diabetes occurs relatively late in the disease and is an independent predictor of mortality of chronic pancreatitis. Ketoacidosis and diabetic neuropathy were relatively uncommon in this form of diabetes. This is probably due to the fact that in pancreatic diabetes, insulin secretion is not completely lost, and glucagon secretion is reduced. Although some patients may respond to oral hypoglycemics, insulin is often needed in most patients. Insulin requirements are usually lower than for most patients with genetic diabetes because insulin receptors usually are not down-regulated and insulin antibodies initially are not present. Overzealous control of blood glucose should be avoided because this may result in life-threatening hypoglycemia.

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CHAPTER 96

John P. Duffy and Howard A. Reber

NONENDOCRINE TUMORS OF THE PANCREAS

DUCTAL ADENOCARCINOMA OF THE PANCREAS

Approximately 29,000 new cases of pancreatic cancer occur every year in the United States, and almost all of these patients will eventually die from the disease. Although surgical resection of the tumor offers the only chance for cure, even after a “curative resection,” the median duration of survival is only 18 to 20 months, and the 5-year survival rate is 15%. [1] Although these overall statistics are sobering, certain groups of patients fare better. If the tumor is removed with clear surgical margins and no lymph node metastases are present, the 5-year survival is as high as 25%. [2] Patients with well-differentiated tumors may have as much as a 50% chance for surviving 5 years. Unfortunately, only a minority of patients fall into these categories.

A number of new and exciting developments have occurred in the field. They include a rapid growth in the characterization of the genetic abnormalities associated with the disease, the appearance of new diagnostic techniques that have improved our ability to rapidly diagnose and stage it, and effective new therapeutic modalities, which allow nonsurgical palliation in many patients. Curative surgery is performed with operative mortality rates of only 1% to 2%, compared to 10 times that figure a decade ago. A number of new operations exist, which hold the promise of more effective treatment and less morbidity. In this chapter, we discuss these advances and present a broad overview of the diagnosis and management of patients with pancreatic neoplasms.

EPIDEMIOLOGY AND RISK FACTORS

The peak incidence of pancreatic carcinoma occurs in the seventh decade of life, and there is a slight male to female predominance. [3] The overall incidence of the disease is 30% to 40% higher in the African-American population than in Caucasians. The highest incidence occurs in Maori men of New Zealand, in whom the likelihood of developing pancreatic cancer is twice that of Caucasians in the United States or Europe. [4]

A variety of observations support the notion that there are hereditary (genetic) factors that influence the development of pancreatic cancer. Reports of the disease in multiple family members of the same generation and within extended families show an autosomal dominant inheritance pattern. [5] Certain genetic disorders predispose to pancreatic cancer. They include multiple endocrine adenomatosis type I, hereditary nonpolyposis colorectal cancer, familial adenomatosis polyposis, Gardner syndrome, familial atypical multiple mole melanoma syndrome, Peutz-Jeghers (PJ) syndrome, and von Hippel-Lindau syndrome. [5], [6] However, genetic factors appear to play a role in only about 5% of all newly diagnosed cases of pancreatic cancer. [5]

Smoking

Around the world, the risk factor most strongly linked to pancreatic cancer is cigarette smoking. [6], [7] and [8] It approximately doubles the chance of developing the disease. The relation between number of pack years of smoking and the risk of pancreatic cancer is not as strong as it is with lung or oropharyngeal cancers, but it is generally true that the risk is greater with more cigarettes smoked. It is also curious that the risk of pancreatic cancer rapidly decreases when individuals discontinue cigarette use. The relative risk falls to about 1 only 10 to 15 years after quitting. The smoking of cigars and other forms of tobacco probably does not increase risk. Autopsy studies of smokers have revealed hyperplastic changes in the pancreatic ducts, alterations that have been noted to increase in proportion to cigarette consumption. [4]

Pancreatitis

A case control study in the United States Veterans Administration compared the occurrence of chronic pancreatitis in 2639 patients with pancreatic cancer to a

matched control group of 7774 subjects. A history of pancreatitis doubled the risk of developing pancreatic cancer. [9], [10] Other epidemiologic studies suggest that the relative risk of developing pancreatic cancer in patients with chronic pancreatitis is increased by up to 15 times when compared to control populations. This increase in risk also is seen in kindreds with hereditary pancreatitis. [11] This suggests that the etiology of the pancreatitis is not a factor, but that the changes associated with chronic inflammation and fibrosis in the pancreas are more important. Nevertheless, chronic pancreatitis accounts for only a small fraction of patients who have pancreatic cancer. [12] A multicenter international study found that 1.8% of patients with chronic pancreatitis developed pancreatic cancer during a mean follow-up of 7.4 years. [10]

Surgery

In one study, patients who had undergone a partial gastrectomy had a five to seven times greater risk of developing pancreatic cancer. [13] Another study found that there was an approximately threefold elevation in the risk of pancreatic cancer in patients who had undergone previous gastrectomy for peptic ulcer. [14] The apparent increase in incidence may be caused by altered metabolism of ingested carcinogens by the remaining stomach and small intestine following surgery.

Ethanol

A large number of epidemiologic studies in both the United States and Europe failed to find any consistent etiologic association between alcohol consumption and the development of pancreatic cancer. The evidence at the present time does not support the view that it is a risk factor. [7], [8]

Coffee

In an Italian multicenter study, consumption of more than three cups of coffee per day increased the risk for pancreatic cancer even when the data were controlled for potential confounding factors such as cigarette smoking or alcohol use, and when the analysis was restricted to nonsmoking coffee drinkers. [15] However, a case-control study in Japan and a metaanalysis of data published between 1981 and 1993 did not support the notion that either caffeinated or decaffeinated coffee or tea were risk factors for pancreatic cancer. [16]

Diet

In general, a high total energy intake correlates with an increased risk of pancreatic cancer. Products of plant origin and dietary fiber, in particular, appear to be protective. Meats and foods of animal origin increase the risk. Protein and fat consumption have been shown to be promoters of pancreatic carcinogenesis in experimental animals. [3] In Japan, two clinical studies showed a correlation between meat consumption and pancreatic cancer. [4] Another study in the United States found a 2.5-fold increased risk of developing pancreatic cancer in men consuming protein, largely in the form of meat. [17]

Obesity

Prospective cohort studies including 46,648 men (ages 40–75) and 117,041 women (ages 30–55) showed that a body mass index (BMI) of at least 30 kg/m ^{2} was associated with an increased risk for pancreatic cancer. [18] In addition, total physical activity was shown to be inversely related to pancreatic cancer risk for those with a BMI of 25 or greater. [18] In the same study height was also found to be associated with increased risk. The increased risk associated with obesity is thought to be related to abnormal glucose metabolism, insulin resistance, and hyperinsulinemia commonly seen in obese individuals.

Genetic Factors

It is estimated that familial factors and genetic susceptibility play significant roles in the development of around 10% of pancreatic cancer cases. A recent review of the National Familial Pancreas Tumor Registry (NFPTR) demonstrated an 18-fold increase in the risk for malignancy among first-degree relatives of patients with familial pancreatic cancer compared to those with relatives with sporadic disease. [19] For those with three or more family members with familial disease, the risk increased a remarkable 57-fold. [19] Thus, first-degree relatives of patients with familial pancreatic cancer are at significant risk and may benefit from early screening methods.

MOLECULAR GENETICS

In recent years, many exciting advances have been made in the understanding of pancreatic cancer at its genetic level. Ductal adenocarcinomas of the pancreas appear to evolve from normal ductal epithelium that develops into benign, flat, mucinous duct lesions, a transition characterized by transformation of the normal, low-cuboidal ductal epithelium to a columnar morphology. [20] Upon microdissection, about half of these flat ductal lesions are found to contain *K-ras* mutations. [20] These lesions progress to papillary lesions without atypia, to papillary lesions with atypia, and finally to invasive adenocarcinoma. [20] The available evidence suggests that the cells that progress through this sequence have been genetically altered in ways that favor unrestrained growth. Such alterations can occur as mutations in oncogenes, which normally code for proteins that are critically involved in cellular transcription and growth, or in tumor suppressor genes, whose protein product functions to suppress cellular proliferation. Oncogene mutations—through point mutation, amplification, or chromosomal translocation—confer upon the pancreatic cancer cell a “gain in function,” usually manifested as a constitutively active and unrestrained growth-promoting protein. Mutations in tumor suppressor genes, on the other hand, usually result in “loss of function” for the transformed pancreatic ductal cell, as the altered or deleted protein cannot effectively control cellular growth and suppress tumor formation. Mutations in oncogenes and tumor suppressor genes are encountered in pancreatic ductal adenocarcinoma.

Genetic alterations associated with pancreatic cancer can be divided into two groups, those that occur with a high frequency or a low frequency.

High Frequency Alterations

Four genetic mutations represent the most frequent alterations in established pancreatic cancers:

- activation of the *K-ras* oncogene in 80% to 95% of cases
- inactivation of the tumor suppressor genes *p16* (80% to 90%)
- *p53* (75%)
- *DPC4* (50%). [20], [21], [22], [23], [24] and [25]

The *K-ras* mutations, which occur most commonly in codon 12, also have been found in the majority of noninvasive proliferating ductal lesions before cancer is seen. This suggests that the genetic alterations responsible for ductal cell malignant transformation occur at an early stage of carcinogenesis. [21]

K-ras is a member of the membrane-associated, guanine nucleotide-binding, protein family involved in signal transduction stimulated by growth-promoting effectors at the cell surface. [21] When *K-ras* is mutated, usually through point mutation, it is transformed into an oncogene whose abnormal protein product gains the capacity for unrestrained mitogenic signal transduction. Evidence indicates that *K-ras* mutations occur more frequently in pancreatic carcinomas than in other variants of periampullary carcinoma, suggesting a molecular difference may underlie their distinct tumor biology and clinical behavior. [21], [24] It is clinically significant that the *K-ras* mutation has been discovered in many benign human neoplasms and early lesions, indicating that it is not specific for invasive malignancy. This fact has hindered its development as a reliable screening tool for pancreatic ductal adenocarcinoma. [20]

p16, a tumor suppressor gene located on chromosome 9p, encodes a protein that serves as a cyclin-dependent kinase (CDK) inhibitor. Its interactions with cyclin-complexes effectively arrest cells in the G1 phase. [20], [21] Mutations in *p16*– deletions, missense, nonsense, frameshift, and hypermethy- lations—lead to loss of its restrictive action on cell growth. A germline mutation in *p16* associated with familial melanoma also increases risk of developing pancreatic carcinoma. [21]

p53 is a tumor suppressor gene located at chromosome 17p which encodes a 53 kd protein that negatively regulates cell growth and proliferation. This nuclear-binding protein regulates the transcription of factors important in cellular processes such as cell-cycle modulation, DNA surveillance and repair, differentiation, and apoptosis. [21] Nearly all mutated *p53* protein products lose the ability to bind DNA. [20] *p53* expression has been found to be induced by hypoxia, a

condition especially profound in pancreatic carcinomas. [22]

Cloned by Hahn and colleagues in 1994, *DPC4* (for *D*eleted in *P*ancreatic *C*ancer, Locus *4*) is a tumor suppressor gene related to the *Smad* gene family. [20] Its protein product is a cytoplasmic protein which, in response to transforming growth factor-ß (TGF-ß) binding to its receptor TGF-ßR $_2$, translocates to the nucleus, binds the *Smad*-binding element of DNA, and regulates transcription. [20] Germline mutations in *DPC4* have been shown to be associated with juvenile polyposis. [20] Interestingly, recent immunohistochemical analysis of 79 resected pancreatic intraductal papillary mucinous tumors (IPMTs) demonstrated uniform (100%) expression of the *DPC4* product, compared to only 70% and 45% expression in high-grade pancreatic intraepithelial neoplasms and infiltrating adenocarcinomas, respectively. [26] Thus, the *DPC4* deletion is thought to occur as a relatively late event in the process of ductal cell transformation; the presence of its protein product in IPMTs may contribute, in part, to the more favorable prognosis and more indolent course of these tumors.

Low Frequency Alterations

These less-frequent mutations comprise alterations in oncogenes, tumor suppressor genes, and growth factors and their receptors. The signal transducer AKT2 is a target oncogene thought to be involved in the amplification of chromosome 19q13, a change found in 10% to 20% of pancreatic cancers. [20] The MYB oncogene has been observed in 10% of cases of pancreatic cancer. [20]

The *BRCA2* tumor suppressor gene has been found to be inactivated in about 7% of pancreatic cancers. [21], [23] This mutation is especially significant in that it is a germline mutation which represents, as yet, the most common inherited predisposition to pancreatic carcinoma. [21] Patients with PJ syndrome, characterized by hamartomatous intestinal polyposis, have an increased risk of pancreatic and other gastrointestinal cancers. [21], [23] This syndrome is associated with mutation of the *LKB1/STK11* gene, a tumor suppressor gene which encodes a serine/threonine kinase. [20] About 5% of pancreatic cancers unrelated to the PJ syndrome contain this mutation. [20]

In addition to these gene mutations, there is also evidence that various growth factor receptors (e.g., epidermal growth factor [EGF] receptor, c-erbB2, c-erbB3, TGF-ß receptor 1–3), growth factors (e.g., EGF, TGF-a, TGF-ß $_{1-3}$, fibroblast growth factor [aFGF], bFGF], and intercellular and endothelial-leukocyte adhesion molecules (e.g., intercellular adhesion molecule-1, endothelial leukocyte adhesion molecule-1) are up-regulated in human pancreatic adenocarcinomas. [21] These changes may stimulate further tumor growth and enhance the metastatic behavior of pancreatic cancer cells, thus possibly contributing to shorter postoperative survival following tumor resection. [21], [24] Insulin-like growth factor-1 has been shown to enhance the growth of human pancreatic cancer cells. [27] Expression of the fibroblast growth factor receptor has been found to correlate with metastases and tumor stage. [21], [28] Finally, patients with familial recurrent pancreatitis have been found to express a mutated serine protease gene, *PRSS1,* also known as the *cationic trypsinogen gene*. A carrier of this mutation has an overall lifetime risk of developing pancreatic cancer of 40%, usually at an earlier age (average age of onset 39 years). [20] This increased cancer risk is thought to be related to the mitogenic response of ductal cells to multiple episodes of inflammatory damage. [20], [23]

PATHOLOGY

Ductal adenocarcinoma and its variants make up more than 90% of all malignant exocrine pancreatic tumors. Of the remainder, some are acinar cell carcinomas, some are sarcomas, a few are malignant lymphomas, and some are of uncertain histogenesis.

About two thirds of ductal adenocarcinomas occur in the head of the gland; the rest occur in the body or tail, or diffusely throughout the pancreas. They are characterized by an intense desmoplastic reaction in which ductlike structures of varying degrees of differentiation are seen. Because of their proximity to the intrapancreatic portion of the common bile duct, tumors in the head usually produce jaundice since they compress and obstruct the bile duct as they grow. They often obstruct the pancreatic duct as well, and although steatorrhea may result, there may not be any obvious symptoms. Tumors of the head of the pancreas are usually at least 2 cm in diameter when they are first diagnosed. Most tumors that are resected have a median diameter of 2.5 to 3.5 cm. Tumors of the body and tail commonly are larger (5–7 cm), and more advanced when they are discovered because they do not produce symptoms as early as head tumors do. The symptoms from tumors in the body and tail are usually caused by malignant infiltration of the retroperitoneal structures and nerves, which produces pain. By the time the diagnosis is made, almost all are unresectable. Nevertheless, there is evidence that resectable body tumors have a similar prognosis to resectable tumors in the head of the gland.

Approximately 70% to 80% of adenocarcinomas of the head of the pancreas have metastasized to regional lymph nodes by the time they are discovered, which worsens the prognosis but does not preclude cure. These tumors also commonly invade lymphatic channels and perineural spaces. The prognosis is also influenced by the degree of tumor differentiation and by the presence of invasion of the retroperitoneal tissues adjacent to the cancer. The best outcome is seen in patients who have well-differentiated neoplasms, without retroperitoneal invasion or lymph node metastases. Distant metastases (e.g., lung) may occur, but pancreatic cancer typically infiltrates locally into the adjacent structures (e.g., stomach, duodenum, colon, transverse mesocolon, portal and superior mesenteric veins, superior mesenteric artery). The liver is the most common site of intra-abdominal metastasis, and peritoneal seeding of the tumor is also seen. In patients without distant spread, vascular invasion by tumor is the most common reason for unresectability.

The most widely accepted staging system for pancreatic cancer is the Union Internationale Contre le Cancer (UICC) System, which is shown in Table 96-1. Since pancreatic cancer has usually spread to lymph nodes by the time the diagnosis is made, the majority of patients have at least stage III disease. Patients with stage IV disease (distant metastases) cannot be cured and are considered unresectable.

TNM CLASSIFICATION	CHARACTERISTICS
T1	Limited to the pancreas
T2	Direct extension to duodenum, bile duct, or peripancreatic tissues
T3	Direct extension to stomach, spleen, colon, adjacent large vessels
N0	Regional lymph nodes not involved
N1	Regional lymph nodes involved
M0	No distant metastasis
M1	Distant metastasis present
TNM STAGING SYSTEM	**CHARACTERISTICS**
Stage I	T1-2, N0, M0
Stage II	T3, N0, M0
Stage III	T1-3, N1, M0
Stage IV	T1-3, N0-1, M1

TNM, tumor nodes, metastasis.

TABLE 96-1 Staging of Carcinoma of the Pancreas

CLINICAL MANIFESTATIONS

The most common symptoms in patients with cancers in the head of the pancreas are jaundice and abdominal pain, usually with some degree of weight loss. In those with tumors of the pancreatic body, jaundice is unusual. Abdominal pain and back pain are common, and weight loss may be profound.

Pain

The cause of the pain associated with cancer in the head of the pancreas may be gallbladder and bile duct distension associated with biliary obstruction, pancreatic duct distension associated with pancreatic duct obstruction, and invasion of retroperitoneal or somatic nerves. Abdominal or back pain may occur. Abdominal pain is most commonly felt in the epigastrium (46%), but it also may be felt on either side or in the lower quadrants. Persistent severe back pain is likely to reflect unresectability, and is caused by tumor invasion of the retroperitoneal nerves.

Jaundice

Jaundice, accompanied by pain, is the presenting symptom in 80% to 90% of patients with cancer of the head of the pancreas. Painless jaundice is uncommon. Jaundice in the absence of any other symptoms is associated with a better prognosis, perhaps because the tumor is strategically located near the bile duct where, even at a small size, it has obstructed the duct early in its development.

Weight Loss

By the time the diagnosis is made, weight loss of more than 10% ideal body weight is common. There are several possible explanations for such weight loss. The pain associated with the tumor produces anorexia in almost all patients, so decreased food intake is the most common cause. Malabsorption also may occur, especially in patients with pancreatic duct obstruction in the head of the pancreas, as digestive enzyme output from the pancreas may be prevented from entering the duodenum. It is less likely in those with pancreatic duct obstruction from body tumors, since enzymes from the pancreatic head drain normally, and are enough to maintain normal fat and protein absorption. Finally, some workers hypothesize the existence of a humoral substance secreted by the tumor that increases catabolic activity, but it has yet to be characterized. [29]

Diabetes Mellitus

Diabetes sometimes appears as an early manifestation of pancreatic cancer, occurring many months before the tumor becomes evident. It has been postulated that the tumor may release a substance, as yet uncharacterized, that stimulates the secretion of islet amyloid polypeptide (IAPP) from the beta cells of the uninvolved islets. IAPP causes insulin resistance. In some patients with pancreatic cancer, diabetes, and elevated serum IAPP, the diabetes and high IAPP levels disappeared once the tumor was resected. [30] A case-controlled study showed no increase in the prevalence of long-standing diabetes mellitus among 116 patients with pancreatic cancer, 24% of whom had diabetes at the time of cancer diagnosis. [31] Thus, diabetes may be a consequence of cancer development rather than a risk factor for pancreatic cancer.

Other Clinical Features

Light-colored stools and dark urine are characteristic of obstructive jaundice. Other symptoms include pruritus, nausea, vomiting, and weakness. Vomiting may be caused by duodenal or gastric outlet obstruction caused by tumor invasion. In some cases, when there is no apparent obstruction, it may be caused by abnormal gastric motility for as yet obscure reasons. Less common findings include superficial thrombophlebitis and gastrointestinal bleeding. In advanced stages, gastrointestinal bleeding may occur from direct invasion of the tumor into the duodenum, stomach, or colon.

PHYSICAL FINDINGS

Physical examination may reveal hepatomegaly, a palpable abdominal mass, or ascites. In the past, a distended palpable gallbladder (Courvoisier sign) was thought to be pathognomonic for bile duct obstruction caused by a tumor. When gallstones caused the bile duct obstruction, gallbladder distention was thought to be less likely because chronic inflammation of the gallbladder wall caused scarring and contracture of the viscus. However, the gallbladder is palpable only in about one half of jaundiced patients with pancreatic cancer, so Courvoisier sign is of little practical diagnostic value. [32] In those patients with pruritus, persistent scratching produces excoriation and lichenification of the skin. Rarely, an abdominal bruit can be heard if the tumor compresses the aorta or splenic artery.

DIAGNOSTIC INVESTIGATIONS

Routine Clinical Tests

In patients with biliary obstruction, the total serum bilirubin and alkaline phosphatase concentrations are elevated. The total bilirubin level tends to be greater with malignant obstruction than obstruction from benign causes (>15 vs. 5 mg/dL). Serum amylase elevation is seen in only 5% of patients with pancreatic cancer.

Immunologic Tests: Tumor-Associated Antigens

A variety of tumor-associated antigens have been studied with the hope that one or more would be specific for pancreatic cancer, and sensitive enough to allow for early diagnosis when the tumor was still small and curable. The most sensitive (80%) and specific (90%) is CA 19-9, but it still falls short of the ideal since it is almost never positive with small tumors (<1 cm). CA 19-9 values also may be abnormal with other cancers (gastric, colorectal cancer) and with some benign conditions (cholangitis). Determination of serum CA 19-9 levels may be useful to provide some assurance that the tumor has been resected in its entirety, to signal the presence of recurrent disease after resection, and to determine the response to adjuvant therapy. [33], [34]

RATIONALE OF THE WORKUP IN PATIENTS WITH SUSPECTED PANCREATIC CANCER

There are two main goals of the diagnostic workup in patients who are thought to have a pancreatic malignancy. The first is to establish the diagnosis with a high degree of certainty. In patients who are candidates for surgery, this usually does not require histological proof of malignancy. The second is to decide whether the patient should undergo an operation in an effort to either resect or palliate the disease. A number of tests are available to help with these decisions, but it is rare that all of them are required in every patient. The clinician should strive to make the workup as cost- and time-efficient as possible, arriving at a treatment plan promptly. [35]

Noninvasive Techniques

Ultrasound (US) may be useful as a first-line investigation in patients with jaundice to distinguish between intrahepatic and extrahepatic causes. Extrahepatic obstruction from a pancreatic (or any periampullary) cancer would be expected to show dilated intrahepatic and extrahepatic biliary radicles (Fig. 96-1). The ducts are not dilated with obstruction at the intrahepatic level. US can also identify gallstones that may be the cause of obstruction. However, abdominal wall musculature, bony structure, and overlying bowel gas can obscure transabdominal visualization of the pancreas through a sonographic window. Although US provides some information about the relationship of the tumor to surrounding structures, computed tomography is much better in this regard. US is a relatively inexpensive test, with a sensitivity of 70% and a specificity of more than 90% for the diagnosis of pancreatic cancer. [36] In up to 25% of patients, however, the examination is unsatisfactory owing to body habitus or overlying bowel gas.

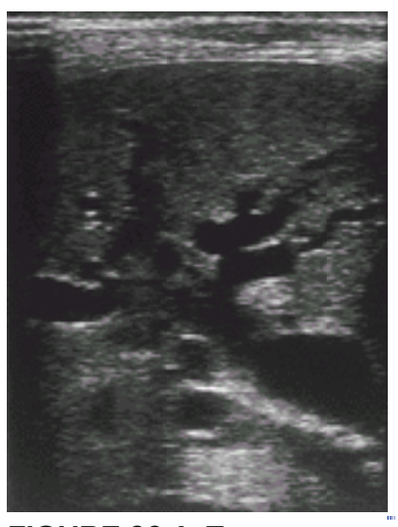

FIGURE 96-1. Transcutaneous ultrasound in a patient with common bile duct obstructed by a tumor in the head of the pancreas. The intra- and extrahepatic bile ducts

are dilated.

Currently, helical computed tomography (CT) is the best overall study for diagnosis, as well as the preoperative staging of pancreatic cancer. It provides information about the nature and site of the lesion (e.g., pancreatic vs. other periampullary tumors, hilar bile duct tumors), its resectability (e.g., presence of hepatic metastases, vascular invasion), and vascular anatomy. It is important to stress that the helical CT is the state-of-the-art procedure. When the helical CT is compared with the conventional technique, and especially when imaging of both the pancreas and liver is done separately according to published protocols, the differences in the sensitivity and specificity of the two approaches is dramatic. [37] The primary lesion itself, as well as liver metastases, may be evident with the helical CT, and these may not be seen with the standard scan (Fig. 96-2). Extraordinary detail is provided about vascular anatomy and vascular invasion by tumor (Fig. 96-3). If an experienced radiologist indicates that the tumor is unequivocally “unresectable” because of major vascular invasion, this is about 98% reliable. If the tumor is judged to be “resectable” on the basis of helical CT, this prediction is accurate in about 80% of cases. [35], [38]

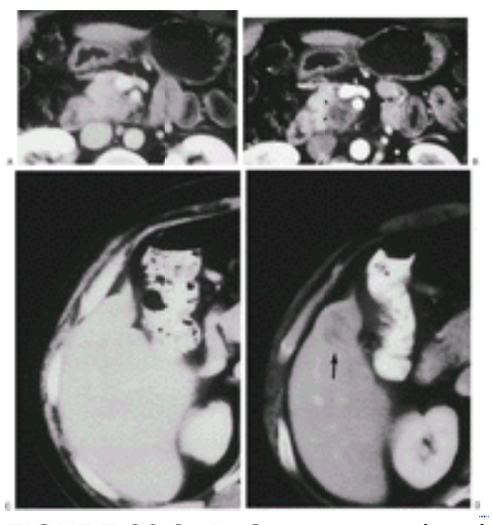

FIGURE 96-2. A: On a conventional computed tomography (CT) scan, the primary tumor in the uncinate process of the pancreas is not seen. **B:** The helical (spiral) CT scan performed 4 days later reveals the tumor (*arrowheads*). **C:** On a conventional CT scan, no liver metastases are seen. **D:** The helical CT performed 11 days later reveals an obvious metastatic lesion (*arrow*). (Courtesy of David Lu, M.D., UCLA Department of Radiology, Los Angeles.)

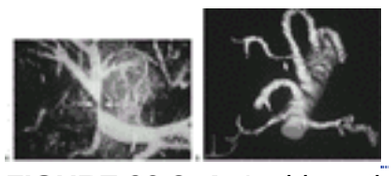

FIGURE 96-3. A: In this patient with unresectable pancreatic cancer, the helical computed tomography (CT) scan shows that the tumor has compressed the portal vein (*arrows*). **B:** In another similar patient, the tumor is compressing the hepatic artery at its origin from the celiac axis (*arrow*), which indicates that the tumor is unresectable. In this case, computer reconstruction of the CT images was performed to demonstrate the detailed anatomy of the aorta and its branches. (Courtesy of David Lu, M.D., UCLA Department of Radiology, Los Angeles.)

CT scan is able to detect tumors about 2 cm or greater in diameter, which appear as lucent areas, because they are less well perfused with blood than the adjacent pancreatic tissue. The technique has a false-positive rate of less than 10%, which is usually caused by focal pancreatitis, or variations in normal pancreatic anatomy that are mistaken for tumor.

Magnetic resonance imaging (MRI) may provide slightly better pancreatic tissue contrast than CT, but spatial resolution is inferior. It was hoped that MRI would be superior to CT in outlining the blood vessels, but such an advantage has not been demonstrated. This may change as better contrast agents are developed. In a recent study, MRI was shown to predict resectability with an accuracy of 77%. [39]

Endoscopic retrograde cholangiopancreatography (ERCP) has a sensitivity of 95% and a specificity of 85% for the diagnosis of pancreatic cancer. [40] The procedure can be performed successfully in over 90% of patients and it detects some tumors not seen on CT. The classic finding that suggests pancreatic cancer is obstruction of both the bile and pancreatic ducts in the head of the pancreas, the so-called double-duct sign (Fig. 96-4). In tumors of the body or tail of the pancreas, only the pancreatic duct is abnormal (Fig. 96-5). At the time of ERCP, brushings for cytology may be obtained from the pancreatic duct, which has a sensitivity of about 60% in proving the diagnosis of malignancy. Some have argued that ERCP should be done routinely in these patients, since it may be possible to determine the origin of the tumor (e.g., ampulla of Vater instead of the pancreas). Although this may be true in some cases, it usually does not affect subsequent management since the operation that is required is the same for all of the periampullary malignancies. At the time that the ERCP is done, a stent also can be placed into the obstructed common bile duct, which has an important role in the palliation of patients who do not require surgery. Stents, however, should not be used routinely for preoperative decompression (see section “ Nonsurgical Palliation of Biliary and Duodenal Obstruction ”).

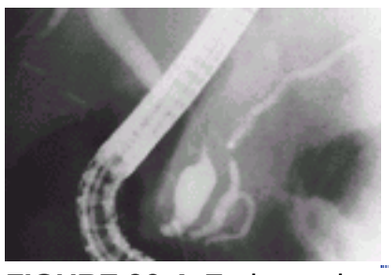

FIGURE 96-4. Endoscopic retrograde cholangiopancreatography (ERCP) of a patient with cancer of the head of the pancreas shows that both the pancreatic and bile ducts are compressed by the tumor. This produces the “double-duct sign.”

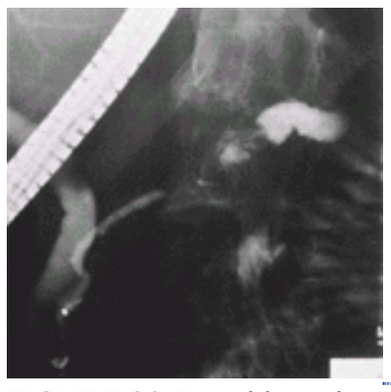

FIGURE 96-5. In this patient with cancer of the body of the pancreas, the duct is compressed in the body of the gland, and the obstructed ductal segment proximal to that point is dilated.

In spite of its value, ERCP is not required in all patients with pancreatic cancer. If a patient has a history typical for pancreatic cancer (e.g., pain, jaundice, weight loss) with a mass in the head of the pancreas evident on CT scan, then an ERCP is unnecessary for the diagnosis. An ERCP at this point generally adds nothing to the workup that is of value to either the patient or the surgeon. If the CT scan does not show a mass or raises questions about the diagnosis, ERCP should be done. If ERCP is done first, however, all patients still require a CT scan.

Endoscopic ultrasound (EUS) is not likely to be used as a first-line diagnostic study in most patients with pancreatic cancer. Once the diagnosis is suspected, however, it can provide valuable information that can aid in tumor staging and determination of resectability. With surgical pathology as the gold standard, recent studies have shown that EUS accuracy in tumor staging and estimation of lymph node involvement ranges from 69% to 85% and 54% to 75%, respectively. [39], [41], [42] Lymph nodes infiltrated by carcinoma usually appear as round, hypoechoic masses more than 1 cm in diameter. EUS can provide information about whether the tumor invades the portal, splenic, or superior mesenteric veins, the celiac axis, or superior mesenteric arteries. Common EUS criteria for vascular involvement include abnormal vessel contour, loss of vessel-parenchymal interface, tumor visible within the vessel lumen, and the presence of dilated peripancreatic venous collaterals. [43] When results are compared to surgical findings, EUS accuracy ranges from 69% to 93%, [42], [43], [44] and [45] and its ability to predict tumor resectability ranges from 72% to 93%. [42], [45]

EUS can detect tumors smaller than those currently detectable by CT scanning techniques (<2.0 cm diameter), with sensitivity and specificity for tumor detection as high as 99% and 100%, respectively. [43] The technique also permits accurate placement of biopsy needles (e.g., EUS-guided fine-needle aspiration [FNA] cytology) into a pancreatic mass when there is need to establish the diagnosis (Fig. 96-6). EUS-FNA has also been used to sample nearby lymph nodes to aid in local staging, with a low complication rate of 1% to 2%. [43]

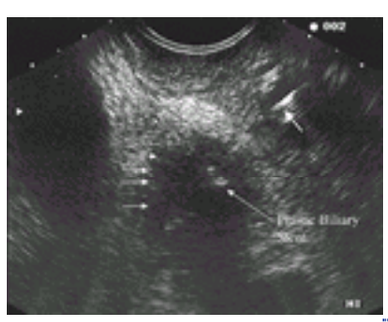

FIGURE 96-6. Endoscopic ultrasound and fine-needle aspiration (FNA) of a 1.5-cm localized hypoechoic mass in the head of the pancreas. The FNA needle is seen in the top right hand corner (*solid arrow*). A biliary stent is seen in the center of this mass, which was not visualized on helical computed tomography scan of the pancreas. Cytologic analysis of the FNA revealed adenocarcinoma. (Courtesy of James J. Farrell, M.D., UCLA Department of Medicine, Division of Digestive Diseases, Los Angeles.)

The eventual role of EUS in the workup of these patients continues to evolve. Limitations of EUS and its applications include the considerable expertise required to perform it (still not widely available), interobserver variability in scan interpretation, inability to examine distant organs for metastasis, and a difficulty in distinguishing focal pancreatitis from a pancreatic mass. For the patient who presents with symptoms and signs suggestive of pancreatic carcinoma (jaundice, abdominal discomfort, weight loss), helical CT scan remains the best initial study for establishing the diagnosis, evaluating extent of disease, and determining resectability, making EUS unnecessary. In a patient with a negative CT scan but a history that suggests pancreatic cancer, EUS is a useful adjunctive examination given its increased sensitivity for detecting small tumors. Likewise, EUS may be useful to estimate resectability of the tumor if the CT scan does not resolve that issue, and to obtain tumor tissue to confirm the diagnosis if that seems appropriate.

Invasive Techniques

Percutaneous transhepatic cholangiography (PTC) is useful in patients with proximal bile duct obstruction (e.g., from a tumor at the hepatic duct bifurcation) to delineate the bile duct anatomy on the hepatic side of the obstruction. The intrahepatic ducts are opacified, which defines the degree of their involvement, an important determinant of resectability. In patients with periampullary tumors, ERCP is preferred to PTC if the bile duct anatomy needs to be defined.

Preoperative fine-needle aspiration (FNA) for cytology can be obtained percutaneously using a fine-gauge needle under CT or US (or EUS) guidance. Characteristic signs of malignancy are single or irregularly arranged clusters of cells exhibiting pleomorphism, large vesicular nuclei, and prominent nucleoli (Fig. 96-7). Complications include hemorrhage, pancreatitis, pancreatic fistula, and seeding of the needle tract with cancer cells, all of which are uncommon. FNA should not be done routinely in the workup of patients with suspected pancreatic cancer. It is uncomfortable for the patient, it is associated with considerable cost, and in patients who are operative candidates, it does not change management. FNA is indicated in situations where the cytologic proof of malignancy will alter management. For example, in a patient with an unresectable tumor in the body of the pancreas who has no symptoms for which surgical palliation is required, FNA could confirm the diagnosis, and chemotherapy, radiation, or both could be given. In a patient with obstructive jaundice and a mass in the head of the pancreas, who may not be a candidate for resection because of coexisting medical problems, FNA would be useful to confirm the diagnosis. Then, a stent could be placed to palliate the jaundice, and surgery, simply to obtain tissue for diagnosis, would have been avoided. *It is important to stress that a negative FNA never rules out the possibility of malignancy.*

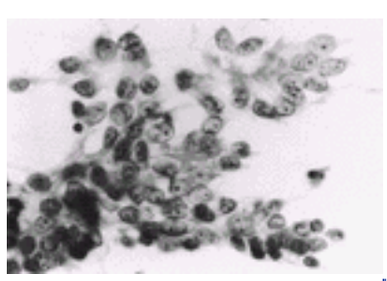

FIGURE 96-7. Fine-needle aspiration of a mass in the body of the pancreas shows cells characteristic of ductal adenocarcinoma of the pancreas. The tumor invaded the retroperitoneal structures and was unresectable.

The argument that preoperative knowledge of the diagnosis of cancer simplifies or speeds up the operation may be valid if the surgeon is inexperienced. However, most pancreatic surgeons do not require histological proof of malignancy before proceeding with resection. Resection is performed on the basis of the patient's history, the information from preoperative tests, and the gross findings at operation. Of course, this issue must always be discussed preoperatively with the patient. With this approach, experienced surgeons err less than 10% of the time (i.e., a resection is performed for what turns out to be benign disease). This is acceptable since the mortality rate of resection is quite low (1%–2%), and the majority of patients who undergo resection for benign disease have chronic pancreatitis for which resection is also appropriate.

Angiography, which was performed more commonly in the past, is unnecessary. Helical CT scans provide similar information about the presence of vascular anomalies, as well as displacement or constriction of major peripancreatic vessels (e.g., celiac or hepatic arteries, portal or superior mesenteric veins), which suggests unresectability.

Chronic Pancreatitis

Chronic pancreatitis is an independent risk factor for pancreatic cancer (see previous discussion), and the two diseases may share clinical (pain, weight loss, endocrine/exocrine dysfunction) and imaging characteristics (ductal abnormalities, mass, or cystic lesions). [46], [47], [48], [49] and [50] Because pancreatic cancers usually cause some degree of pancreatic duct obstruction, most patients with pancreatic cancer also have areas of chronic pancreatitis adjacent to the tumor. And pancreatic cancers themselves grow in a desmoplastic fashion that grossly may be indistinguishable from chronic pancreatitis without associated malignancy. Because there is no serologic or imaging study that can reliably distinguish chronic pancreatitis from pancreatic carcinoma, and a previous diagnosis of chronic pancreatitis can delay treatment of resectable pancreatic carcinoma, [46], [47] the clinician must maintain a high index of suspicion in patients known to have the benign disease. For example, any patient with a previously established diagnosis of chronic pancreatitis who experiences a significant alteration in symptomatology—accelerated weight loss or increasing pain—should be evaluated thoroughly for the presence of malignant disease.

TREATMENT

Surgery

Preoperative Management As in all patients about to undergo major surgery, it is important to optimize cardiac, pulmonary, and renal function preoperatively. This usually can be done in the outpatient setting. Although these patients have often lost weight, most are still adequately nourished to safely undergo the operation. However, if the serum albumin is less than 3 g/dL, or if surgery must be delayed for more than several weeks after the diagnosis is established, supplemental nutrition is often appropriate. Pancreatic enzyme replacement may also help. Nutrition can usually be provided enterally, but occasionally, such as with gastric outlet obstruction, parenteral alimentation is indicated. Because obstructive jaundice can cause defects in hepatic, renal, and immune function, it was hoped that preoperative relief of the jaundice would correct these defects, and decrease postoperative morbidity and mortality rates. Unfortunately, several randomized controlled studies have proven that routine preoperative bile duct stenting does not help. [51], [52], [53] and [54] However, if operation must be delayed several weeks or more, for any reason, or if cholangitis occurs, a plastic biliary stent (at least 10 French, if possible) should be placed endoscopically in jaundiced patients. *Metal Wallstents* (Boston Scientific, Boston) *should never be placed in patients who are candidates for resection.* These stents incite a severe inflammatory reaction and eventually may be incorporated into the bile duct wall. This can complicate or even prevent the resection.
Operative Management: Resection The first successful resection for a periampullary cancer was performed as a two-stage operation by the German surgeon Kausch in 1909. In 1935, Whipple performed a similar procedure for an ampullary carcinoma, which he had perfected to a one-stage resection by 1942. That operation has been called a pancreaticoduodenectomy, or Whipple resection, and it is similar to the one performed today (Fig. 96-8). It involves a partial gastrectomy (antrectomy), cholecystectomy, and removal of the distal common bile duct, head of the pancreas, duodenum, proximal jejunum, and regional lymph nodes. Reconstruction requires a pancreaticojejunostomy, hepaticojejunostomy, and a gastrojejunostomy. In the hands of experienced pancreatic surgeons around the world, the operative mortality rate of this procedure is now 2% or less.

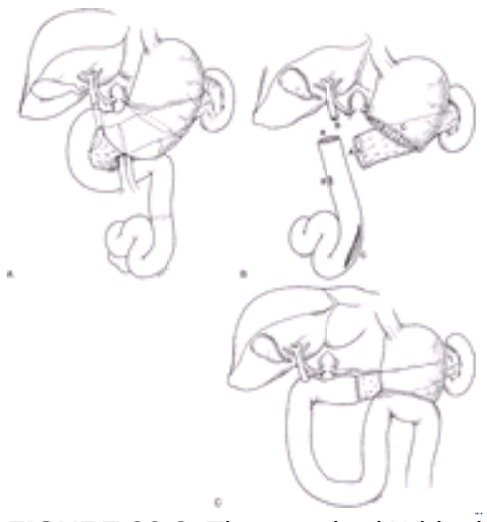

FIGURE 96-8. The standard Whipple pancreaticoduodenectomy. **A:** *Dashed lines* indicate the resection margins in the typical operation for a tumor in the head of the pancreas. **B:** The specimen has been resected. *A, B,* and *C* represent the sites for subsequent anastomoses between the bowel and the pancreas, bile duct, and stomach. **C:** The completed anastomoses are shown.

Resectable tumors in the body or tail of the pancreas require a distal pancreatectomy and splenectomy. The head of the pancreas, gallbladder, bile duct, stomach, and duodenum can usually be preserved since the tumor and its lymphatic drainage do not involve these structures. Mortality and morbidity rates are lower than after a Whipple resection.

Modifications of the Pancreaticoduodenectomy

Pylorus Preserving Whipple Resection Because of concern that the standard Whipple resection was associated with excessive weight loss and nutritional disturbances, many surgeons adopted this modification in which the stomach, pylorus, and first 3 to 4 cm of duodenum are preserved (Fig. 96-9). This maintains gastric reservoir function, and postoperative gastric emptying is closer to normal. The operation was first described in 1944, but it did not become popular until the 1970s when it was described for patients with chronic pancreatitis; later it became popular for pancreatic cancer as well. [55], [56] This less extensive procedure does not appear to compromise the chance for cure of the cancer. However, the few studies that compared it with the standard Whipple in regard to the amount of weight loss, various nutritional parameters, and overall quality of life associated with each operation, yielded conflicting results. Many surgeons perform the two procedures interchangeably, often reserving the standard Whipple for patients with larger, more extensive tumors.

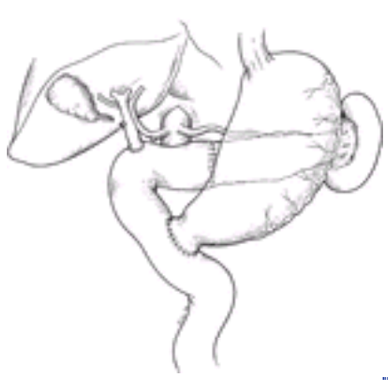

FIGURE 96-9. The pylorus preserving modification of the standard Whipple pancreaticoduodenectomy is shown. The entire stomach, the pylorus, and several

centimeters of duodenum are retained.

Radical (Extended) Whipple Resection Some Japanese surgeons have advocated the use of a more radical pancreatic resection to improve cure rates. [57] The standard Whipple is modified by removal of more peripancreatic soft tissue and lymph nodes, often with resection of segments of the superior mesenteric and portal veins, when they appear to be involved by tumor. Although some of these studies have reported improved survival, there have been no properly designed, prospective trials that compare this operation with the standard resection. Most surgeons in the United States are skeptical that the radical operation cures more patients, and it is rarely done in this country.

Determination of Resectability at the Time of Operation

Although the majority of patients who undergo resection for pancreatic cancer are not cured by the operation, it is performed only in circumstances where cure appears to be possible. Thus, resection is not done in the presence of liver or peritoneal metastases, or metastases to lymph nodes, which are not normally removed as part of the Whipple operation. The operation is begun by carefully examining the peritoneal cavity and its contents, and by obtaining biopsies of any areas suspicious for metastasis. Frozen-section diagnoses are usually available within 20 minutes, and a preliminary decision can be made about resectability. In the absence of distant metastases, resectability usually depends on whether the tumor has invaded any major blood vessels. Assessment of vascular involvement requires mobilization of the tumor from surrounding structures, which is done next. Involvement of the superior mesenteric, celiac, or hepatic arteries precludes resection. In most cases, so too does invasion of the superior mesenteric or portal vein. If the vessels appear to be free of tumor, the resection proceeds.

It is unusual for vascular involvement to be found at the time of operation if it was not already suspected on the basis of helical CT scan or EUS done preoperatively. It is more common (10%–15% of cases) to find small hepatic or peritoneal metastases, which were not evident from the preoperative studies. For this reason, some surgeons prefer to begin the operation with laparoscopy, which permits examination of the liver and peritoneal surfaces, and biopsy of any suspicious areas. If metastatic tumor is found, laparotomy may be avoided, unless gastric and biliary bypasses are required for palliation. Even these may be done laparoscopically, in some cases. The major drawbacks of laparoscopy are the additional time and expense required for the procedure and the inability to determine the presence of vascular invasion. The latter requires more extensive dissection and is aided by the tactile senses available only during laparotomy.

Laparoscopy is indicated whenever there is a high likelihood of unresectability that has not been confirmed preoperatively. Examples include some patients with pancreatic cancer and CT evidence of liver or other metastases, which have not been proven with FNA; patients with pancreatic body or tail cancers, all of whom have a very low chance of having a resectable lesion; and patients with pancreatic cancer and ascites, which is probably caused by unrecognized peritoneal metastases.

A number of other factors have been discussed in relation to their influence on resectability. Advanced age is not a contraindication. Each patient must be assessed individually and an evaluation made of the associated risks caused by coexistent cardiovascular, pulmonary, and renal disease. Many patients who undergo Whipple resection are octogenarians; some patients under age 65 may be unacceptable operative risks. Large tumor size is also not a contraindication to resection. Although small pancreatic tumors (<2 cm diameter) are more likely to be resectable than larger ones, no patient should be denied the opportunity for cure because the tumor is “too large.” Indeed, in several major centers in the United States, most of the Whipple resections for cancers in the head of the gland are done for tumors between 3 to 5 cm in diameter. [58]

Experienced pancreatic surgeons usually proceed with pancreaticoduodenectomy without a biopsy of the primary tumor to confirm the diagnosis of malignancy. Indeed, if the history and clinical picture, the preoperative tests, and the operative findings are all consistent with the diagnosis, then the chance that cancer is present is over 90%. In those cases where cancer is not the cause of the symptoms, the most likely diagnosis is chronic pancreatitis, for which pancreaticoduodenectomy is also appropriate. In part, this approach has evolved because it may be difficult to establish the diagnosis of pancreatic cancer on frozen-section biopsy material, and to distinguish it from the surrounding zone of chronic pancreatitis. This is especially true when the cancer is small and most likely to be cured by resection.

SURGICAL PALLIATION

During exploration, when resection of the primary tumor is not possible, the surgeon must decide whether any palliative procedures should be done. Because most patients with cancer in the head of the pancreas are jaundiced at the time of operation, a bypass to relieve the biliary obstruction is the most common procedure. An anastomosis between the gallbladder and jejunum (cholecystojejunostomy) or common bile duct and jejunum (choledochojejunostomy) are both effective bypass techniques [32] (Fig. 96-10). Jaundice is relieved in about 90% of patients. In about 10% of patients however the bilirubin does not return to normal because of impaired hepatic function. This may be caused by long-standing obstruction or extensive hepatic metastases. [32] If the gallbladder is used for decompression, the surgeon should be certain that the cystic duct enters the common bile duct distant from the tumor. If the junction of the cystic and common ducts is close to the tumor, the former may become obstructed as the cancer grows. Then the cholecystojejunostomy cannot function and the jaundice will recur. In patients who have had a biliary stent placed preoperatively, the stent should be removed and a cholecystojejunostomy should be done, if it is technically possible. This eliminates the need for stent changes during the remaining months of the patient’s life. If it is not possible, a choledochojejunostomy should only be done if the common duct is still dilated (e.g., >1 cm diameter), which makes the operation easier. Otherwise, the stent, which also provides effective palliation, can be left in place, and a surgical biliary bypass is not done.

FIGURE 96-10. A: Choledochojejunostomy. **B:** Cholecystojejunostomy. Each of these operations effectively relieves biliary obstruction by diverting the bile into the small intestine proximal to the tumor.

At the time of the initial exploration, it is unusual that patients have duodenal obstruction from the tumor, but 15% to 20% develop it sometime before they die. Thus, most surgeons perform a prophylactic gastrojejunostomy at the same time that a biliary bypass is done (Fig. 96-11). This is not associated with increased morbidity or mortality rates. [59] Occasionally, patients complain of vomiting preoperatively, and no objective evidence can be found for duodenal obstruction. In these patients, a gastrojejunostomy usually does not relieve the symptom, and the patient may still be unable to eat. The problem is probably caused by abnormal gastric motility, perhaps because of tumor infiltration of the nerve plexus. Although prokinetic agents may help, the symptom is often refractory to all forms of treatment.

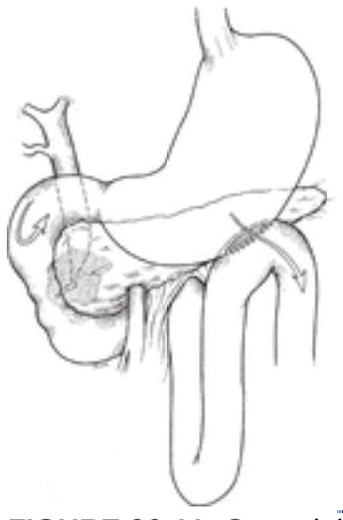

FIGURE 96-11. Gastrojejunostomy. The stomach can now empty into the small intestine directly when the tumor obstructs the duodenum.

NONSURGICAL PALLIATION OF BILIARY AND DUODENAL OBSTRUCTION

The development of techniques for endoscopic stenting of biliary obstruction represents a major advance in management, which allows many individuals with unresectable pancreatic cancer to avoid operation altogether. The advantages and problems associated with both surgical and endoscopic approaches have been defined in several prospective studies that compared the two. [60], [61], [62] and [63] In general, operative biliary bypass is more expensive, because of charges associated with the surgical procedure itself, and the hospitalization required for recovery. However, surgical bypass is more likely to provide permanent relief of the jaundice than are the endoscopically placed stents. The initial charges associated with endoscopic stenting are lower, but stents often become obstructed by bile pigment after 3 to 4 months and require replacement at intervals. Thus, the cumulative charges may exceed those of surgery, and there is added morbidity and inconvenience associated with recurrent jaundice and cholangitis, and multiple endoscopic procedures. Both approaches are equally effective, and have similar morbidity and mortality rates. As advances in both fields continue however, the advantages of each must be reassessed. For example, the metallic Wallstents developed are larger than the 10- to 12-French plastic stents in common use, and they are less likely to become obstructed. Laparoscopic techniques for both biliary bypass and gastrojejunostomy may decrease the cost and discomfort of the surgical approach, and permit more rapid hospital discharge. [64], [65] Duodenal obstruction is still usually treated surgically, but preliminary experience with duodenal stents suggests that this may be an option in selected patients. [66], [67] and [68]

MALABSORPTION

Some patients with unresectable pancreatic cancer experience fat malabsorption because the tumor obstructs the pancreatic duct, and insufficient amounts of pancreatic enzymes enter the duodenum. The result is diarrhea and steatorrhea, which interfere with the ability to gain weight and the quality of life. About 25% of patients who undergo pancreaticoduodenectomy also may suffer from malabsorption, which requires treatment. This is probably the result of poor mixing of the gastric chyme with the pancreatic enzymes that enter the gut at some distance from the gastrojejunostomy. Impaired stimulation of pancreatic secretion may also play a role.

Although fat absorption itself cannot be restored to normal, the goal should be to stop the diarrhea, restore adequate nutrition, and maintain body weight. The principles of treatment require that about 30,000 to 40,000 U of lipase be supplied with each of four to five meals spread throughout the day. A total of 100 g fat/day divided, equally among the meals, is usually tolerated. Patients who have undergone pancreatic resection almost always respond well to treatment; those with unresected cancer often do not. This is probably because of other poorly understood effects of the tumor for which medical management is still unsatisfactory.

PAIN

Conventional treatment of the pain associated with pancreatic cancer includes both opioid and nonopioid analgesics as well as nonsteroidal antiinflammatory drugs, tricyclic antidepressants, and antineoplastic agents. [69] However, pain from advanced pancreatic cancer is often difficult to control, and narcotics may be ineffective in providing relief. If this occurs, visceral pain may be managed by celiac plexus block, either at the time of exploratory surgery or later by a percutaneous approach. Pain is relieved in up to 90% of patients, and significant complications are uncommon. [70] Thoracoscopic pancreatic denervation for pain control in unresectable pancreatic cancer has also been reported. The morbidity was low with this minimally invasive approach. [71] For relief of somatic pain, an intercostal nerve block may be beneficial. [72]

ADJUVANT THERAPY

5-Fluorouracil (5-FU), combined with supervoltage radiation, confers a modest prolongation of survival in patients with locally advanced pancreatic cancer. This was demonstrated in 1969 in a study in which unresected patients receiving radiation alone had a median survival of 6.3 months, compared to 10.4 months for patients who received 5-FU plus radiation. The Gastrointestinal Tumor Study Group (GITSG) confirmed these observations in 1981. The median survival of patients receiving radiation alone was 4.5 months; in those who received both 5-FU and radiation, it was about 10 months. The GITSG also demonstrated a survival advantage (10.9 vs. 21 months) for patients who were treated with this same combination following curative resection of pancreatic cancer. [73] The treatment regimen was a split course of 40 Gy, with a 2-week separation between each 20 Gy segment. [74] 5-FU, 500 mg/m ^{2} of body surface area, was given as a bolus dose daily for the first 3 days of each radiation cycle and weekly thereafter for 2 years. [74] While more recent European studies (European Organization for Research and Treatment of Cancer [EORTC], European Study Group for Pancreatic Cancer [ESPAC]) confirmed the value of chemotherapy, they cast doubt on the value of radiation. [75], [76]

Gemcitabine is being used increasingly in the management of pancreatic cancer. [77], [78] In several studies, where it was used as a single agent, it appeared to slightly increase survival in a small fraction of those treated, when compared to 5-FU alone. However, in about 25% of patients it reduced pain, increased weight, and improved the overall quality of life. The mechanism of action was unclear. [79]

PROGNOSIS

Surgical resection provides the only chance for cure, but the median survival after resection is only 18 to 20 months, and the overall 5-year survival is 15%. However, if the tumor is removed with clear surgical margins and no lymph node metastases are found, the 5-year survival may be as high as 25% to 30%.

Various prognostic indicators for survival after resection for pancreatic cancer have been evaluated. One of the strongest factors is negative lymph node status. In one center, a median survival of more than 4.5 years was seen in patients with negative nodes versus 11 months for patients with positive nodes. Unfortunately, even small tumors have metastasized to nodes in 75% to 80% of resected cases. Poorly differentiated tumors are associated with a worse prognosis than well-differentiated ones (10% vs. 50% 5-year survival). Patients with smaller tumors (<2.5 cm in diameter) tend to have a better prognosis than patients with larger tumors. Another important prognostic factor is whether the resection margins of the resection specimen are involved with tumor. For this reason, the pancreatic and bile duct margins are routinely evaluated during the operation. If tumor is present, more tissue is resected if possible, until negative margins are obtained.

Operative mortality rates for the Whipple resection are now 2%, or less, in major centers around the world. There is increasing evidence that the best outcomes (operative mortality rates and long-term survival figures) and lowest treatment costs in patients with pancreatic cancer are achieved at institutions with the most experience (>20 Whipple resections a year). This appears to be related not only to the surgical expertise, but to that of the numerous other ancillary services (e.g., nursing, radiology, gastrointestinal endoscopy) that all gain experience in the management of large numbers of these difficult-to-treat patients.

LESS COMMON EXOCRINE PANCREATIC TUMORS

Mucinous Cystadenoma and Cystadenocarcinoma

Mucinous cystic neoplasms are large (often >5 cm), bulky, uni- lobular, or multilobulated cysts containing mucin (Fig. 96-12). The cysts are lined with mucinous columnar epithelium that forms papillary projections, which may contain foci of dysplastic cells or invasive carcinoma. Radiographic studies reveal calcification within the cysts in about 10% of cases; calcification almost never occurs in pseudocysts. Because mucinous cystadenomas are premalignant, they should be resected. When the tumors occur in the body or tail of the pancreas, the appropriate treatment is distal pancreatectomy. When they occur in the head of the pancreas, a Whipple resection is required. These neoplasms comprise 1% to 2% of exocrine tumors and are more common in women (female-to-male ratio 6:1). The peak age of occurrence is 40 to 60 years. [80] They appear to have a better prognosis than ductal pancreatic cancers; the 5-year survival after a curative resection of a mucinous cystadenocarcinoma is at least 50%.

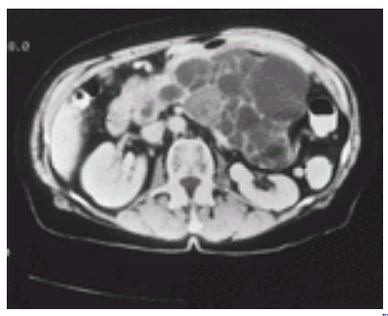

FIGURE 96-12. Computed tomography (CT) scan of a patient with a malignant mucinous cystic neoplasm, which was resected. Note the large cystic structure with internal septations creating multiple large cysts.

Serous Cystadenoma and Cystadenocarcinoma

These lesions are typically large, well-demarcated, multiloculated cystic tumors filled with watery fluid. They usually have a characteristic honeycomb appearance, and the individual cysts are 1 cm or less in diameter, which is smaller than the individual cysts in mucinous cystic neoplasms (Fig. 96-13). They account for about 1% of neoplastic pancreatic lesions and usually occur in older adult women in the sixth and seventh decades of life. These so-called microcystic adenomas are usually asymptomatic and are found incidentally when a US or CT scan is done for some other reason. One third of the tumors occur in the head of the pancreas. [81], [82] and [83] Although malignant transformation of serous lesions is uncommon, it does occur. These tumors also should be removed if removal can be done safely. These neoplasms, as well as the mucinous cystic tumors previously discussed, should not be confused with pseudocysts, where internal or external drainage procedures are acceptable forms of treatment.

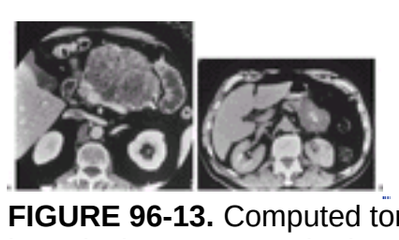

FIGURE 96-13. Computed tomography (CT) scans of patients with benign serous cystadenomas, both of which were resected with a distal pancreatectomy. **A:** This large lesion suggests the honeycombed appearance of this tumor, which typically contains many very small cystic structures. It is to be contrasted with the large cysts seen in Figure 96-12. **B:** This smaller lesion demonstrates the central calcification, which is sometimes seen.

Solid and Papillary Epithelial Neoplasm

Solid and papillary epithelial neoplasms appear to be increasing in frequency. They are most common in adolescent girls and young women, and have a characteristic appearance on CT scan (Fig. 96-14). The tumors are often large and may produce vague abdominal discomfort, which leads to their discovery. They have a favorable prognosis, and most patients are cured by resection. Nevertheless, some of these tumors may recur locally and there is one report of liver metastasis. The majority of these tumors occur in the body or tail of the pancreas, where they are treated with distal pancreatectomy. When they occur in the head, pancreaticoduodenectomy is required.

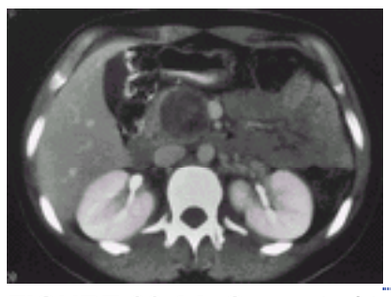

FIGURE 96-14. Computed tomography (CT) scan of a solid and papillary neoplasm in a 21-year-old woman who underwent a Whipple resection. The tumor was removed completely.

Intraductal Papillary Mucinous Tumors (IPMT, Mucinous Ductal Ectasia)

Patients with IPMT often present with repeated episodes of inflammation from underlying chronic pancreatitis. In fact, these patients are likely to present with abdominal pain as the principal complaint. The pancreatitis is caused by obstruction of the pancreatic duct by intraductal tumor growth and inspissated mucus, which these tumors secrete (Fig. 96-15). CT scans reveal a dilated pancreatic duct. ERCP confirms the ductal dilation and often shows intraductal mucus, which appears as filling defects. A glob of mucus emanating from the gaping orifice of the ampulla of Vater is characteristically seen at the time of ERCP (Fig. 96-16). Approximately half the lesions show papillary malignant changes, but even these tumors have a better prognosis than usual ductal adenocarcinoma. Treatment is required both to remove any premalignant or malignant disease that may be present and also to relieve the episodes of pancreatitis. This may require total pancreatectomy because the entire pancreatic duct may be affected.

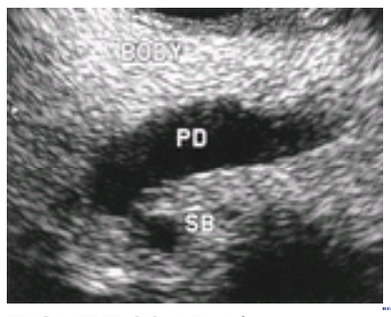

FIGURE 96-15. Linear array endoscopic ultrasound image of the pancreatic body showing dilated main pancreatic duct (PD) and dilated pancreatic ductal side branch (SB). Surgical resection confirmed main duct and side branch intraductal papillary mucinous tumor. (Courtesy of James J. Farrell, M.D., UCLA Department of Medicine, Division of Digestive Diseases, Los Angeles.)

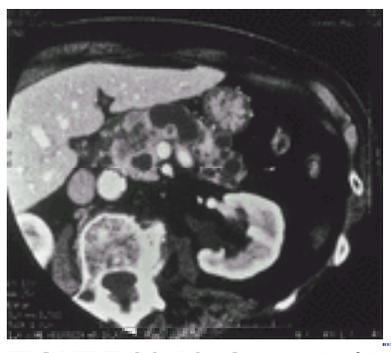

FIGURE 96-16. Computed tomography (CT) scan of a patient with mucinous ductal ectasia of the pancreas. In addition to multiple cystic lesions throughout the gland, there is a hypodense mass in the head of the pancreas consistent with a malignant neoplasm. The patient was 89 years old. Because a total pancreatectomy would have been required, surgery was not performed in this woman, who had multiple medical problems.

A recent review of a single center's experience with 60 resected IPMTs revealed that these tumors are being recognized more frequently. [84] In this series, 37% of the resected IPMTs were associated with an adjacent invasive adenocarcinoma, and the 5-year survival for those patients undergoing IPMT resection was 57%. [84]

Giant Cell Tumors

These rare tumors are characterized by the presence of bizarre giant cells and sarcomatoid cells supported by minimal fibrous tissue. Giant cell tumors have a poor prognosis, even worse than common ductal adenocarcinoma.

Acinar Cell Tumor

Acinar cell carcinomas are uncommon and characterized by acinar arrangement of cells supported by minimal fibrous stoma. Zymogen granules are present, which may be identified by electron microscopy. Patients may have elevated serum lipase levels and associated nonsuppurative panniculitis of the extremities and bone marrow and manifest subcutaneous nodules and poly- arthritis. These tumors usually occur in adults in their sixth to eighth decades of life.

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CHAPTER 97

Robert T. Jensen

ENDOCRINE NEOPLASMS OF THE PANCREAS

Endocrine tumors of the pancreas are classified according to the type of clinical syndrome present (Table 97-1). In general they share a number of common features including various aspects of pathology, natural history, considerations in treatment, localization studies, and treatment of metastatic tumors. In each case, except for nonfunctioning pancreatic endocrine tumors (PETs), the principal clinical manifestations of the endocrine tumor are due to the extensive release of hormones by the tumor into the circulation. Each of the various hormones released in the different syndromes occur naturally and are important in mediating various physiological processes (see Chapter 4). However, in the PET syndromes these hormones are not under normal physiological regulation and are released autonomously by the tumor. PETs are classified as either functional or nonfunctional (see Table 97-1) depending on whether a clinical syndrome due to an ectopically released hormone is present (i.e., gastrinoma, insulinoma, VIPoma, glucagonoma, somatostatinoma, GRFoma, ACTHoma, PETs causing carcinoid syndrome, or hypercalcemia). Nonfunctional PETs frequently ectopically release hormones and peptides (pancreatic polypeptide [PP], chromogranin, neurotensin, neuron-specific enolase); however, these cause no distinct clinical syndromes. [1], [2], [3] and [4] The term *nonfunctional PET* is commonly used and will be retained here to mean any PET not secreting products that cause a distinct syndrome even though some actually release peptides and are, strictly speaking, functioning. The term *pancreatic endocrine tumor (PET)* is also retained in this chapter to indicate the tumors listed in Table 97-1, even though it is also a misnomer in that a number of these tumors also occur outside the pancreas (i.e., gastrinomas, VIPomas, somatostatinomas, GRFomas, ACTHomas [see Table 97-1]). These tumors are also frequently called islet cell tumors; however it is unproven that they originate from pancreatic islets. [5], [6] It is important to realize that even though these tumors are usually slow growing in many cases, some are aggressive, [2], [7], [8] and [9] therefore, effective therapy will require treatment of both the autonomous hormone overproduction as well as treatment directed at the tumor itself. In this chapter the common features of these tumors will be dealt with together and the distinctive features separately. Gastrinoma was discussed in Chapter 67 and will, in general, not be discussed here.

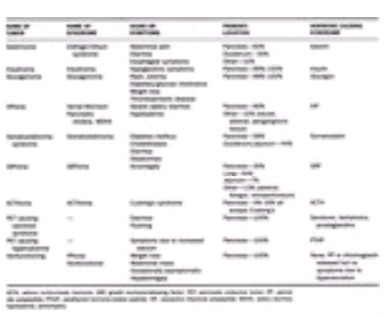

TABLE 97-1 Endocrine Tumors of the Pancreas

EPIDEMIOLOGY

In general, PETs are uncommon, having a prevalence of less than 10 per million population. [2], [10], [11] In some older series insulinomas were reported to be the most common PET with an incidence in various series of 0.8 to 0.9 per million population per year, whereas gastrinomas were reported to occur in 0.1 to 0.4 per million per year. [12] However, in more recent studies [10], [12] gastrinomas were as common as insulinomas (1–3 new cases/year/million population). The remaining symptomatic PETs each occur in less than 0.5 per million per year. [10], [11] Approximately 400 cases of glucagonoma have been reported worldwide, [13], [14], [15] and [16] 250 cases of VIPomas, [17], [18] and [19] less than 180 cases of somatostatinomas, [10], [20], [21], [22] and [23] whereas the number of cases of GRFomas is unclear. The relative frequency of these tumors is insulinomas, nonfunctional PETs, and gastrinomas occurring approximately with equal frequency, 2 to 8 times as common as VIPomas and 17 to 30 times as common as glucagonomas. [2], [10], [11] Somatostatinomas were equal in frequency to glucagonomas in some studies, but less common in others. [2], [10], [11] In various series PETs had an incidence equal to, [11] or 50%, [24] of that seen with carcinoid tumors. In autopsy studies, PETs occur in 0.5% to 1.5% of cases; [7], [11], [25], [26] however, in less than 1 of 1000 cases was a functioning PET thought to occur. [7], [11], [25] In surgical studies nonfunctioning PETs are reported to comprise 15% to 33% of all PETs removed, [1], [10], [27], [28] and [29] and in a large pathology series to account for 36% of all PETs. [6] In addition to the established PET listed in Table 97-1, it has been proposed [30] that PETs secreting calcitonin cause a distinct syndrome with diarrhea. However, too few cases are described to include this syndrome. Furthermore, with other causes of hypercalcitonemia, such as with medullary thyroid cancer, only 25% to 42% of patients develop diarrhea and studies suggest it is not secondary to the increased calcitonin levels per se, but due to a motility disorder of which the causative factor(s) is unknown. Therefore, until more patients are studied it remains unclear if this should be considered a specific PET syndrome. [30], [31], [32] and [33]

PATHOLOGY AND TUMOR BIOLOGY

PETs share a number of pathological features. [5] These tumors frequently contain ductular structures [5], [6] and produce hormones not normally produced in the adult pancreas. Therefore they are thought to originate from an immature stem cell. [5], [6] Because endocrine cells bud off from ductules during ontogenesis of the pancreas and ductular structures are present in these tumors, it has been suggested these cells may originate from ducts. [34] The tumors are thought to originate from cells that are part of the diffuse neuroendocrine cell system. [5], [6], [35], [36] These cells share cytochemical properties and these tumors, together with carcinoid tumors, medullary carcinoma of the thyroid, melanomas, and pheochromocytomas, [5], [6], [36], [37], [38] and [39] have been called APUDomas (an acronym for *a*mine *p*recursor *u*ptake and *d*ecarboxylation). [2], [5], [6], [36], [37] and [38] The tumors are composed of monotonous sheets of small round cells with uniform nuclei and cytoplasm (Fig. 97-1). Mitotic figures are uncommon. Ultrastructurally, the tumors demonstrate electron dense granules which contain various peptides, amines, neuron-specific enolase, synaptophysin, and chromogranin A and C. [2], [5], [6], [38] PETs are frequently multihormonal when examined by immunocytochemical studies. [5], [6], [12], [27], [40], [41] In various series more than 50% of all PETs contained more than one hormone by immunocytochemistry. [5], [6], [12], [40], [41] and [42] In insulinomas, for example, glucagon has been identified by immunocytochemical methods in 0% to 44%, somatostatin in 0% to 18%, gastrin in 3% to 11%, PP in 18% to 39%, and ACTH-like immunoreactivity in 11%. [27], [38] Particularly common are PP cells which have been identified in 22% to 39% of insulinomas, 0% to 67% of glucagonomas, and 50% to 75% of VIPomas. [27] In some cases numerous hormones are released into the circulation whereas in others only one peptide may be detectable. [27], [40], [41] At present it is not apparent why

patients with a given PET syndrome present only with a syndrome characteristic of hypersecretion of only one of these peptides; even though the tumor contains multiple peptides. Possible explanations include that only one of the peptides produced is released, only one is released in sufficient quantity to cause symptoms, only one peptide produced is biologically active, or all the peptides are released but some have antagonistic physiological actions. [43]

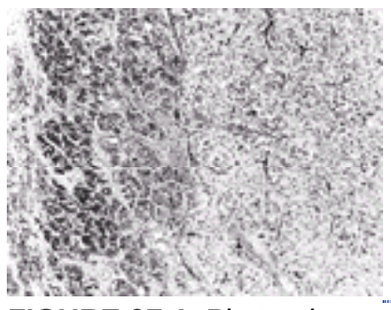

FIGURE 97-1. Photomicrograph of an insulinoma. This insulinoma (**right**) demonstrates a typical histological pattern seen with pancreatic endocrine tumors (PETs) with monotonous sheets of small round cells with uniform nuclei and cytoplasm. The tumor is well marginated and interfaces with the normal pancreas (**left**) by a distinct margin. Hematoxylin and eosin stain, original magnification ×125.

In addition to producing and frequently releasing multiple gastrointestinal peptides, PETs produce a number of products that are characteristic of neuroendocrine differentiation and are used to identify these tumors. These general markers are neuron-specific enolase, chromogranins (A,B,C), synaptophysin, epitope Leu-7, protein gene product (PGP)-9.5, and the alpha or beta subunit of human chorionic gonadotropin (a-hCG, ß-hCG). [3], [11], [44], [45] and [46] Chromogranins are acidic glycoproteins that are present in almost all endocrine or neuronal tissue. [3], [5], [44] Chromogranin is released into the plasma in patients with PETs or carcinoid tumors, [24], [44], [45], [47], [48] and [49] and elevated plasma levels of chromogranin A, B, or C are reported in more than 90% of such patients. [3], [24], [44], [45], [49] Increased plasma levels of a-hCG and ß-hCG also occur in PETs and have been reported by some to be suggestive of malignancy; however, this remains controversial. [2], [3], [12], [27], [45], [46] Neuron-specific enolase (NSE), the r-r dimer of the glycolytic enzyme enolase, occurs in the cytoplasm of most neuroendocrine cells, is present in most PETs as well as carcinoids and other APUDomas, [5], [36], [39] and is frequently released into the plasma and can be used as a tumor marker. [45], [50] Synaptophysin is a calcium-binding vesicle membrane glycoprotein that is synthesized independently of the other neuroendocrine proteins. [11], [39] The presence of chromogranin immunoreactivity in the tumor is the most generally used marker to identify a pancreatic tumor as endocrine in origin. [5]

Because patients with PETs are now living longer with increasingly effective therapy, the development of a secondary symptomatic PET syndrome may become more common. [40], [41] One study suggests a secondary symptomatic PET syndrome is common, occurring in 7.5% of patients with a mean follow-up of 19 months, [40] whereas another study suggests it is uncommon, occurring in only 2% of patients with gastrinoma with a mean follow-up of 11 years. [41] Because of the frequent presence of multiple peptides by immunocytochemistry in the PET, it has become increasingly difficult, if not impossible in most cases, to determine by immunocytochemistry, which of the hormones found in the tumor are clinically important in a patient. [6], [12], [38] Therefore, a functional PET syndrome should only be diagnosed if the clinical symptoms are present and plasma elevations are documented, and not on the basis of immunohistochemical results only.

In general, histological classification of PETs has failed to predict the growth pattern of the tumor or determine whether it is malignant. [6], [12], [38] Furthermore, there is no definite correlation between the histological pattern and the type of clinical syndrome the tumor is associated with. [12], [38], [51] Therefore, at present malignancy can only be unequivocally established in those patients who are demonstrated to have metastatic tumor that has spread either to lymph nodes or the liver, gross invasion or infiltration into adjacent organs, or clear blood vessel invasion. [5], [6], [12], [38] Because of this it is not completely established what percentage of PETs are malignant. The benign nature can only be established by long-term follow-up. In general, 5% to 10% of insulinomas are reported as malignant, whereas in various series 50% to 90% of the other tumors are reported as malignant. [2], [7], [12], [16], [17], [20], [23], [38], [51], [52]

The size of the tumor is usually not related to the severity of the hormonally induced symptoms; however in general, there is a correlation of size with the occurrence of malignancy. [6], [7], [8] and [9], [42], [51], [53] Whereas insulinomas, like gastrinomas, are generally small (<2 cm), glucagonomas and the other PETs are larger at the time of detection (frequently >5 cm). [12], [16], [17], [20], [23], [51], [52] The majority of the primary PETs are solitary and encapsulated when not associated with the multiple endocrine neoplasia type I (MEN I) (discussed subsequently). [5], [6] When metastatic spread occurs it is usually to regional lymph nodes first and later to the liver. Late in the course of the disease, metastases to bone may also occur. [8], [54]

PETs may occur either in a nonfamilial form (sporadic) or be associated with an inherited syndrome. [55], [56], [57] and [58] Inherited syndromes associated with PETs include MEN I [58], [59], [60] and [61] and three phakomatoses (von Recklinghausen's disease, von Hippel-Lindau syndrome, tuberous sclerosis). [11], [55], [56], [57] and [58] MEN I or Wermer syndrome is an autosomal dominant disorder characterized by tumors or hyperplasia of multiple endocrine organs with hypercalcemia due to hyperparathyroidism being the most common abnormality (i.e., occurring in 97% of patients). [57], [59], [60] Nonfunctional PETs are the second most frequent abnormality, occurring in 80% to 100% of patients. [60] Functional PETs occur in 80% of patients with MEN I. [59], [60] and [61] The incidence of gastrinomas, insulinomas, glucagonomas, and VIPomas in MEN I is 54%, 21%, 3%, and 1%, respectively. [60] Pituitary and adrenal cortical adenomas also occur but are less common. [60], [61] In MEN I, the chromosome defect mapped to chromosome 11q13, [59] and a study [62] using positional cloning identified the *MEN-I* gene in this region. The *MEN-I* gene contains 10 exons and encodes a unique ubiquitously expressed 610-amino acid protein, called menin, a nuclear protein that interacts with the AP1 transcriptional factor, Jun D [59] and with SMAD3, a protein important in mediating the cell growth inhibition mediated by transforming growth factor-ß (TGF-ß). [63] Numerous studies suggest that the *MEN-I* gene is a tumor suppressor gene, functioning in a similar manner to that suggested by Knudson for retinoblastoma. [58], [59], [64] In such a circumstance, the affected individual inherits one altered copy of the responsible gene from an affected parent, and then acquires a mutation on the remaining normal allele. Studies [58], [65], [66] suggest a similar double acquired loss of the *MEN-I* gene may be important in sporadic (noninherited) PETs because 32% to 38% of these have loss of heterozygosity at the 11q13 locus. [58], [66] Furthermore, 27% to 39% of sporadic PETs have mutations in the *MEN-I* gene, suggesting some sporadic PETs have similar pathogenesis to PETs in patients with MEN I. [58], [66] The exact percentage of patients with PETs who have MEN I varies in different series and with different tumors from less than 5% to 25%. [12] MEN I is present in 20% to 25% of patients with gastrinomas, [12], [67] 4% to 6% with insulinomas, [52], [60] 13% to 17% with glucagonomas, [15], [16] 6% to 11% with VIPomas, [17], [68] 7% with somatostatinomas, [23] and 33% with GRFomas. [20] The recognition that the PET can be part of the MEN-I syndrome is important because multiple PETs are frequently present. [6], [12] Furthermore, screening of family members for various features of the syndrome is indicated. [12], [59], [60] and [61]

Almost all insulinomas either occur in the pancreas (see Table 97-1) or are attached to it, [52], [69], [70] and [71] although rarely an aberrant insulinoma may be found in carcinoid tumors in the duodenum, ileum, lung, or mesentery. [52], [69] Multiple insulinomas occur in only 10% to 13% of cases and in these patients the presence of MEN I should be suspected. [51], [52] Glucagonomas occur almost entirely within the pancreas, usually in the pancreatic body and tail. [6], [14], [15] and [16], [51], [72] In contrast to insulinomas or gastrinomas, the majority of glucagonomas are usually large (3–10 cm) at the time of diagnosis. [14], [15] and [16], [51], [72] VIPomas in adults are almost all pancreatic in location (i.e., >90%); however, occasionally the VIPoma syndrome is reported in association with a pheochromocytoma or carcinoid tumor of the intestine. [17], [18] and [19] In children, the VIPoma syndrome is often due to a ganglioneuroma or ganglioneuroblastoma. [19], [73], [74] VIPomas are usually large tumors. [17], [19] Somatostatinomas, similar to gastrinomas, frequently are found in extrapancreatic locations (see Table 97-1). [12], [20], [21], [23] In various reviews, [20], [23] 47% to 56% of somatostatinomas are in the pancreas, usually in the pancreatic head and 44% to 52% are extrapancreatic, usually in the small intestine (duodenum or jejunum). Similar to glucagonomas or VIPomas, somatostatinomas tend to be large (mean size = 5 cm). [21], [23], [75] Nonfunctioning PETs and PPomas occur in the pancreas, and are usually greater than 5 cm in diameter. [1], [4], [6], [29] Other less common functioning PETs such as those associated with acromegaly (GRFomas), [21], [76], [77] carcinoid syndrome, [78], [79] paraneoplastic hypercalcemia, [27], [80], [81] and [82] or Cushing syndrome, [27], [83], [84] and [85] are usually large tumors within the pancreas.

In addition to the MEN-I syndrome, some studies report PETs in patients with von Recklinghausen's disease, [22], [55], [86] von Hippel-Lindau syndrome, [55], [87] and tuberous sclerosis. [55], [88], [89] Duodenal somatostatinomas have been reported in a number of patients with von Recklinghausen's disease [22], [23], [55], [90] as well as

Zollinger-Ellison syndrome. [86] In von Hippel-Lindau syndrome, 10% to 17% of patients have a PET, which is usually nonfunctional, but occasional insulinomas and VIPomas are described. [55] Somatostatinomas have also been reported in von Hippel-Lindau syndrome. [91] Three patients with tuberous sclerosis were reported with insulinomas and one was reported with a nonfunctioning PET. [88], [89]

The molecular pathogenesis of PETs, until recently, was largely unknown. [55], [56], [58] In contrast to most common nonendocrine tumors (pancreatic, colonic, gastric adenocarcinomas, etc.), alterations in neither common tumor suppressor genes (retinoblastoma gene, *p53* gene) nor common oncogenes (i.e., *jun, src, ras, fos, myc,* etc.) are generally present in PETs or the closely related carcinoid tumor, [55], [57], [58] suggesting they are not generally important in their molecular pathogenesis. Studies suggest alterations in the *MEN-I* gene, as discussed previously in this section, in the *p16/MTS1* tumor suppressor gene, *DP64/SMAD 4* gene, amplification of the *HER-2/neu* protooncogene, and loss of an unknown tumor suppressor gene on chromosome 1 or 3p could also be important. [7], [55], [57], [58], [92], [93] Mutations in the *MEN-I* gene occur in one third of sporadic PETs, [58], [66] and alterations in the *p16/MTS1* gene in 50% to 92% of PETs, [92], [93] suggesting these are particularly important. Whether the presence of these alterations carries a worse prognosis is unclear.

CLINICAL FEATURES AND DIAGNOSIS

In each PET syndrome except nonfunctioning tumors, early symptoms are caused by the excessive hormone release, whereas late symptoms are primarily due to the metastatic spread of the tumor per se (cachexia, pain, bleeding).

Insulinoma

Insulinomas are endocrine tumors that originate in the pancreas, secrete excessive amounts of insulin, and cause a distinct syndrome characterized by symptoms due to hypoglycemia.

Insulinomas rarely occur in adolescence. The average age of presentation in most series is between 40 and 50 years of age, with 60% of the insulinomas occurring in women. [51], [52], [70], [71], [94], [95]

The clinical manifestations of insulinomas are due to the hypoglycemia secondary to the excess unregulated insulin secretion by the tumor. The symptoms are typically associated with fasting (i.e., the attacks characteristically occur before breakfast, when a meal is delayed, or hours postprandially). The majority of patients have symptoms of central nervous system dysfunction secondary to hypoglycemia (neuroglycopenic symptoms) which include headaches, confusion, lightheadedness, visual disturbances, irrational behavior, drowsiness, or even confusion resulting in coma. Patients with insulinomas misdiagnosed as psychiatric or neurological problems have been reported. [11] The majority of patients also have symptoms due to catecholamine excess secondary to the hypoglycemia such as sweating, tremor, palpitations, and irritability. [51], [94] The average time from onset of symptoms to diagnosis is 3 years, although some studies suggest this to be decreasing. [94], [95]

The presence of an insulinoma should be suspected in all patients with hypoglycemia, especially patients with a history of fasting hypoglycemia or with a family history of MEN I. Fasting hypoglycemia can be mediated by a number of different conditions (Table 97-2). [51], [70], [96], [97] and [98] It should be remembered that insulinoma is only one cause of fasting hypoglycemia and other causes should also be suspected such as inadvertent or deliberate administration of insulin, ingestion of oral hypoglycemia agents, alcoholism with severe liver disease or malnutrition, or other extrapancreatic tumors (see Table 97-2). The diagnosis of insulinoma requires the demonstration of hypoglycemia combined with an inappropriate elevation of plasma insulin concentration and ultimately a PET.

I. Mediated by endogenous insulin or insulin-like factors
  A. Pancreatic islet disease (insulinoma, hyperplasia, nesidioblastosis)
  B. Spontaneous autoimmune antiinsulin antibody syndrome
  C. Autoantibodies to insulin receptor
  D. Noninsulin tumor-associated hypoglycemia
II. Reduced Hepatic Glucose Output
  A. Deficient gluconeogenesis or glycogen storage
  B. Hormonal deficiencies (e.g., adrenal insufficiency)
  C. Enzymatic defects (e.g., glucose-6 phosphatase deficiency)
  D. Ethanol consumption and poor nutrition
  E. Severe liver disease
III. Drugs or Other Pharmacological Causes
  A. Sulfonylureas or biguanides
  B. Insulin administration
  C. Ingestion of akee fruits (hypoglycin)
  D. Other drugs (aspirin, pentamidine, haloperidol, quinine)

Modified from refs. 51, 70, 98.

TABLE 97-2 Causes of Fasting Hypoglycemia

The key to establishing the diagnosis is suspecting the symptoms could be due to hypoglycemia, and Whipple's triad, [99] described in 1938, was long used to diagnose insulinomas. This triad consisted of hypoglycemic symptoms, detecting hypoglycemia (<50 mg/dL), and relief of symptoms with glucose ingestion. Unfortunately the symptoms are not specific for insulinoma [100] and were present in only 43% of cases of insulinomas in a large review of 1085 cases. [52] A fast with monitoring of plasma insulin, glucose, C-peptide, and proinsulin values is now the study usually used to establish the diagnosis of insulinoma. [70], [71], [94], [98] A 72-hour fast in the hospital (with free access to water) is usually used, [51], [98] although one study proposes a 48-hour fast is sufficient. [101] The blood studies mentioned should be monitored at regular intervals (2–4 h) and more frequently if blood sugar decreases to less than 50 mg/dL. If at any point the patient becomes symptomatic, repeat serum levels of the blood studies should be obtained before administering glucose. The test is terminated for neuroglycopenic symptoms or for persistent glucose levels less than 40 mg/dL. Over 90% of insulinomas reported have been detected in this fashion and 70% to 80% of patients will develop hypoglycemia during the first 24 hours of fasting and 98% will develop the condition by 48 hours. [51], [94], [95] In normal nonobese subjects, serum insulin concentrations decrease to less than 6 µU/mL when blood glucose decreases to 40 mg/dL or less and the ratio of serum insulin to glucose remains less than 0.3 (in mg/dL). [51], [95], [97], [98] An insulinoma is considered present if serum insulin concentrations remain constant or increase during hypoglycemia. One set of proposed criteria are:

- documented glucose levels near or below 40 mg/dL (2.2 mM)
- concomitant insulin levels greater than 6 µU/mL (43 pmol/L);
- elevated C-peptide levels (greater than 0.2 nmol/mL in a patient without elevated sulfonylureas in the plasma). [70]

Some add a requirement for a serum proinsulin =5 pmol/L [71], [102] or the insulin/glucose ratio more than 0.3 [51], [98] In one study all 114 patients with insulinoma operated on between 1980 and 1995 had a positive fast, [70] whereas in another review all 132 patients with insulinomas fulfilled the biochemical criteria, but 4 of 132 patients (3%) did not become symptomatic. [103] In the very small percentage of patients with a negative test (i.e., serum insulin concentrations decreasing to less than 6 µU/mL with hypoglycemia), yet insulinoma is still suspected, additional tests (i.e., various provocative tests) may have to be used. [27], [51], [70], [97], [98], [103]

The diagnosis of insulinoma can be difficult because an inappropriately elevated serum insulin concentration associated with hypoglycemia can be observed in a number of different conditions (see Table 97-2). [104] In addition to insulinomas, ß-cell hyperplasia or nesidioblastosis, factitious or inadvertent administration of excessive insulin or oral hypoglycemia agents and autoantibodies against insulin or the insulin receptor can also cause those findings (see Table 97-2). [51], [98], [104] ß-Cell hyperplasia or nesidioblastosis consisting of a proliferation of insulin-producing cells is a leading cause of hyperinsulinism in newborns and has also occasionally been reported in adults. [98], [105] Of these varying causes of hypoglycemia and hyperinsulinemia (see Table 97-2), surreptitious use of insulin or sulfonylureas is the most difficult to distinguish from a patient with insulinoma. [97], [98], [102], [106] To differentiate these conditions a combination of studies consisting of the determination of serum concentrations of proinsulin, C-peptide, antibodies to insulin, and sulfonylurea need to be done. [97], [98], [102], [106]

Insulin is synthesized as proinsulin, which consists of a single chain molecule containing both a 21-amino acid a-chain and a 30-amino acid ß-chain connected by a 33-amino acid connecting peptide (C-peptide). [107], [108] Insulin and C-peptide are liberated in equimolar amounts, and because proinsulin or its intermediate are also found in granules, it is also detected in small quantities in the serum with insulin. [98], [107], [108] Normal subjects have less than 25% of the serum insulin as proinsulin.

Serum proinsulin has been reported to be elevated in at least 80% to 90% of patients with insulinoma. [97], [98], [108] Since high proinsulin values are not found in other hypoglycemic disorders (elevated levels can occur in cirrhotics and patients with type-2 diabetes), an elevated value is very helpful in the diagnosis of insulinoma. However, a normal level does not exclude the diagnosis. In insulinoma the characteristic findings are either a normal or elevated serum concentration of C-peptide, antibodies to insulin are not present, and sulfonylurea is not detected in the blood. [97] In a typical patient with surreptitious use of insulin the C-peptide level is decreased, proinsulin is normal or decreased, antibodies to insulin may be present, and serum sulfonylurea determinations are negative. [97], [98], [104] In patients with surreptitious use of sulfonylureas, the serum concentration of proinsulin is usually normal, antibodies to insulin are not present, and sulfonylurea is detected in the serum or the urine. [97], [98] It is important to remember that the methods usually used for measuring second generation sulfonylureas are insensitive. [71]

VIPoma

The VIPoma syndrome is due to an endocrine tumor usually located in the pancreas that secretes excessive amounts of vasoactive intestinal polypeptide (VIP) which causes a distinct syndrome characterized by fasting, large volume diarrhea, hypokalemia, and hypochlorhydria (Table 97-3).

SIGN/SYMPTOM	FREQUENCY (%)
Secretory diarrhea	89–100
Hypokalemia	67–100
Weight loss	33–72
Dehydration	44–95
Hypochlorhydria	34–76
Hyperglycemia	20–50
Hypercalcemia	25–50
Flushing	13–28
Dilated, atonic gallbladder	Unknown

From refs. 17, 19, 68, 115, 131.

TABLE 97-3 Clinical Features of the Vasoactive Intestinal Polypeptide Syndrome

The VIPoma syndrome is also called the Verner-Morrison syndrome because of its original description by these two authors in two patients with profuse watery diarrhea, hypokalemia, and dehydration with a non-ß-cell pancreatic islet cell tumor. [109] It has also been termed pancreatic cholera [110] and the WDHA syndrome [111] (for the *w*atery *d*iarrhea, *h*ypokalemia, and *a*chlorhydria that some patients develop). [18], [19], [74] While early in the presentation the diarrhea may be episodic or intermittent, characteristically the diarrhea is large in volume (>1 L/d), secretory in nature, and persists during fasting (see Table 97-3). [18], [19], [27] The diarrhea fluid frequently is described as having the appearance of weak tea. An increased net secretion of sodium and chloride into the small intestine has been demonstrated in these patients. [112], [113] The sum of fecal sodium and potassium concentrations multiplied by two will equal the osmolality, thus there is no osmotic gap. [19] In general these patients are reported not to develop steatorrhea (only 16% do so [19]) or have abdominal cramping pain, but some studies suggest pain may be more common than once was typically reported. [19], [27], [68] Studies assessing malabsorption are usually normal with a normal fecal fat excretion in 84%, normal D-xylose in 94%, and normal Schilling's test in 87%. [19] The increased fecal loss frequently leads to dehydration (see Table 97-3). [19], [68] Hypokalemia is present in most patients (see Table 97-3) [17], [18], [19], [68], [114], [115] and is the result of the large losses of potassium in the diarrhea fluid. Hypochlorhydria rather than achlorhydria is found in 28% to 76% of patients with VIPoma syndrome (see Table 97-3). [18], [68], [114], [115] This has been attributed to the inhibitory effect of VIP on gastric acid secretion. [18], [18], [27] Flushing is seen in 13% to 28% of patients with the VIPoma syndrome [17], [18], [68], [114], [116] and has been attributed to the vasodilatory effects of VIP (see Table 97-3). [18], [117] The fact that prolonged infusion with VIP in humans results in a gradual decrease in flushing suggests the development of tachyphylaxis. [11] This phenomenon has been used to explain why only a minority of patients develop flushing. [18] Hyperglycemia is noted in 20% to 50% of patients with VIPomas [19], [68] and it has been attributed [18] to the glycogenolytic effect of VIP on the liver (see Table 97-3). [118] Hypercalcemia also is reported in 25% to 76% of patients with VIPoma. [18], [19], [27], [68] At present the mechanism of hypercalcemia in most cases is not established; however, it seems to be related to the tumor itself because it is relieved by complete tumor removal. [19] Only 6% to 11% of patients with VIPomas have a history of MEN I, which is why MEN I is rarely the cause of the hypercalcemia. [17], [19], [68]

Beside VIP a number of substances, [18], [119] including secretin, [11] gastric inhibitory polypeptide, [120] pancreatic polypeptide, [121] and prostaglandins [122] are reported to cause diarrheagenic secretion in some patients. Controversy arose in early studies whether VIP was the sole cause of the diarrhea in WDHA because it was not found to be elevated in all patients by different investigators and exogenous VIP infusions for 1 or 2 hours did not cause diarrhea in humans. [123] However, in more recent studies, only VIP has been found to be elevated consistently in patients with the WDHA syndrome. [18], [73], [119], [124], [125] Furthermore, continuous infusion of VIP for 10 hours in normal subjects to achieve plasma VIP concentrations similar to those seen in patients with WDHA produced profuse watery diarrhea within 6 to 7 hours. [124], [126] The diarrhea produced in these patients is similar to that seen in patients with WDHA. The ability of VIP to cause intestinal secretion is consistent with its known mechanism of action. VIP has been shown to cause net chloride secretion associated with increased short circuit current, to bind to specific receptors on intestinal epithelial cells, and to activate adenylate cyclase and increase cyclic AMP in intestinal cells. [27], [127] Occasionally patients with the WDHA syndrome have normal plasma concentrations of VIP. [18] Another VIP-related peptide, peptide histidine isoleucine (PHI), is reported to be elevated in the plasma of patients with WDHA. [19], [27], [128], [129] Tumor tissue obtained from patients with the WDHA syndrome also demonstrated PHI-like immunoreactivity which was produced by the same cells as VIP. [19], [27], [128], [129] Furthermore, analysis of a WDHA tumor demonstrated that VIP and PHI sequences are in the same gene. [127], [129] PHI infusions also caused intestinal secretion although PHI was 32-fold less potent than VIP [130] and therefore, even though PHI was reported to coexist in 22 of 24 VIPomas, [128] because of the low potency, it remains to be proven whether PHI is contributing to the diarrheal state. [19]

The definite diagnosis of VIPoma requires the establishment of a secretory diarrhea, demonstration of elevated serum concentrations of VIP, and identification of a PET. The likely possibility that VIPoma and not some other cause is responsible for the diarrhea may be suggested by measuring the stool volume. [18], [19], [27] In 70% to 80% of patients with VIPomas the stool output is greater than 3 L/d, [19], [131] in 100% of 52 patients it was more than 1000 L/d. [19] When the stool output is less than 700 mL/d the diagnosis of VIPoma is excluded. [18] In addition the diarrhea in patients with VIPomas persists during fasting [18], [19] and the sum of the concentration of stool sodium and potassium multiplied by two equals the isotonicity. [19]

A number of diseases can cause chronic secretory diarrheas with this volume and need to be differentiated from VIPomas. [32] The diarrheas due to midgut carcinoids in the carcinoid syndrome or medullary thyroid carcinoma are primarily due to motility disturbances and rarely reach this volume. [19], [32] However, high-volume fasting diarrhea occurs in patients with Zollinger-Ellison syndrome [12], [32], [67] and diffuse islet-cell hyperplasia, [19] as well as among patients with surreptitious use of laxatives or the pseudopancreatic cholera syndrome. [19], [73], [132] The serum gastrin concentration is elevated in patients with Zollinger-Ellison syndrome but not in patients with VIPoma. Furthermore, with effective treatment of the gastric hypersecretion the diarrhea stops in patients with Zollinger-Ellison syndrome. [12], [32], [67], [133]

To differentiate patients with VIPoma from those with pseudopancreatic cholera syndrome or laxative abuse a reliable VIP radioimmunoassay is required. [18], [19], [73] The normal fasting plasma VIP concentration in a number of laboratories is 0 to 190 pg/mL. [18], [73], [119] It is best that the fasting plasma VIP concentration be determined at the time when diarrhea is present because occasionally between diarrhea periods in some patients with VIPoma, VIP levels may be normal. [18] The mean concentration of plasma VIP in one series of 29 patients with the VIPoma syndrome was 956 pg/mL with a range of 225 to 1850 pg/mL. [18], [27] In another series [74] of 52 cases of VIPoma the mean value was 702 pg/mL with a range from 159 to 2530 pg/mL. In two reviews, [17], [68] 94% to 100% of patients with VIPoma had elevated plasma VIP levels. Normal plasma VIP levels are reported to occur in 12% of cases with a pancreatic tumor and in 82% of cases with pancreatic islet-cell hyperplasia. [19] In most recent studies plasma VIP levels are not elevated in patients with laxative abuse. [18], [19], [73], [114], [119], [120] Patients with chronic secretory diarrhea due to chronic laxative abuse can be difficult to detect, but should be suspected if plasma VIP levels are normal or a laxative screen is positive. [134] Because of the difficulty in diagnosing laxative abuse, a laxative screen should be performed in any patient with unexplained chronic diarrhea. [135]

Elevated plasma VIP levels should not be the sole basis for the diagnosis of a VIPoma. [18], [136] Other conditions including prolonged fasting, inflammatory bowel disease, small bowel resection, radiation enteritis, and chronic renal failure can occasionally elevate plasma VIP levels. [18], [136] In one study of 193 patients with chronic diarrhea, [136] eight different gastrointestinal hormones (motilin, PP, neurotensin, somatostatin, substance P, VIP, gastrin-releasing peptide, and calcitonin) were assessed and 45% of the patients had one or more elevated levels; however, no patient had a PET. This study shows false-positive elevated plasma levels for those hormones reported to cause diarrhea, including VIP, are not uncommon and therefore tests need to be ordered selectively and carefully interpreted in light of other

clinical and laboratory findings to establish a correct diagnosis. [136]

Glucagonoma

Glucagonomas are endocrine tumors of the pancreas that secrete excessive amounts of glucagon and cause a distinct syndrome characterized by dermatitis, glucose intolerance, weight loss, and anemia (Table 97-4).

SIGN/SYMPTOM	FREQUENCY (%)
Dermatitis (migratory necrolytic erythema)	64–90
Hypoaminoacidemia	41–100
Glucose intolerance or diabetes mellitus	38–90
Weight loss	56–96
Anemia	33–85
Diarrhea	14–29
Thromboembolism	12–35
Glossitis, cheilitis	14–40
Psychiatric disturbance	0–17

from refs. 13–16, 72, 141–143, 152.

TABLE 97-4 Clinical Features of the Glucagonoma Syndrome

The first description of what we know as the glucagonoma syndrome was reported in 1942 [137] in a patient with a skin rash associated with pancreatic cancer. McGavran and colleagues, in 1966, reported the first well-described case of a patient with the glucagonoma syndrome. [138] The patient had elevated immunoreactive glucagon in plasma, diabetes mellitus, a skin rash, and a PET. In 1973, Wilkinson [139] introduced the term *necrolytic migratory erythema* for the skin rash associated with PETs. Mallinson and colleagues in 1974 [140] specifically established the association of glucagonomas with the skin rash when they reported nine cases with the clinical glucagonoma syndrome consisting of dermatitis, diabetes mellitus, unexplained weight loss, hypoaminoacidemia, anemia, and a glucagon-producing tumor of the pancreas.

The peak incidence for the glucagonoma syndrome is 45 to 70 years of age; the syndrome has not been described in children. [13], [14], [15] and [16], [51], [72] The characteristic skin rash associated with the glucagonoma syndrome, called necrolytic migratory erythema is present in the vast majority of cases (see Table 97-4). [14], [15] and [16], [72], [139], [141], [142] and [143] The skin lesion starts as an annular erythema at intertriginous and periorificial sites. It is normally found on the buttocks, groin, perineum, and thighs. The erythema subsequently becomes raised and the central parts form superficial bullae. The top of the bulbous lesion frequently detaches, leaving eroded areas that become crusted (Fig. 97-2; see also Color Fig. 97-2). Lesions frequently become confluent. The extent and the severity of the lesions may wax and wane. Healing in 2 or 3 weeks results in hyperpigmentation (see Fig. 97-2). Histologically, early skin lesions demonstrate a superficial spongiosis and necrosis and subcutaneous blister formation. [139], [141], [142] and [143] The pathogenic mechanism by which the glucagonoma syndrome produces this characteristic skin rash is unclear. It is not established that the skin rash is associated with the hyperglucagonemia because numerous patients have been given large doses of glucagon for long periods of time without developing the characteristic rash. [141] Glucagon-induced hypoaminoacidemia may be one mechanism because correction of the hypoaminoacidemia has been shown to eliminate the dermatitis without changing plasma glucagon concentrations. [141], [144], [145] The resemblance of the lesions to those seen with zinc deficiency have resulted in trials of therapy with zinc, with some responses. [15] However, resolution of the rash has been reported following simple hydration with glucose and saline; [141], [146] therefore, it is not known that either hypoaminoacidemia or zinc deficiency are necessarily causative in all patients. A number of other clinical features are also characteristic of the glucagonoma syndrome (see Table 97-4). Angular cheilitis (see Fig. 97-2) is a common feature as is nail dystrophy and thinning of the hair. [14], [15] Some patients develop a painful glossitis (see Table 97-4).

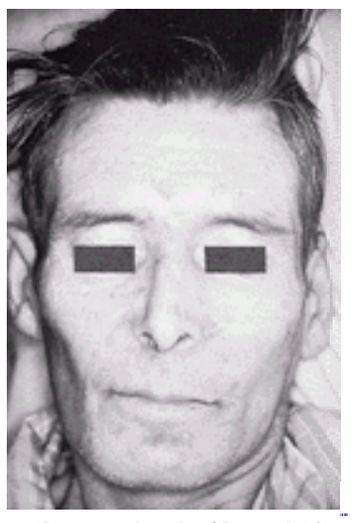

FIGURE 97-2. (See Color Fig. 97-2) Migratory necrolytic erythema involving the face in a patient with metastatic glucagonoma. These typical skin lesions usually start on the extremities or intertriginous or periorificial sites. The lesions initially are erythematous and scaly, later become raised and bullous, and finally become crusty as is evident in this patient. Healing results in hyperpigmentation. Angular cheilitis, a common feature in patients with glucagonoma, is also present in a mild form in this patient. The patient also demonstrates loss of the buccal fat pad and temporal muscle wasting indicative of the generalized wasting these patients characteristically develop.

Hypoaminoacidemia occurs in most patients and, as originally pointed out by Mallinson, the levels may vary with the intensity of the disease. [16], [140], [141] The hypoaminoacidemia is thought secondary to the hyperglucagonemia because glucagon infusions have been demonstrated to have a controlling influence on amino acid metabolism by altering gluconeogenesis. [141], [147] It has been demonstrated in human subjects that glucagon deficiency caused by either suppression with somatostatin or total pancreatectomy increases plasma amino acid concentrations. [27], [148], [149] On the other hand, the administration of glucagon decreases plasma amino acid concentrations. [149] Metabolic studies [150], [151] in patients show an increase in amino acid catabolism, a 15% increase in whole body lipolysis, and a 15% increase in hepatic glucose production; the energy expenditure, as well as rate of protein breakdown, were normal.

Diabetes mellitus and glucose intolerance occur frequently in patients with the glucagonoma syndrome (see Table 97-4). [13], [16], [141], [143], [152] In one study [14] however diabetes was present in only 38% of patients at presentation and developed in an additional 38% as the glucagonoma progressed. The relationship of the hyperglucagonemia to the diabetes mellitus or glucose intolerance remains unclear. Glucagon levels in patients with glucagonomas do not correlate well with the degree of glucose intolerance. [51], [141], [142] and [143], [152] Furthermore, various treatments have resulted in definite changes in plasma glucagon concentrations but have not produced correlated changes in blood glucose. For example, tumor resection and normalization of blood glucose may not result in the correction of the glucose intolerance. [51], [72], [141] The somatostatin analog, octreotide, may depress plasma glucagon levels, but frequently does not improve the diabetes. [11], [14], [15], [153] In some patients however, removal of the glucagon-producing tumor has improved glucose tolerance. [140], [141] Whether or not glucagon causes glucose intolerance appears to depend to a large degree on the patient's insulin reserve. If it is intact, hyperglucagonemia is present without glucose intolerance. [14], [51], [141], [152]

Weight loss is a common feature of patients with the glucagonoma syndrome and it may be profound (see Table 97-4, Fig. 97-2). [13], [14], [15] and [16], [51], [72], [141], [142] and [143] A number of observations suggest weight loss is a specific feature of the glucagonoma syndrome. Weight loss is prominent in patients with small tumors as well as those with metastatic tumors. It is not seen early in other PETs unless malabsorption is present, suggesting the cachexia is an intrinsic feature of the glucagonoma syndrome. [141] The weight loss has been attributed to the catabolic effects of glucagon. [141], [151] However, in an experimental study [154] in which glucagonomas from a pluripotent rat islet tumor were transplanted into normal animals, severe anorexia and adipsia developed. Studies show bilateral vagotomy had no effect on the

development of anorexia, that the satiety peptides, cocaine and amphetamine-regulated transcript (CART) or leptin, are unlikely to be the mediators, and that the unknown mediator can override the potent appetite-stimulatory effect of neuropeptide (NPY). [155], [156] These results [154] suggest that the anorectic effects of glucagonoma are due to the production of a novel substance by the glucagonoma and not due to glucagon per se because other transplanted glucagonomas producing similar levels of plasma glucagon elevation do not cause anorexia. [154]

Thromboembolic phenomena are more common in patients with the glucagonoma syndrome (see Table 97-4). [13], [14] and [15], [51], [72], [141], [142] and [143] Both deep vein thrombosis and pulmonary emboli have been reported to occur in a significant number of cases and have been the cause of death in some patients. [13], [51], [141], [142] and [143] Glucagon is not known to affect coagulation parameters, however the pathophysiology of the thromboembolic events in glucagonoma has not been systematically studied. A normochromic, normocytic anemia is also reported to be a frequent finding in patients with glucagonomas (see Table 97-4). [14], [15] and [16], [51], [72], [141], [142] Serum iron, serum vitamin B_{12}, and serum folate concentrations are usually normal, and the anemia does not respond to therapy with any of these agents. It, however, does respond to resection of the tumor. [141], [157], [158] and [159] Further evidence that the anemia may be due to glucagon excess is that prolonged therapy with a long-acting glucagon preparation decreases erythropoiesis in rats and mice. [159] Psychiatric disturbances, particularly depression, have been reported in a number of patients with the glucagonoma syndrome, however the exact frequency or even whether they are more common in patients with glucagonoma has not been established (see Table 97-4). [14], [15], [72], [137], [139], [141]

Glucagonoma is usually suspected in patients with chronic unexplained and therapy-resistant dermatitis or elevated sedimentation rates and is associated with glucose intolerance, thromboembolic phenomenon, or the MEN-I syndrome (see Table 97-4). [11], [15], [141], [142] and [143], [152] Diagnosis rests on demonstrating a pathological elevation of the plasma glucagon concentration. In almost all patients with glucagonoma the plasma glucagon concentration is elevated (>150 pg/mL), [16], [141], [142], [152] with a mean value of 2110 ± 334 pg/mL (±1SEM) in 23 patients with proven glucagonoma in one study. The highest value in a patient without glucagonoma was 409 ± 29 pg/mL. In a review of 58 published cases of glucagonoma, plasma glucagon levels exceeded 1000 pg/mL in 90%, were between 500 and 1000 pg/mL in 7%, and less than 500 in 3%. [13], [72] In a study from the Mayo Clinic of 21 cases, [14] 48% of patients had values greater than 1000 pg/mL and 86% of patients had values greater than 500 pg/mL. It has been suggested that a plasma glucagon concentration of more than 1000 pg/mL is diagnostic of glucagonoma [141] because other causes of elevated plasma glucagon levels do not cause this degree of elevation. In this study 3 of 21 patients (14%) had plasma glucagon levels less than 500. [14] A high degree of suspicion for the disease therefore should be maintained, especially in patients with small tumors or MEN I. [15] Other conditions that are associated with increased plasma glucagon concentrations include renal insufficiency, acute pancreatitis, hypercorticism, hepatic insufficiency, severe stress (trauma, exercise, bacteremia, diabetic ketoacidosis), prolonged fasting, familial hyperglucagonemia, or other PETs. [15], [16], [51], [72], [142], [152], [160], [161] In two studies, one involving 71 patients with fasting hyperglucagonoma at least two times the upper limit of normal (i.e., >120), [152] and the other [16] with 407 cases of glucagonoma from the literature, only 21% and 57% of the patients included, respectively, had a clinical glucagonoma syndrome. Therefore, hyperglucagonemia without the glucagonoma syndrome is not necessarily uncommon. In one series [152] the hyperglucagonemia occurred with polyfunctional PETs in 69% of the cases, especially with patients with Zollinger-Ellison syndrome, insulinomas, and carcinoid tumors. In almost all of these cases the hyperglucagonemia was not associated with the glucagonoma syndrome. In general, in these nonglucagonoma conditions causing hypergastrinemia, the plasma glucagon is not elevated above 500 pg/mL except in patients with cirrhosis. [51], [72], [142] For the occasional patient with glucagonoma with a fasting plasma glucagon concentration less than 1000 pg/mL that may overlap with other conditions, there is no proven provocative test that will clearly distinguish these conditions. [51], [72] Usually, however, these nonglucagonoma conditions can be excluded on clinical grounds. In various series from 0% to 20% of patients with glucagonomas had MEN I. [13], [14] and [15], [142], [143] These patients can be difficult to diagnose because they have minimal symptoms early in their disease course. [15] It is important to remember that necrolytic migratory erythema [162] can be observed in patients without an associated glucagonoma. The most common conditions this is reported to occur in are celiac disease, malabsorptive conditions, cirrhosis, malignancies, and pancreatitis. [162]

Somatostatinomas

Somatostatinomas are endocrine tumors of the pancreas or intestine that secrete excessive amounts of somatostatin which causes a distinct syndrome characterized by diabetes mellitus, gallbladder disease, diarrhea, and steatorrhea (Table 97-5).

SIGN/SYMPTOM	FREQUENCY (%)		
	Somatostatinoma *Pancreatic*	*Intestinal*	Somatostatinoma *Syndrome**
Diabetes mellitus	90–95	21	95
Gallbladder disease	90–94	43	68
Diarrhea	60–97	10–40	37
Steatorrhea	83	12	47
Hypochlorhydria	86	17	26
Weight loss	30–90	20–45	68

*Somatostatinoma refers to a pancreatic endocrine tumor which shows somatostatin-like immunoreactivity and may occur without (89%) or with (11%) the somatostatinoma syndrome which is due to somatostatin secretion by the tumor.[21,22]

Modified from refs. 20, 21, 23.

TABLE 97-5 Clinical Features of the Somatostatinoma Syndrome

In 1977 the first two cases of somatostatinomas were independently reported. [163], [164] On the basis of these reports and the known actions of somatostatin, it was proposed that the clinical somatostatinoma syndrome consisted of mild diabetes mellitus, gallbladder disease, weight loss, and anemia. [165] Diarrhea, steatorrhea, and hypochlorhydria became additional features as other cases have been described (see Table 97-5). [75], [166], [167]

There is no general agreement on the definition of what is a somatostatinoma. In the literature this term is generally used to indicate a PET possessing somatostatin-like immunoreactivity and it may (11% to 45%), or may not (55% to 89%), be associated with clinical symptoms due to ectopic release of somatostatin. [21], [22] and [23] In this chapter the term *somatostatinoma syndrome* will be used to indicate a somatostatin-producing PET associated with clinical features due to ectopic somatostatin release (see Table 97-5), and the term *somatostatinoma* will be used to include a PET containing somatostatin immunoreactivity.

Somatostatin is a tetradecapeptide originally isolated and purified from ovine hypothalamic tissue. [168] It inhibits the release of almost all other hormones, and has direct effects on a number of gastrointestinal functions including being a potent inhibitor of basal and pentagastrin- or meal-stimulated acid secretion, cholecystokinin-stimulated pancreatic enzyme secretion, and intestinal absorption of amino acids. [168], [169] A number of studies have suggested that somatostatin has a paracrine effect on antral gastrin release. [168] Somatostatin also has marked effects on gut motility and transit time. [168], [169]

Somatostatinomas primarily occur in the pancreas (46% to 75%) or in the upper small intestine (25% to 54%). [20], [21], [23], [75] In the pancreas 62% to 78% of the somatostatinomas occur in the pancreatic head. [20], [23], [75] Tumors not in the pancreas occur in the duodenum (43% to 90%), ampullary area (48%), jejunum (5%), or cystic duct area. [20], [23] Most somatostatinomas (>96%) are solitary and vary from less than 1 cm to 10 cm in diameter with a mean of 4 to 5 cm. [23], [75] Pancreatic tumors are larger than duodenal tumors (5 cm vs. 2 cm) ($P < 0.0001$), [23] and are more malignant than intestinal tumors (92% vs. 6%) in one study, [20] but not in another [23] (i.e., both 50%). With duodenal somatostatinoma the occurrence of lymph node metastases correlates with primary tumor size. [170] Electron microscopic studies show typical D-cell secretory granules in 52% to 89% of the tumors. [20], [23], [170]

The mean age of patients with pancreatic somatostatinomas or intestinal somatostatinomas is 50 years with a range of 30 to 84 years. [20], [75] In one series, [20] in the patients with pancreatic somatostatinomas there was a 2:1 female predominance whereas 60% of the patients with intestinal somatostatinoma are male; [20] however, in another review somatostatinomas occurred with equal frequency in both genders. [23]

In assessing the symptoms and signs due to somatostatinomas it is important to differentiate between those due to the PET per se, which are similar to any PET (pain, icterus, weight loss, hepatomegaly, abdominal tumor, etc.) from those that are due to the increased somatostatin secreted by the PET. In one large literature review of 173 cases of somatostatinomas, [23] only 18 cases (11%) were associated with the somatostatinoma syndrome (see Table 97-5). In one study [23], 93% of

patients with somatostatinomas had symptoms or signs of the condition; the most frequent being abdominal pain (40%), weight loss (26%), icterus (23%), abdominal tumor (12%), diarrhea (18%), nausea/vomiting (16%), and hepatomegaly (10%). Diabetes mellitus or glucose intolerance occurs in 63% to 90% of patients with somatostatinomas. [20], [21], [75] However, diabetes mellitus occurred in 90% to 95% of the patients with pancreatic somatostatinomas and only 21% of those with intestinal somatostatinomas (see Table 97-5). [20] The diabetes is usually mild and severe hypoglycemia and ketosis, although reported, are uncommon. [21], [27], [167] The development of diabetes mellitus or glucose intolerance is secondary to the ability of somatostatin to inhibit insulin and glucagon release [20], [21], [168] and the replacement of functional islet tissue by tumor. [75] This latter factor may be responsible for explaining why diabetes mellitus is more common in patients with pancreatic than intestinal somatostatinomas (see Table 97-5) [20] In patients with the somatostatinoma syndrome, 95% had diabetes mellitus (see Table 97-5). [23]

Gallbladder (cholelithiasis) disease is reported in 65% to 90% in patients with somatostatinomas. [20], [21], [75] It is reported in 90% to 94% of patients with pancreatic somatostatinoma and 43% of patients with intestinal somatostatinoma (see Table 97-5). [20] Cholelithiasis was present in 68% of patients with the somatostatinoma syndrome (see Table 97-5). [23] The high incidence of gallbladder disease in patients with somatostatinomas and the lack of occurrence in patients with other PETs suggests a causative association between the gallbladder disease and somatostatinoma. This conclusion is supported by studies which demonstrate that somatostatin analogs inhibit gallbladder emptying, alter the motility of the sphincter of Oddi, alter hepatic bile secretion, and have effects on the bile composition. [21], [168], [169], [171], [172] and [173] It is also supported by studies of patients with acromegaly or PETs treated long-term with octreotide. [169], [174], [175] The latter studies [169], [171], [175] demonstrate that 13% to 66% of patients treated long-term with octreotide develop gallstones.

Diarrhea and steatorrhea were reported in 36% to 90% of patients in various studies. [20], [21], [23], [75] The diarrhea characteristically consists of 3 to 10 foul-smelling stools per day and the steatorrhea varies from 20 to 76 g/d. [20], [75] Diarrhea and steatorrhea are much less common in patients with intestinal (10% to 40%) rather than pancreatic somatostatinomas (60% to 97%) (see Table 97-5). [20], [23] In some, but not all cases, the severity of the diarrhea and steatorrhea correlates with the size and degree of metastatic spread of the tumor and improves with tumor resection. [75] Somatostatin has been shown to inhibit pancreatic enzyme and fluid secretion as well as gallbladder motility. [168], [169] In addition somatostatin also inhibits intestinal absorption of lipid, [176] D-xylose, vitamin B_{12}, and folate. These biologic actions of somatostatin may be responsible for the maldigestion, steatorrhea, and diarrhea observed in these patients. [75] The fact that patients with intestinal somatostatinomas have less diarrhea or steatorrhea has been attributed to their lower plasma somatostatin levels or the lack of local effects of the somatostatinoma within the pancreas (See Table 97-5). [20] Diarrhea is present in 37% and steatorrhea in 47% of patients with the somatostatinoma syndrome (see Table 97-5). [23]

Hypochlorhydria was found in 33% of patients in one study, [75] in 53% of patients in a second study, [20] and 70% in a third study. [23] Hypochlorhydria occurred in 86% of patients with pancreatic somatostatinoma and 17% of patients with intestinal somatostatinoma (see Table 97-5). [20] Hypochlorhydria was present in 26% of patients with the somatostatinoma syndrome (see Table 97-5). [23] Infusion of somatostatin has been shown to inhibit gastric secretion in normal human subjects as well as basal and food-stimulated gastrin release. [168] As with the glucagonoma syndrome, weight loss is reported to be common in patients with somatostatinomas. [21] Weight loss was reported in 32% to 90% of patients with pancreatic tumors and in 20% to 45% of patients with intestinal tumors [23], [75] (see Table 97-5), with weight loss ranging from 9 to 21 kg. Weight loss was reported in 68% of patients with somatostatinoma syndrome (see Table 97-5). [23] The weight loss may be secondary to the diarrhea and malabsorption. Other less common features reported in patients with somatostatinoma include mild to moderate anemia in 15% to 67% of patients with somatostatinomas, [23], [75] and in 21% of patients with the somatostatin syndrome. [23] Also, the occurrence of various associated endocrine disorders is reported in patients with somatostatinomas. [75] In one study, four patients were reported to have hypoglycemic episodes with normal or elevated plasma insulin concentrations and were iagnosed as having insulinomas. [75] In two patients with elevated plasma insulin concentrations the tumors did not contain increased amounts of insulin, only somatostatin. Tumor tissue was not available from the other two cases; however, immunocytochemical studies demonstrated mostly D-cells (somatostatin-producing cells) with some ß-cells (insulin-producing) in one patient and 20% to 30% D-cells with 30% to 40% ß-cells in another patient. [75]

The diagnosis of somatostatinoma has been applied to numerous PETs in the literature based only on immunohistochemical results which, in most cases (55% to 89%), is not associated with the somatostatinoma syndrome due to ectopic somatostatin release. [21], [22] and [23] The diagnosis of the somatostatinoma syndrome requires the demonstration of increased concentrations of somatostatin-like immunoreactivity (SLI) in the resected tumor and the plasma as well as at least some features of the appropriate clinical syndrome (see Table 97-5). The establishment of SLI only in a resected tumor is not sufficient for the diagnosis of the somatostatinoma syndrome. Minimal increases in plasma/SLI levels need to be interpreted with caution because they can occur in nonendocrine disorders. [21] Furthermore, while plasma SLI levels are frequently elevated with pancreatic tumors, in small intestinal somatostatinomas the SLI levels are frequently normal. [21], [22] SLI is frequently detected as a subpopulation of cells in a variety of PETs, especially nonfunctional tumors. [21], [177] High plasma SLI levels have been reported with endocrine tumors outside of the pancreas or intestine, as with small cell lung cancer, bronchial oat cell carcinoma, medullary thyroid carcinoma, pheochromocytomas, and other catecholamine-producing extra-abdominal paragangliomas. [21] It is likely in the future that the syndrome will be diagnosed earlier because of the increased clinical awareness of the somatostatinoma syndrome (see Table 97-5) and the greater availability of reliable assays for the determination of SLI in blood. Presently these assays are complicated by the need for extraction of the plasma and are not widely available.

The diagnosis of somatostatinoma at a time when plasma SLI concentrations are only marginally elevated and the tumor has not yet metastasized will require the development of reliable provocative tests. Tolbutamide and arginine have been reported to stimulate plasma SLI increases; [75], [178] however, arginine is a well established stimulant for normal D-cells and thus is unlikely to differentiate normal from supranormal secretion. [75] Furthermore even though tolbutamide stimulates SLI increases in animals, [178] it is reported not to cause changes in plasma SLI concentrations in normal human volunteers. [179] Until an adequate provocative test is developed, if the somatostatinoma syndrome is to be diagnosed earlier, it may be necessary to determine plasma SLI concentrations on all patients with diabetes mellitus without a family history and with any other clinical features of the somatostatinoma syndrome, or all patients with unexplained diarrhea. [166]

In most studies somatostatinomas have been found more or less by accident. [75] Because most somatostatinomas (i.e., 89%) [23] do not produce the somatostatinoma syndrome and the most common presenting symptoms are nonspecific (abdominal pain, weight loss, icterus, diarrhea, nausea/vomiting), [23] the correct diagnosis is rarely suspected prior to the discovery of a PET at laparotomy for cholecystectomy, during imaging studies for nonspecific complaints, or after a biopsy for metastatic liver disease discovered during a workup for nonspecific gastrointestinal symptoms. [20], [21], [23], [75] The presence of psammoma bodies in a duodenal tumor should particularly raise the suspicion that the tumor could be a somatostatinoma. [21], [23], [180] These lesions are found in approximately one half of duodenal somatostatinomas, [21], [22] and [23] but are uncommonly found (i.e., 2% to 5%) in pancreatic somatostatinomas or in other types of duodenal carcinoids or duodenal gastrinomas. [21], [180] Duodenal somatostatinomas are increasingly associated with von Recklinghausen's disease. [21], [22], [55] Duodenal somatostatinomas in patients with von Recklinghausen's disease are similar to duodenal somatostatinomas in patients with sporadic duodenal somatostatinomas in that they are rarely associated with symptoms of the somatostatinoma syndrome (i.e., 2%), infrequently cause elevated plasma SLI levels, often contain psammoma bodies (37% to 66%), [22], [23] and are less likely to be malignant (30% duodenal vs. 70% for pancreatic somatostatinomas). [22], [23]

GRFomas

GRFomas are endocrine tumors that frequently originate in the pancreas but also occur in other extra-pancreatic sites and secrete excessive amounts of growth hormone-releasing factor (GRF) that causes acromegaly.

Tumors releasing growth hormone-releasing factor (GRFomas) were described in 1982. [181], [182] Growth hormone-releasing factor is a 44-amino acid peptide originally isolated from human PETs causing acromegaly and is structurally similar to vasoactive intestinal polypeptide. [21], [181] The true frequency of this syndrome is still not established but a review in 1997 included 40 cases. [77] In one immunocytochemical study [183] of PETs, 4 of 9 (44%) contained GRF-like immunoreactivity, although only one patient had acromegaly. In another study [184] 6 of 24 (25%) PETs, of which 16 were associated with various clinical syndromes (8 with insulinoma; 5, gastrinoma; 2, VIPoma-2; 1, Cushing syndrome), were found to have GRF-immunoreactive material. None of these patients had clinical acromegaly. GRFomas occur in the pancreas in 29% to 30% of cases; 47% to 54% occur in lung tumors with the majority in the right lung; 8% to 10% occur in the small intestine; and up to 13% occur in other sites including adrenal gland, foregut, and retroperitoneum (see Table 97-1). [21], [77], [185], [186] The patients were 15 to 66 years of age with an average age of 38 to 39 years. [77], [185], [186] A female predominance (78%) is seen with pancreatic GRFomas as well as all GRFomas (73%). [77], [185], [186] The clinical features can be divided into three categories: acromegalic features due to ectopically released GRF causing excess growth hormone secretion; clinical features due to hormones other than GRF co-released, such as gastrin causing Zollinger-Ellison syndrome or ACTH causing Cushing syndrome; and local symptoms due to mass effect of the hormone. [21], [185], [186] The mean duration from symptom onset to diagnosis is 5 to 6 years. [185], [186] The acromegalic features of GRFomas are indistinguishable from those of classical acromegaly. [21], [185], [186] Patients usually have a large pancreatic tumor (>6 cm; range 1–25 cm) and in 33% to 39% of cases it is metastatic at the time of

diagnosis. [77], [185] The majority of pancreatic GRFomas occur in the pancreatic tail. [77], [186] In one series [185] metastatic disease was present in 30% of the patients with pancreatic GRFomas and two of the three patients with intestinal GRFomas. [186] No relationship between the presence or absence of metastases, tumor size, or plasma GRF level was found. [186] Multiple pancreatic GRFomas occurred in 30% in one series, [186] usually in patients with MEN I. [21] Forty percent of the patients with pancreatic GRFomas have associated Zollinger-Ellison syndrome; 40% have Cushing syndrome, and 33% have associated MEN-I syndrome. [27], [186]

GRFomas are usually suspected in a patient with acromegaly and an elevated growth hormone level with hepatic metastases or with abdominal complaints. [21], [76], [181], [182], [185], [186] Pancreatic GRFomas are often associated with a gastrinoma and Zollinger-Ellison syndrome or Cushing syndrome. Therefore, any patient with Cushing syndrome or peptic ulcer symptoms, diarrhea, a pancreatic mass, or with symptoms suggestive of chronic esophageal reflux who presents with acromegalic features, should be suspected of having a GRFoma. Furthermore, pancreatic GRFomas have occurred in a number of patients with the MEN-I syndrome. Any patient with acromegaly with hyperparathyroidism or a family history of MEN-1 should also be suspected of having a GRFoma. The diagnosis should be suspected in any patient with acromegaly without a pituitary adenoma, with acromegaly associated with hyperprolactinemia which is seen in 70% of GRFomas, with paradoxical growth hormone response to thyroid-stimulating hormone (TSH), or during an oral glucose tolerance test. [21], [77], [185], [186] A GRFoma is an uncommon cause of acromegaly. In one study [187] of 177 unselected patients, none had acromegaly. The diagnosis is confirmed by performing a plasma assay for GRF and for growth hormone. [21], [76], [77], [185], [186] Elevated plasma levels of growth hormone (usually >5 µg/L in men and 10 µg/L in women) and an elevated plasma GRF confirm the diagnosis. A recent review proposes an empirical diagnostic threshold value for GRFomas of 300 pg/mL for the plasma GRF level. [21] Healthy subjects have GRF levels less than 50 to 100 pg/mL [77] and patients with pituitary adenomas causing acromegaly have plasma GRF levels less than 200 pg/mL. [21], [187] In addition to growth hormone and GRF, in patients with GRFomas plasma levels of insulin-like growth factor 1 (IGF-1) are elevated similar to patients with acromegaly due to a pituitary adenoma. [185]

Nonfunctioning PETs

Nonfunctioning PETs are endocrine tumors that originate in the pancreas and either do not secrete any peptide or their secreted products do not cause clinical symptoms. The symptoms that occur are due to the effects of the tumor per se.

Nonfunctioning PETs differ from the PETs in their clinical presentation, as their symptoms are entirely due to the tumor per se and not to the secreted products. [1], [6], [20], [28], [29] Nonfunctional PETs frequently release peptides which cause no clinical symptoms including chromogranin A (69% to 100%), chromogranin B (100%), pancreatic polypeptide (50% to 100%), a-hCG (40%), ß-hCG (20%), synaptophysin, and neuron-specific enolase; some release neurotensin. [1], [3], [20], [43], [45], [50] Immunocytochemically, these tumors often contain multiple peptides. [1], [38] In one study of 30 such tumors, [38] 50% had immunoreactivity for insulin, 30% for glucagon, 40% to 75% for PP, 13% for somatostatin, and 13% for none of these. Nonfunctional PETs are usually solitary, large tumors (72% >5 cm in one study [188]) and occur primarily in the pancreatic head (14 were in the head, two were in the body, and three were in the tail in one study [188]). Nonfunctioning PETs are frequently diagnosed after the patient presents with symptoms or signs of metastatic tumor in the liver (cachexia, abdominal pain, hepatomegaly) and a liver biopsy is performed which reveals metastatic endocrine tumor. [1], [20], [28], [29] Infusions of PP into animals has been shown to have numerous effects including inhibition of pancreatic secretion, a relaxant effect on the gallbladder, weak inhibition of pentagastrin-stimulated acid secretion, and various stimulating effects on gastrointestinal motility. [189] At present it is unclear why patients with PETs with very high plasma concentrations of PP do not have symptoms due to the increased PP per se. Typically the patient with a nonfunctioning PET is 40 to 60 years of age. [1], [20], [28], [29] They occur equally in men and women. [1], [28], [29], [188] Patients with nonfunctioning PETs frequently present with abdominal pain (36% to 56%), 28% to 40% with jaundice, 24% to 46% with weight loss, 8% to 40% with abdominal masses, and in 16% of the tumors were found incidentally at surgery in asymptomatic patients. [29], [188] In most series, [1], [28], [29], [190] but not all, [191], [192] more than 60% (range, 64%–92%) of patients with nonfunctioning PETs have metastatic disease at the time of diagnosis. [1], [20], [29], [188] The median delay in time from first symptoms to diagnosis varies from 0.5 to 2.7 years. [1]

Nonfunctioning PETs are usually not differentiated from any other malignant tumor of the pancreas prior to histological studies. [1], [20] In one series [28] the diagnosis of a nonfunctioning PET was not made in a single patient prior to surgery. Furthermore, 20% of the patients were asymptomatic and the tumors were found incidentally during other operative procedures. [28] Any patient with a long survival (>5 years) previously diagnosed as having metastatic pancreatic adenocarcinoma should be suspected of possibly having a nonfunctioning PET. An elevated plasma chromogranin A or B level (69% to 100%), PP (50% to 100%), a-hCG (40%), ß-hCG (20%), neuron-specific enolase (31%), or positive somatostatin receptor scintigraphy (SRS) is strong evidence to suggest that a pancreatic mass is a PET. [1], [3], [20], [45], [50], [193], [194] In 53 patients with adenocarcinoma of the pancreas, none had elevated plasma levels of PP. [20] However, elevated levels of PP do not establish the diagnosis of a PPoma even when a pancreatic mass is present because plasma PP levels are increased in 22% to 71% of patients with functional PETs [1], [41], [195], [196] as well as in nonpancreatic carcinoid tumors. In one study [196] plasma PP levels were greater than 1000 pg/mL in 32% of patients with gastrinomas, 21% with insulinomas, 57% with glucagonomas, 74% with VIPomas, 33% with somatostatinomas, and 45% with carcinoid tumors. Furthermore, plasma PP elevations can occur with renal failure, older age, postbowel resection, alcohol abuse, certain infections or inflammatory conditions, acute diarrhea, diabetes, pancreatitis, or with eating. [196] To improve the specificity an atropine suppression test has been proposed. [196] In a study of 48 patients with elevated plasma PP levels, atropine (1 mg IM) did not suppress the levels in any of 18 patients with PETs, but did suppress it in all patients without PETs. In another study of 28 pancreatic ductular cancers [193] none were positive with SRS whereas 65% of proven PETs were positive. Furthermore, 42% of long-term survivors of pancreatic adenocarcinoma (>3 years) were positive by SRS, suggesting they were really PETs that had been misdiagnosed. [193] In a study of 61 nonfunctional PETs, [192] 56 (92%) were well differentiated, 5 (8%) poorly differentiated, and 34 (56%) demonstrated metastases. The presence of vascular or perineural invasion, a K_i 67 proliferative index greater than 2%, a mitotic rate =2, size =4 cm, capsular penetration, nuclear atypia, lack of progesterone receptors, and presence of calcitonin all correlated with malignancy. [192] These factors had prognostic significance for survival. [192] At present there are no data that suggest that nonfunctioning PETs producing PP or other peptides differ in biologic behavior from tumors not producing these peptides. [190]

Other Tumors (Cushing Syndrome, etc.)

Cushing syndrome associated with a PET (ACTHoma) is usually [83], [164], [197], [198] and [199] recognized in patients with another symptomatic PET. In various studies 4% to 16% of all cases of ectopic Cushing syndrome were due to PETs secreting adrenocortico- tropic hormone (ACTH). [84] Cushing syndrome occurs in 19% of patients with MEN I with Zollinger-Ellison syndrome, is due to pituitary production of ACTH, and the disease in these patients is usually mild. [83] Cushing syndrome occurs in 4% to 5% of patients with sporadic Zollinger-Ellison syndrome. [8], [83], [197], [198] and [199] In patients with the sporadic form of Zollinger-Ellison syndrome, the symptoms of Cushing syndrome are severe, usually occur in patients with metastatic disease, and the Cushing syndrome is due to ectopic production of ACTH. [83], [198], [199] In one study [198] each patient with ectopic Cushing syndrome resulting from a PET had metastatic disease in the liver, although rare patients with nonmetastatic disease with Zollinger-Ellison syndrome with Cushing syndrome due to ACTH release by the gastrinoma are reported. [85] These patients usually respond poorly to chemotherapy. [83] In one prospective study [8] the development of Cushing syndrome in patients with gastrinomas was an independent predictor of poor survival, with patients living a mean of only 1.7 years after its onset. [83] The occurrence of Cushing syndrome only as a clinical manifestation of a PET is uncommon. [200]

Paraneoplastic hypercalcemia due to a PET releasing PTH-related peptide (PTHrP), a PTH-like immunoreactive material, or an unknown hypercalcemic substance has been reported rarely. [80], [81], [201], [202] In most, [80], [201], [202] and [203] but not all, [80], [117] cases describing paraneoplastic hypercalcemia with a PET [201], [202] and [203] the tumors are large and are metastatic to the liver at the time of diagnosis. In a review [80] of 23 cases of PETs associated with hypercalcemia, 30% occurred in patients with VIPomas. The tumors were generally large (7–18 cm) and in 7 of 8 patients PTHrP was demonstrated immunohistochemically. [80]

It has been proposed that there is a neurotensinoma syndrome associated with PETs. [20], [27], [43], [204], [205] and [206] Neurotensin is a 13-amino acid peptide first extracted from bovine brain and subsequently also isolated from the human gastrointestinal tract. [207] Neurotensin has a number of pharmacological effects including hypotension, tachycardia, cyanosis, stimulating pancreatic protein and bicarbonate secretion, affecting intestinal motility, and stimulating jejunal and ileal fluid and electrolyte secretion. [207] The clinical features of the patients described with neurotensinomas with PET include diarrhea with hypokalemia, weight loss, and in some cases diabetes, cyanosis, hypotension, and flushing. [20] In the six cases included in one review, [20] three were cured by tumor resection and three responded to streptozotocin treatment. [20], [27], [204], [205] and [206] Others [41], [204] have raised the question of whether a specific neurotensinoma syndrome actually exists. In one study [204] of 180 patients with functional PETs an elevated plasma neurotensin level was found in six patients with VIPomas and the clinical symptoms of those patients did not differ from those without an elevated plasma neurotensin level. In a second study, [41] 19% of patients with Zollinger-Ellison syndrome were found to have an elevated plasma neurotensin level and their symptoms did not differ from those without an elevated plasma neurotensin level, therefore it is not apparent that a distinct

neurotensinoma syndrome exists.

PETs causing the carcinoid syndrome can occur. [78], [79] These tumors are characteristically malignant (68% to 88%) and are large. [56], [78], [79] Pancreatic PETs [24] are foregut carcinoids which may lack dihydroxyphenylalanine (DOPA) decarboxylase, the enzyme that converts 5-hydroxytryptophan to serotonin (5-hydroxytryptomine); [24] however, 84% of patients with pancreatic PETs associated with the carcinoid syndrome have increased urinary 5-hydroxyindule acetic acid (5-HIAA) levels which can be used for their detection. [56]

TUMOR LOCALIZATION

The techniques used for tumor localization and the rationale for using various localization methods is similar for all the PETs. Tumor localization studies need to be undertaken for a number of reasons once the proper radioimmunoassays and functional studies establish the particular type of PET. For appropriate management of the patient the tumor's location and extent must be established. [208], [209] The only possible long-term cure of these syndromes is by surgical excision. To establish whether the tumor is potentially resectable, tumor localization studies are essential. In all cases except insulinoma, the majority, if not all of these tumors if followed long enough, may be malignant, and therefore if metastatic spread is not present at the time of diagnosis, an attempt should be made to surgically cure these patients. Gastrinomas and insulinomas frequently present as small tumors. [42], [52], [210] Up to 50% of PETs that occur frequently extrapancreatically, such as gastrinomas, [12], [42], [210], [211] and 10% to 20% of PETs that occur almost entirely within the pancreas, such as insulinomas, [51], [95] are not found at surgery. Therefore, detailed localization studies may help the surgeon in finding the tumor. The tumor can also be multiple with multifocal tumors reported to occur in 10% of insulinomas [51], [98] and up to 50% of some PETs such as gastrinomas. [12], [211] Furthermore, except for insulinoma, the other PETs are frequently diagnosed only after metastatic spread to the liver has occurred. [12], [16], [17], [20], [23], [51], [75], [141] To determine whether treatment should be started for the metastatic disease and to assess the results of such treatment, tumor localization methods are essential. [2], [12], [212], [213] and [214]

Abdominal ultrasound, [2], [12], [208], [215], [216], [217], [218], [219] and [220] computed tomography (CT scan) (Fig. 97-3, Fig. 97-4, Fig. 97-8, Fig. 97-9), [2], [12], [208], [215], [216], [217] and [218], [220], [221], [222], [223], [224] and [225] selective abdominal angiography (Fig. 97-5, Fig. 97-6), [2], [12], [208], [218], [220], [223], [226] magnetic resonance imaging (MRI) (see Fig. 97-4, Fig. 97-7), [2], [208], [215], [216], [225], [227], [228] radiolabeled SRS (Fig. 97-7, Fig. 97-8), [215], [221], [229], [230], [231], [232], [233], [234] and [235] endoscopic ultrasound (see Fig. 97-9), [215], [221], [222], [235], [236], [237], [238] and [239] selective venous sampling for hormones from portal venous tributaries using a transhepatic approach (i.e., portal venous sampling, PVS) (Fig. 97-10), [12], [140], [240], [241], [242] and [243] hepatic venous sampling for hormones after intraarterial injection of secretin (gastrinomas) or calcium (insulinomas) (see Fig. 97-10), [243], [244] and [245] and intraoperative ultrasonography (Fig. 97-11) [216], [228], [246], [247] have all been reported to be useful for localizing PETs.

FIGURE 97-3. Computed tomographic (CT) scan in a patient with an insulinoma. The insulinoma is in the pancreatic tail (*arrow*). The splenic vein courses around the tumor. The CT scan with intravenous contrast will localize 17% to 40% of insulinomas.

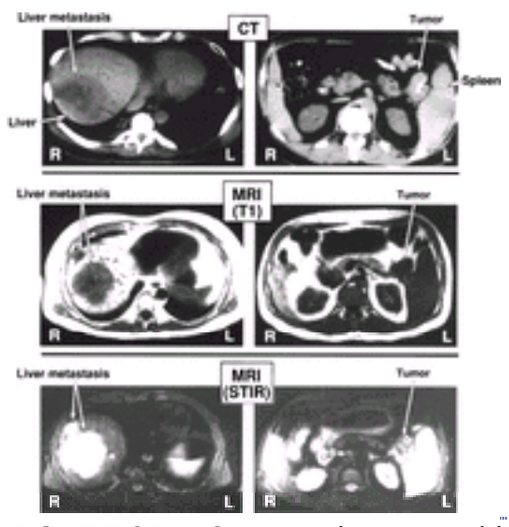

FIGURE 97-4. Computed tomographic (CT) scan (**top panels**), magnetic resonance imaging (MRI) T-1 weighted image (**middle panels**), and a short inversion time-inversion recovery (STIR) MRI (**bottom panels**) in a patient with a pancreatic endocrine tumor (PET) metastatic to the liver producing parathyroid hormone-related peptide (PTHrP) causing hypercalcemia. In the left panels the CT scan (**top**) demonstrates a solitary, large metastasis in the right lobe of the liver (*arrow*) which is not well outlined and on the T-1 (**middle**) and the STIR (**bottom**) MRIs two metastases are clearly seen (*arrow*) in the right lobe of the liver. In the **right panels** more caudal views show the primary tumor in the pancreatic tail. The CT scan (**top, right**) demonstrates clearly the 4-cm pancreatic tail tumor with central calcification, whereas it is less well seen on the MRI T-1 (**middle**) and STIR images (**right**). This figure demonstrates that metastases from PETs are often much more easily identified on MRI, especially the STIR images, because the tumor has a characteristically bright appearance. [12], [364] In contrast the primary tumor is frequently seen better on the CT scan.

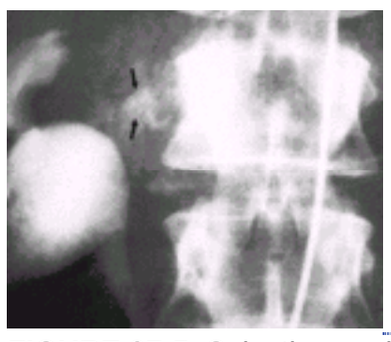

FIGURE 97-5. Selective angiographic localization of the pancreatic endocrine tumor (PET) in a patient with insulinoma. The insulinoma (1 cm) is localized in the pancreatic head by a selective celiac artery injection. Insulinomas, as other PETs, are characteristically hypervascular as demonstrated in the insulinoma in this patient. Because of this hypervascularity even small pancreatic primary lesions are frequently only seen on angiography. Angiography localizes 60% of insulinomas and 68% of the primary tumors in other PET syndromes.

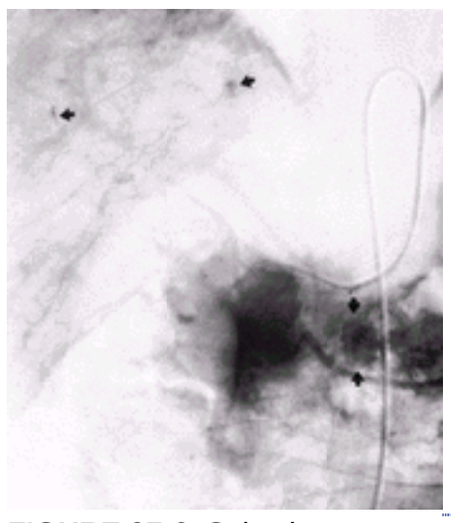

FIGURE 97-6. Selective common hepatic artery injection in a patient with metastatic insulinoma. The primary insulinoma is in the pancreatic head (*double arrows*) and two hepatic metastases are located in the liver (*small arrows*). Tumors are malignant in only 10% to 15% of patients with insulinomas, whereas in other pancreatic endocrine tumors (PETs) the majority are malignant. For the conventional localization methods (ultrasound, computed tomography scan, magnetic resonance imaging [MRI], angiography), angiography and MRI are the most sensitive modalities for localizing liver metastases (see Table 97-6). (Courtesy of Dr. Jeffrey A. Norton, University of California, San Francisco.)

TABLE 97-6 Studies Assessing the Ability of Various Modalities to Localize Insulinomas and Other Pancreatic Endocrine Tumors

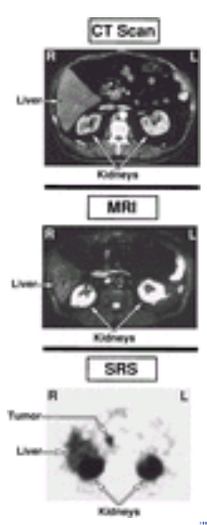

FIGURE 97-7. Comparison of the ability of computed tomography (CT) scan, magnetic resonance imaging (MRI), and somatostatin receptor scintigraphy (SRS) to localize a pancreatic endocrine tumor (PET). This patient previously had a duodenal PET secreting chromogranin A and gastrin resected and underwent reexamination when the serum chromogranin A level increased 1-year postresection. Neither the CT scan (**top**) nor the MRI (**middle**) demonstrated a lesion, whereas the SRS demonstrated a tumor in the pancreatic head area. At surgery a 1.5-cm pancreatic head lymph node containing tumor was found and resected. This study demonstrates the greater sensitivity of SRS for localizing all noninsulinoma PETs [169], [230], [231], [232] and [233] and why it is now the initial imaging study of choice in these patients.

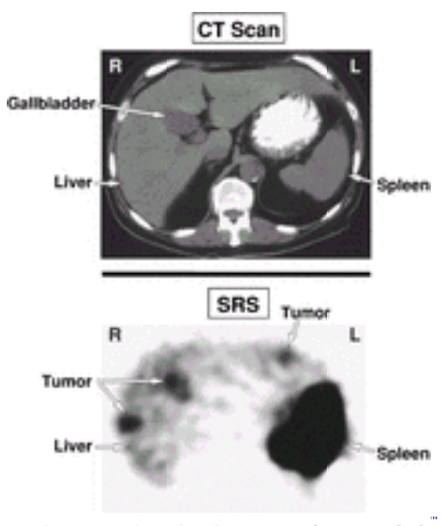

FIGURE 97-8. Comparison of the ability of computed tomography (CT) scan and somatostatin receptor scintigraphy (SRS) to localize metastatic pancreatic endocrine tumor (PET). This patient had right upper quadrant pain with an elevated serum chromogranin level and gastrin level. The CT scan (**top**), magnetic resonance imaging, ultrasound, and angiography were negative; however, the SRS (**bottom**) showed three liver metastases. A percutaneous biopsy demonstrated metastatic PET. This study demonstrates the greater sensitivity of SRS for detecting metastatic disease, both in the liver and other distant sites, in patients with malignant PETs. [169], [230], [231], [232] and [233], [251], [255], [258]

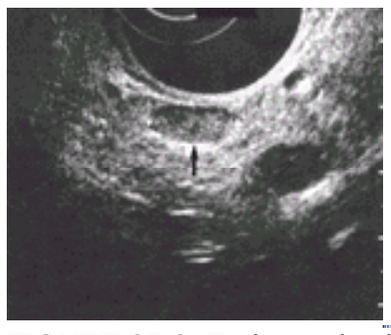

FIGURE 97-9. Endoscopic ultrasound of a patient with an insulinoma. This image of the head of the pancreas was taken using a 12-MHz probe at the level of the duodenal bulb. It shows a 1.3 × 0.7 cm sharply demarcated relatively hypoechoic nodule (*arrow*) which was an insulinoma that was later enucleated at surgery. The splenic vein is seen below the tumor in the 5 o'clock position. The patient was a 29-year-old woman with clinical history and laboratory values diagnostic of insulinoma, but with negative dynamic computed tomography and selective angiography studies. This result shows the sensitivity of endoscopic ultrasound for localizing even small, intrapancreatic endocrine tumors. (Courtesy of Dr. Charles J. Lightdale, Director of Clinical Gastroenterology, Columbia-Presbyterian Medical Center, New York, NY.)

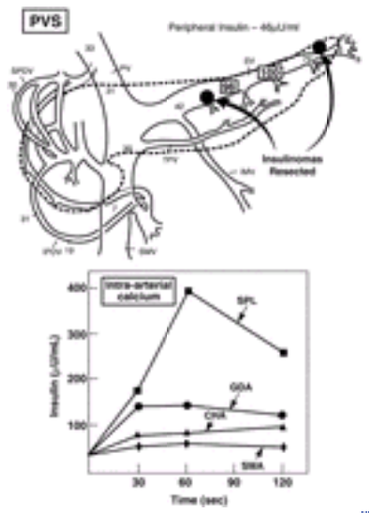

FIGURE 97-10. Functional location of an insulinoma by selective venous sampling for insulin concentrations in portal venous tributaries (**top**) or by selective hepatic venous sampling for insulin concentrations after selective intraarterial calcium injections (**bottom**) in a patient with multiple insulinomas in the setting of multiple endocrine neoplasia type I (MEN I). In the **top panel** a sampling catheter was passed across the liver and venous samples obtained from the indicated locations and simultaneous peripheral venous samples were obtained which were assayed for insulin concentration. The step-up to 100 μU/mL is seen in the midsplenic vein, suggesting a tumor in the pancreatic body or tail. (Abbreviations: *PV,* portal vein; *SMV,* superior mesenteric vein; *SPDV,* superior pancreaticoduodenal vein; *IPDV,* inferior pancreaticoduodenal vein; *IMV,* inferior mesenteric vein; *TPV,* transverse pancreatic vein; *SV,* splenic vein.) In the **bottom panel** calcium gluconate (0.01 mEq Ca^{2+}/kg) [244] was selectively injected into the indicated arteries and hepatic venous samples obtained prior to injection and at 30, 60, and 120 seconds postinjection and analyzed for insulin concentration. There was a marked increase after the splenic artery injection (*SPL*), whereas after injection into the gastroduodenal artery (*GDA*), common hepatic artery (*CHA*), and superior mesenteric artery (*SMA*) there was a much smaller increase. Two insulinomas were found at surgery (see location by *arrows* in **top panel**) in the pancreatic body and tail, which is in the area localized by the portal venous sampling (PVS) as well as the area supplied by the splenic artery. The intraarterial calcium method is more sensitive than PVS [243], [244] and has the advantage of requiring less expertise, can be done at the time of angiography, and therefore, a separate procedure is not required. In some studies [244] the intraarterial calcium is the most sensitive modality for localizing small insulinomas. [Courtesy of Dr. John Doppman, NIH.]

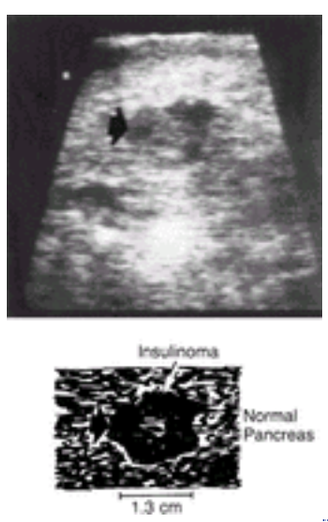

FIGURE 97-11. Intraoperative ultrasound localization of an insulinoma. **Top:** An intraoperative ultrasound image of an insulinoma (*arrow*). The image was taken with a 10-MHz transducer. **Bottom:** A drawing of the ultrasound image showing a tumor (*arrow*). A 1.3-cm, nonpalpable insulinoma was found at surgery in the pancreatic head. Studies [216], [228] demonstrate intraoperative ultrasound will localize 88% of insulinomas. In this patient the use of intraoperative ultrasound guided the successful enucleation of this tumor which could not be identified at surgery by any other means. (Courtesy of Dr. Jeffrey A. Norton, University of California, San Francisco.)

Small PETs (<1 cm) have been the most difficult to localize. The ability of conventional imaging studies (ultrasound, CT scanning, MRI scanning, and selective angiography) to image a PET have been shown to be dependent on the size of the tumor. [224], [226], [227] and [228] For example, using any of the imaging modalities (ultrasound, CT scanning, selective angiography, or MRI) less than 10% of tumors smaller than 1 cm, 30% to 40% of tumors 1 to 3 cm, and 70% to 80% of tumors larger than 3 cm were detected. [12], [248] Therefore most VIPomas, glucagonomas, and somatostatinomas which are usually clinically suspected only when the tumor is large (frequently >4 cm), will be detected. One study demonstrates that small PETs (gastrinomas) (<1 cm) also were less frequently detected than large tumors by SRS. [231] Table 97-6 summarizes the results of studies on the ability of the various modalities to localize insulinomas which are characteristically small (<1 cm), as are most duodenal gastrinomas at the time of clinical presentation compared to other PETs which are frequently larger. [15], [16], [17] and [18], [23], [70], [195] The results with insulinomas are presented separately, not only because of their smaller size at presentation, but also because they are almost exclusively within the pancreas, whereas gastrinomas, VIPomas (in children), and somatostatinomas can frequently be extrapancreatic. [16], [17], [23], [52], [69], [231], [249] Ultrasound, CT scanning (see Fig. 97-3), and MRI (10% to 30%) are relatively insensitive, whereas selective abdominal angiography 60% (see Fig. 97-5, Fig. 97-6), selective PVS for insulin 80% (see Fig. 97-10), intraoperative ultrasound 89% (see Fig. 97-11), endoscopic ultrasound 82% (see Fig. 97-9), and hepatic venous sampling for insulin after calcium injection 94% (see Fig. 97-10) are sensitive methods to localize insulinomas. Insulinomas differ from other PETs in that they possess a low density of somatostatin receptors that bind octreotide with high affinity (subtypes 2 and 5, sst_2, sst_5). This makes SRS much less sensitive in localizing insulinomas (i.e., 25%, see Table 97-6). [209], [232], [233] Results for localization of other PETs except insulinomas are listed in Table 97-6 and compared to the values for insulinomas. These results are primarily based on studies of gastrinomas because the other PETs are uncommon, and frequently the patients with these tumors present with advanced metastatic disease so that routine surgical exploration is not performed.

Similar localization data involving large numbers of patients are not available with other PETs within the pancreas. However, because VIPomas, GRFomas, and

glucagonomas are almost entirely intrapancreatic it is likely that similar results to those for insulinomas of similar size will be obtained except for the SRS, which will be higher in these tumors. For somatostatinomas, which are frequently extrapancreatic, it is likely that results similar to those obtained with gastrinomas will be found. Ultrasound and MRI are relatively insensitive at localizing noninsulinoma PET primaries (24% to 25%); however, CT localizes 50%, angiography finds 68%, [12], [208], [226] selective venous sampling for gastrin 70%, [208], [240], [243] intraoperative ultrasound 91%, [208], [216], [246] endoscopic ultrasound 70%, [208], [236] and SRS 70%. [215], [230], [234], [235], [250], [251]

All PETs are hypervascular and therefore angiography is frequently the most sensitive conventional imaging modality. [208], [226], [230] Figure 97-5 and Figure 97-6 show the typical hypervascular appearance of both a primary PET (insulinoma) (see Fig. 97-5) and hepatic metastases from a malignant insulinoma (see Fig. 97-6).

Two newer localization methods that are being increasingly used in patients with PETs are SRS and endoscopic ultrasonography (see Table 97-6). PETs, in addition to a number of other neuroendocrine tumors (carcinoids, medullary thyroid cancer, pituitary adenomas, small cell lung cancer, paragangliomas) and other nonendocrine tumors (lymphomas [Hodgkin's disease, non-Hodgkin's lymphoma], central nervous system tumors [meningiomas, astrocytomas], breast cancer, prostate cancer) and granulomatosis diseases (sarcoidosis, tuberculosis, Wegener's granulomatosis), frequently possess high densities of somatostatin receptors which can be used for localization studies with radiolabeled somatostatin analogs. [169], [232], [233] For SRS, both [^{111}In-DTPA-DPhe [1]]octreotide and [iodine 123 (^{123}I)-Tyr [3]]octreotide have been used [169], [233] and in the United States the former agent is now increasingly used for tumor localization studies. [^{111}In-DTPA-DPhe [1]]octreotide has the advantage over [^{123}I-Tyr [3]]octreotide in that it is primarily excreted in urine instead of bile, allowing better visualization of right upper quadrant masses; has a larger half-life (2.8 days vs. 13.2 hours) allowing longer scanning time and is easier to label. [169], [233], [252] In vitro autoradiographic studies demonstrate 100% of gastrinomas, glucagonomas, nonfunctional PETs, 61% to 67% of insulinomas, 88% to 96% of carcinoids, and 0% of pancreatic adenocarcinomas bind radiolabeled octreotide. [230], [232], [233] Cloning studies demonstrate that there are five subtypes of somatostatin receptors and that radiolabeled octreotide binds with high affinity with somatostatin receptor subtypes 2 and 5 to subtype 3 with intermediate affinity and to subtypes 1 and 4 with low affinities. [169], [253] The overexpression of somatostatin receptors seen on autoradiography in PETs is consistent with the results of SRS in various studies. SRS visualizes a tumor in 87% to 96% of patients with a carcinoid tumor, 73% to 100% with a gastrinoma, 46% to 61% with an insulinoma, 82% to 89% with a nonfunctional PET, 100% with a glucagonoma, and 50% to 80% with a VIPoma in two reviews consisting of 477 patients. [232], [233] In a comparative study [230] of conventional modalities and SRS in 80 patients with gastrinomas, SRS identified a possible primary tumor in 58% and was superior ($P < 0.001$) to angiography (28%), MRI (30%), CT scan (31%), or ultrasound (9%). It was equal to the combination of all conventional imaging studies combined (48%). Furthermore, SRS [230] only identified a primary tumor in 20% of patients. Another study [231] confirms the greater sensitivity of SRS over conventional imaging studies, but it demonstrates that SRS can also frequently miss (>50%) PETs under 1 cm in diameter. The former study [230] concluded that SRS was now the imaging study of choice to localize noninsulinoma PETs. Figure 97-7 demonstrates the increased sensitivity of SRS over the CT scan or MRI in detecting tumor recurrence in a lymph node in a patient with a malignant PET. A number of additional studies have compared the ability of SRS and other imaging studies to localize tumors in patients with insulinomas [215], [221], [222], [238] and other PETs. [215], [230], [234], [235], [238], [254] In these studies SRS localized an insulinoma in 10% of patients and a gastrinoma in 71% to 81% of patients, whereas CT scan, ultrasound, and MRI detected 20% of insulinomas and a primary tumor in 38% of patients with other PETs.

Three studies [251], [255], [256] and [257] have examined the ability of SRS to alter clinical management in patients with PETs, which is the most important test of its potential use. In one study [255] involving 40 patients with gastroenteropancreatic tumors, SRS altered clinical management in 28% of patients; in the other two studies [251], [256] involving 122 and 85 patients, it altered management in 47% and 24%, respectively, of cases. SRS altered management primarily by clarifying equivocal lesions seen on conventional imaging studies, being the only modality to identify liver metastases, a primary tumor, or distant metastases which altered management. [251], [255], [256] and [257] One particularly important category of clarifying lesions was in regard to small hemangiomas of the liver (<2 cm). [258] These have a prevalence of 7.3% to 15% in autopsy studies, appear indistinguishable from small liver metastases from PETs on MRI or angiography, and cannot be distinguished easily by red blood cell (RBC) scanning or any imaging modality. [258] In one study [258] SRS was positive in 93% of patients with small liver metastases (mean 1.3 cm) from PETs and negative in all small hepatic hemangiomas (mean 1.3 cm). SRS had a significantly better sensitivity, accuracy, and positive and negative predictive value than any other modality for identifying the liver metastases (see Fig. 97-8). [258]

Numerous studies demonstrate that endoscopic ultrasound (EUS) is a sensitive method to detect PETs, especially if intrapancreatic (see Fig. 97-9). [215], [221], [222], [235], [237], [238] and [239] In ten different studies, endoscopic ultrasound localized an insulinoma in 57% to 92% of patients, [215], [221], [222], [236], [239] and was almost equally sensitive to calcium provocative testing (i.e., 88%), [239] whereas CT scan only localized a tumor in 0% to 20%, and SRS in 12% to 14%. Similarly, EUS localized a gastrinoma overall in 40% to 100% of patients, [239] a pancreatic gastrinoma in 75% to 100% of cases in four studies, [215], [222], [237] but a duodenal gastrinoma in 0% to 67% in four studies. [215], [235], [237], [238] Therefore, EUS was particularly sensitive for localizing PETs within the pancreas and with insulinomas is more sensitive than SRS, whereas with other PETs in the pancreas it is at least equal in sensitivity to SRS. [215], [221], [239] Figure 97-9 shows the ability of EUS to localize an insulinoma in a patient. EUS requires considerable expertise to obtain optimum results, whereas SRS can be performed in most radiology departments. However, to obtain optimum results with SRS, single photon emission computed tomography (SPECT) scanning is needed. [232], [234] Some studies suggest SRS in combination with EUS in patients with gastrinomas is more sensitive than either alone. [235], [259] Neither the EUS nor the SRS localize many small extrapancreatic PETs, especially duodenal gastrinomas. [210], [231], [234], [235] SRS has the advantage over EUS of allowing total body at one time, therefore it is especially useful for identifying distant metastases. With both EUS and SRS false-positive results can be obtained; however, the exact rate is not well defined. SRS can be positive in thyroid disorders, breast disease, lymphomas, granulomatosis diseases (sarcoid, tuberculosis, etc.) due to gallbladder filling, wound infections, accessory spleen, various arthritides, and other neuroendocrine tumors besides PETs, such as gastric carcinoids. [169], [233], [260], [261] In a prospective study, [261] SRS had a false-positive rate of 12% for localizing a gastrinoma; however, when the clinical context was carefully considered, the percentage in which false-positive SRS results altered clinical management was 3%. There are no comparable studies on the false-positive rate of EUS for small PETs, especially those that are extrapancreatic in location.

For metastatic disease to the liver, SRS is more sensitive than conventional imaging studies (see Table 97-6). [169], [230], [234], [251], [255], [258] This is shown in Figure 97-8 for a patient presenting with right upper quadrant pain with an elevated serum chromogranin level but normal CT scan (see Fig. 97-8) as well as other conventional imaging (MRI, angiography, ultrasound). The SRS demonstrated three liver metastases that were proven to be metastatic PETs. In a comparative study [230] of 24 patients with proven hepatic metastases and gastrinomas, SRS was positive in 92% of the patients which was significantly better than ultrasound (46%), CT scanning (42%), angiography (62%), and was equal to MRI (71%). The combination of all conventional imaging studies was equal to SRS alone (83% vs. 92%, respectively).

Functional localization by venous sampling for hormonal gradients from various portal venous sites or peripheral veins has been used for both insulinomas and other PETs (see Fig. 97-10). [12], [140], [240], [241], [242] and [243] This procedure requires considerable expertise and was associated with complications in 20% of the patients in one large study. [241] A simplified method has been developed that has replaced direct venous sampling from portal venous tributaries and does not require a transhepatic catheterization. Studies [210], [245], [262] demonstrated that gastrinomas can be functionally localized by measuring hepatic venous samples for gastrin gradients after selective intraarterial injection of secretin. This method was more sensitive than PVS, required less expertise, and had lower morbidity. A similar procedure has been developed for localizing insulinomas [244], [263] using calcium gluconate (0.01–0.025 mEq Ca^{2+}/kg). In one comparative study [244] in patients with insulinomas the intraarterial study with selective hepatic venous sampling for insulin levels was positive in 88% of the patients, ultrasound in 9%, CT scan in 17%, MRI in 43%, angiography in 36%, and PVS in 67%. Calcium infusions may also stimulate the release of hormones from other PETs, so this approach may be useful in these tumors. [264], [265] and [266] A typical result is shown in Figure 97-10 where a much larger increase in insulin concentration in the hepatic vein is seen after the splenic arterial injection; at surgery insulinomas were found in the pancreatic tail in the area supplied by this artery.

Intraoperative ultrasound can be a valuable aid in localizing PETs at the time of surgery (Fig. 97-11). [216], [228], [246], [247] Intraoperative ultrasound is particularly useful for localizing small intrapancreatic PETs, but is less sensitive for extrapancreatic tumor. [216], [246] Intraoperative ultrasound may also suggest whether the PET is malignant, define the proximity of a PET to the pancreatic duct, and further define the anatomy of a nodule palpated at surgery. [216], [228], [246], [247] The use of intraoperative ultrasound at surgery will be discussed further in the treatment section under "Surgical."

Bone metastases have been described in patients with advanced PETs. [54] In a one study [54] of 115 patients with gastrinomas, bone scanning, MRI of the spine, and SRS were compared in their ability to identify bone metastases. Eight patients (7%) were shown to have bone metastases and SRS and MRI had the highest sensitivity and specificity. [54] Bone scanning [54] had a low specificity and was less sensitive than SRS or MRI of the spine. Bone metastases occurred in 31% of the patients with liver metastases, only occurred in patients with liver metastases, were usually in the axial skeleton initially, and their detection changed management in all cases. [54] Figure 97-12 shows an example of the ability of SRS to image the entire skeleton at one time and localize bone metastases in a patient with a malignant

PET. In this patient (see Fig. 97-12) SRS detected more spinal metastases than MRI, demonstrating its high sensitivity.

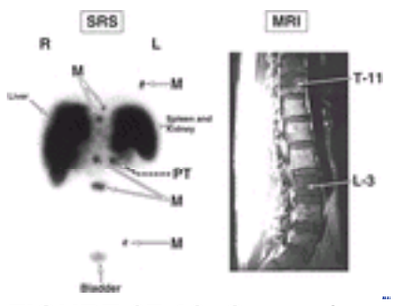

FIGURE 97-12. Comparison of the ability of somatostatin receptor scintigraphy (SRS) and magnetic resonance imaging (MRI) to localize bone metastases in a patient with a malignant pancreatic tail pancreatic endocrine tumor (PET) secreting glucagon and gastrin. In this patient with back pain, MRI demonstrated T-11 and L-3 metastases (**right panel,** *arrows*) whereas SRS (**left panel**) demonstrated numerous metastases (*M*) in the thoracic and lumbar spine, ribs, and right pelvis. The primary tumor (*PT*) is shown on SRS (*broad dotted arrow*). This patient's results show the high sensitivity of SRS for localizing bone metastases and its ability to allow a complete body scan at one time.

Figure 97-13 summarizes the currently recommended approach to a patient with a PET that is not an insulinoma. In such a patient both the location of the primary tumor and possible occurrence of liver or distant metastases need to be assessed. Because of its increased sensitivity and ability to image the entire body in one study, the SRS is recommended as the initial study with SPECT imaging. If the SRS is positive for liver metastases, the extent and number of lobes need to be established to determine whether surgical resection of all tumor that can imaged is possible as discussed in the next section, "Treatment." If diffuse metastases are present, a CT or ultrasound-guided percutaneous liver biopsy should be done (see Fig. 97-13). Because it is difficult to evaluate increases in tumor size by SRS, an MRI or CT scan should be performed to evaluate changes in the size of liver metastases with time. [212] If no metastases are present and no primary tumor is seen, endoscopic ultrasound should be performed by an experienced endoscopist. If a primary tumor is still not seen, a selective angiogram should be performed and the use of intraarterial calcium with determination of hepatic venous hormonal gradients considered (see Fig. 97-10, Fig. 97-13).

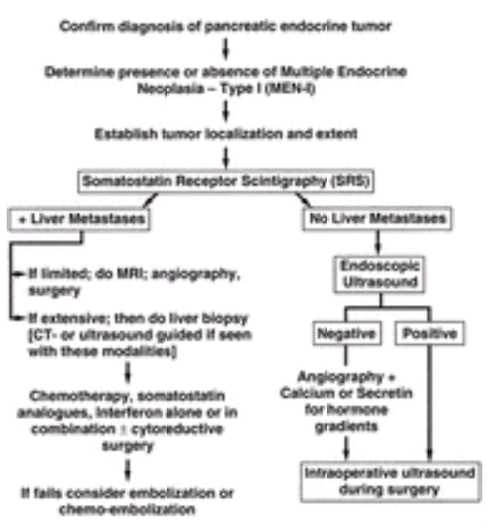

FIGURE 97-13. Algorithm summarizing the approach to tumor localization in a patient with a pancreatic endocrine tumor (PET) that is not an insulinoma. Because of its increased sensitivity for detecting the primary and metastatic disease both in the liver and distant sites [169, 230, 233, 251, 255] and its ability to cover all body areas in one study, somatostatin receptor scintigraphy (SRS) with single photon emission computed tomography (SPECT) imaging is the initial localization method of choice. SRS will localize greater than 90% of patients with liver metastases. [169, 230, 233, 251, 255] If no liver metastases are seen, endoscopic ultrasound (EUS) is the most sensitive method to localize the pancreatic primary. [236, 259] If the EUS is negative, selective angiography with intraarterial injection of calcium (insulinoma, somatostatinoma, glucagonoma) and sampling of hormonal levels in hepatic venous samples should be done. At surgery intraoperative ultrasound should always be used because it will identify lesions not seen by other methods. [216] Up to 15% of patients with metastatic disease in the liver may have resectable disease and should be considered for surgery. [213, 214, 272, 276]

TREATMENT

All patients with functioning PETs have two problems that are interrelated but often have to be dealt with separately. Acutely the symptoms due to the hormone excess state must be controlled, and ultimately the tumor itself must be dealt with. Although excision of the PET would solve both clinical problems, this is frequently not possible either because there is metastatic disease at the time of diagnosis, the primary PET cannot be found at surgery, or curative resection is not achieved. Furthermore, the clinical condition of the patient may have deteriorated because of complications of the hormone excess state necessitating treatment prior to surgery. [2, 247]

Even with the increased ability to control the symptoms of the hormone excess state with various medical therapies, a number of factors suggest that all patients with PETs should be considered for possible surgical resection of the tumor. First, even though only 5% to 15% of insulinomas are reported as malignant, with the other PETs at least 50% are malignant and in many reports this percentage of patients or higher have metastases at the time of diagnosis. [2, 267] For example, in various studies up to 90% of all gastrinomas, [12, 42, 268] 90% to 100% of nonfunctional PETs, [1, 28, 269, 270] 82% of glucagonomas, [13] and 50% to 60% of VIPomas [18, 19] were malignant. Furthermore, although in some studies large tumors are more likely to be malignant, [7, 8, 51, 53, 271] metastases occasionally occur with small tumors; therefore, the fact that no tumor is found on the initial imaging studies or that the tumor is small does not establish that metastatic disease will not occur. Although a number of patients with metastatic glucagonomas, [51, 72, 141] gastrinomas, [7, 8, 12, 53] and somatostatinomas [13, 21, 75] have been reported to have long-term survivals, recent detailed studies with PETs, such as gastrinoma in which there has been very effective therapy for the gastric hypersecretion for a more than 15 years, suggest that long-term prognosis is much worse in patients with advanced disease than originally thought. [7, 8, 12, 72, 271, 272] Results from the study primarily of patients with gastrinomas have provided a number of insights into prognostic factors and determinants of survival in these patients which are likely also important with other less common PETs because of their similar biologic behavior, histopathology, growth patterns, and metastatic pattern (Table 97-7). [7, 8, 42, 53, 212, 271] These studies demonstrate that various PETs in different patients frequently grow at very different rates. [7, 8, 17, 53, 212, 271, 273] In approximately 75% of patients with gastrinomas the tumor demonstrated either no growth or indolent growth and in 25% it demonstrates aggressive growth. [8, 53] Furthermore, even in patients with liver metastases, aggressive growth occurred in less than 50%, however, all the deaths occurred in this subgroup of patients. [212]

I. Clinical Features	
	Female gender
	Absence of MEN-1 syndrome
II. Laboratory Features	
	Increased chromogranin A in some studies
	Increased gastrin level in gastrinomas
	Lack of progesterone receptors
III. Tumor Extent/Size	
	Presence of liver metastases
	Increased extent of liver metastases
	Presence of lymph node metastases
	Presence of bone metastases
	Primary tumor site
	Primary tumor size (≥3 cm—worse prognosis)
IV. Tumor Behavior	
	Growth of liver metastases
	Incomplete tumor resection
	Nonfunctional tumor
	Development of ectopic Cushing syndrome
	Increased depth of tumor invasion
V. Histological Features	
	High nuclear atypia
	Poor tumor differentiation
	High growth indices [high K_i 67 index (>2%), Proliferating cell nuclear antigen expression]
	Capsular invasion
	Vascular or perineural invasion
VI. Other	
	Flow cytometric features (i.e., aneuploidy)
	Ha-Ras oncogene or *p53* overexpression

Data are from refs. 7, 8, 53, 169, 192, 271, 274.

TABLE 97-7 Prognostic Factors in Pancreatic Endocrine Tumors for Decreased Survival

The presence or absence of liver metastases either initially, or the development of liver metastases during follow-up, is the single most important prognostic factor (Fig. 97-14). [8] , [17] , [53] , [271] For example, in one study [8] the 15-year survival in patients with liver metastases present during initial assessment was 26%, whereas in patients without liver metastases it was 96%. The extent of liver metastases (one lobe, two lobes, diffuse) (see Fig. 97-14), presence of lymph node metastases, bone metastases, a large primary tumor size, primary location (pancreatic worse prognosis than duodenal gastrinoma), various histological features, tumor marker levels (chromogranin A, specific hormone such as gastrin level), and flow cytometric features are all important prognostic factors (see Table 97-7, Fig. 97-14). [2] , [7] , [8] , [53] , [169] , [192] , [271] , [274] Studies demonstrate that survival is directly related to tumor extent. In patients with primary tumors so small that no tumor was found at the time of surgery, the 5- or 10-year survival was 90% to 100%. [12] , [72] , [272] In patients with complete tumor resection the 5- or 10-year survival was also 90% to 100%. In patients whose tumors are incompletely resected or who have developed a recurrence postoperatively, the 5-year survival varied from 14% to 76% and 10-year survival was 20%. In patients with unresectable PETs, such as gastrinomas metastatic to the liver, the 5-year survival varied from 20% to 75% and the 10-year survival in one study was 30% (see Fig. 97-14). [12] , [72] , [272] These data suggest that once metastatic disease occurs the prognosis is significantly decreased. An important study [275] demonstrated for the first time that resection of the primary tumor (i.e., gastrinomas) significantly decreases the rate of development of liver metastases. In this study [275] survival was also almost significantly improved ($P = 0.08$). Furthermore, because therapy for advanced disease is in many cases only partially effective, [2] , [12] , [211] , [213] it is important to attempt to resect the PET while still localized. Unlike gastrinoma, the long-term medical therapy of the other various PETs is not well established and it is not known what percentage of patients will not respond to medical treatment. Therefore, in functional PET syndromes not including gastrinomas, cytoreductive surgery may be helpful because it may improve symptom control. [141] , [213] , [214] , [276] , [277] Lastly, the long-term toxicity of most medical therapies except for gastrinomas is not well established.

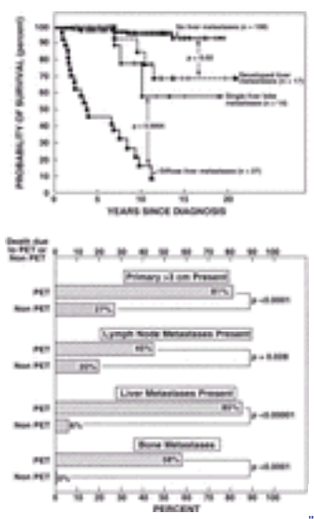

FIGURE 97-14. Effect of the extent or development of liver metastases from a pancreatic endocrine tumor (PET) (**top panel**), presence or absence of lymph node or bone metastases or PET primary size on disease-related survival. In the bottom panel, the percentages of patients with a PET-related (n = 33) or non–PET-related (n = 34) death with the indicated tumor size or metastases are shown. Data are from 158 patients and are modified from refs. [8] (**top**) and 42 (**bottom**). These results demonstrate that the presence of liver, bone, or lymph node metastases, a large primary PET size, and increased extent of liver metastases are all important predictors of a tumor-related death.

Nonmetastatic Disease

Medical Treatment

General considerations. The initial management of patients with insulinomas and the long-term management of those patients with insulinomas who are not surgically cured, is directed at controlling the hypoglycemia. The simplest form of nonsurgical control is dietary management. Frequent small meals may alleviate symptoms. [70] , [98] In severe cases intravenous glucose combined with various drug therapies may be required. In general, 10% dextrose should be avoided because of the electrolyte imbalances generated by the combination of elevated serum concentrations of glucose and hyperinsulinemia. [98] Some investigators have used slowly absorbed nutrients to maintain normal blood sugars prior to surgery. This may be preferable to diazoxide because of reports of sudden hypotension in some patients receiving diazoxide prior to surgery. [278] However, diazoxide (150–800 mg/d) has been used successfully to manage patients with insulinoma prior to surgery as well as in long-term treatment. [70] , [98] , [279] Diazoxide is a benzothiadiazide that has a plasma half-life of 28 hours and a peak hyperglycemic effect 12 hours after an oral dose or 4 hours after an intravenous dose. Its hyperglycemic action is attributed to directly inhibiting insulin release by adrenergic stimulation and by enhancing glycogenolysis. [51] , [70] The main side effects of diazoxide are sodium retention (47%) gastrointestinal symptoms such as nausea, and occasionally, hirsutism. [51] , [70] , [98] , [279] Sodium retention can be minimized by administration of diazoxide with a diuretic and restricting sodium intake. The gastrointestinal side effects can be decreased by taking the drug with meals. [51] , [98] Diazoxide is primarily excreted by the kidneys and is known to displace various protein bound drugs and increase their free concentrations in the serum. [98] Diazoxide therapy should be initiated with low doses and increased to a maximum of 600 to 800 mg/day in two or three divided doses as tolerated or until symptoms are controlled. In general approximately 50% to 60% of patients will respond. [51] , [70] , [98] , [279] , [280] Patients have been treated for up to 22 years with diazoxide, [279] demonstrating it can be effective long term. The calcium channel blocker verapamil [70] , [281] , [282] and diphenylhydantoin [283] have also been used to control hypoglycemia, with some success in some patients when diazoxide has failed. The long-acting somatostatin analog, octreotide (Sandostatin) has been shown to be effective in 36% of patients for the acute and long-term control of hypoglycemia in patients with insulinoma. [27] , [116] , [153] , [169] This will be discussed further in the next section. In the initial management of patients with VIPomas the most important issue is to replace the large volumes of fluid and electrolytes lost, and correct the hypokalemia and acidosis [18] , [19] This is particularly important because acutely a common cause of death in these patients is renal failure associated with hypokalemia and hypokalemic nephropathy. [18] Drug therapy previously included the use of high doses of prednisone (60–100 mg/d) [18] which decreased diarrhea in 40% to 50% of patients, [18] , [18] and clonidine, [284] angiotensin II, [285] norepinephrine, [285] , [286] indomethacin, [287] , [288] lithium carbo-nate, [289] phenothiazines, [290] , [291] propanolol, [27] metoclopra-mide, [292] lidamidine, [293] and loperamide, [293] all of which decreased the diarrhea in a small number of patients. These agents primarily enhanced sodium absorption in the proximal small intestine or inhibited intestinal secretion. [18] Newer studies demonstrate that the long-acting somatostatin analog octreotide is now the drug of choice and will control diarrhea in 87% of all patients with VIPomas. [27] , [169] This will be considered in detail in the following section. In patients with glucagonoma a migratory necrolytic erythema rash may be severe. Some studies suggest that correction of the hypoaminoacidemia with parenteral amino acids and normalization of the plasma amino acid levels will improve the rash. [72] , [144] , [145] However, another study noted disappearance of the rash after surgical resection of the glucagonoma but not dietary normalization of the plasma amino acids. [146] Octreotide [116] , [153] , [169] improves symptoms in 84% of patients with

glucagonomas and its use is discussed in detail in the next section.

Medical therapy with octreotide. As pointed out earlier, somatostatin has been shown to inhibit the release of a number of hormones in both animals and in human volunteers, as well as to have direct actions on a number of target organs including inhibition of gastric and pancreatic secretion, intestinal absorption, and gastrointestinal motility. [168], [169] Administration of natural somatostatin by continuous infusion to a number of patients with functioning PETs demonstrated symptomatic improvement in patients with VIPomas, insulinomas, gastrinomas, and glucagonomas. [27], [43], [116], [169] However, natural somatostatin has a very short half-life ($t_{0.5}$ 2–3 min) [11], [169] and, therefore, has limited therapeutic efficacy because it has to be given by continuous intravenous infusion. The synthetic octapeptide analog of somatostatin, octreotide acetate (Sandostatin), has a half-life ($t_{0.5}$ 100 min) 33 times longer than somatostatin and thus can be administered 2 to 4 times a day. [116], [169], [294], [295] In the rat, it was 70-fold more potent than somatostatin at inhibiting growth hormone release, 3-fold more potent than native somatostatin at inhibiting insulin release, 23-fold more potent at inhibiting glucagon release, and 80-fold more potent than native somatostatin at inhibiting acid secretion. [169], [294], [295] Octreotide is usually started at a dose of 50 to 150 µg bid to tid subcutaneously given by self-administration and the dose increased if it is not effective. Doses up to 750 µg tid have been used in patients with the carcinoid syndrome and PETs. [116], [153], [169] There have been reports in small numbers of cases where intermittent subcutaneous administration of octreotide was ineffective or only partially effective and the continuous infusion of octreotide was more effective. [169], [296], [297] One long-term study [298] reported on 7 patients with various PETs treated for a median period of 20 months (range 13–54 months) in which octreotide was initially effective in all. Worsening of symptoms and rising plasma hormone concentrations occurred over a median period of 5 months after starting therapy and was initially reversed by increasing the octreotide dose. After a median of 13 months (range 5–34 months) symptoms returned and were no longer responsive to higher doses of octreotide. In 55 patients with insulinoma drawn from various series, [169] octreotide improved some symptoms in 31% and plasma insulin concentrations decreased in 41%. However 65% of the patients received octreotide for less than 1 week because most went to curative surgery. Octreotide also decreases growth hormone secretion and plasma glucagon levels and it may worsen hypoglycemia. Reduction in hypoglycemia attacks was noted only occasionally. [299] Octreotide was given for more than 1 month in 10 patients. In short-term studies octreotide appears to benefit 36% of patients with insulinoma, however the true percentage of patients with insulinomas that continue to respond to octreotide is not clear. [153] Five subtypes of somatostatin receptors exist and octreotide has high affinity for subtypes 2 and 5, intermediate affinity for sst_3, and low affinity for sst_1 and sst_4. [169], [253] The response rate of insulinomas to octreotide is likely lower than other PETs because they frequently possess low densities of somatostatin receptors, whereas the other PETs possess high densities of high affinity sst_2 and sst_5 in 80% to 90% of cases. [169], [232], [253], [300], [301] This finding may also explain why insulinomas appear to be less responsive to octreotide than glucagonomas, VIPomas, gastrinomas, or GRFomas, and are imaged less frequently by SRS. [169], [232], [233], [300] Octreotide improved diarrhea in 27 of 31 patients (87%) with VIPomas in one review [169] and plasma VIP concentrations decreased in 87% of the patients, which is similar to the 78% to 90% response rate in other reports. [17], [19], [116], [153], [169] In one review [169] tumor size was evaluated in 18 patients with VIPomas and a decrease in size occurred in 4 patients. All patients demonstrated a decrease in diarrhea within the first 24 hours but in 3 patients the effect lasted only a few days. [116] Octreotide was given for more than 1 month in 90% of the patients and remained effective for more than 6 months in 50% of all patients. However, some patients developed decreased responsiveness to octreotide with time and the dose had to be increased. In 87% of patients there was a decrease in the VIP plasma level with octreotide treatment although in many cases it did not return to within the normal range. [169], [302], [303] In up to 25% of patients the diarrhea may respond without a decrease in plasma VIP levels. [153] The apparent discrepancy in response and plasma hormone levels may be partially explained by the fact that multiple forms of VIP occur in the plasma and after octreotide treatment only the native peptide may disappear, which may represent a small fraction of the total VIP immunoreactivity and, therefore, no change in plasma VIP is detected. [303] An example of such a patient is shown in Figure 97-15. This patient [302] had severe secretory diarrhea of 5816 ± 185 g/d with daily losses of 616 mmol of sodium and 348 mmol of potassium, and required 3 L/d of fluid with 462 mmol of sodium and 400 mmol of potassium to maintain serum potassium at 3.4 mmol/L and bicarbonate at 15 to 24 mmol/L (see Fig. 97-15). Octreotide, 100 µg twice a day, was started and stool output fell to 292±55 g/d, with fecal loss of 5 mmol of sodium and 25 mmol of potassium. Serum potassium and bicarbonate levels rose. This VIPoma was secreting VIP, motilin, PP, and neurotensin which, with octreotide treatment, decreased 94% for PP, 76% for VIP, 88% for neurotensin, and 70% for motilin (see Fig. 97-15). [302] This patient's diarrhea was controlled for 3 months at which time she died of respiratory failure. [302] The cost effectiveness of long-term octreotide treatment in patients with VIPomas has been studied. [304] It was concluded that long-term octreotide treatment decreases medical costs for a patient with VIPoma by 50%. [304]

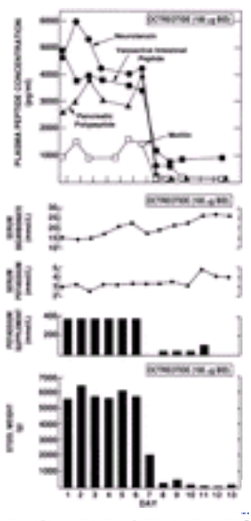

FIGURE 97-15. Effect of octreotide treatment on hormonal levels, stool volume, serum electrolytes and potassium replacement in a patient with a metastatic VIPoma. This patient [302], [303] had a VIPoma secreting vasoactive intestinal polypeptide (VIP), neurotensin, pancreatic polypeptide (PP), and motilin. Values for the different parameters for 6 days prior to and 7 days after receiving octreotide 100 µg bid SC are shown. The hormone levels all decreased by more than 70%, the need for bicarbonate and potassium supplementation were decreased by 94%, and stool weight decreased by more than 95% (5816 ± 185 g/d to 292 ± 55 g/d). The patient was maintained for 3 months on octreotide, remaining asymptomatic until death of unrelated causes.

In 32 patients with glucagonoma, octreotide improved the skin rash in 24 patients and reduced plasma glucagon concentrations in 78% of patients. [14], [15], [27], [43], [72], [116], [153], [157], [169] Weight loss, pain, and diarrhea improved in patients with these symptoms, but octreotide may not improve the diabetes mellitus. [14], [72], [157] Octreotide usually does not decrease tumor size, decreasing the tumor size in only 8% of cases in one study [14] and only 4% of cases in the literature. [14], [169] Decreases in plasma glucagon concentration occur in 80% to 90% of patients but do not always parallel the improvements in the rash. [72], [116], [153] Octreotide was administered for more than 1 month in 94% of patients and remained effective for more than 6 months in 75% of all patients. In one study [14] the duration of treatment ranged from 2 months to 3.5 years. Bromocriptine, a dopamine agonist, is effective in patients with classic acromegaly; however, it only reduces GRF levels in 25% of patients with GRFomas. [77] Octreotide has become the agent of choice for GRFomas. [21], [77], [169], [185] Octreotide reduces symptoms of both growth hormone excess and growth hormone-releasing hormone (GHRH) plasma concentrations in patients with GRFomas. [21], [77], [153], [169], [305] In most patients reported with GRFomas, octreotide improved symptoms and reduced the GHRH levels in all patients, [21], [77], [169], [185] although in one study [306] /SUP>continuous infusion was required. Eight of the nine patients received treatment with octreotide for more than 6 months. [153], [169], [305] Pituitary shrinkage occurred with octreotide treatment in some studies. [185] In GRFomas, the suppression of growth hormone secretion is mainly due to a suppressive effect at the pituitary level because plasma GRF levels do not become undetectable. [77] A small number of patients with somatostatinomas were treated with octreotide. [153], [307] In three of four treated patients plasma somatostatin-like immunoreactivity levels decreased and symptoms due to the somatostatinoma syndrome (diarrhea and diabetes) were improved in two patients. [307] Long-term treatment with octreotide has a number of unresolved issues. Many patients require increasing doses of octreotide with time and some patients require very high doses. [153], [169], [308] When the octreotide was stopped and restarted in a small number of patients, the symptoms were controlled at a lower dose of octreotide, [153] whereas other patients did not respond. Furthermore, it is unclear why, in some cases, intermittent subcutaneous (SC) injections are not effective, whereas continuous low-dose subcutaneous infusions are effective. It is also unclear if intermittent SC octreotide injections are not effective, whether the sustained release preparations (octreotide–long-acting release [LAR], lanreotide-slow release [SR]) [309] will be effective. In addition to reducing the ectopic release of hormone, octreotide also decreases growth rate in a significant proportion of PETs and decreases tumor size in a small percentage of patients. [2], [153], [169], [273], [310] This will be discussed later in the chapter but it is not established that the dose of octreotide is the same for both effects. With short-term treatment side effects have been recorded in approximately 50% of patients treated with octreotide but have rarely been serious enough to stop therapy. [116], [153], [169] Most patients only experience some pain or discomfort at the SC injection site (9%). [116] The most common other side effects are gastrointestinal and include nausea (15%), vomiting (1%), constipation, crampy abdominal pain, and diarrhea (13%) which may improve with time. [153] Steatorrhea can occur with octreotide. [169] At low doses of octreotide steatorrhea is usually mild and transient, however at high doses gross steatorrhea can occur. [311] Dunne and colleagues [116] reported 13% of patients developed diarrhea, 6% fatty stools, and 8% loose stools; in nine patients elevated fecal fat levels were demonstrated. Octreotide rarely has to be stopped because of steatorrhea. Octreotide occasionally may impair glucose tolerance and unmask diabetes mellitus. [312] Occasional postprandial hyperglycemia has been reported. [153] Reactions were graded as mild in 48%, moderate in 38%, and severe in 15%. [116] Most of the side effects were short-lived and did not result in stopping octreotide treatment. During long-term treatment a number of studies have assessed the safety and adverse effects of octreotide,

especially on gallstone formation and gallbladder function, particularly in patients with acromegaly. [169], [171], [174], [175], [313] In one study [174] of 103 acromegalic patients treated with octreotide (SC) for a mean of 30 months, 66% of patients developed an adverse event the first year and 46% the second. Most were related to the gastrointestinal tract including diarrhea (58%), abdominal discomfort (44%), loose stools (36%), nausea (30%), flatulence (13%), and constipation (9%). With continued treatment the adverse events usually disappeared and only 5% of patients discontinued treatment because of adverse events. In various studies in acromegalic patients the incidence of gallstones ranges from 5% to 80% with a mean of 29%. [169], [171], [174], [313] In one study [175] of 33 patients with carcinoid syndrome or PETs treated long-term with octreotide, the overall incidence of cholelithiasis and/or gallbladder sludge was 52%. Seven percent of the patients had symptomatic disease and required an emergency cholecystectomy. The incidence of cholelithiasis varied with octreotide dose, [175] being 35% in patients on low dose (150 µg tid) and 61% with high dose (500 µg tid). The incidence of acute cholecystitis was 12% in the low-dose group and 5% in the high-dose group. [175] Eleven percent of patients underwent elective cholecystectomy. Long-term treatment can also alter glucose tolerance significantly in some patients, which indicates glucose levels should be monitored. [169] Long-term treatment with synthetic somatostatin analogs has been greatly simplified by the availability of sustained-release forms. [58], [169], [172], [309], [314], [315], [316], [317] and [318] Both a long-acting octreotide formulation (octreotide LAR [30 mg IM every month], available in the U.S. and Europe) and a slow-releasing lanreotide (lanreotide SR [30 mg IM every 10–24 days], available in Europe) are now being used for the treatment of acromegaly, carcinoid syndrome, and PETs. [169], [172], [309], [314], [315], [316], [317] and [318] Following a 30-mg IM injection of octreotide-LAR, a plasma level of 1 mg/mL is achieved and maintained for more than 25 days, indicating this formulation can be given monthly. [309] The principal side effects of octreotide-LAR are similar to that discussed for the shorter-acting formulation and include pain at the injection site and gastrointestinal symptoms (usually mild and frequently transient—pain, cramping, diarrhea). [309], [314], [315], [319], [320] Octreotide-LAR and lanreotide-SR impair gallbladder emptying [172], [173] and octreotide-LAR has been shown to inhibit CCK release. [173] In various studies long-term treatment with octreotide is reported not to increase the incidence of gallstones (at 12 mos), [316] or to cause 15% to develop asymptomatic biliary sludge, but none developed gallstones (up to 3 y). [318] Lanreotide-SR is reported to cause new gallstones in 10% to 26% of patients. [314], [317], [321]

Surgical All patients with PETs should undergo exploratory laparotomy for possible cure. [2], [210], [216], [247], [277], [322] Exceptions are patients with unresectable metastatic disease or the occasional patient whose symptoms are well controlled by medical management yet who also has a concurrent medical problem that would make the risks of surgery prohibitive. It is particularly important that the surgery only be done by a surgeon with experience in operating on patients with PETs. [12], [98] Even very experienced general surgeons and many endocrine surgeons have little experience with these rare tumors and therefore will frequently miss small tumors, perform unnecessary major resections, or failure to perform the appropriate resection to palliate symptoms. As outlined previously (see Fig. 97-13), prior to surgery all patients should undergo localization studies and if the tumor localization or extent is still unclear, then functional localization studies (selective hormone sampling of portal venous tributaries) or calcium infusion with hepatic venous sampling (insulinomas, somatostatinomas, VIPomas) should be considered. Even if localization studies are negative, provided metastatic disease or a prohibitive concurrent illness are not present, the patient should undergo exploratory laparotomy. [2], [12], [210], [216], [231], [247], [277] The surgical approach has been reviewed, [210], [223], [277] and is guided by the tumor location. At exploration the liver should be carefully examined for evidence of metastatic disease. In the case of PETs that are frequently extrapancreatic in location the entire abdomen, especially the proximal small intestine, should be carefully explored to locate extrapancreatic tumors. [210], [223] An extended Kocker maneuver should be performed to allow thorough examination of the pancreatic head area. The entire pancreas must be carefully explored, even if one PET is found, because there may be multiple tumors. [98] A detailed exploration for multiple endocrine tumors is particularly important in those patients with MEN I. [12], [323] Isolated small insulinomas should be removed by enucleation because they are rarely malignant. Isolated noninsulinoma PETs in the pancreatic body or tail should be removed either by enucleation or en bloc by distal pancreatectomy because they are occasionally malignant. [210], [216], [223], [247] Pancreatic head and proximal pancreatic body tumors require enucleation with careful dissection to avoid damage to the main pancreatic duct and its attendant morbidity. If at surgery an isolated hepatic metastasis is found and can be removed without increased risk, then it should be excised with negative margins. [2], [223] Aggressive surgery such as a proximal cephalic pancreaticoduodenectomy (Whipple operation) at present is indicated only in carefully selected patients because it has not been established that the increased risk of morbidity may not outweigh the potential benefit in most patients. [2], [12], [210] Pneumococcal vaccine (Pneumovax) should be administered preoperatively to lower the risk of postsplenectomy sepsis. At the time of surgery the use of intraoperative ultrasound may be helpful and is recommended. [12], [216], [282], [283] Studies show this technique will identify a certain number of PETs that would not otherwise be identified and therefore should be used routinely (see Fig. 97-11). [2], [12], [216], [228], [246], [247], [324] In seven studies [216] intraoperative ultrasound localized 89 of 101 (88%) insulinomas. Intraoperative ultrasound changed operative management in 10% of all cases of gastrinomas [246] either by localizing additional tumors or by determining a tumor was malignant. Intraoperative ultrasound is particularly helpful for intrapancreatic lesions. [216] In one study [246] intraoperative ultrasound localized 96% (22 of 23) of pancreatic gastrinomas and in a review of four studies [216] it localized 91% of 34 pancreatic gastrinomas. However, it is less sensitive for localizing duodenal tumors, localizing only 30% of duodenal gastrinomas in two studies. [246] In 12 consecutive patients with occult insulinomas not visualized by any conventional imaging study, [228] intraoperative ultrasound identified 10 of 11 (91%) of the insulinomas that were correctly identified including 5 pancreatic head insulinomas not identified by palpation. The use of intraoperative ultrasound not only localizes primary tumors not detected by other modalities, it reduces complications like fistulae, pancreatitis, and pseudocysts by allowing better definition of the anatomy prior to a procedure. [216] At the time of surgery in patients with insulinomas there have been occasional reports of hypoglycemia during manipulation of the tumor. Use of the artificial pancreas (Biostator) has documented an increased glucose requirement during resection of the insulinoma. [325] A number of centers document successful tumor removal by demonstrating rebound hyperglycemia. However, a study has shown that rebound hyperglycemia did not occur in 23% patients with successful tumor excision [326] and therefore, its use is not recommended. In most studies 75% to 98% of patients with insulinomas will be cured by surgery. [51], [52], [70], [98], [103], [218], [223], [277] With glucagonomas there have been a number of reports of successful excision of the tumor [11], [43], [72], [141], [327], [328] and in many cases follow-up has been significantly long enough to suggest cure. However, at least 50% to 95% of the patients with glucagonomas have metastases at the time of diagnosis and a significant additional percentage at the time of surgery. [14], [16], [72], [141] A number of cases of glucagonomas have shown recurrence during long-term follow-up, despite what appeared to initially have been a complete resection of all tumor. [141] Therefore, the true long-term cure rate is not established. With VIPomas, somatostatinomas, GRFomas, and PPomas many patients present with metastatic disease and, while curative resections do occur, the exact percentage of patients cured by surgical excision of the tumor has not been adequately studied. [4], [29], [68], [277] If the patient is found to have more extensive disease with either local invasion or metastases or at time of surgery or metastatic disease prior to surgery, then the question of debulking or cytoreductive surgery often arises, especially if the patient has a functional PET syndrome. [213], [214], [272], [276], [277], [329] This will be dealt with more extensively in the following section “Metastatic Disease.”

Metastatic Disease

Metastatic PETs have been characterized as slow growing. [7], [11] In fact, until recently there was very little data on the rate of growth of untreated metastatic disease in patients with PETs because many patients required antitumor therapy to control symptoms. Three studies [8], [212], [273] provide some insights into the natural history of metastatic disease. One study [212] of 19 patients with proven metastatic gastrinoma in the liver provides some important insights into the natural history. In 26% of these patients the tumor demonstrated no growth over a 29-month period, 32% had slow growth (1% to 50% volume increase/mo) and 42% had rapid growth (>50% volume increase/mo). [212] These results agree with another study [273] which demonstrated that 78% of patients with metastatic neuroendocrine tumor to the liver had progressive disease over a 6-month period. The third study [8] showed that 50% of patients with gastrinomas die from tumor-related causes, with the extent of metastatic disease and presence of bone metastases being important prognostic factors. These studies demonstrate that in most patients metastatic PETs continue to increase in size and in approximately 50% the growth is rapid. [212] Survival studies also support these findings. [8], [212] In one study [212] 62% of patients with liver metastases with rapid growth died and none of the patients with liver metastases showed either no growth or slow growth over a mean follow-up period of 5.5 years. No clinical or laboratory parameter correlated with growth rate; however, the growth rate was highly predictive of death from the tumor. [212] In one recent study [328] involving 85 patients with various functional PETs, none of the patients without metastatic disease died (follow-up 3–18 years, median 8 years). However, for the 41 patients with metastatic disease, 66% died from tumor progression with an overall 5- and 10-year survival rate of 54% and 28%, respectively. In patients with metastatic disease not to the liver (primarily lymph nodes), [328] the 5-year survival was significantly better ($P < 0.001$) (90%) than those with hepatic metastases at diagnosis (23%). The 5- and 10-year survival rates [328] for patients with malignant insulinomas is 50% to 88% and 50% to 75%, gastrinomas 47% to 53% and 18% to 30%, glucagonomas 52% to 60%, and VIPomas 20% to 88% and 25%, respectively. [16], [17], [52], [68], [328] The natural history of metastatic VIPomas, glucagonomas, somatostatinomas, or insulinomas is not completely clear because until recently effective therapy for the hormone excess state did not exist in most cases and patients frequently died of resulting complications from the hormone hypersecretion. However, with the availability of the long-acting somatostatin analog octreotide, this may change. Excellent long-term survival data are now available for patients with gastrinomas, for which effective therapy has existed for more than 20 years (see Fig. 97-14). [8], [12], [53], [210], [211] Overall, the 5- and 10-year survival rates are 74% and 60%, respectively, in 6 series, [12], [53] whereas for patients with a tumor completely resected it is 100% and 90%, [53] with a primary tumor so small that no tumor was found at surgery it is 95% and 84%, [12], [53] for patients in whom the tumor was resected but they are not cured or had a recurrence it is 75% and 56%, respectively, [53] and in patients with unresectable tumors it is 44% and 33%, respectively. These data clearly demonstrate that overall survival correlates with the extent of the disease. Furthermore, two large studies [8], [53] of 185 patients demonstrates the single most important determinant of long-term survival in patients with gastrinoma is the presence of liver metastases. Moreover, of patients with metastatic disease who subsequently died, 50% to 70% of the deaths were due to tumor progression. [8], [12] At present there are no data to suggest that the other metastatic PETs will differ in their natural history from gastrinoma and, in fact, the study of survival of patients with other PETs [328] previously cited suggests a similar natural history. This conclusion is further supported by the fact that similar percentages of patients with each noninsulinoma PET have metastatic or malignant disease, suggesting that their malignant potential is similar as are growth patterns. [18], [20], [51], [141] Therefore, most authorities would agree that treatment directed at the metastatic disease is indicated. [2], [330], [331] Disagreement occurs over the type of therapy, the efficacy of the therapy, and when therapy should begin. Chemotherapy, [12], [211], [270], [330], [332], [333], [334], [335], [336], [337], [338], [339], [340], [341], [342], [343], [344], [345], [346], [347], [348], [349], [350], [351], [352] and [353] possible cytoreductive surgery, [12], [213], [214], [272], [276], [354], [355], [356] and [357] hepatic arterial embolization (for functional tumors metastatic to the liver with or without chemotherapy [chemoembolization]), [337], [358], [359] interferon, [360], [361], [362], [363], [364], [365], [366] and [367] the use of the long-acting somatostatin analogs, [153], [273], [310], [360], [368], [369], [370], [371], [372] and [373] liver transplantation, [56], [374], [375], [376], [377], [378] and [379] and somatostatin receptor-mediated

radiotherapy [58], [380], [381] and [382] have all been reported to have positive effects on metastatic PET growth. Each will be considered in subsequent sections.

Chemotherapy A number of different chemotherapeutic regimens have been evaluated in patients with metastatic PETs (Table 97-8). In most studies, because of their rare occurrence, chemotherapy for PETs has been considered together as a group or in other cases included in series with metastatic carcinoid tumors. [2], [12], [270], [343], [348], [351] Although some studies [270], [334], [343] report no difference in responsiveness to chemotherapeutic agents between different types of PETs, it has been pointed out that the sample size of the various tumors included in these studies was too small to establish that each type of PET responds equally to chemotherapy. [12], [342] In fact, some clinical observations suggest there may be significant differences in their responsiveness. For example, in some studies as many as 90% of metastatic VIPomas are reported to respond to streptozotocin, often with a complete response. [16] In contrast, only 5% in one prospective study [340] and 40% in another prospective study [342] of patients with metastatic gastrinomas increasing in size prior to treatment responded to streptozotocin plus 5-fluorouracil (5-FU) with or without doxorubicin and in no case was there a complete response. Furthermore, a high proportion of metastatic glucagonomas are reported to respond to dimethyltrizenoimidazole carboxamide (DTIC) including several complete remissions in some studies, [2], [156], [333], [383], [384] and [385] whereas in other PETs, DTIC had a low response rate (i.e., 6%; see Table 97-8).

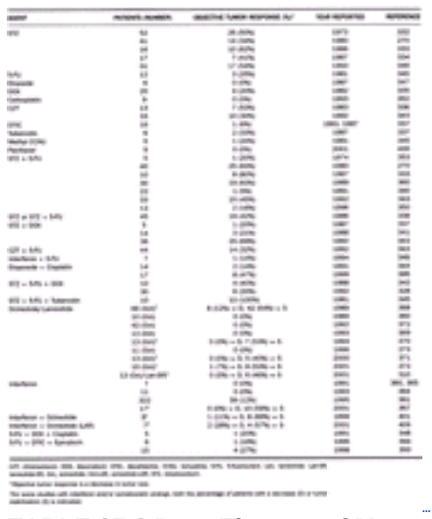

TABLE 97-8 Drug Therapy of Metastatic Pancreatic Endocrine Tumors

In almost all studies, streptozotocin is used either alone or in combination with 5-FU and/or doxorubicin. [2] Streptozotocin is a nitro-urea antibiotic originally isolated from a *Streptomyces*. [386], [387] In early studies it was observed to induce diabetes mellitus. [388] The first report of clinical effectiveness of streptozotocin in a PET was in 1968. [389] Since then it has been generally used in the treatment of PETs either alone or in combination with other agents (see Table 97-8). In various series streptozotocin alone has been reported to cause objective remissions in 34% to 62% of PETs and other single-agent therapy has been relatively ineffective with 5-FU, etoposide, doxorubicin, carboplatin, methyl CCNU, tubercidin, and DTIC giving an objective response in 0% to 33% (see Table 97-8). Chlorozotocin (CZT), a chloroethyl analog of streptozotocin, was discovered in 1976. [389] Compared to streptozotocin, CZT is reported to have only mild to moderate gastrointestinal toxicity and myelosuppression is the dose-limiting side effect, [390], [391] Furthermore, it has the practical advantage of only having to be administered in a single intravenous dose during each course of chemotherapy. CZT alone in two studies had a response rate of 30% to 53%, which is similar to streptozotocin alone (see Table 97-8). Streptozotocin with 5-FU or doxorubicin or both has been reported to cause objective responses in 5% to 80% of patients with a mean response rate of 43% in the different reports (see Table 97-8). The current chemotherapeutic agents of choice for metastatic PETs are the combination of streptozotocin and doxorubicin. [343] This recommendation is based on an Eastern Cooperative Oncology Group (ECOG) comparative study [343] of streptozotocin and doxorubicin versus streptozotocin plus 5-FU or CZT in 105 patients with metastatic PETs. Streptozotocin and doxorubicin had a 69% response rate and this was significantly higher than the 45% response rate with streptozotocin and 5-FU or 30% response rate with CZT alone. Furthermore, streptozotocin [343] plus doxorubicin had a significant advantage in terms of survival ($P = 0.0004$). A previous prospective study [270] done by the ECOG had also demonstrated the superiority of double-agent therapy over streptozotocin alone, with streptozotocin plus 5-FU having a higher response rate of 63% compared to 40% with streptozotocin alone in patients with metastatic PETs. At present it is not clear why the response rate in some more recent prospective studies [340], [342], [350] is significantly lower than the response rate reported in these two studies. In three more recent prospective studies with streptozotocin with 5-FU with or without doxorubicin in patients with metastatic gastrinomas or PETs to the liver, response rates of 5%, 16%, and 40% were reported. [340], [342], [350] In no study were there any complete responders and in one study there was no difference in survival between responders and nonresponders. [342] At present it is not clear whether this difference in response rates from those previously reported with PETs is due to either differences in the tumors themselves or to differences in extent of metastatic disease. The use of both streptozotocin and CZT is associated with significant side effects. In prospective studies 74% to 100% of patients experienced nausea/vomiting and 20% to 40% developed renal toxicity with increased serum creatinine, proteinuria, or both. Nausea and vomiting are frequently severe but now can be controlled in almost every patient with 5-hydroxytryptamine receptor antagonists such as ondansetron. [2], [392] These side effects are reversible in most cases including the early renal effects; however, in the 1992 ECOG study, [343] nine patients developed chronic renal failure and seven patients required dialysis. Serum creatinine and urine protein must be assessed prior to each cycle of chemotherapy. [270], [342] Various triple-drug combinations with or without streptozotocin have been tried in small numbers of patients with variable response rates including streptozotocin with 5-FU and doxorubicin [328], [342] or with tubercidin, [345] 5-FU, doxorubicin, and cisplatin, and 5-FU with DTIC and epirubicin (see Table 97-8). [348], [393], [394] In one study the combination of etoposide and cisplatin [344] was used in 13 patients with metastatic carcinoid tumors, 14 patients with metastatic PETs, and 18 patients with anaplastic tumors (see Table 97-8). None of the carcinoid tumors, 14% of the PETs, and 67% of the anaplastic neuroendocrine tumors demonstrated a partial to complete regression. [344] Two subsequent studies [395], [396] confirmed the effectiveness of this combination in undifferentiated or poorly differentiated carcinoids or PETs, with a 54% and 42% response rate. Most experts agree that chemotherapy, chemoembolization, hormonal therapy, or interferon is only indicated in patients with extensive metastatic disease (usually to the liver) [2], [12], [211], [213], [330], [333], [338], [358] and is not indicated for treatment of the primary tumor when surgery is not curative. There is no agreement, however, about when chemotherapy, chemoembolization, hormonal therapy, or interferon should be begun. [2], [12], [211] Some groups recommend treatment only when the patient develops symptoms due to the PET, [270] whereas others recommend it only when the tumor is demonstrated to be increasing in size. [2], [12], [342]

Other Treatments of Metastatic Disease

Interferon. Because of the toxicity and relatively poor response rates of chemotherapy in patients with extensive metastaticPETs, other approaches have been investigated. Interferon (human leukocyte, a-interferon) is reported to be effective in controlling symptoms in a number of patients with PETs and carcinoid tumors. [349], [361], [362], [365], [397], [398] One review [399] analyzed results from 383 patients from 13 studies of patients with various neuroendocrine tumors (carcinoid tumors, PETs) treated with interferon (a-2a recombinant, a-2b recombinant, a-2c recombinant, natural leukocyte a-IFN). A biochemical response (decreased tumor plasma or serum marker) occurred in 44%, decreased tumor size in 11%, [399] and tumor stabilization in 35%. Of the 58 patients with PETs in another review of 322 patients, [361] 51% had a biochemical response and 12% had a decrease in tumor size occurring with a mean duration of treatment of 20 months (range, 2–96 months). [361] Patients with VIPomas had a higher response rate (83%, 10 of 12) than other PETs. Twenty-eight of the 58 patients had received chemotherapy prior to treatment with interferon and their biochemical response rate was not different from those without previous chemotherapy (55% vs. 44%). [361] Disease stabilization occurred in 14 of 58 (24%) patients with a mean duration of 16 months. In a number of studies a-interferon treatment rarely resulted in a decrease in size of either a malignant PET (see Table 97-8) or carcinoid tumor (0–12%); however, in a number of studies interferon treatment inhibited further tumor growth in 24% to 80% of patients with metastatic neuroendocrine tumors (see Table 97-8). [361], [399] These studies suggest a-interferon as a primary tumoricidal agent is minimally effective; however, it has tumoristatic activity in a significant percentage of patients. [361], [364] Another study [400] reports a-interferon can stimulate an increase in bcl-2 expression in neuroendocrine tumors (NETs) (i.e., carcinoids and PETs) and this effect may stabilize the cells at G_0 and contribute to this tumoristatic effect. Whether a-interferon treatment extends survival is unknown. Interferon has also been used in combination with a number of agents, such as 5-FU, [349] but it is not clear that this increases its antitumor effect (see Table 97-8). Also, a-interferon is increasingly being used in combination with somatostatin analogs [401], [402] (see Table 97-8). This is discussed in the following section.

Somatostatin analogs. A number of studies in animals have demonstrated that somatostatin analogs can inhibit the growth of various tumors under various experimental conditions. [169] Studies demonstrate that except for insulinomas, more than 90% of PETs possess somatostatin receptors. [232], [233], [403] In one review of cases reported up to 1988, [368] hormonal therapy with the long-acting somatostatin analog octreotide was reported to cause a decrease in tumor size in 12% of patients with NETs (see Table 97-8). In more recent studies 0% to 8% of patients were reported to experience decrease in NET size. [58], [273], [319], [370], [372], [398], [404], [405], [406] and [407] However, disease stabilization is reported in 25% to 88% of patients with metastatic NETs treated with somatostatin analogs. [315], [319], [320], [406], [407] These results demonstrate that octreotide, similar to interferon, has minimal tumoricidal activity alone in PETs. However, in a significant number of patients it stabilizes the disease, preventing additional progression, but it remains unclear whether it causes stabilization for a significant period of time to increase survival. Tumor stabilization is also reported with depot-sustained release formulations of somatostatin analogs. [319], [320], [405] A study [408] demonstrates somatostatin analogs can induce apoptosis in NETs and this may be an important factor in its tumoristatic effects.

The combination of somatostatin (either SC or long-acting forms) with a-interferon is reported to cause disease stabilization in 57% to 89% of patients with malignant NETs, some of whom did not previously respond to either agent alone (see Table 97-8). [401] , [409]

Hepatic artery embolization/chemoembolization. Hepatic artery embolization with or without postocclusion chemotherapy (streptozotocin, doxorubicin, 5-FU, or DTIC) has been used successfully in patients with various PETs. [2] , [337] , [358] , [359] , [410] , [411] , [412] , [413] , [414] , [415] , [416] and [417] Because the liver derives only 20% to 25% of its blood supply from the hepatic artery and 75% to 80% from the portal vein, and because most PETs are vascular with an arterial supply, hepatic artery embolization can be used if the portal vein is patent to treat metastatic disease to the liver. In some studies 60% to 100% of patients with metastatic NETs have symptomatic improvement and 33% to 80% have an objective response. [11] , [413] , [416] , [418] Chemoembolization with doxorubicin in iodized oil combined with gelatin or sponge particles, or with streptozotocin, 5-FU, cisplatin, or mitomycin C is reported to improve symptoms in 68% to 100% of patients and to decrease tumor size and/or hormone levels in 37% to 100% of patients. [350] , [412] , [415] , [416] , [417] , [418] , [419] and [420] Mean duration of objective responses lasts from 6 to 42 months. [412] , [416] , [418] This procedure is not without significant side effects. In the literature [410] the mortality overall for this procedure is less than 3% of cases, but pain develops in almost 100%, fever and leukocytosis are seen in at least 50% of patients, and 5% to 15% are reported to have severe complications including hepatic failure, bleeding, infection, or acute renal failure. [358] , [419] In almost all patients some abdominal pain, nausea, vomiting, and fever occur, which lasts up to 10 days. [58] , [412] , [416] , [420] Furthermore, in patients with metastatic PETs, in up to 31% of patients with metastatic gastrinoma to the liver, metastases to bone can occur, [54] suggesting that procedures directed at the liver, such as embolization, may be of limited value to control metastatic spread in some patients with extensive disease. However, in a patient with a hormonal syndrome that cannot be controlled by other therapies, this procedure may be helpful. [2] , [24] Hepatic artery embolization has been combined with treatment with a-interferon [11] , [24] , [421] in patients with metastatic carcinoid tumors, which closely resemble PETs in responsiveness and histology. Five of seven patients (71%) one year after the treatment continued to demonstrate a decrease in the metastatic disease in the liver, whereas with interferon alone (n = 10) only 10% demonstrated a decrease. In another study [421] involving 36 patients with metastatic carcinoid tumors, interferon alone resulted in a 24% response rate (decrease in 5-HIAA secretion) and survival rate was 40% after 5 years. In contrast [421] with interferon and hepatic artery embolization, the response rate was 60% and 5-year survival rate was 75%.

Cytoreductive surgery. The systematic removal of all resectable tumor (i.e., cytoreductive or debulking surgery) may prolong the life expectancy of some patients with some functional PETs. [2] , [11] , [12] , [29] , [213] , [214] , [272] , [276] , [277] , [322] , [354] , [355] Systematic removal of all resectable disease has been reported to help control symptoms in patients with malignant insulin-omas, [277] , [328] , [422] glucagonomas, [19] , [51] , [141] , [277] VIPomas, [18] , [19] , [277] GRFomas, [21] somatostatinomas, [277] and nonfunctional PETs [1] , [29] and has led to the suggestion that resection should be performed if possible, if extensive tumor is identified at the time of surgery and resection can be done without increased risk. No studies have evaluated cytoreductive surgery in a controlled, prospective manner. It is important to distinguish between the possible benefit of such surgery in patients who are asymptomatic (nonfunctional tumors) or have symptoms well controlled (gastrinomas, effective medical control of other tumors), from patients in whom symptoms are not controlled. In the former group cytoreductive surgery is performed to extend survival, but no controlled studies have established its usefulness. In the latter case there are numerous examples previously cited with each of the symptomatic PETs where cytoreductive surgery improved symptoms. A number of studies [214] , [276] , [329] , [423] and two reviews [213] , [416] have provided data regarding this approach. It is important to remember that surgical removal of all visible tumor is only possible in a very small proportion of all patients with PETs with metastatic disease (9% in one study, [423] 5% and 15% in two others [214] , [272]). In one study [214] of 80% of cases with metastatic disease deemed possibly surgically resectable, the tumor was completely excised at surgery and survival was 79% at 5 years with a mean follow-up of 3 years. In this study [214] patients with extensive metastatic disease postresection had a 5-year survival equal to that of patients with inoperable tumors of 28%, whereas patients with limited metastatic disease at surgery had a significantly prolonged survival ($P = 0.019$). In another study [276] 36 extended hepatectomies or hepatic lobectomies and 38 nonanatomic liver resections were performed in patients with advanced disease with malignant NETs. Perioperative mortality was 2.7%, morbidity 24%, and 4-year survival was 73%, with 90% of patients having symptomatic improvement. [276] Norton and colleagues [272] reported the successful resection of all metastatic disease in 5 of 20 (25%) patients with metastatic gastrinomas with extensive disease, including two patients who maintained normal fasting gastrins postresection. The conclusion of each of these studies [213] , [214] , [272] , [276] , [329] was that resection of metastatic disease should be considered in selected patients with metastatic NETs if all gross disease can be resected safely. It is important to remember, however, that this approach is only possible in approximately 10% of patients. It has not been systemically evaluated, and it remains unclear whether patients identified with metastatic disease by localization studies prior to surgery and with symptoms well controlled by medical management will benefit long-term by debulking surgery. In fact one review [355] injects a note of caution in the use of aggressive cytoreductive surgery, pointing out that this surgery can be associated with a high complication rate and that many of these patients survive a prolonged period of time even with advanced disease. In this review [355] the authors recommend cytoreductive surgery be performed in patients with metastatic PETs only if the PET is potentially fully resectable. If it is not fully resectable at surgery, they recommend refraining from performing a partial resection.

Liver transplantation. Liver transplantation is increasingly being used in patients with metastatic NETs (PETs and carcinoid tumors). [11] , [56] , [58] , [374] , [375] , [376] , [377] , [378] and [379] , [424] , [425] The use of hepatic transplantation for many metastatic tumors, such as colorectal carcinoma, cholangiocarcinoma, or metastatic sarcomas, has been largely abandoned because of early recurrence. [378] However, because of their more indolent biologic behavior, patients with metastatic NETs are considered more likely to benefit from liver transplantation. [378] , [424] The reports are generally confined to small numbers of patients with limited follow-up. [56] , [374] , [375] and [376] , [424] In a review [424] of reports of liver transplantation in patients with malignant NETs (n = 103) (48 PETs, 43 carcinoids), the 5-year survival rate was 45%. Occasional long-term cures are reported. [424] , [425] It was concluded that liver transplantation may be justified in younger patients with metastatic disease confined to the liver. [424]

Somatostatin receptor-mediated radiotherapy. The presence of high densities of somatostatin receptors on almost all NETs (PETs and carcinoids), [169] , [232] , [233] , [300] which is now widely used to localize these tumors, [169] , [232] , [233] , [257] is now being investigated as a means of possible antitumor treatment using radiolabeled analogs. [58] , [380] , [381] and [382] , [426] , [427] Both [^{111}In-DTPA ^{0}]octreotide, which emits conversion and auger electrons as well as somatostatin analogs usually coupled by a DOTA group (1, 4, 7, 10,-tetraazacyclododecane-N, N', N'', N''' tetraacetic acid) to yttrium 90 (^{90}Y), which strongly emits ß-particles, or lutetium 177 (^{177}Lu) which is a ß- and ?-emitter, are reported to have antitumor effects in animal studies and small studies in humans. [381] , [382] , [426] , [427] , [428] , [429] , [430] , [431] , [432] , [433] and [434] Of 20 patients [380] with advanced NETs who received at least 20 Gbq of [^{111}In-DTPA ^{0}]octreotide, in 14 patients (70%) either a decrease (30%) or tumor stabilization (40%) occurred. In two reports involving 20 patients and 39 patients with various malignant tumors including NETs, [430] , [434] administration of a ^{90}Y-labeled octreotide analog resulted in 80% to 86% showing a tumor growth response with 21% to 25% demonstrating a decrease in size and 49% to 55% tumor stabilization. Renal toxicity occurred in 3% to 5%. [430] , [434] Lutetium 177-DOTA-Tyr ^{3}-octreotide was used in 26 patients mostly with metastatic NETs, who received at least 300 mCi. [433] Eighty-eight percent demonstrated tumor stabilization (54%) or a decrease in size (34%). Although the use of ^{111}In-, ^{90}Y-, or ^{177}Lu-or labeled somatostatin analogs represents an exciting new approach to target tumor cytotoxicity, at present, too few patients with NETs have been treated to assess either its toxicity or efficacy.

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CHAPTER 98

David C. Whitcomb

HEREDITARY DISEASES OF THE PANCREAS

GENETIC PREDISPOSITION TO PANCREATIC DISEASE

Determining genetic predisposition to pancreatic disorders was recently recognized as one of the most important areas of research and clinical practice related to the pancreas. The importance of this rapidly developing field centers on the recognition that most patients with idiopathic pancreatitis have a strong genetic predisposition and that other forms of pancreatic disease, including alcoholic pancreatitis, appear to be strongly influenced by gene-environment interactions. [1] A number of pancreatic disorders are now known to be associated with germ-line mutations (Table 98-1). The focus of this chapter is on the genetic factors underlying various types of pancreatic disease, with application of these findings to major disorders. In addition, the management of disease-specific problems is addressed.

TABLE 98-1 Pancreatic Disorders Associated with Germline Mutations

Definitions and Nomenclature

Clear understanding of the relationship between gene mutations and pancreatic disease requires use of several specific terms. Key terms for this discussion include hereditary pancreatitis, familial pancreatitis, mutation, DNA sequence variation, polymorphism, gene, modifier gene, monogenic and polygenic traits or disorders, orthologous genes, and synteny. In addition, the primary nomenclature systems are reviewed (see Online Links at the end of this chapter for up-to-date information on nomenclature).

Hereditary pancreatitis specifically refers to otherwise unexplained pancreatitis in an individual from a family in which the pancreatitis phenotype appears to be inherited through a disease-causing gene mutation expressed in an autosomal dominant pattern. [2], [3] *Familial pancreatitis* refers to pancreatitis from *any* cause that occurs in a family with an incidence that is greater than would be expected by chance alone, given the size of the family and incidence of pancreatitis within a defined population. Familial pancreatitis may or may not be caused by a genetic defect. Hereditary pancreatitis therefore defines a narrow, specific term, whereas familial pancreatitis is a broad, general term that encompasses hereditary pancreatitis. These distinctions are especially important from prognostic and genetic counseling perspectives because of the very high risk and clear clinical course of hereditary pancreatitis.

The term *mutation* refers to an event, wherein a DNA base or bases are substituted, inserted, or deleted, resulting in a new genomic DNA base sequence, or the process by which a gene undergoes a structural change. [4] The term mutation can also be used to describe a modified gene resulting from a mutation. [4] The result of a mutation is *DNA sequence variation*. A *polymorphism* refers to the existence of two or more genetically different classes (e.g., DNA sequence variances) in the same interbreeding population. [4] A *s*ingle genomic DNA *n*ucleotide *p*olymorphism is *SNP* (pronounced “snip”). The terms polymorphism and mutation are often used with more specific intent. For example, the term *polymorphism* may refer to commonly observed DNA sequence variation (e.g., seen in greater than 1% of a population), and the term *mutation* is commonly used to describe a DNA sequence variation that is disease-associated.

The location of a genomic DNA sequence variation is usually communicated using the nucleotide number of a reference sequence with the change following the number. For example, the cationic trypsinogen R122H mutation could be designated by the GenBank accession number (U66061, 133283G>A), or preferably an official National Center for Biotechnology Information (NCBI) Reference Sequences (see RefSeq http://www.ncbi.nlm.nih.gov/LocusLink/refseq.html) for mRNA (NM_002769, 367G>A), or a genomic contig (NT_007769, 133283G>A).

A *gene* is a DNA segment that contributes to phenotype, function, or both. [5] If there is an absence of demonstrated function, a gene may be characterized by sequence, transcription, or homology, [5] homology meaning similar structure, function, or both. [6] Human gene mutations are preferably described using the HUGO Mutation Database nomenclature. [7] (see HGMD, Human Gene Mutation Database: http://www.uwcm.ac.uk/uwcm/mg/hgmd0.htm). In this system, the location of a specific DNA sequence variation in a gene is determined using the reference sequence, and then numbering the sequential nucleotides of the gene beginning at the A of the ATG of the initiator Met codon. The designation of the position of a nucleotide change is as above with the caveat that a “g.” should precede the designation for genomic sequence and “c.” for cDNA. Deletions are designated by “del” after the nucleotide number (e.g., 1997delT) and insertions by “ins” after the nucleotide interval number (e.g., 1997-1998insT). [7] For amino acid nomenclature, the single-letter amino acid symbol is preferred, and the format is R122H (arginine at codon 122 substituted by histidine). The “wild-type” amino acid is given before and the mutation-associated amino acid after the codon number. Therefore, there is no confusion as to the significance of G, C, T and A in the nomenclature. [7] For intronic variations, the “IVS” designation is used. For example, IVS4+1G>T denotes the G to T substitution at nucleotide +1 of intron 4, and IVS4-2A>C denotes the A to C substitution at nucleotide -2 of intron 4. [7] For further details, see the “Recommendations for a Nomenclature System for Human Gene Mutations.” [7]

A *monogenic* disorder means that the genetic disorder occurs through mutations in a single gene. A monogenic disease could be either dominant or recessive. A *modifier gene* refers to the co-inheritance of another mutated gene that alone is not disease causing, but confers unique phenotypic features to a genetic disorder

(see the later section “ Meconium Ileus”). Polygenic disorders result from the combined action of mutant alleles from more than one gene when neither of the mutant genes alone is disease causing.

When similar genetic information is compared between species (e.g., mice and humans) several additional terms are used. The term *orthologus* means the same or homologous gene sequences in different species (e.g., rat versus human cationic trypsinogen), whereas the term *synteny* properly refers to the gene loci on the same chromosome in different species. [8] However, the term syntenic region is often used to compare regions of similar gene content between species when they are located within different chromosomal regions (e.g., the *trypsinogen gene cluster* on human chromosome 7, mouse chromosome 6, and rat chromosome 4).

Classification and Organization of Genetic Information

The explosion of information on genes and genetics spawned the creation of a number of important resources. Online Mendelian Inheritance in Man (OMIM) is a computerized database of human genes and genetic disorders supported by the NCBI, National Library of Medicine, National Institutes of Health. OMIM focuses primarily on inherited, or heritable, genetic diseases and serves as a phenotypic companion to the human genome project. [9]

The UniGene system was created by the NCBI to consolidate all of the redundant expressed sequences tags (short DNA sequences derived from reverse transcription of RNA extracted from various tissues). The system was designed to partition automatically all GenBank sequences, including expressed sequences tags, into a nonredundant set of gene-oriented clusters. Each UniGene cluster contains sequences that represent a unique gene and is linked to related information, such as the tissue types in which the gene is expressed. Each gene is identified by a unique descriptive name and UniGene symbol and number, which are coordinated with other genetics databases. In this chapter, the common names for genes or proteins are often used. If the common name of a gene (e.g., trypsinogen) and the UniGene name and symbol differ (e.g., protease, serine 1; *PRSS1*), then both are given with the initial use in the text.

Clinically Important Genetic Mutations

Mutations in three different genes clearly predispose to most gene-associated exocrine pancreas disorders in humans. The genes include the cationic trypsinogen (protease, serine, 1; *PRSS1*) gene, the pancreatic secretory trypsin inhibitor (serine protease inhibitor, Kazal-type, 1; *SPINK1*) gene, and the cystic fibrosis transmembrane conductance regulator (*CFTR*) gene. Although the mechanism and inheritance pattern of the disease-associated mutations in these genes are markedly different, they all cause *common* and *clinically significant* pancreatic disorders.

Determining whether a mutation is clinically important requires understanding of several interacting factors. [3] These factors include the *severity* of the mutation, the *prevalence* of the mutation in specific populations, and the *physiologic function* of the wild-type gene. The severity of a mutation reflects a change in the function of the encoded protein by altering the amount or location of expression, the protein function, the regulatory elements, or the ability to interact properly with other factors. [3] In general, more severe mutations are more likely to cause disease in an individual patient. The prevalence of a mutation in a population (or DNA sequence variation) also determines clinical importance. One can envision a large variety of very severe gene mutations that would disrupt the function of key proteins, but if they are never seen in patients, then they are of little clinical importance. [3] The physiologic function of the gene product also plays a critical role in determining whether the mutation is clinically important. In other words, does the gene play a pivotal role in a critical process? Are there adaptive or redundant mechanisms in place to lessen the impact of a mutation? Does the gene product play a key role in development, regulation, protection, or repair? Does the disease appear as an autosomal dominant, autosomal recessive, or complex genetic pattern, and does it require environmental factor or mutations in a second gene in order to become clinically evident? Indeed, growing experience with gene knock-out mice reveals that the importance and independence of many genes must be reconsidered. Thus, clinically important mutations must be determined by identifying mutations in well-characterized patient populations combined with adequately sized genetic epidemiology studies. In the following sections on pancreatic disease, recent data on the severity and prevalence of known mutations and on the physiologic role of the gene product are presented.

MAJOR GENE MUTATION–ASSOCIATED PANCREATIC DISORDERS

Cationic Trypsinogen (PRSS1) Gene Mutation–Associated Pancreatitis

Cationic Trypsinogen The first pancreatitis-associated mutation in the cationic trypsinogen gene (*PRSS1* OMIM *276000) was discovered in 1996 through efforts to identify the cause of hereditary pancreatitis. [10] The role of trypsin in pancreatic physiology and the protective mechanisms employed by the pancreas to control prematurely activated trypsin are covered as a background to understanding mutation-associated pathophysiologic mechanisms predisposing to pancreatitis.
Physiology of trypsin. Cationic trypsinogen is the most abundant of three trypsinogen isoforms synthesized in the pancreas and represents about 23% of total pancreatic secretory proteins in humans. [11] The other two isoforms, anionic trypsinogen (protease, serine, 2; *PRSS2;* OMIM *601564) and mesotrypsinogen (protease, serine, 3; *PRSS3*) make up about 16% and less than 0.5% of the total pancreatic secretory protein by weight, respectively. [11], [12] The specific amino acid targets for trypsin are the arginine (symbol Arg or R) or lysine (symbol Lys or K) residues within the target peptides or proteins. Trypsinogen becomes enzymatically active *trypsin* after the intestinal brush-border brush enzyme enterokinase or another active trypsin molecule cleaves the 8–amino acid N-terminal peptide fragment called *trypsinogen activation peptide* (TAP) (Fig. 98-1). Trypsin activates not only trypsinogen but also the other pancreatic digestive proenzymes in an activation cascade. These pancreatic digestive enzymes digest the major, complex nutrient molecules within a meal. The pancreatic enzymes are then destroyed, usually before the mid-ileum.

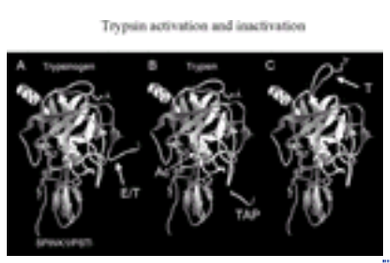

FIGURE 98-1. Trypsin activation and inactivation. This figure shows three variations of the trypsin-SPINK1 complex. **A:** Trypsinogen is the top portion of the complex with arginine 122 (extending from the loop at the top). The *arrow* shows the site of the TAP portion of the trypsinogen molecule where either enterokinase (E) or trypsin (T) can cleave this peptide and activate trypsinogen to trypsin. **B:** Trypsin is generated when TAP is released and the molecule undergoes conformational changes to become active (Ac, active site). **C:** The side chain containing the critical R122 site can be attacked by another trypsin (T) when it is in a susceptible conformation (From ref. [14]).

Pancreatic acinar cell protection from trypsin. Because of the efficiency of the activation cascade and the destructive potential of pancreatic digestive enzymes unleashed in the wrong location, it is imperative that trypsin activity within the pancreas be severely limited. The pancreatic acinar cells synthesize pancreatic secretory trypsin inhibitor (*SPINK1*) (see Fig. 98-1), which is regulated as an acute-phase reactant. [13] *SPINK1* provides the first line of defense against prematurely activated trypsin within the pancreas. [10] A second protective mechanism is trypsin self-destruction or autolysis beginning at arginine 122 of the trypsin primary amino acid chain (see Fig. 98-1). The autolysis site is often inaccessible to hydrolysis, especially with elevated calcium, so that it cannot be attacked by a second trypsin. [14], [15] Thus, trypsin both activates trypsinogen by cleaving TAP and later inactivates trypsin by cleaving the side chain at R122. Additional protective mechanisms are also operational. [12], [16]
Pancreatitis-Associated Trypsinogen Mutations Several mutations have been identified within the cationic trypsinogen gene that cause acute and chronic pancreatitis. [17] The number of mutations is limited and the type of mutations specific because they cause pancreatitis by altering the protein through a gain-of-function mechanism by enhancing the activation of trypsinogen or preventing inactivation of trypsin. [18] Gain-of-function mutations often result in an autosomal dominant inheritance pattern because only one of the two alleles must be affected to express the abnormal phenotype; that is, half of the protein is out of control. Loss-of-function mutations are usually autosomal recessive because both copies of the gene product must be *lost* to cause the disease phenotype (e.g., cystic fibrosis). In the case of hereditary pancreatitis, most families have identifiable gain-of-function mutation in the cationic trypsinogen gene, with R122H and N29I being the most prevalent. These two mutations represent the causative mutation in about two thirds of the families with classical hereditary pancreatitis [19] and currently are the only mutations for which genetic testing is recommended. [20] (Table 98-2 and Table 98-3).

1. Recurrent (two or more) attacks of acute pancreatitis for which there is no explanation (anatomic anomalies, ampullary or main pancreatic strictures, trauma, viral infection, gallstones, alcohol, drugs, hyperlipidaemia, etc.)
2. Unexplained (idiopathic) chronic pancreatitis
3. A family history of pancreatitis in first-degree (parent, sibling, child), second-degree, (aunt, uncle, niece, nephew), or third-degree (grandparent, first cousin) relative
4. An unexplained episode of documented pancreatitis occurring in a child that has required hospitalization and where there is significant concern that hereditary pancreatitis should be excluded
5. For a patient with pancreatitis eligible for an Ethics Committee– or Institutional Review Board (IRB)–approved research protocol

Modified from ref. 20.

TABLE 98-2 Indications for Genetic Testing for *PRSS1* Mutations

Before genetic testing, the patient should understand:
1. Why the test has been suggested and the need for documented informed consent
2. The implications of finding a pancreatitis-related mutation in the *PRSS1* gene for the health and medical care of that patient
3. How their genetic test result will be communicated to them and who else will be informed of their result (i.e., the clinician that has requested that test, other involved pancreatic specialists, the family doctor, hospital medical records, insurance carrier)
4. The availability of genetic counseling after the test result is known
5. The pancreatic cancer risk and the possible adverse health and life insurance and employment consequences for the patient (if not safeguarded against by state or national legislation)
6. The implications of a positive genetic test result for their relatives
7. Whether or not they wish for their test sample to be used for any research project and by what (anonymized) route this will occur

Modified from ref. 20.

TABLE 98-3 Pregenetic Test Patient Information for *PRSS1* Mutations

Clinical Characteristics of Hereditary Pancreatitis About two thirds of the families with hereditary pancreatitis appear to have cationic trypsinogen mutations, and one third have unidentified mutations. [19] The clinical phenotype of the trypsinogen-associated mutations and noncationic trypsinogen associated mutations are similar, and thus the topic is addressed here.

Hereditary pancreatitis disease phenotype. Hereditary pancreatitis is a genetic disorder with high disease penetrance; about 80% of individuals with a cationic trypsinogen R122H or N29I genotype in most studies develop pancreatitis at some time in their life (80% disease penetrance). The reason for the incomplete penetrance is unknown and appears to be independent of major environmental factors or modifier genes. [21] The clinical presentation is usually recurrent episodes of acute pancreatitis beginning in childhood (median age of onset, 10 years), although the range is from infancy to the fifth or sixth decade of life. [22] Episodes of acute pancreatitis are similar to acute pancreatitis from other causes and may vary from mild abdominal discomfort to severe life-threatening episodes with pancreatic necrosis, splenic vein thrombosis, pseudocyst formation, and death. However, in some patients, an episode of acute pancreatitis may be prolonged. Half of the patients with episodes of acute pancreatitis progress to chronic pancreatitis. [19], [23] Patients with hereditary pancreatitis-associated chronic pancreatitis suffer from all of the common complications of alcoholic and idiopathic chronic pancreatitis, including unrelenting pain, parenchymal and ductal calcifications, duct distortion, fibrosis, maldigestion, and diabetes mellitus. [22], [24], [25], [26], [27] and [28] These features make hereditary pancreatitis indistinguishable from other causes of acute and chronic pancreatitis except for the relatively early age of onset, the autosomal dominant inheritance pattern, and the lack of other identifiable etiologies. [18] A high incidence of pancreatic cancer is another striking feature of hereditary pancreatitis, beginning 30 to 40 years after the onset of pancreatitis. [29], [30] The estimated accumulated risk for pancreatic cancer to age 70 in these families is about 40 percent. [28], [29], [30], [31] and [32] A remarkable gene-environment interaction is related to tobacco smoking. The age- and sex-adjusted odds ratio is doubled by tobacco smoking, and the median age of diagnosis of pancreatic cancer is 20 years earlier in the smokers. [29], [30] Thus, the evidence is very strong for a connection between hereditary pancreatitis and pancreatic cancer.

Diagnosis of hereditary pancreatitis. The diagnosis of hereditary pancreatitis remains an area of controversy. Reasons include lack of consensus on a precise definition and that the implications of this diagnosis will persist throughout life. Furthermore, the diagnosis has implications for future descendants and other family members and may affect social and reproductive choices, employment, and insurability. [33], [34] and [35] Before genetic testing, the diagnosis of hereditary pancreatitis was based solely on evaluation of the patient's extended family and exclusion of other causes of pancreatitis. [36] Genetic testing offers a new and powerful tool because it is highly accurate, provides precise risk assessment, and can detect or exclude the likelihood of pancreatitis *before* symptoms develop. [33] Furthermore, genetic testing can detect hereditary pancreatitis–associated mutations in individuals without a clear family history of pancreatic disease. Indeed, the cationic trypsinogen mutations N29I and R122H have been detected in families that appear to have autosomal dominant, autosomal recessive, or familial patterns or patients with idiopathic (sporadic) pancreatitis. [19] Therefore, it is currently our practice to diagnose patients with hereditary pancreatitis only if they have a clear family history or have unexplained pancreatitis or recurrent pancreatitis-like pain with confirmed cationic trypsinogen R122H, N29I, or equivalent mutations.

Genetic testing for cationic trypsinogen mutations. Genetic testing should be approached with caution. [33], [34] The results of genetic testing are highly accurate but may have broad implications for the patient's future health, family, employment, and insurability. The clinician must therefore understand the implications of testing, be prepared to provide pretest and posttest counseling to the patient (or refer the patient to a genetic counselor), and ensure that informed consent is obtained before testing. [33] Genetic testing is now available for the cationic trypsinogen R122H and N29I mutations [1] through an approved laboratory (in the United States an approved laboratory would require a Clinical Laboratory Improvement Act license [33], [35]). Guidelines for testing have recently been adopted by an expert consensus conference group [20] and are summarized in Table 98-2 and Table 98-3. The primary indications for cationic trypsinogen mutation testing include recurrent idiopathic acute pancreatitis, idiopathic chronic pancreatitis, verification of a clinical suspicion in a family member of a kindred with known mutations, to help a patient understand or validate his or her condition, and to assist individuals in making lifestyle decisions (e.g., reproduction, diet, smoking) based on the known risk for pancreatitis and potentially pancreatic cancer. [1], [29], [33] Genetic testing is also used in children with unexplained pancreatitis or episodes of pancreatitis-like pain when there is significant concern about the possibility of hereditary pancreatitis. Indeed, identification of an established pancreatitis-associated gene mutation can be valuable in expediting an expensive and prolonged evaluation of recurrent pancreatitis in children and precludes further evaluation of elusive causes of pancreatitis in adults. The positive and negative predictive values of a genetic test in identifying specific mutations are almost perfect with properly applied modern techniques. [33], [35] Interpretation of test results and explanation of their meaning to the patient continue to be a central issue because positive or negative test results have lifelong implications for the patient as well as the patient's extended family. A positive test result in a clinically *unaffected* person is interpreted as conferring a significant increased risk for pancreatitis, with this risk possibly diminishing with age. A negative test result in a family with a known mutation essentially eliminates the risk for this genetic form of pancreatitis. If a mutation has not been previously identified in the family, then a negative test result in an unaffected person is considered noninformative because one cannot distinguish whether the tested individual is free from genetic risk or has inherited a different pancreatitis predisposing gene mutation. [1] The genetic testing of children raises unique issues. [20] Unlike an adult patient, a child legally cannot provide informed consent. Thus, the decision for a child is essentially left to the parents or legal guardian. For children 7 years and older, a parent or legal guardian may provide consent for genetic testing, although these older children should provide assent, or agreement to the testing. The testing of purely asymptomatic children is strongly discouraged because there is no clear medical benefit in identifying carriers at a young age. [20], [33] Testing for the purpose of intervention with diet, medication, or surveillance for complications of a genetic disorder (e.g., undertaking repeated colonoscopies for patients with the familial adenomatous polyposis syndrome) has been advocated. [37] In hereditary pancreatitis families, alcohol, emotional stress, and fatty foods have been reported to precipitate pancreatitis attacks, [25] and smoking increases the risk for both pancreatitis [38], [39] and [40] and pancreatic cancer. [30] Testing for the purpose of encouraging mutation-positive older children to avoid these excesses is advocated by some caregivers. However, avoidance of fatty foods, alcohol, and tobacco represents excellent general medical advice and therefore provides no compelling reason for genetic testing. [33] In either case, the personal desires of older children to postpone testing or to proceed with testing to relieve their own anxieties and learn more about their own personal health must also be carefully considered. Ownership of test results in children must be addressed. [1]

Treatment of hereditary pancreatitis. No specific treatment exists for the prevention or treatment of hereditary pancreatitis. However, some patients report that vitamins, antioxidants, or digestive enzyme supplements are helpful. [41] Symptomatic improvement was documented in a controlled trial of three children given antioxidants, vitamins, and selenium. [42] Symptomatic treatment for pancreatic duct obstruction or other sources of pain is also helpful and should be handled in a manner similar to other forms of pancreatitis. [43] Definitive recommendations for specific treatments await clinical evidence from well-designed trials. [41] A major concern for patients with hereditary pancreatitis is pancreatic cancer. Tobacco smoking appears to be the most important independent risk factor [29], [30] and should be strongly discouraged. Unfortunately, no good screening test exists for the early diagnosis of pancreatic cancer in high-risk groups. [32], [44] A recent consensus conference concluded that at the present time, screening for pancreatic cancer should be performed in high-risk individuals within multicenter Institutional Review Board–approved protocols. [45] If a patient in a high-risk group is to undergo an attempted curative resection of the pancreas for a suspected tumor, the whole pancreas should be removed. [45]

Pancreatic Secretory Trypsin Inhibitor (SPINK1) Gene Mutation–Associated Pancreatitis

An association between mutations in the *SPINK1* gene and idiopathic chronic pancreatitis in children was first described in 2000. [46] The predominant *SPINK1* mutation was N34S, which is present in about 1% to 2% of the general population. *SPINK1* mutations are commonly associated with familial pancreatitis and idiopathic pancreatitis developing before the age of 20 years. [47] *SPINK1* mutations are also associated with other forms of alcoholic pancreatitis (6%) [48] and occur in a high percentage of patients with fibrocalculous pancreatic diabetes, a subtype of tropical pancreatitis. [49], [50] *SPINK1* mutations are seen to a lesser extent in tropical calcific pancreatitis [51] and even in early-onset non–insulin-dependent diabetes mellitus in Bangladesh. [51]

Physiology of the Pancreatic Secretory Trypsin Inhibitor, *SPINK1* *SPINK1* appears to play a major role in protecting the pancreas from prematurely activated trypsinogen. [12] The *SPINK1* gene codes for a 56–amino acid mature peptide (see Fig. 98-1) that is synthesized in pancreatic acinar cells as well as other sites. [12] *SPINK1* normally inhibits trypsin by directly blocking the active catalytic site. A lysine target substrate, or "bait," for active trypsin is located at the apex of the substrate-inhibitor binding loop. The lysine fits into trypsin's specificity pocket, and *SPINK1* forms a stable complex with a covalent bond between the catalytic serine residue of the enzyme and the lysine carboxyl group of the reactive site of the trypsin inhibitor. [12] The binding is reversible, and *SPINK1* is also slowly digested by trypsin. [12] *SPINK1* inhibits anionic trypsin (*PRSS2*) but not mesotrypsin (*PRSS3*). [12] It has been estimated that the pancreas contains enough inhibitors to neutralize about 20% of total potential trypsin activity, [12] although this ratio probably varies depending on physiologic conditions. Because *SPINK1* and trypsinogen are both synthesized within the acinar cells, it is likely that *SPINK1* provides the first line of defense against prematurely activated trypsinogen (Fig. 98-2). Therefore, a *loss* of *SPINK1* function should hypothetically increase the chance of free trypsin activity inside the pancreas by allowing active trypsin to overcome more easily the first line of defense. [46], [47]

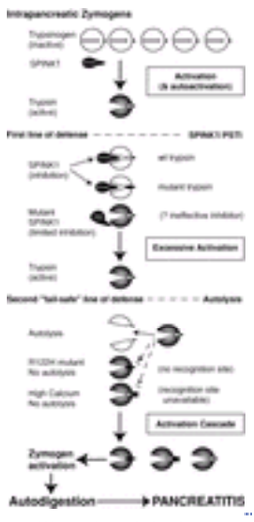

FIGURE 98-2. Mechanistic model of trypsinogen activation and inactivation within the pancreas. Tyrpsinogen (inactive) and *SPINK1* (also known as pancreatic secretory trypsin inhibitor, or *PSTI*) are synthesized together within the pancreatic acinar cells at a 5:1 ratio (*top*). Trypsinogen activation occurs within the acinar cell and threatens to initiate the zymogen activation cascade, leading to pancreatic autodigestion and pancreatitis. The first line of defense is trypsin inhibition by *SPINK1/PSTI*. *SPINK1* effectively inhibits up to 20% of potential trypsin (including mutant trypsin). If there is excessive trypsin activation (>20%) or ineffective inhibition by mutant *SPINK1*, then free trypsin activity rises and again threatens to initiate the activation cascade. The second line of defense is trypsin autolysis. This process begins with hydrolysis of the side chain connecting the two globular domains of trypsin at arginine 117 (R117 using the chymotrypsinogen numbering system [208]) coded for by codon 122 (codon numbering system [7]). Autolysis fails in hereditary pancreatitis with R122H mutation and possibly others, or under high calcium conditions. As free trypsin levels rise, the zymogen activation cascade is activated, leading to pancreatic autodigestion and acute pancreatitis. In hereditary pancreatitis and possibly other conditions, repeated attacks of acute pancreatitis lead to chronic pancreatitis. [209] (From ref. [1].)

Pancreatitis-Associated *SPINK1* Gene Mutations Based on the likely physiologic role of *SPINK1* in protecting the pancreas from active trypsin, it is predicted that pancreatitis-associated *SPINK1* mutations would be loss-of-function mutations. Therefore, a large number of disease-associated mutations are possible. The *SPINK1* N34S is the most commonly identified mutation in nearly all control populations, with prevalence of up to about 2% of control population (about 1% alleles frequency). [46], [47], [52], [53] This means that the *SPINK1* N34S mutation is *many* times more prevalent than chronic pancreatitis (1 in 16,000 or 0.006% of the population) and especially more common than idiopathic chronic pancreatitis in children. Patients with idiopathic pancreatitis may have *SPINK1* genotypes that are heterozygous, compound heterozygous, or homozygous for *SPINK1* mutations without a clear difference in age of onset or disease severity. [47] Therefore, the common *SPINK1* N345S mutation acts as an important *risk factor* or susceptibility factor for pancreatitis, potentiating the harmful effects of other genetic or environmental insults to the pancreas that actually initiate episodes of pancreatitis. [47], [54], [55]

Clinical Characteristics of Patients with *SPINK1* Mutations

Clinical phenotypes of patients with SPINK1 mutations. Unlike patients with cationic trypsinogen mutations, patients with *SPINK1* mutations and pancreatic disease do not have a single phenotype. [54] The highest incidence of *SPINK1* mutations is in patients with idiopathic chronic pancreatitis beginning before the age of 20 years. [47], [56] Depending on the regional population, between 15% and 25% of children have identifiable *SPINK1* mutations (usually N34S), [46], [47], [56] whereas in adults with idiopathic chronic pancreatitis or alcoholic chronic pancreatitis, the prevalence is about 5% to 10%. [47], [56]

Clinical presentation. A single clinical presentation for patients with *SPINK1* mutations has not been fully defined. The age of onset is usually younger than 20 years. In the United States, children may present with recurrent attacks of acute pancreatitis or with abdominal pain that is eventually determined to be due to chronic pancreatitis. In the Indian subcontinent, the first symptom may be diabetes associated with fibrocalculous pancreatic diabetes (see earlier). The number of attacks of pancreatitis and number of hospitalizations reported by patients with *SPINK1* mutations are similar to reports of patients with hereditary pancreatitis, with nearly 90% hospitalized more than once and 60% hospitalized more than 5 times. Review of the cases from the University of Pittsburgh database suggest that pancreatic calcifications are present at diagnosis in 16% of patients, pseudocysts in 19%, and dilated ducts in 26%. Insulin-dependent diabetes mellitus was also present in 13% of patients with pancreatitis associated with *SPINK1* mutations. These data suggest that the clinical characteristic of these patients is similar to other types of idiopathic chronic pancreatitis.

Genetic testing for SPINK1 mutations. Testing for *SPINK1* mutations in individuals with early-onset chronic pancreatitis may provide important information on predisposing causes of pancreatitis for the concerned patient. [1] However, most experts do not advocate genetic testing for *SPINK1* mutations at this time. [20] Furthermore, because fewer than 1% of patients with a heterozygous *SPINK1* mutation alone are likely to develop pancreatitis, the major reasons are lacking to do *presymptomatic* testing. If genetic testing is performed, appropriate pretest and posttest genetic counseling must be provided.

Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) Gene Mutation–Associated Diseases, Including Cystic Fibrosis and Pancreatitis

The *CFTR* gene (OMIM *602421) was discovered during the search for the cause of cystic fibrosis (CF). CF (OMIM #219700) has been the most common lethal genetic defect of white populations. CF is caused by severe mutations in the *CFTR* gene located on chromosome 7q32 and is inherited as an autosomal recessive disorder. [57]

Physiology of *CFTR* The *CFTR* gene contains more than 4300 nucleotides, divided into 24 exons that code for a single protein of 1480 amino acids. [57] The protein forms an anion channel for chloride and, to a lesser degree, bicarbonate. CFTR has 12 membrane-spanning regions divided into 2 membrane-spanning domains, 2 nucleotide-binding folds, and a regulatory domain (R domain) that is activated by cyclic adenosine monophosphate (cAMP). *CFTR* also regulates other ion channels in the same cell, including the amiloride-sensitive epithelial sodium channel. [58], [59] In the pancreas, the *CFTR* channel is located on the apical (lumenal) membrane of the pancreatic duct cell. [60] Binding of secretin to secretin receptors or vasoactive intestinal polypeptide receptor [61] on the basolateral membrane of the duct cells results in elevation of cAMP. cAMP activates protein kinase A (PKA) which, in the presence of ATP, interacts with the R domain of *CFTR* to open the *CFTR* anion channel. Opening of the *CFTR* channel results in efflux of intracellular chloride and depolarization of the duct cell. [62] *CFTR* activation is tightly linked to pancreatic bicarbonate secretion. [63] With cell depolarization, bicarbonate and sodium enter the duct cell through the basolateral sodium-bicarbonate cotransporter. [62], [64], [65] It is believed that bicarbonate secretion occurs on the apical side through a chloride-bicarbonate exchanger, [66] although *CFTR* is also permeable to bicarbonate with a Cl^- to HCO_3^- permeability ratio of 5:1, [67] and other bicarbonate channels probably exist. [67] *CFTR* also plays a key role in chloride and bicarbonate secretion in the

airways, intestine, bile ducts, and other organs.

***CFTR* Mutations in Typical and Atypical Cystic Fibrosis** Major mutations in *both* of the *CFTR* alleles result in complete loss of *CFTR* function and typical CF. The effects on cell and organ function include inability to hydrate mucus and other macromolecules adequately, leading to accumulation of viscid material and inspissated glands. This results in progressive organ destruction of the respiratory system and pancreas and to dysfunction of the liver, intestine, sweat glands, and other sites where epithelial cell secretion plays an important physiologic role. About 1000 mutations and sequence variations have been identified in the human *CFTR* gene. About 70% of white patients with CF have a 3—base pair deletion of the phenylalanine-coding codon 508 (?F508). [71] African Americans may have their own set of common CF mutations that originate from the native African population, including the 3120+1G>A mutation, which occurs with a frequency of 12.3% in a representative population. [68] Although there are many reported *CFTR* mutations, about 70 specific mutations represent more than 90% of all CF-causing mutations. [69]

Classification of CFTR mutations. The mutations in the *CFTR* gene have been classified based on their functional impact on the CFTR protein. Mutations affecting synthesis (class 1), maturation and trafficking (class 2), or activation (class 3) yield little or no functional protein and are considered severe mutations. [58, 70] Mutations that alter conductance (class 4) and protein abundance (class 5) and diminish, but do not eliminate, *CFTR* function are considered mild mutations and are often associated with pancreatic sufficiency or atypical CF. [58, 70] In most cases, so-called mild mutations are clinically mild only in regard to the severity of the pancreatic involvement, not overall disease severity or longevity. Some mutations affect the ability of *CFTR* to regulate other channels (class 6). [58] Finally, there are numerous silent mutations (no amino acid base change), intronic and exonic polymorphisms, and sequence variations with unknown significance. In general, the *less* severe mutation dictates the phenotype of autosomal recessive disorders such as CF and thus determines the phenotypic classification. Class 4 and 5 mutations deserve special mention because preservation of some *CFTR* function alters the eventual disease phenotype. For example, patients who have one or two mild *CFTR* mutations, such as R117H, R334W, R347P, A455E, or P574H, and therefore retain more than 1% *CFTR* function, are pancreatic sufficient. [71] If less than 1% of *CFTR* function exists, then the patient will develop typical CF with pancreatic insufficiency [69, 72] (Table 98-4). Sweat chloride becomes abnormal when less than 5% of function is preserved. With 5% of *CFTR* function or less, the patient also becomes susceptible to progressive pulmonary infection, even though the patient may maintain pancreatic sufficiency. Other features of CF, such as congenital bilateral absence of the vas deferens (CBAVD) with azoospermia, nasal polyps, or recurrent pancreatitis may occur any time *CFTR* function falls below 10% of normal.

PERCENTAGE OF NORMAL CFTR FUNCTIONS*	MANIFESTATIONS OF CYSTIC FIBROSIS
<1	Pancreatic exocrine deficiency (plus manifestations listed below)
<4–5	Progressive pulmonary infection (plus manifestations listed below)
<5	Clinically demonstrable sweat abnormality (plus manifestations listed below)
<10	Congenital absence of the vas deferens
10–49	No known abnormality
50–100	No known abnormality (this range represents the levels in asymptomatic heterozygotes and normal persons)

*The percentage of normal CFTR function is approximate.
Modified from ref. 69.

TABLE 98-4 Relation between the Amount of Functional *CFTR* Produced and the Phenotype

Some *CFTR* mutations may modify other *CFTR* mutations. Examples include the R553Q-?F508/?F508 phenotype, which partially reverses the ?F508 phenotype, or the R117H-intron 8 5T allele (*in cis*), which confers tissue-specific variations in expression of the mild R117H mutant CFTR protein. [58] Thus, a variety of genotype-phenotype combinations exists, and these can be broadly categorized based on the overall functional level of *CFTR*.

Other germline mutations that modify the cystic fibrosis genotype. Recent animal model and clinical epidemiologic evidence demonstrate that mutations in genes other then *CFTR* strongly influence the clinical features of CF. [73] The risk for meconium ileus was localized to a chromosomal region in mice, [74] which is syntenic to a locus on human chromosome 19 that was determined to be associated with meconium ileus in humans. [75] Other modifier gene mutations increase the risk for liver disease in CF patients. [73] The risk for liver disease appears to be independent of *CFTR* genotype, is associated with meconium ileus, [76] and is also associated with mutations in some protease inhibitors, [77] including a $_1$-antitrypsin. [73] The risk for pulmonary disease may also be influenced by modifier genes. For example, an a $_1$-antitrypsin enhancer polymorphism appears to be a genetic modifier of pulmonary outcome in CF. [78] Various combinations of *CFTR* mutations, modifier genes, and environmental factors also result in atypical CF features, including chronic pancreatitis, CBAVD with male infertility, disseminated bronchiectasis, chronic sinusitis, or other disorders without typical pancreatic and pulmonary manifestations of CF. [58, 79, 80]

Clinical Characteristics of Cystic Fibrosis The incidence of CF is 1 in 3200 white American newborns, 1 in 15,000 African American newborns, and 1 in 31,000 Asian American newborns. [81] On the other hand, CF is very rare in Japanese, with only 34 patients identified in a national survey of CF in Japan conducted in 1999. [82] CF is diagnosed within the first year of life in more than 70% of patients and in more than 85% by age of 5 years, but 8% remain undiagnosed until after the age of 10 years. [81] Presenting features during infancy include meconium ileus, maldigestion and malabsorption with frequent foul stools, recurrent pneumonia, failure to thrive, and rectal prolapse. When an infant presents with pancreatic insufficiency or meconium ileus, the etiology is usually CF caused by two severe *CFTR* mutations. Pulmonary function is normal at birth in patients with CF but later accounts for much of the morbidity and almost all of the mortality associated with CF. A number of factors other than the *CFTR* genotype contribute to the severity of lung disease, including chronic infection with *Pseudomonas aeruginosa,* nutritional status, environmental factors such as exposure to second-hand smoke, [83] and probably the effects of unidentified modifier genes. In older patients, the presenting symptoms of CF may include various pulmonary symptoms, nasal polyps, male infertility, liver disease, recurrent acute pancreatitis, or chronic pancreatitis, although the prevalence of *CFTR* mutations in patients with these common disorders is much lower than the prevalence of *CFTR* mutations in infants with pancreatic insufficiency or meconium ileus. A careful family history may also provide important clues to the diagnosis of CF. Features of CF that are commonly present at the time of diagnosis are given in Table 98-5.

FEATURE	PERCENT
Acute or persistent respiratory symptoms	50.5
Failure to thrive, malnutrition	42.9
Steatorrhea, abnormal stools	35.0
Meconium ileus, intestinal obstruction	18.8
Family history	16.8
Electrolyte imbalance	5.4
Rectal prolapse	3.4
Neonatal screening	2.3
Nasal polyps, sinus disease	2.0
Genotype	1.2
Hepatobiliary disease	0.9
Prenatal diagnosis	0.8
Other	1.2
Unknown	1.9

*Not mutually exclusive.
Data from the Cystic Fibrosis Foundation National Patient Registry. Modified from Table 1, CFF consensus conference Vol VII (section 1) March 25, 1996.

TABLE 98-5 Presenting Features in 20,096 Patients with Cystic Fibrosis

Organ-Specific Effects of *CFTR* Mutations

The pancreas in cystic fibrosis

Pancreatic pathology. Eighty-five to 90% of patients with CF have exocrine pancreatic dysfunction. [84] This illustrates the central role of *CFTR* in human pancreatic physiology compared with the *CFTR* knock-out mouse, which has relatively mild pancreatic changes. [85] In humans, pancreatic insufficiency may become increasingly evident in early life because pancreatic function is progressively lost. [86] In patients with typical CF, the pancreas becomes shrunken, cystic, fibrotic, and fatty. The histological appearance resembles other forms of chronic pancreatitis except for the inspissated secretions in the pancreatic ducts. Acinar cell atrophy, ductular hyperplasia, mild inflammatory changes, and progressive fibrosis gradually replace the pancreatic lobules. The islets of Langerhans are spared in most cases until late in the process and appear concentrated in the shrinking pancreas. [87] Radiographically, the appearance of the pancreas in CF can be quite variable. Ultrasonography, magnetic resonance imaging (MRI), and computed tomography (CT) scanning can document the progression of the chronic pancreatitis. Radiographically, the pancreas can appear normal, with incomplete or complete lipomatosis, as a cystic pancreas, macrocystic pancreas, or atrophic pancreas. [88, 89] and [90] The greatest sensitivity is provided either by MRI or CT scanning, but even with these methods, the correlation of abnormalities with the degree of exocrine dysfunction is poor. [88]

Exocrine pancreas dysfunction in cystic fibrosis. The progressive pancreatic damage occurring in patients with CF often begins in utero. On the other hand, the pancreas of an infant with severe *CFTR* mutations may appear normal. [86, 91] One of the first signs of pancreatic pathology in infants is hypertrypsinemia [92, 93] related to destruction of the pancreatic parenchyma. Pancreatic function progressively deteriorates and, in most cases, ends in complete pancreatic insufficiency. Pancreatic insufficiency in CF is usually suspected after observing abnormal stools and heralds complete pancreatic exocrine failure. Patients with retained pancreatic function may develop recurrent pancreatitis. Pancreatitis tends to be more problematic in older patients, with the reported incidence among patients older than 30 years being about 2.4%. [94, 95] In addition to pancreatic exocrine dysfunction, about one third of adults with CF have glucose intolerance, and about 10% develop cystic

fibrosis–related diabetes mellitus (CFRD) [96], [97] because the islets of Langerhans are destroyed. [98] CFRD remains a major problem, and a consensus conference report on diagnosis and management has recently been published. [99]

Recurrent acute and chronic pancreatitis associated with CFTR mutations. As noted earlier, compound heterozygous *CFTR* mutations with *CFTR* ^{SEVERE}/*CFTR* ^{MILD} genotype may have the CF phenotype with abnormal sweat chloride levels and pulmonary disease, while maintaining adequate pancreatic function for digestion. [71], [100] These patients are susceptible to acute pancreatitis. [95] Recent evidence suggests that many patients diagnosed with idiopathic chronic pancreatitis may harbor a combination of one severe mutation, such as ?F508, plus a milder mutation, such as *CFTR* R117H, which allows enough *CFTR* activity for most organs to function. [101] However, this combination of severe (class 1 to 3) and mild (class 4 to 5) *CFTR* mutations clearly predisposed individuals to recurrent acute and chronic pancreatitis. [101], [102] The diagnosis may be difficult in these cases because many mild *CFTR* mutations are not included in commercial *CFTR* testing (see later). Confirmation of diminished *CFTR* function should be done with nasal bioelectric potential difference measurements.

Pancreatic cancer risk. There is growing evidence that chronic pancreatitis from any cause is a risk factor for the development of pancreatic cancer. [103] Recent reports suggest an increased incidence of both pancreatic and intestinal cancers among CF patients compared with control populations. [104], [105] Cancers tended to occur in the third decade of life and often involved the esophagus, small and large intestines, stomach, liver, biliary tract, pancreas, and rectum. [105] Issues of prevention and screening likely will arise as more CF patients survive well into adulthood. However, at this time, there are no data on effective screening modalities for patients with chronic pancreatitis and high risk for pancreatic cancer. [45]

Gastrointestinal track disorders in cystic fibrosis. Patients with CF suffer from a wide variety of gastrointestinal problems extending from the esophagus to the rectum. [106], [107] The most common problems include gastroesophageal reflux and regurgitation, meconium ileus, distal intestinal obstruction syndrome, constipation, and rectal prolapse. The risk for gastrointestinal bleeding is increased because of unbuffered gastric acid, liver disease, and vitamin deficiencies. Symptoms of altered bowel habits, nausea and vomiting, heartburn, gastroesophageal reflux, bloating, flatus, constipation, and abdominal pain are common. [106]

Gastroesophageal reflux. Symptoms of gastroesophageal reflux occur in 20% of patients with CF. [108], [109] and [110] Esophagitis has been documented in up to 50% of patients with significant respiratory problems. [110], [111] Barrett esophagus has also been seen in a number of patients. [112] The reasons for reflux and esophagitis in CF are many, including excessive stomach acid and impaired acid buffering, body positioning during postural drainage of pulmonary secretions, diminished intrathoracic pressure related to lung disease, increased dietary requirements, and others. Treatment is identical to gastroesophageal reflux without CF.

Small intestinal complications of cystic fibrosis. The lumenal surface of the intestine is covered by a massive epithelial surface area. These cells express *CFTR,* and ion transport is affected by *CFTR* mutations. [61], [113], [114] The importance of *CFTR* in gut function is especially evident in mouse models, in which nearly all mice die of intestinal pathology unless there is specific intervention. [85] The CF knock-out mouse intestinal pathology parallels much of human intestinal pathology, including abnormal electrophysiologic profiles, goblet cell hyperplasia, mucin accumulation, crypt dilation, and intestinal obstruction similar to human meconium ileus. [85] Abnormal duodenal bicarbonate secretion is seen in CF patients, which partially explains the lower postprandial pH in the proximal duodenum of these patients compared with normal controls. [115], [116] and [117] Decreased activity of certain cytoplasmic peptide hydrolases in intestinal mucosa and reduced uptake of phenylalanine, isoleucine, and glycine have been found in patients with CF in comparison with control subjects, [118] but xylose absorption remains normal. [119] The small intestinal epithelial cells express a wide variety of ion channels, including non- *CFTR* chloride channels. On the other hand, a number of other transport abnormalities are also present, and may include other ion channels that are regulated by *CFTR*. [59] The variety of chloride channels in the colon is limited compared with the small intestine and respiratory system, [120] so that the defect in colonic function closely relates to the *CFTR* genotype. The loss of pancreatic digestive enzymes and resulting maldigestion aggravates the intestinal pathobiology. The lumen of mucosal glands in the small intestine of patients with CF often contains inspissated secretions. The changes may increase with age. The most severe alterations in the intestinal glands of the small bowel are usually found in patients with meconium ileus. [121] Mucus in CF is more abundant, stains more intensely, and contains more weak acid groups and protein than normally seen. [87] On radiographic examination, the duodenal folds usually appear thickened with nodular filling defects, mucosal smudging, dilations, and redundancy. [122] Radiographically, similar changes occur in the more distal small bowel, including thickening and distortion of jejunal folds and variable dilation of intestinal loops from the jejunum to the rectum. [123]

Meconium ileus. Clinically, the most severe complication of CF in the intestine is meconium ileus. However, distal intestinal obstruction syndrome, intussusception, rectal prolapse, and malabsorption also present common and significant problems. Meconium ileus is the presenting symptom in 10% to 20% of infants with CF and appears to be related to both *CFTR* genotype [100], [124] and a modifier gene on human chromosome 19. [75] The meconium is characterized by a striking decrease in the content of water and the presence of undegraded serum proteins, intestinal disaccharidases, and some lysosomal enzymes. The role of diminished pancreatic enzymes remains controversial because pancreatic involvement may be mild or normal in infants with meconium ileus. [91] Meconium ileus rarely occurs in infants without CF. The focal point of meconium ileus is the distal ileum, which appears narrowed, beaded, and filled with pellets of inspissated meconium. [125] Proximally, the ileal wall is hypertrophied and may become dilated with sticky, dark green-black meconium. Distally, the colon is unused. In addition, meconium ileus may be complicated by volvulus, atresia, or meconium peritonitis. [87] Meconium ileus classically manifests with signs of intestinal obstruction within 48 hours of birth in an infant who is otherwise well. In simple meconium ileus, no meconium passes, and there is progressive abdominal distention and, eventually, bilious vomiting. Dilated, firm, rubbery loops of bowel are visible and palpable through the abdominal wall, particularly in the right lower quadrant, and rectal examination is tight, productive of only a small mucous plug or a small amount of sticky meconium. [87] The complications of “complicated” meconium ileus involve volvulus, ileal atresia, a ruptured viscus, meconium peritonitis, or a combination of these.

Distal intestinal obstruction syndrome and constipation. The distal intestinal obstruction syndrome differs from meconium ileus and may reflect accumulation of undigested food residues, abnormal motility, dilation of the bowel, and dehydration. The distal intestinal obstruction syndrome may affect 3% to 10% of patients with CF. [107], [126], [127] Intussusception of the ileocolic region is a common complication of the distal intestinal obstruction syndrome. [107], [128] Distal intestinal obstruction syndrome may even be the presenting symptom of the disease. Symptoms include abdominal pain caused by constipation or fecal impaction, palpable cecal masses that may eventually pass spontaneously, and complete obstruction of the bowel by firm, putty-like fecal material in the terminal ileum, right colon, or both. [126], [129] The plain radiograph of the abdomen characteristically shows the proximal colon and distal small bowel packed with bubbly-appearing fecal material.

Rectal prolapse. Significant rectal prolapse occurs in 1% to 2% of patients with CF, [130] although it has been seen in up to 18% of patients in some large studies. [131] Onset of rectal prolapse is usually in the first 3 years of life, is often the presenting symptom of CF, and is usually recurrent. [131] A sweat test has been advocated for any child who has had even a single episode of rectal prolapse. [131] Medical management is almost always successful, and adequate replacement of pancreatic enzymes usually results in rapid improvement. However, up to 10% of patients with significant rectal prolapse may require surgical correction.

Liver disorders in cystic fibrosis. Liver abnormalities are frequently observed in patients with CF, but the frequency and severity appear to be decreasing. About 15% of patients have a liver-related problem. However, with modern medical management and emphasis on nutrition, less than 5% develop significant liver injury. [94], [132], [133] The three predominant forms of liver disease in CF include neonatal cholestasis, manifesting with or without meconium ileus or intestinal atresia [134]; fatty liver syndrome; and cirrhosis manifested by portal hypertension or liver failure. Clinical findings include palpable liver (11%), elevated levels of liver enzymes (2.4%), abnormal serum albumin levels (7.4%), cirrhosis with portal hypertension (2.5%), fatty liver (7%), neonatal liver disease (6%), and palpable spleen (2.2%). [130] Asymptomatic hypertransaminasemia or abnormal ultrasonographic findings may be the only manifestations of liver disease, although detection of hepatomegaly or splenomegaly remains a critical component of the physical examination. Esophageal varices or ascites can also be manifestations of hepatic involvement in CF. These may precede evidence of functional impairment (hepatocellular failure) by years. The prevalence of liver abnormalities in CF patients with pancreatic sufficiency is markedly lower. Most patients with mild liver abnormalities do not progress to liver failure, and the abnormal liver injury test results seen in infants may spontaneously resolve. [133] About 10% of patients develop some degree of cirrhosis, and this usually occurs before or during puberty. [133] A familial tendency to develop cirrhosis has been seen [135] and may be associated with one or more modifier genes (see earlier). Other risk factors include neonatal liver disease, pancreatic insufficiency, and possibly human leukocyte antigen class [135], [136] and meconium ileus. [76] Malnutrition and essential fatty acid deficiency may also predispose patients to hepatic steatosis, [133] but nutritional status does not appear to be a major risk factor for liver disease. [137] Histologically, the liver of infants may contain excessive biliary mucus associated with mild periportal inflammation and early fibrosis. Cholestasis from excessive biliary mucus with mild periportal changes may develop and persist for some time. Later in life, focal biliary fibrosis, characterized by inspissated granular eosinophilic material in ductules, bile duct proliferation, chronic inflammatory infiltrates, and variable fibrosis, may develop. Occasionally, the focal lesions coalesce and progress to multilobular biliary cirrhosis. [138] Bile acid metabolism is disturbed in patients with CF and exocrine pancreatic insufficiency. [139], [140] and [141] Fecal losses are high and may approach those of patients with ileal resection. Pancreatic enzyme replacement reduces fecal bile acid excretion and corrects steatorrhea and azotorrhea. The fractional turnover rate of the bile acid pool is increased and the total bile acid pool size diminished in the absence of pancreatic enzymes, [141] whereas the biliary lipid composition and saturation index approach those of patients with cholelithiasis. [140] Treatment with pancreatic supplements returns abnormal values toward normal. Patients with hepatomegaly and abnormal liver test results, with or without abdominal pain, should undergo an assessment of the status of the liver and biliary system. After routine laboratory screening, an ultrasound examination, MRI, or other imaging study may be warranted. Liver biopsy in patients with CF should be undertaken when indicated by the clinical course.

Malnutrition in Cystic Fibrosis Malnutrition occurs in CF through diminished nutrient intake, maldigestion, malabsorption, altered energy utilization, and increased energy expenditure. Intake may be diminished because of abdominal pain, severe gastroesophageal reflux, anorexia, and vomiting or regurgitation and aggravated by depression, physical fatigue, smell and taste aversion, or an altered body image. [142] Maldigestion and malabsorption occur because of pancreatic insufficiency, inadequate bile salt secretion, excess intestinal mucus, infectious enteritis, bacterial overgrowth of the small bowel, and other small bowel pathology. [143] Altered energy utilization is primarily related to diabetes mellitus and liver disease. Increased energy expenditure frequently accompanies severe respiratory disease and is

accentuated by chronic infections, fever, increased respiratory effort, and bronchodilator medications. [142] Severe respiratory symptoms are also associated with anorexia, nausea, and vomiting, which further aggravate the malnutrition cycle. The risk for undernutrition and malnutrition in patients with CF and pancreatic insufficiency is therefore high and potentiates the morbidity and mortality from CF-related problems. [144], [145], [146] and [147]

Diagnosis and Treatment of Cystic Fibrosis

Diagnosis of typical and atypical cystic fibrosis. Once the possibility of CF is considered, the final diagnosis should be confirmed or excluded in a timely fashion and with a high degree of accuracy. This will help prevent unnecessary testing, provide appropriate therapeutic interventions and prognostic and genetic counseling, and ensure access to specialized medical services. On the other hand, the diagnosis should not be taken lightly or made prematurely because it has implications for daily physiologic therapy, medications, special meals, insurance, and employment. Furthermore, a "diagnosis" of CF is difficult to undo. Clinical confirmation of the diagnosis rests on demonstration of elevated concentrations of chloride in sweat (chloride concentration at least 80 mmol/L) [69], [148] or demonstration of an abnormal nasal bioelectric response in specific testing protocols, [69], [149], [150] reflecting abnormal *CFTR* function. When performed correctly, these tests are reliable. However, both false-positive and false-negative sweat test results are seen in newborns, in patients with malnutrition, in the presence of some medications, or if inadequate sweat is obtained. [69] Thus, most experts insist on utilization of standardized methods performed at CF centers that perform the test frequently. A Cystic Fibrosis Foundation consensus panel recently recommended that the diagnosis of CF could be made by the presence of one or more characteristic clinical features, a history of CF in a sibling, or a positive newborn screening test result with confirmation by laboratory evidence of *CFTR* dysfunction. [81] Furthermore, they recommended that either sweat chloride or nasal bioelectrical responses should be abnormal on 2 separate days before the diagnosis is confirmed by one of these methods. [81] Nasal potential difference measurement is difficult to perform, very labor intensive, and available only in a few centers, but it can be especially helpful in cases of atypical CF or borderline features. [69], [101], [151] Genetic testing is also commercially available to confirm the clinical diagnosis (two disease-causing mutations must be identified), but these results cannot always be interpreted apart from the clinical context and functional testing, especially in cases with atypical presentation, because between 2% and 15% of patients with CF may have one or two unidentified mutant alleles.

Treatment. The treatment of CF should be initiated early and should be aggressive, especially in young patients. The effects of delayed diagnosis and inadequate treatment may be long lasting and irreversible in some cases. Therefore, vigilance is warranted.

Pancreatic enzyme replacement therapy. The treatment for pancreatic digestive enzyme deficiency is enzyme replacement. [152], [153] and [154] However, the timing, amount, and other cofactors play important roles in proper delivery of enzyme supplements aimed at restoring digestive function. [155] Enzymes are given with all protein- and fat-containing foods and milk products, including predigested formulas and breast milk. Microspheres, minimicrospheres, or microtables are preferable to granules because the acid-resistant enteric coating protects the enzyme from acid degradation in the stomach and protects against the mouth and perianal excoriation that was previously seen with raw, uncoated enzyme powders. [156], [157] Enzymes should be taken about 15 minutes before each meal or snack. For prolonged meals, additional enzymes should be taken also during the meal. Parents and adults should be taught to adjust the enzyme dosage according to the anticipated amount of fat in a meal. Generic enzymes may not be bioequivalent to proprietary enzymes. [158] Therefore, apparent treatment failure should include investigation of the brand of enzymes that were dispensed. Another cause of failure is destruction of the digestive enzymes by gastric acid. Although enteric coating of pancreatic enzymes may protect pancreatic enzymes in the stomach, the intestines remain more acidic (1 to 2 pH units) in patients with *CFTR* mutations than those with other types of pancreatitis because of loss of duodenal bicarbonate secretion. [115], [116] In the past, treatment of the gastric and intestinal acidity included the use of sodium bicarbonate (baking soda) and histamine-2 receptor antagonists. However, the availability, efficiency, and safety of the proton pump inhibitors have resulted in widespread use of these products. Enteric-coated products remain effective when used with proton pump inhibitors. The dose of enzymes is usually calculated according to lipase content. A usual dose of pancreatic enzymes contains 1000 to 2500 units lipase/kg/meal. Adequacy of treatment is typically determined on clinical grounds. Frequent, bulky, fatty stools, excessive bloating and flatus, excessive appetite, and inadequate growth velocity are signs of inadequate treatment. Calculation of a coefficient of fat absorption is used for clinical studies, but rarely in clinical practice. Although the human fecal elastase-1 test accurately predicts pancreatic insufficiency, [159], [160] and [161] it is of no value in determining the adequacy of enzyme replacement because it is specific for *human* elastase.

Treatment of meconium ileus and distal intestinal obstruction syndromes. Treatment of complicated meconium ileus is surgical. There is a very low operative mortality rate, and long-term survival approaches 100% for uncomplicated meconium ileus. [124], [162] Nonoperative relief of uncomplicated meconium ileus may be possible with diatrizoate (Gastrografin) enemas. [107], [124] A diagnostic barium enema usually precedes therapeutic Gastrografin enemas in children, [91], [124], [163] but a Gastrografin enema can be used directly in adult patients with distal intestinal obstruction syndrome. Infants with CF and meconium ileus who survive beyond 6 months of age have the same prognosis as any patient with CF and do not tend to have more severe disease. [124] The treatment of distal intestinal obstruction syndrome is primarily medical. A stepwise approach with therapeutic trials of more than one modality should be used in each patient before a consideration of surgery. [125] Vigorous medical therapy includes regular oral doses of pancreatic enzymes and stool softeners, oral or rectal administration of 10% *N*-acetylcysteine, and Gastrografin enemas. Maintenance treatment with oral doses of *N*-acetylcysteine, increased doses of pancreatic enzymes, and lactulose has been successfully used to prevent recurrent episodes of the syndrome. Treatment of this disorder with balanced intestinal lavage solutions has also proved helpful. [127], [164] New polyethylene glycol preparations without electrolytes (e.g., MiraLax) also appear to be safe, effective, and well tolerated in adults [165] and children, [166] although its utility in CF has not yet been established.

Treatment of liver complications. The treatment of symptomatic liver disease in CF is a challenge and usually requires a team approach. Treatment of cholestasis is probably best accomplished with ursodeoxycholic acid (20 mg/kg/day), although controlled clinical trials have not confirmed as yet that ursodeoxycholic acid can prevent the progression of liver disease. In patients with cirrhosis, infections (spontaneous bacterial peritonitis and cholangitis) necessitate treatment with appropriate antibiotics. Encephalopathy is extremely uncommon in the cirrhosis of CF but should be treated with protein restriction, lactulose, or neomycin, especially in preparation for liver transplantation. Endoscopic screening should be considered for patients with evidence of cirrhosis to diagnose complications of portal hypertension, including gastropathy and gastric and esophageal varices. Gastrointestinal bleeding should be treated vigorously upon diagnosis. Endoscopic banding or sclerotherapy of bleeding esophageal varices is the most effective and rapid form of therapy. Adrenergic beta-blockers (e.g., propranolol, atenolol) have not yet been widely used in patients with CF liver disease. If severe lung disease is not a contraindication for surgery and the clinical status of the patient is acceptable, end-stage liver disease in CF is an indication for liver transplantation. [167] In rare cases, combined liver-lung transplantation may be indicated, but few such procedures have been reported. [168]

Assessment and treatment of malnutrition. Maintenance of adequate nutrition remains one of the most important challenges in treating patients with CF. The key concepts for nutritional management include proper assessment of nutritional requirements, taking into consideration age, height, weight, and anthropometrics and severity of lung disease as well as anorexia, pancreatic insufficiency, other intralumenal phase abnormalities, and mucosal dysfunction. [142], [169], [170] The physician must therefore determine whether the patient has inadequate nutritional intake and the degree of maldigestion, malabsorption, altered energy requirements, or other medical complications such as diabetes mellitus or liver disease. If the nutritional targets of maintaining or gaining weight are missed, or biochemical evidence of malnutrition emerges, then a detailed evaluation is warranted. Nutritional targets begin with determination of the patient's ideal weight compared with his or her actual weight. Further adjustments are made for age, growth patterns, specific nutritional deficiencies, and other factors. For adult patients, the body mass index (BMI) is used to assess appropriate weight, calculated as weight in kilograms divided by height in square meters. [156] In pediatric patients, the BMI percentile should be used [156] (see appropriate charts on the Center for Disease Control website at http://www.cdc.gov/growthcharts). The older method was to calculate the percentage ideal body weight (IBW), calculated as the actual weight divided by ideal weight-for-height multiplied by 100. [170], [171] The percentage IBW should also be recorded if this method was used before development or use of the BMI percentile charts. [156] /SUP>If the patient is losing weight or a child appears to develop a weight plateau but maintains weight above 90% of IBW, [156] or if the BMI is between the 10th and 25th BMI percentile, he or she is at risk for nutritional failure. Nutritional failure occurs when weight plateaus for more than 3 months in children younger than 5 years of age or for more than 6 months in children older than 5 years of age, or when the percentage IBW is less than 90% or the BMI percentile is less than 10th percentile. [156] Inquiry about reproductive health, including menstrual regularity in females, may also provide clues as to nutritional health. The optimal dietary intake for a patient with CF is greater than the recommended daily allowance of healthy children and adults. Ideally, a normal diet for age should be encouraged, except that fat intake should achieve 35% to 40% of calories, balanced with adequate pancreatic replacement therapy. [106], [156] However, a daily intake between 110% and 149% of the standard Recommended Daily Allowance (RDA) for age is needed for calories and protein to promote normal growth, and children with end-stage lung disease may require in excess of 150% of the RDA. [156], [172] Nutritional intervention begins with addition of high-calorie foods to the usual diet and use of nutritional supplements. [156] When this fails, enteral feedings should be started. This may be achieved through nasogastric tubes, gastrostomy, or jejunostomy tubes. The presence and severity of gastroesophageal reflux disease symptoms may influence the decision on route. Standard formulas are usually well tolerated. Nocturnal infusion is encouraged to promote normal eating patterns during the day. Initially, 30% to 50% of the estimated caloric needs should be provided overnight. Very-low-fat, elementary formulas may be used without enzyme supplements for patients with enteral feeding tube and should be given by continuous infusion. [156] Pancreatic enzyme supplements taken orally in the usual premeal dose are recommended before all nocturnal enteral feedings. Patients with enteral feedings should be monitored for carbohydrate intolerance by measuring blood glucose levels 2 to 3 hours into the feeding and at the end of the feeding on at least 2 separate nights. Insulin may be required to prevent hyperglycemia, and the dosage may need to be adjusted during illness, steroid use, or other changes in health status. All patients with CF should receive a multivitamin preparation daily, and many require vitamin A, E, K, and D supplements. [170]

Clinical practice guidelines for cystic fibrosis management. Advances in medical treatment and nutrition have markedly improved the prognosis of patients with CF, with more than half of all patients living beyond 30 years of age. [130], [173] The treatment necessary to surpass this milestone requires attention to a wide range of issues involving multiple organ systems, nutritional challenges, developmental problems, social issues, and other considerations. These are usually best met by well-coordinated multidisciplinary teams. Care guidelines have been defined and refined within the numerous CF centers, which typically focus on infant and pediatric patients. In many cases, the pediatric CF specialists continue to care for adult patients with CF. However, there is growing

consensus among experts that these patients should transition to physicians specializing in adult patient care sometime between the age of 18 and 21 years, depending on the medical condition, the emotional maturity of the patient, and the readiness of the family. [174] Adult patients with CF suffer from the same matrix of problems recognized in the pediatric population, plus additional problems. These medical problems include pancreatitis, adequate nutrition, cirrhosis with portal hypertension, diabetes with its long-term complications, osteopenia, and reproductive issues. [97], [175], [176] and [177] Gallbladder disease, peptic ulcer disease, pancreatitis, and cirrhosis with portal hypertension are more common in adults than in children. [130], [178] Furthermore, pulmonary disease is more severe in adults than in children, and malnutrition remains a prominent problem in about 35% of adults with CF. [130] Increasingly, these patients will require evaluation for potential malignancies of the digestive tract and evaluation for liver disease or other complications that will necessitate the specialized attention of a gastroenterologist. The complexity and severity of these problems requires the coordinated care of a multidisciplinary team that is typically headed by pulmonologists and includes gastroenterologists, endocrinologists, urologists, and the allied health care of nurses, dietitians, respiratory and physical therapists, and social workers. [174] The adult patient should be seen at least four times a year, with annual investigation of nutritional status (including vitamin levels), liver function, and other disease-related screening tests (e.g., oral glucose tolerance test hemoglobin A1c). Routine clinical visits should include qualitative dietary history; assessment of appetite, eating, and related behavioral patterns; and supplemental feeding practice since the last visit, including enteral and parenteral nutrition. [174] The nutritional assessment may be expedited with assistance of a qualified registered dietitian or others with skills in obtaining a 3-day quantitative dietary record and assessment of energy balance. Measurement of mid-arm circumference and triceps skin-fold thickness provides clinical information on both lean body mass and subcutaneous fat stores. Standardized techniques and equipment should be used for body measurements, and special training is recommended to ensure accuracy and consistency. [156] The pulmonary problems are beyond the scope of this chapter and require the attention of a pulmonologist with experience in CF. Also, CFRD should be managed by the endocrinologists. If the CF-related training and experience of an adult medical specialists is limited, then a CF Care Center Director or other pediatrician with more extensive experience with CF should remain on the multidisciplinary team, or referrals should be made to centers with more experience in adult CF medicine. [174]

RARE SYNDROMES WITH MAJOR PANCREATIC MANIFESTATIONS

Shwachman-Diamond Syndrome

Shwachman-Diamond syndrome (SDS, OMIM *260400) remains the second most frequently recognized cause of pancreatic insufficiency in children, representing about 3% of causes of pancreatic dysfunction. [86] SDS is an autosomal recessive disorder [179] with a disease gene at the centromere of chromosome 7. [180], [181] Rather than being a pancreas-destroying disease like CF and hereditary pancreatitis, the pancreas in patients with SDS appears to have fatty replacement of acinar cells. [182], [183] The parotid gland amylase secretion may also be defective, suggesting that the acinar cell dysfunction is generalized. [184]

SDS is characterized by exocrine pancreatic insufficiency with normal sweat electrolytes and significant hematologic abnormalities. [185], [186] and [187] Skeletal defects, including short ribs with broadened anterior ends and metaphyseal dyschondroplasia on the femoral head, short stature, myelodysplastic syndromes, and acute leukemias, are common. [185], [186] Other abnormalities include elevated liver enzymes, hepatomegaly, and developmental delay.

The pancreatic insufficiency associate with SDS is quite variable and differs from CF because it improves with time in up to 50% of patients. [86], [185], [186], [188] The lesion appears to be a developmental failure of the acinar cells because enzyme secretion is drastically reduced, whereas duct cell function is normal. [86] Pancreatic insufficiency can be diagnosed with fecal fat measurements, pancreatic function testing for lipase levels, low immunoreactive serum trypsinogen levels, or imaging studies such as MRI or CT that reveal lipomatous pancreas. The treatment of pancreatic insufficiency is supportive, including pancreatic enzyme replacement.

Cyclic neutropenia is the most common hematologic disorder, but other cellularity can be involved. [185], [186], [188] The cyclic neutropenia leads to recurrent infections, especially respiratory problems. The human granulocyte colony-stimulating factor has been successfully used in patients with SDS with severe neutropenia and frequent infections. [186], [189] This seems to control the neutropenia, even when weekly doses are given. The dose should be the minimum necessary to achieve control of infection. [186] The problem with this treatment is that there are no clear guidelines for the duration of use or for whether it is advisable in the presence of cytogenetic alterations. [186]

Another major problem in SDS patients is myelodysplastic syndrome and eventually acute myeloid leukemia. [185], [186], [190], [191] Although the prevalence has not been established, acute myeloid leukemia is reported to occur in 0% to 30% of patients. [179], [185], [186], [191] Some specific abnormalities are common, especially isochromosome 7q10. [180] Allogenic bone marrow transplantation should be considered for patients who develop leukemia. [186], [192], [193] and [194]

Johanson-Blizzard Syndrome

Johanson-Blizzard syndrome (OMIM *243800) is a very rare autosomal recessive syndrome of unknown etiology. Johanson-Blizzard syndrome [195] is also known as nasal alar hypoplasia, hypothyroidism, pancreatic achylia, and congenital deafness syndrome. [9] It is characterized by pancreatic insufficiency and growth retardation with lipomatous transformation of the pancreas. Histologically, the pancreatic ducts and islets are preserved but are surrounded by connective tissue and a total absence of acini. Pancreatic ductal function remains normal. [196] Additional features include thyroid dysfunction, cardiac anomalies, genitourinary malformations, midline ectodermal scalp defects, dental anomalies, and imperforate or anterior anus. [195], [197] On the other hand, there may be milder forms presenting with isolated pancreatic enzyme deficiencies. [197] The treatment of the exocrine pancreas deficiencies is similar to other forms of pancreatic insufficiency.

Pearson Marrow-Pancreas Syndrome

Pearson marrow-pancreas syndrome (OMIM #557000) is a rare, autosomal dominant mitochondrial DNA (mtDNA) breakage syndrome. [198], [199] Clinical features include pancreatic insufficiency, sideroblastic anemia, and diabetes mellitus. The pancreatic insufficiency appears to be due to pancreatic fibrosis. [200] The molecular defect in Pearson marrow-pancreas syndrome was initially identified as a 4977–base pair deletion of mtDNA, although a variety of mtDNA defects have been discovered. [201], [202] and [203] The clinical features and severity of disease appear to correlate with the proportion of abnormal mtDNA, and multiple organ systems may be involved.

RARE DEVELOPMENTAL ABNORMALITIES

A variety of congenital abnormalities of the pancreas have been observed, reflecting defective embryogenesis. Pancreatic divisum, annular pancreas, common channel syndrome, choledochal cysts, heterotopic pancreatic tissue, and annular pancreas are included in this group, but the genetics is not well understood.

Pancreatic agenesis is extremely rare and is probably caused by a homozygous nucleotide deletion in the *PDX1* gene, resulting in premature termination of the *PDX1* gene production. [204] The clinical features include intrauterine growth retardation, insulin-dependent diabetes, and pancreatic exocrine insufficiency. Serum C-peptide and glucagon are undetectable, and the pancreas is absent on imaging studies. Survival into childhood is possible with proper diagnosis and insulin and enzymatic supplementation. [204], [205] Pancreatic hypoplasia is thought to be a variant of pancreatic agenesis, and isolated agenesis of the dorsal or ventral pancreas has also been observed. Agenesis of the dorsal pancreas [206] and ventral pancreas [207] has occasionally been seen, but the cause is unknown.

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CHAPTER 99

Diane M. Simeone

GALLBLADDER AND BILIARY TRACT: ANATOMY AND STRUCTURAL ANOMALIES

The extrahepatic biliary tract has a great number of anomalies and variations, with a wide range of anatomic variations also seen in the vascular structures surrounding the extrahepatic biliary tree. Increasing use of diagnostic and interventional radiology as well as therapeutic endoscopy has made it essential for a wide variety of physicians to become familiar with the anatomy of the extrahepatic biliary tract. This review of anatomy and anomalies of the extrahepatic bilary tree will assist surgeons, gastroenterologists, and radiologists in understanding and recognizing the anomalies they may encounter.

EMBRYOLOGICAL DEVELOPMENT

The biliary tract develops from the primitive gastrointestinal tract, in the distal foregut, as a ventral sacculation; it is first apparent at week 5 of gestation, or when the embryo is 3-mm long (Fig. 99-1A). This sacculation grows and extends into the ventral mesentery, dividing into two buds: the cranial bud develops into the liver and intrahepatic bile ducts and the caudal bud develops into the gallbladder and cystic duct (Fig. 99-1B). The base of the diverticulum ultimately becomes the common bile duct. Another small bud develops from the proximal aspect of the caudal bud and grows inferiorly, ultimately developing into the ventral pancreas (see Fig. 99-1A, Fig. 99-1B).

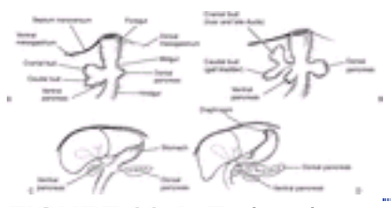

FIGURE 99-1. Embryology of the developing biliary tract. **A:** At the 3-mm stage of the embryo, the ventral bud enters the mesogastrium and soon divides into a cranial and a caudal bud. A smaller caudad bud represents the origin of the ventral pancreas. **B:** As the embryo reaches 5 mm, the cranial bud, which will form the liver and intrahepatic biliary tract, moves toward the septum transversum, pulling the caudal bud, which will form the gallbladder and extrahepatic bile ducts. **C:** When the embryo reaches 7 mm, the right and left lobe of the liver occupy the position under the septum transversum. The ventral pancreas and the extrahepatic biliary tract are visible. As the ventral pancreas rotates to reach the dorsal pancreas, it pulls the lower end of the common bile duct with it. **D:** At the 12-mm embryonic stage, the ventral pancreas has rotated, and the anatomic relations between the bile ducts and the gastrointestinal tract have assumed their mature forms.

The cranial bud divides into two smaller buds that grow upward toward the septum transversum (future diaphragm). These eventually become the right and left lobes of the liver. The caudal bud is carried superiorly by the growth of the cranial bud and stops at the undersurface of the cranial bud to become the gallbladder and cystic duct (Fig. 99-1C). Rarely, continued advancement of the caudal bud too far superiorly will result in an intrahepatic gallbladder, which is almost always embedded within the right lobe of the liver.

As the cranial and caudal buds are advancing, the ventral pancreatic bud rotates 180 degrees from right to left, allowing it to fuse with the dorsal pancreatic bud to form the complete pancreas. Fusion of the two parts of the pancreas occurs during week 7 of gestation. Because the lower end of the common bile duct is attached to the ventral pancreatic bud, the rotation results in fusion of the junction of the common bile duct and the duodenum on the posteromedial duodenal wall, posterior to the dorsal pancreatic duct (Fig. 99-1D).

The ventral sacculation from which the biliary tract arises is originally composed of a solid cord of endodermal cells and contains no lumen. Beginning at week 7 of gestation, vacuolization begins to occur and a completely open lumen is formed in the gallbladder, cystic duct, hepatic ducts, and common bile duct within 1 week. By the third month of gestation, bile flow is demonstrable through the canalized biliary tract into the duodenum. [1]

ANATOMY OF THE GALLBLADDER

Gross Anatomy

The gallbladder is a pear-shaped sac that is located along the inferior surface of the liver in the line of division of the anatomic left and right lobes; it lies in a depression known as the gallbladder fossa (Fig. 99-2). The adult gland is typically 7 to 10 cm in length and has an average capacity of 30 mL. The gallbladder is intimately attached to the liver by loose connective tissue, which contains small veins and lymphatics that connect between the gallbladder and liver. Occasionally, one or more small accessory ducts from the liver traverse this connective tissue to enter the gallbladder directly. Inadequate ligation of an accessory duct may account for the occasional bile leak that occurs after cholecystectomy.

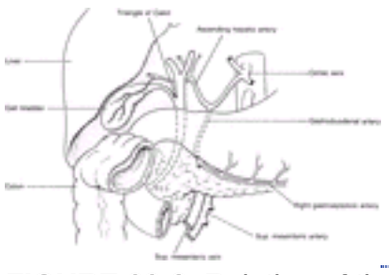

FIGURE 99-2. Relation of the gallbladder and the extrahepatic biliary tract to the liver, duodenum, colon, and pancreas.

The remainder of the gallbladder not in direct contact with liver is covered by peritoneum reflected from the liver. The gallbladder is divided into four anatomic areas: fundus, body, infundibulum, and neck. The fundus is the rounded end of the gallbladder that normally extends 0.5 to 1 cm beyond the liver margin and is covered with peritoneum. The fundus rests on the duodenum and hepatic flexure of the colon (see Fig. 99-2) and is in direct contact with the anterior abdominal wall, usually near the lateral border of the right rectus abdominus muscle at approximately the level of the ninth costal cartilage; however, its position may vary. The fundus may occasionally possess a kink, known as a phrygian cap deformity, which has no pathological significance.

The fundus merges imperceptibly into the body, the largest part of the gallbladder, and the segment that is closely attached to the liver on one side. This intimate contact is responsible for early direct spread of gallbladder carcinoma into the liver. The infundibulum represents a transitional region between the body and neck. It is notable because occasionally a shallow diverticulum may be present on the inferior surface of the infundibulum, referred to as a Hartmann pouch. Gallstones may become impacted within the Hartmann pouch, causing cystic duct obstruction and thus acute cholecystitis.

The neck is a short (usually 5–7 mm) region that tapers into the cystic duct. The neck occupies the deepest part of the gallbladder fossa and lies in the free border of the hepatoduodenal ligament.

Arterial Blood Supply

The blood supply to the gallbladder is usually from a single cystic artery that arises from the right hepatic artery, traverses the triangle of Calot, and divides into two branches close to the gallbladder wall [2] (Fig. 99-3A). One branch runs along the peritoneal surface of the gallbladder and the other branch in the gallbladder fossa between the gallbladder and liver. When the cystic artery arises from the right hepatic artery, its course is often parallel and medial to the cystic duct. Double or accessory cystic arteries have been reported in up to 20% of people. Variations in the site of origin and course of the cystic artery are common. The cystic artery typically arises from the right hepatic artery (95%), but also may arise from the left hepatic artery, the common hepatic artery, an aberrant right hepatic artery that arises from the superior mesenteric artery, the gastroduodenal artery, the superior mesenteric artery, or celiac axis directly (see Fig. 99-3). The cystic artery may be short or long, and may pass either behind the hepatic duct (84%) or may cross the hepatic duct anteriorly.

FIGURE 99-3. Common variations in the origin of the cystic artery. **A:** It originates most commonly from the right hepatic artery; it traverses the triangle of Calot, and, upon reaching the gallbladder, it divides into two main branches. **B:** Occasionally the two branches come off the right hepatic artery independently. The cystic artery may cross the hepatic duct anteriorly (**C**), come off the left hepatic artery (**D**), or, more rarely, come directly from the celiac axis (**E**). (From Lindner H. Embryology and anatomy of the biliary tree. In: Way LW, Pellegrini CA, eds. Surgery of the gallbladder and bile ducts. Philadelphia: WB Saunders, 1987.)

The cystic artery, when it originates from the right hepatic artery, may closely parallel the right hepatic artery for a distance before reaching the gallbladder. In 5% to 10% of cases, the cystic artery does not arise from the right hepatic artery until just before it enters the right lobe of the liver, thus creating a very short (only a few millimeters) cystic artery. Failure to recognize this may lead to inadvertent ligation of the right hepatic artery during cholecystectomy.

Venous and Lymphatic Drainage

The gallbladder lacks a major cystic vein but rather drains through a network of small veins. These veins run either from the hepatic surface of the gallbladder directly to the liver or run toward the cystic duct and join venous collaterals from the common bile duct before ultimately draining into the portal vein.

The lymphatic drainage of the gallbladder parallels the venous drainage pattern. Lymph flows directly from the gallbladder to the liver or may drain toward the cystic duct into a single node or group of nodes. From these lymph nodes, lymph may ultimately drain into several nodes along the surface of the portal vein and common bile duct.

Nerve Supply

The gallbladder is innervated by branches of both the sympathetic and parasympathetic nervous systems, which pass through the celiac plexus. Preganglionic sympathetic nerves arise from the T8 and T9 levels. Postganglionic sympathetic nerves originate at the celiac plexus and travel along the hepatic artery and portal vein to the gallbladder. Parasympathetic nerves arise from branches of the vagal trunks. Unlike the branches of the posterior vagal trunk that pass through the celiac plexus, branches of the anterior vagal trunk reach the gallbladder by way of the gastrohepatic ligament (Fig. 99-4). Visceral pain caused by gallbladder wall distention or inflammation is conducted through afferent sympathetic fibers and is referred to the epigastric, right subcostal, or right scapular regions. The nerve supply to the common bile duct is the same as for the gallbladder.

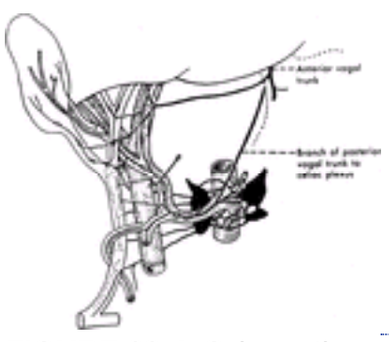

FIGURE 99-4. Schematic representation of the innervation of the gallbladder and extrahepatic biliary tract. The nerves originate from both vagi and from the celiac axis. They reach the biliary tract, traveling along the walls of the hepatic artery, except for the direct vagal branches of the anterior vagus, which cross through the gastrohepatic ligament.

Although gallbladder contractility is primarily mediated by cholecystokinin (CCK), innervation of the gallbladder also modulates gallbladder contractility. Vagal activity contributes to normal gallbladder tone and enhances the ability of subthreshold doses of CCK to promote gallbladder mobility. [3] Sympathetic fibers enhance gallbladder relaxation. In addition to the classic parasympathetic and sympathetic nervous systems, a large number of peptide-containing neurons exist within the gallbladder. The peptides identified thus far by immunocytochemical techniques include CCK, [4] vasoactive intestinal polypeptide, [4] gastrin-releasing peptide, [5] neuropeptide Y, [6] pituitary adenylate cyclase–activating polypeptide (PACAP), [7] calcitonin gene–related peptide (CGRP), [7] and substance P. [7] The physiologcial functions of these peptidergic nerves within the gallbladder remain to be elucidated.

Histology and Ultrastructure

The gallbladder has five layers: epithelium, lamina propria, muscularis, perimuscular connective tissue layer, and serosa. The mucosa, lined with a simple columnar epithelium, consists of many folds that increase the efficiency of the gallbladder in concentrating bile. The apical surface of the epithelium is covered with abundant microvilli. The basal surface of the epithelium is in contact with the lamina propria, which is rich in loose connective tissue, elastic fibers, blood vessels, and lymphatics. The muscularis is an arrangement of circular, longitudinal, and oblique fibers without any well developed layers. Ganglia may be found between the muscle fibers of the muscularis. The subserosa contains loosely arranged collagen and elastic fibers, as well as larger blood vessels and lymphatics. This thick subserosal layer attaches the gallbladder to the liver. Dissection during cholecystectomy usually takes place between the subserosa and serosa, which in normal circumstances is an avascular plane. The serosa is absent on the surface of the gallbladder that is in direct contact with the liver. Mucus is secreted into the gallbladder from tubular alveolar glands found in the mucosa lining the infundibulum and neck of the gallbladder.

At the ultrastructural level, the gallbladder epithelium is well suited for its physiological role of concentrating bile. The columnar epithelial cells have basally located nuclei and apices covered with numerous microvilli that protrude into the gallbladder lumen. Numerous mitochondria are present within the epithelial cells, which facilitate the active process of bile concentration. Epithelial cells are tightly joined together along their lateral membranes by tight junctions, which separate the gallbladder lumen from the intercellular spaces. During active bile concentration, the intercellular spaces become distended with water and electrolytes. Water extracted during this process passes into a rich capillary network in the lamina propria.

ANATOMY OF THE EXTRAHEPATIC BILIARY DUCTS

Hepatic Ducts

The common hepatic duct is formed by the union of the right and left hepatic ducts. In 95% of cases, this union takes place outside the liver, just below the level of the porta hepatis. In 5% of cases, the left and right hepatic ducts join within the substance of the liver. The lengths of the right and left hepatic ducts vary from 0.5 to 2.5 cm in length, with the left hepatic duct usually longer than the right. The ducts may join at a wide or acute angle, depending on their extrahepatic length; however, they usually join about 1 cm below the porta hepatis to form the common hepatic duct. The confluence of the right and left hepatic ducts lies anterior to the portal venous bifurcation, usually overlying the origin of the right branch of the portal vein. An accessory right hepatic duct is present in 10% of people. The common hepatic duct passes downward in the superior portion of the hepatoduodenal ligament and lies anterior to the portal vein and to the right of the hepatic artery (Fig. 99-5). In cases when the cystic duct inserts on the right hepatic duct or at the junction of the union of the right and left hepatic ducts, there is no common hepatic duct. The length of the common hepatic duct varies from 2 to 6.5 cm. It joins the cystic duct to form the common bile duct.

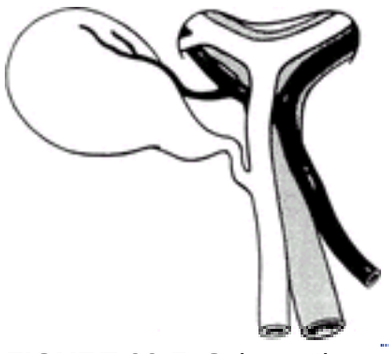

FIGURE 99-5. Schematic representation of the hilum of the liver. The right anterior and posterior hepatic ducts join to form the right hepatic duct. This duct and the left duct join outside the liver capsule to form the common hepatic duct. The left duct is usually longer and more superficial than the right. The triangle of Calot, with the right hepatic artery and the cystic artery, is also clearly displayed. (From Lindner H. Embryology and anatomy of the biliary tree. In: Way LW, Pellegrini CA, eds. Surgery of the gallbladder and bile ducts. Philadelphia: WB Saunders, 1987.)

The course of the right hepatic artery is variable in relation with the right hepatic duct and common hepatic duct. Usually the right hepatic artery passes posterior to the common hepatic duct near the junction of the right and left hepatic ducts (64%). In 24% of people, the right hepatic artery or the cystic artery may pass anterior to the common hepatic duct. In the remaining patients (12%), the right hepatic artery arises from the superior mesenteric artery and runs parallel and to the right of the common bile duct and posterior to the cystic duct. Because of the inconstant location of the right hepatic artery and its close proximity to the cystic artery, the right hepatic artery may be mistaken for the cystic artery and is particularly vulnerable to injury during biliary tract surgery.

The triangle of Calot, originally described in 1891, is bounded superiorly by the hilum of the liver, on the right by the cystic duct, and on the left by the common hepatic duct [8] (see Fig. 99-5). Dissection of the triangle of Calot during cholecystectomy must be carried out with care because many important structures run through this triangle: the right hepatic artery, the cystic artery, 90% of accessory hepatic ducts, and 95% of aberrant right hepatic arteries. In cases when there is a replaced right hepatic artery originating from the superior mesenteric artery, the vessel travels toward the liver in a position posterolateral to the common bile duct and just behind the cystic duct. Here it is vulnerable to injury. Complete dissection of the triangle of Calot, with separation of the base of the gallbladder from the liver bed, allowing identification of the only two structures that should be emanating from the gallbladder, the cystic duct and cystic artery, is critical to avoid injury during cholecystectomy.

Cystic Duct

The cystic duct arises from the neck of the gallbladder and joins the common hepatic duct. Its lumen usually measures 1–3 mm, but it may occasionally be as large as 10 mm, allowing larger gallstones to enter the common bile duct. While the cystic duct typically joins the common hepatic duct directly (70%), the site of the cystic duct junction with the extrahepatic biliary tree may vary from the right hepatic duct down to the level of the ampulla (Fig. 99-6 and Fig. 99-7). Thus the length of the cystic duct may vary from 0.5 to 8 cm. The cystic duct may join the right hepatic duct, may run parallel to the common hepatic duct for a distance with connective tissue ensheathing both ducts, and, in some instances, may not enter the common hepatic duct until passing behind the duodenum. Additionally, the cystic duct may spiral around the common hepatic duct, passing either dorsal or ventral to the common hepatic duct before joining it. These anatomic variations may lead to bile duct injury during cholecystectomy, especially if dissection is carried out to clearly define the union of the cystic duct and common bile duct, a practice that is unnecessary during cholecystectomy.

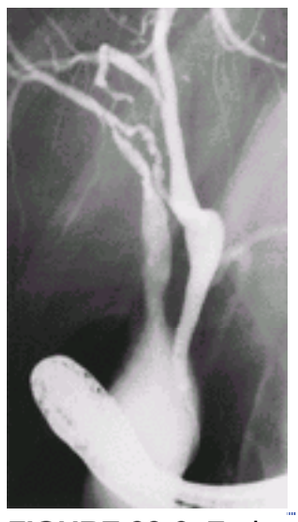

FIGURE 99-6. Endoscopic retrograde cholangiopancreatogram demonstrating an anomalous junction of the cystic duct with an accessory right hepatic duct.

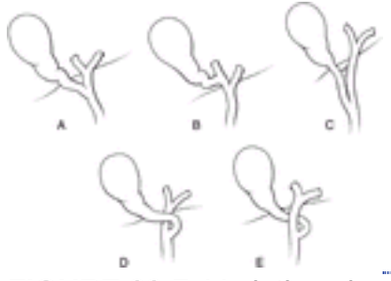

FIGURE 99-7. Variations in cystic duct anatomy. **A:** Cystic duct joins common hepatic duct directly (most common). **B:** Cystic duct joins the right hepatic duct. **C:** Low junction of cystic duct with common hepatic duct. **D:** Anterior spiral of cystic duct before joining common hepatic duct. **E:** Posterior spiral of cystic duct before joining common hepatic duct.

Within the cystic duct, the mucosa forms a series of 5 to 10 crescent-shaped folds, known as the spiral valves of Heister. They span the length of the cystic duct and project into the cystic duct lumen. These valves serve to prevent excessive distention or collapse of the gallbladder with changes in cystic duct pressure and may function to block passage of gallstones into the common bile duct. They may make catheterization during intraoperative cholangiogram difficult.

Common Bile Duct

Union of the cystic duct and common hepatic duct forms the common bile duct. Its length is approximately 7.5 cm, but can vary depending on the relative lengths of the cystic and common hepatic ducts. The mean diameter of the common bile duct is about 6 mm, but it may dilate significantly in the face of distal obstruction. The common bile duct is divided into four anatomic segments: supraduodenal, retroduodenal, pancreatic, and intraduodenal. The supraduodenal segment is usually 2.5 cm in length and is located in the right border of the hepatoduodenal ligament. It lies anterior to the portal vein and to the right of the ascending common hepatic artery. This anatomic relationship allows the surgeon to compress these three structures between an index finger inserted through the foramen of Winslow into the lesser sac and a thumb placed anteriorly across the hepatoduodenal ligament. This maneuver is referred to as the Pringle maneuver and allows rapid occlusion of the blood supply to the liver, which is useful for controlling major liver hemorrhage.

The retroduodenal segment lies posterior to the first part of the duodenum and is about 2.5 to 4 cm long. As it runs to the inferior surface of the duodenum, it moves right to left, lying just to the right of the gastroduodenal artery. The retroduodenal common bile duct is loosely attached to the duodenum by a thin layer of areolar tissue. Because of the close relationship of this portion of the common bile duct and the duodenal bulb, it may occasionally be involved in the inflammatory process of a posterior penetrating duodenal ulcer and may be inadvertently injured during antrectomy.

The pancreatic segment of the common bile duct extends from the lower border of the first part of the duodenum to the posteromedial wall of the second portion of the duodenum where the duct penetrates the duodenal wall. It may be entirely retropancreatic, or it may lie within the substance of the head of the pancreas. Obstruction of this segment of the common bile duct is common with cancers of the pancreatic head, which often initially present with painless jaundice.

The common bile duct then turns 90 degrees to the right to enter the posteromedial wall of the descending duodenum. The final, or intraduodenal, segment of the common bile duct is about 2 cm long and travels obliquely through the duodenal wall with the main pancreatic duct. The common bile duct may join the main pancreatic duct outside the duodenum, or the ducts may form a common channel as they traverse through the duodenal wall, in both of these cases opening through a single ostium on the major ampulla of Vater. This site is typically 7 to 10 cm from the pylorus. From the inside of the duodenal lumen, the termination of the ampulla appears as a small, protruding, nipple-like structure marked by a longitudinal duodenal fold. In 29% of cases, a septum persists between the two ducts, and the ducts empty on the papilla as separate ostia.

Ampulla of Vater

Union of the common bile duct and the main pancreatic duct forms the ampulla of Vater. The length of the ampulla is variable, and if there is no junction of the common bile duct and main pancreatic duct, there is no true ampulla of Vater. Rienhoff and Pickrell [9] studied the pancreatic duct system in 250 autopsy specimens. They found an ampulla longer than 2 mm in 46% of cases (range from 3 to 14 mm), an ampulla less than 2 mm in 32% of cases, and no junction of the pancreatic and bile ducts in 29% of cases.

The common bile duct narrows significantly as it passes through the wall of the duodenum, and the ampulla narrows before it enters the duodenal lumen. These narrowings are frequent sites for stones to lodge and cause either biliary or pancreatic obstruction. Additionally, these are potential sites of injury when instrumented during common bile duct exploration.

Sphincter of Oddi

The intraduodenal segment of the common bile duct and the ampulla is surrounded by a sheath of smooth muscle fibers referred to collectively as the sphincter of Oddi. The sphincter of Oddi is a unique group of muscle fibers that arise from the bile duct wall and manometric studies have verified that the sphincter acts independently of the duodenal musculature. The resting pressure of the sphincter of Oddi is approximately 13 mm Hg above duodenal pressure. [10] Regulation of bile flow is primarily controlled by the sphincter of Oddi. Relaxation of the sphincter occurs with CCK stimulation and is facilitated by parasympathetic stimulation. Sympathetic stimulation causes increased sphincter tone.

The preampullary portion of the common bile duct is invested in a sheath of ircular muscle referred to as the sphincter choledochus (sphincter of Boyden) (Fig. 99-8). The distal main pancreatic duct may have a short sphincter called the sphincter pancreaticus. If present, the sphincter pancreaticus and sphincter choledochus may intertwine in a figure-of-eight manner. The smooth muscle sheath surrounding the ampulla is called the sphincter of the ampulla; if there is no ampulla, the distal sphincter is simply called the sphincter of the papilla. During endoscopic sphincterotomy, the sphincter of Oddi is divided using electrocautery to relieve common bile duct obstruction from a common duct stone.

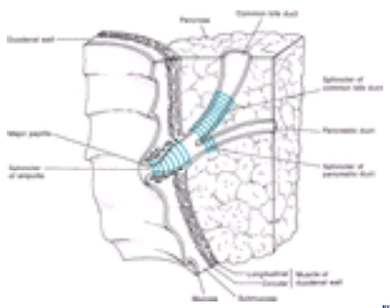

FIGURE 99-8. The muscular apparatus at the terminal end of the common bile duct. The bile duct is closely associated with the pancreatic duct, and they both enter the medial wall of the duodenum tangentially. Each duct has its own sphincter, which is poorly developed in the pancreatic duct.

Arterial Blood Supply

The arterial blood supply to the extrahepatic bile ducts is segmental. Because of the segmental nature of the blood supply, extensive mobilization of the extrahepatic bile ducts may lead to ischemic injury and development of postoperative biliary stricture and thus should be avoided. [11] The hepatic ducts and the supraduodenal portion of the common bile duct are nourished by small arterial branches from the cystic artery (Fig. 99-9). The retroduodenal portion of the common bile duct is supplied by branches of the retroduodenal and posterior superior pancreaticoduodenal arteries The pancreatic and intraduodenal segments of the common bile duct are supplied by both the anterior and posterior superior pancreaticoduodenal arteries.

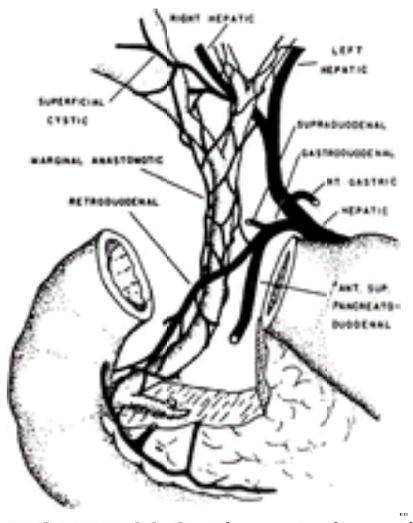

FIGURE 99-9. The extrahepatic biliary tract is supplied by a rich vascular net of vessels from the hepatic and gastroduodenal arteries. The relationship between the hepatic arteries and the extrahepatic biliary tree is evident.

Venous and Lymphatic Drainage

The veins of the hepatic ducts and proximal common bile duct, like those from the gallbladder and cystic duct, enter the liver directly. Veins from the lower portion of the common bile duct drain into the portal vein. The lymphatic drainage from the hepatic ducts and upper common bile duct flows superiorly along the course of the common bile duct to lymph nodes in the porta hepatis. Some lymphatic drainage arising from the inferior portion of the common bile duct may reach the deep pancreatic nodes near the origin of the superior mesenteric artery. All lymphatic drainage ultimately reaches the celiac lymph nodes.

Histology and Ultrastructure

The mucosa of the extrahepatic bile ducts contains columnar epithelium surrounded by a layer of connective tissue. The epithelium contains many mucous glands. Muscle fibers in the hepatic ducts and proximal common bile duct are relatively few and discontinuous, and may be arranged in either a longitudinal or circular direction. As the common bile duct approaches the duodenum, it begins to develop a more substantial muscle layer, which merges into the sphincter of Oddi complex, where distinct bundles of muscle fibers are evident.

CONGENITAL VARIATIONS AND MALFORMATIONS

Gallbladder and Cystic Duct

Congenital anomalies of the gallbladder may be classified into three different categories: anomalies of number, anomalies of form, and anomalies of position. Duplication of the gallbladder is thought to occur in approximately one of every 4000 human gallbladders. [12] With complete duplication, the gallbladders are separate, and the cystic ducts may join one another before entering the common duct or each cystic duct may enter separately. Partial duplications can result in bilobed gallbladders (Fig. 99-10A), in which the two gallbladder cavities are separate at the fundus but joined at the neck and drain via a common cystic duct. Gallbladders may also be septate (divided by a partial or complete septum) with septa present in either a longitudinal or transverse direction, the latter referred to as an hourglass gallbladder (Fig. 99-10B). Septate gallbladders are thought to be a result of incomplete vacuolization of the solid endodermal cord during development. None of these abnormalities of the gallbladder by themselves have clinical significance unless they are involved in a pathological process.

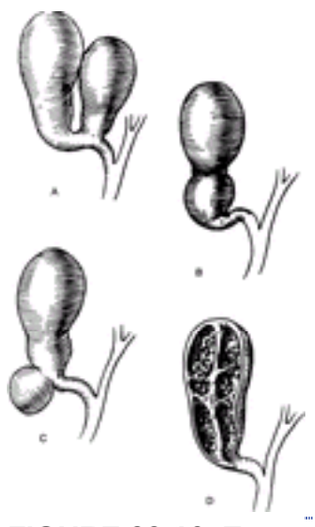

FIGURE 99-10. Two gallbladders (**A**), a bilobed gallbladder (**B**), a diverticulum at the neck (**C**), and a septated gallbladder (**D**) are all anatomic variations that relate to the embryological development of the biliary tract. (From Lindner H. Embryology and anatomy of the biliary tree. In: Way LW, Pellegrini CA, eds. Surgery of the gallbladder and bile ducts. Philadelphia: WB Saunders, 1987.)

Lack of development of the gallbladder bud results in agenesis of the gallbladder. Most cases are associated with extrahepatic biliary atresia. [13] In the absence of associated biliary atresia, an intrahepatic gallbladder or left-sided gallbladder must be ruled out before the diagnosis is made.

Abnormal migration of the caudal bud (future gallbladder and cystic duct) of the ventral diverticulum during development may cause an anomalous gallbladder position. If the caudal bud advances beyond the cranial bud (future liver), it may become buried in the liver substance, creating an intrahepatic gallbladder. These are usually identified by ultrasonography prior to cholecystectomy and may make cholecystectomy slightly more difficult. If the caudal bud lags behind the cranial bud, a floating gallbladder is created. A floating gallbladder is a gallbladder that is completely covered with peritoneum; it is usually suspended from the liver by a mesentery. A floating gallbladder is at a higher risk for torsion. Left-sided gallbladders are rare, but they have been described in the setting of situs inversus (all abdominal viscera are reversed right for left) or in the setting in which the gallbladder alone is transposed. [12] The gallbladder has also been found in other unusual locations such as the falciform ligament, [14] the abdominal wall, [15] and the retroperitoneum. [16] Regardless of the anomalous position of the gallbladder, it is important to realize that in almost all cases the cystic duct will inevitably join the common bile duct in a relatively normal position.

Abnormal development of the gallbladder bud may also cause anomalies in the cystic duct. Double cystic ducts may drain a single, nonseptated gallbladder and join the biliary system at the common bile duct or right hepatic duct. There may be absence of the cystic duct, and, in this case, the neck of the gallbladder drains directly into the common bile duct. Failure to recognize this anomaly has resulted in excision of a segment of the common bile duct during cholecystectomy in which the common bile duct has been mistakenly identified as the cystic duct. Liberal use of intraoperative cholangiography has been emphasized when intraoperative anatomy is confusing or anomalies are present.

Hepatic Ducts and Common Bile Duct

True accessory hepatic ducts result from the development of an extra bud from the biliary anlage during development. Accessory ducts are much more common on the right than the left. They may drain directly into the gallbladder, the right or left hepatic ducts, the cystic duct, or the common bile duct. Accessory hepatic ducts are present in about 10% of individuals. Cystohepatic ducts that drain bile directly from the liver to the gallbladder are rare. Aberrant hepatic ducts, unlike true accessory ducts, result from an extrahepatic course of a duct that is normally contained within the liver parenchyma. Accessory ducts provide a second drainage route from a region of the liver, in contrast to an aberrant duct, which represents the only drainage to a particular region of the liver, but lies in an unusual location. Almost all aberrant hepatic ducts represent the right anterior segmental duct, which joins the right posterior segmental duct outside the liver. This extrahepatic union occurs in about 10% of cases and may result in mistaken identification of the right anterior segment duct as the cystic duct during cholecystectomy, with inadvertent ligation. [17] Both aberrant and true accessory hepatic ducts are at equal risk for injury during cholecystectomy.

Anomalies in the common bile duct do exist but are quite uncommon. The common bile duct may contain two lumens but appears to be single externally. The common bile duct may be completely duplicated, with one duct draining the right lobe of the liver and one duct draining the left lobe. Duplicated ducts usually empty separately into the duodenum. [18], [19] Rarely, the common bile duct may open into the gastrointestinal tract at an ectopic site. The common bile duct has been reported to drain into the stomach [20] and the duodenal bulb, [21] as well as the distal duodenum. [22], [23]

Biliary Atresia

Biliary atresia is defined as the obliteration of the extrahepatic and/or intrahepatic bile ducts, with many different variations described (Fig. 99-11). The incidence of biliary atresia is 1 per 15,000 births. Ten to 15 percent of patients with biliary atresia have associated anomalies of the inferior vena cava (absence), portal vein (preduodenal portal vein), intestine (intestinal malrotation), and spleen (polysplenia). [24] Most patients have a form of biliary atresia with complete obstruction of the gallbladder and extrahepatic bile ducts, whereas 10% to 15% of patients have obstruction of the proximal hepatic ducts with patency of the gallbladder and distal common bile duct. Only 1% to 2% of patients have proximal hepatic duct patency.

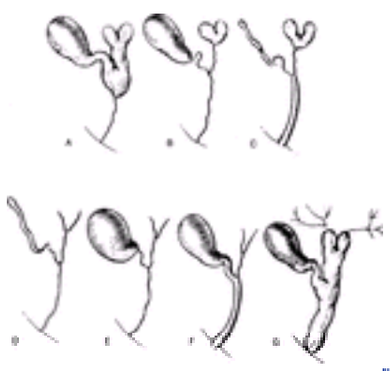

FIGURE 99-11. The different forms of biliary atresia. Biliary atresia may be partial, affecting the intra- or extrahepatic portions of the biliary tract, or it may be a complete process (**D**). (From Lindner H. Embryology and anatomy of the biliary tree. In: Way LW, Pellegrini CA, eds. Surgery of the gallbladder and bile ducts. Philadelphia: WB Saunders, 1987.)

In biliary atresia, the bile ducts are replaced by dense fibrotic tissue containing evidence of both an acute and chronic inflammatory process. [25] Typically, there is obliteration of the entire extrahepatic biliary tract, including the gallbladder. Extrahepatic biliary atresia frequently extends into the intrahepatic bile ducts, probably as an end result of a destructive process leading to fibrosis and obliteration of the biliary tree, with development of secondary biliary cirrhosis. [26] If left untreated, the typical course is one of progressive hepatic insufficiency, with an average survival of 12 to 19 months. [27]

The pathogenesis of biliary atresia is not known. Existing data seem to contradict the theory that failure of recanalization of the bile ducts during development is the cause. Data accumulated from the surgical experience using portoenterostomy have provided some insight into the disease process. Early establishment of bile drainage, typically before 60 days, can reverse the liver injury and is associated with long-term survival, while delay in establishing bile drainage until after 120 days is often associated with progression of cirrhosis and a poor outcome. In addition, infants who develop biliary atresia are rarely jaundiced at birth and frequently have bile-stained meconium; biliary atresia has rarely been demonstrated in autopsy studies of fetuses. [28] These observations suggest that biliary atresia is a dynamic process targeting the extrahepatic bile ducts, with progressive destruction rather than a static process. While the events that initiate this destructive process are unknown, experimental data suggest that neonatal Reovirus type 3 or other infectious stimuli may be causative agents. [29], [30] and [31]

The clinical presentation of biliary atresia is progressive neonatal jaundice with an onset in the first few weeks of life. Jaundice usually becomes visible in the first 2 to 4 weeks of life. Biliary atresia must be differentiated from neonatal physiologcial jaundice, as well as congenital infectious causes, such as Cytomegalovirus and rubella, and genetic diseases, including a1-antitrypsin deficiency.

Technetium 99m–iminoacetic acid (^{99m}Tc-IDA) hepatobiliary scanning is the imaging test of choice for biliary atresia, especially when preceded by a 5- to 7-day course of phenobarbital to increase bilirubin conjugation and excretion. The sensitivity of the test for biliary atresia is 100%, with a specificity of 94%. [32] An abdominal ultrasound should also be performed as a standard part of the evaluation of the jaundiced infant and is helpful to evaluate the possibility of other sources of biliary obstruction. Liver biopsy is nondiagnostic in up to 25% of patients and is often unnecessary to make a diagnosis.

Because of the near 100% mortality associated with medical management of biliary atresia, surgical therapy has become the treatment of choice. The standard operation, hepatic portoenterostomy, termed the Kasai procedure, was first described by Kasai in 1959 [33] following his original observation of residual microscopic bile channels in the fibrous tissue of the porta hepatis in patients with biliary atresia. The Kasai procedure consists of exploratory laparotomy with excision of the occluded extrahepatic biliary system and a Roux-en- enteric anastomosis to the transected fibrotic cord at that level of the porta hepatis where microscopic bile ducts have

been documented by frozen section. Primary hepatic transplantation is reserved for patients with a delayed diagnosis and advanced cirrhosis at presentation or patients with failed Kasai procedures. [34]

Most series report a 40% to 60% 5-year survival rate following portoenterostomy. [35], [36] In general, one third have good long-term results with minimal or no liver dysfunction and normal growth and development, one third do poorly and are dependent on immediate liver transplantation for survival, and one third have slow, progressive liver dysfunction over a period of months to years and subsequently need liver transplantation. Although the failure rate is fairly high, the fact that one third of patients will do well and never require transplantation and another subset will experience normal growth for a period of years before potentially requiring transplantation, in addition to the shortage of transplantable organs, has prompted centers to use the Kasai procedure as first-line therapy for biliary atresia. [37], [38] and [39]

Choledochal Cyst

See Chapter 102 (“Cystic Diseases”) for detailed discussion.

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CHAPTER 100

Sum P. Lee and Cynthia W. Ko

GALLSTONES

Autopsy studies of Egyptian and Chinese mummies have shown that gallbladder stones have existed for more than 35 centuries. Today, over 20 million Americans have gallstones, and more than 700,000 cholecystectomies are performed annually in the United States. [1] Gallstone-related symptoms and complications are among the most common gastroenterologic disorders requiring hospitalization, at an estimated annual cost of almost $6.5 billion. [2] This chapter summarizes the pathogenesis, clinical manifestations, and evolving treatment of gallstones.

EPIDEMIOLOGY

Gallstones are classified as cholesterol stones and pigment stones. In most Western countries, more than 75% of gallstones are the cholesterol type. The prevalence of gallstones varies widely in different countries and among different ethnic groups in the same country. Epidemiologic and animal studies have implicated genetic factors to account for this wide variation. [3], [4] Animal models are being developed that may help this line of investigation. [5] In addition, lifestyle factors, such as diet, obesity, weight loss, and low levels of physical activity have been implicated. [6], [7], [8], [9] and [10] Gallstone prevalence increases with age, and gallstones are more prevalent in females of any age group. [1]

Because the disease is nonfatal and often asymptomatic, the diagnosis of gallstones is not always made before death. Autopsy studies correlating with clinical information suggest that approximately two thirds of the population with gallstones are asymptomatic. [11] Methods most commonly used to determine gallstone prevalence include autopsy studies, determination of cholecystectomy rates, and ultrasonography. A Danish screening ultrasound survey showed new gallstone formation in 3% of the population older than 40 years of age in each 5-year period. [12]

The wide scatter in gallstone prevalence among different nations and ethnic groups is strong evidence that genetic factors are involved in gallstone formation. Those with the highest rates of gallstone formation are the Pima Indians of North America, Chileans, and Caucasians in the United States. [3], [11], [13] Next are the populations of Sweden, Germany, and Austria, followed by New Zealand, Great Britain, Norway, Ireland, and Greece. [14], [15], [16], [17] and [18] At the bottom of this list are the Asian populations in Singapore and Thailand. [19], [20]

The type of gallstone also varies among cultures. In developed countries, cholesterol gallstones are most common and usually occur in the gallbladder. In Africa and Asia, where the prevalence of gallstones is low, pigment gallstones are most common, and stones also can be found in the bile ducts.

Progressive increases in the rate of gallstones during the 20th century support a role for lifestyles and dietary factors in the pathogenesis of gallstones. Cholecystectomy rates partly reflect the provision of medical services, and autopsy gallstone rates may vary even within a country during a given time period. Nevertheless, several studies have shown increases in the prevalence of gallstones over time. [17], [21], [22] and [23] Post–World War II westernization in Japan is one example. Since the late 1940s, the prevalence of gallstones in Tokyo has more than doubled, and there has been a change from pigment to cholesterol gallstones. [22] In Saudi Arabia over a 10-year period, the frequency of cholecystectomy has increased sharply, a rise not matched by the increase in population, use of medical services, or number of other surgical procedures. A shift to a more Western diet has been suggested as a possible explanation. [24]

ETIOLOGY

Pathogenesis of Cholesterol Gallstones

The total body pool of cholesterol is supplied by de novo synthesis from acylcoenzyme A (acyl-CoA) and dietary absorption. [25], [26] Most of this pool is solubilized and secreted unmodified in bile or converted to bile acids (Fig. 100-1). Approximately 20% of biliary cholesterol comes from new hepatic synthesis. [27], [28] The remainder originates from a preformed pool within the liver. [29], [30] Important contributors into this pool include hydrolysis of cholesteryl ester stores, dietary sources such as chylomicrons, and direct hepatic and extrahepatic synthesis of the lipoproteins high-density lipoprotein (HDL), low-density lipoprotein (LDL), and very-low-density lipoprotein (VLDL). Total hepatic cholesterol is tightly regulated within a narrow range. [31], [32] The association of gallstones with serum cholesterol levels is not straightforward; although gallstones may be linked to decreased serum HDL cholesterol and increased serum triglyceride levels, no such association has been found for total serum cholesterol levels. [33], [34]

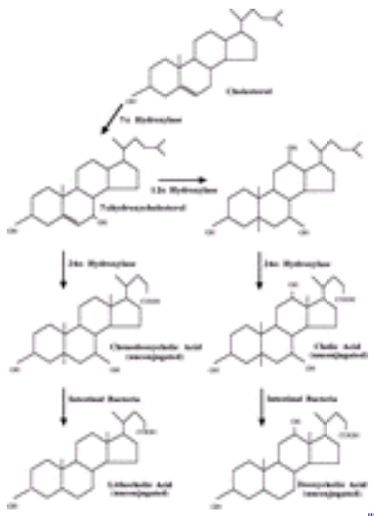

FIGURE 100-1. Schematic representation of bile acid synthesis and metabolism. The initial step in bile acid synthesis is hydroxylation of cholesterol by the enzyme 7a-hydroxylase. Further conversion of this metabolite by 12a-hydroxylase and 26a-hydroxylase results in synthesis of the primary bile acids, cholic acid, and chenodeoxycholic acid. Following conjugation with glycine or taurine, secretion, and transport to the intestine, bacteria metabolize the primary bile acids to the secondary bile acids, lithocholic acid and deoxycholic acid.

Various defective metabolic states can disrupt this regulatory balance, resulting in absolute biliary cholesterol hypersecretion or relative bile acid hyposecretion. Both defects can coexist, as in the case of the Pima Indians. [35]

Several mechanisms may be operative in biliary cholesterol hypersecretion. [36], [37], [38], [39] and [40] Nonobese gallstone patients may have higher intracellular levels of total and free cholesterol, suggesting enhanced intracellular cholesterol transport. [41] Clinical situations associated with excessive secretion of biliary cholesterol include obesity, aging, drug effects, and hormonal therapy. [42] Table 100-1 summarizes some of these examples and the proposed underlying pathogenesis. Recent animal studies have indicated that there are specific genes that determine the incidence of cholesterol stone formation in mice fed a lithogenic diet. [4] For example, the *Lith1* gene is associated with gallstone disease in a dominant fashion. [5] Further work has identified the bile-salt–export pump as a candidate protein for the *Lith1* gene. [43] Other candidate genes associated with gallstones in mice include HMG CoA reductase, canalicular multispecific organic anion transporter (*Cmoat*), intracellular lipid transporters (*Pctp* and *Fabp6*), lipoprotein lipase, and lecithin-cholesterol acyltransferase. [4] Although the human homologs to these animal genes are still being identified, the *ApoE4* genotype has been linked to an increased incidence of cholesterol gallstones in humans. [44], [45] and [46] ApoE, a lipid transfer protein, is reported to be associated with stones with high cholesterol content, [45] faster dissolution when treated with ursodeoxycholic acid, and higher recurrence rates after lithotripsy. [46] ApoE may have a role in regulation of biliary cholesterol secretion. [47] The role of apoE, and its interactions with dietary factors, deserves further study in humans.

CLINICAL EXAMPLES	PATHOGENESIS
Obesity, hyperlipoproteinemia	Increased hydroxymethylglutaryl-coenzyme A activity leading to increased mevalonic acid and cholesterol synthesis
Progesterone, oral contraceptives, clofibrate	Inhibition of hepatic acyl-coenzyme A cholesterol acyltransferase activity leading to decreased conversion of cholesterol to cholesteryl ester stores
Estrogens	Increased lipoprotein receptors B and E leading to increased hepatic cholesterol uptake; inhibition of 12α-hydroxylase leading to decreased synthesis of chenodeoxycholic acid
Selected nonobese Japanese	Increased hepatic intracellular transport of cholesterol
Selected nonobese Caucasians	Reduced activity of 7α-hydroxylase leading to a defect in conversion of cholesterol to bile acids; increased enterohepatic cycling of bile acids, leading to increased biliary deoxycholate levels
Age	Age-related decrease (?) in 7α-hydroxylase activity
Marked weight reduction	Mobilization of tissue cholesterol

TABLE 100-1 Risk Factors Associated With Cholesterol Hypersecretion in Bile

Alternatively, biliary supersaturation may occur with relative bile acid hyposecretion [48], [49] and [50] (Table 100-2). A diminished bile acid pool and secretion rate may result from excessive intestinal losses or decreased production. In nonobese patients with cholesterol gallstones, cholic acid and chenodeoxycholic acid pools are reduced, and deoxycholic acid is often increased in bile. [50] Conversion of cholic acid to deoxycholic acid, possibly in the small intestine or by colonic bacteria, may account for these changes. [51], [52] With an expanded deoxycholate pool in bile, biliary cholesterol secretion rises. [53]

CLINICAL EXAMPLES	PATHOGENESIS
Ileal disease, bypass, or resection	Impaired bile acid absorption or excessive losses
Cerebrotendinous xanthomatosis	Inherited 26-hydroxylase deficiency leading to incomplete oxidation of the cholesterol side chain and decreased bile acid production
Congenital 12-hydroxylase deficiency	Decreased cholate and deoxycholate synthesis
Selected nonobese Caucasians	Excessive feedback suppression of bile acid synthesis leading to a decreased pool, hepatic return, and secretion
Primary biliary cirrhosis Chronic cholestasis	Decreased bile acid secretion

TABLE 100-2 Risk Factors Associated With Relative Bile Acid Hyposecretion

Cholesterol Solubilization

Free cholesterol is virtually insoluble in aqueous solution. Bile acids, because of their unique amphipathic properties, are able to solubilize cholesterol and phospholipids in mixed micelles. [54], [55] Phase equilibrium diagrams can be constructed to characterize cholesterol solubility in a defined milieu of various lipid concentrations. Using these data, the Cholesterol Saturation Index (CSI) can be calculated. [56] CSI is the ratio of the actual amount of cholesterol in a given bile sample to the maximal cholesterol carrying capacity of that sample determined in vitro. Bile that has a CSI greater than 1 is considered supersaturated. Supersaturated bile that does not form cholesterol crystals is called metastable.

In addition to classic micelle packaging, cholesterol can be solubilized with phospholipid (principally phosphatidylcholine) as unilamellar vesicles ranging in size from 40 to 100 nm. [57], [58], [59] and [60] Vesicles consist of a bilayer of phospholipid interdigitated with cholesterol, without associated bile acids. These vesicles are thought to be assembled within the hepatocyte and then transported to the biliary canaliculus. [61] Vesicles comprise a separate and distinct carriage system that probably represents the major mode of cholesterol transport from the hepatocyte.

Bile acid secretion is the primary driving force behind the biliary secretion of cholesterol and phospholipid. [62], [63] As bile acids are actively secreted against a gradient

into the canaliculus, they induce the subsequent secretion of cholesterol-phospholipid vesicles into bile. Hydrophobic bile acids are more effective in provoking lipid secretion, although the maximum rate of biliary lipid secretion is similar for all bile acids. [64] Within the canaliculus, the bile acid concentration rises, reaching a micellar concentration that allows conversion to mixed micellar carriers. This interchange continues in a dynamic fashion within the ductules, ducts, and gallbladder. [65] The relative percentage of cholesterol transported in the micellar fraction can vary dramatically, depending on the physiological conditions. For example, at low bile acid secretion rates, as with fasting, the predominant form of cholesterol carriage in hepatic bile is vesicular. [57], [58] Conversely, the higher bile acid concentration in the gallbladder favors a shift toward micelle formation [57], [58] (Fig. 100-1 and Fig. 100-2).

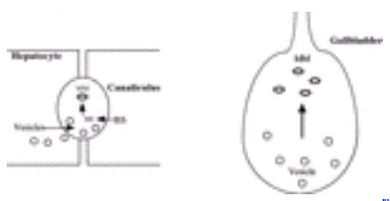

FIGURE 100-2. Schematic representation of biliary lipid secretion. At the biliary canalicular membrane of the hepatocyte, vesicles of cholesterol and phospholipids are secreted. Bile salts (*BS*) are also secreted by a different mechanism. Inside the canaliculus, mixed micelles (*MM*) of bile salts, cholesterol, and phospholipids are formed when bile salts reach a critical micellar concentration. As bile is further concentrated in the gallbladder, more vesicles are converted to mixed micelles. Mixed micelles transfer more phospholipid than cholesterol in solubilizing biliary lipids. Vesicles in the gallbladder are relatively depleted in phospholipid and enriched in cholesterol. These vesicles are more unstable and prone to nucleation.

Nucleation of Cholesterol Crystals

An early and indispensable step in cholesterol gallstone formation is nucleation, the emergence of solid cholesterol crystals in saturated bile (Fig. 100-3). Antecedent vesicular fusion and aggregation may be crucial for crystal generation. Nucleation of crystals results in a diminution of vesicular cholesterol without effect on micellar cholesterol. [57] Under video-enhanced microscopy, crystals appeared to originate from aggregated vesicles. [66], [67] and [68] Thus, cholesterol in vesicles may be more unstable and prone to precipitate than that in mixed micelles.

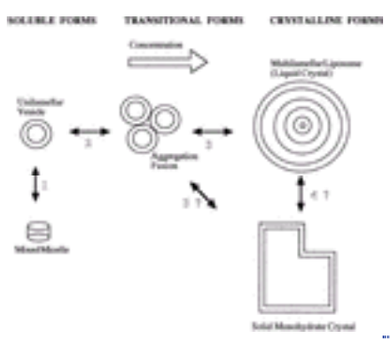

FIGURE 100-3. Schematic representation of cholesterol nucleation. In bile, cholesterol exists in two soluble forms, vesicles and mixed micelles. These forms are in dynamic equilibrium (*1*) depending on the total bile salt and lipid concentration. With concentration, vesicles are depleted of phospholipid and enriched in cholesterol, resulting in a tendency for vesicles to aggregate and fuse (*2*). Pronucleators may also cause vesicular fusion. The fused vesicles lead to formation of multilamellar liposomes or liquid crystals (*3*). Cholesterol separates from the liquid to the solid crystalline form during nucleation (*4*). Whether vesicles can nucleate into solid cholesterol crystals is unclear.

Vesicles in bile do not represent a homogeneous population. Gallbladder bile vesicles are likely to participate in nucleation; vesicles in hepatic bile are much more resistant to nucleation. The discrepancy in their behavior partly is caused by the difference in their relative cholesterol-to-phospholipid content. Vesicles that have increased ratios of cholesterol to phospholipid aggregate, fuse, and nucleate more readily. [68] The concentrated bile acids within the gallbladder may preferentially remove vesicular phospholipid over cholesterol during the dynamic shift to the micellar phase. The remaining vesicles are relatively cholesterol enriched and prone to nucleation. Conversely, hepatic vesicles have lower cholesterol-to-phospholipid contents and are therefore more stable.

Many people secrete supersaturated bile without developing gallstones. [69] Gallbladder bile taken from patients with cholesterol stones nucleates more rapidly than equally saturated bile from controls. Together, these observations suggest that factors in addition to cholesterol supersaturation must be involved in nucleation. Considerable efforts have been made to identify specific nucleating factors. Biliary proteins have generated the most attention, as total biliary protein content is increased in bile with cholesterol crystals compared with samples without crystals. [70]

Burnstein and colleagues provided the first major evidence for a potent pronucleating agent in bile. [71] A small-molecular-weight glycoprotein in bile binds to concanavalin A and exhibits promoting abilities. [72], [73] Hepatic bile from patients with cholesterol gallstones contained proteins associated with vesicles; when purified, these vesicles had potent cholesterol-nucleating activity. [74] A small, pigment-associated, anionic protein appears to be important in the regulation of calcium salts and in cholesterol precipitation in bile. [75], [76] and [77] Other putative pronucleators include a $_1$-acid glycoprotein aminopeptidase N, immunoglobulins G and M, haptoglobin, fibronectin, and a $_1$-antichymotrypsin. [78], [79], [80], [81], [82], [83], [84] and [85] However, the role of these proteins in vivo remains controversial. [86], [87] and [88] In contrast to these proteins, other biliary protein fractions, such as apolipoproteins AI and AII, a biliary glycoprotein heterodimer, and immunoglobulin A (IgA) inhibit in vitro nucleation assays. [89], [90], [91] and [92]

Other research has focused on the role of gallbladder mucin, a high-molecular-weight glycoprotein that is the major organic constituent of gallbladder mucus. It consists of a polypeptide core with multiple oligosaccharide side chains. These peptide cores also contain hydrophobic domains that bind pigment, cholesterol, and phospholipid, entrapping vesicles and micelles in its binding sites. [93], [94] With this close contact, membrane fusion and mixing can occur with formation of multilamellar vesicles. Addition of mucin to supersaturated bile will greatly accelerate cholesterol crystal nucleation. [88], [95] Mucin may become incorporated into the core of the growing gallstone, providing an architectural framework for crystal growth and stone formation. [96] Thus, mucin can act as a pronucleator. [97] Mucus hypersecretion before stone formation appears to be a universal antecedent in a number of animal models of lithogenesis. [98], [99] The stimulus for mucin hypersecretion is not clear, although prostaglandins may play a role. For example, the hypersecretory response can be inhibited by the administration of large doses of oral aspirin in the human gallbladder, [100] and aspirin ingestion can completely abolish subsequent formation of cholesterol crystals and gallstones. [101] However, inhibition of prostaglandin synthesis with indomethacin did not prevent mucin hypersecretion in a prairie dog model. [102], [103] Although prostaglandin activity may play a role in the pathogenesis of mucin hypersecretion, the role of other factors such as lithogenic bile or hydrophobic bile salts awaits study. [104], [105] and [106] More recently, oxy- sterols have been identified in bile of patients with pigment gallstones, [107] and the presence of these oxysterols may also affect mucin secretion. [108] The gallbladder can also secrete in response to gastrointestinal peptides such as secretin, assisting the emptying of precipitate. [109]

Calcium complexes are also important, as they are present in the central matrices of many, if not all, cholesterol stones, where they may serve as potential nidi for cholesterol crystallization. [110], [111], [112] and [113] Calcium has been shown to bind to mixed micelles, simple micelles, and simple vesicles in vitro, potentially explaining its role in cholesterol crystal nucleation. [114] Calcium may also regulate gallbladder secretion of electrolytes and glycoproteins. [115], [116] Various studies have found conflicting data about levels of biliary calcium in gallstone patients. [117], [118] and [119] Free and total calcium increased in the bile in obese patients who developed

gallstones during weight reduction. [120]

Role of the Gallbladder

The importance of the gallbladder in cholesterol gallstone pathogenesis must be emphasized. Cholecystectomy essentially cures recurrent de novo cholesterol stone formation. Gallbladder mucosal function plays an important role in this pathogenic process. The gallbladder can absorb excess water and electrolyte and concentrate bile. [121] It also normally secretes hydrogen ions and mucin. Sodium and chloride ion fluxes are altered in an animal model of lithogenesis. [122]

Alterations of lipid metabolism in gallbladder epithelium that might increase cholesterol content have also been studied. Hydroxymethylglutaryl-CoA reductase activity, governing cholesterol synthesis, is lower in the gallbladder mucosa than in hepatic tissue, and acyl-CoA:cholesterol acyltransferase (ACAT) activity catalyzing the esterification of cholesterol is several times higher than in the liver. [123] A study also suggests that normal human gallbladders can absorb phospholipids and cholesterol from bile in vitro, resulting in bile becoming less lithogenic. [124] In contrast, gallbladders from patients with cholesterol stones were less able to absorb cholesterol and phospholipids, resulting in potentially more lithogenic bile. This impaired ability to absorb biliary lipids may be an additional pathogenetic factor for gallstones.

In addition to epithelial cell function, gallbladder motility is of vital importance. Gallbladder stasis is a risk factor for lithogenesis, as mucin gels to accumulate with prolonged bile storage. The viscosity of these gels may interfere with mechanical emptying. Increased cholesterol nucleation and crystal growth as well as enzymatic and nonenzymatic hydrolysis of bilirubin conjugates could occur with prolonged stasis. The resulting unconjugated bilirubin may precipitate with calcium.

Altered gallbladder contractility before cholesterol stone formation has been shown in a lithogenic animal model. [125] In humans, prolonged total parenteral nutrition (TPN) induces gallbladder hypomotility and profound stasis, predisposing to subsequent biliary sludge and possible stone development. [126] High spinal cord injury is linked to gallbladder stasis and gallstone formation. [127], [128] Pregnancy, oral contraceptives, obesity, diabetes mellitus, and octreotide therapy are also associated with impaired emptying and cholelithiasis. [129], [130] Hormonal influences and prostaglandins are postulated mediators of gallbladder contractility. [131]

Biliary Sludge

Precipitates in bile have been called by many names—biliary sludge, microlithiasis, and pseudolithiasis. Sludge is best diagnosed by microscopic examination of a fresh sample of gallbladder bile. [132] Sludge also can be seen ultrasonographically as low-amplitude echoes without postacoustic shadow that layer with gravity. [133], [134] In certain clinical conditions, sludge may evolve into cholelithiasis.

Biochemically, sludge is composed of calcium bilirubinate granules and cholesterol monohydrate crystals embedded in a mucus gel. Ceftriaxone may precipitate as a calcium salt that appears ultrasonographically as sludge. [135] These calcium precipitates, with cholesterol crystals 50 µm or more in diameter, produce the characteristic ultrasonic echoes in sludge. The deformable mucin gel accounts for its unique layering and flow characteristics. [134]

In a prospective study of patients receiving prolonged TPN, 6% of the patients developed sludge during the initial 3 weeks. By 4 to 6 weeks, 50% of the patients had sludge, and after 6 weeks, its appearance was universal. [136] Stones developed in 43% during follow-up. Significantly, sludge resolved with reinstitution of oral feedings in all the patients by the end of 4 weeks. Although stasis was undoubtedly the major factor in sludge formation, these patients represented a medically ill population with complicating metabolic derangements.

Gallbladder sludge may also appear spontaneously in selected individuals. The clinical outcome of a group of patients who presented with upper abdominal pain and had sludge documented on initial ultrasound examination was examined. [134] In approximately 18%, the sludge resolved over a 2-year period. It had a disappearing and reappearing course in about 60%. Gallstones also developed in 14%. Another retrospective study demonstrated similar findings. [137] In most, sludge disappeared spontaneously after 2 months. omplications including stones or acute acalculous cholecystitis developed in 19.6%. These collective cohorts may represent patients with de novo cholesterol stone formation who have been detected at an early stage.

One scenario of cholesterol lithogenesis begins first as a defect in the hepatic secretion of biliary lipids, resulting in cholesterol supersaturation of bile. Gallbladder mucin hypersecretion and stasis develop, perhaps through the action of chemical mediators. As the bile is progressively concentrated, the vesicular fraction becomes relatively cholesterol enriched. These vesicles aggregate and fuse. When the balance of the opposing promoting and inhibitory factors favors nucleation, cholesterol crystals form. The crystals formed are retained within the gallbladder, where they grow and conglomerate with mucin glycoprotein and other constituents such as calcium and bilirubin. Continued stasis and impaired emptying of cholesterol precipitates lead to macroscopic stone formation.

Pathogenesis of Pigment Gallstones

Chemical Composition and Chemical Associations Pigment gallstones are subclassified into brown and black types, which differ in morphology, pathogenesis, and clinical associations (Table 100-3). In contrast to cholesterol gallstones, ethnic origin is not an important determinant of pigment gallstones. Black pigment stones can occur in persons with no predisposing factors. In the United States, black pigment gallstones are found in the gallbladder and are not associated with bacterial infection.

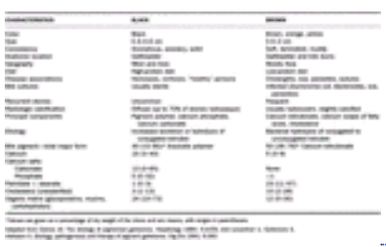

TABLE 100-3 Clinical Associations and Compositions of Black and Brown Pigment Gallstones

Factors associated with the formation of black pigment stones include chronic hemolysis (e.g. sickle cell disease), thalassemia, cardiac valvular prosthesis, advancing age, long-term TPN, and cirrhosis. [138], [139], [140] and [141] Black pigment stones seldom coexist with cholesterol stones in the same gallbladder. A calcium-regulatory protein that may mediate calcium precipitation has been isolated from black pigment stones. [142] In Asia, brown pigment stones are often found in the bile ducts and are strongly associated with bacterial infection. [143] There is a high incidence of infection with *Escherichia coli,* with the bacterial presence within stones easily demonstrated. [144], [145] and [146] Biliary secretory IgA is decreased. [147] Infected bile exhibits high bacterial ß-glucuronidase activity, often associated with a juxtapapillary duodenal diverticulum. [148], [149]

Deconjugation of Bilirubin Bilirubin, a tetrapyrrole-like cholesterol, is insoluble in water. After glucuronidation, bilirubin is secreted, mostly as the diglucuronide (75%–80%) or the monoglucuronide (20%), with a small amount of unconjugated bilirubin (3%). Because black and brown pigment stones contain calcium-bilirubin salts, the pathogenesis of pigment stones involves the deconjugation and precipitation of bilirubin. In patients with chronic hemolysis, there is a tenfold increase in the excretion of conjugated bilirubin in hepatic bile. [150] In an animal model of ileal resection, bilirubin circulates enterohepatically during the early postsurgical period, with increases in biliary levels of this molecule. [151] In patients with Crohn's ileitis or who had undergone ileal resection, concentrations of conjugated and unconjugated bilirubin were increased compared to patients with Crohn's or ulcerative colitis. In these studies, it has been hypothesized that unconjugated bilirubin is absorbed from the colon in the presence of ileal disease. These findings may help explain the increased incidence of pigment stones in patients with ileal disease. Bile acid mixed micelles and an acidic milieu promote bilirubin solubilization. The role of biliary ß-glucuronidase in the pathogenesis of pigment gallstones has been controversial. Studies employing molecular genetic techniques have implicated bacterial infection in producing brown pigment stones. [152], [153] It has been hypothesized that bacterial ß-glucuronidase hydrolysis of conjugated bilirubin into insoluble bilirubin leads to infection-related brown pigment stones. However, this hypothesis does not seem to be relevant to black pigment gallstones. [154] ß-Glucuronidase activity in uninfected bile has been demonstrated, suggesting that this enzyme may also originate in the epithelium. [155], [156] In support of this, a study showed that gallbladder bile from patients with pigment, cholesterol, or mixed gallstones has a higher proportion of bilirubin monoglucuronide than that of controls. [157] However, another risk factor, gallbladder stasis, must be present for precipitates of calcium bilirubinate to form.

Stasis, similar to the scenario described for cholesterol nucleation, can provide an opportunity for nonenzymatic hydrolysis of bilirubin diglucuronide and subsequent precipitation. [158], [159] Prolonged TPN, with attendant gallbladder stasis, can result in the formation of gallbladder sludge and black pigment stones. [136], [160] The gallbladder may contribute to pigment gallstone formation in other ways. Calcium homeostasis is intimately linked to black and brown pigment gallstones, because both kinds contain predominantly calcium bilirubinate. Hepatic bile is supersaturated with calcium carbonate. The gallbladder epithelium can acidify bile, increasing the solubility of calcium carbonate. Inability of an inflamed gallbladder to acidify bile may contribute to pigment gallstone formation. Gallbladder epithelium secretes a matrix of mucous glycoprotein into bile that binds to bilirubin and other hydrophobic lipids and is contained in black and brown pigment gallstones. [93] A working hypothesis can be made of pigment gallstone formation. Bilirubin excretion is increased. Gallbladder stasis or bile infection allows the hydrolysis of bilirubin diglucuronide to less soluble forms. Inability of an inflamed gallbladder mucosa to acidify bile perturbs calcium and bilirubin olubility. Mucus secreted by the gallbladder epithelium buffers hydrogen ions and decreases acidity enabling calcium carbonate, phosphate, and bilirubin to precipitate. For reasons poorly understood, the calcium bilirubinate in brown pigment stones remains in the monomeric form, but undergoes significant cross-linking to become polymeric and crystalline in black pigment stones.

CLINICAL MANIFESTATIONS

Biliary Colic

Approximately one third of patients with gallstones are symptomatic. Biliary colic is the main complaint in 70% to 80% of symptomatic patients. This is a visceral pain of tonic spasm resulting from transient obstruction of the cystic duct by a stone. Implicit in the term biliary colic is the fact that the gallbladder mucosa does not show features of acute inflammation. The pain of biliary colic is thought to be caused by functional spasms around an obstructed cystic duct; pain of acute cholecystitis is caused by gallbladder wall inflammation.

Although sometimes quite variable, biliary pain often has specific characteristics. The pain is episodic and severe, and it is located in the epigastrium or less frequently in the right upper quadrant, left upper quadrant, the precordium, and the lower abdomen. [161], [162] The pain may be precipitated by eating a large meal, but often no single food is the lone culprit. Pain may develop without any precipitating events.

Typically, the pain has a sudden onset and rises steeply in intensity over a 15-minute interval to a steady plateau lasting as long as 3 hours. Resolution of pain is slower. Pain lasting more than 6 hours should raise the suspicion of cholecystitis. The pain may radiate to the interscapular region or, rarely, to the right shoulder tip. Vomiting and diaphoresis are not uncommon. The patient is usually restless and unable to find a comfortable position. Residual tenderness in the upper abdomen may persist after an attack. The interval between attacks is unpredictable and may last weeks, months, or years.

True biliary colic should be differentiated from nonspecific dyspepsia. Flatulence, pyrosis, aerophagia, vague abdominal discomfort, and fatty food intolerance are common complaints of many patients, whether or not they have cholelithiasis. [163] Whether patients with gallstones experience increased distress from fatty foods has been difficult to prove.

The ability to differentiate true biliary pain from nonspecific abdominal symptoms significantly impacts the success of treating gallstone disease. For example, a cholecystectomy performed for gallstone-induced biliary colic is usually curative, but symptoms often persist if it is done for patients with nonspecific dyspepsia and cholelithiasis. [164], [165] It is worthwhile to clarify prevalent misunderstandings about biliary pain. First, biliary colic is a misnomer, because the pain is steady, not intermittent or fluctuating. Second, the pain is primarily epigastric. It is inappropriate to interpret pain not located in the right upper quadrant as atypical of gallstone symptoms. Third, fat intolerance is not a feature of biliary colic. Fourth, despite the availability of many imaging techniques to demonstrate the presence of gallstones, the diagnosis of biliary colic is ultimately based on clinical judgment.

Acute Cholecystitis

The most common cause of acute cholecystitis is obstruction of the cystic duct by gallstones, resulting in acute inflammation of the gallbladder. Approximately 90% of cases are associated with cholelithiasis. Obstruction likely damages the gallbladder mucosa, initiating an inflammatory reaction involving prostaglandins and other chemical mediators. [166], [167] Hydrolysis of biliary lipids such as lecithin and reabsorption of bile salts may also play a role. [168], [169] Although bacteria are commonly found in the gallbladder bile of patients with acute cholecystitis, this is probably a secondary event. [170], [171] Secondary bacterial infection can progress to empyema.

Most patients with acute cholecystitis have had previous attacks of biliary pain. The pain of acute cholecystitis typically lasts longer than 3 hours and, at the end of 3 hours, shifts from the epigastrium to the right upper quadrant with the emergence of localized tenderness. With time, the intensity of pain may diminish, but tenderness increases. Vomiting is common. Fever is common, but is rarely higher than 101°F. The sequence of clinical features represents visceral pain of ductal impaction by stones, proceeding to gallbladder inflammation with parietal pain.

In older adults, presenting signs and symptoms may be especially mild. On physical examination, the Murphy sign, an abrupt arrest in inspiration secondary to pain elicited during direct palpation of the right upper quadrant, may be present. In 30% to 40% of patients, a palpable mass may be present, consisting of the gallbladder and perhaps adherent omentum. Fifteen percent of patients with acute cholecystitis will be jaundiced, even without choledocholithiasis and obstruction. The pathogenesis may involve edema and compression around the inflamed cystic duct. Pyogenic or amebic liver abscesses are in the differential diagnosis.

Chronic Cholecystitis

Patients with chronic cholecystitis usually have gallstones and have had repeated attacks of biliary pain or acute cholecystitis. This results in a thickened and fibrotic gallbladder, which shows evaginated mucosal pouches (Rokitansky-Aschoff sinuses) on histological examination. It is uncommon for the gallbladder to be palpable during an attack of pain. The patient may have fewer symptoms referable to the gallbladder but may present with associated gallstone complications such as recurrent pancreatitis, choledocholithiasis, and cholangitis.

Choledocholithiasis and Cholangitis

Gallstones passing into the common bile duct from the gallbladder may proceed to the duodenum if the stone is small. They may also remain in the common bile duct and give rise to complications. The majority of stones in the common duct have the same composition as those in the gallbladder. However, some are softer and more brownish because of deposits of calcium bilirubinate and other calcium salts, including fatty acid complexes. [139] Common duct stones are frequently associated with infected bile, which may or may not be clinically apparent.

Choledocholithiasis is one of many causes of obstructive or “surgical” jaundice, which must be differentiated from hepatocellular or “medical” jaundice. Obstruction of the passage of bile results in jaundice and pruritus. The pathogenesis of pruritus is unclear, although retention of bile salts with irritation of sensory nerve endings or retention of endogenous opiate agonists have been proposed as mechanisms. Sometimes pruritus can be the predominant or incapacitating symptom. With biliary obstruction, the feces may be hypocholic or acholic. Light-colored or clay-colored stools are uncommon in bile duct obstruction resulting from gallstones because the obstruction is rarely complete, but are more commonly observed with malignant strictures of the common bile duct.

Unlike malignant obstruction of the common bile duct, a palpable and nontender gallbladder due to cholelithiasis is uncommon. The biliary obstruction is usually incomplete, and the gallbladder itself is likely to be fibrotic, scarred, and nondistensible secondary to chronic cholecystitis. However, the exceptions to this rule (Courvoisier law) are substantial, with frequent false-positive and false-negative results.

Obstruction of the common bile duct also causes increased intraductal pressure, inhibiting bile flow and causing the reflux of bile from the canaliculus into the sinusoids. The normal pressure in the common bile duct is 10 to 15 cm H_2O; with obstruction, the pressure can rise to above 40 cm H_2O. [172], [173] This also distends the biliary tree. Dilation of the extrahepatic and intrahepatic bile ducts is a valuable sign of common bile duct obstruction and is best seen using ultrasonography or computed tomography (CT) scan. Clinically, the only manifestations may be mild hepatomegaly or right upper quadrant tenderness.

With prolonged biliary obstruction, secondary hepatic parenchymal damage may set in. With increased fibrogenesis induced by bile duct obstruction, secondary biliary cirrhosis may occur. The propensity for developing cirrhosis varies with the completeness and duration of obstruction. The average time for choledocholithiasis

to result in secondary biliary cirrhosis is about 5 years. [174] Patients may present with portal hypertension or hepatic failure. Patients with incomplete obstruction more commonly present with variceal bleeding, and those with complete obstruction with hepatic failure. [175] Even if the patient has cirrhosis, every effort should be made to relieve the obstruction, since reversal of portal hypertension and secondary biliary cirrhosis have been reported. [176]

A common complication of choledocholithiasis is cholangitis. For bacterial infection to occur, obstruction or bile stasis is always present, although obstruction is not always associated with overt bacterial infection. For example, cholangitis is extremely common in choledocholithiasis, not uncommon in sclerosing cholangitis, and relatively uncommon (10%–15%) in malignant bile duct strictures.

The typical clinical picture, occurring in 70% of cases, consists of biliary pain, jaundice, and chills and rigors (Charcot triad). Pain, usually characteristic of biliary colic, occurs in 90% of patients. Bacteremia with chills and fever occurs in 95%. Clinical jaundice is present in 80%. Clinical signs are nonspecific, with mild hepatomegaly, tenderness, and occasionally rebound tenderness. [177] Depending on the progress of the illness, shock, multiple liver abscesses, or multi-organ failure may result. In contrast, for many patients, cholangitis is a short and self-limited illness complicating choledocholithiasis or sclerosing cholangitis.

Blood cultures are often positive and reflect the organisms infecting the bile ducts. The most commonly found organisms are *E. coli, Klebsiella, Pseudomonas,* and enterococci. Co-infection with anaerobes occurs in 15%. [178], [179] and [180]

Acute pancreatitis may be precipitated by the passage of stones or sludge in the common bile duct, and sludge or microscopic stones may account for a significant proportion of cases of idiopathic pancreatitis. [132], [181]

DIFFERENTIAL DIAGNOSIS

Clinical Signs and Symptoms

The clinical symptoms and signs of biliary tract disorders are not highly specific. The patient's history, physical findings, and laboratory data should be carefully examined. A clinical impression of biliary colic should be confirmed by imaging studies. Because gallstones are found commonly and may coexist with other disorders, the finding of gallstones does not exclude other diseases contributing to or complicating the patient's clinical picture. Disorders of other visceral organs, including the upper gastrointestinal tract, colon, kidneys, and pancreas, should be excluded by appropriate diagnostic tests. Extra-abdominal disorders that may produce a similar clinical picture include angina pectoris, dissecting aortic aneurysm, spinal neuralgia, pleuritis, pericarditis, and uncommon metabolic disorders such as hereditary angioedema (C1 esterase inhibitor deficiency) and acute intermittent porphyria.

In addition to pain, patients with acute cholecystitis usually present with symptoms and signs of local inflammation (e.g., right upper quadrant mass, tenderness) and systemic toxicity (e.g., fever, leukocytosis). The differential diagnosis includes other causes of intra-abdominal inflammation or infection. Acute appendicitis can cause a difficult diagnostic problem. The periumbilical pain shifting to the right lower quadrant with an inflamed mass may be confused with biliary colic with an inflamed gallbladder. A gallbladder can be low lying or an appendix can be subhepatic. Fever and leukocytosis are features of both. Ultrasonography or hepatobiliary scintigraphy may be helpful.

It may be difficult to differentiate acute pancreatitis from cholecystitis because the two conditions produce tenderness in an overlapping area. Gallstones may cause acute pancreatitis, and cholecystitis and pancreatitis therefore may coexist. Acute cholecystitis alone can be associated with hyperamylasemia, but pancreatitis often has higher enzyme levels. Biliary scintigraphy and imaging techniques such as ultrasound and CT scans are helpful diagnostically.

Perforated peptic ulcer usually produces more dramatic pain and peritoneal signs. Plain abdominal x-ray films or CT scans often show free intra-abdominal air. If free air cannot be visualized and ulcer perforation is still suspected, an urgent radiographic study with water-soluble contrast should demonstrate the perforation.

Diagnostic Studies

Laboratory Tests In uncomplicated biliary colic, there are usually no accompanying changes in hematologic and biochemical tests. In acute cholecystitis, leukocytosis with a "left shift" is usually seen, but this does not differentiate it from other intra-abdominal infections. The amylase level may be elevated because of transient obstruction of the pancreatic duct by a common duct stone. Edema and inflammation of the gallbladder can partially obstruct the common bile duct, causing mild elevation of the serum aminotransferases and alkaline phosphatase. Elevation of serum bilirubin can also be associated with the enzyme level changes, especially if the common hepatic duct or common bile duct is involved in the inflammatory reaction. Bilirubin elevation is proportional to the degree of obstruction. Bile canalicular and ductular cells produce alkaline phosphatase, the elevation of which does not depend on the magnitude or cause of obstruction. Abdominal pain, fever, and jaundice are often the presenting features of choledocholithiasis. In such cases, the bilirubin level is usually between 2 and 10 mg/dL, and the alkaline phosphatase is less than five times normal. If the level of bilirubin is above 15 mg/dL, a neoplastic obstruction should be strongly suspected. A cholangiogram should be obtained to define the cause and level of obstruction if ductal dilation is documented by ultrasonography or CT scans.

Radiologic Studies Plain abdominal films are often obtained. They are rarely useful in biliary colic because only 13% to 17% of gallstones contain sufficient calcium to be radiopaque. In acute cholecystitis, x-ray films are obtained to exclude other intra-abdominal causes of abdominal pain, fever, and leukocytosis, such as perforated ulcer or intestinal obstruction. Occasionally, when emphysematous cholecystitis is present, intramural gas outlining the gallbladder can be seen, but ultrasonography is a better source to show this. Ultrasonography has high specificity and sensitivity for the diagnosis of gallstones and should be a routine examination in the evaluation for gallstone disease. Stones appear as high-amplitude echoes with postacoustic shadow (Fig. 100-4). Other findings include thickening of the gallbladder wall (>2 mm), intramural gas, and a pericholecystic collection of fluid. The latter two suggest active gallbladder inflammation or infection. Biliary sludge may also be found and is common in extrahepatic biliary obstruction. In the absence of distal obstruction, sludge can be associated with abdominal pain (i.e., biliary colic), acute cholecystitis, or pancreatitis and should be regarded as part of the spectrum of gallstone disease. [133], [134] Dilation of intrahepatic or extrahepatic bile ducts suggests distal obstruction. However, a diagnosis of choledocholithiasis is not excluded by the absence of sonographic demonstration of a stone. [182]

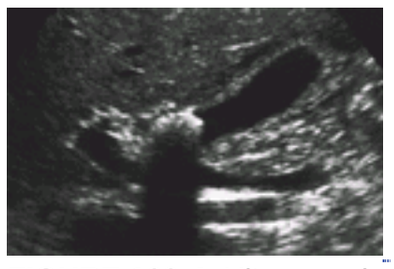

FIGURE 100-4. Ultrasound scan showing gallstones. The stones generate high-amplitude echoes and are large and dense enough to constitute substantial acoustic impedance in the path of the ultrasound beam. Hence, there is a void behind the stone, known as the postacoustic shadow. Some edema of the gallbladder wall is also seen, suggesting acute cholecystitis.

Hepatobiliary scintigraphy can confirm or exclude the diagnosis of acute cholecystitis with a high degree of sensitivity and specificity. After a 2- to 4-hour fast, the patient is given an intravenous injection of a technetium 99 (^{99m}Tc)-labeled iminodiacetic acid derivative (IDA agent), which is excreted into the bile ducts and is sequentially imaged under a gamma camera. Several ^{99m}Tc-IDA compounds have been developed and differ with respect to their degree of hepatic uptake and time to reach a peak concentration in bile. In a normal study, images of the gallbladder, common bile duct, and small bowel appear by 30 to 45 minutes. A normal ^{99m}Tc-IDA scan virtually rules out the diagnosis of acute cholecystitis in a patient who presents with abdominal pain. Failure to visualize the gallbladder by 90 minutes despite adequate views of the liver, common bile duct, and small bowel strongly suggests acute obstructive disease. False positives can result from either nonfasting or prolonged fasting states. Chronic cholecystitis is another cause of false-positive results. Delayed repeat scanning after 4 or more hours decreases the false-positive rate and provides sensitivity and specificity values of approximately 97% and 90%, respectively. Administration of low-dose morphine at 1 hour may obviate the need for delayed repeat scans. Morphine may be used as an adjunct to scintigraphy, as it causes contraction of the sphincter of Oddi, increasing common bile duct pressure and enhancing filling of the cystic duct. Hepatobiliary scintigraphy cannot provide anatomic information and cannot directly identify gallstones. Oral cholecystography is seldom used nowadays as a primary diagnostic test for the detection of gallstones or acute cholecystitis. The study involves the administration of

oral contrast tablets such as iopanoic acid the night before. The contrast is absorbed from the small bowel, conjugated and excreted by the liver, and concentrated within the gallbladder. The following morning, abdominal x-rays are obtained. Extrabiliary conditions such as small bowel disease, hepatic dysfunction, or prolonged fasting can all lead to gallbladder nonvisualization. [183], [184] Inadequate gallbladder visualization can occur in 15% to 50% of patients after a single dose. The chief advantage of oral cholecystography over ultrasound is its ability to assess the patency of the cystic duct and gallbladder function, which may be useful if medical therapy of gallstones is being considered. Moreover, the size, number, and degree of calcification of stones can be accurately determined. CT can also be used in the detection of cholelithiasis. Although superior to ultrasound in other clinical situations, CT is not as reliable in diagnosing gallbladder stones. However, CT is useful in demonstrating dilated bile ducts and mass lesions and can be considered the test of choice if clinical suspicion of a tumor (e.g., pancreatic cancer) obstructing the common bile duct is strong. When a more definitive view of the biliary system is needed, cholangiography should be performed by endoscopic retrograde cholangiopancreatography (ERCP) or through a percutaneous transhepatic cholangiography (PTC) (Fig. 100-5). The selection of either approach depends on the expertise at a particular institution and on the level and nature of the suspected lesion. In general, ERCP is used to demonstrate the lower limit of an obstruction and has the advantage of the ability to sample tissue. PTC demonstrates the upper limit of an obstructive lesion or a proximal obstruction better. Both tests have a risk of introducing infection in the presence of obstruction.

FIGURE 100-5. Endoscopic retrograde cholangiogram shows a stone within the common bile duct and a small stone or debris in the cystic duct. (Courtesy of Scott Schulte, M.D.)

Magnetic resonance (MR) cholangiography, a promising new technique for imaging the biliary tree, has been shown to be over 90% sensitive and specific for the diagnosis of bile duct stones, when compared to ERCP. [185], [186] and [187] It also has the advantage of being noninvasive and potentially allowing diagnostic imaging without the risks of PTC or ERCP. Helical CT has also been studied, and found to have sensitivity and specificity of 80% to 85%. [188], [189] Addition of an oral contrast agent for CT cholangiography may provide sensitivities and specificities comparable to MR cholangiography. [190] With the availability of many tests, the future challenge to the clinician will be to avoid unnecessary or redundant investigations.

CLINICAL COURSE AND COMPLICATIONS

Natural History of Asymptomatic Gallstones

It is estimated that 60% to 80% of all gallstones are asymptomatic at a given time. [191], [192] and [193] The rate with which gallstones develop varies. In patients on TPN or obese persons with rapid weight loss, the interval can be weeks. In Pima Indians, the progression from cholesterol supersaturated bile to formation of gallstones occurs in 5 to 10 years. [194] When gallstones do form, the risk factors for developing symptoms are unknown. However, the rate at which stones give rise to symptoms and complications is relatively small. In contrast, once gallstones start to cause biliary-specific symptoms, the risk of continuing problems is relatively high.

Studies by Wenckert and Robertson [195] and Lund [196] followed a total of 1307 patients with gallstones as long as 20 years and concluded that 50% of patients remained asymptomatic, 30% had biliary colic, and 20% had complications. Some have cited such data as indicating prophylactic cholecystectomy for all patients with gallstones, regardless of the severity of symptoms. However, the patients in these studies, although asymptomatic at the time of entry to the follow-up study, had previously been hospitalized for symptoms related to gallbladder disease.

Gracie and colleagues [197] followed a cohort of 123 asymptomatic persons with gallstones found by oral cholecystography. These 110 men and 13 women (average age, 54 years) were followed for 11 to 24 years. New-onset biliary colic developed at a rate of 2% per year for the first 5 years, but the cumulative incidence over time plateaued, such that the total incidence was 15% at 10 years and 18% at 15 and 20 years. Three patients (2%) developed complications. All were preceded by repeated attacks of biliary colic and all had uneventful cholecystectomies. No deaths related to gallbladder disease occurred in this cohort.

The Rome Group for Epidemiology and Prevention of Cholelithiasis (GREPCO) study has reported follow-up of 161 patients with gallstones. [198] For patients who were initially asymptomatic, biliary colic developed in 11.9% at 2 years, 16.5% at 4 years, and 25.8% at 10 years. The subjects were also followed for development of complications of gallstones, including cholecystitis, pancreatitis, and biliary obstruction. After 10 years, 3% of those who were initially asymptomatic developed complications. One patient of this group developed an adenocarcinoma of the gallbladder.

A 20-year population-based survey of mortality in Pima Indians with and without gallstones was reported. [199] The overall age- and sex-adjusted death rate was higher in those with a history of gallstones, with a ratio of 1:9. The death rate attributed to malignancies was 6.6 times higher in those with gallstones. Of the 20 fatal malignancies in patients with gallstones, 11 were cancers of the gastrointestinal tract, of which 6 were malignancies of the gallbladder or bile ducts.

These studies highlight the question of whether a link between gallstones and malignancies exists. A statistical association between colon cancer and gallstones has been reported, although this may represent selection bias because patients with colon cancer are more likely to have their gallbladders evaluated. [200] Increased rates of gallbladder carcinoma or carcinoma of the extrahepatic bile ducts in patients with gallstones have been found in an autopsy series and in a case-control study. [201], [202] This issue deserves further study.

Natural History of Symptomatic Gallstones

Once an episode of biliary colic has occurred, there is a high risk of repeated attacks of pain. Cohort studies following symptomatic gallstone patients indicate that 58% to 72% of patients have ongoing symptoms and complications. [196], [203] More than 90% of complications, such as cholecystitis, cholangitis, and pancreatitis, are preceded by attacks of pain. The most serious complications are gangrene and perforation of the gallbladder, occurring in about 10% of cases of acute cholecystitis.

When the gallbladder perforates, the outcome depends on the anatomic relation with the neighboring structures. It can localize to form an abscess; it can be a free perforation with peritonitis; or it can communicate with another hollow viscus as a fistula. A localized perforation with confined spillage sealed off by omentum and adherent adjacent viscera is the most common form of perforation. A pericholecystic abscess is formed. This should be suspected when acute cholecystitis is slow to resolve, especially with a second episode of fever, right-sided abdominal pain, or the appearance of a right upper abdominal mass. Ultrasonography and CT may show the pericholecystic abscess, but this is not dependable. If the degree of clinical suspicion is high, the diagnosis should be confirmed at surgery. This is especially so in older patients or patients on long-term steroid treatment in whom fever and the inflammatory response may be minimal.

In 1% to 2% of perforations, especially in the fundus of the gallbladder, free perforation occurs and carries a mortality rate of at least 30%. Bile and purulent peritonitis are almost always associated with persistent or progressive abdominal pain. When the suspicion is raised, no test can establish the diagnosis without further delaying intervention and causing additional tissue injury. Antibiotics should be given and emergency surgery performed.

When the gallbladder perforates into the adjacent intestine, an attack of acute cholecystitis often subsides as the inflamed organ is decompressed. In descending

order of frequency, the duodenum, hepatic flexure of the colon, stomach, and jejunum are the sites of the cholecystoenteric fistula. If the gallstones are completely discharged and are small enough to pass rectally, an uncomplicated cholecystoenteric fistula results. However, if stones are still present in the gallbladder or in the common bile duct, chronic symptoms may arise.

When a fistula develops and the gallstone is too large to traverse the intestine, it causes obstruction. In such cases, the diameter of the stone is greater than 2.5 cm and the site of obstruction is usually in the small intestine, most commonly at the ileocecal valve. Gallstone ileus sometimes has a prior history of acute cholecystitis, but most of the stones erode slowly through the gallbladder, and the symptoms may be minimal, especially in older patients. Gallstone ileus should always be considered in an older patient with intestinal obstruction. [204], [205] Plain abdominal x-ray films may show air in the biliary tree, and barium studies often reveal the site of communication. The gallbladder does not opacify with oral cholecystography. Ultrasound scans can detect air in the biliary tree but not the site of the fistula. CT scans are less useful in detecting gallstones and fistulae, although they may show air in the biliary tree.

TREATMENT

Because gallstones are common and often asymptomatic, the treatment of gallstones should consider the patient's clinical presentation (Fig. 100-6). For asymptomatic gallstones, before active treatment is instituted, the following questions should be considered: What is the rate at which new-onset symptoms emerge? If symptoms develop, what is the risk of these evolving to complications, and what is the risk of morbidity and mortality from such complications? What is the risk of prophylactic treatment? What are the short- and long-term costs of prophylactic treatment compared with no or delayed treatment?

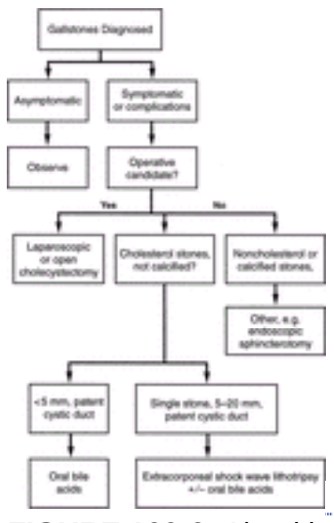

FIGURE 100-6. Algorithm for management of biliary sludge and gallstones.

The fractional and cumulative risk of developing symptoms is not great (2%–5% per year). Complications develop after the emergence of symptoms. Risk-benefit analyses of elective cholecystectomy have shown no advantage to aggressive prophylactic cholecystectomy for asymptomatic gallstone patients. Once complications develop, cholecystectomy is a safe procedure with a mortality rate of 0.1% to 0.2% for open procedures. [206], [207] The surgical mortality rate of 0.9% to 1.8% is higher for patients older than 60 years of age. [208], [209] The mortality rate for laparoscopic cholecystectomy appears comparable. [210] With common bile duct exploration, the mortality rate is approximately two to three times higher.

Generally, the indications for cholecystectomy should not change, and asymptomatic patients do not need to undergo surgery despite the widespread availability of laparoscopic cholecystectomy. However, in certain subsets of patients, it may be prudent to make exceptions and proceed with a prophylactic cholecystectomy. These groups include children, patients with sickle cell disease, and the morbidly obese. [211], [212] In the latter group, a cholecystectomy is recommended if the patient undergoes bariatric surgery such as a gastric bypass. [213] Patients who are immunosuppressed on a long-term basis may represent another group, as signs of acute cholecystitis may be "masked." A calcified or "porcelain" gallbladder is associated with a higher risk of cancer. [214] It is also agreed that patients with choledocholithiasis, even if asymptomatic, should undergo surgery. This is reinforced by postmortem studies, which showed, even in asymptomatic patients, ductal stones contributed significantly to death. [215], [216]

Patients with diabetes mellitus are a controversial group. It has been stated that diabetics have a higher risk of complications, morbidity, and mortality from gallstones. However, they may have an increased operative risk with elective and emergency cholecystectomy, largely because of concomitant cardiovascular disease. [217], [218] The issue of prophylactic cholecystectomy in diabetics continues to be debated.

Symptomatic gallstones should be treated. Options include surgery, dissolution, or fragmentation of symptomatic gallstones (Table 100-4).

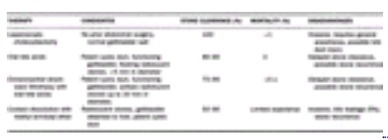

TABLE 100-4 Therapeutic Options for Symptomatic Gallbladder Stones

Surgical Treatment

Cholecystectomy is the only definitive treatment. Elective cholecystectomy is usually simple, safe, and curative; is indicated for most patients with symptoms; and is the first choice when complications ensue. Nonsurgical modalities of treatment are less favored with the advent of laparoscopic cholecystectomy.

Laparoscopic Cholecystectomy

In 1901, Georg Kelling reported laparoscopic examination of the abdominal cavity using a cystoscope in a dog. [219] Technical advances, such as the Veress needle used to induce pneumoperitoneum, was introduced in 1928 and is still in use today. [220] Laparoscopic techniques were first routinely incorporated into surgical practice by gynecologists in the 1960s. With the introduction of videolaparoscopy in the 1980s, general surgeons began to use the technique to perform cholecystectomies. [221]

Laparoscopic cholecystectomy was first performed in France in 1987. [222] The procedure has since gained remarkably rapid widespread use and acceptance in Europe and the United States, becoming the standard of care for the treatment of symptomatic cholelithiasis and acute or chronic cholecystitis. In fact, the overall rate of cholecystectomy increased rapidly with the introduction of the laparoscopic approach, possibly due to a wider patient and physician acceptance of this method of treating gallstones. [223] There is also some evidence that the indications for cholecystectomy have broadened since introduction of the laparoscopic approach. [224] This rapid increase, however, has shown signs of leveling off or diminishing. [225] Due in part to the rapid acceptance of the procedure by patients and physicians, there have been few randomized, controlled trials comparing laparoscopic with open cholecystectomy. [226], [227] However, early publications compiling results for more than 5000 patients have documented its safety and efficacy. [228] Retrospective comparative studies have shown that laparoscopic cholecystectomy produced fewer complications, shorter hospital stays, more rapid returns to normal activities, and minimal use of postoperative analgesia. [229], [230] and [231] Additionally, overall mortality

appears to be reduced. [225]

The technique involves general anesthesia with endotracheal intubation. A pneumoperitoneum is established, and the laparoscope with video camera attached is inserted through an umbilical incision. Accessory trocars and cannulas are inserted through separate sites. The gallbladder, cystic duct, and artery are identified. An intraoperative cholangiogram can be performed by cannulation of the cystic duct. This allows for the detection of common bile duct stones and delineation of the ductal anatomy. The blood supply to the gallbladder is identified and controlled, and then the cystic duct is ligated. The gallbladder is dissected from the underlying liver bed, using electrocautery or laser energy. The gallbladder is removed through the incision site. Stones may need to be mechanically crushed within the gallbladder before removal. Stones spilled into the peritoneal cavity are retrieved. [232]

The primary indication for laparoscopic cholecystectomy is identical to that for open cholecystectomy: symptomatic cholelithiasis. Absolute contraindications include an inability to tolerate general anesthesia and uncontrolled coagulopathy. Relative contraindications include scarring or inflammation, precluding access to the gallbladder using the transperitoneal approach, and diffuse peritonitis. As surgeons have accumulated more experience, the technique has been applied to a variety of patients. The procedure has been successfully performed in anticoagulated patients, obese patients, children, and pregnant women prior to the third trimester. [233], [234], [235] and [236]

Examination of the results of large series illustrates the pertinent aspects of the procedure. A representative analysis of 1518 laparoscopic cholecystectomies performed at academic and private hospitals has been published. [237] In 4.7% of the patients, the procedure was converted to open cholecystectomy, most often because of difficulties in identifying anatomy due to inflammation or bile duct injury. Of the 1518 patients in this series, 96.5% had ultrasound examinations before surgery to document cholelithiasis, and preoperative cholangiography was performed in 4.2% of patients. Some surgeons routinely used intraoperative cholangiography and others used it selectively or not at all. Intraoperative cholangiograms were obtained in 29.3% of patients, with a failure rate of 23.3%.

Complications, including delayed discharge, undesired change in therapy, readmission, biliary injury or leakage, infection, hemorrhage, or conversion to open cholecystectomy, occurred in 5.1% of cases. Superficial wound infection was the most common complication. Injuries to the common bile duct or the hepatic duct occurred in 0.5%. Similar to other groups, the rate of bile duct injury was higher early in a given surgeon's experience with the procedure, illustrating the steep learning curve associated with the procedure. The mean hospital stay was 1.2 days, with a range of 6 hours to 30 days. One patient died. [210], [228]

Several aspects of laparoscopic cholecystectomy are currently being investigated. This includes debates about the selection of patients for preoperative or intraoperative cholangiography. Some argue for routine operative cholangiography to look for unsuspected common bile duct stones and to identify ductal anatomy, anomalies, and injuries. Intraoperative cholangiography is usually safe and successful. [238] Others argue for a more selective approach. Preoperative clinical and ultrasonographic assessment can be used to predict the likelihood of common duct stones. A history of jaundice or pancreatitis, dilated common bile ducts on ultrasound, and elevated levels of bilirubin, aminotransferases, or alkaline phosphatase can be used as an indication for preoperative ERCP with possible sphincterotomy or intraoperative cholangiogram. [229], [239] Preoperative ERCP and sphincterotomy does not interfere with laparoscopic cholecystectomy or increase the operative complication rate. [240] Statistical models to predict which patients have choledocholithiasis, and therefore might benefit from preoperative ERCP or intraoperative cholangiogram, have been developed. [241], [242]

If unsuspected choledocholithiasis is discovered intraoperatively, several options are available. The surgeon may convert to an open procedure and carry out a common bile duct exploration with transcystic duct retrieval of stones. Bile duct exploration can also be carried out using laparoscopic or endoscopic techniques. [243] Another common option is postoperative ERCP with sphincterotomy. In experienced hands, laparoscopic bile duct exploration appears to have high success rates (75%–91%). [244], [245] It also has the potential advantages of using only one procedure for treatment of these patients and avoiding the delay inherent in postoperative ERCP. For those patients in whom the procedure is not successful, postoperative ERCP with sphincterotomy remains an option. The modalities of therapy in this area are evolving rapidly, and it is not yet clear which of these techniques will ultimately prove most efficacious and cost-effective. The perioperative approach to common bile duct stones should depend on local availability and expertise.

A serious direct complication of laparoscopic cholecystectomy is injury to the common bile or hepatic ducts. This may result from misidentification of the common duct for the cystic duct, resection of part of the common and hepatic ducts, or associated right hepatic arterial injury. The estimated incidence of bile duct injury varies from 0.3% to 2.7%. [246] In contrast, biliary tract injuries are estimated to occur in 0.25% to 0.5% of open cholecystectomies. [247] The major risk factor for bile duct injury is the experience of the surgeon. Bile duct injuries appear to be much more common early in a surgeon's experience with the technique. Other risk factors appear to be the presence of aberrant biliary tree anatomy and the presence of local acute or chronic inflammation. [246] There does not appear to be a correlation between the incidence of bile duct injury and the frequency of operative cholangiography. [210] Bile leaks may also develop due to displacement of the operative clips on the cystic duct.

Bile duct injury leads to two clinical manifestations: bile leakage into the peritoneum, with resulting abdominal pain and bile peritonitis, and biliary obstruction due to partial or complete hepatic or common duct obstruction from ductal ligation or stricture. Patients can present 3 to 7 days after surgery with fever, abdominal pain, anorexia, ileus, ascites, nausea, or jaundice. Patients with late-onset stricture may present months later with jaundice. Biliary scintigraphy using ^{99m}Tc-IDA can be used to diagnose bile leakage and may show activity in the right paracolic gutter. [248] ERCP with stent placement or sphincterotomy can be used both diagnostically and therapeutically. ERCP appears to be the treatment of choice for less severe lesions, such as minor lacerations of the common bile duct. [246], [249] ERCP may also be used to treat bile leakage due to displaced cystic duct clips. Lesions in the proximal biliary tree may be more amenable to percutaneous transhepatic approaches. [250] Surgical repair may be necessary in some patients, such as those with transected common bile ducts. [246], [250], [251] and [252] Intra-abdominal bile collections may need to be drained percutaneously.

These results bring up certain issues that merit further elaboration. The management of patients with acute cholecystitis, and in particular, the timing of surgery has been controversial. Although laparoscopic cholecystectomy has been performed successfully in acute cholecystitis, this group of patients has a higher incidence of common bile duct stones, and the procedure is technically more difficult and often lasts longer. [253] In this setting, intraoperative cholangiograms may be more difficult to obtain and bile duct injuries more common. Two randomized trials comparing early (<3 days) versus delayed (4–6 weeks) surgery for acute cholecystitis showed no benefit to delaying surgery. [254], [255] Although patients in the early surgery group had longer operating times, they had similar rates of conversion to open cholecystectomy and shorter hospital stays. Thus, early operation for acute cholecystitis may be beneficial.

If the operative field is not adequately visualized or variant anatomy is encountered, conversion to an open cholecystectomy should be undertaken. Conversion to an open procedure should not be considered a complication. In most series, conversion rates are higher with emergency operations. Reported rates range from 1.5% to 15%, with most reporting rates around 5%. [210] With experience, the operative conversion rate appears to fall.

The cost-effectiveness of laparoscopy has been examined. Although the direct operating room and recovery room costs are higher for laparoscopic cholecystectomy, the shortened length of hospital stay leads to a net savings. [256], [257] More rapid return to normal activity may lead to indirect cost savings. Not all such studies have demonstrated a cost savings, however. [229] In fact, with the higher rate of cholecystectomy in the laparoscopic era, the costs in the United States of treating gallstone disease may actually increase.

Although laparoscopic cholecystectomy has replaced open cholecystectomy as the standard of care for gallstones, many patients still present with complications of gallstone disease. Glasgow and colleagues [258] found that 1.3 per 1000 people underwent cholecystectomy for symptomatic gallstone disease in California in 1996. Of these, 56% had biliary colic, and the remainder more severe complications such as acute cholecystitis, gallstone pancreatitis, or cholangitis. Patients presenting with biliary colic were more likely to undergo laparoscopic surgery electively, while those with the more severe complications were more likely to require an emergency or open procedure. In many patients, surgery was significantly delayed after the development of initial symptoms. Patients being treated for complicated gallstone disease had significantly longer hospital stay and costs.

Many studies suggest that laparoscopic cholecystectomy has become common and that indications for the procedure are widening. [224] Cholecystectomy may be performed in some whose symptoms are not clearly biliary in origin. In contrast, some patients with symptomatic gallstones may not receive surgery in a timely fashion. Thus, there is evidence for both underuse and overuse of laparoscopic cholecystectomy. Future work might center upon clarification of the appropriate indications for surgery, as well as ensuring timely recognition and treatment of gallstone symptoms.

Endoscopic Treatment

Stones may be removed from the common bile duct by means of ERCP with sphincterotomy. In the postoperative period, endoscopic and radiographic techniques may be used to extract stones through a T-tube tract. Stones may be extracted or fragmented with the help of a choledochoscope. In all of these modalities, the gallbladder is left in situ, allowing the possibility of recurrent stone formation. Some small studies have shown that patients undergoing sphincterotomy alone for treatment of their gallstone disease have a much higher rate of future biliary complications than patients undergoing open cholecystectomy. [259]

Medical Dissolution

With the advent of laparoscopic cholecystectomy, medical therapy for gallstone disease has fallen somewhat out of favor. These therapies are now limited to patients who are not candidates for laparoscopic or open cholecystectomy.

Oral Bile Acid Litholysis Two bile acids, chenodeoxycholic acid (3a,7a-dihydroxycholanoic acid; CDCA) and its 7ß epimer (3a,7ß-dihydroxycholanoic acid), ursodeoxycholic acid (UDCA), have been used for gallstone dissolution. [260], [261], [262], [263], [264] and [265] These two bile acids induce the secretion of undersaturated hepatic bile, and do not increase the absolute amount of bile acids secreted. The biologic effects of administration of CDCA and UDCA include suppression of de novo hepatic cholesterol synthesis by inhibiting hydroxymethylglutaryl-CoA reductase and enhancement of the activity of 7a-hydroxylase, leading to an increase in bile acid synthesis. [266], [267] Intestinal absorption or reabsorption of cholesterol is down-regulated. [268] Because CDCA has more associated side effects and a lower success rate in dissolving gallstones, UDCA, the major bile acid of the Himalayan black bear, is now the bile acid of choice for gallstone dissolution. UDCA is a remarkably safe compound. The usual dose is 8 to 12 mg/kg/d. The proper selection of patients is a main factor affecting the success of bile acid therapy. The selection criteria include gallstones predominantly consisting of cholesterol; pigment stones or calcified stones are excluded. Small stones that are buoyant in an oral cholecystogram examination have a 90% chance of dissolution. Stones should have a large surface area in relation to the mass, and those with a diameter larger than 1.5 cm are not suitable. Oral cholecystogram or hepatobiliary scanning can be used to document the patency of the cystic duct.
Contact Solvents Solvents can be delivered directly to dissolve gallstones when a catheter can be used to access them. Several cholesterol solvents are available, although none is a simple, safe, and effective method of treating gallstones. The most commonly used agent is methyl- *tert*-butyl ether (MTBE). To dissolve gallstones, the gallbladder is punctured by percutaneous transhepatic approach. A catheter is inserted into the lumen of the gallbladder, and MTBE is introduced and withdrawn from the catheter. [269], [270] Dissolution can be achieved within a few hours. [271] In well-selected patients in specialized centers, this method is effective (90% success rate) and safe. Complications include those caused by the percutaneous puncture procedure and side effects if the MTBE drains into the duodenum (e.g., hemolytic anemia, erosive, or hemorrhagic duodenitis, aspiration pneumonia, somnolence). Small cholesterol gallstones without a thick calcified rim are suitable for this therapy. Calcified or pigment gallstones, cirrhosis of the liver with portal hypertension, and coagulopathy are contraindications.
Extracorporeal Shock Wave Lithotripsy Energy generated externally can be directed to fragment gallstones. This method of treatment combines two approaches: oral bile acid therapy and fragmentation of gallstones. Extracorporeal shock wave lithotripsy (ESWL) requires accurate localization of the gallstones within the gallbladder, usually achieved by three-dimensional ultrasonography. This spatial localization is fed into a computer, which directs the energy to focus sharply on the gallstone. The source of energy varies with different methods. The energy waves, traveling in phases of compression and rarefaction, converge on the gallstone without causing tissue damage because no acoustic impedance occurs. Acoustic impedance and differential tensile forces develop, causing surface disintegration or cavitation. Fragmentation of a gallstone into small fragments (<3 mm) results from successful ESWL. These fragments can be discharged into the duodenum spontaneously or can be rapidly dissolved by UDCA. Depending on the type of lithotriptor, it takes one to several sessions of ESWL before optimal fragmentation can be achieved. UDCA, usually given before and concurrently with ESWL, is continued for several months until stones have completely disappeared. [272] If patients are properly selected, ESWL is effective and safe. Outpatient treatment has been reported. [273] Selection criteria of patients for ESWL are similar to those listed for oral bile acid therapy. Early reports indicated a promising success rate of more than 90% with single, small cholesterol stones. [274], [275] However, larger stones and multiple stones have a lower fragmentation rate of 34% to 71%. [276] The best results are obtained with small, single stones. [277] About 20% of patients developed biliary colic for several weeks after ESWL (25% of these patients required cholecystectomy), and 1% had pancreatitis. [274], [275] and [276] Other side effects include local discomfort, petechiae or bruising, transient noncontraction of the gallbladder, and microscopic hematuria. ESWL is currently not used in the United States for uncomplicated gallbladder stones. However, problematic common bile duct stones not cleared by ERCP with sphincterotomy may be amenable to a combined approach using ESWL. [278]

Rational Approach to Treating Symptomatic Gallstones

Surgery remains the definitive curative method of gallstone treatment. The advent of laparoscopic cholecystectomy and its widespread acceptance has revolutionized surgical treatment of gallstones. Laparoscopic cholecystectomy is now regarded as the treatment of choice for symptomatic gallstones. For patients unable or unwilling to pursue this route, nonsurgical methods such as oral bile acid therapy or lithotripsy are available (Fig. 100-6).

When stones are removed and the gallbladder is left behind, stones may recur. With oral bile acid therapy, the cumulative recurrence rate is about 15% in 2 years and 50% in 10 years. [279], [280] Patients with multiple stones have higher recurrence rates than those who have solitary stones. The long-term recurrence rate after ESWL is also high, 15% at 1 year and 60% at 5.5 years. [46] One of the major challenges of nonsurgical treatment modalities of gallstones is preventing recurrence. It is not generally agreed whether long-term, low-dose UDCA can protect against recurrence. Cholecystectomy is curative for gallbladder stones.

To our knowledge, no formal cost comparison has been made between surgical and nonsurgical treatment. Surgery has a high immediate cost and low long-term cost. Nonsurgical means of treatment may have a low immediate cost but high long-term cost because of recurrent or persistent disease. In selecting patients for nonsurgical treatment, a diagnosis of gallstones by ultrasonography is insufficient. The physician must also determine gallstone composition and gallbladder function. Regular monitoring by ultrasonography during nonsurgical treatment may be necessary. These additional tests increase cost.

In approaching a patient with symptomatic gallstones, an elective cholecystectomy remains the gold standard. However, for those who cannot or will not have surgery, there are other alternatives. One of the nonsurgical methods may be selected if the patient satisfies all the criteria and if expertise is available.

ACALCULOUS CHOLECYSTITIS

Acute cholecystitis can present as an acalculous disorder. Acalculous cholecystitis represents 5% to 10% of the patients with acute cholecystitis and may be increasing in frequency. [281], [282] This entity is distinct from gallstone-related disease in that it usually occurs in the setting of major surgery, critical illness, trauma, or burn-related injury. The patients tend to be predominantly male and older than 50 years of age. Many of them are on TPN, and bile inspissation or sludge formation may occur. Hypotension and sympathetic vasoconstriction may contribute, predisposing the patient to ischemic injury as well. Extensive small vessel occlusion has been shown in cholecystectomy specimens from these patients. [283] Another postulated risk factor is mechanical ventilation with positive end-expiratory pressure (PEEP). [281] PEEP may decrease portal perfusion and increase resistance in the common bile duct, with subsequent stasis. [284], [285] The pathogenesis of acalculous cholecystitis probably involves some combination of bile stasis, chemical inflammation, and mucosal ischemia. Gallbladder ischemia may be the only predisposing factor found in many cases. Bacterial infection, usually by gram-negative bacteria and anaerobes, appears to occur secondarily.

P>In acalculous cholecystitis, complications such as gallbladder gangrene and perforation, emphysematous cholecystitis, or empyema, may develop more rapidly than in calculous cholecystitis. [281] One retrospective study revealed that 70% of patients had gangrene, empyema, or perforation of the gallbladder evident at the time of surgical exploration. [286] This increased complication rate occurring in the setting of advanced age and other illnesses contributes to the higher mortality rate associated with this disorder. For these reasons, it is important to suspect and pursue an early diagnosis. It should be suspected in patients who are critically ill with signs of sepsis, but with no obvious source. Findings on physical examination and laboratory studies are not necessarily helpful. Potential diagnostic modalities include ultrasonography, CT, hepatobiliary scintigraphy, and laparoscopy.

Ultrasonography is usually the initial modality chosen. Criteria for acalculous cholecystitis include a thickened gallbladder wall (>4 mm), pericholecystic fluid, subserosal edema without ascites, intramural gas, sloughed mucosal membranes, or a sonographic Murphy sign. Reported sensitivity rates range from 67% to 92%, with specificity over 90%. [287] Experience with CT is also favorable. Diagnostic criteria are similar to ultrasonography. Its reported sensitivity and specificity are in excess of 95% when compared with findings at the time of surgery. [287] Another advantage of CT is the ability to look for other intra-abdominal pathology. Results with hepatobiliary scintigraphy are variable, with reported sensitivity rates ranging from 68% to 91% and frequent false-positive results. Adjunctive use of morphine to enhance gallbladder filling may improve the accuracy of this test. [288] Patients in whom the diagnosis is uncertain or with generalized peritonitis may benefit from

diagnostic laparoscopy or exploratory laparotomy. In small series of patients, diagnostic laparoscopy appears to be accurate and safe. [289], [290] Some suggest empiric percutaneous cholecystostomy if the diagnosis is suspected but cannot be confirmed with other modalities. [291]

Rare causes of acute cholecystitis include specific infections, such as *Salmonella* or *Candida* infection. [292], [293] Cytomegalovirus and *Cryptosporidium* can infect the biliary system and produce cholecystitis and cholangitis in severely immunocompromised patients, such as those with acquired immunodeficiency syndrome (AIDS) or after marrow transplantation. [294], [295] In patients with AIDS, infection with microsporidia such as *Enterocytozoon bieneusi, Pneumocystis carinii,* and *Isospora belli* have been documented. [296] However, acute calculous cholecystitis may also occur in these patients. No specific symptoms can differentiate these two, and patients may have coexisting gallstones and opportunistic infection. In these patients, acalculous cholecystitis probably represents part of the spectrum of AIDS-related cholangiopathy.

Once a diagnosis of acalculous cholecystitis is made, the gallbladder must be drained or removed. Choosing the treatment modality may be difficult as patients often have severe co-morbid conditions. Options now include percutaneous or surgical cholecystostomy, surgical cholecystectomy, or endoscopic transpapillary cholecystotomy. [291], [297], [298] and [299] Percutaneous cholecystostomy has been gaining favor recently. Its advantages include the use of local anesthesia and the ability to be performed at the bedside. Also, percutaneous cholecystostomy can be the definitive therapy in acalculous cholecystitis. The overall success rate is 63% to 94% with a low complication rate of approximately 10%. [300] Complications include liver laceration, hemorrhage, infection, hemobilia, bile leak, bile peritonitis, and catheter dislodgment. Other disadvantages of cholecystostomy include a limited ability to look for other causes of sepsis and potentially inadequate drainage in cases of pericholecystic abscess, gangrene, or gallbladder perforation. Surgical cholecystostomy or cholecystectomy are the traditional therapies, but patients may be poor operative candidates. Endoscopic transpapillary drainage has been reported in small series and needs to be further studied before it can be recommended for general use.

RECURRENT PYOGENIC CHOLANGITIS

Recurrent pyogenic cholangitis, also known as Oriental cholangiohepatitis or hepatolithiasis, is a syndrome primarily seen in southeast Asian populations such as Taiwan, China, Japan, and Korea. Patients present clinically with recurrent attacks of abdominal pain, fever, and jaundice. Previous reports found this entity to be among the leading causes of acute abdominal emergencies in Hong Kong. [301] The prevalence of intrahepatic stones is likely decreasing in many parts of Asia. [302] Primary intrahepatic stones are generally calcium bilirubinate or mixed stones. Intrahepatic calcium bilirubinate stones contain less bilirubin and more cholesterol than pigment stones in the extrahepatic bile ducts. [303] The pathogenesis of these stones is complex, and likely involves a combination of bile infection, malnutrition or dietary factors, biliary stasis, and possibly parasitic infestation. It is generally believed that bacterially mediated hydrolysis of bilirubin by ß-glucuronidase is crucial to formation of pigment stones. This leads to formation of unconjugated bilirubin, which can precipitate as calcium bilirubinate. The ß-glucuronidase enzyme often originates from *E. coli* or other bacteria in the biliary tree. There has been a debate about whether bacterial infection precedes or is a sequela of stone formation. However, in at least some patients with bilirubinate stones, bile infection precedes stone formation. [148] ß-Glucuronidase can be inhibited by glucarolactone, whose levels are decreased in patients with a low protein and low fat diet. [304] It has therefore been suggested that dietary factors contribute to formation of intrahepatic stones. The role of infection by parasites such as *Ascaris lumbricoides* or *Clonorchis sinensis* has been controversial. The parasites or their ova have been found as nidi for intrahepatic stones in earlier studies. [303] However, infection with these parasites is widespread, but recurrent pyogenic cholangitis is uncommon outside southeast Asia. Even within endemic countries, there is not a direct correlation between the prevalence of parasitic infection and the occurrence of this syndrome. These lines of evidence suggest that factors in addition to parasites are important for the development of these stones.

Recurrent pyogenic cholangitis frequently presents in patients under 50 years of age. Men and women are affected equally. It is more common with lower socioeconomic status and malnutrition. Symptoms occur when the stones cause partial or complete biliary obstruction, either with or without associated infection. Many patients will present with epigastric or right upper quadrant pain or discomfort. Acute cholangitis, with fever, right upper quadrant pain, and jaundice, is also common. Physical examination may show right upper quadrant tenderness, hepatomegaly, or an enlarged gallbladder. Laboratory results are generally consistent with the degree of biliary obstruction and infection. In advanced cases, biliary cirrhosis and its sequelae may develop.

Recurrent pyogenic cholangitis is often diagnosed clinically in endemic areas. Delineating the extent and location of stones and strictures is essential to planning further treatment. Abdominal plain films are rarely useful as the stones are usually radiolucent. CT scan may be useful to assess ductal dilation. Percutaneous or endoscopic retrograde cholangiography may provide the most accurate information. The left intrahepatic duct is preferentially involved. Frequent findings on cholangiography include stones, biliary strictures, and ductal dilation. The intrahepatic ducts may also appear “pruned” with decreased arborization. [305] Histologically, increased fibrous tissue around the portal tracts may be found with bile ductular proliferation.

Surgery is the primary method of treatment for this syndrome. The role of endoscopic treatment is relatively limited because of the difficulty in treating intrahepatic strictures and stones. Surgery should aim to remove as many stones as possible and to achieve adequate drainage of obstructed ducts. [305] In rare cases, hepatic resection can be undertaken if the stones and strictures are localized. If strictures cannot be managed operatively, many surgeons will create a Roux-en-choledochojejunostomy with the end of the loop left in the subcutaneous abdominal wall tissue. [306] This allows easier access for later endoscopic or interventional radiologic treatment.

Interventional radiology techniques are also important in management of recurrent pyogenic cholangitis. If percutaneous transhepatic drainage can be established, cholangioscopy can be used to direct attempts at electrohydraulic or laser lithotripsy, stricture dilation, and stone clearance. [307] Solvents such as MTBE are ineffective, because the stones are not cholesterol stones. Surgical and interventional radiologic techniques to achieve adequate drainage and stone clearance will be required by many patients.

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CHAPTER 101

Jayant A. Talwalkar and Russell H. Wiesner

PRIMARY SCLEROSING CHOLANGITIS AND OTHER CHOLANGIOPATHIES

Primary sclerosing cholangitis (PSC) is a chronic cholestatic liver disease characterized by inflammation and fibrosis of the intrahepatic and extrahepatic bile ducts. Histological characteristics include the presence of portal tract inflammation, periductal fibrosis, and loss of small bile ducts. Disease progression is almost universal among subjects, resulting in biliary cirrhosis and hepatic failure. Immunologic and genetic alterations have been cited as factors in its pathogenesis. [1], [2] Understanding the natural history of PSC has become important because of the success of liver transplantation as an effective therapeutic modality.

EPIDEMIOLOGY

With the increasing availability of endoscopic retrograde cholangiography (ERCP), a growing body of knowledge about the clinicopathologic features of PSC has emerged. Fifty to 70% of affected individuals are of male gender, with an average age of 40 years at diagnosis. [3], [4], [5], [6], [7], [8], [9] and [10] The male predominance seen in PSC has not been observed in inflammatory bowel disease (IBD) or similar autoimmune liver diseases as primary biliary cirrhosis (PBC) and autoimmune hepatitis (AIH). However, a significant proportion of female subjects have been reported among select cohorts. [8], [9] The suggestion of a reduced survival outcome from PSC in female subjects, however, has yet to be confirmed. [9]

The close association between PSC and IBD is well established. Among northern Europeans, 70% to 80% of PSC patients have chronic ulcerative colitis (UC). [11], [12], [13] and [14] Lower prevalence rates of IBD in PSC, however, have been noted elsewhere. [15], [16] These wide variations appear related to influences not explained by clinical practice. The use of surveillance colonoscopy with mucosal biopsy assessment, although improving the detection of subclinical IBD, is nonetheless required if accurate prevalence estimates for IBD in PSC are to be determined. No meaningful difference in PSC-related survival, however, has been observed among patients with or without IBD. [9] Ulcerative pancolitis or colonic involvement alone by Crohn's disease imparts the greatest risk for PSC. [10], [11] Conversely, about 2.4% to 4% of patients with UC [10], [12], [13] and [14] and 1.4% to 3.4% with Crohn's disease [11], [14] have or develop PSC. From descriptions in the general population, an estimated prevalence rate of 13 to 140 cases of PSC per 1 million population exists. [10] The only population-based epidemiologic investigation of PSC revealed annual incidence and cross-sectional prevalence rates of 1.3 and 8.5 per 100,000 population. [17] Geographic areas outside of Norway, however, have reported PSC at lower rates with less well-defined methods. [18] The accuracy of prevalence data is limited by several factors, including the failure to identify asymptomatic patients, those with fluctuations in liver biochemical parameters such as serum bilirubin, and those without coexisting IBD. From an estimated prevalence rate of 40 to 225 cases per 100,000 United States population for UC, the prevalence of PSC is estimated between 2 and 11 cases per 100,000 population with UC.

Although a negative relationship between smoking history and UC activity is well established, [19] similar investigations in PSC have also been conducted. By comparing UC patients with age- and gender-matched controls, a lower prevalence rate of PSC (4.9%) was observed among individuals who smoked compared with nonsmoking controls (26.1%). The coexistence of UC could not support this finding. [20]

PATHOGENESIS

Immune Mediated

Immune System Alterations Immunologic and genetic alterations have been cited as factors in the pathogenesis of PSC as well as disease progression. Cellular immune abnormalities have been described focusing primarily on the significance of lymphocyte function in PSC. CD4 lymphocyte recognition of antigens found on biliary epithelia is hypothesized to initiate the immune-mediated histological damage seen in PSC. [21], [25] T-cell reactivity studies have demonstrated activation of both suppressor and cytotoxic lymphocyte populations with exposure to major histocompatibility complex (MHC) antigens in vitro. [25] Recent investigations suggest that hepatic T cells in PSC have oligoclonal restriction different from populations in peripheral blood. [22] Conflicting results exist, however, regarding the significance of altered circulating CD8 and CD4 T lymphocytes as both reductions and elevations in their ratios have been observed. [23] Increased numbers of hepatic B-cell lymphocytes reported among PSC patients with advanced histological disease is not recognized with earlier stages of disease, suggesting that cellular infiltration may be a secondary phenomena related to chronicity. [24] Historically, there have been no accepted animal models that characterize PSC. Orth and colleagues [26] described a bacteria-free rat model of sclerosing cholangitis derived from the injection of 2,4,6-trinitrobenzesulphonic acid into bile ducts. The cholangiographic pattern in these animals was consistent with PSC as well. The presence of CD3-positive T-cell infiltrates and increased bile duct expression of class II MHC molecules was also consistent with PSC observed in humans. Further studies are required to validate the model's reproducibility. A number of clinical investigations have also confirmed the presence of numerous humoral immune markers in the sera of patients with PSC. Non–organ-specific markers such as immunoglobulin G and M, antinuclear antibody (ANA), and anti–smooth muscle antibody (ASMA) are often seen in low but detectable titers. [2], [28] Their relevance in the pathogenesis of PSC may occur among patients displaying clinical features of AIH and PSC. Elevated levels of circulating immune complex molecules have also been reported in up to 80% of PSC cases and other HLA-B8–and HLA-DR3–associated immune-mediated diseases. [27], [28] Selected cross-reactive antibody species among patients with UC and PSC are not seen in cases of PSC alone, suggesting a shared mechanism for immune-mediated damage. [29] Perinuclear antineutrophil cytoplasmic antibody (pANCA) has been reported in 26% to 85% of PSC subjects and in 70% of UC patients without PSC. [30], [31] and [32] The antigenic and subsequent pathogenic role of pANCA, however, is unknown. [33] Seropositivity for ANCA in PSC has been reported with extensive involvement of the biliary tree, hepatic dysfunction, and severe cholestasis. [34]
Genetics An increased prevalence of HLA-A1, -B8, and -DR3 alleles in PSC has been described but remains nonspecific given their occurrence in other organ-specific autoimmune diseases. [35] Greater than expected frequencies of the HLA-DRw52a antigen and HLA-DRB3*0101 were initially reported among 100% of patients undergoing liver transplantation at a U.S. center. [36] Subsequent investigations have not supported the DRw52a allele as an important susceptibility marker for disease development. [37] HLA-DRB3*0101 appears to be the most common susceptibility locus in 55% to 65% of PSC patients [21] and may be universal in end-stage liver disease from PSC. [13] In Scandinavia, however, the HLA alleles DRB1*0301 and DRB1*1301 have been reported as the primary susceptibility loci in PSC. [38] Of potential significance is the reported association between HLA-DRB3*0101 and reduced survival among subjects with PSC. [37] Rapid disease progression has also been suggested with the existence of HLA-DR4, in which a protective effect was once previously proposed. [39] A multicenter investigation found the HLA-DR3,DQ2 heterozygous genotype associated with more rapid disease progression in PSC, whereas a protective effect resulted from HLA-DQ6. [41] Although controversial in nature, these findings remain to be confirmed among larger populations to verify the predictive effect of HLA alleles on disease outcome in PSC. A number of non-MHC candidate genes may be related to the immune dysregulation in PSC. Serum cytokines, including interleukin-1 (IL-1) and IL-10, have been examined as inflammatory response mediators in PSC. No association between IL-1 and IL-10 gene polymorphisms with susceptibility or resistance to PSC has been observed. [40], [45] Class I MHC chain A (*MICA*) and B (*MICB*) genes have also been studied in PSC. *MICA5.1* (90% versus 74%) and *MICB24* (58% versus 29%) were significantly increased among PSC patients compared with controls. [42] The frequency of PSC patients carrying all four alleles was also significantly greater than controls (49% versus 18%). Among two independent PSC populations, [43] the MICA*008 allele was more common in PSC than controls (66% versus 48%), whereas the MICA*002 allele had a protective effect from PSC. The influence of a matrix metalloproteinase-3 functional polymorphism has also been reported in PSC. [44] Although reductions in allele 6A homozygosity were observed in PSC compared to controls, the development of portal hypertension was significantly associated with 5A homozygosity in PSC.

Non–Immune Mediated

The primary target of immune-mediated injury in PSC is the biliary epithelium. Increased expression of class II MHC molecules on biliary epithelium in PSC to support their role as antigen-presenting cells remains controversial.

The strong association between PSC and IBD has suggested that an infectious etiology may be present in specific individuals. [46] Alterations in diseased bowel mucosa facilitating portal bacteremia may be responsible. Increased serum levels of tumor necrosis factor (TNF) have been observed in animal models from portal bacteremia, resulting in activated hepatic Kupffer cells. [47] Clinical findings in humans, however, are noted for the relative lack of significant colonic inflammation in patients with PSC and after total proctocolectomy for refractory UC. [48] As with PBC, the role of *Helicobacter* species infection in developing PSC has been examined. The significance of an increased rate of *Helicobacter* species DNA in patients with PSC (especially those with UC) compared with noncholestatic liver disease requires further study. [49] An increased rate of a-Hemolytic streptococci from explanted liver tissue [50] and bile [51] have also been found in PSC patients. Among PSC patients with or without previous ERCP, a-hemolytic streptococci were the most frequent isolate despite similar positive culture rates. The impact of a-hemolytic streptococci on etiopathogenesis or disease progression in PSC, however, remains unknown. Viral infections such as cytomegalovirus among immunocompromised hosts can result in PSC-like changes. [52]

Immunodeficiency syndromes have been identified with cholangiographic changes suggestive of PSC including familial immunodeficiency syndrome, X-linked immunodeficiency, angioimmunoblastic lymphadenopathy, and acquired immunodeficiency syndrome (AIDS). [53]

Bile acid metabolite hepatotoxicity has been observed in animal models after chenodeoxycholic acid breakdown to lithocholic acid by bacterial 7-a-dehydroxylation in the colon. No major bile acid abnormalities among human subjects with PSC have been identified. [54]

Biliary tree injury resulting from hepatic artery chemoembolization with fluorodeoxyuridine, 5-fluorouracil, and mitomycin can yield cholangiographic features compatible with PSC. [55] Direct toxicity or ischemia to bile ducts is hypothesized.

CLINICAL MANIFESTATIONS

Asymptomatic elevations in serum liver biochemistries or the development of symptoms such as fatigue (75%) and pruritus (70%) are observed with the diagnosis of PSC (Table 101-1). Jaundice (65%) and weight loss (40%) are less common and should raise the suspicion of advanced disease or a complication related to PSC. [1], [2], [4], [8] The average duration of symptoms before diagnosis is 2 years. Fever and abdominal pain in a patient with PSC are highly suggestive of bacterial cholangitis, which can manifest at initial presentation. [56] A previous history of IBD and elevated serum liver biochemistries often prompts investigations to exclude PSC. Among patients with refractory UC requiring total proctocolectomy, it has been shown that PSC may still develop. [48]

FINDING	PREVALENCE (%)
Symptom	
Fatigue	75
Pruritus	70
Jaundice	65
Weight loss	40
Fever	35
Asymptomatic	10
Sign	
Hepatomegaly	55
Jaundice	50
Splenomegaly	30
Hyperpigmentation	25
Xanthelesma	4

TABLE 101-1 Symptoms and Signs at Initial Presentation in Primary Sclerosing Cholangitis

Asymptomatic Disease

Although symptomatic PSC is commonly associated with jaundice or pruritus, an increasing number of asymptomatic cases are being identified. Asymptomatic patients have represented between 15% and 25% of most reported observational cohorts examined. [4], [5] and [6], [9], [57] Recent reports, however, have observed prevalence rates of greater than 40% for asymptomatic PSC. [8]

Progression of liver disease can also occur among asymptomatic patients, yet existing data are conflicting. Over a 6-year period, advancing liver disease was noted among 34 (76%) of 45 asymptomatic subjects with a 31% rate of liver transplantation or death. Minimal histological abnormalities (stage I or II) on liver biopsy with no cases of cirrhosis were found in most subjects at study entry. The estimated median survival rate for asymptomatic patients without liver failure was 75% at 7 years versus 96% for comparable age- and sex-matched healthy controls. [57] Among 13 (31%) of 42 PSC patients who were asymptomatic at initial presentation, Helzberg and colleagues [4] reported that 11 individuals remained symptom free for a mean duration of 37 months, resulting in an overall survival rate of 75% at 9 years. No significant decompensation occurred over a 4.4 year period among 27 asymptomatic subjects. [3]

Despite a silent or subclinical presentation at diagnosis, there is evidence to support that asymptomatic PSC becomes symptomatic. Fatigue, pruritus, jaundice, or abdominal discomfort develop in 60% of cases. [4], [5] and [6], [8] The similar frequencies in clinical deterioration among asymptomatic and symptomatic patients, however, emphasizes the lack of predictive ability associated with subjective complaints at diagnosis. [4], [6] Differences in criteria for defining the asymptomatic state may also explain the discrepancy in published results. Fatigue and weight loss have been reported in up to 30% and 24% of asymptomatic subjects, respectively. In addition, advanced histological features have been demonstrated in 30% to 43% of subjects considered asymptomatic at diagnosis. [3], [57]

Symptomatic Disease

Most subjects in reported investigations have been described as symptomatic based on findings at diagnosis. [3], [4], [5], [6], [7], [8] and [9] Helzberg and colleagues [4] first reported an 18% mortality rate among patients with PSC over a mean follow-up period of 56 months. All deaths occurred in symptomatic individuals. A similar mortality rate of 14% over 51 months has been observed with a reduced median survival for symptomatic individuals as well (112 months versus 144 for the entire cohort). [8] Two larger series, however, have provided evidence to suggest a more aggressive disease course for symptomatic PSC. Among 174 PSC subjects followed at the Mayo Clinic, [5] a 41% combined rate for liver failure, cholangiocarcinoma, and need for transplantation was observed over 6.25 years of follow-up. Median survival for the entire cohort was 11.9 years. For symptomatic patients with PSC, the median survival was reduced to between 8 and 9 years. Similar results were reported from Kings College, [6] where a median survival of 12 years among 126 subjects over a 6.9 year period of follow-up was observed.

Biochemical Features

Serum hepatic biochemical parameters usually reflect a cholestatic profile. Alkaline phosphatase values often predominate, with elevations up to three times the upper limit of normal in nearly 95% of cases. [1], [2], [3], [4], [5], [6], [7], [8] and [9] Serum alanine and aspartate transaminase levels can also be found between two and three times the upper limit of normal. [3], [4], [8], [9], [56] Of note, serum total bilirubin is normal in 60% of individuals. [1], [2], [56] Increased levels of total bilirubin, which often fluctuate in PSC, are worrisome for advanced disease when persistent elevations occur. No significant differences in serum liver biochemistry profiles have been seen among asymptomatic and symptomatic individuals at diagnosis. [57] Normal serum albumin and prothrombin time levels reflecting hepatic synthetic function are found in most cases at diagnosis. [3], [4], [5], [6], [7], [8] and [9]

Serologic Features

Serum immunoglobulin levels and autoantibodies are associated with PSC. Elevations in IgG and IgM in up to 40% and 60% of individuals are observed. [2], [7] ANA and smooth muscle antibody ASMA can also be detected but are in lower titers compared to AIH. [2], [7], [58] Antimitochondrial antibodies (AMAs) are exceedingly rare in PSC. [5], [58] pANCAs are detected in the serum of PSC patients at rates between 30% and 80% [30], [31] but lack specificity because they are also observed in PBC, AIH, and UC. [31], [59] Angulo and colleagues [60] have observed a greater rate of seropositivity for ANA, anticardiolipin, antithyroperoxidase, pANCA, and rheumatoid factor among PSC patients compared with control subjects. Ninety-seven percent of PSC patients were found to have one or more autoantibodies, whereas 81% were found with three or more. The presence or absence of IBD was not significantly associated with autoantibody history.

Radiographic Features

Cholangiography The diagnosis of PSC is made by cholangiography. Both ERCP and percutaneous transhepatic cholangiography (PTC) have been employed in a nonoperative setting. PSC commonly affects the entire biliary tree in a heterogeneous manner. Intrahepatic duct involvement is nearly universal among patients with PSC. [61] Only 20% of patients have hilar duct involvement with sparing of the remaining extrahepatic duct. Segmental fibrosis of both intrahepatic and extrahepatic ducts with subsequent saccular dilation of normal intervening areas results in the characteristic beads-on-a-string appearance seen in PSC (Fig. 101-1). Bile duct strictures are of varying lengths but can be up to several centimeters. The serrated appearance of bile duct walls and occasional diverticulum are also identified in extrahepatic portions of the biliary tree in 25% of cases. [62]

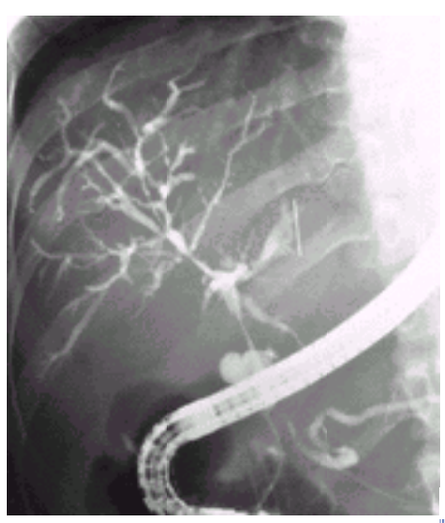

FIGURE 101-1. Cholangiographic features in primary sclerosing cholangitis. Multifocal strictures with intervening saccular dilation (beads-on-a-string appearance) of both intrahepatic and extrahepatic bile ducts are shown.

The initial use of magnetic resonance cholangiopancreatography (MRCP) for suspected PSC has been increasingly recognized. [63], [64] Among 23 of 73 patients identified with PSC, [63] an overall diagnostic accuracy of 90% for MRCP compared with 97% for invasive cholangiography was reported. Similar conclusions were observed among 34 subjects with PSC in an independent study. [64] Advantages of MRCP over invasive cholangiography include visualization of bile ducts proximal to obstructed areas and hepatic parenchymal assessment to exclude features of cirrhosis and portal hypertension. The inclusion of subjects with advanced PSC and overestimation of stricture length by MRCP have been observed as potential biases in both investigations.

Cross-Sectional Imaging Computed tomography (CT) [65], [66] and [67] and magnetic resonance imaging (MRI) [67], [68] and [69] have been used in the evaluation of PSC. Radiologic features of PSC detected by cross-sectional imaging include segmental dilation of intrahepatic bile ducts which tapers toward the hilum. [65] Gallstones have been identified in up to 25% of patients with PSC, whereas asymmetric thickening of the gallbladder wall independent of cholelithiasis may also be observed. [66] Marked hypertrophy of the right and caudate hepatic lobes with simultaneous atrophy in the remaining liver is often seen in cirrhotic-stage PSC. [67] As observed for other etiologies of cirrhosis, the presence of confluent hepatic fibrosis and regenerative nodule formation may be confusing for hepatocellular carcinoma. With MRI, the presence of abnormal T2-weighted signal intensity from periportal areas is common and correlates with fibrosis. [68] Enlarged areas (greater than 1.5 cm) of abnormal T2 signal within portal areas are considered suspicious for cholangiocarcinoma. [69]

Histological Features

Liver biopsy is required for staging disease severity and determining prognosis in PSC. Periductal fibrosis with inflammation, bile duct proliferation alternating with obliteration, and ductopenia constitute the main histological findings. [70] Fibroobliterative cholangiopathy, which is considered the most diagnostic finding on liver biopsy PSC, is present in only 10% of all cases. [71] However, similar findings may occur in varying stages of other conditions, such as PBC, AIH, or chronic extrahepatic bile duct obstruction. The histological abnormalities in PSC are classified into four stages. Stage I (portal stage) is associated with a nonspecific inflammation confined to the limiting plate that surrounds the portal tract. Fibrosis is usually not seen at this stage. Stage II (periportal stage) is associated with inflammation extending beyond the limiting plate with expansion of the portal tracts. Fibrosis is seen involving portal and periportal areas. Stage III (septal stage) is characterized by septal or bridging fibrosis spanning portal tracts in conjunction with bridging necrosis (Fig. 101-2). Stage IV, or cirrhosis, is when complete septal fibrosis in association with nodular regeneration is present. [70]

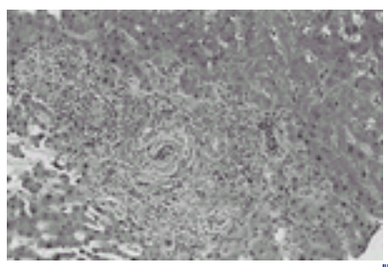

FIGURE 101-2. Histologic features of primary sclerosing cholangitis. Portal tract expansion with chronic nonspecific inflammation, edema, and periportal fibrosis consistent with stage II disease is shown.

DIFFERENTIAL DIAGNOSIS

The identification of serum hepatic biochemical elevations consistent with cholestasis in a male patient with concurrent IBD is strongly suggestive of PSC (Table 101-2). Excluding structural causes for biliary obstruction including choledocholithiasis by ultrasonography or cross-sectional imaging is required. Cholangiography by direct or cross-sectional imaging methods that reveals the typical features of PSC is considered the gold standard for diagnosis. Although liver biopsy is not mandatory for confirming the existence of PSC, it is helpful for staging and excluding other causes of chronic cholestasis.

Extrahepatic biliary tract obstruction
Choledocholithiasis
Strictures
Malignancy
Primary biliary cirrhosis
Autoimmune hepatitis
Pyogenic hepatic abscesses
Fungal cholangitis
Acquired immunodeficiency syndrome cholangiopathy
Choledochal cyst
Cystic fibrosis
Cirrhosis
Intrahepatic hepatocellular carcinoma
Cholangitis glandularis proliferans
Eosinophilic cholangitis

TABLE 101-2 Differential Diagnosis of Primary Sclerosing Cholangitis

PBC more commonly affects women than men (9:1 ratio) and is rarely associated with IBD. The diagnosis is confirmed by elevated serum titers of AMA. Early histological involvement is characterized by lymphoplasmacytic portal infiltration or granulomatous inflammation (florid duct lesion). [72] AMA-negative PBC or autoimmune cholangitis is clinically similar to PSC in the absence of cholangiography. With normal biliary tree visualization, a liver biopsy is required to distinguish this condition from small duct PSC, in which involvement with ductular fibrosis is more prominent. [73]

AIH is distinguished from PSC by (1) predominant elevations in serum aminotransferase levels compared with alkaline phosphatase and bilirubin; (2) the presence of elevated serum ANA or ASMA titers; (3) extensive interface or lobular lymphoplasmacytic inflammation; and (4) normal cholangiography. [74] In children, the initial presentation of PSC often resembles AIH; thus, cholangiography is necessary for accurate diagnosis. [75] Overlap syndromes between AIH and PSC among pediatric and adult populations have been recognized in 2% to 20% of cases based on various definitions. [58], [76], [77] Cholangiography in these patients, however, must demonstrate features of PSC before consideration of an overlap state is made. Potential benefit from corticosteroid therapy among select individuals may be attained.

The development of pyogenic hepatic abscesses with invasion of adjacent bile ducts can result in irregularities reminiscent of PSC. [78] Frequent attacks of ascending cholangitis in a native of Southeast Asia are consistent with the diagnosis of recurrent pyogenic cholangitis (oriental cholangiohepatitis). Cholangiography is noted for dilation of intrahepatic and extrahepatic ducts, decreased arborization with tapering of peripheral ducts (arrowhead sign), and straightening of the intrahepatic ducts from periductal fibrosis. [79] Systemic fungal infection may colonize or invade the biliary tree, resulting in cholangiographic features similar to PSC. [80] AIDS cholangiopathy presents with striking elevations in serum alkaline phosphatase with or without symptoms of jaundice and abdominal pain. Extrahepatic bile duct dilation and papillary stenosis are most commonly observed. Thickened bile duct walls rather than saccular dilatations in AIDS cholangiopathy distinguish this entity from PSC. [81]

Choledochal cyst involvement of intrahepatic bile ducts can also resemble PSC. Ultrasonography usually shows marked areas of ductal dilation, and cholangiography is almost invariably diagnostic. [82] Cystic fibrosis, which involves intrahepatic bile duct cystic dilation, can produce a cholangiogram typical of PSC. [83]

Diffuse abnormalities of the biliary tree without discrete extrahepatic stricturing may also be observed with conditions such as cirrhosis and intrahepatic hepatocellular carcinoma. In cirrhosis, the presence of distal duct tapering is noted when cholangiography is performed. Cross-sectional imaging and liver biopsy (if indicated) will confirm the presence of hepatic neoplasia when suspected. Biliary duct changes similar to that in cirrhosis are also observed if cholangiography is performed. [84]

Cholangitis glandularis proliferans [85] (a rare disorder similar to adenomyomatosis of the gallbladder) and eosinophilic cholangitis [86] can present with nonstricturing intrahepatic duct dilation resembling PSC.

ASSOCIATED CONDITIONS

An increased prevalence for concurrent autoimmune disorders has been recognized in PSC (Table 101-3) but remains less common than in PBC. UC remains the most commonly associated autoimmune disorder in PSC (70% to 80%) [2], [5], [6], [7] and [8], [10], [11], [12], [13] and [14] with colonic involvement from Crohn's disease in 5% to 8% of cases. [11], [14] Other diseases include pancreatitis (10% to 25%), [87], [89] diabetes mellitus (5% to 15%), [88] and autoimmune thyroid disease (3% to 5%). [88] Rare associations include Sicca syndrome, [89] immune thrombocytopenic purpura, [90] mediastinal or retroperitoneal fibrosis, [91], [92] pseudotumor orbit, [92] Peyronie disease, [93] rheumatoid arthritis, [94] ankylosing spondylitis, [95] Hodgkin disease, [96] membranous nephropathy, [97] glomerulonephritis, [98] autoimmune hemolytic anemia, [99] celiac disease, [100], [101] histiocytosis X, [102] hypereosinophilia, [86] focal nodular hyperplasia, [103] and Down syndrome. [104]

DISEASE	PREVALENCE (%)
Ulcerative colitis	70–80
Crohn's colitis	5–8
Pancreatitis	10–25
Diabetes mellitus	5–15
Autoimmune thyroid disease	3–5

TABLE 101-3 Selected Extrahepatic Autoimmune Diseases Associated with Primary Sclerosing Cholangitis

DISEASE-RELATED COMPLICATIONS

Fatigue

The frequency of reported fatigue by individuals with PSC averages between 60% and 70%. [56], [68] Its level of intensity appears independent of histological and cholangiographic disease severity. [1], [2], [3], [4], [5], [6], [7] and [8] Reported fatigue has been associated with reductions in health-related quality of life in PSC. [105] Knowledge of the exact mechanism responsible for fatigue in PSC remains unknown.

Pruritus

In PSC, the prevalence of pruritus ranges from 40% to 60% on average in reported observational cohort studies. [1], [2], [3], [4], [5], [6], [7] and [8] Proposed mechanisms of action have included bile salt retention with skin deposition [106] and the accumulation of endogenous opioid ligands and receptors within the central nervous system. [107] As with fatigue, the severity of pruritus in PSC appears to be independent of histological stage, although extrahepatic stricturing can influence symptom intensity. [1], [2], [3], [4], [5], [6], [7] and [8]

A number of medical therapies have been evaluated for symptom relief of pruritus. Initial therapy with antihistamines is often associated with residual sedation and only limited success. [72] Cholestyramine (a bile acid–binding resin) may decrease the intensity of pruritus but is rarely met with complete symptom resolution. [108] Among subjects with intact gallbladders, the use of 4 g before or after breakfast is thought to maximize bile acid sequestration and symptom alleviation. Divided doses over 24 hours in subjects after cholecystectomy are more effective than once- or twice-daily dosing. Administration of cholestyramine several hours apart from other medications is recommended to prevent elimination without gastrointestinal absorption.

Hepatic enzyme inducers, including phenobarbital [109] and rifampin, [110], [111] and [112] have been associated with improvements in bile acid flow experimentally and in the treatment of pruritus. Excessive sedation at higher doses has limited the clinical utility of phenobarbital. [109] Rifampin in doses of 300 to 600 mg/d is associated with a rapid onset of action and symptom relief in PSC. [110] Although well-tolerated, this medication is also associated with cholestatic liver injury and bone marrow aplasia in up to 10% of cases. [111] Ursodeoxycholic acid (UDCA) at doses of 13 to 15 mg/kg/d has not been shown to be effective in the treatment of pruritus. [112] Other therapeutic options used for pruritus in PSC, including oral naltrexone, [113] activated charcoal hemoperfusion, [114] and plasmapheresis, [115] have been sporadically applied with good results. Among patients with intractable pruritus refractory to medical therapy, liver transplantation is the only existing therapeutic option. [72]

Metabolic Bone Disease

Metabolic bone disease in PSC is related to osteopenia rather than osteomalacia (defective bone mineralization) among North American patients. [116] Decreased bone formation (osteoporosis) rather than increased resorption is thought to be an important mechanism in the pathogenesis of bone disease. [117] Calcium and vitamin D metabolism is usually abnormal among anicteric patients with PSC. [118] The recognition of improved bone mineral density about 6 months after liver transplantation also suggests the presence of an unidentified substance creating osteopenia. [121] About 50% of PSC patients have osteopenia (i.e., a Z score of less than -1.0 by lumbar spine bone mineral densitometry), whereas osteoporosis (i.e., a Z score of less than -2.5) is less common than in PBC (10% versus 35%). [119], [120] As many as 50% of liver transplant recipients have bone mineral densities below the fracture threshold. [121] Reasons for this difference compared to PBC include a greater proportion of male patients with higher baseline bone mass and a slower rate of bone loss compared with women. Osteoporosis in PSC should be suspected among postmenopausal women and individuals with advanced age, a long duration of IBD, and the presence of advanced hepatic disease. [119], [120]

Ensuring adequate intake of calcium (1000 to 1200 mg/d) with weight-bearing activity is recommended as an initial treatment for all individuals. Measurement of serum vitamin D levels is also an essential part of the preventative management in PSC. The presence of vitamin D deficiency is variable among patients with metabolic bone disease. Oral replacement therapy is indicated if measured serum levels are reduced compared with local normal values. The 25-hydroxylation of vitamin D is intact among PSC patients; therefore, vitamin D can be used rather than 1,25-dihydroxyvitamin D or 25-hydroxyvitamin D. Dosing is generally between 25,000 and 50,000 IU two to three times per week. The efficacy of bisphosphonate (alendronate, etidronate) therapy in PSC remains unknown. No benefit from calcitonin was observed compared with placebo among PSC patients after liver transplantation. [121]

Steatorrhea

Impairment in bile acid delivery resulting in insufficient critical micellar concentrations in the small intestine is the most common cause of steatorrhea with advanced hepatic disease from PSC. [1] The coexistence of celiac disease with PSC has been reported in an increasing number of patients. [100], [101] Exocrine pancreatic insufficiency has also been observed as a cause of steatorrhea related to overall glandular dysfunction, which may be associated with chronic pancreatitis. [87], [89] Determining the exact cause is important because specific therapeutic options can be employed for symptom relief. In patients with decreased bile acid concentrations, the oral replacement of medium-chain triglycerides for long-chain compounds is usually of benefit. Adherence to a gluten-free diet and pancreatic enzyme replacement therapy are recommended for patients affected by celiac disease and pancreatic insufficiency, respectively.

Micronutrient Deficiency

Malabsorption of fat-soluble vitamins is common with advanced hepatic disease from PSC. Intrahepatic cholestasis and impaired bile acid delivery to the small intestine are putative causes. Vitamin A deficiency, observed in 20% to 50% of cases, is often clinically asymptomatic. When symptomatic, the presence of night blindness is observed and should be corrected by oral replacement therapy with 25,000 to 50,000 IU two to three times a week. As discussed previously, vitamin D deficiency can occur and is the next most common fat-soluble vitamin deficiency in 15% to 50% of cases. [118]

Vitamin E deficiency is a rare occurrence in PSC. When present, it may be associated with ataxia from neuropathic involvement of the posterior vertebral columns of the spinal cord. Oral replacement therapy with 400 IU daily is indicated in asymptomatic patients. [122]

Vitamin K deficiency is most commonly related to severe cholestasis, cholestyramine treatment for pruritus, or significant hepatic dysfunction from end-stage PSC. The initiation of water-soluble oral vitamin K, 5 mg daily, with repeat measurement of prothrombin time is effective in determining the extent of malabsorption. If correction of prothrombin time is encountered, daily oral doses between 5 mg and 10 mg per day should be initiated.

Biliary Calculi

Biliary calculi have been reported in up to 40% of patients with PSC. [1], [2], [56], [66] Cholelithiasis is common and often requires cholecystectomy when symptomatic biliary colic occurs in nonadvanced hepatic disease. [66] Choledocholithiasis is reported at frequencies of 5% to 15% and more commonly among symptomatic individuals with biliary strictures. Most calculi are pigmented in nature and involve both central and peripheral bile ducts. [123] Symptoms, including intractable abdominal pain or recurrent bacterial cholangitis, have been successfully treated by endoscopic [124], [125] or percutaneous [126] methods to provide biliary decompression and stone extraction. Biliary surgery, including stone extraction and bilioenteric anastomosis, [126], [127] may be required only in cases refractory to endoscopic and percutaneous approaches.

Dominant Stricture

Dominant strictures or stenoses occur in 15% to 20% of PSC patients. [128] Clinical manifestations include a sudden increase in serum alkaline phosphatase or bilirubin in the asymptomatic patient, progressive jaundice, and bacterial cholangitis. [1], [2], [56] Cholangiography is required to determine the nature and extent of existing strictures. [128] Balloon dilation of identified stenoses by endoscopic [124] or percutaneous [128] approaches often provides significant clinical improvement. For cases with incomplete response to balloon dilation alone, the use of endoscopic stents for 3 to 6 months to prevent recurrent stricture formation has been advocated. [129], [130] Reports of infectious complications, however, have raised questions about the efficacy of this approach. [131] Cytologic brushings with pinch biopsies of all dominant strictures is required to help exclude the presence of cholangiocarcinoma. [132]

Bacterial Cholangitis

Bacterial cholangitis in PSC is most commonly associated with a previous history of biliary tract surgery, [56] bile duct calculi, [123] or ductal obstruction from a dominant stricture. [128] Once suspected, empiric broad-spectrum intravenous antibiotic therapy should be initiated. Cholangiography (with ERCP if possible) should then be considered to exclude obstruction and provide biliary decompression. The use of prophylactic antibiotic therapy (oral fluoroquinolone, trimethoprim-sulfamethoxazole), especially in the setting of short-term biliary stent placement, may reduce the risk for recurrent cholangitis. Recurrent episodes of bacterial cholangitis in PSC can be an indication for liver transplantation. [56]

Cholangiocarcinoma

A measurable proportion of subjects with PSC will develop cholangiocarcinoma as a complication of disease. Prevalence rates appear to vary between 4% and 20% in most reported series. [3], [5], [6], [7], [8] and [9], [133] Between 30% and 45% of individuals with PSC are diagnosed with cholangiocarcinoma at the time of liver transplantation [134], [135], [136], [137], [138], [139], [140], [141] and [142] and autopsy. [3], [143], [144] PSC may be simultaneously present in 30% to 50% of cases of diagnosed cholangiocarcinoma. The anatomic distribution of cholangiocarcinoma in PSC primarily involves the hilum (75% of cases), intrahepatic ducts (16%), and gallbladder (8%). [145]

Risk factors for cholangiocarcinoma in PSC remain incompletely understood. No association with stage of hepatic disease in PSC is observed, with coexisting cirrhosis affecting only 20% of patients with cholangiocarcinoma. [145], [146] Advanced age and longer duration of IBD [144] have also been cited. Formal epidemiologic investigations (case-control) have been performed in two independent populations. [147], [148] Previous or current cigarette smoking has been suggested to be associated with cholangiocarcinoma in only one study. [148] The presence of colorectal dysplasia and carcinoma in patients with PSC and UC has been noted as a possible risk factor for cholangiocarcinoma. [143]

The presence of oncogenic mutations, inactivation of tumor suppressor genes, and apoptosis have been postulated as mechanisms for developing cholangiocarcinoma in PSC. K- *ras* mutations found in the biliary secretions of PSC patients have been associated with cholangiocarcinoma, [149], [150] yet this remains a nonspecific finding. Aberrations of the *p53* tumor suppressor gene occur in 30% to 80% of PSC cases complicated by cholangiocarcinoma [151] but are often seen as a late finding. Chromosomal loss of 9p21 and *p16* tumor suppressor gene inactivation have also been reported. [152] The recognition of biliary dysplasia as a precursor to cholangiocarcinoma has been inconsistently suggested. [153], [154]

The diagnosis of cholangiocarcinoma in PSC remains challenging despite recent technological advances. Using the Mayo PSC model among 48 subjects undergoing liver transplantation, the incidence of biliary tract malignancy was significantly associated with model risk scores higher than 4.4 independent of clinical status. [155] Subsequent investigations to confirm this observation have not been performed. Marked bile duct dilation in association with a polypoid structure larger than 1 cm on cholangiography has been reported in association with cholangiocarcinoma. The distinction between benign and malignant biliary strictures with less prominent findings, however, remains difficult. Comparisons between ultrasound, CT, and MRI have also demonstrated inadequate ability to distinguish between PSC patients with and without cholangiocarcinoma. [156] Emerging techniques such as positron emission testing, which show initial promise, require further examination. [157]

Serum tumor markers, including carbohydrate antigen 19-9 (CA 19-9) and carcinoembryonic antigen (CEA), have been employed for the early detection and diagnosis of cholangiocarcinoma. CA 19-9 levels higher than 100 U were found to have a sensitivity of 89% and specificity of 86% for detecting cholangiocarcinoma in PSC. [158] The combination of both markers (CA 19-9 + CEA × 40) exceeding values of 400 U was reported to have an 86% diagnostic accuracy despite a sensitivity of 67%. [159] Similar results using this formula were reported in an independent study. [160] The prospective evaluation of four tumor markers (CA 19-9, CEA, CA 50, and CA 242) among 75 patients failed to achieve high test performance characteristics for predicting cholangiocarcinoma. [146] Serum CA 19-9 levels of more than 100 U/mL for detecting cholangiocarcinoma among PSC patients have recently been noted with further limitations in diagnostic sensitivity (52%) and specificity (76%). [161]

The performance of endoscopic biopsy and cytologic brushing for the diagnosis of cholangiocarcinoma is noted for good specificity but poor sensitivity. [132], [162] Among 47 patients with dominant strictures identified at ERCP, a 60% sensitivity and 89% specificity using biopsy and brushings was observed. [132] False positivity with cellular atypia from chronic inflammation may also occur. [162] DNA aneuploidy detection by flow cytometry has been reported in 80% of individuals with cholangiocarcinoma versus 12% with benign strictures. [163]

Therapeutic options for cholangiocarcinoma in the setting of PSC are limited. For extrahepatic biliary resection in PSC, operative complication and morality rates exceed 30% and 6%, respectively, in the absence of bile duct cancer. [164] Highly selected patients, however, may derive benefits from surgical resection. [165] Curative attempts with liver transplantation to date have resulted in poor outcomes from residual tumor at surgical margins and local hepatic lymph node involvement. [136], [139], [141], [142] However, the presence of cholangiocarcinoma in situ (tumor smaller than 1 cm) found incidentally at liver transplantation has been associated with long-term survival rates similar to those of patients with PSC alone. [138], [166] To improve outcomes after liver transplantation, the implementation of preoperative radiation and chemotherapy in highly selected patients with unresectable cholangiocarcinoma and PSC has yielded a short-term (less than 5-year) survival of greater than 80% at the Mayo Clinic. [167]

Portal Hypertension

The development of biliary cirrhosis from progressive PSC can result in the typical complications of portal hypertension associated with other chronic liver diseases. These include ascites, spontaneous bacterial peritonitis, hepatic encephalopathy, esophagogastric variceal bleeding, portal hypertensive gastropathy, hepatorenal syndrome, and hepatopulmonary syndrome. A unique complication is peristomal variceal bleeding, which occurs among patients with IBD who have undergone proctocolectomy with ileostomy formation. [168] Newer surgical techniques, including ileal pouch–anal anastomosis at the time of proctocolectomy, are indicated among patients with identified PSC to prevent this complication. [169] Pharmacological (ß-blocker) and endoscopic (sclerotherapy) therapies are without long-term efficacy. [168] Transjugular intrahepatic portosystemic shunt placement has been shown to be effective in treating uncontrolled bleeding and preventing further episodes. [170]

Colonic Dysplasia and Carcinoma

The association between an increased risk for colonic dysplasia or carcinoma and PSC in the setting of UC has gained interest in recent times and remains controversial. Decreased bile acid secretion and increases in secondary bile acid exposure to colonic mucosa are considered potential mechanisms. [171] In addition, a greater proportion of right-sided colon neoplasia has been observed in UC patients with PSC versus UC alone. [172], [173] and [174] Absolute cumulative risks for colorectal dysplasia and carcinoma of 5% (at 10 years), 31% (20 years), and 50% (25 years) have been observed in patients with PSC and UC, compared with 2%, 5%, and 10% risks for UC alone, respectively. [143] Other studies, however, have failed to confirm a significant risk from PSC for colonic dysplasia and carcinoma. [175], [176] Given the potential association between bile acid exposure and colorectal neoplasia, the use of UDCA was strongly associated with a decreased risk for colonic dysplasia among 59 PSC patients undergoing colonoscopic surveillance for UC. This association remained after adjustment for duration and severity of PSC. A younger age at IBD onset, however, was also associated with an increased dysplasia risk. [177] Confirmation of findings in a controlled trial setting is warranted. Annual surveillance colonoscopy with biopsy is recommended in patients with PSC and UC, especially after liver transplantation among patients with UC in the absence of

proctocolectomy. [135], [178], [179] and [180]

DISEASE-MODIFYING THERAPIES

Liver transplantation is the only effective treatment for end-stage hepatic disease resulting from PSC. Because of the variable nature of disease progression, the development of randomized clinical trials for assessment of therapies has been difficult. As a result, there is no effective medical therapy identified for the treatment of PSC (Table 101-4).

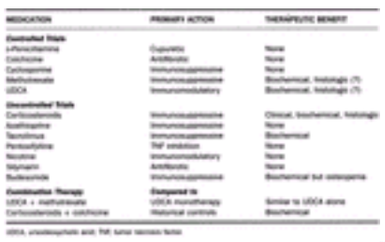

TABLE 101-4 Medical Therapies Evaluated in Primary Sclerosing Cholangitis

Medical Therapy

D-Penicillamine An increase in hepatic copper found among patients with chronic cholestatic liver disease including PSC provided the rationale for examining D-penicillamine. Among 70 patients randomized to active drug or placebo, there was no reported benefit in terms of biochemical improvement, histological stabilization, or survival free of liver transplantation after 3 years of follow-up. Major toxicities, including renal disease, resulted in drug discontinuation among 20% of subjects. [181]

Colchicine A randomized controlled trial of 84 subjects with PSC (44 given colchicine 1 g/d, 40 placebo) revealed no differences in biochemical, histological, or survival between groups after 36 months of follow-up. [182] Combination therapy with prednisolone and colchicine has been associated only with initial biochemical improvement after 24 months of treatment. [183]

Corticosteroids The report of beneficial results from corticosteroid use has only been demonstrated in uncontrolled trials. Patients with early-stage histological disease may have symptomatic and biochemical improvement. [184] The use of oral budesonide, a systemic corticosteroid with significant first-pass hepatic metabolism and reduced toxicity, in 21 PSC patients was associated with improved hepatic biochemistries alone at the expense of worsening osteopenia in a pilot investigation. [185] The use of prednisone or budesonide in combination with UDCA was not associated with significant clinical or biochemical improvement in a double-blinded, randomized controlled trial. [186] A combination of prednisone, azathioprine, and UDCA in 15 patients was noted for significant biochemical and histological improvement. These results were also observed among individuals with partial biochemical responses to UDCA. [187] Case reports detailing beneficial effects from topical corticosteroid therapy by endoscopic delivery through a nasobiliary drain were complicated by an increased rate of bacterial cholangitis. [188], [189]

Azathioprine No randomized controlled trial for azathioprine monotherapy has been reported to date. Open-label pilot studies are noted for mixed results using biochemical parameter responses. [190], [191]

Cyclosporine A single randomized controlled trial of cyclosporine among 34 patients [192] was noted for the absence of significant symptomatic, biochemical, or histological improvement. Toxicity from cyclosporine, including hypertension and renal insufficiency, was minimal.

Methotrexate Data from a double-blind, randomized controlled trial comparing oral pulse methotrexate (15 mg/wk) with placebo among 24 patients [193] is noted only for reductions in serum alkaline phosphatase. No improvements in total bilirubin, histology, or survival were observed. Early-stage histological disease was associated with possible symptom and biochemical improvement. [194] Combination therapy with methotrexate and UDCA in 19 patients was associated with toxicity (from methotrexate) without biochemical improvement when compared to UDCA alone in 9 patients. [195]

Ursodeoxycholic Acid The mechanism of action attributed to UDCA in the treatment of chronic cholestatic liver disease is multifaceted. In addition to suppressing cytotoxic endogenous bile acid secretion, [196] there is evidence to suggest that UDCA is associated with membrane stabilization [197] and reduced aberrant HLA type I expression on hepatocytes. [198] Improved mitochondrial dysfunction induced by hydrophobic bile acids, decreased cytokine production, and inhibition of apoptosis have also been recently identified in association with UDCA. [199] Early investigations suggested a beneficial effect from UDCA on PSC. [200], [201] Three randomized, controlled trials comparing UDCA with placebo, however, did not show significant improvements in histology or survival despite biochemical parameter response. [202], [203] and [204] Two recent investigations of high-dose UDCA (25 to 30 mg/kg/d) reported improvements in biochemical, [205], [206] cholangiographic, and histological parameters [206] when compared with standard doses. Results from placebo-controlled studies examining high-dose UDCA in PSC are awaited.

Novel Therapies Pilot investigations of oral nicotine [207] and pirfenidone [208] have failed to demonstrate improvements in serum hepatic biochemistries or Mayo PSC risk score. Despite properties of anti-TNF attenuation, the use of oral pentoxifylline was not associated with clinical or biochemical improvement among 20 patients with PSC. [209] No investigation of parenteral anti-TNF antibody use in PSC has been reported to date. Tacrolimus (FK506), a calcineurin-based inhibitor with pharmacological properties similar to cyclosporine, has been associated with significant improvements in biochemical profiles among all 10 subjects in an open-label pilot trial. Mean serum aminotransferase levels among these patients, however, were up to 5-fold greater than normal values, which is not seen in patients with typical PSC. No effect on histology based on limited follow-up was also observed. [210] The nucleoside analog cladribine usually was associated with progression of histological fibrosis despite improvements in periportal inflammation among four PSC patients after 24 months of follow-up. [211]

Endoscopic Therapy

Endoscopic therapy in PSC has been successful in alleviating cholestasis aggravated by mechanical biliary obstruction. Controversy remains, however, regarding the safety and efficacy of ERCP based on the limited numbers of patients reported and variety of endoscopic methods used. Among PSC patients undergoing diagnostic ERCP, a 2% complication rate has been reported compared with 14% among patients with jaundice or cholangitis. [212] With advances in technology and endoscopic development, a number of options for therapeutic intervention are possible, including endoscopic sphincterotomy, hydrostatic balloon dilation, nasobiliary tube (NBT) drainage, and biliary stent placement. The current use of endoscopic dilation with sphincterotomy or stenting is associated with clinical response rates from 70% to 90%. [125], [213]

Among 53 patients requiring ERCP for the evaluation of biliary obstruction, [124] a technical success rate of 88% was observed with 15 subjects (28%) experiencing procedure-related complications. Twenty-eight patients (56%) reported symptomatic improvement after ERCP for a median of 31 months. No change in cholangiographic findings, however, was seen in more than 50% of cases. In the largest series reported to date of 106 PSC patients, [214] dominant stenoses were observed in 43 instances over a median follow-up of 5 years. Most of these stenoses were treated using balloon dilation. Complications included pancreatitis (5.2%), cholangitis (3.3%), and bile duct perforation (0.5%). Survival after balloon dilation of the first recognized stenosis was significantly better when compared with predicted survival using the Mayo multicenter prognostic model.

NBT placement for drainage and saline irrigation has been employed to prevent recurrent biliary obstruction. [188], [189] Among 12 asymptomatic patients with high-grade strictures treated initially by hydrostatic balloon dilation followed by NBT irrigation, [215] an improvement in clinical, biochemical, and cholangiographic features was documented in 75% of cases over 23 months. One patient had no change in clinical status from unresponsive disease. Progressive disease in 3 subjects required treatment by liver transplantation. Further use of NBT irrigation among symptomatic patients has not been reported.

A retrospective examination of 25 symptomatic patients treated by biliary stent placement is noted for an endoscopic success rate of 84%. [129] Clinical and biochemical improvement in all cases was noted after 6 months. Additional procedures for stent exchange was associated with a complication rate of 14% over a 29-month period. During follow-up, only 12 patients (57%) remained asymptomatic with normal hepatic biochemistries. The use of short-term stenting (mean, 11 days) of dominant strictures among 32 patients has been associated with an 83% improvement rate from symptomatic cholestasis (fatigue, pruritus) and biochemical improvement over 4 years. [130] Reintervention rates at 1 year (80%) and 3 years (60%) were observed. A recent investigation advocating the use of aggressive stricture dilation, stenting, or

both, combined with UDCA therapy, in PSC was based on results in an uncontrolled trial setting. [216]

Biliary Reconstructive Surgery

Among pre-cirrhotic patients with extrahepatic biliary strictures refractory to endoscopic and percutaneous therapy, the use of intraoperative resection has been performed successfully. [164], [217], [218] and [219] Common sites of involvement in this setting include the common bile duct (CBD) and perihilar regions. Early reports involving 17 of 22 patients undergoing choledochoenteric anastomosis noted good to excellent results in 13 cases (76%). [217] Transhepatic stenting was employed in 6 cases. An overall survival rate of 82% at 52 months after surgery was observed. Among 31 patients undergoing extrahepatic bile duct resection and long-term stenting, [218], [219] excellent results among 26 pre-cirrhotic individuals were reported. A 20% survival rate over 5 years, however, was noted in patients with cirrhosis.

Comparisons between operative and nonoperative therapies have been reported involving 104 patients treated surgically (n = 54) or using endoscopic balloon dilation (n = 35) and percutaneous stenting (n = 19). [164] In this retrospective investigation, no differences in procedure-related morbidity and mortality were recorded. Improvements in serum bilirubin up to 3 years after resection were significantly better compared with nonoperative therapies. Among 40 patients with cirrhosis at surgical resection, an overall 5-year survival rate of 85% (including liver transplantation in 21 patients) was noted. Transplant-free survival, however, was 78% at 5 years among subjects without decompensated cirrhosis undergoing resection. A 59% overall and 46% transplant-free 5-year survival rate, respectively, was observed among medically treated individuals. Cholangiocarcinoma in 9% of medically treated patients was reported, in contrast to no cases among surgical patients.

Concerns regarding operative bile duct resection complicating the performance of liver transplantation have been raised. [134], [136], [139], [140], [141] and [142] The association between prior bile duct surgery and poor outcomes, including decreased survival, however, has not been conclusively determined. [135], [138] Among select individuals with PSC, a history of prior surgery has been associated with longer operative procedure times, [134], [135], [164] greater intraoperative blood loss, [134], [135] and [136] and increased risks for biliary complications after transplantation. [134], [135], [137]

Liver Transplantation

Among patients with end-stage liver disease from PSC, the current treatment of choice is liver transplantation. PSC is the fourth most common indication for liver transplantation in the United States, accounting for nearly 10% of all procedures. Indications for liver transplantation are similar to other hepatic diseases, including complications from portal hypertension (ascites, variceal bleeding, encephalopathy). Significant impairments in health-related quality of life from fatigue, pruritus, and painful osseous fractures may also be considerations for liver transplantation. Unusual circumstances prompting liver transplantation referral in PSC include recurrent cholangitis despite medical therapy, severe extrahepatic biliary obstruction among individuals with advanced hepatic disease precluding operative repair, and uncontrolled peristomal variceal bleeding. Reports from single centers performing liver transplantation for PSC are noted for patient survival rates of 90% to 97% at 1 year and 83% to 88% at 5 years (Table 101-5). Improvements in operative technique and immunosuppression management have greatly contributed to improvements in patient survival. Long-term graft survival, however, has been affected by a higher incidence of rejection (acute cellular and chronic ductopenic) and hepatic artery thrombosis. [220]

TABLE 101-5 Patient Survival after Liver Transplantation for Primary Sclerosing Cholangitis

Among patients with coexistent UC undergoing liver transplantation, it has been recognized that gastrointestinal symptoms may improve with the initiation of immunosuppressive therapy. Exacerbations of UC, however, are reported in up to 30% to 50% of patients, with some individuals requiring proctocolectomy for disease control. [221], [222] and [223] An increased prevalence of pouchitis among PSC patients after liver transplantation compared with UC patients who undergo transplantation for other hepatic disease states has also been reported. [224] Patients with intact colons appear to be at increased risk for colorectal neoplasia after liver transplantation. [135], [178], [179] and [180], [225] Among 198 post–liver transplantation patients with IBD, [225] a 7% prevalence rate for colorectal cancer was observed in most cases diagnosed within 30 months of liver transplantation. Duration of IBD greater than 9 years was noted in 93% of cases in this series. The cumulative risk for colorectal cancer among individuals transplanted for PSC at the Mayo Clinic [178] was calculated at 1% per year. The cumulative incidence for colonic dysplasia, however, was increased to 15% at 5 years and 21% at 8 years.

An increasing body of evidence suggests that PSC is a recurrent disease after liver transplantation in select patients. [225], [226], [227] and [228] Quantifying the prevalence rate for recurrent PSC, however, has been limited by the absence of uniform diagnostic criteria that exclude known causes of biliary tract dysfunction including ABO incompatibility, chronic ductopenic rejection, early and late hepatic artery thrombosis, and prolonged cold ischemic time. Among 120 PSC patients undergoing liver transplantation at the Mayo Clinic, [228] 24 (20%) developed evidence of recurrent PSC using strict criteria. Cholangiography was diagnostic for recurrent PSC in 92% of cases, whereas 46% of liver biopsy results were compatible with recurrence. An independent study found prevalence rates of recurrent disease closer to 10%. [225] The identification of sustained elevations in serum alkaline phosphatase levels (more than two times the upper limit of normal) beyond 2 years from liver transplantation appears associated with an increased risk for disease recurrence. [228]

NATURAL HISTORY AND PROGNOSIS

Most published investigations to date support the contention that PSC is a progressive disease which can ultimately lead to liver failure or death. A median survival of 9 to 12 years in the absence of liver transplantation appears independent of geographic and environmental influences [3], [4], [5] and [6], [9], [10] Subsets of patients with PSC, however, appear to follow different patterns of disease progression. Because of this significant variation in natural history, the use of mathematical prognostic models has allowed for improvements in estimating risks for disease progression and PSC-related survival independent of liver transplantation.

Subject age, serum total bilirubin, hemoglobin, a history of IBD, and histological stage on liver biopsy were observed as independent predictors of death among PSC patients at the Mayo Clinic. [5] A similar prognostic index including subject age, hepatomegaly, splenomegaly, elevated serum alkaline phosphatase, and advanced histological stage has also been reported. [6] To address variations among independent populations, a multicenter prognostic model was developed using 426 patients from five centers. [229] Subject age, serum bilirubin, splenomegaly, and histological stage emerged as independent predictors of death in PSC. More recently, a revised version of the original Mayo PSC model has eliminated histological stage in favor of variceal bleeding history, avoiding the need for invasive liver biopsy in estimating prognosis. [230]

Examination of the Child-Turcotte-Pugh (CTP) classification as a prognostic model in PSC has yielded mixed results. [231], [232] Significant differences in 7-year survival have been demonstrated among CTP class A (89.8%), class B (68%), and class C (24.9%) patients with PSC. [231] Subject age and CTP classification were independent predictors of mortality. A subsequent investigation of 147 patients, [232] however, was noted for limitations in predicting survival among CTP class B and C patients. CTP classification was not observed as an independent predictor of death in this cohort, in contrast to the Mayo PSC risk score.

Attempts at defining the optimal timing for liver transplantation in PSC have examined the application of existing prognostic models. Among 216 PSC patients who underwent liver transplantation between 1981 and 1990, [139] a dramatic survival benefit was observed compared with predicted survival using the Mayo PSC model (73% versus 28% at 5 years; $p < 0.001$). Long-term survival between subjects grouped by risk score, however, was similar. The occurrence of cholangiocarcinoma was associated with a significant decline in survival independent of pretransplantation Mayo risk score. The presence of ascites, previous upper abdominal surgery, elevated serum creatinine, IBD, and history of biliary tree malignancy have been described as predictors of reduced posttransplantation survival as well. [233]

Additional investigations surrounding outcomes after liver transplantation in PSC have focused on health-related quality of life and resource use. [234], [235] and [236] Among

436 patients with cholestatic liver disease including PSC (n = 208), the independent predictors of postoperative morbidity were subject age, renal failure (defined as serum creatinine higher than 2 mg/dL), Child-Pugh classification, and United Network of Organ Sharing status. [234] Increased resource use based on hospital charges has been associated with a Karnofsky score of less than 40, poor nutritional status, and renal failure. [235] A subsequent multicenter investigation, however, demonstrated that the CTP classification is a significantly better predictor of clinical and economic outcomes than the Mayo risk score. [236]

VARIANT SYNDROMES

Small Duct Primary Sclerosing Cholangitis

A subset of patients with typical features of PSC may have normal-appearing intrahepatic and extrahepatic bile ducts by cholangiography. [237], [238] The diagnosis of small duct PSC is based on (1) serum biochemical profile consistent with cholestatic liver disease; (2) the presence of IBD; (3) normal cholangiogram; and (4) appropriate findings on liver histology. Characteristic features described by liver histology assessment include mild portal mononuclear inflammation, ductular proliferation, and nonsuppurative fibrous obliterative cholangitis. [238] Fibrous-obliterative cholangitis, although considered pathognomic for PSC, occurs in only 10% to 40% of cases. [70] Granulomatous cholangitis (more commonly seen in primary biliary cirrhosis) is seen in 8% of cases. [1] Disease progression is characterized by periportal fibrosis, ductopenia, and eventual biliary cirrhosis. [70] Previously described as part of *pericholangitis*, the prevalence of small duct PSC is estimated between 15% and 35% among adults with UC. [237], [238] As many as 33% of patients with eventual large duct PSC are identified with normal cholangiograms during the course of disease despite histological evidence for PSC. Isolated small duct involvement at diagnosis with eventual recognition of large duct PSC occurs in 40% of cases. [237] Although uncommon, the progression to end-stage liver disease requiring hepatic transplantation has been associated with small duct PSC. [139] Specific reference to small duct PSC in terms of medical therapy has not been widespread given its rare occurrence. The natural history of small duct PSC appears to reflect outcomes related to large duct PSC. [237], [238] Cholangiocarcinoma has been rarely observed among patients with small duct PSC. [239]

Autoimmune Hepatitis–Primary Sclerosing Cholangitis Overlap Syndrome

Patients with PSC and clinical features reminiscent of AIH are classified as having an *overlap syndrome* between the two conditions. [240], [241], [242] and [243] Greater than expected serum aminotransferase elevations (5 to 10 times) and increased titers of serum autoantibodies, including ANA, ASMA, and pANCA, are observed in most cases. [241], [242] and [243] The presence of typical biliary stricturing and dilation seen with PSC is often accompanied by increased lymphoplasmacytic portal or periportal infiltrates on liver biopsy consistent with AIH. In PSC, the presence of overlap with AIH was previously thought to range between 35% and 54% using the original International Autoimmune Hepatitis Group (IAHG) scoring system criteria. [240] Using the revised IAHG scoring system, a decrease in prevalence of AIH-PSC overlap between 1.4% and 11% has been reported. [241], [242] In contrast, van Buuren and colleagues observed similar prevalence rates of AIH-PSC overlap based on either conventional diagnostic criteria (23%) or the revised IAHG scoring system (19.5%). [243] Empiric immunosuppressive therapy for potential benefit among patients with AIH-PSC overlap has been recommended. [240]

Idiopathic Adulthood Ductopenia

Idiopathic adulthood ductopenia (IAD) is a syndrome described by the presence of (1) biochemical parameters consistent with cholestasis; (2) histological evidence of ductopenia involving more than 50% of bile ducts; (3) no evidence for the diagnoses of PSC and IBD; and (4) the exclusion of other known etiologies for cholestasis, including PBC and drug-induced hepatotoxicity. [244] Variation in extent of bile duct loss likely reflects a spectrum in natural history. [238] Other diagnostic possibilities include an unidentified chronic viral cholangitis, small duct PSC in the absence of IBD after colonoscopic investigation, or advanced AMA-negative PBC with resulting ductopenia. [244]

Among 29 reported cases of IAD, [238], [244] a median age of 27 years (range, 15 to 67 years) at diagnosis is observed. Male-to-female gender distribution is estimated at 2:1. Jaundice and pruritus are seen at initial presentation in 33% of cases. Disease progression reported among 10 patients occurred over a median duration of 4 years (range, 5 months to 15 years). Seven of 23 (30%) patients with follow-up greater than 6.5 years died from liver disease, whereas 4 patients required liver transplantation. Corticosteroid or UDCA therapy was associated with clinical improvement in 3 of 4 cases. Among subjects with features similar to IAD despite less than 50% bile duct loss, [245] a benign course in 3 of 24 individuals using sequential liver biopsies over 12 years was reported. Four of 5 patients treated with UDCA had normalization of serum liver biochemical parameters.

ACQUIRED IMMUNODEFICIENCY SYNDROME CHOLANGIOPATHY

Epidemiology

Biliary tract disease in AIDS (AIDS cholangiopathy) was first described soon after the identification of the human immunodeficiency virus (HIV). [246] AIDS cholangiopathy describes a syndrome that resembles sclerosing cholangitis associated with papillary stenosis. [247], [248] Pathogens involved with the development of AIDS cholangiopathy do not appear to affect patients who are immunocompromised for other reasons. The mean age of affected patients is reported at 36.5 years, with a preexisting diagnosis of AIDS for 1 to 2 years before symptomatic biliary tract involvement. [248], [249] CD4 T-cell counts of less than 50/mm ^{3} have also been associated with the risk for AIDS cholangiopathy and significant mortality within 1 year of infection. [249]

Pathogenesis

Infection of the biliary or duodenal epithelium is the primary cause of ductal abnormalities in AIDS cholangiopathy. Among pathogens identified, *Cryptosporidium* species are the most common identifiable etiology and often found without coexistent infection. [247], [248], [250], [251] Among 82 HIV-infected patients exposed to cryptosporidia in a water-borne outbreak, [250] the development of abdominal pain associated with biliary tract disease occurred in 29% of cases and was confirmed by cholangiography in 40%. Simultaneous bouts of new onset diarrhea from cryptosporidia were also reported. If undetected by blood or stool cultures, the next most common organisms are *Microsporidia* species. [252], [253] Other recognized infections include cytomegalovirus (CMV), [254] *Enterocytozoon bieneusi*, [255] *Septata intestinalis*, [256], [257] *Mycobacterium avium–intracellulare*, [258] *Isospora belli*, [259] and possibly HIV infection itself. [258] Hepatic CMV inclusions have been observed in 5% to 44% of patients at autopsy. Among AIDS patients with CMV and cholestatic liver disease, as many as one third have bile duct abnormalities. [254] Ultimately, as many as 50% of patients with AIDS cholangiopathy have no identifiable opportunistic infection. [248], [252], [254], [256] Malignancies such as lymphoma [260] and Kaposi sarcoma [260] have been associated with cholangitis but rarely present as a cholangiopathy. Histological findings in AIDS cholangiopathy observed on bile duct and liver biopsy include moderate to severe inflammation and fibrosis. [261]

Clinical Manifestations

The most common presentation is epigastric or right upper quadrant abdominal pain (64% to 88%) with fever (20% to 65%). Jaundice, however, is distinctly uncommon. Diarrhea is also present in most cases related to small intestinal involvement with the characteristic pathogens. Superimposed bacterial cholangitis is a rare complication. [248], [249], [262] Serum liver biochemistries are noted for elevations in alkaline phosphatase greater than three to five times the upper limit of normal in more than 75% of cases. Milder increases in serum transaminases (less than threefold) are also noted. Normal biochemical parameters have also been observed in 20% of instances with cholangiographic abnormalities. [248], [249], [252], [263] Ultrasonography or CT imaging can detect biliary duct dilatation (intrahepatic or extrahepatic) in most instances. [264], [265] Cholangiography is required for the diagnosis of AIDS cholangiopathy. Left-sided biliary duct system involvement is observed with an increase in intrahepatic duct involvement with saccular dilatations, debris, and pruning. Irregular stricturing of the entire CBD with wall thickening in the absence of dilation is common. [248], [262], [265] The most common characteristic biliary tree manifestation is papillary stenosis with intrahepatic sclerosing cholangitis in 50% of cases. Other variations include intrahepatic and extrahepatic sclerosing cholangitis without papillary stenosis, papillary stenosis alone, intrahepatic sclerosing cholangitis alone, or long (more than 1 cm) extrahepatic strictures. [248], [267], [268] Among individuals with severe pain and associated cholangitis, the identification of papillary stenosis is nearly universal. [248], [263], [269] The presence of isolated CBD strictures should raise the suspicion for primary lymphoma [260] or pancreatic disease. [262]

Differential Diagnosis

The differential diagnosis includes choledocholithiasis, acalculous cholecystitis, periampullary tumors including pancreatic and bile duct cancer, and PSC. Choledocholithiasis will also produce intrahepatic duct dilation above the level of obstruction where biliary duct dilation is distinctly uncommon in AIDS cholangiopathy. The presence of elongated stricturing with intrahepatic and peripheral bile duct dilation is more often observed with periampullary carcinoma but can resemble findings when papillary stenosis alone is present. Direct visualization and biopsy of the ampulla is required to exclude carcinoma. Global duct narrowing, diffuse stricturing, and intrahepatic stone formation characterize PSC. The finding of concomitant IBD also suggests PSC, even in subjects with AIDS.

Treatment

Treatment is directed at identified pathogens as well as biliary tree abnormalities. Antimicrobial therapy against *Cryptosporidia* species with trimethoprim-sulfamethoxazole is indicated, especially among patients with coexistent diarrheal symptoms. [251] Effective treatment against *Microsporidia* species remains elusive. [253] Ganciclovir for CMV [254], [267] and multidrug regimens against *M avium–intracellulare* [258] should also be provided. Albendazole for *S intestinalis* infection has been shown to result in symptom improvement when present. [256] Successful therapy for malignancy-related cholangiopathy with reversal of biliary tract abnormalities has been reported with B-cell non-Hodgkin lymphoma in anecdotal instances. [260] There is no indication for prophylactic treatment in patients unaffected by AIDS cholangiopathy.

Symptomatic patients with a high degree of suspicion for biliary tract disease require cholangiography by ERCP. [247], [248], [262] For patients with papillary stenosis alone, endoscopic sphincterotomy has been associated with improvement in abdominal symptoms. [249], [267], [269] Often, there is little to no change in serum hepatic biochemical parameters in most cases. [262] A continued rise in serum alkaline phosphatase levels, however, suggests the presence of intrahepatic duct involvement as well. [248], [262] The performance of balloon dilation or stent placement in cases of papillary stenosis is without dramatic benefit over sphincterotomy. [262] UDCA therapy for symptomatic disease is not highly efficacious. [270] A diagnosis of acalculous cholecystitis requires cholecystectomy. [271]

POSTTRANSPLANTATION CHOLANGIOPATHIES

Bone Marrow Transplantation

Epidemiology The most frequent etiologies of cholangiopathy within the first 80 days of bone marrow transplantation (BMT) include acute graft-versus-host disease (GVHD), cholangitis lenta, and drug-induced cholestasis. [272], [273] Chronic GVHD is the most common cause beyond 80 days. [273] Risk factors for developing cholestatic-mediated injury after BMT have been identified. The use of conditioning therapies before BMT, including combinations of irradiation and immunosuppressive agents, has most often been associated with hepatic venoocclusive disease (VOD). [274], [275] and [276] Allogeneic donor stem cell recipients are considered to be at increased risk for acute and chronic GVHD. [273] Related sibling donors with identical HLA typing impart the lowest risk for acute or chronic GVHD among all donor source types. [277] The use of immunosuppressive agents for acute GVHD prophylaxis is also associated with an increased risk for cholestatic injury. [272], [278] Antimicrobial prophylaxis against viral and fungal infections in the post-BMT period, [272], [279] although reducing the risk for hepatobiliary infections, may also contribute associated toxicities resulting in liver injury.

Pathogenesis A number of pathophysiologic mechanisms have been implicated in BMT-related cholangiopathies. In cholangitis lenta, the release of bacterial cell wall components, [280] as well as proinflammatory cytokines including IL-1, IL-6, and TNF-a, results in bile flow impairment. [281] Biliary epithelial cells, zone 1 hepatocytes, and periductular glands have been recognized as intrahepatic targets for donor-derived lymphoid cells in acute GVHD. [282] Cell death mediated by apoptosis has also been observed, yet exact mechanisms for this process after BMT are not well understood. [283] Abnormalities in cellular and humoral immunity have been implicated in chronic GVHD. [272] Impaired bile salt and bilirubin transport is associated with hepatotoxicity from cyclosporine. [284] Bile canaliculi injury, hepatocyte necrosis, and elevated serum hepatic biochemistries are observed with supratherapeutic blood levels of cyclosporine. [272], [285] Intrahepatic thromboses of terminal venules and central veins are observed in conjunction with clinical manifestations of VOD. [274]

Clinical Manifestations Acute GVHD occurs exclusively among recipients of allogenic bone marrow and stem cell infusions. The development of a skin rash, evidence for gastrointestinal dysfunction (including nausea and diarrhea), and elevated serum hepatic enzymes are typical manifestations. Hepatic GVHD may also occur independently of skin and the gut manifestations but is uncommon. Average time to onset is about 15 to 21 days after BMT. A delay in time to onset for acute GVHD has been reported with the use of immunosuppressive prophylaxis and non-HLA mismatched grafts. Increases in serum aminotransferase and alkaline phosphatase levels (2- to 10-fold each) with hyperbilirubinemia are characteristic. Decompensated liver disease (ascites, coagulopathy) is uncommon in the early stages of acute GVHD. A rapid increase in hyperbilirubinemia may suggest concomitant sepsis, cholangitis lenta, or VOD. [272] Liver biopsy is often required in this setting for diagnosis. Histological findings of acute GVHD include bile duct epithelial cell necrosis and apoptosis, mild to moderate hepatocyte necrosis, and cholestasis. Ductopenia has been reported in severe cases. [286] The probability of developing chronic GVHD from acute GVHD is estimated at 50% or higher. Progressive hepatic GVHD has been reported as an independent predictor of mortality after BMT. Clinical evidence of portal hypertension is often noted in the absence of histological cirrhosis. [272] Cholangitis lenta is associated with fever and mild hyperbilirubinemia in the early posttransplantation phase and the use of immunosuppressive treatment for acute GVHD. The time interval for developing a preceding infection may be days to weeks. Elevations in serum alkaline phosphatase may be observed, but isolated hyperbilirubinemia is the most common biochemical finding. Total bilirubin levels can reach values as high as 15 mg/dL, and yet hepatic synthetic function is generally intact. Liver histology is noted for cholestasis within pericentral canaliculi and periportal areas in the absence of significant portal inflammation from immunosuppression. [272], [280] Drug-induced cholestasis can occur by idiosyncratic (cyclosporine) [287] or dose-dependent (methotrexate) [272] mechanisms. Among 38 patients receiving cyclosporine at 3 mg/kg/d, [288] an increase in serum bilirubin levels was reported compared with patients given 1.5 mg/kg/d despite therapeutic serum trough levels (less than 400 ng/mL). In doses up to 20 mg/kg/d, most patients treated for acute leukemia developed jaundice and hyperbilirubinemia, which resolved after drug cessation in 2 to 5 days. [289] Azathioprine [290] and fluconazole [291] may be associated with hepatocyte necrosis yet more commonly result in asymptomatic serum aminotransferase elevations. Antibiotics, including amoxicillin–clavulanic acid, [292] flucloxacillin, [293] and trimethoprim-sulfamethoxazole, [294] have been reported in association with cholestatic hepatitis and chronic liver disease in rare instances. Ceftriaxone has been implicated in the pathogenesis of cholelithiasis. [295] Cholestasis is also observed after chronic total parenteral nutrition (TPN). Elevated serum liver biochemistries, including total bilirubin, serum aminotransferases, and alkaline phosphatase, can be attributed to TPN. The association between prolonged TPN and liver failure, however, remains quite rare. Reversal of liver biochemical abnormalities often occurs after TPN discontinuation and resumption of oral intake. Other associated complications in the presence or absence of chronic TPN administration include biliary sludge, cholelithiasis, and acalculous cholecystitis. [296] Chronic GVHD of the liver has been associated with the rapid onset of bile duct destruction and subacute development of intrahepatic cholestasis. It may occur de novo or result from prior acute GVHD. The estimated time to onset after BMT ranges between 40 and 50 days. Hepatic involvement occurs in up to 80% of cases. Biochemical features typically include progressive elevations in serum alkaline phosphatase (as high as 15 times normal) and hyperbilirubinemia. Rapid-onset chronic GVHD may present with elevated serum aminotransferases during tapering of immunosuppressive medications. [272], [273] Histological features on liver biopsy include ductopenia, lymphoplasmacytic portal inflammation, periportal fibrosis, and cholestasis. In select cases, a paucity of bile ducts may be observed (vanishing bile duct syndrome). [272], [286] The development of jaundice, hepatomegaly, and weight gain over 5% of ideal body weight is consistent with the diagnosis of VOD. The average time of onset after BMT is between 15 and 30 days. Persistent jaundice after an episode of acute GVHD can also be from VOD in the setting of fluid retention. Biochemical features include isolated hyperbilirubinemia as the most common presentation. [272], [274], [297] Ultrasonography is commonly negative for biliary tract obstruction or hepatic vein thrombosis. Most patients with VOD have mild involvement and eventual spontaneous resolution. Progressive VOD is associated with liver failure in select cases. [297]

Treatment Prevention of acute GVHD is the most important aspect of treatment because eventual multiple organ involvement is often fatal. Conventional prophylactic strategies have included combination pharmacological therapy with corticosteroids, [272], [276] cyclosporine, [272], [276] methotrexate, [272], [276], [278] tacrolimus, [278] or antilymphocyte antibodies. [278] Depletion of donor T lymphocytes by elution or monoclonal antibody therapy is associated with reductions in the incidence and severity of acute GVHD. Reports of Epstein-Barr virus–associated lymphoproliferative disorder from depletion and elution techniques, however, have also been noted. [302] Systemic corticosteroid therapy (prednisone, 2 mg/kg/d) is the treatment of choice in acute GVHD with hepatic involvement. [298] Treatment failures (defined as persistent skin and gut symptoms) are managed with increasing corticosteroid doses (4 to 8 mg/kg/d) [299] and the addition of tacrolimus [301] or antithymocyte globulin. [300] Empiric treatment of cholestasis-associated pruritus with UDCA may be of benefit in select cases. [303] No specific therapy exists for cholangitis lenta. Treatment of the underlying infection often results in a reduction of hepatic biochemical elevations and improvement in bile flow. [280] Discontinuation of potential offending agents is the initial step in treating drug-induced cholestasis. Improvements in cholestasis and serum hepatic biochemistries occur within 5 to 7 days of cyclosporine discontinuation. [272], [289] Dose reduction of cyclosporine or substitution with other anti–T-lymphocyte agents is recommended in the presence of acute GVHD to minimize worsening bile duct injury. [272] The use of immunosuppressive therapy including prednisone and cyclosporine is required for the treatment of chronic GVHD. Salvage therapy options include tacrolimus, thalidomide, or sirolimus. [272], [273] Empiric UDCA to modulate symptoms from intrahepatic cholestasis may result in serum hepatic biochemical improvement. [303] Mild to moderate cases of VOD are treated by supportive measures. Diuretic therapy may be useful in patents with symptomatic

fluid overload. [272], [274], [297] For severe cases of VOD, the use of thrombolytic therapy has been shown to improve hyperbilirubinemia in up to 30% of patients. The risk for significant hemorrhage among patients with preexisting thrombocytopenia, including a fatality rate approaching 15%, is associated with thrombolytic therapy. [297], [304] Transjugular intrahepatic portosystemic shunt placement to improve sinusoidal portal hypertension has been associated with an increased mortality rate in patients with severe VOD. [305]

Liver Transplantation

Cholangiopathies associated with liver transplantation can involve either intrahepatic or extrahepatic bile ducts. The two most common forms of surgical biliary anastomoses performed in liver transplantation are choledochocholedochostomy and choledochojejunostomy. Factors involved in choosing one type of anastomosis over another include underlying hepatic disease and size match between recipient and donor organ.

Epidemiology Intrahepatic cholangiopathies after liver transplantation primarily involve the biliary epithelium. One condition in liver transplantation associated with bile duct injury is chronic allograft rejection. Histological criteria for chronic allograft rejection include the loss of intralobular bile ducts in 50% or more identified portal tracts. [306] Recurrent allograft PSC occurs in up to 20% of cases in which strict criteria are applied to exclude other potential causes of intrahepatic biliary disease. [226], [227] and [228] Features of biliary obstruction including cholangitis and fibroobliterative lesions have been reported in allograft biopsies of PSC patients. [228] Extrahepatic cholangiopathies after liver transplantation include anastomotic strictures, hilar strictures, ischemic-type bile duct strictures, bile leaks, choledocholithiasis, and sphincter of Oddi dysfunction. The overall incidence of biliary complications ranges from 9% to 15%, and complications usually develop 2 to 6 months after liver transplantation. Anastomotic strictures occur in 3% to 7% of cases and may be early or late. [306], [307], [308] and [309] Hilar strictures after liver transplantation are associated with local ischemia or hepatic artery thrombosis. Prolonged cold ischemia time and ABO incompatibility have also been reported as risk factors. [310], [311] Bile leaks occur in up to 25% of liver transplantation recipients. [307], [308] Sites of injury include the biliary anastomosis, T-tube insertion site, reduced-size parenchymal graft cut edge, or unrecognized collateral bile duct. [307] Bile leaks associated with the choledochojejunostomy anastomosis occur at the reconstruction site in the early posttransplantation period. [309] T-tube removal in association with choledochocholedochostomy anastomosis is reported to constitute more than 50% of all bile leaks. [307], [308] The finding of choledocholithiasis after liver transplantation is generally associated with a biliary stricture. [307] Sphincter of Oddi dysfunction has been recognized after choledochocholedochostomy reconstruction. Cholangiography is noted for a dilated CBD tapering to its origin. [307], [308]

Clinical Manifestations Patients with intrahepatic or extrahepatic cholangiopathies after liver transplantation often present with asymptomatic elevations in serum alkaline phosphatase or total bilirubin. Mild elevations in serum transaminases can occur as well. Alternatively, the presence of fever or right upper quadrant abdominal pain may be consistent with bacterial cholangitis. [307], [308] and [309] Jaundice and pruritus are uncommon but can be associated with chronic ductopenic rejection. [306] Imaging studies with ultrasound or CT are sensitive for the detection of intrahepatic duct dilation. Examination of hepatic arterial blood flow by Doppler ultrasonography is mandatory in the setting of biliary stricture to exclude thrombosis and stenosis. [310], [311] If a T tube is present, cholangiography is performed to determine the location and extent of biliary strictures when suspected. [307], [308] and [309] ERCP has developed into the initial method of cholangiography in liver transplantation patients for diagnostic and therapeutic interventions. [312] The use of PTC may be indicated when distal bile duct dilation is not significant or evidence for a proximal biliary obstruction above the bifurcation is observed by imaging studies. [307], [314] Histological examination of the allograft by liver biopsy is warranted if cholangiography is unremarkable. [306]

Treatment Ensuring adequate immunosuppression in the presence of chronic ductopenic rejection is mandatory. Conversion from cyclosporine to tacrolimus (FK506) is often recommended because resulting bile flow impairment and cholestasis reduces effective gastrointestinal absorption of cyclosporine. [306] Treatment options for recurrent PSC deal primarily with stricture dilation with or without long-term biliary tube stenting. [228] Hepatic retransplantation in this population is the only available definitive solution if allograft failure ensues. [228], [313] Surgical conversion to a choledochojejunostomy for biliary strictures associated with the choledochocholedochostomy anastomosis was previously considered the treatment of choice after liver transplantation. [307], [309] Recent advances in endoscopic and radiologic techniques have demonstrated satisfactory results from balloon dilation, stenting, or both. Patients who fail to respond after these nonoperative measures may then be considered candidates for biliary anastomosis reconstruction. The use of expandable metal stents may also be considered in subjects who are poor candidates for surgery after liver transplantation. [312], [314], [315] Focal areas of stricturing in association with indwelling T tubes, often related to postoperative edema, usually resolves with observation. [307] The correction of aberrant hepatic artery flow (by angioplasty or surgical revision) may be beneficial to prevent further bile duct injury and diffuse stricturing in the early postoperative period. Hilar strictures are difficult to manage by operative intervention based on previous manipulation of the porta hepatis. Interventions with balloon dilation and percutaneous indwelling stent placement are often required. Problems with recurrent biliary obstruction from debris and mucosal sloughing (especially when ischemic cholangiopathy is present) require frequent biliary stent manipulations and changes. [315] The efficacy of bilateral endoscopically placed stents (polyurethane and expandable metal) compared to percutaneous drainage is unknown. Diffuse biliary stricturing can initially be managed with nonoperative methods, including balloon dilation followed by stenting of dominant strictures. [307], [315] Similar to hilar strictures, the occurrence of stent occlusion has limited the long-term success of this strategy. [315] No benefit with UDCA or prophylactic antibiotic therapy for the prevention of biliary stricturing has been shown. Hepatic retransplantation in eligible candidates is often required for definitive therapy. [313] The management of choledocholithiasis is similar to non–liver transplantation scenarios with the use of balloon dilation and stone removal by ERCP or PTC. Endoscopic sphincterotomy may or may not be required. [307], [312] Sphincter of Oddi dysfunction, when confirmed using accepted criteria, is also treated by endoscopic sphincterotomy despite the absence of abnormal manometric findings. [307]

IATROGENIC CHOLANGIOPATHIES

Operative Bile Duct Injury

Epidemiology Operative bile duct injury is most commonly associated with cholecystectomy (90% of instances). The incidence of bile duct injury with open cholecystectomy is estimated between 0.1% and 0.2%, which includes minor duct (of Luschka) injuries. [316] Initial reports of bile duct injury from laparoscopic cholecystectomy in 1% to 2% of cases have now been reduced to frequencies equivalent to those of open cholecystectomy given the accumulation in clinical experience. [317] Risk factors for bile duct injury include limited operative experience, acute or chronic inflammation involving the biliary tree and gallbladder fossa, and aberrant anatomy. [318] A classification system for bile duct injuries has been proposed by Strasberg and colleagues. [318] Type A injuries affect the minor ducts without the loss of biliary tree continuity, including the cystic duct and gallbladder fossa. Type B and C injuries involve aberrant right hepatic duct injuries. Occlusion of the cystic duct in this scenario is termed a type B injury, whereas complete cystic duct transection is termed a type C lesion. Type D injuries involve incomplete lacerations of the primary bile ducts (common bile, common hepatic, left and right hepatic). Evolution of these lesions to type E injuries can occur. Type E injuries include five variations with lesions located between the common hepatic duct and above the bifurcation. Type A, D, and E bile duct injuries are most often reported.

Clinical Manifestations The timing and extent of injury often determine clinical presentation. Bile duct injuries are often not recognized at the time of surgery. The median time to symptoms is 5 to 7 days but may be weeks. Fever, abdominal pain, and wound drainage can be observed. Jaundice may represent distal CBD occlusion or transection. A minor proportion of patients are asymptomatic at presentation. Imaging studies, such as hepatobiliary scintigraphy (HIDA scan), can document the existence of bile leakage in 90% to 95% of cases. Identification and localization of bile duct injuries by cholangiography is required before repair. [317], [318] Late complications, including biloma formation and biliary stricture, may occur. [319]

Treatment Endoscopic or percutaneous methods to treat bile duct injuries have been employed with success. These measures are effective in the treatment of active bile leaks as well as benign postoperative drainage strictures. Endoscopic sphincterotomy with or without NBT or stent placement is equally effective for bile leak closure. Removal of stents in 3 to 4 months with repeat cholangiography to document bile leak closure is recommended. The disconnection between distal and proximal bile duct systems requires PTC for diagnosis and therapy. [320] Injuries noted at surgery require immediate repair. Techniques include suturing of linear lacerations, end-to-end anastomosis, formation, and Roux-en- biliary reconstruction. Damage involving the common hepatic duct bifurcation is treated with biliary-enteric reconstructive techniques. Facilitation of surgical repair can be provided by preoperative stent placement. Continued stent placement for up to 3 to 6 months is advised. [320] Long-term results from surgical bile duct repair are good and associated with the nature and extent of injury, type of surgical reconstruction performed, and center experience. [319]

Secondary Sclerosing Cholangitis

The development of cholangiographic changes similar to PSC have been reported as secondary phenomena resulting from complications of therapeutic interventions. The most well-known cause of secondary sclerosing cholangitis (SSC) is related to hepatic artery infusion with floxuridine (FUDR). Prevalence rates for FUDR-associated SSC remain unchanged between 17% and 56%. Involvement of the common hepatic duct bifurcation with sparing of the CBD is characteristic. The arterial supply of extrahepatic bile ducts is derived primarily from the gastroduodenal arcade, which is often excluded from hepatic artery infusion. Ischemia, rather than direct FUDR toxicity to biliary epithelium, appears to be the underlying mechanism of action. [321] Although injury appears to be contained within the biliary system,

liver failure associated with FUDR-induced SSC is related to coexistent obstructive vasculopathy. [320] The injection of 20% formaldehyde with sodium chloride or ethanol for the treatment of hydatid disease has also been associated with SSC. [322] Direct sclerosant-type effects on biliary epithelia is the proposed mechanism of action.

MISCELLANEOUS CHOLANGIOPATHIES

Biliary Stricture from Chronic Pancreatitis

Epidemiology Biliary strictures from pancreatic disease occur based on anatomic proximity between the distal CBD and posterior pancreas. The presence of fixed structures, including pancreatic cysts and masses, are associated with extrinsic CBD compression. Biliary stenosis associated with the inflammation of acute and chronic pancreatitis is usually transient. Chronic biliary obstruction is noted among 4% to 10% of patients with alcoholic pancreatitis. Biliary stricture after operative therapy for chronic pancreatitis is observed between 60% to 70% of cases. [323], [324]
Clinical Manifestations In chronic pancreatitis, there are no specific clinical features that suggest the presence of biliary obstruction. Serum hepatic biochemistries are noted for elevations in serum alkaline phosphatase without similar degrees of hyperbilirubinemia. Mild serum aminotransferase (78%) and amylase (47%) elevations can occur as well. Among patients with postoperative bile duct strictures, as many as 80% of cases are identified within the first postoperative year. Late manifestations may occur years after initial surgery. [323], [324] Among 987 patients with benign postoperative biliary strictures followed over 25 years, the most common symptoms at presentation were fever (64%), abdominal pain (49%), and jaundice (44%). Elevations in serum alkaline phosphatase and bilirubin occurred in 50% of cases. [325]
Diagnosis Ultrasonography and cross-sectional imaging techniques (CT, MRI) are useful for documenting biliary dilatation or biloma formation in select cases. Cholangiography (ERCP or PTC) can define both location and extent of stricture involvement. [323], [324] Smooth, long stricturing of the distal CBD between 2 and 4 cm in length is most characteristic. Hepatic histology can also identify biliary obstruction in 80% of cases involving chronic alcoholic pancreatitis with cirrhosis occurring in 29% of patients. [324]
Course and Complications Complications associated with biliary stricture from chronic pancreatitis include cholangitis, choledocholithiasis, and secondary biliary cirrhosis. As many as 20% of reported patients have developed biliary cirrhosis or liver failure after biliary stricture formation from operative injury. [324]
Treatment In cases of biliary stricturing from chronic pancreatitis, the optimal management has yet to be defined. Surgical decompression is employed for high-grade obstruction complicated by jaundice or cholangitis. [325] Endoscopic approaches with stricture dilation and stenting have been more recently performed. Nearly 25% of patients experience stricture regression after stenting for up to 3 months, whereas incomplete responders require operative decompression. [326] Among patients with abnormal serum hepatic biochemistries and no symptoms, the pursuit of operative therapy is controversial. Liver biopsy features of progressive biliary fibrosis favor surgical decompression.

Biliary Fistulas

Spontaneous Enteric Biliary Fistula

Epidemiology. Prevalence rates between 0.5% and 5% after biliary tract operations have been reported. Gallstones are responsible for 90% of cases, followed by peptic ulcer disease (6%) and trauma (4%). Cholecystoduodenal (75%), cholecystocolonic (15%), and cholecystogastric (6%) fistulas are the most frequent sites of tract formation. Cholecystoduodenal fistulas from gallstones almost exclusively involve the periampullary area. Fistulas may be obstructive or nonobstructive. The female-to-male ratio for developing both fistula types is 2:1 to 3:1. Gallbladder carcinoma has been associated with enteric biliary fistulas in 15% of cases. [327]
Clinical manifestations. Nonobstructive fistulas are often diagnosed as incidental findings at cholecystectomy or cholangiography. Consequences of these fistulas include ascending cholangitis, weight loss, and bleeding. Obstructive fistulas present with symptoms reminiscent of bowel obstruction, which may be complete or partial. [327] Gallstone ileus can present with acute small bowel obstruction from gallstones of diameters greater than 25 mm. The ileocecal area is the most common site of obstruction (75%), followed by the jejunum (20%). Abdominal x-ray findings of pneumobilia are observed in only 30% of enteric biliary fistulas. Biliary reflux of oral contrast is more suggestive of biliary fistula. [328]
Treatment. The treatment of choice when possible is surgical. For gallstone ileus, the performance of cholecystectomy, fistula excision, and operative cholangiography to exclude choledocholithiasis is recommended. Mortality rates approaching 20% in previous reports have now been reduced below 5%. [328]
Operative Biliary Fistula Most commonly observed in association with bile duct injury after cholecystectomy, the development of a cutaneous or internal bilioperitoneal fistula occurs in 33% of postoperative cases. Hepatobiliary scintigraphy (i.e., HIDA scan) can detect bile leaks and fistulas with greater than 90% sensitivity. Cholangiography may ultimately be required for diagnosis confirmation. Endoscopic therapy with NBT placement or stents has been successful in selected cases. Treatment failures require surgical intervention. [329]
Bronchobiliary Fistula The overall prevalence rates of bronchobiliary fistula are unknown but range between 4% and 10.5% among patients with hepatic and subphrenic abscesses, respectively. Other etiologies responsible for bronchobiliary fistula formation include blunt abdominal trauma, bile duct stenosis, choledocholithiasis, and congenital causes. Clinical manifestations include bilioptysis, jaundice, and cholangitis. The diagnosis has been made by cholangiography and hepatobiliary scintigraphy. The treatment of choice is surgical. [330]
Miscellaneous Biliary Fistula Vascular fistulas with the biliary tree may occur and include both arterial and venous systems. Causes include trauma, infection, or iatrogenic (operative, transjugular intrahepatic portosystemic shunt) complications. Hemobilia is the most common sign at presentation. Biliobiliary fistulas involving the gallbladder, cystic duct, or secondary biliary radicle with the main biliary tree can also occur. Compression by stone impaction in the cystic duct or gallbladder neck (Mirizzi syndrome) may result in erosion through the CBD, resulting in fistula formation. [331]

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CHAPTER 102

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CYSTIC DISEASES OF THE BILIARY TRACT

Cystic dilations occur throughout the biliary system. Most common are cystic dilations of the common bile duct (CBD) (i.e., choledochal cysts), but cystic dilations involving the intrahepatic ducts alone or in combination with other cystic abnormalities of the extrahepatic ducts are increasingly recognized. The terminology and classification are tremendously confused. The term *choledochal cyst* has commonly referred to all cystic abnormalities of the biliary tree; however, this term is best restricted to only cystic abnormalities of the CBD. Despite the many variations of these cystic abnormalities, some underlying features unify them.

EPIDEMIOLOGY

Biliary cysts are rare, with about 4000 cases reported worldwide. [1] The incidence of biliary cysts is much higher in Japan, where one half to two thirds of all reported cases have originated. [1], [2] and [3] The Japanese incidence (1 in 13,000 births) is about 150 times more common than in the West (1 in 2,000,000 births). [3] The incidence is much higher among women, with a female-to-male ratio of 3:1 or 4:1. Between 40% and 60% of patients are diagnosed before 10 years of age, 52% to 76% before 20 years of age, and 83% to 90% before 30 years of age. [1], [2], [3] and [4] This is mainly a disease of children and young adults.

CLASSIFICATION

Todani and associates [5] proposed the most complete and practical classification that recognizes five types of bile ducts cysts (Fig. 02-1).

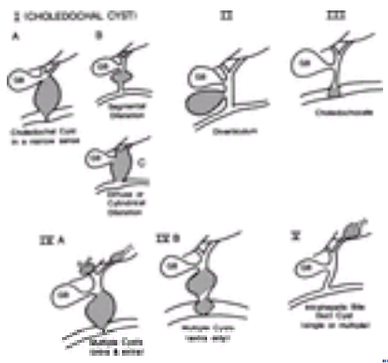

FIGURE 102-1. Todani's classification of biliary cysts based on location. *Hatched areas* represent cystic dilations.

Type I cysts include choledochal cysts in a narrow sense, segmental choledochal dilations, and diffuse or cylindrical dilations. Seventy-five to 85% of cases are type I.
Type II or diverticulum cysts are found anywhere in the extrahepatic ducts and make up 2% to 3% of reported cases.
Type III or choledochocele cysts, also rare, represent 1.4% to 5.6% of reported cases. In 1989, Sarris and Tsang [6] reviewed 48 cases in the English literature and proposed a further anatomic classification of choledochoceles. In type A, the ampulla opens into the choledochocele, which communicates with the duodenum through another small opening. Type A choledochoceles can be further subclassified into A1, in which the pancreatic duct and CBD share a common opening into the cyst (33% of cases); A2, in which the openings are distinct (4% of cases); and A3, in which the choledochocele is small and entirely intramural (25% of cases). In type B, the ampulla opens directly into the duodenum, with the choledochocele communicating only with the distal CBD (21% of cases). This scheme is useful because of important therapeutic implications.
Type IV cysts are also subclassified. Type IVA multiple cysts in the intrahepatic and extrahepatic ducts account for 18% to 20% of reported cases. [3] Type IVB multiple cysts occur only in the extrahepatic duct and are much less common.
Type V intrahepatic bile duct cyst (single or multiple) are also quite rare. They probably represent Caroli disease as originally described.

Serena and colleagues [7] proposed a type VI biliary cyst, which is cystic dilation of the cystic duct with otherwise normal intrahepatic and extrahepatic ductal systems. Three cases of type VI cysts have been described in the literature up to 1999. [8]

Anatomically based classification is of great practical importance in planning surgical management. Other classifications emphasize clinical presentation rather than ductal anatomy.

PATHOGENESIS

The cause of biliary cysts has been disputed. The major debate concerns whether cysts are congenital or acquired. The possibility of genetic factors has also been raised. First, this disease is much more common in Japanese than in Europeans. Familial occurrence has been reported in one pair each of mother and daughter, mother and son, father and daughter, and a pair of sisters. However, a study of a pair of monozygotic twins showed discordance for both anomalous pancreaticobiliary junction and biliary cyst. [9] Thus, this is not compatible with single gene control.

Many early investigators, including Douglas and Alonzo-Lej, favored the congenital malformation theory. [10] Several observations support this theory. First, biliary cysts appear in all age groups, and antenatal diagnosis has been reported as early as 15 weeks' gestation, making a congenital origin likely. There are also many associated anomalies of the biliary system, such as double CBD, double gallbladder, multiseptate gallbladder, biliary atresia, congenital hepatic fibrosis, and annular pancreas. [11] In addition, many cysts fail to shrink after decompressive surgery, suggesting inherently abnormal ductal wall development. [2]

Of the theories proposing acquired cysts, that of Babbitt and associates [12] has received the most attention. They proposed that choledochal cysts develop because of an anomalous arrangement of the distal pancreaticobiliary tree. In all seven of their patients with choledochal cysts, they found that the CBD entered the pancreatic duct at a right angle at an abnormally long distance (more than 2 cm) from the ampulla of Vater (Fig. 102-2). This abnormal anatomy impaired normal sphincteric function at the pancreaticobiliary junction. Because the maximal secretory pressure of the pancreas is 30 to 50 cm H_2O and that of the liver is 25 to 30 cm H_2O, pancreatic juice flows into the bile duct in the absence of a sphincter mechanism. This anomaly is associated with recurrent cholangitis, edema, fibrosis, obstruction of

the bile ducts, and carcinoma. These complications are believed to result from chronic reflux of pancreatic juice and subsequent activation in bile of various pancreatic proteinases and lipases. In support of this are findings of high levels of amylase that are frequently found in the cyst fluid and the demonstration that pancreatic juice in the CBD of animals with an anomalous arrangement can cause cystic dilation of the CBD. [4], [13] Although amylase has been found in the cyst fluid, this enzyme probably does not play a pathogenetic role. Amylase is not detectable in the duodenal fluid of newborns and infants, but trypsin activity is the same in newborns and older children. [14] In addition to increased amylase activity in bile, Shimada and colleagues [15] also showed markedly increased concentration and proportion of lysophosphatidylcholine in bile (especially in gallbladder) of patients with anomalous pancreaticobiliary ductal junction, most of whom had biliary cyst. This was due to increased hydrolysis of phosphatidylcholine, a normal component of bile, by increased activity of phospholipase A_2. Because lysophosphatidylcholine is cytotoxic, this may play a role in the pathogenesis of biliary cyst by adversely affecting an already weakened bile duct wall and in the later development of malignant degeneration.

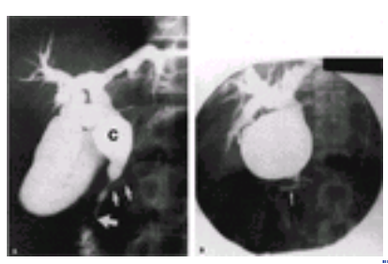

FIGURE 102-2. Choledochal cyst—anomalous ductal relationship. **A:** Cystic enlargement of the common bile duct (*C*) with associated dilation of the common hepatic and intrahepatic ducts. The abnormal junction of the pancreatic duct (*small arrows*) and common bile duct and long common channel (*large arrow*) is displayed. **B:** A large cyst involves the common hepatic and common bile duct accompanied by intrahepatic ductal dilation. A long common channel (*arrow*) results from the abnormally high junction with the pancreatic duct.

Anomalous pancreaticobiliary ductal anatomy occurs in 65% to 80% of patients with choledochal cysts. [10] The anomalous pancreaticobiliary ductal union is not unique to patients with biliary cysts; it is also associated with biliary malignancies (especially gallbladder carcinoma) and abnormal pancreatic ducts. [16] In a large study of 1325 patients examined with direct cholangiography for symptoms suggestive of hepatobiliary and pancreatic disease, 29 patients had the anomaly (2.2%), of whom 21 patients had associated choledochal cyst. Surprisingly, the frequencies of gallbladder carcinoma in patients with the anomaly were 50% and 5% in the absence or presence of associated choledochal cyst, respectively. None of the patients with gallbladder carcinoma had gallstones. The overall frequency of carcinoma of the biliary tract was 14% in patients with choledochal cyst and 50% in those without. This striking difference led the authors to recommend prophylactic cholecystectomy in patients with anomalous pancreaticobiliary ductal junction. [17] Similar findings have been reported by Sugiyama and Atomi. [18] Based on these studies, the authors concurred with the recommendation of prophylactic cholecystectomy.

Many investigators emphasized that cystic dilation results from a combination of two factors: segmental weakness of the duct wall and distal obstruction. [19] The weakness of the wall and the distal obstruction may be congenital or acquired. Several animal experiments support the importance of distal obstruction. [2], [20] Ligation of the distal CBD in adult rabbits, rats, and dogs gives rise to diffuse dilation of the intrahepatic and extrahepatic ducts. This also occurs in neonatal lambs. [20] After CBD ligation, all six studied lambs developed choledochal cysts, and all four adult sheep developed isolated dilation of the gallbladder.

The etiologic importance of these distal ductal abnormalities is unclear. The animal experiments may not be applicable to humans. The distal obstruction theory also would not explain solely intrahepatic duct cysts. Not all choledochal cysts have demonstrable distal obstruction.

The infantile form of choledochal cysts appears in some cases to result from an obliterative process similar to that producing biliary atresia. Twenty-four (13%) of 188 patients with biliary cysts were also found to have biliary atresia. [21] However, well-documented cases of acquired biliary cysts in adults have appeared, including seven attributed to recurrent attacks of pancreatitis. [22] More recent studies suggest many other factors may also be involved. One is the finding of abnormal sphincter of Oddi pressure in patients with choledochal cysts. [23] Other studies have added viral infection and genetic mutations to possible causes of choledochal cysts. [24], [25] and [26] Tyler and coworkers found reovirus in 13 of 23 tissues from patients with extrahepatic biliary atresia and 7 of 9 tissues from patients with choledochal cysts, prevalences that were much higher than in patients with other hepatobiliary diseases (21%) or autopsy cases (12%). [24] This is intriguing because reovirus can produce a disease in weanling mice with similar pathological features to human choledochal cysts. [24] However, the number of patients with choledochal cysts examined was small, and causality remains to be established. Two cases of choledochal cysts in association with rare genetic syndromes have been described recently. [25], [26] One was in association with familial adenomatous polyposis syndrome [25] and the other in association with Simpson-Golabi-Behmnel syndrome. [26] These investigators raised the possibility that mutations in their respective genes may have contributed to the development of the choledochal cyst.

Two recent studies advanced our understanding of the pathogenesis of Caroli syndrome (type V biliary cysts in association with congenital hepatic fibrosis; see section on Clinical Presentations). First is the identification of impaired protein *N*-glycosylation due to phosphomannose isomerase deficiency as a cause of congenital hepatic fibrosis, a disorder of ductal plate malformation often associated with type V cysts. [27], [28] These patients often present with protein-losing enteropathy, which responds to D-mannose treatment. [28] Whether the hepatobiliary abnormalities will improve is unknown. These recent findings also suggest that normal protein glycosylation is important in the formation of ductal plates. Another advance is the development of a novel animal model with features of polycystic kidney, multiple intrahepatic bile duct dilations, and congenital hepatic fibrosis. [29] These animals exhibit increased proliferative and decreased apoptotic activities in the biliary epithelial cells during development as compared with controls, suggesting that the remodeling defect in immature bile ducts associated with the imbalance of cell kinetics might play a role in the development of the hepatobiliary abnormalities. [29] This animal model should provide important insights into the pathogenesis of this syndrome.

It is clear that many factors are important in the pathogenesis of biliary cysts, which encompass a wide spectrum of lesions.

CLINICAL PRESENTATIONS

Type I or Choledochal Cysts

Patients with choledochal cysts present with two typical clinical constellations determined mostly by the patient's age at presentation. [2] In infancy, jaundice with or without acholic stools is the most common finding, occurring in as many as 80% of patients. [2] The presentation is often indistinguishable from biliary atresia, which is sometimes associated with choledochal cysts. [21] Pain may or may not be a factor, but vomiting and failure to thrive have been reported in as many as 50% of patients. [2] Hepatomegaly is often found, and 30% to 60% of patients have a palpable abdominal mass. [2] The mass is usually in the right hypochondrium; is soft, elastic, round, and mobile laterally; and may follow the movements of the diaphragm. [5] The classic clinical triad of pain, jaundice, and a palpable abdominal mass has been found only in 11% to 63% of large series. [2] Frequently, biliary cirrhosis and portal hypertension complicate the management. [21]

The noninfantile form (patients older than 2 years of age) covers a wide age range, with the oldest patient reported in the eighth decade. Asymptomatic patients have been described. [30] Chronic and intermittent pain appears to be the most common presenting symptom, reported in 50% to 96% of patients. [31] Intermittent jaundice and recurrent cholangitis are also common, reported in 34% to 55%. Abdominal mass is much less common; it occurs in about 10% to 20% of cases. The classic triad of pain, jaundice, and a palpable mass has been reported in 3% to 13% of patients. [31] Cirrhosis and portal hypertension are less often encountered. Recurrent pancreatitis has been reported only in the noninfantile form, although whether there is actual pancreatic inflammation is debatable. [32] When these patients present with painful attacks, they often show slight elevation of serum bilirubin levels and an increase in the degree of the choledochal dilation, possibly caused by biliary obstruction. The terms *fictitious pancreatitis* and *pseudopancreatitis* have been applied to these patients. [32] One possible explanation is that amylase in the biliary tract has ready access to the bloodstream. This would also explain why hyperamylasemia is almost never observed in patients younger than 1 year of age with choledochal

cysts: the amylase levels are usually very low in bile because acinar growth of the pancreas is not complete until 1 to 2 years of age. [32]

The other presentation that has been reported only in patients older than 10 years of age is that of carcinoma associated with choledochal cysts. [33]

Type II or Diverticulum Cysts

More than 50 cases of type II or diverticulum cysts have been reported in the world literature, representing 2% to 3% of large series. [31], [34] Presenting symptoms generally reflect the compression of nearby structures by the cyst.

Type III or Choledochocele Cysts

About 125 cases of choledochocele have been reported. [31], [35], [36] and [37] More than 73% of reported patients were older than 20 years of age, unlike with type I cysts, which are found in 60% of the patients with biliary cysts younger than 10 years of age. [36] There is also less female preponderance, with a female-to-male ratio of 1.4:1. [36] Common presenting symptoms include intermittent episodes of abdominal pain and obstructive jaundice. Abdominal mass is not found. Pancreatitis is much more common in choledochocele than in any other type of biliary cyst and is reported in 30% to 70% of these patients, presumably caused by reflux of bile or obstruction. [37] Stones in the cele or the CBD are also much more common, reported in 25% to 35% of these patients, in contrast to only 8% in type I cysts. [31], [37] Rarely, intestinal (duodenojejunal) intussusception may be the presenting feature of choledochocele. [35]

Type IV and V Cysts or Caroli Disease

Caroli and associates [38] originally described this disease as a congenital malformation of intrahepatic bile ducts, characterized by segmental cystic dilation of the intrahepatic ducts; increased incidence of biliary lithiasis, cholangitis, and liver abscesses; absence of cirrhosis and portal hypertension; and association of renal tubular ectasia or similar renal cystic disease.

After the first report, Caroli [39] recognized that two distinct disease entities are associated with intrahepatic duct cysts: the simple type, which was his original description, and the periportal fibrosis type. The simple type is associated with medullary sponge kidney in 60% to 80% of the cases. The periportal fibrosis type is also known as *Caroli syndrome*. In addition to intrahepatic cystic dilation, congenital hepatic fibrosis, cirrhosis, portal hypertension, and esophageal varices are frequently seen. Hepatic function is usually well preserved in the periportal fibrosis type; the major clinical manifestations are recurrent cholangitis, liver abscess formation, and portal hypertension. It is often associated with the renal abnormalities of autosomal recessive polycystic kidney disease. [27]

It is now apparent that Caroli disease, including the simple and periportal-fibrosis types, represents only part of the spectrum of cystic disease of the intrahepatic ducts. In an excellent review by Barros and associates [40] of 46 well-documented cases with hepatic histopathology and biliary tree studies available, only 13% of patients had the simple type. Thirty-five percent had the periportal-fibrosis type, 22% had intrahepatic and extrahepatic cystic dilation (i.e., type IVA of Todani's classification), and 30% had all three abnormalities: cystic dilation of the intrahepatic and extrahepatic bile ducts and congenital hepatic fibrosis.

The term *congenital hepatic fibrosis* refers to a unique congenital liver histology characterized by bland portal fibrosis, hyperproliferation of interlobular bile ducts within the portal areas with variable shapes and sizes of bile ducts, and preservation of normal lobular architecture. [40] The prevalence of biliary cysts in patients with congenital hepatic fibrosis probably has been underestimated because most patients did not have cholangiographic studies performed.

The aforementioned forms of intrahepatic biliary cystic disease should be differentiated from polycystic liver disease because of significant dissimilarities. Polycystic liver disease is an autosomal dominant genetic defect, but the manner of inheritance of Caroli disease remains poorly defined. [27] Polycystic liver cysts are large, contain serous fluid, and do not communicate with the biliary tree. Caroli disease cysts communicate with the biliary tree. Portal hypertension is rare in polycystic liver disease. The prognosis for polycystic liver disease patients is determined by the effect of cystic disease on the kidneys and not the liver. [41] The incidence of malignant transformation in the intrahepatic parenchymal cysts of polycystic liver disease is 1.3%, compared with 7% in Caroli disease. [42]

Until 1984, 162 cases of Caroli disease had been reported. [42] Males and females are equally affected, unlike the female predominance in choledochal cysts. Symptoms usually appear in early adult life, with recurrent episodes of fever, chills, and abdominal pain due to cholangitis. More than 80% of patients present before 30 years of age. Biliary lithiasis is found in 34% of these patients and may predispose to recurrent cholangitis. [40] Occasionally, the disease is diagnosed later in life, with the sequelae of portal hypertension, most commonly bleeding esophageal varices. [39], [42]

DIAGNOSIS

Diagnosis of biliary cysts requires a high index of suspicion. The major reason for missed diagnosis is that biliary cysts are usually not included in the differential diagnosis of recurrent abdominal pain, pancreatitis, obstructive jaundice, or cholangitis. Most patients do not present with the classic triad of pain, jaundice, and abdominal mass. Biochemical tests only reflect the degree of biliary obstruction and infection. Correct diagnosis depends on the use of imaging studies.

Ultrasonography is the best screening method for type I, II, IV, and V cysts. [1], [2] More than 17 cases of antenatal diagnosis of choledochal cyst by means of ultrasonography have been reported. [43] When a choledochal cyst is suspected on prenatal ultrasonography, repeat ultrasonography is indicated immediately after birth so that expeditious treatment can be provided. The major drawback to ultrasonography is that it provides little anatomic or functional information.

Hepatobiliary scintigraphy has been advocated as a sensitive, noninvasive tool for the diagnosis of choledochal cyst. This technique is useful for all except type III cysts. [44] The characteristic findings are biliary tract dilation and tracer retention after 24 hours. [44] It provides information about excretory patterns and has therefore been recommended by many for postoperative patient follow-up.

Ultrasonography and scintigraphy are complementary and provide a sound basis for preoperative diagnosis in patients younger than 2 years of age. For older patients, computed tomography has been advocated by some to be superior to ultrasound. [44], [45] Until recently, percutaneous transhepatic cholangiography (PTC) and endoscopic retrograde cholangiopancreatography (ERCP) have been regarded as providing the best detailed examinations. PTC is especially useful in defining complex anatomic cysts of the intrahepatic ducts, but ERCP provides the best visualization of the distal portion of the biliary tree, especially the relation of the biliary and pancreatic ducts. [2] This notion has been challenged in recent years by improvements made in magnetic resonance imaging. [46], [47] Several studies have shown that magnetic resonance cholangiopancreatography (MRCP) is as good as ERCP in the evaluation of biliary cysts, especially in adults. [46], [47] Because MRCP is noninvasive and not operator dependent, it is likely to replace ERCP and PTC as the imaging study of choice in patients with biliary cysts.

ERCP is likely to remain useful in the diagnosis of type III cysts (i.e., choledochoceles). The typical features are the "clubbed" appearance of the distal CBD and a round, cystlike, contrast-filled structure in the terminal CBD, which often protrudes into the duodenal lumen. Emptying of contrast material is often delayed. Choledochoceles are easily differentiated from duodenal diverticula and duodenal duplication cysts by filling during cholangiography but not during upper gastrointestinal contrast studies (Fig. 102-3). Duodenal diverticula fill on upper gastrointestinal series but not on cholangiography. Duplication cysts do not fill with either method. [48]

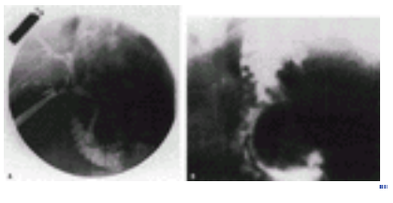

FIGURE 102-3. Choledochocele—type III cyst. **A:** Cholangiography reveals a club-shaped enlargement of the distal common bile duct bulging into the duodenum. **B:** Upper gastrointestinal series reveals a smoothly rounded filling defect in the second portion of the duodenum.

Oral cholecystography and intravenous cholangiography are of no use in diagnosing biliary cysts. [3], [31] Other nonspecific findings include a mass displacing the gas shadows on plain abdominal radiographs and displacement of antrum, usually anteroinferiorly and to the left, on upper gastrointestinal series.

The definitive diagnosis of biliary cyst and its classification is secured by intraoperative cholangiography at the time of operative intervention. Delineation of the precise anatomy is critical in preventing injury, especially to the pancreatic duct because of the high frequency of the anomalous union and in providing the best operative management possible. [2]

PATHOLOGY

The wall of the cyst is usually thickened because of productive fibrosis and inflammation. In histology, dense connective tissue, with fibrocollagenous and sometimes smooth muscle and elastic elements, is seen. There is no epithelial lining, but islets of cylindrical or columnar epithelium may be preserved. A case of biliary papillomatosis arising in a choledochal cyst wall was reported. [49] Duodenal mucosa covers choledochoceles externally, but the internal surface of choledochoceles is variably lined by duodenal mucosa (63% of cases), bile duct mucosa (21% of cases), or unclassified glandular epithelium (13%). [50]

Liver biopsy is abnormal in 60% of patients with biliary cysts, demonstrating biliary cirrhosis, portal fibrosis, or evidence of biliary atresia. Other abnormal findings include liver abscesses, especially in cases of Caroli disease, and sludge or stones in the CBD. In newborn patients with biliary cysts, the histology is usually normal or shows mild bile duct proliferation consistent with biliary obstruction. [10]

TREATMENT

Biliary cyst treatment is surgical, and primary cyst excision is the procedure of choice. [5] Internal drainage procedures, such as choledochoduodenostomy and Roux-en- choledochojejunostomy, were widely employed, especially in Western countries. [2], [22] There are three major drawbacks to internal drainage procedures compared with cyst excision. First, the morbidity rate is much higher, up to 88% with internal drainage procedures compared with 2% to 8% with cyst excision. [21] The usual complications are recurrent pain, jaundice, stricture formation, and cholangitis. These appear to be more common after choledochoduodenostomy than after Roux-en- choledochojejunostomy. [2], [21] The second drawback is the need for reoperation because of the high complication rate. In large series, 13% to 70% of patients have required reoperation after internal drainage, but a 0% to 10% reoperation rate was reported after cyst excision. [21], [51] The third drawback is the risk for malignant transformation, which is not diminished after an internal drainage procedure (see the section on Clinical Course and Complications). Patients who develop biliary carcinoma after excision of choledochal cyst are extremely rare; only three were reported up to 1984. [52] The fundamental reason for poor outcome after internal drainage is that diseased tissue remains and is used for biliary reconstruction. The cyst wall is inherently inflamed and fibrous, precluding normal healing. Biliary stasis persists and predisposes to recurrent cholangitis, stone, and stricture formation.

The aforementioned techniques are applicable to choledochal cysts (i.e., types I and IV cysts involving the CBD). Type II cysts should be excised. Treatment of type III cysts depends on the anatomy (see Fig. 102-1). Sarris and Tsang [6] recommend excision of the duodenal lumenal portion of the cyst, leaving the medial portion containing the ampulla intact as the preferred treatment in most cases of type IIIA cysts (i.e., A1 and A2). Transduodenal sphincteroplasty and ERCP with papillotomy have been advocated as treatment for type IIIA3 cysts. Type IIIB cysts should be treated by excision and sphincteroplasty. The malignant potential of choledochocele appears to be very low. [36], [37] However, Schimple and colleagues [53] pointed out that carcinoma has only been reported in choledochoceles that were lined by biliary or undifferentiated epithelium internally (up to 20% in some reports). They advocate complete excision of the choledochocele by separation and reinsertion of the common bile duct and pancreatic duct into the duodenal wall if the choledochocele is lined by biliary or undifferentiated epithelium, whereas sphincteroplasty should be reserved only for those lined by duodenal epithelium.

Intrahepatic cyst (i.e., type IVA or V) treatment depends on the degree of involvement. When segmental cystic disease is confined to one lobe (more often the left lobe), lobectomy is usually curative. [42], [54] If both lobes are involved, some have advocated establishing a permanent-access hepaticojejunostomy after partial hepatectomy to allow easy biliary tree access when necessary. [54] It is uncertain how effective this approach will be. If extrahepatic cysts coexist with intrahepatic cysts (type IVA), they should be excised as previously described. Chronic antibiotic therapy in multilobar Caroli disease may have some benefit. Ultimately, if attacks of cholangitis are frequent and quality of life poor, hepatic transplantation may be a therapeutic option. A summary of the recommended surgical treatments is given in Table 102-1.

TYPE OF BILIARY CYST	SUGGESTED TREATMENT
I	
A	Cyst excision with Roux-en-Y hepaticojejunostomy or jejunal interposition hepaticoduodenostomy
B	
C	Choledochojejunostomy or sphincteroplasty if excision is impossible
II	Excision if possible
III	
A_1	Excision
A_2	
A_3*	Sphincteroplasty or endoscopic retrograde cholangiopancreatography with papillotomy
III B	Excision and sphincteroplasty
IV	
A	Extrahepatic cyst excision with biliary reconstruction as in IA and IB. Lobectomy if intrahepatic cyst is confined to one lobe, or permanent access hepatocojejunostomy if intrahepatic cysts are diffuse. Transplantation in selected cases.
B	Cyst excision as in IA, IB
V	Hepatic resection for localized disease or permanent access hepaticojejunostomy for diffuse disease. Transplantation may be an option.

* Complete excision of the choledochocele by separation and reinsertion of the common bile duct and pancreatic duct into the duodenal wall if the choledochocele is lined by biliary or undifferentiated epithelium.

TABLE 102-1 Suggested Treatment for Biliary Cysts

Cholecystectomy should be performed at the time of cyst excision because continuous free bile flow minimizes the risk for reflux and cholangitis and because the gallbladder has shown clinical and histological evidence of cholecystitis in patients in whom secondary operations have been necessary, and the gallbladder is predisposed to malignant change in patients with biliary cysts. [55]

If the definitive surgical procedure is successful, the prognosis is generally excellent. Reversal of cirrhosis has even been reported. [56]

CLINICAL COURSE AND COMPLICATIONS

Complications of biliary cysts include recurrent cholangitis before treatment and after internal drainage procedures, stone formation, stenosis and stricture, pancreatitis, biliary cirrhosis, portal hypertension due to cirrhosis or portal vein thrombosis, liver abscess, especially in Caroli disease, and cyst rupture. [6], [21], [57] Pregnancy can precipitate or aggravate symptoms of choledochal cyst, specifically cyst rupture. Twenty-six cases have been reported to rupture during pregnancy or labor. [58] Pregnant women with symptomatic choledochal cyst should avoid labor by having cesarean sections as soon as the fetus is mature.

The most feared complication is malignancy. [55], [59] Carcinoma afflicts predominantly adults. The youngest patient with carcinoma during initial choledochal cyst surgery was 10 years of age. [59] The mean age for detecting carcinoma after enteric drainage was 35 years, with a mean interval of 10 years after the enteric drainage procedure. [59] The mean age for detecting carcinoma at the initial surgery for biliary cyst was 50 years; this is two decades earlier than the mean age for bile duct

carcinoma in the general population. [59] Even though the overall incidence of biliary carcinoma in association with biliary cysts is 2.5%, it is actually 14% to 18% in the adult patients (older than 20 years of age) and 50% by 50 years of age. [2], [33], [59] In contrast, the overall incidence of biliary carcinoma is 0.012% to 0.48% in the general population. [60] The female-to-male ratio is 2.5:1, slightly less than that seen with choledochal cysts.

Carcinomas occur not only in the cyst wall but also in the remainder of the hepatobiliary and pancreatic tree. Of 154 cases of carcinoma in cases of biliary cysts, only 58% were actually in the cyst wall, 40% were gallbladder, 1.3% were pancreatic, and 0.6% were intrahepatic carcinomas. [59]

The pathogenesis of malignant change in biliary cysts is unknown. Stones are not thought to be important because they are found only in 13.8% of patients [59] (Fig. 102-4). Adenocarcinoma is the most common type, constituting 70% to 84% of all types. Other histological types are squamous (4% to 9%) and undifferentiated or anaplastic (7% to 21%), and one case of small cell carcinoma was reported. [59]

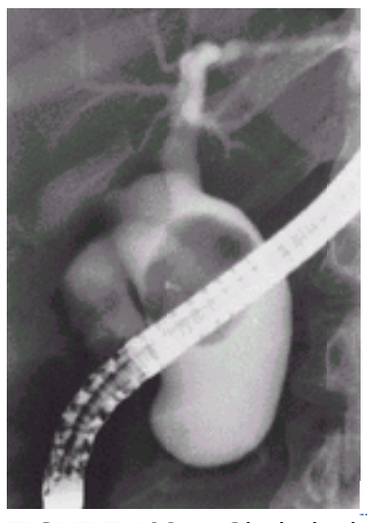

FIGURE 102-4. Choledochal cyst—type IA. Endoscopic retrograde cholangiogram shows cystic dilation of the common bile duct with a very large filling defect due to a calculus. Stones are uncommon complications of biliary cysts, reported in only 13.8% of cases (Courtesy of Dr. Hartley Cohen, USC School of Medicine, Los Angeles.)

Isolated intrahepatic cysts (i.e., Caroli disease) are also associated with a higher incidence of carcinoma. Overall, 7% of patients with Caroli disease were found to have cholangiocarcinoma, with no female predominance. [61] The average age at the time of diagnosis of carcinoma was 51 years. [61]

The prognosis of carcinoma related to biliary cysts is dismal. Almost all patients die soon after diagnosis. [59], [61] One preventive therapy is total cyst excision during the initial operation. However, cyst excision does not completely eliminate the future risk for carcinoma because the rest of the pancreaticobiliary tree appears to also be predisposed to malignant change.

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CHAPTER 103

Jeffrey A. Drebin and Steven M. Strasberg

TUMORS OF THE BILIARY TRACT

CHOLANGIOCARCINOMA

Cholangiocarcinoma is cancer of biliary epithelium. It may arise from intrahepatic or extrahepatic bile ducts. Sporadic cholangiocarcinoma is quite uncommon, but cholangiocarcinoma has a high incidence in the presence of certain benign conditions. Information on cholangiocarcinoma is mostly derived from small case series gathered over many years and therefore tends to be descriptive. Consequently, claims for the best form of investigation or treatment must be interpreted cautiously.

Classifications

There are several suggested classifications. Clinically, intrahepatic and extrahepatic cholangiocarcinomas are distinct entities (Table 103-1) but are linked by cell of origin and etiologic factors. Intrahepatic cholangiocarcinoma arises from small ducts or ductules and presents as an intrahepatic mass, the differential diagnosis of which usually is other intrahepatic tumors such as hepatomas. Extrahepatic cholangiocarcinoma arises from large ducts and usually presents as biliary tract obstruction. The extrahepatic tumors may be further subdivided into upper duct tumors (also called perihilar, hilar, or Klatskin tumors) and lower duct tumors. [1] The division into upper and lower duct cholangiocarcinomas is logically based on staging and treatment considerations. Lower duct tumors are similar to other periampullary tumors, such as adenocarcinomas of the head of the pancreas, in terms of diagnosis and treatment.

NAME	SYNONYMS
Intrahepatic	Small duct, intraparenchymal, peripheral[a]
Extrahepatic—upper duct	Hilar cholangiocarcinoma, perihilar cholangiocarcinoma, Klatskin tumor, proximal[a] bile duct cancer
Extrahepatic—lower duct	Distal[a] bile duct cancer

Note: The classification is the authors'. Intrahepatic and extrahepatic are the main division in the classification per the TNM system. Extrahepatic tumors are divided into two rather than three anatomic zones per ref. 1.

[a] These terms are undesirable, particularly "proximal" and "distal." Distal means away from the center of the body and proximal means near. Note that in pancreatic terminology, distal refers to the part of the duct farthest away from the termination in the duodenum; whereas in biliary terminology, the opposite is true. The terms are properly applied to limbs, and their use in biliary-pancreatic terminology should be abandoned.

TABLE 103-1 Classification of Cholangiocarcinoma

Extrahepatic cholangiocarcinoma has been classified, for staging purposes, by a classification introduced and modified by Bismuth [2] into four types based on the level of ductal involvement (Fig. 103-1). Cholangiocarcinomas are also classified by the TNM system, [3] which groups intrahepatic cholangiocarcinomas with other intrahepatic malignancies and considers extrahepatic cholangiocarcinomas as a separate group (Table 103-2). A potentially useful modification of the staging system for intrahepatic cholangiocarcinomas has recently been proposed. [4]

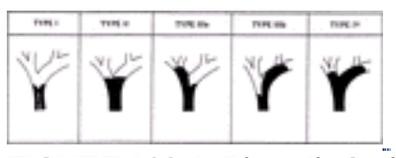

FIGURE 103-1. Bismuth-Corlette classification of hilar cholangiocarcinomas.

TABLE 103-2 TNM Classification for Cholangiocarcinoma and Gallbladder Carcinoma

Incidence, Etiology, and Pathogenesis

The true incidence of bile duct carcinoma is difficult to determine because reports of incidence and survival statistics from these cancers are usually included with liver and gallbladder cancers or pancreatic carcinomas. Recent reports suggest that rates of intrahepatic cholangiocarcinoma may be increasing in Great Britain [5] and the United States. [6] The incidence peaks at about 65 years of age, and the tumor is more common in men than women.

Sporadic cholangiocarcinoma is very uncommon. The tumor usually arises in association with an underlying condition. Underlying conditions are of two types: those that have a high frequency in localized populations, such as parasitic infestation with *Opisthorchis viverrini* in Northeast Thailand; and those that have a much lower frequency but a more global distribution, such as primary sclerosing cholangitis (PSC) (Table 103-3). In Northeast Thailand, liver fluke–associated cholangiocarcinoma is a leading cause of death, [7] whereas in the United States, cholangiocarcinoma accounts for less than 1% of malignancies. The diverse predisposing causes of cholangiocarcinoma usually result in chronic inflammation of the biliary tract.

Predisposing causes with a high incidence in localized areas of the world
Infestation with *Opisthorchis viverrini* in Northeast Thailand
Infestation with *Chlonorchis sinensis* in China
Intrahepatic stones
Predisposing causes with a low but more global incidence
Primary sclerosing cholangitis
Choledochal cysts
Thorotrast[*]
Ulcerative colitis

[*]A number of other agents have been claimed to be associated with cholangiocarcinoma, but the evidence is weak.

TABLE 103-3 Predisposing Causes of Cholangiocarcinoma

Opisthorchis species– and *Chlonorchis* species–related cholangiocarcinoma are caused by eating raw fish [7] containing the larval stage of flukes. Because eating raw fish is more common among males in affected populations, liver fluke–associated cholangiocarcinoma is more common in males. The risk for cholangiocarcinoma in people infested with *Opisthorchis* species is related to the degree of infestation, as measured by stool egg count. [8] Infestation is associated with increased nitrate levels in body fluids, which is reversible upon treatment with praziquantel. [9] There is suggestive evidence from human [9] and animal [10] studies that *N*-nitroso compounds may be the carcinogens involved. Liver fluke infestation is also associated with development of intrahepatic stones. Intrahepatic gallstones are also associated with an increased risk for cholangiocarcinoma. [11] Cholangiocarcinoma may develop even after stone removal, if bile duct fibrosis induced by the stone disease leads to stasis and infection. [11]

Cholangiocarcinoma occurs in about 10% of patients with PSC. [12] Also, cholangiocarcinoma occurs 10 times more frequently in patients with chronic ulcerative colitis than in the general population. [13] PSC and ulcerative colitis often coexist; in such patients, as might be expected, the incidence of cholangiocarcinoma is even higher. [14] Cholangiocarcinoma also occurs in about 10% of patients with untreated choledochal cyst disease. [15], [16] Malignancy of the bile ducts and gallbladder in patients with choledochal cysts appears to be related to the presence of an anomalously high pancreatic duct–bile duct junction. This is discussed in the section on Carcinoma of the Gallbladder. Thorotrast-related cholangiocarcinoma is now largely a disease of the past because the agent has not been used since about 1940.

Molecular Genetics Studies in recent decades have shown that cancer results from the accumulation of multiple genetic abnormalities in a single cell. [17] This process of multistep carcinogenesis involves genes that control cell cycle progression and apoptotic cell death. Many of the specific genetic events responsible for neoplastic transformation in colon and pancreatic malignancies have been identified. [18], [19] Relatively less is known about the events responsible for neoplastic transformation in the biliary tract. The *erbB2* oncogene (also called the *HER2* oncogene or the *neu* oncogene) encodes a cell surface protein that interacts with several distinct cellular receptors, including the receptor for epidermal growth factor. [20] Overexpression of *erbB2* has been identified in tumors of the breast, ovary, stomach, pancreas, lung, and prostate. Several studies have examined expression of *erbB2* in cholangiocarcinomas. [21], [22] and [23] Although an early report suggested that expression of *erbB2* was present in less than 10% of cholangiocarcinomas, [21] studies from several other laboratories have identified *erbB2* expression in 67% to 73% of cholangiocarcinomas. [22], [23] Overexpression of the epidermal growth factor receptor has also been identified in some cholangiocarcinomas. [24] The *ras* genes constitute a distinct family of oncogenes that transduce proliferative signals from cellular receptors to the cell interior. [25] One member of this family, termed K- *ras* has been shown to be mutated in a significant fraction of colon, pancreas, and lung carcinomas. Several studies have demonstrated K- *ras* expression in cholangiocarcinomas. [24], [26], [27] and [28] Point mutations involving this gene may occur in more than half of all cholangiocarcinomas. [27], [28] Several other genes, including the *myc* gene [29] and *p53* gene, [30] have been studied in cholangiocarcinomas. These genes encode proteins that reside in the cell nucleus and control transcription of growth regulatory genes. Expression of the *myc* gene appears to be an almost universal feature of cholangiocarcinomas, [23] whereas *p53* expression has been identified in 50% to 80% of cholangiocarcinomas. [30], [31], [32] and [33] The studies cited here have just begun to characterize the genetic events responsible for the development of cholangiocarcinoma. It is interesting that the genes that have been linked to cholangiocarcinoma thus far, particularly *erbB2* K- *ras* and *p53* also appear to be involved in a high percentage of pancreatic carcinomas. [19] It is possible that this reflects exposure to common carcinogens in the biliary and pancreatic epithelium.

Pathology

Malignant tumors of the bile ducts are almost exclusively epithelial tumors (carcinomas). Tompkins and associates [34] reported that the distribution of extrahepatic bile duct cancers is 49% in the upper third, 25% in the middle third, 19% in the lower third of the duct; 7% of these cancers were diffuse. The Johns Hopkins group reported a distribution of 6% intrahepatic; 67% extrahepatic, upper duct (“perihilar”); and 27% extrahepatic, lower duct (“distal”). [1] It must be stressed that all such figures arising from surgical case series are heavily influenced by referral patterns and may not be representative of the disease generally.

Grossly, extrahepatic bile duct cancers can be divided into three types: polypoid or nodular masses, sclerosing, and diffusely infiltrating. Some cancers may present as only a thickening of the bile duct wall, which appears to be involved in dense fibrous scar. Polypoid or papillary cancers have the best prognosis.

Histological variants of adenocarcinomas of the bile duct include well-differentiated, pleomorphic, giant cell, adenosquamous, oat cell, and colloid carcinoma. Bile duct cancers have a tendency to spread along the bile duct. The longitudinal extent of cancers along the bile duct is often difficult to determine by gross examination. Shimada and colleagues [35] examined 29 cases by histological examination after resection and found that the mean distance of microscopic invasion beyond the gross margin was 16.8 mm toward the liver and 6.5 mm toward the duodenum.

Histological diagnosis of malignant bile duct cancers may be difficult at times. Well-differentiated cancers with little invasion are difficult to differentiate from the bile duct involved in scar formation or PSC. Cancers arising in the upper duct tend to be better differentiated with more sclerotic stroma that contains chronic inflammatory cells. [36] Perineural invasion (i.e., malignant cells within nerve sheaths), a prominent feature of bile duct cancers, is not present in PSC and is the single most important diagnostic criterion of malignancy. Other problems in differential diagnosis occur with cancers of the lower bile duct that involve the ampulla, duodenum, and pancreas. Because all of these sites may give rise to malignant tumors with similar characteristics, it may be difficult to determine site of origin conclusively. An important point is that bile duct cancers, as opposed to pancreatic adenocarcinomas, are somewhat less likely to metastasize widely. As a result, aggressive treatment of bile duct tumors is associated with a somewhat more favorable outcome.

Hilar tumors often arise in one of the main bile ducts and produce asymptomatic obstruction of that duct before progressing to obstruct the other main duct, which leads to jaundice. Unilateral obstruction results in atrophy of the affected side of the liver and hypertrophy of the other side. This phenomenon will also occur if there is unilateral portal vein occlusion. The clinical importance of atrophy has been emphasized by Blumgart and Stain. [37] Atrophy may result in axial rotation of structures in the hepatoduodenal ligament. For instance, with atrophy of the left hemiliver and hypertrophy of the right, the structures rotate in a clockwise direction when viewed from below, which hides the hepatic artery, rotates the bile duct to the left, and exposes the portal vein on the right side of the hepatoduodenal ligament in the usual

position of the bile duct. The effect of atrophy must be taken into account when adding liver resection to extrahepatic duct resections for cholangiocarcinoma.

Cholangiocarcinomas must also be differentiated from benign inflammatory tumors, [38] also called *hepatic inflammatory pseudotumors* and *benign fibrosing disease*. [39] These inflammatory masses mimic extrahepatic and intrahepatic cholangiocarcinomas but consist of chronic inflammatory cells and fibrosis. Benign inflammatory tumors appear to occur most frequently in extrahepatic upper ducts but also occur intrahepatically, [40] and less commonly in lower ducts. In a recent series of upper duct "tumors," this inflammation accounted for 13% of cases. [39] The failure to appreciate this diagnosis may lead to inappropriate therapies, such as long-term stenting or hepatic resection.

Bile duct cancers usually spread by direct extension to involve adjacent organs and tissues. Upper duct cancers may directly invade the liver. Cancers in the common bile duct or cystic duct may directly invade the gallbladder. Regional lymph node metastases are common, liver metastases distant from the primary occur occasionally, but distant metastases are unusual.

Intrahepatic cholangiocarcinomas grossly may be well or poorly demarcated, single or multiple. Mucin production, fibrosis between the acini of tumor tissue, and a more overtly glandular pattern are the main differentiating characteristics from hepatoma. Immunohistochemical staining for specific cytokeratins may also be helpful. Unlike hepatomas, some cholangiocarcinomas stain positively for CA 19-9 or carcinoembryonic antigen (CEA). It may be very difficult to distinguish between a liver metastasis of colonic or pulmonary origin and an intrahepatic cholangiocarcinoma.

Clinical Presentation

Extrahepatic upper duct and extrahepatic lower duct tumors are not usually separable on the basis of presenting symptoms. Most patients present with painless jaundice and its clinical constellation of dark urine, light stool, and pruritus. Nonspecific gastrointestinal symptoms such as anorexia and nausea, as well as mild weight loss and fatigue, are not unusual. Mild abdominal pain without jaundice may be the sole presenting symptom in upper duct cholangiocarcinoma; many patients with upper duct cholangiocarcinoma have had recent cholecystectomy, presumably because it was believed that gallstones were the source of these symptoms. Cholangitis is uncommon unless the biliary tree is instrumented. On examination, patients are usually jaundiced, and in the case of tumors arising below the insertion of the cystic duct, the gallbladder may be palpable.

Laboratory tests suggest extrahepatic biliary obstruction. Characteristically, the bilirubin rises over several weeks to a level of 20 mg/dL if the jaundice is unrelieved and the direct reacting fraction accounts for more than 50% of the total bilirubin. Marked elevation of serum alkaline phosphatase and ?-GT levels and mild elevation of transaminase levels are also usual.

Cholangiocarcinoma in patients with PSC is often associated with deterioration in clinical status and liver function tests. However, these signs are unreliable and late markers of cholangiocarcinoma. Cholangiocarcinoma may be present without any change in traditional liver function tests. [41] Furthermore, screening of patients at risk, such as those with PSC, using computed tomography (CT) scanning, ultrasound, and even endoscopic retrograde cholangiopancreatography (ERCP) is ineffective. [42] CA 19-9 is emerging as a better diagnostic marker. The Mayo group has reported that a cutoff of 100 IU was 89% sensitive and 86% specific for cholangiocarcinoma in PSC. [43] The King's College group recommend the use of an index that incorporates both CEA and CA 19-9 values and that has attained similar sensitivities for cholangiocarcinoma in PSC. [42] Bile CEA levels are reportedly elevated in cholangiocarcinoma, but not in benign diseases, other than intrahepatic stones. [44]

Intrahepatic cholangiocarcinomas characteristically present with nonspecific right upper quadrant pain. Other symptoms include weight loss and nausea. An intrahepatic cholangiocarcinoma may occasionally be critically situated in the central area of the liver and, as a result, may cause jaundice owing to obstruction of the main ducts by external pressure.

Diagnosis and Staging

Extrahepatic, Lower Duct Tumors The most common cause of painless jaundice or jaundice with mild pain in the cancer age group is pancreatic carcinoma. Consequently, both lower and upper duct cholangiocarcinomas are often initially suspected to be pancreatic carcinoma. The question of which is the best initial investigation of painless jaundice is controversial. Ultrasound, ERCP, and CT scan have all been advocated. Ultrasound has the advantages of low cost and lack of invasiveness, and it is the best test for detection of choledocholithiasis and cholecystolithiasis. In young patients with any pain or in older patients with pain that has characteristics of biliary colic, ultrasound is probably still the best first examination. Conversely, in the typical patient with painless jaundice, CT scan may be the best initial investigation. This is so because in most instances, when a periampullary mass is detected without evidence of metastatic spread, CT scan is the only investigation needed before proceeding to surgery or staging laparoscopy with ultrasound. When no mass is present on CT scan, a diagnostic ERCP is performed to determine the site and type of biliary obstruction.
Staging laparoscopy with ultrasound. Preoperative staging of the tumor is also of great importance. Ideally, staging locates and sizes all tumors in the body. The immediate goal of staging for the surgeon is to determine whether the tumor has spread beyond the limits of operative resection; this may be due to distant metastases or to the local extent of the tumor. For staging purposes in lower duct cholangiocarcinoma, *local extent* means the macroscopic limits of the tumor in relation to the portal vein and superior mesenteric artery. The value of staging laparoscopy with ultrasound in hepatobiliary-pancreatic malignancies has recently become evident. The importance of laparoscopy in staging hepatobiliary-pancreatic malignancy was demonstrated by Warshaw's group [45] and by John and associates. [46] We have found that additional staging information, proving that resection for cure was not possible, was obtained by this technique in 22 of 50 patients with hepatobiliary-pancreatic malignancies, who had been completely evaluated by conventional studies. [47] Of the remaining 28 patients, 26 were actually resectable for cure at the time of surgery. Staging lap- aroscopy permits evaluation of the peritoneal surfaces without laparotomy, and frequently metastatic implants are found. Laparoscopic ultrasound can detect vascular and distant nodal invasion, which makes the tumor inoperable [47] (Fig. 103-2). Others have found endoscopic ultrasonography useful in determining the extent of tumor involvement. [48] The role of positron emission tomographic scanning in staging cholangiocarcinoma is still investigational. [49], [50]

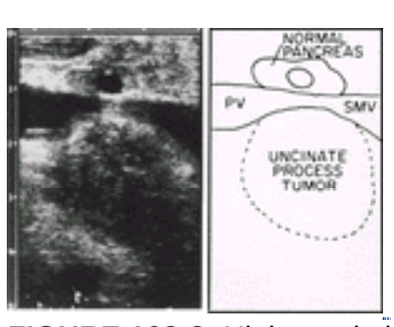

FIGURE 103-2. High-resolution laparoscopic ultrasound image demonstrating a 2.5- to 3-cm inhomogeneous hypoechoic mass in the uncinate process consistent with a pancreatic cancer. The tumor abuts the superior mesenteric vein (*SMV*) and portal vein (*PV*) posteriorly behind the neck of the pancreas, which itself appears to be normal. No evidence of actual vascular invasion is seen. At laparotomy, this adenocarcinoma was resected. (From ref. [47].)

Extrahepatic Upper Duct Tumors Altemeier and colleagues [51] and Klatskin [52] wrote classic papers on perihilar cholangiocarcinoma in an era in which the biliary tree was inaccessible to preoperative imaging. Consequently, both stressed the difficulty of diagnosing this tumor. Today, there are many imaging techniques that can display the tumor or its effects on adjacent vessels, including transabdominal ultrasound, CT scan, CT with angioportography, magnetic resonance imaging (MRI), percutaneous cholangiography (PTC), ERCP, positron emission tomography, angiography, endoscopic ultrasound, and laparoscopy with laparoscopic ultrasound examination. Diagnosis is usually based on history and imaging characteristics. Any patient who has a perihilar stricture, without evidence of ductal disease elsewhere in the biliary tree suggestive of PSC, and who has not had previous biliary surgery that might have resulted in stricture, is considered to have upper duct cholangiocarcinoma. Inflammatory pseudotumor is not ruled out by these considerations, and a confident diagnosis depends on pathological examination of tissue obtained at surgery or cytologic brushings obtained by ERCP or PTC. In a recent report covering 74 patients with biliary and pancreatic strictures, the sensitivity and specificity of brush cytology were 56% and 100%, and the positive predictive value was 100%. [53] Therefore, brush cytology is diagnostic and very useful when positive but of little value when negative. Because perihilar tumors are less accessible than lower duct or pancreatic neoplasms, it is unlikely that a sensitivity of 50% is being achieved in these tumors. Mucobilia on ERCP is an uncommon finding but highly suggestive of a papillary cholangiocarcinoma, which may be intrahepatic or

extrahepatic. [54] Fine-needle aspiration (FNA) is also useful if the mass can be seen on ultrasound examination or on CT scan. Another technique that we have used with success is to direct the biopsy needle at a stent placed through the site of duct narrowing. Staging is also of great importance in upper duct tumors. For staging purposes, *local extent* means the macroscopic limits of the tumor in relation to the biliary tree, especially the upper level of the stricture, and whether and to what degree invasion of adjacent vessels has occurred. The upper level of the stricture provides the information for the Bismuth stage. Blumgart [37] has described the indicators of unresectable bile duct cancers: bilateral intrahepatic bile duct spread, involvement of the main trunk of the portal vein, involvement of both branches of the portal vein or bilateral involvement of the hepatic artery and portal vein, or a combination of vascular involvement on one side of the liver with extensive bile duct involvement on the other. With vascular replacement, it may be possible to resect some cholangiocarcinomas previously considered unresectable. The local extent of the disease along the biliary tree has usually been determined by direct cholangiography—either ERCP or PTC (Fig. 103-3). Because the upper margin of disease is of most interest in these tumors, PTC has been generally favored over ERCP. However, today, ERCP has progressed to the point that a complete picture of the biliary tree can often be obtained even when the obstruction is complete. Magnetic resonance cholangiography is a new, less invasive method of evaluation of the biliary extent of perihilar cholangiocarcinoma. Although promising, it is too early to determine whether it will routinely provide the detail obtained by direct cholangiography (Fig. 103-4).

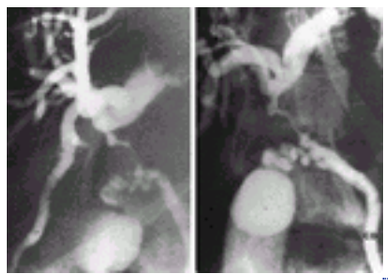

FIGURE 103-3. Cholangiogram shows a Klatskin tumor. The intrahepatic bile ducts are dilated and the size of the common bile duct (*CD*) is normal.

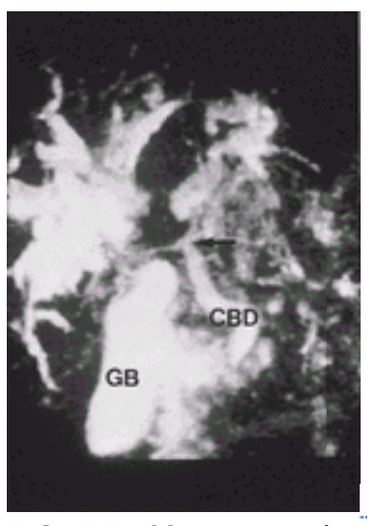

FIGURE 103-4. Magnetic resonance cholangiogram showing extrahepatic and intrahepatic biliary tract. Narrowing of bile duct produced by a hilar tumor is shown (*arrow*). GB, gallbladder; CBD, common bile duct. (Courtesy of Dr. Joseph T. Ferrucci, Department of Radiology, Boston University School of Medicine.)

Vascular invasion has been assessed by angiography, color-flow Doppler ultrasound, and MRI. Angiography is the gold standard for assessing vascular involvement but is also the most invasive. Our preference is for color-flow Doppler ultrasound because the tumor and its extravascular extent, as well as its effect on vessels, are seen in real time, and multiple detailed views of the tumor and its relationships may be obtained. Vascular deviation without narrowing is not evidence of unresectability. Color-flow Doppler ultrasound is very dependent on the skill of the ultrasonographer and on equipment. It has been claimed that Doppler ultrasound is effective in evaluating portal vein involvement (Fig. 103-5) but less so in examining the hepatic artery. [55]

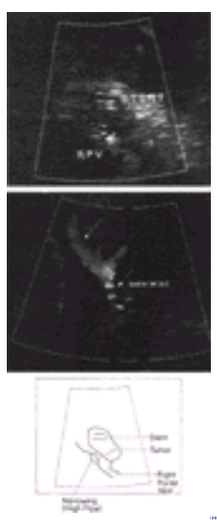

FIGURE 103-5. Flow Doppler ultrasonograms showing encasement of right portal vein by tumor. A stent that had been placed through the tumor can also be seen. Note that in the area of vein encasement, the color is yellow and blue, indicating higher-velocity flows than the red areas, in which the vein is not compressed.

Additional information on local extent of disease in or around the biliary tree can be obtained by laparoscopy with laparoscopic ultrasound, which permits close contact between the ultrasound probe and the tumor. [47] The best position for the probe is often the anterior surface of the liver, looking through a 2- to 4-cm thickness of liver into the porta hepatis. Using this method, we have been able to identify tumor extending along the intrahepatic ducts above the level that was evident by the upper level of the biliary stricture identified on direct cholangiography. Vascular encasement and lymph node involvement may be detected (Fig. 103-6). Bilateral biliary involvement to the point that all four sectional ducts [47] are involved precludes curative resection.

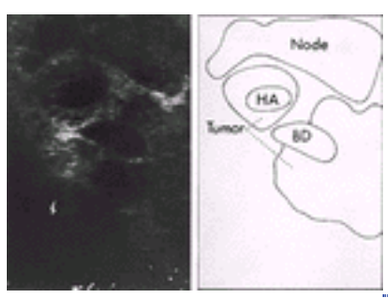

FIGURE 103-6. Laparoscopic ultrasonogram showing encasement of the hepatic artery (*HA*) and bile duct (*BD*). An enlarged lymph node that contains tumor is also seen.

Several factors preclude dogmatic statements regarding selection and order of investigation in extrahepatic upper duct cholangiocarcinoma. There are no good comparative studies examining this issue; there are so many potentially valuable investigations, and further developments in imaging are occurring so rapidly that it is

unlikely that good data will be realized; equipment and expertise vary from center to center; patients are often referred with several studies already completed; upper duct cholangiocarcinoma is often at presentation not distinguishable from lower duct cholangiocarcinoma or other malignancies causing complete obstruction of the lower duct, and consequently, initial investigation cannot be directed specifically to the perihilar region. The important considerations are that a combination of investigations must be used to achieve the goals of staging—evaluating the local extent of disease and whether it is metastatic—and that clinicians have a variety of algorithms to choose from to achieve staging. Our approach is given below.

Summary of our present algorithm for staging in extrahepatic cholangiocarcinoma. In the typical case of painless jaundice, a CT scan is obtained first. If there is a periampullary mass, the next step is staging laparoscopy with ultrasound unless therapeutic decompression of the biliary tree by ERCP or percutaneous stenting is indicated (see later). If no mass is seen, ERCP with brushings is performed when ductal dilation extends into the lower bile duct or when there is no ductal dilation. Either ERCP or PTC cholangiography with brushings is performed when only intrahepatic ducts are dilated, suggesting an upper ductal lesion. In the case of lower duct lesions, detection of a single stricture by ERCP is taken as evidence of a periampullary cancer, possibly cholangiocarcinoma, and the next investigation is staging laparoscopy with ultrasound. In the case of upper duct strictures, compatible with hilar cholangiocarcinoma, the next step is to evaluate vascular involvement by color-flow Doppler ultrasound; FNA is done at this time. In some cases, MRI is useful to assess liver invasion or vascular involvement not clearly identified on CT or flow Doppler ultrasound. Staging laparoscopy with ultrasound then follows for those patients who are still potentially resectable. The utility of staging laparoscopy in the operative assessment of extrahepatic cholangiocarcinomas has been recently validated in a large series from the Memorial Sloan Kettering Cancer Center. [56]

Intrahepatic Tumors Intrahepatic cholangiocarcinoma is in the differential diagnosis of the intrahepatic mass. Except for events such as mucobilia or tumor embolizing into the extrahepatic bile ducts, an event that may cause pain, jaundice, or even pancreatitis [57] (Fig. 103-7), intrahepatic cholangiocarcinoma does not produce highly characteristic findings. On axial imaging, the tumor may be single or multiple. Neither CT or MRI appears superior in characterizing these rare lesions. [58] On CT scan, the lesion is usually irregular or unencapsulated and of low attenuation with only mild enhancement seen with contrast. [58] The tumors are low attenuation on CT angioportography. [59] On T1-weighted MRI, the intrahepatic cholangiocarcinoma is usually of low intensity, but on T2-weighted images, they are of high intensity. [58], [60] Local vascular invasion is often seen by CT, MRI, [60] or angiography, [61] and ductal dilation is often present peripheral to the tumor. Centripetal filling in after gadolinium administration has been noted. [60] Liver biopsy is useful when focal nodular hyperplasia is likely on the basis of imaging studies, or the tumor is inoperable because of metastatic spread or extent in the liver. Otherwise, in potentially resectable cases, the next investigation after axial imaging by either CT or MRI is staging laparoscopy with ultrasound as for other hepatic masses. [46], [47]

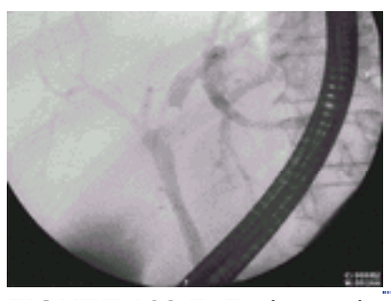

FIGURE 103-7. Endoscopic retrograde cholangiogram showing a bullet-shaped filling defect in the left hepatic duct. At surgery, this was found to be a tumor embolus from an intrahepatic cholangiocarcinoma. This patient presented with pancreatitis, presumably caused by such an embolus.

Preoperative decompression of the biliary tree in jaundiced patients. Three randomized studies have shown that jaundiced patients do not benefit from preoperative decompression of the biliary tree. [62], [63] and [64] However, these studies were performed in an era in which the most common surgical procedure performed after preoperative decompression was palliative surgical bypass of the biliary tree—a relatively short procedure associated with limited morbidity. No study has examined the potential benefit of preoperative decompression when the following operative procedure is either pancreaticoduodenectomy or liver resection. A recent study found that operative morbidity was greater in patients with hilar cholangiocarcinoma who had an aspartate aminotransferase level of more than 90 IU than in patients with lower levels. [65] Until definitive studies are available, preoperative decompression is still an option to be considered. In our opinion, it should be done if hemihepa- tectomy for cholangiocarcinoma is planned in a jaundiced patient or if a pancreaticoduodenectomy is to be done in a patient with longstanding or severe jaundice. Other preoperative preparations include correction of vitamin K deficiency and bowel preparation.

Treatment

Lower Duct Tumors Lower duct cholangiocarcinoma is one of the four types of periampullary tumors treated by pancreaticoduodenectomy, the others being pancreatic, ampullary, and duodenal cancers. The extent of the procedure is the same as that used for pancreatic carcinoma and will not be described in detail in this chapter. The procedure has reached a “near-zero” mortality rate in expert hands at tertiary care centers specializing in hepatobiliary-pancreatic surgery. [66] Often, the tissue of origin of the tumor, that is, whether pancreatic, ampullary, or bile duct in origin, is uncertain until after the specimen is examined pathologically, and even then, doubt can remain. The main complication of this procedure is a fistula from the pancreatic-jejunal anastomosis, which occurs in 5% to 10% of patients. [66] Biliary fistulas occur in about 2% of patients. Patients rarely die from these complications today because of improvements in diagnostic and interventional radiology, intensive care, and treatment of infection. [66] Resection of part of the stomach is no longer required for lower duct cholangiocarcinoma, although there is little or no difference in short-term outcome or quality of life [67] between the pylorus-preserving and standard types of pancreaticoduodenectomy. Many patients require pancreatic enzyme replacement after this procedure, but few become diabetic. Palliative treatment of unresectable lower duct tumors is by stenting and a combination of chemotherapy and radiation. The only randomized trial of surgical bypass versus endoscopic intubation favored the latter. [68] When unresectability is determined only after open exploration has been performed, a double bypass is performed to decompress the biliary tree and bypass the duodenum, which may become obstructed. Palliative resection of periampullary tumors may be unavoidable because the full extent of the tumor may not be detectable until the procedure is nearing completion; however, compared with upper duct tumors, this is an uncommon problem. Palliative resections for visceral cancers is an accepted procedure, for example, for colonic and gastric cancers, but has been avoided for periampullary tumors because of the risk of the procedure. Recently, however, an argument has been made in favor of palliative resection because the procedure can be now done with little risk. [69]

Upper Duct Tumors The goals of surgical resection are to remove the tumor with negative resection margins. This may be accomplished with or without an associated hepatic resection depending on the extent of the tumor. In some cases, it will be known preoperatively or by intraoperative staging with ultrasound that a liver resection will be required because the disease will have spread into the liver on one side. At other times, the need for a liver resection becomes apparent only intraoperatively, after exploration, or after the extrahepatic bile ducts are resected and microscopic disease is discovered on one side at the highest possible point of extrahepatic resection. Unfortunately, it is also at this point in the operation, when because of the discovery of bilateral microscopic involvement, it becomes clear that a curative resection has not been accomplished and is not possible. The same considerations apply to the discovery of microscopic involvement of vessels. Operative management is further complicated by the fact that frozen-section diagnosis of the presence or absence of tumor at the resection margin may be very difficult in these fibrotic cancers. A description of the operative technique is beyond the purposes of this text; however, the principal steps in the operation are as follows. The bile duct is divided at the level of the pancreas and a negative margin confirmed by frozen section. The bile duct and gallbladder are dissected off and away from the portal vein and hepatic arteries, leaving the latter structures bare; that is, all nodes and soft tissue are taken with the specimen. The bile ducts and surrounding tissue are freed to above the bifurcation where the tumor is situated, until macroscopically normal ductal tissue is encountered in the right and left bile ducts. These ducts are divided and frozen sections obtained. Coring out of liver tissue in the porta may permit excision to the level of the first branches of the right and left bile ducts. If the tumor has grown too far up one side to perform an extrahepatic resection, the bile duct is divided on the side to be retained, and the liver, bile ducts, and vascular structures are removed on the other side. Reconstruction is by a Roux-en- limb, 60 cm in length, to avoid reflux of intestinal contents into the biliary tree. Cholangiocarcinoma commonly invades caudate bile ducts, and caudate resection is routinely performed by some surgeons for upper duct cholangiocarcinoma. This area must be carefully evaluated at surgery and caudate resection performed if there is any question of caudate invasion. The important trends in surgery of upper duct cholangiocarcinoma are that a higher percentage of upper duct tumors are being resected, [70] largely owing to the more common use of liver resection, [71] vascular resection and reconstruction are becoming more common, and surgical morbidity and mortality are decreasing. Mortality is 5% or less. [72], [73] and [74] Because liver resection combined with hilar duct resection is performed for more advanced disease, comparisons between local resection and local resection plus liver resection for outcomes, such as percentage of procedures obtaining negative margins and survival, are meaningless. The results of surgical resection depend highly on whether negative resection margins are achieved. [65], [75], [76], [77], [78] and [79] There are almost no 5-year survivors when resection margins are positive. [65], [79], [80] Other negative prognostic variables are tumor stage, [78] nodal disease, [54], [78] tumor grade, [65], [80] bilirubin concentration, [65], [80] and absence of mucobilia. [54] When negative margins are obtained, the 5-year survival rate varies from 11% to 43% in recent series. [1], [65], [78], [79] and [80] Again, it must be stressed that these results are in small numbers of highly selected patients. The median survival of patients with tumor-free margin is 3 to 3.4 years, compared with a median survival of 1 to 1.2 years for patients with disease at the resection margins. Palliation of unresectable upper duct tumors may be by percutaneous [81] or endoscopic intubation or by surgical bypass. There have been no randomized trials. Metallic stents have extended the functional period of stents. Plastic stents typically clog at 3 to 4 months, whereas metal stents last 8 [82] to 10 [83] months. Surgical treatment is by biliary enteric bypass, usually employing the duct to segment III, the so-called Bismuth-Corlette procedure. [84] Bypass to

right-sided ducts is more difficult and less successful. Surgical implantation of large-bore tubes through the tumor has been used in the past but is rarely employed now.[85, 86] Both surgical bypass and internal stenting have proponents.[70, 78, 87] Stenting has the advantage of initial low morbidity and the disadvantages of occasional failure to clear jaundice, bouts of cholangitis, and the need for repeated procedures to deal with stent obstruction. With improvement in techniques of stenting, one may expect resolution of jaundice in 80% of patients, with a 10% major morbidity rate, a 30-day mortality rate of about 5%, and a median survival of 8 months.[88] Surgical bypass requires a major open surgical procedure and has corresponding early morbidity,[70] but the complete clearance of jaundice is usual, and cholangitis is uncommon. There are many nonrandomized comparisons of these techniques, but all are contaminated by patient selection and consequently are of little value in determining which technique is superior. For instance, greater success in clearing jaundice by surgical bypass may partly reflect an earlier stage of disease. Patient selection is probably quite appropriate in this disease. Younger, healthier patients, in whom bypass to segment III will decompress 40% to 50% of the liver, probably should have surgical decompression; whereas older patients, especially those with intercurrent disease or in whom decompression will provide biliary drainage from less than 40% of liver mass, should be treated by nonsurgical means.[89] Radiation therapy has been used alone or in combination with other techniques. Its value is unclear because most series are small and no randomized comparisons exist. External-beam irradiation was successful in clearing jaundice in 10 of 11 patients in a recent report; no other decompressive measures were used.[90] Brachytherapy has been applied through percutaneous tubes, with a median survival of 23 months.[91] The combination of surgery and radiotherapy was reported to provide a median survival of 14 months in unresectable or recurrent disease.[92] Another series reported no benefit to radiation.[93]

Intrahepatic Tumors The principles of treatment are as for other malignant intrahepatic hepatic lesions.[94] The tumor must be resected with a margin of normal tissue to obtain microscopically free resection margins (a 1-cm tumor-free resection margin is the goal) yet leave enough normally functioning liver tissue behind for the patient to have adequate liver function in the postoperative period. The size of the resection may vary from a single segment or less to resection of three of the four hepatic sections.[95] Like upper duct tumors, results depend on whether negative resection margins are achieved. There are few case series that separate out these uncommon tumors from other cholangiocarcinomas. One recent series obtained a 5-year survival rate of 44%[1] in 18 patients, whereas another reported a 32% 2-year survival rate in 19 patients.[96] Liver transplantation has been performed for intrahepatic and upper duct cholangiocarcinoma, but the results have been disappointing. In Klintmalm's series, only 3 of 14 patients (21%) lived more than 28 months after the procedure,[97] whereas Shimoda's series of 25 patients reported a 3-year survival of 32%.[98] Although these are not poor results for a visceral cancer, they must be evaluated with the knowledge that many patients with end-stage chronic liver disease are dying while on a waiting list for liver transplantation, and the comparative survival rate in this group of patients would be expected to be about 90%. In general, liver transplantation is not a treatment for this disease at the present time.[78, 97]

BENIGN BILE DUCT TUMORS

Benign tumors are uncommon. Most information comes from case reports. The clinical presentation is similar to that of choledocholithiasis: pain, jaundice, and less commonly, cholangitis or pancreatitis. Adenomas are the most common benign tumors of the bile ducts. They may be single, multiple, tubular, papillary, or mixed. Multicentricity has been reported but is uncommon. Cystadenoma and granular cell tumors are rare benign tumors of the extrahepatic bile ducts. Cystadenomas are similar to and may coexist with cystadenomas of the liver and pancreas.[99] Granular cell tumors are unusual, occur most commonly in black women, and may be associated with granular cell tumors at other sites.[100, 101] The malignant potential of these lesions is unknown. Reported benign mesenchymal tumors include fibromas, lipomas, leiomyomas, and neurilemomas.

Papillomatosis is a rare condition in which multiple papillary lesions involving a large amount of the bile duct mucosa may be encountered. Because malignant transformation or progression to invasive carcinoma has been reported, papillomatosis of the bile duct mucosa should be considered a premalignant lesion.[102, 103] This lesion should be treated by complete excision if possible; if not, regrowth is common.

CARCINOMA OF THE GALLBLADDER

Carcinoma of the gallbladder is the fifth most common malignancy of the gastrointestinal tract.[104] Like information on cholangiocarcinoma, studies of gallbladder cancer tend to be small case series covering many years, and claims for the best orm of investigation or treatment must be cautiously interpreted. Cancers arising from gallbladder mucosa appear to behave in a fashion similar to other adenocarcinomas of the gastrointestinal tract; premalignant to invasive malignant changes can be found, metastatic spread occurs by lymphatic and vascular routes, diagnosis is often delayed, and survival is related to stage. Interestingly, at the population level, mortality is also inversely related to cholecystectomy rates.[105]

Incidence

The highest incidence of gallbladder cancer occurs in Chileans and Bolivians.[106] Gallbladder cancer is the fourth leading cause of all cancer deaths in Chile; mortality from gallbladder cancer (5.2%) in Chile is the highest in the world.[107] Other commonly affected population groups include Native Americans, Hispanic American women, Latin American women, Japanese women in Japan, African Zimbabwian women, European immigrant women in Israel, and northern Europeans.[108] In contrast to the general population of the United States, in whom the incidence is low at about 3 per 100,000, gallbladder cancer is the most common gastrointestinal malignancy in Native Americans. Low rates of gallbladder cancer occur among Nigerians, New Zealand Maoris, and Chinese natives and immigrants.[108]

Cancer of the gallbladder in the United States is a disease of elderly people, with the greatest incidence occurring after 65 years of age. It is three times more common in women than in men, and it is more common in whites than in blacks. Genetic differences are exceedingly important. In North America, the highest incidence of gallbladder cancer occurs among Native Americans and Mexican Americans. In North and South America, gallbladder cancer is related to Indian rather than Spanish heritage because the incidence of gallbladder cancer in Spain, Cuba, and Puerto Rico is low. In Mexico, mestizos (people of mixed ancestry) have the highest incidence, whereas in Bolivia, there appears to be a difference based on tribal origin (language).[109]

Etiology

Etiologic factors that appear to be or have been claimed to be important for the development of gallbladder cancer include genetic characteristics, gallstone disease, bile composition, calcification of the gallbladder wall, anomalous junction of the biliary and pancreatic ducts, congenital biliary cysts, some infections, environmental carcinogens, and drugs. Geographic differences may reflect racial and genetic differences, although cultural differences possibly play a role. A progression from dysplasia to early cancer to metastatic cancer has been observed,[110] the progression from dysplasia to cancer taking about 15 years.

Gallstone disease is associated with gallbladder cancer, but the mechanism is unknown. Most reports of the relationship between gallstones and gallbladder cancer have come from operative or autopsy studies; however, a prospective cohort study of patients with gallstones found that gallstones did increase the risk for gallbladder cancer, but the actual incidence was very low (9 per 10,000 per person-years), and the absolute number of cases of gallbladder cancer (5 of 2583 people) in this population was low.[111] The incidence of gallbladder cancer in patients with cholecystolithiasis ranges between 0.5% and 3%.[112] Cholecystolithiasis is present in 70% to 90% of patients with gallbladder cancer. Duration of gallstones, patient age, size of gallstones, and possible carcinogenic effects of gallstones, such as from the chemical composition or bacteria within the stones, may be important,[113] although in a recent study, patients with cancers did not have larger stones, nor were their stones of higher cholesterol content.[109]

Duration of cholecystolithiasis appears to be a crucial factor for the development of cancer. Patients who have had cholecystolithiasis for longer than 40 years have a significantly higher incidence of gallbladder cancer than those who have had gallstones for a shorter time. Glenn and Hays[114] suggested that 1% of patients older than 65 years of age with gallstones would develop gallbladder cancer. This more likely reflects the duration of stone disease rather than an effect of age. However, the possibility of cancer is not an indication for cholecystectomy in a patient with asymptomatic stones. In many countries, gallbladder cancer is unknown, which is probably related to the rate of cholecystectomies performed for asymptomatic gallstones or early in the course of patients with symptomatic stones.

Little is known about the pathophysiology of gallbladder cancer in patients with cholecystolithiasis. Chronic irritation of gallbladder mucosa by stones, followed by repeated episodes of epithelial repair over a period of years, may lead to malignant transformation. The chemical composition of stones or bile may be related to development of gallbladder cancer. Indeed, because bile from high endemic areas is more mutagenic than that from low endemic areas,[115] there is probably some factor excreted by the liver into bile that contributes to malignant transformation. Cholesterol stones are the most common type associated with gallbladder cancer. Bile acids can act as co-carcinogens, and there is a structural similarity to the known carcinogen, methylcholanthrene. In the most comprehensive analysis yet, Strom and colleagues[109] performed a case-control study in Mexico and Bolivia that compared patients with gallbladder cancer, patients with gallstones, and patients with no

biliary tract disease. Their prior hypotheses that patients with tumor would have increased levels of lithocholate was not confirmed, but bile glycoursodeoxycholate levels were unexpectedly elevated in the patients with cancer. This paper must be read to appreciate the difficulty of doing and interpreting such a combined epidemiologic and biochemical study. For instance, the authors were unsure at the end of their study whether their unexpected findings in bile acid composition were a cause or effect of the tumor. There are no conclusive data linking bile composition of bile acids to carcinoma of the gallbladder. Because bile is the main route of excretion of dietary or chemical metabolites with molecular weight of more than 500, there are a very large number of potentially carcinogenic compounds.

A number of recent studies from Japan have linked anomalous pancreatic-biliary duct junction (APBJ) to gallbladder and extrahepatic bile duct cancer. [116], [117] and [118] There is a much higher incidence of biliary tract cancer in patients with APBJ. About 2% of Japanese patients examined had APBJ. About 75% of these cases were associated with a choledochal cyst, the rest having normal-caliber bile ducts. In those associated with choledochal cyst, cancer may arise in the extrahepatic duct or gallbladder, whereas in those without ductal dilation, the cancer seems almost always to occur in the gallbladder. [116], [117] Cancer of intrahepatic ducts does not appear associated with APBJ. Patients with APBJ get cancer at a younger age than patients with sporadic gallbladder cancer. [117] Pediatric patients with APBJ frequently have epithelial hyperplasia of the gallbladder. [119] The nonmalignant areas of the gallbladder of patients with APBJ-associated gallbladder cancer show increased hyperplastic changes in the gallbladder mucosa compared with patients with sporadic gallbladder cancer. [120] In a cat model, side-to-side biliary-pancreatic anastomosis produced hyperplastic changes in the gallbladder within 6 months. [121]

Another factor that has been associated with gallbladder cancer is partial or complete calcification of the gallbladder wall (porcelain gallbladder). Although an incidence of coexistent gallbladder malignancy of up to 25% has been claimed in patients with porcelain gallbladder, recent data suggest that this is less common. [122] Others have questioned whether porcelain gallbladder is linked to gallbladder carcinoma at all. [123] *Salmonella typhi* carriage; exposure to toxic environmental factors in the automotive, rubber, textile, and metal industries [36], [106], [124]; previous gastric operations [125]; and elevated body mass index [109] have been associated with gallbladder cancer.

Molecular Genetics There are relatively few studies examining the molecular events in carcinoma of the gallbladder. As in cholangiocarcinomas, the *erbB2* oncoprotein is expressed in a significant fraction of gallbladder cancers. [126] Furthermore, transgenic mice that express *erbB2* in the gallbladder epithelium develop gallbladder carcinomas. [127] Data regarding the expression of *ras* and *myc* genes in gallbladder cancer are conflicting, [126], [128], [129] but it appears that the expression of these genes is substantially less than in cholangiocarcinomas. Similarly, point mutations involving the K- *ras* gene appear to be less common in gallbladder cancer than in cholangiocarcinomas. [129] Several studies have identified *p53* expression in gallbladder cancers; about two thirds of such tumors express *p53*. [33], [130] Such overexpression is not seen in adenomas.

Pathology

As noted, there is a progression from dysplasia to carcinoma in situ to invasive cancer in the gallbladder epithelium that appears to take about 15 years. [110] Premalignant lesions are associated with gallbladder cancer, and it is thought that chronic inflammation may play a role in development of premalignant lesions. [36] Two types of metaplasia, intestinal and squamous, have been found in patients with gallbladder cancer. The relation of intestinal metaplasia to subsequent development of gallbladder cancer has not been determined. Squamous metaplasia, in which squamous epithelium replaces the normal gallbladder epithelium, is a rare premalignant lesion that has been found with squamous cell cancer of the gallbladder. Cholecystitis follicularis, a rare type of inflammation, has been reported in a few cases of gallbladder cancer, but its premalignant potential is unclear. [36]

The classification of malignant lesions of the gallbladder is shown in Table 103-4. Sixty percent of cancers of the gallbladder are found in the fundus, 30% in the body, and 10% in the neck. They may be isolated or involve the entire gallbladder through intramural spread, analogous to linitis plastica of the stomach.

Malignant Epithelial Tumors
Adenocarcinoma
Squamous cell carcinoma
Oat cell carcinoma
Others
Malignant Mesenchymal Tumors
Embryonal rhabdomyosarcoma
Leiomyosarcoma
Malignant fibrous histiocytoma
Miscellaneous
Carcinosarcoma
Carcinoid
Lymphoma
Melanoma
Others

TABLE 103-4 Malignant Neoplasms of the Gallbladder

Most malignant neoplasms of the gallbladder are adenocarcinomas, which behave in a manner similar to adenocarcinomas arising from other epithelial tissues. They originate as mucosal lesions, and as growth progresses, they invade the wall of the gallbladder. The anatomy of the gallbladder wall partly explains the advanced stage of disease at which most patients with gallbladder cancer present. The lack of a well-defined muscularis leads to early entry of invasive gallbladder cancer into the perimuscular connective tissue. Lymphatic, neural, and hematogenous invasion occurs earlier with gallbladder cancer than with cancers of the gut.

Gallbladder cancer frequently extends beyond the gallbladder to involve the liver and the extrahepatic biliary tree. Most patients with liver involvement have direct extension of the disease; fewer than 10% of patients have liver metastasis without full-thickness invasion of the gallbladder wall. The routes of lymphatic drainage of the gallbladder have been studied in an attempt to rationalize aggressive surgery for cancer of the gallbladder. Uesaka and colleagues [131] injected a carbon particle suspension into the primary drainage nodes—the cystic and peri- choledochal nodes. The main route of drainage was on the right side of the bile duct to the superior retropancreatic node and from there directly to the paraaortic nodes or indirectly to the paraaortic nodes through posterior pancreatic nodes. Drainage to the left side of the bile duct to the celiac nodes and drainage upward to the hilar nodes and into segment IV of the liver were much less common modes of drainage. Lymph node metastasis to the cystic, pericholedochal, peripancreatic, and celiac nodes occurs early; more than 50% of patients are found to have lymph node metastases at the time of diagnosis. Direct invasion of the duodenum and the colon also occur. Intraperitoneal spread, Krukenberg tumors (i.e., ovarian metastases), and hematogenous dissemination occur.

A uniform staging system for gallbladder cancer has not been widely applied. Nevin and colleagues [132] developed the first clinically useful staging system. The TNM system, described by the American Joint Committee on Cancer, is the preferred staging system. [3] Survival of patients with gallbladder cancer is related to stage and histological type of cancer. Lymph node involvement is rare in stage T1 tumors [132], [133]; that is, lymph node involvement almost never occurs until the muscularis has been penetrated. After that lymph node involvement is common, occurring in about 50% of stage II patients and in 70% to 80% of patients in stage III and IV. [132], [133] There is a close correlation between lymph node involvement and prognosis. [134], [135] and [136] Most long-term survivors are patients with well-differentiated tumors that were minimally invasive. These are usually found incidentally at or immediately after cholecystectomy (Fig. 103-8).

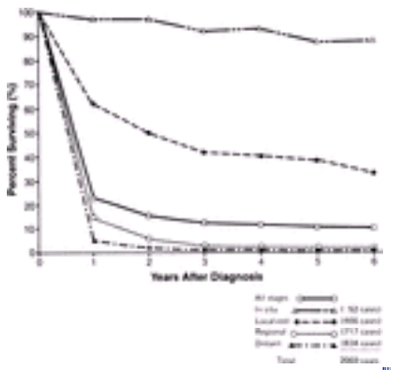

FIGURE 103-8. Gallbladder cancer survival rates by stage in all races and genders from 1973 to 1981. (From ref. [36].)

Clinical Presentation

Signs and symptoms of gallbladder cancer are nonspecific. This explains the delay in diagnosis in most patients with gallbladder cancer. In TNM stage I and II symptoms often mimic those of cholelithiasis and cholecystitis. [137] Indeed, in early stages, the cause of symptoms may be the associated cholelithiasis. Pain is the most common initial complaint; it may be dull and aching, colicky, sharp, constant, or intermittent, and it may or may not radiate to the back. Other symptoms include nausea, vomiting, and anorexia. In later TNM stages, weight loss, jaundice, hepatomegaly, a palpable mass, or ascites may develop. Jaundice develops in 30% to 60% of patients and is a poor prognostic sign because it is caused by extension of the tumor beyond the gallbladder, with obstruction of the extrahepatic bile ducts; 85% of patients with jaundice have unresectable tumors. [112] The patients may also have obstruction of the duodenum or colon. Malignant cholecystoenteric fistula may be the first indication of gallbladder cancer.

Diagnosis and Staging

In most cases, the diagnosis is not made preoperatively [112]; this is true even in recent series. [138] Laboratory findings are not diagnostic but are related to abnormalities associated with bile duct obstruction. Patients frequently have increased alkaline phosphatase and bilirubin levels. Blood levels of CEA or CA 19-9, another tumor marker, may also be elevated. These findings are not diagnostic because they may have other causes.

Diagnostic techniques include oral cholecystography, ultrasonography, CT scanning, and possibly MRI. The most widely employed diagnostic study for biliary tract disease is ultrasound. Findings on ultrasonography that are suggestive, but not diagnostic, of gallbladder cancer include thickening of the wall, a mass projecting into the lumen, calcification of the gallbladder, multiple masses or a fixed mass in the gallbladder, a mass extending into the liver, or an extracholecystic mass. In a comparison of patients with unsuspected gallbladder cancer and patients with benign gallbladder disease, it was found that solitary stone, displaced stone, intralumenal mass, and invasive mass were more commonly associated with cancer. [139] A report of mucosal thickening should also be viewed with suspicion. CT scanning may help in the diagnosis but is of limited help in staging the disease except in estimating the extent of hepatic invasion [140] (Fig. 103-9).

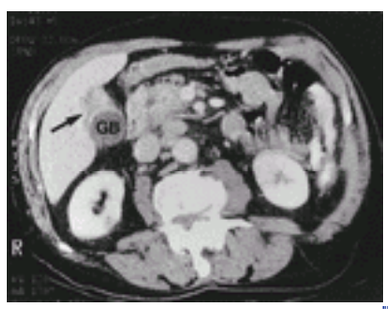

FIGURE 103-9. Computed tomography scan of patient with carcinoma of the gallbladder (*GB*) showing invasion of hepatic parenchyma (*arrow*).

Gallbladder polyps may be malignant but are rarely so when smaller than 1 cm in diameter. In a recent series of 103 patients with gallbladder polyps, none was malignant if less than 1 cm in diameter, but 23% of polyps larger than 1 cm were malignant. [141] In another series, 88% of polyps larger than 1 cm in diameter were malignant, and polyps larger than 1.8 cm were more likely to contain a more advanced stage of cancer. [142] FNA appears to be an accurate way of distinguishing between polyps due to cholesterolosis and neoplastic polyps, [143] especially when the polyps are larger than 1 cm in diameter [144]; however, it is much less accurate in determining whether a neoplastic polyp is an adenoma or carcinoma. [144] Duplex Doppler ultrasound imaging of blood flow within a neoplastic polyp may also be useful in distinguishing these lesions from cholesterol polyps.

Many of the staging considerations are similar to those discussed under cholangiocarcinoma. Staging laparoscopy with ultrasound appears to have many of the same advantages for staging of gallbladder cancer that it has for other hepatobiliary malignancies. [46], [47], [145]

Treatment

Both treatment and prognosis of gallbladder cancer depend on stage of disease. [132] Unfortunately, most cancers are diagnosed when symptomatic, and at that time, the stage of disease is usually TNM III or IV. [135], [146] Because most patients in advanced stages are not resectable, the stage distribution at the time of open surgery has been more heavily weighted to later stages than the stage distribution of patients actually resected for cure. For instance, in a German series of 81 operated patients, the TMN stage distribution (I to IV) was 7%, 12%, 15%, and 66%, [135] whereas the stage distribution of 106 radically resected cases in a Japanese series was 14%, 43%, 24%, and 19%. [133] It is to be hoped that staging laparoscopy with ultrasound will alleviate this disparity and reduce the number of patients having open exploration without resection. Of the 106 patients in the Japanese series, curative resection was achieved in 74. Most of the noncurative resections were in patients with stage IVb cancer. [133] In a recent American series containing 149 patients, 58 cancers were explored, and of those, 23 were resected for cure. [135] As previously noted, these types of figures may not be representative of the broad population of patients contracting the disease.

There are no established principles of treatment because no definite trials establishing these exist. At best, there are rational guidelines based on the facts that T1 lesions rarely have metastasized to lymph nodes but T2 to T4 lesions have usually done so [132], [133] and that radical surgery achieving negative tumor margins has resulted in acceptable results in more advanced disease. [147] There are several levels of resection. In increasing order of magnitude they are (1) extraserosal cholecystectomy or cholecystectomy with wedge resection of the liver, (2) extraserosal cholecystectomy combined with portal lymph node dissection and sometimes extrahepatic bile duct resection, (3) liver resection with cholecystectomy and lymph node dissection and sometimes extrahepatic bile duct resection, and (4) the most radical—liver resection with cholecystectomy and lymph node dissection combined with pancreaticoduodenectomy.

Beginning with the earliest stages, the recommendation for treatment may be summarized as follows. When gallbladder cancer is suspected, an open procedure should be performed. If there is no evidence of spread outside the gallbladder, an extraserosal cholecystectomy should be done. An extraserosal cholecystectomy excises the gallbladder on a deeper plane than a standard cholecystectomy, so that the gallbladder and all connective tissue down to actual liver tissue are removed. This can be done laparoscopically; however, it is our opinion that it should not be attempted because gallbladder perforation and bile spillage are more common in the laparoscopic technique. The negative consequences of tumor implantation or incomplete excision far outweigh any benefit of minimally invasive surgery. The excised specimen should be inked. If resection margins are negative and if the tumor has not penetrated the muscularis (T1 lesion), the procedure is considered to be complete. If margins are positive or if the muscularis has been penetrated, then resection of segments 4b and 5 and a lymph node dissection are performed. If the cystic duct resection margin is positive, then, in addition, the extrahepatic bile duct is excised to clear margins.

When there is obvious penetration to the serosal layer on the deep surface of the gallbladder by intraoperative ultrasound or palpation, then resection of the gallbladder with segments 4b and 5 and a lymph node dissection with or without extrahepatic bile duct resection is the recommended procedure. Sometimes, because of the local depth of penetration into the liver, a more extensive liver resection is required. Stated otherwise, a cholecystectomy is usually sufficient for T1 lesions and usually results in cure. For T2 to T4 lesions, the extent of resection depends on the route of direct extension, whether into the liver or bile duct; a lymph node dissection is always done.

Results in recent case series [133], [135], [147], [148] appear to show marked improvement over classic reports. Mortality is infrequent, even for major resections, but there is a complication rate of about 25%, including minor complications. Exact comparisons among series are not possible because different operations have often been done for the same disease stage. For long-term survival, a curative resection with negative margins must be obtained. If negative resection margins are achieved, 5-year survival rates according to stage are about as follows: stage I, 90%; stage II, 80%; stage III, 40%; and stage IV, 15%. Prognosis in stage IV appears best when the tumor is placed in this category because of local hepatic invasion, rather than because of lymph node involvement. [135] Lymph node involvement is an ominous sign, [135] although 5-year survivors have been reported in some series. [133], [148] Based on the pattern of lymph node drainage, a pancreaticoduodenectomy has been performed for T2 to T4 disease. [149] However, the results have still been poor in advanced stages.

Palliation of gallbladder cancer mainly requires treatment of obstructive jaundice. The considerations are the same as for cholangiocarcinoma, except that percutaneous or endoscopic means of decompression are preferable in this rapidly progressive state. Chemotherapy is experimental and of no proven benefit. [150] Although some gallbladder cancers may be radiosensitive, [151] radiation therapy provides little survival benefit because of the extent of tumor at the time of diagnosis. Palliation of jaundice has been reported, and intraoperative irradiation may be beneficial. [152], [153] Although these types of treatments should be considered, their role has not been defined.

Incidental Gallbladder Cancer at Laparoscopic Cholecystectomy Gallbladder cancer may be an incidental finding at laparoscopic cholecystectomy, as it was at open cholecystectomy in the past; the incidence ranges from 0.3% to 1%. The difference in the present era is that port-site implantation may occur. [154], [155] Port-site implantation may simply be due to contact between the malignancy and the tissues at the perimeter of the port site at the time of gallbladder extraction. However, positive-pressure pneumoperitoneum appears to be another factor. [156] Preoperative suspicion should be raised by the ultrasonographic findings listed previously. Extraction of the gallbladder in a plastic bag is prudent when these signs exist or when there is evidence of gallbladder wall thickening at surgery. Every gallbladder should be inspected at the time of extraction and questionable areas resected for biopsy. If a cancer is discovered, it should be treated at that time using the principles stated previously. The tissue surrounding the trocar ports should also be excised as part of the treatment because seeding may have occurred. Early reoperation is the approach of choice when the tumor is discovered on later pathological examination, when surgery at the time of the initial procedure could not proceed because of uncertainty of diagnosis or degree of penetration of the tumor, or when the surgeon believes that the proposed procedure must be discussed with the patient. Stage I gallbladder carcinomas that have clear resection margins probably require no further surgical treatment.

BENIGN TUMORS OF THE GALLBLADDER

Benign tumors of the gallbladder are uncommon and infrequently symptomatic. Benign epithelial tumors include adenomas and mixed tumors. Adenomas may be tubular, papillary, or mixed. One third are multiple, and fewer than 50% are associated with stones. Benign tumors may be symptomatic, causing biliary colic, presumably as a result of shedding of tumor, which temporarily obstructs the cystic duct. Treatment is laparoscopic cholecystectomy when the mass is symptomatic and less than 1 cm in diameter. Cholecystectomy is performed for either symptomatic or asymptomatic tumors when the mass is larger than 1 cm in diameter because of the possibility of malignancy and for the same reason the procedure is performed open. FNA may permit the diagnosis of cholesterolosis, in which case surgery is necessary only if symptomatic and laparoscopic surgery may be performed. (see earlier).

Other problems with which benign tumors of the gallbladder are confused are much more common than benign tumors of the gallbladder. These include cholesterolosis and adenomyomatosis. The former is a disease of unknown etiology in which cholesterol accumulates focally, segmentally, or diffusely in the gallbladder. Focal cholesterolosis may present as a polyp and be confused with a tumor. Adenomyomatosis goes by a variety of other names as well. It is a condition in which there is nonneoplastic ingrowth of gallbladder glands into the wall of the gallbladder and even penetration of the wall. The cause is unknown. It may be focal or diffuse. When focal, the site is usually in the fundus, producing the characteristic fundal adenomyoma. When diffuse, it produces wall thickening. Both types may be confused with malignancy. There are characteristic changes on ultrasound and contrast cholangiography, which help differentiate the process form cancer. Benign mesenchymal tumors are rare and include leiomyoma, hemangioma, and lipoma.

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CHAPTER 104

Glen A. Lehman and Stuart Sherman

SPHINCTER OF ODDI DYSFUNCTION (POSTCHOLECYSTECTOMY SYNDROME)

Sphincter of Oddi dysfunction (SOD) is a major consideration in patients with postcholecystectomy syndrome, which may be broadly defined as persistence or recurrence of upper abdominal symptoms, especially right upper quadrant pain, after cholecystectomy. If the cholecystectomy was done for vague or nonspecific symptoms, which were in reality not pancreatobiliary in origin, persistence of symptoms is not surprising. If, however, the cholecystectomy was done for clinically apparent biliary colic or cholelithiasis, then recurrence of symptoms is of more concern for organic disease, such as common duct stones or SOD. Common duct stones are discussed in Chapter 102. This chapter focuses primarily on the discussion of SOD.

DEFINITIONS

We assume that patients with postcholecystectomy syndrome, upper abdominal pain suggestive of biliary colic, undergo appropriate invasive and noninvasive evaluation to rule out common duct stones, tumors, or strictures near the cholecystectomy site. The residual group of patients has a high frequency of SOD. SOD refers to benign, noncalculous obstruction to flow of bile or pancreatic juice through the pancreaticobiliary junction, that is, the sphincter of Oddi (SO). [1] SOD may be manifested clinically by pancreaticobiliary pain, pancreatitis, or deranged liver function tests. SO dyskinesia refers to a motor abnormality of the SO, which causes a hypertonic sphincter. In contrast, SO stenosis refers to a structural alteration of the sphincter, probably from an inflammatory process, with subsequent fibrosis. Because it is often impossible to distinguish patients with SO dyskinesia from those with SO stenosis, the term SOD has been used to incorporate both groups of patients. In an attempt to deal with this overlap in etiology, and also to determine the appropriate use of SO manometry (SOM), a clinical classification system has been developed for patients with suspected SOD [2] (Hogan-Geenen SOD classification system; Table 104-1) based on clinical history, laboratory results, and endoscopic retrograde cholangiopancreatography (ERCP) findings. A variety of less accurate terms—such as papillary stenosis, ampullary stenosis, biliary dyskinesia, and postcholecystectomy syndrome—are listed in the medical literature to describe this entity. The latter term is somewhat of a misnomer because SOD may clearly occur with an intact gallbladder.

TABLE 104-1 Hogan-Geenen Sphincter of Oddi Classification System Related to the Frequency of Abnormal Sphincter of Oddi Manometry and Pain Relief by Biliary Sphincterotomy

ANATOMY, PHYSIOLOGY, AND PATHOPHYSIOLOGY

The SO is a small complex of smooth muscles surrounding the terminal common bile duct, the main (ventral) pancreatic duct of Wirsung, and the common channel (ampulla of Vater), when present. It has both circular and figure-of-eight components. The high-pressure zone generated by the sphincter is variably 4 to 10 mm in length. Its roles appear to be to regulate the flow of bile and pancreatic exocrine juice and to prevent duodenum-to-duct reflux (i.e., to maintain sterile intraductal environment). The SO possesses both a variable basal pressure and phasic contractile activity. The former appears to be the predominant mechanism for regulating outflow of pancreaticobiliary secretion into the intestine. Although phasic SO contractions may aid in regulating bile and pancreatic juice flow, their primary role appears to be maintaining a sterile intraductal milieu. Sphincter regulation is under both neural and hormonal control. Phasic wave frequency of the sphincter is closely tied to the migrating motor complex (MMC) of the duodenum. Phase III of the MMC is associated with the most rapid phasic wave activity of the sphincter. Innervation of the bile duct does not appear to be essential because sphincter function has been reported to be preserved after liver transplantation. [3] Although regulatory processes vary among species, cholecystokinin and secretin appear to be most important in causing sphincter relaxation, whereas nonadrenergic, noncholinergic neurons, which at least in part transmit vasoactive intestinal peptide and nitric oxide, also relax the sphincter. [4] The role of cholecystectomy in altering these neural pathways needs further definition.

Wedge specimens of the SO obtained at surgical sphincteroplasty from SOD patients show evidence of inflammation, muscular hypertrophy, fibrosis, or adenomyosis within the papillary zone in about 60% of patients. [5] In the remaining 40% with normal histology, a motor disorder is suggested. Less commonly, infections with cytomegalovirus or *Cryptosporidium* species, as may occur in patients with acquired immunodeficiency syndrome, or *Strongyloides* species have caused SOD.

It is suspected that SOD causes pain by impeding the flow of bile and pancreatic juice resulting in ductal hypertension. Alternatively, ischemia arising from spastic contractions and hypersensitivity of the papilla has been proposed.

EPIDEMIOLOGY AND FREQUENCY

SOD may occur in pediatric or adult patients of any age; however, patients with SOD are typically middle-aged women. Although SOD most commonly occurs after cholecystectomy, it may be present with the gallbladder in situ. In a survey of functional gastrointestinal disorders, SOD appeared to have a relevant impact on quality

of life because it was highly associated with work absenteeism, disability, and health care use. [6]

The frequency of manometrically documented SOD in patients before cholecystectomy has received limited study. Guelrud and colleagues [7] studied 121 patients with symptomatic gallstones and a normal common bile duct diameter (by transcutaneous ultrasound) by SOM before cholecystectomy. An elevated basal sphincter pressure was found in 14 patients (11.6%). SOD was diagnosed in 4.1% of patients with a normal serum alkaline phosphatase level (4 of 96) and in 40% with an elevated serum alkaline phosphatase level (10 of 25). Ruffolo and associates [8] evaluated 81 patients with symptoms suggestive of biliary disease but normal ERCP and no gallbladder stones on transcutaneous ultrasound by scintigraphic gallbladder ejection fraction and endoscopic SOM. Fifty-three percent of patients had SOD, and 49% had an abnormal gallbladder ejection fraction. SOD occurred with a similar frequency in patients with an abnormal gallbladder ejection fraction (50%) and a normal ejection fraction (57%).

Postcholecystectomy pain, resembling the patient's preoperative biliary colic, occurs in at least 10% to 20% of patients. [9] The frequency of diagnosing SOD in reported series varies considerably with the patient selection criteria, the definition of SOD, and the diagnostic tools employed. In a British report, SOD was diagnosed in 41 (9%) of 451 consecutive patients being evaluated for postcholecystectomy pain. [10] Roberts-Thomson and Toouli [11] evaluated 431 similar patients and found SOD in 47 (11%). In a subpopulation of such patients with a normal ERCP (except dilated ducts in 28%) and recurrent pain of more than 3 months duration, SOD was diagnosed in 68%. Sherman and colleagues [12] used SOM to evaluate 115 patients with pancreaticobiliary pain with and without liver function test abnormalities. Patients with bile duct stones and tumors were excluded from analysis. Fifty-nine of 115 patients (51%) showed abnormal basal SO pressure greater than 40 mm Hg. These patients were further categorized by the Hogan-Geenen SOD classification system (see Table 104-1). The frequencies of abnormal manometry of a single sphincter segment were 86%, 55%, and 28% in type I, II, and III patients, respectively. These abnormal manometric frequencies are very similar to those reported by others for type I and type II patients. [13], [14] In type III patients, the finding of an abnormal basal sphincter pressure has varied from 12% to 55%. As noted, patient selection factors may be one explanation for this great variability. When both the pancreatic and biliary portions of the sphincter are studied, the detection rate for SOD increases.

Dysfunction may occur in the pancreatic duct portion of the SO and cause recurrent pancreatitis. Manometrically documented SOD has been reported in 15% to 59% of patients with recurrent pancreatitis, previously labeled as idiopathic. [15]

TYPICAL CLINICAL PRESENTATION

Abdominal pain is the most common presenting symptom of patients with SOD. The pain is usually epigastric or right upper quadrant, may be disabling, and lasts for minutes to hours. In some patients, the pain is continuous with episodic exacerbations. It may radiate to the back or shoulder and be accompanied by nausea and vomiting. Food or narcotics may precipitate the pain. The pain may begin several years after a cholecystectomy was performed for a gallbladder dysmotility or stone disease and is similar in character to the pain leading to the cholecystectomy. Alternatively, patients may have continued pain that was not relieved by a cholecystectomy. Jaundice, fever, or chills are rarely observed. Physical examination is characterized only by mild epigastric or right upper quadrant tenderness. The pain is not relieved by trial medications for acid-peptic disease or irritable bowel syndrome. Laboratory abnormalities consisting of transient elevation of liver function tests, typically during episodes of pain, are present in less than 50% of patients. [12] After initial evaluation, patients are commonly categorized according to the Hogan-Geenen SOD classification system (see Table 104-1). Patients with SOD may present with typical pancreatic pain (epigastric or left upper quadrant radiating to the back) and recurrent pancreatitis.

Clinical Evaluation

The diagnostic approach to suspected SOD may be influenced by the presence of key clinical features. However, the clinical manifestations of functional abnormalities of the SO may not always be easily distinguishable from those caused by organic ones (e.g., common bile duct stones) or other functional nonpancreaticobiliary disorders (e.g., irritable bowel syndrome).

General Initial Evaluation

Evaluation of patients with suspected SOD (i.e., patients with upper abdominal pain with characteristics suggestive of a pancreatobiliary origin) should be initiated with standard serum liver chemistries, serum amylase or lipase, abdominal ultrasonography, or computed tomography (CT) scans. The serum enzyme studies should be drawn during bouts of pain, if possible. Mild elevations (less than two times the upper limits of normal) are frequent in SOD, whereas greater abnormalities are more suggestive of stones, tumors, and liver parenchymal disease. CT scans and abdominal ultrasounds are usually normal, but occasionally a dilated bile duct or pancreatic duct may be found (particularly in patients with type I SOD). Standard evaluation and treatment of other, more common upper gastrointestinal conditions, such as peptic ulcer disease and gastroesophageal reflux, should be done simultaneously. In the absence of mass lesions, stones, or response to acid suppression therapeutic trials, the suspicion for sphincter disease is increased.

DIAGNOSTIC METHODS (NONINVASIVE)

Because SOM (considered by most authorities to be the gold standard for diagnosing SOD) is difficult to perform, invasive, not widely available, and associated with a relatively high complication rate, several noninvasive and provocative tests have been designed in an attempt to identify patients with SOD.

Morphine-Prostigmin Provocative Test

Morphine has been shown to cause SO contraction, as assessed manometrically. Prostigmin (neostigmine), 1 mg subcutaneously, is added as a vigorous cholinergic secretory stimulant to morphine (10 mg, subcutaneously) to make this challenge test. The morphine-prostigmin test, historically, had been used extensively to diagnose SOD. Reproduction of the patient's typical pain associated with a fourfold increase in aspartate aminotransferase, alanine aminotransferase, alkaline phosphatase, amylase, or lipase levels constitutes a positive response. The usefulness of this test is limited by its low sensitivity and specificity in predicting the presence of SOD and its poor correlation with outcome after sphincter ablation. This test has largely been replaced by tests believed to be more sensitive.

Ultrasonographic Assessment of Extrahepatic Bile Duct and Main Pancreatic Duct Diameter after Secretory Stimulation

After a lipid-rich meal or cholecystokinin administration, the gallbladder contracts, bile flow from the hepatocytes increases, and the SO relaxes, resulting in bile entry into the duodenum. Similarly, after a lipid-rich meal or secretin administration, pancreatic exocrine juice flow is stimulated, and the SO relaxes. If the SO is dysfunctional and causes obstruction to flow, the common bile duct or main pancreatic duct may dilate under secretory pressure. This can be monitored by transcutaneous ultrasonography. Sphincter and terminal duct obstruction from other causes (stones, tumors, strictures) may similarly cause ductal dilation and need to be excluded. Pain provocation should also be oted if present. Limited studies comparing these noninvasive tests with SOM or outcome after sphincter ablation [16], [17], [18] and [19] show only modest correlation.

Quantitative Hepatobiliary Scintigraphy

Hepatobiliary scintigraphy assesses bile flow through the biliary tract. Impairment to bile flow from sphincter disease, tumors, or stones (as well as parenchymal liver disease) results in impaired radionuclide flow. The precise criteria to define a positive (abnormal) study remain controversial, but duodenal arrival time greater than 20 minutes and hilum-to-duodenum time greater than 10 minutes are most widely used. [20], [21] Most studies are flawed by lack of correlation with SOM or outcome after sphincter ablation. Thomas and colleagues [22] recently reported that the use of morphine during nuclear scintigraphy increased sensitivity to 83% and specificity to 81% when compared with abnormal basal sphincter manometry. Overall, it appears that patients with dilated bile ducts and high-grade obstruction are likely to have a positive scintigraphic study. Esber and colleagues [23] found that patients with lower-grade obstruction (Hogan-Geenen classification types II and III) generally have normal scintigraphy, even if done after cholecystokinin provocation.

In the absence of more definitive data, we conclude that noninvasive testing for SOD has a relatively low or undefined sensitivity and specificity and is, therefore, not recommended for general clinical use, except in situations in which more definitive testing (manometry) is unsuccessful or unavailable.

DIAGNOSTIC METHODS (INVASIVE)

Because of their associated risks, invasive testing with ERCP and manometry should be reserved for patients with clinically significant or disabling symptoms. In general, invasive assessment of patients for SOD is not recommended unless definitive therapy (sphincter ablation) is planned if abnormal sphincter function is found.

Cholangiography

Cholangiography is essential to rule out stones, tumors, or other obstructing processes of the biliary tree that may cause symptoms identical to those of SOD. Once such lesions are ruled out by a good-quality cholangiographic study, ducts that are dilated or drain slowly suggest obstruction at the level of the sphincter. A variety of methods to obtain a cholangiogram are available. For noninvasive imaging, magnetic resonance cholangiography is most promising, but quality varies greatly from center to center. Software development continues, and quality of images continues to evolve. Direct cholangiography can be obtained by percutaneous methods, intraoperative methods, or more conventionally, ERCP. Although some controversy exists, extrahepatic ducts that are greater than 12 mm in diameter (postcholecystectomy) when corrected for magnification, are considered dilated. Drainage of contrast is influenced by drugs that affect the rate of bile flow and relaxation or contraction of the SO. Such drugs must be avoided to obtain accurate drainage times. Because the extrahepatic bile duct angulates from anterior (the hilum) to posterior (the papilla), the patient must be supine to assess gravitational drainage through the sphincter. Although definitive normal supine drainage times have not been well defined, [24] a postcholecystectomy biliary tree that fails to empty all contrast media by 45 minutes is generally considered abnormal.

Endoscopic evaluation of the papilla and peripapillary area can yield important information that can influence the diagnosis and treatment of patients with suspected SOD. Occasionally, ampullary cancer may simulate SOD. The endoscopist should do tissue sampling of the papilla (preferably after sphincterotomy) in suspicious cases. [25]

Radiographic features of the pancreatic duct are also important to assess in the patient with suspected SOD. Dilation of the pancreatic duct (more than 6 mm in the pancreatic head, and more than 5 mm in the body) and delayed contrast drainage time (9 minutes in the prone position) may give indirect evidence for the presence of SOD.

Sphincter of Oddi Manometry

The most definitive development in our understanding of the pressure dynamics of the SO came with the advent of SOM. SOM is the only available method to measure SO motor activity directly. Although SOM can be performed intraoperatively and percutaneously, it is most commonly done in the ERCP setting. SOM is considered by most authorities to be the gold standard for evaluating patients for sphincter dysfunction. [26], [27] The use of manometry to detect motility disorders of the SO is similar to its use in other parts of the gastrointestinal tract. Unlike other areas of the gut, SOM is more technically demanding and hazardous. Questions remain as to whether these short-term observations (2- to 10-minute recordings per pull-through) reflect the 24-hour pathophysiology of the sphincter. Despite some problems, SOM is gaining more widespread clinical application.

Technique and Indications SOM is usually performed at the time of ERCP. All drugs that relax (anticholinergics, nitrates, calcium-channel blockers, and glucagon) or stimulate (narcotics or cholinergic agents) the sphincter should be avoided for at least 8 to 12 hours before manometry and during the manometric session. Data indicate that benzodiazepines do not affect the sphincter pressure and therefore are acceptable sedation for SOM. Meperidine, at a dose of 1 mg/kg, does not affect the basal sphincter pressure (although it does increase the phasic wave frequency). [28] Because the basal sphincter pressure is generally the only manometric criterion used to diagnose SOD and determine therapy, it was suggested that meperidine could be used to facilitate conscious sedation for manometry. If glucagon must be used to achieve cannulation, an 8- to 10-minute (at least) waiting period is required to restore the sphincter to its basal condition. Size 5-French catheters should be used because virtually all standards have been established with these catheters. Triple-lumen catheters are state of the art and are available from several manufacturers. A variety of catheter types can be used. Catheters with a long intraductal tip may help secure the catheter within the bile duct, but such a long nose is commonly a hindrance if pancreatic manometry is desired. Over-the-wire (monorail) catheters can be passed after first securing one's position within the duct with a guidewire. Whether this guidewire influences basal sphincter pressure is unknown. Some triple-lumen catheters accommodate a 0.018-inch diameter guidewire passed through the entire length of the catheter and can be used to facilitate cannulation or maintain position in the duct. A recent study in our unit showed that stiffer-shafted nitinol-core guidewires used for this purpose commonly increase basal sphincter pressure by 50% to 100%. To avoid these artifacts, such wires need to be avoided, or very-soft-core guidewires must be used. Guidewire-tipped catheters are being evaluated. Aspiration catheters in which one recording port is sacrificed to permit both end and side hole aspiration of intraductal juice are highly recommended for pancreatic manometry [29] (Fig. 104-1). Most centers prefer to perfuse the catheters at 0.25 mL/channel using a low-compliance pump. Lower perfusion rates give accurate basal sphincter pressures but not accurate phasic wave information. A new water-perfused sleeve system, similar to that used in the lower esophageal sphincter, awaits more definitive trial in the sphincter of Oddi. [30] The perfusate is generally distilled water, although physiologic saline needs further evaluation.

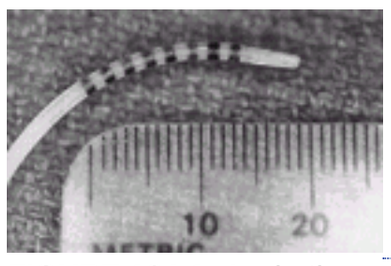

FIGURE 104-1. Aspiration manometry catheter: 5-French manometry catheter (Wilson Cook Model No. SOM-21-S-Lehman, Wilson Cook, Winston-Salem, North Carolina), which allows for aspiration from end and side holes while pressures are accurately recorded from the two remaining ports.

SOM requires selective cannulation of the bile duct or pancreatic duct. The duct entered can be identified by gently aspirating on any port (Fig. 104-2 and Color Fig. 104-2). The appearance of yellow fluid in the endoscopic view indicates entry into the bile duct. Clear aspirate indicates that the pancreatic duct was entered. A small amount of dilute contrast can be injected, but this may hinder accurate intraductal pressure measurements and offers no advantage over aspiration. One must be certain that the catheter is not impacted against the wall of the duct to ensure accurate pressure measurements. Once deep cannulation is achieved and the patient acceptably sedated, the catheter is withdrawn across the sphincter at 1- to 2-mm intervals by standard station pull-through technique. Ideally, both the pancreatic and bile ducts should be studied. Data indicate that an abnormal basal sphincter pressure may be confined to one side of the sphincter in 35% to 65% of patients with abnormal manometry. [31], [32], [33] and [34] Thus, one sphincter may be dysfunctional whereas the other normal. Raddawi and colleagues [31] reported that an abnormal basal sphincter was more likely to be confined to the pancreatic duct segment in patients with pancreatitis and to the bile duct segment in patients with biliary-type pain and elevated liver function tests.

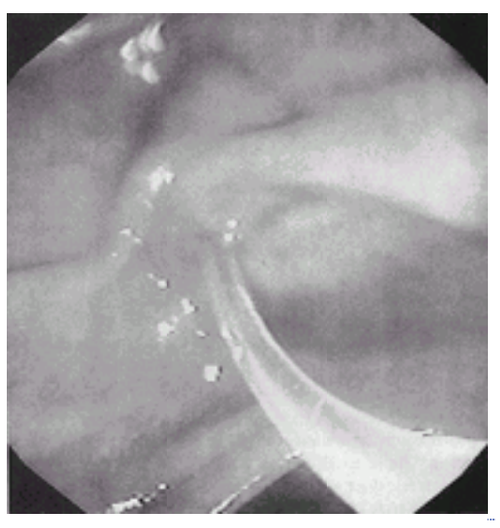

FIGURE 104-2. (See Color Fig. 104-2.) The duct entered during sphincter of Oddi manometry can be identified by aspirating the catheter. Dark-colored fluid (*top right*) signifies entry into the bile duct, whereas clear fluid indicates pancreatic duct entry.

Abnormalities of the basal sphincter pressure should ideally be observed for at least 1 minute in each lead and be seen on two or more separate pull-throughs. From a practical clinical standpoint, we settle for one pull-through (from each duct) if the readings are clearly normal or abnormal. During standard station pull-through technique, it is necessary to establish good communication between the endoscopist and the manometrist who is reading the tracing as it rolls off the recorder. This permits optimal positioning of the catheter to achieve interpretable tracings. Alternatively, electronic manometry systems with a television screen can be mounted near the endoscopic image screen to permit the endoscopist to view the manometry tracing during endoscopy. Once the baseline study is done, agents to relax or stimulate the sphincter can be given (e.g., cholecystokinin) and manometric or pain response monitored. The value of these provocative maneuvers for everyday use needs further study before widespread application is recommended. Criteria for interpretation of an SO tracing are relatively standard; however, they may vary somewhat from center to center. Some areas where there may be disagreement in interpretation include the required duration of basal SO pressure elevation, the number of leads in which basal pressure elevation is required, and the role of averaging pressures from the three (or two in an aspirating catheter) recording ports. [2] Our recommended method for reading the manometry tracings is first to define the zero duodenal baseline before and after the pull-through. Alternatively, intraduodenal pressure can be continuously recorded from a separate intraduodenal catheter attached to the endoscope. Identify the highest basal pressure (Fig. 104-3) that is sustained for at least 30 seconds. Take the four lowest amplitude points in that zone and take the mean of these readings as the basal sphincter pressure for that lead for that pull-through. Average the basal sphincter pressure for all interpretable observations and take this as the final basal sphincter pressure. The amplitude of phasic wave contractions is measured from the beginning of the slope of the pressure increase from the basal pressure to the peak of the contraction wave. Four representative waves are taken for each lead and the mean pressure determined. The number of phasic waves per minute and the duration of the phasic waves can also be determined. Most authorities read only the basal sphincter pressure as an indicator of pathology of the SO. However, data from Kalloo and colleagues [35] suggest that intraductal biliary pressure, which is easier to measure than SO pressure, correlates with SO basal pressure. In this study, intrabiliary pressure was significantly higher in patients with SOD than those with normal SO pressure (20 versus 10 mm Hg; $p < 0.01$). This study needs to be confirmed but supports the theory that increased intrabiliary pressure is a cause of pain in SOD.

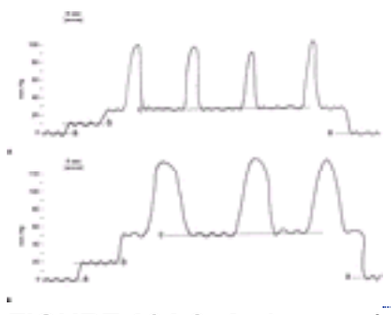

FIGURE 104-3. A: A normal station pull-through of one lead at sphincter of Oddi manometry. The study has been abbreviated to fit onto one page. *(a)* Baseline duodenal 0 reference. *(b)* Intraductal (biliary) pressure of 10 mm Hg (normal). *(c)* Basal biliary sphincter pressure of 28 mm Hg (normal). Phasic waves are about 70 mm Hg in amplitude and less than 6 seconds' duration (normal). **B:** An abnormal station pull-through of one lead at sphincter of Oddi manometry. *(a)* Baseline duodenal 0 reference. *(b)* Intraductal biliary pressure of 20 mm Hg (abnormal). *(c)* Basal biliary sphincter pressure of 52 mm Hg (abnormal). Phasic waves are about 70 mm Hg in amplitude (normal) but have an abnormal duration of 10 to 12 seconds.

The best study establishing normal values for SOM was reported by Guelrud and associates. [36] Fifty asymptomatic control patients were evaluated and repeated on two occasions in 10 subjects. This study established normal values for intraductal pressure, basal sphincter pressure, and phasic wave parameters (Table 104-2). Moreover, the reproducibility of SOM was confirmed. Various authorities interchangeably use 35 mm Hg and 40 mm Hg as the upper limit of normal for mean basal SO pressure.

Basal sphincter pressure*	>35 mm Hg
Basal ductal pressure	>13 mm Hg
Phasic contractions	
Amplitude	>220 mm Hg
Duration	>8 s
Frequency	>10/min

Values were obtained by adding 3 standard deviations to the mean (mean obtained by averaging the results on 2–3 station pull-throughs). Data combine pancreatic and biliary studies.

* Basal pressures determined by (1) reading the peak basal pressure (i.e., highest single lead as obtained using a 3-lumen catheter); (2) obtaining the mean of these peak pressures from multiple station pull-throughs.

From ref. 36.

TABLE 104-2 Suggested Standard for Abnormal Values for Endoscopic Sphincter of Oddi Manometry Obtained from 50 Volunteers without Abdominal Symptoms

Several studies have indicated that pancreatitis is the most common major complication after SOM. [29] Using standard perfused catheters, pancreatitis rates as high as 31% have been reported. Such high complication rates have initially limited more widespread use of SOM. These data also emphasize that manometric evaluation of the pancreatic duct, particularly in patients with chronic pancreatitis, is associated with a high complication rate. Rolny and associates [37] reported an 11% incidence of pancreatitis after pancreatic duct manometry. Twenty-six percent of chronic pancreatitis patients undergoing SOM developed pancreatitis. A variety of methods to decrease the incidence of postmanometry pancreatitis have been proposed. These include (1) using an aspiration catheter; (2) performing gravity drainage of the pancreatic duct after manometry; (3) decreasing the perfusion rate to 0.05 to 0.1 mL/lumen/min; (4) limiting pancreatic duct manometry time to less than 2 minutes (or avoiding pancreatic manometry); (5) using the microtransducer (nonperfused) system [38]; and (6) placing a pancreatic stent after manometry or sphincterotomy. In a prospective randomized study, Sherman and colleagues [29] found that the aspirating catheter (this catheter allows for aspiration of the perfused fluid from end and side holes while accurately recording pressure from the two remaining side ports) reduced the frequency of pancreatic duct manometry-induced pancreatitis from 31% to 4%. The reduction in pancreatitis with the use of this catheter in the pancreatic duct and the very low incidence of pancreatitis after bile duct manometry lend support to the notion that increased pancreatic duct hydrostatic pressure is a major cause of this complication. Thus, when the pancreatic duct sphincter is studied by SOM, aspiration of pancreatic juice is strongly recommended. In a prospective randomized trial, Tarnasky and associates [39] showed that stenting the pancreatic duct decreased post-ERCP pancreatitis from 26% to 6% in a group of patients with pancreatic sphincter hypertension undergoing biliary sphincterotomy alone. This observation, however, was not confirmed by another study. [40] SOM is recommended in patients with idiopathic pancreatitis or unexplained disabling pancreaticobiliary pain with or without hepatic enzyme abnormalities. An attempt is made to study both sphincters, but clinical decisions can be made when the first sphincter evaluated is abnormal. An ERCP is usually performed (if an adequate study is not available) immediately after the SOM to exclude other potential causes for the patient's symptoms. Indications for the use of SOM have also been developed according to the Hogan-Geenen SOD classification system (see Table 104-1). In type I patients, there is a general consensus that a structural disorder of the sphincter (i.e., sphincter stenosis) exists. Although SOM may be useful in documenting SOD, it is not an essential diagnostic study before endoscopic or surgical sphincter ablation. In fact, sometimes SOM may produce misleading results in this group of patients. Such patients uniformly benefit from sphincter ablation regardless of the SOM results (see section " Stent Trial as a Diagnostic Test"). Type II patients demonstrate SO motor dysfunction in 50% to 65% of cases. [41] In this group of patients, SOM is highly recommended because the results of the study predict outcome from sphincter ablation. Type III patients have pancreaticobiliary pain without other objective evidence of sphincter outflow obstruction. SOM is mandatory to confirm the presence of SOD. Although not well studied, it appears that the results of SOM may predict outcome from sphincter ablation in these patients.

Stent Trial as a Diagnostic Test

Placement of a pancreatic or biliary stent on a trial basis in hope of achieving pain relief and predicting the response to more definitive therapy, that is, sphincter ablation, has received only limited application. Pancreatic stent trials, especially in patients with normal pancreatic ducts, are strongly discouraged because serious ductal and parenchymal injury may occur if stents are left in place for more than a few days. [42] Goff [43] reported a biliary stent trial in 21 type II and III SOD patients with normal biliary manometry. Size 7-French stents were left in place for at least 2 months if symptoms resolved and removed sooner if they were judged ineffective. Relief of pain with the stent was predictive of long-term pain relief after biliary sphincterotomy. Unfortunately, 38% of the patients developed pancreatitis (14% were graded severe) after stent placement. Because of this high rate of complications, biliary stent trials are strongly discouraged.

THERAPY FOR SPHINCTER OF ODDI DYSFUNCTION

Therapy for SOD is evolving. Historically, most emphasis has been placed on definitive intervention, that is, surgical sphincteroplasty or endoscopic sphincterotomy. This appears appropriate for patients with high-grade obstruction (type I as per Hogan-Geenen criteria). In patients with lesser degrees of obstruction, the clinician must carefully weigh the risks and benefits before recommending invasive therapy. Most reports indicate that SOD patients have a complication rate from endoscopic sphincterotomy of at least twice that of patients with ductal stones. [40], [44]

Medical Therapy

Medical therapy for documented or suspected SOD has received only limited study. Because the SO is a smooth muscle structure, it is reasonable to assume that drugs that relax smooth muscle might be an effective treatment for SOD. Sublingual nifedipine and nitrates have been shown to reduce the basal sphincter pressures in asymptomatic volunteers and symptomatic patients with SOD. [1], [45] Khuroo and colleagues [46] evaluated the clinical benefit of nifedipine in a placebo-controlled crossover trial. Twenty-one of 28 patients (75%) with manometrically documented SOD had a reduction in pain scores, emergency room visits, and use of oral analgesics during short-term follow-up. In a similar study, Sand and associates [47] found that 9 of 12 (75%) type II SOD (suspected; SOM was not done) patients improved with nifedipine. Although medical therapy may be an attractive initial approach in patients with SOD, several drawbacks exist. [1] First, medication side effects may be seen in up to one third of patients. Second, smooth muscle relaxants are unlikely to be of any benefit in patients with the structural form of SOD (i.e., SO stenosis), and the response is incomplete in patients with a primary motor abnormality of the SO (i.e., SO dyskinesia). Finally, long-term outcome from medical therapy has not been reported. Nevertheless, because of the relative safety of medical therapy and the benign (although painful) character of SOD, this approach should be considered in all type III and less severely symptomatic type II SOD patients before considering more aggressive sphincter ablation therapy. Guelrud and colleagues [48] have demonstrated that transcutaneous electrical nerve stimulation lowers the basal sphincter pressure in SOD patients by a mean of 38% but generally not into the normal range. This stimulation was associated with an increase in serum vasoactive intestinal peptide levels.

Surgical Therapy

Surgery was the traditional therapy of SOD. The surgical approach, most commonly, is a transduodenal biliary sphincteroplasty with a transampullary septoplasty (pancreatic septoplasty). Sixty to 70% of patients were reported to have benefited from this therapy during a 1- to 10-year follow-up. [49], [50] Patients with an elevated basal sphincter pressure, determined by intraoperative SOM, were more likely to improve from surgical sphincter ablation than those with a normal basal pressure. [50] Some reports have suggested that patients with biliary-type pain have a better outcome than patients with idiopathic pancreatitis whereas others suggested no difference. [49], [50] However, most studies found that symptom improvement after surgical sphincter ablation alone was relatively uncommon in patients with established chronic pancreatitis. [50]

The surgical approach for SOD has largely been replaced by endoscopic therapy. Patient tolerance, cost of care, morbidity, mortality, and cosmetic results are some of the factors that favor an initial endoscopic approach. Surgical therapy is reserved for patients with restenosis after endoscopic sphincterotomy and when endoscopic evaluation or therapy is not available or technically feasible (e.g., Roux-en- gastrojejunostomy).

Endoscopic Therapy

Endoscopic Sphincterotomy Endoscopic sphincterotomy is the standard therapy for patients with SOD. Most data on endoscopic sphincterotomy relate to biliary sphincter ablation alone. Clinical improvement after therapy has been reported to occur in 55% to 95% of patients (see Table 104-1). These variable outcomes are reflective of the different criteria used to document SOD, the degree of obstruction (type I biliary patients appear to have a better outcome than type II and III), the methods of data collection (retrospective versus prospective), and the techniques used to determine benefit. Rolny and colleagues [51] studied 17 type I postcholecystectomy biliary patients by SOM. In this series, 65% had an abnormal SOM (although not specifically stated, it appears that the biliary sphincter was studied alone). Nevertheless, during a mean follow-up interval of 2.3 years, all patients benefited from biliary sphincterotomy. The results of this study suggested that because type I biliary patients invariably benefit from biliary sphincterotomy, SOM in this patient group is not only unnecessary but also possibly misleading. The results of this study, however, have never been validated at another center. Although most of the studies reporting efficacy of endoscopic therapy in SOD have been retrospective, two notable randomized trials have been reported. In a landmark study by Geenen and associates, [49], [52] postcholecystectomy, type II, biliary patients were randomized to biliary sphincterotomy or sham sphincterotomy. SOM was performed in all patients but not used as a criterion for randomization. During a 4-year follow-up, 95% of patients with an elevated basal sphincter benefited from sphincterotomy. In contrast, only 30% to 40% of patients with an elevated sphincter pressure treated by sham sphincterotomy, or with a normal sphincter pressure treated by endoscopic sphincterotomy or sham sphincterotomy, benefited from this therapy. The two important findings of this study were that SOM predicted the outcome from endoscopic sphincterotomy and that endoscopic sphincterotomy offered long-term benefit in type II biliary patients with SOD. Confirming data were seen in a 2-year follow-up study by Toouli and colleagues. [53] Sherman and associates [54] reported their preliminary results of a randomized study comparing endoscopic sphincterotomy and surgical biliary sphincteroplasty with pancreatic septoplasty (with or without cholecystectomy) to sham sphincterotomy for type II and III biliary patients with manometrically documented SOD. The results are shown in Table 104-3A and Table 104-3B. During a 3-year follow-up period, 69% of patients undergoing endoscopic or surgical sphincter ablation improved, compared with 24% in the sham sphincterotomy group ($p = 0.009$). There was a trend for type II patients to benefit more frequently from sphincter ablation than type III patients: 13 of 16 (81%) versus 11 of 19 (58%) ($p = 0.14$). Evidence is now accumulating that the addition of a pancreatic sphincterotomy to an endoscopic biliary sphincterotomy in such patients may improve the outcome. Long-term outcome studies, preferably in randomized trials, are awaited.

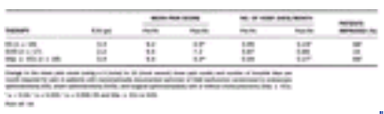

TABLE 104-3A Randomized Controlled Trial for Sphincter of Oddi Dysfunction: Outcome of Therapies

SOD TYPE*	NO. OF PATIENTS IMPROVED/TOTAL NO. OF PATIENTS		
	ES	S-ES	Slp ± CCx
Type II	5/6 (83%)†	1/7 (14%)	8/10 (80%)†
Type III	8/13 (62%)	3/10 (30%)	3/6 (50%)

*SOD type based on Hogan-Geenen SOD Classification System.
†$p < 0.02$; ES and Slp ± CCx vs S-ES.
From ref. 54.

TABLE 104-3B Clinical Benefit Correlated with Sphincter of Oddi Dysfunction (SOD) Type

These results clearly indicate that the response rate and enthusiasm for sphincter ablation must be correlated with patient presentation and balanced against the high complication rates reported for endoscopic therapy of SOD. Most studies indicate that patients undergoing endoscopic sphincterotomy for SOD have complication rates two to three times higher than patients undergoing endoscopic sphincterotomy for ductal stones. [44] Pancreatitis is the most common complication, occurring in up to 20% of patients. At least in one study, the frequency of complications appears to be dependent on the bile duct diameter. [40], [55] Endoscopic techniques are being developed (e.g., pancreatic duct stenting before combined pancreaticobiliary sphincterotomy) to limit such complications. [39]

Balloon Dilation and Stenting

Balloon dilation of strictures in the gastrointestinal tract has become commonplace. In an attempt to be less invasive and possibly preserve sphincter function, adaptation of this technique to treat SOD has been described. Unfortunately, because of the unacceptably high complication rates, primarily pancreatitis, this technology has little role in the management of SOD. [56] Similarly, although biliary stenting might offer short-term symptom benefit in patients with SOD and predict outcome from sphincter ablation, it too has unacceptably high complication rates and cannot be advocated in this setting [43] (see previous section, “ Stent Trial as a Diagnostic Test”).

Botulinum Toxin Injection

Botulinum toxin, a potent inhibitor of acetylcholine release from nerve endings, has been successfully applied to smooth muscle disorders of the gastrointestinal tract such as achalasia. In a preliminary clinical trial, toxin injection into the SO resulted in a 50% reduction in the basal biliary sphincter pressure and improved bile flow. [57] This reduction in pressure may be accompanied by symptom improvement in some patients. A small series of patients [38] showed an excellent correlation between pain resolution with botulinum toxin injection into the major papilla and symptom resolution with subsequent biliary sphincterotomy. Such an approach requires two endoscopies to achieve symptom relief. Further studies are needed before recommending this technique.

SPHINCTER OF ODDI DYSFUNCTION IN RECURRENT PANCREATITIS

SOD has been manometrically documented in 15% to 59% of patients with recurrent pancreatitis, previously labeled as idiopathic. [15] Biliary sphincterotomy alone has been reported to prevent further pancreatitis episodes in more than 50% of such patients. From a scientific, but not practical, viewpoint, care must be taken to separate out subtle biliary pancreatitis, [58] which will similarly respond to biliary sphincterotomy. The value of ERCP, SOM, and sphincter ablation therapy was studied in 51 patients with idiopathic pancreatitis. [59] Twenty-four (47.1%) had an elevated basal sphincter pressure. Thirty were treated by biliary sphincterotomy ($n = 20$), or surgical sphincteroplasty with septoplasty ($n = 10$). Fifteen of 18 patients (83%) with an elevated basal sphincter pressure had long-term benefit (mean follow-up, 38 months) from sphincter ablation therapy (including 10 of 11 treated by biliary sphincterotomy), in contrast to only 4 of 12 (33.3%; $p < 0.05$) with a normal basal sphincter pressure (including 4 of 9 treated by biliary sphincterotomy). Guelrud and colleagues, [60] however, found that severance of the pancreatic sphincter was necessary to resolve the pancreatitis. In this series, 69 patients with idiopathic pancreatitis caused by SOD underwent treatment by standard biliary sphinctero- tomy ($n = 18$), biliary sphincterotomy with pancreatic sphincter balloon dilation ($n = 24$), biliary sphincterotomy followed by pancreatic sphincterotomy in separate sessions ($n = 13$), or combined pancreatic and biliary sphincterotomy in the same session ($n = 14$). Eighty-one percent of patients undergoing pancreatic and biliary sphincterotomy had resolution of their pancreatitis, compared with 28% of patients undergoing biliary sphincterotomy alone ($p < 0.005$). These data are consistent with the theory that many such patients who benefit from biliary sphincterotomy alone have subtle gallstone pancreatitis. The results of Guelrud and colleagues [60] also support the anatomic findings of separate biliary and pancreatic sphincters, and the manometry findings of residual pancreatic sphincter hypertension in more than 50% of persistently symptomatic patients who undergo biliary sphincterotomy alone. The best method to treat residual pancreatic sphincter stenosis (after biliary sphincterotomy) awaits further study. Patients with idiopathic pancreatitis who fail to respond to biliary sphincterotomy alone should have their pancreatic sphincter reevaluated and be considered for sphincter ablation if residual high pressure is found.

FAILURE TO ACHIEVE SYMPTOMATIC IMPROVEMENT AFTER BILIARY SPHINCTEROTOMY

Table 104-4 lists several potential explanations as to why patients may fail to achieve symptom relief after biliary sphincterotomy is performed for well-documented SOD. First, the biliary sphincterotomy may have been inadequate, or restenosis may have occurred. Although the biliary sphincter is commonly not totally ablated, [61] Manoukian and associates [62] indicate that clinically significant biliary restenosis occurs relatively infrequently. If no cutting space remains in such a patient, balloon dilation to 8 to 10 mm may suffice, but long-term outcome from such therapy is unknown.

1. Residual or recurrent biliary sphincter dysfunction
2. Pancreatic sphincter (major papilla) dysfunction
3. Chronic pancreatitis—subtle, pancreatogram normal
4. Other obstructive pancreatobiliary pathology (stones, strictures, tumor, pancreas divisum)
5. Nonpancreatobiliary disease—especially gut motor disorders or visceral hypersensitivity.

TABLE 104-4 Causes for Failure to Achieve Symptom Relief after Biliary Sphincterotomy for Sphincter of Oddi Dysfunction

Second, the importance of pancreatic sphincter ablation is being increasingly recognized, as noted in the data preliminarily reported by Guelrud and colleagues. [60] Soffer and Johlin [63] reported that 25 of 26 patients (mostly type II), who failed to respond to biliary sphincterotomy, had elevated pancreatic sphincter pressure. Pancreatic sphincter therapy was performed with overall symptomatic improvement in two thirds of patients.

Third, patients may fail to respond to sphincterotomy because they have chronic pancreatitis. These patients may or may not have abnormal pancreatograms. Intraductal pancreatic juice aspiration, after secretin stimulation, may help make this diagnosis. [64], [65] Endoscopic ultrasound may show parenchymal and ductular changes of the pancreas in some of these patients, suggesting chronic pancreatitis. [66], [67]

Fourth, some patients may have pain from altered gut motility or hyperalgesia [68] of the stomach, small bowel, or colon (irritable bowel or pseudoobstruction variants). There is increasing evidence that upper gastrointestinal motility disorders [69], [70] and visceral hyperalgesia [71] may masquerade as pancreatobiliary-type pain (i.e., discrete right upper quadrant pain). Multiple preliminary studies show disordered duodenal motility in such patients. [69], [70] This area needs further study to determine the frequency, significance, or coexistence of these motor disorders associated with SOD.

SUMMARY

Our knowledge of SOD and the manometric techniques available to assist in this diagnosis are evolving. Successful endoscopic SOM requires good general ERCP skills and careful attention to the main details listed previously. If SOD is suspected in a type III or mild to moderate pain level type II patient, medical therapy should generally be tried. If medical therapy fails or is bypassed, ERCP and manometric evaluation are recommended. The role of less invasive studies remains uncertain owing to undefined sensitivity and specificity. Sphincter ablation is generally warranted in symptomatic type I patients and type II and III patients with abnormal manometry. The symptom relief rate varies from 55% to 95%, depending on the patient presentation and selection. Initial nonresponders may require thorough pancreatic sphincter and pancreatic parenchymal evaluation. More studies are needed to evaluate this complex issue. SOD patients have relatively high complication rates after invasive studies or therapy. Thorough review of the risk-to-benefit ratio with individual patients is mandatory.

Acknowledgments

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CHAPTER 105

Gary C. Kanel

ANATOMY, MICROSCOPIC STRUCTURE, AND CELL TYPES OF THE LIVER

The liver is a unique organ having a wide range of both structural and physiologic functions. Understanding the arrangements of all the individual components of the liver on both gross and light microscopic examination assisted by electron microscopic and immunoenzymatic features allows a better understanding of liver histopathology and pathophysiology. Study of the embryologic evolution of the hepatic and hepatobiliary elements also assists in understanding many of the key concepts in developmental abnormalities seen in a wide variety of both pediatric and adult liver diseases. This chapter outlines the gross and microscopic characteristics of the liver and briefly addresses liver function, with an introductory presentation of embryologic development.

EMBRYOLOGY

The hepatic primordium anlage first appears toward the end of the third week of gestation and is seen as a hollow midline outgrowth stalk (hepatic diverticulum) of the endodermal epithelium at the distal aspect of the foregut (future duodenal loop). [1], [2], [3], [4] and [5] By the fourth week, the diverticulum enlarges by way of proliferation of the endodermal cell strands (hepatoblasts) and projects cranially into the mesoderm of the septum transversum (the mesodermal plate between the pericardial cavity and the yolk sac stalk), eventually giving rise to the hepatic parenchyma and intrahepatic duct structures (Fig. 105-1). The cephalic end ultimately develops into the right and left hepatic lobes. The stalk between the diverticulum and foregut narrows, forming the extrahepatic biliary system and gallbladder. The proliferating endodermal cells initially form solid cords, which then anastomose and form isolated vesicles and cribriform tubules with centrally located lumenal structures (biliary canaliculi). The cords eventually become confluent and form channels that subdivide the cords by small capillaries, which co-mingle with the developing vitelline and umbilical veins. These channels ultimately form the hepatic sinusoids. The individual hepatoblasts develop into the mature hepatocyte, with those cells immediately adjacent to the portal mesenchyme becoming the ductal plates. [6] The rapid growth rate of the hepatic cords allows sheets several cells thick, termed *muralium multiplex* to develop; these persist until birth, after which the cell sheets narrow to two cells (*muralium duplex*) and eventually evolve into a one cell thick trabecular cord (*muralium simplex*) by 5 years of age. [7], [8]

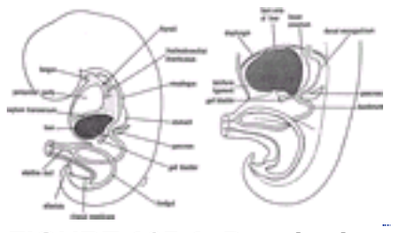

FIGURE 105-1. Developing Embryo. The drawing to the *left* represents a 9-mm embryo, estimated at about 36 days' gestation. The liver is derived from the hepatic diverticulum (midline outgrowth stalk of the distal foregut) and projects cranially into the septum transversum and caudally into the abdominal wall. The drawing to the *right* represents an embryo that is slightly older, with the falciform ligament noted between the hepatic parenchyma and the anterior abdominal wall. (From ref. [3].)

The mesoderm of the septum transversum initially surrounds the liver and is in continuity with the lesser curvature of the stomach, duodenum, and ventral body wall. The mesoderm becomes stretched between the liver and lesser curvature, forming the lesser omentum, and between the diaphragm and ventral abdominal wall, forming the falciform, coronary, and triangular ligaments. A portion also develops into the hepatic (Glisson) capsule. The developing hepatic artery and vagus nerve branch follow the mesoderm along and adjacent to the portal vein. The mesoderm on the liver surface also is in continuity with the peritoneum, whereas that portion in contact with the future diaphragm remains uncovered ("bare area"). The mesoderm is the main focus for the development of hematopoiesis, which begins at about 6 weeks and becomes most active during the sixth and seventh months, then rapidly regresses owing to developing bone marrow activity. The hematopoiesis is responsible for the prominent liver size in the fetus (up to 10% body weight by week 10, with the liver occupying most of the abdominal cavity) but regresses at birth so that only a few islands of hematopoietic cells are noted. The liver then weights about 5% of body weight. Hematopoiesis is usually absent by 4 weeks of age. Initially, the enlarged fetal liver has symmetric right and left lobes; with time, however, the growth rate diminishes but affects the left lobe more than the right, causing the size difference seen at birth. The caudate and quadrate lobes develop as subdivisions of the right lobe.

The *vascular network* is originally derived from the development of both the vitelline and umbilical veins and occurs at the same time as proliferation of the hepatoblasts. The cords and vessels anastomose, forming the hepatic sinusoids. The sinusoidal plexus initially receives blood through the vitelline vein and is drained into the sinus venosus, but by the fifth week, the right and left umbilical veins also supply blood to the sinusoidal plexus. By the fifth week, most of the major vessels are identified and include the right and left umbilical vein, the transverse portal sinus, and the ductus venosus, which shunts the blood directly from the umbilical vein into the inferior vena cava. At this time, the left umbilical vein becomes the main source of blood flow, with the right umbilical vein becoming atrophic by the fifth week. The portal vein develops from the vitelline vein and then subdivides into the right and left branches. The hepatic and portal vein branches divide the parenchyma into lobules and acini. The left umbilical vein blood flow has three major branching routes: (1) through the sinusoidal plexus of the left half of the liver, (2) through the sinusoidal plexus of the right half of the liver by retrograde flow from connections with the left branch of the portal vein, and (3) through the ductus venosus. At birth, a sphincter mechanism closes the ductus venosus, causing cessation of blood flow through the umbilical vein, with the liver now receiving blood from the left branch of the portal vein. The resultant closed segment of the umbilical vein then becomes the umbilicus, and the fibrotic ductus venosus becomes the ligamentum venosum.

The *biliary apparatus* develops from membranous infolding occurring between the junctional complexes between individual hepatoblasts and appears initially as intercellular spaces with no distinct wall. The biliary canaliculi are first seen at week 6, with bile synthesis occurring by week 9 and bile secretion by week 12. The hepatoblasts immediately adjacent to the mesenchyme of the portal tracts form the ductal plate, which is two layered; by 3 months, a lumen is seen within the ductal plate, with formation of double-layered tubular structures. Invasion of these cells into the portal mesenchyme occurs, forming an anastomosing network of portal duct structures. These ducts are not fully developed at term, [9] after which remodeling forms the true interlobular bile ducts that assume their major role as a linkage between the canalicular biliary apparatus and the extrahepatic biliary tree. This biliary network is fed by a hypervascular complex of arterioles and capillaries formed from the peribiliary plexus. The immature structures at birth may persist for up to 1 months. The extrahepatic biliary tree develops from the stalk of the original hepatic outgrowth.

The individual cell functions become apparent at different but early times in the embryologic development. [7], [10], [11] The alpha-fetoprotein, which is in high quantities at birth, initially is present by 1 month's gestation. Glycogen may be seen by 2 months, with glycogen synthesis becoming most apparent by 3 months; at birth, the amount of glycogen rapidly diminishes owing to rapid and active glycogenolysis. Fatty change within the hepatocyte also parallels that of glycogenesis. Hemosiderin is usually visible in early stages and becomes most marked as intrahepatic hematopoiesis decreases; it then gradually decreases but may still be seen at birth in the

periportal hepatocytes. The perisinusoidal cells and Kupffer cells appear by 3 months' gestation.

GROSS ANATOMY

The liver takes up the majority of the right upper abdominal cavity and extends from the right lateral aspect of the abdomen 15 to 20 cm transversely toward the xiphoid. [1], [12], [13] and [14] The weight of the adult liver varies from 1200 to 1800 g, dependent on the overall body size, and constitutes about 1.8% to 3.1% of the total body weight; however, at birth, the liver is larger compared to adjacent thoracic and abdominal viscera and constitutes about 5% to 6% of the body weight. [1], [15], [16] The liver anatomically has four lobes: right, left, caudate, and quadrate. The right lobe accounts for one half to two thirds the total liver volume; however, functionally, the right and left lobes are of about equal size and are divided by a line extending from the inferior vena cava superiorly to the middle of the gallbladder fossa inferiorly. A total of eight functional segments are present, each demarcated by the vascular and biliary drainage [1], [7], [12], [17], [18] and [19] (Fig. 105-2): the lateral (segments VI and VII) and medial (segments V and VIII) divisions of the right lobe, the medial (segment IV) and lateral (segments II and III) divisions of the left lobe, and the caudate lobe (segment I), the latter a "watershed" area of the right and left lobe vasculature.

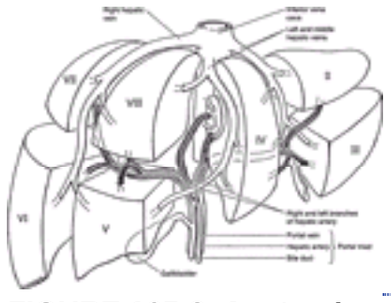

FIGURE 105-2. Anatomic and Functional Subdivisions. The liver is divided into eight functional anatomic segments, each having its own vascular flow and biliary drainage. These include the right lateral (segments VI and VII) and medial (segments V and VIII) divisions, the left medial (segment IV) and lateral (segments II and III) divisions, and the caudate lobe (segment I). (From Moore KL, Dalley II AF. Clinically oriented anatomy. 4th ed. Philadelphia: Lippincott Williams & Wilkins, 1999.)

The hepatoduodenal ligament connects the liver to the superior aspect of the duodenum and supports the hilar vessels and duct structures. The transverse fissure separates anteriorly the right lobe from the caudate lobe, whereas the umbilical fissure is located to the left of the quadrate lobe, which itself is bordered on the right by the gallbladder. The peritoneal layers forming the falciform ligament, which extends between the liver and the anterior abdominal wall, separate to form the superior layer of the coronary ligament and the left triangular ligament. The ligamentum teres is located along the lower edge of the falciform ligament and contains the obliterated umbilical vein remnant. The total surface area of the liver is structured by direct continuity with the surrounding abdominal organs, ligaments, and fascia.

The *portal vein* is the main route of vascular drainage of the gastrointestinal tract and is formed through the merger of the superior mesenteric and splenic veins. It also receives blood from the coronary and cystic veins. The portal vein is located along the hepatoduodenal ligament posterior to the hepatic artery and common bile duct and ends at the porta hepatis at the main lobar fissure, dividing into the right and left main branches. The right branch divides early into anterior and posterior segments, whereas the left branch divides into two segments: the *pars transversus* which extends to the left in the porta hepatis, and the *pars umbilicus* which descends into the umbilical fossa in line with the left segmental fissure. The caudate lobe veins arise from both the right and left main portal vein branches (Fig. 105-3).

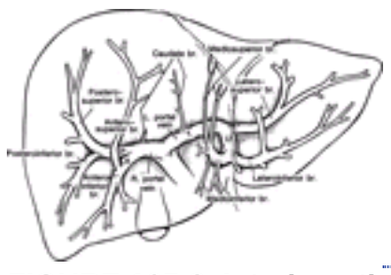

FIGURE 105-3. Intrahepatic Branches of the Portal Vein. T, pars transversus; U, embryonic ductus venosus; P, posterior segment; A, anterior segment. (From Skandalakis LJ, Colborn GL, Gray SW, et al. Surcical anatomy of the liver and extrahepatic biliary tract. In: Nyhus LM, Baker RJ, eds. Mastery of surgery. 2nd ed. Boston: Little, Brown, 1992.)

The *hepatic vein* is composed of three major tributaries (right, middle, left), each having intrahepatic branches. The middle and left hepatic eins often converge to form a single outflow vessel before draining into the inferior vena cava, whereas the right hepatic vein opens through a separate ostium. The caudate lobe drains directly into the inferior vena cava.

The *hepatic artery* is a branch of the celiac artery and ascends along the hepatoduodenal ligament and eventually divides into the right and left main branches. The right hepatic artery is usually seen behind the common hepatic duct after giving rise to the cystic artery, and it eventually divides into the anterior and posterior segmental branches. The left hepatic artery obliquely passes upward and to the left in the porta hepatis, eventually dividing into the medial and lateral segmental branches. The quadrate lobe is fed by a middle hepatic artery branch, whereas the caudate lobe is fed by both right and left hepatic artery branches.

The *biliary system* originally arises from the bile canaliculi and can grossly be demonstrated in the larger interlobular branches. The biliary drainage of the right lobe is derived from anterior and posterior segmental branches that merge to form the right hepatic duct. Lateral and medial segmental branches merge to form the left hepatic duct that drains the left lobe. The caudate lobe is drained from three duct branches directly into the right and left hepatic ducts (Fig. 105-4). The smaller ducts do not have a distinct wall, but the larger septal branches demonstrate a thin wall secondary to periductal collagen fibers and can more easily be demonstrated on gross inspection. The bile duct structures are directly fed by the hepatic artery and its branches, which parallel the ducts as they course through the various hepatic segments.

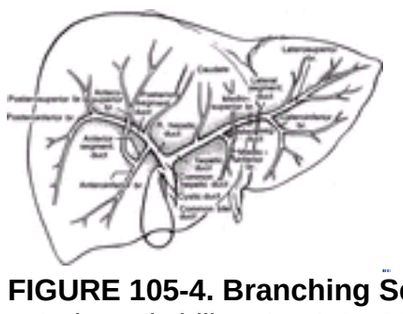

FIGURE 105-4. Branching Segments of the Intrahepatic Bile Ducts. (From Skandalakis LJ, Colborn GL, Gray SW, et al. Surgical anatomy of the liver and extrahepatic biliary tract. In: Nyhus LM, Baker RJ, eds. Mastery of surgery. 2nd ed. Boston: Little, Brown, 1992.)

The *lymphatic channels* provide both sympathetic and parasympathetic innervation and are divided into *deep* and *superficial* branches. The deep lymphatic branch parallels the portal and hepatic vein branches, whereas the superficial lymphatic structures arise from the Glisson's capsule and drain through the adjacent falciform ligament, diaphragm, esophagus, and hilar lymph nodes. [1], [20]

The *nerve supply* parallels the main hepatic artery and portal vein and is divided into *parasympathetic* and *sympathetic* fibers. The former are preganglionic and derived from the vagus nerve, whereas the latter are postganglionic and receive their nerve supply from T7 through T10. The nerve supply enters the hepatic hilum through both anterior and posterior routes and feeds the arteries and bile ducts through sympathetic innervation. The nerve fibers branch through the main portal tracts, with smaller unmyelinated branches feeding the periportal hepatocytes. In addition, many of the nerve fibers terminate on endothelial cells lining the smallest arterioles and along Kupffer cells, stellate (fat-storing) cells, and hepatocytes. [21]

MICROANATOMY

Normal Histology

Architecture The basic architectural arrangement of the portal structures, sinusoids, and outflow vessels is evenly spaced throughout all lobes of the liver and is best assessed under low- and medium-power microscopy. [22], [23] Although the portal inflammatory and cellular components and the size of the liver cell plates vary, dependent on the developmental stage of the liver, the architectural framework can be seen in fetal liver as well. Although the architecture may at times be difficult to assess when acute liver injury is present, upon recovery, the arrangements universally return to normal; on the other hand, distortion of the architecture seen in advanced chronic liver disease is for the most part irreversible.

Portal Tracts The portal tracts are often referred to as *portal triads;* this term is misleading in that more than three components are present. The *interlobular bile ducts* number from one to two per portal structure, although in infants, the ducts early on appear to be slightly less frequent. The ducts are usually seen immediately adjacent to the *hepatic arterioles* which are responsible for their blood supply. The hepatic arterioles are usually singly present. The *portal venules* similarly are a single structure, although transverse cuts of the portal tracts in biopsy material can mistakenly appear as if two or more ducts and vessels are present. True increase in portal venules and lymphatics are characteristic of portal hypertension and are usually seen in conjunction with portal fibrosis or cirrhosis. The *fibrous tissue* that supports the major portal components varies in amount, dependent on the distance of the portal tract from the hepatic hilum, and errantly can be felt to be "fibrotic" when the biopsy specimen is taken toward the hilum. The infiltrating *cellular inflammatory components* within the fibrous tissue consist of scattered lymphocytes, which are scanty but often present to some degree even in normal livers.

Parenchyma In adults, the hepatic lobules constitute about 80% of the total hepatic volume and are composed predominantly of *liver cell cords* one cell thick made up of polyhedral hepatocytes. The adjacent sinusoids are lined by both *endothelial cells* and *Kupffer cells*. The perisinusoidal space is located between the endothelial cells and hepatocytes. *Stellate cells* and *collagen fibers* are also present along the perisinusoidal space, but on routine hematoxylin and eosin staining of normal liver tissue, these structures are usually inconspicuous. The sinusoids drain from the portal venule and hepatic arterioles into the terminal hepatic venules (Fig. 105-5).

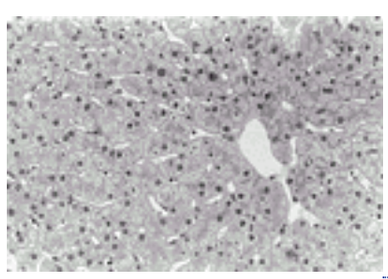

FIGURE 105-5. Parenchyma. The liver cell plates are one cell thick and are composed of polyhedral hepatocytes with centrally placed nuclei. These cells are bordered by sinusoids lined by both Kupffer and endothelial cells. The sinusoids directly drain into the terminal hepatic (central) venules.

Parenchymal Cellular Components

Hepatocyte The *hepatocyte* composes about two thirds of the total number of cells within the liver and about four fifths of the total liver volume. [1], [24], [25] and [26] The cells on the average measure from 25 to 40 µm in diameter, dependent on their zonal location and patient age, and are polyhedral and multifaceted. The cells in the adult are arranged in cords that are one cell thick and have three distinct cell boundaries: *sinusoidal lateral* (intercellular), and *canalicular* membranes. [7], [24] The *sinusoidal* or basolateral surface area constitutes about 60% to 70% of the total surface area. Microvilli measuring about 0.5 µm in length can be seen along the surface extending into the perisinusoidal spaces. The *lateral* membranes lie between adjacent hepatocytes and constitute about 15% of the surface area. They are divided into a number of junctional subunits. [1], [24] The *gap junctions* represent the two adjacent membranes having a distinct 2- to 4-µm gap, whereby communication between cells for transport of metabolites occurs by way of projected particles ("bobbins") directed from each cellular membrane. No microvilli are seen. The *desmosomes* are attached to the intermediate filaments and play a role in the resilience of the cell membrane. The *intermediate junctions* are attached to various cytokeratin filaments that also aid in cellular resilience. The *tight junctions* are zones of contacted membranes and represent a permeability barrier to macromolecules. The *canalicular* membranes constitute about 15% of the membrane surface area, lie toward the center of the intercellular junction, and represent the origin of biliary drainage (Fig. 105-6 and Fig. 105-7). They are lined by microvilli and measure in diameter from 0.5 µm in the perivenular zone (zone 3 of Rappaport) to 2.5 µm in the periportal zone (zone 1).

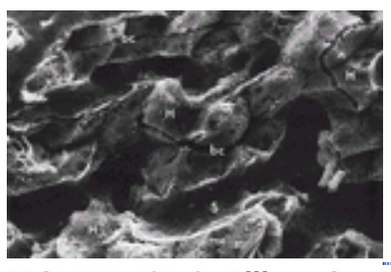

FIGURE 105-6. Biliary Canaliculi. By scanning electron microscopy, the liver cell plates and adjacent sinusoids (*S*) are seen with the bile canalicular (*bc*) network located between the adjacent hepatocytes (*H*). (From ref. [24].)

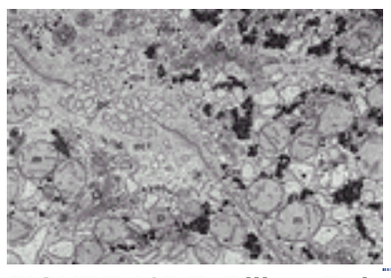

FIGURE 105-7. Biliary Pole. By electron microscopy, the bile canaliculi (*bc*) can be seen to the left of the field, surrounded by microvilli (*mv*) and pericanalicular ectoplasm (*ect*). Golgi complexes (*G*), vesicles (*ves*), mitochrondria (*m*), peroxisomes (*p*), and lysosomes (*lys*) can also be seen in the adjacent liver cell cytoplasm. (From ref. [24].)

Nucleus. The liver cell *nucleus* is centrally located within the hepatocytes, measures about 10 µm in diameter, and contains clumped chromatin and nucleoli. [24], [27] The nuclear membrane is composed of two envelopes separated by a narrow zone. These membranes have numerous apertures or pores that provide

cytoplasm-nuclear communication. The outer membrane often is lined by ribosomes, which are in direct communication with adjacent rough endoplasmic reticulum fibers. The intranuclear chromatin fibers are composed of *heterochromatin* which is round to irregular dense metabolically inactive granules, and *euchromatin* which actively transcribes RNA. [24] Anywhere from one to six nucleoli can be seen, their number and size in direct proportion to their degree of activity (ribosomal RNA function). The hepatocyte usually contains one nucleus, although bilobed forms may also be seen that are more often identified in the elderly population. Mitotic figures are infrequent but may also be prominent in instances of active liver cell regeneration (e.g., resolving acute viral hepatitis). Various nuclear inclusions may be identified [24], [28], [29] and include glycogen (frequently seen in diabetic patients), lipid, and cytoplasmic invaginations (pseudoinclusions, or nuclear membrane irregularities).

Cytoplasm. The liver cell *cytoplasm* constitutes about 90% of the volume of the hepatocytes and contains numerous functionally important organelles (Fig. 105-8). The cytoplasm and intracellular components vary in size, ultrastructure, and function, dependent on the zonal location of the hepatocytes (see " Functional Components: Liver Cell Heterogeneity" and Table 105-1 and Table 105-2). The superstructure is maintained by the cytoskeleton of the hepatocyte and includes three major subdivisions: the microfilaments, microtubules, and intermediate filaments. [7], [24], [30], [31] and [32]

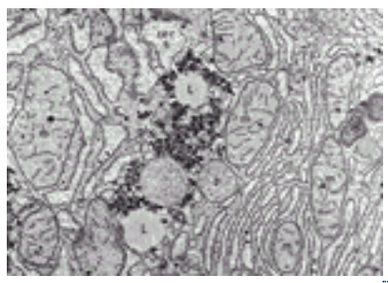

FIGURE 105-8. Liver Cell Cytoplasm). As seen by electron microscopy, the cytoplasm is composed of rough endoplasmic reticulum (*rer*), mitochondria (*m*), glycogen (*gly*), lipid droplets (*L*), and peroxisomes (*p*). (From ref. [24].)

TABLE 105-1 Liver Cell Structural Zonal Variations

ZONE 3 (PERIVENULAR)	ZONE 1 (PERIPORTAL)
Glycolysis	Gluconeogenesis
Glycogen synthesis from glucose	Glycogen synthesis from lactate
Lipogenesis	β Oxidation of fatty acids
Removal of ammonia from blood by glutamine	Amino acid catabolism
Detoxification, biotransformation of the majority of drugs and toxins*	Urea synthesis
Ketogenesis	Cholesterol synthesis
Bile acid synthesis	Bile acid secretion
Bile salt–independent fraction of bile formation; bile acid uptake (sodium independent)	Bile salt–dependent fraction of bile formation; bile acid uptake (sodium dependent)
Glucuronidation	
Mixed-function oxidase	
Increase in Kupffer cell phagocytic activity	

* Note that certain drugs and toxins (e.g., allyl formate, phosphorus) are metabolized and may cause liver cell injury in zone 1 because of different pathophysiologic mechanisms.

TABLE 105-2 Liver Cell Functional Zonal Variations

The *mitochondria* are one of the most prominent intracellular organelles, [7], [24] averaging up to 2200 per hepatocyte. They have numerous critical functions that include oxidative phosphorylation and fatty acid oxidation and contain components essential for the urea and citric acid cycles. They are larger in the perivenular zone but more numerous in the periportal zones and generally maintain a constant volume within each hepatocyte. They are oval to oblong in shape, and measure anywhere from 0.4 to 3.5 µm in diameter. They may be divided into outer and inner membranes separated by a gap. The *endoplasmic reticulum* [24], [33] is composed of a convoluted network of cisternae, saccules, tubules, and vesicles that are distributed throughout the liver cell cytoplasm and constitute about 15% of the cytoplasmic volume. It is divided into two components, the *rough endoplasmic reticulum* and *smooth endoplasmic reticulum.* The rough endoplasmic reticulum is seen predominantly surrounding the nucleus and biliary channels, whereas the smooth endoplasmic reticulum forms a meshwork of small tubules that are devoid of ribosomes and often communicate with the Golgi apparatus. The *Golgi apparatus* [7], [24], [33], [34] is composed of highly polarized parallel flattened dilated saccules or vesicles, is about 1 µm in diameter, and is most often present adjacent to the liver cell nucleus and biliary canaliculi. Anywhere from 40 to 60 Golgi can be seen within the cytoplasm. The Golgi function includes production of secretory vacuoles for transportation and excretion of synthesized proteins into the sinusoids, bile secretion, incorporation of carbohydrates to proteins, and membrane synthesis and repair. *Lysosomes* [7], [24], [35], [36] appear as electron-dense pleomorphic single-membrane–bound vesicles containing various enzymes, such as acid phosphatase, esterases, proteases, and lipases; they are most frequently identified adjacent to the canalicular membrane. They are divided into *primary* and *secondary* lysosomes, which are actively involved in phagocytosis and eventual degradation of various cellular components. *Peroxisomes* are single-membrane intracellular organelles, also termed *microbodies* that vary in size from 0.2 to 1.3 µm. [7], [24], [37], [38] They are round to oval, exhibit a finely granular homogeneous matrix, contain various oxidases and catalases, and primarily function in the oxidation and degradation of numerous substrates with formation of hydrogen peroxides. They are also involved in oxidation of long-chain fatty acids. There are various *intracellular components* readily seen by light microscopy that contribute to the function and appearance of the liver cell. [22], [23] *Lipochrome* pigment appears as fine to coarse brown granules and is derived from increase in lysosomal activity and intracellular condensation of various cellular remnants. Two other intracellular pigments that can resemble lipochrome include bile and hemosiderin. *Bile* is present as clumped green to green-yellow globules that stain positively with the Hall stain for bilirubin. Usually, the presence of intracellular bile is also accompanied by intracanalicular bile. *Hemosiderin* pigment is coarsely granular and golden brown, is best highlighted by the Perl iron stain, and represents red blood cell degradation remnants. Intracellular *lipids* appear as clear, distinct, rounded vacuoles and are composed predominantly of neutral triglycerides. The size of the fat droplets varies, with the extremely small variants (termed *microvesicular* or "foamy") often difficult to assess on routine light microscopy unless thin 1-µm sections are prepared or fat stains (oil red O) on frozen sections are performed. Intracellular *glycogen* is distributed throughout the cytoplasm but is more frequently seen adjacent to the smooth endoplasmic reticulum. Glycogen is easily demonstrated by periodic acid-Schiff stain on frozen-section material. Nuclear glycogen, when abundant, gives the nucleus a clear appearance and may be seen in a number of liver diseases, including diabetes and Wilson disease.

Kupffer Cell The *Kupffer cells* are sinusoid lining cells that function as tissue macrophages; they represent more than 75% of fixed macrophages throughout the body and take up about one third of the hepatic sinusoidal cell volume. [7], [24], [39], [40], [41] and [42] Although originally derived from the circulation, they eventually rest along the sinusoidal borders and maintain the ability to divide. Kupffer cells have oval to elongated nuclei and abundant pyramidal stellate cytoplasm that contains numerous lysosomes; they measure up to 9 µm in length (Fig. 105-9). They overlie but do not form junctional complexes with the smaller endothelial cells. Kupffer cells may be identified, however, in gaps between adjacent endothelial cells, with cytoplasmic processes extending through endothelial fenestrations. Their primary functions relate to phagocytosis and eventual clearance of particulate material, clearance of endotoxins and degenerating cellular components, synthesis and catabolism of lipids, clearance of senescent erythrocytes, sequestration of antigens, and clearance of immune complexes.

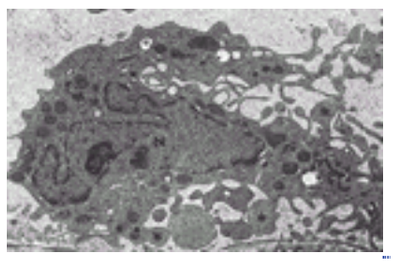

FIGURE 105-9. Kupffer Cell. By electron microscopy, the Kupffer cell exhibits a cleft nucleus (*N*), pseudopodia (*), and numerous lysosomes (*lys*). (From ref. [24].)

Endothelial Cell The *endothelial cells* are flattened, elongated sinusoidal cells ranging in size from 50 to 80 nm; they represent almost one half of the sinusoidal cell volume [24], [39], [42], [43] and [44] (Fig. 105-10). Numerous cytoplasmic projections and clustered fenestrae or gaps that range in size from 0.1 to 0.2 µm are present. These fenestrae function as a filtration barrier. The individual cells are loosely attached. A main function of the endothelial cell relates to filtering of the sinusoidal blood of various molecules, enabling substances such as glycoproteins and polysaccharides direct contact with the hepatocyte, but excluding and protecting the liver cell from numerous larger cellular components and various soluble compounds by (1) receptor-mediated endocytosis (ligand binding to coated pits on the cell surface), (2) pinocytosis (trapping of extracellular fluid droplets), and (3) phagocytosis (plasma membrane encircling and internalizing particles). [45]

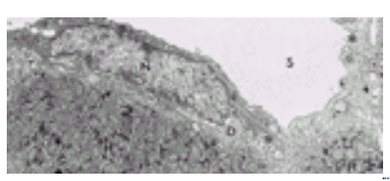

FIGURE 105-10. Endothelial Cell. This photomicrograph by electron microscopy of a sinusoid (*S*) shows an endothelial cell with an elongated nucleus (*N*) and scanty cytoplasm (*). The space of Disse (*D*) and adjacent cytoplasm of two hepatocytes (*H*) are noted. (From ref. [24].)

Stellate (Perisinusoidal) Cell Also termed *fat-storing cells* and *Ito cells* the *stellate cells* are located within the perisinusoidal liver cell recesses along the space of Disse [24], [39], [42], [46], [47] and [48] (Fig. 105-11) and constitute about one fifth of the sinusoidal cell volume. These cells range in size from 2 to 10 µm in diameter and contain small star-shaped nuclei without prominent nucleoli. The cytoplasm often contains variably sized lipid droplets, which contain a high concentration of vitamin A (retinoyl palmitate) that can easily be demonstrated as intensely green on frozen sections by rapidly fading fluorescence when excited at 328-nm wavelength. [45] Besides being the major source of vitamin A storage, the cells synthesize extracellular matrix by way of cytokine activation and resultant transformation to myofibroblasts in response to liver injury, with enhancement of protein and collagen synthesis. [49], [50]

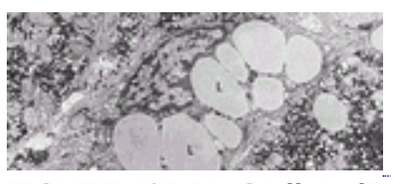

FIGURE 105-11. Stellate (Perisinusoidal) Cell. These cells, also termed *Ito* cells (*IC*), are seen by electron microscopy along the perisinusoidal recesses and characteristically demonstrate cytoplasmic lipid droplets (*L*) containing vitamin A. Adjacent hepatocytes (*H*) are noted. (From ref. [24].)

Pit Cell Also termed *liver-associated lymphocytes* and *large granular lymphocytes pit cells* are nonparenchymal T cells distributed within the sinusoidal lumen in loose contact with the endothelial or Kupffer cells; they occasionally can be seen within portal tracts. [45], [51], [52], [53] and [54] These cells function as natural lymphocyte-activated killer cells. They are often called pit cells because of the resemblance of the cytoplasmic granules to grape seeds. [1], [55] These cells are often seen in direct contact with the endothelium in response to various immunologic mechanisms and can be targeted in viral hepatitis, acute posttransplantation cellular rejection, and various primary and metastatic neoplastic processes. [7], [45]

Stroma (Extracellular Matrix) The *stroma* constitutes about 10% of the liver volume and overall supports the basic hepatic architectural arrangement, produces intercellular cohesion and communication, and effects cellular differentiation. The Glisson's capsule, composed of dense hypocellular collagen, encapsulates the liver and extends at the hilum into the hepatic parenchyma, forming the tensile structure of the portal tracts. Extension within the sinusoids into the space of Disse as reticulin fibers maintains the intralobular framework. Five basic types of collagen are seen, with types I and III representing more than 95% of the total collagen. Type I represents mature collagen strands, whereas type III represents new collagen (reticulin fibers). Type IV is present in the basal lamina (membrane) around small vascular structures and ducts and represents about 1% of the total hepatic collagen. The noncollagenous proteins are numerous matrix glycoproteins and include (1) *laminin* the major glycoprotein component within the basement membranes; (2) *fibronectin* synthesized by peri- sinusoidal cells and responsible for collagen adhesion; and (3) *elastin* which stabilizes blood vessel walls. Collagen deposition is often triggered by activation of the stellate cells in certain instances of liver cell necrosis and impaired or delayed regeneration of hepatocytes.

Biliary Network

The main function of the *biliary tract* is to transport bile synthesized in the hepatocyte into the gastrointestinal tract by way of the extrahepatic biliary network. In addition, the transport proteins synthesized by biliary epithelium and located within the microvilli aid in both (1) the secretion of bicarbonate-rich fluid and (2) the reabsorption of various fluids and solutes that overall enhance bile flow. [56] It can be divided into its structural components, [57] the smallest of which are the *biliary canaliculi*. These are located along the intercellular spaces between hepatocytes, range in size from 0.5 to 1 µm in diameter, and are lined by microvilli. The canaliculi have numerous anastomotic connections and may undergo contractions secondary to actin, myosin, and tropomyosin, [58] enabling and enhancing forward bile flow.

The canaliculi that enter the portal tracts are called the *terminal ductules periportal cholangioles* or *canals of Hering*. These duct structures are derived from hepatocytes located at the limiting plate and form communication with the interlobular bile ducts. The ducts at this point develop a basement membrane and have both liver cell and ductal ultrastructural and histochemical features. Although in the normal liver, these ducts are usually inconspicuous on routine light microscopy, their prominent proliferation (*duct transformation duct metaplasia*) is most apparent in instances of severe liver cell necrosis that extends to and involves the periportal hepatocytes.

The *interlobular bile ducts* range in size from 15 to 20 µm in diameter within the smaller portal structures and are lined by a single layer of cuboidal cells having discrete round nuclei, usually inconspicuous nucleoli, and scanty eosinophilic cytoplasm. The duct cytoplasm contains sparse mitochondria, free ribosomes, and abundant intermediate cytokeratin filaments. The lumenal surface contains numerous pinocytotic vacuoles. Adjoining duct epithelium shows complex interdigitations. The lumenal surface also contains microvilli. A basement membrane is apparent and easily demonstrated on periodic acid-Schiff stain. Although the smaller ducts have no apparent wall, the larger interlobular ducts, which measure up to 100 µm in diameter, develop a small periductal fibrous sheath. Anastomoses of the smaller interlobular ducts is frequent. The main blood supply is the smaller branches of the hepatic artery and the peribiliary plexus, which run in parallel with the duct structures. These ducts express class I major histocompatible antigens, with cytokine-mediated class II expression in instances of liver allograft rejection and certain chronic biliary tract diseases that attack ducts, such as primary biliary cirrhosis.

The larger *interlobar and septal ducts* measure more than 100 µm in diameter, have a fibrous wall, and are lined by a single layer of cuboidal to columnar epithelium with nuclei located toward the basement membrane. Some degree of periductal fibrous tissue is common but should not be confused with the distinct periductal concentric collagenosis seen in instances of long-term bile duct obstruction.

The interlobar and septal ducts lead into the *segmental ducts* that measure up to 800 µm in diameter, which eventually form the major *hilar ducts* that measure up to 1.5 mm in diameter. The hilar ducts ultimately branch into the main right and left hepatic ducts. The hilar ducts are lined by columnar mucus-secreting epithelium, have a distinct fibromuscular wall, and are associated with both intramural and extramural seromucinous peribiliary glands, which communicate with the bile duct lumen. The same type of peribiliary glands are demonstrated around the intrahepatic large bile ducts as well. [59]

Vascular Network

The major blood vessels that supply the liver are the *portal vein* and *hepatic artery*. The *portal vein* sequentially develops interlobar, segmental, interlobular veins and preterminal branches, with the terminal portal venules measuring about 20 to 30 µm in diameter and seen in the smaller triangular portal tracts. The vascular inflow from the portal veins is controlled by various sphincters. The *hepatic artery* branches accompany the portal vein and divide within the smaller portal tracts into two segments: (1) the *periportal plexus* which branches around the portal vein and drains into the sinusoids, and (2) the *peribiliary plexus* which provides blood supply to the accompanying interlobular bile ducts by way of small capillaries that are layered around the ducts. Various connections are seen between the small arterioles and the sinusoids that are most prominent in the periportal zone (zone 1 of Rappaport). The arterial flow is dependant on both sphincters and contractile mechanisms that define the degree of arterial versus portal venous blood flow, with the arterial flow varying inversely with the portal venous circulation.

The *hepatic acinus* can be divided into three segments: simple, complex, and acinar agglomerate. The *simple acinus* is the smallest functional parenchymal unit and centers around the preterminal portal venules, hepatic arteriole, and terminal bile ductules (Fig. 105-12). The acinus is divided into three zones (zones of Rappaport): (1) *periportal* (zone 1), which includes the limiting plate; (2) *midzone* (zone 2); and (3) *perivenular* (zone 3), with the terminal hepatic venule at its outer lateral margin. There is no discrete demarcation separating these zones from one another. The watershed areas occur at the periphery of the acini where blood can be derived from the smallest arteriolar branches and portal venules from adjacent acini. Biliary drainage runs parallel to the vascular sinusoidal circulation. The *complex acinus* is derived from three adjacent simple acini fed by a preterminal portal vein and arterial branches. A small aggregate or sleeve of liver cells (*acinuli*) directly surrounds the preterminal branch. The *acinar agglomerate* is composed of about four complex acini and is fed by a portal venous branch measuring 300 to 1200 µm in diameter.

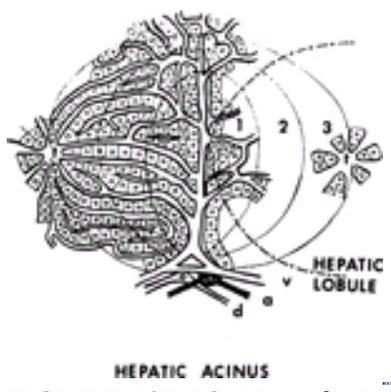

FIGURE 105-12. Hepatic Acinus. This drawing represents a *simple* acinus and its relationship with the preterminal portal venule (*v*), small hepatic arteriole (*a*), and ductule (*d*) and is divided into three zones (zones of Rappaport) with the terminal hepatic venule (*t*) located along its outer margins. (From Sternberg SS. Histology for pathologists. New York: Raven Press, 1992.)

The *space of Disse* lies between the hepatocyte and the endothelial cells, measures 0.2 to 1.0 µm in width, and forms a space that is not appreciated on routine light microscopy on biopsy material but that can sometimes be seen in autopsy livers secondary to liver cell shrinkage from autolysis. Numerous microvilli can be seen projecting from the liver cell membrane into the space of Disse. The discontinuity of the adjacent endothelial cells enables plasma to have easy permeability to the liver cell membranes. The space of Disse contains reticulin fibers, which can best be appreciated on special reticulin stains and which are easily seen on electron microscopy. The stellate or perisinusoidal cells (Ito cells) also protrude into this space. The sinusoids eventually drain into the terminal hepatic venules, which have no fibrous wall. These vessels then drain into the terminal hepatic veins and into the sublobular intercalated veins; they then exit the liver from the three main hepatic vein branches into the inferior vena cava.

Most *hepatic lymph* is derived from the space of Disse, whereas about 10% comes from capillary leakage from the peribiliary plexus. Its main function is to drain excess proteinaceous fluid from the interstitial hepatic spaces. [1, 60] The hepatic lymph drainage within the space of Disse travels into the smallest lymphatic vessels within the portal tracts by way of “endothelial massaging” by circulating erythrocytes and leukocytes within the sinusoids. The terminal branches form plexuses lined by endothelial cells and are accentuated around the hepatic arterioles in the smaller portal structures, although lymphatics are also seen adjacent to portal veins and bile duct tributaries in larger portal tracts. Small branches can also be seen along the hepatic venous outflow branches. A lymphatic plexus is also identified within the Glisson’s capsule and communicates with the intrahepatic lymphatics through anastomotic channels. Most lymphatics leave the liver at the porta hepatis, although lymphatic drainage is prominent through the Glisson’s capsule in instances when the hepatic venous drainage is impaired (e.g., acute and chronic hepatic venous outflow obstruction, cirrhosis). The larger lymphatic channels have an identifiable wall and are valved.

Neural Network

The *nerve fibers* are composed of both parasympathetic and sympathetic branches and release neurotransmitters from the intrasinusoidal fibers that contribute to modulation of liver cell function. Small nerve segments can be seen within the larger portal tracts, but smaller unmyelinated fibers can be discerned by way of electron microscopy and immunohistochemical studies within the space of Disse.

Functional Components: Liver Cell Heterogeneity

The hepatocytes within the various parenchymal zones have many different specialized physiologic functions that often parallel the light microscopy, ultrastructural features, and enzyme distribution patterns [1, 21, 61, 62, 63, 64, 65] and [66] (see Table 105-1 and Table 105-2). These functions are manifestations of (1) the nutrient and hormonal gradients delivered to the various zones, (2) the sinusoidal vascular perfusion and oxygen concentration gradients, (3) the availability of innumerable substrates and cofactors, and (4) the expression of various enzyme activities through gene expression and local (zonal) genetic variations. As a consequence, functional and morphologic zonal liver cell changes and injury are end-stage manifestations of modifications, alterations, and impairment of many of these critical and complex parameters.

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CHAPTER 106

Marc G. Ghany and T. Jake Liang

ACUTE VIRAL HEPATITIS

Five viral agents have been associated with acute hepatitis. These viruses were denominated A, B, C, D, and E in order of their discovery. They are a genetically diverse group of viruses linked by their tropism for the liver, their primary site of replication. They all cause an acute hepatitis, which can range in clinical severity from an asymptomatic illness to fulminant hepatitis. Hepatitis B, C, and D viruses have the propensity to progress to chronic hepatitis. These hepatotrophic viruses account for 90% to 95% of all cases of acute viral hepatitis in the United States. [1] Data from the Centers for Disease Control and Prevention (CDC) Sentinel Counties Study of acute viral hepatitis indicated that hepatitis A virus (HAV) was the cause for almost half the reported cases, hepatitis B virus (HBV) a third, hepatitis C virus (HCV) 15%, and the remainder to non-A, non-C viruses. [2] In the United States, acute viral hepatitis has a low case fatality rate, 1% to 2%, but is associated with substantial morbidity. Death from end-stage liver disease, a long-term consequence of acute viral hepatitis, ranks in the top 10 of all causes of mortality in the United States.

Other viruses from the families *Arenaviridae* (e.g., Lassa fever virus), *Bunyaviridae* (e.g., Rift Valley fever and Hanta viruses), *Flaviviridae* (e.g., yellow fever and dengue viruses), *Filoviridae* (e.g., Marburg and Ebola viruses), and *Herpesviridae* may cause hepatitis as part of a systemic illness but are not considered primary hepatotrophic viruses.

EPIDEMIOLOGY

Overview

The hepatotrophic viruses are found worldwide, and there are distinct geographic regions where each virus is highly endemic. The global distribution of each virus is closely linked to its mode of transmission. Enterically transmitted viruses, HAV and hepatitis E virus (HEV), are more common in regions of the world that lack proper sanitation and have limited access to fresh water. In contrast, parenterally transmitted viruses, HBV, HCV and hepatitis D virus (HDV), predominate in areas of the world where social or customary practices facilitate their transmission, explaining the high rate of HCV infection in injection drug users in the United States and HDV in the South American Indians of the Amazon basin. Despite the institution of public health measures to educate the general public on transmission risks, the number of cases of acute viral hepatitis has not declined substantially in recent years. This may be partly because risk factors for acute viral hepatitis have not been identified in 30% to 40% of cases. [1] If elimination of infection is the ultimate goal, further studies on the various modes of transmission are necessary.

Hepatitis A Virus

Prevalence HAV infection occurs worldwide. The prevalence of antibody to hepatitis A (anti-HAV) correlates inversely with the socioeconomic status and the prevailing standard of hygiene of a region. In highly endemic areas, the prevalence of anti-HAV is almost universal by 5 years of age, whereas in industrialized nations, it is generally less than 5% for all age groups sampled. This finding has important public health implications. Outbreaks of HAV infection are more common in low-prevalence areas because of a susceptible population and rare in endemic areas because of natural herd immunity. This explains the cyclic epidemics observed every 10 to 15 years in areas of intermediate prevalence, such as the United States. Additionally, a paradoxical increase in the number of cases of clinical hepatitis is observed in low- and intermediate-prevalence areas owing to a greater number of adult infections. About one third of the U.S. population has serologic evidence of past HAV infection. Prevalence was found to increase with age according to data obtained from the Third National Health and Nutrition Examination Survey (NHANES-III, 1988–1994). [3] This finding can be explained by infections that occurred during childhood when sanitary conditions were poorer than present-day standards and travel to endemic regions by adults. The age-adjusted prevalence of anti-HAV was highest in Mexican Americans (70%) and lowest in non-Hispanic whites (23%). This difference is probably related to socioeconomic factors rather than racial susceptibility to infection. The incidence of acute HAV infection in the United States has remained fairly constant over the past several decades.

Transmission HAV is transmitted primarily by the fecal-oral route through person-to-person contact or ingestion of contaminated food or water. Viral titers are highest in stool and can reach 10^8 virions per milliliter. [4] This is because HAV lacks a lipid envelope and is readily secreted by hepatocytes into bile and the intestinal tract. [5] The virus is detected in stool during the incubation period, and the viral titer peaks 2 weeks before the onset of symptoms. [6] Therefore, the highest period of infectivity is before the onset of symptoms. In most cases, viremia becomes undetectable after the peak of the alanine aminotransferase (ALT) level. [7] However, experimental

transmission studies performed in human volunteers and chimpanzees revealed that viremia may persist for long periods during the convalescent phase of the illness. [6], [7] and [8] This suggests that blood obtained during the convalescent phase may be potentially infectious. Furthermore, the virus could be demonstrated by reverse transcriptase polymerase chain reaction (RT-PCR) in convalescent stool and may be a source of infection in sporadic cases. Viral titers in serum are about twofold less than those of stool; therefore, parenteral spread is not a significant route of transmission. Transmission has been reported occasionally in blood transfusion recipients, patients who require clotting factors, and injection drug users who share needles. [9], [10] and [11] Transmission has not been associated with saliva, semen, or urine. [4] Sexual transmission of hepatitis is reported in both heterosexuals and homosexuals. Outbreaks in homosexual men seem to be related oral-anal contact during the sexual encounter. [12], [13] Nosocomial spread from undiagnosed cases has been reported. [14] In the United States, the highest rates of spread are reported in household contacts, in intimate contacts of an index case, and in child day-care centers (person-to-person spread). [15] Data from the Viral Hepatitis Surveillance Program at the CDC in 1995 showed that contact with another person with hepatitis A was overall the most frequently identified risk factor. Attendees or employees of day-care centers accounted for 10% to 15% of cases. However, nearly 50% of cases in both 1994 and 1995 reported no known risk factor. [15] It has been suggested that transmission from an asymptomatic infant or child may account for most of the adult cases of HAV infection. [16] Outbreaks of HAV have been associated with ingestion of shellfish, [17], [18] raw onions, [19] salads, [20] and frozen strawberries, [21] but these account for a small number of cases overall.

Hepatitis B Virus

Prevalence Hepatitis B occurs worldwide; the areas of highest prevalence are China, Southeast Asia, sub-Saharan Africa, and Alaska. Almost 50% of the world's population live in these regions, where the prevalence of hepatitis B surface antigen (HBsAg), a serologic marker of chronic infection, can exceed 8%. [22] Another 40% of the world's population live in areas with intermediate prevalence, 3% to 5%, such as Japan and India. In low-prevalence areas such as the United States, Western Europe, and Australia, the rate of HBsAg positivity is 0.1% to 2%. In endemic areas, most infections occur in children, whereas in areas of low seroprevalence, most infections occur in adults. The prevalence of HBsAg was found to increase with age in the U.S. population and is highest in people aged 20 to 29 years, probably reflecting transmission through injection drug use or sexual exposure. [23] Certain U.S. populations, such as Eskimos, have higher rates of HBsAg positivity. [24] The incidence of acute HBV infection has declined dramatically in the United States, primarily owing to the use of the hepatitis B vaccine in high- risk groups, behavioral changes as a result of the human immuno- deficiency virus (HIV) epidemic, and federal regulations introduced to limit nosocomial spread.
Transmission Hepatitis B is transmitted primarily by the percutaneous route. Not surprisingly, the highest titers of virus are seen in serum and blood; intermediate titers are seen in semen, vaginal fluid, and saliva; and lowest titers are observed in tears, urine, feces, and breast milk. The infectivity of other body fluids is 100- to 1000-fold less compared with blood. Transfusion of blood products used to be the major route of transmission before the introduction of blood screening in 1985. Studies of multitransfused hemophiliac patients before 1985 indicated that 40% to 50% had markers of previous or active HBV infection. [25] Transfusion-related HBV infection has been virtually eliminated by screening of blood donors. The transmission of hepatitis B varies in endemic and nonendemic regions. In the United States and Western Europe, areas of low endemicity, injection drug use and sexual exposure to an acute case or chronic carrier are the major modes of transmission. Data from the CDC Viral Hepatitis Surveillance Program showed that sexual exposure was the predominant risk factor in the United States; 26% of cases reported sexual contact with a person known to have HBV. Injection drug use was the next most common (13%). The risk for sexual transmission was directly related to number of lifetime sexual partners, paid sex, and prior history of sexually transmitted diseases. The risk for transmission appears to be greater from men to women. In contrast, in endemic regions, perinatal (vertical transmission) and person-to-person spread (horizontal transmission) are more common modes of spread. Perinatal spread is responsible for the high rate of infection seen in China and Southeast Asia. [26] Perinatal spread is more efficient if the mother is HBeAg positive. Fetal exposure to blood during passage through the birth canal is thought to be the cause of infection rather than transplacental spread. In sub-Saharan Africa, another highly endemic area, the major route of spread is horizontal transmission, occurring in young children between the ages of 4 and 6 years, presumably through close physical contact or intrafamilial spread. The reason for the lower rate of vertical transmission in the African population may be due to the low prevalence of hepatitis B e antigen (HBeAg) in the serum of females of childbearing potential. [27] Health care workers were at major risk for exposure in the 1980s; however, widespread use of the hepatitis B vaccine has reduced the incidence of acute hepatitis markedly, from 9% in 1985 to 0.8% in 1994–95. [28] Nosocomial infection continues to remain a source of infection, and transmission has been reported from physician to patient, from contaminated multiuse vials and medical instruments, in hemodialysis units, and through organ transplantation, especially in the case of antibody to hepatitis B core antigen (anti-HBc)–positive donors after orthotopic liver transplantation (OLT).

Hepatitis C Virus

Prevalence HCV infection occurs worldwide, and it is estimated that 3% of the world's population is infected based on blood donor studies. [29], [30] The geographic prevalence of antibody to HCV (anti-HCV) is fairly consistent throughout the world, ranging from 0.5% to 2%. However, the prevalence of HCV is disproportionately high in Egypt and Japan. In Egypt, this has been explained by the use of mass parenteral therapy for schistosomiasis with reusable glass syringes during the period between 1961 and 1985 and in Japan through the use of blood transfusions during the Second World War. [31], [32] and [33]. Data from the NHANES-III study found a prevalence of anti-HCV of 1.8% in the general U.S. population. [34] Prevalence was higher in African Americans and Hispanics. Groups with higher prevalence than the general population included hemophiliacs, injection drug users, patients receiving hemodialysis, and people attending sexually transmitted disease clinics. Currently, injection drug users are at highest risk for acute HCV infection. Since 1989, the incidence of acute HCV infection has been on the decline, primarily owing to screening of the blood supply and a decrease in injection drug use.
Transmission HCV is transmitted by percutaneous, sexual, and perinatal routes. The highest viral titers are seen in blood and serum. Before the introduction of blood-screening procedures (anti-HBc and ALT levels were used as surrogate markers for non-A, non-B hepatitis beginning in 1985 until they were replaced by anti-HCV testing in 1991), transfusion of blood or blood products was a major source of infection, accounting for up to 20% of cases of infection. HCV is efficiently transmitted by blood. Sixty to 95% of hemophiliacs who received clotting factor before 1985 tested positive for anti-HCV. [35], [36] With the introduction of screening measures, the risk for acquiring hepatitis C through blood transfusion in the United States is now estimated to be 1:400,000. [37] Transmission of HCV through the blood supply still occurs in countries where testing for HCV is not routinely performed. Injection drug use is a highly efficient means of transmission and now the major route of transmission of HCV in the United States. The prevalence of anti-HCV in active injection drug users who share needles is 70% after 1 year and greater than 90% after 8 years. Prevalence of anti-HCV in this group has been shown to be associated with duration of injection drug use and sharing needles with a long-time drug user. Intranasal cocaine use has been suggested as a mode of transmission. [38] A cross-sectional study of volunteer blood donors in the United States reported that a history of intranasal cocaine use was independently associated with anti-HCV positivity. [38] HCV can be transmitted sexually. Epidemiologic case-control and cross-sectional studies report independent associations between HCV infection and exposure to an infected sex partner, increasing number of sex partners, failure to use a condom, and sexual activities involving trauma. [34], [39] The presence of other sexually transmitted diseases (e.g., herpes and syphilis) has been shown to facilitate the transmission of HCV. The rate of sexual transmission from an infected case to a partner is low in monogamous relationships, 1.5%, but increases with duration of the partnership and averages 3% to 5%. [40], [41] The rate of sexual transmission is the same for heterosexual and homosexual relationships. Overall, sexual transmission accounts for a relatively small percentage of acute cases, less than 10%. Perinatal transmission occurs but is low and is significantly lower compared with HBV. The rate of transmission is estimated to be 3% to 5% for mothers not infected with HIV-1 and increases to 12% to 14% in HIV-1–positive women. [42], [43] Perinatal transmission of HCV was shown to correlate with viral level; thus, the higher viral loads in HIV-positive women may account for their higher rate of perinatal transmission. [44], [45] Transmission of HCV through breast-feeding has not been reported. [46] It was suggested that cesarean section may be associated with a lower rate of perinatal transmission. [47] Mother-to-child transmission occurs but is uncommon. [41], [48] Nosocomial transmission of HCV occurs but is not a major cause of new infections. [49], [50] Transmission through contaminated multidose vials, through endoscopes, from physician to patient, and through surgical procedures were identified as potential sources of infection. [51], [52], [53] and [54] Needle-stick injury is a risk factor for health care workers (see later).

Hepatitis D Virus

Prevalence HDV, the infectious agent of delta hepatitis, is found throughout the world. Its prevalence mimics that of HBsAg owing to its dependency on HBV for its life cycle. [55] However, there are areas where there is discordance between the prevalence of HDV and HBV, such as in China, where the rate of HBV infection is high but HDV low. The reason for this is unknown. HDV is endemic in the southern Mediterranean, Amazon basin, and tropical and subtropical Africa. It is estimated that about 5% of HBsAg carriers are infected with HDV. Prevalence of HDV is low in the United States. [56] Recent studies indicate that the seroprevalence of HDV in southern Europe is declining. Studies in Italy confirmed a decrease in incidence of acute cases from 3.1 per 100,000 population in 1987 to 1.2 per 100,000 population in 1992. [57], [58] This decline was attributed to the introduction of hepatitis B vaccination programs and a decrease in injection drug use. [57]
Transmission Hepatitis D is transmitted in a similar fashion to hepatitis B. The single most important factor influencing transmission of HDV is the HBsAg status of an individual. HDV cannot be transmitted in the absence of HBsAg. Exposure to blood and blood products used to be the major route of transmission, but this has been virtually eliminated by screening of blood for HBV. [55] In developed countries, injection drug use and sexual transmission are the major routes of spread. The mode of spread in populations from Africa and the Amazon Basin is not well understood, but tribal practices that expose individuals to blood or sexual exposure may play a role. Perinatal transmission is rare.

Hepatitis E Virus

Prevalence HEV is the major causative agent of enteric non-A, non-B hepatitis and the primary cause of sporadic hepatitis in the developing world. [59], [60] HEV is found worldwide, and the highest prevalence is found in areas of the world where there is inadequate sanitation. It is endemic in Southeast and Central Asia. Epidemics of HEV infection, usually involving many thousands of people, occur in endemic areas. In endemic areas, the prevalence of antibody appears to increase with age; about 5% of children younger than 10 years old have antibody to HEV, but 10% to 40% of adults are seropositive. [61], [62] This suggests that HEV infection does not occur frequently in children in endemic areas. This discrepancy may be in part due to the sensitivity of the test used for serologic testing. The prevalence of anti-HEV in the United States is low.

Transmission Hepatitis E is transmitted by the fecal-oral route. Highest attack rates are seen in individuals aged 15 to 40 years. [62] In endemic areas, consumption of contaminated water accounts for most new cases of HEV. Person-to-person spread is rare, and secondary attack rates appear to be distinctly uncommon, occurring in 0.7% to 2.2% of household contacts of hepatitis E, compared with rates of 50% to 75% in susceptible household contacts of hepatitis A. [62] There is no evidence for parenteral or sexual transmission. Vertical transmission is known to occur but accounts for only a small number of cases. [63] HEV is responsible for sporadic outbreaks of hepatitis. However, the reservoir of HEV during sporadic outbreaks is unclear. Recent data based on the detection of HEV in the feces of domestic swine in Nepal and the United States and anti-HEV in the sera of pigs, cattle, sheep, and rodents suggests that HEV may be a zoonosis. [64], [65] and [66] Cases of HEV infection in the United States are rare and usually associated with a history of travel to an endemic area. Infection has been reported without a history of travel, lending support to the zoonosis theory.

VIROLOGY

The five known hepatitis viruses, A to E, belong to completely different virus families. Although all of them can cause acute hepatitis, only HBV, HCV, and HDV infection can lead to chronic infection. The virologic characteristics of each virus are summarized in Table 106-1 and described in detail in this section.

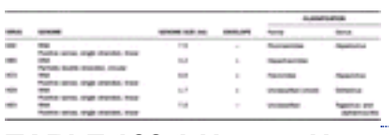

TABLE 106-1 Human Hepatitis Viruses

Hepatitis A Virus

HAV is classified in the *Hepatovirus* genus in the *Picornaviridae* family. It has one serotype but multiple genotypes. The virus is nonenveloped and stable to acid and heat treatment. The HAV has a linear, positive-sense, single-stranded RNA genome of about 7.5 kilobases (kb). The genome has a protein covalently attached to its 5' end and a poly A tail at the 3' end. [67] The genome is organized similarly to other picornaviruses in that it codes for a single polyprotein of about 2235 amino acids [68] (Fig. 106-1). The polyprotein is divided into three main functional domains from the 5' end, P1, P2, and P3. Individual proteins are cleaved from the functional domains, with P1 encoding the viral capsid proteins, whereas P2 and P3 encode the nonstructural proteins. The 5' untranslated region (UTR) is the most conserved segment of the genome [69] and contains an internal ribosomal entry site that functions to initiate protein synthesis from two in-frame AUG codons. [70] Mutations in this region appear to enhance the adaptation of virus growth in cell culture but do not appear to affect in vivo virulence. [71]

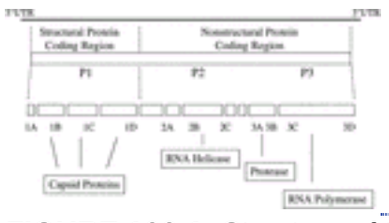

FIGURE 106-1. Structure of the hepatitis A virus and genetic organization. The *top line* represents the hepatitis A virus RNA genome. Translation of the genome yields the polyprotein; the *lower diagram* illustrates the three regions of the polyprotein and the individual protein products.

Cleavage within the P1, P2, and P3 domains of the polyprotein gives rise to the individual viral proteins. Four capsid proteins, designated 1A to 1D, are encoded in P1, with the P2 and P3 regions encoding proteins 2A to 2C and 3A to 3D, respectively. The 3B protein covalently attaches to the 5' terminus of the genome and serves as the primer for replication. [67] The functions of 2A, 2B, and 3A are unclear. The 3C is probably a serine proteinase [72] involved in cleavage of the polyprotein. 3D is the putative viral RNA polymerase. [73]

The replication process of HAV has been inferred from studies of other picornaviruses. [74] Entry of the virus into the host is mediated by a cell surface receptor, which has been recently proposed to be a mucin-like class 1 integral membrane glycoprotein. [75] Viral entry is followed by uncoating and initiation of viral protein synthesis. [76] Viral RNA synthesis proceeds from negative to plus strand and occurs in the cytoplasm. Viral assembly follows a sequence similar to that of picornaviruses in a cellular membrane compartment. [77]

Hepatitis B Virus

HBV has been classified into seven genotypes, A to G, based on sequence divergence. [78], [79] Three types of viral particles are visualized in infectious sera under electron microscopy. Two of the viral particles are smaller spherical structures with a diameter of 20 nm and filaments of variable length with a width of 20 nm, both of which vastly outnumber the Dane particles in infectious serum. [80] The spheres and filaments are composed of HBsAg and host-derived lipids and are noninfectious owing to the absence of nucleic acid. [81] The infectious HBV virion (Dane particle) has a 42-nm spherical, double-shelled structure, consisting of a lipid envelope containing HBsAg that surrounds an inner nucleocapsid. The HBcAg complexes with viral-encoded polymerase and viral DNA genome to form the nucleocapsid. [82] The genome of HBV is a partially double-stranded circular DNA of about 3.2 kilobase pairs. The viral polymerase is covalently attached to the 5' end of the minus strand. [83]

The viral genome encodes four overlapping open reading frames (ORFs) (Fig. 106-2). The S ORF encodes the viral surface envelope proteins, the HBsAg, and comprises the pre-S1, pre-S2, and S regions. The core gene consists of precore and core regions. Multiple in-frame translation initiation codons are a feature of the *S* and *C* genes, which give rise to functionally distinct proteins. HBeAg and the viral nucleocapsid HBcAg are translated from initiation at the precore and core regions, respectively (Fig. 106-3). The core protein has the intrinsic property to self-assemble into a capsid-like structure and contains a highly basic cluster of amino acids at its C terminus with RNA-binding activity. [84] The precore ORF codes for a signal peptide that directs the translation product to the endoplasmic reticulum, where the protein is further processed to form the secreted HBeAg. The role that HBeAg serves is still unknown because mutations inactivating the precore region do not appear to affect viral replication. [85] One speculative role for HBeAg is an immune tolerogen, whose function is to promote persistent infection. [86] The polymerase (pol) is a large protein (about 800 amino acids) and is functionally divided into three domains: the terminal protein domain (TP), which is involved in encapsidation and initiation of minus-strand synthesis; the reverse transcriptase domain, which catalyzes genome synthesis; and the RNase H domain, which degrades pregenomic RNA and facilitates replication. The HBX is a 16.5-kd protein and has been attributed with multiple functions, including signal transduction, direct transcriptional activation, DNA repair, and inhibition of protein degradation. [87], [88] and [89] The mechanism of this activity and the biologic function of HBX in the viral life cycle remain largely unknown.

However, it is well established that HBX plays an essential role in HBV infection in vivo [90], [91] and may contribute to the oncogenic potential of HBV. [92]

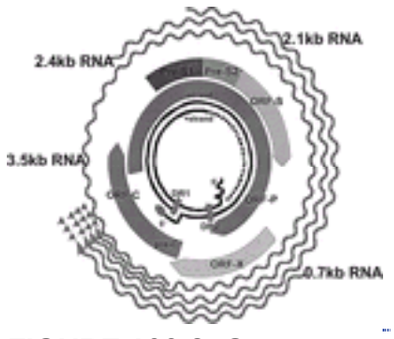

FIGURE 106-2. Genome structure and organization of hepatitis B virus. The hepatitis B virus open reading frames: pre-C and C (precore and core proteins), P (polymerase protein), pre-S1, pre-S2, and S (L, M, and S surface envelope proteins), and X protein are shown. The viral genome structure is composed of the full length (-)-DNA strand and variable length (+)-DNA strand (*solid* followed by *dashed line*). The pol protein is covalently attached to the 5' of the (-) strand and a capped oligoribonucleotide (*angulated line*) to the (+) strand. Direct repeats 1 and 2 (*small rectangular boxes*) are shown on the genome. The *outer lines* represent the four transcripts all terminating at a common polyadenylation site.

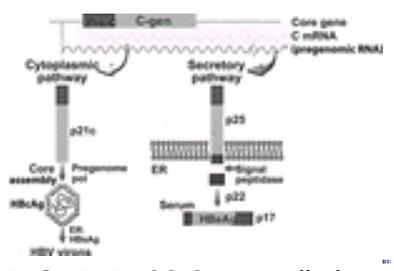

FIGURE 106-3. Two distinct processing pathways of *HBV* core gene. Two species of core RNAs are transcribed from the core gene. One starts before the pre-C and the other in the middle of the pre-C region. The former codes for the pre-C/C protein and is directed to the secretory pathway through the signal peptide sequence in the pre-C. After further proteolytic cleavage in the endoplasmic reticulum, HBeAg is formed and secreted. The latter RNA codes for the core protein, which is involved in the nucleocapsid assembly.

Other functionally important elements within the HBV genome include two direct repeats (DR1 and DR2) in the 5' ends of the minus and plus strands, which are required for strand-specific synthesis during replication. [93] Two enhancer elements, designated as En I and En II, confer liver-specific expression of viral gene products. [94] A glucocorticoid-responsive sequence within the S domain, [95] a polyadenylation signal within the core gene, [96] and a posttranscriptional regulatory element overlapping En I and part of HBX ORF have been described. [97]

The HBV replication pathway has been studied in great detail and is summarized in Fig. 106-4. The initial phase of HBV infection involves the attachment of mature virions onto host cell membranes, likely involving the pre-S domain of the surface protein. [98] Various cellular factors have been proposed to be the viral receptors, but only the carboxypeptidase D has been shown to play an essential role in viral entry for the duck HBV. [99] Mechanisms of viral disassembly and intracellular transport of viral genome into the nucleus are not well understood and probably involve modification of nucleocapsid core protein. [100] After entry of viral genome into the nucleus, the single-stranded gap region in the viral genome is repaired by the viral pol protein, and the viral DNA is circularized to the covalently closed circular form. [100] This form of HBV DNA serves as the template for transcription of several genomic and subgenomic RNAs. Structurally, the transcripts are unspliced, polyadenylated, and possess a 5' cap structure. The 3.5-kb genomic transcripts consist of two species with different 5' ends: the pregenomic and the precore RNAs. The pregenomic RNA (pgRNA) serves as the template for reverse transcription and the mRNA for core and polymerase; the precore RNA directs the translation of the precore gene product. [101] The polymerase is initiated at the pol start codon of the pgRNA, probably as a result of a ribosomal scanning mechanism. [102] The large HBsAg (LHBsAg) protein is translated from the 2.4-kb subgenomic RNA, the middle (M) and small HBsAg (SHBsAg) proteins from the 2.1-kb RNA, and the HBX protein from the 0.7-kb RNA.

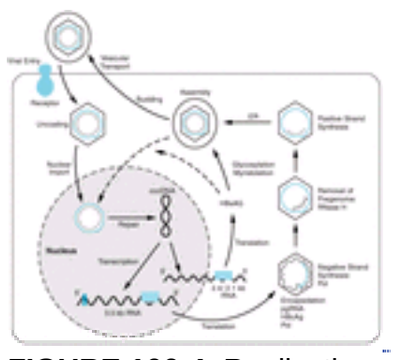

FIGURE 106-4. Replication of hepatitis B virus. Infectious virions probably attach to hepatocytes through the pre-S1 domain of the L protein. Upon entering, the nucleocapsid is delivered to the nucleus, and the viral genome is repaired to the covalently closed circular form (cccDNA). Viral transcripts are translated in the cytoplasm, and the core and polymerase proteins interact with the genomic length RNA to form the nucleocapsids. Reverse transcription occurs, and the mature virions are assembled in the endoplasmic reticulum, where they acquire the surface proteins. The virion is then secreted through vesicular transport. The encapsidation signal (dark box) and posttranscriptional regulatory element (light box) on the *HBV* transcripts are shown as *rectangular boxes*.

The S protein accounts for most *S* gene products, with the L protein constituting 1% to 2% and the M protein 5% to 15%. Each surface protein has a glycosylation site in the S domain. Additional modifications of the M and L proteins occur at the pre-S2 domain with an N-linked oligosaccharide and a myristic acid at the amino-terminal glycine residue of the pre-S1 domain. [103] The distribution of the three envelope glycoproteins varies among the types of viral particles, with little M and L protein in the 20-nm particles but relatively more L protein in the Dane particles.

Assembly of HBV begins with encapsidation of the genome. The packaging signal is a *cis*-acting element referred to as epsilon, which contains a stem-loop structure. [104] The terminal protein of the pol interacts with the epsilon and in concert with the core protein forms the nucleocapsid. [105] After encapsidation, the pol mediates the reverse transcription of the pgRNA to minus-strand DNA and subsequent positive-strand synthesis. The circular form of the DNA is completed through several complicated steps of strand transfer. [106] The nucleocapsid then interacts with the envelope proteins in the endoplasmic reticulum to assemble into mature virions.

Hepatitis C Virus

HCV is an enveloped virus and has been classified into six genotypes, 1 to 6. The virus circulates predominantly in complex with immunoglobulins and lipoproteins. [107], [108] It is a member of the *Flaviviridae* family and has a positive-sense, single-stranded RNA genome about 9.6 kb in length with a single large ORF and highly conserved UTRs at the 5' and 3' ends [109], [110] and [111] (Fig. 106-5). The 5' UTR contains an internal ribosomal entry site that mediates translation. [110] The 3' UTR has a

variable length with a 5' to 3' sequence followed by a polypyrimidine tract and finally by a highly conserved 98 nucleotide sequence. [111] The 3' UTR is also predicted to have extensive secondary structures that probably play a role in viral replication. In the large ORF, the structural genes for core and two envelope glycoproteins, E1 and E2, are located at the N-terminal end and the nonstructural genes NS2, NS3, NS4A, NS4B, NS5A, and NS5B at the C-terminal end.

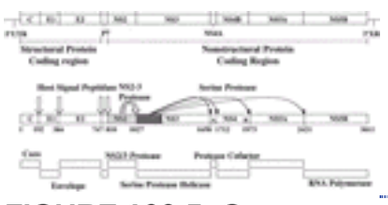

FIGURE 106-5. Genome organization of hepatitis C virus. The 5' and 3' untranslated regions (UTR) flanking a polyprotein open reading frame are shown at the *top*. Numbering refers to nucleotide positions of genes, based on the sequence of an *HCV* genotype 1a infectious clone. The middle panel shows HCV polyprotein processing with cleavage sites of host signal peptidase (*open arrows*), NS2-3 protease (*gray arrow*), and NS3 serine protease (*thin arrows*). Numbering denotes amino acid position upstream of cleavage sites. The processed HCV proteins are shown at the bottom.

The HCV polyprotein of about 3000 amino acids is processed co- and posttranslationally by cellular and viral proteases to produce the individual gene products. [112], [113] Cellular proteases in the endoplasmic reticulum catalyze the cleavage of the structural proteins, whereas viral encoded proteases cleave the nonstructural proteins. [114]

The highly conserved core protein is the putative viral nucleocapsid and encompasses the first 191 amino acids of the polyprotein. The E1 and E2 are envelope glycoproteins with C-terminal hydrophobic transmembrane domains. [115] The 27 N-terminal amino acids of E2 have been designated the hypervariable region 1, owing to significant amino acid variations among all isolates. [116] Studies have suggested that sequence variations of the hypervariable region 1 result from immune pressure as a mechanism to evade host immune response.

The NS2 region encodes a metalloproteinase that requires cell membrane and N-terminal NS3 for efficient function. [117] The NS2-3 protease mediates autocatalytic cleavage between the NS2 and NS3 (Fig. 106-5). The NS3 region encodes a multifunctional protein with an N-terminal serine protease and a C-terminal RNA helicase and nucleotide triphosphatase (NTPase). [118] The NS3 protease, distinct from NS2-3 protease activity, is involved in processing the downstream polyprotein. [119] The NS4A interacts with and acts as a cofactor for the NS3 protease. [120] The crystal structure of NS3 protease has a trypsin-like structure but becomes more chymotrypsin-like when co-crystalized with NS4A. [121] The function of NS4B is unknown. The NS5A may play a role in sensitivity to interferon. [122] The NS5B is the RNA-dependent RNA polymerase that mediates viral replication. Similar to replication of other *Flaviviridae* family members, HCV replicates in the cytoplasm, presumably in a membrane-associated compartment (Fig. 106-6). The NS5B recognizes a structure in the 3' UTR and initiates minus-strand RNA synthesis in a primer-independent template-specific process. [123] The synthesis of plus strand is presumably carried out by NS5B in a similar manner. HCV replication in vivo probably occurs in the context of multiple viral nonstructural proteins. The process of viral assembly is poorly understood and probably requires the interactions among viral genome, core, and E1 and E2 proteins.

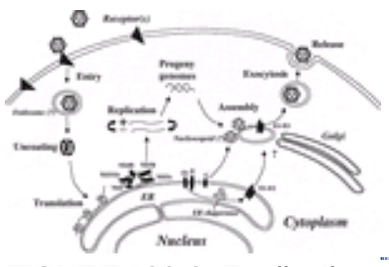

FIGURE 106-6. Replication of hepatitis C virus. The virion attaches and enters the susceptible cell through pathways that are not yet completely defined. The viral genome is then directed to a membranous component in the perinuclear endoplasmic reticulum region and serves as template for HCV protein synthesis. The nonstructural proteins form a replication complex with the genomic RNA and direct RNA replication (to negative and then plus strands). The structural proteins, which are retained in the endoplasmic reticulum, interact with the progeny genomes and assemble into virions. The virions are then secreted through an unknown exocytotic pathway, probably not passing through the Golgi compartment.

Hepatitis D Virus

HDV requires co-infection with HBV for replication. [124] Delta antigen is the inner ribonucleoprotein (RNP) component of a subviral particle that is enveloped by HBsAg. HDV has many features in common with viroids, the subviral agents of plants. [125] The RNP complex consists of small (SHDAg) and large (LHDAg) delta antigens and a single-stranded circular RNA genome 1.7 kb in length that has extensive self-complementation to form a rodlike structure (Fig. 106-7). HDV attachment to the host cell is mediated by HBsAg. [126] The HDV genome uses host RNA polymerase II to carry out RNA-directed RNA synthesis that is dependent on the small delta antigen. [127], [128] Both genomic and antigenomic RNAs possess ribozyme activities that catalyze RNA self-cleavage and self-ligation. Similar to plant viroids, transcription and replication are integrated into a single process using a double rolling-circle mechanism. [125] After entry into cells, HDV genome serves as a template for replication, resulting in the production of multimeric antigenomes. Nascent antigenomes, through their intrinsic ribozyme activities, form circular monomeric RNAs that, in turn, serve as templates for the production of HDV genomes. Alternatively, the elongating product can be cleaved and released as polyadenylated mRNAs, which then direct delta antigen synthesis. [129]

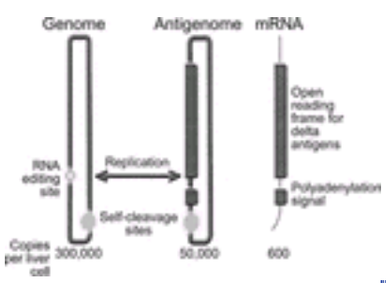

FIGURE 106-7. Genomic organization of hepatitis D virus. The RNA genome has a rodlike structure and contains an RNA editing and a self-cleavage site (*circle*). The antigenome is synthesized from the genomic RNA and is the template for HDV mRNA encoding the delta antigens. The antigenome also serves as the template for genome synthesis. The estimated copy numbers of the RNA species in the infected liver are shown *below*.

Translation of HDV mRNA yields the small and large delta antigens, which differ by the addition of 19 amino acids in the C-terminal end of LHDAg. [130] The small and large delta antigens have distinct functions during HDV replication. The additional C-terminal amino acids of the LHDAg contain an isoprenylation signal that appears to be important for interaction with HBsAg during viral assembly. [131] The RNA editing process results in two mRNA species coding for the two forms of HDAg and

involves the double-stranded RNA-activated adenosine deaminase. [132] This RNA editing activity results in the C-terminal extension of the LHDAg. [133]

HDV assembly begins with the association of the delta antigens with the newly synthesized genome to yield an RNP complex. [134] The RNP complex is transported from nucleus to cytoplasm, presumably mediated by the nucleocytoplasmic shuttling function of delta antigens. The LHDAg of the RNP interacts with HBsAg to facilitate assembly. Large delta antigen is required for particle assembly, whereas small delta antigen is co-packaged but not required for particle formation. [135]

Hepatitis E Virus

HEV is a nonenveloped virus and is currently unclassified but is similar to togavirus and alphavirus. The viral genome is a single-stranded, positive-sense RNA genome of about 7.5 kb. It is polyadenylated and has a capped 5' end. [136] The genome is organized into three overlapping ORFs flanked by noncoding regions (Fig. 106-8). ORF 1 appears to encode the nonstructural gene products, ORF 2 the capsid protein, and ORF 3 a protein with possible nucleocapsid function. [137] Based on sequence analysis and comparison with other viral genomes, ORF 1 contains several functional domains: methyl transferase, papain-like cysteine protease, helicase, and an RNA-dependent RNA polymerase. [138] ORF 2 begins 38 nucleotides downstream from the 3' terminus of ORF 1 and contains arginine-rich regions believed to be involved in genome encapsidation. ORF 3 overlaps one 3' nucleotide of ORF 1 and extends into ORF 2.

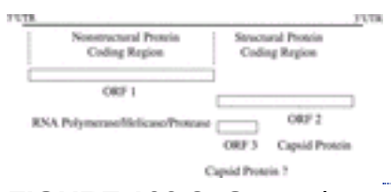

FIGURE 106-8. Genomic organization of hepatitis E virus. The open reading frames (ORF) are flanked by 5' and 3' noncoding regions and a 3' polyadenylation. Within ORF 1: MT (methyl transferase), X and Y (unknown functions), Pro (protease), Hel (helicase), H (proline-rich hinge region), Pol (RNA-dependent RNA polymerase). ORF 2 codes for the capsid, and ORF 3 codes for a protein of unknown function.

Replication of HEV has not been characterized because of a lack of similarity to other viruses and efficient cell culture systems. The mechanisms of viral attachment, entry, and uncoating are unknown. The released genome likely uses the host translation machinery to synthesize viral gene products. Processing of the ORF 1 polyprotein is carried out by either cellular proteases or a viral papain-like proteinase encoded by ORF 1. The viral encoded polymerase mediates synthesis of plus- and minus-strand genomes as well as subgenomic RNA species, which serve as templates for translation of viral antigens. The mechanism of viral assembly is unknown but probably involves the capsid function of ORF 2 and ORF 3.

PATHOGENESIS

Acute infection with hepatitis viruses is associated with a spectrum of liver diseases and systemic manifestations, ranging from asymptomatic infection, to self-limited hepatitis, to fulminant hepatitis. In addition, three of the viruses (B, C, and D) are associated with persistent infection and chronic hepatitis whose clinical manifestations are variable. The outcome, extent, and severity of acute viral hepatitis depend on a variety of viral and host factors. Although host responses play a major role in viral clearance and disease pathogenesis, the outcome of infection is often predicated on a variety of viral adaptive mechanisms. The pathogenesis of acute viral hepatitis involves a complicated set of virus-host interactions and is likely a result of diverse host immunologic responses to the viral infection. In this section, the pathogenesis of acute viral hepatitis is discussed in the general context of viral infection and host responses and with respect to relevant issues specific to each of the viruses. Among the hepatitis viruses, the pathogenetic mechanisms have been much better studied in HBV and HCV than in the others; therefore, we focus more on the former viruses. Furthermore, the pathogenesis of acute hepatitis is often inferred from the various studies during chronic infection (for B, C, and D), and we attempt to highlight these issues during the course of discussion in this section.

Primary Infection

After gaining entry into a susceptible host, the hepatitis viruses are carried by the blood to the liver, which is the major site of viral tropism. For HAV and HEV, the viruses enter through the gastrointestinal epithelium, and the uptake mechanisms remain largely unknown. It is possible that a round of local replication may occur at the site of entry before the virus is further spread to the liver through the portal circulation. Because HBV, HDV, and HCV have been reported to infect nonhepatocytes, the existence of an intermediate compartment of replication before infection of the liver has also been proposed but remains controversial. [139], [140] Many initial events of the viral infection have been characterized in the animal models for each virus. HBV and probably HCV replicate in a noncytopathic manner without causing injury to hepatocytes, whereas the cytopathicity of other viruses is unknown.

Immune Responses

During primary infection, the initial pathway of antiviral immune response is largely unknown. Initial viral infection is associated with activation of innate immunity in the liver. Recognition of infected hepatocytes by resident natural killer or natural killer T cells leads to activation of these cells and induction of antiviral cytokines, including interferons. [141], [142] This phase of innate immunity leads to the initial control of viral replication. Because this antiviral response is likely associated with a noncytopathic mechanism, little or no hepatocellular injury is evident. The innate immunity also plays a critical role in the activation of the adaptive immunity, including humoral and cellular responses. Induction of humoral immune response with production of neutralizing antibodies prevents viral spread and leads to subsequent elimination of circulating viruses. [143] This mechanism is particularly relevant for HAV and HBV infections because of the high level of viral replication and viremia. Neutralizing antibodies against the capsid (for HAV and HEV) or envelope (for HBV) can be induced and lead to viral clearance and protection from subsequent exposure. [144], [145] and [146] This constitutes the basis for the successful formulation of HAV (probably HEV as well) and HBV vaccines based on the capsid and envelope proteins, respectively. In HAV, HBV, HDV, and HEV, immunoglobulin M (IgM) class of antibodies is detected first and serves as a useful marker for acute infection. IgA class of antibodies has been detected in HAV and HEV infections, but its implication in viral clearance and protective immunity remains unknown. [144] On the other hand, neutralizing antibodies probably play little or no role in viral clearance during acute HCV infection. [147] A clearly defined anti-HCV IgM response has not been identified, and the antibody response, [148] especially against the envelope proteins, tends to occur much later than other hepatitis viral infections. [149]

For HBV, the antibody response to the envelope proteins is a T-cell–dependent process. [150] A minority of patients infected with HBV fail to produce antienvelope antibodies, therefore contributing to the establishment of persistent infection. This deficiency resides at the level of T-cell nonresponsiveness, probably as a result of tolerance or anergy induction toward the HBV proteins. [151] The molecular mechanisms underlying this deficiency, however, are largely undefined. For HAV infection, whether the relapsing or prolonged phase is secondary to transient failure of humoral immune response is unknown. On the other hand, antiviral antibodies may play a role in viral pathogenesis. Cell surface forms of viral antigens on infected hepatocytes could be a potential target for antibody-directed cell cytotoxicity. [152], [153] However, this mechanism has not been shown to be operative in hepatocellular injury associated with any of the viral infections. In contrast, immune complex diseases with complement fixation, including glomerulonephritis, vasculitis, and cryoglobulinemia, which are major extrahepatic manifestations of HBV and HCV infections, can result from abnormally activated antibody response to viral antigens.

The other limb of immune response, the cell-mediated immunity (CMI), is critical for the long-term control of viral infections, including the hepatitis viruses. Although this aspect has only been studied in great detail for HBV and HCV, it is not unreasonable to suspect that CMI may play a role in viral clearance and pathogenesis for HAV, HDV, and HEV. Much of the information for hepatitis B has been gleaned from studies in mouse models, chimpanzees, and infected humans. [151], [154] Nonprimate animal models that are susceptible to hepadnaviral infection (woodchucks, ground squirrels, ducks) have also provided utility in this venue of research. In acute HBV infection, individuals can mount a vigorous, multispecific, and polyclonal cellular immune response to HBV. In contrast, chronically infected patients have a weak or barely detectable anti-HBV response. [151] This is true for both CD4 and CD8 responses. During acute hepatitis B, a vigorous HLA class II–restricted, $CD4^+$, helper T-cell response to multiple epitopes of HBc/eAg predominates in virtually all patients. Such a response to HBV envelope antigens, however, is much less evident. The class II–restricted, $CD4^+$ T cells specific for nucleocapsid play an important immunoregulatory role in the control of viral infection. [151], [155] By helping B cells produce neutralizing antienvelope antibodies and activating HBV-specific cytotoxic T lymphocytes (CTL), this $CD4^+$ helper T-cell population may direct the initial antiviral

response. During chronic HBV infection, the once strong nucleocapsid-specific CD4 $^{+}$ T-cell response becomes barely detectable, presumably owing to a switch to T-cell exhaustion or nonresponsiveness. During exacerbation or successful treatment of chronic hepatitis B, these nucleocapsid-specific T-cell responses could reemerge. [156], [157]

In most viral infections, the activation of virus-specific CD8 $^{+}$ CTLs is critical for viral clearance. These cells can detect and eliminate or cure virus-infected cells through recognition of viral peptides in the context of HLA class I molecules. Using short synthetic peptides mimicking processed viral antigens and cells genetically engineered to express viral proteins, CTL responses to HBV in humans have been studied extensively. [151] These studies indicate that patients acutely infected with HBV develop a strong, polyclonal, HLA class I–restricted CTL response that is directed against multiple epitopes in all viral proteins. Similar to the class II–restricted CD4 responses in a minority of infected people, the class I–restricted CTL responses disappear and are barely detectable during the chronic phase of HBV infection. [155], [158]

Studies of immune responses in HCV revealed that both CD4 and CD8 responses can be detected against various viral proteins during acute infection, but the vigor and character of these responses appear to be directly correlated with the outcome of the viral infection. [147] A vigorous and multispecific CD4 and CD8 response is associated with viral clearance in both human and experimental chimpanzee infections. [159], [160] However, in most acute HCV infections, the infected hosts appear to exhibit little or no CMI and progress to chronic infection. This limited immunity suggests an “immune avoidance” model for HCV infection. A recent study has pointed to a defect in dendritic cells in antigen presentation, presumably as a result of infection of these cells by HCV. [161] Other mechanisms, such as the immunomodulatory effects of HCV core protein and the interferon inhibitory property of the NS5a protein, may also contribute to an overall weak immunity to this viral infection. [162], [163]

In chronic HBV and HCV infections, the detection of virus-specific CD4 and CD8 cells, albeit with low frequency and weak activities, in the peripheral blood and liver of chronically infected individuals suggests a pathogenic relationship between the indolent cellular immune response and necroinflammatory liver disease associated with chronic hepatitis. [151] Therefore, the CMI is a double-edged sword: vigorous response leads to viral clearance, whereas ineffective response results in chronic hepatocellular injury.

For HBV infection, the molecular and cellular mechanisms of viral clearance and hepatocellular injury have been elucidated in the animal models, including both transgenic mice and chimpanzees [151], [154] (Fig. 106-9). Adoptive transfer of murine CD8 $^{+}$, class I–restricted HBsAg-specific CTLs into syngeneic transgenic mice expressing HBV envelope protein in the liver leads to acute hepatitis. [164], [165] The CTLs first target the liver through interaction between the HBV-specific T-cell receptors and the antigen-presenting HLA class I molecules on the hepatocytes and cause scattered apoptosis of hepatocytes. By secreting cytokines, including interferons, the CTLs recruit a variety of antigen-nonspecific inflammatory cells into the liver, resulting in more extensive necroinflammatory injury of the liver that resembles acute hepatitis B in humans. The predominant infiltrating effector cells are the macrophages, which probably mediate most of the hepatocellular injury in this model. [166] The CTLs, although not primarily responsible for most of the hepatocellular injury, initiate the cascade of immunologic events leading to hepatitis in this model. A major consequence of these immunologic events is the marked inhibition of hepatocellular *HBV* gene expression and viral replication. [167] Although the CTLs may contribute to a small proportion of inhibition by direct cytolysis of *HBV*-expressing hepatocytes, a noncytolytic mechanism likely plays a substantial role in this effect. [167] Interferon-a and tumor necrosis factor-a produced by activated CTLs and other effector cells are the key cytokines involved in this mechanism. [167] A recent study in chimpanzees [154] also provided support to the noncytopathic mechanism as the major antiviral mechanism responsible for viral clearance during acute hepatitis. The resident natural killer or natural killer T cells, as discussed previously, provide the first line of defense; the CTLs, together with other inflammatory cells and production of antiviral cytokines, likely constitute the final assault on the virus. The CTL response appears to be long-lived because patients recovered from acute HBV infection harbor HBV-specific CTLs many years after viral clearance. [158] The CTLs have been postulated to persist as a result of continuous stimulation by low-level viral replication and in the meantime keep the infection in check. It is not clear how prevalent is this occult HBV infection, but low-level HBV virus can be frequently detected in patients with serologic evidence of recovery. [168], [169] and [170] Furthermore, active HBV infection can be reactivated in these patients upon immunosuppressive therapy, and their serum or organs are capable of transmitting HBV infection. [171], [172] and [173]

FIGURE 106-9. The immunopathogenesis of hepatitis B. See text for details.

In HCV infection, long-term persistence of virus-specific T cells has also been demonstrated despite evidence of recovery. [174] However, low-level viral infection is probably not the reason for this persistence. It is possible that these virus-specific memory T cells have unusual longevity in vivo and can be rapidly activated upon viral antigen stimulation years after infection. Whether these virus-specific T cells can offer protective immunity to subsequent viral infection is unknown. The observation that chimpanzees can be reinfected despite recovery from previous infection [175], [176] suggests that protective immunity may not persist after viral clearance.

The cellular immune responses to HAV, HDV, and HEV are largely unknown. Although HAV-specific CD8 CTLs have been detected in liver biopsies during acute hepatitis A, their role in viral clearance and pathogenesis remains undefined. [177], [178]

CLINICAL MANIFESTATIONS

Symptoms related to acute viral hepatitis are variable and nonspecific. It is impossible to distinguish one form of viral hepatitis from the other based solely on clinical features. The clinical course of hepatitis varies widely from an asymptomatic phase recognized only by elevation in aminotransferase levels to fulminant hepatitis with frank jaundice and hepatic coma. [179], [180] and [181] About 20% of cases of acute hepatitis present with jaundice. There is a variable incubation period for each virus, ranging from 1 week to 6 months. The initial presentation is dependent on age at exposure, integrity of the host immune system, presence or absence of underlying liver disease, and the level of viral inoculum. Overall mortality is low. An overview of the clinical features of each of the five hepatotrophic viruses is given in Table 106-2.

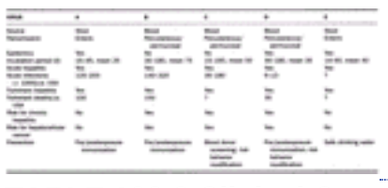

TABLE 106-2 Clinical Overview of the Five Viral Hepatidides

Before the onset of symptoms, there is a prodromal phase that can last from 1 day to 2 weeks and averages 5 to 7 days, during which there are nonspecific constitutional symptoms. By far the most common symptom reported is loss of appetite. Patients report a distinct aversion for food that may worsen as the day

progresses. [182] They may also lose the taste for tobacco and alcohol. Fatigue and weakness are common symptoms reported by 90% of patients and may be severe enough to confine them to bed. Before the onset of jaundice, two thirds to three fourths of patients complain of low-grade fever and flu-like symptoms. Mild headache is reported by 50% to 70% of patients. Vomiting and nausea are experienced by some patients but are not predominant features of acute viral hepatitis, and their continued presence should alert one to consider alternative diagnoses. Diarrhea is not a common feature of acute hepatitis and occurs in less than 25% of cases of HAV infection. Patients often complain of a dull ache under the right rib cage or even frank pain. Closer to the onset of jaundice, the urine may begin to darken.

The prodromal phase is followed by the icteric phase. The onset of jaundice usually coincides with the peak of the serum ALT. The duration of jaundice is variable, ranging from 4 days to several months, but averages 2 to 3 weeks. Patients initially notice a yellow discoloration of the mucous membranes and sclerae, followed later by a sallow discoloration of the skin. The urine becomes intensely dark owing to the release of conjugated bilirubin from necrosed hepatocytes, and stools become pale owing to an inability of the hepatocytes to conjugate bilirubin to its secretory form. During this phase, half of patients may experience itching, which is usually mild and transient. Weight loss of 2 to 10 kg is not uncommon. A minority of patients experience depression.

During the convalescent phase, most symptoms resolve. Jaundice disappears, with a return of normal skin, urine, and stool color. The return of an appetite and weight gain are favorable signs indicating recovery. Fatigue may persist for up to 2 to 6 months.

The physical examination during acute viral hepatitis may be normal, but almost all patients have some degree of hepatic tenderness. Most patients have mild hepatomegaly (12 to 14 cm), and the liver edge should feel soft and smooth. On occasion, it may feel firm to the examining hand, but it should never feel hard or nodular. If these findings are elicited, underlying cirrhosis should be suspected and an acute on chronic hepatitis considered. Splenomegaly and spider angiomas are features not commonly observed with acute hepatitis, but they may occur rarely. Jaundice is present in icteric cases, involving the mucous membranes, sclerae, and skin. The total bilirubin level usually has to be greater than 2.5 mg/dL before jaundice is clinically evident.

Extrahepatic Manifestations

Extrahepatic manifestations may occur in patients with acute viral hepatitis and result from direct infection of another organ or as an immune-mediated phenomenon. A serum sickness–like syndrome may be present in 5% to 15% of patients with acute HBV infection and less so in those with HCV infection. It presents as a low-grade fever, rash, arthralgias, and angioneurotic edema. [183], [184] The rash is usually urticarial but may be maculopapular. The arthralgias tend to involve the large joints, such as the elbows, wrists, knees, and ankles. Symptoms are thought to be immune complex mediated and resolve with onset of hepatitis.

Polyarteritis nodosa (PAN) is well described in association with HBV infection. In one series, PAN occurred in almost one third of newly diagnosed cases of HBV. [185] PAN is thought to result from the deposition of antigen-antibody complexes in the intima of the vessel wall; HBsAg is the suspected antigen. [186], [187] Symptoms include arthralgias, fever, abdominal pain, renal disease, mononeuritis, and skin rash. PAN occurs more often with chronic HBV infection. [188] The mortality rate ranges from 30% to 50%.

Essential mixed cryoglobulinemia (EMC) is a systemic vasculitis characterized by palpable purpura, arthralgias, and weakness. EMC has been linked to hepatitis A, B, and C based on the detection of viral antigens, antibody, and virus in the cryoprecipitate and the vascular lesions. [189], [190] and [191] The association of EMC and viral hepatitis is strongest with HCV infection. [192] Cryoglobulins are proteins that precipitate in the cold and, in the case of HCV infection, may result from HCV infection of B cells [193], [194] with the production of both IgG and IgM antibody; the IgM cross-reacts with rheumatoid factor. [195] The prevalence of cryoglobulins in unselected patients with hepatitis C is unknown. Most cases of HCV-related EMC are associated with chronic infection and do not correlate with viremia or genotype but appear to be related to the duration of infection and to more advanced stages of liver disease.

Renal disease is often seen in patients with acute viral hepatitis, and specific pathology is best characterized for HBV and HCV. Manifestations of renal involvement range from mild proteinuria and abnormal urinary sediment, including red blood cell and hyaline casts, to frank glomerulonephritis with renal failure. The renal lesion is thought to result from the deposition of immune complexes in the glomerular basement membrane. HBsAg, HBeAg, and HCV antigen have been demonstrated in the immune complexes from the glomerular basement membrane. [196], [197] and [198] The most common renal lesions seen in HBV infection were membranous [199] and membranoproliferative glomerulonephritis. [200] Membranous glomerulonephritis occurs mainly in children from endemic regions. The natural history of membranous glomerulonephritis is not well defined. [201] Nephrotic syndrome and proteinuria are the most common presentations. Children are more likely to be asymptomatic and to have spontaneous resolution of the nephrotic syndrome. [199] Progressive renal failure usually develops in adults. Most patients with glomerulonephritis have evidence of active viral replication, but the severity of the renal disease does not correlate with the activity of the liver disease or the viral level in serum. Membranoproliferative glomerulonephritis is the most common renal lesion seen in HCV infection, usually in association with EMC. [202] In the presence of cryoglobulins, 98% to 100% of patients with membranoproliferative glomerulonephritis have detectable anti-HCV. In the absence of cryoglobulinemia, patients with chronic HCV infection may have a variety of renal lesions suggestive of primary glomerular disease including nephrotic syndrome and nonnephrotic proteinuria.

Papular acrodermatitis (Gianotti syndrome) has been described in children with acute hepatitis B. [203], [204] The syndrome is most commonly seen in young children aged 1 to 6 years. It is characterized by erythematous papules on the arms, legs, and face with truncal sparing and lymphadenopathy. The mucous membranes are spared. It is not related to the infecting subtype of HBV. It usually runs a benign course, improving with the onset of hepatitis.

Other organ systems affected during viral hepatitis, although rarely, include the heart and gastrointestinal and nervous systems. Cases of myocarditis, pericarditis and bradycardia, and pericardial and pleural effusions have been described. Pancreatitis is commonly reported, especially in children, [205] and may be due to direct infection of the gland in the case of HAV infection. Neurologic manifestations have included encephalitis, meningoencephalitis, mononeuritis multiplex, transverse myelitis, and Guillain-Barré syndrome. [206], [207] and [208] A variety of hematologic disorders may follow viral hepatitis, including pure red cell aplasia, pancytopenia, aplastic anemia, and hemolytic anemia.

DIAGNOSIS AND DIFFERENTIAL DIAGNOSIS

The diagnosis of acute hepatitis is based on the constellation of clinical, biochemical, histological, and serologic findings. The CDC case definition of acute viral hepatitis is an illness with a discrete date of onset, accompanied by jaundice or elevated serum aminotransferase levels greater than 2.5 times the upper limit of normal. The clinical symptoms associated with viral hepatitis were described in the previous section and are nonspecific for etiology. The specific diagnosis depends on the results of serologic testing. The sensitivity of current serologic assays should allow the etiology of acute viral hepatitis to be established in almost all cases by the time patients become symptomatic. In instances in which there may be a late or weak antibody response, sensitive RNA and DNA assays may be helpful in the diagnosis.

Serology

In the setting of a known exposure and appropriate symptoms with elevated serum aminotransferases, acute hepatitis A is diagnosed by the detection of IgM antibody to HAV (anti-HAV IgM) and the absence of other viral markers in serum. [209], [210] Anti-HAV IgM is usually present in serum 5 to 10 days into the incubation period and remains detectable for up to 6 months after infection, when IgG type antibody rises to high titer and persists for life [211] (Fig. 106-10). Commercial assays for total anti-HAV measure both IgG and IgM, and thus a positive test result is not helpful in distinguishing acute from chronic infection. HAV RNA can also be detected in serum and stool during the incubation phase but is primarily a research tool and is not used for diagnostic purposes.

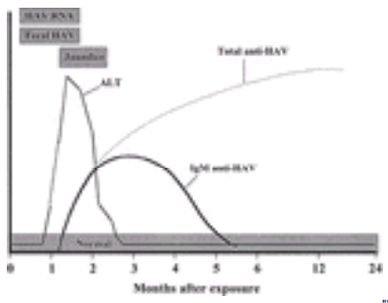

FIGURE 106-10. Typical serologic course of acute hepatitis A virus infection.

A number of viral antigens and their respective antibodies can be detected in serum after infection with HBV, and their interpretation is often the source of great confusion to physicians and patients alike. HBsAg and HBeAg are the first two viral proteins detected in serum [146] (Fig. 106-11). HBsAg may be detected as early as 1 to 2 weeks or as late as 11 to 12 weeks after infection, and its persistence is a marker of chronicity. HBeAg is considered a marker of HBV replication and infectivity. It is more important for assessing risk for transmission in chronic HBV infection. [212] The level of HBcAg in serum is usually too low to detect but can be visualized by immunostaining of liver tissue. A cytoplasmic distribution of HBcAg in hepatocytes was shown to correlate with disease activity.

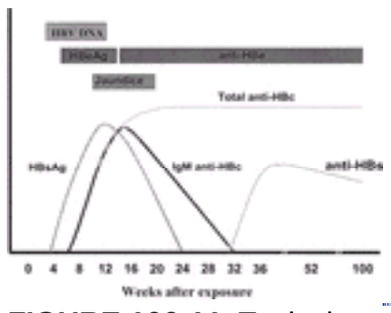

FIGURE 106-11. Typical serologic course of acute hepatitis B virus infection.

Antibodies to three of the viral proteins (HBsAg, HBeAg, and HBcAg) can be detected in convalescent sera. Anti-HBc IgM class is the first to appear, followed by antibody to HBeAg (anti-HBe) and finally antibody to HBsAg (anti-HBs), which is the marker of recovery. Anti-HBc IgG type persists after recovery and is a useful marker for prevalence studies of past exposure to HBV. [213] It should not be used to diagnose acute or chronic infection. HBsAg and anti-HBc IgM are the most frequent markers present during the symptomatic phase of the illness. [146] , [214] Rarely during acute HBV infection, HBsAg is rapidly cleared from serum, and anti-HBV IgM may be the only marker detectable; this scenario is referred to as the “window” period. This situation may also be seen during the convalescent phase of the illness when HBsAg and aminotransferase levels are declining and anti-HBs is not yet detectable. Given the sensitivity of current assays for HBsAg, this window period is rarely observed in clinical practice. Diagnosis of acute hepatitis B rests on the demonstration of anti-HBV IgM and HBsAg and negative tests for anti-HAV and anti-HCV. HBV DNA is present during the incubation and symptomatic phases of infection. Like HBeAg, it correlates with disease activity and infectivity. Current PCR-based assays have a cutoff of 100 virions per milliliter of blood, but routine testing for HBV DNA is not recommended unless the patient is immunocompromised. [215]

Acute hepatitis C infection is difficult to recognize because fewer than 20% of cases present with jaundice. [216] Anti-HCV can be detected as early as 4 weeks after acute infection and in the majority of cases remains detectable lifelong [217] , [218] (Fig. 106-12). The diagnosis of acute hepatitis C infection should be based on a history of exposure, absence of prior liver disease, negative tests for other hepatitis viruses, and a positive test for anti-HCV. [219] A supplemental confirmatory test, such as the radioimmunoassay blot assay, should be performed. HCV RNA is detectable within 2 weeks of established infection and is also used to confirm acute infection. [220] However, routine HCV RNA testing is not currently advocated owing to the lack of standardization of the test. It is recommended in the investigation of immunocompromised patients who may have a low or absent antibody response.

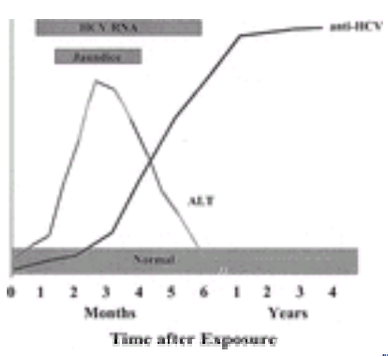

FIGURE 106-12. Typical serologic course of acute hepatitis C virus infection.

Acute delta hepatitis may occur in two settings: simultaneously with HBV (co-infection) or after infection of a chronic hepatitis B carrier (superinfection). The serologic pattern differs in each case. During co-infection, markers of HBV are usually detected first, followed later by markers for HDV. The presence of anti-HBc IgM is an important finding of HDV co-infection and a discriminating marker for HDV superinfection. Early in the course of co-infection, hepatitis D antigen (HDAg) is not detectable unless the hepatitis is severe, but within 1 to 2 weeks, antibody to hepatitis D (anti-HDV) IgM should be detectable (Fig. 106-13). Anti-HDV IgG is usually delayed for several weeks after the onset and in some cases is present only transiently during the convalescent phase. The late and poor antibody response in acute delta co-infection makes the diagnosis difficult. It is advisable to perform repeat testing for anti-HDV IgM to confirm HDV co-infection. HDV RNA can be detected early in the course of co-infection, but commercial assays are not currently available. Thus, the diagnosis of acute delta co-infection depends on the detection of anti-HDV IgM, HBsAg, and anti-HBc IgM.

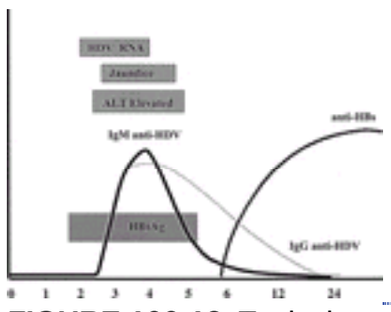

FIGURE 106-13. Typical serologic course of acute hepatitis D virus co-infection.

Acute delta superinfection occurs in the setting of chronic hepatitis B. HDAg and HDV RNA can be detected early in the course of infection by experimental assays. In contrast to HDV co-infection, anti-HDV IgM and IgG are both present early during the symptomatic phase of acute delta superinfection (Fig. 106-14). Anti-HBc IgM is usually absent or present in low titer. Diagnosis of acute delta superinfection thus rests on the detection of anti-HDV and HBsAg and absence of anti-HBc IgM.

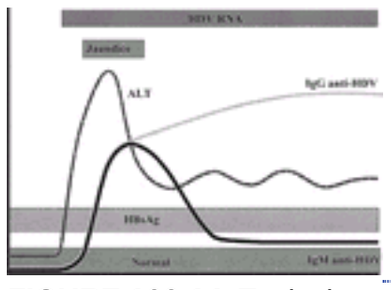

FIGURE 106-14. Typical serologic course of acute hepatitis D virus superinfection.

Diagnostic tests for antibody to HEV (anti-HEV) are now available. In outbreak settings due to HEV, anti-HEV IgM is detectable in greater than 90% of patients' sera during the symptomatic phase and remains detectable for a period of 2 months after the onset of illness (Fig. 106-15). Anti-HEV IgG persists after recovery and appears to be protective against reinfection, at least in the short-term. In the appropriate setting, diagnosis of acute hepatitis E is confirmed by the demonstration of anti-HEV IgM.

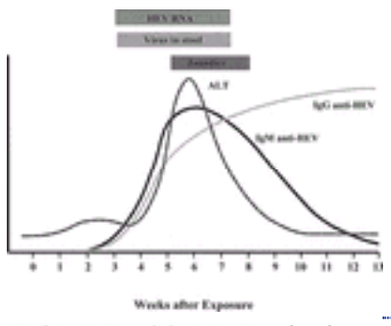

FIGURE 106-15. Typical serologic course of acute hepatitis E virus infection.

Laboratory Features

The laboratory hallmark of acute viral hepatitis is an elevation in the serum aminotransferase level. [221], [222] The serum ALT level is more specific for hepatocyte necrosis and is usually higher than the serum aspartate aminotransferase level. During an acute hepatitis infection, levels range from 10 times the upper limit of normal to greater than 20 times the upper limit of normal and peak with the onset of jaundice. The height of the ALT or aspartate aminotransferase elevation usually correlates with the level of destruction of hepatocytes but not with clinical outcome. The serum alkaline phosphatase level, a marker of biliary epithelial cell damage, may rise to two to three times the upper limit of normal but in most cases remains in the normal range. The conjugated and unconjugated serum bilirubin fractions are elevated. The level is dependent on the extent of hepatocyte damage but typically ranges from 85 to 340 µmol/L (5 to 20 mg/dL). In fulminant cases, laboratory markers of hepatic function become markedly abnormal: the prothrombin time prolongs, serum albumin falls, and higher bilirubin levels are seen. These are ominous signs that indicate impending hepatic failure and should prompt early patient referral to a liver transplantation center. During the acute phase of the illness, leucopenia with neutropenia and lymphopenia may be observed. Rarely, aplastic anemia may complicate acute viral hepatitis.

Histology

A liver biopsy can distinguish acute from chronic hepatitis and viral from nonviral causes but is rarely necessary to establish the diagnosis. Regardless of the etiology, acute viral hepatitis has certain characteristic findings.

There is an acute inflammatory response involving the entire liver. [223], [224] Hepatocyte damage is greatest near the terminal hepatic venules (zone 3). Here, the classic histological features of acute hepatitis are observed. The hepatocytes appear swollen (ballooning change). Ballooned hepatocytes have a granular, pale-staining cytoplasm with enlarged nuclei and are occasionally multinucleated. Numerous perivenular hepatocytes undergo acidophil change as a result of apoptosis. Their cell cytoplasm becomes densely stained and irregular in shape with pyknotic nuclei. These cells are thought to be the precursor of the acidophil body (Councilman body). The acidophil body is not specific for viral hepatitis but is a characteristic feature when present in abundance. The remaining hepatocytes show regenerative hyperplasia. The combination of cell swelling, cell shrinkage, apoptotic body formation, and cell loss gives rise to disruption of the liver cell plate pattern or lobular disarray. [225] When only a few hepatocytes are involved, the damage is designated *focal necrosis*. When groups of adjacent hepatocytes are involved, it is termed *confluent necrosis*.

Inflammatory cells are present within the hepatic lobule, among the damaged cells. Lymphocytes and macrophages are the major cell types. Plasma cells, neutrophils, and eosinophils are present to varying extent. Kupffer cell hyperplasia is recognizable. The mononuclear cell infiltrate also involves the portal tract. The inflammatory infiltrate may spill out of the portal tract, but this should not be confused with piecemeal or interface necrosis, a feature of chronic viral hepatitis. Surprisingly, despite the extensive destruction of liver tissue, the reticulin framework of the liver remains remarkably preserved. Cholestasis may or may not be present. Bile ductular proliferation may be seen but is not a prominent feature. Mild fatty change is seen more frequently with HCV infection. Liver fibrosis is generally absent but may be seen if the biopsy is done late in the course of the illness.

In some cases, the confluent necrosis is severe enough to involve the entire central zone of the acinus. The necrotic cells are replaced by the condensed reticulin framework and inflammatory cells, predominantly macrophages forming central-to-portal bridging necrosis. This entity may be the precursor to fulminant hepatitis, and in one series, its presence was associated with a poorer outcome. [226] In extreme cases, there may be involvement of all zones of the acinus, referred to as *massive necrosis* with total loss of hepatocytes. The parenchyma is replaced by the collapsed reticulin framework and inflammatory cells. This is a manifestation of severe hepatitis seen in the clinical setting of fulminant hepatitis and coma.

The histological changes associated with acute viral hepatitis persist for several weeks after onset. Thus, a nearly normal liver biopsy 2 to 3 weeks after an episode of jaundice effectively rules out acute hepatitis. Late biopsy findings include a decrease in the degenerative changes, macrophages that stain intensely with periodic acid-Schiff stain, and numerous mitotic figures.

Staining the liver tissue with antibodies for hepatitis B surface and core antigens in the case of hepatitis B and delta antigen for delta hepatitis can help with the identification of the etiologic agent of the hepatitis.

Differential Diagnosis

An array of diseases may mimic acute viral hepatitis and should always be considered in the differential diagnosis while awaiting the results of serologic testing. As with other acute medical illnesses, rapid diagnosis permits the prompt institution of specific therapy that may prevent an adverse outcome. The differential diagnosis

of acute viral hepatitis is summarized in Table 106-3.

TABLE 106-3 Acute Viral Hepatitis: Differential Diagnosis, Diagnostic Testing and Specific Therapy

The prevalence of drug-induced liver disease is probably underestimated in clinical practice. Drug-induced liver disease should be considered in every case of acute hepatitis until the cause is firmly established. A wide variety of over-the-counter drugs are associated with hepatic toxicity, which may result in a clinical picture indistinguishable from acute viral hepatitis. Therefore, it is mandatory to obtain a detailed drug history from every patient who presents with acute hepatitis. Clinical clues to the diagnosis are the timing of the rise in serum aminotransferase in relation to the onset of jaundice and presence of a serum eosinophilia. In drug-induced liver disease, the jaundice tends to lag behind the rise in aminotransferases, whereas in viral hepatitis, the serum aminotransferase level tends to peak with the onset of jaundice.

Autoimmune hepatitis may present as an acute hepatitis in up to 30% of cases. [227], [228] The typical presentation is that of a young woman with a high titer of antinuclear antibody and smooth muscle antibody and other clinical evidence of autoimmune disorders such as arthralgias and autoimmune hemolytic anemia. The globulin level may not be elevated during an acute presentation. Testing for the HLA markers A1, B8, and DR3 may be helpful in confirming the diagnosis of autoimmune hepatitis.

Wilson disease is “the great imitator” and has diverse presentations, including that of acute hepatitis. [229] The diagnosis is difficult to confirm during an acute presentation and requires a high index of suspicion. It is important to establish the diagnosis early because of an extremely high mortality rate. In a recent series of fulminant hepatitis, no patients with Wilson disease referred to a transplantation center survived despite undergoing OLT. [230] The usual diagnostic criteria may be normal during an acute presentation. Ceruloplasmin level may be normal because of widespread release from necrosed hepatocytes, and urinary copper excretion may be low owing to altered renal clearance. Diagnostic clues may be the young age of the patient, a low alkaline phosphatase level, and the presence of a hemolytic anemia.

The liver may be an innocent bystander in infections that have multisystem involvement. Yellow fever, malaria, and Q fever may involve the liver and cause a hepatitis-like picture. Epstein-Barr virus (EBV), cytomegalovirus (CMV), herpesviruses, and adenoviruses may cause severe infection in immunocompromised patients and those at the extremes of age. These agents should be suspected when routine serologic testing is negative in an immunocompromised patient or when a history of recent travel to an endemic area is obtained from the patient.

Acute cardiovascular failure with hypotension from pump failure or sepsis can result in ischemic injury to the liver, so-called shock liver. In this setting, there can be massive hepatocyte destruction with serum aminotransferase levels greater than 20-fold the upper limit of normal, mimicking acute viral hepatitis. Bilirubin levels usually are only mildly elevated. The history of hypotension and aminotransferase levels that rapidly normalize should establish the diagnosis.

Given the vast number of metabolic functions carried out by the hepatocyte, it is surprising that toxic injury is not seen more commonly. Mushroom poisoning from ingestion of *Amanita phalloides* and exposure to carbon tetrachloride are the best examples of toxic injury leading to markedly elevated aminotransferase levels. The diagnosis depends on a careful medical history indicating ingestion or exposure.

Rarely, biliary tract obstruction (choledocholithiasis or acute cholecystitis) can be confused with acute hepatitis. [231] Usually, other clinical features are present to suggest the diagnosis, such as an elevated serum alkaline phosphatase level and more intense right upper quadrant pain.

Finally, in special populations such as bone marrow transplant recipients, unique diseases should be considered during the investigation of an acute rise in serum aminotransferase levels. These include acute graft versus host disease, which typically occurs within the first 28 days after bone marrow transplantation, and venoocclusive disease, which occurs within the first 2 to 3 weeks. [232], [233] and [234] In pregnancy, acute fatty liver and the HELLP syndrome (*h*emolysis, *e*levated *l*iver enzymes, and *l*ow *p*latelets) should be considered. [235], [236]

COURSE AND COMPLICATIONS

Course

Most cases of acute hepatitis are uncomplicated and resolve without sequelae. This is the expected outcome after infection with HAV and HEV in 99% of instances. One to 2% of cases are complicated by a fulminant course with a high mortality rate. Hepatitis B, HCV, and HDV may progress to a chronic hepatitis in 10% to 90% of cases depending on the infecting virus, age at infection, and immune status of the host. This is recognized by persistent elevation in the aminotransferase levels 6 months after the onset of infection and detection of specific serologic or virologic markers. Chronic hepatitis is best assessed by liver biopsy. Occasionally, an acute hepatitis has a prolonged course over 4 months and up to 1 year, typified by the prolonged course of HAV infection. This variant needs to be differentiated from chronic hepatitis, and the best means of doing so is watchful waiting until the aminotransferase levels return to normal.

The prolonged form of HAV infection is characterized by pruritus, fever, diarrhea, and weight loss with serum bilirubin levels greater than 10 mg/dL and a clinical course lasting a minimum of 12 weeks. [237], [238] It is seen more commonly in males and older individuals. Importantly, all patients recover without lasting sequelae. Some patients with this variant of hepatitis A may benefit from corticosteroid therapy. [238]

In addition to the prolonged variant of HAV infection, a relapsing variant is seen in up to 20% of HAV cases, distinguishable from the protracted form by its biphasic presentation. [239] Patients initially present with an acute hepatitis followed by apparent recovery only to have a recrudescence of hepatitis 5 to 10 weeks later. The second phase tends to be associated with higher bilirubin levels and to have a more prolonged course compared with the first phase. [240] Steroid therapy during the early phase of the illness and alcohol consumption have been identified as potential risk factors for relapsing HAV infection. [239]

Despite these complications, complete resolution is the rule. Serologic evidence of HAV IgM in serum during recrudescence of the illness and detection of HAV in stool during relapse are helpful tests for excluding other cause of hepatitis. The pathophysiology of these atypical presentations is currently unknown. A recent study in children reported an association between the protracted form of hepatitis A and HLA-DRB1*1301, a marker for pediatric autoimmune hepatitis. [241] Case reports have noted that acute hepatitis A infection may trigger the onset of autoimmune hepatitis in adults. [242], [243]

Other atypical courses of viral hepatitis have been described, which sometimes lead to diagnostic confusion. A biphasic pattern of hepatitis may also be observed after hepatitis D co-infection, although cholestasis is not a feature. The first peak is related to HBV infection and the second to HDV. On occasion, HBsAg-negative delta hepatitis is encountered during acute infection, owing to suppression of HBV replication by HDV. [244] This presentation can be quite perplexing for the clinician, and repeat testing for anti-HBc IgM and markers of HDV infection are recommended in suspected cases. Flares of chronic hepatitis B or reactivation are well recognized in association with chemotherapy or immunosuppression. It is important to distinguish reactivation from acute infection or nonviral causes of hepatitis because the management is different. The diagnosis of reactivation can be tricky. In most cases, the clinical picture is recognizable by elevated aminotransferase levels with detectable HBsAg and absent anti-HBV IgM in serum. However anti-HBV IgM may be detectable in 25% of cases of reactivation, leading to confusion with acute hepatitis B. The only way to resolve this dilemma is to obtain a history from the patient of previous HBV infection or to perform a liver biopsy, which should

demonstrate changes of both acute and chronic hepatitis in the case of reactivation.

Complications

Fulminant hepatitis is a complication of acute hepatitis characterized by a rapid progression to hepatic failure with associated hepatic encephalopathy. Only 1% to 2% of all cases of acute viral hepatitis result in fulminant hepatitis, but 75% of fulminant cases are caused by viral hepatitis. Fulminant hepatitis has been reported with all of the hepatotrophic viruses. [245], [246], [247] and [248] Fulminant hepatitis has an extremely high mortality rate; survival has improved with the widespread availability of OLT. [249]

Older individuals with hepatitis A may be at increased risk for fulminant hepatitis. [250] A report of a high rate of fulminant hepatitis A in patients with underlying chronic hepatitis C [251] has not been substantiated by other studies. [252], [253]

Epidemics of fulminant hepatitis B have been reported with variant strains of the virus. In several studies, molecular characterization of the virus revealed mutations that led to inactivation of the gene encoding HBeAg. [254], [255], [256] and [257] However, other studies showed a low rate of these mutations in fulminant hepatitis B, and the mutations were also found to arise de novo in patients with chronic HBV infection who did not develop fulminant hepatitis B, suggesting that other factors play a role. [258], [259], [260] and [261] Fulminant hepatitis B has also been reported with emergence of the YMDD variant under lamivudine therapy. [262], [263]

Fulminant hepatitis may occur more frequently with HDV co-infection. [264], [265] In addition, periodic epidemics of a severe fulminant form of delta hepatitis have been described in the Amazon basin, referred to locally as *LaBrea hepatitis* and may be related to the particular genotype of HDV, genotype 3. [266]

Fulminant hepatitis E occurs at surprisingly high rates in pregnant women during the third trimester, ranging from 15% to 25%. [267], [268] The reason for this is currently unknown. Survival is poor.

A hepatitis-associated aplastic anemia and pure red cell aplasia may complicate acute hepatitis and is associated with a high mortality rate. [269], [270] and [271] It appears to be more common in the Far East and in adolescent boys and young men. [272]

Chronic hepatitis is a complication seen only with HBV, HCV, and HDV. Rates of chronic infection depend on immune status of the host and age at time of infection and vary from 10% to 90%. The causes of chronicity are unknown but undoubtedly involve a failure of the cellular immune response.

Other long-term complications of acute viral hepatitis include hepatocellular carcinoma, especially in the case of HBV and HCV infection, and lymphoma associated with HCV infection.

PREVENTION AND IMMUNOPROPHYLAXIS

Hepatitis A Virus

Hepatitis A infection can be prevented by three strategies: active immunization through vaccination, which has the advantage of lifelong immunity; passive immunization through the administration of antibody made from pooled human sera processed by cold ethanol fractionation; and a combination of the two strategies.

Passive Immunization Immunoglobulin provides protection against hepatitis A infection by the passive transfer of antibody. It can be used to provide immunity before and after exposure. The recommended dose is 0.02 mL/kg of immunoglobulin administered intramuscularly for both preexposure and postexposure prophylaxis. If used for preexposure prophylaxis, a dose of 0.02 mL/kg provides protection for less than 3 months. A higher dose of 0.06 mL/kg will provide protection for about 5 months. For postexposure prophylaxis, administering immunoglobulin within 2 weeks of the exposure is greater than 85% effective. [273] Later administration is ineffective in preventing infection but will attenuate the course of disease. [274] Serious adverse events are rare after administration of immunoglobulin, but anaphylaxis has been reported after repeat administration to patients with known IgA deficiency. [275] It is safe to administer during pregnancy or breast-feeding and can be coadministered with inactivated vaccines, oral polio vaccine, and yellow fever vaccine. [3] Immunoglobulin can interfere with the immune response to other live attenuated vaccines, and these vaccines should be postponed for about 2 weeks. [3]

Active Immunization Two commercially inactivated hepatitis A vaccines are available in the United States: HAVRIX and VAQTA. Both vaccines are highly immunogenic in adults, adolescents, and children, with 94% to 100% achieving protective levels of antibody 1 month after a single dose of vaccine. [276] The lower limit of antibody required to prevent HAV infection is unknown, but in vitro studies indicate that levels of less than 20 mIU/mL can be neutralizing. [276], [277] The efficacy of both vaccines was demonstrated in clinical trials. HAVRIX was shown in a large double-blind randomized controlled trial to have an efficacy of 94% after two doses 1 month apart. [278] Similarly, VAQTA was evaluated in a double-blind, placebo-controlled, randomized trial and had an efficacy of 100% after one dose of vaccine. [279] The vaccine has not been evaluated in the postexposure setting. A small randomized trial showed that hepatitis A vaccine was 79% efficacious in preventing HAV IgM positivity after household exposure to hepatitis A compared with no treatment. [280] Hepatitis A vaccine has not been directly compared to immunoglobulin in postexposure prophylaxis. The vaccine appears to confer long-term protection against infection in studies, with follow-up ranging from 6 to 7 years. [281], [282] Hepatitis A vaccination provides preexposure protection against HAV infection, and it is recommended for people who are at increased risk and for anyone wishing to obtain immunity. Individuals who are at increased risk for hepatitis A infection and who should be routinely vaccinated are summarized in Table 106-4. Both vaccines appear to be quite safe. In Europe, Asia, and the United States, about 8 million doses of hepatitis A vaccine have been administered, and serious events, anaphylaxis, Guillain-Barré syndrome, brachial plexus neuropathy, transverse myelitis, multiple sclerosis, encephalopathy, and erythema multiforme, although rare, have been reported. [278]

Group at Risk
People traveling to or working in countries that have high or intermediate endemicity of infection
Men who have sex with men
Illegal drug users
People who have occupational risk factors for infection
People who have clotting factor disorders

TABLE 106-4 People for Whom Hepatitis A Vaccine Is Recommended

The hepatitis A vaccine can be safely coadministered with the hepatitis B vaccine without affecting the immunogenicity of either vaccine or increasing the frequency of side effects. [283] Safety has not been evaluated in pregnancy, but because the vaccine is inactivated, the risk to the fetus is expected to be low. Similarly, no special precautions are required when vaccinating immunocompromised patients. [284] The vaccine is safe and effective when administered to patients with chronic liver disease. [285], [286] Prevaccination serologic testing for susceptibility is not recommended because of the additional costs and because vaccination poses no risk to a person with a history of hepatitis A infection. Postvaccination testing is not recommended because of the high rate of response. A combined hepatitis A and B vaccine is now commercially available.

Hepatitis B Virus

The goals of vaccination against hepatitis B are similar to those for hepatitis A: to prevent infection in susceptible individuals and to prevent spread of infection and chronic liver disease. Additionally, vaccination against hepatitis B can prevent infection with delta virus and prevent hepatocellular carcinoma related to HBV infection. HBV infection can be prevented by passive or active immunization or a combination of the two.

Passive Immunization Hepatitis B immune globulin (HBIG) is prepared from plasma that contains high-titer anti-HBs. In the United States, HBIG has an antibody titer of more than 100,000 IU/mL by radioimmunoassay. HBIG is used in conjunction with hepatitis B vaccination for postexposure prophylaxis to prevent transmission of HBV from a HBsAg-positive mother to her child or alone to prevent reoccurrence of HBV infection after OLT.

Active Immunization Two yeast-derived recombinant hepatitis B vaccines are available in the United States. A plasma-derived vaccine that was licensed for use in the United States in 1982 is no longer available. The vaccine is part of the routine immunization program in many countries and is very effective in reducing the incidence of HBV infection in endemic areas of the world. The recommended schedule is a series of three intramuscular doses of hepatitis B vaccine into the deltoid

muscle of adults and anterolateral thigh of neonates and infants. The recommended dosing schedule induces an antibody response (anti-HBs titer higher than 10 mIU/mL) in more than 90% of adults and more than 95% of children and adolescents. [287], [288] Increasing the interval between the first and second doses of HBV vaccination has little effect on immunogenicity or final antibody titer. The third dose confers optimal protection. Longer intervals between the second and third doses result in higher final titers of anti-HBs. [289] Administration of hepatitis B vaccine does not interfere with the antibody response to other vaccines. [290] The immune response is comparable to a standard course of vaccination if the product of one manufacturer is used with that of another. Hemodialysis patients usually require a higher dose or a greater number of vaccinations to achieve an adequate immune response. [291], [292] This probably also applies to immunocompromised individuals but has not been confirmed. Clinical trials in the United States and other countries have shown that the hepatitis B vaccine is 80% to 95% effective in preventing HBV infection in susceptible people. [288], [293], [294] and [295] If a protective antibody response is elicited, then the vaccine is 100% effective. Studies suggest that even patients who fail to develop an adequate immune response to the vaccine (anti-HBs titer less than 10 mIU/mL) are probably protected against infection. Likewise, individuals who lose detectable antibody are probably protected because of lasting immunologic memory. The vaccine appears to afford long-term protection. Studies in which the plasma-derived vaccine was used indicate that 13% to 60% of patients may lose antibody after 9 years of follow-up. [296], [297] and [298] The efficacy of the hepatitis B vaccine to prevent transmission of HBV was shown in several studies after introduction of vaccine programs in endemic regions. Before vaccination, the carrier rate of HBV infection in Taiwan was estimated to be 15% to 20%. A program of universal vaccination was initiated in 1984. This program was highly effective in reducing the rate of chronic HBV infection. Over the next decade, the rate of chronic HBV infection fell from 10.5% to 1.7% in children aged 6 years, and the annual incidence of hepatocellular carcinoma fell from 0.7 per 100,000 to 0.36 per 100,000. [299], [300] and [301] These findings were confirmed by other studies. [302] Vaccination is not contraindicated in pregnant women or in those who are breast-feeding. Postvaccination testing is not recommended after routine vaccination of children or adolescents. Testing for immunity is advised only for people whose subsequent management depends on their immune status, such as infants born to HBsAg-positive mothers, dialysis patients and staff, and patients with HIV infection. [298] Currently, booster doses are not recommended for vaccinated individuals, nor is routine serologic testing to determine immune status, unless a person was exposed to the virus. Revaccination of nonresponders to a primary course of vaccine should be considered because 15% to 25% develop an adequate antibody response after one dose and 30% to 50% after three doses of vaccine. [296] For hemodialysis patients, vaccine-induced immunity may wane, and patients should probably undergo testing for antibody levels annually and receive a booster dose if anti-HBs levels fall below 10 mIU/mL. [298] Pain and fever are the most common side effects associated with HBV vaccination; however, in placebo-controlled trials, these side effects occurred no more frequently than with placebo. [287], [288], [293], [295] Over 1 billion doses have been administered worldwide, and serious adverse events occur rarely. [22] A possible association between the plasma-derived vaccine and Guillain-Barré syndrome and demyelinating diseases has been reported. [303] Two recent studies failed to confirm an association between hepatitis B vaccine and multiple sclerosis. [304], [305] In addition, there are large differences in the geographic prevalence of HBV and incidence of multiple sclerosis. Furthermore, analysis of postmarketing studies in the United States showed no increase in multiple sclerosis after hepatitis B vaccination, and reanalysis of the French data by the Viral Hepatitis Prevention Board and World Health Organization concluded there was no association between hepatitis B vaccination and multiple sclerosis. [306] Variant viruses, with “escape mutations” in the immunodominant “a” determinant region of the HBsAg gene after vaccination, have been described. These variants have been reported worldwide. Therefore, a theoretical concern of hepatitis B vaccination is the replacement of wild-type virus by the variant strain to which vaccinated individuals in the community will be susceptible. A report from the Taiwan vaccination program described an increased prevalence of vaccine escape variants from 8% to 28% over a 10-year period, lending credence to this concern. [307] However, this finding has not been confirmed in other studies. [308], [309] Furthermore, chimpanzees vaccinated with commercial hepatitis B vaccines were protected from infection after challenge with an infectious strain of the variant virus. [310] Hepatitis B vaccination is recommended for the prevention of perinatal HBV infection, for infants born to HBsAg-positive mothers, and as part of the universal vaccination of all preschool-aged children. [298] Individuals who are at high risk for acquiring HBV infection should receive hepatitis B vaccination and are listed in Table 106-5. The recommendations for use of the hepatitis B vaccine in the postexposure setting are outlined in Table 106-6.

Group at Risk
People with occupational risk
Clients and staff of institutions for the developmentally disabled
Hemodialysis patients
Recipients of clotting factor concentrates
Household contacts and sex partners of HBV carriers
Adoptees from countries where HBV infection is endemic
International travelers
Injection drug users
Sexually active men and women
Inmates of long-term correctional facilities

TABLE 106-5 People for Whom Hepatitis B Vaccine Is Recommended

EXPOSURE	RECOMMENDATION
Perinatal	Vaccination + HBIG
Sexual (acute case)	HBIG +/− vaccination
Sexual (chronic carrier)	Vaccination
Household (acute case)	None
Household (acute case, known exposure)	HBIG +/− vaccination
Household contact (chronic carrier)	Vaccination
Inadvertent percutaneous/permucosal	Vaccination +/− HBIG

HBIG, hepatitis B immunoglobulin.

TABLE 106-6 Recommendations for Postexposure Immunoprophylaxis for Hepatitis B

Hepatitis C Virus

No effective vaccine is available to prevent hepatitis C infection. This is primarily due to a lack of neutralizing antibody, the high mutation rate of the viral genome, existence of viral quasispecies, and a lack of knowledge about protective immunity. Protection against infection after challenge with a homologous strain has been demonstrated in chimpanzees using hyperimmune serum against region 1 of the E2 protein but not against heterologous strains. [311], [312] and [313] Studies in chimpanzees using a DNA vaccine encoding the E2 protein failed to protect against infection with a homologous strain of virus but may have attenuated the course of infection and prevented chronicity. [314]

Studies in the late 1970s showed that use of immunoglobulin failed to prevent transmission of non-A, non-B hepatitis (HCV). [315], [316] Intravenous infusions of immunoglobulin may attenuate the severity of non-A, non-B hepatitis (HCV). [317], [318] Until further studies are carried out, use of immunoglobulin as postexposure prophylaxis for HCV infection cannot be recommended.

Hepatitis D Virus

The dependence of HDV replication on HBV means that the same preventative and postexposure measures use to prevent HBV infection can be employed to prevent hepatitis D co-infection. Risk behavior modification is the only current measure to prevent HDV superinfection.

Hepatitis E Virus

A recombinant vaccine for HEV is being evaluated in Nepal. Immunoglobulin has not been evaluated for postexposure prophylaxis.

Needle-Stick Injury Transmission of blood-borne pathogens through needle-stick injury is an important issue for health care workers. It is a significant contributor to job-related injuries and emotional stress for health care workers. Each year, about 600,000 to 800,000 needle-stick injuries occur in the United States. Infection with HBV, HCV, or HIV constitutes the major risk for the health care worker after needle-stick injury. The risk for HBV infection is primarily related to the degree of contact with blood in the work environment and to the HBeAg status of the source. In studies of nosocomial transmission of HBV through needle-stick injury, the risk for clinical hepatitis if the source was HBsAg and HBeAg positive was 22% to 31%; the risk for serologic evidence of HBV infection was 37% to 62%. In contrast, the risk for developing clinical hepatitis from a contaminated needle from an HBsAg-positive, HBeAg-negative individual was 1% to 6%, and the risk for HBV seroconversion 23% to 37%. [319] Although percutaneous exposure is an efficient mode of transmission of HBV, it presently accounts for a relatively small number of nosocomial infections as a result of increased hepatitis B vaccine use in health care workers. HCV is not transmitted efficiently by needle-stick injury. The rate of seroconversion to anti-HCV after needle-stick injury averages 1.8% per injury and ranges from 0% to 7%. [320], [321] About 2.4% of new cases of HCV infection are due to needle-stick injury. A deep injury, use of a hollow-bore needle, and visible blood on the needle have been identified as risk factors for transmission of HCV. [320] Exposure

prevention is the primary strategy to reduce HBV and HCV transmission by needle-stick injury. Health care workers should be educated concerning the risk for and prevention of blood-borne infections, including vaccination against HBV. Health care workers were required to adopt universal precautions regulations that were introduced in 1987. [322] Despite these recommendations, needle-stick injuries continue to occur. Regulations to prevent needle-stick injury should be enforced in the workplace. General management after a needle-stick injury involves washing the wound with soap and water. The person whose blood was the source of exposure should be evaluated for evidence of HBV, HCV, and HIV infection. No special precautions are necessary to prevent secondary transmission during the follow-up period. However, exposed people should refrain from donating blood, plasma, organs, tissue, and semen. The exposed person does not need to modify sexual practices or to avoid becoming pregnant. Employers should follow all federal and state requirements for recording and reporting occupational injuries and exposures. [323], [324] Specific recommendations for postexposure management are outlined by the U.S. Public Health Service. [325] Any unvaccinated person who sustains a needle-stick injury should receive the hepatitis B vaccine series. The recommendations for management of exposure based on HBsAg status of the source and vaccine status of the exposed individual are shown in Table 106-7. Both HBIG and hepatitis B vaccine can be administered simultaneously.

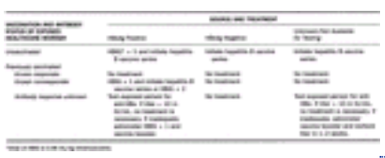

TABLE 106-7 Recommendations for Postexposure Prophylaxis for Health Care Workers Following Exposure to HBV

No specific therapy is available for prevention of HCV infection after needle-stick injury. Recommendations for postexposure management are intended to achieve early identification of chronic cases and, if present, to refer to a specialist for treatment options. The source of exposure should be tested to determine anti-HCV status. Baseline anti-HCV and ALT testing should be performed on the exposed individual and repeated in 4 to 6 months. If desired, HCV RNA testing can be performed after 4 to 6 weeks. All positive tests should be confirmed by supplemental testing using the recombinant immunoblot assay, for example. Immunoglobulin administration is not recommended because of the absence of neutralizing antibody and the failure of immunoglobulin to prevent transmission of HCV. [315], [318] Antiviral agents are not recommended for postexposure management. Counseling should be provided to exposed people.

TREATMENT

Treatment of acute viral hepatitis is largely supportive and directed to ensuring adequate nutrition and hydration and to monitoring patients for the development of fulminant hepatitis. Drug therapy is rarely indicated for acute viral hepatitis A and E because 90% to 95% of patients are expected to recover without sequelae. Hepatitis B and D can progress to chronic infection, but given the current response to antiviral therapy, none is indicated. Based on the high response rate to interferon therapy, it may be reasonable to treat patients with clearly documented acute HCV. [326]

General Recommendations

Most patients with acute hepatitis can be managed outside of the hospital setting as long as they can maintain adequate hydration and intake of calories. The old therapy of bed rest is no longer advocated unless the patient experiences severe fatigue. [327] No specific dietary measure or supplement has been shown to be effective. Protein should be restricted only in patients exhibiting clinical signs of hepatic encephalopathy. During the convalescent phase, a high-protein diet may be necessary to aid in recovery. Alcohol should be avoided, and use of medications should be kept to a minimum. Drugs that are metabolized by the liver, such as benzodiazepines, should be avoided or dose reduced if absolutely essential. Patients should be assessed weekly during the early phase of the illness and followed through recovery. They should be monitored for symptoms and signs of encephalopthy, such as increased somnolence, drowsiness, and asterixis and, if present, assessed with serial trail tests. Monitoring of the serum prothrombin time is a useful marker for assessing hepatic decompensation and for deciding when to refer to a liver transplantation unit. The monitoring of serum aminotransferase levels is not helpful for assessing hepatic function because levels may return to normal in fulminant hepatic failure after massive destruction of hepatocytes. Antiemetics can be used to improve symptoms of nausea. Patients who exhibit clinical features of fulminant hepatitis should be rapidly referred to a liver transplantation unit for management. OLT may be a life-saving procedure for patients who decompensate after acute hepatitis (see Chapter 48 on fulminant hepatitis). It is not necessary to isolate patients with acute hepatitis. People caring for patients with acute HAV and HEV infection should wash their hands with soap and water. Close contacts of people with acute HBV infection should receive hepatitis B vaccine.

Specific Therapy

The rationale for antiviral therapy in acute viral hepatitis is to prevent fulminant hepatitis and progression to chronic hepatitis. The indications for antiviral therapy have not been clearly delineated. Interferon-a was compared with placebo in retrospective fashion for the treatment of acute hepatitis B. A higher rate of clearance of HBsAg and HBeAg and loss of HBV DNA were observed in the group that received interferon-a, 80% versus 54%. Therapy was well tolerated, but the concern remains that interferon therapy may worsen the hepatitis. To date, there have been no randomized, prospective trials evaluating interferon therapy for acute hepatitis, B and use of this agent should be on a case-by-case basis using sound clinical judgment. Lamivudine, a nucleoside analogue, was shown to be effective in treatment of chronic HBV infection and may hold promise as a therapeutic agent for acute infection. This is based on the benefit in a single case of fulminant hepatitis B, and reports of success in treating reactivation of hepatitis B after chemotherapy and acute hepatitis B after OLT. [328], [329] and [330] Studies on therapy of acute delta hepatitis are not available. Until further data are available, specific antiviral therapy is not recommended for acute HBV or HDV infection. Lamivudine should be used to prevent reactivation in patients receiving chemotherapy or high-dose immunosuppressive therapy.

Therapy for acute hepatitis C is controversial. The rate of chronic hepatitis C ranges from 50% to 85%, and therapy during the chronic phase leads to sustained viral clearance in about 50% of cases. [331] This is the rationale for treatment of acute cases of hepatitis C but must be weighed against the risks of therapy. Small published studies indicate that treatment of acute hepatitis C using interferon monotherapy reduces severity of the acute hepatitis and reduces the rate of chronic infection. [332], [333], [334] and [335] The sustained loss of HCV RNA ranged from 39% to 64%, compared with 0% to 20% in untreated controls. A metaanalysis of nine controlled trials assessing the efficacy of a short-course (3 months) of low-dose interferon (3 MU three times per week) for acute hepatitis C showed that therapy was associated with a higher rate of normal ALT levels and clearance of virus at the end of treatment. [336] Long-term follow-up is not available from any of the published series. The argument against treatment of acute hepatitis C is based on the fact that a substantial proportion of acute cases, 25% to 30% resolve spontaneously and that treatment is problematic. It remains to be proved whether antiviral therapy initiated during the acute stage is superior to treatment started shortly after the infection becomes chronic. Based on a recent report showing a high response rate to interferon-a in acute hepatitis C, it may be reasonable to treat bona fide cases of acute HCV. [326]

OTHER VIRUSES

Other nonhepatotrophic viruses can cause hepatitis as part of a generalized systemic infection. Members of the Herpesviridae family are covered here because of their worldwide prevalence and their special role in bone marrow transplant recipients, patients receiving chemotherapy, and post-OLT patients. The identification of other putative hepatotrophic viruses and their role in acute hepatitis are briefly discussed.

Herpes Simplex Virus Types 1 and 2

Infection with herpes simplex virus (HSV) occurs worldwide and affects people of all ages. [337] The clinical manifestations of primary disease depend on the age and immune status of the individual. Primary infection in children tends to be a mild and subclinical illness in almost all cases. Severe infections occur more frequently in infants and immunocompromised patients. HSV 1 classically causes infection above and HSV 2 below the waist. The clinical manifestations of primary disease include vesicular and ulcerative gingivostomatitis and anogenital ulceration. [337] Hepatitis due to HSV 1 and 2 infection tends to occur in the setting of generalized primary infection or reactivation. HSV hepatitis is not a major cause or non-A to non-E hepatitis. [338] Rates of fulminant hepatitis and mortality are high in infants, patients receiving chemotherapy, bone marrow transplant recipients, and women during the third trimester of pregnancy. [339], [340] and [341]

Fever and abdominal pain are the major symptoms of hepatic involvement, and patients are frequently appear ill. The characteristic mucocutaneous lesions usually seen in primary infection are present in less than one third of cases; therefore, a high index of suspicion is required to make the diagnosis. Hepatomegaly may be present on clinical examination. There is often discordance between the aminotransferase levels and serum bilirubin level, with marked elevations in

aminotransferases but minimal jaundice. There may be an associated leucopenia, thrombocytopenia, and coagulopathy. Liver biopsy shows nonspecific hepatocellular necrosis and a mild, scattered inflammatory infiltrate. Multinucleated hepatocytes and nuclear inclusion bodies are specific for the disease.

The diagnosis depends on positive viral cultures from body fluids and PCR detection of viral DNA from serum. Serology is not helpful because the prevalence of antibody is high in the general population. A fourfold rise in IgG titer between acute and convalescent titer is diagnostic but is not practical in acutely ill patients. Immunostaining or PCR analysis of liver tissue is rapid and specific for the diagnosis. When the diagnosis is suspected in acutely ill patients, liver biopsy for culture and immunostaining is recommended for rapid diagnosis. [342]

There have been no clinical trials evaluating therapy for acute HSV hepatitis. Acyclovir, 5 mg/kg, is the treatment of choice based on the success of numerous case reports. [343] Therapy should be initiated promptly while awaiting diagnostic tests because of the high mortality rate. Successful resolution has been reported with vidaribine, and it may be used as an alternative agent to acyclovir. OLT may be a life-saving procedure. [344]

Epstein-Barr Virus

EBV occurs worldwide. Most cases in childhood are asymptomatic, whereas in adults, the typical symptoms of infectious mononucleosis are present: fever, malaise, headache, and cervical and axillary lymphadenopathy. The virus is lymphotrophic, and after initial infection and replication in epithelial cells of the pharynx, infects B lymphocytes. The liver is frequently involved in primary infection, but in contrast to HSV 1 and 2 infection, the hepatitis is usually mild and subclinical. Severe hepatitis has been described in immunosuppressed individuals but is uncommon. Most severe cases are due to reactivation of latent infection.

The clinical features of EBV-related hepatitis in adolescents and adults include fever, malaise, pharyngitis, abdominal pain, and mild adenopathy. The aminotransferase level is mildly elevated and peaks during the second to fourth week of infection. [345] No pattern is specific for EBV. Occasionally, the serum alkaline phosphatase level may be markedly elevated, greater than 15 times the upper limit of normal. A dissociation between the serum alkaline phosphatase and bilirubin, with an elevated alkaline phosphatase and normal serum bilirubin, has been observed. Jaundice is present in about 10% of cases and is usually mild (bilirubin less than 5 mg/dL). The coexistence of a hemolytic anemia may lead to higher than usual bilirubin levels. Most cases resolve in a few weeks, but aminotransferase levels may remain elevated for several months. Splenomegaly is frequently detected in primary infection (50%) and hepatosplenomegaly in 25% of cases. In children, a common presentation is fever, hepatomegaly, and disseminated intravascular coagulation. EBV hepatitis has been described after OLT but is infrequent and not a major cause of graft loss. [346] It may be associated with use of antilymphocyte preparations in the OLT setting. Chronic EBV hepatitis has not been reported. Cases of fulminant hepatitis occur but are rare. [347]

For practical reasons, the virus is not cultured in diagnostic laboratories, and primary infection is diagnosed by demonstration of nonspecific heterophile antibody using Paul-Bunnell or slide agglutination reactions (monospot test). Heterophile antibodies may only be detected after the second week of infection and may be present only for a short period. This test has been replaced by the monospot test, which is quite specific and sensitive for EBV. If more specific and sensitive tests are required, immunofluorescent tests for detection of specific antibodies to EBV proteins are available. Anti–viral capsid antigen IgM and IgG antibodies are usually present at the time of onset of clinical symptoms. EBV DNA can be detected in blood but is primarily a research tool. The liver biopsy shows a mononuclear cell infiltrate involving the portal area and sinusoids with minimal hepatocyte necrosis and Kupffer cell proliferation. The liver biopsy findings may be nonspecific. To aid histological diagnosis, liver tissue can be stained for EBV nuclear antigen 1 or probed for EBV-encoded small nuclear RNA by in situ hybridization. [348], [349]

No controlled trials have been done to assess treatment of EBV hepatitis because of the mild nature of the hepatitis. Successful outcome of severe cases has been reported with acyclovir. Based on these case reports, acyclovir should be tried in severe cases of EBV hepatitis. OLT can be life-saving when medical management fails. In the post-OLT setting, EBV hepatitis should be managed with reduction in immunosuppressive medication and judicious use of intravenous immunoglobulin. [350]

Cytomegalovirus

CMV infection occurs worldwide. In cross-sectional studies, more than 50% of the population exhibited past evidence of infection with CMV. [351] Most infections are asymptomatic, especially in children. Routes of transmission are intrauterine, perinatal, and sexual through contact with blood, genital fluid, or saliva.

In infants, CMV infection can cause a neonatal hepatitis syndrome characterized by jaundice, hepatomegaly, hemolytic anemia, and thrombocytopenic purpura with mild elevations in aminotransferase level. The diagnosis may sometimes be confused with extrahepatic biliary atresia, and indeed CMV infection has been proposed as a causative agent. [352] Most cases resolve without long-term sequelae.

Primary infection in adults presents with symptoms similar to those of infectious mononucleosis. The liver is commonly involved in primary infection, but the hepatitis tends to be mild; jaundice is rarely present. [353] Patients at risk for severe hepatitis include neonates, bone marrow and liver transplant recipients, and those receiving chemotherapy. In severe cases, the level of aminotransferases may mimic acute hepatitis. Distinguishing features are prolonged fever and atypical mononuclear cells on peripheral smear.

The liver biopsy usually shows multinucleated giant cells in neonatal hepatitis. Intranuclear inclusions are observed in hepatocytes, Kupffer cells, and biliary epithelial cells. Hepatic lobular microabscesses are reported in liver transplant recipients. [354]

The presence of atypical mononuclear cells on peripheral blood smear may suggest the diagnosis. More definitive diagnosis rests on recovering the virus from infected body fluids. The shell vial assay, which uses a combination of viral culture and immunologic detection of immediate to early gene expression, is the test of choice. IgM is sensitive for the detection of CMV macroglobulins but does not reliably indicate acute infection. Presence of IgG antibody is not specific for acute infection. A fourfold rise in IgG antibody between acute and convalescent sera is diagnostic but is seen in less than 50% of cases. Liver tissue can be stained with monoclonal antibody against CMV early and late antigen, and PCR techniques can yield a faster diagnosis than routine culture. [355], [356]

There are no controlled trials of therapy for CMV hepatitis. There are several case reports citing the benefits of gancyclovir for severe hepatitis. Patients with normal immunity generally have a mild course and do not require treatment. Treatment should be reserved for immunocompromised patients because of associated toxicity.

Recently Discovered Viruses

Studies indicate that in 10% of cases of transfusion-related hepatitis, 50% of cases of fulminant hepatitis, and 20% of community hepatitis, no cause can be identified. [357] This has led to the search for other hepatotrophic viruses. Using molecular techniques, this search began in earnest, and three candidate viruses have been discovered. Hepatitis G virus (HGV) or GB virus C (GBV-C), basically the same virus, was the first described, TT virus (TTV) (named after the initials of the patient from whom it was discovered) was the second, and SEN virus (SENV) the third.

Hepatitis G Virus HGV is a member of the *Flaviviridae* family and is distantly related to hepatitis C. Based on blood donor studies, the virus appears to have worldwide prevalence, with a rate of viremia of 1% to 2%. It can be transmitted by blood transfusion, and other modes of transmission, such as the percutaneous route, may be possible. It appears to have a high rate of persistence (91%). Initial studies suggested this was a causative agent of non-A to non-E hepatitis, but this has not been substantiated in other studies. [358], [359] Rates of HGV infection appear to be similar in transfusion recipients who developed hepatitis and those in whom hepatitis was not seen. [358] Additionally, most cases were not associated with a rise in aminotransferases. HGV may not even be hepatotrophic because inoculation studies in a chimpanzee failed to demonstrate liver disease. Based on these studies, it has been concluded that HGV is not a cause of acute or chronic hepatitis.
TT Virus TTV was initially discovered in 1997 from the serum of a patient with non-A to non-G hepatitis. [360] TTV is a single-stranded DNA virus belonging to the *Circovirus* family. Studies of multitransfused individuals and blood donors suggest a worldwide prevalence, with rates of viremia ranging from 5% to 90%. [360], [361] and [362] Initial studies from Japan indicated that the virus was related to hepatitis in three of five patients after blood transfusion, suggesting that it was the cause for non-A to non-E hepatitis. [360] However, a study of blood donors and prospectively followed transfusion recipients with and without hepatitis indicated a similar rate of newly acquired TTV infection in patients with (23%) and without (22%) hepatitis. [362] Transmission using serum and stool from two infants with acute TTV infection was observed in chimpanzees, but clinical and histological evidence of hepatitis was not demonstrated. [363] These data support the conclusion that TTV is not a hepatotrophic virus.
SEN Virus SENV is the most recently identified candidate virus as a potential cause for non-A to non-E hepatitis. SENV is a single-stranded DNA virus probably

belonging to the *Circovirus* family and distantly related to TTV. Five strains have been identified (A, B, H, D, and E), but only strains SENV-D and SENV-H had low enough rates in donor populations and high rates in transfusion-related hepatitis to qualify as potential agents of acute hepatitis. Preliminary data supporting SENV as an infectious hepatitis agent included a high prevalence of SENV in injection drug users and multitransfused patients, and the prevalence of SENV was high in cases of non-A to non-E hepatitis but low in control populations. [364] In a prospective study of transfusion-related hepatitis, there was a strong association between SENV with transfusion-associated non-A to non-E hepatitis (92%), compared with transfused controls who did not develop hepatitis (24%). [365] Persistent viremia was demonstrated for up to 12 years, and chronic elevation of aminotransferases was observed, but chronic hepatitis was not documented by liver biopsy. However, most patients with SENV infection did not have hepatitis, and replication within the liver has not been established. [365] These data appear to indicate that SENV-D and SENV-H may have a causative role in non-A to non-E hepatitis. In summary, HGV and TTV are not causative agents of non-A to non-E hepatitis. They can be transmitted by blood transfusion, but it is debatable whether they cause liver disease after infection or are hepatotrophic at all. It is possible they may cause liver disease in certain settings, as in immunocompromised patients infected with CMV and EBV, but this remains to be proved. They do not appear to worsen underlying viral or nonviral liver disease. SENV, another transfusion-transmitted virus, may lead to persistent infection, but more definitive studies are awaited before it can be added to the hepatitis alphabet.

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CHAPTER 107

Robert G. Gish and Stephen Locarnini

CHRONIC HEPATITIS B VIRAL INFECTION

There are estimated to be more than 400 million carriers of hepatitis B virus (HBV) worldwide. [1] The prevalence of chronic HBV varies. In the Western world, HBV is usually an adult acquired disease with a prevalence ranging from 0.2% to 1% of the population and is responsible for less than 10% of chronic liver disease. In Asia, Africa, and the Middle East, chronic HBV is acquired at birth or early in life, has a prevalence from 5% to 20%, and is among the leading causes of death in those regions. At least 20% to 30% of those carriers die from complications of chronic liver disease, including cirrhosis and liver cancer. [2], [3] The World Health Organization places HBV in the top 10 causes of death worldwide. [4] The estimated viral burden in the United States is 1.5 million people, with the disease concentrated in ethnic subgroups and populations with high-risk behavior and health care costs to the United States economy that exceed $350 million per year. Although the United States is considered an area of low prevalence for hepatitis B infection, the incidence of new cases, the prevalence of carriers, and the burden of acute and chronic disease maintains hepatitis B among the important communicable diseases. It was estimated that more than 430,000 new cases of hepatitis B infection occurred each year in the United States in the 1980s, compared with a much lower estimate of 185,000 per year in 1997. [5], [6] These acute infections lead to 27,000 to 42,000 chronic carriers, 17,000 hospitalizations, and ultimately 4000 to 5500 deaths per year from cirrhosis and primary liver cancer as well as the morbidity and cost of liver transplantation in about 350 patients per year. [7], [8] and [9] The risk for chronic HBV infection after acute exposure ranges from 1% to 5% for adults and greater than 90% for infants born to infected mothers.

Understanding the prevalence of HBV, patients at risk, clinical presentation, serologic (Table 107-1) and molecular tests, as well the current standards of monitoring and treatment is an essential part of all clinical physicians' practices. Advances in molecular biology and virology, as well as developments in antiviral research, have led to much greater understanding of this virus, allowing implementation of new therapies and more appropriate testing. Sharing of knowledge between investigators and clinicians worldwide will continue to advance our understanding of this extensive health problem.

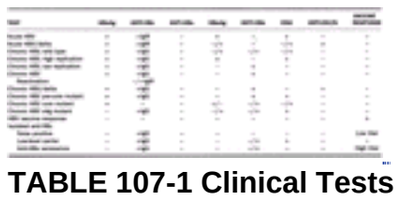

TABLE 107-1 Clinical Tests Used for Hepatitis B Virus

The serious consequences of end-stage liver disease and liver cancer confront patients and physicians throughout the world. Communicating the high rate of severe consequences of this disease is essential to encourage patients to participate in monitoring and follow-up. [10], [11], [12], [13] and [14] Testing patients at risk for HBV, interpreting the serologic tests available, distributing patient education, and implementing regular follow-up are all-important steps in the management of infected patients. Vaccination, the key step in eliminating this global scourge, must be applied to all infants, [15] household contacts of HBV-infected individuals, health care workers, and adolescents and individuals who are sexually active with more than one sexual partner. [16], [17] and [18] Vaccination of all newborns has been calculated to be cost-effective in the developed countries [19], [20] and may become cost-effective in developing countries, depending on the cost of vaccine and methods of administration. Regularly evaluating patients and counseling them about the monitoring process is very important because of the revolution in antiviral therapy taking place today for patients who are infected with HBV.

TERMINOLOGY AND ASSAYS

Terminology and testing for HBV have confounded many students of medicine since the discovery of HBV [19], [20], [21] and [22] because of confusing and misleading nomenclature. Inconsistent and inaccurate terms, such as the oxymoron "healthy carrier," must be removed from our terminology. These outdated terms, often used to reassure patients or physicians, do not represent the true clinical outcome of any patient group and also cannot be applied to any individual with any level of confidence.

The liver biopsy remains a very important step in evaluating patients with chronic HBV infections but is not required for all patients. The outpatient liver biopsy is safe, with a risk for serious complications being less than 1 in 2000 and the risk for death being less than 1 in 10,000. [23], [24] The liver biopsy determines the *stage* of disease (fibrosis) and the extent or level of inflammation (*grade*) (see Chapter 143). This determination then allows the patient and physician to decide on the necessity and urgency of treatment and the need for and frequency of liver cancer screening as well as monitoring of viral replication and liver panels. With the arrival of new antiviral medications aimed at inhibiting HBV replication, increasing rates of resistance to medications have been observed. The clinical significance of these resistant viruses is still being evaluated. Because combination therapy is currently in development, awaiting new information about treatment is reasonable as part of a patient treatment decision for some patients. Conversely, patients with advancing liver disease or impending liver failure usually require immediate treatment with our current best therapy, such as lamivudine.

DEFINITIONS

Chronic Hepatitis B Virus Disease

Chronic HBV disease is defined as hepatic necroinflammation resulting from the presence of HBV. These patients are positive both for hepatitis B surface antigen (HBsAg) and for HBV DNA with serum levels of HBV greater than 100,000 copies per milliliter or the presence of hepatitis B core antigen (HBcAg) staining in the liver. Liver enzyme levels are either persistently elevated or intermittently elevated over time periods of 6 months or longer. The World Health Organization case definition of chronic disease requires the detection in serum of HBsAg on two occasions over a 6-month period. Liver biopsy is an important component in defining the severity of liver disease in these patients. Abnormal liver enzyme levels are defined as an alanine aminotransferase (ALT) of more than 27 IU/mL in women and 30 IU/mL in men when individuals have a normal body mass index. [25] Patients with chronic HBV disease can be divided into two subgroups: hepatitis B e antigen (HBeAg) positive and negative, which will be discussed further in the section on HBV variants. Patients with normal ALT levels but high levels of HBV DNA and necroinflammation on liver biopsy are also included in the group owing to similarity in natural history. [10], [12], [13] and [14] Persistent HBV DNA levels detected over the first 1 to 4 weeks after acute hepatitis B disease indicate a strong likelihood of not clearing HBV infection, and consideration should be given to intervention with antiviral therapy. [26] "Flares" of hepatitis B infection occur in some patients and are defined as intermittent elevations of serum aminotransferase levels to more than 10 times the upper limit of normal. These events occur in 10% to 15% of patients each year and may be asymptomatic and undetected.

Inactive Carrier

Inactive HBsAg carrier state is defined as the presence of HBsAg in serum, no detectable HBeAg, low levels of HBV DNA (less than 100,000 copies per milliliter), and persistently normal ALT levels for periods of longer than 6 months. A liver biopsy can be performed in such patients but is not a usual practice, and the hepatitic activity index is typically less than 3 on a 1 to 22 scale. (See Chapter 143 on liver biopsy and histology scoring systems.) These patients are often described as immunotolerant and remain at risk for HBV DNA integration into hepatocyte nuclear DNA with the attendant risk for liver cancer at a later point in life.

Reactivation

Reactivation of hepatitis B disease occurs when a person is in the inactive HBsAg carrier state and develops an elevation of liver enzyme levels and a rise in HBV viral DNA levels. The HBeAg can become positive in such patients. The liver biopsy typically has an hepatic activity index score of 4 or greater. [27]

Resolved Infection

Resolved hepatitis B infection is defined as serologic tests showing HBsAg negative and HBV DNA negative in a person with a history of acute or chronic HBV infection and currently normal levels of liver enzymes. These patients may remain anti-HBc positive for 5 to 10 years or longer and are at risk for transmitting disease on rare occasions (such as the donation of solid organ tissue) or of reactivating HBV disease if treated with immunosuppressive medications. In addition, those patients who appear to have resolved HBV infection may develop active liver disease if they are treated with immunosuppressive medications such as prednisone, methotrexate, or chemotherapy.

Co-infection

Co-infection with hepatitis delta virus (HDV), hepatitis C virus (HCV), and human immunodeficiency virus (HIV) is defined as the presence of active viral replication with one or more of the previously defined viruses by molecular assays. These infectious problems are discussed in detail later.

SEROLOGIC DIAGNOSIS

As a general principle of laboratory-based investigation for the virologic cause of chronic liver disease, testing should include all the major etiologic agents (HBV, HBV plus HDV, HCV) [28] because these are the common infections in high-risk groups such as injecting drug users, multiply transfused patients, and those who are sexually active with different partners. The principal screening assay for acute and chronic hepatitis B, as well as the screening of blood and organ donors, detects HBsAg in serum. [29]

The typical serologic profiles found in current or historical HBV infection are shown in Table 107-1. Hepatitis B core-specific immunoglobulin G (IgG) antibody (anti-HBc IgG) is almost always present in the serum of patients with chronic hepatitis B and may play a role in immunomodulation of fetal serologic responses to HBV after perinatal transmission from HBeAg-positive mother to her neonate. These antibodies cross the placenta in utero and thus the newly infected neonate has high titers of maternal anti-HBc IgG at birth (indicating a strong helper T-cell type 2 (Th 2 response), partly contributing to its "tolerized" state to the HBV. These antibodies are not neutralizing and do not appear to influence viral replication. The detection of anti-HBc IgM is generally diagnostic of acute HBV infection. [30]

Most patients with underlying inflammatory activity in the liver demonstrate one or some markers of virus infection. Typically, HBV DNA is detected and measurable by molecular techniques [31], [32] (Table 107-2). Relative levels of HBV DNA often correlate inversely with the degree of necroinflammatory activity in the liver, reflecting attempts by the host's immune response to control and eliminate the virus.

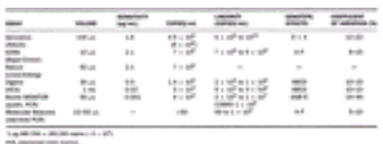

TABLE 107-2 Hepatitis B Virus DNA Viral Load Measurement

HBV has a number of genetic variants, described as genotypes A, B, C, D, E, F, and G. Genotyping is specified by sequencing the HBV genome or by using serologically based antibody testing. Understanding the clinical significance of HBV genotypes is evolving (Table 107-3). Genotype assessment and nucleic acid sequence analysis may allow epidemiologists to track the movement of HBV infections through the world or regions of the world and establish other epidemiologic behavior of HBV infection, including the investigation of common source outbreaks. [33]

CHARACTERISTIC	HEPATITIS B GENOTYPES		
Region	Europe, United States	Asia	Africa
Genotypes	A, D, F, G	B, C	A, D, E
Subject	Adult	Infant	Children
Transmission	Horizontal	Vertical	Vertical
Carrier	20%	30%	80%
Anti-HBe positive and increased ALT	Yes	Yes	Unknown
Response to interferon	30% (G–A)	5%	Unknown
HCC	Low	High	High
Ethnic group	White	Asian	African

ALT, alanine aminotransferase; HCC, hepatocellular carcinoma.

TABLE 107-3 Clinical Characteristics of Hepatitis B Genotypes

Depending on the predominant HBV genotype in the particular geographic setting, patients infected with genotypes other than A often present to the liver clinic seropositive for anti-HBe rather than HBeAg. These patients are infected with the precore G1896 stop codon precore mutation (“precore mutant”). Genotype A HBV infected patients still tend to present to the clinic seropositive for HBeAg.

Prevalence

Regions of high, intermediate, and low endemicity have been identified. High levels of endemicity (positive HBsAg) (at least 8% of population infected) represent more than 45% of global population, intermediate rates of infection (2% to 7%) represent another 43% of global population, and low prevalence (less than 2%) represents the final 12% of global population. Five percent of the U.S. population demonstrates exposure or past infection with HBV. About 1 of 200 people in the United States is currently infected with HBV. Some ethnic subgroups in the United States have up to 7% to 10% prevalence rates that closely approximate their country of origin. HDV infection and its prevalence are discussed later.

Transmission

Modes of transmission vary depending on the country of origin; ethnic background; local, traditional, and Western medicine practices; and most importantly, in developed countries, risk behavior. Understanding each of the major routes of transmission is the key step in preventing HBV transmission and can lead to the eradication of disease through education, prevention, and vaccination. [21], [34] In populations that acquired HBV as adults, heterosexual contact (42%), men having sex with men (15%), and injection drug use (21%) represent the most common modes of acute transmission and may well represent the most common causes of chronic disease.

Vertical transmission is believed to be the leading cause of HBV transmission worldwide. This pathway, as a major cause of HBV transmission, has recently been brought into question with identification of transmission through iatrogenic routes and the application of folk medicine in young populations. Even taking these newly identified risks into account, vertical (predominantly perinatal) transmission probably accounts for more than 50% of all childhood-acquired disease. HBV is believed to infect most commonly neonates at the time of delivery. There has also been a proposal that HBeAg traversing the placenta during pregnancy may induce tolerance to HBV that infects the child during the birthing process, resulting in chronic infection that may persist lifelong in many patients.

Nosocomial and occupational exposure and transmission are also an important issue. Health care workers exposed to patients who carry HBV and patients exposed to medical practitioners who carry HBV infection are both at risk. [35], [36] and [37] Renal dialysis patients are, historically, considered at high risk for acquiring or transmitting HBV because many had extensive blood exposure, and further exposure to other patients took place in the dialysis setting. [35], [36], [37] and [38] Some states require, by law, that practitioners must remove themselves from practice if they perceive that they pose a risk (in this case infectious risk) to their patients. Health care workers that are exposed or potentially exposed to needle-stick injuries or blood-splash exposure are also at risk for acquiring HBV. [39] The most important step is vaccination as a preventative step before risk exposure, immediate testing for immunity to hepatitis B if exposure takes place, and application of hepatitis B immunoglobulin (HBIG) and HBV vaccine if a health care worker who is not immune is exposed to blood or sharps. [35], [36] and [37], [40] Hepatitis B vaccination is strongly advised for all individuals who work in a health care setting that may expose them to blood or body fluids. Elimination of reused needles in developing countries may lead to a marked reduction in childhood, adolescent, and adult-acquired HBV infections. [41]

Household transmission has been clearly identified as a risk for acquiring HBV. [7] The route of exposure is not clear, but HBV is a DNA virus that can exist outside the body on fomites for prolonged periods. Avoiding the sharing of household utensils that may carry blood, such as toothbrushes and razor blades, is also important. The transmission of HBV with common kitchen items and food sharing has not been described. All household members living with an HBV carrier must be tested for HBV and vaccinated if not immune. Recent studies have even suggested bed bugs as a source of HBV transmission. [42], [43]

HBV is a sexually transmitted disease. [7], [44], [45] Individuals attending clinics treating sexually transmitted disease commonly have evidence of exposure to HBV as indicated by the presence of anti-HBs and anti-HBc, and a subset of patients also have HBsAg in their serum. Many of these patients had asymptomatic disease, although some patients present with acute HBV after sexual exposure.

Blood transfusion is a very unlikely source of HBV in Western countries, where effective screening of blood takes place. Screening of all blood units for HBV as well as pre-donation screening for risk behavior should be standard throughout the world. The risk for acquiring HBV through a blood transfusion in the United States is less than 1:400,000 units. The current test used to screen blood for HBV infection is the anti-HBc assay. [21], [46] The reason any risk is present from blood products relates to the fact that some patients donate blood before the onset of an immune response to HBV infection or clinical evidence of hepatitis. Genomic testing is emerging as a method to make the blood supply even safer. [47] A number of developing countries do not screen blood or blood products for HBV, posing a major risk to patients who may undergo blood product transfusion in those countries.

About 20% to 35% of individuals with adult acquired HBV infection in the United States have no known risk factor or easily identifiable risk factor. [7] Possible transmission through sexual contact, medical or dental treatment, sharing household utensils, and blood exposure in an occupational etting must be considered.

Co-infection

In patients who are infected with HBV, co-infection with HCV, [48] HIV, or both exists in certain risk groups, especially those with adult high-risk behavior. All patients with HBV infection must be tested for HCV infection, and if a risk history of sexual exposure, injecting drug use, or intravenous drug abuse is discovered, HIV infection testing is necessary. HIV infection clearly influences the response to HBV vaccine. HDV co-infection is also an important medical issue. HDV infection is specific also to certain world regions, such as Amerindians in South America and high-risk populations in the Mediterranean area.

Prevention

Vaccination is the only medical intervention to date that can markedly change the incidence and prevalence of this disease. The list of HBV vaccine products available worldwide is extensive, and individuals should review local product availability in their own country. All current products appear to be very similar in terms of immune response and level of anti-HBs titer after a series of vaccinations. Worldwide vaccination for HBV has finally come of age, with most countries using or planning a broad-based HBV vaccine program. [8], [49], [50] and [51] Vaccination has already resulted in a marked decline in childhood prevalence of HBV disease in Taiwan and more importantly has already demonstrated a major decrease in hepatocellular carcinoma (HCC), even in this young age group. [52], [53] and [54] This is the ultimate in cancer treatment: prevention through vaccination. All individuals who are exposed to blood or blood products, plan travel to developing countries, live in a household or have intimate contact with an HBV carrier, work in a health care setting with patient contact, are born to HBV-positive mothers, or have chronic liver disease should be vaccinated against HBV infection. Safe sexual practices, needle exchange or nonreusable needles, thorough blood screening, and medical instrument sterilization are all essential components of a prevention program.

For children born to HBV-infected mothers, prevention of vertical transmission with injection of HBIG and vaccine or vaccine alone is recommended or considered the standard of care in most parts of the world. [55] HBIG administration to the infant at the time of birth (in combination with vaccination), because of expense, is reserved for use in countries with higher per-person health care expenditures. Vaccine appears to protect more than 80% to 90% of infants when administered at birth (without HBIG) and followed by two additional doses within the next 6 to 12 months. HBIG intramuscular injection, when used with HBV vaccine, has an even higher efficacy, protecting more than 98% of infants. Some special populations have a low to very low response to conventional vaccines, leading to the development of more

immunologically active vaccines or adjuvants to current vaccines to promote an increased response rate. Examples of such populations include older patients, organ transplant recipients, and other immunosuppressed populations, including patients co-infected with HIV and dialysis patients. Hepatitis A vaccination of carriers with HBV is emerging as the standard of care in many countries owing to the high mortality rate for acute HAV infections in chronic HBV carriers as demonstrated in outbreaks such as the Shanghai epidemic. [56]

Postvaccination testing is only for those individuals who need to know their HBV immune status for their medical management or for potential occupational exposure risks. Revaccination results in an antibody response in more than 30% of all people who do not respond to a primary vaccination series. [57] HBV vaccine levels remain above the level perceived to be protective, (10 IU/mL) in abut 50% of patient for time intervals of up to 15 years. It is also clear that patients who do not have measurable antibodies can mount an amnestic response and be protected from HBV infection. [58], [59] and [60] There is no evidence that HBV vaccine is associate with any form of arthritis or neurological sequelae. [61], [62] and [63]

Based on the previously described epidemiologic behavior, the populations to screen for HBV and subsequently to vaccinate if not infected with HBV are those patients who are born in endemic regions of the world, patients with a history of high-risk sexual activity or more than 50 lifetime sexual partners, men having sex with men, any person with a history of injecting drug use, individuals who have or have had sexually transmitted diseases, household contacts of HBV carriers, sexual contacts of HBV carriers, people who have had medical care or injections in a developing country, any person with acutely or chronically elevated liver enzymes, and carriers of HIV or HCV infection.

Children are a special subgroup of patients who need to be considered as carriers if born to a mother who is an HBV carrier and who did not receive HBV vaccination (with or without HBIG) vaccination at birth. Also, all children who came from orphanages in developing countries should be tested for HBsAg. All immigrants from developing countries where there is a significant prevalence of HBV need to be screened for HBsAg as well. Both pediatric and adult travelers to developing countries are at risk for HBV infection and need to be tested if evidence of liver disease is present.

HEPATITIS B VARIANTS AND MUTANTS

Although HBV is a DNA virus, replication is through an RNA-replicative intermediate requiring an active viral reverse transcriptase polymerase enzyme (Fig. 107-1). The reverse transcriptase of HBV lacks a conventional proofreading function, which is found in other higher-order polymerases. Therefore, HBV exhibits a mutation rate more than 10-fold higher than other DNA viruses and more closely resembles retroviruses such as HIV. The number of nucleotide substitutions varies depending on the stage of disease. The natural evolutionary rate for the HBV genome in chronic hepatitis B is about 1.4 to 3.2 × 10^{-5} substitutions per site per year; however, in the liver transplantation setting, it is almost 100-fold higher. This higher substitution rate may also be a result of the immunosuppression regimen associated with transplantation, especially the use of steroids and the positive enhancing effect on the HBV glucocorticoid response element in the viral genome, resulting in further up-regulation of viral replication.

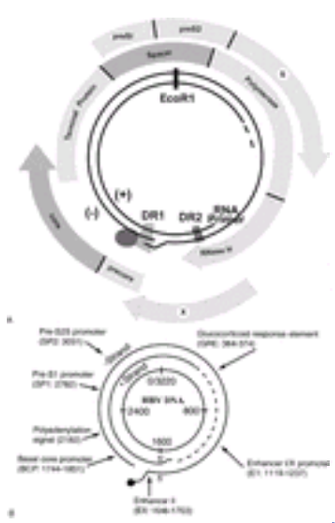

FIGURE 107-1. Molecular structure of HBV virus showing **(A)** the sites of surface, core, and e antigen production, and **(B)** primer sites and areas involved in mutations.

Pre-C and C Gene Mutants

The HBV core gene contains two in-frame start codons that control the synthesis of HBcAg and HBeAg. Both proteins are targets for immune-mediated viral clearance mechanisms. The two major groups of mutations that affect HBeAg synthesis are precore region mutations (G1896A) and mutations in the basal core promoter (BCP) at nucleotides 1762 and 1764, all resulting in diminished production of HBeAg and a resultant increased host immune response. Precore mutations frequently occur temporally related to core gene mutations and deletions. Mutations in the BCP are likely to increase viral replication and enhance disease activity.

X Gene Mutants

The X protein is a nonstructural protein encoded by the *X* gene, which can transactivate a variety of cellular and viral promoters by activating signaling pathways. [64] X has been reported to interact with a number of nuclear proteins involved in cell cycle regulation and DNA repair and transcription. Persistently high levels of the HBx expression have been associated with the development of liver tumors in transgenic mice, and X protein can transform nontumorigenic cell lines in vitro.

Pre-S and S Gene Mutants

The *S* envelope gene contains three in-frame start codons, which divide the gene into the three surface proteins of HBV: The pre-S1 region, the pre-S2 region, and the S region. Three corresponding mRNAs serve for the synthesis of these proteins. The pre-S1 promoter (SPI) is located upstream of the *S* gene, whereas transcription of the pre-S2 and S mRNAs is directed by the S promoter (SPII), which is located within the pre-S1 region. Surface mutations have obvious clinical relevance in the prevention of HBV owing to impact on prophylactic vaccination as well as laboratory-based diagnosis. [19], [20], [65], [66] Several large HBV vaccination programs in endemic regions have revealed a 2% to 3% incidence of vaccine escape mutants resulting from alterations in the HBsAg protein. Generally, the amino acid (aa) substitution of glycine for arginine at aa 145 of the S protein makes this epitope unlikely to bind to antibodies generated to wild-type HBsAg. The “a” determinant of the HBsAg is a peptide sequence located between aa 116 and 160 of the S protein, which represents the major immune target of polyclonal antibody to the HBsAg. This antibody reactivity is directed mainly against the second loop of this determinant, located in aa 139 to 147. In patients infected with HBV exhibiting surface mutations affecting the a determinant, the mutant HBsAg may not be detectable by commonly used HBsAg assays, especially if based on monoclonal antibody “capture” reagents. As a result, these mutants do represent a potential public health problem because patients harboring HBV with these surface mutants remain infectious but do not exhibit readily detectable HBsAg and hence may escape diagnosis, and contacts may not be protected from viral infection. [6], [67], [68], [69] and [70]

Polymerase Gene Mutations

The polymerase open reading frame (ORF) is the longest, covering almost 80% of the whole genome and overlapping the other three ORFs. The HBV polymerase protein mediates encapsidation of the pregenomic RNA into the core particle and synthesizes the HBV DNA genome therein. It has four enzymatic activities: DNA synthesis priming activity, RNA-dependent reverse transcriptase (RT) and DNA-dependent DNA polymerase activity, and RNaseH (RNA degradation) activity.

HBV quasispecies with mutations in the polymerase gene have been detected in patients undergoing antiviral therapy (Table 107-4). These mutations significantly

decrease the in vitro sensitivity of the polymerase to lamivudine-associated chain termination and competition for enzymatic inhibition. Similarly, after famciclovir therapy, polymerase mutations in codon L180M markedly decrease famciclovir efficacy. [71]

FCV MUTATIONS	HBsAg MUTATIONS
H436N/Y	A domain no change
V519L	B domain E164D
P523L	B domain no change
L526M/V	B domain no change
T530S	Not published
V553I	C domain W199 stop/no change
N/S/H584K	D domain after end HBsAg
R588K	D domain after end HBsAg
LMV MUTATIONS	**HBsAg MUTATIONS**
I399S	Not published
F512L	B domain A157D
V519L	B domain E164D
L526M/V	B domain no change
T530S	Not published
A546V	Not published
M550I	C domain W196S, W196L
M550V	C domain I195M
V553I	C domain M198I/W199S
S559T	Not published
S565P	S210R
L575V	Not published
ADV MUTATIONS	
None to date	

TABLE 107-4 Mutations in Hepatitis B Polymerase Protein Associated with Drug Resistance to Lamivudine (LMV) and Famciclovir (FCV) with the Corresponding Change in Hepatitis B Surface Antigen (HBsAg)

Genotypes and Serotypes

HBV has a number of genetic variants described as serotypes (adw, ayw, and adr) and genotypes A, B, C, D, E, F, and G. Genotyping is specified by sequencing the HBV genome or by using serologically based antibody testing. Understanding of the clinical significance of HBV genotypes is evolving (see Table 107-3). Serotyping and genotype assessment may allow epidemiologists to track the movement of HBV infections through the world or regions of the world and establish other epidemiologic behavior of HBV infection and potentially establish specific clinical behaviors to each subgroup.

Depending on the predominant HBV genotype in the particular geographic setting, patients infected with genotypes other than A (B, C, D, and E) often present to the liver clinic seropositive for anti-HBe rather than HBeAg. These patients are infected with the precore G1896 stop codon precore mutation (“precore mutant”). Genotype A HBV-infected patients still tend to present to the clinic seropositive for HBeAg and so have HBeAg-positive chronic hepatitis B (wild-type) infection. The molecular virologic basis for this genotype preference is discussed in more detail below under HBV mutants.

CLINICAL MANIFESTATIONS

Clinical manifestations of chronic HBV infection range from no signs or symptoms of disease (most patients) to the warning signs of end-stage liver disease. [72] The clinical manifestations of end-stage liver disease are the same as those seen in patients with other causes of liver failure. Some patients with HBV develop extrahepatic manifestations of infection, including periarteritis nodosa, leukocytoclastic vasculitis with the increased likelihood of major organ damage through immune complex injury with target organs, including peripheral nerves, kidneys, brain, and gut through immune complex deposition. [73], [74] Examination of the creatinine level in each patient with HBV infection and assessment for proteinuria are important. Peripheral skin rashes can be assessed by punch biopsy to look for signs of vasculitis. Some patients need a visceral and renal angiogram to identify angiographic signs of vasculitis in those organs due to periarteritis nodosa.

Cirrhosis may manifest early and subtly with changes in sensorium, memory loss, and day-night reversal. The signs of sequential liver synthetic test result abnormalities (low albumin, elevated bilirubin, prolonged International Normalized Ratio, low cholesterol, high ammonia) and progression to marked signs of portal hypertension are not peculiar to HBV infection. The presence of portal hypertension is manifested by ascites, varices, thrombocytopenia, leukopenia, and coagulopathy, as seen in patients with cirrhosis of other causes. Acute flares of disease in patients with HBV are more common than conditions such as HCV infection or autoimmune diseases in which the inflammatory process is often “burned out.”

Laboratory tests are an integral part of evaluating patients with chronic HBV infection. It is very important to differentiate liver enzymes from liver function tests, although these two terms are often confused or, unfortunately, combined. Liver enzymes are markers of liver inflammation and do not reflect liver function. The ALT, aspartate aminotransferase (AST), alkaline phosphatase, and gamma-GT are useful markers of liver inflammatory activity only. These tests, if elevated, should lead the clinician to a full laboratory evaluation for HBV infection as well as investigation of other causes of liver disease (fatty liver, herbal medications, alcohol-induced liver disease, [39], [75] over-the-counter and prescription medications, HCV, and hemochromatosis). Elevated liver enzyme levels in a patient who is HBsAg positive provide a compelling indication for liver biopsy and consideration of treatment.

Liver function tests commonly include cholesterol, albumin, International Normalized Ratio, bilirubin, and ammonia. When results of liver function tests become abnormal, there is usually severe liver injury present, which could indicate end-stage liver disease or a flare of hepatitis superimposed on chronic liver disease. A flare, or marked rise in liver enzyme levels, and a decline in liver function occurs as an increased immune response directed at HBV occurs. This flare of disease activity could also represent superimposed infection such as HCV or HDV, worsening HIV disease if HIV infection is present, and potentially progression to end-stage liver disease. When results of liver synthetic tests become abnormal in a patient with chronic HBV, a clinician should strongly consider consulting with a transplantation center if the patient has no contraindications to transplantation. There should also be a strong consideration for immediate initiation of antiviral therapy with a nucleoside or nucleotide analog. In the setting of liver synthetic abnormalities, interferon is relatively contraindicated, unless used by expert hands.

Laboratory surrogates to assess for the severity of liver disease include signs of hypersplenism, such as a low platelet or white blood cell count. Platelet count levels below 100,000 to 150,000 should lead one to a strong suspicion of advanced fibrosis. An elevated alpha-fetoprotein (AFP) level is usually only found in advancing liver disease or much more rarely in the presence of liver cancer. The degree of AFP elevation provides information regarding the likely presence of hepatocellular carcinoma. A high ferritin and iron saturation could make one suspicious for hemochromatosis, although abnormalities in biochemical markers of iron overload are also common in patients with marked fibrosis or cirrhosis. A “reversed” AST-to-ALT ratio, in which the AST is higher than the ALT, also supports advanced fibrosis or cirrhosis.

Patients with chronic HBV infection may have a normal to moderately reduced quality of life compared with those without HBV infection. Patients with end-stage liver disease have a markedly reduced quality of life, although this is true for all patients with advanced liver disease and is not specific to HBV infection. Patients who undergo treatment with interferon often have a reduced quality of life during treatment. If the patient clears HBV infection, the patient’s quality of life may improve after treatment to a normal level, and treatment may be not only cost-effective but also cost-saving. [76], [77]

Hepatocellular carcinoma, hepatoma, and primary liver cancer are serious risks for all HBV carriers (see Chapter 118). The estimated lifetime risk for HCC in patients who are chronically HBsAg positive is estimated to be about 20%. This risk is markedly increased in patients with cirrhosis, active liver disease as demonstrated by biopsy, elevated liver enzyme levels, elevated HBV DNA level, co-infection with HCV, a strong history of alcohol abuse, confirmation as a carrier of HBV for more than 50 years, an exposure to aflatoxins, a family history of HCC, and an elevated AFP level. [78], [79], [80] and [81] Issues such as prevention of HCC by vaccine is covered in other sections, and prevention of HCC by use of interferon and new antiviral therapies is discussed later in the section “Treatment.” Patients who are chronic HBV carriers need to be informed of the risk for HCC and need to be considered for screening, depending on resources and available treatments, for HCC. The initiation and frequency of screening can take place according to relative risk for HCC in special groups of patients with HBV, most importantly, in patients with cirrhosis.

DIFFERENTIAL DIAGNOSIS

All patients with elevated levels of liver enzymes need to be screened for fatty liver, medication toxicity, elevated iron, HBV and HCV infection, and alcohol abuse. The

differential diagnosis of chronic HBV infection includes considering acute HBV infection, which usually can be differentiated by the presence of anti-HBc IgM (see Table 107-1). Patients with elevated liver test results and detectable presence of anti-HCV in serum can be evaluated by the use of molecular testing. If HBV DNA is not measurable and HCV RNA is found to be elevated, HCV is considered the dominant disease, and therapy should focus on the HCV infection. Autoimmune disease rarely occurs concomitantly with HBV disease and is also rarely caused by HBV. One must rule out biliary obstruction if the gamma-glutamyl transpeptidase or alkaline phosphatase is higher than the AST or ALT serum levels. A baseline ultrasound is useful in patients with elevated levels of liver enzymes to rule out other diseases, provide indirect information concerning the presence or absence of portal hypertension, and provide reassurance to the patient about the absence of liver cancer. Examination of the liver biopsy for plasma cells and testing for antinuclear antibodies and anti–smooth muscle antibody is important, especially if serum globulin levels are greater than 4 gm/dL. Patients with elevated levels of liver enzymes who are HBsAg positive with very active liver disease need to be assessed for delta hepatitis infection. [82], [83], [84] and [85]

COURSE AND COMPLICATIONS

Each subgroup of patients with HBV infection has a different clinical course. The chronic carrier with consistently elevated liver enzymes is the patient with the highest risk for progression to cirrhosis and the subsequent complications. The next highest risk patient with risk for progression is the patient who presents with flares of liver enzymes and reactivation of liver disease. Each cycle of liver enzyme elevation purportedly is due to immune activation and attempts, often ill fated, to clear HBV infection (Fig. 107-2). Spontaneous seroconversion from HBeAg to anti-HBe occurs in 1% to 10% of chronic hepatitis B carriers per annum, but seroconversion from HBsAg to anti-HBs, with clearance of HBV from the liver, is very uncommon (at or less than 1% per year). Only about 1% of patients with chronic HBV spontaneously clear infection (convert from HBsAg positive to negative) each year. [86] The rate of HBsAg seroconversion is higher in patients with more active liver disease as measured by liver enzymes than in those with persistently normal liver enzymes but is in general less than 15% over any 3-year interval. [11], [12] During these immune "attacks" or flares, there is ongoing hepatocellular damage and commonly progressive fibrosis. Thus, in general, the degree of liver enzyme elevation, as well as the number of flares, often correlates, over time, with the severity of liver injury. In specific patients, testing and evaluation by sequential liver biopsies may be necessary to determine the urgency of treatment. The liver biopsy also allows a medical practitioner to provide a full level of informed consent to patients before starting therapy. Blood testing for progressive liver disease and screening for HCC should also take place more frequently in those patients with established cirrhosis than in patients with mild or no fibrosis.

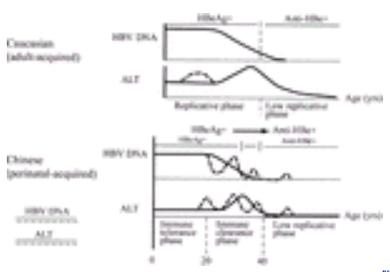

FIGURE 107-2. Natural history of HBV infection. (From ref. [14].)

The chronic carrier with a persistently normal level of liver enzymes (less than 30 IU/mL) is at a much lower risk for progressive liver disease as well as cancer, although the overall risk for HCC is still significant. These patients commonly have minimal liver disease on liver biopsy. The frequency of blood testing of these patients is an important issue because annual testing may be insufficient to document a stable patient or define inactive disease. For such patients, the frequency of screening with blood tests could occur at least biannually, and a more thorough evaluation, testing every 3 months, may be justified.

During regular laboratory testing of liver enzymes, HBV DNA, anti-HBe, and HBeAg testing can determine whether a precore mutant virus has emerged. This viral mutant can be identified by the presence of moderate to high HBV DNA serum levels in the absence of HBeAg. In rare circumstances, patients who appear to clear HBsAg but do not develop anti-HBs may have developed surface mutant viruses. The clinical significance of these mutant viruses is not clear. Long-term studies are in progress to find the specific markers that allow us to identify which patients are at risk for progressive liver disease. As therapy evolves and becomes increasingly complex, the options to treat such patients will expand.

Hepatitis D Virus

The patient with dual HBV and HDV infection has a typical serologic profile of HBsAg and anti-HBe positive (rarely, HBeAg positive) as well as IgG anti–hepatitis delta (anti-HD) seropositivity (see Table 107-1). Specific immunoassays are available for detection of both IgM-(for both acute and active infection) and IgG-specific anti-HD (seen in chronic disease as well as resolved infection). Serum assays for detecting HDAg and HDV RNA are restricted and can typically only be requested from central reference laboratories. HDV infection is easy to miss, and testing should include HDV RNA and anti-HD IgG and IgM as well as markers of HBV infection. These should be repeated in suspected cases.

In the United States, HDV infection occurs predominantly with intravenous drug users in large cities and with high-risk sexual activity. [87] Ethnic populations that may carry HBV from the Mediterranean region or Middle East need to be tested for HDV infection as well. Delta infection leads to more severe liver disease in those patients who acquire HDV and HBV at the same time than in those who acquire acute infection with HBV alone (see Chapter 106 on acute hepatitis). [82], [83], [84] /SUP>and [85] Patients with acute co-infection have a very high rate of viral clearance of both HBV and HDV. When HDV infection is superimposed on chronic HBV infection, the disease commonly accelerates to cirrhosis in a much shorter time. Patients with HDV and HBV co-infection, compared with patients with chronic HBV infection alone, may also be at greater risk for liver cancer, probably because cirrhosis develops as a higher rate. [88]

HDV is a serious disease including both acute infection simultaneously with HBV and superinfection of HDV superimposed on chronic HBV. Acute infection simultaneously with HBV and HDV commonly results in severe disease with a high risk for fulminant liver failure. If the patient survives the acute disease, viral clearance of both HBV and HDV will probably take place. HDV infection can be treated with interferon, although results are discouraging, with patients rarely clearing HDV and HBV long term. Therapy requires at least 1 year of treatment. Nucleoside analog therapy, such as lamivudine, does not suppress HDV infection. Liver transplantation in patients with acute HDV infection is often followed by milder liver disease if HBV and HDV infection persist, compared with HBV graft infection without HDV co-infection. HBIG therapy to prevent graft infection with HBV can also prevent HDV infection of the new graft. [89]

Patients with HDV infection typically have very low levels of HBV DNA owing to the suppressive effect of HDV on HBV viral replication. HBeAg and anti-HBe are variable. HDV co-infection probably results in a higher risk for HCC, although this may be attributable to more active or aggressive liver disease in these patients as opposed to an oncogenic effect of the HDV virus. The key serologic markers include anti-HBc IgM and anti-HD IgM in the acute phase, whereas markers of chronicity (IgG and anti-HBc and anti-HBe) are not detected (see Table 107-1). Thus, the profile is that HBsAg, HBeAg, and HBV DNA become detectable in serum along with HDAg and HDV RNA. Anti-HD IgM becomes detectable soon after onset of illness but is rapidly followed by anti-HD IgG (see Table 107-1). Current assays for anti-HD IgM and IgG tend to detect antibody only transiently, so that the most reliable marker for establishing HDV infection is the detection of HDV RNA by polymerase chain reaction in serum.

Hepatitis D Superinfection in Chronic Hepatitis B

HDV superinfection in chronic hepatitis B is defined as primary virus infection on a preexisting persistent HBV replication. Serologic markers such as titers of HBsAg and HBV DNA can show transient reduction during superinfection. Thus, serial testing is again required. The typical serologic profile is HBsAg positive as well as anti-HD (IgG and IgM) positive, but negative for IgM-specific anti-HBc (see Table 107-1). Again, the diagnosis of persistent infection is established by repeated testing for HDV RNA by polymerase chain reaction. The diagnosis of chronic hepatitis D is readily made when HDAg is detected by immunohistochemistry in the nucleus of hepatocytes of the liver biopsy specimen.

Pathogenesis of Hepatitis D Infection

The manner by which HDV causes liver injury remains controversial. Histological observations of microvesicular steatosis and cytoplasmic eosinophilia in the absence of a mononuclear cell or lymphocytic infiltrate in the liver biopsy specimen of patients with acute HDV infection are consistent with a direct cytotoxic effect of HDV. In vitro molecular studies have supported this view with experimental evidence that the smaller form of HDAg (SHDAg) expression in cells results in the production of pyknotic nuclei and shrunken eosinophilic cytoplasm as well as a reduced cellular nucleic acid synthesis. In contrast, evidence supporting an immune-mediated cytotoxic pathway is sparse, and a direct link between HDV-associated disease expression and the immune response is lacking.

Hepatitis B Interference by Hepatitis D

Suppression of markers of HBV replication during HDV superinfection has been observed in humans as well as in the woodchuck and chimpanzee animal models. In more than 90% of hepatitis B patients superinfected with HDV, seroconversion occurs from an HBeAg-positive to an HBeAg-negative and anti-HBe–positive state. Serum HBV-specific DNA polymerase activity and intrahepatic HBcAg expression can no longer be detected in these patients. On rare occasions, HDV superinfection has been associated with termination of the HBsAg carrier state and seroconversion to anti-HBs. The mechanism for this interference has not been conclusively established.

Hepatitis B Co-infection with Human Immunodeficiency Virus

HIV co-infection with HBV results in more aggressive (progression to cirrhosis and liver failure) liver disease in some patients, although other patients have minimally active liver disease with little or no evidence of progressive liver disease. Why certain patients have progressive liver disease in this setting and others do not is unclear and is not correlated with any specific clinical factor or laboratory test. The discrepancies may be explained by an interaction between the host immune system, helper T-cell subtypes present (Th 1 versus Th 2), host factors that predict fibrosis, and levels of HBV and HIV viral replication. Treatment with lamivudine may slow the progression of liver disease but is problematic from the perspective of increased resistance compared with patients who are not co-infected. [90]

Hepatitis B Co-infection with Hepatitis C

HCV co-infection with HBV results in a higher risk for cirrhosis and a higher risk for liver cancer. Patients often have one dominant viral disease. Treatment focused on the dominant virus, as revealed by blood tests of viral replications, is probably the best management step. [91], [92], [93], [94], [95] and [96]

Acute HAV infection sometimes complicates chronic HBV infection. These patients are at higher risk for severe liver disease. [97] Vaccination for HAV is advocated in all patients with chronic HBV infection. [97]

TREATMENT

Definitions of response to antiviral therapy are very important when evaluating patients who are about to undergo antiviral therapy and for those patients who have been previously treated who appear to need subsequent treatment. Definitions of response are defined as biochemical (liver enzymes), virologic (viral replication), and histological (liver biopsy).

Biochemical response is defined as a fall in liver enzyme levels into the normal range. These events, like the virologic definition given later, can be initial (during therapy), at the end of therapy, or sustained. Sustained response means that the liver enzymes were normal at the end of therapy and at least 6 to 12 months after cessation of therapy. Partial biochemical response means marked fall (greater than 50%) in liver enzyme levels to the near-normal range, reductions typically associated with improvement in histology.

Virologic response means a decrease of HBV DNA to less than 100,000 copies per milliliter when pretreatment levels were greater than 100,000 or by a greater than 2 log reduction from baseline. The reduction in serum HBV DNA viral levels should correlate with HBeAg loss if treatment is given, and finally conversion from anti-HBe negative to positive would complete what is termed *triple seroconversion*. Virologic response can also be termed initial, end of treatment, and sustained if there is loss of viral markers or replication for 6 to 12 months after treatment. HBeAg seroconversion is the terminology used when the HBeAg is lost (negative) long term.

Histological response is defined as an improvement in liver histology on paired liver biopsies. Improvement is usual defined using the histology activity index, the Ishak score or the Metavir score, or one of the modifications. There is no international consensus about which scoring system is best. A significant improvement is based on a decrease in number by one to three points and is usually based on improvement in necroinflammation because the fibrosis score typically lags behind changes in inflammation in the liver.

Complete response means normalization of liver enzymes, virologic response, and loss of HBsAg. HBeAg must be negative, and HBV DNA must be negative by the most sensitive molecular test available, which typically can measure down to 100 copies per milliliter (see Table 107-2). This is thereby synonymous with resolved or cured HBV infection. This term does not require the presence of anti-HBs and does not require a follow-up liver biopsy.

Interferon

The term *interferon* is used to describe a large family of naturally occurring peptides that are included within an even larger group of molecules, termed *cytokines* that modulate immune function. This family of proteins is often involved in the host's control or elimination of acute and chronic viral infections. Interferon directly decreases viral replication by interference with DNA and RNA replication at the genome level. Interferon also indirectly modulates viral replication and viral protein synthesis by activating other immune pathways and modulating the level of other cytokines. The increased synthesis of enzymes, such as 2'5'-oligoadenylate synthetase, further activates the immune system. By enhancing major histocompatibility complex (MHC) antigen expression, interferon increases in the identification of infected cells with HBV and leads to cytolysis and viral clearance.

About 20 natural forms of interferon exist in humans, including interferon-a-2a (produced in lymphocytes), -a-2b, -ß (produced in fibroblasts), -? produced in T cells), and -?. A number of these interferons have been isolated and have been shown to have direct antiviral effects on HBV replication in humans. [98] Interferon is the only medication that has been proved to reduce and eliminate HBV infection in chronically infected patients in randomized controlled trials [99] and is not associated with drug resistance. The patients with HBeAg-positive and HBeAg-negative chronic HBV infection both appear to benefit from treatment, with documented improved outcomes in clinical trials (Fig. 107-3 and Fig. 107-4). Over time, there is an accumulative increase in patients who are HBV DNA and HBeAg negative and a decrease in the number of patients with decompensated liver disease.

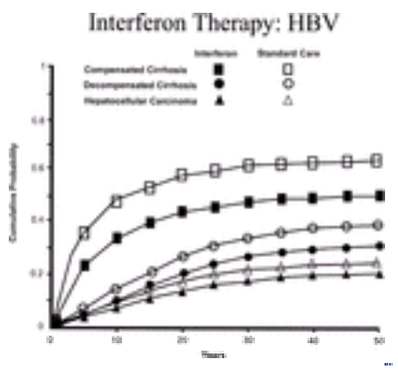

FIGURE 107-3. Short- and intermediate-term outcomes of interferon (IFN) treatment. (From ref. [99].)

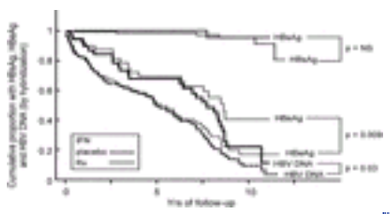

FIGURE 107-4. Long-term outcomes after interferon treatment. (From ref. 57.)

Prednisone has been studied in clinical trials and practice and appears to be beneficial only for certain rare subgroups of patients, such as those with modest viral replication and near-normal levels of liver enzymes. 100 In summary, only an “ideal” subset of patients responds to interferon and clears HBsAg (patients with low levels of liver enzyme elevation). Interferon appears to be efficacious only in patients who have an ongoing native immune response. The presence of elevated levels of liver enzymes and the presence of hepatic inflammation identify these ideal patients who have an inherent immune response to chronic HBV infection. One form of interferon, interferon-a-2b (Intron-A, Schering, Kenilworth, NJ), has been approved for the treatment of chronic HBV infections in many countries throughout the world, including adults and children in the United States. Interferon-a-2b treatment results in HBeAg clearance in 30% to 50% of selected patients and HBsAg clearance in up to 30% of patient with long-term follow-up. 101 Interferon-a-2a (Roferon, Hoffman La Roche Laboratories, Basel, Switzerland) and interferon-a-con-1 (Infergen, Intermune, Brisbane, CA) may also be used to treat HBV infection but have not been approved for this indication by the U.S. Food and Drug Administration. New pegylated forms of interferon-a-2a and -a-2b are undergoing phase III trials to determine whether the longer half-life of this compound imparts any improvement in therapeutic outcome over standard interferon preparations.

The ideal dosing for the different forms of interferon range from 15 to 35 million units (MU) weekly. Interferon is administered by subcutaneous or intramuscular injection three times a week, although some clinical studies have used daily dosing regimens. Prolonged treatment up to 32 weeks may be useful in patients who have an initial reduction in viral levels of HBV DNA without DNA clearance. The side-effect profile of interferon is significant, with a wide variety of side effects that are well described and hopefully will be reduced with newer forms of interferon or with lower doses used in combination with other forms of less toxic therapy. Some examples of interferon side effects (which are often dose related) are bone marrow depression, psychiatric disturbances, alopecia, flu-like syndromes, nausea, weight loss, an increased risk for bacterial infections, and hyperglycemia. The ideal patients who have the best antiviral response to interferon are those with high levels of liver enzymes, moderate levels of serum DNA, and recent or adult-acquired infection ith normal immune response to HBV. Patients who fit this profile may have HBsAg seroconversion that may exceed 30% in long-term follow-up. 101 Other immunomodulators including thymosin-a-1 (Zadaxin, Sciclone Pharmaceuticals, Redwood City, CA) have been shown to be associated with HBeAg seroconversion and triple seroconversion as a single agent and more importantly may be useful in combination with interferon.

Nucleosides and Nucleotides

Modified nucleosides, such as adenosine arabinoside (ara-A) and nucleoside analogs, have been studied during the past few decades. Ara-A was the initial nucleoside analog extensively studied for the treatment of HBV infection. Treatment with ara-A resulted in a decrease in levels of HBV DNA in the serum but had significant untoward side effects, including, in some patients, a permanent peripheral neuropathy.

Toxicity and weak antiviral effects were the major limiting factors behind the failure of ara-A, didanosine (ddI), and similar medications. Fialuridine, a fluorinated nucleoside, has been studied more recently and was found in humans to have major mitochondrial toxicity that resulted in pancreatitis, acidosis, liver failure, and death. Lobucavir was also studied in humans recently, but clinical trials were halted when this agent was associated with gynecologic cancers in animals. These complications support close short- and long-term scrutiny of these powerful medications that can disrupt host cellular homeostasis.

Another important perspective on HBV treatment is the question whether HBV infection is ever cured. HBV exists in a very stable form known as covalently closed circular DNA, or cccDNA, that has a very long intracellular half-life. There are no clearly identified antiviral effects of nucleoside or nucleotide analogs on cccDNA. Clearance of this stable intracellular form of the HBV genome has not been conclusively shown with any antiviral agent.

Currently, adefovir (Hespera®, Gilead, Foster City, CA) lamivudine (3TC, 3-thiacytidine, Epivir, Zeffix, GlaxoSmith-Kline Research Triangle Park, NC) are the only U.S. Food and Drug Administration–approved oral medications that have proven efficacy against HBV, including HBeAg seroconversion and triple seroconversion. Lamivudine is also approved in many foreign countries, including China. Data to date concerning efficacy include a rate of 30% to 35% HBeAg loss at 1 year of therapy. Seventeen percent of patients underwent triple seroconversion, and rare (less than 6%) HBsAg seroconversion to negative has also been reported with most patients remaining HBeAg and HBV DNA negative long term. 18, 102, 103 and 104 (Fig. 107-5). Resistance to lamivudine is very common after 1 year of treatment (greater than 20%) and is associated with mild elevations in levels of liver enzymes compared with the period before emergence of the resistant mutant virus. The presence of the lamivudine-resistant mutant may rise as high as 60% after three years of continuous use of lamivudine. This emergence is associated with more active liver disease as measured by serum levels of liver enzymes. A few patients return to their baseline level of liver disease or viral replication after the emergence of the lamivudine-resistant virus. The lamivudine-resistant mutant virus and other resistant variants are discussed later. This site in the HBV reverse transcriptase or “polymerase” is the most common site of lamivudine resistance, although other sites of mutations associated with resistance have been identified. The rate of viral resistance after 3 years is 50% to 70%. 105 Other nucleosides that confer some level of anti-HBV activity include famciclovir and ganciclovir, but these medications have found little use to date owing to lower levels of efficacy. 106, 107

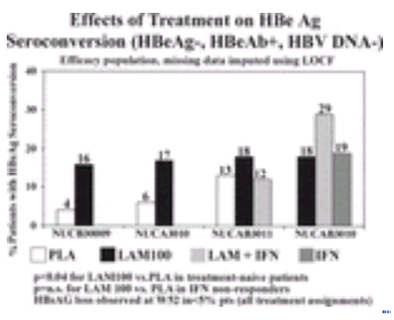

FIGURE 107-5. Outcomes of lamivudine (LAM) treatment for chronic HBV infection. (From refs. 102, 103, 104 and 105, 111.)

Future medications that are in development to treat chronic HBV infection include pegylated interferons, interferons fused to albumin, 5-fluorothiacytidine, tenofovir, entecavir, clevudine L-d4FC, L-ß-thymidine, L-ß-adenine, and L-ß-cytosine, 19, 20, 108 as well as ribozymes and therapeutic vaccines. Other more far-reaching treatments may include dominant negative viral mutants, antisense DNA and RNA, intracellular antibodies, or delivery of current medications through new and advanced deliver methods, such as liposomes or methods that include molecules that bind to specific cellular receptors. Treatment may also take place with DNA vaccines that may provide ongoing internal immune suppression through production of HBV-derived proteins. 42, 109 Liposomes may eventually be used to deliver vaccines such as those originally developed for hepatitis B by oral route or intramuscular injections.

GENOTYPIC RESISTANCE OF HEPATITIS B TO NUCLEOSIDE ANALOGS

To date, the emergence of HBV mutants that show resistance to nucleoside analogs as a result of genetic mutation has been observed only as a consequence of the clinical use of lamivudine or famciclovir. The discussion that follows concerns important amino acid changes in the HBV POL sequence that have been detected in clinical isolates and shown to confer or modify resistance to lamivudine, famciclovir, and other nucleoside analogs.

There has been some confusion over the numbering system used for the HBV POL (see Table 107-4). This inconsistency is the direct result of the heterogeneity between the seven different genotypes of HBV (see Table 107-3). Recently, a standardized numbering system for the HBV POL has been proposed by Stuyver and colleagues, and a consensus is emerging 110 that will be used in this chapter. In this proposal, the HBV reverse transcriptase domain starts with the highly conserved EDWGPCDEHG motif, which includes 344 amino acids and the lamivudine resistance mutations

are designated as HBV POL L180M (previously aa 515, 525, 526, and 528) and HBV POL M204V/I (previously 539, 549, 550, or 552).

Lamivudine Resistance

Initial clinical trials established that treatment of chronic HBV infection with lamivudine was (1) safe and well tolerated, and (2) caused rapid and substantial decreases in viremia. However, decreases in viremia were not stable and rebounded in most cases after treatment stopped. [111], [112], [113] and [114] As long-term trials progressed, it became apparent that during treatment a “rebound” could occur, most of which has been attributed to the development of drug resistance. This resistance was observed at a greater frequency and at an early time point in the transplantation setting [115], [116] and [117] compared with immunocompetent patients. [118], [119], [120] and [121] Lamivudine-resistant populations of HBV were also detected in patients who were co-infected with both HBV and HIV and who were receiving lamivudine as antiretroviral therapy. [122], [123] and [124] Although varying frequencies of resistance and different times preceding the detection of resistance have been reported, the incidence of resistance in immunocompetent patients and immunosuppressed patients clearly correlated with treatment duration. During the first year of lamivudine therapy, resistance developed in 14% to 32% of cases studied, [125], [126] and [127] a frequency that increased to greater than 50% after 2 years. [120]

Sequencing of the HBV POL ORF from drug-resistant isolates revealed mutations in a methionine codon, which caused substitution of isoleucine or valine for methionine 204 (M204V/I) (see Table 107-4). These substitutions affect the catalytic site, changing the conserved YMDD tetrapeptide in motif C from to YIDD or YVDD; these are analogous to substitutions for M184 in the HIV reverse transcriptase, which likewise confers lamivudine resistance. [128] Mutations that alter the YMDD motif appear to have multiple effects on properties of the polymerase, but, in general, they appear to increase fidelity and reduce the ability to process the viral genomic code. Three-dimensional models of the HBV polymerase predict that substitution of ither isoleucine or valine, both of which are smaller than methionine, would increase the size of the dNTP-binding pocket and decrease binding affinity for lamivudine triphosphate. [129], [130]

A number of different mutational patterns appear to be responsible for the lamivudine resistance, but most cases of HBV resistance to lamivudine affect the M204 codon (see Table 107-4). One pattern is associated with two amino acid substitutions (L180M + M204V), whereas a second pattern causes a single substitution (M204I). [115], [117] A variety of other mutations, which cluster mainly in the polymerase B and C motifs, have also been described in association with lamivudine resistance (see Table 107-4).

Nucleotide changes, which produce the M204V/I polymerase mutations also, alter codons in the overlapping surface antigen ORF (see Fig. 107-1). The nucleotide change responsible for the M204V change also results in an isoleucine-to-methionine substitution at residue 195 (I195M) HBsAg. Several nucleotide substitutions can cause the M204I change in the polymerase. Depending on the specific nucleotide substitution, there are three possible alterations to the surface antigen: (1) tryptophan-to-serine change at position 196 (W196S), (2) a tryptophan-to-leucine change at position 196 (W196L), or (3) the introduction of a stop codon, which would truncate the surface antigen. It is unlikely that these surface antigen changes affect drug resistance, but they may perhaps affect antigenicity and subsequent binding of anti-HBs, either natural or through therapeutic administration of HBIG directed at the a determinant. Mutations that generate L180M are silent in the surface antigen ORF.

Other mutations that alter the amino acid sequence of HBV POL have emerged under selection pressure of lamivudine in vivo. Although their functional consequences remain largely unknown, two obvious possibilities exist: changes may enhance (1) drug resistance or (2) replication efficiency. Presumably, increase in fidelity and processivity (respectively) are major contributors to each process.

Resistance to Famciclovir

The antiviral activity of famciclovir against HBV was established in both the liver transplantation setting and chronically infected patients. [107], [131], [132] and [133] In general, treatment with famciclovir is less effective than treatment with lamivudine and results only in single log $_{10}$ decrease in viremia. [107] Furthermore, a significant proportion of patients fail to respond to famciclovir treatment. Nonresponse may be due to natural polymorphisms in the virus that affect antiviral sensitivity, or to variability in host metabolism that affects the production or intracellular stability of penciclovir triphosphate. [134] Resistance to famciclovir appears to develop more rapidly than lamivudine resistance and has been described both in liver transplant recipients and in immunocompetent patients. [134], [135], [136] and [137]

As with lamivudine resistance, efforts to determine the molecular basis of famciclovir resistance focused predominately on sequencing the reverse transcriptase portion of the polymerase. In contrast to lamivudine resistance, which is almost invariably associated with mutation of the codon for M204, famciclovir resistance does not affect the active site (YMDD) motif, nor does it map predominately to a single locus. Furthermore, M204 mutants are not selected by famciclovir. [134], [136]

Antiviral Cross-Resistance of Hepatitis B Variants

Clinically, a variety of drug-resistant HBV mutants have emerged under the selective pressures of lamivudine and famciclovir therapy (Table 107-5). Rational design of effective chemotherapeutic strategies for the future will require the determination of the sensitivities of these mutants to novel nucleoside and nucleotide analogs as they become available. This will be necessary both to provide effective treatment to patients who are already infected with drug-resistant HBV and to prevent, or at least minimize, the possibility of drug resistance arising in treatment-naive individuals.

Lamivudine Resistance	
Group 1: L526M + M550V	(B plus C domain)
Group 2: M550I	(C domain)
Group 3: F512L + L526M +/– M550V	(B and/or C domain)
Group 4: L426V/I + M550I	(A plus C domain)
Group 5: L526M + M550I	(B plus C domain)
Groups 1–3 isolates resistant to famciclovir	
All groups sensitive to adefovir dipivoxil	
Famciclovir Resistance	
A, B, and C domain, typically L526M	
Isolates sensitive to lamivudine, but lamivudine resistance emerges more quickly	
Isolates sensitive to adefovir dipivoxil	
Adefovir Dipivoxil Resistance	
None to date	
Entecavir Resistance	
None to date	

TABLE 107-5 Antiviral Cross-Resistance: Clinical Studies

Recent clinical reports already suggest two ominous conclusions: (1) development of lamivudine resistance is accelerated by preexisting famciclovir resistance, [136], [137] and (2) lamivudine-resistant HBV does not respond to subsequent treatment with either famciclovir or combination chemotherapy with lamivudine and famciclovir. [138], [139] and [140] In vitro assays have confirmed that lamivudine resistance confers resistance to penciclovir.

Fortunately, convincing evidence indicates that adefovir dipivoxil and entecavir inhibit replication of lamivudine- and famciclovir-resistant HBV. Cell-free polymerase assays show that the sensitivities of wild-type and genetically engineered mutant HBV polymerases to adefovir diphosphate are not significantly different. [141], [142] Analyses from several independent laboratories have confirmed that adefovir dipivoxil and entecavir are equally effective as inhibitors of wild-type virus and lamivudine-resistant HBV in cell culture. Furthermore, recent clinical experience shows that treatment with adefovir dipivoxil can suppress the replication of lamivudine-resistant HBV in vivo. [143], [144]

MANAGEMENT OF SPECIAL POPULATIONS

Alcohol use in patients chronically infected with HBV increases the risk for progression to cirrhosis, results in more rapidly progressive liver disease, and also increases the risk for liver cancer. [19], [20], [145] Patients with chronic liver disease should not drink alcohol, and patients with documented chronic HBV infection should be educated about the risks of alcohol use and advised not to drink any alcohol because the threshold for complications of alcohol use are unknown.

Dialysis patients need to be tested for HBV infection, and those without evidence of immunity need to be vaccinated with a double dose of vaccine. Patients who are chronically infected have been historically isolated from other patients in dialysis centers, but at this time, the sterile techniques used, the exchange of equipment in dialysis centers, and other advances in therapy do not mandate

complete separation of such patients.

Treating chronic HBV infection in dialysis patients should focus on the presence or absence of progressive liver disease as demonstrated by biopsy. Interferon probably has less efficacy in dialysis patients owing to the relative state of immunosuppression in these patients. Lamivudine can also be used, but dose adjustments must take place to take into account the dominant renal clearance of these medications.

The management of post–liver transplantation HBV infection is complex. [19] The most important step is to prevent graft infection or reinfection, which can start before liver transplantation by screening donors for anti-HBc and recipients for HBV DNA replication as well as by suppressing HBV replication to low levels using nucleoside or nucleotide analogs before the patient enters the operating room. Perioperative and postoperative HBIG therapy is used to bind circulating virus and prevent graft infection [146] and results in improved survival and a much lower rate of HBV graft infection, thereby preventing aggressive liver disease. Lamivudine maintains viral replication at a very low level and, when used in combination with HBIG, results in a risk of less than 5% for graft infection in compliant patients. [147] Newer nucleoside and nucleotide analogs may also be used in a similar clinical setting. The use of two oral agents may obviate the need for the quite costly use of HBIG. The emergence of mutant viruses that are resistant to lamivudine is a major problem in the treatment of HBV disease after liver transplantation, with more than 30% to 40% of patients developing resistance, and with more aggressive liver disease seen once resistance emerges. [148]

Other solid organ transplant recipients, such as bone marrow, heart, and kidney recipients, are at risk for reactivation of HBV disease after organ transplantation. [149], [150], [151], [152] and [153] An expert in hepatitis B management must evaluate these patients before they undergo transplantation. If active HBV replication is present, avoiding or delaying organ transplantation is advised unless successful suppression of HBV disease can be achieved. Some transplantation centers will not perform organ transplantations in patients who are HBsAg positive. Another clinical setting is the transmission of HBV from the organ donor to recipient, in which the risk for transmission is nearly 100% if the donor is HBsAg positive. Donation is rarely performed in this circumstance. Another risk is the presence of anti-HBc in the donor, which poses a small (less than 3%) risk for HBV transmission in organ transplant recipients other than liver recipients, with a significant risk to liver transplant recipients that ranges up to 70%. [154], [155] Finally, recipients who are anti-HBc positive can reactivate nascent HBV disease once immunosuppression is initiated. [156] Any patient who develops acute liver disease after solid organ transplantation needs to be assessed by HBsAg testing. Conversely, bone marrow transplantations have resulted in HBsAg clearance by adoptive immunity from the donor's immune cells. [157]

In summary, chronic HBV infection continues to pose a very serious health care problem in all countries of the world. There is emerging molecular, clinical, and treatment information that will markedly enhance our ability to manage patients with chronic HBV disease. Hopefully, with the advent of universal infant vaccination, treatment of all forms of chronic HBV disease, and new screening methods and treatment modalities for HBV-induced cirrhosis and liver cancer, we will markedly decrease the incidence and prevalence of this worldwide endemic disease.

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CHAPTER 108

Aijaz Ahmed and Emmet B. Keeffe

HEPATITIS C VIRUS INFECTION

EPIDEMIOLOGY

Chronic hepatitis C virus (HCV) infection has reached pandemic proportions, with an estimated 170 million individuals infected worldwide. [1], [2], [3] and [4] Data from population-based studies demonstrates that chronic HCV infection accounts for 40% of chronic liver disease and results in an estimated 8000 to 10,000 deaths annually in the United States. [5] In addition, there is a marked geographic and temporal variation in the incidence and prevalence of HCV infection. [1]

Incidence

Chronic hepatitis C is the most common chronic blood-borne infectious disease in the United States, even though the number of new infections per year has declined since 1989 by more than 80%, to 36,000 cases in 1996. [5] Current reports on new infections demonstrate that HCV infection is particularly common among young Hispanic men between 20 and 39 years of age. [3] Injection drug use is the most common risk factor currently associated with the transmission of HCV infection and is responsible for as many as 38% of newly diagnosed cases. [5]

Prevalence

The worldwide prevalence rates of chronic HCV infection vary from as low as 0.4% in the Western European countries to as high as 22% in Egypt and other parts of Africa. [6], [7], [8], [9], [10], [11] and [12] About 85% of patients develop chronic liver disease after acute HCV infection. Therefore, the past incidence of acute HCV infection is a key predictor of the current prevalence of chronic hepatitis C. [13] The Third National Health and Nutrition Examination Survey (NHANES-III), conducted from 1988 to 1994, provided data on the past incidence of acute HCV infection using national seroprevalence and age-specific incidence data from the sentinel counties surveillance program. [6], [7] The overall national prevalence of antibody to HCV (anti-HCV) was 1.8 percent, which corresponds to an estimated 3.9 million individuals (95% confidence interval, 3.1 million to 4.8 million) infected with HCV. About 65% of these individuals were 30 to 49 years of age. [6], [7] Serum HCV RNA was detectable in 74% cases, which corresponds to an estimated 2.7 million actively infected individuals nationwide (95% confidence interval, 2.4 million to 3.0 million). Of these chronically infected individuals, 74% were infected with genotype 1 (57% with genotype 1a, and 17% with genotype 1b).

Demographic Characteristics

Infection with HCV has no boundaries and affects individuals from all walks of life, ranging from children to elderly people. The highest incidence of acute hepatitis C is found among individuals between 20 to 39 years of age, with males outnumbering females. [5] African Americans and whites have a similar incidence of acute HCV infection, whereas individuals with Hispanic background have the highest incidence rate. In the general population, the highest prevalence rates of HCV infection are found among individuals between the ages of 30 to 49 years, with males predominantly affected. [4] In contrast to the rate of acute HCV infection, African Americans have a substantially higher prevalence rate of chronic HCV infection when compared with whites. A study conducted to estimate seroprevalence rates of human immunodeficiency virus (HIV), hepatitis B virus (HBV), and HCV among individuals with severe mental illness in the United States showed a 20% prevalence rate of HCV infection, which is 11 times the overall estimated prevalence rate for HCV infection in the general population. [14] Higher rates of HCV infection were also noted in patient groups with HIV and HBV infection. Individuals with asymptomatic HCV infection frequently fail to receive appropriate treatment to eradicate viremia and retard the progression of liver damage and thus may be a source of infection to others. [14]

Infection with HCV may differ in the pediatric age group (as compared with adults) with respect to transmission, natural history, and response to treatment. The prevalence of chronic HCV infection in children ranges from 0.05% to 0.4% [15]. The major mode of acquisition has shifted from parenteral transmission to maternal-infant transmission. However, the actual rate of vertical transmission is low. [15]

Transmission Patterns

Based on age-specific prevalence data, at least three distinct transmission patterns of HCV infection have been established. First, in developed nations such as the United States and Australia, chronic HCV infection mainly affects individuals between the ages of 30 and 49 years, indicating that the risk for HCV infection was at its

peak in the recent past (10 to 30 years ago) and primarily affected young adults. [1] Second, in nations such as Japan and Italy, HCV infection mainly affects older individuals, reflecting that the risk for HCV infection peaked in the distant past. Finally, Egypt is a classic example of an ongoing risk for acquiring HCV: high rates of infection are observed in all age groups. In countries with the first pattern, injection drug use is the most common risk factor associated with chronic HCV infection, whereas in those with the second or third pattern, unsafe injections and contaminated equipment used in health care facilities are primarily responsible for HCV transmission. [1] The contribution of different risk factors predicts the variability in the transmission patterns of HCV infection between various regions and countries.

Modes of Transmission

In the United States, the relative importance of the two most common exposures associated with transmission of HCV, blood transfusion and injection drug use, have undergone a substantial change. [16] Blood transfusion, which accounted for a significant proportion of HCV infections acquired more than 10 years ago, has been almost completely eliminated as a risk factor for HCV infection owing to improved screening of blood and organ or tissue donors. Since 1990, the risk for transfusion-transmitted HCV infection has been almost nonexistent, as demonstrated by the inability of Centers for Disease Control and Prevention (CDC) Sentinel Counties Viral Hepatitis Surveillance System to report any transfusion-associated cases of acute hepatitis C. [6], [7] Therefore, the risk for transfusion-related transmission of HCV is negligible. In contrast, injection drug use has consistently been associated with a significant proportion of chronic HCV infections and currently accounts for 60% of HCV transmission in the United States. Other known exposures (occupational, hemodialysis, household, perinatal) together account for about 10% of HCV transmission. In up to 10% of individuals with chronic HCV infection, no recognized source of infection can be identified, and most individuals in this category come from a low socioeconomic status. Case-control studies have found no association of increased HCV transmission with military service. [16]

The variation in HCV prevalence rates in a certain population is determined by the mode of transmission. [15] The highest prevalence rate of HCV infection is associated with high-dose or recurring direct percutaneous blood exposure, which has occurred in injection drug users, hemophiliacs who received clotting factor concentrates produced before 1987, and transfusion recipients of HCV-positive blood or blood products before 1992. [12], [13], [16], [17] Moderate prevalence rates of HCV infection are associated with smaller direct percutaneous exposures, such as experienced by individuals receiving long-term hemodialysis. Lower prevalence rates are associated with percutaneous or mucosal exposures due to high-risk sexual behavior or with low-dose percutaneous exposures experienced by health care workers. The lowest prevalence rate of HCV infection is found among individuals who lack high-risk characteristics, as exemplified by the screened volunteer blood donor population.

Injection Drug Use In the early 1990s, the decrease in the incidence of HCV infection was associated with a sharp fall in HCV infection among injection drug users. [4], [5] Up to 65% of injection drug users have detectable anti-HCV within 1 year of starting this high-risk behavior. [17], [18] Despite the high proportion of new infections due to injection drug use, for reasons that are unclear, the dramatic decline in incidence of acute hepatitis C since 1989 correlates with a decrease in cases among injection drug users. [4], [5] Several differences in risk factors associated with HCV prevalence rates were observed in two populations of young injection drug users from the same city (New York City), suggesting that indirect transmission of HCV may occur. [17] Among subjects 17 to 59 years of age, the strongest factors independently associated with HCV infection were injection drug use and high-risk sexual behavior. Independent drug-related risk factors included frequent injections, heavy crack smoking, injecting in a shooting gallery, and syringe-mediated sharing. Sharing of drug preparation equipment has been implicated as a possible route of HCV transmission. [5], [19] Intranasal cocaine use can also lead to transmission of HCV infection. [20] Other factors independently associated with HCV infection included poverty, having had 12 or fewer years of education, having been divorced or separated, and residence in an urban area as compared with the suburbs. [21], [22]
Blood Transfusion The current risk for acute infection with any pathogen associated with transfusion has decreased to less than 5%. [4] The exact risk for transfusion-related HCV infection is 0.6% per patient and 0.03% per unit of blood transfused. [23], [24], [25] and [26] Among donors whose blood units passed all screening tests, the risks of donating blood during an infectious window period were estimated as follows: for HIV, 1 in 493,000; for human T-cell lymphotropic virus, 1 in 641,000; for HCV, 1 in 103,000; and for HBV, 1 in 63,000. [25] The incidence of posttransfusion HCV infection has decreased appreciably since the implementation of donor screening using surrogate markers initially and then tests for anti-HCV. [4], [5], [19], [23], [27], [28] and [29] In 1984, the sharp decline in the incidence of transfusion-associated hepatitis was noted when donors were screened with alanine aminotransferase (ALT) and anti-HBc. In 1990, anti-HCV testing was added to the screening protocol and resulted in a further decrease in the incidence of transfusion-related hepatitis. [30] Before 1984, transfusion was the most common route of HCV infection; in particular, patients receiving multiple transfusions had the highest risk. [31], [32], [33], [34], [35], [36], [37], [38], [39], [40], [41], [42], [43], [44], [45] and [46] In hemophiliacs, unheated and dry heat–treated clotting factor concentrates carried a high risk for transmitting HCV infection. [41] In comparison, clotting factor concentrates inactivated by vapor heating had a significantly lower, close to zero, risk for transmitting HCV infection. [41], [47]
Chronic Hemodialysis The transmission of HCV infection is common among hemodialysis patients. Transmission of infection in this setting has been related to the number of blood transfusions, the duration of hemodialysis, and nosocomial transmission of HCV in the dialysis unit. [48], [49], [50], [51] and [52] Studies have failed to determine the most likely mode of transmission in dialysis units; therefore, emphasis is placed on strict adherence to blood collecting and handling using universal safety precautions, careful attention to hygiene in the dialysis units, and sterilization of dialysis machines in order to minimize the risk for HCV transmission. [52], [53] Nosocomial transmission of HCV not related to blood transfusions has been reported between patients dialyzed on the same shift and not sharing the same dialysis machine. [54] In dialysis units in Italy and France, the incidence and prevalence rates of HCV infection are 1% to 2% and 20% to 60%, respectively. [55]
Occupational The reported prevalence of HCV infection in health care workers such as orthopedic and oral surgeons is 1% to 2%, which is comparable to that in the general population. [56], [57], [58], [59], [60] and [61] However, some surveys have demonstrated seroprevalence rates ranging between 0.6% and 4.5% in health care workers, exceeding the seroprevalence rate of HCV infection in blood donors at the same facility by up to 4.5 times. [61], [62] and [63] This observation has been supported by other studies that have demonstrated significantly higher prevalence of HCV in dentists and oral surgeons as compared with controls. [64] There are no reports documenting transmission of HCV through mucus membranes. However, accidental needle-stick exposure and blood splashes to the conjunctiva have been associated with transmission of HCV infection. [60], [65], [66] The incidence of HCV transmission by accidental needle-stick exposure varies from 0% to 10% and has been associated with the viral load in the inoculum, depth of inoculation, and size of needle. [5], [67], [68] and [69] The risk may be higher with needles that have hollow channels as compared with nonhollow sharps. [68] Nosocomial transmission of HCV infection has been suspected, and health care workers have been reported to transmit HCV infection to their patients. [54], [70] HCV was detected in the biopsy suction channel after an endoscopic procedure in 27% of patients with chronic HCV infection, and HCV RNA was detected on the biopsy forceps in 6% of cases. [71] A thorough disinfection procedure appears to be effective in eliminating the risk for HCV transmission. Percutaneous transmission of HCV has been documented due to lack of sterile practices and reuse of contaminated supplies in traditional medicine, folk medicine, tattooing, body piercing, commercial barbering, circumcision, and scarification. [72], [73], [74] and [75] On the other hand, there are case-control studies that show no evidence of increased HCV transmission associated with exposures resulting from medical, surgical, or dental procedures; tattooing; acupuncture; ear piercing; or foreign travel. [16] HCV transmission does not appear to be associated with military service. [16]
Sexual Transmission It is estimated that up to 20% of patients with chronic hepatitis C and no identifiable risk factor may have acquired HCV sexually. [76], [77] HCV RNA has been detected in the semen of as many as one third of HCV viremic men. [76] Although seminal viral loads are low, semen could be infectious and play a role in the spread of HCV infection. The risk for acquiring HCV rises in men who have sex with men and bisexual men, [78], [79] in heterosexuals with multiple sexual partners, [78] with the number of sexual contacts, [80] with other sexually transmitted diseases such as syphilis and HIV, [81] in partners of injection drug users, [11] in prostitutes and their contacts, [82] with failure to use condoms, [78], [79] with sexual activities involving trauma, [78], [79], [82] and with divorce and separation. [2] Patients with HCV and HIV co-infection have a fivefold greater risk for transmitting HCV to their partners. [83] Male-to-female transmission of HCV is higher than female-to-male transmission. [84], [85] The risk for sexual transmission of HCV is significantly lower than that of HBV, HIV, and other sexually transmitted diseases. [79]
Vertical Transmission The rate of mother-to-child transmission of HCV is low. [86], [87] However, based on the prevalence of HCV infection in pregnant women in the Western world (0.09% to 0.25%), a reasonable number of pediatric HCV infections may occur. [87] Prospective trials have shown that the transmission rate of mother-to-infant HCV infection is 7.8% in anti-HCV–positive mothers. [88] It has been demonstrated that high maternal viral load, vaginal delivery, HCV and HIV co-infection, and negative anti-NS4 antibody are significant risk factors for vertical transmission of HCV. [89], [90], [91], [92] and [93] The placenta acts as a barrier and prevents free, direct communication between maternal and fetal blood. [94], [95] and [96] This barrier is broken at birth, resulting in free communication between the maternal and fetal blood. [97] Elective cesarean delivery is associated with lowest amount of microtransfusion from mother to fetus. [97] Therefore, it has been proposed that patients with high HCV viral load and other risk factors for vertical transmission of HCV infection might benefit from elective cesarean section. [97], [98]

MOLECULAR VIROLOGY AND PATHOGENESIS

Molecular Organization and Characteristics

HCV RNA was isolated in 1989 using a random-primed cDNA library established from the plasma of patients with suspected non-A, non-B hepatitis infection. [99], [100]

and [101] Using electron microscopy, HCV has been visualized in chimpanzee hepatocytes and human T and B cells. [102] HCV genome is a 9.6-kb long, single-stranded RNA molecule. The genomic size and molecular organization suggest close association with flaviviruses and pestiviruses. [103] Data from filtration studies indicate that the HCV ranges in size from 30 to 60 nm in diameter. [103] The virus is coated with a bilayer lipid envelope. [104] The crucial structural constituent of the HCV genome is the core protein, which is found at the amino terminal of the polyprotein. The core protein is strictly conserved in HCV genotypes. The coating glycoproteins that form the HCV envelope are referred to as E1 and E2. [105] The E2 glycoprotein has two hypervariable (HVR) segments: HVR-1 and HVR-2. These hypervariable segments provide HCV with the basic mechanisms for evading the host immune defenses and establishing chronic infectivity. [106], [107], [108] and [109] Anti–HVR-1 antibodies provide protection against HCV infection, supporting the fact that this region harbors a crucial signal for a neutralization factor. HVR-1 activates helper T-cell responses during HCV infection. It was noted that the frequency of anti–HVR-1 T-cell responses was more pronounced in patients who recovered after interferon therapy than in patients who did not respond. [110], [111] The HCV must bind to a receptor on the cell membrane to facilitate entry into the cell. It has been shown that E2 envelope proteins have the ability to bind cell membranes. [112] The entry into the cell and exposure to the intracellular acidic environment causes irreversible structural changes in envelope proteins coating the HCV genome. [113] This is followed by release of genomic RNA, triggering protein translation. Furthermore, multiple independent adaptive mutations cluster in the HCV nonstructural protein NS5A and confer increased replicative ability in vitro. [114]

Genotypes and Quasispecies

The open reading frame of HCV RNA demonstrates polymorphism and encodes a polyprotein precursor consisting of about 3000 amino acids, varying among different genotypes. HCV is classified into four hierarchical levels: genotypes, subtypes or subgenotypes, isolates, and quasispecies. [115], [116], [117], [118], [119], [120], [121] and [122] HCV appears to exist simultaneously within an individual as a series of related, but immunologically distinct, variants called *quasispecies*. The existence of these quasispecies may provide a mechanism for the virus to escape the host immune response. The HCV genotype, however, is stable and does not change over the course of infection in an individual. The evolutionary dynamics of the HCV quasispecies during the acute phase of hepatitis C predict whether the infection will resolve or persist. [123] Individuals infected with HCV can develop superinfection with other HCV genotypes, leading to eradication or suppression of the primary genotype. Simultaneous infection with multiple HCV genotypes may result in concomitant infection or eventual dominance by one genotype. [124] The genotype distribution showed no relation to the severity of the liver disease or to the duration of the disease. [125]

Pathophysiology and Immunopathogenesis

The mechanistic pathways involved in the pathogenesis of chronic HCV infection include direct HCV-induced cytopathic insult to hepatocytes or injury resulting from specific or nonspecific immune responses. [126] It has been demonstrated that cytotoxic T cells and cytokines produced by both $CD4^{+}$ (helper T cells) and cytotoxic T cells may trigger the inflammatory response in the hepatocyte. [126] The helper T-cell type 2 (Th 2) immunity, as well as weak HCV-specific T-cell response, is associated with viral persistence. [127] The production of interleukin-12 and nitric oxide is critical for the induction of Th 1 and innate immunity. [127] Patients infected with HCV mount a humoral immune response to epitopes of HVR-1. However, HCV infection may be difficult to eradicate owing to the rapidly evolving nature of HVR-1, which results in the generation of variants that are not recognized by preexisting antibodies. [128] Treatment-induced control of viremia is associated with the development of HCV-specific T-cell responses with enhanced interferon-? and low interleukin-10 production. [129] The greater efficacy of combination therapy using interferon-a plus ribavirin may be related to its ability to suppress HCV-specific interleukin-10 production. [129] The resolution of HCV infection is not followed by protective immunity against reinfection with homologous or heterologous strains. [130]

Histopathology

Histopathologically, chronic hepatitis C does not have characteristic pathognomic features and demonstrates wide variation of pathologic findings. [131] The mechanism underlying hepatocyte and bile duct injury in HCV infection results from host immune response to the virus and direct viral cytopathogenecity. Acute hepatitis C is histologically indistinguishable from other causes of acute viral hepatitis. [131], [132] HCV infection can present histologically as a triad of lymphoid aggregates in portal tracts, epithelial damage of small bile ducts, and microvesicular and macrovesicular steatosis. [131] Antibodies directed against HCV antigens allow identification of viral proteins by immunohistochemistry using monoclonal or polyclonal antibodies, particularly when frozen tissues are employed. [132] Detection of HCV RNA sequences in fixed liver tissues by in situ hybridization appears to be a more sensitive method than immunohistochemical staining. [131], [132] Immunostaining should be performed to rule out other chronic liver diseases.

A number of numeric scoring systems have been used to grade the amount of inflammation and stage and degree of fibrosis in liver biopsy specimens from patients with chronic hepatitis C. The histological activity index was the original scoring system and has been used in many clinical investigations. [133] Biopsies are graded in four categories: periportal necrosis, intralobular necrosis, portal inflammation, and fibrosis. [133] The numeric designations used for the degree of fibrosis in the various scoring systems are generally as follows: (1) stage 1—portal fibrosis; (2) stage 2—periportal fibrosis with few septa; (3) stage 3—bridging fibrosis with many septa; and (4) stage 4—cirrhosis. [133], [134], [135] and [136] These scoring systems provide definitive end points for statistical analysis of serial changes in liver histology and offer an alternative to the use of traditional pathological descriptions in following the natural history and treatment responses of asymptomatic chronic active hepatitis. The terms chronic active hepatitis, chronic persistent hepatitis, and chronic lobular hepatitis are no longer used. [135]

SCREENING AND DIAGNOSIS

Screening

Primary care physicians should routinely screen new patients for a history of risk factors associated with HCV infection. During the history and physical examination, findings suggestive of chronic liver disease include encephalopathy, ascites, edema, spider angiomas, palmar erythema, a firm liver edge, and splenomegaly. Most patients with chronic HCV infection have nonspecific constitutional symptoms, such as fatigue and decreased energy level. Elevated serum ALT levels can predict the prevalence of HCV infection in some populations. [137], [138] and [139] In summary, a test for anti-HCV should be performed if an elevated ALT level is found or a positive history of risk factors for HCV infection or physical findings suggest the presence of chronic liver disease. A qualitative polymerase chain reaction (PCR) test for HCV RNA is warranted in patients who test positive for anti-HCV, particularly those with normal ALT levels or no HCV risk factors, to confirm HCV infection and rule out a false-positive test or recovery from past HCV infection. Serologic tests for anti-HCV are probably the best screening tests for dialysis patients.

Chronic HCV infection may be a multisystem disease, and patients should be screened for cryoglobulinemia by checking serum cryoglobulins, serum creatinine, and a urinalysis. More than 50% of patients with chronic HCV infection have cryoglobulinemia, but only 1% to 5% experience related symptoms. Additional baseline laboratory tests that are commonly obtained after the initial diagnosis of chronic hepatitis C include a standard liver panel, complete blood cell count, prothrombin time, tests for co-infection with HBV or HIV, antinuclear antibody to exclude coexistent autoimmune hepatitis, alpha-fetoprotein level, and abdominal ultrasound. If treatment is being considered, a quantitative rather than qualitative serum HCV RNA test and HCV genotype are also obtained. In acute hepatic injury, prothrombin time and, to a lesser extent, total bilirubin are the best indicators of severity of disease. Although ALT is useful for detecting acute and chronic hepatic injury, it is not related to severity of acute hepatic injury and is only weakly related to severity of chronic hepatic injury. [140]

Diagnosis

The discovery of HCV in 1989 led to the development of diagnostic serologic and virologic tests. [141], [142], [143], [144], [145], [146], [147], [148] and [149] The diagnostic tests used to detect HCV infection can be divided into two main classes: (1) serologic tests or assays that detect anti-HCV; and (2) molecular tests or assays that detect, quantify, and characterize HCV RNA. [142], [143] and [144]

Serologic assays have been subdivided into screening tests for anti-HCV, such as the enzyme immunoassay (EIA), and supplemental tests, such as the recombinant immunoblot assay (RIBA). Three generations of anti-HCV tests have been developed, and each generation has provided higher sensitivity for detecting anti-HCV. Third-generation anti-HCV tests (EIA-3 and RIBA-3) contain antigens from the HCV core, nonstructural 3, nonstructural 4, and nonstructural 5 genes. The EIA-1 contained a single HCV recombinant antigen derived from a nonstructural gene. The specificity of EIA-1 was 30% to 50% in low-risk populations, such as blood donors, and the development of detectable antibodies after acute infection required several months. [142] The use of additional antigens in EIA-2 and EIA-3 tests has increased the sensitivity and shortened the time to detection after acute infection to a median of 7 to 8 weeks, but false-positive anti-HCV by EIA results still occur in low-risk populations. EIA-3 did not detect more HCV-infected individuals in a donor population that previously tested negative in EIA-2, but it did detect HCV

antibodies earlier in some patients with acute HCV infection. EIA-2 and EIA-3 are significantly more sensitive than RIBA-2 and RIBA-3. [145], [146] Supplemental anti-HCV tests are designed to resolve false-positive testing by EIA and are appropriately used in low-prevalence settings in which false-positive anti-HCV tests remain a problem. [144], [145] and [146] In practice, confirmation of HCV infection is performed by detection of HCV RNA by PCR or the branched-chain DNA (bDNA) assay. [145], [146] and [147] The EIA-3 has an estimated sensitivity of 98.9% and specificity of 100% in patients with chronic HCV infection, whereas the sensitivity RIBA-3 is 78.8% in the hemodialysis population. [148] These data provide evidence for the good sensitivity and specificity of EIA-3 assays, particularly in high-risk patient groups, and confirm their use for screening in these populations. [147], [148]

Detection of serum HCV RNA provides evidence of active HCV infection and is useful as a confirmatory test. [142], [143], [144], [145], [146] and [147] Optimal HCV RNA assays using PCR at present have a sensitivity of less than 100 copies/mL. There are several methods for assessing HCV RNA levels, including various PCR assays, the bDNA, and transcription-mediated amplification. Quantitative PCR is the most sensitive test for quantifying hepatitis C viral load, whereas the bDNA test appears to be the most specific method. However, it must be recognized that these molecular tests have their own limitations, which include inadequate dynamic range, high variability of PCR assays, and poor sensitivity of the bDNA test. [142], [149] Commonly used serum HCV RNA qualitative and quantitative second-generation PCR assays are Amplicor HCV v2.0 and Amplicor HCV Monitor v2.0, respectively. Second-generation assays are 1 log more sensitive and genotype independent, compared with their respective first-generation tests (10^{2} copies/mL versus 10^{3} copies/mL for qualitative tests; 10^{3} copies/mL versus 10^{4} copies/mL for quantitative tests). [143], [144]

An anti-HCV–positive result in a low-risk setting, such as blood donation, should be confirmed with an analytical antibody test. Subsequently, a serum HCV RNA test should be performed on donors with a positive or indeterminate confirmatory test result. On the other hand, an anti-HCV–positive test result in high-risk populations is likely to be true positive. A quantitative HCV RNA test and genotyping should be performed if therapy is being considered. [148]

According to data from the CDC, 200,000 to 700,000 new cases of acute viral hepatitis occur in the United States each year. [3], [4], [5] and [6], [149] In 1995, these cases included 180,000 new hepatitis A virus (HAV) infections, 128,000 HBV infections, and 28,000 HCV infections. From these cases, there are about 100 deaths per year from fulminant hepatitis A and 150 deaths from fulminant hepatitis B; hepatitis C has rarely been implicated as a cause of fulminant hepatic failure. Although the death rate from acute viral hepatitis is low, many adult patients with HBV infection (5%) and HCV infection (85%) become chronically infected, and a significant percentage of these patients ultimately develop cirrhosis or hepatocellular carcinoma (HCC) over two or more decades. Because 75% of acute viral hepatitis results from either HAV or HBV infection, initial laboratory investigation should include serologic tests to exclude HAV or HBV infection. In the event these studies are negative, further testing should be performed to rule out acute HCV infection. Serum HCV RNA is detectable 1 to 2 weeks after the onset of infection, whereas antibodies to HCV are not detected until 8 to 10 weeks after infection with HCV. In clinically stable patients, it may be plausible to wait and check antibodies to HCV.

NATURAL HISTORY

Acute Infection

Acute infection with HCV is usually asymptomatic and therefore not diagnosed and underestimated. Acute HCV infection is the third most common cause of acute hepatitis associated with jaundice. The most common cause of acute hepatitis in United States is acute hepatitis A, which accounts for 49% of all cases. [29], [150] This is followed by acute hepatitis B (35% of cases) and acute HCV infection (16% of cases). [29] As many as 43% of cases of acute hepatitis C are associated with intravenous drug use, 17% with sexual exposure to an infected individual or with high-risk sexual behavior, and less than 10% with other risk factors also known to transmit HCV infection. [29], [151] No known risk factors can be identified in 31% of patients with acute HCV infection. [29]

The incubation period from infection to onset of symptoms can range from 2 to 12 weeks, with an average of 6 to 7 weeks. [6], [152], [153] and [154] Serum HCV RNA is detectable within 7 to 21 days after an acute infection. [154] Serum HCV RNA, in titers of 10^{5} to 10^{7} copies/mL, can be detected at the onset of jaundice. [155] Aminotransferase levels are elevated between 14 to 28 days after infection, and anti-HCV becomes detectable about 60 days (varies from 20 to 150 days) after the onset of infection. [154]

Preicteric Period A wide constellation of clinical features, including low-grade fever, decreased energy level, fatigue, nausea, vomiting, right-sided abdominal pain, myalgias, and arthralgia, may develop in 15% to 20% of patients. This clinical syndrome occurs within 2 to 12 weeks of acute HCV infection and also lasts for 2 to 12 weeks. Systemic symptoms may be followed by jaundice. In rare circumstances, patients can develop rash, hives, and arthralgias that are most likely secondary to a serum sickness syndrome precipitated by immune complexes. [152], [153] and [154] Most patients with acute hepatitis C, however, remain asymptomatic.
Icteric Period The nonspecific symptoms, which develop in the preicteric phase, usually persist during the period of jaundice. Infrequently, jaundice can be a presenting symptom of acute HCV infection and typically lasts for 1 to 2 weeks. Serum HCV RNA usually becomes detectable with the onset of jaundice. Fulminant liver failure after HCV infection has been reported but is a rare occurrence. [29], [154]
Resolution Versus Chronicity After the acute onset, HCV infection is self-limiting and resolves 3 to 6 months later in 15% to 30% of cases, with return of ALT levels to normal and disappearance of serum HCV RNA, although anti-HCV remains detectable. During convalescence, patients report resolution of jaundice, improved appetite, weight gain, and feeling of well-being. However, nonspecific clinical features may persist for a few months in a small minority of patients who recover from acute HCV infection. A higher rate of HCV eradication is noted in patients with clinically symptomatic acute HCV infection as compared with those who are asymptomatic. Therefore, the outcome of HCV infection is dependent on the viral host immunologic interactions in the first few weeks of infection. Patients who recover from acute HCV infection usually do so within the first 12 weeks of infection. Nearly 10% of patients who develop anti-HCV response eventually lose the nonneutralizing antibody and evidence of past acute infection. [156] Some patients may not develop anti-HCV response with short-duration, acute hepatitis C; it is difficult to estimate the proportion of patients with this response. Spontaneous clearance and resolution of HCV infection is rare after 6 months of persistent infection. [157] Up to 75% to 85% of adults and 55% of children develop chronic HCV infection. [152], [153] and [154], [158], [159], [160] and [161]

Chronic Infection

Chronic HCV infection develops in as many as 85% of patients after acute infection. [162] Most patients with chronic infection have asymptomatic elevation of aminotransferase levels and lack physical findings suggestive of liver disease. [163], [164] Fatigue is the most common symptom reported by patients with chronic HCV infection. [163] The correlation of symptoms with histological severity of liver disease is poor in patients with chronic hepatitis C; patients may have fatigue associated with either mild or severe disease. [163] Vague and intermittent right upper quadrant pain is the second most frequent symptom noted in patients with chronic HCV infection. Other less common nonspecific symptoms include low-grade fever, nausea, vomiting, myalgia, and arthralgia. Up to 15% to 20% of patients with chronic HCV infection have psychological or psychosocial problems, which complicate initial assessment and later decisions regarding treatment. One third of patients with chronic HCV infection have normal ALT levels, and 25% have ALT levels that are elevated to less than twice the upper limit of normal. [162], [165] ALT elevations are persistent in 26% of cases and intermittent in 40% to 70% cases, with resolution to normal range in 17% of patients on long-term follow-up. [162], [165], [166] Self-limited, intermittent ALT flares have been noted and may be associated with an increase in serum HCV RNA, but not with viral eradication. [167] A 10-fold rise in ALT levels has been associated with severe piecemeal necrosis. [162], [163], [168] The titer of serum HCV RNA is not associated with histological severity.

Hepatitis C–Related Cirrhosis

Several studies have now confirmed that chronic HCV infection demonstrates minimal clinical progression in the initial two decades after infection. [164], [169], [170], [171], [172], [173], [174] and [175] Chronic infection with HCV leads to cirrhosis in at least 20% to 25% of immunocompetent patients within 20 years of the onset of infection.

Risk Factors Associated with Rapid Progression to Hepatitis C–Related Cirrhosis Cirrhosis and end-stage liver disease may develop more rapidly under certain circumstances. [161] Factors influencing the rate of progression of fibrosis include gender (male sex), genetic background (Japanese at higher risk than Americans), age at exposure (higher risk for cirrhosis with age greater than 40 years at time of HCV infection), duration of HCV infection (longer), route of transmission (blood transfusion greater risk than injection drug use), and coexisting conditions, including alcoholic liver disease (daily alcohol consumption of 50 g or more), chronic tobacco use, HIV infection, and HBV infection. [176], [177], [178], [179], [180], [181] and [182] Between 10% and 20% of patients with chronic hepatitis B have detectable anti-HCV. [183], [184] Alternatively, 2% to 10% of patients with anti-HCV demonstrate markers of HBV infection. Co-infection with HBV and HCV results in more severe liver disease than does isolated HBV or HCV infection. The median duration of chronic HCV infection required for the development of cirrhosis can vary from 13 years in men who use alcohol and are older than 40 years of age at the time of infection to as long as 42 years in women who avoid alcohol and are less than 40 years of age at the time of infection. [185] The overall risk for cirrhosis is less than 10% over a 10-year period in patients with mild chronic hepatitis, 44% in those with moderate hepatitis,

and 100% in those with severe hepatitis with bridging fibrosis. [180]

Clinical Presentation The mean age of patients presenting with HCV-related cirrhosis is 55 years, as compared with a mean age of 42 years in patients presenting with chronic HCV infection without cirrhosis and of 66 years in patients with HCC. [186] Other studies have also confirmed a mean difference of at least 10 years between the diagnosis of cirrhosis and detection of HCC in patients with chronic HCV infection. [187], [188] and [189] HCV-related cirrhosis affects men more commonly than women, with a ratio ranging from 1.3:1 to 3.2:1. [165], [190], [191] The diagnosis of HCV-related compensated cirrhosis is often made incidentally during screening of blood donors or at the time of routine laboratory testing. [165] A wide spectrum of nonspecific symptoms can be noted in patients with compensated and decompensated cirrhosis, including fatigue in 75%, abdominal pain in 24%, and anorexia in 13%. [162] Systemic symptoms are more severe in patients with decompensated cirrhosis, and the annual risk for decompensation is 3.9%. [165] The clinical presentation can be dramatic after hepatic decompensation and manifests with ascites in 48%, variceal bleeding in 22%, hepatic encephalopathy in 8%, jaundice in 6%, or combination of complications in 17% of patients. [165] Other clinical features noted with hepatic decompensation include lower extremity edema, easy bruising, pruritus, muscle wasting, muscle cramps, and sexual dysfunction. Patients with HCV-related compensated cirrhosis have a mean survival rate of 80% at 10 years, compared with a mean survival rate of 50% at 5 years once decompensation occurs. [165] Physical findings suggestive of cirrhosis include a palpable firm liver edge in 79%, hepatomegaly in 50% to 78%, splenomegaly in 25% to 37%, and cutaneous stigmata of advance liver disease in 31%. [165] Most patients with HCV-related cirrhosis have mild to moderate elevation of aminotransferase levels, with less than 5% of patients having ALT levels 10-fold higher than the upper limit of normal. [165] The association between ALT levels and histological severity of chronic HCV is poor. [168], [192] Coagulopathy, hypoalbuminemia, hyperbilirubinemia, and thrombocytopenia are associated with a decrease in survival rate at 10 years. [165] The annual risk for HCC in patients with chronic hepatitis C with cirrhosis is 1.4% to 3.3% in the United States and Europe and as high as 6.9% in Japan and Italy. [193] The mean length of time from HCV infection to the development of HCC is 29 years. [162], [165]

CLINICAL MANIFESTATIONS AND COMPLICATIONS

Chronic HCV infection can be associated with extrahepatic organ involvement and can lead to complications primarily involving the hepatic parenchyma. Most of these manifestations are directly associated with HCV infection; however, HCC nearly always develops in the setting of HCV-related cirrhosis. Other complications associated with HCV-related cirrhosis were discussed earlier in the respective section.

Extrahepatic Clinical Manifestations

The prevalence of extrahepatic clinical manifestations is relatively low but can be associated with significant morbidity and even mortality. The precise pathogenesis of these extrahepatic manifestations has not been determined, although most represent the clinical consequences of an immune-mediated response. [194], [195]

Autoantibodies There is a high prevalence of a variety of autoantibodies, usually in low titers, associated with chronic HCV infection. Antinuclear antibody, anti–smooth-muscle antibody, and antithyroid antibody are detectable in 40% to 65% of patients with chronic HCV infection. [196], [197] The clinical significance of most of these autoantibodies is negligible, except for liver-kidney-microsomal type 1 antibodies (anti-LKM1). [197], [198] The patients with anti-LKM1 can be differentiated into two groups, anti-HCV positive and anti-HCV negative. [199], [200] Type 2 autoimmune hepatitis with high anti-LKM1 titers and negative anti-HCV primarily affects young women with moderate to marked elevation of ALT levels. These patients are responsive to immunosuppressive therapy and are HLA-DR3 positive. In contrast, patients with chronic HCV infection and a positive anti-HCV associated with a positive anti-LKM1 have distinctive features, such as an equal distribution between men and women, mildly elevated ALT levels, low anti-LKM1 titers, and responsiveness to interferon therapy. [201] Finally, patients with classic type 1 autoimmune hepatitis can have a false-positive anti-HCV, which can be confirmed with the finding of a negative serum HCV RNA. [202], [203]

Renal Disorders A wide array of renal disorders can occur in association with chronic HCV infection. [204], [205], [206], [207], [208] and [209] The pathogenesis of HCV-related renal disorders remains undefined; however, deposition of circulating immune complexes in the subendothelial space and mesangium in the glomeruli plays a central mechanistic role. These disorders may be manifest as cryoglobulinemic systemic vasculitis, proteinuria, microscopic hematuria, nephrotic syndrome, or acute renal failure. Uremic patients with chronic HCV infection and those undergoing regular dialysis frequently develop immunologic abnormalities. The most common HCV-related renal disorder is type I membranoproliferative cryoglobulinemic glomerulonephritis with a chronic course interspersed by acute recurrent episodes. The membranoproliferative cryoglobulinemic glomerulonephritis is usually associated with type II cryoglobulinemia with immunoglobulin M (IgM) rheumatoid factor, polyclonal IgG anti-HCV antibodies, and HCV RNA IgM as the principal constituents in cryoprecipitate. The treatment of renal disorders associated with chronic HCV infection is discussed in the section on “Treatment.” [210], [211], [212], [213], [214] and [215]

Endocrine Disorders Infection with HCV may contribute to the development of diabetes mellitus. Both disorders are common; therefore, cross-sectional studies were performed to confirm the close association. [216], [217] and [218] Patients with chronic HCV infection have an increased prevalence of type 2 diabetes mellitus. HCV-related cirrhosis can induce insulin resistance and predispose patients to diabetes mellitus. [216], [217] and [218] However, the increased prevalence of diabetes is independent of cirrhosis. [217] Further investigation is needed because the pathogenesis may be multifactorial and unique to HCV. [217] In the United States, type 2 diabetes mellitus occurs more often in individuals with chronic HCV infection who are older than 40 years of age. [218] Autoimmune thyroiditis can be associated with HCV-related liver disease. [219], [220] Antithyroid antibodies are found in 5% to 13% of patients with chronic HCV infection, predominantly affecting older women. [219], [220] Autoimmune thyroiditis is manifested as hypothyroidism in 3% to 6% of patients with chronic HCV infection. Interferon-related thyroid disease is more frequently noted in patients with preexisting thyroid antibodies. However, interferon can induce the formation of antithyroid antibodies. Discontinuation of interferon therapy is usually accompanied by resolution of the thyroid disorder. Salivary and lacrimal gland involvement is common in HCV-infected individuals, but HCV antigens are not detectable in affected glands. [221] Testicular atrophy and sexual dysfunction can be associated with HCV-related cirrhosis.

Rheumatologic Disorders Rheumatologic disorders associated with chronic HCV infection are common and include mixed cryoglobulinemia, vasculitis, sicca symptoms, myalgia, arthritis, and fibromyalgia. [221], [222], [223] and [224] There is a wide spectrum of clinical manifestations that can be noted and are not characteristic of HCV infection. However, a characteristic joint syndrome is associated with the presence of mixed cryoglobulinemia and characterized by intermittent monoarticular or oligoarticular, nondestructive arthritis affecting large and medium-sized joints. Treatment of patients with HCV-associated arthritis is limited and remains controversial.

Cutaneous and Mucosal Lesions The prevalence of anti-HCV varies from 4% to 65% in patients with cutaneous lichen planus or oral lichen planus. [225], [226] These estimates were obtained from populations with a high overall prevalence of HCV infection, such as southern Europe and Japan. [226] Other lesions noted in association with HCV infection include porphyria cutanea tarda, necrotizing cutaneous vasculitis, and Mooren corneal ulcer. [222] The high prevalence of HCV infection in patients with porphyria cutanea tarda is based on epidemiologic data. [222] The precise role of HCV in the pathogenesis of porphyria cutanea tarda is unknown.

Cardiac Dysfunction Recent studies have demonstrated an association between chronic hepatitis C and hypertrophic cardiomyopathy or dilated cardiomyopathy. [227] In a collaborative research project of the Committees for the Study of Idiopathic Cardiomyopathy, the prevalence of anti-HCV was 11% in patients with hypertrophic cardiomyopathy and 6% in those with dilated cardiomyopathy, both of which were significantly higher than the prevalence rate of HCV infection in volunteer blood donors in Japan. [227] Various other cardiac abnormalities were found, of which arrhythmias were the most frequent.

Miscellaneous Extrahepatic Manifestations Many systemic disorders can be associated with chronic HCV infection, including polyarteritis nodosa–type systemic vasculitis, [228] colonic vasculitis, [229] idiopathic thrombocytopenic purpura, [222] celiac sprue, [230] Sjögren syndrome, [222] polymyositis, [231] and multiple myeloma. [232] Systemic sclerosis has been reported in patients with chronic HCV infection, manifested by severe Raynaud phenomenon, progressive skin thickening, painful fingertip ulcers, dysphagia, and Sjögren syndrome. [233] The prevalence of chronic HCV infection in patients with systemic lupus erythematosus is greater than the prevalence of HCV in blood donors from the same population. [234] Patients with coexistent systemic lupus erythematosus and HCV infection tend to manifest a lower frequency of cutaneous systemic lupus erythematosus lesions and anti- double-stranded DNA (dsDNA) antibodies and a higher rate of hepatic involvement, hypocomplementemia, and cryoglobulinemia. [234]

Lymphoproliferative Diseases

The role of HCV infection in the etiology and pathogenesis of lymphoproliferative diseases is supported by several studies reporting high seroprevalence of HCV in patients with B-cell non-Hodgkin lymphoma. [235], [236] and [237] HCV demonstrates high affinity to lymphoid tissue. HCV is associated with type II mixed cryoglobulinemia, a benign monoclonal lymphoproliferative disorder that can transform to overt B-cell non-Hodgkin lymphoma.

Primary Hepatic Lymphoma

The etiologic association of HCV and primary hepatic lymphoma is unclear. [238] Numerous reports of primary hepatic lymphoma of B-cell origin in association with HCV infection have been published. [238] Primary hepatic lymphoma is typically a large cell, high-grade malignant B-cell lymphoma. Only a few cases of T-cell lymphoma

have been described. [238]

Hepatocellular Carcinoma

The current incidence of HCC in United States is 2.4 cases per 100,000 individuals, but the incidence of HCC is as high as 120 cases per 100,000 in China, the Far East, and sub-Saharan Africa. [193], [239], [240], [241] and [242] The occurrence of HCC in association with HCV-related liver disease shows marked geographic variation. [240], [241] and [242] The rate of detectable anti-HCV in patients with HCC varies from 60% to 70% in Spain and Italy, 50% in Japan, and 27% in United States to less than 10% in Hong Kong. [193] Within the United States, the association of HCC with a positive anti-HCV has a much higher prevalence in geographic areas with a large Hispanic population. [242], [243] and [244] About 20% to 25% patients with chronic HCV infection develop cirrhosis within 20 years of the onset of infection. HCC develops at a rate of 1.5% to 9% per year in patients with chronic hepatitis C and cirrhosis. [165], [244] Therefore, there is a 5% risk for developing HCC in patients with HCV infection over a course of 20 years. The risk factors associated with development of HCC in the setting of HCV infection include age greater than 60 years, male gender, alcohol use, co-infection with HBV, porphyria cutanea tarda, increased hepatic iron content, and severity of cirrhosis. [243], [244], [245] and [246] The incidence of HCV-related HCC is steadily rising, and HCV accounts for nearly half of all cases of HCC in the United States. [242], [243]

Ultrasound of the abdomen and serum alpha-fetoprotein are reasonable tests in a screening strategy for early detection of HCC in patients with chronic HCV infection and cirrhosis. [246] There is no consensus on the frequency with which these tests should be performed, but most experts recommend an interval of every 6 to 12 months. Screening is recommended only for patients with known or suspected cirrhosis because the risk for HCC is quite low in patients who do not have cirrhosis.

Cholangiocarcinoma

There is also a somewhat higher incidence of cholangiocarcinoma in patients with HCV-related cirrhosis, and cholangiocarcinoma is also associated with poor prognosis. [247] and [248]

TREATMENT

In general, the treatment of patients with chronic hepatitis C, including patients with compensated cirrhosis, is antiviral therapy with interferon-based therapies. Patients with decompensated cirrhosis should be referred to a liver transplantation center for consideration for liver transplantation, which is the treatment of choice when liver failure complicates chronic HCV infection.

Interferon-Based Antiviral Therapy

The best candidates for antiviral therapy of chronic hepatitis C are patients at the greatest risk for progression to cirrhosis. [181], [249] These patients are characterized by persistently elevated ALT levels for more than 6 months, detectable serum HCV RNA, and either portal or bridging fibrosis with at least moderate degrees of inflammation and necrosis on liver biopsy. Patients treated in clinical trials typically are required to have elevated ALT levels of more than 1.5 times the upper limit of normal, clinically compensated liver disease, and no other significant medical or psychiatric illness that contraindicates therapy (Table 108-1). Clinical guidelines on the management of chronic HCV infection were established at a National Institutes of Health (NIH) consensus conference in 1997 [250] and confirmed at a European consensus conference 2 years later. [251] More recently, these guidelines were confirmed and further modified. [252]

Hematologic
- Hemoglobin <12 g/dL in women and <13 g/dL in men
- White blood cell count $<1500/mm^3$
- Platelet count $<100,000/mm^3$

Hepatic
- Normal aminotransferase levels
- Decompensated cirrhosis

Cardiovascular
- Severe coronary artery disease
- Unstable angina

Endocrine
- Poorly controlled diabetes mellitus

Autoimmune
- Untreated thyroid disease

Neuropsychiatric
- Seizure disorder
- Severe psychiatric illness

Obstetric
- Pregnant or unable to practice contraception

TABLE 108-1 Contraindications: Interferon and Ribavirin Therapy

Virologic and Biochemical Response Responses to antiviral therapy of chronic HCV infection may be defined on the basis of the biochemical, virologic, and histological outcomes. The persistence of a biochemical (normal ALT) or virologic (undetectable HCV RNA) response for 6 months or more after cessation of therapy is the operational definition of a *sustained response* (SR). [253] Several studies, including a mean 4-year follow-up of 80 patients and a 10-year follow-up of 5 patients who all had initial posttreatment 6-month sustained responses showed long-term virologic sustained response rates of 96% and 100%, respectively. [254], [255] In addition, there was improvement of liver histology, including evidence of regression of hepatic fibrosis in noncirrhotic patients. These studies suggest that some cases of chronic HCV may indeed be "cured" and that a benefit on mortality should be expected if cirrhosis, with its risk of HCC, can be prevented. A posttreatment elevated ALT level and detectable HCV RNA that had been normal and undetectable, respectively, during treatment define *relapse* after a course of therapy. *Nonresponse* to antiviral therapy is defined as persistently elevated ALT levels or detectable serum HCV RNA, and a variant of nonresponse called *breakthrough* is characterized by an initial decrease of ALT levels into the normal range or the disappearance of detectable HCV RNA with subsequent elevation of ALT or presence of detectable HCV RNA while still receiving treatment. [256], [257] Failure to clear HCV RNA after 24 weeks of combination therapy predicts the lack of a sustained virologic response and has been used as a stopping rule for antiviral therapy. However, histological improvement may occur with longer therapy, and thus continuation of therapy may be beneficial in selected cases on an experimental basis.

Aims of Antiviral Therapy The primary goal of antiviral therapy in patients with chronic HCV infection is long-term viral eradication as determined by undetectable HCV RNA in serum and liver. [250], [251] and [252] Secondary objectives of treatment include improvement and prevention of liver parenchymal inflammation and fibrosis with normalization of ALT levels and reduced inflammation and fibrosis on liver biopsy.

Selective Role of Liver Biopsy The NIH and European consensus conferences recommended that a liver biopsy be performed when considering treatment for chronic HCV infection to distinguish patients most likely to benefit from therapy, that is, those with moderate histological disease, from those who may be less likely to benefit, that is, those with mild disease and no or minimal fibrosis or those with advanced disease and cirrhosis. A decision to perform a liver biopsy can be influenced by many factors, such as patient preferences, cost-effectiveness, presence of relative contraindications, and suspicion of coexistent liver diseases suggested by laboratory abnormalities like high-titer antinuclear antibody with hypergammaglobulinemia or abnormal iron studies. Liver biopsy is clearly the most reliable method of establishing the severity of liver disease due to chronic HCV infection but provides no superiority to overall clinical assessment in ruling out other unsuspected coexisting liver diseases. [258] Apprehension and anxiety are frequently noted in patients before a liver biopsy is performed. A percutaneous liver biopsy is a blind procedure, which can cause pain in 30%, severe complications in 0.3%, and death in 0.03% of patients. [259], [260] In addition, liver biopsy incurs both direct health care–related and indirect work loss–related costs. Based on a cost-effectiveness analysis, it has been recommended that the most suitable strategy in the management of chronic HCV infection is to initiate therapy in all patients and avoid a liver biopsy. [261] Considering these arguments, there is a trend to initiate therapy without performing a liver biopsy. The role of liver biopsy might be thought of as selective rather than mandatory in identifying candidates for antiviral therapy, such as for the patient who prefers to defer therapy if mild disease with no or minimal fibrosis is present. [250], [251] and [252] A number of studies have been performed in patients with chronic HCV infection to investigate the utility of laboratory tests, such as the aspartate transaminase (AST)-to-ALT ratio or the platelet count, to establish the severity of liver disease, but none of the proposed predictors is as sensitive and specific as liver biopsy. [262], [263], [264] and [265] In one study, a more complex formula correctly classified 46% of patients with or without significant fibrosis into these two respective categories. [265] Unfortunately, this formula required three assays that are not routinely performed in patients with chronic HCV infection (a $_2$-microglobulin, haptoglobin, and apolipoprotein A $_1$), in addition to three routine tests (?-globulin, ?-glutamyltranspeptidase, and total bilirubin). The future treatment of chronic hepatitis C may include antiviral drugs and also antifibrotic agents, which might necessitate the performance of a liver biopsy or the identification of reliable serum markers of fibrosis to establish the end points of therapy. A wide variety of serum fibrosis markers have been studied to predict the stage of fibrosis in HCV-related liver disease. [266], [267], [268] and [269] The serum markers that have been studied include procollagen type III *N*-terminal peptide, 7S fragment of type IV collagen, hyaluronan, matrix metalloproteinase-1 and -2, and tissue inhibitor of

metalloproteinase-1. [266], [267], [268] and [269]

Interferon-Induced Viral and Immune Kinetics The potency of interferon is determined by measuring antiviral activity in biologic assays relative to a number of international reference standards. Further insight into interferon-induced viral kinetics and host immune modulation has been provided by recent studies. [270], [271], [272], [273] and [274] It has been demonstrated that multitarget, potent $CD4^+$ proliferative T-cell responses are maintained long term after eradication of HCV infection, as compared with focused and weak responses in patients with persistent HCV infection. [273] On the other hand, the HCV-specific $CD8^+$ T-cell response is quantitatively weaker in both responders and nonresponders. [273]

Standard Interferon and Ribavirin Combination Treatment of Naive Patients The four types of interferon-a that have been evaluated in a large number of patients with chronic HCV infection include interferon-a-2b, [275] -a-2a, [276] -a-n1 (lymphoblastoid interferon), [277] and a-con-1 (consensus interferon). [278] In the United States, as of late 2001, interferon-a-2b, interferon-a-2a, interferon-a-con-1, combination interferon-a-2b with ribavirin, and peginterferon-a-2b with ribavirin are licensed and marketed for the treatment of chronic HCV infection. The U.S. Food and Drug Administration originally licensed interferon in 1991 for the treatment of chronic hepatitis C, and the initial strategy of treating patients for 6 months yielded 10% to 15% sustained response rates. It was later found that 12 to 18 months of therapy increased the response rates to 20% to 25%. [253] Interferon and ribavirin combination antiviral therapy became the standard treatment for chronic HCV infection from in 1998. [252] Ribavirin, 1000 to 1200 mg/day in two divided doses, combined with interferon-a-2b, 3 million units (MU) three times weekly for 6 months, was found to improve significantly the biochemical and virologic sustained response rates compared with interferon alone in an early pilot experience. [279], [280] Phase III American and European trials comparing standard interferon monotherapy with combination interferon-a-2b plus ribavirin therapy showed a significant improvement in response rates for all measures of efficacy, including virology, ALT levels, and histology in treatment-naive patients with chronic HCV infection. [281], [282] Serum HCV RNA was eradicated in 31% to 35% patients after 6 months of combination therapy and in 38% to 43% patients with 12 months of combination therapy, as compared with 13% to 19% with interferon monotherapy. [281], [282] These studies also showed important differences in the outcome based on genotype: about 30% of patients with genotype 1 versus about 65% with genotype 2 or 3 had sustained virologic responses. In addition, patients with genotype 1 had increased rates of sustained virologic response with 48 versus 24 weeks of therapy, whereas patients with other than genotype 1 achieved no additional benefit after 24 weeks of combination therapy. These studies dictate that combination therapy be administered for 12 months to patients with genotype 1 but only 6 months to those with genotypes 2 and 3 to achieve maximal sustained response rates. Multiple factors, [281], [282] in addition to genotype, are known to predict the likelihood of successful therapy with ribavirin and interferon and may influence the decision of whether to undergo a course of therapy.

Retreatment of Relapsers and Nonresponders Retreatment studies have been performed in patients who received interferon monotherapy and were nonresponders at the end of treatment or relapsed after treatment was discontinued. [278], [283], [284], [285] and [286] Retreatment response rates have been highly variable, depending in large part on the response to the initial course of the treatment. The only treatment strategies that have shown some favorable results with interferon monotherapy are use of a higher dose, treatment for a longer period of time, or use of a different interferon. In general, biochemical end-of-treatment rates are much higher in relapsers (79% to 85%) than nonresponders (14% to 30%). [283] The best predictors of a sustained response to retreatment of relapsers were a negative serum HCV RNA at the end of the first course and an initial treatment course of only 6 months. [283] Retreatment of relapsers with interferon monotherapy, such as interferon-a-con-1, 15 µg three times weekly for 12 months, achieved a 58% sustained virologic response. [278], [285] In contrast to the reasonably good results of retreatment of relapsers, the cumulative data indicate that nonresponders to interferon therapy achieve only modest responses with retreatment, such as 13% sustained virologic response to interferon-a-con-1, 15 µg three times weekly for 12 months. Furthermore, some recent studies suggest that a small proportion of patients who failed to respond to previous interferon monotherapy (i.e., noncirrhotic patients with low levels of virus and genotypes other than 1) may also benefit from retreatment with combination interferon and ribavirin. [287]

Pegylated Interferons Polyethylene glycol (PEG) is a large, inactive water-soluble polymer attached to interferon with a covalent bond. [288], [289] and [290] The process of pegylation results in reduced clearance of pegylated interferon and up to 10-fold increase in drug half-life, allowing once a week administration. [288], [289] and [290] Pegylated interferon has improved efficacy by maintenance of a steady serum level but may suffer a variable loss in interferon activity determined by the size of the PEG molecule and site of pegylation. [271], [288], [289] and [290] The two PEG interferons that have been studied include the long-branched 40-kd peginterferon-a-2a (Pegasys, Roche, Nutley, NJ), which is metabolized by the liver, and the short linear 12-kd peginterferon-a-2b (PEG-Intron, Schering, Kenilworth, NJ), which is excreted by the kidneys. [291], [292] Pilot studies demonstrated that the safety profile of pegylated interferon administered in ascending subcutaneous doses to healthy subjects was comparable to that of standard interferon. [291], [292], [293] and [294] Pegylated interferon monotherapy was more efficacious than standard interferon monotherapy in a dose-dependent fashion. [291], [292], [293] and [294] In one clinical trial, 531 interferon-naive patients with chronic HCV infection were treated for 48 weeks with peginterferon-a-2a, 180 µg once weekly, and showed a sustained virologic response rate of 39% at 6 months after discontinuation of therapy, in comparison to 19% sustained response with standard interferon-a-2a three times weekly. [291] In another study, 1219 interferon-naive patients with chronic HCV infection were treated with peginterferon-a-2b weekly using a weight-based dosing of 1 or 1.5 µ/kg weekly and interferon-a-2b three times weekly in the control group for 48 weeks. [292] The sustained virologic response rates were 25% with pegylated interferon-a-2b, 1 µg/kg once weekly, 23% with pegylated interferon-a-2b, 1.5 µg/kg once weekly, and 12% with interferon- a-2b three times weekly. Therefore, monotherapy with either peginterferon has twice the efficacy of standard interferon monotherapy in terms of sustained virologic response. [291], [292], [293] and [294] The response rates were reduced in patients with genotype 1 and higher viral loads. Both peginterferons were tolerated as well as the standard nterferons, with discontinuation rates ranging from 6% to 11% in all treatment groups. Peginterferon-a-2b was approved and licensed by the U.S. Food and Drug Administration in February 2001, and approval of pegylated interferon-a-2a is expected in 2002. There are no clinical trials comparing the efficacy of the two peginterferons face to face; however, based on available data, it is expected that any differences between the two drugs are not significant. [291], [292], [293] and [294] The combination of peginterferon with ribavirin has yet further improved the efficacy of antiviral therapy of chronic HCV infection. [295], [296] Peginterferon-a-2b at a weekly dose of 1.5 µg/kg, plus ribavirin, 800 mg/day, showed a significantly higher response rate when compared with standard interferon plus ribavirin combination in a large phase III trial. [295] The overall sustained virologic response rate was 54% (genotype 1, SR 42%; genotype 2 or 3, SR 82%). The group treated with the peginterferon plus ribavirin combination reported higher frequency of fever, nausea, and injection-site reaction with erythema as compared with the control group. The rate at which the drugs were discontinued was similar in two arms of the study. The dose of ribavirin may not have been ideal, with higher doses achieving yet better results. A similar large, phase III study of peginterferon-a-2a plus ribavirin reported a sustained viral response rate of 56% (genotype 1, SR 46%; genotype 2 or 3, SR 76%). [296] These results are probably not significantly different than the results using peginterferon-a-2b, making these two drugs interchangeable. Therefore, based on these efficacy data, the use of pegylated interferon monotherapy is only appropriate if there are contraindications to ribavirin use. Based on data from these recent trials, peginterferon and ribavirin combination therapy will become the standard of care in treating patients with chronic HCV infection. [295], [296] Table 108-2 compares the sustained virologic response rates reported with interferon monotherapy, standard interferon plus ribavirin therapy, and treatment with peginterferon plus ribavirin.

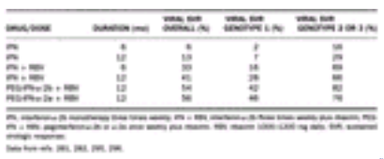

TABLE 108-2 Comparison: Sustained Virologic Response Rates in Treatment-Naive Patients

The use of pegylated interferon has also been investigated in patients with more advanced chronic hepatitis C. Peginterferon-a-2a monotherapy was studied in 271 patients with chronic HCV infection with bridging fibrosis or cirrhosis. [293] These patients were randomized to standard interferon-a-2a three times weekly; peginterferon-a2a, 90 µg once weekly; or peginterferon-a-2a, 180 µg once weekly for 48 weeks with a 6-month posttreatment follow-up period. [293] Intolerance to therapy was not significantly different in the three treatment arms of the study. The sustained virologic response rates with clearance of HCV RNA 24 weeks after treatment were 8%, 15%, and 30% in the three treatment groups, respectively. The biochemical sustained response rate and histological improvement were also better in the peginterferon groups. Thus, the efficacy of 180 µg of peginterferon-a-2a administered once weekly is significantly better than standard interferon-a-2a three times weekly in patients with HCV-related cirrhosis. [293] In summary, the evolution of interferon-based therapy for chronic hepatitis C, particularly combination therapy with ribavirin and pegylation of interferon, has resulted in incremental improvement in the sustained virologic response rate: (1) 10% to 15% with interferon monotherapy for 6 months; (2) 15% to 25% with interferon monotherapy for 12 to 18 months; (3) 40% with combination interferon and ribavirin; and (4) 55% with peginterferon and ribavirin.

Adverse Effects of Interferon and Ribavirin Common side effects associated with ribavirin and interferon treatment, most of which are mild to moderate but may necessitate discontinuation of therapy in 10% to 15% of patients, are listed in Table 108-3. [295], [296], [297] and [298] The major side effect of ribavirin is a dose-dependent hemolytic anemia, which is reversible and usually stabilizes after 5 to 6 weeks of treatment. If severe anemia develops, treatment must be discontinued.

Nonspecific
Flu-like symptoms, including headache, fatigue, rigors, and fever
Cutaneous
Injection-site reaction, pruritus, rash, dry skin, and hair loss
Respiratory
Cough and dyspnea
Gastrointestinal
Anorexia, nausea, vomiting, and dyspepsia
Neuropsychiatric
Depression, insomnia, anxiety, and irritability

TABLE 108-3 Adverse Effects: Interferon and Ribavirin Therapy

There are several contraindications to combination therapy with interferon and ribavirin (see Table 108-1). Patients with preexisting anemia usually cannot tolerate the degree of hemolysis that occurs with ribavirin therapy and it can be dangerous. Patients with significant cardiovascular disease are particularly at risk should severe anemia develop during therapy. Patients with chronic hepatitis C and premorbid conditions preventing the use of ribavirin, particularly anemia, can be treated with peginterferon monotherapy.

Treatment of Variant Clinical Manifestations

Acute Infection A 3-month course of interferon monotherapy at a dose of 3 MU three times weekly for 3 months is associated with higher virologic and biochemical end-of-treatment and sustained response rates compared with no treatment in patients with acute hepatitis C. [249], [253], [299], [300] In a metaanalysis, the virologic sustained response rate was 32% versus 4% in controls ($p < 0.001$). [300] A recent study of 44 patients with acute hepatitis C treated with interferon-a-2b, 5 MU daily for 4 weeks and then three times weekly for another 20 weeks, showed a remarkable 98% sustained virologic response rate 6 months after completion of therapy; that is, prevention of chronic hepatitis C. [301] Further studies are needed to confirm the role of induction with a higher initial dose and daily therapy versus combination interferon and ribavirin therapy or peginterferon therapy.

Children Several trials have examined the role of interferon monotherapy in children with chronic HCV infection and showed an overall sustained response ranging from 0% to 45%. [302] It is reasonable to extrapolate that combination interferon and ribavirin therapy may further improve the rate of HCV clearance. Further studies are needed to study the role of combination therapy in children with chronic HCV infection, particularly to focus on quality-of-life issues, including performance in school.

Normal Alanine Aminotransferase Levels As many as 25% of patients have persistently normal ALT levels in the setting of chronic HCV infection. [303] Fortunately, histologically advanced liver disease is rarely found in these patients. [304], [305] and [306] However, mildly abnormal liver histology may be encountered in these patients, and the reported rate of cirrhosis varies from 0.3% to as high as 10% in patients with normal ALT levels. [304], [307] In these patients, advanced fibrosis was associated with coexistent significant amount of alcohol consumption. [308] Some viremic patients with normal ALT levels develop elevation of ALT levels induced by interferon therapy. [306] The clinical significance of this ALT elevation is unclear. In a recent study, combination therapy with interferon and ribavirin for 6 months was associated with a 33% sustained response rate. [309] The recommendations from the NIH consensus conference in 1997 suggested that patients with normal ALT levels associated with chronic HCV infection should not be treated and followed conservatively. However, these guidelines were made when interferon monotherapy was the standard of care. The improved response rate with combination interferon and ribavirin therapy suggests that these guidelines should be modified, especially in patients with biopsy-proven moderate hepatic inflammation and fibrosis and in patients with genotypes 2 or 3. [309], [310] and [311]

Mixed Cryoglobulinemia and Glomerulonephritis Interferon monotherapy has been shown to be beneficial in the treatment for essential mixed cryoglobulinemia and glomerulonephritis associated with HCV infection. [211], [212], [312] Long-term therapy is usually required to prevent relapse of the underlying disorder. [210], [211] and [212] In the absence of significant renal impairment, combination therapy with interferon-a and ribavirin provide much better efficacy and should be the treatment of choice. [210] Other forms of therapy used in the management of HCV-induced cryoglobulinemic glomerulonephritis include cytotoxic therapy and immunosuppressive therapy. [213], [214] and [215] Cyclophosphamide and corticosteroids, often in combination with plasma exchange, have been used with some success. [213], [214] and [215]

Autoimmune Liver Disease Interferon-based therapy can precipitate a flare-up of coexistent otherwise quiescent autoimmune chronic hepatitis. It is recommended that patients with coexisting chronic autoimmune hepatitis and HCV infection should initially be treated with immunosuppressive therapy to prevent acute exacerbation of autoimmune liver disease. [313], [314]

Immunocompromised Status The efficacy of interferon-based treatment of HCV infection after liver transplantation, end-stage renal disease, or renal transplantation, and HIV co-infection is suboptimal but needs further investigation. [315], [316] There are currently no published guidelines on how to best manage patients who have HCV and HIV co-infection. End-stage liver disease is now the leading cause of death in hospitalized HIV-seropositive patients. [317] Because of similar routes of transmission, an increasing number of patients with either HCV or HIV are being identified as co-infected with both viruses. It has been shown that HCV and HIV co-infected patients have a faster progression rate (up to threefold) of liver fibrosis than HCV-positive patients without HIV. [318] There is a plethora of data now indicating that both HCV RNA levels and survival are directly influenced by HIV infection. These results suggest that HIV infection-induced CD4 depletion is independently associated with severity of liver fibrosis in chronic HCV infection. [318], [319] Because data are limited, a reasonable approach to co-infected patients is to enroll such patients in ongoing clinical trials. At least two of the protease inhibitors used for the treatment of HIV infection, ritonavir and indinavir, can cause serious liver toxicity. [320] Ritonavir leads to late liver toxicity, normally due to triglyceride abnormalities. Indinavir, on the other hand, usually causes only a Gilbert-like syndrome, although in some cases, it may cause a more significant toxicity. The protease inhibitors, saquinavir and nelfinavir, have only minimal liver toxicity. The use of highly active antiretroviral therapy has prolonged the healthy lifespan of patients infected with HIV, and deaths among individuals with acquired immunodeficiency syndrome have declined for the first time in 1996 with the institution of this therapeutic approach. [321]

Novel Investigational Treatment Strategies and Agents

High-dose, initial induction therapy of chronic HCV infection with daily interferon has been suggested as a potentially efficacious regimen. [322], [323] Because of the rapid HCV replication rate, early intervention with an aggressive high-dose interferon regimen may prove the most effective way to prevent HCV persistence.

The role of amantadine in the treatment of HCV as a single agent, [324] in combination with interferon, [325] and as triple therapy with interferon, ribavirin, and amantadine has been evalua- ted. [326] Amantadine monotherapy is not efficacious, [324] but triple therapy has shown some promise. [326] The combination of interferon with other novel agents, such as thymosin-a-1, macrophage colony-stimulating factor, ursodeoxycholic acid, and iron-depleting measures has been studied, but showed minimal or no benefit compared with interferon monotherapy. [327], [328], [329] and [330] Silymarin, extracted from the milk thistle plant, is safe and undergoing study for the treatment of chronic hepatitis C. [331]

Preventive and Safety Measures

Patient should be informed that, although the risk for transmitting HCV infection by sexual contact is low (3% to 5%), HCV is potentially transmissible. Patients should be careful about blood exposure of any type to partners and family contacts. Open wounds must be covered, and razors or toothbrushes should not be shared. Although sexual or intrafamilial HCV transmission is rare, testing sexual partners or other family members if there is a concern regarding infection usually provides reassurance. Patients with chronic HCV infection should be counseled to avoid excessive consumption of alcohol, which can accelerate the progression of hepatitis C when used in moderate or large amounts. It has been recommended that the less alcohol consumed the better and that complete abstinence is ideal. [250], [251] and [252] Finally, it is recommended that hepatitis A and B vaccine be given to patients with chronic HCV infection who are not immune because acute hepatitis A or B superimposed on chronic HCV infection may be more severe. [332] The safety and efficacy of these vaccines have been demonstrated in patients with mild to moderate chronic HCV infection, [333] and a more convenient combined hepatitis A and B vaccine was licensed in 2001. The inability to culture the virus and its genetic heterogeneity are obstacles to developing a vaccine against HCV infection. [334] Investigational vaccination candidates, including recombinant proteins, peptides, virus-like particles, naked DNA, and recombinant viruses, are being explored. [334]

Economic Implications

Current estimates of medical and work loss costs of HCV-related acute and chronic liver disease are in excess of $600 million annually based on unpublished CDC data. In 1995, the outcomes of hospitalizations were analyzed, focusing primarily on inpatient mortality and health care cost. [335] It was estimated that HCV-related liver disease resulted in 26,700 hospitalizations and 2600 deaths in acute, nonfederal hospitals in the United States. The cumulative cost of these hospitalizations was $514 million.

Cirrhosis The sustained response to interferon monotherapy, 3 MU three times weekly for 6 to 12 months, ranged from 5% to 10% in patients with HCV cirrhosis. [336] The sustained virologic response was genotype dependent, that is, 4% in genotype 1 and 10% with genotypes 2 and 3. [337] Results from a metaanalysis of six randomized controlled trials of combination interferon and ribavirin therapy versus interferon monotherapy in 75 patients with compensated cirrhosis showed a 22%

sustained biochemical response with combination therapy in comparison to 4% with interferon monotherapy. [338] Patients with cirrhosis who received combination therapy had a significantly higher sustained virologic response (17% versus 0%). The sustained response was genotype dependent (7% to 10% with genotype 1 versus 24% to 33% with genotypes 2 and 3). The overall data are suggestive of a better outcome of treatment of HCV in cirrhotic patients. [339] Interferon-based therapy reduces the rate of progression of fibrosis and lowers the incidence of HCC by 13% and the mortality rate from HCC by 16%. [339] The persistence of serum HCV RNA after 24 weeks of combination antiviral therapy was predictive of a lack of sustained virologic response in patients with cirrhosis and also in those without cirrhosis. [281], [282] The use of maintenance interferon therapy has been associated with a decline in the incidence of HCC in patients with compensated cirrhosis, even in the absence of end-of-treatment or sustained virologic responses. [188], [340], [341] and [342] Other complications of end-stage liver disease may also be retarded with long-term interferon use in patients with chronic hepatitis C and compensated cirrhosis. [188] The efficacy of 180 µg of peginterferon-a-2a is significantly better than that of standard interferon-a-2a given three times weekly in patients with HCV-related cirrhosis. [293] Patients who eradicate HCV theoretically should have a better prognosis after liver transplantation; however, antiviral therapy for HCV infection is poorly tolerated in decompensated patients. Therefore, interferon therapy alone or interferon plus ribavirin therapy in decompensated liver disease should only be used in the setting of a clinical trial. [343], [344]

Liver Transplantation

In the United States, about 8000 patients die each year from complications of HCV-related cirrhosis. [13], [16] Decompensated chronic hepatitis C with cirrhosis, alone or in combination with alcoholic liver disease, is the most common indication for liver transplantation in the United States and Europe. [345] Based on statistics obtained from the United Network for Organ Sharing (UNOS), 23% of patients who underwent liver transplantation between 1994 and 1998 carried the diagnosis of HCV infection. [346] There has been a dramatic increase in the number of patients with HCV infection undergoing liver transplantation, and HCV currently accounts for up to 50% of liver transplantations both in the United States and Europe. [347]

Candidacy for Liver Transplantation Patients who are being considered for liver transplantation must meet one of the following minimal listing criteria: (1) Child-Pugh class B or C (Child-Pugh score of 7 or greater) in the setting of cirrhosis; (2) history of gastrointestinal bleeding caused by varices, or a single episode of spontaneous bacterial peritonitis irrespective of Child-Pugh score; or (3) small unifocal or multifocal HCC (one lesion less than 5 cm in diameter or three lesions less than 3 cm involving the same lobe of the liver) with no portal vein invasion or metastases. [348] Patients who fulfill the UNOS specified listing criteria for liver transplantation have an estimated 1-year survival rate from chronic liver disease of less than 90%, which is the likely outcome with liver transplantation at major institutions. [349] In patients with compensated HCV-related cirrhosis, the 4- to 5-year survival rate is 91%, and the estimated risk for decompensation is 18% to 20%. [188], [350] Liver transplantation is the best treatment for chronic hepatitis C with decompensated cirrhosis, but HCV reinfection poses challenging management issues that may arise either early or late after transplantation. [351], [352]

POSTTRANSPLANTATION RECURRENT INFECTION

Recurrent HCV infection presenting as acute lobular hepatitis is documented in 75% of patients by liver biopsy by 4 months after transplantation. [347], [353] Most patients (80% to 85%) with recurrent HCV infection develop mild liver disease without life-threatening complications in the short- to medium-term follow-up. The remaining 15% to 20% of patients can have a more severe course with rapidly progressive disease resulting in cirrhosis and decompensation. [347], [353] Recurrent HCV infection can develop anytime during the first year after liver transplantation, but rejection is more common in the first 2 months. A liver biopsy must be performed to differentiate recurrent hepatitis C from rejection based on certain histological characteristics. [354], [355] and [356] Recurrent HCV infection is universal after transplantation, and thus measurement of serum HCV RNA is not helpful.

Risk Factors

The lack of HCV-specific T-cell responses is a crucial underlying defect that results in the pathogenesis of HCV-related graft injury after transplantation. [357] Humoral immunity does not play a role in preventing recurrent HCV after transplantation. [358] A variety of host, viral, and immunosuppressive therapy responses are associated with recurrent HCV infection after transplantation [359], [360], [361], [362], [363], [364], [365], [366], [367], [368], [369], [370], [371], [372], [373], [374], [375], [376], [377], [378] and [379] (Table 108-4). The association of HCV genotype 1b with rapidly progressive liver disease after transplantation has been noted by some investigators, [352], [366], [371], [379] but this observation has not been confirmed by others. [379] High pretransplantation serum HCV RNA levels [360], [363] and early detection of high posttransplantation serum HCV RNA levels were associated with more severe recurrent HCV infection. [364], [377] Fibrosing cholestatic hepatitis is a severe life-threatening variant of recurrent posttransplantation HCV infection associated with very high viral loads. [380] Pretransplantation HCV quasispecies may not be detectable postoperatively but can predict severity of recurrent HCV after transplantation. [381] Increasing age and absence of pretransplantation co-infection with HBV are associated higher allograft reinfection. [366] The posttransplantation course in patients with HCV, HBV, and HDV co-infection has been associated with a slower replication rate of HCV and an innocuous histological necroinflammatory response. [184] Although HGV has been shown to persist after transplantation, it has no influence on the course of recurrent hepatitis C. [382] The role of cytomegalovirus (CMV) and posttransplantation recurrence of HCV is controversial. In the initial 12-month period after transplantation, neither CMV viremia nor CMV disease was noted to have any influence on the histological outcome of HCV recurrence. [383] On the other hand, some observations have associated CMV with the severity of recurrent HCV infection. [359], [369], [375] Reducing the level or dosage of immunosuppression more rapidly than usual after transplantation is crucial to limit HCV replication. [368], [372] High-dose and potent immunosuppressive therapy, including the number of methylprednisone boluses, cumulative steroids dose, or use of OKT3 or mycophenolate, are associated with severe recurrence and rapid progression of recurrent hepatitis C. [361], [377] Extended rewarming time during allograft implantation is associated with higher incidence of recurrent HCV infection and should be minimized. [383]

Host
HLA matching
Age
Nonwhites
Cytomegalovirus co-infection
Absence of pretransplantation co-infection with HBV
Presence of IgM anti-HCV core antibodies after transplantation
Viral
Genotype 1b
Pretransplantation HCV quasispecies
High (> 1 × 10^6 Eq/mL) pretransplant HCV RNA levels
High serum or liver, early posttransplantation HCV RNA viral load
Allograft
Prolonged rewarming time during allograft implantation
Year of transplantation (recent year associated with rapid rate of progression)
Early posttransplantation histological injury
Episodes of rejection
Drugs
High level of immunosuppression
Mycophenolate use
OKT3 use
Methylprednisone boluses
High cumulative steroid dose

TABLE 108-4 Risk Factors: Severe Recurrence of Hepatitis C after Transplantation

Natural History

The observations from short- to medium-term (5 years or less) follow-up studies in patients with recurrent HCV infection after transplantation showed survival rates that were comparable to those in patients who underwent liver transplantation for other indications. [352], [384] It is suspected that longer follow-up after recurrent HCV infection of the allograft will demonstrate a decline in survival rate. [181]

Reinfection with HCV after transplantation is universal in patients with detectable serum HCV RNA before transplantation. [351], [354], [385], [386] Up to 70% of patients with pretransplantation viremia can develop graft hepatitis after transplantation. [351], [352], [386] Data from programs with annual protocol liver biopsies suggest that 20% to

28% patients will develop HCV-related cirrhosis by 5 years after transplantation. [352], [373] Liver biopsy performed at the end of the first year after transplantation is predictive of the future risk for cirrhosis. [352], [373] Less than 10% of patients with mild hepatitis at 1 year showed evidence of cirrhosis at 5 years, whereas two thirds of patients with moderate hepatitis at 1 year progressed to cirrhosis at 5 years. [347], [373], [376] On the other hand, studies with favorable outcomes have been reported with posttransplantation graft and patient survival rates in patients with recurrent HCV infection that are similar to those of uninfected controls. [352], [363], [366], [387], [388] The HCV serologic status after transplantation did not affect 10-year graft or patient survival rates. [387]

The cumulative probabilities of decompensation in patients with recurrent HCV-related allograft cirrhosis were 8%, 17%, and 42% at 1, 6, and 12 months, respectively, after the diagnosis of cirrhosis. Graft and patient survival rates were 100%, 85%, and 71%, and 100%, 92%, and 74% at 1, 6, and 12 months, respectively. Patient survival rates decreased significantly after decompensation (93%, 61%, and 41% at 1, 6, and 12 months, respectively). Variables associated with decompensation, retransplantation, and mortality included a high Child-Pugh score (more than 6), low levels of serum albumin, and a short interval between liver transplantation and posttransplantation cirrhosis.

Treatment

In contrast to the success in preventing HBV reinfection after liver transplantation, there is as yet no effective antiviral strategy to prevent HCV reinfection. [389], [390] Recurrent hepatitis C may be accelerated in immunosuppressed liver transplant recipients, progressing to cirrhosis in 8% to 16% of patients within 5 years in some reports. [352], [391] The rapid rate of progression to liver failure has been observed in a subset of HCV-infected transplant recipients who develop a cholestatic pattern of liver injury. [368], [372], [392] Cholestatic hepatitis is an infrequent cause of jaundice in HCV-infected liver transplant recipients and should be a diagnosis of exclusion. [354], [380] Cholestatic hepatitis C ranges in severity and does not always signal impending graft failure. The natural history of cholestatic hepatitis C differs dramatically from that of cholestatic hepatitis B, which has been shown to lead to death or need for retransplantation within several weeks. [393]

Antiviral Therapy The use antiviral therapy can be preemptive (prophylactic) or after histologically documenting recurrent hepatitis C. The argument in favor of preemptive antiviral therapy is based on the fact that recurrent allograft infection with HCV is histologically noted in more than 50% patients after transplantation. The negative consequence of preemptive interferon-based therapy is the immunomodulatory effect of interferon that may be associated with an increased risk for acute allograft rejection. Interferon has been shown to up-regulate expression of HLA antigens in the allograft, resulting in an increased incidence of rejection in up to 35% of recipients after transplantation. [394] The experience with preemptive antiviral therapy consists of anecdotal case reports and uncontrolled, nonrandomized studies with a limited number of patients. Multicenter, controlled randomized trials are needed to assess the benefit of preemptive antiviral therapy. Treatment of histologically documented recurrent HCV infection with interferon has resulted in a biochemical response with normalization of serum ALT levels in only 10% to 30% of patients. [394], [395] and [396] Patients with low viral loads have higher rates of favorable response. [352] Sustained biochemical and virologic responses are less likely due to the ongoing viral replication facilitated by immunosuppression. Interferon and ribavirin combination therapy has demonstrated improved efficacy as compared with interferon monotherapy in the treatment of patients with recurrent HCV infection after transplantation. [375]

Judicious Immunosuppressive Therapy The intensity of immunosuppression defined by the potency and level of immunosuppressive agents can play a pivotal role in preventing both recurrence of HCV and rejection of allograft. [397] There was no significant difference in graft and recipient survival rates with cyclosporine versus tacrolimus-based induction therapy. [352], [363], [398] Treatment of allograft rejection should be established by performing a liver biopsy. Trial of antirejection therapy must be avoided in patients with inconclusive liver biopsies. It is recommended that the least aggressive antirejection therapy be instituted in patients with documented rejection. The risk for severe, rapidly progressive HCV recurrence in allograft recipients is associated with increasing number of methylprednisolone boluses, mycophenolate use, and OKT3 use. [361], [373], [374], [377], [399]

Retransplantation The number of HCV-related liver transplant recipients who develop allograft failure requiring retransplantation rose from 6.5% in 1990 to 38.4% in 1995. [356] Most patients develop allograft cirrhosis and decompensation due to recurrent HCV infection. Other causes of graft failure may include chronic rejection, biliary complications, and hepatic artery thrombosis. [356] The initial data with liver transplantation in patients with end-stage liver disease secondary to chronic HCV infection demonstrated that less than 10% develop severe allograft damage requiring retransplantation. [366], [388] Unfortunately, for reasons that are not completely clear, current experience and a multicenter trial show a decline allograft survival, with up to 20% of patients developing cirrhosis within 5 years after transplantation. [352], [359], [373], [388] After retransplantation, the 12-month survival rate was less than 50% in patients who developed allograft failure due to recurrent HCV infection. [400] Severe hyperbilirubinemia and renal failure were associated with poor graft survival after retransplantation. [401], [402] Patients who underwent retransplantation before the onset of severe hyperbilirubinemia or renal failure had a 12-month graft survival rate of greater than 75%. [401], [402] and [403] Multicenter, controlled randomized are needed to identify prognostic markers associated with improved graft and host survival after retransplantation.

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CHAPTER 109

William M. Lee and Frank V. Schiødt

DRUG-INDUCED LIVER DISEASE

EPIDEMIOLOGY

Drug-induced hepatotoxicity is a frequent cause of acute liver injury of exceptional severity, comprising more than 50% of all cases of acute liver failure (ALF) in the United States [1], [2] (Fig. 109-1). Hepatotoxicity has been described for a large number of drugs, [3] although the number of cases is quite low, given the number of prescriptions written.

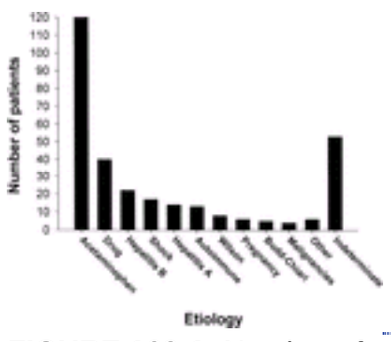


FIGURE 109-1. Number of cases of acute liver failure among 308 patients grouped according to etiology in a multicenter study between 1998 and 2001 at 14 sites around the United States participating in the Acute Liver Failure Study Group. Acetaminophen and idiosyncratic drug reactions were the presumed cause in 160 (52%) cases of acute liver failure in this series.

Different agents cause liver injury in different ways at different rates. The majority of reactions are directed against hepatocytes but biliary injury as well as combined hepatocyte/biliary injury or damage to specific organelles (e.g., mitochondria) produce the different disease patterns observed. While some agents such as isoniazid cause liver injury in as many as 1 in 100 people and fatality in 1:10,000, other agents result in liver damage in only 1:50,000 or may never cause liver injury.

Few data on the epidemiology of drug-induced liver disease are available. A national European registry on adverse drug reactions described an almost doubling of reported cases of drug-induced liver disease from the decade 1968–1978 [4] to the following decade 1978–1987, the last period accounting for 22 cases per million people per year. [5] In developing parts of the world, drug-induced liver disease is much less common and related to very few drugs. [6] However, probably only a small fraction of actual cases are reported, and a true estimate of the incidence of drug-induced liver disease may be impossible to obtain. This is due in part to the difficulty in establishing the diagnosis, as well as inadequate reporting systems.

The exact number of drug-induced liver injuries per year in the United States is unknown, but the severity of many of these cases and the tragedy involved in a presumed preventable injury makes it imperative that all sensible precautions be taken to avoid such incidents.

DRUG METABOLISM AND MECHANISMS OF HEPATOTOXICITY

The liver, situated between the absorptive surface of the gastrointestinal tract and target of drug effects throughout the body, is central to the metabolism of foreign substances. Since hepatocyte metabolism is required for virtually every drug, it is remarkable how seldom injury to liver cells occurs. Most drugs and xenobiotics cross the intestinal brush border because they are lipophilic. *Biotransformation* is the process whereby lipophilic therapeutic agents are rendered more hydrophilic by the hepatocyte, resulting in drug excretion in urine or bile. In most instances, biotransformation changes a nonpolar to a polar compound through several steps. Foremost is an oxidative pathway (e.g., hydroxylation) mediated by the cytochromes P450 (CYPs). [7] This is typically followed by esterification to form sulfates and glucuronides, which results in addition of highly polar groups to the hydroxyl group. These two enzymatic steps are referred to as phase I (P450 oxidation) and phase II (esterification). Other important metabolic pathways involve glutathione S-transferase, acetylating enzymes, and alcohol dehydrogenase, but the principal metabolic pathways for most pharmacological agents involve P450 and subsequent esterification.

The exact details of the pathogenesis of liver injury remain unclear for most drugs. A single drug may cause its toxic effects in several ways. An oversimplified approach suggests that high-energy unstable metabolites of the parent drug, the result of P450 activation, bind to cell proteins or DNA and disrupt cell function. Perhaps the best example is acetaminophen. Although used universally for nonnarcotic pain relief, acetaminophen taken in large quantities causes profound centrilobular necrosis. [8] The metabolic pathway for acetaminophen involves both phase I and phase II reactions, glutathione detoxification, and the formation of reactive intermediates (Fig. 109-2). Glucuronidation and sulfation occur as the initial detoxifying step since the parent compound contains a hydroxyl group. Since glucuronidation and sulfation capacity greatly exceeds daily needs, even patients with far-advanced liver disease continue to have adequate glucuronidation capacity, which explains why no obvious enhancement of toxicity is observed in patients with cirrhosis taking acetaminophen. [9]

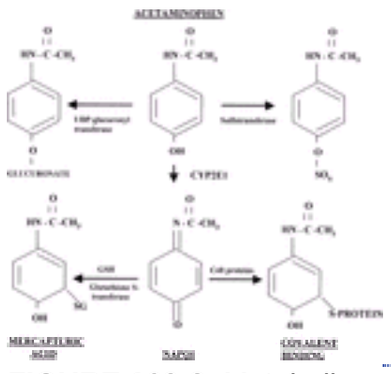


FIGURE 109-2. Metabolic pathway for acetaminophen

Enzyme Polymorphism

The rarity of drug toxicity begs the question of how an infrequent event (1:10,000) occurs. Genetically variant CYP iso-enzymes, such as are observed with metabolism of debrisoquine, partially explain observed individual variation in responses to drugs. Debrisoquine is an antihypertensive drug marketed in Europe which is hydroxylated by CYP2D6, an iso-form that is totally lacking in 5% of healthy individuals, greatly prolonging the half-life of the parent compound in affected individuals. [10] Fast and slow acetylator patterns are observed to affect whole races, and have been implicated in isoniazid metabolism, which includes an acetylation step. [11] Genetic variants, which occur relatively frequently, cannot explain the formation of a toxic intermediate in only a rare individual. While there might be other metabolic variant P450 species that are even rarer, little evidence for these has been found in affected patients. Other explanations are necessary.

Most drugs are small organic compounds that are unlikely to evoke an immune response. While some toxic drug reactions are associated with an obvious allergic response, most are not. Nevertheless, immune mechanisms not associated with systemic allergic immunoglobulin E (IgE) reactions or skin hypersensitivity might be involved. Studies suggest that the very products of CYP metabolism, the highly reactive intermediates formed within the microsomes, covalently bind to the enzyme itself to form a drug-hapten adduct that disables the enzyme and injures the cell. [12] Haptenization then evokes an immune response directed against the newly formed antigen or neo-antigen. P450s have been shown to traffic to the plasma membrane allowing the P450-drug adduct to become the target of a subsequent cytolytic attack (Fig. 109-3). Whether these adducts or smaller peptides processed and presented via the major histocompatibility complex (MHC) class I and class II schemes are the targets remains unclear. Still, the association of neo-antigens, autoantibodies, and hepatotoxic drugs implicates an immunologic mechanism.

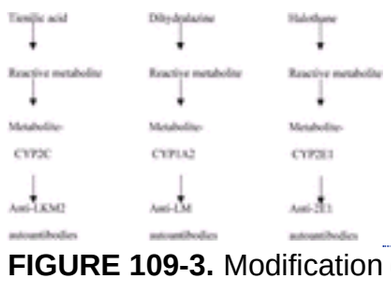


FIGURE 109-3. Modification of host P450 enzymes renders them immunogenic. Autoantibodies can be detected, which recognize the enzyme-metabolite adducts and the enzyme itself. (Adapted from ref. [12].)

Whether the drug causes significant cell necrosis or not, the P450-drug adducts can evoke the immune response. Any subsequent P450-drug adduct present on the hepatocyte surface would evoke a further response. Responses may be antibody-mediated or occur from direct cytolytic attack by primed T cells [12], [13] (Fig. 109-4).

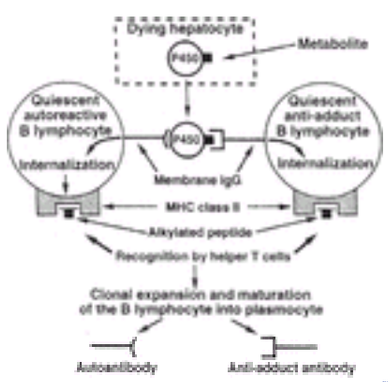


FIGURE 109-4. Possible models of liver injury due to drugs. Direct toxicity may play a role but immune mechanisms are also operative. In this model, re-challenge or continuing to receive the same drug would simply elicit a preformed cytolytic response. (From ref. [12].)

Such a combined toxic/immunologic mechanism is involved in the liver injury caused by halothane. Halothane was a widely used fluorinated hydrocarbon anesthetic which causes severe, often fatal liver injury after multiple exposures. [13] Other fluorinated hydrocarbons also occasionally result in the same response. [14], [15] and [16] While halothane has never been withdrawn, its use has been limited by the advent of safer agents. Both direct cytotoxicity and immune-mediated toxicity are observed in keeping with the clinical observations, that severe halothane toxicity occurs with repeat exposures. Still, evidence of injury can usually be identified within a week of the first exposure. As befits an immune reaction, the interval to toxicity is shortened and the damage more severe with each successive exposure.

Specific genetically determined components of the immune response may be important. For example, the binding of peptides for antigen presentation depends on human leukocyte antigen (HLA) configurations that are genetically determined. This variation among individuals is thought to underlie the diverse responses observed in patients encountering the hepatitis B virus. The highly variable severity of reactions observed depends on the fit of antigen peptides in the HLA groove. A specific HLA haplotype has been associated with amoxicillin-clavulanate–induced hepatitis, being found in 57.1% of patients versus 11.7% of controls. [17] Polymorphisms have also been identified for the interleukin 10 (IL-10) promoter and for tumor necrosis factor-a (TNF-a). These variations in immune responsiveness could modulate the severity of the responses observed. For example, different IL-10 promoter phenotypes are recognized. A C-to-A substitution at position 627 is linked to severe asthma and, by inhibiting IL-10 secretion, an up-regulation of immune reactions of the Th2 type. This same phenotype has been linked to hepatitis C-related liver injury and to the severity of alcoholic liver injury. [18], [19] Variant TNF-a phenotypic expression has been implicated in determining the severity of drug reactions related to acetaminophen. [20] A multistep, immune-based mechanism would best explain both the rarity of idiosyncratic reactions, and their severity, as well as the findings of mild, nonprogressive liver injury in some patients—those with “protective” phenotypes. While an immunologic explanation for many reactions is plausible, the exact mechanism to account for most drug reactions remains obscure. Both cell necrosis and apoptosis have been recognized, and sinusoidal lining cells as well as Kupffer cells are part of the process. [21]

There should be little doubt that the metabolic fate of any compound is a complex process. There are other important environmental and host variables outlined in Table 109-1. Often multiple factors are at play simultaneously, including drug interactions, either induction or competition. Common inducing agents include ethanol, phenobarbital, and phenytoin, but cigarette smoke is also a potent inducer of certain P450 species. Induction or substrate competition for available enzyme may not result in hepatotoxicity but strongly impacts plasma drug levels. For example, the effect of ketoconazole on enhancing cyclosporin levels is the result of induction, [22]

while competitive inhibition by ketoconazole increases serum levels of astemizole (Hismanal) with resulting torsades de pointes. [23]

Age:	Drug reactions appear to impact the elderly more often, P450s vary.[127] Adults more susceptible than children in some scenarios (acetaminophen, halothane, isoniazid) and less susceptible in others (e.g., aspirin, valproic acid).
Gender:	Females are more prone to drug reactions statistically, mechanism unknown.
Size/weight:	Effects on women relate to intrinsic gender differences but also to size.
Pregnancy:	Effects of drugs in pregnancy have been poorly studied.
Preexisting Liver Disease:	Hepatic disease may *protect* against idiosyncratic reactions and may enhance toxicity in dose-dependent hepatotoxins (such as acetaminophen).[126]
Renal Disease:	Slowed disappearance of parent compound yields higher concentrations and affects P450.[128] Tetracycline toxicity is enhanced in renal disease.
Certain Foods:	Grapefruit has an unknown substance that interferes with metabolism.[129,130]
Concomitant Drugs:	Drug-drug interactions are common causes of adverse effects (e.g., valproate and chlorpromazine together lead to enhanced cholestasis).
Genetic Factors:	Enzyme polymorphisms (e.g., enhanced phenytoin liver disease in patients with defective epoxide hydrolase activity), HLA phenotypes (nitrofurantoin susceptibility).

HLA, human leukocyte antigen.

TABLE 109-1 Factors Influencing the Metabolic Fate of Drugs

Other Mechanisms

In drug-induced cholestasis, disruption of specific transport proteins or processes in hepatocytes or cholangiocytes may be the event that results in cholestasis. Bile salt transport from plasma into the hepatocytes is provided for by two basolateral (sinusoidal) transport systems: the sodium-taurocholate cotransporter (NTCP), and the organic-anion transporting polypeptide (OATP), whereas several canalicular export pumps have been identified. [24] Estrogen may cause multiple canalicular membrane transport changes, [25] affecting, among others, the canalicular bile salt pump. [26]

Uncoupling or inhibition of mitochondrial respiration may in some instances lead to microvesicular steatosis. [27] Mitochondrial ß-oxidation of fatty acids is impaired and this may decrease cellular energy supply leading to severe liver dysfunction. The mitochondrial ß-oxidation may be affected either directly or by impairment of mitochondrial respiration. [28]

Drug-induced liver injury may be modulated and enhanced by inflammatory mediators that may trigger hepatocyte apoptosis. [29] Hepatocyte apoptosis is complex and may be controlled by the intracellular energy status [30], [31] and by the redox state of the cell. [32] How the specific drugs involved in hepatotoxicity affect hepatocyte apoptosis remains to be studied.

The new techniques of pharmacogenomics may be helpful in predicting an individual's risk of hepatotoxicity for a given drug. [33], [34]

CLASSIFICATION OF HEPATOTOXIC AGENTS

Two main categories of drugs can produce liver disease. One group consists of intrinsic (predictable) drugs, whereas a second group consists of idiosyncratic (unpredictable) drugs. Unfortunately, the vast majority of drugs involved in liver disease belong to the idiosyncratic, and thus unpredictable, group.

Intrinsic (Dose-Dependent) Agents

Hepatotoxins of this group produce liver disease in most patients in a dose-related fashion if toxic amounts of the drug are ingested. Furthermore, similar lesions can often be found in animal models. [35] Hepatotoxicity may be caused by the drug itself or, most frequently, by toxic effects of its metabolites. Table 109-2 provides a list of known dose-dependent drugs.

DRUG	RESPONSE
Acetaminophen	Total dose, single vs. multiple time points
Amiodarone	Total dose over time
Bromfenac	Toxicity only occurs after extended use
Cocaine	Dose-related vascular collapse
Cyclophosphamide	Dose-related, worse with previous ALT elevations
Cyclosporine	Cholestasis with toxic blood levels, CYP3A phenotype
Methotrexate	Aminotransferase/fibrosis; single dose/total dose
Niacin	Large doses yield vascular collapse
Oral contraceptives	Prolonged usage yields hepatic adenomas
Tetracycline	Total dose, renal dysfunction
Toxins (yellow phosphorus, carbon tetrachloride, amanita toxin, bacterial toxins)	Total dose

ALT, alanine aminotransferase.

TABLE 109-2 Drugs/Toxins in Which a Dose-Response Effect Is Observed

Until recently, occupational hepatotoxic agents, such as carbon tetrachloride [36] and yellow phosphorus, [37] were frequently encountered. In the last two decades, however, these agents were rarely seen. Instead, acetaminophen (paracetamol) has emerged as the most dominant intrinsic hepatotoxic drug. Taken in small doses (4 g or less per day) acetaminophen is an extremely safe drug. However, its therapeutic index is low since only as little as 10 to 12 g may cause extensive hepatic necrosis. [38], [39]

In acetaminophen metabolism, the phase II reactions predominate, with only a small fraction of acetaminophen metabolized by cytochrome P450, until the quantity of acetaminophen exceeds phase II capacity, at which point significant amounts of a toxic intermediate, *N*-acetyl-p-benzoquinoneimine (NAPQI), are formed primarily via CYP2E1 (see Fig. 109-2). [40], [41] NAPQI binds covalently to cell macromolecules disrupting mitochondrial and nuclear function. [35] Antibodies to nitrotyrosine residues can be detected as evidence of oxidative stress in livers of patients (or experimental animals) demonstrating toxicity. [42] These residues are formed by the rapid reaction of superoxide and nitric oxide formed by Kupffer cells reacting to form peroxynitrite, unless covalent bonding of NAPQI is prevented by its conjugation (via glutathione-S-transferase) to form mercapturic acid, a harmless water-soluble product excreted by the kidney. [43] Depletion of glutathione lowers this last defense against the formation of NAPQI-related intracellular adducts. Thus, starvation and chronic alcohol intake by depleting glutathione enhance toxic injury, [44], [45], [46], [47] and [48] while *N*-acetylcysteine, by replenishing glutathione, protects against acetaminophen-induced injury. [49] This direct toxic reaction occurs predictably in all individuals and is not an allergic reaction. The final step leading to cell death remains unclear but may involve an increase in levels of cytosolic calcium altering the cytoskeleton and membrane integrity, leading to "blebbing" of the cell membrane and loss of its integrity. [50] Dose-related necrosis (lysis) of hepatocytes occurs but apoptotic pathways are also implicated. A finding that peroxisomal proliferator activation prevents the liver injury associated with acetaminophen links this liver damage to apoptosis but does not preclude a combined necrosis/apoptosis effect. [51], [52]

Acetaminophen overdose is the most common cause of acute liver failure with hepatic encephalopathy in several Western countries including the United Kingdom and the United States. [39], [53], [54] Although the prognosis for acetaminophen-induced acute liver failure is relatively good, with a spontaneous survival (i.e., survival

without liver transplantation) of approximately 57%. [1] It is still the leading cause of acute liver failure death of in the United States. [2]

Except for acetaminophen-induced liver disease, intrinsic drug cases are rare.

Idiosyncratic Reactions

While acetaminophen is a dose-related toxin, most drug reactions are idiosyncratic, occurring from 1 in 1000 to 1 in 50,000 patients. The etymology of “idiosyncratic” from the Greek loosely translated is “the unique composite of the self”—the particular features of a given individual. This places the emphasis appropriately on the patient’s characteristics rather than on the drug itself. Idiosyncratic reactions are not due to the drug itself, since almost everyone can tolerate them, but to something unique about the patient who ingests them and gets a toxic reaction. Theories abound to explain these reactions. Any theory of pathogenesis must “explain” the features shown in Table 109-3. The proportion of drug-induced liver disease varies greatly among drug classes as evidence of class effect. Nonsteroidal agents, antibiotics, and anticonvulsants are highly associated with drug-induced liver disease, whereas hormones, antihypertensive drugs, digoxin, and antiarrhythmic drugs are very rarely associated. Idiosyncratic reactions occur in small numbers such that some drugs continue to be used when usefulness or uniqueness makes the risk acceptable. Isoniazid is such a drug, virtually the only drug implicated in developing countries, where drug-induced liver injury is otherwise unheard of. [55] Some 15% to 20% of individuals receiving isoniazid as a single agent for tuberculosis prophylaxis may develop increased transaminases, but these usually stabilize or improve, so that less than 1% may develop severe hepatic necrosis. [56] More recently, a lower estimate of severe liver injury of 1:1000 has been given for isoniazid from a large tuberculosis public health clinic, [57] since 11 of 11,141 patients developed isoniazid-induced liver disease. This is still a high rate of injury compared to other idiosyncratic drug reactions, yet the usefulness of the drug has precluded its withdrawal.

1. Occur rarely
2. A pattern consistent for each drug
3. Similar drugs exhibit similar features called “class effects”
4. Individual drugs in a class still vary considerably
5. Reactions occur at varying time intervals after beginning ingested
6. Reactions vary in severity, but typically severe and fatal if drug continued
7. Mild injury can sometimes disappear with continued use
8. Rarity of most reactions suggests possibility of multiple hits
9. Re-challenge is virtually always met with greater severity, shorter latency

TABLE 109-3 Idiosyncratic Drug Reactions

Aside from isoniazid, nonsteroidal analgesics may be the most commonly associated drug class capable of inducing idiosyncratic reactions. [58] The newer cyclooxygenase-2 (COX-2) inhibitors have been implicated. [59], [60] It is important to recognize that certain classes are known to be associated with toxic injury while others are much less likely.

TYPES OF DRUG REACTIONS, CLINICAL PICTURES

While most liver injury involves direct hepatocyte necrosis/apoptosis, some drugs primarily injure bile ducts or canaliculi, causing cholestasis without significant hepatocyte damage. Others affect sinusoidal cells or present a particular pattern of liver injury affecting multiple cell types (mixed type). In a rough way, drug reactions can be grouped as hepatocellular, cholestatic or mixed, but these are only very general terms and do not apply to all circumstances. An additional way to categorize drug reactions emphasizes the histological changes involved as well as the cell type (Table 109-4).

Hepatocellular: isoniazid, trazodone, diclofenac, nefazodone, venlafaxine, lovastatin
Cholestatic: chlorpromazine, estrogen, erythromycin
Mixed: amoxicillin/clavulanate, carbamazepine, herbs, cyclosporin, methimazole
Immunoallergic: halothane, phenytoin, sulfamethoxazole
Granulomatous: allopurinol, diltiazem, nitrofurantoin, quinidine, sulfa drugs
Steatohepatitis: amiodarone, perhexiline maleate, tamoxifen
Autoimmune: nitrofurantoin, methyldopa, lovastatin
Fibrosis: methotrexate, vitamin A excess
Vascular collapse: nicotinic acid, cocaine, ecstasy
Venoocclusive disease: busulfan, herbal medicines

TABLE 109-4 Types of Drug Reactions, With Examples

A distinction between the various types of liver injury can be made based on the liver tests alanine aminotransferase (ALT) and alkaline phosphatase (AP). [61] The liver injury is considered *hepatocellular* when ALT alone is more than two times above upper limit of normal (ULN) range, or when the ratio (ALT/ULN)/(AP/ULN) is greater than or equal to 5. The liver injury is termed *cholestatic* if AP alone is more than two times above ULN, whereas the term *mixed* designates a situation when both ALT and AP are above two times ULN and the ALT/ULN:AP/ULN ratio is between 2 and 5.

Hepatocellular Reactions

Hepatocellular reactions are the most common type of drug-induced liver disease, constituting up to 90% of cases. [62] They are characterized by a hepatocellular pattern of serum liver tests, as defined above. Many drugs have been implicated in hepatocellular type drug-induced liver disease (Table 109-5). Usually, improvement is quick after discontinuation of the drug (1–2 months), and only a few patients develop fulminant, acute liver failure with hepatic encephalopathy. [63]

Acetaminophen
CCl_4
Cloxacillin
Diclofenac
Halothane
Isoniazid
Lovastatin
Nefazodone
Trazodone
Venlafaxine

TABLE 109-5 Agents That May Cause Liver Injury of Hepatocellular Type

Histological findings include necrosis and cellular infiltration. Necrosis may be zonal (e.g., acetaminophen- or CCl_4-induced) or diffuse (e.g., halothane-induced), and the inflammatory response consists of lymphocytes or eosinophils. Massive necrosis formation may cause acute liver failure and death. [8]

Acetaminophen has been the best understood example of direct hepatocyte toxicity. Liver injury occurs predictably after intentional suicidal overdose, [64] and during the “therapeutic misadventure,” in which acetaminophen used in therapeutic or excessive doses for pain relief leads to severe liver injury. [44], [65] Enhanced toxicity occurs due to the enzyme induction and glutathione depletion by alcohol as well as fasting as outlined previously. [45] Acetaminophen toxicity is the most common form of acute liver failure observed in the United States. [2] In a medical record review from an urban county hospital in the United States 71 patients admitted in a 39-month period had actual or *potential* hepatotoxicity due to acetaminophen. [54] Those who ingested acetaminophen accidentally, without suicidal intent fared worse because they presented for treatment late, and did not realize that they had done anything harmful. This group was more likely to develop severe liver injury and to die from the

episode. By contrast, those with suicidal intent took larger doses, presented to a hospital earlier, and received *N*-acetylcysteine, an effective antidote. One fifth of the suicidal cases had severe injury and the potential for a fatal outcome should not be underestimated. Key features in the accidental group were excessive chronic alcohol intake (usually more than 6 drinks/day), and the use of acetaminophen for a specific pain problem, but generally in excess of package recommendations. The extremely elevated aminotransferase values (often greater than 6,000 IU/L and sometimes as high as 30,000 U/L) observed in suicidal and accidental acetaminophen ingestion help distinguish these cases from viral hepatitis or other drug injury. [54] *N*-acetylcysteine should be given by nasogastric tube on admission, and for the ensuing 72 hours, to provide glutathione substrate. In Europe, intravenous *N*-acetylcysteine is the standard treatment. [39] Expected survival is greater than 80%, although transplantation is occasionally indicated. The incidence of acetaminophen poisoning varies widely throughout the world, but is becoming more frequent and widespread as indicated by a report from Taiwan. [66] Acetaminophen poisoning is an increasing cause of ALF in children. [67], [68] and [69] As in adults, two forms are seen: teenagers may overdose with suicidal intent, but more ominous are instances of inappropriate overdosing of acetaminophen in small infants due to parents' ignorance of the high risks involved.

The U.S. Acute Liver Failure Study Group, a national registry established in 1998, reviewed the experience with 108 patients who developed ALF defined as altered mentation and coagulopathy, due to acetaminophen toxicity (Larson AM, et al, unpublished data). The majority (79%) were women, although the gender breakdown for other etiologic categories of ALF is similar, and accidental overdose comprised 58% of patients.

Cholestatic Reactions

Cholestatic reactions have been described for a number of drugs, some of which are listed in Table 109-6. Cholestasis is best defined as failure of bile to reach the duodenum, [70] and common symptoms are jaundice and pruritus. *Pure cholestasis* with no signs of hepatocellular necrosis is almost exclusively seen in patients taking oral contraceptives, anabolic steroids, or sex hormone antagonists such as tamoxifen. [71], [72] Acute *cholestatic hepatitis* is histologically characterized by cholestasis (dilated canaliculi, brown granules in cytoplasm of hepatocytes) and some degree of liver cell necrosis as well as bile duct injury and inflammatory infiltration with polymorphonuclear leukocytes. Drugs related to this type of reaction include carbamazepine, [73], [74] trimethoprim-sulfamethoxazole, [75] captopril, [76] and ticlopidine. [77]

Pure cholestasis	Anabolic steroids
	Tamoxifen
	Estrogens
Cholestatic hepatitis	Allopurinol
	Amoxicillin/clavulanate
	Azathioprine
	Barbiturates
	Captopril
	Carbamazepine
	Chaparral (herbal)
	Chlorpromazine
	Clindamycin
	Erythromycin
	Flucloxacillin
	Gold salts
	Phenytoin
	Ticlopidine
	Trimethoprim-sulfamethoxazole

TABLE 109-6 Drugs Involved in Cholestatic Drug Reactions

Generally, drug-induced cholestasis needs a longer time to resolve than the hepatocellular type of drug reactions. [3] In some instances progressive destruction of segments of the intrahepatic biliary tree may occur, the so-called *vanishing bile duct syndrome* [78] that occurs after a protracted course (more than 6 months) of drug-induced cholestasis. The result is a state of chronic cholestasis, resembling primary biliary cirrhosis. [79] Approximately 30 drugs have so far been implicated in the vanishing bile duct syndrome, including among them chlorpromazine [80] and ajmaline. [81], [82]

A sclerosing cholangitis-like syndrome with jaundice caused by intra- and extrahepatic strictures of the bile ducts is sometimes observed in patients receiving intraarterial floxuridine chemotherapy for hepatic metastases of colorectal cancer. [83]

Immunoallergic Reactions

Drugs may be associated with reactions that are definitely allergic in nature. Hypersensitivity reactions such as fever, eosinophilia, or rash are common. Halothane induces fever, eosinophilia, and antimitochondrial antibodies. [13] Halothane was formerly widely used as an inhalation anesthetic. However, it has been implicated in a high number of very severe cases of liver disease. [4], [5], [84] Halothane causes a hepatocellular injury as evidenced by findings of necrosis—ranging from spotty necrosis to bridging hepatic necrosis and multilobular necrosis—in liver biopsies. [85]

Phenytoin (Dilantin) induces the simultaneous onset of fever, rash, lymphadenopathy, or eosinophilia. [86] The mechanisms responsible for the combined allergic and hepatotoxic reaction are unknown, but the slow resolution of the illness suggests that the allergen remains on the hepatocyte surface for weeks or months. With phenytoin, a mononucleosis-like picture may also be seen and frequently is confused with a viral illness or streptococcal pharyngitis. [87] When the offending agent is not discontinued promptly, despite signs of developing hepatitis, a severe Stevens-Johnson drug eruption and prolonged fever may result. [88] As with any therapeutic agent, rapid recognition of the presence of a toxic drug reaction and immediate discontinuation of the compound are the keys to limiting hepatic damage. It is important to remember that features of an allergic reaction may not be obvious. Even in the absence of systemic signs of allergy, eosinophilia or granulomas may be present on liver biopsy.

Sulindac is a nonsteroidal antiinflammatory drug (NSAID) that causes immunoallergic liver disease with a cholestatic pattern of injury in two thirds of liver injury cases. [89] Sulindac may also cause hepatocellular liver injury and is more frequently implicated in drug-induced liver disease than other NSAIDs, such as ibuprofen. [5]

Steatohepatitis

Steatosis in the liver can be present either in a microvesicular or in a macrovesicular pattern. Macrovesicular steatosis is the most common form and is histologically characterized by hepatocytes containing a single vacuole of fat filling up the hepatocyte and displacing the nucleus to the cell's periphery. [27] Macrovesicular steatosis is typically caused by alcohol, diabetes, or obesity. Sometimes drugs such as corticosteroids or methotrexate may cause these hepatic changes. [90]

In microvesicular steatosis hepatocytes contain numerous small fat vesicles, not displacing the nucleus. [27] These lesions are associated with disruption of mitochondrial DNA with resulting anaerobic metabolism leading to lactic acidosis in the most severe cases. Not uncommonly, macrovesicular and microvesicular lesions are observed concomitantly and in these cases the microvesicular lesions should be given higher attention since they are associated with a more severe prognosis. [28] Hepatocellular necrosis may also be present. Acute fatty liver of pregnancy [91] and Reye syndrome [92] are two examples of severe liver diseases caused by microvesicular steatosis.

Drugs involved in microvesicular steatosis include valproate, [93] tetracycline, [94] fialuridine, [95] and others (Table 109-7). Aspirin use in children has been associated with Reye syndrome, [96] and the incidence of Reye syndrome has decreased since warnings concerning aspirin use in children were issued. [97], [98] The exact causal relationship between aspirin and microvesicular steatosis remains to be studied. [92]

Aspirin
Ethionine
Fialuridine
Nucleoside analogs
Valproic acid
Tetracycline (intravenous administration)
Troglitazone

TABLE 109-7 Agents That May Cause Microvesicular Steatosis of the Liver

Other Drug Reactions

There are several other types of drug reactions involving the liver, which are of lesser importance in terms of number and severity. These include granulomatous reactions, fibrosis, ischemic injury, and chronic autoimmune liver injury (see Table 109-4). The type of reaction observed can be helpful in determining the likely agent, since most drugs have a specific injury profile.

A pattern of venoocclusive disease (VOD) with obliteration of small intrahepatic veins, sinusoidal congestion, and necrosis is observed frequently in patients receiving chemotherapy (e.g., busulfan) following bone marrow transplant. [99], [100] and [101] Symptoms include rapidly accumulating ascites, painful hepatomegaly, and jaundice occurring shortly after the conditioning regimen has been instituted. Rarely, herbal medicines may cause VOD. [102]

TREATMENT

Prompt discontinuation of a suspected drug is mandatory. General supportive therapy may be necessary according to the state of the patient, ranging from none to intravenous fluid replacement to the very intensive monitoring and treatment of patients with acute liver failure with hepatic encephalopathy. [63], [103], [104] Liver transplantation is performed in more than 50% of patients with idiosyncratic drug-induced acute liver failure since spontaneous survival in this setting is less than 20%. [1]

CRITERIA FOR CAUSAL ASSESSMENT OF DRUG-INDUCED LIVER INJURY

How do we decide whether a certain drug is responsible for the liver disease encountered in a certain patient? It is important to understand that we are always trying to assess a possible association from the *past* unlike most other scoring models that try to predict the *future*. [105] A standardized reporting form developed by an international panel working in France provides a worthwhile causality assessment scoring system, [61] called the RUCAM (*R*oussel *U*claf *c*ausality *a*ssessment *m*ethod) (Table 109-8). These guidelines outline the steps an experienced clinician uses to assess likelihood of drug reactions. [106] Causality assessment methods typically include temporal relationship, course after cessation of drug, risk factors, concomitant drugs, a search for nondrug causes (viral hepatitis), previous information concerning the drug, and response to re-challenge, which is usually not available. Validation of the RUCAM scoring system suggests that a RUCAM score could be classified as *highly probable* (RUCAM score >8), *probable* (score 6–8), *possible* (score 3–5), *unlikely* (score 1–2), or *excluded* (score =0). [107]

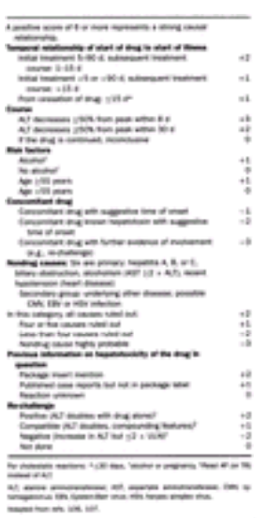


TABLE 109-8 Scoring System for Assessing Causality for Hepatocellular Types of Reactions

A newer model called the clinical diagnostic scale (CDS) was developed in Portugal [108] and tested in the United Kingdom. [109] Features from the CDS include most of the features from the RUCAM plus extrahepatic manifestations such as rash, fever, arthralgia, cytopenia, and eosinophilia. [108] The British study suggests using a CDS score of 9 or more as a very strong indicator of drug-induced liver disease. [109] However, a recent Spanish comparison study in 215 patients suggested that the RUCAM system is more accurate than the CDS model. [110]

LESSONS FROM DRUGS WITHDRAWN FROM THE MARKET

Two drugs withdrawn due to severe and fatal liver injury were bromfenac and troglitazone. There are important lessons from each one, illustrating the difficult issues surrounding drug-induced liver injury. NSAIDs as a class have been associated with considerable hepatotoxicity. [111], [112] Bromfenac was an NSAID marketed as Duract, and introduced in 1997 as a short-term analgesic for orthopedic pain. Aminotransferase elevations had been noted during clinical testing of bromfenac, but no instances of severe toxicity had been reported. However, increased transaminases were associated with longer use, although the total number of patients undergoing long-term treatment was small. No instances of ALF were observed, but less than 1000 patients were studied. Because of concerns that long-term use might be hazardous, approval was given with the limitation that the drug should only be used for intervals of 10 days or less. Once released, bromfenac was associated with more than 50 cases of severe liver injury and the drug was withdrawn in June 1998. All patients in whom toxicity was observed had been taking the drug for more than 8 weeks. [113], [114] With hindsight, a drug that effectively relieved pain would not be prescribed or used reliably only for a 10-day period. Risk/benefit ratio must be an important consideration in the approval process for any new drug—bromfenac offered little that was new and on that basis should have met an even stricter test for safety.

Troglitazone (Rezulin) was approved by the FDA in January 1997, the first of a new class of compounds, the thiazolidinediones. As an agonist of the nuclear regulatory factor peroxisomal proliferator activator receptor-? (PPAR-?), troglitazone reduces insulin resistance and increases insulin-stimulated glucose disposal, resulting in improved glycemic control for patients with type II diabetes. In clinical trials, reversible elevations of aminotransferase levels were observed, occasionally reaching more than eight times ULN, but, again, no examples of ALF. However, once the drug was approved, several reports of severe and fatal liver injury appeared. [115], [116], [117] and [118] Troglitazone has a variable time frame for demonstrating toxicity, from a few days to several months. [119] The pathogenesis is not understood. Unlike bromfenac, troglitazone was not immediately removed. As a new agent, its benefits were initially thought to outweigh the risks. The drug was recalled in the United Kingdom in 1999. The renewed warning in the package insert (in a black box), mandated monthly monitoring during the first 6 months of use. However, the number (>90) and severity of these cases (68 fatal, 10 transplanted) as well as doubts over the efficacy of monitoring led to drug withdrawal in 2000, 3 years after its approval. A factor in the FDA decision was the arrival of two new PPAR-? agonists, rosiglitazone (Avandia) and pioglitazone (Actos), approved, respectively, in May and July 1999. Although of the same class, these agents do not seem to have the same degree of toxicity. Thus far, neither drug has shown the 1:50,000 incidence of severe hepatotoxicity seen with troglitazone. Although there may be intrinsic differences in the drugs, more careful screening of patients by physicians, monitoring of ALT levels, and early discontinuation of moderately severe cases may play a part in limiting the severe toxicity associated with this class of drugs. [119]

THE DRUG APPROVAL PROCESS

How can new drugs get through the drug approval process and still lead to so many deaths that they are ultimately withdrawn? In brief, drug development is divided into three stages: initial research and development, clinical testing for new drug application (NDA), and the post-marketing experience.

Research and Development

The initial stage of drug development includes drug discovery and initial testing for efficacy, or toxicity in animals or in vitro model systems. Most new compounds fail to make it through this stage, either because of toxicity or lack of efficacy. Compounds may be “discovered” in several ways: synthesized to resemble previous compounds, discovered in the field by purification of naturally occurring peptides (e.g., cyclosporin A), or generated by computer modeling. A compound shown to have a desirable effect in vitro or in vivo, then undergoes extensive preclinical testing in a variety of animals using doses up to 50 times that predicted to be useful in humans to ascertain the types of toxicity that might be expected. While metabolic pathways differ in some specific aspects, the similarities between lower mammals and humans are quite notable. Animals are euthanized after short-term experiments and all organs examined; those dying during experiments undergo necropsy to determine cause of death. Long-term exposure studies are performed looking for carcinogenicity or other delayed effects. Preclinical testing, which may take 5 to 6 years to complete, is still a crude technique and no substitute for clinical trials in humans. The use of massive dosing in animals may in part compensate for metabolic differences between species, but human trials are ultimately needed. Toxicogenomics offers some promise of early identification of “toxicity” gene expression profiles (i.e., signatures).

Clinical Trials

In phase I testing, progressively larger doses of the test medication are given to well-paid healthy volunteers. Routine monitoring includes vital signs, electrocardiogram (ECG), assessment of reported side effects, and blood measurements including serum aspartate aminotransferase/alanine aminotransferase (AST/ALT), amylase, and creatine phosphokinase (CPK). In phase II testing, patients are exposed for the first time and the emphasis shifts from safety alone to safety *and* efficacy. Depending on the intended use of the medication and the prevalence of the disease to be treated, from 500 to 5000 study patients may test the medication for periods of up to a year. In early phase II trials, a progressive dosing scheme identifies the maximal dose that is effective and still safe. If a given dose is effective, it is then determined if there are any short- or long-term side effects. In one example, adefovir dipivoxil, a nucleotide analog was being given for human immunodeficiency virus (HIV) infection. Unanticipated renal failure was noted at 60 and 120 mg daily doses, [120] and the dose was then lowered to 30 and finally to 10 mg/day. However, adefovir was not effective at 10 mg and only partially so at 30 mg, so the drug did not gain approval for HIV. Adefovir was recently approved for hepatitis B where its efficacy appears to be better than that observed for HIV infections. Such an example of frequent renal or hepatotoxicity is easy to spot—serum creatinine levels rose in many patients taking adefovir within the first few months on treatment. Several NSAIDs have made it to phase II trials only to be withdrawn due to frequent aminotransferase level increases. However, this is where safety concerns regarding idiosyncratic reactions founder. It is easy to pick up common reactions but hard to pick up the truly rare drug injury during the pre-approval process.

Dr. Hyman Zimmerman stated that if a drug causes enough liver injury to lead to jaundice, even rarely, then 10% of affected patients will develop acute liver failure (“Hy’s rule”). [90] Put another way, any drug that in phase II or III testing demonstrates not only aminotransferase elevations, but increases in bilirubin or jaundice will likely lead to ALF when larger numbers of patients are exposed. This sounds like a very imprecise “rule” but it has served quite well over the years, and there does not seem to be anything better.

How certain can we be that clinical studies identify instances of liver injury? First, all studies are conducted according to previously established guidelines of good clinical practice. In each clinical study, a detailed assessment of liver biochemical parameters are part of every company’s NDA filing. Data supporting the safety of the drug include placebo-controlled trials where the incidence of abnormalities must be shown to be similar to that observed in the placebo group. Aminotransferases exceeding three times the ULN generally require discontinuation of the drug. Increased aminotransferase levels *without* bilirubin elevations may not lead to discontinuation during a phase III trial, but frequent or more severe aminotransferase increases (>8 times ULN) or accompanying increases in bilirubin will likely bring a new drug trial to a halt. If any case of ALF occurs, the trial is discontinued, as was the case with fialuridine.

The FDA approves approximately 50 new drugs each year. The approval process takes between 6 months to a year, once the NDA is filed. Approval brings with it instant widespread, intense marketing efforts and the necessity for all U.S. pharmacies to stock the drug. As noted previously, the number of prescriptions frequently rises rapidly into the millions, within a year or less. This fact explains why some drugs only demonstrate problems once they receive FDA approval. Idiosyncratic events occurring in only 1:50,000 patients are not going to be recognized in a study of 4500 patients. The “rule of threes” applies: to reliably identify a single case of liver injury due to a drug with 95% confidence, there must be three times the number of patients studied as the incidence of the drug reaction. In other words, a 1:1500 reaction requires 4500 patients to reliably detect a single case; a 1:50,000 reaction would require 150,000 patients. No clinical trial will reliably pick up rare drug reactions. Approval by the FDA provides a wider experience than the limited exposure of the carefully controlled clinical trial. Thus, it should not be surprising that drug reactions are observed in the post-marketing period and not before. However, post-approval drug recall still takes time to evolve while the drug continues to be prescribed despite the recognition of adverse events.

Post-Marketing Surveillance

The greatly increased number of patients receiving a new drug ensures that untoward or unusual drug effects will be observed. In addition to increased numbers, a wider range of patients than the defined clinical trial population is exposed. For example, most studies do not include patients with renal failure, heart failure, HIV/AIDS, pregnant women, the elderly, or children. Any of these groups may show enhanced toxicity. Even the best-randomized controlled clinical trial is not a “real-life” experience. The difficulty is in identifying these drug reactions quickly and accurately, once the product is released. During clinical trials and the after market period, pharmaceutical companies must report serious adverse events (SAEs) to the FDA within 24 hours. An SAE is any unexpected medical occurrence that at any dose results in death, is life threatening, requires hospitalization, or results in permanent disability or a birth defect. Pharmaceutical manufacturers maintain a safety monitoring force which gathers reports, assesses likelihood of the reaction being attributed to their product, and issues a report to the FDA and to clinical investigators if there is still an ongoing trial. However, there is bound to be a bias toward any new product, just as there is bias built in to the design of clinical trials. [121]

There are several additional shortcomings to the effectiveness of post-marketing surveillance. First, the reporting system is passive. Physicians and pharmacists are under no obligation to report adverse events. The FDA introduced the Medwatch program to improve surveillance, asking physicians and pharmacists to report on a standardized form all drug reactions they observe. However, it is estimated that less than 10% of severe adverse drug reactions are reported to the company or the FDA. Reasons for under-reporting include: failure to recognize “hepatitis” as being due to a drug, concern about malpractice implications, reluctance to get involved, complacency (“too busy”), and so forth. Reports received from pharmacists or drug representatives (or physicians for that matter) seldom contain full clinical information. Privacy issues may preclude further inquiries and raise concern regarding possible legal implications. For all these reasons, a passive reporting system is inadequate. Nevertheless, the main source of information is the Medwatch system, plus case reports. [122]

HEPATOTOXICITY IN THE PATIENT WITH CHRONIC LIVER DISEASE

Hepatologists are frequently asked “Is the patient with liver disease more susceptible to liver injury?” Intuitively, this makes sense, until we realize that hepatotoxic reactions represent the culmination of hepatic enzyme activity. If liver function is impaired, one might predict diminished activity of certain enzyme systems. Patients with liver disease do not appear to be at increased risk for hepatic injury compared to their counterparts without underlying liver problems. Dr. Zimmerman put it best: “A stubborn [misconception] has been the view that patients with pre-existing hepatic disease are more likely than others to suffer hepatic injury on exposure to drugs that cause liver damage. There is virtually no evidence for this view.” [90] What do we know of the liver function of patients with cirrhosis? Many enzyme systems are well preserved even in advanced disease. For example, patients with terminal alcoholic hepatitis still are able to conjugate most of their bilirubin. Therefore, enzyme activity in many instances exceeds the daily requirement, so that even severe liver injury would not be expected to lead to an adverse drug reaction. In general, phase I reactions may be diminished but this is not uniformly so. In severe liver disease the activity of CYP2C19 is greatly decreased while that of CYP2D6 is intact. [123] In nonalcoholic steatohepatitis (NASH) enzyme cytochrome CYP2E1 is increased, particularly in the centrilobular region, so that acetaminophen toxicity should be enhanced in patients with NASH. [124] Thus far, this has not been appreciated clinically.

Drug metabolism in patients with cirrhosis can be reduced as much as 50%. Whether the cells in a patient with cirrhosis are sick or simply reduced in number but functioning normally is not clear. Neither answer is exactly correct. It appears that the physiological changes seen with fibrosis along the hepatic sinusoids results in a widening of the barrier between the bloodstream and the hepatocyte. In support of this, patients with cirrhosis with comparably diminished metabolism of acetaminophen and theophylline normalize theophylline dis- posal, but not acetaminophen, with oxygen supplementa- tion. [125] The metabolism of theophylline uses CYP1A1 and CYP1A2, which requires oxygen as substrate, unlike acetaminophen conjugation (phase II). The limitation to metabolism is the barrier to oxygen absorption. These studies support the “intact hepatocyte/sick membrane” hypothesis. In summary, dosage adjustments may need to be made in patients with

cirrhosis, but these individuals do not appear to have an abnormally sensitive hepatic metabolic system, just less reserve if an hepatotoxic insult were to occur. [125], [126]

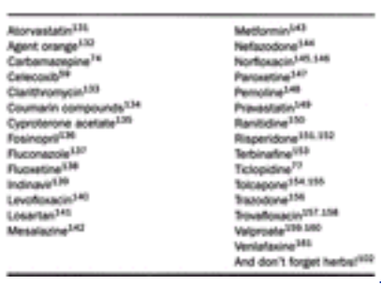
In general, patients with liver disease suffer more renal than hepatic insults. They are particularly prone to nephrotoxicity due to the altered renal circulation of the cirrhotic patient. Nephrotoxicity of aminoglycosides, radiocontrast, and prostaglandin inhibitors such as indomethacin are a frequent problem for patients with cirrhosis, but doses of antibiotics, antipsychotics, and so forth, are seldom adjusted, although any medication with sedating effects may be a problem if metabolism is slowed.

Avoiding further liver injury in the patient with preexisting liver disease is a difficult task. Antituberculous therapy cannot be withheld from patients just because they have alcoholic cirrhosis. In these instances, frequent monitoring appears to be helpful, but the value of this monitoring has not been proven in controlled trials, is seldom adhered to and can prove very expensive. Despite surveillance using liver enzyme levels, acute liver failure has developed in patients treated with isoniazid. In many instances, the presence of preexisting liver disease is subclinical, (e.g., in patients with NASH). Whether the diabetic population is more at risk for troglitazone hepatotoxicity due to their diabetic fatty liver is still debated. Nevertheless, a healthy regard for the possibility of increased hepatotoxic reactions in patients with preexisting liver disease, and the use of periodic surveillance during treatment should allow the maximum chance to avoid prescribing drugs that are harmful.

CLINICIAN'S GUIDE TO HANDLING NEW DRUGS

The best advice in prescribing new pharmaceutical agents is not to prescribe them. It is wise to defer embracing new drugs during their first year of introduction particularly if they demonstrate no unique advantages over accepted formulations. Marketing hype exceeds real-life experience with any new agent. Physicians must strive to instill in their patients a healthy level of alertness with regard to drug-induced liver injury, particularly for agents with known hepatotoxicity. Physicians and pharmaceutical companies should strike a careful balance between alerting patients to the potential for severe reactions without frightening them so that they avoid needed medications. Monitoring aminotransferase levels is suggested for known hepatotoxins such as isoniazid or diclofenac on a monthly basis but is unlikely to be cost-effective when an adverse reaction occurs in only one in 50,000 patients. Since many drug reactions develop within days, monitoring provides no guarantee. Most fatal drug reactions could be prevented if the offending agent were withdrawn immediately, at the first sign of illness. The patient most likely to be harmed is the one who believes in the complete safety of drugs, doesn't realize that drug-induced injury is possible, or is encouraged to be compliant when signs of toxicity are beginning.

New drugs should be prescribed with caution, keeping an eye out for case reports. Some of the newer agents implicated in acute liver necrosis are listed in Table 109-9.



Atorvastatin[131]	Metformin[143]
Agent orange[132]	Nefazodone[144]
Carbamazepine[74]	Norfloxacin[145,146]
Celecoxib[59]	Paroxetine[147]
Clarithromycin[133]	Pemoline[148]
Coumarin compounds[134]	Pravastatin[149]
Cyproterone acetate[135]	Ranitidine[150]
Fosinopril[136]	Risperidone[151,152]
Fluconazole[137]	Terbinafine[153]
Fluoxetine[138]	Ticlopidine[77]
Indinavir[139]	Tolcapone[154,155]
Levofloxacin[140]	Trazodone[156]
Losartan[141]	Trovafloxacin[157,158]
Mesalazine[142]	Valproate[159,160]
	Venlafaxine[161]
	And don't forget herbs![162]

TABLE 109-9 Current List of Newer Drugs Reported to Cause Severe Toxicity

The diagnosis of drug-induced liver injury necessitates determining the precise timing of the drug ingestion, making a careful record of all drugs ingested, and being particularly suspicious of known hepatotoxic agents begun within 3 months of the onset of illness. After withdrawal of the offending agent, improvement should be rapid, within days. Cautious re-challenge may be made *only* if the toxicity observed was highly questionable and if no other drug is available for a serious problem. If jaundice, coagulopathy, or any degree of encephalopathy is present initially, then hospitalization is required since drug reactions worsen quickly, and fatal outcomes are common. [1], [2]

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CHAPTER 110

E. Jenny Heathcote

AUTOIMMUNE HEPATITIS

When first described [1], [2] autoimmune hepatitis (AIH) was thought to be a potentially fatal form of chronic hepatitis which predominately affected young women. It is now recognized that this disease may affect all age groups [3], [4] and men are affected in a ratio to women of 1:4. All racial groups are at risk of AIH. Its prevalence in Europe is about 1.9 per 100,000. [5] Spontaneous remission may occur but subsequent relapse is usual; only rarely is the disease curable. Although this disease can be fatal if severe and left untreated, [6], [7] and [8] earlier recognition of the manifestations of AIH has shown that in some instances the disease may be asymptomatic and its course perhaps more benign than was originally thought.

PATHOGENESIS

There are many features of AIH, which suggest the disease is immune-mediated. Certain human leukocyte antigen (HLA) class II alleles are associated with both disease “susceptibility” and treatment responsiveness. [9] In individuals from North America and Europe the class II HLA DRB * 0301 and DRB * 0401 alleles are more common in individuals with type I AIH. In South America and Japan the HLA pattern associated with AIH is different. A common antigenic peptide which recognizes a positively charged amino acid at positions 67-72 on the DRB allele may be the link. [10] The immune response is complex, involving several promoters such as costimulatory signals for T-cell activation, adhesion between immune synapses, cytokine production, and T-cell receptor configuration. Genetic polymorphisms of one or more of these promoters may be relevant to the pathogenesis of AIH. [11]

AIH may be associated with low serum complement levels and a null allotype at the C4A or C4B gene location (a silent C4AQ+0 gene is identified in such patients). [12] This C4A gene deletion is associated with both relapse on therapy and increased mortality. [13]

Immunoglobulin levels are elevated two- to threefold in active AIH, indicating marked B-cell activation, the cause being multifactorial. [14], [15] Several reports imply that the onset of AIH may be precipitated by an acute viral infection [16], [17], [18] and [19] in genetically susceptible individuals. The induction of AIH in a few patients being treated with interferon for chronic hepatitis C has also been described. [20] Certain drugs (e.g., minocycline) may also induce an AIH-like disease. [21], [22] and [23]

The nonorgan-, nonspecies-specific antibodies typically associated with AIH are antinuclear antibodies (ANAs) and smooth muscle (specifically antiactin) antibodies (SMAs). The detection of these antibodies in up to a third of individuals with other liver diseases not thought to have an autoimmune basis [24] makes it unlikely that they have any pathogenic role on their own. Another autoantibody-antisoluble liver antigen (anti-SLA) is described in 11% of patients with type 1 AIH. This antibody is identical to liver/pancreas antibody. [25], [26] Anti-SLA may also be detected in 14% to 30% of patients given a diagnosis of cryptogenic cirrhosis. This latter observation supports the concept that some patients with a diagnosis of cryptogenic cirrhosis may have an autoimmune basis for their liver disease. [27]

There is another distinct form of AIH (type 2) which has been most often described in Europeans but does occur in North America. [28] In these individuals an antibody against liver and kidney microsomes (anti-LKM1) (specifically the cytochrome P450-2D6) can be detected; serum generally tests negative for ANA and SMA. It remains unknown as to whether this antibody has any pathogenic significance. If the immunofluorescence slides are not read carefully this antibody could be misinterpreted as being an antimitochondrial antibody (AMA). [29] Sometimes anti-LKM1 can be detected in the serum of persons with chronic hepatitis C. [30]

There are many other autoantibodies described in association with AIH. Asialoglycoprotein receptor (ASGP-R) antibodies can be detected in 90% of patients with all forms of AIH. This antibody is directed at the receptor for $CD4^{+}$ T cells. [31] Antineutrophil cytoplasmic antibodies (ANCAs) were initially believed to be a hallmark for primary sclerosing cholangitis (PSC) but subsequent case series indicate that these antibodies are not specific and are also common in AIH. [32]

A rare condition known as autoimmune polyglandular syndrome type 1 (APS-1) is associated with AIH in 10% to 20% of patients. APS-1 is associated with a homozygous mutation in an autoimmune regulator (AIRE) gene, which encodes a nuclear transcription factor. A survey of individuals with a variety of autoimmune liver disease did not indicate that this same mutation was present in individuals with AIH. [33]

Some patients thought to have an AIH are found to test positive for only anti-SLA, and have been categorized as having type 3 AIH. However, more than likely as the symptoms are so similar, these rare patients can be categorized as having type 1 AIH.

AIH type 1 is the form most frequently encountered in North America; type 2 appears to be more common in Europe. The latter more commonly affects young persons, generally presents acutely, and is more frequently associated with cirrhosis.

The HLA patterns in type I and type 2 AIH are also distinct but this may be partially explained by the very different geographic distribution of these two forms of AIH. The HLA pattern which appears to confer resistance to AIH type I, confers susceptibility to AIH type 2 (i.e., DRB1 *07 and DRB1 *15 and DQB1 *06) [34] suggesting that the genetic susceptibilities to these two forms of AIH are quite separate. Type 1 and type 2 AIH can often be distinguished clinically by the natural history of the disease, the laboratory parameters for each type, and the different responses to treatment [35] (Table 110-1).

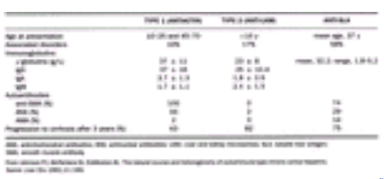

TABLE 110-1 Comparison of the Clinical and Immunologic Features of Types 1 and 2 Autoimmune Chronic Active Hepatitis

CLINICAL MANIFESTATIONS

Presenting Symptoms

At first presentation the diagnosis of AIH may be missed as the symptoms of a mild self-limiting, acute hepatitis are very nonspecific. [36] Most often AIH is diagnosed in individuals who are found to have markedly abnormal biochemical tests of the liver when they present with jaundice. Occasionally presentation may be as an acute [37] and sometimes fulminant hepatitis. [38] Careful history taking may reveal past nonspecific but relevant symptoms such as episodes of malaise, nausea, and arthralgias. There may be a family history of other autoimmune diseases. Sometimes the disease may present de novo with hepatic decompensation (e.g., ascites, variceal hemorrhage). On occasion a complication of AIH (e.g., secondary amenorrhea or delayed menarche) may be the only complaint which leads to a visit to a physician. AIH is sometimes entirely asymptomatic, diagnosed as the result of blood work done as part of a routine physical or as part of the workup of another autoimmune disease.

As with most chronic liver diseases fatigue is the most common symptom; jaundice is a presenting symptom in more than half. Other gastrointestinal complaints include anorexia, diarrhea, sometimes pruritus, and occasionally right upper quadrant discomfort. Associated autoimmune diseases include thyroiditis, inflammatory bowel disease, and rheumatoid arthritis. In children there is a very strong link between AIH and sclerosing cholangitis; [39] up to 50% are found to have both diseases simultaneously at presentation. [40] This dual presentation is much less frequently observed in adults.

Physical Examination

Generally, hepatomegaly is present and jaundice is frequent but there may be no abnormal physical findings noted, particularly if cirrhosis is absent. The spleen may be palpable (50%) and spider nevi may be present but they do not necessarily indicate cirrhosis. Hepatic decompensation is unusual but occasionally patients with “burned out” disease may present with ascites (20%), hepatic encephalopathy (14%), or bleeding varices (8%). The liver is generally small by this time. [36]

Laboratory Investigations

The alteration in liver enzymes in AIH is typically hepatocellular. Elevation in the serum aminotransferase levels may be quite marked, greater than 1000 IU/L in some cases. A conjugated hyperbilirubinemia is common. Further evidence of liver dysfunction may be present, such as a coagulopathy and hypoalbuminemia. The blood film may reveal a pattern typical of hypersplenism (low total white cell count and/or platelet count and/or hemoglobin) even at presentation in those with already established cirrhosis.

Infection with hepatitis A, hepatitis B, and hepatitis C must be ruled out through serological testing. Other viruses may affect the liver as part of a systemic infection, (e.g., Epstein-Barr, adenovirus, parvovirus, and Cytomegalovirus [in the immunocompromised individual]) and these should be sought if an infectious etiology is considered likely.

If viral hepatitis is excluded, testing for serum ANA and SMA should be requested: the immunofluorescence technique is sufficient. Anti-LKM1 and ANA/SMA are nearly always mutually exclusive, thus anti-LKM1 should be requested once serum tests negative for ANA and SMA. If there is concern that anti-LKM1 may have been misread, then an enzyme-linked immunosorbent assay (ELISA) can be used to confirm the identity of AMA. It is probably not clinically helpful to screen for any other autoantibodies. Only when ANA and/or SMA are associated with an at least twofold increase in immunoglobulin G (IgG) should a diagnosis of type 1 AIH be seriously considered. These two antibodies, generally in low titer, are detected in up to a third of individuals with other forms of chronic liver disease but the hypergammaglobulinenemia is much greater in type 1 AIH. In type 2 AIH the hypergammaglobulinemia may not be as profound, even low immunoglobulin A (IgA) levels may be observed. [34]

LIVER HISTOLOGY

To confirm a diagnosis of AIH liver histology is essential even though the pattern of disease is not entirely specific. Both viral hepatitis and drug-induced hepatitis may be histologically indistinguishable from AIH. If there is no evidence of hepatic decompensation, liver biopsy can be performed via the percutaneous route, otherwise the transjugular route is necessary. The liver biopsy should be reviewed by an experienced hepatopathologist. The portal tracts and hepatic lobules need to be examined in minute detail looking for evidence of an interface hepatitis at the junction of the portal region and the liver lobule and a lobular hepatitis caused by infiltration with lymphoplasmacytic chronic inflammatory cells (Fig. 110-1). The intrahepatic bile ducts generally appear normal. If there are cholangitic features then a strong suspicion of an associated cholangitis or a misdiagnosis should be entertained. Copper retention within hepatocytes is one of the hallmarks of Wilson disease but may also be seen to a lesser degree in individuals with AIH with or without cholangitic features. Staining for a-1 antitrypsin globules with the periodic acid-Schiff stain (found then to be diastase-resistant) should be negative and there should be no excess iron noted. In those who present with severe disease, large areas of bridging necrosis may be noted. Even when the clinical presentation is acute, it is common for patients to have evidence of chronic liver disease, possibly even established cirrhosis. The composite histological picture is 81% specific and the positive predictive value is 68% [40] of AIH when compared with tissue from individuals with viral hepatitis and cryptogenic cirrhosis.

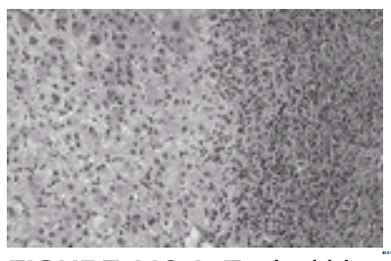

FIGURE 110-1. Typical histological picture of autoimmune hepatitis.

DIFFERENTIAL DIAGNOSIS

There are other liver diseases that may present in a similar fashion to AIH and therefore need to be routinely excluded before a confident diagnosis of AIH can be made. Measurement of serum ceruloplasmin level is essential in those individuals who present when young (<40 years). If the levels are reduced, measurement of 24-hour urine copper and slit-lamp examination of the eyes for Kayser-Fleischer rings are necessary so as to exclude *Wilson disease.* Falsely low values for ceruloplasmin may be seen in patients with severe hepatic decompensation from any cause. Falsely high urine copper may result from severe cholestasis but rarely to the degree (10- to 20-fold elevated) associated with Wilson disease. There are so many genetic mutations associated with this disease that no one mutation can be used as a specific hallmark.

There are several drugs that may cause the clinical, biochemical, serologic, and histological features typical of AIH. The drugs currently most often associated with typical features of type 1 AIH are minocycline [23] and nitrofurantoin. [22] The picture of type 2 AIH may be mimicked by tienilic acid. [2] Withdrawal of the drug may not be instantly associated with biochemical improvement, however, resolution should occur over a matter of months, not years.

Fortunately there are now sensitive and specific tests to identify *viral hepatitis.* Nevertheless hepatitis C may mimic both type 1 and type 2 AIH. [41], [42] In individuals from areas where the prevalence of viral hepatitis is high it is possible for chronic viral hepatitis and AIH to coexist.

Cryptogenic cirrhosis is the diagnosis given to patients in whom no obvious etiology can be identified. Czaja and colleagues [43] compared 12 such cases with 94 cases of clear-cut AIH and showed that there was no statistical difference between the two diseases with respect to gender, age, pattern of histological activity at presentation, or frequency of concurrent autoimmune disorders. However the level of total gammaglobulin and the IgG concentrations were significantly lower in those with a diagnosis of cryptogenic cirrhosis. Patients with cryptogenic cirrhosis had no ANA, SMA, anti-LKM-1, AMA, thyroglobulin, or microsomal thyroid antibodies in

their serum. However HLA class I and II patterns were similar between the groups, suggesting that the etiology in these individuals was likely autoimmune. All patients were treated with corticosteroid therapy and improved. Other authors have described a similar population. [44] However so-called cryptogenic cirrhosis may be caused by other silent diseases (e.g., nonalcoholic steatohepatitis) [45] and it is probably unwise to treat "cryptogenic" cirrhosis with immunosuppressive therapy unless there are some autoimmune features present.

AUTOIMMUNE HEPATITIS/PRIMARY SCLEROSING CHOLANGITIS OVERLAP SYNDROMES

Particularly in children, overlapping features of AIH and primary sclerosing cholangitis (PSC) are common. [39], [40] It is unclear whether the cholangitis visible on radiological delineation of the biliary tree in children with otherwise obvious AIH is the same cholangitis that may be diagnosed in adults. Both are frequently associated with chronic inflammatory bowel disease. There may be no distinguishing features between individuals with AIH and those with AIH plus PSC (i.e., serum alkaline phosphatase/gammaglutamyltranspeptidase and immunoglobulin values may be elevated in both). The therapeutic response cannot distinguish between the two. Thus a high index of suspicion is needed. Recent data suggest that in children it is probably appropriate to examine the biliary tree via endoscopic retrograde cholangiopancreatography (ERCP) or magnetic resonance imaging (MRI), even if the primary diagnosis is AIH. [39] The cholangitis may affect only the intrahepatic bile ducts, although in most, progression to extrahepatic disease occurs later in life.

When Boberg and associates [46] applied the first AIH scoring system to 114 adults given a primary diagnosis of PSC they found that two individuals scored "definite" and eight "probable" for AIH. Subsequently the application of the modified AIH scoring system [47] has reduced the number of patients with radiological proven PSC who score "probable" for AIH. Two publications indicate that the rate of PSC/AIH overlap ranges from 4% [48] to 8% of patients with PSC. [49] In the latter study the patient population was younger, so it may be reporting the pediatric PSC/AIH overlap described by Gregorio and colleagues. [40]

The *AIH scoring system* can be employed to determine the strength of evidence that an individual has AIH (Table 110-2). Factors considered important (by a panel of experts) when making the diagnosis have been weighted either positively or negatively. They include gender, alkaline phosphatase (ALP):aspartate aminotransferase (ALP:AST) (or alanine aminotransferase [ALT]) ratio, height of IgG, titer ANA, SMA or LKM1, AMA status, viral serology, drug history, average daily alcohol intake, liver histology (interface hepatitis, predominant lymphoplasmacytic infiltrate, rosetting of liver cells, biliary changes), and other autoimmune diseases. Additional measures can include seropositivity of other autoantibodies, HLA DR3 or DR4, and response to immunosuppressive therapy. [47]

TABLE 110-2 Summary of Autoimmune Hepatitis Scoring Revised Sheet

NATURAL HISTORY OF AUTOIMMUNE HEPATITIS

Now that immunosuppressive treatment is the standard of care for symptomatic AIH, the natural history of this disease is rarely observed. Examination of the data from subjects who received placebo in the randomized controlled trials of corti- costeroids ± azathioprine reported in the early 1970s indicate that AIH is a rapidly progressive disease with a 5- and 10-year survival rate of only 50%, and 10%, respectively [4], [5] and [6] (Fig. 110-2). Death is generally due to liver failure often complicated by sepsis. Hepatocellular carcinoma is rare. [50] However it is important to note that 30 years ago, when these trials were conducted most patients presented to their physician with hepatic decompensation and thus the majority had well-established cirrhosis at baseline. With the introduction of routine laboratory checks of liver biochemistry and a heightened awareness of liver disease in general, it is likely that the proportion of individuals diagnosed with less advanced disease is increasing. Sometimes AIH may be diagnosed in the absence of any symptoms. Currently there are no published data on the natural history of asymptomatic AIH.

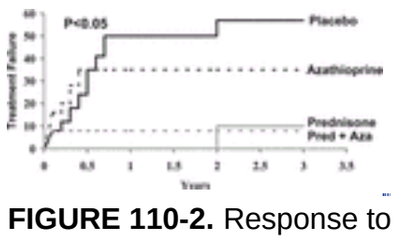

FIGURE 110-2. Response to immunosuppressive therapy. (From ref. [8].)

Treatment, successful in more than 80% of affected individuals, has allowed most patients with AIH to lead a normal life with a life expectancy similar to age- and gender-matched control populations (i.e., 98% at 10 years). [51] This study also indicated that the 10-year survival in AIH is not dependent on baseline liver histology (Fig. 110-3). For the most part young women with this disease, if treated appropriately, may have successful pregnancies (corticosteroids and/or azathioprine should not be stopped during pregnancy). If flare-ups do occur they are nearly always in the postpartum period. [52]

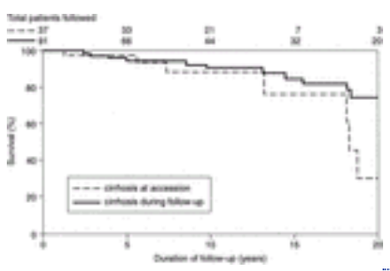

FIGURE 110-3. Survival of patients with type 1 autoimmune hepatitis: comparison of survival, using death and orthotopic liver transplantation as end points, from development of cirrhosis at accession versus follow-up. (From ref. [51].)

Unfortunately most of the complications of AIH are due to side effects from long-term immunosuppressive therapy needed to control liver disease. It is essential to monitor systemic blood pressure, bone mineral density, and blood or urine glucose. Regular eye examinations looking for cataracts should also be performed.

Patients with AIH may develop other autoimmune diseases during follow-up, (e.g., thyroiditis and rheumatoid arthritis). Thus patients given a diagnosis of AIH need to recognize their need for lifelong follow-up. The complications of liver failure are as for all liver diseases, namely, ascites, hepatic encephalopathy, and variceal hemorrhage. It must be determined if liver failure is due to a flare-up of disease in which case medical therapy is appropriate or due to “burned out” end-stage disease in which case liver transplantation is probably the best option.

TREATMENT (Table 110-3)

Initial therapy: Prednisone 30–40 mg/d monotherapy or prednisone 30 mg/d and azathioprine 50–100 mg/d combined therapy
Dose reduction: Slowly reduce prednisone (2.5–5 mg every 1–3 mos) if ALT remains <1.5 uln
Maintenance: Minimum dose prednisone and/or azathioprine to maintain ALT <1.5 uln
Stop therapy: If ALT <1.5 uln for 1–2 y +/−, liver biopsy indicates inactivity
Retreatment of relapse: Reintroduce therapy as for initial treatment
Failure to respond: To above, use either high dose prednisone 40–60 mg daily or another immunosuppressant, or consider liver transplant if appropriate

ALT, alanine aminotransferase; uln, upper limit normal.

TABLE 110-3 Suggested Therapeutic Regimen: for Autoimmune Hepatitis

The first therapeutic trials in patients with AIH conducted in the late 1960s and early 1970s indicated a highly significant improvement in survival following therapy with corticosteroids. [4], [5] and [6] The studies from the Mayo Clinic indicated that treatment with high-dose corticosteroid (60 mg daily initially with 20 mg daily as maintenance) and lower dose corticosteroid combined with azathioprine (30 mg prednisone daily initially and reducing the dose to 10 mg daily as maintenance plus 50 mg/d of azathioprine) were equally effective. Azathioprine 100 mg/d monotherapy was no better than placebo. [7], [8] Several lessons were learned from these trials. First, high-dose corticosteroids are associated with the rapid onset of severe side effects (osteoporosis, diabetes, systemic hypertension, and cataracts) and thus it is wise to treat symptomatic patients from the start with a lower dose of steroids (30–20 mg/d) combined with the steroid-sparing agent azathioprine. As azathioprine takes at least 6 weeks to take effect it makes sense to start this agent when steroid treatment is initiated. The dose of steroid can be tapered once the serum aminotransferase levels have fallen to less than a twofold elevation. Tapering by 2.5 to 5 mg every 1 to 3 months with careful monitoring of the aminotransferase levels is appropriate. Most patients have a rapid symptomatic and somewhat slower biochemical response within the first few weeks of therapy. There are some patients whose serum aminotransferase levels do not return to normal for many months. A fall in the level of IgG is also a good indicator of response. Liver histology does not improve until the biochemical tests have been normal for at least 6 months. About 20% respond poorly with 13% achieving only a partial response and only 7% who do not respond to treatment at all. [53] Poor response to immunosuppressive therapy is more common in patients whose liver biopsy shows areas of submassive necrosis. [54] Patients with cirrhosis, even if decompensated, generally respond well to therapy. Therefore even patients being considered for liver transplantation, if they have active disease, should be given immunosuppressive therapy without delay. No randomized control trials of therapy in patients who have asymptomatic disease have been conducted. An unsuccessful attempt was made, but patients refused to be included, put off by the long list of side effects attributed to corticosteroids (Dr. P. Gregory, personal communication, 1997).

Once the serum aminotransferase levels have returned to near-normal values it may be possible to wean the patient off corticosteroids altogether (this was not done in the early trials) and maintain patients on azathioprine alone in a dose of 1 to 2 mg/kg/d. [55], [56] Such a strategy reduces the deleterious consequences of long-term corticosteroid therapy. The white blood cell count needs to be regularly checked because of the potentially toxic effects of azathioprine on the bone marrow. Some patients cannot tolerate azathioprine, mainly because of its gastrointestinal side effects. Alternate-day immunosuppressive therapy is ineffective in AIH.

It is generally advised that immunosuppressive therapy be continued for up to 2 years after the liver biochemical tests have normalized before cessation of immunosuppressive therapy is contemplated. This timing is arbitrary; however it is likely that at least 1 year of therapy is essential. Some would advocate that prior to stopping treatment another liver biopsy should be performed to confirm that the hepatic inflammation has disappeared. A reduction in fibrosis may be observed [57], [58] but unfortunately half of those patients not cirrhotic at presentation will progress to cirrhosis despite ongoing immunosuppressive therapy and an apparent biochemical response. [59]

In those rare patients with AIH in whom standard therapy with a combination of prednisone and azathioprine fails, other immunosuppressive agents have been tried. Alternatives to classic treatment are also needed for children who may experience devastating growth retardation due to the use of high-dose corticosteroids. However, carefully monitoring corticosteroid therapy is probably still the best treatment in children, although cyclosporin has been used with benefit in children [60] both with type 1 and type 2 AIH. [61], [62] A newer agent used to prevent transplant rejection, mycophenolate mofetil (MMF), may be preferable as it has less nephrotoxicity—but it often causes diarrhea. [63] Methotrexate has been reported to be of benefit, as has cyclophosphamide in refractory type 1 AIH. [64], [65]

Budesonide is a corticosteroid which has a 90% first pass effect in the liver, but nevertheless appears to have a beneficial effect at this site, causing aminotransferase levels to normalize in AIH. [66] Unfortunately these encouraging results were not confirmed in another study; [67] in addition steroid side effects were observed in this latter study likely because intra- or extrahepatic portosystemic shunting was present thus obviating the beneficial properties of budesonide.

Once it is believed that the disease is in long-term remission (sustained normalization of serum aminotransferase levels, with histological evidence on liver biopsy of quiescent disease) immunosuppressive therapy may be stopped. Unfortunately the majority (80%) of individuals with AIH who are weaned off treatment will relapse, [68] generally within the first few months after treatment cessation but sometimes not until several months or even years later. Reinstitution of immunosuppressive therapy is generally successful.

It is now quite unusual for patients who have been treated for AIH to progress to end-stage liver disease. Risk factors for disease progression are a DR4-negative phenotype and the presence of anti-LKM1 antibodies as well as massive necrosis on first biopsy and low albumin and high prothrombin time at present. [69] However when liver transplant is necessary, it is extremely successful (>90% 5-year survival), although a pattern of recurrent liver disease similar to AIH may recur in the transplanted liver in up to 40%. [70], [71] In most cases this responds to increased immunosuppressive therapy. For this reason attempts at withdrawal of anti-rejection therapy in those who received a liver transplant for previous AIH are not recommended.

In addition to specific therapy for liver disease, individuals with AIH may require additional supportive therapy. All should be encouraged to lead an entirely normal life but certain precautions need to be taken, as is the case for anyone on long-term immunosuppressive therapy. In addition the complications of long-term corticosteroid therapy should be anticipated and prevented if possible. Those intending foreign travel should receive appropriate advice from travel medicine specialists, particularly with regard to appropriate vaccinations and how to avoid certain parasitic infections, (e.g., *Strongyloides*). All patients on immunosuppressive treatment should learn to recognize any infection early in its course and rapidly seek medical attention.

In young women who are successful in becoming pregnant it is important to check for the presence of esophageal varices in those who are cirrhotic, as nonselective ß-blocker therapy may well be required during pregnancy. Advice to the obstetrician to avoid a prolonged second stage of labor is also important. It is quite safe to maintain corticosteroid and/or azathioprine therapy during pregnancy; sometimes requirement for immune suppression may lessen during the pregnancy. Flare-ups, if they occur, are most often observed following delivery although they may occur during pregnancy. [52]

Baseline measurement of bone mineral density is wise. Adequate intake of calcium and vitamin D should be recommended to all individuals with AIH given corticosteroids. Preventive biphosphonate therapy has been shown to be of value.

SUMMARY

Although AIH is one of the less common causes of a chronic hepatitis it is essential that the diagnosis not be missed, as therapy is so successful, even curative in up to 20%. Most patients once in remission can be maintained in good health on azathioprine monotherapy with no untoward side effects. Treatment of AIH with immunosuppressive agents does not always prevent progression to cirrhosis, but it does significantly reduce the risk of dying of liver failure.

Key Points

- Two types AIH, distinguished by autoantibody status
- Presentation may be with a silent, or a symptomatic or even fulminant hepatitis
- Differential diagnosis: drug induced hepatitis, Wilson disease, viral hepatitis
- Immunosuppressive therapy markedly improves survival but is curative in only 20% of patients.

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CHAPTER 111

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PRIMARY BILIARY CIRRHOSIS

Primary biliary cirrhosis (PBC) is a chronic liver disease that is defined by its clinical presentation, histological features, and serologic findings. The underlying abnormality in PBC is a slowly progressive, nonpurulent inflammatory destruction of the biliary epithelial cells lining the small to medium-sized interlobular ducts. [1] The larger biliary ducts are spared. The pathophysiology of bile duct destruction is poorly understood, but the process is thought to be autoimmune in nature. Autoantibodies are a characteristic feature of the disease; over 90% of patients with PBC have antimitochondrial antibodies (AMAs) and about 30% have antinuclear antibodies (ANAs). [2] Loss of normal biliary drainage eventually leads to a clinical picture of chronic hepatic cholestasis and its complications.

PATHOGENESIS

PBC: An Autoimmune Disease?

PBC is presumed to be an autoimmune disease, although the evidence to support that hypothesis is largely inferential. Clinically, PBC shares features with other autoimmune diseases such as predominance in middle-aged women and frequent concurrence with other autoimmune disorders. [2] Epidemiologic studies have failed to provide evidence for a clear mode of transmission that could be compatible with an infectious etiology, such as parenteral exposure or fecal-oral contamination. [3] Microscopically, bile duct destruction is associated with T-cell infiltrates and expression of biliary epithelial cell surface markers of immune activation such as adhesion molecules and human leukocyte antigen (HLA) class I and II molecules. [4] In many ways, the histological picture resembles that of graft versus host disease, a condition known to be caused by T-lymphocyte–dependent immune responses directed against alloantigens. [5] In addition, virtually 100% of patients with PBC have circulating autoantibodies, often to mitochondrial or nuclear antigens. [2] The response to immunosuppressive therapy has been modest. Liver tests improve with immunosuppression, but the disease does not vanish, as it does in some other autoimmune diseases. Experimental models of PBC are not well established, which has hampered study of the pathogenesis. The best-defined features of the immunopathogenesis of PBC are the T-lymphocyte response in the liver and specific immune response directed at mitochondrial antigens.

Alternative hypotheses regarding the pathogenesis of PBC include a graft versus host disease related to fetal microchimerism or an immune response to endogenous retroviruses. Evidence of persistent fetal cells has been detected in the liver of 70% of mothers with PBC. [6] However, fetal microchimerism has also been detected by sensitive polymerase chain reaction (PCR) in an equal fraction of controls. Mason and colleagues [7] have demonstrated antibody reactivity to the human retroviral intracisternal A type particle in 51% of patients with PBC and have amplified and cloned endogenous retroviral sequences from PBC liver. These retroviruses, however, do not appear to be entirely specific for PBC, since 58% of patients with systemic lupus and 20% of patients with chronic viral hepatitis have the same antibodies.

Pathogenicity of Antimitochondrial Antibodies

In 1987, Gershwin and colleagues [8] identified the primary structure of the major target antigen of the PBC AMAs. This autoantigen was identified as the human mitochondrial enzyme, dihydrolipoamide acetyl transferase, also referred to as the E2 component of the pyruvate dehydrogenase complex (PDC-E2). PDC belongs to a family of multienzyme complexes involved in cellular respiration called the 2-oxo-acid dehydrogenase complexes. Although PDC-E2 is the major autoantigen in AMAs, patients with PBC may have antibodies to any of the 2-oxo-acid dehydrogenase complexes, including branched chain 2-oxo-acid dehydrogenase complex (BCOADC) and 2-oxoglutarate dehydrogenase complex (2-OGDC), or other components of the PDC complex (E1, E3, X) [9] (Table 111-1).

AUTOANTIBODY	PREVALENCE BY IMMUNOFLUORESCENCE (%)	PREVALENCE BY IMMUNODIFFUSION, ELISA, OR IMMUNOBLOTTING (%)
Antimitochondrial	85–95	
Anti-PDC-E2*		95–97
Anti-PDC-E1		41–66
Anti-PDC-E3		38
Anti-PDC-X		95–97
Anti-OGDC-E2		39–88
Anti-BCOADC-E2		53–55
Antinuclear	25–70	50–81
Nuclear rim/peripheral/perinuclear	26	
Anti-gp210 (nuclear pore complex)*		10–30
Anti-lamin B (nuclear membrane)*		1
Homogeneous/diffuse	10	
Antihistone		80
Anti-DS DNA		10
Centromere	10–24	
Speckled	10–20	
Multiple nuclear dot	4–17	
Anti-sp 100/anti-PML*		5–40
Coarse nuclear speckled/nucleolar		
Anti-Smith (Sm) (small RNP)		24
Anti-Scl-70 (DNA-topoisomerase 1)		24
Fine nuclear speckled		
Anti-Ro (SS-A) (small RNP)		25–30
Anti-La (SS-B) (small RNP)		21
Negative by immunofluorescence		
Anti-SS DNA		71
Anti–smooth muscle	20	
Antiactin		13–52
Antiliver membrane	42	
Anti-asialoglycoprotein receptor		15–23
Anti–carbonic anhydrase		8–45
ANCA	16–26	
Antiplatelet		40

*Autoantibodies relatively specific for primary biliary cirrhosis.

TABLE 111-1 Prevelance of Autoantibodies in Primary Biliary Cirrhosis

The role of these autoantibodies in the pathogenesis of bile duct destruction has been the subject of considerable debate. Experimental models have thus far failed to demonstrate that generation of AMAs leads to chronic destruction of bile ducts in PBC. Rats, guinea pigs, rabbits, and rhesus monkeys have all been immunized with

the recombinant form of PDC-E2. Antibodies to PDC-E2 were readily induced, but none of the animals developed abnormal liver tests or abnormal histology after 8 months of follow-up. [10] PDC-E2 is a mitochondrial enzyme that is expressed in the intracellular compartment of all aerobic cells. Tissues with high energy requirements, such as muscle, and even hepatocytes express higher levels of PDC-E2 per cell than cholangiocytes. [11] Thus, it is difficult to reconcile the apparent cholangiocyte specificity of cellular injury in PBC with the ubiquitous presence of mitochondrial antigens.

In 1991, Neuberger and colleagues reported staining of intact bile duct epithelial cells with antibodies to PDC-E2 in explanted PBC liver. [12] Similar aberrant staining was not seen in normal liver or liver disease controls. The events leading to this expression of PDC-E2-reactive material are still not understood. One hypothesis is that cholangiocytes are first injured by another process, allowing release of intracellular antigens that are then displayed on the cell surface. Another hypothesis is that mitochondrial antigens are carried to the surface complexed with immunoglobulin A (IgA) during the normal route of IgA excretion into bile. Alternatively, the abnormal surface staining could result from surface expression of an abnormal or partial-length PDC-E2, or a molecule that is cross-reactive with antibodies generated against PDC-E2. Reactivity to such a cross-reactive molecule could, in theory, originate from exposure to an infectious organism, since PDC enzymes are well conserved across species. In particular, the rough mutant of *Escherichia coli* expresses a PDC epitope that is recognized by PBC AMAs, [13] and these bacteria have been found in PBC urine and feces more frequently than in liver disease controls. [14], [15]

Clinically, AMAs appear years before other clinical signs of the disease, and thus may be present at the initiation of the disease process. However, there is no evidence that antibody and complement-dependent cytotoxicity is occurring near the small bile ducts in PBC liver. Deposits of C3d (derived from the terminal complex of complement) are not found near bile ducts [16] and immune complexes are not consistently identified in patients with PBC. [17] Furthermore, AMA titer does not correlate well with disease severity. Hence, many questions regarding the role of AMA in the pathogenesis of PBC remain unanswered.

T-lymphocyte–Mediated Destruction of Biliary Epithelial Cells

A rapidly growing body of evidence supports the hypothesis that bile duct destruction occurs through T-cell–mediated mechanisms in PBC. This is not surprising considering the abundance of T-lymphocytes in PBC portal tract infiltrates. [18] The mechanisms leading to T-cell activation are not well understood. Classically, T cells are activated after recognition of an antigen in the context of major histocompatibility complex (MHC) and costimulation by molecules on the surface of professional antigen-presenting cells. Indirect evidence suggests cholangiocytes may be able to serve as antigen presenting cells in PBC. The biliary epithelial cells of patients with PBC exhibit increased expression of HLA class I, HLA class II, and intracellular adhesion molecule 1 (ICAM-1), and have been noted to express the costimulatory molecule, B7, by some investigators. [4] Collectively, these cell surface markers create the appropriate environment in which T cells could make direct contact with cholangiocytes and recognize antigens. However, antigen presentation by cholangiocytes has not been demonstrated in vitro. [19]

Both $CD4^+$ and $CD8^+$ T cells have been implicated in the pathogenesis of PBC. In vitro, cytotoxic $CD8^+$ cells are the major lymphocyte subset that binds to cholangiocytes. [19] Aberrant staining for HLA class II on biliary epithelium, however, suggests that $CD4^+$ T-cell recognition of antigens on cholangiocytes may be important. Clinical studies examining HLA associations with PBC have reached a variety of contradictory conclusions. The most frequently observed association across studies is a weak link with HLA-DR8, with a recent study suggesting that HLA-DR8 is a risk factor for disease progression. [20] In North America and Europe, 11% to 36% of Caucasian females with PBC carry the HLA-DR8 antigen, as opposed to 4% to 9% of controls. In Japan, where HLA-DR8 is present in 23% of the normal population, HLA-DR8 is found in up to 79% of patients with PBC.

PDC-specific $CD4^+$ T cells have been demonstrated in the blood and liver of patients with PBC. About 55% of patients with PBC have circulating T cells reactive to the purified E2/X component of PDC, as compared to 1% to 13% of controls. [21], [22] Between 3% to 8% of liver-derived T-cell clones from patients with PBC are also specific for PDC. [22] In addition, T cells within PBC liver have been found to consist of oligoclonal populations, in agreement with the notion that they are antigen-specific T cells. [23] The identification of PDC-E2/X specific $CD4^+$ T cells in blood and liver of patients with PBC along with linkage of PBC to selected HLA-DR alleles supports a role for T-cell responses to mitochondrial antigens presented by cholangiocytes in the pathogenesis of PBC.

The Th1 cytokine interferon-? (IFN-?) has been found to be expressed at higher levels in PBC liver than in control livers from patients with autoimmune hepatitis or in normal liver tissue from transplant donors. [24], [25] Patients with PBC are also more likely to have an interleukin 1ß (IL-1ß) gene allele associated with high IL-1ß production. [26] It remains to be determined whether these cytokines play an important role in biliary cell death via macrophage or cytotoxic lymphocyte activation. Biliary cell death likely occurs primarily through induction of apoptosis. Fas antigen is present on biliary epithelium, and biliary epithelial cells from patients with PBC have been observed to undergo apoptosis more frequently than control biliary epithelial cells. [26]

CLINICAL MANIFESTATIONS

Clinical Presentation

The diagnosis of PBC is suspected when a patient presents with evidence of chronic cholestasis. In the early stages, this may be an asymptomatic elevation of serum alkaline phosphatase levels and/or ?-glutamyl transpeptidase. Over time, fatigue or clinical signs of chronic cholestasis such as pruritus or hypercholesterolemia may develop. Jaundice occurs late in the disease process and is a poor prognostic sign. Early reports of patients with PBC primarily described patients with jaundice and advanced disease. [27], [28] Over time, clinical case series have included a decreasing percentage of jaundiced patients and an increasing number of asymptomatic patients. [29] This phenomenon is attributed to increasing physician awareness of the disease as well as widespread use of serum alkaline phosphatase and specific autoantibody tests to help establish an early diagnosis. With earlier diagnosis, a long presymptomatic stage of PBC has become well recognized. In one study of 45 patients with PBC identified during this asymptomatic stage, 84% remained asymptomatic 5 years after the diagnosis had been originally suspected. [29] Patients with PBC may experience symptoms from associated conditions, such as sicca complex, thyroid disease, or arthritis before symptoms of the liver disease itself. [30] Eventually, however, the typical patient experiences the insidious onset of fatigue or pruritus. [31]

Signs/Symptoms

Fatigue is the most common complaint of patients with PBC. Its etiology is obscure, and its severity may be disproportionate to other disease manifestations. [31] No specific medical therapy is yet available, but lifestyle changes such as adequate rest and regular exercise may offer some benefit. [32] Concomitant depression or anxiety is not uncommon, and these co-morbidities may benefit from pharmacological therapy.

The generalized pruritus of PBC is characterized by periodic exacerbations and improvements. Pruritus is often more noticeable in the evenings, when distractions are minimal. Dry skin and decreased ultraviolet exposure during the winter months or intense heat and sweating during warm weather may intensify symptoms. Pruritus is more common in females than males, [33] and pregnancy or exogenous estrogens may precipitate or worsen the itching. The etiology of the pruritus is still obscure. The serendipitous discovery that bile acid sequestrants improve symptoms has led to the hypothesis that elevated bile acid levels are responsible. However, a poor correlation between serum or skin levels of bile acids and symptoms argues against this. [34] A centrally mediated mechanism of pruritus, potentially mediated by endogenous opioids, was proposed by Bergasa and colleagues, [35] who was able to elicit scratching in monkeys by injecting PBC plasma into their brains. Endogenous opioid levels are increased in cholestatic patients, and symptoms of pruritus are relieved by administration of opioid receptor antagonists. Interestingly, this intervention is often accompanied by symptoms of narcotic withdrawal. [34]

Right upper quadrant discomfort is present in about 17% of patients with PBC. [36] It may be present in the absence of hepatomegaly, and it has no correlation with levels of alkaline phosphatase, bilirubin, or aminotransferases, or with histological stage. It is typically nonspecific in character, is not progressive in nature, and, in fact, often disappears spontaneously.

Xanthomas and hyperpigmentation are the primary skin findings expressed in patients with PBC. [31] Xanthomas are most often seen around the eyes (xanthelasma), but may also develop over tendons and in palmar-digital creases. Hyperpigmentation, which results from increased melanin deposition, is most often found on the trunk and arms. Skin may also darken in areas that are repetitively scratched, which may result in a butterfly pattern of sparing in the middle of the back. Both xanthomas and hyperpigmentation are more often seen in patients with prolonged cholestasis but can also be seen before the onset of cirrhosis. [37]

Serologic Findings

The laboratory abnormalities seen in PBC reflect its cholestatic and immunologic nature. Elevated alkaline phosphatase and ?-glutamyl transpeptidase are the predominant feature, although aminotransferases may also be moderately elevated. Hyperbilirubinemia ensues in later stages of the disease. Prothrombin times may be prolonged due to either malabsorption of vitamin K or deteriorating hepatic synthetic function. Hypercholesterolemia often occurs as a consequence of reduced biliary excretion of sterols and impaired feedback regulation of retained bile acids. [38] In early stages of the disease, it primarily consists of high-density lipoprotein (HDL) particles. Elevated serum and hepatic copper levels also develop as a consequence of chronic cholestasis. Although impressive copper deposition can be seen in the liver, there is no evidence that this contributes to disease progression. [39] As with other etiologies of portal hypertension and cirrhosis, decreased albumin and platelet counts reflect underlying hepatic synthetic function and hypersplenism, respectively. Elevated erythrocyte sedimentation rates are not uncommon, but are not useful in following clinical status.

Almost 100% of patients with PBC have circulating autoantibodies (see Table 111-1). The most sensitive and specific autoantibodies are the AMAs, present in 90% to 95% of patients with PBC and rarely found in other conditions. [40] ANAs are not as sensitive or specific, but are also present in 25% to 70% of patients with PBC. The ANAs found in patients with PBC are comprised primarily of anti-Sp100, anti-Gp210, anticentromeres, and occasionally antilamins. Of note, some investigators have found antinuclear pore complex (anti-gp210, antilamin B receptor) or nuclear dot (anti-sp100, anti-PML) to be relatively specific markers for PBC, [41], [42] In addition, the anticentromere antibodies are specifically associated with patients that also have the CREST syndrome (calcinosis, Raynaud's phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasia). [43] It is not uncommon to find other autoantibodies that are usually associated with other connective tissue diseases, but these autoantibodies are usually an isolated finding that is not accompanied by other clinical features of the connective tissue disease. Hyperglobulinemia is not as prominent as in autoimmune hepatitis, but selective elevations of the immunoglobulin M (IgM) fraction are found in 80% to 95% of patients with PBC. [2] IgM levels may also be increased in up to 20% of patients with common bile duct obstruction and 70% of patients with alcoholic cirrhosis, but in these diseases IgM elevations are usually accompanied by similar elevations in the IgA and IgG levels. [44]

A positive serum test for AMA is characteristic of patients with PBC. When present in higher titers (=1:80), it is the most specific laboratory abnormality of this disease. Using indirect immunofluorescence, approximately 90% to 97% of patients with PBC will be AMA-positive. [40] AMAs detected by immunofluorescence are rarely associated with diseases other than PBC, but have been reported in association with syphilis, myocarditis, tuberculosis, collagen vascular disease, and iproniazid therapy. [45] Low titer (=1:40) AMAs have also been reported in individuals with other liver diseases such as autoimmune hepatitis. [46] Low titer AMAs may also be found in PBC, and these are sometimes transient. [47] Therefore, repeat testing is warranted when the clinician suspects a diagnosis of PBC, but the AMA test is initially negative. AMAs that are identified in seemingly healthy individuals may represent subclinical PBC and deserve to be followed up. Most of these asymptomatic individuals will eventually develop an elevated alkaline phosphatase, histological findings compatible with PBC, and fatigue or pruritus. [48]

Histology

The hallmark histological finding of PBC is focal and segmental nonsuppurative cholangitis. Various authors have developed histological staging systems that group the histological abnormalities into four distinct stages [49], [50] (Fig. 111-1). As described by Ludwig and colleagues, [49] stage I is characterized by lymphocytes infiltrating the portal tracts. In stage II the degree of inflammation is greater, extending beyond the limiting plate. In all systems, stage III is defined by the presence of bridging fibrosis, and the presence of cirrhosis represents stage IV.

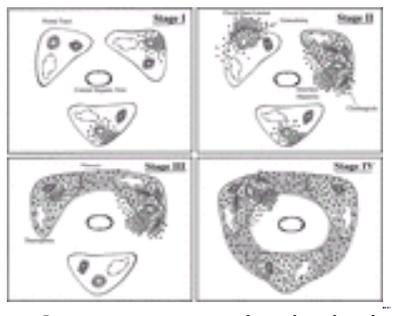

FIGURE 111-1. Histological progression of primary biliary cirrhosis (PBC) is categorized into four stages. Characteristic features of each stage are illustrated here, but the lesions are often patchy in nature. Stage I: mononuclear portal tract infiltrates, sometimes involving the bile duct. Stage II: intense mononuclear portal tract infiltrate with interface hepatitis, a florid duct lesion (granulomatous involvement of bile duct), and pseudoductular (cholangiolar) proliferation. Stage III: bridging fibrosis, mononuclear portal tract infiltrates with interface hepatitis, pseudoductular proliferation, and ductopenia. Stage IV: cirrhosis, mononuclear portal tract infiltrates with interface hepatitis, pseudoductular proliferation, and ductopenia.

In earlier stages, bile ducts may show evidence of damage, but are still present in adequate numbers. The most specific (although not pathognomonic) histological finding is the "florid duct lesion," in which the bile duct is surrounded by an intense lymphocytic or granulomatous infiltrate, and the basal integrity of the bile duct has been breached by individual lymphocytes. Granulomas in close proximity to the bile duct are also highly suggestive of PBC. Because of the patchy nature of bile duct involvement in PBC, many portal tracts remain normal or display a nonspecific pattern of mononuclear cell infiltration. The mononuclear infiltrate of PBC consists primarily of T cells, but plasma cells and eosinophils may be seen. The presence of a large number of polymorphonuclear cells, however, should prompt consideration of extrahepatic obstruction. Because interface hepatitis (piecemeal necrosis) and foci of lobular inflammation may be present in PBC, the presence of bile duct involvement is an important factor in distinguishing PBC from other forms of chronic inflammatory liver diseases such as autoimmune hepatitis. Bile staining and cholate stasis with feathery degeneration of hepatocytes is evident in some patients, and Mallory's hyaline may be found in the affected hepatocytes. Bile ductular proliferation (cholangioles or pseudo-ducts) is very common, particularly along the periphery of the portal tract, and is used as a defining characteristic of stage II PBC in the Scheuer staging system. [50] As the disease advances, ductopenia becomes evident. It is essential to have an adequate number of portal tracts on the biopsy to evaluate the presence of ductopenia. A minimum sample of 4 portal tracts (with all 4 portal tracts containing arterioles but no ducts) or ideally 20 portal tracts (with =10 portal tracts containing arterioles but no ducts) are needed to make a diagnosis of ductopenia. [51] Rhodamine and orcein stains for copper and copper binding protein are often positive in PBC liver. The intensity of staining correlates with disease severity, and copper retention may reach levels comparable to that seen in Wilson disease. [52]

Epidemiology

The prevalence of primary biliary cirrhosis in the United States and Europe is estimated to be 3.5 per 100,000 individuals (range 0.7–24) with a yearly incidence of 0.9 per 100,000. [3], [53] Some of the highest prevalences have been reported in England and Sweden, where interested researchers have conducted careful epidemiologic studies. PBC has been infrequently reported in native Africans [54] and Asians. [55] However, a higher incidence has been reported in persons from India and southern Asia who have migrated to Western countries. [56] In addition, the prevalence in Japan (5.4 per 100,000) appears to be comparable to that in many European countries. [57] Over the last two decades, the reported prevalence of PBC in North America and Europe has been increasing more rapidly than for any other autoimmune disease. [53] This increase began shortly after the discovery of the AMA and the widespread use of automated chemistry panels, suggesting that greater physician awareness and easier access to diagnostic tests is leading to an increase in the number of patients who are identified at an earlier, frequently asymptomatic phase. [29] Most patients are still identified during middle age, with a median age of presentation between 40 and 55 years of age.

Women are affected with PBC much more frequently than are men. The reported female:male ratio ranges from 3:1 to 22:1, with most studies reporting about 12:1. [3] The reason for such a strong female predilection is not known. One hypothesis is that estrogens play an important role in the development and/or clinical expression of PBC, as have been noted for other autoimmune diseases. The fact that PBC has not yet been reported in a prepubertal female also supports this hypothesis. Of note, while many autoimmune diseases improve during pregnancy, the symptoms of PBC may worsen due to an increase in cholestasis. While most features of PBC

in men are similar to those in women, men have been found to have less pruritus and fewer other autoimmune-associated conditions. [33]

The prevalence of PBC in family members of affected individuals reported from North America and Europe is near 4% (range 1%–6.4%), [3], [58] which translates into a prevalence rate nearly a thousand-fold higher than control rates. Of first-degree relatives with PBC, mother-daughter pairs are the most common. This familial clustering has been considered evidence for a genetic predisposition to PBC. A positive family history of other autoimmune disorders is quite common, and the prevalence of AMA in family members of probands with PBC is increased. [59]

Most epidemiologic studies have found neither evidence for geographic clustering nor associations with previous surgeries, pregnancies, infections, vaccinations, medications, or alcohol ingestion. [60] Only a very weak association with past smoking has been reported. Thus, the genetic and environmental factors that may be important in the development of PBC remain elusive.

Extrahepatic Conditions Reported to Be Associated With PBC

PBC is frequently accompanied by other extrahepatic conditions, many of which are also believed to be autoimmune in nature. Up to 84% of patients with PBC manifest features of other autoimmune diseases (Table 111-2).

EXTRAHEPATIC DISEASE	PREVALENCE (%)
Sjögren syndrome	30–58
Gallstones	30–50
Decreased pulmonary diffusion capacity	40–50
Renal tubular acidosis	20–33
Osteoporosis	15–40
Bacteriuria	11–35
Arthropathy	4–38
Rheumatoid arthritis	3–26
Hypothyroidism	11–32
Raynaud phenomenon	7–14
CREST* syndrome	3–6
Autoimmune thyroiditis	3–6
Autoimmune anemias	1–2
Psoriasis	1–13
Lichen planus	0.5–6
Ulcerative colitis	0.5–1

*CREST: syndrome of calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, and telangiectasias.

TABLE 111-2 Extrahepatic Diseases Associated With Primary Biliary Cirrhosis

The most common symptomatic co-morbid condition by far is Sjögren syndrome, which occurs in 30% to 58% of patients with PBC. [2], [28], [61], [62] Xerostomia and xerophthalmia of sicca syndrome are the usual manifestations. Vasculitis, renal, pulmonary, or neurological involvement are rarely clinically significant problems. Evidence of renal tubular acidosis is also present in one third of patients with PBC, but this association appears to be independent of Sjögren syndrome. [63]

The CREST syndrome is present in about 5% of PBC patients. [2], [28], [33], [62], [64] The most common features seen are Raynaud syndrome and telangiectasias. Isolated Raynaud syndrome occurs in an additional 7% to 14%. Esophageal dysmotility usually presents as gastroesophageal reflux disease or symptomatic hiatal hernia, but frank dysphagia may be present and can be worsened by concomitant xerostomia from Sjögren syndrome. Progressive systemic sclerosis, with involvement of other internal organs such as kidney and heart, is actually rare in patients with PBC.

About 10% to 20% of patients with PBC complain of arthralgias, [2], [61], [65] which may be due to a variety of causes. Because PBC often occurs in older individuals, osteoarthritis is the most common reason for joint pains. Rheumatoid arthritis is, however, more common in patients with PBC than in the general population. One study found that 53% of patients with PBC had circulating rheumatoid factor, although only 7% met established criteria for the diagnosis of rheumatoid arthritis. [66] Radiologic surveys of patients with PBC demonstrate that erosive arthritis is common and may not always be accompanied by a positive rheumatoid factor. Hypercholesterolemic arthropathy and hypertrophic osteoarthropathy are also potential reasons for joint disease in this population. [67]

Thyroid disease is another frequent co-morbid condition, present in about 20% of patients with PBC. [2], [28], [33], [61], [68] Usually this manifests as hypothyroidism, but Hashimoto thyroiditis is also common. Fatigue is a frequent symptom of PBC. However, hypothyroidism should always be excluded before attributing the fatigue to PBC itself.

Osteoporosis is observed in up to 40% of patients with PBC referred for transplant. [69] The severity of bone disease correlates with the severity of the liver disease, and therefore patients with stage IV disease should be screened with bone densitometry at regular intervals. In an unselected group of stable patients with PBC, osteopenia (defined as T score of -1.5 to -2.5) was present in 45%, and osteoporosis (defined as T score <2.5) was present in 15%. [70] Osteoporosis in patients with PBC is due to both increased resorption and decreased formation of bone. [71] Vitamin D deficiency due to malabsorption in later stages of disease may also lead to osteomalacia.

Decreased pulmonary diffusion capacity is very common in PBC, but it is usually asymptomatic. It is disputed as to whether this finding is related to Sjögren syndrome, CREST syndrome, or the presence of cirrhosis. [72] Symptomatic pulmonary disease is present with a low frequency (<1%) and has been reported to be due to a variety of conditions, including sarcoidosis, [73] interstitial pulmonary fibrosis, [74] fibrosing alveolitis, [29] bronchiolitis obliterans with organizing pneumonia (BOOP), [75] primary pulmonary hypertension, and hepatopulmonary syndrome.

Urinary tract infections are frequent, recurrent, and often asymptomatic in female patients with PBC. Up to 61% of female patients with PBC report prior urinary tract infections. [71] Upon random screening, about 20% of female patients with PBC will have bacteriuria, as compared to about 7% of age-matched female patients with other liver diseases, 5% of healthy age-matched females, and 20% of age-matched female cirrhotics. [76] At least half of patients with PBC with bacteriuria are asymptomatic. When followed over time, an additional 10% will develop bacteriuria over a 3-month interval, and 65% will develop pyuria over a 12-month interval. Recurrent infections are often with a different organism, and treatment of asymptomatic infections does not decrease recurrence.

Despite the frequent occurrence of circulating immune complexes, immune complex disease has only occasionally been reported to occur in PBC. Systemic lupus erythematosus, [77] temporal arteritis, [78] and other vasculitis syndromes can be found in patients with PBC, but are not particularly increased in frequency. Several reports of membranous glomerular nephritis and leukocytoclastic vasculitis indicate that IgM complexes can be found in the kidney and skin, suggesting that the elevated IgM levels may play a role. [79] In addition, more than a dozen cases of polymyositis with PBC appear in the literature, many of which are in Asian patients. [80]

Numerous case reports testify that celiac sprue can occur in patients with PBC. A single study from Ireland found that 11% of patients with PBC had anti-endomysial antibodies, which were often accompanied by villous atrophy. [81] However, three larger studies examining prevalence of anti-endomysial antibodies in unselected patients with PBC have failed to find a valid association. [82]

Gallstones are found by radiologic imaging in 30% to 50% of patients with PBC, particularly in later stages of the disease when pigment stones are more likely to be formed. [83] Their presence often leads to confusion and delays in diagnosis of PBC, when the patient presents with cholestatic liver test abnormalities, right upper quadrant discomfort, and gallstones, and a presumptive diagnosis of extrahepatic biliary disease is made.

Autoimmune anemias are occasionally seen (1%–2%), and anemia can be due to pernicious anemia, celiac disease-related iron deficiency anemia, or autoimmune hemolytic anemia. The autoimmune hemolytic anemias can be warm, cold, or mixed.

Inflammatory bowel disease has been reported in several patients with PBC that have been carefully characterized. [84] The coexistence of PBC and ulcerative colitis is much less frequent than in primary sclerosing cholangitis, and Crohn’s disease has not been reported as frequently as ulcerative colitis.

Skin disorders that are considered autoimmune have been reported in association with PBC. The most common is psoriasis, [60] but also lichen planus, [2] pemphigoid, [85] dermatitis herpetiformis, and cutaneous sarcoid [86] have been reported. In large screening studies of patients with psoriasis or lichen planus, PBC is very rare or nonexistent, probably reflecting the differences in disease prevalence. [87] Some reports of lichen planus may have been attributable to penicillamine treatment.

The incidence of extrahepatic cancer in PBC has been studied because of numerous case reports of cancers found in patients with PBC, as well as reports from the 1980s that the relative risk of breast cancer was increased 4.4-fold compared to other individuals. [88] More recent population studies, however, have failed to find an increased risk of extrahepatic malignancies in PBC. [89], [90]

The clinical course of associated conditions is not altered by treatment of PBC. [61] The clinician must maintain a high index of suspicion for those conditions that are commonly encountered in patients with PBC so that these diseases may be recognized and managed appropriately.

DIFFERENTIAL DIAGNOSIS

The differential diagnosis of a patient who presents with chronic cholestatic liver tests may initially include extrahepatic obstruction or intrahepatic cholestatic liver diseases, as well as granulomatous and infiltrative diseases of the liver.

Extrahepatic obstruction, from gallstones, tumors, or sclerosing diseases of the bile ducts should first be excluded with good sonographic imaging or cholangiography. Serologic testing for AMA will help to differentiate PBC from other intrahepatic cholestatic liver diseases. In patients who are AMA-negative, cholangiography is usually necessary. Liver biopsy is recommended for patients who are suspected to have PBC in order to confirm the suspected diagnosis, stage the patient, and rule out other etiologies of liver disease. In patients who are AMA-negative, liver biopsy is essential to demonstrate a nonsuppurative cholangiopathy and to exclude other disorders. [32]

A variety of diseases may resemble PBC clinically and/or histologically, but they do not lead to AMAs. Patients who have had prolonged extrahepatic obstruction may also develop a secondary biliary cirrhosis which histologically resembles primary biliary cirrhosis. A rare variant of primary sclerosing cholangitis affects only the small intrahepatic ducts. [91] However, these biliary lesions are characterized by an exuberant fibrotic reaction (“onion skinning”) and less lymphocytic infiltration. Sarcoidosis or idiopathic granulomatous hepatitis may mimic PBC with similar biochemical liver tests, right upper quadrant discomfort, hepatomegaly, and hepatic granulomas. Typically, the granulomas of sarcoidosis are larger, well-formed granulomas found in the liver parenchyma rather than involving the bile ducts. [92] It is possible to have both conditions concurrently; there are several well-documented cases of patients with AMA-positive PBC who also have multiorgan involvement with sarcoidosis. [73] A rare form of chronic hepatitis C that is cholestatic has also been reported, [93] and cirrhotic patients with hepatitis C may present with fatigue and pruritus. Because infection with the hepatitis C virus is so prevalent, viral infection with hepatitis C should be excluded in all patients with suspected PBC. It is also important to recognize that a variety of drugs have been implicated as causes of nonsuppurative cholangitis and ductopenia, which in some cases may progress to biliary cirrhosis. [94] Other rare causes of intrahepatic cholestasis, such as lymphoma and idiopathic ductopenia, are best excluded by examining liver tissue.

Differentiating primary biliary cirrhosis from autoimmune hepatitis (AIH) can be difficult because of occasional overlapping histological and serologic features. However, the distinction is essential in order to develop a safe and effective treatment program for each patient. A published scoring system by the International Autoimmune Hepatitis Group can be used to facilitate the distinction and is particularly helpful when the AMA test is negative. [95] ANAs detected by routine immunofluorescence are common in both diseases and their titer does not help to distinguish between AIH and PBC.

Autoimmune Cholangitis/AMA-Negative PBC

It is clear that a small number of patients have a clinical and histological picture similar to that of primary biliary cirrhosis, except they have atypical antibody profiles including the absence of AMAs, higher prevalence of ANAs (70% to 100%), and lower (although still elevated) total IgM levels. [96] This syndrome has been called “autoimmune cholangitis” or “AMA-negative PBC.” It is unclear whether the pathogenesis of this condition is distinct from that of AMA-positive PBC, or whether the serologic differences between the two syndromes merely reflect individual diversity of the humoral immune response. Large series of patients with autoimmune cholangitis have not been extensively examined because the frequency of this disorder is so low that few centers see more than a handful of cases. Interpretation of studies is further complicated by the lack of uniform diagnostic criteria. The limited studies that have been conducted suggest that the natural history and response to treatment of this disorder is quite similar to that of AMA-positive PBC. [97] Similar immunologic disturbances, including the presence of PDC-E2-reactive material on the surface of bile ducts and oligoclonality of T-cell populations, have been noted in both conditions. [98], [99] In fact, there is currently reluctance among many experts to embrace the hypothesis that this syndrome represents a distinct entity from the syndrome of AMA-positive PBC.

COURSE AND COMPLICATIONS

Clinical Course

PBC may exist for relatively long periods of time in an asymptomatic state. [31], [100] Indeed, data from a natural history study suggest that a fraction of asymptomatic patients will never develop symptoms during their natural life span. [100] However, most asymptomatic individuals eventually become symptomatic, and most individuals with established disease eventually progress to cirrhosis. Clinical progression is foretold by worsening of specific biochemical parameters, particularly serum bilirubin levels. The mean survival of patients with bilirubin values over 2 mg/dL is 4 years, and for those with bilirubin values over 6 mg/dL, mean survival drops to 2 years. [101] Development of variceal bleeding is not a good indicator of prognosis, since portal hypertension may develop before the onset of cirrhosis. In those cases, portal fibrosis, portal granulomas, or nodular regenerative hyperplasia are thought to cause pre-sinusoidal portal hypertension. [102] Several mathematical models exist that incorporate bilirubin and other parameters into formulas that are clinically useful to predict mortality and judge timing for transplant referral of individual patients. [103], [104] The most extensively validated and commonly used survival model is the Mayo Risk Score. [103] The Mayo Risk Score calculates an R score based on the following formula (adapted from original to use international normalized ratio [INR], personal communication with Dr. E.R. Dickson and Dr. Terry Therneau, Mayo Clinic, April, 2001):

$R = 0.871 \log_e(\text{bilirubin mg/dL}) + -2.53 \log_e(\text{albumin g/dL}) = 0.039 \text{ age (years)} = 0.881 \log_e(\text{INR}) = 5.92 = 0.859$ (edema score of 0, 0.5, or 1.0 for none, mild, or moderate edema).

The computed R score is then used to predict the fraction of patients expected to survive for:

- 1 year = 0.970 (R-5.07)
- 2 years = 0.941 (R-5.07)
- 3 years = 0.883 (R-5.07)
- 4 years = 0.833 (R-5.07)
- 5 years = 0.774 (R-5.07)
- 6 years = 0.721 (R-5.07)
- 7 years = 0.651 (R-5.07).

Of note, in all prognostic models, serum bilirubin values play a prominent role in predicting survival. Serum albumin, age, and time are found to be independent predictors of survival in some but not all prognostic models. The Mayo Risk Score has been demonstrated to be useful in judging the appropriate time for transplantation. However, in some populations, it has been found to overestimate mortality. [105] An updated version of the model, which evaluates interval changes in the patient’s condition to estimate short-term survival, is less likely to overestimate long-range mortality in patients with early disease. [106]

Complications

Once cirrhosis is established, the clinical complications of portal hypertension and deteriorating liver function are similar to those of patients with cirrhosis from other causes. The risk of hepatocellular carcinoma in patients with PBC is greatly increased compared to healthy individuals. [89] This risk is almost exclusively limited to cirrhotics, and is higher for men. In general, the reported risk of hepatocellular carcinoma in PBC is significantly less than the reported risk of hepatocellular carcinoma in other liver diseases such as hereditary hemochromatosis, hepatitis B, or chronic hepatitis C. One possible exception is the subgroup of men with advanced PBC in whom incidences as high as 20% have been reported. [90]

Pruritus is a common and often a vexing symptom of patients with PBC. Its severity often fluctuates and is not correlated with disease progression. However, in the terminal stages of the disease, pruritus often disappears. Its etiology is unknown, but elevated bile acid levels and endogenous opioid levels have been implicated. [107]

Decreased osteoblastic and increased osteoclastic activity contribute to accelerated osteoporosis in patients with primary biliary cirrhosis. [71] The risk increases with disease severity, and regular screening of bone density in cirrhotic patients facilitates treatment. Preserving bone mass is particularly important for patients who ultimately undergo liver transplantation, because bone mass decreases further in the immediate posttransplant period of heavy immunosuppression.

Like osteoporosis, deficiencies of the fat-soluble vitamins are common in patients with PBC, and correlate with disease severity. For this reason, asymptomatic cirrhotics should be screened. Poor bile salt excretion is the primary reason for deficiencies, which may be made worse by treatment with bile acid sequestrants or antibiotics. Vitamin A is frequently the first fat-soluble vitamin to become deficient, and patients may not complain of night blindness. Vitamin D deficiency may cause osteomalacia, and vitamin K deficiency may result in prolonged coagulation times. Vitamin E serum levels may be falsely elevated in patients with hyperlipidemia. A serum vitamin E to total lipid ratio of less than 0.8 mg/g is considered deficient.

TREATMENT

Therapy for PBC

Therapy with ursodeoxycholic acid (ursodiol), at a dose of 13 to 15 mg/kg/day has become the mainstay of therapy for PBC (Fig. 111-2). The medication may be administered in three divided doses or as a single evening dose. The potential mechanisms of ursodiol's beneficial effect on PBC are multiple. [108] Ursodiol is a bile acid that is less toxic to hepatocytes than other more hydrophobic bile acids. It is normally present in human bile at low concentrations (1%), but when ingested at a daily dose of 13 to 15 mg/kg, it replaces about 40% of the bile acid pool. It increases intracellular and canalicular transport of hydrophobic bile acids, thereby decreasing the potentially toxic effects of bile on the liver during cholestasis. Ursodiol stabilizes hepatocyte membranes, decreases rates of apoptosis of biliary epithelial cells, and has antioxidant properties such as the ability to inhibit nitric oxide synthetase. Ursodiol decreases expression of adhesion molecules (LFA-I, LFA-II, and ICAM-1), HLA class I and II expression in the liver, and T-cell reactivity and cytokine production.

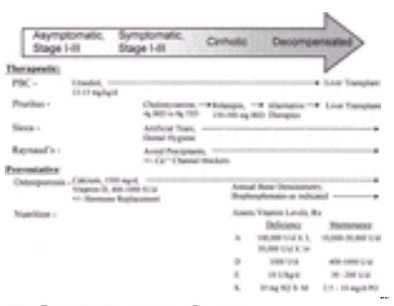

FIGURE 111-2. Management of PBC and associated conditions. (Adapted from Heathcote, J. AASLD practice guidelines: management of primary biliary cirrhosis. Hepatology 2000;31:1005.) Complications of portal hypertension (not shown) are managed in the same manner as in other forms of cirrhosis.

Five large, randomized, double-blind controlled trials have demonstrated the beneficial effects of ursodeoxycholic acid therapy. [109], [110], [111], [112] and [113] Collectively, these studies randomized 890 individuals to either ursodiol or placebo and followed them for an average of 2 years. In all trials, ursodiol produced a rapid and impressive improvement in serum liver tests, primarily alkaline phosphatase, bilirubin, aminotransferases, and IgM levels. Biochemical improvement was usually noted within the first 1 to 2 months of therapy and continued to improve, with a maximal response seen by about 6 months. Effects on symptoms were unimpressive, although a small number of patients reported an improvement in itching. Four of the five trials reported beneficial effects on histology, primarily improvement in inflammatory features and less ductopenia in the treated groups. No effect on the development of cirrhosis or portal hypertension, or death/transplantation could be detected at 2 years in any of the individual trials. However, in follow-up of one of the study groups, ursodiol therapy was associated with a delay in the onset of varices. [114] In addition, a combined analysis of patients from three of the trials, which included patients that were switched from placebo to ursodiol after 2 years, reported an improvement in transplant-free survival after 4 to 6 years in patients originally randomized to ursodiol. [115] The conclusions from this combined study, however, have been disputed by a subsequent metaanalysis of 11 randomized trials that examined outcomes of both the initial randomized treatment periods and the follow-up periods. This metaanalysis failed to show any difference between ursodiol and placebo in the incidence of death or transplantation, or development of liver disease complications. [116] The difficulty in clearly demonstrating survival benefits of ursodiol is not surprising considering the slowly progressive course of PBC and the relatively short duration of the placebo phase in most randomized controlled trials. Importantly, ursodiol was extremely well tolerated and safe in all trials. Therefore, because of its excellent safety profile, proven ability to improve markers of cholestasis, and suggestion of long-term benefits, ursodiol is recommended as treatment for all stages of PBC.

Because the prevailing viewpoint is that bile duct destruction is immune-mediated, immunosuppressive and antiinflammatory drugs have historically been selected as potential medical treatments for PBC. Unfortunately, the results of clinical trials have been disappointing or inconclusive for almost all of them. In striking contrast to ursodiol therapy, the major problems encountered in the use of immunosuppressive therapy have been their serious side effects, which have an important cumulative impact on therapy of a slowly progressive, lifelong disease.

Chlorambucil was evaluated in a controlled trial of 24 patients followed for 4 years. [117] Modest improvements in bilirubin levels, IgM levels, and inflammatory scores were seen in the chlorambucil group. However, bone marrow depression, requiring cessation of treatment in 17% of patients, was a significant problem. It was thought that the potentially dangerous side effects of chlorambucil, combined with its modest effects on the disease, made it an impractical therapeutic choice.

D-Penicillamine has been tested in 3 large controlled clinical trials on 472 patients with long-term follow-up. [118], [119] and [120] Between 22% and 46% of patients experienced serious side effects necessitating discontinuation of the drug. Despite its antiinflammatory, immunosuppressive, antifibrotic, and copper-chelating properties, D-Penicillamine was found to have no therapeutic benefit in PBC.

Prednisolone was evaluated in a controlled trial of 36 patients followed for 3 years. [121] Significant improvements in alkaline phosphatase, aminotransferases, and immunoglobulin levels were seen. Improvements in fatigue, pruritus, and bilirubin levels were transient, and there was a trend toward an improvement in liver histology. These modest benefits were outweighed by the deleterious side effects of prednisolone, especially with respect to accelerated loss of bone density. Budesonide, a corticosteroid with a high first pass hepatic metabolism, has been tested as co-therapy with ursodeoxycholic acid and compared to ursodeoxycholic acid monotherapy in two controlled clinical trials in a total of 42 patients followed for 2 years. [122], [123] Divergent conclusions were reached in the two studies. The European study found significant improvement in liver histology, liver enzymes, and immunoglobulin levels with no significant increase in bone loss in the budesonide group. The American trial, which included a larger percentage of patients with advanced disease, found marginal improvements in liver tests and significant loss of bone mass in the budesonide group.

Cyclosporine was evaluated in a multicenter controlled trial in which 349 patients were followed for up to 6 years. [124] Significant improvements in bilirubin, alkaline phosphatase, albumin, aminotransferases, and pruritus were seen, but liver histology was not affected. Survival (time to death or transplant) was modestly prolonged

in the cyclosporine group, but complications such as hypertension and renal toxicity were severe enough to discourage cyclosporine from further consideration.

Azathioprine has been tested in two controlled clinical trials [125], [126] on a total of 293 patients followed for 2 to 3 years. Both trials failed to see a survival difference between those treated with azathioprine and those treated with placebo. However, both trials also included a large percentage of jaundiced patients with advanced disease. In the European trial, when patients with higher bilirubin levels were excluded from the analysis, a slight survival advantage was seen in the azathioprine group. Azathioprine was well tolerated by patients with PBC in both studies. Nevertheless, after these trials, azathioprine was largely abandoned as a therapy for PBC.

Colchicine has both antiinflammatory and antifibrotic properties, and thus has been seriously considered as a potential therapy for PBC. Three controlled trials have compared colchicine to placebo with similar results. [127], [128] and [129] Serum levels of bilirubin, albumin, alkaline phosphatase, cholesterol, and aminotransferases were modestly improved, but symptoms and histological progression were not affected. Colchicine was not associated with any severe side effects, although some patients discontinued the drug because of gastrointestinal complaints. In the trial by Kaplan and colleagues, [127] a trend toward decrease in liver mortality was seen at 4 years. However, larger studies with longer follow-up are needed to determine the true long-term effects of colchicine.

The use of methotrexate as a therapeutic agent is still under investigation. Small, uncontrolled trials have reported dramatic biochemical and histological responses to low-dose weekly methotrexate. [130] One controlled trial of 60 patients randomized to either methotrexate or placebo for 4 to 6 years noted improvements in alkaline phosphatase, aminotransferases, and immunoglobulin levels. [131] Trends toward a decrease in hepatic fibrosis were seen. The safety of long-term methotrexate in primary biliary cirrhosis is unknown and is a valid concern. Conflicting reports exist regarding the potential pulmonary toxicity and effect on hepatic fibrosis. [131], [132]

Thus, at present, ursodiol is the only approved recommended therapy for PBC. While it is hoped that ursodiol therapy will delay liver disease progression and prolong survival, it is clear that ursodiol does not obviate the need for liver transplantation. Patients with complicated cirrhosis or poor estimated survival (Mayo Risk Score =7) should be considered for transplant referral. Ursodiol therapy does not invalidate the use of serum bilirubin or Mayo Risk Score to estimate the appropriate timing for transplant referral. [32] In general, patients with PBC have excellent posttransplant outcomes, with a 75% 5-year survival and with 95% of survivors returning to work. AMAs frequently persist after transplant, although often at lower titers than pretransplant. Recurrence of PBC in the transplanted liver is difficult to distinguish histologically and biochemically from chronic rejection and effects of immunosuppressant drugs, but it has been reported to occur in a minority of patients. Fortunately, due to slow progression of recurrent disease, it is not a significant clinical problem.

Management of Complications

Managing the complications of cholestasis comprises a considerable portion of the clinician's treatment of the patient with PBC (see Fig. 111-2).

Bile acid sequestrants, such as cholestyramine and colestipol, are effective in controlling 80% of cholestatic pruritus. Because of their efficacy and safety profile, they are the first line of medical treatment for cholestatic pruritus. [32] Unfortunately, compliance with these drugs is sometimes impaired by dislike of the powdered drug delivery system or the difficulty in timing other medication doses so that the sequestrant does not inhibit drug absorption. (Manufacturers recommend that all other medicines be taken at least 1 hour before or 4 hours after taking the sequestrant.) Because the sequestrants are not absorbed, side effects are limited to the gastrointestinal tract. Rifampin has also been demonstrated to relieve cholestatic itching in small controlled trials. [133], [134] The mechanism is unknown but is postulated to be mediated via enhancement of the mixed function oxidase system leading to an increase in the rate of bile acid metabolism and excretion and/or competition for hepatic bile acid uptake. The latter may also lead to an increase in serum bilirubin levels in some patients, which may make monitoring of disease progression difficult. At a total dose of 300 to 600 mg/day given in divided doses, the mean time to relief is 2 days, and it is also effective long term. It has been demonstrated by one group to be superior to phenobarbital therapy. [135] Phenobarbital, however, is also more effective than placebo. The mechanism of action is likely related to hepatic enzyme induction and/or sedation. The drug dosage (15–180 mg/d) is titrated to balance efficacy and sedation, but usually much lower doses are required than for the treatment of epilepsy. Antihistamines are often used to treat itching in patients with PBC, but no controlled trials have demonstrated their efficacy. In fact, there is a lack of evidence that cholestatic pruritus is histamine-mediated. However, anecdotal success in patients with mild itching may be attributable to their sedating properties. Naloxone, naltrexone, and nalmefene, opioid receptor blockers, have been demonstrated to be superior to placebo in alleviating cholestatic pruritus. [136] Unfortunately, these drugs can elicit an opioid withdrawal syndrome, which can limit the tolerability of these drugs. Several small studies have investigated *ondansetron,* a serotonin 5HT3 receptor blocker, as a potential antipruritic agent in PBC, with results ranging from dramatic to marginal. [137] The potential mechanism of action is unknown, but may be related to crossing of the serotonergic and opioid pathways. Larger controlled trials are needed to assess its efficacy. Plasmapheresis has been attempted as a desperate measure to control difficult pruritus of cholestasis. In a very small number of patients, transient success has been reported lasting anywhere from 24 hours to several months. Phototherapy has also been anecdotally successful in ameliorating cholestatic pruritus, but its effectiveness has never been adequately tested in clinical trials. Finally, some patients have intractable itching, which has been an independent indication for liver transplantation. [32]

Accelerated osteoporosis can be a devastating complication of chronic cholestasis, and careful monitoring and preventive treatment is the best therapy. Calcium supplementation (1500 mg/day for postmenopausal women and 1000 mg/day for men and premenopausal women) is well tolerated and widely used. Estrogen replacement for postmenopausal women is also well tolerated. Estrogens inhibit canalicular transport of bilirubin and have the potential to worsen cholestasis. However, the majority of patients with PBC can benefit from transdermal or low-dose oral estrogen replacement without experiencing a clinically significant increase in cholestasis. [138] The ability of bisphosphonates to increase bone density has been demonstrated in patients with PBC, so these agents are indicated when bone density is already decreased. [32] Slow fluoride has also been demonstrated in one trial to increase bone density in patients with PBC. [32] However, less overall experience and increased side effects makes this drug a less attractive choice compared to bisphosphonates.

Deficiencies in fat-soluble vitamins should be replaced. Due to potential toxicity from overdoses, levels of vitamin A and D should be monitored in patients receiving supplements.

Serum cholesterol levels are typically elevated in patients with PBC, even in early stages of the disease. However, two retrospective studies indicate that this does not translate into increased mortality from cardiovascular events. [139], [140] Potential reasons for this include:

- onset of disease during middle age rather than childhood resulting in less time for plaque formation
- a favorable HDL/total cholesterol ratio during early stages of the disease
- a favorable lipoprotein profile including the presence of lipoprotein X and low lipoprotein A levels.

Therefore, the decision to intervene medically should be based upon factors such as unfavorable lipoprotein profiles, significant personal or family cardiac history, or the presence of painful xanthomas. Patients with PBC are often resistant to dietary management of hypercholesterolemia, and they may respond paradoxically to clofibrate therapy. [141] Almost all controlled trials of ursodiol have shown significant decreases in serum cholesterol levels with ursodiol therapy. Case reports have claimed efficacy with HMG-coenzyme A reductase inhibitor, [142] or low-density lipoprotein (LDL) apheresis, [143] but no large randomized clinical trials have been published.

Many patients with PBC have sicca syndrome and are troubled by xerophthalmia nd xerostomia. Saline moisturizers (artificial tears) may be used to relieve irritation of the cornea and help prevent ulceration. Xerostomia may be relieved with adequate access to fluids, use of sugar-free hard candies, and prescription moisturizers when necessary. It is important to have regular evaluation by a dental professional, since decreased saliva secretion leads to decreased clearance of food particles and increased dental caries.

Treatment of Autoimmune Cholangitis/AMA-Negative PBC

In autoimmune cholangitis, ursodiol is effective at improving liver biochemistries, and 3-year clinical outcome is similar to that of AMA-positive PBC. [97] However, the ability of ursodiol to improve transplant-free survival in autoimmune cholangitis has not been specifically investigated. Steroid therapy may modestly improve liver biochemistries in autoimmune cholangitis, as it does in AMA-positive PBC, [144] but steroids have been associated with accelerated bone density loss and other undesirable side effects in cholestatic patients. Thus, ursodiol appears to be better tolerated as a long-term therapy than corticosteroids. Rarely, some individuals will have overlapping features of both autoimmune hepatitis and autoimmune cholangitis. Some of these patients who meet criteria for possible or probable autoimmune

hepatitis and who do not respond well to an initial trial of ursodiol may respond to corticosteroids. [144] Thus, clinical judgment must be used to determine the predominant syndrome and initiate therapy accordingly.

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CHAPTER 112

Paul C. Adams

HEMOCHROMATOSIS

Hemochromatosis is the most common genetic disease in populations of European ancestry. Despite estimates in different countries ranging from 1 in 100 to 1 in 300, many physicians consider hemochromatosis to be a rare disease. The diagnosis can be elusive because of the nonspecific nature of the symptoms. With the discovery of the hemochromatosis gene (*HFE*) in 1996, [1] new insights into the pathogenesis of the disease and new diagnostic strategies have come forth.

A fundamental issue has arisen since the discovery of the *HFE* gene. This issue concerns whether the disease “hemochromatosis” should be defined strictly on phenotypic criteria, such as the degree of iron overload (transferrin saturation, ferritin, liver biopsy, hepatic iron concentration, iron removed by venesection therapy), or whether it should be defined as a familial disease in Europeans most commonly associated with the C282Y mutation of the *HFE* gene and varying degrees of iron overload. Since the genetic test has been widely used as a diagnostic tool, most studies now use a combination of phenotypic and genotypic criteria for the diagnosis of hemochromatosis. It is important to realize that there are many causes of iron overload other than hemochromatosis (Table 112-1).

HFE-related hemochromatosis
C282Y homozygotes (95%)
C282Y/H63D compound heterozygotes (4%)
H63D homozygotes (1%)
Non-HFE related hemochromatosis
Transferrin receptor-2 mutation
Ferroportin mutation
Non-familial (may be a heterogeneous collection of conditions resulting in iron overload)
Juvenile hemochromatosis (young adults with cardiac and endocrine dysfunction)
Neonatal hemochromatosis
Miscellaneous iron overload
African-American iron overload
African iron overload
Polynesian iron overload
Transfusional iron overload
Insulin resistance–related iron overload
Aceruloplasminemia
Alcoholic siderosis
Iron overload secondary to end-stage cirrhosis
Porphyria cutanea tarda
Post-porta caval shunt

TABLE 112-1 Differential Diagnosis of Iron Overload

IRON ABSORPTION AND PATHOGENESIS OF HEMOCHROMATOSIS

In a normal human, most dietary iron is absorbed from the proximal duodenum. Both ionic iron and heme iron are absorbed across the enterocyte at the tip of the intestinal villi. Iron transport proteins at the brush border include DMT1 (divalent metal transport protein 1) and Dcytb. Iron transport within the enterocyte has not been well defined, but a number of iron-related proteins including transferrin, transferrin receptor, ferritin, iron regulatory peptide, and hepcidin have been described. The transfer of iron from the enterocyte into the portal circulation involves another series of transport proteins including hephaestin and ferroportin (IREG1).

The *HFE* gene produces a major histocompatibility complex (MHC) class 1 protein that is expressed in many cells but has a high concentration in the duodenal crypts. It interacts with transferrin receptor to facilitate iron uptake into cells. In hemochromatosis, patients have a mutated HFE protein resulting in a conformational change in the protein and this interaction is impaired. A current hypothesis is that the duodenal crypt cells develop a relative iron deficiency and when they migrate to the tip of the villi another gene is stimulated (DMT1) which increases iron absorption (Fig. 112-1). The data on HFE function and DMT1 expression have not built a conclusive case for the pathogenesis of hemochromatosis. Other iron-related genes and proteins are being investigated and it is likely that a cascade of events follows the mutation in HFE protein resulting in iron overload. [2], [3]

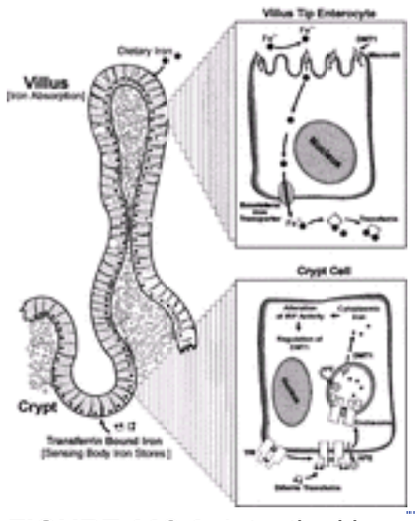

FIGURE 112-1. Intestinal iron absorption in hemochromatosis. (From Bacon BR, Powell LW, Adams PC, et al. Molecular medicine and hemochromatosis: at the crossroads. Gastroenterology 1999;116:197.)

CLINICAL FEATURES

Liver Disease

Although hemochromatosis is often classified as a liver disease, it should be emphasized that it is a systemic genetic disease with multisystem involvement. The liver has been central both in the diagnosis and prognosis in hemochromatosis. Hepatic iron deposition is common in hemochromatosis because it is the first stop on the pathway after enhanced intestinal iron absorption. The liver has a great capacity to accumulate iron within hepatocytes initially without any obvious sequelae both in terms of clinical symptoms or abnormal liver biochemistry. Since hepatic iron presumably accumulates from birth in this genetic disease a relationship between iron and age seems predictable. However, this may only apply serially within an individual patient since the correlation coefficient between age and hepatic iron concentration was not significant in the 410 homozygotes ($r = 0.07$, $p = 0.12$). [4] Hepatomegaly remains one of the more common physical signs in hemochromatosis but may not be present in the young asymptomatic homozygote. In older studies in which patients presented with clinical features of chronic liver disease in the 5th or 6th decade, cirrhosis was invariably present. As patients are detected as young adults through pedigree studies or population screening studies, the prevalence of cirrhosis is much lower. In a study of 410 homozygotes from Canada and France, 22% had cirrhosis of the liver at the time of diagnosis. The mean aspartate aminotransferase (AST) and alanine aminotransferase (ALT) were within the normal range within these 410 patients. Cirrhotic patients and patients with concomitant alcohol abuse were more likely to have abnormal liver enzymes. [5] A study of the critical hepatic iron concentration associated with cirrhosis in C282Y homozygotes using receiver operating characteristic curve analysis suggested that the critical iron concentration was 283 µmol/g (normal 0–35). [6] However, there were many patients with much higher liver iron concentrations who did not have cirrhosis. Liver damage at lower iron concentrations was usually associated with other risk factors such as alcohol abuse or chronic viral hepatitis. Therefore, it seems likely that there are factors other than iron overload that contribute to cirrhosis in hemochromatosis. It should also be emphasized that women can have significant liver involvement in hemochromatosis. In a study of 176 women, matched with men for year of birth, there was no difference in the hepatic iron concentration. [7] Many of these studies are subject to referral bias with only the sickest patients being sent for medical evaluation. Population screening studies have rarely identified patients with cirrhosis but large studies are in progress to resolve this issue. In fact, a large screening study suggested that the prevalence of symptoms in screened C282Y homozygotes does not differ from a control population. [5] Another approach has been to study hemochromatosis in relatives of the proband case. A large study from Utah demonstrated that 38% of men and 10% of women relatives had at least one hemochromatosis-related condition. [8]

Hepatocellular carcinoma has been described in 18.5% of cirrhotic patients with hemochromatosis. [9] It has rarely been described in noncirrhotic hemochromatosis patients. The relative risk is approximately 200-fold, which is similar to cirrhotic patients with chronic viral hepatitis. The effect of iron-depletion therapy has usually been stabilization of the liver disease. This may account for the relatively small number of C282Y homozygotes that have required liver transplantation. Reversal of cirrhosis has rarely been described with iron depletion. This has been previously questioned on the basis of sampling errors but evidence is increasing in other liver diseases (treatment of chronic viral hepatitis) that cirrhosis can be reversed. Posttreatment liver biopsies have been uncommonly reported; [10] they are not recommended.

Diabetes

The presence of diabetes in hemochromatosis is directly related to the presence of liver disease. [4] Many patients with cirrhosis of any etiology have glucose intolerance or diabetes and this is true for hemochromatosis as well. Earlier morphologic studies of iron deposition in the pancreas suggested pancreatic damage as the cause of diabetes. However subsequent studies have demonstrated that high-circulating insulin levels (insulin resistance secondary to liver disease) are more common than low-circulating insulin levels related to islet cell damage. Metabolic studies of insulin and glucose have not clearly demonstrated a reversal of these changes with iron depletion. This is consistent with the clinical observation that diabetes rarely resolves with therapy. [11], [12] Insulin resistance in obese patients has also been associated with iron overload in patients without the typical genetic profile for hemochromatosis. [13] Most of these cases have a moderate elevation in serum ferritin with a normal transferrin saturation.

Cardiac Disease

Cardiac disease in hemochromatosis includes both cardiomyopathy and arrhythmias. In a series of 410 patients, cardiac disease was only present in 10% of probands and 3% of discovered cases. [4] Dyspnea is the most common symptom associated with the cardiomyopathy. The cardiac iron concentration is significantly lower than the liver iron concentration. Transvenous cardiac biopsies have occasionally missed the diagnosis of hemochromatosis and should not be considered to have excluded the diagnosis. Uncommon cases of young adults presenting with life-threatening cardiac disease have been reported. These cases have been called “juvenile hemochromatosis” and preliminary studies would suggest that they are not homozygous for the *HFE* gene. [14] The putative gene for juvenile hemochromatosis has been localized to chromosome 1 in several Italian pedigrees but the gene has not yet been identified. [15] These patients can also have life-threatening ventricular arrhythmias requiring implantable defibrillators or potentially hepatotoxic medications such as amiodarone.

Arthropathy

Arthralgias are perhaps the most common symptom of hemochromatosis. Joint complaints have been found in 32% of probands and 21% of discovered cases. [4] The classic description is in the proximal interphalangeal joints of the hands but wrist, shoulder, knees, and feet are commonly affected. The features of the arthropathy are more similar to osteoarthritis and less commonly chondrocalcinosis. Radiologic features are often nonspecific but, on occasion, the diagnosis is suggested by an astute radiologist. Joint complaints are particularly common in women with hemochromatosis. Arthritis has been demonstrated to be the major factor affecting quality of life in hemochromatosis. [16] Since arthritis is common, it has been difficult to attribute the arthritis to hemochromatosis in an aging population. [17] This relationship between the background prevalence of arthritis in the female population is illustrated in Figure 112-2.

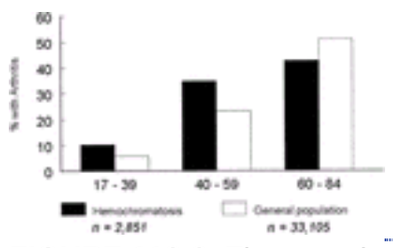

FIGURE 112-2. The prevalence of arthritis in hemochromatosis is compared to the general population in women in various age ranges. (Adapted from a survey of 2851 hemochromatosis patients and 33,105 patients in the general population. [17])

DIAGNOSIS

A paradox of genetic hemochromatosis is the observation that the disease is underdiagnosed in the general population and overdiagnosed in patients with secondary iron overload.

Underdiagnosis

Preliminary population studies using genetic testing have demonstrated a prevalence of homozygotes of 1 in 200 in patients of Northern European ancestry. The fact that many physicians consider hemochromatosis to be rare implies either a lack of penetrance of the gene (nonexpressing homozygote) or a large number of patients who remain undiagnosed in the community. Until larger genetic population studies allow us to estimate the percentage of homozygotes without iron overload, it is assumed that both of these factors are contributory.

A major problem in the diagnosis of hemochromatosis is the lack of symptoms and the nonspecific nature of symptoms. An elderly patient who presents with joint symptoms and diabetes is not often considered to have genetic hemochromatosis. Many patients assume that the routine blood tests done frequently at ambulatory clinics would have tested their iron status but this is uncommonly done unless iron deficiency is suspected. The presenting features vary depending on age and gender but fatigue is the most common complaint. Women are more likely to have fatigue, arthralgia, and pigmentation rather than liver disease. [7]

Diagnostic Tests for Hemochromatosis

A number of diagnostic algorithms based on laboratory tests have been proposed for the diagnosis of hemochromatosis. [18] These should be used as guidelines for the clinician and do not replace clinical judgment based on history and physical examination, imaging studies, pathology, and pedigree studies.

Transferrin Saturation The transferrin saturation is often elevated in patients with hemochromatosis. It is a two-step test that is widely available. It can be calculated by using the serum iron and one of the following: total iron binding capacity, unsaturated iron binding capacity, or transferrin. The transferrin saturation has been reported to have a sensitivity of greater than 90% for hemochromatosis. However, this has previously been part of the diagnostic criteria. The sensitivity of transferrin saturation is lower in population screening studies designed to detect C282Y homozygotes. A fasting value has even greater predictive value but may not always be practical. The fasting transferrin saturation is most useful in excluding false-positive cases. The transferrin saturation is often elevated in young adults with hemochromatosis before the development of iron overload and a rising ferritin. The threshold to pursue further diagnostic studies has varied from 45% to 62% in previous studies. A lower threshold picks up more patients with hemochromatosis but also leads to more investigations in patients without hemochromatosis. A higher threshold leads to fewer investigations overall with a greater possibility of missing some patients. A common threshold used in screening studies is more than 45% in women and more than 50% in men.
Unsaturated Iron Binding Capacity The unsaturated iron binding capacity (UIBC) is a one-step colorimetric assay that has been used in many reference laboratories to calculate the transferrin saturation. It is an inexpensive test compared to transferrin saturation and has been demonstrated to be a promising initial screening test for hemochromatosis. [19], [20] A UIBC less than 27 µmol/L has been proposed as a screening threshold for hemochromatosis. [21]
Serum Ferritin The relationship between serum ferritin and total body iron stores has been clearly established by strong correlations with hepatic iron concentration and amount of iron removed by venesection. [22] However, ferritin can be elevated secondary to chronic inflammation and histiocytic neoplasms. A major diagnostic dilemma in the past was whether the serum ferritin is related to hemochromatosis or another underlying liver disease such as alcoholic liver disease, chronic viral hepatitis, or nonalcoholic steatohepatitis (with or without insulin resistance). It is likely that some of these difficult cases will now be resolved by genetic testing.
Liver Biopsy Liver biopsy has previously been the gold standard diagnostic test for hemochromatosis. Liver biopsy has shifted from a major diagnostic tool to a method of estimating prognosis and concomitant disease. The need for liver biopsy seems less clear now in the young asymptomatic C282Y homozygote where there is a low clinical suspicion of cirrhosis based on history, physical examination, and liver biochemistry. A large study conducted in France and Canada suggested that C282Y homozygotes with a serum ferritin of less than 1000 µg/L, a normal AST, and without hepatomegaly have a very low risk of cirrhosis. In this multivariate model, it was more difficult to predict the presence of cirrhosis rather than the absence of cirrhosis. [23] Patients with cirrhosis have a 5.5-fold relative risk of death compared to noncirrhotic hemochromatosis patients. [10], [24] Liver biopsy is considered in typical C282Y homozygotes with liver dysfunction, and in iron-overloaded patients without the typical C282Y mutation. Simple C282Y heterozygotes, compound heterozygotes (C282Y/H63D), and patients with other risk factors (alcohol abuse, chronic viral hepatitis) with moderate to severe iron overload (ferritin more than 1000 µg/L) may be considered for liver biopsy.
Hepatic Iron Concentration and Hepatic Iron Index The traditional method of assessing iron status by liver biopsy uses the semiquantitative staining method of Perls. This is adequate when there is no iron staining or massive parenchymal iron overload. However, when moderate iron overload is present, the degree of iron overload can be difficult to interpret. Iron concentration can be measured using atomic absorption spectrophotometry. The normal reference range for hepatic iron concentration is 0 to 35 µmol/g (<2000 µg/g). The hepatic iron concentration (µmol/g) divided by age (years) is the hepatic iron index. This was demonstrated by Bassett and colleagues [25] to be a useful test in differentiating the patient with genetic hemochromatosis from the patient with alcoholic siderosis. The index remains a useful test in this clinical setting but has been extrapolated to be a diagnostic criterion for hemochromatosis. The hepatic iron index will become less useful with the advent of genetic testing. It will remain a tool to aid the clinician in clinical judgment about an individual case. It may be most useful in the unusual hemochromatosis patient that is negative by conventional genetic testing but clinically seems to have genetic hemochromatosis.

Imaging Studies of the Liver

Magnetic resonance imaging (MRI) can demonstrate moderate to severe iron overload of the liver. The technology is advancing and it is possible that eventually it may be as precise as hepatic iron determination.

Genetic Testing for Hemochromatosis

A major advance that stems from the discovery of the hemochromatosis gene is the use of a diagnostic genetic test. The original publication reported that 83% of a group of patients with suspected hemochromatosis had the characteristic C282Y mutation of the *HFE* gene. A more detailed analysis of the non-C282Y cases revealed other explanations for the iron overload in some cases resulting in more than 90% of the typical hemochromatosis patients being C282Y homozygotes. In the original report, the gene was called *HLA-H* but this name was later changed to *HFE*. [1] The C282Y mutation is also reported as 845A in some laboratories reflecting the base pair change rather than the amino acid change. Subsequent studies in well-defined hemochromatosis pedigrees reported that 90% to 100% of typical hemochromatosis patients had the C282Y mutation. [26], [27], [28], [29] and [30] The presence of a single mutation in most patients is in marked contrast to other genetic diseases in which multiple mutations were discovered (cystic fibrosis, Wilson disease, a1-antitrypsin deficiency). A second minor mutation, H63D, was also described in the original report. [1] This mutation does not cause the same intracellular trafficking defect of the *HFE* protein. Compound heterozygotes (C282Y/H63D) and less commonly H63D homozygotes [31] may resemble C282Y homozygotes with mild to moderate iron overload (Fig. 112-3A and Fig. 112-3B). These genotypes are much more common than C282Y homozygotes in the general population yet are not commonly reported in large series of typical hemochromatosis patients. This suggests that most compound heterozygotes and H63D homozygotes have normal iron studies. A polymorphism on intron 4 of the *HFE* gene (5569A) was independently reported by several laboratories to lead to false-positive genetic testing in which a C282Y heterozygote appears to be a homozygote. [32], [33] This should be considered during the evaluation of a “nonexpressing” C282Y homozygote. The correct diagnosis can be confirmed by direct DNA sequencing. Pedigrees with iron overload have been described with mutations in the transferrin receptor 2 gene and the ferroportin gene. [34], [35] and [36] It is likely that more mutations will be found but they will only be relevant to a minority of patients. The interpretation of the test in several settings is shown in Table 112-2.

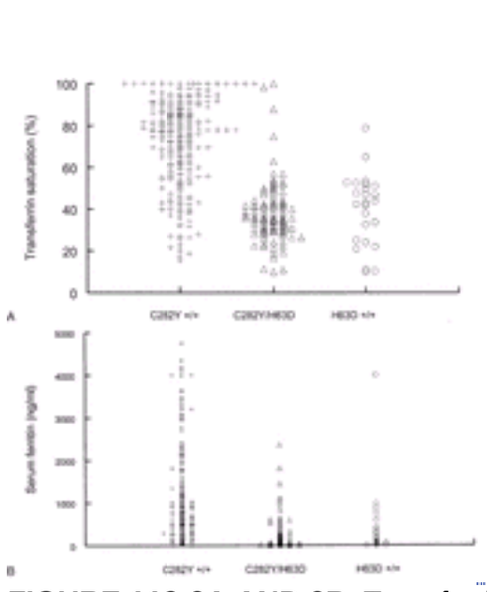

FIGURE 112-3A AND 3B. Transferrin saturation and serum ferritin in referred C282Y homozygotes, compound heterozygotes (C282Y/H63D), and H63D homozygotes. Iron studies in screening studies are usually normal in compound heterozygotes and H63D homozygotes.

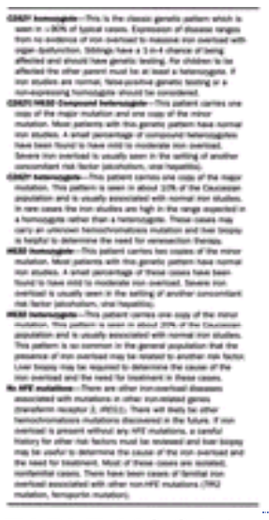

TABLE 112-2 Interpretation of Genetic Testing for Hemochromatosis

The genetic test can also be done on DNA extracted from buccal smears or from paraffin-embedded tissue such as liver explants. Studies of explanted livers have demonstrated that many liver transplant patients classified as hemochromatosis are negative for the C282Y mutation. [37] This suggests that those patients may have had iron overload secondary to chronic liver disease rather than hemochromatosis. Therefore any interpretation of iron reaccumulation after liver transplant for hemochromatosis must be cautious.

Genetic discrimination is a major concern with the widespread use of genetic testing. A positive genetic test even without iron overload could disqualify a patient for health or life insurance. [38] In the case of hemochromatosis, the advantages of early diagnosis of a treatable disease outweigh the disadvantages of genetic discrimination.

If we define the presence of homozygosity for the C282Y mutation as the new gold standard for hemochromatosis, it provides for the first time a benchmark for the assessment of the phenotypic diagnostic tools that have been used for decades. The transferrin saturation and serum ferritin with 3 HFE genotypes are shown in Figure 112-3.

Nonexpressing Homozygotes As genetic testing has become more widespread an increasing number of persons have been found with the hemochromatosis gene without iron overload. This includes siblings within well- defined hemochromatosis pedigrees. [29] The prevalence of these nonexpressing homozygotes is not yet determined from population studies but this may explain the wide discrepancy between the gene frequency and the clinician's perception that this is an uncommon disease. Pooled estimates from 14 studies have suggested that 50% of C282Y homozygotes may not have iron overload. [39] Patients who are homozygous for the C282Y mutation should be considered at risk of developing iron overload. If there are no abnormalities in transferrin saturation or ferritin in adulthood however, it seems more likely that these individuals are nonexpressing homozygotes rather than patients who will develop iron overload later in life. It seems appropriate to repeat the serum ferritin and transferrin saturation every 5 years in non–iron-loaded C282Y homozygotes to understand more about their natural history. It is important to exclude a false-positive genetic test result as previously described. In most nonexpressing homozygotes, there has been no evidence of chronic blood loss or reduced dietary intake of iron. The study of the nonexpressing homozygote may provide additional information about new genes that counteract the effect of the hemochromatosis gene.

Family Studies in Hemochromatosis Once the proband case is identified and confirmed with the genetic test for the C282Y mutation, family testing is imperative. Siblings have the highest chance of carrying the gene and should be screened with the genetic test (C282Y and H63D mutation), transferrin saturation, and serum ferritin. [40] Phenotypic expression can vary widely between siblings and I have identified identical twins with hemochromatosis with a marked difference in iron loading which suggests that environmental factors are contributory. Patients are usually very concerned about their children and may have difficulty with the concept of autosomal recessive transmission. The risk to a child is dependent on the prevalence of heterozygotes in the community and is probably greater than 1 in 20 and much lower if the spouse is non-Caucasian. A cost-effective strategy now possible with the genetic test is to test the spouse for the C282Y mutation to assess the risk in the children. [41] If the spouse is not a C282Y heterozygote or homozygote, the children will be obligate heterozygotes. This assumes paternity and excludes another gene or mutation causing hemochromatosis. This strategy is particularly advantageous when the children are geographically separated or may be under a different health care system than the parents. Genetic testing in general raises many ethical questions such as premarital testing, in utero testing, and paternity issues which have not been tested yet in hemochromatosis. Preliminary studies at our center have demonstrated that genetic testing is well accepted by patients and does not lead to an increase in anxiety. [42] If an isolated heterozygote is detected by genetic testing, it is recommended to test siblings. Extended family studies are less revealing than a family study with a homozygote but more likely to uncover a homozygote than random population screening.

DIAGNOSIS OF NON-*HFE* HEMOCHROMATOSIS AND SECONDARY IRON OVERLOAD

It is important to remember that there will be patients with a clinical picture indistinguishable from genetic hemochromatosis that will be negative for the C282Y mutation. Most of these patients will be isolated cases although a few cases of familial iron overload have been reported with negative C282Y testing. [34], [43] In non-Caucasians, iron overload is not commonly associated with HFE mutations. Therefore, the role of screening for *HFE* mutations in African Americans and Asians has not been clinically useful. Hispanic populations have been found to have HFE mutations, which is likely related to their Spanish heritage. Iron overload has been described in African Americans [44] but many of these cases had other risk factors for iron overload and pedigree studies have not been commonly reported. The iron overload described in sub-Saharan Africans may be related to another iron-loading gene [45] and a linkage to iron overload in African Americans is an intriguing hypothesis that awaits the identification of that gene.

A negative C282Y test should alert the physician to question the diagnosis of genetic hemochromatosis and reconsider secondary iron overload related to cirrhosis, alcohol, viral hepatitis, or iron-loading anemias. If no other risk factors are found, the patient should begin venesection treatment similar to any other patient with hemochromatosis. The decision to classify this group of patients as non-HFE hemochromatosis or idiopathic iron overload is a matter of semantics and ideology surrounding case definition. Quantification of iron burden by hepatic iron concentration or quantitative phlebotomy will be important to further characterize this group of patients.

In most cases of secondary iron overload, the patient has anemia with iron overload from increased iron absorption or multiple transfusions. These patients will not tolerate venesections and will require parenteral chelation therapy. Oral iron chelators such as deferiprone have been controversial because of potential hepatotoxicity.

Some patients with hepatitis C or alcoholic liver disease will be misdiagnosed as hemochromatosis. An elevated serum ferritin is common in these conditions but the hepatic iron concentration is often within normal limits. Venesection therapy of these conditions has been studied and is not recommended in patients with a normal hepatic iron concentration.

TREATMENT

The treatment for hemochromatosis continues to rely on the medieval therapy of periodic bleeding. At the University of Western Ontario, patients attend an ambulatory care facility and the venesections are performed by a nurse using a kit containing a 16-gauge straight needle and collection bag (Blood Pack MR6102, Baxter, Deerfield, IL). The patient lies in the reclining position for 15 to 30 minutes while blood is removed. A hemoglobin test is done at the time of each venesection. If the hemoglobin decreased to less than 10 g/dL the venesection schedule is modified to 500 mL every other week. Venesections are continued until the serum ferritin is approximately 50 g/L. The concomitant administration of a salt-containing sport beverage (e.g., Gatorade) is a simple method of maintaining plasma volume during the venesection. Maintenance venesections after iron depletion of three to four venesections per year are done in most patients although the rate of iron reaccumulation is highly variable. [46] The transferrin saturation will remain elevated in many treated patients and will not normalize unless the patient becomes iron-deficient. The prognosis of hemochromatosis is primarily dependent on the presence of cirrhosis at the time of diagnosis (Fig. 112-4). In some cases (particularly non-HFE related iron overload), a component of the serum ferritin elevation is related to inflammation rather than iron overload, so the ferritin does not decrease with

treatment and the patient becomes anemic. In these cases, a careful review of the liver biopsy including hepatic iron concentration may be helpful in deciding to discontinue treatment or decrease the frequency of phlebotomies.

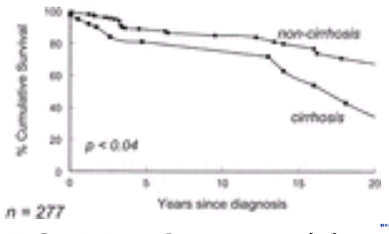

FIGURE 112-4. Actuarial survival of treated C282Y homozygotes in cirrhotic and noncirrhotic patients.

Chelation therapy with desferrioxamine is not recommended for hemochromatosis. The therapy is expensive, inefficient, cumbersome, and potentially toxic. Oral chelators such as deferiprone have side effects such as agranulocytosis and possible hepatotoxicity. Erythrocytophoresis has been used but is more expensive than simple phlebotomy therapy. Patients are advised to avoid oral iron therapy and alcohol abuse but there are no dietary restrictions. Patient support groups are discouraged by the practice of iron fortification of foods, but much of this iron is in an inexpensive form with poor bioavailability.

POPULATION SCREENING

Identification of persons at risk of developing the sequelae of hemochromatosis is preferred. The plan is to screen a population of asymptomatic individuals, with no personal or family history to suggest that they are at higher risk of the disease than the rest of the population. It aims to detect disease in presymptomatic individuals in order to provide more effective treatment in the early stages of disease. [47] Since screening programs are initially associated with increased health care costs, it is imperative that planning of screening protocols consider all the risks and benefits, and the diagnostic strategy relevant to the disease being considered prior to implementation. Population screening for hemochromatosis meets most of the criteria established by the World Health Organization for screening for medical disease.

Target Populations

It is inadequate to wait for the onset of symptoms to diagnose hemochromatosis. Many of the symptoms are unrecognized and organ damage is usually present at the time of clinical presentation. [48] In our database, 43% of referred male probands had cirrhosis of the liver (irreversible complication) at the time of diagnosis. In contrast, among the asymptomatic siblings that were then investigated, cirrhosis was present in only 6%. The ideal patient to screen is the young adult with Northern European ancestry.

Phenotypic Screening

Sporadic screening studies have been performed to establish the use of various tests and the prevalence of hemochromatosis in a target population. Target populations have included blood donors, hospital inpatients, outpatients, employees, diabetics, and military recruits. Initial testing and test thresholds also varied and have included serum iron, UIBC, transferrin saturation, ferritin and combinations of these tests. All of these studies have used phenotypic screening rather than genotypic screening. The prevalence of hemochromatosis in most of these studies is similar to studies using genetic testing which suggests that most cases will show biochemical expression of the disease.

Genetic Population Screening

A large study was conducted on 2375 Australian neonates. A prevalence of 1 in 150 were homozygous for the C282Y mutation. [49] A population study in Western Australia demonstrated a frequency in adults of 1 in 189. [50] In this study, inhabitants of Busselton, Western Australia had annual examinations and blood tests as part of a long-term epidemiologic project. When 12 C282Y homozygotes were discovered in the screening study, it was possible to determine the serum ferritin drawn 4 years previously. This is a fascinating glimpse into the natural history of untreated hemochromatosis detected at an early stage in the asymptomatic population. The serum ferritin in these homozygotes is shown in Figure 112-5. It is apparent that most but not all of these homozygotes are accumulating iron over the observation period. Another large study screened 5211 Canadian blood donors and directly compared transferrin saturation, UIBC, and C282Y genotyping. In this study, the UIBC detected more homozygotes than the transferrin saturation at a lower cost and with fewer false-positive cases. [19] A screening study of 41,038 adults attending a health appraisal clinic in San Diego, demonstrated a prevalence of C282Y homozygotes of 1 in 271. The transferrin saturation was more than 50% in only 75% of male and 40% of female C282Y homozygotes. An elevated ferritin was present in approximately 76% of male and 54% of female C282Y homozygotes. Patients were asymptomatic with a low iron burden. [5]

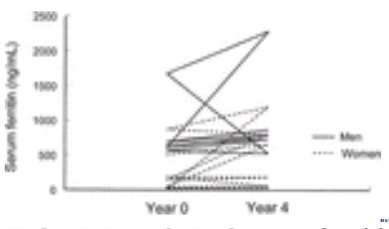

FIGURE 112-5. Serum ferritin in untreated C282Y homozygotes observed over a 4-year period. (Adapted from a population survey screening for hemochromatosis in Busselton, Australia. [50])

Since many patients have now been described homozygous for the C282Y mutation without iron overload, the role of the genetic test as the initial screening test will greatly depend on the prevalence of the nonexpressing homozygote in the general population. The preferred approach for screening at the present time is initial screening for iron overload with transferrin saturation or UIBC, with C282Y genotyping as the second line test in those with abnormal iron tests. An example of this approach is a study in 65,238 Norwegians attending a health appraisal clinic. [51] Genetic testing was only done in cases with an elevated transferrin saturation. Cirrhosis of the liver was detected in 3.7% of men and none of the women. This approach allows for the genetic testing to be focused on those with iron abnormalities. In general, the screening studies to date have demonstrated high prevalence but low morbidity. [52], [53] The natural history of untreated disease remains unknown. If untreated cases would never develop significant morbidity the use of population screening is greatly decreased.

A strong case for initial screening with the genetic test can be made in countries with a high prevalence of disease, low cost of the genetic test, and a lower risk of genetic discrimination (e.g., Ireland).

Periodic health examination in asymptomatic people has been discouraged by many health care systems. Screening in diabetic and arthritis clinics has also been studied. By the time that diabetes is present, organ damage is also evident and arthritis is a common cause of an elevation in serum ferritin which will increase the number of false-positive patients that will go on to have invasive testing. The use of genetic testing within an arthritis clinic would likely improve the screening algorithm.

It seems that population screening studies will continue using combinations of phenotypic and genotypic testing. The challenge for the future will be to devise an optimal method of sampling the population and financial support from health care systems. Physicians and third party payers need to be convinced about the

morbidity of hemochromatosis before accepting mass screening.

SUMMARY

Hemochromatosis is a common and often underdiagnosed disease. Early diagnosis and treatment results in an excellent long-term prognosis. The development of a diagnostic genetic test has improved the feasibility of the goal of prevention of morbidity and mortality from hemochromatosis.

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CHAPTER 113

Ronald J. Sokol

METABOLIC DISEASES OF THE LIVER

Hepatic involvement is a common feature in inborn errors of metabolism that are manifest in childhood and in adult life. In these disorders, it is frequent that a gene controlling a single enzyme in an important metabolic pathway or coding for a transport protein has been mutated, causing a significant block in the metabolism or transport of the substrate. Both deficiencies of important products of the enzyme and accumulation of toxic precursors lead to the disease phenotypes. In many of the genetic disorders affecting the liver, an environmental exposure to the specific substrate or toxin (galactose, fructose, copper, protein load) are important modifiers of the onset and severity of the disease phenotype. Many prior cryptogenic acute and chronic liver diseases have now been attributed to specific metabolic defects, and there are certainly many more to be discovered. It has become apparent that modifier genes most certainly play a major role in determining the clinical phenotype, severity of disease, and onset of illness for many metabolic and genetic liver diseases that were previously thought to be caused by single gene mutations.

Metabolic liver disease may present in children in a variety of clinical phenotypes, including the appearance of acute hepatitis, bacterial sepsis, metabolic coma, or as an ingestion of a toxin or drug overdose. A high index of suspicion is necessary in order to perform the correct metabolic testing and establish the diagnosis of a metabolic disorder. Some of the clinical features suggesting a metabolic liver disease include:

- hepatomegaly or splenomegaly
- cirrhosis
- hepatic steatosis
- acute fulminant liver failure
- the presence of neuromuscular or cardiac findings
- hypoglycemia, hyperammonemia
- organic acidemia
- hepatic synthetic functional failure
- mildly elevated aminotransferases
- episodes of recurrent vomiting or severe gastroesophageal reflux
- developmental delays
- failure to thrive
- chronic diarrhea
- cataracts
- abnormalities of skin or hair
- family history of similarly affected infants or children, or of consanguinity.

Most metabolic liver diseases are inherited in an autosomal recessive manner; however, several segregate into autosomal dominant and x-linked inheritance patterns. Diseases with maternal inheritance (mitochondrial DNA [mtDNA] diseases) also commonly involve the liver.

Metabolic liver diseases that present during childhood follow three patterns. A number of diseases present as a neonatal cholestasis syndrome (Table 113-1) which requires rapid diagnostic evaluation so that effective therapy can be instituted to avert permanent injury to the liver or other organs. A second mode of presentation is that of chronic liver disease with progressive hepatic fibrosis, portal hypertension, and eventually cirrhosis, with or without splenomegaly. Finally, metabolic liver diseases may present as a “metabolic syndrome,” including hypoglycemia, encephalopathy, metabolic acidosis, variable ketonemia, or lactic acidosis and/or hyperammonemia. A complete categorization of all metabolic liver diseases affecting children and adults is beyond the scope of this chapter. The reader is referred to textbooks on liver disease in children for more complete coverage of this topic. [1] This chapter will focus on five major areas of metabolic disease: Wilson disease, a1-antitrypsin deficiency, Reye syndrome, porphyria, and a group of important metabolic liver diseases presenting in early childhood.

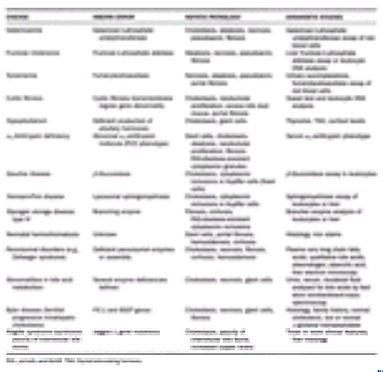

TABLE 113-1 Metabolic and Genetic Causes of Neonatal Cholestasis

WILSON DISEASE

Clinical Features

Wilson disease is caused by mutations in *ATP7B* gene which lead to progressive accumulation of copper in the liver followed by copper deposition in other organs, leading to the varying clinical features of Wilson disease. Although copper accumulates from birth, clinical symptoms are rarely apparent prior to 4 to 5 years of age. [2] Copper deposition in the liver initially causes asymptomatic biochemical abnormalities. The youngest reported patient presenting with hepatic clinical symptoms was 4 years old. [3] After the storage capacity of the liver is exceeded, circulating nonceruloplasmin-bound copper levels increase, distributing copper systemically. This redistribution of copper to other organs parallels the development of clinical manifestations of Wilson disease. Prior to 10 years of age, 80% of patients present with hepatic and 17% with neuropsychiatric manifestations. Between 10 and 18 years of age approximately 50% of patients present with either hepatic or neuropsychiatric symptoms. After 18 years of age, only 25% present with hepatic symptoms, with 75% having neuropsychiatric symptoms. [4] After hepatic saturation with copper, copper may also accumulate in the ocular cornea, leading to the characteristic Kayser-Fleischer (K-F) ring, a greenish-brown pigment at the periphery of the cornea on its posterior surface in Descemet membrane. [5] A less common ophthalmologic feature of Wilson disease is the “sunflower cataract” caused by deposits of copper in the anterior and posterior lens capsule. Wilson disease may also lead to a variety of renal disturbances including proteinuria, glucosuria, phosphaturia, generalized amino aciduria, and microscopic hematuria. Kidney stones are also common in Wilson disease, [6] and patients with the fulminant presentation of Wilson disease present with acute renal failure. Intravascular hemolysis is frequent and may be the presenting abnormality in approximately 15% of patients. Cardiac involvement has been recognized in Wilson disease with echocardiographic abnormalities present in up to one third of adults, [7] as well as arrhythmias, orthostatic hypotension, and histological changes in the heart. [7] Skeletal manifestations of Wilson disease include bone demineralization, rickets, spontaneous fractures, osteochondritis desiccants, chondromalacia patellae, and degenerative arthritis. [8] Skin pigmentation may be increased, particularly on the anterior aspect of the lower legs. [9] Various endocrinologic abnormalities have also been reported. [10], [11]

The liver is both the site of the Wilson disease biochemical defect in copper metabolism that leads to systemic disease, as well as the initial target of copper toxicity. Hepatic involvement in Wilson disease can present in one of four clinical subtypes: acute hepatitis, fulminant hepatic failure, chronic active hepatitis, and cirrhosis. Occasionally elevated aminotransferase levels are detected in asymptomatic patients for whom a chemistry panel is drawn, or in siblings of a patient with Wilson disease. It is essential that the clinician investigate children and young adults for Wilson disease if aminotransferase levels remain elevated and more common diseases are excluded.

Acute hepatitis is the mode of presentation for approximately 25% of patients with Wilson disease, with typical findings of jaundice, anorexia, nausea, malaise, pale stools, and dark urine, mimicking the features of infectious hepatitis. Laboratory tests show typically elevated aminotransferases, a normal serum albumin, and prothrombin time, with conjugated hyperbilirubinemia. However, if serologic testing for viral hepatitis is negative, Wilson disease needs to be considered. A mild hemolytic anemia or a depressed serum uric acid level (due to renal tubular losses) may be key features to alert the clinician to this diagnosis. However, the patients generally recover from this presentation, and the diagnosis may be missed.

Fulminant hepatic failure is a much more dramatic presentation of Wilson disease, although less common. It typically occurs in the adolescent or young adult who develops an acute icteric hepatitis that rapidly evolves to liver failure in a few days to several weeks with extreme jaundice, fatigue, hepatic coma, severe coagulopathy, ascites, renal failure, and invariably death, if liver transplantation is not performed. A Coombs-negative hemolytic anemia is caused by the rapid release of massive amounts of copper from the necrosing liver, and is the single most important diagnostic feature suggesting Wilson disease in this clinical setting. Interestingly, serum aminotransferase levels are not as high as in other patients with acute liver failure, alkaline phosphatase is generally very low for the age of the patient, and serum aspartate aminotransferase (AST) is significantly more elevated than the alanine aminotransferase (ALT). [12] Serum copper levels are actually elevated above normal in this presentation of Wilson disease because of the massive release of copper from the liver. K-F rings are present in no more than half of patients with this presentation, so their absence does not exclude the presence of Wilson disease. Prompt referral of patients with this presentation to a center that can perform liver transplantation is essential in order to save the patient’s life. A similar presentation of acute fulminant liver failure and death has been described in patients with Wilson disease who had been successfully chelated with medications, but who unadvisedly discontinued therapy or became noncompliant for as little as 8 months.

Chronic active hepatitis is a presentation of Wilson disease in up to 30% of patients, generally occurring during adolescence or young adulthood. Cirrhosis is common at the time of diagnosis. Typical findings of malaise, anorexia, fatigue, abdominal pain, nausea, and weight loss may precede the onset of jaundice in this scenario. Amenorrhea, delayed onset of puberty, edema, polyarthralgia, ascites, and clubbing may also indicate the chronic nature of this hepatitis. Physical examination invariably reveals hepatomegaly and usually splenomegaly. Liver biopsy shows characteristic findings of chronic active hepatitis with variable degrees of fibrosis. The histology can mimic autoimmune or chronic viral hepatitis, so a high suspicion for Wilson disease is necessary to establish this diagnosis. Importantly, histochemical stains for copper-associated proteins in the liver may be negative in patients with Wilson disease, and should never be used as the diagnostic criteria for copper overload. K-F rings are present 50% of the time, but their absence does not exclude Wilson disease. For this reason, it is imperative that biochemical screening for Wilson disease be undertaken in all patients with chronic active hepatitis who present younger than age 40 to 45 years.

Cirrhosis and its complications are relatively common features of Wilson disease in childhood and during adulthood. Virtually all adults with the neuropsychiatric presentation of Wilson disease have underlying compensated cirrhosis. Patients with cirrhosis may have an insidious presentation with cutaneous signs of chronic liver disease, splenomegaly, or onset of ascites as the initial clinical abnormalities. Other than the findings of a mild hemolytic anemia, a K-F ring if sought, positive family history, or neuropsychiatric symptoms, there is little to clinically distinguish this cause of cirrhosis from others.

Cholelithiasis is relatively common in adolescents with Wilson disease because of ongoing hemolysis and the presence of cirrhosis. Interestingly, hepatocellular carcinoma has been quite unusual in patients with Wilson disease, even those with advanced cirrhosis.

Neuropsychiatric Presentation

The onset of neurological or psychiatric symptoms is the first indication of illness in approximately 40% to 50% of patients with Wilson disease. Neurological onset has been recorded in children as young as 6 years, and in adults as old as 52 years of age. [13] Neurological symptoms generally involve the motor system presenting as manifestations of extrapyramidal or cerebellar dysfunction. [14], [15] Symptoms may be broken down into those of “pseudoparkinsonism,” pseudosclerosis, and dyskinesia, each of which correlates with specific magnetic resonance imaging features of the brain. [16] Common extrapyramidal symptoms include facial grimaces, drooling, a fixed grin, dysphasia, and contractures of the jaw and extremities. Bradykinesia, cognitive impairment, cogwheel rigidity, and an organic mood syndrome characterize the pseudoparkinsonism features. Ataxia, tremor, and reduced functional capacity are present in patients with the pseudosclerosis presentation. Because intelligence is unaffected, patients generally become frustrated and depressed with the neurological presentations of Wilson disease. The neurological basis for these abnormalities is the dysfunction of the basal ganglia and cerebellum, although corticospinal and corticobulbar pathways are also affected.

It is well appreciated that patients with Wilson disease can have a primary psychiatric presentation, including organic dementia, neurotic behavior, bipolar disorder, schizophrenic psychosis, or antisocial behavior. In children and adolescents, there may be a major change in affect or personality that impairs socialization skills and school performance. Psychiatrists must maintain a high index of suspicion for Wilson disease and measure the plasma ceruloplasmin and obtain an eye examination

for a K-F ring in all young patients with psychiatric disorders. K-F rings are present in over 90% of patients with the neuropsychiatric presentation of Wilson disease.

Hepatic Histology

Early histological findings in asymptomatic children with Wilson disease include periportal glycogen-filled, swollen nuclei [17] and hepatic microvesicular and macrovesicular steatosis. [18] In adolescents and young adults, the macrovesicular steatosis may be confused with alcoholic liver disease or nonalcoholic steatohepatitis. As the disease progresses, mononuclear cell infiltrates of the portal tracts become evident with increasing periportal fibrosis over time. Focal hepatocyte necrosis with interface hepatitis may be present in the chronic active hepatitis form of the disease. Eventually, mixed micro- and macronodular cirrhosis develops. The fulminant hepatitis lesions are characterized by microvesicular fat, coagulative cell necrosis, pigment laden Kupffer cells, collapse of stroma, and drop out of hepatocytes, with underlying well established cirrhosis. Histochemical stains for copper or copper-associated proteins may be negative despite massive accumulation of copper in the liver, and thus may be misleading. There is no correlation between histochemical staining of copper and quantitative copper measurements of liver biopsy samples in Wilson disease.

Electron microscopy may be helpful in Wilson disease because of characteristic ultrastructural lesions. In the early stage of Wilson disease, hepatocellular mitochondria are pleomorphic and large, and show widened intracristal spaces, increased matrix density, large granules, and sometimes crystalline, vacuolated, or dense inclusions in the mitochondrial matrix. There is usually microvesicular steatosis present at the same time. These mitochondrial changes diminish with progression of the lesion toward cirrhosis.

Genetics

In 1993, *ATP7B* was identified by three groups as the gene affected in Wilson disease. [19], [20], [21] and [22] By 2000, over 170 mutations in this gene have been identified from patients with Wilson disease, most being small deletions or missense mutations. FONT size=-1> [23] , [24] Unfortunately, there are no dominant mutations, although specific mutations appear to be more common among certain ethnic groups. [23] Thus, the most common mutation (*His1069Glu*) may be present in as many as 40% of cases in specific ethnic populations. Consequently, genotyping of suspected patients is a tedious task and is performed primarily in research laboratories. Haplotype analysis (microsatellite markers) may be particularly helpful in evaluating relatives of a known case in which microsatellite markers are informative. [25] *ATP7B* is located on the long arm of chromosome 13.

Pathogenesis and Biochemistry

The *ATP7B* gene is highly expressed in the liver and kidney, with lower expression in the lung, placenta, and other tissues. The encoded protein is a P-type cation-transporting ATPase, and includes domains for copper binding sites and ATP binding. *ATP7B* is located in the trans-Golgi compartment of the hepatocyte and appears necessary for the transport of copper into vesicles bound for lysosomes and eventual excretion in the bile canaliculus. *ATP7B* also makes copper available for synthesis of ceruloplasmin. When *ATP7B* is mutated in Wilson disease, both copper secretion into bile and copper incorporation into ceruloplasmin are impaired, resulting in accumulation of toxic levels of copper within the hepatocyte. The function of the hepatic copper chaperone proteins that deliver copper into intracellular targets appear to be normal in Wilson disease. Ceruloplasmin is the primary carrier by which copper is secreted from the liver into the systemic circulation. Ceruloplasmin functions to mobilize iron from tissues by oxidizing ferrous iron for transfer into transferrin. Although ceruloplasmin levels in plasma are very low in Wilson disease, the ceruloplasmin gene is unaffected.

More than 80% of absorbed copper from the diet each day is excreted in bile. Therefore, impaired biliary excretion of copper in Wilson disease leads to accumulation of this toxic metal in the liver. Copper also accumulates in the liver in other conditions that cause cholestasis; however it is primarily lysosomal-bound in those conditions and may not necessarily be hepatotoxic. In Wilson disease, the copper appears to be free to enter other compartments, such as the mitochondria. The precise intracellular target for the toxic action of copper is not known; however, recent data suggest that mitochondrial oxidative injury caused by copper overload may be one of the primary pathogenetic events in Wilson disease. [26], [27] and [28] Several studies suggest that hepatocyte apoptosis may be the primary mode of cell death in copper toxicity. [29], [30]

Diagnosis

The diagnostic criteria for Wilson disease are dependent on the clinical presentation. In patients presenting with neurological or psychiatric symptoms, the absence of a K-F ring, and a normal ceruloplasmin level virtually exclude the diagnosis of Wilson disease. However, in one series, 2 of 20 patients with neurological symptoms did not have K-F rings. [31] In the presence of normal neurological or psychiatric function, the absence of a K-F ring does not exclude Wilson disease. The common biochemical tests used for diagnosis of Wilson disease are plasma ceruloplasmin, hepatic copper concentration, and 24-hour urine copper excretion (basal and after penicillamine challenge) (Table 113-2). For patients presenting with liver dysfunction, ceruloplasmin is generally less than 20 mg/dL, 24-hour urine copper is greater than 100 µg, and hepatic copper concentration is more than 250 µg/g dry weight. Other conditions that can give rise to increased hepatic copper concentration (see Table 113-2) must be excluded with appropriate testing. Two studies [32], [33] demonstrated excellent discrimination between Wilson disease and other liver disorders when a 24-hour urine copper analysis was measured after a penicillamine challenge (500 mg given orally immediately before and repeated 12 hours into the urine collection). Values above 1575 µg of copper in this 24-hour urine collection indicated Wilson disease. Liver biopsy samples must be obtained in copper-free stainless steel needles. Quantitative hepatic copper determination is essential, with normal copper content <50 µg/g dry weight, [34] and in Wilson disease, it is virtually always above 250 µg/g dry weight, [2] if not considerably higher.

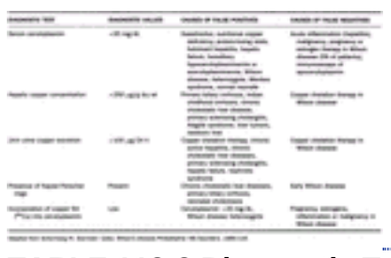

TABLE 113-2 Diagnostic Tests for Wilson Disease

Unfortunately, the assays used to analyze serum for ceruloplasmin have changed over recent years to the immunologic technique, which is in common use. Apo-ceruloplasmin is recognized by this assay, resulting in higher values than those previously measured by oxidase techniques. Therefore, a ceruloplasmin level between 20 and 30 mg/dL cannot be used on its own to definitively exclude Wilson disease. If there is a high suspicion of Wilson disease, the 24-urine copper excretion, slit-lamp examination, and perhaps liver biopsy should be performed despite a low normal level of ceruloplasmin. If a sibling has Wilson disease, microsatellite (haplotype) analysis may be another potential means to diagnose Wilson disease. In the past, incorporation of copper 64 (^{64}Cu) into ceruloplasmin could be determined; however, it is now difficult to obtain this radiolabeled copper. Finally, it may be possible to obtain genotyping of *ATP7B* from one of the research laboratories performing this analysis. If the diagnosis still remains in doubt, genotype and phenotype approaches to diagnosing Wilson disease have been published. [4]

In patients with fulminant hepatic failure, specimens to establish the diagnosis of Wilson disease may be difficult to obtain. Standard liver biopsy may be contraindicated because of coagulopathy, and the patient may be in acute renal failure, so a collection of 24-hour urine copper concentrations may not be possible. However, the diagnostic features of a severe hemolytic anemia, extremely high bilirubin, elevated serum copper values, acute renal failure, low serum alkaline phosphatase, and inappropriately mild elevations of aminotransferase levels are consistent with this diagnosis. Because these patients will either succumb to their illness or undergo a liver transplant, the explanted or autopsied liver should be evaluated for histology and for copper content.

Treatment

Wilson disease is uniformly fatal without effective therapy. Therefore the hallmark of medical management is the reduction (or chelation) of the accumulated burden of copper and preventing reaccumulation of copper. For symptomatic patients, most authorities recommend copper chelating agents and a low copper diet. For maintenance therapy, copper chelators or oral zinc therapy may be effective.

When the diagnosis of Wilson disease is established in a symptomatic patient, treatment should begin with either triethylenetetramine dihydrochloride (trientine) or D-penicillamine. In recent years it has become evident that trientine is probably better tolerated in most patients than D-penicillamine. The dose of trientine in adults and adolescents is approximately 1 to 2 g daily in 3 divided doses 1 hour prior to meals. The dose of penicillamine is also 1 to 2 g/day, given at least 30 minutes before or 2 hours after meals. [2] In young children, the dose for each of these drugs is approximately 20 mg/kg/day rounded to the nearest multiple of 250 mg given in divided doses over 24 hours. After stabilization, the dose of trientine or D-penicillamine can be divided into 2 or 3 daily doses, and reduced to approximately 1 g/day in adults. D-penicillamine may have an antipyridoxine effect, [2] thus all patients should also receive 25 mg of pyridoxine daily. It is recommended that D-penicillamine or trientine be started at one fourth to one half the desired dose and increased gradually over 1 to 2 weeks' time. Within the initial month of therapy the patient should be monitored for signs of acute allergic reactions or other toxicity. Thus a complete blood count, urinalysis, and renal and liver tests should be obtained weekly for the first month. If the patient responds appropriately, monitoring is then performed each 1 to 3 months for the first year, and then every 6 to 12 months thereafter. Some experts recommend following 24-hour urine excretion of copper to monitor chelation therapy, obtained several times in the first 6 months and then yearly thereafter. Several grams of copper should initially be excreted over 24 hours, but after several years of chelation, as little as 300 µg of copper should be excreted per day. [2] If urinary copper excretion is very low or very high, it suggests poor compliance with copper chelators. If the K-F rings had initially been present, then serial ophthalmologic examinations yearly should be performed to be certain that these lesions disappear with time. The failure of disappearance again suggests poor compliance. Many authorities recommend lifelong chelation therapy with trientine or D-penicillamine in patients who responded well to therapy. A sudden discontinuation of chelation therapy can result in fulminant liver failure which is usually fatal. [35]

It has been demonstrated that up to 30% of patients [36] will have some worsening of neurological symptoms when treated with D-penicillamine. The cause of this neurological worsening has been proposed to be a transient increase in blood copper, and then brain copper exposure as the penicillamine mobilizes hepatic copper stores; [37], [38] or alternatively, that penicillamine may form a complex of intracellular copper that is more toxic. [38] Neurological exacerbation has also been reported following initiation of treatment with trientine, [39] thiomolybdates, [40] and zinc; [41] however the frequency seems to be lower than with D-penicillamine. For these reasons, Brewer and colleagues [42] propose the use of ammonium tetrathiomolybdate for initial therapy in neurologically affected patients. This remains an investigational compound which complexes with dietary copper, thus preventing its absorption, and when absorbed complexes with copper and albumin in blood preventing cellular uptake and decreased intracellular copper stores. Concerns have been raised regarding bone marrow suppression and other toxicities of ammonium tetrathiomolybdate; this drug is under investigation.

D-penicillamine may produce both allergic and toxic effects in up to 20% of patients. Trientine appears to be associated with a lower frequency of side effects, and therefore is recommended by some authorities as a safer initial chelator for Wilson disease. Side effects of penicillamine include fever, skin rash, lymphadenopathy, pancytopenia, proteinuria, nephrotic syndrome, drug-associated systemic lupus erythematosus, Goodpasture syndrome, optic neuritis, and bone marrow suppression. Long-term therapy with D-penicillamine may cause a dermatopathy. [43], [44]

Patients who present with fulminant hepatic failure caused by Wilson disease rarely, if ever, survive unless orthotopic liver transplantation is performed. These patients require critical care therapy in a liver transplantation center fully equipped to use hemodialysis, plasmapheresis, and other means of sustaining the patient until liver transplantation.

In recent years the use of zinc has been proposed as maintenance or adjunctive [45] therapy for Wilson disease. Zinc inhibits intestinal absorption of copper by increasing metallothionein binding of copper in intestinal epithelia, which are sloughed in the gut. Some authorities recommend starting zinc acetate 50 mg of elemental zinc 3 times a day [46] or zinc sulfate 150 to 220 mg 3 times a day. [46] The zinc must be given between meals, and compliance may limit the effectiveness of this therapy. Zinc therapy is generally not used as initial therapy of symptomatic patients because of its slower onset of action. It may play a role in presymptomatic patients or siblings (especially young children) who are diagnosed with Wilson disease. Iron status needs to be monitored on patients who are undergoing chronic zinc therapy.

Finally, the use of antioxidants as adjunctive therapy in Wilson disease has not been fully explored. Because copper induces oxidative damage to the hepatocyte, [27], [47] and because a-tocopherol levels may be low during copper overload, [27], [47] it has been proposed that a-tocopherol might be an adjunctive therapy in symptomatic patients with Wilson disease. [4]

The role of orthotopic liver transplantation is well established in patients with fulminant liver failure caused by Wilson disease. In addition, patients with end-stage liver disease caused by Wilsonian cirrhosis should be considered for liver transplantation, as would other patients with cirrhosis. Long-term survival is excellent following liver transplantation. Neurological features generally improve following liver transplantation, however this currently is not recommended to treat the sole neurological presentation of Wilson disease.

REYE SYNDROME

In the 1960s and 1970s, reports of children developing an apparently new disease called encephalopathy with fatty liver appeared in Australia, the United States, and Europe. Mortality was 40% to 70%, and the disease tended to occur in epidemics linked to influenza B, varicella infection, and other viral infections. The incidence of Reye syndrome peaked in the early 1980s, rapidly falling during the late 1980s and 1990s following the restriction of aspirin use in children. [48] The decline in incidence was also associated with the discovery and identification of numerous inherited metabolic defects of fatty acid transport and mitochondrial fatty acid oxidation, which harbor similar clinical features to Reye syndrome. Therefore, it is not completely clear whether the near disappearance of Reye syndrome was the result of reduction of aspirin use or the reclassification of many patients into newly discovered inborn errors of metabolism.

Clinical Features

Reye syndrome is characterized by a prodromal febrile illness, usually a viral respiratory illness, followed by the abrupt onset of protracted vomiting after it appears that the child was in a recovery phase from the viral illness. Although cases have most often been linked to epidemics of influenza A and B and varicella infections, Reye syndrome has been reported in association with almost every viral illness. The vomiting occurs within 3 to 7 days of the onset of the viral illness, and leads to dehydration and hyperpnea. Patients then rapidly pass through graded stages of encephalopathy, progressing to seizures, cerebral edema, coma, and possibly death. In over 90% of cases in the United States, a history of aspirin use for the treatment of the initial viral illness is obtained. Physical examination reveals an anicteric state with mild to moderate hepatomegaly, neurological dysfunction, overly brisk deep tendon reflexes, and varying degrees of dehydration. Biochemical testing reveals elevated AST and ALT from several 100's to several 1000's, a normal serum bilirubin and albumin, elevated prothrombin time and international normalized ratio (INR), hyperammonemia, and hypoglycemia, particularly in young children. The Centers for Disease Control case definition of Reye syndrome includes:

- the presence of acute, noninflammatory encephalopathy with a normal cerebral spinal fluid cell count and protein
- liver involvement characterized by microvesicular steatosis, AST/ALT elevation of threefold or greater, or hyperammonemia
- no other known explanation for the patient's illness.

During epidemics, patients with Reye syndrome were generally school-age to teenage children who were previously healthy, however a subgroup of children presented within the first few years of life, frequently with underlying failure to thrive and hypoglycemia in the presentation. Interestingly, respiratory alkalosis was present, lactic acidosis was rare, but dicarboxylic organic aciduria was common.

Hepatic Histology

Reye syndrome causes extensive hepatic panlobular microvesicular steatosis, characterized by foaminess of hepatocytes with centrally placed nuclei, which may

require special fat stains to identify the microvesicular steatosis. Cholestasis is absent, and inflammation is unusual. Electron microscopic examination of the liver shows striking mitochondrial morphologic abnormalities. Variable degrees of mitochondrial swelling, pleomorphism, loss of dense bodies, and matrix edema are present, correlating with the severity of the disease. Striking microvesicular steatosis is present early in the course, progressing to macrovesicular steatosis during the recovery phase. Hepatocytes do not undergo necrosis, but rather suffer from temporary metabolic (mitochondrial) failure. The central nervous system shows marked cerebral edema, particularly in fatal cases, with changes in neuronal mitochondria resembling those in the liver. Kidney lesions also show lipidosis and mitochondrial abnormalities. Thus, Reye syndrome is believed to be caused by generalized mitochondrial dysfunction, and was one of the first well-described mitochondrial hepatopathies.

Genetics

Reye syndrome occurs in children generally between the ages of 4 and 12 years, with only very rare cases being reported in adults. Occasional cases of recurrent Reye syndrome were reported in very young children, now believed to have been caused by fatty acid oxidation defects and other metabolic disorders that were not identified at the time. Familial cases of Reye syndrome were also reported, most likely caused by autosomal recessively inherited inborn errors of metabolism.

Pathogenesis and Biochemistry

Generalized acute mitochondrial dysfunction underlies the biochemical and clinical abnormalities observed in patients with Reye syndrome. [49] The etiology of the mitochondrial lesions is most likely a combination of viral-induced, cytokine-mediated cellular injury, the effect of aspirin on mitochondrial oxidative phosphorylation in, perhaps, genetically predisposed individuals. Because Reye syndrome has been reported in patients with collagen vascular disorders receiving large doses of aspirin who did not have apparent viral infections, isolated cases may have been purely caused by the effects of aspirin. The similar clinical presentation of fatty acid oxidation defects suggests that many cases of Reye syndrome were undiagnosed defects in this important pathway for cellular energy metabolism.

The epidemiologic association between aspirin intake and Reye syndrome was established in the United States in the late 1970s. [50], [51] Subsequently, in 1982, the U.S. surgeon general advised against the use of salicylates in children with febrile illnesses, and warning labels were placed on all aspirin products. A dramatic reduction in the use of aspirin in children followed over the next several years, with a coincident decline in the number of reported cases of Reye syndrome. However, in other countries in the world, such as Australia and Asian countries, aspirin use was not associated with Reye syndrome, yet there was dramatic reduction in the number of cases. Thus, it has been proposed by some [52] that the discovery of new metabolic diseases (such as fatty acid oxidation defects) and closer examination of cases resulted in a clinical reclassification of cases that would have previously been called Reye syndrome. Others claim that the effects of salicylates on mitochondrial oxidative phosphorylation may have brought to clinical recognition cases of subclinical metabolic liver disease.

Diagnosis

The diagnosis of Reye syndrome is made clinically, based on a prodromal viral illness, an intake of aspirin products, acute onset of vomiting with progressive encephalopathy, and cerebral edema. [53] Hyperammonemia, elevated AST and ALT, prolongation of prothrombin time, and normal or only slightly elevated bilirubin are characteristic. A normal cerebral spinal fluid excludes infectious or other causes of similar illnesses. Toxin ingestions need to be excluded, as do fatty acid oxidation defects, organic acidopathies, adrenal insufficiency, urea cycle enzymopathies, and intracranial lesions resulting from brain tumors, nonaccidental trauma, and so forth. The finding of microvesicular steatosis in a noninflamed liver with characteristic mitochondrial changes on electron microscopy is consistent with the diagnosis, but may be seen in other inborn errors of metabolism as well. [54]

Treatment

Management of Reye syndrome requires early diagnosis and a high index of suspicion for the disease. Initial treatment includes controlled rehydration for the vomiting, and administration of 6 to 8 mg/kg/min of concentrated solutions of dextrose plus electrolytes, to maintain blood sugars well above 100 mg/dL. Excessive sedation is avoided and the treatment of hyperammonemia varies among institutions. Intracranial pressure monitoring is necessary for patients who reach the encephalopathy stage of delirium and combativeness, with treatment aimed at carefully reducing intracranial pressure to maintain a cerebral perfusion pressure above 45 to 50 mm Hg (intravenous mannitol, pentobarbital coma, hypothermia, etc.). [55] Complications such as pancreatitis, acute renal failure, sepsis, pulmonary hemorrhage, or cerebral hemorrhage increase the risk of mortality. Unfortunately, most contemporary cases of Reye syndrome present in stage 3 to 4 coma, and do not have acceptable control of intracranial pressure resulting in sub-optimal nervous system outcome, despite survival. It should be pointed out that the metabolic liver failure in Reye syndrome undergoes spontaneous recovery (consistent with the effects of a toxin) during the first 2 to 3 weeks of illness, except for continued steatosis.

ALPHA-1 ANTITRYPSIN DEFICIENCY

a1-Antitrypsin deficiency affects 1 in 600 to 1 in 2000 live births. The PiZZ homozygous phenotype is an autosomal recessive disorder in which there is defective secretion of a1-antitrypsin from the liver, resulting in serum levels of only 10% to 20% of normal. The mutant a1-antitrypsin Z protein is trapped in the endoplasmic reticulum of the hepatocyte, causing liver injury in 10% to 20% of affected individuals. [56] The protein is in the family of circulating serine protease inhibitors called *serpins,* and is an approximately 55-kd secreted glycoprotein which inhibits neutrophil proteases and elastases. For reasons that are not clear, only 10% to 20% of PiZZ affected individuals develop clinically significant liver disease during the first 20 years of life. In adults, emphysema is common in affected homozygotes, particularly in association with cigarette smoking. a1-antitrypsin deficiency is the most common genetic cause of liver disease in children that leads to cirrhosis, and hence, is the leading genetic indication for orthotopic liver transplantation.

Clinical Features

Persistent neonatal cholestatic jaundice is the most common childhood presentation of a1-antitrypsin deficiency. Some infants occasionally present with acholic stools and may mimic the clinical appearance of biliary atresia. Affected infants tend to have been small for gestational age at birth. Serum aminotransferase levels, alkaline phosphatase, and ?-glutamyl transpeptidase may all be elevated. Hepatomegaly associated with progressive hepatic fibrosis may or may not be present on physical examination. Occasionally patients present with a secondary vitamin K deficiency coagulopathy causing bleeding in the first several months of life. Other affected infants and young children will present abruptly with symptoms of portal hypertension, such as esophageal variceal bleeding or hypersplenism. A very small number of patients present in acute fulminant liver failure during infancy. In adults, a1-antitrypsin deficiency may cause chronic hepatitis, cirrhosis, portal hypertension, or present as hepatocellular carcinoma. [57] There is also a slight increase in the risk of cryptogenic cirrhosis in adults who are PiMZ heterozygotes.

Sveger [58] conducted a prospective study, examining a1- antitrypsin phenotype in 200,000 newborn infants in Sweden. Of those 200,000 infants, 127 PiZZ infants were detected, 14 of which had cholestatic jaundice in infancy, 9 that had severe liver disease. Approximately 50% of the PiZZ individuals had abnormal aminotransferase levels without any other indication of liver involvement. Through childhood, more than 85% of the 127 patients had normalization and persistently normal aminotransferase levels with no evidence of liver disease or liver dysfunction. This prospective study in Swedish children suggests that only 5% to 10% of all PiZZ children will develop significant liver disease. However, it is well appreciated that subclinical hepatic fibrosis and even cirrhosis can occur in this disease without elevated liver blood tests.

Several other alleles of a-1 antitrypsin have been associated with liver disease. Children who are phenotype PiSZ appear to develop liver disease in a similar manner to PiZZ affected children. PiSS affected children do not develop liver disease. Other rare forms of a1-antitrypsin deficiency have occasionally been associated with liver disease. [59], [60]

It is rare for emphysema or any significant pulmonary involvement to affect children with a1-antitrypsin deficiency. In one study there were no significant differences between pulmonary function of PiZZ children and that of an age-matched control group between 13 and 17 years of age. [61] In adults over the age of 25 years, approximately 60% to 70% with a1-antitrypsin deficiency may develop variable degrees of emphysema, generally in the 4th or 5th decade.

Hepatic Histology

Liver histology in PiZZ homozygotes ranges from a benign-appearing, almost normal liver to one of extensive cirrhosis. The classic hallmarks of this deficiency are the

periodic acid-Schiff–positive (PAS), diastase-resistant globules that appear in periportal hepatocytes. These globules represent retained a1-antitrypsin material in the endoplasmic reticulum of hepatocytes. Globules may not be easily recognized in PiZZ infants until up to 6 months of age. [62] In the neonate, a giant cell hepatitis with multinucleated giant cells, lobular disarray, cellular, and canalicular cholestasis is commonly observed in affected infants. Occasionally, bile duct proliferation suggests biliary atresia. As the child becomes older, a more benign appearance of bridging hepatic fibrosis with minimal or no giant cell transformation is observed. The liver can also have the appearance of a chronic active hepatitis, or show increased iron deposition. Approximately 10% to 30% of infants with cholestatic jaundice evolve into a mixed macro- and micronodular cirrhosis. Bile ducts are generally normal; however, in 5% to 10% of children who present with neonatal cholestasis, paucity of interlobular bile ducts will develop, causing a chronic cholestatic disease.

Genetics

a1-Antitrypsin is encoded by a 12.2-kb gene located on human chromosome 14 q31-32. [2], [63], [64] Structural variances of the a1-antitrypsin protein are classified by the protease inhibitor phenotype system defined by agarose electrophoresis or isoelectric focusing of plasma. [64] A letter is assigned to variants according to the position of migration of a1-antitrypsin in these gel systems. The more common normal variant migrates in an intermediate isoelectric point. It has been designated M. The most severe deficiency form migrates to a high isoelectric point, and has been designated as Z. More than 100 allelic variants of a1- antitrypsin have been reported, all inherited in an autosomal recessive fashion. With some variants there is no a1-antitrypsin detectable in serum, the so-called null variants. The variants that have been associated with low serum concentrations of a1- antitrypsin are known as deficiency variants. Not all of these are associated with clinical disease, such as the S variant; emphysema and liver disease have been associated with the PiSZ, PiZZ, M $_{malton}$, and M $_{duarte}$ variants. The heterozygote state does not confer disease susceptibility, except for the small possibility that the PiMZ heterozygote state increases risk for cryptogenic cirrhosis in adults.

Pathogenesis and Biochemistry

The pathogenesis of liver injury in a1-antitrypsin deficiency has not been completely defined. It is clear that the accumulation of mutant a1-antitrypsin protein in the endoplasmic reticulum of liver cells is involved in its pathogenesis. In fact, transgenic mice carrying the mutant Z allele of the human a1-antitrypsin gene developed PAS-positive, diastase-resistant globules in the liver as well as liver injury. [65], [66] It is difficult, therefore, to understand why only 10% to 15% of patients develop significant disease, since all PiZZ affected individuals have globules present in their liver. Perlmutter and colleagues have demonstrated delay of degradation of mutant a1-antitrypsin Z protein in fibroblasts from PiZZ individuals with liver disease as compared to those without liver disease, [67] providing evidence that other factors affecting the fate of mutant a1-antitrypsin molecules may play a role in determining the genetic susceptibility to liver disease. This important role for a modifier gene may help explain the 10% to 15% incidence of significant liver disease in this and other single gene mutation metabolic disorders. [68]

Diagnosis

Diagnosis is established by isoelectric focusing or agarose electrophoresis of a patient's serum a1-antitrypsin. Generally, total serum levels of a1-antitrypsin are found to be 10% to 20% of the normal values with the PiZZ phenotype. Establishing the exact phenotype assists in determining the risk of the patient for liver disease. For example, the PiSS phenotype elicits no risk for liver disease, whereas PiSZ and PiZZ do pose this risk. All infants with neonatal cholestatic jaundice require a1-antitrypsin testing, inasmuch as it may be very difficult to clinically or histologically identify this disease at that time. Infants with a1-antitrypsin deficiency may mimic the presentation of biliary atresia, including acholic stools, failure of biliary excretion on nuclear scintigraphy, and liver biopsy findings that are similar. It is said that a1-antitrypsin concentrations may increase during the acute phase response, however serum concentrations never exceed 50 to 60 mg/dL in patients with the PiZZ phenotype, allowing one to use serum concentrations as a screen for deficiency. The finding of PAS-positive, diastase-resistant globules on liver biopsy should never in itself be used to either diagnose or exclude the diagnosis of a1-antitrypsin deficiency.

Treatment

There is no specific therapy for a1-antitrypsin deficiency–associated liver disease; however, several measures are taken in infants with the cholestatic presentation. Fat-soluble vitamin supplementation and an infant formula containing medium chain triglyceride oil are administered until the cholestasis resolves. In addition, ursodeoxycholic acid is frequently given to increase bile flow and reduce liver injury from cholestasis, as in other neonatal cholestatic diseases. Although early studies suggested that breast feeding might be beneficial, [69] subsequent data from the Swedish Nationwide Screening Study did not support this contention. [70] Approximately 10% to 30% of infants diagnosed with a1-antitrypsin deficiency who have neonatal cholestasis progress within the first 5 years to cirrhosis and end-stage liver disease. The remaining 70% to 90% of patients either show complete recovery from liver injury, or have variable degrees of chronic elevated aminotransferases and progressive hepatic fibrosis.

Complications of portal hypertension are treated as they are in other childhood liver diseases. Orthotopic liver transplantation is required for children with end-stage liver disease, in whom hepatic synthetic functional failure or severe complications do not respond to standard therapy.

Patients with emphysema caused by a1-antitrypsin deficiency are now treated with replacement therapy with purified or recombinant plasma a1-antitrypsin, either by intratracheal aerosol administration or by intravenous administration. [71] Improvement in serum concentrations of a1-antitrypsin without significant side effects results from these therapies. One nonrandomized trial suggested that there was slower decline in forced expiratory volume in 1 second in patients on replacement therapy. [72] Lung transplantation has been performed in a number of patients with severe emphysema. Actuarial survival is approximately 50% at 5 years. [73] a1-antitrypsin is certainly a disease that could benefit from successful gene replacement therapy: continued investigation in this arena will take place over the next decade.

Finally, siblings of all affected patients with the PiZZ phenotype should be screened by isoelectric focusing or agarose gel electrophoresis to determine their phenotype. If identified as a homozygote, strict avoidance of cigarette smoke and other air pollutants may help prevent the onset of emphysema later on in life.

PORPHYRIA

Inherited deficiencies or toxic inhibition of the enzymes involved in the heme synthesis pathway leads to eight disorders called the porphyrias. Each of these porphyrias is characterized by the accumulation of specific substrates or precursors of heme that provide characteristic biochemical footprints in blood, urine, and feces, which allow for differentiation of the type of porphyria. The varied accumulated compounds, rather than the decrease in synthesis of heme, are responsible for the cutaneous, hepatic, neuropsychiatric, and hematologic manifestations of the porphyrias. Three of the porphyrias are inherited in autosomal recessive fashion, and five by autosomal dominant inheritance. Most patients present during or after puberty; however, onset in infancy or early childhood has been reported. [74], [75], [76], [77], [78], [79] and [80] Five of the porphyrias are expressed exclusively in the liver, two in both liver and erythroid cells, and one type has only erythroid expression. [74], [75], [76] and [77] Porphyrias are generally classified into the acute porphyrias and the cutaneous porphyrias. In the acute porphyrias, aminolevulinic acid (ALA) is increased. In the cutaneous porphyrias, porphyrins rather than ALA accumulate. Diagnostic studies and clinical features of the major porphyrias are outlined in Table 113-3.

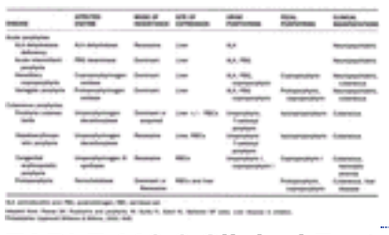

TABLE 113-3 Clinical Features of Human Porphyrias

Acute Porphyrias

Four hepatic porphyrias present with acute attacks of potentially life-threatening neurological dysfunction. These attacks generally begin around puberty, and may

diminish after the 5th decade of life. The most common symptoms include abdominal pain, constipation, and vomiting, presumably caused by autonomic nervous system dysfunction of the gastrointestinal tract. The peripheral neuropathy may develop into a flaccid quadriplegia with respiratory compromise in severe cases. Psychiatric symptoms may include depression, psychosis, and hysterical behavior. [74], [75] and [76], [81], [82], [83], [84], [85], [86] and [87] The pathogenesis of neurological symptoms is related to the ALA blood levels, although the exact mechanism by which the ALA functions as a neurotoxin has not been determined. Drugs and toxins that increase cytochrome P450 activity may precipitate acute attacks; relative iron deficiency and cyclical progesterone excretion in menstruating females may be responsible for the higher frequency of acute attacks in women compared to men.

Hepatic abnormalities in the acute porphyrias are characterized by mild AST and ALT elevations during attacks. Histological examination of the liver reveals steatosis and increased iron deposition. Intramitochondrial crystalline inclusions have also been described. [81], [82], [88], [89], [90], [91] and [92] A disturbing high incidence of hepatocellular carcinoma has been described in patients with inducible porphyrias. [93]

Acute Intermittent Porphyria

The most common form of acute porphyria is acute intermittent porphyria (AIP). The defective enzyme is porphobilinogen (PBG) deaminase and the disease is of autosomal dominant inheritance. The prevalence of the disorder is approximately 1 in 1000 to 2000. [81], [94] Diagnosis is established by the determination of large amounts of ALA and PBG in the urine during acute attacks, with PBG excretion above that of ALA. A characteristic of AIP is a black or red color of a patient's urine after exposure to light or air. In asymptomatic patients, there is no increase in PBG or ALA excretion in the urine. A diagnosis is then established by enzyme analysis in liver tissue, red blood cell lysates, or genetic analysis of leukocyte DNA. [82], [95]

Treatment of mild AIP attacks includes management of fluids and electrolytes, avoidance of drugs that may precipitate symptoms, and observation. Severe neurological symptoms are treated with hematin, which inhibits hepatic ALA-synthetase activity, yielding lower circulating levels of ALA. [96], [97] Liver transplantation has been used in very unusual circumstances for severe, unremitting symptoms. [98]

Other Acute Porphyrias

Hereditary coproporphyria (HCP), variegate porphyria (VP), and ALA dehydratase deficiency comprise the other acute porphyrias. HCP and VP are autosomal dominant. The urine co-proporphyrin excretion is elevated in these two disorders, with fecal protoporphyrin being present in VP. Cutaneous manifestations predominate over neurological symptoms in these conditions. The accumulation of porphyrins in the skin is responsible for the cutaneous lesions. Neurological symptoms can resemble those of AIP. ALA dehydratase deficiency is a very rare autosomal recessive disorder, yielding exclusive excretion of ALA in the urine. The disease is characterized by severe acute neuropsychiatric porphyria episodes that are poorly responsive to medical treatment. [99] Treatment of neurological attacks in all of these conditions follows the protocol for the treatment of AIP attacks.

Cutaneous Porphyrias

Cutaneous lesions without neurological or psychiatric involvement characterize four of the porphyrias. However, significant hepatic involvement may be observed. These diseases include porphyria cutanea tarda (PCT), hepatoerythropoietic porphyria (HEP), congenital erythropoietic porphyria (CEP), and protoporphyria. Two types of skin photosensitivity develop in the cutaneous porphyrias. Skin lesions appear to be the result of photoexcitation of porphyrins located in the upper levels of the epidermis or walls of dermal blood vessels with resulting reactive oxygen species generation leading to tissue injury by a variety of proposed mechanisms. [100], [101], [102], [103], [104] and [105] Patients with PCT, HEP, and CEP show increased skin fragility in light exposed areas, manifested by the formation of bullous lesions, secondary scarring, and hypertrichosis. Patients with protoporphyria show immediate photosensitivity with itching, burning, and erythema soon after light exposure. Although they do not develop bullae, chronic skin changes evolve into leathery, hyperkeratotic skin in light exposed areas.

Porphyria Cutanea Tarda

PCT is the most common form of porphyria in North America and Europe, inherited as an autosomal dominant disorder or acquired during chronic hepatitis C and other infectious diseases. PCT may also be associated with toxins, particularly halogenated hydrocarbons. The genetic form of PCT is caused by deficiency of uroporphyrinogen decarboxylase. Urine and fecal uroporphyrins are elevated (see Table 113-3), however PBG levels are normal. Most patients with PCT present in adulthood, with photosensitivity skin reactions, scarring, infection, and lichenification of the skin. Because many individuals with the identical enzyme deficiency remain asymptomatic, others have sought coexisting abnormalities that must be present in order to develop overt disease. These include iron overload, a mutated allele in the *HFE* (hereditary hemochromatosis) gene, the presence of hepatitis C viral infection, or Alagille syndrome. Ingestion of ethanol, toxins, and estrogens may also be involved in the development of sporadic cases of PCT.

Liver involvement is a frequent finding in PCT, manifested by hepatomegaly, focal inflammation, granuloma formation, and hepatic hemosiderosis. Progression to cirrhosis is not uncommon. [106], [107] and [108] It remains controversial whether there is an increased risk for hepatocellular carcinoma in patients with PCT. [109], [110]

Therapy for PCT includes phlebotomy or chelation therapy to reduce the iron burden. Chloroquine has been used to form complexes with uroporphyrin to enhance removal of these deposits from skin. These treatments may be combined in patients with significant liver disease, however their efficacy is still unclear. [76], [79], [106], [111]

Protoporphyria

Erythropoietic protoporphyria is one of the most common forms of porphyria in North America, caused by a defect in ferrochelatase. The enzyme can be measured in liver and bone marrow cells, being less than 50% of normal in almost all cases. Affected patients have elevated levels of erythrocyte, plasma, and fecal protoporphyrin. [112], [113] and [114] The primary clinical manifestation of protoporphyria is photosensitivity, which generally appears in childhood as burning, pruritus, erythema, and edema in light exposed areas. Scarring and lichenification of the skin develop with time. Avoidance of light, use of sunscreens, and oral administration of carotenoids appear to be the most efficacious treatment for the skin lesions. Approximately 5% to 10% of affected patients develop significant hepatic involvement, with reports in children as young as 10 years of age. Jaundice is generally the first indication of hepatic dysfunction, however rapid progression of decompensated cirrhosis may lead to liver failure within several years of clinical presentation. [115], [116], [117], [118] and [119] The liver is black in color, enlarged, and exhibits fibrosis and macronodular cirrhosis. Dark brown pigment, which represents protoporphyrin deposits, is found in hepatocytes, Kupffer cells, and small biliary ducts. Soluble protoporphyrin appears to plug small bile ducts interrupting bile flow and may initiate oxidative injury to hepatocytes. It is unclear why only 5% to 10% of patients develop progressive liver disease, but other gene modifiers or environmental factors must be involved. Other manifestations of protoporphyria include hemolysis, neurological dysfunction, and gallstones.

Treatment of individuals with liver disease is aimed at reduction of protoporphyrin levels in the liver. Iron supplementation and blood transfusions have been used in the past to diminish erythropoiesis, thereby reducing protoporphyrin production. Hematin has similarly been shown to decrease plasma protoporphyrin in several patients. [120] Interestingly, administration of chenodeoxycholic acid has been demonstrated to increase protoporphyrin excretion into bile. [76], [77], [78] and [79], [111] Cholestyramine and activated charcoal both bind protoporphyrin, interrupting the enterohepatic circulation and increasing fecal excretion. [82], [121] These modalities do decrease circulating levels of protoporphyrin, however they have not been shown to conclusively affect the course of the liver disease. Although orthotopic liver transplantation has been performed in several patients with end-stage liver disease, protoporphyrin production continues in the bone marrow, depositing protoporphyrins in the newly transplanted allograft. [122], [123] In addition, skin lesions and neurological symptoms may continue, complicating the postoperative course.

Congenital Erythropoietic Porphyria

CEP is an autosomal recessive disorder caused by a deficiency of uroporphyrinogen III co-synthase. It is quite rare with fewer than 100 patients reported. The major manifestation is photosensitivity, hypertrichosis, and hemolysis. Liver disease is present and may progress to cirrhosis, with iron overload playing a significant role. [76], [79], [111], [124] Treatment includes limiting light exposure, sunscreens, and beta carotene. Hematin or blood transfusions to decrease erythropoiesis have also been attempted in patients with liver disease.

METABOLIC DISEASES OF THE NEONATE AND OF CHILDHOOD

Hepatic involvement may be the presenting symptom of a variety of metabolic disorders occurring primarily or exclusively in infancy and childhood. Because effective treatments for many of these diseases have been identified over the past decades, patients are now surviving into adulthood. Disorders of amino acid, carbohydrate, lipid, ammonia, and mitochondrial metabolism may all have hepatic involvement as the primary clinical manifestation. It is beyond the scope of this chapter to describe in detail all childhood metabolic liver diseases, and the reader is referred to other sources for further details. [1] This chapter will focus on several of the most common metabolic liver diseases presenting in infancy and early childhood.

Hereditary Tyrosinemia

Hereditary tyrosinemia is caused by a deficiency of the enzyme fumarylacetoacetate hydrolase (FAH), the terminal enzyme in the pathway of phenylalanine and tyrosine degradation. Although the gene for the enzyme has been cloned, there are no definite genotype-to-phenotype correlations in severity of the disease. A number of metabolites of tyrosine accumulate proximal to the FAH block, including tyrosine and succinylacetone. [125], [126] Succinylacetone and succinylacetoacetate inhibit enzymes such as porphobilinogen synthase, which causes the build-up of ALA and symptoms similar to that of acute intermittent porphyria. [127], [128] It is unclear how accumulated toxic compounds initiate liver injury in tyrosinemia.

Tyrosinemia may present in infancy as acute hepatic failure, neonatal cholestasis, rickets, failure to thrive, or as compensated or decompensated cirrhosis later in childhood. There is also a very high incidence of hepatocellular carcinoma in affected children. Episodes of acute peripheral neuropathy may lead to pain, respiratory depression, and death. [129] Children who present during infancy have decreased survival compared to those with a later presentation in childhood. [130]

Liver histology is characterized by macrovesicular steatosis, pseudo-acinar formation of hepatocyte rosettes, hemosiderosis, and varying degrees of hepatocyte necrosis and apoptosis. A fine diffuse fibrosis develops, eventually progressing into a micronodular cirrhosis. Regenerative nodules are frequently present, and frank hepatocellular carcinoma may occur in patients as early as 2 years of age. The incidence of tyrosinemia is approximately 1 in 100,000; however, there are pockets of high frequency secondary to a founder effect, such as 1:1800 live births which occurs in the Saguenay-Lac Saint Jean region of Quebec, Canada. [131]

Hereditary tyrosinemia diagnosis is suggested by features of metabolic liver disease in infants or older children, including diminished hepatic synthetic function (hypoglycemia, coagulopathy, hypoalbuminemia) in the face of only mildly elevated aminotransferases. Bilirubin may be elevated or normal, and hypophosphatemic rickets may be present. Renal tubular dysfunction may cause glycosuria, proteinuria, amino aciduria, and hyperphosphaturia. Serum a-fetoprotein levels are usually extremely high for age. The diagnosis of tyrosinemia is established by the finding of succinylacetone in the urine. In some research laboratories, FAH activity can be measured in red blood cells. Genetic diagnosis is generally not necessary, however in Quebec, 100% of patients have a single mutation of the FAH gene. [132] Hypertyrosinemia is a nonspecific finding and is not used to establish this diagnosis. Neonatal screening is being carried out in high incidence regions, such as Quebec.

Treatment of hereditary tyrosinemia involves a diet restricted in phenylalanine and tyrosine, although this diet does not prevent liver disease but may have beneficial effects on kidneys. Orthotopic liver transplantation reverses the metabolic abnormalities and reduces the risk for hepatocellular carcinoma, prevents neurological disease, and is usually associated with stabilization of kidney disease. This generally had been considered the preferred option for children with tyrosinemia over the age of 1 to 2 years. However, a recent metabolic inhibitor, NTBC [2-(2-nitro-4-trifluoromethylbenzoyl)-1,3-cyclohexanedione] has been shown to be effective in many cases with tyrosinemia. This compound inhibits 4-hydroxyphenyl-pyruvate dioxygenase, the second enzyme involved in tyrosine degradation, which, when inhibited, prevents the production of succinylacetone. Therefore, toxic precursors do not accumulate, liver disease improves dramatically, patients exhibit reduction of a-fetoprotein levels, liver synthetic function improves, and renal tubular damage may resolve. To date, well over 150 patients have been treated with NTBC in an ongoing investigation of its long-term effectiveness. The major concern is that mice who were homozygous for the FAH gene mutation, and who develop tyrosinemia, respond to NTBC but still develop hepatocellular carcinoma. [133] Whether or not treatment with NTBC will reduce the high risk for development of hepatocellular carcinoma in children with tyrosinemia is unknown. The current recommendation is for continued surveillance for hepatic masses in children under long-term NTBC therapy. Neurological symptoms are treated with intravenous dextrose, pain control, NTBC, or hematin.

Galactosemia

Classic galactosemia is an autosomal recessive disorder resulting from a deficiency of the enzyme galactose 1-phosphate uridyltransferase. Patients develop chronic cholestatic liver disease, failure to thrive, mental retardation, and cataracts. Two other conditions result in elevated plasma galactose concentrations, however, without observable liver disease. These include uridine diphosphate 4-epimerase deficiency and galactokinase deficiency, the latter causing only juvenile cataracts. Classic galactosemia is a disease in which toxicity of the liver results from ingested dietary galactose and its failure to be converted to glucose. Most cases are now detected by newborn screening programs, such that it is an unusual patient who presents with neonatal liver disease.

The classic presentation of galactosemia is that of vomiting, diarrhea, and failure to thrive following the institution of lactose-containing infant feedings—either breast milk or lactose-containing infant formula. Within weeks to months of birth, hepatomegaly, cholestatic jaundice, and cataracts develop in the affected infant. If left completely untreated, mental retardation as well as cirrhosis of the liver will develop. Another classic presentation is that of *Escherichia coli* urinary tract infections and bacteremia, sometimes with a fatal course. Affected females may show ovarian failure due to in utero injury from the galactose toxic products. Laboratory findings include elevated serum aminotransferase levels, prolonged prothrombin time, conjugated hyperbilirubinemia, and hypoalbuminemia. Hemolytic anemia occurs frequently. Renal tubular dysfunction is characterized by generalized amino aciduria in combination with proteinuria and galactosuria.

Pathogenesis Galactose-1-phosphate accumulates in classic galactosemia, causing injury to the affected organs. Galactose may also be reduced to galactitol, which causes cataracts when it accumulates in the lens of the eye. It appears that galactose-1-phosphate or galactosamine accumulation is responsible for the renal, ovarian, hepatic, and brain abnormalities. Galactose-1-phosphate may also interfere with phosphoglucomutase, an enzyme that allows the release of glucose from glycogen. Liver biopsy reveals macrovesicular steatosis in the first months of life, with pseudoacinar transformation of hepatocytes, but giant cell transformation is generally absent. If left untreated over the first several months of life, portal fibrosis and the development of cirrhosis accompany bile duct proliferation. The diagnosis of galactosemia is suggested by the detection of reducing substances in the urine with a negative glucose-oxidase paper test on the urine. Since vomiting or poor intake of lactose-containing formula or breast milk may limit galactose excretion in the urine, false-negative results of urine reducing substance tests may occur in galactosemia. More specific diagnosis is based on measurements of erythrocyte galactose 1-phosphate uridyltransferase. If an infant with galactosemia has received a red blood cell transfusion, the red cell enzyme level may be raised into the normal range and the diagnosis missed. Newborn screening programs have been established across the United States and in many areas of the world. Measurement of enzyme activity for galactose compounds is the analysis performed on newborn blood spots.

Treatment Treatment of galactosemia is based on the complete elimination of dietary galactose. Infant lactose and galactose-free formulas have been marketed around the world and are instituted as soon as the diagnosis is made. As the child becomes older, a complete lactose and galactose-free diet must be maintained. [134], [135] In recent years, it has been suggested that endogenous production of some galactose may be the mechanism that underlies some of the delayed toxicity present in children maintained on a lactose and galactose-free diet. [136] Follow-up studies have revealed that some patients treated from infancy still suffer poor growth, learning disabilities, and primary ovarian failure in females. [136]

Hereditary Fructose Intolerance

Hereditary fructose intolerance is an autosomal recessive disorder caused by deficiency of the fructose-1-phosphate aldolase B enzyme activity in the liver. Symptoms occur when fructose is added to the infant's diet, usually in the form of fruit juices or infant formulas that contain sucrose. [137], [138] Infants develop vomiting, irritability, hypoglycemia, and even convulsions following the ingestion of fructose. If fructose is not removed from the diet, hepatomegaly, failure to thrive, chronic diarrhea, conjugated hyperbilirubinemia, and metabolic dysfunction ensue, including proximal renal tubular abnormalities. [139] Older children may have a self-aversion to sweets, knowing that these tend to make the child feel ill. Growth delay, consequences of repeated episodes of hypoglycemia on brain development, and metabolic acidosis eventually complicate the clinical picture.

Liver histology is characterized by diffuse macrovesicular steatosis, scattered hepatocellular necrosis, bile duct proliferation, and periportal fibrosis. Cirrhosis has also been described with complications of portal hypertension. The toxic moiety leading to liver injury is accumulated fructose-1-phosphate. Both hypoglycemia and ATP

depletion in hepatocytes are caused by the accumulation of this compound. [139]

Diagnosis of hereditary fructose intolerance is suggested by the onset of hypoglycemia, a profound hypophosphatemia, and hyperuricemia following fructose ingestion with detection of fructose in the urine by chromatography. Several common mutations in the gene for this enzyme allow for genetic diagnosis in most cases. [140] The incidence of the disease is approximately 1 in 20,000 newborns. [141], [142] Because the enzyme is also present in jejunum, it has been suggested that jejunal biopsies can be used for enzyme analysis to establish the diagnosis.

Treatment of hereditary fructose intolerance is based on the complete and lifelong elimination of fructose, sucrose, and sorbitol-containing foods, juices, medications, or other compounds from the diet. [137], [143], [144] Low levels of chronic fructose ingestion may lead to growth failure [143] and persistent hepatic steatohepatitis.

Glycogen Storage Diseases

Excessive accumulation of glycogen or abnormal synthesis of glycogen in the liver or muscle tissue is the underlying basis for a group of diseases called the glycogen storage diseases (GSD). Seven of the glycogen storage diseases primarily involve the liver causing a variety of hepatic manifestations. Types Ia, Ib, Ic, and III lead to hypoglycemia and characteristic metabolic derangements; types III, IV, and IX are associated with periportal fibrosis; and types VI and IX are associated with benign hepatomegaly. Understanding the underlying enzymatic defects in each of these disorders has led to a better understanding of the pathophysiology and the development of treatment modalities (Table 113-4). The hepatic glycogen storage diseases will be discussed in the remainder of this chapter.

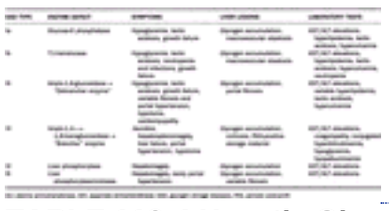

TABLE 113-4 Hepatic Glycogen Storage Diseases

GSD Type I There are three forms of GSD type I involving the glucose-6-phosphatase enzyme system. Glucose-6-phosphatase catalyzes the last step in the enzymatic degradation of glycogen with release of glucose into the blood. Failure of proper function of glucose-6-phosphatase leads to profound hypoglycemia during the fasting state. GSD type Ia involves deficiency of the glucose-6-phosphatase enzyme, GSD type Ib involves abnormalities in the translocase responsible for movement of glucose-6-phosphate across microsomal membranes to the site of action of glucose-6-phosphatase. GSD type Ic has been reported with a deficiency of the phosphate-pyrophosphate translocase. [145] GSD type Ia (von Gierke disease) is characterized by recurrent episodes of profound and severe hypoglycemia, lactic acidemia, hyperuricemia, hypertriglyceridemia, and hypercholesterolemia. The hypoglycemic events frequently start during infancy, when the interval between feedings becomes prolonged beyond 2 to 3 hours, or when the child starts sleeping for longer durations at night. The child generally suffers from growth failure and short stature, extreme hepatomegaly without splenomegaly, bleeding episodes caused by platelet dysfunction, renomegaly, and a cherubic appearance secondary to the deposition of “baby fat” in the face and trunk. The abdomen is protuberant, and arms and legs are frequently thin. Impaired glucose production through glycogenolysis leads to overproduction of glucose-6-phosphate which is shunted into pathways for synthesis of lactate, triglycerides, cholesterol, and uric acid. [146] As a result, glucagon levels are high and insulin levels are low. Patients suffer from growth failure if untreated or poorly treated, and eventually multiple hepatic adenomas, which may develop into hepatocellular carcinoma. As glucose-6-phosphatase is also expressed in the kidney, enlarged kidneys are common, and nephrocalcinosis or glomerulosclerosis may also develop over time. [147] Hyperuricemia may contribute to chronic renal disease as well as to gouty arthritis. [148] Progressive hepatic fibrosis and portal hypertension virtually never occur in this disorder, however hepatomegaly is extremely prominent. The diagnosis is established by demonstrating elevated hepatic glycogen content and low activity of glucose-6-phosphatase on frozen liver tissue. Liver histology shows markedly enlarged hepatocytes with pale cytoplasm causing a “mosaic” appearance histologically. There is also macrovesicular steatosis and an accumulation of glycogen in both cytoplasm and nucleus under electron microscopy. There is no hepatic fibrosis, cirrhosis, or chronic inflammation. Treatment of GSD-1a is directed at providing a continuous source of exogenous glucose to prevent hypoglycemia and the resultant hormonal and metabolic changes. Frequent daytime high starch meals along with uncooked cornstarch supplements are provided. Cornstarch is given in doses of 1.5 to 3 g/kg every 3 to 6 hours as needed. In order to maintain blood sugar at night, constant drip feedings of a high glucose elemental formula are provided through either a nasogastric tube or a gastrostomy tube. The aim is to maintain blood sugars of at least 70 mg/dL. As children become older, many opt to discontinue constant drip feedings at night, and ingest uncooked cornstarch at bedtime, awakening every 4 hours at night to drink a dose of cornstarch. Either of these techniques can effectively maintain blood sugar, reverse metabolic abnormalities, and provide adequate growth. [149], [150], [151], [152], [153], [154] and [155] However blood sugars need to be monitored periodically. It should be pointed that the hypoglycemia is better tolerated when the children are hyperlipidemic and acidotic than when they are adequately treated with cornstarch or constant drip feedings. However, once the patients are well controlled with these modalities, the children may react severely to episodes of hypoglycemia, with neurological depression, convulsions, and even death. For patients who cannot be controlled with these therapeutic modalities, and for patients with multiple hepatic adenomas, liver transplantation has been advocated. [156] However, the vast majority of patients can maintain a high quality of life with their native liver.

GSD Type Ib GSD-Ib presents in a similar manner as GSD-Ia, however patients also have neutrophil dysfunction and frequently are neutropenic. Therefore, they have a propensity toward recurrent bacterial infections, inflammatory bowel disease-like lesions, and oral ulcerations. [157], [158], [159], [160] and [161] The diagnosis of GSD-Ib is suggested by the finding of neutropenia in a patient who appears to have all of the other classic features of GSD type Ia (hypoglycemia upon fasting, lactic acidosis, elevated AST, ALT, hypertriglyceridemia, hyperuric acidemia, hepatomegaly, growth failure). This disorder is diagnosed by measurements of liver glucose-6-phosphatase activity. In GSD type Ib, which involves the glucose-6-phosphate T_1 translocase enzyme, fresh liver homogenate will yield defective activity, whereas frozen liver homogenate will have normal glucose-6-phosphate activity. The reason for this finding is that freezing disrupts the microsomal enzyme barrier, allowing access of the substrate to glucose-6-phosphatase, thereby bypassing the need for the translocase enzyme. In fresh homogenate tissue, the microsomes are intact, thus necessitating the requirement for the translocase enzyme. This disease is treated similarly to GSD type 1a, with the addition of granulocyte colony stimulating factor to elevate neutrophil counts.

GSD Type III GSD type III is due to the deficiency of debrancher enzyme activity, [146] also known as amylo-1,6-glucosidase. This enzyme degrades the 1,6 branch points of the glycogen molecule after hepatic phosphorylase has hydrolyzed the outer branches. Patients with GSD type III have either liver and muscle involvement (type IIIa), or liver involvement alone (type IIIb), the latter being the minority of patients. The patients may present in identical manner to GSD type Ia, with hepatomegaly, hypoglycemia, hyperlipidemia, hyperuricemia, growth retardation, and similar biochemical features. However, blood lactate and uric acid levels may be normal in GSD type III, with moderate elevations of serum AST and ALT. Patients have the potential to develop progressive portal fibrosis, leading to cirrhosis and complications of portal hypertension. [162] Myopathic weakness may worsen as the patient ages into adulthood. Cardiomyopathy with ventricular hypertrophy may also occur in type III patients. GSD type III is inherited in an autosomal recessive manner, the human gene being an 85-kb gene composed of 35 exons located on chromosome 1p21. Mutations in this gene have been described in both types IIIa and IIIb GSD. Liver histology in GSD type III resembles that found in type Ia, however, there is less steatosis, and periportal fibrosis progressing to cirrhosis may be present. [163] Treatment of type III GSD depends upon the symptomatology. In patients with hypoglycemia, acidosis, and hypertriglyceridemia, treatment is similar to that in type Ia GSD. However, many patients with GSD type III can be managed on a high starch diet, without the need for cornstarch therapy. There is one report of a high protein diet and continuous nocturnal infusion of a high protein formula improving muscle strength in a patient with myopathy. There is no other effective treatment for the progressive myopathy or cardiomyopathy.

GSD Type IV GSD type IV is caused by a deficiency of the brancher enzyme, amylo-1,4,6-transglucosidase, present in leukocytes and in liver. An abnormal glycogen is formed that contains few branch points, which appears to be toxic to hepatocytes. [164] Inheritance is autosomal recessive. The human gene coding for this enzyme is on chromosome 3p12. There appears to be a phenotype-genotype correlation which may explain clinical variability. [165] Patients may present with three clinical scenarios:

1. Progressive liver injury and cirrhosis, leading to liver failure and death during the first 5 years of life
2. Chronic liver disease without progressive fibrosis
3. Abnormal neuromuscular development with hypotonia, muscle atrophy, and absent deep tendon reflexes.

Cardiomyopathy may also develop because of deposition of amylopectin in myocardial cells. Adults with milder symptoms, but with similar muscle histology have been described. [165] Children with early onset liver failure present with failure to thrive, hepatosplenomegaly, cholestatic jaundice, and occasionally hypotonia. Complications of cirrhosis such as ascites and liver synthetic dysfunction prevail and death usually occurs by the 2nd to 4th year of life. [166] Diagnosis is established by finding the

characteristic abnormal structure of glycogen on electron microscopy in muscle, cultured fibroblasts, or liver. Verification of deficient branching enzyme activity is determined in muscle, leukocytes, cultured fibroblasts, or liver. Liver histology reveals micronodular cirrhosis and hepatocytes that appear to contain excessive glycogen, which is PAS-positive and diastase-resistant. [167] Electron microscopy shows large irregular areas of glycogen fibrils, and finely granular material. [167] Liver transplantation appears to be the only effective therapy for children with progressive liver failure and cirrhosis. [168], [169] Although many patients have done well following liver transplantation without cardiac or muscle involvement, a single report suggests that cardiac disease may be a posttransplantation issue. [170]

GSD Type VI and IX GSD type VI and IX involve deficiencies of the phosphorylase system, which stimulates degradation of hepatic glycogen. Deficiency of hepatic phosphorylase causes GSD type VI, of which there are many subtypes. Patients generally present with asymptomatic hepatomegaly, elevated aminotransferases, variably with hypoglycemia or hyperlipidemia. Growth retardation occasionally occurs, but many children are found to be normal in stature. Lactic acidosis is rare and the prognosis is generally very good. Hepatomegaly improves as the patient reaches adulthood, and growth also is generally normal by adulthood. Liver biopsy findings include glycogen-filled hepatocytes, but generally the absence of inflammation, cell injury, and fibrosis. GSD type IX is caused by phosphorylase b kinase deficiency. This disorder is characterized by asymptomatic hepatomegaly, mild elevation of aminotransferases, glycogen-filled hepatocytes on liver biopsy, and variable degrees of hepatic fibrosis. Slow progression to cirrhosis has been reported in several cases with the development of portal hypertension. Diagnosis is established through enzyme analysis of liver and erythrocytes. [171], [172] Phosphorylase b kinase deficiency is more common than the true phosphorylase deficiency. Rarely, patients with type IX GSD develop biochemical features of type I or type III GSD. Treatment is generally not necessary for GSD type IX.

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CHAPTER 114

David W. Crabb and Lawrence Lumeng

ALCOHOLIC LIVER DISEASES

EPIDEMIOLOGY

Alcoholic liver disease (ALD) continues to be a major cause of cirrhosis and death around the world. In the United States, there are at least 14 million problem drinkers and alcoholics, [1] and alcohol abuse is one of the most important causes of chronic liver disease. Cirrhosis accounts for the majority of all medical deaths among alcoholics. In the past, ALD was attributed to malnutrition (hence the term *Laënnec nutritional cirrhosis*). However, alcoholic cirrhosis occurs in well-nourished persons, and cirrhosis does not occur in patients with simple malnutrition. In fact, there is some evidence that the rate of ALD has increased in Japan despite significant increases in the fat and caloric content of the diet. [2] Epidemiologic studies have shown a direct correlation between national per capita consumption of alcohol and prevalence of ALD, and between individual alcohol consumption and the risk of alcoholic hepatitis or cirrhosis. [3], [4] and [5] Historically, a dramatic decrease in ALD was associated with diminished alcohol intake during war rationing and prohibition. Both the amount of ethanol ingested and duration of heavy drinking are important factors in the induction of ALD. Alcohol intake is best quantified in terms of grams of ethanol per day, and interestingly, each common unit of drinking contains about 10 to 15 g of ethanol: one ounce or shot of distilled spirits, a 12-oz bottle of beer, or a 4-oz glass of wine. There is no difference in risk of ALD associated with use of any of these forms of beverage alcohol or with the pattern of drinking, with the exception that drinking alcohol at times other than at meals is associated with increased risk of liver injury. [6] The amount of ethanol that can be safely consumed is estimated to be up to 20 g (two drinks) per day for men and up to 10 g (one drink) for women, and in fact this amount of alcohol consumption is associated with reduced risk of myocardial infarction. [7] A daily intake of ethanol as low as 40 g in men or 20 g in women over more than 10 years results in a significant increase in the incidence of cirrhosis. The peak incidence of ALD is in the age range of 40 to 55 in men, but a decade earlier in women. Not only do women develop liver disease more rapidly, they do so with a lower daily alcohol intake, and thus lower cumulative exposure to ethanol, [8], [9], [10] and [11] and there is evidence that a larger percentage of women progress to more advanced liver disease. [12], [13] and [14] The male-to-female ratio of alcoholism is about 10:1, but for alcoholic liver injury it is about 3:1. This strongly supports the clinical impression of an increased susceptibility of women to ALD, and emphasizes the need to help women control abusive drinking at a younger age. It remains an unsolved question why only a subset of alcoholics develops serious liver injury (no more than 35% of heavy drinkers develop alcoholic hepatitis and only 20% develop cirrhosis). Indeed, even in the alcohol-fed baboon model, only a fraction of the animals developed cirrhosis. A better understanding of this phenomenon could lead to identification or modification of risk factors. Some clues have been gained from studies of the pathogenesis of this disease.

PATHOGENESIS

It may be argued that the ultimate treatment for ALD is the prevention or treatment of alcohol abuse. However, a better understanding of the hepatotoxicity of alcohol may lead to treatments of fatty liver and alcoholic hepatitis, and thus prevention or delay of occurrence of cirrhosis. It may also foster an understanding of the adverse interaction between alcohol consumption and hepatitis C virus (HCV) infection. Furthermore, insights gained from studying alcoholic liver injury may extrapolate to nonalcoholic fatty liver and steatohepatitis.

Metabolism of Ethanol

Ethanol is rapidly absorbed from the gastrointestinal tract and most of it is metabolized in the liver. When moderate amounts (e.g., 10–30 g) of ethanol are consumed by normal adult men in the fed state, a sizeable first pass metabolism of ethanol can be easily detected. That is, the increase in blood alcohol level is less when alcohol is taken orally than when the same dose is given intravenously. Whether the stomach or liver or both is responsible for this phenomenon is debated. In favor of gastric metabolism are the facts that gastric mucosa contains alcohol dehydrogenases and peak blood alcohol levels are higher in individuals with decreased gastric alcohol dehydrogenase activity (e.g., women and patients with a gastrectomy, chronic gastritis, or who are taking cimetidine). [15], [16], [17], [18], [19], [20] and [21] However, the total ethanol oxidizing capacity of the stomach is rather low, suggesting a hepatic contribution. [22], [23] It has been suggested that the observed alterations in first pass metabolism seen with the various stomach conditions reflect concomitant changes in rates of alcohol absorption and delivery to the liver. [24] First pass metabolism is most evident when low doses of alcohol are ingested with or soon after a meal.

After absorption, ethanol is distributed in the body water space, and is largely metabolized in liver to acetaldehyde by alcohol dehydrogenase (ADH) in the cytosol and the cytochrome P450IIE1 (CYP2E1) in microsomes (Fig. 114-1). ADH is responsible for the bulk of ethanol oxidation. The ADH reaction rate is controlled by the amount of ADH, the acetaldehyde level, and the free reduced nicotinamide adenine dinucleotide/nicotinamide adenine dinucleotide $^{+}$ (NADH/ NAD $^{+}$) ratio in the cytosol. [25] The amount of ADH in the liver is not induced by chronic drinking; however, with fasting, protein malnutrition and liver disease, ADH activity and the ethanol elimination rate are decreased. [26] Humans have seven ADH gene loci, two of which are polymorphic (Table 114-1). People of different racial groups inherit different sets of ADH isoenzymes. The kinetic properties of the various ADH isoenzymes differ widely in vitro, [26] but the effects of inheriting different ADH isoenzymes on

alcohol pharmacokinetics are relatively small. African Americans who carry a copy of the *ADH2 * 3* gene, encoding an enzyme with high maximum velocity, have a somewhat faster rate of alcohol metabolism. [27]

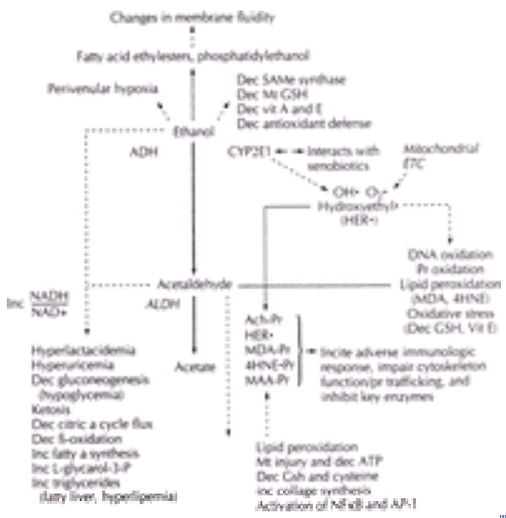

FIGURE 114-1. Pathway of alcohol metabolism and its effect on metabolism of hepatocytes. Alcohol is metabolized by alcohol dehydrogenase (ADH) or cytochrome P4502E1 (CYP2E1) to acetaldehyde. Acetaldehyde is then oxidized by aldehyde dehydrogenase (ALDH). The ADH and ALDH reactions generate free reduced nicotinamide adenine dinucleotide (NADH) more rapidly than it can be reoxidized by the mitochondria. The increased reduced nicotinamide adenine dinucleotide/nicotinamide adenine dinucleotide $^{+}$ (NADH/NAD $^{+}$) ratio modifies many intermediary metabolism pathways, indicated in the lower left, and contributes to formation of reactive O_2 species by the mitochondrial electron transfer chain (ETC). CYP2E1 contributes to the formation of reactive O_2 species and hydroxyethyl radical (HER·), which promote oxidation and peroxidation of DNA, proteins, and lipids. Acetaldehyde (*Ach*) and aldehyde products of lipid peroxidation (*4HNE,* 4-hydroxynonenal; *MDA,* malondialdehyde) participate in the formation of various aldehyde-protein adducts (*MAA,* malondialdehyde-acetaldehyde adducts; *Pr,* protein). Other effects include changes in membrane fluidity and depletion of antioxidants (including *SAMe,* S-adenosylmethionine). These effects are important mechanisms that lead to liver cell injury and cell death. Other abbreviations: *Dec,* decrease; *Fe,* iron; *Inc,* increase; *Mt,* mitochondrial. (From Lumeng L, Crabb DW. Alcoholic liver disease. Curr Opin Gastroenterol 2001;17:211, with permission.)

ALCOHOL METABOLIZING ENZYMES	INTRACELLULAR LOCATION	POLYMORPHISMS
Alcohol dehydrogenase	Cytosolic	*ADH2*1, 2*2, 2*3* *ADH3*1, 3*2*
Cytochrome P450IIE1	Endoplasmic reticulum	Promoter variants
ACETALDEHYDE METABOLIZING ENZYMES		
Aldehyde dehydrogenase 2	Mitochondrial	*ALDH2*1, ALDH2*2*
Aldehyde dehydrogenase 1	Cytosolic	None known

TABLE 114-1 Ethanol Metabolizing Enzymes in Humans

CYP2E1 is associated with reduced nicotinamide adenine dinucleotide phosphate (NADPH)-cytochrome P450 reductase in the microsomal membrane (this complex was originally designated the microsomal ethanol oxidizing system [MEOS]), and reduces molecular oxygen to water as ethanol is oxidized to acetaldehyde. [28] CYP2E1 is inducible by chronic drinking, [29] especially in the perivenular zone, [30], [31] and it may contribute to the increased rates of alcohol elimination in heavy drinkers. CYP2E1 can also be induced in fasting and diabetes and by a diet high in fat. [29] CYP2E1 has a high K_m for ethanol and may assume a greater role in ethanol metabolism at high blood levels. CYP2E1 is unusually “leaky” and generates reactive oxygen species (ROS) including hydroxyl radical (OH·), superoxide anion (O_2), hydrogen peroxide (H_2O_2), and hydroxy ethyl radical (H_2). Thus, CYP2E1 is a major source of oxidative stress. [32], [33] This P450 enzyme also has capacity to activate many xenobiotics to toxic metabolites, most notably acetaminophen. [34] In addition to CYP2E1, other P450 enzymes such as CYP1A2 and 3A4 may also play a role in ethanol oxidation. [29]

Acetaldehyde is converted by aldehyde dehydrogenases (ALDH) to acetate, which is released from the liver and metabolized by heart and muscle. [35] The most important ALDH isozyme in acetaldehyde metabolism is the mitochondrial form, designated ALDH2 (Table 114-1). ALDH2 is inhibited by disulfiram (Antabuse). Consumption of alcohol while taking disulfiram or one of a number of other drugs (metronidazole, and some sulfonylureas and cephalosporins) results in the accumulation of acetaldehyde, producing vasodilation, facial flushing, tachycardia, nausea, and vomiting. [36], [37] and [38] This reaction is also observed in about half of Asians when they drink. [39] It is the result of inheriting an ALDH2 allele (*ALDH2 *2*) that contains a point mutation at position 487. This allele acts as a dominant negative, resulting in markedly reduced activity of ALDH2 in heterozygotes. [40]

A minor pathway of alcohol metabolism is the formation of fatty acid ethyl esters. [41], [42], [43] and [44] During the hydrolysis of triglyceride or phospholipid, ethanol can substitute for water, forming the ethyl ester. This metabolite can form in tissues that do not otherwise participate in ethanol oxidation, such as heart and pancreas. It is suggested that these lipids disturb membrane function, and may thus contribute to organ dysfunction.

Nutritional Aspects of ALD

It is important to emphasize that ethanol itself, rather than a nutritional deficiency in the alcoholic patient, can cause ALD. Baboons chronically given large amounts of ethanol, despite a nutritionally complete diet, developed fatty liver and significant fibrosis. [45] About 30% of the baboons developed cirrhosis after 1 to 5 years. Rats fed by constant intragastric infusion of a nutritionally adequate liquid diet containing ethanol comprising 50% of total calories developed fatty liver, necrosis, and fibrosis. However, there are interactions between diet and alcohol in the development of liver injury. The most obvious is that severe liver injury in the rat model (in particular activation of stellate cells and fibrosis) required a high content of polyunsaturated fatty acids (PUFA) in the diet; [46] fatty liver and injury are in fact prevented by substituting saturated fat or medium chain triglycerides for the PUFA. [47] This has been suggested to be due to increased susceptibility of the PUFA to oxidative damage. It is interesting that there appears to be an inverse relationship between saturated fat and cholesterol intake, and risk of ALD. [48] Administration of ethanol in a liquid diet that is very low in carbohydrate also causes liver inflammation and severe fatty liver in rats. [49] Lastly, obesity has been found to be a risk factor for the development of cirrhosis in humans. [50]

Genetic Predisposition to Alcoholism and ALD

Twin, adoption, and high-risk familial clustering studies have all proven genetic predisposition to alcoholism (see Chapter 55). The strongest genetic associations identified to date are those between risk of alcoholism and genes encoding alcohol-metabolizing enzymes. In particular, individuals having the genes encoding high activity ADH (ß2 ADH encoded by *ADH2 *2*) or the dominant negative allele for ALDH2 found in Asians (*ALDH2 *2*) are at reduced risk of alcoholism. [51], [52] This finding is quite robust, having been replicated by a number of groups. *ADH2 *2* is very common in Asians, and it has also been identified in about 20% of Israelis. This gene locus also appears to reduce the risk of alcohol abuse in Israelis. [53] The effect of these variants can best be explained by either increased rates of formation (high activity ADH) or decreased rates of clearance of acetaldehyde (ALDH2 deficiency), which can cause aversive reactions to drinking. However to date, high circulating levels of acetaldehyde have not been documented in the patients with *ADH2 *2*. An additional polymorphism in the promoter of the *ALDH2* gene has been reported. [54], [55] The polymorphic site, either an A or G, is adjacent to an element bound by transcription factors belonging to the steroid receptor family. The A allele, which is less active in vitro, was less common in a group of Japanese alcoholics with active ALDH2. [54] This would be consistent with the notion that a less active promoter results in synthesis of less ALDH2 enzyme and reduced ability to remove acetaldehyde. This variant was found in all ethnic groups examined. [55] It will be

very interesting to see if the observations on the association of the A allele with protection from alcoholism can be extended to Caucasians and Africans.

The evidence for genetic risk factors for ALD is less strong than that for alcoholism. The largest existing study is the U.S. Veterans Administration Twin Panel Study. Analysis of the dataset in 1981 showed a substantially higher concordance for cirrhosis in monozygotic twins than in dizygotic twins, indicating the presence of a genetic component to the risk. [56] This database was reanalyzed in 1994. The analysis again supported the notion that concordance for cirrhosis was higher in the monozygotic twins, but found that most, but not all, of the genetic liability for cirrhosis was the result of shared risk for alcoholism. [57] This study is limited by lack of information on the prevalence of hepatitis C in the cohort. No specific candidate genes that confer risk or protection against ALD have been firmly established. Specifically, none of the ADH polymorphisms have been clearly identified as predisposing to the development of ALD. Polymorphisms in CYP2E1 have been sought as a clue to predisposition to ALD, since this enzyme may contribute to the oxidative stress of ethanol metabolism. An initial report suggested a significant association of the c2 allele (one that lacks the Rsa I restriction site) with ALD. This promoter variant is known to increase the rate of transcription (compared to the wild type c1 allele) when this 5'-regulatory construct was coupled to a reporter gene in an in vitro expression system. [58] Although this initial report drew much attention, most subsequent studies have refuted any relationship between the c2 allele and the risk of ALD. [59] Case-control studies have indicated the possibility of increased genetic risk for ALD through inheritance of *ALDH2 *2,* and polymorphisms of the tumor necrosis factor-a (TNFa) and interleukin-10 (IL-10) promoters, as well as glutathione S-transferase, [60], [61], [62] and [63] but these require confirmation.

Genesis and Consequences of Fatty Liver

Fatty liver has long been recognized as the earliest response of the liver to chronic alcohol consumption. This was attributed to the effect of increased levels of cytosolic and mitochondrial NADH, which inhibits oxidation of fatty acids and stimulates their synthesis (see Fig. 114-1). Increased levels of L-glycerol-3-phosphate and fatty acids, as well as induction of a number of enzymes involved in de novo fatty acid synthesis, result in accelerated synthesis of triglycerides. The liver secretes some of this fat as very-low-density lipoprotein (VLDL), which can result in hyperlipidemia. The rate of secretion of VLDL apparently does not keep pace with the increased rate of synthesis of triglycerides, so the liver cells become filled with fat. Because the high periportal to low perivenous oxygen gradient normally present within the liver acinus is made steeper by chronic alcohol ingestion, perivenous hepatocytes become much more hypoxic. [64] As a result, the effect of alcohol metabolism on the redox state and on fat accumulation is most severe in perivenous cells. Fat accumulation does not continue indefinitely because the redox-state changes are attenuated with continuing chronic alcohol use. [65]

Additional support for this model comes from cell model studies. Cell lines expressing high levels of ADH metabolize alcohol and experience a more reduced redox potential in the cytosol, as reflected by an increase in the lactate/pyruvate ratio in the medium. Coincident with this, the cells accumulate large amounts of triglyceride and free fatty acids. As in perfused liver or isolated hepatocytes, this was associated with inhibition of fatty acid oxidation and an increased rate of fatty acid synthesis. [66] Addition of tocopherol to the medium did not prevent fat accumulation, arguing that oxidative stress was not involved. On the other hand, methylene blue lowered the lactate/ pyruvate ratio and attenuated the accumulation of fat in the cells. These results argue that a high NADH/NAD $^{+}$ ratio alone is sufficient to initiate the accumulation of fat and the development of fatty liver.

In addition to these effects of ethanol, another mechanism has been suggested. The levels of fatty acids in the liver appear to be sensed by the peroxisome proliferator-activated receptor (PPARa). [67], [68] This receptor forms heterodimers with retinoid X receptor, binds consensus response elements, and activates gene transcription. Many genes involved in fatty acid binding (fatty acid binding protein), fatty acid oxidation (peroxisomal fatty acyl-coenzyme A [CoA] oxidase, mitochondrial medium chain fatty acyl-CoA dehydrogenase, microsomal lauryl hydroxylase), and lipoprotein synthesis (apo CIII and AI) appear to be regulated by PPARa. Mice in which the genes for PPARa or fatty acyl-CoA oxidase are disrupted develop steatohepatitis. [69], [70] PPARa mRNA was also reported to be decreased in the liver of rats fed ethanol chronically, [71] and certain PPARa-regulated genes (fatty acid binding protein, malic enzyme, and acyl-CoA oxidase) were repressed by alcohol feeding of mice. [72] Moreover, alcohol metabolism by hepatoma cells impairs the function of transfected PPARa. [73] Further work will be required to understand the role of this interesting receptor in the development of fatty liver.

An interesting variant of fatty liver is alcoholic foamy degeneration. Pathologically, there is microvesicular steatosis, especially prominent in the perivenous zone of the liver. Microvesicular fatty change typically suggests damage to mitochondria, and indeed, this lesion has been associated with deletions in the mitochondrial genome. [74] This may be related to the oxidative stress on the mitochondrion discussed subsequently.

Fatty liver is not as benign as once considered. Some patients with alcoholic fatty liver may develop fibrosis or cirrhosis without ever experiencing an attack of alcoholic hepatitis, as may some with nonalcoholic fatty liver. Animals with fatty liver are exquisitely sensitive to lipopolysaccharide (LPS), [75] and LPS has an important role in the pathogenesis of experimental ALD, as discussed in more detail later in the chapter. Thus, prevention or reversal of fatty liver may reduce the hepatotoxicity of ethanol.

Roles of Oxidant Stress

The redox hypothesis of alcoholic fatty liver has been challenged by studies in which antioxidants prevented development of fatty liver in animals receiving large, single doses or chronic feeding of alcohol. In addition, with chronic alcohol feeding of baboons, the redox shift in the liver becomes less severe, but the fat accumulation persists. This suggests that over time, the redox state is no longer the primary cause of alcoholic fatty liver. [65] A candidate for the perpetuation of fatty liver at this stage is oxidative stress. There are two primary ways by which chronic alcohol use can induce oxidative stress: induction of CYP2E1 and reductive pressure on the mitochondrial electron transfer system (see Fig. 114-1). In either case, increased rates of single electron transfers (at the sites of flavin coenzymes in the cytochrome catalytic cycle or the electron transfer system) facilitate the transfer of electrons to oxygen. CYP2E1 is induced by chronic alcohol use and is “leaky,” in that electrons transferred to it from CYP450 reductase can be transferred to molecular oxygen in the absence of substrate, forming superoxide. [76] The preferential expression of CYP2E1 in the central zone of the liver coincides with the prominence of alcohol-induced injury to this zone. Exposure of CYP2E1-expressing HepG2 cells to high concentrations of ethanol produced morphologic changes and DNA fragmentation, activation of caspases 1 and 3, and apoptosis. [77] These changes were not observed in control HepG2 cells, and could be prevented by an inhibitor of CYP2E1, by antioxidant compounds, or by inhibition of the caspases. Thus, oxidative stress mediated by CYP2E1 can injure cells via activation of caspases and apoptosis. The situation is less clear in vivo; CYP2E1 inhibitors attenuated liver injury in alcohol-fed rats; [78] however, mice with the CYP2E1 gene knocked out were still sensitive to ethanol hepatotoxicity. [79] It is possible that other ethanol-metabolizing CYPs are important in the CYP2E1 knockout animals.

Other groups have shown that ethanol metabolism is capable of causing acute oxidative stress (as demonstrated by dichlorofluorescein fluorescence) in perfused liver and isolated hepatocytes. [80], [81] The mitochondrion appears to be important in this phenomenon, and it is well known that chronic alcohol use causes abnormalities of mitochondrial morphology and function. Among, the most prominent antioxidant defenses in the mitochondrion is the presence of glutathione and glutathione peroxidase. It is noteworthy that mitochondrial glutathione is preferentially depleted in the alcohol-fed baboon or rat. [82] Alcohol use also adversely affects antioxidant defense in hepatocytes by decreasing the activity of S-adenosylmethionine synthase and depleting the stores of vitamins A and E. [83]

An end result of oxidative stress is lipid peroxidation, a chain reaction of lipid radical formation that follows from the removal of an electron from PUFA. The lipid free radical undergoes addition of oxygen to form a lipid hydroperoxide, and then decomposes. This leads to production of reactive aldehydes such as malondialdehyde (MDA) and 4-hydroxynonenal (4-HNE). Lipid peroxidation has been demonstrated in experimental animals exposed to alcohol. [84] In humans, ethanol use increases the formation of lipid peroxides and is associated with reduced levels of vitamin E, the chief antioxidant in membranes systems [85] and increased exhalation of pentane (a product of lipid peroxidation) has been reported in ALD. [86] However, a trial of vitamin E supplementation in alcoholic cirrhosis did not show beneficial effects on mortality or hospitalization rate. [87]

Role of Protein Adduct Formation

Protein adducts are covalent linkages formed between the amino acid side chains (typically lysine) of a number of liver proteins and a low-molecular-weight reactive chemical such as acetaldehyde, hydroxyethyl radical (HER $^{\cdot}$), and peroxidative aldehydes like 4-HNE and MDA [88] (see Fig. 114-1). Adducts are more likely to form when the level of the reactive aldehydes are increased. Acetaldehyde levels are higher in alcoholics than in social drinkers due to the adaptive increase in ethanol oxidation and reduction in the ability of mitochondria to oxidize acetaldehyde after heavy use of alcohol. [89] HER $^{\cdot}$ forms when ethanol itself accepts an electron and forms a free radical. [90] The peroxidative aldehydes form during lipid peroxidation. The modified proteins identified to date include tubulin, [91] ? [4] -3-ketosteroid

5ß-reductase (initially identified as the 37-kd protein), [92] low-density lipoproteins, [93] cytochrome C, [94] CYP2E1, [95] and various liver cell membrane proteins. In the hepatic stellate cell, Jun N-terminal kinase has been shown to form adducts with 4-HNE. [96] Undoubtedly there are other as yet undiscovered adducts. There are two important biologic consequences and one practical consequence of adduct formation. First, the adducts are recognized as neo-antigens. Antibodies directed against membrane and soluble proteins of the hepatocyte modified by acetaldehyde and by HER [-·] are detected in alcoholics and they constitute a possible immune component to liver injury. [97] Among the most provocative studies are those that showed increased sensitivity of the liver to ethanol in animals immunized with protein-acetaldehyde adducts, then fed ethanol. The immunized, alcohol-fed animals developed hepatic inflammation and fibrosis, while animals that were only immunized or only fed alcohol did not. [98] Second, the adducted protein maybe dysfunctional. For example, tubulin adducts are abnormal in terms of protein trafficking and this probably contributes to protein retention and swelling of the liver cells. A practical ramification is that the detection of adducts may ultimately be used as a test for alcohol abuse and to monitor for abstinence.

Cytoskeletal Changes in ALD

The accumulation of fat in the liver is accompanied by induction of microsomal proteins and fatty acid binding protein, and by retention of secretory proteins (due to impaired cytoskeleton function and protein trafficking pathways caused by formation of acetaldehyde-protein adducts) and water. [99] In addition, continuous intragastric infusion of ethanol caused a striking decrease in alkaline protease activity in liver as well as a decline in sodium dodecyl sulfate (SDS)-activated 20S proteasome proteases (i.e., chymotrypsin-like (ChT-L) and peptidylglutamyl peptide hydrolase activities). [100] The inhibition of proteolysis under this condition was abrogated by administration of an inhibitor of CYP2E1, suggesting that oxidative stress caused this inhibition. Western blot studies also demonstrated a marked reduction of ubiquitin and ubiquitin conjugates, and levels of ubiquitin mRNA, in rats given ethanol. [101] Ubiquitin is necessary for the targeting of cytosolic proteins for degradation in the proteasome, and deficiency of ubiquitin could contribute to the accumulation of cytosolic proteins. This causes the hepatocytes to swell and is called balloon or hydropic degeneration. The sheer size of the cells may increase portal pressure and interferes with blood flow to the perivenous zone. [102] There is also increased oxygen consumption in the liver of rats after prolonged alcohol consumption. Increased oxygen consumption coupled with decreased blood flow may induce perivenular hypoxia; this may be a signal for apoptosis or necrosis and fibrosis in the perivenous zone. Ethanol may also affect the interactions of hepatocytes with extracellular matrix. Tuma and colleagues [103] reported that alcohol-feeding preferentially impaired cellular attachment and spreading of perivenous hepatocytes but spared the periportal counterparts.

The Role of Lipopolysaccharide and the Kupffer Cell

There is a growing appreciation that portal vein lipopolysaccharide (LPS) and activation of the Kupffer cells are important in alcoholic liver injury (Fig. 114-2). Studies show that alcohol feeding increases LPS levels in the portal vein, that chronic ethanol feeding induces the expression of the LPS binding protein and its receptor present on the Kupffer cell (CD14), and that inactivation of Kupffer cells by gadolinium chloride ameliorates the injury seen with chronic ethanol administration in the Tsukamoto-French model. [104] Kupffer cells can also be activated by ethanol oxidation by CYP2E1, [105] which appears to act via the oxidative stress-sensitive nuclear transcription factor, NF-?B. This factor coordinately increases the expression of a wide range of genes, including those for cytokines and chemokines. Among these are:

- TNFa, which is a cause of hepatocyte apoptosis and oxidative stress
- transforming growth factor-ß1 (TGFß1), the most important stimulus for collagen synthesis by stellate cells
- IL-8, which attracts neutrophils to the liver (plasma IL-8 levels are markedly increased in patients with alcoholic hepatitis on admission and decreased thereafter with recovery)
- IL-1 and IL-6, which elicit an acute phase response
- eicosanoids, which are implicated in the increased rate of hepatocyte oxygen consumption during ethanol metabolism
- chemokines that stimulate migration of leukocytes into the liver
- platelet-derived growth factor (PDGF), a major stimulator of hepatic stellate cell (HSC) proliferation [106] (discussed later in the chapter).

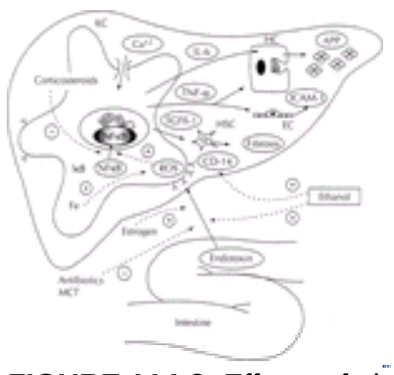

FIGURE 114-2. Effects of alcohol ingestion on endotoxin levels, the activation of Kupffer cells (KC). Ethanol consumption increases the absorption of lipopolysaccharide (endotoxin) from the gut, and induces expression of CD14, the endotoxin receptor, on KC. This action is more pronounced in females, due to effects of estrogen. Antibiotics and medium chain triglycerides (*MCT*) may reduce endotoxin absorption and action. Binding of endotoxin to the CD14 molecule leads to activation of nuclear factor ?B (NF?B), which in turn activates numerous inflammation-related genes, including those for tumor necrosis factor-a (*TNF*a), interleukin 6 (*IL-6*), and transforming growth factor-ß (*TGFß-1*). These in turn have effects on the hepatocytes (HC), such as synthesis of acute phase proteins (*APP*) and generation of reactive oxygen species (*ROS*), endothelial cells (*EC*), such as expression of intercellular adhesion molecule-1 (*ICAM-1*), and hepatic stellate cells (*HSC*), leading to proliferation and collagen synthesis. Formation of ROS may independently activate NF?B. KC contain calcium channels that are involved in activation; calcium channel blockers reduce the activation of these cells in experimental models. Other abbreviations: *I?B,* inhibitor of NF?B *Fe,* iron. (From Lumeng L, Crabb DW. Alcoholic liver disease. Curr Opin Gastroenterol 2001;17:211, used with permission.)

TNFa appears to play a particularly important role in ALD. Knock-out mice, which lack the TNF type 1 receptor, [107] and rats given antibody against TNFa [108] are protected against liver injury in the Tsukamoto-French model. This cytokine may contribute to liver injury by stimulating the formation of superoxide in the mitochondria, [109] particularly when mitochondrial glutathione is depleted. It is also interesting to note that the mere presence of fatty liver (even without ethanol consumption) increases the sensitivity of the liver to the effects of LPS. [75] Two reviews by Diehl [110] and McClain and colleagues [111] summarized the role of cytokines in the pathogenesis of ALD.

The relationship between pathological liver injury, endotoxemia, diet, lipid peroxidation, and NF?B activation and imbalance of the expression of pro- and antiinflammatory cytokines has been examined in rats. [112] The animals were given ethanol in a diet containing medium-chain triglycerides, palm oil, corn oil, or fish oil by chronic intragastric infusion. Electrophoretic mobility shift assay was used to evaluate NF?B activation and mRNA assays were performed for proinflammatory cytokines (TNFa, IL-1ß, interferon-? [IFN-?], and IL-12), C-C chemokines (MCP-1, MIP-1, and RANTES), C-X-C chemokines (CINC-1, MIP-2, IL-10, and ENA-78) and antiinflammatory cytokines (IL-10, IL-4, and IL-13). The greatest necroinflammatory injury was observed in rats fed fish oil or corn oil plus ethanol. These animals had activation of NF?B, increased expression of proinflammatory C-C and C-X-C chemokines, and also the highest levels of LPS and lipid peroxidation. IL-10 and IL-4 mRNA levels were decreased in these two groups of alcohol-fed rats that exhibited the most extensive necroinflammation.

Pathways of Hepatic Stellate Cell Activation

The final element in alcoholic liver injury is activation of the hepatic stellate cell (HSC) and production of collagen. [106], [113] This fibrogenic process requires the interaction of Kupffer cell-derived cytokines with the HSC (Fig. 114-2 and Fig. 114-3). An area of great interest is the control of the earliest stage of HSC activation. One of the very earliest events is the activation of NF-?B in the HSC, but the signal responsible for this is as yet unknown. [114] This might relate to changes in the matrix which these cells contact in the liver (e.g., by modification by acetaldehyde), formation of protein adducts in the HSC, or oxidative stress originating in the hepatocytes or within the stellate cells themselves (HSC are known to express ADH and ALDH, but not CYP2E1). [115], [116] Acetaldehyde, MDA, and 4-HNE can also

induce stellate cells to express collagen genes. [117], [118] The ? isoform of PPAR may participate in the control of HSC differentiation. Expression of this receptor falls during HSC activation, and treatment of the cells with PPAR? ligands retards the activation of the cells. [119]

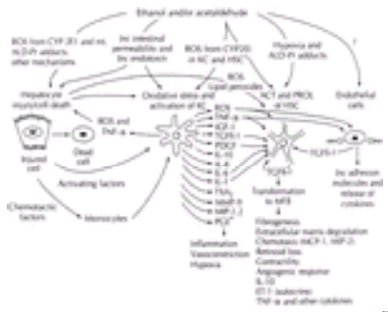

FIGURE 114-3. Effects of hepatocyte injury, endotoxin, and ethanol- or acetaldehyde-induced changes (including hypoxia and oxidative stress) on autocrine and paracrine pathways of activated Kupffer cells (*KCs*), hepatic stellate cells (*HSCs*), and endothelial cells. Although not shown, other cell types in liver (e.g., bile duct epithelium, lymphocytes, and platelets) are also involved. Fibrogenesis includes synthesis of matrix proteins (e.g., type 1 and 3 collagens, proteoglycans, hyaluronate, and several specialized glycoproteins including fibronectin), release of metalloproteinases and secretion of tissue inhibitors of metalloproteinases (*TIMPs*). Abbreviations: *ALD-Pr,* aldehyde-protein; *ROS,* reactive oxygen species; *TNFa,* tumor necrosis factor-alpha; *IGF-1,* insulin growth factor-1; *TGFß-1,* transforming growth factor beta-1; *PDGF,* platelet-derived growth factor; *IL,* interleukin; *TXA* $_2$, thromboxane A $_2$; *MMP,* matrix metalloproteinase; *MIP,* macrophage inflammatory protein; *MCP,* monocyte chemotactic protein; *ET,* endothelin; *PGE,* prostaglandin E; *MFB,* myofibroblast. *ACT,* activation; *PROL,* proliferation; *mt,* mitochondria; *Inc,* increased. (From Lumeng L, Crabb DW. Alcoholic liver disease. Curr Opin Gastroenterol 2001;17:211, with permission.)

Once NF-?B is activated, the stellate cells express receptors for and can be stimulated to divide by PDGF. They also can be stimulated to secrete collagen by TGFß1 and possibly IL-1. [106] With time, this activation may become irreversible, leading to progressive fibrosis and cirrhosis. The irreversible activation may be driven by the changes the cells make in the extracellular matrix, such as deposition of type I collagen, production of TGFß1 by the HSC themselves (resulting in a positive feedback loop), and depletion of retinyl ester stores in the HSC. In addition, the HSC also make intercellular adhesion molecule-1 (ICAM-1), macrophage inflammatory protein-2 (MIP-2), platelet-activating factor (PAF), stem cell factor (SCF-1), and monocyte chemotactic factor (MCP-2), chemokines and adhesion molecules that may be important in the migration of leukocytes into the liver. [106] All of these factors may lead to progressive liver damage, even if the patient stops drinking.

Gender Differences in the Response to Alcohol

Given the new knowledge about the pathogenesis of ALD, it is worth revisiting the differences in responses to alcohol between men and women. [120] Studies using volume estimates by computed tomography (CT) scan have shown that men and women have livers that are similar in size. This results in a larger liver mass/body mass ratio for women. When the rate of alcohol metabolism is normalized to liver mass, men and women have similar metabolic rates. [121] However, blood alcohol levels after comparable doses of alcohol will usually be higher in women than in men because of the women's smaller body mass and lean body mass, resulting in a lower volume of distribution in women. It has also been found that female rats are more susceptible to alcohol-induced liver injury in the Tsukamoto-French model. [122] This is likely due to more pronounced accumulation of fat in their livers, lower levels of fatty acid binding protein (and thus higher concentrations of unbound free fatty acids), high levels of plasma LPS, more striking expression of the LPS receptor (CD14) in Kupffer cells and LPS binding protein, [123] more pronounced central hypoxia, and more marked activation of NF-?B in the HSC. [124] Female rats fed ethanol showed a significantly higher level of CINC-1 mRNA (the rat equivalent of IL-8, the major neutrophil chemokine) in the liver than male counterparts in response to LPS. Oophorectomy abolished the gender difference in CINC-1 mRNA expression. Estrogens also modulate the oxidative stress of ethanol metabolism. [125] Thus, many mechanisms may conspire to make women alcoholics more prone to develop alcoholic hepatitis and cirrhosis.

Apoptosis in ALD

Cell death in ALD can occur by either apoptosis or necrosis. Traditionally, these processes are distinguished by the lack of inflammation in the former. However, recent concepts dispute this distinction (i.e., massive cell death via apoptosis can overwhelm phagocytic clearance and can incite inflammation including chemotaxis for neutrophils). While there is no doubt that hepatocyte necrosis takes place due to alcohol-induced liver injury, data indicate that apoptosis is also important. Because apoptosis is a rapid process (varying from minutes to a few hours), hematoxylin and eosin staining for apoptotic bodies underestimates the extent of apoptosis; thus, the TUNEL assay and immunohistological methods are needed to evaluate apoptosis. With these latter methods, the apoptotic index was found to correlate with Maddrey discriminant function [126] and histological severity. [127]

There are two major pathways in apoptosis that ultimately activate effector caspases and cell death. One involves the death ligands and death receptors (e.g., ligand-receptor binding involving TNFa and FAS-ligand) and the other involves opening of mitochondrial membrane transition pores and release of cytochrome C. [128] The release of TNFa from Kupffer cells has been mentioned; ROS are also able to induce apoptosis. Apoptosis in ALD has important consequences. Activation of Fas induces IL-8, probably a key factor in the neutrophilic infiltration of the liver in alcoholic hepatitis. Moreover, apoptosis, followed by phagocytosis of the dead cell, can present intracellular neoantigens, such as aldehyde-protein adducts, to the immune system, with the formation of cytotoxic T cells directed toward these epitopes. Thus, it may be possible to formulate therapeutic modalities that modulate the toxicity of ethanol by altering either the pro-apoptotic signals or the anti-apoptotic defenses of the liver cells.

CLINICAL MANIFESTATIONS

The spectrum of ALD ranges from asymptomatic hepatomegaly to hepatocellular failure from alcoholic hepatitis or end-stage cirrhosis. Increasingly, patients are referred for evaluation of abnormal laboratory tests or radiographs. For example, macrocytosis is common in alcoholics and its presence in an otherwise healthy person suggests occult alcohol abuse. [129] This may be due to either thick macrocytes caused by folate deficiency or thin, target cell macrocytes due to a toxic effect of alcohol on the bone marrow, or changes in the lipid composition of the red cell membrane in cirrhosis or cholestasis. These thin target macrocytes disappear very slowly with improvement in liver function. On the other hand, spur cell anemia is seen in patients with severe ALD. The red cells of these patients have increased free cholesterol, form multiple irregular projections, and are cleared from the circulation by the spleen. Thus, the patient presents with hemolytic anemia. [130] It carries a poor prognosis. Abdominal ultrasound examinations may detect unsuspected alcoholic fatty liver exhibiting increased echogenicity.

Clinical diagnosis of the three major stages of alcoholic liver injury is difficult in that these stages may occur in any combination in the same patient, emphasizing the importance of liver biopsy. For purposes of instruction, it is useful to divide ALD into its three major types: alcoholic fatty liver, alcoholic hepatitis, and cirrhosis, and discuss clinical, laboratory, and pathological features of each. A number of less common variants are listed in Table 114-2 and discussed within the following sections.

1. Alcoholic fatty liver with perivenular fibrosis
2. Alcoholic foamy degeneration (alcoholic microvesicular steatosis)
3. Sclerosing hyaline necrosis
4. Alcoholic cirrhosis with chronic hepatitis*
5. Cholestasis
6. Hepatic iron overload
7. Lipogranuloma
8. Massive hepatic necrosis (due to acetaminophen ingestion)
9. Focal fatty liver or focal fat sparing

*A significant portion of these patients likely have concomitant hepatitis B or C infection.

TABLE 114-2 Less Common Pathological Features of Alcoholic Liver Disease

Alcoholic Fatty Liver

Consumption of ethanol in sufficient quantities (in the range of 120–150 g/d for 2 to 3 weeks) will result in fatty liver. Abstinence for 4 weeks leads to resolution. [131], [132] The development of fatty liver occurs despite consumption of an otherwise adequate diet. Stigmata of alcoholism (e.g., Dupuytren contractures, testicular atrophy, loss of the male pattern of body hair, palmar erythema, spider angiomata, and gynecomastia) may also be present. Evidence of hypogonadism and feminization are evident in alcoholic males before the onset of liver disease. It results from toxic effects of ethanol on Leydig cells leading to decreased testosterone production; impairment of hypothalamic-pituitary function resulting in low luteinizing hormone levels; induction of aromatase in adipose tissue and increased conversion of androgens to estrogens; and ingestion of nonsteroidal estrogens derived from plants (called phytoestrogens) in alcoholic beverages which then can lead to increased estrogenic effect. [133], [134] and [135]

Some cases of biopsy-proven alcoholic fatty liver are associated with constitutional symptoms of weakness, cachexia, fever, anorexia, nausea, vomiting, jaundice, hepatic tenderness, splenomegaly, or ascites. Most likely, such cases represent a combination of fatty liver and alcoholic hepatitis, since the classic lesions of alcoholic hepatitis are difficult to find in livers with panlobular fat, or may be missed because of sampling error. Another clinical variant is alcoholic foamy degeneration. [136] It has been noted in about 2% of a series of over 300 patients in Spain. [137] These patients typically present with hepatomegaly, an increased international normalized ratio (INR), jaundice, and often hyperlipemia. Encephalopathy and leukocytosis are uncommon. A transient sharp increase in transaminases followed by more prolonged evidence of cholestasis is described. This may be the explanation for cases of apparent alcoholic fatty liver that is complicated by cholestasis and hepatic failure. [138]

Laboratory abnormalities are generally mild. Bilirubin is elevated in about one fourth of cases but is usually less than 5 mg/dL. As in all stages of ALD, serum aspartate aminotransferase (AST) and alanine aminotransferase (ALT) are usually less than 300 IU/L (with the exception of foamy degeneration). The AST is usually higher than serum ALT (with the AST:ALT ratio =2 in 80% of the cases) in ALD, while the ALT is usually higher than AST in viral and toxic hepatitis and nonalcoholic steatohepatitis (NASH). Alkaline phosphatase (AP) is usually no more than 300 IU/dL. The ratio of ?-glutamyltranspeptidase (GGT) to AP activity is at times greater than 5, as a result of disproportionate induction of GGT by alcohol abuse per se. This ratio is more specific than sensitive provided that the patient is not on drugs that also induce GGT (e.g., anticonvulsants). In alcoholic fatty liver, serum albumin and globulin levels are expected to be normal in the absence of coincidental medical conditions, including malnutrition.

The fatty liver is enlarged, firm, and may be pale yellow. Microscopically, there is great variation in the number of cells affected, with a tendency for fat accumulation in the perivenous and midzones of the liver lobule. Occasionally, hepatocytes rupture to form a fatty cyst, and then a lipogranuloma. Generally, there is sparse cell necrosis or inflammation. Intrahepatic cholestasis and mild cholangiolitis may be seen in the absence of extrahepatic biliary obstruction. [139] Perivenular fibrosis may be present and has been shown to be a precursor of cirrhosis. Alcoholic foamy degeneration is characterized by perivenous hepatocytes filled with foamy, microvesicular fat, and focal cell necrosis, but no inflammation. [136], [137] There are yet other variants of fatty liver diagnosed radiographically as focal fatty liver and focal fat sparing. As these names imply, the former is due to focal accumulation of fat while the rest of the liver is not steatotic. The latter indicates the reverse. These lesions may be confused with mass lesions on ultrasound or abdominal CT scan. In the past, these conditions could be diagnosed by liver spleen scan. [140], [141] Magnetic resonance imaging (MRI) will provide definitive diagnosis of these conditions and will distinguish them from mass lesions.

Alcoholic Hepatitis

Patients with alcoholic hepatitis are usually symptomatic with anorexia, nausea, malaise, weakness, abdominal pain, icterus, weight loss, and fever. Most also show obvious evidence of protein-calorie malnutrition. [142], [143] The most severe cases are those with deep jaundice, ascites (often infected), azotemia, and hepatic encephalopathy, and may progress rapidly to death. Once set into motion, alcoholic hepatitis seems to progress despite stopping drinking. The importance of LPS in animal models of alcoholic liver injury suggests that infection, with endotoxinemia, may precipitate an attack of alcoholic hepatitis, and thus infection should be sought and treated in patients presenting with alcoholic hepatitis. Physical examination reveals the findings summarized in Table 114-3. Laboratory abnormalities are more severe in alcoholic hepatitis than in alcoholic fatty liver. [144] Anemia occurs in 50% to 70% of patients, secondary to toxic effects of ethanol on the bone marrow, impaired pyridoxine metabolism, folic acid deficiency, iron deficiency from blood loss, or hypersplenism. Leukocytosis is observed in many, and may assume leukemoid proportions, probably driven by increased levels of IL-8. Leukopenia and thrombocytopenia are present in 10% to 15% and may reflect folate deficiency, alcohol-induced marrow suppression, or hypersplenism. Serum AST and ALT are elevated in all cases of alcoholic hepatitis and AP is elevated in 80% of cases. If AST levels exceed 300 IU/L, sclerosing hyaline necrosis (discussed subsequently), coincidental viral hepatitis, or acetaminophen overdose should be suspected. An elevation of the AST:ALT ratio greater than 2 is characteristic of, but certainly not diagnostic of, alcoholic liver injury. [145], [146] Bilirubin is usually increased, while albumin is usually decreased and the prothrombin time (PT) is often prolonged. Electrolyte abnormalities are frequent in alcoholic hepatitis, and include hyponatremia, hyperchloremia due to renal tubular acidosis, hypokalemia from poor dietary intake, vomiting or diarrhea, hypomagnesemia from increased urinary loss, and respiratory alkalosis.

FINDING	FREQUENCY (%)
Hepatomegaly	95
Hepatic tenderness	50–70
Signs of portal hypertension (splenomegaly, prominent abdominal veins and ascites)	40–70
Stigmata of chronic parenchymal liver disease and alcoholism (bruising, leukonychia, palmar erythema, spider angiomata, edema, parotid gland enlargement, gynecomastia, and testicular atrophy)	30–60
Jaundice	55
Fever	50
Upper gastrointestinal bleeding	30
Hepatic encephalopathy	20

TABLE 114-3 Frequency of Clinical Findings in Alcoholic Hepatitis

In alcoholic hepatitis there is ballooning degeneration, focal hepatocyte necrosis, and a neutrophilic inflammatory infiltrate. The latter two are considered absolute criteria for the diagnosis of alcoholic hepatitis. More than 30% of patients develop alcoholic hyaline (i.e., Mallory bodies, aggregates of perinuclear, eosinophilic, amorphous material that are composed of intermediate filaments). Although highly suggestive of ALD, hyaline is also seen in many other liver diseases (Table 114-4). Other pathological findings in alcoholic hepatitis include steatosis and fibrosis or cirrhosis. [139], [147] On a case-by-case basis, it is difficult to distinguish alcoholic hepatitis from NASH on biopsy; however, periportal fibrosis, bile duct proliferation, and cirrhosis are more frequently seen in alcoholic steatohepatitis. Pericellular and perisinusoidal fibrosis are seen in both alcoholic steatohepatitis and NASH and give rise to a characteristic “chicken-wire” appearance, and perivenular sclerosis leads to deposition of a collar of collagen in the wall of the central vein. [148], [149], [150] and [151] Clinically, patients with alcoholic hepatitis are much sicker than those with NASH. In a few cases of alcoholic hepatitis, there is sclerosing hyaline necrosis (i.e., severe, irregular, confluent, centrilobular necrosis with obliteration of the central vein by collagen). [151] Some type of venoocclusive lesions are commonly seen in patients with alcoholic hepatitis and alcoholic liver cirrhosis. [152] Ninety-three percent of 15 patients with severe alcoholic hepatitis and 87% of 15 patients with alcoholic cirrhosis exhibited mild focal stenosis to severe stenosis or obstruction. Severe venoocclusive lesions were associated with recalcitrant ascites and variceal bleeding, and portended a high mortality.

Alcoholic cirrhosis (especially with continued drinking)*
Steatohepatitis
Alcoholic steatohepatitis*
Nonalcoholic steatohepatitis*
Chronic hepatitis C
Parenteral nutrition-induced liver disease
Drug-induced liver disease (e.g., griseofulvin, nifedipine, nicardipine, didanosine, diethylstilbestrol, tamoxifen, tetracycline, valproic acid, vitamin A, diltiazem, glucocorticoids, methotrexate, or amiodarone)
Chronic cholestatic disorders
Primary biliary cirrhosis*
Primary sclerosing cholangitis
Chronic biliary obstruction
Extrahepatic biliary atresia
Copper overload conditions
Wilson disease*
Indian childhood cirrhosis*
Congenital hepatic fibrosis
Copper-associated liver disease of childhood
Hepatocellular neoplasms/masses
Hepatocellular carcinoma
Hepatic adenoma
Dysplastic hepatocellular nodule
Focal nodular hyperplasia
Adenomatous hyperplasia
Weber-Christian disease
Inherited diseases of metabolism:
Abetalipoproteinemia
Alpha$_1$-antitrypsin deficiency
Glycogen storage disease, type IV*
Bloom syndrome
Postinfantile giant cell hepatitis
Juvenile Parkinson's disease

*Indicates that Mallory bodies are found occasionally to commonly; in the other conditions, they are rare. For more information, see ref. 221.

TABLE 114-4 Differential Diagnosis of Liver Diseases Exhibiting Mallory's Hyaline

Alcoholic Cirrhosis

Alcoholic cirrhosis may be asymptomatic in 10% to 20% of patients but commonly presents with the complications of chronic liver disease and the usual stigmata. The stigmata related to hypogonadism and feminization, such as spider nevi, are most prominent in patients with ALD and hereditary hemochromatosis. Complications of alcoholic cirrhosis include cachexia, coagulopathy, anasarca, spontaneous bacterial peritonitis, hepatorenal syndrome, hepatic encephalopathy, hepatocellular carcinoma, and gastrointestinal bleeding from esophageal and gastric varices. Other less frequent causes of bleeding are ectopic varices and portal hypertensive gastropathy.

The liver test abnormalities in alcoholic cirrhosis are less pronounced than those in alcoholic hepatitis. In fact, in compensated cirrhosis, many of the liver tests are nearly normal. Laboratory abnormalities may include mild increases in AST, ALT, and AP, depression of serum albumin, polyclonal elevation of serum globulins (greater than 4 g/dL), prolongation of PT, leukopenia, thrombocytopenia, and anemia. In fact, thrombocytopenia is an excellent clue to the presence of large varices (and therefore cirrhosis) in patients evaluated for abnormal liver tests. [153] Hypersplenism by itself rarely leads to platelet counts lower than 40,000/µL and splenectomy or splenic embolization is almost never indicated.

The pathological end-stage of ALD is of course cirrhosis, characterized by fibrous bands connecting portal triads with central veins, and by regenerative nodules. The nodules are typically small (1–3 mm) and uniform in size. [147] Although fibrosis may be reversible, regression is impossible when significant nodular regeneration occurs. Ethanol is known to suppress hepatic regeneration; [154] thus, with prolonged abstinence, nodular regeneration becomes more vigorous and may result in mixed micro/macronodular or macronodular cirrhosis. Superimposed fatty change and alcoholic hepatitis are often seen with cirrhosis. There may also be increased stainable iron in biopsy samples. Iron overload in alcoholic cirrhosis can be differentiated from hereditary hemochromatosis by genetic analysis for *HFE* mutations or by quantitative iron analysis. In hereditary hemochromatosis, the hepatic iron index (hepatic iron content expressed as µmol/g dry weight divided by the patient's age) in the cirrhotic liver is more than 1.9 while that in the cirrhotic liver of alcoholics is less than 1.9. In some patients with alcoholic cirrhosis, there is portal inflammation with piecemeal necrosis. This feature may reflect an autoimmune component of ALD, unsuspected Wilson disease (particularly in young patients), or most commonly, coexistent chronic viral hepatitis (particularly HCV infection).

DIFFERENTIAL DIAGNOSIS

There are numerous pitfalls in the diagnosis of ALD, among them failing to consider ALD in patients not fitting the stereotype of the skid-row alcoholic (such individuals account for only a minority of alcoholics), and assuming that abnormal liver tests in an alcoholic patient are due to ALD. It should be noted that more than 50% of patients with alcohol abuse develop liver disease because of HCV or HBV infection. Alcoholism should be considered in any patient with liver disease, and a careful drinking history should be obtained from the patient and a reliable third party such as a family member. The history should be augmented by the simple CAGE questionnaire. [155], [156] The CAGE test employs the following questions: Have you felt the need to *Cut down* on drinking? Are you *Annoyed* by references to your drinking? Do you feel *Guilty* about your drinking? Do you ever need an *Eye-opener* (i.e., do you drink in the morning to relieve withdrawal symptoms)? The phrase "eye-opener" is no longer widely used, and the concept usually needs to be explained to the patient. A positive response to two or more of these questions, however, strongly suggests alcohol abuse and should prompt a more thorough history investigating consequences of alcohol use. These include problems with the law, loss of jobs, family disruption, and injury while drinking, as well as the better known medical complications such as withdrawal syndromes. The central feature of alcoholism is that drinking takes precedence over other activities in the patient's life. The sensitivity of the CAGE test for alcohol abuse is 70% to 96%, the specificity is 91% to 99%, and the positive predictive value is about 80%. The history should also include an estimate of the grams of ethanol consumed daily and the duration of drinking.

A liver biopsy is necessary to differentiate fatty liver, alcoholic hepatitis, and cirrhosis reliably and to distinguish ALD from other causes of liver disease. Since more than 50% of patients with presumed ALD are infected with HCV or HBV, these infections must be excluded in any alcoholic. Prior to the discovery of HCV, it was reported that up to 20% of liver biopsies done on alcoholic patients showed nonalcoholic (and sometimes treatable) liver diseases. This percentage is likely lower now, but the biopsy continues to provide information about the presence of fibrosis and cirrhosis that cannot otherwise be obtained. Appropriate blood tests and biopsy should be performed to exclude nonalcoholic causes of liver disease and to define the severity of disease. However, tense ascites, severe thrombocytopenia (<80,000/µL) or prolonged PT (INR >1.4), may preclude percutaneous needle biopsy. A safer but more expensive alternative is a transjugular liver biopsy.

Alcoholic Fatty Liver

As outlined in Table 114-5, fatty liver can result from many processes, and of course the patient may have more than one cause. When one encounters massive hepatomegaly (the liver span exceeds 13 cm in the midclavicular line), the differential diagnosis includes fatty liver, right heart failure, constrictive pericarditis, Budd-Chiari syndrome, infiltrative processes (e.g., amyloidosis, myeloproliferative disorders, reticuloendotheliosis, and lipid storage diseases), and neoplasms. In evaluating the cause of hepatomegaly, a helical CT scan with dual contrast and arterial/venous phases should be performed to exclude neoplasms, as this is more sensitive in detecting focal hepatic masses than either CT scan done only in the venous phase, ultrasound, or sulfur colloid scan.

Macrovesicular fat
Alcoholic steatohepatitis
Nonalcoholic steatohepatitis
Hepatitis C
Toxins and drugs: methotrexate, halogenated hydrocarbons, niacin, HIV protease inhibitors, glucocorticoids, heavy alcohol use
Nutritional disorders: obesity, diabetes, choline deficiency, systemic carnitine deficiency, celiac disease, kwashiorkor, parenteral nutrition
Inherited metabolic disorders:
Lipodystrophy
Wilson disease
Abetalipoproteinemia
Inflammatory bowel disease
Weber-Christian disease
Q fever
Microvesicular fat*
Reye syndrome
Parenteral alimentation
Yellow fever
Heat stroke
Alpers disease
Toxins and drugs: valproic acid, intravenous tetracycline, toxic shock syndromes, salicylate overdosage in children, Jamaican vomiting disease, FIAU, nucleoside reverse transcriptase inhibitors, mycotoxins
Metabolic diseases: cholesterol ester storage disease, galactosemia, Wolman disease, long chain 3-hydroxyacyl-CoA dehydrogenase deficiency, medium chain acyl-CoA deficiency
Complications of pregnancy: acute fatty liver of pregnancy, eclampsia, HELLP syndrome

CoA, coenzyme A; FIAU, fialuridine; HELLP, hemolysis, elevated liver enzymes, and low platelet syndrome; HIV, human immunodeficiency virus.

*Microvesicular fatty liver is usually the result of mitochondrial damage and defects in lipid oxidation pathways; however, there is significant overlap with many disorders occasionally producing microvesicular fatty liver. See ref. 221 for a more detailed discussion.

TABLE 114-5 Differential Diagnosis of Fatty Liver

Alcoholic Hepatitis

The distinctive histological appearance of alcoholic hepatitis usually establishes the diagnosis. However, NASH can mimic it. Clinically, patients with alcoholic steatohepatitis are usually much sicker than those with NASH. In one study, [147] patients with NASH had a mean AST:ALT of 0.9 (range 0.3–2.8) and patients with alcoholic hepatitis had a mean ratio of 2.6 (range 1.1–11.2). Thus, the AST:ALT ratio is a useful but not foolproof parameter to distinguish NASH from ALD. Since fever, leukocytosis, right upper quadrant pain, jaundice, and cholestasis can occur in alcoholic hepatitis, and patients with cirrhosis have an increased risk of cholelithiasis, imaging of the liver and biliary tree by ultrasound or CT may be necessary to exclude extrahepatic cholestasis. These noninvasive tests are more specific (85%–100%) than sensitive (50%–95%) in detecting extrahepatic biliary obstruction. Therefore, magnetic resonance cholangiography (which depends on a strong T2 signal from stationary fluid and thus cannot be done in the presence of ascites), endoscopic retrograde cholangiopancreatography (ERCP), or percutaneous transhepatic cholangiography (PTC) may be required to exclude obstruction.

Alcoholic Cirrhosis

Alcoholic cirrhosis is usually micronodular but it can be macronodular. Thus, the differential diagnosis of alcoholic cirrhosis includes posthepatitic cirrhosis (whether due to chronic viral infection, drugs, metabolic disorders, or autoimmune hepatitis), the cirrhotic stage of primary or secondary biliary cirrhosis, Wilson disease, hemochromatosis, and cirrhosis caused by NASH. As with NASH, the degree of fatty infiltration of the liver may decrease as cirrhosis develops. These diagnoses require a careful history of drug use (including herbs and over the counter medications) and exposure to hepatitis viruses. They also require tests for hepatitis B surface antigen, hepatitis C antibody, autoantibodies (antinuclear, anti-DNA, antimitochondrial, and anti-smooth muscle antibodies), serum ceruloplasmin, a1-antitrypsin level, copper and iron studies, and the measurement of the metal content of the liver biopsy. Alcoholic cirrhosis with features of chronic active hepatitis may reflect superimposed hepatitis B or C or an autoimmune process incited by alcoholic injury. Disease processes that mimic cirrhosis (e.g., constrictive pericarditis, Budd-Chiari syndrome, venoocclusive disease, idiopathic portal hypertension, portal vein thrombosis, and myeloid metaplasia) should be considered.

Special Considerations

HCV and ALD Soon after the introduction of serologic tests for HCV antibodies, it became apparent that HCV infection was common among alcoholics with liver disease (which would previously have been considered “pure” ALD). The prevalence of HCV markers in alcoholic patients without liver disease ranges from 0 to 14%; in alcoholic patients as a whole (with and without liver disease) the prevalence ranges from 11% to 35%; and in alcoholic patients with chronic liver disease the prevalence ranges from 18% to as high as 51%. [157], [158] This increased prevalence may reflect an increased likelihood of experimenting with injection drugs among alcoholics, but other factors cannot be excluded. The liver injury caused by alcohol plus HCV is more than additive—likely it is synergistic—in promoting the progression of fibrosis and the incidence of hepatocellular carcinoma. [159] Alcohol abuse may increase HCV viral load, [159] but this is controversial. [160] Alcohol use is believed to lead to a significant decrease in response of HCV to interferon therapy. [161] The mechanism for this interaction remains poorly understood but has been attributed to ethanol’s effect on hepatic iron content, hepatic regeneration, impairment of the immune system, and activation of stellate cells. It has been reported that alcoholic patients exhibited significantly greater quasispecies complexity than nonalcoholic controls. It has been suggested that the higher quasispecies complexity in alcoholic patients explains the poor response to interferon therapy. [162]

Hepatic Iron Stores and ALD Excessive deposition of iron in the liver is seen in at least one third of alcoholic patients. The mechanisms involved in this phenomenon remain largely unknown. [163] It is widely believed that iron overload can lead to hepatic damage in ALD and hereditary hemochromatosis by facilitating generation of reactive oxygen species, as ferrous iron can transfer electrons to oxygen, or catalyze the formation of hydroxyl radical from hydrogen peroxide. As a reflection of this, protein adducts with MDA or 4-HNE were much more prominent in ALD and hemochromatosis than in other forms of liver disease. [164] The protein-adducts were found predominantly in zone 3 in ALD and in zone 1 in patients with hemochromatosis. The role of HFE (the product of the gene that is mutated in hemochromatosis) mutations in ALD with iron overload has been investigated by groups in Japan and the United Kingdom. In Japanese, in whom the HFE C282Y and H63D mutations are uncommon, alcoholic patients still develop iron overload, and those with iron overload do not carry the mutations. [165] The British study [166] included 257 patients with ALD and 117 matched healthy volunteers. About 15% of fibrotic/cirrhotic patients were C282Y heterozygotes compared with approximately 14% of controls. Of the patients without the C282Y mutation, 15% carried the H63D mutation as compared with 17% of the controls. Thus, the cause of iron overload in ALD is unrelated to the mutations causing hemochromatosis.

COURSE AND COMPLICATIONS

Alcoholic Fatty Liver

Alcoholic fatty liver is thought to be benign unless accompanied by perivenular fibrosis or foamy degeneration. Patients with these conditions have a high risk of developing hepatic failure or cirrhosis. However, in animals, fatty livers are highly susceptible to toxicity from LPS, and it is well known that donor livers that are fatty may fail when transplanted; thus, fatty liver may be less benign than previously thought. Furthermore, obesity is a risk factor for the development of cirrhosis. Treatment includes abstinence from alcohol and correction of nutritional deficits. Bed rest has no proven value. Under this regimen, fatty liver should be expected to regress in 3 to 6 weeks.

Alcoholic Hepatitis

The early mortality rates for alcoholic hepatitis vary from 19% to 78% (mean 49%) within 2 months. In the subset of patients with severe disease and spontaneous hepatic encephalopathy, the early mortality rate was higher (59%). Not surprisingly, the worst prognosis has been observed in patients with severe jaundice, encephalopathy, renal failure, ascites, and variceal bleeding. [167], [168] Several laboratory tests are prognostic indicators: PT of more than 4 seconds above control despite vitamin K treatment, total bilirubin greater than 5 mg/dL, and serum creatinine that increases more than 0.6 mg/dL during the first 10 days of hospitalization. The severity of fibrosis or cirrhosis found on liver biopsy is also a prognostic indicator (i.e., with little fibrosis, the 5-year survival was 72% but with severe fibrosis, it was less than 50%). However, many patients are too ill for conventional liver biopsy. Patients with alcoholic hepatitis who continue to drink have a very high risk of persistent hepatitis or progression to cirrhosis, and a 5-year survival of only 50%. Among those able to stop drinking, 70% reverted to normal, and the remainder either had persistent alcoholic hepatitis or cirrhosis. As expected, their mortality rate decreased to 24%. Unfortunately, it appears that women are less likely to have a complete recovery from an attack of alcoholic hepatitis. [13]

For the planning of therapy, it is of value to stratify patients with alcoholic hepatitis according to severity and prognosis. Several scoring systems have been evaluated, including the Child-Turcot-Pugh system, the combined clinical laboratory index of the University of Toronto, [167] and Maddrey discriminant function, [169] for their ability to predict 30-day mortality of patients with alcoholic hepatitis. The simple Maddrey discriminant function (4.6 × [PT (sec) - control] + bilirubin (mg/dL)) exhibits the highest predictive value. A discriminant function of more than 32 predicted a less than 50% 30-day survival. The test suffers from the fact that it was developed and validated before the standardization of the PT as the INR. The presence of hepatic encephalopathy is also a poor prognostic indicator.

Alcoholic Cirrhosis

The prognosis of alcoholic cirrhosis depends upon drinking history, the presence of alcoholic hepatitis, and the severity of the disease. [170], [171], [172] and [173] Older literature (pre-liver transplant) indicated that the 5-year survival was nearly 85% for abstainers without jaundice, ascites, or gastrointestinal bleeding. Continued drinking lowered survival to 60%. The presence of jaundice or ascites reduced the survival to 50% in abstainers and to 30% in drinkers. Gastrointestinal bleeding carried the worst prognosis, with a 5-year survival of 35% in abstainers and 20% in drinkers. Therefore, it is prudent to urge complete abstinence for patients with any form of ALD. There are many therapies for cirrhotic patients that have become established since these data were analyzed (preventive treatment for varices and spontaneous bacterial peritonitis, endoscopic and transjugular intrahepatic porto-systemic shunt (TIPSS) treatment for varices, screening for hepatocellular carcinoma). However, for many of these therapies it has been difficult to demonstrate improvement in long-term survival.

Complications of Alcoholism and ALD

Metabolic Disorders The interaction between induced activity of CYP2E1 in alcoholics and acetaminophen is clinically very important. [174] Although therapeutic doses of acetaminophen are harmless when consumed by alcohol-naive individuals, in alcoholics such doses can produce serious liver disease with extremely high aminotransferase levels, massive perivenular hepatic necrosis, pancreatitis, and renal failure. Thus, heavy drinkers should be warned to avoid acetaminophen, and in

particular to read medication labels of over-the-counter medications to be sure they do not inadvertently consume more than 2 g per day. On the other hand, small doses of acetaminophen are safer in cirrhotic patients than nonsteroidal antiinflammatory drugs (NSAIDs), which have adverse effects on platelet and renal function. Cyclooxygensae-2 (COX-2) inhibitors have less effect on platelet function than traditional NSAIDS, but probably offer no benefit with regard to the risk of renal complications. Two problems unique to alcoholic patients are alcoholic ketoacidosis and hypoglycemia. Oxidation of ethanol is coupled to the production of NADH. The high NADH concentration inhibits fatty acid oxidation in the mitochondria. Alcoholics who are fasting while drinking heavily exhibit very high free fatty acid levels, low insulin levels, and high levels of glucagon and catecholamines. When they reduce their drinking (e.g., as a result of intercurrent illness), fatty acid oxidation is unrestrained, and there is accelerated ketogenesis with the concentration of ß-hydroxybutyrate (not detected by urine dipsticks) characteristically exceeding that of acetoacetate. [175] By administering glucose and fluids to these patients, insulin levels rise, lipolysis is curtailed, and the ketosis abates. The high NADH concentration also favors the formation of reduced metabolites (e.g., conversion of pyruvate to lactate, and dihydroxyacetone-phosphate to L-glycerol-3-phosphate). Increased lactate formation results in mild hyperlacticemia and contributes to hyperuricemia due to impaired renal excretion of urate. The reduction in pyruvate and dihydroxyacetone phosphate concentrations, two major carbon sources for glucose synthesis, inhibits gluconeogenesis, and may result in hypoglycemia. Alcoholic hypoglycemia occurs during fasting, when blood glucose levels are maintained by gluconeogenesis. [176] Hypoglycemic cerebral symptoms may be confused with alcohol intoxication. Alcoholic hypoglycemia is also readily reversible by administration of fluids and glucose, and subsequent resumption of eating. Most patients are phosphate- and magnesium-deficient as well and need to have these electrolytes replaced.

Infections Alcoholics are more prone to infections than other liver patients as a result of malnutrition, impaired phagocyte function and other immune defects, [177] and impaired sensorium (with its risk of aspiration). The most severe infections may be heralded by only a deterioration in mental status, hypotension, acute renal failure with features of prerenal azotemia or hepatorenal syndrome, or nonspecific symptoms, and frequently are not accompanied by fever. Bacteremia and spontaneous bacterial peritonitis (SBP) are common in cirrhotics. Other infections include pneumonia and lung abscess, bacterial meningitis, and urinary tract infections. [178] Patients with cirrhosis are also at increased risk of sepsis from opportunistic fungal infections, *Listeria monocytogenes,* or vibrios (especially *Vibrio vulnificus* when they consume raw seafood such as oysters [179]). Patients with possible infections should have a chest radiograph, urinalysis, surveillance blood cultures, and perhaps lumbar puncture. A sample of ascites, if present, should be analyzed for serum ascites-albumin gradient, cell count, differential, and cultured in blood culture bottles at the bedside. An absolute neutrophil count over 250/µL indicates infection. [180] Direct inoculation of ascitic fluid into blood culture bottles at the bedside substantially improves the chances of culturing an organism, as the concentration of bacteria in the fluid is quite low. [181] The presence of *Bacteroides* species or multiple organisms strongly suggests the possibility of a perforated viscus. Metaanalysis of many studies now supports the prophylactic use of antibiotics active against gram-negative rods for all cirrhotics with gastrointestinal bleeding, whether variceal or not. [182], [183], [184] and [185]

Altered Neurological Status It is difficult to evaluate sick, intoxicated alcoholic patients. The major differential diagnoses are stroke; head trauma; chronic or acute subdural hematoma; meningitis; hypoglycemia; hyponatremia; postictal state; drug, ethylene glycol, or methanol ingestion; Wernicke-Korsakoff syndrome; hepatic encephalopathy; and alcohol or drug withdrawal syndromes. [186] In most patients with impaired consciousness, a noncontrasted head CT will be performed early to exclude hematomas, tumor, and other mass lesions. Head CT is highly desirable before performing lumbar puncture, unless the clinical suspicion of meningitis is high, in which case lumbar puncture should be done promptly and antibacterial therapy initiated. Alcoholic patients often abuse drugs other than alcohol and they may ingest toxic substances by mistake. Depressant drugs, including narcotics, and phenothiazines can be detected by rapid blood and urine tests. Methanol or ethylene glycol poisoning is suggested by an anion gap metabolic acidosis (often with an osmolar gap) in the absence of ketosis. Because the requirement for thiamine is increased by carbohydrate, alcoholic patients should be given thiamine upon admission to the hospital before receiving dextrose solutions. Supplementation of other water-soluble vitamins, especially pyridoxine and folate, is advisable. Vitamin K should be given if the PT is prolonged. Hepatic encephalopathy may be difficult to diagnose. [187] Generally, there is physical evidence of chronic liver disease. The presence of fetor hepaticus (the odor of methionine catabolites) is distinctive, but not a sensitive diagnostic test. Venous or arterial blood levels of ammonia are usually elevated. Asterixis is an indicator of metabolic encephalopathy but is not specific or sensitive, and it disappears with deep coma. An electroencephalogram may help to distinguish hepatic encephalopathy from alcohol withdrawal. When hepatic encephalopathy is diagnosed, a careful search must be made for the factors that precipitated it (e.g., gastrointestinal bleeding; electrolyte and acid-base imbalance; high protein intake; severe constipation; noncompliance with medications; drug use, including acetaminophen, sedatives, and narcotic analgesics; infection; renal failure; and hepatoma). The use of centrally acting sedating drugs does not cause hepatic encephalopathy; rather the patients appear to have increased sensitivity to the sedating effects. Drug therapy of patients with liver disease must take into consideration alterations of pharmacodynamics and pharmacokinetics, [188], [189] as outlined in Table 114-6.

INCREASED DRUG METABOLISM	INCREASED DRUG/ CHEMICAL TOXICITY	INCREASED DRUG TOLERANCE
Pentobarbital	Acetaminophen	Anesthetics
Meprobamate	Vitamin A	Barbiturates
Warfarin	Isoniazid	Meprobamate
Tolbutamide (and other sulfonylureas)	Phenylbutazone	Benzodiazepines
Phenytoin	Halothane	
Cocaine	Enflurane	
Rifampin	Carbon tetrachloride	
Aminopyrine	Benzene	
Methadone	Nitrosamines	
	Cocaine	
INCREASED PHARMACODYNAMIC EFFECT		**ANTABUSE-LIKE REACTIONS**
H_1 antihistamines		Tolbutamide
Barbiturates		Metronidazole
Benzodiazepines		Griseofulvin
Chloral hydrate		Quinacrine
Meprobamate		Pargyline
Narcotics		Reserpine
Phenothiazines		Phenylbutazone
Phenytoin		Moxalactam, and several other second- and third-generation cephalosporins

TABLE 114-6 Interactions Between Chronic Alcohol Abuse and Drug Actions and Metabolism

Renal Dysfunction Patients with ALD may have renal dysfunction at the time of admission, or more likely, it may develop during hospitalization. If the patient has advanced, chronic liver disease with tense ascites or has been treated vigorously with diuretics, the hepatorenal syndrome (HRS) should be considered, [190] but other causes of renal failure should be excluded. Urine studies will allow one to distinguish acute tubular necrosis from either prerenal azotemia or hepatorenal syndrome. Typically in the latter, the urine sodium is low (<10 mEq/L), the fractional excretion of sodium (FE_{Na}) is less than 1%, the urine:serum creatinine ratio is over 30, and the urine osmolality is high (urine osmolality more than 100 mOsm/L greater than serum osmolality), with no proteinuria or casts, and an acellular urinary sediment. In order to distinguish prerenal azotemia (intravascular volume depletion) from HRS, careful physical examination and monitoring of chest radiographs must be employed as one judiciously infuses crystalloid and colloid. Ultimately, measurement of pulmonary artery pressure might be needed to ensure that the intravascular volume is adequately expanded. Other potentially reversible renal disease must also be considered (e.g., urinary tract obstruction, a drug side effect, radiocontrast-induced nephropathy, sepsis, rhabdomyolysis, hepatitis B- or C-associated cryoglobulinemia, and toxicity from inhalants such as toluene). The therapy for suspected HRS should include paracentesis, careful crystalloid and colloid administration, and continuous venovenous hemofiltration (preferred over hemodialysis). Ultimately orthotopic liver transplantation is the only definitive therapy for HRS and end-stage liver disease. Peritoneovenous shunts have not been shown to improve survival in HRS, [191] and have fallen out of favor because of the high rate of shunt occlusion. The role of TIPSS in the treatment of HRS is being assessed, [192], [193] but is not likely beneficial. The use of liver support systems is experimental.

Worsening Liver Disease Alcoholic patients may have historical or physical findings that suggest that previously diagnosed liver disease has worsened. Of course, it cannot be assumed that the abnormalities are all due to alcohol abuse. The patient may have superimposed viral hepatitis, drug toxicity, hepatoma, or biliary obstruction (up to 30% of patients with ALD have gallstones, usually bilirubin pigment stones [194]). The patient should be closely questioned about ingestion of drugs, toxins, herbal medications and OTC medicines, and exposure to viruses (e.g., by sexual contact, blood and blood product transfusions, or intravenous or nasal drug use). The physical examination should be directed toward finding new evidence of congestive heart failure or pericardial constriction, changes in liver size, abnormal masses, hepatic bruits, visible or palpable gallbladder, abdominal tenderness, Murphy's sign, and so on. Hepatomegaly suggests fat, tumor, mass, or infiltrative lesions, but is also common in uncomplicated cirrhosis. The liver tests may help with the diagnosis (transient high transaminases with abdominal pain suggests passage of a common duct stone; very high transaminases and new jaundice suggests a drug toxicity such as acetaminophen or viral hepatitis), and lead to evaluation for parenchymal liver diseases (hepatitis serology, autoimmune markers, markers of iron and copper overload). Predominant elevation of the alkaline phosphatases indicates cholestasis, but does not differentiate intrahepatic from extrahepatic causes. Radiologic imaging by CT or ultrasound should be performed to look for dilated ducts or intrahepatic masses. Magnetic resonance cholangiograms are usually not useful in patients with end-stage liver disease because ascites interferes with this imaging method. Masses usually require radiologically guided biopsy, but alternatively, selective intraarterial injection of an oily contrast material known as lipiodol may provide the diagnosis of malignancy. If the ducts are dilated, a cholangiogram should be obtained to find the site and possibly the cause of the obstruction. If the patient has symptoms suggestive of intermittent obstruction (attacks of pain or symptoms of biliary infection) but no dilation of the ducts, a cholangiogram should also be performed. If neither biliary obstruction nor masses are present, primary biliary cirrhosis and infiltrative diseases should be considered and the liver should be biopsied.

TREATMENT

The treatment of ALD should be divided into treatment of the underlying alcoholism, and measures to address the three major stages of ALD. The latter includes attempts to prevent or slow fibrosis, to control the inflammation of alcoholic hepatitis, and finally the treatment of complications that are common to other forms of end-stage liver disease.

Treatment of Alcoholism

The success of therapies for alcohol abuse varies from 30% to 90%. Results are improved when the abuse is detected at an early stage when the patient still has support from family and employers. Patients are first detoxified, usually as outpatients, then enter a phase of maintenance of sobriety. Current methods for monitoring alcohol consumption in patients who are under treatment are not satisfactory, despite the availability of the carbohydrate-deficient transferrin (CDT) test for well over a decade. [195], [196] and [197] This form of transferrin lacks the normal number of sialic acid moieties due to effects of ethanol on its posttranslation modification in the Golgi apparatus, and on its rate of removal from the plasma. Abstinence is sought through cognitive therapy and behavior modification counseling, augmented by pharmacological therapy. There is evidence from animal and clinical studies that opioid antagonists can improve the results of alcoholism treatment, and naltrexone has been approved for use in treatment of alcoholism, in conjunction with behavioral or psychological therapy. Additional medications to reduce craving, such as acamprosate and ondansetron, have shown promise in clinical trials. For most patients, optimal success will entail a combination of behavior modification, regular involvement in support groups such as Alcoholics Anonymous, and pharmacological therapy. Co-morbid psychiatric problems such as depression also need to be addressed. Alcohol abuse and its treatment are discussed in more detail in Chapter 55.

Treatment of ALD

McCullough and colleagues [198] have published a set of practice guidelines on the management of ALD. They summarized that treatment remains limited to the following:

- abstinence is crucial in the therapy for ALD
- there is general consensus to use corticosteroids to treat only those patients with severe alcoholic hepatitis
- there is good scientific basis to maintain good nutritional status at all stages of ALD
- liver transplantation should be considered in chronic ALD
- management of complications of chronic liver disease (e.g., ascites, portal hypertension-associated bleeding, hepatic encephalopathy, and hepatocellular carcinoma) is the same as for other causes of cirrhosis.

Alcoholic fatty liver responds well to abstinence and is not thought to cause lasting sequelae, thus it will not be discussed further. In addition to the therapies discussed in the following paragraphs, patients should be vaccinated against hepatitis A and B to prevent the often disastrous consequences of hepatitis superimposed on ALD, and should receive the polyvalent pneumococcal vaccine.

Alcoholic Hepatitis

Cessation of alcohol drinking is paramount in the treatment of alcoholic hepatitis, and is best accomplished by the usual measures discussed previously. Protein-calorie malnutrition is a common finding in patients with ALD. [142] In the VA Cooperative Study on Alcoholic Hepatitis, more than 75% of patients with severe liver disease exhibited signs of kwashiorkor, marasmus or both and malnutrition worsened the prognosis of alcoholic hepatitis. Several small controlled trials have tested the effect of enteral or parenteral amino acid therapy in alcoholic hepatitis. [199], [200], [201] and [202] These trials have shown faster improvement in serum bilirubin, serum albumin, and ascites, and no deterioration in hepatic encephalopathy in the patients provided amino acids. Only one of the controlled trials has demonstrated improved mortality rate, but most have been limited in this regard by insufficient statistical power. Certainly, most patients with alcoholic hepatitis are malnourished and this should be addressed either enterally or parenterally. [203] For these patients, the “routine” prescription for a 2-g sodium, 40- to 60-g protein diet is inappropriate. Although marketed aggressively, amino acid mixtures enriched in branched-chain amino acids are recommended only for patients who have not responded to conventional nutritional therapy. [204]

Corticosteroids modulate the immune system (including the production of the cytokines involved in liver injury) and inhibit the activation of HSC and fibrogenesis. The use of corticosteroids (prednisone, prednisolone, or 5-methylprednisolone in the dose range of 35 to 80 mg/d for 4 to 6 weeks) in the treatment of alcoholic hepatitis has been extensively studied in numerous randomized control trials and several metaanalyses. [205], [206], [207], [208] and [209] They demonstrated that:

1. Only patients with hepatic encephalopathy or Maddrey score of more than 32 should be treated.
2. Treatment should exclude patients with gastrointestinal bleeding, active infection, renal failure, or acute pancreatitis. The problem of unrecognized infection, or infection that occurs during the course of therapy, is of major concern.
3. Corticosteroids will reduce mortality risk by 25%.
4. One has to treat seven patients in order to avoid one death.

Overall, it appears that corticosteroid therapy benefits some patients with severe alcoholic hepatitis in the absence of contraindications, but this still remains controversial.

A randomized controlled trial [210] compared the therapeutic efficacies of corticosteroid therapy (40 mg/d) versus enteral nutrition (2000 kcal/d). Outcomes were assessed at 28 days (the end of therapy) and 1 year later or until death. Sixty-two percent of patients treated by enteral nutrition and 39% of patients treated with corticosteroids were alive at the end of 1-year follow-up. The difference was not statistically significant. Mortality during the 28 days of therapy occurred earlier with enteral nutrition but mortality beyond the first 28 days was higher with corticosteroid therapy, largely due to infections. Perhaps by deploying both therapies in combination the respective shortfalls of corticosteroid and enteral nutrition therapies can be offset.

Anabolic-androgenic steroids such as oxandrolone have been tested in an attempt to enhance recovery (i.e., to increase the removal of hepatic fat, to stimulate protein synthesis, and to accelerate cell repair and hepatic regeneration). Oxandrolone was tested in a VA cooperative study in a 30-day trial in patients with moderate to severe alcoholic hepatitis. Oxandrolone appeared to be most beneficial in patients with moderate malnutrition, particularly in those who ingested sufficient calories during the trial. [211] This emphasizes the importance of attention to the patients’ nutrition and the difficulty of performing such studies. Oxandrolone has not been widely used despite this trial; additional data on subsets of patients who will benefit most would probably increase physicians’ interest in this drug.

Propylthiouracil (PTU) has been used to treat alcoholic hepatitis, possibly by decreasing the hypermetabolic state induced by alcohol and protecting perivenous cells from hypoxic injury. [212], [213] It also inhibits neutrophil myeloperoxidase and may thereby reduce oxidative stress from hypochlorous acid made in the neutrophils. In one study, PTU improved symptoms and laboratory tests; [214] but in another series of more severely ill patients, PTU was not effective. [215] In the largest controlled trial, 310 patients with ALD were randomized to either PTU or placebo and followed for up to 2 years. PTU therapy resulted in a cumulative mortality rate that was half that of the placebo group among moderately ill patients, and in a subgroup of severely ill patients. [216] Thus, PTU therapy may benefit patients with moderate to severe alcoholic hepatitis. Other inhibitors that block myeloperoxidase but do not interfere with thyroxine production may ultimately be of value. Pentoxifylline has been reported to improve outcomes in alcoholic hepatitis, in particular it reduced the frequency of HRS. [217] The absolute risk reduction of HRS in this trial was an impressive 40%. This may be due to inhibition of the actions of TNFa and other cytokines and chemokines.

Alcoholic Cirrhosis

Alcoholic cirrhosis has been treated with corticosteroids without effect on survival. Colchicine (1 mg/d, 5 d/wk) improved survival in one study with a 14-year follow-up. [218] Oddly, much of the mortality in the placebo-treated group was not due to liver disease. The actions of colchicine are being studied further in an ongoing VA cooperative study.

Several other agents including polyunsaturated lecithin and S-adenosylmethionine (SAM) are being tested in multicenter trials. Lieber [219] has reviewed the actions of

polyunsaturated lecithin in the treatment of liver fibrosis and determined:

- it protects alcohol-fed baboons against alcohol-induced fibrosis and cirrhosis and prevented the depletion of hepatic dilinoleoylphosphatidylcholine
- it attenuates the transformation of HSCs into myofibroblasts
- it augments the defenses against oxidative stress, reduces the levels of CYPE21, and normalizes 4-HNE, F2-isoprostanes (an index of lipid peroxidation), and glutathione (GSH) levels
- it stimulates collagenase production in HSCs, thereby decreasing collagen accumulation in the liver.

A randomized double-blind multicenter trial that includes 800 patients with ALD with or without HCV infection is still ongoing.

SAM administration has been shown to be useful in experimental animals by replenishing this transmethylating agent, enhancing the synthesis of polyamines, and providing cysteine for glutathione synthesis. [220] The results of a large clinical trial were recently published. [221] SAM may reduce mortality or need for transplantation in cirrhosis, especially in those with less advanced liver disease. Future therapies may be suggested based on the mechanisms of injury discussed previously. These include better antioxidants, inhibitors of CYP2E1, inhibitors of NF-?B activation or of eicosanoid production by Kupffer cells, antagonists or antibodies against PDGF, TNFa, and TGFß, and inhibitors of apoptosis in hepatocytes.

Patients with advanced (Child's B or C) alcoholic cirrhosis can be treated by liver transplantation provided that they do not have other alcohol-related co-morbid disorders, they have undergone intensive outpatient treatment and rehabilitation, and have at least a 6-month period of documented abstinence. [222], [223] and [224] It has been of interest to use the CDT to test for abstinence in this population. However, CDT has only recently been evaluated in patients before and after liver transplantation. [225] Elevated CDT was found in a number of abstinent patients who had long-term, solid evidence of abstinence proven by negative biochemical short-term tests and negative psychological evaluations. After liver transplantation, patients with both ALD and non-ALD had significantly lower CDT values than before transplantation. The results suggest that CDT is not useful in evaluating patients before transplantation but may be useful in posttransplant settings.

Guidelines for treatment for alcoholism prior to transplantation vary from center to center, but the 2-year survival of carefully selected patients compares quite well with those transplanted for other indications. Additionally, the quality of life of these transplanted patients is excellent with many patients returning to part-time or full-time employment. The rates of return to drinking after transplantation vary between 10% to 30% depending on candidate selection process and criteria for relapse. ALD now accounts for approximately 25% of adult liver transplantations in the United States.

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CHAPTER 115

Oliver F. W. James

NONALCOHOLIC FATTY LIVER DISEASE

As Maher has stated “NASH [nonalcoholic steatohepatitis] has become a household word in hepatology.” [1] The clinical and pathological features of nonalcoholic fatty liver disease have been recognized for 40 years. [2], [3] It was not until 1980 however that Ludwig and colleagues coined the term *nonalcoholic steatohepatitis (NASH)* to describe “the pathological and clinical features of non-alcoholic disease of the liver associated with the pathological features commonly seen in alcoholic liver disease itself.” [4] While the acronym NASH has been useful in drawing attention to this important liver disease the term *steatohepatitis* excluded the appearance of fat alone in the liver, yet pure fatty liver is part of the spectrum of nonalcoholic fatty liver disease as a whole and pure fatty liver is probably the precursor of steatohepatitis and fibrosis. [5] In 1999 McCullough and colleagues coined the term *nonalcoholic fatty liver disease (NAFLD)* to describe the whole spectrum of disease from simple fat through steatohepatitis and fibrosis to cirrhosis. This account refers to the whole spectrum of this disease as NAFLD and confines the term NASH to steatohepatitis with or without fibrosis or cirrhosis.

It is becoming clear that NAFLD is an important part of the insulin resistance, obesity, hyperlipidemia, hypertension, constellation known as syndrome X (the metabolic syndrome) and like the other features is rapidly becoming more common in overnourished, underexercised Western society. [6]

EPIDEMIOLOGY

There are no epidemiologic studies of the exact incidence or prevalence of NASH but there are a number of clues from several countries that suggest that it is becoming one of the commonest liver diseases in Western countries. In a study of Canadian office hepatology practices NASH was the second most common liver disease encountered (after hepatitis C). [7] In N-HANES-3 (USA) cryptogenic elevation in serum ALT [alanine aminotransferase] was seen in 2.6% of the population after exclusion of other possible causes. This was independently associated with increased waist: hip ratio and indices of insulin resistance (summarized in ref. [6]). In an ultrasound-based study in northern Italy 75% of obese individuals were found to have fatty liver; furthermore, 16% of nonobese, nonheavy drinking (by Italian standards) individuals were also found to have fatty liver on ultrasound. [8] While significant obesity and type 2 diabetes are becoming more common in adults, possibly more importantly, this is also occurring alarmingly in children. For example, in children in the United Kingdom between 1984 and 1994, obesity more than doubled. Almost 20% of Scottish girls between 9 and 11 were overweight in 1994, twice the prevalence of 10 years earlier. [9]

How many individuals who have fatty liver alone will go on to develop steatohepatitis and what proportion of these may develop cirrhosis is still quite unclear, but it may be a significant proportion.

PATHOGENESIS

Since fatty liver appears to be a necessary but not sufficient condition for the subsequent development of steatohepatitis and fibrosis, it has been suggested that the development of fatty liver is the first hit in a “two-hit” model of the pathogenesis of NASH. [10] Over 50% of obese individuals have fatty liver, and the more obese the more likely the fatty liver. Furthermore, central obesity is more closely related to fatty liver. [11] It is suggested that free fatty acids (FFAs) are mobilized more readily from visceral than from subcutaneous fat and visceral fat drains this increased load of FFAs directly into the liver, thus supplying increased hepatic triglyceride. [12]

Insulin Resistance and NAFLD

As well as increased delivery of FFA to the liver, insulin resistance appears to be necessary for the development of fatty liver. Of 46 NALFD (diagnosed by ultrasound), nondiabetic individuals, Marchesini and colleagues [13] found that 100% all had insulin resistance.

In careful studies, Sanyal and colleagues [14] used a two-step hyperinsulinemia euglycemic clamp to examine insulin resistance in patients with NASH, those with fatty liver alone, and controls. Patients with known diabetes or with cirrhosis were excluded. Both patients with NASH and fatty liver demonstrated insulin resistance. Using steady state enrichment of [6,6- $_2$H] glucose hepatic output was assessed before and during clamping by measuring differences between total enrichment and glucose infusion rate. This showed marked suppression of hepatic glucose output in both fatty liver and NASH subjects during insulin infusion. The Sanyal group thus demonstrated hepatic as well as peripheral insulin resistance. [14] Combined peripheral and hepatic insulin resistance in both pure fatty liver subjects and patients with NASH was confirmed by Marchesini and colleagues, [15] who found glucose disposal in patients with NAFLD was similar to well-controlled type 2 diabetics. Interestingly, among the NAFLD patients there was no difference in glucose disposal between patients with body mass index (BMI) less than 25 kg/m ^{2} and those with BMI over 25 kg/m ^{2}. This again points to the necessity of insulin resistance rather than mere generalized obesity in the pathogenesis of nonalcoholic fatty liver.

Development of Steatohepatitis

Two main pathways have been implicated in the development of steatohepatitis—these are oxidative stress caused by lipid peroxidation and release of reactive oxygen species (ROS) on the one hand, and the release of the endotoxin-cytokine proinflammatory cascade on the other. As we will see the two are by no means mutually exclusive and FFAs are important in both. FFAs are endogenous ligands for peroxisomal proliferator activated receptors a (PPARa) and ? (PPAR?). The severity of NASH has been shown to correlate with levels of serum FFAs (KD Lindor, personal communication, October 2000). Because as we have seen there may be hepatic as well as peripheral insulin resistance there is not only an increased supply of FFAs to the liver but increased mitochondrial ß-oxidation which itself generates reactive oxygen species.

Oxidative Stress In mammalian cells peroxisomes oxidize long chain and very long chain FFAs. Mice lacking the gene encoding fatty acyl-coenzyme A oxidase (AOX), the initial enzyme of the inducible peroxisomal ß-oxidation system, developed severe microvesicular NASH [16]— suggesting that an excess of long chain and very long chain acyl-coenzymes A (CoAs) can lead to the lesion. In contrast AOX -/-mice which also lack PPARa develop only mild steatosis suggesting first that the

long chain and very long chain acyl-CoAs can function as endogenous PPARa ligand and second that PPARa-induced genes must play a part in development of steatonecrosis/hepatitis. [17] PPARa regulates the transcription of some CYP450 enzymes involved in long chain FFA oxidation, including AOX and CYP4A family members. This metabolism via these extra mitochondrial pathways generates ROS—either hydrogen peroxide in the case of peroxisomal ß-oxidation or superoxide in the case of CYP4A catalyzed w-oxidation. Metabolism of long chain FFA by CYP4A also generates dicarboxylic fatty acids that are toxic to mitochondria leading to apoptosis and necrosis. [18] In the lipid-rich methionine-choline deficient (MCD) diet in the mouse, expression of two major CYP4A proteins (4A10 and 4A14) is greatly increased, hence, potentially increasing the generation of dicarboxylic fatty acids toxic to mitochondria and increasing apoptosis and necrosis. [19] Just as ethanol induces CYP2E1 so fatty acids are both substrate for and inducers of CYP2E1, and CYP2E1 activity is induced in diabetes. Microsomal CYP2E1 is an important microsomal catalyst of lipid peroxidation in the MCD mouse model and in patients with NASH. Studies have demonstrated marked induction of hepatic CYP2E1 correlated with measures of lipid peroxidation. [20], [21]

Endotoxin Cytokine-Mediated Injury

The human “model” of accelerated NASH provided by the series of patients given jejunoileal bypass (JIB) surgery for obesity was the initial clue to a second putative mechanism for steatonecrosis. [22] This “blind loop” provided an ideal source of bacterial endotoxin to be absorbed into the already fatty liver. Interestingly, early studies showed that this could be prevented or reversed in some patients by the antibiotic metronidazole. [22] A few patients have been described with bacterial contamination of the small bowel associated with NASH and it has been suggested that up to 50% of patients with NASH may have small intestinal bacterial overgrowth and raised serum tumor necrosis factor-a (TNFa); [23] these observations, however, need to be confirmed. In the mouse model it has been demonstrated that obesity increases sensitivity to endotoxin-induced liver injury and that there are altered hepatic lymphocyte subpopulations in obesity-related murine fatty liver. [24], [25] In addition FFAs are ligands for PPARa and these ligands inhibit macrophage function.

Fibrosis

Oxidative stress and the proinflammatory cascade lead to necroinflammation but independently excess fat has been shown to activate stellate cells leading to fibrosis. The greater the degree of fatty change in human fatty liver biopsies, the greater the number of activated stellate cells. [26] Since hypertension (part of the metabolic syndrome) is probably associated with NAFLD the finding that angiotensin II enhanced the production of transforming growth factor-ß1 (TGF-ß1) which contributes to stellate cell activation offers another putative mechanism for enhanced fibrosis. [27], [28] Very recently, leptin resistance, associated with insulin resistance, has also been implicated in fibrogenesis (AASLD Consensus, Atlanta, Sept. 2002).

In summary, the pathogenesis of NASH is complex. It offers obvious parallels with the pathogenesis of alcoholic steatohepatitis. In NASH insulin resistance and increased supply of FFA appear to be essential. The varying roles of oxidative stress, the endotoxin cytokine-mediated proinflammatory response and, more specifically, fibrotic stimuli probably determine which individuals develop NASH/cirrhosis and which merely retain a fatty liver.

Genetic Determinants

The mechanisms described for the development of NASH give clear indications of where we should look for genetic determinants both of the development of fatty liver and of NASH although these will be extraordinarily complex. Familial tendency to NASH and insulin resistance is now being described. [29] What is known, or rather what is not known, has been summarized by Day and Daly. [30] It has been shown that over-expression of the enzyme 11ß-hydroxysteroid dehydrogenase type 1 (11ß-HSD-1), which controls glucocorticoid activation in adipose tissue, leads to a phenotype of central obesity—at least in the mouse. [31] This may offer an important clue to the causation of central obesity in humans and hence, if variations of expression of 11ß-HSD-1 are confirmed, a clue to genetic susceptibility to NASH. This is speculative but indicative of current research thinking.

CLINICAL MANIFESTATIONS

Most patients with NASH have few symptoms or signs suggestive of chronic liver disease. Because case series are heterogeneous in their derivation—some being derived from groups of obese individuals for example—it is difficult to give precise figures. However, up to 30% of patients complain of persistent fatigue and lethargy, up to 30% have persistent right upper abdominal discomfort in the absence of gallstones, and probably about 50% of all patients are asymptomatic. Older series suggested that over 75% of patients were female and markedly obese but more recently the proportion of men reported with NAFLD or NASH has increased, many without marked obesity. [32] It is clear that increasing levels of obesity are, not surprisingly, accompanied by increasing likelihood of more severe histological change. Clinical examination is almost always unremarkable. The liver may be palpable but there are no other signs to suggest liver disease until the complications of cirrhosis develop. The most important features of examination of a patient with putative NAFLD are to measure patients’ weight and height—hence BMI and, if practiced, measure waist:hip ratio as an assessment of central obesity.

History

Paramount in history taking for NASH is careful documentation of alcohol consumption, including events in the medical or social history of the patient that might suggest previous heavy alcohol use. A history or the presence of other diseases associated with the metabolic syndrome—type 2 diabetes (noninsulin-dependent diabetes mellitus, NIDDM), hyperlipidemia, and hypertension should be recorded. A history of previous obesity and its duration, together with recurrent weight-reducing diets may also be important.

Although NAFLD has thus far only been discussed in the context of the metabolic syndrome, there are a number of other important rare potential causes of NAFLD (Table 115-1).

Surgical procedures for obesity
Jejunoileal bypass (JIB)
Gastroplasy
Biliopancreatic diversion
Surgery leading to major weight loss
e.g., massive intestinal resection
Total parenteral nutrition (adults)
more likely after intestinal resection
Weight cycling/bulimia
Bacterial contamination of small bowel
e.g., Jejunal diverticula
Medications
Amiodarone
Perhexiline
(Coralgil)
?? Calcium channel blockers
Tamoxifen
Environmental
? Dimethylformamide
? Toxic oil syndrome
Syndromes of severe insulin resistance
Lipodystrophies (complete or partial)
Insulin receptor mutations

TABLE 115-1 Causal Associations With Nonalcoholic Steatohepatitis

Previous Weight-Reducing Surgery Ten percent or more of patients undergoing JIB developed steatohepatitis, liver fibrosis, and, not infrequently, hepatocellular failure or the complications of cirrhosis—usually within 1 or 2 years but occasionally many years postoperatively. [33] More modern weight reduction surgery—for example gastroplasty—is now the surgical treatment of choice for morbid obesity. NASH has been shown to be present in up to a one fourth of these patients at surgery, although very severe deterioration or complications postoperatively are now much rarer than with JIB. [34]
Weight Cycling Occasionally individuals have been described who have made rapid weight reduction with “crash diets” only to regain weight again. At least one patient died of hepatic decompensation and cirrhosis in these circumstances. [35]
Total Parenteral Nutrition Total parenteral nutrition (TPN), particularly following small bowel resection, is followed by NASH in perhaps 15% of adult patients.

Occasionally this leads to cirrhosis. Presumably this may be related to weight loss with associated increased delivery of FFAs, possibly accompanied by small bowel bacterial contamination leading to release of endotoxin-TNFa, but this is speculative. [36]

Drugs A number of drugs have been implicated in the development of NASH. All of these reactions are rare and associated with other manifestations of drug hepatotoxicity. The most well re- cognized of these among currently used drugs is with amiodarone where “pseudo-alcoholic” liver disease has been detected in up to 1% patients receiving amiodorone, almost all taking the drug for over a year. This was in addition to more familiar iodine deposition and phospholipidosis. [37] More recently the estrogen-receptor ligand tamoxifen has been implicated in the development of not only fatty liver but NASH as well. However, a number of the reports of this apparent association have been among obese women so that the relationship between tamoxifen and true NASH is still rather unclear (summarized in ref. [20]). A recent report from Brazil suggests a relationship between exposure at work to petrochemicals in the environment and development of NASH; in some of these individuals liver function and histology improved following removal from the environment. [38] In general, however, the relationship between industrial or environmental exposure to, for example, petrochemicals, and development of NASH, is highly speculative.

Familial Lipodystrophy Severe insulin resistance is present in patients with complete or partial lipodystrophy. A number of these individuals, including some children, develop steatohepatitis with Mallory bodies and liver cell degeneration. [39] Very rare insulin receptor mutations are also associated with steatohepatitis.

Ethanol While it is vital to emphasize exclusion of excessive alcohol ingestion when making the diagnosis of NAFLD the relationship between ethanol consumption and NAFLD is interesting. While “excess” ethanol ingestion is almost certainly synergistic with other risk factors for fatty liver and steatohepatitis—obesity and insulin resistance—the exact meaning of “excess” is still unclear. Certainly, excess weight is perceived as a risk factor for the development of severe alcoholic liver disease. [40] Dixon and colleagues, however, have suggested that mild (probably less than 20 g/d) ethanol consumption among obese individuals reduced the risk of development of NASH, possibly by increasing insulin sensitivity but this is highly speculative. [27] It is recommended that patients with NASH, or steatohepatitis in which obesity/insulin resistance and higher ethanol consumption may each contribute to the liver disease should consume less than 20 g (2 U) ethanol/d.

DIAGNOSIS

The diagnosis of NAFLD/NASH is based upon the demonstration of a fatty liver ± steatohepatitis and fibrosis, rigorous exclusion of significant ethanol consumption, and exclusion of other conditions in which fatty liver can occur—for example hepatitis C. Patients with NAFLD often present with “cryptogenic” persistent elevation of ALT.

Laboratory

The standard range of liver function tests and other blood tests used in assessment of patients with liver disease are employed in evaluation of patients with possible NASH. There are a few features of these tests that help to distinguish NASH from ALD although none is really specific. These are shown in Table 115-2A. In mild NASH the aspartate aminotransferase:alanine aminotransferase (AST:ALT) ratio is often less than 1 whereas in alcoholic liver disease the reverse is true. In more severe NASH however AST is often higher than ALT. Note that ?-glutamyltranspeptidase (?GT) is elevated in both NASH and alcoholic liver disease. Very few patients with NASH have elevated red cell mean corpuscular volume (MCV) whereas macrocytosis is a common indicator of excess alcohol consumption.

TEST	NAFLD	ALCOHOLIC
AST/ALT	Often <1 (if mild)	>1
Bili	Normal	May be ↑ or ↑↑
Alk Phos	Usually normal until late	May be ↑
Albumin	Normal until late	Often ↓ or ↓↓
PT	Normal until late	Often ↑ or ↑↑
Gamma GT	↑	↑ or ↑↑
MCV*	Normal	↑ or ↑↑
Blood ethanol	Absent	Often present
Lipids	May be ↑ or ↑↑	May be ↑ or ↑↑
Ferritin	May be ↑ (slight)	May be ↑ or ↑↑

Alk Phos, alkaline phosphate; AST/ALT, aspartate aminotransferase/alanine aminotransferase; GT, glutamyl transpeptidase; MCV, mean corpuscular volume.

*Personal look back has shown 0/35 nonalcoholic steatohepatitis (NASH) patients with raised MCV. Ratziu et al.[42] had raised MCV in 0/93 NAFLD patients.

TABLE 115-2A Nonalcoholic Fatty Liver Disease (NAFLD) Laboratory Investigations

Other liver diseases are excluded by conventional means (see Table 115-2B). The presence of anti-hepatitis C virus (HCV) excludes a diagnosis of NASH alone. While the appearance of fatty liver in patients with genotype 3HCV may be virally induced, fatty liver and HCV may coexist in other genotypes and should not be regarded as mutually exclusive. Fat on biopsy in patients with HCV suggests the possibility of accelerated progression of fibrosis in these individuals. [41] An autoimmune profile should be checked although a few NASH patients have been described with low titers of anti-smooth muscle antibody by Bacon and colleagues [42] and Teli and colleagues. [43] The subject of iron markers is slightly confused since some groups believe that excess liver iron may be one of the factors promoting liver inflammation and fibrosis, through oxidative stress, in patients who have fatty change. [44] While some patients with NASH do have slightly raised serum ferritin this is probably a reflection of inflammatory change in the liver rather than of increased iron stores in most cases. HFE genotype now allows definitive diagnosis or exclusion of hemochromatosis where any doubt exists.

HBV and HCV markers	Negative[†]
Iron saturation	Normal and *HFE* gene wild type
Autoimmune profile	Normal[*]

HBV, hepatitis B virus; HCV, hepatitis C virus.

*Note a few NAFLD patients described with low titer smooth muscle antibody.

†Note HCV and NAFLD may coexist.

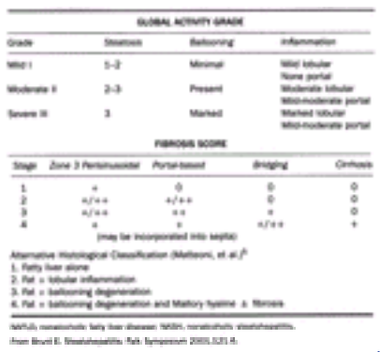
TABLE 115-2B Nonalcoholic Fatty Liver Disease (NAFLD) Laboratory Investigation: Exclusions

Liver Imaging

The most inexpensive, quickest, and probably still the most effective mode of liver imaging in patients with suspected fatty liver is with ultrasound. This confirms the presence of fat. Importantly, while ultrasound can demonstrate the likelihood of cirrhosis by gross changes in liver outline and signs of portal hypertension, liver fibrosis and steatosis can have similar sonographic appearances which can be indistinguishable. [45] Thus NASH cannot be “staged” by ultrasound or any other imaging modality. Small areas of focal fat within the liver should not be regarded as suggestive of NASH.

Liver Biopsy

Liver biopsy is essential to determine the presence and severity of steatohepatitis and extent of fibrosis-cirrhosis, and to give the best idea of likely prognosis. [46] It is also mandatory for the conduct of good clinical trials. Histopathologists may argue about the grading and staging of liver disease, but the suggestion of Brunt and colleagues to provide a global activity grade and a fibrosis stage seems sensible and its reproducibility appears reliable (AD Burt, personal communication, April 2002). This system, or something like it will probably be widely adopted [47] (Table 115-3). As implied in the original definition of NASH by Ludwig and colleagues, [4] there are really no distinguishing histological features between alcoholic and nonalcoholic steatohepatitis.



GLOBAL ACTIVITY GRADE			
Grade	Steatosis	Ballooning	Inflammation
Mild I	1–2	Minimal	Mild lobular None portal
Moderate II	2–3	Present	Moderate lobular Mild-moderate portal
Severe III	3	Marked	Marked lobular Mild-moderate portal

FIBROSIS SCORE				
Stage	Zone 3 Perisinusoidal	Portal-based	Bridging	Cirrhosis
1	+	0	0	0
2	+/++	+/++	0	0
3	+/++	++	+	0
4	+	+	+/++	+

(may be incorporated into septa)

Alternative Histological Classification (Matteoni, et al.)[3]
1. Fatty liver alone
2. Fat + lobular inflammation
3. Fat + ballooning degeneration
4. Fat + ballooning degeneration and Mallory hyaline ± fibrosis

NAFLD, nonalcoholic fatty liver disease; NASH, nonalcoholic steatohepatitis.
From Brunt E. Nonalcoholic fatty liver disease. NASH. nonalcoholic steatohepatitis.

TABLE 115-3 Histology: Grading and Staging NASH/NAFLD

In the context of a younger patient with no overt clinical liver disease, with ultrasound appearances diagnostic of fatty liver and persistent isolated elevation of ALT, it may be argued that outside the setting of a clinical trial liver biopsy is unnecessary. In view of lessons from the natural history of the disease (discussed subsequently) this author's current recommendation would be that unless there are clinical indications to the contrary, liver biopsy is unnecessary under the age of 40 to confirm a diagnosis of NASH. Under the age of 40, biopsy is unlikely to yield important prognostic information. In view of lessons from the natural history and of the inability of ultrasound to distinguish fat from inflammation/fibrosis, the author's present practice is to recommend liver biopsy in all patients over age 50 for information on grading and staging, which will inform physician and patient of prognosis.

COURSE AND COMPLICATIONS

Our knowledge of the natural history of NASH was, until a few years ago, based upon case series (summarized in ref. [48]). Three large cross-sectional studies and one quasi-longitudinal study, however, have allowed us to begin to understand the natural of history of NASH in adults. Pediatric NASH, the most common cause of abnormal liver function tests (LFTs) in adolescents, is outside the scope of this chapter. A cross-sectional study from the Mayo Clinic examined 144 patients in whom liver biopsy had been carried out and a diagnosis of NAFLD had been made after conventional exclusions of other liver disease. The diseases the patients had ranged from simple fatty liver to severe cirrhosis. A range of clinical and laboratory parameters were examined as predictors of severity of histological change. In these patients age was the most significant predictor of degree of fibrosis ($p < 0.001$). Only 4% of patients under age 45 (1 individual) had marked fibrosis, no nonobese, nondiabetic patient under age 45 had severe fibrosis. [49] Ratziu and colleagues [50] examined 93 obese patients with abnormal LFTs who had received liver biopsy and were found to have varying degrees of NAFLD. They found that age over 50 correlated independently with septal fibrosis (odds ratio 14:1 age over 50 vs. age under 50). They also found that if BMI was less than 30, age less than 50, and ALT elevated less than twofold then no patient had septal fibrosis or cirrhosis. They did find that 10 patients (12%) had cirrhosis on biopsy that had not been clinically evident in any way. In a series of 105 consecutive very obese (BMI > 35 kg/m ^{2}) patients having laparoscopic gastric banding, mean age 41, mean BMI 47 kg/m ^{2}, many of whom had normal LFTs, Dixon and associates [27] found 19% had steatohepatitis, 10 had advanced fibrosis, 1 had cirrhosis. Seventy-five individuals had steatosis ± nonspecific inflammation alone. The mean age of this group was 41. Of importance, the presence of NASH correlated with the presence of hypertension and measures of central obesity most closely. The longitudinal study from the Cleveland Clinic underlined the importance of the presence or absence of NIDDM. Whereas 25% of 44 patients with diabetes in this series developed cirrhosis (with a mean follow up of 9 years) only 10% of 88 patients without type 2 diabetes had cirrhosis on an initial biopsy or developed it at follow-up. [5]

At the "fat alone" end of the NAFLD spectrum Teli and colleagues [43] followed up 40 patients with simple fatty liver, most around the age of 40, in whom alcohol had been rigorously excluded. None developed significant clinical liver disease over an 11-year follow-up. In the Cleveland Clinic study only one patient out of 25 (4%) with simple fatty change went on to develop cirrhosis. Thus, simple fat with no evidence of necroinflammation, ballooning degeneration, or fibrosis, appears to have an excellent prognosis particularly in younger individuals, and for at least 10 years.

Cryptogenic Cirrhosis

At the other end of the spectrum of NAFLD there is now increasing intriguing evidence that NASH may be the most common precursor of cryptogenic cirrhosis. Two studies have shown that 70% of patients with cryptogenic cirrhosis, mean age just over 60, had one or more of the following: previous obesity, NIDDM, or hyperlipidemia, and may well, therefore, have had "burnt-out" NASH. This was in contrast to around 30% of patients of comparative age with other forms of cirrhosis. [51], [52] The reasons why most patients with cryptogenic cirrhosis do not demonstrate much fat in the liver may be because of the undernutrition of cirrhosis and because of portosystemic shunting of lipids "avoiding" delivery to the liver in the portal blood. To add some support to this concept, it is now reported that following transplantation for cryptogenic cirrhosis, a proportion of individuals, most with returning obesity or with NIDDM, developed fatty liver and classic histological features of NASH (J Ong, personal communication, Nov. 2000). The clinical course of patients with cryptogenic cirrhosis associated with obesity has been compared with that of HCV-related cirrhosis. [53] In this study patients with cryptogenic cirrhosis all over age 60, severe (Child-Tureot-Pugh grade B and C) cirrhosis was as frequent in cryptogenic as in HCV patients and survival of these cryptogenic obese patients was lower than in untreated age- and sex-matched HCV cirrhotic "controls" at 30 months ($p < 0.02$). Finally, hepatocellular cancer (HCC) was detected in 8 out of 27 (27%) of these patients. A second study has confirmed the relationship between cryptogenic cirrhosis, associated with previous features of the metabolic syndrome and development of HCC. [54] Median age for development of HCC was around 70 years.

We can now see a picture of the natural history of NAFLD (Fig. 115-1) in which obese individuals or those with insulin resistance develop fatty liver. At some stage over age 40, presumably particularly in individuals with genetic susceptibility, factors promoting oxidative stress, the proinflammatory cascade, and fibrosis are brought to bear and in this unknown proportion of initially "fat-alone" patients progressive liver disease develops. It appears that in patients with absolutely no histological features of necroinflammation, ballooning degeneration, or fibrosis, the prognosis is good—presumably these individuals have "good" genes and are less susceptible to insult by oxidative stress and so forth. At the other end of the spectrum, once cirrhosis has developed a proportion of patients lose the glycogen vacuoles and are left with "burnt-out" cirrhosis, typically around age 60. A proportion of these patients subsequently go on to develop HCC—like most other cirrhotic conditions.

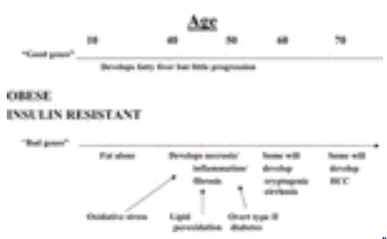


FIGURE 115-1. The fatty liver continuum.

TREATMENT

This consideration of treatment will focus on "classic" NAFLD associated with insulin resistance and obesity. Clearly, where there is an association between NASH and a rare causative factor, such as a medication, then treatment will be aimed at removing the underlying precipitating putative cause. Although the clinical course of NAFLD is variable and, at present, unpredictable, treatment should be aimed at stopping disease progression, certainly preventing development of cirrhosis, and if possible, returning hepatic architecture to normal. [55] Almost no biopsy- confirmed controlled trials of treatment for NAFLD have been reported.

We may consider treatment in two parts—reversal of the underlying metabolic defects—hence reduction of hepatic fat and reversal of insulin resistance, and intervention against such "attack" mechanisms as oxidative stress, proinflammatory cascade, and fibrosis itself.

Weight Loss and Exercise

There is probably nothing more difficult than long-term change of an individuals lifestyle; nonetheless, a number of uncontrolled pilot studies or case reports have suggested that weight reduction, restricted animal fat diet, and exercise do improve or normalize LFTs in patients with NASH. [56], [57] A number of control trials with liver biopsies are currently underway in respect of lifestyle modification.

In view of the importance of insulin resistance in genesis of fatty liver, drugs that improve insulin sensitivity are obvious candidates for use in treatments of NASH. Metformin reverses fatty liver disease in obese, leptin-deficient mice. [58] A pilot study of metformin in 20 patients with NASH showed reduction in transaminases (returning to normal values in half) and reduction in liver volume over 4 months. [59] Interestingly, the Ob/Ob grossly obese mice developed gross hepatic steatosis and steatohepatitis but not fibrosis suggesting that leptin may modulate development of fibrosis. The thiazolidinedione troglitazone was subjected to a pilot study in NASH treatment with liver biopsies before its withdrawal for other toxic side effects. Interestingly, there was little improvement in steatohepatitis. [60] Other PPAR? ligands (other thiazolidinediones) significantly increased severity of steatosis in diabetic mice. [61] Nonetheless, the use of newer thiazolidinediones may prove effective not only for their role in improving insulin resistance but because they may also reduce central obesity. [31]

Lipid lowering agents have been the subjects of pilot studies. In one of the very few control trials in which before and after biopsies were carried out, Laurin and colleagues [62] found no benefit either to LFTs or liver biopsies after 1-year treatment with clofibrate. A short pilot study of gemfibrozil showed improvement in LFTs but no biopsy evidence. [63]

Antiinflammatory/Antifibrotic

Because of its possible membrane-stabilizing and cytoprotective effects ursodeoxycholic acid (UDCA) was also subjected to a pilot, biopsy-proven, control trial evaluation. [62] There were improvements in LFTs but little real improvement was seen in liver biopsies. This treatment is now subject to a larger multicenter control trial whose results are awaited.

A variety of putative antioxidants—betaine, N-acetyl-cysteine (NAC), vitamin E, and silymarin have all been or are currently under pilot assessment (summarized in ref. [55]).

Because of the possibility that there is hypertension-associated increased angiotensin II activity leading to TGFß1 stimulation of stellate cells, and hence fibrosis in patients with NASH, a trial of angiotensin-converting enzyme inhibitors (e.g., captopril), conceivably in conjunction with one or more of the other treatments outlined previously, might be reasonable.

One can summarize by saying there is no proven treatment for NAFLD/NASH. End-stage cirrhosis is, of course, treated as for all other chronic liver disease. In individual patients outside the context of a controlled trial, gradual weight loss, return to an American Heart Association recommended dietary balance, modest regular exercise, and possibly an antioxidant would seem sensible. In those with frank NIDDM, metformin may be the treatment of choice if there are no other contraindications. In patients with known hyperlipidemia, treatment with a statin would seem reasonable. The results of control trials throughout the world are awaited eagerly.

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CHAPTER 116

Michel Mendler, John Donovan, and Andres Blei

CENTRAL NERVOUS SYSTEM AND PULMONARY COMPLICATIONS OF END-STAGE LIVER DISEASES

Portal hypertension, after a critical level of portal pressure is reached, results in portal-to-systemic shunting of splanchnic blood. Substances present in portal blood gain access to the systemic circulation as the consequence of both portosystemic shunting and an impaired metabolism by the cirrhotic liver. An imbalance of humoral mediators is implicated in the pathogenesis of complications of cirrhosis and portal hypertension. Among these pathologies are hepatic encephalopathy, hepatopulmonary syndrome, and portopulmonary hypertension. These complications of the portal hypertensive state and, as discussed elsewhere, variceal bleeding, ascites, and hepatorenal syndrome represent the major manifestations of end-stage liver disease.

Hepatic encephalopathy, hepatopulmonary syndrome, and portopulmonary hypertension can be the cause of significant morbidity and poor quality of life. In patients with cirrhosis and portal hypertension, medical therapies may often attenuate the severity of symptoms associated with each of these conditions but are not considered curative. Liver transplantation, if not otherwise contraindicated, may be indicated as the treatment of choice in the selected patient.

HEPATIC ENCEPHALOPATHY

Hepatic encephalopathy (HE) is a multifaceted neuropsychiatric syndrome observed in the setting of hepatic insufficiency or portosystemic shunting. Central nervous system (CNS) abnormalities in patients with liver failure were recognized by Hippocrates (quoted in refs. [1] and 237): “those who are mad on account of phlegm are quiet, but those on account of bile are vociferous, vicious, and do not keep quiet.” A graphic description of what is recognized today as HE was made by Frerichs in the late 19th century. [3], [4] The first assessment of HE in the era of modern medicine was made by Sherlock in 1954. [5] The onset of HE is a sign of poor prognosis both in acute or chronic liver disease. The diagnosis of HE relies on clinical indices in most instances, made in the setting of a confusional syndrome in a patient with liver failure or portosystemic shunting. *Minimal hepatic encephalopathy* (MHE) is a more recent concept of unrecognized, subtle abnormalities diagnosed by neuropsychiatric testing and identified by sophisticated imaging modalities. Multiple definitions and grading schemes for HE have been proposed, leading to confusion among clinicians when staging HE and to difficult interpretation of clinical data by researchers. In an attempt to remedy this lack of consensus, a team of experts recently elaborated guidelines for the nomenclature of HE and the performance of clinical trials. [6] The pathophysiology of HE is still incompletely understood. A clear requisite is the decreased hepatic clearance of potential neurotoxins, especially ammonia; as a result, neurotransmission abnormalities arise in the brain. Astrocyte swelling may also have an important pathogenic role. The factors involved in HE fluctuate over time, especially in the setting of cirrhosis; thus, it is common to observe precipitating factors and rapid regression of symptoms after treatment. Acute HE in the setting of acute hepatic failure (AHF), however, signals a grave prognosis requiring liver transplantation in selected cases.

Definition

HE can be defined by the presence of a wide range of neuropsychiatric disturbances secondary to either chronic liver disease (cirrhosis) or acute injury (fulminant hepatic failure [FHF]), or in a patient with either intrahepatic or extrahepatic portosystemic shunting (congenital, acquired, or iatrogenic). *Portosystemic encephalopathy* is the preferred term of some authors. HE is a diagnosis of exclusion with multiple differential diagnoses: metabolic encephalopathies, intracranial structural disorders, psychosis, Wernicke encephalopathy, drug- or toxin-induced alterations of mental status, seizure disorders, infectious diseases of the CNS. A clinical episode of HE can be overt and often easily diagnosed. HE can also be subclinical (MHE). Finally, HE can be episodic or persistent (Table 116-1).

A: Encephalopathy associated with acute liver failure.
B: Encephalopathy associated with Portosystemic bypass and no intrinsic liver disease.
C: Encephalopathy associated with cirrhosis and portal hypertension

1. *Episodic*
   a. Precipitated
   b. Spontaneous
   c. Recurrent
2. *Persistent*
   a. Mild
   b. Severe
   c. Treatment dependent
3. Minimal (formerly subclinical)

TABLE 116-1 Classification of Hepatic Encephalopathy

Pathophysiology

The pathophysiology of HE is complex and still incompletely understood despite extensive research in animal models and in humans. [7], [8], [9], [10], [11], [12] and [13] The mechanisms involve neurochemical alterations and changes in the functional status of cerebral neurotransmitter systems. Permanent structural disorders of the brain are not present. The pathogenic models developed to date attempt to explain the functional nature of HE, its reversible nature, and its precipitation by a host of clinical factors (see later). Current views support a role for multiple mechanisms participating together to different degrees in the production of neurological symptoms. The following is a summary of current hypotheses.

Role of Gut-Derived Neurotoxins The principal toxin implicated is ammonia. In the absence of liver disease, about 40% of ammonia is derived by colonic bacteria acting on lumenal contents; 60% is produced from catabolism of dietary protein, metabolism of glutamine, and deamination and transamination pathways of amino acids. Constant low levels of circulating ammonia are maintained by the production of urea (liver) and the production of glutamine from glutamate by glutamine synthetase (liver, muscle, and brain). In the brain, only astrocytes contain glutamine synthetase. In patients with cirrhosis or acute liver failure, reduction in hepatocellular function is thought to explain the observed elevations of blood ammonia. Portosystemic shunting increases ammonia levels by bypassing hepatic

metabolism. The deleterious effect of ammonia cannot be construed from the venous or arterial levels, which correlate poorly with symptoms of HE. The facilitated passage of ammonia across the blood–brain barrier (BBB) complicates the interpretation of blood levels. [14], [15] In animal models, coma stages of HE are achieved at brain ammonia concentrations of 1 to 5 mmol/L, as compared with normal blood levels of 0.05 to 0.1 mmol/L. The neurotoxicity of ammonia involves multiple mechanisms leading to the development of Alzheimer type II astrocytosis, regarded as the cardinal pathological CNS change associated with HE. These cells are recognized by their large pale nuclei, prominent nucleoli, abundant mitochondria, rough endoplasmic reticulum, and cytoplasmic vacuolation. [16] At low levels, ammonia suppresses inhibitory postsynaptic potential formation and depolarizes neurons. At high concentrations, ammonia increases the resting membrane potential and inhibits axonal conductance and excitatory postsynaptic potential formation, thereby depressing CNS function. These abnormalities involve the inhibition of cellular chloride channels by ammonia. Other mechanisms whereby ammonia may play a role in HE include a reduction in brain glucose and oxygen consumption, an increase in brain glutamine, a rise in serotonin by increased cerebral tryptophan uptake, and an alteration in glutamatergic neurotransmission [10] (Table 116-2). Oral therapies for HE, such as lactulose or antibiotics that lower gut-derived ammonia, support the ammonia hypothesis. However, the lack of a strong correlation between arterial levels and the severity of HE, along with the paradoxic excitatory effects of ammonia on the CNS in contrast to the state of neurodepression observed in HE, suggests that ammonia is not the sole player in HE. Other neurotoxins may act synergistically with ammonia. Methanethiol, a mercaptan derived by gut bacterial metabolism, may exert adverse effects on neuronal function by synergism with ammonia and short-chain fatty acids. [17] Decreased elimination of phenols derived from bacterial metabolism of tyrosine and phenylalanine in the gut may also act synergistically with ammonia.

	AMMONIUM ACETATE TREATED	
METABOLITES (LOCALIZATION)	*Precoma*	*Coma*
Ammonia (brain)	↑	↑
Ammonia (CSF)	↑	↑
Energy metabolites		
Lactate (brain)	↑	↑
Lactate (CSF)	↑	↑
α-Ketoglutarate (brain)	NC	NC
ATP	NC	↓
ADP	NC	NC
ATP/ADP ratio	NC	↓
Amino acids		
Glutamine (brain)	NC	↑
Glutamine (CSF)	↑	↑
Glutamate (brain)	NC	↓
γ-Aminobutyric acid (brain)	NC	NC
Alanine (brain)	↑	↑
Alanine (CSF)	↑	↑
Monoamine transmitters and metabolites		
Homovanillic acid (brain)	↑	↑
Dopamine turnover (brain)	↑	↑
Serotonin (brain)	↓	↓
Serotonin (extracellular)	NC	↑
5-Hydroxyindoleacetic acid (brain)	↑	↑
Serotonin turnover (brain)	↑	↑

NC, no change.

Adapted from ref. 10.

TABLE 116-2 Changes in Brain and Cerebrospinal Fluid (CSF) Metabolites in Ammonia-Precipitated Encephalopathy in Chronic Liver Disease

Role of Altered Neurotransmitters Alteration in excitatory (glutamatergic-mediated) and inhibitory (?-aminobutyric acid [GABA]-mediated) neurotransmitters may underlie the pathogenesis of HE. In recent years, the GABA-ergic neurotransmission hypothesis received considerable attention. This hypothesis involves the interplay of GABA, so-called endogenous or natural benzodiazepines (BZ), and the diazepam-binding receptor. Normally, GABA acts as the primary inhibitory CNS neurotransmitter, counterbalancing excitatory glutamatergic tone by binding to $GABA_A$ receptors, triggering increased chloride currents into the neuron and neuroinhibition by hyperpolarization of the postsynaptic membrane. In animal models of HE, visual-evoked responses typical of HE can be induced by drugs that potentiate GABA-mediated neurotransmission, drugs that normally induce seizures by decreasing GABA-ergic tone are ineffective, and $GABA_A$-receptor complex antagonists reverse electrophysiologic and neurological aspects of HE. These phenomena in HE could be explained by an up-regulation and increase in number of $GABA_A$ receptors. There is little evidence, however, supporting the role of gut-derived GABA, decreased hepatic metabolism of GABA, or increased uptake of GABA through the BBB. [18] Rather, there is evidence suggesting that natural BZ receptor agonists may be elevated in HE and are responsible for the GABA-mediated responses observed experimentally. [8], [19], [20] Many of these compounds are classic 1,4-BZs, such as diazepam and *N*-desmethlydiazepam, but many others are yet to be characterized. The source of these compounds and their relation to cirrhosis or portosystemic shunting is still unclear. Hypothetical sources include dietary sources (wheat, potatoes, soy, milk, beans, rice, and mushrooms) and bacterial conversion of BZ precursors in the gut or by the diseased liver after absorption. Indirect evidence for the role of natural BZ in HE is the reversal, albeit incomplete, of HE by BZ receptor antagonists in animal models and in humans. [21], [22] Despite supporting evidence for the natural BZ hypothesis in HE, many issues remain unresolved (Table 116-3). Finally, peripheral benzodiazepine receptors (PBRs) located in glial mitochondria, [23] distinct from $GABA_A$ receptors, may play a role in HE. An increase in cerebral ammonia is thought to increase the density of these PBRs. Activation of these receptors in turn induces the synthesis of neurosteroids (tetrahydroprogesterone and tetrahydrodeoxycorticosterone), which are potent agonists of $GABA_A$ receptors, enhancing GABA-ergic neurotransmission. [24] Indirect evidence for the involvement of PBRs in HE is the effect in animal models of pregnenolone and dehydroepiandrosterone, endogenous neurosteroid antagonists. [25], [26]

Evidence Supporting the Concept
Studies in animal models
BZ receptor antagonist (BZRA)-induced ameliorations of HE
In HE, BZRAs induce excitation of CNS neurons in vitro.
In HE, occupancy of BZ receptors by BZ agonists is demonstrated.
In HE, the BZ levels in CSF, blood, and brain tissue are increased.
Levels of BZs correlate with severity of HE.
Findings in humans
BZRA-induced ameliorations of HE
In HE, increased levels of BZs in CSF, blood, and postmortem brain tissue
Levels of BZs correlate with severity of HE.
Unresolved Issues
The source of these BZs is unknown.
The identity of many BZ receptor ligands in HE is unknown.
The properties of many BZ receptor ligands in HE are unknown.
Whether BZ levels in HE are sufficient to depress the CNS is unknown.
The precise role of BZs in the pathogenesis of HE is unclear.
Interactions between BZs and other putative toxins in HE is yet unexplored.

BZ, benzodiazepine; CSF, cerebrospinal fluid; HE, hepatic encephalopathy.

Adapted from ref. 19.

TABLE 116-3 Concept that Natural Benzodiazepines Contribute to Hepatic Encephalopathy

In normal conditions, glutamate is an excitatory neurotransmitter that counterbalances the inhibitory effect of the $GABA_A$ receptors. Regulation of brain extracellular glutamate levels involves presynaptic and postsynaptic control but is predominantly under the influence of reuptake and conversion into glutamine by perineuronal astrocytes. There is evidence suggesting that, in HE, hyperammonemia down-regulates glutamate binding sites on postsynaptic neurons and impairs glutamate reuptake, thus explaining the increase in extracellular cerebral glutamate [27] observed in HE. A different pool of glutamate, localized in astrocytes, contributes to the detoxification of ammonia through the synthesis of glutamine. This reaction is catalyzed by glutamine synthetase, solely localized to astrocytes in brain. [16] Accumulation of glutamine correlates with the degree of HE, as assessed in human cerebrospinal fluid. [28] Abnormalities in cerebral catecholamine transmitters may play a small role in HE. Noradrenaline levels are decreased in animal models of HE, and serotonin-synaptic defects may explain the high levels of cerebral serotonin metabolites (tryptophan and 5-hydroxindoleacetic acid). Serotonin is also increased in HE due to an increase in uptake of tryptophan across the BBB. Down-regulation and loss of D_2-dopamine receptors in the pallidum in HE may be the consequence of manganese accumulation and could explain the decreased magnetic resonance signal observed in this region of the brain and the extrapyramidal symptoms sometimes observed in HE. The origin of elevated manganese in cirrhotic patients may be due to both impaired biliary manganese excretion and portosystemic shunting. Finally, the “false neurotransmitter” hypothesis described by early authors is based on the observation that brain tyrosine hydroxylase is inhibited in HE. Certain amines (tyramine, octopamine, and ß-phenylethanolamine) can be synthesized from tyrosine through alternative pathways. It was hypothesized that these amines acted as weak neurotransmitters, competing with standard catecholamines. None of these hypotheses alone can fully explain the symptoms of HE. Synergism between each individual pathway most likely occurs. As shown in Table 116-2, hyperammonemia could directly produce HE but also induces a cascade of proencephalopathic mechanisms. Basile and Jones [8] recently put forth a concept uniting the ammonia and GABA-ergic concepts, which they consider as primordial in HE. They argue that ammonia can directly potentiate inhibitory GABA-ergic neurotransmission and synergistically augment the actions of natural BZ receptor agonists (Fig. 116-1). This could explain why some patients with HE have normal ammonia levels or do not respond to BZ receptor antagonists.

FIGURE 116-1. Postulated interrelationships between elevated brain concentrations of ammonia and increased GABA-ergic neurotransmission in the pathogenesis of hepatic encephalopathy. The mechanisms depicted, alone or in combination, can account for a substantial increase in GABA-ergic neurotransmission, which is a major factor contributing to the manifestations of hepatic encephalopathy. (From ref. [8].)

Modifications in Energy Metabolism Severe HE is accompanied by reductions in cerebral blood flow and in cerebral oxygen and glucose consumption. These abnormalities have been demonstrated in HE by positron emission tomography and magnetic resonance spectroscopy techniques and were shown to correlate with abnormalities of neuropsychiatric testing (see later). Rather than a cause of HE, these abnormalities appear to be secondary to severe HE.

Astrocyte Swelling Alzheimer type II astrocytes in HE are related to hyperammonemia and glutamine metabolism. [16] Additional abnormalities in astrocyte osmolarity [29], [30] have prompted a recently developed hypothesis whereby disturbances in astrocyte cell volume homeostasis are a critical event in the development of HE. [13] Magnetic resonance spectroscopy studies have shown that an astrocytic osmolyte, *myo*-inositol, is depleted in HE. This phenomenon is observed in cirrhosis, [29] experimental portacaval shunting, [31] and after transjugular intrahepatic portosystemic shunt (TIPS) [30] and is reversible, especially after liver transplantation. The extent of these changes may correlate to the severity of HE. This depletion in *myo*-inositol is a response to the increase in intracellular glutamine [32] triggered by hyperammonemia. Other organic osmolytes, such as taurine and a-glycerophosphorylcholine, exit the astrocyte to counteract astrocyte swelling. Furthermore, other phenomena known to precipitate HE and often encountered in cirrhotic patients (see later), namely hyponatremia (overdiuresis), elevated tumor necrosis factor-a (during sepsis), BZ, [32] and neurosteroids [33] can cause astrocyte swelling. This low-grade astrocyte swelling has multiple cascading consequences, including activation of extracellular regulated protein kinases, elevation of intracellular calcium concentration, up-regulation of PBR (and hence up-regulation of GABA-ergic tone), and alkalinization of endocytotic vesicles affecting receptor density and neurotransmitter processing. Finally, swelling may also induce changes in endothelial plasma membrane transporters responsible for the modifications in the BBB encountered in HE. As shown in Fig. 116-2, the authors of this novel pathogenic model suggest that this common astrocytic pathway could explain both the role of multiple factors triggering HE and the multiple abnormalities in neurotoxins and neurotransmitters described previously. The authors speculate that this model is applicable to acute HE as well, only with differences in kinetics and degree of swelling.

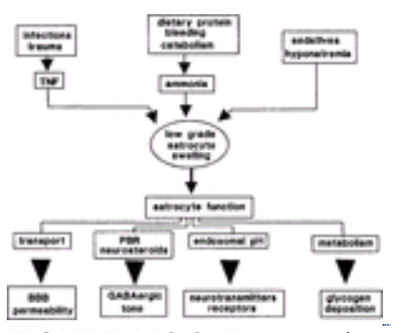

FIGURE 116-2. Proposed mechanism through which various precipitating factors can induce hepatic encephalopathy. This model views a disturbance of astrocyte hydration (low-grade cerebral edema) as a key event and one (not the only) major mechanism leading to astrocyte dysfunction and the clinical picture of hepatic encephalopathy. (From ref. [13].)

Mechanisms Involved in Fulminant Hepatic Failure The mechanisms described previously for chronic HE apply as well to the sudden onset of HE observed in the setting of FHF. Cerebral ammonia levels are generally very high, in excess of 4 mmol/L, [34] and correspond to high glutamine levels; abnormalities in the BBB may also play a pivotal role. The principal distinguishing factor of HE in FHF, however, is massive astrocyte swelling and cerebral edema. The causes of cerebral edema are twofold [35]: massive astrocyte glutamine uptake in response to hyperammonemia, [36] and cerebral vasodilation. The latter could be the result of systemically related cerebral vasodilation (through gut-derived endotoxins, proinflammatory cytokines, and stimulation of nitric oxide synthase) and locally induced cerebral hyperemia (through the release of lactate in response to hypoxia and the release of prostaglandins by the cerebral endothelium).

Clinical Aspects

Overt Hepatic Encephalopathy: Signs and Symptoms Overt hepatic encephalopathy (OHE) can present with a wide variety of signs and symptoms. [4], [37], [38], [39] and [40] The most typical are changes in consciousness, intellectual function, and behavior as well as neuromuscular abnormalities. Onset is most often rapid, and clinical features can worsen or improve rapidly within hours or days. Impaired consciousness can range from subtle changes in personality and inversion of the sleep-wake cycle to lethargy, stupor, and coma. Personality changes can be subtle, only apparent to the patient's friends and family. Abnormalities in intellectual function include unusual behavior; disorientation to time, place, or person; impaired antegrade or retrograde memory; and confusion. The most prominent neuromuscular abnormality is flapping tremor, a form of asterixis. Asterixis is defined as an inability to maintain posture actively due to deregulation of agonist and antagonist muscle tone. Flapping tremor is demonstrated by the development of rapid involuntary flexion and extension movements of the wrist when the patient is instructed to maintain wrists dorsiflexed, fingers spread, and arms outstretched anterior to a horizontal plane. This position is achieved when instructing the patient to push against an imaginary wall in front of them. Flapping tremor should be differentiated from fine tremulations of the hands seen in alcohol withdrawal. Instructing the patient to close his or her eyes can increase the sensitivity of the examination. In patients who are bedridden, the arms should not be outstretched too high above the plane of the body because this leads to a drop in the arms because of fatigue and can be falsely interpreted as flapping tremor. Other signs of asterixis are abrupt loss of tone during tongue protrusion, pedal dorsiflexion, and fist clenching. Asterixis is not demonstrable in the patient who is comatose. At this stage, abnormal ocular movements, a diffuse pyramidal syndrome, or even decerebrate posturing may be observed. Other neuromuscular abnormalities of OHE include ataxia, hypomimia, dysarthria, bradykinesia, and choreo- athetotic movements. Whether partial or generalized seizures are part of the clinical spectrum of OHE is debated. [41] Most cases are due to alcohol withdrawal or an underlying metabolic abnormality (hyponatremia) or are drug-induced. Typically, seizures due to OHE are observed in the acute setting of fulminant liver failure when cerebral edema is involved. One or more of these findings in a patient with chronic or acute liver failure or portosystemic shunting is diagnostic for OHE, after excluding differential diagnoses. In this clinical setting, there is seldom need to proceed with special blood tests, brain imaging, or neuropsychological testing to confirm the diagnosis. Foetor hepaticus can be detected in liver failure and portosystemic shunting and can be observed in patients with or without OHE. A sweet, musty breath reminiscent of rum, thought to be produced by volatile mercaptans and sulfides derived from methionine, characterizes foetor hepaticus. The pungent odor can be potent, easily detected when entering the patient's immediate environment; in other instances, it may be subtle and require that the patient exhale deeply in proximity to the examiner to be recognized. Foetor hepaticus can be associated with a peculiar body odor, known as fish-odor syndrome, due to impaired metabolism of trimethylamine. [42] Multiple schemes have been proposed to grade OHE. Grading of OHE is useful for clinicians to assess objectively patient status; it is an absolute necessity in clinical research to appreciate the effects of treatments and the diagnostic value of special tests, or in mechanistic studies. By far the most widely used and accepted system is the West Haven criteria of altered mental state in HE [43] (Table 116-4). This is a relatively simple semiquantitative 0 to IV point scale using readily identifiable clinical signs. This system does not give criteria for the diagnosis of stage 0 (MHE) or of stage IV (coma). Stage IV is best assessed by the Glasgow coma scale [44] (Table 116-5). The Portosystemic Encephalopathy Index [45] (PSEI) has been used in many clinical trials. This additive grading system is the ratio (0 to 1) of the sum of scores for mental state, asterixis, number connection test, [46] plasma ammonia levels, and electroencephalogram (EEG) grade over a total possible 28 points. The PSEI is not superior to the West Haven criteria in assessing OHE and suffers from the use of arbitrary unit multiplicators and the inclusion of plasma ammonia levels that do not correlate to symptoms of HE. Another system proposed by Gitlin [47], [48] combines the clinical features of the West Haven criteria with EEG features but is not widely used. Both the PSEI and Gitlin grading systems, therefore, incorporate testing for MHE. The simplest grading system is that which is included in the Child-Pugh score for severity of cirrhosis [49], [50]: absent (0), moderate or responsive to treatment (2), or severe or unresponsive to treatment (3). This three-point scale, however, is far too ambiguous and subjective. In 1998, a working party of experts in HE came to the following consensus regarding the grading of OHE [6]: only clinical grading should be used for quantification of HE; the West Haven criteria (I to IV) should be used for changes in consciousness, intellectual function, and behavior; the Glasgow coma scale can be used in addition for patients in stages III and IV.

Stage 0:	Lack of detectable changes in personality or behavior. Asterixis absent.
Stage 1:	Trivial lack of awareness. Shortened span. Impaired addition or subtraction. Hypersomnia, insomnia, or inversion of sleep pattern. Euphoria or depression. Asterixis can be detected.
Stage 2:	Lethargy or apathy. Disorientation. Inappropriate behavior. Slurred speech. Obvious asterixis.
Stage 3:	Gross disorientation. Bizarre behavior. Semistupor to stupor. Asterixis generally absent.
Stage 4:	Coma.

From ref. 43.

TABLE 116-4 West Haven Criteria for Altered Mental State in Hepatic Encephalopathy

EYES OPEN		BEST MOTOR RESPONSE		BEST VERBAL RESPONSE	
Spontaneously	4	Obeys verbal orders	6	Oriented, conversant	5
To command	3	Localizes painful stimuli	5	Disoriented, conversant	4
To pain	2	Painful stimulus, flexion	3	Inappropriate words	3
No response	1	Painful stimulus, extension	2	Inappropriate sounds	2
		No response	1	No response	1

*The best ocular, verbal and motor responses are summed. The best score is 15, the worst is 3. Severe hepatic encephalopathy is defined by a score of less than 12.

From ref. 44.

TABLE 116-5 Glasgow Coma Scale*

Minimal Hepatic Encephalopathy: Definition and Diagnosis The term *subclinical hepatic encephalopathy* (SHE) was coined in the 1970s when researchers determined that patients with cirrhosis who did not have OHE performed poorly in neuropsychiatric and electrophysiologic testing. [2], [4], [6], [40], [51], [52], [53], [54], [55] and [56] There has since been much debate and research on which tests best describe this condition, whether or not it is relevant in affecting patient prognosis or function, and whether SHE represents either HE at a stage at which it is not detectable by clinical observation or a pathologically distinct entity from HE. More recent research has provided conclusive evidence that SHE is clinically relevant and most likely an early expression of the clinical spectrum that constitutes HE. The experts in the field of HE now prefer the term *minimal hepatic encephalopathy*, or MHE, to subclinical encephalopathy. [6], [57] Dozens of neuropsychological tests have been used to characterize or have been proposed as methods to diagnose MHE. [2], [58] The choice of which constitutes the best test or tests is challenging. Analysis of performance is hampered by inconsistencies in use of controls or appropriate controls; lack of correction for the influence of age, gender, social status, education, and cultural background; data from small patient groups; and lack of validation. Certain tests are time consuming or require professional expertise in neurology or psychiatry that may not be attainable by an internist or gastroenterologist-hepatologist; and certain batteries of tests are more suitable for research purposes than for routine testing. The current consensus for clinical trials [2], [6] is to perform at least two of the following standardized tests: the number connection test (NCT) A and B, digit-symbol test (DST), or block-design test (BDT). There still is no consensus as to the most accurate tests to use in routine clinical practice. Conn [46] and Zeegen and colleagues [55] were the first to use a modified Reitan trail-making test, which became the NCT and the most widely used paper-and-pencil neuropsychological test for MHE (Fig. 116-3). The NCT tests cognitive dysfunction. In the NCT-A, the subject must connect in the correct order as quickly as possible a sequence of scattered circles numbered 1 through 25. The score is the time required for the subject to complete the trail, including time to correct errors. A low score indicates a good performance. The concept of the NCT-B is similar, except for an alternating sequence of scattered numbered and lettered circles, 1-A through 13-L. The subject must be given the opportunity to practice either test before doing a timed run, thereby decreasing training bias in follow-up testing. With a normal upper time limit of 30 seconds [46] for NCT-A and 100 seconds for NCT-B, the sensitivity and specificity in detecting SHE are estimated to be 56% and 100%, respectively, for the NCT-A and 68% and 99.2%, respectively, for the NCT-B. [2] The accuracy of the tests improves when taking into account age and educational level and when only considering results above 2 standard deviations (SD) from normal populations as being pathological: the sensitivity of NCT-A increases to 77% and that of NCT-B to 87%. [59] It has been clearly demonstrated that using an NCT uncorrected for age as a screen significantly overdiagnoses MHE. [60] The threshold for normal scores is further modified by the version of the test used. [45]

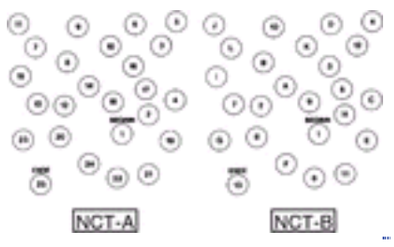

FIGURE 116-3. An example of number connection tests (*NCT*) A and B used to assess minimal hepatic encephalopathy. (From ref. [59].)

The DST is a subset of the Wechsler Adult Intelligence Scale [61] and measures motor speed and accuracy. The subject is given a list of nine line symbols (- =) associated with digits from 1 to 9. A "stimulus page" consisting in series of four rows of 20 blanks associated with random digits from 1 to 9 must be filled with the symbols that correspond to each digit. The test score is the total number of correct sequential matchings of digits to symbols in a 90-second interval. A high score indicates a good performance. Little data are available on the accuracy of this test. When considering scores 2 SD below those of normal populations, the sensitivity is reported to be 90%, with 95.8% specificity. [2] The BDT is not a paper-and-pencil test. The subject is given 1-inch-square red and white cubes with which patterns from either demonstration blocks or pictures must be produced with either four or nine blocks. It is recommended that the BDT be administered as part of a battery with the NCT and DST. [47], [62], [63] In fact, the best accuracy is obtained with a battery of paper-and-pencil–only tests coined the Psychometric Hepatic Encephalopathy Score (PHES) [2] or Portosystemic Encephalopathy Syndrome Test, [6] consisting of the NCT-A and -B, the DST, and two other less frequently used tests: the Line Drawing Test [62] and the Serial Dotting Test. [58] With this combination, diagnostic sensitivity for MHE is 96% and specificity is 100%. [2] Although the proponents of such a system state that it can be carried out effectively in 10 to 20 minutes, it is clear that psychometric tests in general must be administered methodically, in a quiet room with sufficient light, with the subject in a sitting position and that they require careful interpretation and adaptation to age, education, and learning effect. In an effort to overcome these limitations, Groeneweg and associates [64] studied correlations among clinical and laboratory parameters, quality-of-life assessment, the NCT and DST, and EEG assessment in patients with cirrhosis. Variables independently associated with MHE were male gender, a Child-Pugh score of more than 7, the presence of esophageal varices, and an affirmative response to 5 specific statements of 36 from the Sickness Impact Profile questionnaire. [65] For a given patient, the sum of each relative risk (total ranging from 0 to 14.7) establishes the SHE Probability Score, corresponding to a sigmoid curve of MHE probability (Fig. 116-4). A threshold score of 3.3 yields a sensitivity of 90% and a specificity of 77% in diagnosing MHE. The authors suggest that this score could be easily performed at the bedside or in an ambulatory setting to select only those patients with a score of 3.3 or higher for further psychometric or neurophysiologic testing to confirm the diagnosis of MHE. An obvious source for the low specificity is that impairment in quality of life in patients with cirrhosis may be due to factors other than HE. [66] The SHE Probability Score merits further validation in patients with liver disease, with and without cirrhosis.

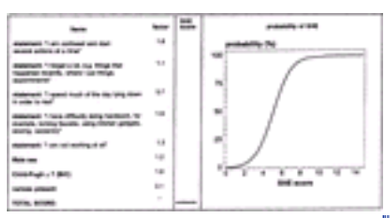

FIGURE 116-4. The subclinical hepatic encephalopathy score (SHE), proposed to screen for minimal hepatic encephalopathy (MHE). The score is the sum of points (regression coefficients) for each positive item. The probability of MHE can be derived by plotting the score on the sigmoid curve. (From ref. [64].)

The many drawbacks of neuropsychological testing have led researchers to evaluate different neurophysiologic methods to achieve an objective diagnosis of MHE. These tools include EEG, brainstem auditory-evoked potentials (BAEPs), visual-evoked potentials (VEPs), somatosensory-evoked potentials (SSEPs), P300 event-related potentials (P300), and critical flicker frequency. These methods are objective, are not biased by education or learning effects, and for the most part do not depend on age. They do, however, require sophisticated equipment and highly trained personnel to interpret results. In the opinion of the expert working party, [6] none of these tools is specific and can be used to diagnose MHE at this time. The key problem in the interpretation and comparison of these methods is the lack of consensus regarding the gold standard definition of MHE. The EEG is the neurophysiologic test most frequently used in routine clinical practice. EEG abnormalities in

patients with cirrhosis or liver failure can be due to multiple causes, including HE, electrolyte abnormalities, infection, alcohol withdrawal, seizure disorder, cerebral edema, cerebral ischemia, central pontine myelinolysis, and neurodegenerative processes. [41], [67] This explains in part why the reported frequency of abnormal EEG findings among cirrhotic patients without HE is 8% to 35% [68] and that in MHE there is no overlap between EEG and abnormal psychometric tests. [60], [69] On the other hand, the test may explore different aspects of HE. EEG patterns typical of HE are not epileptiform, but rather a continuous slowing of background rhythm and presence of atypical triphasic (theta) waves. [70] Conventional EEG analysis is inadequate to localize these patterns objectively in a reproducible fashion; quantitative spectral EEG analysis is the current standard test. [69], [71], [72] and [73] Evoked potentials are electrical signals generated by excitable regions of the brain subsequent to external stimulation. Different stimuli induce evoked potentials in different regions of the brain. Evoked potentials can describe abnormal sensory function qualitatively and quantitatively in the absence of overt clinical manifestations. They are routinely used to characterize multiple sclerosis and toxic-metabolic disorders, the extent and topography of brain disorders, and CNS dysfunction in comatose patients. BAEP, VEP, and SSEP testing use the exogenous components of evoked potentials and are influenced by the physical properties of the mode of stimulus. P300 evoked potentials are endogenous event-related cerebral potentials, generated in response to a change in visual or acoustical stimulus ("target stimulus"). It is thought that the P300 response correlates to cognitive processes and thus detects the impaired cerebral function of MHE demonstrated by psychometric testing. BAEP abnormalities correlate poorly with MHE in general [74], [75] and [76]; they are present in grade IV HE in nonalcoholic cirrhosis and may reflect the accumulation of toxic substances and portosystemic shunting. BAEP abnormalities are observed in earlier stages of HE in alcoholic liver disease and even in the absence of cirrhosis, possibly owing to ethanol-induced demyelination. Both flashlight-stimuli VEPs and pattern-reversal mode VEPs have been studied in HE; extremely heterogeneous results have been reported with poor sensitivity in detecting both OHE and MHE. SSEP [77], [78], [79] and [80] appears to be sensitive in detecting MHE but not specific; abnormalities can be detected in alcoholic cirrhotic patients without HE, possibly in relation to peripheral neuropathy. P300 peak event-related auditory and visual potentials have been studied in alcoholic and nonalcoholic cirrhotic patients with control populations and correlations to psychometric testing. [69], [81], [82], [83] and [84] P300 testing is limited to HE grades 0 to II because it requires cooperation of the patient and appears to be extremely sensitive in MHE; however, its sensitivity is decreased by recent alcohol abuse. In the opinion of the expert working party, [30] P300 testing holds the greatest promise of these EP tests in detecting and characterizing MHE. Kircheis and colleagues [85] recently presented the use of critical flicker frequency (CFF) in HE. CFF analysis is thought to reflect the efficiency of the visual apparatus and the functional competence of the cerebral cortex and has been used extensively to evaluate multiple sclerosis, Alzheimer's disease, and the effects of psychoactive drugs. Kircheis and colleagues [86], [87] hypothesized that CFF abnormalities in HE reflect astrocyte swelling incriminated in hepatic retinopathy. Their well-designed study included a large number of alcoholic and nonalcoholic cirrhotic subjects, an adequate control population, and correlations to an extensive battery of psychometric tests. The authors determined a threshold flicker frequency capable of diagnosing MHE with 100% specificity; however, sensitivity was low (55%). These data will most likely spark new investigations into the usefulness of this technique.

Imaging Modalities Routine imaging modalities of the brain, such as computed (CT) scanning and magnetic resonance imaging (MRI), are of little utility in making a positive diagnosis of HE, but rather are used to exclude differential diagnoses. [4], [6], [88], [89] Such diagnoses would include intracerebral infarction, hemorrhage, abscess or tumor, intraventricular hemorrhage, normal-pressure hydrocephalus, central pontine myelinolysis, and cortical atrophy. In most of these instances, the clinical presentation is not typical of HE and points to a localized CNS defect eventually in association with a risk factor such as head injury, coronary heart disease, sepsis, or rapid changes in serum sodium. Systematic CT screening for intracranial abnormalities in patients with suspected HE and typical risk factors but without any signs of CNS localization is not warranted. In typical HE, CT scan in nonalcoholic patients is typically normal, and CT frequently shows cortical atrophy in alcoholic patients. In fulminant hepatitis, CT is a quasi-obligatory procedure to rule out an unexpected intracranial process that may contraindicate liver transplantation; otherwise, it might reveal cerebral edema related to acute HE. CT cannot identify structural abnormalities that correlate to MHE. MRI findings in HE include symmetric T1 hyperintensities in the pallidum, nigral substance, and the dentate cerebellar nucleus, possibly due to accumulation of manganese. [90], [91] These findings may become more prominent as liver function worsens and tend to regress after transplantation [92]; nevertheless, they do not correlate to the degree of neurological dysfunction, [93] and such isolated findings in a cirrhotic patient cannot be used alone to diagnose MHE or to correlate overt abnormal consciousness to HE. In recent years, research has intensified in the use of magnetic resonance spectroscopy (MRS) to explore various features of HE. The same radiofrequency signals emitted by nuclear magnetic resonance–sensitive nuclei (^{1}H, ^{31}P, ^{13}C) can be either transformed into an image (MRI) or into a spectrum of frequencies (MRS) corresponding to chemical entities. To date, only ^{1}H-MRS (proton MRS) has shown promising results by demonstrating associations between HE and pathophysiologic mechanisms involving osmolytes in astrocyte swelling and cerebral ammonia detoxification. The most consistent findings are decreased levels of *myo*-inositol and increased levels of glutamine in the brain. [29], [30], [92], [94] Whereas the degree of variation in these neurometabolites does appear to correlate with the severity of OHE and response to treatment, they are not sensitive enough to detect MHE in asymptomatic cirrhotic patients. [95] ^{31}P-MRS has been used to characterize altered brain energy metabolism in HE with notable abnormalities in brain phosphocreatinine, inorganic phosphate, and adenosine triphosphate suggestive of decreased glucose use and modifications in the BBB [96], [97] and [98]; however, data on correlations with disease severity and the diagnosis of MHE are conflicting. The use of MRS in HE is still in its infancy and is reserved for clinical research; although this technology appears promising, it requires specialized equipment and highly trained personnel to operate the equipment. It also remains very time consuming and therefore is not suitable for routine clinical practice. Single photon and positron emission tomography have been used to study features of mechanisms of HE involving cerebral blood flow and metabolic rate, behavior of central BZs and GABA receptors and response to treatment, and cerebral ammonia metabolism. [15], [99], [100], [101], [102] and [103] This technology has not been studied in terms of its ability to diagnose or grade HE.

Biochemical Markers In the setting of OHE, routine blood biochemistry completes the clinical presentation of underlying liver disease in the form of suspected cirrhosis or hepatic failure, showing elements in favor of decreased hepatic synthesis (decreased serum albumin, prolonged prothrombin time) and portal hypertension (thrombocytopenia). In patients with HE due to portosystemic shunting alone, such as those with portal thrombosis without cirrhosis, routine blood work may be normal. Abnormalities associated with a precipitating factor can be observed, such as hypokalemic metabolic alkalosis secondary to diuretic treatment, elevated blood urea nitrogen relative to creatinine secondary to a gastrointestinal hemorrhage, and leukocytosis secondary to sepsis. Routine blood work may also aid in making a differential diagnosis of HE, such as uremia, hypoglycemia, or hypercapnia. In line with the hypothesis of cerebral ammonia in the pathophysiology of HE, venous and arterial levels of ammonia have been studied as a marker of HE. Venous ammonia levels are significantly lower than arterial ammonia to which the brain is exposed, [36], [104] owing principally to ammonia metabolism by muscle. [105] Both venous and arterial ammonia levels vary from fasting to feeding states and are affected by the hypercatabolic state, with negative nitrogen balance observed in cirrhotic patients with wasting. [106] Finally, correct arterial ammonia determination requires that specimens be placed on ice and analyzed within 20 minutes of blood draw. [107], [108] These limitations may explain the conflicting results reported for the diagnostic accuracy of peripheral ammonia and HE. Although arterial ammonia levels are increased in up to 90% of patients with HE and decrease with clinical recovery and increase with severity of liver failure, clinical manifestations may lag by 24 hours, and there is a poor correlation between the severity of HE and ammonia levels. [109] Earlobe capillary ammonia correlates closely with arterial ammonia and may be an alternative to facilitate routine testing but requires validation. [110] Partial pressure of arterial ammonia (pNH_3), which depends on arterial ammonia and pH, is thought to better represent the unionized ammonia that crosses the BBB and has been shown in one study to correlate better with the clinical stages of HE than do arterial ammonia levels. [111] Despite the limited accuracy of venous ammonia levels in diagnosing HE, they continue to be used in the setting of emergency medicine to screen for HE in patients with unexplained abnormal consciousness without evidence of underlying liver disease.

Hepatic Encephalopathy of Acute or Fulminant Hepatic Failure HE associated with AHF is characteristically rapid in onset and in progression toward coma. In a previously healthy patient with AHF, the onset of HE within 8 weeks of onset of symptoms of liver disease defines FHF and gravely compromises prognosis. This rapid progression without a precipitating factor (other than liver failure itself) and the absence of reversibility by medical treatments (see later) differentiate HE of FHF from that of cirrhosis or portosystemic shunting. The pathophysiologic mechanisms of FHF HE involve high levels of cerebral ammonia that cross the BBB, leading to the synthesis of glutamine and the accumulation of osmolytes, which result in astrocyte swelling, cerebral edema, and ultimately death by cerebral herniation. [36], [112] The key to management is the early detection of MHE by the methods discussed previously. In the presence of OHE, high arterial ammonia values above 200 µmol/L may be of value in predicting fatal intracranial hypertension. [112] Cerebral edema can be detected by CT scan and intracranial pressure monitored by epidural transducers. Medical treatment is based on the same principles as HE of cirrhosis (discussed later), in addition to therapies specifically aimed at decreasing cerebral edema and intracranial pressure, pending liver transplantation (if not contraindicated).

Hepatic Encephalopathy without Liver Disease HE without underlying liver failure is observed in the setting of either spontaneous or iatrogenic extrahepatic portosystemic shunt. In a patient with abnormal consciousness and no known underlying spontaneous portosystemic shunt, the diagnosis of HE can be quite difficult. Clues to HE are the absence of common causes of abnormal consciousness (see later) and signs of portal (sometimes segmental) hypertension, such as abdominal or lower-back superficial collaterals, splenomegaly, esophageal or gastric varices, and ascites. Etiologies of spontaneous extrahepatic portosystemic shunt include congenital vascular malformations, which are rare, and the development of shunts secondary to partial occlusion of the portal system at the extrahepatic or intrahepatic level. Thrombosis or occlusion of the portal vein or its branches can occur in the setting of previous abdominal trauma, a primary or secondary hypercoagulable state (carcinoma, myeloproliferative disease, circulating lupus anticoagulant or antiphospholipid antibody, factor V Leiden or prothrombin mutations, or a mass effect or invasion to the porta hepatis, such as metastasis, hepatocellular carcinoma cholangiocarcinoma, or lymphoma). MHE is not a consideration in this setting because it is OHE that will alert the physician. HE is a frequent complication of portosystemic shunt surgery in cirrhotic patients, estimated at a rate of 30% to 80%, depending on type and diameter of shunt. [113] Lower rates of HE are obtained with distal splenorenal shunts and small side-to-side portacaval shunts, which preserve some degree of prograde portal flow. [114], [115] In addition to high rates of HE, surgical shunts can result in high mortality rates in the setting of liver failure and have been largely (although not completely) replaced by the TIPS procedure. [116] Initially used to treat uncontrolled esophageal bleeding in cirrhotic patients as a bridge to liver transplantation, indications for TIPS now include refractory ascites, hepatohydrothorax, Budd-Chiari syndrome, and possibly hepatorenal syndrome, irrespective of ensuing liver transplantation. As in side-to-side surgical shunts, TIPS is plagued by an increased rate of postprocedure HE, estimated initially at a rate

of 3% to 80%. Improvements in techniques (smaller-diameter stents) and a better understanding of predictive factors of post-TIPS mortality have decreased the rate of post-TIPS HE to about 30%. The mechanisms of post-TIPS HE are twofold: circumvention of hepatic detoxification by the shunt and aggravation of hepatic insufficiency secondary to decreased portal flow. Predictive factors of new-onset or worsening post-TIPS HE include pre-TIPS HE, nonalcoholic cirrhosis, female gender, age, hypoalbuminemia, stent diameter larger than 10 mm, or a decrease in portacaval gradient of less than 10 mm Hg. [117], [118], [119], [120] and [121] More recently, it has been shown (as could be expected) that post-TIPS HE can persist in the form of MHE and that pre-TIPS MHE can progress to OHE after the procedure. [122], [123] Despite the relatively high rate of post-TIPS HE, most authors describe rapid onset within 1 month of the procedure and a gradual decay of symptoms after 3 months, presumably owing to gradual narrowing of the stent by endothialization. Furthermore, when carefully selecting recipients, debilitating persistent post-TIPS HE is infrequently observed, and symptoms usually respond to standard medical therapy. Recently, stent-narrowing techniques have been developed [120], [124] that may alleviate refractory cases.

Precipitating Factors In a patient with cirrhosis or portosystemic shunting, HE can develop spontaneously and persist without treatment. Spontaneous HE is typically observed in acute liver failure or in large-bore portosystemic shunts that may be either iatrogenic (surgical shunt and TIPS) or spontaneous. [39], [48] Lam and colleagues [125] observed that patients with large-bore spontaneous shunting (demonstrated by collateral circulation emptying into the inferior vena cava) have a significantly higher incidence of HE and spontaneous HE than do cirrhotic patients with small spontaneous shunts or with Cruveilhier-Baumgarten murmurs. In most instances, however, symptoms of HE are precipitated by a number of factors that exacerbate MHE. The treatment or correction of these factors may suffice in some instances to help resolve the episode of HE. The most frequent precipitating factors involve a sharp increase in arterial blood ammonia. Digestive sources leading to increased ammonia levels include excess dietary protein, severe constipation, and gastrointestinal hemorrhage. Any bacterial infection (in particular, spontaneous bacterial peritonitis) or massive blood transfusions (hemolysis) can exacerbate ammonia levels as well as azotemia and hypokalemia. Systemic alkalosis can increase the diffusion of ammonia across the BBB. Multiple phenomena can trigger HE by inducing transient hepatic anoxia resulting in reduced hepatic metabolism of toxins. These include dehydration, hemodynamic shock, severe anemia, and hypoxemia. Circumstances typically observed in cirrhotic patients are overuse of diuretics, fluid restriction and paracentesis, severe gastrointestinal bleed, and rarely, overuse of laxatives. All classes of psychoactive drugs, even at minimal doses, can precipitate HE by amplification of their CNS depressant effect owing to impaired hepatic drug metabolism. BZs are notorious for putting patients into encephalopathic coma by activating central GABA-BZ receptors. Finally, rapidly progressive or acute decrease in hepatic functional reserve can precipitate HE. The TIPS procedure not only produces an intended portosystemic shunt but also leads to decreased hepatic perfusion. Hepatocellular carcinoma can cause HE by replacement of functional hepatocytes, and this effect can be compounded by portal vein thrombosis due to encroachment or reduction of viable hepatocytes by transarterial hepatic embolization or surgical resection. Frequently, multiple mechanisms are involved in precipitating HE. Diuretics, the mainstay of ascites therapy, can cause hypokalemia, azotemia, and dehydration. Gastrointestinal bleeding from ruptured esophageal varices leads to decreased hepatic perfusion and increased gastrointestinal protein content. When assessing a patient with HE, these precipitating factors must be actively sought and corrected; the risk for iatrogenic precipitation of HE should be considered. Diuretics should be decreased in dose or withheld, especially if azotemia or hypokalemia is present. Family members should be questioned about a recent high-protein meal. Sepsis should be contained rapidly. Gastrointestinal hemorrhage should be controlled by standard therapies and bowel cleansing initiated even before the onset of HE. Coma after the use of psychoactive drugs can be due to either their sedative effects or to HE; these drugs should be avoided whenever possible, and in some instances, an antidote may be useful. When performing tolerable invasive procedures such as gastrointestinal endoscopy, minimal doses of sedative drugs (such as the commonly used midazolam) should be administered. [126] TIPS should not be performed in a patient who has a significant history of HE.

Prognosis In the setting of cirrhosis, HE completes the clinical and biochemical picture of liver failure and therefore in itself is a sign of poor prognosis. The Child-Pugh score recognizes this feature and includes stages of HE in the empiric scoring system to grade the severity of cirrhosis. [49], [50] The survival rate after a first episode of OHE in patients with cirrhosis, irrespective of Child-Pugh score or etiology, is about 40% at 1 year and 20% at 3 years. [127] In patients with HE, factors independently associated with poor outcome are male gender, elevated alkaline phosphatase, bilirubin, blood urea nitrogen and potassium, and hypoalbuminemia. Interestingly, neither the severity of the initial encephalopathic episode, the precipitating factor, nor the presence of an iatrogenic portosystemic shunt significantly modifies survival. Estimates of OHE in cirrhotic patients vary widely from 20% to 80% of cohorts and those of MHE from 30% to 84%, depending on the tests used. It was initially thought that MHE had little impact on daily life and represented an esoteric diagnostic entity. Sophisticated studies show conclusively that MHE significantly impairs daily functioning. As compared with healthy controls and cirrhotic patients without MHE, patients with SHE are significantly impaired when driving a car or performing at work [128] and have higher SIP [65] scores than control populations. [129] Furthermore, the onset of MHE may be predictive of a high risk for ensuing OHE. HE associated with FHF is of very poor prognosis, especially if HE develops rapidly after the onset of jaundice and is rapidly severe and prolonged. Unrelenting intracranial hypertension and loss of the oculovestibular reflex are associated with fatal outcome. [130]

Differential Diagnosis The differential diagnosis of HE is wide, depending on the clinical circumstances [4], [48] (Table 116-6). In a patient with known cirrhosis, with or without spontaneous or iatrogenic portacaval shunting, the onset of abnormal consciousness concomitant to a recognized precipitating factor of HE usually leaves little doubt as to the positive diagnosis of HE. In an alcoholic patient, a withdrawal syndrome should be considered if there is a history of recent alcohol cessation, the patient has hand tremors rather than asterixis, and auditory or visual hallucinations are present. Korsakoff psychosis is gradual and persistent and involves characteristic antegrade amnesia. Wernicke encephalopathy, occurring in the setting of thiamine deficiency and intravenous glucose infusion, can have a rapid onset but will not respond to usual therapy for HE. Non–alcohol-related differential diagnoses of HE in the setting of cirrhosis include: cerebral vascular accidents and intracranial hemorrhage secondary to head trauma, typically recognized by a compatible history and a localizing motor or sensory defect and diagnosed by brain CT scan; central pontine myelinolysis observed in the setting of an iatrogenic correction of hyponatremia with an increase in natremia of more than 10 mmol/L per 24 hours diagnosed by brain MRI; or rare instances of intracranial infection.

Metabolic encephalopathies
Hypoglycemia
Electrolyte imbalance
Hypoxia
Carbon dioxide narcosis
Azotemia
Ketoacidosis
Toxic encephalopathies
Alcohol
Acute intoxication
Withdrawal syndrome
Wernicke-Korsakoff syndrome
Psychoactive drugs
Salicylates
Heavy metals
Intracranial lesions
Subarachnoid, subdural, or intracerebral hemorrhage
Cerebral infarction
Cerebral tumor
Cerebral abscess
Meningitis
Encephalitis
Epilepsy or postseizure encephalopathy
Neuropsychiatric disorders

Adapted from ref. 48.

TABLE 116-6 Differential Diagnoses of Hepatic Encephalopathy

The onset of abnormal consciousness in a patient with acute liver failure should be considered as HE unless proved otherwise. CT scan of the brain can show cerebral edema and rarely shows spontaneous intracranial hemorrhage due to a hypocoagulative state. Gross metabolic abnormalities observed in the setting of FHF such as hypoglycemia and azotemia can also cause abnormal consciousness mimicking HE. Rare degenerative disorders of the CNS should be considered when neurological symptoms are atypical of HE and do not respond to standard treatment. Hepatocerebral degeneration and transverse myelitis can occur in protracted cirrhosis and portosystemic shunting. Histological changes include hyperplasia of the protoplasmic astrocytes and neuronal loss; however, the mechanisms involved and the role of hepatic failure or portosystemic shunting are not well understood. [131] Hepatocerebral degeneration can present with progressive signs of chronic cerebellar and basal ganglia involvement, signs of parkinsonism, focal cerebral symptoms, epilepsy, and dementia. Alper disease, described as a degeneration of the cerebral gray matter in infancy, is an inherited autosomal recessive disorder and has been observed in association with cryptogenic cirrhosis or subacute hepatitis with steatosis. [132], [133] and [134] Transverse myelitis can lead to paraplegia. Two cases have been described in the setting of primary biliary cirrhosis and Sjögren syndrome. [135], [136] Wilson disease is an inherited autosomal recessive disorder. It results in excessive accumulation of copper in various organ systems, particularly in the liver and brain, leading to hepatolenticular degeneration. The signs of hepatic dysfunction (FHF, chronic hepatitis, cirrhosis) tend to occur before those of neurological dysfunction; however, neurological Wilson disease can occur without signs of liver disease. Neurological signs are initially subtle and become chronic and progressive if the disease process is unrecognized or left untreated. The neuropsychiatric signs are numerous and varied and can be classified into three distinct subsets according to lesions of the CNS [137]: pseudoparkinsonian (dilation of the third ventricle leading to bradykinesia, rigidity, cognitive impairment, organic mood syndrome); pseudosclerosis (focal thalamic lesions leading to ataxia, tremor, reduced functional capacity); and dyskinesia (focal abnormalities of the putamen and globus pallidus leading to dys- kinesia, dysarthria, organic personality syndrome).

Diagnostic Approach The approach to the diagnosis of HE depends for the most part on the clinical setting. In patients with cirrhosis or portosystemic shunting, stereotypical abnormalities of OHE are sufficient to make the diagnosis, especially when symptoms follow a precipitating factor. Neuropsychometric or electrophysiologic testing in this setting is redundant, but differential diagnoses such as severe hypoxia, hypoglycemia, azotemia, and hyponatremia should be

excluded. Systematic CT scan of the brain is not justified and should only be performed if CNS defects are suspected on the clinical exam. Symptoms of OHE usually respond rapidly to the usual oral therapies, confirming the diagnosis. Organic brain syndromes usually have a clinical presentation that is not typical of OHE and do not respond to these therapies. When OHE is absent, patients should be screened for MHE as described earlier given the prognostic implications. The diagnosis of OHE in a patient with abnormal consciousness but unknown liver disease requires the demonstration of clinical or biochemical signs of acute or chronic liver failure and the systematic exclusion of differential diagnoses. The extent to which patients should be evaluated for differential diagnoses (see Table 116-6) depends of course on the clinical presentation. In this setting, increased arterial blood ammonia may be suggestive of HE but is not 100% sensitive or specific (may be elevated in azotemia).

Current Nomenclature

Surveys performed by the Organisation Mondiale de Gastroentérologie in 1998 showed that there is considerable discrepancy among specialists on terms used to define the different clinical settings of HE. [6] The term *portosystemic encephalopathy* is used for HE in cirrhotic patients and in patients with portosystemic shunting without liver disease. *Acute encephalopathy* is interpreted as either HE of AHF or a reversible episode in patients with cirrhosis. *Chronic encephalopathy* is used to describe either recurrent episodes or continuous HE. The working party of experts has elaborated a multiaxial nomenclature and proposes that it be adopted by clinicians to assess patients and by researchers for clinical endpoints. This novel nomenclature describes three HE types: A, B, and C. Type A HE refers to encephalopathy associated with **a**cute liver failure. Type B HE refers to encephalopathy associated with portosystemic **b**ypass and no intrinsic hepatocellular disease. Type C HE refers to encephalopathy associated with **c**irrhosis and portal hypertension or portosystemic shunts. Type C is characterized by three subcategories and other subdivisions: episodic HE that may be precipitated, spontaneous, or recurrent; persistent HE that may be mild, severe, or treatment dependent; and MHE (Table 116-1).

Treatment of Type B or C

As described previously, in most instances, a precipitating factor of HE can be found. A rigorous search to identify such a factor must be undertaken and removed if possible. [7], [39], [48], [138]

Meanwhile, measures to decrease the nitrogenous load should be implemented. These measures include nutritional management and reduction of gut-derived ammonia. Nutritional management should include careful limitation of animal proteins and supplementation of any vitamin or mineral deficiencies, especially in zinc. In theory, dietary protein restriction can significantly reduce ammonia load; however, protracted nitrogen restriction leads to malnutrition, especially in hypercatabolic cirrhosis, thus aggravating prognosis. In fact, a positive nitrogen balance can improve HE by increasing the muscle's capacity to detoxify ammonia. The recommendations are for 1 to 1.5 g protein/kg/day, mostly in the form of vegetables and dairy products, which provide a higher calorie-to-nitrogen ratio than meat proteins. Vegetable sources have the added benefit of providing fiber that improves intestinal transit and acidifies stools, thereby reducing the formation of toxic fatty acids and ammonia from colonic amino acids. Proteins should be withheld from the diet at onset of OHE for 24 hours and reintroduced rapidly by increments of 20 g/day.

Reduction of gut-derived ammonia or other putative compounds of HE is the mainstay and first-line treatment of HE. Most of the ammonia load involved in the pathogenesis of HE enters the circulation from the intestinal tract, derived from colonic bacteria and deamination of glutamine in the small intestine. The ammonia is absorbed by passive diffusion. Other putative substances are derived from the gut: BZ-like substances or precursors, toxic short-chain fatty acids, phenols, and mercaptans. The therapeutic goal is to shorten bowel transit in order to eliminate ammonia-producing bacteria and to lower the available absorbable colonic ammonia. This can be achieved with nonabsorbable disaccharides, either lactulose or lactitol. These agents promote the incorporation of lumenal ammonia into the bacterial protein wall and promote catharsis by acidification of the colon as the result of the bacterial production of acetic and lactic acid by bacterial fermentation. This decrease in pH is hostile to urease-producing bacteria and further reduces the production of ammonia in the colonic lumen. The inconveniences of these agents are flatulence, abdominal cramping, and diarrhea. Some patients cannot tolerate the excessively sweet taste of lactulose. In a noncomatose patient who is not fasting, lactulose is given orally and titrated from 15 to 45 mL every 8 to 12 hours to achieve two or three loose and acidic (pH < 6.0) stools daily. Diarrhea should be avoided because this will reduce patient compliance and may exacerbate HE by promoting dehydration or electrolyte imbalances. In patients who are comatose or maintained fasting because of variceal hemorrhage, lactulose can be given by enemas (300 mL in 1 Liter of tepid water) every 4 to 8 hours as needed to achieve resolution of HE. [139] Despite the lack of any large-scale study of nonabsorbable disaccharides in HE, [45], [140] most experienced physicians attest to the rapid efficacy of these agents (most often within 24 hours), especially if the precipitating factor can be removed. After an episode of HE, lactulose can be administered preventively at minimal effective doses. Lactitol, available in Europe, appears to be equally as effective as lactulose at the dose of 30 to 45 g/day and reportedly has the advantages of being associated with less flatulence and is more palatable. [141], [142] Lactose can be used in lactase-deficient patients at the dose of 100 g. [143] The efficacy of lactulose in MHE warrants further study. [144]

The use of oral antibiotics to eliminate the urease-producing bacteria should be a second-line treatment. Despite their comparable efficacy to nonabsorbable disaccharides, certain undesirable effects limit their use. Neomycin is the most widely used antibiotic for this indication, at a dose of 2 to 6 g/day. Despite its limited intestinal absorption (3%), long-term use can expose patients to nephrotoxicity and ototoxicity, especially in instances of renal failure. The most frequent complications are malabsorption and bacterial superinfection. For these reasons, neomycin should be tapered rapidly after 2 to 3 days of treatment for a total of 1 to 2 weeks of treatment. An alternative to neomycin is metronidazole, 500 mg/day for 1 week, but this has a risk for neurotoxicity, especially in cases of severe liver failure. Despite evidence suggesting that *Helicobacter pylori* infection could cause HE by generating ammonia, [145] there is insufficient evidence to date to warrant a systematic eradication in patients with HE who are *H. pylori* seropositive, unless they have upper gastrointestinal symptoms or lesions that otherwise warrant this strategy. The effectiveness of a combination of nonabsorbable disaccharides and antibiotics is still debated. There is limited evidence that they could be synergistic, [146] and logically this would occur if the nonabsorbable disaccharide acts on bacteria resistant to the antibiotic. However, in most cases, this is most likely not the case, and therefore the antibiotic will eradicate bacteria thereby bypassing the desired therapeutic effect of lactulose, which is to incorporate the ammonia into the bacterial proteins. [147] In fact, one study suggested that a combination of neomycin and lactulose resulted in an effect comparable to placebo. [140] Hence, the consensus is to start antibiotics only after failure of lactulose alone and to stop lactulose if stool pH increases above 6.0.

L-Ornithine-L-aspartate has been proposed in Europe for the treatment of HE with the hypothesis that this molecule decreases ammonia by recycling through the urea cycle and by increasing glutamine synthesis. Two placebo-controlled studies, one with an intravenous formulation [148] and the second with an oral formulation, [149] show promising results. Nitrogen excretion can also be increased by benzoate (by conjugating glycine to form hippuric acid) and by phenylacetate (by conjugating with glutamine to form phenylacetylglutamine). Benzoate has been successfully used to treat inborn errors in the urea cycle. [150] One controlled trial showed that benzoate was just as effective as oral lactulose [151] in treating HE. Benzoate can be given intravenously, which can be of benefit in severely confused or comatose patients.

Studies of treatments targeting false neurotransmitters such as branched-chain amino acids (BCAA), levodopa, and bromocriptine have not shown beneficial results of significance that warrant formal recommendations on their use. The theory that increased concentrations of aromatic amino acids produce false neurotransmitters forms the basis for trials of the BCAAs leucine, isoleucine, and valine. Increased levels of tyrosine, phenylalanine, and tryptophan enter the brain in exchange for glutamine generated from astrocytic ammonia detoxification. These aromatic amino acids in excess would then be metabolized into false neurotransmitters (octapamine and phenylethanolamine) and serotonin. Administration of BCAAs in theory could therefore reverse this excess in aromatic amino acids. Results have been conflicting: short-term crossover studies have shown no benefit of BCAAs in EEG or psychometric testing [152], [153]; one long-term trial found an improvement in mental state, blood ammonia levels, and psychometric testing. [154]

Flumazenil (1 mg intravenous bolus) may be beneficial in selected patients with severe HE, [21], [155] and no oral preparation is available for long-term treatment. This drug can also be considered when BZ use is suspected.

Finally, supportive care is also an essential part of the management of HE. Care must be taken in preventing bodily harm in confused patients and complications of decubitus in comatose patients; adequate intravenous or nasogastric hydration and feeding must be implemented when oral intake is contraindicated. Airway protection should be considered in deeply comatose patients if the prognosis warrants such a measure. In patients with persistent and progressively worsening HE, such as can be observed in severe acute alcoholic hepatitis with or without cirrhosis, the prognosis is irremediably poor, and therefore, minimally invasive life support is the most compassionate choice. In otherwise stable patients, next of kin should be informed of the reversible nature of HE and should be educated on how to recognize the beginning stages of HE once the patient has left the hospital. Compliance to diet and to preventive therapy with lactulose is greatly improved by repeated education of the patient and family through interviews by the attending physician, nursing staff, and dietitian.

HEPATOPULMONARY SYNDROME

Hepatopulmonary syndrome identifies a group of patients with cirrhosis exhibiting severe hypoxemia as a result of intrapulmonary vascular dilation. The first descriptions of patients with chronic liver disease accompanied by digital clubbing and cyanosis can be traced to the 1880s. [156] Although several clinical reports were published after 1950, the term *hepatopulmonary syndrome* was first used in the late 1970s to describe patients with cirrhosis that exhibited exercise-aggravated hypoxemia and orthodeoxia. [157] The increasing recognition of milder forms of the syndrome allows detection of compatible abnormalities in up to 17% of patients with cirrhosis awaiting liver transplantation. [158]

Definition

The classic triad of the hepatopulmonary syndrome encompasses liver disease, a gas-exchange abnormality, and intrapulmonary vascular dilations. The gas-exchange abnormality has been defined as one that results in a partial pressure of oxygen of less than 70 mm Hg. Mild hypoxemia is common in cirrhosis, seen in up to one third of patients. In a series of 100 subjects, PaO_2 values of less than 70 mm Hg were seen in 28% of patients, whereas PaO_2 values of less than 60 mm Hg were seen in 8% of individuals. [159] In the hepatopulmonary syndrome, hypoxemia can be severe. In a series of 22 patients, 13 of 22 subjects had a PaO_2 of less than 60 mm Hg. [160]

As patients with cirrhosis may have higher respiratory rates, an arterial-alveolar oxygen gradient of more than 20 mm Hg (accounting for alveolar carbon dioxide) may be a better reflection of functional impairment. [159] Age adjustments are also important when evaluating the alveolar-arterial oxygen difference. The syndrome was defined with two elements [159]: an alveolar-arterial oxygen difference greater than 15 mm Hg while breathing room air in an upright position, irrespective of the degree of arterial hypoxemia, and evidence of intrapulmonary vascular dilations using contrast-enhanced echocardiography.

In an earlier definition, the absence of underlying lung conditions was required for the diagnosis of the hepatopulmonary syndrome. [161] With the high prevalence of smoking and associated lung disease, as well as other liver-related lung problems (hepatic hydrothorax, pulmonary hypertension), this definition may be too narrow. Hepatopulmonary syndrome can coexist with other lung conditions. The increasing ability to detect the presence of intrapulmonary vascular dilations by noninvasive tools allows a definition based on anatomic abnormalities.

Clinical Features

Most patients with hepatopulmonary syndrome have underlying cirrhosis with variable degrees of hepatic decompensation. The severity of liver disease is not related to the degree of hypoxemia. [162] An association with other hepatic conditions has also been described. These conditions include entities with portal hypertension, such as inferior vena cava webs and nodular regenerative hyperplasia. The presence of portal hypertension or portosystemic shunting is important for the expression of the syndrome.

Patients describe the insidious and progressive onset of dyspnea. Although dyspnea is aggravated by exercise, it has unique features: An increase of symptoms while standing (platypnea) is the result of redistribution of blood to lower lung lobes (more perfusion, lower oxygenation). A corresponding sign is orthodeoxia, a worsening of oxygen saturation upon standing that is relieved by recumbency. Platypnea and orthodeoxia are associated with more severe forms of the syndrome.

Digital clubbing and cyanosis reflect the impact of chronic hypoxemia and are weakly sensitive but highly specific for the diagnosis of hepatopulmonary syndrome. [163] Spider nevi are commonly observed. Most series describe the presence of such angiomata in 80% to 90% of cases, raising the possibility of a similar pathogenic influence on the development of pulmonary vascular dilations. However, the specificity of spider nevi for the diagnosis of the hepatopulmonary syndrome does not appear to be high, 63% in a report on 112 patients. [163] Patients with spider nevi do exhibit more prominent features of the hyperdynamic circulatory state, [164] with a high cardiac output (CO) and low mean arterial pressure as a result of a decreased peripheral vascular resistance.

Hepatopulmonary syndrome with progressive hypoxemia is a debilitating condition. Mortality in these subjects is high, and appropriate candidates should be referred for liver transplantation.

Diagnosis

Abnormal Gas Exchange Lung function can be normal in patients with the hepatopulmonary syndrome. In cirrhosis, a restrictive lung pattern can reflect the presence of ascites or hepatic hydrothorax. Alveolar diffusion abnormalities are commonly seen in patients awaiting liver transplantation. In two large series, a decrease in the diffusion capacity for carbon monoxide was observed in more than half of the subjects. [165], [166] In a recent study, [158] a reduced carbon monoxide diffusing capacity (DL_{CO}) was more commonly seen in subjects with the hepatopulmonary syndrome. However, the mechanisms responsible for altered oxygen and carbon monoxide diffusion do not appear to be equal. After liver transplantation, a reduced DL_{CO} can still be observed in the presence of normalization of oxygen diffusion. [167] An alveolar-arterial gradient of 15 mm Hg [158] or 20 mm Hg [163] has been used to diagnose the hepatopulmonary syndrome. Determination of arterial oxygen levels is best obtained with the patient standing. In one series, the mean fall in PaO_2 was 12 mm Hg when going from the supine to the standing position. [168] Breathing 100% oxygen can also be used to demonstrate the presence of anatomic arteriovenous shunting, where there is perfusion without ventilation. Pulmonary vasodilation without anatomic shunts (excess perfusion with normal ventilation) can be corrected with 100% oxygen. This test appears specific, but not sensitive, in the diagnosis of the hepatopulmonary syndrome.

Pulmonary Vasodilation A mottled appearance of the chest radiograph has been described in patients with clubbing and dyspnea. [169] Bilateral interstitial markings, especially in the lower lobes, may reflect the presence of arterial vasodilation. The diagnosis of vasodilation requires more precise instruments.

Contrast echocardiography. With this approach, microbubbles derived from agitated saline are injected intravenously. Under normal circumstances, they are not visible in the left atrium because they cannot traverse the pulmonary circulation. Although the immediate appearance of bubbles (after one heart beat) suggests the presence of a patent foramen ovale or an atrial septal defect, the diagnosis of pulmonary vasodilation is supported by the appearance of bubbles within three to five beats. The transthoracic approach is preferred over transesophageal echocardiography because the marginal improvement in imaging does not justify the higher cost and effort. There is an improved sensitivity of the transesophageal approach in cases of early hepatopulmonary syndrome, [170] but such initial changes are unlikely to change management. There is disparity between findings at contrast echocardiography and blood gas exchange. In up to 40% of cirrhotic patients, an abnormal contrast echocardiogram is not accompanied by evidence of hypoxemia. [171] Compensatory mechanisms to vasodilation, including hyperventilation, maintain oxygenation in these circumstances. When other pulmonary abnormalities exist, such as hydrothorax or obstructive disease, contrast echocardiography is not an accurate tool. Hypoxemia can arise from the alternative condition, and a positive bubble echo may not be sufficiently specific. Contrast echocardiography is a sensitive tool, valuable for screening, but lacking specificity.

Radionuclide lung perfusion scanning. Injection of technetium-labeled macroaggregated albumin intravenously results in homogenous lung imaging as the macroaggregates lodge in pulmonary arterioles. When macroagreggates pass through the pulmonary vasculature, they are detected in extrapulmonary tissues. When the brain is scanned, less than 6% of total counts can be detected under normal circumstances. [172] In their series of 25 subjects with the hepatopulmonary syndrome, all patients with positive scans (more than 6% brain uptake) had hypoxemia with PaO_2 values below 60 mm Hg, with a positive correlation between the degree of shunting and arterial hypoxemia. The lung perfusion scan provides specificity to the diagnostic workup. It is well suited for patients with intrinsic lung problems, in whom we have reviewed the difficulties in using contrast echocardiography. As all specific tests, it is not appropriate when used as a screening tool. Contrast echocardiography should be used in such a role; in addition, this technique also provides information on cardiac function and pulmonary pressure, important for the differential diagnosis.

Pulmonary angiography. This invasive diagnostic procedure lacks sensitivity for the diagnosis of the hepatopulmonary syndrome. Two types of abnormalities have been described: a diffuse “spongiform” appearance of pulmonary vessels during the arterial phase and a distinct arteriovenous communication. [172] The latter can be suspected in patients with severe hypoxemia and a poor response to 100% oxygen. Such anatomic communications have been closed by embolization [173] and may even persist after liver transplantation. A less invasive approach to anatomic visualization is high-resolution chest CT. One report noted a good correlation between dilated pulmonary vessels and hypoxemia in patients with hepatopulmonary syndrome. [172] A diagnostic algorithm has been proposed (Fig. 116-5). It uses arterial blood gases, pulmonary function tests, and contrast echocardiography as the initial tools. Lung scanning is reserved for those individuals with hypoxemia, positive bubble echo, and abnormal pulmonary function tests.

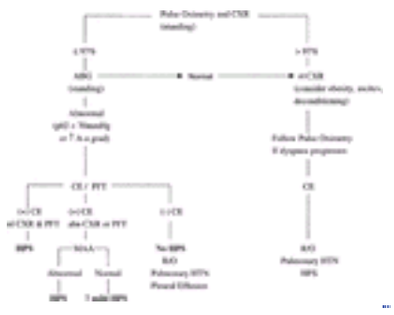

FIGURE 116-5. A diagnostic approach to the hepatopulmonary syndrome. (From ref. [163].)

Pathogenesis

The hyperdynamic circulation, a characteristic feature of the portal hypertensive state, arises from the effects of peripheral vasodilation. Pulmonary vasodilation could be viewed as part of this generalized hemodynamic picture. However, several observations indicate specific elements in its development. Although the degree of peripheral vasodilation worsens with the extent of liver failure, the development of the hepatopulmonary syndrome appears to be independent of the degree of hepatic dysfunction. Furthermore, only a subset of patients develop this unique pulmonary picture, in comparison to the more generalized prevalence of the hyperdynamic circulation.

Anatomic and Physiological Alterations Pathological studies point to a structural alteration of the pulmonary microcirculation. Among salient findings is vasodilation at the precapillary level, especially in gas-exchange areas. These were observed with postmortem injections of microopaque gelatin into the pulmonary vascular tree.[174] Spider nevi were seen on the pleura in almost half of the patients. One subject had a fully developed arteriovenous communication. No structural abnormalities were detected in the alveolar wall or connective tissue. Of interest, spider nevi have also been described in the pleura of patients dying from FHF.[175] Arteriovenous communications are likely to arise from the opening of preexisting anatomic pulmonary vascular communications as a result of increased blood flow. In children with tricuspid atresia, in whom a cavopulmonary communication is constructed to bypass the atretic segment, an angiographic picture similar to the hepatopulmonary syndrome develops in up to 25% of subjects, with dilated vessels and arteriovenous communications. Of interest, inclusion of hepatic vein flow directly into the pulmonary arterial bed appears to reduce the development of such complications.[176] The functional impairment can be defined as an alveolar-capillary oxygen disequilibrium.[177] In this concept, oxygen from the alveolar side has a longer path to traverse to reach the center of the capillary, where red cells constitute the center stream, resulting in hypoxemia. Although oxygenation would be improved by administering 100% oxygen, this measure would not correct hypoxemia if defined arteriovenous communications were present. A reduced transit time in the pulmonary capillary bed as a result of an increased CO further limits blood oxygenation, explaining the aggravation of symptoms with exercise. The input of multiple factors, including arteriovenous communications, cardiac performance, minute ventilation, and limitations of oxygen diffusion, explains the clinical spectrum of this condition. At one end of the spectrum are patients with severe hypoxemia have prominent pulmonary arterial vasodilation, on occasion with discrete arteriovenous communications. At the other end are patients with a more modest degree of shunting, in whom activation of compensatory mechanisms allows for maintenance of normal oxygenation.

Mechanisms of Vasodilation A critical role for nitric oxide has been established in the pathogenesis of the hyperdynamic circulation.[178] In the pulmonary circulation, a vasodilatory role for nitric oxide is also suggested by the demonstration of increased exhaled nitric oxide.[179] In 18 patients, a doubling of exhaled nitric oxide was detected, which normalized after transplantation. The normalization of the alveolar-arterial oxygen gradient was related to the reduction in exhaled nitric oxide. The effects of intravenous methylene blue, an inhibitor of guanylate cyclase (normally activated by nitric oxide to increase the level of cyclic guanosine monophosphate), further support a pathogenic role for nitric oxide.[180] In seven patients with severe hepatopulmonary syndrome (PaO_2 of 58 mm Hg), methylene blue improved PaO_2 to 74 mm Hg, associated with a rise of pulmonary and systemic vascular resistance (SVR). In a rat model of cirrhosis secondary to common bile duct ligation, intrapulmonary vasodilation and features of the hepatopulmonary syndrome (hypoxemia and increased alveolar-arterial oxygen gradient) can be observed. More recent studies demonstrate an increased exhaled nitric oxide in this model[181], [182] with demonstration of higher pulmonary total and calcium-dependent nitric oxide synthase activities. An increase in inducible nitric oxide synthase activity could be explained as a result of accumulation of intravascular macrophages. Treatment with an inhibitor of nitric oxide synthase for 6 weeks normalized features of the hepatopulmonary syndrome in this model. Other isoforms of nitric oxide synthase have been incriminated. Endothelial nitric oxide synthase was selectively increased in the lungs of rats with common bile duct ligation.[183] The possibility has been raised that production of endothelin-1 by the diseased liver, with subsequent increase in delivery to the lung vasculature, could be the stimulus for endothelial activation.[163] Endothelin-B receptors are present in the lumenal surface of the pulmonary vascular endothelium, and their activation results in vasodilation. Regardless of which isoenzyme is activated (pointing at different mechanisms of stimulation), it appears that pulmonary production of nitric oxide plays a critical role in the development of the hepatopulmonary syndrome. Further therapeutic interventions to inhibit its production are expected.

Treatment

Oxygen therapy is less likely to help those individuals who need it the most; severe hypoxemia is indicative of more conspicuous vasodilation. Nonetheless, a trial of oxygen therapy is often attempted in these patients.

A large number of medications have been tried, without definitive results. These include vasoconstrictors, such as phenylephrine[184] and somatostatin analogues.[160] Aspirin and indomethacin showed mild improvement in 4 subjects.[185], [186] More puzzling was the response to garlic powder: 6 of 15 subjects had improvements in PaO_2 of greater than 10 mm Hg.[187] The vasoactive component of garlic powder has not been identified. We have reviewed the experience with intravenous methylene blue; in one patient, L-NAME, an oral inhibitor of nitric oxide synthase, did not result in improvement.[188]

Hepatopulmonary syndrome occurs in the setting of portal hypertension. Relief of portal hypertension with TIPS results in complete portosystemic shunting with an increase in cardiac preload. The response of the hepatopulmonary syndrome to TIPS has been examined in a few patients. In one case,[189] an improvement of oxygenation of 20 mm Hg was detected 6 months after the procedure; the persistence of an abnormal lung scan suggested a mechanism of improvement whereby the CO was redirected to areas with better ventilation-perfusion characteristics. Worsening of oxygenation parameters has also been reported.[190] TIPS should not be used for the treatment of the hepatopulmonary syndrome unless more convincing evidence is provided.

In the 1980s, the presence of the hepatopulmonary syndrome was viewed as a contraindication to liver transplantation. Postoperative hypoxemia results in a more difficult recovery, and mortality is increased after the procedure. A PaO_2 of less than 50 mm Hg puts the patient at high risk for a morbid course.[191] Unique postoperative complications include the difficulty in discontinuing mechanical ventilation and the development of cerebral embolic hemorrhages. Newer approaches to postoperative management have been proposed, including the use of inhaled nitric oxide. However, when these patients are followed over a prolonged period, normalization of hypoxemia occurs. In 81 transplant recipients, improvement or normalization occurred in 82% of patients.[191] The recovery process may last more than 1 year.[192] At this time, liver transplantation is the only proven therapy for the hepatopulmonary syndrome.

The rise in pulmonary vascular resistance (PVR) that is seen after liver transplantation can unmask the presence of primary pulmonary hypertension.[193], [194] The coexistence of both entities can be difficult to diagnose before the procedure.

PORTOPULMONARY HYPERTENSION

Pulmonary artery hypertension complicating portal hypertension, known as *portopulmonary hypertension* (PPHTN), can be the cause of serious comorbidity and mortality in the patient with advanced hepatic disease. Like the hepatopulmonary syndrome, PPHTN is a pulmonary complication of portal hypertension. Hepatopulmonary syndrome results from pulmonary vasodilation and causes hypoxia. In contradistinction, PPHTN is a consequence of pulmonary vasoconstriction and is not a usual cause for hypoxia.

Mantz and Craige, in 1951, first described cor pulmonale in a patient with portal vein thrombosis, portal hypertension, and a spontaneous portosystemic shunt.[195] Later observations established that PPHTN can occur with cirrhosis and portal hypertension, extrahepatic portal hypertension and, rarely, after a surgically created portosystemic shunt. Portopulmonary hypertension may often be difficult to manage by medical therapy, can contraindicate an otherwise curative liver transplantation, and may persist after liver replacement. The reported mean survival of 15 months after diagnosis and a 50% 6-month mortality rate is poorer than the mortality

associated with primary pulmonary artery hypertension. [196] This emphasizes the dire significance of this diagnosis. Before the diagnosis of PPHTN can be made, other causes of elevated pulmonary artery pressure (PAP), including intracardiac shunts, valvular heart disease, chronic hypoxia, chronic pulmonary venous thrombosis, and connective tissue disorders must be excluded by relevant diagnostic testing.

Incidence

The reported prevalence of PPHTN in patients with portal hypertension is dependent on the patient population studied, the method of diagnostic detection, and the criteria selected for diagnosis. In an autopsy series of 17,901 patients, McDonnell and colleagues [197] recorded a 0.73% prevalence of pulmonary artery hypertension in patients with cirrhosis; in a comparative group of patients without cirrhosis, the prevalence was 0.13%. Hemodynamic study of cirrhotic patients with portal hypertension but without suspicion of pulmonary artery hypertension by Hadengue and associates [198] identified PPHTN in 2% of those patients. Ramsey and colleagues [199] reported a mean pulmonary arterial pressure (MPAP) greater than 25 mm Hg in 8.5% of patients and PVR greater than 120 dynes/sec/cm $^{-5}$ in 12.5% of patients evaluated for liver transplantation. A greater prevalence of PPHTN among patients evaluated for liver transplantation suggests that development of PPHTN is related to the duration and severity of portal hypertension.

Pathophysiology

The exact pathogenesis of PPHTN is not well understood. A current and best hypothesis is that the portosystemic shunting of splanchnic blood and impaired hepatic function allows arteriopathic and vasoactive mediators access to the pulmonary circulation. Endothelial cell injury may initiate a cascade of histological changes that are an early component of pathogenesis. [200] Eventually, a plexogenic arteriopathy of small precapillary pulmonary arterioles marked by laminar intimal fibrosis, medial hypertrophy, and concentric fibrosis characterizes the pathology of PPHTN. [201] A relative excess of humoral vasoconstrictors like endothelin and thromboxane A_2, and a relative decrease in mediators of vasodilation, like nitrous oxide and prostacyclin, favor vasoconstriction and the development of PPHTN. [200] Microvascular thrombosis because of excess thrombin, deficient fibrinolysis, and intrinsic platelet dysfunction caused by cirrhosis contribute to further increases in PVR. [202]

Most patients with cirrhosis and portal hypertension have a hyperdynamic circulation characterized by a high CO, elevated cardiac index, and a low SVR. Normally, flow-related increases in PAP caused by increases in CO induce pulmonary vascular dilation and recruitment of new pulmonary vasculature. PPHTN results when these adaptive mechanisms are attenuated by the progressive pulmonary arteriopathy. Further increases in hyperdynamic flow result in worsening severity of PPHTN.

Diagnosis

The diagnosis of PPHTN is based on clinical suspicion, the results of noninvasive diagnostic testing, and hemodynamic measurements allowed by a confirmatory right heart catheterization.

Clinical Considerations The patient with PPHTN may be asymptomatic. Usually, the diagnosis of PPHTN is made after the diagnosis of portal hypertension is established. Robalino and Moodie [196] described the clinical findings in cases of PPHTN. The most common presenting symptom was exertional dyspnea, which was present in most symptomatic patients. Other symptoms, including syncope, chest pain, fatigue, hemoptysis, and orthopnea, occurred infrequently. Systemic arterial hypertension, a pronounced pulmonic second sound (P_2), or a right ventricular heave predicted PPHTN in a group of patients with cirrhosis and portal hypertension being evaluated for liver transplantation. [203] Abnormal chest radiograph findings that are more common to cirrhotic patients with PPHTN include the prominence of the main and central pulmonary vessels, right ventricular enlargement, and peripheral vascular pruning that are also common to patients with primary pulmonary hypertension. [204] Electrocardiographic abnormalities of right ventricular hypertrophy, right atrial dilation, right bundle branch block, and premature atrial contractions in the patient with portal hypertension should alert the observer to the possibility of coexisting PPHTN.

Echocardiography Transthoracic Doppler echocardiography is the most important noninvasive test used to establish the diagnosis of PPHTN. Results of this examination can often exclude ventricular failure, intracardiac shunting, and valvular dysfunction as causes of elevated PAP. Echocardiographic indicators of PPHTN are right atrial dilation, right ventricular hypertrophy, interventricular septal wall thickening, and bulging of the interventricular septum into the left heart. Measurement of the tricuspid valve regurgitant jet velocity by echocardiography allows an indirect estimation of the MPAP. Importantly, most patients with PPHTN have tricuspid valve regurgitation. Torregrosa and associates [205] studied 107 patients with echocardiography who had cirrhosis and were referred for possible liver transplantation. Seventeen patients had suspected PPHTN when echocardiography diagnosed a systolic MPAP greater than 40 mm Hg or a pulmonary acceleration time of less than 10 ms. Hemodynamic monitoring confirmed PPHTN in only 5 of these patients.

Hemodynamic Assessment Right heart catheterization is the diagnostic gold standard used to establish the diagnosis of PPHTN. Catheterization allows direct measurements of PAP, CO, and pulmonary capillary wedge pressure (PCWP) and indirect calculations of PVR and SVR. Criteria from right heart catheterization that establish a diagnosis of pulmonary artery hypertension include an elevated MPAP of greater than 25 mm Hg at rest, an elevated PVR of greater than 120 dynes/second/cm $^{-5}$, and a PCWP of less than 15 mm Hg. [206] The high CO and low SVR characteristic of the hyperkinetic circulation of cirrhosis and portal hypertension is usually confirmed. Volume overload or diminished left ventricular function as a cause of the elevated PAP is excluded by a PCWP less than 15 mm Hg. The severity of PPHTN is classified as mild, moderate, or severe. Complete unanimity does not exist in terms of specific levels of MPAP defining each category. Generally, PPHTN is classified as mild when MPAP is 25 to 35 mm Hg, moderate when PAP is between 35 and 50 mm Hg, and severe when MPAP is greater than 50 mm Hg. Kuo and associates [204] compared selected hemodynamic and clinical data of 30 patients with PPHTN to similar data in equivalent patient groups with only cirrhosis and portal hypertension or primary pulmonary hypertension. Patients with PPHTN demonstrated hemodynamic findings also observed in patients with cirrhosis and portal hypertension or with only primary pulmonary hypertension. Patients with PPHTN had the hyperdynamic circulation common to cirrhosis. Portopulmonary hypertension patients demonstrated increases in PVR similar to those found in the patients with primary pulmonary artery hypertension. Echocardiography, chest radiography, and electrocardiographic findings suggesting pulmonary artery hypertension were similar in patients with PPHTN and primary pulmonary artery hypertension. Patients with PPHTN had a significantly more severe respiratory alkalosis than patients with primary pulmonary hypertension or cirrhosis alone. In patients with cirrhosis, a PCO_2 of less than 30 was an especially specific indicator for PPHTN.

Treatment

Medical therapy for the patient with PPHTN involves the administration of pharmacological agents that, by vasodilator effect, reduce PAP. When administration of a vasodilator agent during a pulmonary artery catheterization reduces PVR and MPAP (diagnostic response), a therapeutic trial of vasodilator therapy should be considered. [207], [208] Long-term treatment may result in progressive hemodynamic improvements as evidenced by repeat right heart catheterization. Liver transplantation may be considered in selected patients with mild PPHTN or patients with more severe PPHTN that have responded to vasodilator therapy.

Epoprostenol Therapy Although acute reduction of PAP has been reported to result from the administration of several different agents, including isosorbide-5-mononitrate and inhaled nitrous oxide, the contemporary medical treatment of PPHTN is mainly limited to epoprostenol (formally PGI_2 or prostacyclin) infusion therapy. [209] Epoprostenol is a potent vasodilator agent with anti–platelet-aggregating properties that is naturally produced by vascular endothelial cells. Instability of epoprostenol at a pH of less than 10.5 and a short half-life require constant intravenous infusion by way of long-term vascular access for chronic therapy. Drug treatment–related side effects can include systemic hypotension, headache, nausea, and venous catheter-related complications. [210] Effective epoprostenol treatment of patients with primary pulmonary artery hypertension has been reported. [210] Immediate vasodilation followed by promotion of vascular remodeling that causes further reduction of PAP is proposed as an explanation for this medication's therapeutic effect. Patients with at least moderate PPHTN (MPAP higher than 35 mm Hg) who have had a diagnostic response to epoprostenol are candidates for a trial of longer-term epoprostenol therapy. Kuo and associates [209] reported improved hemodynamic parameters in four patients with PPHTM while treated with epoprostenol for a period of 6 to 14 months. Krowka and associates [211] treated 10 patients with an MPAP higher than 35 mm Hg for 8 days to 30 months. Six patients showing improvements in PVR, MPAP, and CO after 1 hour had further reduction of PVR when treated with continuous therapy. Available data do not permit firm conclusion as to the long-term benefit of epoprostenol therapy, nor how often the beneficial effect of therapy might persist after the cessation of treatment. Epoprostenol therapy is most appropriate for the cirrhotic patient who would, except for moderate to severe PPHTN, be a candidate for liver transplantation. [212]

Newer Therapies Newer and still to be developed vasoactive agents will undoubtedly be of benefit as future treatments for PPHTN. Treatment with treprostinil, a prostacyclin analogue, is of reported benefit in patients with primary and secondary pulmonary artery hypertension. Administration by a subcutaneous infusion, rather than the continuous intravenous route required by the less stable epoprostenol, reduces the risk for adverse complications due to long-term venous access. [213], [214] The efficacy of treprostenol in the treatment of patients with PPHTN has not been described. Bosentan is an orally administered drug and endothelin receptor agonist with reported efficacy in the treatment of primary pulmonary artery hypertension. [215] Unfortunately, bosentan-associated hepatotoxicity contraindicates its use in patients with preexisting liver disease.

Liver Transplantation Krowka and associates [216] reviewed the results of orthotopic liver transplantation in 43 patients with PPHTN. The 30-day mortality rate was 22%. A preoperative MPAP greater than 50 mm Hg was associated with a 100% mortality rate. Four additional patients died from cardiopulmonary failure between 5 and 30 months after operation. Mortality in patients with an MPAP of 35 to 50 mm Hg before surgery was 50%. All patients with a preoperative MPAP of less than 35 mm Hg survived. Ramsey and associates [199] reported that only 2 of 7 patients with an MPAP of more than 60 mm Hg experienced a good quality of health and life after liver replacement. [199] During liver transplantation, morbidity and mortality in patients with PPHTN is caused by the hemodynamic stresses of reperfusion. Intraoperative events that contribute to depressed right ventricular function include hypothermia, hyperkalemia, hypercapnia, and acidosis. The potential liver transplant recipient must be evaluated for possible PPHTN by the described methods. In general, an MPAP of greater than 35 to 40 mm Hg is recognized as an absolute contraindication to liver transplantation. Dobutamine-assisted stress echocardiography documenting left ventricular dysfunction in patients with PPHTN further excludes orthotopic liver transplantation consideration. Patients with an MPAP of less than 35 mm Hg are candidates for further consideration of liver transplantation. Patients without other contraindications to liver transplantation who have moderate to severe PPHTN are candidates for epoprostenol therapy. [217] The goal of epoprostenol therapy is reduction of MPAP to a level that allows further consideration for transplantation. A reasonable reduction in MPAP is to no more than 35 to 40 mm Hg. Patients successfully transplanted after chronic preoperative epoprostenol treatment require continued epoprostenol infusion therapy after liver transplantation. [212] Eventual reversal of PPHTN after liver transplantation may allow discontinuation of treatment. [211], [218] The required duration of postoperative therapy varies and is dependent on the resolution of PPHTN that is documented by repeat right heart catheterization. [217] Persistence, or worsening, of elevated PAP after liver transplantation is an indication to continue epoprostenol therapy. [219] Recurrent PPHTN accompanying graft failure has been reported. [220] More radical surgical treatment that includes simultaneous heart, lung, and liver transplantation has been described but is experimental. [221]

Summary

Portopulmonary hypertension is a pulmonary complication of portal hypertension that has a poor prognosis. The diagnosis requires confirmatory right heart catheterization. Vasodilator therapy with epoprostenol in selected patients may result in hemodynamic improvement. Patients with mild PPHTN (MPAP of less than 25 mm Hg) or patients with moderate and severe PPHTN responsive to epoprostenol therapy may benefit from liver transplantation.

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CHAPTER 117

Francis Y. K. Yao and Nathan M. Bass

LIVER TRANSPLANTATION

In the fourth decade since the inception of human orthotopic liver transplantation, [1] this procedure has become a highly successful treatment for patients with life-threatening, advanced liver disease. The 1-year patient survival according to the United Network for Organ Sharing (UNOS; Richmond, VA) registry for all adult liver transplants performed in the United States over a 2-year period from 1998 to 2000 was 87%, [2] compared to 79% from 1988 to 1994. [3] Many liver transplant centers have now achieved 1-year patient survival rates of about 90%, and 5-year survival rates of 70% to 80%. The success of liver transplantation is, however, limited by the serious crisis of donor shortage. [4] As of June 2001, a total of 17,983 patients in the United States were placed on the waiting list for liver transplantation, whereas only 4934 liver transplantation operations were performed in the year 2000 in over 120 liver transplant centers across the country [2] (Fig. 117-1). The rapid rise in the number of patients on the waiting list for liver transplantation has led to increased waiting time and a greater number of deaths each year among patients awaiting liver transplantation [2, 4, 5] (see Fig. 117-1). Organ donation has, however, remained relatively stagnant in recent years in spite of the concerted efforts to increase donation. [4]

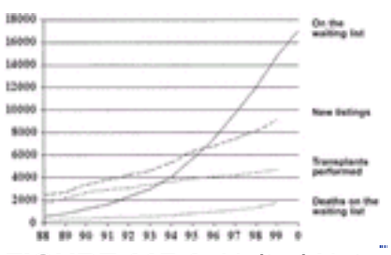

FIGURE 117-1. United Network for Organ Sharing (UNOS) data showing the trends in the liver waiting list, liver transplantation performed, and waiting list deaths over the past decade in the United States. Data for listed patients are for the end of each year up to and including 2000. (From UNOS data, March 2001; courtesy, John P. Roberts, M.D., Department of Surgery, University of California, San Francisco.)

This chapter reviews the surgical technique, organ preservation, indications and evaluation for liver transplantation, disease-specific issues and outcomes, and management of posttransplant complications.

SURGICAL TECHNIQUE AND PRESERVATION

Matching of an appropriate donor organ with a liver transplant recipient requires compatibility of the ABO blood type but not the human leukocyte antigen (HLA) tissue type. The conventional recipient operation involves orthotopic transplantation of the donor organ (i.e., in the normal anatomic location). The procedure is conventionally divided into three phases: the hepatectomy phase (removal of the recipient native liver), the anhepatic phase, and the reperfusion phase. The latter follows the complete vascular reconnection of the liver. The order of anastomoses of the donor liver usually commences with revascularization of the donor organ (suprahepatic vena cava, infrahepatic vena cava, portal vein, and hepatic artery) followed by biliary reconstruction. However, numerous variations exist in the technique of implanting the donor organ, based upon variations in anatomy, type of recipient liver disease, and surgical preference. [6] The hepatic arterial reconstruction is commonly achieved with an end-to-end anastomosis of the donor celiac axis with a Carrel patch of aorta to the recipient common hepatic artery or celiac axis. Vascular grafts directly from the recipient aorta may be used in some cases to ensure adequate arterial inflow. The usual approach to biliary reconstruction employs a direct duct-to-duct anastomosis (choledochocholedochostomy) except in instances where the recipient duct is diseased (primary sclerosing cholangitis, biliary atresia), a large discrepancy in size exists between the recipient and donor ducts, or there is inadequate length of bile duct for a tension-free anastomosis. In these cases, as well as in most very small children, a choledochojejunostomy with a Roux-en- limb of jejunum is performed. Finally, the donor gallbladder is removed. In order to avoid significant hemodynamic instability and congestion of the bowel and kidneys during the anhepatic phase, some centers employ a venovenous bypass circuit, which connects the portal vein and inferior vena cava to the left subclavian vein. [6]

In light of the critical shortage of donor livers in recent years, strategies that make use of segmental grafts have been developed and include splitting of cadaveric livers between two recipients, and the use of living donors. [7, 8] The latter has become almost routine in the case of the pediatric liver recipient in whom a left lateral segment, usually obtained from a blood-type–compatible parent, is sufficient, and is also a relatively safe procedure for the donor. [7] More recently, living adult-to-adult donor procedures have been developed and increasingly used. [8, 9] and [10] In order to provide a minimal functioning liver mass for the recipient, this procedure usually involves removing the right lobe from the donor and transplanting it orthotopically into the recipient (Fig. 117-2; Color Fig. 117-2). In both the donor and the recipient, there is rapid regeneration of the liver, with full size regained over 6 to 8 weeks. Despite the potential advantages of living-related liver transplantation, it carries inherent risks for the healthy donor, the full extent of which is yet to be fully elucidated. [9, 10] To date, there have been two known donor deaths among close to 1000 adult-to-adult living donor liver transplants performed in the United States. The American Society of Transplant Surgeons has published a position paper on adult-to-adult, living-related liver transplantation, [11] and practice guidelines have been established at a recent national consensus conference on living donor organ transplantation. [12]

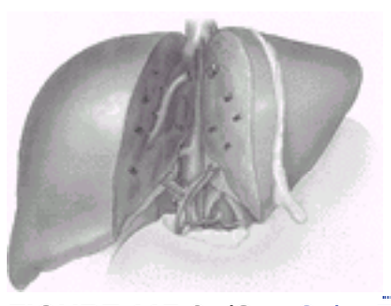

FIGURE 117-2. (See Color Fig. 117-2.) Illustration of technique of living donor right hepatic lobectomy. (Courtesy John P. Roberts, M.D., Department of Surgery, University of California, San Francisco.)

A major advance in the preservation of donor organ viability and extension of the geographic distance over which organs can now be procured and transported was the development of the University of Wisconsin (UW) cold storage solution with which the donor organ is flushed and cooled prior to removal and transportation. [13] The UW solution has extended the cold ischemia time for liver preservation for up to 24 hours, although it is uncommon for organs that have been preserved for longer than 16 hours to be used on a routine basis. Key principles of the UW solution are a high K^+ and Mg^{2+} and low Na^+ and Ca^{2+}. This solution will maintain intracellular ion concentrations, adenosine as a substrate for ATP synthesis, hydroxyethyl starch as an oncotic solute to reduce interstitial edema, and allopurinol and glutathione as antioxidants.

INDICATIONS FOR LIVER TRANSPLANTATION

Non–Disease-Specific Complications and Prognostic Models

Liver transplantation should be considered for patients with cirrhosis when one or more signs of hepatic decompensation develop. In a study from Barcelona, [14] the probability of decompensation was 58% at 10 years after the diagnosis of cirrhosis. Ascites was the most common manifestation of hepatic decompensation, followed by jaundice, hepatic encephalopathy, and variceal bleeding. The median survival with compensated cirrhosis was 8.9 years, compared to only 1.6 years for decompensated cirrhosis. [14]

Ascites and Related Complications Approximately 50% of patients with cirrhotic ascites succumb in 2 years. [15] Up to 10% of patients with cirrhotic ascites are refractory to standard diuretic therapy. [16] Treatment options for refractory ascites include serial therapeutic paracentesis, [17], [18] peritoneal venous shunt, [18], [19] and the transjugular intrahepatic portosystemic shunt (TIPS). [20] Survival for patients with refractory ascites is only about 50% at 1 year regardless of the type of treatment they have received. [17], [18], [19] and [20] Ascites may be associated with spontaneous bacterial peritonitis and hepatorenal syndrome, both of which further shorten survival. The 1-year probability of survival was reported in one study to be 66% for patients with cirrhosis and ascites without spontaneous bacterial peritonitis, compared to only 38% for patients with this complication. [21] In one third of patients with spontaneous bacterial peritonitis, renal impairment develops despite treatment of the infection. [22], [23] Hepatorenal syndrome is a particularly ominous complication of cirrhosis, associated with a median survival of only 1.7 weeks after its onset. [24] Renal failure prior to liver transplantation is a strong predictor of death [25], [26] and increased cost [27] after liver transplantation.

Portal Hypertensive Bleeding Bleeding from esophageal or gastric varices carries a high 30-day mortality ranging from 30% to 50%. [28] The risk of recurrent variceal bleeding is about 60% to 70% within 2 years of the index hemorrhage. [28] Better understanding of the basic pathophysiology culminating in portal hypertension and bleeding has led to important advances in pharmacological therapy including ß-blockade for primary and secondary prophylaxis of variceal bleeding, and vasopressin or somatostatin for the initial control of hemorrhage. [28] Endoscopic sclerotherapy has been the cornerstone of management of esophageal variceal hemorrhage, but endoscopic banding ligation appears to produce superior results with a lower incidence of rebleeding, and a trend toward lower mortality compared to sclerotherapy. [29] Another major advance in the treatment of variceal bleeding is TIPS which has a technical success rate of greater than 90% in experienced hands. [30], [31] Compared to endoscopic treatments, TIPS confers a lower rate of rebleeding but no survival advantage. [32], [33] The current accepted indications for TIPS include (a) emergency (rescue or salvage) therapy for uncontrolled, acute portal hypertensive bleeding and (b) prevention of recurrent portal hypertensive bleeding for which endoscopic or pharmacological therapy has either failed or cannot be applied. [28], [33] The Mayo Clinic group proposed a model based on serum bilirubin, serum creatinine, prothrombin time, and etiology of liver disease, which appears to be strongly predictive of 30-day mortality after TIPS. [34] Severe hyperbilirubinemia occurring after the creation of TIPS is a sign of rapid hepatic decompensation, associated with a particularly ominous short-term prognosis. [35] Encephalopathy develops in about 30% of patients after TIPS and is usually reversible with medical therapy. [28] A major problem with TIPS is the high incidence of shunt stenosis or occlusion, estimated to be 30% over the first year and 50% at 2 years. [36] TIPS has not been shown in a study from the National Institute of Diabetes and Digestive and Kidney Diseases (NIDDK) Liver Transplant Database to have any significant impact on liver transplantation in terms of total operative time, blood product requirements, survival, or length of hospital stay. [37] Consequently, preoperative portal decompression solely for facilitating liver transplantation is not an acceptable indication for TIPS. Occasionally, TIPS also creates technical difficulties for liver transplantation secondary to stent migration or extension into the extrahepatic portal or hepatic vein. [28], [33] Portosystemic shunt surgery provides more definitive control of bleeding compared to sclerotherapy [28], [38], [39] or TIPS [40] in patients with adequate hepatic function to tolerate surgery, but does not improve survival and is also associated with an increased incidence of encephalopathy. Distal splenorenal shunt is the preferred surgical approach in consideration for future liver transplantation since the hepatic hilum is not invaded.

Hepatic Encephalopathy Hepatic encephalopathy is a complex neuropsychiatric syndrome which may be present in 50% to 70% of all patients with cirrhosis, including those with subtle abnormalities only demonstrable by psychometric testing. [41] Hepatic encephalopathy is aggravated by portosystemic shunting via collaterals secondary to portal hypertension, and is also associated with surgical portosystemic venous shunting and the creation of TIPS. [29] Nonabsorbable disaccharides such as lactulose, and antibiotics including neomycin with activities against urease-producing bacteria are effective in reversing hepatic encephalopathy. The efficacy of lactulose and neomycin in the treatment of hepatic encephalopathy appears to be equal. [42] Symptomatic hepatic encephalopathy is an indication for consideration of liver transplantation. Uncommonly, some patients with hepatic encephalopathy have progressive, debilitating neurological or neuropsychiatric syndromes such as dementia, spastic paraparesis, cerebellar degeneration, and extrapyramidal movement disorders, associated with structural abnormalities of the central nervous system. [41] If very advanced, neurological injury may not be reversible with liver transplantation, although one report has suggested that there may be gradual improvement after liver transplantation. [43]

Child-Turcotte-Pugh Score The Child-Turcotte-Pugh (CTP) score was originally used to stratify patients according to the risks for undergoing portal decompressive shunt surgery. [44] The CTP score has also become the predominant method for predicting prognosis of patients with cirrhosis irrespective of etiology. [45], [46] This scoring system is based on five variables: stage of encephalopathy, severity of ascites, serum bilirubin level, serum albumin level, and the international normalized ratio (INR) for prothrombin time (Table 117-1). In a natural history study from Innsbruck University, Austria involving 620 cirrhotic patients followed for 15 years, [45] the estimated 1- and 5-year survival rates were 95% and 75%, respectively, for Child class B, and 85% and 50%, respectively, for Child class C. Patients with Child class A cirrhosis had significantly better survival probabilities exceeding 90% for up to 10 years of follow-up. [45] According to the minimal listing criteria for liver transplantation implemented by UNOS in 1997, [47] the key criterion for eligibility of listing for liver transplantation is an estimated 90% or less chance of 1-year survival, which equates with a CTP score of 7 or higher (Child class B or C). In addition to using the CTP score for determining eligibility of listing for liver transplantation (minimal listing criteria), [47] the UNOS guidelines previously relied on this scoring system to determine the priority in organ allocation among patients with chronic liver disease. [48] Patients with fulminant liver failure, who are at greatest risk for short-term mortality, are classified in a separate category and listed with the highest priority (status 1) for liver transplantation. [48]

	1 POINT	2 POINTS	3 POINTS
Encephalopathy (stages)*	0	1–2	3–4
Ascites	None	Slight	Moderate
Bilirubin (mg/dL)	<2	2–3	>3
(PBC and PSC)	<4	4–10	>10
Albumin (g/dL)	>3.5	2.8–3.5	<2.8
PT prolonged (sec)	1–4	5–6	>6
(INR)	<1.7	1.8–2.3	>2.3

*Encephalopathy stages: 0 to 4 (ref. 45).

INR, international normalized ratio; PBC, primary biliary cirrhosis; PSC, primary sclerosing cholangitis; PT, prothrombin time.

Notes: The individual measurements are allocated 1, 2, or 3 points, and the points from the 5 variables are added to give a total score ranging from 5 to 15 points.

Child's classes A, B, and C correspond to Child-Turcotte-Pugh score of 5–6 points, 7–9 and 10–15, respectively.

Adapted from ref. 44.

TABLE 117-1 The Child-Turcotte-Pugh Classification

Model for End-Stage Liver Disease The Model for End-Stage Liver Disease (MELD) prognostic score has been developed using a formula based on serum creatinine, serum bilirubin, and INR for prothrombin time to predict short-term mortality for patients with end-stage chronic liver disease [49] (Table 117-2). This model was based on multivariate analysis of readily and objectively measurable variables originally developed to assess the short-term prognosis of patients with cirrhosis undergoing the TIPS procedure. [34] The MELD scoring system was subsequently shown to also be useful in grading the risk of death at 1 week, 3 months, and 1 year for patients with end-stage chronic liver disease [49] (Table 117-2). The MELD score has been implemented by UNOS as a disease severity index to determine priority for organ allocation, [50] replacing the previous system based on the CTP score and other specific clinical criteria. [48] Using the MELD model, the patient with the highest score reflecting the greatest risk for short-term mortality will receive liver transplantation first, and the length of waiting time will be a minimal factor in determining organ allocation. [50]

TABLE 117-2 The Model for End-Stage Liver Disease (MELD) Scoring System Determining Disease Severity and Priority for Organ Allocation

Quality-of-Life Issues Encephalopathy, ascites, and debilitating fatigue are some of the common manifestations of end-stage liver disease that markedly impair a patient's quality of life. Even subclinical hepatic encephalopathy, determined only by psychometric analysis, can cause significant impairment of daily functioning. [51] Liver transplantation improves quality of life, and facilitates return to gainful employment in over half of the patients. [52], [53] and [54] Patients with severe impairment of quality of life but not meeting the listing criteria based on the CTP score may be listed for liver transplantation on exceptional grounds upon approval of a regional UNOS-appointed review board. [47]

Disease-Specific Issues and Outcome After Liver Transplantation

More than 60 distinct diseases have been treated with liver transplantation. [55] The relative frequencies of disease indications for liver transplantation in the United States based on recent UNOS data [5] are presented in Figure 117-3. Hepatitis C-related cirrhosis is currently the most common indication for liver transplantation in the United States, accounting for over 20% of all liver transplants. [5] The guidelines for liver transplantation, special considerations, the controversial aspects of certain disease indications, and posttransplant outcome for specific types of liver disease are selectively reviewed (Table 117-3). Many of the less common types of liver disease will not be discussed in detail in this chapter.

FIGURE 117-3. Liver transplantation in the United States according to the primary diagnosis. Data from ref. [5] for the year 1998. *ALD,* alcoholic liver disease; *PBC,* primary biliary cirrhosis; *PSC,* primary sclerosing cholangitis.

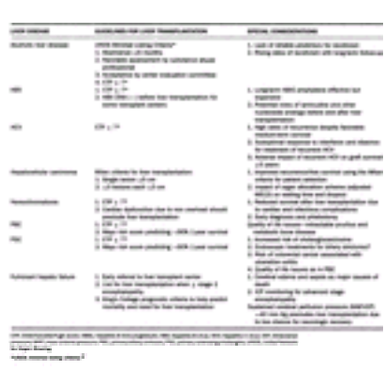

TABLE 117-3 Summary of Disease-Specific Guidelines for Liver Transplantation

Alcoholic Liver Disease Once considered a highly controversial indication for liver transplantation on both medical [56] and ethical grounds, [57] alcoholic liver disease is second only to hepatitis C as the most common indication for liver transplantation in the United States (see Fig. 117-3). Survival following liver transplantation for alcoholic cirrhosis has been reported to be at least as good as for most other indications. [58], [59], [60] and [61] The obvious concern, however, is the possibility of returning to alcohol consumption after liver transplantation. The patient transplanted for alcoholic liver disease who drinks again creates a highly emotional and demoralizing issue and is often perceived as a treatment failure, even though the absolute alcohol intake may be low and graft function normal. There is insufficient data to fully understand the impact of moderate alcohol consumption after liver transplantation, and the extent and pattern of graft injury. The reported incidence of recidivism following liver transplantation has typically ranged from 10% to 20% at 12 months after liver transplantation. [62] With longer follow-up, however, the recidivism rate for any alcohol use has exceeded 30%. [62], [63], [64] and [65] Better insight into patient selection, closer monitoring, and ngoing counseling for these patients after liver transplantation are therefore needed. There is a strong consensus based on a survey of liver transplant centers and a National Institute of Health workshop to require an established period of abstinence, typically 6 months, before approval for listing for liver transplantation. [66], [67] The required period of abstinence is useful since abstinence alone can often lead to sustained clinical improvement, [68] thereby obviating the need for liver transplantation. The 6-month rule for abstinence as a predictor of long-term sobriety remains controversial, [63], [69], [70] and should be used in conjunction with other psychosocial and psychiatric parameters in the patient selection process. The University of Michigan alcoholism prognosis scale [61] and several other models [62] have been proposed to predict the risk of recidivism, but the value of these models as a tool for patient selection has not been clearly established. The UNOS minimal listing criteria for alcoholic liver disease include:

- abstinence of at least 6 months
- favorable assessment by a substance abuse professional
- approval by the transplant center evaluation committee, in addition to fulfilling the medical criteria to be listed for liver transplantation. [47]

Hepatitis B Less than a decade ago, liver transplantation for patients with cirrhosis due to chronic hepatitis B infection was considered a relative contraindication due to high rates of hepatitis B recurrence and graft failure. [71], [72] and [73] The most aggressive form of hepatitis B recurrence, known as fibrosing cholestatic hepatitis, often leads to death within weeks of onset of graft reinfection. [74] Subsequently, the use of high-dose intravenous hepatitis B immunoglobulin (HBIG) after liver transplantation significantly reduced the incidence of hepatitis B recurrence and improved survival. [75], [76] and [77] The benefit of HBIG appears to be greatest among patients with negative hepatitis B DNA and hepatitis B e-antigen prior to liver transplantation. [75] Samuel and colleagues from France [78] reported their long-term survival and hepatitis B recurrence data on 89 patients with hepatitis B-cirrhosis who received liver transplantation and intravenous HBIG prophylaxis. Despite an excellent actuarial 10-year survival of 72.8%, the 5-year actuarial hepatitis B recurrence rate was still high at 65% with HBIG prophylaxis. [78] Graft reinfection despite HBIG prophylaxis may be caused by inadequate neutralization because of overwhelming amounts of hepatitis B viral replication or breakthrough infection by hepatitis B surface antigen protein escape mutants. [79], [80] Another concern of intravenous HBIG is its high cost, estimated to be $30,000 to $50,000 in the United States during the first year of therapy after liver transplantation. [81] Lamivudine, an oral nucleoside analog, has been used both in the prophylaxis and treatment of recurrent hepatitis B infection after liver transplantation. [81] Based on preliminary experience, lamivudine alone started preemptively before transplantation and continued thereafter appears to be less effective than HBIG for prophylaxis against hepatitis B recurrence after liver transplantation. [81], [82] A potentially more effective strategy for hepatitis B prophylaxis, based on limited data, is a combination of lamivudine with intramuscular or intravenous HBIG. [83], [84] and [85] Randomized, controlled studies are currently underway to evaluate these different strategies. Preliminary data have also suggested that HBIG can be safely discontinued after short-term (6–12 months) use

following liver transplantation, with continued long-term lamivudine. [86] Lamivudine is also effective in the treatment of recurrent hepatitis B infection after liver transplantation. In the largest published series by Perrillo and associates [87] using lamivudine for 52 weeks, 60% of the 52 patients cleared hepatitis B viral DNA, 31% cleared hepatitis B e antigen, 6% lost hepatitis B surface antigen, and 71% had normalization of alanine transaminase levels. In patients with decompensated hepatitis B-cirrhosis awaiting liver transplantation, data have emerged suggesting the effectiveness of lamivudine in stabilizing liver function and improving the CTP score, [88], [89] and [90] which may also confer a survival benefit. [90] An important concern of long-term use of lamivudine beyond 6 to 9 months, regardless of the clinical setting, is the high rate of emergence of lamivudine-resistant mutant virus (YMDD mutant), estimated to be about 20% to 30% per year in posttransplant patients. [81] This mutation may lead to deterioration of graft function in some patients. [81] Other nucleoside or nucleotide analogs, such as adefovir dipivoxil, may become available to serve as “rescue” therapy should lamivudine resistance occur with deterioration of liver function. [91], [92]

Hepatitis C Despite almost invariable persistence of serum hepatitis C RNA after liver transplantation and a rate of graft re-infection (histological evidence of hepatitis) of 40% to 75%, [93] survival up to 5 years is similar to that for other indications for liver transplantation. [94], [95], [96] and [97] While prospective long-term survival data following liver transplantation for hepatitis C infection are very limited, it is increasingly recognized that the course of posttransplant recurrent hepatitis C is more rapid when compared with that prior to liver transplantation. [93] About 10% to 20% of patients with recurrent hepatitis C infection progress to cirrhosis within the first 5 years after liver transplantation. [93], [94], [96] A severe form of recurrence, resembling the syndrome of fibrosing cholestatic hepatitis in hepatitis B-infected patients, has also been reported. [98] Factors predictive of more severe recurrent disease and worse outcome after liver transplantation have not been clearly elucidated, but include a high pretransplant hepatitis C RNA titer (at least 1 million mEq/mL) [95] and genotype 1b. [97], [99], [100] Other studies, however, have not supported these associations. [96], [101] Patients with severe and multiple rejection episodes may also have early and more severe recurrent hepatitis C, presumably related to more intense immunosuppression. [102], [103] and [104] The type of immunosuppression (cyclosporine versus tacrolimus) does not clearly appear to have any significant impact on the incidence or severity of hepatitis C recurrence, [105], [106] but additional studies to further evaluate the impact of these and other new immunosuppressive agents on the course of recurrent hepatitis C are needed. Treatment of recurrent hepatitis C after liver transplantation with either interferon or ribavirin alone has yielded disappointing results. [107], [108] and [109] Combination therapy with interferon and ribavirin for recurrent hepatitis C appeared promising initially in a pilot study, especially among patients treated early in the course of the disease. [110] However, subsequent studies using this therapeutic combination have shown suboptimal virologic response and poor patient tolerance. [111], [112] Factors contributing to the latter include renal insufficiency, anemia, and thrombocytopenia commonly observed in this population. The dose of ribavirin used in these patients is typically less than that in the nontransplant setting, [111] in large part due to reduced glomerular filtration rate associated with both cyclosporine and tacrolimus. Another approach has been to use interferon with or without ribavirin early after liver transplantation for prophylaxis against hepatitis C recurrence. [113] and [114] In a study by Sheiner and colleagues, [113] a 1-year course of interferon alone commenced within 2 weeks after liver transplantation significantly reduced the incidence of hepatitis C recurrence, particularly for patients with low hepatitis C RNA levels. [113] Multicenter, randomized controlled trials have been initiated in Europe and in the United States to assess the optimal timing and efficacy of interferon and ribavirin for posttransplant hepatitis C infection. Transplantation of a hepatitis C-positive donor liver into a recipient with hepatitis C-cirrhosis does not appear to result in a worse outcome in the initial years after liver transplantation, [115] and may prove to be an important strategy for expanding the donor pool for liver transplantation.

Hepatocellular Carcinoma Liver transplantation for hepatocellular carcinoma (HCC) in unselected patients, including those with extensive and bulky tumors, was associated with dismal outcome primarily due to aggressive tumor recurrence. More recent experience, however, has suggested excellent recurrence-free survival after liver transplantation in patients with small tumors, with 5-year survival rates consistently above 70%. [116], [117], [118], [119] and [120] In a study by Bismuth and associates from France, [116] patients with no more than 3 tumor nodules and none greater than 3 cm in largest diameter had a 3-year disease-free survival of 83% following liver transplantation compared to only 18% treated with hepatic resection. In a subsequent study by Mazzaferro and colleagues from Milan, Italy, [117] 35 patients who met the specific criteria of a solitary tumor not exceeding 5 cm or no more than 3 tumor nodules, none greater than 3 cm had excellent overall and recurrence-free survival rates of 85% and 92%, respectively, at 4 years after liver transplantation. [117] The recurrence-free survival in patients with small HCC was identical to that of patients without tumor. [118] The Milan criteria [117] have been incorporated into the UNOS guidelines for liver transplantation for HCC. Poorly differentiated tumor grade and microvascular invasion appear to correlate with advanced tumor-stage based on the size and number of lesions, and are also potential predictors for poor outcome after liver transplantation. [121], [122] Despite excellent survival after liver transplantation for selected patients with HCC, tumor growth in the face of increased waiting time for liver transplantation in this country presents a considerable challenge, as eventually, acceptable criteria for liver transplantation may be exceeded. Advances in living-related liver transplantation may be a potential solution for this major problem by overcoming the waiting time obstacle in those patients who have a suitable donor. [123] However, there are still logistic and ethical concerns about this option. [9], [10] In order to control tumor growth, many liver transplant centers use chemoembolization, percutaneous ethanol injection, or radiofrequency ablation as “bridging procedures” to liver transplantation. [120], [124] However, the benefit of these procedures in slowing the rate of tumor growth and ultimately preserving the chance for successful liver transplantation is unproven. [120], [124] Due to the high dropout rate related to tumor growth in patients on the waiting list, the current MELD system for organ allocation is adjusted to give higher priority to patients with HCC who are at increased risks for exclusion based on tumor characteristics. [125] This system is designed to reduce the waiting time and thus the rate of dropout from the waiting list. If the waiting time for liver transplantation can be significantly reduced for patients with HCC, it may also be possible to modestly expand the tumor size criteria, as proposed in a study from the authors’ institution, [120] without compromising survival after liver transplantation.

Hemochromatosis and Other Metabolic Liver Diseases Patients with hereditary hemochromatosis are at increased risks for the development of cirrhosis, liver cancer, cardiomyopathy, and diabetes. [126] Several studies have indicated a less favorable outcome following liver transplantation for patients with hemochromatosis. [127], [128], [129] and [130] In a retrospective analysis of 37 patients who underwent liver transplantation for hemochromatosis, [127] the 1- and 5-year survival rates were only 58% and 40%, respectively, compared to 62% 5-year survival for all patients reported to the UNOS registry over the same period. Infectious complications were the cause of 53% of all deaths within the first year after liver transplantation, whereas cardiac complications accounted for 50% of the late mortality. [127] A high incidence of hepatic malignancies (27%) was also observed at the time of liver transplantation. [128] The diagnosis of hemochromatosis was often not made until after liver transplantation, [129], [130] a factor that might have contributed to the poor outcome. Once the diagnosis of hemochromatosis is confirmed, early initiation of phlebotomy may diminish the likelihood of posttransplant cardiac complications. Thorough cardiac evaluations with right heart catheterization and possibly endomyocardial biopsy are recommended before liver transplantation. [130] Cardiac dysfunction due to excessive iron infiltration may preclude liver transplantation in view of the high perioperative and late postoperative cardiac mortality. [127], [130] The hallmark of Wilson disease, a relatively rare inherited disease of copper metabolism, is its highly variable clinical presentation. [131] The diagnosis of Wilson disease is suggested by a young age at presentation and the presence of low serum ceruloplasmin, neuropsychiatric symptoms, and Kayser-Fleischer rings on slit-lamp ophthalmologic examination, [131] although atypical clinical presentations and the absence of these classic signs are also common. Wilson disease may present as fulminant hepatic failure with a high mortality when managed with medical treatment alone. [132], [133] Such patients can be listed at UNOS status 1 for liver transplantation. [48] The presence of a Coombs-negative hemolytic anemia and a serum alkaline phosphatase to serum bilirubin ratio of less than 2 is suggestive, but not diagnostic, of fulminant Wilson disease. [129] Nazer and coworkers have also developed a useful prognostic score (based on serum bilirubin, serum transaminase levels, and prothrombin time) which stratifies patients with Wilson disease according to the risk of death without liver transplantation. [134] The neurological and psychiatric manifestations of Wilson disease improve in most but not all patients after liver transplantation. [129], [133] A long list of other diseases in the category of inborn errors of metabolism has been successfully treated with liver trans plantation. [55], [129] The indication for liver transplantation is end- stage cirrhosis caused by some of these diseases, including a-1-antitrypsin deficiency, [135] glycogen storage disease (types I and IV), [136] cystic fibrosis, [137] erythropoietic protoporphyria, [138] and tyrosinemia. [139] At the other end of the spectrum, liver transplantation has been performed solely for the correction of nonhepatic manifestations of certain metabolic diseases in which the genetic defect is expressed in the liver in the absence of significant hepatic dysfunction. Examples include Crigler-Najjar syndrome, [140] primary hyperoxaluria, [141] familial amyloid polyneuropathy, [142] and severe familial hypercholesterolemia. [143]

Autoimmune Hepatitis Progression of autoimmune hepatitis to cirrhosis and liver failure requiring liver transplantation is usually seen in patients refractory to conventional immunosuppressive therapy and those who present late in the course of disease without prior diagnostic confirmation or appropriate therapy. [144] Although the outcome of liver transplantation for autoimmune hepatitis is generally excellent, it is now increasingly recognized that there is a risk for disease recurrence. [145], [146] Bearing in mind a significant limitation with respect to the small number of patients in single-center reports, and the differences in the definition of recurrent disease based on histology, autoantibodies, liver biochemical profiles, or a combination of these factors, the recurrent rate is estimated to be between 20% to 30%. [146], [147], [148], [149], [150], [151] and [152] The diagnostic criteria for recurrent autoimmune hepatitis have not been well established. The use of monitoring autoantibodies after liver transplantation is unclear, and most studies have found that these markers persist to some extent in all patients transplanted for autoimmune hepatitis. Published data on titer or persistence of the autoantibodies are currently insufficient for predicting recurrence or disease activity after liver transplantation. [146] Histological features of recurrent autoimmune hepatitis may be difficult to differentiate from that in acute cellular rejection. Risk factors for recurrent disease include suboptimal immunosuppression and HLA haplotypes DR3 and DR4. [146] Recurrent autoimmune hepatitis rarely develops in the first year after liver transplantation, presumably because of the higher level of immunosuppression used during this period. [146], [147] The course of recurrent autoimmune hepatitis is typically indolent, and the histological features for recurrent disease rapidly resolve in most cases when adequate immunosuppression is restored. [146] One study, however, reported graft failure requiring re-transplantation in 6 of 24 patients with recurrent autoimmune hepatitis and more than 1 year of follow-up. [147]

Nonalcoholic Fatty Liver Disease Nonalcoholic fatty liver disease (NAFLD) includes a spectrum of histological lesions ranging from steatosis to nonalcoholic steatohepatitis (NASH), [153] the latter being increasingly recognized as a significant cause of cirrhosis. [153], [154] and [155] Many patients with cryptogenic cirrhosis may, in

fact, have NAFLD as the etiology. [154] Several reports have suggested that NAFLD may recur after liver transplantation. [155], [156], [157] and [158] This phenomenon has also been described in patients with cryptogenic cirrhosis following liver transplantation, with the presumption that NAFLD is the unrecognized underlying cause of their liver disease prior to liver transplantation. [159] Recurrent NAFLD after liver transplantation highlights the importance of host-dependent factors, as opposed to liver-dependent factors, in the pathogenesis of the disease.

Cholestatic Liver Disease Patients with cholestatic liver disease, chiefly primary biliary cirrhosis (PBC) and primary sclerosing cholangitis (PSC), have traditionally been considered the best candidates for liver transplantation based on superior survival statistics. [3], [160] In addition to the CTP score (Table 117-1), other prognostic models have also been used to predict survival and optimal timing for liver transplantation in these patients. [160] Quality-of-life indications for liver transplantation in patients with cholestatic liver disease include pruritus [161] and metabolic bone disease with osteopenia predisposing to fractures. [162] Intractable pruritus in the absence of overt liver failure has been considered an acceptable indication for liver transplantation. No therapy other than liver transplantation has been shown to increase bone mineralization in patients with cholestatic liver disease. [163] Although bone loss continues in the first 3 to 6 months after liver transplantation, bone density then returns back to pretransplant levels at 1 year, and may be restored toward normal within 2 to 3 years after liver transplantation. [163] Liver transplantation is also effective for xanthomatous neuropathy, an unusual complication of cholestatic liver disease. Biliary atresia is the most common indication for pediatric liver transplantation (see Fig. 117-3).

Primary biliary cirrhosis. Primary biliary cirrhosis (PBC) is characterized by progressive destructions of small intrahepatic bile ducts leading to eventual cirrhosis and liver failure. [164] The Mayo Clinic risk score has been validated and widely used as a prognostic model for predicting survival without therapeutic intervention. [165], [166] This model is based on five parameters: serum bilirubin, serum albumin, age, prothrombin time, and edema. Serum bilirubin is the most important prognostic variable for PBC. Bilirubin level above 10 mg/dL is associated with a particularly poor prognosis. [167] Liver transplantation has clearly been shown to improve survival for PBC as compared with the survival predicted by the Mayo Clinic model. [160], [168] Referral for liver transplantation should be initiated when the 1-year probability for survival falls below 90%, which correlates with a Mayo risk score of 6.0 or higher. [160] For practical purposes, a CTP score of 7 or higher is more widely used to determine eligibility for listing. The Mayo risk score now plays no role in identifying patients with PBC in need of liver transplantation and prioritizing them on the waiting list. This function is now served by the MELD scoring system [50] (see Table 117-2). Ursodeoxycholic acid (UDCA) improves the biochemical profiles, especially the bilirubin level in patients with PBC. Some concerns were raised that using the Mayo risk score and possibly also the MELD score in UDCA-treated patients might underestimate their mortality. [47], [169] Although existing data are still inconclusive, UDCA may slightly prolong the disease course before needing liver transplantation. [170] Whether PBC recurs after liver transplantation has been a controversial issue. Data emerging in the last few years appear to suggest that recurrence does happen in some patients. [145] The incidence of recurrence among the larger studies is reported in the 10% to 20% range, [145], [171], [172], [173], [174] and [175] with some centers reporting recurrence rates approaching 50% at 10 years. [176] Ambiguity in defining recurrence may explain the variations in the reported incidence of recurrence. The diagnosis of recurrent PBC can only be made based on histological criteria, which may be difficult to differentiate from other causes of bile duct damage in the allograft such as acute or chronic rejection and biliary complications. To date, disease recurrence has had little impact on the quality of life or graft survival, although a few patients have required re-transplantation. [145], [176] Preliminary data with a small number of patients have suggested that recurrence occurs earlier and more frequently in patients receiving tacrolimus as opposed to cyclosporine. [145], [176] This observation needs to be confirmed in larger studies. Whether UDCA alters the rate of progression of recurrent PBC has not been studied.

Primary sclerosing cholangitis. PSC typically involves both the intrahepatic and extrahepatic bile ducts; with segmental biliary strictures, beading, and dilations. [177] A number of prognostic models have been proposed for predicting survival in PSC, [160], [177] including the Mayo risk score which is based on four variables: age, serum bilirubin level, splenomegaly, and cirrhosis on liver biopsy. [178] The CTP score and MELD score now serve the function of determining eligibility for liver transplant listing for patients with PSC and prioritizing them on the waiting list, respectively. [50] No medical therapy, including UDCA, [179] has clearly been proven to be effective in improving the natural history of PSC. [177] High-dose UDCA may help some patients with earlier disease. [180] One of the indications for liver transplantation in PSC is recurrent bacterial cholangitis, which is often associated with bile-duct stones or obstructing strictures. [177] Endoscopic stenting or balloon dilation of strictures may benefit some patients, [177], [181], [182] although controlled data are limited. Because of extensive scarring of the diseased common bile duct, anastomosis to a Roux-limb of jejunum is the standard surgical approach at liver transplantation. Patients with PSC are also at risk for developing cholangiocarcinoma, with an estimated incidence of up to 20% to 30% at the time of liver transplantation. [183], [184] Establishing the diagnosis of cholangiocarcinoma in patients with PSC is often very difficult. Neither brush cytology [185] nor serum tumor markers including CA 19-9 [186] are reliable diagnostic tools. Preliminary data on positron emission tomography for detecting small cholangiocarcinoma have shown some promise. [187] Survival after liver transplantation is very poor when cholangiocarcinoma is present. [184], [188] About 70% of patients with PSC also have inflammatory bowel disease, [189] and are at risk for developing co- lorectal cancer. There have been several reports of colorectal cancer diagnosed after liver transplantation in patients with PSC and ulcerative colitis. [190], [191] and [192] Continued surveillance by yearly colonoscopy after liver transplantation to detect dysplasia is recommended. The reported incidence of recurrent PSC after liver transplantation has ranged from 10% to 40%. [193], [194], [195] and [196] Distinguishing cholangiographic features of recurrent disease from some other conditions, such as ischemic strictures related to prolonged cold ischemia time, ABO incompatibility, and hepatic artery thrombosis can be extremely difficult. [145] Compelling evidence for recurrent disease comes from the reports by Graziadei and associates, [195], [196] in which nonanastomotic strictures of intrahepatic and extrahepatic bile ducts developing more than 90 days after liver transplantation defined cholangiographic recurrence, whereas fibro-obliterative lesions and fibrous cholangitis were consistent with histological recurrence. The incidence of recurrence was 18% among 120 patients with PSC receiving liver transplantation, versus 1% of a control group of 415 patients transplanted for other indications. [195] Patients with hepatic artery thrombosis, chronic rejection, ABO incompatibility, and anastomotic strictures were excluded in the study. [195] Recurrent PSC does not appear to impact adversely on patient or graft survival over the intermediate term. [196]

Fulminant Hepatic Failure Fulminant hepatic failure is traditionally defined as the development of hepatic encephalopathy within 8 weeks after the de novo onset of jaundice. [197] Subacute or subfulminant hepatic failure is observed in a small subgroup of patients in which encephalopathy develops later, up to 26 weeks after the onset of illness. [198] The predominant cause of fulminant hepatic failure is acetaminophen-induced hepatic injury. [199], [200] Idiopathic liver failure possibly from an unidentified virus, other types of known viral hepatitis (hepatitis A, B, D, and E but not hepatitis C), and idiosyncratic drug-induced liver injury comprise the next most common group, [199], [200] although differences exist between Europe, the United States, and Asia. [199], [200] and [201] Patients with fulminant hepatic failure typically have no preexisting liver disease, but there are well-recognized examples of hitherto asymptomatic chronic liver disease, such as Wilson disease, presenting as fulminant hepatic failure. [132] Historically, only 20% to 30% of patients with fulminant hepatic failure have survived without liver transplantation. [197], [200] A variety of medical therapies, including charcoal hemoperfusion, hemodialysis, plasmapheresis, prostaglandin analogs, and corticosteroids have failed to improve outcome in controlled clinical trials. [202] The well-validated King's College prognostic criteria identify the subgroup of patients at highest risk of dying who should undergo liver transplantation as soon as possible [203] (Table 117-4). A coagulation factor V level of less than 20% also appears to have significant discriminatory prognostic value in nonacetaminophen-induced fulminant hepatic failure. [204], [205] Early referral of patients with fulminant liver failure to a liver transplant center is of paramount importance, and these patients should be listed for liver transplantation with the highest priority (UNOS status 1) as soon as they develop clinically evident hepatic encephalopathy. Rapid deterioration and death, most commonly from cerebral edema and sepsis, can occur within days after the development of stage 3 or 4 encephalopathy. [202] Intracranial pressure monitoring is commonly instituted in patients with grade 4 hepatic encephalopathy. [206], [207] and [208] At the authors' institution, patients deteriorating beyond stage 3 hepatic encephalopathy are electively intubated and ventilated with adequate sedation both to protect the airway and to prevent surges in intracranial pressure related to psychomotor agitation. Sustained elevation of intracranial pressure with cerebral perfusion pressure (mean arterial pressure-intracranial pressure) below 40 mm Hg for at least 2 hours despite treatment with mannitol and pentobarbital is associated with a very poor prognosis secondary to irreversible neurological damage and precludes liver transplantation. [206], [207] However, exceptions to this observation have been reported. [208] Monitor dysfunction must be carefully excluded since false readings may result in erroneous decisions with respect to the patient's candidacy for liver transplantation. [208] The usual protocol for patients in hepatic coma with intracranial monitoring is to maintain cerebral perfusion pressure =50 mm Hg and intracranial pressure below 20 mm Hg prior to and during liver transplantation to ensure full neurological recovery after surgery. [208]

ACETOMINOPHEN-INDUCED FULMINANT LIVER FAILURE	
Prognostic Factors	*Predicted Mortality Rate (%)*
pH < 7.3	95
Grade 3 or 4 encephalopathy	67
Prothrombin time >35 sec	
Serum creatinine >3.5 mg/dL	
Liver transplantation is indicated for pH <7.3 irrespective of the grade of encephalopathy; or prothrombin time >35 sec and creatinine >3.5 mg/dL with grade 3 or 4 encephalopathy.	
NONACETAMINOPHEN-INDUCED FULMINANT LIVER FAILURE	
Prognostic Factors	*Predicted Mortality Rate (%)*
Prothrombin time >35 sec	100
Age < 10 or > 40	96
Bilirubin >17.5 mg/dL	93
Jaundice for >7 d before encephalopathy	97
Unfavorable etiology	90
Idiosyncratic drug reactions	
Wilson disease	
Non-A, non-B hepatitis	
Halothane hepatitis	
Liver transplantation is indicated for prothrombin time >35 sec or any three of the remaining variables irrespective of the grade of encephalopathy.	

Data from ref. 203. The prothrombin time is converted to equivalent values in the United States.

TABLE 117-4 King's College Prognostic Criteria for Fulminant Hepatic Failure

The 1-year survival rates following liver transplantation for fulminant hepatic failure have traditionally ranged from 60% to 70%, [199], [207] while a much higher 1-year survival rate of 92% was reported from the authors' institution where aggressive monitoring and treatment of intracranial hypertension had been the rule. [209] More recent data from UNOS also revealed an improved overall patient survival rate approaching 90% at 1 year in the United States, [2] perhaps reflecting advances in the care of critically ill patients and improved patient selection. Liver transplantation for fulminant hepatic failure is, however, limited by the severe shortage of cadaveric organ donors and by the very brief time available to obtain suitable organs. There has been considerable interest in the use of extracorporeal liver support, including the bioartificial liver-assist device (BAL; Circe Biomedical, Inc., Lexington, MA) and the extracorporeal liver-assist Device (ELAD; Hepatix, Inc., Houston, TX), for providing temporary liver support and serving as a "bridge" to liver transplantation. [199] These devices differ in the nature of the hepatocyte component (porcine vs. human hepatoblastoma cell line) and bioreactor design. [199] Preliminary results of two controlled studies were, however, disappointing in that no survival benefit could be demonstrated among patients treated with these devices compared with those receiving supportive care only prior to liver transplantation, [210], [211] thus dampening the initial enthusiasm regarding this treatment. Extracorporeal liver perfusion using cadaveric livers judged unsuitable for transplantation, [212] or porcine livers [213] have been reported in a small number of patients. Auxiliary orthotopic liver transplantation [214] is a special procedure in which part of the native liver is left in situ and the reduced-size graft provides temporary support until subsequent regeneration of the native liver. Thereafter, the auxiliary graft is either surgically removed or allowed to atrophy following withdrawal of immunosuppression. In a series of 30 patients from Europe who underwent this procedure for fulminant hepatic failure, [214] 63% survived and 43% had restored normal native liver function after a mean follow-up of 18 months. Although auxiliary liver transplantation has the advantage of not subjecting patients to lifelong immunosuppression with its attendant risks, technical difficulties have prevented its widespread use for fulminant hepatic failure.

EVALUATION FOR LIVER TRANSPLANTATION

Although the approach to the evaluation of patients for liver transplantation may vary according to the individual liver transplantation center, the principles are generally similar (Fig. 117-4). The evaluation process typically involves a multidisciplinary team including hepatologists, transplant surgeons, transplant nurse coordinators, social workers, and individuals with expertise in substance abuse issues. The use of other consultants varies according to individual transplant centers and patient needs. The main roles of the transplant hepatologist are to

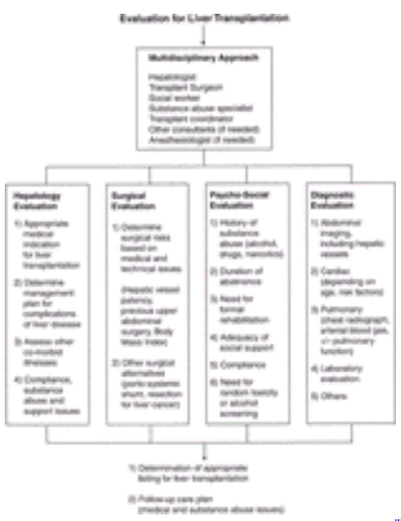

FIGURE 117-4. Principles in the evaluation of potential candidates for liver transplantation.

- confirm that the patient has appropriate medical indications for liver transplantation
- implement a plan for the management of the complications of liver disease
- evaluate disease-specific issues that may potentially impact on outcome after liver transplantation
- assess other co-morbid conditions and possible contraindications to liver transplantation.

Surgical risks and technical considerations for liver transplantation need to be carefully evaluated, primarily by the transplant surgeon. Extensive portal and mesenteric venous thrombosis, previous abdominal surgery near the hepatic hilum, and severe obesity can make the transplant operation difficult, if not impossible. In some patients with a TIPS, extension or migration of the stent into the extrahepatic portal vein, the hepatic vein, or vena cava may compromise the success of liver transplantation. [28] Patients should be fully informed of the risks for liver transplant surgery, the possibility and likelihood of recurrent disease after liver transplantation, and the potential alternatives to liver transplantation. For instance, some patients with well-compensated or mildly decompensated cirrhosis may benefit from portal decompressive surgery for refractory variceal bleeding. Surgical resection for HCC may be an alternative to liver transplantation in some patients with well-compensated cirrhosis, a single lesion less than 5 cm, and no portal hypertension. [120] Psychosocial assessment is an integral part of the liver transplant evaluation process to address substance abuse issues, the risk for recidivism, compliance, and adequacy of social support. In additional to a thorough history and physical examination, an abdominal imaging study (ultrasonography with Doppler study, computed tomography, or magnetic resonance imaging) to exclude intra- or extrahepatic tumors and to evaluate hepatic vessel patency, and laboratory tests are part of the routine evaluation process.

Cardiac evaluation prior to liver transplantation is intended to exclude coronary artery disease, valvular heart disease, and cardiac failure from various etiologies, the extent of which may also depend on the cardiac risk factors present in the individual patient. Special attention should be given to patients with alcoholic liver disease and hemochromatosis, since they are at increased risk for cardiomyopathy. A chest radiograph and arterial blood gas analysis are routinely done in most transplant centers. Patients with a1-antitrypsin deficiency, chronic tobacco use, or known or suspected intrinsic lung diseases should undergo full pulmonary function testing. It is also important to identify two pathophysiologically distinct pulmonary syndromes that are uncommon but clinically important among patients with cirrhosis. These are hepatopulmonary syndrome and portopulmonary hypertension. [215] Hepatopulmonary syndrome is characterized by arterial hypoxemia (PaO_2 <70 mm Hg or alveolar-arterial gradient >20 mm Hg) and pulmonary capillary bed dilation supported by contrast echocardiography and technetium macroaggregated albumin lung scanning. [215] Hepatopulmonary syndrome is reversible after liver transplantation, but patients with a baseline PaO_2 of =50 mm Hg undergoing liver transplantation have a perioperative mortality of about 30%. [215] Portopulmonary hypertension is defined as a mean pulmonary arterial pressure (PAP) of more than 25 mm Hg and pulmonary capillary wedge pressure of less than 15 mm Hg. [216] The perioperative mortality associated with liver transplantation was reported to be 50% among patients with moderate portopulmonary hypertension (mean PAP between 35 and 50 mm Hg) and 100% for those with severe portopulmonary hypertension (mean PAP of =50 mm Hg). [216] Right heart catheterization is indicated to confirm the diagnosis when the estimated mean PAP is =35 mm Hg by echocardiography.

Contraindications to Liver Transplantation

The commonly cited absolute and relative contraindications to liver transplantation, which also reflect the authors' opinion rather than a true consensus, are listed in Table 117-5. The absolute contraindications include severe co-morbid medical illness, such as cardiac and pulmonary diseases that are not reversible and adversely impact the patient's short-term life expectancy, and extrahepatic malignancies excluding certain skin cancers. Patients with advanced HCC and cholangiocarcinoma are generally excluded from liver transplantation, although some centers have performed liver transplantation on these patients under certain experimental treatment protocols. As already described, patients with severe portopulmonary hypertension should not undergo liver transplantation due to an unacceptably high perioperative mortality. [215], [216] At the authors' institution and some other transplant centers, selected patients with moderate or severe pulmonary hypertension are often placed on intravenous epoprostenol (prostacyclin) [217] under an experimental protocol in an attempt to decrease the PAP before liver transplantation. Systemic infections must be adequately treated before liver transplantation. Psychiatric and psychosocial contraindications include active substance abuse, high recidivism risk, noncompliance, and poorly controlled psychiatric illness. Poor social support is also a relative contraindication to liver transplantation. Technical contraindications include extensive thrombosis involving both the portal and mesenteric veins. Severe obesity, defined as a body mass index of = 35, was shown in a recent analysis of UNOS data to be associated with a significantly worse survival after liver transplantation. [218] Thus it is important for these patients to lose weight prior to liver transplantation.

Absolute contraindications
Severe, irreversible co-morbid medical illnesses that adversely impact short-term life expectancy
Severe pulmonary hypertension (mean PAP ≥50 mm Hg)*
Extrahepatic malignancy (excluding some skin cancers)
Extensive hepatocellular carcinoma or with macrovascular or lymph node invasion*
Cholangiocarcinoma*
Uncontrolled systemic sepsis
Extensive portal vein and mesenteric vein thrombosis
Active alcohol or drug abuse
Noncompliance
Unacceptable risks for recidivism from drugs or alcohol
Severe, uncontrolled psychiatric disease
AIDS (HIV)*
Relative contraindications
Moderate pulmonary hypertension (mean PAP between 35 and 50 mm Hg)*
Severe hepatopulmonary syndrome with Pa_{O_2} of ≤50 mm Hg
Severe obesity (body mass index ≥ 35)
Poor social support
Advanced age (≥70 y)

AIDS, acquired immunodeficiency syndrome; HIV, human immunodeficiency virus; PAP, pulmonary arterial pressure.

*Liver transplantation has been performed in some centers under an experimental treatment protocol.

TABLE 117-5 Contraindications to Liver Transplantation

It is generally agreed that patients with established acquired immunodeficiency syndrome (AIDS) should not be considered for liver transplantation. However, individuals infected with the human immunodeficiency virus (HIV) without an AIDS-defining illness now survive longer with current advances in antiviral therapy. Whether they should be considered for liver transplantation is a highly controversial issue. Data on liver transplantation for HIV-infected patients are very limited. A number of early deaths from aggressive hepatitis C recurrence in co-infected patients who underwent liver transplantation have been reported. [219] At present, liver transplantation should not be performed in HIV-infected individuals outside of clinical trial protocols.

POSTTRANSPLANT MANAGEMENT

Factors Affecting Allograft Function and Survival

Immunosuppression and Rejection The calcineurin inhibitors, first cyclosporine and subsequently tacrolimus, have been the mainstay of immunosuppression for liver transplantation for two decades. [220], [221] Immunologic mechanisms of action for these and other immunosuppressive drugs used today are described in greater detail elsewhere. [220], [221] The first available formulation of cyclosporine (Sandimmune®) is dependent on bile for absorption leading to erratic absorption and suboptimal drug levels in the early posttransplant period when given orally. The introduction of a microemulsion formulation of cyclosporine (Neoral®) led to improved bioavailability and absorption. The usual induction immunosuppressive regimen in the early posttransplant period is high-dose intravenous corticosteroid therapy with tapering to oral doses of 20 mg per day by the end of the first week. Although the calcineurin inhibitors are usually started within the first 2 days after liver transplantation, they are nephrotoxic and there is a need to allow renal function to recover after liver transplantation before initiating these drugs in patients with impaired renal function. Daclizumab, an anti-interleukin-2 (IL-2) receptor antibody, and rapamycin (also known as sirolimus) are two new immunosuppressive agents that are nonnephrotoxic and can be used in place of the calcineurin inhibitors in this setting to reduce the risk for rejection. Defining the role of these new agents in liver transplantation either alone or in combination with a calcineurin inhibitor is currently the goal of active investigations. [220], [221] The typical maintenance immunosuppressive regimen consists of a calcineurin inhibitor and prednisone, with or without azathioprine or mycophenolate mofetil. Mycophenolate mofetil is a relatively new immunosuppressive agent, which works by reversible inhibition of inosine monophosphate dehydrogenase. This drug appears to be more potent than azathioprine with more specific enzyme inhibition. At the authors' institution, a primary regimen of tacrolimus, mycophenolate mofetil, and prednisone is used. The 5-year patient and graft survival rates are comparable for cyclosporin- and tacrolimus-based immunosuppressive regimen. [106] Although management of immunosuppression differs according to the individual transplant center's preference, most patients can be maintained on lower doses of immunosuppression with time and in the absence of acute rejection. The ultimate level of maintenance immunosuppression also depends on the pretransplant diagnosis of liver disease—lower level of maintenance immunosuppression in general for chronic hepatitis B or C and higher for patients with autoimmune hepatitis, primary biliary cirrhosis, and possibly fulminant hepatic failure. Prednisone can be withdrawn with close monitoring in some patients at 6 months to 1 year after liver transplantation. [222] Acute cellular rejection occurs in 30% to 60% of all liver transplant recipients within the first year after transplantation, [220], [221], [222], [223] and [224] and is thought to be secondary to mismatches in class I and II major histocompatibility complex (MHC) antigens. [225] In the study from the NIDDK Liver Transplant Database, [224] acute rejection was not shown to exert an overall detrimental impact on patient and graft survival. Only the subgroup of patients with a severe episode of rejection defined by histological criteria was at higher risk for death or re-transplantation. [224] These findings were in contrast to that in renal transplant recipients, in whom even a single acute rejection episode was associated with a significantly reduced graft survival. In the past decade, advances in immunosuppressive drugs have resulted in a progressive decline in the incidence of acute rejection episodes after liver transplantation. [226] A primary immunosuppressive regimen of a calcineurin inhibitor, mycophenolate mofetil, and prednisone is associated with a rejection rate of only about 30%. [226] Acute rejection is most commonly seen within the first 6 weeks, especially between day 4 and 14 after liver transplantation. [220], [221], [222], [223] and [224] The incidence of acute rejection generally decreases with time after liver transplantation. The diagnosis is suggested by nonspecific elevations in the liver enzymes. Fever, leukocytosis, and eosinophilia may be present, but are also nonspecific. The other diagnostic considerations for elevated liver enzymes in the early postoperative period include "preservation/reperfusion" injury, hepatic artery thrombosis, and biliary complications. Histologically, acute cellular rejection is characterized by the triad of portal mixed cellular inflammation with predominantly lymphocytes, bile ductular injury, and subendothelial inflammation of the portal or terminal hepatic venules (endotheliitis). [227] At least two of the three features are required for the diagnosis. [227] Eosinophils are commonly present in the mixed cellular infiltrate although this is a nonspecific feature. The initial treatment for acute cellular rejection is usually high-dose methylprednisolone at 1000 mg for 2 doses one day apart. This is followed by a 6-day taper of oral prednisone from 200 mg to 20 mg per day for a total "re-cycle" dose of 3 g. Some centers use a total of only 2 g of methylprednisolone. Steroid-resistant rejection occurs in 10% to 20% of patients treated with cyclosporine, but the incidence is reported to be less than 10% with a tacrolimus-based immunosuppressive regimen. [106], [223] Muromonab-CD3 (also known as OKT3), a murine monoclonal antibody to the CD3 receptor on T-lymphocytes, is most commonly used for the treatment of steroid-resistant acute cellular rejection. Muromonab-CD3 use is almost always accompanied by a cytokine-release syndrome, which includes fever, chills, and other symptoms within the first hour after administration. This resolves within 4 to 6 hours. Patients are given methylprednisolone, antipyretics, and antihistamines as a premedication regimen about 1 hour prior to receiving muromonab-CD3 to relieve some of these symptoms. Aseptic meningitis is associated with muromonab-CD3 in 3% to 5% of cases. The use of muromonab-CD3 is associated with an increased incidence of serious infections, particularly cytomegalovirus (CMV), and rarely, a capillary leak syndrome that can present as pulmonary edema. Antilymphocyte globulin may also be effective in steroid-resistant rejection. Hyperacute rejection is a rare and devastating event mediated by preformed antibodies that bind to allogenic epitopes on donor endothelial cells, leading to rapid graft failure within hours to days after liver transplantation. [225] It is most commonly due to ABO incompatibility. Chronic ductopenic rejection [228] is seen in about 10% of all liver transplant recipients, usually occurring beyond 60 days after transplantation. It is characterized by progressive loss of interlobular bile ducts and an obliterative arteriopathy. [228] Chronic rejection usually develops after multiple episodes of acute rejection, but may be seen after

an unresolved episode of acute rejection or indolently over months to years after liver transplantation without a definite rejection episode. Although chronic rejection is generally irreversible and requires re-transplantation, some cases of reversal after switching from cyclosporine to tacrolimus have been reported. [228], [229] Chronic rejection may also result in diffuse stricturing of bile ducts in the allograft.

Vascular Complications Hepatic artery thrombosis occurs in about 7% of all adult liver transplants and in 10% to 40% of pediatric transplants. [226], [230] Hepatic artery thrombosis usually occurs within the first 30 days after liver transplantation. Risk factors for hepatic artery thrombosis include small hepatic arterial size, complex anatomy, the need for extension grafts to perform arterial anastomosis, prolonged ischemic time, and a hypercoagulable state. [226] The cause of hepatic artery thrombosis is often unknown. [231] Since the bile ducts receive blood supply exclusively from the hepatic artery early after liver transplantation, hepatic artery thrombosis results in diffuse biliary strictures, often associated with cholangitis, and less frequently, hepatic abscesses. If hepatic artery thrombosis is identified early after liver transplantation, immediate arterial reanastomosis may reverse the long-term consequences to the allograft. [231] Nevertheless, re-transplantation is ultimately necessary in almost all cases. [226] Some patients, particularly in whom this complication occurs later following liver transplantation, may benefit from long-term, repeated biliary stenting and dilation. The second most common vascular complication is portal vein thrombosis, which may present with ascites, recurrent variceal bleeding, or hepatic encephalopathy. [226] In some cases, this condition is only associated with asymptomatic liver test abnormalities. Patients at risk for portal vein thrombosis include those requiring interposition of grafting during portal vein anastomosis or those with a history of hypercoaguable state or portosystemic surgical shunts. Balloon dilation and stenting may be effective in some cases of portal vein stenosis, thus abrogating the need for surgical revision or re-transplantation. [232] Thrombosis or stenosis of the hepatic vein or the inferior vena cava is uncommon. This complication has been reported in patients with recurrent Budd-Chiari syndrome following liver transplantation [233] and recipients of partial or split liver transplants either from a cadaveric or living donor. [234]

Biliary Complications Choledochocholedochostomy (end-to-end anastomosis) is the most frequently used biliary anastomosis in adult liver transplantation. A T-tube is often placed across the anastomosis to decrease the risk of anastomotic stricture. The T-tube is usually clamped between day 3 to 7 after liver transplantation and removed 6 weeks to 6 months later as the tract has matured. Bile leak at the T-tube exit site is nevertheless a frequent occurrence. [235], [236] At the authors' institution and some other transplant centers, the standard T-tube placement in end-to-end anastomosis has been abandoned and replaced by direct end-to-end anastomosis without an associated increase in adverse consequences. [237] The other type of anastomosis is the Roux-en- choledochojejunostomy, which is used mainly in patients with preexisting bile duct abnormalities such as PSC and biliary atresia or in whom technical difficulty is encountered in fashioning a conventional duct-to-duct anastomosis. The overall incidence of biliary complications after liver transplantation falls within the 10% to 15% range. [238] In a series of almost 1800 patients reported from the University of Pittsburgh, [239] 80% of biliary complications were identified within the first 6 months after liver transplantation, and one third were diagnosed in the first month. Bile leaks and biliary strictures account for 80% of biliary complications. [238], [239] and [240] Most of the biliary complications after liver transplantation can now be managed by nonsurgical techniques. [235], [236], [238] Bile leaks occur most commonly at the anastomotic site, T-tube exit site, or cut edge of reduced-size liver grafts. Bile leaks may result in a large biloma, bile peritonitis, and super-infection. Management with biliary stent placement and percutaneous drainage of biloma is successful in most cases. [235] Surgical revision is indicated when a large leak is present with severe disruption of the bile duct. Bile leaks from Roux-en- anastomosis more often require surgical revision. [235] Biliary strictures occur most often at the site of anastomosis, possibly related to poor local blood supply from hepatic artery branches, and edema. Endoscopic biliary stenting or balloon dilation is often adequate without surgical intervention. [238] A more difficult problem is the diffuse, "ischemic" type of strictures, which are most often due to hepatic artery thrombosis, ABO-incompatible allografts, or possibly prolonged, cold ischemia before graft revascularization. [241] However, no specific underlying cause can be identified in many cases. In summary, although the overall impact of biliary complications on mortality is small due to improvements in patient management, as well as diagnostic and therapeutic techniques for these complications, [238] they are still common and potentially serious, resulting in graft loss in 1% to 3% of liver transplant recipients. [238]

Recurrent Disease in the Allograft Disease-specific recurrence following liver transplantation has already been reviewed. In general, recurrent autoimmune hepatitis and cholestatic liver disease have a minor impact on graft survival, [145] whereas recurrent viral hepatitis has been a highly important cause of graft failure within the first 5 years following liver transplantation. [226] The use of HBIG, and potentially the addition of a nucleoside or nucleotide analog, have diminished the adverse impact of hepatitis B recurrence on graft survival. [81] The magnitude of the adverse impact of recurrent hepatitis C on graft survival, on the other hand, has become increasingly evident in the last few years. Interferon plus ribavirin therapy remains the only proven treatment for hepatitis C, but this treatment has been suboptimal thus far for the control of recurrent disease after liver transplantation. [110], [111] and [112] The problem related to liver transplantation for hepatitis C infection will likely worsen as many patients with hepatitis C infection are now at risk for cirrhosis, liver failure, and HCC as the disease advances to the 3rd and 4th decade following acute exposure of the chronically infected population. Liver re-transplantation is generally associated with a worse survival compared to primary liver transplantation, [242] and further depletes a limited donor pool. Despite improved outcome in recent years for liver transplantation among patients with HCC, [120] many patients still succumb to recurrent HCC, which tends to be highly aggressive in immunosuppressed, posttransplant patients and is rarely curable by surgical resection or re-transplantation. Better understanding of the mechanisms and predictors of tumor recurrence, refinements of the selection criteria for liver transplantation, and modification of organ allocation policies to reduce waiting time and the pretransplant risk of tumor dissemination may further improve the outcome for HCC. The course of recurrent NAFLD has not been well characterized due to the small number of patients studied. It appears that the course is relatively benign, although some cases progress to cirrhosis and graft failure. Finally, alcoholic liver disease may recur with heavy alcohol intake in the posttransplant period. [62] Surreptitious alcohol consumption should be considered in the diagnostic evaluation of unexplained graft dysfunction, especially among those with a history of heavy alcohol consumption prior to liver transplantation.

Medical Complications of Immunosuppression

The immunosuppressive drugs used today for liver transplantation have many inherent risks. Some of the well-known direct adverse effects of these immunosuppressive drugs are listed in Table 117-6. Nephrotoxicity, neurotoxicity, impaired glucose metabolism, and gastrointestinal disturbances are common with both cyclosporine and tacrolimus (calcineurin inhibitors). Both neuropathy and gastrointestinal adverse effects are more common with tacrolimus. Hyperkalemia is also more often seen with tacrolimus and is usually managed with fludrocortisone. [222] Hypertension and altered lipid profiles are more frequently associated with cyclosporine than tacrolimus. Hirsutism and gingival hyperplasia are specifically associated with cyclosporine and not tacrolimus. [223] It is important to recognize that a long list of drugs can interfere with the metabolism of the calcineurin inhibitors (Table 117-7). Most interactions occur as a result of either competitive inhibition or induction of metabolism at the level of cytochrome P450 3A4. Increased drug levels can lead to toxicities whereas decreased drug levels can precipitate rejection. A major adverse effect of both azathioprine and mycophenolate mofetil is bone marrow suppression. Gastrointestinal toxicity, including a hemorrhagic gastritis is also associated with mycophenolate mofetil. Frequent side effects of rapamycin include hyperlipidemia, thrombocytopenia, and leukopenia. Interstitial pneumonitis has been linked to the use of rapamycin.

CORTICOSTEROIDS	AZATHIOPRINE	MYCOPHENOLATE MOFETIL	RAPAMYCIN
Infection	Leukopenia	Leukopenia	Leukopenia
Poor wound healing	Thrombocytopenia	Thrombocytopenia	Thrombocytopenia
Osteonecrosis	Hepatotoxicity	Gastrointestinal	Hyperlipidemia
Cataracts	Gastrointestinal	Nausea/vomiting	Interstitial pneumonitis
Diabetes	Pancreatitis	Abdominal pain	
Cushingoid habitus	Cough	Ulceration/Gastritis	
Peptic ulcer	Arthralgia	Pancreatitis	
Hypertension	Retinopathy	Arthralgia	
Obesity	Hypersensitivity		
CNS symptoms			
Growth retardation			

	CYCLOSPORINE	TACROLIMUS
Nephrotoxicity	++	++
CNS toxicity	++	++
Headaches		
Tremor		
Paresthesia		
Confusion		
Nightmares		
Seizure		
Hypertension	++	+
Glucose intolerance	++	++
Hyperkalemia	+	++
Hypomagnesemia	++	++
Hyperuricemia	+	+
Gastrointestinal	++	++
Diarrhea		
Nausea/vomiting		
Abdominal pain		
Anorexia		
Hirsutism	++	−
Alopecia	−	+
Gingival hyperplasia	+	−
Hyperlipidemia	++	+

TABLE 117-6 Side Effects of Immunosuppressive Drugs

Potential inhibitors of metabolism resulting in increased levels*
Macrolide antibiotics
Erythromycin
Azithromycin
Clarithromycin
Anti fungal agents
Ketoconazole
Fluconazole
Itraconazole
Calcium-channel blockers
Verapamil
Diltiazem
Nicardipine
Cimetidine
Doxycycline
Danazol
Potential inducers of metabolism resulting in decreased levels*
Antiseizure medications
Phenobarbital
Phenytoin
Carbamazepine
Anti tuberculosis medications
Isoniazid
Rifampin

*Most interactions occur as a result of either competitive inhibition or induction of metabolism at the level of cytochrome P450 3A4.

TABLE 117-7 Potential Drug Interactions with Cyclosporine and Tacrolimus (Partial List)

Among the most important short- and long-term complications of immunosuppression are opportunistic infections and malignancies, as well as cardiovascular, metabolic, and renal diseases. These are discussed in greater detail below.

Infectious Complications Over 50% of liver transplant recipients will experience one or more episodes of infection in the postoperative period. [243] Historically, infectious complications account for most cases of death in the first year after liver transplantation. [243] The timetable of infection in all forms of solid-organ transplantation is most easily organized into three periods according to Fishman and Rubin: [244] the first month, 1 to 6 months, and more than 6 months after transplantation. In the first month after liver transplantation, conventional bacterial and fungal nosocomial infections predominate. The typical bacterial infections in the early postoperative period are wound and intravenous line infections, as well as pneumonia. The most common bacterial pathogens are *Staphylococcus* and *Enterococcus* species. [243] Vancomycin-resistant *Enterococcus* has emerged in the past few years as an important nosocomial pathogen in liver transplant recipients. [245] After the first month, opportunistic infections, such as viruses (cytomegalovirus, herpes, Epstein-Barr virus, and others), fungal infections, *Nocardia, Listeria,* tuberculosis, and *Pneumocystis carinii* pneumonia (PCP) are important concerns. [244] Fungal infections usually present 1 to 6 months after liver transplantation. *Candida albicans* represent the most common fungal pathogen. [246] Invasive aspergillosis carries a high mortality, and its incidence among liver transplant recipients may be rising. [247] Other mycoses, such as cryptococcosis, coccidioidomycosis, and histoplasmosis are rare except in cases of exposure in specific geographic endemic areas. [243], [244] Oral nystatin or clotrimazole are the most frequently used agents for fungal prophylaxis. Fluconazole has also been shown to be effective as a fungal prophylactic agent, [248] but carries a risk of increasing the level of calcineurin inhibitors (see Table 117-7). PCP is now very rare in liver transplant recipients due to highly effective long-term prophylaxis with trimethoprim-sulfamethoxazole. [249] Beyond 6 months after transplantation, opportunistic infections are less common, as the degree of immunosuppression decreases in most patients. Community-acquired infections or various recurrent or persistent infections, such as viruses (Epstein-Barr virus, hepatitis B and C) may develop. Cytomegalovirus (CMV) is one of the most important infectious complications after organ transplantation, and has been suggested to be an important cause of reduced survival [250] and high resource use among liver transplant recipients. [251] Distinction should be made between CMV "infection," which refers to the detection of the virus in tissue or body fluids, versus CMV "disease," defined as CMV infection with symptomatic clinical manifestations. [251] About 50% of liver transplant recipients with CMV infection develop symptomatic CMV disease. The incidence of symptomatic CMV disease is estimated to be 15% to 40% after liver transplantation. [251] Patients at highest risk for CMV infections include those who are CMV-seronegative who received a CMV-positive donor liver, and those treated with muromonab-CD3 or antilymphocyte globulin. [252] CMV disease is most commonly seen within the first 3 months after liver transplantation, with a median time to onset of 6 weeks. CMV is uncommon beyond 6 months after transplantation except in profoundly immunosuppressed individuals. Many patients present with a "CMV syndrome" characterized by fever, malaise, arthralgia, myalgia, and hematologic abnormalities such as leukopenia and thrombocytopenia. In liver transplant recipients, clinical manifestations include fever, hepatitis, pneumonia, gastroenteritis, and chorioretinitis in descending order of frequency. [252] Conventional CMV cultures from tissue or blood are relatively insensitive, and the results may not be available for 1 to 2 weeks. The shell vial assay using a monoclonal antibody against the early CMV antigen has replaced the conventional culture methods in many laboratories because of improved sensitivity and the advantage of rapid detection of CMV as early as 24 hours. Histologically, the classic intranuclear inclusion bodies (Cowdry type A) are not always identified. A number of diagnostic assays for the early detection of CMV infection are now available, including:

- CMV antigenemia assay which uses monoclonal antibodies to detect the viral pp65 antigen expressed on blood leukocytes
- polymerase chain reaction (PCR)
- hybrid capture CMV DNA assay. [251], [253]

While these tests are highly sensitive and specific for detecting CMV infection, the specificity for predicting CMV disease is relatively low, in the 30% to 60% range. [251], [253] The CMV antigenemia assay is the predominant diagnostic test used at the authors' institution due to the rapid turnaround time compared to the PCR assay. Standard treatment of CMV disease is intravenous ganciclovir for 14 to 21 days. There is no evidence from controlled trials in solid organ transplantation that the addition of CMV immunoglobulin increases its efficacy. [251] Clinical response to intravenous ganciclovir is seen in two thirds of patients. Among the 20% of patients with recurrent CMV infections, about two thirds respond to a second course of ganciclovir treatment. [254] There have not been any reported cases of ganciclovir-resistant CMV infection in liver transplant recipients, although this possibility certainly exists. There is currently no consensus regarding the best regimen for CMV prophylaxis after liver transplantation, [251], [255] although a small number of randomized controlled trials are shedding some light on the optimal prevention regimen. At the authors' institution, intravenous ganciclovir is used for the first week posttransplantation in all recipients. Thereafter, oral acyclovir is used for CMV-positive recipients or CMV-negative recipients of CMV-negative livers, and oral ganciclovir is used in CMV-negative recipients who receive CMV-positive livers. Preemptive therapy using intravenous ganciclovir following detection of CMV viremia was shown in one study to be effective in preventing CMV disease. [256] Valganciclovir, the oral prodrug of ganciclovir, has much higher bioavailability compared to oral ganciclovir and is currently being tested in clinical trials for prophylaxis as well as treatment of CMV infections.

Posttransplant Malignancies De novo malignancy developing after transplantation has been a well-known complication of liver and other solid-organ transplantation. [257], [258] The most common is skin cancer, followed by lymphoma, cancer of the lip, Kaposi sarcoma, vulval and perineal cancers, hepatobiliary cancers, and sarcoma. [258] Posttransplantation lymphoproliferative disease (PTLD) develops in about 2% of liver transplant recipients in the first 5 years after transplantation under both cyclosporine and tacrolimus immunosuppressive regimens. [106], [222], [258] PTLD most often occurs within the first year after liver transplantation, and involves the liver allograft in one third of cases. The usual histology is B-cell derived, large cell non-Hodgkin's lymphoma. The association between PTLD and Epstein-Barr virus infection has been well documented. [259] Management of PTLD includes withdrawal of immunosuppression, intravenous ganciclovir, interferon, or chemotherapy.

Cardiovascular, Renal, and Metabolic Complications These complications are discussed together because they are closely related. Hypertension is common after liver transplantation and may be related to renal insufficiency or immunosuppressive drugs. The incidence of hypertension appears to be lower with tacrolimus than cyclosporine. [222] The first-line treatment for hypertension after liver transplantation has been the dihydropyridine calcium-channel blockers (nifedipine, amlodipine, isradipine, felodipine, and nicardipine). [260] With the exception of nicardipine, this group of calcium channel blockers has no significant interactions with the calcineurin inhibitors, in contrast to that observed with the other groups of calcium channel blockers such as verapamil or diltiazem (see Table 117-7). Second-line antihypertensive agents include ß- and a1-adrenergic agents, loop diuretics, and angiotensin-converting enzyme inhibitors. [260], [261] The use of the a-adrenergic agents such as clonidine and doxazosin is limited by their central nervous system toxicity, particularly drowsiness. It is best to avoid the angiotensin-converting enzyme inhibitors in the early posttransplantation period because they may aggravate renal dysfunction and hyperkalemia. [260] About one third of patients achieve effective control of their hypertension with a single agent, and another one third require the addition of a second agent. [261] Renal insufficiency following liver transplantation may be either acute or chronic, and multiple etiologies have been described. [260], [261] Direct toxicity of the calcineurin inhibitors is believed to be the cause in more than 70% of patients with renal insufficiency. [260] Other causes include unresolved renal dysfunction related to preoperative hepatorenal syndrome, acute tubular necrosis, and various forms of glomerulonephritis. [260] The incidence and mechanism of nephrotoxicity appear to be similar between cyclosporine and tacrolimus. [222], [262] The greatest decline in renal function occurs in the first 18 months after liver transplantation. Renal function then remains stable in most cases despite maintaining cyclosporine at therapeutic levels. [263] Even withdrawal of cyclosporine in the setting of established azotemia may not result in an improvement of renal function, but may increase the risk of chronic rejection. [264] One approach in the management of patients with chronic renal insufficiency after liver transplantation is to reduce the dose of the calcineurin inhibitors to a minimum and add either mycophenolate mofetil or rapamycin. The incidence of end-stage renal failure necessitating hemodialysis or kidney transplantation increases with time following liver transplantation, reaching about 5% at 5 years and 10% after 10 years. [260] Obesity, diabetes,

and hyperlipidemia are common after liver transplantation. [265], [266] In an analysis of about 800 adults from the NIDDK Liver Transplant Database, approximately 20% of nonobese liver transplant recipients became obese over the 2 years after liver transplantation. [267] The cause of posttransplant obesity, diabetes, and hyperlipidemia are multifactorial and interrelated. Diabetes is present in about 5% to 15% of patients with advanced cirrhosis awaiting liver transplantation. [265] These patients are at higher risks for posttransplant diabetes than those without diabetes prior to liver transplantation. The management of diabetes after liver transplantation is not substantially different than in the nontransplant setting, although the steroid tapering process may result in improved diabetic control with time after liver transplantation. Conflicting results have been reported with respect to the impact of diabetes on medium- to long-term survival. [265] One study revealed a fourfold higher mortality among diabetic patients, [268] whereas another study showed a higher survival rate in diabetics versus nondiabetics. [269] A more consistent finding, however, is a higher incidence of bacterial and fungal infections after liver transplantation among diabetic patients compared to nondiabetic patients. [265] The intriguing observation from a few studies that the prevalence of diabetes after liver transplantation is higher among patients with hepatitis C infection needs to be further investigated. [270], [271] Hyperlipidemia may be associated with obesity, diabetes mellitus, cyclosporine, rapamycin, and corticosteroids. Treatment of hyperlipidemia may be problematic after liver transplantation. Cholestyramine interferes with gastrointestinal absorption of calcineurin inhibitors and therefore should be administered at a separate time from the calcineurin inhibitors. Niacin has multiple side effects and can be hepatotoxic. Preliminary data regarding the safety and efficacy with HMG-coenzyme A reductase inhibitors in liver transplant recipients appear to be favorable. [272] Patients on tacrolimus may have a lower cardiovascular risk profile, based on hypertension and hyperlipidemia, than patients on cyclosporine. [273] Switching from cyclosporine to tacrolimus may benefit some patients with poorly controlled hyperlipidemia. Despite the increase in cardiac risk index for many of these patients after liver transplantation, long-term follow-up data regarding the risks and incidence for cardiovascular events and mortality following liver transplantation are only beginning to emerge. One study found that among patients who survived longer than 5 years after liver transplantation, the prevalence of cardiac disease was no higher than that in the nontransplant population in the United States. [274] Two other studies, on the other hand, reported cardiovascular events as the cause of 14% to 24% of deaths beyond one year after liver transplantation. [275], [276] Clearly, more studies with longer follow-up are needed. At present, it is important to try to correct potentially reversible cardiovascular risk factors following liver transplantation.

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CHAPTER 118

Lewis R. Roberts and Gregory J. Gores

HEPATOCELLULAR CARCINOMA

EPIDEMIOLOGY

Hepatocellular carcinoma (HCC) is the fifth most common cancer and the third most frequent cause of cancer death worldwide, with an estimated 560,000 new cases per year. [1] Estimates from the International Agency for Research on Cancer for the year 2000 show worldwide age-specific incidence ratios of 14.97 cases per 100,000 per year in men and 5.51 cases per 100,000 per year in women. [1] Because of the low overall cure rate for HCC, the worldwide age-specific mortality ratios are nearly identical to the incidence ratios at 14.41 deaths per 100,000 per year in men and 5.46 deaths per 100,000 per year in women. There are strong etiologic associations with chronic hepatitis B virus (HBV), chronic hepatitis C virus (HCV), alcoholic cirrhosis, other causes of chronic liver disease, and dietary aflatoxin exposure. Because of the strong associations with diseases and risk factors that have distinct geographic distributions and different frequencies within different ethnic groups, there is also a preferential distribution of cases of HCC to certain geographic regions and specific at-risk populations.

International Variation

There is considerable international variation in rates of HCC. By region, the highest incidence is in Eastern Asia, with age-specific ratios of 35.46 per 100,000 in men and 12.66 per 100,000 in women. China, with the largest population in the region, had an estimated 306,000 cases of HCC in 2000, representing 54% of the total number of cases worldwide. Other regions of high incidence are sub-Saharan Africa, South Eastern Asia, and Southern Europe. The lowest incidence rates are found in Northern Europe; North, Central, and South America; South Central Asia; and Australia/New Zealand. The highest incidence regions in Asia and Africa have a high prevalence of chronic HBV infection as well as high-level exposure to dietary aflatoxins. These combined risk factors play a synergistic role in the development of HCC. Other combinations of risk factors that are important in the epidemiology of HCC include co-infection with HBV and HCV and alcohol use in individuals with chronic hepatitis from viral or other causes. Cigarette smoking and long-term use of oral contraceptives also appear to be independent risk factors for HCC. The risk for HCC increases with age. The age distribution of cases of HCC also varies in different populations. A significant proportion of HCCs occurs in young and middle-aged individuals at the peak of their productive lives. This results in a high public health and economic impact, particularly in the countries with the highest incidence rates. There is a 15- to 40-year lag time between development of chronic hepatitis or cirrhosis and diagnosis of HCC. This lag time is shorter in patients with active chronic hepatitis C; therefore, the increasing prevalence of chronic hepatitis C has led to a decrease in the age of onset of HCC in many populations. For HBV-induced HCC, the age at acquisition of HBV infection is important because individuals who become infected in the perinatal period or in early childhood are more likely to develop chronic hepatitis or become chronic carriers, with an attendant increased risk for cirrhosis and HCC. [2], [3]

Gender Variation

There is a significant effect of gender on the risk for HCC. The worldwide male-to-female incidence ratio is 2.7:1. In general, the male-to-female ratio is highest in the regions with the highest incidences of HCC. Higher levels of androgen signaling may be associated with the increased risk for HBV-related HCC in men. [4]

North America

In the United States, there has been a well-documented increase in the number of cases of HCC since the 1980s. Analysis of data from the Surveillance, Epidemiology, and End Results (SEER) database of the National Cancer Institute showed a 71% increase in incidence of histologically proven HCC from 1.4 per 100,000 population for the period from 1976 to 1980 to 2.4 per 100,000 for the period from 1991 to 1995. [5] In particular, the incidence of HCC increased significantly among younger people, 40 to 60 years old, during the period from 1991 to 1995 as compared with earlier periods. Between 1981–1985 and 1991–1995, there was a 41 percent increase in the overall mortality rate from HCC. There is also an effect of gender on the incidence of HCC in the United States, with an incidence in men that is three times the incidence among women. There is a striking effect of race on the incidence of HCC in the United States. For the period from 1991 to 1995, Hispanics, Asians, Pacific Islanders, and Native Americans had the highest age-adjusted incidence of HCC, 7.4 per 100,000. For black men, the incidence was 6.1 per 100,000 per year, whereas for white men, it was 2.8 per 100,000. The incidence of HCC is highest among recent Asian immigrants to the United States, with age-standardized rates of 23.9 per 100,000 in Korean men and 5.5 per 100,000 in Korean women. These immigrant populations have a high prevalence of chronic HBV infection. Subsequent United States–born generations typically have lower rates of chronic HBV infection and a lower risk for HCC. [5] In general, people of lower social class in urban U.S. communities have a higher risk for HCC. This is likely related to the inverse relationship of social class with prevalence of HBV infection and

cigarette smoking.

Analysis of hospitalization rates for HCC in a population of U.S. veterans showed that the recent increase in incidence of HCC was primarily due to chronic HCV infection. Between 1993–1995 and 1996–1998, age-adjusted hospitalization rates for HCC associated with HCV increased threefold, from 2.3 per 100,000 in 1993–1995 to 7.0 per 100,000 in 1996–1998. During the same time periods, the age-adjusted hospitalization rates for HCC associated with HBV or alcoholic cirrhosis and in patients without known risk factors remained relatively stable. [6,7]

Despite improvements in early diagnosis and management of HCC, survival of patients with HCC in the United States has not improved significantly since the 1980s. A recent analysis of data from the SEER database showed that the overall 1-year relative survival rate increased from 14% in the period from 1977 to 1981 to 23% in the period from 1992 to 1996. [8] However, 5-year relative survival rates did not change as much, increasing from 2% to 6%. Median survival after diagnosis increased only slightly, from 0.57 years in the period from 1977 to 1981 to 0.64 years in the period from 1992 to 1996. Because most of the improvement in survival occurred in the first year after diagnosis, the apparent improvement is likely to be due to lead-time bias resulting from earlier diagnosis of HCC rather than from improvements in long-term survival resulting from more effective treatment. There were no significant influences of gender or ethnicity on survival. Most patients with HCC had advanced disease at the time of diagnosis. In the period from 1987 to 1991, only 0.8% of patients with HCC underwent radical surgery with curative intent; these patients had a significantly better survival than patients with untreated advanced disease, with a 1-year relative survival rate of 60% and a 5-year relative survival rate of 40%. In the same period from 1987 to 1991, 7.4% of patients underwent palliative surgery; these patients had a 1-year survival of 50% and a 5-year survival of 20%. The proportion of patients undergoing radical surgery in the later period from 1991 to 1996 was unchanged from the earlier period at 0.8%. These data suggest that screening programs aimed at identifying patients with HCC early enough to allow radical surgery either were not widely practiced, were ineffective, or were not coupled with widespread availability of the specialized surgical skills needed for optimal radical surgery. The proportion of patients with HCC able to undergo radical surgery is probably also limited by the recognition in recent years that surgical outcomes in patients with cirrhosis and clinically significant portal hypertension are very poor. [9] There is an urgent need to develop methods of identifying the individuals who are at highest risk for HCC, to determine the optimal and most cost-effective screening methods to allow early diagnosis of HCC, and to provide access to specialized multidisciplinary management of HCC. Recent improvements in the management of HCC, particularly the increasing availability of liver transplantation for patients with early disease, may result in further improvements in survival.

The Alaskan Native population has a relatively high age-adjusted incidence of HCC of 7.9 per 100,000 in men and 1.4 per 100,000 in women. For Eskimos, the incidence rates are 13.1 per 100,000 in men and 2.2 per 100,000 in women. [10] Most cases of HCC in this population are related to chronic HBV infection. There is also significant familial clustering of HCC in the Alaskan Native population. In many cases, individuals develop HCC at a very young age, often as early as the second decade of life, suggesting a familial predisposition to HCC. In addition, there is no evidence of dietary contamination with fungal aflatoxins, and *p53* mutations have not been demonstrated in any HCCs tested so far in this population; this contrasts with the 29% rate of *p53* mutations in HCCs worldwide. [11] The available evidence, therefore, strongly suggests an inherited predisposition to HCC, potentiated by the high rates of chronic HBV infection. A hepatitis B immunization program that includes prevention of perinatal HBV infection, routine infant vaccination, and catch-up vaccination of older children and adults has been used to eliminate new chronic HBV infections in the Alaskan Native population. [12] Presumably, as has been documented in Taiwan, the decrease in HBV infections will result in a decrease in incidence of HCC in this population. [13]

ETIOLOGY AND RISK FACTORS

Chronic Hepatitis B

There are 350 million chronic HBV carriers worldwide. The prevalence of HBsAg positivity ranges from high (more than 8%) in Africa, Asia, and the Western Pacific, to intermediate (2% to 7%) in Southern and Eastern Europe, and low (less than 2%) in Western Europe, North America, and Australia. Fifty million new cases of HBV infection occur each year; of these, 5% to 10% of adults and up to 90% of infants become chronically infected. In the United States, at least 250,000 people contract hepatitis B each year. The predominant mode of viral transmission in a population varies with the prevalence of HBV infection. In low-prevalence countries, sexual contact among high-risk adolescents and adults is the predominant mode of transmission. In highly endemic countries, most HBV infections are contracted perinatally or in early childhood. One fourth to one third of individuals with chronic hepatitis B develop progressive liver disease. Cirrhosis develops in 0.1% to 2% of patients with chronic hepatitis B each year, depending on the duration of HBV replication, the severity of disease, and the presence of coexisting infections or alcohol use. In individuals who develop cirrhosis, the risk for progression to HCC varies from 2% to 10% per year. [14] Both host and viral factors influence the natural history of chronic hepatitis B. Certain HBV strains predominate in HCCs, suggesting that particular genotypes are more effective at initiating or promoting hepatocarcinogenesis. [15] Vaccination of infants and at-risk individuals decreases the prevalence of chronic HBV infection and the incidence of HCC. [13]

Chronic Hepatitis C

An estimated 170 million people are infected with HCV worldwide, and in some countries, HCV has overtaken HBV as the major cause of HCC. HCV is an RNA virus that causes acute and chronic hepatitis. It is contracted through parenteral exposure to infected material. Those at highest risk for acquiring hepatitis C are recipients of contaminated blood transfusions, injection drug users, people who snort cocaine with shared straws, and health care workers at risk for needle-stick and other exposures. Although the incidence of acute HCV infection has fallen dramatically in the United States since the 1990s, the prevalence of infection remains high (about 2.7 million Americans) because chronic hepatitis C develops in about 75% of those infected. Both acute and chronic hepatitis C are asymptomatic in most patients. Chronic hepatitis C is a slowly progressive disease that eventually results in severe morbidity in 20% to 30% of infected patients. Virologic features of HCV, such as genotype and viral load, are associated with progression and response to therapy. [3] There is a marked difference in the geographic distribution of HCV genotypes, with types 1, 2, and 3a being most frequently found in Western countries. Genotype 1b is associated with more severe liver disease and with lower response rates to antiviral therapy. The development of quasispecies and escape mutants enables viral persistence and the development of chronic liver disease. Persistent hepatitis, with high average annual alanine aminotransferase levels, is a significant predictor of the development of HCC. [16] Patients with active hepatitis, those with co-infections with HBV or human immunodeficiency virus (HIV), and those with other causes of chronic liver disease such as alcoholic liver disease are more likely to develop liver-related complications. Patients with combined hepatitis B surface antigen positivity and chronic HCV infection are at particular risk for HCC. [17] HCV-positive individuals who are hepatitis B core antibody positive also have a higher prevalence of cirrhosis, lower HCV RNA levels, and an impaired ability to respond to interferon treatment. [18]

Because of shared routes of transmission, patients infected with the HIV virus are at risk for co-infection with HCV. With the advent of highly active antiretroviral therapy, there has been decreased morbidity and better survival of patients with HIV infection. Co-infection with HIV may also be associated with more rapid progression of chronic HCV. As a result, hepatic cirrhosis, end-stage liver disease, and HCC due to chronic infection with HCV are important causes of both morbidity and mortality in co-infected patients. [19]

After surgical resection of HCC, the presence of active hepatitis and hepatitis C viremia are risk factors for tumor recurrence. Prior treatment of hepatitis C with interferon is protective against the initial development of HCC or recurrence after surgical resection. The protective effect of interferon is most marked in patients who respond to interferon treatment by clearing the virus, but it may occur to a lesser degree even in patients who do not clear the virus. [20]

Dietary Aflatoxin Exposure

Dietary exposure to fungal aflatoxin is an important risk factor for development of HCC. Aflatoxin (derived from *a* spergillus *fla* vus *toxin*) is a mycotoxin produced by two fungal species, *Aspergillus flavus* and *Aspergillus parasiticus*. *A. flavus* is widespread in nature, occurring in soil and decaying vegetation, hay, and grains, particularly in conditions of high moisture content and high temperature. There are at least 13 different types of aflatoxin, with aflatoxin B1 considered the most toxic. Epidemiologic studies have indicated a relationship between aflatoxin intake and incidence of liver cancer in several countries. [21]

Aflatoxins are metabolized in the liver by the cytochrome P450 and glutathione S-transferase enzyme systems. Aflatoxin B1 is a procarcinogen that is converted in the liver to the mutagenic metabolite aflatoxin B1-8,9-exo-epoxide by hepatic microsomal cytochrome P450. Glutathione S-transferases and other phase II enzymes subsequently detoxify this DNA-reactive metabolite. [22] Interindividual differences in the biotransformation of dietary aflatoxins result in differing susceptibility to aflatoxin-induced carcinogenesis. The glutathione S-transferase M1 (*GSTM1*) gene exhibits genetic polymorphism, with a proportion of normal individuals having a homozygous deletion of the gene. Individuals living in regions where dietary aflatoxin levels are high who have the *GSTM1* null genotype are at increased risk for HCC. [23] Aflatoxin exposure predisposes to mutations in the *p53* gene, particularly a G-to-T transversion at codon 249 of the *p53* gene, an event that contributes to the pathogenesis of HCC. [24] In many countries, particularly in tropical regions, grains, peanuts, beans, and other foods are harvested and stored under conditions that

allow fungal growth and accumulation of high levels of aflatoxin. Measures are being instituted in many countries to decrease contamination of foodstuffs with aflatoxin. There are also efforts to develop drugs that enhance aflatoxin detoxification and excretion. [25]

The carcinogenic potential of dietary aflatoxins is significantly increased by the presence of concomitant chronic HBV infection. In one study, patients testing negative for HBV had 0.01 cases of HCC per 100,000 population per year per nanogram of aflatoxin B1 ingested per kilogram of body weight per day. In contrast, patients testing positive for HBV had 0.3 cases per year per 100,000 population, a 30-fold higher risk than in the absence of hepatitis B surface antigen in the serum. Individuals with chronic HCV infection are also probably at increased risk from dietary aflatoxin. [26]

Cirrhosis from Alcohol and Other Chronic Liver Diseases

Cirrhosis from any cause, including chronic alcohol abuse, increases the risk for HCC. The HCC risk is higher for individuals with chronic hepatitis and ongoing hepatocellular injury. Therefore, individuals who stop drinking significantly reduce their risk for HCC. Similarly, patients with nonalcoholic steatohepatitis and cirrhosis who achieve reduced hepatic steatosis and those with hereditary hemachromatosis and cirrhosis who are iron depleted by phlebotomy have a reduced risk for HCC. Certain causes of cirrhosis, such as autoimmune hepatitis, carry a relatively low risk for HCC. [27]

Familial and Genetic Influences on Risk for Hepatocellular Carcinoma

Particularly in areas of the world with a high incidence of HCC, family history of HCC is a significant risk factor for HCC. [28] The biologic basis for this phenomenon is unknown. Epidemiologic studies have suggested either the presence of a recessive allele that contributes to risk for HCC or a familial predisposition to a prolonged HBV replication phase. [29], [30] Genetic polymorphisms of the carcinogen-metabolizing enzymes cytochrome P450 (CYP), glutathione S-transferase (GST) M1, and *N*-acetyltransferase (NAT2), as well as p53 polymorphisms, may contribute to familial risk for HCC. [31]

PATHOGENESIS

Oncogene activation and tumor suppressor inactivation play important roles in carcinogenesis. Oncogene activation occurs through transcriptional activation, gene amplification, or development of activating mutations of genes involved in cellular growth control. Tumor suppressor inactivation can occur through genomic deletions, translocations, or mutations or through epigenetic modifications such as promoter methylation. Normal human somatic cells have a finite lifespan in vivo and undergo senescence after a predictable number of cell divisions. Cellular senescence is triggered by the activation of two interdependent mechanisms. One mechanism leads to induction of cell cycle arrest due to activation of two tumor suppressor genes, *p53* and *pRb*. The second mechanism is activated by a critical shortening of chromosomal ends due to the progressive shortening of the telomeres with each cell division. When both cell growth regulatory mechanisms are inactivated, unchecked cell division and progression to cancer occurs. Cells that are turning over at higher than normal rates because of local growth factor up-regulation or chronic cell injury and regeneration have a higher likelihood of developing genomic alterations that lead to oncogene activation or tumor suppressor inactivation. Environmental exposure to carcinogens can also cause oncogene activation or tumor suppressor inactivation (Fig. 118-1). A number of approaches have been used to elucidate the molecular mechanisms of liver carcinogenesis. These include (1) the use of chemical tumor initiators and promoters in animal models; (2) studies of growth factors and their signaling pathways; (3) transgenic mouse models overexpressing cytokines, growth factors, or oncogenes; (4) studies of immune-mediated mechanisms of hepatocellular injury; (5) analysis of the molecular genetic changes that occur in HCCs, including studies of chromosomal allelic imbalance, comparative genomic hybridization, restriction landmark genomic scanning, and gene expression analysis using cDNA microarrays; and (6) studies of the molecular consequences of HBV integration and the interaction of the protein products of HBV and HCV with host cell processes. These studies have contributed to our growing appreciation of the multiplicity of mechanisms and pathways that may contribute to the carcinogenic process in toxin-affected, chronically inflamed, or otherwise injured liver tissue. In cirrhotic livers, macroregenerative nodules with foci of hepatocyte dysplasia have been identified as preneoplastic lesions of HCC. Histologically, dysplastic lesions are classified as small cell or large cell lesions or as foci of adenomatous hyperplasia; available evidence suggests that small cell dysplasia and adenomatous hyperplasia are the predominant preneoplastic lesions. [32] Unfortunately, we are still far from a unified, comprehensive understanding of liver carcinogenesis. This is in part because HCC is initiated in multiple genetic and environmental contexts and almost certainly emerges as a consequence of multiple possible pathways. The lack of a comprehensive view of the pathogenesis of HCC has also prevented the development of effective, targeted, preventive or therapeutic interventions that are elegant and also simple enough to be applicable to most patients with this disease.

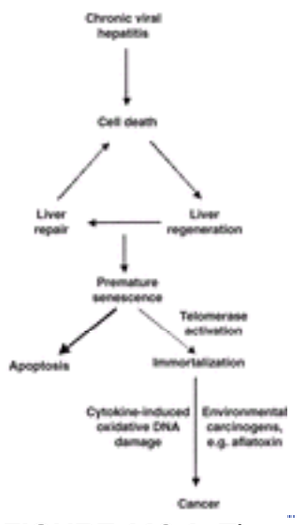

FIGURE 118-1. The role of chronic hepatitis with recurrent liver injury, regeneration, and repair in development of hepatocellular carcinoma. Premature senescence of liver cells occurs by apoptosis. Cells undergoing telomerase activation escape apoptosis. The resulting immortalized cells are susceptible to the influence of oxidative and environmental carcinogen-induced DNA damage, leading to the eventual development of transformed neoplastic hepatocytes.

Tumor Suppressor Genes

Tumor suppressor genes are normal cellular genes that, when homozygously inactivated, can contribute to tumor development. Tumor suppressor genes generally operate in a recessive manner, requiring loss of both copies for tumorigenesis, in contrast to oncogenes, which can exert their effects in a dominant fashion. Loss of heterozygosity (LOH) is loss of one allele in a tumor cell from a chromosomal region for which the individual's normal cells are heterozygous. LOH is detected using polymorphic DNA markers that can distinguish between the two alleles. If one allele of a tumor suppressor gene is inactivated by mutation, then deletion of the second allele, detected by LOH, is associated with loss of expression of the gene. Promoter hypermethylation also leads to decreased expression of tumor suppressor genes. DNA hypermethylation on CpG islands and DNA hypomethylation on pericentromeric satellite regions are early events during hepatocarcinogenesis. CpG island hypermethylation leads to decreased tumor suppressor gene expression, whereas pericentromeric hypomethylation results in chromosomal instability and deletions. [33]

Known or putative tumor suppressor genes shown to be mutated in HCC include *p53,* [34] retinoblastoma (*Rb*), [35] *p16,* [36] *axin1,* [37] *Smad2,* [38] *Smad4,* [38] *M6P/IGF2R,* [39] *BRCA2,* [40] and *DLC-1*. [41] In human HCCs, LOH has now been reported in multiple chromosomal regions, including 1p, 1q, 2q, 4q, 5q, 6q, 8p, 8q, 9q, 10q, 11p, 12p, 13q, 14q, 16q, and 17p. [42], [43] A number of these regions contain known tumor suppressor genes, such as at 17p13.1 (*p53*), 6q26-27 (*M6P/IGF2R*), 8p21.3-22 (*DLC-1*), and 13q12-q32 (*Rb* and *BRCA2*). For a few of these chromosomal loci, clinicopathological associations have been demonstrated, such as the association of LOH on chromosome 1p with early-stage HCC [44] and that of LOH on chromosome 16q with progression of HCC. [45] In addition, patients with LOH at multiple regions have more advanced stage of disease, less well-differentiated tumors, higher serum alpha-fetoprotein levels, and a worse prognosis. [46] Chronic HBV infection and integration of the HBV genome are also associated with high rates of genomic instability. [47] Comparative genomic hybridization (CGH) allows the identification of gains and losses of DNA sequences across the entire tumor genome. Analysis of HCC tumors and cell lines by CGH has revealed genomic DNA copy number gains on chromosomal arms 1p, 1q, 6p, 7p, 7q, 8q, 11q, 12q, 17q, 20p, and 20q. Recurrent losses were found on 3p, 3q, 4p, 4q, 5q, 6q, 8p, 9p, 11q, 13q, 14q, 15q, 16q, 17p, 18q, and 21q. [48], [49] and [50] Significantly, most of the overrepresented regions harbor known protooncogenes, and half of the underrepresented regions coincide

with sites of known or putative tumor suppressors. [49] Notably, a recent comparison of CGH analysis of HCCs due to HBV infection, as compared with HCCs from nonviral etiologies, showed a lower frequency of loss at 4q, 16q and 17p in nonviral HCC samples, suggesting that these abnormalities are associated with HBV infection. [51] CGH analyses also reveal a similar pattern of chromosomal alterations in HBV- and HCV-induced HCCs. Thus, a subset of alterations may preferentially contribute to virus-induced carcinogenesis. In general, LOH and CGH studies have shown variable rates of chromosomal instability. Patient populations with low rates of virus-induced HCC have lower levels of chromosomal instability than populations with high rates of virus-induced HCC. Despite this variability, there is remarkable consistency in the locations of chromosomal instability, suggesting that particular gene targets are affected by genomic instability in HCC.

Investigation of gene mutations in the tumor suppressor gene *p53* in HCC has been particularly revealing because HCCs from patients with high dietary exposure to fungal aflatoxin B1 have a high frequency of point mutations at the third position of codon 249 of the *p53* gene, resulting in a G:C-to-T:A transversion. [52] The frequency of this mutation in HCCs increases proportionally to the level of dietary exposure to aflatoxins. A G:C-to-T:A transversion at the second position of codon 249 is often found in patients with chronic HBV or HCV infection. The occurrence of this mutation correlates with oxyradical exposure. Both of these mutations lead to decreased binding of *p53* to its nuclear DNA targets. [53] Mutations in *p53* have been demonstrated in nonneoplastic liver cells in subjects from communities with high dietary aflatoxin B1 exposure, suggesting that the mutations are early events in neoplastic transformation (Fig. 118-2). However, *p53* mutations can also occur late in tumor progression. The HBV HBx gene product has also been shown to interact with *p53* and strongly inhibit *p53* sequence-specific binding, leading to inhibition of *p53*-mediated apoptosis. Abnormalities in the *Rb* gene have been noted in association with *p53* mutations in advanced HCCs, particularly in poorly differentiated tumors, and *p53* and *Rb* mutations may have an additive effect in liver carcinogenesis. [54] HCCs from different populations show different frequencies of *p53* mutations. In Alaskan Natives, *p53* mutations are rare or absent, suggesting that a difference in environmental or host factors leads to a different mechanism of hepatocarcinogenesis. [11] Molecular genetic studies show that HCCs appear to fall into two groups, with either low or high levels of chromosomal changes. [55] In general, the tumors with low-level chromosomal instability also are more likely to have disruption of the Wnt signaling pathway, with ß-catenin mutations, low-grade histology, and a better long-term prognosis (Fig. 118-3). In contrast, tumors with high-level chromosomal instability are more likely to have *p53* mutations.

FIGURE 118-2. Major molecular pathways of hepatocarcinogenesis. Hypermethylation of the *p16* gene promoter is one of the earliest changes in development of hepatocellular carcinoma. In regions with high dietary aflatoxin exposure, the development of *p53* mutations also occurs early. In areas with low aflatoxin exposure, *p53* and *pRb* pathway disruption occur late in hepatocarcinogenesis.

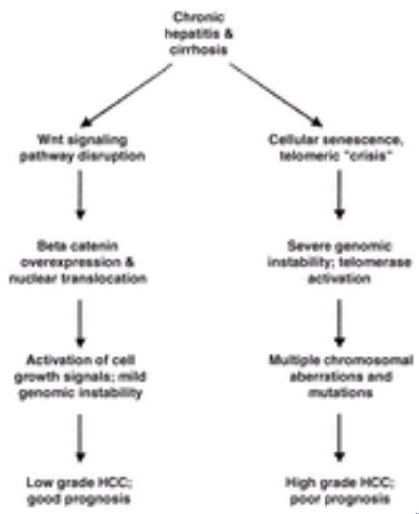

FIGURE 118-3. Two distinct pathways of liver carcinogenesis. Disruption of the Wnt signaling pathway occurs in about 25% of hepatocellular carcinomas. ß-Catenin mutations prevent degradation of ß-catenin and lead to accumulation of nuclear ß-catenin. ß-Catenin cooperates with the Tcf transcription factor to activate cellular growth control genes. Tumors with ß-catenin mutations tend to have lower rates of genomic instability and to be lower-grade tumors with a better prognosis.

Promoter hypermethylation is an important cause of loss of expression of tumor suppressor genes in hepatocarcinogenesis. Genes inactivated by this mechanism include *p16,* [56] *SOCS,* [57] *TSLC1,* [58] and E-cadherin. [59] In particular, *p16* inactivation by hypermethylation appears to be one of the earliest genetic events in hepatocarcinogenesis [60] (see Fig. 118-3).

Oncogenes

Protooncogenes are cellular genes involved in the control of cell growth. Mutation, overexpression, or amplification of these genes leads to oncogenic activity that contributes to neoplastic transformation. Oncogenes known to be overexpressed in HCC include *c-fos* and *c-myc,* [61] *c-erbB-2,* [62] *c-met,* [63] ß-catenin, [64] and gankyrin. [65] Overexpression of *c-myc* is frequently a consequence of amplification of the *c-myc* locus in HCC. [48] Activation of the *c-myc* and *c-fos* oncogenes also occurs as a consequence of the transactivating function of the HBV x protein (HBx) and the carboxyl-terminal truncated middle hepatitis B surface protein (MHBst). [66] Oncogene activation may also occur through promoter hypomethylation, [67] through reactive oxygen species–mediated pathways, or through protein kinase C or mitogen-activated protein kinase pathways. Reactive oxygen species– or lipid peroxidation–mediated processes are also important in the pathogenesis of chronic liver injury induced by alcohol, genetic hemachromatosis, and a $_1$-antitrypsin deficiency.

Growth Factors and Growth Factor–Signaling Pathways

A number of different growth factors are involved in hepatocarcinogenesis. Insulin and insulin-like growth factors (IGFs) I and II promote hepatocyte growth. IGF-II is frequently overexpressed in HCCs. In addition, insulin receptor substrate 1 (IRS-1), a cellular mediator of IGF signaling, is frequently overexpressed in HCCs. [68] Signaling events downstream of IRS-1 lead to activation of the mitogen-activated protein kinase cascade and cell proliferation through up-regulation of cellular growth genes and inhibition of apoptosis. [69] Transforming growth factor-a (TGF-a) and the structurally related epidermal growth factor are another potent class of hepatocellular growth factors. TGF-a is overexpressed in the liver of patients with chronic hepatitis. Because TGF-a levels are often increased in HCCs, it is likely that it contributes to cellular proliferation in cancer, and it may be a factor in tumor initiation or progression in patients with chronic hepatitis. In a transgenic mouse model, TGF-a dramatically enhances *c-myc*–induced hepatocarcinogenesis by promoting proliferation and survival of HCC cells. [70] The TGF-a/ *c-myc* transgenic mouse model also shows hyperphosphorylation of the retinoblastoma protein pRb, which results in inactivation of pRb and release of E2F transcriptional activity, resulting in cell cycle progression and enhanced mitosis. [71] Transforming growth factor-ß $_1$ (TGF-ß $_1$) inhibits cell proliferation and promotes cellular differentiation, fibrogenesis, and apoptosis. Increased TGF-ß $_1$ levels may create an environment in which selection of hepatocyte clones resistant to TGF-ß $_1$–induced apoptosis occurs. The *Smad* genes are involved in TGF-ß signaling, and mutations in the *Smad2* and *Smad4* genes have been described in HCC. [38] Hepatocyte growth factor, the ligand for the *c-met* oncogene, activates the Akt/PI-3 kinase pathway to suppress Fas-mediated cell death in human HCC. [72] High-molecular-mass forms of basic fibroblast growth factor have also been shown to have a mitogenic effect on hepatocytes. [73] Clearly, multiple growth-promoting molecules are involved in hepatocellular carcinogenesis.

Telomere Length and Telomerase Activity

Telomeres are specialized protein-DNA structures at the ends of chromosomes that contain long stretches of TTAGGG hexameric repeats. Telomeres prevent degradation of chromosome ends and end-to-end fusion with other chromosomes. Aging of somatic cells is associated with reduction in telomere length, owing to the inability of traditional DNA polymerases to replicate completely the end of the chromosomal DNA. In contrast, germ-line and neoplastic cells express telomerase, an enzyme that restores telomere length. There is progressive shortening of telomeres during progression from chronic hepatitis to cirrhosis and eventually to HCC. [74] This occurs as a consequence of the multiple cycles of cell injury, death, and regeneration that occur in injured liver, leading to premature hepatocellular senescence. Telomere shortening beyond a critical length leads to a proliferative block, referred to as *telomeric crisis,* which is characterized by chromosomal instability, end-to-end fusions, and cell death. Stabilization of the telomeric DNA through either telomerase activation or the activation of an alternative mechanism of telomere maintenance is essential if the cells are to survive and proliferate indefinitely [75] (see Fig. 118-2). There is cross talk between the oncogene and telomerase pathways because *c-myc* has been shown to activate the telomerase gene promoter. [76] Hepatocarcinogenesis is characterized by the evolution of clones of hepatocytes with increased telomerase expression and an immortalized phenotype. [77] Therefore, almost all HCCs show reactivation of telomerase activity. [78]

Microsatellite Instability

Microsatellite instability (MSI) is defined as a change of any length due to either insertion or deletion of repeating units in a DNA microsatellite within tumor genomic DNA when compared with normal tissue. This form of genomic instability is associated with defective DNA mismatch repair in tumors, which is important in the pathogenesis of the hereditary nonpolyposis colon cancer syndrome (HNPCC) and associated malignancies. [79] To date, six mismatch repair genes have been identified in humans: *hMLH1, hMSH2, hPMS1, hPMS2, hMSH3,* and *hMSH6*. Colorectal cancers are classified as having high-frequency MSI (MSI-H) if greater than 30% to 40% of more than five microsatellite loci analyzed show instability, as having low-frequency MSI (MSI-L) if less than 30% to 40% of more than five loci analyzed show instability, or as being microsatellite stable (MSS) if 0% of loci show instability. Of sporadic colorectal cancers, 70% are MSS, 15% are MSI-L, and 15% are MSI-H. MSS or MSI-L colorectal cancers do not have an associated defective mismatch repair phenotype or the clinicopathological features of HNPCC tumors; instead, they behave in the same manner as sporadic colon cancers. Clear criteria have not been defined for MSI in noncolonic tumors. To date, a number of studies have demonstrated either no instability or a relatively low frequency of low-level MSI in HCC. [80], [81] As is the case with other noncolonic tumors, it is unclear whether this finding is of significance in the etiology or pathogenesis of HCCs or is instead simply a consequence of the generalized genomic instability found in cancer. It has been suggested that cumulative low-level MSI at multiple loci leads to tumor progression. Further investigation is needed to resolve this potentially important question.

Chromosomal Fragile Sites

Chromosomal fragile sites are specific genetic loci that are susceptible to forming gaps, breaks, and rearrangements in metaphase chromosomes of cells cultured under conditions that inhibit DNA replication, such as treatment with the DNA a-polymerase inhibitor, aphidicolin. Fragile sites are grouped into “common” or “rare” classes based on their frequency of occurrence and the culture conditions required for their expression. Thus far, 89 common and 28 rare chromosomal fragile sites have been identified. Common fragile sites are present in all individuals. The most frequently observed common chromosomal fragile sites occur at 3p14.2 (FRA3B), 16q23 (FRA16D), 6q26 (FRA6E), 7q32 (FRA7H), and Xp22 (FRAXB). [82] Common fragile sites span a distance of 250 to 1500 kilobases and display characteristics of unstable, highly recombinogenic DNA in vitro. In particular, they are preferred sites for sister chromatid exchanges, chromosomal deletions and rearrangements, integration of viral sequences and transfected plasmid DNA, and initiation of bridge-breakage-fusion cycles, which lead to gene amplification. Because of these characteristics and the frequent coincidence of fragile sites with chromosomal breakpoints in malignant cells, it has been hypothesized that fragile sites are involved in carcinogenesis. [83] The most convincing evidence of the potential significance of the fragile sites in carcinogenesis is the location of FRA3B, the most highly inducible common fragile site, at chromosome 3p14.2. This chromosomal region is frequently deleted in lung cancer, renal cell carcinoma, and pancreatic cancer and is also the location of the fragile histidine triad (*FHIT*) gene. [84] The *FHIT* gene has been proposed to be a tumor suppressor and has recently been identified as a preferential target in HCC. [85] The cloning of additional fragile sites, particularly ones such as FRA16D (16q23) and FRA6E (6q26) that are located in regions at which there is known LOH in HCC, should provide additional information about the potential role of fragile sites in hepatocarcinogenesis.

Angiogenesis

New vessel formation is required for the continual growth of the tumor and provides a gateway for cells to escape the confines of the primary tumor. HCCs are usually hypervascular and show evidence of active angiogenesis. Arteriolar density in the tumors correlates with angiographic vascularity and also with the proliferating cell nuclear antigen labeling index. [86] It has been shown that IGF-II expression is stimulated by hypoxia, and that IGF-II in turn activates expression of vascular endothelial growth factor, an important regulator of tumor angiogenesis. [87] IGF-II has also been shown to stimulate angiogenesis directly. The hypoxia inducible factor (HIF-1) also plays a role in angiogenesis in HCC. [88] HIF-1 stimulates angiogenesis by activating transcription of the gene encoding vascular endothelial growth factor. In parallel with the increase in angiogenic stimuli, it has been shown that the expression of collagen XVIII, the precursor of the antiangiogenic molecule endostatin, is decreased in larger and more vascular HCCs. [89] Antiangiogenic agents have been shown to be effective against HCC in a mouse model, and it is likely that clinical trials of antiangiogenic agents in human HCC will be performed and reported soon. [90]

Hepatitis B Virus

Multiple mechanisms appear to contribute to HBV-induced hepatocarcinogenesis. Although persistent hepatitis with regenerative hepatocellular turnover and cirrhosis is presumed to play a significant role in HBV-induced carcinogenesis, a significant percentage of HBV-related HCCs occur in the absence of cirrhosis, suggesting a specific role for HBV in the development of these cancers. HBV integrates into the host genome in almost all patients with chronic hepatitis. Viral integration almost invariably precedes the development of HCC. HBV DNA appears to integrate into random sites in the human genome; however, with recent advances in molecular genetics, there is emerging evidence that, as with other viruses, viral integration preferentially targets specific genes or common chromosomal fragile sites. [91] Integrated HBV is thought to contribute to the pathogenesis of HCC through a number of potential mechanisms: (1) by local genomic effects due to production of genomic instability and focal deletions and translocations; (2) by interruption of genes, leading to overexpression, suppression, or production of novel fusion transcripts; and (3) by the transactivating effects of viral proteins, particularly the *X* and *preS* gene products. Viral integration has been shown to result in deletions, translocations, and gene amplification at the site of integration. [92] In a number of cases, HBV integration has been shown to occur adjacent to or within genes involved in cell growth or apoptosis. Genes found at HBV integration sites include the retinoic acid receptor-ß gene (*RARB*), [93] the cyclin A gene, [94] and the sarco/endoplasmic reticulum calcium ATPase (*SERCA*). [95] In the case of the *RARB* and cyclin A integrations, expression of a fusion transcript results in oncogenic effects. [96], [97] Integration of HBV into the *SERCA* gene leads to accumulation of chimeric HBx/SERCA 1 proteins that induce apoptosis. HBx protein inhibits the function of the tumor suppressor *p53* and activates cytoplasmic mitogenic signaling cascades, including the mitogen-activated protein kinase (MAPK) and the Janus family tyrosine kinase (JAK)/signal transducer and activators of transcription (STAT) pathways. HBx also interacts with transcription factors in the nucleus to transactivate multiple genes involved in cellular growth control, including the nuclear factor (NF)-?B, activator protein 1, and serum response element pathways. [98] The HBx protein has been shown to play a role in fibrogenesis. HBx interacts with *Smad4* and stabilizes the complex of *Smad4* with components of the basic transcriptional machinery, leading to enhanced transcriptional activity in response to TGF-ß. In addition, confocal microscopy studies suggest that HBx facilitates and potentiates the nuclear translocation of Smads, further enhancing TGF-ß signaling. This suggests that HBx-enhanced Smad-mediated signaling may contribute to HBV-associated liver fibrosis. [99] The hepatitis B surface and core genes and proteins may also play significant roles in hepatocarcinogenesis. It has been shown that a truncated carboxyl-terminal variant of the HBV S protein directly interacts with DNA and transactivates several cellular and viral promoters, including the *c-jun* and *c-fos* promoters. [100] HCC has also been associated with HBV strains that have deletions or insertions in the HBV core promoter region, suggesting a role for changes in core protein expression in persistence of HBV infection, HBV integration, and carcinogenesis. [101]

Hepatitis C Virus

In patients with chronic HCV viremia, persistent liver damage with abnormal alanine aminotransferase levels plays an important role in the development of HCC. This may be due to repeated cycles of liver cell injury and regeneration, resulting in premature cellular senescence and the development of genomic aberrations. However, in addition, it has been shown that individuals with persistent liver damage develop a population of HCV quasispecies that changes over time, whereas individuals with persistently normal transaminases have a stable infection with a single species of HCV. [102] Even within the same patient, tumor samples show greater variability in the core region of HCV isolates than is found in adjacent benign samples. [103] This phenomenon may be important in hepatocarcinogenesis, by allowing for selection

of more oncogenic variants of the HCV core protein. In a transgenic mouse model, HCV proteins suppress Fas-mediated hepatocellular apoptosis and may use this mechanism to maintain persistence of HCV infection. [104] Of the seven HCV proteins, core and nonstructural (NS) proteins NS2, NS3, NS4A, NS4B, NS5A, and NS5B, the core protein is the most potent cellular signaling activator, causing activation of the NF-?B, AP-1, and SRE pathways and the mitogen-activated protein kinase MAPK/ERK cascade. [105] Activation of these cascades leads to cell proliferation. The HCV core protein has been shown to interact with the 14-3-3 protein, leading to activation of Raf-1 kinase. [106] The NS4B protein also activates the NF-?B pathway, and the NS5A protein has been shown to repress the interferon-induced protein kinase, PKR. The envelope protein E2 is transcribed from the most variable region of the HCV genome and is thought to allow the virus to escape from neutralizing antibodies; in addition, E2 inhibits PKR and contributes to interferon resistance of HCV. [107]

SURVEILLANCE

Surveillance in a population assumes (1) the disease has an asymptomatic stage but if left alone will produce significant morbidity or mortality; (2) a high-risk population can be identified; (3) surveillance tests are inexpensive, noninvasive, and accurate; (4) clearly defined recall procedures exist, and (5) effective therapy is available (Table 118-1). For HCC, almost all of these criteria can be met in principle. Patients with cirrhosis from alcohol abuse, viral hepatitis, hemochromatosis, a $_1$-antitrypsin deficiency, and primary biliary cirrhosis all have an increased incidence of HCC. [108], [109], [110], [111] and [112] Asians and Africans who are chronic HBV carriers are also at significant risk for HCC. [113] In contrast, HCC is unusual in Wilson disease, autoimmune hepatitis, and primary sclerosing cholangitis. [114], [115] and [116] In high-risk cirrhotic patients, the incidence of HCC is 2% to 6%, greater than the calculated incidence of 1.5% thought to be the cut-off point in order for surveillance to be cost-effective. [117] Ultrasonography is relatively inexpensive, noninvasive, and accurate in the diagnosis of early-stage HCC, and, therefore, it is recommended that ultrasonography be used to screen for HCC at 6-month intervals in patients with cirrhosis. This radiographic technique has a sensitivity of 70% and a specificity of 90% for identifying HCC in an asymptomatic population. [118] The 6-month interval time schedule comes from data on the doubling time of these cancers. [119], [120] The utility of serum alpha-fetoprotein determinations in screening for HCC remains controversial [121]; however, an elevated serum alpha-fetoprotein does identify patients at increased risk for HCC. [122] Recall procedures (the definition of an abnormal result and the procedure for investigating abnormal results) are less well established. Most experienced clinicians would follow any lesion greater that 0.5 cm with a spiral enhanced computed tomography (CT) scan of the liver (see section, " Differential Diagnosis, Diagnosis, and Staging"). Based on this analysis, surveillance of individuals with cirrhosis for HCC is generally encouraged. However, evidence-based data for instituting screening for HCC are less compelling. Several retrospective, uncontrolled or nonrandomized trials show little benefit to surveillance. [118], [123], [124] and [125] These studies have been fraught with problems, such as being based entirely on hospital populations as opposed to non–referral-based populations, lack of established recall procedures, poor compliance, and failure to offer liver transplantation as a curative procedure for patients with small HCCs. Modeling data are more encouraging. We have developed a Markov model to assess the cost-effectiveness of screening for HCC (Fig. 118-4). The data demonstrate that screening for HCC is cost-effective and can be defended as a clinical strategy.

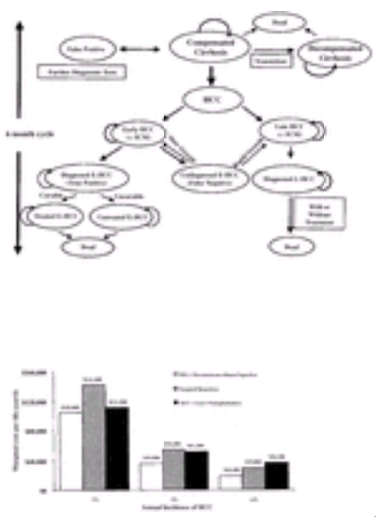

FIGURE 118-4. A Markov model for screening for hepatocellular carcinoma. The **upper panel** shows the different clinical states programmed in the model. The **lower panel** shows the cost per life-year saved for patients treated by percutaneous ethanol injection, surgical resection, or orthotopic liver transplantation.

CRITERIA	CRITERIA MET FOR HEPATOCELLULAR CARCINOMA
Asymptomatic stage	Yes
Late stages are incurable and cause mortality	Yes
High-risk population	Yes, cirrhosis
Surveillance testing is inexpensive and noninvasive.	Yes, ultrasonography
Established recall procedures	Not well developed
Effective therapy	Liver transplantation, surgical resection, ablative procedures*

*These therapies can only be applied on a selective basis, and the non-transplantation therapies are fraught with recurrent disease.

TABLE 118-1 Criteria for Instituting Population Surveilance

DIFFERENTIAL DIAGNOSIS, DIAGNOSIS, AND STAGING

The differential diagnosis of liver mass lesions is thoroughly reviewed in Chapter 47. Therefore, this section is limited to the differential diagnosis of liver mass lesions in the presence of cirrhosis. The major problem in cirrhosis is differentiating HCC from a macroregenerative nodule. Macroregenerative nodules (MRNs) are large nodules that form in the liver as part of the cirrhotic process. The presence of large MRNs is itself a risk factor for the development of HCC, and therefore, the distinction between MRNs and HCC is difficult. Indeed, current nomenclature suggests that instead of MRN the term *dysplastic nodule* be applied. [126] The imaging hallmark of HCC is increased vascularity of the lesion as compared with the surrounding tissue and is used to help make the diagnosis of HCC radiographically (Fig. 118-5). For lesions larger than 2 cm, a definite diagnosis needs to be established because the lesion is still amenable to therapy at this stage (see later). If the lesion is identified by two imaging studies and manifests contrast enhancement on spiral CT or dynamic magnetic resonance imaging (MRI) studies in the presence of cirrhosis, the diagnosis of HCC can be comfortably established. [127] Also, if the lesion is identified by two imaging studies and the alpha-fetoprotein level is higher than 400 ng/mL, the diagnosis of HCC also is likely. [127] If these strict criteria are adopted, imaging misdiagnosis is very unlikely. In a very robust study, the diagnosis of HCC was established preoperatively without biopsy with an accuracy of 99.6%, sensitivity of 100%, specificity of 98.9%, and positive and negative predictive values of 99.3% and 100%, respectively. [128] In the absence of these noninvasive criteria for the diagnosis of HCC (Table 118-2), histologic or cytologic verification of the malignancy should be obtained. However, the risk for tumor seeding has been estimated to be 3.4% to 5.1%, [129], [130] and a biopsy should only be performed if it will change therapy. In many instances, expert clinicians may elect to proceed with surgery or an ablative therapy based on the presence of a clear-cut mass lesion in a cirrhotic liver because it is difficult to interpret a negative biopsy. Lesions can be missed despite ultrasound guidance of the biopsy, and it is extremely difficult for the pathologist to distinguish between a dysplastic nodule and an early well-differentiated HCC. Finally, given the propensity of dysplastic nodules to evolve into cancer, ablative therapy for a dysplastic nodule is not necessarily inappropriate. The detection of small (less than 1 cm) hypoechoic or hyperechoic nodules during ultrasonography of the liver should raise the suspicion of HCC. Hypoechoic nodules are consistent with neoplastic growth of tissue, whereas hyperechoic nodules suggest the presence of steatosis. It is now well established that many small HCCs contain fat, which is subsequently lost as the cancer progresses. [131] Therefore, both hypoechoic and hyperechoic lesions must be considered malignant in a cirrhotic liver until further characterized. However, pathological studies have shown that only about half of such nodules smaller than 1 cm are neoplastic. These small lesions, unfortunately, are too small to assess their vascularity by imaging studies or to obtain tissue reliably for pathological examination. It is recommended that these lesions be followed with serial imaging studies at least 3 to 4 months apart. Growth of the lesion or the development of contrast enhancement on cross-sectional imaging studies would signify the development of HCC. Ultrasound and spiral CT scans are

the standard imaging modalities for the follow-up of these early and diagnostically challenging lesions.

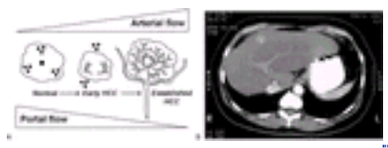

FIGURE 118-5. A: Schematic representation of arterialization during hepatocellular carcinoma development. **B:** Computed tomography scan showing contrast enhancement of an early hepatocellular carcinoma. This pathological feature of hepatocellular carcinoma allows more accurate detection of malignant nodules. The technique of transarterial chemoembolization also relies on the preferential arterial vascularization of hepatocellular carcinomas as compared with the normal hepatic parenchyma.

Radiographic criteria
Two concident imaging studies
Arterial hypervascularization
Combined criteria
One imaging study with hypervascularization
Alpha-fetoprotein >400 ng/mL

TABLE 118-2 Noninvasive Criteria for Diagnosis of Hepatocellular Carcinoma

Natural History

An understanding of the natural history of HCC is necessary to interpret the results of the published treatment trials. The natural history of HCC is dependent on the severity of the underlying cirrhosis, tumor characteristics (i.e., size, multicentricity, presence or absence of vascular invasion, pathological grade, and metastases), symptoms as manifest by the performance status of the patient, comorbid diseases, and the efficacy of treatment interventions. Four prognostic classifications for HCC have been developed (Table 118-3). All of these are imperfect, in part, because they do not take into account specific information on the tumor (e.g., information on the genetic changes or specific mechanisms involved in pathogenesis of the tumor); are based on information at the time of the initial diagnosis and, therefore, do not incorporate changes in prognostic variables over time; and are based on retrospective analysis. The first was developed by Kunio Okuda and co-workers and is simply known as the Okuda classification. [132] This classification takes into account hepatic factors (bilirubin, albumin, and ascites) and a single tumor factor (extent of liver replacement more 50% or less than 50%). Given its relatively gross assessment of tumoral factors and absence of patient health parameters, this classification is seldom used in clinical assessment. The other three prognostic classifications take into account more refined assessments. The Barcelona Clinic Liver Cancer (BCLC) prognostic assessment takes into account the patient performance status, presence or absence of constitutional symptoms, vascular invasion, and extrahepatic spread. [133] The Cancer of the Liver Italian Program (CLIP) uses the Child-Turcotte-Pugh Score, tumor size and multicentricity, vascular invasion, and serum alpha-fetoprotein to establish a prognosis. [134], [135] The Group d'Etude et de Traitement du Carcinoma Hepatocellulaire (GETCHC) takes into account the Karnofsky index, serum bilirubin, alpha-fetoprotein, serum alkaline phosphatase, and vascular invasion. [136] It is clear from an assessment of these models that vascular invasion, poor performance status, and an elevated alpha-fetoprotein with tumor multicentricity are uniform poor prognostic parameters. More importantly, these studies also predict which patients will do well for extended periods of time. For example, using data from patients randomized to no treatment in well-designed randomized control trials, the BCLC group found that patients with unicentric disease, no symptoms, and no evidence of vascular invasion nor extrahepatic spread had a 50% 3-year survival rate [137] (Fig. 118-6). This subgroup of patients with HCC are frequently those selected for locoregional therapies (see later), and the survival in the treatment groups is frequently no better than that in this untreated group of patients (Fig. 118-7). This information on the natural history of HCC needs to be kept in mind when counseling patients and helping them select treatment options.

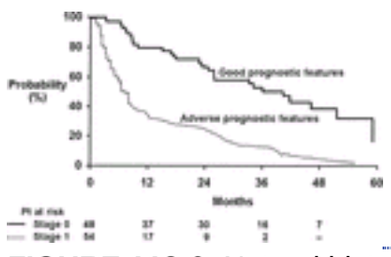

FIGURE 118-6. Natural history of hepatocellular carcinoma. Patients with good prognostic factors and early detection of hepatocellular carcinoma have a significantly better short- to mid-term outcome compared with patients with adverse prognostic features at the time of diagnosis. (Modified from ref. [137].)

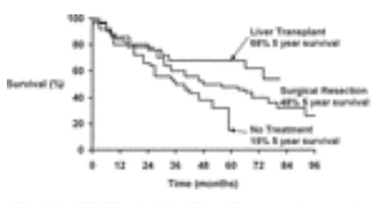

FIGURE 118-7. Results of surgical resection and orthotopic liver transplantation for hepatocellular carcinoma. Survival after surgical resection is limited by the development of recurrent hepatocellular carcinoma in the remaining cirrhotic liver. Liver transplantation results in improved long-term survival because of replacement of the liver with a noncirrhotic donor liver. (Adapted from ref. [9].)

CLASSIFICATION	PARAMETERS
BCLC	Performance status
	Constitutional symptoms
	Vascular invasion
	Extrahepatic spread
CLIP	Child-Turcotti-Pugh stage
	Tumor size or nodularity
	Vascular invasion
	Alpha-fetoprotein ≥400 mg/mL
GETCHC	Karnofsky index <80%
	Bilirubin ≥3 mg/dL
	Alpha-fetoprotein >35 mg/mL
	Alkaline phosphatase > 2 × upper limit of normal
	Vascular invasion
Okuda	Ascites
	Tumor size >50% of liver
	Albumin <3 g/L
	Bilirubin >3 mg/dL

TABLE 118-3 Prognostic Stage Classification Systems for Hepatocellular Carcinoma

Clinical Manifestations

With better institution of surveillance strategies, most HCCs will be identified at an asymptomatic stage. However, many patients continue to present with advanced HCC. The clinical manifestations of advanced HCC are protean and include decompensation of cirrhosis, tumoral symptoms, acute abdominal catastrophe, cholestasis, fever of unclear etiology, paraneoplastic phenomena, and the metastatic presentation (Table 118-4). HCC may result in decompensation of cirrhosis by vascular invasion of the major portal vein or its large branches. This event worsens portal hypertension and frequently results in the development of ascites and refractory variceal bleeding. Infiltrating cancers in the liver also increase portal hypertension at the level of the sinusoids, promoting ascites formation and variceal bleeding. Also, the diffuse infiltrating cancers further worsen hepatic function, resulting in hyperbilirubinemia, coagulopathy, and encephalopathy. Even small cancers may be associated with hepatic decompensation because either advanced cirrhosis is frequently associated with HCC or the small cancers have effects on the immune system or hepatic function in general. For example, about 20% of patients hospitalized with spontaneous bacterial peritonitis have HCC. [138] Because of this association, all hospitalized cirrhotic patients should have appropriate imaging studies to look for HCC.

PRESENTATION	CLINICAL SYNDROME
Tumor	Malaise, anorexia, weight loss, night sweats
Hepatic decompensation	Jaundice, ascites, variceal bleeding
Acute abdominal pain	Tumor bleeding, capsular extension
Pyrexia	Fever and leukocytosis
Metastases	Brain, lung, or bone metastases
Cholestasis	Large bile duct obstruction or diffuse infiltrating cancer
Paraneoplastic	Increased red blood cell mass, Ca^{++}; decreased glucose; diarrhea

TABLE 118-4 Clinical Manifestations of Symptomatic Hepatocellular Carcinoma

Tumoral and acute presentations of HCC are less common than the presentation of hepatic decompensation. The tumoral presentations include anorexia, weight loss, general malaise, and painful hepatomegaly. The acute presentation is associated with spontaneous rupture of the cancer into the peritoneum or extension through the capsule. Mortality is high with hemoperitoneum due to exsanguination. Selective acute arterial embolization of the bleeding artery is the preferred treatment for this complication. Patients with pain from tumor extension through the capsule likely have pain from this rupture event. The pain is usually acute, limited in time (less than 24 hours), and assumed to be biliary colic. The screening ultrasound of the liver often identifies unexpectedly a liver mass when the patient seeks medical attention.

Cholestasis may result from tumor-associated extrinsic compression of the large bile ducts, tumor invasion of the large bile ducts, or miliary metastasis throughout the liver. Tumor invasion of the bile ducts can be associated with hemobilia and gastrointestinal bleeding. [139] In our experience, the miliary pattern of metastases is usually associated with disproportionately high serum transaminases in addition to hyperbilirubinemia and jaundice.

Fever of unknown etiology is an unusual presentation of HCC. [140] Interestingly, the febrile episodes may be discrete episodes and associated with leukocytosis. Tumors need not be large to generate this symptom complex. Given the frequent difficulty in diagnosing HCC by a single imaging study, multiple imaging studies may be necessary to make the diagnosis of HCC in this context. In contrast to fever, night sweats are common in advanced HCC and portend a poor prognosis.

Paraneoplastic phenomena associated with HCC are rare but include hypercalcemia, erythrocytosis, hypoglycemia, thrombophlebitis migrans, arterial hypertension, and the syndrome of watery diarrhea, hypokalemia, and achlorhydria. The hypercalcemia is due to production of a parathyroid hormone–like molecule, the erythrocytosis is due to erythropoietin generation by the tumor, and hypoglycemia is mediated by insulin-like growth factors. [141] Arterial hypertension is uncommon in advanced cirrhosis, and its presence, therefore, should prompt imaging studies for HCC. In addition to the rare syndrome of watery diarrhea, hypokalemia, and achlorhydria, diarrhea without the electrolyte abnormalities is very common in some populations with HCC. [141] In children with hepatoblastomas, isosexual precocity may be a paraneoplastic syndrome. [141] In adults, feminization and gynecomastia are also associated with HCC, but these syndromes are often present in cirrhosis without HCC and are, therefore, difficult to attribute to HCC. Cutaneous manifestations of HCC are equally rare and include dermatomyositis, pemphigus foliaceus, and Leser-Trélat sign. The latter is the sudden onset of seborrheic dermatitis and freckles, with a rapid increase in the number of the lesions accompanied by pruritus. Pityriasis rotunda (round or oval hyperpigmented scaly lesions) may be associated with HCC in South African blacks. [141]

Metastatic complications of HCC are not dissimilar from the metastatic complications of other neoplastic diseases. However, HCC does have some unique metastatic presentations. One metastatic complication of HCC is dyspnea due to tumor pulmonary emboli, and another is headache associated with pituitary metastases. [142] Common sites for HCC metastases include the diaphragm with associated pleural effusions, bone metastases with pain, and regional lymph node metastases, which may be associated with abdominal discomfort. In general, abdominal pain in patients with HCC is a poor prognostic symptom and usually indicates advanced disease.

Staging of Hepatocellular Carcinoma for Selection of Therapy

The staging procedure depends in part on the intensity of the selected treatment. All patients should undergo a chest radiograph to exclude pulmonary metastases and at least two imaging studies to stage intrahepatic disease and exclude vascular invasion (ultrasound, contrast CT, contrast MRI); in addition, many centers will perform a CT scan of the chest and a bone scan if liver transplantation is being contemplated. It is important to note that abdominal lymphadenopathy is common in cirrhosis, especially perihilar lymph nodes and even more generalized lymphadenopathy in hepatitis C. Thus, one has to be very cautious in attributing lymphadenopathy to metastatic disease. In suspicious cases, we have found endoscopic ultrasound with fine-needle aspiration (FNA) of hypoechoic lymph nodes useful for detecting metastatic involvement of lymph nodes. Finally, the dilemma of what to do with a small HCC remote from a portal vein thrombosis remains problematic. If tumor thrombosis is present, radical or ablative therapies are not effective; if the thrombus is bland, then these therapies may be contemplated. Many experienced centers use color-flow Doppler and ultrasound-guided FNA of the thrombus to make a decision. [143], [144] and [145] The appearance of an arterial signal on the color-flow Doppler is most indicative of tumor thrombus because these lesions are frequently arterialized despite their residence in a vein. A negative color-flow Doppler along with a negative FNA would be strong evidence in favor of a bland thrombus.

LOCOREGIONAL THERAPIES

Locoregional approaches include surgical and percutaneous ablative techniques. The outcome of these therapies for HCC is dependent on several readily identifiable prognostic factors. Poor prognostic factors identified in numerous studies include unicentric tumors larger than 5 cm, multicentric disease with more than three 3 lesions, satellite lesions adjacent to the main tumor (a marker for tumor invasion and therefore metastatic potential), and vascular invasion. [133] In particular, invasion of the main portal vein or one of its segmental branches, a striking feature of HCC, is a relative contraindication to radical therapy.

Surgical Approaches

Surgical resection is commonly accepted as the initial treatment of choice for localized HCC worldwide. Despite the acceptance of surgery for the treatment of HCC, there are no randomized controlled trials demonstrating its efficacy. This approach is limited in its application because few patients are suitable for partial hepatectomy because of either tumor size, vascular invasion, multicentricity, the presence of portal hypertension, or overall hepatic functional reserve. Intrahepatic recurrence of HCC is common among patients who undergo apparent curative resection because the neoplastic potential of the nonresected liver remains unchanged. Indeed, the 5-year recurrence rate is 52% because of either de novo tumor recurrence or the progression of previously unrecognized intrahepatic metastases. [9] Hepatic decompensation is high in patients with any elevation of the serum bilirubin or evidence of portal hypertension [146] (Fig. 118-8). A wedged hepatic venous pressure gradient above 10 mm Hg is considered to be a contraindication to resection. However, few liver centers routinely perform this test; instead, many surgeons use a platelet count of less than 100,000 per µl or the presence of varices (indicative of a wedged hepatic venous pressure gradient of at least 12 mm Hg) as contraindications to surgery. Operation-related mortality is also high, especially in patients with limited hepatic functional reserve. Partial hepatec- tomy should therefore be considered only in patients without cirrhosis or in those with mild liver disease (Child A cirrhosis), normal portal pressure, and normal bilirubin. [133] The best outcomes are observed in patients with unicentric disease, no vascular invasion, tumors smaller than 5 cm, and relatively inactive liver disease. Given these selection criteria, it is estimated that only about 5% of patients with cirrhosis and HCC are candidates for surgical resection. [147] In this highly selected patient

population, the best reported outcomes are 50% at 3 years. [133] This outcome is not that much different from the natural history of untreated, asymptomatic, small, unicentric disease without vascular invasion or that obtained with ablative techniques (see earlier). Because of these observations, many hepatologists seldom advocate resection for HCC except in ideal candidates. However, if appropriate adjuvant therapies or secondary chemopreventive agents could be developed, surgery would be a more attractive option. Along these lines, polyprenoic acid has been shown in a single study to decrease recurrence rates after surgical resection of HCC. [148] If these studies can be confirmed, the use of this agent should enhance outcomes after surgery or ablative therapy.

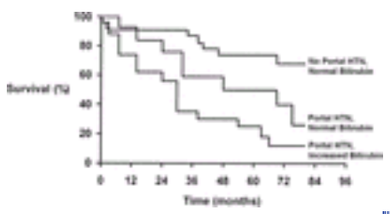

FIGURE 118-8. Hepatic decompensation leads to a worse outcome after surgical resection for hepatocellular carcinoma in patients with clinically significant portal hypertension (HTN) and an increased serum bilirubin. (Modified from ref. [9].)

Orthotopic liver transplantation (OLT) for patients with HCC offers several advantages over partial hepatectomy: (1) it can be employed in patients with all stages of liver disease; and (2) the cancer, the neoplastic potential of the underlying liver disease, and the liver disease itself are all therapeutically addressed at the same time. Indeed, numerous studies have now demonstrated that in patients with unicentric disease less than 5 cm or with multicentric disease of up to three lesions and each lesion 3 cm or smaller, the results with liver transplantation are superb (see Fig. 118-8). For example, a study of 120 cirrhotic patients with HCC showed that transplanted patients with a maximum of two small (less than 3 cm) nodules had improved survival without recurrence (83% at 3 years) compared with patients treated with partial hepatectomy (18% at 3 years) for tumors of the same size. [149] One large recent series found that HCC less than 5 cm in diameter or the presence of three or fewer tumors with a maximal diameter of 3 cm was associated with overall survival rate of 85% and disease-free survival rate of 92% at 4 years. [150] Thus, transplantation centers throughout Europe and the United States have adopted these criteria in HCC patients for OLT. However, limited studies have suggested that unicentric tumors with a capsule and without vascular invasion of up to 6.5 cm may do equally well with OLT. [151] In addition, data from Germany suggest that very-well-differentiated large tumors have acceptable outcomes after OLT. [152] Thus, the current guidelines of one lesion less than 5 cm or three lesions all less than 3 cm are restrictive criteria developed to ensure the best outcomes and need to be reevaluated as more information becomes available. The most important limitation of OLT is an increasing organ shortage, which is problematic for several reasons. OLT for HCC removes livers from the common donor pool and may thus aggravate the situation for patients with end-stage cirrhosis. In addition, a long waiting period for a donor organ often allows a small tumor to become large or even metastatic. Indeed, the dropout rate for patients initially meeting criteria for OLT may be as high as 25% during 6 months of waiting time in the absence of therapy. [9] However, these patients were selected on the criteria that all lesions had to be identified by two imaging modalities, resulting in an understaging bias. In contrast, most transplantation centers in the United States have an overstaging bias, which is to take the maximum number of lesions identified on any given study to be the actual number of tumors. Nonetheless, given the increasing waiting times for OLT, most centers prefer to treat the lesions before OLT to prevent tumor growth and dissemination. Ablative approaches and chemoembolization (CE) therapies are employed (see later). None of these therapies has been subjected to randomized controlled trials in combination with OLT; therefore, the optimal approach remains to be ascertained.

In summary, tumor criteria identifying patients with HCC who are most likely to benefit from surgical approaches (resection and transplantation) have now been established. Many experts feel that patients meeting criteria for surgical approaches should be offered this option as definitive therapy. In patients who are candidates for both resection and transplantation, the best outcomes are obtained with transplantation because of the high recurrence rate after liver resection. [9] However, if effective secondary chemoprevention therapies can be established, surgical resection may become preferred because it avoids the risks and need for long-term immunosuppression. For patients who are not candidates for surgical approaches, locoregional forms of treatment are reasonable. These options include percutaneous alcohol injection (PEI), radiofrequency ablation (RFA), and transcatheter arterial chemoembolization and are discussed in the following paragraphs.

Percutaneous Alcohol Injection

PEI has become a widely accepted form of therapy for small, localized HCC. The advantage of PEI over other local ablative minimally invasive therapies is its relatively simple technique and low cost. PEI can be safely performed in patients who are not candidates for resection or OLT, even in the presence of advanced cirrhosis. [153] It is generally agreed that HCC less than 3 cm in size and fewer than three nodules is suitable for PEI. The destruction of tumor cells by injection of absolute alcohol is probably due to a combination of cellular dehydration, coagulative necrosis, and vascular thrombosis followed by tissue ischemia.

For PEI, sterile ethanol (95%) is slowly instilled throughout the tumor nodule under ultrasound or CT guidance. Because it is important to achieve uniform distribution of the ethanol, the injecting needle is usually placed at the far end of the lesion and then withdrawn in a stepwise fashion during the injection. For small lesions, a single treatment is often sufficient, whereas larger lesions may require several treatments over a number of days. The extent of necrosis achieved is dependent on tumor size. Small nodules less than 3 cm in diameter are usually completely destroyed, larger tumors only partially. [154], [155] Patients should therefore be followed regularly with spiral CT scans. PEI can be repeated for local recurrences and new lesions within the liver. PEI is associated with a complication rate of 1.7% and a 0.1% mortality rate. Patients may experience abdominal pain, which is often due to leakage of ethanol into the peritoneal cavity and can be prevented by leaving the needle inside the lesion for 30 seconds or more after injection. A low-grade fever may be present for 1 to 3 days after the procedure as a result of tumor necrosis. Contraindications to PEI include massive ascites, coagulopathy, and obstructive jaundice, which increase the risk for postprocedural bleeding and bile peritonitis.

The long-term survival rate is as high as 70% in patients with cirrhosis undergoing PEI for a single HCC less than 3 cm in diameter. [154], [155] In the largest series published, Livraghi and colleagues [153] reported survival rates in 746 cirrhotic patients with HCC treated with PEI. In patients with a single tumor less than 5 cm in diameter (*n* = 462), the 3- and 5-year survival rates were 79% and 47%, respectively, for Child A patients, 63% and 29% for Child B patients, and only 12% and 0% for Child C patients. In patients with Child A cirrhosis, survival was much lower in those with multiple HCCs (35% at 5 years), single HCCs larger than 5 cm (30% at 5 years), and advanced HCCs (0% at 5 years). Unfortunately, there have been no prospective randomized controlled trials comparing PEI to surgical resection or transplantation for early stage HCC. In a large retrospective study from Japan in which patients were matched for severity of the liver disease and stage of tumor, there was no significant difference in survival between patients treated with PEI and those who underwent surgical resection. [156] Thus, long-term survival after PEI may be equivalent to survival after surgical resection in a selected subgroup of patients with small tumors and mild cirrhosis. Similar to the results after surgical resection, tumor recurred in 50% of patients after PEI. [157] In summary, PEI is a safe, simple, inexpensive treatment for patients with HCC. Although relatively few reports of long-term results are available, PEI seems to be an effective method for patients with single, small tumors. For this subgroup of patients, PEI might be equal to surgical resection, with fewer complications and less procedure-related morbidity and mortality. However, more controlled studies comparing PEI to other treatment modalities are needed to better define its role in the management of HCC. Treatment of larger lesions is also possible but is less likely to ablate the tumor completely and is therefore associated with higher rates of relapse and decreased survival rates.

Radiofrequency Ablation

Radiofrequency ablation (RFA) is emerging as a new therapeutic method in the management of HCC. The first papers reporting ultrasound-guided RFA of hepatic tissue were published in 1990 and suggested that RFA could prove useful to create focal coagulative necrosis of hepatic tumors while sparing normal liver tissue. [158] Significant advances in equipment design have led to increasing success in the use of this method in the management of HCC. RFA destroys tissue by thermal energy generated with an alternating electric current generator that operates in the radiofrequency range from 200 to 1200 kHz. Like PEI, RFA is usually performed by a percutaneous approach in the outpatient department. After local anesthesia, a needle electrode is introduced through the skin under ultrasonographic guidance and advanced into the center of the tumor. Treatment is guided by impedance measurements between components of the needle electrode. The goal of each procedure is to ablate the tumor and a small margin of normal tissue around the tumor. Depending on the equipment used, this can be achieved in one or two short sessions (less than 15 minutes) for lesions up to 5 cm. Because large blood vessels act as a heat sink, RFA is not effective for lesions adjacent to large veins in the liver. It is also more technically difficult to treat lesions in segment 8 (lesions high in the dome of the liver) with RFA than with PEI. Spiral contrast-enhanced CT scanning is usually used to assess tumor necrosis after RFA, since ultrasound is usually not sensitive enough for this purpose. RFA can be repeated for residual tumor found on

follow-up or for new tumor nodules that develop in the liver.

Although the experience with RFA is still limited, the first results are promising. In 1995, Rossi and colleagues [159], [160] reported initial results from 24 patients with one or more tumor nodules smaller than 3 cm that were treated with intent to cure. Complete tumor necrosis was achieved in all but two patients after one session; both patients successfully received additional thermal ablations. About half the patients had recurrence of the tumor during follow-up, which could be retreated in 70% of these patients. The survival rate was 95% the first year, 84% the second year, 67% the third year, and 45% the fourth and fifth years. Livraghi and associates [161] used a large cohort of patients (*n* = 112) with small HCCs (less than 3 cm in diameter) to compare the effectiveness of RFA with that of PEI. Complete tumor necrosis was achieved in 90% of tumors with RFA in an average of only 1.2 sessions per tumor and in 80% of tumors with PEI in 4.8 sessions. However, in contrast to other series, one major and four minor complications were reported in patients treated with RFA; in contrast, no complications occurred in patients treated with PEI. Finally, a recent study suggests RFA may be associated with a high rate of needle track seeding (12.5%) because of the large needle used in this approach. [162] It is not clear whether this result is generalizable.

In conclusion, RFA appears to be a fast, safe, and effective technique for ablation of small liver tumors. Larger controlled studies are needed to determine whether RFA, rather than PEI, should be the method of choice for small HCCs. Nonetheless, because it usually requires fewer sessions, is better tolerated, and has more easily assessed treatment margins, RFA is the preferred choice in many centers. Until more information becomes available, the availability of experience and equipment in individual centers, location of the lesion within the liver, and closeness of the tumor to large vessels should determine the method used for treatment.

Transcatheter Arterial Chemoembolization

CE is a technique that uses angiography to embolize selectively the arterial supply to an HCC. A cytotoxic agent such as doxorubicin or cisplatin is mixed with lipiodol, Gelfoam, or Ivalon particles to form a suspension that is then injected into the feeding artery of the tumor by angiographic technique through the common femoral artery. This results in both selective ischemic and chemotherapeutic effects on the HCC. Because of the dual blood supply of the liver, CE causes minimal damage to normal liver parenchyma; 80% to 100% of the blood flow to liver tumors but only 20% to 30% of the flow to normal liver is supplied by the hepatic artery. After the procedure, the suspension remains in the tumor, where the cytotoxic agent is slowly released to exert its effect. Although lipiodol serves a dual role in CE by carrying the chemotoxic agent to the tumor and also embolizing the vasculature, it can remain in the lesion for prolonged periods of time and preclude adequate radiographic assessment of tumor necrosis by contrast-enhanced CT scanning. The relative contribution of the chemotherapeutic agents to treatment of HCCs, compared with the simple ischemic effect of occluding the tumor vasculature, has never been established.

Risks associated with CE include contrast allergy, contrast-induced nephropathy, and bleeding or pseudoaneurysm formation at the catheter site. Most patients develop a postembolization syndrome with fever and right upper quadrant pain and are therefore hospitalized. Liver function tests can be elevated for up to 2 weeks. Major complications include hepatic abscess, which is most common in patients with a preexisting biliary-enteric anastomosis; hepatic artery dissection or thrombosis; and ischemic cholecystitis. The morbidity rate of CE was 5.1% and the treatment-related mortality rate 4.1% in one retrospective study. [163] Advanced Child C cirrhosis is a relative contraindication to CE because of the high risk for potentially fatal complications such as liver failure. Other contraindications include hepatic encephalopathy, biliary obstruction, portal vein occlusion, a surgical portosystemic shunt, a transjugular intrahepatic portosystemic shunt device, bilirubin higher than 5 mg/dL, International Normalized Ratio higher than 1.7, or creatinine higher than 1.8.

The efficacy of CE is determined by the size of the lesion. One study reported a survival rate of 100% at 3 years for tumors less than 2 cm in diameter, whereas 3-year survival was 0% for tumors greater than 5 cm. [164] Because CE also avoids the risk for needle track seeding, transplantation centers often use CE as a bridge to transplantation for HCC patients. CE prevents progression of the lesions and potential micrometastases while the patient waits for a donor organ. In a pilot study at our institution, none of the transplant recipients pretreated with CE has developed recurrent HCC during a mean follow-up of more than 2 years. [165] However, the role of CE as a palliative procedure remains controversial. Indeed, one large controlled study that compared CE to conservative treatment for unresectable HCC of any size was stopped because an interim analysis failed to show the expected benefit of CE. [166] These results have been confirmed by several other randomized controlled trials, which did not show improved survival in patients with nonsurgical HCCs. [167], [168] and [169] Based on the current evidence available in the published literature, CE cannot be recommended for the treatment of large, unresectable HCC.

SYSTEMIC THERAPY

Four forms of systemic therapy have been applied to the treatment of advanced HCC, including cytotoxic therapy, immunotherapy, hormonal therapy, and gene therapy.

Cytotoxic Chemotherapy

HCC is extremely refractory to current available cytotoxic therapies. Moreover, the presence of cirrhosis with impaired clearance of drugs metabolized by the liver increases drug toxicity. Portal hypertension with leukocytopenia and thrombocytopenia also exacerbate drug-induced bone marrow toxicity. The overall response rate is less than 20% for systemic chemotherapy given intravenously using either single or combined agents. [170] Intra-arterial therapy also has not been shown to affect disease progression or survival. [171] Thus, there is no current proven chemotherapeutic regimen for HCC.

Hormonal Therapy

The presence of androgen receptors in HCC, the sexual dimorphism of the liver, and the male predominance of HCC led to the use of antiandrogen therapy for this cancer. Unfortunately, studies were negative, and this approach was abandoned. [172] The presence of estrogen receptors in the liver also led to the use of tamoxifen (an antiestrogen) for the treatment of this disease. Initial studies suggested a survival benefit. [173] However, several randomized controlled trials and a metaanalysis of these trials have shown no survival benefit. Finally, HCCs express somatostatin-2 receptors, and administration of somatostatin appeared to improve survival of patients with advanced HCC in a pilot study. [174] Larger randomized trials are needed to evaluate further the use of somatostatin in HCC.

Immunotherapy

Two forms of immunotherapy have been applied in HCC, using systemic therapy with interferon or employing lymphokine-activated natural killer cells. Although interferon was superior to no treatment or doxorubicin (Adriamycin) in one study, this result was not reproduced in a second study. [175] Further, the administration of interferon in doses required for antitumoral effects was associated with a high rate of adverse side effects, necessitating discontinuation of therapy. Lymphokine-activated killer cell therapy is associated with significant toxicity, is expensive, and is difficult to implement. [176] Results with this approach have also been modest, and, therefore, it has not been adopted as a therapeutic option.

Gene Therapy

Gene therapy is rational for HCC because a transient transfection (e.g., liposome-mediated transfer of plasmid DNA constructs) or transduction (e.g., administration of a virus expressing the desired construct) may be sufficient to destroy the tumor cells. It also may be possible to effect selective tumor targeting using the alpha-fetoprotein promoter or angiography. In addition, bystander effects have been observed with all gene therapy strategies, indicating that not all cells have to be transduced. However, these goals have yet to be realized in the clinical context. Several trials, including those attempting to transfect HCC cells with wild-type *p53,* have evolved to human studies, but the results have been disappointing so far. Nonetheless, it is premature to abandon this approach, and further developments are likely.

SUMMARY

The selection of an appropriate treatment strategy for patients with HCC depends on careful tumor staging and assessment of the underlying liver disease (Fig. 118-9). All patients with localized HCC (involvement of one single lobe, no vascular invasion or extrahepatic disease) should be evaluated for the potentially curative therapy options of partial hepatectomy or OLT. Candidates for partial hepatectomy must have no liver disease or Child A cirrhosis, normal portal pressure, and normal serum bilirubin. For patients not meeting these criteria, OLT should be considered if there is a solitary lesion smaller than 5 cm or three or fewer lesions smaller than

3 cm. Local ablative therapies, such as PEI, RFA, and CE, offer palliation for patients with contraindications for surgical approaches. PEI and RFA are minimally invasive and can be used on an outpatient basis, usually for tumor nodules smaller than 3 cm. When used for small tumors, the survival rates can be similar to those achieved by partial hepatectomy. CE may be used as an interim treatment for patients waiting for OLT. Given the paucity of efficacy data, there are no proven systemic chemotherapy regimens, immunotherapy approaches, or hormonal therapies that can be recommended at this time.

FIGURE 118-9. Management algorithm for curative treatment of hepatocellular carcinoma.

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CHAPTER 119

David S. Raiford

LIVER ABSCESS

AMEBIC VERSUS PYOGENIC ABSCESS

This chapter describes the clinical presentation, imaging characteristics, laboratory findings, natural history, and response to treatment of hepatic abscess. Although there is considerable overlap in these features between amebic and pyogenic abscesses, the differences in epidemiology, associated conditions, treatment, and prognosis underscore the need for the physician to distinguish these entities. Effective management depends critically upon prompt and correct definition of the abscess type.

EPIDEMIOLOGY

There are important differences in the epidemiology of amebic abscess, as it presents in the United States, and pyogenic abscess. Because intestinal amebiasis is a necessary prelude to hepatic amebic abscess, patients with amebic abscess typically have emigrated from or traveled to areas where intestinal amebiasis is prevalent. [1] Ethnicity of the local patient population is important. For example, in a large series from Los Angeles, 92% of 457 patients with amebic liver abscess had Hispanic surnames, compared with 37% of the local patient population. [2] In contrast, ethnicity of patients with pyogenic abscess did not differ from the general hospital population. Another difference identified between amebic and pyogenic liver abscess in this series was a striking male predominance (84%) in those with amebic abscess, whereas the gender distribution in those with pyogenic abscess was about equal. The median age of patients with amebic abscess (28 years) was significantly lower than that of those with pyogenic abscess (44 years). [2]

Since the classic review of Ochsner and associates [3] in 1938, and after the introduction of antibiotics in the 1940s, the frequency of pyogenic liver abscess associated with intraabdominal infection has diminished. Although pyelophlebitis (mostly due to appendicitis) accounted for 43% of cases of pyogenic liver abscess in the first decades of the 20th century, [3] a series of patients from 1973 to 1993 indicated that in only 7% of cases was pyogenic liver abscess believed due to portal bacteremia. [4] During the past few decades, biliary obstruction, both benign and malignant, has emerged as the most common etiology of pyogenic liver abscess, now accounting for 50% to 60% in several recent series. [4], [5] and [6] This trend undoubtedly reflects improvements in early diagnosis and management of extrabiliary abdominal infections attributable to modern imaging modalities and use of antibiotics. Importantly, over time, the incidence of cryptogenic hepatic abscess has remained stable at 15% to 20% of cases. [3], [4], [7] Several series suggest that diabetes mellitus may predispose to pyogenic liver abscess because 15% to 25% of cases occur in patients with diabetes. [2], [4], [5], [8] Finally, since the 1980s, it has become apparent that recipients of liver transplant allografts have an elevated risk for pyogenic liver abscess. [9]

CLINICAL MANIFESTATIONS

Fever and right upper quadrant pain are the principal symptoms of hepatic abscess, both amebic and pyogenic. Fever is evident in virtually all patients. Although spiking fever and chills favor pyogenic abscess, these may be seen with amebic abscess. Pain is reported by 75% to 90% of patients, is usually constant, is of variable intensity, and may exhibit pleuritic features with radiation to the right shoulder if diaphragmatic involvement is present. [2], [10], [11] and [12] Most patients have symptoms for less than 2 weeks before seeking medical care. Nonspecific symptoms, such as weakness, anorexia, nausea, and weight loss, are common. About one third of patients with either type of liver abscess report diarrhea, and one fourth have a nonproductive cough.

On physical examination, sequential measurements of body temperature should be made to detect fever. Hepatic enlargement and tenderness are typical, but not invariably present. Jaundice is rare in amebic abscess and, when present, should suggest biliary tract obstruction with pyogenic infection or underlying chronic liver disease. Percussion dullness, diminished breath sounds, or other chest findings at the right lung base are evident in 20% to 30% of patients and suggest involvement of the superior portion of the right hepatic lobe. Occasionally signs of weight loss, dehydration, and anemia are evident. [2], [4], [11], [13]

DIAGNOSIS AND DIFFERENTIAL

Imaging Modalities

The clinical constellation of fever, right upper quadrant discomfort, and hepatic enlargement with tenderness should prompt an imaging study early in the diagnostic assessment. Depending on the age of the patient and level of clinical suspicion for cholelithiasis or biliary obstruction, either ultrasonography (US) or computed tomography (CT) will be performed. These techniques facilitate discrimination of liver abscess from cholecystitis, bile duct obstruction, or pancreatitis. Radionuclide liver-spleen scanning after administration of ^{99m}Tc sulfur-colloid has been supplanted by US and CT because the latter two are more sensitive for small lesions (less than 3 cm) and offer better precision in localizing lesions that may require percutaneous aspiration or drainage. Of note, lesions near the dome of the right hepatic lobe may be difficult to visualize by US. Typically, on US, an abscess appears as a rounded, hypoechoic mass, sometimes with internal echoes [14], [15] (Fig. 119-1). CT scanning with intravenous contrast identifies an abscess as a low-density lesion (Fig. 119-2), often with peripheral enhancement, and may provide better definition of extrahepatic pathology associated with pyogenic abscess (e.g., appendiceal or diverticular abscess). [16] Although both modalities are sensitive for detecting abscesses and biliary obstruction, neither can distinguish reliably between amebic and pyogenic abscesses. [17], [18] and [19] Although less widely used, magnetic resonance imaging (MRI) also has high sensitivity for detection of hepatic abscess. Characteristically, liver abscesses are hypointense on T1-weighted and hyperintense on T2-weighted images, and wall enhancement soon after gadolinium infusion is typical. [20] Thus, MRI may provide complementary data if US and CT findings are ambiguous or when coronal images will help guide management of a lesion in the superior portion of a hepatic lobe (Fig. 119-3). Most abscesses, both amebic and pyogenic, occur in the right hepatic lobe. The presence of multiple abscesses strongly suggests pyogenic infection, as does identification of concomitant biliary tract obstruction. Chest radiographs in patients with an abscess adjacent to the diaphragm may show elevation of the right diaphragm, subpulmonic effusion, and right lower lobe atelectasis or infiltrate. It is well to remember that hepatic tumors may present with necrosis and secondary infection, [21] mimicking a primary abscess.

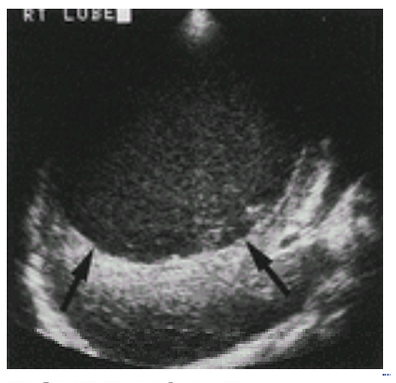

FIGURE 119-1. Transverse sonogram of the right hepatic lobe showing a large hypoechoic amebic abscess (*arrows*). This lesion is homogeneous, as is typical, although any ultrasonographic pattern may occur. The image is from a 26-year-old man who had strongly positive amebic serology and recovered rapidly with metronidazole treatment. (From Reynolds TB. Liver abscess. In: Kaplowitz N, ed. Liver and biliary diseases. 2nd ed. Baltimore: Williams & Wilkins, 1996:463–468.)

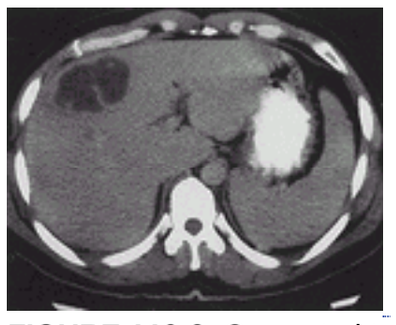

FIGURE 119-2. Computed tomography scan showing a low-density lesion in the anterosuperior segment of the right hepatic lobe of a 34-year-old man. *Klebsiella pneumoniae* was cultured from the blood and from the abscess cavity. The patient recovered with antibiotic treatment and a single abscess aspiration. (From Reynolds TB. Liver abscess. In: Kaplowitz N, ed. Liver and biliary diseases. 2nd ed. Baltimore: Williams & Wilkins, 1996:463–468.)

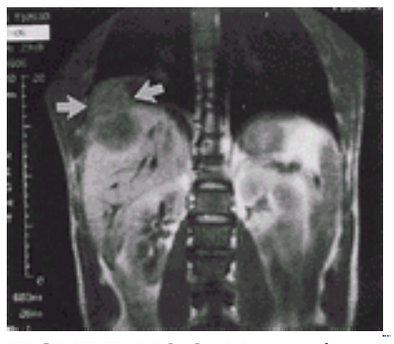

FIGURE 119-3. Magnetic resonance image showing a 6-cm lesion in the dome of the right hepatic lobe with apparent penetration into the thoracic cavity (*arrows*). The patient was a 35-year-old Hispanic man with 3 weeks of pleuritic, right-sided chest pain and fever. Red-brown material was aspirated from the right chest. Amebic serology was strongly positive. He recovered with metronidazole therapy. (From Reynolds TB. Liver abscess. In: Kaplowitz N, ed. Liver and biliary diseases. 2nd ed. Baltimore: Williams & Wilkins, 1996:463–468.)

Laboratory Testing

Although commonly employed blood tests are of limited utility in reaching a specific diagnosis of hepatic abscess, certain abnormalities are typically present. Leukocytosis (more than 10,000/mm ^{3}) is present in more than 90% of patients. Mild anemia is common, with hemoglobin levels of less than 12 g/dL seen in two thirds of patients. Mild to moderate elevation of serum alkaline phosphatase activity is typical but of little discriminating value in the absence of information from imaging studies. [2], [4], [22]

Detection of antiamebic antibodies is of primary importance in diagnosis of amebic liver abscess. Although serologic tests may rarely be negative very early in infection, more than 90% of patients with hepatic abscess develop antibodies to *Entamoeba histolytica* in high titer. A positive serologic response to *E histolytica* indicates tissue invasion by the parasite and not simply intestinal colonization. Both indirect hemagglutination and enzyme-linked immunosorbent assays are available. [23] Importantly, seropositivity does not distinguish current from prior disease. Thus, persistence of antiamebic antibodies after resolution of amebic abscess (usually in diminishing titer) may lead to confusion in the differential diagnosis of a subsequent liver lesion, especially in areas in which infection is endemic. [1], [24], [25] and [26]

A key element in diagnosis and treatment of pyogenic liver abscess is identification of the organisms in the abscess. Blood cultures are positive for bacteria in about 50% of patients. Thus, at least two separate specimens of blood should be taken for culture before administration of antibiotic agents. Most pyogenic abscesses are caused by enteric gram-negative aerobic rods, streptococci, and anaerobes. Although blood cultures typically identify a single organism, cultures from abscess aspiration are frequently polymicrobial. [2], [28], [29] Needle aspiration of an abscess is the best and most direct method to distinguish amebic from pyogenic abscess. Material from an amebic abscess is brown-red in color and typically is not particularly malodorous. A pyogenic abscess yields material that is creamy, tan-green in color, and often putrid, reflecting anaerobic infection. Gram stains of amebic abscess contents show neutrophils but no bacteria, unless secondary infection is present. Smears of pyogenic abscess contents usually identify at least one bacterial form. Meticulous handling of aspirated material to avoid exposure to air enhances recovery and identification of anaerobic species. Reliable and complete identification of infectious agents ensures proper selection of an antibiotic treatment regimen. [29]

COURSE AND COMPLICATIONS

Amebic Abscess

Without treatment, amebic liver abscess tends to enlarge and ultimately extend through the diaphragm or rupture into the peritoneal cavity. Pleuropulmonary involvement is the most common complication. [10], [30], [31] and [32] Upon extension through the diaphragm, amebic empyema or lung involvement with development of hepatobronchial fistula may occur. Rupture into the right pleural cavity is less frequent than into the lung, probably because of fusion of the diaphragmatic and visceral pleura induced by inflammation. Erosion into a bronchus may be followed by expectoration of amebic pus and by amelioration of symptoms. This mechanism of abscess drainage affords an opportunity for spontaneous resolution. Less frequently seen is rupture into the pericardium, typically a complication of amebic abscess within the left hepatic lobe. This may be accompanied by retrosternal pain, a pericardial friction rub, and findings of cardiac tamponade, such as jugular vein distention with pulsus paradoxicus. Historically, rupture into the pericardium has carried a high mortality. [10], [30]

When an amebic abscess ruptures abruptly into the peritoneum, diffuse abdominal pain and signs of peritonitis occur. Less frequently encountered is a contained abscess adjacent to the site of hepatic rupture. In neither instance is surgery required because cure typically results from antimicrobial therapy alone. [10], [31] Other rare intraabdominal complications include rupture into bowel and hemobilia due to formation of an arteriobiliary fistula.

Pyogenic Abscess

The mortality rate associated with untreated pyogenic liver abscess approaches 100%. [3], [5], [7] As with amebic abscess, complications of pyogenic liver abscess include rupture and extension into surrounding tissues, with pleuropulmonary involvement most common. In contrast to amebic peritonitis, should rupture into the peritoneum occur from pyogenic liver abscess, mortality is very high without surgical management. Abscess-associated thrombosis of the portal vein or a hepatic vein can lead to residual portal hypertension or the Budd-Chiari syndrome after otherwise successful treatment of the abscess. Patients with large or multiple pyogenic abscesses are at increased risk for developing infections at remote body sites due to bacteremia. Patients with polymicrobial abscesses have a higher mortality rate than do those with monomicrobial infections. In one series, all 14 patients who had two or more organisms isolated from blood cultures died. [5] Mortality appears to be higher in patients with pyogenic abscesses of biliary origin, probably because these patients tend to have multiple abscesses. Even with prompt diagnosis and treatment, the mortality rate from pyogenic liver abscess remains high, ranging from 10% to 30%. [2], [5]

TREATMENT

Liver abscess should be considered in any patient with fever, leukocytosis, pain in the right upper quadrant, and tenderness over the liver or right lower chest wall. Depending on the age of the patient and the nature of the discomfort and associated symptoms elicited by history, either US or CT should be obtained. Either modality will detect reliably one or more hepatic lesions, with imaging characteristics suggesting abscess. Should jaundice or evidence for biliary obstruction be discovered, consideration of cholangiography and biliary decompression is appropriate. Regardless of whether amebic or pyogenic abscess is believed more likely, at least two sets of blood cultures should be obtained before administration of antibiotic agents.

Amebic Abscess

If the patient is a young man who has emigrated from or traveled to an area where amebiasis is endemic, who has a single lesion on an imaging study, and who does not appear toxemic, then amebic abscess is the more likely diagnosis. Stool specimens should be examined for amebic cysts and trophozoites and serum tested for *E histolytica* antibodies. Treatment in adult patients should then be initiated with metronidazole, 750 mg given orally three times daily for 10 days. [1], [29] An alternative regimen is chloroquine phosphate, 1000 mg (600 mg chloroquine base) given orally daily for 2 days, and then 500 mg (300 mg chloroquine base) given orally daily for 2 to 3 weeks. [1], [33] Clinical improvement is expected within three days. After treatment with either of these tissue amebicides, therapy with a lumenal amebicide is appropriate to eradicate amebas within the bowel because these may cause relapsing or persisting infection. Iodoquinol, 650 mg given orally three times daily for 20 days, is the drug of choice for this purpose. [33] Aspiration of the abscess is indicated if there is no clinical improvement within several days, if the diagnosis is in doubt, or if the abscess is large or located such that rupture is believed likely. Amebic pleural effusions are treated by aspiration and one of the amebicidal regimens. Amebic pericarditis is treated by pericardiocentesis and by aspiration of the (usually left lobe) hepatic abscess in addition to amebicidal antibiotics. Surgery is not indicated for uncomplicated amebic liver abscess but may be appropriate in some instances for rupture or, rarely, for failure of medical therapy alone. [34] US-guided percutaneous drainage is effective and should obviate the need for surgical management in most cases. [35] In uncomplicated amebic liver abscess, the prognosis is good, with a mortality rate of about 1%. [10] Extension into the chest or pericardium increases mortality significantly, to 6% and 25% to 30%, respectively. [10], [31]

Pyogenic Abscess

If the patient is older, lacks a history suggesting risk for amebiasis, or has multiple lesions or bile duct obstruction on an imaging study, pyogenic abscess is more likely. After blood cultures have been obtained, treatment with a broad-spectrum antibiotic regimen is appropriate. Because of the mixed nature of many pyogenic infections, the initial empiric regimen should provide effective coverage against aerobic enteric bacilli, microaerophilic streptococci, and enteric anaerobes. Suitable regimens include ampicillin-sulbactam, piperacillin, cefoxitin, imipenem, or a third-generation cephalosporin plus metronidazole. [29] Previous instrumentation or obstruction of a patient's biliary tree and local patterns of bacterial antibiotic resistance should prompt broader coverage or antibiotic selection to strengthen coverage against specific pathogens such as *Enterococcus* and *Pseudomonas* species. [4] In most instances, diagnostic aspiration for bacterial culture is appropriate. Ideally, this should occur before antibiotics are administered, but treatment should not be delayed injudiciously if prompt aspiration is not feasible. Many choose to leave a drain in place after aspiration. Several series have indicated that aspiration (with or without catheter drainage) and antibiotics are curative in most instances. [2], [4], [6], [13], [36], [37], [38], [39], [40], [41] and [42] Antibiotic therapy alone should be employed only for those with multiple small abscesses not amenable to aspiration and for those with a contraindication to aspiration and drainage (e.g., coagulopathy). [43] Surgical drainage, either open or laparoscopic, [44] is appropriate for patients who fail to improve with percutaneous drainage and antibiotic therapy and for those who have an extrahepatic or biliary source of hepatic abscess in need of surgical treatment. Surgical intervention may ultimately be necessary in up to one third of patients with pyogenic abscess. [45], [46] Although percutaneous drainage and current antibiotic regimens have reduced mortality from pyogenic liver abscess significantly, it remains high (20% to 40%) in patients with significant comorbidity, multiple abscesses, malignant biliary obstruction, and inadequate drainage. [4], [5]

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I am grateful for the use of Figure 119-1, Figure 119-2, and Figure 119-3. *These appeared previously in Reynolds TB. Liver abscess. In: Kaplowitz N, ed. Liver and biliary diseases. 2nd ed. Baltimore: Williams & Wilkins, 1996:463–468.*

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CHAPTER 120

Laurie D. DeLeve

VASCULAR DISEASES OF THE LIVER

SYSTEMIC CIRCULATORY DISEASE

Ischemic Hepatitis

Ischemic hepatitis may occur as a result of reduction in systemic blood pressure with or without hypoxemia. Ischemic hepatitis often occurs in patients with underlying heart disease, [1], [2] and [3] and it has been suggested that right-sided heart failure with hepatic venous congestion predisposes to ischemic hepatitis due to systemic hypotension. [3] Decreased cardiac output may be due to cardiac causes such as myocardial infarction, severe congestive heart failure, cardiac arrhythmias, severe cardiomyopathy, cardiogenic shock, constrictive pericarditis, and pericardial tamponade. Systemic causes of hypotension include septic shock or hypovolemia due to massive hemorrhage, extensive burns, or extravasation of fluid into the peritoneum, such as in acute pancreatitis. Primary pulmonary causes of hypoxemia may include massive pulmonary embolus, end-stage chronic obstructive lung disease, or restrictive lung disease.

The diagnosis is made on clinical grounds when there is systemic hypotension and severe and rapidly reversible elevation of serum aminotransferase in the absence of other causes of acute hepatitis. In addition to these features, a marked, rapid elevation of lactate dehydrogenase (LDH) [4] with a low ratio of serum aminotransferase to LDH is helpful in distinguishing ischemic hepatitis from acute viral hepatitis. [5] Serum aminotransferases are most commonly 20 or more times the upper limit of normal, and an elevation less than 20 suggests the need for further confirmation of the diagnosis. [6] Concomitant renal hypoperfusion with elevation in creatinine and blood urea nitrogen are also usually present. The classic histological feature is centrilobular necrosis.

Congestive Hepatopathy

Causes of congestive hepatopathy include chronic right-sided heart failure due to cardiomyopathy or valvular heart disease, constrictive pericarditis, and pulmonary hypertension. Clinical findings include a dull ache in the right upper quadrant, hepatomegaly, increased jugular venous pressure, and hepatojugular reflux. Liver tests are usually normal. Congestion causes sinusoidal dilation in the centrilobular sinusoids with preservation of normal architecture in the periportal areas. Histologically, this pattern is referred to as a “nutmeg” appearance. Centrilobular fibrosis may occur with long-standing disease, but frank cardiac cirrhosis is rare.

Treatment of both ischemic hepatitis and congestive hepatopathy is directed toward the underlying cardiopulmonary disease.

BUDD-CHIARI SYNDROME

Classic Budd-Chiari syndrome is defined as obstruction or occlusion of the hepatic veins due to thrombus, fibrous obliteration, or tumor invasion. In the West, classic Budd-Chiari is more common, whereas in South Africa, India, Japan, Nepal, and China, membranous obstruction of the hepatic portion of the inferior vena cava (IVC) is more frequent. The symptoms of these two forms of hepatic vein obstruction are the same, but etiology and management differ. [7], [8] For the sake of clarity, the former will be referred to as *Budd-Chiari syndrome,* and the latter will be referred to as *membranous obstruction of the IVC*.

Clinical Features

Budd-Chiari syndrome may present with hepatomegaly, abdominal pain and tenderness, ascites, mild jaundice, and eventually, liver failure. The presentation may be acute, subacute, or chronic, although the latter two are more common. Severity of symptoms depends on the extent of the involvement of the hepatic veins, the time course over which the obstruction develops, and the duration of untreated disease. Slower development of the obstruction or occlusion allows the formation of collaterals, which alleviates sinusoidal congestion. There is a wide spectrum in presentation of the disease. Patients with an acute presentation are more likely to present with fulminant hepatic failure with coagulopathy, encephalopathy, and hepatorenal syndrome. Patients with a chronic presentation may present with chronic liver disease with (near-) normal aminotransferases, low albumin, and a prolonged prothrombin time. If there is extensive formation of collaterals or involvement of a single hepatic vein, the patient may have preservation of normal liver function and no ascites. Budd-Chiari syndrome may be asymptomatic in up to 25% of cases. The most common presentation is a history of vague complaints for less than 6 months and onset of ascites. The serum ascites–albumin gradient is greater than 1.1 g/dL.

The caudate lobe drains through the right inferior hepatic vein into the IVC, which thromboses less frequently, so that flow through the caudate lobe may become an important route of blood flow when there is occlusion of two or all three main hepatic veins. This increased flow leads to caudate lobe hypertrophy in about half of the patients with Budd-Chiari syndrome. Significant caudate lobe hypertrophy may compress the IVC behind it, complicating decompression by portacaval shunts (which are below the level of compression of the IVC).

Risk Factors

Most cases of Budd-Chiari syndrome can be categorized into two groups: hepatic vein thrombosis and mechanical outflow obstruction. Risk factors can therefore be divided into these two categories. The major risk factors for hepatic vein thrombosis (Table 120-1) are chronic myeloproliferative diseases and prothrombotic coagulation disorders. About 25% to 30% of patients have multiple concurrent risk factors. [9], [10] Myeloproliferative disorders are the leading causal factor in 50% of

patients. [9], [11], [12] and [13] The chronic myeloproliferative disorder most closely associated with hepatic vein thrombosis is polycythemia vera, whereas essential thrombocythemia is a less common cause. In a large series, 10% of patients with polycythemia vera and 13% of patients with essential thrombocythemia developed hepatic vein thrombosis. [14] None of the 232 patients with chronic myelogenous leukemia in this series developed thrombosis of the major abdominal vessels. The increased risk for hepatic vein thrombosis is linked to both overt and latent myeloproliferative disease.

RISK FACTORS FOR HEPATIC VEIN THROMBOSIS	MECHANICAL OUTFLOW OBSTRUCTION
Myeloproliferative diseases	Hepatocellular carcinoma*
Polycythemia vera	Renal cell carcinoma*
Essential thrombocythemia	Adrenal carcinoma*
Prothrombotic coagulation disorders	Leiomyosarcoma*
	Right atrial myxoma*
Factor V Leiden mutation	Hydatid cysts*
Protein C deficiency	Amebic or pyogenic abscess*
Antiphospholipid antibodies (prothrombin G20210A mutation)	Sarcoidosis
	Trauma*
	Aspergillosis
Oral contraceptives, pregnancy, puerperium	Abdominal surgery
Hepatocellular carcinoma	
Behçet disease*	
Paroxysmal nocturnal hemoglobinuria	

*More commonly associated with membranous obstruction of the hepatic portion of the inferior vena cava.

TABLE 120-1 Risk Factors for Budd-Chiari Syndrome

In a recent series, one or more prothrombotic risk factors were identified in 87% of patients with hepatic vein thrombosis. [9] Two recent case series have identified factor V Leiden as an important risk factor, [9], [10] but these studies differed as to whether protein C deficiency was a risk factor. The prevalence of antithrombin III deficiency, protein S deficiency, the G20210A prothrombin gene mutation, and the methylene-tetrahydrofolate reductase (*MTHFR*) gene mutation in patients with hepatic vein thrombosis is similar to that expected for a healthy control population. Once Budd-Chiari syndrome causes chronic liver disease, diagnosis of protein C deficiency, protein S deficiency, or antithrombin III deficiency may be misleading because synthesis of these coagulation factors decreases as liver function declines. Antiphospholipid antibodies are a risk factor for hepatic vein thrombosis and may be primary or related to one of the various connective tissue diseases. Oral contraceptives, pregnancy, and the puerperium are associated with an increased risk for hepatic vein thrombosis, but there most commonly is a concurrent risk factor. Hepatocellular carcinomas that invade the hepatic veins may cause hepatic vein thrombosis or may cause a mechanical obstruction, usually at the level of the IVC. Uncommon risk factors for hepatic vein thrombosis include paroxysmal nocturnal hemoglobinuria, [15] postoperative complications, and Behçet's disease. Although Behçet disease may cause hepatic vein thrombosis, it is more likely to cause obstruction of the IVC. [16], [17]

Procoagulants and anticoagulants circulate systemically throughout the vasculature. If the hemostatic balance of coagulation factors were the only determinant of thrombosis, then any change in the balance would have an impact on all vascular beds. As noted in the preceding paragraph, some of the prothrombotic disorders do and others do not cause hepatic vein thrombosis. Predisposition to thrombosis in a particular vascular bed is a characteristic feature of prothrombotic disorders. This selectivity of thrombosis for discrete segments of the vasculature may be due to endothelial cell–dependent procoagulant and anticoagulant activities. [18]

The classic definition of Budd-Chiari syndrome includes both hepatic vein thrombosis and obstruction or occlusion of the hepatic veins that is nonthrombotic. The major causes of mechanical outflow obstruction are cancers and infections (see Table 120-1). Some of the causes listed in Table 120-1 may cause mechanical obstruction at the level of the hepatic veins, but they are more commonly associated with obstruction of the hepatic portion of the IVC. Uncommon causes of mechanical obstruction of hepatic veins are trauma with formation of an intrahepatic hematoma, granulomata due to sarcoidosis, and aspergillosis invading hepatic veins. [19]

Diagnosis

Imaging in conjunction with liver biopsy establishes the diagnosis of Budd-Chiari syndrome but also allows the hepatologist to plan management based on the anatomy and the severity of liver disease. Initial evaluation includes noninvasive imaging with ultrasound, dual-phase computed tomography (CT), or magnetic resonance angiography. These studies determine whether there is obstruction or absence of flow at the level of the major hepatic veins or IVC; intrahepatic, capsular, or systemic collateral formation; hepatic parenchyma with variegated appearance and nodularity; caudate lobe enlargement; ascites; or splenomegaly. If normal hepatic veins with phasic flow are detected, the diagnosis of Budd-Chiari syndrome is excluded. The definitive diagnosis requires angiography with visualization of the hepatic veins and IVC and pressure measurements. Alternatively, percutaneous transhepatic venography can be used to measure hepatic vein pressure. In addition to the diagnosis of Budd-Chiari, the diagnostic workup needs to uncover the risk factors. Because there are frequently two or more causes, one of which may be an occult myeloproliferative disorder, [9], [10] bone marrow biopsy is mandatory.

A liver biopsy is needed to establish whether there is necrosis or fibrosis, features that are deciding factors in management. On histopathology, the thrombi in the hepatic veins have multiple layers, consistent with ongoing or recurrent thrombosis. Portal vein thrombosis occurs secondarily in some of the small branches of the portal vein and in the main portal vein in about 25% of cases. In acute Budd-Chiari syndrome, there is centrilobular and sometimes midlobular sinusoidal congestion, acute hemorrhage, and liver cell ischemia. Chronic outflow obstruction leads to bridging fibrosis between central veins with sparing of the portal tracts. Commonly, there is marked heterogeneity of the lobular changes. Areas of the liver subject to the most severe outflow obstruction demonstrate sinusoidal fibrosis. Benign regenerative nodules are common.

Management

A decision tree for management is shown in Fig. 120-1. Medical therapy may not alter progression of liver disease or improve survival, but diuretics and paracentesis can be used to alleviate symptoms. In selected cases of recent thrombosis, thrombolytic agents such as tissue plasminogen activator and streptokinase have been used successfully, but the literature on this is limited to case reports. Increased sinusoidal pressure and decreased liver perfusion in Budd-Chiari syndrome lead to parenchymal necrosis, fibrosis, cirrhosis, and portal hypertension. Patients must be monitored by serial liver biopsies to detect persistent necrosis and early fibrosis. Biopsy from a single liver lobe may be misleading because not all three major hepatic veins may be equally affected, so that a bilobar liver biopsy is more reliable. Sinusoidal decompression performed before significant fibrosis and loss of liver function may stabilize disease progression. Other indications for decompression include refractory ascites and uncontrollable gastrointestinal hemorrhage. Options for decompression of obstruction in the hepatic vein include transjugular intrahepatic portosystemic shunt (TIPS) and surgical shunt. TIPS may also benefit individuals with fulminant hepatic failure. [20], [21] and [22] Given the high rate of reocclusion, TIPS is perhaps best suited as a bridge therapy for liver transplantation in Budd-Chiari syndrome. The goal of a surgical shunt is to create hepatofugal flow through the portal vein. The choice of surgical shunt often depends on the preference of the surgeon. If there is significant compression of the IVC by caudate lobe hypertrophy or substantial obstruction of the IVC by thrombosis or a web, pressure in the retrohepatic IVC may be higher than in the portal vein. Under these conditions, shunts proximal to the obstructed IVC (portacaval or mesocaval shunt) may fail, and a mesoatrial shunt should be considered. Side-to-side portacaval shunts, with direct anastomosis of the side of the portal vein to the side of the vena cava, have the highest patency rate but may make future liver transplantation more difficult: a portacaval shunt will require extensive dissection around the porta hepatis, and reversal of the shunt is a major undertaking. Mesocaval shunts are technically less demanding and may facilitate surgery at the time of liver transplantation, but when performed with a synthetic graft, they have a higher thrombosis rate than a side-to-side portacaval shunt.

FIGURE 120-1. Management of Budd-Chiari syndrome.

Liver transplantation is the treatment of choice for acute Budd-Chiari syndrome with fulminant hepatic failure and for end-stage liver disease after a chronic course. In more recent series of patients who underwent liver transplantation for Budd-Chiari syndrome, survival rates have been much improved over earlier series. Three-year survivals of 78% to 88% and 5-year survival rates of 75% have been reported. [23], [24] The improved survival is likely due to earlier treatment after onset of symptoms and the recognition of the need for lifelong anticoagulation in most patients. Liver transplantation may cure one of the underlying predispositions, such as factor V Leiden, protein C deficiency, or antithrombin III deficiency, but a thorough diagnostic workup may reveal the presence of two or more remaining prothrombotic risk factors that still require anticoagulation.

In addition to management of the liver disease, the predisposing factor needs to be addressed. In some cases, risk factors may be avoided, such as oral contraceptives or pregnancy. Phlebotomy alone increases the risk for thrombosis in patients with polycythemia vera, whereas one nonrandomized trial has suggested a reduced risk for thrombosis in patients receiving phlebotomy plus hydroxyurea. [25], [26] Hydroxyurea reduces the risk for thrombosis in patients with essential thrombocythemia. [27]

OBSTRUCTION OF THE INFERIOR VENA CAVA

In non-Western countries, IVC obstruction is more common than hepatic vein obstruction. Membranous obstruction of the hepatic portion of the IVC is one of the entities frequently seen in countries such as South Africa, India, Japan, Nepal, and China, whereas in the West, it is uncommon. The original theory was that the membrane was congenital, but current thinking is that it is a consequence of thrombus organization. [28], [29] One of the initiating factors for the initiating thrombophlebitis or thrombosis may be an infectious process. Behçet disease is more likely to cause obstruction in the hepatic portion of the IVC than in the hepatic veins. [16], [17] Patients with prothrombotic disorders also have a predisposition to obstruction of the IVC across the hepatic portion, but it is unknown why that particular section of the IVC is more vulnerable to thrombosis.

A characteristic difference between hepatic vein obstruction and IVC obstruction is the development of collateral venous circulation across the abdomen, chest, and back in the latter disease. IVC obstruction predisposes to hepatocellular carcinoma, even in the absence of cirrhosis. The reported incidence of hepatocellular carcinoma in these patients varies widely in the literature. [8] In contrast, hepatocellular carcinoma does not seem to be a consequence of hepatic vein thrombosis, but tumor involvement of the hepatic veins may lead to Budd-Chiari syndrome.

Management of IVC obstruction differs somewhat from that for Budd-Chiari syndrome. If there is a thin occluding membrane, this may be perforated by angioplasty or surgically. Portacaval shunt is not indicated, but decompression may be achieved by TIPS or by cavoatrial or mesoatrial shunt.

PORTAL VEIN THROMBOSIS

Clinical Presentation

Most patients have splenomegaly and some have a tender enlarged liver. Unless the patient has underlying cirrhosis, ascites resolves over time and is therefore an uncommon finding.

Most commonly, the course of portal vein thrombosis is chronic. Initially, patients develop extensive collaterals throughout the upper gastrointestinal tract, portal hypertensive gastropathy, splenomegaly, and ascites. Blood flow to the liver from the hepatic artery and from perihepatic varices is usually adequate to maintain normal liver function. Recanalization of the portal vein clot leads to formation of small tortuous veins, so-called cavernomatous transformation. The most common presentation in patients with chronic portal vein thrombosis is gastrointestinal hemorrhage due to varices, although the presenting complaint in cirrhotic patients may be worsening ascites. [30] An alternate presentation to the more chronic course described earlier is an acute course with abdominal pain. This may signify propagation of the clot into the mesenteric circulation with bowel ischemia or infarction.

Risk Factors

Hepatoma, pancreatic cancer, and cirrhosis are common risk factors for portal vein thrombosis. When patients with cancer or cirrhosis are excluded, one or several factors associated with increased risk for thrombosis may be identified in 70% to 80% of patients. [9], [10] Primary myeloproliferative diseases, notably polycythemia vera and essential thrombocythemia, are a common risk factor. As noted earlier for hepatic vein thrombosis, many of the patients have latent myeloproliferative disease that is not detected in peripheral blood smears. Two inherited disorders that have been identified relatively recently, the prothrombin gene mutation (factor II G20210) and factor V Leiden, appear to be risk factors for portal vein thrombosis, although there is some discrepancy in the literature on this. [9], [10] Inherited protein C, protein S, and antithrombin III deficiency need to be distinguished from acquired deficiency due to impaired hepatic synthesis in liver disease, but it is again unclear from the literature whether the inherited forms occur more commonly in this population than in controls. [9], [10] Local precipitating factors play a common role in portal vein thrombosis. [10] Intra-abdominal infection with pylephlebitis, malignancy, abdominal trauma, or surgery, either alone or in concert with other risk factors, may precipitate portal vein thrombosis. Oral contraceptive use, Behçet disease, [31] and antiphospholipid antibodies are other risk factors for portal vein thrombosis. Portal vein thrombosis is a rare complication of liver transplantation. Septic portal vein thrombosis can occur in appendicitis, diverticulitis, or *Bacteroides* bacteremia.

Analogous to hepatic vein thrombosis, most patients have two or more risk factors for portal vein thrombosis. Portal vein thrombosis in patients with malignancy may be due to local tumor invasion or to a hypercoagulable state. Pancreatitis may cause portal vein thrombosis either through extrinsic compression of the portal vein by a pseudocyst or through pylephlebitis secondary to acute pancreatitis. Cirrhosis may predispose to portal vein thrombosis because of sluggish blood flow through the liver in intrahepatic portal hypertension.

Diagnosis

Doppler ultrasound can detect size and patency of the portal vein and cavernous transformation of the portal vein. The latter diagnosis is made when a cluster of small irregular collateral vessels are present instead of a normal portal vein. Contrast-enhanced CT can determine patency of the portal vein. If orthotopic liver transplantation is planned for patients with portal vein thrombosis, it is crucial to determine the extent of thrombosis. This can be determined more accurately with double-helical CT than with standard CT.

After the diagnosis is established, it is crucial to determine the underlying risk factors for thrombosis, which often requires a hematology consult. Among the risk factors listed earlier, the ones most easily missed are the primary myeloproliferative disorders, polycythemia vera and essential thrombocythemia. These disorders are frequently occult (i.e., not manifested in peripheral blood) at the time the patient presents with portal vein thrombosis. Diagnosis of occult myeloproliferative disorders therefore requires a bone marrow biopsy.

Management

The most common presenting symptom of portal vein thrombosis is variceal hemorrhage. Acute variceal bleeding may be managed endoscopically. Prevention of recurrent variceal bleeding can be addressed with endoscopic band ligation and beta-blockers. The two options for intractable variceal hemorrhage are shunt surgery and TIPS. Shunt surgery in this population carries an increased risk for thrombosis of the shunt and should be performed with larger veins. [32] TIPS is technically difficult in this setting and should be considered for intractable bleeding. [22] Portal vein thrombosis is an infrequent complication after liver transplantation. Two case

reports have described the use of a combination of local thrombolytic therapy, angioplasty, and stenting in this setting. [33], [34]

Liver transplantation in patients with portal vein thrombosis requires a special approach to the vascular reconstruction. Options include combined liver and small bowel transplantation, interposition of a venous graft between the donor portal vein and the recipient portal vein proximal to the thrombus, cavoportal hemitransposition (portal perfusion with inflow from the IVC), or arterialization of the portal vein. [35], [36] and [37]

Given the risk for variceal hemorrhage on one hand and extension of thrombosis on the other, the question arises as to the risk-benefit ratio of anticoagulation. In a recent retrospective study in a cohort of patients without underlying malignancy or cirrhosis, the benefit of anticoagulation was found to outweigh the risk. [38]

SINUSOIDAL OBSTRUCTION SYNDROME (HEPATIC VENOOCCLUSIVE DISEASE)

In 1920 and again in 1945, papers were published describing a novel liver disease. [39], [40] The most striking histological feature was occlusion of the central veins, and it was subsequently named *hepatic venoocclusive disease*. [41] Clinical studies have demonstrated that involvement of the central veins is not essential to the disease, [42], [43] and experimental studies, discussed later in this chapter, have shown that the disease process originates in the sinusoids. [44], [45] Based on these findings, investigators in this field have proposed renaming this *sinusoidal obstruction syndrome* (SOS). [46]

Pathogenesis

In contrast to most liver diseases, SOS presents with features of portal hypertension, and evidence of parenchymal dysfunction follows later. This is indicative of the primary circulatory nature of the disease. Experimental studies indicate that the drugs that cause SOS target the sinusoidal endothelial cell. [44], [47], [48] and [49] Sinusoidal endothelial cells round up, and gaps form within and between these cells. [44] This permits blood to enter into the space of Disse and dissect off the sinusoidal lining, which then embolizes downstream and blocks sinusoidal flow. [45]

Clinical Presentation

The originally described cases of SOS were due to ingestion of herbal teas (“bush tea disease”) or foodstuffs contaminated with pyrrolizidine alkaloids, particularly in protein-malnourished individuals. In many parts of the world, ingestion of pyrrolizidine alkaloids is still the major cause of SOS. In North America and Western Europe, the most frequent cause of SOS is treatment with a conditioning regimen for hematopoietic stem cell transplantation (bone marrow transplantation), either high-dose combination chemotherapy or chemotherapy plus irradiation. [43], [50], [51] and [52] SOS may occur after long-term immunosuppression with azathioprine therapy, such as in kidney or liver transplantation patients. [53], [54], [55], [56] and [57] SOS is also seen after treatment with chemotherapeutic agents unrelated to stem cell transplantation and has been associated with drugs such as actinomycin D, dacarbazine, cytosine arabinoside, 6-thioguanine, and urethane. [58] More recently, SOS has been reported in patients treated with a novel drug for acute myeloid leukemia called gemtuzamab ozogamicin (Mylotarg). [59], [60], [61] and [62].

The classic features of SOS are hyperbilirubinemia, right upper quadrant pain of liver origin or hepatomegaly, and weight gain. The course of disease is protracted in SOS owing to chronic exposure to pyrrolizidine alkaloids, whereas SOS in the setting of stem cell transplantation evolves more rapidly. In patients treated with cyclophosphamide-containing conditioning regimens, onset is within 10 to 20 days of initiation of treatment. [51] In other conditioning regimens, onset of SOS may occur later than for cyclophosphamide-containing regimens. [63]

Diagnosis

In the setting of stem cell transplantation, a tentative diagnosis can be made based on the presenting features, but other conditions need to be ruled out. Clinical criteria for SOS after stem cell transplantation have been published by two groups of investigators (Table 120-2). The differential diagnosis in this population includes acute or hyperacute graft-versus-host disease, sepsis, medication-induced cholestasis, congestive heart failure, and tumor infiltration. Imaging studies can confirm hepatomegaly and ascites and can exclude biliary obstruction and tumor invasion into the hepatic parenchyma or vasculature, but there are no definitive diagnostic features by imaging. The main differential diagnosis that might warrant a liver biopsy is acute or hyperacute graft-versus-host disease.

SEATTLE CRITERIA[65]	BALTIMORE CRITERIA[52]
Diagnosis requires two of three criteria within 20 days of transplantation: Bilirubin >2 mg/dL Hepatomegaly or pain of liver origin >2% weight gain due to fluid retention	Hyperbilirubinemia (>2 mg/dL) plus at least two of the following three findings: (Tender) hepatomegaly >5% weight gain Ascites

TABLE 120-2 Criteria for Diagnosis of Sinusoidal Obstruction Syndrome

Prevention and Treatment

Prevention is largely limited to identification of those at high risk for SOS, who may then be limited to less hepatotoxic alternatives. The major risk factors are hepatitis C, hepatic fibrosis or cirrhosis, previous exposure to a myeloablative regimen, and previous history of SOS. Strategies to reduce the risk for SOS might be use of a nonhepatotoxic, nonmyeloablative regimen, [64] lower dose of total-body irradiation, [65] reversing the order of drugs in the busulfan-cyclophosphamide regimen (i.e., cyclophosphamide first), [66] or avoiding cyclophosphamide-containing regimens altogether.

Adjustment of busulfan dosing based on plasma concentrations has been shown in some studies to be beneficial, but this could not be confirmed in a number of studies (see review [67]). The value of therapeutic monitoring of busulfan may depend on other factors, such as the age of the patient, the underlying disease, and the other drugs in the conditioning regimen. [67]

Prophylactic treatment has been tried with heparin, low-molecular-weight heparin, prostaglandin E, pentoxifylline, and ursodeoxycholic acid, but none of these compounds has been shown to reduce the incidence of fatal SOS (see review [46]).

Most patients with SOS recover spontaneously. For those who require therapy, the approach is largely supportive. Ascites is treated with sodium restriction, diuretics, and therapeutic paracentesis when indicated for discomfort or respiratory impairment. In patients with severe SOS with multiorgan failure, hemodialysis and mechanical ventilation may be used but are unlikely to affect the outcome. [51], [68], [69] and [70]

For patients with a poor prognosis (see section “ Prognosis”), tissue plasminogen activator (t-PA) plus heparin should be considered. Just less than 30% of patients with severe SOS treated with t-PA plus heparin may show improvement. [71], [72] and [73] t-PA plus heparin should not be used in patients with increased risk for intracerebral or pulmonary hemorrhage, and patients who already have renal or pulmonary failure may not benefit. [72]

Defibrotide is an experimental compound that has been tried for a variety of vascular diseases. It is a single-stranded polydeoxyribonucleotide with antithrombotic, antiischemic, and thrombolytic effects and reduces leukocyte accumulation. Uncontrolled clinical trials in moderate to severe SOS have been promising.

TIPS decompresses portal pressure and relieves ascites but does not improve outcome of SOS. [35], [74], [75] Liver transplantation may be considered in patients who develop SOS after stem cell transplantation for a benign disease or in those whose underlying malignancy is expected to have a good outcome (e.g., chronic myelogenous leukemia in chronic phase). However, liver transplantation is not indicated when the risk for recurrent malignancy is high.

Prognosis

Definitions of SOS and of fatal SOS vary among transplantation centers. These differences may account for the wide range in published case-fatality rates, which range from 0% to 67%. [51], [52], [76], [77] and [78] Two large studies with cyclophosphamide-containing regimens reported a 70% recovery rate from SOS, [51], [76] and a study with SOS due to other alkylating agents reported an 84% recovery rate. [63] For patients who develop SOS due to cyclophosphamide-containing regimens, outcome can be predicted based on bilirubin level and weight gain using published graphs. [79]

NODULAR REGENERATIVE HYPERPLASIA

Nodular regenerative hyperplasia (NRH) is an uncommon disorder in which there are nodules of hyperplastic hepatocytes surrounded by areas of hepatocyte atrophy. In contrast to cirrhosis, there are no fibrous septa surrounding the nodules. Most commonly, the nodules are scattered diffusely throughout the liver. Two large autopsy studies have reported prevalence rates of 2.1% and 2.6%. [80], [81]

Etiology

The etiology is not known, but the widely accepted hypothesis is that NRH is initiated by impaired perfusion of areas of the liver. [82] Hepatocytes in areas of hypoperfusion become atrophic or apoptotic, [83] and parenchyma in areas where perfusion is maintained develops reactive hyperplasia.

Underlying predispositions to NRH include collagen vascular diseases, several hematologic malignancies, some immunologic disorders, and a variety of drugs and toxins (Table 120-3). Although the diseases and predisposing factors for NRH vary greatly, a shared element is the ability to impair the circulation at the level of the portal vein or sinusoids. Inflammation of the hepatic artery in collagen vascular diseases or immune complex diseases may lead to inflammatory destruction of adjacent portal veins. [84], [85] Prothrombotic disorders, such as agnogenic myeloid metaplasia, polycythemia vera, or antiphospholipid syndrome, may cause thrombosis in portal veins. [86] Damage to sinusoidal endothelial cells by long-term azathioprine therapy, [48], [53] such as in renal or liver transplant recipients, or after conditioning therapy for stem cell transplantation [42], [87] may lead to NRH.

Collagen vascular diseases
Rheumatoid arthritis
Scleroderma
Systemic lupus erythematosus
Polyarteritis nodosa
Glomerulonephritis
Hematologic diseases
Polycythemia vera
Essential thrombocythemia
Agnogenic myeloid metaplasia
Chronic myeloid leukemia
Hodgkin disease
Non-Hodgkin lymphoma
Multiple myeloma
Immunologic disorders
Cryoglobulinemia
Antiphospholipid syndrome
Myasthenia gravis
Drugs and toxins
Anabolic steroids
Azathioprine
Oral contraceptives
Conditioning regimen for stem cell transplantation
Thorotrast
Toxic oil syndrome
6-Thioguanine

TABLE 120-3 Conditions Associated with Nodular Regenerative Hyperplasia

Clinical Aspects

NRH is most commonly asymptomatic, and the lesion is most likely to be detected incidentally at autopsy. When symptomatic, the disease presents with evidence of portal hypertension with variceal hemorrhage, ascites, splenomegaly, or hypersplenism. Although rare, NRH may present as end-stage liver disease. [88]

Imaging studies may detect the manifestations of portal hypertension, but not the 2- to 5-mm nodules themselves. The diagnosis can be established by liver biopsy. The pathologic appearance is of scattered nodules of hyperplastic hepatocytes surrounded by atrophic parenchyma and the absence of fibrotic septa around the nodules.

In most patients, no treatment is needed for the liver disorder itself, although the underlying predisposing condition or precipitating factor may require therapy. Manifestations of portal hypertension are treated along conventional lines.

PELIOSIS HEPATIS

Peliosis hepatis is a rare abnormality characterized by blood-filled cystic lesions in the hepatic parenchyma distributed irregularly throughout the liver. The peliotic cavities range in size from less than 1 mm to several centimeters. Although most common in the liver, peliotic lesions may also be present elsewhere in the reticuloendothelial system, notably the spleen, abdominal lymph nodes, and bone marrow.

Etiology

Historically, peliosis was found at autopsy in patients with chronic wasting illnesses, in particular, tuberculosis and cancer (Table 120-4). Peliosis was also observed in patients on androgenic anabolic steroids or after long-term treatment with azathioprine. In patients with acquired immunodeficiency syndrome (AIDS), infection with *Bartonella henselae* or *Bartonella quintana* may cause peliosis as well as bacillary angiomatosis.

Acquired immunodeficiency syndrome and *Bartonella* species infection
Tuberculosis
Myeloproliferative diseases
Leukemia
Lymphoma
Multiple myeloma
Macroglobulinemia
Anabolic steroids
Oral contraceptives
Azathioprine
Arsenic
Thorotrast
Vinyl chloride
6-Thioguanine

TABLE 120-4 Conditions Associated with Peliosis Hepatis

The initial change in peliosis hepatitis appears to be sinusoidal dilation, followed by formation of cavities without sinusoidal endothelial cells. [89], [90] Eventually, endothelial lining may be restored in parts of the peliotic cavities. Although it had been suggested that the initial target in peliosis was the sinusoidal endothelial cells, this concept has been most clearly supported by studies of peliosis due to *Bartonella* species. *Bartonella* bacilli can be detected by electron microscopy in sinusoidal endothelial cells of AIDS patients, [91] and there is disruption of the sinusoidal endothelial cell lining. [89] Interestingly, *Bartonella* causes bacillary angiomatosis in the skin, an organ with a continuous endothelial lining, but causes peliosis in the reticuloendothelial system, which has a discontinuous endothelium. [90]

Peliosis due to drugs and toxins has also been attributed to damage to sinusoidal endothelial cells. A few xenobiotics have been linked to SOS, NRH, peliosis hepatis, or sinusoidal dilation, and in some cases, up to all four have been described within the same liver. Azathioprine (all four lesions), urethane (peliosis, SOS), 6-thioguanine (peliosis, NRH, SOS), Thorotrast (peliosis, NRH), oral contraceptives (sinusoidal dilation, peliosis hepatis, NRH) and anabolic steroids (peliosis, NRH)

cause an overlap of these injuries. [53], [80], [92], [93] Damage to sinusoidal endothelial cells and sometimes to hepatic venular endothelial cells seems to be the common link in these four types of liver injury. [53], [94]

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CHAPTER 121

Sareh Parangi and Richard Hodin

ABDOMINAL CAVITY: ANATOMY, STRUCTURAL ANOMALIES, AND HERNIAS

EMBRYOLOGY OF THE ABDOMINAL CAVITY

Abdominal Cavity

Understanding of the embryology of the abdominal contents is imperative to understanding the pathophysiology of the various congenital and structural anomalies of the abdominal cavity. The abdominal or peritoneal cavity is formed from a large intraembryonic coelomic cavity during the fourth week of embryonic development. The cells of the somatic mesoderm lining the intraembryonic coelomic cavity become mesothelial and form the parietal layer of the serous membranes lining the outside of the peritoneal, pleural, and pericardial cavities. In a similar manner, the cells of the splanchnic mesoderm layer form the visceral layer of the serous membranes covering the abdominal organs, lungs, and heart. [1] During the fifth to seventh weeks of development, the pleuroperitoneal folds fuse with the septum transversum, and the mesentery of the esophagus thus separates the thoracic cavity from the abdominal cavity. In the cephalic and caudal parts of the embryo, the primitive gut (an endodermal lined cavity) forms a blind-ending tube, the foregut and the hindgut, respectively. The middle part, the midgut, remains temporarily connected to the yolk sac by means of the vitelline duct or yolk stalk. The upper gastrointestinal tract develops from the foregut and includes the esophagus, the stomach, the duodenum, the liver, the gallbladder and the pancreas. The midgut develops into the small bowel, the right and transverse colons. The hindgut development forms the left colon, sigmoid, rectum, and the upper part of the anal canal.

Development of the Foregut, Midgut, and Hindgut

During embryonic development, the foregut rotates 90 degrees clockwise around its longitudinal axis, causing the left side of the stomach to face anteriorly and its right side, posteriorly (Fig. 121-1). The foregut blood supply is the celiac artery.

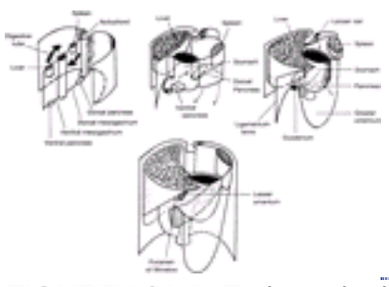

FIGURE 121-1. Embryonic development of the foregut. The organs of the upper gastrointestinal tract rotate 90 degrees clockwise (when reviewed from above) in a transverse plane. Originally, ventral structures like the liver and ventral pancreas are situated to the right, and posterior organs such as the spleen and dorsal pancreas and the dorsal aspect of the future stomach are positioned to the left side of the abdominal cavity. The dorsal mesogastrium elongates and folds to the left to form the greater omentum.

The development of the midgut is characterized by rapid elongation of the gut and its mesentery, resulting in the formation of the primary intestinal loop. Over its entire length, the midgut is supplied by the superior mesenteric artery. As a result of this rapid elongation and the simultaneous expansion of the liver, the abdominal cavity of the fetus becomes too small to contain all the intestinal loops. These loops enter the extraembryonic coelomic cavity in the umbilical cord, forming a physiologic umbilical herniation during the sixth week of development (Fig. 121-2). Coincident with this rapid growth, the midgut rotates 270 degrees counterclockwise around the axis of the superior mesenteric artery. At about the third month, the herniated intestinal loops return to the abdominal cavity. The proximal portion of the jejunum is the first part to reenter, and it comes to lie on the left side, whereas the primitive cecum is the last to enter back and descends into the right iliac fossa.

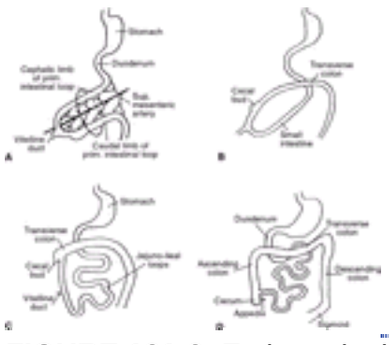

FIGURE 121-2. Embryonic development of the midgut. **A:** Schematic drawing of the primary intestinal loop before rotation (lateral view). The superior mesenteric artery forms the axis of the loop, and the *arrow* indicates the counterclockwise rotation around this axis. **B:** Similar views as in **A,** showing the primary intestinal loop after 180-degree counterclockwise rotation. The transverse colon is shown as passing in front of the duodenum. **C:** Anterior view of the intestinal loop after the 270-degree counterclockwise rotation. Note the cecal bud in the right upper quadrant **D:** Similar views as in **A** with the intestinal loops in the final position. Cecum and appendix are now in the right lower quadrant. (Adapted from ref. [1].)

The hindgut gives rise to colon and rectum past the distal third of the transverse colon as well as the upper part of the anal canal. The junction of the endodermal and ectodermal parts of the anal canal is formed by the pectinate line, which is found just below the anal columns. The endoderm of the hindgut also gives rise to the

internal lining of the bladder and urethra. The hindgut blood supply is the inferior mesenteric artery.

Mesenteries of the Abdominal Cavity

During the development of the peritoneal cavity, the splanchnic mesoderm covers the developing gut. Eventually, most of this ventral mesentery is resorbed, except for a small portion between the liver and the stomach that persists as the gastrohepatic ligament (or lesser omentum) (see Fig. 121-1), and the ligaments of the liver, that is, the coronary, falciform, and teres. The dorsal mesogastrium remains and changes in position and size; the spleen is positioned to the left, the pancreas is situated posteriorly, and the space within the mesogastrial fold becomes the lesser sac. The dorsal mesogastrium enlarges, forming the greater omentum, which drapes anteriorly. As the midgut elongates during development, the dorsal mesodermic attachment elongates as well and forms the mesentery. The right colon adheres to the posterolateral abdominal wall and becomes retroperitoneal; the rest of the midgut is intraperitoneal.

ANATOMY OF THE ABDOMINAL CAVITY

The abdominal cavity in the adult is defined by the diaphragm superiorly, the abdominal walls laterally, and the pelvis inferiorly. The peritoneal covering is the endodermal investment of the surface of the organs of the abdominal cavity. The pancreas, parts of the duodenum, right and left colon, and the rectum, being retroperitoneal, are only covered anteriorly by peritoneum. The organs are supported by thickened bands of peritoneum: the gastrohepatic, gastrosplenic, splenorenal, and splenocolic ligaments. The liver is suspended to the diaphragm also by the falciform and coronary ligaments as well as to the abdominal wall by the ligamentum teres that contains the remnant of the umbilical vein.

The abdominal cavity has three compartments: lesser sac, supramesocolic, and inframesocolic. The lesser sac is limited by the stomach and gastrocolic ligaments anteriorly; by the spleen, splenorenal, and gastrosplenic ligaments on the left side; and by the pancreas and duodenum posteriorly (see Fig. 121-1). The lesser sac communicates with the rest of the abdominal cavity through the foramen of Winslow. The boundaries of the foramen are the liver superiorly, hepatic pedicle anteriorly, duodenum and pancreas inferiorly, and the retroperitoneum posteriorly.

The supramesocolic compartment lays cephalad to the transverse mesocolon and contains the duodenum, pancreas, liver, spleen, and lesser sac. The inframesocolic compartment contains the small intestine, the colon, and the suspensory ligaments of the spleen and liver. The area between the colon and the inferior margin of the liver is called the *Morison pouch* and acts as a potential space for fluid accumulation. In addition, fluid can accumulate along the pouches created by the right and left parietocolic peritoneal coverings.

The inferior limit of the peritoneum covers the pelvic urogenital organs and the inguinal region. Most inferiorly is the pouch of Douglas, a peritoneally covered pouch between the rectum and uterus in females and the rectum and bladder in males.

The anatomy of the inguinal region (the groin) is complex and is discussed in the section “Groin Hernias.”

CONGENITAL ANOMALIES

In the embryo, the abdominal cavity is too small to accommodate the intestines, and a physiologic herniation occurs into the umbilical cord with return of the intestines to the abdominal cavity by the 10th week. An *omphalocele* (Fig. 121-3A) is failure in the growth of the coelomic walls with a large herniation of the abdominal viscera into the base of the umbilical cord. The herniated viscera are covered by a sac composed of amnion. The incidence of omphalocele is estimated to be 1 in 6000 to 10,000 live births. *Gastroschisis* (see Fig. 121-3B) is a similar anomaly, but the defect in the abdominal wall is located lateral to the umbilicus and is caused by failure of closure of the abdominal wall. No sac is present to cover the herniated intestines, and the size of the defect is often smaller. In gastroschisis, the intestines can be inflamed and thickened, and the mesentery may be foreshortened. The incidence of gastroschisis is 1 in 3200 to 10,000 births, with some male predominance; associated congenital anomalies include incomplete rotation and fixation of the midgut in 40% of cases. At least 50% of infants with omphalocele have associated anomalies of the skeleton, gastrointestinal tract, or nervous or genitourinary system. [3] Ultrasound can detect these defects prenatally, and vaginal delivery is safe for all newborns but those with the biggest omphalocele, which may contain liver. [4] Both conditions are surgical emergencies of the newborn. The airway should be managed first, and a nasogastric sump tube should be placed to prevent intestinal distention. If present, the sac is painted with antiseptic solutions, and the herniated contents are kept moist and covered with a warm moistened saline soaked gauze and plastic to prevent dehydration and desiccation. Alternatively, the entire lower torso can be covered by a plastic intestinal bag. Once stable, an operation is needed to reduce the herniated viscera and perform a fascial closure. If necessary, the viscera are wrapped in a sheet of silicone (Silastic chimney) that is sutured to the abdominal wall, allowing time for growth of the abdominal wall and eventual reduction of the herniation.

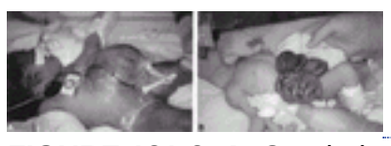

FIGURE 121-3. A: Omphalocele with large herniation of the abdominal viscera. Note the amnionic sac covering the viscera. **B:** Gastroschisis with defect lateral to the umbilicus. Note that the herniated intestines are engorged and discolored and that no sac is present.

Remnants of the omphalomesenteric (vitelline) duct may present as abnormalities related to the abdominal wall or remain asymptomatic for long periods. An *umbilical sinus* results from the continued presence of the umbilical end of the duct and can result in persistent drainage and infection at the umbilicus. Persistence of the entire duct is heralded by passage of enteric contents from the umbilicus, and excisional treatment avoids complications of intussusception or volvulus. Cystic remnants should be excised if seen at operation or if presenting as an infection at the umbilicus. *Meckel diverticulum* results when the intestinal end of the vitelline duct persists. This is a true diverticulum, composed of all layers of the intestinal wall.

CONGENITAL DIAPHRAGMATIC HERNIAS

The diaphragm appears in the third week of development as a septum transversum that separates the thorax from the abdomen. The diaphragm is derived from the following structures: the septum transversum, which forms the tendinous part of the diaphragm; the pleuroperitoneal membranes; the muscular components from the lateral and dorsal body walls; and the mesentery of the esophagus, which forms the crura of the diaphragm (Fig. 121-4A). A diaphragmatic hernia is one of the more common malformations of the newborn (1 in 2200) and is most frequently caused by failure of the pleuroperitoneal membranes to separate completely the peritoneal and pleural cavities posteriorly. [5]

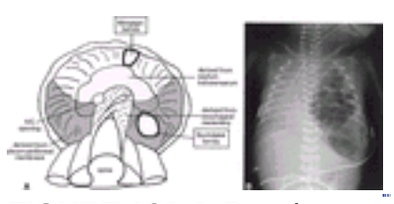

FIGURE 121-4. Development of the diaphragm and congenital diaphragmatic hernia. **A:** Abdominal surface of the diaphragm and the derivation of the components during development. The pleuroperitoneal membranes, the septum transversum, and the esophageal mesentery form the diaphragm. A Bochdalek hernia forms when there is a posterolateral defect. Morgagni hernias are less common and are present anteriorly. **B:** Chest radiograph of a child with a congenital posterolateral (Bochdalek) diaphragmatic hernia on the left. The mediastinum is displaced to the right by the intestinal loops present in the left chest.

Posterolateral hernia defects (hernia of Bochdalek) are usually large and on the left side; the intestinal loops, stomach, spleen, and part of the liver may enter the thoracic cavity (see Fig. 121-4). The presence of abdominal viscera in the chest results in compression of the heart and hypoplasia of the lung. Animal models have shown that long-term compression of the lung leads to abnormal development of the lung and the pulmonary vascular bed. [6] The neonate presents with acute respiratory distress; the heart, mediastinum, and lungs are displaced to the right, and the abdomen may be scaphoid. Respiratory sounds are absent on the affected side, and heart sounds may be audible in the right chest. The diagnosis is easily made with a chest radiograph showing one or more air-fluid levels in the left thorax, displacement of the mediastinum, and loss of the sharp diaphragmatic line separating the thorax and abdomen (see Fig. 121-4B). Radiologic studies with water-soluble contrast show the intra-abdominal contents in the left chest. Lung hypoplasia results in acute respiratory distress and is the main cause of death in these patients. Bowel obstruction, strangulation, and other gastrointestinal malformations can result in other complications. Mortality depends on the age of the patient, associated malformations, and most importantly the degree of lung hypoplasia. Mortality rates are as high as 50% in those infants requiring emergency operations during the first week of life but decrease to 10% in infants with a higher degree of lung maturity. Extracorporal membrane oxygenation is used in some instances to support the maturing lungs before and after repair of the hernia. [7]

Occasionally, a small part of the muscular fibers of the dia- phragm fails to develop, and a hernia may remain undiscovered until later in life. Such a defect is seen in the anterior portion of the diaphragm and is known as a *parasternal hernia* (hernia of Morgagni; see Fig. 121-4A). These hernias are usually small and can contain stomach, omentum, colon, or small intestine with a peritoneal covering.

UMBILICAL HERNIAS of CHILDHOOD

Herniation of intra-abdominal viscera through the umbilicus is an *umbilical hernia*. A congenital defect in the umbilical ring during development of the abdominal wall results in this hernia. Congenital umbilical hernias are more common in black and male infants, most not reaching maximal size until the infant is 1 month of age. The younger the neonate, the more common this congenital deformity, reaching 20% in premature infants. Predisposing conditions include Down syndrome, gargoylism, amaurotic familial idiocy, cretinism, Beckwith-Wiedemann syndrome, and diseases that increase intra-abdominal pressure. This type of hernia has a high propensity to reduce and heal spontaneously, so that repair is generally not indicated in children younger than 3 years of age. [8] Large or symptomatic hernias, especially with incarceration or strangulation, require immediate attention.

HERNIAS OF THE ABDOMINAL CAVITY IN ADULTS

“Hernia is a protrusion of any viscus from its proper cavity” (Sir Astley Cooper, 1804). Hernias are composed of a herniated viscus, the hernia sac, and the opening of the hernia (the hernial ring) (Fig. 121-5). The hernial sac is the internal wall of the hernia lined by peritoneum. An external hernia is comprised of a viscus abnormally located outside the abdomen; an internal hernia is when the viscus is abnormally located within the intra-abdominal space. Any viscus can become part of a hernia, but most commonly, a segment of intestine is partially or completely herniated.

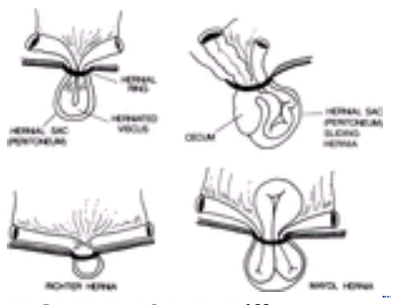

FIGURE 121-5. Different types of intestinal hernias.

A *Richter hernia* occurs when only a portion of the wall of the herniated bowel is included in the hernia (see Fig. 121-5). Such a hernia is unique in that it can strangulate without causing a complete small bowel obstruction. A herniation of two adjacent loops through the same hernial opening is called a *Maydl hernia* (see Fig. 121-5). This kind of hernia is dangerous because the closed loop may perforate, leading to peritonitis. A *sliding hernia* is when a partially retroperitoneal structure (such as colon) is part of the wall of the hernial sac (see Fig. 121-5). The symptoms of hernias depend on a number of factors, such as the exact viscus that is herniating, the location of the herniated bowel (i.e., jejunum or ileum), and whether there is a full or partial strangulation. When omentum alone is strangulated within a hernia, obstruction of the gut does not occur.

A *reducible hernia* occurs when the hernial sac contents return to the abdominal cavity spontaneously or with external manipulation. The major complications of hernias are incarceration, bowel obstruction, and bowel strangulation. A complication is more likely to occur if the hernial opening is small and in hernias in which the bowel has access to the sac. Bowel incarceration greatly increases the chance that strangulation will occur. An *incarcerated hernia* occurs when the hernial contents cannot be dislodged from the sac. Incarceration occurs when there are adhesions from the hernial contents or around the opening of the hernia, making reduction impossible, or when there is edema or contortion of the hernial contents such that hernial contents once entered into the sac cannot exit. A hernia, reducible or incarcerated, becomes a *strangulated hernia* when the blood supply to the herniated viscus is compromised.

Epidemiology

Hernias are common occurrences. Five percent of people have a spontaneous abdominal hernia during their lifetime. Many people also develop secondary hernias, most frequently at the site of a previous incision or injury to the abdominal wall. Hernia is the most common cause of intestinal obstruction worldwide, and in the United States, it is the third most common cause. In surgical practice, 10% to 15% of all procedures are related to hernias. Of all hernias, 90% are incisional, umbilical, or inguinal hernias. Groin hernias account for 85% of all hernias (Table 121-1). Indirect inguinal hernias are the most common, followed by direct inguinal hernias. Incisional hernias develop in 3% to 5% of all abdominal operations and represent 7% of abdominal wall hernias. Internal hernias are uncommon and have been seen in 0.5% of autopsies.

TYPE	OCCURRENCE
Inguinal	80%
Indirect	60%
Direct	30%
Both	10%
Femoral	5%
Inguinal and femoral	2%
With a sliding component	12%
Sigmoid	8%
Cecum	4%
Maydl hernia	2%

*Groin hernias account for 85% of all hernias.

Adapted from Fonkalsrud EW, Ament ME. Hernias of the abdominal wall. In: Fonkalsrud EW, Ament ME, eds. Hernias of the Abdominal Wall. Philadelphia: WB Saunders, 1980, and Nyhus LM, Condon RE, eds. Hernia. Philadelphia: JB Lippincott, 1978.

TABLE 121-1 Epidemiology of Groin Hernias*

There is a male-to-female prevalence in inguinal hernias of about 7:1, whereas there is female dominance in femoral, umbilical, and incisional hernias of about 2:1. Although femoral hernias are seen more commonly in females, the most common hernia in women still remains an indirect inguinal hernia.

Pathophysiology /H4>

Our understanding of the pathophysiology of inguinal hernias has changed over the years. In the past, indirect hernias were attributed at least in part to a patent processus vaginalis, which is present in many infants and up to 25% of young children. However, more recent studies have shown the inguinal ring and any patent processus vaginalis to close with a valvelike effect because of tension on the abdominal wall musculature when there is increased intra-abdominal pressure. [9] In cases of indirect inguinal hernia, this valvelike effect may becomes ineffective, allowing a viscus to enter the patent processus vaginalis. [10] Chronic injury in the form of traumatic overstretching of muscles and aponeurotic structures undoubtedly contributes to hernia formation, although rigorous scientific studies have not been performed. Clinicians often see the onset of herniation related to such activities as extreme athletics, conditions associated with increased intra-abdominal pressure such as pregnancy, chronic cough, chronic obstructive pulmonary disease, obstructive uropathy, massive ascites, or obstipation from colorectal cancer.

In cases of incisional hernias, infection or tension during the wound healing or scar remodeling phase can lead to weakened musculoaponeurotic tissue, which in turn leads to herniation at a later date. Some studies have shown altered collagen formation and breakdown in tissue samples from the inguinal floor of patients with hernias. [11]

Diagnostic Considerations

Most hernias are detected using a history and physical examination. The patient needs to be questioned regarding the presence of a bulge, conditions that make the bulge appear and disappear, and any associated pain or gastrointestinal symptoms. History of any previous hernias, abdominal wall incisions, or a family history of any connective tissue disorder such as Marfan syndrome should be sought. A review of systems to include any of the above-mentioned causes of intra-abdominal pressure should be obtained because failure to recognize these factors will increase the chance for recurrence if repair is undertaken. A complete physical examination of the abdomen and rectum should be done, including a prostate exam and testing of the stool for occult blood. All scars should be noted. The exam is best initiated with the patient in the standing position because gravity tends to pull the intra-abdominal contents into hernias. If no bulge is obvious, a Valsalva maneuver is used to increase intra-abdominal pressure. If a bulge is seen, a gentle attempt to reduce the hernia can be made with the patient standing or in the recumbent position. Examination of the area in the recumbent position often allows better palpation of the hernial defect and opening.

Radiographic studies are rarely needed but may be useful in cases in which the abdominal wall fat layer prevents accurate diagnosis. Ultrasound is rarely helpful, but in some cases, it can delineate the abdominal wall musculature and note the presence of air or peristalsing bowel. Computed tomography (CT) is good at detecting abdominal and pelvic defects; use of Valsalva may sometimes be helpful during imaging. [9], [12]

Specific Types of Hernias

Epigastric Hernias Epigastric hernias occur in the midline of the abdominal wall between the umbilicus and the xiphoid in 5% of the population (males more than females). Most epigastric hernias are asymptomatic. An area of congenital weakness in the linea alba allows protrusion of bowel, omentum, or just preperitoneal fat. Symptoms range from a small painless nodule below the xiphoid to epigastric pain with exertion or rarely small bowel obstruction. Diagnosis is easy if the hernia is large but can be difficult if there is a small hernia in an obese patient. Tenderness while palpating the linea alba with elevation of the head can sometimes be produced in small hernial protrusions. These hernias should be surgically repaired. About 20% of patients have multiple hernial defects in other locations along the linea alba, [13] and this should be taken into consideration at the time of repair.

Umbilical Hernias Umbilical hernias in adults often occur in obese people, multiparous women, and patients with ascites. The diagnosis is usually self-evident with a protuberant mass at the umbilicus. Incarceration of omentum or small bowel is common, and up to 30% of cases present with strangulation of bowel. Forty percent of patients with ascites and end-stage liver disease have umbilical hernias. In this setting, ulceration and perforation of the hernia can lead to peritonitis with a high mortality rate. [14] Discoloration or ulceration of overlying skin heralds imminent rupture. Umbilical hernias in adults generally need surgical repair. Small defects can be repaired primarily if there is no tension; defects larger than 3 cm should be repaired with prosthetic mesh or by using some other method to ensure tension-free repair (e.g., release of the lateral fascial attachments). Patients with ascites should have their ascites controlled before repair; otherwise, the repair will likely fail. In some cases of refractory ascites, portacaval shunt or transjugular intrahepatic portacaval shunt allows for a successful surgical herniorrhaphy after the ascites has disappeared. Alternatively, concomitant peritoneovenous shunt and herniorrhaphy should be considered to prevent breakdown of the repair. [15] Liver transplantation is another alternative in the appropriate candidate.

Groin Hernias Hernias of the groin are the most common of all hernias. The inguinal anatomy is defined superiorly by the arch of the aponeurosis of the transversus abdominis muscle and inferolaterally by the upper ramus of the pubis and the psoas muscle. The inguinal canal is about 4 cm in length and is located just above the inguinal ligament between the internal and external rings (Fig. 121-6). The inguinal canal contains the spermatic cord in males (vas deferens, spermatic artery and vein, and cremasteric muscle) and the round ligament in females, in addition to the ilioinguinal nerve. The superior entrance to the inguinal canal is the internal ring (the deep inguinal ring) and is an outpouching of the transversalis fascia lateral to the inferior epigastric vessels. The external ring is a slitlike opening between the diagonal fibers of the aponeurosis of the external oblique muscle (see Fig. 121-6A). The anterior wall of the inguinal canal is composed mainly by the aponeurosis of the external oblique. The superior wall is formed by the arching fibers of the internal oblique and transverse abdominal muscles, otherwise called the *conjoined tendon*. The inferior wall is formed by the inguinal ligament (the folded aponeurosis of the external oblique) that forms a shallow ledge, and more medially by the lacunar ligament along the pectineal line of the pubis. The inguinal ligament is the landmark between the abdomen and thigh. It is fixed medially to the pubis and laterally to the anterosuperior spine of the ilium and fascia of the thigh. The pectineal ligament (Cooper ligament) lies between the pubic tubercle and the femoral vein. The posterior wall (floor) is formed mainly by transversalis fascia; medially, this wall is reinforced by formation of the conjoint tendon (inguinal falx), the merging of the internal oblique and transverse abdominal aponeuroses. The floor or the posterior wall has no intrinsic strength, and as the fibers of the transversalis fascia deteriorate, a direct hernia results. [2] Hesselbach triangle is bounded by the inguinal ligament inferiorly, the inferior epigastric vessels laterally, and the lateral border of the rectus medially (see Fig. 121-6A). A weakness in the floor of this triangle (the transversalis fascia) results in a direct inguinal hernia. Hernias protruding lateral to Hesselbach triangle are termed *indirect hernias* (see Fig. 121-6B). Inguinal hernias include direct and indirect hernias found above the inguinal ligament, and femoral hernias are found below the inguinal ligament in the femoral canal. The inferior epigastric artery serves as an important defining anatomic landmark. Indirect hernias originate lateral to this artery and protrude into the inguinal canal along the spermatic cord. Direct hernias are located medial to the inferior epigastric artery and come through a weakened inguinal floor composed of the transversalis aponeurosis and fascia (see Fig. 121-6). A *pantaloon hernia* is composed of both direct and indirect components. *Sliding hernias* have one wall composed of colon or bladder. A special type of indirect inguinal hernia containing a Meckel diverticulum is *Littré hernia*.

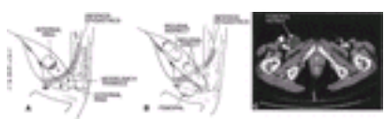

FIGURE 121-6. The right inguinal region. **A:** Schematic anatomy showing internal and external rings and Hesselbach triangle. Spaces where hernias occur include Hesselbach triangle (2), the inguinal canal (1, 2) and the femoral space (3). **B:** The most common groin hernias are direct and indirect hernias above the inguinal ligament and femoral hernias below the inguinal ligament. **C:** Computed tomography of an incarcerated femoral hernia on the right. Note a loop of small intestine is present medial to the femoral vessels; the vessels are marked by a *white arrowhead*.

The main symptom of groin hernia is a bulge in the inguinal area that appears intermittently or with increased intra-abdominal pressure. Pain is usually mild; local pain or colicky abdominal pain is suggestive of incarceration or strangulation. Palpation should be performed in the standing and recumbent position and using Valsalva maneuver to sustain increased intra-abdominal pressure. Both sides of the groin and scrotum should be examined. To perform a digital exam, the index finger is introduced into the inguinal canal through the external inguinal ring by invaginating the skin in the area (Fig. 121-7). Using the index finger, the external inguinal ring is first palpated, and with a small further push, the internal ring can be palpated. Using this simple technique, an indirect hernia can be felt exiting the internal ring at the fingertip, and a direct hernia can be felt more medial to the finger. In femoral hernias, the inguinal canal has no hernia, but a mass is felt below the inguinal ligament in the medial thigh near the saphenofemoral junction, medial to the femoral vein. Obesity makes the examination difficult, especially in detection of femoral hernias, and radiologic studies may be helpful [16] (see Fig. 121-6C).

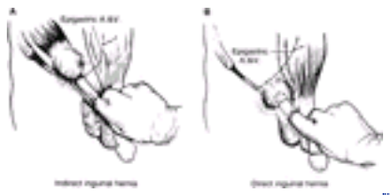

FIGURE 121-7. Digital examination of inguinal hernias. **A:** Indirect inguinal hernias are palpated in the inguinal canal at the fingertip. **B:** Direct inguinal hernias are palpated on the sides of the examiner's finger.

The differential diagnosis of an inguinal bulge includes lipoma, tumors, lymphadenopathy, abscess, cysts, endometrial deposits, testicular torsion in an undescended testicle, hydrocele, varicocele, pseudoaneurysm, and saphenous varix. Strangulated hernia can present with signs and symptoms of small bowel obstruction with the addition of a painful, tender, or tense swelling in the groin. Without an operation, it is difficult to determine whether bowel loops are strangulated; therefore, all painful, tense hernias should be considered strangulated and repaired emergently. Because both strangulated inguinal and femoral hernias require urgent operations, differentiating them is of intellectual but not practical interest. [17] Surgical treatment is generally recommended for most inguinal and femoral hernias, because of the chance of strangulation—5% in indirect hernias and up to 30% in femoral hernias. Elective repair can be undertaken as outpatient surgery using general anesthesia, spinal anesthesia, intravenous sedation, or local anesthesia alone. Repair of the hernia requires delineation of anatomy, identification of the hernia sac, and reduction of sac contents. Indirect hernia sacs are ligated at the level of the internal inguinal ring; direct hernia sacs are pushed below the repair site. Suitable native tissue can be used for repair. The modified Bassini repair apposes the shelving edge of the inguinal ligament to the conjoint tendon (inguinal falx), the merging of the internal oblique and transverse abdominal aponeuroses. The Shouldice repair is similar but uses an overlapping technique with two rows of sutures. The McVay repair involves opening the inguinal floor and approximates the conjoint tendon and transversalis fascia to the pectineal ligament (Cooper ligament); this repair is suitable for femoral hernias. Relaxing incisions of the anterior rectus sheath are used to reduce tension on repairs using native tissue. Polypropylene mesh repair is now widely used as standard repair for all direct and larger indirect hernias. The Lichtenstein repair involves overlay of the mesh along the inguinal floor underneath the spermatic cord; the mesh is sutured to the shelving edge of the inguinal ligament, the pubis medially, and the internal oblique fascia superiorly. A slit in the mesh allows passage of the spermatic cord. [18] Recurrence rates are thought to be somewhat lowered by the use of mesh, but a higher risk for infection must be taken into consideration. [19] The laparoscopic approach to repair of hernias has recently been popularized. A transabdominal or preperitoneal approach can be taken, and knowledge of anatomic considerations is key [20] (Fig. 121-8A). The transabdominal preperitoneal and the totally extraperitoneal repairs are the most commonly used laparoscopic approaches. [21] After reducing the hernia contents, the repair is performed by securing a sheet of polypropylene mesh over the internal aspect of the inguinal floor and internal ring area using surgical clips or staples (see Fig. 121-8B, Fig. 121-8C). Proponents of laparoscopic inguinal hernia repair cite advantages such as improved cosmesis, less postoperative discomfort, earlier return to work, and easier repair of bilateral hernias. The laparoscopic approach also has disadvantages. Complications related to laparoscopy, such as intestinal or vascular injury during trocar insertion, potential for adhesions, the need for general anesthesia, and increased cost, are potential downsides of laparoscopic repair. Prospective randomized trials have been performed showing advantages of laparoscopic repair for one or more of the above-cited parameters. [22], [23] and [24] However, many surgeons still feel the potential risks associated with laparoscopy and the increased cost outweigh the advantages and do not routinely recommend laparoscopic hernia repair. The laparoscopic approach has been recommended by some surgeons as ideal for repair of recurrences, for bilateral hernias, and for patients who need to return to a very active lifestyle rapidly. [19], [25]

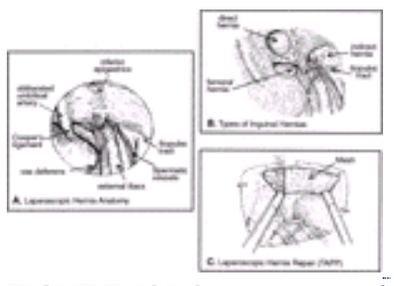

FIGURE 121-8. Laparoscopic repair of inguinal hernias. **A:** Anatomy of the inguinal region as seen through the laparoscope using the transaabdominal preperitoneal (TAPP) approach. **B:** Spaces through which hernias commonly occur as seen through the laparoscope. **C:** After reduction of the hernia, the polypropylene mesh is secured over the inguinal floor using surgical clips or staples.

Pelvic Hernias The intestine can herniate through the pelvic floor (weakness, perhaps, from multiparity or previous trauma) in areas such as the obturator foramen, the greater or lesser sciatic foramina, or the perineal muscle. The most common of these hernias is the *obturator hernia*. The obturator foramen is covered by a membrane except for one small area, and herniation occurs when the intestine (usually ileum) enters the foramen through this small defect and into the obturator region of the thigh (Fig. 121-9). This hernia is seen more commonly in females (male-to-female ratio of 1:6), on the right side, and in the sixth decade of life. Bilateral hernias can be seen, albeit rarely. [26] Most obturator herniations present with acute intestinal obstruction with no previous signs or symptoms. Occasionally, dysesthesias occur in the medial thigh because of compression of the obturator nerve. The Howship-Romberg sign (pain in the medial thigh radiating to the knee or hip exacerbated with extension, adduction, or medial rotation of the thigh) is present in 50% of the patients. A tender mass may be palpated near the obturator canal on rectal or vaginal exam. Radiologic studies may show a gas shadow in the region of the obturator foramen; a small loop of intestine in this region can be also seen on CT. [26] Surgical treatment using a transabdominal approach is indicated in all cases. [27], [28]

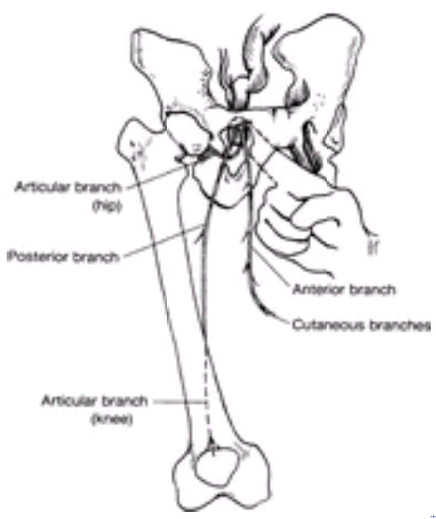

FIGURE 121-9. Rectal or vaginal palpation of the obturator foramen may confirm the existence of an obturator hernia. (Adapted from Nyhus LM, Condon RE, eds. Hernia. Philadelphia: JB Lippincott, 1978.)

Sciatic hernias are extremely rare and can present with a slowly enlarging mass in the gluteal fold area. A CT scan is often needed for diagnosis, and treatment is surgical using a transabdominal or combined transabdominal and gluteal approach. *Perineal hernias* occur through the pelvic floor musculature, and symptoms can vary but are usually related to a protruding mass in the perineal area, worst with standing. These hernias can be seen after removal of the rectum in abdominoperineal resection for rectal carcinoma. Repair is undertaken using a transabdominal approach; polypropylene mesh may be needed.

Lumbar Hernias The posterior abdominal wall has two naturally weak areas. The superior triangle of Grynfelt-Lesshaft, also called the *lumbocostoabdominal triangle,* is the most common site for lumbar herniation. This area is bordered by the 12th rib, the posterior border of the internal oblique, and the anterior border of the sacrospinalis. The inferior triangle of Petit or lumboiliac abdominal triangle is limited by the iliac crest, the anterior border of the latissimus dorsi, and the posterior border of the external oblique muscle. Lumbar hernias are seen most commonly in men and are either congenital (20%), acquired (50%), or due to trauma (25%), for example, rib fractures or removals, iliac crest fractures, or removal of a segment of iliac crest for bone grafting. [13] These hernias are usually left sided. Symptoms usually include a tender mass or pain in the lumbar area or radiating to the pelvis. Lumbar hernias should be surgically fixed when first noted because they tend to enlarge, and larger hernias are more difficult to repair.

Spigelian Hernias A Spigelian hernia is rare, occurring through the linea semilunaris, lateral to the rectus abdominis, with protrusion through the external oblique fascia. Spigelian hernias occur in elderly people, are often small and difficult to diagnose, and always present below the arcuate line of Douglas. Symptoms include local pain or discomfort worsened by increased intra-abdominal pressure. Diagnosis can be made with ultrasound or CT scan if physical examination is difficult

because of obesity or if differentiation is needed from abdominal wall tumor. [29] Surgical repair is needed because of the risk for incarceration; the results are excellent.
Diaphragmatic Hernias Acquired diaphragmatic hernias are the result of blunt or penetrating trauma. Most hernias occur on the left, and herniated viscera can include stomach, spleen, colon, or the left lobe of the liver. Herniation and traction on the mesentery can lead to nausea, vomiting, diaphoresis, and general abdominal discomfort. Respiratory symptoms occur if there is lung or mediastinal compression. Some patients present years after a traumatic injury. [30] The diagnosis is made based on physical examination with dullness to percussion along the left chest; and chest radiograph shows abdominal viscera or a nasogastric tube curled in the thorax (Fig. 121-10). CT scans can be useful in visualizing defects with protruding viscera. [31] If the index of suspicion is high for a diaphragmatic injury, normal radiologic studies should not deter one from laparoscopy or surgical exploration of the diaphragm. [32] All diaphragmatic hernias should be fixed upon diagnosis to avoid strangulation or acute respiratory distress. Chronic herniations may be best approached transthoracically to allow release of intrathoracic adhesions, but most can be repaired transabdominally.

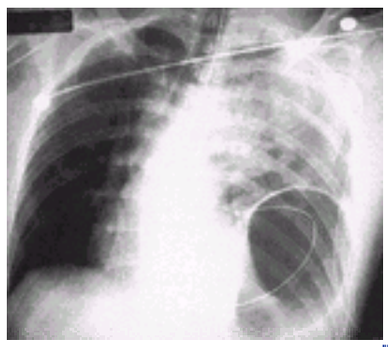

FIGURE 121-10. Traumatic hernia of the diaphragm on the left. The diaphragmatic line has disappeared and the gastric fundus is in the left thoracic space. The contour of the nasogastric tube should be noted in the left chest.

Internal Hernias An internal hernia is a protrusion of any intraperitoneal viscus into a compartment within the abdominal cavity. There is no hernia sac, and most often, the herniated viscus is entering a known anatomic space or foramen; some hernias occur in surgically created or congenital defects. In one review, 0.2% to 0.9% of cadavers had incidentally noted internal hernias, most commonly paraduodenal (53%), followed by pericecal (13%) and the foramen of Winslow (8%), and supravesical and intersigmoid hernias (6% to 7% each). [33] Most symptoms are intermittent, and patients often present with small bowel obstruction or signs and symptoms of strangulation. Radiologic studies are not always helpful, and the differential diagnosis must include foreign bodies, intestinal volvulus, adhesions, and tumors. [34] , [35] Surgical treatment is needed and includes reduction of the herniated viscus and repair of the defect.
Paraduodenal Hernias The left paraduodenal fossa of Landzert is a peritoneal pocket located lateral to the fourth portion of the duodenum and posterior to the inferior mesenteric vein. A left paraduodenal hernia (75%) results from a dorsal malrotation of the midgut resulting in a hernial opening with the inferior mesenteric artery covering the anterior margin. A right paraduodenal hernia (25%) is formed by incomplete rotation of the midgut such that the small intestine is in a sac covered anteriorly by the mesocolon and the superior mesenteric artery. The cecum remains in the right upper quadrant and is attached to the right abdominal wall by thickened parietocolic peritoneal bands of tissue called *Ladd bands*. Signs and symptoms are elusive and can include any signs associated with chronically incarcerated hernias. Contrast radiographs show stasis and delay in transit with large portions of small bowel trapped in one anatomic position. Surgical correction is necessary, and damage to the small bowel vasculature should be carefully avoided.
Hernias of the Foramen of Winslow Herniation of a mobile cecum and distal ileum can occur into a larger than usual foramen of Winslow. Signs and symptoms are vague but can include crampy epigastric pain relieved with flexion of the torso. Radiographic studies show a gas-filled loop of bowel behind the stomach and pushing the stomach anteriorly and to the left. Surgery with reduction and cecopexy may be needed.
Pericecal Hernias, Intersigmoid Hernias The distal ileum can pass through a defect in the cecal mesentery occupying the right paracolic gutter. Most present with right lower quadrant pain with or without a small bowel obstruction. Intersigmoid hernias are composed of herniated small bowel present in the peritoneal pocket between the two loops of sigmoid. These hernias are usually picked up incidentally at operation or during a radiologic study such as barium enema or CT. [35]
Transmesenteric and Transomental Hernias About 5% to 15% of internal hernias occur through defects in the mesentery or the omentum. Transmesenteric hernias occur most commonly in children, along the mesentery of the jejunum. Prenatal ischemic events may be the causative agent because the area of defect is often associated with atretic segments of intestine. Symptoms of small bowel obstruction may develop, and surgery is required at diagnosis.
Iatrogenic Hernias Iatrogenic hernias result from alteration of intra-abdominal anatomy, most often at surgery. Seventy-five percent of iatrogenic hernias manifest in the first postoperative year.
Retroanastomotic hernias. A retroanastomotic hernia occurs through any defect in the mesenteric space left open after completion of an intestinal anastomosis. Hernias can occur after a Billroth II anastomosis in the space between the gastrojejunostomy and the posterior abdominal wall. Most patients present with intestinal obstruction and require immediate surgical correction.
Incisional hernias. Incisions of the abdominal wall result in future herniation 2% to 5% of the time. Multiple factors are responsible for a postoperative incisional hernia, including defective suture material, undue tension on the sutured fascia, obesity, previous incisions, infections, seromas or hematomas, malnutrition, and smoking. [36] Different incisions have different rates of future herniation. Midline incisions have the highest rates; paramedian and transverse incisions have lower rates of herniation. Incisional hernias can range in size from 1 cm to large hernias containing nearly all the intestines. With increasing popularity of laparoscopic surgery, attention needs to be paid to all trocar site defects larger than 0.5 cm to avoid future herniation. The diagnosis of incisional hernia is usually evident by history as well as physical exam. The hernial ring is often palpable with the edges of the muscle retracted laterally. Incisional hernias often incarcerate but tend not to strangulate. Operative repair should be undertaken in most patients. For many years, the repair of incisional hernia was associated with a high recurrence rate. Most recurrences are thought to be due to infection, insufficient dissection and exposure of other defects, or closure under excess tension. Tension should be avoided at the time of repair, and prosthetic mesh is often needed. In more recent years, the introduction of synthetic prosthetic materials has provided the opportunity to perform a tension-free repair, thereby reducing the rate of recurrence. [36] In addition, more recently, musculoaponeurotic flaps are used along with relaxing incisions in cases in which prosthetic material is best avoided. In addition, natural tissue substitutes, such as fascia lata or pig small intestinal submucosa, are being used to repair incisional hernias. [37]

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CHAPTER 122

Frederic E. Eckhauser, James A. Knol, Michael W. Mulholland, and Alicia M. Terando

INTRA-ABDOMINAL ABSCESSES AND FISTULAE

INTRA-ABDOMINAL ABSCESSES

An *intra-abdominal abscess* is a well-defined collection of pus that is isolated from the rest of the peritoneal cavity by inflammatory adhesions, omentum, abdominal viscera, and loops of intestine. Abscesses may or may not contain viable bacteria and represent successful but incomplete attempts of normal host defense mechanisms to eradicate peritoneal infection. Complete resolution of an abscess is impaired by the inaccessibility of its contents to host cellular defenses.

Intra-abdominal abscesses may occur within the peritoneal cavity or the retroperitoneum; they may be either within or adjacent to abdominal viscera. Visceral abscesses usually result from lymphatic or hematogenous spread of bacteria to the organ. Intraperitoneal, nonvisceral abscesses most commonly follow perforation of an abdominal viscus or anastomotic dehiscence. Less frequently, nonvisceral abscesses may occur after incomplete resolution of diffuse peritonitis with a loculated, residual site of infection. Retroperitoneal abscesses may occur as a result of perforation of the gastrointestinal tract or through lymphatic or hematogenous spread of bacteria to retroperitoneal organs. Pancreatic abscess is an example of the latter mechanism.

Intra-abdominal abscesses frequently arise in a postoperative context, with one half to three fourths occurring after an abdominal, particularly colonic, operation. [1] Most postoperative abscesses result from a documented anastomotic leak. In decreasing order of frequency, diverticular disease, appendicitis, inflammatory bowel disease, and perforated visceral carcinomas account for primary, noniatrogenic intra-abdominal abscesses.

Anatomic Considerations

The peritoneal attachments of the intra-abdominal organs and the small bowel mesentery divide the intraperitoneal space and have an important influence on abscess formation. The transverse colon, its mesentery, and the attached omentum separate the abdominal cavity into upper and lower spaces. The upper abdominal space, in turn, is divided into left and right compartments by the falciform ligament. The diagonal attachment of the small bowel mesentery divides the lower abdominal compartment into left and right spaces.

In good health, the peritoneal cavity contains a small amount of fluid that circulates within the abdominal cavity in a pattern determined by visceral attachments. Fluid adjacent to the small bowel mesentery moves dependently into the pelvis and then moves caudally along the right and left pericolic gutters. Peritoneal fluid on the right reaches the corresponding subhepatic and subphrenic spaces, whereas left-sided fluid is impeded from reaching the left subphrenic space by the phrenocolic ligament. The peritoneal fluid circulation favors unilateral spread of bacteria within the abdominal cavity. For example, duodenal perforation frequently is associated with right subphrenic collections, whereas a perforated appendicitis may result in right subhepatic abscess. Intraperitoneal fluid is absorbed along the inferior surface of the diaphragm by specialized lymphatic vessels.

The foramen of Winslow is formed by the free border of the gastrohepatic omentum and the posterior parietal peritoneum (Fig. 122-1). The lesser sac communicates with the general peritoneal cavity through the foramen, but the amount of cross-circulation is small, and perforations within the general cavity rarely produce infection within the lesser sac. Conversely, primary lesser sac abscesses remain confined and usually do not produce diffuse peritoneal contamination (Fig. 122-2).

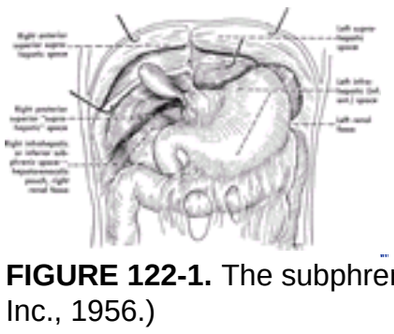

FIGURE 122-1. The subphrenic spaces, the diaphragm and liver being elevated. (From Hollinshead WH. Anatomy for surgeons. Vol 2. New York: Paul B. Hoeble, Inc., 1956.)

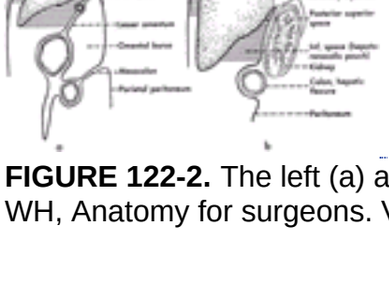

FIGURE 122-2. The left (a) and right (b) subphrenic spaces, as seen in diagrammatic parasaggital sections. The spaces are horizontally lined. (From Hollinshead WH, Anatomy for surgeons. Vol 2. New York: Paul B. Hoeble, Inc., 1956.)

The retroperitoneum is a potential space bordered superiorly by the diaphragm, inferiorly by the pelvic brim, laterally by the quadratus lumborum muscles, and posteriorly by transversalis fascia. The renal fascia separates the retroperitoneum into anterior and posterior compartments. The anterior retroperitoneal space contains the esophagus, pancreas, bile duct, portal and splenic veins, sometimes the appendix and border portions of the duodenum, and the posterior surfaces of the ascending, descending, and sigmoid colonic segments. Posterior perforation in any of these intestinal segments can result in retroperitoneal abscess. The posterior retroperitoneal space contains the kidneys, ureters, gonadal vessels, inferior vena cava, aorta, and lymphatic channels. Extra-abdominal manifestations may occur because of these anatomic relationships. Retroperitoneal perforation of a cecal carcinoma may cause an abscess, which initially develops along the psoas muscle and then passes through the femoral canal to produce an abscess in the right thigh (Fig. 122-3).

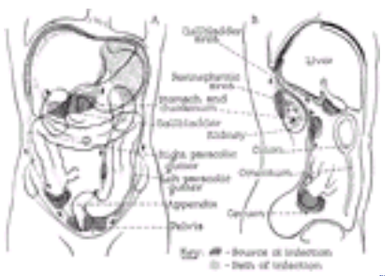

FIGURE 122-3. Etiology and paths of infection causing subdiaphragmatic abscess. (From Anson BJ, Maddock WG. Callander's surgical anatomy. 4th ed. Philadelphia: WB Saunders, 1958.)

Etiology

Primary peritonitis (spontaneous bacterial peritonitis) is defined as inflammation of the peritoneal cavity without a source of contamination from the gastrointestinal tract (see Chapter 46). *Secondary peritonitis* occurs because of necrosis or perforation of the gastrointestinal tract with spillage of enteric contents and bacterial contamination. Intra-abdominal abscesses occur as a consequence of secondary peritonitis. After peritoneal contamination, bacteria are initially cleared by subphrenic lymphatics, undergo phagocytosis by resident peritoneal macrophages, and are sequestered by fibrin deposition. Continued bacterial contamination promotes the influx of polymorphonuclear leukocytes, increases capillary permeability, and results in fluid exudation. These peritoneal defense mechanisms are crucial for effective bacterial clearance and killing, but they also can have adverse effects for the host. Lymphatic clearance of microorganisms can lead to sepsis and development of distal sites of infection. Increased capillary permeability with fluid exudation dilutes bactericidal opsonic activity and inhibits phagocytosis. Fibrin entraps bacteria, inhibiting phagocytic migration and function. Whereas these events help initially to localize peritoneal contamination, they also promote formation of intra-abdominal abscesses.

The normal bacterial flora of the esophagus, stomach, and duodenum is sparse and is composed of swallowed viridans streptococci, microaerophilic streptococci, *Lactobacillus* species, *Fusobacterium* species, and *Candida* species. Bacterial counts increase and enteric organisms proliferate in the presence of achlorhydria or gastric or duodenal obstruction.

The small bowel flora from the proximal jejunum to the distal ileum is characterized by progressively increasing numbers of Enterobacteriaceae, *Enterococcus* species, and anaerobic species. The distal ileum has an endogenous flora that resembles the colon. The colonic lumen contains up to 1012 organisms per gram of stool, with anaerobes outnumbering aerobes by a ratio of 1000:1. Anaerobic organisms include *Bacteroides fragilis, Eubacterium* species, and *Bifidobacterium* species. Aerobic organisms include *Escherichia coli, Klebsiella* species, *Enterococcus* species, and *Proteus* species.

With secondary peritonitis, polymicrobial anaerobic and aerobic infection is expected. Polymicrobial infections are documented in 60% to 80% of cases, with an average of four distinct isolates per patient at the time of initial operation. [2] *B fragilis* is the anaerobic bacteria isolated most often; *E coli* is the most common aerobe. Most chronic intra-abdominal abscesses are polymicrobial, but only aerobes are cultured in more than half of cases. [3] Abscess formation is promoted in the presence of barium, feces, necrotic tissue, or hemoglobin. These adjuvants promote the growth of bacteria by blocking lymphatic clearance, inhibiting bacterial clearance and killing, and providing bacterial nutrients such as iron. [4] Preexisting immune compromise and prior antibiotic exposure may significantly influence the bacteriologic spectrum of intra-abdominal abscesses in seriously ill patients. In two studies of patients in intensive care units, a reduced frequency of *E coli* and *B fragilis* isolates was noted, with an increased frequency of fungi, *Staphylococcus epidermidis, Pseudomonas* species, *Enterococcus* species, and *Enterobacter* species. [5], [6]

Clinical Manifestations

Generalized peritonitis produces intense abdominal pain and obvious physical findings; the diagnosis is seldom in doubt. Patients with intra-abdominal abscesses are usually subacutely to chronically ill, and symptoms and physical findings may be subtle or sometimes equivocal. Fever and chills are extremely common. Malaise and anorexia are universal but also nonspecific. Leukocytosis with a shift to immature forms is usually present. Most other common laboratory tests are noninformative.

An abscess that abuts the parietal peritoneum may produce localized pain, tenderness, and a palpable mass. In contrast, intramesenteric and interloop abscesses usually do not contact the somatically innervated parietal peritoneum and produce diffuse, poorly localized visceral pain; a mass usually cannot be appreciated. Subphrenic abscesses often are associated with diaphragmatic splinting, tachypnea, cough, and hiccup. An associated pleural effusion is frequently present and can be detected by physical examination or chest radiograph. Pelvic abscesses may produce tenesmus, diarrhea, or urinary retention as a result of irritation to the adjacent rectosigmoid colon or bladder. These symptoms should prompt rectal and pelvic examinations to detect mass, fluctuance, or tenderness. Retroperitoneal abscesses produce fewer constitutional symptoms than intraperitoneal processes. Lumbar or iliopsoas muscle spasm may be present, and involvement of psoas muscle may cause pain on flexion of the hip (psoas sign).

Detection of intra-abdominal abscesses in postoperative patients is especially difficult. Incisional pain, the effects of postoperative analgesics and antibiotics, and the impediment of incisions, drains, and bandages to physical examination compound the problem. The mean time postoperatively to clinical signs of abscess is 8 days. [7]

Diagnostic Studies

Plain abdominal radiographs, intralumenal contrast studies, and radioisotope scanning have unacceptably low diagnostic accuracy rates, ranging from 15% to 50%, when used for the detection of intra-abdominal infection. [8] These older modalities have been supplanted by computed tomography (CT) and ultrasonography.

Ultrasonography Ultrasonography has been used frequently as the initial screening and diagnostic tool for investigation of suspected intra-abdominal abscess. Ultrasonography has the advantages of portability, speed, low cost, and lack of radiation exposure. The anatomic areas best suited for ultrasound investigation are the right upper quadrant, the pelvis, and the left upper quadrant if the spleen is present. Ultrasound also has significant limitations for the detection of intra-abdominal abscesses: it is operator dependent in its performance, and the images are not intuitively understandable or accessible to many nonradiologists. Obesity, open wounds, surgical dressings, ostomy appliances, and overlying bone severely limit ultrasound access. Because gas reflects transmitted sound waves, intestinal ileus and overlying lung preclude ultrasound examination of many areas of the abdomen. These limitations are most pronounced in postoperative patients, the group at highest risk for abscess and the group in which clinical signs and physical examination are least reliable. Reported sensitivities of ultrasound for detection of intra-abdominal abscess range from 75% to 80%. [9]

Computed Tomography Computed tomography is the superior mode for investigating patients with suspected intra-abdominal abscesses (Fig. 122-4). CT scanning is much less operator dependent than ultrasonography, the visual information is less abstract, and CT provides better anatomic resolution that is not affected by surgical incisions, bandages, body habitus, or the presence of bowel gas. Intralumenal fluid collections, interloop areas, and retroperitoneal structures all can be visualized.

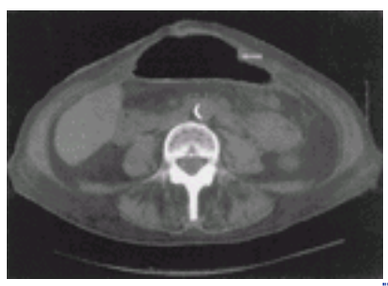

FIGURE 122-4. Computed tomography scanning provides a highly accurate means for evaluating the presence of an abscess and its relationship to contiguous visceral and vascular structures. Localized accumulations of fluid of low attenuation plus the presence of loculations, gas, or debris are highly suggestive of an abscess. The *arrow* points to a large, gas-filled loculation in the mid-upper abdomen that eventually proved to be a localized abscess.

The presence of an abscess is suggested by a localized accumulation of fluid of low attenuation (Fig. 122-5). CT provides information about the size and shape of cavity walls as well as the presence of loculations, gas, or debris. CT has a reported sensitivity of 80% to 100% for the detection of intra-abdominal abscesses. [10] Interloop abscesses are detected less reliably, with about 60% sensitivity.

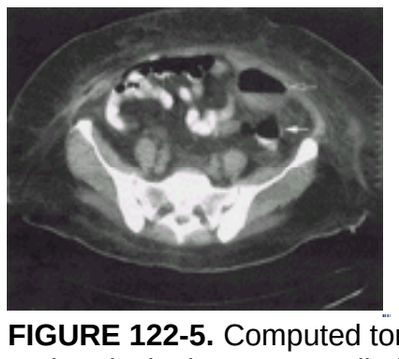

FIGURE 122-5. Computed tomography findings suggestive of diverticular abscess include thickening of the colonic wall (*closed arrow*) along with mesocolic edema and pericolonic or mesocolic fluid accumulations. The *open arrow* points to a localized collection of fluid and gas that represents a pericolonic diverticular abscess.

CT-guided diagnostic aspiration is not complicated and may be performed with the patient under local anesthesia using an 18- to 22-gauge needle. The needle is directed into the fluid collection, often using additional CT imaging for guidance. Selection of a safe route for diagnostic aspiration is crucial. The sampling needle must not transgress intestine before entering the fluid collection. False-positive results may occur if intestinal or colonic contents are inadvertently aspirated. In addition, sterile fluid collections, such as a pseudocyst or hematoma, could be contaminated and converted into an abscess. Samples should be analyzed by Gram stain and for aerobic and anaerobic culture. Usually, a decision regarding therapeutic drain placement can be made at the time of diagnostic sampling on the basis of Gram stain.

Endoscopic Ultrasound Endoscopic ultrasound (EUS) has recently been recommended as an adjunct for localizing pancreatic pseudocysts or abscesses. EUS has been found to be helpful in precisely identifying pseudocysts, thereby facilitating accurate and complete endoscopic or open cyst decompression. [11], [12] The presence or absence of associated varices resulting from portal hypertension can be determined, thereby redirecting the site of decompression and improving the safety of the procedure. EUS can also be helpful to differentiate between a cystic neoplasm and an inflammatory pseudocyst. [13], [14] When combined with cyst fluid aspiration and fine-needle aspiration cytology, EUS can provide useful information to guide subsequent therapy, particularly for lesions that are not amenable to CT or ultrasound-guided drainage because of their location or anatomy. This can be especially useful in patients with evidence of occult, undrained infection and loculated fluid collections.

Therapy

Successful management of patients with intra-abdominal abscesses is based on the following principles: control of the source of intraperitoneal or retroperitoneal contamination; drainage of established abscess cavitiesAelimination of residual bacterial infection through use of appropriate antibiotics; and physiologic support of the patient. Patients with acute secondary peritonitis require immediate operation to resect necrotic or perforated tissue, to remove blood or intestinal contents, and to eliminate continuing contamination. In contrast, patients with established intra-abdominal abscesses, especially those with postoperative sepsis, present more difficult decisions. Although delayed therapy is associated with poor outcome, so too is an inappropriate or poorly executed operation. Patient selection is greatly aided by cross-sectional imaging techniques combined with the judicious use of percutaneous fluid aspiration and continuous external catheter drainage. CT-guided therapy has permitted many patients to avoid laparotomy and has helped to convert others from emergent operation to staged, urgent, or elective procedures.

Established intra-abdominal abscesses can be drained percutaneously using CT or ultrasound guidance, by transperitoneal laparotomy, or by an extraperitoneal surgical approach. Most studies comparing percutaneous drainage with surgical drainage are flawed in that they are not prospective or randomized and include patients with widely varying etiologies of abscess. Nonetheless, the overall conclusion is that percutaneous drainage of intra-abdominal abscesses can be performed as safely as surgical drainage and with similar long-term clinical outcome. [15] The most important issue is proper patient selection and not the technique used; severity of illness and the nature of underlying intra-abdominal pathology determine outcome.

Percutaneous catheter drainage requires a safe drainage route that avoids solid organs and bowel. A safe passage can be identified in 85% to 90% of cases. [16] Complex or loculated cavities, suggested by persistent fever and leukocytosis, may require the placement of additional catheters. Necrotic tissue, thick pus, and infected hematomas respond less well to catheter drainage. Fungal abscesses, which have thick pus and invade surrounding tissues, usually contraindicate percutaneous drainage. Extensive collections; interloop, intramesenteric, splenic, and pancreatic abscesses; and abscesses that communicate with bowel have lower success rates and require individualized judgment. The success rates for percutaneous CT-guided drainage of established, well-defined, unilocular intra-abdominal abscesses is 80% to 90%. [17] The rates for complex abscesses, as defined herein, are significantly lower, about 50%.

Complications relating to the performance of percutaneous drainage occur in 5% of patients with simple abscesses and 20% of those with complicated fluid collections. [18] Bacteremia is the most common complication. Catheter manipulation and irrigation increase the risk for bacteremia; prophylactic intravenous antibiotics should be administered before catheter changes. Bleeding can occur because a catheter traverses a vessel during insertion or erodes a vessel over time. Inappropriate placement trajectories that pass through the pleural space may result in pneumothorax, empyema, or pleural effusion. The complication rates for percutaneous drainage are generally lower than for surgery when equivalent patient groups are compared. Surprisingly, mortality rates are similar, suggesting that preexisting illness and the underlying cause of abscess are more important determinants of mortality than the technique of drainage. [19]

Surgery

Operative therapy should be individualized for each patient and depends on the underlying pathology as well as the resulting intra-abdominal abscess. Surgical treatment of established intra-abdominal abscesses requires either a transperitoneal or an extraperitoneal surgical approach. Drainage of a subphrenic abscess posteriorly through the bed of the 12th rib is an example of extraperitoneal drainage. Extraperitoneal drainage has the advantages of not violating the peritoneum and spreading bacterial contamination and is generally less physiologically stressful. Extraperitoneal procedures do not permit detailed evaluation for coexisting intraperitoneal pathology, which is present in up to one fourth of these patients. [20]

Patients with coexisting intraperitoneal pathology, such as intestinal infarction, anastomotic dehiscence, or extensive infected hematoma, require transperitoneal laparotomy to address the primary problem. In these patients, a number of additional techniques have been proposed to treat the septic component more aggressively, including radical peritoneal debridement, intraoperative lavage, continuous postoperative lavage, and open abdominal drainage. In controlled, prospective trials, none of these techniques has conferred significant benefit. [21], [22], [23], [24] and [25]

Appendiceal Abscess

An abdominal mass is be present on physical examination in about 2% to 3% of patients with appendicitis at their initial evaluation. [26] A periappendiceal mass, which

is formed by inflamed omentum and adherent loops of intestine, usually indicates appendiceal perforation and signifies the development of a phlegmon or an abscess. The risk for perforation is increased if the patient has been symptomatic for more than 24 hours, temperature exceeds 38°C, or the white blood cell count is greater than 15,000/mL. The wound infection rate is 1.8% after an uninflamed appendix is discovered at laparotomy, 8.5% if the appendix is inflamed but not perforated, and 17% with appendiceal perforation. [27] Pelvic abscesses occur postoperatively in 15% of patients with appendiceal perforation.

An appendiceal mass or abscess may be treated by both operative and nonoperative means, although immediate operation is not favored because the intense inflammatory reaction increases the risk for injury to adjacent bowel or ureter. CT is not justified as a routine procedure in the diagnosis of acute appendicitis, but it is essential in the nonoperative management of a periappendiceal mass. Initial management should include percutaneous drainage of periappendiceal fluid collections and intravenous antibiotics. Elective interval appendectomy, which can be performed after the resolution of periappendiceal inflammation, is associated with a 0.2% mortality rate and a major complication rate of only 1.2%. [28]

Diverticular Abscess

CT, the preferred initial test when a diverticular abscess is suspected, [29] has several advantages: it causes less discomfort than barium enema, the risk for inducing a perforation is negligible, and pericolonic complications of diverticulitis can be evaluated. CT findings that indicate diverticulitis include thickening of the colon wall, mesenteric edema, pericolonic and mesenteric abscesses, and pneumoperitoneum (see Fig. 122-5). If diverticulitis is suspected, intravenous contrast should not be administered initially; visualization of contrast material (administered orally or rectally) within the bladder is presumptive evidence for colovesical fistula. The sensitivity of CT is reported to be 65% for uncomplicated diverticulitis and 90% to 100% for detecting the complications of perforation, abscess, or fistula. [30]

The ability of CT to define accurately the complications of acute diverticulitis has had a major impact on clinical management. Cross-sectional imaging of pericolonic abscess permits CT-guided percutaneous drainage (Fig. 122-6), which minimizes the need for colostomy formation, provides optimal conditions for resection with primary anastomosis, and improves patient safety. Small pericolic or intramesenteric abscesses that can be resected en bloc with the sigmoid colon usually do not require preoperative percutaneous drainage. Diverticular abscesses associated with fecal peritonitis, pneumoperitoneum, or colonic obstruction should be treated by emergent operation. The diseased colonic segment should be resected and the abscess debrided. The formation of a proximal colostomy and closure of the rectum (Hartmann procedure) are the safest surgical options. The colostomy is closed in 2 to 3 months after resolution of the septic process.

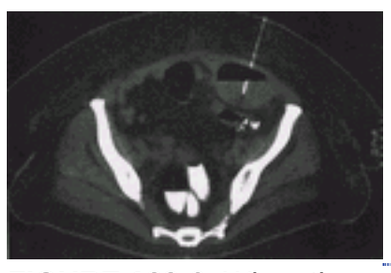

FIGURE 122-6. When they are localized, pericolonic diverticular abscesses may be amenable to percutaneous aspiration and continuous catheter drainage. The suspected abscess depicted in this figure is being safely aspirated using computed tomography guidance.

Patients treated by percutaneous drainage of diverticular abscesses should experience definite clinical improvement within 24 to 48 hours, with decreased pain, defervescence, and diminished leukocytosis. Interval catheter injection is used to demonstrate progressive collapse of the abscess cavity. In a selected series of patients, percutaneous drainage was associated with resolution of sepsis in 89%. [31] Fistulous communication with the colon was demonstrable radiologically in 47% but clinically significant in only 16%. In three fourths of treated patients, single-stage sigmoid resection with primary anastomosis was performed subsequently without complication.

Pancreatic Abscess

CT of the pancreas is used to estimate the severity of acute pancreatitis and to distinguish acute pancreatitis from pancreatic abscess. [32] Glandular enlargement, loss of internal structure, and blurring of pancreatic margins characterize mild disease. Increasing severity is demonstrated by the development of peripancreatic fluid collections. Severe cases are associated with fluid collections in the lesser sac, perirenal and posterior pararenal space phlegmons, and pancreatic ascites.

Contrast-enhanced CT is the preferred method of identifying necrotizing pancreatitis, which may serve as a precursor for pancreatic abscess. In enhanced CT pancreatography, thin tomographic cuts of the pancreas are obtained during infusion of intravenous contrast medium. Contrast-deficient areas correlate with regions of pancreatic nonperfusion and necrosis. The sensitivity of dynamic CT for detecting pancreatic necrosis has been reported to be 85% to 95%. [33], [34]

Pancreatic abscess represents one form of intra-abdominal abscess in which operative drainage remains the primary therapeutic modality. CT imaging is essential for diagnosis and as a guide for the timing of therapy, but it has a lesser therapeutic role than for other forms of intra-abdominal abscesses. In pancreatic abscess, interventional radiology has a supportive rather than primary role. Pancreatic abscesses tend to be poorly localized and to contain solid, devitalized material that cannot be evacuated by catheter (Fig. 122-7). Operative therapy for a pancreatic abscess is more properly termed a debridement rather than an abscess drainage. In one study, percutaneous drainage was successful in only 10% of pancreatic collections. [35]

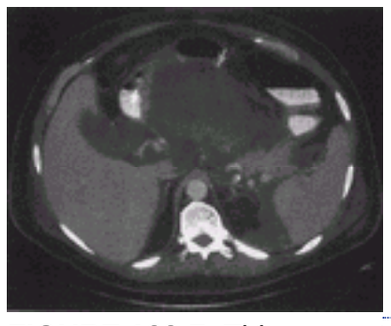

FIGURE 122-7. This computed tomography scan of a patient with a suspected pancreatic abscess (*arrow*) shows fairly typical findings: relatively poor localization, surrounding inflammation, and poor contrast enhancement suggesting devitalized pancreas or peripancreatic fat and debris. These lesions generally are not amenable to percutaneous catheter drainage because they contain solid debris and are best managed with surgical debridement and drainage.

GASTROINTESTINAL FISTULAE

A *fistula* is defined as an abnormal communication between the lumen of a hollow viscus and another hollow organ or the integument. Abdominal fistulae generally are classified by their sites of origin and termination, by the volume and composition of drainage, and by their etiology. They pose difficult management problems because of the resulting fluid, electrolyte, and protein losses. These losses, coupled with the increased metabolic stress of recent surgery, infection, or malignancy, may lead to significant malnutrition. Before 1970, mortality and complication rates associated with abdominal fistulae exceeded 50%. Advances in parenteral and enteral forms of nutrition and the refinement of surgical techniques have improved the rate of fistula closure and reduced associated comorbidity and mortality.

Etiologies

Gastrointestinal fistulae develop from multiple factors, including complications of abdominal surgery (anastomotic dehiscence, erosion by an adjacent drain, unrecognized iatrogenic injury), blunt or penetrating trauma, inflammatory bowel disease including Crohn's disease and diverticulitis coli, malignancy, and radiation. Prosthetic mesh materials used to reconstruct the large abdominal wall or pelvic floor defects have been associated with enterocutaneous fistulae, presumably by erosion of the mesh through the adjacent bowel wall. Most enteric fistulae develop as complications after surgery.

Clinical Manifestations

The major clinical problems of gastrointestinal fistulae include fluid and electrolyte losses, skin excoriation, hypermetabolism, abscess formation, sepsis, and malnutrition. Loss of gastric juice, containing high concentrations of hydrogen and chloride ions, can result in hypochloremic, hypokalemic metabolic alkalosis. Loss of alkaline pancreatic secretions can produce significant metabolic acidosis. Large-volume fluid losses result in dehydration and oliguria, and, if uncorrected, may progress to altered renal function. The presence of activated digestive enzymes in the fistula effluent may cause skin excoriation, especially when the external opening of the fistula is located at the base of a skin crease or in a surgical wound that impairs secure adherence of a protective dressing.

Sepsis frequently accompanies fistula development. A localized fistula may rupture freely into the peritoneal cavity or cause an abscess in distant areas such as the pelvic, subhepatic, or subdiaphragmatic spaces. A fistula also may masquerade as a wound infection and become apparent only when enteric contents breach the fascial closure and appear on the dressings. Operative or radiologic drainage of a postoperative abscess also may demonstrate an unsuspected fistula. Rarely, fistulae may develop when an abscess erodes into adjacent bowel.

The importance of aggressive nutritional support in patients with gastrointestinal fistulae cannot be overemphasized. Malnutrition adversely affects reparative and immune processes. Several factors contribute, including protein and nutrient losses from the fistula, the anorexia and decreased food intake associated with malignancy and chronic illness, and the increased metabolic demands imposed by recent operation. Before 1970, the reported mortality and morbidity rates associated with enterocutaneous fistulae ranged from 40% to 65%. [36] These figures have been reduced to 15% or less with regimens that use aggressive parenteral or enteral nutrition. [37]

Therapy

Fistula management is multifaceted. Adequate external drainage, control of infection, and intensive nutritional support are the keys to improved patient survival. Total parenteral nutrition (TPN) should be used initially, but enteral support with elemental or polymeric diets can be used during the later phases of treatment. Enteral support requires about 1250 cm of intact small intestine irrespective of the fistula location. The caloric and nutrient requirements for enteral nutrition are the same as those for TPN. Immediate surgical correction of the fistula is not generally a priority and may represent an unacceptable risk to an already septic, immunocompromised patient. Undrained abscesses should be searched for aggressively with CT and drained percutaneously whenever feasible with CT or ultrasound guidance. Once the patient has been stabilized, the source organ and extent of the fistula should be evaluated radiographically, preferably with water-soluble contrast media if there is question of continued communication between the fistula tract and the peritoneal cavity.

Intensive nutritional support should be continued for 4 to 8 weeks in anticipation of spontaneous closure of the fistula. Several factors decrease the likelihood of spontaneous closure (Table 122-1) and alter the surgical strategy and require delaying definitive reoperation. The general principles of surgical management include resection, serosal patching or internal drainage of the fistula, correction of any associated pathology, and provision of adequate wound coverage. These fundamentals are essential for decreasing the potential lethality of this condition.

Partial or complete obstruction
Persistent inflammation or infection
Foreign body in fistula tract
Cancer
Inflammatory bowel disease
Radiation enteritis
Fistula tract < 2 cm long
Fistula volume > 500 mL/24 h

TABLE 122-1 Factors that Impair Fistula Closure

Effective management of gastrointestinal fistulae requires establishment of treatment priorities. Dehydration should be corrected rapidly along with any derangements in electrolyte or acid-base balance. Sterile central venous access should be established to initiate and maintain parenteral nutrition. A separate peripheral intravenous catheter should be used to replace measured fistula losses, nasogastric suction volumes, and third-space losses resulting from peritoneal inflammation or hypoalbuminemia.

Control of Sepsis Uncontrolled sepsis is the major determinant of mortality in patients with gastrointestinal fistulae. [38] Control of infection requires systemic antibiotics coupled with surgical or radiologic drainage of abscesses. Signs of occult sepsis, including rigors or chills, unexplained leukocytosis, acid-base disturbances, or evidence of single or multiple organ dysfunction, suggest undiagnosed abscesses that should be investigated aggressively using ultrasonography or CT and percutaneously drained whenever feasible. Patients commonly present with multiple or loculated abscesses that may recur after successful percutaneous or surgical drainage. Antibiotics should be used selectively, based on appropriate culture and sensitivity testing.

Control of Drainage Drainage along the fistula tract also should be established to prevent pooling of secretions, to permit accurate quantification and analysis of fluid losses, and to minimize excoriation of surrounding skin. A fistulogram should be obtained to identify the source organ and to establish optimal drainage. Soft catheters with multiple side holes can be positioned radiologically into the proximal and distal arms of the fistula for collection and reinfusion of gut secretions. Such catheters provide access for enteral nutrition, although their presence will retard and ultimately prevent spontaneous closure of the fistula. Suction catheters should be positioned carefully adjacent to but not through the bowel opening so as not to impair spontaneous closure of the fistula. Failure to configure a reliable device for collection of fistula drainage compromises wound care and complicates fluid and electrolyte management, especially in patients with high-output fistulae. Fastidious care of the skin and tissues surrounding the fistula site is essential. Uncontrolled leakage of gastrointestinal secretions containing activated digestive enzymes causes severe skin excoriation and is both painful and unsanitary for the patient. The help of a qualified and experienced enterostomal therapist can be invaluable in difficult cases.

Management of Nutrition Prompt initiation and maintenance of adequate nutrition are imperative for the successful treatment of patients with gastrointestinal fistulae. Formulations vary, but the essential components include 2000 to 5000 Kcal, 70 to 200 g of amino acids, and 500 to 1000 mL of 10% fat emulsion per 24 hours, plus vitamins, trace minerals, and electrolytes as needed to meet daily requirements. Before the development of TPN, many patients died of malnutrition and starvation. Complete bowel rest and TPN are recommended for patients with proximal or high-volume fistulae. When the fistula is distal and of low volume, enteral nutritional support may be feasible. A variety of nutritional formulations can be used. Enteral nutrition is more economic and physiologic than TPN, avoids the risk for catheter-related and metabolic complications, and yields comparable results. [39] Enteral feeding also prevents gut atrophy and bacterial translocation, potentially reducing the risk for portal pyemia and septic complications. Enteral feeding is associated with well-recognized complications, the most serious of which is aspiration pneumonia; this occurs at a fairly low fixed rate, irrespective of the type of access (nasal or percutaneous) or the terminal position of the catheter (stomach or small intestine). Diarrhea usually can be managed by altering the type, rate, or concentration of the enteral formulation.

Management of Fistula Drainage Decreasing fistula output is an important component of therapy. This can be accomplished by giving the patient nothing by mouth, inserting a nasogastric tube to evacuate gastrointestinal secretions and ensure bowel rest, and administering drugs or hormones to decrease gastric and pancreaticobiliary secretions. Complete and long-term bowel rest may control fistula output and encourage natural closure but may result in hepatic cholestasis. In one clinical study of patients with enterocutaneous fistulae, collection of effluent from the proximal limb and reinfusion through a soft catheter into the distal limb of the fistula prevented or reversed biochemical evidence of intrahepatic cholestasis. [40] Reinfusion of fistula effluent is generally innocuous and should be considered whenever feasible. Supportive management of gastrointestinal fistulae with TPN, skin care, and control of infection results in spontaneous closure rates of 60% to 75%, although the chances of cure vary according to the anatomy of the fistula and underlying condition or conditions. [41] The characteristics that predict a low likelihood of spontaneous fistula closure are outlined in Table 122-1. In most studies, the mean time to fistula closure with TPN alone is 4 to 5 weeks. [39] The major detractors from conservative treatment are its long duration, high cost, and patient inconvenience. Hormonal suppression of gastrointestinal fistulae with somatostatin,

a tetradecapeptide with potent inhibitory actions on gastrointestinal and pancreaticobiliary secretions, [42] was facilitated by the development of long-acting synthetic analogs (octreotide; SMS 201-995) that are resistant to degradation. Several retrospective studies showed that subcutaneous administration of somatostatin effectively reduces the volume of fistula output and reduces the mean time to spontaneous closure, [42], [43] particularly in patients with high-output fistulae in whom spontaneous closure with conservative treatment rarely exceeds 50%. In one study, subcutaneous administration of SMS 201-995 [42] and octreotide (both at 100 µg/8 hours) rapidly reduced fistula output regardless of the fistula site or pretreatment output and achieved spontaneous closure in 77% of patients, within 14 days [43] (mean treatment duration, 5.8 days). [42] The final closure rate was the same for patients with high- or low-volume output fistulae. Fistula output was not a reliable predictor of fistula closure. Serum albumin levels below 3 g/dL and a history of chemotherapy or radiation had a negative influence on fistula closure.

Nonoperative Management

Endoscopic obliteration with fibrin sealant has been used with varied success in patients with upper gastrointestinal tract fistulae that are small-caliber, straight fistulae of relatively short duration with little or no associated infection. [44] Permanent occlusion is not always achieved, but repeat endoscopic occlusion can be attempted with little risk to the patient. Fibrin sealants stimulate fibroplasia, unlike other compounds, such as the tissue adhesives prolamine or *N*-butyl-2-cyanoacrylate (Histoacryl), which induce a foreign-body reaction. Based on recent reports, large fistulae and fistulae associated with radiation injury or neoplasia respond poorly to this technique.

Operative Management

Despite the development of parenteral nutrition, innovative pharmacological or hormonal therapies, and interventional radiographic techniques, about 20% to 30% of patients with gastrointestinal fistulae require operative closure, usually combined with treatment of associated conditions (e.g., bowel obstruction, foreign body, cancer, damaged bowel). Any experienced surgeon knows these corrective operations can be quite formidable, with high morbidity for the patient. One of the keys to successful fistula treatment is individualization of care. The clinical course of patients with gastrointestinal fistulae tends to be protracted, but continued improvement and eventual resolution can be anticipated if serious management errors are avoided. [45]

The surgical management of gastrointestinal fistulae has changed dramatically since the pre-TPN era, when heroic operations were often necessary to close fistulae and establish gastrointestinal continuity. Adherence to the principles of nonoperative care has been associated with a decreased need for surgical correction. Now, 50% to 80% of simple (single) external fistulae close spontaneously within 4 to 6 weeks. [38], [46] Complex or multiple fistulae occurring within an eviscerated wound are associated with increased mortality and morbidity and invariably require surgical correction. The optimal timing of surgical intervention in these patients must be individualized, but delayed reconstruction is often advisable, and a staged, multidisciplinary approach may be necessary. Despite the complexity of these problems, eventual closure can be achieved in most patients. [47] Many patients require extensive mobilization of skin flaps and the use of skin grafts to close large abdominal wall defects. The optimal timing of reoperation in complex fistula patients is undefined, but delays of 4 to 6 months may be advisable to allow for resolution of intra-abdominal inflammation and adhesions. As inflammatory processes resolve, the abdominal wall generally becomes softer and more compliant.

Gastric Fistulae

Gastric fistulae are uncommon, constituting fewer than 15% of all gastrointestinal fistulae. [48] They may develop after stomach surgery for benign peptic ulcer disease or malignancy resulting from dehiscence of a surgical anastomosis, or they may be caused by inadvertent injury to an otherwise normal segment of stomach wall after splenectomy, surgery for morbid obesity, or highly selective vagotomy. The first indication of a gastric fistula is usually the development of a subphrenic or lesser-space abscess. Diagnosis can be made by oral water-soluble contrast study or CT examination. Urgent laparotomy is warranted in patients with free dye extravasation and signs of generalized peritonitis, but percutaneous CT-directed catheter drainage may be feasible in patients with localized, perigastric fluid collections. The bacteriology of gastric fistulae includes anaerobic bacteria from the oropharynx and fungi, which commonly are identified in conditions associated with hypochlorhydria or malignancy.

The reported success rate for nonoperative treatment of gastric fistulae ranges from 43% to 100%, but there are few reliable criteria that predict success. [45] Radiologic positioning of a nasoenteric feeding tube beyond the fistula site into the distal duodenum affords access for enteral nutrition and avoids the need for TPN in most patients. Definitive reoperation should be delayed for at least 6 to 8 weeks after diagnosis and initial drainage. Surgical options include debridement and two-layer closure of the stomach wall defect or the use of a Roux-en-Y serosal patch or anastomosis. [49] Most gastric fistulae can be closed successfully (about 90%), but the success rate varies with the underlying condition and whether the fistula is simple or complex (e.g., involves other organs, such as the colon or pancreas). [50]

Pancreatic Fistulae

Pancreatic fistulae develop as complications of severe pancreatitis, penetrating or blunt abdominal trauma, or surgery directly on or around the pancreas. The major consequences of pancreatic fistula are metabolic, including bicarbonate loss with resulting acidosis and malabsorption caused by a loss of digestive enzymes. Diversion of alkaline pancreatic secretions from the duodenum also may predispose to mucosal ulceration and warrants treatment with H_2-receptor antagonists or proton pump inhibitors. Additional medical treatment includes pancreatic enzyme supplementation and octreotide acetate to decrease fistula losses. Pancreatic fistulae that require operation are uncommon, especially if there is no evidence of proximal pancreatic duct obstruction or malignancy, but closure may be protracted if the fistula tract is tortuous and associated with a residual abscess cavity. [51]

Endoscopic retrograde cholangiopancreatography is used to identify variations in duct anatomy or duct obstruction that would affect closure. In selected cases, placement of a pancreatic duct endoprosthesis may improve ductal drainage and expedite fistula closure. [52], [53] Pancreatic endoprostheses have been used successfully to treat a variety of chronic inflammatory conditions, with an estimated complication rate of about 20% and rare procedure-related deaths. The major complications are related to obstruction or migration of the stent or erosion of the duodenal wall by a malpositioned endoprosthesis. Additional diagnostic studies, including a CT scan and a fistulogram, should be obtained to detect pathology that might alter management. Nonoperative therapy for pancreatic fistulae is generally successful, and only 10% to 30% of patients ultimately require reoperation. Interestingly, fistula output has not proved to be a reliable predictor of spontaneous fistula closure.

The surgical options for managing a pancreatic fistula include internal drainage of the fistula into a defunctionalized Roux-en-Y limb of jejunum or pancreatic resection. Resection is appropriate for a fistula arising from the pancreatic tail and carries little risk for endocrine and exocrine dysfunction. In contrast, resection of a pancreatic head fistula (Whipple operation) is rarely indicated. The perioperative use of octreotide acetate may decrease the risk for fistula recurrence. Despite the complexity of these problems, reoperative success rates of 85% or greater have been reported from various institutions. [54], [55]

Duodenal Fistulae

Duodenal fistulae usually arise after gastric surgery for peptic ulcer disease or malignancy. The mortality rate of duodenal fistulae remains high (10% to 35%), with uncontrolled sepsis accounting for most deaths. [41] Unlike other gastrointestinal fistulae, early reoperation to establish adequate drainage and abort septic complications should be considered a high priority in duodenal fistula patients.

There are two types of duodenal fistulae. End fistulae result from dehiscence of the duodenal stump after distal gastrectomy and Billroth II reconstruction. The incidence of this complication in postgastrectomy patients ranges from 1.5% to 5%. [56] Lateral fistulae develop after blunt or penetrating trauma or result from inadvertent duodenal wall injury after operations in the right upper quadrant involving the colon, kidney, or biliary tract. Although uncommon, duodenal perforation also can occur after endoscopic interventions for biliary disease, including papillotomy and retrograde stone extraction.

The combined use of TPN, nasogastric suction, proton pump inhibitors, and octreotide acetate markedly diminishes fistula output and facilitates management. Early attempts at definitive reoperation should be resisted because the surrounding inflammatory response will jeopardize a successful result. The reported rates of spontaneous duodenal fistula closure range from 40% to 73%. [45]

Definitive reoperation should be considered after 6 to 8 weeks of conservative therapy. A contrast study of the fistula tract and the distal bowel should be obtained to detect unsuspected obstruction. For simple fistulae, a Roux-en-Y serosal patch technique achieves excellent results. [57] For large defects or dehisced duodenal

stumps, the end of the Roux limb can be anastomosed to the freshened end of the defect. [58] Primary repair of the defect is generally unsuccessful, and resection with primary anastomosis is unnecessary and hazardous, especially for defects located close to the ampulla of Vater.

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CHAPTER 123

B. Mark Evers

DISEASES OF THE PERITONEUM, RETROPERITONEUM, MESENTERY, AND OMENTUM

PERITONEUM

Embryology, Anatomy, and Physiology

The peritoneum is the mesothelial lining of the peritoneal cavity and its contained viscera. Embryologically, it develops from the primitive coelom, which is formed by a splitting of the lateral mesoderm into somatic and splanchnic layers. [1] The parietal peritoneum, derived from the somatic layer, lines the abdominal cavity, diaphragm, and pelvis. The visceral peritoneum, from the splanchnic layer, covers the intraperitoneal organs and forms the mesenteries by which they are suspended.

The peritoneum and mesentery are supplied mainly by the splanchnic blood vessels and, to a lesser extent, by branches of the lower intercostal, lumbar, and iliac arteries. Differences arise in the innervation of the visceral and parietal peritoneum, which lead to different perceptions of painful stimuli. The visceral peritoneum receives afferent innervation from the autonomic nervous system and responds primarily to traction and pressure; painful stimuli are perceived as a poorly localized, dull pain. In contrast, the parietal peritoneum is innervated by both somatic and visceral afferent nerves; noxious stimuli are perceived as a localized, sharp pain with rebound tenderness.

The peritoneum, with a total surface area of about 2 m^2, serves as a bidirectional dialysis membrane through which the passage of both high- and low-molecular-weight solutes is accomplished predominantly by simple passive diffusion. [2], [3] Factors that can alter absorption include intra-abdominal pressure, temperature, pH, increased portal pressure, lymphatic blockade, and scarring of the peritoneum. [4], [5] The peritoneum contains a complex multileveled defense system that provides an innate immune system to protect against inflammatory processes. [6], [7] This system consists mainly of mechanical clearance of the peritoneal cavity; sequestration mechanisms, such as fibrin trapping of bacteria activation of complement, particularly C3a and C5a, which stimulate neutrophil chemotaxis; and the bacteria-killing actions of polymorphonuclear neutrophils and macrophages. The disease processes of the peritoneum discussed in this section include inflammation of the peritoneum (i.e., peritonitis), primary mesothelioma, and pseudomyxoma peritonei.

Peritonitis

Peritonitis involves a local or generalized inflammation of the parietal and visceral peritoneum. With the development of broader-spectrum antibiotics, newer anesthetic techniques, and modern preoperative and postoperative monitoring in the intensive care unit, the mortality from peritonitis has been greatly reduced; however, despite these advances, peritonitis remains a formidable challenge confronting physicians.

Peritonitis can be classified as either *primary* or *secondary*. Primary peritonitis is less common and involves a spontaneous infection of preexisting ascites in the absence of any obvious intra-abdominal source. [8], [9] The spectrum of bacteria causing this syndrome and the patient population primarily affected have changed. Spontaneous bacterial peritonitis is now more common in adults than in children and shows no differential gender incidence. The group most commonly affected, formerly children with nephrosis, is now adults with cirrhosis. Whereas gram-positive organisms formerly caused most of these infections, gram-negative enteric bacteria now account for 60% to 80% of organisms in ascitic fluid cultures, with *Escherichia coli* and *Klebsiella pneumoniae* the most common species. Primary peritonitis is discussed in Chapter 46.

Secondary peritonitis is caused by diseases of or injury to the intra-abdominal organs. Acute suppurative peritonitis, granulomatous peritonitis, and chemical (aseptic) peritonitis belong to this category and are discussed in this section.

Acute Suppurative Peritonitis

Etiology. Spillage of intestinal contents into the peritoneal cavity as a result of primary intra-abdominal disease (e.g., perforated peptic ulcer, appendicitis, diverticulitis, perforated carcinoma), penetrating trauma, or iatrogenic perforation after instrumentation or radiologic procedures is the usual cause of acute suppurative peritonitis. [10], [11]

Clinical manifestations. Abdominal pain, which may be sudden in onset if associated with a perforated viscus, is the predominant symptom. [12] Characteristically, patients with peritonitis lie supine with knees flexed and with frequent and limited intercostal respirations. Examination of the abdomen of a patient with generalized peritonitis reveals distention and bowel sounds that are either hypoactive or absent. Of the various physical findings of peritonitis, tenderness and muscle guarding are the most important. Tenderness is often maximal over the organ in which the process originated; however, as the inflammation spreads, tenderness may be present over the entire extent of the peritoneum. Rebound tenderness, defined as the sudden and severe pain produced with the release of the hand after deep abdominal palpation, is often described as a useful adjunct in the diagnosis of the acute abdomen. This test, however, elicits no more information than that obtained from a careful and gentle examination of the abdomen and, furthermore, may cause unexpected and undue pain to the patient. Symptoms associated with acute suppurative peritonitis include anorexia, nausea that is often accompanied by vomiting, fever (38 to 40°C), and signs of hypovolemia (e.g., tachycardia, dry mucous membranes, hypotension). Typical diagnostic findings in patients with acute peritonitis include leukocytosis, often with an increased number of bands in the differential count. Roentgenographic findings are usually nonspecific, with evidence of a paralytic ileus of both the small bowel and colon in some patients. Free air under the diaphragm may be seen in an upright chest radiograph if a ruptured viscus is the cause (Fig. 123-1). Generally, the diagnosis of acute suppurative peritonitis is based on clinical manifestations and a thorough physical examination; laboratory and radiologic procedures serve mainly a confirmatory role.

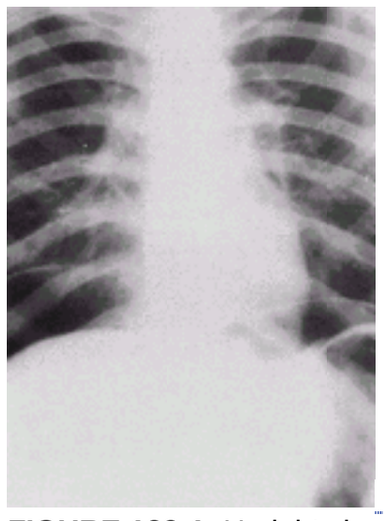

FIGURE 123-1. Upright chest film demonstrating free air under both diaphragms after perforation of a duodenal ulcer. (Courtesy of Charles J. Fagan, M.D., Galveston, TX.)

Clinical course and complications. Usually, suppurative peritonitis has an abrupt onset and a relatively short course with a rapid progression. Mortality results from fluid shifts and systemic endotoxin, which may cause hypovolemia and septic shock. Early diagnosis with prompt surgical intervention and aggressive preoperative and postoperative management is essential to reduce the morbidity and mortality from multiple organ system failure resulting from untreated peritonitis. [13], [14], [15] and [16]

Treatment. The hallmarks of treatment include resuscitation with intravenous fluids to restore normal physiology, broad-spectrum antibiotics, and operative management to control the source of peritoneal contamination and irrigate the peritoneal cavity (Table 123-1).

Restore normal physiology
Fluid resuscitation
Correct electrolyte imbalances
Supplemental O_2
Monitoring (e.g., vital signs, urine output, central venous pressure, pulmonary capillary wedge pressure)
Broad-spectrum antibiotics
Operative intervention
Control source of infection
Irrigate peritoneal cavity
Planned reoperations if source of infection not controlled
Investigational adjuvant therapies
Immunostimulation (e.g., administration of biologic response modifiers, vaccination)
Immunorepletion (e.g., administration of antiendotoxin antibody)
Immunoabrogation (e.g., cytokine antagonism, inhibition of prostaglandin synthesis)

TABLE 123-1 Treatment of Acute Suppurative Peritonitis

Resuscitation involves aggressive administration of isotonic crystalloid fluids (e.g., Ringer lactate) to correct hypovolemia and electrolyte imbalances. Careful monitoring of the response to rapid fluid resuscitation is necessary, particularly if large fluid volumes are required or the patient has impaired cardiac reserve. Monitoring usually entails frequent assessment of vital signs, hourly measurement of urine output, and frequent determination of central venous pressure or, preferably, placement of a Swan-Ganz catheter for pulmonary capillary wedge pressure and pulmonary artery pressure determinations. Oxygen may be administered to overcome the mild hypoxemia that is commonly present, and nasogastric intubation is required to decompress the stomach. Antibiotics play an important, albeit adjuvant, role in the management of patients with peritonitis. To be effective in the treatment of peritonitis, antibiotic therapy must be initiated before and continued during and after surgical therapy. In addition, broad-spectrum antimicrobial therapy should be directed toward both aerobic and anaerobic pathogens. An aminoglycoside (e.g., gentamicin, tobramycin, amikacin), in combination with either clindamycin or metronidazole, provides coverage for most aerobic and anaerobic organisms encountered in the gastrointestinal tract. [17], [18], [19] and [20] Because of concerns about toxic effects, most notably nephrotoxicity, third-generation cephalosporins, newer broad-spectrum penicillins, and a new class of ß-lactam antibiotics, called *carbapenems,* are being evaluated as single agents for the treatment of bacterial peritonitis. Prospective randomized clinical trials indicate that these single agents may be as effective as the combination of an aminoglycoside and clindamycin. [21], [22] Expeditious surgical intervention remains the mainstay of treatment. [23] After an adequate period to ensure resuscitation, laparotomy should be performed to irrigate the peritoneal cavity copiously and to repair the rupture. Aerobic and anaerobic cultures should be obtained to ensure appropriate antibiotic coverage. If the source of infection is not eliminated, the debridement is incomplete, or definitive abdominal closure cannot be accomplished, staged laparotomies may be required for re-debridement, removal of abdominal packs, and repeat irrigation until the source of infection is controlled and abdominal closure obtained. [18] In this situation, the placement of a large abdominal zipper can greatly facilitate abdominal opening and closing. Other procedures that have been described, but found not beneficial, include continuous postoperative irrigation with Tenckhoff catheters and radical peritoneal debridement. Recently, laparoscopy has been used as both a diagnostic and therapeutic modality in selected patients with peritonitis. [24] The future role and possible advantages of laparoscopy compared with a formal laparotomy remain to be defined. Novel agents directed at immunomodulation are under investigation and may become clinically useful in the treatment of sepsis. One investigational therapy attempted to neutralize the deleterious effects of lipopolysaccharides (LPS) released from the bacterial wall using either monoclonal or polyclonal antibodies directed to different regions of the LPS molecule. LPS, one of the most potent bacterial toxins, is an important trigger of the cytokine cascade. The acute exaggerated release of cytokines (e.g., tumor necrosis factor, interleukins, interferons) may produce many of the hemodynamic manifestations of septic shock; chronic production leads to tissue wasting and cachexia. Two anti-LPS monoclonal antibodies are being studied: HA-1A, a human monoclonal antibody directed against the lipid A domain of LPS; and E5, a murine monoclonal immunoglobulin M antibody. [25], [26] Initial prospective randomized trials suggest some efficacy with this treatment. Other areas of active research include strategies to neutralize the effects of cytokines using either neutralizing antibodies or specific cytokine receptor antagonists. [27], [28] A receptor antagonist to interleukin-1 (IL-1ra) has been developed and has proved useful in experimental models of shock and cachexia. [29], [30] Finally, responses elicited by cytokines appear to be mediated by secondary agents (e.g., eicosanoids, nitric oxide) acting at the effector sites. A competitive inhibitor of nitric oxide, N^G-methyl-L-arginine, protected against the hypotensive effects of tumor necrosis factor in a canine model, which suggests that the effect of this cytokine on hemodynamic stability may be mediated through nitric oxide and further suggests a potential clinical use of nitric oxide inhibitors in the treatment of sepsis. [31]

Granulomatous Peritonitis

Etiology. Granulomatous peritonitis is a disease process characterized by peritoneal inflammation that is associated with formation of granulomata and an increased incidence of adhesions. The disease most commonly associated with granulomatous peritonitis is tuberculosis. [32], [33] Other, less common causes include fungal (e.g., *Candida* species, histoplasma), amebic, and parasitic infections. [34], [35] and [36] Iatrogenic causes of granulomatous peritonitis also occur, usually related to the presence of glove lubricants (e.g., talc, cornstarch) or cellulose fibers from gauze, surgical drapes, or gowns within the peritoneum. [37]

Tuberculous Peritonitis

Epidemiology. After years of a decreasing incidence of tuberculosis in the United States, there has been a general reemergence of this disease secondary to an increase in the cases of acquired immunodeficiency syndrome (AIDS) and an increase in the number of immigrants. [38], [39], [40], [41], [42] and [43] Since 1985, cases of tuberculosis have increased 18% nationwide, with the highest increases occurring in certain highly populated areas of the country. Particularly disturbing is the recent appearance of multidrug-resistant strains of tuberculosis. [43], [44] and [45] In addition to patients with AIDS, other groups at high risk include poorly nourished, debilitated patients and those with cirrhosis.

Etiology. Tuberculous peritonitis is a form of abdominal tuberculosis that can involve the omentum, intestinal tract, liver, spleen, or female genital tract in addition to the parietal and visceral peritoneum. The overall incidence of abdominal tuberculosis in the United States is about 0.5% to 1% of all cases of tuberculosis. [32], [33] Tuberculous peritonitis usually is associated with a primary focus of tuberculosis elsewhere. This primary focus is usually the lung; however, only about one third of cases have clinical or radiographic evidence of pulmonary tuberculosis. The pathologic organism, *Mycobacterium tuberculosis,* can gain entry to the peritoneal cavity by one of three mechanisms: transmurally from diseased bowel, from tuberculous salpingitis, or, more commonly, by hematogenous spread from a pulmonary focus. [32], [33]

Clinical manifestations. Generally, the onset is insidious, with more than 70% of patients having had symptoms for more than 4 months before definitive diagnosis. The most common symptoms are constitutional and include fever, anorexia, weakness, malaise, and weight loss. Abdominal distention caused either by ascites or by partial obstruction may be present. On examination, the abdomen is diffusely tender in most patients; however, the classic doughy abdomen is rarely found. Tuberculous peritonitis should be suspected in high-risk or immunocompromised patients with ascites, fever, unexplained generalized symptoms, and diffuse abdominal pain or tenderness. [32], [33]

Differential diagnosis and diagnostic studies. Routine laboratory and radiographic studies are rarely diagnostic. A normal leukocyte count is present in most

patients, and anemia is found only occasionally. Tuberculin skin tests are usually positive in patients with tuberculous peritonitis; however, a negative result is of no help in excluding the disease. Chest radiographs are abnormal in 80% of patients and include findings of pleural effusions or pulmonary infiltrates. Radiographs of the abdomen are seldom of benefit; however, a computed tomography (CT) scan may be useful in identifying thickened bowel and ascites (Fig. 123-2).

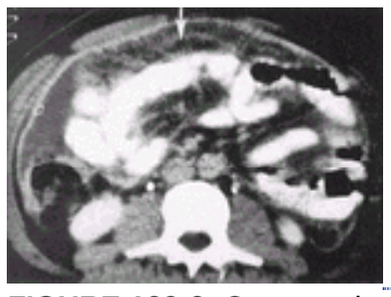

FIGURE 123-2. Computed tomography scan demonstrating ascites (*open arrow*) and small bowel thickening (*closed arrow*) in a patient with tuberculous peritonitis. (Courtesy of Charles J. Fagan, M.D., Galveston, TX.)

Examination of the peritoneal fluid may prove useful (see Chapter 46). In patients with tuberculous peritonitis, the protein content characteristically exceeds 3 g/dL, and glucose concentration is below 30 mg/dL in more than 80%. Most patients have a leukocyte count higher than 250 cells/mm ^{3} and a relative lymphocytic pleocytosis. Acid-fast stains are rarely positive unless at least 1 L of ascitic fluid is obtained, concentrated by centrifugation, and then examined. To compound the diagnostic problem, bacterial cultures of *M tuberculosis* often require 4 to 6 weeks of incubation. For these reasons, much attention has been given to the development of a rapid diagnostic test for tuberculosis. The most promising approach involves amplification and detection using the polymerase chain reaction procedure. Although not in widespread use, this procedure requires about 24 to 48 hours and can detect as few as 10 to 100 bacilli in a sample. [46], [47] The diagnosis of tuberculous peritonitis often is suggested by findings at laparoscopy or laparotomy. Tuberculous peritonitis is characterized by stalactite-like fibrinous masses from the parietal peritoneum; in addition, the peritoneum may be studded with small granulomata. The differential diagnosis of tuberculous peritonitis is variable, depending on the acuteness of the symptoms. In patients with a prolonged history, tuberculous peritonitis most commonly is confused with Crohn's disease or carcinoma. In patients presenting acutely, the differential diagnosis must include such entities as acute appendicitis, cholecystitis, perforated ulcer, and salpingitis. In summary, the diagnosis of tuberculous peritonitis is often difficult to make and requires an initial suspicion based on the clinical presentation (i.e., fever, anorexia, and abdominal pain) in combination with findings of a nonexudative ascites. It is crucial that a suspected diagnosis be confirmed by open or laparoscopy-assisted biopsy before committing the patient to a long-term treatment regimen with multiagent therapy.

Clinical course and therapy. Before the advent of chemotherapy, the mortality rate from tuberculous peritonitis was as high as 60%; now the disease is, for the most part, readily curable with the available agents. Therapy with isoniazid in combination with one or two additional drugs for 18 to 24 months is the treatment of choice. Although not proven experimentally, corticosteroids for 2 to 3 months may be beneficial in preventing the formation of dense fibrous adhesions, which could lead to bowel obstruction. The most worrisome feature of the resurgence of tuberculosis has been the recent outbreaks of multidrug-resistant isolates that often fail to respond to both isoniazid and rifampicin, the two cornerstone antituberculosis drugs. [38], [39], [40], [41], [42], [43], [44] and [45]

Chemical (Aseptic) Peritonitis

Etiology. Peritoneal inflammation can result from spillage of irritant materials that are initially sterile; with time, secondary bacterial contamination occurs with signs and symptoms similar to those of acute suppurative peritonitis. Most of the substances that cause chemical peritonitis are capable of acting as adjuvants to promote the growth of bacterial contaminants. These agents include bile, usually resulting from open or laparoscopic biliary tract operations without adequate external drainage, from inadvertent injury to the biliary tract, or from external trauma [48], [49] and [50]; urine, from intraperitoneal bladder rupture [51]; and chyle, which occurs secondary to injuries of large lymphatic vessels during operations on retroperitoneal organs. [52] In addition, spillage of barium sulfate, secondary to perforation of the gastrointestinal tract from diagnostic procedures, results in a severe peritoneal irritation, and, in combination with accompanying enteric bacteria, poses a lethal threat to the patient. [53], [54]

Treatment. The same principles that apply to the management of acute suppurative peritonitis also apply to the treatment of chemical peritonitis. Adequate intravenous fluids to replace the peritoneal fluid sequestration and antibiotics are followed by laparotomy to irrigate the abdomen and control the source of the peritoneal contamination.

Peritonitis as a Complication of Chronic Peritoneal Dialysis

Etiology. Continuous ambulatory peritoneal dialysis (CAPD) has gained increasing acceptance as an efficacious and cost-effective alternative to chronic hemodialysis in patients with end-stage renal disease. It is estimated that, in the United States alone, about one fourth of the more than 100,000 dialysis patients use CAPD. Despite its obvious advantages, the most common complication of CAPD is infectious peritonitis resulting from bacterial contamination of the peritoneal cavity. [55], [56] and [57] Even with better patient education regarding sterile techniques and proper cleaning around the catheter site, peritonitis in this group of patients is the major source of morbidity and the largest single cause of patient failure on CAPD. Although the incidence of peritonitis varies with the institution, peritonitis as a complication of CAPD averages 1.4 episodes per patient-year of treatment. [55], [56] and [57]

Epidemiology. The most frequent portals of entry for bacteria in CAPD patients include the exit site of the dialysis catheter and the tunnel through which the catheter traverses the abdominal wall. This usually occurs as a result in a break in aseptic technique with secondary contamination. Other possibilities for contamination include hematogenous or lymphatic dissemination from a septic focus originating from the patient. In contrast to other types of peritonitis, CAPD-related peritonitis usually is caused by a single organism, in most cases by gram-positive cocci (*Staphylococcus epidermidis, Staphylococcus aureus, Streptococcus* and *Enterococcus* organisms). [55], [56], [57], [58] and [59] Gram-negative bacteria account for about 25% of all cases of CAPD peritonitis, with the most common being *E coli* and *Pseudomonas aeruginosa*. The presence of anaerobic organisms or a mixed flora should raise suspicion of an intestinal perforation or other intra-abdominal disease as the cause of peritonitis. Fungal peritonitis is relatively uncommon but remains the most serious type of infective peritonitis in CAPD patients because of treatment difficulties. [60] At least 20 different fungal species have been cultured; however, *Candida albicans* and *Candida tropicalis* remain the most common.

Clinical manifestations. The severity of symptoms is dependent on the infecting organism; however, the signs and symptoms in most patients with CAPD peritonitis are clinically less severe than those in patients with suppurative peritonitis. Some cases may even be asymptomatic and detected only by the presence of a cloudy effluent. The predominant symptom found in patients is a diffuse abdominal pain, usually associated with a low-grade fever and leukocytosis. Other symptoms may include hyperhydration, diarrhea, hypotension, and pressure pain above the catheter tunnel. Characteristically, a turbid dialysis effluent is found in nearly all cases of CAPD peritonitis.

Diagnosis. The diagnosis of CAPD peritonitis is based on both clinical and laboratory findings. [55], [56] and [57], [61] The clinical diagnosis should be based on the presence of two of three criteria: abdominal pain or tenderness, a turbid dialysate containing more than 100 neutrophils/mm ^{3}, and a positive culture from the peritoneal fluid. Even if the patient's history and examination are consistent with the diagnosis of peritonitis, it is essential that the dialysate be evaluated to confirm the diagnosis and to identify the causative organisms. In some centers, the use of bacteria-concentrating techniques yielded positive cultures in more than 90% of cases.

Therapy and clinical course. Of patients with CAPD peritonitis, 70% to 80% can be treated successfully on an outpatient basis without hospitalization or interruption of dialysis. [55], [56], [57], [58], [59] and [60], [62], [63] CAPD peritonitis is treated most effectively by intraperitoneal administration of antibiotics. The initial choice of antibiotics should be broad enough to cover the most common pathogens causing CAPD peritonitis and then changed according to susceptibility testing. Vancomycin, cephalosporins, and aminoglycosides are the agents most commonly used to treat CAPD-associated peritonitis. In addition to antibiotics, heparin also is added to the dialysis bag to reduce fibrin formation and, thereby, the incidence of postinfective adhesions. Indications for catheter removal include persistence of peritonitis after 4 to 5 days of treatment and the presence of fungal or tuberculous peritonitis, fecal peritonitis, or severe skin infection at the catheter site.

Peritonitis in the Immunocompromised Host

Etiology and diagnosis. The AIDS epidemic and increasing use of immunosuppressive therapy for cancer, autoimmune disease, and multiple organ transplantation procedures have combined to increase greatly the incidence of opportunistic infections of the gastrointestinal tract. [64], [65] The differential diagnosis of abdominal pain in AIDS is quite broad and involves both AIDS-related and non–AIDS-related disorders [64], [65] and [66] (see Chapter 124). Perforation of the small bowel or colon secondary to cytomegalovirus enteritis is a frequent cause of peritonitis in AIDS patients. [67] Other organisms reported to cause peritonitis in these patients include *Mycobacterium avium–intracellulare, M tuberculosis, Cryptococcus neoformans*, *Strongyloides* organisms, and *Leishmania* organisms. [64], [65], [66], [67], [68], [69] and [70]

Clinical manifestations and treatment. Immunocompromised patients with peritonitis usually present with severe abdominal pain as the predominant symptom, like patients with suppurative peritonitis. Principles of management include emergent laparotomy and resection of the involved portion of bowel, intravenous fluid resuscitation, and broad-spectrum antibiotics.

Primary Mesothelioma

Incidence and Etiology Primary mesotheliomas, arising from mesenchymal and epithelial components, are uncommon. The reported incidence in the United States

is only 2.2 cases per million population; however, studies suggest that the incidence is increasing throughout the world and especially in industrialized countries. [71], [72], [73] and [74] Only 20% to 40% of mesotheliomas occur in the peritoneum, with most of these tumors originating from the pleura. Malignant mesothelioma is linked to exposure to asbestos, and there is a strong association with occupations that require workers to handle or work in proximity to asbestos, such as those in the textile, insulation, building, demolition, and shipyard industries. [73], [74], [75], [76] and [77] There is a latency period of 20 to 40 years from significant asbestos exposure to development of mesothelioma; two thirds of the patients are diagnosed at 45 to 64 years of age, and there appears to be a male predominance. Crocidolite is considered the most oncogenic type of asbestos, whereas the oncogenic potential of chrysotile is still debated. Although asbestos is the predominant cause of this disease, other etiologic agents may be associated with primary mesothelioma, including use of the angiographic contrast material Thorotrast (thorium dioxide), radiation exposure, and recurrent peritonitis.

Clinical Manifestations The clinical presentation is nonspecific, with the predominant complaint being abdominal pain present in the epigastrium or right upper quadrant. Other symptoms include nausea and vomiting, malaise, fever, weight loss, diarrhea, and anemia. Ascites is the most common physical finding and occurs in 90% of patients. Peritoneal mesotheliomas can produce and secrete a variety of ectopic hormones, including antidiuretic hormone, growth hormone, and insulin-like factors, which can produce associated paraneoplastic syndromes with symptoms of hypoglycemia, hyponatremia, thrombocytosis, and increased production of fibrin-degradation products. [71], [72], [73] and [74] Interestingly, peritoneal mesothelioma has been described in association with other synchronous neoplasms such as colorectal cancer.

Differential Diagnosis and Diagnostic Studies Malignant mesothelioma is a difficult malignancy to diagnose early. [71], [72], [73] and [74], [78] Mesotheliomas are generally firm, white tumors that present as individual nodules studding the peritoneal surface and tend to spread over the surface of the intra-abdominal organs, eventually encasing the viscera. Ultrasound and CT studies can suggest the diagnosis, demonstrate the extent of tumor, and also aid in directed biopsy of suspicious lesions (Fig. 123-3); however, in most cases, the diagnosis is confirmed after diagnostic laparoscopy or laparotomy.

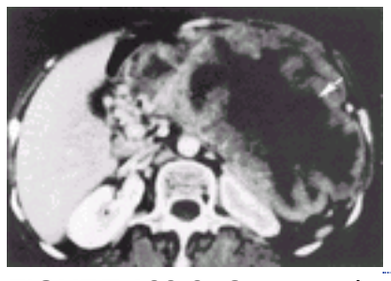

FIGURE 123-3. Computed tomography scan demonstrating diffuse mesenteric and peritoneal involvement of a soft tissue mass (*arrow*) causing displacement of intra-abdominal organs in a patient with peritoneal mesothelioma. (Courtesy of Charles J. Fagan, M.D., Galveston, TX.)

Microscopic and immunohistochemical studies are available to aid in the diagnosis. [79], [80] Immunohistochemistry is useful for two reasons: (1) to determine whether the mesothelial cells present are benign or malignant, and (2) to distinguish mesothelioma from an adenocarcinoma. The typical analysis of a suspected mesothelioma should include electron microscopy, periodic acid-Schiff (PAS) stain, Alcian blue stain to detect hyaluronic acid, and an anticarcinoembryonic antigen (anti-CEA) antibody stain. Ultrastructural features on electron microscopy include long, slender microvilli and diffuse cytoplasmic or perinuclear distribution of tonofilaments. Mesotheliomas lack intracellular mucin on PAS stains, should produce negative results on anti-CEA antibody stain, and often stain positively for hyaluronic acid. In contrast, adenocarcinomas often stain positively for mucin and CEA and do not contain hyaluronic acid. Well-characterized monoclonal and polyclonal antibodies hold great promise for improving the diagnostic accuracy. [81] Recent studies indicate that calretinin, a member of the family of calcium-binding proteins, and cytokeratin 5/6 stains appear to be the most sensitive and specific positive members for distinguishing between malignant epithelial mesotheliomas (positive staining) and metastatic adenocarcinomas (negative staining). [80] Among the antibodies considered negative members for mesothelioma, MOC-31, CEA, and BG-8 appear to be the best diagnostic discriminators. Other useful negative markers include B72.3, Leu-M1, Ber-EP4, and thrombomodulin. Immunostaining for N-cadherin and E-cadherin appears to be promising in the diagnosis of mesothelioma. Another useful marker is epithelial membrane antigen, which has been shown to be of value in distinguishing between benign and malignant mesotheliomas. Additional molecular studies that may serve as useful adjuncts to the diagnosis include the identification of high levels of the platelet-derived growth factor (PDGF) in malignant mesotheliomas. [81] Less common primary peritoneal neoplasms that should be included in the differential diagnosis include benign papillary mesotheliomas, benign fibrous mesotheliomas, adenomatoid tumors, and multicystic peritoneal mesotheliomas. [73], [74]

Genetic Factors With the advances in molecular techniques and the ability to identify genetic abnormalities in various tumors, studies have focused specifically on potential genetic abnormalities in mesotheliomas. [71], [72], [75], [76] and [77], [82], [83] The accumulation of numerous chromosomal deletions in a number of mesotheliomas suggests a multistep process of tumorigenesis, characterized by the loss or inactivation of various tumor suppressor genes. Genetic abnormalities that have been identified include frequent deletions of specific sites within chromosome arms 1p, 3p, 6q, 9p, 13q, 15q, and 22q. [82], [83] Loss or mutation of the *p16 Ink4a* tumor suppressor gene at 9p21 has been noted with high frequency (about 85%) and may contribute to the pathogenesis of these tumors. In addition, another recurrent theme of genetic abnormalities in mesotheliomas is mutation or allelic loss of neurofibromatosis type 2 (*NF2*), a tumor suppressor gene, at 22q12. Recent studies have identified the presence and expression of simian virus 40 (SV40) in a number of mesotheliomas. [84] SV40 large T antigen has been shown to inactivate the tumor suppressor gene products *Rb* and *p53,* suggesting the possibility that asbestos and SV40 could act as co-carcinogens in mesothelioma. Furthermore, circulating antibodies to SV40 antigens have been described, raising the possibility that the immune system may be modulated to treat patients with mesothelioma. Finally, expression of growth factors, including PDGF-A and -B, insulin-like growth factors I and II, and transforming growth factor-a and -ß, have been identified in these tumors. A possible paracrine and autocrine role for PDGF has been postulated because the PDGF receptor is expressed in a number of mesotheliomas. Continued studies are required to determine whether these genetic abnormalities play a role in the pathogenesis and progression of the mesothelioma tumors and may serve as potential targets for novel treatment strategies.

Treatment and Prognosis Therapy for mesothelioma has proved disappointing, mainly as a result of the advanced stage of the disease at diagnosis and the local aggressiveness of the tumor. [85], [86] Rarely can surgery be performed for a curative resection; rather, laparotomy serves to establish a tissue diagnosis and to perform palliative procedures such as bypassing obstructive lesions or tumor debulking. Chemotherapy offers little improvement in overall patient survival. Doxorubicin, alone or in combination with other antineoplastic agents, achieves the best, albeit minimal, response. Studies suggest that surgical cytoreduction and multimodality therapy, especially with paclitaxel and cisplatin chemotherapy, may confer a survival advantage. In addition, there may be an advantage in using hyperthermic intraperitoneal infusion of the chemotherapeutic agent in patients with primary peritoneal mesotheliomas. [85], [87] Radiation therapy has been used in combination with intracavitary instillation of various chemotherapeutic agents or with systemic chemotherapy, but with only minimal success. Future directions for the management of mesothelioma include the use of interleukin-2, interferon-?, tumor necrosis factor-a, monoclonal antibodies, and cancer vaccines, which may prove beneficial when used in combination with other therapies. The results of recent trials suggest potential benefits of chemo-immunotherapy for the treatment of mesothelioma. Novel treatment strategies currently under investigation include the introduction of the herpes simplex virus thymidine kinase gene into the mesothelioma, so that growth is inhibited after administering ganciclovir, and the transfection of antisense oligonucleotides to the growth factor PDGF-A or -B. [71], [72], [88] Multiple obstacles, including transfection efficiency and proper vector construction, must be overcome before these types of gene therapy protocols will be clinically beneficial. The prognosis for mesothelioma of the peritoneum is poor. Death usually occurs within 12 to 14 months of the diagnosis from progressive gastrointestinal tract obstruction and debility.

Pseudomyxoma Peritonei

Incidence and Etiology Pseudomyxoma peritonei is a rare condition manifested by diffuse, gelatinous implants of the peritoneal cavity and omentum arising from mucinous neoplasms of either the appendix or ovary. The reported incidence of pseudomyxoma peritonei is about 2 in 10,000 laparotomies. Most female patients with pseudomyxoma peritonei have disease in both the appendix and ovary; therefore, there is no general agreement regarding the tissue of origin. Recent applications of immunocytochemistry and genetic analysis by polymerase chain reaction strongly suggest that most cases are due to metastases from appendiceal neoplasms. [89], [90]

Clinical Features and Diagnosis Women between 45 and 55 years of age make up about 75% of patients with pseudomyxoma peritonei. This is usually an unexpected diagnosis made at laparotomy for a preoperative diagnosis of appendicitis or ovarian tumor. Patients can present with an increasing abdominal girth secondary to mucinous ascites or intestinal obstruction, abdominal pain, or a mass in the abdomen. [89], [91], [92] Histologically, pseudomyxoma peritonei is characterized by a benign appearance with simple columnar epithelium containing mucin-filled vacuoles. [93], [94] Certain features noted on ultrasonography or CT scan are characteristic for this disease. Ultrasonographic findings include multiple intraperitoneal multilocular cysts and ascitic septation. A characteristic CT finding of pseudomyxoma peritonei is “scalloping” of the hepatic and bowel margins secondary to extrinsic compression by ascitic spaces containing gelatinous material (Fig. 123-4). The role of magnetic resonance imaging (MRI) in diagnosing pseudomyxoma remains to be defined. T2-weighted images accentuate differences between mucinous and fluid ascites; however, a major disadvantage of MRI is the added expense compared with CT scan.

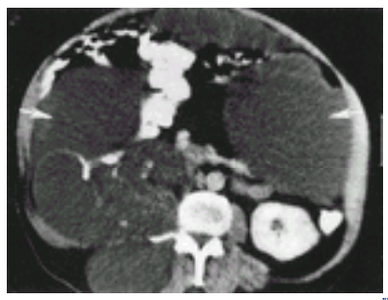

FIGURE 123-4. Computed tomography scan demonstrating large gelatinous masses (*arrows*) in a patient with pseudomyxoma peritonei. (Courtesy of Charles J. Fagan, M.D., Galveston, TX.)

Treatment and Prognosis Pseudomyxoma peritonei is a low-grade malignancy that rarely metastasizes or invades contiguous viscera. The treatment of pseudomyxoma peritonei is primarily surgical, with aggressive debulking of all intra-abdominal tumor and omentectomy. [95], [96] In addition, because of the usual involvement of both the appendix and the ovaries, it is recommended that appendectomy and bilateral oophorectomy be performed in all patients. Recurrence occurs in about 75% of patients; repeat laparotomies are indicated for recurrent disease. Recently, new strategies in the treatment of this disease have included a chemotherapy regimen with surgical treatment. [95], [97], [98] These reports suggest a survival advantage using early postoperative chemotherapy (intraperitoneal or systemic) combined with cytoreductive surgical treatment, compared with repeated surgical procedures in the absence of chemotherapy. Radiation treatment, mucolytic agents, and phototherapy have been described but appear to be of little benefit. [89] Long-term survival is about 54% at 5 years, with death occurring usually as a result of extensive peritoneal disease and intestinal obstruction.

RETROPERITONEUM

The retroperitoneum is the space behind the abdominal cavity extending from the posterior peritoneum and the mesenteries to the spinal column and the psoas, quadratus lumborum, and transversus abdominis muscles. It extends superiorly from the diaphragm and inferiorly to the levator muscles of the pelvis. The retroperitoneum is divided into the anterior compartment, which contains the pancreas, duodenal loop, and ascending and descending colon, and the posterior compartment, which contains the perinephric space with the kidneys, ureters, and adrenal glands and an extensive network of vessels, lymphatics, and neural structures. The space also contains fatty and areolar tissue and connective tissue.

Modern diagnostic techniques, such as ultrasonography, CT scan, MRI, and arteriography, have greatly enhanced our ability to diagnose and treat pathologic processes in the retroperitoneum; however, this region remains elusive. Early clinical manifestations are nonspecific, thus allowing pathologic processes to become relatively advanced before producing symptoms. Retroperitoneal disease processes often present as a mass. (The differential diagnosis of retroperitoneal masses and other disease processes is shown in Table 123-2.) This section specifically discusses retroperitoneal hemorrhage and infections, idiopathic retroperitoneal fibrosis, and primary neoplasms of the retroperitoneal space. In addition to these clinical entities, fluid collections consisting of lymph, gastrointestinal secretions (e.g., duodenal, pancreatic), bile, or urine also may collect in this space, usually secondary to iatrogenic or traumatic injuries. [99], [100], [101] and [102] A CT scan or ultrasound may be a useful diagnostic imaging study; percutaneous drainage with fluid analysis may aid in further establishing the diagnosis and, in certain instances, may be useful in the treatment.

Fluid collections
Lymph
Duodenal succus
Pancreatic juice
Bile
Urine
Hematoma
Spontaneous hemorrhage
Traumatic hemorrhage
Fat necrosis
Acute necrotizing pancreatitis
Abscess
Primary
Secondary: extension from abdomen, kidney, vertebra
Neoplasm
Benign
Cyst
Benign soft tissue tumor
Malignant
Sarcoma
Germ cell–derived neoplasm
Lymphoma
Lymph node metastases
Aortic aneurysm
Retroperitoneal fibrosis

TABLE 123-2 Differential Diagnosis of Retroperitoneal Masses and Disease Processes

Retroperitoneal Hemorrhage

The most common etiology for retroperitoneal bleeding and hematoma is secondary to traumatic injuries associated with pelvic or vertebral fractures, avulsion of the vascular pedicle of the kidney, or penetrating injuries. [102], [103], [104], [105] and [106] Other causes for hemorrhage may be secondary to anticoagulation therapy, acute pancreatitis, spontaneous hemorrhage into an adrenal gland or retroperitoneal tumor, ruptured aortic aneurysm, and ruptured ureteral or ovarian veins during pregnancy. In addition, massive retroperitoneal hemorrhage has been associated with femoral vein catheterization and liposuction. [107], [108]

If hemorrhage occurs quickly and in significant amounts, signs of hypovolemic shock may be present. Signs of retroperitoneal hemorrhage that may occur late include pain felt most in the abdomen and back and ecchymosis of the flank (Grey Turner sign). Laboratory examination may demonstrate a low hematocrit. Evaluation of the retroperitoneal hemorrhage may include plain films of the abdomen, which can show obliteration of the psoas shadow. A CT scan may be useful in the stable patient to evaluate the possible etiology and extent of the hemorrhage (Fig. 123-5).

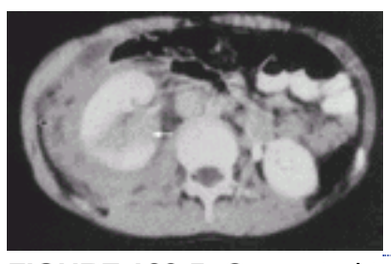

FIGURE 123-5. Computed tomography scan demonstrating a large retroperitoneal hematoma (*open arrow*) due to blunt injury to the kidney (*closed arrow*). (Courtesy of Melvyn H. Schreiber, M.D., Galveston, TX.)

Treatment of retroperitoneal hemorrhage is secondary to the specific etiology. For example, retroperitoneal hemorrhage secondary to a pelvic fracture may be treated by fracture stabilization or arteriography and embolization of the bleeding vessel. Vascular injuries or a ruptured abdominal aortic aneurysm require emergent surgical intervention and control of the ongoing hemorrhage.

Retroperitoneal Infections

Abscesses in the retroperitoneal space are normally secondary to diseases of surrounding abdominal organs, the urinary tract, or the vertebral column. [109], [110], [111] and [112] Most commonly, the abscesses result from perforated diverticular disease or colonic cancer, appendicitis, acute pancreatitis, penetrating duodenal ulcer, or biliary tract disease. A psoas abscess may occur secondary to intra-abdominal or vertebral infections; extension from Crohn's disease, appendicitis, colon diverticulitis, and cancer are the most frequent causes. Primary bacterial infections in the retroperitoneal space can occur but are rare. Except for the Gerota fascia, there are no anatomic boundaries to the retroperitoneal space; therefore, abscesses can be large, extend bilaterally, and present late in their course. As a result, retroperitoneal abscesses may be associated with higher mortality rates.

Clinical manifestations include fever, chills, anorexia, and weakness; pain in the lower back and abdomen, thigh, or flank is present in about 60% of the patients. A palpable mass may be present on physical examination. Leukocytosis is often noted as the most common laboratory abnormality. The CT scan is the most useful test to establish the diagnosis (Fig. 123-6); CT-guided percutaneous needle aspiration of the abscess may allow for bacteriologic examination, so that an appropriate antibiotic treatment regimen can be chosen based on the specific organisms and sensitivities. Depending on the etiology, percutaneous drainage may be effective treatment for some of these collections; surgical drainage is required secondary to inadequate fluid evacuation. Coliform organisms and *S aureus* are the most common organisms encountered. Other organisms can occur rarely and include tuberculosis, which most commonly develops in the urinary tract or spine; opportunistic fungi (e.g., *Candida, Aspergillus, Cryptococcus* organisms) may spread to the retroperitoneum, particularly in patients who are immunocompromised. Actinomycosis or retroperitoneal infections with *Nocardia* organisms can be the result of secondary extension from a primary focus. For example, actinomycosis may be secondary to perforation of the appendix or colon and nocardiosis associated with a pulmonary infection.

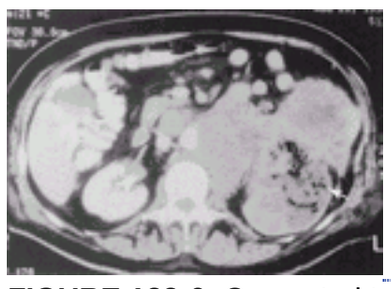

FIGURE 123-6. Computed tomography scan demonstrating a retroperitoneal infection and abscess (*arrow*) with air bubbles present originating from infection of the left kidney. (Courtesy of Melvyn H. Scheiber, M.D., Galveston, TX.)

Retroperitoneal Fibrosis

Etiology and Epidemiology Retroperitoneal fibrosis is an uncommon disease process characterized by progressive nonspecific inflammation and fibrosis of connective and adipose tissue in the retroperitoneal space. [113], [114], [115] and [116] This process originates just below the level of the aortic bifurcation near the sacral promontory and then spreads throughout the retroperitoneum with extension bilaterally along the aorta and inferior vena cava. Symptoms are the result of gradual compression of the tubular structures in the retroperitoneal space, particularly the ureter. Retroperitoneal fibrosis was first described by the French urologist Albarran in 1905. [113] The initial description of retroperitoneal fibrosis in the English literature is credited to Ormond in 1948. [114] In fact, idiopathic retroperitoneal fibrosis is often referred to as *Ormond disease*. Retroperitoneal fibrosis is relatively uncommon, with an instance of 1 in 200,000. There appears to be an overall male predominance of 2:1 or 3:1, with presentation usually in the fifth and sixth decades of life. The etiology of this disease is, for the most part, not known. A number of factors have been postulated to contribute to this disease process, including extravasation of urine, surgery, and nonspecific gastrointestinal inflammatory diseases, including Crohn's disease; however, most cases of retroperitoneal fibrosis are considered idiopathic (Table 123-3). Various medications have been associated with retroperitoneal fibrosis as a side effect. Methysergide, an antiserotonin drug, is the best described agent in which the occurrence of retroperitoneal fibrosis has been clearly linked. [117] Other drugs implicated in this fibrotic process include ergotamine, pergolide, hydralazine, methyldopa, and, most recently, ß-adrenergic blocking agents such as metoprolol. [118] An immune etiology for retroperitoneal fibrosis has been suggested, particularly because an association with other immune-mediated connective tissue disorders, such as systemic lupus erythematosus, Raynaud disease, systemic vasculitis, and ankylosing spondylitis, has been demonstrated in certain cases. [119] In addition, amelioration of symptoms can occur with corticosteroid treatment. In some patients, retroperitoneal fibrosis can occur secondary to severe retroperitoneal infection (e.g., pancreatitis) or inflammatory aortic aneurysms and in response to neoplasms, most commonly Hodgkin lymphoma, sarcomas, carcinoid tumors, and certain adenocarcinomas. [120], [121]

Primary
Idiopathic
In conjunction with other fibrotic processes (e.g., mediastinal fibrosis, mesenteric fibrosis, sclerosing cholangitis)
Drug-associated (e.g., methysergide, ergotamine, hydralazine, and β-adrenergic drugs)
Paraneoplastic (e.g., sarcoma, Hodgkin disease, carcinoid tumor)
Secondary
Radiotherapy
Retroperitoneal infection
Retroperitoneal fluid collection or hematoma
Inflammatory abdominal aortic aneurysm

TABLE 123-3 Etiologies of Retroperitoneal Fibrosis

Pathology Retroperitoneal fibrosis appears grossly as a woody, white, fibrous plaque that, as it progresses, surrounds the ureters and vascular structures in the retroperitoneum. [115], [116] Early microscopic features include active chronic inflammation occurring with abundant lymphocytes, plasma cells, and macrophages interspersed with fibroblasts and collagen bundles. [115], [116], [122] As the disease progresses, the tissues become relatively acellular with scattered calcifications, an abundant collagen deposition, and fibrous scarring.

Clinical Manifestations The symptoms of retroperitoneal fibrosis are generally nonspecific, the most common symptom being a dull, constant pain localized to the back or flank. [113], [114], [115] and [116] This pain may radiate to the lower abdomen, groin, or anteromedial aspect of the thigh. Other symptoms include anorexia, nausea, diarrhea, fever, and weight loss; patients may present with hypertension. Laboratory abnormalities may include leukocytosis, an elevated erythrocytic sedimentation rate, and elevated alkaline phosphatase levels. As the disease progresses, late complications of retroperitoneal fibrosis occur as the result of encasement and compression of the tubular retroperitoneal structures, most commonly involving the ureters, and eventually leading to azotemia, renal insufficiency, and possible renal failure. Lower extremity edema may occur from compression of lymphatics and venous obstruction. [123] The aorta can be involved, albeit uncommonly, with symptoms consistent with arterial insufficiency (e.g., claudication, rest pain). Rarely, this fibrotic process may involve the duodenum, common bile duct, or colon, resulting in obstruction. [115], [116], [124], [125]

Differential Diagnosis and Diagnostic Studies The differential diagnosis of retroperitoneal fibrosis includes retroperitoneal hemorrhage, primary neoplasms, and metastasis to the retroperitoneum. Imaging studies useful in establishing the diagnosis of retroperitoneal fibrosis include an intravenous pyelogram, in which a triad of classic findings have been described: delayed excretion of contrast material with hydronephrosis, narrowing of the ureters at about the L-4 to L-5 level, and a medial displacement of the ureters [115], [116] (Fig. 123-7). In fact, this medial deviation of the ureters was considered a pathognomonic sign of retroperitoneal fibrosis, in contrast to retroperitoneal neoplasms, in which deviation of the ureters normally occurs laterally. Recent studies, however, indicate that medial deviation is not a constant finding in patients with retroperitoneal fibrosis. The CT scan has emerged as the procedure of choice for the diagnosis and follow-up of patients with retroperitoneal fibrosis. [116], [126] Findings include a periaortic soft tissue mass of variable thickness that envelops the retroperitoneal structures, with obliteration of the fat plane between the mass and adjacent psoas musculature. The MRI also has been used to assess retroperitoneal fibrosis, particularly using T2-weighted images to delineate the stage of the disease more clearly [127], [128] and [129] (Fig. 123-8). The advantages of MRI compared with CT are a more exact anatomic definition because of multiplanar capability and the avoidance of nephrotoxic contrast agents.

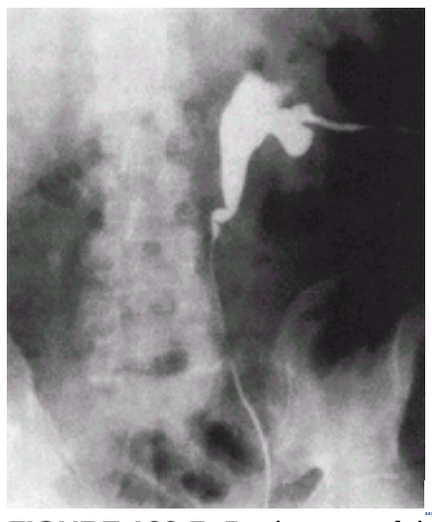

FIGURE 123-7. Pyelogram of the left kidney through a nephrostomy tube demonstrating marked hydronephrosis, narrowing of the ureter at the L4 level, and medial displacement of the ureter. (Courtesy of Melvyn H. Schreiber, M.D., Galveston, TX.)

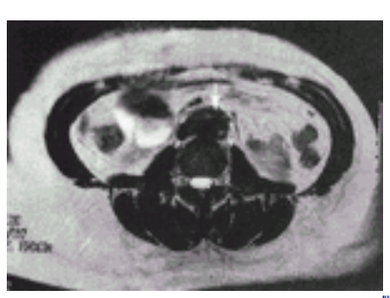

FIGURE 123-8. Axial T2-weighted magnetic resonance scan demonstrating a periaortic soft tissue mass (*arrow*) that obliterates the fat plane in a patient with retroperitoneal fibrosis. (Courtesy of Greg Chalchub, M.D., Galveston, TX.)

Treatment Goals of treatment include establishing the diagnosis, relieving the obstruction (usually involving the urinary tract), and preventing progression of the disease. Medications associated with retroperitoneal fibrosis should be discontinued. Surgical management is directed at ureterolysis for ureteral obstruction with intraperitoneal transposition of the ureters. Other maneuvers described to prevent recurrence of the ureteral obstruction include wrapping the ureters in omental fat or interposing retroperitoneal fat between the ureters and the fibrosis. [130], [131] The use of laparoscopic ureterolysis in selected cases has been described with good results at short-term follow-up. [132] Symptoms caused by venous obstruction are best treated nonoperatively, with elevation and elastic support of the leg eventually resulting in the development of collateral circulation. The medical management of retroperitoneal fibrosis includes corticosteroids, which are effective in the early, active inflammatory stage. Other immunosuppressive drugs, such as azathioprine and cyclophosphamide, also have been used with varying success. [133] A recent case report demonstrated remission of an advanced case of retroperitoneal fibrosis using a combination of prednisone and the new immunosuppressive agent mycophenolate mofetil, a selective inhibitor of T and B lymphocytes. [134] Recent studies suggest that tamoxifen, a nonsteroidal antiestrogen agent, may be effective in regression of the fibrotic process and amelioration of symptoms secondary to increased synthesis and secretion of transforming growth factor-ß, an inhibitory growth factor. [135] In patients with idiopathic retroperitoneal fibrosis without renal compromise and effective ureterolysis, the prognosis is excellent, with long-term success rates exceeding 90%. [115]

Retroperitoneal Neoplasms

Etiology and Pathology Tumors in the retroperitoneal space arise from mesodermal, neuroectodermal, or embryonic remnants, including fat, connective tissue, fascia, muscle, nervous tissue, lymph vessels, and lymph nodes. Malignant tumors are about four times more prevalent than benign neoplasms, with the most common neoplasms being malignant lymphomas or lymphosarcomas. [136], [137] Other malignant neoplasms in this area include other types of sarcomas, such as liposarcoma and leiomyosarcoma. [138], [139] and [140] Of the benign tumors, the most frequent are lipomas. Most patients presenting with primary retroperitoneal tumors are in the fifth or sixth decades of life, although about 15% of these tumors are found in children aged younger than 10 years. In addition to primary tumors of retroperitoneal structures, the retroperitoneum is also a common site for metastasis, primarily lymphatic metastasis from neoplasms of abdominal organs.

Clinical Manifestations Retroperitoneal tumors can grow to a large size before becoming symptomatic. [136], [137] Early symptoms are normally vague or lacking and may include fatigue, weakness, weight loss, abdominal discomfort, and a sense of fullness or abdominal heaviness. Nausea and vomiting and a change in bowel habits may occur with intestinal compression. Swelling of the lower extremities may occur secondary to obstruction of lymphatics or venous return. Larger tumors can cause pain radiating to both thighs as a result of involvement of the lumbar and sacral nerve roots. Rarely, patients may present with hypoglycemia. The most common physical finding is the presence of a nontender abdominal mass.

Differential Diagnosis and Diagnostic Studies Primary retroperitoneal tumors should be differentiated from other masses in this region, including renal lesions, pancreatic cysts and tumors, malignancies of the gastrointestinal tract, ovarian tumors, and cysts of the omentum and mesentery. The CT scan is a useful imaging modality to identify the mass, determine its size and origin, and assess its relationship, proximity, and invasion of surrounding structures [141] (Fig. 123-9). In addition, CT-guided needle biopsy often can provide a histological diagnosis preoperatively. MRI has become increasingly important as a diagnostic tool for retroperitoneal neoplasms. [129], [142] This study provides a better degree of definition between the mass, surrounding muscle groups, and vascular structures compared with CT (Fig. 123-10). Before surgery, angiography provides important information about the vasculature supplying the neoplasm. The main value of barium studies is to delineate primary intestinal malignancies from retroperitoneal tumors.

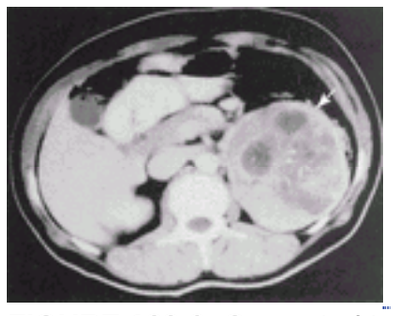

FIGURE 123-9. Computed tomography scan of a large retroperitoneal leiomyosarcoma (*arrow*). (Courtesy of Melvyn H. Schreiber, M.D., Galveston, TX.)

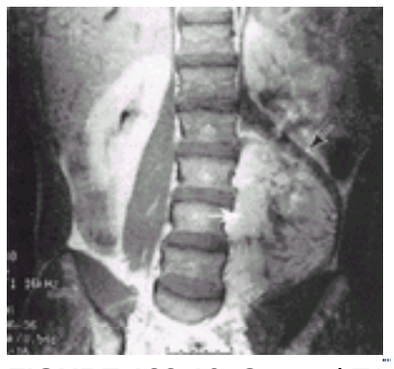

FIGURE 123-10. Coronal T1-weighted magnetic resonance scan after administration of the contrast medium gadolinium shows a large retroperitoneal sarcoma (*closed arrow*) with displacement of the left psoas muscle laterally (*open arrow*). (Courtesy of Greg Chalchub, M.D., Galveston, TX.)

Treatment Surgical excision offers the greatest prospect for cure and remains the best option for treatment of these retroperitoneal malignancies. [143], [144] Some retroperitoneal tumors are benign and are cured by complete excision. Treatment of malignant tumors involves surgical resection or radiation or a combination of the two modalities. With the exception of lymphomas, chemotherapy appears to offer little to the multimodality management of these tumors. It is important to remember that even though the overall prognosis for malignant retroperitoneal tumors is relatively low, en bloc resection of a primary malignancy can achieve 5-year survival rates of greater than 60%. Furthermore, debulking procedures can provide symptomatic relief and may offer a better response rate to radiation therapy in the postoperative period in patients with tumors that cannot be excised because of size or invasion of surrounding structures. Malignant tumors have a recurrence rate in the range of 30% to 50% over a 3-year period.

MESENTERY AND OMENTUM

During fetal development, the splanchnic mesoderm forms the mesentery covering the developing gut. [1] The greater omentum, which extends from the greater curvature of the stomach caudally to fuse with the transverse colon and mesocolon, develops from the dorsal mesogastrium. The omentum and mesentery are rich in lymphatics and blood vessels and provide a major source of macrophages and lymphocytes to aid in removal of foreign material or infection in the abdominal cavity. The small bowel mesentery contains the lymphatic and vascular network responsible for transporting all of the nutrients absorbed from the small bowel. Primary diseases of the mesentery and omentum are relatively uncommon. This section describes some of the diseases that primarily or prominently involve the omentum, mesentery, or both mesenchymal structures (Table 123-4).

Mesenteric Diseases
Primary mesenteric inflammatory diseases
Mesenteric panniculitis
Retractile mesenteritis
Mesenteric cysts
Embryonic and developmental cysts
Traumatic or acquired cysts
Neoplastic cysts
Infective and degenerative cysts
Mesenteric tumors
Benign tumors
Lipoma
Hemangioma
Leiomyoma
Ganglioneuroma
Malignant tumors
Leiomyosarcoma
Liposarcoma
Rhabdomyosarcoma
Metastatic disease
Mesenteric fibromatosis
Omental Diseases
Mass lesions
Primary tumors and cysts
Metastatic disease
Vascular lesions damaging blood supply
Torsion
Primary
Secondary: hernia, adhesion, tumor
Infarction
Primary
Secondary: torsion, incarceration in hernia

TABLE 123-4 Classification of Mesenteric and Omental Diseases

Mesenteric Panniculitis and Retractile Mesenteritis

Incidence and Etiology Mesenteric panniculitis (also called primary liposclerosis, lipogranuloma, isolated lipodystrophy, and mesenteric Weber-Christian disease) is a nonspecific, rare inflammatory process involving the adipose tissue of the mesentery. [145], [146] This process includes a spectrum of diseases, from inflammatory to fibrotic lesions, which have been called *mesenteric panniculitis* and *retractile mesenteritis,* respectively. Possible etiologies include trauma, autoimmunity, infection, ischemia, previous abdominal surgery, or abdominal malignancy, but in most cases, the precise etiology is not known.
Pathology Gross pathologic findings include a diffusely thickened and rubbery mesentery, usually as a solid mass in the root of the mesentery or as several adherent masses. Microscopic findings include excessive growth of normal fat, with subsequent degeneration, fat necrosis, and xanthogranulomatous inflammation with lipid-laden macrophages, infiltration with histiocytes, lymphocytes, and occasional foreign-body giant cells, leading to fibrotic scarring and calcification. [145]
Clinical Manifestations Patients with mesenteric panniculitis generally present in middle or late adulthood (average age, 53 years). There is a slight male predominance. When symptomatic, patients may present with cramping abdominal pain, which may be local or generalized; weight loss; nausea and vomiting; and low-grade fever. Patients may occasionally present with findings consistent with an acute abdomen. About 60% of patients present with a palpable abdominal mass noted by physical examination; the remainder of patients present with an incidental mass noted at examination or laparotomy. [145], [146] The differential diagnosis includes a wide range of mesenteric pathologies, such as mesenteric lymphoma, fat necrosis, primary or secondary mesenteric tumors, and mesenteric fibromatosis. In certain instances, an abdominal aortic aneurysm may be considered, owing to transmission of aortic pulsations to the anterior abdominal wall. [147] This diagnosis can be suspected by various radiographic studies. Plain radiographic findings include displacement of intestinal segments and extrinsic compression of the bowel. Findings by CT scan include fat density masses arising from the mesentery, surrounding mesenteric vessels, displacing bowel loops without evidence of invasion, a left-sided orientation, a hypodense fatty halo surrounding nodules and vessels, and a hyperattenuated stripe [148] (Fig. 123-11). Ultrasonography has also been successfully used as a diagnostic tool in these patients. [149]

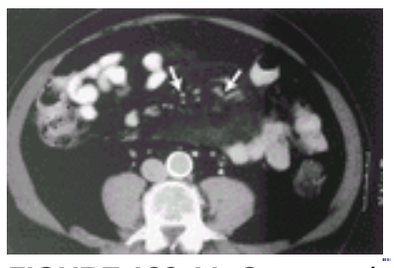

FIGURE 123-11. Computed tomography scan of mesenteric panniculitis demonstrating a fat density mass surrounding mesenteric vessels with a characteristic “fatty halo” surrounding the vessels (*arrows*). (Courtesy of Luis B. Morettin, M.D., Galveston, TX).

Over time, some cases of mesenteric panniculitis progress to retractile mesenteritis characterized by a thickened mesentery that is fibrotic and retracted. There may also be mesenteric pseudocysts from fat necrosis. Rarely, there is involvement of the colonic mesentery, and fewer than 5% of patients have retroperitoneal involvement. Symptoms include continuous abdominal pain, fever, and weight loss. Some patients develop small bowel obstruction, mesenteric thrombosis, and lymphatic obstruction with ascites, steatorrhea, or protein-losing enteropathy.
Treatment and Prognosis Patients with mesenteric panniculitis should undergo biopsy of the lesion to rule out malignancy. This may be accomplished laparoscopically or by an open procedure. Resection of the entire mass is usually not possible and should not be attempted. In patients with symptomatic progressive sclerosing or retractile mesenteritis, dramatic responses have been reported after treatment with prednisone and azathioprine or with cyclophosphamide. [150], [151] Experience with these immunosuppressive agents is too limited to determine their efficacy in this usually self-limiting disease. Surgical diversion or bypass is

indicated for symptomatic relief in cases in which extrinsic bowel obstruction due to mesenteric inflammation and scarring has occurred. The prognosis for mesenteric panniculitis is usually good but depends on underlying disease processes.

Mesenteric Fibromatosis

Incidence and Etiology Fibromatous proliferation of the mesentery, called *mesenteric fibromatosis* or *mesenteric desmoid,* is a rare and noninflammatory condition.[152] These mesenteric lesions can infiltrate the surrounding structures, and similar to desmoid tumors in other locations, they tend to recur locally, but they never metastasize. Most of the reported cases have been associated with familial adenomatous polyposis syndromes (in particular, Gardner syndrome). Mesenteric fibromatosis affects about 3% of patients with familial adenomatous polyposis and 14% to 29% of patients with Gardner syndrome.[153],[154] This process has also been reported after abdominal surgery, trauma, prolonged estrogen intake, and pregnancy.[152] In addition, it may occur as a primary condition (primary mesenteric fibromatosis) in the absence of predisposing factors.
Clinical Manifestations Patients with mesenteric fibromatosis may present with an asymptomatic abdominal mass on physical examination or a mesenteric mass at laparotomy. Symptoms of intestinal obstruction are the most common complaint. Aggressive mesenteric desmoids can become infiltrative and can involve mesenteric vessels, leading to intestinal infarction and perforation. The radiographic features of mesenteric fibrosis are generally nonspecific. The desmoid tumor appears as a solid mass by ultrasound. CT scan shows a nonenhancing mass with soft tissue density. At laparotomy, desmoids are found to involve the mesentery of the small bowel or transverse mesocolon, and the most common site is the base of the small bowel mesentery.[155],[156] These lesions can be difficult to differentiate from low-grade sarcoma and can lead to misdiagnosis at operation and extensive small bowel resection.
Treatment and Prognosis In the past, the treatment of choice for patients with mesenteric desmoids has been wide local excision, but, unlike extra-abdominal desmoids, no clear evidence exists showing a benefit from negative margins on the rate of recurrence of mesenteric fibromatosis. Furthermore, the severity and frequency of complications, recurrence rates as high as 85%, and relatively good long-term survival in unresectable patients lead some to reserve resection for patients suffering from complications secondary to desmoid tumors (e.g., obstruction, perforation, or fistula formation).[157],[158] There are anecdotal reports of successful treatment with prostaglandin inhibitors (i.e., sulindac or indomethacin) and antiestrogens (e.g., tamoxifen) alone and in combination.[159],[160] and[161] Cytotoxic chemotherapy with dactinomycin, vincristine, and cyclophosphamide and radiotherapy have been used, individually or in combination, with variable success.[162],[163] and[164]

Cysts and Tumors of the Mesentery and Omentum

Mesenteric and Omental Cysts

Incidence and etiology. Cysts of the mesentery and omentum are uncommon lesions, with an incidence of about 1 case for every 140,000 general hospital admissions and about 1 per 20,000 pediatric hospital admissions.[165],[166] These cysts represent benign proliferation of ectopic lymphatics lacking communication with the normal lymphatic system.[167] Although they are most commonly found in the small bowel mesentery, they can occur anywhere along the gastrointestinal tract. Some authors differentiate between mesenteric or omental cysts and cystic lymphangiomas.
Clinical manifestations. Cystic lymphangiomas characteristically are found in children (mean age, 6 to 10 years) and are frequently large and almost always symptomatic, usually presenting with abdominal distention, pain, or vomiting and occasionally with an acute abdomen mimicking appendicitis.[168] Lymphangiomas are most often located in the small bowel mesentery but occasionally may be found in the mesocolon, the omentum, or, more rarely, the retroperitoneum. They may be single or multiple. In contrast to cystic lymphangiomas, the mean age of presentation of patients with nonlymphangiomatous mesenteric cysts is 44 years.[169] These cysts include nonpancreatic pseudocysts, enteric cysts, and mesothelial cysts. As many as one third of the nonlymphangiomatous cysts may be located in the omentum, and only 25% are symptomatic.
Diagnosis and treatment. The identification of an intra-abdominal cystic mass can be obtained by radiographic studies. Plain radiographs often demonstrate a mass displacing bowel gas and may demonstrate proximal dilation of the bowel. Ultrasonography, CT, and MRI may demonstrate the multilocular or unilocular nature of the cysts, which may have homogeneous or nonhomogeneous contents (Fig. 123-12). Of the three imaging modalities, ultrasonography probably yields the most information for the least expense.

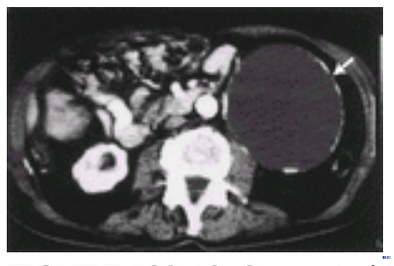

FIGURE 123-12. Computed tomography scan of a large lymphatic cyst arising from the mesentery (*arrow*). (Courtesy of Luis B. Morettin, M.D., Galveston, TX.)

Surgical resection represents the definitive diagnosis and treatment for these lesions. Percutaneous aspiration and biopsy are not warranted because they are not likely to provide a definitive diagnosis and may lead to infection or rupture of the cyst. Total excision of the cystic lymphangioma requires resection of adjacent attached abdominal organs (usually segments of small bowel) in most cases, but most other types of mesenteric cysts simply can be excised.[167],[169] Rarely, the location of the cyst precludes complete resection and requires drainage or marsupialization. However, this is usually associated with a high recurrence rate.[170],[171]

Solid Tumors of the Omentum and Mesentery

Incidence and etiology. Primary tumors of the omentum and mesentery are exceedingly rare and may arise from any of the cellular elements that constitute these tissues.[155],[172] By far the most common types of tumors that involve the omentum and mesentery are malignant tumors that arise from intra-abdominal viscera, such as the ovaries, stomach, colon, or pancreas, and secondarily involve the omentum or mesentery through metastasis or direct invasive extension. In some instances, the tumor arises from the mesenchymal elements in the wall of the gut segment; for example, leiomyoma, schwannoma or gastrointestinal stromal tumors (GISTS) are derived from smooth muscle, neural sheath, or interstitial cells of Cajal, respectively.[173],[174]
Clinical manifestations. Patients may present with a constellation of nonspecific or vague complaints; however, the most consistent presenting feature of omental and mesenteric tumors is a palpable abdominal mass that is often movable, often associated with pain, and sometimes associated with ascites or vomiting.[155],[172],[175] Plain abdominal radiographs and gastrointestinal contrast studies can demonstrate intestinal displacement and are useful in excluding the intestine as the origin of the tumor. A CT scan usually identifies the location of the mass within the mesentery or omentum and can exclude other organs as the origin of the mass but is nonspecific with regard to the diagnosis.[176]
Treatment. The treatment of primary omental and mesenteric tumors is surgical excision, if possible. The malignant tumors of the mesentery and omentum spread by local invasion and by peritoneal implantation and kill by involvement of vital abdominal organs. Surgery always should be considered in patients with symptoms of bowel obstruction or ischemia. Metastatic tumors of the mesentery are more common than primary mesenteric tumors and are usually the result of enlarged lymphomatous or carcinomatous lymph nodes. In addition, malignant midgut carcinoid tumors often metastasize or spread directly to the mesentery and exhibit pronounced fibrosis causing mesenteric shortening, angulation, and fixation of the bowel, which can result in bowel obstruction and intestinal infarction.[177],[178] and[179] Surgical palliation of patients with metastatic carcinoid tumors is warranted because of the indolent behavior of these tumors. Mesenchymal tumors (e.g., leiomyomas or GISTS) of the gut wall are also treated by surgical resection. The GISTS may express gain-of-function mutations of the *c-kit* protooncogene, which encodes a tyrosine kinase receptor. If the stromal tumor stains positive for *c-kit*, the tumor appears to be responsive to the inhibitor G1eevec, and studies are underway to assess the responsiveness of these tumors to this form of chemotherapy.

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CHAPTER 124

Phillip D. Smith and Edward N. Janoff

GASTROINTESTINAL COMPLICATIONS OF THE ACQUIRED IMMUNODEFICIENCY SYNDROME

The acquired immunodeficiency syndrome (AIDS) has become the greatest worldwide pandemic in history, surpassing even the bubonic plague of the 14th century in magnitude and number of deaths. Originating in chimpanzees in central Africa, [1] AIDS was first recognized in 1981 in men who have sex with men in the United States. [2], [3] and [4] The disease rapidly achieved epidemic proportions in this group and then intravenous drug users. Subsequently, human immunodeficiency virus type 1 (HIV-1), the causative agent of AIDS, spread to heterosexual populations, and, by the mid 1990s, heterosexual contact was recognized as the mode of transmission in 75% to 85% of infected people worldwide. [5]

In the 1990s, the AIDS pandemic was also recognized in developing countries throughout the world, but particularly those of sub-Saharan Africa. More than 70% of people infected with HIV-1 now live in these resource-poor nations, where in some countries, an astonishing 25% of the population is infected. [6] In these countries, vertical transmission has emerged as a major problem, and thousands of young children have been orphaned by the epidemic. In Botswana, for example, the prevalence of HIV-1 infection among pregnant women exceeds 30%. [6] By 2002, 20 million people worldwide were estimated to have died of AIDS and 40 million to be infected with the virus. The devastating impact of AIDS on the well-being of individuals and their families, societies, and national economies throughout the world is incalculable.

In the late 1990s, improved treatment options in the form of highly active antiretroviral therapy (HAART) and more effective agents for the prevention and treatment of opportunistic infections caused a decline in AIDS-related deaths in the United States and an increase in the prevalence of people living with HIV-1 infection. However, these therapeutic agents are prohibitively expensive throughout most of the world, and drug resistance is a serious potential problem. Moreover, development of an effective vaccine for HIV-1 infection has lagged far behind the development of therapeutic agents. Thus, until HIV-1 infection can be cured through antiviral therapy or prevented through an effective vaccine, clinicians throughout the world will continue to care for patients with AIDS.

The gastrointestinal tract mucosa plays a fundamental role in the pathogenesis and clinical manifestation of AIDS. Consequently, it is not surprising that gastrointestinal complications are common in patients with AIDS (Table 124-1). Infectious processes cause most of these complications and are responsible for relentless morbidity and, in some cases, mortality. [7] Among HIV-1–infected patients, diarrhea significantly diminishes the quality of life and is associated with an increased risk for death. The effective care of patients with AIDS requires a clear understanding of the gastrointestinal infections associated with HIV-1 disease and the most efficient and effective approaches for the evaluation and therapy of these infections. Therefore, this chapter focuses on the intestinal infections and key gastrointestinal syndromes associated with HIV-1 disease and presents effective approaches for their evaluation and treatment.

Diarrhea	Acute HIV-1 syndrome, infectious enteritis and colitis, drug-induced and AIDS enteropathy
Abdominal pain	Infectious organ disease, non-Hodgkin lymphoma, pancreatitis, and typhlitis
Hepatobiliary disorders	Acalculous cholecystitis, sclerosing cholangitis, and papillary stenosis
Pancreatitis	Drugs (pentamidine and dideoxyinosine), infections, and neoplasms (non-Hodgkin lymphoma and Kaposi sarcoma)
Gastrointestinal bleeding	Upper tract: infectious esophagitis, Kaposi sarcoma and lymphoma Lower tract: infectious colitis, idiopathic colonic ulcers, non-Hodgkin lymphoma and Kaposi sarcoma
Neoplasms	Non-Hodgkin lymphoma and Kaposi sarcoma
Anorectal disease	Anus: condylomata, infection, fistula, fissure and squamous cell carcinoma Rectum: nonspecific proctitis, infectious proctitis and perirectal abscess
Opportunistic infections	Inflammation, ulceration and inflammatory mass

AIDS, acquired immunodeficiency syndrome; HIV-1, human immunodeficiency virus type 1.

TABLE 124-1 Gastrointestinal Complications of AIDS

ROLE OF THE GASTROINTESTINAL MUCOSA IN HUMAN IMMUNODEFICIENCY VIRUS TYPE 1 INFECTION

All HIV-1 infections, excluding those acquired parenterally, are acquired across a mucosal surface. For heterosexual transmissions, the genital mucosa is the most common site of virus entry. In vertical transmission, HIV-1 is acquired through the upper gastrointestinal tract during the swallowing of infected amniotic fluid in utero, infected blood and secretions intrapartum, and infected breast milk postpartum. [8] Among men who have sex with men, the rectum is the portal of entry in anogenital contact and the upper tract the site of entry in orogenital contact. [9], [10]

In addition to their role in HIV-1 transmission, mucosal surfaces participate in many key events in HIV-1 pathogenesis. [11] First, mucosal cells likely are involved in selecting from the pool of inoculated viruses the phenotypically and genotypically restricted HIV-1 that is transmitted in acute infections. [12], [13] and [14] Second, lamina propria lymphocytes appear to be the initial target cells for HIV-1 infection. [15], [16] Third, HIV-1 replication occurs in the mucosa, a microenvironment containing an abundance of microorganisms and cytokines capable of enhancing HIV-1 replication in infected cells. [17], [18], [19] and [20] Fourth, the subsequent decline in the number of circulating $CD4^+$ T lymphocytes, a fundamental characteristic of AIDS, begins in the gastrointestinal mucosa. [21], [22], [23] and [24] Fifth, the local and systemic reduction in $CD4^+$ T cells predisposes the gastrointestinal tract mucosa to an array of opportunistic infections and neoplasms.

DIARRHEAL DISEASES

HIV-1-related diarrhea may first occur during the syndrome of primary HIV-1 infection 1 to 4 weeks after virus inoculation. This mononucleosis-like syndrome, characterized by fever, sore throat, rash, lymphadenopathy, fatigue, weight loss, and myalgias, occurs in up to 90% of patients acutely infected with HIV-1 and lasts 1 to 3 weeks. [10], [25], [26], [27], [28] and [29] Acute, self-limited diarrhea occurs in about one third of patients with the syndrome. [25], [29] Severe, but less common, gastrointestinal manifestations of acute HIV-1 infection include thrush and ulcerations of the oral cavity, esophagus, and rectum as well as enteropathy and profound weight loss. [29], [30] High titers of circulating virus coincide with the clinical syndrome, eventually eliciting an antibody response (seroconversion) that is followed by a rapid decline in the level of circulating virus. [27], [28] Resolution of acute HIV-1 syndrome is followed by a prolonged period of clinical latency that often lasted 8 to 10 years before the use of HAART. [31]

In addition to the diarrhea associated with acute HIV-1 syndrome and *opportunistic infections* (see later), medication-induced diarrhea is a frequent side effect of many antiretroviral and antimicrobial agents currently used to treat HIV-1 and opportunistic pathogens. [32], [33] Nucleoside analogs and protease inhibitors also may impair pancreatic function, causing steatorrhea. [32] Discontinuation, dose reduction, or accommodation to the offending drug are usually accompanied by resolution of symptoms.

Chronic diarrhea, defined as two or more loose or watery stools per day for at least 1 month, is common in patients with clinically apparent AIDS. Previously, in North America, diarrhea occurred in about 50% of HIV-1–infected patients [34], [35]; in developing African countries, the incidence of diarrhea may approach 90% of infected patients. [36], [37] HIV-1–associated diarrhea is typically chronic, often severe, and frequently associated with cramps, anorexia, fever, weight loss, and wasting. [35], [38], [39] Patients with lower CD4 $^{+}$ T-cell numbers are more likely to develop these symptoms. [35] The wasting is a consequence of the malabsorption of fat, carbohydrate, and probably protein [39], [40], [41], [42], [43] and [44] and reduced nutritional intake. [45] Tumor necrosis factor-a (TNF-a), a proinflammatory cytokine produced by activated or infected monocytes and macrophages, may be involved in the pathogenesis of AIDS-associated cachexia through its effects on lipid metabolism. [46]

Chronic diarrhea and *malabsorption* in HIV-1-infected patients cause fatigue and inanition, a marked decrease in the sense of well-being, and an impaired ability to conduct social and work-related activities of daily life. [47], [48] In African AIDS patients, diarrheal illness is more severe and accompanied by profound cachexia. [49] Referred to as *slim disease,* this severe manifestation of AIDS-associated diarrhea likely reflects limited access to medical care and the presence of opportunistic infections superimposed on traditional tropical infections. Patients with AIDS and diarrhea have lower numbers of circulating CD4 $^{+}$ T lymphocytes and a higher incidence of extraintestinal opportunistic infections, suggesting that infected patients who develop diarrhea have a greater degree of immunosuppression than those who do not. [50] Diarrhea in HIV-1–infected patients is associated with decreased survival in adults and infants. [44], [51], [52] In developed countries, the total cost of care for HIV-1–infected patients with diarrhea is 50% higher than for patients without diarrhea. [47] The increased outpatient costs reflect more office visits and diagnostic tests, and the increased inpatient costs reflect, in part, longer hospitalizations. [48] The high cost of providing health care and the related loss of ability to work are major drains on the financial resources of families and ultimately society in developed as well as developing countries.

Diagnostic Evaluation

The decision to begin an extensive evaluation of diarrhea in HIV-1–infected patients is tempered by the epidemiology and microbiology of potential enteric pathogens, the cost and feasibility of stool examination versus an invasive endoscopic procedure, the ability of the patient to undergo sedation and endoscopic evaluation, and the therapeutic implication of identifying certain pathogens. [38], [53] Because pathogen-specific therapy can often reduce the volume and frequency of the diarrhea, [50], [54] a vigorous diagnostic evaluation in HIV-1–infected patients with severe chronic diarrhea is usually justified. Increased awareness of the infectious processes involving the gastrointestinal tract (Table 124-2) and improved diagnostic techniques have led to the identification of enteric pathogens in 44% to 85% of AIDS patients with diarrhea. [50], [54], [55] and [56] A gastrointestinal pathogen is more frequently identified in symptomatic patients who also have weight loss and less than 100 CD4 $^{+}$ T cells/mm ^{3} than in those without weight loss and more than 100 CD4 $^{+}$ T cells/mm ^{3}, indicating that patients with more severe illness and greater immunosuppression are more likely to have an identifiable enteric pathogen. [56], [57] Two or more enteric pathogens may coexist in about 25% of AIDS patients with diarrhea. [58]

ORGAN	PATHOGENS
Esophagus	*Candida albicans*, herpes simplex virus, cytomegalovirus
Stomach	Cytomegalovirus, *Mycobacterium avium* complex
Biliary tract	Cytomegalovirus, *Cryptosporidium parvum*, microsporidia, *Isospora belli*
Gall bladder	Cytomegalovirus, *Crytosporidium parvum*, microsporidia, *Isospora belli*
Pancreas	Cytomegalovirus, *Mycobacterium avium* complex, herpes simplex virus
Small intestine	*Cryptosporidium parvum*, microsporidia, *Isospora belli*, *Cyclospora cayetanensis*, *Mycobacterium avium* complex, *Salmonella* species, *Campylobacter jejuni*
Colon	Cytomegalovirus, *Mycobacterium avium* complex, *Shigella* species, *Clostridium difficile*, *Campylobacter jejuni*, *Histoplasma capsulatum*, adenovirus, enteroaggregative *Escherichia coli*
Rectum	Herpes simplex virus, lymphogranuloma venereum, papilloma virus

HIV-1, human immunodeficiency virus type 1.

TABLE 124-2 Sites of Infections in the Gastrointestinal Tract in Patients Infected with HIV-1

Step 1 Evaluation begins with a thorough history and physical examination. The history should include a detailed review of the patient's drugs. In North America and Europe, antimicrobial agents and antiretroviral drugs are among the most common causes of gastrointestinal symptoms and diarrhea [33] (Table 124-3). In addition, the increased use of antimicrobial agents in HIV-1–infected patients may be associated with increased rates of *Clostridium difficile*–associated diarrhea. [59], [60] Thus, a review of current medications and their temporal relationship to symptoms is an increasingly important component of the diagnostic evaluation. The geographic regions in which a patient has resided or traveled are important because some AIDS-defining pathogens are endemic or more common in certain geographic regions. For example, *Histoplasma capsulatum* is endemic in the Mississippi valley and is more common in HIV-1–infected patients who have resided in or traveled to that region. [61] *Cryptosporidium parvum* and *Isospora belli* are more common in patients in developing countries than in developed countries. [62], [63] Rotavirus, which is not a common pathogen in AIDS patients in the United States, [50], [64] /SUP>has been identified in 18% of AIDS patients with diarrhea in Australia. [65]

TABLE 124-3 Gastrointestinal Complications of Antiretroviral Therapy and Potential Interactions with other Medications used in Gastrointestinal Medicine

Step 2 After the history and physical examination, we use the following protocol to evaluate diarrhea in HIV-1–infected patients. [66], [67] Up to three fresh stool specimens are cultured for *Salmonella* species, *Shigella* species, and *Campylobacter jejuni,* the most common bacterial diarrheal pathogens identified in patients with and without HIV-1 infection. In contrast, *Aeromonas* species, *Yersinia* species, and *Escherichia coli* 0157:H7 are not more common among patients with AIDS, and therefore, stool is not routinely cultured for these bacteria. Each specimen is also examined for ova and parasites using saline, iodine, trichrome, and acid-fast preparations of direct or concentrated samples of stool. Stool is assayed for *C difficile* toxin by immunoassay, but because about 10% of non-AIDS patients with *C difficile* infection are toxin negative, stool is cultured for *C difficile* when the bacterium is suspected despite a negative toxin assay. [68] The identification of enteric pathogens by stool examination alone in 48% to 55% of HIV-1–infected patients with diarrhea [54], [69] underscores the importance and cost-effectiveness of stool examination as the initial diagnostic test. [70] When fever is present, blood cultures are obtained for *Salmonella* species, *Shigella* species, and *C jejuni* because bacteremia often accompanies enteric infection in HIV-1–infected patients.

Step 3 When stool examination does not reveal a potential pathogen or when therapy for the identified pathogen does not improve symptoms, we perform esophagogastroduodenoscopy (EGD) and colonoscopy. In addition to visual inspection of the mucosa, tissue biopsies are obtained for histopathological examination and microbiologic evaluation for the presence of bacteria, protozoa, and viruses. Because small bowel fluid from HIV-1–infected patients rarely contains parasites [71] or excessive levels of bacteria (more than 104/mL), [72] we no longer routinely aspirate fluid from the small intestine to diagnose parasitic infection or bacterial overgrowth. The use of endoscopic procedures to evaluate HIV-1–infected patients with diarrhea was previously controversial, [70] but their usefulness has been confirmed in the following studies. Among HIV-1–infected patients with diarrhea and pathogen-negative stool studies, EGD alone yielded a diagnosis in 26% of patients, [71] EGD plus sigmoidoscopy a diagnosis in 50%, [44] and EGD plus colonoscopy a diagnosis in 44%. [56] The diagnostic yield of an endoscopic procedure is significantly greater for patients whose number of CD4$^+$ T cells is less than 100 cells/mm^3 and whose diarrhea is severe. [56], [73] A lower tract endoscopic procedure provided a diagnosis in 64% of patients with weight loss of more than 10 kg but yielded no diagnosis in patients with stool volumes of less than 400 mL/day or less than 5 kg lost weight. [74] Colonoscopy is usually superior to flexible sigmoidoscopy for the detection of pathogens and neoplasms; in one study, [73] 30% of pathogens and 75% of lymphomas were detected only in the proximal colon, well beyond the reach of the flexible sigmoidoscope. Thus, endoscopic procedures are an important adjunct to the diagnostic evaluation of unexplained chronic diarrhea in HIV-1 disease, and the diagnostic yield is greater for patients with more profound immunosuppression and severe diarrhea.

Step 4 When the evaluation of tissue specimens also does not yield an identifiable pathogen and when therapy for an identified pathogen is ineffective, glutaraldehyde-fixed tissue specimens obtained during step 3 can be evaluated by electron microscopy for microsporidian (duodenal biopsies) adenovirus and enteroaggregative bacteria (colonic biopsies). The cost required to perform electron microscopy should be weighed against the low prevalence and lack of an effective therapeutic agent for these pathogens. When no treatable pathogen is identified, the therapeutic approach should emphasize reducing symptoms rather than a continuous diagnostic evaluation.

Approach to Therapy

We approach the treatment of gastrointestinal infections in HIV-1–infected patients using the following principles. [67] First, during the diagnostic evaluation of diarrhea, many patients benefit from supportive therapy with fluids, electrolytes, and antimotility agents. Second, because the array of opportunistic and nonopportunistic pathogens that cause diarrhea in HIV-1–infected patients is extensive (Table 124-4) and because drug-related side effects and drug interactions are common, specific antimicrobial therapy is preferable to empiric therapy. Third, diarrhea and often other gastrointestinal syndromes, such as AIDS cholangiopathy, may be multifactoral in etiology, and, therefore, treatment may be only partially beneficial. Fourth, many microorganisms, including cytomegalovirus (CMV), herpes simplex virus (HSV), some enteric bacteria, and *Mycobacterium avium* complex (MAC), may be resistant or develop resistance to currently used antimicrobial agents. Consequently, infections with these pathogens require close monitoring with appropriate in vitro drug sensitivity testing. Fifth, long-term suppressive therapy is often necessary for infections that reactivate or recur after treatment. Sixth, specific antimicrobial therapy and supportive care can enhance the quality of life and duration of survival in many patients. Seventh, HAART may induce remission of some gastrointestinal infections, obviating the need for antimicrobial therapy for cryptosporidiosis and microsporidiosis. However, some infectious processes, such as CMV retinitis (when the CD4$^+$ T-cell number increases to more than 200 cells/mm^3), chronic hepatitis B infection and MAC adenitis may be exacerbated by HAART. HIV-1–infected patients with these processes should be followed closely for possible flare-up of their opportunistic infection. Finally, appetite enhancers may be helpful, but the role of total parenteral nutrition and anabolic steroids is uncertain.

	OPPORTUNISTIC	NONOPPORTUNISTIC
Bacteria	*Salmonella* species *Shigella flexneri* *Campylobacter jejuni* *Mycobacterium avium* complex Enteroaggregative *Escherichia coli*	*Clostridium difficile*
Protozoa	*Cryptosporidium parvum* *Enterocytozoon bieneusi* *Septata intestinalis* *Cyclospora cayetanensis* *Isospora belli*	*Giardia lamblia* *Entamoeba dispar*
Viruses	Cytomegalovirus Herpes simplex virus Adenovirus Picobirnavirus and astrovirus[†]	Rotavirus* Norwalk virus
Fungi	*Histoplasma capsulatum* *Candida albicans*[‡]	

*Nonopportunistic in United States and Africa, but more common in HIV-1–infected patients in Australia and Europe.

†More common in HIV-1–infected patients with diarrhea; pathogenesis under investigation.

‡Esophageal disease only.

HIV-1, human immunodeficiency virus type 1.

TABLE 124-4 Opportunistic and Nonopportunistic Pathogens in the Gastrointestinal Tract in Patients Infected with HIV-1

Highly Active Antiretroviral Therapy

A major advance in the therapeutic management of HIV-1 infection has been the development of HAART. HAART combines reverse transcriptase inhibitor (RTI) and protease inhibitor (PI) drugs into a powerful and highly effective antiviral regimen. RTIs include nucleoside analogs (NRTIs), such as zidovudine, and nonnucleoside analogs (NNRTIs), such as nevirapine, which inhibit the ability of HIV-1 RNA to encode viral DNA. PIs block the cleavage of HIV-1 Gag-Pol polyprotein precursor into structural core proteins and enzymes (reverse transcriptase, integrase, and protease), causing the production of abnormal viral proteins and noninfectious virions. Current HAART regimens typically include three or more agents, usually two NRTIs and one PI, to diminish or ablate viral replication, prevent the development of resistance, enhance recovery of immunologic competence, and improve clinical status. [75], [76], [77] and [78]

HAART has dramatically reduced the rate of death among patients with HIV-1. Among patients with advanced HIV-1 disease in the United States, mortality declined between 1995 and 1997 from 29.4

to 8.8 per 100 person-years. [79] The dramatic effect of HAART on mortality has been accompanied by an equally impressive decline in the incidence of many systemic and gastrointestinal opportunistic infections, [80], [81] and [82] most likely due to the inhibition of HIV-1 replication and the resultant recovery of immune function. Because the recovery of immune function takes several months, rates of opportunistic infections decline most prominently after 2 months of antiretroviral therapy. [83] Thus, prophylactic therapy against many such infections may be discontinued in patients on HAART whose $CD4^+$ lymphocytes have increased in number to 200 cells/mm^3 for more than 12 weeks. [83], [84], [85], [86], [87] and [88]

Cryptosporidiosis was the first AIDS-associated opportunistic infection shown to improve clinically and microbiologically in response to antiretroviral therapy. [89], [90], [91], [92] and [93] Patients with microsporidiosis also improve on HAART, although parasite-associated diarrhea may recur when the patient fails to respond to antiretroviral therapy. [93] The incidence of CMV gastrointestinal disease, esophageal candidiasis, and idiopathic esophageal ulceration also declines dramatically in association with HAART. [82] HAART appears to induce sustained clinical improvement in some patients with AIDS enteropathy. [92]

A spectrum of toxicities are associated with antiretroviral drugs [33] (see Table 124-3). Nausea, vomiting, abdominal pain, and diarrhea occur early in the course of therapy and may abate over time. Gastrointestinal toxicity significantly limits the initial tolerance to protease inhibitors and necessitates dose titration and sequential addition of other medications, such as RTI nucleoside analogs. [33] Hepatic toxicity is more often associated with long-term therapy and is typically mild, although severe hepatic toxicity, particularly with the PI ritonavir, may require discontinuation of the medication. [94] Pancreatitis is a more specific complication of dideoxyinosine. Toxic reactions ascribed to each drug are often confounded by the administration of two or more drugs at the same time. Increased rates of adverse events, such as pancreatitis or hepatic toxicity, may result from simultaneous administration of medications with overlapping toxicities. Distinguishing between symptoms of drug toxicity and opportunistic infection requires careful evaluation of the timing of events and the presence of comorbid findings.

HAART toxicities occur more frequently among patients with advanced HIV-1 disease ($CD4^+$ T cells less than 200 cells/mm^3) and may limit use of the antiretroviral drugs. [33] In patients taking NRTIs, gastrointestinal symptoms include nausea, vomiting, abdominal pain, and diarrhea in more than 25% of patients. Long-term therapy may be associated with myopathic, neuropathic, hepatic, pancreatic, and hematologic complications. [94], [95] and [96] Among patients taking NNRTIs, gastrointestinal and hepatic toxicities are not common. Mild rashes are the predominant side effect. Drug interactions occur primarily with inducers of and substrates for the cytochrome P450 system. The most frequent dose-limiting toxicities with protease PIs are diarrhea, nausea, and abdominal discomfort. Hepatic toxicity is most often associated with ritonavir. The presence of chronic hepatitis B or C virus may increase the rate but not necessarily the severity of hepatic effects [94]; these infections should not limit the initiation of HAART. Metabolic complications, particularly with PI-containing regimens and less often with NRTIs, include glucose intolerance, elevations in triglycerides and cholesterol, and lipodystrophy syndrome (increased abdominal fat, loss of peripheral subcutaneous fat of the face, arms, legs, and buttocks, and the presence of a “buffalo hump”). Women receiving therapy may experience breast enlargement.

The management of HAART toxicity is complicated by the need to identify the causative agent in a multi-drug regimen. Many of the toxic effects of antiretroviral medications are dose-dependent, and dose reduction may allow the continuation of therapy. Because optimal antiretroviral activity is usually dose-dependent, particularly for PIs, changing antiretroviral agents may be preferable to lowering doses. Because the development of HIV-1 resistance may occur, clinical and virologic status should be monitored. The management of overlapping toxicities of different medications and their effects on drug metabolism may allow continuation of the most critical agents. Severe or prolonged complications and the development of hypersensitivity reactions with abacavir require discontinuation of individual agents. For gastrointestinal side effects, antidiarrheal agents, such as loperamide or diphenoxylate atropine, may be useful for drug-related diarrhea, and antiemetics may reduce drug-induced vomiting. Taking medications with or after eating can limit nausea. Histamine receptor blockers and antacids may affect absorption of some drugs and usually do not alter symptoms.

Bacterial Infections

Salmonella* species, *Shigella* species, and *Campylobacter jejuni The gram-negative bacteria *Salmonella typhimurium* (less frequently, *Salmonella enteritidis*), [97], [98], [99], [100], [101], [102] and [103] *Shigella flexneri,* [103], [104], [105], [106], [107] and [108] and *C jejuni* [109], [110] cause a similar clinical illness in HIV-1–infected patients characterized by chronic or recurrent diarrhea, abdominal cramps, and fever. During symptomatic infection, the stool frequently contains fecal leukocytes and either gross or microscopic blood. Each of these bacterial infections occurs more often, causes more prolonged or recurrent diarrhea, and is more commonly associated with bacteremia and antibiotic resistance in HIV-1–infected patients than in seronegative patients. *C jejuni* may cause visible mucosal inflammation. *Campylobacter* species other than *C jejuni* have been detected in HIV-1–infected patients with diarrhea, [111] but a causative role for these species in the diarrhea awaits confirmation.

Diagnosis. These bacteria are diagnosed by culture of the organism in a stool specimen. The blood should also be cultured when diarrhea is accompanied by fever.

Treatment. The drug of choice for infection with *Salmonella* species is ceftriaxone or ciprofloxacin; for *Shigella* species, ciprofloxacin; and for *C jejuni,* erythromycin or ciprofloxacin (Table 124-5). Because recurrence is common and drug resistance may develop, repeat culture of stool and blood (when bacteremia occurs) and drug sensitivity testing are used to monitor therapeutic response.

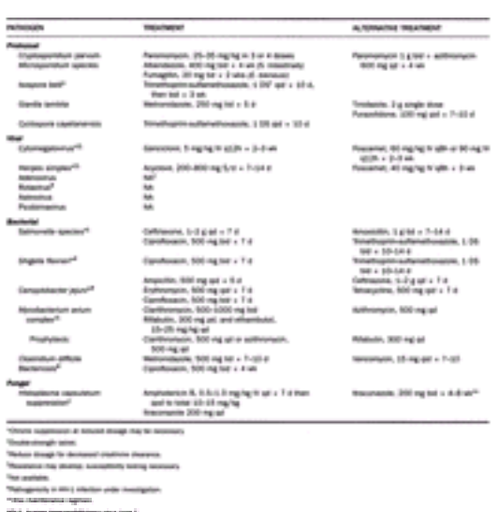

TABLE 124-5 Therapy for Intestinal Infections in HIV-1–Infected Adults

***Mycobacterium avium* Complex** MAC comprises two acid-fast, obligate, intracellular bacteria: *M avium* and *Mycobacterium intracellulare*. Exposure is common because the organisms are ubiquitous in the environment. In immunocompetent hosts, MAC is not an enteric pathogen, but immunosuppressed HIV-1–infected patients ($CD4^+$ T cells less than 100 cells/mm^3) are highly susceptible to infection. Macrophages are an important reservoir for the bacteria, and during HIV-1 infection, macrophages phagocytose, but appear unable to kill, MAC. The organism up-regulates macrophage HIV-1 production in dually infected cells [112] and alters macrophage function. [113] The gastrointestinal tract is the likely portal of entry for MAC. MAC is the most common cause of systemic bacterial infection in HIV-1–infected patients in the United States. MAC is much less common in Africa and Haiti. End-organ involvement and bacteremia usually do not occur until the number of circulating $CD4^+$ T cells declines to less than 100 cells/mm^3. Gastrointestinal tract disease, which is usually a manifestation of disseminated infection, may be associated with diarrhea, abdominal pain, malabsorption, weight loss, and fever, with or without night sweats. [114], [115] Concomitant hepatomegaly and splenomegaly with anemia due to MAC infection are often present. The numerous mycobacteria typically present in gastrointestinal tract mucosa suggest that MAC contributes to the associated gastrointestinal symptoms. Endoscopic abnormalities are nonspecific and include erythema, friability, erosions, and fine white nodules. Occasionally, an inflammatory mass, which can be mistaken for a tumor, is the major gastrointestinal manifestation. The small intestine is reported to be involved more often than the colon, [114] but this may reflect easier endoscopic access. Histological examination shows diffuse infiltration of the lamina propria by macrophages filled with bacilli. Granulomata are usually absent or poorly developed, possibly owing to the impaired chemotactic activity of monocytes in patients with AIDS. [116]

Diagnosis. The diagnosis of MAC is based on the identification of typical acid-fast organisms in stool or gastrointestinal biopsy specimens (fixed and touch preparations). [114], [115] Culture of gastrointestinal tissue may increase the positivity rate compared with acid-fast staining alone. [117] Isolation and speciation are achieved by culture of organisms from stool and biopsy specimens. Although the incidence of both pulmonary and extrapulmonary *Mycobacterium tuberculosis* infections is increased among HIV-1–infected patients, intestinal involvement is uncommon.

Treatment. Although initial trials of antituberculosis agents proved disappointing, improved results with newer multidrug regimens provide reason for cautious optimism in treating MAC. [84], [118], [119] Combination regimens have produced more sustained benefit, but such regimens are expensive, have adverse effects, and often are difficult to tolerate. The current recommended three-drug therapy is two antimycobacterial agents, including a macrolide azole agent and rifabutin, and a third agent, such as ethambutol. [120] When the $CD4^+$ T cell number falls below 50 cells/mm^3, clarithromycin or clarithromycin plus rifabutin is effective prophylactic therapy; MAC disease occurred in 9% and 7% of patients treated with clarithromycin alone or combination therapy, respectively. [121] For patients on

HAART who respond with an increase in the number of CD4 + lymphocytes to greater than 100 cells/mm 3 for more than 3 to 6 months with sustained suppression of HIV-1 plasma RNA for the same period, prophylactic therapy may be discontinued.

Clostridium difficile The gram-positive bacteria *C difficile* is an important pathogen in the differential diagnosis of diarrhea in patients with HIV-1 infection. Risk factors for *C difficile*–associated diarrhea in HIV-1–infected patients include recent antimicrobial therapy (particularly clindamycin and pyramethamine) and hospitalization, [122] risk factors similar to those in immunocompetent patients. [123] Although asymptomatic carriage occurs, *C difficile* infection in HIV-1–infected patients is commonly associated with profound watery diarrhea, abdominal pain and tenderness, fever, leukocytosis, and the presence of fecal leukocytes and occult blood. [124] However, the severity of the clinical illness, the response to therapy, and the frequency of recurrent or persistent infection in HIV-1–infected patients are similar to those of seronegative subjects. [125], [126] Thus, *C difficile* is a frequent, but nonopportunistic, pathogen associated with diarrheal illness in HIV-1–infected patients.

Diagnosis. The diagnosis of *C difficile*–associated diarrhea or colitis is based on the presence of diarrhea during antimicrobial use and the detection of either *C difficile* toxin or organisms in stool or endoscopically confirmed pseudomembranous colitis. The potential usefulness of stool culture is underscored by the higher rates of positive stool cultures (90% to 97%) compared with positive toxin assays (70% to 73%) in HIV-1–seronegative subjects with *C difficile* infection. [68], [127] Newer toxin assays show increased sensitivity for toxin A or are capable of detecting toxin A deficient strains of *C difficile*. [128]

Treatment. Metronidazole and vancomycin are highly effective for the treatment of *C difficile* infection in HIV-1–infected patients [124], [125] and [126] (see Table 124-5).

Enteroaggregative Bacteria Intestinal infection with enteroaggregative *E coli* has been reported in HIV-1–infected patients in the United States and Africa. [129], [130], [131] and [132] The bacterium adheres to enterocytes predominantly in the colon and ileum, where focal areas of epithelial cell injury (effacement, displaced microvilli, necrosis) are associated with attached bacteria. [129], [130] Accumulation of inflammatory cells in the epithelium or lamina propria is minimal or absent. Enteroaggregative *E coli* have been identified more frequently in stool from HIV-1–infected adults with diarrhea than in stool from asymptomatic seropositive subjects [132]; the mechanism of the diarrhea is not known. Therapeutic eradication of the bacteria with ciprofloxacin has been associated with restoration or improvement of diarrheal symptoms. [133]

Protozoan Infections

Cryptosporidium parvum *C parvum* is a unicellular, spore-forming coccidian protozoan taxonomically related to *Isospora, Toxoplasma, Sarcocystis,* and *Cyclospora*. [134], [135] Before the advent of HAART, cryptosporidia were present in about 20% of AIDS patients with chronic diarrhea in the United States [50], [54], [134], [136] but in as many as 55% of AIDS patients in developing countries such as Zaire and Haiti. [62], [137] Persistent infection occurs predominantly in HIV-1–infected patients with less than 200 CD4 + T cells/mm 3. [138], [139] A relatively common cause of self-limited mild diarrhea in immunocompetent people, [134], [140], [141] cryptosporidiosis can cause chronic, debilitating diarrhea in patients with AIDS. Typically, the diarrhea is voluminous, nonbloody, and watery; abdominal cramps, anorexia, malaise, and weight loss are also prominent symptoms. [142] Asymptomatic carriage may occur, [143] but symptoms of dehydration and wasting are more common. Malabsorption of both fat and carbohydrate is associated with cryptosporidiosis. Rarely, cryptosporidia may spread to the biliary tract, leading to biliary tract obstruction, [144], [145] or to the esophagus, causing distal esophagitis. [146] The parasite has been identified throughout the gastrointestinal tract, developing within a parasitophorous vacuole beneath the epithelial cell membrane but outside the cytoplasm. Despite its ability to cause severe disease, the spectrum of *Cryptosporidium* species–associated symptoms is broad, may vary over time, and is strongly influenced by the level of immunosuppression. [147], [148]

Diagnosis. Cryptosporidia are easily identified microscopically in stool with a modified acid-fast stain. Concentration of stool by zinc sulfate or sucrose flotation enhances detection of rare or infrequent oocysts during intermittent shedding. A single stool examination was sufficient to diagnose 96% of cases in one retrospective study. [149] When *Cryptosporidium* infection is suspected despite negative stool examination, hematoxylin and eosin–stained tissue specimens obtained by endoscopy can reveal the parasite; biopsy of the terminal ileum has a higher sensitivity (91%) than that of the duodenum (53%) for the detection of the parasite. [150] Organisms may also be identified on small and large intestinal brush-border biopsy specimens by electron microscopy, but this technique is rarely necessary.

Treatment. Despite reports of successful treatment of cryptosporidiosis in HIV-1–infected patients with spiramycin, [151], [152] paromomycin, [148], [153], [154] and [155] bovine colostrums, [156], [157] transfer factor, [158] somatostatin, [159], [160] and zidovudine, [89] therapy with these agents has not been consistently effective, and successful controlled clinical trials have not yet been performed. A course of paromomycin, a nonabsorbable aminoglycoside, with or without azithromycin (Table 124-5) is reasonable based on preliminary studies reporting symptomatic improvement and oocyst clearance in some patients. [148], [153], [154], [155] and [155a] Somatostatin may reduce the frequency and volume of *Cryptosporidium*–associated diarrhea; [159], [160] therapy is complicated by requirement that the drug be given parenterally, but long-acting preparations now allow once-a-day or less frequent injection. To date, HARRT is the most effective therapy for reducing the incidence, severity, and duration of AIDS-associated *Cryptosporidium* infections. [91], [92], [161], [162] and [163]

Microsporidia Microsporidia are spore-forming, obligate intracellular protozoa that infect the small intestine (*Enterocytozoon bieneusi*), small intestine with dissemination (*Septata intestinalis*), liver (*Encephalitozoon cuniculi*), and cornea (*Encephalitozoon hellem*). The parasite has distinctive ultrastructural features; many organisms at different stages of evelopment can be identified in the same enterocyte, causing cytopathic effect [164] (Fig. 124-1; Color Fig. 124-1). *E bieneusi* infects only enterocytes in the small intestine. *S intestinalis,* which is ultrastructurally distinct from *E bieneusi,* also infects epithelial cells but can penetrate into the lamina propria, infect macrophages, and disseminate to other organs such as the kidney. [165], [166], [167], [168] and [169] Microsporidia were initially identified exclusively in HIV-1–infected patients, but the parasite has now been diagnosed in seronegative liver and renal transplant recipients. [170], [171] and [172] and infrequently in immunocompetent people. [173], [174] and [175]

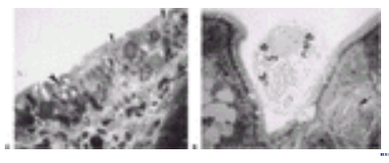

FIGURE 124-1. (See Color Fig. 124-1). **A:** Densely stained microsporidial spores are detected in the cytoplasm of several epithelial cells by light microscopic examination of an intestinal biopsy from a patient with microsporidiosis (semi-thin plastic section; methylene blue–azure II and basic fuchsin stain, original magnification × 630). **B:** Electron micrograph shows a necrotic intestinal enterocyte containing six microsporidial spores in the final stage of being sloughed into the lumen (original magnification × 10,000). (Courtesy of Dr. Jan M. Orenstein.)

Infection with the parasite is associated with chronic, watery, nonbloody diarrhea of variable severity, frequently with substantial fluid and weight loss. [176], [177] and [178] Microsporidiosis has been reported to cause 40% to 50% of the cases of unexplained diarrhea among patients with HIV-1 infection, [179], [180] but microsporidia infection is not invariably accompanied by diarrhea. [181], [182] *E bieneusi* was equally common in HIV-1–infected patients with and without diarrhea. The highest density of *E bieneusi* is located in the jejunum, [183] where the parasite infects enterocytes and may cause villous atrophy. [57] The organism has been associated with AIDS-related biliary tract disease, including acalculous cholecystitis, cholecystitis with cholelithiasis, and cholangitis, [184], [185] presumably by ascending the proximal small intestine to infect the biliary tract. *S intestinalis* also can cause biliary tract disease, including necrotizing cholangitis. [186]

Diagnosis. The diagnosis of microsporidiosis is based on the identification of the parasite in small intestinal tissue or fluid. Biopsy should be performed at the most distal site, where the parasite burden is the highest. Because of their small size, poor staining qualities, and minimal associated inflammation, microsporidia were formerly diagnosed exclusively by transmission electron microscopy. Microsporidia can be identified by light microscopy in semi-thin plastic sections of small intestinal biopsy specimens stained with methylene blue–azure II–basic fuchsin, (Fig. 124-1A; see also Color Fig. 124-1A) [171] touch preparations of mucosa stained with Giemsa, [187], [188] and stool specimens stained with a small modified trichrome (chromotrope 2R) stain. [187] One light microscopic technique appears to obviate the need for electron microscopy in more than 50% of cases, and a combination of any two techniques obviates the need in more than 75% of cases. [58] A rapid fluorescence technique based on Uvitex 2B binding to chitin, a component of the spore wall, is reportedly effective for detecting microsporidia. [180] However, because Uvitex 2B binds to any chitin-containing microorganism, morphologic evaluation may be necessary to confirm the diagnosis. Polymerase chain reaction using primers for the rRNA gene of *E bieneusi* and *S intestinalis* has been developed to identify microsporidia in tissue specimens, [189] but this diagnostic tool is not widely available.

Treatment. Curative therapy for microsporidiosis is not currently available. Albendazole, a benzimidazole derivative related to mebendazole, has been used to treat patients with both intestinal species of microsporidia. [189] (see Table 124-5). In open-label trials, [190], [191] albendazole was associated with cessation of weight loss. Albendazole appears to be more effective for *S intestinalis* than *E bieneusi* infections [192]; the drug has provided resolution or improvement in diarrheal symptoms and weight gain for patients with disseminated *S intestinalis* infections. [168], [169] Fumagillin is the

preferred drug for *E bieneusi.* 169a Patients on HAART have shown both improvement and resolution of diarrheal symptoms due to microsporidia infection. 161 , 163

Isospora belli *I belli* is an obligate, intracellular coccidian parasite that resembles *Cryptosporidium* in its life cycle and small intestinal habitat. In the early years of the AIDS epidemic, *I belli* was identified in fewer than 3% of AIDS patients in the United States but in up to 15% of patients in developing nations such as Zambia and Haiti. 63 , 193 Isosporosis is typically a chronic illness characterized by profuse, nonbloody, watery diarrhea that may be indistinguishable from the diarrheal illness associated with *Cryptosporidium* and microsporidia infections. Weight loss of at least 10% may occur during the months before diagnosis. 63 , 194 Nausea and abdominal cramps also typically accompany the illness; fever and vomiting are less frequent. Dehydration requiring hospitalization has been reported in nearly 50% of Haitian patients. 63 Steatorrhea and eosinophilia may also be present. Histology usually shows inflammation associated with some degree of villous atrophy; in contrast to *Cryptosporidium, I belli* may invade the mucosa. Although concentrated in the small intestine, *I belli* has been identified throughout the gastrointestinal tract and has been associated with biliary disease, specifically acalculous cholecystitis. 195 Rarely, the organism disseminates to extraintestinal sites, including mesenteric and tracheobronchial lymph nodes. 196

Diagnosis. The diagnosis of *I belli* is established by the identification of the typical highly refractile, spherical oocysts (containing two sporoblasts) in stool using the modified Kinyoun acid-fast stain. Stool concentration may be necessary to detect infrequent or rare oocysts.

Treatment. The most effective therapeutic agent for *I belli* is trimethoprim-sulfamethoxazole (see Table 124-5). Because infection recurs in as many as 50% of cases after treatment, prolonged or repeat therapy with this agent may be required to suppress clinical illness.

Cyclospora cayetanensis *Cyclospora* are obligate, intracellular coccidian parasites with morphologic and staining characteristics of a cyanobacterium-like organism. 197 , 198 The spherical cyst-like organisms measure 8 to 10 µm in diameter and, on modified acid-fast staining, resemble Cryptosporidia. 199 The *Cyclospora* oocyst has two sporocysts, each containing two sporozoites, distinguishing the parasite from the larger *I belli*. 199 Both sexual and asexual forms occur in small intestinal epithelial cells, suggesting the organism can complete its life cycle in a single host. 200 The organism has been identified in patients with diarrhea from Latin America (primarily Peru), the Caribbean, Southeast Asia, Eastern Europe, and the United States. In immunocompetent people, the diarrhea is typically abrupt in onset, watery, and prolonged (lasting a mean of 6 weeks) but self-limited. 201 The histopathology of the small intestine in patients with *Cyclospora*–associated diarrhea shows acute and chronic inflammation, surface epithelial disarray, and varying degrees of villous atrophy and crypt hyperplasia. 202 *Cyclospora* infection in patients with AIDS is associated with prolonged or relapsing diarrhea and weight loss as great as 10%. 203 , 204 Diffuse, cramping abdominal pain accompanies the diarrhea in more than one half of patients and fever in one third. 204 *Cyclospora* outbreaks have been associated with contaminated drinking water and imported raspberries. 205 , 206

Diagnosis. Modified Kinyoun stain is used to identify *Cyclospora* oocysts in a concentrated fresh stool sample; oocysts appear as 8- to 9-µm, acid-fast, granular spheres.

Treatment. Trimethoprim-sulfamethoxazole appears to be effective therapy for *Cyclospora* infection. 204 (see Table 124-5).

***Giardia lamblia* and *Entamoeba* species** In immunosuppressed HIV-1–infected patients, *Giardia lamblia* does not cause more frequent or severe infection than in seronegative patients and responds appropriately to antimicrobial therapy with metronidazole. 207 In addition, neither colitis nor liver abscess caused by *Entamoeba histolytica* infection appear to be more frequent or more severe in HIV-1–infected patients. The *Entamoeba* species usually detected in HIV-1–infected patients is *Entamoeba dispar,* a nonpathogenic species. 208

Viral Infections

Cytomegalovirus CMV is a double-stranded DNA virus in the herpesvirus group. After initial infection, which in immunocompetent patients is usually asymptomatic, the virus enters a latent, nonproductive phase in circulating mononuclear cells. 209 When the host becomes immunosuppressed, virus is reactivated, leading to replication and cell lysis. Replication occurs in the nuclei of permissive cells, and virus accumulates in nuclear and cytoplasmic inclusions, producing cell enlargement and the characteristic nuclear and cytoplasmic inclusions. In the immunosuppressed host, CMV has been detected in intestinal mononuclear, epithelial, endothelial, and smooth muscle cells. 210 The local inflammatory response appears to involve CMV-induced release of proinflammatory cytokines. 211 , 212 The clinical manifestations of CMV disease in the gastrointestinal tract include esophagitis, 213 gastritis, 214 small intestinal enteritis, 214 and, less frequently, acalculous cholecystitis, 214 papillary stenosis, 145 sclerosing cholangitis, 145 pancreatitis, 215 and appendicitis. 216 Colitis appears to be the most common enteric manifestation of enteric CMV disease 217 , 218 and 219 ; among AIDS patients with colitis or enteritis, CMV has been identified in biopsy specimens from 45% of patients. 218 CMV colitis is characterized by diarrhea, fever, and weight loss. Abdominal pain and hematochezia are also frequently present and help distinguish CMV colitis from the protozoan diarrheal illnesses described earlier. The colon appears to be particularly susceptible to progression to ischemic necrosis and perforation during CMV infection. 220 Regardless of whether diarrhea is present, CMV-associated disease of the biliary tract (usually without icterus or pruritus) should be included in the differential diagnosis of abdominal pain, nausea, and vomiting in patients with AIDS. Endoscopic findings that suggest CMV-induced disease range from localized hyperemia to hemorrhagic erythema to superficial or deep ulceration (Fig. 124-2; Color Fig. 124-2A).

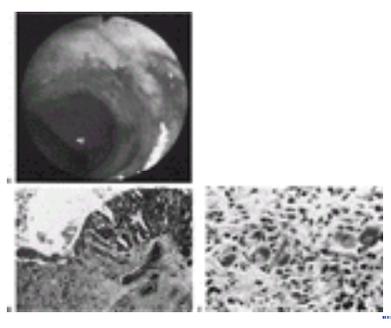

FIGURE 124-2. (See Color Fig. 124-2.) **A:** Endoscopic visualization of mucosal ulceration and inflammation in a patient with cytomegalovirus colitis. **B:** Light microscopic view of a colon biopsy from the same patient shows ulceration and hemorrhage. **C:** A higher magnification shows infiltration by inflammatory cells, many containing cytomegalic inclusions. (**B, C,** hematoxylin and eosin; **B,** original magnification × 30; **C,** original magnification × 62). (From ref. 211 .)

Diagnosis. CMV should be suspected when endoscopic or colonoscopic visualization shows discrete, often hemorrhagic, erosions or ulcerations with normal intervening mucosa in an HIV-1–infected patient. Colitis may be patchy in about 40% of cases and involve only the right colon or cecum in 18%, 220 underscoring the importance of colonoscopy in the diagnostic evaluation of colitis in an HIV-1–infected patient. The diagnosis of CMV gastrointestinal disease is established by the histopathological identification of large (cytomegalic) mononuclear, endothelial or epithelial cells containing intranuclear or cytoplasmic inclusions with surrounding inflammation 218 , 222 (see Fig. 124-2B, Fig. 124-2C; see also Color Fig. 124-2B, Color Fig. 124-2C) and confirmed by immunocytochemical staining, in situ hybridization, or DNA amplification.

Treatment. Ganciclovir and foscarnet, currently the drugs of choice (see Table 124-5), provide variable clinical responses. Because these agents are virustatic and not virucidal, recurrent CMV disease is common after the cessation of therapy. Ciclovovir is a new nucleoside analog with potent prolonged in vivo activity against CMV in patients with CMV retinitis, but the agent has not been studied in patients with gastrointestinal CMV disease.

Herpes Simplex Virus HSV is also a double-stranded DVA virus in the herpesvirus group. Latent in sensory nerve ganglia, HSV reactivation during immunosuppression leads to spread of the virus along sensory nerve pathways to mucocutaneous sites, where the virus next spreads among epithelial cells by lysis, releasing virions that infect and lyse adjacent cells. Gastrointestinal HSV disease is confined to the perianal region, rectum, and esophagus. The perianal lesions are typically chronic, cutaneous ulcers that cause localized pain but not diarrhea. 222 Involvement of the rectum (proctitis) is often associated with perianal disease. Proctitis manifests as severe anorectal pain, tenesmus, constipation, inguinal lymphadenopathy, and less often, difficulty with urination and sacral paresthesias. Such symptoms may also be associated with proctitis in seronegative homosexual men. 223 Diarrhea is not associated with typical proctitis, although the mucopurulent discharge may be misinterpreted as diarrhea. Proctocolitis, which occurs when the proctitis extends proximally into the distal sigmoid colon, can cause mild diarrhea associated with hematochezia, but the predominant symptoms are generally those of the proctitis. Overall, HSV proctocolitis is an infrequent cause of diarrhea in HIV-1–infected patients. 218

Diagnosis. Anoscopy and sigmoidoscopy are required to diagnose HSV proctitis and proctocolitis, respectively. Typical lesions begin as small vesicles and progress to erosions that often coalesce into diffuse ulcers. Diagnosis is predicated on the cytologic identification of intranuclear (Cowdry type A) inclusions in cells within the lesion and is confirmed by virus isolation.

Treatment. Acyclovir and related agents are the drugs of choice (see Table 124-5) for HSV infections. Because recurrent HSV is common, long-term suppressive therapy may be necessary.

Adenovirus In HIV-1–infected patients, adenovirus has been isolated from various body sites and may induce hepatic necrosis, 224 , 225 and 226 but intestinal involvement is rare. In the United States, adenovirus excretion is equally common in HIV-1–infected patients with and without diarrhea. 54 , 64 In contrast, 23% of Australian HIV-1–infected patients with diarrhea reportedly excrete adenovirus, compared with 5.4% without diarrhea. 65 Adenovirus has been identified with transmission electron microscopy and culture in inflamed colonic tissue of AIDS patients with chronic, watery, nonbloody,

nonmucoid diarrhea.[224, 227] Weight loss was also a prominent symptom. Endoscopically, the colonic mucosa showed areas of discrete, often raised, erythematous lesions that were several millimeters in diameter. Light microscopy showed chronic inflammation surrounding epithelial cells containing large, amphophilic intranuclear, but not cytoplasmic, inclusions. Adenovirus appeared to infect only epithelial cells, especially goblet cells, sparing lamina propria cells, which are frequent targets of CMV. At the ultrastructural level, adenovirus was associated with degeneration, death, and focal necrosis of infected epithelial cells, some of which had been extruded into the lumen (Fig. 124-3). Although adenovirus can cause pathogenic changes in the colonic mucosa, a causal relationship between adenovirus and diarrhea has not been established. Effective therapy is not available.

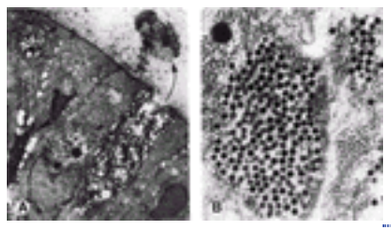

FIGURE 124-3. A: Transmission electron micrograph of a colonic biopsy specimen from a patient with adenovirus colitis shows a nucleus from a necrotic enterocyte extruded into the lumen. Adjacent cells have foci of disrupted cytoplasm containing secondary lysosomes (original magnification × 34,000). **B:** Higher magnification of a portion of the extruded nucleus in **A** shows numerous hexagonal virions characteristic of adenovirus (original magnification × 49,000). (From ref. [224].)

Astrovirus, Picobirnavirus, and Rotavirus Prospective evaluation of chronic diarrhea in HIV-1–infected patients indicated an association between the presence of astrovirus and picobirnavirus and diarrhea,[228] but further epidemiologic and clinical studies are needed to define the role of these viruses in HIV-1 disease. Rotavirus is not associated with diarrhea in HIV-1–infected patients in the United States[54, 64, 218, 228] or Africa (Zaire)[229, 230] but is the predominant virus detected in the stools of Australian HIV-1–infected homosexual men with diarrhea[65] and is present in the stool in about 14% of HIV-1–infected adults in Germany.[231] These findings suggest geographic variation in the prevalence of rotavirus-associated diarrhea in HIV-1–infected patients, but additional studies are needed.

Fungal Infections

Histoplasma capsulatum *Histoplasma capsulatum* is a dimorphic fungus that exists as a mycelial form with septate branching hyphae that contain readily airborne and infectious spores and a yeast form present in tissue macrophages. The organism is endemic in the central region of the United States and some tropical, subtropical, and temperate zone areas, where bird excrement provides microfoci of infection. Disseminated *H capsulatum* is as an important opportunistic infection in HIV-1–infected patients who reside in endemic areas.[232, 233] Gastrointestinal involvement generally reflects disseminated disease, which is due primarily to reactivation of quiescent infection. Among patients with disseminated *H capsulatum,* gastrointestinal involvement has been detected in about 70% of cases by evaluation of biopsy specimens, but gastrointestinal symptoms are present in only about 10% of patients.[234] Symptoms include diarrhea, weight loss, fever, and abdominal pain. Whether *H capsulatum* itself causes these symptoms has not been proved. Most patients with gastrointestinal manifestations of *H capsulatum* have colonic involvement. Colonoscopy in such patients may show inflammation, ulcerations, or fungating mass lesions.

Diagnosis. The histological examination of Giemsa-stained sections shows small yeast-like cells within macrophages or histiocytes. When infection is intense, the organism is not difficult to identify. However, the diagnosis of histoplasmosis is established by culture. Serologic detection of complement-fixing antibodies to *H capsulatum* mycelial antigens provides supportive evidence for infection.

Treatment. Amphotericin B is the drug of choice for disseminated histoplasmosis (see Table 124-5). The recommended therapy in HIV-1–infected patients is amphotericin B, 0.5 to 1.0 mg/kg intravenously daily for 7 days, then every other day to a total of 10–15 mg/kg followed by itraconazole 200 mg qd for chronic suppression.[235] Long-term therapy with amphotericin B, 50 to 80 mg intravenously every 2 weeks, is also highly effective in suppressing relapses after initial treatment.[233] The role of caspofungin acetate, a fungicidal echinocandin, is under evaluation.

ABDOMINAL PAIN

Abdominal pain is less common than diarrhea in HIV-1–infected patients. In two cohorts of patients with HIV-1 infection, the prevalence of severe abdominal pain was 12% among patients attending a surgical outpatient clinic and 15% among consecutive patients admitted to a hospital.[236, 237] The causes of abdominal pain in patients with HIV-1 infection are dependent on the severity of the immune suppression. Among HIV-1–infected patients with relatively preserved immune function ($CD4^+$ T cells more than 200 cells/mm^3), pain is typically due to traditional gastrointestinal disorders, such as peptic ulcer disease, biliary tract disease, inflammatory bowel disease, pancreatitis, and less commonly, appendicitis and diverticulitis. In contrast, among patients with AIDS, pain is commonly the consequence of a neoplastic process or an opportunistic infection.[236, 237, 238] and[239] The most common diseases associated with abdominal pain in patients with AIDS are *non-Hodgkin lymphoma* involving the gastrointestinal tract or abdominal cavity, CMV disease of the small intestine and colon, and *pancreatitis*. *Hepatobiliary disease* due to opportunistic infections (see later) is the leading consideration in the differential diagnosis of right upper quadrant pain in patients with AIDS. Less common causes of AIDS-associated abdominal pain include *typhlitis* or *necrotizing enterocolitis* with or without neutropenia[240, 241] and unusual manifestations of opportunistic infections, such as CMV-induced peritonitis[215] and appendicitis.[216]

AIDS-associated conditions that may present as an acute abdomen requiring emergent laparotomy include intestinal or colonic perforation due to CMV disease, intestinal obstruction or perforation caused by Kaposi sarcoma or lymphoma, intussusception associated with neoplasm or infection, and appendicitis caused by infection and neoplasm.[210, 220, 242, 243] and[244] Aside from the expanded array of causes for abdominal pain in patients with AIDS, the evaluation and management of abdominal pain in HIV-1–infected patients is otherwise similar to that in seronegative patients. Although emergent abdominal surgery was accompanied by high rates of complication and mortality early in the AIDS epidemic,[236, 242] improved awareness of the spectrum of AIDS-related abdominal complications and better surgical approaches have brought these rates into the acceptable range,[243] making emergent surgical intervention appropriate management for the acute abdomen in patients with HIV-1 infection and AIDS.

HEPATOBILIARY DISORDERS

HIV-1-infected patients and patients with AIDS may develop *cholecystitis* and *cholangitis* due to cholelithiasis and the same gram-negative bacteria that cause biliary tract disease in seronegative patients. In addition, HIV-1–infected patients are susceptible to a spectrum of biliary tract disorders related to *opportunistic pathogens* (see Table 124-2) and *lymphoma,* referred to collectively as the *AIDS cholangiopathy*. These disorders became apparent early in the AIDS epidemic when several patients with AIDS presented with right upper quadrant abdominal pain, jaundice, and fever and were shown to have acalculous cholecystitis, biliary tract obstruction due to stenosis of the distal common bile duct, and narrowing and irregularity of the intrahepatic and extrahepatic bile ducts.[144, 214] The identification of *Cryptosporidium* species infection or CMV in the gallbladder and bile ducts led to the conclusion that these pathogens had an etiologic role in the disorder. Endoscopic retrograde cholangiography further defined the underlying lesion as consistent with papillary stenosis and sclerosing cholangitis.[145] Besides *Cryptosporidium* infection and CMV, other opportunistic pathogens implicated etiologically in the syndrome include *E bieneusi, E intestinalis, I belli,* and *Pneumocystis carinii*.[144, 145, 184, 185, 195, 245, 246, 247] and[248]

Endoscopic retrograde cholangiography is the most effective diagnostic tool for evaluating the extent of the disease and excluding other processes, such as cholelithiasis, and sphincterotomy is an effective therapeutic modality for relieving the obstruction and pain.[145, 249] Rarely, HIV-1–associated non-Hodgkin lymphoma may cause sclerosing cholangitis, resulting in a similar clinical syndrome.[250] The long-term prognosis of HIV-1–infected patients with hepatobiliary cholangiopathy is similar to that of patients without the syndrome.[251, 252]

Hepatitis C virus (HCV) is the leading cause of chronic hepatitis among HIV-1–infected patients in the United States. Intravenous drug users and patients with hemophilia compose most of these co-infected patients; 40% of HCV-infected hemophiliac men are co-infected with HIV-1.[253] The natural history of patients infected with both HCV and HIV-1 differs from that of patients with HCV alone. Patients with both infections have high HCV RNA levels in blood and liver tissue, more rapid progression of liver disease, and possibly increased rates of death related to hepatic disease.[254, 255, 256] and[257] HCV viral loads are typically increased with advanced immunosuppression and are inversely proportional to $CD4^+$ T-cell number.[94, 254, 257] Despite the accelerated course of HCV replication and associated liver disease, HCV does not usually promote progression of HIV-1 disease or increase mortality.[258, 259] However, an increased risk for death and for new AIDS-defining illnesses and delayed recovery of $CD4^+$ T cells with antiretroviral therapy have been reported in some HIV-1–infected patients with HCV infection.[260] These increased risks were primarily associated

with intravenous drug use and unrelated to more progressive liver disease.

The response of HCV to antiviral therapy with interferon (IFN) in patients with HIV-1 co-infection appears to be similar to the response in HIV-1–seronegative subjects. About one fourth of patients show a sustained reduction in the level of virus 1 year after discontinuing therapy. [261], [262] and [263]; most responders have high CD4 + T-cell numbers (more than 500 cells/mm 3). Preliminary data suggest that the antiviral effects of IFN therapy plus ribavirin may be greater than with IFN alone among both HIV-1–infected and seronegative patients, [264], [265] although anemia is an important complication.

Another therapeutic option for patients with HCV and HIV-1 is HAART. In several series, [253], [266], [267], [268], [269] and [270] levels of plasma HCV and transaminases showed transient and clinically unapparent increases with HAART, whereas HCV RNA decreased to below the limits of detection, albeit in a minority of patients. Although both subclinical and severe hepatitis can occur with the initiation of HAART, [94], [270], [271] and [272] the presence of co-infection with HCV and HIV-1 should not be a primary deterrent for beginning antiretroviral therapy. The relatively infrequent “immune restoration” syndrome, characterized by deleterious inflammatory responses or even reactivation of infections (e.g., CMV and MAC) on HAART, [273], [274] and [275] appears not to affect HCV infection. Efforts to reduce the impact of HCV infection should include limiting the use of alcohol, vaccination for hepatitis A if the patient is seronegative, and therapy for HCV to eradicate infection or limit progressive liver injury in those with significant hepatic pathology.

PANCREATITIS

Pancreatitis is an important gastrointestinal complication in patients with HIV-1 infection. Autopsy studies have identified an array of *opportunistic pathogens,* including viruses (CMV and HSV), fungi (*Cryptococcus neoformans*), bacteria (*Mycobacteria* species), and protozoa (*Toxoplasma gondii*), as well as *neoplasms* (non-Hodgkin lymphoma and Kaposi sarcoma), in the pancreas. [276] Although CMV and tuberculosis may cause clinical pancreatic disease, [215], [277], [278] most infectious and neoplastic processes involving the pancreas in HIV-1–infected patients are subclinical. In contrast, *drug-induced pancreatic disease* is a common cause of clinical pancreatitis among patients with HIV-1 infection. [279] The two most common drugs implicated in HIV-1–associated pancreatitis are pentamidine and dideoxyinosine. Pentamidine-induced disease, which may occur with either inhaled or parenterally administered drug, causes mild to severe and even fatal pancreatitis. Dysregulation of glucose metabolism is common in this setting, although pentamidine itself may cause dysregulation in the absence of clinical pancreatitis. [280] Dideoxyinosine therapy may induce pancreatitis in up to 10% of patients [281], [282]; occasionally, dideoxyinosine-induced pancreatitis is fatal. Low-level hyperamylasemia (serum amylase level less than three times the upper limit of normal) may occur in patients with AIDS despite the absence of underlying pancreatic disease. [283], [284] After antiretroviral drug-induced pancreatitis has been excluded, the presentation, evaluation, and management of acute pancreatitis in HIV-1–infected patients are similar to those in seronegative patients.

GASTROINTESTINAL BLEEDING

Gastrointestinal bleeding is uncommon in HIV-1–infected patients with and without AIDS. In one prospective study, [285] upper gastrointestinal bleeding occurred in 6% of HIV-1–infected patients during a 14-month period. HIV-1–infected patients are as likely to bleed from HIV-1–related etiologies as *non–HIV-1-related causes*. The most common HIV-1–related causes are *viral esophagitis, gastroduodenal lymphoma,* and *MAC disease* of the proximal small intestine. [285], [286] and [287] Regarding viral esophagitis, HSV is the most common cause of bleeding; CMV esophagitis is rarely associated with significant bleeding. [213], [288], [289] Thrombocytopenia and other coagulopathies infrequently cause bleeding from minor lesions.

Similar to the low prevalence of bleeding in the upper gastrointestinal tract, intestinal and colonic bleeding in HIV-1–infected patients is uncommon. Although the differential for lower gastrointestinal tract bleeding includes traditional causes, such as diverticulosis, angiodysplastic lesions, and hemorrhoids, *opportunistic infectious processes* are, in our experience, the most common etiology for bleeding from the lower gastrointestinal tract in patients with HIV-1 infection. In contrast to the esophagus, however, where HSV is the most common infection associated with bleeding, lower tract bleeding is more frequently caused by CMV. [219], [220], [290] Among HIV-1–infected patients with CMV colitis, bleeding is reported to occur in up to 10%. [220] Typically, bleeding is slow and associated with abdominal pain and diarrhea; bleeding from CMV colitis is rarely severe. [291] Other HIV-1–related causes of lower gastrointestinal tract bleeding include neoplasms, such as non-Hodgkin lymphoma; unusual infections, such as *H capsulatum*; and idiopathic ulcerations. The approach to diagnosis and management of gastrointestinal bleeding in HIV-1–infected patients is similar to that of seronegative patients. Gastrointestinal bleeding is associated with reduced survival in patients with HIV-1 infection owing to the comorbidity associated with late-stage AIDS. [287]

GASTROINTESTINAL NEOPLASMS

Kaposi sarcoma and *non-Hodgkin lymphoma* are the most common gastrointestinal neoplasms associated with HIV-1 infection. Recognized formerly as a rare neoplasm of the skin in older men of Jewish or Eastern European origin, Kaposi sarcoma emerged in the 1980s as one of the most common opportunistic processes heralding the AIDS epidemic. Although the incidence of Kaposi sarcoma has declined in recent years, [292] the tumor remains an important gastrointestinal malignancy in HIV-1–infected, immunosuppressed, homosexual and bisexual men; Kaposi sarcoma occurs rarely in those who acquire HIV-1 intravenously or through heterosexual contact. Epidemiologic and molecular studies have implicated human herpesvirus type 8 (HHV-8), also termed *Kaposi sarcoma herpesvirus,* etiologically in the development of Kaposi sarcoma. [293], [294], [295], [296] and [297] Histologically, the tumor is characterized by the proliferation of spindle-shaped endothelial cells in a stroma of proliferating abnormal vessels, fibroblasts, and infiltrating leukocytes. The local presence of angiogenic growth factors, HHV-8, and impaired immunosurveillance mechanisms appear to predispose the gastrointestinal tract to Kaposi sarcoma formation.

Endoscopically, Kaposi sarcoma begins as a small hyperpigmented plaque or macule, progressing to a violaceous nodule and eventually a tumor-like mass. In the early stage of the AIDS epidemic, visible tumor was identified in the gastrointestinal tract in 40% to 50% of patients with AIDS. [298], [299] Although the tumor may be more common in the upper tract, Kaposi sarcoma occurs throughout the gastrointestinal tract. Oropharyngeal Kaposi sarcoma is highly suggestive of upper tract involvement. Most patients with enteric Kaposi sarcoma are asymptomatic; consequently, therapy for gastrointestinal lesions is rarely necessary. IFN-a and doxorubicin treatment have been associated with tumor regression. [300], [301] Unusually large tumors may cause bleeding, obstruction, or perforation and can be treated emergently with radiation therapy.

Among HIV-1–infected patients with non-Hodgkin lymphoma, the most frequently involved gastrointestinal sites are the colon (46%), ileum (39%), and stomach (23%). [302] Symptoms associated with gastrointestinal lymphoma include abdominal pain, weight loss, obstruction, and bleeding. [302] /SUP>, [303] and [304] The presence of latent and replicating Epstein-Barr virus DNA in the lymphomas of patients with AIDS suggests that lymphoproliferative lesions in the gastrointestinal tract could represent the outgrowth of Epstein-Barr virus–transformed mucosal B [305] Chemotherapy, surgery, radiotherapy, or combination therapy have not substantially improved survival of HIV-1–infected patients with gastrointestinal lymphoma.

ANORECTAL DISEASE

Anal complications associated with HIV-1 disease includ*perianal condylomata, infection, fissure, fistul*, and *squamous cell carcinom*. These complications occur predominantly in HIV-1–infected homosexual and bisexual men. Homosexual men, particularly those who are HIV-1 positive, have a high incidence of anal infection with human papillomavirus (HP[306] which has been implicated as a risk factor for anal condylomata and anal squamous cell carcino[307] The link between HPV, particularly types 16 and 18, and anal carcinoma may be through the ability of the virus to induce local dysplasia in a high proportion (15%) of immunosuppressed, HIV-1–infected homosexua[308] Perianal infection is commonly caused by HSV, but rectal involvement is rare. Rectal complications in HIV-1-infected patients include nonspecific proctitis (rectal inflammation in the absence of an identifiable patho[309] and infectious proctitis due to *Chlamydia trachomatis, Treponema pallidum, Neisseria gonorrhoea* or HSV.[310] Perirectal abscess in HIV-1–infected patients is often idiopathi[311] but may be associated with HP[312], [313] or CMV.[314]

The initial presentation of anorectal disease includes dyschezia (pain on defecation), bleeding, drainage from the anal canal, palpable mass, and sepsis syndrome (local pain and fever). Dyschezia usually results from ulcerative lesions in the anal canal due to infection, abscess, fissure, hemorrhoids, or neoplasm. A palpable mass suggests neoplasm. When the mass is tender and accompanied by fever, perirectal abscess leads the differential diagnosis. Tenesmus, urgency, and frequent small-volume stools suggest proctitis. Perianal drainage is usually caused by a fistulous tract or, less commonly, severe proctitis. Careful inspection, palpation of the perianal area and anal canal, and visualization of the rectum are mandatory components of the anorectal evaluation. Biopsy is performed for microbiologic evaluation and histological examination, and computed tomography is helpful for staging neoplasms and evaluating perirectal abscess. Symptomatic treatment, pathogen-specific antimicrobial therapy, and surgery in selected patients [315] are effective for most nonmalignant anorectal lesions. Anorectal neoplasms are treated with radiation therapy, chemotherapy, and, when localized, surgical res[316] ion.

In summary, the gastrointestinal tract plays a critical role in HIV-1 infection at virtually all stages of disease. As the largest lymphoid organ in the body, the intestinal mucosa is an important site of virologic and immunologic events that eventually lead to local immunosuppression and the gastrointestinal complications of AIDS, particularly the acquisition of opportunistic infections. New insights into the natural history of these infections, improved diagnostic modalities, new antiretroviral therapy, and more effective antimicrobial agents provide sound reasons for optimism in caring for patients with the gastrointestinal complications of AIDS.

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CHAPTER 125

Ellen Li and Samuel L. Stanley, Jr.

PARASITIC DISEASES: PROTOZOA

Protozoan parasitic infections of the gastrointestinal tract are being increasingly recognized as an important public health problem in the United States. They were the most frequently identified etiologic agents in water-borne disease outbreaks in 1991 through 1994. [1] The reasons for the apparent increased prevalence of intestinal parasites in the United States include the continuing immigration of people from developing countries in southeast Asia, the Caribbean, and Central America; the expanded use of day-care facilities, which represent breeding grounds for any fecally-orally transmitted pathogens; a growing problem with fecally contaminated food and water supplies; the increased recognition of parasites as pathogens of patients with acquired immunodeficiency syndrome (AIDS); and the development of improved stool examination techniques. [2]

The intestinal protozoan pathogens include both parasites that are extracellular, such as *Giardia lamblia, Blastocystis hominis,* and *Entamoeba histolytica,* and the intracellular spore-forming coccidian parasites *Cryptosporidium parvum, Cyclospora cayetanensis,* and *Isospora belli.* This latter group of organisms has emerged as important pathogens in immunocompetent individuals as well as in patients with AIDS. The most commonly detected protozoan pathogen in stool specimens in the United States is *G lamblia* [2] (Table 125-1). *B hominis* is the next most commonly detected protozoan in stool, although its role in intestinal disease remains controversial. The reported prevalence of the coccidian organisms, such as cryptosporidia, is probably underestimated because the appropriate procedures necessary to detect them may not be routinely performed in many clinical laboratories. Conversely, the prevalence of clinically significant *E histolytica* infections may be overestimated, based on the recent recognition that organisms identified as *E histolytica* by microscopic examination could be the morphologically indistinguishable nonpathogenic *E dispar*. In some cases in which infection is suspected on clinical grounds, stool examination may not be revealing, and endoscopy may be necessary to make the diagnosis. It is important for the gastroenterologist to know what the likely organisms are, where and what to sample, and how specimens should be processed to optimize the diagnostic yield (Table 125-2).

ORGANISM	STOOL SPECIMENS CONTAINING THE ORGANISM (%)
Giardia lamblia	7.2
Blastocystis hominis	2.6
Entamoeba histolytica	0.9
Dientamoeba fragilis	0.5
Cryptosporidium species	0.2

Note: The results of 216,275 stool specimens examined for intestinal parasites by state public health laboratories in 1987 are shown. Only organisms detected at a level of more than 0.1% of the stools are listed.

Adapted from ref. 2.

TABLE 125-1 Intestinal Protozoan Parasites Detected by Stool Examination in the United States

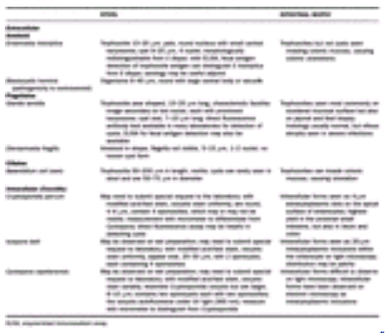

TABLE 125-2 Morophology of Human Gastrointestinal Protozoan Parasites

EXTRACELLULAR PROTOZOAN PARASITES

Amebiasis: Entamoeba histolytica

E histolytica is the causative agent of human amebiasis. The clinical manifestations of human amebiasis range from asymptomatic disease to invasive disease manifested as amebic colitis or liver abscess. Although Brumpt postulated in 1925 that there were two morphologically indistinguishable species of *E histolytica*—one that could cause disease and a second that could not [3]—only in the 1990s have molecular genetic analyses confirmed data from isoenzyme studies and clearly established that there are two genetically distinct but morphologically identical species [4], [5]: *E histolytica,* which can cause invasive amebiasis, and *E dispar,* a harmless commensal.

The life cycle of *E histolytica* consists of two stages: the cyst (host infective stage) (Fig. 125-1) and the trophozoite (host tissue invasive stage) (Fig. 125-2). No host other than humans is implicated in the life cycle, although natural infection of primates has been reported. [6] The disease is most commonly acquired by ingestion of food or water contaminated with feces containing the cyst form of the parasite, but venereal transmission through fecal-oral contact also occurs. Under still undefined stimuli in the intestinal tract, the cysts excyst and then form by nuclear and cytoplasmic division a total of eight trophozoites. The trophozoites are the motile feeding form of the parasite and contain a single nucleus and pseudopodia. Trophozoites are the causative agents of invasive disease but play no role in the transmission of disease, owing to their rapid degeneration outside the body and destruction by gastric acidity. *E histolytica* trophozoites multiply by binary fission, and they encyst, producing uninucleate cysts, which then undergo two successive nuclear divisions to form the characteristic quadrinucleate cysts.

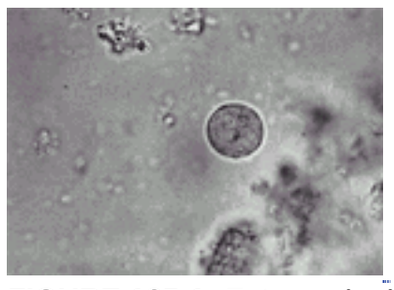

FIGURE 125-1. *Entamoeba histolytica* cyst from stool specimen stained with iodine. The cyst contains four nuclei and measures 9 to 25 µm in diameter. (Courtesy of Centers for Disease Control and Prevention, Atlanta, GA.)

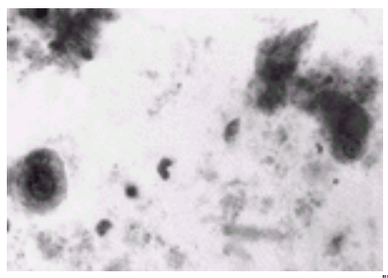

FIGURE 125-2. *Entamoeba histolytica* trophozoites in stool. Note ingested red cells and the single nucleus (trichrome stain). (Courtesy of Centers for Disease Control and Prevention, Atlanta, GA.)

Epidemiology It has been estimated that 500 million people are infected with either *E histolytica* or *E dispar.* [7] In asymptomatic individuals, the prevalence of *E dispar* infection is about 7- to 10-fold higher than *E histolytica* infection. Forty million individuals infected with *E histolytica* develop disabling colitis or extraintestinal abscesses, resulting in 50,000 deaths annually. Among parasitic diseases, only malaria and schistosomiasis result in more deaths than amebiasis. [7] High prevalence rates of amebiasis (both *E histolytica* and *E dispar*) are found in developing countries, including Central and South America, the Indian subcontinent and Indonesia, and the sub-Saharan and tropical regions of Africa. [7], [8] and [9] A recent prospective study of a cohort of almost 300 children living in a slum region of Dhaka revealed that almost 40% acquired a new *E histolytica* infection during the 12-month study period, but only 3% of the infections were associated with dysentery. [8] A serologic study of the inhabitants of Mexico City demonstrated that 8.4% had been infected with *E histolytica* in the past 5 to 10 years. [9] The prevalence of colonic disease is equal between men and women, but amebic liver abscesses and other extraintestinal disease are 3 to 10 times more common in men. Children, especially neonates, and pregnant and postpartum women have an increased risk for severe disease and death. Malignancy, malnutrition, and treatment with corticosteroids are other risk factors for more severe disease. In developed countries of Europe and North America, amebic infection is generally confined to certain high-risk groups, including recent travelers and immigrants, inmates of mental institutions, and sexually active homosexual men. [10] However, it should be noted that even in these populations, most individuals are infected with *E dispar* rather than *E histolytica*. [11], [12] There is conflicting evidence regarding naturally acquired immunity to amebiasis. In the study of Bangladeshi children, some children showed resistance to the acquisition of a new *E histolytica* infection that was associated with the presence of stool immunoglobulin A antibodies to an *E histolytica* antigen. [8] However, the effect was relatively short lived, and almost 20% of the children in the study had more than one episode of amebiasis during the 1-year study period. [8] These data, along with anecdotal reports of multiple episodes of amebic liver abscess in the same individual, suggest that naturally acquired immunity to amebiasis is, at best, short lived and may occur only in a small proportion of infected individuals. [13], [14]

Pathogenesis and Pathology Invasive disease is characterized by ulcerations of the intestinal mucosa, which extend into the submucosa. Trophozoites invading laterally through the submucosa, relatively sparing the overlying epithelium, give rise to the classic flask-shaped ulcer of amebiasis. [15], [16], [17], [18] and [19] In some cases, amebic invasion through the mucosal and submucosal layers and subsequent dissemination through the portal circulation to the liver result in the formation of liver abscesses. Adhesion of amebic trophozoites to host epithelial cells through a galactose-binding lectin, [20] release of cysteine proteases, [21], [22], [23], [24] and [25] amebic contact-dependent cytolysis of host cells through amebapore (a pore-forming protein that resembles NK-lysin from natural killer T cells), [26], [27] and amebic phagocytosis and motility have all been implicated in tissue invasion and destruction by the organism. *E histolytica* can induce severe inflammation that can be confused clinically with inflammatory bowel disease. [28] The molecular mechanisms underlying amebic induction of gut inflammation and disease have recently been delineated. [29], [30] These studies suggest that *E histolytica* cysteine proteinases play a key role in amebic invasion of intestinal tissue and may stimulate gut inflammation by activating host interleukins-1 (IL-1). [25] Recent studies using human cell lines and a murine model of amebic liver abscess indicate that *E histolytica* trophozoites can also induce apoptosis in host cells. [31], [32] This may be particularly important for the pathogenesis of amebic liver abscess. The interaction of *E histolytica* trophozoites with the intestinal mucosa has been modeled in animals, [19], [33] colonic explants, [34], [35] and [36] intestinal cell lines, [37], [38] and [39] and human intestinal xenografts. [25], [40], [41] In addition to disruption and penetration of trophozoites through the epithelium, changes in epithelial permeability may occur before morphologic disruption, which could facilitate transport of amebic toxins or secretagogues across the epithelium before direct trophozoite invasion. [39] Ultrastructural studies have shown blunting of microvilli in regions where amebas are in close proximity to the brush border. [39], [42] Neutrophils in close proximity to amebas often exhibit a variety of degenerative changes, and it has been suggested that substances released from lysed neutrophils contribute to the tissue damage. This was confirmed by recent studies in human intestinal xenografts in severe combined immunodeficient mice, in which depletion of neutrophils from mice lessened intestinal xenograft damage early in amebic infection. [41] Trophozoites also may cause diarrhea by stimulation of intestinal secretion: amebic lysate applied to the serosal but not the mucosal side of stripped colonic mucosal preparations produces a rapid increase in anion secretion. [35], [43] *E histolytica* trophozoites introduced into human intestinal xenografts induce the production of the proinflammatory mediators IL-1, IL-8, and cyclooxygenase-2 by the intestinal epithelial cells. [25], [44] Cytokine production by intestinal epithelial cells is mediated by *E histolytica* activation of the transcription factor NF-?B, and blockade of NF-?B synthesis greatly reduces inflammation and tissue damage early in amebic infection. [41] These studies suggest that the inflammatory response, although critical for controlling *E histolytica* infection, may contribute to tissue damage early in disease.

Clinical Manifestations

Intestinal disease. The clinical manifestations of intestinal amebiasis range from asymptomatic infection to mild diarrhea to classic dysentery with abdominal pain, tenesmus, and bloody stools. [45], [46] The presentation is generally subacute, with most individuals symptomatic for less than 1 month. Dysentery is a hallmark of invasive amebic infection, and stools that are not grossly bloody almost invariably contain occult blood, consistent with the invasive pathophysiology. Rarely, patients may present with a more fulminant colitis with severe bloody diarrhea and fever. [47] These patients have diffuse abdominal tenderness but usually do not present with a rigid surgical abdomen, although up to 75% of these patients may have peritonitis secondary to leakage through the severely diseased colon. Fulminant colitis is associated with a high (more than 50%) rate of mortality. The risk for developing this syndrome is higher in children, pregnant women, and patients taking corticosteroids. Toxic megacolon is a rare but often fatal complication that is associated with administration of corticosteroids to patients with intestinal amebiasis. Unusual presentations of intestinal amebiasis include chronic nondysenteric infection—in which symptoms of diarrhea, abdominal pain, and weight loss persist for years—and appendiceal involvement. The differential diagnosis of invasive intestinal amebiasis includes infection with *Shigella, Campylobacter,* and other invasive bacteria, as well as pseudomembranous colitis secondary to *Clostridium difficile,* cytomegalovirus colitis, ischemic colitis, and inflammatory bowel disease. Because of the similarities in symptoms between amebiasis and inflammatory bowel disease, [28] amebiasis should be excluded by examination of stools or amebic serology in all patients before a diagnosis of inflammatory bowel disease is made and especially before corticosteroid therapy is begun. Rarely, a localized amebic colonic infection results in a segmental mass of granulation tissue forming an ameboma. Amebomas are found, in decreasing order of frequency, in the cecum, ascending colon, rectosigmoid, transverse colon, and descending colon, and they can be detected on physical examination as a tender palpable mass. The ameboma can be mistaken for a carcinoma on barium enema. Amebic strictures are most commonly observed in the anus, rectum, or sigmoid colon and must be differentiated from those due to lymphogranuloma venereum (chlamydia) or malignancy.

Extraintestinal disease. The most common manifestation of extraintestinal amebiasis is amebic liver abscess. [48], [49], [50] and [51] Patients often present with fever and right upper quadrant pain, leukocytosis, and abnormal liver test results, with an elevated alkaline phosphatase level the most common finding (more than 75% of individuals). Hyperbilirubinemia is uncommon, and severe jaundice may indicate bacterial superinfections of the abscesses. Concurrent diarrhea occurs in only one third of the patients. In about half the patients with amebic liver abscess, hepatomegaly and point tenderness of the liver are prominent on physical examination. Most patients have a single abscess, usually in the right lobe. Multiple abscesses are observed more commonly in patients with an acute presentation. With early diagnosis and treatment, the mortality of uncomplicated liver abscess is less than 1%. Most of the complications are related to rupture of the amebic liver abscess, which occurs most commonly into the chest (10% to 20%), but also into the peritoneum (2% to 7%) or pericardium (rare, often from left lobe abscess). The development of a bronchopleural fistula presents as a cough productive of the odorless, brown anchovy paste–like necrotic contents of the liver abscess. Amebas are usually not detectable in the necrotic material. Before the availability of effective antiamebic therapy, formation of such a fistula was a good prognostic sign for a spontaneous

cure because it provided drainage of the abscess. Amebic brain abscess occurs rarely but has a high associated rate of mortality. There have been case reports describing genitourinary involvement, including perinephric abscesses, splenic abscesses, infected rectal cysts, rectovaginal fistulas, cervical ulcers, uterine involvement, and vaginal lesions. [51] Cutaneous amebiasis is a rare complication and generally involves the perineum and genitalia as a complication of intestinal disease. [52]

Diagnosis The diagnosis (see Table 125-2) of intestinal amebiasis still rests primarily on microscopic demonstration of cysts or trophozoites in the stool or in scrapings or biopsy samples obtained by sigmoidoscopy or colonoscopy. Because organisms may be excreted intermittently or may be unevenly distributed in the fecal specimen, at least three stool specimens on separate days should be examined to detect 85% to 95% of the infections. Barium studies, laxatives, bismuth-containing preparations, antidiarrheal agents, and antibiotics can interfere with detection of the organism. Problems with microscopy include the misidentification of other *Entamoeba* species and leukocytes that have ingested red cells for *E histolytica* and the inability to distinguish between *E histolytica* and *E dispar*. In patients with mild bowel complaints, whose symptoms do not appear consistent with invasive amebiasis, care must be taken in the interpretation of a positive microscopic examination for amebic cysts or trophozoites. Fecal antigen detection assays that can distinguish between *E histolytica* and *E dispar* infections have been developed for commercial use, and their efficacy has been established in areas where disease is not endemic. [53], [54] and [55] Because *E dispar* infections are not associated with seroconversion, amebic serology may be a useful adjunct in assessing the patient, especially in areas where *E histolytica* is not endemic. Sigmoidoscopy should generally be performed in patients presenting with dysentery. Endoscopy should be performed without previous bowel preparation in order to increase the chance of detecting amebas in the colonic mucus. [56] Discrete shallow-based ulcers covered with yellow or white exudate with intervening areas of edematous mucosa are often found in invasive amebic colitis. However, the endoscopic appearance of amebic colitis may be indistinguishable from inflammatory bowel disease, diffuse mucosal erythema, granularity, and friability, or from pseudomembranous colitis secondary to *C difficile* infection, with pseudomembranes. [47] Multiple biopsy samples or scrapings should be taken from the ulcer edge, exudate, and intervening mucosa. [47], [56] The diagnostic yield from endoscopic biopsy samples or scrapings is high. Trophozoites are usually easily identified in these specimens by routine light microscopy, but immunohistochemical staining for *E histolytica* may further increase the yield. [47] Serology is an important tool in the diagnosis of amebic liver abscess and can help confirm the diagnosis of intestinal amebiasis. [57] Because antiamebic antibodies may persist for months and years after the eradication of infection, a positive serology requires more rigid clinical and diagnostic correlation in endemic areas. Indirect hemagglutination is the most sensitive assay, and it yields positive results in 90% to 100% of subjects with liver abscess, 75% to 90% of subjects with symptomatic intestinal infection, and 5% to 50% of subjects with asymptomatic infection. Enzyme-linked immunosorbent assay (ELISA) kits for the serodiagnosis of amebiasis are also available. Because serologic tests may be negative early in the course of an acute infection, it is important to repeat the serology 5 to 7 days later. Recombinant antigens have been used for diagnosis and may offer improved specificity for acute infection. [58], [59] and [60] An imaging procedure should be performed in all suspected cases of amebic liver abscess. Right upper quadrant ultrasonography is usually the least expensive initial procedure. The usual appearance is a round or oval-shaped hypoechoic lesion with no wall echoes. On abdominal computed tomography, the abscesses usually appear as well-defined low-density lesions. The differential diagnosis of space-occupying lesions in the liver includes bacterial (pyogenic) liver abscess, hydatid cyst, or tumor. Amebic serology usually establishes or excludes the diagnosis of amebic liver abscess. Ultrasonographic or computed tomography–guided needle aspiration of the abscess can be performed to rule out a pyogenic abscess if the results of serology are nondiagnostic or if the clinical condition of the patient requires an immediate diagnosis. Concomitant pyogenic and amebic infections are unusual at the time of the initial presentation, but bacterial superinfection may occur after repeated aspirations or open drainage. Amebas are rarely recovered in the aspirate. Because of the risk for spillage and anaphylaxis, aspiration should be avoided if there is suspicion of a hydatid cyst.

Treatment Patients with *E histolytica* infection should receive antiamebic therapy, but there is no evidence that patients with *E dispar* infection require treatment. [46], [61] If one does not have access to assays that distinguish between infection with *E histolytica* and *E dispar,* treatment decisions must be based primarily on the clinical presentation. The goals of treatment are to cure invasive disease and eradicate intestinal carriage of the organism. Metronidazole is the drug of choice for patients with invasive intestinal disease or amebic liver abscess (Table 125-3). Metronidazole is highly efficacious (90% cure rate in many studies) and is relatively well tolerated. Despite widespread use of this drug, *E histolytica* resistance to metronidazole has not been a problem to date. Intestinal disease is generally treated for 5 to 10 days. Antibiotic therapy of liver abscess is similar; however, 2.4 g of metronidazole taken as a single dose for 2 days has been reported to be more effective in a small group of patients. [62] Common side effects of metronidazole include nausea, headache, metallic taste, and abdominal discomfort; ataxia, confusion, insomnia, and paresthesias may occasionally occur. Patients should avoid alcoholic beverages owing to metronidazole's disulfiram-like properties. The most serious side effects are central nervous system effects (psychosis, seizures), which mandate cessation of the drug. Rapid recurrence of these symptoms after readministering the drug has been reported. [63] The drug is available for intravenous administration in patients unable to take medications by the oral route. Other nitroimidazoles—tinidazole and ornidazole—may have less toxic effects but are not currently available in the United States. Tinidazole is reported to be effective as a single dose. [64]

DRUG	ADULT DOSAGE	PEDIATRIC DOSAGE
Lumenal Agents[1]		
Paromomycin (Humatin)	30 mg/kg/d orally × 7 days in 3 divided doses	25 mg/kg/d orally × 7 days in 3 divided doses (maximum, 2 g/d)
Iodoquinol (Yodoxin)	650 mg orally tid × 20 days	30–40 mg/kg/d × 20 days in 3 divided doses (maximum, 2 g/d)
Agents for Invasive Amebiasis[2]		
Metronidazole	750 mg orally tid × 5–10 days 500 mg IV every 6 h × 5–10 days	30–50 mg/kg/d × 5–10 days orally in 3 divided doses; 15 mg/kg IV load followed by 7.5 mg/kg every 6 h (maximum, 2250 mg/d)
Dehydroemetine[3] (Mebadin)	1–1.5 mg/kg/d IM × 5 days (maximum, 90 mg/d)	1–1.5 mg/kg/d IM × 5 days (maximum, 90 mg/d)

[1]Lumenal agents are used primarily for the treatment of nondysenteric amebiasis and to eradicate intestinal colonization after treatment of invasive amebiasis (colitis and liver abscess).

[2]Treatment of amebic colitis, liver abscess, and other invasive disease is followed by a complete course of a lumenal agent to eliminate intestinal colonization.

[3]Available only through the Centers for Disease Control and Prevention: telephone, 404-639-3670 or 404-639-3356; nights and emergencies, 404-639-2888.

tid, three times daily; IM, intramuscularly.

TABLE 125-3 Antibiotic Therapy for Amebiasis

In the critically ill patient with fulminant amebic colitis, there may be some advantage in adding a second agent, dehydroemetine, for the first 2 to 3 days of therapy. Although there are no controlled trials to support such combination therapy, dehydroemetine offers a rapid amebicidal effect that could be beneficial in severe disease. The short course of therapy is designed to lessen the risk for cardiotoxicity. Dehydroemetine, which is preferred to the parent compound, emetine, because it may be less toxic, is available only from the Centers for Disease Control and Prevention. Because bacterial peritonitis resulting from leakage of the diseased colon is a frequent complication of fulminant amebic colitis, antibacterial therapy should also be initiated in these patients. [46] As in other patients with dysentery, antidiarrheal agents should not be administered. After treatment of invasive intestinal or extraintestinal amebic disease is complete, a lumenal agent is generally begun to eradicate intestinal carriage of the organism. Three drugs have proven efficacy as lumenal agents: diloxanide furoate (no longer available in the United States), paromomycin, and iodoquinol (see Table 125-3). The side effects of paromomycin are diarrhea and gastrointestinal side effects. Rare dose-related neurological toxicity, including optic neuritis, has been reported with iodoquinol. It is controversial whether a lumenal agent is necessary in conjunction with treatment with metronidazole because courses of metronidazole of 10 days or longer may be effective in eradicating intestinal carriage. [65] Because about 10% to 15% of patients treated with any of these agents fail to eradicate intestinal carriage, a follow-up stool examination after completion of therapy is recommended. Most cases of colonic and extracolonic amebiasis can be managed medically. Even in cases of fulminant colitis, it is unclear whether these patients benefit from surgery unless toxic megacolon is present. [46] Further interventions are generally reserved for prevention of complications resulting from perforation of the colon and rupture of liver abscesses. Possible indications for aspiration of an amebic liver abscess as an adjunct to medical therapy include extremely large abscesses in which rupture is felt to be imminent, abscesses in the left lobe of the liver (because of the higher risk for rupture into the pericardium), and treatment failure, in which fever and pain persist after 3 to 5 days of appropriate therapy. Aspiration can generally be accomplished percutaneously. Surgical drainage should be reserved for large left lobe abscesses that cannot be reached percutaneously. It is important to note that amebic liver abscess cavity may resolve slowly and that successfully treated abscess cavities may increase in size over the first few weeks. Most abscesses resolve by 6 months, but 10% of patients may have abnormal ultrasonographic findings more than 1 year after therapy. The persistence of a cavity alone is not an indication for aspiration or for administering another course of antibiotics. Positive ameba stool examinations may be encountered in patients with minimal bowel complaints as well as in AIDS patients with diarrhea who do not have symptoms or findings consistent with invasive disease. Most of these cases represent infection with *E dispar,* rather than *E histolytica,* and do not require therapy. However, it is recommended that asymptomatic cyst passage of pathogenic strains of *E histolytica* be treated because these patients may subsequently develop invasive disease or transmit the organism to close contacts. At the present time, our recommendation is to treat all patients in areas of nonendemicity whose stool examination results are positive, particularly because stool examinations are rarely conducted in the absence of any intestinal symptoms. This practice may change after widespread acceptance of tests that can differentiate between *E histolytica* and *E dispar*. Despite legitimate concerns about administering metronidazole in pregnancy, women who develop symptomatic amebic colitis during pregnancy should receive metronidazole because of their increased risk for fulminant colitis. A long-term follow-up study of women given metronidazole for therapy of trichomonas during pregnancy reported no adverse outcomes. [66] However, in the case of the pregnant woman with a positive stool

examination result, but without evidence of invasive disease (a setting in which infection with *E dispar* is most likely), treatment with paromomycin, a nonabsorbable aminoglycoside, could be initiated with close follow-up of the patient. [67]

Giardia lamblia

Giardia lamblia (also termed *intestinalis* and *duodenalis*) is a flagellated protozoan initially described by Van Leeuwenhook in 1681 and by Lambl in 1859. The clinical spectrum of human giardiasis includes asymptomatic carriage, acute self-limiting diarrhea, and, rarely, persistent diarrhea that may fail to respond to appropriate therapy even in immunocompetent individuals. *Giardia* infection can cause intestinal malabsorption, and in children, this may be associated with retardation of growth and development.

Giardia species represent one of the most primitive of all eukaryotes. [68] The life cycle consists of two stages: the trophozoite and the cyst (Fig. 125-3). Infection is initiated by ingestion of as few as 10 to 100 cysts. [69] The cysts excyst in the proximal small intestine, releasing two trophozoites that multiply by binary fission. Trophozoites attach to the intestinal mucosa through an attachment organelle called the *ventral disk*. The life cycle is completed by encystation, an event that may be induced by exposure to intralumenal conjugated bile salts or decreasing levels of cholesterol. [70], [71]

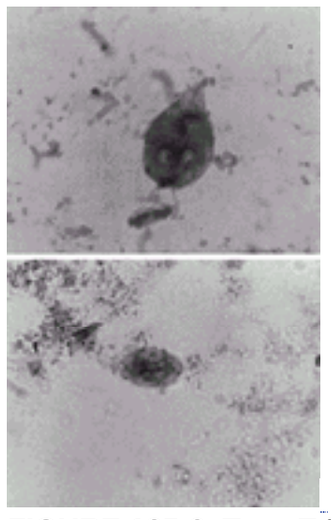

FIGURE 125-3. Top: Trichrome stain of a *Giardia lamblia* trophozoite, demonstrating the characteristic facelike image secondary to the two nuclei, each containing a prominent karyosome and four symmetrically placed flagella. **Bottom:** Trichrome stain of a *Giardia lamblia* cyst, 7 to 10 µm in length, and ovoid in shape. (Courtesy of Patrick Murray, Ph.D., St. Louis, MO.)

Epidemiology *Giardia* is cosmopolitan in distribution, and prevalence rates are estimated to be as high as 2% to 5% of populations in the industrialized world and a staggering 20% to 30% of individuals in some regions of the developing world. *Giardia* is now the most frequently identified intestinal parasite in the United States and was the most commonly implicated pathogen in water-borne disease outbreaks between 1978 and 1991. [2] Although recognized as a common pathogen, it is less well appreciated that *Giardia* can cause illness serious enough to require admission to the hospital, with the number of admissions for giardiasis rivaling that for shigellosis. [72] High-risk groups for giardiasis include infants and young children, particularly those attending day-care centers, individuals who have traveled outside the United States, people exposed to infected water sources, and homosexual men. [73] The prevalence of *Giardia* infections among children in day-care centers varies from 17% to 90%. Day-care centers are emerging in the United States as a major site of endemic *Giardia* infection and as a nidus for spread of the organism to the surrounding community. [74] *Giardia* has been implicated in multiple outbreaks of diarrheal illness associated with ingestion of contaminated drinking water and recreational water sources. [1], [75] Many of these outbreaks were associated with unfiltered water systems in which the only treatment was chlorination, a tribute to the ability of the cysts to survive in fresh water and their relative insensitivity to chlorination. Contamination of recreational water sources may occur from animals; strong epidemiologic data, and more recently genetic data, indicate that giardiasis is a zoonosis. [76], [77] This provides an immense reservoir of infection that can make disease control more difficult. Transmission of *Giardia* does not occur exclusively by ingestion of contaminated water; food-borne transmission has been described in several outbreaks.

Pathogenesis and Pathology Despite intensive investigation, the pathogenesis of human giardiasis is poorly understood. Adherence to intestinal epithelial cells is a critical step in infection. [78] Adhesion is inhibitable by drugs that block actin polymerization and microtubule formation, which interfere with attachment by the ventral sucking disk of the parasite. Adhesion is also inhibited by D-mannose and other mannose derivatives, suggesting that parasite adherence may be mediated in part by *Giardia* lectin. [79], [80] and [81] Inhibition of *Giardia* adherence to artificial substrates by human neutrophils and monocytes indicates one possible mechanism by which host innate immunity could alter infection. [82] How *Giardia* actually causes diarrhea remains unclear. Animal models suggest a secretory mechanism, with net secretion occurring by a Ca^{2+}-dependent mechanism, [83], [84] malabsorption secondary to a decrease in brush-border surface area, [85] and local zinc depletion by *Giardia* cysteine-rich proteins resulting in interference with intralumenal digestion. [86], [87] Malabsorption of fat and vitamins, including vitamin A, vitamin B_{12}, and folate, has been documented in some patients. [88] The extent of villous atrophy seen in histological sections has correlated with the severity of diarrhea and malabsorption in some reports. [89] However, in most individuals with giardiasis, minimal, if any, changes are seen in biopsy samples by light microscopy, although distortion of microvilli has been observed at the ultrastructural level. [90] It has been suggested that malabsorption associated with giardiasis may be associated with reductions in intralumenal bile salt concentration and intralumenal enzyme levels. [76] Infecting strains may differ in their virulence. [91], [92] One phenotypic difference between *Giardia* strains is their susceptibility to host intestinal proteinases, and resistance may depend on the form of variant surface protein expressed by the parasite. [93] Host factors also play a role in whether infection with *Giardia* results in disease. Killing of *Giardia* trophozoites exposed to human milk is dependent on the presence of bile salt–stimulated lipase. [94] Host macrophages can phagocytose *Giardia* trophozoites, suggesting that cellular immunity could play some role in controlling disease. [95] There is strong experimental and clinical evidence that secretory antitrophozoite antibodies are important and contribute to clearance of the parasite from the intestinal tract. [96] For example, individuals with immunoglobulin deficiencies are at significantly higher risk for chronic *Giardia* infection. [97], [98] Although recent animal studies suggest that cell-mediated immunity could play a role in host defense against *Giardia,* [99] the clinical course of giardiasis does not appear to be altered in patients infected with the human immunodeficiency virus (HIV). Ingestion of maternal antibody in breast milk appears to provide infants with protection from symptomatic disease but does not stop the establishment of *Giardia* infection. [100]

Clinical Manifestations Studies of human volunteers suggest that the incubation period for symptomatic disease is 1 to 2 weeks. [69], [92] It is important to note that the onset of symptoms may precede the excretion of cysts in stool by more than 1 week. [101] The clinical presentation is varied and can be classified in one of three syndromes: asymptomatic carrier state, acute self-limiting diarrhea, and chronic diarrhea. Asymptomatic infection is probably the most common outcome of *Giardia* infection. [102], [103] In individuals who develop symptoms, the most common presentation is that of diarrhea with watery, foul-smelling stools, often with abdominal distention, flatulence, nausea, flatus, anorexia, and vomiting. Blood and mucus in the stool are rare. Low-grade fever also may be observed, and in some individuals, fatigue and malaise or headache may be prominent. Although infection is self-limiting in most healthy individuals, a significant proportion (25% to 30%) come to medical attention because of chronic diarrhea, often with features of steatorrhea and weight loss. Malabsorption of vitamins A and B_{12}, proteins, and D-xylose is not unusual, and more than 50% of symptomatic individuals report weight loss of greater than 10 pounds. [103], [104] It is important to note that secondary lactase deficiency may persist for several weeks after eradication of *Giardia* infection, necessitating the dietary restriction of lactose. This can be confused with recurrent or relapsing infection. Protracted disease is more frequent in young children and in those individuals with an underlying humoral immunodeficiency syndrome and nodular lymphoid hyperplasia. This syndrome is most commonly associated with patients with common variable immunodeficiency and occasionally with selective immunoglobulin A deficiency. [97], [105] However, *Giardia* infection also has been associated with nodular lymphoid hyperplasia in the absence of overt humoral immunodeficiency. [106] Extraintestinal disease is rare, but there are case reports of individuals with gastritis, reactive arthritis, biliary tract disease, and urticaria. [107], [108] and [109] Gastric giardiasis may be seen in patients with achlorhydria.

Diagnosis The diagnosis of *Giardia* infection (see Table 125-2) should be considered in any patients with prolonged diarrhea, particularly if they have been exposed to children in day care, were exposed to potentially contaminated water, or report a history of recent travel. The diagnosis has been based primarily on detection of *Giardia* cysts or trophozoites in stool or of trophozoites in the upper intestine. Examination of a single stool may detect 50% to 70% of cases, and sensitivity increases to 85% when three concentrated stools collected on different days are examined. Because symptoms may precede excretion of stools by more than 1 week, one stool should be obtained 1 to 2 weeks after the initial one. Even with the examination of three stools, however, some infections may be missed owing to periodic excretion

of cysts, with as much as 20 days elapsing between positive stools. The prior use of antibiotics, antacids, and antidiarrheals may interfere with detection of the organism. Barium studies are not useful and may even hamper detection of the organism in stool. The experience of the laboratory staff in the microscopic recognition of *G lamblia* also has an important impact on the sensitivity of microscopic examination of stools. A direct fluorescent antibody test, now being used in clinical laboratories, allows for the detection of a low number of organisms in a shorter period of time and can facilitate the differentiation of *G lamblia* cysts from other cysts and cystlike bodies. [110] ELISAs for the detection of *Giardia*-specific antigens in fecal specimens appear to have specificity and sensitivity rates that compare favorably with microscopy. [111], [112] and [113] When the diagnosis cannot be made by examination of stool and the index of suspicion for *Giardia* infection is high, intestinal aspiration, intestinal biopsy, or brush cytology may be indicated. One advantage of biopsy is that it may provide additional information regarding associated bowel pathology, such as nodular lymphoid hyperplasia. An alternative approach is to treat empirically for giardial infection before subjecting the patient to more invasive tests. The nonspecificity of symptoms associated with chronic *Giardia* infection and the similarity to symptoms observed in irritable bowel syndrome may contribute to delayed and missed diagnoses. The differential diagnosis includes infection by other protozoan organisms, enteric viruses, and noninvasive bacteria. In patients with malabsorption, the differential diagnosis includes other causes of steatorrhea and malabsorption as well as inflammatory bowel disease. Serology has not proved helpful in the diagnosis of giardial infections.

Treatment Metronidazole and tinidazole (not available in the United States) are currently the drugs of choice (Table 125-4) for the treatment of giardiasis. Quinacrine exhibits similar efficacy but is no longer available within the United States. Furazolidone is somewhat less effective than metronidazole, but it is available in a liquid preparation that is widely used in children in the United States. More recently, albendazole has been shown to exhibit some efficacy in the treatment of giardiasis, and it may prove useful for administration to children in undeveloped countries for the simultaneous treatment of intestinal helminths and *Giardia.* [114] None of the above agents are recommended for use during pregnancy. Paromomycin, a nonabsorbable aminoglycoside with antigiardial activity, has been suggested as a possible drug for use in pregnant women.

	ADULT DOSAGE	PEDIATRIC DOSAGE
Metronidazole	250 mg tid × 5–7 days	5 mg/kg/d × 10 days
Tinidazole	2 g single dose	50–75 mg/kg single dose
Quinacrine	100 mg tid × 5–7 days	2 mg/kg tid × 5–7 days
Furazolidone	100 mg qid × 7–10 days	2 mg/kg tid × 7–10 days

tid, three times daily; qid, four times daily.

TABLE 125-4 Drug Treatment of Giardiasis

A major problem in treating patients with *Giardia* infection is the recurrence of symptoms after standard courses of therapy. This may be secondary to persistent infection because of resistant organisms, or may occur in patients in whom reinfection cannot be documented and in whom an immunodeficiency, such as hypogammaglobulinemia, is not present. [115], [116] One should consider secondary lactase deficiency, which may persist despite eradication of infection. Relapse of symptoms after an initial course of therapy is frequent enough that some physicians have adopted the practice of routinely prescribing a second course of therapy. Patients who have persistent symptoms after single-drug therapy may benefit from combined therapy. [117], [118] Our current approach to individuals with recurrent symptoms after treatment is to give a second course of metronidazole (250 mg three times daily for 10 to 14 days), whether or not a microbiologic diagnosis of recurrent infection can be made. If symptoms persist after this second course of treatment, and lactase deficiency is ruled out, stools are examined again for *Giardia* parasites. If parasites are detected, a search for possible immunodeficiency is made, and combination therapy with metronidazole and furazolidone is administered. Although symptomatic patients infected with *Giardia* should receive therapy, the treatment of asymptomatic carriers is not routinely recommended. This particularly pertains to control of infection within day-care centers. A longitudinal study of asymptomatic children in a day-care center indicated that excretion of *Giardia* cysts is well tolerated, and in the setting of a high rate of reinfection, treatment of asymptomatic children may lead to recurrent courses of therapy of potentially toxic drugs. [119]

Blastocystis hominis

Blastocystis hominis is a commonly detected intestinal protozoan parasite, found in 10% to 25% of stools examined [120] (see Table 125-2). The pathogenicity of *B hominis* continues to be controversial. [120], [121], [122], [123], [124] and [125] Mild diarrhea, nausea, anorexia, and fatigue may be seen in those individuals with a high parasite load. However, in a recent study, symptomatic patients with high concentrations of *Blastocystis* parasites [more than 10 organisms per high power (400×) field] were more likely to be co-infected with other enteric pathogens. [122] This may explain why the treatment of *Blastocystis* with iodoquinol and metronidazole appears to be more effective in relieving the symptoms of infected patients than in eliminating the infection. [123] Alternatively, there may be distinct strains of *B hominis* differing in their pathogenic potential, which could explain the poor correlation between infection and symptomatic disease. A recent study suggested that *Blastocystis* infection was associated with disease only in immunocompromised individuals. [124] At this point, symptomatic *Blastocystis* infection should be considered a diagnosis of exclusion, and if symptoms persist after therapy, there should be a further investigation for other causes of intestinal disease. [125] Optimum therapy has not been established, but metronidazole, iodoquinol, and furazolidone have efficacy in anecdotal reports.

Dientamoeba fragilis

Dientamoeba fragilis is a flagellated protozoan parasite that is being increasingly identified in stool samples [126] (see Table 125-2). There is no known cyst form of *D fragilis,* and recent studies suggest that it shares a common evolutionary history with trichomonads. [127] Symptoms ascribed to *D fragilis* infection include mild diarrhea, abdominal pain, anorexia, and fatigue. [128] Fever, irritability, weight loss, nausea, and vomiting are occasionally reported, and an associated eosinophilia has been described. [126] Co-infection of *D fragilis* and the pinworm, *Enterobius vermicularis,* is nine times higher than expected based on random distribution of each organism, increasing the possibility that pinworm eggs or larvae could be responsible for the transmission of *D fragilis* infection. [129] The pathogenic potential of this organism remains uncertain, but symptomatic individuals and experimentally infected volunteers become free of symptoms when it is eradicated. [128], [130] Adults should be treated with diiodohydroxyquin (650 mg three times daily for 3 weeks), or tetracycline (500 mg four times daily for 1 week) as an alternative. [130], [131] In children, metronidazole is recommended (30–50 mg/kg/day orally in three divided doses for 5 to 10 days). [131] Paromomycin (25 mg/kg/day orally in three divided doses for 5 to 7 days) is also a reasonable alternative.

Balantidium coli

Balantidium coli is a ciliated protozoan parasite and is a rare cause of intestinal disease (balantidiasis) in humans (see Table 125-2). Disease outbreaks in humans are associated with close exposure to pigs and their feces. [132] Outbreaks in institutions associated with poor hygiene or coprophagia have also been reported, [133] as have water-borne outbreaks. Humans are probably relatively resistant to *B coli* infection, but malnutrition, achlorhydria, and poor hygiene predispose to disease. The spectrum of disease ranges from asymptomatic carriage, to watery diarrhea, to active colitis with bloody, mucus-laden stools. *B coli* infection is also a rare cause of appendicitis. [134], [135] *B coli,* like *E histolytica,* can invade the colonic mucosa and spread through submucosal layers, causing colonic ulcerations. Misdiagnosis as ulcerative colitis has been reported. [136] Rarely, colonic perforation occurs with resultant peritonitis. [137] Extraintestinal disease, with lung or liver involvement, is a rare complication. [136] Diagnosis is made by observing the motile trophozoites in stool saline mounts under low-power microscopy. [132] Cysts are much less frequently seen in human infections. Optimum therapy remains uncertain, but tetracycline (500 mg four times daily for 10 days) is recommended by most authorities. Iodoquinol (650 mg three times daily for 3 weeks) is an appropriate alternative.

INTRACELLULAR PROTOZOAN PARASITES (COCCIDIA)

The AIDS epidemic has played an important role in the recognition of Coccidia (phylum Apicomplexa, class Sporozoasida, subclass Coccidiasina) as important gastrointestinal protozoal pathogens. Organisms from the genera *Cryptosporidium, Cyclospora,* and *Isospora* can cause an acute, self-limited diarrheal illness in immunocompetent hosts and a severe chronic diarrhea in patients with AIDS and other forms of immune dysfunction. The similar life cycles of these organisms may account for their similar clinical presentations. *Cryptosporidium, Cyclospora,* and *Isospora* are all intracellular pathogens that can replicate asexually within the enterocyte to produce a merozoite stage, which can autoinfect other enterocytes, leading to a rapid spread of the infection within the epithelium. All three pathogens also undergo a sexual stage that leads to the generation of cysts or spores, which are sloughed into the gut lumen and excreted in the stool. For *Cryptosporidium,* formation of the infectious sporozoite can take place within the host, which can autoinfect other enterocytes. However, for *Cyclospora* and *Isospora,* formation of the infectious sporozoite must take place outside of the host.

Of note, not all laboratories routinely include screening for coccidial organisms in their analysis of stool samples for parasites. Organisms of the three genera *Cryptosporidium, Cyclospora,* and *Isospora* possess distinct morphologic characteristics, although it may be difficult to distinguish cryptosporidia from *Cyclospora*

organisms (see Table 125-2). The modified acid-fast stain may be the most useful for distinguishing among the three genera (Fig. 125-4). The distinction is important because infections with *Cyclospora* and *Isospora,* but not *Cryptosporidium,* respond well to antibiotic treatment.

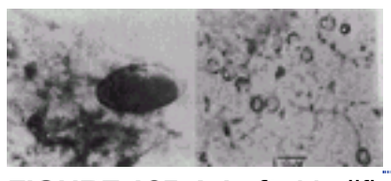

FIGURE 125-4. Left: Modified Kinyoun stain of *Isospora belli* cyst in stool. Note that the ellipsoid shapes are larger (20–33 × 12–15 µm) than the oocysts of *Cryptosporidium* or *Cyclospora* (4–10 µm). Immature oocysts contain a single sporocyst; mature oocysts contain two. (**Right**) Modified Kinyoun stain of *Cryptosporidium* oocyst in stool. (From DeHovitz JA, Pape JW, Boncy M, Johnson WD. Clinical manifestations and therapy of *Isospora belli* infection in patients with the acquired immunodeficiency syndrome. N Engl J Med 1986;315:88.)

Microsporidia are spore-forming protozoa that have been recognized as causing widespread infection in patients with AIDS (see Chapter 124). Because they do not appear to be significant pathogens in immunocompetent hosts, they are not discussed further in this chapter.

Cryptosporidium parvum

Cryptosporidium species were originally described in the stomach and intestine of asymptomatic mice by Tyzzer in 1907. [138] *C parvum* was recognized as a cause of diarrhea in animals but was not identified as a human pathogen until 1976. [139], [140] Initially, most of the reported *C parvum* infections were in patients with AIDS. [141] However, with improved methods of stool diagnosis and better understanding of the clinical disease, cryptosporidiosis has emerged as an important cause of diarrhea in normal hosts as well. The importance of *Cryptosporidium* infection as a public health problem in the United States was underscored by several recent large water-borne outbreaks of disease. *C parvum* is considered the predominant species causing disease in humans but is capable of infecting all species of mammals. [142]

Epidemiology Human cryptosporidiosis is found worldwide in both industrialized and developing countries and in both rural and urban areas. The parasite is transmitted by ingestion of oocysts that are excreted in the feces of infected humans or animals, from person-to-person spread, by ingestion of contaminated food and water, and, as a zoonosis, by contact with infected (primarily farm) animals. Direct person-to-person transmission may occur by fecal-oral spread related to sexual practices. The prevalence of *Cryptosporidium* infection is highest in young children and immunocompromised individuals. The stool positivity rate is higher in developing countries (1% to 22%) than in industrialized countries, where cryptosporidia are identified in the stool of 0.3% to 4% of individuals with diarrheal illnesses. [143], [144], [145] and [146] In developing nations, there is a high rate of asymptomatic carriage, and in some studies, the prevalence of stool positivity for *C parvum* in control patients approached or exceeded that observed in children with diarrhea. [145] Based on serologic studies, exposure to *Cryptosporidium* is relatively common even in industrialized countries, with as many as 15% to 36% of individuals seropositive. [147], [148], [149] and [150] The highest prevalence rates in these studies were seen in individuals with occupational exposures, such as dairy farmers. [150] In developing countries, the seroprevalence is even higher, with 65% to nearly 100% of individuals possessing antibodies to *C parvum* in portions of Latin America. [151], [152] Cryptosporidiosis is among the most common causes of diarrhea in patients with AIDS, with an annual incidence of 5% to 10%. [142] Severe cases of cryptosporidiosis also can be seen in individuals with other forms of immunodeficiency, such as hypogammaglobulinemia, and in those patients who are receiving immunosuppressive drugs for malignancies, autoimmune diseases, or organ transplantation. [145] The reader is referred to Chapter 124 for a more detailed discussion of *Cryptosporidium* infection in immunocompromised hosts. In developing countries, a significant portion of disease transmission may arise from person-to-person spread within families, with transmission rates comparable to those of *Shigella.* [152] In industrialized countries, day-care centers represent a source for outbreaks and sporadic infections. A cross-sectional study showed that 1.8% to 3.8% of children in day-care centers are infected, but rates are much higher (27% to 59%) among children in centers experiencing an outbreak of *C parvum* disease. [153] Infections occur primarily in diaper-aged children, who serve as a potential nidus for secondary infection of the surrounding community. The risk for secondary transmission within a household is low (5%) when the index case involves an adult. [154] Person-to-person spread within the hospital setting also has been reported. [155] Water-borne outbreaks of *C parvum* are a major threat to public health. *Cryptosporidium* was identified as the agent responsible for eight water-borne disease outbreaks involving drinking water between 1991 and 1994. [1], [156], [157] In 1993, an estimated 403,000 people in the Milwaukee area became infected with *C parvum* due to a failure in water treatment and purification. [157] This represents the largest community outbreak of diarrhea due to any defined organism ever reported in the United States. [154], [157] *C parvum* also has been identified as the causative agent in water-borne outbreaks associated with swimming pool water. [158], [159] These outbreaks underscore the resistance of *C parvum* oocysts to chlorination and the difficulties in removing the organism by filtration. Food-borne outbreaks of cryptosporidiosis have been reported; in several instances, the outbreaks were associated with ingesting unpasteurized apple cider. [160], [161]

Pathogenesis and Pathology The mechanisms by which cryptosporidia cause intestinal disease are still poorly defined and include disruption of the epithelial barrier, [162], [163] resulting in malabsorption, [164] induction of inflammatory mediators, or release of parasite enterotoxins that stimulate intestinal secretion. [165], [166] However, no product of *C parvum* has yet been identified that possesses enterotoxic or cytotoxic activity. On histological examination of intestinal biopsy specimens from individuals with *C parvum* infection, a variety of nonspecific changes can be seen, including villous atrophy (partial or complete) and crypt hyperplasia with increased numbers of lymphocytes, macrophages, plasma cells, and, less commonly, neutrophils in the lamina propria. [167] However, many patients show normal duodenal biopsy specimens, despite chronic diarrhea. [167] There appears to be an association between the extent of malabsorption and intestinal injury and the intensity of the infection (based on oocyst excretion). [168] Recent studies in a human intestinal xenograft model of disease and in human volunteers have shown that *C parvum* infection induces production of tumor necrosis factor-a from intestinal cells, but the contribution of cytokine production to disease remains unclear. [169], [170] and [171] The prolonged infection in immunocompromised patients and in animal models suggest that CD4 (helper) T cells and interferon are critical to the control of infection. [172], [173] and [174] One of the remarkable consequences of the availability of highly active antiretroviral therapy has been a significant reduction in the cases of cryptosporidiosis seen in HIV-infected individuals. [175]

Clinical Manifestations The clinical spectrum of the disease is wide and includes asymptomatic carriage, a self-limited diarrheal illness in immunocompetent individuals, and a relentless, severe, watery diarrhea in some immunocompromised individuals. In a study of 29 healthy volunteers, the median infective dose was estimated to be 132 oocysts, with a mean incubation period of 9 days (range, 5 to 22 days). [148] As few as 30 oocysts caused infection in some subjects. The infectivity was therefore comparable to the infectivity of *Giardia* cysts. Subjects with diarrhea excreted more oocysts over the course of the infection than did subjects without diarrhea. [176] The predominant symptom in intestinal cryptosporidiosis is diarrhea, which is often accompanied by abdominal cramps and varies greatly in severity from individual to individual. Some patients have scant, intermittent diarrhea, whereas others (more often, immunocompromised individuals) experience voluminous watery diarrhea that can approach the quantities seen with cholera (more than 10 L/day). Mucus may be present in stools, but bloody diarrhea is rare, and white blood cells are usually not found in stools from patients with cryptosporidiosis. Accompanying symptoms may include nausea, fatigue, general malaise, anorexia, vomiting, and low-grade fever. The duration of illness in immunocompetent individuals ranged from 2 to 26 days in one study, [177] with a mean of 12 days, but more prolonged illness is not unusual. [152] Transient relapse of watery diarrhea has been observed in immunocompetent hosts, particularly in the massive outbreak of water-borne *Cryptosporidium* infection that occurred in Milwaukee. [154] Extraintestinal disease is rare in immunocompetent individuals, but pancreatitis and cases of reactive arthritis have been reported in previously healthy adults. [178], [179] Cholecystitis is seen in as many as 10% of patients with cryptosporidiosis and AIDS but is rare in immunocompetent patients. The differential diagnosis for cryptosporidiosis in immunocompetent hosts includes other protozoan infections (e.g., *C cayetanensis, G lamblia,* and *I belli*), bacterial infection (e.g., *Salmonella, Shigella, Campylobacter, Yersinia, C difficile,* and enterotoxigenic *E coli* in patients with a history of travel to developing countries), and viral infections (e.g., rotavirus and Norwalk viruses).

Diagnosis The diagnosis of *Cryptosporidium* infection should be suspected in individuals with diarrhea lasting longer than 3 days and currently rests primarily on the identification of the oocysts in stool samples [180] (see Table 125-2 and Fig. 125-4). It is important to make a diagnosis in the interest of public health, although disease is generally self-limiting in immunocompetent hosts. Patients should be questioned with respect to recent travel, exposure to children in day care, their drinking water, ingestion of unpasteurized milk or fruit juices, exposure to recreational water, and exposure to animals, particularly cattle. Stool specimens from individuals suspected of having cryptosporidiosis should be preserved in formalin as soon as possible and should be concentrated by either a sugar flotation or formalin-ethyl acetate method. At least three different stool concentrates and five or six acid-fast stained samples should be examined before the diagnosis of cryptosporidiosis is excluded. There are now immunology-based tests that may facilitate the diagnosis, especially in settings in which skilled microscopists are not available [146], [181] (see Table 125-2). In most patients with diarrhea and suspected cryptosporidiosis, stool examination, as outlined earlier, is probably as sensitive as small intestinal biopsy and is the diagnostic procedure of choice. [182], [183] For those unusual cases in which *Cryptosporidium* infection is suspected, but the major symptoms are abdominal cramps or

abdominal pain without significant diarrhea, stool specimens are more likely to be negative, and a duodenal aspirate with biopsy, combined with lower endoscopy and terminal ileum biopsy, may have a higher diagnostic yield. [183]

Treatment Effective treatment for cryptosporidiosis remains an elusive goal. For immunocompetent individuals, the disease is self-limiting, and symptomatic treatment (replacement of lost fluids) is the cornerstone of therapy. Nonspecific antidiarrheal therapies, such as antimotility agents, may provide some relief for individual patients. A recent study in patients with AIDS suggested that combination oral therapy with azithromycin (600 mg daily) and paromomycin (1 g twice daily) may have some efficacy, but not all patients responded. [184]

Cyclospora cayetanensis

In the early 1990s, diarrhea associated with an organism variously described as a large *Cryptosporidium,* blue-green algae, cyanobacterium-like body, or coccidian-like body was reported. [185], [186], [187], [188] and [189] The organism was identified as a *Cyclospora* species by Ortega and colleagues, [190] who successfully induced sporulation and excystation of oocysts obtained from individuals with diarrhea. Electron microscopic studies of the organisms in enterocytes revealed that they possessed morphologic features of coccidians of the phylum Apicomplexa, [191] and a molecular phylogenetic analysis confirmed that *Cyclospora* is a coccidian parasite closely related to protozoa of the *Eimeria* genus, a known pathogen of birds. [192]

Epidemiology *Cyclospora* species are distributed widely—the organism has been implicated as the causative agent in diarrheal illnesses in the Americas, Southeast Asia, the Indian subcontinent, the Caribbean, Africa, Australia, and Europe. [1] Originally, disease was described primarily in travelers to endemic areas, [188], [193], [194] but with greater awareness of the parasite, indigenous disease in endemic areas and outbreaks in areas of low endemicity are being more widely recognized. [195], [196] and [197] Prevalence rates as high as 11% have been reported in nonnative residents of an endemic area, and 18% of young children (1 month to 2 years of age) in poor regions of Peru had oocysts in their stool. [196], [198] In the United States, surveys of stool specimens suggest a low prevalence of only 0.3% to 0.5%. [199], [200] and [201] The ingestion of water containing sporulated oocysts of *Cyclospora* is the major mode of transmission, [185], [197], [202], [203] but food-borne outbreaks also have been described. [204], [205] In the United States and Canada, a major outbreak of *Cyclospora* infection (1500 cases involving 20 states, the District of Columbia, and two provinces) was associated with the consumption of raspberries harvested in Guatemala. [206] *Cyclospora* may be a relatively common cause of diarrhea (11%) in patients with AIDS in developing countries but is relatively rare in patients with AIDS in the United States, possibly because of widespread use of trimethoprim-sulfamethoxazole (TMP-SMX) as prophylaxis against *Pneumocystis carinii.* [207]

Pathogenesis and Pathology Limited information is available on how *Cyclospora* causes disease in humans. Disease appears to involve primarily the small intestine, and documented impaired D-xylose absorption suggests that the proximal small intestine may be an important target for infection. [188], [208] Electron microscopic studies of jejunal biopsy samples have demonstrated that the sporozoite, trophozoite, schizont, and merozoite stages of the organism are present within parasitophorous vacuoles in the cytoplasm of infected enterocytes. [191], [194] Histological findings on examination of biopsy specimens from patients with active *Cyclospora* infection are similar to those seen with cryptosporidiosis and with tropical sprue. [194]

Clinical Manifestations Prolonged, nonbloody diarrhea (99%), associated with loss of appetite, fatigue, and weight loss (90%), are the major manifestations of *Cyclospora* infection and are similar to those observed for cryptosporidia. [206] The incubation period is estimated to be 7 to 10 days, but shorter (1-day) incubation periods have been reported. [185], [197] The diarrhea can be prolonged, with a mean duration of illness of 14 days (1 to 60 days) reported in the 1996 outbreak, but other series have reported a mean duration of as long as 56 days. [198], [200], [206], [207], [208] and [209] The mean duration of oocyst shedding was 23 days. [190] Symptoms of diarrhea and fatigue may persist after organisms are no longer detectable in stool samples; conversely, individuals may continue to shed oocysts after symptoms have disappeared. Although cyclosporiasis can be prolonged and frequent relapses are common, disease appears to be self-limited in immunocompetent hosts. In contrast, in immunosuppressed hosts, cessation of infection may not occur without drug therapy. [207] Extraintestinal manifestations of *Cyclospora* infection appear to be rare, but acalculous cholecystitis has been reported in patients with AIDS. [209]

Diagnosis *Cyclospora* infection should be suspected in patients with prolonged diarrhea, particularly those with a recent history of travel. Diagnosis depends on the detection of oocysts by the microscopic examination of stool samples (Fig. 125-5; see Table 125-2). Because excretion of oocysts may be intermittent, multiple stool samples should probably be examined before excluding the diagnosis of *Cyclospora* infection. The diameter of oocysts in stained specimens can distinguish *Cryptosporidium* oocysts (4 to 6 µm) from the larger (8 to 10 µm) *Cyclospora* oocysts.

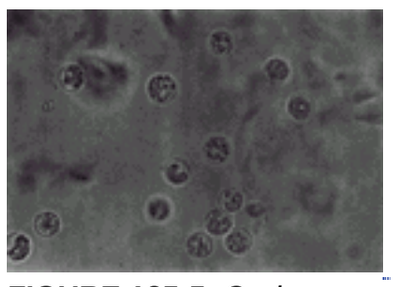

FIGURE 125-5. *Cyclospora cayetanensis* oocysts (saline, wet preparation). Note 8- to 10-µm diameter spheres containing numerous refractile globules. (Courtesy of Earl G. Long.)

Diagnosis of cyclosporiasis also has been made by light microscopic examination of duodenal and jejunal aspirates obtained during endoscopy and examined as wet mounts, or, probably more efficiently, as acid-fast–stained samples. Smears also can be examined under ultraviolet fluorescence microscopy looking for the characteristic blue fluorescence. Identification of the parasite by light microscopy in a duodenal biopsy specimen has been reported, but in general, this has not been a high-yield approach for diagnosis, and electron microscopy probably offers a more sensitive method in biopsy specimens. [191]

Treatment Immunocompetent patients can be treated successfully with few or no relapses with TMP-SMX (160 mg TMP/800 mg SMX) twice daily for 7 to 10 days. [207], [210] Individuals seropositive for HIV with cyclosporiasis were treated with TMP-SMX (160/800 mg) taken orally four times daily for 10 days and had cessation of diarrhea within 1 to 5 days (mean, 2.5 days) of initiation of therapy and negative stools at day 10. [207] About 40% of the patients developed recurrence of cyclosporiasis and required retreatment with TMP-SMX. Continued prophylactic therapy with TMP-SMX orally three times weekly prevented further relapse in all but one of these patients. Few data are available on potential alternatives to TMP-SMX in sulfa-allergic patients, but pyrimethamine and the newer macrolides may be effective.

Isospora belli

Isospora belli is a coccidian protozoan that was first recognized as an intestinal parasite of humans more than 80 years ago in military forces stationed overseas and in outbreaks of diarrhea in institutionalized individuals. [211], [212] *Isospora* infection is more common in individuals residing in developing countries and is relatively rare in immunocompetent individuals in the United States. Sporadic small-scale outbreaks of *Isospora* infection have been seen in mental institutions and day-care centers within the United States, but more widespread outbreaks, as have been seen with *Cryptosporidium* and *Cyclospora,* have not been reported. [211], [213], [214] Isospora is an important cause of diarrhea (about 15%) in individuals with AIDS in developing countries [215], [216], [217], [218], [219] and [220] but accounts for only 0.2% of diarrhea cases in AIDS patients within the United States. [221] Isospora is sensitive to TMP-SMX, and one reason for the lower prevalence of isosporiasis in AIDS patients within the United States may be the widespread use of TMP-SMX as prophylaxis for *P carinii* infection.

The manifestations of *Isospora* infection are similar to those seen with both *Cyclospora* infection and cryptosporidiosis: watery, nonbloody, noninflammatory diarrhea that is often accompanied by abdominal cramps, fatigue, weight loss, anorexia, and occasionally fever. [212], [214], [222], [223] and [224] The incubation period is relatively short (2 to 3 days), and infection is generally self-limited but may be quite prolonged in immunocompetent hosts. [223], [224] In contrast to cryptosporidiosis and cyclosporiasis, a peripheral eosinophilia may be seen in individuals with isosporiasis. [223] In immunosuppressed hosts, most prominently those with AIDS, disease is more severe, and recurrent episodes of profuse watery diarrhea are common. [216], [225], [226] Signs and symptoms of malabsorption may be prominent. [224] Extraintestinal disease is rare, but acalculous cholecystitis, reactive arthritis, and disseminated infection have been described in individuals with AIDS. [227], [228] and [229]

Diagnosis is made by detection of the *Isospora* oocysts in feces using light microscopy (see Table 125-2). Shedding of oocysts is intermittent, and multiple stool samples may need to be examined. Charcot-Leyden crystals and eosinophils are occasionally seen in stool samples from individuals with isosporiasis. [223], [224] There are case reports of individuals who had negative stool tests but had organisms detected by histological examination of small intestinal biopsy specimens. [224]

The drug of choice for the treatment of *Isospora* infection is TMP-SMX (160/800 mg four times daily for 7 days, followed by 10 days of 160/800 mg twice daily).

Patients with AIDS may require more prolonged initial therapy; once infection has been eradicated, they should be placed on prophylactic TMP-SMX (160/800 mg three times weekly) or sulfadoxine-pyrimethamine (500 mg sulfadoxine/25 mg pyrimethamine given once weekly) to prevent relapses. [215], [225] Alternative agents in the sulfa-allergic patient include pyrimethamine alone, nitrofurantoin, and furazolidone.

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CHAPTER 126

Richard D. Pearson

PARASITIC DISEASES: HELMINTHS

Intestinal helminths are prevalent throughout the world in areas where sanitation and public health measures are poor. Although many infested persons are asymptomatic, the impact of helminthic infections on health and childhood development in endemic areas is substantial. In industrialized countries, *Enterobius vermicularis,* the pinworm, is common among children, and other intestinal helminths are encountered among immigrants, returning travelers, and occasionally local residents.

The pathogenic intestinal helminths (Table 126-1) are divided into the roundworms (Nemathelminthes of the class Nematoda) and the flatworms (Platyhelminthes), which are subdivided into the Cestoda, or tapeworms, and the Trematoda, or flukes. [1], [2] This classification system is useful clinically because members of these groups tend to have similar life cycles, metabolic pathways, and susceptibilities to chemotherapeutic agents. Photographs of adult helminths and eggs are available in the *Atlas of Gastroenterology* (see Chapter 80).

TABLE 126-1 Helminthic Infections Involving the Human Gastrointestinal Tract and Their Treatment

The degree to which intestinal helminths compromise health depends on multiple factors, including their pathogenicity, environmental factors, and the genetically determined susceptibility and immune responses of their hosts. In general, tissue damage correlates with the magnitude of the parasite burden, which varies widely among persons, even in areas of intense transmission. In a study of *Ascaris lumbricoides* in Nepal, genetic components accounted for between 30% and 50% of the variability in worm burden and environmental factors between 3% and 13%. [3] In some instances a single worm or a limited number of worms or larvae produce life-threatening disease.

Helminths typically have complex life cycles. Most cannot multiply in their hosts and have finite life spans. The common intestinal nematodes usually die after several years, although some of the flukes, such as *Clonorchis sinensis,* can survive for decades. In the case of *Strongyloides stercoralis, Capillaria philippinensis,* and *Hymenolepis nana,* autoinfection can result in large parasite burdens and persistent infection.

The mechanisms of immunity to intestinal helminths in humans are poorly understood. Data from animal models suggest that $CD4^+$ lymphocytes and their cytokines are important. In general $CD4^+$ Th2-type cytokine responses (Il-4, Il-5, Il-9, and Il-13) are associated with expulsion of worms from the gastrointestinal tract, while Th1-type responses (Il-12 and interferon-?) are found in chronic infections. [4] The precise interplay among these cytokines, the effector mechanisms, and the role of eosinophils and immunoglobulin E (IgE)-mediated responses remain to be defined.

Exposure in an endemic area along with clinical evidence of disease should alert the clinician to the possibility of a helminthic infection. Eosinophilia may be present, but it is neither a sensitive nor a specific test for helminths. A species-specific diagnosis usually depends on identification of adult worms, larvae, or ova in stool or tissue. Serologic tests provide presumptive evidence of infection in many situations. A number of effective drugs are available for the treatment of helminthic infections (see Table 126-1). [5], [6]

INTESTINAL NEMATODES (ROUNDWORMS)

Intestinal nematodes, or roundworms, are prevalent in areas of the world where sanitation is poor and indiscriminate defecation occurs. It is not uncommon for residents of endemic areas to be infected with more than one helminthic species. The intensities of these infections vary depending on genetically determined, but poorly understood, host factors and micro-environmental variables. [3], [4] Children tend to have the highest worm burdens.

The life cycles of intestinal nematodes can be divided into three general patterns. In many cases infection follows ingestion of ova (Fig. 126-1), which excyst

releasing larvae in the gastrointestinal tract. *Trichuris trichiura* and *E vermicularis* develop to adulthood within the lumen of the bowel, whereas *Ascaris lumbricoides* larvae invade the intestinal wall, pass through the venous circulation to the lungs, migrate into the alveoli, ascend to the pharynx, are swallowed, and then reach maturity in the intestinal tract. Hookworms and *S stercoralis* invade the skin, pass through the venous circulation to the lungs, and then follow a pattern comparable to that of *Ascaris*.

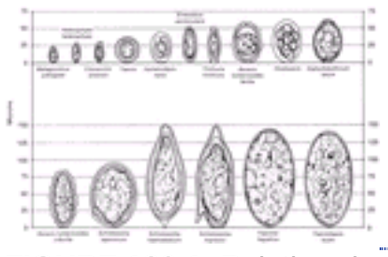

FIGURE 126-1. Relative sizes and shapes of helminth ova. (Adapted from Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.)

The clinical manifestations of these helminthic infections generally depend on the organ systems involved and the magnitude of the worm burden. Children living in endemic areas have demonstrated improved growth and activity following treatment of roundworms. [7], [8] and [9] The disability-adjusted life years lost to helminthic diseases has been estimated to be 39 million. [10] This has led to the implementation of mass treatment programs with the broad-spectrum anthelmintic albendazole in areas of high prevalence.

Trichuris trichiura (Whipworm)

The whipworm, *T trichiura,* is prevalent in developing areas in tropical and temperate climates. [11] Humans become infected when they ingest embryonated ova. After excystation, larvae penetrate the intestinal mucosa with their threadlike anterior ends, molt, mature, and reattach as adults to the mucosa of the cecum and the rest of the colon. Mature female worms release 2000 to 6000 eggs (see Fig. 126-1) per day into the feces. Maturation requires a period of 10 to 14 days in the soil. About 3 months elapse between the ingestion of eggs and the production of ova by mature whipworms. Adults can persist in the colon for several years.

Epidemiology *T trichiura* has a worldwide distribution. Various *Trichuris* species infect animals, but only *T trichiura* is found in humans. Ova require warm, moist, shaded soil for development. Infection is most frequent in areas without latrines and in communities where untreated human fecal material is used for fertilizer. The severity of infestation also varies within a community resulting in an aggregated, or clumped, distribution. The subset of people with heavy infections is more likely to show manifestations of disease.

Clinical Manifestations Mild *T trichiura* infections are often asymptomatic. [11] Heavy infections are most common in children between 2 and 10 years of age and may be associated with diarrhea, anemia, and growth retardation. [12] A small percentage of those infected present with a dysentery syndrome. [13] Rectal prolapse is a rare but potentially serious consequence of heavy infection [11] (see Fig. 126-2, Color Fig. 126-2). Cases of colonic obstruction and perforation have been reported. [14] Eosinophilia is frequently present, even in light infections.

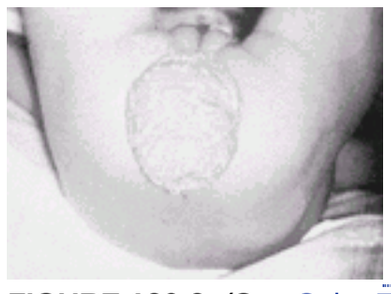

FIGURE 126-2. (See Color Fig. 126-2.) *Trichuris trichiura* associated with rectal prolapse in a child. Adult *T trichiura* are seen as white threads on the mucosal surface. (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.)

Diagnosis, Treatment, and Prevention The diagnosis of trichuriasis is confirmed by identifying ova in the stool or adult worms in the colonic mucosa. Mebendazole or albendazole are recommended for therapy (see Table 126-1). A single 400-mg dose of albendazole is used in mass treatment programs in endemic communities, [7], [8] but in persons with heavy infections, it is advisable to administer it daily for 3 days. [5], [6] A single dose of albendazole combined with ivermectin has been reported to be more effective than albendazole alone. *T trichiura* infection can be prevented by adequate sanitary disposal of human feces, hand washing, and proper preparation of food.

Enterobius vermicularis (Pinworm)

E vermicularis is endemic in both temperate and tropical climates. [11], [15] It is encountered most frequently among school-age children living in areas of high population density. People become infected when ingested ova hatch in their upper small bowel, and larvae mature as they migrate to the ileum. Adults live 7 weeks (males) to 13 weeks (females). The earliest that eggs appear in the stool is 5 weeks. An infected person may harbor anywhere from a few to several hundred adult pinworms.

At the time of oviposition, the adult female (Fig. 126-3) migrates out through the anus to lay her eggs on the perianal or perineal skin. Eggs (see Fig. 126-1) are expelled by uterine contraction, by death and disintegration of the adult worm, or by disruption of worms during scratching. The shell of the ovum has a thick outer albuminous layer, which plays a role in adherence to objects in the environment.

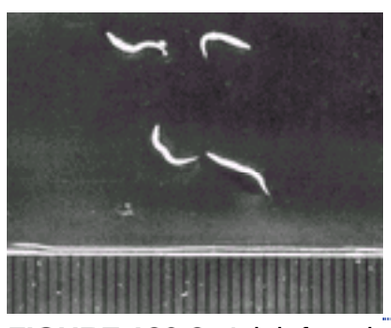

FIGURE 126-3. Adult female *Enterobius vermicularis* (pinworms) may be found on the perianal skin or occasionally on the surface of stools. Adult female pinworms are 8 to 13 mm long and 0.3 to 0.5 mm wide. (The scale is in millimeters.) (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.)

Epidemiology *E vermicularis* can infect people of all ages, but it is most prevalent among children between the ages of 5 to 10 years. [11], [15], [16] Finger sucking, [17] poor personal hygiene, and exposure to infected family members or peers in schools or orphanages are contributing factors. Although one time as many as 30% of elementary school students in the United States were infected, the prevalence has probably decreased in recent decades. [18] Enterobiasis also has been reported

among male homosexuals, who become infected through anilinction.

Clinical Manifestations The most important consequence of *E vermicularis* infection is irritation of the perianal or perineal skin, [11], [15], [16] although many infected persons are asymptomatic. Scratching, self-induced trauma to the skin, and, in some persons, secondary bacterial dermatitis may result. Vulvovaginitis may develop in prepubertal girls when adult pinworms migrate to the vagina, [19] and may result in secondary enuresis [20] or introital colonization leading to bacterial urinary tract nfection. [21] Adult pinworms occasionally are found in the appendix after surgical excision. Although they have been associated with abdominal pain, they do not appear to cause acute appendicitis. Eosinophilic ileocolitis has been associated with *E vermicularis* in persons presenting with abdominal pain, melena, and diarrhea. [22], [23] On rare occasions, adult female pinworms reach the peritoneum by traversing the female genital tract or by migrating through a perforation in the bowel associated with appendicitis, diverticulitis, or intestinal malignancy to produce ectopic peritoneal disease. [24], [25] Granulomatous reactions to dead worms or eggs may result in small peritoneal nodules, which can be confused grossly with metastatic carcinoma. Granulomatous inflammation in response to worms or eggs also has been observed in the vulva, vaginal wall, cervix, endometrium, salpinx, and ovaries, as well as in the liver and epididymis. [26]

Diagnosis, Treatment, and Prevention The discovery of ova (Fig. 126-2 and Color Fig. 126-2) or pinlike adult female worms on the perianal skin (Fig. 126-3) is indicative of enterobiasis. The most successful diagnostic approach uses a strip of transparent tape that is held with the adhesive side out affixed to a tongue depressor. [16] When a person first arises in the morning, before bathing or defecation, the buttocks are spread and the tape is pressed against the perianal skin several times. The strip then is transferred to a microscope slide with the adhesive side down. Eggs are prominent at low power. A single smear detects about 50% of those infected; 90% can be detected with three swabs. Stool examination for ova and parasites is positive in only 10% to 15% of cases. On rare occasions, ova are found in vaginal smears treated with Pap stain or in the urine sediment. *E vermicularis* is susceptible to a number of anthelmintic drugs (see Table 126-1). Pyrantel pamoate, administered as a dose of 11 mg/kg body weight (maximum, 1 g) and repeated in 2 weeks, is available over the counter in the United States. Albendazole and mebendazole are also effective. Spread of *E vermicularis* can be reduced by hand washing and by treating infected children. Because the home environment is often contaminated with viable ova, it is advisable to launder clothes and linen; to vacuum around beds, curtains, and other potentially contaminated areas; and to cover food to avoid dust-borne eggs. Anthelmintic treatment of the entire household may be necessary to interrupt transmission.

Capillaria philippinensis

C philippinensis, a small intestinal nematode, causes severe diarrhea and malabsorption. [11], [27], [28] It was initially described among residents of the Philippines, but cases also have been reported from Thailand, Indonesia, Japan, Taiwan, Korea, India, Egypt, Iran, and Colombia. Infection follows ingestion of raw or inadequately cooked freshwater or brackish fish, the intermediate host, containing infectious larvae. Fish-eating birds are the usual definitive hosts.

Adult *C philippinensis* invade the mucosa of the small intestine of humans causing inflammatory changes in the lamina propria. [27], [28] Symptoms include watery diarrhea (often severe), abdominal pain, borborygmus, constitutional complaints, malabsorption, and weight loss. Infection also has been associated with jejunal stricture and chronic intestinal pseudoobstruction. Some ova are passed in the stool; others excyst in the intestine and complete the life cycle resulting in autoinfection and a large worm burden. Deaths occur as a result of malnutrition, electrolyte imbalance, and secondary infections in debilitated patients.

The diagnosis of *C philippinensis* is made by identifying ova, larvae, or adult worms. Multiple stool examinations often are necessary because ova are shed sporadically and in low numbers. Albendazole (see Table 126-1) is the treatment of choice; mebendazole is an alternative. Supportive care with fluid, electrolytes, and nutritional replacement is often necessary. Thorough cooking of fish prevents infection.

Capillaria hepatica, a common hepatic pathogen of rodents and other animals, has been associated with hepatomegaly, fever, and eosinophilia in a very limited number of persons, primarily children, around the world. [29], [30] The pathology is characterized by prominent granulomatous reactions to eggs in the liver. The diagnosis usually is made by liver biopsy. Albendazole has been used for treatment.

Trichostrongylus Species

Trichostrongylus species are common parasites of herbivorous animals such as sheep, horses, and goats. [1] Human infections occur most prevalently in the Middle East and Asia. Ova are passed in the feces of animals, hatch in 1 or 2 days under favorable conditions, and pass through 3 free-living stages before becoming infective after approximately 3 days. Although larvae can invade through the skin, human infection usually occurs when they are ingested in contaminated food or water. Larval migration through the lungs is not necessary for maturation. Adult worms reside in the duodenum or upper jejunum with their heads embedded in the mucosa.

Most human infections are asymptomatic, but epigastric pain, diarrhea, and flatulence have been reported. Rarely, anemia or emaciation develops. Eosinophilia is present in some cases. The diagnosis is made by identifying *Trichostrongylus* ova in stool. *Trichostrongylus* ova are larger and more pointed on the ends than hookworm ova. Albendazole, mebendazole, or pyrantel pamoate can be used for treatment (see Table 126-1). Vegetables should be cooked thoroughly and water boiled before ingestion in endemic areas.

Ascaris lumbricoides

A lumbricoides is the most prevalent and the largest of the intestinal nematodes that infect humans; [31] it is the only nematode that truly looks like a “worm.” Adult female ascarids can grow to a length of 40 cm. They produce a prodigious number of ova—up to 200,000 each day (see Fig. 126-1). Fertilized eggs develop in moist, warm, shaded soil and become infectious in 2 to 4 weeks. After ingestion in contaminated food or water, eggs hatch in the duodenum. Larvae subsequently penetrate the intestinal wall, enter the venous circulation, and migrate to the lungs. They break into the alveoli, ascend to the trachea, are swallowed, and complete their development in the small intestine. The time from ingestion of ova to the production of eggs by mature females is 10 to 12 weeks.

Epidemiology An estimated 1.3 billion people are infected with *Ascaris* worldwide, resulting in 12 million cases of acute illness and 10,000 deaths each year. [32] Only cold, arid climates are spared. Transmission of *Ascaris* is continuous in the tropics and intermittent in temperate areas. While the majority of the population may be infected in areas of poor hygiene, only a subset, usually children younger than 10 years of age, harbor a large number of adult worms at any given time. [33] Genetic and shared environmental factors account for the variation. [3]

Clinical Manifestations The clinical manifestations of ascariasis can be divided into those associated with the migration of larvae through the lungs and those associated with adult worms in the gastrointestinal tract. [31] Migrating larvae in the lungs can elicit pulmonary hypersensitivity reactions, including bronchospasm, hypersecretion of mucus, and bronchiolar inflammation. Sputum may contain Charcot-Leyden crystals and, on occasion, larvae. The presence of eosinophilia with pulmonary infiltrates leads to the clinical diagnosis of Löffler syndrome. Urticaria and other manifestations of hypersensitivity may also be present during the pulmonary phase. *Ascaris* antigens are among the most potent allergens. Hypersensitivity reactions have been a major problem for investigators working with *Ascaris* in vitro. Accurate characterization of the gastrointestinal manifestations of *Ascaris* infection is confounded by the high frequency of concurrent infestations with other intestinal parasites and enteropathogenic bacteria. Patients harboring *Ascaris* have reported abdominal pain, nausea, anorexia, and diarrhea, but it has been difficult to determine how frequently *Ascaris* is responsible for these symptoms. The precise impact of Ascaris infection on nutrition is difficult to quantify in people harboring multiple enteric pathogens, [34], [35] but children infected with *Ascaris* and other common intestinal nematodes have demonstrated gains in growth and activity after treatment with albendazole. [7], [8] and [9] One of the most serious complications with *Ascaris* occurs when a large number of worms become intertwined to form a bolus that causes partial or complete intestinal obstruction in young children (Fig. 126-4; see also Color Fig. 126-4). [36] It has been estimated to occur in as many as 2 per 1000 children infected with *Ascaris* per year. [37] Those affected present with nausea, vomiting, abdominal pain, and occasionally a palpable abdominal mass. [36], [37] and [38] The presentation may mimic an acute abdomen. Rarely, a mass of worms results in intussusception or volvulus of the small bowel. Bowel perforations with peritonitis and localized abscesses have been reported, but the role of *Ascaris* in these cases remains uncertain. Intestinal pseudoobstruction has been reported with *Ascaris*-associated inflammatory lesions. [39] Adult *Ascaris* have also been found in the appendices of persons presenting with of acute appendicitis. [40]

FIGURE 126-4. (See Color Fig. 126-4.) Intestinal obstruction caused by a mass of adult *Ascaris lumbricoides* is seen in this autopsy specimen. Intestinal obstruction is an unusual complication of heavy *Ascaris* infection. (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.)

On occasion, adult *Ascaris* migrate to the oropharynx or nasopharynx, much to the consternation of the host or parents, or more importantly, they may enter the common bile duct, producing biliary colic, acute cholecystitis, ascending bacterial cholangitis, secondary liver abscess, perforation of the bile duct, intrahepatic or bile duct calculi, [41], [42] or acute pancreatitis. [43], [44]

Diagnosis, Treatment, and Prevention Embryonated or unembryonated ova in feces are indicative of *A lumbricoides* infection (see Fig. 126-1). On occasion, migrating adult worms, which range from 10 to 40 cm in length, are found in feces; are detected radiographically or sonographically in the gastrointestinal tract, bile duct, pancreatic duct, or intestine; [41], [42], [43] and [44] or emerge from the oropharynx or nose. By ultrasound they produce echogenic, nonshadowing images that appear as single or multiple strips. The digestive tract of the worm appears as an anechoic inner tube. *Ascaris* can also be visualized by computed tomography. On contrast-enhanced radiographic studies, they appear as cylindrical filling defects; the intestinal tract of the worm may be seen as a thin thread of contrast material within a tubular filling defect. Albendazole, 400 mg as a single dose, mebendazole, or pyrantel pamoate are effective in the treatment of *Ascaris* (see Table 126-1). Therapy for the complications of *Ascaris* depends on the organs involved. Intestinal obstruction may require surgery, but most patients are treated successfully with rehydration and anthelmintic drugs. Endoscopic extraction and sphincteroplasty have been used to remove adult worms from the biliary tract, but chemotherapy and supportive therapy are effective in many cases. In principle, ascariasis can be prevented by the sanitary disposal of human feces and good personal hygiene. In practice, this is difficult to achieve. *Ascaris* eggs are relatively stable in the environment and can persist for years. Community-based mass treatment programs are effective, but treatment must be repeated, usually at 3- to 4-month intervals, due to re-infection. [45]

Trichinella spiralis

Trichinella spiralis is cosmopolitan in its distribution. [46] Humans become infected when they ingest inadequately cooked meat containing infective larvae. The encysted larvae are released in the small intestine and molt 4 times within 36 hours to become adults. After mating, females remain embedded in the mucosa. In about 5 days, viviparous females begin to deposit larvae, which invade the mucosa, enter the lymphatics or bloodstream, and ultimately encyst in skeletal muscle cells. They remain there until ingested by another carnivore or until their death from senescence after several years. A single female worm produces approximately 1500 larvae during her life.

Epidemiology *T spiralis* is responsible for the majority of cases of trichinosis, but some are caused by *T britovi, T pseudospiralis, T nativa* in arctic regions, and *T nelsoni* in Africa. Approximately 50 cases of trichinosis occur in the United States annually. [47] Trichinosis can follow ingestion of contaminated pork or sausage from domestic pigs that have had access to garbage or wildlife carcasses, or from inadequately cooked meat from wild boar, bear, walrus, cougar, horse, or other animals. [48], [49], [50] and [51] Horses have become infected from protein supplements made from contaminated meat. [51] As the number of cases of trichinosis due to commercial pork has decreased in the United States and other developed countries, the proportion of cases due to wild game has risen. Cases of trichinosis are often sporadic, but small outbreaks are not uncommon. Infections in North America have been reported among immigrants from Southeast Asia who prefer rare or undercooked pork; trichinosis has also been reported among international travelers who have eaten inadequately cooked pork or other infested meat. [52]

Clinical Manifestations During the initial phase of trichinosis, nausea, vomiting, abdominal pain, or diarrhea may occur as the result of larval invasion of the intestinal mucosa. The systemic phase, which typically begins 1 to 3 weeks after ingestion of contaminated meat, is characterized by fever, myalgia, facial or periorbital edema, headache, conjunctivitis, and occasionally a rash. [46], [47], [48], [49], [50], [51] and [52] Extraocular muscles are frequently affected. Cardiac and central nervous system involvement occur in a minority of cases, and can be life threatening. Trichinosis among native Inuits living in northern Canada differs in that prolonged diarrhea is the dominant symptom in previously infected people who become re-infected. [50]

Diagnosis, Treatment, and Prevention The diagnosis of trichinosis is suggested by the constellation of fever, myalgia, eosinophilia, and a history of eating raw or inadequately cooked, potentially contaminated meat. Creatine phosphokinase or aldolase is elevated in about half of the cases. The clinical diagnosis is eventually supported by serologic evidence. This includes a positive enzyme-linked immunosorbent assay (ELISA), bentonite flocculation test, or indirect immunofluorescence test. However, anti- *Trichinella* antibodies are usually not detectable until the third week of infection or later. [46] The diagnosis can be confirmed by muscle biopsy, but that is seldom necessary. Either albendazole [53] or mebendazole [5] can be used for the treatment of acute trichinosis, along with corticosteroids when symptoms are severe (see Table 126-1). Bed rest is prescribed and nonsteroidal antiinflammatory agents are administered for symptomatic relief of fever and myalgias. Trichinosis can be prevented by cooking pork or other potentially contaminated meat to at least 76.6°C (170°F). Meat less than 6 inches thick can be rendered safe if frozen at -15°C (5°F) for 20 days, unless it is infected with *Trichinella nativa,* which is resistant to freezing. [50]

Hookworms

The hookworms, *Ancylostoma duodenale* and *Necator americanus,* rank among the major causes of iron deficiency anemia in the world. Adult hookworms are small, creamy white nematodes. [11], [54] The anterior portion of *N americanus* is sharply curved in the direction opposite the curve of the body, producing a hooklike appearance. Fourth-stage larvae and adults anchor themselves in the mucosa of the small intestine and employ complementary strategies to inhibit the coagulation of the host's blood. They secrete *Ancylostoma* anticoagulant peptide, which inhibits factor Xa, a factor(s) that inhibits platelet aggregation and adhesion, and fibrinogenolytic enzymes. [55], [56] and [57] Hookworms periodically change their location. Bleeding from the former site of attachment contributes to the total blood loss. *N americanus* infection results in the loss of approximately 0.03 mL/d of blood per adult hookworm, and *A duodenale* infections results in the loss of 0.15 to 0.26 mL/d. [58], [59]

Ova (see Figure 126-1) are released into the intestine and excreted in feces. They excyst in the soil liberating rhabditiform larvae, which feed actively on organic debris and bacteria. After increasing in size, they molt twice to become infectious filariform larvae, which migrate in response to pressure, carbon dioxide, or warmth and invade the epidermis through fissures or hair follicles, most commonly between the toes or on the dorsum of the feet. Infection also may be acquired through the interdigital spaces of the hands. Invading larvae exsheath as they enter the host and pass through the venous circulation to the lungs. There they break out of the alveoli, ascend to the pharynx, and are swallowed. Although invasion is principally through the skin, filariform larvae of *A duodenale* occasionally are ingested and complete their life cycle in the gut. In addition, data from animal models suggest that larvae can enter a developmentally arrested state in somatic tissues, including mammary glands, and could infect breast-feeding infants after resuming development.

Epidemiology *A duodenale* and *N americanus* are widely distributed in tropical and subtropical areas. Hookworm infections are limited to areas where the climate allows development of larvae in the soil. They are readily killed by desiccation or freezing. *A duodenale* is found in the Middle East, North Africa, India, and China. *N americanus* frequently overlaps *A duodenale* in Africa, India, and China and is the dominant hookworm in sub-Saharan Africa, southern China, southern India, and Southeast Asia. *N americanus* is the predominant species in the New World, where there are also focal sites of *A duodenale*. *N americanus* infections were once common in the southeastern region of the United States, but treatment and control measures resulted in a dramatic lowering of prevalence by 1965. [60] Human infection with *A ceylonicum* has been reported from Southeast Asia and India. In general the prevalence and intensity of hookworm infection increases through childhood, reaching a plateau in young adults. Several factors favor the spread of hookworms: poor sanitation and agrarian practices; walking barefoot; and the use of human excrement as fertilizer.

Clinical Manifestations The clinical manifestations of hookworm infection correlate with the life cycle of the organism and the intensity of infection. [11], [54] Penetration of the skin by filariform larvae can produce a papular eruption with intense pruritus, vesiculation, and local edema (“ground itch” or “dew itch”). As larvae pass through the lungs, they can elicit hypersensitivity reactions with cough, wheezing, infiltrates, and eosinophilia (Löffler syndrome). Epigastric pain, flatulence, and abdominal tenderness accompany the early intestinal phase of infection in some. Humans experimentally infected with *N americanus* [61] experienced abdominal pain and flatulence 35 to 40 days after exposure to filariform larvae, eosinophilia peaked between 38 and 64 days, and eggs first appeared in the stool during the sixth week. At times, abdominal pain was severe enough to suggest peptic ulcer disease and occasionally was accompanied by diarrhea with mucus or blood in the stool. In young children with heavy primary hookworm infections, massive invasion by filariform larvae can result in acute gastrointestinal hemorrhage, a condition that is rare but potentially life threatening. [62] The hallmark of chronic hookworm disease is iron deficiency anemia. [11], [54], [63] The infecting species of hookworm; the parasite burden;

the age, sex, diet, iron requirements, and iron reserves of the host; and concurrent malaria are important variables. Hookworm disease with severe anemia is common in young children, potentially stunting growth and intellectual development, and in women during pregnancy, increasing the risk of maternal and neonatal mortality. [64] Hypoalbuminemia is often present and generally correlates with the degree of anemia. In areas of high iron intake and bioavailability, even heavy hookworm infections may not produce anemia.

Diagnosis, Treatment, and Prevention Hookworm disease should be considered in any person from an endemic area who presents with the clinical and laboratory findings of iron deficiency anemia. Eosinophilia frequently accompanies hookworm infection. [65] Direct fecal examination for ova is adequate to detect clinically significant infections, except in the case of the naive host who presents with eosinophilia or abdominal symptoms in the prepatent stage of infection prior to the onset of ova production. It is not easy or necessary clinically to differentiate between the eggs of *N americanus* and *A duodenale*. Hookworm ova can hatch in stool specimens that have been allowed to stand at warm temperature, and it is occasionally necessary to differentiate the rhabditiform larvae of hookworms from those of *S stercoralis*. Hookworm larvae have a long buccal tube, in contrast to the short buccal tube of *S stercoralis*. The treatment of hookworm disease includes iron repletion as well as anthelmintic therapy (see Table 126-1) with albendazole, mebendazole, or pyrantel pamoate. [66] During pregnancy, iron deficiency anemia is treated with ferrous sulfate alone, and anthelmintic therapy, which is potentially teratogenic, is administered after delivery. Empirical therapy is occasionally administered to returning travelers who present with eosinophilia and abdominal symptoms after a recent exposure who may be in the prepatent period. The transmission of hookworm infection can be interrupted by sanitary disposal of human feces, use of footwear, and treatment of infected people.

Strongyloides stercoralis

S stercoralis is in an important cause of morbidity and mortality, but it is less prevalent than *A lumbricoides* or the hookworms. It differs from them in its ability to complete its life cycle in the soil and in its capacity to produce autoinfection, which can result in infections that last for decades and may progress to life-threatening hyperinfection in immunocompromised hosts. [67] Like the hookworms, *S stercoralis* enters the skin from soil contaminated by feces, migrates through the venous circulation to the lungs, penetrates alveoli, ascends the trachea, is swallowed, and ultimately reaches maturity in the small bowel. Adults reside in the superficial mucosa of the duodenum or jejunum and produce ova, which quickly hatch releasing rhabditiform larvae. Autoinfection occurs when a subset of rhabditiform larvae converts to filariform larvae in the gut or on the skin. Filariform larvae can penetrate the intestinal mucosa or skin and go on to complete the life cycle. *Strongyloides* infections can persist for decades because of autoinfection. The conversion of rhabditiform to filariform larvae is enhanced in immunocompromised person and those receiving corticosteroids. [67] Rhabditiform larvae are excreted in fluctuating numbers in the stool. After reaching the soil, they convert to infectious filariform larvae or to free-living adults, which in turn produce rhabditiform larvae that convert to filariform larvae. *S stercoralis* is the only human geohelminthic pathogen that can complete its life cycle entirely in the soil.

Epidemiology *S stercoralis* is endemic in tropical areas of Africa, Asia, and Latin America as well as the southern part of the United States and Eastern Europe. Prevalence rates of 3% to 21% have been reported from Nigeria. Autochthonous cases are reported from Appalachian regions of West Virginia, Virginia, and Tennessee. [68], [69] and [70] A number of U.S. military personnel who were exposed in Southeast Asia during World War II or the Vietnam War remained infected decades later. [71], [72] and [73] *Strongyloides fülleborni,* primarily a pathogen of infrahuman primates, is recognized as an occasional cause of human strongyloidiasis in Papua, New Guinea and Africa. [74] An unexpectedly high prevalence of *S stercoralis* and increased severity of disease has been observed in persons concurrently infected with human T-cell lymphotrophic virus type 1 (HTLV-1). [75], [76] and [77] The interactions between the two pathogens are complex. It is thought that the spontaneous secretion of high levels of interferon-? by HTLV-1- infected mononuclear cells may suppress potentially protective Th2 responses. [77] In addition, chronic infection with *S stercoralis* may hasten the development of T-cell lymphoma or leukemia in persons infected with HTLV-1. [78] *S stercoralis* can produce life-threatening hyperinfection in persons with suppressed immunity and those receiving steroids. [67] The syndrome had been reported in association with HTLV-1, organ transplantation, malignancies (especially lymphoma and leukemia), corticosteroid and other immunosuppressive therapy, protein-calorie malnutrition, visceral leishmaniasis, lepromatous leprosy, and other debilitating conditions. Hyperinfection has been associated with human immuno- deficiency virus infection, [79] but it has not been as frequent as originally predicted. Exogenous steroids appear to be homologs of molecules that stimulate conversion of filariform to rhabditiform larvae. No cases of hyperinfection have been reported among transplant recipients receiving cyclosporin, which has been demonstrated in animals to have anti- *Strongyloides* activity.

Clinical Manifestations Most people are asymptomatic at the time of cutaneous penetration by larvae, although some develop a maculopapular rash or linear urticaria, called larva currens, [67], [80] which also may be seen as a consequence of external autoinfection. Eosinophilia is common. On rare occasions infected persons may present with generalized, chronic prurigo nodularis or lichen simplex chronicus. The pulmonary phase of strongyloidiasis can be associated with cough, shortness of breath, wheezing, fever, transient pulmonary infiltrates, and eosinophilia. [81] Once *S stercoralis* reaches the gut, it can produce epigastric or diffuse abdominal pain or diarrhea. [67], [82] With heavy infections of the upper small bowel, vomiting, malabsorption, steatorrhea, and weight loss can occur. In some cases the symptom complex may be confused with inflammatory bowel disease. [83] Eosinophilic granulomatous enterocolitis and inflammatory mass lesions have also been reported. [84] Rarely, intestinal obstruction develops. [85] Of note, about one third of chronically infected World War II veterans diagnosed with *S stercoralis* decades after exposure were asymptomatic; the other two thirds reported recurrent larva currens, gastrointestinal complaints, or pulmonary symptoms. *Strongyloides* hyperinfection is characterized by multisystem involvement and the presentation varies. [67] Some patients present with pulmonary symptoms including cough, shortness of breath, wheezing, and hemoptysis. They may progress to the acute respiratory distress syndrome. Infiltrates are seen on chest radiograph. Others have intestinal symptoms. Concurrent polymicrobial bacterial infections often occur as a result of bacteria on the surface of invading filariform larvae. Sepsis, meningitis, peritonitis, and endocarditis have been reported. The mortality rate with *S stercoralis* hyperinfection exceeds 50%.

Diagnosis, Treatment, and Prevention The diagnosis of strongyloidiasis is confirmed by identifying larvae in stool, tissues, or secretions. Eosinophilia is frequently present in immunocompetent patients, but it may be absent in immunocompromised hosts. The agar plate method is the most sensitive way to identify larvae in stool. It has replaced the Baermann funnel and other techniques. [86] In people with the hyperinfection syndrome, larvae are also frequently found in sputum or tissue. Several serologic tests, including ELISA, have been developed and provide presumptive evidence of infection. [87] Ivermectin has emerged as the treatment of choice for uncomplicated strongyloidiasis (see Table 126-1). [5], [88] Thiabendazole, once the drug of choice, was associated with frequent side effects and is no longer manufactured in the United States, but some physicians prefer it for the treatment of patients with hyperinfection. Relapses have been reported with both drugs. Albendazole, 400 mg daily for 3 days, cures about half of those treated. [89] Prevention of strongyloidiasis depends on improved standards of living and personal hygiene. It is advisable to screen immunocompromised persons, particularly those receiving corticosteroid therapy, who may have been exposed to *S stercoralis* in endemic areas.

Other Syndromes Caused by Intestinal Nematodes

Cutaneous Larva Migrans Also known as *creeping eruption,* this is a distinctive form of parasite-induced dermatitis characterized by serpiginous, papulovesicular, erythematous, pruritic lesions. [90] It is a consequence of migrating larvae of intestinal nematodes of animals that cannot complete their life cycle in humans (i.e., hookworms of dogs and cats, primarily *A braziliense* and less commonly *A caninum,* and other nematodes). Cutaneous larva migrans is prevalent in tropical and subtropical areas of the Caribbean, Africa, Latin America, and the southern Atlantic and Gulf of Mexico coasts of North America. Infection is acquired when people come in contact with infected soil in playgrounds, beaches, or crawl spaces under houses. A single 12 mg dose of ivermectin, or alternatively, albendazole, 400 mg daily for 3 days, can be used as treatment.

Eosinophilic Enteritis *A caninum,* the dog hookworm, has been identified as a cause of eosinophilic enteritis, generally a rare condition but quite common in northeastern Australia. [92], [93] The findings include eosinophilic infiltration of the wall of the intestine, aphthous ulceration of the ileum, regional lymphadenopathy, and in some patients peripheral blood eosinophilia. Many of those infected are asymptomatic, whereas others experience abdominal pain. The diagnosis is made by finding a sexually immature adult worm at endoscopy or by serological evidence of *A caninum* infection. Although optimal chemotherapy has not been identified, albendazole or mebendazole are likely to be effective.

Visceral Larva Migrans and Ocular Toxocariasis Visceral larva migrans is a systemic syndrome resulting from invasion of human organs by larvae of the dog ascarid, *Toxocara canis,* the cat ascarid, *Toxocara cati,* [94], [95] or less commonly, other nematodes of animals. Infection follows ingestion of ova originating in animal feces. The syndrome is encountered most commonly in children with a history of pica, but on occasion it is seen in adults, some of whom have a history of eating clay. Larvae are released and penetrate the intestine. Because these helminths are unable to complete their life cycle in humans, they wander through various organs. *Toxocara* species are common throughout tropical and temperate areas. The clinical course of visceral larva migrans is highly variable, ranging from no symptoms to fever, cough, abdominal pain, arthralgia, oligoarticular arthritis, urticarial skin rash, or neurological abnormalities. Physical examination may reveal hepatomegaly, pneumonitis, dermatitis, or encephalitis. Persons with ocular toxocariasis may present with an intraocular mass lesion suggestive of a retinoblastoma. Eosinophilia is common and often pronounced. Hypergammaglobulinemia and high levels of IgE are also observed. These findings have been attributed to the dynamic nature of the larval membrane and the release of antigens that stimulate Th2 responses. The characteristic pathological lesion consists of a granulomatous inflammatory response with eosinophils. The triad of hepatomegaly, eosinophilia, and hypergammaglobulinemia suggests visceral larva migrans. Ultrasound or magnetic resonance imaging can be used to identify hepatic granulomas. An ELISA is available to detect anti- *Toxocara* antibodies. High levels of isohemagglutinins against A and B blood group antigens are also frequently observed. Diethylcarbamazine is used for treatment (see Table 124-1); [5] albendazole is an alternative. Corticosteroids are also administered in severe cases. Control measures include worming dogs, covering sandboxes, and preventing promiscuous defecation of dogs in other areas where

children play.

Anisakiasis Also known as anisakidosis, this syndrome is caused by members of the family Anisakidae, which are nematode parasites of sea mammals. The common intermediate hosts include codfish, herring, rockfish, sardine, anchovy, salmon, tuna, mackerel, Pacific pollock, Pacific red snapper, and squid. [96], [97], [98] and [99] Anisakine larvae normally reside in the viscera of fish, but they migrate to muscle if fish are not promptly filleted after being caught. Anisakiasis occurs after contaminated fish are eaten raw or inadequately cooked. It is most common in Japan and Europe, areas where fish is consumed raw as sushi or sashimi. Sporadic cases occur in the United States. Anisakine larvae are prevalent in susceptible types of fish along the California coast and in the northern Atlantic Ocean. People who ingest infested fish may be asymptomatic, but the presentation can be dramatic when anisakine larvae attempt to invade the mucosa. In acute gastric anisakiasis, severe abdominal pain, chest pain, nausea, vomiting, and occasionally gastric bleeding develop 12 to 24 hours after contaminated fish is ingested. [97] Involvement of the greater curvature of the stomach is most common. In intestinal anisakiasis, symptoms mimicking acute appendicitis, regional enteritis, or small bowel obstruction or perforation develop several days after ingestion. Eosinophilic inflammatory responses develop at the site of attempted larval penetration, but peripheral eosinophilia is not common. Intestinal inflammation and edema may result in luminal narrowing, thumbprinting, or mass effect and may be confused with a neoplasm. Symptoms can persist for weeks to months in chronic cases. The allergic consequences of exposure to *Anisakis* species, which include urticaria, angioedema, erythema, bronchospasm, and anaphylaxis, have been increasingly recognized in recent years. They have been reported in persons ingesting contaminated fish, either raw or cooked, as well as among fisherman and fishmongers. [99], [100] Allergic symptoms can develop up to a day after contaminated fish is ingested. Since the life cycle cannot be completed in humans, neither anisakine larvae nor ova are passed in the stool. Visualizing worms at endoscopy typically leads to a diagnosis of anisakiasis. Larvae have been extracted successfully from the stomach and duodenum by using biopsy forceps. [96], [97] and [98] Surgical resection is occasionally necessary to relieve intestinal obstruction, to repair a perforation of the bowel, or to rule out malignancy, but most patients can be managed conservatively. Anisakiasis can be prevented by filleting freshly caught fish before larvae have a chance to penetrate muscle, by cooking fish to 60°C (140°F) for at least 10 minutes, or by freezing for 24 hours at -20°C (-4°F). Allergic symptoms can occur in patients ingesting properly cooked as well as frozen fish and correlate with the presence of IgE antibodies to *Anisakis simplex.*

Gastrointestinal Angiostrongyliasis *Angiostrongylus costaricensis* is endemic among rodents from California to South America. Adult worms reside in mesenteric arteries in the ileocecal region of rodents. They occasionally infect the same site in humans. Human disease is typically sporadic and observed among children in Central America. Rare cases have been reported in the United States, and an outbreak occurred among adults who ate contaminated mint. [101], [102] Humans and rodents acquire infection by ingesting infected slugs or their slime in vegetation. Adult worms and ova elicit a granulomatous arteritis with eosinophilic infiltration in the ileocecal region that can result in thrombosis, local infarction, and bowel perforation. The clinical manifestations vary from a visceral larva migrans-like syndrome to those of an acute abdomen. Nausea, vomiting, right lower quadrant pain, and fever are common. A right lower quadrant mass may be palpable, suggesting acute appendicitis or malignancy. Radiographic studies may document a mass lesion in the ileocecal region. Eosinophilia and leukocytosis are common. Neither ova nor larvae are found in the stools of humans. Surgery is often necessary, and the diagnosis is made when adult worms or ova are identified in surgical specimens. Cooking slug-infested vegetables and hand washing can prevent infection.

CESTODES (TAPEWORMS)

The major pathogenic cestodes are *Taenia solium* (pork tapeworm), *Taenia saginata* (beef tapeworm), *Diphyllobothrium latum* (fish tapeworm), *Hymenolepis nana* (dwarf tapeworm), *Echinococcus granulosus,* and *Echinococcus multilocularis*. [103], [104] Cestodes have complex life cycles, living as adults in the gastrointestinal tract of their definitive mammalian hosts and as solid or bladder larvae in tissues of vertebrate or invertebrate intermediate hosts. Adult tapeworms are ribbonlike, with a scolex at the anterior end that is responsible for attachment in the small intestine, a connecting neck region, and the strobila, which is a chain of progressively developing segments known as proglottids. The number of proglottids ranges from 3 to 4000, depending on the cestode species, and the total length of adult tapeworms varies from several millimeters in the case of *E granulosus* to several meters in the case of *T saginata*.

Humans are the definitive host for *T saginata, T solium, H nana,* and *D latum,* and adult tapeworms are found in their gastrointestinal tract. On rare occasions, humans serve as a definitive host for *Hymenolepis diminuta,* which is primarily a parasite of rats and mice, and *Dipylidium caninum,* a dog tapeworm. Both of the latter infections are uncommon, light, and usually of little consequence. Humans serve as intermediate hosts for *E granulosus, E multilocularis,* and *T solium,* and cysts are found in liver, lung, brain, or other organs. In general, adult tapeworms produce little evidence of disease in their human hosts, although *D latum* can result in vitamin B_{12} deficiency. In contrast, cestodes can produce severe, life-threatening disease when humans serve as intermediate hosts.

Taenia saginata

The beef tapeworm, *T saginata,* is renowned for its length, up to 4 to 6 meters, and the occasional finding of strobila up to several feet long in the stool. [103], [104] Humans become infected when they ingest inadequately cooked beef or meat from other herbivores, such as camels, that contains the bladder or cysticercus stage. As the meat is digested, the cysticercus breaks down, releasing a scolex that typically attaches in the upper jejunum. Development of the adult worm is followed by the appearance of proglottids and ova in the stool. Cattle become infected when they ingest ova from human feces. Larvae are released in the intestine of the cow, penetrate the mucosa, and find their way to muscle, where they develop into cysticerci, completing the life cycle.

Although nonspecific symptoms such as abdominal discomfort, epigastric pain, nausea, vomiting, diarrhea, and weight loss have been attributed to *T saginata,* most infected persons are asymptomatic. There is usually no evidence of weight loss or malnutrition. The diagnosis of *Taenia* infection is made by finding ova or proglottids in the stool. The ova of *T saginata* are indistinguishable from those of *T solium* (see Fig. 126-1), and a species-specific diagnosis requires examination of the proglottids (see *Atlas,* Chapter 80). Praziquantel is the treatment of choice (see Table 126-1). [5] Niclosamide is an alternative, but resistance has been reported.

Taenia solium

Humans can serve as both the definitive host and as an intermediate host for the pork tapeworm, *T solium.* [103], [104] Infection is endemic in areas of Latin America, Africa, and Asia where hogs are raised and sanitary conditions are poor. People become infected as the definitive host when they ingest inadequately cooked pork containing infective cysticerci cellulosae. The scolex is released and attaches to the small intestine. Adult worms usually cause few, if any, symptoms, although a number of nonspecific gastrointestinal complaints have been attributed to them. Eosinophilia may be present. Mature proglottids and ova of *T solium* are released in the stool. Pigs, the normal intermediate host, become infected when they ingest ova from human feces.

The major risk with *T solium* is cysticercosis, [105], [106], [107] and [108] which develops when humans ingest ova from human feces or, theoretically, if internal autoinfection occurs in persons harboring adult worms. Ova excyst in the human or pig intestine. Larvae invade the mucosa, disseminate, and form cysticerci in the brain, subcutaneous tissue, skeletal muscle, eye, or other organs. Neurocysticercosis is the most common parasitic disease of the central nervous system in the world. It is prevalent in many Latin American countries where it is a leading cause of epilepsy and hydrocephalus. It is increasingly encountered in the United States and other industrialized countries among immigrants from endemic areas, and on rare occasion, in international travelers. Autochthonous cases of neurocysticercosis also have occurred among residents of the United States who have neither traveled internationally nor been exposed to pigs, but who have had close contact with immigrants harboring adult tapeworms. [107]

Intestinal infection is documented by finding *Taenia* ova (see Fig. 126-1) or proglottids (see *Atlas,* Chapter 80) in the stool. The ova of *T solium* and *T saginata* are indistinguishable from one another by conventional microscopy. The diagnosis of neurocysticercosis usually is based on a history of exposure in an endemic area, the clinical presentation, and computed tomographic and magnetic resonance findings. The identification of antibodies in serum or cerebrospinal fluid provides further support for the diagnosis. [108]

Praziquantel is recommended for the treatment of intestinal *T solium* infection (see Table 126-1). Niclosamide can be used, but it is associated with disintegration of the adult worm, [103] raising the theoretic possibility of autoinfection. Some physicians recommend a purge after therapy to expedite expulsion of proglottids, but there are no data to prove that it is necessary.

The guidelines for the treatment of neurocysticercosis are controversial. Both albendazole and praziquantel are effective in killing parenchymal cysticerci in the brain. [108] They have been used to treat patients with multiple, non-enhancing cysticerci. Corticosteroids are usually given concurrently to prevent cerebral edema, which may develop in response to parasite antigens released from dying cysticerci. It is important to note that corticosteroids reduce the concentration of praziquantel in the cerebrospinal fluid while increasing that of albendazole. For that reason most physicians favor the latter drug. Anticonvulsants are used in patients with seizures. The results of anthelmintic therapy in patients with extraparenchymal neurocysticercosis have been disappointing. Surgical procedures are necessary to remove intraventricular cysts and to divert cerebrospinal fluid in those who develop hydrocephalus. Albendazole and praziquantel are contraindicated in persons with

cysticerci in the spinal cord or eye.

Infection with *T solium* can be prevented by the proper disposal of human feces, treatment of infected people, inspection of pork, and thorough cooking of pork to at least 50°C before it is eaten. Freezing below -12°C (10°F) is an effective way to kill cysticerci, but they can remain viable for as long as 2 months in meat frozen at 0°C or F to -2°C (30°F). [109] Attempts are underway to develop a vaccine that would reduce or eliminate cysticercosis in pigs.

Diphyllobothrium latum

The fish tapeworm, *D latum,* is known for its ability to compete with its human host for vitamin B_{12}. [104], [110] The parasite has a complex life cycle. Adult worms reside in the human gastrointestinal tract. Ova are passed in feces. Free-swimming coracidia are released in fresh water and infect small crustaceans (copepods). When freshwater fish ingest infected copepods, procercoid larvae are released and penetrate into them. Humans become infected when they ingest raw or inadequately cooked fish, including pike, salmon, trout, whitefish, or turbot. *D latum* is found in regions of northern Europe, northern Asia, northern North America, and temperate areas of South America, such as Chile and Argentina. Some people have become infected while preparing gefilte fish, as they sampled the raw fish while adding condiments.

A number of minor gastrointestinal complaints have been associated with *D latum,* [104], [110] but most infected people are asymptomatic. Some present with macrocytic anemia due to vitamin B_{12} deficiency. The diagnosis of *D latum* is made by identifying ova or the typical broad, rectangular proglottids in the stool. The treatment of choice is praziquantel; niclosamide is an alternative (see Table 126-1). Thorough cooking or freezing of fish (-10°C [14°F] for 24 hours) prevents infection.

Hymenolepis nana

The dwarf tapeworm, *H nana,* infects as many as 20 million people worldwide. It has been diagnosed among immigrants to the United States from a number of endemic areas, including Southeast Asia. Natural hosts include humans, mice, and rats. [103], [104]

Infection with *H nana* follows ingestion of ova (see Fig. 126-1). Transmission is often hand to mouth, particularly among children, or by ingestion of food or water contaminated by feces. An oncosphere is liberated from each ova in the intestine. It penetrates a villus and develops into a cercocystis. At maturity, the cercocystis ruptures, releasing a scolex, which becomes anchored in the small intestine. Some eggs are passed in the stool. Others excyst in the lumen of the small intestine and invade villi to produce internal autoinfection.

Light *H nana* infections are usually asymptomatic or associated with vague abdominal complaints. Heavy infections may be associated with loss of appetite, abdominal pain with or without diarrhea, anorexia, flatulence, or weight loss. [104], [111] The diagnosis is made by identifying eggs in the feces (see Fig. 126-1). Praziquantel is the treatment of choice (see Table 126-1). [5], [6] Spread of *H nana* could be interrupted by improved sanitation.

***Echinococcus* Species**

Echinococcus granulosus Echinococcosis is endemic in scattered areas around the world. Adult *E granulosus* reside in the intestine of canines, their definitive hosts, and as hydatid cysts in sheep, humans, or other mammals that ingest ova from the feces of infected dogs. [104], [112] *E granulosus* is found in areas where dogs, sheep, and humans live in close proximity under conditions of poor sanitation. Infection is found in areas of Australia, New Zealand, North Africa, the Middle East, and South America. Scattered autochthonous cases have been reported from the western part of the United States. In some areas of Canada, moose and caribou serve as intermediate hosts, and the wolf is the predominant definitive host. Humans become infected with *E granulosus* when they ingest ova. Transmission is often hand to mouth from the fur of infected dogs or by way of food or water contaminated with dog feces. Ova excyst in the human intestine. The embryo invades through the intestinal wall and is carried to the liver, lung, or other organs though the portal system or lymphatics. Approximately two thirds of the cysts develop in the liver (Fig. 126-5), one fifth in the lung, and the remainder in bone, brain, muscle, eye, heart, or other organs. The typical *E granulosus* cyst contains an external hyaline cuticula, an inner germinal membrane with brood capsules, protoscoleces, which are known as hydatid sand, and daughter cysts that are replicas of the mother cyst.

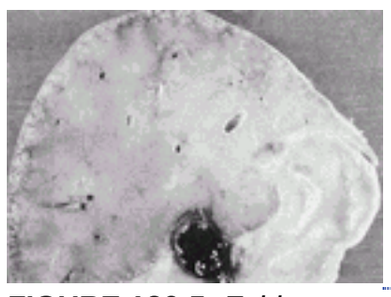

FIGURE 126-5. *Echinococcus granulosus* (hydatid) liver cysts have well-defined capsules, each with an irregular lining. These capsules contain a semi-opaque, tan liquid with hydatid sand composed of protoscoleces, each of which can mature to one adult worm. (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.)

The natural history of *E granulosus* in humans has not been defined fully. Hydatid cysts followed prospectively may shrink, remain stable, or grow slowly at a rate of approximately 0.3 cm per year. They usually do not cause symptoms until they are 10 cm or larger. Many cysts are silent or associated with vague abdominal pain. Tissue damage occurs secondary to pressure from the enlarging hydatid cyst or rupture into the biliary tract, [112] peritoneum, or lung. Obstructive jaundice, cholangitis, or biliary pain occur on rare occasions. Intra-abdominal spillage of cyst contents following spontaneous rupture can result in anaphylaxis or the seeding of the peritoneal surface. Cysts outside the liver often pose diagnostic dilemmas. The findings may be suggestive of neoplasm, particularly when cysts occur in the testes, ovary, urinary bladder, retroperitoneum, heart, or eye. The diagnosis of *E granulosus* is based on a history of exposure in an endemic area and identification of a typical hydatid cyst by ultrasound, computed tomography, or magnetic resonance imaging. [113], [114] Various serologic tests are available to detect antibodies, but they are negative in a substantial number of cases, and the specificity varies. [115] The diagnosis is confirmed histopathologically by identification of cysts or cyst contents. The World Health Organization (WHO) Informal Working Group on *Echinococcosis* recommends surgery as the treatment of choice for *E granulosus* cysts. [116], [117] Percutaneous, ultrasound-guided, cyst puncture with drainage and instillation of a scolicidal agent, such as hypertonic saline or alcohol, followed by aspiration has been advocated as an alternative for simple, uncomplicated cysts. [118], [119] Anaphylaxis has been reported as a result of leaked cyst material, but it is uncommon. [120] Laparoscopic approaches [121] have also been used. Injection of formalin into cysts should be avoided as it has been associated with sclerosing cholangitis and death. [122] Of major concern with any drainage procedure is spillage of cyst contents, resulting in anaphylactic shock or peritoneal implantation. Treatment with high doses of albendazole or mebendazole before surgery or percutaneous drainage reduces the risk of peritoneal implants. [123] Albendazole, administered at high doses for several months, is an alternative to surgery or percutaneous drainage. It suppresses cyst growth in the majority of cases and cures approximately one fourth of cases. It is the treatment of choice for patients with inoperable lesions. [116], [124] Mebendazole also has activity, but it is less well absorbed than albendazole. Liver transplantation has been performed successfully in rare cases. Progress has been made in controlling transmission of *E granulosus* in endemic sheep-raising areas by treating intestinal infections in dogs and keeping them from eating the uncooked viscera of sheep or carrion. *E granulosus* has a relatively low reproductive rate in its domestic life cycle, and it is thought to be relatively unstable and potentially susceptible to eradication. [125]

Echinococcus multilocularis This *Echinococcus* species is endemic in northern regions of North America, Central Europe, northern Asia, and Japan, and has a life cycle involving foxes, sled dogs, and other canines as definitive hosts, and mice, voles, and other rodents as natural intermediate hosts. [104], [112] Humans, who serve as intermediate hosts become infected by eating wild plants or berries contaminated with canine feces or by hand-to-mouth spread from the fur of infected dogs. *E multilocularis* has been recognized as an emerging zoonosis in Central Europe where infection has been documented among foxes, which are increasing in number, as well as domestic dogs and cats. [126] In humans, *E multilocularis* is typically found as an alveolar hepatic cyst, a spongy mass of fibrous tissue and small cysts with a jellylike matrix. These cysts resemble neoplasms in their ability to spread locally, metastasize, and invade bile ducts and vessels. The initial symptoms of *E multilocularis* are often vague. Hepatomegaly, abdominal pain, obstructive jaundice, and focal necrosis follow. Allergic complications are rare. Late complications include secondary bacterial infections, biliary cirrhosis, chronic Budd-Chiari syndrome, and portal thrombosis. The diagnosis is based on a history of exposure in an endemic area and ultrasonographic, computed tomographic, or magnetic resonance findings. The presence of anti- *Echinococcus* antibodies is suggestive of the diagnosis, but a negative serologic test does not exclude the diagnosis. [127] Surgical resection is the only reliable means of curing *E multilocularis* infections. [116] Prolonged albendazole treatment usually is administered to those who undergo surgery as well as to those who have inoperable lesions (see Table 126-1). [116] Liver

transplantation has been necessary in rare cases. *E multilocularis* can be prevented by avoiding uncooked, potentially contaminated wild fruits, by worming sled dogs, and by washing hands after exposure to potentially infected domestic animals.
Other *Echinococcus* Species A limited number of cases of polycystic hydatid disease resulting from *Echinococcus vogeli* and *Echinococcus oligarthus* have been reported from Central and South America. [128] *E vogeli* is a zoonotic parasite that infects wild canines and rodents in rural areas. Surgical resection of cysts is the only proven therapy.

TREMATODES (FLUKES)

Trematodes have complex life cycles with humans, and in some instances other animals, serving as the definitive host. Fresh-water snails are the primary intermediate hosts. *Schistosoma* species, which are often referred to as blood flukes, are among the most prevalent of the trematodes that infect humans. *Schistosoma mansoni* and *Schistosoma japonicum,* which live in mesenteric venules, are major causes of intestinal and liver pathology. Less prevalent are *Schistosoma intercalatum* and *Schistosoma mekongi*. *Schistosoma haematobium* primarily affects the urinary tract, although it occasionally involves the appendix or bowel. *Schistosoma* species enter the skin when humans come in contact with fresh water. Other pathogenic trematodes live in the biliary tract or the small intestine. They include the liver flukes *Clonorchis sinensis, Opisthorchis viverrini, Fasciola hepatica,* and *Metorchis conjunctus,* and the intestinal flukes *Fasciolopsis buski, Heterophyes heterophyes,* and *Metagonimus yokogawai*. They are acquired by eating the second intermediate hosts, fresh water fish, or in the case of *F hepatica* and *F buski,* water plants.

Schistosoma Species

As many as 200 million persons are infected with schistosomiasis worldwide. [129] *Schistosoma* species have complex life cycles. The delicate adult worms live as pairs within mesenteric venules in their definitive human host (see *Atlas,* Chapter 80). The adult female spends her life in the gynecophoric canal of the male. From 300 eggs in the case of *S mansoni* to 3500 in the case of *S japonicum* are produced daily. Lytic enzymes secreted by the ova allow them to penetrate the wall of the vessel to reach the lumen of the intestine. They are excreted in the stool. Miracidia are released when ova hatch in fresh water.

The next phase of infection begins when miracidia penetrate snails of the appropriate genera. After asexual development, fork-tailed cercariae emerge. They enter human skin when contact is made with fresh water during bathing, fishing, or other water-related activities. The tails of the cercariae are lost, and the resulting schistosomulae make their way through the vasculature to the portal circulation. Approximately 3 weeks later, *S mansoni* migrates predominantly to the superior mesenteric veins and *S japonicum* to the inferior mesenteric veins. Adult worms can live as long as 30 years in humans. [130]

Epidemiology *S mansoni* is endemic in Africa and many areas of Latin America. Transmission frequently is associated with dams, reservoirs, and irrigation systems. Scattered foci are present in the Middle East. *S intercalatum* is found in areas of Western and Central Africa. *S japonicum* is endemic in the Far East, China, coastal river valleys in Japan, numerous islands in the Philippines, and Indochina. Its epidemiology differs from other *Schistosoma* species in that it has a number of nonhuman hosts, including cows, dogs, and pigs. [131], [132] *S mekongi* is found along the Mekong River in Indochina and is similar to *S japonicum*.
Clinical Manifestations Invasion of skin by cercariae is usually asymptomatic, but a mild, pruritic dermatitis may result. Acute schistosomiasis, or Katayama fever, was first described among people with primary *S japonicum* infections. It can occur, however, with *S mansoni* and other *Schistosoma* species. [133] The time from invasion by cercariae to onset of systemic symptoms ranges from 6 to 8 weeks with *S japonicum* to 3 to 8 weeks with *S mansoni* and coincides with the start of egg production. Katayama fever is a serum sickness–like syndrome characterized by fever, malaise, urticaria, abdominal discomfort, diarrhea, weight loss, cough, mild hepatosplenomegaly, lymphadenopathy, and eosinophilia. Symptoms can persist for several months. Chronic schistosomiasis develops as a consequence of the inflammatory response directed against schistosome eggs that are deposited in mesenteric vessels, carried to the liver through the portal circulation, or reach the lungs or other sites by way of the systemic circulation. The degree of pathology depends on the worm burden and the host's genetically determined immune response to the eggs, a complex mix of Th1 and Th2 cytokines that result in varying degrees of inflammation and fibrosis. [134] Inflammation, hypertrophy, and ulceration of the mucosa characterize intestinal disease caused by *Schistosoma*. It is often asymptomatic, but infected persons may experience abdominal pain, diarrhea, and occult or gross blood in their stools. Intestinal polyps and strictures may be encountered late in disease. [135] Eggs that are swept to the liver through the portal circulation elicit a granulomatous inflammatory response and “Symmers” periportal fibrosis, which varies in intensity. [136] A subset of persons progress to severe “pipe-stem” portal fibrosis with presinusoidal portal hypertension. [137] Hepatomegaly and splenomegaly are common in heavily infected persons. The pathology in the liver is predominantly mesenchymal; the parenchyma and liver function are preserved until late in the disease. Bleeding from esophageal varices and ascites are complications of advanced hepatosplenic schistosomiasis. Heavy infections in children can be associated with malnutrition and growth retardation. [138], [139] Portosystemic anastomoses in persons with hepatosplenic schistosomiasis can result in shunting of eggs to the pulmonary vasculature, where granulomas and fibrosis may progress to chronic obliterative arteritis, pulmonary hypertension, and eventually cor pulmonale. [140] Granulomatous responses to eggs occasionally are encountered in other organs, including the gallbladder, where they have been associated with cholecystitis and cholelithiasis. Ectopic *S mansoni* egg-related granuloma have been identified in the spinal cord, [141], [142] and *S japonicum*-related granuloma in the central nervous system. [143] A small subset of patients infected with *S mansoni* develop immune complex glomerulonephritis that can progress to the nephrotic syndrome. This complication is seldom observed with *S japonicum.* Schistosomiasis also has been associated with prolonged or relapsing *Salmonella* bacteremia. The bacteria apparently colonize adult worms and are thus protect them from the effects of systemically administered antibiotics. [144] *S haematobium* resides primarily in venules of the vesical plexus, but on occasion it too involves the gastrointestinal tract. *S haematobium* has been found in association with acute appendicitis, although its role in pathogenesis is uncertain, [145], [146] and it has been reported as a cause of asymptomatic appendiceal calcification. [147]
Diagnosis, Treatment, and Prevention The diagnosis of schistosomiasis is confirmed by identifying ova in feces or biopsy material. Direct smears of stool are of relatively low sensitivity; Kato thick smears using 20 to 50 mg of fecal material are more sensitive. [148] Diffuse or patchy mucosal hyperemia, friability, and occasionally schistosomal polyps may be observed at colonoscopy. [149] A rectal biopsy may be necessary to document light infections. Ultrasonography has been widely used to assess hepatosplenic involvement and responses to chemotherapy. [150], [151] Several serologic assays are available and provide suggestive evidence of schistosomiasis, [152] but they are of no help in determining the severity or duration of infection. Praziquantel is recommended for the treatment of all *Schistosoma* species (see Table 126-1). [5], [6] It is effective in killing the parasite and can decrease periportal fibrosis, hepatomegaly, and splenomegaly, [150], [151] but treatment does not reverse all of the effects of chronic schistosomiasis. Concern has been raised about the possible emergence of resistance to praziquantel. [153] Oxamniquine is an alternative for the treatment of *S mansoni*. Endoscopic sclerotherapy or surgical decompression have been used successfully to treat bleeding esophageal varices due to portal hypertension. [154], [155] Control measures in endemic areas include improved sanitation and disposal of human feces, avoidance of contaminated fresh water, use of molluscacides, and mass treatment programs. [132] Despite these efforts, schistosomiasis remains prevalent in many endemic areas. There is hope that an effective vaccine can be developed, but one is not currently available.

Liver Flukes

The liver flukes, *Clonorchis sinensis* and *Opisthorchis viverrini,* are found in Japan, China, Indochina, South Korea, and Taiwan among those who eat raw freshwater fish. [156], [157] and [158] It has been estimated that 6 to 7 million people are infected in northeastern Thailand alone. Adult liver flukes live in the biliary tract. Eggs are released into the bile and passed in the stool. Snails are the first intermediate host, and freshwater fish are the second. Humans become infected when they eat raw or incompletely cooked fish. Freed larvae enter the common bile duct from the duodenum. These flukes have life spans of 10 to 30 years. In the United States, *C sinensis* and *O viverrini* are periodically encountered among immigrants from Southeast Asia.

Slight liver enlargement and tenderness may be observed at the onset of infection. The major pathological changes of chronic infection are in the bile ducts. Initially, there is desquamation of the biliary epithelium, followed by hyperplasia and eventually adenomatous proliferation. Fibrosis and stricture of the bile ducts may follow.

The precise relationship of infection to gastrointestinal symptoms is uncertain. Light infections are usually asymptomatic. In heavy infections, dyspepsia, abdominal pain, anorexia, hepatomegaly, icterus, edema, or diarrhea may occur. Pyogenic cholangitis, cholelithiasis, chronic cholecystitis, pancreatitis, and cholangiocarcinoma are potential long-term complications. [159], [160]

The diagnosis is confirmed by identifying ova in the stool or bile (see Fig. 126-1). Ultrasonographic examination may reveal liver enlargement; dilation, sludge, and thickening of the wall of the gallbladder; dilation of intrahepatic bile ducts; or evidence of cholangiocarcinoma. [161] Endoscopic retrograde cholangiopancreatography may show small, irregular filling defects in the common bile duct, tortuosity and irregular dilation of the intrahepatic ducts, or blunting of the terminal branches of the biliary tree. [162] Praziquantel is the drug of choice for the treatment of *C sinensis* and *O viverrini* (see Table 126-1).

Fasciola hepatica, which is responsible for “liver rot” in sheep and other herbivorous mammals, including humans, is cosmopolitan in sheep- and cattle-raising areas of the world. [156] Snails are intermediate hosts. Humans become infected when they eat wild watercress contaminated with encysted metacercariae. Larvae excyst in the small intestine, penetrate the gut wall, migrate through the peritoneal cavity, and enter the liver through the capsule. They may wander through the human liver for periods of 6 to 9 weeks before finally penetrating into the bile ducts, where they spend their adult lives. Acutely infected people may experience fever, hepatomegaly, abdominal pain, weight loss, anemia, and eosinophilia. [163] Jaundice and biliary colic are manifestations of chronic disease. Serologic assays provide presumptive evidence of infection. The diagnosis is confirmed by identifying ova in bile, duodenal aspirates, or stool. Multiple low-density areas may be observed in the liver by computed tomography. They are often subcapsular in location, arrayed in a tractlike fashion, and evolve slowly. [164] Unlike the other liver flukes, therapeutic failures are common in patients with *F hepatica* treated with praziquantel. Bithionol is used for treatment (see Table 126-1); [5], [6], [165] triclabendazole, a benzimidazole used for veterinary infections, is also effective.

On rare occasion, *Opisthorchis felineus* or other liver flukes that primarily infect animals are found in humans. *Dicrocoelium dendriticum,* a liver fluke of sheep and cattle, has been reported in humans who have ingested infected field ants, the second intermediate host. Most infections are asymptomatic, but hepatobiliary disease and eosinophilia can result. A common-source outbreak of acute infection caused by the North American liver fluke, *Metorchis conjunctus,* occurred in Montreal, Canada, among persons who ate raw fish (sashimi) prepared from white suckers. [166] The illness was characterized by persistent upper abdominal pain, low-grade fever, eosinophilia, and elevated liver enzymes. Ova were present in the stool. The symptoms resolved after treatment with praziquantel.

Intestinal Flukes

The intestinal fluke, *Fasciolopsis buski,* has as its definitive hosts humans, hogs, and occasionally dogs. [167] It is endemic in Asia. The adult flukes attach to the mucosa of the upper small intestine and release ova into the feces. Snails are the intermediate host. Cercariae encyst on water plants such as the water caltrop, water hyacinth, water chestnut, or water bamboo, which are ingested raw by humans. Inflammation, ulceration, and mucosal abscesses may develop in the small intestine. Epigastric pain, nausea, and diarrhea of varying severity are reported, but many people are asymptomatic. The diagnosis is made by identifying ova in the stool. Praziquantel is recommended for treatment (see Table 126-1). [5], [6]

Many species of the trematode family Heterophyidae, including *Heterophyes heterophyes* and *Metagonimus yokogawai,* are parasites of humans and other fish-eating mammals. The first intermediate hosts are snails, and the second intermediate hosts are freshwater or brackish-water fish. Humans become infected from eating raw or inadequately cooked fish. *H heterophyes* is encountered in Egypt, the Middle East, and Asia. It does relatively little damage to the intestine. Colicky pain, abdominal tenderness, and diarrhea are experienced by some, particularly those with heavy infections. Eosinophilia may be present. A case of *Heterophyes* infection was reported in the United States in a woman who ate sushi imported from the Orient. [168] *M yokogawai* is found primarily in the Far East, where it has infected American travelers, causing diarrhea. [169] *M yokogawai* on rare occasions is associated with the formation of egg-related granuloma at distant foci, such as the heart or central nervous system. Intestinal fluke infections are typically diagnosed by finding ova in the feces. Praziquantel is the treatment of choice (see Table 126-1). [5], [6] Infection can be prevented by eating only thoroughly cooked fish.

On rare occasions, people living in the Pacific Northwest or Siberia become infected with *Nanophyetus salmincola* when they ingest raw or inadequately cooked salmonid fish. Abdominal pain, diarrhea, nausea, and vomiting may result, along with eosinophilia. [170] Praziquantel has been used for therapy (see Table 126-1). Infection with one of many *Echinostoma* species may also occur after ingestion of raw, infected snails, fish, or amphibians, but symptoms, when present, are usually mild. [171]

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CHAPTER 127

Elizabeth M. Petty

GASTROINTESTINAL AND HEPATIC MANIFESTATIONS OF SPECIFIC GENETIC DISORDERS

Genetic factors contribute to the pathogenesis of many gastrointestinal (GI) and hepatic disorders. Additionally, many inherited multisystemic conditions and syndromes are associated with specific GI or hepatic pathology. While the impact of genetic contributions to these conditions varies, genetic and nongenetic factors (e.g., environmental, epigenetic) interact at least to some degree to influence phenotypic expression of most conditions with GI or hepatic manifestations. Clinicians need to be aware of genetic contributions to these conditions. Recognition of a highly penetrant well-understood genetic contribution to a disease could impact clinical management of the patient and often has significant implications for other family members. To help recognize genetic contributions to GI or hepatic disorders, it is helpful to review a comprehensive family history, relevant medical records (including pathology reports, laboratory studies, and clinical summaries) of the patient, and, in some cases, medical records of other affected family members.

As discussed in Chapter 54, the family history may reveal a specific Mendelian inheritance pattern (e.g., autosomal dominant, autosomal recessive, X-linked recessive, or X-linked dominant) or non-Mendelian mode of inheritance (mitochondrial/maternal/cytoplasmic or imprinted) that suggests a discrete inherited genetic contribution. It is sometimes difficult to recognize genetic contributions when there is a small family, genetic heterogeneity, common phenocopies (sporadic conditions that mimic inherited disorders), variable expressivity of manifestations, or incomplete penetrance. Determining the genetic contribution to a GI or hepatic disease that is multifactorial (e.g., where significant environmental or epigenetic factors interact with an individual's underlying genetic constitution) or polygenic (e.g., where several genetic factors, including modifying genes, interact to contribute to the disease) is even more challenging. Despite these challenges, it is clinically relevant to assess genetic contributions to the disease process.

The purposes of this chapter are to:

Provide a basic framework as to when, how, and why to consider genetic factors.
Review several well-known genetic syndromes that may present with significant GI or hepatic manifestations, most of which are highly penetrant single-gene disorders with Mendelian patterns of inheritance that have additional non-GI manifestations.

Some of these syndromes present early in life (e.g., inborn errors of malabsorption), while others are characterized by their adult-onset presentation (e.g., dominant colon cancer syndromes). It is important to be aware of these syndromes to ensure that patients are appropriately diagnosed and offered the most appropriate medical management. For example, in an adult with Ehlers-Danlos syndrome (EDS) type IV (often referred to as the "vascular type" of EDS), an autosomal dominant syndrome characterized by type III collagen abnormalities, who presents with acute severe abdominal pain, rupture of the bowel and dissection of an abdominal artery must be immediately suspected to reduce disease-associated morbidity and early mortality.

Most of the diseases presented in this chapter are summarized in an abbreviated table format for easy reference. More comprehensive discussions of the pathophysiology and treatment for common GI and hepatic diseases mentioned in this chapter are covered in more detail elsewhere in this text (see the references for each topic, Chapter 128, and the). For more information about less common genetic conditions several excellent genetic textbooks and on-line genetic databases are referenced for additional, more detailed, information about the particular syndromes and diseases. [1], [2], [3], [4] and [5] Specifically, the *Online Mendelian Inheritance of Man* (OMIM) (http://www3.ncbi.nlm.nih.gov/Omim/), GeneTests-GeneClinics Web site (http://www.geneclinics.org/), and the Center for Disease Control's HuGE Net (http://www.cdc.gov/genetics/hugenet/reviews.htm) are excellent on-line database references that provide regularly updated information about many genetic syndromes and include more detailed up-to-date reference lists. All information for this chapter was obtained through these on-line searches of these databases with review of their listed references and genetic textbooks. [4], [5] Genetic contributions to common GI and hepatic conditions that have complex multifactorial or polygenic etiologies (e.g., inflammatory bowel disease, cirrhosis) are covered in other disease-specific chapters throughout this text (see).

PRACTICING GENETIC MEDICINE (See Also Chapter 54)

Basic genetic principles must be well understood by the clinician in order to provide accurate diagnostic, prognostic, and therapeutic services for individuals with genetic conditions and other at-risk family members. Once the genetic basis of a disease is recognized, preventive screening, medical or surgical management, genetic counseling, patient and family education, and referrals can be implemented as appropriate. DNA testing is now available for dozens of conditions that have prominent GI or hepatic manifestations and the list will continue to grow. With completion of the Human Genome Project and identification of the genetic codes for all of our estimated 30,000 to 35,000 human genes, genetic discoveries relevant to gastroenterologists and hepatologists will continue to be revealed at an unprecedented pace. Genetic testing can serve various purposes, from molecular confirmation of a suspected disease to facilitate more appropriate medical management and accurate recurrence risk counseling as in the case of hemochromatosis, or, predictively, to more accurately predict one's own risk of developing colon cancer. Thus, this new era of molecular medicine has already made an undeniable impact in medicine, especially as related to predictive genetic testing.

An example of this is seen in familial adenomatous polyposis (FAP), classically a highly penetrant, autosomal dominant condition caused by deleterious mutations in the adenomatous polyposis coli gene (*APC*) located on the long arm of chromosome 5 (see Chapter 90). In its classic form, individuals with one copy of a mutated *APC* gene will develop colon cancer in early to mid-adulthood after having hundreds to thousands of polyps that began developing in adolescence. At-risk individuals benefit from genetic counseling, education, and testing to better understand their risks of developing cancer. In over 80% of affected individuals, causative *APC* mutations can be detected, allowing robust and reliable predictive testing for at-risk family members. Specific preventive management strategies (e.g., serial colonoscopies, colectomies) can greatly reduce the risk of colon cancer in mutation carriers. Individuals lacking the mutation that segregates in affected family members, are spared the high risk of developing early-onset colon cancer and do not need the surveillance and surgical interventions recommended for mutation carriers. Thus, it is both clinically and economically desirable to offer predictive genetic testing to individuals at risk for a highly penetrant, late adolescent/early adult-onset disorder such as FAP, where interventional management can significantly reduce morbidity and early mortality. In contrast to highly penetrant *APC* mutations responsible for classic FAP, recognition of a relatively common polymorphism associated with a hypermutable tract of DNA in the *APC* gene in Ashkenazi Jews confers only a mildly increased risk of colon cancer in individuals with the polymorphism. Thus, understanding the relative risks for colon cancer associated with various mutations is critical for patients as it may have profound effects on their clinical management. With a better understanding of genetic mechanisms in health and disease, the number and use of genetic tests available will continue to grow and, ultimately, individually targeted molecular-based therapies may become a therapeutic reality for many GI and hepatic disorders.

Issues in genetic counseling, education, and testing will be raised throughout this chapter. As discussed more fully in Chapter 54, genetic counseling involves discussing diagnostic, prognostic, recurrence risk and medical management strategies, genetic testing issues, and related psychosocial concerns with referral to

appropriate support systems and educational resources. Although board-certified geneticists and genetic counselors currently provide genetic counseling, the growth of genetic knowledge will require that virtually all health care professionals will need to provide some basic level of genetic counseling in the future. For complex cases and rare genetic conditions specialized genetic services are available through most academic medical centers.

Since genetic testing evolves with laboratory methods becoming increasingly more sensitive, specific, and cost-effective, detailed discussion of specific genetic tests will not be addressed here. It is important to remember, however, that each test has its own strengths and weaknesses, and the specificity and sensitivity will vary considerably depending on the particular genetic test. For some disorders, such as hereditary nonpolyposis colorectal cancer (HNPCC), an adult-onset, autosomal dominant disease where genetic heterogeneity exists, clinicians must now choose between a number of different kinds of available genetic tests to identify the most appropriate, sensitive, specific, and cost-effective test for any one individual or family. On-line genetic testing laboratory databases, such as GeneTests, for health care professionals, provide up-to-date information on laboratories that offer research or clinical genetic testing for diseases with a genetic component. [2] Individuals needing more information should consult with local genetic centers and gather up-to-date information provided by national disease-associated organizations.

While DNA tests may be the most appropriate diagnostic test to offer an individual being evaluated for a genetic condition, this is not always the case. Consider, for instance, a disease like neurofibromatosis type 1 (NF1), an autosomal dominant, neurocutaneous disorder that may be associated with GI symptoms caused by the mass effect of internal plexiform neurofibromas or by episodes of vascular ischemia caused by vascular lesions in the intestinal bed. Close to 100% of older children and adults with NF1 have a clinically recognizable diagnosis while current molecular testing for NF1 is only about 70% sensitive for identifying mutations in affected individuals—much less than a good clinical genetics examination. DNA testing in a clinically diagnosed individual who presents for management of their GI symptoms currently offers no benefit to the patient as there are relevant genotype/phenotype correlations that would offer insight into their prognosis or management. Molecular testing for NF1, however, may be quite useful if the same individual is interested in prenatal testing or if the patient has very young children and the diagnosis cannot be firmly established by clinical examination.

In all cases, genetic testing encompasses more than a simple laboratory analysis and generally includes pretest counseling and education, provision of informed consent, accurate interpretation and discussion of implications, follow-up after test results, and post-test education, management, and support (Table 127-1). This is especially true when molecular testing is used to more accurately determine a healthy individual's genetic risk for developing a disease, as in predictive testing of an asymptomatic individual who, by virtue of their family history or their ethnicity, is at risk of having inherited a particular mutation. Such testing is currently employed in the evaluation of inherited colon cancers, as described previously.

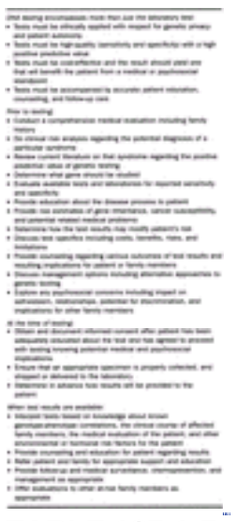

TABLE 127-1 Foundations for the Appropriate Use of Genetic Tests

WELL-DEFINED GENETIC DISORDERS WITH PROMINENT GASTROINTESTINAL OR HEPATIC PRESENTATIONS

Malabsorption Disorders (See Also Chapter 77)

Some genetic syndromes involve defects of intestinal absorption (Table 127-2). Most are disorders of brush border enzymes or transport proteins that transfer sugars, amino acids, electrolytes, lipids, vitamins, and minerals from the lumen of the gut into our circulation. They may involve malabsorption of a singular, specific substance, such as in lactase deficiency, or general malabsorption of several substances, such as in Hartnup disease. The other large group of genetic malabsorption syndromes involves disorders of the exocrine pancreas. Cystic fibrosis is most common, followed in frequency by the Shwachman-Bodian syndrome and the much rarer Johanson-Blizzard syndrome.

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TABLE 127-2 Genetic Disorders of Intestinal Malabsorption

Defects in Carbohydrate Metabolism

Congenital lactase deficiency. Patients with congenital lactase deficiency (i.e., disaccharide intolerance II) present with watery diarrhea, usually after the first milk feeding. Lactase, an enzyme with lactase and phlorizin hydrolase activity, appears to be absent in the duodenal mucosa of affected infants, but the duodenum is otherwise histologically and enzymatically normal. The DNA sequence of the human lactase-phlorizin hydrolase gene, *LCT,* maps to chromosome 2q21.

Later onset lactase deficiency. Lactase activity is generally around 10% of maximal activity in adults with lactase deficiency. Approximately three fourths of non-Caucasian adults are lactase-deficient and 5% to 30% of Caucasians, depending on their ethnicity, have some degree of lactase deficiency. In ethnic groups where late-onset lactase deficiency is common (e.g., >50% prevalence), affected individuals between 1 and 5 years of age often present with diarrhea and crampy abdominal pain after the ingestion of lactose-containing foods. In Caucasians with hypolactasia, symptoms generally arise during adolescence. The diagnosis is most often confirmed by the noninvasive techniques of oral lactose tolerance tests or breath tests. Since adult lactase activity variation in different ethnic groups is independent of lactose intake, the differences are largely genetic. The molecular defect is a decreased amount of active brush border lactase protein, which is probably regulated at the level of gene expression. Two phenotypes have been described: type I is characterized by a decrease in mRNA coding for lactase-phlorizin hydrolase; type II is characterized by abnormal posttranslational processing of lactase-phlorizin hydrolase, with retention of the molecule in the endoplasmic reticulum. Since hypolactasia is autosomal recessive it would be expected that only siblings would be affected in families. However, parents are commonly affected due to the high prevalence of the gene for hypolactasia in the population, which results in homozygotes marrying heterozygous carriers.

Congenital lactose intolerance. Congenital lactose intolerance is a serious autosomal dominant disorder characterized by vomiting, failure to thrive, dehydration, renal tubular acidosis, disaccharidiuria, aminoaciduria, liver damage, cataracts, and brain damage in infants. Unlike congenital lactase deficiency, it can be fatal unless diagnosed and managed promptly. The disease is caused by the abnormal absorption of lactose in the stomach rather than the duodenum. Lactose is hypothesized to be toxic to the liver and brain, analogous to the toxicity of galactose-1-phosphate in galactosemia. Treatment consists of lactose avoidance. The disorder spontaneously resolves between 12 and 18 months of age.

Sucrose isomaltose malabsorption/disaccharide intolerance I. The autosomal recessive sucrose-isomaltose malabsorption disorder (i.e., sucrase– *a*-dextrinase deficiency) presents with symptoms similar to lactase deficiency, except that they are evoked by sugar, not milk. Symptoms range from severe diarrhea in infants to

occasional stomach upset in adults. Ingestion of sweetened foods and fruits provokes symptoms but tolerance to the sugars increases with age. Starch is generally well tolerated. In general, homozygotes have severe enzyme deficiency with lifelong symptoms, but heterozygotes with intermediate enzymatic activity may have mild symptoms as infants and no symptoms as adults. The disorder affects 10% of the Inuit of Greenland but only 0.2% of North Americans. The diagnosis is made using the same techniques as used in lactase deficiency. Ingesting live yeast (fresh baker's yeast has sucrase activity), preferably on a full stomach, can ameliorate the symptoms after sucrose ingestion. Studies suggest that the primary defect is in the sucrase with secondary effects in isomaltase. Defects in intracellular processing of the enzyme complex have been demonstrated: accumulation in the endoplasmic reticulum, blocked transport in the Golgi, and transport of a catalytically altered enzyme to the cell surface, illustrating the heterogeneity of this relatively uncommon, single-gene disorder. The sucrase-isomaltase gene (*SI*) maps to chromosome 3q25-26.

Glucose-galactose malabsorption. The autosomal recessive condition of intestinal monosaccharide transport deficiency presents similarly to intestinal disaccharidase deficiency. In glucose-galactose malabsorption, diarrhea and dehydration occur after ingestion of carbohydrates containing one or more of these sugars. Tolerance to these sugars improves with age. Acidic stools, containing glucose and galactose, suggest the diagnosis. Some patients also have a mild defect in renal tubular reabsorption of glucose. It is clinically distinguished from disaccharidase deficiency as there is no significant rise in plasma glucose during a glucose-galactose oral tolerance test. Treatment consists of substituting fructose for other dietary carbohydrates. The transport mechanism in the small intestine is dysfunctional where the mucosa is unable to take up glucose and galactose. Mutations in the glucose-galactose transporter gene (*SGLT1,* also known as *SLCA5A1*—solute carrier family 5) which maps to chromosome 22q13 are responsible for the condition.

Trehalase deficiency. Trehalase deficiency, an autosomal recessive disorder characterized by diarrhea and vomiting after ingestion of trehalose-containing foods, is rare in Caucasian Americans but is found in 10% to 15% of the Inuit of Greenland. Trehalose is a disaccharide found in insects and mushrooms. Deficiency can be detected by oral tolerance tests. The gene encoding human renal trehalase (*TREH*) encodes a brush border membrane glycoprotein and maps to 11q23.

Defects in Amino Acid Transport Proteins are normally absorbed by two major mechanisms: transport of liberated free amino acids by group-specific, sodium-dependent amino acid transport systems and uptake of unhydrolyzed peptides independent of the specific entry mechanisms. At least three major group-specific active transport systems exist: monoamine monocarboxylic (neutral) amino acids; dibasic amino acids and cystine; and dicarboxylic (acidic) amino acids. The genetic disorders of amino acid absorption involve deficiencies in one or more of these transport mechanisms.

Iminoglycinuria. Iminoglycinuria is due to benign autosomal recessive inborn errors of imino acids (i.e., proline and hydroxyproline) and glycine absorption in the kidneys and consists of five subtypes. Two subtypes also involve the intestine. This autosomal recessive malabsorption condition is often asymptomatic but has been associated with mental retardation and gyrate atrophy of choroid and retina. When any of the three amino acids is ingested, excretion of the other two increases, indicating a group-specific membrane transport defect. Heterogeneity in the disorder is suggested by the fact that some homozygotes show no proline transport defect. Obligate heterozygotes may be hyperglycinuric or not. At least four alleles for the disorder are suspected, but it is not known whether they are all at the same locus. They occur most often in Ashkenazi Jews and the frequency among Caucasians is about 1 in 15,000, giving a heterozygote frequency of about 2%. The existence of an autosomal dominant form of glycinuria with oxalate nephrolithiasis, seen most often in Ashkenazi Jews, may be a particular heterozygote form of iminoglycinuria that predisposes some persons to urolithiasis.

Hartnup disease. Hartnup disease is a rare heterogenous autosomal recessive disorder with an incidence of 5 cases per 100,000 individuals. It causes virtually no symptoms in more than 90% of affected persons in the United States, probably reflecting the adequate diet that most of this population consumes. In symptomatic individuals, an intermittent, red, scaly, pellagra-like rash after sun exposure and attacks of cerebellar ataxia, sometimes accompanied by psychiatric disorders and seizures, develop in childhood. Low-normal or subnormal intelligence has also been reported. The main feature, and the only diagnostic test, is massive aminoaciduria involving neutral and aromatic monoamine monocarboxylic amino acids sharing a common renal reabsorption mechanism in the proximal tubule. Patients have diminished capacity for jejunal absorption of these amino acids because of a brush border transport defect, causing retention of the amino acids in the intestine for abnormally long periods which allows intestinal bacteria to convert them into dipeptide decomposition products, some of which are absorbed. There is inadequate nicotinamide synthesis because of diminished tryptophan absorption, which causes pellagra. The decomposition is toxic to the central nervous system, and coupled with pellagra, produces the cerebellar and psychiatric manifestations. A general nutritional deficiency results from the diminished availability of essential amino acids. Liver biopsy shows extensive fatty liver without infiltration of inflammatory cells or fibrosis. Patients are treated by the administration of niacin. The gene, *HND,* maps to chromosome 5p15 based on the isolation of the mouse gene hyperphenylalaninemia (*hph2*) that produces a murine disorder similar to human Hartnup disease.

Cystinuria. Cystinuria is an autosomal recessive disorder characterized by cystine, lysine, ornithine, and arginine transport abnormalities in the renal tubule and intestinal epithelium. The clinical manifestations are confined to nephrolithiasis resulting from the insolubility of cystine in the urine, and expression of the disease peaks in the second and third decades. Men are more severely affected than women. It is relatively rare, with an incidence in the United States of 1 in 15,000 individuals (based on newborn screening studies) but has an incidence of 1 in 2500 among Israeli Jews of Libyan origin. Three forms of cystinuria are recognized, and investigation of the radiopaque stones that present as hexagonal cystine crystals in the urine in all types leads to the diagnosis. Classification is based on excretion rates of amino acids in obligate heterozygotes. In type I, heterozygotes excrete no excess amino acids, in type III, excretion of up to twice the normal amount is noted, and in type II, the rarest form of the disease, excretion of 9 to 15 times the normal amount is noted. Type I cystinuria maps to chromosome 2p16 where the culprit gene, *SLC3A1,* was identified. The type II and III cystinuria gene, *SLC7A9,* maps to chromosome 19q13. Treatment is directed at reducing the concentration of cystine in urine by increasing urine volume, increasing solubility by alkalinization, and, as needed, reducing free cystine excretion by medical therapy, historically D-penicillamine.

Lysinuric protein intolerance/dibasic aminoaciduria II. Lysinuric protein intolerance, or dibasic aminoaciduria II, is a rare serious autosomal recessive disorder beginning in infancy when cow's milk is introduced. It is characterized by abnormal transport of dibasic amino acids across the basolateral membranes of intestinal and renal tubular cells. Symptoms include vomiting, diarrhea, failure to thrive, severe growth retardation, hepatosplenomegaly with ultimate cirrhosis, lens opacities, mental retardation, sparse and brittle hair, osteopenia, attacks of stupor, and sometimes hyperextensible skin and joints. It is caused by the malabsorption of arginine, lysine, and ornithine as demonstrated after oral protein loading. Reduced tubular reabsorption of these amino acids also occurs in the kidney with demonstrable lysinuria. Some heterozygotes manifest partial intestinal malabsorption after an oral protein load. Quantitative plasma amino acids reveal low levels of arginine, lysine, ornithine, leucine, and tyrosine; normal serine and citrulline levels; and elevated glutamine and alanine levels. Dibasic aminoaciduria without cystinuria, impaired intestinal absorption of basic amino acids, and protein intolerance are diagnostic features. Hyperammonemia results from deficient hepatic uptake of the amino acids producing a deficiency of some intermediate products of the urea cycle. Treatment consists of protein restriction with citrulline and lysine supplements. Some of the abnormalities resolve on this therapy. It is caused by mutations in the amino acid transporter gene, *SLC7A7,* which maps to chromosome 14q11.

Blue diaper syndrome. Blue diaper syndrome is a rare autosomal or X-linked recessive disorder of intestinal, but not renal, tryptophan malabsorption. Infants present with failure to thrive, recurrent fever, infections, irritability, and constipation in addition to the bluish discoloration of the diapers. Hypercalcemia and nephrocalcinosis also occur because of increased absorption of calcium. The unabsorbed tryptophan is converted to indoles in the intestine by bacteria. The indoles are absorbed and converted in the liver to indican, which, when oxidized, becomes indigo blue in the urine. Included in the differential diagnosis is the blue-green discoloration that can occur from a pigment elaborated by *Pseudomonas aeruginosa*.

Methionine malabsorption syndrome (oasthouse urine disease). This autosomal recessive condition of methionine absorption is often called oasthouse urine disease because affected patients excrete urine containing *a*-hydroxybutyric acid, a product of bacterial metabolism of methionine that has the odor of an oasthouse, a kiln for drying hops. The clinical features include white hair, hyperpnea, convulsions, diarrhea, and mental retardation. Individuals will have a positive ferric chloride test on analysis of their urine in addition to a-hydroxybutyric aciduria. A methionine-free diet improves the symptoms.

Disorders of Electrolyte Absorption Four genetic causes of intestinal electrolyte and water are well characterized: microvillus inclusion disease, congenital chloride diarrhea, congenital sodium diarrhea, and disorders of aldosterone metabolism causing significant salt wasting.

Congenital chloride diarrhea. Congenital chloride diarrhea is an autosomal recessive disorder that may present in utero with polyhydramnios, presumably caused by in utero diarrhea. The infant may be premature and exhibit abdominal distention. Voluminous, watery, high-chloride–containing stools are produced, leading to a hypocholuria, hypokalemia, and metabolic alkalosis. The secretory diarrhea in this condition results from a deficiency in intestinal brush border chloride-bicarbonate exchange. With later presentations, or inadequate replacement therapy, growth failure is noted. The high volume of diarrhea can be illustrated by the family of one young boy with the disorder who required donations of disposable diapers by a diaper manufacturer as this was the boy's largest non-reimbursable "medical" expense. Affected adults have a history of watery diarrhea since birth with numerous hospital admissions for dehydration and electrolyte abnormalities. Additional adult-onset features included glucose intolerance, gout, proteinuria, and mild renal function impairment. The chloride content of the stools and characteristic serum electrolyte changes are diagnostic. Treatment consists of administering prostaglandin inhibitors and potassium chloride. The gene for this condition, *DRA,* maps to chromosome 7q22-q31.

Congenital sodium diarrhea. Congenital sodium diarrhea is an autosomal recessive disorder that presents similarly to congenital chloride diarrhea with polyhydramnios, prematurity, and voluminous, watery, high sodium and bicarbonate-containing stools in the first days of life. Treatment consists of replacement of sodium bicarbonate. Perfusion studies show that the jejunum is in a net secretory state, with intact hexose transport but with abnormal intestinal sodium-hydrogen exchange activity.

Microvillus inclusion disease. Microvillus inclusion disease is an autosomal recessive disorder involving the brush border membrane which results in severe congenital fluid and electrolyte malabsorption and causes severe refractory diarrhea in early infancy. Unlike sodium and chloride diarrheas, polyhydramnios is not present prenatally. Microvillus inclusion disease likely results from either defective brush border assembly and differentiation or from an error of intracellular transport. Localization of the cytoplasmic inclusions to the microvillus membrane in villus cells is revealed by electron microscope examination. Rectal biopsy is used for early diagnosis. Therapies such as octreotide, clonidine, and epidermal growth factor have had limited success. It has been suggested that the only therapy to reduce fluid losses may be enterectomy. Survival is unusual after 2 years of age. Small bowel transplantation may become the best available treatment.

Intestinal Defects in Protein Absorption Many genetic syndromes demonstrate selective protein malabsorption as outlined in Table 127-2. Most involve

abnormalities of the intestinal lymphatic system with intestinal lymphangiectasia, protein-losing enteropathy, and vascular hyalinosis. A rare association of protein-losing enteropathy with neurofibromatosis has been described. This patient had massive protein loss from a localized segment of small intestine with possible neurofibromatosis involvement of the mesenteric vessels.

Intestinal Defects in Lipid Absorption Several important genetic disorders may present in adults with lipid malabsorption. Many are caused by inborn errors of apolipoprotein or lipid metabolism. Familial amyloidosis polyneuropathy is also an important disorder with onset in adulthood that may present with lipid malabsorption. The clinical features common to most of these syndromes are caused by the deficiency of fat-soluble vitamins. Neurological symptoms may be present because of a deficiency of vitamin E, and a coagulopathy may be caused by vitamin K deficiency.

Familial amyloidosis polyneuropathy. The amyloid protein in familial amyloidosis polyneuropathy type 1 is composed of abnormal transthyretin molecules. Transthyretin is a plasma protein that transports thyroid- and retinol-binding proteins. Systemic deposition of this abnormal protein causes a progressive stocking sensorimotor neuropathy that usually begins in the lower limbs. The autonomic nervous system may also be involved, causing orthostatic hypotension, impotence, and abnormal GI motility presenting as initial constipation followed by episodic diarrhea. Malabsorption of bile acids is common and appears to be the result of GI motility dysfunction from the autonomic neuropathy, with resultant bacterial overgrowth, rather than from the amyloid deposits in the intestinal mucosa. This is an autosomal dominant condition that can be presymptomatically diagnosed by looking for mutations in the transthyretin gene, *TTR,* on chromosome 18q11-12.

Abetalipoproteinemia. The principal GI symptom of abetalipoproteinemia, an autosomal recessive disorder, is malabsorption of fat, often beginning in infancy with steatorrhea and poor weight gain. The disorder is often misdiagnosed as celiac disease. Impaired absorption of fat-soluble vitamins is responsible for many of the clinical features. Histopathological analysis of the small bowel shows normally formed villi with lipid-engorged enterocytes. Treatment consists of avoidance of fat and the administration of fat-soluble vitamins. Neurological symptoms usually begin in the second decade and include peripheral neuropathy and cerebellar ataxia. Decreased night and color vision, caused by an atypical retinitis pigmentosa, may also develop as does mild to moderate anemia, usually with hemolysis where most circulating red cells are acanthocytes. There may be an iron or folate deficiency because of malabsorption and there usually is a coagulopathy caused by decreased vitamin K stores. The total cholesterol values are in the range of 20 to 50 mg/dL and triglyceride values are also very low, with little postprandial increase. There are no chylomicrons, very-low-density lipoproteins, low-density lipoproteins, or apolipoprotein B particles, the major structural apolipoprotein for these lipids. Heterozygotes are asymptomatic with normal plasma lipids. The genetic defect is not in the apolipoprotein B gene itself but instead in the microsomal triglyceride transfer protein gene, *MTP,* on chromosome 4q22-24 that mediates the intracellular transport of membrane-associated lipids. *MTP* mutations cause defects in the ability of the liver and intestines to assemble or secrete apolipoprotein B.

Anderson disease/lipid transport defect of the intestine. Individuals with Anderson disease, an autosomal recessive lipid transport defect of the intestine characterized by hypobetalipoproteinemia with accumulation of apolipoprotein B-like protein in intestinal cells, present with fat malabsorption with steatorrhea and growth retardation, but without the significant acanthocytosis and the neuroocular symptoms of abetalipoproteinemia. Low-density lipoprotein particles are present in the plasma, but there is a failure of chylomicron formation. Enterocytes from intestinal biopsies show numerous fat droplets, and monoclonal antibody staining reveals the presence of apolipoprotein B48, the form produced by the intestine. Defective glycosylation of apolipoprotein B48 has been observed, which may underlie the absence of formation and secretion of chylomicrons.

Familial hypobetalipoproteinemia. Familial hypobetalipoproteinemia is thought to be an autosomal dominant disorder caused by low levels of apolipoprotein-ß. Truncated versions of the normal apolipoprotein B gene, *APOB,* on chromosome 2p24 have been reported in some families where affected individuals have low levels of low-density lipoproteins, normal amounts of triglycerides, low levels of high-density lipoproteins, mild fat malabsorption, and a defect in chylomicron clearance. There were no reports of neuroocular abnormalities. In other families with similar clinical presentations linkage to *APOB* has been excluded. Linkage analysis suggests that another locus (*FHBL2*) may map to 3p22-p21, confirming the genetic heterogeneity of this condition.

Wolman disease and cholesterol ester storage disease/lysosomal acid lipase deficiency. Wolman disease is a severe autosomal recessive disorder that is often fatal in infancy. Cholesterol esters and triglycerides accumulate in the liver, adrenal gland, spleen, lymph nodes, bone marrow, small intestine, lungs, and thymus because of lysosomal acid lipase deficiency. Death is thought to be the result of intestinal malabsorption. Cholesterol ester storage disease is a late-onset disorder with milder manifestations. It also involves lysosomal acid lipase deficiency and is allelic with Wolman disease. The culprit lysosomal acid lipase gene (*LIPA*) maps to chromosome 10q24-25. Prenatal diagnosis has been performed by enzyme assay in cultured amniocytes. DNA-based testing may be available soon.

Exocrine Pancreas Abnormalities (See Also Chapter 98) Defects in protein and fat absorption are more commonly caused by abnormalities of the exocrine pancreas. Examples include deficiencies of specific pancreatic exocrine enzymes, cystic fibrosis, the Shwachman-Bodian syndrome, and the Johanson-Blizzard syndrome. Table 127-2 lists genetic syndromes manifesting with exocrine pancreas insufficiency.

Cystic fibrosis. Cystic fibrosis (CF) is the most common lethal autosomal recessive disorder in the Caucasian population, occurring with a frequency of about 1 in 2500 births. The disorder is characterized by viscous secretions and dysfunction of multiple exocrine glands, leading to pancreatic insufficiency, malabsorption, chronic pulmonary infection with emphysema, and a high chloride concentration in sweat. Milder mutations of the gene, *CFTR,* result in male infertility owing to congenital bilateral absence of the vas deferens or chronic pancreatitis. Only the GI manifestations of CF are reviewed here. Because of the often life-threatening nature of the pulmonary complications of classic CF, less emphasis is sometimes paid to the GI complications, but their importance is underscored by the fact that CF remains a leading cause of malabsorption in Caucasian children in developed countries. More than one third of patients with CF survive to adulthood and develop many of the GI complications with time. In fact, the majority of patients with classic CF have some degree of pancreatic insufficiency leading to achylia. Many are born with insufficiency and require enzyme replacement therapy by their first birthdays. Because the pancreatic secretions are abnormal, the pancreatic ducts become plugged, leading to destruction of distal ducts. Proximal to these clogged ducts, autodigestion and inflammation lead to fibrosis and ultimately to exocrine insufficiency. The result is malabsorption, primarily of fats and fat-soluble vitamins, which causes malnutrition, susceptibility to hemorrhage caused by lack of vitamin K, tetany caused by hypocalcemia from losses in the stool, and neurological problems caused by vitamin E deficiency. Treatment consists of pancreatic enzyme replacement. The islets of Langerhans may become secondarily affected by fibrosis of the pancreas, leading to glucose intolerance or clinical diabetes that is similar to non–insulin-dependent diabetes in approximately 50% of patients with CF. Approximately 25% to 30% of CF patients also develop focal biliary cirrhosis. In 2% to 5% of these cases, multilobular biliary cirrhosis with portal hypertension may develop. Thick secretions plug the bile ducts, leading to inflammation and fibrosis. There usually is a history of pancreatic insufficiency in these cases. The earliest clinical manifestation of CF may be meconium ileus, occurring in approximately 10% to 15% of patients, caused by the lack of fluid in the small intestine because of a lack of pancreatic secretion or defective intestinal epithelial secretion. Gastrografin enema or oral *N*-acetylcysteine may resolve the obstruction; if not, surgery is necessary. A similar abnormality occurring later in life, called the distal intestinal obstruction syndrome, may occur in 10% to 20% of CF patients, causing symptoms of colicky pain and constipation. These symptoms may be relieved with Golytely, mineral oil, an increase in pancreatic enzymes, or *N*-acetylcysteine. Rectal prolapse occurs in approximately one fourth of patients before 2 years of age, but it usually does not occur in patients receiving pancreatic enzyme replacement therapy. The mechanism for the prolapse is unknown. The responsible gene, *CFTR,* was mapped to chromosome 7q31 by positional cloning strategies. The putative protein product for the gene, called the cystic fibrosis transmembrane regulator protein, forms a cAMP-regulated chloride channel. More than 500 *CFTR* mutations have been described, which account for over 90% of all recognised mutant chromosomes detected by linkage analysis. The sensitivity of mutation testing varies depending on an individual's ethnicity, however, with up to 96% of mutant chromosomes detectable in affected Ashkenazi Jews and less than 70% of mutant chromosomes detected in African Americans with most commercially available mutation testing products. The most common mutation in Caucasians of Northern European descent is dF508, a deletion of the amino acid residue phenylalanine at codon 508, occurring in 70% of CF patients. The dF508 deletion is located near an apparent binding site for ATP, and it may interfere with the phosphorylation of the CF protein or with some other need for energy provided by ATP. Most of the non-dF508 mutations are relatively rare, affecting less than 1% to 10% of CF patients. Some *CFTR* genotypes are associated with pancreatic sufficiency or insufficiency, suggesting that the genotype at the *CFTR* locus is responsible for susceptibility to pancreatic insufficiency. *d*F508 and the common mutation among Ashkenazi Jews, *W1282X,* are examples of pancreatic insufficiency alleles. Preconceptual and prenatal testing are readily available. As of 2001 the American Colleges of Medical Genetics and Obstetrics and Gynecology recommend that all Caucasian couples considering children and pregnant women be routinely offered DNA carrier testing.

Shwachman-Diamond/Shwachman-Bodian syndrome. The Shwachman-Diamond/Shwachman-Bodian syndrome, also called congenital lipomatosis of the pancreas, is an autosomal recessive disorder and the second most frequent pancreatic cause of malabsorption in childhood. It has an incidence of 1 in 250,000 births. Symptoms include poor growth, intestinal malabsorption, steatorrhea, and recurrent respiratory infections, which present within the first few years of life. Dysplastic changes in the metaphyses of long bones lead to a moderate dwarfism. Patients may suffer from a pancytopenia caused by bone marrow dysfunction and are at increased risk for hematologic malignancies. The pancreatic enzyme replacement required in infancy may not be necessary later. The locus for this condition maps to the centromeric region of chromosome 7.

Johanson-Blizzard syndrome. In addition to pancreatic insufficiency with malabsorption, Johanson-Blizzard syndrome, an autosomal recessive syndrome, is characterized by multiple anomalies including aplasia or hypoplasia of the nasal alae, upswept frontal hair, widely spaced teeth, congenital deafness, hypothyroidism, postnatal growth retardation, mental retardation, midline ectodermal scalp defects, and absent permanent teeth. Genitourinary and anorectal abnormalities have also been described. Autopsy findings have shown almost complete replacement of the pancreas with fat and a paucity of islet cells. Death usually occurs in childhood from malabsorption-related complications. Survival to adulthood is rare but possible if continuous enzyme replacement is instituted. Insulin-requiring diabetes is a feature in adulthood, similar to the situation for CF patients, presumably because of the loss of ß-cells.

Cartilage-hair hypoplasia/metaphyseal chondrodysplasia, McKusick type. Cartilage-hair hypoplasia (i.e., metaphyseal chondrodysplasia, McKusick type) is an incompletely penetrant autosomal recessive dwarfing syndrome (average adult height is 107–157 cm) with radiologic changes called metaphyseal dysostosis. Other features include fine, sparse, light-colored hair, anemia, malabsorption caused by pancreatic insufficiency, Hirschsprung disease, and a susceptibility to chicken pox. Malignancies, specifically lymphoma, are more frequent in affected individuals. The *CHH* gene maps to chromosome 9p13.

Asphyxiating thoracic dystrophy/Jeune syndrome. Asphyxiating thoracic dystrophy, also called Jeune syndrome, is an autosomal recessive skeletal dysplasia that is often lethal in the perinatal period caused by involvement of the rib cage leading to severe asphyxia. Mildly affected individuals survive to adulthood. Other features include polydactyly, chronic nephritis, and intestinal malabsorption because of pancreatic insufficiency. Cystic changes occur in the kidney, liver, and

pancreas. This syndrome shares clinical features seen in two other genetic syndromes, Ellis-van Creveld and renal-hepatic-pancreatic dysplasia syndromes.

Disorders of Vitamin and Mineral Assimilation This heterogeneous group of disorders displays widely variable manifestations dependent on the specific vitamin or mineral involved. Some disorders of vitamin and mineral metabolism can be appropriately treated by adequate replacement therapy.

Imerslünd-Grasbeck syndrome (see also Chapter 77). Imerslünd-Grasbeck syndrome is an autosomal recessive disorder of chronic relapsing megaloblastic anemia caused by malabsorption of vitamin B_{12}, which is not corrected with exogenous intrinsic factor. No antibodies to intrinsic factor or gastric parietal cells are detected. The defect appears to be located between the attachment of B_{12} to the surface of the ileal cell and the binding to transcobalamin II. In some cases, the defect is a selective failure of intrinsic factor transcytosis; in others, it may be the absence of an ileal receptor; and in others, the defect is unknown. Associated features include proteinuria and malformations of the urinary tract and persistent proteinuria. The gene for this disorder, *MGA1,* maps to 10p12.2. This syndrome should be differentiated from intrinsic factor deficiency and transcobalamin II deficiency. The disease does not respond to administration of intrinsic factor. Treatment consists of monthly intramuscular doses of 250 µg of hydroxycobalamin or intramuscular doses every 2 to 3 months of 1000 µg of hydroxycobalamin. The proteinuria does not usually respond. Because mental retardation may ensue from delay or lack of treatment, the siblings of an affected person should be screened.

Menkes syndrome (see also Chapter 77). Menkes syndrome, also called kinky hair disease, is an X-linked recessive disorder of copper metabolism caused by a defect in the intestinal absorption of copper owing to mutations in the *ATP7A* gene on chromosome Xq12-13. This affects copper-dependent enzymes in the body, producing a phenotype characterized by light-colored and kinky hair, growth retardation, severe neurological impairment with focal cerebral and cerebellar degeneration, changes in the metaphyses of the long bones, tortuosity of vessels, and bladder diverticula. Most patients die between 6 months and 3 years of age but mildly affected patients with presumably allelic forms of the disease have survived to adolescence. Low serum copper levels in the appropriate clinical setting is a simple and reliable diagnostic method. Heterozygotes may be detected by the finding of pili torti (i.e., kinky hair).

X-linked cutis laxa/occipital horn syndrome. X-linked cutis laxa/occipital horn syndrome mutations are caused by mutations in *ATP7A,* on chromosome Xq12-13, making the syndrome allelic with Menkes syndrome. The syndrome is characterized by occipital bony prominences (occipital horns), short broad clavicles, fused carpal bones, pectus deformities, a hooked nose with a long philtrum, multiple large diverticula of the bladder, varicosities, inguinal hernias, joint laxity, and diarrhea. Serum levels of copper and ceruloplasmin are low, similar to Menkes syndrome.

Acrodermatitis enteropathica (see also Chapter 76). Acrodermatitis enteropathica is an autosomal recessive disorder of zinc malabsorption and presents with intermittent diarrhea, dermatitis, and failure to thrive. Additional features include alopecia of the scalp, eyebrows, and eyelashes. At autopsy islet cell hyperplasia, absence of the thymus, and absence of germinal centers with plasmacytosis of the lymph nodes and spleen are noted. Laboratory findings include low serum and urinary levels of zinc and serum alkaline phosphatase (i.e., a zinc metalloenzyme). Oral zinc therapy has been successful in ameliorating symptoms. Low serum zinc levels are helpful, but not 100% sensitive, in making the diagnosis. Ultrastructural abnormalities of the Paneth cells of the small intestine (i.e., cells that usually contain high concentrations of zinc) or a trial of oral zinc supplementation with good clinical response may provide evidence for the diagnosis in cases with normal serum zinc levels. The locus has been mapped to chromosome 8q24.

Hemochromatosis (see also Chapter 112). Hemochromatosis is one of the most common autosomal recessive disorders with a carrier frequency of 1 in 10 individuals and affecting between 1 in 200 to 400 individuals of Northern European descent. An entire chapter, Chapter 112, is devoted to this condition. In homozygotes, increased absorption of iron in the small intestine results in an excess of iron deposited in many organs, ultimately resulting in organ failure in some individuals. Serious manifestations of hemochromatosis include cirrhosis, diabetes, cardiomyopathy, arthritis, hepatomas, and hypogonadotropic hypogonadism. Symptoms vary considerably among individuals. These may be minimal in individuals with chronic or episodic blood loss (e.g., menstruating women) and more severe in men with a high content of iron in their diet. Generally, symptoms begin in adulthood, after several years of increase in iron absorption. The gene, *HFE,* maps to the human leukocyte antigen (HLA) region of chromosome 6p21, and has been cloned. One predominant mutation, Cys282Tyr, accounts for between 80% to 90% of mutant chromosomes, making DNA testing relatively straightforward. Homozygotes diagnosed early or when asymptomatic can be closely monitored and managed to prevent symptoms of iron overload. In addition to DNA diagnosis, liver biopsy along with serum iron, total iron-binding capacity, and ferritin levels may be helpful in the diagnosis. Treatment continues to include phlebotomy for individuals with increased iron levels. Other forms of hemochromatosis, caused by other factors, likely exist.

Chronic Diarrhea Syndromes Not Characterized by Malabsorption (See Also Chapter 42)

Several genetic disorders featuring chronic, recurrent diarrhea are not associated with a specific malabsorption syndrome. Many of the inherited immunodeficiency disorders involving T-cell antigens, T-cell antigen receptors, B cells, and immunoglobulin and complement deficiencies are characterized by a chronic diarrhea. Inherited colitis syndromes, neurological disorders, and endocrinopathies may also feature chronic or recurrent diarrhea as a common manifestation. Many of the polyposis syndromes may present with complaints of chronic diarrhea. Table 127-3 briefly summarizes those disorders that may be more commonly encountered by the clinician.

TABLE 127-3 Diarrhea Syndromes

Recurrent Gastrointestinal Bleeding Syndromes (See Also Chapter 130)

Recurrent acute or occult GI hemorrhage should raise suspicion for a variety of genetic disorders (Table 127-4). These include structural abnormalities, such as the polyposis syndromes (e.g., Peutz-Jeghers syndrome, Gardner syndrome); vascular abnormalities, such as those that occur in vascular malformation syndromes and connective tissue disorders (e.g., hereditary hemorrhagic telangiectasia syndrome, Ehlers-Danlos syndrome); and disorders of the hematologic system that may involve platelets, clotting factors, or abnormalities in fibrinolysis. The polyposis syndromes are reviewed elsewhere (see Chapter 90).

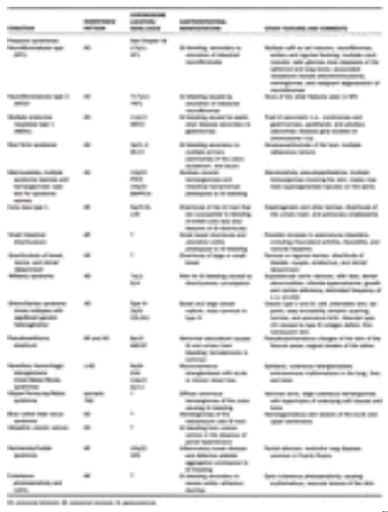

TABLE 127-4 Recurrent Gastrointestinal Bleeding Syndromes

Many of the structural and vascular abnormality syndromes that cause GI bleeding belong to a group of disorders called the phakomatoses or genodermatoses, including neurofibromatosis type I, basal cell nevus syndrome, Gardner syndrome, hereditary hemorrhagic telangiectasia, Klippel-Trenaunay-Weber syndrome, and Peutz-Jeghers syndrome. *Phakos* means spots, and these disorders have characteristic spotty or patchy cutaneous lesions. Other common features include a major vascular component to some lesions, involvement of the skin and central nervous system, and autosomal dominant inheritance patterns, often with significant variable expressivity and a high new mutation rate.

Many genetic hematologic disorders predispose to GI bleeding. Deficiencies of clotting factors may result in a bleeding diathesis. von Willebrand disease and combined deficiency of factor VII and VIII are autosomal dominant disorders that may present with recurrent GI bleeding. The most common factor deficiency is factor VIII deficiency, better known as hemophilia A, an X-linked condition occurring with a frequency of 1 in 10,000. This disorder usually presents with hemarthroses that result in a chronic arthropathy, deep muscle hematomas, frank hematuria, and spontaneous intracranial bleeds. GI bleeding is not a common complication.

Abnormalities in fibrinolysis may also predispose to GI bleeding. Deficiencies of the anticlotting factors, protein C, protein S, and antithrombin III, can cause portal vein thrombosis, which can be complicated by ruptured esophageal varices.

Several genetic disorders are associated with abnormal platelet number or function predisposing to increased GI bleeding. Glanzmann thrombasthenia (i.e., platelet glycoprotein IIb-IIIa deficiency) is an autosomal recessive bleeding diathesis with deficient clot retraction and abnormal platelet morphology. Mucosal bleeding is typical, and gingival bleeding, epistaxis, and laryngeal bleeding are common features. An autosomal dominant form of Glanzmann thrombasthenia has similar features. Bernard-Soulier syndrome (i.e., platelet glycoprotein Ib deficiency) is an autosomal recessive bleeding disorder characterized by unusually large platelets and a prolonged bleeding time. The platelet membrane is abnormal and lacks a receptor, glycoprotein Ib, involved in the adhesion of platelets to subendothelium and von Willebrand factor; patients are susceptible to mucosal bleeding. Thromboxane synthase deficiency, leading to platelet aggregation abnormalities, is an autosomal recessive disorder that may present with petechiae, bruising, epistaxis, hematuria, or massive GI bleeding. The Hermansky-Pudlak syndrome is also characterized by abnormal platelets and is discussed in a later section of this chapter.

Neurofibromatosis (See Also Chapter 73) There are two major types of neurofibromatosis (NF): NF type 1 (NF1) and NF type 2 (NF2). NF2 is not typically associated with significant GI manifestations. NF1 and a probable allelic variant of NF1, previously called NF3, known as familial intestinal neurofibromatosis, can be associated with GI hemorrhage because of ulceration of neurofibromas in the lumen of the bowel. Major characteristics of NF1 include multiple cutaneous café au lait macules, multiple neurofibromas or plexiform neurofibromas, axillary or inguinal freckling, optic gliomas, multiple Lisch nodules (i.e., iris hamartomas), and bony dysplasia of the sphenoid or long bones. Occasionally, an individual with NF1 may present with obstructive GI symptoms secondary to having large internal plexiform neurofibromas extending throughout their thoracoabdominal cavity. Arterial occlusive disease also has been reported in NF and may be the underlying etiology of GI complaints in affected individuals. NF1 is an autosomal dominant disorder displaying significant intrafamilial and interfamilial variability and occurs in 1 per 4000 persons of all ethnic groups. New mutations are thought to account for up to 50% of affected individuals. The large NF1 gene maps to chromosome 17q12. DNA-based diagnostic testing relies on a protein truncation assay, which detects approximately 70% of mutations, or on linkage analysis when multiple family members are available to set the appropriate phase. The National Neurofibromatosis Foundation (and its local chapters) provides educational materials, support, and newsletters to families and physicians.

Hereditary Hemorrhagic Telangiectasia (HHT)/Osler-Weber-Rendu Syndrome Hereditary hemorrhagic telangiectasia (HHT) is a genetically heterogeneous autosomal dominant disorder of high penetrance characterized by mucocutaneous telangiectases. Two major genetic forms are recognized. The usual presenting feature is epistaxis occurring at puberty. The cutaneous telangiectases appear later, usually between 5 and 20 years of age. Blood loss from the mucosal lesions in the gut can be chronic or acute. Larger vessel involvement is common. Pulmonary arteriovenous malformations occur in approximately 15% to 20% of individuals, especially in those families with HHT1 whose disease is caused by mutations in the endoglin gene, *ENG,* on chromosome 9q34, and may lead to embolic complications or high-output cardiac failure. HHT2 maps to 12q13 where mutations have been identified in *ALK-1*. Arteriovenous malformations occur less often, in the cerebral and hepatic vessels.

Klippel-Trenaunay-Weber Syndrome The Klippel-Trenaunay-Weber syndrome is a sporadic, but presumably autosomal dominant, disorder that features varicose veins and large, cutaneous hemangiomas with hypertrophy of the underlying bones and soft tissues. Diffuse cavernous hemangiomas of the colon are found in 1% to 12.5% of patients and can sometimes cause significant GI bleeding. Partial colectomy and endoscopic laser ablation of residual abnormal colonic mucosa has been successful in the management of this complication.

Blue Rubber Bleb Nevus Syndrome The blue rubber bleb nevus syndrome is an autosomal dominant disorder characterized by the presence of easily compressible bladderlike hemangiomatosis skin lesions that refill quickly after compression and have the look and feel of rubber nipples. The sites most commonly involved are the trunk and upper extremities, but the mucosa of the nasopharynx and GI tract, particularly the small bowel, can be affected. Bleeding is an important complication of these lesions. Nocturnal pain and regional hyperhidrosis may also be features of this disorder.

Ehlers-Danlos Syndrome Connective tissue disorders in which the integrity of structural tissues is impaired may predispose to GI bleeding. Ehlers-Danlos syndromes (EDS) are a heterogenous group of connective tissue disorders affecting the skin, ligaments, joints, blood vessels, and internal organs, depending on the structural protein or modifying enzyme affected. There are six major subtypes of EDS that have been described, most of which are autosomal dominant, although the dermatospraxis and kyphoscoliosis forms are autosomal recessive. The nomenclature for EDS subtypes has been revised to define the “classic” forms, EDS I and EDS II (where EDS II is a milder form) as characterized by soft, velvety, hyperextensible skin, lax joints, easily bruised skin, atrophic scarring, venous varicosities, hernias, and prematurity caused by rupture of fetal membranes. GI bleeding caused by hollow organ (i.e., bowel) and arterial rupture is not a usual manifestation but may occur. Some cases of EDS I and II (a milder variant similar to EDS I) have demonstrated to alterations in the type V collagen gene, *COL5AI,* which maps to chromosome 9q. The “vascular” form of EDS, EDS IV, is associated with increased morbidity and early mortality caused by bowel and large vessel ruptures. Individuals most often have very thin, fragile skin with easily visible veins and friable tissues. They bruise easily, have tight skin over the fingers, face, and ears (acrogeria), and hyperextensible distal interphalangeal joints. Defects in the type III collagen gene, *COL3A1,* which maps to chromosome 2q31, are responsible for the phenotype. Unfortunately, no specific treatment is available for EDS IV and it is not uncommon for individuals to die during late adolescence or early adulthood owing to rupture of the bowel, large arterial vessels, or uterus during pregnancy. Cases have been reported where the ruptured bowel could not be appropriately repaired at surgery owing to the extensive friability of the tissue that was unable to adequately hold sutures. For most disorders in the EDS family of syndromes, analysis of skin fibroblasts for collagen synthesis and secretion are the diagnostic methods of choice. Once a specific DNA mutation is identified in a family, sensitive DNA-based mutation testing can easily be done. Unfortunately, even with early diagnosis of type III collagen abnormalities, no specific therapy is available to prevent rupture of hollow organs and vessels. However, appropriate genetic counseling about risks and strategies to minimize catastrophic complications, especially during pregnancy, can be offered to individuals.

Pseudoxanthoma Elasticum Pseudoxanthoma elasticum primarily affects the skin of the neck, axillae, and other flexural areas; the Bruch membrane of the eye, resulting in angioid streaks on funduscopic examination; and the arteries, producing GI and urinary tract hemorrhage, precocious calcification, and occlusive vascular changes. Recurrent, severe hematemesis was the presenting feature of one of the first reported cases. Hematemesis is much more common than melena; superficial hemorrhages and erosions may be seen in the stomach. Bleeding tends to occur as early as the first decade of life, before the skin and eye changes are evident. Pregnant women are more susceptible to bleeding. The diagnosis is made on the basis of clinical examination but there is wide variable expressivity. Allelic autosomal dominant and autosomal recessive forms have been reported. The dominant types are characterized by *peau d’orange* skin changes, severe vascular complications, severe choroiditis, angioid streaks, retinal detachment, macular or focal changes in the skin which may be very stretchy, myopia, a high-arched palate, blue sclerae, and loose joints. The recessive types may be clinically similar. Patients have been reported with generalized skin changes with no ocular or vascular abnormalities. Mutations in the multidrug resistance gene, *ABCC6* (ATP-binding cassette, sub-family C, member 6), on chromosome 6p13 have been identified in

individuals with both autosomal dominant and autosomal recessive forms of the disease.

Syndromes Characterized by Hamartomas, Macrocephaly, Multiple Lipomas, and/or Hemangiomas (See Also Chapter 90) A collection of several autosomal dominant syndromes, including Bannayan-Zonana syndrome, Bannayan-Riley-Ruvalcaba syndrome, Cowden syndrome, Riley-Smith syndrome, and Ruvalcaba-Myhre-Smith syndrome, present with similar features but exhibit genetic variability and, likely, both allelic and locus heterogeneity. Typical features include macrocephaly, pseudopapilledema, multiple lipomas, multiple hemangiomas involving the skin and viscera, thyroid nodules, and intestinal hamartomatous polyposis. The latter two features create a susceptibility to GI hemorrhage. Men may have hyperpigmented macules on the penis. High birth weight and length is typical, but growth levels off at 6 to 7 years of age. There may be delayed motor development, autistic features, cognitive delays, and lifelong problems with coordination. Seizures have been reported from intracranial hemorrhage caused by arteriovenous malformations. Arteriovenous malformations may also lead to extremity overgrowth, necessitating amputation. In adults a higher incidence of breast, thyroid, colon, renal, endometrial, and skin cancer have been reported, making this a cancer predisposition syndrome for which preventive screening and management should be employed. Mutations in the *PTEN* gene, a tumor suppressor gene first identified in association with Cowden syndrome, which maps to chromosome 10q23, have also been demonstrated in individuals with Bannayan-Riley-Ruvalcaba, Bannayan-Zonana, Riley-Smith, and Ruvalcaba-Myhre-Smith syndromes, demonstrating that these are indeed, allelic conditions. Mutations in the *BMPR1A/ALK3* gene, a member of the transforming growth factor-ß/ *BMP* superfamily, on 10q22, have been identified in some families with these conditions. They have also been noted in some kindreds with the inherited juvenile polyposis syndrome, a similar but distinct hamartoma syndrome with an increased predisposition to colon cancer. Mutations in the *MAD4/DPC4* gene located on 18q21 have also been associated with familial juvenile polyposis and some reports have suggested that juvenile polyposis can be caused by mutations in the *PTEN* gene. Atypical juvenile polyps with mixed features of hamartomas and adenomas are characterized by a variant of the juvenile polyposis syndrome known as the hereditary mixed polyposis syndrome. This form maps to chromosome 6q16. Further molecular characterization of the hamartomatous syndromes is needed to more precisely clarify the relationships between these conditions. It has been suggested that if *PTEN* mutations are identified in a patient that the most useful diagnosis may be the " *PTEN* hamartoma syndrome" to reduce the confusion associated with the multiple syndromes that have been associated with *PTEN* alterations.

Cutis Laxa Several forms of cutis laxa have been described and the relationship between these forms at a molecular level is not yet understood. Cutis laxa type I is an autosomal recessive disorder characterized by abnormalities of the elastic tissue and features diaphragmatic and other hernias; diverticula of the GI and urinary tract, which are susceptible to bleeding; and pulmonary emphysema, which may lead to cor pulmonale. Death usually occurs in the first year of life. Cutis laxa I has been mapped to the long arm of chromosome 5. The second less severe form is characterized by prenatal and postnatal growth deficiency, large fontanels with delayed closure, congenital hip dislocation, and lax joints. Diminished elastin production has been verified in these patients, and quantitation reveals decreased elastin mRNA, perhaps caused by decreased rates of transcription or degradation of unstable transcripts. A dominant form of cutis laxa does not have the visceral manifestations characteristic of the recessive forms. X-linked cutis laxa is discussed in "Disorders of Vitamin and Mineral Assimilation" as it is associated with copper abnormalities. The syndrome also features diverticular disease of the GI tract, increasing the possible risk for GI hemorrhage.

Hermansky-Pudlak Syndrome Hermansky-Pudlak syndrome is an autosomal recessive disease that occurs most frequently in Puerto Rico, where the prevalence is 1 in 2000 individuals. It is a storage disorder of a ceroidlike substance and is associated with features of partial albinism, restrictive lung disease, and a platelet abnormality. There is a deficiency of the granule storage pool in platelets, causing defective ADP release, which leads to a prolonged bleeding time and defective platelet aggregation. Inflammatory bowel disease with an onset between the ages of 12 and 30 years is also common in the Puerto Rican patients. The combination of inflammatory bowel disease and defective platelets contributes to the occurrence of GI bleeding in these patients. The gene for this disorder, *HPS,* maps to chromosome 10q23.

Intestinal Motility or Pseudoobstruction Disorders (See Also Chapter 41, Chapter 73)

Intestinal pseudoobstruction may result from intrinsic enteric myopathies and neuropathies or from generalized muscular and neurological disorders. This section reviews the genetic disorders that feature abnormal intestinal motility or pseudoobstruction in each of these categories. These disorders are summarized in Table 127-5.

TABLE 127-5 Intestinal Motility and Pseudoobstruction Disorders

Intestinal pseudoobstruction is suggested clinically by recurrent signs and symptoms of intestinal obstruction in the absence of mechanical obstruction, as proven by appropriate radiologic, endoscopic, or surgical investigations. The presence and severity of complications depend on the individual's age (i.e., infant or adult), site of involvement, and degree of functional impairment. In the adult, a long-standing history of symptoms suggesting GI motility dysfunction—such as dysphagia, early satiety, and constipation or diarrhea—may precede the onset of acute symptoms. Patients usually present in the second to fourth decades of life with complaints of abdominal pain, distention, and sometimes vomiting. Malnutrition is common and results from decreased food intake, impaired digestion, or malabsorption. Infants with these disorders usually present with acute, severe, obstructive symptoms of abdominal pain and distention after feeding. Reflux esophagitis and aspiration pneumonia caused by impaired gastroesophageal motility is also common in infants.

After mechanical obstruction has been ruled out, diagnostic studies to evaluate pseudoobstruction include esophageal and intestinal manometry, gastric emptying studies, and full-thickness biopsy with silver stains. Esophageal manometry shows aperistalsis and incomplete relaxation of the lower esophageal sphincter. Intestinal manometry usually shows a hypomotile pattern. Gastric emptying studies show a delay for solids and a variable pattern for liquids. Silver stains delineate neuropathic from myopathic forms. Detection of subclinically affected family members at risk can be accomplished with these diagnostic techniques.

Visceral Myopathy Syndromes Clinical GI manifestations of the visceral myopathic and neuropathic forms of intestinal pseudoobstruction are similar. In the myopathic form, there may be other evidence of myopathy, such as megacystis, vesicoureteral reflux, ophthalmoplegia, ptosis, and small intestine diverticulosis. Pathogenetic mechanisms of GI manifestations include degenerative changes and thinning of intestinal smooth muscle, especially of the longitudinal layers; the enteric neurons are normal. A mitochondrial myopathy associated with intestinal pseudoobstruction caused by a visceral myopathy has been described. Some forms of visceral myopathy may be mitochondrial myopathies. Diagnosis of a mitochondrial myopathy can often be made by a Gomori trichome–stained skeletal muscle biopsy, which shows the characteristic ragged, red fibers, or by analysis of respiratory chain enzymes. There are no well-documented reports of a mitochondrial DNA mutation responsible for intestinal pseudoobstruction. The reported familial cases are autosomal dominant or recessive, lacking the maternal pattern of transmission seen with mitochondrial DNA mutations. If a mitochondrial myopathy is involved, it is likely that the disease gene responsible in most cases resides in the nucleus.

Duchenne muscular dystrophy. Duchenne muscular dystrophy is an X-linked disorder characterized by skeletal muscle degeneration causing atrophy and weakness. The incidence in male births is about 1 in 3500. Onset of symptoms usually begins before 5 years of age, with progressive symptoms of muscle weakness, toe walking, and difficulty in rising from sitting and in climbing stairs. Affected boys have a waddling gait, and pseudohypertrophy of the calves is typical. By puberty, most require use of a wheelchair; joint contractures and scoliosis then develop. Death usually occurs in the late teens or early 20s from respiratory infections or heart failure. Laboratory studies show elevated serum levels of aldolase, lactate dehydrogenase, and creatine kinase. Common GI symptoms include abdominal pain and distention, delayed gastric emptying, acute gastric dilation, and chronic intestinal pseudoobstruction. The large disease gene, encompassing over 2 Mb of DNA, responsible for Duchenne muscular dystrophy maps to Xp21 and encodes a protein found in skeletal muscle called dystrophin, which helps stabilize the cytoskeleton of skeletal muscle. This gene is lacking in affected boys and partially deficient in female carriers. DNA diagnosis is readily available. The majority of mutations are large deletions that can be easily detected by polymerase chain reaction-based DNA diagnostic methods. Prenatal DNA diagnosis, followed by pregnancy termination if the fetus is affected, is therefore an option for families wishing to prevent the birth of an affected child. Obviously, improved treatments are desperately needed and many hope that efforts in gene therapy in mouse models will lead to human trials in the near future.

Myotonic dystrophy. Myotonic dystrophy is an autosomal dominant disorder characterized by delayed muscular relaxation, facial weakness and muscle atrophy, ptosis, cataracts, gonadal atrophy, cardiac conduction abnormalities, and male pattern baldness. Significant GI complications are caused by abnormal functioning at all levels of the GI tract, from the pharynx to the anal sphincters; this results from degenerative changes in smooth muscle and fatty infiltration, causing problems with swallowing, aspiration, and constipation. The more severe phenotype of congenital myotonic dystrophy only occurs with alleles of maternal origin. The congenital form, often inherited from a mildly affected mother, features mental retardation, hypotonia, facial diplegia, gastroparesis, subacute intestinal obstruction, megacolon, and constipation. The disease gene maps to 19q13 and involves a CTG trinucleotide repeat in the three untranslated regions of the gene. Unaffected individuals have

5 to 27 copies of the repeat, whereas affected individuals have minimally 50 or more repeats with severity increasing as the repeat copy number increases to several kilobases. Expansion of the repeat can occur in subsequent generations, providing a molecular basis for the clinical phenomenon of anticipation (i.e., earlier clinical onset of disease in subsequent generations) in this disorder. DNA testing is sensitive, specific, and relatively easy to perform, thereby providing a specific way to confirm a suspected clinical diagnosis noninvasively. Other disorders presenting with myotonia caused by different genetic alterations can present with GI symptoms similar to those previously described.

Kearns-Sayre syndrome. Kearns-Sayre syndrome is a mitochondrial disorder characterized by progressive external ophthalmoplegia, pigmentary retinopathy, cardiomyopathy, and cardiac conduction defects; weakness of facial, trunk, and extremity muscles; and deafness, small stature, electroencephalographic changes, and increased cerebrospinal fluid protein. GI symptoms relate to the pharyngeal muscle weakness experienced by affected individuals, causing swallowing difficulties and dysphagia. The mitochondrial mutations characteristic of this disorder are deletions ranging in size from 2 to 7 kb and can be easily detected by analysis of mitochondrial DNA.

Visceral neuropathy syndromes. The visceral neuropathic forms of intestinal pseudoobstruction are characterized by abnormal pupillary reflexes, ataxia, and dysarthria; peripheral nervous system abnormalities, including absent deep tendon reflexes and impaired vibratory and position sense; and autonomic neurological abnormalities such as inappropriate blood pressure responses to phenylephrine, Valsalva maneuver, upright posture, and lack of sweating on warming. One type is also associated with basal ganglia calcification and mental retardation. Histological sections of tissues stained with a silver stain reveal degenerative changes of myenteric plexus neurons.

Hirschsprung disease (see also Chapter 82). Hirschsprung disease is probably a multifactorial disorder that usually occurs as an isolated congenital anomaly but may be a syndromic association of several genetic conditions (see Table 127-4). With isolated Hirschsprung disease, the risk to family members increases with severity (i.e., length of GI tract involved) and the number of affected relatives, especially female relatives. Empiric risk estimates are 7.2% for the siblings of affected women and 2.6% for the siblings of affected men. Complex segregation analysis demonstrated Mendelian inheritance patterns when the length of affected bowel was considered. Cases with aganglionosis beyond the sigmoid colon had a mode of inheritance that was compatible with an autosomal dominant trait with incomplete penetrance. Cases with aganglionosis no more proximal than the sigmoid colon had inheritance patterns consistent with multifactorial inheritance or autosomal recessive inheritance with very low penetrance. Isolated, nonsyndromic familial forms of the disease have demonstrated mutations in the *RET* protooncogene, which maps to chromosome 10q11.

Familial achalasia and diffuse esophageal spasm. Familial occurrence of achalasia, an autosomal recessive disorder, involves the esophagus, causing symptoms of progressive dysphagia, retrosternal discomfort, and intermittent regurgitation. The cause of achalasia is unknown. Diverticulosis of the esophagus may also occur. The severity of the disorder exhibits intrafamilial variability. Esophageal ganglion cells and dorsal motoneurons of the vagal nucleus are decreased, and there is the wallerian degeneration of the vagus nerve.

Familial dysautonomia. Familial dysautonomia, also called Riley-Day syndrome, is an autosomal recessive disorder, involving the autonomic nervous system, that is most prevalent in the Ashkenazi Jewish population, with an incidence of about 1 in 3500 individuals. It is characterized by autonomic instability, including lack of tearing, emotional lability, paroxysmal hypertension, increased sweating, cold hands and feet, corneal anesthesia, red blotching of the skin, and drooling. GI manifestations include oropharyngeal incoordination, abnormal esophageal motility and decreased lower esophageal sphincter pressure, prolonged gastric emptying, and gastroesophageal reflux, all of which result in dysphagia, emesis, and aspiration pneumonia. Affected individuals are reluctant to eat and suffer from dehydration and failure to thrive. Fundoplication and gastrostomy has some documented benefit in these cases. Diagnosis is determined by a lack of axon flare after intradermal injection of histamine, absence of fungiform papillae on the tongue, miosis of the pupil after instillation of methacholine chloride (2.5%), absent deep tendon reflexes, and diminished tear flow. The disease gene, *IKBKAP,* maps to chromosome 9q31 and linkage disequilibrium is present within Ashkenazi Jews, with a high-risk haplotype recognized.

Mechanical Obstruction and Malformation Syndromes

Most genetic disorders that cause mechanical obstruction are related to congenital malformations, and in general they have more importance in the pediatric population than they do for adults. However, as noted earlier in this chapter, some disorders with potential mass effects, such as large abdominal plexiform neurofibromas in NF1, may cause mechanical obstructive symptoms in rare instances. Another more common genetic cause for mechanical obstruction not caused by malformation is meconium ileus seen in 10% to 15% of cystic fibrosis patients. The anatomic site or sites of developmental malformation in these syndromes is usually restricted to derivatives of the embryologic foregut, midgut, and hindgut. In a rostral to caudal distribution, Table 127-6 reviews genetic malformation syndromes that may present with symptoms secondary to a mechanical obstruction.

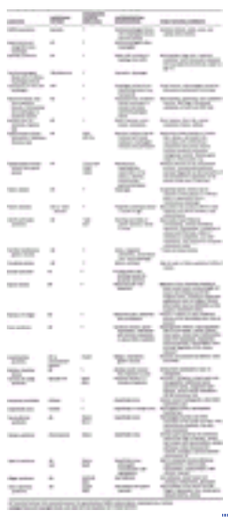

TABLE 127-6 Mechanical Obstruction and Malformation Syndromes

Chronic Abdominal Pain Syndromes (See Also Chapter 128)

Many genetic syndromes can present with recurrent bouts of abdominal pain, including familial Mediterranean fever (FMF), Fabry disease, hereditary angioneurotic edema, hereditary fructose intolerance, and the hereditary pancreatitis syndromes (Table 127-7). Although these syndromes (except for FMF) are relatively rare; making the correct diagnosis in a symptomatic individual may prevent unnecessary and invasive procedures and may similarly benefit other family members. The porphyrias are also associated with significant recurrent abdominal pain as discussed in Chapter 49.

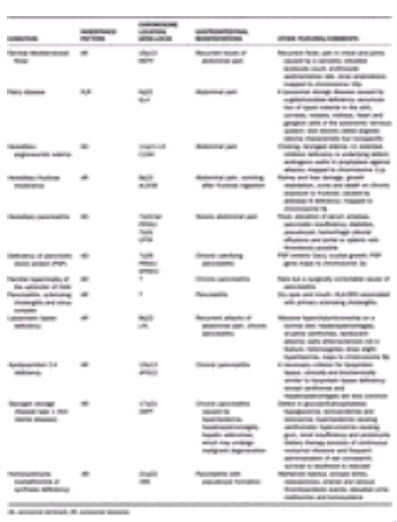

TABLE 127-7 Chronic or Recurrent Abdominal Pain Syndromes

Familial Mediterranean Fever FMF is an autosomal recessive disorder with protean manifestations and an obscure cause that has a frequency of approximately 1 in 2700 in Israel, a country in which it has been studied intensively. Half of affected individuals are of Sephardic Jewish descent, about 20% are Armenian, 20% are Turkish or Arabic, and the remainder are Italian, Greek, or Ashkenazi Jews. FMF occurs rarely in Northern Europeans. Approximately 50% of patients do not have a positive family history. Manifestations usually appear during childhood or adolescence and are characterized by brief episodic febrile attacks, recurring in varying intervals and associated with painful inflammation involving a variety of serosal surfaces, including the abdomen, chest, joints, and skin, lending the alternative name of recurrent familial polyserositis. Attacks last typically for 1 to 2 days and occur once or twice each month. The natural history of the attacks can vary considerably, even in an individual patient. Clinical features include fever, abdominal pain, and signs of peritonitis in most patients. Because the peritonitis is nonspecific, other acute febrile conditions must be considered, such as appendicitis, cholecystitis, pancreatitis, and intestinal obstruction. Less constant features include pleuritic pain, mild arthritis of the large joints, and a transient erysipelas-like skin lesion on the lower extremities. In 1% of patients, meningitis also occurs. Arthritis is an episodic monarthritis or oligoarthritis of the large joints, mimicking oligoarthritic forms of juvenile rheumatoid arthritis with attacks usually lasting days to weeks. They may last for months and can be associated with radiographic changes of periarticular osteopenia without erosions. Laboratory test results are nonspecific during the attacks of pain and fever. The leukocyte count averages 16,000 cells/μL but may be as high as 40,000 cells/μL. The erythrocyte sedimentation rate increases, as do other acute-phase reactants. Albuminuria and microscopic hematuria also occur. Radiographic studies may show bowel edema and air-fluid levels in the small bowel, causing confusion with obstruction. The diagnosis is made in patients with the appropriate ethnic background, who have typical, self-limited, and recurrent attacks of fever and abdominal pain. The most severe feature of this disease is the progressive accumulation of a specific protein known as amyloid fibrillar protein, AA, in the kidney, which manifests clinically as a nephropathy. There is considerable ethnic variation in the incidence of amyloidosis in FMF. Amyloidosis occurs least frequently in Armenians with the disorder, even though FMF occurs most commonly in Armenians (1 in 400). Amyloidosis occurs commonly in non-Ashkenazi Jews (1 in 2400) and is most common in Moroccan Jews in Israel (up to 30%). In some patients, the amyloid develops before any other clinical sign. The genetic or environmental factors that account for these differences in the incidence of amyloidosis are not clear. The amyloidosis appears to be a phenomenon secondary to the periodic inflammation, because its occurrence is reduced by colchicine therapy. Colchicine (0.6 mg), administered orally, two to three times per day, is an effective treatment in the prevention of the acute febrile attacks of the disease. A low-fat diet (20 g/d) may also prevent the attacks. Colchicine does not allay symptoms if taken during attacks. Some patients use it intermittently to forestall attacks when they recognize that one is imminent. The cause of FMF is unknown. It seems to involve a genetically determined defect in the regulation of inflammatory responses. Research into the cause of FMF has included studies of suppressor T cells, leukocyte chemotaxis, lysosome release from neutrophils, and immunoglobulins, all without a definitive identification of the pathophysiological defect. The FMF susceptibility gene, *MEFV* or the pyrin gene, maps to chromosome 16p13, and both diagnostic and predictive testing of at-risk family members is possible. Although clinical heterogeneity exists, as reflected by the different prevalence rates of renal amyloid in different ethnic groups, there is no evidence for locus heterogeneity, because the FMF gene is linked to the same locus in the non-Ashkenazi Jewish and Armenian populations.

Fabry Disease/Diffuse Angiokeratoma Fabry disease, also called diffuse angiokeratoma, is an X-linked recessive lysosomal storage disorder caused by a deficiency of a-galactosidase. Deposition of lipoid material in several tissues is responsible for the characteristic findings. These include: attacks of pain in the abdomen and extremities (probably caused by lipid changes in ganglion cells of the autonomic nervous system), vascular lesions in the fundi of the eye and the kidney, corneal opacities, a hypertrophic cardiomyopathy, and characteristic skin lesions of a vascular nature called angiokeratoma. Symptoms may present in adulthood, and death usually occurs after renal failure in mid-adulthood. Female heterozygotes may have renal insufficiency and proteinuria, and rarely have skin lesions. a-Galactosidase activity is deficient in the leukocytes of male patients, and carrier females can be identified because of decreased activity. Several mutations in the a-galactosidase gene, *GLA,* at Xq22 in patients with Fabry disease have provided the definitive evidence to indicate that this is the gene for Fabry disease. Enzyme replacement therapy is now available to treat affected individuals.

Hereditary Angioneurotic Edema Hereditary angioneurotic edema is an autosomal dominant disorder that affects the respiratory and GI tracts. Symptoms of choking occur because of airway edema that can be precipitated by acts of trauma, such as tracheal intubation. GI symptoms include abdominal pain, nausea, vomiting, and diarrhea. Barium studies during an acute attack show bowel wall edema. Associated findings include an increased incidence of autoimmune disorders. Deficiency of C1 esterase inhibitor is the underlying pathophysiological defect in these patients. This protein regulates the first component of complement (C1) by inhibition of the proteolytic activity of its subcomponents C1r and C1s, preventing activation of C4 and C2 by C1s and inhibiting other serine proteases, including plasmin, kallikrein, and the coagulation factors XIa and XIIa. Low levels of C4 and normal levels of C1 are characteristic of hereditary angioneurotic edema. These low levels of C4 are responsible for the increased incidence of systemic lupus erythematosus, glomerulonephritis, and vasculitis among these patients. Heterozygotes have about 10% to 20% of the normal levels of C1 inhibitor rather than the expected 50%, possibly because of complex formation of the activated C1 inhibitor and activated C1 or because of increased catabolism of the protein. Androgens are effective in stimulating increased synthesis of C1 inhibitor from the normal allele and are useful for prophylaxis against attacks. Concentrates of C1 inhibitor are useful in acute attacks. The C1 inhibitor gene, *C1NH,* maps to chromosome 11q11-q13. Two clinically indistinguishable types of hereditary angioneurotic edema are described. Type 1 patients have a deletion of the C1 inhibitor gene or a truncated transcript, and type 2 patients have a single base substitution.

Hereditary Fructose Intolerance Hereditary fructose intolerance (HFI) is an autosomal recessive disorder caused by a deficiency of aldolase B (i.e., fructose-1-phosphate aldolase). This enzyme is expressed in tissues that metabolize fructose: the liver, kidney, and small intestine. It catalyzes the reversible cleavage of fructose-1-phosphate into dihydroxyacetone phosphate and glyceraldehyde. After fructose, sorbitol, or sucrose ingestion, patients with HFI present with severe abdominal pain, vomiting, and hypoglycemia. Liver and kidney damage, growth retardation, coma, and even death may result from chronic ingestion of fructose. Most patients with HFI learn to avoid fructose-containing foods. Problems arise when they are unable to avoid them, as during infancy, or if given intravenous fructose- or sorbitol-containing infusions for dehydration resulting from diarrhea and vomiting associated with illness, a practice carried out in some European countries. Intravenous fructose loading tests and measurement of aldolase B activity in liver biopsy specimens are methods used for the diagnosis of HFI. The aldolase B gene, *ALDOB,* maps to chromosome 9q22. It consists of nine exons that code for 363 amino acids. At least 10 mutations have been described; three missense mutations of *A149P, A147D,* and *N334K* account for 87% of alleles in the European HFI population and 68% in the North American population. The *A149P* mutation is the most common and widespread, accounting for 58% of HFI alleles in Europe and 55% in North America. Linkage studies suggest that the *A149P* allele arose once during evolution and spread through the mechanism of genetic drift, rather than occurring several times in different populations. Testing for the common mutations with amplified DNA specimens against a panel of allele-specific oligonucleotides would be an appropriate initial diagnostic approach that could identify most patients in a noninvasive manner.

Hereditary Pancreatitis Syndromes (See Also Chapter 98) Hereditary pancreatitis is an autosomal dominant disorder that is similar clinically to nonfamilial chronic pancreatitis. Steatorrhea and recurrent attacks of severe abdominal pain, fever, and marked elevation of serum amylase levels are characteristic. The elevated amylase level differentiates the disorder from FMF. The mean age at onset is 13 years. Between 5% and 10% of affected persons have pancreatic insufficiency, diabetes, and pseudocysts. As in nonsyndromic pancreatitis, emotional stress, alcohol, or a diet rich in fat can precipitate an attack. The disorder exhibits 80% penetrance. The gene for hereditary pancreatitis is the cationic trypsinogen gene, *PRSS1,* which maps to chromosome 7q32-qter. One predominant mutation in codon 117 results in an arginine to a histidine substitution that can easily be detected by restriction digestion. Codon 117 is a trypsin-sensitive site and when the arginine 117 cleavage site is replaced by histidine, trypsin continues to activate trypsinogen and other zymogens, ultimately leading to pancreatitis secondary to autodigestion of the pancreas. Mutations in the cystic fibrosis gene, *CFTR,* have also been associated with chronic pancreatitis when present in heterozygotes in association with the variable number of thymidines in intron 8, specifically the *5T* allele. Other genetic syndromes involving obstruction of the pancreatic duct, such as secretory pancreatic stone protein deficiency, familial hypertrophy of the sphincter of Oddi, or sclerosing cholangitis, pancreatitis, and sicca complex, can present as cases of familial or recurrent pancreatitis. Recurrent pancreatitis caused by a submucosal ampullary tumor has also been described in a patient with neurofibromatosis. Cases of hereditary or recurrent pancreatitis may be caused by genetic disorders of triglyceride metabolism, leading to hypertriglyceridemia, such as lipoprotein lipase deficiency, apolipoprotein C-II deficiency, and familial hypertriglyceridemia.

Gastrointestinal Neoplasm Syndromes (See Also Chapter 34, Chapter 62, Chapter 69, Chapter 80, Chapter 90, and Chapter 91)

Exciting and important applications of genetics to oncology and gastroenterology has led to the elucidation of genetic contributions to many GI malignancies. Details about GI and hepatic malignancies are further described in Chapter 62, Chapter 69, Chapter 80, and Chapter 91. Genetic disorders have been associated with malignancies in the esophagus, stomach, small and large bowel, pancreas and rectum as summarized in Table 127-8A and the general features that suggest an inherited predisposition to cancer are presented in Table 127-8B. Although most cancers are sporadic, approximately 10% are caused by the inheritance of an autosomal dominant germline mutation in a tumor suppressor or DNA recognition and repair gene leading to a significantly increased risk of developing cancer at a relatively early age. The clinical features of GI malignancies are covered in many other chapters of this volume and will not be discussed further here. An outline of genetic syndromes that are associated with increased risks for GI tract neoplasms are listed in Table 127-9. Predictive genetic testing and early preventive management may be useful for several of these conditions as illustrated by examples in Table 127-10. Increasingly, prognostic genetic tests will be used in oncology to help direct clinical management of patients.

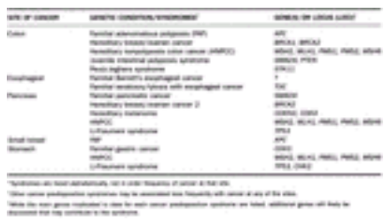

TABLE 127-8A Specific Cancer Sites in the Gastrointestinal Tract of the Hereditary Cancer Predisposition Syndromes*

- A clear inheritance pattern is suggested in the family history
- One particular type of cancer, or a limited number of different cancers, segregates in the family
- Cancer diagnoses occur at earlier ages when compared to the onset in the general population
- Family member has an identified genetic mutation associated with a cancer predisposition syndrome
- There is an unusual constellation of tumor(s)
  - —Multiple primaries in a single individual
  - —Bilateral cancers in paired organs of an individual
- Occurrence of relatively rare cancers not commonly seen in the general population clustering within a family
- Multiple recognized precursor lesions are present in the individual or family members
- Cancers are associated with other nonmalignant physical features or medical problems
- Constellation of cancers or other features that are part of a known cancer predisposition syndrome

TABLE 127-8B Findings Suggestive of an Inherited Cancer Predisposition Syndrome

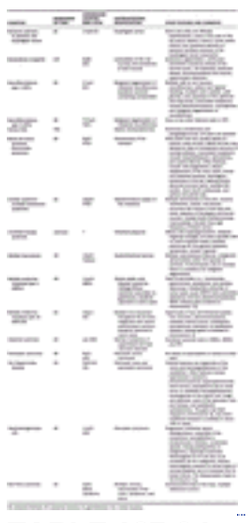

TABLE 127-9 Intestinal Neoplasm Syndromes

TABLE 127-10 Examples of the Use of DNA Testing and Ancillary Studies in the Evaluation of Cancer Predisposition Syndromes

Syndromes Associated With Benign Hepatic Masses and/or Cysts

Many genetic conditions are associated with benign hepatic masses and/or cysts. While oftentimes the hepatic manifestations do not result in significant clinical symptoms, recognition of the hepatic manifestations may be helpful in making the diagnosis of the genetic syndrome. Of the many multisystemic genetic syndromes that may be associated with benign hepatic lesions, the autosomal dominant disorders polycystic kidney disease, tuberous sclerosis complex, and von Hippel-Lindau syndrome are three conditions where clinical recognition is of paramount importance to insure that affected patients receive appropriate medical management. Von Hippel-Lindau syndrome is presented in Table 127-9.

Polycystic Kidney Disease There are both autosomal dominant and autosomal recessive polycystic kidney diseases. In general, autosomal recessive polycystic kidney diseases present very early in life, whereas the autosomal dominant forms generally present later with progression to end-stage renal disease around the fifth decade of life. While the main manifestations of polycystic kidney disease involve multiple cysts in the kidneys bilaterally with renal failure, hepatic cysts and colon diverticula may also develop. In addition, patients with the autosomal dominant form of adult polycystic kidney disease may have vascular complications including a high risk of intercranial aneurysms and an increased prevalence of cardiac valve disease. In general, the hepatic cysts are asymptomatic but the kidney cysts progress to end-stage renal disease in later adulthood. There is locus heterogeneity for autosomal dominant polycystic kidney disease with mutations in the *PKD1* gene at chromosome 16p13 and *PKD2* gene at chromosome 4q21-q23. In addition, there have been some families reported that do not map to either the chromosome 16 or chromosome 4 locus suggesting that there is at least one additional gene that predisposes to autosomal dominant polycystic kidney disease. In general, management of the disease consists of management of the renal complications with dialysis and kidney transplants.

Tuberous Sclerosis Complex Tuberous sclerosis (TS) complex is a multisystemic autosomal dominant disease with significant variable expression among affected individuals, even those within the same family. Several cutaneous features, including facial angiofibromas, hypopigmented macules, Shagreen patches, subcutaneous nodules, café au lait spots, and subungual fibromas, suggest the diagnosis. Retinal patches and astrocytomas can be identified on ophthalmology examinations. Dental examinations reveal pitted dental enamel and gingival fibromas. More serious complications include cardiac rhabdomyoma which may present early in childhood or infancy and neurological problems due to subependymal nodules, cortical tubers, and intracranial calcification that can be detected with head imaging studies and may lead to infantile spasms, seizures, and mental retardation in a significant number of patients. Cysts and angiomyolipomas are often seen in the kidneys and may also be identified in the liver of patients. The lesions in the liver are usually asymptomatic. In addition, lymphangiomyomatosis can also be identified in the lung and are important to identify in women of childbearing age. Patients are at increased risk for renal carcinoma. There is locus heterogeneity for this autosomal dominant condition with mutations in the *TSC1* gene on chromosome 9q34 and mutations in the *TSC2* gene at chromosome 16p13 accounting for the vast majority of cases. Although this condition is autosomal dominant, many patients are thought to possess new mutations and, therefore, there may be no family history of this disease. The *TSC* genes encode proteins that function as tumor suppressors.

Disorders of Bile Pigment Metabolism (See Also Chapter 44)

Several genetic disorders of bile pigment metabolism have been described. Although they vary in severity and clinical symptoms, most present with some degree of jaundice. The most commonly observed disorders are presented in Table 127-11.

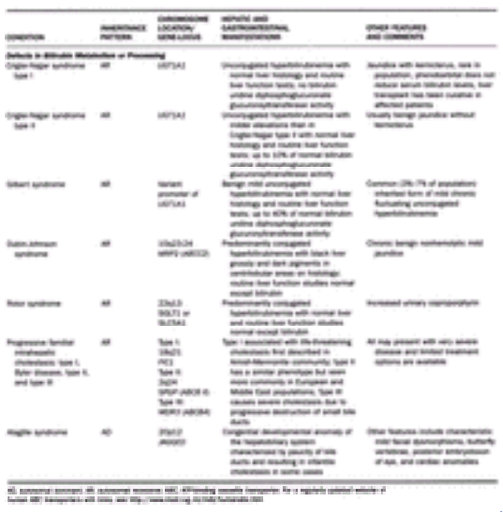

TABLE 127-11 Genetic Disorders of Bile Pigment Metabolism

Examples of Inborn Errors of Metabolism With Hepatic Manifestations (See Also Chapter 113)

Most of the inborn errors of metabolism are autosomal recessive or X-linked recessive disorders that present with multisystemic complications. Many are associated with a variety of hepatic findings including storage of precursor metabolic products, cirrhosis, and/or hepatomegaly. Discussion of classic metabolic diseases involving the GI tract and the liver are presented in Chapter 113. Many so-called metabolic disorders have been presented earlier in this chapter (i.e., hereditary fructose intolerance, Fabry's disease, hemochromatosis, etc.). A few additional diseases will be discussed subsequently to further illustrate the scope of different hepatic and GI manifestations that may be associated with inborn errors of metabolism. Although many inborn errors of metabolism present early in infancy or childhood, some do not present until later years.

Wilson Disease Wilson disease is an autosomal dominant disorder caused by mutations in the ATPase Cu $^{++}$ transporting ß-polypeptide gene (*ATP7b*) which is associated with high non–ceruloplasmin-bound serum copper levels. High urinary copper levels are also seen, as well as a low serum ceruloplasmin. Patients present with a variety of neurological and abdominal complaints. Often the onset of symptoms is in late childhood, adolescence, or early adulthood. Neurological symptoms include dementia, poor motor coordination, personality changes, dystonia, dysarthria, tremors, dysphagia, and decreased concentration. The main abdominal findings that can occur involve the liver and may include atypical hepatitis, cirrhosis, hepatomegaly, hepatic coma, and high liver copper levels. Esophageal varices can also arise. Renal tubular dysfunction and kidney stones may be seen. Loss of protein, amino acids, glucose, uric acid, phosphates, and calcium is often identified in the urine. Hypoparathyroidism and hemolytic anemia may also be observed. One characteristic ophthalmologic finding is Kayser-Fleischer rings. The diagnosis is based on analyzing serum copper levels and ceruloplasmin. *ATP7b* gene mutations lead to a dramatic build up of intracellular hepatic copper with subsequent and significant hepatic and neurological abnormalities. The gene maps to 13q14-q21. Treatment involves reducing serum copper levels. Agents including penicillamine and zinc have been successful in decreasing plasma copper levels.

Medium-Chain Acyl-Coenzyme A Dehydrogenase (MCAD) Deficiency MCAD is a relatively common autosomal recessive inborn error of fatty acid metabolism frequently seen in populations with the homozygote frequency estimated to be 1 in 6500 to 1 in 20,000, depending on ethnic background. Caucasians of Northern European descent exhibit the highest frequency of MCAD-deficient genotypes. MCAD-deficient patients are at high risk of hypoglycemia, vomiting, lethargy, encephalopathy, cardiorespiratory arrest, hepatomegaly, seizures, apnea, and sudden death in infancy. In fact, patients with this condition often die unrecognized with sudden infant death syndrome. Long-term outcomes may include developmental and behavior abnormalities, muscle weakness, cerebral palsy, and attention deficit disorder. Most often, infants are born healthy but present with symptoms after a fast or illness. It is in these times of metabolic stress caused by fasting or infection when the demands for fatty acid oxidation are high and patients may present with progressive symptoms. The MCAD gene is located on chromosome 1p31 and encodes an enzyme responsible for the metabolism of meaty enchained fatty acids. Newborn screening for this condition is now offered in several states, as recognition of the condition at the time of birth may allow for prevention of symptoms and sudden infant death syndrome by treating infections promptly and avoiding fasting conditions. Supplemental treatment including carnitine therapy may also be beneficial.

Tyrosinemia Tyrosinemia type 1 is an autosomal recessive condition due to mutations in the fumarylacetoacetate hydrolase (*FAH*) gene, which maps to chromosome 15q23-q25 and is required for the metabolism of tyrosine. Build up of tyrosine in the body can create multiple problems including failure to thrive, hypertrophic cardiomyopathy, renal Fanconi syndrome, renal failure, kidney stones, rickets, muscle weakness, episodic paralysis and peripheral neuropathy, abnormal blood coagulation studies, and a variety of abdominal problems. Hepatic problems that may occur include hepatomegaly, liver failure, cirrhosis, and associated ascites. Pancreatic islet-cell hypertrophy may also occur, as may splenomegaly. Gastrointestinal bleeding and paralytic ileus may also occur. Analysis of plasma amino acids is diagnostic. In addition, elevated a-fetoprotein, hypoglycemia, and hypophosphatemia can also be detected. Symptoms are highly variable and patients may have a progressive course that rapidly leads to hepatic failure while others may have acute and chronic hepatic crises. Hepatocellular carcinomas occur at high frequency in affected patients. The highest incidence of this condition is in the Saguenay-Lac St. Jean region of Quebec, Canada and northern Europe. Patients with this condition are often described as having an unusual "cabbage-like" odor.

Alpha-1-Antitrypsin (AAT) Deficiency AAT is an autosomal recessive condition with multiple allelic forms. Homozygotes for the PiZ form and some other much rarer variants are at a high risk of developing emphysema, particularly if they smoke, in early adulthood. These patients account for approximately 1% of all patients with emphysema. About 10% of the ZZ homozygotes develop liver failure that often leads to fatal childhood cirrhosis. This is due to an abnormal polymerization of the Z antitrypsin where the abnormal molecule has been poorly secreted in forms insoluble intracellular inclusions that are presumably toxic. In general, the infantile liver abnormalities occur when there is a less than 20% deficiency of the enzyme. Laboratory findings include a low serum a-1-antitrypsin (Pi) deficiency and abnormal liver function tests. Mutations in the protease inhibitor 1 gene on chromosome 14q32 are responsible for the phenotype. Again, multiple different mutations have been identified and different alleles are associated with different degrees of severity of disease. In the most severe form with hepatic failure in infancy, liver transplant has been used to treat this condition.

COMMON GASTROINTESTINAL AND HEPATIC DISORDERS WITH COMPLEX GENETIC CAUSES

In addition to the many monogenic, mendelian genetic disorders with GI and hepatic manifestations previously reviewed in this chapter, there are several common disorders whose genetic susceptibilities are inherited in a more complex or multifactorial mode. These disorders include conditions such as pernicious anemia, celiac disease (Chapter 76), inflammatory bowel disease (Chapter 83), cirrhosis (Chapter 116), gallstone disease (Chapter 100), peptic ulcer (Chapter 66), and sporadic colon cancer (Chapter 91). Common diseases often show familial aggregation caused by genetic factors or the combination of genetic and environmental factors. They also exhibit associations with certain genetic markers, often with specific HLA genotypes. Studies of family aggregation, twin data, subclinical markers, and genetic markers have been essential in demonstrating the importance of genetic determinants in the development of these common diseases. Many of the common complex gastrointestinal conditions are seen more frequently in specific genetic syndromes. For example, inflammatory bowel disease has been reported in individuals with Turner syndrome, Hermansky-Pudlak syndrome, glycogen storage disease type Ib, various immunodeficiency disorders, secretory immunoglobulin deficiency, gammaglobulinemia and hypogammaglobulinemia, selective immunoglobulin A deficiency, Chédiak-Higashi syndrome, deficiency of C2, hereditary angioedema, and chronic granulomatous disease. Further elucidation of genetic contributions to common non-mendelian GI and hepatic conditions will undoubtedly improve our understanding of their pathophysiology and may ultimately lead to susceptibility testing and preventive management for some of these conditions, at least in some populations in the near future. All of these common complex GI and hepatic conditions are discussed in great detail in other chapters of this text (see) and will not be further discussed here.

Acknowledgments

The author wishes to gratefully acknowledge the significant contributions of Drs. Maren Scheuner, Huiying Yang, and Jerome I. Rotter, whose related chapter in the second edition of this text formed the foundation for the current revision. The original words and tables of that chapter continue to live on in part of this revised chapter.

REFERENCES

The following on-line genetic databases and comprehensive genetic textbooks were used as the sources for all information contained in this overview chapter that summarizes salient features. Specific detailed references for each disease are found in these sources with the OMIM database being most comprehensive and up-to-date. Information from OMIM and its links were used to generate all tables.

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CHAPTER 128

Joel S. Levine

GASTROINTESTINAL MANIFESTATIONS OF SYSTEMIC DISEASES

Systemic diseases are commonly manifested by gastrointestinal signs or symptoms. The liver or gut may be the principal targets of the disease process or simply indirectly affected; in either case, the gastrointestinal manifestations may be the cause for seeking medical attention. The purpose of this chapter is to examine the systemic disorders that have well-recognized gastrointestinal manifestations and to stress their more important effects on the digestive system.

CARDIOVASCULAR DISEASES

Aortic Stenosis

Aortic stenosis, a relatively common condition affecting predominantly men in middle to late life [1], has been associated with an increased incidence of gastrointestinal bleeding, [2] often attributed to angiodysplasias of the gut (Heyde syndrome). [3], [4] Although case series suggest that 15% to 41% of patients with bleeding angiodysplasias also have hemodynamically significant aortic stenosis, no such relationship is found when aortic stenosis is sought in patients with nonbleeding colonic angiodysplasias. [5] Thus, the validity of the association between the two conditions has been questioned, [6] and probably lies more with the now recognized changes in blood coagulation associated with the abnormal valve that makes the angiodysplasias bleed in older adult patients.

Although most bleeding angiodysplasias in patients with aortic stenosis are found in the right colon, gastric and small bowel lesions also have been described. [7], [8] Lesions identified by angiography or endoscopy can be incriminated as the bleeding source if active hemorrhage is observed. [9] Endoscopic cautery, angiographic embolization, and intestinal resection remain standard therapy for active bleeding. [9] Aortic valve replacement has resulted in the cessation of bleeding in some cases, [10], [11] but the angiodysplasias persist. [12] A probable mechanism is that aortic stenosis can be complicated by a deficiency of the largest multimers of von Willebrand factor and impaired shear-induced platelet aggregation, and this is corrected by porcine valve replacement. [13], [14] The use of oral contraceptives has been suggested as an effective medical alternative to surgery. [15] These latter approaches to management need validation.

Congestive Heart Failure

Impairment of the heart's ability to fill or empty the left ventricle results in inadequate perfusion of tissues and a consequent inability to maintain tissue metabolic demands. Prevalent causes include hypertension, valvular heart disease, cardiomyopathies, anemia, and hypermetabolic states (e.g., thyrotoxicosis and sepsis). [16] Heart failure leads to congestion of the splanchnic venous bed, causing anorexia, nausea, bloating, and abdominal pain. Mesenteric ischemia of the bowel due to low flow states is not uncommon in such patients. [17] Rarely, venous congestion is sufficiently severe to produce weight loss, diarrhea, malabsorption, protein-losing enteropathy, [18], [19], [20] and [21] and the clinical picture of cardiac cachexia. Hepatic congestion secondary to right heart failure (Fig. 128-1) may mimic biliary tract disease or hepatitis because of jaundice, right upper quadrant pain, hepatomegaly, abnormal liver enzyme levels, low protein ascites, and prolongation of the prothrombin time. [22] Prolonged congestion rarely progresses to cardiac cirrhosis. [23]

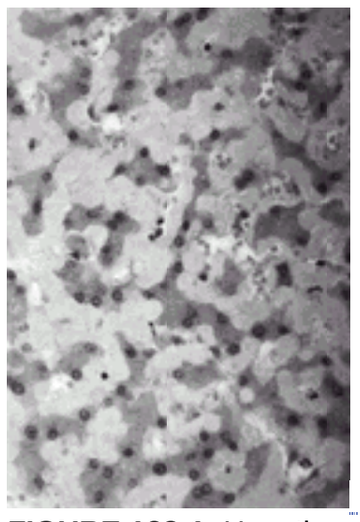

FIGURE 128-1. Hepatic congestion and cirrhosis secondary to congestive heart failure. (Hematoxylin and eosin stain; original magnification ×100.)

The medical therapy of heart failure can produce gastrointestinal side effects (Table 128-1). Digoxin causes anorexia, nausea, and vomiting in many patients. [24] Digoxin-associated constriction of splanchnic vessels may contribute to intestinal ischemia. [25] Quinidine and other agents used to control arrhythmias may produce nausea, anorexia, and diarrhea. Constipation or pseudoobstruction can result from diuretic-induced hypokalemia and hypomagnesemia. Severe pancreatitis has been attributed to a variety of diuretics, including thiazides, furosemide, and ethacrynic acid. [26] Oral potassium supplements have been implicated in esophagitis, ulcers,

and, rarely, intestinal strictures. [27] Angiotensin-converting enzyme (ACE) inhibitors are generally well tolerated, but may cause mild diarrhea. Rarely, ACE inhibitors have been associated with bowel obstruction and ascites due to angioedema of the intestine, as well as pancreatitis. [28]

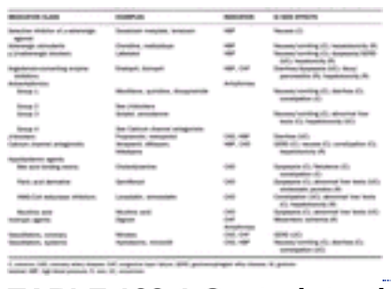

TABLE 128-1 Gastrointestinal Side Effects of Drugs for Cardiovascular Disease

The differential diagnosis of many of these complaints is long, and many patients are subjected to prolonged, fruitless investigation before the actual cause is discovered. Treatment is aimed at improving cardiac function. Cardiac medications should be changed or the dosage reduced when drug toxicity is suspected.

Coronary Artery Disease

Although the incidence of coronary artery disease (CAD) has decreased markedly since 1990, it remains the most prevalent cause of death in the United States. It is not uncommon for the initial presenting symptoms of angina or infarction to mimic gastrointestinal disease (epigastric pain or pressure, heartburn), which at times is confusing to both patient and physician. Once the diagnosis of CAD is certain, its treatments may lead to gastrointestinal problems.

Smooth muscle relaxants (calcium channel antagonists, nitrates) improve coronary blood flow but also relax the lower esophageal sphincter. Some suggest that the epidemic of esophagitis and esophageal carcinoma over the past two decades of the 20th century may relate to the common use of anti-anginal as well as other medications. [29] Constipation is also particularly prevalent with the calcium channel antagonists, and attention to bowel function is important. Lipid-lowering medications also are associated with constipation and elevations of transaminases, but most have mild and transient gastrointestinal symptoms that only rarely require discontinuation of the medication.

Large prospective trials have demonstrated the efficacy of low-dose aspirin in the secondary [30] and primary prevention [31] of myocardial infarction. In all trials, there is a small but consistent increase in the risk of gastrointestinal bleeding in the aspirin-treated groups. Although the risk of gastrointestinal bleeding is dose-related, even daily doses below 100 mg/d cause irreversible inactivation of platelet prostaglandin synthetase for 7 to 10 days. [32] Epidemiologic studies suggest that the concurrent use of nitrovasodilators reduce this risk substantially. [33] /SUP>Ticlopidine and clopidogrel, other antiplatelet agents, appear to be more effective than aspirin, are effective in preventing restenosis of coronary stents, are used with increasing frequency, and have a lower risk of gastrointestinal bleeding. [34]

More than 600,000 cardiac surgeries are now performed each year in the United States. Complications of cardiac surgery include acalculous cholecystitis, acute pancreatitis, mesenteric ischemia, hepatic necrosis, and others. [35] Gastrointestinal complications are recognized more frequently as management of these patients becomes more aggressive, as attested by reports of gastrointestinal bleeding after transesophageal echocardiography [36] and acute ischemic hepatic failure after malposition of an intraaortic balloon pump. [37]

CHROMOSOMAL ABNORMALITIES AND OTHER GENETIC DISORDERS

Abetalipoproteinemia

Abetalipoproteinemia is an autosomal recessive condition that causes severe hypolipidemia, diarrhea, fat malabsorption, growth failure, retinitis pigmentosa, acanthocytosis, and cerebellar ataxia. The growth failure and diarrhea can be improved with a diet modified to contain medium-chain triglycerides. Children with the disease can develop severe neurological disease from vitamin E deficiency, and fat-soluble vitamins must be monitored and replaced. [38] The disorder is a result of mutations of the microsomal triglyceride transfer protein, [39] and with current molecular technology it can be differentiated from a variety of hypobetalipoproteinemias caused by truncation producing mutations of the apolipoprotein B-100 gene [40] and from chylomicron retention disease.

Anderson-Fabry Disease

Anderson-Fabry disease, an X-linked glycosphingolipidosis, is caused by mutations in the gene encoding the lysosomal exoglycosidase, a-galactosidase A, with the resulting deposition of ceramide trihexose in the lysosomes of endothelial, neural, and smooth muscle cells. Diagnosis is made by skin biopsy, measurement of a-galactosidase in leukocytes, and genetic studies. [41] Gastrointestinal manifestations are present in most affected men and in 29% of female carriers and include episodic constipation, diarrhea, nausea and vomiting, and abdominal pain, and, rarely, intestinal ischemia and perforation. The cause of these symptoms may be related, in part, to impaired autonomic function and vascular insufficiency. [42]

Down Syndrome

Down syndrome (chromosome 21 trisomy) can be associated with multiple gastrointestinal anomalies, the presence of which predicts higher mortality. [43] Infants with duodenal stenosis and atresia present with vomiting, abdominal distention, and the “double-bubble” sign on abdominal radiography. Less complete obstructions from fenestrated duodenal membranes may present later in life as a cause of vomiting. [44] Esophageal abnormalities include tracheoesophageal fistula, esophageal stenosis, and gastroesophageal reflux. Imperforate anus, Hirschsprung disease, and malrotation of the intestine have been described. Celiac disease has a relatively high prevalence in these patients. [45] The high incidence of hepatitis A and B infection in patients with Down syndrome should be reduced with early vaccination.

Familial Mediterranean Fever

Familial Mediterranean fever (recurrent polyserositis) is a recessive disease that is common in populations of Eastern Mediterranean origin. The gene responsible for familial Mediterranean fever has been cloned and designated *MEFV*. Three missense mutations have been identified and can be screened for using an amplification refractory mutation system assay. [46] The target organ appears to be the blood vessel, and vasculitis can be present. Clinical manifestations begin in childhood and include self-limited attacks of fever, synovitis, peritonitis, or pleurisy. [47]

The diagnosis is suspected when patients of Mediterranean extraction with a positive family history present with recurrent episodes of chest or abdominal pain, peritoneal signs, arthritis, fevers, and leukocytosis. The patient may have undergone previous fruitless diagnostic workups, including laparotomy. Frequent complications include amyloidosis, degenerative arthritis, renal vein thrombosis, and narcotic addiction. [47] Treatment with colchicine has been beneficial in preventing and ameliorating acute attacks. [48]

Gaucher Disease

Type 1 (nonneuronopathic or “adult”) Gaucher disease is a rare autosomal recessive disease caused by a deficiency of glucocerebrosidase and resulting in the amassing of an insoluble glucocerebroside in the lysosomes of macrophages. Adults present with hepatosplenomegaly, thrombocytopenia, and pathological bone fractures. Ascites or esophageal varices, which occasionally may bleed massively, have been described (Fig. 128-2; see also Color Fig. 128-2). Definitive diagnosis relies on assaying ß-glucocerebrosidase activity. [49] Enzyme replacement therapy with small doses of mannose-terminated glucocerebrosidase (alglucerase) or the recombinant form of glucocerebrosidase imiglucerase can offer the hope of a productive life for many patients. [50]

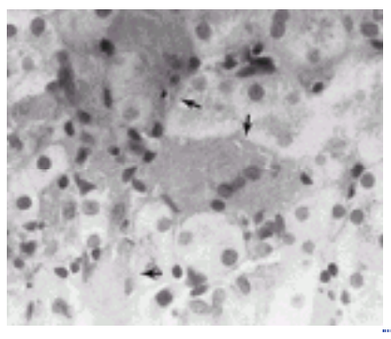

FIGURE 128-2. (See Color Fig. 128-2.) Gaucher disease of the liver. Multinucleated Gaucher cells (*arrows*) are present. (Hematoxylin and eosin stain; original magnification × 400.)

Hepatic Porphyrias

The hepatic porphyrias are a group of inherited metabolic disorders of heme synthesis. The acute hepatic porphyrias include acute intermittent porphyria, variegate porphyria, hereditary coproporphyria, and d-aminolevulinic acid (ALA) dehydratase deficiency. They are characterized by attacks of abdominal pain (commonly with vomiting and constipation) associated with hypertension, neuropathy, and neuropsychiatric disturbance that may last days to months. The abdominal pain is severe, may be associated by signs of peritoneal inflammation on physical examination, and can lead to unnecessary abdominal surgery. The peripheral neuropathy is manifest as pain in the extremities but may progress to a motor neuropathy. Photosensitivity only occurs in variegate and coproporphyria. Attacks can be precipitated by medicines (barbiturates, Dilantin, alcohol), menstruation, malnutrition, or other medical or surgical illnesses. [51], [52]

Sequence analyses of the genes for the enzymes of heme biosynthesis have revealed the porphyrias as highly heterogeneous, with multiple mutations underlying each type. All but ALA dehydratase deficiency are inherited as an autosomal dominant trait and are associated with increased urinary excretion of porphobilinogen (PBG) and ALA (Table 128-2). ALA dehydratase deficiency has normal excretion of PBG with elevated ALA. [53] Once the diagnosis is made, genetic counseling should be undertaken; patients are told to avoid precipitating factors and are placed on a high carbohydrate diet. Acute attacks may be ameliorated by infusion of glucose, hematin (being replaced by heme arginate), [54] and cimetidine 400 mg twice daily, [55] which suppress ALA synthetase. Treatment of seizures has been difficult because many anti-epileptic medications increase the hepatic metabolism of heme and can precipitate attacks. Gabapentin does not have this effect and seems to be safe and effective as a treatment of seizures in this setting. [56]

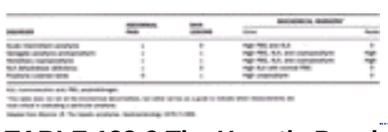

TABLE 128-2 The Hepatic Porphyrias

Porphyria cutanea tarda (PCT) is the hepatic porphyria inherited as an autosomal dominant trait, but its clinical presentation as a disease may depend on a number of other genetic and environmental factors. Characteristics include prominent photodermal and mechanical dermal sensitivity without abdominal pain. It is associated with alcoholic liver disease, hemochromatosis (or other disorders of iron overload), and hepatitis B and C. PCT has normal urinary excretion of PBG and is differentiated from variegate and coproporphyria by the increased urinary excretion of uroporphyrins. Abstention from alcohol, phlebotomy or desferrioxamine to reduce iron overload, low-dose chloroquine, and interferon-ribavirin treatment of hepatitis C all have reduced symptoms. [57]

Hereditary Angioedema

Hereditary angioedema is a rare autosomal dominant inherited deficiency of activated C1 inhibitor, a plasma protein that prevents uncontrolled intravascular activation of complement. Measurement of serum C4 screens for the disease, and an assay showing low serum C1 inhibitor confirms the diagnosis. [58]

The major clinical feature is episodic, self-limited, nonpitting, swelling of the orofacial region, larynx, extremities, and gastrointestinal tract. Angioedema is distinguished from urticaria in that it is asymmetric and burns rather than itches. Edema in the cervical region can compromise the upper airway. Abdominal attacks are characterized by colic, bloating, and nausea and vomiting secondary to enteric mucosal edema and can occur in the absence of peripheral or orofacial angioedema. Barium studies have demonstrated “thumbprinting” and other signs of mucosal edema. [59] Bradykinin is believed to be the main, if not the sole, mediator responsible for the edema. Treatment with synthetic androgens (e.g., danazol or methyltestosterone) increases the levels of C1 esterase inhibitor and can ameliorate or prevent attacks. Where available, infusions of C1 inhibitor concentrates are the treatment of choice for emergent attacks. [58] Premedication with C1 inhibitor concentrate or antifibrinolytic agents (e.g., e-amino caproic acid, tranexamic acid) should be considered before oropharyngeal manipulations such as upper endoscopy. [60]

Hyperlipidemias

The familial hyperlipidemias are a collection of inherited disorders of lipid metabolism, characterized by elevated plasma lipid levels. The epidemiologic association with atherosclerotic vascular disease is clear. Types I (hyperchylomicronemia) and V (hyperlipoproteinemia) are also associated with recurrent episodes of pancreatitis, as are patients with a deficiency of lipoprotein lipase. Patients with type IV hyperlipidemia also may be subject to an increased incidence of gallstones. Only disorders that predispose to high levels of triglycerides (>1000 mg/dL but usually >2000 mg/dL) are associated with acute pancreatitis. [60], [61] Many of these patients, particularly if overweight or diabetic, will develop fatty liver associated with abnormal serum liver enzyme levels, and will have nonalcoholic steatonecrosis on biopsy. The third trimester of pregnancy and the use of oral contraceptives are associated with an increased risk of developing severe hypertriglyceridemia. [62]

In the past 5 years multiple studies have demonstrated the beneficial effect of lipid lowering medication on the secondary prevention of coronary disease and stroke, with increasing evidence of benefit from primary prevention in patients with multiple risk factors. Statins (3-hydroxy-3-methylglutaric acid [HMG-CoA] reductase inhibitors) are commonly employed, and are associated with benign transaminase elevations in less than 1% of patients on higher doses. Hepatotoxicity is more common in patients who are also receiving drugs metabolized by the cytochrome P-450 enzyme systems, such as clarithromycin, fluconazole, fluoxetine, and cyclosporin. The symptoms of hepatitis are usually mild fatigue, anorexia, and weight loss. Aminotransferases are usually elevated two to three times the upper limit of normal. Although symptoms subside almost immediately on discontinuation of the statin, the liver tests make take several weeks to return to normal. Asymptomatic elevations of aminotransferases to 1.5 times the upper limit of normal can just be followed without stopping medication. Tests of liver function should be performed within the first 1 to 2 months of initiating therapy and every 6 months thereafter. [63]

Bile acid binding resins such as cholestyramine cause abdominal fullness, flatulence, and constipation in 30% of patients. These symptoms usually respond to dose reductions or the addition of psyllium or prune juice. Nicotinic acid, especially at doses more than 2000 mg/d, has been implicated in more common hepatotoxic reactions than statins. Gemfibrozil may cause severe abdominal pain, and is associated with increased frequency of gallstones. [63]

Niemann-Pick Disease

Niemann-Pick disease is a rare, inherited lipid storage disease that principally afflicts people of Jewish ethnicity. The enzyme sphingomyelinase is absent or nonfunctional, leading to the accumulation of sphingomyelin in the lysosomes of reticuloendothelial cells. Affected infants manifest severe neurological disease, hepatosplenomegaly, and hepatic failure and do not survive to adulthood. Survival of patients with type B Niemann-Pick disease to adulthood is common but may be associated with cirrhosis and portal hypertension. [64]

Tangier Disease

Tangier disease is a rare codominant inherited deficiency of plasma a-lipoprotein. The genetic defect has been localized to chromosome 9q31. Affected patients exhibit reduced total cholesterol, low

high-density lipoproteins, and elevated plasma triglycerides. Accumulation of cholesterol in the reticuloendothelial system results in enlarged orange or red tonsils, lymphadenopathy, peripheral neuropathy, hepatosplenomegaly, and yellow-orange patches in the colonic mucosa. [65]

Turner Syndrome

Turner syndrome (X chromosome monosomy) occurs in 1:2000 live female births and presents with a variety of external somatic features of which short stature and gonadal dysgenesis are almost invariably present. Gastrointestinal hemorrhage resulting from intestinal vascular malformations, an increased incidence of inflammatory bowel disease, and a greater risk of anorexia nervosa have been described. [66] Mild elevation of alkaline phosphatase may be seen. This cholestasis is not related to the chronic use of estrogens for the gonadal dysgenesis. [67]

Von Hippel-Lindau Disease

Von Hippel-Lindau (VHL) disease is an autosomal dominant inherited disorder characterized by hemangioblastomas of the retina and central nervous system, renal cell carcinoma, pheochromocytoma, and endolymphatic sac tumors with marked phenotypic variability. Heterogeneous germline mutations lead to the absence of the VHL protein resulting in uncontrolled production of vascular endothelial growth protein and the formation of blood vessels. [68] Pancreatic disease is found in three fourths of the affected individuals. The most common lesions found were cysts, serous cystadenomas, and neuroendocrine tumors. These lesions rarely cause symptoms and only require surgical intervention when very large. [69]

CONNECTIVE TISSUE DISEASES

Ehlers-Danlos Syndrome

Ehlers-Danlos syndrome (EDS) is a collection of inherited diseases affecting collagen synthesis. Nine different subtypes are described, although gastrointestinal involvement is common only in types I (gravis) and IV (arterial). Patients with type I EDS have fragile, hyperelastic skin and hyperextensible joints. They often complain of bleeding gums and easy bruising after even minor trauma. Severe gastrointestinal bleeding from mucosal lesions occasionally occurs. [70]

EDS type IV is unique in that hyperelastic skin is lacking and joint hyperextensibility is less prominent. Ecchymoses, thin skin with visible vessels, and “cigarette paper” scars are common, however. Cultured fibroblasts that synthesize abnormal type III procollagen molecules or the identification of a mutation in the gene for type III procollagen (*COL3A1*) confirms the diagnosis. Most complications occur after age 20. The defect in type III collagen synthesis leads to multiple aneurysms, including in the arteries of the splanchnic system. Rupture of these aneurysms results in intra-abdominal bleeding. Spontaneous perforation of the bowel, particularly the sigmoid colon, is common and a cause of death. Tissue fragility and poor wound healing with dehiscence contributes to surgical mortality. Repeated perforations are seen. [71]

Mixed Connective Tissue Disease

Mixed connective tissue (MCT) disease, or overlap syndrome, is a heterogeneous autoimmune disorder that may present with clinical features of lupus, scleroderma, and polymyositis. Gastrointestinal manifestations may resemble any of those described (see below) for the aforementioned rheumatoid diseases. Esophageal symptoms of heartburn and dysphagia are most common, however, with either proximal or distal esophageal motility distorted. Treatment with corticosteroids may improve the swallowing abnormality. [72]

Polymyositis and Dermatomyositis

Polymyositis and dermatomyositis are rheumatologic diseases characterized by inflammation of striated and, to a lesser extent, smooth muscle. Muscle biopsy is needed for diagnosis. A characteristic skin rash accompanies dermatomyositis. The gastrointestinal tract may be affected throughout its entire length, but the proximal esophagus usually is involved. Gastric and esophageal emptying is compromised in many patients, who may complain of dysphagia, aspiration, regurgitation, early satiety, and bloating. [73]

Colonic pseudodiverticula and pneumatosis intestinalis also may develop. Constipation is a common complaint. Suggested contributing causes include neurological dysfunction and diminished smooth muscle contractility as a consequence of muscle atrophy, fibrosis, or inflammation. Acute inflammation of smooth muscle may result in gut wall edema, ulceration, or perforation. An association of dermatomyositis and malignancy has long been postulated. In older adult patients colon cancer seems to be prevalent, and colonoscopic screening is appropriate. [74]

Progressive Systemic Sclerosis

Progressive systemic sclerosis (PSS) or scleroderma is characterized by vasculitis of small arteries and fibrosis of skin and other organs. Gastrointestinal manifestations are found in most patients. Tightening of the perioral skin with restricted ability to open the mouth and impaired taste sensation may contribute to malnutrition. Esophageal involvement, present in 70% to 80% of patients, is associated with more severe Raynaud’s phenomenon, skin abnormalities, and autonomic dysfunction. [75], [76] Smooth muscle atrophy leads to absent or low-amplitude esophageal contractions that are most prominent in the mid- and distal esophagus and weakening of the lower esophageal sphincter, leading to both reflux of acid and retarded clearance of the refluxed material. In addition, gastric emptying is commonly delayed, further increasing acid reflux. [77] The result is the most severe type of symptomatic reflux and erosive esophagitis. Evaluation of dysphagia may be difficult in advanced cases because it may be impossible to distinguish primary dysmotility from stricture. Endoscopy should be the primary diagnostic modality. Dilation of a stricture can substantially improve swallowing. The risk of developing strictures is high, and patients should be maintained on a proton pump inhibitor at a dose sufficient to suppress heartburn. Use of the more potent esomeprazole (40 mg/d) may be cost-effective in this setting.

Gastrointestinal hemorrhage is not particularly more common in PSS patients. In those with upper gastrointestinal bleeding, endoscopy may reveal telangiectasia [78] or gastric antral vascular ectasia (watermelon stomach). Chronic iron deficiency anemia, persistent occult blood in the stool, or recurrent melena and hematemesis are common manifestations. Cautery with heater probe or laser may reduce recurrent bleeding. [79]

The small intestine frequently is involved, causing severe morbidity. Hypomotility, dilation, pseudodiverticula, and thickening of the bowel are common. Stasis, pseudoobstruction, and bacterial overgrowth may produce anorexia, abdominal distention, postprandial periumbilical pain, malabsorption, diarrhea, steatorrhea, and weight loss. The bacterial overgrowth may be worsened by the need for effective inhibition of acid secretion because of concomitant gastroesophageal reflux. Erythromycin, cycled antibiotics, and octreotide [80] may reduce symptoms, but many patients require home parenteral nutrition.

Delayed colonic transit and impaired anal sphincter function have been demonstrated in patients with PSS. [81] Pseudodiverticula (Fig. 128-3) and telangiectasias are common. Constipation, pseudoobstruction, and volvulus may develop. Fecal incontinence and rectal prolapse are particularly disabling symptoms. Pneumatosis cystoides intestinalis may be found and is probably due to excessive hydrogen production by intestinal bacteria altering the partial pressure of nitrogen in the intestinal wall. [80], [82] Careful attention to bowel function has an important role in extending the patient’s functionality. Patients with the CREST (calcinosis, Raynaud phenomenon, esophageal dysmotility, sclerodactyly, telangiectasia) variant of PSS are at risk for the development of primary biliary cirrhosis.

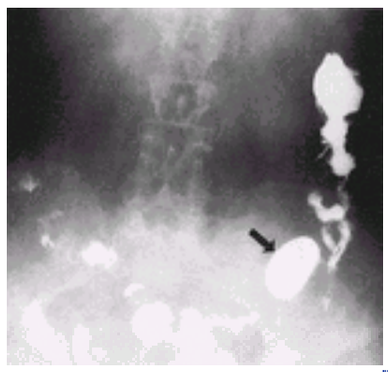

FIGURE 128-3. Wide-mouthed pseudodiverticula are present in a patient with scleroderma.

Pseudoxanthoma Elasticum

Pseudoxanthoma elasticum is a rare inherited disorder of connective tissue synthesis, characterized by plaques resembling xanthoma on the skin and angioid streaks on the fundus. Degeneration of elastic fibers in visceral blood vessels results in ineffectual vasoconstriction and vessel retraction after injury, predisposing to gastrointestinal bleeding. [83] Yellow raised plaques similar in appearance to the skin lesions have been identified in the stomach during endoscopy. [84] The best therapy for bleeding is uncertain, but arterial embolization has successfully controlled hemorrhage. [85]

Rheumatoid Arthritis

Rheumatoid arthritis is a chronic systemic disease characterized by a deforming symmetric polyarthritis and the presence of serum rheumatoid factors. It most commonly afflicts women between the 3rd and 7th decades of life and has a prevalence of about 1% in the United States. [86] Gastrointestinal manifestations are common and protean. Involvement of the temporomandibular joint impedes chewing. Sicca syndrome, when present, is accompanied by stomatitis and gingivitis. Heartburn and dysphagia are not as common as in PSS or MCT disease. Secondary amyloidosis may occur in patients with long-standing rheumatoid arthritis, may involve any portion of the gastrointestinal tract, may be manifest by chronic diarrhea, and can be diagnosed by endoscopic gastroduodenal biopsies. [87]

Vasculitis is less common than in other rheumatic diseases; however, both small and large vessels may be involved in the setting of severe arthritis, subcutaneous nodules, high titers of rheumatoid factor, and depressed serum complement levels. Necrotizing vasculitis of the mesenteric vessels may result in intestinal ischemia, bleeding, and infarction. [88] Cholecystitis, appendicitis, [89] perisplenitis, splenic infarction, pancreatitis, and hepatic arteritis have been described. [90]

Iatrogenic illness results in the most common gastrointestinal manifestations in rheumatoid arthritics. Chronic administration of salicylates or nonsteroidal antiinflammatory drugs (NSAIDs) that inhibit both cyclooxygenase 1 and 2 (COX-1 and COX-2) produce gastroduodenal erosions and ulcers, as well as hypocoagulability. COX-2 is an inducible enzyme that is associated with joint inflammation; its suppression leads to decreased pain, inflammation, and fever. COX-1 is a constitutive enzyme in the gastric mucosa, where it is necessary for the maintenance of normal mucosal integrity, and in platelets, where it is needed for normal aggregation. Although these lesions can be associated with anorexia, nausea, and abdominal pain, many patients are asymptomatic. [91] Larger, chronic ulcers of the stomach and duodenum will cause significant gastrointestinal bleeding or perforation in about 1% to 2% of patients each year. [92] The risk of bleeding is increased substantially (up to 9%–10%) in frail older patients who

- are taking certain NSAIDs (e.g., piroxicam)
- are taking higher doses of any NSAID
- have a previous history of gastrointestinal bleeding or ulcers
- are using steroids or aspirin with an NSAID
- have concomitant cardiopulmonary disease. [93]

Under ideal circumstances the patient with rheumatoid arthritis would be treated with joint-sparing immunotherapy alone (e.g., methotrexate) and not require chronic NSAIDs. In practice, however, many continue to have pain and inflammation that only responds to NSAIDs. For these individuals, the use of selective COX-2 inhibitors (celocoxib, rofecoxib) has been associated with decreased dyspepsia and bleeding. [94] Only patients who have multiple risk factors and who cannot use the selective COX-2 inhibitors should be considered for expensive prophylactic therapy with proton-pump inhibitors, or misoprostol. [95] Any of these approaches, however, can heal active ulcers if nonselective NSAIDs must be continued.

Methotrexate is commonly used to modify the disease progression of rheumatoid arthritis. Nausea and vomiting are common side effects during the first year of therapy and limit the use of this efficacious medication. Decreased renal function (either because of kidney disease or aging) increases the risk of side effects, and low-dose folic acid supplementation may be protective. [96] Although chronic therapy with low-dose methotrexate can result in hepatic fibrosis, when liver biochemistries are monitored the risk of developing cirrhosis is small in younger patients who do not drink alcohol. Hence, the clinical value of obtaining periodic liver biopsies is controversial. [97], [98] The toxic effects of gold preparations are discussed in the section on “Heavy Metal Toxicity.” Patients with Felty syndrome (rheumatoid arthritis, splenomegaly, and leukopenia) can develop intra-abdominal sepsis for reasons that are undefined as well as portal hypertension and variceal hemorrhage. [99]

Seronegative Spondyloarthropathies

The seronegative spondyloarthropathies are a collection of rheumatic diseases including ankylosing spondylitis, psoriatic arthritis, the reactive arthritides (e.g., Reiter syndrome), the arthritis associated with inflammatory bowel disease, and Whipple disease. Serum rheumatoid factor is lacking, but most patients are human leukocyte antigen (HLA) B27-positive, and many have recurrent oral and genital ulcers. [100] Patients with reactive arthritis frequently have an antecedent gram-negative enteric or chlamydial infection. [101] Many have chronic gastrointestinal symptoms (e.g., mild diarrhea, abdominal cramps), and most patients have endoscopic or histological evidence of ileocolonic inflammation that sometimes resembles Crohn’s disease. [102]

Sjögren Syndrome

Sjögren syndrome results from immune-mediated destruction of exocrine glands and is characterized by keratoconjunctivitis sicca (dry eyes and mouth). Atrophy of the salivary glands leads to a lack of saliva, which can, in turn, impair swallowing. In addition, esophageal dysmotility and mucosal webs are found. [103] Gastric atrophy and celiac disease [104] are reported with higher frequency in these patients than in age-matched controls. There is a strong association with the development of primary biliary cirrhosis. [105] Pancreatic acinar atrophy may result in pancreatic insufficiency and steatorrhea in some patients. [103]

Systemic Lupus Erythematosus

Systemic lupus erythematosus (SLE) is a systemic autoimmune disease that is most common among young and middle-aged women. Its characteristics include arthritis, myalgias, fever, rash, lymphadenopathy, renal disease, polyserositis, and central nervous system involvement. SLE is associated with abdominal pain of multiple causes. Inflammation of serosal surfaces may produce lupus peritonitis, [106] which usually is self-limited but may be associated with ascites. Self-limited peritonitis is difficult to distinguish from intestinal ischemia or perforation secondary to mesenteric vasculitis. [107] Pancreatitis, sometimes severe, perhaps related to vasculitis, has been associated with SLE. Intussusception, enteritis, and pneumatosis intestinalis may be seen. In particular, SLE may present with terminal ileitis, and it can be confused with Crohn’s disease. [108] Such vasculitis is usually associated with active disease in other organs. Malabsorption and protein-losing enteropathy with lymphangiectasia are documented. [109] Although clinically significant liver disease is uncommon in SLE, many patients have hepatomegaly and abnormal liver enzyme levels. Drug-induced hepatotoxicity is probably the most common underlying cause. [110] Abnormal liver enzyme levels and hepatocellular necrosis have been attributed to aspirin therapy, even when salicylate levels are within the therapeutic range. [111] Stopping aspirin therapy usually results in prompt regression of this syndrome. Other drugs, such as azathioprine, may produce similar abnormalities. Hepatic vasculitis is a rare cause of liver disease, which in extreme cases has resulted in liver rupture. [112]

DERMATOLOGIC DISEASES

Ataxia-Telangiectasia

Ataxia-telangiectasia is an autosomal recessive disorder that is characterized by progressive ataxia, telangiectasia on the bulbar conjunctiva and sun-exposed regions, sinopulmonary infections, immunodeficiency, and a propensity for lymphoreticular and other malignancies. [113] Rarely, hepatic venoocclusive disease has been reported in the absence of bone marrow transplantation or coincidental hepatitis infection. [114]

Blue Rubber Bleb Nevus Syndrome

Blue rubber bleb nevus syndrome is characterized by rubbery, blue nevi on the skin and gastrointestinal hemangiomas. The gastrointestinal hemangiomas can result in hemorrhage and iron deficiency anemia, may be the lead point of intestinal intussusceptions, and are commonly identifiable at the time of upper or lower endoscopy. Actively bleeding lesions can be cauterized, or the vessel can be occluded during angiography. [115], [116] and [117]

Cowden Syndrome

Cowden syndrome is an autosomal dominant disease characterized by ectodermal, mesodermal, and endodermal hamartomatous neoplasms. Patients manifest small papular lesions on the skin and oral mucosa. Diffuse esophageal glycogenic acanthosis may be characteristic of the syndrome. [118] There is a strong propensity for thyroid goiter, thyroid cancer, and, in women, breast cancer. Other organ systems may be involved. [119] Multiple hamartomatous polyps of various sizes can develop from the esophagus to the colon, but have a low potential for malignant degeneration. The hamartomas can cause chronic gastrointestinal blood loss and partial intestinal obstruction [120] (see Chapter 90).

Epidermolysis Bullosa

Epidermolysis bullosa is a group of diseases in which minor trauma disrupts the cohesion between epidermis and dermis, resulting in the formation of vesicles or bullae. Epidermolysis bullosa lethalis is an autosomal recessive disorder of the newborn period associated with extensive bullae formation and congenital pyloric atresia. [121] Other forms of this entity include epidermolysis dystrophica with either dominant or recessive inheritance and epidermolysis simplex of dominant inheritance.

Lingual adhesions and microstomia occur as a consequence of injury to the oropharyngeal mucosa during the eating of solid foods. [122] Clinically significant esophageal disease is seen more commonly in recessive epidermolysis dystrophica. Odynophagia and dysphagia are common because esophageal bullae lead to erosions, ulcers, pseudodiverticula, shortened esophagus, webs, strictures, and obstruction, most commonly in the cervical esophagus. Eating a soft or pureed diet may limit mechanical injury to the esophagus, but endoscopic placement of a gastrostomy tube may be needed. Strictures respond to frequent dilation with inflatable balloons. Bougienage probably should be avoided because the shearing forces could perpetuate injury. [123], [124]

Hereditary Hemorrhagic Telangiectasia

Hereditary hemorrhagic telangiectasia (Osler-Weber-Rendu disease) is an autosomal dominant condition found in 1 to 2 per 100,000 people. It is associated with the formation of telangiectasias, aneurysms, and arteriovenous malformations throughout the body. Cutaneous telangiectasia usually appears after the second or third decade of life and is located most frequently on the face, lips, nares, tongue, ears, and hands. The most prominent clinical feature of this disease is recurrent bleeding from mucosal telangiectasias. [125]

Epistaxis is the most common form of bleeding, with gastrointestinal hemorrhage developing in 10% to 40% of patients. Genitourinary, pulmonary, and intracerebral bleeding also may occur. [126] Hepatic vascular malformations cause arterioportal and intrahepatic portosystemic shunts, resulting in high-output congestive heart failure. Hepatomegaly, hemobilia, portal hypertension, esophageal varices, hepatic encephalopathy, bruit, and cardiomegaly have been noted. Elevated alkaline phosphatase and bile duct lesions similar to Caroli disease or sclerosing cholangitis can be encountered. [127]

Iron deficiency anemia is common and may require continuing oral or parenteral iron replacement or even transfusions. Endoscopic coagulation can effectively control life-threatening hemorrhage from a visible intestinal vascular malformation. In many cases, however, the bleeding site cannot be localized, and chronic hormonal therapy is initiated. If possible all antiplatelet medications (e.g., aspirin, NSAIDs) should be returned to the physician, as in such patients, estrogen-progesterone therapy can reduce the need for repetitive transfusions. [15] When large intrahepatic arteriovenous fistulae result in high-output congestive failure, embolization of these vessels or ligation of select branches of the hepatic artery may result in improvement of the failure, but infarction of the liver. [128]

Neurofibromatosis

Neurofibromatosis (von Recklinghausen disease) is an autosomal dominant inherited disease. The gene frequency is 1 in 3000 births. Cutaneous café au lait spots and neurofibromas rarely transform into sarcoma. [129] Polypoid neurofibromas can be present throughout the gastrointestinal tract, and it has been suggested that the Auerbach myenteric plexus may be the source of the tumors. [130] Rarely, the liver and gallbladder also are affected. [131]

Neurofibromas occasionally cause gastrointestinal bleeding and obstruction presenting as melena and abdominal pain. Gastrointestinal leiomyomas, sarcomas, and neurogenic neoplasms also are reported; [131] a somatostatin-rich carcinoid tumor of the periampullary region of the duodenum containing psammoma bodies also has been reported. [132]

Pemphigus

Pemphigus is a group of diseases characterized histologically by acantholysis resulting in bullae formation. Onset is usually between the 5th and 7th decade of life. D-Penicillamine, which is used in the management of Wilson disease, can cause pemphigus, although this is rare. Pemphigus vulgaris can involve the esophagus. Endoscopy shows blisters or erosions in the upper esophagus and linear red streaks along the entire organ, but clinically significant disease is uncommon. [133]

Psoriasis

Psoriasis is not associated with gastrointestinal disease; however, difficult psoriasis often is controlled by prolonged methotrexate administration. At the doses used, this drug carries the risk of potentially serious hepatotoxicity, including fatty liver, hepatic fibrosis, and cirrhosis. [134], [135] Serial liver biopsies are required to detect early hepatic damage because biochemical changes poorly reflect the extent of liver injury.

Stevens-Johnson Syndrome

Stevens-Johnson syndrome is an uncommon, but severe, hypersensitivity reaction characterized by a diffuse rash (erythema multiforme) that may progress to an exfoliative dermatitis. It is fatal in about 5% of cases. [136] This syndrome often is accompanied by fever and mucositis of both the gut and the respiratory tract. Etiologic agents include drugs (e.g., sulfonamides, allopurinol, anticonvulsants, NSAID) and infectious agents. The oropharyngeal mucosa frequently exhibits erosions and mucosal sloughing, although the entire gut may be involved in severe cases. Oropharyngeal pain may be severe enough to limit oral intake. Mucosal injury may result in dysphagia, odynophagia, abdominal pain, and gastrointestinal bleeding. [137] Esophageal stricture has been reported as a sequela of the Stevens-Johnson syndrome. [138] The syndrome is commonly associated with abnormal liver tests and hepatotoxicity.

Management is similar to that provided for major burns, and patients with extensive loss of skin should be managed in intensive care. Treatment includes identification and elimination of the inciting factors, rehydration, nutritional support, antibiotics, and vigilance for respiratory problems and secondary infections. Complete recovery may take several weeks. [136], [137] and [138]

Sweet Syndrome

Sweet syndrome is an uncommon condition, seen in women more than men, characterized by the acute eruption of tender, erythematous, non-ulcerating plaques and nodules. Nodules on the legs can mimic erythema nodosum. Diagnosis is defined by the histological presence of neutrophilic infiltration and leukocytoclasis on skin biopsy. The latter findings also are found in pyoderma gangrenosum, which, in contradistinction, is represented clinically by ulcerating plaques with violaceous edges. As with pyoderma, a strong association of Sweet syndrome with both ulcerative colitis and Crohn's

disease has been noted. Also reported are mouth ulcerations and an association with myeloma, myeloid leukemia, and adenocarcinoma of the colon. [139]

Tylosis

Tylosis is a rare autosomal dominant disorder characterized by thickening of the skin on the palms and soles (focal non-epidermolytic palmoplantar keratoderma) (Fig. 128-4; see also Color Fig. 128-4). Late onset tylosis (type A) is associated with squamous cell carcinoma of the esophagus. The causative locus, located on chromosome 17q25.1, has been termed the tylosis esophageal cancer (TOC) gene, [140] and has also been associated with sporadic esophageal cancer. [141] The esophageal mucosa forms papillomas. Affected persons have about a 90% probability of developing esophageal carcinoma by the age of 65 years, and should be screened with endoscopy and biopsies. [142]

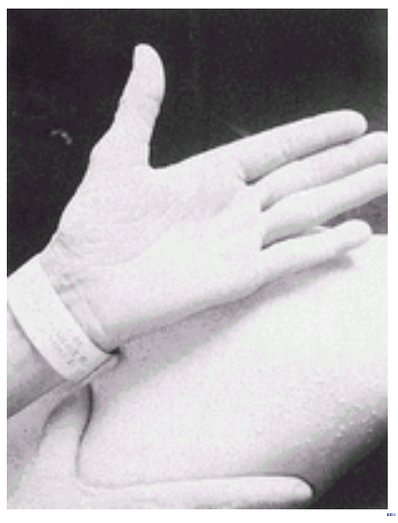

FIGURE 128-4. (See Color Fig. 128-4.) Hyperkeratosis of the palm and leg may be indicative of tylosis.

Urticaria

Urticaria is a reactive inflammatory vascular dermatosis characterized by vasodilation, increased vascular permeability, and extravasation of protein and fluids into the dermis, resulting in the characteristic peau d'orange wheals. Urticaria of less than 6 weeks' duration usually is related to inciting factors, such as contact allergy, medications, or food allergy. Chronic cases are usually idiopathic. Only a minority of patients with chronic urticaria will have systemic mastocytosis, which may be signaled by the more classic urticaria pigmentosa, flushing, and diarrhea. [143] Initial reports of an association with *Helicobacter pylori* infection appear unfounded. [144] Implicated in the pathophysiology of both acute and chronic urticaria is the release of histamine and other inflammatory mediators. Gastrointestinal manifestations have been reported in a number of patients suffering from chronic urticaria and include abdominal pain, gastroduodenitis, peptic ulcer, and enteric mucosal "hives." [143]

Urticarial vasculitis is a distinct entity in which the cutaneous lesions resemble urticaria but histologically show vasculitis. The persistence of urticaria longer than 24 hours and the presence of residual bruising suggest its diagnosis. It may accompany a collagen vascular disease, a familial complement deficiency, or an infection. Involvement of the gastrointestinal tract produces transient crampy abdominal pain, nausea and vomiting, and diarrhea. These symptoms may appear with cutaneous urticaria and are more in keeping with mucosal edema than with ischemia. Hypocomplementemia is frequently evident during the acute attack. [145]

ENDOCRINOLOGIC DISORDERS

Acromegaly

Acromegaly is a disorder whose clinical manifestations result from excessive growth hormone (GH) and insulin-like growth factor-1 production, usually a result of a pituitary adenoma. Rarely, carcinoid and pancreatic islet cell tumors can secrete a GH releasing factor that leads to acromegaly. Although patients manifest enlargement of the tongue, enlargement of the other organs of the digestive system is unusual, and hepatosplenomegaly thus would warrant further investigation. [146] It is generally accepted that patients with acromegaly are at greater risk for colorectal neoplasia, although this idea is controversial, and therefore should undergo screening by colonoscopy after the age of 50. [147], [148]

When GH secretion cannot be normalized by ablation of the adenoma, octreotide can control symptoms. As a consequence of the chronic use of the somatostatin analog, patients are at increased risk of developing gallstones; for this reason, reducing the lithogenicity of the bile with ursodeoxycholic acid may be considered. [149]

Addison Disease

Addison disease results from insufficient production of both mineralosteroids and corticosteroids by the adrenals. Gastrointestinal symptoms are common in untreated Addison disease. The prominent clinical features include anorexia, vomiting, weight loss, abdominal pain, apathy, hypotension, hyponatremia, and hyperkalemia. [150] Mucosal and cutaneous pigmentation occur in primary adrenal failure. Addison disease of autoimmune etiology is associated with atrophic gastritis, antibodies against K^+-H^+-ATPase and intrinsic factor, and uncommonly achlorhydria and pernicious anemia. Subclinical Addison disease may present with increased transaminase levels. [151]

Cushing Syndrome

Overproduction of adrenal corticosteroids caused by oversecretion of corticotropin from a pituitary adenoma, or ectopically produced in small cell lung cancers, leads to the typical redistribution of fat (buffalo hump, central obesity, cushingoid facies) that identify Cushing syndrome. Gastrointestinal manifestations, other than transaminase elevations from hepatic steatosis, are unusual. However, an uncommon type of a corticotropin-independent, food-dependent Cushing syndrome deserves comment. Hypercortisolism in these patients is induced by an overexpression of gastric-inhibitory polypeptide (GIP) receptors in adrenal adenomas and adrenal hyperplasia. Eating stimulates GIP secretion, GIP binds to the receptors in the adrenal gland, and abnormal cortisol synthesis and secretion are initiated. [152]

Diabetes Mellitus

Diabetes mellitus is a common metabolic disease resulting in primary hyperglycemia. Insulin-dependent diabetes mellitus (IDDM) is caused by a genetically mediated immune destruction of the pancreatic ß-cells, whereas the eight times more common non–insulin-dependent diabetes mellitus (NIDDM) is caused by insulin-resistance at the effector cell. NIDDM is more prevalent in minority groups, particularly Hispanic groups of Mexican origin, Native Americans, and some Asian-Pacific Islander groups. [153] Diabetics are always at risk of acute complications resulting from uncontrolled hyperglycemia, such as ketoacidosis or hyperosmotic coma and infections. After one or two decades of overt disease, complications often develop. Arteriosclerosis may result in myocardial infarction, stroke, or vascular compromise of the extremities. Other frequent complications include nephropathy with resulting renal failure, retinopathy progressing to blindness, and various sensory or motor neuropathies. For a variety of inadequately understood social, genetic, and cultural reasons, diabetes-related morbidity and mortality are also more common among minorities. [153] Abdominal complaints (Table 128-3) are common, nonspecific to diabetes, and may not relate directly to the diabetes. [154]

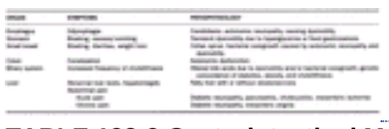

TABLE 128-3 Gastrointestinal Manifestations of Diabetes Mellitus

Acute abdominal pain associated with nausea and vomiting is a frequent accompaniment of acute diabetic ketoacidosis and resolves with normalization of blood sugar. Persistent, severe abdominal pain may signal that the ketoacidosis is the consequence of some other serious intra-abdominal process. Acute pancreatitis, cholecystitis, intestinal ischemia, appendicitis, and diverticulitis always must be considered.

Later stages of the disease may entail gastrointestinal symptoms of a chronic and irreversible nature. Many of the chronic symptoms are believed to result from visceral autonomic neuropathy, which is thought to be a consequence of a metabolic dysfunction of peripheral nerves caused by prolonged hyperglycemia. [155] Studies in which blood sugar is maintained within a normal range by using frequent daily administration of insulin demonstrated a delay in the onset of neuropathy and improvement of established neuropathy. [156]

Although clinical manifestations of esophageal dysfunction, often mild pyrosis, and dysphagia are unusual, abnormal esophageal motor function is common in both IDDM and NIDDM. [157], [158] Esophageal dilation, reflux, and delayed esophageal emptying may be seen radiographically. Changes demonstrated manometrically may include feeble peristaltic pressure waves that are at times multipeaked or multiphasic, increased frequency of nonperistaltic contractions, and decreased lower esophageal sphincter pressure.

New-onset odynophagia in the diabetic patient should suggest the presence of *Candida* esophagitis. Oral thrush accompanies *Candida* esophagitis in no more than 50% of patients, but when present it is an indication for an empirical trial of an antifungal agent such as fluconazole. If odynophagia persists, or if oral thrush is not present, diagnostic upper endoscopy should be undertaken. The most common visual finding is that of cream-colored plaques scattered throughout the esophagus. The diagnosis is made through endoscopic biopsy and brushing of the plaques. Other causes of odynophagia in the diabetic patient include herpes simplex esophagitis and esophageal spasm. Rarely, severe gastroesophageal reflux disease (GERD) secondary to decreased lower esophageal sphincter (LES) pressure can present with odynophagia.

Nausea and vomiting are common in patients with uncontrolled hyperglycemia and do not necessarily mean that the patient has the fixed vagal autonomic neuropathy seen in IDDM patients with gastroparesis diabeticorum. [158] Hyperglycemia, even in healthy volunteers using the glucose clamp technique, disrupts normal myoelectric activity in the stomach. There is an inverse relationship between gastric emptying and blood glucose concentration. [159] These alterations in gastric myoelectric activity result in gastric dilation, slow peristalsis, and delayed gastric emptying. Increased pyloric resistance and small intestinal dysmotility [160] also slow transpyloric flow. The patient's diabetes must be under optimal control before investigations are undertaken for gastroparesis.

Symptoms of gastroparesis include early satiety, bloating, heartburn, persistent nausea, and paroxysmal vomiting. Typically, the vomiting occurs 3 to 4 hours after meals and includes undigested food. Physical examination may demonstrate a succussion splash as well as findings of peripheral neuropathy. Patients are also predisposed to the formation of gastric bezoars from indigestible food. Yet most diabetic patients who have diabetes with abnormalities of gastric emptying have no symptoms. [161] Typical gastroparesis is defined by finding more than 1000 mL of gastric contents after awakening in the morning or by demonstrating a massively dilated stomach on routine abdominal radiographs or upper gastrointestinal series. Endoscopy supports the diagnosis by confirming the absence of structural gastric outlet obstruction and revealing a substantial gastric residual after an overnight fast. Other causes of gastroparesis that may be more treatable, such as hypocalcemia, hypothyroidism, or medications (e.g., narcotics, tricyclic antidepressants), must be excluded. Subtle gastric emptying dysfunction can be documented by gastroscintiscanning but must be interpreted with caution as abnormalities are found commonly in asymptomatic diabetics. [161]

Central to the treatment of diabetic gastric emptying disorders is the optimal normalization of blood glucose with appropriate diet, insulin, or hypoglycemic agents. Modification of concomitantly prescribed medications and correction of any electrolyte abnormalities are also important. In young women with IDDM eating disorders are common, and are associated with noncompliance, underdosing of insulin to achieve weight loss, and poor metabolic control. Symptoms can mimic gastroparesis. If the eating disorder is not treated progression of microvascular complications is more common. [162]

Therapy with prokinetic agents can be disappointing in advanced gastroparesis. The central dopaminergic and peripheral cholinergic effects of metoclopramide to accelerate gastric emptying may be short-lived because of tachyphylaxis. Although not available in the U.S., domperidone (up to 20 mg four times daily) can have longer-lasting improvement in symptoms despite a variable effect on tests of emptying, without the risk of the parkinsonian side effects of metoclopramide. [163] Although erythromycin improves gastric emptying in diabetic patients, [164] it has not been helpful in the long-term management of gastroparesis. Part of the unimpressive response to oral prokinetic agents may be due to the necessity for drugs to empty into the small intestine in a timely manner to achieve adequate blood levels. Surgery to improve gastric drainage should not be considered. Repeated measurements of gastric emptying are not useful because they do not discriminate well between symptomatic and asymptomatic patients and do not predict the response to treatment. [165]

Diarrhea and fecal incontinence are common complaints in advanced IDDM. [166] The diarrhea is watery, painless, and commonly nocturnal. It can remit after days or months, but recurrence is frequent. Although intestinal autonomic neuropathy may be the cause in as many as 50% of cases, other, more treatable, causes must be excluded. If steatorrhea is present, celiac disease, which is found more commonly in patients with IDDM, can be screened for with antiendomysial antibodies. [167] Pancreatic insufficiency and bacterial overgrowth also should be considered in the differential diagnosis. Fecal incontinence is commonly due to anal sphincter dysfunction and decreased rectal sensation, [168] but a flexible sigmoidoscopy will exclude proctitis.

Once other causes have been cleared, idiopathic diabetic diarrhea is treated with good glucose control, opiates, and bulk-forming agents containing psyllium. Some patients may improve with clonidine hydrochloride, an a_2-adrenergic agonist, which possibly stimulates an increase in intestinal fluid absorption by enhancing adrenergic activity; [169] however, this medication may induce hypotension. Other patients with suspected bacterial overgrowth may respond to cycled antibiotic therapy. Diarrhea control and biofeedback training [170] may provide complete or partial relief of fecal incontinence.

Severe constipation associated with colonic motor abnormalities [171] occurs in 20% of patients with diabetes and neuropathy. The onset of constipation is often insidious. Management includes the use of a variety of nonstimulant laxatives.

Diabetics can develop chronic abdominal pain secondary to thoracic radiculopathy. The pain is present constantly, is unilateral, and may or may not be associated with paresthesia or hypesthesia. Electromyographic examination may provide evidence of impairment of thoracic or lumbar nerve roots. Antidepressant drugs such as amitriptyline or desipramine may help control painful diabetic neuropathy. [172]

Diabetes may lead to a large, poorly functioning gallbladder that has a predilection for cholelithiasis. [173] Contrary to previous assumptions, patients with diabetes with acute cholecystitis may not have a substantially increased risk of complications compared with the general population. [174] Thus, diabetes mellitus is not an indication for prophylactic cholecystectomy.

Hepatomegaly and fatty liver are frequent findings, especially in diabetics with poorly controlled blood sugar, hyperlipidemia, and obesity. In a minority of such patients nonalcoholic steatonecrosis can develop, presenting with mild elevation of transaminases. Progression to cirrhosis is recognized, but weight reduction and diabetes control can normalize liver tests. [175]

Endometriosis

Endometriosis is defined as the presence of endometrial glands and stroma outside of the uterine cavity and musculature. The disorder is common to women during the years they menstruate, and it is associated with severe perimenstrual lower abdominal pains and infertility. Since bowel function is commonly abnormal around the menstrual period, the pains can be difficult to distinguish from irritable bowel syndrome or diverticulitis. Implants of endometrial tissue on the gastrointestinal tract are uncommon and usually asymptomatic. Most endometriosis takes the form of plaques of tissues on the serosal surface of the bowel lying in the pelvis in close proximity to the uterus and ovaries.

When intestinal endometriosis does cause symptoms, the symptoms can mimic a diversity of gastrointestinal illness. [176], [177] Deeper infiltration of the bowel wall can lead to obstruction of either the small or large bowel. Penetration of the mucosa can cause lower gastrointestinal bleeding, which may occur only during menstruation, but most patients do not characterize symptoms as cyclic. Rarely, perforation is reported. Radiographic and endoscopic findings may be difficult to distinguish from inflammatory bowel disease, diverticulitis, or colorectal cancer. Treatment is surgical and may

require bowel resection if implants are deep. [176], [177]

Hyperparathyroidism

Hyperparathyroidism is a disorder of calcium and phosphate metabolism due to the excess secretion of parathyroid hormone. Primary hyperparathyroidism is caused by adenomatous or hyperplastic change in the parathyroid gland and may be associated with inherited multiple endocrine adenopathies (see " Multiple Neoplasia Syndromes"). Rarely, carcinomas secrete a parathyroid-like hormone that can induce a similar syndrome. Primary hepatic tumors have been reported to do this more commonly. [178] Secondary forms of the diseases occur when there is a tendency toward hypocalcemia, such as chronic renal failure. Symptoms that are probably consequences of high serum calcium concentrations include anorexia, nausea, vomiting, constipation, and abdominal pain. Hypercalcemia induces gastric hypersecretion, and likely predisposes to peptic ulcer formation. [179] Pancreatitis can develop when hypercalcemia is moderate to severe. [180]

Hypoparathyroidism

Hypoparathyroidism, or low serum ionized calcium from whatever cause, can lead to neuromuscular irritability. Gastrointestinal manifestations can include abdominal pain, intestinal tetany, diarrhea, steatorrhea, and intestinal pseudoobstruction. [181] The steatorrhea may be secondary to pancreatic insufficiency due to insufficient endogenous cholecystokinin release following a meal. Hypoparathyroidism is present in most patients with the autosomal recessively inherited autoimmune polyendocrinopathy-candidiasis-ectodermal dystrophy syndrome. Associated manifestations include adrenocortical failure, gonadal failure, gastric parietal cell atrophy and pernicious anemia, and malabsorption with steatorrhea. [182]

Hyperthyroidism

Excessive circulating thyroid hormone causes hyperthyroidism. Although several disorders can induce this syndrome, 60% to 80% of the cases are a consequence of Graves disease, which is of autoimmune etiology. The symptoms of hyperthyroidism include nervousness, palpitations, heat intolerance, excessive perspiration, fatigue, weakness, dyspnea, and eye complaints. Despite having an enhanced appetite and hyperphagia, patients usually have weight loss because of their hypermetabolic state. [183] The most common physiological effect on the gastrointestinal system is an alteration in intestinal, but not gastric, motility, resulting in shortened small bowel and colonic transit time. This, as well as the increased consumption of fat, may result in steatorrhea and increased stool frequency, but rarely frank watery diarrhea. [184] Because of the higher prevalence of hyperthyroidism and irritable bowel syndrome in women than in men, the diagnosis of hyperthyroidism should receive strong consideration when a new diagnosis of irritable bowel syndrome is contemplated, especially in female patients. There is also a high incidence of atrophic gastritis and hypochlorhydria or achlorhydria. [185] Approximately 30% of patients have autoantibodies to the gastric parietal cell.

Moderate to severe hyperthyroidism can be associated with hepatic dysfunction. Increases in blood alkaline phosphatase, transaminases, and bilirubin have been attributed to liver injury. In many cases, passive congestion of the liver secondary to heart failure could be the underlying mechanism. [186] Autoimmune-type thyroid diseases can flare up when chronic hepatitis is treated with interferon. [187] Propylthiouracil, used in the treatment of thyrotoxicosis, can induce serious liver damage. [188]

Hypothyroidism

Hypothyroidism results from a deficiency of thyroid hormone. Most cases are a consequence of thyroid gland dysfunction resulting from Hashimoto thyroiditis or therapy of hyperthyroidism. Symptoms include weakness, dry skin, swelling of the extremities, mental aberrations, coarse voice, paresthesias, and muscle cramps. Patients have a decrease in metabolism and experience anorexia with moderate weight gain. Motility is slowed throughout the gastrointestinal tract. Dysphagia and heartburn may develop related to decreased esophageal peristalsis and diminished lower esophageal sphincter pressure. [189] Intestinal transit is slowed; it usually manifests as constipation and abdominal distention. Rarely, intestinal pseudoobstruction or megacolon results. [190] The gallbladder also may dilate and contract poorly. Other gastrointestinal manifestations include impairment in salivary, gastric, intestinal, and pancreatic secretion. [191]

Ascites, usually having a high protein concentration, is a rare complication of myxedema. It can be associated with histological changes in liver architecture suggestive of central congestive fibrosis, without congestive heart failure. [192] The ascites resolves after appropriate thyroid replacement therapy. [193]

GRANULOMATOUS DISEASES

Granulomas are dynamic, usually focal inflammatory processes composed of macrophages and other inflammatory cell types. Many factors such as infectious agents, silica, metals complexed to macromolecules, and immune complexes can evoke a granulomatous response. Even when there is a known nidus, the cause of a granuloma is not always evident on histological examination. Granulomas caused by an antigen that induces a delayed hypersensitivity reaction are called hypersensitivity granulomas. Diseases such as schistosomiasis, leprosy, tuberculosis, histoplasmosis, and berylliosis are hypersensitivity granulomas. Lesions evoked by substances that do not stimulate a hypersensitivity response are termed foreign body granulomas; examples are granulomas that develop in response to mineral oil and talc.

Many infectious and noninfectious diseases are associated with granuloma formation in the liver and intestines (Table 128-4). [194], [195] The significance of these associations is complicated by the observation that granulomas are present in 3% to 10% of liver specimens obtained at abdominal surgery or autopsy. The etiology of granulomas can, at times, be ascertained when bacteria, ova, or mineral crystals are identified within individual lesions. Some granulomas have distinctive morphology, such as the ring granuloma that is commonly found in hepatic Q fever, although ring granulomas have been described in other conditions rarely. [196], [197] Serologic testing, culture of the involved tissue, or clinical presentation may provide the diagnosis in other circumstances.

TABLE 128-4 Some Conditions Associated with Granulomatous Inflammation of the Liver (L) or Intestines (I)

In industrialized societies, the illnesses most commonly associated with hepatic granulomas are sarcoidosis and tuberculosis (discussed subsequently) (Table 128-5). Primary biliary cirrhosis, drug-induced liver disease, and occasionally other primary liver diseases account for some of the remaining cases. In many patients, it is not possible to identify an etiologic factor or an associated disease. Patients may be asymptomatic or may present with fever, fatigue, hepatosplenomegaly, and jaundice. Granulomatous hepatitis is suggested by a disproportionately high alkaline phosphatase with a normal biliary system on ultrasound. Diagnosis requires a liver biopsy that is cultured and examined histologically. The process rarely progresses to extensive hepatic fibrosis and cirrhosis, although sarcoidosis can lead to portal hypertension.

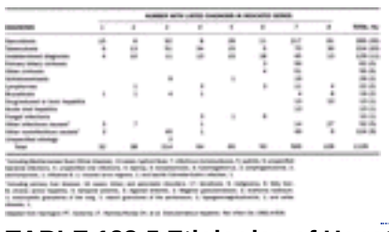

TABLE 128-5 Etiologies of Hepatic Granulomas in Several Reported Series from Around the World

Causes of granulomatous inflammation of the intestines are less well characterized (see Table 128-4). In the United States, Crohn's disease and lymphoma are overwhelmingly the most frequent diagnoses, but a rare case of intestinal tuberculosis may be encountered. All these diseases can have a chronic course, presenting with abdominal pain, fever, fatigue, weight loss, diarrhea, and gastrointestinal bleeding. All have a predilection for the terminal ileum and can result in intestinal strictures. [198]

When a specific cause of the granulomatous inflammation is defined, treatment is directed at eliminating the offending agent. Lesions that defy diagnosis or that are of unknown cause present special problems. Many patients are asymptomatic and require no therapy. Unusual conditions such as idiopathic granulomatous hepatitis frequently respond to corticosteroid administration.

Sarcoidosis

Sarcoidosis is a systemic disorder that is characterized by noncaseating granulomas in pathological specimens. The age-adjusted annual incidence of sarcoidosis in the United States is 10.9/100,000 for Caucasians and over three times more common in African Americans. In addition the disease affects blacks more acutely and more severely than people of other races. Although the cause of sarcoidosis is unknown, clustering of cases has suggested exposure to a common infectious or environmental agent. Genetic factors appear to play a part in the pathogenesis as it is likely that the host develops an exaggerated cellular immune response and the formation of granulomas when exposed to a variety of antigenic stimuli. [199]

Although the most common sites of involvement are the lungs and the intrathoracic lymphatic system, evidence of the disease will be found elsewhere in 40% to 70% of cases. Hepatic sarcoidosis is usually manifest by fever, hepatomegaly, and elevated alkaline phosphatase, and may be the most obvious clinical manifestation when pulmonary disease is subtle. [199], [200] Typically the noncaseating granulomas are scattered throughout the liver, but confluent granulomas may be present. In the patient with severe hepatic sarcoidosis and confluent granulomas scarring, portal hypertension, and hepatic dysfunction can result. A chronic cholestatic syndrome mimicking primary biliary cirrhosis is due to a granulomatous cholangiopathy with consequent ductopenia. Imaging studies may be normal, show a diffuse hepatopathy, demonstrate nodular lesions that can be mistaken for malignancy, and rarely show extrahepatic biliary obstruction due to lymphadenopathy or bile duct strictures. [201], [202] Treatment commonly includes steroids, but progression to cirrhosis and liver failure many ensue. Liver transplantation can be considered, but recurrence of sarcoidosis in the allograft is the norm. [203]

Although symptomatic disease in the liver, spleen, and abdominal lymphatics is common in sarcoidosis, involvement of the gastrointestinal tract is rare. The stomach is the most common site and may be defined by thickened gastric folds, nodularity, ulcers, or erosions. [204] As small bowel and colon disease is so rare, the demonstration of typical granulomatous ileocolitis in a patient with known sarcoidosis should still be followed and treated as presumed Crohn's disease.

Tuberculosis

Approximately one third of the world's population is infected with the causative organism of tuberculosis (TB), *Mycobacterium tuberculosis,* with over 8 million new cases reported annually. Despite an average annual decline in incidence in the United States of 5.8% per year from 1953 to 1985, the number of new cases of tuberculosis increased 20% over the next 7 years as a result of the human immunodeficiency virus/ acquired immunodeficiency syndrome (HIV/AIDS) epidemic, increased immigration, and diminished TB control. [205] Although extrapulmonary TB occurs in about 20% of cases, in the United Stated the gastrointestinal tract is involved in only 2%. [206]

TB may involve any portion of the gastrointestinal tract. At presentation abdominal pain, fever, and weight loss are the most common symptoms, and have usually been present for months. A mass is palpable in the right lower quadrant in 25% of cases. In the United States Crohn's disease, colon cancer, lymphoma, and even amebiasis will be considered before TB. The diagnosis of tuberculosis should be considered if:

- the disease has been documented elsewhere in more typical locations (e.g., the chest radiograph shows characteristic lesions, exudative ascites and peritoneal disease are recognized, tubercular tuboovarian disease is present)
- the patient is considered high risk (e.g., malnourished, immigrant from a developing country, patient with AIDS, alcoholic patient).

Tubercular granulomas may have distinct histological features: they tend to coalesce; they often contain giant cells that are numerous and large; and caseation, although rare in the liver and intestine, should suggest tuberculosis. In the unusual case, histological staining, polymerase chain reaction, or culture may identify acid-fast bacilli. Treatment follows accepted protocols. [207], [208] and [209]

Peritoneal disease with ascites is common in countries in which TB is endemic. In the United States TB peritonitis is associated with poverty, homelessness, alcoholism, and malnutrition. Abdominal distention, fever, weight loss, and abdominal pain in a patient with the appropriate risk factors should raise the suspicion for the disease. The chest x-ray will be abnormal in 50%, but active pulmonary TB will be present in only 15%. Paracentesis will usually show a high protein (>2.5 g/dL) fluid with a low albumin gradient (<1.1g/dL). Examination of the fluid for acid-fast bacilli is positive in less than 3% of cases, and cultures are positive in 20%. Adenosine deaminase activity greater than 33 U/L has excellent sensitivity and specificity. In nonendemic countries laparoscopy with peritoneal biopsies may be needed for diagnosis, whereas a therapeutic trial is commonly employed in areas with a higher prevalence of TB. [210]

HEAVY METAL TOXICITY

Lead Poisoning

Lead poisoning remains the most common form of heavy metal intoxication, often producing vague, nonspecific, and transient symptoms. Inner-city children in major metropolitan areas are at particularly high risk because of lead-based paint ingestion. The diagnosis should be considered in patients with abdominal complaints and a history of occupational or environmental exposure, such as ingestion or removal of lead-based paints, manufacture of batteries or jewelry, welding, automobile radiator repair, and eating from painted dishes acquired abroad. [211]

Common symptoms include recurrent severe abdominal pain (lead colic), oral ulcers, constipation, stocking-glove paresthesias, psychiatric symptoms, and a metallic taste in the mouth. Physical findings include a gingival lead line (absent in edentulous patients) and peripheral neuropathy. Anemia and renal dysfunction also may be evident at presentation. [212] Mild acute hepatitis that responds to chelation therapy has been described in the setting of lead intoxication. [213] Lead inhibits enzymes involved in the synthesis of heme, and secondary porphyria, characterized by elevated ALA and other porphyrin metabolites in the urine, may result.

Blood lead concentrations are an unreliable predictor of body lead stores, as they reflect only recent exposure. The diagnosis of lead intoxication is best established by measuring 72-hour urine lead levels after administration of calcium disodium edetate. [212] Treatment of lead poisoning with parenteral [204] or oral [214] chelating agents is usually successful if the diagnosis is made in time.

Arsenic Poisoning

Arsenic poisoning is endemic in many parts of the world, including North America, as a result of environmental contamination. Arsenic compounds continue to be used commercially, particularly in the preservation of wood and the manufacture of glass and metal. [215]

Acute arsenic poisoning results in a reversible inactivation of sulfhydryl-containing enzymes. Cellular oxidative processes are blocked and tissue hypoxia ensues. Patients present with severe colicky abdominal pain, nausea, vomiting, profuse diarrhea, malodorous ("garlicky") breath, dysphagia, hepatomegaly, jaundice, and circulatory collapse. Quantities of 100 to 300 mg may be fatal in an adult.

Quantitative measurement of 24-hour urinary arsenic excretion is the only reliable test to confirm arsenic poisoning. Treatment includes gastric lavage, chelation therapy (e.g., British anti-Lewisite [BAL, dimercaprol] or D-penicillamine), and cardiorespiratory support. [216], [217]

Chronic arsenic toxicity often manifests as nonspecific symptoms, including weakness, nausea, diarrhea, or constipation. Macular skin pigmentation, leukoderma, keratotic lesions of the palms and soles, pancytopenia, edema, peripheral vascular disease, neuropathy, cirrhosis and accompanying presinusoidal portal hypertension may appear alone or in combination. [218] The diagnosis of chronic arsenic intoxication is made by Gutzeit or Reinsch tests or by measuring arsenic levels in tissue samples (e.g., hair, nails). An association with cutaneous, hematologic, respiratory, and hepatic malignancies (angiosarcoma) also has been suggested. [219]

Gold

Gold preparations (e.g., gold sodium thimalate, auranofin) have long been used as disease-modifying treatment of rheumatoid arthritis. Adverse effects include aplastic anemia, proteinuria, rash, stomatitis, and diarrhea. [220] The last effect is particularly common with auranofin, an oral preparation that increases intestinal permeability and decreases intestinal transit time.

Rarely, gold can induce a severe enterocolitis, with markedly ulcerated small and large intestinal mucosa. The clinical picture can mimic ulcerative jejunoileitis seen in celiac sprue, or small intestinal lymphoma. The clinical course may be protracted, and if the relationship to gold therapy is not appreciated, a need for laparotomy. Gold should be withdrawn as soon as the condition is recognized. Corticosteroid therapy is of uncertain benefit. [221] Reversible cholestatic hepatitis also has been reported. [222]

HEMATOLOGIC DISORDERS

Hemoglobinopathies

Sickle cell anemia is an autosomal recessive disease of hemoglobin synthesis (hemoglobin SS). Eight percent of the African-American population of the United States are heterozygotes; hence, as many as 15 of every 100,000 African Americans may be affected. Under conditions of low oxygen tension, erythrocytes containing hemoglobin S-S deform (sickle), and become lodged in capillary beds, causing widespread venous congestion, thrombosis, and microinfarction and leading to the clinical vasoocclusive “crisis.” [223]

In addition to diffuse pains throughout the long bones of the body, abdominal pain and intestinal ileus are common during the acute crisis. It is often difficult to distinguish a sickle cell crisis from other gastrointestinal emergencies such as cholecystitis, appendicitis, bowel infarction, and bowel obstruction. Moreover, these intra-abdominal illnesses themselves may provoke a sickle cell crisis. The absence of bone or pleuritic pain is suggestive of a primary intra-abdominal cause. Sickle cell crises often resolve in less than 4 days. Hence, abdominal symptoms of longer duration also should raise the possibility of a primary abdominal illness. [224] Endoscopy and computed tomography (CT) scan of the abdomen can be helpful in distinguishing between the many potential causes of pain. [225]

The liver and spleen are frequently involved in sickle cell anemia. Acute crisis can selectively affect the liver with hepatic infarcts, causing pain, fever, and transaminase elevation. Chronic sickling causes cholelithiasis, cholecystitis, and chronic passive congestion. The common use of hypertransfusion programs to lower the risk of recurrent stroke places these patients at high risk of chronic hepatitis C and hemosiderosis. [226] A combination of these factors can lead to progressive liver injury, with significant fibrosis and cirrhosis developing by adulthood. [227] Splenomegaly is frequent in the early stages of the disease, but “autosplenectomy” caused by repeated splenic infarctions often results in a shrunken, fibrotic, or calcified spleen. Hyposplenism will be documented by the presence of Howell-Jolly bodies in the peripheral blood smear and signals an increased risk from encapsulated organisms, such as pneumococci and *Bacteroides.* [223]

Compared with hemoglobin S, patients with hemoglobinopathy C may have similar but milder attacks. In this disease, autosplenectomy is less common, and splenomegaly usually persists into adulthood. The risk for hepatobiliary disease remains high.

Patients with ß-thalassemia are characterized by a relative overproduction of a-globulin chains, consequent ineffective hematopoiesis, and severe anemia. Current therapy has depended on transfusion therapy. Iron overload of tissue, which is fatal if not treated, is the most important complication of this disease. In patients not receiving transfusions the enormous erythroid expansion leads to abnormally regulated iron absorption and increases in body iron burden ranging from 2 to 5 g/y. Regular transfusions may double this burden. [228] In the absence of chelating therapy iron progressively accumulates in the liver, heart, and endocrine organs, with resulting dysfunction. Iron-induced liver disease, often aggravated by chronic hepatitis C, is a common cause of death in adults. This can be delayed by adequate desferrioxamine therapy monitored by frequent quantitative measurement of liver iron stores with liver biopsy. [229]

Hemolytic Uremic Syndrome/Thrombotic Thrombocytopenic Purpura

The hallmarks of the hemolytic uremic syndrome (HUS) are hemolysis, thrombocytopenia, and acute renal failure. Although most common in children, all age groups are affected. Indeed, thrombotic thrombocytopenic purpura (TTP) is likely the same illness manifesting with fever and central nervous system sequelae in patients over 40 years of age. Both are now grouped under the thrombotic microangiopathies. Antecedent gram-negative enteric infections with *Salmonella, Shigella,* and *Campylobacter* species have been associated with HUS/TTP. [230], [231] In the past decade, verotoxin-producing strains of *Escherichia coli,* especially serotype O157:H7, appear to be responsible for most reported cases. It must be remembered that only a small minority (5% of patients with symptomatic enteritis in a recent outbreak) of patients with these food-borne illnesses develop HUS/TTP. [232] TTP has also been reported in patients undergoing solid organ transplantation, associated with the use of cyclosporin or tacrolimus. [233]

The pathophysiology connecting these enteric infections with the hematologic consequences is poorly understood. Endothelial cell injury (perhaps mediated by endotoxins or cytokines) followed by intravascular coagulation and, finally, thrombotic microangiopathy in the glomerulus and the gastrointestinal mucosa are consistent features (Fig. 128-5) [234], [235] Similar thrombotic angiopathy has been reported in gastric antral vascular ectasia (GAVE) syndrome (see Chapter 130). Thus, TTP should be included in the differential diagnosis of cases with endoscopic changes of GAVE and in whom biopsies show microscopic thromboses. [236]

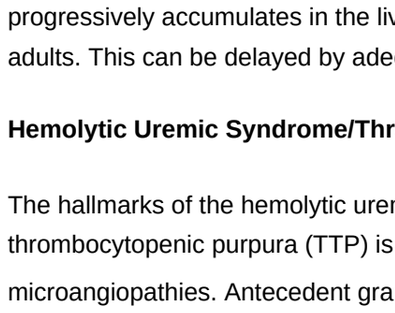

FIGURE 128-5. Colonic biopsy specimen from a patient with hemolytic uremic syndrome shows intravascular coagulation and thrombotic microangiopathy. (Hematoxylin and eosin stain; original magnification ×16.)

The spectrum of gastroenterologic symptoms ranges from gastroenteritis, to segmental colitis mimicking Crohn’s disease or ischemia, to fulminant colitis with toxic megacolon or colonic perforation. Bowel disease almost always precedes the development of HUS. [235] The disease is usually self-limited, although the course may be prolonged, and fatalities occur. There is now evidence that early treatment of the enteric infection with antibiotics increases the risk of developing HUS/TTP, and should be avoided. [237] Treatment focuses on general support and dialysis when needed. In adults with neurological changes (e.g., TTP) plasmapheresis can be life-saving. [234], [235]

Hereditary Spherocytosis

Hereditary spherocytosis is a disease of autosomal dominant inheritance characterized by small, fragile, spherical erythrocytes. Hemolysis leads to indirect hyperbilirubinemia, jaundice, and

splenomegaly in infancy. Pigment gallstones may be evident even in childhood. Splenectomy has been advocated for children and young adults, but it is of doubtful benefit to older, well-compensated patients. [238]

Hypercoagulable States

Hypercoagulability is an important cause of gastrointestinal morbidity. Spontaneous thrombosis of the mesenteric veins can cause intestinal ischemia and infarction (see Chapter 131) of the portal vein can lead to portal hypertension and variceal hemorrhage (see Chapter 33 and Chapter 147). Infarction of the hepatic veins can result in ascites and the Budd-Chiari syndrome (see Chapter 46). Oral contraceptives, pregnancy, SLE, myeloproliferative disorders (discussed later in the chapter), cancer, and surgery [239] all are associated with a hypercoagulable state and a consequent propensity for venous thrombosis. Antithrombin III deficiency [240] and protein C deficiency are inherited (autosomal dominant) disorders associated with recurrent venous thrombosis. Factor V Leiden mutation resulting in thrombophilia attributable to activated protein C resistance is a common genetic difference that has been associated with small bowel infarctions, [241] hepatic vein occlusion during pregnancy, [242] and an increased risk of venous thrombosis in patients with inflammatory bowel disease. [243] Once the cause is identified, most affected patients require lifelong therapy with warfarin to prevent recurrent vascular catastrophe.

Hypocoagulable States

Deficits of Coagulation Factors Deficiencies of coagulation factors may be inherited or acquired and when severe are associated with bleeding tendencies. The best known of the inherited factor deficits is hemophilia A, an X-linked disorder of factor VIII synthesis. Other inherited forms include hemophilia B (Christmas disease or factor IX deficiency) and factor XI deficiency. Gastrointestinal bleeding occurs in all these diseases. It may be mucosal, intramural, or intra-abdominal and may involve virtually any part of the gastrointestinal tract. Bleeding can be spontaneous or follow trauma or surgery. Upper gastrointestinal hemorrhage has multiple potential causes in these patients, including varices, because of the increasing incidence of chronic liver disease from blood-borne hepatitis viruses. Endoscopy can be undertaken safely in hemophiliac patients after correction of factor VIII level to 0.4 U/mL. [244, 245] Acquired or iatrogenic coagulation factor deficiencies are seen commonly as a consequence of vitamin K deficiency, heparin or warfarin therapy, or liver disease. Another cause is disseminated intravascular coagulation. In conditions such as SLE and cancer, circulating anticoagulants of endogenous origin can appear and interfere with normal coagulation.

Platelet Abnormalities Platelet defects also may result in severe bleeding diatheses that manifest as gastrointestinal bleeding. von Willebrand disease, the Bernard-Soulier syndrome, and Glanzmann thrombasthenia are inherited diseases of platelet adhesion and aggregation associated with prolonged bleeding times. Aspirin and other drugs (e.g., penicillins, cephalosporins) interfere with platelet function by irreversibly inhibiting COX-1 activity and prolong the bleeding time for up to 5 to 7 days. [246] Patients should be instructed to avoid the use of these drugs before and after elective endoscopic surgery and liver biopsy. In contrast, the effect of other NSAIDs on platelet function is reversible in 1 to 2 days, and endoscopic surgery can be performed safely shortly after discontinuing the drug.

Plummer-Vinson-Kelly Syndrome

Dysphagia, esophageal webs, and iron deficiency anemia are characteristic of Plummer-Vinson-Kelly syndrome. Middle-aged women are most commonly affected. Glossitis, dyspepsia, atrophic gastritis, diarrhea, hoarseness, and paresthesias are frequently present. [247] An association with postcricoid esophageal cancer has been reported. [248] The syndrome is uncommonly seen, which may reflect the improved identification and treatment of iron deficiency anemia.

Hypopharyngeal or esophageal webs of squamous epithelium cause the dysphagia. Inflammation is frequently present in the submucosa and may contribute to the dysphagia by producing fibrosis. The webs usually are thin (less than 2 mm thick) and will only be visualized when the esophagus is fully distended during esophageal cineradiography or endoscopy.

Although the significance of iron deficiency anemia with regard to the pathogenesis of Plummer-Vinson-Kelly syndrome is uncertain, iron and vitamin supplementation is indicated. Therapy includes dilation of the webs and fibrous tissue to alleviate the dysphagia. A careful endoscopic evaluation of the hypopharynx and upper esophagus also should be undertaken to exclude cancer.

METABOLIC DISORDERS

Capillary Leak Syndrome

Capillary leak syndrome is an episodic illness of variable severity characterized by increased small vessel permeability and fluid shifts. [249] Its association with systemic toxicity, sepsis, and administration of a number of recombinant protein medications, including interleukin-2 and granulocyte-macrophage colony-stimulating factor (GM-CSF), supports the contention that it is mediated directly by cytokines. [249, 250] In addition to hemoconcentration, hypotension, and a monoclonal gammopathy, patients can experience severe nausea, vomiting, and diarrhea secondary to impaired fluid absorption in the gastrointestinal tract. Therapy with aminophylline and terbutaline has been reported to ameliorate the syndrome in some patients. [251]

Systemic Amyloidoses

The systemic amyloidoses are a group of diseases characterized by the widespread extracellular deposition of insoluble fibrillar proteins. Classification of amyloidosis is based on the precursor plasma proteins that form the fibril deposits. Two types of primary systemic amyloidosis are recognized; AL (immunoglobulin-light-chain–related in patients with plasma cell dyscrasia) and ATTR (familial transthyretin-associated). Secondary amyloidosis is formed from serum amyloid A, an acute-phase reactant formed in response to inflammation. It is termed AA and caused by chronic infectious or inflammatory diseases, such as tuberculosis and rheumatoid arthritis. [252]

AL amyloidosis has the widest spectrum of organs infiltrated, with the kidney and heart most commonly involved. The gastrointestinal tract may be involved at any site. Infiltration of the tongue results in macroglossia in as many as 20% of cases, a feature that is uncommon in ATTR and AA forms of the disease. The esophagus is infiltrated in two thirds of patients, but clinical symptoms are uncommon. [253] Gastric infiltration can produce prominent gastric folds, but resulting gastric outlet obstruction, ulcer, and bleeding are uncommon. [254]

Amyloid can accumulate between the muscle layers of the intestine, causing coarsening of the valvulae conniventes (Fig. 128-6; see also Color Fig. 128-6). Mesenteric retraction and intestinal obstruction also can result from amyloid infiltration. Pseudoobstruction and autonomic dysfunction can lead to stasis, bacterial overgrowth, and malabsorption in some patients. [255] This is also a common feature of the ATTR type. Intestinal ischemia resulting in pain, enterocolitis, bleeding, and infarction also has been reported. [256]

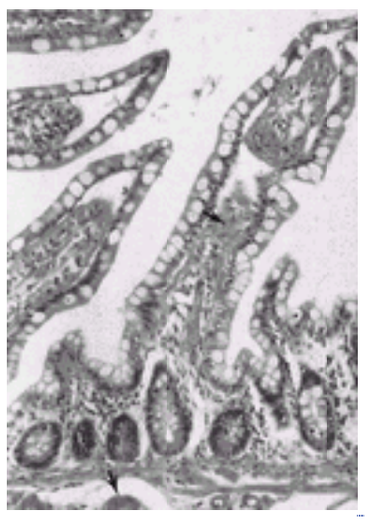

FIGURE 128-6. (See Color Fig. 128-6.) Amyloid infiltration of the small intestine (*arrows*). (Congo red stain; original magnification ×200.)

Hepatomegaly is common in AL amyloidosis but not splenomegaly. Hepatosplenomegaly is more common in AA amyloidosis, but liver failure and portal hypertension are uncommon. [257] Ascites may result from nephrotic syndrome-induced hypoproteinemia. Pancreatic involvement occasionally produces exocrine pancreatic insufficiency and steatorrhea.

The diagnosis of amyloidosis is based upon clinical suspicion and tissue biopsy of the affected organ. A safe and simple procedure, obtaining a sample of subcutaneous fat, has replaced the blind rectal biopsy recommended in the past. [252] Treatment of the underlying disease process, if present, may result in amyloid regression, but this is not the rule. [258]

NEOPLASTIC DISORDERS

Cancer Cachexia

Cancer cachexia, a common manifestation of a variety of advanced neoplastic diseases, is characterized by marasmus with anorexia, early satiety, weight loss, anemia, and edema. [259] Nutritional evaluations find an elevated basal metabolic rate and increased energy expenditure associated with systemic catabolism of muscle and adipose tissue. [260] Such tumor-induced, treatment-related malnutrition is the proximate cause of death in 20% to 30% of patients with cancer, but it also complicates the delivery of adequate antineoplastic therapy and markedly impairs the quality of life.

In exploring the cellular mediators of cancer cachexia, two groups of compounds have been suggested. Materials with hormonelike properties that result in the direct catabolism of host tissues such as endogenous corticosteroids or a lipid-mobilizing factor produced by tumors form one group. [261] The second group is the proinflammatory cytokines such as tumor necrosis factor-a, interleukin-6 (and 1), and interferon-?. [262] Since cytokines are also cellular growth factors, they may also increase tumor proliferation. Treatments aimed at these compounds might slow tumor growth as well as reduce cachexia.

Beneficial outcomes of the treatment of cancer cachexia could be just weight gain, but treatment would be more helpful if there was also an improved state of well being, improved function (Karnowsky index) and perception of quality of life, or improved survival. Simply replacing nutrients via gastrostomy or parenterally has not been helpful. The literature regarding the efficacy of nutrition support in patients with cancer has been critically reviewed, and no clear benefit was found. [263] Megestrol acetate has been shown to improve appetite and modestly increase weight. [264] In a trial both indomethacin and prednisone stabilized weight and the Karnowsky index, while patients treated with placebo worsened. Of interest was an increased survival (510 vs. 250 days) and decreased pain in the indomethacin group compared to both prednisone and placebo. [265] Such trials raise the hope that improving cachexia may not only offer palliative support, but also impact survival.

Consequences of Chemotherapy

Constipation Patients with cancer commonly develop constipation as a consequence of factors such as decreased physical activity, reduced fluid intake, depression, drugs used in treatment, and changes in diet. [266] Effective pain management may lead to the use of potent narcotics, all of which lead to constipation. Close attention to bowel function as opioids are being started will reduce the number of later problems with obstipated and impacted patients, who occasionally require hospital admission. [267] Although attention to diet is important, there are limitations to the amount of fibrous food the cancer patient will tolerate. Reversal of established opioid constipation requires agents that increase peristalsis, such as senna derivatives (see Chapter 43). Metabolic disorders and intestinal lumenal narrowing secondary to tumor growth or radiotherapy also can cause constipation. Constipation may result from hypercalcemia related to ectopic production of parathyroid-like hormone by neoplasms. Rarely, malignancy, particularly small cell carcinoma of the lung, produces a paraneoplastic visceral neuropathy that results in aberrant motor activity throughout the gastrointestinal system. [268]

Diarrhea Diarrhea is common immediately after potent chemotherapy. In adults, diarrhea may be a self-limited consequence of mucositis, but other causes need to be considered. *Clostridium difficile* colitis is a common cause. The spore-forming organism is found on the environmental surfaces of oncology units, [269] and antibiotics are ubiquitous. Because sigmoidoscopy is avoided in neutropenic patients, identification of the toxin forming *C difficile* in stools should lead to treatment with metronidazole or vancomycin. Several uncommon tumors are associated with the development of chronic diarrhea and, at times, steatorrhea that may be difficult to distinguish from chemotherapy-induced symptoms. [270] They release one or more of a variety of agents into the circulation that affect gastrointestinal function. These include carcinoid tumors, gastrinomas, glucagonomas, somatostatinomas, medullary thyroid carcinomas, pheochromocytomas, and vasoactive intestinal polypeptide (VIP) tumors. Diarrhea can be a rare consequence of some more common tumors, such as bronchogenic carcinoma.

Nausea and Vomiting

Nausea and vomiting, common after chemotherapy, exacerbate inanition and malnutrition. Inadequate management of these symptoms is a common reason for discontinuation of potentially effective chemotherapy or the unwanted conversion of outpatient to inpatient chemotherapy. [271] Cancer chemotherapy causes two main phases of vomiting: an intense, acute phase of emesis that occurs almost immediately following administration of anticancer drugs; and a milder, delayed phase of nausea and vomiting of longer duration. [272] It appears that the acute vomiting is mediated by serotonin (5-hydroxytryptamine, 5-HT) binding to 5-HT3 receptors in the central nervous system, triggering emesis. High doses of intravenous metoclopramide and the 5-HT3 receptor antagonists (e.g., ondansetron, granisetron) are most effective in treating acute-phase vomiting. [273] A wide variety of antiemetics of differing effects and costs is available to the clinician. [274], [275] and [276] Because the cost of newer pharmaceuticals needs to be matched against the increased total cost of therapy when a patient is admitted to the hospital with dehydration and intractable vomiting, clinicians may find an approach to acute and delayed emesis valuable (Table 128-6).

TABLE 128-6 Graduated Approach to Treatment of Chemotherapy-Associated Emesis*

Anticipatory vomiting can be difficult to control once initiated. The development of emesis induced by the sights and sounds of the setting in which chemotherapy is provided is more likely to occur in patients with pretreatment anxiety, posttreatment dizziness, delayed nausea and vomiting, and severe nausea and vomiting. [277] Behavioral therapy has been shown to be effective once anticipatory vomiting has developed. [278]

Hematologic Malignancies

Heavy Chain Diseases The heavy chain diseases are related neoplastic disorders of B cells. The neoplastic B cells produce abnormal monoclonal heavy chains. a-Heavy chain disease (Mediterranean lymphoma), the most common of these entities, is seen in two forms: pulmonary and enteric. The former is more common in the United States, and it rarely exhibits gastrointestinal manifestations. [279] The latter is found principally in the Mediterranean region in countries with poor sanitation. Infiltration of the small intestine and abdominal lymph nodes by malignant B cells and a-heavy chains produces abdominal pain and masses, vomiting, weight loss, malabsorption, steatorrhea, and hypocalcemia. [280] Additional complications include obstruction, intussusception, and intestinal perforation. Diagnosis is established by small bowel biopsy. Treatment includes initial staging laparotomy and debulking of tumor load if appropriate, followed by chemotherapy or radiotherapy. [281]

Leukemias Both acute and chronic leukemias can involve the gastrointestinal tract. The gastrointestinal manifestations result chiefly from leukemic infiltration of the lumenal gut and liver, or they are a consequence of therapy. Gingivitis is caused by leukemic invasion of the gums and causes oral pain and bleeding. Massively enlarged tonsils in acute lymphocytic leukemia rarely result in oropharyngeal dysphagia. Leukemic infiltration of the esophagus, stomach, intestine, or colon produces mucosal or intramural lesions that may cause dysmotility, obstruction, bleeding, or enterocolitis. [282], [283] Hepatosplenomegaly and portal hypertension are additional complications of leukemic infiltration. Rarely, an illness indistinguishable from fulminant hepatitis occurs. [284] Subcapsular splenic hemorrhage and splenic rupture have been reported, particularly in acute lymphoblastic leukemia. [285] Induction chemotherapy for leukemia can induce an often fatal syndrome characterized by acute

abdomen, paralytic ileus, diarrhea, and gastrointestinal bleeding. [286] Development of this syndrome often is attributed to necrotizing enterocolitis or typhlitis, a severe inflammatory process that involves the distal small bowel, cecum, or appendix (Fig. 128-7). [284], [285] It is seen most often in the setting of postchemotherapy neutropenia, but similar lesions have been described in other immunocompromised states. The etiology of the disease is unknown. Diarrhea (sometimes bloody), right lower quadrant pain or tenderness, nausea, and vomiting are the usual presenting features, developing over hours to days. The differential diagnosis includes infectious colitis caused by *C difficile,* Cytomegalovirus, and other organisms, appendicitis, Ogilvie syndrome, and bowel obstruction. Patients at risk must be observed carefully for evidence of perforation.

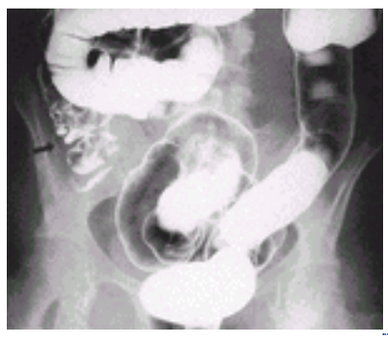

FIGURE 128-7. Typhlitis involves terminal ileum and cecum in a patient with acute myelogenous leukemia.

Lymphoma As many as 10% of patients with non-Hodgkin lymphoma originating from a nongastrointestinal site develop involvement of the gastrointestinal tract. [287] Obstruction, bleeding, abdominal masses, and perforation are among the more frequently recognized clinical features. Invasion of the liver and spleen is frequent, commonly presenting with fever and elevated alkaline phosphatase. Lymphoma in the liver can mimic acute hepatitis [288] and fulminant hepatic failure. [289] Mantle cell lymphomas can present as multiple lymphomatous polyposis and resemble the familial adenoma syndromes until biopsy is obtained. [290] Primary digestive tract lymphoma of the stomach (see Chapter 69) and the intestine (see Chapter 80) are discussed elsewhere. Clinically evident gastrointestinal involvement in Hodgkin disease is infrequent, although a substantial proportion of patients who die from this illness are found at autopsy to have hepatic involvement [291] (Fig. 128-8). Presinusoidal portal hypertension can result from either increased intrahepatic blood flow as a consequence of splenomegaly or intrahepatic infiltration with malignant cells. When the lumenal gut is involved, thickened mucosal folds are common (Fig. 128-9).

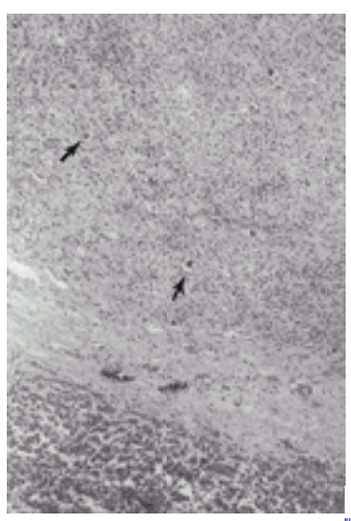

FIGURE 128-8. Hodgkin's disease infiltrating the liver. Reed-Sternberg cells are present (*arrows*). (Hematoxylin and eosin stain; original magnification × 25.)

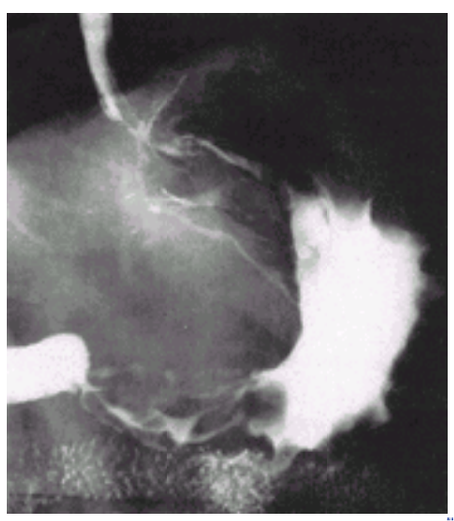

FIGURE 128-9. Hodgkin disease infiltrating the stomach. Thick folds in antrum and fundus.

Multiple Myeloma Multiple myeloma results from a monoclonal neoplastic proliferation of plasma cells and usually affects middle-aged and older people. Osteolytic lesions of the skeleton, hypercalcemia, anemia, monoclonal gammopathy, and renal disease are characteristic findings. [292] When the gastrointestinal tract is invaded, plasmacytomas form, with attendant abdominal pain, ulceration, bleeding, or obstruction. The lesions can be detected radiographically or endoscopically [293] and may look like peptic ulcer disease or colon cancer. Some patients have hyperviscosity syndromes as a result of elevated serum immunoglobulin. Visceral ischemia and thrombosis may result. Amyloidosis is a well-recognized complication of myeloma (see section " Metabolic Disorders"). Involvement of the liver and spleen results in hepatosplenomegaly and, rarely, portal hypertension. Jaundice can be caused by infiltration of the liver parenchyma or obstruction of the biliary tree secondary to myeloma in the pancreatic head. [294]

Myeloproliferative Disorders Myelofibrosis with myeloid metaplasia is characterized by ineffective erythropoiesis, dysplastic megakaryocyte hyperplasia, and an increase in the ratio of immature granulocytes to total granulocytes. This myeloproliferative disorder is associated with bone marrow fibrosis and extramedullary hematopoiesis in the spleen and liver, and presents with marked splenomegaly and progressive anemia. Portal hypertension, associated with ascites or variceal hemorrhage, occurs in about 7% of patients. The portal hypertension can be due to the increased portal flow through the enlarged spleen and obliteration of small portal venules, and is resolved with splenectomy. Patients can also develop acute left upper quadrant abdominal pain due to splenic infarction. [295] Paroxysmal nocturnal hemoglobinuria is a myeloproliferative disorder characterized by intravascular hemolysis that is manifest by episodes of hemoglobinuria and life-threatening venous thrombosis. In a large cohort of such patients seen over a 30-year period at one hospital, [296] the most common sites of venous thrombosis were the hepatic and mesenteric veins. This was also the most common cause of death. All such patients should be on long-term anticoagulation. Polycythemia vera is a myeloproliferative disorder that affects middle-aged people and is characterized by an increased red cell mass. Many patients have associated thrombocytosis. The erythrocytosis and hyperviscosity syndrome lead to plethora, headache, vertigo, dizziness, visual disturbances, vascular ischemia, and venous thrombosis. [297] Budd-Chiari syndrome may develop as a consequence of hepatic vein thrombosis. [298] Hepatosplenomegaly resulting from extramedullary hematopoiesis produces upper abdominal fullness in some patients. Bleeding esophageal varices are secondary to portal vein thrombosis and increased blood flow through the giant spleen. Primary or essential thrombocytosis is a myeloproliferative disorder characterized by platelet counts greater than $10^6/mm^3$ but without an increase in the red cell mass. This condition must be differentiated

from the secondary thrombocytosis that accompanies inflammation and iron deficiency. Platelet aggregation defects predispose to gastrointestinal bleeding or thrombosis. Hepatosplenomegaly is evident in many patients [299] and is caused by extramedullary hematopoiesis or Budd-Chiari syndrome.

Waldenström Macroglobulinemia Waldenström macroglobulinemia is an immunoglobulin M (IgM)-secreting variant of multiple myeloma characterized by the presence of macroglobulin (monoclonal IgM) in the serum. Osteolytic lesions are seen less commonly than in myeloma, whereas lymphadenopathy and hyperviscosity are more prominent. Massive hepatosplenomegaly results from infiltration by plasma cells. The deposition of large amounts of IgM in the intestinal lamina propria and mesenteric lymph nodes can impair absorption, [300] resulting in diarrhea and steatorrhea.

Nonhematologic Malignancies

Gynecologic Malignancies The treatment of cancer of the cervix, depending on stage of disease, usually involves a combination of a hysterectomy, chemotherapy, and radiation therapy. Because any invasion of the bowel can abort attempts at exenteration procedures, [301] a change in bowel habit or blood in the stool should lead to a colonoscopy before operation. Most gastrointestinal consequences of cervical cancer relate to the sequelae of radiation therapy. The risk of more severe radiation-induced bowel complications, such as obstruction or fistula formation, are doubled when adjuvant radiotherapy follows, rather than precedes, hysterectomy. [302] Although most radiation-induced bowel disease occurs in the first two years after radiotherapy, illness can occur at any time in the next several decades (see Chapter 132). Ovarian cancer can produce gastrointestinal complications by local extension and frequently encases the abdominal viscera. Intestinal obstruction, either because of direct invasion of bowel by peritoneal metastases or as a consequence of radiotherapy, is a common and disabling clinical complication. [303] In many terminally ill patients, palliation of vomiting can be a distressing management problem that is beyond the capabilities of reconstructive surgery. Octreotide at doses of 0.3 to 0.6 mg/d, in a small group of patients, rapidly relieved symptoms, and allowed removal of the nasogastric tube. [304] Refractory malignant ascites also can be an important cause of ovarian cancer morbidity, requiring repeated paracentesis or the placement of a peritoneovenous shunt for palliation. [305] Metastases to the ovary from gastrointestinal malignancies (Krukenberg tumors) may be difficult to distinguish from primary ovarian cancer by noninvasive imaging modalities. Typically, these tumors are bilateral, asymmetrically large, and solid. Almost all are mucinous signet-ring tumors from the stomach and colon, all with poor prognosis. [306]

Malignant Melanoma Malignant melanoma is a common malignancy whose prevalence is increasing. Although metastases to the gastrointestinal tract are commonly revealed at autopsy, fewer than 9% of patients with melanoma are diagnosed with gastrointestinal involvement while alive. [307] The small intestine, stomach, and colon are most commonly involved, but the liver, pancreas, and gallbladder also can be affected. Although most patients remain asymptomatic, complications include abdominal pain, anorexia, gastrointestinal hemorrhage, and intestinal perforation. In appropriate circumstances, surgical intervention to alleviate significant gastrointestinal complications is warranted and may prolong survival. [308]

Mastocytosis Mastocytosis is a rare disorder characterized by the proliferation of mast cells into the skin (urticaria pigmentosa) and other organs, including the intestines and liver. Common symptoms are headache, pruritus, flushing, dizziness, wheezing, and tachycardia. Urinary histamine excretion often is elevated. Gastrointestinal dysfunction develops more commonly than previously thought, resulting in abdominal pain, nausea, diarrhea, fecal urgency, malabsorption, gastritis, and peptic ulcers. [309] Rubbing cutaneous lesions or drinking alcohol may precipitate symptoms. Symptoms probably develop secondary to the release of many mast cell products, such as histamine, leukotrienes, and platelet-activating factor. [310] Many patients have liver disease, manifested by hepatosplenomegaly, portal hypertension with ascites, and abnormal liver tests. Liver biopsy shows portal fibrosis, nodular regeneration, and venoocclusive disease. Infiltration with mast cells is common but is seen only when special stains (toluidine blue and chloracetate esterase) are used. [311] Treatment with cromoglycate, or histamine receptor antagonists provides relief in some patients. [312]

Metastatic Disease to the Gastrointestinal Tract Intestinal metastasis from tumors originating in locations outside the gastrointestinal system can induce significant symptoms. Abdominal pain, obstruction, and bleeding are common consequences. Other than melanoma, malignancies derived from organs such as the breast, lung, and thyroid [313] can metastasize to the intestines and on occasion are diagnosed by endoscopic biopsies. In approximately half of the patients with Kaposi sarcoma, a frequent accompaniment of AIDS, intra-oral or intestinal involvement develops [314] (Fig. 128-10; see also Color Fig. 128-10).

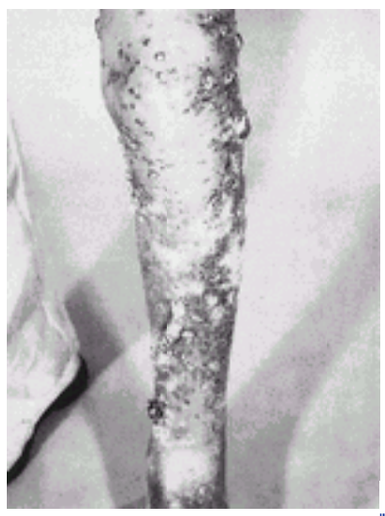

FIGURE 128-10. (See Color Fig. 128-10.) Kaposi sarcoma of the lower extremities.

Multiple Endocrine Neoplasia Syndromes Two main types of multiple endocrine neoplasia (MEN) are distinguished. MEN-1 is characterized by the combined occurrence of tumors of the pituitary gland, pancreatic islets, and parathyroid. Islet tumors may secrete gastrin, insulin, glucagon, VIP, and corticotropin. Resulting clinical syndromes will depend on the hormone that is pathologically secreted. [315] Inactivation of the MEN-1 gene locus is associated with the development of multifocal gastric carcinoids. [316] Treatment of such patients with somatostatin analogs (octreotide) is associated with the regression of the gastric carcinoids. [317] MEN-2A is characterized by pheochromocytoma, medullary thyroid carcinoma, and hyperparathyroidism. [315] MEN-2B is similar to type 2A but without involvement of the parathyroid glands. It is associated with the development of alimentary tract ganglioneuromas throughout the gastrointestinal tract and a characteristic facial appearance consisting of patulous lips and thickened tarsal plates of the eyelids. Gastrointestinal symptoms are common, particularly severe obstipation with megacolon that may be mistaken for Hirschsprung's disease. [318] Both MEN-2A and 2B have been associated with a mutation in codon 664 of the *RET* protooncogene. [319] Octreotide is used as a radiolabeled tracer to localize these tumors and for symptomatic therapy by reducing the release of secretory products from the tumors. [320] The use of octreotide is associated with an increase in gallstone formation.

Renal Cell Carcinoma Renal cell carcinoma (hypernephroma) can directly invade into, or metastasize to, the intestinal tract, causing bleeding [321] and obstruction. Resection of the primary tumor has been associated with regression of metastatic lesions. Nonmetastatic hepatic dysfunction (Stauffer syndrome) is common, usually manifested by abnormal alkaline phosphatase and prolongation of the prothrombin time, [322] and it is associated with hepatic sinusoidal dilation. [323] It is possible that in some cases the elevated phosphatase derives from the tumor itself and does not denote hepatic dysfunction.

NEUROMUSCULAR DISORDERS (Table 128-7)

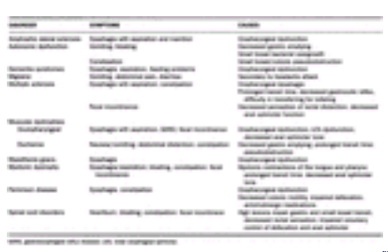

TABLE 128-7 Gastrointestinal Manifestations of Neuromuscular Disorders

Autonomic Dysfunction

The autonomic nervous system supplies every organ in the body, and dysfunction is associated with orthostatic hypotension, erectile problems, urinary retention, and temperature deregulation. Abnormal gastrointestinal function of the sympathetic and parasympathetic pathways in primary neurological disorders such as Parkinson disease, pandysautonomia, [324] or paraneoplastic syndrome [325] are manifested by vomiting, small bowel bacterial overgrowth, intestinal pseudoobstruction, fecal incontinence, and constipation. A number of gastrointestinal diseases can develop dysautonomic symptoms, particularly advanced liver disease. [326] Antibodies to ganglionic acetylcholine receptors have been found in a group of patients with idiopathic dysautonomia, suggesting an autoimmune etiology for some. [327]

A common association with the neurological diseases characterized by autonomic dysfunction is postprandial hypotension. Marked reduction in blood pressure after a meal may result in syncope, weakness, angina pectoris, nausea, and falls. The pathophysiology is complex and includes inadequate sympathetic nervous system compensation for meal-induced splanchnic pooling of blood, impaired baroreflex function, and peripheral vasodilation secondary to vasodilatory gastrointestinal peptides and insulin secretion. Similar symptoms are also seen in older adults without more global dysautonomia. [328]

Dementia Syndromes

Dementia syndromes, such as Alzheimer disease, multi-infarct dementia, and Huntington disease, demonstrate gastrointestinal symptoms related to oropharyngeal dysfunction, with dysphagia for solids and liquids and an increased risk of aspiration and progressive inanition and malnutrition. Diminished recent memory and disorientation impede rapid diagnosis of common and often easily recognized medical problems.

In advanced terminal neurological diseases, difficult decisions must be made pertaining to providing or withholding enteral feedings that must be delivered by nasogastric tube, gastrostomy, or jejunostomy. These techniques of enteral alimentation permit more effective, longer administration of defined enteral diets that can prolong the end stage of patients in a severely demented or “vegetative” state. Enteral support through feeding tubes is a form of medical treatment that is not necessarily mandatory for all patients in such condition, and indeed would not be the choice of most competent patients living in a nursing home. [329], [330] and [331] Percutaneous endoscopic gastrostomy is an easily applied, safe, and popular method of administering enteral feeding. Jejunostomies should be used only in patients with severe gastroesophageal reflux, in whom the procedure may provide additional protection against aspiration pneumonia.

Hiccups

Hiccups are intermittent, involuntary, spasmodic contractions of the diaphragm and accessory muscles of respiration that lead to abrupt ending of inspiration by airway closure at the glottal level. Transient hiccups are a common phenomenon often associated with overindulgence of food or drink, and they can be dealt with effectively by a variety of home nostrums and traditional remedies. Hiccups become a medical problem when they are prolonged and unresponsive to the usual approaches. Prolonged hiccups lasting for weeks or months are seen more commonly in men and can lead to insomnia, inanition with weight loss, and occasionally suicide. [332]

Particularly if the hiccups continue while the patient is sleeping, an organic cause should be sought. The potential causes of protracted hiccups are extensive and include foreign bodies in the ear canal, cervical tumors or adenopathy, central nervous system disease, metabolic disease (diabetes, uremia, hyponatremia), alcoholism, and chest (tumors or inflammation in the mediastinum, pericardium, pleural space, or diaphragm) disease. Gastrointestinal diseases associated with protracted hiccups include gastroesophageal reflux, gastric obstruction with distention, and stomach or esophageal tumors. Many of these problems can be defined with a careful history and physical examination, plain radiograph films of the chest or abdomen, and routine blood work. Further diagnostic studies may require CT of the chest, abdomen, and central nervous system, endoscopy, and otolaryngology and neurology consultations. For many patients, a specific causal etiology may not be found. [333]

In a practical sense, the most immediate problem confronting the clinician is how to abort the protracted hiccups. Assuming that the usual home remedies (e.g., breath holding, drinking water) have been unsuccessful, local stimulation of the pharynx with a nasogastric tube might be tried. Continued hiccups would lead to sequential trials of medications, none of which has been evaluated in randomized trials. Baclofen, a centrally acting antispasticity agent, has been effective when administered orally, 5 to 60 mg/d. [334] The dopaminergic antagonists (chlorpromazine, metoclopramide, haloperidol), amitriptyline, carbamazepine, diphenylhydantoin, quinidine, and nifedipine all have successfully stopped protracted hiccups. [335]

Migraine

Migraine is a periodic headache syndrome that usually begins early in life. Well-defined visual, sensory, or motor dysfunction may precede or accompany the headache. Gastrointestinal manifestations are almost universal and include nausea, vomiting, crampy abdominal pain, and diarrhea. The nausea and vomiting may impede the use of oral medications during an attack. A commonly employed antimigraine medication, sumatriptan, a selective 5-hydroxytryptamine-1 receptor agonist, has been associated with the development of ischemic colitis. [336]

Migraine equivalents such as cyclic vomiting [337] and abdominal migraine [338] are encountered, particularly in childhood. The diagnostic criteria include recurrent stereotypic attacks of either intractable vomiting or upper abdominal pain, no abdominal symptoms between attacks, a family history of migraine, and response to specific migraine therapy. These symptoms are only occasionally accompanied by headache or prodrome. Nortriptyline has been used with some efficacy in adults to prevent attacks of vomiting. The differential diagnosis of paroxysmal, repetitive abdominal pain includes the much less commonly encountered abdominal epilepsy, which is diagnosed by demonstration of spike and wave activity on the electroencephalogram.

Multiple Sclerosis

Multiple sclerosis (MS) is a focal demyelinating disorder of the central nervous system. Although oropharyngeal dysphagia occurs, in population-based studies, constipation and fecal incontinence are the most serious and disabling problems for patients with MS. Constipation is a problem for more than two thirds of MS patients, and about 25% have fecal incontinence weekly. [339] Causes of constipation are multiple, including difficulty in transferring to commode, increased segmenting contractions that lead to prolonged transit time, [340] and diminished gastrocolic response to a meal. Abnormalities of both anal sphincter function and diminished perception of rectal distention contribute to the incontinence. [341] The clinical use of improving outcomes of testing such as defecography or anal rectal manometry is unproved.

For patients with MS, careful attention to bowel function can markedly improve quality of life. Having an occupational therapist evaluate the home environment to improve access to commode, judicious use of fiber in the early stages of disease, and a strict bowel regimen that uses digital stimulation or glycerin suppositories and enemas to avoid impaction are all useful. Stimulant laxatives may increase bowel spasm and cause unpredictable bowel explosions that always are associated with incontinence and further lack of control.

Muscular Dystrophies

Muscular dystrophies are inherited myopathies featuring progressive weakness and muscle wasting. Although generally thought of as involving only the skeletal muscle, several forms have been associated with motor disturbances of the gastrointestinal system. Ptosis and dysphagia are the principal manifestations of oculopharyngeal muscular dystrophy. Radiographic abnormalities reflecting the global degeneration of skeletal muscle include altered clearance of barium from the pharynx, delayed initiation of swallowing, aspiration, and nasopharyngeal regurgitation. [342] More recent esophageal manometric studies also suggest abnormalities of the body of the esophagus and lower esophageal sphincter. [343] The external anal sphincter is also nonfunctional.

Duchenne muscular dystrophy is an X-linked recessive disorder that usually presents with initial proximal skeletal muscle weakness by the age of 5 years, progressing to incapacitation and death by the 3rd decade of life. Gastrointestinal symptomatology includes nausea, vomiting, abdominal distention, and constipation. Acute gastric dilation [344] and intestinal pseudoobstruction have been reported. [345] Gastric hypomotility occurs frequently in advanced disease. [346]

Myotonic Dystrophy

Myotonic dystrophy is an autosomal dominant, slowly progressive condition, with symptoms presenting in the 3rd decade of life. The distinctive features are delayed relaxation of muscle after initial contraction, muscle wasting of a characteristic pattern, frontal baldness in men, testicular atrophy, dysarthria, and cataracts. Studies have found abnormal muscle function throughout the gastrointestinal tract. Common symptoms are abdominal pain, dysphagia, emesis, chronic or episodic diarrhea, coughing while eating, and fecal incontinence. One fourth of patients with myotonic

dystrophy consider their gastrointestinal symptoms the most disabling consequence of the disease. [346]

Weak or myotonic contractions of the tongue and pharynx can result in oropharyngeal dysphagia and aspiration. Weakness of the upper esophageal sphincter and low-amplitude esophageal peristaltic contractions are frequent motor disturbances but may have little correlation with symptoms. [346] Delayed gastric emptying, prolonged intestinal transit, and abnormal anal sphincter function also may develop. [347], [348] Although rarely of clinical significance liver tests such as the alkaline phosphatase and transaminases may be mildly elevated. [349]

Parkinson Disease

Parkinsonism is characterized by progressive rigidity, tremor at rest, bradykinesia, and diminished postural reflexes. Autonomic dysfunction is common. Although 10% to 15% of cases occur in the 3rd and 4th decades of life, the disease increases exponentially in older adults. Over 3% of those over the age of 65 have Parkinson disease. Asians and African Americans are less likely to develop the disease than Caucasians. Regularly observed pathological changes include degeneration of pigmented brainstem nuclei, which contain distinctive eosinophilic intracytoplasmic inclusions termed Lewy bodies. Dementia is common and loss of sympathetic ganglia and parasympathetic neurons in the enteric nervous system has been documented. [350]

Gastrointestinal dysfunction is common and clinically important in patients with Parkinson disease. Dysphagia, bloating, nausea, constipation, and defecatory dysfunction are common symptoms. Over 50% of patients have swallowing disorders. For many, defined pharyngeal swallowing dysfunction is documented on swallowing studies and is similar to that seen after stroke or with amyotrophic lateral sclerosis. Although a variety of esophageal manometric abnormalities have been noted, they are not distinctly different from age-matched controls. [351] Gastric-emptying is disordered in many patients, and is associated with abnormal myoelectric activity. [352] The gastroparesis not only leads to disabling symptoms that may lead to malnutrition, it also may be responsible for some of the motor fluctuations seen with levodopa therapy. The drug therapy that appears to be most effective in reducing these symptoms, domperidone, [353] is not available in the United States.

Constipation characterized by colonic inertia is associated with depletion of dopaminergic neurons in the muscularis externa, [354] decreased colonic motility, and alteration of puborectalis and anal sphincter function. [351] Constipation is aggravated by the anticholinergic effects of anti-parkinsonian medications. The use of stimulant laxatives is common. Pelvic floor dyssynergia is frequently documented, and straining to defecate complicates the constipation. [355] A daily bowel regimen with the use of glycerin suppositories can be helpful.

Spinal Cord Disorders

Acute spinal cord injury is commonly associated with ileus, and patients are at increased risk of gastrointestinal bleeding. [356] Nasogastric suction and prophylaxis with continued acid suppression or sucralfate are warranted. After surviving the acute injury, constipation and fecal incontinence are almost universal and are a major source of disability and depression. [357] Over time laxative dependence, dilated colon, and increasing time to generate a movement occupies a great deal of the patient's day. Rectal sensation, voluntary control of defecation, and normal anal sphincter function are lost. [358] Abnormal colon compliance and motor activity are also present. [359] A carefully designed bowel program that includes laxatives, manual distention of the rectum to stimulate the intact defecation reflex, and careful attention to assistance in transfer to commode or low toilet can be helpful. A few patients may be aided by anterior sacral root stimulators. [360] High spinal cord lesions rarely can impair gastric and small bowel motility, which predisposes to gastric distention, esophageal reflux, and adynamic ileus. [361]

Other Neuromuscular Diseases

Amyotrophic lateral sclerosis (ALS) is a neurodegenerative disorder characterized by progressive loss of motor neurons, muscle atrophy, and weakness. Dysphagia ultimately will be present in all patients and is due to discoordination of the entire pharyngeal and upper esophageal swallowing mechanisms. Rapid nutritional disintegration is common, increasing the potential for respirator dependence. Studies demonstrate that placement of a percutaneous endoscopic gastrostomy (PEG) is safe, rapidly improves nutrition, and seems to improve quality of life in ALS patients. [362], [363] Delayed gastric emptying has been reported, and if present might necessitate the use of jejunal feeding.

Residual swallowing problems are common in patients who have had a stroke. The consequences are impaired nutritional intake, decreased quality of life, and a risk of aspiration pneumonia. Pharyngeal motor dysfunction and a delay in swallowing initiation are common after acute stroke, whereas secondary oropharyngeal sensory dysfunction is uncommonly seen on video swallowing studies. Vocal cord mobility is reduced, with resulting reduction in airway protection. Swallowing therapy may help maintain oral intake in some cases. [364] Commonly, a choice must be made with dysphagic stroke patients as to whether to provide nutrition via nasogastric feeding or percutaneous gastrostomy. One study compared these two methods of nutritional delivery and found that the endoscopic gastrostomy group had a lower 6-week mortality (13% vs. 57%) and a greater weight gain (77% vs. 12%) than the nasogastric feeding group. [365] Quality of life was not measured.

Myasthenia gravis is an autoimmune disorder of the neuromuscular junction that is a treatable cause of oropharyngeal dysphagia. [366] Some patients have coexisting immunodeficiency states and may have intestinal villus atrophy and increased susceptibility to enteric infections. [367] Intestinal pseudoobstruction that has occurred in patients with myasthenia gravis and thymoma has resolved with treatment of the myasthenia, suggesting that the gastrointestinal dysfunction may be a paraneoplastic syndrome associated with thymoma. [368]

Stiff-man syndrome consists of symmetrical, progressive stiffness and painful spasm of axial musculature. Dysphagia may develop, possibly related to muscle spasm of the cricopharyngeus and upper esophagus. [369] Whether epilepsy and abnormal brain discharges can cause intestinal symptoms (e.g., nausea, vomiting, pain) without overt seizures developing is controversial. [324]

NUTRITIONAL DISTURBANCES

Malnutrition

Malnutrition results from famine, poverty, poor dietary habits, and chronic disease. Kwashiorkor identifies the protein-deficient patient with an adequate intake of calories, exemplified in developed countries by the catabolic patient after trauma, infection, or burn. Typically such patients are characterized by a persistent edematous state. Marasmus syndromes are related causally to a deficiency in total caloric intake (i.e., the starved patient). [370] Diarrhea and infection are common manifestations of the terminal malnutrition. Malnutrition causes both humoral and cellular immunodeficiency, which renders the mucosa susceptible to bacterial overgrowth and infection by various pathogens, such as parasites, toxigenic bacteria, and viruses. Atrophy of the intestinal mucosa and pancreas contributes substantially to development of the diarrheal state. Hepatomegaly may develop in conjunction with fatty liver and pancreatic enzyme secretion may be reduced by over 80%. [371] In Western countries, chronic alcoholism and chronic liver disease are common causes of malnutrition. [372]

The indications for the delivery of nutrients by the enteral and parenteral route as well as the efficacy of nutritional supplementation in malnourished, hospitalized patients are unproved. [373] Recent weight loss of more than 10% of body weight, the inability to eat for 5 to 7 days in an undernourished patient or for 10 to 14 days in a well-nourished patient, and a hypercatabolic state (e.g., burn) are generally proposed criteria for initiation of nutritional supplementation, [263] especially in critically ill patients. Both parenteral and enteral formulations can provide all essential nutrients, but the enteral route is preferred whenever the gastrointestinal tract is functional. [374]

Refeeding of extremely malnourished patients should be done gradually to permit mucosal and metabolic adaptation and to avoid exacerbation of diarrhea. Many patients will need pancreatic enzyme supplementation. Too rapid refeeding with high-calorie solutions that are rich in carbohydrates can lead to death by precipitating severe hypophosphatemia, hypokalemia, and hypomagnesemia and by intensifying other metabolic disturbances. [375]

Obesity

Almost 50% of adult Americans weigh more than 10% above ideal body weight, and obesity-related conditions are estimated to contribute to more than 300,000 deaths annually. Detailed evaluation of this topic can be found in Chapter 21.

ORGAN TRANSPLANTATION AND COMPLICATIONS

Bone Marrow Transplantation

As a consequence of the induction regimen (lethal doses of radiation therapy and chemotherapy), impairment of the immune system, and graft-versus-host disease (GVHD), gastrointestinal complications are common after bone marrow transplantation. [376] It is usual for mucositis to ensue, resulting in oropharyngeal pain, nausea, vomiting, abdominal pain, diarrhea, and gastrointestinal

bleeding. These symptoms usually appear within a few days of induction and resolve after 3 to 4 weeks. Overt gastrointestinal bleeding is uncommon (7.4%) in such patients when sucralfate or histamine H2 antagonists are given prophylactically. Endoscopy usually does not define an isolated lesion and carries a 19% risk of inducing clinically relevant bacteremia. [377] Antibiotic prophylaxis should be given, and the risk and benefit of endoscopy should be considered before initiating the procedure.

A suppressed immune system predisposes the marrow transplant recipient to life-threatening infections. Bacterial and fungal infections are more common during the initial 30 days, whereas viral and parasitic infections are observed more often after day 30. [378] Oropharyngeal and esophageal thrush occur frequently. Cytomegalovirus (CMV) and herpesvirus infections are common and may involve any portion of the gastrointestinal tract. Acute diarrhea most commonly is caused by GVHD, but *C difficile* infection can cause severe, prolonged diarrhea. [379] Diagnosis depends on appropriate stool studies.

Graft-Versus-Host Disease Acute GVHD (see Chapter 129) often complicates allogeneic bone marrow transplantation and may be either acute (onset <100 days posttransplant) or chronic (onset >100 days posttransplant). In GVHD, donor cytotoxic T-lymphocytes destroy host cells in a variety of organs, including the skin, liver, and gut. [380] The earliest sign of acute GVHD is often the onset of a severe watery, secretory diarrhea. Anorexia, nausea, vomiting, jaundice, gastrointestinal bleeding, and abdominal pain may also be present. Some 30% to 50% of allogeneic bone marrow recipients develop acute GVHD, and of these 10% to 20% have either severe hemorrhage or perforation. [381] Although less common, chronic GVHD also may demonstrate gastrointestinal symptoms. The diagnosis is best made by endoscopy with biopsy, a relatively safe and simple procedure (Fig. 128-11). Tissue typically demonstrates apoptosis and gland destruction, sparse inflammatory infiltrate, and granular eosinophilic debris in dilated glands. The granular debris may be the most specific feature but is not seen commonly. [382] The use of oral beclomethasone dipropionate and prednisone are more effective than prednisone alone in treating intestinal GVHD. [383]

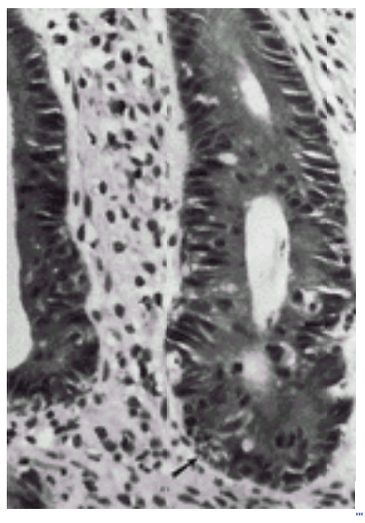

FIGURE 128-11. Rectal biopsy specimen shows crypt cell degeneration, which is the characteristic lesion of graft-versus-host disease. (Hematoxylin and eosin stain; original magnification ×100.)

Hepatic Venoocclusive Disease Liver damage secondary to cytoreductive therapy is common after bone marrow transplantation. The most prominent site is the terminal hepatic venule. Venoocclusive disease manifests by hyperbilirubinemia, hepatomegaly or right upper quadrant pain, and sudden weight gain within 20 days of transplantation without other cause. Pretransplant aminotransferase elevations, high-dose cytoreductive therapy, and persistent fever are independent risk factors. The incidence of venoocclusive disease after allogeneic bone marrow transplantation prepared with busulfan and cyclophosphamide was reduced by 60% in patients receiving ursodeoxycholic acid prophylaxis. [384] There is no clearly defined therapy for venoocclusive disease, which, when severe, is associated with multiorgan failure and death. [385]

Heart Transplantation

More than half of patients undergoing cardiac transplantation develop acute gastrointestinal problems. [386] The most common severe complications include upper gastrointestinal bleeding, pancreatitis, cholecystitis, and bowel perforation. [387] An increased risk for pancreaticobiliary disease persists for 1 year after the operation. [388] Although there is no evidence of an increased risk of colorectal cancer or adenomatous polyps in patients posttransplant, the chronic immunosuppression dictates a vigilant approach to colorectal cancer screening.

Liver Transplantation

The direct hepatobiliary consequences of liver transplantation are not unexpected and relate to performance of the transplanted liver and the patency of the created vascular and bilioenteric anastomoses. [389] The latter problems are commonly amenable to radiologic or endoscopic interventions. [390] Postoperatively, most infections are bacterial and relate to the surgery, ventilation, and intravenous lines. [391] CMV infection is common, any organ in the gastrointestinal tract can be infected, but infection may not be associated with clinical disease. Esophagitis with dysphagia, gastritis with delayed gastric emptying, [392] and colitis resulting from CMV have been recognized. Because effective therapies (e.g., ganciclovir) for CMV are available, an aggressive endoscopic diagnostic approach with multiple biopsies is warranted. Studies are being undertaken to determine whether risk-group–specific prophylactic therapy is useful. [393]

Gastrointestinal bleeding occurs in almost 10% of patients after liver transplantation, and endoscopy will reveal a source in most patients. Gastroduodenal ulcers are the most common cause, followed by enteritis, persisting portal hypertension, and bleeds at the Roux-en-Y anastomosis. [394] Acute pancreatitis is not a rare event after liver transplantation and is associated with a high mortality. [395] A strong correlation exists between the development of pancreatitis and the presence of hepatitis B, extensive surgical dissection around the pancreas, and the number of liver grafts received by the same patient. [396]

Lung Transplantation

The risks of lung transplantation are similar to those noted after other solid organ transplants; however, several reports have described a high frequency of postoperative colonic perforation. [397, 398] Because mortality is related directly to the duration of delay in diagnosis after symptoms are noted and symptoms may be blunted secondary to immunosuppression, careful evaluation for abdominal distention with plain films of the abdomen may help to guide clinical decisions. Because of the extensive mediastinal dissection required during heart-lung transplants, vagal injury can occur, leading to dysphagia and gastric stasis. Fortunately, there are few important clinical sequelae of the nerve injury. [399] Fatal hyperammonemia has been reported to occur uncommonly after lung transplantation, and is associated with hepatic glutamine synthetase deficiency. [400]

Renal Transplantation

More than 16% of patients who undergo renal transplantation develop gastrointestinal complications. [401] Whereas peptic ulcer disease with hemorrhage or perforation were common when high doses of prednisone were used, they are much less common in the cyclosporine era. More effective medical therapies exist for ulcers, making surgical intervention less common. [402] Intestinal ischemia, diverticulitis with perforation, CMV-induced colitis, intestinal tuberculosis, and intra-abdominal abscesses are increasingly common. [403] Infection with hepatitis C and B are common in dialysis patients who develop chronic renal failure. After transplantation, the immune suppression may accelerate the course of chronic hepatitis (particularly hepatitis B) and fulminant hepatitis may occur. [404]

PREGNANCY

Pregnancy is accompanied by a variety of physiological changes, many of which have been attributed to the effects of progesterone or a-human chorionic gonadotropin on gastrointestinal motility and displacement of the viscera by the gravid uterus. Common gastrointestinal symptoms of pregnancy include altered appetite, pica, ptyalism, gingivitis, vomiting, dyspepsia, heartburn, constipation, and hemorrhoids. Management of symptoms with pharmaceuticals is complicated by a paucity of clinical studies in humans demonstrating safety for the fetus. These issues are discussed in detail in Chapter 52.

Liver Disease

Although liver function is normal in pregnancy, palmar erythema and spider nevi are often observed, perhaps as a result of elevated steroid hormones. Serum alkaline phosphatase and leucine amino peptidase levels are increased, attributable to placental production. Other liver enzyme levels are normal. Jaundice is infrequent, but when it occurs it is most often the result of intrahepatic cholestasis of pregnancy (ICP). [405] Cholestasis presents in the 2nd or 3rd trimester as severe generalized pruritus that worsens at night. Marked increases in alkaline phosphatase and serum bile acids with normal ?-glutamyl transferase (?GT) are characteristic. A subgroup with raised ?GT is associated with heterozygous MDR3 mutations. [406] In Chile, ICP has been associated with a decrease in blood selenium level, suggesting environmental causes. [407] There is an increased risk of stillbirths and premature births. Ursodiol can help control the pruritus, but its effect on the outcome of pregnancy is uncertain. [408] Prophylactic vitamin K should be administered parenterally to avoid postpartum bleeding. [405]

The HELLP syndrome is a variant of severe preeclampsia characterized by hemolysis, elevated liver enzymes, and low platelets. Its features suggest a thrombotic microangiopathic process, and it must be distinguished from acute fatty liver of pregnancy, HUS, and TTP. [409] HELLP and acute fatty liver have been associated with the presence of long-chain 3-hydroxyl CoA dehydrogenase deficiency in the infants of these mothers. [410] Marked elevations of transaminases are unusual and may suggest complicating hepatic vein thrombosis or fulminant liver failure, or spontaneous intrahepatic hemorrhage and hepatic rupture with an increased mortality. [411] Despite studies suggesting high fetal and maternal mortality with the HELLP syndrome, [409], [412] other studies have noted greatly improved outcomes when interdisciplinary teams (that included a hepatologist) have managed the patients aggressively with expeditious delivery. [413], [414]

Acute fatty liver of pregnancy resembles Reye syndrome and usually occurs in young primigravida or multiparous women in the 3rd trimester. Vomiting, right upper quadrant pain, and a viral-like syndrome are common early in the course of disease. Jaundice, encephalopathy, and acute renal failure then supervene. As opposed to the HELLP syndrome hypofibrinogenemia, hypoglycemia, and hypoprothrombinemia are common. [414] Microvesicular fat accumulated in the central zone is seen on liver biopsy, but the diagnosis is based on clinical presentation and biopsy is rarely needed. When acute fatty liver is present, the pregnancy should be terminated promptly. Maternal and fetal mortality has been reduced to less than 5%, and survivors have no long-term liver disease. [413], [414]

Conception and viable pregnancy in patients with chronic liver disease is uncommon and fraught with complications. Patients with established portal hypertension have an increased risk of variceal hemorrhage during the third trimester of pregnancy with increase in both fetal and maternal mortality. Prophylactic variceal ligation with ablation of varices or transvenous intrahepatic portal shunt should be considered. [415]

PSYCHOLOGICAL DISORDERS

Anxiety and Stress

Anxiety and stress can alter gastrointestinal function and produce a broad range of gastrointestinal symptoms in otherwise healthy persons. It is not surprising, then, that patients with underlying gastrointestinal illness (e.g., inflammatory bowel disease, gastroesophageal reflux) might notice a worsening of their symptoms at those times; but this does not mean that stress is the cause of the disease. In some patients, however, emotional factors may be the cause of or may contribute to the symptoms of globus syndrome, esophageal spasm, nutcracker esophagus, nonulcer dyspepsia, and irritable bowel syndrome. [416] The close epidemiologic association between sexual and physical abuse as a child and the development of severe and refractory functional gastrointestinal illness as an adult is clear. [417] The mechanisms for the association are complex and probably relate to both psychological (somatization, reinforcement of abnormal illness behavior) and physiological (enhanced visceral sensitivity) factors. In addition, sexual abuse is a common feature of many psychiatric diagnoses (e.g., somatization disorders, depression) associated with functional bowel disorders. [418] Thus, the uniqueness of sexual abuse as an associated finding has not been established. One study has found striking hyperactivity of the hypothalamic-pituitary axis and autonomic nervous system is such patients when compared to age-matched controls. This supports the hypothesis that abnormal secretion of central nervous system corticotropin-releasing factor may mediate the association between early life stress and anxiety disorders in adults. [419]

The finding that altered emotional states increase the frequency of swallowing, and thus aerophagia, may provide insight into the common association of abdominal bloating with stressful life events. [420] Common diseases of unknown origin such as fibromyalgia or chronic fatigue syndrome are commonly associated with functional gastrointestinal complaints, such as nausea, abdominal pain, and disturbed bowel function, as well as defined gastrointestinal illnesses such as irritable bowel syndrome and hepatitis C. [421], [422] and [423] Low-dose tricyclic antidepressants can be quite helpful. The skillful and caring clinician always takes the patient's emotional state into account in the management of any patient with gastrointestinal illness.

Eating Disorders

Anorexia Nervosa Anorexia nervosa is a potentially fatal eating disorder characterized by severe voluntary weight loss, altered body image, and morbid fear of obesity that primarily affects adolescent girls. [424] Anorexia nervosa may carry a mortality of up to 9%. Gastrointestinal complaints are seen in more than half of patients, but multiple systems are involved secondary to the starvation. Delayed gastric emptying that contributes to a feeling of bloating and early satiety is common, but it usually resolves with a return to normal body weight. [425] Serum levels of trace metals such as zinc and copper are commonly depressed, and hypophosphatemia may supervene during refeeding. In severe cases, parenteral feeding may be needed. Details of evaluation and management are found in Chapter 35.

Bulimia Bulimia is a distinct syndrome characterized by cyclic binge eating, followed by self-induced vomiting, fasting, laxative abuse, or excessive exercising to promote weight loss. [424] As opposed to anorexia, severe weight loss and multisystem involvement are rare. Common gastrointestinal symptoms include bloating, flatulence, constipation, abdominal pain, borborygmi, and nausea. Binge eating causes acute gastric dilation, which can result in gastric rupture. Vomiting is associated with dental enamel erosion, parotitis, esophagitis, Mallory-Weiss tears, and esophageal rupture. [426] Although pancreatitis may occur, hyperamylasemia in patients with bulimia most often is caused by increased salivary-type amylase. Treatment is directed at the underlying psychiatric diagnosis.

Rumination Although frequently reported in children, rumination is uncommon in adults who are not developmentally disabled. It is characterized by daily effortless regurgitation of undigested food and may be associated with prominent weight loss. Screening for other eating disorders is important. Delays in diagnosis and the concomitant overuse of diagnostic tests are common. Esophageal and upper gastrointestinal studies are normal. There is no clearly effective medical therapy other than reassurance and psychiatric consultation. [427]

Münchhausen Syndrome

Münchhausen syndrome is the repeated seeking of medical care for factitious and purposefully self-generated illness. In contrast to hysteria, the symptoms are subject to "conscious" control. Patients have feigned many gastrointestinal diseases, including abdominal emergencies, hematemesis, hematochezia, and inflammatory bowel diseases. [428] Satisfactory treatment of this syndrome is rare.

PULMONARY DISORDERS

a_1-Antitrypsin Deficiency

a_1-Antitrypsin (aAT) deficiency is an autosomal recessive inherited condition that is the most common genetic cause of liver disease in children. It is also associated with chronic obstructive pulmonary disease, chronic liver disease, and hepatocellular cancer in adults. The ZZ phenotype has been associated most frequently with severe disease. [429] Adult patients can be screened effectively by serum aAT levels. Those with an aAT level below 80 mg/mL should undergo aAT phenotyping. [430] Liver disease tends to manifest late in life and is associated with inclusion bodies containing entangled polymers of mutant Z aAT within hepatocytes in the periportal areas on liver biopsy, a finding unrelated to the severity of the liver disease. [431] Liver transplantation is an effective form of therapy for patients with severe liver disease. [429]

Asthma/Chronic Cough

Asthma is a common cause of disability and death that is characterized by varying degrees of bronchial constriction. GERD is present in three fourths of asthmatic patients [432] and may be worsened by the use of asthma medications (theophylline, ß-adrenergic agonists) that lower esophageal sphincter pressure, but GERD also may substantially complicate the course of the lung disease. Worsened wheezing is thought to result from the aspiration of gastric contents, although reflex bronchial constriction also has been suggested as a mechanism. [433] Pyrosis as well as other symptoms of GERD, asthma onset in middle age, and the absence of coincident allergy or eosinophilia are clues to a possible association with GERD. When GERD-induced asthma is suspected, 24-hour ambulatory pH monitoring can confirm severe reflux. [434] Therapy of GERD in this setting usually requires prolonged potent acid inhibition with proton pump inhibitors. [435] Because of uncertainties in

demonstrating a causal relationship between asthma and GERD, antireflux surgery should be considered carefully. [436]

Chronic idiopathic cough is commonly associated with GERD. [437] Even in the absence of heartburn, acid may trigger nocturnal cough or laryngospasm. Laryngospasm may be mistaken for asthma unless appropriate pulmonary function tests are employed. Evaluation and management are the same as for asthma.

Consequences of Mechanical Ventilation and Respiratory Failure

Respiratory failure and mechanical ventilation may be complicated by gastrointestinal bleeding from erosive gastritis and ulcer disease. Associated coagulopathy further increases the risk of overt hemorrhage. It is important to emphasize the infrequency of this complication when it is monitored prospectively in large populations. [438] This said, giving antacids, H2 antagonists, and barrier-enhancing drugs prophylactically to critically ill patients reduces the incidence of stress ulcerations [439] and should be provided to ventilated patients. It has been suggested, however, that raising the gastric pH might permit colonization of the stomach by pathogenic bacteria, which, in turn, may predispose to nosocomial pneumonia. This is controversial, and careful attention to a general preventive strategy is likely to be more helpful than arbitrarily switching to sucralfate. [440] Administering sucralfate alone through a nasogastric tube may expose the patient to an increased risk of erosive esophagitis associated with a nasogastric tube because gastric pH remains low. [441]

Acute exacerbation of chronic respiratory failure is a common cause of ischemic hepatitis. In contrast to the severe hypertransaminitis seen after circulatory arrest, where severe hypotension is the proximate cause, in patients with acute respiratory compromise superimposed on chronic respiratory failure, the cardiac index is increased. The cause of the hepatic damage is a combination of severe arterial hypoxemia and elevated central venous pressure. [442] This has also been noted in a patient with severe obstructive sleep apnea. [443]

Cystic Fibrosis

Cystic fibrosis (CF) is the most common inherited disease of Caucasians in North America, with an incidence of about 1 per 2000. Mutations in the *CF* transmembrane conductance regulator gene [444] at the long arm of chromosome 7 lead to an underlying defect in mucosal cAMP ion and water transport. A net shift in fluid from the lumen results in viscous secretions and duct obstruction in the respiratory tract, salivary glands, pancreas, intestinal lumen, and biliary tree. [445]

The presence of viscous secretions in the intestinal lumen has important clinical consequences. Affected infants may present with meconium ileus, intussusception, distal intestinal obstruction syndrome (DIOS), intestinal atresia, volvulus, and perforation. [445] Chronic constipation is common and may lead to rectal prolapse. Mineral oil or acetylcysteine may be effective. [446] Recurrent use of gastrointestinal lavage with Golytely also may be helpful in reducing the symptoms of bloating and distention that characterize DIOS. [447]

Symptomatic gastroesophageal reflux is common in children who have CF. Contributing factors are decreased saliva production, gastrointestinal hypomotility, chronic cough, and the need for postural drainage of pulmonary secretions. [448] Standard antireflux therapy is usually effective. As more patients survive to adulthood, complications of GERD, such as stricture and Barrett esophagus, [449] will be seen with greater frequency.

Essentially all patients have severe nutritional impairment caused by pancreatic insufficiency and manifested by steatorrhea and malabsorption of nonfat nutrients. Protein-calorie malnutrition is aggravated by the catabolic state induced by chronic respiratory tract infections but also may contribute to worsening lung disease. Improvement of exercise tolerance and pulmonary function can result from enteral/parenteral nutritional support, pancreatic enzyme replacement, and fat-soluble vitamin supplementation. [450], [451] The use of large doses of high-potency pancreatic enzymes has caused a fibrosing colitis manifested by bloody diarrhea associated with diffuse narrowing of the colon with rectal sparing. [452] As survival to adulthood becomes more common, new problems are recognized. Symptomatic osteomalacia is a common sequela of chronic steatorrhea, 25-hydroxy vitamin D levels should be monitored, and supplemental calcium and vitamin D should be provided. [453] A greater than expected incidence of malignancies of the digestive tract has been found. [454] Thus persistent or unexplained gastrointestinal symptoms should be investigated carefully.

Focal biliary cirrhosis is common and asymptomatic, but about 10% of children with CF develop confluent biliary cirrhosis and progressive portal hypertension. [455] If variceal bleeding occurs, banding or portosystemic shunting would be the therapeutic modalities of choice. Ursodiol improves liver function tests, but it is unclear whether fibrosis is retarded. Improvements in surgical techniques have allowed end-stage cirrhotics with CF to undergo liver or combined liver and lung transplantation successfully. [455]

RENAL DISORDERS

Chronic Renal Failure

Chronic renal failure is associated with a number of gastrointestinal disturbances. Dysgeusia, a metallic taste in the mouth, and fetor uremia are often noted by uremic patients. Parotitis and sicca syndrome occur commonly in uremic patients, further limiting compliance with fluid restriction and impairing nutrition.

Renal failure commonly is associated with anorexia, nausea, vomiting, and epigastric pain. Endoscopy should be done to exclude ulcer or gastritis. Because uremia is associated with gastric emptying abnormalities, [456] however, symptoms may be alleviated by using agents such as metoclopramide. Gastrointestinal bleeding has multiple possible causes, [457] but in contrast to patients with normal renal function, arteriovenous malformations are particularly prevalent. Estrogen therapy may be helpful. [458] Peptic ulcer disease, gastritis, esophagitis, and duodenitis may be managed with H2 antagonists or antacids, with lower doses needed to prevent drug accumulation and magnesium antacids avoided to prevent hypermagnesemia. The diagnosis of *Helicobacter pylori* is difficult in uremics and relies on histological methods rather than the breath test. [459] Duodenal pseudomelanosis is an idiopathic condition in which pigmentation develops in the proximal duodenal mucosa (Fig. 128-12; see also Color Fig. 128-12). This syndrome has no known symptoms attributable to it.

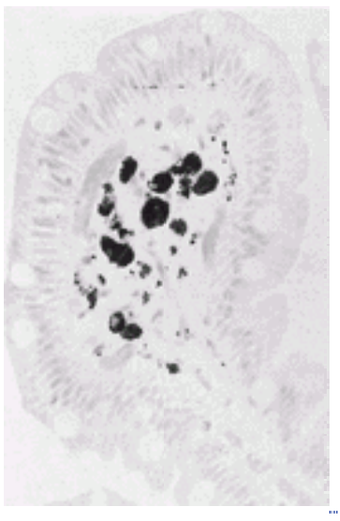

FIGURE 128-12. (See Color Fig. 128-12.) Duodenal biopsy specimen from a patient with pseudomelanosis duodeni shows the deposition of pigment within lamina propria macrophages.

Uremic patients may develop abdominal pain, constipation, pseudoobstruction, and intussusception. Important factors in the development of constipation include oral fluid restriction and the use of antacids that contain aluminum hydroxide. Diarrhea secondary to a variety of causes is common. Fecal impaction must be excluded. Although conditions such as bacterial overgrowth and pseudomembranous colitis must be considered, the cause of many cases remains idiopathic. The incidence of pancreatitis in chronic renal failure is unknown. [456] Elevation of serum amylase is common but should not be attributed necessarily to impaired renal clearance, especially if the level exceeds twice the upper limit of normal. [460]

Uremic patients can develop hepatic friction rubs without other manifestations of liver disease. Elevated alkaline phosphatase in such patients can be of intestinal origin and may not be indicative of

bone disease or cholestasis. Chronic hepatitis from hepatitis B and C is common, and may complicate the ability to provide effective dialysis and kidney transplantation. [461] Treatment with lamivudine for hepatitis B and interferon/ribavirin for hepatitis C can be undertaken with caution in such patients.

Hemodialysis

Inability to gain weight and diarrhea may be manifestations of inadequate dialysis. A few patients on long-term hemodialysis can develop severe obstipation, colonic pseudoobstruction, and spontaneous perforation of the colon. [462] Serum lipase levels can increase significantly after heparin administration and thus have reduced specificity for the diagnosis of pancreatitis in this setting. [463] Idiopathic refractory exudative ascites occurs in a few patients, is difficult to treat medically, and resolves with kidney transplantation. [464]

Peritoneal Dialysis Patients undergoing continuous ambulatory peritoneal dialysis are commonly subject to an easily treatable form of bacterial peritonitis. Differentiating this condition from peritonitis associated with important intra-abdominal pathology (bowel perforation, ischemic bowel, appendicitis) can be difficult because polymicrobial infections can occur without important gastrointestinal pathology, [465] and pneumoperitoneum is common after dialysis. Pneumoperitoneum not associated with perforated viscus was found in 4% of one series, and may take weeks to resolve. [466]

Polycystic Kidney Disease

Polycystic kidney disease (PKD) is an autosomal dominant disorder whose abnormal genes (PKD1, PKD2) have been localized to chromosome 16. PKD accounts for about 10% of the chronic dialysis population. [467] Hepatic cysts are an important manifestation of PKD. The frequency of coincident hepatic cysts increases with age, and the large cysts are more common in women who have had multiple pregnancies. [468] Despite large cysts, hepatic parenchymal volume is preserved, as is liver function. [469] Hepatic cysts can become infected, large cysts can obstruct the biliary tree, and early intervention with both antibiotics and percutaneous drainage is needed. [470] An association with congenital hepatic fibrosis has been reported. [471]

SUBSTANCE ABUSE

Alcohol

Alcohol consumption can enhance LES pressure, reduce the frequency of esophageal peristaltic contractions, and delay esophageal emptying. High-amplitude peristaltic contractions also have been noted (nutcracker esophagus). [472] These alterations in function may result in chest pain and esophagitis. Chronic alcohol ingestion increases the risk of esophageal cancer.

Alcohol produces acute and chronic gastritis by disruption of the gastric mucosal barrier. Binge drinking can result in gastric erosions and severe hemorrhage. Chronic use, however, seems to protect against development of serious mucosal injury. Epidemiologic studies suggest that the concomitant use of alcohol and NSAIDs [473] or alcohol and warfarin [474] potentiated the risks of severe GI bleeding or perforation from the use of either alone.

Alcoholism may result in diarrhea and malabsorption. Alcohol enhances small intestinal transit, decreases brush border enzyme activity, and impairs absorption of nutrients. Acute exposure of the small bowel to alcohol causes altered mucosal and microvascular permeability. [475] Chronic pancreatitis, the spectrum of chronic alcoholic liver disease, protein-calorie malnutrition, and vitamin deficiencies are contributing factors (Chapter 95).

Amphetamines

The social use of ecstasy (3,4-methylenedioxymethamphetamine, MDMA) and amphetamines has become widespread in the United States. They induce euphoria, intensify emotions, and have become a staple of the “rave” culture. In excess the use of these drugs causes anxiety, hallucinations, chest pain, palpitations, coma, subarachnoid hemorrhage, and death. [476] A panarteritis that is similar to polyarteritis nodosum (but negative for antineutrophil cytoplasmic antibody, or ANCA), with associated mesenteric ischemia, is reported. [477] As with cocaine hepatotoxicity is common, caused by both immune-mediated mechanisms and hyperthermic liver injury, and therapy for severe cases is largely supportive. [478]

Cocaine

The use of cocaine and related metabolites is widespread. Anorexia and diarrhea are common. Cocaine use can result in gastric ulcerations with perforation, [479] retroperitoneal fibrosis, and intestinal ischemia with infarction. [480] Severe liver injury is almost always associated with rhabdomyolysis and hyperthermia. The zone of hepatic necrosis will vary depending on other drugs being used. [481] The practice of smuggling cocaine in condoms concealed in the digestive system can result in intestinal obstruction and death.

Narcotics

Narcotic administration for abuse or for pain relief frequently results in physical dependency, with resulting physical symptoms on withdrawal. Gastrointestinal symptoms of withdrawal include anorexia, nausea, vomiting, and abdominal pain. These symptoms may be difficult to distinguish from structural gastrointestinal disorders. Narcotics slow colonic peristalsis and thus frequently induce constipation that can be a long-standing problem. [482] Clonidine therapy may ameliorate the symptoms of narcotic bowel syndrome.

Parenteral administration of illicit drugs can result in chronic viral hepatitis, cirrhosis, and hepatic abscess. Injected contaminants such as talc can cause hepatic granulomas.

VASCULITIDES

Behçet Disease

Behçet disease is a systemic illness of unknown etiology characterized by necrotizing vasculitis and most commonly described in people of Middle Eastern and Japanese extraction. Common manifestations include orogenital ulcerations, eye inflammation, skin lesions, arthritis, and migratory thrombophlebitis. Gastrointestinal tract involvement is uncommon. [483] A curious and virtually pathognomonic sign is the development of pustules at the site of needle punctures.

Oral lesions resemble aphthous ulcers and may be single or multiple. Esophageal ulcerations are common, but infrequently cause symptoms. [484] The most frequent extra-oral sites of gastrointestinal involvement are segmental inflammation and ulceration of the ileocecal region and colon, resembling Crohn’s disease. Intestinal bleeding or perforation may result. Therapy is similar to that of Crohn’s disease, with immunomodulating medications often ameliorating the disease. Resection of severely diseased segments of the gut may be required, but postoperative recurrence is common. [485]

Other than symptomatic Budd-Chiari syndrome, involvement of the liver and spleen are rare. Typically, such patients have cavernous transformation of the portal vein, associated with portal vein, inferior venal caval, and hepatic venous thrombosis. Definable coagulopathies have not been defined as the cause of the thrombosis, suggesting the primary importance of vasculitis. Once established the presence of Budd-Chiari in the patient with Behçet disease indicates a poor prognosis. [486], [487]

Churg-Strauss Syndrome

Churg-Strauss syndrome is a systemic vasculitis characterized by the presence of asthma, hypereosinophilia, and necrosis with extravascular eosinophil granulomas. Common symptoms other than asthma are weight loss, fever, myalgias, arthralgias, and vasculitic skin lesions. Small to medium-size vessels are involved, and ANCAs are commonly detected. Corticosteroids alone or in combination with cyclophosphamide will usually induce clinical remission, but recurrence is common. [488], [489]

Gastrointestinal manifestations are common, and when present are associated with a bad prognosis. [488] They include nausea, vomiting, abdominal pain, gastrointestinal hemorrhage, perforation, and cholecystitis, are related to mesenteric vasculitis, and are similar to those found in polyarteritis nodosa. Mesenteric ischemia with ulceration and perforation is reported. On occasion a case may be diagnosed with endoscopic biopsy. [490]

Cryoglobulinemia

Cryoglobulinemic vasculitis is caused by the localization immune complexes of mixed cryoglobulins in the walls of venules, capillaries, and arterioles, which incites acute inflammation. Mixed cryoglobulins, rheumatoid factor, and low levels of C_4 are typically found in the serum. The most frequent clinical manifestations are purpura, arthralgias, and nephritis (the main cause of morbidity). Cryoglobulinemia occurs in a variety of clinical settings, such as lymphoproliferative disorders, infections, and autoimmune diseases, or as an idiopathic process. However, cryoglobulinemic vasculitis is always associated with chronic hepatitis C. [488]

Gastrointestinal illness, such as recurrent episodes of crampy abdominal pain, enterocolitis, and, rarely, small or large bowel ischemia have been reported. It is likely that effective treatment of the hepatitis C infection with interferon-ribavirin will abolish the vasculitis. [491]

Giant Cell Arteritis

Giant cell arteritis is a granulomatous disease that usually involves large-sized arteries, including the aorta and the temporal arteries. The disease usually occurs in older adults and is associated with the clinical syndrome of polymyalgia rheumatica. Disseminated fibrinoid necrosis of smaller vessels can result in visceral ischemia, abdominal pain, nausea, anorexia, weight loss, bleeding, and perforation. Disturbances of liver function are common and have been attributed to hepatic artery involvement [492] and hepatitis associated with fibrin-ring granulomas. [493]

Henoch-Schönlein Purpura

Henoch-Schönlein purpura is a vasculitis syndrome with a peak age of onset in childhood, characterized by systemic deposition of immunoglobulin A (IgA)-dominant immune complexes in small vessels. The classic triad of symptoms includes a palpable purpuric rash, abdominal cramping, and hematuria typically begins after an upper respiratory infection. [488] ANCA is negative. Most children have a self-limited disease without chronic sequelae. Any part of the gastrointestinal tract may be affected. Gastrointestinal symptoms include abdominal pain, vomiting, diarrhea, intestinal obstruction, or intussusception and can precede the onset of the characteristic purpuric rash. [494] The bowel may be edematous and show evidence of submucosal or subserosal hemorrhage. Radiographic findings include thickening of small bowel folds (Fig. 128-13), colonic wall edema, and intussusception. Whether corticosteroids actually improve outcomes is controversial. [495]

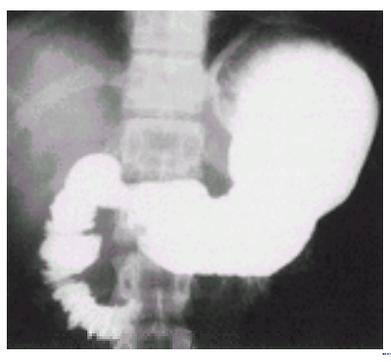

FIGURE 128-13. Henoch-Schönlein purpura. Thickened folds in the second part of the duodenum.

Polyarteritis Nodosa

Polyarteritis nodosa (PAN) is a systemic necrotizing vasculitis that affects small and medium-size arteries of nearly every organ system, giving rise to a wide spectrum of clinical findings. Recently individualized from PAN, microscopic polyangiitis is an ANCA-positive systemic vasculitis of small-size vessels smaller than arteries characterized by rapidly progressive glomerulonephritis and pulmonary involvement that is usually absent in PAN. The absence of necrotizing granulomatous inflammation separates this entity from Wegener granulomatosis and Churg-Strauss syndrome. The gastrointestinal manifestations of the two disorders is similar. [488]

Many cases of PAN are associated with hepatitis B infection. [496] Fever, weight loss, arthralgias, subcutaneous nodules, hypertension, and peripheral neuropathy are common. Gastrointestinal complaints are present in about one third of patients, usually secondary to visceral ischemia. PAN is the most common cause of localized vasculitis of the gastrointestinal tract. [497] The most common clinical manifestations are epigastric pain, nausea, anorexia, mucosal ulceration, intestinal bleeding, and diarrhea. [498] Appendicitis, cholecystitis, pancreatitis, [499] cataclysmic intra-abdominal hemorrhage, bowel obstruction, and liver infarction have been reported. Typical aneurysmal dilation of small and medium-sized arteries on arteriogram (Fig. 128-14) may provide a diagnosis. Corticosteroid and cyclophosphamide therapy have improved the overall survival of these patients dramatically. [500]

FIGURE 128-14. Polyarteritis nodosa. Typical aneurysmal dilation of small and medium-size arteries.

Wegener Granulomatosis

Wegener granulomatosis is an ANCA-positive, small vessel vasculitis characterized by granulomatous inflammation involving the respiratory tract and necrotizing vasculitis of small to medium-size vessels. Glomerulonephritis is common. Manifestations of upper airway disease are sinus pain and purulent drainage, nasal mucosal ulceration, and otitis media. Tracheal inflammation causes stridor and may lead to dangerous airway compromise. Chest x-ray commonly shows nodular radiographic densities, and unlike Churg-Strauss syndrome there is no asthma. [488]

Odynophagia secondary to esophageal vasculitis manifesting as punched out ulcers in the esophagus has been reported. [501] Granulomatous inflammation of the stomach may be mistaken for Crohn's disease. [502] The associated vasculitis may result uncommonly in intestinal or colonic ischemia, bleeding, and perforation. [503]

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CHAPTER 129

Fergus Shanahan

GASTROINTESTINAL MANIFESTATIONS OF IMMUNOLOGIC DISORDERS

IMMUNODEFICIENCY DISEASES AND THE GUT

The gut epithelium represents the largest surface area where the host interacts with the environment, separating the lumen from the internal milieu. This structure is vulnerable to access by infectious material. Strategic mucosal defense requires a precisely regulated mucosal immune system supported by a variety of nonimmunologic protective factors, such as gastric acidity, digestive enzymes, peristalsis, mucus secretion, epithelial cell regeneration, and the mutually competitive interactions of the gut flora. [1], [2]

The importance of effective mucosal defense is underscored by the frequency of mucosal infections. Mucosal infections are a major cause of mortality in children under 5 years of age; *Helicobacter pylori* is the most common infection in humans; and diarrheal infectious disease alone accounts for up to 5 million deaths among children worldwide annually. A wide spectrum of opportunistic infections occurs in patients with immune deficiencies. Patients with milder, more selective forms of immune deficiency (e.g., immunoglobulin A [IgA] deficiency), however, are frequently free of infectious complications. The nonimmunologic mucosal defenses as well as the functional reserve of the immune system, probably account for the resistance of these patients to infection. In addition immune deficiencies may predispose to autoimmunity and opportunistic cancers.

CLASSIFICATION OF IMMUNODEFICIENCY DISORDERS

Immune deficiency disorders are categorized into primary and secondary groups. The primary immune deficiencies may be congenital or acquired and result from intrinsic defects in the cellular components of the immune system or their secretory products. In many cases the specific metabolic defect or alteration in gene regulation results in aberrant signal transduction within lymphocytes. [3] Approximately 100 inherited immunodeficiency disorders have been identified; [4] those of particular gastroenterologic importance are discussed in this chapter. For convenience, this heterogeneous group of conditions will be discussed under the following headings: Predominantly B-Cell (Antibody) Defects, Predominantly T-Lymphocyte Defects, Combined B- and T-Lymphocyte Defects, Phagocytic Cell Defects, and Complement Deficiency.

Secondary immunodeficiencies are far more common than the primary disorders. Causes include protein-calorie malnutrition, protein-losing enteropathy, cancer, immune senescence with aging, and the increasingly recognized iatrogenic immunodeficiencies associated with organ transplantation and immunosuppressive agents. The most important secondary immunodeficiency disorder is acquired immunodeficiency syndrome (AIDS), which is discussed separately in Chapter 124.

Predominantly B-Lymphocyte (Antibody) Defects

X-Linked (Congenital or Bruton) Hypogammaglobulinemia This is the prototypic syndrome of a pure B-cell or humoral immunodeficiency. [2] It occurs in males, although rarely in females. The genetic defect results from mutations in the cytoplasmic signal transduction molecule, Bruton tyrosine kinase (Btk), although why the *Btk* gene defect results in arrest of B-cell development is unresolved. [5] The basis for genetic, clinical, and immunologic heterogeneity observed in this condition is unclear; additional genetic and environmental factors may determine the clinical phenotype. Serum levels of all immunoglobulins are less than 100 mg/dL, and there is an inability to make functional serum antibodies. An intrinsic B-cell defect with a maturation block in pre–B-cell to B-cell differentiation is present. [5] The pre–B cells fail to express functional immunoglobulin M (IgM) on the cell surface because of an inability to rearrange immunoglobulin light-chain genes. Pre–B cells are present in bone marrow in normal numbers, but circulating B cells are absent and peripheral lymphoid tissue is hypoplastic with rare or absent plasma cells. The thymus and T-cell function are normal. Patients usually present with recurrent pyogenic infection during infancy (after disappearance of maternal immunoglobulin G [IgG]) or in early childhood. The respiratory and gastrointestinal tracts are the most frequent sites of infection. [6], [7] and [8] Gastrointestinal infections occur in 30% of patients. [6] These are usually infectious diarrheal episodes commonly caused by *Campylobacter* species. Giardiasis can cause severe mucosal damage and steatorrhea in these patients, but it is responsive to therapy and is surprisingly uncommon. Perirectal abscess formation and small-bowel bacterial overgrowth also may occur. In asymptomatic patients, rectal biopsy characteristically reveals an absence of plasma cells and a mild lamina propria neutrophil infiltrate with early crypt abscess formation. [8] This may lead to an erroneous diagnosis of chronic colitis. Although cellular immunity is normal, these patients are susceptible to certain virus infections, notably hepatitis and enteroviral infections including chronic meningoencephalitis and poliomyelitis. [5] There is an increased risk of malignancy, particularly lymphomas and leukemias. Treatment consists of parenteral immunoglobulin replacement. [9]

Selective Immunoglobulin A (IgA) Deficiency Selective IgA deficiency is the most common primary immune deficiency with a variable prevalence in different ethnic groups, reported as high as 1 per 500 population in the Western world (range 1/500–1/3000). [10], [11] Its occurrence is usually sporadic, but familial cases have been reported. Most patients lack both serum and secretory IgA1 and IgA2. The basic defect is not known, and there may be etiologic heterogeneity as well as clinical heterogeneity. Occasionally IgA deficiency may be transient, and a large number of drugs including phenytoin, penicillamine, and sulfasalazine have been linked with reversible deficiency. [10] Although IgA deficiency is associated with a wide range of conditions, [10], [11] and [12] mainly recurrent infections of mucosal tissues and autoimmune disorders (Table 129-1), the majority of IgA-deficient individuals are asymptomatic and are free of a complicating disease. The associations with celiac disease and pernicious anemia may be due in part to a common linkage of IgA and such diseases with HLA-B8/DR3. In one case, IgA deficiency with malabsorption and villous atrophy was associated with an IgG antibody to intestinal epithelial cells and a favorable response to cyclophosphamide. [13]

Gastrointestinal manifestations and associations:
None (usually)
Infections, especially giardiasis (may not be any more common than in general population)
Gluten-sensitive enteropathy (IgA prevalence in celiac disease is 1:50)
Pernicious anemia
Vitamin B_{12} deficiency secondary to bacterial overgrowth
Intrinsic factor deficiency
Nodular lymphoid hyperplasia
Food allergy (increased serum antibodies to food antigens but clinical disease apparently rare)
Crohn's disease (IgA deficiency prevalence reported to be 1:73)
Disaccharidase deficiencies (unproven association)
Extraintestinal manifestations and associations of IgA deficiency:
Non–organ-specific autoimmune disorders (e.g., collagen vascular diseases)
Atopy
Malignancy (lymphomas, carcinoma reported but extent of risk is unclear and probably low)
Risk of anaphylaxis if given IgA-containing solutions, including blood

TABLE 129-1 Disorders That May Occur in Association With Immunoglobulin A (IgA) Deficiency

When infections occur in IgA deficiency, they are usually recurrent bacterial and viral sinopulmonary disorders; the gastrointestinal tract is seldom involved. Giardiasis occurs in IgA-deficient subjects, but unlike pan-hypogammaglobulinemia, its frequency is probably the same as in the general population. When persistent or recurrent infections occur, it suggests associated immunologic defects, such as deficiencies of IgG subclasses, particularly IgG2 and IgG4, which have been found in some patients with IgA deficiency. [10] Jejunal biopsy is usually morphologically normal in selective IgA deficiency, with an absence or paucity of IgA-producing cells and an increase in IgM-secreting cells. This may be an important compensatory response because, like IgA, IgM can bind to a secretory component. Most patients with IgA deficiency require no treatment. Replacement with serum IgA is futile because only locally produced IgA is transported into the lumen. Parenteral administration of IgA-containing blood products may be contraindicated because of the risk of anaphylactic reactions due to anti-IgA antibodies. Whether patients should be screened routinely for IgA antibodies is unclear, as the titer of antibody is not well correlated with risk of anaphylaxis to blood products. Prophylactic IgG therapy for IgA deficiency is either hazardous or ineffective, although some clinicians have reported a beneficial effect. [14] Patients with recurring infections should be screened for IgG subclass deficiency, but can usually be managed with prophylactic or periodic antibiotics without requiring immunoglobulin replacement. [10]

Secretory Component Deficiency Secretory component (SC) is a glycoprotein receptor on the basolateral surface of the mucosal epithelial cell that is essential for the transepithelial delivery of IgA and IgM from the lamina propria to the lumen. Deficiency of SC is very rare, and affects not only lumenal IgA levels but also any compensatory effects of IgM. In isolated case reports, it has been associated with intestinal candidiasis and diarrhea. [15], [16] Administration of IgA-rich bovine colostrum may result in symptomatic improvement. [15]

Common Variable Hypogammaglobulinemia ("Acquired" or Late-Onset Hypogammaglobulinemia) Common variable hypogammaglobulinemia is second to selective IgA deficiency as the most common primary immunodeficiency in adults and is the most common symptomatic primary immunodeficiency. Its variable phenotype and immunologic heterogeneity have hindered identification of the underlying defect(s). It comprises a group of disorders in which there is defective terminal differentiation of B-lymphocytes in the majority of patients. [10], [17] An intrinsic defect within B cells has not been demonstrated yet, and the fundamental defect(s) might reside within T cells or an anomaly of T- and B-cell communication, but not from excessive T-cell suppressor activity. Most cases are sporadic, but the condition can be familial. Clinically, common variable hypogammaglobulinemia may be similar to X-linked (Bruton) agammaglobulinemia, but differences include the later age of onset, less severe infections, the fact that the lymphoid tissue (tonsils, lymph nodes, and spleen) may be normal or enlarged, and the association with autoimmune and granulomatous disease in some patients. [17], [18] The chronic granulomatous disorder resembles the noncaseating lesions of sarcoidosis but is distinct from chronic granulomatous disease associated with defects in neutrophil function (vide infra). Patients present usually in the second or third decade with either recurrent respiratory tract infections (most commonly pneumococci, staphylococci, and *Haemophilus influenzae*) or diarrhea and steatorrhea [10], [17], [18], [19] and [20] (Table 129-2). Up to 60% of patients have chronic recurrent diarrhea; two thirds of these have malabsorption. Enteroviral infection of the central nervous system is rare. Giardiasis is a particularly common cause of symptoms in common variable hypogammaglobulinemia, but unlike in normal individuals, it may be persistent and lead to extensive mucosal damage with steatorrhea and malabsorption. [21] It is usually reversible with appropriate anti- *Giardia* therapy. Other parasitic infections, such as cryptosporidiosis and strongyloidiasis, occur less commonly. Patients with common variable hypogammaglobulinemia are susceptible to bacterial enteric pathogens including *Shigella, Salmonella,* and *Campylobacter* species (*C jejuni* and *C fetus*), which occasionally may mimic ulcerative colitis. Bacterial overgrowth with anaerobes has been described. [18]

Gastrointestinal:
Giardia lamblia infection
Small bowel bacterial overgrowth
Viral gastroenteritis (e.g., rotavirus)
Infectious diarrheas (Campylobacter, Salmonella, Shigella)
Secondary disaccharidase deficiency
Pernicious anemia, atrophic gastritis, gastric carcinoma
Gluten-sensitive sprue
Sprue refractory to gluten-free diet
Nodular lymphoid hyperplasia
Extraintestinal:
Increased incidence of generalized lymphomas
Increased incidence of autoimmune diseases
Recurrent respiratory tract infections
Chronic granulomatous disorder

TABLE 129-2 Disorders That May Occur in Association With Common Variable Hypogammaglobulinemia

In some patients, sprue-like changes may be found on intestinal biopsy, and giardiasis should always be ruled out. True gluten-sensitive enteropathy has not been well documented in common variable hypogammaglobulinemia. Jejunal biopsy generally shows a paucity of plasma cells in the lamina propria, unlike sprue. When the mucosa is flat, an enteropathy refractory to a gluten-free diet is likely. Other intestinal inflammatory disorders that have been reported in common variable hypogammaglobulinemia include Crohn's disease and idiopathic ulcerative jejunitis. [7], [9], [20] In one third of patients with common variable hypogammaglobulinemia, atrophic gastritis and pernicious anemia develop, with a markedly increased risk of gastric cancer. [7], [18] Differences from classic pernicious anemia include the absence of mucosal plasma cells, lack of autoantibodies, involvement of the entire gastric mucosa, and normal rather than elevated serum gastrin levels. Defective gastrin release in response to bombesin or food has been reported as a highly specific means of distinguishing common variable hypogammaglobulinemia from other forms of hypogammaglobulinemia. [22] Chronic liver disease has occurred in 10% to 15% of patients with common variable hypogammaglobulinemia [7], [20] due to hepatitis C or another non-A, non-B hepatitis virus acquired from prior plasma infusion; it has not been a problem with the use of the licensed intravenous immunoglobulin preparations. [20] Cholelithiasis is a frequent feature of common variable hypogammaglobulinemia. [19] Other potential causes of hepatobiliary disease include autoimmune hepatitis, sclerosing cholangitis, and biliary cryptosporidiosis. Nodular lymphoid hyperplasia is frequently found in common variable hypogammaglobulinemia, but it does not occur in X-linked hypogammaglobulinemia and is rare in selective IgA deficiency. The lymphoid nodules usually involve the small intestine but may affect the colon, rectum, and stomach. The nodules consist of large lymphoid follicles with germinal centers within the lamina propria. Plasma cells are either absent or markedly diminished. The lymphoid hyperplasia is thought to reflect B cells unable to undergo full differentiation to immunoglobulin-screening plasma cells. [23] Localized forms of nodular lymphoid hyperplasia may occur in apparently immunocompetent individuals, particularly in the large bowel. In children and adolescents, a self-limited lymphoid hyperplasia in the terminal ileum is a frequent radiologic finding. Although there is an increased incidence of lymphomas in common variable hypogammaglobulinemia, [24] intestinal nodular lymphoid hyperplasia per se does not appear to be premalignant. Almost all patients with common variable hypogammaglobulinemia require parenteral immunoglobulin replacement therapy.

Miscellaneous B-Cell Defects A variety of other syndromes associated with immunoglobulin abnormalities have been reviewed. [3], [4] Gastrointestinal manifestations may occur but are seldom a dominant component of the clinical picture.

Predominantly T-Lymphocyte Defects

Congenital Thymic Hypoplasia (DiGeorge Syndrome) DiGeorge syndrome, the most common contiguous-gene deletion syndrome in humans, is associated with defective formation of the third and fourth pharyngeal pouches during embryogenesis. It is characterized by absent T-lymphocyte function, hypoparathyroidism, and cardiovascular abnormalities, particularly of the aortic arch. The syndrome is due to deletions in the short arm of chromosome 22, and several studies have implicated the transcription factor TBX1 as a key candidate gene, although other genes have a contributory role. [25] Presentation is usually with neonatal tetany or seizures. There may be unusual facies. Other structures developing at the same stage of embryogenesis also may be affected; for example, esophageal atresia may occur. Other gastrointestinal manifestations include candidiasis, chronic diarrhea, and partial villous atrophy with malabsorption. [7]

Chronic Mucocutaneous Candidiasis Chronic mucocutaneous candidiasis (CMCC) represents a spectrum of clinical syndromes in which there is an increased susceptibility to chronic *Candida* infections of the mucosa, skin, and nails. [26], [27] and [28] Most patients become infected with other organisms in addition to *Candida* species. There is variability in mode of inheritance (autosomal recessive and sporadic), age of onset (early or late), and frequency of association with autoimmune disorders such as pernicious anemia and endocrinopathies, including Addison disease, diabetes mellitus, hypothyroidism, and hypoparathyroidism. The latter subset

of patients comprise the autoimmune polyglandular syndrome type 1 (APS1) or autoimmune polyendocrinopathy-candidiasis-ectodermal dystrophy (APECED), a recessive disorder due to mutations in the *AIRE* (autoimmune regulator) gene.[29] Most patients develop infections before the age of 5 years, but some may have the onset of *Candida* infections in adulthood. *Candida* infection most commonly involves the oropharyngeal mucosa, but involvement of the esophagus also may occur and may be present in the absence of oral lesions. Esophageal strictures, nutritional deficits, and the endocrinopathies account for most of the morbidity and mortality associated with the disease. More commonly the skin lesion is a sharply demarcated scaly erythematous macular rash; less commonly it may be granulomatous. Nails of the hands and toes are frequently involved. The *Candida* infection is usually superficial, involving the epidermis, but does not penetrate the dermis or become systemic. A variety of defects in T-cell function have been described. Autoantibodies may precede the development of endocrinopathies. The development of endocrine abnormalities is often insidious, and it is important to evaluate patients regularly for endocrine function. CMCC does not respond well to topical antifungal therapy; systemic agents are usually required. Immunologic reconstitution with therapeutic benefit has been achieved with transfer factor alone or in combination with fetal thymus transplantation.

Miscellaneous T-Cell Defects The molecular basis of several primary disorders of T-lymphocytes has been recognized.[3,30] These rare immunodeficiencies have revealed that although one phenotype may be the result of several gene mutations, a single gene mutation may lead to multiple phenotypes because of the interaction of environmental factors and modifier genes. In many cases, the defects reside in the signaling or transduction pathways for T-lymphocyte activation. Intractable diarrhea caused by diffuse inflammatory enteropathy similar to that seen in genetically engineered mice lacking cytokines such as interleukin-2 (IL-2) and IL-10 is common in disorders associated with T-cell dysfunction.

Combined B- and T-Lymphocyte Defects

Severe Combined Immunodeficiency Syndromes Severe combined immunodeficiency (SCID) is the most severe form of primary immunodeficiency and represents a syndrome caused by several distinct genetic defects, characterized by profound deficiency of T- and B-lymphocyte function and sometimes natural killer (NK) cells.[3,31] At least seven different forms of SCID in humans are now recognized, and the responsible genetic defect has been identified in some of these.[3] The most common form of SCID (60% of cases) is X-linked.[3] It is caused by defective cytokine signaling due to a mutation of the common cytokine receptor ?-chain (?c), a component of the receptors for IL-2, IL-4, IL-7, IL-9, and IL-15. The remainder of SCID cases are autosomal recessive, most of them due to deficiencies of purine-degradation enzymes, such as adenosine deaminase (ADA) and nucleoside phosphorylase. Mutations in the gene for Janus kinase 3 (*Jak3*) which transduces the receptor signal from the common ?c, also cause autosomal recessive SCID, as do mutations in the gene encoding the a-chain of the IL-7 receptor. Another variant of SCID is reticular dysgenesis, in which there is a coexisting deficiency of granulocytes. Clinically, the syndrome presents in early infancy with severe life-threatening infections, chronic diarrhea, malabsorption, and failure to thrive. Graft-versus-host disease (GVHD) due to transplacentally acquired maternal lymphocytes may occur during the neonatal period. There is increased susceptibility to viral infections, including cytomegalovirus, and chronic rotavirus infection may account for the intractable diarrhea in some patients. Jejunal biopsy reveals absent plasma cells, and in some cases there is partial villous atrophy with numerous periodic acid-Schiff–positive macrophages in the lamina propria. A high incidence of gastroesophageal reflux has been reported in association with SCID.[32] Without a bone marrow graft, the clinical course is rapidly fatal. In cases of ADA deficiency, enzyme replacement with irradiated erythrocytes or enzyme conjugated to polyethylene glycol is possible. Results with gene therapy have been encouraging, despite suboptimal initial results in ADA deficiency.[33,34]

Wiskott-Aldrich Syndrome Wiskott-Aldrich syndrome is an X-linked recessive condition characterized by eczema, thrombocytopenia, recurrent infections, progressive defect in T-cell function, and a poor antibody response to polysaccharide antigens. Atopy with elevated immunoglobulin E (IgE) levels is common. The underlying defective gene encodes the Wiskott-Aldrich syndrome protein, a member of a family of proteins responsible for signal transduction from the cell membrane to the actin cytoskeleton in hemopoietic cells.[35] Gastrointestinal manifestations include hemorrhage, chronic diarrhea, malabsorption, and nonspecific colitis. Without treatment, patients die from hemorrhage or the development of Epstein Barr virus–induced lymphoma. Stem cell transplantation is the only curative therapy for Wiskott-Aldrich syndrome.

Ataxia Telangiectasia Ataxia telangiectasia is an autosomal recessive, multisystem disorder. There are defective DNA repair mechanisms, cell cycle abnormalities, frequent chromosomal abnormalities, increased sensitivity to ionizing radiation, and predisposition to cancer. The gene responsible for this syndrome (*ATM*) encodes a product similar to phosphatidylinositol-3-kinases, which are involved in signal transduction and cell-cycle control.[36] The most prominent clinical features are progressive cerebellar ataxia, oculocutaneous telangiectasia, chronic recurrent sinopulmonary infections, and a high incidence of malignancy. The immune deficiency is variable and affects both cellular and humoral elements. IgA deficiency occurs in over 50% of patients and may be associated with IgG2 subclass deficiency. Malignancies are the most common cause of death. They usually are lymphoreticular, but adenocarcinomas also occur. Other gastrointestinal complications are not common in ataxia telangiectasia. Mild abnormalities of liver function may occur, and levels of a-fetoprotein are elevated.

Phagocytic Cell Defects: Chronic Granulomatous Disease

Intrinsic defects in neutrophils, monocytes, and macrophages include several disorders of cell locomotion, chemotaxis, killing, and metabolism. Numerically, the most important of these and the most frequently encountered in gastroenterology is chronic granulomatous disease.

Chronic granulomatous disease represents a group of disorders of phagocytic cell oxidative metabolism. There are several variants of the basic defect in oxidase function, and more than one mode of genetic transmission is involved.[37,38] In its classic form, inheritance is X-linked and is caused by mutations in the gene coding for the ß-subunit of cytochrome ß558, which is the catalytic redox entity of nicotinamide adenine dinucleotide phosphate (NADPH) and responsible for generating superoxide in phagocytes. More rarely, chronic granulomatous disease may be transmitted in an autosomal recessive or dominant pattern caused by mutations in the a-subunit of cytochrome ß558 or in cytosolic proteins required for activation of NADPH.

The respiratory burst associated with stimulation of phagocytes is lacking. This results in defective microbicidal activity because of the failure to generate toxic oxygen metabolites such as hydrogen peroxide. There is a markedly increased susceptibility to pyogenic and fungal infections, including staphylococci, *Serratia marcescens, Salmonella,* gram-negative enterococci, and *Candida* and *Aspergillus* species. There is particular susceptibility to infections with catalase-positive organisms. In contrast, catalase-negative organisms such as pneumococci, streptococci, and lactobacilli are not major pathogens in chronic granulomatous disease because they do not destroy their endogenous hydrogen peroxide and thereby contribute to their own demise.[37]

The condition classically presents during infancy, although variants of the disease may present later. Every organ is vulnerable to infection, and the associated granulomas occur throughout the body. Their formation is not well understood. They consist of plasma cells, lymphocytes, macrophages, and occasional multinucleated giant cells without organisms. A second type of granuloma occurs in which hyphal elements of fungi are found in giant cells. Pigmented, lipid-bearing tissue histiocytes also are found.[39]

Hepatic and gastrointestinal disorders are a prominent feature of chronic granulomatous disease. Hepatomegaly is very common, with liver abscess formation in more than 30% of patients. *Staphylococcus aureus* is the most common organism in the abscesses, which often require surgery and may be recurrent.

Gastrointestinal presentations of chronic granulomatous disease may mimic Crohn’s disease, and commonly include chronic gingivitis and stomatitis, perianal abscesses with fistulae, *Salmonella* gastroenteritis, diarrhea, and malabsorption.[37,39] The characteristic granulomas and pigmented histiocytes may be found in patients with no gastrointestinal symptoms. In some patients the granulomatous reaction leads to stricture formation, most commonly in the gastric antrum. Although surgical treatment may be required, resolution of strictures following antibiotic therapy is well known, and aggressive antimicrobial therapy is the treatment of first choice.[37,40]

Complement Deficiency: Hereditary Angioedema (C1 Esterase Inhibitor Deficiency and Nondeficiency Syndromes)

Deficiency states involving individual complement proteins are rare. In general, they are associated with a high incidence of infectious and autoimmune disorders.[41] Deficiency of C1 esterase inhibitor is the most common complement deficiency state and the major one in which gastrointestinal symptoms are a prominent feature. Defective C5a inhibitor protein in serosal fluid in patients with familial Mediterranean fever also occurs.[42]

Hereditary angioedema is characterized by recurrent, self-limited attacks of circumscribed nonpitting, subepithelial edema in the skin and mucous membranes.[43] The skin lesions are painless, and, unlike urticaria, pruritus is absent. The onset of symptoms is variable, although most patients have their first episode in childhood. A positive family history is common. Attacks may develop over hours and may last from hours to several days, often precipitated by minor local trauma, dental extractions, infections, and surgery. There is frequently initial awareness of a tingling sensation in the affected area, and a faint macular or serpiginous erythema may precede the swelling. The most serious manifestation is laryngeal edema, which may lead to fatal airway obstruction. Gastrointestinal involvement is common and includes colicky abdominal pain, vomiting, and watery diarrhea. Significant intestinal fluid loss can occur, leading to hypotension and shock. Fever and leukocytosis are notably absent. The abdomen may be tender but not rigid. Bowel sounds may be increased. Barium studies during an attack may show the “stacked coin”

appearance of mucosal edema. The lesions consist primarily of circumscribed edema without inflammatory cellular infiltrates. The edematous segment of bowel can cause intussusception. When gastrointestinal symptoms precede other manifestations, or occur in isolation, the diagnosis is often delayed, and many of these patients have been subjected to unnecessary abdominal surgery. [44]

Hereditary angioedema is transmitted in an autosomal dominant manner. [45] In approximately 85% of patients, the levels of C1 esterase inhibitor (C1-INH) are low due to heterozygosity for a non-expressed C1 inhibitor allele. In a minority of patients, levels of the inhibitor protein are normal but it is functionally defective due to heterozygosity for a nonfunctional C1 inhibitor allele. A third, estrogen-dependent, inherited form of angioedema has been described with no defect in C1-INH. [46], [47] and [48] This is transmitted in an autosomal dominant manner but may be X-linked in some cases. Episodes of angioedema are indistinguishable from those associated with other forms of the disease, but occur only during pregnancy or with the use of exogenous estrogens. Acquired forms of angioedema due to increased catabolism of C1-INH are clinically similar to the hereditary disorders occurring particularly in patients with lymphoproliferative disorders and collagen vascular diseases. [49] Drugs such as angiotensin-converting enzyme inhibitors may also cause angioedema.

The pathogenesis of angioedema is not fully understood, but the increased vascular permeability may be mediated by kinins. C1-INH prevents autoactivation of C1 and inhibits activated Hageman factor and kallikrein. The diagnosis, in most cases, is confirmed by quantitative and qualitative analysis of C1-INH. In addition, levels of C4 are secondarily reduced between attacks and nearly absent during attacks. Infusion of C1-INH concentrate is safe and effective treatment for severe attacks of hereditary angioedema. [50], [51] For prophylaxis, attenuated androgenic steroids, such as danazol and stanozolol, are effective. These drugs restore levels of C1-INH and C4, but may have other effects because patients often respond to low doses that do not raise levels of C1-INH and C4. The dose used should be tailored mainly to clinical rather than biochemical response.

Immunodeficiency Secondary to Gastrointestinal Disease: Protein-Losing Enteropathy and Intestinal Lymphangiectasia

Immunodeficiency due to protein loss may be a component of any severe inflammatory condition in the gastrointestinal tract; examples include protein-losing gastroenteropathy (PLGE) (e.g., Ménétrier disease) and extensive Crohn's disease. [52] However, the more severe cases of gastrointestinal protein loss occur in the setting of primary lymphangiectasia or secondary lymphatic obstruction [53] (see Chapter 81).

Other Secondary Immunodeficiencies of Gastroenterologic Importance

Protein-calorie malnutrition leads to significant depression of cell-mediated immune function. This may occur in association with malignancy, particularly lymphoreticular tumors. Iatrogenic immunosuppression is an increasingly common cause of secondary immunodeficiency (e.g., the occurrence of reversible IgA deficiency with drugs such as sulfasalazine, penicillamine, and phenytoin or in patients undergoing organ transplantation) (see Chapter 51). The increased susceptibility to *Candida* and other infections with corticosteroids is well known, but it is also important to be aware that corticosteroids are the most common cause of superinfection with *Strongyloides stercoralis.* [54]

Within developed countries, older adults are increasingly at risk of immunodeficiency. Systemic immune deficiency has been associated with increasing age, and infectious diseases are an important cause of morbidity and mortality in older patients. Studies of age-related changes in immune function have yielded conflicting results, however, and there is a lack of conclusive information on mucosal immune senescence in humans. The significance of age-related alterations in immune function is uncertain; reduced mucosal resistance and risk of bacterial overgrowth in older adults are probably multifactorial. Nonimmunologic risk factors include motility disturbances, achlorhydria, co-morbidity, polypharmacy, and antibiotic use with altered enteric flora. [55]

Neutropenic Enterocolitis Neutropenic enterocolitis is commonly a complication of chemotherapy for hematologic malignancy but may occur in patients with neutropenia of any cause. [56], [57] and [58] It is increasingly recognized, probably because of greater use of potent chemotherapy, occurring in 6% of patients treated for leukemia. [57] Neutropenic colitis is also called necrotizing enterocolitis, ileocecal syndrome and typhlitis (from the Greek *typhlos* meaning "blind sac" or cecum). It usually begins 7 to 10 days after chemotherapy, and affects primarily the ileocecal region with clinical manifestations ranging from abdominal discomfort, vomiting, distension, diarrhea, and tenderness to local or generalized peritonitis after perforation. Secondary sepsis due to bacterial translocation is common. Diagnosis requires a high index of suspicion—many cases are diagnosed postmortem. Bowel wall thickening may be evident on plain radiography and on ultrasonography or computerized tomography. Barium studies are generally avoided because of the risk of perforation. Management includes fluid and electrolyte replacement, broad-spectrum antibiotic coverage, and restoration of neutrophil count if possible. Depending on the severity, surgery may be required. [56], [58]

FOOD ALLERGY (HYPERSENSITIVITY)

Increasing public perception of potential adverse reactions to foods and food additives requires that physicians have a clear understanding of the true significance of food allergies. Although immunologic hypersensitivity to food stuffs was demonstrated almost 70 years ago, the issue of food allergy has been controversial. [59], [60] Much confusion has been created by the failure to use appropriate terminology distinguishing food allergy from other forms of adverse food reaction and to use an unbiased approach to the investigation of potential adverse reactions to foods. The development of scientifically sound experimental approaches has advanced our understanding of the pathogenesis of food allergy and improved diagnostic management of the problem. [61], [62] Reviews by the American Academy of Allergy and National Institutes of Health [63] and the American Gastroenterological Association [64], [65] have clarified and standardized terminology regarding adverse reactions of foods.

Classification and Prevalence

The terms *food allergy* and *food hypersensitivity* may be used interchangeably but should be reserved for those reactions that have been shown to be mediated by the immune system (Table 129-3). *Food intolerance* is a term that should be used to describe nonimmunologically mediated adverse reactions to food or food additives that may closely mimic acute allergic reactions. Reactions to food additives, such as sulfites, salicylates, and tartrazine, may be more common and as important as allergy to natural food components (see Table 129-3). Some foods may produce adverse reactions by multiple mechanisms in different individuals, including allergic reactions to milk proteins or, rarely, to contaminant antibiotics, or they may result from lactose intolerance or fat intolerance.

Immunologically mediated:
Early: Immunoglobulin E–mediated
Delayed: Late-phase reactions, immune (complex-mediated or cell-mediated immunity (e.g., celiac disease, most cases of cow's milk allergy)
Nonimmunologically mediated:
Idiosyncratic
Gastrointestinal (e.g., lactase deficiency)
Systemic (e.g., glucose 6-phosphate-dehydrogenase deficiency)
Pharmacological (e.g., caffeine, tyramine, serotonin, alcohol, histamine)
Toxic (e.g., aflatoxins, botulism, mushroom toxins)
Infectious (e.g., salmonellosis)
Other contaminants and additives
Antibiotics, pesticides
Dyes (e.g., tartrazine)
Flavorings and preservatives (e.g., monosodium glutamate [Chinese restaurant syndrome], sulfites, benzoate, nitrites, and nitrates)
Gastrointestinal disorders (e.g., peptic ulcer, cholelithiasis)
Psychological (e.g., phobias, avoidance, faddism)

*This list is intended to be representative and is not comprehensive.

TABLE 129-3 Differential Diagnosis of Adverse Reactions to Foods*

The spectrum of nonimmunologically mediated food intolerance is wide (see Table 129-3) and includes food avoidance, which may arise for various reasons, including faddism, anorexia nervosa, or even dysgeusia. Common causes of dysgeusia include medications (mostly halogen salts), menopause, and depression.

Community surveys indicate that at least 20% of adults believe that they have an adverse reaction to some food. When tested objectively, using blinded food challenges, the majority of complaints cannot be reproduced; estimates closer to 1% are more realistic. [66], [67] and [68] Most adverse reactions to food are probably not

immunologically mediated. The prevalence of true food allergy is more common in atopic individuals and in children, but declines with age. Estimates of the frequency of food allergies in children are about 5%, although cow's milk allergy may be more common. [65] In adults, food allergy is a well-documented entity, [66] but is uncommon. True food allergy is a most unlikely diagnosis in adults presenting with new onset of gastrointestinal symptoms in the absence of a strong atopic background or without a history of food allergy since childhood.

Immune Response to Dietary Antigens

Small amounts of antigenic macromolecules normally penetrate the gastrointestinal mucosal barrier and reach the systemic circulation, occurring more in early life and in conditions causing disruption of the mucosal defenses. Uptake of antigenic material and presentation to the mucosal immune system may occur by three main routes. [2] First, the specialized microfold or M cells that overlie lymphoid follicles are adapted to sampling lumenal antigens and delivering them to dendritic cells for presentation to T cells. [70] Second, the surface enterocytes lining the intestine are also capable of presenting antigens, and can selectively stimulate suppressor T cells in vitro. [71], [72] Although the significance of enterocyte-mediated uptake and processing of antigens in vivo is unclear, preferential stimulation of suppressor cells could be an important mechanism preventing hypersensitivity to dietary antigens. Finally, antigen sampling occurs across the epithelium by dendritic cells, which are capable of extending to the lumen without disrupting epithelial tight junctions. [73]

When dietary antigen engages the immune system, there may be a local secretory IgA antibody response, but a systemic immune response seldom occurs. Most commonly, a state of immunologic tolerance develops. Oral tolerance (sometimes termed the Sulberger-Chase phenomenon) is a state of specific immunologic unresponsiveness that occurs following oral ingestion of antigen, and significantly regulates immune responses to dietary antigens. [2], [74] Immune responses that are most easily suppressed or tolerated by feeding proteins are IgE-mediated and delayed-type hypersensitivity responses. Not surprisingly, these are the major immunologic reactions mediating food allergy. Stated differently, food allergy or hypersensitivity represents a breakdown in oral tolerance. Mechanisms responsible for the induction and maintenance of oral tolerance appear to involve activation of regulatory T cells. The molecular mediators include suppressive cytokines, particularly transforming growth factor-ß. [75] Moreover, the liver appears to down-regulate systemic immune responses to dietary antigens, [59] and the enteric flora have an obligatory role in the establishment of immune tolerance. [76]

Susceptibility to food allergy is genetically influenced but environmental determinants also play a role. All forms of allergy, including food allergies, are more common in developed countries, linked with improved sanitation and changes in the pattern of exposure to childhood infections. Some enteric infections may have a protective effect against food allergy, [76] although the relationship is complex since enteric infections may also act as an adjuvant for responses to ingested antigens. [77]

Pathogenesis of Food Allergy

The majority of food hypersensitivity reactions are caused by a relatively small number of foods, [64], [65] especially milk, eggs, nuts, fish, shellfish, soybeans, and wheat. For several of these foods, the specific allergen has been isolated or cloned. [78] Many food allergens are heat- and acid-stable, are resistant to proteolysis, and have a relatively low molecular weight. Their immunogenicity may be retained with food processing and digestion, and they have the appropriate size for bridging IgE on the surface of mast cells.

Since food-labeling regulations vary in different countries, allergens may be hidden in processed foods. [79] With the development of transgenic foods, it is also important to be aware that food allergens may be transferred by genetic manipulation. [80]

Food-allergic or hypersensitivity reactions are either immediate (IgE-mediated, type I hypersensitivity) or delayed (late-phase IgE-mediated, immune complex–mediated, and cell-mediated). Immediate or IgE-mediated hypersensitivity reactions account for the majority of immunologically mediated adverse reactions to foods. Delayed-type hypersensitivity is involved in the pathogenesis of gluten-sensitive enteropathy (see Chapter 76).

The wheal and flare reaction that occurs in some individuals after intradermal injection of a food allergen is due to the presence of food-specific IgE antibodies on the surface of cutaneous mast cells; adjacent IgE molecules are cross-linked by the allergen and trigger degranulation of the mast cell. This phenomenon could be passively transferred by a factor now known to be IgE. [81] When normal subjects were injected with serum from fish-sensitive patients, they developed wheal and flare reactions at the injection site upon ingestion of fish, showing that such antigens can reach and trigger mast cells at extragastrointestinal sites.

The central role of mast cell degranulation (Fig. 129-1) in the pathogenesis of food allergy is supported by the increased plasma levels of histamine in patients with cutaneous, gastrointestinal, and respiratory symptoms elicited by double-blind, placebo-controlled food challenge but not in patients with negative food challenges. [61], [62] Peripheral blood mononuclear cells from some patients with food allergy may spontaneously produce an IgE-dependent histamine-releasing factor that is associated with high spontaneous release of histamine in vitro from the patient's basophils and mast cells. [82] Studies in mast cell-deficient murine models of food hypersensitivity confirmed the central role of mast cells and their functional interaction with nerves and epithelial cells. [83] Eosinophils may partially mediate the late phase of food allergic reactions within the gut mucosa in experimental animals [84], [85] and may contribute to allergy-related dysmotility syndromes. [65] Interleukin-5 and the eosinophil chemokine ecotaxin are the primary mediators of eosinophil recruitment to the mucosa. [84], [85] and [86]

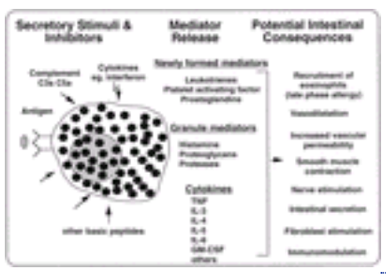

FIGURE 129-1. Mast cell mediators and potential consequences of their release. Mediator release is triggered when adjacent IgE antibodies on the surface of the mast cell are bridged by binding to an allergen. Mast cell secretion may also be stimulated by a variety of other endogenous and exogenous factors, including activated complement components (C3a and C5a), neuropeptides, bee venom, polymyxin, ionomycin, and opiates.

Clinical Features of Food Allergy

Symptoms usually occur within minutes of ingestion but may be delayed several hours. Systemic anaphylaxis is the most serious manifestation of food hypersensitivity and may be the first presentation. Co-factors such as aspirin ingestion or exercise may have a profound effect on the clinical expression of food hypersensitivity. Postprandial, exercise-induced systemic anaphylaxis has been well documented. More common, a variety of less severe and usually transient symptoms may be experienced. The organs primarily affected are the skin (eczema, urticaria), the respiratory tract (rhinitis, asthma), and the gut. [65], [87]

Gastrointestinal symptoms are nonspecific and range from edema and pruritus of the lips, buccal mucosa, and pharyngeal mucosa to vomiting, cramping, distension, and diarrhea, depending on the level of the gut primarily affected. Symptoms primarily involving the buccal or lingual mucosa (oral allergy syndrome) are particularly common and may not be associated with other gastrointestinal manifestations. Chronic diarrhea and malabsorption may arise as a result of non–IgE-mediated delayed hypersensitivity reactions, such as in food-protein enteropathies and celiac disease (see Chapter 76). Cows' milk hypersensitivity in infants may cause occult bleeding or frank colitis. [65], [88] Food allergy is not responsible for most cases of irritable bowel syndrome but should be considered in the small subset of patients with a strong history of atopy. [89] The importance of taking a careful history is confirmed by studies showing that symptoms can discriminate irritable bowel from food allergy. [90] There is no convincing evidence for food allergy in the pathogenesis of inflammatory bowel disease. Links between food allergy and other syndromes including esophageal reflux and dysmotility appear important in children and have been reviewed elsewhere. [65]

Evaluation of the Patient With Suspected Food Allergy

Only a small number of adult patients referred with a supposed food allergy have a reproducible, immunologically mediated adverse reaction to food. [67], [91] A careful and objective assessment is important not only to detect preventable disease in such patients but also to offset inappropriate and potentially harmful dietary restriction in others. Although the differential diagnosis of food-related symptoms is broad (see Table 129-3), a detailed history and physical examination will often indicate the correct diagnostic category. The most definitive criteria for the diagnosis of food allergy are:

- ingestion of the implicated food reproducibly induces the patient's symptoms evidence that an immunologic mechanism is involved.

A general diagnostic guideline is shown in Figure 129-2. [64] In practice the diagnostic approach should be individualized, and the rigor with which one attempts to prove the diagnosis will be influenced by several factors, such as the age of the patient, the nature and nutritional importance of the food implicated, the likely reliability of the patient's history and compliance with challenge test protocols, and the severity of the symptoms. For example, patients who are thought to be allergic to several major food groups require an accurate confirmation of the diagnosis because of the inconvenience and potential danger of malnutrition with prolonged, complicated restriction diets. In contrast, patients with only episodic symptoms triggered by uncommonly encountered or nonessential foods should be instructed to avoid such foods and do not require rigorous food challenges. For reliable patients with a clear history of objectively verifiable symptoms and signs, an open challenge may suffice. The patient with vague or subjective complaints, self-diagnosed as an adverse reaction to food, poses the greatest difficulty. In adults, particularly the nonatopic individual, the likelihood of such symptoms being due to food allergy is small. Prolonged investigative evaluation beyond simple screening tests may not be appropriate or desirable for these patients. [91]

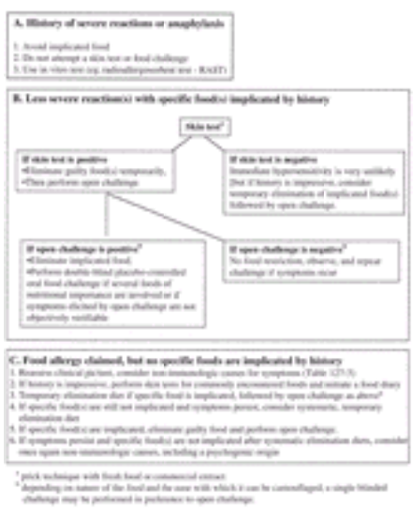

FIGURE 129-2. General guidelines for the investigation of adult patients with suspected food allergy.

Food challenges and skin testing with food antigens can trigger potentially serious and even fatal anaphylaxis in a small percentage of patients. Such diagnostic tests should be performed only by experienced physicians and only in a facility equipped to address serious allergic reactions.

Skin Tests and In Vitro Assays In vitro tests for cellular immunity to food antigens and assays for food antigen-antibody complexes are still under investigation and not recommended for routine diagnostic use. For practical purposes, the demonstration that an adverse reaction to food is immunologically mediated involves the demonstration of IgE antibodies specific for the implicated food allergen. Direct skin testing with the prick technique with food extract and control solution is a simple and sensitive method of detecting specific mast cell–bound IgE antibodies. [65], [69] If properly performed, a negative skin test indicates that immediate hypersensitivity to the food tested is highly unlikely. However, a positive test (wheal >3 mm in diameter larger than that of the control wheal) merely indicates the presence of sensitizing antibodies and not necessarily the presence of clinical food hypersensitivity, which can be confirmed conclusively only by food challenge (see Fig. 129-2). Skin testing should not be attempted if the clinical history indicates there is a substantial risk of anaphylaxis; an in vitro assay for the presence of allergen-specific IgE in the serum should be used. Assays for IgE antibodies to several of the known food allergens include the commercially available radioallergosorbent test and enzyme-linked immunosorbent assay (ELISA) kits. Less commonly used is the basophil histamine release assay, which is performed mainly in research laboratories. There is no place for the nonvalidated, so-called cytotoxicity food allergy test, which is still widely advertised in the lay press. [64], [65]

Elimination Diets Systematic elimination of different foods and the use of a food/symptom diary may help identify foods to which a patient might be allergic. Resolution of symptoms and recurrence with reintroduction of selected foods is consistent with a diagnosis of food hypersensitivity. If multiple or several major food groups are implicated, confirmation should be obtained by controlled food challenge. The design of the diet, particularly the degree of food restriction, will depend on the patient's history and the number of potential food allergens thought to be involved. Detailed discussion and examples of different elimination diets are available. [63], [65] Ideally, the design of the diet should involve an experienced dietitian. Reintroduction of the implicated food(s) should be supervised by a physician; anaphylaxis was reported in 5% of patients with atopic dermatitis during food challenge following elimination trials. [92] The most restrictive elimination diet is an elemental diet. Elemental diets are unpleasant to take, but on a short-term basis they may be diagnostically useful in patients claiming to have allergies to multiple foods, or where the possibility of food allergy is a major concern but no specific food has been implicated by the clinical history. Persistence of symptoms on such a diet excludes food allergy from the differential diagnosis. If symptoms resolve on an elemental diet, various foods may be reintroduced in a systematic fashion.

Food Challenge A double-blind, placebo-controlled oral food challenge is the most reliable and unbiased method for determining if symptoms are related to a specific food. However, like direct skin testing, oral food challenge is contraindicated in patients with a history of food-related anaphylaxis. Dehydrated foods are most convenient for diagnostic challenges because they can easily be disguised in a variety of vehicles or gelatin capsules, and are commercially available. Detailed guidelines for the performance of food challenges have been published. [93], [94] Because the double-blind, placebo-controlled food challenge is time consuming and expensive, "single-blinded" or "open" challenges are convenient initial tests in selected cases to rule out food allergy or to help identify those who require a double-blind challenge (see Fig. 129-2). Direct challenge of the intestinal mucosa with food antigen extracts using colonoscopy has been used to identify patients with intestinal allergy. Mucosal wheal and flare reactions are recorded semiquantitatively and tissue biopsy specimens are examined for mast cell and eosinophil activation. As with skin testing, positive and negative controls are required and similar safety considerations must be applied. The most suitable gastrointestinal site for the challenge has not been determined and may vary in different individuals and depend on the nature of the allergen. [95]

Treatment and Prevention of Food Allergy

The majority of patients can be reassured they will outgrow their food allergy. Peanut allergy, the most common cause of severe food allergy in adults, may be an exception, although it is outgrown in approximately 20% of cases. [96] The only acceptable treatment of food allergy is avoidance of the offending food. Patient education is the best means of ensuring dietary compliance. Expert instruction and supervision by an experienced dietitian reduce the likelihood of inadvertent ingestion of allergens. Drug therapy with antihistamines and corticosteroids has only a secondary role in the management of symptoms due to inadvertent ingestion of food allergens. [97] Parenteral immunotherapy (hyposensitization) with food extracts has been disappointing and has no role at present. There is some evidence to suggest that breast-feeding until infants are 6 months old may have a protective effect against the development of food allergy. The potential prophylactic role of altering the enteric microflora with probiotics has become an area of investigation. [98]

EOSINOPHILIC GASTROENTERITIS

Pathophysiology

Eosinophilic gastroenteritis is probably not a single entity but rather a heterogeneous collection of disorders of varied causes with similar clinicopathologic features. [65], [99], [100] Allergic phenomena, particularly food hypersensitivity, have been proposed to account for eosinophilic gastroenteritis but the evidence for this is limited in

many patients. Although food hypersensitivity (usually to milk) may be found in children with eosinophilic gastroenteritis, the response to dietary elimination is often disappointing. Some patients have no atopic symptoms, and fewer than half of all patients with eosinophilic gastroenteritis have a positive family history of allergy. Studies of experimental models of allergic intestinal disorders have demonstrated the role of the eosinophil chemokine, ecotaxin, and of eosinophils in mediating intestinal injury including neuronal damage. [84], [85] Other cytokines including IL-3, granulocyte-macrophage colony-stimulating factor, and IL-5 promote persistence of the eosinophil infiltration within the gut. [86], [101], [102]

Clinical Manifestations

The clinical manifestations of eosinophilic gastroenteritis depend on the gastrointestinal site primarily affected and the layer of bowel wall predominantly involved. [65], [99] The stomach and intestine are most commonly involved, but the disease may affect any part of the gastrointestinal tract as well as the pancreatic and biliary tree. When the predominant involvement is mucosal, the condition behaves like other forms of inflammatory bowel disease, with diarrhea, cramping, postprandial nausea, vomiting, and periumbilical pain. If the eosinophilic infiltration is extensive, there may be malabsorption, weight loss, protein-losing enteropathy, blood loss, and anemia. In preadolescents, growth retardation may occur. A low level of peripheral eosinophilia occurs in most cases. Stools may be positive for occult blood, although frank hematochezia is rare. Charcot-Leyden crystals secondary to lumenal extrusion of eosinophils may be seen. Detection of these distinctive hexagonal bipyramidal crystals may be easier with fluorescence microscopy than conventional light microscopy. Although characteristic of eosinophilic involvement in inflammation, they may also be derived from basophils and are not specific for allergic reactions, being found in parasitic and other causes of inflammation including idiopathic inflammatory bowel disease. Quantification of mediators such as eosinophil cationic protein and eosinophil protein X within stool samples promises to be an alternative diagnostic strategy. [103] Definitive diagnosis of mucosal eosinophilic gastroenteritis is made by barium roentgenography and gastric and small bowel biopsy. A diffuse mucosal pattern with nodules or intralumenal masses and a sawtooth mucosal pattern may be noted in the small bowel. A widening of small bowel segments may be seen secondary to mesenteric nodal involvement. Gastric lesions may have a cobblestone appearance in the antrum and thickening of mucosal folds. Small bowel biopsies may be diagnostic, showing a diffuse infiltration of the mucosa with eosinophils. However, because eosinophilic gastroenteritis may present as a patchy intestinal disease, multiple biopsies at different levels of the gastrointestinal tract should be performed.

When eosinophilic gastroenteritis predominantly involves the muscularis layer of the intestine, the clinical presentation consists of intestinal obstruction with nausea, vomiting, pain, and abdominal distention. Barium studies of the upper gastrointestinal tract will usually demonstrate irregular narrowing of the distal antrum or small bowel. Endoscopic mucosal biopsy may not be helpful. Occasionally, isolated infiltration of esophageal muscle may be seen with symptoms and manometry evaluations suggestive of achalasia. Rarely, the eosinophilic infiltration is predominantly serosal and may be associated with eosinophilic ascites or pleural effusions.

Some cases of gastroesophageal reflux, particularly in children, may represent a localized form of eosinophilic infiltration—eosinophilic esophagitis. [65] This suggests that food allergy or other hypersensitivity phenomena should be considered in the management of children with reflux. Whether there is a similar process in adults is less clear and probably uncommon.

Differential Diagnosis

Diseases associated with peripheral eosinophilia in addition to mucosal eosinophilic infiltrates may mimic idiopathic mucosal eosinophilic gastroenteritis. The most frequently encountered is the hypereosinophilic syndrome, characterized by diffuse gastrointestinal involvement, frequent peripheral eosinophilia, and eosinophils in several other organs. Periarteritis nodosa may be associated with a peripheral eosinophilia, and abdominal pain and nodular masses in the stomach and small bowel may be seen on radiography. This condition may be distinguished from eosinophilic gastroenteritis by the systemic nature of the inflammatory process and by the eosinophilic infiltrate localized to the perivascular region. The nodular polypoid mucosal findings on radiograph in periarteritis nodosa are secondary to ischemia and inflammation rather than eosinophilic infiltration. Intestinal parasitism (hookworm, *Ascaris, Strongyloides, Toxocara, Trichuris, Capillaria,* and *Trichinella* species) causes peripheral eosinophilia and gastrointestinal symptoms, including diarrhea and abdominal pain. The diagnosis is usually suggested by the clinical history and confirmed by stool analysis and examination of a duodenal aspirate. Protozoal infections such as amebiasis and giardiasis are not associated with eosinophilia. Finally, intramural collections of mature eosinophils, most commonly in the stomach, are often seen in eosinophilic granuloma. These patients present with symptoms of gastric obstruction. The isolated nature of the granuloma may help distinguish this disorder.

The second major category of diseases that may mimic eosinophilic gastroenteritis clinically and radiologically includes inflammatory and infiltrative disorders, such as gastrointestinal lymphoma, scirrhous carcinoma of the stomach, and Crohn's disease. These conditions may be associated with a mild increase in eosinophils histologically.

Treatment

There is no specific treatment for eosinophilic gastroenteritis. An attempt to identify a specific environmental or food allergen may be considered if there is a convincing history of symptoms triggered by food and if there is a strong background of atopy. In most adult patients, a specific food will not be implicated, and prolonged investigation with elimination diets is not appropriate. If necessary, a trial of an elemental diet can quickly eliminate a role for food hypersensitivity. Oral corticosteroids, 20 to 40 mg prednisone/d are effective in most patients. A short course of 7 to 10 days usually produces clinical remission, although repeat courses may be required. Some patients may require more prolonged treatment. The long-term prognosis for patients with eosinophilic gastroenteritis is generally favorable, and the disease course is one of waxing and waning severity. Occasionally, the use of total parenteral nutrition is required to manage patients with severe disease and symptoms that are refractory to steroids. Segmental resection of the intestine is not curative.

GASTROINTESTINAL COMPLICATIONS OF ORGAN TRANSPLANTATION

Gastrointestinal complications are common in patients undergoing transplantation, particularly bone marrow transplantation. Complications include toxicity of induction protocols (radiation or chemotherapy) prior to bone marrow grafting; acute and chronic GVHD; and opportunistic infections due to immunodeficiency associated with both the induction procedure and graft-versus-host reactions. [104], [105] and [106] Some of the clinical and diagnostic characteristics that help distinguish these entities in the setting of bone marrow transplantation are shown in Table 129-4.

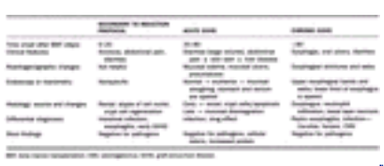

TABLE 129-4 Gastrointestinal Complications of Bone Marrow Transplantation

Intestinal Disease Secondary to Induction Protocols

Induction protocols are varied and include multiple agents (both chemotherapeutic agents and high-dose radiation), which are capable of injuring rapidly dividing cell populations and producing intestinal cell death (see Table 127-4). Enterocyte death occurs immediately during the induction and requires up to 3 weeks before there is mucosal regeneration. Both the small and large intestine are damaged during this process, although the small intestinal epithelium is more susceptible to this form of cell death. [107] Anorexia, cramping, and abdominal pain, with watery diarrhea, may develop. During healing, there is atypia of cell nuclei and regeneration of crypt cells, but the surface epithelium is normal. [108] This histopathological picture is important in helping distinguish this entity from acute GVHD or secondary viral infections. The time of onset following bone marrow transplantation is a helpful clue, and rectal biopsy with negative stool cultures are the most important diagnostic tests (see Table 129-4). Treatment consists of supportive measures and parenteral nutrition.

Graft-Versus-Host Disease

GVHD is most frequently seen in the setting of bone marrow transplantation but may occur with other transplants. Acute GVHD is characterized by epithelial cell death mainly in the gastrointestinal tract, liver, and skin, whereas chronic GVHD is characterized by fibrosis and atrophy of these and other organs. [109] GVHD is a multistep process in which the induction phase involves primarily CD4 T-lymphocytes transferred with the donor graft which recognize disparities with the recipient's major histocompatibility complex (MHC) antigens as foreign. Different subsets of memory T cells expressing distinct adhesion olecules home separately to the gut and skin. [110] Activation of these alloreactive donor T cells within the tissues is followed by clonal expansion (expansion phase). This is followed by cytokine and chemokine release ("cytokine storm") which leads to recruitment of additional inflammatory cells (effector phase). [111] The immunopathology is consistent with T-cell–mediated tissue injury, [112], [113] with predominance of tumor necrosis factor-a (TNF-a) and other T_H1 cytokines. [114], [115]

Acute GVHD A moderate to severe degree of acute GVHD develops in 30% to 50% of patients with sustained bone marrow engraftment. Acute GVHD may be staged on the basis of skin (dermatitis), gut (mucositis, enteritis), and liver damage. [104], [105] The onset or continuance of profuse, watery diarrhea 3 weeks following bone marrow transplantation is indicative of intestinal acute GVHD. The severity of the intestinal condition generally parallels that of the skin and liver involvement, although profound gastrointestinal symptoms can occur without any gross skin or liver changes. Altered mucosal barrier function due to conditioning protocols is associated with increased translocation of bacterial lipopolysaccharide (LPS) and the responsiveness of donor cells to LPS appears to predict the severity of acute GVHD. [116] The clinical picture, when severe, includes anorexia, vomiting, buccal mucositis, abdominal pain, intestinal bleeding, protein loss, and secondary infection (see Table 129-4). High-volume diarrhea may occur, and the amount of fluid generated is an index of extent and severity of disease activity. Extensive disease may be associated with up to 10 L of diarrheal fluid loss per day. The large stool volume may lead to distention and pain. Distention of the bowel may be exacerbated by opiate analgesics, which should be used with caution. Because the symptoms of GVHD are not specific, vigilance is required for other complications, particularly infections. The most common cause of gastrointestinal bleeding in these patients is esophagitis and gastric erosions. However, when a discrete ulcer develops, cytomegalovirus infection is an important consideration. Medical treatment of such lesions is often unsatisfactory, and if bleeding is persistent, surgical resection may be required. Features that distinguish acute GVHD from the enteritis associated with the induction protocol include an erythematous, maculopapular skin rash over the palms, soles, and trunk, as well as liver test abnormalities, including hyperbilirubinemia. The diagnosis of acute GVHD may be made with confidence approximately 3 weeks following bone marrow transplantation if there is evidence of a functioning graft and if there is watery diarrhea accompanied by a diffuse skin rash and jaundice (see Table 129-4). The patient who presents with gastrointestinal symptoms but without jaundice or skin rash represents a much more difficult diagnostic challenge. Other diagnostic considerations include an opportunistic intestinal infection and the residual effects of the chemoradiotherapy conditioning lasting beyond the usual 3-week period. A combination of stool examinations, barium radiographic studies, endoscopy, and histopathology may be helpful (see Table 129-4). The stool usually contains large amounts of cellular debris and red and white blood cells. Protein loss in the stool may be sufficiently severe to lead to profound hypoalbuminemia. The absence of pathogens in the stool is important but does not necessarily rule out an infection of the gastrointestinal tract. Barium roentgenography may be helpful in the differential diagnosis and in establishing extent of disease. Widespread changes may occur, including mucosal and submucosal edema, pneumatosis cystoides intestinalis, and mucosal ulcerations. These radiologic findings may also be consistent with other conditions, such as cytomegalovirus enteritis and the effects of acute radiation. The acute radiologic features may return to normal, or in some cases they may progress to a chronic segmental, ribbonlike appearance, usually in the small intestine. Endoscopic appearances may be normal or may show patchy erythema or extensive mucosal sloughing. These lesions are most prominent in the ileum, cecum, and ascending colon, with relative sparing of the gastric and rectal mucosa. Histological changes seen in biopsy samples from advanced or severe lesions may not be as diagnostically useful as those from early lesions. Therefore, endoscopic biopsy samples should be taken from intact mucosa as well as from areas involved with gross inflammation. The earliest change seen on light microscopy is apoptosis of individual cells in the intestinal crypts (Fig. 129-3). This is diagnostic, if obtained from normal-appearing mucosa at least 20 days following transplantation. [117] Inflammatory cells or microorganisms are not present in the adjacent mucosa. Later, the histopathology can progress to a total denudation of the mucosa; the apoptosis lesion is no longer evident, and changes are not specific.

FIGURE 129-3. FIGURE Rectal biopsy specimen taken after bone marrow transplantation from a patient with acute graft-versus-host disease shows apoptotic bodies (*arrows*). This necrosis of individual cells within the crypt is the first change seen on light microscopy and is characteristic of acute graft-versus-host disease if found after day 20, when damage from chemoradiation therapy has resolved.

Management of intestinal acute GVHD consists of nutritional support, maintenance of fluid and electrolyte balance, steroid and immunosuppressive treatment, and vigilance for secondary infectious complications. Common opportunistic pathogens include astrovirus, *Clostridium difficile,* and adenovirus. [118] For patients with severe high-volume diarrhea, subcutaneous administration of octreotide may provide relief. To improve long-term survival, doses ranging up to 2 mg/kg of prednisone, for up to 1 to 2 weeks, may be used. Cyclosporine may be used in combination with specific anti–T-cell monoclonal antibodies and inhibition of TNF-a is promising. [114] A subset of patients with intestinal GVHD do not progress to severe multisystem disease and can be managed more conservatively with less toxic drug strategies, including topically active steroids, thereby minimizing the risk of opportunistic infections. [106], [119] Prevention of GVHD can be achieved by prior removal of mature donor T-lymphocytes from the bone marrow graft. Unfortunately, this is associated with reduced graft survival. To circumvent this, several new experimental strategies for bone marrow transplantation and prevention of GVHD are under investigation. [120], [121] and [122]

Chronic GVHD Chronic GVHD is a multisystem disorder that occurs 80 to 400 days after allogeneic transplantation (see Table 129-4). The majority of patients have had prior acute GVHD. However, approximately one fourth of the patients with chronic GVHD have had no clinical indications of any prior acute graft-versus-host illness. The skin, liver, and gastrointestinal tract are predominantly involved. The clinical features resemble those of sicca syndrome and systemic sclerosis. Gastrointestinal involvement occurs particularly in the oral mucosa (mucositis), esophagus, and small intestine. Esophageal symptoms of dysphagia with weight loss are common presenting gastrointestinal manifestations. [123], [124] Patients with skin involvement (hyperpigmentation and scleroderma-like changes) frequently, but not always, have esophageal involvement. Dysphagia with chronic esophageal reflux can lead to chronic lung disease. Small bowel involvement with chronic GVHD is characterized either by a patchy fibrosis of the lamina propria and submucosa, or by bacterial overgrowth as a result of stasis and dysmotility. This latter abnormality responds to oral broad-spectrum antibiotics. As with other immunosuppressed conditions, opportunistic gastrointestinal infections are a constant threat posttransplantation and must always be considered in the differential diagnosis. The combination of radiology, esophagoscopy with biopsy, and manometry may be required to distinguish between esophageal involvement with chronic GVHD and reflux peptic disease of the esophagus. Endoscopic lesions of chronic GVHD can range from generalized desquamation of the upper and midesophagus to weblike fibrous bands. [123] Manometry with pH monitoring shows a nonspecific motor abnormality and a decreased ability to clear acid from the esophagus. In contrast to peptic esophagitis, the distal esophagus is usually spared in chronic GVHD. [124] Histological changes within the esophagus include infiltration with neutrophils and lymphocytes with necrosis of individual cells of the basal mucosa, similar to the changes found in the skin and oral mucosa. Submucosal fibrosis may be found, but there are no muscle or neural abnormalities. In contrast to acute GVHD, the treatment of chronic GVHD is frequently satisfactory. Early diagnosis and drug therapy have prevented much of the disability associated with this condition. Prednisone alone or in combination with azathioprine has been effective. Aggressive antireflux therapy is important for reducing the symptoms associated with esophageal involvement. However, fibrosis tends to be progressive, and esophageal dilation may be required for treatment of webs and strictures.

The Liver in GVHD

In acute GVHD, liver involvement is manifested by cholestasis and mild hepatocellular necrosis. Hepatic failure is uncommon. [104] Although liver abnormalities may be the presenting feature of acute GVHD, skin and intestinal GVHD are usually evident by the time jaundice occurs, and diagnosis is relatively clear when these multisystem manifestations arise at least 3 weeks following bone marrow transplantation. Within the first 2 to 3 weeks after transplantation, venoocclusive disease

secondary to chemoradiotherapy may be a more important diagnostic consideration, whereas throughout the posttransplant period, GVHD must be distinguished from, or may coexist with, several other causes of hepatotoxicity. These include parenteral nutrition, viral infections, drug-induced liver damage (such as immunosuppressants, antibiotics, and antifungals), hypotension, and sepsis.

Biochemical abnormalities due to liver complications of GVHD include a marked elevation in serum alkaline phosphatase followed by hyperbilirubinemia, but serum transaminases are elevated to a lesser extent. Liver biopsy is hazardous because of the risk of bleeding due to thrombocytopenia in bone marrow transplant patients, but may be helpful particularly where additional causes of liver disease are suspected. Histological changes of GVHD within the first week after the onset of biochemical abnormalities are nonspecific and consist of a mild lobular hepatitis with variable eosinophilic necrosis of hepatocytes and necrosis of biliary epithelium. Subsequently, the changes become more characteristic with abnormal interlobular bile ducts followed by destruction of small bile ducts and proliferation of ductules. The histological picture may evolve into one of profound chronic cholestasis with portal inflammation and fibrosis. Progression to liver failure may occur but most patients with severe GVHD die of infection or other complications before this. At least half of the patients who survive severe acute GVHD develop chronic GVHD and most of these will have liver involvement. In contrast to the mixed hepatocellular-bile duct damage of acute GVHD, the liver damage in chronic GVHD is predominantly portal with dense plasmacytic infiltration, loss of small bile ducts, and cholestasis. [105]

SMALL BOWEL TRANSPLANTATION (See AlsoChapter 79)

Transplantation of the small bowel has become a realistic therapeutic strategy for patients with intestinal failure where continued parenteral nutrition is no longer a safe option. [125], [126], [127] and [128] These will usually be patients with complications of parenteral nutrition such as advanced liver disease or loss of venous access due to catheter sepsis or venous thrombosis. Intestinal failure in adults is generally due to short bowel syndrome following resection for conditions such as Crohn's disease or infarction associated with trauma or hypercoagulable states. In children, congenital disorders such as atresia or necrotizing enterocolitis are the most common indications for small bowel transplantation. Improvements in the success rate for intestinal transplantation have been due, in large part, to the introduction of potent immunomodulatory agents such as tacrolimus, [129] and 5-year survival rates for graft and patient have been reported in the range of 32% to 52% and 33% to 38%, respectively. [130] Unfortunately, graft survival is not equivalent to freedom from parenteral nutrition; results from the international intestinal transplant registry indicate full graft function has been achieved in only 78% of survivors, with partial or nonfunctioning grafts in 10% and the requirement for removal of the graft in the remainder. [131]

Contrary to what had been expected, GVHD has not been a major obstacle. [128], [129], [130], [131] and [132] However, the procedure still has relatively high mortality and morbidity because of rejection, sepsis, and posttransplant lymphoproliferative disorder. Rejection predisposes to sepsis because of loss of mucosal barrier function. This is compounded by potent immunosuppression with the risk of opportunistic infections, particularly cytomegalovirus enteritis, and a subsequent lymphoproliferative disorder that is probably related to Epstein-Barr virus infection. [129]

Distinguishing rejection from infections and other pathology may be difficult, particularly because the rejection process is often focal or patchy. Multiple endoscopic biopsies are required to monitor the progress of the graft. Crypt cell apoptosis characterizes acute rejection, whereas chronic rejection is associated with fibrosis, focal ulceration, and obliterative arteriopathy. [133] Recipient lymphocytes infiltrate the lamina propria of the graft and replace donor lymphoid cells within weeks to months. The rejection reaction is associated with local production of T_H1 cytokines including TNF-a and interferon-?, and targets mainly the crypt epithelium and local endothelium. [134] Neither the morphology nor the cytokine response is specific to the rejection process. Clinical details including preventive and therapeutic strategies for rejection have been reviewed. [130] Vigilance and careful attention to detail are required, and the degree of immunosuppression to prevent rejection must be balanced against risk of opportunistic infection. Gene therapy of the graft may in the future render it resistant to immune assault [135] and normal mechanisms of tolerance and immune privilege might be used to prevent graft rejection. [136]

With longer survival of intestinal grafts additional complications may be anticipated (e.g., recurrence of Crohn's disease in the intestinal graft). [137] This contrasts with reported resolution of Crohn's disease following bone marrow transplantation. [138] Thus, substitution of host immunity rather than the target organ is required, consistent with the concept that the fundamental defect in Crohn's disease does not reside exclusively within the target organ and probably reflects a dysregulated immune response to enteric flora.

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CHAPTER 130

Mitchell S. Cappell

GASTROINTESTINAL VASCULAR MALFORMATIONS OR NEOPLASMS: ARTERIAL, VENOUS, ARTERIOVENOUS, AND CAPILLARY

The classification of gastrointestinal vascular lesions has been confusing due to variable classification schemes and inadequate consideration of anatomic and pathophysiologic principles. [1] Figure 130-1 presents a simple classification based on anatomic and pathophysiologic principles. Vascular lesions are divided into neoplastic lesions and nonneoplastic malformations, the latter including inflammatory (vasculitis) and structural malformations. Structural malformations are divided anatomically according to the most affected vascular component: arteriovenous, arterial, capillary, and venous. Only the entities highlighted in color in Figure 130-1 are discussed in this chapter.

FIGURE 130-1. Classification of vascular gastrointestinal lesions. Diagrammatic outline of a new classification of vascular gastrointestinal lesions according to pathophysiology and according to the most affected vascular structure, whether arterial, venous, arteriovenous, or capillary. Diseases highlighted blue are discussed in this chapter.

The nomenclature of these lesions has also been confusing owing to redundancy, application of erroneous pathophysiologic mechanisms, use of the same name for disparate diseases, and inconsistent application of names to disparate diseases. For example, the Dieulafoy lesion has been called an exulceratio simplex, cirsoid aneurysm, caliber persistent artery, and submucosal arterial malformation; the Dieulafoy lesion is not an aneurysm. [2] The term phlebectasia has been used to describe both small hemangiomas or isolated venous varicosities. [3] The most commonly used name is employed in this chapter, provided the name is pathophysiologically reasonable. The vascular lesions described herein are rare, except for angiodysplasia, the Dieulafoy lesion, and gastric antral vascular ectasia (GAVE). However, it is important to recognize even the rare vascular diseases because they are frequently misdiagnosed and mistreated. For example, until recently, most rectal hemangiomas were misdiagnosed and mistreated as internal hemorrhoids. [4]

DIEULAFOY LESION

Pathology and Pathophysiology

Although uncommon, the Dieulafoy lesion is important because it tends to bleed repeatedly and massively. Arteries normally progressively narrow in the arterial tree as successive branches approach the mucosa to terminate as submucosal end arteries. In the Dieulafoy lesion, the submucosal end artery is abnormally large; the alternative name of caliber-persistent artery is based on this abnormality. The lesion usually, but not always, occurs in the stomach, within 6 cm of the gastroesophageal junction and near the lesser curvature. The propensity for this location is explained by the absence of the usual submucosal arterial anastomotic gastric plexus in this region. [5] In three large clinical series, this lesion was in the proximal stomach in 75% of patients, the distal stomach in 13%, and the duodenum in 12%. [6], [7] It occurs rarely in the esophagus. [8]

The Dieulafoy artery is vulnerable to relatively minor mechanical trauma because it is large and superficial. Pulsations of the artery, peristaltic contractions, or passage of the food bolus may cause gastrointestinal bleeding. [9] Alternatively, the large artery might cause hypoperfusion, ischemia, and erosion of overlying mucosa by shunting of blood (steal phenomenon). Both theories are compatible with the pathological findings of a thrombus attached to a large superficial artery overlying a

small nonexcavated mucosal defect, with a normal vascular endothelium, without atherosclerosis, inflammation, or aneurysm. [2], [9], [10] Whether the lesion is congenital or acquired is unknown. [2], [11]

Epidemiology and Clinical Presentation, Upper Gastrointestinal Lesion

The incidence is about 1.5% among patients with upper gastrointestinal bleeding. [6], [12], [13] and [14] The male-to-female ratio is about 2:1, [7], [10], [14] and the mean age at diagnosis is 55 years, with a range of 1 to 95 years. [7], [10] The lesion is not associated with use of alcohol, nonsteroidal antiinflammatory drugs, or tobacco, nor with the presence of peptic ulcer disease or *Helicobacter pylori* infection. [7], [10], [15], [16]

Patients present with acute, usually severe, gastrointestinal bleeding, often associated with manifestations of hemodynamic compromise such as hypotension, orthostasis, tachycardia, and prerenal azotemia. The bleeding is rarely occult and usually presents as hematemesis, melena, or hematochezia; about 50% of patients manifest both hematemesis and melena. [10] Other gastrointestinal complaints, such as abdominal pain, are atypical, and their presence should suggest another diagnosis, such as peptic ulcer or esophagitis. [7]

Diagnosis, Upper Gastrointestinal Lesion

The lesion is usually diagnosed by endoscopy, which reveals a pigmented protuberance, representing the vessel stump, with minimal surrounding erosion nd no ulceration (Fig. 130-2; see also Color Fig. 130-2). The lesion is typically small; sometimes only a tiny necrotic spot is found in the proximal stomach. A pigmented protuberance within an ulcer represents a visible vessel in a peptic ulcer, not a Dieulafoy lesion. Stigmata of recent hemorrhage, such as frank bleeding or an adherent clot, commonly occur. The lesion should be suspected when a fresh densely adherent clot in the proximal stomach has a narrow point of attachment to a small mucosal defect. The lesion may be missed at endoscopy because of small size, concealment between gastric rugae or underneath blood in the gastric lake, or careless gastric retroflexion. [17] Indeed, in one series of 177 cases, the lesion was missed in 51% of the initial esophagogastroduodenoscopies (EGDs) and then detected in 33% of cases on repeat EGD. [10] Meticulous endoscopic technique—including careful examination during gastric retroflexion, aspiration of the gastric lake, and adequate gastric insufflation to efface the gastric rugae—should increase the sensitivity of endoscopy.

FIGURE 130-2. (See Color Fig. 130-2.) Endoscopic appearance of Dieulafoy lesion. Videophotograph shows a gastric cardial Dieulafoy lesion as a small, pigmented protuberance, which represents the vessel stump, with minimal surrounding erosion and no ulceration. (From: Lee JG, Leung JWC. Stigmata of recent hemorrhage in Dieulafoy's lesion. Gastrointest Endosc 2000;51:191.)

Angiography is indicated for ongoing upper gastrointestinal bleeding after a nondiagnostic EGD and was found to be diagnostic in 11 of 14 patients. [10] Angiography is suggestive of this lesion when extravasation of contrast is detected from a point source in the proximal stomach after a nondiagnostic EGD. Angiography may also demonstrate arterial tortuosity in the territory of the left gastric artery without other vascular abnormalities. [18] Angiographic findings of arteriovenous shunting and an early-filling or late-draining vein are consistent with angiodysplasia and not a Dieulafoy lesion. The Dieulafoy artery is too small in caliber to be recognized as enlarged by angiography and, besides, is often obscured by extravasated contrast.

Recent anecdotal reports suggest endoscopic ultrasound can confirm the diagnosis. In one series of eight patients, an abnormally large, 2- to 3-mm wide, tortuous gastric vessel penetrated through the muscularis propria and coursed through the submucosa for at least 2 cm [19], [20] (Fig. 130-3).

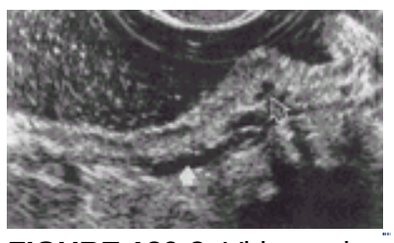

FIGURE 130-3. Videoendosonographic image of a Dieulafoy lesion. A relatively large gastric vessel courses outside the muscularis propria (left of figure, *solid arrow*), penetrates the muscularis propria (middle of figure), and runs tortuously through the submucosa (right of figure, *open arrow*). (From ref. [6].)

Therapy, Upper Gastrointestinal Lesion

Most patients require vigorous fluid resuscitation and transfusion of three or more units of packed erythrocytes to treat the severe bleeding. The lesion is not related to gastric acidity, and gastric acid suppression has no proven therapeutic role.

Endoscopic therapy for this point source of bleeding is particularly attractive, and fatal hemorrhage may occur without it. The most experience has accumulated with endoscopic injection therapy with epinephrine or polidocanol, [13] which has been shown to achieve hemostasis in 27 of 28 patients. [21] There may be a higher risk for gastric perforation from sclerotherapy for a Dieulafoy lesion than for a gastric ulcer [22] owing to a lack of inflammation and fibrosis around a Dieulafoy lesion. [6] Other endoscopic therapies include bipolar electrocoagulation, thermocoagulation, laser photocoagulation, endoscopic band ligation, and hemoclips. [23], [24] and [25] In three reviews, involving 79 to 111 patients, long-term hemostasis was achieved in about 85% of patients by various endoscopic therapies. [6], [10], [26] Savides and Jensen recommend endoscopic epinephrine injection to reduce lesion vascularity before electrocoagulation with a Gold probe. [27]

Surgery was the treatment of choice before the development of endoscopic therapy, but it is currently restricted to patients who fail endoscopic therapy. Surgical therapy has included electrocoagulation with vessel ligation, wedge resection, or proximal partial gastrectomy. Bleeding may recur after electrocoagulation or limited wedge resection because of the long span of the abnormal artery. [28] On the other hand, gastrectomy is theoretically unnecessary because the abnormal artery is usually restricted to the proximal stomach. [29] A wide wedge resection is a reasonable compromise between resecting too little and resecting too much. [7], [15]

Therapeutic angiography with selective left gastric artery embolization before surgery for ongoing bleeding is supported by anecdotal evidence, but the hemostasis may only be transitory. [30] Although this lesion used to have a grave prognosis, the mortality rate decreased to about 25% with the advent of endoscopic therapy in the 1980s [7] and has recently declined further. [26]

Lower Gastrointestinal Lesion

Dieulafoy lesion of the jejunum, ileum, colon, and rectum has been increasingly reported. In a recent review, this lesion occurred in 0.3% of about 3000 patients with gastrointestinal bleeding. [31] The lesion also tends to bleed suddenly and severely, [32], [33], [34] and [35] manifesting as bright red blood per rectum, rather than melena, with hemodynamic instability. [31]

Dieulafoy lesions of the jejunum, ileum, and colon are diagnosed preoperatively by endoscopy or angiography. [36] Lesions are more commonly detected in the colon and rectum than the jejunum and ileum, probably because of the more frequent use of colonoscopy than enteroscopy and the easier identification of lesions at colonoscopy because of a wider endoscopic field. The incidence of jejunal and ileal lesions may therefore be underestimated. [37] The colonoscopic diagnosis is often complicated by severe lesion hemorrhage, which increases the risk and decreases the yield of colonoscopy. [38] However, rectal lesions are readily diagnosed by sigmoidoscopy. [38], [39] The endoscopic appearance and histology of these lower gastrointestinal lesions are similar to those of gastric lesions. [37]

Surgery has been the traditional therapy for these lower gastrointestinal lesions, [38], [39] but colonoscopic hemostasis is being increasingly attempted, with the same modalities as are used for gastric lesions. Numerous small case studies suggest that endoscopic therapy should be initially performed for a bleeding colonic lesion and surgery reserved for refractory bleeding. [31], [39] The colonic Dieulafoy lesion is not a polyp and should not be removed by snare polypectomy because this can cause massive postpolypectomy hemorrhage. [40]

EHLERS-DANLOS SYNDROME

The Ehlers-Danlos syndrome is a rare heterogeneous group of about 10 genetic disorders of collagen metabolism that cause skin hyperextensibility, articular hypermobility, and tissue fragility. The syndrome is diagnosed by the characteristic clinical presentation, family pedigree analysis, and demonstration of genetic or biochemical defects. [41] Type IV is the most commonly associated with vascular lesions. [41], [42] Patients with type IV disease have thin transparent skin, wide thin (papyraceous) cutaneous scars overlying bony prominences, and cutaneous ecchymoses from spontaneous arterial rupture due to vascular and perivascular connective tissue friability. [43], [44] They have an increased risk of intramural intestinal hematomas, [43] colonic diverticular bleeding, [43], [45] gastrointestinal bleeding, [46], [47] and rarely intestinal perforation [43], [48] due to dissection or rupture of intestinal arteries. The syndrome does not predispose to peptic ulcer disease, but patients with this syndrome who have coincidental peptic ulcers often develop severe bleeding due to the weakness of perivascular supportive tissue. [41] Type IV disease is diagnosed by biochemical analysis of collagen. [46] Type IV disease carries a high mortality rate from repeated gastrointestinal bleeding. [47] Gastrointestinal surgery produces poor results owing to tissue fragility and a bleeding diathesis.

PSEUDOXANTHOMA ELASTICUM

Pseudoxanthoma elasticum is a rare genetic disorder of elastin synthesis characterized by yellowish xanthomatoid papules (pseudoxanthomas) on the skin, angioid streaks in the ocular fundi, and intermittent claudication and weak peripheral pulses due to vascular occlusion. [49] Gastrointestinal bleeding occurs frequently; a rate of 13% has been observed in 200 patients. [49] The hemorrhage typically commences in young adulthood. [49], [50] Most commonly, patients bleed from a characteristic lesion: numerous petechiae or erosions as well as yellowish xanthomatoid nodular lesions that resemble the cutaneous lesions are predominantly found in the gastric fundus. [51], [52], [53] and [54] Bleeding is attributed to elastin degeneration in small gastric arteries (Fig. 130-4) and is frequently severe and recurrent. [55] The bleeding arterioles do not vasoconstrict normally in response to hemorrhage owing to intimal calcification and elastin degeneration and are refractory to available pharmacological therapy. [52] The lesions also may not be amenable to endoscopic therapy because of their diffuseness. Gastrectomy is the primary therapy.

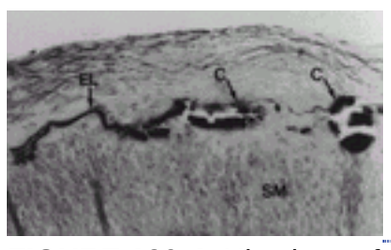

FIGURE 130-4. Histology of pseudoxanthoma elasticum. An elastin stain of a cross section of the proximal stomach from a patient with pseudoxanthoma elasticum, who underwent total gastrectomy for massive refractory gastric bleeding, reveals a submucosal gastric artery with focal intimal thickening, fragmentation of the elastin lamina (EL), and intramural calcifications (C). SM, smooth muscle; L, lumen. (From ref. [52].)

ANGIODYSPLASIA

Epidemiology

Angiodysplasia is an important cause of acute and chronic gastrointestinal bleeding, particularly from the lower gastrointestinal tract. About 1% of asymptomatic healthy subjects have colonic angiodysplasia, [56] whereas about 3% to 6% of lower gastrointestinal bleeders have angiodysplasia. [57], [58], [59] and [60] The frequency of asymptomatic upper gastrointestinal angiodysplasia in healthy subjects is unknown, but angiodysplasia accounts for about 2% to 5% of acute upper gastrointestinal bleeding. [61], [62] and [63] About 3% to 5% of gastrointestinal bleeding arises from the jejunum and ileum, [64] and angiodysplasia is the most common cause of bleeding in this location. [64], [65] and [66] The recognition of angiodysplasia has increased markedly because of greater use of endoscopy, causing the incidence to increase more than sixfold during the 1980s at one university hospital. [67] Angiodysplasia is increasingly recognized also in the jejunum and ileum with greater use of enteroscopy. [64], [65]

The incidence is unaffected by sex or race. [60], [67] Angiodysplasia is not familial but may be confused with hereditary hemorrhagic telangiectasia (HHT), which is familial. Patients with bleeding from colonic angiodysplasia are generally elderly, with an average age of about 70 years, [58], [61], [67], [68] and [69] but it occasionally occurs in infants, children, and young adults. [70]

Pathology

Colonic angiodysplasia occurs most commonly in the right colon, especially the cecum, [71] but up to 40% of colonic lesions occur distal to the hepatic flexure. [56], [72], [73] The predilection for the cecum is explained by the higher cecal mural tension owing to its larger lumenal diameter, according to the LaPlace law. [74] Upper gastrointestinal lesions occur most commonly in the stomach, sometimes in the duodenum, and rarely in the esophagus. [61], [67], [71], [75]

Angiodysplasias are often multiple [76] and tend to be clustered [69] (Fig. 130-5; see also Color Fig. 130-5). Despite clustering, 15% to 20% of patients with colonic angiodysplasia have upper gastrointestinal angiodysplasia, and vice versa. [69], [77]

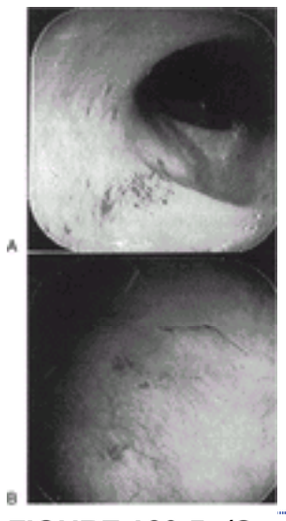

FIGURE 130-5. (See Color Fig. 130-5.) Endoscopic photo of cluster of angiodysplasia. **A:** Colonoscopic videophotography demonstrates two adjunct clusters of angiodysplasia in the descending colon in an elderly female with iron deficiency anemia; colonoscopy revealed no other angiodysplasia in the rest of the colon. **B:** Close-up views demonstrate the characteristic endoscopic appearance of angiodysplasia: an intensely red color, an intricate reticulonodular structure, and communication with prominent feeding or draining vessels. (From ref. [69].)

Unlike the lesions of HHT, which occur in all layers of the bowel wall, angiodysplasia occurs in the mucosa and submucosa. [64] Histologically, angiodysplasia consists of dilated, distorted, tortuous, and thin-walled vessels lined by endothelium with no or little smooth muscle and no inflammation, fibrosis, or atherosclerosis. [61], [68], [78] Initially, submucosal veins are dilated, but in older lesions, mucosal veins and capillaries and even arteries become dilated [74], [78] (Fig. 130-6). Congenital arteriovenous malformations, in contrast to angiodysplasia, typically have thick-walled arteries.

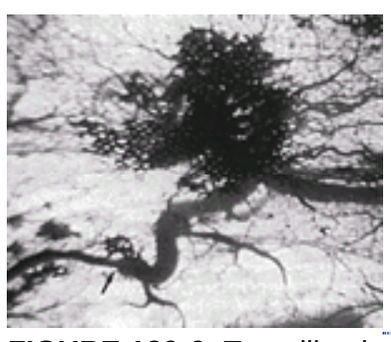

FIGURE 130-6. Transilluminated view of angiodysplasia. Photomicrograph through a dissecting microscope of a transilluminated, cleared colon after intravascular silicone injection reveals a mucosal ectatic vascular tangle surrounded by normal colonic crypts. The vascular tangle is drained by ectatic venules that lead to a large, distended, and tortuous underlying submucosal vein (*arrow*). (From Boley SJ, Brandt LJ, Mitsudo SM. Vascular lesions of the colon. Adv Intern Med 1984;29:301.)

Associations and Pathophysiology

Angiodysplasia is reported as the most common cause of upper gastrointestinal bleeding (19% to 32%) in patients with *chronic renal failure,* as compared with only about 3% to 5% in the general population. [79], [80], [81] and [82] Similarly, colonic angiodysplasia is a frequent cause of lower gastrointestinal bleeding in patients with chronic renal failure. [83]

The association with chronic renal failure is, however, somewhat controversial because several studies report no association. [84], [85] and [86] The reported discrepancies could arise from several sources. First, the risk of angiodysplasia appears to increase with the duration and severity of renal dysfunction, [79] and studies showing no association may include patients with a short duration and mild severity of renal insufficiency. Second, many studies that show a lack of an association were published in the early 1980s when angiodysplasia was an obscure diagnosis. Third, the presence of an association may be artifactual: coagulopathy associated with renal insufficiency may promote bleeding from and detection of otherwise clinically silent angiodysplasia. [79] Alternatively, a coagulopathy would promote erosions and ecchymoses during endoscopy that could be misinterpreted as angiodysplasia. In conclusion, angiodysplasia appears to be associated with chronic renal failure, but a prospective, controlled, investigator-blinded endoscopic study is needed to resolve the controversy.

The pathophysiology of this apparent association is speculative. Chronic, intermittent fluid overload in patients with renal failure may result in venous hypertension, microvascular venous dilation, and retrograde capillary dilation. [81] Renal failure patients might be older and have asymptomatic angiodysplasia, which could clinically manifest as a result of the bleeding diathesis associated with renal failure. [82] Hypertension and vascular disease, common in renal failure, could also augment bleeding from angiodysplasia. [61]

Several case reports have suggested that congenital or acquired *von Willebrand disease* may be associated with gastrointestinal bleeding from angiodysplasia. [87] Duray and associates reported two patients and reviewed eight other cases in the literature, [88] but Gostout and colleagues failed to find any cases of von Willebrand disease in a prospective study of 22 patients, published as an abstract. [89]

After an association between *aortic stenosis* and unexplained gastrointestinal bleeding was proposed in 1958, [90] several retrospective studies showed that up to 25% of patients with angiodysplasia had aortic stenosis. [71], [74], [76], [80], [91], [92] In many of these studies, aortic stenosis was diagnosed by clinical criteria without confirmation by echocardiography or cardiac catheterization. [93] An association was further strengthened by several case reports of cessation of bleeding from angiodysplasia after successful aortic valve replacement. [94], [95] and [96] In two recent prospective endoscopic studies of 29 to 40 patients with gastrointestinal angiodysplasia, none had aortic stenosis detected by two-dimensional Doppler echocardiography. [57], [97] Thus, an association is unproven. [98]

A number of case reports have noted an association between gastrointestinal angiodysplasia and *scleroderma* or the *CREST syndrome* (calicinosis cutis, Raynaud phenomenon, esophageal dysfunction, sclerodactyly, and telangiectasia). [99], [100], [101] and [102] Although this association has not been proven in a large controlled trial, the known occurrence of cutaneous telangiectasia in scleroderma renders an association with gastrointestinal telangiectasia both credible and likely. Several cases of angiodysplasia have been reported in association with *Turner syndrome* [103], [104] and *portal hypertension*. [105], [106] These associations, however, are unproven.

The pathogenesis of angiodysplasia is unknown and may vary according to the associated condition and the age of onset. [107] A popular theory is that angiodysplasia is a degenerative lesion of aging caused by chronic intermittent low-grade obstruction of veins, capillaries, and arterioles that supply the mucosa [74] (Fig. 130-7). A neurovascular (or hormonal) mechanism could contribute to angiodysplasia. [94] In response to low-grade local hypoperfusion, sympathetic nerves may stimulate intestinal vascular smooth muscle relaxation to increase flow and reverse the hypoxia. With chronicity, local vascular overload, dilation, and ectasia pathologically develop. [94] This theory could explain the disappearance of angiodysplasia with intravenous meperidine injection (a problem with endoscopic diagnosis) [108] or cold water colonic perfusion [97] and reappearance with naloxone administration. Increased expression of angiogenic factors [109] and deficient production of type IV collagen [110] have also been proposed as pathogenetic factors. Conceivably, the same genetic lesion that occurs in germ cells in HHT, as discussed later, may occur focally in somatic cells in sporadic angiodysplasia. This hypothesis, although speculative, is easily tested by molecular genetic analysis of tissue from isolated angiodysplasia.

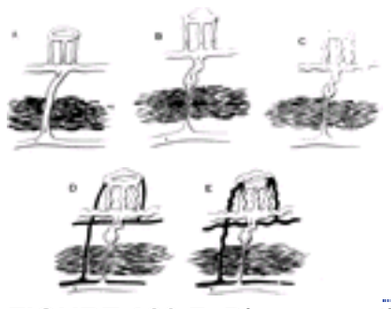

FIGURE 130-7. Diagrammatic illustration of a popular theory of angiodysplasia pathogenesis. **A:** Normally patent distal veins within the muscular layer of gut drain proximal mucosal vessels, including the capillary ring. **B:** These distal veins become partially obstructed due to muscular contractions or increased intralumenal pressure. **C:** Chronic repeated intermittent obstruction of these distal veins results in proximal venous dilation and tortuosity. **D:** With time, gut veins become progressively more dilated and tortuous, and the lesion propagates proximately. **E:** Ultimately, the capillary ring becomes dilated, the precapillary sphincter becomes incompetent, and a small arteriovenous communication occurs through the ectasia. (From ref. [74].)

Clinical Presentation

Bleeding from angiodysplasia is usually low grade and recurrent, and always painless. About 10% to 15% of patients present with acute severe hemorrhage. [91] The bleeding pattern may depend on the lesion stage, with more advanced lesions causing more frequent and more severe bleeding. [68] The clinical presentation depends on the bleeding site, chronicity, and severity. Acute upper gastrointestinal bleeding most commonly manifests as melena, and occasionally as hematemesis [67]; acute jejunal and ileal bleeding as melena [64]; and acute lower gastrointestinal bleeding as hematochezia, and rarely as melena. [61], [67]

Chronic bleeding presents with fecal occult blood or iron deficiency anemia. Patients bleeding from angiodysplasia tend to have a longer history of gastrointestinal bleeding than other patients owing to greater difficulty in diagnosis. [63], [111] Patients sometimes present with prior gastrointestinal bleeding without an endoscopic diagnosis, although this phenomenon is occurring less frequently with increasing endoscopic lesion recognition. [67], [107]

Physical examination is unrevealing except for signs of gastrointestinal blood loss. Although, in the past, angiodysplasia was associated with severe gastrointestinal bleeding, [71] angiodysplasia is increasingly associated with mild bleeding, probably owing to earlier identification and recognition of milder cases. [67]

Diagnosis

Endoscopy is the diagnostic procedure of choice. [72], [111] In a study of 80 patients, colonoscopy had a sensitivity of 81% when compared with angiography and pathologic examination of the resected colon. [112] Angiodysplasia may be missed behind folds, at sharp turns, or when covered by stool. True angiodysplasia can be carelessly dismissed as endoscopic erosions or fresh blood. [75] Angiodysplasia may be inconspicuous in a patient with hypotension or severe anemia and may become obscured after meperidine administration for endoscopy. [108] Although endoscopy should generally be performed after correction of anemia or hypotension with fluid resuscitation or blood transfusion, this principle is particularly important when angiodysplasia is suspected. When colonic angiodysplasia is suspected, complete colonoscopy with careful cecal examination is important. Although endoscopy is generally superior to barium studies in the evaluation of any cause of chronic gastrointestinal bleeding, this is particularly true when angiodysplasia is suspected because angiodysplasia is mucosal and macular and not visualized with barium contrast, and barium studies preclude subsequent angiography. [113]

Endoscopy is a fairly specific test provided strict diagnostic criteria are applied. At endoscopy, angiodysplasia appears as a dense macular and reticular network of vessels (vascular tuft), which is typically 2- to 8-mm wide and is intensely red because of the high oxygen content in erythrocytes within vessels supplied by arteries without intervening capillaries (Fig. 130-5 and Fig. 130-8; see also Color Fig. 130-5 and Color Fig. 130-8). Sometimes a prominent feeding artery or draining vein is observed. [57], [112], [114] Some endoscopists note a pale (anemic) mucosal halo surrounding angiodysplasia due to shunting of blood (vascular steal) from surrounding tissue by the low-resistance angiodysplastic shunt. [92]

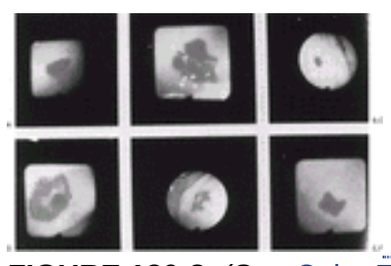

FIGURE 130-8. (See Color Fig. 130-8.) Spectrum of endoscopic appearance of upper gastrointestinal angiodysplasia. The characteristic endoscopic findings of angiodysplasia are intense erythema, well-demarcated margin, irregular stellate (fern-like) margin, fine internal reticular (fern-like) structure, and macular mucosal location. Lesions range from 2 × 3 mm to 5 × 7 mm in size. All angiodysplasia are from the stomach, except for **A,** which is from the duodenum. (From ref. [80].)

It is important to assess at endoscopy whether observed angiodysplasia is the source of blood loss. In two large studies of patients with angiodysplasia, 30% to 44% had other significant gastrointestinal lesions, such as colonic polyps, colon cancer, inflammatory bowel disease, or peptic ulcer disease. [59], [114] Bleeding is attributed to angiodysplasia when it is actively bleeding or has an overlying clot. Otherwise, the diagnosis is made only by exclusion of other causes.

The hot biopsy technique is preferred to reduce the risk of bleeding from endoscopic biopsy, but endoscopic biopsy is generally not recommended because of the low diagnostic yield and the increased risk of provoking hemorrhage. [61], [76] Pathological examination of an endoscopic biopsy supports the endoscopic diagnosis in only about 40% of cases because endoscopic biopsies are superficial, and the most consistent and characteristic histological abnormalities are submucosal.

The angiographic hallmarks of angiodysplasia are (1) a vascular tuft or tangle resulting from the local mass of irregular vessels, best visualized in the arterial phase; (2) an early and intensely filling vein resulting from a direct arteriovenous connection without intervening capillaries; and (3) persistent opacification beyond the normal venous phase (slowly emptying vein), possibly resulting from venous tortuosity [115] (Fig. 130-9). About 60% to 90% of patients have each of these angiographic signs. [73], [115] Angiodysplasia bleeds only intermittently, and extravasation of contrast material from angiodysplasia is detected in only about 10% of angiograms. [68] Angiography has the advantage of detecting lesions of the small intestine as well as colonic angiodysplasia and can be performed during severe hemorrhage when colonoscopy is ineffective. [61] Small intestinal angiodysplasia can, however, be missed at angiography owing to misidentification as normal vascular arcades. [116] Recently, helical computed tomographic angiography has been proposed as a minimally invasive technique to image colonic angiodysplasia. [117]

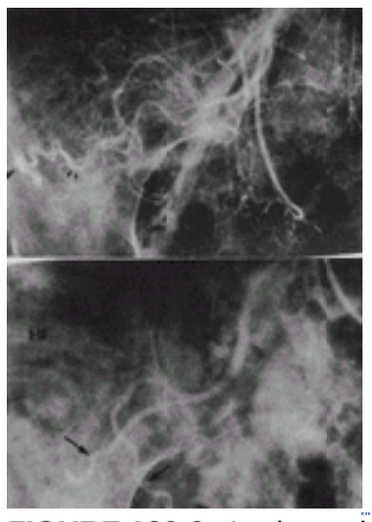

FIGURE 130-9. Angiographic findings in angiodysplasia. **Top:** Arterial phase shows a vascular tuft (*large arrow*) and two early filling veins (*small arrows*) that have filled with contrast before other veins. **Bottom:** Late phase reveals two densely opacified, dilated, and tortuous cecal veins (*arrow*) that still retain contrast after other veins have cleared. (From ref. [115].)

Angiodysplasia is seldom detected at laparotomy by visual inspection or by manual palpation of the serosal surface of gut because of its small size and mucosal location. [61] Richardson and colleagues [73] correctly identified angiodysplasia in only one of 39 laparotomies. hus, preoperative diagnosis by endoscopy or angiography is critical to avoid blind bowel resection for gastrointestinal bleeding. Intraoperative enteroscopy can help localize angiodysplasia to minimize the extent of small bowel resection. [118]

Differential Diagnosis

At endoscopy, angiodysplasia can be differentiated from other red lesions, such as gastritis, Barrett esophagus, inflammatory bowel disease, tumor, phosphosoda enema-induced lesions, and other vascular lesions. Phlebectasia and hemangiomas are blue or wine-colored submucosal vascular masses that collapse with endoscopic air insufflation. Kaposi sarcoma presents as a violaceous submucosal nodule or mass. A Dieulafoy lesion is typically elevated with one visible vessel. GAVE is distinguished from angiodysplasia by a characteristic antral location and a characteristic arrangement of red lesions on longitudinal ridges. Portal gastropathy is distinguished from angiodysplasia by a distinctive pale reticular border surrounding polygonal intensely erythematous patches located in the proximal stomach.

The endoscopic appearance of gastrointestinal telangiectasias of HHT may be identical to that of angiodysplasia, except that the lesions of HHT tend to be greater in number, occur in all bowel wall layers, and occur in all bowel segments. Patients with HHT may also have cutaneous telangiectasia, a positive family history, and a history of epistaxis. The diagnosis of scleroderma, CREST, or renal failure in association with angiodysplasia is based on the clinical manifestations of these conditions. The endoscopic findings in radiation colitis, aside from telangiectasia, include fibrosis, exudation, erosions, and mucosal friability, findings not compatible with angiodysplasia. [119]

Red mucosal lesions at endoscopy resembling angiodysplasia can be created iatrogenically by nasogastric aspiration or by endoscopic abrasion or suction. Lesions from nasogastric aspiration are round, colinear, equidistant, and relatively uniform in size, owing to the geometric arrangement of the nasogastric tube apertures, and are at the same stage of lesion evolution because of their simultaneous creation. [120] Lesions of endoscopic trauma are not seen initially during endoscopic intubation, but typically identified during endoscopic extubation. As compared with endoscopic trauma lesions, angiodysplasia has a finely reticular (fernlike) internal structure owing to a vascular tuft, an irregular (stellate) border as opposed to a round border with trauma, an abrupt lesion margin as opposed to an indistinct margin with trauma, and lesions lying flush (coplanar) with mucosa (see Fig. 130-5 and Fig. 130-8; see also Color Fig. 130-5 and Color Fig. 130-8). Thus, angiodysplasia is best diagnosed by detailed mucosal examination during endoscopic insertion with minimal use of suction. [91] The angiographic findings of angiodysplasia are occasionally mimicked by tumor or inflammatory bowel disease, [61] but these other conditions are differentiated by endoscopy.

Therapy

Endoscopic Therapy At EGD and colonoscopy, isolated, actively bleeding angiodysplasia is treated by endoscopic thermocoagulation, electrocoagulation, or photocoagulation. When bleeding, angiodysplasias are sometimes first injected with alcohol or epinephrine to slow the bleeding. These endoscopic therapies are relatively safe and highly successful at achieving short-term hemostasis when performed by an experienced endoscopist. Gostout and colleagues [77] reported cessation of bleeding in 72 of 83 patients (87%) after laser photocoagulation during a mean follow-up period of 12 months. These therapies are also relatively effective long term: among 47 patients receiving laser photocoagulation, 61% had no significant gastrointestinal bleeding during 54 months of follow-up. [121] Enteroscopic therapy may be similarly effective for accessible small intestinal angiodysplasias provided the technical expertise is available. [122] The major complication of therapy is gastrointestinal perforation which occurs in about 4% of cases. [77], [121] The highest risk for perforation is in the thin-walled cecum, the most common location of angiodysplasia. Laser therapy has the advantage of not requiring probe contact with the lesion, whereas the other endoscopic therapies entail a risk for probe adherence to the lesion after application. Laser therapy, however, has the important disadvantage of greater depth of tissue destruction and a possibly higher perforation rate. [107] This appears to be less of a problem with argon as compared with Nd:YAG lasers. [107] Most endoscopists prefer thermocoagulation or electrocoagulation over photocoagulation because of lower equipment costs and simpler technique. Recently, endoscopic band ligation has also been reported to be effective. [123]

Angiographic Therapy At angiography, active bleeding may be arrested by intra-arterial vasopressin infusion or by transcatheter embolization. This may obviate the need for surgery or may slow bleeding enough to permit elective rather than emergent surgical resection. Vasopressin infusion has an initial success rate of about 50% to 90%, with about a 33% chance of rebleeding. [124], [125] Although embolization used to produce a high rate of intestinal infarction, the frequency of this complication has recently decreased markedly as a result of improved catheter design and application of distal embolization. Two recent studies reported a high rate of hemostasis with embolization without complications. [126], [127] Angiography offers an alternative nonoperative therapy for jejunal or ileal angiodysplasia when enteroscopy is unavailable.

Surgical Therapy Surgery is reserved for severe bleeding from well-characterized and localized lesions that are refractory to endoscopic or angiographic therapy. Endoscopy of both the upper and lower gastrointestinal tract should be performed preoperatively to exclude distant synchronous angiodysplasia or other lesions. [69] Right hemicolectomy is indicated for refractory bleeding from cecal angiodysplasia. [91] Even if angiodysplasia were identified exclusively in the cecum, the entire right colon should be resected to ensure that no angiodysplasia is left behind. In right-sided angiodysplasia, none of the left colon should be removed even if colonic diverticulosis is present to avoid the increased morbidity and mortality of subtotal colectomy. [91] This resection strategy yields a significant but manageable long-term risk of postoperative rebleeding. In one study, 6 of 16 patients (37%) rebled after right hemicolectomy during an average follow-up of 3.6 years [71]; in another study, 4 of 17 patients (24%) rebled. [111] Early rebleeding, which accounts for half or more of the cases, is usually due to unappreciated lesions in the remaining colon and is usually successfully treated by subtotal colectomy. [91]

Medical Therapy After reports of their use to control epistaxis from intranasal telangiectasia in HHT, estrogen and progesterone have been used to control chronic recurrent gastrointestinal bleeding from angiodysplasia. These hormones are believed to promote vascular integrity. [128] In seven patients bleeding from angiodysplasia associated with renal failure, blood transfusion requirements decreased from 1.2 units/month to 0.21 units/month after instituting hormonal therapy. [129] In a study of four patients with angiodysplasia and six with telangiectasia from HHT, estrogen and progesterone decreased transfusion requirements from 10.9 to 1.1 units during a 6-month period. [130] However, in a study of 64 patients with small bowel angiodysplasia, no difference was noted in the rate of bleeding cessation or in the number of units transfused between patients treated with estrogen and progesterone versus untreated controls. [128] Hormonal therapy is currently reserved for patients with chronic bleeding from numerous or widespread angiodysplasias and from angiodysplasias in the jejunum or ileum that are relatively inaccessible to endoscopic therapy and in patients who either fail or cannot tolerate endoscopic or surgical therapy. [128], [131] Hormonal therapy can cause feminization in male patients. [132] Other side effects include vaginal bleeding, fluid retention, thromboembolism, and congestive heart failure. [133]

Natural History

In a retrospective comparison of angiodysplasia and other causes of gastrointestinal bleeding, patients with angiodysplasia had a milder hospital course with fewer transfusions of packed erythrocytes, shorter hospitalizations, and lower mortality rates. [67] Yet, patients occasionally have severe acute bleeding from angiodysplasia requiring surgery. Among 31 patients with angiodysplasia presenting with bleeding or anemia treated conservatively by blood transfusions alone, 26% rebled within 1 year and 46% within 3 years. [114] Thus, endoscopic therapy for angiodysplasia should be performed for gastrointestinal bleeding, even when the bleeding manifests as fecal occult blood or mild chronic anemia, because of the significant risk of rebleeding. In patients with numerous lesions, the endoscopist should concentrate on the largest and most accessible lesions, especially on those with endoscopic stigmata of recent hemorrhage.

Many angiodysplasias are asymptomatic and incidentally found at endoscopy, as, for example, during screening for colon cancer. Elderly patients with no history of bleeding frequently have small angiodysplasias detected in resected colonic specimens removed for unrelated colonic disease. [74] In a review of 41 patients with upper gastrointestinal angiodysplasias, 21 (51%) were incidental endoscopic findings. [92] Management guidelines for asymptomatic angiodysplasia should depend on their risk of subsequent bleeding. In two series of 9 or 15 subjects with asymptomatic angiodysplasia, none experienced gastrointestinal bleeding during a mean follow-up of 2 to 3 years. [56], [114] These findings suggest a conservative approach for asymptomatic angiodysplasia. [61], [134]

HEREDITARY HEMORRHAGIC TELANGIECTASIA

The term telangiectasia denotes vascular ectasia, which is part of a systemic disease or syndrome, in contradistinction to angiodysplasia, which is nonsyndromic. [3] Gastrointestinal telangiectasia, which appears nearly identical to angiodysplasia, is associated with HHT, scleroderma, CREST syndrome, and possibly Turner syndrome. CREST and Turner syndrome were aforementioned with angiodysplasia.

HHT, a disease with autosomal dominant transmission, produces a syndrome of multiple cutaneous telangiectasias, especially on the face, lips, tongue, oral mucosa, and hands, together with multiple mucosal telangiectasias, especially in the nose and gastrointestinal tract (see Chapter 49). The cutaneous lesions are usually a minor cosmetic problem, whereas the nasal and gastrointestinal lesions frequently bleed significantly and repeatedly. [135] This bleeding tendency is explained by the thin and fragile vascular wall that lacks an elastic lamina or muscle layer, vulnerability to trauma because of vessel superficiality, and perhaps intralesional venous hypertension due to arterial shunting of blood. The syndrome is caused by at least two different genetic mutations that impair blood vessel growth and repair, resulting in irregular, tortuous blood spaces lined by a thin single layer of endothelial cells. [136]

The disease prevalence is about 10 per 100,000 population. [137] Both sexes are affected equally. [138], [139] More than 80% of patients experience epistaxis, spontaneously or after minimal trauma, which typically begins during the teenage years. [137] About 25% of patients with epistaxis require blood transfusions. [133]

About 25% of patients experience clinically significant gastrointestinal bleeding, which typically begins during middle age. [140], [141] Upper gastrointestinal bleeding is more common than lower gastrointestinal bleeding because upper gastrointestinal telangiectasias are more common. [141] Gastrointestinal bleeding typically manifests as melena and occasionally as hematemesis. The differential diagnosis of melena in these patients includes swallowed blood from epistaxis. The gastrointestinal bleeding is typically painless, chronic, episodic, and progressive. [139] Among 25 patients with gastrointestinal bleeding, one half required blood transfusions, and one fourth required transfusion of more than 6 units. [140]

About 15% of patients have pulmonary arteriovenous malformations or fistulae, which can cause hypoxemia or hemoptysis, [142] and about 10% have cerebrovascular malformations, which can cause a hemorrhagic stroke or brain abscess. [143] Symptomatic liver involvement is uncommon but important in that it can cause high-output heart failure, portal hypertension, biliary disease, or liver failure. [144], [145]

The diagnosis is straightforward in patients with the triad of telangiectasia, recurrent epistaxis, and a compatible family history. [146] The cutaneous telangiectasias, unlike spider nevi, only partly blanch by diascopy because of vessel tortuosity. The site and source of gastrointestinal bleeding is diagnosed by endoscopy. The endoscopic appearance of telangiectasia resembles that of nonsyndromic angiodysplasia and resembles the cutaneous telangiectasia occurring in this syndrome. Lesions tend to be widespread throughout the gastrointestinal tract. As for angiodysplasia, other causes of gastrointestinal bleeding should be excluded at endoscopy, [139], [141] barium contrast radiography should not be used for diagnosis, [139] and angiography may be useful to diagnose the extent of bowel telangiectasia. [147]

The therapy resembles that for nonsyndromic angiodysplasia, with endoscopic thermocoagulation, electrocoagulation, or photocoagulation, [77], [121] but treatment is complicated by lesion multiplicity, dissemination, and progression. Isolated actively bleeding telangiectasias are usually successfully treated, but patients often rebleed from other, untreated gastrointestinal telangiectasias and, therefore, require multiple endoscopic sessions. [148] Segmental bowel resection is reserved for active, severe, and well-localized bleeding refractory to medical or endoscopic therapy because of the tendency to rebleed subsequently from telangiectasias at other sites. [111], [114], [138] When available, angiographic embolization can obviate the need for surgery.

In theory, systemic or generalized therapy should be favored over local therapy because of the typically large number and widespread extent of the telangiectasias. A few studies have reported a markedly decreased incidence in chronic gastrointestinal bleeding after estrogen and progesterone therapy. [130], [149]

GASTRIC ANTRAL VASCULAR ECTASIA

Clinical Presentation

GAVE is uncommon, reported in only three cases among 10,000 consecutive endoscopies in 1984. [150] The reported incidence appears to have increased somewhat since then owing to greater lesion recognition. The mean age at presentation is 70 years (range, 50 to 90 years), [151], [152] and [153] with a female-to-male ratio of about 5:1. [150], [152], [153] and [154] Patients typically present with iron deficiency anemia from chronic occult gastrointestinal bleeding [150], [153], [155] but occasionally have melena from acute bleeding. [151], [152] Hematemesis is unusual. The occult bleeding and anemia may be longstanding because of delayed diagnosis. [152] The lesion is painless. Physical examination reveals pallor and fecal occult blood. [153]

Diagnosis

Endoscopy reveals multiple parallel prominent longitudinal folds that traverse the antrum and converge to the pyloric sphincter. The folds contain intensely erythematous linear streaks at their apices [150] (Fig. 130-10; see also Color Fig. 130-10). Occasionally, the streaks extend into the proximal stomach. [156], [157] The term *watermelon stomach* derives from the resemblance of these erythematous linear streaks to the stripes on a watermelon rind. Despite lesion vascularity, endoscopic biopsy appears safe in that it can cause slight but not severe bleeding. [150], [158] Histological analysis of endoscopic biopsy samples taken from the apices of the longitudinal folds reveals hypertrophied mucosa, dilated and tortuous mucosal capillaries that are often occluded by bland fibrin thrombi, and dilated and tortuous submucosal veins. [150], [158], [159] Computed tomography may reveal focal mural thickening of the gastric antrum. Double-contrast upper gastrointestinal series may reveal prominent, scalloped antral folds radiating to the pyloric sphincter. [160] Angiography may reveal mild antral hypervascularity. [161] All these radiologic procedures are seldom necessary because the endoscopic appearance and endoscopic biopsy findings are generally diagnostic.

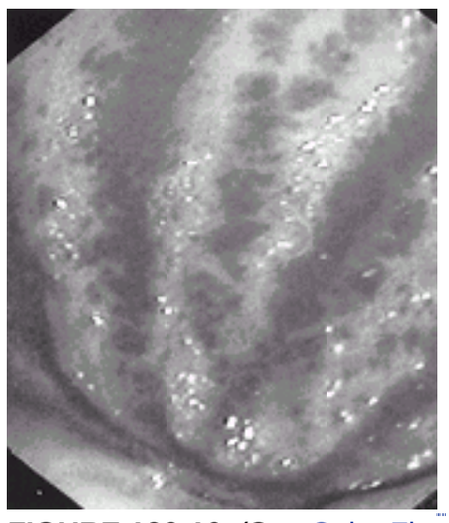

FIGURE 130-10. (See Color Fig. 130-10.) Videophotograph shows the characteristic endoscopic finding in gastric antral vascular ectasia (GAVE) of linear, intensely erythematous lesions at the apices of longitudinal antral folds radiating to the pylorus. The alternative lesion name of *watermelon stomach* derives from the resemblance of these erythematous linear streaks to the stripes on a watermelon rind. (From Clouse RE. Vascular ectasias, tumors, and malformations. In: Yamada T, Alpers DH, Laine L, Owyang C, Powell DW, eds. Atlas of gastroenterology. 2nd ed. Philadelphia: Lippincott Williams & Wilkins, 1999:550.)

Etiology

The etiology is unknown. The disease is associated with gastric hypochlorhydria, [154] cirrhosis, [162], [163] bone marrow transplantation, [164] and scleroderma, [157], [165] but these associations do not appear to provide a clue to the etiology. A trophic etiology has been proposed [166] because of the finding of elevated blood or mucosal tissue concentration of gastrin, [167] prostaglandin E2, [168] and 5-hydroxytryptamine, [169] but these abnormalities have not been conclusively proven. [162]

Alternatively, the lesions may arise from repeated antral mucosal trauma from pyloric prolapse. [150] The lesions are generally restricted to the antrum, the only gastric region subject to peristaltic contractions, and generally occur on longitudinal folds, the antral area subjected to the most trauma during peristaltic contractions. [153] Affected antral mucosa appears to be abnormally mobile and loosely attached to the underlying muscularis propria, as would be expected if mucosa had been chronically pulled from the underlying muscularis propria. [150] The vascular ectasia resembles that seen in other traumatic gastrointestinal lesions, such as the solitary rectal ulcer syndrome or prolapsed hemorrhoids. [153], [170] Cirrhotic patients might develop the lesion as a result of antral dysmotility. [171]

Therapy

Patients require transfusion for acute gastrointestinal bleeding and iron replacement for chronic bleeding. The underlying lesions may be treated by medical, endoscopic, or surgical therapy. Estrogen and progesterone therapy has been successfully used in anecdotal reports. [172], [173] Even though antrectomy removes the lesion and cures the disease, [150], [154] endoscopic therapy is usually attempted first because it has lower morbidity and mortality. Antrectomy is reserved for patients with refractory bleeding. [174] Endoscopic lesion ablation requires multiple endoscopic sessions because of the extensiveness of the lesion. Laser therapy has been most commonly used because it requires fewer endoscopic sessions than other modalities, [152], [157], [174], [175] but the heater probe has also been successfully used. [153] From 87% to 100% of patients have stable hematocrits without transfusions for up to 2 years after endoscopic therapy. [152], [157], [176] Endoscopic complications, such as perforation, stenosis, ulceration, and hemorrhage, are infrequent. [151], [152] Transjugular intrahepatic portosystemic shunt does not stop bleeding from GAVE in cirrhotic patients. [177]

HEMANGIOMAS

Capillary and Cavernous Hemangiomas

Intestinal hemangiomas are rare, with an incidence of about 1 per 15,000 patients. [178] About one half of these hemangiomas are associated with cutaneous hemangiomas, most commonly in the blue rubber bleb nevus syndrome and rarely in Klippel-Trenaunay syndrome, as described later. [179] Intestinal hemangiomas are either capillary, cavernous, or mixed. Capillary hemangiomas consist of small (capillary-sized), thin-walled, closely packed vessels; cavernous hemangiomas consist of large, dilated, blood-filled, thin-walled vessels; and mixed hemangiomas contain both types of vascular lesions. [180], [181] Capillary hemangiomas tend to be small and well circumscribed, whereas cavernous hemangiomas are often large and poorly circumscribed. In both types of hemangiomas, vessels are lined by well-differentiated, flattened, and hyperplastic endothelial cells. Capillary hemangiomas tend to be located in the perianal skin, whereas cavernous hemangiomas tend to be located in the rectum and distal colon. [4], [182]

Intestinal cavernous hemangiomas are clinically more significant owing to their propensity to bleed because of their frequent large size and large blood volume. Thrombocytopenia due to platelet sequestration and consumption within large hemangiomas also predisposes to bleeding. [183], [184] Multiple lesions often occur within the gastrointestinal tract. [185]

Colonic cavernous lesions may be asymptomatic (10%) or may produce acute gastrointestinal bleeding that manifests clinically as hematochezia or melena (72%), [182], [186], [187] or chronic gastrointestinal bleeding that manifests clinically as iron deficiency anemia. [188], [189], [190] and [191] The bleeding is painless. [192] It is typically episodic, recurrent, and progressive. Bleeding usually begins in childhood or young adulthood but occasionally begins in old age. [193] Bleeding lesions are generally large and more likely to be cavernous than capillary hemangiomas. Gastrointestinal lesions occasionally intussuscept and cause gastrointestinal obstruction. [194], [195] Rectal hemangiomas may produce tenesmus and rectal urgency. [181], [196] Hemangiomas occasionally occur in other organs and manifest as hematuria, epistaxis, hemoptysis, or menorrhagia, according to their location. [190], [193], [197]

Abdominal radiographs demonstrate phleboliths from thrombosis and calcification within about 50% of hemangiomas [4], [198] (Fig 130-11A). Barium contrast studies typically reveal multiple polypoid filling defects in the stomach, small bowel, or colon but occasionally reveal an annular intestinal constriction [186], [197], [199], [200] and [201] (see Fig. 130-11B). Computed tomography may demonstrate a large, thickened mesentery containing large vacuoles next to an abnormal intestinal segment with a thickened wall. [4]

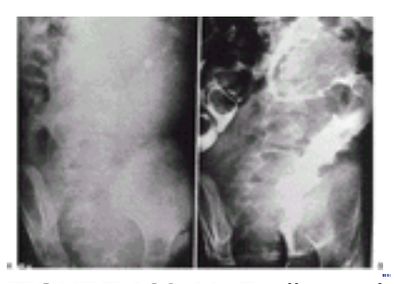

FIGURE 130-11. Radiographic appearances of intestinal hemangiomas. **A:** Abdominal radiograph of a 16-year-old white male patient with an extensive distal colonic cavernous hemangioma demonstrates a cluster of radiopaque phleboliths partly outlining the distal colonic wall. **B:** Barium enema demonstrates irregular narrowing in the rectosigmoid and “thumbprinting” in the sigmoid and descending colon caused by intramural hemangiomatous masses. Although the rectosigmoid narrowing resembles an apple-core lesion found in colon cancer, the radiographic findings of phleboliths and intramural masses point to the diagnosis of cavernous hemangioma. (From ref. [212].)

EGD and colonoscopy have high diagnostic sensitivity and specificity in patients with characteristic dermatologic syndromes (see later), but the lesion can be misdiagnosed at endoscopy in patients without the dermatologic syndromes. At endoscopy, intestinal hemangiomas appear as violet-blue, sessile, polypoid lesions. [189] (Fig. 130-12A, Fig. 130-12B; see also Color Fig. 130-12A, Color Fig. 130-12B). It is often difficult to determine at endoscopy which lesion bled because of lesion multiplicity and multifocality. [200] Endoscopic biopsy should be performed cautiously, and only if necessary, owing to the risk of profuse hemorrhage. [4] Pathological examination reveals typical findings of a capillary or cavernous hemangioma: numerous ectatic dilated blood-filled spaces lined by a thin layer of cuboidal endothelium and surrounded by scant fibrous stroma with occasional smooth muscle cells. [190] Lesions lack cellular atypia or dysplasia and do not undergo malignant transformation.

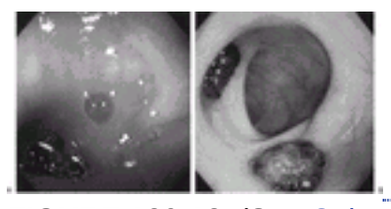

FIGURE 130-12. (See Color Fig. 130-12A, Color Fig. 130-12B.) Endoscopic appearance of colonic hemangiomas. **A:** Videophotograph of a 3-mm colonic capillary hemangioma adjacent to a mid-sigmoid diverticulum. **B:** Videophotograph from the colon shows the characteristic endoscopic appearance of a cavernous hemangioma as a bluish, round, smooth, and well-circumscribed sessile polypoid lesion. (**A,** Courtesy of T.G. Tietjen, M.D., Petoskey, Michigan; **B,** From ref. [206] .)

The differential diagnosis includes colon cancer, adenomatous polyps, hemorrhoids, and ulcerative colitis. [202] Colonic hemangiomas are distinguished from colonic polyps at endoscopy by their bluish color, typical submucosal location, compressibility, and poorly defined margins. [203] Rectal hemangiomas are distinguished from internal hemorrhoids by a nodular, as opposed to serpiginous, shape; submucosal versus superficial location; and extension beyond the anorectum. Colonic hemangiomas are differentiated from pseudopolyps of ulcerative colitis by the presence of a bluish color, submucosal location, and lesion compressibility and by the absence of mucosal granularity, of blunting of the vascular pattern, and of continuous involvement on colonoscopy. [179] The correct diagnosis is critical for proper management. Polypectomy may be inadvisable because of the risk for postpolypectomy hemorrhage from a cavernous hemangioma. Hemorrhoidectomy is ineffective therapy for bleeding from rectal hemangiomas. [202]

Patients typically require frequent blood transfusions for chronic gastrointestinal bleeding. Surgery for gastrointestinal bleeding is limited by the tendency for the lesions to be multiple and widespread and by the frequent evolution of new lesions. [204] Surgery is reserved for severe well-localized bleeding. [197] , [199] Recently, endoscopic hemostasis by coagulation, [187] , [205] band ligation, [206] or endoscopic polypectomy [204] , [206] has been reported without complications.

Blue Rubber Bleb Nevus Syndrome

The blue rubber bleb nevus syndrome, although rare, should be suspected when characteristic dermatologic lesions occur in conjunction with gastrointestinal bleeding (see Chapter 49). Patients present with multiple violet-blue, slightly raised, and elastic cutaneous hemangiomas resembling a rubber nipple [207] and ranging in size from 0.5 to 5 cm in diameter, together with multiple gastrointestinal hemangiomas, which are commonly located from the stomach through the rectum and rarely in the esophagus or oropharynx [188] , [194] , [199] (see Fig. 130-12A, Fig. 130-12B and Color Fig. 130-12A, Color Fig. 130-12B). The cutaneous lesion has a wrinkled surface and can be emptied of blood by manual pressure, leaving a wrinkled blue or white sac that slowly refills when the pressure is released. [189] , [193] Lesions usually begin in early childhood, and they tend to increase progressively in number and sometimes in size with age. [197] , [200] , [206] Most cases are sporadic, but several families have been reported with disease transmission in an autosomal dominant genetic pattern. [197] A candidate gene has been identified on chromosome 9. [208]

The clinical presentation, diagnosis, and therapy of the gastrointestinal lesions were aforementioned in the section “Capillary and Cavernous Hemangiomas.” Cutaneous lesions rarely require treatment because they rarely bleed and never become malignant. [199]

Intestinal Hemangiomas in Klippel-Trenaunay Syndrome

Klippel-Trenaunay syndrome is characterized by port-wine cutaneous hemangiomas, bony and soft tissue hypertrophy, superficial varicose veins, and abnormal—atretic, hypoplastic, or occluded—deep veins, usually affecting one lower extremity but occasionally affecting both lower extremities or one or both upper extremities [209] , [210] (Fig. 130-13). A family history of similar lesions is usually lacking. [209] In *Parkes-Weber syndrome,* patients additionally have arteriovenous fistulae.

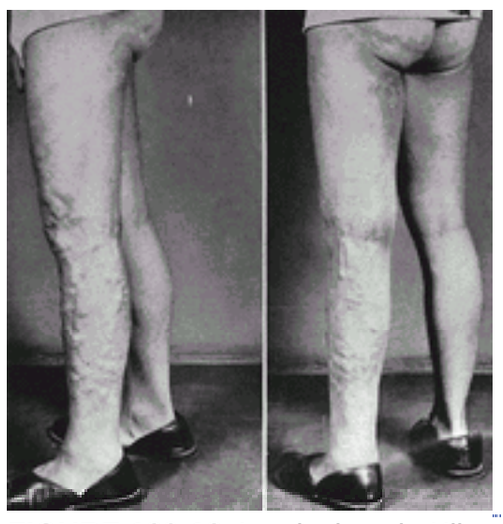

FIGURE 130-13. Leg lesions in Klippel-Trenaunay syndrome. A 23-year-old man has the characteristic physical findings in one lower extremity of superficial varicose veins (see left calf), an increased girth (compare size of ankles and calves in the two legs), an increased length, and a port-wine cutaneous hemangioma (see left calf). (From ref. [209] .)

This syndrome is associated with extensive infiltrative cavernous hemangiomas of the distal colon [211] , [212] , [213] and [214] and, rarely, the small intestine. [215] Patients present with chronic intermittent hematochezia and chronic iron deficiency anemia in childhood or young adulthood. [212] , [216] Thrombocytopenia due to platelet sequestration and consumption may contribute to the bleeding, [213] as may mesenteric venous hypertension due to the congenital obstruction of lower extremity veins and consequent overload of the internal iliac veins. [214] The frequency of rectal bleeding associated with this syndrome is poorly characterized [212] but reported as 1% to 13%. [214] , [217] Diagnostic features found on plain abdominal radiographs, barium enema, colonoscopy, computed tomography, and selective inferior mesenteric angiography are essentially the same as those for multiple cavernous hemangiomas (see earlier). [211] , [212] , [215] Abdominoperineal resection of the colonic hemangioma is the definitive therapy. Endoscopic hemostasis may provide a nonsurgical alternative therapy in the future.

Disseminated Hemangiomatosis

Rarely, patients present with more than 50 intestinal hemangiomas, which range in size from 2 to 20 mm, as part of a syndrome of disseminated hemangiomatosis involving three or more organ systems. [218] Patients typically present at birth with numerous cutaneous hemangiomas. Untreated patients have about a 60% mortality rate during infancy from massive gastrointestinal bleeding, hemorrhagic stroke, or high-output cardiac failure due to arteriovenous shunting. [219], [220] Corticosteroids and interferon-a have been successfully used to promote lesion regression. [219], [220]

Phlebectasias

Intestinal phlebectasias are venous varicosities consisting of a markedly dilated tortuous vein, with a normal vascular wall and scant connective tissue stroma unassociated with portal hypertension. [221] Endothelial cells within the vessel appear normal. They are generally classified as multiple, small hemangiomas, but this classification is somewhat controversial. [181], [183] They occur primarily in elderly men. [222] Phlebectasias may also occur in the oral cavity, mostly at the base of the tongue, where they are called caviar spots (varices) or sublingual phlebectasia, and in the genitalia, where they are called Fordyce lesions or scrotal phlebectasia. [221] These cutaneous and oral findings are clues to the presence of intestinal lesions in elderly men with gastrointestinal bleeding.

Intestinal lesions are characteristically dark bluish-black, range from several millimeters to 10 mm in size, are soft and compressible, blanch with pressure, are multiple, and are located in the submucosa. [222] Although usually asymptomatic and an incidental finding, they occasionally produce chronic occult or acute gross intestinal bleeding. [221], [222] and [223] Because of their location, small intestinal phlebectasias must be diagnosed by enteroscopy. [223] Angiography can occasionally demonstrate the site of bleeding by contrast extravasation but does not diagnose the specific lesions. [221] Surgery is currently the only definitive therapy but is reserved for severe ongoing bleeding because of the multiplicity and extent of the small bowel lesions.

ANGIOSARCOMAS

Angiosarcomas are rare, highly vascular malignancies derived from endothelial cells, representing less than 1% of sarcomas. They may be divided into hemangiosarcomas and lymphangiosarcomas. [224] They commonly arise in the skin and subcutaneous tissue, and rarely in the gut. [178] Chronic lymphedema and prior local radiotherapy are risk factors for cutaneous angiosarcomas, [225] and exposure to thorium dioxide (Thorotrast) during angiography, arsenic in insecticides, or polyvinyl chloride in manufacturing plastics are risk factors for hepatic angiosarcomas. [226]

Grossly, lesions may appear as a hemorrhagic mass. Histologically, angiosarcomas contain numerous interconnecting vascular channels (vasoformative structures) that are irregular in size and shape and contain atypical cells with some mitotic activity or sheets of pleomorphic cells that display prominent mitotic activity. Poorly differentiated angiosarcomas are differentiated by immunohistochemistry from poorly differentiated adenocarcinomas by the presence of factor VIII–related antigen. [227]

Patients are generally elderly. [228], [229] Lesions have been reported in the esophagus, stomach, small intestine, colon, and rectum. [178], [228], [229] and [230] Symptoms are similar to those with cavernous hemangiomas. [229], [230] Endoscopy may demonstrate an ulcerated mass or tight asymmetric stricture. Surgical resection is the standard therapy. Angiosarcomas are often multifocal at diagnosis and typically biologically aggressive. [228], [229] The prognosis appears to be poor as a result of early metastasis. [225]

HEMANGIOPERICYTOMAS

Hemangiopericytomas are uncommon, richly vascular neoplasms composed of pericytes, the mesenchymal cells normally closely apposed to endothelial cells. The stomach is the most commonly involved gastrointestinal organ, [231], [232] but the small intestine, [233], [234] and [235] colon, [236], [237] rectum, [238] and even mesentery or retroperitoneum [239] can be involved. The neoplasm occurs primarily in adults. Symptoms include gastrointestinal bleeding, abdominal pain, constipation, abdominal distention, nausea and vomiting, [231], [236], [237] and occasionally intestinal intussusception. [233], [234], [235] and [236] Large tumors are rarely associated with hypoglycemia. [240]

Barium studies typically reveal a smoothly contoured polypoid lesion. [231], [232] Angiography demonstrates a richly vascular mass with a diffuse capillary blush. [240] Endoscopy may reveal a solitary, fairly well-circumscribed submucosal mass or polyp covered by normal or ulcerated mucosa. Pathological examination of endoscopic biopsy specimens reveals tightly packed spindle-shaped neoplastic pericytes surrounding intercommunicating vascular channels of variable caliber and shape. [236], [241]

Most tumors are benign and slow growing, and many are bulky. [231], [236], [240] Malignancy is characterized histologically by increased cellularity, prominent mitotic activity, hyperchromatism, cellular pleomorphism, and foci of necrosis or hemorrhage. [240], [242] Wide local surgical resection is the primary therapy. [242]

HEMANGIOENDOTHELIOMAS

Hemangioendotheliomas are rare, borderline malignancies with a histological appearance between benign hemangiomas and malignant angiosarcomas. Hemangioendotheliomas display more cellularity and mitotic activity than hemangiomas but less mitotic activity and cellular atypia than angiosarcomas. In epithelioid hemangioendothelioma, the most important subtype, neoplastic endothelial cells focally line small extracellular vascular spaces or contain small intracellular spaces or vacuoles. [243] The tumor often arises from a vein, and like angiosarcoma, immunohistochemistry shows the presence of factor VIII–related antigen. Hemangioendotheliomas are sometimes locally invasive but rarely metastasize.

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CHAPTER 131

Julián Panés and Josep M. Piqué

INTESTINAL ISCHEMIA

Intestinal ischemia is not rare, accounting for about 1 in 1000 hospital admissions. [1] The increase in the incidence of this disorder in recent years is secondary to multiple factors, such as heightened awareness for the diagnoses, the advanced mean age of the population, and the increasing number of critically ill patients. Despite better insights into the pathophysiology of intestinal ischemia, these syndromes continue to have high morbidity, as evidenced by the greater than 60% rate of mortality from acute ischemia of the small intestine. [2] *Diagnosis before the occurrence of intestinal infarction is the most important factor in improving survival for patients with intestinal ischemia*. Thus, the objectives of this chapter are to provide knowledge of the basic anatomic and pathophysiologic mechanisms underlying intestinal ischemic damage, the guidelines for identification of patients who require prompt and aggressive evaluation, and delineation of the optimal diagnostic workup and type of therapy for each patient.

ANATOMY OF THE INTESTINAL CIRCULATION

Generally, the involved segment of ischemic intestine has a linear correspondence to the location and extent of vascular occlusion. Knowledge of the vascular anatomy is therefore crucial for the understanding of the pathophysiology, clinical presentation, and therapy of intestinal ischemia.

Embryology

The mesenteric vessels arise from the primitive ventral segmental arteries. As development proceeds, there is regression of all but three primitive communications, with only the precursors to the three major mesenteric vessels remaining. The 10th segmental artery gives rise to the celiac artery that supplies the foregut, the 13th artery gives rise to the superior mesenteric artery (SMA) to supply the midgut, and the 21st or 22nd artery gives rise to the inferior mesenteric artery (IMA) to supply the hindgut. Most variations in vascular anatomy can be traced to either persistence or incomplete regression of primitive segments. [3]

Celiac Artery

The celiac artery arises from the abdominal aorta at about the level of the T-12 or L-1 vertebral body. It usually courses anteriorly and slightly inferiorly 1 to 2 cm before branching into the left gastric, common hepatic, and splenic arteries. The left gastric artery supplies the stomach and provides small branches to the esophagus that anastomose with aortic esophageal branches, and small branches that anastomose with the right gastric artery, a branch of the hepatic artery. The common hepatic artery divides into the hepatic artery, which supplies the liver; the right gastric artery, which supplies the stomach along the lesser curvature; and the gastroduodenal artery, which divides into right gastroepiploic and superior pancreaticoduodenal arteries to supply the antrum, duodenum, and pancreas. The splenic artery supplies the spleen and body and tail of the pancreas. [4]

Superior Mesenteric Artery

The SMA arises from the aorta about 1 cm below the celiac artery, usually at the level of L-1. It courses inferiorly and toward the right to terminate at the level of the cecum as the ileocolic artery (Fig. 131-1A). The inferior pancreaticoduodenal artery is one of the first branches of the SMA; it supplies the pancreatic head, uncinate process, and descending duodenum, and anastomoses with the corresponding superior branches from the celiac axis. Generally, there are more SMA branches to the distal small bowel than to the more proximal portions, thus providing greater potential for distal anastomotic communication. The middle colic artery arises from the proximal SMA to supply the transverse colon and communicates with branches of the IMA. The right colic artery may arise as a common trunk with the middle colic artery or have a separate origin from the SMA; it supplies the distal ascending colon and right colonic (hepatic) flexure. The ileocolic artery is the terminal branch of the SMA and supplies the distal ileum, cecum, and ascending colon.

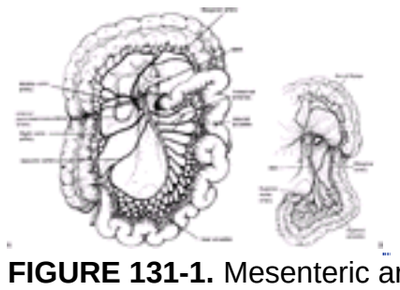

FIGURE 131-1. Mesenteric arterial circulation. **A:** The superior mesenteric artery (SMA) exits the aorta 1 cm below the celiac artery. It supplies the entire small intestine except for the superior part of the duodenum and supplies the right colon and part of the pancreas. Branches include the inferior pancreaticoduodenal artery, numerous jejunal and ileal branches, the ileocolic artery, and the middle colic artery. **B:** The inferior mesenteric artery (IMA) arises from the aorta 3 cm proximal to the aortic bifurcation. The artery branches into the left colic, several sigmoid (inferior left colic) arteries, and the superior rectal (hemorrhoidal) artery. The arcades of the SMA and IMA interconnect at the base and border of the mesentery. The connection at the base of the mesentery provides a potential collateral channel between SMA and IMA, called the *arc of Riolan*. The connection along the mesenteric border provides another potential collateral channel, called the *marginal artery of Drummond*. (From Marston A, Pegington J. Macroscopic anatomy. In: Marston A, Bulkley GB, Fiddian-Green RG, Haglund UH, eds. Splanchnic ischemia and multiple organ failure. London: Edward Arnold, 1989:13.)

Inferior Mesenteric Artery

The IMA is the smallest of the mesenteric vessels and arises from the ventral aorta about 6 to 7 cm below the SMA at the level of L-3 (see Fig. 131-1B). The artery branches into the left colic artery, which supplies the left (distal) transverse colon and splenic flexure; several sigmoid (inferior left colic) arteries, which supply the sigmoid and descending colon; and the superior rectal (hemorrhoidal) artery, which supplies a large part of the rectum. The inferior rectal (hemorrhoidal) arteries, derived from the internal pudendal arteries, and the small and inconstant middle rectal (hemorrhoidal) arteries, derived from the internal iliac arteries, also supply the rectum.

Arterial Collaterals

Extensive anastomotic and collateral circulation exists within each and among the three major intestinal vessels as well as between the mesenteric vessels and the nonmesenteric systemic circulation. Major branches of the celiac artery interconnect in the stomach and duodenum. A series of arcades interconnect neighboring branches of the SMA. [3] A similar series of arcades interconnect neighboring branches of the IMA. The primary potential pathway of collateral flow between the celiac artery and SMA is through the gastroduodenal and pancreaticoduodenal arteries (Fig. 131-2). As previously noted, there are extensive communications between the gastroduodenal artery and branches of the SMA, with the inferior pancreaticoduodenal being the most common. Additionally, omental branches of the SMA can communicate with branches of the celiac artery by the arc of Barkow. [4] An uncommon but well-described communication is the arc of Bühler, a direct communication between the celiac artery and SMA that represents a persistence of the embryonic ventral segmental arteries.

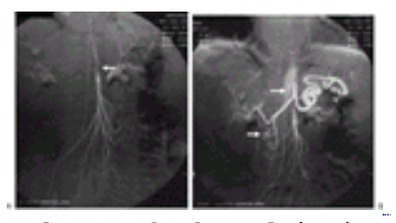

FIGURE 131-2. A: Selective digital subtraction angiography (DSA) of the superior mesenteric artery (SMA), showing a long and high-grade stenosis of the proximal artery (*arrow*). **B:** DSA injection of the celiac axis (*thick arrow*) of the same patient demonstrating collateral filling of the distal SMA through pancreaticoduodenal collaterals (*thin arrow*).

The arcades of the SMA and IMA interconnect at the base (away from the colon) and border (near the colon) of the mesentery (see Fig. 131-1B). The connection at the base of the mesentery provides a potential collateral channel between the SMA and IMA, called the *meandering mesenteric artery,* or *arc of Riolan*. [3] The connection along the mesenteric border provides another collateral channel called the *marginal artery of Drummond*.

Despite the presence of collateral channels, the vasculature has several points susceptible to poor perfusion (watershed areas). The middle part of the small intestine, in the middle of the area perfused by the SMA, is far away from collaterals from the celiac axis to the proximal SMA and from collaterals from the IMA to the distal SMA. This area is the most vulnerable to develop ischemia from SMA occlusion. Narrow terminal branches of the SMA supply the splenic flexure, and the rectosigmoid junction is supplied by terminal branches of the IMA. These two watershed areas are most vulnerable to develop ischemia during systemic hypotension if the collateral anastomoses are small or tenuous. [5]

Venous Anatomy

The venous system generally parallels the arterial distribution. The inferior mesenteric vein (IMV) drains the rectum through the superior rectal vein, the sigmoid through the sigmoid veins, and the descending colon through the left colic vein. The superior mesenteric vein (SMV) drains the small intestine, cecum, ascending colon, and transverse colon through the jejunal, ileal, ileocolic, right colic, and middle colic veins. [6] The SMV also drains the greater omentum and distal stomach with the right gastroepiploic vein and drains part of the pancreas and duodenum through the inferior pancreaticoduodenal vein. The IMV drains into the splenic vein, which then joins the SMV to form the portal vein.

MECHANISMS OF ISCHEMIC INJURY

When blood flow to the intestine decreases below a critical point, important metabolic and structural alterations appear in a variety of intestinal cells that can eventually cause cell destruction and death. Despite the fact that restitution of blood flow is necessary to limit the progression of cellular injury associated with decreased oxygen and nutrient delivery, it has become apparent that restoration of blood flow and oxygenation to the ischemic intestine can result in a paradoxical enhancement of tissue injury; a process that has been termed *reperfusion injury*. The mechanisms that are believed to participate in ischemic and reperfusion injury include the infiltration of postischemic tissues by inflammatory cells, an increased production of reactive oxygen species (ROS), alterations in vascular permeability, and inhibition of local cytoprotective mechanisms (Fig. 131-3). For a detailed description of the structure and functions of intestinal microcirculation under physiologic conditions, see Chapter 22, Gastrointestinal Blood Flow.

FIGURE 131-3. Mechanisms involved in ischemia-induced intestinal damage. Ischemia results in an increased production of oxidants, with a corresponding reduction in the synthesis of nitric oxide (NO) by endothelial NO synthase (eNOS). The enhanced generation of oxidants results in the activation of endothelial cells and leukocytes. Firm adhesion of leukocytes, which is mediated by ß2-integrins (CD11/CD18), is induced by engagement of activated leukotriene B_4 and platelet-activating factor with their receptors on rolling leukocytes. Sustained rolling and adhesion of leukocytes on endothelial cells are ensured by an oxidant-dependent synthesis of endothelial cell adhesion molecules, such as P-selectin and intercellular adhesion molecule 1. The inflammatory responses to ischemia are further amplified by oxidants derived from reduced nicotinamide adenine dinucleotide phosphate (NADPH) oxidase in leukocytes and by mediators released from mast cells and macrophages that normally reside in close proximity to postcapillary venules.

Inflammatory Response

Recruitment of circulating leukocytes toward the ischemic intestine is a key event in the pathophysiology of intestinal ischemic injury. Both leukocyte and endothelial adhesion molecules participate in slowing the leukocyte as it exits the capillary and enters the postcapillary venule, which is the major site of leukocyte–endothelial cell adhesion. [7] The initial event, known as the *rolling phenomenon,* consists of a transient adhesive interaction and is mainly mediated by the selectin family of adhesion molecules: L-selectin, expressed on all leukocytes; P-selectin, detected on activated platelets and endothelial cells; and E-selectin, exclusively found in the latter. Several studies have reported an important up-regulation of P-selectin in the intestine submitted to ischemia, [8], [9] and this molecule appears to play a key role in mediating leukocyte infiltration and organ dysfunction during low-flow states or transient interruption of intestinal blood flow. [10] In addition to P-selectin, L-selectin has also been implicated in the process of tissue infiltration and organ failure resulting from ischemia. [11], [12]

Firm leukocyte adhesion and eventual emigration to the perivascular interstitium are mediated mainly by the intercellular adhesion molecule 1 (ICAM-1), a constitutively expressed immunoglobulin located on endothelial cells, and by its counter-receptors on neutrophils, the ß $_2$-integrins CD11a/CD18 and CD11b/CD18. Endothelial expression of ICAM-1 has been found to increase in tissues subjected to ischemia [13], [14] and also in distant organs, suggesting a possible role for this molecule in the pathogenesis of multiple organ failure. [15] Moreover, the administration of an antibody against ICAM-1 or against the common ß $_2$ subunit CD18 significantly prevents leukocyte accumulation and microvascular dysfunction during reperfusion. [16] Other strategies, such as administration of ICAM-1 antisense oligonucleotide [17] or the disruption of ICAM-1 or CD18 genes, [18] also prevent ischemia-induced neutrophil accumulation. Trafficking of leukocytes in venules is a determinant of the extent of ischemia-induced endothelial barrier dysfunction. The magnitude of albumin leakage in postischemic venules is highly correlated with the number of adherent and emigrated leukocytes, and adhesion molecule–directed antibodies that effectively blunt leukocyte adherence and emigration also exert an attenuating action on ischemia-induced albumin leakage. [16]

Reactive Oxidants

The increased production of ROS by both endothelial cells and leukocytes in postischemic tissues appears to be a key determinant of reperfusion injury to the microvasculature. An important source of endothelial cell–derived superoxide and hydrogen peroxide is the enzyme xanthine oxidase. Intestinal endothelial cells are enriched with the enzyme xanthine oxidase. Ischemia promotes the conversion of the nicotinamide adenine dinucleotide (NAD)-reducing dehydrogenase form of the enzyme to the oxygen-reducing oxidase form. The accumulation of hypoxanthine during ischemia allows for a burst of superoxide and hydrogen peroxide production by the enzyme when oxygen is reintroduced into the blood vessel at the time of reperfusion. [19] Although xanthine oxidase contributes to the initial oxidant stress elicited in venules after a few minutes of reperfusion, adherent leukocytes account for the substantially greater oxidant stress that occurs thereafter. [20] This contention is supported by studies that demonstrate a diminished oxidant stress in postischemic venules of animals in which postischemic leukocyte recruitment is abrogated by means of blocking monoclonal antibodies directed against endothelial or leukocyte adhesion molecules. [19], [21]

Imbalance of Proinflammatory and Protective Mechanisms

It has been suggested that ischemia induces an acute inflammatory response and microvascular dysfunction by altering the balance between nitric oxide (NO) and superoxide production. There are several lines of evidence that implicate NO bioavailability as an important determinant of the inflammatory responses observed after ischemia. These include (1) a reduction in the activity of the Ca^{2+}-dependent isoform of NO synthase in postischemic tissues; (2) the ability of NO synthase inhibitors to mimic most of the microvascular alterations normally produced by ischemia, that is, leukocyte recruitment, platelet-leukocyte aggregation, mast cell degranulation, and an increased albumin leakage; and (3) the protection against ischemia-induced leukocyte recruitment and microvascular dysfunction that is afforded by NO-releasing compounds, which replenish tissue NO levels in postischemic tissues. [22], [23]

Under normal conditions, the flux of NO greatly exceeds the rate of superoxide production. This allows for NO to scavenge the low intracellular levels of superoxide. However, within minutes after reperfusion of ischemic tissues, the balance between NO and superoxide is tipped in favor of the latter. This imbalance results from a profound increase in the production of superoxide by endothelial cells and adherent leukocytes with a corresponding decline in the synthesis of NO from endothelial NO synthase. The relatively low levels of NO are insufficient to oppose leukocyte–endothelial cell interactions and to maintain optimal tissue blood flow.

The accumulation of superoxide that occurs in the absence of NO after ischemia allows for an enhanced generation of hydrogen peroxide. The two reactive oxygen metabolites (and H_2O_2) can rapidly initiate or exacerbate the inflammatory state in venules (1) by eliciting the production of platelet-activating factor (PAF) through phospholipase activation; (2) by mobilizing the stored pool of P-selectin to the endothelial cell surface, where it mediates leukocyte rolling; and (3) by activating nuclear transcription factors, such as NF-?B and AP-1, which in turn activate genes that encode adhesion molecules such as P-selectin, E-selectin, and ICAM-1 [23] (see Fig. 131-3).

INFLUENCE OF RISK FACTORS

It is often assumed that common risk factors for ischemic disease, including hypercholesterolemia, diabetes, and hypertension, render organs such as heart, brain, or intestine more susceptible to ischemia by increasing atherosclerosis-induced flow restriction in arterial vessels. However, there is a growing body of evidence suggesting that the risk factors for ischemic disease may also exert their deleterious action by enhancing the vulnerability of the microvasculature to the injurious effects of ischemia.

Hypercholesterolemia

Elevated levels of plasma cholesterol appear to exacerbate the dysfunctional responses elicited by ischemia in all segments of the microvasculature. Endothelium-dependent relaxation of arterioles is impaired in even mild hypercholesterolemia. [24] This response is reversed by superoxide dismutase, suggesting that an accelerated production of superoxide by arteriolar endothelial cells leads to inactivation of endothelial cell–derived NO. [25] This possibility is supported by measurements of an enhanced production of superoxide by arterial endothelial cells from hypercholesterolemic animals. [24] A larger number of rolling, adherent, and emigrated leukocytes; platelet-leukocyte aggregates; and more albumin extravasation have been observed in postischemic venules of low-density lipoprotein receptor deficient ($LDLr^{-/-}$) mice and rats placed on a high cholesterol diet, compared with their normocholesterolemic controls. [26] One of the factors contributing increased leukocyte recruitment in hypercholesterolemic animals is an altered expression of adhesion molecules; in that regard, an enhanced expression of P-selectin has been demonstrated in postischemic intestinal vasculature of hypercholesterolemic rats, as compared with normal animals. [27]

Diabetes

Diabetes mellitus is another risk factor for ischemic tissue disease that appears to influence the responses of the microvasculature to ischemia. Experimentally induced diabetes is also associated with a significant enhancement in the recruitment of rolling, adherent, and emigrating leukocytes; more albumin leakage; and an accelerated formation of ROS in mesenteric venules exposed to ischemia, when compared with venules of control rats submitted to the same ischemic insult. [21], [28], [29] The enhanced recruitment of rolling leukocytes is mediated by P-selectin, whereas the exaggerated firm adhesion of leukocytes is mediated by interactions between CD11/CD18 on leukocytes and ICAM-1 on endothelial cells. Although constitutive expression of ICAM-1 is similar in diabetic and nondiabetic animals, a more profound increase in ICAM-1 after ischemia has been observed in diabetic animals than in controls. [30]

Diabetes, like hypercholesterolemia, predisposes postischemic tissues to an oxidant stress. Measurements of ROS production within and surrounding mesenteric venules of normal and diabetic animals exposed to ischemia have revealed a more intense oxidant stress in postischemic venules of diabetic animals, which is abolished by interventions (e.g., oxypurinol, anti–ICAM-1 antibody, PAF receptor antagonist) that block the exaggerated recruitment of leukocytes observed in the diabetic condition. [21] Furthermore, PAF- or leukotriene B_4 (LTB_4)-stimulated neutrophils isolated from diabetic rats produce significantly higher amounts of superoxide than their counterparts in control (nondiabetic) animals. [21] Overall, these findings suggest that leukocytes, which may be primed for activation by lipid mediators such PAF and LTB_4, are the primary source of the large fluxes of oxidants generated in postischemic venules of diabetic animals. Unlike the responses observed in hypercholesterolemic animals, wherein the exaggerated albumin extravasation from postischemic venules is tightly linked to the accompanying enhancement of leukocyte–endothelial cell adhesion, a leukocyte-independent mechanism appears to mediate the ischemia-induced endothelial barrier dysfunction in diabetes. [28]

Hypertension

Relatively little is known about the influence of hypertension on the responses of the microvasculature to ischemia. Although a number of studies suggest that hypertension is associated with altered inflammatory responses, it remains unclear whether chronic arterial hypertension enhances or attenuates the responses elicited by inflammatory stimuli, such as ischemia. [20] For example, some reports describe a greater proportion of basally activated granulocytes in the blood of spontaneously hypertensive rats (SHR) compared with normotensive (WKY) control rats, and an increased spontaneous production of xanthine oxidase–derived oxidants in the microvasculature of SHR and hypertensive Dahl salt-sensitive rats. [31] However, these observations contrast with reports describing an impaired leukocyte–endothelial cell adhesion in venules of SHR (relative to WKY) exposed to inflammatory stimuli, [32] or lack of P-selectin up-regulation in response to ischemia and reperfusion in hypertensive animals. [27] When the inflammatory and microvascular responses to ischemia were compared in mesenteric venules of SHR and WKY rats, it was noted that the number of firmly adherent and emigrated leukocytes and platelet-leukocyte aggregations in postischemic venules of SHR were quite similar to those measured in WKY. However, albumin leakage from venules was more profoundly enhanced by reperfusion in SHR than in WKY. [33]

DEFINITIONS OF CLINICAL SYNDROMES

Under the term *intestinal ischemia,* a number of clinical disorders are included in which blood supply to the intestine and mesentery is impaired, thus limiting tissue oxygenation and nutrient availability. This can be a consequence of reduced splanchnic or mesenteric blood flow in the overall territory or in a regional bed.

The spectrum of intestinal ischemia comprises number of syndromes, including *acute mesenteric ischemia* (AMI) as a result of emboli, arterial or venous thrombi, vasoconstriction secondary to low-flow states, or small bowel loop strangulation; *chronic mesenteric ischemia* (also named *intestinal angina*) due to transient, recurrent episodes of inadequate intestinal blood flow to sustain metabolic needs or to support increased metabolic demand such as that associated with digestion; and *ischemic colitis,* in which a circulatory insufficiency of the colon results in varying degrees of local tissue necrosis and systemic manifestations.

ACUTE MESENTERIC ISCHEMIA

Acute insufficiency of the blood supply to the intestine can result from emboli, arterial and venous thrombi, or vasoconstriction secondary to low-flow states.

Arterial Embolism

Embolization to the SMA constitutes about 5% of peripheral emboli and accounts for roughly 50% of all cases of AMI. The SMA is anatomically susceptible to embolism because it has a large caliber and arises from the aorta at a narrow angle. [34] In contrast, the IMA rarely suffers embolic occlusion because it has a small caliber, and the celiac axis rarely suffers embolic occlusion because it arises from the aorta at a right angle. Risk factors for mesenteric arterial embolism are listed in Table 131-1. Most mesenteric arterial emboli originate from the left side of the heart, commonly from a thrombus in the left atrium or left ventricle, or from a lesion on the mitral or aortic valves. [35] Less commonly, emboli originate from an ulcerated aortic atherosclerotic plaque or a thrombosed aortic aneurysm. The most common precipitant of thrombus dislodgment and embolization is a cardiac arrhythmia, especially atrial fibrillation. Anticoagulation markedly decreases the risk for embolization from atrial fibrillation. [36] Myocardial dyskinesia, cardioversion, and cardiac catheterization are other clinical settings for embolization. The sudden occurrence of abdominal pain in these settings should prompt evaluation for SMA embolization. A substantial fraction of patients have prior emboli, and about 20% have synchronous emboli. [35]

Embolus (55% of cases)
Cardiac arrhythmia (atrial fibrillation)
Myocardial dyskinesia
Prosthetic valve
Cardioversion
Cardiac catheterization
Recent myocardial infarction
Prior or simultaneous emboli
Thrombus (15% of cases)
Prior arterial insufficiency
Coronary
Cerebrovascular
Peripheral
Old age
Low-flow state
Diabetes
Hypercholesterolemia
Hypertension
Congestive heart failure
Hypercoagulable states
Vasculitides
Aortic or mesenteric artery aneurysm
Trauma
Nonocclusive mesenteric ischemia (30% of cases)
Cardiogenic shock
Hypovolemic shock
Congestive heart failure
Pulmonary edema
Aortic insufficiency
Major cardiac or abdominal surgery
Dialysis
Vasoconstrictive drugs

TABLE 131-1 Factors Predisposing to Intestinal Arterial Ischemia

About 15% of emboli impact at the origin of the SMA, whereas most lodge distally 3 to 10 cm in the tapered segment of the SMA, past the origin of the middle colic artery. [37] Emboli tend to obstruct mesenteric flow acutely and completely and provide insufficient time to develop protective mesenteric collaterals. Intestinal ischemia due to embolic arterial occlusion can be compounded by reactive mesenteric vasoconstriction, further reducing collateral flow and worsening the ischemic insult. [2]

Arterial Thrombosis

Thrombosis accounts for about 15% of cases of acute intestinal ischemia. Thrombosis of the SMA or celiac artery is generally associated with a preexisting stenosis, usually at the origin of the arteries. Typically, the SMA plaque slowly progresses to a critical stenosis over years, and the residual lumen suddenly thromboses during a period of low flow. Patients usually have diffuse atherosclerotic disease, with prior coronary, cerebrovascular, or peripheral arterial insufficiency (see Table 131-1). About 30% of patients have histories consistent with chronic mesenteric ischemia, including postprandial pain, malabsorption, and weight loss before the acute episode. [34]

Nonocclusive Ischemia

Nonocclusive mesenteric ischemia (NOMI) causes 20% to 30% of episodes of AMI. [38] Mesenteric ischemia without anatomic arterial or venous obstruction is due to mesenteric vasospasm that can occur during periods of relatively low mesenteric flow, especially if there is underlying atherosclerotic disease. Such low-flow states can result from cardiogenic or hypovolemic shock (see Table 131-1). The vasoconstriction is produced by sympathetic activity, possibly mediated by vasopressin and angiotensin. [39] Moreover, mesenteric vasospasm may persist and perpetuate the ischemic injury even after correction of the precipitating event. [2] Vasoconstrictive

drugs, particularly a-adrenergic agents, vasopressin, ergotamine, diuretics, and digitalis glycosides, can contribute to NOMI. Digitalis, even at nontoxic levels, can cause paradoxical and sustained contraction of both arterial and venous smooth muscle cells in the mesenteric circulation. [40]

To make the diagnosis of NOMI, physicians must have a high index of suspicion because these patients may not present with the classic symptom complex of severe abdominal pain. Predisposing factors include myocardial infarction, congestive heart failure, pulmonary edema, aortic insufficiency, dialysis, [41] and major cardiac or intra-abdominal surgery. [42] The mortality rate of this specific subset of patients is relatively high irrespective of treatment because of the underlying medical conditions and the frequent delays in diagnosis.

Venous Thrombosis

Thrombosis of the SMV causes 5% to 10% of cases of AMI. [43], [44] Symptomatic SMV thrombosis is about 20-fold more common than symptomatic IMV thrombosis because of the larger caliber and flow of the SMV. The mean reported patient age ranges from 47 to 60 years, [6] which is younger than that for superior mesenteric arteriopathy because of the association of arterial disease with atherosclerosis.

Although previously less than 50% of patients had a recognized cause, etiologic factors are currently identified in about 80% of cases. [44] Risk factors for mesenteric venous thrombosis are listed in Table 131-2; they include hypercoagulable syndromes (protein C deficiency, protein S deficiency, factor V Leiden mutation, antithrombin III deficiency, antiphospholipid syndrome), portal hypertension, abdominal infections, perforated viscus, pancreatitis, and trauma. [45] The life-threatening complications of this syndrome are induced by the resulting bowel wall edema and increased outflow resistance secondary to venous thrombosis and increased blood viscosity, which can impede arterial inflow, leading to submucosal hemorrhage, venous capillary congestion, and bowel infarction.

Hypercoagulable states
Factor V Leiden mutation
Antithrombin III deficiency
Hyperfibrinogenemia
Antiphospholipid syndrome
Protein S deficiency
Protein C deficiency
Hormones or pregnancy
Neoplasms
Hyperviscosity syndromes
Myeloproliferative disorders
Sickle cell disease
Infection or inflammation
Pancreatitis
Appendicitis
Diverticulitis
Intra-abdominal abscess
Portal hypertension
Trauma

TABLE 131-2 Causes of Mesenteric Venous Thrombosis

Clinical Presentation

Abdominal pain out of proportion to physical findings is a cardinal symptom in patients with AMI and is commonly manifest after embolic or thrombotic occlusion of the SMA. [46] The pain may initially be colicky but becomes constant with progression of ischemia. The pain may be localized or diffuse. The duration of pain is typically short. A history of chronic postprandial abdominal pain is present when acute arterial thrombosis is superimposed on chronic mesenteric ischemia. [37] Abdominal pain may not be appreciated in confused or critically ill patients, and it is absent in 20% of patients with NOMI. [45] The symptoms of acute mesenteric venous thrombosis are usually less severe than those of acute arterial ischemia. Typically, these patients have a diffuse, intermittent abdominal pain of several days' or even weeks' duration.

In the absence of pain, unexplained abdominal distention and gastrointestinal bleeding may be the earliest signs of ischemia and impending intestinal infarction. Fever, diarrhea, nausea and vomiting, and diminished bowel sounds are other common but nonspecific manifestations of AMI. Diffuse or localized abdominal tenderness, rebound, and rigidity are ominous signs and usually herald transmural bowel infarction. [47] Infarction also leads to hypotension from fluid loss and septicemia, decreased urine output from hypovolemia and renal hypoperfusion, and hyperventilation from hypoxia and acidosis. [45]

Diagnosis

Laboratory Tests Serum laboratory tests are not very helpful in making the diagnosis of AMI; no serum marker is sensitive or specific enough to establish or exclude the diagnosis. Moreover, elevations in the levels of serum markers most suggestive of intestinal ischemia usually occur only after transmural bowel infarction develops and therefore cannot be used to diagnose AMI at its early stages, when improved survival would be possible. Leukocytosis, neutrophilia, and a shift to immature leukocytes in the leukocyte differential are common, [37], [48] but these laboratory findings are nonspecific markers of inflammation and infection. About 50% of patients have metabolic acidosis, an ominous finding suggestive of intestinal infarction, and 25% have hyperamylasemia. [49] Hemoconcentration may develop from fluid loss intramurally as interstitial edema, intraperitoneally as ascites, and intralumenally from fluid transudation. [37] Intestinal necrosis is associated with elevations of serum lactate, phosphate, and alkaline phosphatase levels as well as prerenal azotemia, hypoxemia, and bacteremia. [34], [37]
Radiography Plain abdominal radiography is most useful to exclude other potential causes of abdominal pain rather than confirming the diagnosis of AMI. Completely normal plain radiographs have been reported in more than 25% of patients with mesenteric ischemia. [50] Subtle signs of AMI on plain abdominal radiographs include adynamic ileus and distended, air-filled loops of bowel, but these abnormalities are most commonly the result of other causes, such as pancreatitis, mechanical obstruction, or colonic pseudoobstruction. More specific radiographic findings occur in 25% of cases, usually with advanced disease. These findings include mural thumbprinting resulting from edema or hemorrhage (Fig. 131-4). In advanced stages of ischemia, pneumatosis of the bowel wall can be detected. Specifically, portal vein gas on abdominal radiography portends an extremely poor prognosis. [51]

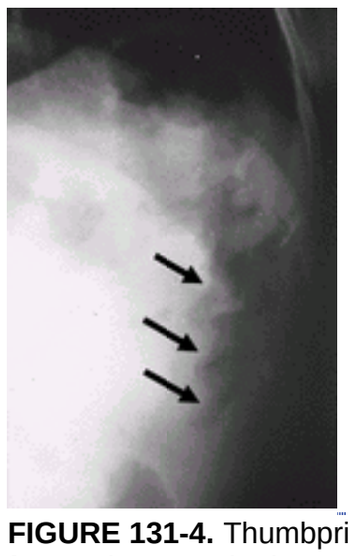

FIGURE 131-4. Thumbprinting. Detail of a plain abdominal radiograph showing semiopaque projections of the wall of the left colon into the lumen (*arrows*) resulting from submucosal edema and sometimes hemorrhage. Although thumbprinting is highly suggestive of intestinal ischemia, it occurs in relatively advanced lesions.

Intralumenal barium contrast evaluations are contraindicated because residual contrast can limit visualization of the mesenteric vasculature during diagnostic angiography. On the rare occasion when barium studies are performed in a patient with abdominal pain not initially thought to have AMI, thumbprinting, stricture, or ulcerations can be found.
Duplex (Doppler) Ultrasonography Duplex ultrasonography may be of some benefit in visualizing flow in the SMA and celiac axis. With expert technical assistance,

these tests can document proximal stenoses in the SMA or celiac axis or complete occlusion of these vessels with a high specificity (92% to 100%) but relatively low sensitivity (70% to 89%). [52] Unfortunately, duplex ultrasonography is of no value in detecting emboli beyond the proximal main vessel, nor in diagnosing NOMI. Moreover, identification of significant arterial stenosis does not establish the diagnosis of intestinal ischemia because total occlusion of two or even all three splanchnic vessels can be present in asymptomatic patients.

Computed Tomography Abdominal computed tomography (CT) is only sometimes useful in diagnosing intestinal ischemia caused by arterial occlusion or NOMI but is useful in excluding mesenteric venous thrombosis and other abdominal disorders in the differential diagnosis. [51] As on plain radiography of the abdomen, most abnormalities on CT associated with AMI are nonspecific and occur late in the course of the disease. Highly suggestive findings for AMI, including portal venous gas and pneumatosis intestinalis, are seen only after infarction has developed. A comparison of plain x-ray films of the abdomen and abdominal CT in patients with proven intestinal infarction showed specific findings in 30% and 39%, respectively. [50] Both abdominal plain x-ray films and CT scans showed only nonspecific abnormalities in 35% of patients with infarcted bowel, indicating that such studies cannot be used to exclude the diagnosis of AMI even after infarction has occurred. [50] In contrast to the limited role of CT in the diagnosis of AMI caused by SMA occlusion or NOMI, CT diagnosis of mesenteric venous thrombosis has proved more valuable (Fig. 131-5). All studies report sensitivities ranging from 90% to 100%. [2], [6] In a series of 20 patients who were stable and did not have peritonitis at the time of the diagnostic workup, the most common positive finding was the demonstration of thrombus in the SMV. The CT scan can demonstrate thrombus in the portal or splenic vein as well. Abnormal bowel characteristics on CT (bowel wall thickening, pneumatosis) can strongly suggest the diagnosis of acute mesenteric venous thrombosis. CT has shown thrombosis of the SMV without associated bowel abnormalities in asymptomatic patients. Such studies have broadened our understanding of the whole spectrum of mesenteric venous thrombosis, which ranges from chronic mesenteric venous thrombosis with no symptoms to acute mesenteric venous thrombosis with bowel infarction.

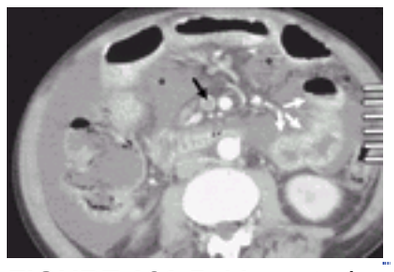

FIGURE 131-5. Mesenteric venous thrombosis in a patient with acute pancreatitis. Contrast-enhanced computed tomography scan shows small bowel wall thickening (*thick arrows*) and a hypoattenuating thrombus of the superior mesenteric vein (*thin arrow*). Ascites is also present (*asterisks*).

Although CT can be considered the primary diagnostic tool for patients suspected of having AMI from mesenteric venous thrombosis, its general use for those suspected of having any form of AMI is not supported in the literature. It has been suggested that contrast-enhanced CT scan should be the initial imaging study in patients suspected of having AMI who have risk factors for mesenteric venous thrombosis, such as a history of deep vein thrombosis or thrombophlebitis or a family history of a hypercoagulable state. [2] Spiral CT and CT angiography may be even more useful for evaluation of the splanchnic vessels, although experience with these techniques is limited. [53], [54]

Magnetic Resonance Angiography Magnetic resonance angiography (MRA) with and without gadolinium enhancement has been evaluated experimentally and clinically for the diagnosis of intestinal ischemia. High sensitivity and specificity were found for MRA in the detection of severe stenoses or occlusion of the origins of the celiac axis and SMA. [55] In patients with thrombosis of the portal or mesenteric veins the overall sensitivity, specificity, and accuracy of MRA for the detection of thrombosis have been reported to be 100%, 98%, and 99%; similar to those of digital subtraction angiography; suggesting that noninvasive contrast-enhanced MRA has the potential to replace intra-arterial digital subtraction angiography as the standard method to assess the whole intestinal venous system. [56] However, this modality is limited in its ability to identify more peripheral occlusions and NOMI.

Angiography Angiography should be performed early when acute mesenteric arterial occlusion is suspected. Because the diagnosis of AMI before bowel ischemia becomes irreversible is the most important factor in improving patient survival, and only angiography or surgery enables such early diagnosis, angiography has become a cornerstone in the evaluation of patients with abdominal pain who are at high risk for AMI. Although early angiography produces a considerable number of negative results, it is essential if diagnoses are to be made early enough to improve survival. The high sensitivity (64% to 100%) and specificity (100%) in most series [57], [58] seems to justify the reliance placed on this test. However, prompt laparotomy should be performed in patients with suspected AMI in whom expeditious angiography is not available. More controversial is the need for angiography in a patient with suspected AMI and signs of peritonitis. Because the latter indicate presence of infarcted bowel, the most compelling reason for angiography (i.e., diagnosis while the effects of intestinal ischemia are still reversible) is no longer a consideration. However, angiography may still provide valuable information in these patients because it may establish a definitive diagnosis of AMI and its cause; identify all vascular lesions, which will guide revascularization procedures; and offer the possibility to administer intra-arterial vasodilators in cases of NOMI. In cases of SMA embolization, most emboli are impacted 3 to 10 cm from the origin of the SMA, distal to the origin of the middle colic artery. The classic meniscus sign can often be visualized at the point of occlusion, which is different from the planar defect produced by a thrombus. [59] However, when angiography is performed more than 24 hours after embolization, the embolic filling defect becomes less sharp and less round because of proximal and distal clot propagation. Contrast is typically poorly visualized distal to emboli because the rapidity of embolic occlusion provides insufficient time to develop collateral flow. Other angiographic findings favoring embolus over thrombus include minimal atherosclerosis in mesenteric vessels, multiple lesions in mesenteric arterial branches, and simultaneous extramesenteric emboli. [59] Symptomatic thrombosis of the SMA is generally associated with a high-grade stenosis or occlusion of the celiac axis. The stenosis usually slowly progresses and permits reconstitution of vascular flow because of development of collaterals (see Fig. 131-2). Aortography is essential in these patients to evaluate potential inflow and outflow sites for bypass grafts as well as to clarify the extent and location of other atherosclerotic lesions in the iliac and IMAs. When the SMA distal to thrombotic occlusion fills from large collaterals, the SMA thrombus is probably chronic and not the cause of acute symptoms. In patients with NOMI, angiography usually reveals multiple areas of narrowing and irregularity in major branches. The small and medium arterial branches may be decreased or absent, producing a “pruned” arterial tree, and there is also impaired intramural vascular filling. [58] Acute mesenteric embolic or thrombotic obstruction may also produce mesenteric vasospasm, so that NOMI is diagnosed only when acute vascular obstruction is absent. The diagnosis of NOMI is strongly favored over the diagnosis of arterial thrombosis by an absence of extramesenteric atherosclerosis and by an increase in SMA caliber after transcatheter vasodilator therapy. [59] In mesenteric venous thrombosis, selective angiograms may reveal reflux of contrast material back into the aorta owing to extremely slow flow and heightened outflow resistance. A prolonged arterial phase with accumulation of contrast and thickened bowel walls is also characteristic. Extreme cases may demonstrate contrast extravasation into the bowel lumen, which may be indicative of active bleeding. The definitive diagnosis of mesenteric venous thrombosis is made during the venous phase; either a filling defect is noted within the portal vein, or, in more extensive thromboses, the entire venous phase is absent.

Treatment

General Measures Initial treatment of patients suspected of having AMI is aimed at resuscitation. Fluids are aggressively administered to replace intravascular fluid lost into the interstitium as edema, into the peritoneum as ascites, or into the lumen from fluid exudation or hemorrhage. Adequate hydration and optimization of cardiac function are particularly important in nonocclusive arteriopathy because hypovolemia and hypotension exacerbate mesenteric vasoconstriction. Medications that cause vasoconstriction, such as vasopressin or digitalis, should be discontinued. Electrolyte abnormalities and acid-base disturbances should be corrected before surgery. Broad-spectrum antibiotics are administered intravenously because intestinal ischemia promotes bacterial translocation and sepsis. Gastrointestinal decompression through a nasogastric tube is recommended because increased intralumenal pressure may decrease mucosal perfusion. Urine volume should be monitored with a bladder catheter. In critically ill patients, pulmonary pressures and cardiac output should be monitored with a Swan-Ganz catheter to optimize fluid and drug therapy. Measurements of arterial oxygen concentration will guide the need for oxygen supplementation or mechanical ventilation.

Embolism Various therapeutic approaches have been proposed for SMA emboli, including surgical revascularization, intra-arterial perfusion of thrombolytic agents or vasodilators, and systemic anticoagulation. The therapeutic option should be chosen taking into account the presence or absence of peritoneal signs, whether arterial occlusion is partial or complete, and whether location of the embolus is above the origin of the ileocolic artery or in more distal branches. There is uniform agreement that exploratory laparotomy is mandatory when signs of peritonitis are present and that embolectomy and resection of any infarcted bowel should be performed as necessary. When abdominal contamination is absent, primary anastomosis is generally performed. When extensive bowel segments are questionably viable, only unambiguously necrotic bowel is resected, and a second-look operation is performed about 24 hours later to permit demarcation between viable and nonviable bowel and to minimize the extent of bowel resection. [60] Anticoagulation with heparin is administered postoperatively to prevent recurrent embolization. [2] Several case reports and small series have reported successful nonoperative treatment of acute mesenteric arteriopathy using heparin [61] and thrombolytic agents, including urokinase, streptokinase, and recombinant tissue plasminogen activator. [62], [63] Data from that experience suggest that thrombolytic therapy is most likely to be successful when the embolus is partially occluding or is in one of the branches of the SMA or in the main SMA distal to the origin of the ileocolic artery and when the treatment is applied within 12 hours of the onset of symptoms. An important question is the long-term reocclusion rate after these therapies. Patients treated medically require close surgical observation to detect failure of medical therapy and the rapid progression to infarction. If signs of intestinal infarction develop, immediate surgery is

indicated. [2] There is some controversy about the use of vasodilators as a complementary treatment in patients with SMA embolus. There is evidence showing that vasoconstriction of both the unobstructed and obstructed branches occurs with an SMA embolus. [58] Such vasoconstriction resolves spontaneously if the embolus is removed soon after it develops. However, if the vasoconstriction has existed long enough, it can become persistent. In these cases, intra-arterial infusion of a vasodilator such as papaverine might be beneficial. [2]

Arterial Thrombosis If a diagnosis of acute thrombosis of the SMA is made, emergency surgical revascularization is recommended. Simple surgical thrombectomy is unlikely to be successful in the long term, and it has to be accompanied by revascularization performed through bypass. In a contaminated operative field, an autogenous aortovisceral graft is required, but polyester fluoroethylene prosthesis may be used when contamination is absent. [1] The proximal SMA should be opened through a longitudinal arteriotomy. If thrombectomy is successful, then a temporary intra-arterial SMA shunt can be placed while definitive revascularization is considered. The longitudinal arteriotomy in the SMA can serve as the distal anastomotic site for an antegrade bypass originating in the supraceliac aorta or for a retrograde bypass from the infrarenal aorta or iliac vessels. As for treatment of an embolus, bowel viability is assessed after revascularization to minimize bowel resection. Although thrombolytic therapy and percutaneous angioplasty have been recommended by some investigators for chronic mesenteric ischemia and SMV thrombosis, there a few reports of the use of these modalities for acute SMA thrombosis. [64], [65]

Nonocclusive Ischemia Management of NOMI is essentially pharmacological and is readily achieved by selective infusion of vasodilators into the SMA. Splanchnic vasodilators include papaverine, tolazoline, nitroglycerin, glucagon, prostaglandin E, phenoxybenzamine, and isoproterenol. [38] The greatest clinical experience is with papaverine, which is administered as a continuous infusion into the SMA at 30 to 60 mg/h. This approach has resulted in a reduction in mortality rates from 70% to 90% observed through the 1980s to 50% to 55% during the 1990s. [58], [60] Subsequent management is dictated by the patient's clinical response to vasodilator therapy. If peritoneal signs resolve, arteriography is repeated after a 30-minute saline infusion to document relief of vasospasm. Papaverine infusion should be continued for at least 24 hours thereafter. It is advisable to repeat arteriography before discontinuation of therapy unless precluded by the risk for contrast-induced nephropathy. Certain precautions specific to the use of intra-arterial papaverine are noteworthy. The patient's hemodynamic status should be carefully monitored because significant hypotension can result if the indwelling catheter migrates into the aorta and papaverine is infused into the peripheral circulation. In this event, the papaverine infusion should be substituted with saline and the position of the catheter confirmed with plain film. Immediate laparotomy is indicated during papaverine therapy if peritoneal signs do not rapidly remit with onset of infusion, if peritoneal signs develop during infusion, or if the patient deteriorates clinically as evidenced by increasing leukocytosis, sepsis, gastrointestinal bleeding, or unstable vital signs. [38] At surgery, gangrenous bowel is resected, according to the surgical principles described for an embolus. Intraoperative peritoneal lavage with warm saline and maintenance of a warm operating room temperature moistens and warms the intestine and helps reduce mesenteric vasospasm. The SMA catheter is left in place to provide vasodilator therapy during and after surgery.

Venous Thrombosis In asymptomatic individuals in whom the diagnosis has been made on a CT scan obtained for reasons other than abdominal pain, either no therapy or a 3- to 6-month course of anticoagulation is reasonable [2]; there are no studies to aid in this therapeutic decision. In symptomatic patients in whom an acute thrombosis of the SMV is diagnosed either by CT scan or angiography, treatment is determined by the presence or absence of peritoneal signs. As in all patients with AMI, signs of peritonitis mandate laparotomy and resection of infarcted intestine. In patients with SMV thrombosis, intravenous anticoagulation with heparin has been shown to prevent thrombus propagation and recurrence and to improve survival. [6], [44], [66] In the absence of peritoneal signs, heparin should be immediately initiated, with an initial bolus of 5000 units followed by continuous infusion of at 1000 units/h. Heparin dosage is then adjusted according to the prolongation of the partial thromboplastin time. Heparin is contraindicated in the presence of acute gastrointestinal bleeding from mesenteric ischemia. Heparin is administered for 7 to 10 days; then prolonged anticoagulation with warfarin for 3 to 6 months is recommended. [2] Thrombolytic therapy is rarely used for SMV thrombosis. Thrombolytic agents have been administered successfully in an antegrade fashion through the SMA, [67] retrograde through the internal jugular vein, [68] and transhepatically through the portal vein. [69] Nevertheless, these forms of therapy should be considered experimental for SMV thrombosis.

Prognosis

Until the early 1980s, the mortality rate of acute mesenteric arterial ischemia ranged from 70% to 90%. [70], [71] Early diagnosis and aggressive therapy have resulted in improved survival. In a report of 21 patients with SMA embolus, intestinal viability was achieved in 100% of patients if the duration of symptoms was less than 12 hours, in 56% if it was between 12 and 24 hours, and in only 18% if symptoms were more than 24 hours in duration before diagnosis. [72] During the 1990s, a 50% to 60% survival rate has been reported in large clinical studies. [72], [73] More than 90% survive when acute mesenteric arteriopathy is diagnosed early by angiography before peritoneal signs occur. [37] Causes of death include extensive intestinal necrosis, recurrent SMA thrombus or embolus, emboli to other sites, cardiopulmonary failure, and intestinal hemorrhage.

The mortality rate for acute SMV thrombosis, 25% to 30%, is lower than that for acute mesenteric arterial ischemia. [6], [66] Mortality is correlated with grade of venous occlusion, presence of collaterals, diagnostic delays, and comorbidity. [74] In a report of 53 patients with SMV thrombosis, 55% had major postoperative complications, such as short bowel syndrome, wound infection, sepsis, and recurrent SMV thrombosis. [44] Recurrences usually involve infarction of bowel adjacent to previously resected bowel.

CHRONIC MESENTERIC ISCHEMIA

Chronic mesenteric ischemia (intestinal angina) is a clinical syndrome characterized by recurrent abdominal pain and weight loss as a result of repeated transient episodes of insufficient intestinal blood flow, usually related to the increased metabolic demand associated with digestion.

Intestinal angina is an uncommon process that only occurs when severe atherosclerotic narrowing of a major splanchnic vessel exists in association with occlusion of one or two of the remaining vessels. This is because the very efficient collateral circulatory network in the small bowel and colon can successfully overcome the single occlusion of a major vessel. In fact, intestinal angina is very uncommon despite the finding at autopsy, performed in unselected cases, of significant stenosis of any of the three major splanchnic vessels in 29% in the overall series, and in 67% in those patients older than 80 years of age at the time of death. [75]

This ischemic syndrome is seen in middle-aged and elderly people and predominantly in women (female-to-male ratio of 3:2). Direct or indirect evidence of atherosclerosis is common in these patients, with more than one third having hypertension, coronary artery disease, peripheral arterial disease, or cerebrovascular disease. Patients with diabetes and smokers are at increased risk.

Clinical manifestations are related to limitations of the ability of the celiac artery or SMA to supply increased blood flow in response to metabolic demands after a meal, mainly because of atherosclerotic vascular lesions. Alternatively, it has been suggested that a steal phenomenon from the intestinal to the gastric circulation occurs when food reaches the stomach.

Clinical Presentation

Recurrent abdominal pain and weight loss are the two characteristic symptoms. Pain is epigastric or periumbilical, eventually radiates to the back, and appears shortly after meals, lasting for 1 to 3 hours. The intensity of the pain is related to the quantity of the meal or to the fat content, is crampy in nature, and has to be distinguished from pancreatic, biliary, or ulcer-related pain. As a consequence of the pain, patients tend to diminish food intake (sitophobia, or fear of eating), and with time, weight loss becomes obvious. Other nonspecific symptoms, such as nausea, vomiting, diarrhea, and constipation, are present in one third of patients. Isolated cases have been documented of malabsorption associated with chronic mesenteric ischemia, [76] and resolved gastroparesis has been reported after revascularization of occluded mesenteric vessels. [77]

Physical findings are not specific. Weight loss with signs of malnutrition is the most common alteration, although some patients maintain their weight despite the high frequency and intensity of the pain. Abdominal tenderness without rebound may be present during an episode of severe pain. Indirect signs of atherosclerotic vascular disease may be found, mainly diminished peripheral pulses, carotid or femoral bruits, or stigmata of past stroke.

Diagnosis

The diagnosis is mainly based on a characteristic clinical picture, presence of an occlusive lesion in the splanchnic vessels documented by angiography, and the absence of other common causes of abdominal pain. In most cases, patients undergo an extensive workup for obscure chronic abdominal pain, including routine laboratory evaluation, plain abdominal radiographs, upper and lower gastrointestinal endoscopy, small bowel barium series, and ultrasonography or abdominal CT. Laboratory abnormalities related to malnutrition, such as anemia, hypoalbuminemia, and hypocholesterolemia, may be present. When common causes of abdominal pain have been ruled out and suspicion of intestinal ischemia is well established from epigastric pain after meals and substantial weight loss, a selective angiographic

study of all three major splanchnic arteries is warranted, especially in patients with risk factors. Exceptions to this approach are patients with high risk for angiographic adverse events, such as those with renal failure or severe coagulopathy disorders. In about 90% of patients with intestinal angina, at least two major splanchnic vessels are found to be occluded or stenosed, and in up to 50% blood flow in all three vessels is severely compromised. [78] In contrast, isolated occlusion of a single vessel is uncommon in documented intestinal angina.

As is the case for AMI, other tests available for the diagnosis of chronic mesenteric ischemia include duplex ultrasonography, [79] MRI angiography, [80] MRI oximetry, [80] and tonometry. [81] In significant vascular stenosis, duplex ultrasonography shows high flow velocity and poststenotic turbulence reflected by modifications on spectral waveform and mixture of colors in the duplex image (Fig. 131-6). Retrograde hepatic artery blood flow suggests stenosis of the celiac artery. In stenosis of the SMA greater than 50% to 70%, the sensitivity of the technique is above 90%, with marginal improvement with postprandial study. [79] Sensitivity for lesions in the celiac axis are slightly lower, 85% to 87%. When performed by skilled operators, duplex ultrasonography may be a useful screening tool. However, if surgery or interventional radiology is planned, then angiography will be needed to ascertain coexisting lesions of the aorta and renal and iliac arteries.

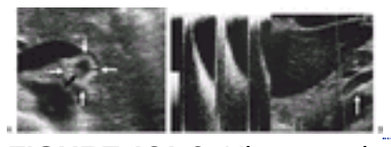

FIGURE 131-6. Ultrasound examination in a patient with celiac artery occlusion and superior mesenteric artery (SMA) stenosis. **A:** Gray-scale ultrasound showing a partial thrombus at the origin of SMA (delimited by *thick arrows*) occluding 50% of the arterial lumen (*thin arrow* shows thrombus). **B:** Pulsed Doppler study of the SMA origin of the same patient. Left half reveals a very high peak systolic velocity (3.84 m/s) resulting from the SMA stenosis (right half, *arrow*).

Tonometry seems a promising technique to evaluate the physiologic adequacy of intestinal blood flow, and the intestinal placement of the balloon has proved to be more sensitive than gastric tonometry, at least in animal studies. [81] However, its use in clinical settings has not been sufficiently evaluated yet.

Treatment and Prognosis

Surgical revascularization has been for many years the treatment of choice for chronic mesenteric ischemia. The techniques used include antegrade and retrograde bypass grafting, transaortic mesenteric endarterectomy, and aortic reimplantation of the SMA. Since the 1980s, percutaneous translumenal mesenteric angioplasty with or without stenting has been introduced.

Patients with symptomatic chronic mesenteric ischemia should be considered for surgery with the goals to relieve symptoms, improve nutrition, and prevent mesenteric infarction. Asymptomatic patients submitted to aortic reconstruction for aortoiliac occlusive disease who also have significant mesenteric occlusion should be considered for bypass grafting because these patients have an increased risk for mesenteric infarction after surgery. [82] The indication for surgery in asymptomatic patients who do not need aortic reconstruction is not clear. It has been proposed that surgery should be considered in such patients when three-vessel occlusion is documented, because of the high risk for developing ischemic symptoms or death. [83] Complete revascularization has been recommended by many surgeons, but several authors have recently proposed that bypass grafting of the SMA alone is an effective and durable procedure for treatment of intestinal ischemia. [84] Series published since the 1980s have reported a wide range of outcome figures for surgical revascularization, but results of more recent publications show mortality rates from 0% to 4%, success rates (defined as relief of symptoms) of more than 90%, and recurrence rates of less than 15%. [2] Advancing age, cardiac disease, hypertension, and additional occlusive disease influence overall mortality. Concomitant aortic replacement, renal disease, and complete revascularization are associated with high postoperative morbidity rates. [85] In patients with severe weight loss, parenteral nutrition before revascularization may reduce surgical morbidity.

The experience with percutaneous translumenal mesenteric angioplasty is not as extensive as with surgery and has been generally applied in high-risk patients. Success and mortality rates appear not to be significantly different from those of surgical revascularization, but recurrence of symptoms is slightly higher than after surgery. [86] Placement of a stent has been successfully used as an adjunctive therapy to angioplasty and, in some selected cases, as a primary method for treatment of chronic mesenteric ischemia. [87]

Based on the current information, it seems reasonable at the present time to offer surgical revascularization to low-risk patients and to perform angioplasty in those with a higher surgical risk. If it can be demonstrated that the addition of stenting to angioplasty can decrease the recurrence rate of symptoms better than angioplasty alone, then this procedure might be a valid alternative to surgery, even in low-risk patients. Surgery rather than angioplasty is indicated for suspected bowel necrosis.

In patients who survive surgical revascularization, the prognosis is excellent; 5-year survival rates approach 80%, and most patients become free from symptoms, gain weight, and resume normal eating habits. [78]

ISCHEMIC COLITIS

Ischemic colitis is the most common form of ischemic injury to the gut and occurs more frequently in elderly people. It can result from either occlusive or nonocclusive events, mainly in the territory of the IMA, in colonic branches of the SMA, and in the SMV and IMV. Thus, ischemic colitis is predominantly seen in the left colon. [88] The splenic flexure and rectosigmoid junction, where low perfusion exists (watershed areas), are commonly affected, whereas the rectum is not usually compromised because of excellent collateral perfusion. Right colon ischemic colitis is rare, [89] is mainly due to low-flow states, and is associated with poor prognosis. [90]

Large arterial vessel occlusion may be caused by thrombi or atheromatous lesions. [88] Embolization of the IMA is uncommon because most emboli are too large to enter this narrow vessel. Aortic surgery can cause ischemic colitis because of unnoticed ligation of the IMA or the occurrence of intraoperative hypoperfusion in the presence of IMA occlusion. [91] Blood flow in the small segmental arterioles may also be compromised by small vessel disease induced by diabetes, radiation, or autoimmune inflammatory arteritis. [92] Venous occlusion may occur as a consequence of hypercoagulability states (see Table 131-2), pancreatitis, or portal hypertension. [92] Vessel occlusion may be secondary to colonic obstruction from colon cancer, benign colonic strictures, adhesions, volvulus, or hernias. [93]

Nonocclusive ischemia may be present in the colon due to low-flow states or vasoconstriction induced by several drugs [94], [95] (Table 131-3). Ischemic colitis has been also reported in long-distance runners. [96]

Estrogen-progesterone	Diuretics	Alosetron
Cocaine	Pseudoephedrine	Barbiturates
Crack	Cyclosporine	Methylsergide
Ergotamine	α-Adrenergic agonists	
Vasopressin	β-Adrenergic antagonists	
Digitalis	Psychotropic drugs	

TABLE 131-3 Drugs Associated with Intestinal Ischemia

Clinical Manifestations

A wide spectrum of disorders can be seen as a consequence of colonic ischemia. These include acute, transient, self-limited ischemia (reversible colopathy characterized by mucosal and submucosal hemorrhage), acute fulminant ischemia (which is transmural and progresses to necrosis), and chronic ischemic colitis (partially reversible vascular disease usually manifested by late colonic stenosis).

Abdominal pain is present in more than two thirds of patients. [97] The pain is moderately severe, crampy, mostly localized in the left lower quadrant, and sometimes associated with diarrhea. Rectal bleeding manifested by either bright red blood in the stools or hematochezia is another common symptom. Bleeding is usually mild and does not require transfusion in most cases. Abdominal distention is also frequent and can be accompanied by nausea and vomiting. Physical signs during the acute episode include abdominal distention, with mild-to-moderate tenderness, fecal occult blood, tachycardia, or moderate fever. Less commonly, massive colon necrosis occurs with obvious signs of peritonitis and shock. Hypertension and history of cancer have been identified as independent prognostic factors of colonic necrosis in elderly people suffering an episode of ischemic colitis. [98] A minority of patients have subclinical acute ischemic disease and develop a colon stricture, which can be asymptomatic or manifested by constipation, diarrhea, or recurrent abdominal pain. [99]

Diagnosis

Although many patients with colon ischemia have no specific cause for such disease, all patients with any of the well-known precipitating factors who develop abdominal pain, bloody diarrhea, and abdominal distention should be investigated for ischemic colitis.

Plain abdominal radiographs usually show nonspecific, air-filled bowel dilation. The specific finding of thumbprinting (see Fig. 131-4) is seen in less than 25% of cases. [100] Colonoscopy or flexible sigmoidoscopy is the method of choice for the diagnosis of ischemic colitis because it allows direct visualization of the mucosa and tissue sampling. [97] Colonoscopy is preferable to sigmoidoscopy because 50% of the ischemic lesions are proximal to the sigmoid colon, except for ischemic injury after aortic surgery, in which lesions always involve the distal colon. The endoscopic examination has to be performed with caution and minimizing air insufflation in order to avoid perforation. The mucosa of the affected segment usually appears edematous, hemorrhagic, friable, and ulcerated. When bowel necrosis is present, colonoscopy reveals cyanotic, gray or black mucosa.

Biopsies often demonstrate nonspecific findings of vascular congestion, submucosal hemorrhage, interstitial edema, inflammatory infiltration, loss of superficial cells, and intravascular platelet thrombi (Fig. 131-7 and Color Fig. 131-7). Hyalinization of the lamina propria, full-thickness mucosal necrosis, presence of atrophic microcrypts, and hemorrhage in the lamina propria are also microscopic signs suggestive of acute ischemic injury, whereas hemosiderin deposition, along with transmural fibrosis and mucosal atrophy is a pathognomonic finding of the disease in its chronic phase. [101]

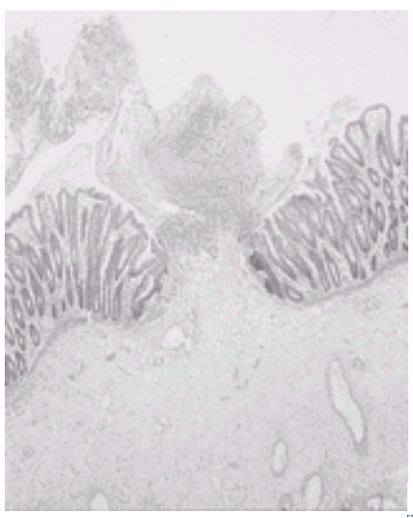

FIGURE 131-7. (See Color Fig. 131-7.) Intestinal ischemic damage. Well-delimited ulceration of the mucosa, with formation of a pseudomembrane. The remaining mucosa is normal. The submucosa has a marked edema and discrete infiltration by inflammatory cells.

Barium enema may be helpful for the diagnosis of ischemic colitis, with a sensitivity that approaches 80%. [102] Thumbprinting is the most characteristic finding, but ulcerations, mural deformity, sacculation, and transverse ridging may also be seen. CT is not usually performed for diagnosis of ischemic colitis, but it can show significant alterations of the bowel wall such as circumferential, symmetric wall thickening with fold enlargement, lumenal narrowing, and polypoid filling defects, and it may be helpful to exclude other diseases. [103]

Angiography is not usually indicated for the evaluation of ischemic colitis because by the time of clinical presentation, colon blood flow may be restored. However, in cases in which distinction between ischemic colitis and AMI is not clear, when only the right side of the colon is affected, and when a more generalized ischemic disorder is suspected, angiography must be performed. In some cases, color duplex ultrasonography is able to distinguish ischemia from inflammatory bowel disease, [104] and absence of arterial flow in the Doppler ultrasonography may be a better early predictor of outcome than clinical and laboratory findings. [105]

Differential Diagnosis

It is not uncommon that the diagnosis of ischemic colitis is established only after repeated clinical, radiographic, or endoscopic studies are performed after symptoms appear. This is because the signs and symptoms of ischemic colitis overlap with those of other colonic diseases or other mesenteric vascular diseases. Infectious colitis, inflammatory bowel disease, colon cancer, radiation colitis, diverticulitis, nonsteroidal antiinflammatory drug–induced colonic lesions, and pancreatitis should be considered.

Some pathogens, such as *Escherichia coli* O157:H7 and cytomegalovirus, may cause vascular damage in the colon, leading to hemorrhagic colitis that can be easily misdiagnosed as ischemic colitis [106], [107]; that is, ischemia may be an associated condition with these infections. [108] Pseudomembranous colitis produced by *Clostridium difficile* may also be mistaken for ischemic colitis. [109] Therefore, stool analysis for bacteria, ova, and parasites and the assay for *C difficile* toxin should be performed in some cases.

The differential diagnosis between ischemic colitis and inflammatory bowel disease can be difficult in some cases. [110] Data supporting the diagnosis of ischemic colitis are age of onset, segmental distribution of injury, abrupt transition between damaged and undamaged mucosa, rectal preservation (only useful for distinction with ulcerative colitis), and rapid resolution of lesions in subsequent colonoscopies. [111] In elderly people, the initial distinction between ischemic and ulcerative colitis may be more difficult. Because of its segmental involvement and rectal sparing, differentiation between ischemia and Crohn's colitis can also be difficult, although terminal ileum involvement and the presence of fistulae or colonic granulomata may help to diagnose Crohn's disease.

Radiation colitis can be easily suspected in patients with antecedents of such treatment, and colon cancer is differentiated mainly by pathological examination of colonic biopsy specimens. [112] Diverticulitis can mimic ischemic colitis, but CT scan is helpful to identify colon diverticula as well as mural thickening, pericolonic inflammation, and eventually fistula or abscess induced by diverticulitis. [113]

In the past decades, nonsteroidal antiinflammatory drug–induced colonic lesions have been identified more frequently than previously suspected. Abdominal pain and diarrhea with or without blood loss is the most common complaint of patients with such colonic lesions. Nonspecific inflammatory changes or erosions and diaphragma-like strictures may be seen at endoscopy, and histology may reveal findings similar to those of ischemic colitis. [114]

Treatment

Patients with reversible or transient ischemic colitis manifested by mild symptoms and signs do not need specific therapy because the event usually resolves spontaneously. In patients with more severe or persistent symptoms, bowel rest and general supportive measures, including parenteral fluids, are recommended. Precipitating factors should be corrected. This includes stopping any medication with potential vasoconstrictor effects on the mesenteric vasculature or narcotics that

may worse colonic distention. Measures should be implemented to correct low-flow states due to either hypovolemia or cardiac dysfunction. Intravenous infusion of broad-spectrum antibiotics is usually recommended, [115] although no definitive clinical evidence is available of the beneficial effects of such policy. Antibiotic use is based in the theoretical protection against bacterial translocation that occurs in severe colonic ischemia, and for early treatment of sepsis that follows massive colon gangrene. [116] In patients treated medically, endoscopy should be repeated during hospitalization to confirm diagnosis and to document recovery.

Patients showing evident signs of peritonitis require urgent surgery. In patients in whom severe symptoms, such as abdominal tenderness or fever, or laboratory findings, such as leukocytosis or metabolic acidosis, persist or deteriorate despite medical treatment, surgery is also warranted. [99] Other infrequent indications for surgery are massive bleeding, persistent ischemic lesions with protein-losing colopathy, and recurrent sepsis in a patient apparently recovered form an acute episode of ischemic colitis. In any case, segmental colectomy of the involved area should be performed, and the resected specimen should be opened in the operative room to assess the extent of mucosal injury. If surgical margins are involved, an additional segment should be resected until margins appear normal. Areas of questionable viability in the colon should also be resected. [88], [92] Recently, the use of intraoperative laser Doppler flowmetry has been proposed to ascertain the viability of colonic segments. [117]

In patients submitted to abdominal aortic graft surgery, several strategies are under investigation to improve rectocolonic microcirculation during and after surgery with the goal of preventing postoperative colonic ischemia. These include intravenous infusion of prostaglandin E_1 [118] and rectal administration of short-chain fatty acids. [119]

In cases of chronic evolution of ischemic colitis, surgery is required only when colonic stenosis becomes symptomatic. [120] Segmental resection is the procedure of choice in such cases, although transendoscopic dilation may be an alternative in selected patients at high risk for surgery.

UNUSUAL CAUSES OF MESENTERIC ISCHEMIA

Obstructing Diseases

An uncommon and controversial cause of mesenteric ischemia is the compression of the celiac artery and eventually the SMA by the median arcuate ligament, especially in thin, young patients, mostly women. It is manifested by upper abdominal pain induced by food intake and associated with weight loss. Diagnosis is made by angiography, duplex ultrasonography, contrast-enhanced CT, or MRA. Surgical division of the obstructing diaphragmatic fibers and denervation of the celiac ganglion, with or without celiac artery dilation or reconstruction, is the recommended treatment. [121]

When a segment of the large bowel is obstructed, the adjacent vasculature could be compromised, thus producing regional colonic ischemia. This phenomenon, which can manifest as an acute or chronic event, may be observed in some cases of sigmoid adenocarcinoma or small bowel carcinoid tumors, incarcerated hernia, sigmoid or cecal volvulus, intussusception, peritoneal adhesions, retroperitoneal fibrosis, neurofibromatosis, or cancer infiltrating the mesentery. [88], [122]

Vasculopathies

Several vasculitides may induce mesenteric ischemia. Polyarteritis nodosa (PAN) is characterized by necrosis of small or medium-sized muscular arteries with the eventual development of aneurysms. PAN may commonly involve the mesenteric circulation. In patients with PAN, gastrointestinal bleeding and bowel infarction with or without perforation are common events with very poor prognosis (mortality rate of about 75% to 100%). Recurrent abdominal pain and steatorrhea with weight loss are also manifestations of this syndrome. Erythrocyte sedimentation rate is usually significantly increased, and diagnosis is confirmed by characteristic angiographic findings. [123] Treatment with corticosteroids and cyclophosphamide provides the best results. [124] Bowel rest, nasogastric decompression, and parenteral nutrition are often needed. Early surgery is indicated when perforation or obstruction occurs.

Unusual cases of mesenteric ischemia have been described in systemic lupus erythematosus and in the antiphospholipid syndrome. [125] Although infrequent, intestinal ischemia may be seen also in some cases of Takayasu disease and Buerger disease, [126], [127] the latter usually presented in young men with heavy smoking. An increased risk for thrombotic events has been documented in Behçet disease and Crohn's disease, [128], [129] and primary or secondary amyloidosis may be associated with intestinal ischemia. [130]

Hematologic Disorders

Mesenteric ischemia can be associated with hypercoagulability states, mainly thrombocytosis, polycythemia, antithrombin III deficiency, antiphospholipid syndrome, proteins C or S deficiencies, 20210A prothrombin gene mutation, and factor V Leiden mutation. [131], [132], [133], [134] and [135] The diagnosis of hypercoagulability in the presence of mesenteric ischemia is crucial to prevent further episodes by means of anticoagulation.

Drugs

Several drugs may precipitate or promote mesenteric ischemia. Estrogen-progesterone combinations used as contraceptives have been described as precipitating factors for intestinal ischemia. [136] However, the frequency and the severity of the contraceptive-related ischemic events have decreased over time, probably because of more careful use of these drugs in patients at risk or because of a lower dose of estrogen included in the new generation of contraceptives. If mesenteric ischemia occurs in a young woman without any other risk factors, past or current use of oral contraceptives should be investigated.

Since the 1990s, an increased frequency of gastrointestinal events associated with cocaine has been reported, including gastric ulcerations, intestinal ischemia, perforations, and retroperitoneal fibrosis. [95] Similar complications have also been documented in crack users (the free-base form of cocaine). [137] The mechanism by which cocaine produces ischemia is uncertain, but it is known that it blocks the reuptake of norepinephrine and enhances the flux of calcium across the endothelial cell membrane, two pathways leading to vasoconstriction. [137], [138] The interval between drug ingestion and onset of symptoms varies from 1 hour to 2 days, and abdominal pain, tenderness, and bloody diarrhea are the most common presentations. Surgery is required in most cases to correct perforations or remove necrotic bowel segments.

Vasopressin used as a therapeutic agent for bleeding esophageal varices is associated with significant risk for systemic and regional ischemic events. [139] Indirect signs of transient intestinal ischemia are seen in some patients treated with vasopressin infusion as a consequence of its vasoconstrictor effect in the splanchnic vascular bed, but this drug also reduces gastrointestinal oxygen extraction. [140]

Recently, ischemic colitis has been described in association with the treatment of irritable bowel syndrome with alosetron, a selective 5-HT_3 antagonist. [141] Other drugs with possible effects promoting intestinal ischemia include ergotamine, digitalis, methysergide, pseudoephedrine, barbiturates, and amphetamines. [88]

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CHAPTER 132

Steven M. Cohn and Stephen J. Bicksto

RADIATION INJURY IN THE GASTROINTESTINAL TRACT

The first reports of radiation-induced injury to the gastrointestinal tract date to the early 20th century shortly after the discovery of x-rays in 1895. [1] The use of ionizing radiation for therapy of malignancies also dates back to the early 20th century and arose out of the observations of Bergonie and Tribondeau, who suggested that "X-rays are more effective on cells which have a greater reproductive activity; the effectiveness is greater on those cells which have a larger dividing future ahead…." [2], [3] They went on to reason that radiation could be effective in destroying tumors while sparing many normal tissues. Although this concept may be somewhat simplistic in today's terms, it led to the expectation that side effects would occur in those tissues, such as the gastrointestinal epithelium, that undergo rapid cell turnover.

The use of multimodality therapies that include radiation have become commonplace in treating many malignancies; in fact, about one half of patients with cancer receive radiation therapy as a component of their treatment. Modern techniques for tomographic localization and fractionation of radiation therapy have significantly reduced short-term and long-term gastrointestinal morbidity resulting from radiation therapy. Nevertheless, most patients experience gastrointestinal symptoms associated with acute radiation therapy, such as diarrhea, abdominal pain, bloating, tenesmus, and bleeding. [4] Chest pain, dysphagia, and odynophagia may be seen when the radiation fields involve the upper gastrointestinal tract. Generally, the acute side effects of therapy can be treated with antispasmodics, antidiarrheals, and topical anesthetics. As a rule, these symptoms resolve shortly after radiation treatment ends. However, up to one fourth of patients who receive radiation therapy also develop some form of chronic injury, defined as symptoms presenting more than 3 months after completion of therapy. [4] Symptoms are usually evident within the first 2 years after initiation of therapy; however, some patients do not develop symptoms for years or even decades. [4] Thus, it is likely that gastroenterologists will continue to encounter and treat patients with acute or chronic radiation injury in their practice.

RADIATION PATHOBIOLOGY

High-energy photons from gamma irradiation, which is used for most types of radiation therapy, can interact with atoms in a number of ways. [5] The most prominent effect leading to radiation-induced injury is Compton scattering, first described by A. H. Compton in 1922. Compton scattering occurs when a gamma photon collides with an outer orbital electron, loses energy, and is deflected, and the energy lost by the photon is imparted to the electron, which is then ejected from the atom. These ejected Compton electrons subsequently cause many of the biologic sequelae of irradiation by secondary ionizations of other atoms or through generation of other highly reactive species, such as free radicals. Such radiochemical interactions can damage a variety of cellular constituents, including DNA, proteins, and lipids. Of these, induction of single- and double-stranded breaks in DNA appears to mediate most of the lethal effects of radiation on cells. Although many cellular pathways for repairing such potentially lethal genetic damage have evolved in humans, failure to repair even a single double-stranded break in DNA may result in the death of that cell.

The amount of radiation-induced damage produced in a tissue or tumor is dependent on the energy of the gamma rays or x-rays, on the absorbed dose of radiation, and on the fractionation schedule of the administered dose of irradiation. [3], [5], [6] The gray (Gy) has supplanted the rad as the accepted unit of absorbed radiation dose and is defined as 1 joule of energy deposited per kilogram of absorbing material. One Gy is an equivalent dose to 100 rad. The dose of radiation absorbed by a biologic tissue is variable and related to the biochemical properties of the tissue, including tissue oxygenation, the activity of pathways that detoxify reactive oxygen species or scavenge free radicals, and the position of cells within the cell cycle. Cells in early S phase or those undergoing mitosis appear to be most sensitive. A minority of DNA damage occurs through direct interaction with high-energy Compton electrons. However, these electrons can interact with other molecules present in the cell, including water and molecular oxygen, to generate peroxides, free radicals, and other reactive oxygen intermediates that are highly reactive with DNA and other cellular acromolecules. Unfortunately, malignant cells that are relatively hypoxic within solid tumors may be resistant to the lethal effects of photon irradiation.

Physical characteristics of the patient, previous abdominal surgeries, and concurrent medical illnesses can also influence the amount of radiation-induced damage to the alimentary tract. Predisposing factors for radiation injury have been identified: higher doses of radiation, patients whose intestines are fixed within the exposed region by adhesions from previous surgery, and those with a narrow anteroposterior diameter are at greater risk. [7] Preexisting vascular disease within the radiation field may predispose patients to increased radiation-induced vascular injury and subsequent ischemia (see subsequent section on " Pathology"). Thus, patients with diabetes, hypertension, and atherosclerotic disease are all more prone to radiation enteritis. [8] Inflammation may also modify the acute response to irradiation through growth factors, cytokines, and other inflammatory mediators produced at sites of tissue injury. For example, fibroblast growth factor-2, prostaglandin-E_2, tumor necrosis factor-a, interleukin-1 (IL-1), and IL-11 all decrease the sensitivity of tissues to radiation injury, suggesting that these mediators may have an adaptive function in cell survival. [9], [10], [11], [12] and [13] IL-12 has a mixed effect, protecting bone marrow–derived cells but sensitizing gut epithelial cells to radiation injury. [13], [14] and [15] Thus, the effects of tissue inflammation on the biologic response to therapeutic irradiation are complex and difficult to predict.

Concurrent pharmacological therapy may also alter the response of both normal and neoplastic tissue to gamma irradiation. Tissue is more susceptible to injury from radiation when it is administered together with chemotherapeutic agents or other pharmaceuticals that sensitize the gut. For instance, combination of gamma irradiation with administration of chemotherapeutic agents that alter cell cycle kinetics or lead to synchronization of the replicating cell populations may potentiate damage to normal tissues or to tumor cells because the lethal effects of radiation-induced damage vary with position in the cell cycle (see earlier discussion). Therefore, the timing of irradiation in relation to administration of these chemotherapeutic agents is an important factor in determining the relative effects on tumor versus normal tissue. Halopyrimidines such as fluorouracil, fluorodeoxyuridine, and iododeoxyuridine may sensitize tumors both by inhibiting effective DNA repair and by increasing the amount of radiation-induced DNA damage. [16], [17] Adjuvant therapy combining fluorouracil and radiation therapy for resected gastrointestinal tumors has improved survival in patients with pancreatic, rectal, and esophageal cancers. [17], [18], [19] and [20] Actinomycin D, doxorubicin, and bleomycin also sensitize cells to radiation-induced DNA damage and cell death.

Drugs with radioprotective activity are under investigation. These compounds have the potential to improve the therapeutic index for radiation therapy. For example, the prostaglandin analogs misoprostol and dimethyl–prostaglandin E_2 both decrease radiation-induced apoptosis in rapidly replicating intestinal epithelial cells and enhance epithelial stem cell survival after irradiation. [9], [12] A number of growth factors and cytokines, including fibroblast growth factor-2, keratinocyte growth factor, and IL-11, also have radioprotective activity in the intestine if administered before irradiation. [11], [21], [22] Free radical scavengers, such as amifostine (WR-2721), have been shown to be radioprotective in both experimental systems and clinical trials. [23] Whether these radioprotective agents will find a role in routine clinical practice remains to be established.

Because DNA damage is thought to be the primary mechanism mediating the lethal effects of radiation, [5], [6] a number of complex intracellular systems have evolved to repair DNA damage induced by radiation as well as damage induced by chemotherapeutic agents. Some of these proteins are specifically involved in recognition and repair of DNA damage. Other enzymes, such as DNA-dependent protein kinase, are involved in DNA replication and V(D)J recombination during immunoglobulin gene rearrangement in normal lymphocytes. These enzymes also play a central role in rejoining double-stranded DNA breaks induced by radiation injury in epithelial

cells in the gastrointestinal mucosa. [6], [24], [25]

The fate of injured cells after irradiation is a complex process. Cells that are not able to repair radiation-induced DNA damage adequately will eventually undergo "mitotic cell death" or mutation due to the genomic instability as a consequence of the unrepaired DNA stranded breaks and other DNA damage. [6] However, in rapidly replicating cell populations, DNA damage induced by relatively small doses of irradiation can also trigger apoptotic cell death, an active, genetically programmed type of cell death. In both humans and rodents, a very low frequency of physiologic apoptosis occurs continuously within the lower regions of the crypt epithelium of the normal small intestine and colon and may be an inherent part of the regulatory mechanism determining stem cell numbers in the normal adult tissue. [26], [27] After even relatively low doses (about 1 Gy) of ionizing radiation, many damaged cells within the rapidly dividing crypt epithelium of the small intestine and colon undergo apoptosis. [26], [27] and [28] Radiation-induced apoptosis occurs with the highest rates in the lower two thirds of the crypt, a region containing the epithelial stem cells and the more rapidly replicating progenitor or "transit" cell population, and reaches a maximum 6 to 12 hours after irradiation. Many of the remaining cells within the proliferative zone that do not undergo apoptosis arrest at checkpoints in G_1 or in late G_2 phases of the cell cycle. [6], [28], [29] These cell cycle checkpoints are thought to provide time for repair of DNA damage and prevent cells from entering S phase or mitosis, when the presence of even a few double-stranded DNA breaks may result in death of the cell or generation of cells with chromosomal abnormalities.

The tumor suppressor gene, *p53,* plays an important role in recognition of DNA damage and coordinating the subsequent cellular response to radiation injury, leading some investigators to describe *p53* as the "guardian of the genome." [30], [31] and [32] It is normally expressed at low levels except during DNA synthesis. Upon DNA damage, expression of *p53* is up-regulated and translocated to the nucleus, where it binds DNA and regulates the transcription of a number of genes, including *p21 cip1, waf1*, *GADD45, mdm2,* and *Bax*. [33], [34] and [35] These genes are involved in regulating apoptosis or cell cycle arrest at checkpoints at the G_1/S boundary or in G_2/M. [36], [37] and [38] Activation of *p53* occurs rapidly in the intestinal crypt epithelium following even low doses of gamma irradiation (0.5 to 1 Gy). This results in a rapid and sustained increase in the transcription of *Bax,* a proapoptotic member of the *Bcl-2* oncogene family that mediates the apoptotic response. The ultimate fate of a cell with DNA damage also depends on the expression of other counterregulatory genes that affect apoptosis. Expression of these counterregulatory genes varies along different regions of the alimentary tract and may depend on extracellular signals as well. For example, antiapoptotic members of the Bcl-2 family, including Bcl-2 and *Bcl-X* $_L$, suppress apoptosis by formation of heterodimers with *Bax*. *Bcl-2* appears to be expressed in the colonic crypts, but not in small intestinal crypt epithelial cells, thus explaining regional differences in the sensitivity of crypt epithelial cells to radiation-induced apoptosis.

Activation of *p53* in response to DNA damage causes cells to arrest at critical checkpoints, [6], [30], [31], [37] a process that is mediated by induction of *p21 cip1, waf1*, *GADD45,* and other gene products. This allows time for the cell to make a decision as to its fate (repair versus cell death) before committing itself to S phase or mitosis. Entry into S phase with damaged DNA or into mitosis in the presence of single-stranded or double-stranded DNA breaks could lead to daughter cells with unstable chromosomal structure, allowing mutations or chromosomal rearrangements to occur. Other genes, such as the gene responsible for ataxia telangiectasia (*Atm*), are thought to regulate the response to DNA damage upstream of *p53*. Thus, cells from these patients have both diminished activation of *p53,* decreased cell cycle arrest at the G_1/S cell cycle checkpoint, an increased sensitivity to ionizing radiation, and increased malignant transformations.

The clonogenic stem cell populations within the intestine and colon appear to be more resistant to radiation-induced cell loss than the more rapidly replicating transit cells. Furthermore, the survival of these clonogenic stem cells may be determined by a mechanism independent of *p53* function. [39] The increased survival and proliferation of the stem cells with radiation-induced DNA damage probably enhances the capacity for restitution of the epithelial function in the short term but also may contribute to the potential for secondary malignancies many years after therapeutic irradiation (see subsequent section on " Pathology").

PATHOLOGY

The histopathological changes in the alimentary tract follow a predictable time course and may be functionally divided into early and delayed clinical syndromes. [40] The early effects primarily involve the mucosa, which is lined by rapidly proliferating epithelial cells that are sensitive to the acute effects of radiation injury. Clinical symptoms include odynophagia, diarrhea, nausea, vomiting, or gastrointestinal bleeding, depending on the location of the radiation field, the dose of radiation, and the fractionation schedule. The delayed effects are more likely to be chronic diarrhea, fibrosis, ulcer formation, or bleeding and are secondary to damage to the vasculature of the organs involved. Although the histological hallmarks seen in tissue specimens are often characteristic of acute or delayed radiation injury, no histological feature is pathognomonic for radiation-induced damage. [40]

The histological and pathological features of acute radiation injury in the human alimentary tract are poorly defined because the acute effects of radiation therapy are generally well tolerated and tissue samples are rarely obtained. Thus, much of what is known about the acute effects of irradiation has been derived from a few studies in humans and more extensive experiments using animal models of radiation injury. [40], [41], [42], [43], [44] and [45] The earliest recognizable histological features following radiation injury to the gastrointestinal tract occur within hours and include apoptosis of lamina propria lymphocytes and epithelial cells, the cessation of epithelial cell replication, and damage to vascular endothelial cells. This results in loss of replicating cells in the squamous epithelium of the esophagus and the columnar epithelial cells in the gastric, small intestinal, and colonic epithelium. Mature, differentiated epithelial cells continue to be lost by extrusion, and in the absence of replacement by replication of the progenitor cells, there is subsequent loss of mucosal function: attenuation and damage to the squamous epithelium in the esophagus; reduced numbers of parietal and chief cells in the gastric epithelium which will decrease gastric secretion; and progressive loss of the small intestinal villous epithelium, which decreases absorptive surface area, and in some patients, leads to transient malabsorption of fats and other macromolecules, or acute diarrhea. In the colon, there are reduced numbers of mitoses and epithelial cell atypia with loss of nuclear polarity in the columnar epithelium lining the crypts. Swelling and loss of goblet cells leads to decreased mucus production. Microabscesses composed of granulocytes and eosinophils may be present. Mucosal and submucosal edema may also be observed in both the small intestine and colon.

An increase in the replicating crypt epithelial cell population in the small intestine and colon, through proliferation of stem cells located near the base of the crypts, is observed beginning about 3 days after low doses of irradiation. This increase in crypt epithelial proliferation is able to compensate for the initial loss of differentiated epithelial cells. [28] However, the survival of the stem cell population is compromised at higher single doses of gamma irradiation, limiting epithelium repair. Fractionated irradiation protocols allow higher total doses to be administered by allowing repair and recovery of replicating epithelial cells and stem cells between fractions. [6] If damage to the stem cell population is severe after high doses of radiation, then epithelial integrity and barrier function are lost, leading to translocation of lumenal bacteria and eventual death from loss of fluids and serum proteins, hemorrhage, and sepsis 7 to 10 days after irradiation. Although rarely, if ever, seen in therapeutic applications of gamma irradiation, death from this syndrome has been observed after exposure of individuals to a single whole-body dose of 10 to 20 Gy as a consequence of radiation accidents or atomic warfare. [40], [46]

Liver biopsies are rarely obtained from patients with acute hepatic injury secondary to therapeutic irradiation for solid neoplasms. However, hepatic venoocclusive disease is not uncommon in bone marrow transplant recipients after cytoreductive therapy with combined chemotherapy and irradiation (see later), [47], [48] and [49] usually occurring before posttransplantation day 30. [48] Hepatic histology shows vascular congestion that is most prominent in centrilobular areas, subendothelial edema, endothelial destruction, sinusoidal dilation, and centrizonal hepatocyte necrosis with attenuation of the hepatocellular cords. [47] Deposition of clotting factors in the walls of hepatic venules suggests that microthrombus formation may be the primary pathogenic mechanism. [50] Occlusion of central veins by thrombus is sometimes observed. As the disease progresses, deposition of collagen in hepatic sinusoids, fibrosis of hepatic venules, and sclerosis of venular walls may be seen. [47], [48] One proposed mechanism for the increased sensitivity to combined chemotherapy and irradiation in this setting involves depletion of glutathione stores in sinusoidal endothelial cells and hepatocytes. [48], [51], [52] and [53]

The delayed effects of radiation injury most commonly occur from 6 months to 5 years after radiation therapy; however, symptoms may appear as early as 3 months or as late as 30 years after radiotherapy in a small number of patients. [40], [54], [55] Vascular injury and regeneration are characteristic of chronic radiation injury and are often observed in pathological specimens. [40] However, these findings are sometimes focal and may be missed unless multiple levels through the specimen are examined. Myointimal proliferation in medium-sized muscular arteries may lead to chronic ischemic injury because of the marked decrease in the lumenal diameter of these vessels (Fig. 132-1). Ulceration of the overlying mucosa occurs in areas of localized ischemia (Fig. 132-2; see also Color Fig. 132-2). Thrombosis is sometimes seen associated with these lesions and is more frequent in vessels with fibrinoid necrosis adjacent to deep ulcerations. The presence of lipophages or foamy macrophages in the intima of small arterioles is also a characteristic finding of delayed radiation injury, although this lesion may also result from other etiologies (Fig. 132-3). Telangiectatic vessels are frequently found in the submucosa and likely account for the diffuse bleeding sometimes observed in radiation enteritis (Fig. 132-4 and Color Fig. 132-4; see later). Sclerosis or medial fibrosis indicative of healing vasculitic lesions may also be observed.

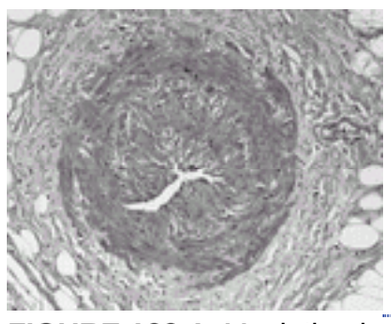

FIGURE 132-1. Myointimal proliferation nearly occludes the lumen of a moderate-sized mesenteric artery. This process usually occurs over several years after radiation injury and may lead to chronic ischemic injury to the intestine. (Original magnification × 100.) (Courtesy of Dr. Christopher Moskaluk.)

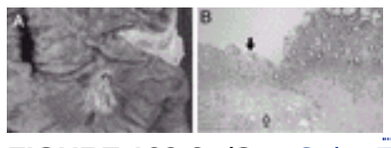

FIGURE 132-2. (See Color Fig. 132-2.) Radiation-induced ulceration in the colon. **A:** Gross appearance of a well-demarcated ulcer present in the rectum years after external radiation for an adjacent neoplasm. **B:** Histological appearance with chronic ulceration, mucosal necrosis (*solid arrow*), and dense submucosal fibrosis (*open arrow*) similar to that seen in ischemic injury. The lesion is notable for the absence of a prominent inflammatory infiltrate. (Original magnification × 20.) (Courtesy of Dr. Christopher Moskaluk.)

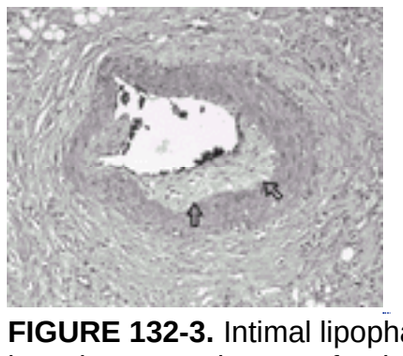

FIGURE 132-3. Intimal lipophage accumulation in intestinal arteriole after irradiation. These foam cells may be seen in the intima of small arteries and arterioles of the intestine several years after irradiation and may lead to lumenal narrowing of these vessels. (H&E, original magnification × 100.) (Courtesy of Dr. Christopher Moskaluk.)

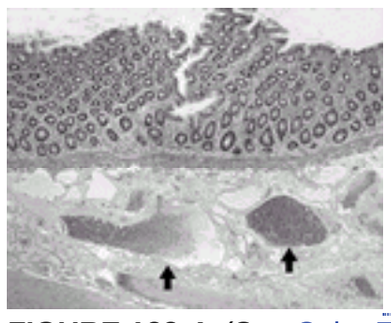

FIGURE 132-4. (See Color Fig. 132-4.) Submucosal telangiectasias (*arrow*) are seen in delayed radiation injury. Dilated venules and lymphatic channels are seen in the submucosa underlying relatively normal-appearing colonic epithelium. (H&E, original magnification × 40.) (Courtesy of Dr. Christopher Moskaluk.)

Changes in the submucosa are frequently observed in delayed radiation injury and are thought to be secondary to the chronic vascular damage described earlier. [40] Increased collagen deposition and fibrosis are often observed with minimal cellular inflammatory infiltrate. The fibrosis may be confined to the submucosa or extend through the muscularis propria and accounts for the thickening or strictures that may occur within any irradiated region of the alimentary tract. Atypical fibroblasts with bizarre nuclei are sometimes found in areas of fibrosis (Fig. 132-5, Color Fig. 132-5). The appearance of these fibrotic lesions in the small intestine may resemble that of Crohn's disease both on gross inspection and on radiologic examination, although fistulae and creeping fat are rarely observed.

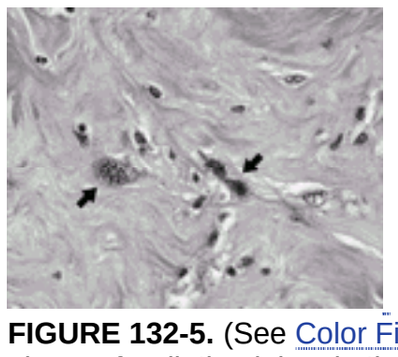

FIGURE 132-5. (See Color Fig. 132-5.) Atypical fibroblasts in radiation injury (*arrows*). Bizarre-appearing, pyknotic fibroblasts are frequently seen in the delayed phase of radiation injury in the alimentary tract. Although these atypical fibroblasts are frequently observed, their presence is not specific for radiation injury. (H&E, original magnification × 400.) (Courtesy of Dr. Christopher Moskaluk.)

Mucosal alterations in the small intestine include villous atrophy, flattening of epithelial cells, and fibrosis of the lamina propria. [40] These findings are often observed in patients with or without symptomatic radiation enteritis and may result in malabsorption when these changes are extensive. Superficial erosions and mucosal ulceration are common, sometimes penetrating deep into the submucosa and muscular propria and occasionally perforating into the peritoneal cavity. Chronic changes in the mucosa of the colon are similar to those seen in the small intestine.

Colitis cystica profunda is a rare delayed complication of radiation injury. [40] The importance of this lesion is in distinguishing it from invasive adenocarcinoma. Its symptoms are similar to colitis cystica profunda secondary to other etiologies. However, its histological appearance may differ from colitis cystic profunda of other

causes in that colonic glands extend deep into the submucosa and sometimes penetrate into the muscularis propria [40], [55] (Fig. 132-6). The presence of glands lined by normal epithelium usually makes it easy to distinguish colitis cystica profunda from adenocarcinoma in pathological specimens.

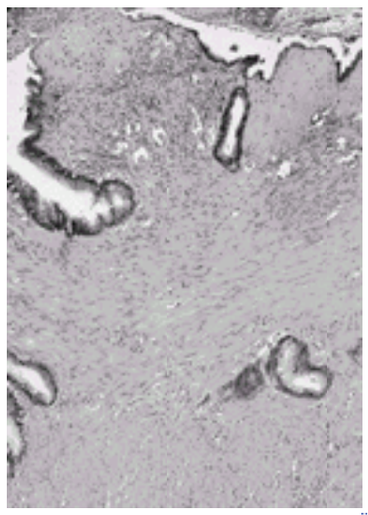

FIGURE 132-6. Colitis cystica profunda. Colonic glands are seen penetrating the muscularis mucosa into the submucosa and muscularis propria. This lesion is distinguishable from carcinoma by the normal cytologic characteristics of the epithelium lining the penetrating glands. (Original magnification × 40.) (Courtesy of Dr. Christopher Moskaluk.)

The development of secondary epithelial or stromal malignancies is also a potential complication of radiation therapy. [40], [56], [57] and [58] However, the number of documented cases is relatively small, and most of these cases generally have occurred after lower doses of gamma irradiation (less than 10 Gy). For example, the frequency of colon cancer is increased in atom bomb survivors 30 years after their exposure (relative risk of 2.4 compared with a matched control population), and an increased risk for colon cancer has been documented in a cohort of women irradiated with relatively low doses (6.25 to 10.5 Gy) for benign uterine bleeding. [40], [59], [60] and [61] Thus, whereas exposure of the colon to radiation may increase the risk for developing a secondary malignancy, the interval between therapeutic irradiation and development of secondary malignancies may be quite long. [40], [59], [60]

SPECIFIC ORGAN INVOLVEMENT

Esophagus

Radiation therapy for thoracic tumors and hematologic malignancies can result in both acute and long-term esophageal injury. The field required to treat thoracic, hematologic, and some pharyngeal malignancies frequently (25% in one series) [62] exposes the esophagus to sufficient radiation to produce pathological changes, but clinical syndromes generally require doses of 6000 cGy. [63] This threshold may be lower in patients whose tissue is sensitized by chemotherapeutic agents. Three major syndromes related to esophageal irradiation have been described: (1) acute inflammation with odynophagia or dysphagia; (2) fibrosis with stricture and dysphagia; and (3) fistula formation with aspiration into the trachea, or lethal bleeding from the aorta. During the acute phase of radiotherapy, symptoms are usually related to mucosal injury and the inflammatory response. Patients typically present with mucositis, which causes odynophagia or dysphagia beginning after the second week of treatment. Frank bleeding is unusual. [64] Acute dysphagia may also reflect a motor disturbance. A pattern of ineffective peristalsis at and distal to the radiation field may occur 4 to 8 weeks after initiation of radiation therapy.

The diagnosis of acute-phase radiation-induced esophagitis is usually evident from the patient's history and symptoms, although other conditions need to be excluded. Acute infection must be considered, particularly for those whose immune systems are impaired by the underlying disease or its treatment. Pill ulceration and reflux esophagitis are also in the differential diagnosis. Endoscopic evaluation is the test of choice, allowing characterization of the lesion and tissue sampling where appropriate. Barium swallow may offer some information in unusual circumstances when endoscopy is not clinically feasible, but these radiologic studies are normal or nondiagnostic most of the time. [65]

Prevention of acute radiation injury should be considered the first line of therapy. Limiting the total radiation exposure and careful selection of the radiation field are important preventive measures. Pharmacotherapy and dietary modifications have also been advocated. [66] If symptoms of odynophagia and dysphagia are sufficiently severe, temporary interruption of radiation therapy for several days to 1 week or enteral nutritional support may be necessary. Pharmacological therapy is often administered on an empiric basis and is directed at controlling symptoms; for example, topical lidocaine is generally given for odynophagia and may be compounded into a variety of "stomatitis cocktails" along with antacids, antihistamines (diphenhydramine), or antifungal (nystatin) ingredients. Antisecretory therapy is frequently used, but, like these cocktails, suffers from the same lack of substantiating data. [64], [67]

Patients with strictures or fistulae typically present well after radiation therapy has been completed (i.e., 6 months or later after initiation of treatment). Diagnosis and therapy of strictures and fistulae typically require a combination of radiologic and endoscopic techniques. Fluoroscopic evaluation is needed to determine the site, whereas either fluoroscopic or combined endoscopic and fluoroscopic methods can be used for subsequent dilation or stent placement. The initial approach to treatment of late esophageal stenosis is careful dilation. Balloon dilation devices may be preferable to rigid dilators because they allow isolated radial expansion of the stricture with minimal axial pressure. The complication rates for the dilation of strictures induced by adiation is not greater than dilation of strictures or other etiologies. [68] However, stent migration of conventional plastic endoprostheses was substantially higher in the radiation group (21%) than in the nonradiation group (3%) (68).

The introduction of the self-expanding metallic stent (SEMS) has made substantial changes in the therapy of esophageal cancer and radiation-induced stenosis and fistula formation. SEMS is typically a cylinder of wire mesh or coil with spring memory for a particular configuration. The first to be introduced was in 1983 [69]; coated and uncoated varieties are now available, and a variety of materials, attachments, and delivery devices are under investigation. The compromised circulation associated with radiation injury can be further worsened by the radial forces from these stents. This may account for the increased perforation rates seen in radiation strictures as compared with esophageal cancers stented before or without associated radiotherapy (36% versus 2.5%) and may contribute to the increased mortality (22% compared to 0%) seen in the same series. [70]

The coated stent is useful for patients suffering tracheoesophageal fistulae. However, enteral nutritional support may be necessary in patients with severe acute symptoms or with difficult-to-manage chronic strictures or fistulae. In patients with uncoated SEMS, later placement of a percutaneous endoscopic gastrostomy is generally feasible because the wire mesh typically embeds securely in the esophagus. Coated SEMS may not be amenable to the same wait-and-see approach.

Stomach

Gastric damage following radiation therapy is rare, but injury can occur when the stomach lies within the radiation field of an adjacent extragastric tumor. [40], [71] Gastric irradiation was used in the past as therapy for peptic ulcer disease because modest doses of radiation could significantly deplete the parietal and chief cell populations inducing gastric atrophy. [40], [72], [73] In an era before effective medical therapy was available, this approach was taken for patients with refractory ulcers. A total dose of 1600 and 1700 cGy given in 10 divided doses resulted in atrophic gastritis with associated decreases in acid and pepsin production. However, in most, acid production returned to normal basal levels within 2 years. [63], [73], [74]

Most patients receiving gastric irradiation are asymptomatic, although hemorrhage due to multiple gastric erosions has been reported as an early complication. [40] Gastric injury from higher exposures can present more dramatically with stenosis, fistulae, and perforation. [75] Strictures are more common in the antrum and cardia

than in the body or fundus. Delayed ulceration may also occur in this setting and is difficult to distinguish from peptic ulcer disease. Radiation-induced ulcers heal poorly because of obliterative arteriolitis within the ulcer bed. Ulceration and gastric strictures usually occur at doses exceeding 4500 to 5000 cGy, and about one half of patients experience clinically significant gastric injury at doses greater than 5500 cGy. [40]

The diagnosis is usually made on the basis of clinical presentation and patient history. Endoscopy may reveal an irregular thickened mucosa similar to the appearance of a gastric neoplasm or an appearance indistinguishable from peptic ulcer disease. Fluorographic upper gastrointestinal studies or computed tomography may be useful in demonstrating the presence of strictures. However, these studies may only show thickening of the gastric wall with an appearance similar to that seen in gastric malignancies. Data are lacking to guide therapy for this uncommon condition. In patients exposed to less than 2000 cGy, simple passage of time may suffice.

Small Bowel

The small intestine is particularly susceptible to radiation injury. Significant epithelial cell death may be observed as early as 4 to 6 hours after irradiation even with the relatively small 1- to 2-Gy fractions commonly used in some radiation therapy regimens. Radiation injury to the small intestine most often occurs in the clinical setting of radiotherapy for gynecologic, urologic, rectal, or retroperitoneal malignancies. The degree of injury appears proportional to radiation dosage delivered to the segment of small intestine that lies within the radiation field. [76], [77] The relatively fixed position of the duodenum, distal ileum, and cecum within the abdominal cavity make these regions particularly vulnerable to radiation-induced injury. [6], [40] At doses of more than 3000 cGy, 90% of patients have significant lesions, 40% at 1000 to 3000 cGy, and 20% at less than 1000 cGy. [78] Previous abdominal surgery that may fix loops of bowel in place is another important risk factor. [7], [79], [80]

Radiation injury to the small bowel can be divided also into acute injury and a chronic syndrome. Nausea, diarrhea, and abdominal discomfort are cardinal manifestations of acute injury. Although many of these symptoms are attributable to epithelial cell death and subsequent loss of mucosal function, symptoms such as nausea and vomiting occurring within in a few hours of irradiation may be mediated by the production of inflammatory cytokines. Fortunately, the epithelium regenerates, and function is restored within 2 weeks, with corresponding resolution of the acute symptoms. [78] This acute syndrome may foreshadow the occurrence of chronic radiation enteritis, at least in children. [81] It is not clear whether this holds for adults, of whom about 5% develop chronic enteritis after exposure. [41]

Symptoms of chronic radiation enteritis typically occur 1 to 2 years after exposure, but presentation 20 years after radiotherapy has been described. [82], [83] and [84] The central mechanism of injury is vascular damage with progressive localized ischemia (see earlier section on “ Pathology”). This leads to ulceration of the mucosa, fibrosis with stricturing, and less frequently, fistula formation or even perforation. Radiographs of the small intestine or colon may reveal evidence of single or multiple strictures or mucosal ulceration, a pattern similar to that observed in Crohn’s disease [4], [85] (Fig. 132-7). Life expectancy is poorer in patients whose enteritis is complicated by fistulae rather than bleeding. [86] Loose stools are the most common symptom and may be caused by a number of abnormalities: bile salt malabsorption in 65%, bacterial overgrowth in 45%, severe fat malabsorption in 40%, rapid intestinal transit in 35%, and lactase deficiency from villous blunting with consequent lactose intolerance in up to 20%. [8], [87]

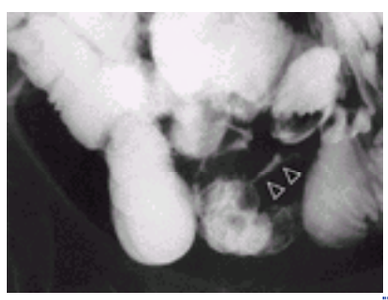

FIGURE 132-7. Barium small bowel radiograph showing an ileal stricture (*arrowheads*) in a patient with diarrhea several years after radiation therapy for cervical cancer. (Courtesy of Dr. Hubert Schaffer.)

Prostheses or mesh material may be used as a preventative measure to displace or protect the small intestine during radiation therapy. [88], [89] and [90] Acute radiation injury is treated supportively and with dose adjustment. [91] Antispasmotics and medications that decrease bowel motility must be used cautiously given that many of these patients are at risk for outright obstruction from underlying tumor or surgical adhesions. Perforation may have a bland presentation in this setting; typical signs of peritonitis, such as fever and severe abdominal pain, may be absent, and adhesions may wall off free air. [92], [93] The data are contradictory for the use of aminosalicylates as a preventative measure. In a prospective double-blind study of sulfasalazine (2 g/day) given to 87 patients undergoing radiation, diarrhea occurred in 55% of the sulfasalazine and 86% of the placebo groups, respectively. [94] However, mesalazine at 4 g/day was not beneficial in another trial. [95]

The treatment of chronic radiation enteritis requires patience and long-term pragmatism. Once symptomatic, this condition is persistent at best and worsens in many, even after surgical resection. [4] Cholestyramine is often a good choice as both a nonspecific antidiarrheal agent and when targeted at bile salt malabsorption. [87] Other antidiarrheals, such as loperamide and diphenoxylate, may also be useful. A newer peripheral opiate antagonist, loperamide- *N*-oxide, was used successfully in a series of 18 patients with radiation enteritis. [96] Treatment was associated with fewer bowel movements and improved bile salt absorption. Bacterial overgrowth can be treated with antibiotics such as amoxicillin clavulanate, ciprofloxacin, or tetracycline. The immunomodulatory effects of these agents may play a role along with their antimicrobial action, but this has not been studied in radiation enteritis.

Specialized enteral formulas, such as elemental diets, are available for treating moderate malnutrition. However, they are costly, and their superiority has not been established. [97] Total parenteral nutrition may be required for severe malnutrition or when enteral feeding is not feasible. Support with total parenteral nutrition allows correction of malnutrition, but recurrent symptoms remain a problem in up to 47% of patients. [98] Methylprednisolone may give additional benefit, but the long-term role of this relatively toxic agent is unknown. [99]

It is prudent to avoid surgery when possible in chronic radiation enteritis, given the likelihood of intraoperative and postoperative difficulties. Surgery is required in about one third of patients for management of strictures causing small bowel obstruction or for perforation. Complications are common (30%), and the mortality rate can be particularly high (more than 5%) for emergency operations. [100] Outright resection of the involved small bowel segment is considered preferable to bypass because of the high incidence of complications such as perforation, bacterial overgrowth, and fistulization. [101]

Colon and Rectum

Radiation proctosigmoiditis is among the most common complications of radiation injury. Many gynecologic and urologic tumors are treated with radiation, and the relatively fixed location of the rectum makes it more susceptible to radiation injury than the rest of the colon. [102] There is a clear dose-related incidence of injury, with acute symptoms common at doses of 3000 to 4000 cGy and with more than one third of patients suffering injury at 6,000 cGy. [103] Complication rates from patient report, rather than record abstraction, are higher still. [104], [105], [106] and [107] When a series of patients undergoing 6 weeks of radiotherapy for nongastrointestinal pelvic tumors was studied at 2-week intervals using sigmoidoscopy, biopsies, and symptom scores, histological and endoscopic abnormalities were both most pronounced after 2 weeks of therapy and improved subsequently. However, symptomatic complaints climbed consistently to the end of the 6-week observation period. [108] One third to one half of patients suffer acute injury with diarrhea and tenesmus during the first months of or after therapy. As a rule, these symptoms resolve over the following 6 months, but about 15% of patients continue to suffer chronic proctitis, manifested by rectal pain, diarrhea, and bleeding. [106] The degree of bleeding has some prognostic value: those patients who do not require transfusion have a high rate of spontaneous remission (70%), low rate of surgery (5%), and low mortality rate (25%); and patients who require blood transfusions or who suffer significant bowel disturbance have a low rate of spontaneous remission (less than 20%), often require surgery (50%), and have substantial mortality (60%). [109]

Many medical approaches have been tried for treatment of patients with acute or chronic radiation injury involving the colon and rectum. Most attention has been paid

to the unfortunate 15% of patients who suffer from chronic radiation proctitis. Unfortunately, no therapy stands out as clearly superior, and risks of each must be weighed against possible benefits. Oral aminosalicylic acid preparations by themselves do not appear effective, although they may help if combined with steroid treatment. [110] Sucralfate enemas by themselves appear effective and well tolerated. [111] Oral sucralfate was beneficial in a series of three patients, but a larger series is needed to evaluate the oral approach. [112]

Short-chain fatty acids are nutrients that promote colonocyte survival and proliferation and have been advocated to accelerate healing in radiation proctitis. In one double-blind study, placebo-treated patients ultimately reached the same level of recovery at 6 months. [113] A crossover study of 20 patients showed encouraging results, but the logistics of drug preparation and the odor of short-chain fatty acids present obstacles to this therapy. [114] Antioxidant therapy has also been investigated as a potential therapy: vitamin E (400 IU three times daily) and vitamin C (500 mg three times daily) were helpful in an open-label series of 20 patients. [115] The application of formalin has been used to treat refractory bleeding from radiation proctitis through a presumed mechanism of obliteration of the telangiectatic vessels through coagulative necrosis. In one series, 50 mL of 4% formalin solution was introduced into patients under sedation and kept in contact with the mucosa for 30 seconds and then cleared away using saline irrigation. Five to six aliquots were used in each session. However, complications were often encountered. [116] In another series of five patients (one treated with topical application of formalin-soaked gauze), severe pain occurred in one patient and acute colitis requiring hydration and antibiotics in another. In another prospective study examining 11 patients treated with four 20-mL aliquots and a total of 15 minutes contact time, all patients had decreased bleeding without reported complications. [117] Hyperbaric oxygen has been used successfully to improve the healing of ulcers in radiation proctitis. [118], [119]

Surgical and endoscopic treatments may be tried when bleeding from telangiectasia is the predominant manifestation of radiation injury (Fig. 132-8). It is critical to weigh potential benefit against risk in this setting. Patients who do not require transfusion generally do not require an interventional approach. Laser therapy can be associated with nonhealing ulcers and may aggravate bleeding or cause outright ulceration. Argon plasma coagulation is designed to create a more controlled level of treatment-induced injury and is used to obliterate natural gastrointestinal telangiectasias. Its introduction as an endoscopic treatment led to some enthusiasm about its use in radiation-related bleeding. In comparison to treatment of classic telangiectasias from other causes, argon plasma coagulation of telangiectasias associated with radiation injury appears to require more sessions to achieve hemostasis but appeared effective in several small series. [120], [121], [122] and [123]

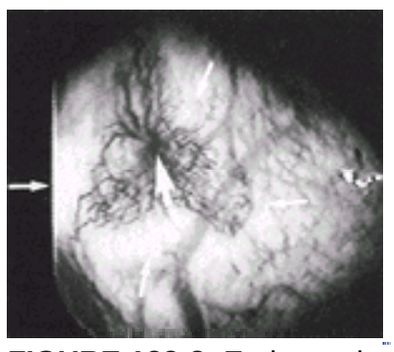

FIGURE 132-8. Endoscopic appearance of telangiectasia (*short arrows*) with prominent central arteriole (*long arrow*) and surrounding pallor in a patient with radiation proctitis.

Individual episodes of bowel obstruction may be treated conservatively with bowel rest, intravenous fluids, and nasogastric decompression, but recurrent obstruction or an unresponsive episode may require surgery. Surgical intervention for stricture generally consists of resection of the diseased segment, with primary anastomosis. Outcome is best if sufficient resection is performed to ensure that one margin is free from radiation injury. [4], [124]

Liver

Older administration techniques made liver injury common when therapeutic radiation was directed at tumors within the hepatic parenchyma. The therapeutic index using these older techniques was unfavorable, with doses of 3000 to 3500 cGy offering consistent injury and inadequate therapeutic effect. Thus, hepatic irradiation lost popularity until more modern technology, such as nonaxial planes of administration and sophisticated modeling, made it possible to deliver higher doses safely. [125]

Hepatic injury following irradiation typically manifests about 4 to 8 weeks after exposure but may occur as early as 2 weeks or as late as 7 months. In reports from the 1960s, more than one fourth of patients with radiation hepatitis had a clinical presentation consistent with Budd-Chiari syndrome or obstruction of suprahepatic veins. [126], [127] Nonspecific symptoms, such as malaise, fatigue, and weight gain, are common. Ascites is often present but may be below the threshold for bedside detection. Right upper quadrant pain is also common, but jaundice is not, in keeping with the characteristic laboratory findings in which alkaline phosphatase levels are elevated with minimal increases in bilirubin and mild elevation of transaminases. [128], [129] The acute illness is generally well tolerated and usually resolves with supportive care, but development of chronic radiation-induced hepatitis has also been described. [130] Computed tomography findings may be helpful. In one series, 23 of 31 patients (74%) showed a low-attenuation area adjacent to the hepatic tumor on tomographic scans in the targeted radiation field. A sharp, straight interface was rarely seen at the treatment margin. These findings were maximal 2 to 3 months after completion of therapy and persisted for up to 3 months. Atrophy in the treated segment or lobe and hypertrophy of the untreated liver were seen in several patients. [131] With proton-beam irradiation, abnormalities noted on computed tomography scans may persist as long as 42 months. [132] Given the setting and frequency with which combined modalities of treatment are used, the differential diagnosis for this condition must always include metastatic disease and drug toxicity.

Liver injury is a particular concern with bone marrow transplantation, in which the combination of total-body irradiation and high-dose chemotherapy for initial cytoreductive therapy is commonly used. Venoocclusive disease, presenting with weight gain (fluid retention), jaundice, and tender hepatomegaly, is common, occurring in 10% to 60% of patients within several weeks after transplantation. [48], [133], [134] Weight gain and increasing liver size with tenderness may herald the onset of venoocclusive disease and usually precede the development of hyperbilirubinemia. [48] Serum aminotransferases may also be elevated. Mortality rates from venoocclusive disease in this setting range from 0% to 67%. [48] Supportive treatment with diuretics and careful fluid and electrolyte management are usually effective in mild to moderately severe venoocclusive disease. Treatment of severe cases of venoocclusive disease has been less successful.

Chemotherapeutic regimens that potentiate hepatic injury when combined with therapeutic irradiation include mitomycin C, MOPP (nitrogen mustard, vincristine, procarbazine, and prednisone), CHOP (cyclophosphamide, doxorubicin, vincristine, and prednisone), CCNU (2-chloroethyl, 3-cyclohexyl, 1-nitrosurea), and pro-MACE (prednisone, methotrexate, doxorubicin, cyclophosphamide, etoposide). [135], [136] Injury from combined therapy is clearly more severe than that seen from radiation alone and carries an overall mortality rate of 30% to 50%.

Treatment of patients with isolated radiation injury to the liver is generally supportive. Salt restriction and diuretics are often helpful. Improvement over months is the rule, with a mortality rate of less than 20%, although those patients who become jaundiced may have a worse course. [126] Treatment is similar with combined injury, but more aggressive therapy may be required.

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CHAPTER 133

Philip S. Schoenfeld*

CLINICAL DECISION MAKING

WHAT IS EVIDENCE-BASED MEDICINE?

Clinical decision making is facilitated by evidence-based medicine. Evidence-based medicine (EBM) is “the conscientious and judicious use of current best evidence from clinical care research in the management of individual patients.” [1]

Conscientious use implies that physicians review articles about clinical research and apply this information to clinical decision making.
Current best evidence from clinical care research implies that physicians systematically appraise the methods and results of clinical research articles. **This is the focus of this chapter.**
Judicious use implies that clinical experience facilitates a thoughtful assessment of an individual patient’s unique situation. [2] The risks and benefits of different tests or treatments must be balanced against an individual patient’s preferences and risks from a specific disease.

The medical literature is expanding at an exponential rate, [2] and the time available for reading may be hurried and fragmented. Thus, many physicians may feel that a systematic review of clinical research is impractical. In recognition of this dilemma, EBM provides frameworks (Table 133-1 and Table 133-2) for the rapid and systematic appraisal of clinical research studies. Studies using improper methodology produce biased results. Use of EBM frameworks (see Table 133-1 and Table 133-2) allows rapid review of abstracts of published research, identifies studies likely to produce biased results, and “tosses out” those studies.

1. When are diagnostic tests necessary?
   a. Is the pretest probability of disease so high or so low that further diagnostic tests are not needed?
   b. How is pretest probability estimated?
2. Assessing study design
   a. Was there a blinded comparison of the diagnostic test to a gold standard test?
   b. Were negative study tests verified by performing the gold standard test?
   c. Was the diagnostic study tested in patients similar to the population in whom the test will be used?
3. Getting from pretest probability to posttest probability
   a. Interpret and apply data about sensitivity and specificity.
   b. Use likelihood ratios to maximize the data from a diagnostic test.
4. Applying the results of clinical research to your patient
   a. Are the results applicable to your patient?
   b. Is the test available with reproducible accuracy?

From ref. 3.

TABLE 133-1 Critical Approach to an Article About a Diagnostic Test

1. Assessing study design
   a. Did the study use concealed random allocation?
   b. Were patients and physicians blinded about allocation to treatment or placebo groups?
   c. Did groups receive equal treatment (co-interventions) except for the experimental study treatment?
   d. Did the study use an intention-to-treat analysis?
   e. Was follow-up of study patients complete?
2. Clinically significant results and statistically significant results
   a. Estimating treatment-effect size: relative risk reduction, absolute risk reduction, and number needed to treat
   b. Evaluating the sample size in a non-statistically significant study: were enough patients entered into the study?
   c. What is the precision of the treatment effect: how large are the 95% confidence intervals?
3. Applying the results of clinical research to your patient
   a. Are the results applicable to your patient?
   b. Are the potential treatment benefits worth the potential side effects, cost, and inconvenience to your patient?

From ref. 19.

TABLE 133-2 Critical Approach to an Article About a Therapy

This chapter outlines systematic frameworks for the critical appraisal of studies about diagnostic tests and therapies; reviews different presentations of study results; and discusses the application of study results to patient care and clinical decision making. Clinical scenarios are used to illustrate the practical application of EBM concepts.

CRITICAL APPRAISAL OF AN ARTICLE ABOUT A DIAGNOSTIC TEST

Case Scenario A—Part I

A 65-year-old woman presents for colon cancer screening after watching a report about this topic on a morning news show. The patient has never been screened for colon cancer. You describe the potential benefits and risks of colonoscopy. However, the patient is hesitant to undergo colonoscopy because she heard that colon perforations can occur during colonoscopy. She has heard that a new “x-ray” test can look for polyps in the colon, and she asks if that would be an appropriate test for

her. Specifically, you consider the following questions:

1. What is the probability of colon polyps in this woman?
2. How accurate is “virtual” or CT colonoscopy for the diagnosis of colon polyps?

You decide to apply EBM frameworks [3] to appraise a recent study [4] that examines the accuracy of CT colonoscopy and answers these questions.

When Are Diagnostic Tests Necessary?

Is the Pretest Probability of Disease So High or So Low that Further Diagnostic Tests Are Not Needed? The pretest probability defines the likelihood that a patient has a specific disorder before any diagnostic test result is available. Diagnostic tests should be ordered when the pretest probability is intermediate. Thus, if the pretest probability of a specific disorder is 50%, then accurate diagnostic tests may rule out the disorder or definitively confirm the presence of the disorder. Conversely, if the pretest probability is very high or very low, then ordering additional diagnostic tests may be unnecessary. Several examples illustrate the concept of pretest probability. What diagnostic tests would you order for a hospitalized patient who suddenly develops diarrhea? Stool tests for ova and parasites are routinely ordered to evaluate diarrhea, but the pretest probability of a parasitic infection in a hospitalized patient with new-onset diarrhea approaches zero. [5] In this situation, the pretest probability is so low that stool tests for ova and parasites should not be obtained. Conversely, patients with peptic ulcers are rarely tested for *Helicobacter pylori* in some countries (e.g., Armenia) because the prevalence of *H pylori* approaches 100%. Thus, these peptic ulcer patients are automatically treated for *H pylori* without performing diagnostic tests to rule in this diagnosis. What constitutes an intermediate pretest probability that suggests the need for diagnostic tests? Clinical judgment governs this decision, based partly on the cost, accuracy, and side effects of the diagnostic test and the consequences of a “missed” diagnosis (i.e., if a missed diagnosis may have fatal consequences, then clinicians will have a lower threshold to order diagnostic tests).

How Is Pretest Probability Estimated? When physicians evaluate a patient’s complaint, they intuitively use their experience and clues from the history and physical exam to estimate pretest probabilities for different medical disorders. However, these estimates are frequently inaccurate. [6] The prevalence of a disorder (the proportion of patients with a specific disorder at a distinct point in time) may provide a more accurate estimate of pretest probability. Valid studies about the prevalence of a particular diagnosis should meet several methodologic criteria. [7] First, the technique for confirming the diagnosis should be explicit and credible. For example, colonoscopy would be an explicit and credible test to estimate the prevalence of colon polyps. However, flexible sigmoidoscopy, which does not examine the right side of the colon, might not be a credible test. Second, the technique for confirming the diagnosis should be applied to consecutive patients who present with a specific complex of symptoms, physical exam signs, or laboratory results. For example, the study about prevalence of colon polyps might require that all study patients are older than 50 years of age and asymptomatic (e.g., no hematochezia or abdominal pain). Finally, clinicians should determine whether the characteristics of their patients are similar to those of the patients examined in the study. For example, if your patient has a family history of colon cancer in a first-degree relative, then this patient’s risk for colon cancer and colon polyps would be higher than among patients without a family history of colon cancer.

Case Scenario A–Part II

A recent abstract estimates the pretest probability for colon polyps in the case scenario patient. [8] In this study, asymptomatic women referred for colon cancer screening underwent colonoscopy. Colonoscopy is a credible diagnostic test to define the prevalence of colon polyps. Consecutive women referred for colon cancer screening were offered colonoscopy. Patients were asymptomatic (e.g., denied history of hematochezia, change in bowel habits, or abdominal pain) and were screened with complete blood cell counts and fecal occult blood tests to rule out anemia or occult gastrointestinal (GI) bleeding. In this trial, the prevalence of colon polyps among asymptomatic women was 21%, and the prevalence of advanced colon polyps (i.e., polyps larger than 10 mm, villous adenomas, adenomas with high-grade dysplasia, or colorectal carcinoma) was only 3%. Although the prevalence of advanced colon polyps is not very high, you recognize that a missed diagnosis of advanced colon polyps could lead to a fatal colon cancer. Therefore, you decide that diagnostic testing for colon polyps is warranted, and you proceed to review the study about the diagnostic accuracy of virtual colonoscopy. [4]

Assessing Study Design

Was There a Blinded Comparison of a Diagnostic Test to a Gold Standard Test? A *gold standard* or reference standard refers to a diagnostic test that definitively establishes the presence or absence of disease. For example, a study examining the diagnostic accuracy of magnetic resonance cholangiography (MRC) for the diagnosis of choledocholithiasis used endoscopic retrograde cholangiopancreatography (ERCP) as the reference test. [9] Reference standards are usually costlier, riskier, or more inconvenient than new diagnostic tests being studied. Otherwise, performing the reference standard would be more sensible. Biopsies, autopsies, surgical pathology, or even prolonged patient follow-up may also be reference standards that determine the presence or absence of disease. The results from the reference standard and the diagnostic test should be examined by investigators who do not know the patient’s history or the results of other tests. This “blinded” comparison is especially important when the interpretation of test results are subjective. For example, in the study assessing the diagnostic accuracy of MRC, [9] the radiologist’s interpretation of the MRC would be biased if he or she knew that the ERCP demonstrated choledocholithiasis. Establishing a reference standard test may be an elusive goal. New technologies may be more accurate than the established reference standard test. If a potentially poor diagnostic test is being used as the reference standard, then the diagnostic accuracy of the new test may appear worse than it truly is. For example, one study evaluated the accuracy of ultrasonography to diagnose cholelithiasis but used oral cholecystograms as the gold standard test. [10] In this study, only patients with abnormal cholecystograms were referred for surgery. Five patients in this study had positive ultrasounds for cholelithiasis, but normal cholecystograms. Based on this study’s analysis, these positive ultrasounds were false-positive test results (i.e., patients did not truly have disease). Ultimately, several of these patients underwent cholecystectomy because of recurrent symptoms, confirming the presence of cholelithiasis and demonstrating that oral cholecystograms may not be an adequate gold standard test.

Were Negative Study Results Verified by Performing the Gold Standard Test? Study results will be distorted if investigators use results of the new diagnostic test to decide whether or not to perform the gold standard test. This *verification bias* may produce biased study results in more than 50% of diagnostic test studies. [11] For example, in the study about the accuracy of MRC for choledocholithiasis, [9] a few patients with normal MRCs actually had choledocholithiasis on ERCP. If ERCPs were withheld from these patients (because investigators assumed choledocholithiasis was absent based on the normal MRCs), then MRC would appear more accurate than it truly is.

Was the Diagnostic Study Tested in Patients Similar to the Population in Whom the Test Will Be Used? Patients with end-stage disease may have grossly abnormal diagnostic test results, making it easy to differentiate healthy people from ill patients. For example, virtual colonoscopy might easily differentiate patients with normal colons from patients with end-stage, near-obstructing colon cancer. The real value of a diagnostic test is its ability to identify patients with early manifestations of disease (e.g., colon polyps) that could be easily confused with a normal finding (e.g., stool). To assess the accuracy of a diagnostic test properly, the test should be studied in a broad range of patients, similar to the patients seen in clinical practice. [12] The best example of this *spectrum bias* may be carcinoembryonic antigen (CEA). CEA was evaluated as a diagnostic test for colorectal cancer. Initially, the test was studied in patients with advanced colorectal cancer and in normal controls. [13] The results demonstrated that almost all (98%) of the advanced colorectal cancer patients had elevated CEA levels, whereas almost all normal controls had low levels of CEA. These initial results raised hope that CEA might be a useful screening tool for colorectal cancer. When this test was studied in a broad population of patients with early-stage colorectal cancer and patients with other GI disorders, the test was inaccurate and unable to differentiate patients with early cancer from patients with other disorders. [14]

Case Scenario A—Part III

The study assessing the diagnostic accuracy of “virtual” or computed tomographic (CT) colonoscopy used conventional colonoscopy as a gold standard, which is appropriate. Conventional colonoscopy was performed in all patients who underwent CT colonoscopy, which eliminates verification bias. However, the accuracy of CT colonoscopy was assessed in patients with an increased risk for colon polyps (i.e., patients with a personal history of colon polyps, family history of colon cancer, or positive flexible sigmoidoscopy or positive fecal occult blood test). Thus, the prevalence of colon polyps in this population may be higher than in the asymptomatic, average-risk woman in the case scenario. However, there is no evidence that colon polyps are larger or more easily identifiable in this population than in asymptomatic patients. Overall, you determine that the article has adequate methodology and you proceed to review the results.

Getting from Pretest Probability to Posttest Probability

Interpreting and Applying Data About Sensitivity and Specificity Sensitivity and specificity can be calculated from the classic 2 × 2 table (Fig. 133-1). The 2 × 2 table is completed by filling in the true-positive test results (positive test result when disease is present), false-positive test results (positive test result when disease is absent), true-negative test results (negative test result when disease is absent), and false-negative test results (negative test result when disease is present). For example, a recent study assessed the accuracy of ferritin for the diagnosis of iron deficiency anemia, using bone marrow aspirates as a gold standard for diagnosis of iron deficiency. [15] This trial found that 150 patients had high ferritin levels (more than 45 µg/L) and 85 patients had low ferritin levels (45 µg/L or less). Of the 85

patients with low ferritin levels, 70 had iron deficiency anemia (true-positive test results) and 15 did not (false-positive test results). Of the 150 patients with high ferritin levels, 135 patients did not have iron deficiency anemia (true-negative test results), and 15 did (false-negative test results).

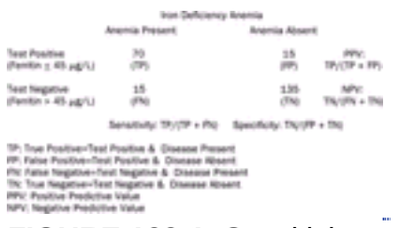

FIGURE 133-1. Sensitivity and specificity of ferritin for the diagnosis of iron deficiency anemia (36% prevalence of iron deficiency anemia) in an elderly population. (From ref. [15].)

In the 2 × 2 table (see Fig. 133-1), the formulas for sensitivity (the percentage of patients with the disease in whom the test results are positive) and specificity (the percentage of patients without the disease in whom the test results are negative) are defined. Using these formulas, the sensitivity of ferritin (with a cutoff point of 45 µg/L of ferritin) for iron deficiency anemia is 82%, and the specificity is 90%. Unfortunately, sensitivity and specificity "work backward" from clinical practice, evaluating patients with known disease and providing data about the presence or absence of certain diagnostic test results. However, patients present with symptoms and diagnostic test results, and we "work forward" with these results to determine the likelihood of disease. The positive predictive value (PPV) and negative predictive value (NPV) from the 2 × 2 table (see Fig. 133-1) provide these data. The formulas for PPV (the percentage of patients with positive test results who have the disease) and NPV (the proportion of patients with negative test results who do not have the disease) are also provided in the 2 × 2 table. Using these formulas, the PPV for low ferritin level (45 µg/L or less) in the diagnosis of iron deficiency anemia is 82%. Hence, 82% of patients with ferritin levels of 45 µg/L or less had iron deficiency anemia. The NPV for high ferritin level is 90%, or 90% of patients with ferritin levels of more than 45 µg/L did not have iron deficiency anemia. Before clinicians apply PPV and NPV to their individual patients, the limitations of these statistics must be recognized. Sensitivity and specificity usually remain relatively constant, although they may vary slightly depending on the severity of disease in a specific patient population. However, PPV and NPV vary widely depending on the prevalence of the disease. For example, consider if all internal medicine admissions to a hospital were screened with ferritin for iron deficiency anemia. In this diverse population, the prevalence of iron deficiency anemia might only be 5%, although the prevalence of iron deficiency anemia was 36% among the elderly anemic patients in the ferritin–iron deficiency anemia study. [15] Assuming that the sensitivity and specificity remain constant, a new 2 × 2 table (Fig. 133-2) can be constructed, producing a significantly lower PPV of 32% and a significantly higher NPV of 99%. Hence, when prevalence of a disease decreases, the PPV decreases, and the NPV increases.

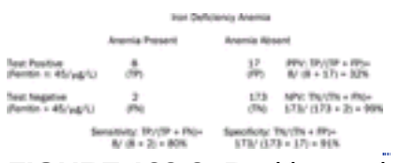

FIGURE 133-2. Positive and negative predictive value of ferritin in the diagnosis of iron deficiency anemia in a hypothetical population of 200 hospitalized patients (5% prevalence of iron deficiency anemia).

Use Likelihood Ratios to Maximize the Data from a Diagnostic Test Likelihood ratios express the likelihood that a particular range of values for a diagnostic test will be found in a patient with a specific disease. They overcome two weaknesses of sensitivity/specificity and PPV/NPV. First, likelihood ratios predict the presence of disease based on a diagnostic test result (similar to PPV/NPV), but likelihood ratios do not change with different disease prevalence (unlike PPV/NPV). Second, studies reporting sensitivity and specificity usually provide data about the accuracy of a diagnostic test around only one value. For example, the ferritin–iron deficiency anemia study [15] calculated sensitivity and specificity of ferritin around a cutoff point of 45 µg/L (i.e., ferritin level less than or equal to 45 µg/L is consistent with iron deficiency anemia and ferritin level greater than 45 µg/L is not consistent with iron deficiency anemia). Intuitively, a patient with a ferritin level of 5 µg/L is more likely to have iron deficiency anemia than a patient with a ferritin level of 40 µg/L, but the sensitivity and specificity cannot differentiate between these two patients. However, likelihood ratios are usually calculated for multiple ranges of diagnostic test results, thereby maximizing the information from a diagnostic test. Thus, likelihood ratios may facilitate the application of diagnostic test results to patient care. Mathematically, the likelihood ratio for a positive test result is as follows:

$$\frac{\text{sensitivity}}{1 - \text{specificity}} \quad \text{or} \quad \frac{\text{true positive rate}}{\text{false positive rate}}$$

The likelihood ratio for a negative test result is as follows:

$$\frac{1 - \text{sensitivity}}{\text{specificity}} \quad \text{or} \quad \frac{\text{false negative rate}}{\text{true negative rate}}$$

In the ferritin–iron deficiency anemia study, [15] likelihood ratios were calculated for four ranges of ferritin: less than or equal to 18 µg/L, 19 to 45 µg/L, 46 to 100 µg/L, and more than 100 µg/L (Fig. 133-3).

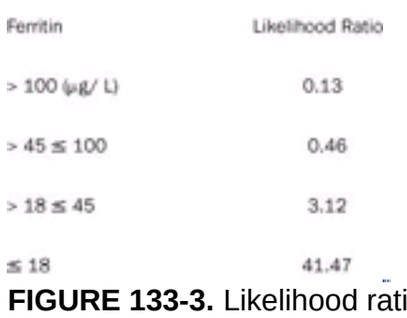

FIGURE 133-3. Likelihood ratios for ferritin in the diagnosis of iron deficiency anemia.

By using a nomogram, [16] likelihood ratios easily convert pretest probabilities to posttest probabilities (Fig. 133-4). Simply place the base of a ruler at the pretest probability and angle the ruler through the likelihood ratio in order to see the posttest probability. For example, clinicians might assume that a 70-year-old patient with a history of myocardial infarction, daily use of aspirin, an MCV of 78, and a hemoglobin level of 11 g/dL has a 50% pretest probability of iron deficiency anemia (moderately higher than the prevalence of iron deficiency anemia among a general population of elderly anemic patients). If this patient has a ferritin level of 5 µg/L, then the pretest probability of 50% and the likelihood ratio of 41 produces a 98% posttest probability that iron deficiency anemia is present. Conversely, if this patient has a ferritin level of 110 µg/L, then the posttest probability is 10%.

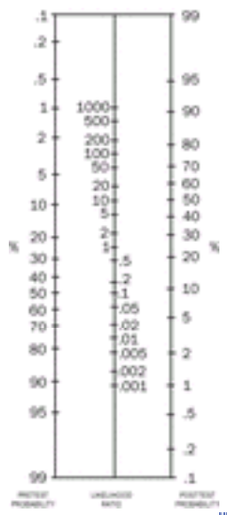

FIGURE 133-4. Nomogram for interpreting diagnostic test results. (Adapted from ref. [16].)

Case Scenario A—Part IV

CT colonoscopy found all three colon cancers in study patients. For patients with large polyps (at least 10 mm), CT colonoscopy produced sensitivity of 96% and specificity of 96% with likelihood ratio for a positive test of 24 and likelihood ratio for a negative test of 0.04. For patients with polyps of any size, CT colonoscopy produced sensitivity of 82% and specificity of 84% with likelihood ratio for a positive test of 5 and likelihood ratio for a negative test of 0.2. The prevalence of large (more than 10 mm) polyps is about 3%, and the prevalence of any colon polyps is about 21%. Therefore, using likelihood ratios, a positive CT colonoscopy for large polyps predicts a 42% posttest probability of large polyps. A positive CT colonoscopy for colon polyps (any size) predicts a 60% posttest probability of colon polyps (any size).

Applying the Results of Clinical Research to Your Patient

Are the Results Applicable to Your Patient? A study may have valid methodology, and the results may indicate that the diagnostic test is accurate. However, if the study patient population is much different than your patients (e.g., geriatric versus pediatric patients, symptomatic versus asymptomatic patients), then the diagnostic test may perform much differently in your patients. If your own patient meets all of the inclusion criteria of a study and the prevalence of disease is similar in your setting, the results are probably applicable to your patient. Physicians need to use their own judgment when there are no definitive research data about the accuracy of the diagnostic test in specific patients. For example, no research is available about the accuracy of CT colonoscopy among asymptomatic individuals referred for colon cancer screening. These average-risk patients may have a lower prevalence of polyps than high-risk patients (e.g., patients with a personal history of colon polyps), but it is unlikely that average-risk patients form different types of polyps than high-risk patients. Thus, results from the CT colonoscopy study [4] should be applicable to the case scenario patient.
Is the Test Available with Reproducible Accuracy? Even when a diagnostic test is adequately described to permit replication in your clinical setting, the test may not be available. Some diagnostic tests require special equipment or skilled examiners, which may not be widely available. For example, CT colonoscopy is not widely available outside academic medical centers in 2003. Many diagnostic tests, including CT colonoscopy, require a subjective interpretation. A well-designed study of a diagnostic test will have the diagnostic test results reviewed by examiners with different levels of expertise. If experienced and inexperienced practitioners produce similar interpretations of diagnostic test results, then you may be assured that study results may be reproduced in your own clinical setting. For example, previous studies [17] have demonstrated that both experienced and inexperienced radiologists can accurately diagnose choledocholithiasis on MRC.

Case Scenario A—Part V

CT colonoscopy is available in your hospital, so you can provide this diagnostic test for your patient. She is an asymptomatic, average-risk patient, and the study examined the accuracy of CT colonoscopy in high-risk patients. However, you doubt that high-risk patients will develop different polyps, so you decide that the results are applicable to your patient. The CT colonoscopy study [4] did not have multiple radiologists review CT colonoscopy results, so you are uncertain whether your hospital's radiologists can produce similarly accurate results with CT colonoscopy. Overall, you decide that CT colonoscopy will not be your routine test of choice for colon cancer screening at this time. Nevertheless, based on your interpretation of your patient's wishes as well as the very high accuracy of CT colonoscopy for large polyps, you decide to proceed with CT colonoscopy for this individual patient.

CRITICAL APPRAISAL OF AN ARTICLE ABOUT A THERAPY

Case Scenario B—Part I

You are seeing a 75-year-old man with osteoarthritis of the hip and a past history of a nonsteroidal antiinflammatory drug (NSAID)– associated bleeding ulcer. His primary care physician has referred this patient because treatment of osteoarthritis with acetaminophen and physical therapy has been ineffective. The primary care physician wants to start the patient on a cyclooxygenase-2 (COX-2)–selective NSAID (coxib), but she is still concerned about the risk for recurrent NSAID-associated bleeding ulcers. Specifically, she asks these questions:

1. Among NSAID-using patients, do coxibs cause fewer serious GI complications (i.e., bleeding, perforation, or obstruction) than conventional NSAIDs?
2. If coxibs do cause fewer serious GI complications, how large is the reduction?

You recently saw an article [18] that addresses this issue. The VIGOR trial randomized more than 8000 rheumatoid arthritis patients to receive rofecoxib or naproxen and examined the frequency of serious NSAID-associated GI complications. You apply EBM frameworks (see Table 133-2) to appraise the study, [19] determining whether the study has appropriate methodology that is likely to produce accurate results.

Assessing Study Design

Did the Study Use Concealed Random Allocation? A patient's response to treatment may be influenced by many factors other than treatment. Age, severity of illness, and comorbid medical problems will affect a patient's prognosis and limit the effect of treatment. Therefore, these factors should be distributed equally between the treatment and placebo group (or between a "new" treatment group and a control group) in order to identify a "true" or accurate estimate about the effectiveness of the treatment. In a randomized trial, every patient has an equal chance of receiving treatment or placebo when they enter the trial, so that these factors are usually distributed equally between the treatment and placebo group. In a nonrandomized trial, physicians determine which patients enter the treatment group and which patients enter the placebo group. For unclear reasons, patients with a good prognosis are disproportionately entered into the treatment group in a nonrandomized trial. Patients with good prognosis are more likely to have a favorable outcome, regardless of the effectiveness of treatment. [20] Nonrandomized trials illustrate that these studies demonstrate larger treatment effects than randomized trials and are more likely to demonstrate a false-positive result. [20] Concealment of allocation maintains the integrity of randomization. In concealed random allocation, researchers who obtain informed consent and enroll patients into a trial do not know whether the next study patient will receive treatment or placebo. If concealment of allocation was used, then the methods section of a study should indicate this (e.g., sealed, opaque, sequentially numbered envelopes were opened after patients gave informed consent; a central coordinating center was called for treatment assignment after a patient gave informed consent). Researchers may subconsciously wish to show that the therapy being studied is superior to the control therapy. Therefore, without concealed allocation, researchers may subconsciously assess a patient's prognosis and guide patients with good prognosis into the treatment or new therapy group and guide patients with a bad prognosis into the placebo or control therapy group.
Were Patients and Physicians Blinded About Allocation to Treatment or Placebo Groups? Blinding simply means that the patients and physicians do not know if the patient received placebo or treatment. This is particularly important when the outcome is subjective. For example, the VIGOR trial [18] assessed the frequency of dyspepsia among study patients. However, the assessment of dyspepsia is quite subjective and variable. Both the patient and the study physicians may assume that coxibs, like rofecoxib, are less likely to cause dyspepsia than conventional NSAIDs, like naproxen, possibly introducing bias into their subjective assessment of dyspepsia. Blinding both the patients and health care personnel (double blinding) is the best method to avoid this bias. Double blinding has been demonstrated to prevent inflated estimates of treatment benefit in randomized trials. [21] *Randomization, concealed allocation, and double blinding are the only techniques that have*

been shown to reduce inflated estimates of treatment benefit in epidemiologic studies. [20], [21] The importance of blinding is self-evident, but it may be difficult to ensure. For example, a recent double-blind, randomized controlled trial compared tegaserod with placebo among patients with constipation-predominant irritable bowel syndrome (IBS). [22] In this trial, the study endpoint was global assessment of improvement in IBS symptoms, which is clearly a subjective outcome. However, tegaserod, a 5-HT $_3$ partial agonist, stimulates colonic motility and rapidly increases the frequency of bowel movements in constipated patients. Thus, patients using tegaserod may have noted the rapid increase in frequency of bowel movements and assumed that they were using tegaserod. This knowledge may have "unmasked" the blinding process and biased the subjective assessment of improvement in IBS symptoms. One possible resolution: ask patients to guess whether they had received tegaserod or placebo at the end of the trial. If 50% of patients receiving tegaserod guess correctly and 50% of patients using tegaserod guess incorrectly, then the blinding process still worked. Even when the study outcome is objective, it is still helpful to maintain double blinding. For example, one study of primary prevention of bleeding esophageal varices with beta-blockers examined overall mortality as a study outcome. [23] This is certainly an objective outcome, but double blinding was still maintained in this study because of the risk for co-interventions. Co-interventions are treatments other than the study treatment that may affect the outcome, especially when the co-interventions are unequally distributed between the treatment and placebo groups. For example, isosorbide-5-mononitrate has been shown to reduce variceal bleeding. [24] Without double blinding, the physician or the patient might be tempted to start isosorbide-5-mononitrate in one group more than the other group. Double blinding limits the unequal use of cointerventions.

Did the Groups Receive Equal Treatment (Co-Interventions) Except for the Experimental Study Treatment? Additional treatments or co-interventions are most problematic when they are very effective, such as the additional use of isosorbide-5-mononitrate in a study about the effectiveness of beta-blockers to prevent variceal bleeding. Although the methodology of a study may be strengthened if all co-interventions are withheld, it may be unethical to withhold effective treatment to patients enrolled in a study. Research does not occur in a vacuum, and patients receive additional treatments or co-interventions to optimize their health while participating in a study. To balance this conflict, the indications to use co-interventions should be clearly described in the Methods section, their use should be limited, and their use should be recorded for later analysis. The VIGOR trial [18] clearly described the use of co-interventions in the Methods section. Patients were allowed to take other treatments for rheumatoid arthritis, including acetaminophen, methotrexate, and corticosteroids, even though concurrent use of corticosteroids with NSAIDs increases the risk for serious NSAID-associated GI complications. [25] The frequency of serious NSAID-associated GI complications among corticosteroid-using patients in patients using rofecoxib and patients using naproxen was recorded for subgroup analysis. Concurrent use of other NSAIDs was forbidden because use of multiple NSAIDs increases the risk for NSAID-associated GI complications. [25] Patients on NSAIDs develop dyspepsia, and over-the-counter preparations to treat dyspepsia are readily available. So, recognizing that trials do not occur "in a vacuum," researchers allowed patients to use antacids or *limited* doses of H $_2$-receptor antagonists. In *limited* doses, these medications are unlikely to affect the occurrence of serious NSAID-associated GI complications, but allowing use of these medications will treat dyspepsia.

Did the Trial Use an Intention-to-Treat Analysis? In almost every study, some patients stop taking the study medication (treatment or placebo). They are noncompliant, or they believe that study medication is causing side effects. An intention-to-treat analysis includes all randomized patients in the final data analysis, regardless of whether the patients completed the study or were compliant. An adherence-to-protocol analysis excludes patients who did not complete the study owing to noncompliance or side effects. An intention-to-treat analysis preserves the value of randomization because some poor-prognosis patients may not be able to complete the study, but these patients should be included in order to understand fully the true effectiveness of a treatment. An example best illustrates the concept of intention-to-treat analysis. In a randomized trial comparing surgical esophageal transection versus standard therapy with sclerotherapy, [26] some patients randomized to surgery never had the operation because they became too ill or refused surgery. These ill and noncompliant patients have a poor prognosis, regardless of whether or not they undergo surgery. If researchers excluded these poor-prognosis patients from the surgery group analysis (or switched these patients to the sclerotherapy group in the middle of the study), then even a useless surgical procedure might appear effective compared with sclerotherapy. Systematic exclusion of patients with a poor prognosis from one treatment group, rather than random loss of patients from both groups, leads to a bias in favor of one therapy. An adherence-to-protocol analysis only includes patients who complete therapy, estimating the likelihood of a good outcome for patients who complete therapy. However, an adherence-to-protocol analysis may lose the value of randomization because patients unlikely to have a good outcome are eliminated from analysis. For example, if a hypothetical therapy cured 50% of patients and caused life-threatening side effects in the other 50% of patients, the adherence-to-protocol analysis would indicate benefit with therapy, but the intention-to-treat analysis would indicate that the hypothetical therapy was much less effective or potentially even dangerous. Because an intention-to-treat analysis includes both compliant and noncompliant patients, it estimates the likelihood of achieving a desired outcome when a patient first starts a treatment, consistent with "real-world" medical practice. Robust studies will present both an intention-to-treat analysis and an adherence-to-protocol analysis, allowing the reader to assess fully the results and to make up their own mind about the benefits of treatment.

Was Follow-Up of Study Patients Complete? When patients drop out of studies, several explanations are possible. The patients may have disappeared because they experienced a side effect or even died from the study treatment, or they may stop follow-up because their symptoms resolved. How can you determine whether the loss to follow-up biased study results? In a treatment study with a positive result, the study results could be recalculated, assuming that all treatment group patients lost to follow-up had a poor outcome and assuming that all control group patients lost to follow-up had a good outcome. If the recalculated results still demonstrate a treatment benefit, then the loss to follow-up did not cause a falsely positive study result. To avoid recalculations, one short cut is available [19]: only rare studies will still demonstrate a positive treatment effect upon recalculation of study results if more than 15% to 20% of patients are lost to follow-up.

Case Scenario B—Part II

The VIGOR trial randomized more than 8000 rheumatoid arthritis patients to receive rofecoxib or naproxen and examined the frequency of serious NSAID-associated GI complications. The study met all criteria for a well-designed study: randomization, concealed allocation, equal and minimal use of co-interventions, use of double blinding, use of an intention-to-treat analysis, and minimal patients lost to follow-up. Based on this analysis, you decide that the study is likely to produce accurate and unbiased results, and you proceed to review the statistical representations of the results.

Clinically Significant Results and Statistically Significant Results

Estimating Treatment-Effect Size: Relative Risk Reduction, Absolute Risk Reduction, and Number Needed to Treat The relative risk reduction (RRR) expresses the decreased risk for an adverse outcome in the treatment group compared with the risk for an adverse outcome in the placebo or control group. For example, in a randomized, double-blind, intention-to-treat trial, patients with endoscopically treated bleeding ulcers received either intravenous proton pump inhibitor (PPI) or placebo. The study endpoint was recurrent peptic ulcer bleeding: 22.5% of patients receiving placebo had recurrent bleeding, whereas 7% of patients receiving intravenous PPI had recurrent bleeding. [27] The RRR may be calculated as follows:

$$\frac{\text{\% placebo patients with bleeding} - \text{\% treatment patients with bleeding}}{\text{\% placebo patients with bleeding}} = \frac{22.5\% - 7\%}{22.5\%}$$

This RRR is 69%. Hence, a patient with an endoscopically treated bleeding ulcer is 69% less likely to develop recurrent bleeding from the ulcer when receiving an intravenous PPI compared with a similar patient not receiving an intravenous PPI. Absolute risk reduction (ARR) is the reduction in adverse outcomes between the placebo group and the treatment group. Although the RRR compares the risk for adverse outcomes between treated and placebo patients, the ARR identifies the actual reduction in adverse outcomes for treated patients. In the study of patients with endoscopically treated bleeding ulcer treated with intravenous PPI, the ARR may be calculated as follows:

$$\text{\% placebo patients with recurrent ulcer bleeding} - \text{\% IV PPI patients with recurrent ulcer bleeding} = 22.5\% - 7\% = 15.5\%$$

Therefore, a patient with an endoscopically treated bleeding ulcer who receives intravenous PPI may decrease their individual risk for recurrent ulcer bleeding by 15.5%. The RRR can be misleadingly large if adverse outcomes are infrequent in patients receiving placebo or no treatment. This concept is best illustrated by comparing the number needed to treat (NNT) and ARR with the RRR. Consider the results from the VIGOR trial. [18] The results from this trial were reported as frequency of serious NSAID-associated GI complications (severe upper GI bleeding, perforation, or obstruction) per 100 patient-years of NSAID use. There were 0.6 complications per 100 patient-years among patients using rofecoxib and 1.4 complications per 100 patient-years among patients using naproxen. Based on these data, the RRR is as follows:

$$\frac{\text{\% naproxen patients with complications} - \text{\% rofecoxib patients with complications}}{\text{\% naproxen patients with complications}} = \frac{1.4\% - 0.6\%}{1.4\%} = 60\%$$

In other words, a patient who uses rofecoxib is 60% less likely to have a serious NSAID-associated GI complication compared with a similar patient who uses naproxen. This sounds impressive until you consider the ARR. The ARR is as follows:

$$\begin{aligned}&\%\ \text{naproxen patients with complications} -\\&\%\ \text{rofecoxib patients with complications} =\\&1.4\% - 0.6\% = 0.8\%\end{aligned}$$

In other words, an average NSAID-using patient who uses rofecoxib reduces his or her individual risk for a serious NSAID-associated GI complication by only about 0.8%. The NNT allows interpretation of study results in terms of patient care, especially when the RRR is large and the incidence of adverse outcomes is small. Specifically, the NNT is the inverse of the ARR or 1/ARR, estimating the number of patients who need to be treated in order to prevent one additional adverse outcome. In the VIGOR trial, the NNT is as follows:

$$\frac{1}{\text{ARR}} = \frac{1}{0.8\%} = \frac{1}{0.008} = 125$$

In other words, for every 125 average patients treated with rofecoxib instead of naproxen for 1 year, one additional serious GI complication will be prevented. Patients and physicians may be less likely to choose a potentially better, but more expensive, treatment if the study results are presented as the ARR or NNT instead of the RRR. [28] Considering all three statistics (ARR, RRR, NNT) helps patients and physicians assess the potential benefits of therapy. Although the ARR and NNT may appear to be more useful than the RRR, the RRR is valuable because of its versatility. It provides the best estimate of treatment benefit among patients with varying risks of adverse outcomes. [29] NSAID-using patients receiving naproxen in the VIGOR trial only had a 1.4% risk for serious NSAID-associated complications per 100 patient-years of use. However, how beneficial would rofecoxib be in a 78-year-old man with a past history of upper GI bleeding? Based on previously published data, [25] this patient would have a significantly higher baseline risk for serious NSAID-associated GI complications, approaching 10 per 100 patient-years of use (i.e., 10% risk per year). Given this baseline risk, applying the RRR decreases the risk for serious NSAID-associated GI complications from 10% to 4%. This produces an ARR of about 6% (10% - 4% = 6%) and an NNT of about 17 (1/6% = 1/0.06 = 17). For this high-risk patient, the added expense of a coxib is outweighed by the significantly improved safety profile. The value of the RRR is that it can be applied to patient populations with different inherent risks for adverse outcomes. [29]

Evaluating the Sample Size in a Non–Statistically Significant Study: Were Enough Patients Entered into the Study? Studies that do not demonstrate statistical significance may be interpreted as negative studies (i.e., the treatment is no more likely than placebo to reduce adverse outcomes). However, an adequate number of patients have to enter a trial in order to demonstrate a statistically significant RRR, regardless of the effectiveness of the treatment. Many trials that do not yield statistically significant results have not entered enough patients into the trial to demonstrate reliably an RRR of 25% or even 50%. [30] When assessing study results, it should be clear if a non–statistically significant result represents a truly ineffective treatment or an inadequate enrollment of patients into a study. When investigators plan a study, multiple outcomes may be analyzed, but the sample size is calculated based on only one outcome. For example, in a study comparing ligation plus octreotide versus ligation alone in the prevention of recurrent bleeding from esophageal varices, [31] the investigators estimated that ligation plus octreotide would reduce recurrent bleeding from varices by 70% compared with ligation alone. The study entered enough patients to demonstrate a statistically significant RRR of 70% for recurrent bleeding, and a statistically significant RRR of 76% was measured in the study. Investigators also evaluated 30-day mortality and found that combined treatment reduced 30-day mortality by 52% compared with ligation alone, but this RRR was not statistically significant ($p = 0.09$). However, only enough patients to demonstrate a statistically significant RRR of 70% for recurrent bleeding were enrolled in this study. Because the study demonstrated a strong trend for reduced 30-day mortality with combined treatment, this therapy may truly be efficacious in reducing 30-day mortality, and this non–statistically significant result is likely due to an inadequate sample size.

What Is the Precision of the Treatment Effect: How Large Are 95% Confidence Intervals? Traditionally, a p value of 0.05 or less indicates statistical significance. Studies with p values of 0.05 or less indicate that there is a 5% or less likelihood that the difference between treatment and placebo occurred due to chance. However, p values do not provide data about the accuracy or precision of study results. Confidence intervals do provide information about the precision of study results. The 95% confidence interval (95% CI) estimates the range within which the true RRR or ARR resides 95% of the time (i.e., if you repeated the same trial 100 times, then the RRR would fall within the 95% CI in 95 of 100 trials). When the lower limit of the confidence interval for RRR is greater than zero, then the treatment is significantly better than placebo. If the upper limit of the confidence interval around the RRR is less than zero, then the treatment is actually harmful or worse than placebo. For example, the RRR in the VIGOR trial for fewer serious NSAID-associated GI complications with rofecoxib is 60% with 95% CI of 20% to 80%. [18] Thus, the RRR for serious GI complications with rofecoxib has a 95% likelihood of being between 20% and 80% with the best estimate being 60%. The magnitude of confidence intervals is determined by the sample size. [32] Studies with large sample sizes have narrower 95% CIs and a more precise estimate of the true RRR. When the upper limit of a CI around an RRR is greater than zero, but the lower limit of the CI is less than zero, it is possible that the treatment could be better, worse, or no different than placebo. If the magnitude of benefit is moderate and the CI is wide and barely crosses zero, then the treatment probably is beneficial and the trial simply did not enter enough patients. For example, one study [33] examined patients with bleeding esophageal varices and compared band ligation to sclerotherapy for reducing rebleeding. The RRR for recurrent variceal bleeding with ligation was 48%, with 95% CI of -15% to 68%; however, only 77 patients were entered into this study, which was not adequate to demonstrate a statistically significant RRR of 48%. A metaanalysis [34] pooled the results from several studies, allowing the analysis of 547 patients. With this larger sample size, the RRR for recurrent variceal bleeding with ligation was 42% (almost the same RRR as the original randomized trial), but with a statistically significant 95% CI of 16% to 60%.

Case Scenario B—Part III

Given these results, you conclude that rofecoxib, a coxib, is less likely than naproxen, a conventional NSAID, to cause upper GI bleeding, perforations, or obstruction. Because your patient is older than 70 years of age and has a past history of NSAID-associated bleeding peptic ulcer, you estimate that his annual risk for a serious

NSAID-associated GI complication is about 10% per year. Given this baseline risk, you estimate that use of rofecoxib instead of naproxen will reduce the baseline risk by 60% (i.e., the RRR = 60%). Therefore, this patient's annual risk will be reduced from 10% to 4%, producing an ARR = 6% (10% - 4% = 6%) and an NNT = 17 (1/6% = 17), which seems very beneficial. However, you realize that you must balance issues of treatment benefit with issues of safety and cost when deciding about treatment.

Applying the Results of Clinical Research to Your Patient

Are the Results Applicable to Your Patient? If your patient meets the inclusion and exclusion criteria of a study, then the results should be applicable to that patient. Even if your patient is a year too old to be included in the study or has a history of a comorbid disease not allowed in the trial, these issues may not prevent the application of study results to your patient. Ultimately, physicians must use their clinical judgment to decide whether a patient differs significantly from study patients, preventing application of study results to that patient. A study may not identify a statistically significant difference between placebo and treatment for all patients entered in a study, but it may find a significant difference for a subgroup of patients. Often, this subgroup of patients has more severe disease with a higher frequency of adverse outcomes. However, physicians should be cautious before applying subgroup analyses to their patients, even if their patients fit into the subgroups. If investigators evaluate multiple outcomes in many subgroups, eventually one subgroup analysis will be statistically significant simply owing to chance. The results of subgroup analyses are more likely to be valid if (1) the treatment effect is large; (2) only a few subgroup analyses were performed; (3) the subgroup analyses were hypothesized a priori (i.e., before performance of the study); (4) it is consistent with current understanding of pathophysiology; and (5) other, independent studies have produced similar findings. For example, the VIGOR trial demonstrated that rofecoxib decreased clinical upper GI events (i.e., symptomatic ulcers, significant upper GI bleeding, perforation, or obstruction) in the subgroup of patients with a past history of clinical upper GI events. This subgroup analysis was one of only a few subgroup analyses; it was hypothesized a priori. Pathophysiology suggests that selective inhibition of COX-2 isoenzymes reduces the inflammatory response without inhibiting COX-1 isoenzymes in GI mucosal cells and platelets, which should prevent clinical upper GI events.

Are the Potential Treatment Benefits Worth the Potential Side Effects, Costs, and Inconvenience to Your Patient? When deciding whether to start a new treatment, both the patient and physician should consider the inconvenience, side effects, and costs associated with the treatment. When balancing the potential benefit of a treatment versus the potential consequences, the NNT is a helpful tool. If the treatment is cheap and convenient and the consequences of the adverse event are potentially severe, then a large NNT may be acceptable. For example, hundreds of health care providers are vaccinated with hepatitis B vaccination in order to prevent one case of hepatitis B. If the treatment is expensive and inconvenient and has potentially significant side effects, then the treatment may still be acceptable if the NNT is small and the consequences of an adverse outcome are life-threatening.

Case Scenario B—Part IV

The VIGOR trial was a multicenter, international study of rheumatoid arthritis patients. Although your patient has osteoarthritis, it is unlikely that this will produce any difference in the frequency of NSAID-associated GI complications among consistent NSAID users. You are concerned about the cost of rofecoxib (average wholesale price of about $2.50/day) compared with the much lower cost of generic naproxen. Also, you note that patients using rofecoxib had a slight, but statistically significant, increase in myocardial infarctions compared with patients using naproxen (0.4% versus 0.1%, respectively). This change may be partly attributed to antiplatelet effect from naproxen. Although you are concerned about these issues, you believe that the risk for serious NSAID-associated GI complications is so high in your patient that

this outweighs concerns about myocardial infarction.

CONCLUSION

Ultimately, decisions about the use of diagnostic tests and treatments are balanced by possible benefits, harms, costs, patient preferences, and availability of these interventions. In this chapter, principles for the systematic appraisal and application of clinical research have been reviewed. This information will provide a basis to interpret the diagnostic and therapeutic applications of GI technology, which will be discussed in ensuing chapters.

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*The opinions and assertions contained herein are the private views of the author and are not to be construed as official or as reflecting the views of the Department of Veteran Affairs.

CHAPTER 134

Dawn Provenzale

ECONOMIC ANALYSIS IN THE DIAGNOSIS AND TREATMENT OF GASTROINTESTINAL DISEASES

In the face of rising national health care expenditures, gastroenterologists have met increased demand to justify their practices and procedures and to demonstrate the value of their services to both private and public consumers. In order to evaluate economic analyses and determine their value for clinical practice, the reader must have an understanding of certain concepts that provide the basis for economic analysis in the medical literature. In addition, the criteria for performing an economic analysis must be clearly understood so that the reader can critically evaluate these economic analyses and determine which can be applied to their practice. [1]

This chapter is divided into three parts: (1) definitions of the terms costs and charges, (2) a discussion of the different types of economic analyses, and (3) an outline of the criteria for performing an economic analysis and the application of these criteria to screening of patients with gastroesophageal reflux disease (GERD) and surveillance of patients with Barrett esophagus.

COSTS AND CHARGES

In the economic sense, the cost of a service is designated by foregone opportunities. An item used for one purpose cannot be used for another purpose. Thus, the cost of a service can be defined as the resources, or items needed to provide the service, the "opportunity costs." Charges, on the other hand, may be regulated or set by the marketplace and may not reflect the true cost of providing a service, [2] but rather profit or loss to the institution, and may be falsely inflated or deflated. [2]

Most economic analyses include direct medical costs. These include the cost of hospital care, medications, laboratory tests, radiologic procedures, and office visits. Indirect medical costs reflect the cost of loss of life, livelihood, absenteeism from work, or decreased earning ability. Because they are difficult to measure, incorporating indirect costs into economic evaluations remains controversial. Intangible costs reflect the costs of pain, suffering, and other nonfinancial outcomes of disease. They, too are difficult to measure and are not typically included in economic analyses, although quality-of-life measures often attempt to capture these effects.

An example will help distinguish between costs and charges and provide information on how charges (prices) are set in the marketplace. We will use as an example two automobile manufacturers, JM and Kryler, as outlined by Finkler. [2] The companies each produce an identical automobile. The cost to each manufacturer to produce this car is $4200. That is, the resources required to build these cars amounts to $4,200. The government imposes a pollution control program, and each company must include a pollution control device in their cars. JM and Kryler each spend $1 billion to solve the pollution control problem, but the impact of this expenditure is different for each manufacturer. JM sells 10 million cars. The increase in cost to produce the JM car is $100 ($1 billion / 10 million cars = $100). Kryler, on the other hand, sells 1 million cars. The increased cost to produce the Kryler cars amounts to $1000 per car ($1 billion / 1 million cars = $1000). Both companies sell their cars for $5000. JM cars now cost $4,300 to produce, so JM makes a $700 profit on each car. Kryler cars now cost $5,200 to produce, and sold at $5000 per car, the company incurs a $200 loss per car. If Kryler had sold the car for the $5,200 production price, the buyers would have gone to the less expensive but comparable manufacturer, JM. Thus, prices, or charges, may be set through a "consensus" to maintain comparability among different companies.

We can now apply this concept to the field of medicine: "There are two hospitals that perform an identical type of open-heart surgery. One hospital, the high-volume hospital, performs 500 operations per year, whereas the second, low-volume hospital, performs 50 operations per year. The cost to perform the surgery at the high-volume hospital is $4300 per patient, whereas the cost to perform the surgery at the low-volume hospital is $5200 per patient. The surgery is more expensive at the low-volume hospital because there are fewer patients to share a variety of equipment and other costs related to open-heart surgery. Both hospitals charge $4500 for this operation. As in the case of the automobile manufacturers, the large-volume hospital makes a profit of $200 per patient or $10,000 per year ($200 × 500 patients), whereas the low-volume hospital incurs a total loss of $35,000 per year ($700 × 50 patients). Both hospitals charge the same amount to perform the surgery. (This consensus approach to setting hospital rates may reflect competition for health maintenance organization contracts or the hospital's desire not to appear as an outlier.) To offset the loss, the low-volume hospital can increase the charge in some area in which the hospital is particularly efficient or can spread the loss over a very-large-volume service where it will have minimal effect. For example, the hospital may charge an extra 7 cents for a particular laboratory test, which, over the hospital's 500,000 volume of laboratory tests, will have a minimal effect. A charge of $30.07 as compared to $30.00 for a laboratory test will cause much less discussion than $5,200 versus $4,500 for open heart surgery." [2]

TYPES OF ECONOMIC ANALYSES

There are several types of economic analyses to which these basic concepts apply. A cost-identification, or cost-minimization, analysis is the simplest. In this type of analysis, the outcomes of the different treatment alternatives for a particular health problem are assumed to be equivalent. For example, if the analyst wished to examine the economic impact of alternative therapies for achalasia (e.g., pneumatic dilation versus surgery), the outcomes of these alternative management strategies (e.g., symptom relief) would be assumed equal. This type of analysis calculates the costs incurred due to the disease and the medical resources used to treat it, providing the opportunity to determine the cost of alternative management strategies for a certain disorder. In a cost-identification analysis, the costs for each management strategy are summed.

The results are expressed in terms of cost per service provided (e.g., cost per cancer detected), or in the case of achalasia, cost per dysphagia-free day, month, or year. The strategy that is least expensive is the preferred strategy. The goal of the cost-identification analysis is to find the least expensive way of treating the disorder in question.

A cost-effectiveness analysis expands on cost-identification to measure the net cost of providing a service and also to measure the outcomes obtained. Typically, in a cost-effectiveness analysis, the outcomes for the alternative management strategies are not equal. One strategy typically has a better outcome than another (e.g., provides a longer life expectancy or more dysphagia-free days). The cost per specified health effect of a technology or program (e.g., costs per life-year gained or cost per case identified) is calculated to determine the net benefits for the net expenditure of costs for a particular practice or program, and an incremental cost-effectiveness ratio (i.e., the additional costs for the additional benefit obtained) is calculated. The incremental cost-effectiveness ratio is compared with ratios

from other interventions. The lower the ratio, the more cost-effective the intervention.

A cost-utility analysis takes the construct of measuring net costs and net benefits one step further by assigning values to different outcomes in order to reflect the relative importance of the various outcomes. For example, liver transplantation for cirrhotic patients may have a substantial effect on quality of life that will not be identified by using only life expectancy or years of life gained from transplantation as the measure of outcome. A cost-utility analysis considers the net expenditure for the net benefit obtained, as in the cost-effectiveness analysis. The net benefits, however, are adjusted to reflect both the quantity and the quality of the outcome; for example, life years gained are adjusted to reflect quality of life. The results are expressed in a ratio such as cost per quality-adjusted life year (QALY) gained, and incremental cost-utility ratios comparing alternative strategies are calculated. A cost-utility ratio is similar to a cost-effectiveness ratio, except that a cost-utility ratio combines the quality and quantity of the outcomes to produce a more robust and meaningful outcome measure. Expressing results in a cost-utility ratio permits comparison to other health care interventions.

A cost-benefit analysis provides an explicit decision about whether the cost of the practice is worth the benefit obtained from it. This is accomplished by expressing cost and benefit in the same units—typically, in monetary terms. This requires that the health effects of a particular practice or strategy (e.g., life years saved, cases of cancer prevented) also be expressed in monetary terms. In cost-benefit analysis, the practice or program is evaluated by subtracting the cost of that program from the benefit to derive the net benefit. If it is positive, the program passes the cost-benefit test. If two or more *mutually exclusive* programs have positive net benefits, one chooses that program with the greatest net benefit. [1]

CRITERIA FOR PERFORMING AND EVALUATING ECONOMIC ANALYSES

In addition to being familiar with the different types of economic analysis, it is important to understand how they are performed and how the results can be applied to clinical practice. The following steps are useful for determining the validity of an economic analysis [3] (Table 134-1). The criteria will be reviewed and discussed. [1]

1. Was a well-defined question posed in answerable form?
2. Was a comprehensive description of the competing alternatives given?
3. Was there evidence that the program's effectiveness had been established?
4. Were all the important and relevant costs and consequences for each alternative identified?
5. Were costs and consequences measured accurately in appropriate physical units?
6. Were costs and consequences valued credibly?
7. Were costs and consequences adjusted for differential timing?
8. Was an incremental analysis of costs and consequences of alternatives performed?
9. Was a sensitivity analysis performed?
10. Did the presentation and discussion of study results include all issues of concern to users?

From ref. 3.

TABLE 134-1 Criteria for Evaluating an Economic Analysis

1. Was a Well-Defined Question Posed in Answerable Form?

The first step in the assessment of an economic analysis is to decide whether a well-defined question has been posed in an answerable form. [3] Alternative strategies for comparison should be stated and the perspective of the analysis (e.g., that of the patient, a health maintenance organization, or society) defined. As an example, the investigator who wishes to examine the cost-effectiveness of screening individuals with GERD for Barrett esophagus might pose the following question: Taking the perspective of an HMO, is screening for Barrett esophagus in patients with longstanding GERD cost-effective compared with common medical practices such as breast and cervical cancer screening?

2. Was a Comprehensive Description of the Competing Alternatives Given?

In the second step of a critical appraisal, the reader should assess the report for a complete description of the competing alternatives (the alternative practices, strategies, interventions). In the case of screening GERD patients, the analysis should explicitly state the results that will trigger further analysis (e.g., columnar epithelium with goblet cells on biopsy) and what additional analysis will be performed (e.g., endoscopic surveillance). The reader should also seek some definition of the events that will result in evaluation of patients in the no-screening strategy (e.g., worsening of GERD symptoms, dysphagia). Enough information should be provided to the reader to judge the generalizability of the program to his or her own practice and to determine whether the relevant costs or consequences have been included.

3. Was There Evidence that the Program's Effectiveness Had Been Established?

The third step in an assessment of an economic analysis is to determine that the effectiveness of the program under study has been established, that is, that a causative link of the program to the outcome exists. In the case of screening for GERD, the reader should be provided with some evidence that screening reduces mortality from esophageal cancer. We are interested in economic evaluations of cost-effective approaches to effective practices. Therefore, if the effectiveness of the practice under study has not been established, the reader should not waste his or her time on the remainder of the report. With respect to screening for GERD, there is no direct evidence (i.e., results of randomized trials) that this practice actually reduces mortality from esophageal cancer, but indirect evidence (i.e., case series) related to early diagnosis and surveillance of patients with Barrett esophagus suggests that screening individuals with GERD may be beneficial. [4], [5], [6], [7] and [8]

4. Were All the Important and Relevant Costs and Consequences for Each Alternative Identified?

The next step in the evaluation requires that all the important and relevant costs and consequences, or benefits, of the alternative strategies be identified. A comprehensive description of the different types of costs and of the distinctions between them is beyond the scope of this chapter, but some definitions are noteworthy. Most economic analyses consider only the direct medical costs of providing a particular service. This would include the costs of equipment and supplies, facility costs, salaries for physicians and support personnel, and other operating and organizational costs. Direct nonmedical costs incurred by patients and their families, such as any out-of-pocket expenses not covered by insurance, may also be included in some analyses, although this is uncommon because direct nonmedical costs are highly individualized and difficult to measure. Indirect costs created by time lost from work or the psychosocial costs reflected in pain or distress suffered by the patient are typically not included in economic analyses because of the difficulty in measuring them.

The costs of screening for GERD would include costs of endoscopy plus biopsies if taken; salaries of technical staff and physicians, endoscopists, pathologists; and any out-of-pocket expenses to the patient. In addition to cost, a summary of the health consequences (risks and benefits) of the program should be listed. These consequences may include changes in the physical, psychological, or emotional functioning of the patient and may be measured in terms of life years gained, disability prevented, or QALYs gained. Thus, a list of relevant consequences of a screening program for GERD patients would incorporate any increase in life expectancy from cancer prevention and some measure of disability or work loss days prevented by this program. With regard to costs, direct medical cost consequences that should be considered include changes in physician visits and hospitalizations and the ordering of follow-up tests such as endoscopy.

5. Were Costs and Consequences Measured Accurately in Appropriate Physical Units?

In the next component of the critical appraisal, the reader must determine whether the costs and consequences were measured accurately and in the appropriate physical units. In this step, the reader should seek a list of the elements of the analysis described under question 4 and the units for their measurement. For example, in a screening program for GERD, endoscopy might be performed for $600 per test. [9] Other screening tests should be similarly listed, and physician visits and fees should be included. The consequences and the units for their measurement should be explicitly stated. In the screening example, the reader should search for evidence that benefits, such as years of life gained from early diagnosis, disability days avoided, or cancers prevented, are valued and are measured in the appropriate units (e.g., days, years, visits).

6. Were Costs and Consequences Valued Credibly?

In the sixth step of a critical evaluation of an economic analysis, the reader must determine whether the costs and consequences included in the analysis are comprehensive and are plausible. The most appropriate concept of the cost of a service or procedure is the economic opportunity cost, which is the value of the components of the service or the items used to produce the service. Costs may be estimated in a variety of ways, and a clear explanation of the data and the methods used to calculate them should be presented.

The newer information systems that measure resource consumption [10] have promoted the accurate assessment of costs for economic analysis. Hospital accounting departments may also provide information about costs. If estimates of costs (of resources consumed) are not available, charges that have been adjusted to reflect costs may be used as proxy for them. One approach to adjusting charges for acute inpatient care is to calculate cost-to-charge ratios (computable from the cost reports submitted by most hospitals to Medicare). These ratios can be used to convert billed charges to resource cost estimates. Third-party reimbursement levels (e.g., the Medicare Diagnosis Related Group [11] payment) have also been used as a proxy for the opportunity cost. Thus, there are diverse methods for obtaining costs. The reader should seek an explanation of the method that was used to determine costs for each analysis. [1]

An explanation of how benefits are calculated should also be provided. In a cost-effectiveness analysis, for example, health consequences are measured in terms of life years saved. Therefore, assigning a value to benefits is straightforward—each life year gained or lost (at a point in time) counts the same. Indirect costs, such as the cost of time lost from work or school, and benefits, as required for one form of cost-benefit analysis, may be more difficult to ascribe. For example, the indirect cost of lost work time may be difficult to calculate for certain groups such as housewives, elderly people, children, and unemployed people. Because indirect benefits are even more difficult to measure, their incorporation into economic evaluations remains controversial. [1]

In cost-utility analysis, a variant of cost-effectiveness analysis, benefits may be measured in terms of QALYs. The analysis examines both the number of years of life gained from a practice and the quality of those years. In other words, a cost-utility analysis considers that certain practices may be associated with discomfort and disability. Outcomes are adjusted to account for these quality-of-life effects. The studies that incorporate quality-of-life measures, also known as *patient preferences* or *utilities,* attempt to quantify how much better the quality of life is in one health state compared with another (e.g., without cancer versus with cancer). The results are reported in quality-adjusted life units (e.g., days, months, or years) gained from a particular strategy. For a GERD screening program, the quality of life of those in the screened and the unscreened strategies would be measured and compared. The quality-of-life measurements should evaluate the short-term inconvenience or disability associated with screening and the long-term disability associated with potential outcomes, including the diagnosis of Barrett esophagus, the risk for adenocarcinoma, and the inconvenience associated with surveillance endoscopy. The results of a cost-utility analysis are reported as an incremental cost-utility ratio.

7. Were Costs and Consequences Adjusted for Differential Timing?

The costs and health consequences for some interventions occur relatively close to the initiation of the program; for example, vaccination for influenza. For other interventions, particularly those involving screening, the timing of the expenditures and the timing of the health benefits of the program may differ significantly. In a cancer screening program, the costs typically arise early on, whereas the benefits of screening (including increased life expectancy) will occur in the future. To adjust for these differences in the timing of outcomes, it is appropriate to apply standard discounting formulas from economics to calculate the present value of both costs and health consequences, that is, to determine their value as viewed by the decision maker today. [12] Discounting considers that a dollar today is worth more than a dollar in the future, and, because life years are valued relative to dollars in economic analyses, they are also discounted. Most economic analyses use discount rates ranging from 3% to 6%, with 3% as the current recommended standard. [13]

8. Was an Incremental Analysis of Costs and Consequences of Alternatives Performed?

To set priorities for resource allocation, it is necessary to consider the additional costs that one program incurs over another, compared with the additional benefits that are produced. The average costs (the total cost for the procedure or program) and average benefits obtained from the program are easily calculated but do not provide the crucial information needed to determine health policy. It is the additional, or incremental, costs and benefits compared to an alternative practice or program that permit the policy maker to maximize health benefits with a limited health care budget. [1] Costs and health consequences (benefits) should be listed, and a cost-effectiveness ratio, the metric for evaluating tradeoffs across treatment alternatives, should be calculated for each strategy being compared. As an example, Table 134-2 lists both the average and incremental cost-utility ratios for alternative strategies for surveillance of patients with Barrett esophagus. Cost-utility ratios are similar to cost-effectiveness ratios except that the outcomes reflect not only the quantity (e.g., life years gained) but also the quality of those years and consider disability and discomfort associated with the health condition under analysis (QALYs). Costs, here, refer to the direct medical costs of the surveillance tests themselves as well any “induced costs” due to complications of surveillance procedures, the treatment costs for cancers that might be detected through surveillance, and the costs for the evaluation and treatment of cancers detected in those who are not undergoing surveillance but who develop symptoms that lead to evaluation for cancer.

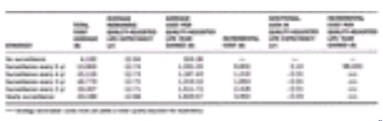

TABLE 134-2 Average Costs and Incremental Costs for Surveillance of Patients with Barrett Esophagus

First, we focus on the average cost-utility ratio. The average cost-utility ratio, or cost per QALY gained, is calculated by dividing the total costs of a particular strategy by the gain in life expectancy for that strategy. For the no-surveillance strategy, which serves as the basis for comparison, the costs amount to $4100 (column 2) and the remaining quality-adjusted life expectancy is 12.64 years (column 3). The average cost-utility ratio is $4100 / 12.64, or $324.36 per QALY gained, as shown in column 4. Moving to the next strategy, surveillance every 5 years, column 2 shows that the cost is $13,900, and column 3 shows that the quality-adjusted life expectancy is 12.74 years. In column 4, the average cost per QALY gained is $13,900 / 12.74, or $1091.05.

Next, we will focus on the calculation of the incremental cost-utility ratio. Column 5 shows that the additional cost for adding surveillance every 5 years compared to no surveillance is $13,900 - 4100 = $9800. The average remaining quality-adjusted life expectancy for the individual who does not undergo surveillance is 12.64 years, whereas the quality-adjusted life expectancy for those undergoing surveillance every 5 years is on average 12.74 years. The years of life gained when surveillance every 5 years is initiated is 12.74 - 12.64 = 0.10 years (37 days) (column 6). The additional 0.10 years is the incremental benefit obtained by performing surveillance every 5 years. The incremental cost per QALY gained, or the incremental cost-utility ratio as shown in column 7, is the incremental cost divided by the additional QALYs gained: $9800 / 0.10 = $98,000, as shown in column 7.

The incremental cost-utility ratios for the more aggressive surveillance strategies (every 1 to 4 years) are not calculated because these strategies are dominated by surveillance every 5 years. In other words, they cost more than surveillance every 5 years and they yield a lower quality-adjusted life expectancy than surveillance every 5 years, as shown in Table 134-2 (columns 2 and 3). It is only with these incremental cost-utility or cost-effectiveness ratios that the policy maker can determine whether the additional benefit is worth the additional cost of adding surveillance compared with a previous policy of no surveillance.

Using these ratios, policy makers who have a fixed budget can allocate funding based on incremental gains and losses. These cost-utility or cost-effectiveness ratios alone, however, cannot identify cost-effective practices. They must be placed in a decision context that is expressed in one of two forms. In the first form, an explicit threshold or maximum amount that the policy maker is willing to spend is stated. For example, the policy maker may be willing to spend $100,000/QALY gained on surveillance of patients with Barrett esophagus. Given our current estimates of cancer risk, surveillance every 5 years would increase quality-adjusted life expectancy and would not exceed the threshold of $100,000/QALY gained. In the second form of decision context, a list of medical practices and their associated cost-utility ratios, also known as a *league table,* is used as a basis for comparison with surveillance of patients with Barrett esophagus. Table 134-3 lists incremental cost-utility ratios for other accepted practices. Surveillance of patients with Barrett esophagus every 5 years has an incremental cost-utility ratio of $98,000/QALY gained, similar to the incremental cost-utility ratio for screening for carotid disease ($130,000/QALY gained) [14] (see Table 134-3). The incremental cost-utility ratio for surveillance

every 5 years of $98,000/QALY gained is lower than the incremental cost-utility ratio for empiric omeprazole therapy in patients with dyspepsia ($780,000/QALY gained), [15] and surveillance of patients with Barrett esophagus would be considered cost-effective compared with this treatment strategy.

	$/QALY GAINED
Prophylaxis for NSAID-associated ulcers with low-dose misoprostol (100 μg 4 times daily) for elderly (>60 yr old) versus no prophylaxis for patients with rheumatoid arthritis taking NSAIDs.	11,000[16]
Dual air bags versus driver's-side only air bags in driving population (and passengers)	69,000[17]
Screening for carotid disease, with carotid endarterectomy if positive versus no screening for carotid disease in 65-year-old men with no symptoms of carotid disease	130,000[14]
Give omeprazole alone empirically versus check serum *Helicobacter pylori* antibody and treat *H pylori* in adults presenting to primary care physicians with diagnosis of dyspepsia	780,000[15]

QALY, quality-adjusted life year.

TABLE 134-3 Incremental Cost-Utility Ratios

Calculating incremental cost-effectiveness or cost-utility ratios for the alternative strategies is essential but does not provide us with a decision rule upon which to base health policy. Critical parameters for decision making include the cost of one strategy compared with another and the resources available to provide surveillance. Those who make health policy must also consider the number of patients affected by Barrett esophagus, the number who might benefit from surveillance, and the number who would benefit from other comparison practices. In addition, the incidence of cancer is paramount in the decision-making process for Barrett esophagus patients. Thus, the policy maker must consider several factors simultaneously when making decisions about funding. These include the budget, or willingness-to-pay threshold, the number of patients who would benefit from surveillance and the benefits to be gained, and the comparison between the cost of surveillance and the cost of other accepted medical practices.

From the patient's perspective, the quality of life associated with surveillance, the short-term discomfort and time lost from daily activities associated with procedures, and the long-term disability associated with esophagectomy are also important factors.

9. Was a Sensitivity Analysis Performed?

As a result of variations in the literature estimates (each of which is affected by sampling and other types of errors) and differences in the opinions of experts in the field, there will be uncertainty surrounding one or more parameters in any economic analysis. To examine the effects of such variation on the results of the analysis, a sensitivity analysis should be performed. In sensitivity analysis, the value of each parameter of interest is varied over a broad range to determine the sensitivity of the results to variations in the parameter. The sensitivity of the preferred strategy to change in the underlying assumptions of the model is a measure of the strength or robustness of the conclusions. If the preferred strategy is altered by sensitivity analysis, then the analysis is said to be sensitive to the value of that parameter. The reader of an economic analysis should seek some evidence that a sensitivity analysis has been performed. In an evaluation of surveillance of patients with Barrett esophagus, a critical sensitivity analysis would be to vary the expected incidence of cancer over a broad range to determine the impact of an increase or decrease in the expected number of cases on the preferred strategy.

10. Did the Presentation and Discussion of Study Results Include All Issues of Concern to Users?

Assumptions and methodologic judgments are an integral part of an economic analysis. The reader should be provided with a list of the assumptions and value judgments that were made to perform the analysis, in order to determine the validity of the study and the applicability of the results to his or her practice. For example, in surveillance of patients with Barrett esophagus, esophageal adenocarcinoma is assumed to occur as a progression from no dysplasia to low-grade dysplasia to high-grade dysplasia and finally to cancer. In ddition, it is assumed that endoscopy is not a perfect test. There may be false-positive or false-negative results for dysplasia or cancer because of sampling error by the endoscopist and misinterpretation by the pathologist. The reader must decide whether these assumptions are credible and the results applicable to his or her practice. In sensitivity analyses, the authors should consider a broad range of parameter assumptions to increase the generalizability of their analysis to populations that differ from their own.

These 10 criteria for economic appraisal provide a framework for reading and evaluating economic analyses in the medical literature. A more detailed discussion of these criteria for economic appraisal can be found in reference 3, which provides a comprehensive review of the principles of health program evaluation.

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CHAPTER 135

Richard A. Rippe, Kevin Behrns, and David A. Brenner

MOLECULAR BIOLOGIC APPROACHES TO THE DIAGNOSIS AND TREATMENT OF GASTROINTESTINAL DISEASES

The development and application of the science and techniques of molecular biology has had a profound impact on the diagnosis and therapy of gastrointestinal disease. Furthermore, molecular biology has revolutionized the way in which these diseases are approached conceptually. Key concepts that are required to understand the regulation of gene expression include deoxyribonucleic acid (DNA) transcription, ribonucleic acid (RNA) translation, posttranslational control, and the effects of DNA mutations on gene expression.

DNA

The genetic material of all known organisms and many viruses is DNA, which consists of chemically linked subunits. Each subunit, called a *nucleotide,* contains a nitrogenous base (a purine [adenosine and guanine] or pyrimidine [cytosine or thymine]), a pentose sugar, and a phosphate group (Fig. 135-1). Nucleotides are linked by phosphodiester bonds forming single-stranded nucleic acids.

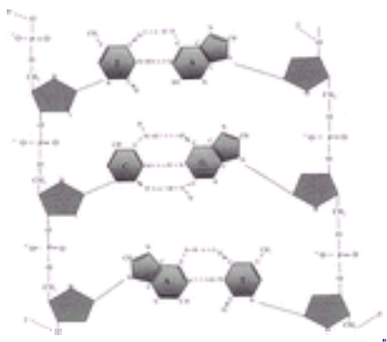

FIGURE 135-1. DNA double helix. A nucleotide consists of the deoxyribose sugar (R) and phosphate backbone with nitrogenous base (A, C, G, or T). Adenonine (A) always pairs with thymidine (T), and guanine (G) always pairs with cytosine (C). The two nucleotide chains run in antiparallel directions, one strand runs in the 5' to 3' direction, and the other strand runs in the 3' to 5' direction. (Reprinted with permission from Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

The DNA double helix consists of two chains that are complementary and antiparallel. They are complementary because adjacent nucleotides always pair in a specific way: adenine to thymine and guanine to cytosine, so that the base sequence of one strand determines that of the other (see Fig. 135-1). They are antiparallel because the 3' to 5' orientation of the sugar-phosphate backbone of one chain is opposite the 5' to 3' orientation of the other chain.

Transcription

Transcription is the synthesis of RNA from its DNA template and is catalyzed by DNA-dependent RNA polymerases. A gene includes all DNA sequences required for the synthesis of the RNA, including transcribed and regulatory sequences. Promoters are the regulatory sequence immediately 5' to the transcription unit and are required for proper transcription initiation. Enhancers are orientation- and position-independent regulatory sequences that increase the transcription rate of a given transcription unit.

Translation

Translation is the synthesis of proteins from the information encoded in its messenger RNA (mRNA). The genetic code consists of groups of three nucleotides called *codons* that specify the amino acid sequence or define sites for the initiation or termination of translation.

TECHNIQUES IN MOLECULAR BIOLOGY

The foundation of recombinant DNA technology involves cutting (restriction digestion) DNA into pieces of interest and then joining them together to generate new recombinant DNA molecules, which can then be mass-produced in bacteria. This process is often referred to as *molecular cloning*. Typically, recombinant constructs consist of vector DNA (DNA molecules that are able to replicate in a given host bacteria) joined to the segment of DNA of interest. Plasmids, the most widely used vector DNA, are extrachromosomal circular DNA molecules that are found primarily in bacteria. They typically contain a gene that confers antibiotic resistance that is used for selection purposes. DNA sequences of interest are inserted into the plasmids and are then grown to high numbers in bacteria, thus generating sufficient quantities for further studies.

Enzymes Used in Molecular Biology Techniques

Most of the methodology used in molecular biology involves the application of specific enzymes. These enzymes can be classified into four groups, including restriction endonucleases, polymerases, ligases, and modifying enzymes. Many current enzymes have been cloned and mutated to enhance performance and reduce cost.

The restriction endonucleases comprise a large group of enzymes that recognize specific nucleotide sequences and cleave double-stranded DNA at precisely these locations. Restriction endonucleases are found naturally in bacteria and are important for degrading foreign DNA. The cleavage sites recognized by the restriction endonucleases are typically four to eight nucleotides in length. Digestion products may have either 5'- or 3'-overhanging nucleotides, also called *cohesive termini* or *sticky ends,* or the digestion may result in blunt-ended molecules. Joining ends of DNA is accomplished with DNA ligases. DNA ligases catalyze the formation of a phosphodiester bond between 5'-phosphate and 3'-hydroxyl ends in duplex DNA. Generating recombinant DNA molecules can be achieved efficiently by digesting double-stranded DNA with appropriate restriction endonucleases, isolating the pieces of interest, and ligating the pieces together to create recombinant molecules (Fig. 135-2).

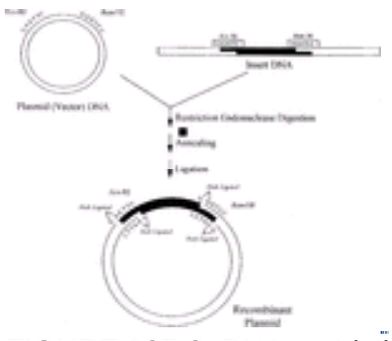

FIGURE 135-2. DNA restriction endonuclease digestion, annealing, and ligation creating a new recombinant molecule. Plasmid DNA and the insert DNA are digested with the same restriction endonucleases (*Eco* RI and *Bam* HI) to create complementary single-stranded nucleotides (“sticky ends”), which anneal together, and the DNA backbone is restored by ligation. (From Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

Another major group of enzymes includes the polymerases, which catalyze the synthesis of nucleic acids from nucleic acid templates. These enzymes are classified by the type of nucleic acid that is synthesized and the type of nucleic acid template replicated. All polymerases synthesize DNA in a 5' to 3' direction and complementary to the template strand. The DNA-dependent DNA polymerases synthesize DNA from DNA templates. In addition to synthesis activity, many of the polymerases possess exonuclease activity, an enzymatic property that removes nucleotides one at a time from the ends of the polynucleotide chain, thereby functioning as a biologic “proofreader.” The exonuclease activity has been used extensively in the laboratory to label double-stranded DNA uniformly.

Klenow fragment is a modified DNA polymerase that also possesses bidirectional exonuclease activities. Because the Klenow fragment possesses both exonuclease activity and polymerase activity, it is useful in DNA cloning protocols. For instance, random priming radiolabeling techniques are used often in Southern and Northern blotting procedures and the creation of these probes utilizes the Klenow fragment. This technique involves first denaturing double-stranded DNA into single strands by heating. Short oligonucleotides then bind in complementary fashion to the denatured DNA, and the fragment is subsequently extended using Klenow enzyme in the presence of deoxynucleotide triphosphates (dNTPs), one of which is radiolabeled (i.e., a ^{32}P-dCTP).

Another group of DNA-dependent DNA polymerases that are used extensively in the polymerase chain reaction (PCR) include the heat-stable DNA polymerases, such as *Taq I*. Originally isolated from *Thermus aquaticus, Taq I* has now been cloned and engineered genetically to improve performance. This enzyme is active over a broad temperature range and is stable with repeated temperature cycling.

The RNA-dependent DNA polymerases synthesize a complementary DNA (cDNA) copy of an RNA template. These enzymes are called *reverse transcriptases* and typically are derived from either retroviruses or the Moloney murine leukemia virus. Reverse transcriptase creates cDNA copies of a population of mRNA molecules. Single-stranded cDNA molecules are converted into double-stranded DNA and cloned into appropriate vectors to generate cDNA libraries. These cDNA libraries are used to isolate genes of interest.

Blotting Techniques

Nucleic acids and proteins are readily detected by fixing these molecules to a nitrocellulose membrane and identifying them by hybridization with specific probes. The detection of specific DNA sequences was described originally by E. M. Southern in 1975 in a technique termed *Southern blotting*.[1] The technique was modified for the detection of RNA sequences and named Northern blotting. Specific proteins are detected by Western blotting.

Southern Blotting Southern blotting involves digesting high-molecular-weight DNA (e.g., genomic DNA) with restriction endonucleases and separating the generated DNA fragments according to size by agarose gel electrophoresis. The DNA fragments are denatured in the gel into single-stranded DNA and subsequently transferred (blotted) from the gel to a solid support, typically a nitrocellulose based membrane. The transferred DNA and a radiolabeled DNA or RNA bind to each other in complementary base-pair fashion to form a hybrid, a process called *hybridization*. The radiolabeled signals are detected by autoradiography, through exposure to x-rays (Fig. 135-3).

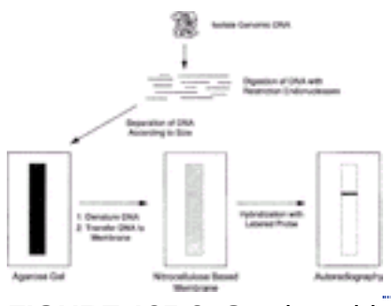

FIGURE 135-3. Southern blotting. Detection of specific DNA sequence in a complex mixture of DNAs (such as genomic DNA) by hybridization with a labeled probe. The hybridization depends on the normal base-pairing of the probe nucleic acid with the complementary genomic DNA sequence. (From Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

Northern Blotting Northern blotting is an adaptation of the Southern blotting method for detection of RNA. RNA is isolated, separated by electrophoresis in formaldehyde-agarose gels, transferred to nitrocellulose-based membranes, and hybridized to specific RNA or DNA probes. Standards of known size allow estimation of the size of RNA of interest. This technique measures the steady-state levels of a particular mRNA under set experimental conditions. Steady-state mRNA levels do not, however, indicate the stage at which gene expression may be regulated. Some conditions might affect the rate at which the gene is transcribed, whereas other conditions may affect the turnover rate, or stability of the mRNA molecule.

Western Blotting Western blotting (immunoblotting) is a method used to detect and quantitate specific proteins using antibodies. The procedure involves extracting proteins from a given source, separating the proteins based on their size by electrophoresis in a polyacrylamide gel and transferring them from the gel to a nitrocellulose-based membrane. Once transferred, the proteins are spotted for binding to a specific antibody. A primary antibody binds to the protein of interest and is subsequently detected using a secondary antibody complexed to an enzymatic detection system. A substrate that undergoes color change when cleaved by the enzymatic detection system allows visualization of the antigen-antibody complexes (Fig. 135-4). The radioimmunoassay and enzyme-linked immunosorbent assay techniques were derived in principle from the Western blot.

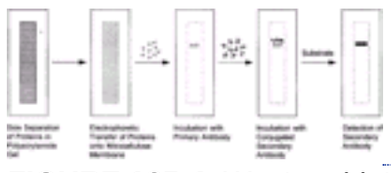

FIGURE 135-4. Western blotting. Detection and quantification of specific proteins using antibodies. Proteins are separated by size on a polyacrylamide gel and transferred electrophoretically to a nitrocellulose membrane. The primary antibody binds to the protein of interest, and the secondary antibody binds to the primary antibody. The secondary antibody contains a detection system that allows identification of the proteins. (From Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

In Situ Hybridization and Immunohistochemical Detection

The study of gene expression often requires the detection of proteins or mRNA within single cells or tissues (in situ analysis). In situ hybridization examines the expression of mRNA, whereas immunohistochemical detection indicates the presence of specific proteins. In situ hybridization provides a sensitive method to locate specific cells that express a gene of interest. The technique involves fixing tissue sections to slides, treating the sections with reagents to increase cellular permeability, and then hybridizing the tissue with either labeled RNA or DNA probes. Probes labeled with enzymes or fluorescent tags (fluorescent in situ hybridization) have excellent resolution, rapid detection, and avoidance of the safety problems associated with radioactive isotopes. In situ hybridization has been used successfully to localize genes on chromosomes, to identify tissues and specific cells in which viruses are replicating, and to locate cells expressing low abundance messages.

Immunohistochemical analysis allows for the localization of proteins to individual cells within tissue samples or to subcellular compartments. Similar to other protein detection methodologies, immunohistochemical analysis uses specific antibodies to bind to the protein of interest, followed by antigen-antibody detection using secondary antibodies directed against the primary antibody.

DNA Sequencing Techniques

Many applications of molecular biology require determination of the DNA sequence, such as a gene or cDNA. Two methods are used to sequence a DNA fragment: chemical sequencing and dideoxy sequencing. Sequencing using chemical degradation was originally described by Maxam and Gilbert. [2]

The second and more popular method of sequencing DNA is called *dideoxy sequencing* or *chain termination sequencing,* as originally developed by Sanger. [3] This method is based on the ability of DNA polymerase to synthesize a complementary strand of DNA from a template (Fig. 135-5). Specific complementary primers are hybridized to the DNA template, and then DNA polymerase is added to allow elongation of the synthesized strand. Four reactions, each containing four dNTPs, one of which is radiolabeled and one of which is a ddNTP, are performed. When the polymerase incorporates the ddNTP in the newly synthesized DNA strand, chain elongation terminates. When one of the four possible ddNTPs (ddATP, ddCTP, ddGTP, ddTTP) is included in each of four reactions, and the reaction products are separated in electrophoretic sequencing gels, a typical sequencing ladder is obtained from which the nucleotide sequence can be read easily. Dideoxy sequencing is now automated. The success of the human genome project depended on high-throughput automated DNA sequencing.

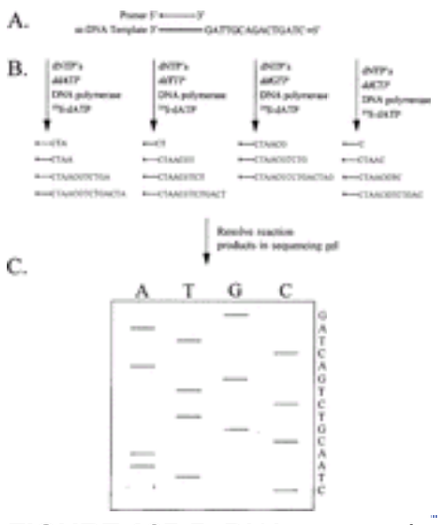

FIGURE 135-5. DNA sequencing by the dideoxy method. Primers specific to the DNA are hybridized to the template (**A**), and then DNA polymerase, dNTPs (dATP, dCTP, dGTP, and dTTP), and a ddNTP are added (**B**). Incorporation of the ddNTP halts chain elongation (**B**). Reaction products are resolved on a sequencing gel (**C**). (From Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

Polymerase Chain Reaction

PCR allows for the rapid amplification of small amounts of nucleic acids. [4] The numerous clinical applications include diagnostic detection of infectious agents, tissue typing, prenatal diagnosis of genetic diseases, and analysis of allelic sequence variations. In the laboratory, PCR methodology is used for DNA sequencing, cloning, gene isolation, and analysis of gene expression.

The basis of PCR is simple but elegant (Fig. 135-6). Two primers are designed that are complementary to the flanking regions of the sequence to be amplified. The primers are added to the template DNA along with a DNA-dependent DNA polymerase that allows new chain synthesis. The reaction mixture is heated to denature (separate the strands) the template DNA, followed by cooling to allow for hybridization of the primers to the template. The DNA polymerase is then allowed to synthesize complementary strands from each primer. Adding the primers in vast excess and repeating the cycles of denaturation, annealing (binding of the primer to the template), and synthesis results in an exponential rate of amplification of the target sequence. The efficiency of PCR has been improved with the discovery of heat-stable DNA polymerases, like *Taq* polymerase, and newer genetically altered enzymes. These heat-stable polymerases retain their enzymatic functions despite thermal cycling. PCR technology has been used extensively for detection of genetic mutations in patients.

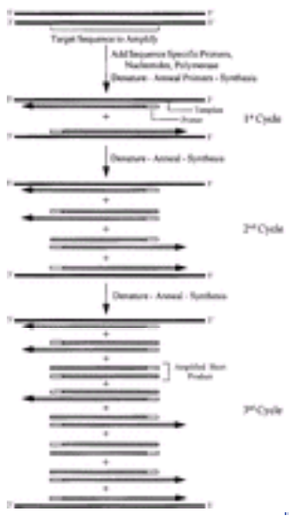

FIGURE 135-6. Amplification of DNA by the polymerase chain reaction (PCR). The template DNA contains the target sequence of interest. Specific DNA primers hybridize to the 5' ends of the target sequence. The reaction also requires a heat-stable DNA-dependent DNA polymerase (such as *Taq I*) and four deoxynucleotides. Repeated cycles of denaturation (usually at 94°C), annealing of primers to template DNA, and DNA synthesis (usually at 72°C) results in the exponential amplification of the target DNA. (From Rippe RA, Behrns K, Brenner DA. Molecular biology for the GI clinician. Gastroenterol Updates 1998;3.)

APPLICATIONS OF MOLECULAR BIOLOGY TO THE CLINICAL LABORATORY

Clinical laboratories are rapidly developing approaches to detect a variety of mutations. The different technical approaches available may be categorized as either targeted or screening mutation detection strategies.

Targeted mutation detection entails straightforward and largely routine procedures by which DNA samples may be analyzed for previously identified mutations using an assay designed for maximum specificity. This approach targets known mutations in potentially large cohorts of clinically indicated patients as well as in disorders marked by one or a few common alleles, and may confirm or establish clinical diagnoses. Furthermore, in families at risk for a particular genetic disease, targeted mutation detection allows for rapid screening of an entire family for the mutation identified in the proband, thereby permitting accurate carrier determinations. Rapid testing of large numbers of patients also permits an assessment of the mutation's frequency among disease-causing alleles, thereby determining which mutations are most prevalent in different patient populations and guiding the creation of effective clinical mutation testing panels.

By contrast, mutation screening analyzes genes for all sequence variants present. By definition, these strategies are not predicated on specificity for specific alleles but rather are designed for highly sensitive detection of all possible variants. In principle, all sequence variants present will be detected without regard to advance knowledge of their pathogenic consequences. Once evidence for a sequence variant is found, the sample must be sequenced to identify it fully. Only when combined with appropriate genetic data and in vitro functional studies can investigators distinguish between disease-causing mutations and polymorphisms without clinical consequence. From a basic research perspective, mutation screening is a critical and obligatory final step toward identifying genes that underlie genetic disease. Screening may also be applied toward the detection of mutations in diseases marked by many different mutations (allelic heterogeneity) and may also aid in the establishment of correlations between genotype and phenotype.

Use of Restriction Endonucleases to Detect Mutations

Mutations represent a fundamental change in the local DNA sequence and may involve single-nucleotide substitutions, small deletions or insertions, or more complex rearrangements. Beyond their potential for clinical consequences, gene mutations may also, by virtue of their changes in nucleotide sequence, create novel recognition sites for restriction endonucleases or destroy preexisting ones. For example, a DNA fragment harboring a mutation might *not* be cleaved with a particular restriction enzyme that normally cleaves wild-type DNA. Conversely, a different mutation might create a novel restriction site not present in the wild-type DNA. In either case, mutated DNA would produce a restriction digestion pattern distinct from that seen with nonmutated DNA when the digestion products are compared after electrophoresis on either agarose or polyacrylamide gels. An example of this commonplace approach applied to mutation detection in cystic fibrosis [5] is shown in Figure 135-7. Other examples include mutation detection in Wilson disease, [6] intrahepatic cholestasis of pregnancy, [7] progressive familial intrahepatic cholestasis, [8] and a $_1$-antichymotrypsin deficiency. [9]

FIGURE 135-7. Restriction endonuclease–mediated detection of the CF causing mutation, W1282X. A restriction recognition site (CCTC/GAGG, *in blue*) present for the endonuclease, *Mnl*1, in the normal *CFTR* exon 20 is ablated by the W1282X mutation (*in blue*). *Arrows* indicate polymerase chain reaction primers. Gel at right shows W1282X-negative (-), W1282X heterozygote (+/-), and W1282X homozygote (+/+) patterns.

Using restriction enzymes to distinguish between alleles is a commonplace technique. This method also has advantages over approaches associated with radioisotope usage and their collateral costs and concerns. The large variety of restriction endonucleases that are commercially available represents a substantial resource to molecular laboratories, and the wide variety of recognition sequences associated with these enzymes affords the investigator many choices for designing straightforward mutation detection strategies.

Despite this large supply of different restriction endonucleases, less than half of known DNA sequence variants independently alter restriction digestion patterns for a commercially available enzyme. Additionally, some enzymes are either unreliable or prohibitively expensive for use in routine, high-throughput analyses.

Single Nucleotide Polymorphisms

Single-nucleotide polymorphisms (SNPs) are the most common variant in the human genome, with a density of at least one common SNP per kilobase pair of DNA. There is now available for research purposes a dense SNP map to study genetic factors important in human diseases, including complex genetic traits. The underlying concept is that genetic markers in close proximity to the responsible mutant genes are in linkage disequilibrium. The technology to perform SNP analysis is rapidly evolving. For example, a microarray chip has been produced that contains more than 1000 SNPs and provides extensive genotype analysis on a patient in one experiment. [10]

Polymerase Chain Reaction–Mediated, Site-Directed Mutagenesis

When confounding factors impede the design of facile restriction-based assays, laboratories may employ a modification of this approach. PCR-mediated, site-directed mutagenesis is a technique by which a novel restriction digestion pattern can be purposefully generated in association with virtually any mutation. This strategy is known by a variety of other names, [11], [12] including *restriction site-generating PCR* [13] and *amplification-created restriction site*. [14] These techniques, identical in approach, involve the design of one PCR primer that abuts the mutation locus and includes typically one or two nucleotides mismatched relative to the template DNA.

If properly designed, this mutagenic primer will bind specifically to its target DNA and support multiple rounds of PCR amplification. All PCR products generated not only will include the mutation locus under study but will also incorporate the base change inherent to the mutagenic primer. The combination of the mutation and the novel base change of this primer creates a novel restriction pattern specific to the different alleles at this locus where none was present previously. This methodology has become commonplace in the literature, including applications for mutations in the *Ras* oncogene, [15] *BRCA1,* [16] ß-thalassemia, [17] medium chain acyl-coenzyme A dehydrogenase deficiency, [18] hereditary hemochromatosis, [19] and a $_1$-antitrypsin deficiency. [20]

Amplification-Refractory Mutation System

In the amplification-refractory mutation system protocol, also known as *allele-specific PCR,* primers are designed that will amplify only one of the alleles present at the locus of interest. [21] This is accomplished with primers that are substantially mismatched relative to one allele, but have sufficient complementarity to anneal to, and amplify, the other allele. Typically, the amplification-refractory mutation system is designed to amplify one allele, whereas a separate reaction will be specific for the other allele at that locus. This technique has also been applied to cystic fibrosis, [22] primary biliary cirrhosis, [23] and the drug-metabolizing liver enzyme, cytochrome P450 CYP2D6 (debrisoquine 4-hydroxylase). [24]

In the absence of any advance knowledge of the likelihood of mutations being present in a DNA fragment, screening strategies are applied. Various technologies are available for the detection of any and all sequence variants that may occur. Many of these approaches will require subsequent DNA sequencing to elucidate the nature of the mutations detected. These approaches, nonetheless, complement targeted mutation detection strategies because all sequence variants will be detected, not just those that have been previously reported and have highly specific assays available.

Single-Stranded Conformational Polymorphism Analysis

Single- and double-stranded DNA fragments may assume distinct secondary structures after they are denatured. These structures are highly dependent on their nucleotide sequences, so that a DNA fragment with a mutation will have an altered structure relative to the normal fragment. The single-stranded conformational polymorphism (SSCP) analysis screening strategy takes advantage of the different rates at which these fragments will migrate through polyacrylamide gels. [25] After amplification of a genomic fragment, the PCR product is typically digested into smaller fragments. It is then partially denatured to single strands and transferred immediately to ice. The sample is then electrophoresed on a nondenaturing polyacrylamide gel. Both double-stranded and single-stranded fragments will be apparent on the gel as distinctly migrating bands. The presence of a mutation or polymorphism within the PCR product will be apparent through band mobility shifts in either the double- or single-stranded fragments or both, when compared with DNA fragments without sequence variants (Fig. 135-8). SSCP is used widely and has been employed for mutation screening in Wilson disease, [26] Alagille syndrome, [27] progressive familial intrahepatic cholestasis, [28] and cystic fibrosis. [29] Other permutations of SSCP, such as conformation sensitive gel electrophoresis, have also been reported. [30]

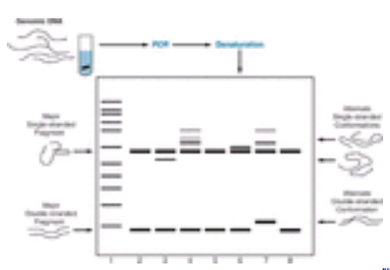

FIGURE 135-8. Single-stranded conformational polymorphism analysis. The polymerase chain reaction product is denatured, placed on ice, and electrophoresed on nondenaturing polyacrylamide gels. Lane 1: size marker. Lanes 2, 5, and 8: no bands of altered mobility are seen in samples with the wild-type sequence. Lanes 3, 4, 6, and 7: bands of altered mobility establish presence of sequence variants. Different variants may yield different electrophoretic patterns. Multiple bands reflect distinct fragment conformations.

Many mutation screening strategies are predicated on heteroduplex formation. Two single-stranded fragments of DNA, such as recently synthesized PCR products, will form fully matched, stable homoduplex structures at appropriate chemical and temperature conditions if they have 100% sequence complementarity. DNA fragments that differ at a small number of nucleotides will also form duplexes, but these fragments will fail to base-pair where the nucleotides are not matched. These heteroduplexes may have subtly altered conformational properties and become substrates for specific chemical as well as enzymatic reactions. When amplifying a DNA fragment from an individual heterozygous for a nucleotide substitution, the final PCR product will include both homoduplex structures for each allele and heteroduplexes derived from the imperfect annealing of dissimilar fragments. Whenever heterozygosity is established, either through gel electrophoresis or through chemical analysis, direct sequence analysis is used to identify the underlying mutation or polymorphism. A heteroduplex detected in a large gene allows DNA sequencing efforts to be targeted toward the fragment that gave rise to the heteroduplex, thus making mutation detection more efficient.

Chemical and Enzymatic Cleavage

Heteroduplex DNA may be cleaved in a sequence-specific manner using either chemical or enzymatic agents. Chemical cleavage of DNA mismatches (CCM) involves agents that modify specific bases when present in the single-stranded form. In imperfectly annealed, heteroduplex fragments, the nucleotide mismatch manifests as a very localized single-stranded region with bases vulnerable to chemical modification. Hydroxylamine modifies unpaired cytosines, whereas osmium tetroxide modifies unpaired thymines. Piperidine is then added to cleave the mismatched products, which are then electrophoresed to resolve the digested fragments. [31] Thus, every possible single-base mismatch, as well as small insertions and deletions, is subject to chemical cleavage. Phenylketonuria, [32] acute intermittent porphyria, [33] and hemophilia A [34] are among the disorders for which CCM has been used. Not only can the presence of a mutation be established, but its approximate location within the fragment also becomes apparent from the size of the digestion products.

An enzymatic protocol for mismatch-directed cleavage using T4 endonuclease VII avoids the chemical toxicity of the CCM reagents. [35], [36] In vivo, T4 endonuclease VII resolves the complicated structures of genetic recombination by recognizing kinks and loops produced in recombining DNA. These transient structures are similar in nature to the mismatch-bearing fragments that typify heteroduplexes. Enzyme mismatch cleavage thus cleaves heteroduplex fragments, allowing the digestion products to be characterized by electrophoresis (Fig. 135-9). Mutation detection using enzyme mismatched cleavage has been described for hereditary nonpolyposis colorectal cancer [37] and for hereditary pancreatitis. [38] As with CCM, some positional information is also generated.

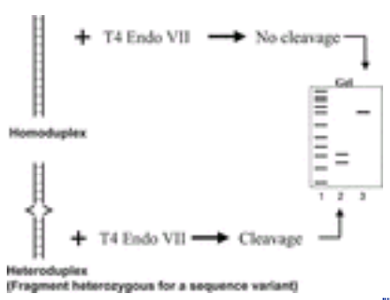

FIGURE 135-9. T4 endonuclease VII–mediated cleavage. Homoduplex polymerase chain reaction (PCR) fragments have no localized single-stranded domains and will not be cleaved by Endo VII and produce a single uncut band after electrophoresis (lane 3). Heteroduplex PCR fragments will be cleaved by Endo VII, giving cleaved fragments (lane 2). Lane 1: size marker.

Heteroduplex Analysis

The conformational changes associated with heteroduplex structures often lead to fragments with altered electrophoretic mobility. Heteroduplex fragments may be separable from homoduplex forms, thereby establishing heterozygosity for a sequence variant in that sample. A method that exploits the differential migration of homoduplexes from heteroduplexes uses mutation detection enhancement (MDE) gels (BioWhittaker Molecular Applications, Rockland, ME). This proprietary gel matrix is formulated to enhance the electrophoretic separation between homoduplex and heteroduplex fragments in a straightforward manner. [39] After PCR, DNA products are renatured slowly to enhance heteroduplex formation and are subsequently electrophoresed on MDE gels. Heteroduplex and homoduplex fragments migrate on a gel with different mobilities, an effect that is directly related to the subtle structural differences between them. This technique has been used for mutation detection in Wilson disease, [40] acute intermittent porphyria, [41] Duchenne muscular dystrophy, [42] and hereditary breast cancer (*BRCA1*). [43]

Denaturing Gradient Gel Electrophoresis

The differential conformational properties of heteroduplex fragments may be exploited by other methodologies. As a result of their mismatched bases, heteroduplexes denature into single strands more readily than their homoduplex counterparts. Denaturing gradient gel electrophoresis (DGGE) uses a chemical gradient within the electrophoretic gel to take advantage of this difference. [44], [45] The chemicals involved, typically urea or formamide, denature PCR fragments from double- to single-stranded forms. PCR of heterozygous samples yields heteroduplex fragments with altered denaturation properties. Once denaturation of a fragment occurs, its migration through the gel effectively ceases. The appearance of these slowly migrating bands on DGGE gels is indicative of heterozygosity for a DNA sequence variant in the underlying sample. This “melting process” is sensitive not only to the nature of the mismatch and the underlying mutation but also to the sequence of the fragment as a whole. DGGE is another widely used technique, having found utility for detecting mutations in cystic fibrosis, [46] erythropoietic protoporphyria, [47] *p53* mutations in hepatocellular carcinoma, [48] tuberous sclerosis, [49] and hereditary breast cancer. [50]

Protein Truncation Test

Cell-free expression systems can be used as a bridge between genomics and proteomics by converting nucleic acid sequence (usually in the form of RNA) into protein sequence. One in vitro expression system is referred to as the *protein truncation test* and can be used to perform mutation analysis, particularly for those mutations that alter the reading frame of the expressed protein, so-called truncating mutations, which lead to a shortened protein product. This method is particularly useful in detecting mutations in large genes that are frequently altered by nonsense and frameshift mutations (the result of small insertions or deletions) in addition to large deletions. Examples of the underlying disorders include familial adenomatous polyposis, [51] cystic fibrosis, [52] Duchenne and Becker muscular dystrophy, [53] neurofibromatosis type 1, [54] and hereditary breast and ovarian cancer defined by tumor suppressor genes such as *BRCA1* and *BRCA2*. [55], [56]

The current method involves reverse transcription PCR, in which total RNA is used to produce single-stranded cDNA. PCR amplification, using a forward primer that includes the gene’s regulatory sequences necessary for transcription and translation (Fig. 135-10), results in a functional protein coding segment. Protein synthesis is completed by adding this segment to an in vitro translation and transcription system that includes radiolabeled amino acids and results in a labeled protein. A novel, lower-molecular-weight band indicates the presence of a truncated polypeptide, representing a truncating mutation in that sample. The position of the band indicates the relative size of the product, from which the position of the genomic alteration can be extrapolated (see Fig. 135-10). Confirmation of the mutation is achieved by genomic sequencing.

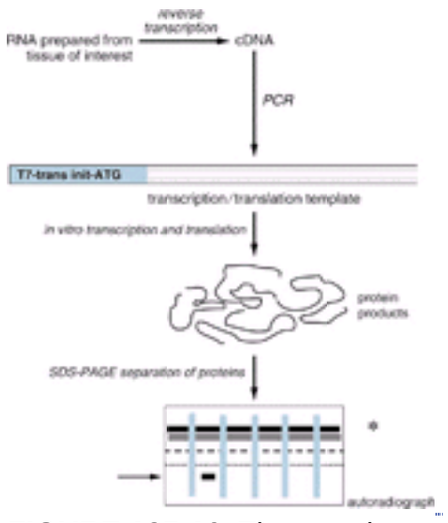

FIGURE 135-10. The protein truncation test. RNA is usually isolated from peripheral blood lymphocytes from whole blood collected in the presence of ACD or EDTA anticoagulants. (The quality of RNA isolated from tissue is dependent on rapid freezing of tissue to -70°C or below.) Coupled in vitro transcription/translation kits, using a rabbit reticulocyte extract, are commercially available. A labeled amino acid, usually ^{35}S-lysine, is added to the in vitro assay to label the synthesized proteins for autoradiography. The *arrow* adjacent to the autoradiograph indicates the presence of a band of lower-than-expected molecular weight, representing a truncated protein. The *asterisk* identifies the full-length, nontruncated protein.

Missense mutations do not usually result in size alterations of proteins; therefore, other procedures must be used for missense detection, so that maximum analytic sensitivity is often combined with scanning techniques for nontruncating mutations. Using this approach, the sensitivity for *BRCA1* and *BRCA2* mutation detection can approach 90%.

Real-Time Polymerase Chain Reaction

Allele-specific mutation detection is also amenable to automation. The Perkin-Elmer 7700 TaqMan allelic discrimination protocol enables high-throughput genotyping using real-time PCR. This strategy entails the use of a short DNA probe with a fluorescent dye attached to one end and a quenching agent linked to the other end. This probe is designed to match perfectly one allele at a particular locus but to be mismatched relative to other alleles. As the synthesis of the PCR product advances, the DNA polymerase 5'-exonuclease activity digests any probe in its path, releasing the fluorescent dye from the quencher and generating a fluorescent signal. If the probe is mismatched relative to the template, its affinity for the template will be greatly reduced under appropriately stringent conditions. Probe not bound to the template will neither be digested nor generate signal. A similar probe may be designed to detect the other alleles, and these assays may be pooled as long as each probe carries a different, separately detectable, fluorescent dye. Real-time PCR approaches to mutation detection have been reported for a $_1$-antitrypsin deficiency, [57] hemochromatosis, [58] Tay-Sachs disease, [59] and the study of mephenytoin metabolism by CYP2C19 (mephenytoin 4'-hydroxylase) variants. [60]

Microarrays

Functional genomics is the study of all genes expressed by a specific tissue and the changes in their expression pattern during a stimulus such as a disease state. Microarrays bring the analysis of gene expression to a genomic scale by permitting the simultaneous measurement of changes in expression of thousands of genes in a single study. Previously, analysis of gene expression was conducted one gene at a time, such as with the Northern blotting technique discussed earlier. Microarrays (also known as *DNA hybridization arrays* and *high-density oligonucleotide arrays*) measure the relative levels of mRNA for thousands of genes. The microarrays have four components: probes, targets, hybridization, and detection. Using the genetic databases available from the Human Genome Project, specific probes are constructed for each human gene to be placed on the array. These probes may be cDNAs that are amplified from plasmids or oligonucleotides synthesized onto the array matrix. The target consists of a labeled cDNA from the tissue or cell of interest. In most cases, an experimental condition (such as a diseased tissue) is compared with a control condition (such as a normal tissue). The cDNAs are labeled either with radioactivity or a fluorescent label. If different fluorescent labels are used for the experimental and control RNA, then both targets may be incubated on the same microarray. The labeled targets are then hybridized to the probes, which

enable the complementary target and probe sequences to bind specifically. The array is then washed to remove nonhybridized probes, reduce nonspecific hybridization, and minimize background. The specific hybridization is then detected by quantitating the amount of label that has annealed to the probe (i.e., either radioisotopic or fluorescence detection) (Fig. 135-11). Massive amounts of data are generated by the detection of hybridization on these arrays. The data must be normalized and then analyzed. New modes of analysis are being developed to analyze effectively this vast volume of information.

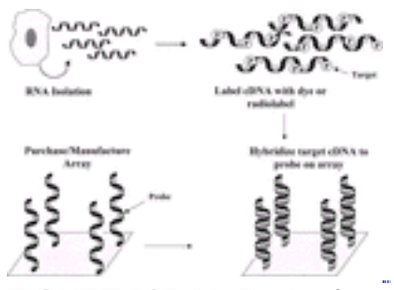

FIGURE 135-11. DNA microarrays for studying gene expression. RNA from a tissue of interest is converted into a labeled cDNA probe and hybridized to thousands of genes. The relative expression of genes representing the entire human genome can be detected on a single chip.

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CHAPTER 136

Gregory Zuccaro, Jr.

REPROCESSING OF GASTROINTESTINAL ENDOSCOPES AND ACCESSORIES

The risks associated with gastrointestinal endoscopy are generally discussed in terms of those directly related to the procedure (e.g., oversedation, bleeding after polypectomy, perforation of the gastrointestinal wall). However, another theoretical risk of endoscopy involves the introduction of pathogens from the endoscope or accessories into patients undergoing a procedure. Because the endoscope and accessories often come into contact with potentially infectious fluids (e.g., blood) from one patient, and then are reused in subsequent patients, pathogens could be passed from patient to patient if not completely removed after each use. Nonpathogenic organisms could theoretically be transmitted to immunocompromised hosts by the same mechanism. Another potential mechanism whereby infection could be introduced through endoscopy is colonization of the endoscope or accessories by organisms in the environment after proper reprocessing, with introduction of these organisms into a subsequent patient. Although apparently rare, clinically significant infections have been documented in the literature by each of these scenarios. In most cases, a breakdown in accepted practices of endoscope and accessory reprocessing can be identified.

Endoscope and accessory reprocessing involves a series of steps that result in the generation of a patient-ready system: a system free of any organisms that might pose a risk for infection to the subsequent patient undergoing endoscopy. In keeping with the concept of universal precautions, each patient is assumed to be potentially infected with a pathogen.

TRANSMISSION OF INFECTION THROUGH ENDOSCOPY

The risk for transmission of infection from patient to patient through endoscopes and accessories appears to be quite small. An American Society for Gastrointestinal Endoscopy (ASGE) review identified 28 cases of transmission of infection through endoscopy since the implementation of standard guidelines for endoscope reprocessing. In each case, the reviewers felt that some breach in endoscope reprocessing techniques occurred (although this may not be true because some cases apparently occurred in the face of accepted reprocessing techniques). They estimated that the risk for infection from endoscopy was about 1 in 1.8 million. [1] This estimate may be understated, in view of the many reports of infection cited in the following paragraphs.

Hepatitis Virus

Transmission of hepatitis C virus from a carrier undergoing colonoscopy to two subsequent patients has been documented. [2] Hepatitis C virus genotyping and nucleotide sequencing established that all three patients were infected with the same isolate. Accepted reprocessing techniques were not performed between each procedure. Transmission of hepatitis B from patient to patient has been reported. [3] Acute hepatitis B infection developed 96 days after endoscopy and therapy for a bleeding ulcer. The endoscope was used immediately before to treat bleeding esophageal varices in a patient with hepatitis B. In this case, however, the endoscope and accessories had been reprocessed carefully following accepted protocols. Serotyping of the hepatitis B virus showed the same subtype in both patients. The patient with the bleeding ulcer did receive blood transfusions; nevertheless, the investigators (and subsequent reviewers [4], [5] and [6]) feel that this case most likely represents transmission of hepatitis B. Several clinical series indicate that the likelihood of hepatitis B virus transmission from patient to patient is extremely low. [7], [8], [9], [10], [11], [12], [13], [14], [15], [16] and [17] In many of these series, the endoscope and accessories were not reprocessed following currently accepted protocols, yet no infection in subsequent patients was evident. Transmission of hepatitis virus from patient to patient through endoscopy, although possible, is exceedingly rare.

Human Immunodeficiency Virus

Transmission of human immunodeficiency virus (HIV) as a result of gastrointestinal endoscopy or bronchoscopy has not been reported, but the virus can be recovered from these instruments after procedures in infected patients. Sampling of the suction and the air and water channels immediately after use in patients with HIV infection revealed HIV-specific DNA by polymerase chain reaction in 7 of 20 endoscopes. [18] In a similar study, all 7 bronchoscopes tested were positive for HIV-specific DNA. [19] HIV was readily eradicated from all contaminated endoscopes and bronchoscopes using standard reprocessing techniques.

Salmonella species

Numerous reports implicate endoscopy in transmission of *Salmonella* infections. [20], [21], [22], [23], [24], [25], [26] and [27] A group of patients developed *Salmonella newport* infection after undergoing colonoscopy subsequent to this procedure being done in a patient with acute *S newport* infection. [20] *S newport* identical to that recovered from the index case was identified in 8 of 48 patients who had procedures before the transmission was recognized. The apparent source of infection was not a colonoscope but the spring from a pair of biopsy forceps. In all of these series, the technique of endoscope reprocessing was less rigorous than currently recommended.

Pseudomonas aeruginosa

Pseudomonas aeruginosa is a ubiquitous organism. It can be found in tap water and can quickly colonize any damp area, including the channel of a reprocessed endoscope, water bottle, or endoscopic automated reprocessor. *Pseudomonas* species have even been identified as contaminants within hospital disinfectants, including chlorhexidine. [28] Therefore, although the prior discussion involved transmission of organisms from patient to patient through a contaminated endoscope, pseudomonal infections involve the introduction of this organism from the environment into a patient.

In one clinical series, 7 patients had *P aeruginosa* bacteremia after endoscopic retrograde cholangiopancreatography (ERCP) in a 1-year period, with 1 death related

to the infection. [29] In each case, the infected patient was the first case of the day in which that duodenoscope was used. In 6 of the 7 cases, the endoscope had been reprocessed more than 48 hours before the endoscopy. Cultures from these endoscopes after prolonged storage were positive for *P aeruginosa, Escherichia coli,* and *Serratia marcescens.* Similar culture results were obtained from other devices in the endoscopy suite, including wash basins, water irrigation bottles, and irrigation syringes. Infection control measures, including alcohol flushing of endoscope channels after reprocessing before compressed air drying to eliminate moisture that would serve as the medium for bacterial growth, resulted in elimination of further cases. The endoscope has been identified as the source of infection in several other series as well. [30], [31], [32], [33], [34], [35], [36], [37], [38], [39] and [40] Patients may become infected even when the pancreatobiliary tree is normal at the time of ERCP. [35]

There may be sources of *P aeruginosa* infection other than the operating channel of an endoscope. Struelens and colleagues [41] noted an increase in post-ERCP bacteremia after the implementation of automated reprocessors in the endoscopy suite. *Citrobacter* and *Pseudomonas* species were among the organisms identified in patients and cultured from duodenoscopes, and the reprocessor also showed evidence of bacterial contamination. Additional steps, including alcohol and air flush of the duodenoscope channels after reprocessing, and disinfection of the automated reprocessor, remedied the situation. Although the literature on transmission of *P aeruginosa* involves primarily duodenoscopes, other endoscopes may be similarly colonized. Alvarado and associates [42] noted a significant increase in the colonization rate of upper gastrointestinal endoscopes with *P aeruginosa* after reprocessing in a recently purchased endoscopic automated reprocessor. When endoscopes that had been manually reprocessed were tested, none were found to be contaminated with *P aeruginosa.* Certain areas of the endoscopic automated reprocessors, including the water inlet hose and air vents, were not accessible to the disinfectant solution and served as a reservoir for the organism. This design flaw in the original endoscope automated reprocessor has been corrected. The problem was corrected in the endoscopy suite by implementation of additional steps to the disinfection process, including rinsing of endoscope surfaces and channels with 70% isopropyl alcohol after machine disinfection, then drying with forced air for 10 to 20 minutes. [43]

Serratia marcescens

Serratia marcescens has been reported as an infectious complication of ERCP, sclerotherapy, and bronchoscopy. Two consecutive patients developed complications of ERCP related to the organism: acute hemorrhagic pancreatitis (patient died) and acute cholecystitis. [44] Cultures from the duodenoscope biopsy channel and intraoperative cultures for both patients grew *S marcescens*. A case of *S marcescens* mediastinitis and empyema after variceal sclerotherapy was reported, although the organism was not recovered from the endoscope or endoscopy suite. [45] Transmission was also reported through bronchoscopy to six patients after the instrument had been used on a patient with *S marcescens* pulmonary infection. [46] The organism was recovered from the bronchoscope on subsequent testing. Before this episode, the disinfection process involved only mechanical cleaning, suctioning water and alcohol through the operating channel, wiping the scope with alcohol, and drying the operating channel by aspirating ambient air. After the consecutive infections were identified, povidone-iodine (Betadine) was added to the operating channel cleaning regimen, and gas sterilization was performed. These additional steps eliminated the organism.

Helicobacter pylori

There are multiple clinical series documenting the patient-to-patient transmission of *Helicobacter pylori*. In 1979, a group of volunteers undergoing studies of gastric acid secretion developed rapid hypochlorhydria. [47] Endoscopic biopsy revealed histological gastritis. Although no organism was recovered, the investigators suspected an infectious etiology. A similar report noted gastritis and hypochlorhydria in 4 of 6 volunteers undergoing gastric acid analysis for a clinical study. [48] Contaminated pH probes rather than endoscopes were the source of infection in these series, emphasizing that proper disinfection must be performed for all gastrointestinal devices (e.g., manometry catheters, pH probes). Gastritis with *H pylori* developed in an individual with normal baseline histology who underwent serial gastric analysis and endoscopy as part of a research protocol. [49] Presumably, the endoscopic procedure was the source of the infection, although no information was offered as to whether the source was the endoscope, the biopsy forceps, or other unidentified agent.

A retrospective review of patient-to-patient endoscopic transmission of *H pylori* with restriction enzyme analysis of bacterial DNA suggested the rate of transmission due to endoscopy and biopsy was 1.1% over 3 years. [50] The method of endoscope disinfection involved mechanical cleaning, a 30-minute wash with 2% glutaraldehyde, sterile water rinse, and hot air dry. Biopsy forceps were not always sterilized between uses but sometimes “cleaned and disinfected.” All of the patients infected with *H pylori* in their series underwent mucosal biopsy, raising the possibility that the forceps was the source of infection. In a study of nine patients with recurrent infection after *H pylori* eradication, two had recurrent infection introduced through endoscopy. [51] The investigators reported that “improved” reprocessing techniques eliminated further transmission. The source of transmission (endoscopy, forceps) was not specified.

Mycobacterium species

The contamination of a flexible endoscope with *Mycobacterium* organisms has been regarded primarily as a problem for bronchoscopists and not gastrointestinal endoscopists. However, with the possibility of transmission of atypical mycobacteria to immunocompromised hosts, a review of this literature is relevant to gastrointestinal endoscopists.

Early reports describe patient-to-patient transmission of *Mycobacterium tuberculosis* through bronchoscopy when disinfection was performed using an iodophor, a chemical disinfectant currently felt to be inadequate for endoscope reprocessing. [52], [53] However, use of a bronchoscope colonized with *M tuberculosis* does not necessarily lead to cross-infection. In one series, 19 patients had bronchoscopy using a bronchoscope colonized with *M tuberculosis*. None of the patients developed symptoms or signs of tuberculosis or positive skin tests.

Continual colonization of bronchoscopes with *Mycobacterium chelonae* led to clinically significant respiratory infection in two immunocompromised patients. [54] Glutaraldehyde disinfection of the bronchoscopes did not remedy the problem, nor did ethylene oxide sterilization. When the bronchoscopes were sent for structural analysis, punctures of the operating channels were found, with gross contamination of proteinaceous material within. The bronchoscopes still were functional despite these internal defects. Clinical experience such as this is the basis for current recommendations regarding leak testing of flexible endoscopes with each use.

M tuberculosis and *Mycobacterium avium* were transmitted through contaminated bronchoscopes, despite use of a rigorous reprocessing protocol involving mechanical cleaning and 2% glutaraldehyde. [55] The endoscope channels were not the source of infection, but persistent mycobacterial infection was present on the suction valve. This infection could not be eradicated with glutaraldehyde, but was eradicated with steam autoclaving. Persistent colonization of endoscopes and bronchoscopes with *Mycobacterium chelonae* has also been linked to persistent colonization within an automated reprocessor. [56] Attempts to disinfect the reprocessor were not successful because of the formation of biofilm within a portion of the reprocessor. Addition of 70% alcohol rinsing to the endoscopes after reprocessing eliminated the problem. A similar case of *Mycobacterium abscessus* transmission was traced to persistent colonization of an automated reprocessor. [57]

Other Organisms

Other microorganisms that have been reported to be transmitted through endoscopy or bronchoscopy include *Listeria, Strongyloides, Rhodotorula, Trichosporon, Enterobacter,* and *Citrobacter* species. [5], [38], [58], [59], [60] and [61]

STERILIZATION AND LEVELS OF DISINFECTION

The Association for Professionals in Infection Control and Epidemiology (APIC) has put forth definitions helpful in a discussion of endoscope reprocessing [62] (Table 136-1). Varying levels of disinfection and sterilization may be achieved when reprocessing an endoscope and accessories. In fact, the same agent may achieve sterilization or high-level disinfection of an endoscope, depending on factors such as the amount of exposure time and the degree to which debris and organic material were removed before exposure to the agent. It is reasonable, therefore, to ask whether an endoscope and accessories should be sterilized between each use, or if a lower standard, such as high-level disinfection, is acceptable.

TERM	DEFINITION
Sterilization	Complete elimination of all forms of microbial life
Disinfection	Elimination of most or all pathogenic organisms
High level	Elimination of all microorganisms except high levels of bacterial spores
Intermediate level	Elimination of most microorganisms; some viruses, fungi, and bacterial spores may not be eliminated
Low level	Elimination of most bacteria, but not more resistant organisms such as mycobacteria and spores
Cleaning	Removal of foreign material (e.g., blood, tissue) from the endoscope and accessories; includes wiping, brushing channels, enzymatic detergents, ultrasonic cleaners

TABLE 136-1 Definitions in Endoscope Reprocessing

In 1968, Spaulding [63] proposed a classification scheme that has been accepted and retained (with minor modifications) by experts in infection control, including the APIC. A device that enters sterile tissue or the vascular system is called a *critical device* and must be sterilized before use. *Noncritical devices* involve contact with intact skin, and intermediate or low-level disinfection is acceptable. A device that comes into contact with mucous membranes, but not sterile tissue or the vascular system, is called a *semicritical device,* and high-level disinfection is deemed sufficient for these items. However, this produces somewhat of a double standard for endoscopy. In many cases, endoscopes simply contact mucosal surfaces, fitting the Spaulding classification scheme nicely. However, a biopsy forceps, sclerotherapy needle, or papillotome may come in contact with the vascular system. Because these devices are passed through the operating channel of the endoscope, should the endoscope be considered a critical device? Although there is no simple answer to this question, most experts in infection control, including the APIC, have recommended high-level disinfection for the endoscope, but sterilization of accessories that come into direct contact with the vascular system. [6]

Problems Specific to the Endoscope and the Endoscopy Unit

The endoscope is a complex structure, with multiple channels and interior surfaces in its internal composition. Areas that cannot be accessed with a cleaning device or disinfectant or sterilant cannot be reprocessed reliably. The operating channel of the endoscope is accessible to a cleaning brush, and manual cleaning of this channel is essential in endoscope reprocessing. The operating channel may become damaged, forming a nidus of persistent colonization. This damage may not be recognizable to the operator because the endoscope may perform normally until the damage is advanced and fluid invasion of the system causes overt malfunction. The air-water channel of the endoscope is smaller and, in many endoscopes, cannot be brushed manually; forcing air through the channel after the procedure, and liquid or gas disinfectants or sterilants that can insinuate themselves into these small areas are used to disinfect this area of the endoscope. The newer, totally immersible endoscopes lend themselves to this aspect of reprocessing. Some newer endoscopes do allow for mechanical cleaning of the air-water channel.

Another aspect of disinfection is the time allotted for the process. Most endoscopy suites have a limited number of endoscopes available and therefore require reasonable turnaround times. A highly effective technique for sterilizing or providing high-level disinfection that requires several hours is not practical and cannot be implemented in most endoscopy suites.

Organisms and Their Resistance to Chemical Disinfectants and Sterilants

Organisms are not equal in their resistance to destruction by chemical disinfectants or sterilants. Among the most susceptible are viruses such as hepatitis B and HIV. These viruses are readily inactivated (i.e., rendered unable to cause infection if injected into a susceptible host) by a number of chemical disinfectants and sterilants. [62] Among the agents effective in hepatitis B inactivation are glutaraldehyde at 1% and 0.1% concentrations (5-minute exposure), 80% ethyl alcohol (2-minute exposure), and 98°C heat for 2 minutes. [64] Hanson and colleagues [65] reported that 2% glutaraldehyde was effective in inactivating HIV in serum within 2 minutes, but 1% glutaraldehyde was ineffective after 15 minutes of exposure, and 70% ethanol did not inactivate the virus after a 10-minute exposure. Resnick and colleagues [66] reported that HIV was rendered noninfective with exposure to solutions of 0.5% sodium hypochlorite (1 minute), 70% alcohol (1 minute), and 0.08% quaternary ammonium chloride (10 minutes). Van Bueren and associates [67] also found inactivation of cell-free and cell-associated HIV with 10-minute exposure to 70% alcohol.

In general, the bacteria that may lead to cholangitis or other infections from endoscopy, including *P aeruginosa, S marcescens,* and *Salmonella* species, are also destroyed by common disinfectants and sterilants. [62] There is a recent report of isolation of a strain of *P aeruginosa* resistant to disinfection by glutaraldehyde. [68] Small viruses, including polio virus and hepatitis A, may be more difficult to inactivate. Chemical disinfectants, including 70% ethanol, 3% and 6% hydrogen peroxide, and dilute aldehydes, are only variably effective against poliovirus. [69] Solutions of 2% glutaraldehyde and high-concentration hypochlorite are necessary to inactivate the virus reliably. Only 0.5% sodium hypochlorite, 0.3% organochlorine, 2% glutaraldehyde, and modified quaternary ammonium compounds effectively inactivated coxsackievirus, adenovirus, parainfluenza virus, and coronavirus. [70] Only 2% glutaraldehyde, quaternary ammonium compounds with high concentrations of HCl, and sodium hypochlorite were effective in hepatitis A inactivation. [71] Among the less efficacious compounds were iodines, alcohols, acetic acids and peracetic acids, and hydrogen peroxide. Peracetic acid and 2% glutaraldehyde are among the chemical sterilants able to inactivate human rotovirus. [72] Among the less efficacious agents were 70% ethyl alcohol and sodium hypochlorite. Protozoan species are also relatively more resistant to destruction. Campbell and colleagues [73] found that *Cryptosporidium* oocysts were resistant to many common disinfectants but were inactivated by some agents, including formol saline and ammonia compounds. The commonly used agents in reprocessing endoscopes were not tested in this series.

Among the most resistant microorganisms are mycobacteria and bacterial spores. Several agents, including quaternary ammonium compounds, dilute sodium hypochlorite, 70% ethanol, povidone iodine, iodophors, and chlorhexidine, are ineffective against *M tuberculosis;* more concentrated sodium hypochlorite and 2% glutaraldehyde are effective. [74] Contact times are important; 2% glutaraldehyde did not destroy the organism in 1 minute but did in 10 to 30 minutes. A study with spores of *Bacillus subtilis* and *Clostridium sporogenes* found that 6% hydrogen peroxide solution was extremely effective against the spores, whereas 2% glutaraldehyde solutions reduced the number of viable spores but did not completely eradicate all spores, even with prolonged exposure times. [75] Glutaraldehyde, peracetic acid, and 10% hydrogen peroxide were the most effective agents in a study of *B subtilis* spores, but none totally eradicated all spores. [76] Some mycobacterial species have been reported to be resistant to even prolonged exposure to glutaraldehyde solutions. [77]

Common Chemical Disinfectants and Sterilants

A large number of chemical disinfectants and sterilants may be used in hospitals or endoscopy suites (Table 136-2 and Table 136-3). Several factors determine the suitability of a given agent for endoscope reprocessing: (1) The agent must be effective against a broad range of viruses, fungi, and bacteria, including mycobacteria. (2) The time of exposure necessary for this activity must not be prohibitively long. (3) The agent must not damage the endoscope or accessories. (4) The agent should not be toxic to staff or patients. (5) The cost must not be prohibitively high.

AGENT	ATTRIBUTES	DISADVANTAGES
Ethylene oxide (gas sterilant)	Highly effective (including action against spores)	Takes hours Potential toxicity to staff
Glutaraldehyde	Exposure times reasonable Relatively inexpensive Large experience with product	Potential toxicity to staff Not sterilant with 20-min exposure May cause colitis in patients
Peracetic acid	Highly effective (including action against spores) Not toxic to staff Effective against organic matter	More costly than glutaraldehyde
Hydrogen peroxide	Highly effective (including action against spores)	May cause colitis in patients

TABLE 136-2 Commonly Used Disinfectants and Sterilants for Endoscope Reprocessing

AGENT	REPORTED DISADVANTAGES
Iodophor solutions	May not reliably eradicate *Mycobacterium tuberculosis*[80] May be difficult to rinse from endoscopes
Hypochlorite solutions	May be less active against organic matter May corrode endoscope components
Quaternary ammonium compounds	Not sufficiently active against some microorganisms, including mycobacteria, some viruses
Alcohols	Flammable; may be a hazard if not properly handled Not effective against spores May not penetrate some proteinaceous material
Phenols	Residual material may remain even after rinsing, posing risk of irritation of mucous membranes

TABLE 136-3 Chemicals Not Typically Used as Sole Disinfectants in Endoscope Reprocessing

Gas sterilization of endoscopes has long been established as an effective method. Ujeyl and coauthors [78] found that as little as a 20-minute exposure time rendered endoscopes free of microorganisms with no apparent damage. Alfa and associates [79] raised concerns about the ability of sterilizers to eradicate completely organisms from the narrow lumens of endoscope channels. Multiple bacteria, including *P aeruginosa,* mycobacteria, and bacterial spores, were inoculated into plastic tubing 3.2 mm in diameter and sterilized with several agents, including ethylene oxide. None of the tested methods completely sterilized the narrow lumen when serum and salt were also present. [79] This result emphasizes the need for mechanical cleaning before disinfection and sterilization with any gas or chemical.

One commonly used chemical sterilant is glutaraldehyde. In the exposure time typical for endoscopy suites, it is an effective high-level disinfectant. If allowed sufficiently long contact times, sterilization is possible. Effective killing of bacteria, tubercle bacilli, and viruses occurs within 10 minutes, and vegetative spores in less than 3 hours. [80] Kovacs and colleagues [81] found that 2% glutaraldehyde achieved high-level disinfection from *M chelonae* in endoscopes with only a 10-minute exposure, but only if mechanical cleaning and alcohol rinse were also performed. However, not all clinical series support effective killing of mycobacteria within 10 minutes. The length of exposure with 2% alkaline glutaraldehyde time to achieve 99% eradication of all mycobacterial species ranged from 10 to 40 minutes (10 minutes for *M tuberculosis*) in another study. [82] These data have led to differing recommendations for the exposure time necessary for 2% glutaraldehyde to achieve high-level disinfection. The U.S. Food and Drug Administration (FDA) had required labeling of Cidex (2% glutaraldehyde, Johnson & Johnson) as effective as a high-level disinfectant only after a 45-minute exposure at 25°C. [83] However, Rutala and Weber [83] and prior APIC guidelines [6] argued that this amount of exposure time was only necessary if no mechanical cleaning of the endoscope had occurred before glutaraldehyde treatment and that exposure times of at least 20 minutes were sufficient to achieve high-level disinfection if mechanical cleaning was performed. Guidelines of several professional societies support a 20-minute exposure time for high-level disinfection, [84], [85] as do the most recent APIC guidelines for infection prevention and control in flexible endoscopy. [86] Testing of endoscopes contaminated with *Enterococcus* species or *H pylori* confirms these recommendations. [87] and [88] Recently, new glutaraldehyde preparations of varying concentrations, intended for use at varying temperatures and exposure times, have been introduced; it is essential that manufacturers' recommendations be reviewed and followed.

Rhinitis, dermatitis, nausea, headache, and respiratory difficulties are among the reported adverse effects on staff of glutaraldehyde exposure. [89], [90], [91], [92] and [93] Epidemiologic studies have suggested that spontaneous abortions are higher among staff performing sterilization with ethylene oxide, but not glutaraldehyde. [94] Adequate ventilation is necessary in areas where this agent is handled; staff should wear appropriate protective equipment, and the atmosphere should be periodically tested to be sure that concentrations are within appropriate parameters.

Glutaraldehyde has been linked to episodes of colitis in patients undergoing endoscopy. In one series, [95] 6 patients experienced low-grade fever, tenesmus, and bloody diarrhea, with symptoms lasting about 5 days. It was felt that the colitis was due to a gradual buildup of glutaraldehyde in the post-reprocessing rinse water and failure of the staff to use forced air to dry the channels after reprocessing. In a similar outbreak, most of the cases were in patients whose sigmoidoscope was reprocessed by a new staff member who did not flush residual glutaraldehyde from the endoscope channels after reprocessing. [96]

Peracetic acid is an effective chemical sterilant, active against all organisms, including bacterial spores. [97], [98] Bradley and colleagues [97] evaluated the Steris System 1 Processor (Stersis Corp., Mentor, OH) (0.2% peracetic acid) in the reprocessing of endoscopes contaminated with vegetative bacteria and spores. All bacteria were killed within 1 minute, and spores within 5 minutes. The investigators noted that the cost per cycle was higher than for reprocessing with glutaraldehyde. Whitbourne and Preston [99] confirmed the efficacy of the Steris processor on 10 medical devices contaminated with high titers of bacterial spores (about 107 colony-forming units/mL). [99] The process involves exposure of the medical device to the peracetic acid for 12 minutes, followed by several automatic rinses with water sterilized by passage through a membrane filter, for a total cycle time of about 25 minutes. Fuselier and Mason [100] reprocessed cytoscopes for 1 year using the Steris System 1 processor, and then used high level disinfection with Voluntary Hospital Association Plus 2% glutaraldehyde solution the next year. They reported no clinical differences between the two systems, but no microbiologic data were presented. Yearly operating costs for the Steris processor totaled $6037, compared with $445 for the HLD system. [100]

Hydrogen peroxide is another agent active against a wide range of microorganisms. Vesley and coauthors [101] tested the efficacy of 2% glutaraldehyde, 6% hydrogen peroxide, and ethylene oxide in endoscopes contaminated with *Pseudomonas* or *Bacillus* species. Ethylene oxide was the most effective agent overall. Hydrogen peroxide was more effective than 2% glutaraldehyde against the *Bacillus* organisms. The exposure times for these liquid chemicals was only 10 minutes, however, not the minimum 20 minutes recommended by the APIC for glutaraldehyde. As with glutaraldehyde, hydrogen peroxide may cause iatrogenic colitis. [102]

ESSENTIAL CONCEPTS IN REPROCESSING OF ENDOSCOPES AND ACCESSORIES

The overriding principle in endoscope reprocessing is that each patient must be assumed to possess potentially an infectious agent that could be passed through the endoscope to a subsequent patient. It is inappropriate for an endoscopy unit to reserve an endoscope or set of endoscopes for patients identified with some known pathogen. Rather, processes must be in place where all endoscopes are reprocessed in such a way that all potential pathogens would be eliminated.

Several professional societies, including the APIC, the Society of Gastroenterology Nurses and Associates (SGNA), and the ASGE have published guidelines for proper reprocessing of endoscopes and accessories. [84], [85] and [86] Essential aspects of these guidelines are presented below and summarized in Table 136-4. These are not intended to be a substitute for the step-by-step detail presented in the guidelines.

1. Inspection for damage should be performed after each case. Leakage testing may identify internal damage not apparent from endoscope performance.
2. Mechanical cleaning before exposure to the disinfectant or sterilant is essential.
3. High-level disinfection is the current standard for endoscope reprocessing.
4. Endoscopic accessories that may contact the vascular system must be sterilized if reused; mechanical cleaning alone or high-level disinfection is inadequate.
5. Channels must be adequately dried after high-level disinfection to prevent colonization with microorganisms.
6. If glutaraldehyde is used for high-level disinfection, the effective concentration must be continually monitored.
7. Continuous quality improvement is essential for each endoscopy unit.

TABLE 136-4 Some Essential Concepts in the Reprocessing of Endoscopes and Accessories

Inspection for Damage

Leaks, tears, and other disruptions to the external or internal integrity of the endoscope may be a nidus of continual colonization by microorganisms. [54] Internal damage to the endoscope may not be apparent to the endoscopist until it is far advanced because the endoscope may perform well. The endoscope must be carefully

inspected before reprocessing. Leak testing may detect internal structural damage at an early stage.

Mechanical Cleaning

Removal of organic material before exposure to chemical disinfectants or sterilants is the most important step in endoscope reprocessing. Scrupulous cleaning of external surfaces, use of enzymatic detergents on external surfaces and internal channels, passing a brush through the operating channel, brushing or forcing air through the more narrow air-water channel, and manual or ultrasonic cleaning of detachable parts of the endoscope are all part of this process. Special attention must be given to difficult-to-access portions of the endoscope, such as elevators and elevator channels of the duodenoscope. The elevator should be mechanically cleaned and the channel flushed.

Removal of organic and foreign material is necessary to achieve high-level disinfection. Alfa and associates [103] studied the suction channels of 10 bronchoscopes, duodenoscopes, and colonoscopes immediately after the procedure. They identified multiple compounds and organisms, including protein, hemoglobin, carbohydrate, endotoxins, and bacteria. Mechanical cleaning eliminated a good deal of this material, but protein and some bacteria remained. Lewis and Arens [104] noted persistence of HIV and vegetative bacteria after high-level disinfection of dental devices when the organism was entrapped in commonly used lubricants. The necessary exposure time of the liquid chemical sterilants to achieve high-level disinfection is decreased by prior mechanical cleaning.

Mechanical cleaning of the endoscope, without exposure to chemical sterilant, eliminated microorganisms in one study from 66 of 68 sites tested. [18] Other studies document the efficacy of mechanical cleaning in the inactivation of hepatitis B virus, [105] hepatitis C virus, [106] *H pylori,* [107], [108], [109] and [110] gram-negative bacteria, [111] and *Clostridium difficile.* [112] Thorough mechanical disinfection plus use of an approved chemical disinfectant or sterilant is highly effective in eliminating bacteria and viruses from endoscopes but is not effective when mechanical cleaning is ineffective. [113]

Use of a Recommended Disinfectant or Sterilant for High-Level Disinfection

Manual high-level disinfection may still be performed, but, with the advent of totally submersible endoscopes, automated reprocessors have become more common. These machines lessen the amount of time staff members must devote to the overall process and lessen exposure to glutaraldehyde, if this is the agent of choice. Vesley and colleagues [101] showed that automated reprocessors were more effective than manual high-level disinfection in killing *B subtilis*. However, Fraser and associates [114] found no difference in overall endoscope contamination rates when endoscopes disinfected manually were compared with those disinfected in automated reprocessors. Automated reprocessors do have potential drawbacks. Persistent infection of the endoscope reprocessor with resultant contamination of the endoscope has been documented. [42] The endoscope must be properly positioned within the machine, with all adapters and covers correctly placed. Furthermore, with endoscopic automated reprocessors using glutaraldehyde, the effective concentration of the disinfectant will decrease with repeated use. [115] Dilute glutaraldehyde is less effective in its action against microorganisms. [116], [117] A test strip or other device must be used at least daily to monitor the concentration of the liquid disinfectant, possibly more frequently depending on procedure volume. Solutions must be changed in accordance with the manufacturer's recommendations. Dilution must also be monitored with the use of hydrogen peroxide. A definite advantage of the Steris processor is that the chemical sterilant is not recycled; therefore, this monitoring process is not necessary. Orthophthalaldehyde is a high-level disinfectant that appears to be stable for up to 14 days of use. [118] Orthophthalaldehyde is approved by the FDA for use in endoscope reprocessing. It is used in some parts of the world, primarily outside the United States. [86]

Rinsing and Drying

The chemical sterilant must be rinsed from the internal and external surfaces of the endoscope. If tap water (which may be colonized with microorganisms) is used, a rinse with 70% alcohol should be performed. Drying with compressed air (or drying cycles on some automated washers) will also decrease the likelihood of residual moisture, which can be a source of colonization. Drying of endoscopes before prolonged storage significantly decreases the rate of bacterial colonization, [119] and forced air drying adds effectiveness to the disinfection process. [120] Alcohol rinse before prolonged storage also may decrease the likelihood of colonization. Endoscopes should be stored with the insertion tubes uncoiled and in a well-ventilated cabinet to facilitate continued air drying.

Reprocessing of Endoscopic Accessories

Because biopsy forceps and other accessories may come into contact with the vascular system, these items should be sterile before each use. Both disposable (single-use) and reusable forceps are commercially available. Improperly reprocessed biopsy forceps may be the source of transmission of infection. Single-use, disposable forceps do not expose patients to this risk. Two recent trials support the use of disposable over reusable forceps in terms of cost and function. [121], [122]

Most accessories for therapeutic ERCP, including papillotomes, guide wires, and extraction balloons, are labeled as single-use items. These items incur a significant cost for the endoscopy suite and are not entirely compensated by technical room reimbursements. [123] Reusable papillotomes are available. These devices function well after multiple uses and sterilizations but cost more than single-use papillotomes. [124], [125] The cost associated with endoscopic accessories has led some centers to reprocess items labeled as single use. Kozarek and colleagues [126] found that most single-use papillotomes performed well with multiple uses and sterilizations. No organisms were recovered from the devices after manual cleaning followed by ethylene oxide sterilization. In a follow-up study, this group found that the single-use papillotomes could be used an average of 3.4 times, and such reuse saved about $66,000 in 1 year for their practice. No infectious complications were attributed to the reuse. [127] These authors have also studied the reuse of other single-use items, such as argon plasma coagulation probes, and assert that sterilization could be achieved and up to 10 uses from a single probe obtained. [128] In a 1995 survey of hospitals in Australia, 58% were reusing at least some medical devices labeled as single use, [129] and a more recent survey documented that reuse is common in the Asia-Pacific region. [130] There are no data to indicate how common this practice is in the United States. The most recent APIC guidelines state only that this practice is "controversial" and that implementation of such a program "requires a major institutional commitment, including a monitoring committee with clearly defined protocols." [86]

Continuous Quality Improvement

Proper reprocessing of endoscopes and accessories requires an organized plan, motivated and educated staff, and a continuous quality improvement process that monitors each step of the process. Breakdowns in reprocessing techniques can lead to clinically significant infections and even death. The process can fail because of technical or human error. Staff and processes must be assessed and improved continually. Each unit should develop its own plan and should generate data on factors including competence of staff and results of random cultures from endoscopes, reprocessors, and accessory equipment.

COMPLIANCE WITH RECOMMENDATIONS FOR ENDOSCOPE REPROCESSING

One concern about endoscope reprocessing is the complexity of the process and the potential that some steps might be omitted or performed inadequately in a busy endoscopic practice. Several investigators have examined the actual practice of endoscope reprocessing, either with surveys or site visits. A 1988 survey of 74 endoscopy suites in Western Europe revealed that if a procedure was just completed in a patient with no known infectious disease, high-level disinfection was not performed after upper endoscopy in any center, in only 13% of centers after colonoscopy, and in only 30% of centers after ERCP. [131] In a 1989 survey, only 45% of Australian hospitals responding followed a process for high-level disinfection after each case. [132] Two years later, a repeat survey indicated that high-level disinfection was satisfactorily performed by 75% of respondents. [133] A 1991 survey in 107 endoscopy units in North Carolina found that glutaraldehyde was the chemical disinfectant used in 91% of units. Only 51% exposed the endoscope to the glutaraldehyde for at least 20 minutes (a 10-minute cycle was used in most other centers), and 54% rinsed with only tap water (not sterile water or alcohol after tap water) after disinfection. [134] A U.S. survey of SGNA members and nonmembers with an interest in endoscope reprocessing found that 93% performed chemical disinfection after each case; glutaraldehyde was the disinfectant used in 88% of respondents' practices. [135]

A more sobering report came from Kaczmarek and coauthors, [136] who directly studied the endoscopic reprocessing practices in 26 endoscopy suites: 24% of endoscopes cultured after high-level disinfection were positive for bacterial growth. A lack of sterilization of all biopsy forceps was observed in 78% of units. Several other basic errors in the disinfection process were observed. Variability was also observed in 18 endoscopy areas in eight Massachusetts hospitals; however, high-level disinfection was practiced in 17 of 18 endoscopy areas. Glutaraldehyde soak times varied from 10 to 45 minutes, and not all areas performed sterilization of biopsy forceps. [137] In a more recent survey, a significant minority of U.S. centers did not completely conform to established society guidelines for reprocessing. [138] In a

1996 survey of 20 Japanese endoscopy suites, endoscopes in all centers were used in consecutive patients with mechanical cleaning but no high-level disinfection. [139]

FUTURE TRENDS

Proper reprocessing of endoscopes demands time and careful attention, is not foolproof, and is associated with significant cost. Therefore, the concept of a sheathed or disposable endoscopic system is very attractive. A new endoscopic system in which the working surfaces are covered with a disposable sheath has been assessed in clinical trials. [140], [141] In a prospective trial, the length of time required to perform endoscopy was slightly longer than with the conventional endoscope, whereas the set-up and reprocessing times were far shorter. [142] Recent reports indicate promise for new techniques of endoscope reprocessing using superoxidized or acidic electrolytic water. [143], [144] and [145]

SUMMARY

Introduction of infection into patients undergoing endoscopy is rare, but it may occur. Infection may be due to patient-to-patient transmission or introduction of microorganisms from the environment into patients. Potential sources of infection include the endoscope, accessories, and automated reprocessors. High-level disinfection is recommended for reprocessing endoscopes, but sterilization is indicated for accessories that may come into contact with the vascular system. The process of high-level disinfection requires close attention to multiple steps. Several clinical series have demonstrated breakdowns in this process, with resultant morbidity and mortality. A continuous quality improvement system is essential for each endoscopy center.

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CHAPTER 137

Martin L. Freeman

SEDATION AND MONITORING FOR GASTROINTESTINAL ENDOSCOPY

Delivery of safe but effective sedation and analgesia can be a challenge for the endoscopist. Cardiopulmonary complications are still one of the leading causes of morbidity and mortality due to endoscopic procedures. To administer sedation properly, it is necessary for endoscopists to understand the definitions and hazards of conscious versus deep sedation, the role of electronic monitoring devices such as pulse oximetry, and the pharmacology of the agonist and reversal agents used. They must develop a systematic approach to sedation and analgesia, in conjunction with well-trained gastrointestinal assistants, and know the limitations of their abilities.

CONSCIOUS VERSUS DEEP SEDATION: DEFINITIONS AND JCAHO REGULATIONS

The term *conscious sedation* has traditionally been used to describe the pharmacological state of restfulness induced for such procedures as endoscopy (Table 137-1). A recent Task Force of the American Society of Anesthesiologists has suggested that *sedation and analgesia* most accurately describes the state that allows patients to tolerate unpleasant procedures while maintaining adequate cardiorespiratory function, and the ability to respond purposefully to verbal or tactile stimulation. [1] They suggest that *conscious sedation* is an imprecise term and should be replaced. *Monitored anesthesia care* (MAC) is another term currently used to differentiate sedation and analgesia without endotracheal intubation from general anesthesia.

1. *Minimal sedation (anxiolysis).* A drug-induced state during which patients respond normally to verbal commands. Although cognitive function and coordination may be impaired, ventilatory and cardiovascular functions are unaffected.
2. *Moderate sedation or analgesia ("conscious sedation").* A drug-induced depression of consciousness during which patients respond purposefully to verbal commands, either alone or accompanied by light tactile stimulation. No interventions are required to maintain a patent airway, and spontaneous ventilation is adequate. Cardiovascular function is usually maintained.
3. *Deep sedation or analgesia.* A drug-induced depression of consciousness during which patients cannot be easily aroused, but respond purposefully following repeated or painful stimulation. The ability to maintain ventilatory function independently may be impaired. Patients may require assistance in maintaining a patent airway, and spontaneous ventilation may be inadequate. Cardiovascular function is usually maintained.
4. *Anesthesia.* Consists of general anesthesia and spinal or major regional anesthesia. It does not include local anesthesia. General anesthesia is a drug-induced loss of consciousness during which patients are not arousable, even by painful stimulation. The ability to maintain ventilatory function independently is often impaired. Patients often require assistance in maintaining a patent airway, and positive-pressure ventilation may be required because of depressed spontaneous ventilation or drug-induced depression of neuromuscular function. Cardiovascular function may be impaired.

From ref. 7.

TABLE 137-1 Defining Levels of Sedation and Anesthesia

Sedation is defined as a reduction in the level of consciousness induced by the medications used to facilitate acceptance of endoscopic procedures. Sedation may range from minimal or no visible change in patient status to loss of consciousness and protective reflexes. For some patients, no sedation may be required, and topical anesthesia may suffice for upper endoscopic procedures. Analgesia is defined as the reduction in pain or perception of nociceptive stimuli induced by the use of medication, primarily opioids. Analgesics are commonly used in low or moderate doses to reduce discomfort without impairment in consciousness, but higher doses can depress respiration and induce sedation. Patients who are given analgesics in higher doses may experience suppression of ventilatory drive, depression of consciousness, and impairment of protective reflexes.

The usual intended goal for sedation and analgesia may be defined as a medically controlled state of depressed consciousness that (1) allows protective reflexes to be maintained, (2) retains the patient's ability to maintain a patent airway independently and continuously, and (3) permits appropriate response by the patient to physical stimulation or verbal commands such as "open your eyes" [2], [3] (see Table 137-1). Conscious sedation, as summarized by the Joint Commission on Accreditation of Healthcare Organizations (JCAHO), is most appropriate for a patient who will accept it willingly and is able to cooperate. On the other hand, it is not suitable for a patient who is uncooperative, mentally impaired, or extremely anxious, and such patients may be best approached with general anesthesia, particularly if a prolonged therapeutic procedure, such as endoscopic retrograde cholangiopancreatography (ERCP), is to be undertaken.

A degree of safety is implied when a state of conscious sedation is achieved compared with deep sedation or general anesthesia. Although one might assume that the degree of care provided to patients under conscious sedation could be less intense or thorough, moving from a state of consciousness to deep sedation is a dose-related continuum that also depends on patient response. The state originally intended might not be the one ultimately achieved. There is a large variability in the pharmacokinetics and pharmacodynamics of sedative drugs in patients; thus, a "standard" dose of sedatives may produce undersedation in some patients and substantial oversedation resulting in anesthesia in others. [4], [5] and [6] Therefore, those who administer sedatives should have the knowledge and equipment, including

monitoring devices and airway management skills, to deal with an oversedated patient.

The JCAHO had previously issued a strict set of standards for anesthesia care that apply when patients, in any setting, receive, for any purpose or by any route, either general anesthesia or sedation (with or without analgesia) that, in the manner used, may reasonably be expected to result in the loss of protective reflexes. [3] Since that time, the JCAHO has developed new definitions of levels of sedation and analgesia. The impetus for this change was in part the recognition that loss of protective reflexes is often a late indicator that a patient receiving conscious sedation is in trouble and in part anecdotal evidence from the field that negative outcomes and even death were occurring as a result of conscious sedation. [7] Following the lead of the American Society of Anesthesiologists, the JCAHO has issued a new set of definitions (see Table 137-1). They emphasize the continuum that exists between the level of sedation intended and that which is achieved. Importantly, for moderate and deep sedation, the JCAHO emphasizes the need for qualified personnel, specifically with respect to competency to manage a compromised airway, to provide adequate oxygenation and ventilation, and to manage an unstable cardiovascular system. The JCAHO guidelines reflect the narrowing gap between practices in the anesthesia and endoscopy communities. The American Society of Anesthesiologists published a set of guidelines for nonanesthesiologists who administer sedation and analgesia. [1] The American Society for Gastrointestinal Endoscopy (ASGE) issued a set of standards of practice specific to monitoring and sedation for gastrointestinal endoscopy. [8] These two documents set forth relatively similar guidelines pertaining to personnel; training; equipment; appropriate preoperative, intraoperative, and postoperative assessment and monitoring; and use of sedative and analgesic agents (Table 137-2).

Preprocedure assessment of the patient, including assessment of eligibility for sedation and analgesia, previous adverse experiences with sedation and analgesia, coexisting medical conditions, allergies, current medications, cardiopulmonary status, and ascertainment of preprocedure fasting
A well-trained gastrointestinal assistant present whose primary role is patient sedation and monitoring
Preprocedure documentation of blood pressure, pulse, respiratory rate, and oxygen saturation
Equipment set up:
Nasal oxygen cannula
Oral suction catheter
Airway resuscitation equipment
Reversal agents immediately available
Establishment of intravenous access
Intraprocedural monitoring of heart rate, blood pressure (frequent, automated monitoring mandatory for high-risk patients), respiratory rate, pulse oximetry, electrocardiogram (mandatory for high-risk patients)
Use of supplemental oxygen as appropriate
Education of personnel regarding pharmacology of agents administered
Titration of sedative or analgesic agents to effect
Documentation of medications delivered, vital signs, respiratory parameters, and level of consciousness throughout procedure
Postprocedural monitoring of level of consciousness, pulse oximetry, and vital signs
Special regimens or consultation with appropriate medical specialty or anesthesiology for patients with special problems such as severe cardiopulmonary disease, uncooperative mental state

From refs. 1 and 8.

TABLE 137-2 Practice Guidelines for Sedation and Analgesia by Nonanesthesiologists

CARDIOPULMONARY COMPLICATIONS OF ENDOSCOPY

Cardiopulmonary complications are said to cause 50% of the morbidity and 60% of the mortality associated with endoscopy. [9], [10], [11], [12], [13], [14], [15] and [16] The true incidence of complications is not known and is likely underestimated because most data are derived from retrospective surveys rather than prospective studies specifically designed to study cardiopulmonary complications. One major recall-based U.S. survey suggested that only 0.5% of ASGE members responding experienced any cardiopulmonary complication annually. [17] Assuming 500 procedures per year per endoscopist (roughly the median for the respondents), this represents a complication rate of 0.01 per 1000 procedures. In contrast, a retrospective study using data entered at the time of endoscopy into an ASGE computer-based management system suggested that a serious cardiopulmonary complication occurred in 5.4 per 1000 procedures, [18] a 500-fold higher incidence; mortality alone was 0.3 per 1000. Another ASGE database study described one respiratory or cardiopulmonary arrest occurring per 1000 procedures. [19] One of the few truly prospective studies [16] found a 30-day procedure-related cardiopulmonary mortality rate of 0.4 per 1000 diagnostic upper endoscopies in the United Kingdom. It has been suggested that this cardiopulmonary complication rate has not decreased since the 1980s, in stark contrast to anesthesia-related deaths, which have decreased to 1 per 26,000 (0.04 per 1000), presumably as a result of better training standards and the widespread adoption of monitoring equipment. [10]

Two large studies have shown that procedures involving administration of opioids such as meperidine (usually given in combination with benzodiazepines) were most likely to be associated with adverse events. [18], [20] ERCP is associated with the highest rate of hypoxemia, although cardiopulmonary outcomes for this procedure in clinical practice outside randomized controlled trials have not been specifically studied. [12], [21], [22] and [23] Therapeutic procedures generally have a higher rate of complications than diagnostic ones, [24], [25] and acute upper gastrointestinal bleeding is associated with a high incidence of aspiration and other complications. [26], [27]

Types of Cardiopulmonary Complications

At least six major types of cardiopulmonary complications may occur as a result of endoscopy. These include respiratory depression, pulmonary aspiration, cardiac arrhythmias, myocardial ischemia, hemodynamic disturbances, and allergic reactions. In addition, indirect morbidity may occur because of inability to complete a therapeutic procedure owing to sedation-related problems. [21]

Respiratory Depression

Respiratory depression can manifest as hypoxemia or CO_2 retention, usually but not always occurring in parallel. [21], [22] A wealth of data show that arterial oxygen desaturation occurs frequently during both upper and lower gastrointestinal endoscopy. [10], [21], [22] and [23], [28], [29], [30], [31], [32], [33], [34], [35], [36], [37], [38], [39], [40], [41] and [42] Defining significant arterial oxygen desaturation as oxygen saturation (SpO_2) less than 90%, these studies show that at some point, up to 40% of patients undergoing upper endoscopy, 50% of patients undergoing colonoscopy, and perhaps 60% or more of patients undergoing ERCP experience significant desaturation. The mechanisms of hypoxemia are varied: opioids and benzodiazepines depress respiration both centrally by blunting ventilatory response to CO_2 and peripherally by blunting hypoxic ventilatory drive. [43], [44], [45], [46], [47], [48] and [49] The physical presence of the endoscope appears to cause minor degrees of hypoxemia, [50], [51] and [52] presumably as a result of coughing or aspiration, or by a reflex mechanism; significant airway obstruction is unlikely because even large-diameter endoscopes do not appear to cause sustained CO_2 retention. [22] Greater degrees of hypoxemia occur with larger-diameter endoscopes [36], [40], [41] and in examinations performed by inexperienced endoscopists. [53] Splinting or looping during colonoscopy may cause transient desaturation. [32] Endoscope-induced desaturation is generally transient or minor; severe and prolonged desaturation, particularly if culminating in respiratory arrest, is almost invariably the result of drug-induced respiratory depression, sometimes occurring after the endoscope has been removed. [21], [19], [22]

The significance of oxygen desaturation during endoscopy has been questioned by some investigators. [54], [55] However, these small series lacking prospective determination of patient outcome do not allow the conclusion that hypoxemia is safe, particularly because adverse events are relatively rare. It is likely that most hypoxic episodes are well tolerated. However, sustained desaturation to less than 90%, the point at which the oxyhemoglobin dissociation curve becomes very steep, may be of concern in elderly patients and in those with ischemic heart disease. [56], [57] Substantial data suggest that hypoxemia during endoscopy may result in tachycardia, [33] ST-segment elevation or depression indicative of ischemia, [33], [36], [37], [58], [59] and [60] and both atrial and ventricular arrhythmias. [36], [61] Hypoxemia is thought to cause most cardiac arrhythmias occurring during endoscopy. [10], [36]

A few studies suggest that patient variables such as increased age, obesity, or pulmonary disease increase the likelihood of desaturation. [23], [39], [62] Most studies, however, have shown that it is impossible to predict by medical illness or age which patients will become hypoxemic. [21], [22], [29], [32], [38], [55]

Use of Supplemental Oxygen

Numerous studies show that routine use of low-flow nasal oxygen during endoscopy prevents or diminishes hypoxemia. Bell and colleagues [31] showed that

administration of 2 L per minute nasal oxygen prevented hypoxemia in patients undergoing upper endoscopy with midazolam. Gross and Long [34] showed that 3 L per minute nasal oxygen reduced by more than half the incidence of hypoxemia in patients undergoing colonoscopy with midazolam and meperidine. Other studies have shown that oxygen reduces but does not abolish hypoxemia during endoscopy, [33], [35] suggesting that the use of oxygen does not eliminate the need to monitor oxygen saturation.

Does oxygen administration prevent deleterious events? During ERCP, patients with saturations below 90% had significantly higher pulse rates than patients who were not hypoxic, suggesting that correction of oxygen may alleviate tachycardia and hence myocardial stress. [33] Another intriguing study showed that administration of oxygen, compared with room air, prevented ST-segment changes indicative of ischemia in patients undergoing upper endoscopy. [63] Based on these findings, and in keeping with standard practice in anesthesia, Bell and others [9], [10] advocate routine preadministration of oxygen for most patients having sedation or endoscopy. Advantages to this approach include low cost (about $3 per patient) and proven prevention of hypoxemia in most cases. Preoxygenation does remove the ability of the oximeter to reflect hypoventilation or apnea, however, as discussed in the next section.

A logical approach would be that recommended by the British Society of Gastroenterology [50]—to preadminister nasal oxygen to those patients who can least tolerate hypoxemia (elderly patients and those with coronary artery disease). With those exceptions, I recommend reserving oxygen through nasal cannula, which should be set up at the bedside before each case, to patients who have sustained desaturation not responsive to verbal encouragement to breathe. Oxygenating mouth guards have been shown to be as effective as nasal cannulas, [64] but they have the distinct disadvantage of being removed immediately after the procedure, a time at which maximal desaturation may occur. [19], [20], [21] and [22] Generally, 2 to 3 L per minute through nasal cannula is adequate. Even higher flow rates occasionally may not correct hypoxemia, especially if both opioids and benzodiazepines are given. [10], [27] Furthermore, higher flow rates of oxygen probably should be avoided because they can mask severe CO_2 retention. [21], [22]

Carbon Dioxide Retention

Pulse oximetry measures arterial oxygen saturation, and not alveolar hypoventilation, which is directly reflected by increases in arterial CO_2 tension. Severe CO_2 retention may occur during endoscopy. [21], [22] The degree of hypoventilation has been shown to be in part related to sedative drug doses, which is logical because the commonly used sedatives or analgesics cause CO_2 retention by centrally depressing ventilatory responsiveness to CO_2. [44], [47], [49] CO_2 retention does not correlate well with the clinically assessed level of sedation or with the presence of underlying medical illnesses such as cardiac or obstructive lung disease. [21], [22] Although oxygen supplementation readily corrects hypoxemia, it does not correct underlying hypoventilation; once oxygen is applied, severe hypoventilation may occur without reliable detection by either pulse oximetry or clinical observation. Transient desaturation in patients already on oxygen generally indicates severe CO_2 retention.

Aspiration

Pulmonary aspiration of gastric contents or blood potentially may result in pneumonia and adult respiratory distress syndrome as well as cardiopulmonary arrest. Inhalation of gastric contents carries a high risk for mortality: 5 deaths were reported among 16 cases of inhalation of gastric contents in a survey of endoscopy in the United Kingdom. [65] Pneumonias occurred in 11 of 1300 patients after upper digestive endoscopy, with 8 deaths [16]; a correlation was seen between use of topical anesthetic sprays and the development of pneumonia. Aspiration is particularly likely when protective reflexes are blunted by excessive sedation or encephalopathy, when significant amounts of fluid or food are still in the stomach, as in patients with diabetic gastroparesis, and in the setting of acute upper gastrointestinal bleeding. In a prospective study, 20% of patients undergoing endoscopy for acute upper gastrointestinal bleeding developed clinically apparent aspiration pneumonia. [27] Aspiration did not occur in patients with endotracheal intubation.

The following precautions should be taken to avoid aspiration during upper endoscopy:

A vigilant gastrointestinal assistant should be prepared to suction the oropharynx with a catheter.
When large quantities of food or fluid are encountered, excessive insufflation should be avoided and the procedure should be terminated unless essential.
Sedation and topical anesthetic sprays should be minimized.

Endotracheal intubation or use of esophageal overtubes is recommended to protect the airway for patients with active upper gastrointestinal bleeding who have ongoing active hematemesis, are unstable, are mentally obtunded, or are uncooperative. [66]

Cardiac and Hemodynamic Complications

Myocardial ischemia and infarction may occur during endoscopic procedures, particularly in those patients with cardiac disease. [25], [58], [59] and [60] There are few prospective data regarding the incidence of clinically significant arrhythmias during gastrointestinal endoscopy. Tachyarrhythmias or bradyarrhythmias may occur as a result of hypoxemia, or from stress due to noxious stimuli. [61] Arrhythmias are more common in patients with a history of cardiopulmonary disease. [36] Although the importance of hypoxemia in producing cardiac arrhythmias and ischemia has been stressed, [9], [10], [61] other factors may be important. In one study of patients undergoing ERCP, Holter monitoring showed that myocardial ischemia correlated better with periods of tachycardia than with periods of hypoxemia. [58] Tachycardia is frequent [33] and may be extreme, with heart rates reported up to 200 beats per minute during stressful, prolonged upper procedures such as ERCP. [67] Tachycardia occurs mostly during upper rather than lower gastrointestinal endoscopic procedures [22] and most commonly occurs in elderly patients and those with cardiac disease. [23] Significant increases and decreases in blood pressure occur during gastrointestinal endoscopy. [32], [37], [68] In a detailed study of 395 patients undergoing ERCP, 7% of patients experienced a peak heart rate and blood pressure double product of greater than 25,000, equivalent to a maximal level cardiac treadmill test. [21] This is an enormous myocardial workload, particularly for the frail and elderly patients often undergoing these procedures.

Hemodynamic disturbances during endoscopy consist primarily of vasovagal reactions and of fluctuations in blood pressure and pulse. Vasovagal reactions usually manifest as perspiration and bradycardia and typically occur as a result of painful stimuli during colonoscopy. Although atropine is widely used to treat vasovagal reactions, few data support its routine use to prevent such events. Hypotension may result from vasodepressor effects of opioids, benzodiazepines, and other medications given during endoscopy.

To avoid the cardiac stress of tachycardia and hypertension during endoscopic procedures, two suggestions come to mind: first, atropine should not be given in upper gastrointestinal procedures unless bradycardia occurs; second, the often-stated axiom that the least sedation possible is the safest [10], [50], [69] may not be true. In a study of patients undergoing endoscopy without premedication, tachycardia occurred in 97% of subjects, ST-segment depression in 14%, supraventricular tachycardia in 6%, and ventricular ectopic complexes in 1.6%. [70] All of these changes were more frequent in cardiac than noncardiac patients. On the other hand, two studies have shown that opioid premedication attenuates endoscopy-induced increases in pulse and blood pressure, [71], [72] suggesting that optimal safety to the patient is a balance between preventing hypoxemia (which can be corrected with nasal oxygen) and preventing endoscopy-induced tachycardia and hypertension.

Procedure Termination as a Complication of Sedation

Procedure termination due to inadequate or excessive sedation has to be considered a sedation-related complication, resulting in morbidity due to delayed therapy, extended hospitalization, repeat or alternative procedures such as surgery, and increased cost. There are few published data on the prevalence of this problem, but it is not uncommon; Diab and others [73] found a 7% incidence of premature procedure termination during upper endoscopy using midazolam alone for sedation, without meperidine. In a prospective multicenter study, 4.1% of all ERCPs (mostly therapeutic) had to be terminated prematurely because of difficulty with sedation. [21] If difficulty is anticipated in sedating a patient for a therapeutic procedure, general anesthesia may be a safer and more cost-effective alternative.

MONITORING

Guidelines issued by the JCAHO, [3] the ASGE, [8] the British Society of Endoscopy, [50] and the American Society of Anesthesiologists [1] state that the most important aspect of monitoring a patient during sedation is a well-trained gastrointestinal assistant closely observing the patient. All of the above organizations now recommend the universal use of electronic monitoring as adjunctive devices, particularly pulse oximetry. The recent availability of compact, portable, all-in-one multichannel

oximeter–electrocardiograph–automated blood pressure units have greatly facilitated electronic monitoring and documentation during endoscopy.

Pulse Oximetry

A pulse oximeter is a spectrophotometric device that detects and calculates the differential absorption of light by oxygenated and reduced hemoglobin to produce a measurement called SpO_2, an estimate of oxygen saturation. [74] The rationale for using pulse oximetry is that it diagnoses hypoxemia before the signs and symptoms become apparent. [75], [76] One study showed that trained anesthesiologists in a well-lit operating room, blinded to oximetry, failed to recognize most major hypoxic events (sustained desaturation to less than 85%), including a number of patients with saturations below 72%. [75] Changes in respiratory rate and heart rate were seen in less than 15% of severe desaturations. A number of investigators have found that changes in respiratory rate or depth, as well as level of sedation, are poor indicators of hypoxemia or CO_2 retention during endoscopy, [21], [28], [77] so that even apparently awake and normally breathing patients may have severe respiratory depression. Pulse oximetry provides an early warning of impending respiratory failure (long before clinical observation) and thus allows the endoscopist to withhold sedation, stimulate the patient physically or verbally, and, if necessary, administer antagonists to reverse hypoventilation or manage the airway before clinically dangerous hypoxemia occurs.

Some endoscopists have resisted the notion that oximetry may be beneficial in endoscopy. [54], [55], [78] A retrospective survey from Switzerland, where pulse oximetry was rarely used (2.5% of procedures), claimed a minuscule cardiopulmonary complication rate (0.1%, with no deaths, in 115,200 endoscopies) and questioned the need for oximetry; the validity of complication data from such surveys, however, is questionable. Despite use of pulse oximetry, Iber and colleagues [79] reported a 7% incidence of major cardiopulmonary complications of sedation during endoscopy. In contrast, the prospective audit of endoscopy in the United Kingdom found a correlation between lack of monitoring and complications. [16] The controversy regarding use of electronic monitoring is fueled by a lack of studies assessing whether oximetry affects outcome in gastrointestinal endoscopy. Such a study is unlikely to be done in the setting of gastrointestinal endoscopy, owing to the relative rarity of adverse events and thus the huge numbers of patients necessary to detect a difference. A prospective, randomized evaluation of pulse oximetry in 20,802 patients undergoing primarily general anesthesia in the operating room was recently reported; using pulse oximetry resulted in a 19-fold increase in detection of hypoxemic events but did not significantly reduce rates of cardiovascular and respiratory complications. [80] However, in anesthesia, unlike endoscopy, airway and ventilation are actively controlled, in conjunction with oxygen supplementation, by trained practitioners not directly involved in the procedure. Pulse oximetry would be more likely to affect patient outcome in endoscopy, in which the patient is required to ventilate spontaneously while being given respiratory depressant drugs by less trained practitioners in a darkened room.

Finger pulse oximetry has a number of practical limitations: motion may cause false alarms when the probe becomes dislodged, and a displayed pulse waveform, or plethysmogram, is essential so that true desaturations can be distinguished from motion artifact (i.e., an arterial waveform correlates with the pulse). Pulse oximeters may be inaccurate with extreme hemodynamic disturbances such as shock or vasoconstriction. [74] As discussed, pulse oximetry may not reflect even severe hypoventilation (CO_2 retention) in a patient receiving supplemental oxygen. [21], [22]

Carbon Dioxide Monitors

Technical limitations presently make noninvasive CO_2 monitoring impractical. Although end-tidal CO_2 is a relatively accurate indicator of arterial $PaCO_2$ in endotracheally intubated patients undergoing mechanical ventilation, monitoring of end-tidal CO_2 through nasal cannula in a spontaneously breathing patient is grossly inaccurate and should not be used in gastrointestinal endoscopy or other settings to estimate arterial PCO_2. [74], [81] Its main utility during sedation and analgesia is to give a graphic display of respiratory rate, so that apnea can be detected in patients receiving supplemental oxygen. Although quite accurate [21], [22], [82], [83] and interesting from a research standpoint, transcutaneous CO_2 monitors are impractical for routine use because they require daily calibration and a 10- to 15-minute equilibration period on each patient. In a randomized controlled trial of CO_2 monitoring during ERCP, CO_2 monitoring effectively prevented the most drastic degrees of CO_2 retention. [21] There was little apparent difference in outcome of the patients as long as oxygen saturation was aggressively maintained with nasal oxygen. As with all studies of cardiopulmonary complications, enormous numbers of patients would be necessary to show a significant difference in clinical outcome.

Blood Pressure Monitoring

Assessment of blood pressure before, during, and after endoscopic procedures is generally considered to be a minimum requirement. [1], [3], [8], [84] Automated blood pressure monitors are being used more often during endoscopy, although their efficacy has not been documented. A prospective randomized study of 618 patients undergoing routine endoscopy found that blood pressure monitoring did not result in a difference in outcome. [68] In that study, patients with coronary artery disease were more prone to hemodynamic disturbances. Again, negative conclusions about the effect of monitoring on outcome are limited in a study of this sample size because adverse outcomes are rare.

During routine endoscopic procedures, blood pressure should be monitored about every 10 minutes. Under special circumstances, more frequent, automated blood pressure monitoring is advisable: blood pressure should be monitored every 3 to 5 minutes in patients with acute gastrointestinal bleeding; every 5 to 10 minutes in patients undergoing prolonged procedures, where hemodynamic disturbances are anticipated owing to higher than usual doses of sedatives; and every 5 to 10 minutes in patients with underlying cardiopulmonary disease who are less likely to tolerate severe fluctuations in blood pressure. Hypotension can be treated by withholding further benzodiazepines or narcotics, by giving intravenous fluids, or by giving reversal agents. Hypertension can be treated by giving more sedatives or analgesics if the patient is undersedated, by decreasing noxious stimuli such as looping of the endoscope, or by switching to a different agent such as droperidol if a prolonged procedure is anticipated.

Electrocardiographic Monitoring

Few specific guidelines have been established for use of electrocardiographic (ECG) monitoring during endoscopic procedures. Numerous studies have documented the occurrence of cardiac arrhythmias during endoscopy, particularly during periods of hypoxemia or tachycardia. Some hospitals require ECG monitoring for all patients undergoing sedation. Although not mandated, ECG monitoring during endoscopy is recommended in the following situations: known cardiopulmonary disease, older age, acute gastrointestinal bleeding, and prolonged procedures such as therapeutic ERCP. [1]

DRUGS FOR ENDOSCOPY

Although major therapeutic endoscopic procedures such as ERCP require sedation and analgesia, routine endoscopy and colonoscopy may be performed safely without sedation. [57], [84], [85], [86], [87], [88], [89], [90], [91], [92] and [93] Practice regarding sedation varies widely in different countries, perhaps related to cultural assumptions, socioeconomic conditions, and other factors. In the United States and the United Kingdom, sedation is widely used for colonoscopy and endoscopy [17], [20], [91]; in France, about 80% of colonoscopies are performed under general anesthesia [86]; whereas in Germany, [92] Finland, [87], [88] and Switzerland, [93] most examinations are carried out without sedation. Physicians' and nurses' assessments of patient satisfaction with sedation for endoscopic procedures correlate poorly with the patients' own assessments. [94], [95] A recent prospective study showed that both topical sprays and intravenous sedation independently improve patients' tolerance of and satisfaction with upper endoscopy. [93] Endoscopy is better tolerated by older than by younger patients, [84], [91] by men than by women, [84], [90], [91], [94] by patients without abdominal pain, [91] by patients with low anxiety scores, [90] and by patients who have had a previous endoscopy. [84] Clues to patient dissatisfaction with sedation for colonoscopy include higher education and longer procedures. [95] Particularly in those groups of patients shown least likely to tolerate upper or lower endoscopic procedures, sedation should be offered to reduce the psychological and cardiovascular stress, enhance tolerance, and improve compliance with subsequent endoscopies. [57] Patients with cardiovascular disease may require adequate sedation to prevent excessive cardiovascular stress; premedication reduces myocardial oxygen demand by attenuating the endoscopy-induced increase in blood pressure and pulse rate, resulting in a decreased rate-pressure product and decreased serum cortisol concentration. [70], [71] and [72]

Endoscopists must carefully appraise their end points for sedation. Patient requirements may be different for different procedures. For example, in upper endoscopy, the goal is usually sedation to alleviate anxiety, gagging, and cardiovascular stress; for colonoscopy, on the other hand, the primary requirement is analgesia to allow some tolerance for distention and endoscope looping. Choice of drugs should be tailored accordingly. Brief procedures such as outpatient endoscopy and most

colonoscopies can be achieved with minimal amounts of either sedation or analgesia, whereas therapeutic procedures such as complex therapeutic ERCP may require deep sedation so that the patient can remain comfortable and cooperative for a prolonged period of time. Another approach to sedation is to allow patients to administer their own sedation: several studies have shown that patient-controlled analgesia is a reasonable alternative to physician-controlled medication. [94], [96]

Topical Agents

Topical agents such as benzocaine (Cetacaine) and lidocaine are frequently sprayed in the pharynx for upper endoscopy. There is controversy regarding whether they improve patient tolerance, but two randomized, double-blind, placebo-controlled studies have shown significant decreases in patient discomfort during intubation and endoscopic examination and improved assessment of patients' tolerance by endoscopists. [93], [97] In one study, most patients indicated that they preferred to have their next endoscopy performed with topical anesthesia. [97] In contrast, another study suggested that topical anesthesia makes little difference in tolerance of the procedure. [98] It is likely that topical anesthesia will provide the most additional benefit in lightly sedated patients. In addition, gag response after topical anesthetic can be a significant predictor of patient tolerance. [99] To be effective, topical anesthesia must be delivered to the posterior pharynx and not merely to the tongue and mouth, sometimes requiring depression of the tongue with a tongue blade or finger. Systemic absorption of topically administered lidocaine has been documented, occasionally resulting in arrhythmias, seizures, and methemoglobinemia. [100] These complications are extremely rare, and in my opinion, carefully administered topical spray is a useful adjunct that may result in decreased necessity for systemic sedation.

Anticholinergics and Antispasmodics

Anticholinergics such as atropine are sometimes used to decrease oral secretions or prevent vasovagal reactions, but they are of uncertain value and have significant potential side effects. A randomized, controlled trial of atropine before colonoscopy showed no statistically significant difference with respect to number or severity of vagal episodes, and neither the endoscopist nor the patients noted any difference in ease or tolerance of the procedure. [101] Atropine may exacerbate the tachycardia that often accompanies upper endoscopy, [102] thus contributing to cardiovascular stress. A randomized, controlled study in pediatric patients found that addition of atropine during upper endoscopy provided no increase in safety or benefits but did decrease oxygen saturation. [103] It is thus recommended that atropine be used only to counteract vasovagal episodes when they occur.

Antispasmodics such as hysoscyamine butylbromide [104] and hysoscyamine sulfate [105] have been shown to be useful in premedication for colonoscopy, with better patient tolerance and easier colonoscope insertion. In contrast, antispasmodics were shown to be of no benefit for flexible sigmoidoscopy. [106] Tachycardia is a potential side effect of these medications.

Benzodiazepines

Benzodiazepines are the most widely used sedatives for gastrointestinal procedures. They are a relatively safe group of drugs with a high therapeutic index. The two drugs used most commonly in the United States and the United Kingdom are midazolam (Versed) and diazepam (Valium). The pharmacology of benzodiazepines is summarized in Table 137-3. These drugs are best titrated using intermittent intravenous injection to achieve a degree of sedation that is considered clinically adequate. Midazolam is about fourfold to fivefold more potent than diazepam on an intravenous dose basis. Numerous studies have demonstrated the equivalency of these two drugs with respect to safety. [11], [18], [20], [48] Despite a significantly longer plasma half-life, recovery time after diazepam is not significantly longer than after midazolam. [11] Potential advantages of midazolam over diazepam include increased amnestic effect and a reduction in local pain and phlebitis on injection. [107] However, other randomized trials have not consistently demonstrated superiority of midazolam over diazepam. [108], [109] and [110] With both drugs, the dosage requirement decreases markedly with increasing age, an effect that may be more pronounced with midazolam than with diazepam. [6], [111]

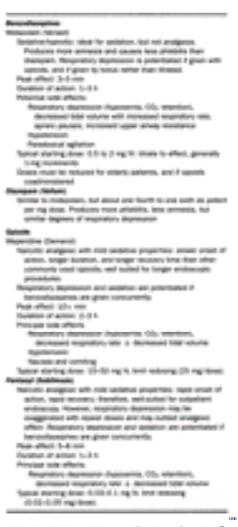

TABLE 137-3 Pharmacology of Commonly Used Sedatives and Analgesics

A starting dose of 0.5 to 2 mg of midazolam should not be exceeded. In older, sicker patients after a single intravenous dose of either drug, maximal central nervous system and respiratory depression are seen within 3 to 5 minutes; further increments, usually of 1 mg (or 0.5 mg in elderly or sensitive patients) can be titrated to effect. Risk for overdose increases significantly if an inadequate period of time (less than 2 to 3 minutes) is allowed between multiple intravenous dose administrations.

The major side effect of benzodiazepines, respiratory depression, results from blunting of central ventilatory response to CO_2 and peripheral response to hypoxemia. [44], [45], [48] Respiratory depression is usually mild when benzodiazepines are titrated carefully as sole agents [112] but is greatly potentiated by coadministration of opioids [113]; thus, doses must be reduced when narcotics are also used. Other side effects include hypotension and occasional paradoxical reactions with agitation and disorientation.

Opioids

Opioids such as meperidine and fentanyl are primarily analgesics with a mild sedative effect and are frequently combined with benzodiazepines to improve patient comfort and decrease the intensity of painful stimuli. The pharmacology of opioids is summarized in Table 137-3. The major side effect is respiratory depression, which is due to central depression of ventilatory response to CO_2 and results in CO_2 retention and hypoxemia due to decreased respiratory rate with or without decreased tidal volume. [47] Used alone, opioids improve tolerance in patients undergoing endoscopy. [71], [72] When used in combination with benzodiazepines, they may result in enhanced tolerance and even amnesia [114], [115] but greatly potentiate respiratory depression. [113] Meperidine (Demerol) is the opioid most widely used by endoscopists in the United States; fentanyl (Sublimaze) is widely used by anesthesiologists and small but increasing numbers of endoscopists. [17] There are significant differences between meperidine and the newer agents, such as fentanyl. Meperidine has a slower onset of opioid effect (5 to 10 minutes versus 2 to 5 minutes), and recovery time for meperidine may be significantly longer than with fentanyl, [115] requiring longer observation and recovery times. Meperidine might provide a slight margin of safety owing to more gradual onset of effects, including respiratory depression. [116] A disadvantage of fentanyl is cumulative respiratory depression with successive doses, despite rapid elimination from the plasma, which might result in dissociation between analgesia and respiratory depression. [47], [117] One study purported to show that meperidine was better than other opioid agents with respect to patient comfort and amnesia during colonoscopy, [115] but the doses of other opioids may not have been equivalent. Overall, fentanyl may be preferable to meperidine for outpatient endoscopy because of rapid onset and recovery. Meperidine is well suited to prolonged procedures such as extended colonoscopy or ERCP, in which gradual onset and sustained analgesia is desired.

Combination of Benzodiazepines and Opioids

Combinations of opioids and benzodiazepines are widely used in the United States [17], [18] and less often in the United Kingdom. [20] The combination of benzodiazepines and opioids results in additive or even synergistic effects. This results in improvements in the induction of anesthesia, [114] in the ability of patients to tolerate upper endoscopy from the physician's standpoint, and in the ability to complete the examination, but in little difference on the patients' reported tolerance of the procedure. [73] As already emphasized, the combination of benzodiazepines and opioids has been shown to cause significantly more hypoxemia and apnea than either class of drug used alone. [113] This phenomenon may explain the observation in two large surveys that additional use of opioids or combinations of agents has been associated with adverse cardiopulmonary outcomes of endoscopy. [12], [20] Therefore, continual vigilance in monitoring is essential when these drugs are coadministered. The doses of each class of drug need to be significantly reduced when used concurrently. It is generally advised to give the opioid first and to allow several minutes for the opioid to take effect before administering the benzodiazepine. [2], [118] The benzodiazepine should then be carefully titrated in smaller doses than when the drug is used alone.

Droperidol

Droperidol is a neuroleptic agent with antiemetic and anxiolytic properties that produces mild sedation and a sensation of indifference or detachment. Droperidol has been reported to be useful in sedating patients who are inadequately sedated with standard combinations or have paradoxical agitation. [119], [120] and [121] Droperidol does not appear to exacerbate opioid-induced respiratory depression [122] but may result in hypotension, so that automated blood pressure monitoring is advised. Droperidol is typically given in a dose of 1 to 2.5 mg, up to 5 mg. Many centers have found this agent to be useful in prolonged endoscopic procedures such as endoscopic ultrasound [120] and ERCP, [123] and in difficult-to-sedate patients, [121] particularly when the patient becomes restless or develops respiratory depression with standard doses of other medications. Randomized, controlled trials have shown that addition of droperidol to standard combinations of benzodiazepines and opioids during complex endoscopic procedures significantly improves patient- and physician-perceived tolerance of the procedure. [120], [121], [122] and [123] However, recent reports of rare serious arrhythmias have resulted in widespread restriction of its use.

Propofol

Propofol is a phenol-derivative short-acting anesthetic with a rapid onset of action. It is used widely in the anesthesia community both for the induction and maintenance of general anesthesia and for less deep stages of monitored anesthesia care. Propofol can be administered either by bolus injection or by continuous infusion. [124] There have been a number of recent reports of the use of propofol for gastrointestinal endoscopy. [125], [126], [127], [128], [129] and [130] Most of these studies have shown that compared with standard sedation with benzodiazepines and opioids, propofol results in improved tolerance of the procedure from both the physician's and the patient's point of view and in faster recovery times. However, the administration of propofol by nonanesthetists, as advocated by some endoscopists, is controversial. [8], [85], [86], [130] This agent has a very rapid onset of action, may result in unconsciousness within 30 seconds of administration, produces significant respiratory depression, [131] and acts synergistically with benzodiazepines [132] and opioids. [133] In one study, propofol administration for ERCP resulted in one case of prolonged apnea requiring mask ventilation. [133] It is notable that in virtually all of the published studies of propofol for gastrointestinal endoscopy, the drug was administered by nurse anesthetists or M.D. anesthesiologists, who, unlike gastrointestinal endoscopy unit personnel, have extensive training and experience with anesthetic administration and airway management, including emergency endotracheal intubation. Even anesthesia personnel find induction and maintenance of deep sedation with propofol without endotracheal intubation to be challenging at times. Although it is possible that gastrointestinal endoscopy unit personnel might be able to administer propofol safely in sufficiently small doses to achieve the light sedation necessary for colonoscopy or endoscopy, it is my opinion, as well as the opinion of a number of others in gastroenterology [85] and anesthesia, [130] that in general, the administration of propofol is best left to anesthetists.

Antagonists

Antagonists to both benzodiazepines and opioids (Table 137-4) are an essential part of the pharmacotherapy of sedation and should be available for immediate administration in all areas where sedation is used. They do not, however, substitute for careful administration of agonist agents and are likely to attenuate, rather than prevent, major complications.

***Flumazenil* (Romazicon)**
Reverses benzodiazepine-induced sedation smoothly, without autonomic stimulation. Effective in reversing benzodiazepine-induced component of respiratory depression.
Peak effect: 3–5 min
Duration of action: 1–2 h
Potential side effects
Resedation (more likely after high-dose benzodiazepine or combination sedation)
Seizures (only in benzodiazepine-habituated patients)
Typical dose: 0.2–0.5 mg IV for reversal of sedation; titrate to effect up to 1-mg total; 1–3 mg IV for benzodiazepine overdose
Naloxone (Narcan)
Reverses opioid-induced analgesia, CNS effects, and respiratory depression. Should not be used routinely because of high potential for untoward side effects. If given nonemergently, lowest possible doses should be used.
Peak effect: 1–2 min
Duration of action: 1–3 h (less than that of meperidine or fentanyl)
Potential side effects
Pain, agitation, nausea and vomiting
Tachycardia, arrythmias, sudden death (primarily in cardiac patients due to catecholamine release)
Pulmonary edema
Withdrawal syndrome in opioid-habituated patients
Typical dose: 0.04 mg (1/10 amp) IV for reversal of analgesia/sedation, titrate to effect 0.4 (1 amp) for narcotic overdose and respiratory arrest

TABLE 137-4 Reversal Agents

Flumazenil (Romazicon) is a benzodiazepine receptor antagonist (see Table 137-4). When given in adequate doses (0.2 to 1 mg), flumazenil reverses benzodiazepine-induced sedation within a few minutes, with minimal side effects. Earlier patient discharges are frequently possible after administration of this drug. [134], [135], [136] and [137] Flumazenil may also be useful to reverse paradoxical reactions to benzodiazepines. [138] There is a potential for resedation because its duration of action is significantly shorter than that of either midazolam or diazepam. In clinical practice, this is rarely a problem unless large doses of benzodiazepine or additional agents have been used. [44], [135] If respiratory depression or deep sedation occurred before administration of flumazenil, the patient should be monitored for extended periods of time (up to 2 hours). Reversal of ventilatory depression due to benzodiazepines is variable and may be delayed, [45], [135] especially if there has been coadministration of an opioid. [139] In patients given combinations of benzodiazepines and opioids, flumazenil effectively reverses the benzodiazepine component of ventilatory depression. [140] Flumazenil may provoke withdrawal or occasionally seizures in patients habituated to benzodiazepines.

Naloxone (Narcan) is the most widely used, and perhaps abused, opioid antagonist. In one large data base study, it was used in 8% to 14% of all endoscopies. [18] Although naloxone can effectively reverse respiratory and central nervous system effects of opioids, the degree and extent of reversal is dose dependent. [47] Naloxone has rapid elimination kinetics such that its duration of effect can be substantially less than that of the opioid, resulting in renarcotization. The major adverse effect of narcotic antagonists is that patients can experience severe pain and catecholamine release, resulting in increased risk for tachyarrhythmias and cardiovascular adverse events, particularly in patients with underlying ischemic heart disease. Unexpected sudden deaths have been reported in healthy patients receiving routine reversal of opioid anesthesia with naloxone. [47] Patients habituated to narcotics may experience severe withdrawal reactions. Therefore, routine reversal of opioid analgesia should be avoided. In a semi-elective situation, small doses (i.e., 0.04 mg, or 1/10 ampule) should be administered and repeat doses titrated to effect; a full ampule, or 0.4 mg, of naloxone should be reserved for emergencies such as respiratory arrest.

Overall Approach to Sedation and Monitoring

It is important to establish a rapport with the patient and explain the process of conscious sedation (see Table 137-2). This may help determine whether the patient

will be likely to cooperate and determine the level of sedation required. The type of procedure should influence the type and depth level of sedation; in general, diagnostic endoscopy usually requires mild sedation; colonoscopy relatively more analgesia; and therapeutic ERCP the most prolonged and deepest sedation to ensure cooperation. The patient's underlying illness and risk factors, particularly cardiopulmonary disease, should be assessed. Before administering medication, baseline vital signs, respiratory rate, and SpO_2 should be noted. Medication should be given in small increments. Clinical observation of the patients' response is a key in determining the subsequent titration of medication. If a combination of a benzodiazepine and an opioid is to be used, it is recommended to begin with an opioid. Before giving the next dose of medication, the patient should be observed for at least 2 minutes (and much longer between meperidine doses) to determine its effect. Whether or not an opioid is given, benzodiazepines should be titrated in small doses.

Prophylactic oxygen should be administered to those patients at higher risk, such as patients with known coronary artery disease or very elderly patients. Before administering further medication, it is useful to observe not only the patient but also the pulse oximeter; even alert and responsive patients may have significant respiratory depression, and on room air, the oximeter can be used as an early warning device. If oxygen saturation begins to decrease, regardless of the patient's appearance, further medication should be delayed. The patient can be stimulated verbally or if necessary physically to encourage breathing. If an SpO_2 below 90% persists more than transiently, nasal oxygen should generally be administered. After administering nasal oxygen, SpO_2 usually returns to baseline or above; further administration can be titrated carefully as needed, with the realization that this patient has declared a sensitivity with respect to respiratory depression from the medications administered. If after 30 to 60 seconds, nasal oxygen does not reverse hypoxemia or hypoxemia worsens, verbal or physical stimulation should be performed, followed by airway assessment, and assisted ventilation or administration of a reversal agent should be considered. If significant desaturation occurs while the patient is already on oxygen supplementation, then special caution must be taken. Even transient desaturation in patients on oxygen supplementation may indicate severe CO_2 retention, and further sedation should be limited or withheld. [21], [22] Such transient desaturation may be the only warning of an impending respiratory arrest.

In all cases after a procedure, patients should be carefully monitored, both clinically and with pulse oximetry and blood pressure monitoring, until they are responsive, oxygen saturation is persistently greater than 90% while breathing room air, and vital signs are stable. It is particularly important to continue monitoring of the patient into the recovery area because maximum hypoventilation and hypoxemia may occur after the procedure is completed.

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CHAPTER 138

Guido N. J. Tytgat

UPPER GASTROINTESTINAL ENDOSCOPY

This chapter reviews the current status of upper endoscopy employed for diagnosis and treatment, and it discusses the use of flexible endoscopes with fiberoptic or video systems. Diagnostic considerations include when to use endoscopy, in which situations it is useful to the patient, how to use biopsy and cytology, and some new approaches to diagnosis. [1], [2] and [3]

In a section that introduces the technical aspects of endoscopy, followed by an analysis of indications, limitations, and pitfalls of upper intestinal endoscopy, some areas of interest are highlighted: dysphagia, gastroesophageal reflux disease, dyspepsia, peptic ulcer disease, upper gastrointestinal bleeding, and upper gastrointestinal neoplasia. This chapter also addresses the risks and contraindications of endoscopy to help the reader understand the cost-benefit analysis of a procedure compared with alternative diagnostic or therapeutic approaches. The final section considers speculations about endoscopy trends in the future and improvements in diagnostic and therapeutic modalities.

TECHNICAL CONSIDERATIONS

A variety of factors are necessary for safe and effective endoscopy. These include properly trained endoscopists; well-trained and clinically experienced assistants; high-quality, well-maintained endoscopic equipment; and a well-designed endoscopy suite containing all the necessary facilities and accessories for endoscopy and for emergency procedures that may occur during endoscopy.

It is mandatory to have equipment that goes to the bedside for endoscopy, especially for acutely ill patients in the intensive care unit. Adequate storage is essential for the many endoscopic accessories. There must be an area for endoscope disinfection and preparation of accessories for sterilization. It is critical to have an area in which to watch patients after endoscopy, especially if they have received sedation or have serious medical illnesses.

Performance of electrocautery requires state-of-the-art electrocautery units. All electric devices should be checked periodically for electrical safety by the hospital electrical safety team. Equipment to document the visual appearance of lesions is also strongly advised.

Fiberoptic Endoscopes

The critical part of a flexible fiberoptic endoscope is the fiber bundle that transmits a coherent image. The imaging bundle consists of several thousand thin glass fibers, 6 to 12 μm in diameter, that transmit light. Each fiber consists of an inner core of glass surrounded by an outer glass layer made of a second type of glass with a lower refractive index. Light entering the fiber reflects off the junction between these two types of glass and propagates down the fiber. A total of 10,000 to 40,000 fibers are grouped together to form the imaging bundle and generate a coherent image. The resolution of the imaging bundle depends on the diameter of the fibers. Fibers may break with excessive bending and torquing or during withdrawal of an accessory, particularly if the tip of the endoscope is flexed. A broken fiber appears as a black dot in the visual image.

The small image at the proximal or viewer's end of the coherent fiber bundle is enlarged with a 15× to 30× magnifying lens in the endoscope ocular. The image cannot be overly magnified because this would make individual fibers visible, interfering with image interpretation. Standard flexible fiberoptic endoscopes have a depth of focus that allows examination of objects 3 to 100 mm away from the tip. The field of view of the imaging system is up to 110 or 120 degrees. Special

high-magnification endoscopes are available.

Endoscope design includes the objective lens system, which focuses the target onto the fiberoptic image bundle; the light source, which illuminates the target; a nozzle for air insufflation; a nozzle for a water jet to clean the viewing objective lens; and a suction channel, which is used to remove gas and fluids and also to pass a variety of accessories by way of the endoscope into the intestinal lumen. Side-viewing endoscopes contain these same features but have a different design that allows the examiner to look sideways; the suction or accessory channel has an elevator, which can change the position of an accessory.

The endoscope control handle contains valves for air and water as well as suction and control knobs. These knobs are attached to wires that go down the length of the flexible insertion tube to the distal bending tip. By maneuvering these wires in an up-and-down or right-and-left direction, it is possible to turn the tip of the endoscope and inspect the entire lumen of the organ being evaluated. In most endoscopes, it is possible to turn the tip of the bending section more than 100 degrees (retroflex) to allow the inspection of surfaces such as the cardia of the stomach, which are difficult to see with the instrument in a straight position.

Endoscopes for the upper intestinal tract are designed to be torque stable. Clockwise or counterclockwise rotation of the control handle results in the same rotation of the tip. The flexibility of the insertion tube varies; often, it is more flexible distally and less flexible proximally.

Videoendoscopes

Videoendoscopy, introduced in the mid-1980s, has changed the field of endoscopy. Videoendoscopy is now used almost universally. The image is generated electronically using a charge-coupled device (CCD) located in the tip of the endoscope. These devices are about 3 mm in diameter.

The first videoendoscopes used a color wheel. Green, red, or blue light was sequentially sent down the illumination bundle of the endoscope and activated the CCD at the tip. It was possible to reconstruct a color image using the three sets of images generated by the colored lights. The videoprocessor displayed a full-color image of the gastrointestinal tract lining. Later videoendoscopes use a color chip that actually obtains the image in color on the tip of the endoscope. These devices use 30,000 to 850,000 pixels of resolution. [4] By incorporating high-pixel-density CCDs, high-resolution endoscopes provide greater mucosal detail. High-resolution endoscopes are capable of discriminating objects 10 to 70 µm in diameter, compared with the naked eye, which is capable of discriminating objects 125 to 165 µm in diameter. Other than the method of delivering the image, the videoendoscope is similar to a fiberoptic endoscope, with the same controls for air, water, and suction; knobs for up-and-down and right-and-left bending of the tip; and a suction channel to remove gas and fluid and to pass a variety of accessories to the tip of the endoscope. A therapeutic gastroscope with a 6-mm accessory channel has been designed for the removal of blood clots and debris. [5] There are also buttons on the videoendoscope control handle to activate videotaping, freeze the image, and obtain a photograph.

There are several advantages inherent in videoendoscopy: the control unit is located away from the endoscopist's face, reducing the chance of contamination by way of splatter from the suction port; everyone present in the room can watch the monitor; the image can be sent outside the room, and multiple television sets can be used for simultaneous observation throughout the hospital, elsewhere in the city, or even by way of satellite to another country; for complicated procedures, the assistants can better assist the primary endoscopist because they can watch the procedure on the monitor simultaneously; and images can be easily documented and stored for teaching or clinical follow-up. Digital images can now be recorded on magneto-optical disks. These images can be recalled from a central image storage system and sent to any location in the endoscopy service. The disks can be used for image processing and for management and reliable storage of endoscopic images and information. [6], [7] The video system is not sensitive to irradiation and is less sensitive to damage caused by bending and torquing than are fiberoptic imaging systems.

Passing the Endoscope

There are two methods of passing an endoscope perorally: the blind method and the direct vision method. The blind method involves passing the tip of the endoscope into the patient's mouth, through a bite guard if necessary, and advancing the tip of the endoscope to about 18 to 20 cm from the incisor teeth. The patient is asked to swallow, which opens the cricopharyngeal sphincter, and the tip of the endoscope is gently advanced into the proximal esophagus. This may be performed by simply bending the tip of the endoscope without placing one or two fingers in the patient's mouth to guide the tip. In some circumstances, one or two fingers may be used to keep the tip of the endoscope midline and guide the endoscope as it is advanced. Care should be taken to avoid a finger bite. The direct vision technique involves passing the endoscope through a bite guard and watching endoscopically to observe the anatomy of the hypopharynx as the tip of the instrument is advanced in the midline into the direction of the closed cricopharyngeal sphincter. The patient is asked to swallow, and under direct vision, the tip of the instrument is passed away from the epiglottis and larynx into the proximal esophagus. This technique is in general more advisable but is especially important in patients with distortion of the hypopharyngeal anatomy (e.g., tumor, prior surgery) or with a suspected proximal diverticulum, in whom blind passage might cause inadvertent injury to the proximal esophagus or perforation of the diverticulum. The direct vision technique also allows an inspection of the hypopharyngeal anatomy. Detailed knowledge of the hypopharyngeal anatomy is mandatory to identify both pyriform sinuses lining the entry to the esophagus. Small-diameter endoscopes (5.3-mm fiberoptic or 6-mm videoendoscopic outer diameter) can also be passed transnasally for unsedated esophagogastroduodenoscopy. [8] Such instruments have a 2-mm accessory channel and are occasionally used for screening office endoscopy.

Most experienced endoscopists prefer to hold the endoscopic controls (e.g., air, water, and suction valves; tip direction wheels) in the left hand. The right hand is used to advance the instrument and intermittently to come back to the endoscope control to turn the control wheels. Torquing the endoscope is accomplished by rotating the instrument control handle, which results in rotation of the entire shaft and tip of the endoscope.

Endoscopic Examination

The endoscopic examination is carried out under direct vision. The esophagus is studied from 20 to 40 cm. At 40 cm lies the ora serrata, which is the junction between the pearly stratified squamous mucosa and the redder gastric columnar epithelium. It is also possible to identify the level of the diaphragm by having the patient sniff. This is important for identification of a hiatal hernia. The esophageal folds can be seen to change with air distention. It is possible to recognize extrinsic pressure on the esophagus from adjacent structures such as the aorta and the left main-stem bronchus.

With slight angulation to the left and anteriorly, the endoscope is passed into the stomach. Air is insufflated to facilitate gastroscopy. The fluid in the gastric pool is aspirated to improve endoscopic inspection and to reduce the likelihood of regurgitation and aspiration during the procedure. It is important to notice the presence of food, bile, or blood in the gastric fluid pool. The gastric mucosa is inspected, and observations are made about the amount of mucus, mucosal appearance, erythema, defects, fold thickness and pliability, and the appearance of blood vessels. Normally, it is not possible to see blood vessels except when the stomach is overdistended with air. The gastric folds begin in the upper portion of the stomach and extend down to the entrance into the antrum. With gentle distention of the stomach, these folds often flatten.

Endoscopy of the stomach is not complete without a careful retroflexion examination in which the tip of the endoscope is bent more than 180 degrees. This retroflexion, together with endoscopic rotation, allows inspection of the lesser curvature, cardia, and fundus. A lesion such as a tumor, Mallory-Weiss tear, or ulcer is often not seen in these areas unless the endoscope is retroflexed. Retroflexion also permits inspection of a hiatal hernia. After retroflexion, the instrument can be rotated clockwise and counterclockwise and gently moved in and out to inspect the entire area of the cardia, lesser curvature, and fundus.

The incisura angularis is a fold that marks the entrance to the antrum. The fold is located on the lesser curvature of the stomach. The mucosa in the antrum does not have rugal folds. Peristalsis begins in the midbody and progresses downward; the frequency is about three contractions per minute. These contractions progress to the pylorus and stop. The pylorus is usually open at rest, but it closes as the contraction wave reaches it. It is useful to watch several contraction waves as they progress toward the pylorus to determine whether there is an area of wall that is less pliable, suggesting an inflammatory or neoplastic infiltration. Peristalsis may be weak or absent in a heavily sedated patient.

After thorough gastric inspection, the tip of the endoscope is placed through the pylorus into the duodenal bulb. It is sometimes necessary to create a loop along the greater curvature before the tip of the endoscope can progress to and through the pylorus. With most end-viewing endoscopes, the entire surface of the duodenal bulb can be inspected. With a small-caliber endoscope, it is possible to inspect the fornices. The endoscope is passed beyond the apex of the duodenal bulb into the second descending portion of the duodenum. Passage from the tip of the bulb into the descending duodenum is often possible under direct vision but occasionally requires looking to the right and down and advancing the instrument gently, even though it is not possible to see the lumen until the tip of the instrument enters the descending duodenum. The duodenal bulb is usually free of folds; duodenal Kerckring folds begin in the descending portion of the duodenum. The endoscope is withdrawn, inspecting the mucosa of the duodenum, stomach, and esophagus. With a cooperative patient, it is often possible to inspect the larynx and hypopharynx

as the instrument is withdrawn.

In transnasal esophagogastroduodenoscopy, a small-caliber endoscope is passed through the most patent side of the nasal cavity after local application of lidocaine gel and a nasal decongestant. After inspection of the pharynx, the scope is introduced into the esophagus after traversing the upper esophageal sphincter under direct vision. Following evaluation of the esophagus, the stomach and duodenum are entered and evaluated. [9]

ACCESSORIES AND METHODS FOR SPECIAL APPLICATIONS

During endoscopy, to increase diagnostic information beyond the visual appearance, a variety of endoscopic accessories can be passed down the biopsy channel. These include measuring devices, biopsy forceps, cytology brushes, needles for cytologic puncture or injection therapy, Doppler probes, probes to obtain an ultrasound image, and probes capable of measuring electrical activity or the pH of fluid at the tip of the endoscope.

Biopsy Forceps

Although all biopsy forceps are similar, they vary in diameter, in whether a central spike is present to impale the tissue and thereby improve the sample size, and in whether the cups are closed or fenestrated [10] (Fig. 138-1). Gentle manipulation is required because excessive force on the control handle can break the forceps. As with other endoscopic accessories, it is important to ensure that the forceps are functioning properly before use. Forceps are available for single or multiple biopsies. Forceps are reusable or disposable.

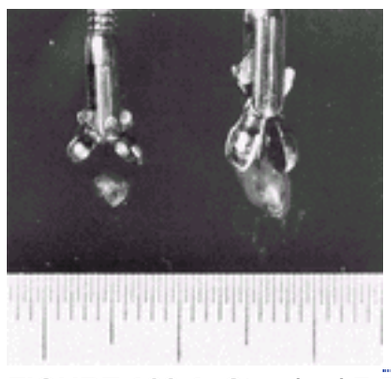

FIGURE 138-1. Standard 7-French (2.3-mm) and large-caliber biopsy forceps.

After obtaining the biopsy specimen, the tissue is removed from the cup, often using the tip of a toothpick. It is possible to orient this material on a mesh to improve the diagnostic quality of the specimen. The larger biopsy forceps are useful to obtain a larger surface area and volume specimen, which may be helpful for diagnosis in gastric lymphoma or linitis plastica.

Most endoscopic forceps obtain mucosa and, occasionally, a small amount of submucosa. To sample more deeply into a lesion such as a submucosal tumor, needle cytology may be applied, or repetitive sampling with a large biopsy forceps should be made in the same area. Another technique for obtaining larger samples involves turning the endoscope toward the mucosa and suctioning the mucosa into the open biopsy forceps to impale the tissue. This turn and suction or aspiration biopsy technique provides a more adequate size of tissue sample. [11]

Alternatively, a large-particle biopsy may be obtained using a polypectomy snare. The snare is placed over the fold or polypoid area, tightened, and gently pulled away from the wall. Prior injection of saline or diluted adrenaline in the submucosa may lift the mucosa to facilitate snaring and to prevent deep mural damage. As the snare is pulled through the lesion, the electrosurgical current causes coagulation and prevents bleeding (Fig. 138-2).

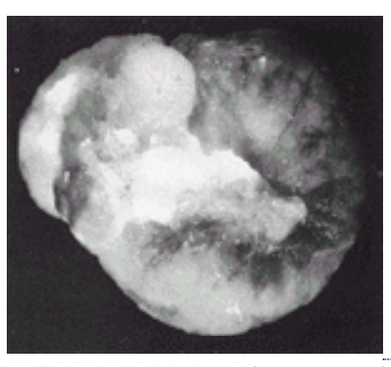

FIGURE 138-2. A large-particle biopsy specimen of a large gastric erosion was taken with an electrosurgical snare.

The lift-and-cut biopsy technique uses an endoscope with two channels to pass a forceps and a snare. The snare is opened, and the forceps is directed through the snare. The tissue is grasped by the forceps and pulled up into the snare. The snare can be tightened, and the tissue can be excised with electrocautery. This technique may result in unexpectedly large, deep biopsy specimens and is not recommended for the inexperienced endoscopist.

Tissue may also become available for histopathological examination through endoscopic mucosal resection (EMR). [12], [13] Most EMR techniques incorporate a submucosal injection to separate the lesion from the muscularis propria. Saline with or without epinephrine is commonly employed. EMR may be carried out using an endoscope fitted with a straight or oblique rimmed transparent cap. A polypectomy snare is passed down the accessory channel and opened inside the rimmed cap. The lesion is suctioned into the cap and the snare tightened around the lesion. The snare is then advanced out of the cap so that the extent of the captured mucosa can be confirmed before transection.

Cytology Brushes

Cytology brushes are used to obtain cell samples from the surface of a lesion for cytologic study or for examination of an exudate to detect, for example, the presence of *Candida*. These brushes are often protected by a plastic overtube or sheath, which allows the examiner to advance the brush out of and pull it back into the sheath. The sheath prevents loss of material and oropharyngeal contamination of the specimen because it covers the brush as it is removed. After removal of the cytology brush from the endoscope biopsy channel, the mucus and fluid are placed on a glass slide and into Carnoy solution or another appropriate fixative solution. [14] These brushes are disposable or can be sterilized. Cytology brushing is especially important in areas that are inaccessible for biopsy, such as narrow strictures in the esophagus.

Material can be aspirated for cytologic examination using an endoscopic needle technique. A 4-mm long, 23-gauge sclerotherapy needle can be placed into the target, such as a submucosal tumor or an ulcerating malignancy, to obtain cellular material from below the mucosa or below the surface exudate, adding to the diagnostic yield of biopsy and brush cytology. [15]

Measuring Devices

Occasionally, it may be important to know the actual size of a target. Endoscopic estimation is often inaccurate. The closer the tip of the endoscope is to the target, the larger the target appears. Measuring methods include using open biopsy forceps with a known distance between the tips of the biopsy cups, using a spring-type measuring probe with gradations of known length, and using a projected grid obtained by employing an argon laser that allows the examiner to see the contour of the target. When image processing is used to correct for distortion, a significant decrease in error can be obtained in vivo. [16]

Measuring Mucosal Blood Flow

Several devices can be passed down the accessory channel to study blood flow and distribution. Doppler ultrasound probes (Fig. 138-3) can detect the flow in blood vessels below the mucosa, such as varices, or in an artery in the base of an ulcer. Doppler devices can be used before and after endoscopic hemostatic therapy, such as injection or thermal therapy for an artery in the base of an ulcer. The Doppler probes have a circumferential radiating pattern (i.e., Doppler signal) and have a controllable depth of acoustic interrogation. [17], [18], [19] and [20] It is also possible to use a laser Doppler device to measure mucosal red blood cell velocity, a parameter thought to correlate with mucosal blood flow. The acoustic and laser techniques require the probe to touch the target mucosa. [21], [22]

FIGURE 138-3. A Doppler ultrasound probe is passed through the instrumentation channel of a side-viewing endoscope.

Reflectance spectrophotoscopy can also be achieved by passing a probe to touch the mucosa gently and measure mucosal oxygen saturation and hemoglobin concentration. [23] Videoendoscopes can be used to calculate the red-to-green signal ratio and estimate mucosal blood hemoglobin. [24] This method assumes that hemoglobin is the predominant light-absorbing pigment in the mucosa and that the degree of absorption of light is proportional to the amount of hemoglobin present.

Endoscopic Ultrasonography

Endoscopists are limited to inspection of the mucosal surface. By combining endoscopy with ultrasound, it is possible to look below the surface at lesions of the intestinal wall and at lesions involving adjacent structures. There are two approaches to this combination of technologies. In the first approach, the endoscope has an ultrasound device built into the tip of the endoscope. In the second approach, an endoscopic catheter is passed down the biopsy channel of the endoscope (see Fig 138-3). Puncture of tissue using ultrasound guidance allows histological and cytologic samples to be obtained from wall abnormalities or possible metastases to adjacent lymph nodes and various therapeutic interventions. [25], [26], [27] and [28] Endoscopic ultrasonography is discussed in Chapter 154.

Chromoscopy

Dye scattering (i.e., chromoendoscopy) is an in vivo technique in which dye is scattered on the mucosa to study and outline the fine detail of the target structure or to enhance the ability to detect a particular lesion (contrast or relief staining endoscopes) or in which the dye is absorbed by certain cell types (vital staining). [29], [30] and [31] The dye solution can be swallowed by the patient before endoscopy, or the dye can be sprayed against the mucosa using a catheter passed down the biopsy channel of the endoscope. The dyes most often reported to be useful are 1% to 3% Lugol solution, 1% to 2% methylene blue, 0.2% to 0.3% indigo-carmine, 1% to 2% toluidine blue, and 0.3% Congo red. Before using any of these dyes, it is necessary to reduce mucus. This is often accomplished using a solution of dimethyl polysiloxane, sodium bicarbonate, and pronase. A 10% solution of *N*-acetylcysteine can also be used before spraying the dye. [32]

Contrast Chromoendoscopy Dyes can be used to accentuate minute alterations in mucosal architecture by accumulating in normal or abnormal mucosal structures, indentations, crevices, or irregularities and accentuating small lesions. This is best achieved using indigo-carmine or methylene blue (Fig. 138-4; see also Color Fig. 138-4).

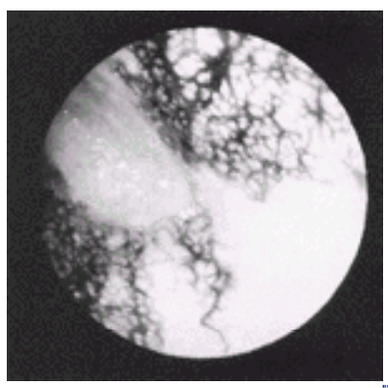

FIGURE 138-4. (See Color Fig. 138-4.) Gluten enteropathy with total villous atrophy. Mosaic appearance of the mucosal lining, accentuated by methylene blue entering deep mucosal crevices.

Vital Staining The dye is absorbed by epithelial cells but may also penetrate necrotic tissue. One or 2 minutes after spraying, the mucosa is washed with water, and the surface can be observed. Lugol solution stains intracellular glycogen in nonkeratinized squamous epithelium. Metaplastic columnar epithelium in the esophagus or injured squamous mucosa does not stain. Esophageal cancer and high-grade dysplasia do not stain with Lugol solution. Methylene blue dye when used for vital staining detects intestinal metaplasia, but not squamous epithelium, in the esophagus or intestinal metaplasia in the stomach. Congo red stains the acid-producing parietal cell mucosa; after the dye is acidified, it turns from red to dark blue. Because toluidine blue stains nuclear DNA more strongly than cytoplasmic RNA, dysplastic or neoplastic cells stain blue. Lugol staining is useful in looking for plaques overlying varices, studying the esophageal mucosa in patients with cancer of the head and neck and in those who are alcoholics and smokers, delineating neoplasms of the esophagus, and excluding a synchronous lesion or an area of dysplasia. Vital staining in the stomach may be useful in a nonhealing ulcer if early gastric cancer is suspected or to study an area of mucosal abnormality such as scarring or discoloration of the mucosal surface. Vital staining may help to detect or exclude areas of dysplasia or malignancy.

Removal of Foreign Bodies

In removing a foreign body, the endoscopist should ensure that the airway is not obstructed and should be certain that the foreign body does not injure the lining of the gastrointestinal tract. During removal of a foreign body, an overtube provides a degree of protection of the airway by preventing dropping of the foreign body during removal, but the only way to ensure complete airway patency is to use a cuffed endotracheal tube.

The mucosa can be protected, especially in the removal of sharp objects, by using an overtube to protect the intestinal lining during removal of a sharp foreign body. It is possible to remove foreign bodies with rigid and flexible endoscopes. Foreign bodies in the area of the hypopharynx and very proximal esophagus are often best removed with an open laryngoscope and grasping forceps. For other foreign bodies, flexible endoscopes are used, preferably with a large instrumentation channel (2.8 to 3.5 mm). The larger channel allows a variety of instruments to be used to grasp and hold the foreign body. These include baskets of the Dormia type, forceps for grasping the foreign body, and polypectomy snares. Alligator forceps with teeth are useful for grasping coins and other types of smooth objects. If an object is

irregular or soft, other devices, such as baskets, snares, and graspers with three prongs, can be used. [33] Experienced clinicians know that flexible endoscopes cannot remove all foreign bodies safely. An irregular, sharp object, an object with a ragged surface, or an object that is impacted in the wall (e.g., dental plate) may require a rigid laryngoscope or esophagoscope to be passed under general anesthesia for removal of the foreign body.

If the foreign body is sharp, a plastic overtube can be used on the endoscope [34] (Fig. 138-5). The endoscope can be withdrawn into the overtube while grasping the foreign body, which protects the mucosa from injury by the sharp foreign body as it is being removed. This also eliminates the possibility of dropping the foreign body in the hypopharynx, causing pulmonary aspiration of the object. The overtube is useful if the endoscope must be passed several times for removal of a foreign body, such as a piece of meat. An alternative to a plastic overtube is a simple latex protector hood fastened to the tip of the endoscope. [35]

FIGURE 138-5. A plastic overtube around the endoscope allows safe retrieval of sharp or pointed foreign bodies.

Most experienced endoscopists practice before attempting foreign body removal. A foreign body similar to that swallowed by the patient is manipulated with the type of instrument available to grasp the foreign body during endoscopy. This time spent in forethought and planning makes the foreign body removal safer and easier. [36]

Endoscopic Control of Upper Gastrointestinal Bleeding

Upper gastrointestinal bleeding is a commonly encountered problem in clinical medicine. For about 85% to 95% of these patients, a precise diagnosis of the cause of the bleeding can be obtained. For a few patients with massive hemorrhage, emergency surgery may be required. In most patients with bleeding, endoscopy can be performed as soon as the patient is stabilized hemodynamically. Endoscopic treatment of nonvariceal and variceal bleeding is discussed in Chapter 147 and Chapter 148.

Polypectomy

Polypectomy is accomplished using a wire snare placed over the polyp (Fig. 138-6) and tightened at the base of the polyp. Snares of a variety of sizes and shapes are available, including a symmetric ellipse, a hexagon, and a slightly hooked configuration. After the handle is closed, the tip of the snare should retract at least 1 cm into the plastic overtube. As the snare is tightened on the lesion, an electrocoagulative current is applied, alone or in combination with a cutting current, during the transection of the polyp.

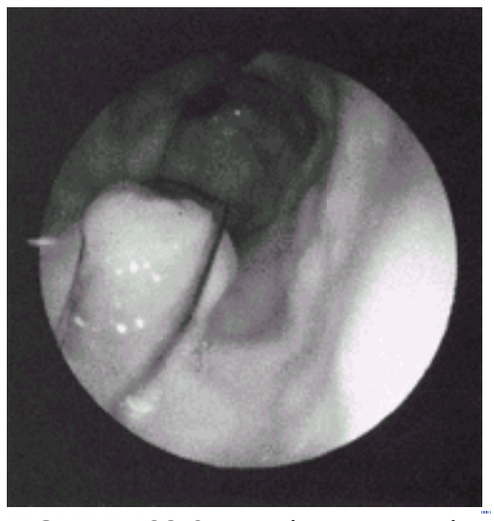

FIGURE 138-6. In polypectomy, the snare loop is positioned around the sessile, adenomatous, gastric polyp.

After the loop has been passed over the polyp, it may be helpful to advance the plastic tube around the wire snare to the spot on the polyp at which the snare will be closed. Thereby, it is possible to control where the transection occurs as the loop is closed. As polypectomy is accomplished, the endoscopist can see a whitish discoloration on both sides of the loop, which indicates that coagulation is taking place.

The complications with gastric polypectomy include hemorrhage and perforation. Because of the thickness of the gastric wall, perforation is unusual, but hemorrhage may be a significant problem. [37] The risk for bleeding is probably greater in large polyps with thick stalks. Injection of the stalk with epinephrine is recommended but may not always prevent bleeding. Mechanical hemostatic devices such as endoloops [38] or clips [39] can prevent bleeding after polypectomy.

Mucosal Resection

Removal of a focus of high-grade dysplasia or early malignancy in the upper intestinal tract through mucosal resection is increasingly performed. The technique is schematically illustrated in Figure 138-7. Usually, the lesion is first examined with endosonography to prove its superficial nature. In principle, the lesion should be limited to the mucosal layer. After proper localization with chromoscopy or fluorescence endoscopy, the lesion is lifted after submucosal injection of saline or diluted epinephrine. Thereafter, the lesion is aspirated, usually into a cap-fitted endoscope or a specially designed overtube, allowing positioning of a polypectomy snare around the aspirated tissue fragment. Transection follows with electrocoagulation, allowing the specimen to be retrieved for histopathological examination. [2], [40], [41], [42], [43] and [44]

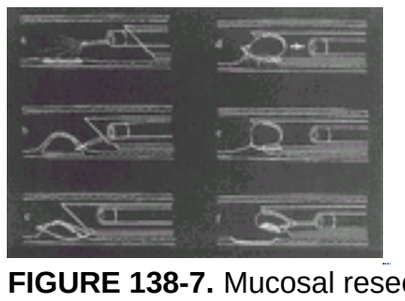

FIGURE 138-7. Mucosal resection of superficial malignancy. **A:** Dye spraying to delineate the lesion. **B:** Lifting the lesion by submucosal injection. **C:** Positioning of the snare. **D:** Aspiration of the polyp-like lesion in overtube. **E:** Transection of the lesion. **F:** Grasping of the transected lesion. (From ref. [40].)

Dilation

Flexible endoscopy is playing an increasingly important role in dilating strictures. Endoscopically passed guidewires are used to guide the tip of the dilating device, especially in treating diverticula, eccentric strictures, tight or tortuous strictures (Fig. 138-8; see also Color Fig. 138-8), and angulated strictures, such as those formed after surgical anastomosis. [45]

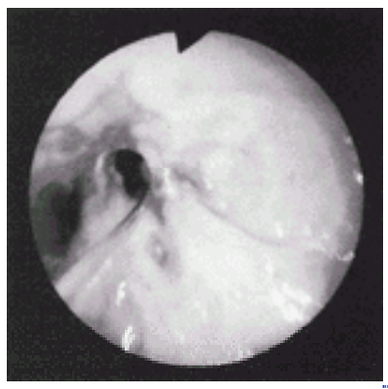

FIGURE 138-8. (See Color Fig. 138-8.) A guidewire has been passed through this eccentric, reflux-induced stricture.

The guidewire with a spring tip is passed by way of the endoscope. If the small-caliber endoscope can be passed through the stricture, the wire can be positioned under direct vision below the narrowed area. If it is not possible to pass the endoscope through the stricture, the wire can be passed under direct endoscopic vision in the stricture if necessary with further fluoroscopic monitoring to ensure that the wire is correctly placed. The guidewire and the spring tip should remain below the area to be dilated throughout the procedure. Several different types of dilators are available. It is also possible to pass catheters with balloon tips through the biopsy channel of an endoscope to dilate strictures. [46] These balloons vary from 4 to 20 mm in diameter when fully inflated. Some balloon catheters have a central channel that can be passed over a guidewire.

Pyloric stenosis is often dilated with through-the-scope balloons that are 3 cm long and have diameters from 9.9 to 17.8 mm (30 to 54 French). These balloons are best passed through a large-caliber instrumentation channel. It is mandatory to monitor passage of the balloon and its inflation with fluoroscopy to be certain that the waist of the balloon, representing the stricture, has been obliterated. [47]

Stents

In some patients with cancer of the esophagus, nonsurgical palliation with stents may be the preferred method of treatment (Fig. 138-9). Endoscopy is critically important to diagnose the malignancy, evaluate the degree of narrowing, measure the length of the tumor narrowing, and determine the distance from the top of the tumor to the incisor teeth, an important factor in placing the prosthesis.

FIGURE 138-9. Esophageal carcinoma before (**A**) and after (**B**) stenting with a Tygon endoprosthesis.

An endoscope usually cannot be passed through the lumen of a patient presenting with an obstructing tumor. In this circumstance, a guidewire is passed through the instrumentation channel under endoscopic guidance. Fluoroscopy is used to follow the tip of the guidewire until it enters the stomach and is positioned along the greater curvature. The endoscope is removed, and the guidewire can be used to dilate the malignant narrowing with metal olives, balloons, or bougies. After the length of the stricture is determined, the prosthesis length should be long enough to include 2.5 to 3 cm proximal to the narrowing and the same distance distal to the narrowing to prevent early restenosis by tumor.

Stents that can be placed in the esophagus include nonexpandable and expandable devices. Nonexpandable stents are inserted using an introducing device, which can be a balloon, a bougie, a Key-Med introducer, or a small-diameter endoscope. [2], [48] Expandable stents are increasingly the therapy of choice, and several covered and noncovered self-expanding metal stents (e.g., Wall, Gianturco, Ultraflex, Song) are commercially available. [49], [50] These devices are passed over a guidewire through the malignant narrowing, within a sheath covering the compressed stent. After the constraining sheath is released, the stent is left in place and expands. The expanding stents have the advantage of producing a wide lumen through the stenosing tumor mass without having to dilate the tumor before placement. However, these stents are difficult to remove or not removable. Stents covered by a silicone membrane may prevent tumor ingrowth.

Laser Tumor Ablation

An endoscope can be used to guide an Nd:YAG laser to open a lumen obstructed by tumor. It is preferable to start coagulating at the distal portion of the tumor so that this tissue can slough and pass down the gastrointestinal tract. This may require dilating the tumor before laser therapy. [51], [52] If it is not possible to dilate the tumor, it may be necessary to begin laser coagulation at the proximal margin. This is more difficult because it may not be clear where the lumen is, and tumorous tissue edema may impair passage of the endoscope to the more distal portions of the tumor. In general, the laser is applied in a circumferential pattern for a tumor that extends around the circumference of the esophagus. To avoid cavitation, the laser beam should not be focused on one spot for a long time. Treated areas appear necrotic and yellow-white. If the tumor does not slough spontaneously, this tissue is gently removed before further treatment with the laser.

An alternative method of applying laser photocoagulation is with contact laser probes, which use lower power. These probes have a tip made of ceramic or sapphire attached to the tip of the quartz fiber delivery system. For small tumors of the esophagus and stomach, photodynamic therapy may be used. A photosensitizing agent, such as a hematoporphyrin derivative, is injected intravenously and is taken up in high concentration in the area of the tumor. A light is used to activate the agent by

selecting an appropriate wavelength for the particular drug. This produces cytotoxic singlet oxygen, resulting in necrosis of the tumor. [53], [54]

Local Irradiation Tumor Therapy

Cesium or iridium can be used to treat a tumor using intracavitary radiotherapy. [55] This therapy requires precise positioning of the applicator into the area of the tumor. Endoscopy is used to place a guidewire, over which the applicator is passed. Fluoroscopy documents the exact locations of guidewire, applicator, and tumor. The applicator probe is fixed in position by attachment to a facemask. The patient is transferred to the treatment area, and the position of the applicator is confirmed by fluoroscopy before passage of the radioactive compound for therapy.

Thermal Tumor Therapy

Direct heat also can be used to treat tumors for palliation. One device for this application is a bipolar device located on a conventional olive dilator. [56] The bipolar electrodes are arranged as stripes around the entire circumference of the olive or a portion of the circumference. Diameters of the probes vary from 6 to 15 mm. A guidewire is passed endoscopically through the narrowed lumen, the endoscope is removed, and the tumor probe is passed over the guidewire. [57], [58] The distal tumor margin is treated by bringing the probe up into the tumor at distances appropriate to cause overlapping coagulation through the narrow tumor segment. Alternative therapies include the use of the argon plasma coagulation device.

PATIENT PREPARATION

Before endoscopy is performed, patients should be asked about relevant medical illnesses, previous endoscopic or radiologic procedures, previous gastrointestinal surgery, and medications, especially those that may be related to gastrointestinal tract inflammation or irritation or interfere with coagulation. It is essential to know whether the patient has a history of dysphagia because this may give an indication that the person has a Zenker diverticulum, another type of diverticulum, or a history of aspiration. Aspiration suggests a fistula in a patient with a cervical or thoracic malignancy or inflammatory disease.

The physician should know about bleeding tendencies in the patient or the patient's family and about any history of drug allergies. If there is a history of bleeding abnormalities, a clotting screen with a prothrombin time, partial thromboplastin time, platelet count, and bleeding time is indicated. If there is evidence of chronic liver disease or other hematologic disorder, it is important to check coagulation.

Diagnostic endoscopy with biopsy is usually safe in patients who are taking oral anticoagulants or aspirin. However, for a patient who is to be treated with electrocoagulation for polypectomy or hemostasis, anticoagulants should be discontinued before the endoscopic procedure and for as long as 2 weeks after the procedure to prevent delayed bleeding. Aspirin-containing compounds and other antiplatelet agents such as nonsteroidal antiinflammatory drugs (NSAIDs) and ticlopidine should be avoided for an appropriate interval (i.e., aspirin for 7 to 10 days; NSAIDs for 2 to 3 days) before an endoscopic procedure that may involve removal of tissue. [59]

Most patients begin fasting after midnight the evening before the endoscopy. The patient should have no liquids for 4 hours before endoscopy and no solids for at least 6 to 8 hours beforehand. Patients with gastric outlet obstruction may be kept on a clear liquid diet for 24 to 48 hours, and lavage of the stomach with a large-bore tube may be necessary to remove retained stomach contents.

The patient must provide written or verbal informed consent before the procedure. Medications may be given before upper endoscopic procedures to anesthetize the posterior pharynx, reduce anxiety, [60] reduce discomfort, and diminish motility and secretions. Which medications are used depends on the clinical situation, including the patient's age, the existence of debilitating diseases, the anxiety level of the patient, the medication, and the patient's history of alcohol use. Upper gastrointestinal endoscopy is almost never performed with general anesthesia, with certain exceptions, such as removal of foreign bodies in small children.

For endoscopic procedures that are expected to be brief and in which small-caliber or pernasal endoscopes are used, no intravenous sedation may be necessary unless the patient is anxious. For therapeutic procedures that involve larger-diameter instruments and longer periods, the patient is usually given intravenous sedation. [61]

Bacteremia may be associated with endoscopic procedures. In a patient with a preexisting heart condition, there may be risk for infectious endocarditis, although this risk is small. The efficacy of antibiotic prophylaxis for endocarditis is unproven, although prophylaxis appears to be effective in patients with prosthetic valves, surgically constructed systemic-pulmonary shunts, and hypertrophic cardiomyopathy. [62], [63] and [64] Prophylactic antibiotics are advised for patients with known endocarditis or prosthetic cardiac valves. For gastrointestinal endoscopic procedures, the standard regimen is 2 g of ampicillin, administered intravenously or intramuscularly, plus gentamicin (1.5 mg/kg, not to exceed 80 mg), administered intravenously or intramuscularly 30 minutes before the procedure, followed by 1.5 g of amoxicillin taken orally 6 hours after the initial medication. Alternatively, the parenteral regimen may be repeated once at 8 hours after the initial dose.

INDICATIONS FOR UPPER ENDOSCOPY

Endoscopy is a highly accurate method of evaluating the mucosal surface of the esophagus, stomach, and duodenum. In addition to the various indications discussed in this section and summarized in Table 138-1, endoscopy is used to evaluate the patient with upper gastrointestinal symptoms before barium radiography is performed. This approach is referred to as *primary panendoscopy*. [65], [66], [67], [68], [69], [70], [71], [72], [73], [74] and [75] In many centers, the cost of this primary endoscopy is reduced because it can often be performed quickly and without any sedation.

Persistent or recurrent dyspepsia or heartburn	
Persistent nausea or vomiting	
Dysphagia or odynophagia	
Hematemesis or melena	
Early satiety or anorexia with weight loss	
Chest pain with negative results on cardiac evaluation	
Iron deficiency anemia with negative results on colon evaluation	
Caustic ingestion	
Suspected malabsorption (for small bowel biopsy)	
Failure to lose weight or recent weight gain after surgery for obesity	
Abnormal or equivocal barium meal radiographic studies	
Mass lesion	
Abnormal fold pattern	
Ulcer crater in esophagus or stomach	
Esophageal or gastric outlet stricture or obstruction	
Deformity or scar in a symptomatic patient	
Survey for Malignancy	
Barrett esophagus	
Familial polyposis or Gardner syndrome	
Follow-up of adenomatous gastric polyp	
Inadequately treated achalasia	
Therapeutic Endoscopy	
Control of bleeding	
Ligation or sclerosis of varices	
Dilation of stricture or narrow stoma	
Percutaneous gastrostomy	
Polypectomy	
Stenting an esophageal tumor	
Laser or tumor probe cauterization of a tumor	
Removal of foreign body	
Placement of feeding tube in the duodenum	
Follow-up Endoscopy	
Gastric or esophageal ulcer, healing or biopsy	
Ligation or sclerosis of varices	
Laser or tumor-probe cauterization of a tumor	
Removal of internal gastrostomy bolster	

Adapted from Morrissey JF, Rechenberger M: Gastrointestinal endoscopy. N Engl J Med 1991;325:1143.

TABLE 138-1 Indications for Upper Gastrointestinal Endoscopy

Dysphagia

Dysphagia is an important symptom in gastroenterology that mandates a full evaluation, and endoscopy is critical in the evaluation of these symptoms. [76] Some endoscopists think a barium esophagogram should be performed before endoscopy, but endoscopy is increasingly used as the first diagnostic test in a patient with dysphagia. In a patient whose symptoms suggest dysphagia caused by Zenker diverticulum, it is essential to have a barium esophagogram study before endoscopy to avoid injury to the diverticulum during passage of the endoscope. Endoscopy in patients with a Zenker diverticulum may be difficult and dangerous because the esophageal lumenal opening becomes almost invisible. To avoid perforation of the diverticular sack, it may be wise first to insert a soft nasogastric tube through the

nose and to use the latter as a guide to locate the laterally displaced esophageal lumen. Endoscopic incision of the septum between esophagus and diverticulum proves to be an efficient and safe method for treating symptomatic disease, especially in high-surgical-risk patients. [77], [78]

If a patient has symptoms referable to the pharynx or neck, enlarged cervical lymph nodes, or neck pain, evaluation should be done first with direct inspection of the larynx, pharynx, and nasal passages. Flexible endoscopy is of limited value in examining the pharynx and hypopharynx. It is possible to view lesions that cause dysphagia endoscopically, including reflux-induced strictures (see Fig. 138-8), caustic injury strictures, strictures resulting from radiation, achalasia, and benign (Fig. 138-10; see also Color Fig. 138-10) and malignant tumors.

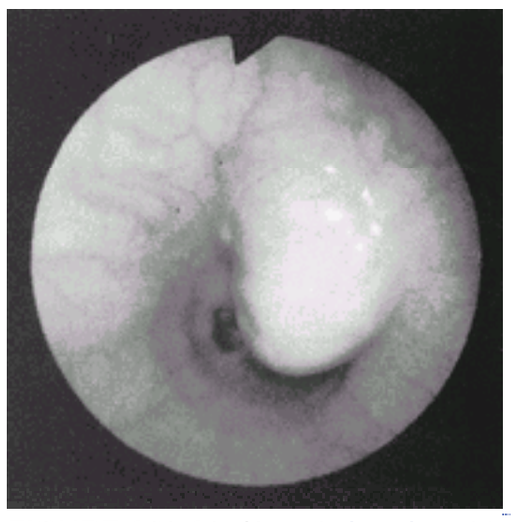

FIGURE 138-10. (See Color Fig. 138-10.) Esophageal leiomyoma.

Esophageal Cancer

A patient often presents with esophageal cancer after the malignancy is far advanced. Advanced cancers have three morphologic appearances. A tumor that is nodular and friable with a hemorrhagic surface with ulcers or erosions is referred to as *exophytic* and has a wide base (Fig. 138-11; see also Color Fig. 138-11). The second type of tumor, in which the lesion is an ulcer with heaped-up surrounding edges, is referred to as *ulcerative*. The third morphologic presentation is a thickened, rigid length of esophageal wall caused by infiltrating cancer, referred to as *infiltrating,* in which the mucosa is often nodular and appears fixed.

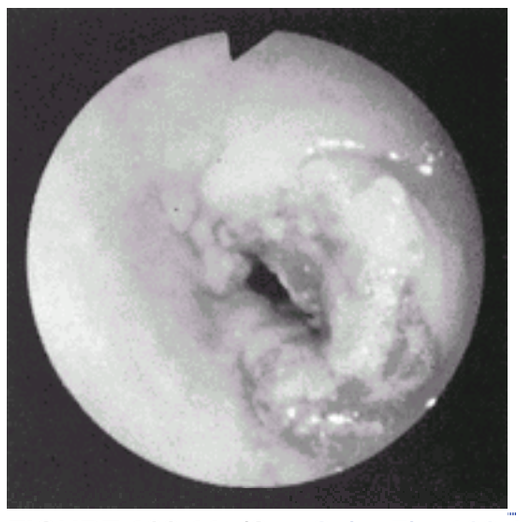

FIGURE 138-11. (See Color Fig. 138-11.) Esophageal carcinoma obstructs the esophagus.

The diagnosis of esophageal cancer requires multiple endoscopic biopsies. Obtaining tissue for a good histological examination is occasionally difficult because of overlying inflammatory exudate, but in most polypoid tumors, biopsies are positive because the proximal tumor edge is visible and can be directly targeted. If the approach to the lesion is tangential, it may help to use biopsy forceps with a central spike. An alternative is to use needle cytology. If nodular lesions are proximal to the main lesion, a biopsy should be performed because such lesions may represent intramural metastasis from the primary tumor.

If the lesion appears as a narrowing or stenosis, it may be difficult to obtain positive biopsy specimens. In this instance, multiple biopsies and brush cytologies may be necessary to make a precise diagnosis. There are two approaches to obtaining tissue from the narrowed area. The first is to use a small-diameter endoscope, which can be passed through the stenotic segment and can obtain biopsy specimens from the entire length of the stenosis. If the stenosis is too tight to permit even a small-caliber endoscope, it may be possible to obtain brush cytology samples from the narrowing and perform a biopsy at the proximal edge. The second approach is to perform dilation of the narrowed segment before performing biopsy and cytology procedures. By dilating up to a diameter of 10 mm, it is possible to pass a small-caliber endoscope through the length of the lesion and obtain biopsy specimens from the proximal and distal edges and along the length of the stenosis.

Overall, a correct diagnosis of esophageal cancer is made in more than 90% of patients if six or more biopsies are performed and if cytologic samples are examined. Multiple biopsies are especially important if there is a large amount of necrotic tumor and exudate. [1]

Gastroesophageal Reflux Disease

Not all patients with esophageal reflux symptoms require endoscopy. If a patient has a chronic or intermittent symptom complex, if the symptoms are not controlled using standard antireflux measures, or if the symptoms occur at night and cause difficulty with coughing, hoarseness, and aspiration, endoscopy may be indicated. Of the patients who present with reflux symptoms, about one half have endoscopic evidence of mucosal injury in the distal esophagus (Fig. 138-12; see also Color Fig. 138-12). The purpose of endoscoping the patient is to detect this injury, to grade the severity of acid-pepsin or bile-induced injury, and to detect esophageal columnar metaplasia in the distal esophagus.

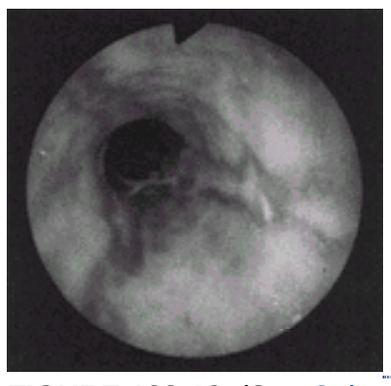

FIGURE 138-12. (See Color Fig. 138-12.) Reflux esophagitis with confluent but noncircumferential breaks or erosions.

It is possible during endoscopy to diagnose a hiatal hernia. The level of the diaphragmatic impression must also be located, if necessary by having the patient sniff during assessment. If, during quiet observation without excessive distention of the esophagus with air, the gastroesophageal junction is more than 2 to 3 cm above the diaphragm, this condition is thought to represent an axial hiatal hernia. The hernia appears endoscopically to be a pouch or widening lined with gastric folds just below the mucosal junction and just above the diaphragm. After the instrument is passed into the stomach and is retroflexed, it is possible to examine the cardia from below and diagnose a hiatal hernia.

Grading of reflux mucosal injury may be accomplished by several schemes, but the modified Savary and the Los Angeles systems [79], [80] and [81] are now often used. One difference between the various systems is whether minor or equivocal changes are included. These changes include erythema, friability, and blurring of the ora serrata.

The sentinel fold or polyp is an abnormality found in some patients with a hiatal hernia. It is a fold with a polypoid proximal margin that is located just distal to the squamocolumnar junction. There is often a small erosion or ulcer on the proximal tip of the fold, which may be seen in a patient with a hiatal hernia and reflux.

Esophageal Columnar Metaplasia

Esophageal columnar metaplasia, also called Barrett metaplasia, is a change in the normal appearance of the esophagus; the mucosa that is normally stratified squamous is replaced by a columnar mucosa. The normal ora serrata or squamocolumnar junction is located more proximally in the esophagus. The only way to determine the anatomic junction of the esophagus and the stomach is that this junction occurs just proximal to the gastric folds. [82], [83] and [84] The distance from the proximal margin of the gastric folds to the squamocolumnar mucosal transition does not vary and should be consistent from one endoscopy to the next.

If the distance from the squamocolumnar junction to the proximal gastric folds in a hiatal hernia is longer than 2 to 3 cm, it usually means that there is a segment of columnar metaplastic mucosa (Fig. 138-13; see also Color Fig. 138-13). A short segment is diagnosed when less than 2 to 3 cm of specialized or intestinal-type columnar metaplastic epithelium is found.

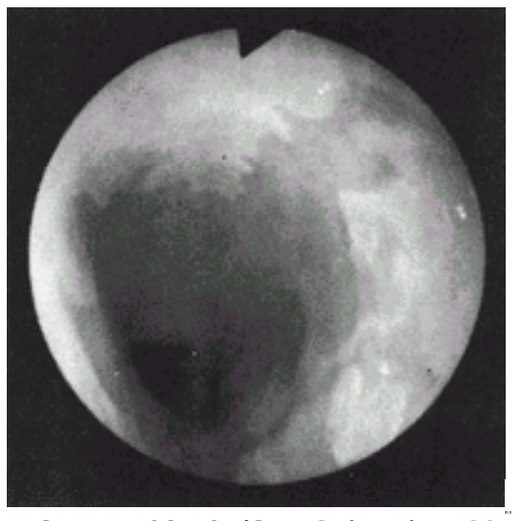

FIGURE 138-13. (See Color Fig. 138-13.) A columnar mucosa–lined esophagus, also known as Barrett esophagus.

Biopsy is not routinely performed in patients with esophageal reflux symptoms unless columnar metaplasia is suspected. In a patient with reflux esophagitis, periodic endoscopy is not required. Exceptions are patients who have not responded to therapy, especially if they have severe esophagitis, and patients who are to undergo any surgery for reflux, in whom presurgical endoscopy is indicated.

Dyspepsia

The symptom complex known as dyspepsia includes pain, burning in the epigastrium, nausea, epigastric fullness, and bloating or distention. Only about one of five patients who present with these symptoms actually has a peptic ulcer. It is now accepted that upper endoscopy is the first-line investigation for patients presenting with significant dyspeptic symptoms. It may be difficult for the clinician to decide whether endoscopy is appropriate. Gastric malignancy and peptic ulcer disease are rare in the dyspeptic patient younger than 45 years of age unless infected with *Helicobacter pylori* or taking NSAIDs. Diagnostic endoscopy might be limited to the latter patients. Now that effective anti– *H pylori* treatments are available, it might be argued that young *H pylori*–positive dyspeptic patients would be better managed by empiric treatment of the infection rather than by referral for endoscopy first. [85], [86]

In some patients with dyspepsia, endoscopy may show patchy erythema, subepithelial hemorrhage, or erosions without distinct ulceration (Fig. 138-14 and Fig. 138-15). Erosions typically have a white base surrounded by a rim of erythema. In evaluating a patient with chronic dyspepsia, not uncommonly resistant to empiric therapy, mucosal biopsies may be taken from the gastric antrum and the corpus. [87], [88] Whether all chronic dyspeptic patients or only those with erosive abnormalities should undergo biopsy remains a controversial issue. *H pylori*–associated inflammation is the dominant factor in the pathogenesis of peptic ulcer disease that is not associated with NSAIDs. [89] The role of *H pylori* infection in functional dyspepsia is still not clarified. [90], [91]

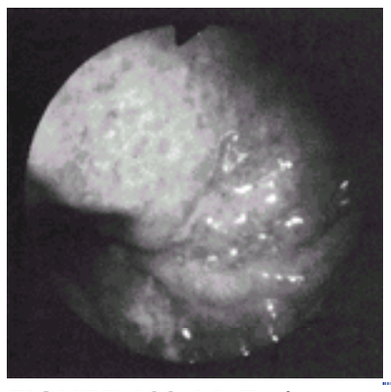

FIGURE 138-14. Endoscopy reveals punctate erythema and swelling.

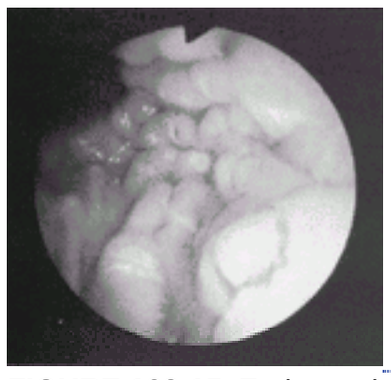

FIGURE 138-15. Endoscopic view of erosive gastropathy.

Gastric Ulcers

Most often, benign gastric ulcers are found close to the incisura or in the distal corpus of the stomach along the lesser curvature. The antrum just proximal to the pylorus is another common area of ulceration, especially of drug-induced ulceration. Ulcers occur in the upper portions of the stomach more commonly in elderly people. If multiple ulcers are present, a drug-induced etiology should be suspected. [92] Most gastric ulcers have diameters less than 3 cm. Typically, an ulcer has a white base with erythematous mucosal margins. These margins are smooth and slightly raised in a benign gastric ulcer. The folds surrounding a benign gastric ulcer usually radiate in an unbroken fashion from the ulcer margin without intervening nodularity; nodularity raises the suspicion of a malignant ulcer.

It is often impossible to be certain from the endoscopic appearance that no malignancy is present in a gastric ulcer. Of those ulcers that appear benign radiographically, 5% to 10% are found to be malignant in some studies. Endoscopy and biopsy are therefore mandatory if a gastric ulcer appears to be indeterminate or suspicious (Fig. 138-16; see also Color Fig. 138-16). Endoscopists should realize that there is an appalling lack of agreement on descriptive terminology and interpretation of ulcer features. [93]

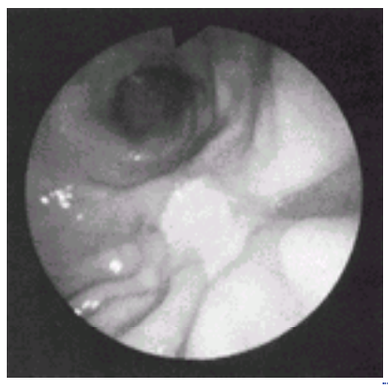

FIGURE 138-16. (See Color Fig. 138-16.) Atypical gastric ulceration along the greater curvature was presumably induced by nonsteroidal antiinflammatory drugs.

Under endoscopic guidance, the physician should obtain multiple biopsy specimens from the margin and the base of a gastric ulcer. [94], [95] At least six biopsy specimens should be taken. It is important to take the biopsy specimens of the ulcer margin as close to the ulcer base as possible to increase the yield if a tumor is present. If the ulcer base is abnormal or appears to have nodules, a biopsy should be performed there. It may be important to follow gastric ulcers until full healing with a repeat endoscopy and multiple biopsies in certain circumstances. It is possible for a malignant ulcer to heal with therapy.

If the patient presented initially with an upper gastrointestinal bleed, biopsies are often deferred. If the endoscopic appearance of the lesion was atypical, it may be important to repeat endoscopy and biopsy after several weeks of therapy.

Duodenal Ulcers

Endoscopy is considered to be the most accurate imaging modality in the evaluation of the duodenal bulb to detect a duodenal ulcer. About 50% of duodenal ulcers are located on the anterior surface of the duodenal bulb. As with gastric ulcers, the mucosa surrounding the ulcer is often red. Adjacent to the ulcer, there may be a second duodenal ulcer, duodenal erosions, or duodenal scarring from previous ulceration. If there is profound scarring from previous ulceration, outpouchings of the bulb may develop, resembling diverticula (Fig. 138-17; see also Color Fig. 138-17). Duodenal ulcers heal at a rate of several millimeters per week. In a patient with a duodenal ulcer, an antral biopsy should be obtained to document the presence of *H pylori* infection. If appropriate, antimicrobial sensitivities can also be obtained. It is not recommended that biopsies be taken directly from a duodenal ulcer unless it is in some way atypical, suggesting the presence of Crohn's disease, malignancy, or infectious or parasitic disease.

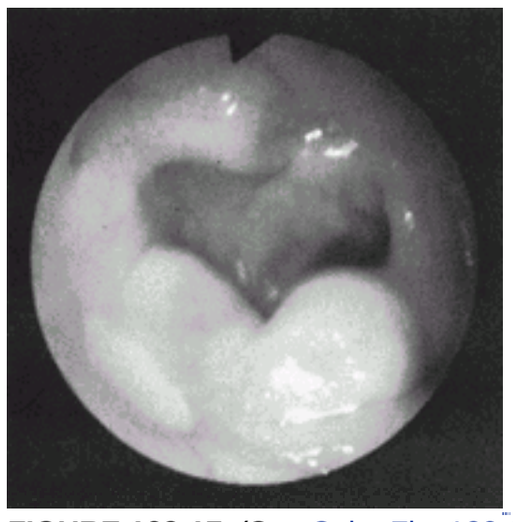

FIGURE 138-17. (See Color Fig. 138-17.) This deformed bulb was caused by scarring after prior ulceration and recurrent duodenal ulcer.

Upper Gastrointestinal Tract Bleeding

In a patient who presents with upper gastrointestinal bleeding, endoscopy is the first diagnostic procedure undertaken. It is usually possible to perform endoscopy on all patients with upper gastrointestinal bleeding unless they have torrential hemorrhage. Most endoscopists stabilize hemodynamically a patient presenting with upper gastrointestinal bleeding and then perform endoscopy. Endoscopy provides valuable information for the diagnosis and planned therapy of the patient, and the advent of therapeutic endoscopy has given the endoscopist the chance to change the natural history in the bleeding patient. Urgent early endoscopy is indicated if the patient presents with hemodynamic stability, if the patient has liver disease, if the patient is rebleeding, or if there is a history of previous aortic-prosthetic surgery because of the possibility of aortoenteric fistula.

The esophagus is examined to determine whether obvious lesions are present that can account for the bleeding, such as esophageal varices or a mucosal Mallory-Weiss tear. In a patient with bleeding varices, both the esophagus and stomach are examined for varices and for portal hypertensive or congestive gastropathy. Varices are assessed by size (e.g., small, medium, large) and the presence of red color signs on the surface of the varix (e.g., red wale markings, hematocystic spots, cherry-red spots). Portal hypertensive or congestive gastropathy may appear as multiple erythematous areas outlined by a white reticular network (mosaic-like pattern), a fine pink speckling (scarlatina-like pattern), or cherry-red spots on a finely granular mucosa with or without spontaneous bleeding in areas of confluence (red-spot pattern). The three patterns are likely to represent particular aspects of a continuous spectrum of congestive mucosal changes. The gastric examination is performed with the instrument straight and retroflexed. Before endoscopy, it is often advisable to lavage the stomach to remove clots; if a clot is still present in the stomach, most endoscopists advance the tip of the endoscope over the clot and into the antrum. If there is brisk duodenal bleeding, blood can be seen refluxing back through the pylorus into the antrum. The endoscope is passed through the pylorus into the duodenal bulb and down into the descending duodenum. If an ulcer is encountered, its size, appearance, associated visible vessel or adherent clot, and bleeding activity are assessed. The instrument is withdrawn into the stomach and retroflexed to examine the lesser curvature, cardia, and fundus. If blood and clots are present, the patient's position may have to be changed to allow the entire gastric mucosa to be inspected. In some instances, it is possible to follow a trail of blood up the mucosa and find the actively bleeding lesion.

In the esophagus, the most important lesions are esophageal varices and Mallory-Weiss tears. Other lesions include cancers, Barrett ulcers, and severe reflux esophagitis. The most common causes of bleeding in the stomach are gastric ulcer, gastric varices (Fig. 138-18; see also Color Fig. 138-18), vascular abnormalities such as telangiectasias (Fig. 138-19; see also Color Fig. 138-19), gastric erosions or hemorrhagic gastropathy (e.g., due to stress, NSAIDs), or a Dieulafoy vascular lesion. In the duodenal bulb, the most common cause of bleeding is duodenal ulcer (Fig. 138-20). Other lesions include erosive duodenitis, often thought to be drug induced. The duodenum may also bleed from vascular abnormalities and from tumors, especially pancreatic tumors infiltrating into the duodenum.

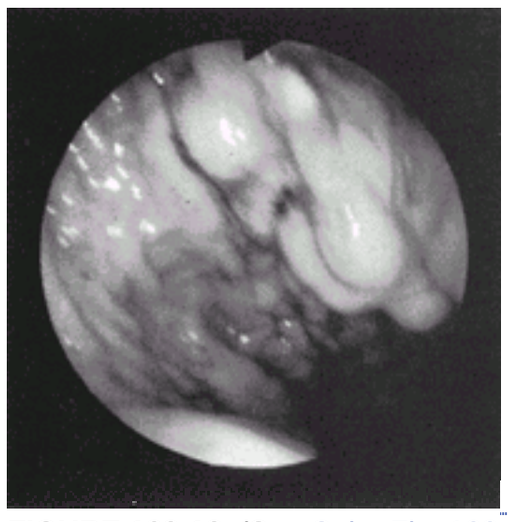

FIGURE 138-18. (See Color Fig. 138-18.) Gastric fundus varices.

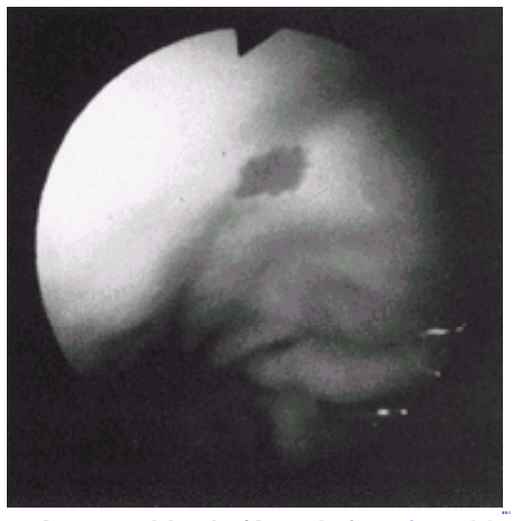

FIGURE 138-19. (See Color Fig. 138-19.) Gastric mucosal vascular ectasia.

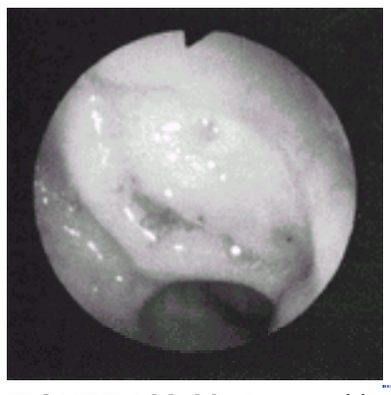

FIGURE 138-20. A vessel is visible in the base of this duodenal ulcer.

Infection of the Esophagus

Candida species infection of the esophagus has several characteristic endoscopic appearances, including a typical white exudate, pseudomembranes, friable mucosa, and rarely, ulcers. The most accurate method of diagnosing fungal infection of the esophagus is endoscopic brushing and biopsy. [97] Brush specimens of the esophageal exudate show mycelia. Some endoscopists obtain a biopsy specimen because the sample may show evidence of deeper tissue involvement with fungus. It is not useful to culture the exudate or the biopsy specimen, because *Candida* species may be demonstrated by culture without significant mucosal infection.

In the differential diagnosis of esophageal infection in the immunocompromised host, several viral infections must be considered, including herpes simplex virus (HSV), varicella-zoster virus (VZV), and cytomegalovirus (CMV). Each of these viruses may be associated with ulcers in the esophagus, usually in patients who are immunocompromised. CMV is thought to affect the deeper, submucosal tissue, and HSV and VZV affect more superficial epithelium.

In HSV infection, vesicles are often seen as the initial lesion. As the lesion progresses, the center of the vesicle sloughs, leaving a small ulcer, the edges of which are usually raised. If these ulcers coalesce, extensive areas may be involved. The diagnosis is made by histological examination and viral culture of brushings or biopsy specimens taken from the edge or center of the ulcers. A necrotizing esophagitis can also occur from VZV infection, especially in immunocompromised patients. As with HSV, lesions may vary from vesicles to larger lesions, and biopsy specimens and brushings are obtained for histological analysis.

The lesions of CMV esophagitis involve the submucosa. The typical endoscopic appearance is that of a geographic pattern ulcer, which may be large, especially in the distal esophagus. [1] Endoscopic brushing of the exudate is less useful for culturing CMV. Biopsy specimens from the center of the ulcer are often required for the diagnosis of CMV. [97] In the immunocompromised host, CMV infection may involve the mucosa and submucosa of the stomach and duodenum in addition to the esophagus.

Removal of Foreign Bodies

Foreign body ingestion occurs in all types of patients, but there is an increased incidence in children, in patients who wear dental plates, and in psychiatrically ill patients or prisoners. Most commonly, adults present with dysphagia or esophageal obstruction caused by a piece of meat or a bone, but children and psychiatrically disturbed patients ingest a variety of objects. [36] The foreign body tends to lodge in the esophagus in areas of physiologic narrowing, such as near the left main-stem bronchus, aortic arch, and just above the diaphragm. [98] Not all ingested foreign bodies require removal, but some foreign bodies that are impacted or have sharp edges should be removed. Of immediate clinical concern is impaction of a foreign body in the esophagus or hypopharynx. Profuse salivation in a patient is considered a clinical sign of esophageal obstruction that must be addressed immediately.

Any foreign object that impacts in the esophagus needs attention, including coins or batteries, which contain corrosive elements. If a radiopaque foreign body passes spontaneously into the stomach, serial abdominal flat films can be used to watch its progress through the gastrointestinal tract. A sharp object, such as an open safety pin or razor blade that remains stuck for more than 2 or 3 days, should be removed endoscopically or surgically.

A coin often lodges in the proximal esophagus just distal to the cricopharyngeal muscle. Posteroanterior and lateral radiographic views of the neck are necessary to identify the exact location of the foreign body and to determine whether the foreign body is in the esophagus or the trachea. A rigid laryngoscope and forceps can be used to remove the coin; alternatively, forceps or a snare is passed by way of a flexible endoscope. The patient can be placed in Trendelenburg position to prevent aspiration of the coin into the trachea if an endotracheal tube is not used. Special care is required if a coin or button battery is pulled through the upper esophageal sphincter because these foreign bodies may become dislodged from a snare or toothed forceps as they are being removed.

A foreign body such as meat is commonly encountered in adults and may totally obstruct the distal esophagus. A polypectomy snare can often be used to grasp and remove the meat. Experience has shown that after a meat bolus has been snared or placed in a basket, it is best to pull the bolus up against the tip of the endoscope as the endoscope is being withdrawn to minimize the likelihood of the bolus becoming dislodged from the snare or basket as it is being removed. If the meat fragments into smaller pieces, an overtube is necessary to reduce patient discomfort. As the endoscope is passed in and out to remove small pieces of the foreign body, the overtube protects the airway and prevents a piece of the foreign body from being dropped in the hypopharynx.

In some instances, a small-diameter endoscope can be passed next to the obstructing bolus to determine whether a stricture is present below the meat. If a stricture is found, it can be dilated with a balloon. The instrument can be withdrawn to above the bolus of meat and can be used to push the bolus of meat gently down into the patient's stomach. However, pushing a foreign body distally without knowing the cause of the obstruction should be avoided because this can cause a perforation.

The greatest difficulty in protecting the airway occurs with sharp objects such as pins and razor blades. If a single-edged razor blade is lodged in the esophagus, an overtube should be used. The razor blade should be pulled into the overtube as it is gently removed, protecting the esophageal mucosa. An overtube is also helpful for removing a pin. If a safety pin is open in the esophagus with the sharp end of the pin pointing proximally, the best management approach is to push the pin into the stomach, rotate it, and pull it out by the hinge with the pointed portion of the pin trailing and pointing away from the endoscope. Other commonly encountered foreign bodies are small batteries. The batteries should be removed to prevent possible serious and even fatal complications if they are stuck in the esophagus. If small batteries rapidly move into the stomach, they usually pass without problems.

After a foreign body has passed the pylorus and is in the small bowel, it usually passes spontaneously. In some instances, a foreign body must be removed surgically from the stomach or from the intestinal tract beyond the stomach if endoscopic removal is not successful. A radiograph should be taken immediately before attempted removal because, in many instances, the foreign body has spontaneously passed out of the stomach, and endoscopy is no longer indicated.

Caustic Injury

Caustic injury may vary from a minimal and superficial burn to severe necrosis involving the entire upper gastrointestinal tract into the duodenum. Ingestion of alkali is often more problematic than ingestion of strong acids. [99] Endoscopy allows the physician to assess the organs injured and, to some degree, determine the extent of the injury. In most instances, endoscopy should be performed early in the patient's evaluation if the patient is stable. If there is evidence of a full-thickness injury to the intestinal tract with possible perforation, shock, or severe injury to the hypopharynx or epiglottis, endoscopy should not be performed. The examination is usually performed with a small-caliber endoscope. The recommended technique of passage is under direct vision with minimal air insufflation. If an area of severe necrosis is visualized, the endoscopy should not proceed beyond this area.

It is possible to divide the extent of the injury caused by caustics into three degrees of injury. [100] The mildest or first-degree injury is characterized by mild friability, erythema, and edema but no evidence of necrosis or ulceration. These superficial injuries often heal completely without long-term sequelae. Second-degree injuries extend into the wall and occasionally to the muscularis propria. Endoscopically, a more severe injury is seen with necrosis, ulcers, exudates, and areas of hemorrhage. These deeper injuries characteristically result in strictures of the severely injured organ, the esophagus or stomach. The most severe injury is a third-degree burn involving the full thickness of the wall. The injury is seen endoscopically as a dark exudate, with sloughing of the mucosa in addition to hemorrhage and ulceration. The organ may appear aperistaltic and dilated. This injury often leads to the late sequelae of esophageal stricture or strictures in the stomach.

Third-degree burns may also be associated with injury to adjacent organs and may present with mediastinitis or peritonitis.

The purpose of early endoscopy is to determine the extent and severity of the burn. A black necrotic area with ulceration suggests an injury through the full thickness of the wall and an increased risk for perforation. In these patients, early surgical intervention must be considered before perforation occurs. If, at the initial endoscopy, an area of full-thickness injury is suspected, it is usually recommended that the endoscopy be discontinued at that point and repeated after 48 hours to determine whether these areas appear to be healing. The patient must be watched for clinical signs of perforation requiring operative intervention.

Drug-Induced Injury

Some medications, such as tetracycline, ascorbic acid, and iron preparations, have an acid pH and directly injure the mucosa. Some medications, such as potassium preparations, are designed for slow release, and if they remain for a prolonged time in the esophagus, they can cause injury. Also, bisphosphonates may damage the esophagus. This problem is made worse if the patient swallows the medication with a small amount of water before going to bed. In elderly patients, a reduced amount of saliva also may contribute to the problem. [101], [102] Endoscopically, these patients have ulcerations of the esophagus characteristically located above areas of physiologic narrowing, where these medications tend to lodge. The injury may vary from friability and erythema to multiple clear-cut ulcerations, often seen as "kissing" ulcers in direct opposition to one another on the esophageal mucosa. These iatrogenic drug-induced injuries may promote esophageal stricture, especially if the medication is taken over a long period.

Mass Lesions

Endoscopy is often used to evaluate masses seen on barium radiographs in the upper gastrointestinal tract, especially in the stomach. Endoscopic ultrasound may prove to be important in the evaluation of these mass lesions. Gastric adenocarcinomas are categorized by the Borrmann classification into four types of tumors. Type 1 is an exophytic mass protruding into the gastric lumen. Type 2 is a protruding mass with associated ulceration. Type 3 is a diffusely infiltrating tumor, often associated with ulceration. Type 4 is an infiltrating mass without associated ulcer. Type 3 lesions can be confused with a benign gastric ulcer. To make this distinction, at least six to eight biopsy specimens from all quadrants of the ulcer are necessary to detect carcinoma. Brush cytology may also be useful. Type 4 adenocarcinoma is often referred to as *linitis plastica* and has diffuse involvement of the gastric wall. This tumor may be mainly submucosal, and biopsy of the mucosal surface over the tumor may reveal relatively normal tissue. It is important to retrieve specimens from any abnormal areas of mucosa associated with these lesions because these are most likely to be the areas in which the tumor has broken through to the surface and to be positive for tumor. Most endoscopists prefer using biopsy forceps of large caliber for these biopsies or needle cytology.

The classification of early gastric cancer was defined by the Japanese Endoscopic Society. Type 1 is a small polypoid lesion. Type 2 can be elevated (2a), flat (2b), or depressed (2c). Type 3 is a lesion that presents as a gastric ulcer with an adenocarcinoma at the margin, often producing a discolored and slightly depressed area. The most common types of early gastric cancer are types 2c and 3. Endoscopic gastric mucosal resection is increasingly employed in early gastric cancer, to remove the tumor with the surrounding mucosa and part of the submucosa after submucosal saline injection to raise the lesion, appropriately stained with indigo-carmine dye scattering to define the extent of the lesion. This technique is especially suitable for cancer confined to the mucosa (with low probability of lymph node metastasis) and with a diameter of 20 mm or less. [103], [104], [105] and [106]

The appearance of gastric lymphoma varies from nodular or polypoid to that of ulcerations surrounded by a nodular and infiltrated mucosa. A deep, volcano-like ulcer is reported, which has elevated surrounding margins. The ulcers are characteristically irregular in their margins and are associated with nodules. Exudate in the base may appear gray. Multiple biopsies often are necessary to diagnose a gastric lymphoma correctly.

ENDOSCOPIC SURVEILLANCE FOR PREMALIGNANT LESIONS

The incidence of neoplasia is increased in several conditions and in some postoperative circumstances in the upper gastrointestinal tract. However, well-designed controlled studies are lacking to define precisely the risk in these conditions of developing malignancy, and it is difficult to determine exactly which conditions require periodic surveillance. [107]

Achalasia

Esophageal cancer develops in 2% to 8% of patients with untreated achalasia after more than 15 years of disease. If the patient is adequately treated with balloon dilation or a surgical myotomy, there may be a smaller increase in the risk for malignancy. [108] There does not appear to be a role for endoscopic surveillance in achalasia. However, any patient with achalasia who experiences a change in signs or symptoms should undergo endoscopy to determine whether a malignancy is present.

Esophageal Columnar Metaplasia

There is an increased risk for adenocarcinoma, in the range of 8% to 10%, in patients with Barrett metaplasia. The premalignant risk does not appear to be reduced by antireflux surgery, despite the fact that this surgery may be associated with a reduction in inflammation. Most endoscopists agree that patients with columnar metaplasia should be screened endoscopically with multiple biopsies, although the exact frequency of surveillance has not been determined. [109], [110], [111], [112] and [113]

Gastric Polyps

Adenomatous gastric polyps are thought to be associated with an increased risk for containing malignancy within the polyp. [114], [115] Hyperplastic polyps are not associated with cancer in the polyp, but adenomatous and hyperplastic polyps may be associated with gastric atrophy, which has been associated with gastric cancer in areas other than the polyp.

Larger adenomatous polyps appear to pose a greater risk for malignancy. It is necessary to remove the entire polyp for adequate histological assessment because random biopsies may miss a focal cancer. Polypectomy may also treat symptoms caused by a polyp, such as bleeding or obstruction. If a polyp is encountered, it should be removed by endoscopy or surgery. The size of the polyp, the number of polyps, and the patient's general medical condition are important factors in determining whether surgery or endoscopy should be used for treatment. If the polyp is hyperplastic, no further surveillance endoscopy is indicated. If the patient has a limited number of small adenomatous polyps (less than 2 cm), endoscopic removal of the polyp is recommended. These patients are followed with surveillance endoscopy to detect recurrent polyps or the development of a carcinoma. If the polyp is adenomatous but is large and sessile, there is an increased risk for complications from endoscopic removal, and surgical therapy is often recommended.

Atrophic Gastritis

Chronic gastritis with severe atrophy is associated with gastric adenocarcinoma (Fig. 138-21; see also Color Fig. 138-21). Only rarely can extensive intestinal metaplasia be identified endoscopically. Mucolysis followed by methylene blue staining may facilitate the recognition of intestinal metaplasia endoscopically and allow targeted biopsy of the lesion. There is no consensus whether regular surveillance should be carried out in such patients.

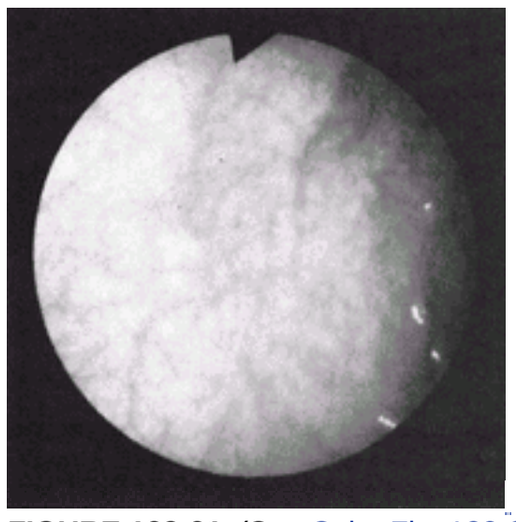

FIGURE 138-21. (See Color Fig. 138-21.) Atrophic gastritis.

Autoimmune Gastritis and Pernicious Anemia

Several population-based studies indicate that the risk for gastric adenocarcinoma and carcinoid tumor is increased in patients with pernicious anemia, but only slightly over that of the general population. Periodic surveillance endoscopy in patients with pernicious anemia is not recommended, except in patients who have known dysplastic changes of the mucosa. [116], [117] and [118] The importance of small carcinoids and endocrine cell hyperplasia in the corpus requires further investigation.

Adenocarcinoma in the Gastric Remnant After Partial Gastrectomy

Several studies have demonstrated that patients undergoing gastric resection for benign gastric or duodenal ulcers are at increased risk for later developing dysplasia and adenocarcinoma. The reported increase in the risk for cancer ranges from 2% to 8.7%. [119] However, other reported series have failed to demonstrate an increased risk for carcinoma in these postoperative patients. The risk for malignancy is not thought to be high enough to warrant routine endoscopic screening of patients who have had gastric resections. In countries in which the risk for postgastrectomy cancer is several times the normal incidence, however, the patients should be screened endoscopically, especially patients who underwent surgery before 50 years of age and those in whom resection was performed at least 15 years ago. If endoscopy is being performed for a different indication on a patient who had gastric resection many years earlier, some endoscopists recommend taking multiple biopsy specimens from the margin of the stoma to look for evidence of dysplasia or early cancer.

RISKS AND CONTRAINDICATIONS

Endoscopic examination of the upper gastrointestinal tract by an experienced endoscopist is a safe procedure even during pregnancy [120] or after esophagogastroduodenal surgery. [121] Cardiopulmonary complications, infection, bleeding, and perforation occur only rarely. Each of these complications is thought to occur in fewer than 0.1% of patients undergoing endoscopy. Overall, the incidence of serious complications with diagnostic upper gastrointestinal endoscopy is estimated to be about 1 in 1000 procedures, with an estimated mortality between 1 and 6 per 20,000 examinations. The smaller-caliber endoscopes are even safer. Several studies have suggested that the complication rate of endoscopy in patients who were experiencing acute gastrointestinal bleeding is higher than in patients who were electively examined. There is, however, no evidence that endoscopy exacerbates bleeding. Cardiopulmonary complications account for more than one half of endoscopy-associated deaths. [122]

Contraindications to endoscopy include suspected perforation, an uncooperative patient, severe shock or respiratory distress, and severe injury to the hypopharynx. Patients with coagulopathies should be examined endoscopically only in life-threatening situations and if the result of the endoscopy is thought to be critical to the patient's care.

ASSESSMENT OF RESULTS

Physicians have for years debated the relative merits of endoscopy and barium contrast radiography in evaluating the esophagus, stomach, and duodenum. [123], [124] and [125] Studies comparing these diagnostic modalities have always suffered from the problem of which technique is to be used as the standard. Radiologists contend that they are able to detect important lesions of the upper gastrointestinal tract in a manner comparable to that achieved with endoscopy. [125] There are distinct advantages to endoscopy. First, endoscopy allows detection of superficial mucosal lesions, such as reflux esophagitis, gastritis, and duodenitis, better than radiography. Endoscopy is thought to be more sensitive and specific for peptic ulcer. Endoscopy permits cytology and biopsy, making endoscopy superior to radiology for diagnosing infectious conditions such as esophagitis with *Candida* species infection or CMV, or for diagnosing gastritis with *H pylori*. However, each type of diagnostic study fails to detect certain lesions, and there are clinical circumstances in which one or the other of these modalities is best or in which a combination of modalities is required.

In upper gastrointestinal bleeding, endoscopy is the diagnostic procedure of choice. Barium radiography has a low rate of detection of the bleeding source, and the barium can interfere with subsequent endoscopy or angiography. Endoscopy offers the possibility of hemostatic therapy. The rapid increase in use of endoscopy is easily understandable because endoscopy is thought to be more accurate, allows the endoscopist to take targeted biopsy specimens, permits brush cytology, and permits the delivery of various types of therapy.

FUTURE TRENDS

The role of endoscopy in clinical medicine will continue to evolve now that new imaging modalities are being developed, [126] such as high-resolution endoscopy, [127] fluorescence endoscopy, [128] optical coherence tomography, [129] and confocal laser scanning microscopy. [130] Ultrasound endoscopy will be used increasingly for diagnosis and for guiding therapy. Wireless capsule endoscopy is on the horizon. Time will tell how useful this technology will ultimately become. [131], [132]

The training of new endoscopists is important, and as part of this education, a standardization of terminology is vital, especially in the use of videoendoscopy to examine areas of inflammation. Issues such as interobserver and intraobserver variability must be considered as we examine the sensitivity and specificity of endoscopy.

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CHAPTER 139

Paul Swain

ENTEROSCOPY

Enteroscopy is the endoscopic evaluation of the distal duodenum, jejunum, and ileum. [1], [2] and [3] Upper endoscopy with conventional forward or side-viewing instruments of a meter in length can reach the junction of the second and third portions of the duodenum, and colonoscopy can intubate and inspect the terminal ileum for a few centimeters. These methods will not be considered in this chapter. There are four methods available for enteroscopy: push enteroscopy, sonde enteroscopy, intraoperative enteroscopy, and wireless capsule enteroscopy. Specialized instruments have been developed for sonde and push enteroscopy (Fig. 139-1).

FIGURE 139-1. A sonde-type enteroscope (Olympus SIF-SW) with the balloon at its tip inflated and a push-type enteroscope (Olympus SIF-10).

PUSH ENTEROSCOPY

Push enteroscopy involves advancing a long endoscope into the small bowel during upper intestinal endoscopy; it has become the most common form of enteroscopy. During push enteroscopy, an endoscope can be pushed beyond the ligament of Treitz into the proximal jejunum. Instruments have been specially designed for this application which have increased tip flexibility, relatively small diameter, improved transmission of force to the endoscope tip, and a conventional biopsy channel. [1], [2] and [3] Initial experience in push enteroscopy was gained using an adult or pediatric colonoscope passed through the mouth. [4], [5], [6], [7], [8] and [9] A colonoscope can be advanced a fair distance into the small bowel, although probably not as far as a push-type enteroscope. A pediatric colonoscope may be superior to an adult colonoscope for enteroscopy because of its smaller outer diameter, but both have been used.

Minor improvements in the design of dedicated push-type enteroscopes, including increased angulation, [1], [2] and [3] better push transmission, [1], [10], [11], [12] and [13] and increased length, [12] combined with greater clinical experience, have led to deeper intubation of the jejunum. As a result, push-type enteroscopy has become much more widely practiced.

Technique

The sedation needed to perform enteroscopy may be slightly greater than that required for gastroscopy because the procedure takes longer and is more uncomfortable. In selected patients, enteroscopy can be performed without sedation. Because the bending section is longer, to allow increased angulation in all directions, and because the instrument is heavier, due to its increased length in comparison to a gastroscope, the instrument is slightly more difficult to handle during esophageal intubation and passage across the pylorus, but the push characteristics and increased flexibility allow for better handling in the jejunum.

A complete examination of the esophagus and stomach should be performed with the instrument before entering the duodenum since looping of the instrument in the stomach may produce red marks, usually on the greater curve, which are difficult to distinguish from vascular ectasias on pull-back. Reducing air nsufflation and consequent distention of the stomach by sucking out the air in the stomach before entering the duodenum may lead to deeper insertion of the instrument. The patient may feel discomfort as the instrument is advanced, distending the greater curvature of the stomach. Paradoxical or arrested tip motion occurs if the instrument loops within the fundus. By exerting pressure and clockwise torque, the endoscopist can generally push through the loop. Abdominal pressure on the left side of the upper abdomen can sometimes help reduce the loop forming in the stomach. As with endoscopic retrograde cholangiopancreatography (ERCP), pulling back once the endoscope has entered the second part of the duodenum will produce “paradoxical” advancement. The ligament of Treitz is usually encountered 85 to 110 cm from the incisors and usually requires full tip deflection to find the lumen. Once around the ligament, the first jejunal loop can be identified by a straight configuration, which points in a caudal direction if seen on x-ray. Buscopan or glucagon can be used to reduce peristaltic activity and may be useful during therapeutic intervention, but it is probably best to avoid administering either until the endoscope has reached as far as possible since peristalsis may help to advance the scope.

Overtube

A variety of methods have been used to try to reduce the tendency of the enteroscope to loop in the stomach. These include the use of abdominal pressure, internal stiffening devices, [1] and overtubes. Overtubes designed for enteroscopy have varied from 60 to 100 cm in length. Some have two ring-shaped metal tip markers to facilitate placement under screening and to prevent the overtube tip from being compressed. Others have a more pliable Gortex tip of 10 cm in length, which may limit mucosal trauma when advancing the overtube over the endoscope. The overtube is initially backloaded onto the shaft of the endoscope. Once the endoscope tip is within the jejunum, the overtube is advanced down the esophagus until its distal tip rests within the second portion of the duodenum for the 60-cm overtubes and to or beyond the ligament of Treitz for the 100-cm overtubes. Fluoroscopy is probably necessary if an overtube is being used since prepyloric placement does little to aid deep intubation. The softer Gortex-tipped overtube may buckle in the stomach and fail to provide sufficient rigidity. The endoscope should be lubricated and the overtube dipped in water before use. As the overtube is advanced, the endoscope shaft needs to be straightened by twisting the endoscope shaft clockwise while pulling it backward. (This is much the same maneuver as positioning a side-viewing duodenoscope within the duodenum during ERCP.) Overtube use is possibly responsible for a majority of the complications reported with enteroscopy. Several experienced enteroscopists have abandoned the use of the overtube, but it may sometimes be helpful in advancing the enteroscope further into the small bowel. Some commercially available enteroscopes are too floppy to be used without an

overtube.

Therapeutic Applications

Push enteroscopes include a standard 2.8-mm biopsy channel that has been used for several therapeutic functions within the proximal small intestine as well as for directed biopsy and delivery of x-ray contrast. [1], [2], [14], [15], [16], [17], [18], [19], [20], [21], [22], [23], [24], [25], [26], [27] and [28] The more the instrument becomes looped as it advances into the small bowel the greater the difficulty in getting instruments to emerge from the biopsy channel. This particularly applies to instruments with a long rigid tip such as a heater or bipolar probes and balloons, rather than snares and laser fibers.

Biopsy Endoscopic forceps–type biopsy has now largely replaced the use of a suction-fired biopsy capsule such as the Crosby capsule as a means of taking small bowel biopsies. The advantages of the endoscopic method are speed, ease, patient comfort, and reliability. Enteroscopic biopsies can reduce the chances of proximal duodenal inflammation and shorter villi in the bulb causing confusion when examining for celiac disease. Enteroscopic biopsy might offer advantages in the workup of patients with malabsorption, particularly if focal specific mucosal abnormalities have been seen in the proximal jejunum on a small bowel x-ray contrast series.
Jejunal Feeding Tube Placement Push enteroscopy has also been used in a variety of ways to place jejunal feeding tubes. Push enteroscopy has been used to carry a transgastric jejunal tube through a previous gastrostomy into the jejunum. [17], [18] Nasojejunal feeding tubes have been placed using the Seldinger technique. [20] The push enteroscope is placed into the jejunum initially and then a guidewire, such as a 0.35 mm-diameter 400-cm–long guidewire, is advanced through the endoscope's biopsy channel. The endoscope is removed, leaving the guidewire in place. A feeding tube is advanced over the wire and then rerouted from the mouth through the nose by passing it back through another tube passed through the nose and retrieved from the back of the pharynx. This method has been combined with a sewing method for attaching the jejunal feeding tube to the stomach to prevent displacement. [1]
Direct Percutaneous Endoscopic Jejunostomy Placement By using push enteroscopy, the "pull" technique for the placement of percutaneous gastrostomies has been extended to permit direct percutaneous jejunostomies. [1], [17], [20], [21] This technique has also been used to place jejunal tubes for small bowel enemas [22], [23] as well as to obtain cholangiograms in patients after Roux-en-Y hepaticojejunostomy. [1], [12], [23]
Polypectomy Polypectomy can be performed in the small intestine via the push enteroscope using snares. [1], [17] Surveillance of patients with a polyposis syndrome can be performed with push enteroscopy, allowing biopsy and polypectomy of larger lesions. Removal of large jejunal hamartomatous polyps in the Peutz-Jeghers syndrome has become part of the screening of this autosomal dominant condition with a malignant potential. The more proximal lesions are relatively easy to remove, but endoscopic removal is not yet possible for all lesions in the jejunum.
Diagnosis and Treatment of Gastrointestinal Bleeding The major indication for push enteroscopy lies in the evaluation and treatment of patients with obscure gastrointestinal bleeding. Reported yields have varied from 3% to 65%. Messer and colleagues [22] reported finding the bleeding site in 20 of 52 patients (38%) with obscure bleeding. Findings included angiodysplasias in 9 and small bowel tumors in 11. Foutch and associates [9] also reported a yield of 38% in their report of examinations of 39 patients. Angiodysplasias were the most common finding, accounting for 80% of the diagnoses. Davies and colleagues [1] reported finding a possible cause of bleeding in 45% of cases referred with occult bleeding. Chong and colleagues [24] reported finding a possible cause of bleeding in 64% of 55 patients using the newer push enteroscopes in combination with an overtube. In addition to diagnosis, push enteroscopy allows cauterization of bleeding sites. Using bipolar cautery, the Foutch group was able to fulgurate angiodysplasias in 11 of 12 patients. Control of bleeding was obtained in 8 of the 11 treated patients. Askin and Lewis [25] assessed the long-term effectiveness of push enteroscopic cauterization of bleeding arteriovenous malformations in 89 patients. Of these patients, 61 were cauterized and 28 were not. The group that was cauterized required significantly fewer blood transfusions than the noncauterized group. In another series [14] of 50 patients undergoing therapeutic push enteroscopy, bleeding was terminated in all patients with isolated angiomas and reduced in more than half of the patients with multiple jejunal angioma as well as in some malignant tumors. In addition, one bleeding duodenal ulcer, three gastric angiomas, linear erosions in a hiatus hernia, two Dieulafoy ulcers, and three watermelon stomachs were found, were treated with bipolar probe or laser, and had no further bleeding. [14] This group had a significantly reduced transfusion requirement when compared to their precauterization status. Heater probes and bipolar probes as well as monopolar probes and laser fibers are available in lengths to fit the newer push enteroscopes.

Complications

Complications are uncommon with push enteroscopy, which is probably nearly as safe as gastroscopy. Most of the complications appear to be associated with the use of a metal-tipped overtube. These include a Mallory-Weiss tear, pancreatitis most likely secondary to papillary trauma, a pharyngeal tear, and three cases in which long strips of gastric mucosa were torn off during advancement of the overtube. [2], [23], [26] These complications with overtubes occurred prior to the addition of a Gortex tip. Some series describe severe patient discomfort, which is usually due to stretching the stomach as a loop is formed along the greater curve while trying to transmit forward pressure to advance the tip in the small bowel.

SONDE ENTEROSCOPY

Sonde enteroscopy uses a long and extremely flexible fiberoptic instrument propelled through the intestine by peristalsis following inflation of a balloon at its tip. [29], [30], [31], [32], [33], [34] and [35] This instrument, in contrast to a push enteroscope, has no biopsy or therapeutic capability and no tip deflection. The endoscopic examination is performed during withdrawal. The instrument allows views of the distal jejunum and ileum, which cannot be reached by push-type enteroscopes. Mucosal visualization is somewhat limited due to the lack of tip deflection capability and occasional uncontrolled instrument withdrawal. Visualization is incomplete even when total small bowel intubation is achieved. Abdominal palpation, inflation and deflation of the tip balloon, and pulling or not pulling on the endoscope at the nose offer some degree of control over the intralumenal view. Lewis and Waye [29] estimated that approximately 50% to 70% of small bowel mucosa is observed during a standard examination. The ileum is reached in about 75% of examinations with 10% reaching the ileocecal valve. The concordance when sonde enteroscopy was compared with intraoperative enteroscopy performed with a colonoscope was 77% in 23 patients with bleeding.

Technique

Pioneers of this technique positioned a sonde enteroscope within the jejunum by passing the instrument transnasally, placing the patient in the right lateral decubitus position and following the patient with sequential fluoroscopy. Endoscopic techniques were subsequently applied to reduce the time the enteroscope took to pass through the stomach into the jejunum. [29] The sonde enteroscope is passed transnasally; then a gastroscope or push enteroscope, passed orally, grasps a suture tied to the tip of the sonde enteroscope with biopsy forceps while both instruments lie within the stomach. The push instrument is advanced into the small bowel, pulling the sonde enteroscope along. Once within the jejunum, the balloon of the sonde instrument is inflated, anchoring it in place as the push enteroscope is removed. Care has to be taken to prevent frictional force, exerted by pulling the larger scope backward, from pulling the sonde enteroscope through the pylorus. The advantage of this technique is that it permits total to near-total small bowel intubation within 8 hours and thus allows the procedure to be performed on an outpatient basis. The original technique averaged 24 hours. Withdrawal times range from 45 to 60 minutes. With present standard transnasal sonde enteroscopes, the new technique of placement has become generally accepted. Some centers use orally passed enteroscopes that are either allowed to pass naturally through the pylorus or are passed over a previously positioned guidewire. [29]

Video recording of the whole procedure with rapid playback helps to reduce the chances of missing lesions. Arranging for a nurse or assistant to visit the patient every 30 minutes to check that all is well and push 6 more inches of the enteroscope into the patient allows the enteroscope to be placed in the duodenum in the morning and removed in the afternoon. A sonde enteroscope is sometimes positioned immediately following push enteroscopy if the latter examination fails to identify the cause of obscure bleeding.

Diagnostic Capability

Sonde enteroscopy identified a small bowel site of bleeding in 33% of 60 patients with obscure gastrointestinal bleeding. [29] In a series of 504 patients with obscure gastrointestinal bleeding, combined push and sonde enteroscopy documented findings in 42% of patients; [31] 18% of the lesions were found in the region covered by push enteroscopy distal to the area examined by standard upper endoscopy and 26% were found in the remaining bowel examined by sonde enteroscopy. Vascular ectasias comprised 80% of the pathological abnormalities found overall, and small bowel tumors accounted for 10%. Several of the tumors discovered occurred in patients after a negative small bowel series. [33] Similar experience using sonde enteroscopy (without prior push enteroscopy) has been reported by Barthel and colleagues with a yield of 27.8% in 18 patients, [34] by Gostout and colleagues with a yield of 26% in 35 patients, [30] and by Morris and colleagues with a yield of 38% in 65 patients. [36]

Complications

The main drawback is the length and discomfort of the procedure, which makes it an unpopular examination with patients, nursing staff, and doctors. Perforations have been described due to balloon overinflation. [36]

INTRAOPERATIVE ENTEROSCOPY

With sonde enteroscopy not routinely available, intraoperative enteroscopy was probably the most common form of total small bowel endoscopic examination. [37], [38], [39], [40], [41], [42], [43], [44], [45], [46], [47], [48], [49], [50] and [51]

Technique

Colonoscopes are usually used for this examination, though a push enteroscope can also be used. The instrument does not need to be sterile, because the technique either involves peroral intubation of the small intestine, or the endoscope is placed into nonsterile bowel through an enterotomy, in which case it can help to place the endoscope in a sterile plastic sheath. It may be helpful to advance the endoscope into the proximal jejunum through the mouth prior to laparotomy, since once the abdomen is open it may be difficult to advance the instrument around the ligament of Treitz because the endoscope forms a loop along the greater curve of the stomach that is not constrained by the anterior abdominal wall. With oral intubation using an adult colonoscope, the endotracheal tube cuff may need to be deflated to permit passage of the wide caliber endoscope. Once the colonoscope has been positioned in the proximal jejunum, laparotomy is performed. It can be helpful to place a noncrushing clamp across the ileocecal valve to prevent distention of the colon with insufflated air because colonic distention can lead to difficulties with subsequent abdominal closure.

The examination is carried out by an endoscopist and a surgeon; the endoscopist looks through the endoscope using minimal air insufflation while the surgeon holds the endoscope tip inside the small bowel so as to inspect a short segment ahead of the endoscope. The surgeon then advances the endoscope into another segment. The view is best seen by dimming the overhead lights, also allowing the surgeon to look at the transilluminated bowel, which can be helpful in finding angiodysplasia. Once examined both internally and externally, the small bowel is pleated onto the shaft of the endoscope and the next section of bowel is examined. Care has to be taken not to overstretch the mesentery on colonoscopes, which are rather stiff for this use. [27] Active bleeding within the small bowel may diminish the effectiveness of this examination. Examination has to be performed during intubation and not on pull-back since mucosal trauma occurs with the pleating-causing artifact that may be confused with the appearance of angiodysplasia. [37] A technique of “reverse transillumination” to help differentiate trauma from vascular ectasias in this setting has been described. [38] This may allow visualization of the feeding vessel of the vascular ectasia. Using nonvideo endoscopic instruments, the endoscopic light source is turned off and the operating room lights are focused on the intestine. Abnormalities identified at enteroscopy are marked with a suture placed on the serosal surface of the small intestine. At the end of the examination, the endoscope is withdrawn and the surgeon can resect the abnormalities identified by the sutures.

An alternative technique is to perform an enterotomy through which a sterilized endoscope or an endoscope covered by a sterile plastic sheath is placed. The enterotomy is generally performed in the mid–small bowel, allowing both proximal and distal intestinal intubation, but it can be performed in the cecum if vascular ectasias are suspected in the cecum or distal small bowel. Retrograde intubation of the small bowel can be performed after the cecum has been inspected. The colonoscope is advanced by the surgeon through the ileocecal valve and up the small intestine.

Diagnostic and Therapeutic Use

Intraoperative endoscopy has been used for a variety of indications [37], [38], [39], [40], [41], [42], [43], [44], [45], [46], [47], [48], [49], [50] and [51] but is probably the endoscopic method most widely used in identifying small intestinal sites of bleeding. [37], [38], [39], [40], [41], [42], [43], [44], [45], [46], [47] and [48] Intraoperative enteroscopy can be successful in identifying the site of blood loss in selected patients with reported yields of 83% to 100%. Vascular ectasias are the most common nonpalpable cause of bleeding, but radiation enteritis, ulceration, malignancies, strictures, and polyps (e.g., multiple polyps in Peutz-Jeghers syndrome) [49], [50] and [51] may require endoscopic identification. Hemostasis can be achieved at intraoperative enteroscopy either by resection of the bleeding lesion or by transendoscopic ablation of the bleeding lesion, usually with a thermal endoscopic method.

In one study involving 71 patients, laparotomy and intraoperative enteroscopy was more successful in defining the source of gastrointestinal bleeding. [38] Preoperative sonde-type enteroscopy may be most useful in recognizing patients with diffuse vascular ectasias in order to avoid operating on such patients.

Complications

Prolonged ileus, sepsis, and perforations (the endoscopist and surgeon should listen for a hissing sound that indicates that perforation has occurred) are common sequlae of operative enteroscopy.

WIRELESS CAPSULE ENTEROSCOPY

Small bowel endoscopy is especially limited by problems of discomfort and failure to advance enteroscopes far into the small bowel. The invention of the transistor made the design of electronic swallowable radiotelemetry capsules possible for the study of gastrointestinal physiological parameters. These capsules were first reported in the late 1950s and included measurements of temperature, [52] pressure, [53] and pH. [54] The development and testing of a new type of radiotelemetry capsule endoscope, which is small enough to swallow (11 × 30 mm) and has no external wires, fiberoptic bundles, or cable has been described and tested [55] (Fig. 139-2). By using a short focal length lens and a CMOS—complementary metal oxide silicon chip—images are obtained as the optical window of the capsule sweeps past the gut wall, without requiring air inflation of the gut lumen. [56], [57]

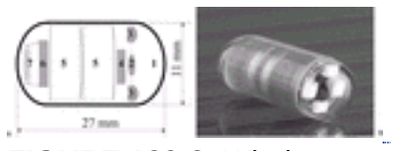

FIGURE 139-2. Wireless capsule endoscope **A:** Schematic diagram. *1*, Optical dome; *2,* Lens; *3*, Illuminating light-emitting diodes (LEDs); *4*, Complementary metal oxide silicon chip (CMOS) imager; *5*, Battery; *6*, ASIC transmitter; *7*, Antenna. **B:** Photograph.

The capsule endoscope is propelled by peristalsis though the gastrointestinal tract and does not require a pushing force to propel it through the stomach, small bowel, or colon. Because the gut is a hollow tube it is unimportant whether the capsule points forward or backward as it passes through.

The video images are transmitted using radiotelemetry (operating in UHF band) to an array of aerials attached to the body. The aerial belt allows image capture and enables the continuous triangulation of the capsule location in the abdomen so that the whole trajectory can be shown on the workstation monitor to help in locating pathologies detected by the capsule imager. The images are stored on a small portable recorder carried on the belt and are subsequently downloaded for analysis. The system allows more than 8 hours of continuous recording of images of the gastrointestinal tract. The patient need not be confined to a hospital or clinic environment during the examination and is free to continue his or her daily routine.

The first human volunteer [55] and patient [58] studies revealed that the capsule was easily swallowed and caused no discomfort. The capsule, propelled by peristalsis, successfully reached the cecum in less than 2 hours. The image window remained clear throughout all of the transmissions with transmission times of up to 6 hours.

Trigonometric analysis of signal strength allowed continuous monitoring of anatomic position of the capsule endoscope.

There is the potential to link this device with radiotelemetry signals indicating the presence of pH, *Helicobacter pylori,* or other relevant physiological or pathological parameters, biosensors, or optical biopsy methods. It is likely that a similar device will form the basis of any robotic attempts to move imaging devices by remote control in the human body. The capsule may provide a better method to examine the small bowel, especially in patients with recurrent gastrointestinal bleeding. Provisional results suggest that this device is nearly as sensitive and specific as push enteroscopy in a randomized study. [59] Unlike all other forms of nonsurgical enteroscopy it can supply images painlessly from the entire small intestine.

CONCLUSION

Minor technical advances in endoscope design and increased use by clinicians have resulted in enteroscopy becoming more useful for diagnosis and therapy. There are still limitations, both in viewing and treating the small bowel, particularly from the distal jejunum to the proximal ileum; these require innovative engineering and clinical understanding if solutions are to be found that will allow enteroscopy to be as successful as upper gastrointestinal endoscopy and colonoscopy have become.

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CHAPTER 140

Jerome D. Waye and Christopher B. Williams

COLONOSCOPY AND FLEXIBLE SIGMOIDOSCOPY

INSTRUMENTATION

Flexible fiberoptic technology began to be used for visualization of the colon several years after fiberoptic gastroscopy was introduced in 1957. The first commercially available fibercolonoscope, the Overholt Coloscope appeared in the early 1960s. Modern colonoscopes, whether fiberoptic or video, give a brilliant, high-resolution, color view of the mucosa through wide-angle (140-degree) optics. The 20- to 30-fold magnification at 5-mm distance rivals the close-up view of a dissecting microscope, but colonoscopes provide excellent distant views as well. Tip angulation in four directions to 160 to 180 degrees allows angulation around most acute flexures. Retroflexion (i.e., J configuration) is possible in the proximal colon or rectum, thus minimizing the risk of missing significant lesions in capacious areas that could not be seen with older, less maneuverable instruments. The length of the instruments varies from the 65-cm flexible sigmoidoscope to the 165-cm colonoscope. The instruments range in shaft diameter from 1-cm pediatric colonoscopes to 15-mm adult instruments. Instrumentation and suction channel sizes range from 2.7 to 4.2 mm in diameter. In most ways, a colonoscope is similar in design to a gastroscope, but it has a more flexible shaft to accommodate the various colonic bends encountered. Smaller-diameter pediatric colonoscopes or standard gastroscopes, which are more maneuverable in the colons of babies and small children, also have a place in examining ileostomies and fixed or strictured adult colons. Their ability to retrovert in the capacious portions of the colon can also make some difficult polypectomies easier. Zoom colonoscopes have the capability of image magnification to levels similar to a dissecting microscope. Enhanced maneuverability is possible with an instrument of variable stiffness. [1] High-resolution optical chips afford greater definition of the visual image.

A full range of endoscopic accessories is available for colonoscopes, including biopsy forceps, electrocoagulating hot-biopsy forceps, cytology brushes, washing and spraying catheters, sclerotherapy needles, a range of dilating balloons and bougies, and various polypectomy snares and retrieving devices. Other standard techniques can be applied, such as the introduction of balloons or tubes over a guidewire and insertion of self-expanding stents for obstructing cancer. Thermal therapeutic devices such as the heater probe, bipolar electrocoagulation, argon plasma coagulator, or laser are as applicable to the lower gastrointestinal (GI) tract as they are to the upper GI tract. Some endoscopists use an overtube as a splinting or stiffening device to control looping in the sigmoid colon or as a temporary stent to facilitate rapid withdrawal and reinsertion of the colonoscope.

Blind rectal ultrasound probes can assess invasion in tumors of the anorectal area. Direct-vision, ultrasound-bearing colonoscopes may also have a role in tumor staging. Small ultrasound probes may be passed through the colonoscope's instrument channel to enable ultrasound as desired at any point throughout the colon [2] (e.g., before removal of sessile lesions, to check the relation of the colon wall to surrounding organs before electrocoagulation).

ANATOMIC BASIS OF COLONOSCOPY

The ease or difficulty of colonoscopy in a given patient is related to the degree to which the supporting mesenteries fuse to the posterior abdominal wall during fetal development. Although the descending and ascending colon become fixed retroperitoneally, in about 10% to 15% of patients they remain mobile on mesocolons similar to the normal anatomy of the sigmoid and transverse parts of the colon. The consequence for the endoscopist is that these colons are mobile and can move around freely and unpredictably within the abdomen, being uncontrollable by the various straightening or rotation maneuvers that are effective with conventional colonic anatomy.

There are no fixed anatomic landmarks between the anus and the ileocecal valve, but there are geographic clues which often are helpful (but may also be misleading). At about 17 cm, the colonoscope leaves the capacious rectum and enters the narrower, extraperitoneal part of the sigmoid colon (Fig. 140-1). The sigmoid has frequent circular haustral infoldings—which may be especially exaggerated with diverticular disease—into which the instrument tip impacts if steering is not accurate.

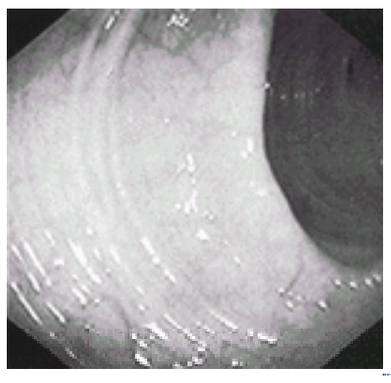

FIGURE 140-1. In the sigmoid colon, the circular outline with the light reflex over arcs of circular musculature clearly indicates the center of the lumen for close-up endoscopic steering purposes.

In contrast to the zigzag path of the sigmoid colon, the descending colon is usually a short, tubular segment. An acute angulation of the instrument tip at the junction of the sigmoid and descending colon may afford a clear view of the tubular descending colon, but forward progress may be impeded by loops in the instrument caused by the tortuous and convoluted sigmoid colon.

The transverse colon (Fig. 140-2; see also Color Fig. 140-2) frequently shows a triangular configuration as a result of its three longitudinal muscles, or teniae coli, that act as tethering boundaries to air distention. The liver is a fairly predictable landmark with the flat, bluish-gray surface and sharply defined edges caused by the balloonlike, air-filled colon contacting the hepatic surface (Fig. 140-3; see also Color Fig. 140-3).

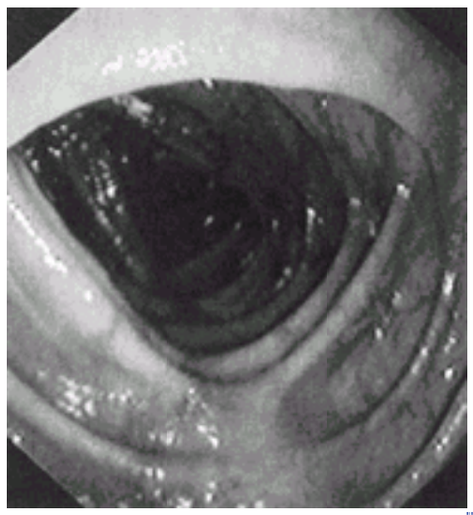

FIGURE 140-2. (See Color Fig. 140-2.) Transverse colon shows the characteristic triangular outline resulting from the relative thickness of the three longitudinal teniae coli. The circular muscle is much thinner.

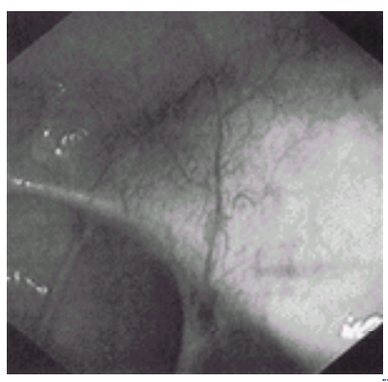

FIGURE 140-3. (See Color Fig. 140-3.) Hepatic flexure shows the dark impression of the liver. Similar discoloration can be caused by other extracolonic viscera at the splenic flexure or in the distal colon. Note the longitudinal impression of a tenia and the transverse haustral folds.

Upon entering the ascending colon, the superior lip of the ileocecal valve may appear in the distance as a short flat segment on the most proximal circular haustral fold (Fig. 140-4; see also Color Fig. 140-4). The upper lip may have a notched bulge or may appear thickened by fat accumulation. Most often the opening into the ileum is slitlike on the inferior side of the fold. The appendiceal orifice, a less definite landmark, is seen as a crescent-shaped slit or a circular convolution of folds at the blind saccular cecal pole (Fig. 140-5; see also Color Fig. 140-5). Multiple, pale reddish, 1- to 2-mm circular halos are often seen in proximity to the appendix when using the videoendoscope. These represent lymphoid follicles. [3]

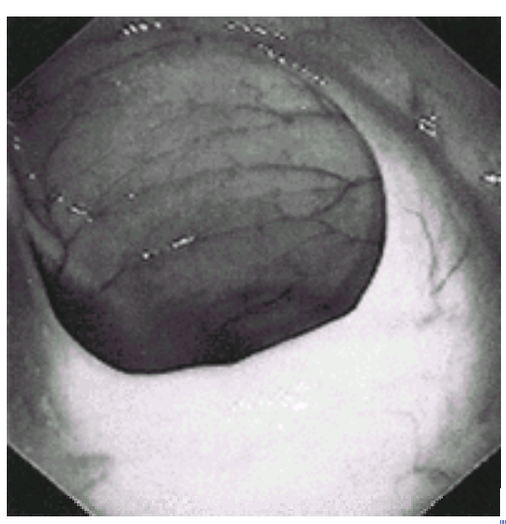

FIGURE 140-4. (See Color Fig. 140-4.) Ileocecal fold shows the characteristic notch on a flattened fold. The notch denotes the superior lip of the ileocecal valve. The opening is just proximal to and below the notch, and cannot be seen without marked angulation of the instrument tip.

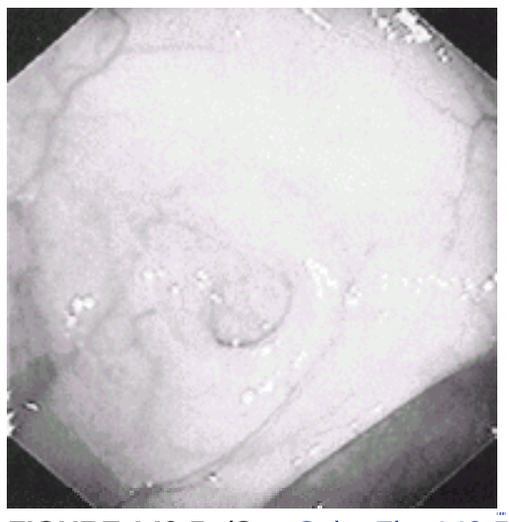

FIGURE 140-5. (See Color Fig. 140-5.) The appendix orifice is variable in configuration. Here, the orifice is round, but it may also be seen as a crescent-shaped slit. It is usually rather insignificant in appearance. The internal aspect remains the same after appendectomy.

The ileal mucosa has a granular appearance when the ileum is air-filled, and short frondlike villi are visible in pools of fluid. The granular appearance is caused by multiple light reflections from the individual villi. One- to 3-mm nodules of lymphoid tissue, sometimes aggregated into 10- to 15-mm plaques of Peyer patches are normal in the terminal ileum, and particularly prominent in children. By contrast, the normal colorectal mucosa is smooth and shiny, although with deflation, fine transverse innominate grooves may show, especially with surface dye spray.

Throughout the colon, a fine, interlacing, vascular pattern is visible through the transparent overlying epithelium. When the normal vascular pattern is hidden by mucosal congestion, visible as diffuse redness of the surface, a pathological process must be considered, but biopsies are needed to differentiate between inflammatory change and traumatic or reactive hyperemia. Opacity of the surface can also be caused by thickening that occurs after chronic colitis or by edema. In the rectum, the vascular pattern is more pronounced, with larger vessels. Hemorrhoidal veins may appear wide and tortuous. A U-turn (i.e., retroflexion) maneuver in the rectum should always be attempted to afford a view of the rectal ampulla, the dentate line (Fig. 140-6; see also Color Fig. 140-6), and internal hemorrhoids, [4] but the rigid anoscope gives a superior view of this area by dilating it maximally.

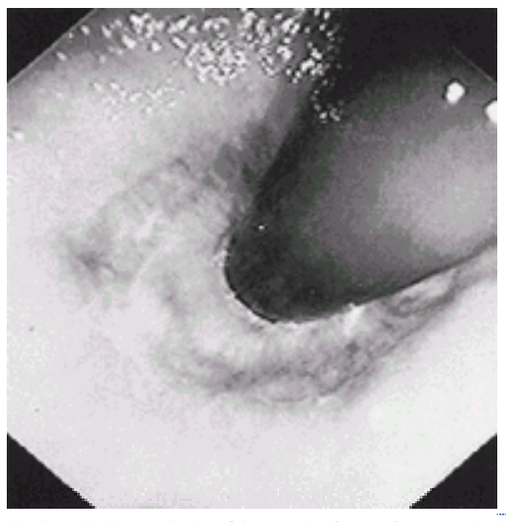

FIGURE 140-6. (See Color Fig. 140-6.) Retroversion of the endoscope in the rectum is a useful maneuver to find or remove small polyps in the distal part of the rectal ampulla. The dentate (pectinate) line is serpiginous at the junction of the squamous mucosa of the anal canal and the columnar rectal mucosa.

Localization During Colonoscopy

The position of the colonoscope can be approximately judged by a combination of factors: the length of instrument inserted, the characteristic appearances of certain areas, including the sharply defined blue color of the liver, the fluid-filled splenic flexure area, and the triangular, air-filled, transverse colon. Most instruments are bright enough to transilluminate the abdominal wall where the bowel approximates to the surface at the splenic flexure (left midaxillary line), transverse colon, or cecum. Fluoroscopic guidance is rarely used, but some find it helpful in the endoscopist's learning phase and in a few difficult cases.

The magnetic imager [5] is a new localizing device whereby small electromagnets can be inserted into the colonoscope shaft, either during manufacture or with a catheter-type wand. External sensors can detect the energy and plot an image of the exact shape and location of the endoscope. There is no radiation, and the computer-generated image is similar to that obtained fluoroscopically. Alternatively, the transabdominal impression of the endoscopist's palpating fingers may often be seen from within. Transillumination or endoscopically visible indentation by finger palpation in the right iliac fossa, however, does not necessarily indicate that the tip of the instrument is located in the cecum, because the instrument tip may stretch almost any portion of the colon into the right lower quadrant during colonoscopic intubation. For instance, mobile portions of the bowel, such as the sigmoid or the midpoint of the transverse colon, can be pushed toward the right iliac fossa.

All localizing judgments that depend on landmarks or palpation can be mistaken. For instance, using the length of scope inserted, in a long or mobile colon, 80 cm of shaft may be looped in the proximal sigmoid; whereas in the same patient during withdrawal, the instrument may be straightened so that the splenic flexure is 40 cm from the anus, and the hepatic flexure (or even the cecum) can be reached with 50 to 60 cm of instrument. The only definitive landmark is the ileum seen through the ileocecal valve, but even this may be difficult to locate, and the valve can be effaced after severe or long-standing inflammatory disease.

Because of the absence of absolute landmarks throughout the colon, even if the endoscopist has reported success in total colonoscopy with identification of the ileocecal valve and cecal pole, there is still the possibility of being grossly mistaken about the location of the instrument tip and in the anatomic localization of pathological findings. [6] Even experienced endoscopists can occasionally mistake the splenic flexure for the hepatic flexure or the hepatic flexure for the cecum, and thereby mislead the surgeon as to the site of an obstructing tumor. [7]

In the absence of fluoroscopy, any lesion seen by colonoscopy can be marked with an intramural deposit of carbon particles using sterilized india ink or a newly developed pure carbon suspension, injected through a long, flexible, injector needle, either for subsequent surgical localization or to enable the endoscopist to find the area on a subsequent examination. [8] A gray-blue, submucosal stain of carbon permanently remains at the injection site. Injection of vital dyes such as methylene blue, indocyanine green, or indigo carmine are rapidly absorbed, disappearing within hours or days. [9] It is possible to mark the proximal extent of tip progress with metal clips, [10] but this technique is often not clinically useful.

BOWEL PREPARATION

Total colonoscopy is rarely possible in the unprepared colon. Patients with profuse watery diarrhea are sometimes mistakenly thought not to require preparation and are often found to have an opaque mucosal coating of fecal material that obscures adequate view of the surface. Even patients with a surgically defunctionalized bowel produce inspissated mucoid residue and require cleansing. A patient with massive colonic bleeding may effectively cleanse the colon of fecal material, but often a residual coating of blood obscures mucosal events during the colonoscopic examination.

Limited Preparation

When it is necessary to examine only the distal colon segments, an adequate preparation can consist of one or two evacuant enemas of tap water, saline, or, more usually, a small-volume hypertonic phosphate or other proprietary enema, given 15 to 60 minutes before the examination begins. This allows enough time for proper bowel clearance but is not so long that proximal contents move distally. Limited bowel preparation assumes normal colonic ability to empty the whole left colon rapidly by mass action. This may not happen in patients with diverticular disease or strictures. In these individuals, even limited examination must usually be preceded by full bowel preparation.

Full Preparation

A wide variety of bowel preparations are available. [11] Ideal attributes for bowel preparation include pleasant taste and avoidance of excessive volume, cramps, or the need for enemas. Certain modifications of medication and diet are necessary. Oral iron medications must be stopped 4 to 5 days before colonoscopy because the formation of organic iron tannates in combination with dietary green vegetable residue renders colon contents black, sticky, and difficult to clear. Constipating agents such as codeine or loperamide should be discontinued 12 hours before colonoscopy. It has been suggested that aspirin and other antiplatelet drugs or anticoagulants be discontinued 7 days before examination to reduce the risk of immediate or delayed bleeding should polypectomy be performed, but this may not be necessary since clinical experience has not borne out any excess bleeding risk in patients on aspirin. [12] No matter what the preparation regimen, colon cleansing is enhanced by a clear or full-liquid diet for 24 to 48 hours before the examination.

Purgative Regimens

Purgative regimens are still the most commonly used form of bowel preparation. Their advantage is that the colon, after a contact laxative such as senna, or bisacodyl, is stimulated to evacuate most of the bowel contents. Disadvantages are that the evacuation stimulus can cause severe cramping abdominal pain, that the cecal region is often poorly cleared, and that some solid residue often remains distally. Therefore, enemas or oral osmotic agents may be required to supplement the cleaning effect of the purgative. More recent bowel cleansing methods do not require enemas. [13]

Osmotic purges can be used as an alternative to enemas, either adding to or supplementing the oral purgative with up to 2 L of an agent such as magnesium salts (e.g., citrate, sulfate) or a nonabsorbed carbohydrate (e.g., mannitol, lactulose, sorbitol). Magnesium sulfate has strong purgative and osmotic effects but also an unpleasant taste; the milder but citrus-flavored magnesium citrate is generally preferred. Mannitol has been the most widely used of the carbohydrate solutions. Its sweet taste can be reduced by cooling, and the likelihood of nausea is prevented by preadministration of an oral antiemetic prokinetic agent such as domperidone or metoclopramide 30 minutes before administration. Five percent mannitol is isotonic, and 2 L are drunk slowly over 1 hour or more. The use of mannitol or other carbohydrate preparations risks explosion during electrosurgical procedures because of the production of hydrogen gas resulting from fermentation by intestinal bacteria. This risk can be effectively eliminated by insufflation of carbon dioxide during the procedure or by meticulously aspirating gas and reinsufflating air before electrosurgery. A low-volume sodium phosphate preparation is commonly used for endoscopy preparation and has been well received by patients. [14] Taken orally the evening before and 4 hours prior to colonoscopy, 45-mL aliquots of sodium phosphate produce a vigorous catharsis. [15] Cleansing enemas are not required. Fluid balance shifts, absorption of sodium, or hyperphosphatemia may cause deleterious effects, especially in the presence of cardiac or renal disease. The noxious taste may be circumvented by dehydrated sodium phosphate in pill form. [16]

Balanced electrolyte solutions, administered in a volume of 4 L orally or by nasal tube, result in an acceptable bowel preparation. The salty, oily taste and required large volume cause about 10% of patients to stop drinking the solution, thereby failing to prepare their colon adequately. Newer formulations with flavoring have improved palatability and may improve patient compliance. Although packaged instructions advise minimal dietary restrictions, better results can be expected from a 24-hour full liquid diet. Simple saline solution (0.9% sodium chloride) is effective and arguably safe for preparation of the majority of patients, but potassium losses and electrolyte fluxes can be avoided by appropriate additional ingredients.

The regimen chosen for whole-colon preparation necessarily varies according to circumstance, patient preference, and clinical indication. Patients need to sleep and must be able to travel without fear of incontinence, so the timing of preparation must also be adjusted according to the time of examination and travel arrangements. [17] Frail, ill, and elderly patients may require modified preparation, sometimes on an inpatient basis if cooperation is uncertain. If obstruction is a possibility, oral preparations may be dangerous or at least should be spread out over a longer period. In almost all circumstances, a patient fit for total colonoscopy is fit for full-bowel preparation. Compromise in reducing the regimen may result in poor preparation and in only being able to perform a limited examination or a less accurate colonoscopy.

Whichever of the different purgative regimens is chosen, it must cause profuse, clear, fluid diarrhea. A more vigorous preparation may be required for the patient with chronic constipation or severe diverticular disease or a patient whose bowel was not properly cleansed with a previous regimen. It is desirable that patients continue to drink fluids up to the time of the examination, and ingest at least some of the preparation fluid within a few hours.

Medication

Sedation Sedation is unnecessary for some examinations, especially those that are limited, or those in patients after sigmoid colon resection or in whom a previous procedure is known to have been easy. A great deal also depends on the patient's attitudes and on local custom. Endoscopists in some countries rarely sedate, whereas those in other countries almost always do. Some routinely use general anesthesia administered by an anesthesiologist, [18] although this may add to the risk of complications by removing feedback from the patient relating to pain from loops or difficult electrosurgical procedures. Using less sedation makes for easier change of position and greater safety. Using no sedation allows the patient to leave rapidly and unescorted after the procedure. Endoscopists who never use sedation may have a lower success rate in total colonoscopy. [19] Some stretching of peritoneal attachments is inevitable, at least transiently, during colonoscopy. This causes gnawing or acute unpleasant visceral pain. Coupled with air distention and the sensation of rectal fullness simulating the desire to defecate, this can make a slow examination difficult to bear for some patients who are mildly or not sedated, although others tolerate it easily. Pain during colonoscopy can usually be rapidly diminished or eliminated by reducing the loops responsible for stretching the mesentery, or by aspirating air, but sometimes further doses of analgesic agents are needed. Normal diet, activities, and medication can usually be resumed as soon as the mild stress of the examination and the effects of medication are over.
Antispasmodics Antispasmodics (glucagon, 0.5–1 mg IV; hyoscine- *N*-butyl bromide, 20–40 mg IV) are thought by some to increase the difficulty of insertion by rendering the colon atonic and therefore more likely to stretch and loop, but they eradicate circular muscle spasm and may increase the accuracy of examinations for small lesions such as polyps or angiodysplasias. [20] Some endoscopists use antispasmodics routinely.
Antibiotics A prosthetic heart valve, history of endocarditis, surgically constructed or congenital systemic-pulmonary shunt, or synthetic vascular graft less than 1 year old places a person at high risk for infection with various procedures. Even in these persons, however, there is insufficient data to strongly recommend antibiotic prophylaxis for colonoscopy with or without biopsy or polypectomy. [21] Antibiotics are not recommended for colonoscopy with or without biopsy or polypectomy in patients with cirrhosis and ascites, immunocompromised patients, those with prosthetic joints or pacemakers, or patients with a history of rheumatic valvular dysfunction, mitral valve prolapse with insufficiency, hypertrophic cardiomyopathy, or for most congenital cardiac malfunctions. If antibiotics are deemed necessary, the usual doses are ampicillin 1 g and gentamicin 80 mg IV 10 to 30 minutes before the procedure.

COLONOSCOPY PROCEDURE

Position and Position Changes

The left lateral position is most commonly used for flexible sigmoidoscopy and colonoscopy, mainly because it gives convenient access to the perineum for insertion. However, colonoscopic examinations can be, and in some centers are, performed in other positions, including lithotomy, knee-chest/elbow, and right lateral positions. Especially if a large-volume fluid preparation regimen has been used, position changes can modify both the configuration of the colon and of the air and fluid within it because of the effects of gravity. When forward progress can no longer be accomplished, a change in the patient's position will aid tip advancement in two thirds of cases. [22]

Insertion of the Endoscope and Flexible Sigmoidoscope

Digital examination of the anorectum is used to lubricate the area generously and to palpate any lesion. A local anesthetic gel may reduce anal discomfort. When

there is rectal stenosis, a fistula, or abnormal anal sensitivity, it may be desirable to use a small-diameter pediatric instrument or a gastroscope. The technique for flexible sigmoidoscopy is the same as for the first part of a colonoscopy, except that it is less important to avoid looping, because the proximal colon is not to be intubated. Characteristically, there is a red-out when the instrument enters the rectum, because the tip abuts on the rectal mucosa. Simultaneous air insufflation and instrument withdrawal are required to disimpact it; and angulation or rotational movements are useful to find the rectal lumen. Movements of the instrument tip should thereafter be slow and deliberate, taking care to steer toward the lumen before pushing inward, so as to avoid constantly losing the view. When the lumen cannot be seen, continuing to push the instrument shaft into the rectum stretches the sigmoid colon into unnecessary and painful loops and rarely results in tip advancement. Once impacted against a haustral fold the tip tends to remain stationary while the rest of the scope flexes into a loop.

Slide-by is a term describing the maneuver when the tip is against the mucosal surface, no part of the lumen can be seen, and the instrument is pushed forward causing the mucosal vessel pattern to visibly slide across the lens. This maneuver should be avoided but is permissible for short distances and only when the instrument is known to be pointing in the correct direction; if tip movement stops and the mucosa blanches, additional pressure risks perforation, and the instrument must be withdrawn to locate the lumen. Withdrawal and disimpaction of the tip is a routine procedure that should be performed every time the view is lost for more than a few seconds.

Diverticular disease is often associated with thickened rings of circular muscle or fixation of the sigmoid colon by pericolic adhesions from previous inflammatory episodes. The resulting distortion and angulations can make it difficult to locate the lumen and coax the instrument through, because the tip is not free to maneuver normally. In a few cases, the angulations are so acute that a small-diameter pediatric colonoscope or a standard gastroscope with the capability for greater tip deflection may allow passage through diverticular disease where the standard colonoscope cannot be passed. Despite the technical problems in intubation, endoscopic examination of diverticular disease can be clinically valuable, mainly because the distorted appearances seen on barium enema may make it impossible to exclude malignancy on radiologic grounds. Similar bowel fixation and angulation may occur after hysterectomy as a result of postoperative adhesions between the sigmoid colon and anterior aspect of the pelvic cavity.

If any pathology is seen during flexible sigmoidoscopy, a biopsy can be easily and safely performed. However, electrosurgery for polyp removal is contraindicated after the limited bowel preparation for flexible sigmoidoscopy because of the significant hazard of explosive gas mixtures.

Total Colonoscopy

The fundamentals of insertion technique are the basis of rapid, relatively pain-free, accurate total colonoscopy. Difficulty during intubation of the proximal colon or passage around the hepatic flexure is invariably related to failure to straighten the colonoscope in the distal colon before attempting the last part of the insertion. Looping in the sigmoid colon is a continual and recurrent problem throughout a colonoscopy but especially when it is necessary to push harder to transmit force around a looped colonoscope to reach the cecum.

During a colonoscopy, there are only a limited number of options available to the endoscopist. These include pushing the instrument in or pulling it out; twisting the shaft to the right or the left; insufflation or withdrawal of air; using tip controls to steer up, down, right, or left; use of abdominal pressure to reduce loops; and changes of patient position. The difference between the slow endoscopist who reaches the cecum infrequently after a traumatic examination and a fast endoscopist who examines the whole colon in over 95% of patients within an average time of 10 minutes is the ability to use the various options logically, quickly, and sequentially without repeating a nonproductive maneuver over and over again.

During the intubation phase, the instrument controls must be continuously manipulated using torque to keep an adequate view of the lumen while the endoscopist concentrates on visual detail to locate and find the direction when it is briefly lost, and backs off quickly if the tip impacts into a bend or fold. The endoscopist can feel when the instrument is passing freely and can be advanced or when it is becoming snarled and should be withdrawn before pushing in again. The biggest mistake of the inexperienced endoscopist is to think that a good view of the lumen is an unqualified signal to push, especially when looped in the sigmoid with a view of the descending colon; pushing under these conditions often results in an expanding sigmoid loop with resultant pain but without tip progress.

Some sigmoid loops, such as the spiral a-loop configuration that may form in the sigmoid colon, are favorable, because they allow the instrument to proceed into the descending colon without forming acute angulations; others, such as the N-loop, which more frequently occurs in the sigmoid, put the tip into an acute angulation at the descending colon junction, making this area difficult and painful to pass through. Fluoroscopy is not necessary to ascertain the presence of a loop, because it is apparent that a loop is forming whenever the shaft is pushed in more than the tip advances. Additionally, once a loop is stretched on its mesentery, additional pushing hurts the patient. Conversely, when the colonoscope shaft is straight, with the colon shortened or pleated over it rather than stretched, only a light touch is needed on the instrument with 1:1 correspondence between shaft and tip during push-pull and rotational movements. When there are no excessive loops, tip angling using the dial controls is also easy and full. Conversely, when the tip is maximally deflected using both control knobs and the lumen is at an acute angle, this indicates that the scope is in a loop configuration. The best maneuver is to withdraw and straighten the shaft, and if that cannot be accomplished, short in-and-out thrusts of the scope often result in forward tip movement.

Because of the multiple bends, folds, and twists of the large bowel, it is inevitable during a colonoscopy that the instrument will loop or start to loop on many occasions. After each advance, by pulling back and restraightening the shaft, the colon is effectively shortened and pleated over the instrument, which is progressively advanced on a “three-steps-forward-and-two-steps-back” basis.

It is usually not possible to keep the instrument straight or the patient completely comfortable all the time because of the unpredictability of the sigmoid colon. There may be a period of difficulty or looping before the tip reaches the fixed retroperitoneal part of the descending colon or the splenic flexure, where the shaft can be fully straightened by pulling back with clockwise rotation to only 50 cm from the anus. Unfortunately, because of the variability of the anatomy and attachments of the sigmoid and descending colon, this first part of a colonoscopy is the most difficult part of the procedure.

Passing the splenic flexure into the transverse colon is usually easy if the sigmoid has been straightened and can be held straight. After maximal shaft withdrawal, the combination of gentle shaft advancement with clockwise twist maintained, simultaneous abdominal pressure over the sigmoid, and avoidance of tip overdeflection usually allows progress into the transverse colon. Overaggressive pushing tends to reform the loop in the sigmoid, and overangulation impacts the tip into the flexure (i.e., the inverted “J” or “walking stick handle” effect). If, after several attempts, the tip does not advance, the instrument should be withdrawn and restraightened; rotating the patient into the right lateral position almost invariably causes the splenic flexure to drop downward, allowing the instrument to slide more easily around to the midtransverse colon. With the patient in the right lateral position, the midtransverse colon often sags down into a rather acute bend, providing a good point at which to withdraw and restraighten the colonoscope once again and also to rotate the patient onto the back or the left lateral position before insertion into the proximal colon.

In the proximal transverse colon or hepatic flexure region, and thereafter in the even more capacious ascending colon, deflation is extremely helpful in advancing the instrument and shortening the distance to the ileocecal region. Aspiration of air causes the colon to deflate concentrically so that the diameter is smaller, but it also results in shortening the colon longitudinally. A few additional maneuvers may be helpful in assisting total intubation, such as rolling the patient into the prone or supine position or asking the patient to inspire to lower the diaphragm. The more difficult it is to reach up or around the hepatic flexure and down to the cecal pole, the less likely it is that aggressive forward pushing will work. The longer and more mobile the colon is, the more certain it is that force will generate loops that will absorb all the motive power applied. With attention to technique and patience, even a redundant colon often shortens to only 50 to 60 cm at the hepatic flexure, permitting the tip to slip down the last few centimeters with ease. Advances in technology have permitted electromagnetic imaging devices to visualize, in real time, the three-dimensional localization of the colonoscope without x-ray involvement, which may significantly change current practice. [5]

Ileocecal Region

Even experienced endoscopists can be mistaken in thinking that a capacious hepatic flexure is the cecal pole, or that tonic contraction of the ileocecal fold represents the appendiceal orifice. The appendiceal orifice is seen often merely as a crescent-shaped slit or a circular convolution of folds. When a crescent orifice is seen, the apex of the crescent almost invariably points toward the ileocecal valve. With the tip in the cecum and the crescentic orifice clearly visible, deflection of the tip in the direction indicated by the apex of the curve will align the scope with the ileocecal valve orifice. Withdrawal of the shaft and deflection of the up/down control dial in the direction of the appendix curve when the first large fold is encountered will often result in ileal intubation. Alternatively, withdrawing the colonoscope 10 cm or so back up the ascending colon and looking for the telltale inward bulge of the valve on the ileocecal fold, which is the first encircling fold back from the cecal pole, is another method to permit the endoscopist to determine where to angulate and enter the ileum. Even so, ileal intubation can be difficult and may require several minutes combined with skill and patience. The anatomy is variable, and both colon and colonoscope can rotate or move during attempted insertion. Some ileocecal valves

show prominent pouting lips; others, however, are the merest slit on the reverse side of the ileocecal fold, which may require visualization by attempting retroversion of the bending section of the colonoscope within the cecal pole.

Terminal Ileum

A skilled endoscopist can enter the terminal ileum with reasonable ease in approximately 80% of cases [23] and can almost invariably obtain a blind forceps biopsy through the valve in the remaining patients. Failures arise mainly when deformity or scarring make angulation impossible or the opening too narrow, but poor bowel preparation and loops in the instrument can be contributory problems. Endoscopic assessment of the mucosa of the terminal ileum is straightforward. Although the polypoid lymphoid follicles that are frequently present may cause the radiologist to mistakenly diagnose Crohn's disease and consider cobblestoning, these nodules and the surrounding mucosa are covered with normal pink mucosa with a granular surface in air or with small villi visible underwater.

The endoscopic view of the terminal ileum is usually limited to 5 to 10 cm. There is rarely a serious indication to attempt further insertion, because ileal Crohn's disease almost invariably involves the terminal part of the ileum, and other lesions such as ileal angiodysplasia are exceedingly rare; a Meckel diverticulum is not endoscopically accessible because it is located 1 m proximally. If deep ileal intubation is attempted, 30 to 50 cm is usually the limit of what can be seen because of the acute angulations that occur in the small bowel.

Role of the Assistant

Colonoscopy must not be performed by one person alone. A properly trained and observant GI assistant is an absolute necessity for the proper performance of colonoscopy. The assistant's functions extend beyond the confines of the endoscopic procedures itself to include setting up the room before the procedure, recording endoscopic photographs, noting biopsy specimens, and filling out requisition forms for the histopathologist. All equipment that may possibly be necessary for the procedure must be assembled and available before the examination. The endoscopic assistant monitors the patient and assists with accessories. If polyps are encountered, the snare and patient return-plate must be plugged into the electrocautery apparatus, which requires setting the proper diathermy levels and testing to ensure that all equipment is functioning properly. During the examination, additional lubricant, gloves, and abdominal pressure may be provided from time to time during various phases. The assistant orders and prepares the necessary pre-endoscopic medications and is responsible for maintenance of the emergency cart, which must include antidotes for the various medications used, cardiac drugs, resuscitation equipment such as an Ambubag, endotracheal tubes and laryngoscopes, intravenous solutions, and oxygen. The cleaning, care, and maintenance of all the endoscopy equipment is in the hands of the assistant, who is responsible for mechanical cleaning and disinfection procedures. Most colonoscopists prefer to advance and withdraw the shaft of the instrument with the right hand, while the left hand manipulates the dial controls as well as the air-water and suction buttons. However, some colonoscopists prefer a two-person technique, whereby the assistant advances the instrument while the endoscopist handles the control portion of the colonoscope.

THERAPEUTIC PROCEDURES

Colonoscopy makes possible a range of therapeutic procedures. Polypectomy is the most obvious, but electrocoagulation, laser photocoagulation, or injection therapy of vascular lesions (e.g., telangiectasias, angiodysplasias, hemangiomas) are all routine. [24] Laser photodestruction also makes possible the ablation of some tumor masses and large sessile polyps if conventional surgical or snare polypectomy techniques are inappropriate. [25] The argon plasma coagulator is a relatively new monopolar device that delivers thermal energy to the colon wall in a noncontact fashion. It is useful for arteriovenous malformations, radiation telangiectasias, and fulguration of residual adenomatous tissue at the base of piecemeal polypectomy sites. [26]

Snare polypectomy of pedunculated or sessile polyps is efficient, rapid, and relatively safe. By constricting the tissue within the closed snare wire, low electrical power (often only 15– 20 watts) is sufficient for transection, with localized tissue heating. Because the colon mucosa is insensitive to thermal injury, the patient should feel nothing during polypectomy or electrocoagulation unless full-thickness heating is occurring, which causes the patient to warn of peritoneal pain at an early stage, before actual damage has occurred. If small (2 to 5 mm) polyps are found, simultaneous biopsy and electrocoagulation with electrically insulated hot-biopsy forceps can be performed. This rapid procedure ensures that a histological specimen is obtained. An alternative technique for small sessile polyps is cold resection, or cheese-wiring across the base by closing the snare in the absence of current. [27] Snared tissue can be retrieved for pathological analysis by aspiration through the instrument channel into a filtered suction trap or into a gauze tissue placed at the connection of the suction tubing with the endoscopic umbilical cord. Larger polyps can be retrieved whole using the snare loop or other grasping devices, although this may necessitate several reinsertions of the endoscope for multiple polyps. A metal basket or nylon-mesh net allows retrieval of several polyps or fragments from the right colon.

Except for polyps with an extremely broad base, almost all benign-appearing polyps encountered can be removed endoscopically. Pedunculated polyps are removed with snare application around the pedicle. Pure coagulating current at low power is used by most endoscopists following snare-loop tightening, providing controlled and localized tissue heating for blood vessel obliteration. [28] Thicker stalks with increased risk of immediate or delayed bleeding can be preinjected with an epinephrine solution to ensure hemostasis before snaring both flat and broad-stalked polyps with early invasive colon carcinoma may be successfully resected by submucosal injection polypectomy (SIP) technique, whereby the polyp is elevated by a fluid injection (3 to 10 [or up to 40] mL of saline or diluted epinephrine solution) into the submucosa prior to transection. [29] A viscous solution of sodium hyaluronate may also be successfully employed. [30] If the polyp is very large and snare placement is difficult, it may be useful to remove segments sequentially until the base can be easily visualized and complete transection can be accomplished. Even very large pedunculated polyps may be removed in piecemeal fashion. Polyps that usually cannot be removed endoscopically are those that cover a substantial area of the colonic surface (i.e., more than 30% of the circumference), extend over two interhaustral folds, are obviously malignant, or involve the appendiceal orifice. [31]

Every effort should be made to retrieve all the resected portions of each polyp and to submit them to the pathologist for histological evaluation. Because of the force of gravity, polyps have a tendency to fall into fluid pools or crevices that hide them from view, making retrieval difficult. There are various techniques for polyp retrieval. Although in clinical circumstances, only about 94% of transected colon polyps are recovered for full histopathological interpretation, those most likely to be lost are small and of little clinical significance.

Tube placement in patients with pseudoobstruction (Ogilvie syndrome) or postoperative ileus can be accomplished by passing a decompression catheter over an endoscopically placed guidewire after withdrawal of the colonoscope, [32] or by carrying the tube up alongside the endoscope during insertion. Immediate decompression occurs, but because of the frequency of redilation, it is wise to leave the tube in situ until it is expelled spontaneously, signaling restoration of normal propulsive contractions. Volvulus can be reduced by placing a wide-caliber tube beyond the twisted segment, provided the colon is not gangrenous, in which case surgery is mandatory. Foreign bodies can be successfully removed endoscopically from all levels of the large bowel. Balloon dilation is possible, usually with through-the-scope (TTS) balloons inserted under direct vision into a stricture, with or without prior guidewire placement. [33] The controlled radial expansion (CRE) balloons exert maximal dilating force over a range of sizes according to the degree of pressure applied. An appropriate-size balloon gives sufficient dilation for most circumstances with minimal risk of splitting the stricture. These balloons have the advantage of allowing passage over an endoscopically positioned guidewire, with correct insertion and dilation observed either endoscopically or on fluoroscopy. Other techniques for stricture dilation include incision into the stricture with electrocoagulation by a needle-knife, the slightly protruded tip of a polypectomy snare, or a papillotome; these methods are however, useful only for relatively thin, short strictures.

Short strictures, such as those after right hemicolectomy for Crohn's disease or when postoperative complications result in localized anastomotic narrowing, respond very well to endoscopic dilation. A single dilation can be permanently effective or can last for a few weeks to many months, depending on the presence of any inflammatory condition likely to cause restenosis. [34] Postoperative anastomotic stenoses may be dilated widely during the first examination because there is usually considerable fibrosis around the anastomotic site which makes the occurrence of free perforation unlikely. Primary strictures in inflammatory bowel disease unrelated to surgery, however, do not have the same surrounding cicatricial reaction, and the initial dilation should be limited to not greater than twice the diameter of the stricture to avoid perforation. Frequently, the lumenal narrowing in Crohn's disease is related to active inflammation with circular muscle spasm and so is rarely ameliorated by attempts at endoscopic dilation techniques. Self-expanding metal stents effectively dilate malignant strictures, and are usually permanently embedded into the tissue once inserted. They may be used for decompression prior to surgery or for symptom palliation. [35]

INDICATIONS

Flexible Sigmoidoscopy

Flexible sigmoidoscopy should be the procedure of first choice for examination of the rectum and sigmoid colon. It is useful for evaluation of complaints that most

likely are caused by conditions affecting the lower bowel, and for population-based screening for colon and rectal cancer, and should replace rigid proctosigmoidoscopy except when the rigid instrument is used for screening examinations of the rectum on grounds of cost, expediency, and the ease of taking larger biopsy specimens. [36] The rigid proctoscope (i.e., anoscope) is preferred for assessment of hemorrhoids and anal canal pathology, which are often poorly seen with a flexible instrument, even in retroversion.

Flexible sigmoidoscopy alone may be considered sufficient examination in patients with minor disturbance of bowel habit, obvious hemorrhoidal bleeding such as blood dripping into the toilet bowl following defecation, or localized left iliac fossa pain. With minimal discomfort and no sedation, the rectum, sigmoid colon, and often the descending colon or splenic flexure can be examined.

Colonoscopy

Colonoscopy can be performed in any patient fit for barium enema and for a similarly wide range of indications (Table 140-1). With the increasingly widespread availability of endoscopic skills and improvements in instrument technology, colonoscopy can often be regarded as a first-line procedure not preceded by barium enema. A prior barium enema rarely helps the endoscopist, because the configuration on x-ray films gives little indication as to how easy or difficult the colonoscopy will be. Most patients find barium enema a more unpleasant experience than colonoscopy because of the sustained inflation required in the double-contrast barium enema (DCBE) [37] and perhaps also because of the sedative-analgesic medications often given for colonoscopy.

- Evaluation of abnormality on barium enema likely to be clinically significant, such as a filling defect or a stricture
- Evaluation of unexplained gastrointestinal bleeding (including occult bleeding)
- Unexplained iron-deficiency anemia
- Screening and surveillance for colonic neoplasia
- Chronic inflammatory bowel disease of the colon if more precise diagnosis or determination of the extent or activity of disease will influence immediate management
- Clinically significant chronic diarrhea of unexplained origin
- Intraoperative identification of a lesion not apparent at surgery (e.g., polypectomy site, location of a bleeding site)
- Treatment of bleeding from such lesions as vascular malformation, ulceration, neoplasia, and polypectomy site
- Foreign body removal
- Excision of colonic polyp
- Decompression of acute nontoxic megacolon or sigmoid volvulus
- Balloon dilation of stenotic lesions (e.g., anastomotic strictures)
- Palliative treatment of stenosing or bleeding neoplasms
- Marking of a neoplasm for surgical localization

Modified from ref. 86.

TABLE 140-1 Indications for Colonoscopy

High-Yield Indications

Abnormal Barium Enema X-Ray The highest diagnostic yield of endoscopic pathology is in patients with tumor, polyps, stricture, or mucosal disease diagnosed on DCBE. Colonoscopy provides histological confirmation or confident exclusion of a questionable abnormality, [38] and allows immediate therapy where appropriate.
Colon Polyps: Benign and Malignant Nearly all polyps can be snare-resected during the procedure, and some radiologically diagnosed carcinomas prove to be benign lesions that are manageable endoscopically. Even with an apple-core lesion on x-ray films, colonoscopy can confirm the histology of the lesion, and in the absence of obstruction, also rule out synchronous polyps and cancers. Five percent of pedunculated or broad-based polyps removed endoscopically are found histologically to contain invasive carcinoma with malignant cells traversing the muscularis mucosae into the submucosal layer. The biologically benign entity in which malignant cells are present only superficial to the muscularis mucosae should not be described as “carcinoma in situ”; but rather by the more innocuous term, *severe dysplasia*. Surgical resection is not necessary in pedunculated polyps with invasive carcinoma if there is a sufficient margin (1–2 mm) between the limit of invasive carcinoma and the resection line on multiple sections, if the carcinoma is well or moderately well differentiated, and if there is no lymphatic or venous invasion. [39], [40] There is controversy as to whether these endoscopic and histological principles should be applied to the local removal of sessile malignant polyps or whether surgery should be recommended unless the patient is a poor operative risk. Because malignant polyps and polypoid carcinomas tend to occur in older, frail patients, and surgery rarely yields resectable lymph nodes without evidence of distant metastases, there is an increasing tendency to use conservative endoscopic management, even in lesions failing to meet the previously outlined criteria, unless the patient is an excellent surgical risk. [41] The use of a permanent colonic tattoo makes it possible to localize a malignant polypectomy site with accuracy in the event that surgical resection is necessary [8] or for subsequent surveillance after an interval of a few months before entering a normal follow-up regimen. [42]
Rectal Bleeding of Undetermined Etiology Rectal bleeding, especially if sustained, dark, or mixed in the stools, is frequently caused by tumor or mucosal pathology. In the subgroup of patients with visible rectal bleeding in whom a barium enema and sigmoidoscopy are normal, about 10% of referral patients have cancer, 15% to 20% have polyps (Fig. 140-7; see also Color Fig. 140-7), and up to 50% have some kind of a visible abnormality, including traumatized, inflamed, or ulcerated mucosa. Blood loss or anemia, particularly in elderly persons, remains the highest-yield indication for colonoscopy. Total colonoscopy should be performed rather than DCBE in the assessment of rectal bleeding, whether overt or occult, because of the obvious bonus of color view in seeing blood, altered blood, or bleeding points, which can be flat and totally invisible on x-ray films. It may also be possible to treat the cause of bleeding endoscopically. [43]

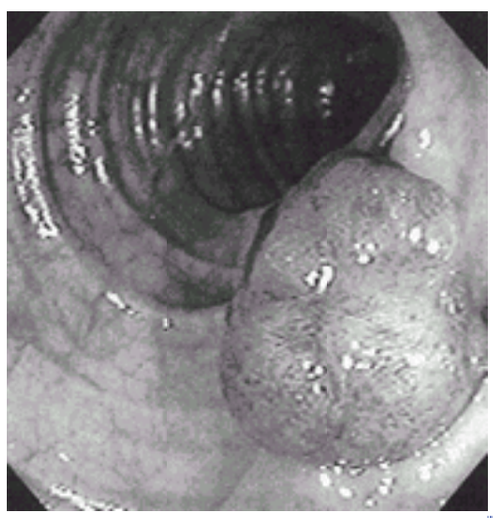

FIGURE 140-7. (See Color Fig. 140-7.) Smooth sessile adenoma is located in the descending colon.

Acute colonic bleeding. Acute colonic bleeding of overwhelming proportions may require surgery and perioperative bowel irrigation with on-table colonoscopy, [44] but bleeding of lesser severity may stop spontaneously. [45] Ten percent of patients admitted to an intensive care unit with a tentative diagnosis of acute colonic bleeding will have an upper GI source, [43] which must be considered in all instances. In most cases of acute bleeding, an attempt at conventional colonoscopy examination is indicated before resorting to angiography, scintigraphy, or other investigational techniques. If possible, oral or nasal tube electrolyte bowel preparation is started immediately after the patient presents because blood coating makes localization of the bleeding site very difficult without preparation or if enemas reflux blood proximally. Colonoscopy should be performed as soon as practical, because the aim is to examine the bowel while fresh bleeding continues. [43] Waiting even an hour or two may miss the opportunity for diagnosis and allow degradation of blood into a dark unpleasant mess. If there is bleeding, adequate visualization can be maintained by changing the patient’s position during the examination. Expertise is needed in this situation, but a cause for bleeding should be apparent in more than half of all cases, and treatment may be possible by polypectomy, electrocoagulation, injection therapy, or other means. [46] Postpolypectomy hemorrhage should be managed by immediate colonoscopy without preparation, since the site of bleeding is known. [47] An algorithm for the approach to acute lower GI bleeding has been published. [43]
Anemia Iron-deficiency anemia or a positive fecal occult blood test should be evaluated by colonoscopy rather than DCBE because flat lesions such as angiodysplasia or minor inflammatory change cannot be diagnosed radiologically. Even if radiographs appear normal, endoscopy is known to find significant numbers of radiologically missed lesions such as cecal carcinomas. In anemic patients, gastroscopy and colonoscopy can be performed at the same visit. The object of

endoscopy in anemic patients is mainly to rule out carcinoma or vascular anomalies with certainty; about 80% have negative examinations, but the endoscopic opinion of normality is considerably more certain than a negative x-ray film result.

Chronic Diarrhea Patients with long-standing bowel diarrhea require an accurate examination with histological biopsies routinely taken and other specimens as appropriate. Rectal evaluation is adequate in many patients, but total colonoscopy and ileal examination complete the investigation and sampling process in one definitive test. Microscopic or collagenous colitis can occur despite normal endoscopic appearances; therefore, biopsy specimens must be taken in any patient with chronic diarrhea or increased bowel frequency. [48] However, the normal colonic mucosa frequently harbors inflammatory cells, and great care should be taken to avoid misinterpretation and overdiagnosis of inflammatory bowel disease when none is truly present, based on an inexperienced pathologist's report of acute or chronic inflammation on biopsy specimens. Collagenous colitis is diagnosed histologically from a thickened intraepithelial collagen layer. Ischemic colitis has a range of appearances, from slight reddening and friability, to varying degrees of ulceration, to frank gangrene; the diagnosis is often possible by combining the typical history of sudden onset of pain, bleeding, and diarrhea with endoscopic evidence of a perisplenic distribution.

Inflammatory Bowel Disease Often, the mucosal appearances are sufficient to make immediate assessment of the extent and type of colitis. [49] Crohn's disease, in particular, shows a rather characteristic pattern of small aphthoid ulcers with intervening normal mucosa quite unlike the generalized redness of early ulcerative colitis. More advanced inflammatory disease of any type, including ulcerative colitis, Crohn's disease, tuberculosis, or amoebic colitis, can be nearly indistinguishable from each other because of the colon's limited range of responses to various diseases that affect the mucosal surface. [50] Once the diagnosis of inflammatory bowel disease has been established, there are only a few indications for follow-up colonoscopy besides malignancy surveillance, unless there is a marked change in symptoms.

Cancer Surveillance or Prevention

The long period preceding cancer during which a focal adenomatous polyp is visible gives the opportunity for visualization, biopsy, and destruction of precancerous lesions, even at sizes of 1 to 2 mm. [51] Accuracy is possible, because color makes it easy to distinguish which small excrescences are fecal, which are air bubbles, and which are polyps. A syndrome of flat adenomas has been described. [52] These adenomas may have a tendency to degenerate into carcinoma while still relatively small in diameter. [53] Detecting their presence may be enhanced by zoom colonoscopy or by dye spraying or "chromoendoscopy." [54]

The recommendations of an expert multidisciplinary panel, published in 1997, [55] included offering colonoscopy every 10 years to asymptomatic persons of average risk who are 50 years of age or older. In addition, the panel suggested annual fecal occult blood tests in this population, with colonoscopy as the investigation of choice if the occult blood test is positive. A flexible sigmoidoscopy is recommended every 5 years, followed by colonoscopy to remove polyps, biopsy cancers, and to examine the rest of the bowel should an adenoma or cancer be seen. Colonoscopy is indicated for any patient with a positive fecal occult blood test and in those who are at increased risk for polyp and cancer formation, including those with first-degree relatives with either colorectal cancer or an adenomatous polyp, in whom screening should begin at age 40. [55] People with a family history of familial adenomatous polyposis whose genetic testing is positive or indeterminate should be offered flexible sigmoidoscopy every 12 months beginning at puberty. [55] Family history of hereditary nonpolyposis colorectal cancer (HNPCC) should trigger an examination of the entire colon every 2 to 3 years starting between the ages of 20 and 30 years.

The interval between surveillance examinations after removal of adenomatous polyps or an operation for colorectal cancer is a matter of debate, [56] but the expert panel [55] has made the following recommendation: Removal of an adenomatous polyp larger than 1 cm in diameter or of multiple polyps requires repeat colonoscopy 3 years after the initial examination, and if that is negative or reveals only a single small tubular adenoma, the surveillance interval can be increased to 5 years. In special circumstances, such as a polyp with invasive cancer, large sessile adenomas, or numerous adenomas, a shorter follow-up interval may be necessary. In patients who have had a colorectal cancer surgically resected with a curative intent, if total colonoscopy had not been performed preoperatively, a complete colon examination should be completed within 1 year of the resection. If this or the preoperative examination was normal, subsequent colonoscopy is indicated in 3 years, and if normal, every 5 years.

Surveillance examinations in patients with extensive ulcerative colitis [57] or Crohn's colitis [58] normally start 8 years after onset of disease and are repeated at 1- or 2-year intervals, with 10 or more biopsy specimens taken at representative sites around the colon. The endoscopist is alert for any nodular, indurated, or plaquelike lesions in the colon, but varying degrees of low- or high-grade dysplasia may be found in relatively normal-looking mucosa.

Intraoperative Colonoscopy

The most common indication for intraoperative colonoscopy is to localize the site of a previously endoscopically resected malignant polyp (not necessary when preoperative tattooing is performed). The site may heal completely in 3 weeks, leaving no external sign of its location, but it can often be endoscopically identified as a scar on the mucosal surface. Another indication is to assist the surgeon in identifying which colonic segment is the source of massive lower GI hemorrhage. The presence of a large volume of blood can completely obscure intralumenal vision. Blood can be rapidly flushed out of the colon by a large-volume saline lavage instilled through a cecostomy and removed with a rectal tube. [48]

Low-Yield Indications and Circumstances When Colonoscopy Is Not Indicated

Conditions manifesting with functional symptoms such as long-standing constipation, bloating, or chronic abdominal pain, may be investigated by x-ray examination if there is no occult or overt colonic bleeding (Table 140-2). On the other hand, the older adult patient may be unable to cooperate with the need to retain barium, resulting in a poor radiographic examination. Bowel symptoms in patients older than 70 years of age may be better investigated by colonoscopy than barium enema. [59]

- Chronic, stable, irritable bowel syndrome or chronic abdominal pain
- Acute diarrhea
- Metastatic adenocarcinoma of unknown primary site in the absence of colonic signs or symptoms when it will not influence management
- Routine follow-up of inflammatory bowel disease
- Upper gastrointestinal (GI) bleeding or melena with a demonstrated upper GI source

Modified from ref. 66.

TABLE 140-2 Diagnostic Colonoscopy Is Usually Not Indicated or Has a Low Diagnostic Yield in the Following Circumstances

A flexible sigmoidoscopy may be the examination of choice when the patient presents with an acute diarrheal syndrome, since the view of the distal colon mucosa often (but not always) mirrors the findings in the remainder of the colon. Other than surveillance for malignancy, repeated colonoscopy for routine follow-up of patients with established inflammatory bowel disease provides little clinically useful information. Colonoscopy need not be performed in the patient with overt GI bleeding in whom an active upper GI source has been demonstrated. Similarly, there is no indication for colonoscopy in the patient with an adenocarcinoma where the primary site of the tumor is unknown, unless the result of colonoscopy will influence the management of the patient.

CONTRAINDICATIONS AND RISKS

Colonoscopy is a relatively stressful physiological experience and a strong vagal stimulus that can produce dysrhythmias, minor electrocardiographic disturbances, and a degree of hypotension; therefore, it is contraindicated for several weeks after myocardial infarction. [60] Both the air pressure involved in distention of the colon and the unavoidable stretching during passage around loops and bends have the potential to increase any existing risk of perforation. Because of this, colonoscopy is contraindicated in the acute or abscess phase of diverticulitis. Severe acute episodes of ulcerative, Crohn's, ischemic, or infective colitis have generally been considered a contraindication for colonoscopy, but some reports suggest that, with due expertise and care, it is safe and may be of value in therapeutic decision making in these situations. [61]

Therapeutic maneuvers increase the hazards of colonoscopy. Balloon dilation in a sick patient may justify antibiotics. Polypectomy carries a small risk of immediate primary bleeding or of delayed secondary hemorrhage for up to 29 days after the procedure. The likelihood of delayed bleeding is especially increased in patients with a coagulopathy or in those taking anticoagulants or antiplatelet therapy. When medical conditions prohibit discontinuation of coumadin, the patient may be

admitted to the hospital for the substitution of heparin for oral anticoagulation. Once the coagulation parameters have returned to normal levels, the heparin dosage should be stopped 4 hours before colonoscopy with polypectomy. If a clean polypectomy has been performed without bleeding, heparin may be restarted 4 hours after the procedure, and oral anticoagulants reinstituted 12 to 24 hours later although the possibility of delayed hemorrhage will remain. The patient must remain on heparin until prothrombin levels have returned to the therapeutic level. If polypectomy is associated with bleeding, an attempt should be made to withhold heparin for 8 to 12 hours if it is considered that the patient can safely remain off anticoagulants for that period. Low-molecular-weight heparin has been used during periods of coumadin discontinuation but this has not been supported by clinical investigations. [62]

Polypectomy can result in perforation by cutting through the bowel wall or by applying sufficient thermal energy to burn through and necrose the full thickness of the colon, resulting in a perforation delayed for between several minutes to many days after the polypectomy. The colon wall is extremely thin, with the normal mean colon wall thickness being about 2.2 mm. [63] Some features of a full-thickness thermal injury to the colon wall, including localized pain, fever, and leukocytosis, may occur 6 to 24 hours after polypectomy. This is known as the postpolypectomy or transmural burn syndrome, [64] and must be distinguished from a perforation. Transmural damage is localized and self-sealing when bowel loops, mesentery, omentum, or other adjacent peritoneal surfaces become adherent, so that conservative management with antibiotics and bed rest is usually sufficient.

When signs of frank perforation occur after colonoscopy, it is safest to advise immediate surgery. If only a small amount of free air is present on a postendoscopy x-ray film and the patient has no symptoms, it may be safe to treat the patient conservatively, with antibiotics but without surgery. In any event, a surgeon should always be involved in observation of the patient and in deciding whether or not surgery is indicated. [65]

Current morbidity and mortality rates for colonoscopy are unknown but are certainly higher than those for barium enema. Reviews from the late 1980s and 1990s indicate that the perforation rate is about 1 in 2500 colonoscopies, [66] with a mortality of 1 in 15,000 colonoscopies; most deaths occur after inappropriate conservative management of suspected perforation. [64] The complication rate is inevitably higher in therapeutic colonoscopy than in diagnostic colonoscopy.

The bleeding rate after polypectomy varies from 0.26% to 1.5%, [66] but likely can be reduced with better attention to hemostatic techniques such as the sole use of coagulating current [28] and slow transection. The use of cutting current may be associated with a higher bleeding incidence. [67] Injection or other means of endoscopic therapy for immediate control of bleeding should avoid the small number of cases in which angiographic or operative intervention was previously necessary. Unexpected x-ray demonstration of free air in the abdominal cavity (pneumoperitoneum) of symptomless patients submitted to routine abdominal x-ray after colonoscopy has been reported. Surgeons also report the existence of hematomas, serosal splits, and manifestations of local colonic trauma in patients coming for laparotomy soon after colonoscopy. These facts serve as a reminder to all endoscopists, particularly those in the learning phase, to be cautious, to be considerate of the patient, to avoid oversedation, and to be prepared to abandon a colonoscopy that appears unreasonably traumatic.

COMPARISON WITH BARIUM ENEMA

Colonoscopy has become a procedure of first choice for most colonic investigations. The ability of the barium enema to diagnose advanced cancers is comparable to that of colonoscopy, but it is less accurate for early colon cancers and polyps. [68], [69] In contrast, it is estimated that it is unusual to miss a polyp = 1 cm in diameter during colonoscopy. [70]

Unfortunately, patients with very long and mobile colons or with advanced diverticular disease who are difficult to endoscope are also difficult for the radiologist. Barium enema can be performed following failed or difficult colonoscopy, but endoscopy is impossible in the presence of barium; it is therefore logical to attempt colonoscopy first in most circumstances, providing that each technique is equally available and that the criteria for performing primary colonoscopy are present.

Technical difficulties of colonoscopy in patients with severe constipation and megacolon make them best managed by modified barium technique, sometimes with no bowel preparation, whereas patients who have reduced mobility, rectal incontinence, prolapse, or stomas are best examined by colonoscopy because of technical difficulty in obtaining proper filling and coating with x-ray contrast material. Barium enema is particularly effective in assessing the configuration and gross morphology of the colon, especially when there are multiple strictures or fistulae that may be impassable or invisible to the endoscopist. The colon in idiopathic or acquired megacolon and Hirschsprung's disease may be almost impossible to prepare and offensive and difficult to endoscope, whereas radiology provides the necessary assessment of colon configuration. In elderly patients with symptoms suggestive of diverticular disease, x-ray can exclude serious pathology in most cases; in the same patients, unless the endoscopist is expert, colonoscopy can be relatively slow, traumatic, and more hazardous, although the endoscopic examination may be more diagnostic. [59]

When colonoscopy proves unreasonably difficult, endoscopy can be abandoned, aspirating residual air as far as possible and proceeding to immediate DCBE. Using CO_2 rather than air insufflation for colonoscopy is an advantage because in 15 minutes there is total absorption of residual CO_2, leaving the radiologist a nondistended bowel that is easy to fill and coat with barium. [71], [72] Endoscopic biopsies can be performed safely before DCBE, [73] and in all probability hot biopsies and snare removal of small stalked polyps may also safely precede the x-ray examination.

Barium enema films are easily stored and retrieved for review and comparison with subsequent examinations, a feature not available with colonoscopy. Videotapes can be recorded continually or intermittently during colonoscopy, but there is no method for endoscopic photographic mapping of the large bowel. Watching 20 to 40 minutes of a colonoscopic videotape is a tedious task that is not likely to be a standard part of any endoscopic review.

VIRTUAL COLONOSCOPY (COMPUTED TOMOGRAPHY OR MAGNETIC RESONANCE COLOGRAPHY)

A computer reconstruction of a rapidly performed abdominal spiral computed tomography (CT) scan provides a simulated tubular view of the intestinal lumen. Infoldings (haustrae), outpouchings (diverticula), and intraluminal protuberances (polyps) can be identified. The process is noninvasive, requiring a single breath-hold while the colon is maximally air-inflated. Cancers and most polyps over 10 mm in diameter can be identified, [74] but smaller polyps are often overlooked when compared to findings at conventional colonoscopy. [75] Retained stool may give a false-positive reading, and flat mucosal lesions are difficult to identify. Technological developments will increase the accuracy of this tool which will require further evaluations before assigning this radiologic procedure a role in the screening algorithm. The use of air as a contrast medium instead of barium means that the "virtual" examination has the ability to either precede or follow the endoscopy.

LIMITATIONS OF COLONOSCOPY

It is pertinent to mention that the endoscopist is capable of gross errors in localization and occasionally of missing large lesions. Blind spots for the endoscopist occur in certain areas, such as the rectal ampulla and flexures (especially the mobile sigmoid-descending colon junction), behind acute bends, and in regions of spastic muscle contractions. Adhesions or strictures can render the endoscope tip immobile and make a proper view for targeted biopsies impossible. Large polyps on long stalks can be missed by the endoscopist because they move around and may spring out of the field of view as the colonoscope is pulled back. Poor preparation and redundant or mobile colons also make the endoscopic examination less accurate.

Careful inspection for possible abnormalities is important on the way in as well as during the withdrawal phase of a colonoscopy because the view of the stretched colon is quite different from that obtained when the bowel is shortened. Several passes may be needed to see all aspects of an angulated or convoluted area properly. There is no room for complacency by the endoscopist over the accuracy of colonoscopy; it takes dexterity and integrity to be sure that an optimum examination has been performed. It can be assumed that 5% to 10% of the mucosal surface is not seen during colonoscopy. This explains the significant detection of lesions up to 5 to 10 mm in diameter during check colonoscopies usually performed within 1 year (or even on the same day) [70] of a previous colonoscopy and polypectomy to establish a "clean colon" that is free of adenomas. It has been estimated that 27% of adenomas in the size range of 1 to 5 mm are missed on colonoscopy, as are 13% of adenomas in the range of 6 to 9 mm, and 6% of those over 10 mm. [70] Colonoscopy can miss colonic cancer in the intramucosal lesions of chronic ulcerative colitis, within strictures, or where there is submucosal involvement by extrinsic neoplasm or metastases.

In addition to diagnostic difficulties, the mechanical limitations of colonoscopy are relevant. An expert manages up to 98% to 99% total colonoscopy, especially if there is no stricture or stenosing lesion to prevent insertion. Less expert endoscopists achieve only up to 70% to 75% total [76] colonoscopy and are more likely to be slow, inaccurate in diagnosis and localization, and more prone to trauma and complications. The lack of proper means of teaching colonoscopy skills limits its clinical

application worldwide.

COSTS

Colonoscopy is highly cost-effective. The instrumentation involved is inexpensive compared to x-ray. A colonoscope should last for at least 500 examinations before major repair or replacement becomes necessary. The actual physician time involved per procedure is greater for colonoscopy than for the barium enema, during which examination films may frequently be taken by a technician. The overall cost is approximately equal if an x-ray and flexible sigmoidoscopy–first schema is compared to a colonoscopy-only approach for the investigation of patients with lower GI symptoms. Many patients with an abnormal sigmoidoscopy or barium enema demonstrating a lesion or probable pathology will require a subsequent colonoscopy for confirmation, polypectomy, or biopsy; whereas only a few with the colonoscopy-first approach will require a barium enema. [77], [78]

FUTURE DEVELOPMENTS

Videocolonoscopy will allow improvements in image resolution as well as potential for computer enhancement and related developments that should improve diagnostic accuracy, localization, and ease of instrument handling. Advances in bowel preparation should permit comfortable clearance in a few hours instead of the 24- to 48-hour preparations currently used. Developments in safe, short-lived analgesia should make insertion easily tolerated while allowing normal activities soon afterward such as driving or returning to work. Computerized interactive teaching models will permit education in manipulative and interpretive skills without imposing an undue burden on and risk to patients during the endoscopist's early training phase. [79], [80]

Development of other diagnostic and therapeutic modalities will allow the endoscopist to apply miniaturized imaging probes to areas of interest and to ablate lesions more completely and safely than can be performed at present. For instance, ultrasound probes passed through the instrumentation channel help to determine the presence and depth of penetration of malignancy. [2], [81] Optical coherence tomography (OCT) uses light instead of sound waves to achieve an "optical biopsy" of the mucosa and submucosa. [82] A new technology is emerging to record spectrophotometric measurements of tissue fluorescence by a sensing device passed through the accessory channel. [83] Differentiation between neoplastic and normal tissue by their inherent differences in level of tissue excitation by laser stimulation may be sufficient for diagnostic purposes or will enable directed biopsy of abnormal areas. [84] With improved photosensitizing agents and laser photocoagulation or locally injected immunotherapeutic agents, colonoscopy should be able to make further inroads into territory that is currently considered the domain of the surgeon.

Wireless "capsule" endoscopy [85] has been developed for small bowel exploration, where peristalsis is the propulsive force for movement. The narrow caliber lumen is sufficiently illuminated by the light to produce a diagnostic visual image. Several inherent differences in the large bowel are impediments to adapting the capsule to colonoscopy: the diameter is wide and the light will be insufficient for adequate visualization; peristalsis is relatively slow in the colon; air insufflation is required to distend the bowel; the capsule tends to tumble and its movements cannot be controlled; and there is no therapeutic capability. Some or all of these factors can be overcome in the future, which may see the development of robotic therapeutic capsules armed with snare, forceps, scissors, and clips.

The future seems limitless, but for now, the colonoscope is the best imaging system for the colon, and has the additional ability to perform therapy including injection, biopsy, decompression, and polypectomy.

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CHAPTER 141

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ENDOSCOPIC RETROGRADE CHOLANGIOPANCREATOGRAPHY, ENDOSCOPIC SPHINCTEROTOMY AND STONE REMOVAL, AND ENDOSCOPIC BILIARY AND PANCREATIC DRAINAGE

Endoscopic cannulation of the major papilla with imaging of the pancreatic duct and biliary tree (endoscopic retrograde cholangiopancreatography [ERCP]) was first successfully accomplished with an end-viewing duodenoscope and reported in 1968. [1] Subsequent development of side-viewing endoscopes with a catheter-deflecting elevator greatly facilitated the technique. Diagnostic studies were supplemented by the first endoscopic sphincterotomies in 1973. [2], [3] Techniques of biliary stone extraction, nasobiliary tube placement, and biliary stent placement soon followed. These developments permitted less invasive diagnostic and therapeutic maneuvers in the pancreatic duct and bile duct previously limited to open surgical and percutaneous techniques.

ENDOSCOPIC RETROGRADE CHOLANGIOPANCREATOGRAPHY

Indications/Contraindications

The role for diagnostic ERCP alone is diminishing as other less invasive/noninvasive imaging techniques (e.g., endoscopic ultrasound, magnetic resonance cholangiopancreatography) become more widely used. Imaging of the ducts without anticipated therapy is clinically helpful in a few clinical settings where other noninvasive or less invasive procedures are negative and the clinical suspicion for pancreaticobiliary disease remains high, such as cholestasis without a dilated duct and chronic abdominal pain with concern for chronic pancreatitis. ERCP is indicated in those clinical settings in which there is significant suspicion of an obstructing, inflammatory, or neoplastic pancreatobiliary lesion, which if detected or ruled out, would alter clinical management. A general classification of indications is listed in Table 141-1.

Suspected Biliary Ductal Disorder
Jaundice or cholestasis of suspected obstructive origin
Acute cholangitis
Gallstone pancreatitis
Clarification of biliary lesion seen on other imaging test
Biliary fistula
Suspected Pancreatic Ductal Disorder
Pancreatic cancer
Mucinous or cystic neoplasm
Unexplained recurrent pancreatitis
Chronic pancreatitis with unrelenting pain
Clarification of pancreatic lesion on other imaging test
Ascites or pleural effusion of suspected pancreatic origin
Pancreatic pseudocyst or fistula
To Direct Endoscopic Therapy
Sphincterotomy
Biliary drainage
Pancreatic drainage
To Direct Endoscopic Tissue/Fluid Sampling
Biopsy, brush, fine-needle aspiration
Bile/pancreatic juice collection
Preoperative Ductal Mapping
Malignant tumors
Benign strictures
Chronic pancreatitis
Pancreatic pseudocysts and ductal disruptions
Mucinous or cystic tumors of pancreas
To Perform Manometry
Sphincter of Oddi
Ductal

ERCP, endoscopic retrograde cholangiopancreatography.

TABLE 141-1 Indications for ERCP

There are few, if any, true contraindications for ERCP. The only truly absolute contraindication is refusal of the patient to undergo endoscopy or the patient's inability to cooperate during the procedure for whatever reason. Even in the latter circumstance, ERCP can be performed under general anesthesia when the study is considered essential for the patient's management. Most contraindications are relative and in such settings, the degree of risk must be balanced against the potential benefit. In many settings—such as acute cholangitis with bile duct stones, malignant biliary strictures, and pancreatic pseudocysts—diagnostic ERCP should not be done unless simultaneously accompanied by a drainage procedure (surgical or endoscopic). ERCP in patients with severe acute pancreatitis, especially with necrosis (when gallstones or trauma are not the etiology), is considered a relative contraindication as bacterial contamination of the pancreatic bed can occur. Other relative contraindications include an unstable cardiopulmonary disease or severe coagulopathy. Patients with co-morbid life-threatening conditions can frequently have their ERCP performed in the intensive care unit (ICU) (with or without fluoroscopy) if deemed medically necessary. For example, in patients with septic shock from bacterial cholangitis, biliary drainage can be lifesaving.

Preparation

Preparation for ERCP involves assembly of a skilled team that includes physician(s), nursing personnel, and a radiology technician. A quality fluoroscopic unit is

needed. A wide variety of catheters, guidewires, stone extraction balloons and baskets, sphincterotomes, stents, drainage catheters, lithotripters, and tissue sampling devices should be available.

Patient preparation includes an updated history and physical and recent complete blood count, serum liver chemistries, serum amylase and/or lipase, coagulation studies, and noninvasive imaging of the upper abdomen, with abdominal ultrasound or a computed tomography (CT) scan. Special risk factors such as anticoagulant therapy, bleeding disorders, prosthetic heart valves, and allergies must be addressed.

Informed consent for ERCP must be obtained. It is both legally and ethically necessary to apprise the patient and family of the risks, benefits, and alternatives of ERCP. Table 141-2 lists the potential complications of diagnostic and therapeutic ERCP and their relative frequency. While legal standards continue to evolve, we recommend that patients not only be informed of the potential complication frequencies but that they also be told that a severe complication may possibly result in a prolonged hospital stay, ICU monitoring, or open surgery and may very rarely result in permanent disability or death. Complication rates vary according to patient and procedure risk factors as well as the disease process being evaluated and treated. [4], [5] Patients with uncomplicated biliary stones, malignancy, or chronic pancreatitis have lower complication rates, while patients with recurrent pancreatitis and sphincter of Oddi dysfunction have two- to threefold higher complication rates. Procedure techniques associated with higher complication rates include repeated cannulation attempts, repeated pancreatic duct injections, pancreatic parenchymal acinarization, and precut sphincterotomy. Attention to details of the technique and patient selection can minimize but not eliminate complications. Early recognition and treatment of complications helps to limit morbidity.

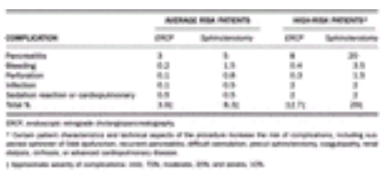

TABLE 141-2 Approximate Frequencies of Complications from ERCP and Sphincterotomy (%)

ERCP can be performed with fiberoptic or videochip instruments that have similar performance characteristics. Video systems offer the advantage of television monitor viewing by all persons in the endoscopy suite. This offers better teaching capabilities and allows better coordination between the endoscopist and nursing assistants. For Billroth II patients, we generally start with a standard side-viewing duodenoscope, but an end-viewing endoscope is occasionally needed. In patients with a long Roux-en-Y gastroenterostomy, a 220-cm enteroscope will reach the site of pancreatic duct or bile duct drainage in approximately half of patients. [6] The lack of a catheter-deflecting elevator and limited compatible accessories make end-viewing endoscopy difficult in these settings.

Technique

The patient is positioned in a prone to slightly left lateral decubitus position on a fluoroscopic table. An antiperistaltic drug (e.g., glucagon or atropine) to inhibit duodenal motility is commonly needed. Initially, a brief endoscopic examination of the esophagus, stomach, duodenum, and major duodenal papilla is done. The finding of a large ulcer or neoplasm may cancel the need for ERCP. Other findings such as varices, a pseudocyst pressing on the gut wall, or edema of the medial wall of the duodenum help to quantitate or localize disease processes. The major papilla is usually located on the medial aspect of the mid-descending duodenum. Prior to attempts at cannulation, fluoroscopic visualization (or a “scout film” radiograph prior to starting the procedure) of the field of interest should be performed to look for calcifications, masses, and old contrast material. The major papilla is then cannulated, usually with a 5F plastic catheter. Orientation of the catheter tip toward the 11 to 12 o’clock position will more likely enter the bile duct; orientation of the catheter toward the 3 to 5 o’clock position will more likely enter the pancreatic duct. Cannulation may be done by gentle impaction of the catheter tip in the papillary orifice. Deep cannulation (greater than 1 cm penetration of the catheter into the duct) more securely establishes an intraductal position which allows contrast injection, fluid aspiration, patient position changes, and endoscope position changes without loss of access to the duct.

Standard contrast media (e.g., meglumine diatrizoate) at a 50% to 60% concentration (full-strength) is used for pancreatography, while 25% to 30% (half-strength) concentration is recommended for cholangiography. Biliary stricture detail is better defined with full-strength contrast, however. Nonionic and lower osmolality contrast media, which are more expensive, offer no safety advantage. Contrast media injection is done with continuous fluoroscopic monitoring. The extent of ductal filling should be correlated with the clinical need to know the ductal anatomy. Complete pancreatography involves filling of the main duct and side branches to the tail. High-resolution fluoroscopy is required to see such detail. In settings where there is excess overlying gas or obesity, underfilling of the pancreatic duct is recommended to avoid acinarization (instillation of contrast media into the pancreatic parenchyma). For an extremely dilated duct, initial aspiration of fluid will allow better contrast visualization without overdistention of the duct. Complete cholangiography requires filling of the peripheral intrahepatic radicles. The left lobe is more dependent in the prone position and fills preferentially. Right lobe filling may require tilting the patient’s head down 15 to 20 degrees on the fluoroscopy table, more forceful injection (a balloon occlusion catheter is helpful), or turning the patient to the supine position. Contrast media mixes slowly with gallbladder bile and final films are best taken in the supine position after endoscope withdrawal (and additional time for mixing with gallbladder contents). Occasionally, delayed gallbladder films taken 4 to 12 hours after completion of the procedure allow for passage of intraluminal gas, giving better diagnostic film quality. In settings of tight biliary strictures, limited contrast filling upstream should be done until catheter access above the stricture is achieved. Similarly, limited pseudocyst filling should be done unless immediate drainage is certain. Several views of each ductal position are recommended both in the limited filling and more completely filled state.

Sphincter of Oddi manometry (SOM) usually is performed at the time of ERCP. All drugs that relax (e.g., anticholinergics, nitrates, calcium channel blockers, glucagon) or stimulate (e.g., certain narcotics, cholinergic agents) should be avoided for at least 8 to 12 hours prior to SOM and during the manometric session. SOM is performed using a low-compliance infusion pump system and a 5F catheter.

Endoscopic sphincterotomy (ES) of the bile duct is commonly performed prior to removing bile duct stones or placing a biliary stent. A pull-type (traction) sphincterotome is advanced into the bile duct and its position confirmed on fluoroscopy. The sphincterotome is pulled back until 5 to 7 mm of cutting wire are passed into the papillary orifice. The instrument is bowed to apply gentle tension in the 11 to 12 o’clock direction. Cautery current at 40 to 60 watts is applied in less-than-one-second bursts to stepwise cut 80% to 90% of the intramural portion of the bile duct. Although blended current is most commonly used, pure cut current may be associated with a lower incidence of pancreatitis without increasing the rate of sphincterotomy induced bleeding. [7] Adequacy of the sphincterotomy is judged by pulling a bowed sphincterotome through the incised sphincter. Small papillae and papillae associated with diverticula may require smaller cuts. Minor bleeding may occur but usually stops spontaneously. Bleeding that limits the endoscopic view should be controlled by hydrostatic balloon tamponade, bipolar cautery, or epinephrine injection.

Precut sphincterotomy involves cutting the papilla to gain deep intraductal access to the biliary tree. This technique should be limited to experienced endoscopists and applied in patients only when there is a high clinical suspicion of obstructive pathology (e.g., jaundiced patient with dilated bile duct on ultrasound) after standard techniques fail. Precutting can be achieved by impaction of a short-nosed, pull-type sphincterotome into the papillary orifice with sequential shallow cephalad cuts until the biliary orifice is identified. Similar sequential shallow cuts can be made with a needle knife. We prefer to place a 3F to 4F, 8-cm long, unflanged polyethylene stent into the pancreatic duct first, if possible, and use the stent to guide needle knife cutting.

Radiology

Figure 141-1 shows a normal pancreatogram. The main duct contour is typically S-shaped but numerous other configurations are common and normal. The upper limits of normal for main duct diameter are 5 mm in the head, 4 mm in the body, and 2 mm in the tail. Side branches are delicate with terminal branching. The accessory duct extends from the genu at the head/body main duct junction to the minor papilla. The minor papilla has a patent orifice in approximately 85% of patients. Pancreas divisum is discussed later in the chapter.

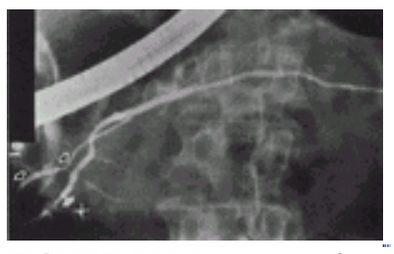

FIGURE 141-1. A normal pancreatogram obtained by placing a metal-tipped cannula (*solid arrow*) in the duct of Wirsung. Note the gradual tapering of the main pancreatic duct and the delicate side branches. The duct of Santorini (*open arrows*) is filled by its connection with the main duct.

A normal cholangiogram is shown in Figure 141-2. The upper limits of normal diameter for the common bile duct is 10 mm. The cystic duct commonly joins the common duct approximately halfway from the hilum to the papilla, but this junction may be quite variable. The intrahepatic radicles have a treelike branch pattern with marked variation in distribution.

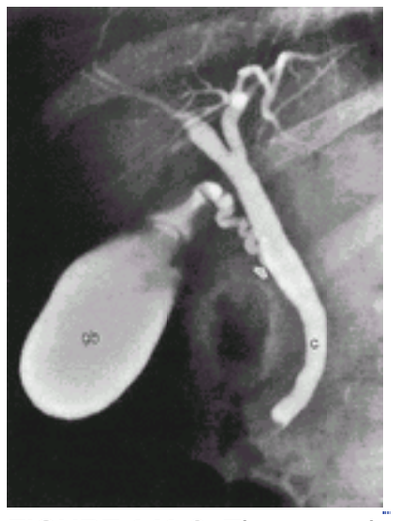

FIGURE 141-2. The normal common bile duct (*c*) becomes the common hepatic duct proximal to the insertion of the cystic duct (*arrow*). The common hepatic duct bifurcates into the right and left hepatic ducts. The cystic duct has a spiral shape owing to the valves of Heister and connects the gallbladder (*gb*) to the bile duct.

Cannulation Success Rates

The papilla is readily identified by experienced endoscopists in nearly all patients with normal anatomy. Difficulties in finding the papilla may arise in cases of large papillary tumors, duodenal stenosis, edematous folds caused by acute pancreatitis, or if the papilla is located inside a diverticulum. Initial cannulation success rates with ductography vary from 80% to 95% depending on operator experience, anatomy, and disease state. Cannulation is easiest in patients with biliary stones but more difficult in patients with periampullary neoplasms, sphincter of Oddi dysfunction, and chronic pancreatitis with obstructing stones. In experienced centers, cannulation success rates exceed 95% even in difficult cases where a prior attempt at cannulation failed. [8], [9] Such high success rates require supplemental use of precut sphincterotomy and minor papilla cannulation in 10% to 25% of cases. In patients with a Billroth II gastrojejunostomy, the success rate should be greater than 80% when the procedure is performed by an expert.

Endoscopic Findings

Duodenal diverticula (usually periampullary) will be present in 10% to 20% of patients. Patients with ductal stones may have edema, enlargement, or inflammation of the major papilla. If a stone has recently passed, as is common in gallstone pancreatitis, the papillary orifice will appear enlarged or lacerated. Stones impacted in the terminal bile duct invariably cause bulging of the papilla but a stone visible in the papillary orifice is uncommon. Pancreatitis commonly causes edema of the medial aspect of the descending duodenum, including the papillae. Pancreatic neoplasms may cause (mass effect) compression from the duodenal bulb down to the major papilla. Duodenal mucosal invasion in pancreatic head cancers occurs in approximately 10% of patients. Tumors of the papilla usually appear as fleshy enlargements with or without ulceration. Pseudocysts will cause compression of the stomach or duodenum in approximately one third of cases.

Complications of ERCP/ES

Complications of ERCP/ES are undesirable outcomes related to some portion of the procedure or sedation required for the procedure. Unsuccessful cannulation, stent placement, stone removal, or making an incorrect diagnosis are failures of the procedure, but generally not included as complications. Table 141-2 lists the more common complications for diagnostic and therapeutic ERCP. Since 1991, a more uniform classification of complications and their severity has been developed. [10]

BILE DUCT STONES

The introduction of ES by Classen and Kawai [2], [3] in 1974 initiated a change in the management of bile duct stones. Before that time, laparotomy with common bile duct exploration was the main therapeutic recourse for the patient with choledocholithiasis. With improvements in equipment and accessories, growth in numbers and skill of biliary endoscopists, and the introduction of laparoscopic cholecystectomy, the clinical settings in which endoscopic management is applied to common duct stones have been broadened considerably (Table 141-3).

1. Postcholecystectomy (no T-tube in)
2. Postcholecystectomy (T-tube in)
Symptomatic patient with immature T-tube tract
Failed T-tube extraction
3. Gallbladder in situ
Elderly patient
Patient with high operative risk
Patients choosing nonoperative management of gallbladder stones
Before laparoscopic cholecystectomy (where laparoscopic common bile duct exploration is unavailable or not selected)
4. Severe gallstone pancreatitis with biliary obstruction
5. Acute cholangitis

ERCP, endoscopic retrograde cholangiopancreatography.

TABLE 141-3 Indications for ERCP and Sphincterotomy for Choledocholithiasis

Methods of Stone Extraction

Standard (Basket and Balloon Catheters) Following identification of a common duct stone, a sphincterotomy is usually performed. The length of the cut is dictated by the length of the endoscopically visible intramural bile duct and by the size of the stone. Balloon catheters are most useful for extracting one or more relatively small stones (<10 mm) in a nondilated duct. They are not as effective for extracting larger stones or small stones in a markedly dilated bile duct since the balloon will often slide past the stone. A major advantage of the stone retrieval balloon (as compared to the basket) is that it cannot become impacted, although the stone can. Catheters with balloons that inflate to 8 to 18 mm are commercially available. The catheter is advanced through the sphincterotomy into the bile duct proximal to the

stone using fluoroscopic guidance. The balloon is then inflated to the diameter of the duct and gentle traction is applied to deliver the stone through the sphincterotomy. Passage of the balloon catheter over a guidewire is often helpful to allow frequent easy catheter passage through the sphincterotomy without trauma, to position the balloon proximal to the stone without pushing the stone proximally, and to avoid repeated cystic duct entry. Stone retrieval baskets with different configurations, length/width, wire types, and number of wires are commercially available. Settings where a basket may be preferred over a balloon include larger stones (>10 mm), intrahepatic stones, smaller stones in a dilated duct, and stones that are larger than the downstream duct (e.g., stone proximal to a stricture) (Fig. 141-3). The basket is advanced through the sphincterotomy and partially or fully opened alongside or above the stone taking care not to push the stone up the duct. The basket is then moved to and fro with the stone adjacent to the widest portion of the basket. Once captured, the stone is removed with gentle traction. Usually there is resistance at the sphincterotomy orifice. By deflecting the scope tip down, extra force in the correct axis is applied, often allowing for successful stone removal. Vigorous pulling on the endoscope is sometimes necessary but has the potential of causing a duodenal tear.

FIGURE 141-3. A Dormia basket has been used to capture a stone (*S*) that is approximately 12 mm in diameter. Two smaller stones (*s*) are seen in the dilated common bile duct, distal to the stone retrieval basket.

In experienced centers, common duct stones can be successfully removed in 80% to 90% of patients after sphincterotomy using standard baskets and balloon catheters. [11], [12] and [13] Difficulty of clearing or failure to clear the common duct of stones may occur for a variety of reasons. In most cases, stone size is the major determinant of success. In one series, [14] stones less than 10 mm in diameter (*n* = 21) were all removed successfully, whereas only 3 of 25 (12%) larger than 15 mm were cleared via stone extraction with balloons and baskets. Stones greater than 15 mm are generally considered to be large; equally important, however, are stone factors such as number, consistency, shape, and location, and ductal factors such as contour, diameter at the level of and distal to the stone, and the presence of coexisting pathology such as a stricture or tumor.

Lithotripsy Techniques A variety of lithotripsy techniques (mechanical, electrohydraulic, laser, and extracorporeal shock wave lithotripsy [ESWL]) and dissolution therapies have been used to facilitate the retrieval of stones not removable by standard methods. [15], [16] The simplest endoscopic adjunct for the management of common duct stones that have failed to be removed by conventional baskets and balloons is the mechanical lithotripter or crushing basket. It is a safe, effective, low-cost procedure that can be performed at the time of the initial ERCP. Although mechanical lithotripters have been available for more than 15 years, the design improvements made over the last several years have been considerable and made these devices relatively easy to use. Using this technique, the stone is captured in a strong wire basket that has been advanced through a metal sheath. Longitudinal traction is then applied by turning a crank handle withdrawing the basket into the metal sheath, resulting in stone fragmentation or wire breakage. There are two types of mechanical lithotripters commercially available. The first is a metal sheath (e.g., Soehendra mechanical lithotripter) that is advanced over a standard basket after the basket handle has been cut and the endoscope removed. This technique is fairly simple once the stone is captured but has the disadvantages of destroying a basket with each use and requiring endoscope removal (and replacement to remove the fragments). This lithotripter is particularly useful for basket impaction and should, therefore, be available for all stone cases. The second lithotripter is a through-the-scope model that has a three-layer system consisting of a basket, plastic sheath, and a larger metal sheath. Using this model, the stone is captured in standard fashion while the metal sheath is in the endoscope channel. If lithotripsy is necessary for stone removal, the metal sheath is advanced and the stone crushed against it. In experienced centers, mechanical lithotripsy allows for removal of more than 85% to 90% of difficult bile duct stones, which are refractory to standard extraction techniques. [17], [18], [19] and [20] Failures of mechanical lithotripsy are typically due to inability to engage the stone within the basket and rarely to insufficient shearing power to fragment the stone. Lithotripsy techniques using ESWL or intracorporeal (laser or electrohydraulic) modalities are acceptable adjuncts to standard endoscopic management in attempting bile duct clearance. [21] The choice between these methods or surgery largely depends on availability as they are usually concentrated in tertiary centers. Intracorporeal lithotripsy can be achieved by producing a shock wave directly on the surface of the stone with either a flexible electrohydraulic probe or a flexible quartz fiber to deliver light from a laser. Both techniques require a fluid medium that is delivered coaxially along the probe or through a nasobiliary tube (NBT). Because of the risk of bile duct injury (electrohydraulic has a greater risk of injury than the pulsed dye laser), intracorporeal lithotripsy is usually performed under direct endoscopic control using the mother-baby endoscope system. In this technique, a small caliber endoscope (baby scope) is advanced through the working channel of the duodenoscope (mother scope) into the bile duct. The laser fiber or electrohydraulic probe is advanced through the working channel of the baby scope and apposition with the stone is assured under direct vision. Laser lithotripsy has been possible under fluoroscopic guidance with the development of a device (smart laser) that can identify bile duct stones by analyzing backscattered light, interrupting the pulse in case of tissue contact. [22] Complete duct clearance rates with intracorporeal lithotripsy techniques range from 80% to 90%. [23], [24], [25], [26] and [27] The main advantages of electrohydraulic lithotripsy over laser technology are its low cost and portability; however, the potential risk of bile duct injury is greater. ESWL is used to treat bile duct stones in a fashion similar to renal and gallbladder applications. Most centers use machines that require fluoroscopy to target the stones and rely on injection of contrast material through an NBT, [28] but some centers have reported good visualization of the stone in 90% of patients with ultrasound imaging. [21], [29] Complete stone clearance can be expected in approximately 80% of patients, but it usually requires a number of ESWL and endoscopic sessions. [21], [28], [29] and [30]

Dissolution Therapy Contact dissolution of biliary stones has been attempted by perfusing the bile duct with solvents administered via an indwelling NBT, percutaneous transhepatic catheter, cholecystostomy tube, or T-tube. The results with these agents (monoctanoin and methyl- *tert*-butyl ether) have been disappointing due to incomplete stone dissolution and the potential for complications from these solvents. [31], [32] Due to their low efficacy and high morbidity, contact dissolution has not assumed an important role in patients with refractory bile duct stones.

Stents and Nasobiliary Tubes When stone extraction is incomplete or failed, biliary drainage should be established to prevent stone impaction and cholangitis. In most situations, this therapy serves as a temporizing measure allowing for improvement in the patient's clinical condition pending repeat attempts at stone removal. The stent (or NBT) is placed so that one limb is above the stone and the other is in the duodenum (in the case of an NBT, the end of the tube is brought out the patient's nose and connected to a drainage bag) (Fig. 141-4). Most authorities recommend double-pigtail stents, although favorable experience is reported with straight 10F stents. [33], [34], [35], [36], [37] and [38] NBTs allow for repeat contrast injection without the need for another ERCP to visualize the biliary tree. However, they are often poorly tolerated, frequently dislodged by a confused or uncooperative patient, and out-of-hospital tube management is not optimal. Endoprostheses are better tolerated but migration or occlusion may occur, leading to recurrent symptoms of biliary obstruction and cholangitis.

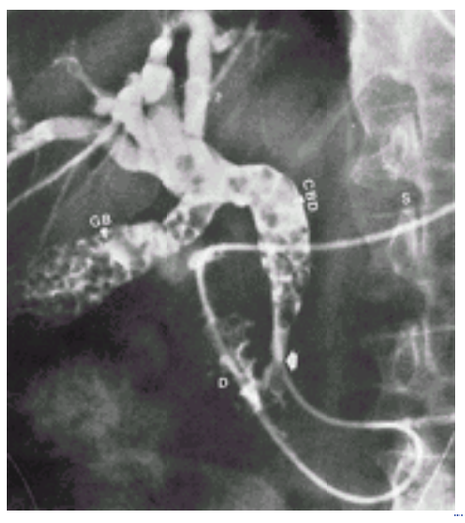

FIGURE 141-4. A nasobiliary tube (NBT) has been placed to provide temporary biliary drainage in this patient with multiple small common bile duct (*CBD*) and gallbladder (*GB*) stones. An NBT traverses the stomach (*S*) and forms a loop in the duodenum (*D*) before entering the papilla (*arrow*).

Biliary stenting not only serves to drain the bile duct but may aid in mechanically fragmenting the stone facilitating subsequent attempts at endoscopic removal. [39], [40] The addition of oral dissolution therapy may also soften and reduce the size of the stone, aiding endoscopic removal. In a study by Johnson and colleagues, [41] 9 of 10 patients with nonextractable stones treated with ursodeoxycholic acid plus stenting had clearance of their bile duct after a mean of 2.7 follow-up procedures in contrast to none of 12 patients treated with stenting alone and a mean of 5.3 follow-up procedures. Long-term internal stenting was believed to be a good palliative measure in elderly and high-risk patients with nonextractable bile duct stones. In several reports, the rate of late complications (primarily cholangitis) was 12% to 15%. [33], [34], [36], [37], [39] The enthusiasm for this approach has been tempered by the results of a study [38] in which 34 complications occurred in 23 of 58 patients (40%) stented for a median of 36 months (range, 1–117 months). There were 9 (16%) biliary-related deaths occurring at a median of 42 months. The rate of late complications was shown to increase proportionally with time (16% at 1 year and 50% at 4 years). The authors advised that permanent stenting be restricted to patients unfit for elective surgical, endoscopic, or percutaneous treatments and in patients with short life expectancy. This recommendation was supported by the results of a randomized study of endoprosthesis insertion versus standard duct clearance techniques in a group of high-risk patients with symptomatic stones. [42] DePalma and Catanzano [43] retrospectively compared the results of endoscopic biliary stenting ($n = 31$) with those of surgery ($n = 37$) in 68 patients over 70 years of age with failed endoscopic bile duct clearance. Although early complications were significantly less frequent in the stented group (12.9% vs 29.7%; $P < 0.0005$), the complications (35.5% vs. 8.1%; $P < 0.001$) and biliary mortality (9.6% vs. 0%) during long-term follow-up were significantly more common in the stented group.

Endoscopic Balloon Dilation of the Biliary Sphincter for Stone Removal Because of the significant risks and unknown long-term effects of sphincter ablation, some authorities have suggested that small common duct stones should be removed after papillary balloon dilation. The main theoretical advantages of not cutting the sphincter are that acute complications might be less frequent and by preserving sphincter function, long-term complications (a particular concern in younger patients undergoing laparoscopic cholecystectomy) may be avoided. Bergman and colleagues [44] reported 36 biliary tract complications in 22 patients (among 93 patients) followed for a mean of 15 years after sphincterotomy and stone removal. The same group [45] later found that, after biliary sphincterotomy, the function of the biliary sphincter was permanently lost and was associated with bacterial colonization, presence of cytotoxic components in the bile, and chronic inflammation of the biliary ductal mucosa. In contrast, manometric studies have suggested recovery of sphincter function within a few weeks of balloon dilation of the sphincter. [46] In this technique, the papilla is dilated with an 8- to 10-mm hydrostatic balloon. Following dilation, the stones are removed using stone retrieval balloons, baskets, or mechanical lithotripsy. Initial reluctance to use this technique arose because of concern for a high risk of postprocedural pancreatitis and cholangitis. A prospective randomized trial comparing ES ($n = 101$) to papillary balloon dilation ($n = 101$) for removal of common duct stones achieved complete stone removal in one endoscopic session in 89% (9% required additional sphincterotomy) after balloon dilation and 91% after sphincterotomy. [47] Mechanical lithotripsy was more frequently required in the balloon dilation group (31% vs. 13%; $P < 0.005$). Early complications were similar for the two groups (17% vs. 24%; $P = 0.29$). Seven percent of patients in each group developed post-ERCP pancreatitis. In another randomized study in patients with Billroth II anatomy, balloon dilation was found to be as effective as sphincterotomy for stone removal with fewer complications. [48] These favorable results for balloon dilation stand in marked contrast to the preliminary results of a multicenter randomized trial conducted in the United States. [49] In this study of 177 patients, procedure success was similar in the balloon dilation (94%) and ES (100%) groups. However, procedure time was significantly longer (51 minutes vs. 40 minutes; $P = 0.011$) and overall morbidity was significantly greater (12% vs. 1%; $P = 0.006$) in the balloon dilation group. Among the 11 episodes of pancreatitis in the balloon dilation group, four were graded severe; two patients died. In another randomized study comparing the two techniques, the frequency of recurrent bile uct stones and cholecystitis at one year were similar. [50] Moreover, at one year after balloon dilation, the common bile duct pressure, sphincter of Oddi basal and peak pressure, and sphincter of Oddi contraction frequency were significantly lower than predilation values. These results suggest that balloon dilation should probably not be used routinely but should be reserved for patients at high risk for sphincterotomy complications such as patients with uncorrectable coagulopathy. [51], [52]

Interface of ERCP and Laparoscopic Cholecystectomy

Laparoscopic cholecystectomy has become the standard, accepted, and preferred technique for the treatment of gallbladder stones. This approach is associated with less postoperative pain, reduced hospitalization time, shorter convalescence, and better cosmetic results than open cholecystectomy. However, the laparoscopic management of common duct stones is much more complex than cholecystectomy alone, requiring advanced surgical skills and sophisticated instrumentation, and it is not widely available. Thus, ERCP plays an integral role in the treatment of common duct stones in the laparoscopic cholecystectomy era. The timing and need for ERCP in relation to the laparoscopic cholecystectomy is dependent on the likelihood of stones being present (low, medium, and high), the skill of the endoscopist, and the ability of the laparoscopist to perform common duct exploration. [53] There appears to be little value to routine ERCP prior to laparoscopic cholecystectomy in patients with a low likelihood of having bile duct stones. When comparing the low yield of detecting clinically important anatomic variants and unsuspected bile duct stones with the generally accepted 3% to 7% ERCP complication rate, the use of routine ERCP prior to cholecystectomy cannot be justified. [54] Patients judged to have a high likelihood of harboring duct stones are likely to benefit from preoperative ERCP and stone extraction (if stones are present). [53] Patients in the medium-risk group create a diagnostic and therapeutic dilemma that must be resolved based on the skills of the endoscopist and laparoscopist at each particular center. Endoscopic ultrasound, magnetic resonance cholangiopancreatography (MRCP), and spiral CT cholangiography appear to have a high sensitivity and specificity for stone detection; however, these imaging modalities [55], [56], [57], [58] and [59] are operator-dependent and limited to select centers. The value of these noninvasive and less invasive techniques in the evaluation of patients with a medium risk of bile duct stones needs further evaluation.

Ultimately, a single laparoscopic procedure that treats both cholelithiasis and choledocholithiasis in a single setting would be the best approach in the majority of patients. This approach appears to be cost-effective and associated with a shorter hospital stay than a two-stage procedure (preoperative ERCP with sphincterotomy followed by laparoscopic cholecystectomy). [60], [61] and [62] When these laparoscopic skills become widely disseminated, the use of ERCP will be relegated to its well established role in the open cholecystectomy era: the treatment of acute cholangitis, severe gallstone pancreatitis, retained common duct stones, and complications of biliary surgery.

Acute Gallstone Pancreatitis

In Western countries, gallstone disease is the leading cause of acute pancreatitis, accounting for 34% to 54% of the cases. [63] Most patients with acute gallstone pancreatitis (AGP) have a mild attack and can be treated conservatively. However, case fatality in severe pancreatitis remains unacceptably high, approaching 10%. In the open cholecystectomy era, urgent surgical intervention for severe AGP did not gain general acceptance because of the increased morbidity and mortality associated with this approach. [64], [65] Coincident with these surgical reports were uncontrolled endoscopic series reporting the efficacy and safety of ERCP and ES in the setting of AGP. [66], [67] Although the results were encouraging, the studies varied in their criteria for patient selection and timing of ES in relation to the acute attack (many were performed in the recovery phase when surgery is also safe). These early series prompted the three randomized controlled trials, [68], [69] and [70] which now serve as the basis for the endoscopic treatment of AGP. The therapeutic principle for ES in AGP is simply removal of the obstructing calculus and reestablishment of bile and pancreatic juice flow. [71]

In a randomized prospective controlled trial from the United Kingdom, 121 AGP patients either received conventional therapy (i.e., gut rest, analgesics, intravenous fluids, and antibiotics) or underwent urgent (within 72 hours after admission) ERCP with ES and stone extraction (if stones were present in the common bile duct at ERCP). [68] Patients were stratified by the predicted severity of their attacks sing the modified Glasgow system. Choledocholithiasis was found in 25% of patients with predicted mild attacks and 63% with predicted severe attacks. The four important findings were:

1. ERCP could be safely performed in the setting of gallstone pancreatitis.
2. There was a significant reduction in the major complications of patients who underwent urgent ERCP and ES.
3. The reduction of morbidity was only apparent in those patients with predicted severe attacks (61% vs. 24%; $P = 0.007$).
4. There was a significant reduction in the hospital stay for those with severe attacks treated with urgent ERCP and ES (median, 9.5 days vs. 17 days, $P = 0.03$).

The mortality rate was improved, but the difference was not statistically significant.

A second randomized controlled study was performed by the Department of Surgery at the University of Hong Kong. [69] One hundred ninety-five patients with acute pancreatitis were randomized to early ERCP (within 24 hours of admission) or conservative therapy. Although the methodology, patient selection, and the assessment of the severity of the acute pancreatitis used in this study differed from the United Kingdom study, the results in the subgroup of patients with gallstone pancreatitis ($n = 127$) were quite similar. Patients with mild pancreatitis had a similar morbidity and mortality regardless of the therapy. In contrast, patients with predicted severe attacks undergoing endoscopic therapy had a lower complication rate (54% vs. 13%; $P = 0.003$) and a lower mortality rate (18% vs. 3%; $P = 0.07$) than patients

treated conservatively.

The third study [70] was a prospective multicenter randomized controlled study from Germany in which 238 patients with AGP and no evidence of severe biliary obstruction (severe biliary obstruction defined as a bilirubin >5 mg/dL) were randomized to ERCP with ES and stone extraction or conservative therapy within 72 hours of symptom onset. This study attempts to address the major criticism of the United Kingdom and Hong Kong studies: the need to exclude patients presenting with concomitant cholangitis because these patients are known to benefit from ERCP. The two treatment groups did not differ significantly for mortality (11% vs. 6% overall mortality; 8% vs. 4% AGP mortality; ERCP vs. conservative therapy) or overall complications (46% vs. 51%; ERCP vs. conservative therapy) regardless of the predicted severity of the pancreatitis. However, respiratory failure was more frequent in the ERCP group (12% vs. 5%; $P = 0.03$), and jaundice was more frequent in patients who received conservative treatment (11% vs. 1%; $P = 0.02$).

Although all three studies conclude that there was no difference in outcomes for patients with mild pancreatitis treated conservatively or by ERCP, only the study from Germany suggested that early ERCP was of no benefit in patients with severe gallstone pancreatitis. Although ERCP is clearly indicated in patients with AGP complicated by cholangitis or biliary obstruction, its role in the setting of severe AGP alone warrants further investigation. A metaanalysis [72] of these three published studies and an abstracted randomized study [73] revealed a statistically significant reduction in morbidity from 38% to 25% and mortality from 9% to 5% in the ERCP/ES group compared with the conservatively treated group. A subgroup analysis based on the severity of pancreatitis was not reported in this metaanalysis.

Acute Cholangitis

Cholangitis is a potentially life-threatening disease that results from bacterial infection of obstructed bile. Systemic toxicity occurs when the intraductal pressure is sufficiently elevated to cause reflux of the bacteria or endotoxin into the blood. [74] Thus, obstruction plays a key role by both increasing intraductal pressure and promoting bacterial overgrowth as a result of bile stasis. The most common cause of acute cholangitis is choledocholithiasis, which occurs in approximately 80% to 90% of unselected cases. [74] Other diseases less commonly associated with cholangitis include malignant obstruction of the bile duct, choledochal cysts, iatrogenic conditions (e.g., postoperative biliary stricture), papillary stenosis, biliary parasites, and the sump syndrome.

Therapy for cholangitis must be individualized because of the spectrum of severity of illness. Antibiotic therapy should be initiated promptly. Analysis of bile and stone cultures indicates that *Escherichia coli, Klebsiella* spp., *Enterobacter* spp., *Enterococcus* spp., and *Streptococcus* spp. are the most commonly isolated bacteria. [75] The antibiotic selected should preferably penetrate an obstructed biliary tree. [75] The majority of patients will respond to conservative management, allowing for a more elective approach to biliary decompression. [76] Urgent decompression is indicated if improvement is not seen within a few hours of initial resuscitation. [77] The latter group will, invariably, have a fatal outcome if conservative treatment is continued. [78]

The options for bile duct decompression include surgical, percutaneous, and endoscopic methods. Endoscopic intervention is now accepted as a definitive therapy for acute cholangitis. The advantages of ERCP are that it can delineate the cause of obstruction, facilitate sampling of bile for cultures, and decompress the biliary tree in a relatively short time with low morbidity. Biliary decompression is the goal of therapy and can be complete (e.g., stone removal) or temporary (e.g., placement of a stent without stone removal) pending more definitive management (this will allow stabilization of an unstable patient). The endoscopic procedure consists of a sphincterotomy with stone extraction or biliary drainage with an NBT or endoprosthesis.

Technical Considerations Ideally the patient should be stabilized or made as stable as possible prior to performing ERCP. Patients with respiratory compromise can have their ERCP performed while on ventilatory assistance. Although the procedure is best performed in a dedicated fluoroscopy room, unstable patients should undergo the ERCP in the ICU. A mobile fluoroscopy unit can be used but reports indicate that ductal decompression can be performed without fluoroscopic assistance in an ICU. [79] Since the intrabiliary pressure is increased in acute cholangitis, contrast injection should be limited to reduce further systemic seeding of bacteria. Enough contrast should be injected to define the anatomy and the cause of obstruction. Alternatively, the bile duct should be aspirated completely of infected bile prior to contrast injection. Aspirated bile should be cultured. In a stable patient, definitive therapy can be performed. In an unstable patient, the length of the procedure should be limited. In such cases, a stent or NBT should be placed and once the patient is stabilized, more definitive therapy can be performed. NBTs are available in a variety of diameters with and without pigtail tips and duodenal a-loops (to maintain the duodenal position). NBT placement is performed in several steps. Following cannulation of the bile duct, a guidewire is advanced through the catheter into the intrahepatic biliary tree. [80] The cannula is withdrawn over the guidewire, keeping the guidewire in position. The NBT is advanced into the biliary tree over the guidewire and the tip usually positioned in an intrahepatic duct. The endoscope is then slowly withdrawn over the NBT by coordinated efforts of the endoscopist and assistant. While watching fluoroscopy, the endoscopist is advancing the NBT and the assistant withdraws the endoscope at 1- to 2-cm increments keeping the NBT in the same position. Although the NBT now emerges through the mouth, it is preferable to bring it out through the nose using a transfer tube. The NBT is taped to the cheek or forehead of the patient and its tip is connected to a drainage bag. Biliary endoprostheses are generally made of polyethylene and are available in a variety of diameters and shapes. The Amsterdam or barbed straight stents are most commonly used for long-term therapy because they offer the best flow characteristics and the longest patency rates. Plastic stents larger than 8.5F require use of a large diameter working channel (e.g., 4.2 mm) duodenoscope. Metal expandable stents, which offer superior bile drainage, are discussed later in the chapter. Double pigtail or straight stents can be used to prevent stone impaction. The equipment needed for endoprosthesis placement include guidewires, guide catheters, pusher tubes, and stents of varying diameters, lengths, and shapes. The initial steps of placing a biliary endoprosthesis are the same as for placing an NBT. After selective biliary cannulation is performed, a guidewire is advanced proximal to the obstruction. The catheter is removed while keeping the guidewire in position. When placing larger diameter stents (usually 10F and 11.5F), stricture dilation (if a stricture is present) with a hydrostatic balloon or dilating catheter may be necessary. If the stent is larger than 7F, a guide catheter (usually 6F) is advanced over the guidewire to stiffen the system to facilitate stent placement. The stent is then advanced over the guidewire and guide catheter using a pusher tube. Through coordinated efforts of the assistant and endoscopist, the tip of the stent is positioned above the stricture (if present) or stone, with the distal end in the duodenum. The stent is deployed by removing the guide catheter and guidewire while keeping the pusher tube against the tip of the stent. Delivery systems that allow passage of the stent, guide catheter, and pusher tube as a single unit have been developed, simplifying stent deployment. The ideal position of the endoprosthesis is for the proximal barb to be located at least 1 to 2 cm above the superior margin of the stricture or stone and the distal end of the endoprosthesis to protrude 1 cm into the duodenum, with the barb preferably at the level of the papilla.

Results The high morbidity and mortality associated with surgical and percutaneous therapy for acute cholangitis prompted evaluation of the safety and use of endoscopic management. [81], [82] and [83] In a retrospective analysis, Leese and colleagues [84] reported on 71 patients with cholangitis due to stones treated by early decompression either surgically ($n = 28$) or by ES ($n = 43$). Early surgery was associated with significantly higher 30-day mortality (21% vs. 5%) and morbidity (57% vs. 8%) than sphincterotomy. The endoscopic group was significantly older and had more medical risk factors than the surgical group and there were no significant differences in the severity of cholangitis. Leung and associates [76] reported their experience in a retrospective analysis of 105 patients with acute calculous cholangitis who did not respond to conservative management and underwent urgent endoscopic decompression at a mean of 1.5 days after admission. Thirty-nine percent of patients had coexisting medical problems, 85% had Charcot triad, and 40% were in shock at the time of admission. Endoscopic drainage was successful in 102 patients (97%). Ninety-seven percent of patients responded with striking improvement of abdominal pain and 93% had resolution of fever within 3 days. The overall 30-day mortality was 5%. Among those in shock, 2 out of 4 drained after 72 hours died, compared with 3 out of 38 drained before 72 hours. There were no deaths in the group without shock irrespective of the timing of drainage. The mortality of 5% compares favorably with that of urgent surgical intervention, which has been reported to be greater than 40% in some series. [85] The ERCP complication rate was 5% and was limited to 5 postsphincterotomy bleeding episodes managed by endoscopic techniques. The safety and efficacy of endoscopic therapy were corroborated in a large retrospective study of 947 patients with cholangitis due to stones ($n = 898$) or stricture ($n = 49$). [86] In a randomized prospective study, Lai and colleagues [87] compared the safety and efficacy of biliary decompression by surgical and endoscopic techniques in 82 patients with severe cholangitis due to stones. Patients treated with laparotomy and common bile duct exploration had a significantly higher morbidity (64% vs. 34%) and mortality (32% vs. 10%) compared to those treated with endoscopic therapy. These and other studies clearly demonstrate the efficacy and safety of biliary decompression either as a definitive therapy or as a temporizing measure pending more definitive intervention once the patient is stabilized.

BILIARY DRAINAGE PROCEDURES

Benign Biliary Strictures

Postoperative Postoperative stricture(s) of the extrahepatic bile duct occur after 0.25% to 1% of cholecystectomies. [88] Most such lesions present as abnormal liver tests, obstructive jaundice, and cholangitis within 2 to 3 months postoperatively, although a much more delayed presentation may occur. In contrast, when the common duct has been completely occluded with a clip, progressive jaundice will become obvious early in the postoperative period. The cholangiogram commonly shows a short, smooth narrowing near the cystic duct stump and surgical clips with proximal duct dilation (Fig. 141-5). Strictures greater than 2 cm in length, those with clips placed securely across the duct, or those associated with resected segments of duct require operative management. Irrespective of the site or pathogenesis of the strictures, the primary aim of endoscopy is to pass a guidewire through the stricture to permit passage of dilators (balloon or catheter) and stents.

Sphincterotomy prior to stricture manipulation permits greater instrument maneuverability. The preferred treatment of short, simple strictures is balloon dilation to 6 to 10 mm with ultimate placement of two to three 10F plastic stents across the stricture and extending into the duodenum. The stricture is redilated and stents are exchanged at 3- to 4-month intervals for 8 to 12 months until the stricture profile is nearly as open as the downstream adjacent duct. The goals of treatment are to render the patient free of symptoms and to achieve sustained normalization of the liver tests after the stents are permanently removed. Because strictures can recur many years after therapy, the long-term outcome following endoscopic intervention is of critical concern. In three uncontrolled endoscopic studies [89], [90] and [91] a good to excellent result was achieved in 70% to 80% of patients followed for an average of 4 years. Assessment of good to excellent outcomes was based on resolution of symptoms and normalization or significant improvement in biochemical tests and radiographic findings. Two long-term follow-up studies of endoscopic stenting of biliary strictures have been reported. [92], [93] Bergman and colleagues [92] reported late complications (median follow-up of 9.1 years) in 34% of 44 patients undergoing biliary stenting with two 10F stents for a maximum of one year. Restenosis at the site of the original stricture occurred in 20% of patients. Costamagna and colleagues [93] undertook a more aggressive endoscopic approach. Each patient received as many large-diameter stents as necessary (based on stricture tightness and bile duct diameter) to eliminate the stricture. Stents were exchanged electively at 3-month intervals and were removed upon complete stricture resolution. There were no symptomatic recurrent biliary strictures among the 40 patients completing the stenting protocol during a 49-month follow-up period. In a nonrandomized study, Davids and associates [94] compared the outcome of 35 patients treated surgically (biliary-enteric anastomosis) and 66 patients managed with endoscopic balloon dilation and stenting (stents changed every 3 months for 1 year). The mean follow-up interval for the surgical and endoscopically treated patients was 50 and 42 months, respectively. Eighty-three percent of patients in both groups attained good to excellent results. The total morbidity rates (surgery, 26%; endoscopy, 35%) and stricture recurrence rates (17% for both groups) were similar for the two groups. Collectively, these data support the use of endoscopic therapy in patients with postoperative biliary strictures. Surgically fit patients who fail initial endoscopic therapy or have recurrent strictures are best managed by a bilioenteric bypass. Although metal expandable stents offer prolonged patency compared to plastic stents, their use for treating benign postoperative biliary strictures should be discouraged. Dumonceau and associates [95] reported mucosal hyperplasia and Wallstent occlusion in all six patients who underwent this therapy.

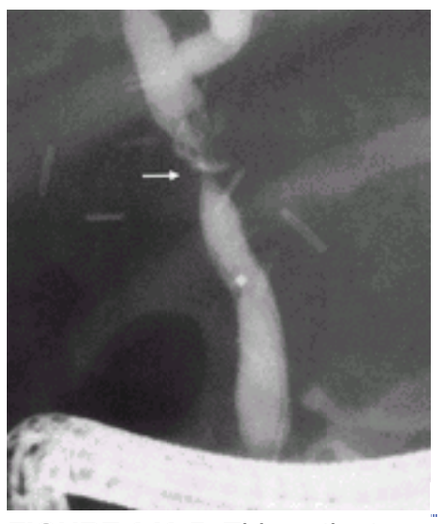

FIGURE 141-5. This patient presented with obstructive jaundice 2 months after laparoscopic cholecystectomy. The cholangiogram shows a common bile duct stricture (*arrow*). Note the clips in the region of the common duct.

Endoscopic techniques have been used to treat biliary strictures complicating orthotopic liver transplantation. [96] Such strictures are often treated in a fashion similar to strictures occurring after other biliary tract surgeries. Anastomotic strictures appear to be more responsive to endoscopic therapy than strictures occurring at nonanastomotic sites. [96]

Distal Common Bile Duct Strictures Secondary to Chronic Pancreatitis Intrapancreatic common bile duct strictures have been reported to occur in 3% to 46% of patients with chronic pancreatitis (Fig. 141-6). Deviere and colleagues [97] evaluated the use of biliary stenting in 25 patients with common bile duct obstruction and significant cholestasis (alkaline phosphatase greater than two times the upper limits of normal) secondary to chronic pancreatitis. Nineteen patients had jaundice, and seven presented with cholangitis. The patients were treated with ES followed by insertion of one or two 10F biliary stents placed across the stricture. The stents were changed when clinical or ultrasonographic evidence of blockage was present. Cholestasis, hyperbilirubinemia, and cholangitis resolved in all patients following stent placement. The late follow-up (mean, 14 months; range, 4 to 72 months) on 22 patients was much less satisfactory. One patient died 1 month after treatment from acute cholecystitis and postsurgical complications, whereas a second died 10 months after stenting of sepsis that was believed to be caused by stent blockage or dislodgment. Stent migration occurred in 10 patients and stent blockage in 8, resulting in cholestasis with or without jaundice ($n = 12$), cholangitis ($n = 4$), or no symptoms ($n = 2$). These patients were treated by stent replacement or surgery ($n = 7$). Ten patients continued to have a stent in place (mean follow-up, 8 months) and remained asymptomatic. Only 3 patients required no further stents because of resolution of their biliary stricture. Other authors have also reported a low stricture resolution rate ranging from 11% to 28%. [98], [99] and [100] The results of the aforementioned studies stand in distinct contrast to those of Vitale and associates [101] who reported that 20 of 25 patients undergoing plastic biliary stenting for a median period of 13.3 months remained stent-free without stricture recurrence during a 32-month follow-up.

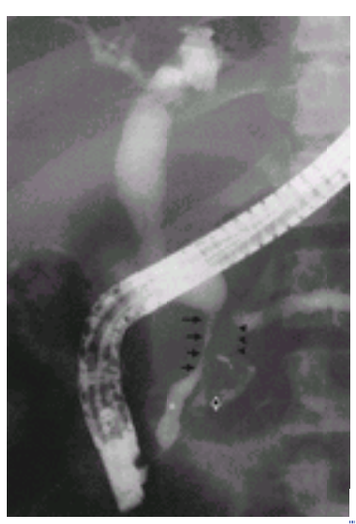

FIGURE 141-6. Chronic pancreatitis-induced common bile duct stricture. The cholangiogram shows a 2-cm common bile duct stricture (*closed arrows*) with proximal dilation. Note the pancreatic duct stone (*open arrow*) and stricture (*arrowheads*).

Because of the disappointing results with plastic stents and the concern for the high morbidity associated with surgically performed biliary drainage procedures in alcoholic (frequently debilitated) patients, the use of uncoated expandable metal stents was evaluated. [102] Twenty patients were treated with a 34-mm long metal stent that becomes 10 mm in diameter when fully expanded. The short length of the stent was chosen so that surgical bypass (e.g., choledochoduodenostomy) would still be possible if necessary. Cholestasis ($n = 20$), jaundice ($n = 7$), and cholangitis ($n = 3$) resolved in all patients. Eighteen patients had no further biliary problems during a follow-up period of 33 months (range, 24 to 42 months). Two patients (10%) developed epithelial hyperplasia within the stent resulting in recurrent cholestasis in one and jaundice in the other. These patients were treated endoscopically with standard plastic stents, with one of these patients ultimately requiring surgical drainage. The authors concluded that this therapy could be an effective alternative to surgical biliary diversion but that longer follow-up and controlled trials will be necessary to confirm these results. The studies cited here and others indicate that plastic biliary stents are a useful alternative to surgery for short-term treatment of chronic pancreatitis-induced common bile duct strictures complicated by cholestasis, jaundice, and cholangitis. This therapy should also be considered for high-risk surgical patients. However, the long-term efficacy of this treatment is much less satisfactory because stricture resolution rarely occurs; operative intervention appears to be a better long-term solution for this problem in patients of average risk. More data on the long-term outcome, preferably in controlled trials, are necessary before the expandable stents can be advocated for this indication. Trials of membrane-coated metal stents and removable coil spring stents are awaited.

Primary Sclerosing Cholangitis

Cholangiographic imaging is the gold standard for diagnosing primary sclerosing cholangitis (PSC), although conditions that mimic PSC must be excluded (Table 141-4). In the variant, small duct PSC, the cholangiogram is normal. [103] Both percutaneous transhepatic cholangiography (PTC) and ERCP can be used to show the characteristic changes of PSC. The choice between PTC and ERCP will depend on local and regional expertise and availability. However, where available, ERCP is the preferred modality because:

- the often small fibrotic ducts of PSC may be difficult to puncture via the percutaneous route
- ERCP has a better safety profile
- the pancreatic duct and cystic duct, which may be involved in up to 20% of PSC patients, can be examined at ERCP.

Neoplasms
Metastatic carcinoma
Cholangiocarcinoma
Lymphoma
Cirrhosis
Polycystic Liver Disease
Caroli Disease
Multiple Hepatic Abscesses
Systemic Fungal Infection
Bacterial Cholangitis
Secondary Sclerosing Cholangitis
Associated with bacterial infection and impacted intrahepatic ductal stones
Associated with congenital immunodeficiency syndromes, opportunistic infections (e.g., cytomegalovirus)
Acquired Immunodeficiency Syndrome (AIDS)
Associated with cytomegalovirus, *Cryptosporidium*, papillary stenosis
Graft-Versus-Host Disease
Hepatic Arterial Ischemia
Posttraumatic
Postsurgical (including intentional or inadvertent ligation of hepatic artery and complications from liver transplantation)
Associated with intraarterial 5-fluorodeoxyuridine (5-FUDR)
Preservation injury of biliary tract in the donor liver
Chronic allograft rejection in liver transplantations

TABLE 141-4 Conditions Producing Cholangiograms That May Mimic Primary Sclerosing Cholangitis

PTC is reserved for ERCP failures and patients with altered anatomy. The role of MRCP and spiral CT cholangiography in the diagnosis of PSC awaits further study. [104]

The classic cholangiographic features in PSC are diffuse multifocal strictures of the intrahepatic and extrahepatic bile ducts (Fig. 141-7). [105] These strictures are usually short with intervening normal or dilated segments giving a beaded appearance. Other frequent findings on the cholangiogram include pseudodiverticula, mural irregularities, and biliary stones and "sludge." [106], [107]

FIGURE 141-7. This patient has primary sclerosing cholangitis involving the extrahepatic and intrahepatic ducts. There are multiple strictures of the common bile duct (*open arrows*) and intrahepatic ducts (*arrowheads*).

The rationale for endoscopic intervention is based on the hypothesis that progressive liver disease and deterioration of liver function may be aggravated or accelerated by back pressure from dominant strictures and stones or debris when present. It is further hypothesized that relief of obstruction may halt, delay, or even reverse progression to cirrhosis and liver failure. [108] Because no medical therapy has been definitively proven effective for PSC, a trial of endoscopic therapy in symptomatic patients seems reasonable. The indications for considering endoscopic management in PSC are the treatment of jaundice or pruritus, symptomatic cholangitis, deteriorating serum hepatic chemistries, and, when the concern for bile duct cancer is high, for purposes of tissue sampling. The most favorable candidates for therapy are those patients with a dominant extrahepatic stricture with or without stones and limited or no intrahepatic involvement. Such ideal anatomy is uncommon.

Endoscopic Techniques If therapeutic skills are not readily available, care should be taken to avoid injecting more than 2 to 3 cc of contrast media proximal to high-grade strictures. Not uncommonly, contrast media will take the path of least resistance and enter the cystic duct and gallbladder; intrahepatic filling is therefore limited. Preferably, a balloon catheter is then manipulated above the cystic duct takeoff, and higher-pressure injection of the intrahepatic radicles is performed with the balloon inflated. Moreover, since the risk of post-ERCP cholangitis in PSC may be as high as 20% to 30% following diagnostic and therapeutic procedures (particularly in patients where obstructed segments are not decompressed), antibiotic prophylaxis has been advocated. All therapeutic procedures aim at improving bile flow. The endoscopic techniques that may be used to achieve this goal are ES, stone/sludge removal, stricture dilation with balloons and catheters, placement of stents and NBTs (with or without instillation of corticosteroids and saline lavage), and combinations of therapy. Many authorities advocate performing ES as this will permit easier access to the biliary tree for other therapeutic maneuvers. Moreover, the sphincter may be involved in the fibrotic process. A potential disadvantage of sphincterotomy is the destruction of the natural barrier between the duodenum and the biliary tree, which protects against bacterial and food contamination. Endoscopic stricture dilation and stenting have been reported in PSC patients since the early 1980s. The techniques have been standardized. Guidewires must first be advanced proximal to the stricture. This may be quite difficult in the PSC setting because of the irregular, high-grade strictures and the presence of pseudodiverticula. Perforation and false passages can occur relatively easily. Such perforations and false passages rarely seem to cause clinical problems, probably because they were made distal to the dominant stricture and the pressure in the bile duct at these sites is low. Once perforation or a false tract is created, however, further attempts to pass the guidewire through the natural stricture are often fruitless. We often put the patient on antibiotics and repeat the ERCP in a few days to a week depending on the clinical situation. Forceful pushing of wires and catheters should never be done. The development of hydrophilic guidewires has facilitated the passage of high-grade strictures. Once the guidewire has been advanced through the stricture, a dilating catheter or balloon can be used to dilate the stricture. The decision of which dilating instrument to use is a personal one as there are no studies that compare the two devices. However, while the diameter of the dilating catheter is dependent on the accessory channel size, no such limitation (for the most part) exists for dilating balloons. Strictures are generally dilated to match the size of adjacent normal ducts (i.e., 2 to 6 mm for intrahepatic ducts and 6 to 10 mm for extrahepatic ducts). Newer higher-pressure balloons permitting inflation to 10 to 12 atm have decreased the frequency of failure to obliterate the stricture waistline but probably carry a higher risk of duct perforation. Another option advocated by some authorities is placement of an NBT. The NBT can be advanced through the stricture and left in place. For the first few days, the drain is used simply for drainage. During this interval, the strictures open up by continuous pressure and movement of the NBT, creating space around the NBT. The catheter can then be used for instillation of saline or drugs (e.g., corticosteroids). Temporary biliary stenting has been advocated by some authorities particularly when dilation fails to

improve stricture patency sufficiently. Others suggest that stents should be avoided owing to the high incidence of cholangitis.

Results The goals of endoscopic intervention in PSC are to relieve jaundice and pruritus, treat cholangitis and theoretically delay the onset of biliary cirrhosis, and buy time prior to liver transplantation. Interpretation of the reported results of endoscopic therapy is difficult because there are no randomized controlled trials, therapies are not uniform, treated patients have variable anatomy, the definition of success varies, studies are generally small, the course of untreated PSC is variable, and there is no long-term follow-up. Johnson and colleagues [109] treated 35 symptomatic PSC patients (29 with cholangitis and 6 with jaundice alone) by dilation (balloon or catheter) with or without biliary stenting. During a mean follow-up period of 24 months, there was a significant reduction in the frequency of hospitalization for cholangitis, bilirubin, and stricture score. Cholangitis occurred shortly after treatment in six patients; five of the six had a biliary stent placed. As a result, these authors recommended avoiding biliary stents in PSC. A retrospective review of 85 PSC patients who underwent 175 ERCP procedures (75 diagnostic and 100 therapeutic) showed that endoscopic therapy was associated with a 15% major complication rate (7% pancreatitis and 8% cholangitis). [110] Clinical follow-up (median, 31 months) was obtained in 50 of 53 patients who underwent 85 therapeutic procedures. Twenty-eight patients improved clinically, while 21 felt the same, and one felt worse. Serum liver chemistries obtained within 3 months of the endoscopic intervention were significantly improved compared with pretreatment values. Overall, 41 of 53 patients (77%) had improvement of their clinical symptoms, liver function tests, or cholangiograms. Van Milligen de Wit and associates [111] reported results of stent therapy in 25 patients with PSC and dominant extrahepatic strictures. Stents were exchanged or removed electively at 2- to 3-month intervals or because of symptoms attributable to stent clogging. Endoscopic therapy was technically successful in 21 patients (84%). In these 21 patients, the results of all serum biochemical liver tests improved significantly within 6 months of stent therapy. During a median follow-up of 29 months (range 2 to 120 months) after stent removal, 12 patients (57%) remained asymptomatic with stable biochemical liver tests and 4 (19%) had clinical and biochemical relapse of disease that responded favorably to repeat endoscopic therapy. Early procedure-related complications occurred in 14% of the procedures. The value of short-term endoscopic stenting (mean, 11 days; range, 1 to 23 days) for 32 patients with dominant strictures has been reported by Ponsioen and colleagues. [112] Cholestatic symptoms improved in 83% and there were statistically significant reductions in abdominal pain, fatigue, and pruritus. Serum liver chemistries were significantly improved. Eighty percent of patients were free of reinterventions at 1 year and 60% at 3 years. Procedure-related complications occurred in 15% but there were no episodes of cholangitis. The authors advocated this technique because of its efficacy and it overcame the complications associated with stent occlusion. Kaya and colleagues [113] reported that stenting after balloon dilation (median stenting interval of about 4.5 months) of dominant strictures provides no additional benefit and is associated with more complications than balloon dilation alone. Baluyut and colleagues [114] found that repeated endoscopic treatments to maintain bile duct patency were associated with a significantly higher observed 5-year survival than predicted by the Mayo Clinic survival model. The value of saline lavage and infusion of steroids via NBT has been studied in small series with inconclusive results. [115], [116] Cholangiocarcinoma is a dreaded complication of PSC, occurring in 9% to 15% of patients. [103] The risk appears to be greatest in those patients with long-standing ulcerative colitis and cirrhosis. Surprisingly, studies suggest that there may be an inverse relation between the duration of PSC and the risk of cholangiocarcinoma. [117], [118] Sudden worsening of jaundice should raise the possibility of the development of cholangiocarcinoma. Cholangiographic findings that suggest malignant transformation include markedly dilated ducts of ductal segments proximal to a stricture, the presence of a polypoid mass, and progressive stricture formation. [106] Comparison with previous ERCP results is essential to signal the presence of complicating cholangiocarcinoma because with PSC uncomplicated by malignancy the cholangiographic appearance frequently remains static for years. [106] Unfortunately, early diagnosis of cancer is difficult because we lack a sensitive, specific serologic marker and bile duct tissue sampling is relatively insensitive. However, tissue sampling of any suspicious lesion found at ERCP is indicated. Although the use of ERCP in helping to make the diagnosis of PSC is clear, its therapeutic efficacy in improving the course of the disease appears highly likely but not definitively established. Clearly, symptomatic patients with dominant extrahepatic strictures are the best candidates for therapy.

Biliary Fistulae

Biliary fistulae most commonly occur as a complication of cholecystectomy, common bile duct exploration, inadvertent operative injury of the bile duct, or as a consequence of a local infection. Rarely, biliary fistulae result from long-standing untreated biliary tract disease. With more widespread use of laparoscopic cholecystectomy, the incidence of bile duct injury, including biliary fistulae, has increased. [119], [120] and [121] Bile leakage from the cystic duct remnant is among the most common injuries reported as a complication of laparoscopic cholecystectomy. The most common cause for cystic duct leaks involves imprecise application of clips on the duct or their subsequent dislodgment during the procedure. [122], [123], [124] and [125] Biliary fistulae may also arise from the intrahepatic ducts and common duct. The duct of Luschka, if present, is quite vulnerable to transection during cholecystectomy. [126] Clearly, distal obstruction from stone, stricture, or papillary stenosis increases the ductal pressure proximally and may promote and maintain the biliary fistula.

Postoperative bile duct leaks usually manifest within a week after surgery. [125], [127], [128] In a series [128] of 62 patients with postcholecystectomy leaks, presenting symptoms included abdominal pain in 89%, abdominal tenderness in 81%, fever in 74%, nausea and vomiting in 43%, and jaundice in 43%. Only 2% presented with a clinically detectable mass or ascites. Biochemical testing is usually nonspecific with variable elevations of the serum hepatic chemistries and white blood cell count.

A high index of suspicion for bile duct injuries after laparoscopic cholecystectomy should be maintained for any patient who fails to follow a smooth, uneventful postoperative course. Patients with suspected biliary fistulae often undergo abdominal ultrasound or CT scan to look for evidence of a biloma and HIDA scan to diagnose the leak. [53] However, direct cholangiography (most often by ERCP) is the most sensitive test to detect a biliary fistula. [128]

The treatment options for biliary leaks include percutaneous or endoscopic placed biliary drains or stents, and surgical drainage and repair of the leak. Patients with large bilomas should undergo percutaneous drainage (unless surgery is performed) of the fluid collection. Endoscopic therapy has been shown to be a definitive therapy in this setting with low morbidity. Patients with leaks from the cystic duct, duct of Luschka, and T-tube tract are optimal candidates for endoscopic treatment. However, patients with injuries of the common bile duct, common hepatic duct, and intrahepatic ducts can also be managed by endoscopic techniques.

The primary goal of endoscopic therapy is to decrease the pressure gradient between the bile duct and duodenum, allowing drainage of bile along the path of least resistance and away from the site of leakage (to allow the defect to seal). This can be accomplished with biliary sphincterotomy alone, stenting alone, NBT alone, or any combination thereof (Fig. 141-8). [53], [121], [127], [128], [129], [130], [131], [132], [133] and [134] Foutch and colleagues [135] compared the various endoscopic treatment options in 23 patients with biliary leaks arising from the cystic duct (*n* = 13), common duct (*n* = 6), and duct of Luschka (*n* = 4). Patients were treated by sphincterotomy alone (*n* = 4), stent alone (*n* = 6), sphincterotomy and stent (*n* = 12), and sphincterotomy and NBT (*n* = 1). The biliary fistula closure rate was 100% and was complicated by minor bleeding in one patient. Davids and associates [134] reported their results of endoscopic management for postoperative biliary fistulae in 55 patients. A sphincterotomy alone was performed when there was a cystic duct stump or hepatic radical fistula but no distal common bile obstruction. Residual stones were removed using conventional techniques. A 10F stent was placed when there was a history of operative biliary trauma, when a benign or malignant stricture was present, or when the common duct could not be completely cleared of stones. The biliary tract and site of leakage were visualized during ERCP in 98% of patients. Endoscopic treatment was attempted in 49 of 54 patients. Five patients had total bile duct obstruction caused by a clip or ligature and could not be treated endoscopically. Successful drainage was achieved in 48 of 49 patients. An excellent outcome (clinical and radiologic resolution of the biliary fistula) was achieved in 43. Despite adequate therapy, five patients had continuing sepsis resulting in death. Although no significant factors predictive of a favorable outcome could be defined, the clinical condition of the patient at the time of referral seemed to be of importance. All poor responders had been admitted to the ICU prior to endoscopic treatment.

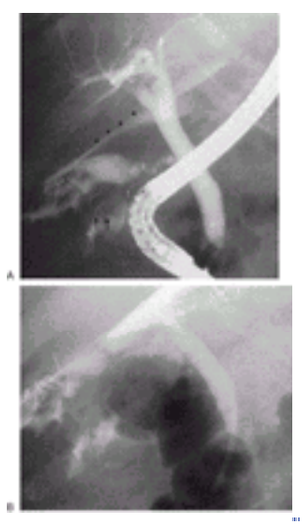

FIGURE 141-8. This patient underwent laparoscopic cholecystectomy for acute cholecystitis. She presented 10 days later with fever, chills, and abdominal pain. **A:** The cholangiogram shows contrast extravasation (*closed arrows*) from a residual gallbladder remnant (*open arrow*). Note subhepatic contrast collection (*arrowheads*). **B:** Endoscopic sphincterotomy was performed, and a 7F stent was placed with the tip of the stent proximal to the cystic duct takeoff.

Patients with clinically evident leaks not identified on cholangiography may have a disconnected duct. Kalacyi and associates [136] showed that MRCP may be helpful in identifying the upstream bile duct and the site of injury.

A sphincterotomy alone can be sufficient treatment for many patients with obstructed bile ducts with associated leaks when the cause for obstruction (papillary stenosis or choledocholithiasis) can be treated with such therapy. [137] However, sphincterotomy as a single treatment has failed when leaks were large. [138] Experience with NBTs to treat biliary leaks is limited, but available data from several investigators have been favorable. [138], [139] Since most fistulae seal in a few days, NBT placement is a reasonable option. The advantages of the NBT include the ability to monitor closure of the leak with repeat cholangiography, maximum decompression can be applied with suction, and the tubes are easily removed without the need for a second endoscopic procedure. However, the risk of infection when improperly cared for, poor patient acceptance and discomfort, and potential electrolyte disturbances from external drainage have been cited as potential disadvantages of this approach. [53] Furthermore, the documentation of closure seen on cholangiogram may not correlate with long-term healing of a disrupted duct. [135]

Biliary stents are a very effective therapy for resolving biliary leaks. The observation from several uncontrolled studies that patients treated with stents alone experience equally good outcomes compared to patients treated with a combination of stents and sphincterotomy suggests that sphincterotomy can be avoided in patients with otherwise unobstructed ducts. [134], [135] Therapeutic efficacy for 7F stents has been high. However, Foutch and associates [135] reported a 22% failure rate with 7F stents; these fistulae resolved by upsizing the stent to 10F. Larger caliber stents are certainly preferred when a concomitant stricture is present. In most reported series, stents were inserted with the proximal end positioned above the leak site. [134], [135] It is assumed that the stent can partially mechanically occlude the leak site favoring more rapid closure. However, Bjorkman and colleagues [133] reported 100% fistula closure rate in 15 patients after placing one short (2 to 3 cm) 10F stent with the stent tip distal to the leak site. The results of this study confirm the importance of eliminating the transpapillary pressure gradient. Most studies that monitor drain output or reassess the fistula by repeat cholangiography report rapid closure of the fistula in most cases with cessation of bile extravasation in 1 to 7 days. [138] The precise time when the fistula site is permanently closed however is difficult to determine from reported series.

The available data suggest that biliary fistulae are likely to heal regardless of the therapy used to decrease the pressure gradient in the direction of the duodenum. Randomized studies comparing sphincterotomy, internal stent, and NBT will be necessary to determine which of these therapies is the safest, most reliable, and most cost-effective management option. Biliary fistulae associated with bile duct strictures will require long-term stenting, preferably with large bore stents (10F and 11.5F stents).

Malignant Bile Duct Obstruction

A variety of palliative options can be offered to the patient with malignant obstructive jaundice including surgical, percutaneous, endoscopic, and medical therapy (chemotherapy and radiation therapy). Certainly the surgically fit patient with a resectable tumor after staging should be offered the option of surgical resection for cure. In the high-risk patient or the patient with an unresectable tumor, endoscopic placement of polyethylene stents has become a widely accepted method of management. [140] Soehendra and Rejinders-Frederix [141] first described endoscopic biliary stenting in 1980. Since then, many advances in stent technology have been made. Despite these developments, stent patency remains a major problem with 10F stents becoming occluded after 3 to 6 months. [142], [143] The problem with stent occlusion has been studied intensively but attempts at altering bile composition with choleretic agents, reducing bacterial load with antimicrobial agents, changing stent material, or influencing mucin production with aspirin have failed to prolong stent patency. [144], [145] and [146] Because deposition of sludge (leading to stent occlusion) may depend on flow rate through the stent, change in stent diameter may influence the process of stent clogging. Theoretically, a small increase in stent diameter may result in an appreciable increase in flow. The limiting factor for insertion of larger plastic biliary stents is the size of the instrumentation channel of the duodenoscope. With today's endoscopes, plastic stents up to 12F can be placed. The question of whether bigger is better still cannot be definitively answered for plastic stents. In some studies, stent patency was significantly longer for large diameter plastic stents than small diameter ones. [147] Others have found no prolongation of stent patency and more complications when larger stents are used. [148] The divergent results of these studies may be explained by study design, patient selection criteria, and sample size.

There is considerable debate as to whether patients with strictures involving the bifurcation require ductal decompression of both the right and left intrahepatic systems. [140] Advocates of a single stent argue that ductal decompression of one lobe improves symptoms of cholestasis and allows jaundice to resolve. [149] Proponents of decompressing both sides of the liver point to the 30% to 40% incidence of cholangitis, increased mortality, and death from sepsis when only one lobe is drained. [150] Our approach to hilar strictures is as follows: Once a guidewire is advanced into an intrahepatic duct, bile is aspirated to limit systemic seeding of any resident bacteria when contrast is injected. Only enough contrast should be injected to define the stricture anatomy. If a good stentable duct is identified, that lobe is drained. When draining only one lobe of the liver, it is imperative to limit contrast injection to the lobe to be drained and avoid manipulation of the other lobe. The other lobe should be stented if the patient develops cholangitis or symptoms of cholestasis persist. Two studies support this approach. [151], [152] and [153] De Palma and associates [152] randomized 157 consecutive patients with malignant hilar obstruction to undergo unilateral or bilateral hepatic duct drainage. In the intention-to-treat analysis, unilateral drainage was associated with significantly higher rates of successful drainage (defined as decreases in bilirubin to less than 75% of the pretreatment values within one month) and lower early complication rates (primarily because of lower rates of cholangitis). Thirty-day morality, late complications, and median survival were similar for the two groups. MRCP can help select the liver lobe to be drained thus avoiding contrast medium injection into the contralateral lobe. [154]

The success rate of plastic stent insertion is about 90%, and it is higher in patients with distal than proximal tumors. [155] If endoscopic stent placement fails, percutaneous drainage or a combined endoscopic radiologic procedure (rendezvous procedure) can be performed. If contrast was injected into the biliary tree, such therapy should be performed urgently to prevent cholangitis. In the surgically fit patient, particularly if duodenal obstruction is present, surgical bypass is a reasonable option. Relief of symptoms can be expected in nearly all patients following successful deployment of a plastic stent. Stenting not only resolves jaundice and pruritus, but also is associated with improvements in quality of life. [156], [157]

Early postprocedure complications, which have been reported in 10% to 20% of patients in most studies, are related to the sphincterotomy or to the insertion of the stent itself. The most frequent early complication is cholangitis, reported to occur in as many as 10% to 15% of patients, and is probably due to the introduction of bacteria during the procedure into the stagnant bile proximal to the stricture. [10], [158] The risk of cholangitis is higher if incomplete drainage is obtained.

The main late complication of biliary stenting is cholangitis as a result of stent occlusion. Stents placed for hilar obstruction appear to occlude faster than stents placed for more distal obstructing lesions. [155], [159] Patients with symptomatic stent occlusion will require stent change and possibly hospitalization for therapy of cholangitis. As a result, some authorities have advocated prophylactic stent changes with the hope of avoiding cholangitis. Sherman and colleagues [159] have demonstrated that nearly 50% of patients undergoing stenting with 10F or 11.5F plastic biliary stents die prior to stent occlusion. Thus, patients with a short life expectancy would be subjected to unnecessary procedures if prophylactic stent changes were performed. Using computer modeling, Tarnasky and associates [160] suggested that indicated stent exchanges are more cost-effective compared to prophylactic stent change at any interval. Prat and associates [161] reported that symptom-free survival was longer for patients undergoing planned stent exchange every 3 months but offered no cost advantage compared to patients undergoing stent exchange for symptomatic occlusion. Preliminary results of a randomized trial comparing scheduled stent change every 4 months to symptomatic stent change revealed no difference in the number of ERCPs per patient, number of stents per patient, mortality rate, need for metal stenting, frequency of surgery, mean stent survival, frequency of cholangitis, and time to death. [162]

One of the major advances in stent technology was the development of the metal expandable stent. Expandable metal stents may offer improved biliary drainage because of their large diameter and small surface area with prolonged patency rates. There are several types of expandable metal stents available, characterized by different insertion devices, methods of deployment, radial forces, and metal composition. To date, most experience has been gained with the Wallstent. This stent is easily inserted over a well-positioned guidewire and successfully deployed in more than 95% of cases. The Wallstent is mounted on a 7.5F delivery device and shortens and expands to 8 to 10 mm as it is deployed. Four prospective, randomized trials [163], [164], [165] and [166] (three endoscopic and one percutaneous) have shown that the metal expandable biliary stent occludes less frequently and less rapidly than the conventional 10F and 11.5F plastic stents. This translated into a reduction in hospitalization requirements (for cholangitis and stent change) and an overall cost savings for the metal stents. Since metal stents are more costly initially, and since approximately half of the patients in most plastic stent series will need a second stent, identification of patients who are likely to outlive their first plastic stent (and

warrant a metal stent) is a major challenge for the managing physician. In a study from Amsterdam, [163] the stent patency curves of Wallstents and plastic stents ran parallel during the first 3 months after stent insertion. After that time the curves diverged in favor of the Wallstent. Therefore, based on data from this study, the authors recommended that only patients with a life expectancy of more than 3 months are potential candidates for use of an expandable metal stent. An additional indication for use of metal stents can be found in the small group of patients who suffer rapid and repeated obstruction of plastic stents. These patients have not been well studied and can, at present, not be identified at the initial stenting session.

When palliation is the goal of therapy for patients with malignant bile duct obstruction, how does endoscopic decompression compare to percutaneous and surgical drainage procedures? In a randomized study comparing percutaneous drainage to endoscopic drainage, [167] endoscopic stenting was associated with more frequent successful drainage (81% vs. 61%; $P < 0.05$), a lower complication rate (19% vs. 67%; $P < 0.05$), and a lower 30-day mortality (15% vs. 33%; $P < 0.05$). Median survival was similar for the two groups (23 vs. 16 weeks). Three prospective, randomized trials [168], [169] and [170] have compared endoscopic to surgical drainage for malignant distal biliary obstruction. Endoscopic stenting and surgery were equally effective palliative treatments with endoscopic treatment having a lower early complication rate and mortality, but a higher risk of late complications, such as stent blockage and gastric outlet obstruction. None of these studies demonstrated a difference in survival rates between treatment groups.

Tissue Sampling at ERCP ERCP frequently provides the first opportunity to obtain a histological or cytologic specimen from an unexplained biliary or pancreatic stricture. A variety of tissue sampling techniques are available to the endoscopist at the time of ERCP. These include bile and pancreatic juice cytology, brush cytology, intraductal forceps biopsy, intraductal fine-needle aspiration, stent cytology, and juice and tissue evaluation for aneuploidy, tumor markers (e.g., CEA, CA 19-9), p53 immunoreactivity, and K *-ras* oncogene mutations. Aspiration of bile or pancreatic juice is the easiest method of obtaining tissue when evaluating biliary and pancreatic strictures for malignancy. Unfortunately, the results have been largely disappointing, with cancer sensitivities in the 6% to 32% range [171], [172], [173] and [174] and frequent findings of acellular specimens. The desmoplastic nature of certain tumors or failure of the neoplasm to invade the ductal epithelium are likely responsible for these results. [174] It has been suggested that endoscopic manipulation of the stricture (e.g., dilation) [175], [176] prior to bile collection may increase tumor exfoliation, making more malignant cells available for diagnosis. However, this has not been confirmed by others. [177] Brush cytology is the most commonly applied method of tissue sampling and most extensively studied. While the technical success rate is high (90% to 95%), most studies demonstrate cancer detection rates in the 20% to 60% range. [171], [178], [179], [180], [181], [182], [183] and [184] Sawada and colleagues [185] have shown that brushing the pancreatic duct may increase the diagnostic yield of brush cytology (compared to brushing the bile duct) in pancreatic cancer. However, pancreatic cancers often disrupt the duct and prevent passage of the brush through the tumor in more than 25% of patients. In an attempt to improve on the sensitivity of brush cytology, other methods have been used. Howell and colleagues [186] originated use of the ERCP endoscopic needle aspiration (ENA) technique and reported a cancer detection sensitivity from biliary samplings of 62% in patients with biliary strictures (including 53% in pancreatic cancer and 80% in cholangiocarcinoma). These impressive results for ENA were not found in three subsequent [177], [183], [187] reports where the sensitivity was 26%, 30%, and 27%, respectively. Endobiliary forceps biopsy allows examination of tissue specimens below the bile duct epithelium. The results of six prospective studies have been encouraging, [171], [177], [179], [180], [183], [188] with a cancer detection rate among 415 patients of 58%. Stents placed for palliation can be removed and sent for cytologic evaluation of adherent cells, as exfoliated malignant cells may become entrapped in the Biofilm and sludge of an occluded endoprosthesis. A few small studies have evaluated this approach, with a wide range of sensitivity results. Foutch and associates [174] reported a sensitivity of 36% (4 of 11 patients), while Leung and associates [189] reported a sensitivity of 79% (11 of 14 patients). Devereaux and colleagues [190] reported a cancer detection rate of only 11% among 101 patients with biliary strictures (57 were confirmed to be malignant). Unfortunately, this method is impractical because the diagnosis of cancer is delayed until the stent is removed. Although it would be preferable to have one technique that would have a cancer detection rate similar to that seen with biopsy of upper gastrointestinal and colonic neoplasms, this goal has not been reached in the pancreaticobiliary tree. Investigators have therefore evaluated the added sensitivity of combining a number of tissue sampling techniques. Jailwala and colleagues [183] reported their results of the cumulative sensitivity of triple tissue sampling at one ERCP session with brush cytology, fine-needle aspiration, and forceps biopsy in 104 patients with malignant bile duct obstruction. Tissue sampling sensitivity varied according to the type of cancer; the highest yield was seen in patients with ampullary cancer. The combination of techniques was superior to individual methods, as the addition of a second or third technique increased cancer sensitivity rates in most instances. It is clear that the cancer detection sensitivity of these standard techniques individually is suboptimal. Methods to improve this sensitivity are therefore being evaluated. Preliminary studies suggest that the yield may be increased by evaluating aspirated fluid and tissue for aneuploidy [191] and tumor markers such as CEA and CA 19-9. Investigations have suggested that the evaluation of tissue or fluid for K *-ras* mutations is more accurate than cytology in the diagnosis of pancreatic cancer. [192], [193], [194] and [195] However, some authors [194] have identified K *-ras* mutations in patients with chronic pancreatitis reducing the specificity of this test. Further study is warranted to determine the role of these new techniques in the assessment of pancreatic and biliary strictures.

Sump Syndrome

The sump syndrome is an infrequent complication of a side-to-side choledochoduodenostomy. Some degree of stenosis of the surgical anastomosis is usually present. Cholangitis, pain, and pancreatitis may occur as food, stones, or other debris accumulate in the common bile duct in the bypassed segment. The reported median time interval between surgery and the appearance of symptoms was 5 years and 6 years between surgery and the diagnosis of sump syndrome. ES with removal of the debris has been shown to be an effective treatment. [196], [197] and [198] Although it may be possible to extract debris and stones through the choledochoduodenostomy, obviating the need for sphincterotomy, this approach puts the patient at risk of recurrent symptoms.

Choledochal Cysts and Anomalous Pancreatobiliary Union

Choledochal cysts are uncommon anomalies of the biliary tree manifested by cystic dilation of the intrahepatic or extrahepatic ducts. [199], [200] These cysts are most often classified by the scheme proposed by Todani and associates. [201] Type I cysts, which involve only the extrahepatic biliary tree, are the most common form, accounting for 80% to 90% of all choledochal cysts. [202] In this form of the anomaly, the cystic duct generally enters the choledochal cyst and the right and left hepatic ducts and the intrahepatic ducts are normal in size. Type II cysts are extrapancreatic bile duct diverticula and make up 2% of reported cases. [203] Type III cysts, accounting for 1.4% to 5% of cases, are choledochoceles and most often involve only the intraduodenal part of the common bile duct but occasionally the intrapancreatic portion. [204] Type IV cysts are subdivided into type IV A, multiple intrahepatic and extrahepatic cysts, and type IV B, multiple extrahepatic cysts. Type IV A cysts account for approximately 19% of reported cases, whereas type IV B cysts are much less common. [205] Finally, the type V cyst, or Caroli disease, consists either of single or multiple intrahepatic cysts. This form of cystic disease within the liver communicates with the biliary system as opposed to fibrocystic disease in which cysts filled with bile do not communicate with the biliary system. [206]

An anomalous pancreaticobiliary union is considered to be present when the common channel is greater than 15 mm in length. In this situation, the pancreatic duct and bile duct junction is outside the duodenal wall and proximal to the sphincter of Oddi promoting reflux of pancreatic juice into the biliary tree. Pancreatic juice reflux has been postulated to be involved in the pathogenesis of carcinoma, which occurs in 2.5% to 17% of patients with choledochal cysts. [207], [208], [209], [210], [211] and [212]

Surgery is the recommended treatment for most patients with choledochal cysts. [212], [213], [214] and [215] Cholangiography is the gold standard for diagnosing choledochal cysts. Although ERCP and PTC are invasive, they can thoroughly assess the cyst anatomy, site of biliary origin, extent of intrahepatic and extrahepatic disease, associated biliary tract anomalies, and complications (e.g., bile duct strictures, stones); and they shed light on possible therapeutic intervention, either definitive or temporizing pending surgery. ERCP is often the preferred modality because it provides detailed evaluation of the pancreatic duct and the pancreaticobiliary union, and is very useful in the diagnosis of type III choledochal cysts (choledochocele).

ERCP has become the procedure of choice to evaluate and treat most patients with type III choledochal cysts. [216] Patients with choledochoceles will commonly present with biliary symptoms (biliary colic, cholestatic jaundice, jaundice) or unexplained pancreatitis prompting ERCP evaluation. The endoscopic features of a choledochocele include:

- the intramural segment of common bile duct protrudes into the duodenum in continuity with an enlarged papilla
- the papilla is soft and smooth
- ballooning of the papilla is noted with contrast injection
- on contrast injection, a cyst-filled structure is apparent on fluoroscopy and in continuity with the common bile duct
- no impacted stone is present.

Several small series have reported the use of endoscopic cyst unroofing and sphincterotomy for both pancreatic and biliary indications. [216], [217], [218], [219], [220] and [221] Ladas and associates [216] identified 15 symptomatic choledochocele patients among 1019 (1.5%) referred for ERCP. Twelve patients were treated by endoscopic

therapy. During long-term follow-up (mean, 26 months; range, 4 to 56 months), 10 of 12 patients were asymptomatic with normal liver tests. One patient had a mild episode of cholangitis and one developed a carcinoma in the choledochocele. This unusually high frequency of choledochoceles may represent overdiagnosis, since several of these patients appeared to have only bile duct and ampulla of Vater dilation associated with ductal stones (not true choledochoceles). Although the risk of cancer in these patients is uncertain, it appears appropriate to recommend long-term follow-up in patients treated by endoscopic therapy alone. How this follow-up should be pursued remains to be clarified. Elton and colleagues [220] described a variant of a choledochocele which they called the dilated common channel syndrome. These patients have enlarged common pancreatobiliary channels which were believed to have developed because of papillary stenosis. Among 77 patients treated with unroofing and sphincterotomy, 77% had complete and long-lasting resolution of symptoms.

The management of anomalous pancreaticobiliary union in the absence of a choledochal cyst is unclear. Because of the high risk of gallbladder cancer, prophylactic cholecystectomy has been recommended by some. [211] In one series [222] of 15 patients with an anomalous pancreaticobiliary union (7 had choledochal cysts) and recurrent pancreatitis or abdominal pain treated by ES, 13 had resolution or reduction in the frequency of pancreatitis and pain. Ng and colleagues [223] similarly reported pain and pancreatitis resolution in five of six patients with a long common channel following endoscopic therapy. It is not yet known whether patients with anomalous junctions without choledochal cysts treated by sphincterotomy need surveillance for cancer.

PANCREATIC DRAINAGE PROCEDURES

Chronic Pancreatitis

Pancreatic duct pressure is generally increased in patients with chronic pancreatitis regardless of the etiology and whether or not the main pancreatic duct is dilated. [224] The aim of endoscopic therapy (and decompressive surgical therapy) for patients with chronic pancreatitis presenting with pain or clinical episodes of acute pancreatitis is to alleviate the obstruction to exocrine juice outflow. Certain pathological alterations of the pancreatic duct, bile duct, or sphincter lend themselves to endoscopic therapy. The techniques (e.g., sphincterotomy, dilation, stenting) and instruments (e.g., sphincterotome, dilating balloon, pancreatic stent) used to treat biliary tract disease have been adapted for use in the pancreatic duct.

Data in this area are often difficult to interpret because of the heterogeneous populations with one or more pathological processes being treated (e.g., pancreatic duct stones, strictures, pseudocysts) and because of the multiple therapies being performed in a given patient (e.g., stricture dilation, stone extraction, bile duct, or pancreatic duct ES). Controlled studies have not been reported to date.

Pancreatic Strictures Benign strictures of the main pancreatic duct may be a complication of previous embedded stone or consequence of acute inflammatory changes around the main pancreatic duct. [225] In Cremer and colleagues' [225] large referral population, only 10% of the patients presented with a stricture without associated calcified pancreatic stones. Pancreatic duct strictures can be treated by stent therapy. If stents larger than 7F are to be used, patients often require both a pancreatic and bile duct sphincterotomy followed by stricture dilation. For optimal results, the therapy must address both the pancreatic duct stricture and duct stones. The best candidates for stenting are those with a distal stricture (in the pancreatic head) and upstream dilation.
Pancreatic Stent Placement Technique The technique for placing a stent in the pancreatic duct is similar to that used for inserting a biliary stent. A guidewire must be maneuvered upstream to the narrowing. Hydrophilic flexible tip wires are especially helpful. Most pancreatic stents are just standard biliary stents with extra side holes at approximately 1-cm intervals to permit better side branch juice flow. In general, the size of stent should not exceed the size of the normal downstream duct. Therefore, 4F to 7F stents are commonly used in small ducts, whereas 10F to 11.5F stents can be used in advanced chronic pancreatitis and grossly dilated ducts. A pancreatic sphincterotomy (major or minor papilla) is often performed prior to (or after) placing a pancreatic stent. [226], [227], [228], [229] and [230] This is done with a standard pull-type sphincterotome or by using a needle-knife to incise the sphincter over a previously placed stent. Some authorities favor performing a biliary sphincterotomy prior to the pancreatic sphincterotomy because of the high incidence of bile duct obstruction and cholangitis, reported by one group, if this is not done. [231] Such complications were not found by others and have been infrequent in our experience. [226], [227] and [228], [232] Performing a biliary sphincterotomy first, however, can expose the pancreaticobiliary septum and allow the length of the cut to be gauged more accurately.
Efficacy of Stenting The results of pancreatic duct stent placement (usually with ancillary procedures) are detailed in Table 141-5. [225], [233], [234], [235], [236], [237], [238], [239], [240] and [241] Successful stent placement was achieved in 82% to 100% of patients. Sixty-six percent of patients were reported to benefit from therapy during an 8- to 39-month follow-up period. Quantification of the degree of improvement is often poorly defined. Partial or complete symptom improvement after stenting suggests that intraductal hypertension was an etiologic factor. Continued symptom relief after stent removal indicates adequate dilation of the narrowing. Differentiation of these two types of improvement is not clarified in some reports.

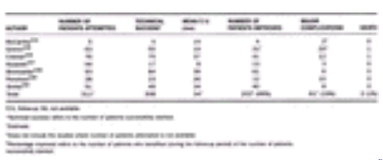

TABLE 141-5 Selected Series Reporting the Results of Pancreatic Duct Stenting for Dominant Strictures

Unfortunately, because the stricture persists in the majority, compliance with long-term use of such plastic stents (i.e., multiple stent changes) would be difficult. As a result, Cremer and colleagues [242] evaluated the expandable metal stent (18F, 23 mm in length) in 29 patients. After 6 months, mucosal hyperplasia resulted in stent occlusion in most patients.

Pancreatic Ductal Stones

It has been postulated that increased intraductal pressure proximal (upstream) to an obstructed focus within the pancreatic duct, as with pancreatic duct stones, is one of the potential mechanisms responsible for attacks of acute pancreatitis or exacerbations of chronic abdominal pain in patients with chronic pancreatitis. [236], [243] Reports indicating that endoscopic (with or without ESWL) or surgical removal of pancreatic calculi results in improvement in symptoms support this notion. [231], [243], [244], [245], [246], [247], [248], [249], [250], [251], [252], [253], [254] and [255] In one series, [249] 32 patients with pancreatic duct stones underwent attempted endoscopic removal. Of these patients, 72% had complete or partial stone removal and 68% improved after endoscopic therapy. Symptomatic improvement was most evident in the group of patients with chronic relapsing pancreatitis (versus those presenting with chronic continuous pain alone; 83% vs. 46%). Factors favoring complete stone removal included:

- three or fewer stones
- stones confined to the head or body of the pancreas
- absence of a downstream stricture
- stone diameter of <10 mm
- absence of impacted stones.

After successful stone removal, 25% of patients had regression of the ductal changes of chronic pancreatitis and 42% had a decrease in the main pancreatic duct diameter. The only complication from therapy was mild pancreatitis in 8%. These data suggest that removal of pancreatic duct stones may result in symptomatic improvement. A longer follow-up will be necessary to determine the stone recurrence rate and whether endoscopic success results in long-standing clinical improvement.

Smits and associates [251] treated 53 patients with pancreatic duct stones primarily by endoscopic methods alone (8 had ESWL). Stone removal was successful in 42 patients (79%), complete in 39, and partial in 3. There was initial relief of symptoms in 38 (90%). Three of 11 patients (27%) with failed stone removal had improvement in symptoms suggesting, as do results of Sherman and colleagues, [249] that some of the clinical response may be related to other therapies performed at the time of attempted stone removal (e.g., pancreatic sphincterotomy). During a median follow-up period of 33 months, 13 patients had recurrent symptoms due to stone recurrence. The stones were successfully removed in 10 (77%). No factor evaluated (etiology of pancreatitis, presentation with pain or pancreatitis, presence of single or multiple stones, location of stones, presence or absence of a stricture) was shown to predict successful stone treatment (defined as complete or partial removal of stones resulting in relief of symptoms).

Cremer and colleagues [252] reported complete stone clearance was achieved in only 18 (45%) of 40 patients with pancreatic duct stones who were treated by endoscopic methods alone, but immediate resolution of pain occurred in 77% of these patients. During a 3-year follow-up, 63% remained symptom-free. Clinical steatorrhea improved in 11 of 15 patients.

ESWL can be used to facilitate fragmentation and stone removal when endoscopic therapy alone fails. [231], [246], [250], [253], [254], [255], [256] and [257] Thus, this procedure is complementary to endoscopic techniques and improves the success of nonsurgical ductal decompression. Patients with obstructing prepapillary concrements and upstream ductal dilation appear to be the best candidates for ESWL. In the largest reported series, [231] 123 patients with main pancreatic duct stones and proximal dilation were treated with the electromagnetic lithotripter, usually before pancreatic duct sphincterotomy. Stones were successfully fragmented in 99% resulting in a decrease in duct dilation in 90%. The main pancreatic duct was completely cleared of all stones in 59%. Eighty-five percent of patients noted pain improvement during a mean follow-up of 14 months. However, 41% of patients had a clinical relapse due to stone migration into the main pancreatic duct, progressive stricture, or stent occlusion. Similar results were reported by Sauerbruch and colleagues [246] in 24 patients using a kidney lithotripter. Complications in series using ESWL were primarily related to the endoscopic procedure. Pancreatoscopy with laser and electrohydraulic lithotripsy of pancreatic duct stones has been successfully performed in small numbers of patients. [258]

Overall, the endoscopist is encouraged to remove pancreatic duct stones in symptomatic patients when they are located in the main duct (in the head or body) and are thus readily accessible. The available data suggest that the clinical outcome after successful endoscopic removal is similar to surgical outcome, with lower morbidity and mortality. [259] However, to date, no comparative trials have been done.

Pancreatic Pseudocysts and Fistulae Pancreatic pseudocysts are defined as encapsulated collections (without an epithelial lining) of pancreatic juice, either pure or containing necrotic debris or blood, which are situated either outside or within the limits of the pancreas from which they arise. [260] A pancreatic pseudocyst arises as a consequence of acute pancreatitis, chronic pancreatitis, or pancreatic trauma. [261] The optimal candidates with pseudocysts for endoscopic drainage have a single mature cyst without pancreatic necrosis, residual adjacent inflammation, or portal hypertension. More complex patients are generally best managed by a multidisciplinary approach with input from surgery, medicine, and interventional radiology. Two endoscopic approaches can be applied depending on whether the cyst communicates with the pancreatic duct. [236], [260], [262], [263], [264], [265], [266], [267], [268] and [269] Cysts communicating with the ductal system can be drained by a transpapillary approach. [262], [263], [265], [266], [267] and [268] The proximal tip of the prosthesis generally has been placed in the cystic cavity. [260] A pancreatic duct sphincterotomy may be required. Noncommunicating pseudocysts can be treated by direct cystoenterostomy via the stomach (endoscopic cystogastrostomy) or duodenum (endoscopic cystoduodenostomy) (Fig. 141-9). The aim of therapy is to create a communication between the cystic cavity and the gastric or duodenal lumen. Two prerequisites should be fulfilled before attempting this treatment: bulging due to the cyst should be obvious during upper endoscopy and the distance between the cyst and the lumen should not exceed 1 cm. [260] This distance can usually be assessed by CT scan, ultrasound, or endosonography, but when the compression is visible, usually the cyst to lumen distance is less than 1 cm. Also, the cyst wall should be mature. A double or triple lumen, beveled tip needle-knife is used to burrow a hole (usually using blended current) into the cyst cavity. A cystoenterotome is commercially available. [270] Many authorities advocate contrast injection via needle puncture of the cyst wall to identify a safe entry site before diathermic puncture. [271] The complication rate of this procedure may also be reduced by using the Seldinger technique without electrocautery. [272] A guidewire is advanced into the cyst and looped 360 degrees to secure positioning. Puncture should be performed perpendicularly to the cyst wall, and thus a duodenoscope is preferred. The newly created tract is then balloon dilated to 8 to 10 mm. Vigorous flow of pseudocyst fluid into the gut lumen generally occurs and this must be aspirated to maintain the endoscopic view. Two or more double-pigtail stents are then placed, bridging the cyst and the intestinal lumen. When significant debris or necrotic tissue is present, a nasocystic drain should be considered to allow for lavage of the cyst cavity. It is appropriate to maintain the patient NPO and on IV broad-spectrum antibiotics for 1 to 3 days if the cyst is >6 cm or contains debris. Diabetics, patients with debris in the cyst, and immunosuppressed patients may need a longer NPO interval as oral intake permits food and a greater concentration of bacteria to enter the residual cyst. The pseudocyst size is followed with ultrasound or CT scan at 4- to 6-week intervals. After resolution (usually in 1 to 2 months), the stents are endoscopically removed. If not done initially, pancreatography is performed to identify and allow treatment of ductal disease, such as stricture or stones, in an attempt to decrease pseudocyst recurrence. Pseudocysts recur in 10% to 20% of endoscopically managed cases, especially in patients with duct cut-off on pancreatogram. Table 141-6 shows the results of endoscopic management from large centers.

TABLE 141-6 Results of Endoscopic Management of Pseudocysts

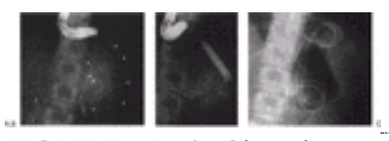

FIGURE 141-9. Chronic pancreatitis with a 7-cm nonresolving pseudocyst. **A:** Puncture of the pseudocyst via transgastric approach. Contrast injection reveals filling of a 7-cm pseudocyst (*arrowheads*) containing filling defects (*arrows*). Note the pancreatic calcifications (*open arrows*) in the region of the pancreas. **B:** The tract between the stomach and pseudocyst is dilated with a 10-mm hydrostatic dilating balloon. **C:** Two 10F, double-pigtail stents were placed, bridging the stomach and pseudocyst.

These excellent results certainly support the use of endoscopic therapy in appropriate candidates. When compared to other endoscopic techniques, this procedure has a relatively high bleeding and perforation rate. Bleeding complications are decreased by use of a hydrostatic balloon (not sphincterotome) to enlarge the tract orifice or by initial puncture with a needle catheter instead of the needle-knife. Nevertheless, the overall complication rate probably compares favorably to surgical series. Coordination with the surgeon is necessary when performing this procedure.

PANCREAS DIVISUM

Pancreas divisum, the most common congenital variant of pancreatic ductal anatomy, occurs when the ductal systems of the dorsal and ventral pancreatic ducts fail to fuse during the second month of gestation. With nonunion of the ducts, the major portion of the pancreatic exocrine juice drains into the duodenum via the dorsal duct and minor papilla. It has been proposed that a relative obstruction to pancreatic exocrine juice flow through the minor papilla could result in pancreatic pain or acute pancreatitis in a subpopulation of patients with pancreas divisum. [273], [274] While several epidemiologic studies dispute the relation of pancreas divisum and pancreatitis, two lines of evidence favor this association:

1. Histological studies and pancreatograms have demonstrated signs of chronic pancreatitis isolated to the dorsal pancreas.
2. Several reports have indicated that symptoms may be relieved by facilitating dorsal duct decompression endoscopically or surgically.

Endoscopic attempts to decompress the dorsal duct in patients with symptomatic pancreas divisum have been performed primarily by dilation, stent insertion, or minor papilla sphincterotomy. [234], [275], [276], [277] and [278] Lans and colleagues [276] reported their results of a randomized controlled trial of long-term (12 months) stenting of the minor papilla in patients with recurrent pancreatitis ($n = 19$). Follow-up continued for at least 12 months after stent removal. Stented patients had fewer hospitalizations and episodes of pancreatitis ($P < 0.05$) and were more frequently judged to be improved (90% vs. 11% for controls, $P < 0.05$). While symptomatic improvement following this therapy has been encouraging, multiple stent changes are generally required and the risks of stent-related complications are considerable. Ertan [277] reported that 21 of 25 patients (84%) with pancreas divisum developed stent-induced ductal changes after stenting periods of 6 to 9 months.

A “more permanent” enlargement of the minor papilla orifice is possible from sphincterotomy. Lehman and colleagues [278] attempted to evaluate the efficacy of minor papilla ES for patients with pancreas divisum ($n = 52$) presenting with disabling pancreatic-type pain ($n = 24$), idiopathic acute recurrent pancreatitis ($n = 17$), or chronic pancreatitis ($n = 11$). A short 4F to 7F stent was placed in the minor papilla and then a 3- to 6-mm sphincterotomy was performed over the stent using the

stent as a guide for cutting and a bridge to prevent edema-induced closure of the cut. The stent was then removed in approximately 2 weeks. The mean duration of symptoms was 5.1 years, and follow-up averaged 1.7 years with all patients being observed at least 6 months after therapy. While 76.5% of the acute recurrent pancreatitis group improved following therapy, only 26% of the chronic pain group ($P = 0.002$) and 27% of the chronic pancreatitis group ($P = 0.01$) benefited. Similarly, as compared to the chronic pain and chronic pancreatitis groups, the acute recurrent pancreatitis group had a significant reduction in the mean pain score and number of hospital days per month required for severe pain or pancreatitis. These discordant results in responsiveness to therapy for the acute recurrent pancreatitis group versus the chronic pancreatitis and chronic pain group were noted in several surgical series evaluating dorsal duct decompression [279], [280] and other endoscopic series. [281], [282] Pancreatitis complicating therapy occurred in 13% but, in general was mild and managed conservatively. Stent-induced dorsal duct changes occurred in 50%. One elderly patient died from complications of a pancreatic abscess that developed after a failed attempt at minor papilla stent placement. Results of these studies suggest that patients with pancreas divisum with acute recurrent pancreatitis are good candidates for endoscopic therapy while patients with chronic pancreatitis or chronic pain alone do not do as well.

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CHAPTER 142

Cyrus E. Rubin and Mary P. Bronner

ENDOSCOPIC MUCOSAL BIOPSY—A MEMORIAL TO RODGER C. HAGGITT, M.D.

Rodger Haggitt passed away on June 28, 2000. This revised chapter is dedicated to him and presents his approach to gastrointestinal biopsy pathology. The sources of this chapter are Dr. Haggitt's prior published versions of this chapter and his other previously published and unpublished work. We also relied on the memories of his closest disciples, collaborators, residents, and other professional colleagues.

Dr. Haggitt early recognized the great clinical importance of endoscopic biopsy. He therefore taught himself how to interpret these small tissue samples and correlate their histological picture with the clinical findings. He developed his skills "on the job," learning much from his patients, fellow pathologists, and consulting gastroenterologists. To an extraordinary degree his approach combined common sense, diagnostic skill, and critical clinical judgment. He was blessed with a superb analytical mind. He was also an unusually able teacher of both pathologists and clinicians and it is our hope that this chapter will keep those teachings and his memory alive.

This chapter describes the clinicohistological approach to gastrointestinal (GI) endoscopic biopsy diagnosis. The endoscope is an excellent tool for visualizing gross mucosal lesions some of which, such as a varix, are obvious diagnostically, but most of which, such as erosions, ulcers, or masses, require biopsy for a specific diagnosis. The normal endoscopic surface appearance of the mucosa does not reveal whether its full thickness is normal or abnormal histologically. When an endoscopic diagnosis of "esophagitis, gastritis, or colitis" is based on subjective endoscopic impressions on which there is poor interobserver consensus, such as "reddening, edema, or granularity," the histology commonly turns out to be completely normal. On the other hand, an endoscopically normal appearing mucosa may

on occasion reveal unsuspected lesions such as dysplasia or infectious processes histologically. For example, every patient with normal appearing colonic mucosa and undiagnosed diarrhea merits at least two rectal and perhaps some proximal biopsies to rule out lesions such as collagenous colitis, lymphocytic colitis, quiescent ulcerative colitis, or Crohn's disease, all of which may not produce endoscopic changes. Because the endoscopic surface mucosal appearance may be misleading, the authors believe, as did Dr. Haggitt, that the mucosa should always be sampled according to a standard protocol during every diagnostic endoscopy.

To interpret the histopathology of endoscopic biopsies accurately, the GI pathologist must know and recognize diagnostic criteria for a broad spectrum of medical and surgical conditions and correlate them with the clinical findings to formulate the diagnosis. Knowing the clinical background of the patient who has been biopsied is essential for an accurate diagnosis because similar histological pictures may be seen in different diseases. To facilitate biopsy interpretation by the pathologist, a detailed endoscopic report should accompany every biopsy request and should include pertinent clinical history, physical findings, relevant diagnostic studies, and an endoscopic photograph of any important findings.

Four factors determine the usefulness of endoscopic biopsy:

1. The skill and diligence of the endoscopist who takes the biopsy.
2. The training and motivation of the technicians who process the biopsies.
3. The specialized experience, training, and skill of the GI pathologist who interprets the biopsies.
4. The quality and quantity of interaction between gastroenterologists and pathologists.

TECHNICAL CONSIDERATIONS

Diagnostic accuracy is greatly enhanced if biopsy samples are large enough, sufficient in number, correctly oriented, separately labeled, optimally fixed, step-serial sectioned, and appropriately stained. Dr. Haggitt said that approximately half the consultations sent to him from pathologists and gastroenterologists were prompted by defects in tissue processing and biopsy technique. In this regard, the suggestions given in the following paragraphs may be helpful.

A 3.4-mm diameter "jumbo" biopsy forceps passed through a large-channel endoscope yields specimens that are twice as large as those taken with standard-size forceps through a smaller endoscope. Smaller diameter endoscopes are now available with larger channels permitting passage of the larger forceps. Large biopsies can be taken safely using the "turn and suck" technique. [1] Larger biopsies are the best for diagnostic purposes because they are easier to orient, contain proportionately less crush artifact, permit more precise evaluation of architecture, and are more likely to detect small, infrequent lesions on serial sectioning. Proper use of a large biopsy forceps is not associated with a higher risk of complications than use of smaller versions. When removing a small lesion by "hot" biopsy or with an electrocautery snare, excessive cauterization can easily distort the tissue beyond recognition or rarely cause perforation. Saline injection submucosally in a sessile lesion can make electrocautery snare removal less dangerous without impairing histological quality.

Endoscopists tend to err on the side of taking too few biopsies for optimal diagnostic accuracy. The specific number of samples that should be obtained depends on whether the disease being evaluated is patchy or diffuse in distribution, and whether it is endoscopically visible and excisable, like a polyp, or invisible, like many dysplasias.

Correct orientation of biopsies is essential for obtaining sections perpendicular to the mucosal surface. Such sections are needed for precise evaluation of villous and crypt architecture, a sensitive but nonspecific indicator of abnormality in a wide variety of mucosal diseases. Ideally, the endoscopy assistant should orient the fresh biopsy onto a substrate which is then placed into fixative. To do this, the specimen is first teased out of the biopsy forceps and placed mucosal surface down on a gloved fingertip; it is then carefully unfolded with a blunt-ended probe to uncover its cut side, which is then gently pressed onto the substrate; the fingertip is then rolled off of the lumenal surface using the side of the probe to keep the specimen attached to the substrate. The mounted biopsy is then dropped into the fixative. Videotapes demonstrating these techniques, produced by Dr. Wilfred Weinstein and Boston Scientific (Natick, MA), may be obtained at no cost by calling toll-free 1-800-225-3226. A less satisfactory alternative is to drop the unmounted biopsy into the fixative without orientation and have a trained technician orient the fixed biopsy in the histology laboratory. If pseudomembranous colitis or amebiasis is suspected, the biopsy should be dropped directly into fixative with minimal manipulation to preserve the adherent diagnostic membranes or amebae.

The cut surface of larger pieces of tissue removed with electrocautery snares can be recognized for orientation by the presence of a fulguration artifact. After preliminary fixation, larger polyps should be divided perpendicular to the cut surface through their long axis so that the center of the polyp can undergo adequate further fixation.

Finally, the locations of biopsy sites should be clearly indicated by placement of the samples in separately labeled bottles of fixative. This simple step not only provides accurate clinicopathological correlation but also guides the endoscopist in rebiopsy of suspicious areas, or the surgeon if resection is indicated.

Despite its wide use, formalin alone is not an ideal fixative. Because of the delicate nature of formalin fixation, the tissue is not protected from the disruptive effects of tissue processing. In addition, formalin provides poor definition of nuclear detail and uneven shrinkage. As formalin requires a minimum of 24 hours for adequate fixation, [2] most tissue is not fixed long enough because of the pressure for rapid turnaround time. Inadequate formalin fixation is a major contributing factor to the generally low quality of histological preparations available in the United States. The advent of microwave rapid processing may solve this problem.

To address these concerns, some pathologists use Bouin or B-5 solution or other formalin-based fixatives containing metallic cations such as mercury, copper, and zinc, or acids such as picric and acetic. These fixatives penetrate tissue more rapidly and precipitate and denature tissue proteins; this hardens the tissue and enables it to better withstand the harsh treatment of embedding and sectioning. The better preservation of nuclear detail provided by these fixatives facilitates the diagnosis of dysplasia and cancer. Decreased shrinkage minimizes artifactual tissue distortion. We use a modified Bouin solution called Hollande solution, which preserves red cell membranes and the eosinophil granules. Specimens should be fixed in Hollande solution for a minimum of 2 hours, but not for more than 3 days because they will become brittle. After sufficient fixation these biopsies can be washed with water and stored in alcohol prior to processing at a later, more convenient time.

We recommend partial serial sectioning of all GI biopsies. From 3 to 5 different biopsy specimens can be oriented on edge together in the same block and at least 10 to 20 sections can be included in each of two ribbons cut from the central, best-oriented portion of the block. Three slides are usually made. Sectioning perpendicular to the mucosal surface provides well-oriented glands, crypts, and villi, whose architecture and surface nuclear maturation are easier to assess, facilitating the diagnosis of dysplasia. Step-serial sectioning helps detect focal lesions like granulomas and dysplasia. Also, biopsies not optimally oriented on the initial sections may be interpreted on deeper cuts. Personal experience has shown that this approach is feasible as a routine procedure in both university and community hospitals if endoscopists and histology technicians are trained and sufficiently motivated.

We routinely stain our biopsies with hematoxylin and eosin (H & E) as well as a HEABS stain (H & E, Alcian blue at pH 2.5, and saffron). The Alcian blue stains acid mucin in goblet cells of intestinal metaplasia of the stomach and Barrett specialized metaplasia. The bright Alcian blue stain marks the occasional malignant signet cell containing acid mucin that may focally invade the lamina propria or deeper layers of the submucosa. Saffron helps identify the collagen deposition characteristic of diseases such as collagenous colitis, chronic ischemia, solitary rectal ulcer syndrome, or collagenous sprue. It may also help differentiate collagen from edema in the lamina propria. We substitute a Genta stain [3] for the HEABS stain in gastric biopsies. The Genta stain is an H & E combined with a silver and Alcian blue stain at pH 2.5. When the Genta stain is performed properly, it helps the pathologist detect *Helicobacter pylori* (HP) with minimal effort and is morphologically diagnostic because of recognizable black dots at each end of its orange body. The Genta stain is difficult to perform well; the Alcian yellow stain is a reasonable alternative. [4] H & E alone may detect the organisms, especially when the critical combination of fixation and hematoxylin is used, but special stains are most helpful when organism load is low. Immunolocalization of various antigens is useful in classification of tumors, hormone secretion, and lymphoid surface antigens. DNA-based assays can now be applied even after use of special fixatives if heavy metal ions are removed by Lugol iodine. [5]

Confusing artifacts affecting mucosal histological study may be induced by rough handling of biopsy specimens or by improper tissue processing. Double-cupped endoscopic biopsy forceps remove specimens by avulsion causing some intramucosal hemorrhagic and crush artifact, especially at the edge of the biopsy where normal cells may be squeezed together mimicking inflammation, fibrosis, dysplasia, or even carcinoma.

Most errors in biopsy diagnosis are related to overinterpretation of normal variations or to unappreciated artifacts.

DYSPLASIA

Esophageal squamous carcinoma, esophageal adenocarcinoma, gastric and cardia adenocarcinoma, and colonic adenocarcinoma occur more frequently in patients with specific diseases predisposing to cancer or in environments where such diseases are more common (e.g., Barrett esophagus predisposes to esophageal adenocarcinoma). Therefore, endoscopic screening for dysplasia or early carcinoma is warranted in such patients.

Glandular Dysplasia

Dysplasia is defined as an unequivocal neoplastic alteration of the epithelium that remains confined within the basement membrane of the gland within which it arose.[6] When dysplastic columnar epithelium proliferates to form a grossly visible polyp, the term *adenoma* may be applied. Dysplasia is recognized histologically by a combination of nuclear and architectural abnormalities.

Dysplastic columnar epithelium has cytologic abnormalities similar or identical to those seen in carcinoma (Fig. 142-1): cytoplasmic mucus is usually reduced or absent. Dysplastic epithelial nuclei are enlarged, often hyperchromatic, variable in size, shape and contour, and may contain prominent or multiple nucleoli; nuclear crowding and stratification are often present. Because of confounding reactive changes, the diagnosis of dysplasia should be made with caution when the mucosa is actively inflamed (Fig. 142-2). The presence of dysplasia should be seriously questioned if the neoplastic nuclear changes do not extend to include epithelia of the mucosal surface [6], [7] (see Fig. 142-1). Dysplastic glands may retain their normal architecture, or more frequently become irregular or even grossly distorted with intralumenal epithelial budding and back-to-back glands [7] (Fig. 142-3).

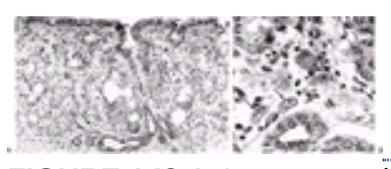

FIGURE 142-1. Low-power (**A**) and high-power (**B**) views of Barrett esophagus with no visible lesion by endoscopy but with high-grade dysplasia and intramucosal carcinoma by biopsy. Marked distortion of the glandular architecture and prominent nuclear abnormalities are seen. Invasion of the lamina propria by individual glandular epithelial cells represents intramucosal carcinoma (*arrows*).

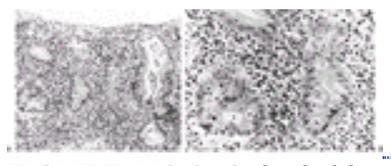

FIGURE 142-2. Colonic biopsy specimen in active ulcerative colitis. **A:** Notice the polymorphonuclear leukocytes within the crypt epithelium, the crypt abscess, the distortion of the crypt architecture, the basal lymphoplasmacytosis, and the marked chronic inflammatory infiltrate within the lamina propria. **B:** Reactive epithelial changes that can be confused with dysplasia are seen. Notice the occasional polymorphonuclear leukocytes within the crypt epithelium with enlarged, crowded, stratified nuclei containing numerous mitotic figures.

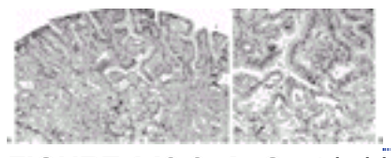

FIGURE 142-3. A: Gastric biopsy specimen shows high-grade dysplasia without invasion in its upper portion, but with gross distortion of glandular architecture forming a complex network of glandlike spaces deeper in the biopsy specimen (*arrow*). **B:** The architectural distortion is better seen at higher magnification of the area with an *arrow* in **A,** and the cytologic abnormalities of the nuclei lining the distorted glands can be better recognized. This gross distortion of the architectural pattern suggests possible invasion of the lamina propria.

Histologically, the degree of abnormality (atypia) within dysplastic mucosa forms a continuous spectrum from slight to marked. In certain situations it may be useful prognostically to grade dysplasia as low or high, based upon the degree of deviation from normal. This will be discussed subsequently.

Among experienced observers there is good inter- and intraobserver agreement on interpretation of high-grade dysplasia (HGD) [8] (Fig. 142-4). The nuclear abnormalities are more pronounced than those seen in low-grade dysplasia (LGD) with more marked nuclear enlargement and more irregular nuclear membranes. In HGD the nuclei may vary markedly in size, shape, and staining characteristics. There is usually loss of nuclear polarity (i.e., some of the elongated neoplastic nuclei that were originally oriented perpendicular to the basement membrane may round out or rotate from a vertical o a more horizontal or jumbled orientation). Invariably, the nuclear changes extend to the epithelial surface, where they are also enlarged and neoplastic in appearance. Goblet cell mucus is usually diminished or absent. Architectural distortion is almost always present and may be marked. Crypts or glands exhibit branching and lateral budding. One may see bridging epithelium extending across glands or crypts to form a cribriform “back-to-back” pattern with epithelial cells of adjacent acini touching one another without intervening lamina propria. A filiform epithelial configuration may also be seen at the mucosal surface. It may be difficult to differentiate HGD from invasive cancer (see Fig. 142-3), if architectural distortion is sufficiently severe, if back-to-back glands are growing into one another, if ill-defined abortive glands are present in the lamina propria, or if the limited depth of the biopsy specimens makes it impossible to determine whether glands are invading into the submucosa [7] (Fig. 142-5). In such situations, an appropriate diagnostic description might be “marked distortion of crypt or glandular architecture such that carcinoma can not be excluded.”

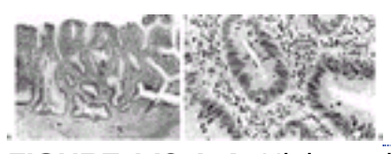

FIGURE 142-4. A: High-grade dysplasia complicating chronic ulcerative colitis. Notice the distorted glandular architecture, almost complete absence of goblet cell mucus, and marked crowding and stratification of nuclei. **B:** At higher power the cytologic abnormalities can be readily identified. The nuclei are markedly enlarged compared with the size of the nuclei in the lamina propria and they vary markedly in size and shape. A beginning loss of nuclear polarity also is seen.

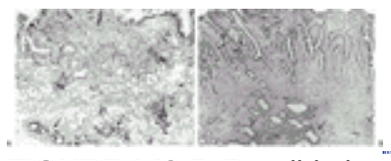

FIGURE 142-5. Possible invasive cancer in a biopsy specimen from a patient with Barrett esophagus. The corresponding area in the surgical specimen shows definite submucosal invasion. **A:** The upper portion of the biopsy specimen above the muscularis mucosae shows only high-grade dysplasia, whereas in the lower portion of the specimen, slightly dilated, darkly stained glands (*arrows*) extend into a widened and splayed muscularis mucosae. Because of the limited depth of the specimen, and the frequent noninvasive trapping of glands into previously ulcerated areas of Barrett esophagus, invasion through the muscularis mucosae into the submucosa cannot be confirmed, but was suspected. **B:** The surgical specimen from the same area clearly shows the dilated glands invading into the submucosa (*arrows*).

In general, there is less agreement on the diagnosis of LGD [8] (Fig. 142-6). The crypt architecture in low-grade dysplasia tends to be preserved, and distortion if present, is mild. The nuclei may be stratified, particularly near the base of the crypts, but the enlargement, hyperchromatism, and crowding is less severe than that seen in HGD. Cytoplasmic mucin is not necessarily diminished. Diagnosis of LGD still requires that the enlarged neoplastic nuclei extend to the epithelial lumenal surface but nuclear polarity is maintained. [7]

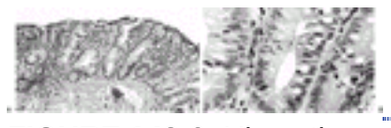

FIGURE 142-6. Ulcerative colitis with low-grade dysplasia. **A:** Notice the relatively well-preserved crypt architecture and the mild chronic inflammatory infiltrate within the lamina propria. There is no evidence of active inflammation. Notice the decreased number of goblet cells and nuclear crowding and stratification involving the crypts and extending to the surface epithelium. **B:** At high-power magnification of the crypts, the nuclear enlargement, hyperchromatism, crowding, and stratification can be readily seen. Notice some dystrophic goblet cells that fail to communicate with the lumenal surface. Such goblet cells are characteristic but not diagnostic of dysplasia.

Squamous Dysplasia

Squamous dysplasia of the esophagus has a similar nuclear neoplastic appearance to that seen in dysplasia of GI organs lined by columnar epithelium. The mucosa has an increased number of epithelial cells per unit area. The nuclei of these epithelial cells are pleomorphic, crowded, hyperchromatic, and overlap each other. The basal epithelial layer often does not “mature” by flattening horizontally and reducing its nuclear size as it approaches the superficial epithelium at the lumenal surface. The cells often assume a jumbled rather than a layered appearance. Squamous dysplasia of the esophagus is rare in the United States without an accompanying endoscopically apparent squamous carcinoma (Fig. 142-7). Most of the epithelial changes that mimic squamous dysplasia are caused by the benign regenerative and inflammatory response of esophagitis.

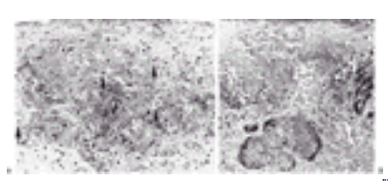

FIGURE 142-7. Esophageal squamous dysplasia and early carcinoma. **A:** Squamous dysplasia. The papillae of the lamina propria are irregular and the mucosa has an increased number of epithelial cells per unit area. The nuclei of these epithelial cells are crowded and hyperchromatic and overlap each other. The cells fail to mature while they move toward the lumenal surface, and numerous mitotic figures are present at all levels of the mucosa (*arrows*). **B:** An esophageal biopsy specimen from another mucosal area of the same patient shows both esophageal squamous dysplasia and early invasion of the lamina propria. This section shows tongues of dysplastic squamous epithelium extending into the lamina propria, some of which have become detached to form discrete nests of early invasive carcinoma.

In both columnar and squamous mucosa throughout the GI tract one must be especially careful in diagnosing dysplasia in inflamed, eroded, or ulcerated tissue infiltrated with polymorphonuclear leukocytes. The nuclear changes caused by inflammation or regeneration may be impossible to differentiate from dysplasia of any grade (see Fig. 142-2). Such changes are often seen in biopsy specimens near the edge of a benign ulcer. When uncertain in such cases, the diagnosis “indefinite for dysplasia” is appropriate. Repeat biopsy evaluation after successful treatment of inflammation can avoid this confusion and lead to the correct diagnosis.

Endoscopic biopsy surveillance for cancer is based upon several assumptions regarding dysplasia: the first assumption is that dysplasia is a histologically recognizable precursor of cancer. As mentioned previously, HGD is a diagnosis with an interpretation that is usually reproducible but the diagnosis of low-grade or indefinite for dysplasia is less so. It may also be difficult to differentiate HGD from early invasive carcinoma because biopsies often do not reach deeply enough into the submucosa to assess invasion through the muscularis mucosae (see Fig. 142-5). In the stomach and esophagus single epithelial cells invading the lamina propria indicate that HGD has progressed to intramucosal carcinoma except in the colon, but these cells are often difficult to identify with certainty. The diagnosis of intramucosal carcinoma is not made in the colon and the reasons for this exception are discussed later in the chapter under “Colonic Polyps.” Examining serial sections may demonstrate the ectopic position within the lamina propria of single invasive epithelial cells (see Fig. 142-1). Considering the difficulty in diagnosing dysplasia or early carcinoma, it is wise to seek an opinion from a second pathologist with considerable experience in the diagnosis of GI dysplasia and cancer before major clinical decisions. More objective techniques such as flow cytometric quantification of DNA may complement the histological diagnosis of dysplasia. [9] However, given the present state of knowledge, it is our belief that clinical decisions should still be based primarily on the histological appearance.

The second assumption is that cancers develop from dysplasia. There is evidence to support this assumption. [10], [11] How soon and often the dysplasia will become cancer in different patients is unpredictable. Prospective data on the natural history of these diseases is accumulating, but much work remains to be done.

The third assumption is that early malignancy can be detected by prospective surveillance while it is still curable by surgical excision. There are prospective studies that support this assumption. [12], [13] It is accepted clinical practice in the United States to place certain patients with Barrett esophagus, colonic adenomas, ulcerative colitis, or Crohn’s disease in surveillance protocols. Older adult patients from certain geographic areas with intestinal metaplasia of the stomach may also merit surveillance because of their increased likelihood of developing intestinal-type adenocarcinoma of the stomach. Some have questioned whether close surveillance is worth the time, effort, expense, and discomfort involved in detecting a relatively low number of patients with dysplasia or early curable carcinoma. Obviously it is worthwhile for those patients whose lives are saved but other, better techniques are needed to select subsets of patients whose high cancer risk warrants the cost and effort of such close surveillance. [14]

ESOPHAGEAL CANCERS

Esophageal Squamous Carcinoma

A history of treated squamous carcinoma of the mouth, pharynx, or nose merits a screening esophagoscopy for esophageal squamous dysplasia (see Fig. 142-7); if found, the patient should undergo frequent and extensive biopsy surveillance. Other rare predisposing diseases such as longstanding achalasia, lye strictures, Plummer-Vinson syndrome, and tylosis may merit screening esophagoscopy. Screening for esophageal squamous dysplasia or early carcinoma is generally not justified in the United States because of its decreasing frequency in many of ethnic groups. When one discovers squamous dysplasia with an early carcinoma of the

esophagus, one must remember that, unlike Barrett adenocarcinoma which is confined to the columnar epithelium, squamous dysplasia is characteristically multifocal throughout the length of the squamous esophagus. Thus, total esophagectomy is mandatory for possible cure.

Esophageal Adenocarcinoma

Adenocarcinoma is now far more frequent than squamous carcinoma of the esophagus in the United States. Barrett-specialized metaplasia is almost always the precursor of these esophageal adenocarcinomas. [15] Barrett is an acquired condition in which chronic reflux of gastric juice erodes the normal stratified squamous esophageal lining and it is replaced by metaplastic columnar epithelium defined by the presence of goblet cells. [7] This metaplasia is potentially precancerous regardless of how short or long the involved esophageal segment may be [7], [16] (Fig. 142-8). Barrett-specialized metaplasia comprises a spectrum of epithelial changes in which the diagnostic sine qua non is the presence of goblet cells containing acid mucus that stains intensely with Alcian blue at pH 2.5. [7], [17] The appearance of the columnar cells between the goblet cells varies from normal gastric surface or foveolar cells containing neutral mucus, to others containing mildly acid mucin staining lightly with Alcian blue, to cells resembling intestinal absorptive cells except for poorly developed brush borders. Barrett-specialized metaplasia is most commonly of the incomplete type. Complete intestinal metaplasia is composed of normal goblet and absorptive cells with Paneth cells at the bottom of some of the crypts. This is the common type of intestinal metaplasia seen in the stomach (Fig. 142-9).

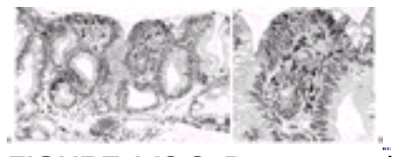

FIGURE 142-8. Barrett esophagus with low-grade dysplasia. **A:** The dysplastic glands to the right of center are irregular in size and shape, and have decreased mucus production. To the left and right, the characteristic goblet cells of the nonneoplastic, Barrett-specialized metaplastic epithelial precursor still can be identified. **B:** Observe the crowding, stratification, and enlargement of the dysplastic nuclei that extend to the lumenal surface. The changes are less severe than those seen in high-grade dysplasia, and there is maintenance of nuclear polarity, where the long-axis of the nuclei remain perpendicular to the basement membrane.

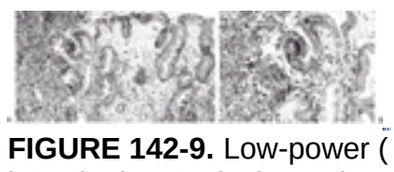

FIGURE 142-9. Low-power (**A**) and high-power (**B**) views of advanced *Heliobacter pylori*–caused multifocal intestinalized pangastritis (MIP) with decreased glands, intestinal metaplasia, and an associated adenocarcinoma. In the right half of the illustrations, the normal gastric glands have been replaced by complete intestinal metaplasia, as evidenced by the goblet cells and normal absorptive cells. To the left in **A** and **B,** sheets of malignant-appearing cells infiltrate the lamina propria.

Thirty-three percent of patients with nonerosive gastroesophageal reflux disease without endoscopic evidence of Barrett esophagus may have intestinal-type goblet cells right at the squamocolumnar junction if both the squamous and columnar epithelium are present in the same biopsy. [18] Biopsies 1 to 2 cm below the esophagogastric junction rarely have goblet cell foci. [18] This common finding of intestinal metaplasia at the normally located esophagogastric junction in gastroesophageal reflux disease should not be called Barrett esophagus because it will stigmatize patients with a false diagnosis of Barrett that will impair insurability.

Barrett esophagus and “columnar-lined esophagus” have been incorrectly considered identical. They are not. Almost all patients with Barrett esophagus have a hiatus hernia lined with normal gastric mucosa. If a biopsy is taken above the diaphragm within a hiatal hernia that is mistaken for tubular esophagus, it will show gastric surface and foveolar columnar epithelium covering fundal or cardiac glands (Fig. 142-10, Fig. 142-11). This should not be misinterpreted as Barrett “columnar” epithelium because what has been biopsied is stomach within a hiatus hernia and does not represent Barrett specialized metaplasia of the esophagus with its precancerous potential. The normal tubular esophagus in its distal 2 or 3 cm may have some cardiac (see Fig. 142-11) or fundal glands covered by columnar epithelium and these too must not be confused with Barrett-specialized epithelium with its goblet cells.

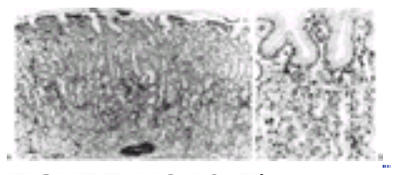

FIGURE 142-10. Biopsy specimen of a typical hiatal hernia pouch. **A:** The gastric fundal (oxyntic) glands are normal. The surface is covered by normal gastric surface mucus cells that dip into normal shallow foveolae. **B:** The lamina propria contains no recognizable inflammatory infiltrate, and numerous parietal cells are visible in the normal oxyntic glands (*arrows*).

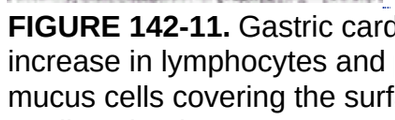

FIGURE 142-11. Gastric cardiac mucosa. **A:** The surface epithelium, foveolae, and glands are lined by cells producing mucus. The lamina propria contains a mild increase in lymphocytes and plasma cells, a feature found in the majority of biopsy specimens from this portion of the stomach. **B:** The appearance of the epithelial mucus cells covering the surface and lining the foveolae at higher magnification illustrates the lengthened columnar cells that are characteristic, but not diagnostic, of cardiac glands.

How can potentially curable adenocarcinoma be detected in Barrett esophagus? The first rule is to recognize that dysplasia and early intramucosal carcinoma (see Fig. 142-1) are usually grossly invisible. [10] The second rule is to methodically and repeatedly biopsy the whole length of Barrett specialized metaplasia within the tubular esophagus from the lower esophageal sphincter to the upwardly displaced squamocolumnar junction. Large, properly processed, and expertly interpreted biopsy specimens increase diagnostic sensitivity and specificity. The finding of dysplasia in Barrett esophagus provides an opportunity to prevent a uniformly fatal adenocarcinoma, yet the proper course of action is unclear when only HGD is found after extensive biopsy. There are three reasons for this uncertainty:

1. The older patient with dysplasia is often a poor surgical risk for esophagectomy.
2. The partial esophagectomy of all the columnar epithelium that eliminates metaplasia still has a sizable mortality, except when performed by exceptionally experienced and skillful surgeons. Even then, there is significant morbidity.
3. How long dysplasia takes to become carcinoma is unpredictable in each individual case.

Recent data on the natural history of Barrett dysplasia by the Seattle and the Hines VA groups indicate that the dysplasia diagnosed at its onset during careful biopsy

surveillance may take a mean of 6 years or longer to progress to early carcinoma that is still surgically curable. [12], [13]

Grading of dysplasia in Barrett esophagus has been shown by prospective study to be the single most important indicator of cancer risk. This is another benefit of endoscopic biopsy surveillance. [12]

If only HGD without adenocarcinoma is found after the extremely thorough sampling of Barrett mucosa recommended in the Seattle protocol, esophagectomy to exclude an undetected carcinoma may not be necessary provided the patient undergoes this compulsive biopsy surveillance at least annually. In our protocol we sample four quadrants of the involved esophagus at 2 cm levels beginning below the lower esophageal sphincter within gastric fundal mucosa, and extending upward throughout the tubular esophagus lined by Barrett-specialized metaplasia to beyond the ora serrata, where columnar epithelium joins the squamous lined esophagus. Any visible lesions are separately and additionally biopsied. In patients with HGD who remain in annual surveillance, the interval is shortened to 1 cm including areas of squamous surface overgrowth. Curable, intramucosal or early adenocarcinoma can be detected by such compulsive annual endoscopic biopsy surveillance. For surgical treatment, the entire columnar-lined portion of the esophagus must be removed, but the squamous-lined portion of the esophagus need not be excised. Alternatives to excision such as endoscopic photoablative treatment are being tested and show promise.

BENIGN ESOPHAGEAL DISEASES

Gastroesophageal Reflux Disease

Gastroesophageal reflux disease (GERD) is one of the most common diseases. It is usually caused by regurgitation of gastric or duodenal contents into the esophagus that injures or even destroys its squamous lining. When the endoscopic appearance of esophagitis has progressed to erosion, ulceration, or stricture, the histological diagnosis of esophagitis is usually obvious because the biopsy specimen shows erosions and active inflammation with intraepithelial polymorphonuclear leukocytes (Fig. 142-12). If the mucosa is intact endoscopically in a patient with reflux symptoms, as is often the case, it may be difficult to find diagnostic histological abnormalities by endoscopic biopsy.

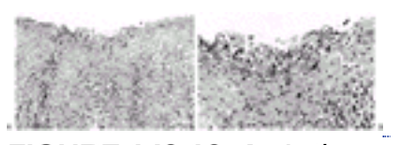

FIGURE 142-12. A: Active esophagitis in gastroesophageal reflux disease in which polymorphonuclear neutrophils and lymphocytes infiltrate the surface of the squamous mucosa and the lamina propria, obscuring the interface between the mucosa and the underlying lamina propria. **B:** Prominent epithelial hyperplasia of the basal layer is evidenced by the enlarged, hyperchromatic, and more numerous nuclei seen best at higher magnification.

Thus, the main problem in diagnosing GERD in patients with reflux symptoms is that many have a normal endoscopy without evidence of esophagitis even by biopsy. Some believe that histological evidence of hyperplasia of the squamous epithelium indicates reflux injury. In squamous hyperplasia, there is lengthening of the papillae of the lamina propria and thickening of the basal layer (Fig. 142-13). Furthermore, there is a significant correlation between the severity of reflux as measured by the 24-hour pH score and the length of the papillae. [19] Unfortunately, squamous hyperplasia as a histological indicator of esophagitis is not useful clinically for several reasons:

Most biopsy specimens taken with the standard-sized forceps are too small to orient for accurate architectural diagnosis of hyperplasia.
More than half of normal persons without GERD have squamous hyperplasia in the distal 2.5 cm of their tubular esophagus and one fifth have it proximally. [20]
Fifteen percent of patients with obvious symptoms of reflux and other positive tests for reflux show no histological evidence of active inflammation or hyperplasia. [21]

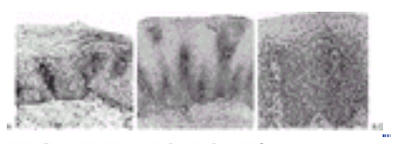

FIGURE 142-13. Biopsy specimens of normal and hyperplastic esophageal mucosa. **A:** Normal squamous mucosa of the esophagus. The papillae of the lamina propria extend through approximately 50% of the thickness of the mucosa, and the basal zone occupies a small portion of the mucosa. **B:** Hyperplasia of the mucosa is indicated by lengthening of the papillae of the lamina propria, some of which extend close to the lumenal surface (*arrow*). In addition, the basal zone is markedly hyperplastic and occupies more than 25% of the thickness of the mucosa. **C:** A profound degree of hyperplasia can be seen with the basal zone occupying two thirds of the thickness of the mucosa and with papillae of the lamina propria extending a similar distance through the mucosa (*arrow*). Note the architectural uniformity wherein the squamous papillae extend to the same depth within the lamina propria. This combined with the epithelial cytologic uniformity are the best features to differentiate benign squamous hyperplasia from dysplasia.

Some believe that the presence of several eosinophils per individual section of squamous epithelium indicates reflux esophagitis. We do not, because data published by Dr. Haggitt on 91 patients indicated that eosinophils did not correlate with a 24-hour pH score unless they were abundant (>6 eosinophils/biopsy). [22]

Active esophagitis with infiltrating polymorphonuclear leukocytes can progress to erosion or ulceration, or it can heal with reepithelialization or with stricture. The eroded squamous epithelium may regrow, or be replaced by specialized metaplastic columnar epithelium (i.e., Barrett esophagus).

Even in those patients with reflux symptoms who have the eroded endoscopic appearance of esophagitis, the histological picture is nonspecific and one must also consider a variety of rare systemic conditions such as eosinophilic gastroenteritis, collagen vascular disease, Stevens-Johnson syndrome, and bacterial stasis. Also, a carcinoma can masquerade occasionally as erosive or ulcerative esophagitis. Thus, esophageal biopsy of erosive esophagitis by endoscopy is not done to confirm GERD but rather to exclude infection, Barrett esophagus, or neoplasia.

Esophageal Infections

Esophageal infections are rare in immunocompetent persons, but immunocompromised hosts are susceptible to cytomegalovirus (CMV), herpes simplex virus (HSV), varicella zoster virus, or *Candida* organisms, all of which have a diagnostic histological pattern [23], [24] (Fig. 142-14). A spectrum of endoscopic appearances is possible with these infections. Biopsies, brushings, and viral cultures are indicated to make these specific diagnoses. Such infections are increasing in frequency due to the immunosuppression caused by acquired immunodeficiency disease (AIDS), or several iatrogenic causes such as chemotherapy or pharmacological prevention of rejection after organ transplantation.

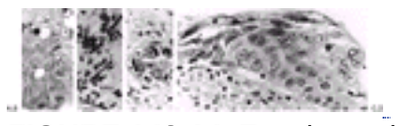

FIGURE 142-14. Esophageal biopsy specimens from various opportunistic infections. **A:** Hematoxylin and eosin (H & E) stained, high-power magnification illustrates esophageal squamous mucosa in which there are budding yeasts (*arrowhead*) and infiltrating pseudohyphae of *Candida* organisms (*arrow*). **B:** Periodic acid-Schiff

stain for carbohydrates highlights pseudohyphae of *Candida* organisms (*arrow*) more clearly than an H & E stain. **C:** A high-power photomicrograph of esophageal mucosa in which cytomegalovirus is present. The *arrowhead* indicates a diagnostic inclusion body with a surrounding halo within the nucleus of an enlarged mesenchymal cell. This cell and the one marked by the arrow also contain granular cytoplasmic inclusions. **D:** The characteristic findings of herpes simplex virus infection: ground-glass nuclei, a Cowdry type A inclusion body (*arrow*), and multinucleated giant cells (*arrowhead*).

Fungal esophagitis is most commonly caused by *Candida albicans* and *Candida tropicalis.* Other fungi are rare. They occur primarily in immunosuppressed, diabetic, or antibiotic-treated patients. They may also be seen in otherwise healthy subjects. *C tropicalis* is more virulent than *C albicans* because of its increased invasiveness. [25] The budding yeasts and pseudohyphae of *Candida* organisms may be recognized in tissue stained with H & E, but are seen far more easily in biopsy specimens stained with periodic acid-Schiff (see Fig. 142-14A, Fig. 142-14B). A few yeast forms within surface debris are extremely common and probably represent oral contamination. The presence of pseudohyphae within the tissue indicates invasive infection, but their presence only in adherent exudate does not exclude it, especially if clinical evidence suggests dissemination.

CMV may be found in the endothelial cells and fibroblasts in the granulation tissue in the base of ulcers and erosions, but also may be identified within columnar epithelial cells (see Fig. 142-14C). Isolated enlarged cells containing CMV may be time-consuming to find histologically but careful histological examination is equal to, or superior to, ancillary techniques for its detection, such as immunohistochemistry. The characteristic features of CMV are cytomegaly with a large viral intranuclear inclusion occupying most of the nucleus except for a surrounding halo and granular inclusions in the cytoplasm. CMV esophagitis alone, or in combination with *Candida* and herpes is relatively common in AIDS. The accurate diagnosis of CMV is important because without early treatment it may be fatal but early effective therapy is available. No single technique for diagnosis of CMV has a very high sensitivity; thus, a combination of diagnostic modalities such as shell viral centrifugation culture and in situ hybridization with specific DNA probes may also be used if the findings on histological examination with H & E staining are uncertain and clinical suspicion is high. [26]

Herpes simplex virus (HSV injury) shows a diagnostic histological picture in the surface squamous epithelium of the esophagus (Fig. 142-14D). Herpetic ulcers are very painful, shallow, sharply punched out, and often surrounded by relatively normal mucosa. Biopsies should be taken from the squamous epithelium at the immediate edge of the lesion in order to be diagnostic. Cells infected by HSV are smaller than those infected with CMV and contain a smaller intranuclear viral inclusion (Cowdry type A). Ground-glass nuclei are made up of multiple small eosinophilic particles producing a smudged appearance; multinucleated giant cells and ballooning degeneration of infected squamous cells may also be seen. HSV type I causes most herpetic esophagitis but it cannot be distinguished on morphologic grounds from the rarer HSV type II or from varicella-zoster. Immunocytochemical staining and in situ hybridization using DNA probes can separate these three organisms.

Less common pathogens may be seen in the immunosuppressed patient such as *Mycobacterium tuberculosis, Mycobacterium avium-complex,* histoplasmosis, or toxoplasmosis. In some patients with AIDS, there is no apparent cause for the esophagitis or ulcer but there is speculation that the human immunodeficiency virus (HIV) itself may be etiologic because it has been detected in the epithelium and lymphocytes of the GI tract. [27]

Other Benign Esophageal Conditions

A variety of other benign conditions are diagnosable by endoscopic biopsy. The most common is the clinically unimportant entity of glycogenic acanthosis. This condition is diagnosed by biopsy of small white nodules or plaques that can be shown to contain squamous epithelium whose cytoplasm is distended with excess cytoplasmic glycogen. The glycogen can be identified by periodic acid-Schiff–stained particles that disappear after glycogenolysis with diastase.

Other rare esophageal diseases that are difficult to diagnose by endoscopic biopsy alone include sarcoidosis, Crohn's disease, Behçet disease, graft-versus-host disease, and the esophageal lesions seen in patients with bullous skin disease.

Esophageal injury caused by prolonged direct mucosal contact with tablets or capsules taken in therapeutic doses occurs frequently. [28] The histological appearance of ulcers and strictures induced by pills is nonspecific but one may see prominent infiltration of eosinophils, or spongiosis and necrosis of squamous epithelium. Birefringent crystalline material may be seen in alendronate-induced injury, [29] and crystalline iron in ferrous sulfate-induced disease. [30] Esophageal erosions and ulcers were found in 20% of patients with arthritis taking nonsteroidal antiinflammatory drugs (NSAIDs), [31] but esophageal injury by NSAIDs may be less common than such injury in other parts of the GI tract.

Benign Esophageal Tumors

Clinically significant benign esophageal tumors are uncommon. The most frequent benign tumor is submucosal leiomyoma, which arises from the muscularis mucosae but it may require a deeper biopsy for diagnosis than is usually provided. A squamous papilloma is a papillary growth composed of fingerlike projections covered by lamellated benign squamous epithelium; it is usually located in the distal third of the esophagus (Fig. 142-15). Inflammatory fibroid polyps contain active inflammation and eosinophils in a prominent fibrovascular stroma; these can become quite large and can be mistaken grossly for carcinoma. A submucosal tumor of polygonal cells filled with abundant, variable-sized granules is almost certainly a granular cell tumor (Fig. 142-16). Lipomas may also be difficult to diagnose by biopsy because they are mostly submucosal.

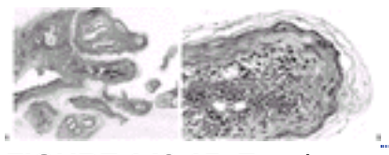

FIGURE 142-15. Esophageal biopsy specimen of a squamous papilloma. **A:** Papillary (fingerlike) projections are characteristic. **B:** The central fibrovascular cores of the papillae are covered by lamellated, benign, squamous epithelium.

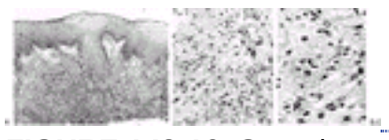

FIGURE 142-16. Granular cell tumor of the esophagus. The lamina propria (**A**) contains sheets of enlarged polygonal cells with granular cytoplasm in **B** and **C**.

GASTRIC CANCERS

Gastric Adenocarcinomas

There are three types of gastric adenocarcinoma: intestinal-type adenocarcinoma, diffuse adenocarcinoma of the stomach, and cardia adenocarcinoma.

Intestinal-type adenocarcinoma is the most common worldwide, although it is decreasing markedly in frequency in the Western world. It is called intestinal-type because it forms glands that resemble those of colonic cancer. It arises in a multifocal intestinalized pangastritis caused by HP (see Fig. 142-9). In an unpublished 15-year longitudinal study [32] of 122 elderly, high-risk Japanese-Americans with intestinal metaplasia undergoing extensive prospective endoscopic surveillance every 3 years, metaplasia progressed to dysplasia followed by intestinal-type adenocarcinoma in 10 patients; in 8 of these patients, the cancer was discovered while surgically curable. The two incurable carcinomas were in patients who failed to return on time for scheduled follow-up. Thus, early intestinal-type adenocarcinoma of

the stomach can be detected even in the United States during close surveillance of a selected population.

Diffuse adenocarcinoma of the stomach is less common. It does not form glands but is made up of diffusely infiltrating single malignant epithelial cells (Fig. 142-17). It offers a diagnostic challenge to pathologists because it can be easily under-diagnosed or over-diagnosed. The infiltrating malignant cells may be sparse, hidden within the lamina propria, and deceptively bland. They may mimic collections of macrophages or xanthelasma within the lamina propria. Histochemical stains for mucin may help identify goblet cells or immunohistochemical markers for cytokeratin can be useful in identifying these infiltrating malignant cells in the lamina propria as epithelial.

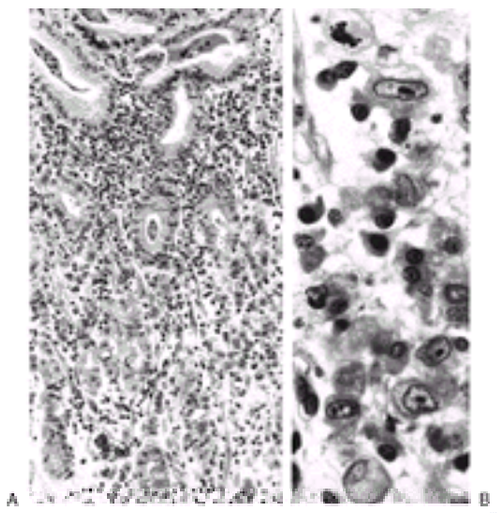

FIGURE 142-17. Gastric biopsy specimen of a minute lesion of diffuse adenocarcinoma that was barely visible endoscopically. **A:** Low-power magnification illustrates how subtle diffuse adenocarcinoma of the stomach can be. The inflamed lamina propria contains infiltrating malignant cells that are difficult to recognize because they are interspersed with inflammatory cells. **B:** The high-power view makes these infiltrating, malignant cells more apparent.

Cardia adenocarcinoma is the third type of gastric adenocarcinoma and it is located in the uppermost stomach involving the gastroesophageal junction. It has clinical and epidemiologic features in common with Barrett esophageal adenocarcinoma, [33] the most rapidly increasing carcinoma in Caucasian American males. [34] Cardia carcinomas and short-segment Barrett adenocarcinomas are probably the same entity. The tumors probably originate in a short distal segment of Barrett-specialized metaplasia that can be easily missed during endoscopy for reflux esophagitis unless several biopsies are always taken at an irregular or jaggedly serrated esophagogastric junction. The finding of even one biopsy with unequivocal dysplasia on initial endoscopy is a red flag indicating the need for extensive and repeated sampling to be certain that early cancer has not been missed. We have diagnosed several early curable cardia carcinomas in this manner (Fig. 142-18).

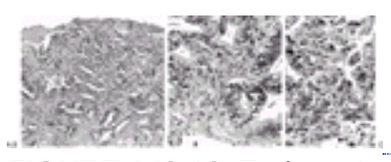

FIGURE 142-18. Early, potentially curable adenocarcinoma of the gastric cardia. **A:** At low-power magnification both high-grade dysplasia and a focus of intramucosal carcinoma (*single arrow*) are present. Residual goblet cells suggest the presence of Barrett-specialized metaplastic epithelium (*double arrows*). **B:** The area previously marked with double arrows shows the enlarged goblet cells that define this dysplastic epithelium as originating from Barrett-specialized metaplasia (*arrowheads*). **C:** In the area marked in **A** with a *single arrow*, malignant cells infiltrating the lamina propria are evident. This patient is alive and well 5 years after local surgical resection of this lesion. Lymph nodes were free of tumor, and the carcinoma did not extend beyond the mucosa.

Gastric Lymphomas

Gastric lymphoma, while far less frequent than adenocarcinoma, is well worth diagnosing while confined to the stomach, especially because low-grade lymphoma of the mucosa-associated lymphoid tissue (MALT) (Fig 142-19) is reversible and potentially curable by a pharmacological regimen for HP eradication. [35] Histologically, it may be difficult to differentiate HP gastritis from low-grade B-cell gastric (MALT) lymphoma. The diagnostic challenge is to distinguish MALT lymphoma from the reactive lymphoid hyperplasia that commonly accompanies HP-induced gastritis. MALT lymphoma is characterized by a dense lymphoid infiltrate, often accompanied by eosinophils and plasma cells. The lymphocytes resemble small, cleaved lymphocytes or monocytoid B-cells. The lymphoid cells infiltrate the gastric epithelium to produce lymphoepithelial lesions, and ultimately destroy the epithelium (see Fig. 142-19). These lymphocytes are thought to be derived from a mantle zone-like B-cell unique to MALT. [36] Histological evaluation remains the most sensitive and specific method for the diagnosis of low-grade lymphoma of MALT. [38] The demonstration of light chain restriction by immunohistochemistry can establish the presence of a clonal proliferation, but the sensitivity of this technique is relatively low, as only about 40% of cases contain detectable cytoplasmic immunoglobulin by immunohistochemistry. Unfortunately, polymerase chain reaction (PCR) analyses for heavy chain rearrangement are also relatively unreliable diagnostically because of detection of small clonal populations that are probably reactive in nature in gastritis with no morphologic or clinical evidence of lymphoma. [39], [40]

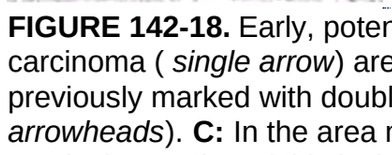

FIGURE 142-19. Gastric mucosa-associated lymphoid tissue (MALT) lymphoma. **A:** Low-power picture could be confused with *Helicobacter pylori* gastritis, but there is a lymphoid infiltrate, a reduction in the number of glands, and lymphoepithelial lesions (*arrow*). **B:** High power of a lymphoepithelial lesion shows small, cleaved lymphocytes infiltrating and destroying the epithelium (*arrows*).

Substantial evidence implicates HP as an etiologic factor in MALT lymphoma. Evidence of HP is present in over 90% of gastric MALT lymphoma cases and the HP infection precedes the lymphoma. Thus, it is not surprising that HP eradication is followed by regression in many patients with these low-grade lymphomas. [35], [37] A reasonable approach to the patient with gastric MALT lymphoma may be eradication of HP with subsequent endoscopic follow-up, with surgical resection reserved for refractory complications (bleeding, gastric outlet obstruction, or high-grade lymphoma). A predominance of large malignant lymphoid cells indicates high-grade transformation, a condition almost always unresponsive to HP eradication alone. How often regression of MALT lymphoma is permanent after HP eradication will require longer prospective studies than are currently available.

Lymphomas can develop after solid organ and bone marrow transplantation. The majority are B-cell Epstein Barr virus-driven proliferations. Histologically, these disorders vary from polyclonal plasmacytic hyperplasia to immunoblastic lymphoma, a monoclonal, frankly malignant lymphoma. Involvement of the GI tract is not uncommon and has been reported to be more common in patients receiving cyclosporine.

Gastric Carcinoid and Enterochromaffin Cell-like Tumors

Microscopically, carcinoids show monotonously similar small round cells growing in cords, glands, or nests, which invade the lamina propria and submucosa (Fig. 142-20). Gastric endocrine cell hyperplasia when it becomes endoscopically visible is considered to be a carcinoid. It is seen in a variety of conditions with a chronically elevated serum gastrin, such as the fundal atrophy of pernicious anemia, Zollinger-Ellison syndrome, or less commonly HP-induced intestinalized pangastritis. Gastric carcinoids associated with hypergastrinemia are indolent and rarely metastasize. Sporadic carcinoids not associated with hypergastrinemia commonly behave much more aggressively, making this distinction important.

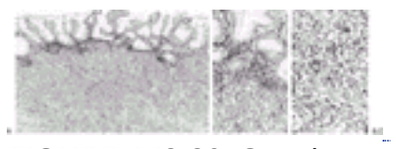

FIGURE 142-20. Gastric carcinoid. **A:** Small nests, glands, and sheets of neoplastic cells have replaced most of the mucosa and invade into the muscularis mucosae and submucosa. **B:** A higher magnification reveals nests of carcinoid cells in the lamina propria beneath the gastric pits. **C:** At the highest magnification the small, round, uniform nuclei with the finely stippled chromatin pattern characteristic of carcinoid can be seen.

Gastric Gastrointestinal Stromal Tumors

Gastrointestinal stromal tumors (GIST) are much more common in the stomach than in the rest of the GI tract. They were originally considered to be of smooth muscle differentiation. They are now recognized to share the phenotype of the interstitial cells of Cajal and as such express the *c-kit* protooncogene product for which a diagnostic immunohistochemical test exists. [41], [42] Grossly, these tumors commonly have surface ulceration. Microscopically GIST is submucosal and deeper in location and does not arise from the muscularis mucosae, as do the esophageal and anorectal true leiomyomas. The tumors may be composed of spindled or epithelioid cells. They may have a vacuolar pattern and other highly variable morphology. They can be benign or malignant and it is difficult to predict their behavior with certainty, although size and mitotic activity, invasion of the lamina propria, or small cell cytology are of help. It is important to diagnose GIST because treatment with the ligand of the *c-kit* receptor, or stem cell factor, is dramatically effective in previously untreatable malignant GIST. [43]

Intramural Extramucosal Tumors

Other rare lesions include intramural, extramucosal tumors derived from muscle, fibrous tissue, nerve, or fat. Endoscopically all may show bridging mucosal folds extending from the top of the tumor to the surrounding mucosa, an appearance reminiscent of the flying buttresses of an ancient cathedral. It is often difficult during biopsy to sample deeply enough of these extramucosal lesions to provide a diagnosis. Many ulcerate and bleed; obstruction occurs in some. A variety of vascular tumors may be seen. Pancreatic nests occasionally are evident on gross inspection.

Kaposi Sarcoma

Kaposi sarcoma in AIDS presents endoscopically as multiple violaceous, flat, macular lesions or as nodules that may have a surface that resembles a strawberry or elephant hide, or infrequently as volcano lesions with superficial erosions at their peaks. It is often missed by shallow biopsies, although experienced endoscopists often suspect the diagnosis grossly in patients with HIV infection. The diagnostic histological picture is one of spindle cells containing clusters or individual hyaline, eosinophilic, cytoplasmic inclusions and erythrocytes within slit-like openings between the streaming spindle cells [44] (Fig. 142-21).

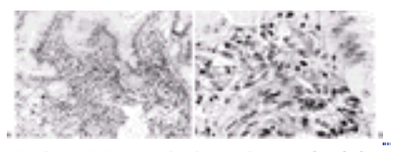

FIGURE 142-21. Gastric biopsy specimen from a patient with Kaposi sarcoma. **A:** The lamina propria contains infiltrating spindle cells of the sarcoma and a dilated vessel at the bottom. The arrow marks the area shown at higher power (**B**). Observe the spindle cells that compose the lesion and the red blood cells in the slitlike spaces between individual spindle cells. The *arrowheads* denote intracytoplasmic hyaline inclusions that are highly characteristic of Kaposi sarcoma. Note the mitotic figure immediately to the right of the arrowheads.

BENIGN GASTRIC DISEASES (Table 142-1)

TABLE 142-1 Diffuse Benign Gastric Mucosal Abnormalities

Gastritides

Gastritis is an inflammatory lesion and contrasts with gastropathy, another common benign gastric mucosal lesion that does not contain much or any inflammation. The classification of gastritis has been greatly advanced because of the recognition that HP causes most of the gastritides, and NSAIDs most of the gastropathies.

The gastric mucosa infected by HP shows neutrophilic infiltration of the gastric epithelium, primarily near the base of the foveolar pits and in the upper part of the glands. Neutrophilic infiltration indicates active inflammation but it should not be designated “acute gastritis” which has a chronologic implication. More appropriate terminology for describing chronic inflammation with neutrophilic epithelial infiltration in the lamina propria separating the foveolae is “chronic active gastritis.” Careful

inspection of the apical region of the surface and pit (foveolar) epithelial cells almost always (=90%) shows HP within the mucus layer, between it and the surface of the epithelial cells, attached to the surface of the epithelial cells, or between epithelial cells. The surface and pit epithelium may show evidence of injury. Lymphoid follicles within the lamina propria are highly characteristic of HP infection. [45]

The construction of a useful classification of gastritis presents many problems: First, there is poor correlation between the endoscopic and histological appearance of the gastric mucosa. Endoscopically normal mucosa may show significant biopsy abnormalities. Conversely, the endoscopic diagnosis of "gastritis" is often made when the endoscopist sees reddening which presumably reflects hyperemia of the mucosa that usually proves normal histologically. [46] Histological evidence of HP gastritis is highly likely to be present (specificity high) if the endoscopist sees antral nodularity, but the nodularity is not present in most adults with HP gastritis (sensitivity low). [46] Thus, the endoscopist should be encouraged to describe observed gross changes rather than to attempt the histological diagnosis of gastritis through the endoscope. Secondly, the topographic distribution of gastritis is important in its classification, but is rarely well documented because of inadequate endoscopic biopsy sampling. Thirdly, ethnic, geographic, and environmental factors appear to be decisive in determining the type of gastritis that a population develops, but these differences are rarely considered in its classification. Finally, there is the problem in classifying two types of coexisting gastritis. For example, in a patient with a benign gastric ulcer it may be impossible to determine the etiologic and histological contribution of NSAIDs that the patient is taking as opposed to HP with which the patient is infected.

These aforementioned problems have resulted in a lack of consensus on the classification of gastritis. The "Sydney" system for classifying gastritis was an attempt to resolve this lack of consensus; [47] however its histological division, if used as recommended, is exceedingly complex, purely descriptive, and not particularly helpful in everyday practice. A group of individuals interested in the pathology of gastritis has attempted to make the Sydney system more user friendly, [48] but it remains a descriptive method of reporting, rather than a nosologic classification correlating clinically different gastric diseases with different histological patterns of gastritis.

Many of the problems in classifying gastritis are caused by different ways of handling the earlier term "chronic nonspecific gastritis." In our opinion, the subdivisions of HP-caused gastritis proposed by Correa [49] make the most sense, and we shall use them with some nosological and histological modifications to reflect our beliefs regarding the relation between the different patterns of gastritis and different diseases. In our present state of knowledge, it is our opinion that all classifications are provisional. Our classification of gastritis patterns is a modification of Correa's. [50] We agree with the classification *diffuse antral-predominant gastritis* (DAG) with HP. We break the *multifocal atrophic gastritis* (MAG) classification of Correa into two different categories: [50] *multifocal intestinalized pangastritis* (MIP) with or without HP and *nonulcer pangastritis* (NUP) with HP. In addition there is the autoimmune gastritis of pernicious anemia— *diffuse corporeal atrophic gastritis* (DCAG) without HP.

In the United States and many other Western countries, the most common pattern of HP gastritis is DAG, a classification that is widely accepted. The presence and extent of intestinal metaplasia (IM) within the antral pyloric gland mucosa in DAG varies in different geographic areas and ethnic groups of the U.S. The antrum is almost always infected by HP and shows a diffuse chronic, active inflammatory infiltrate that expands the lamina propria between the foveolae, pushing the glands apart and basally so that there is an apparent decrease in the number of glands (Fig. 142-22A). Some call this atrophic gastritis; however, there is no real decrease in the number of glands in diffuse antral gastritis, as they will return in their original density and normal position after eradication of HP (see Fig. 142-22B). The gastritis of DAG may extend to varying degree from the antrum along the lesser curvature, but almost all fundal glands in DAG are *free of intestinal metaplasia* and have less gastritis than that present in the pyloric glands of the antrum. A strong association between DAG and both duodenal and juxtapyloric ulcers has been noted, and the risk of intestinal-type gastric adenocarcinoma is low in this setting. However, one must remember that only a minority of patients with any type of HP gastritis, including DAG, develop ulcers. Eradication of HP heals any ulcers that may be present, reverses the gastritis, but has no effect on any intestinal metaplasia that may be present.

FIGURE 142-22. Diffuse antral-predominant gastritis (DAG) associated with duodenal ulcer before (**A**) and after (**B**) successful eradication of *Helicobacter pylori.* **A:** Prominent interfoveolar inflammation with the pyloric glands pushed basally. **B:** A marked reduction in the number of mononuclear cells in the interfoveolar lamina propria. The pyloric glands of the antrum have resumed their normal position, and the antral mucosa has returned to an almost normal appearance.

Both MIP and NUP have gastritis involving the antrum and fundus. In addition IP contains extensive multifocal IM in both antrum and fundus but NUP has less or none. MIP may be associated with benign gastric ulcer and intestinal-type adenocarcinoma (see Fig. 142-9) but NUP is not. Neither NUP nor MIP is associated with duodenal ulcer.

In the United States, MIP is now far less common than DAG although it may be seen more frequently in certain minority populations [51], [52] (i.e., Native Americans and probably Hawaiians, African Americans, and immigrants from Eastern Europe, Asia, and South America). Thus, although HP is strongly associated with MIP, ethnic, environmental and dietary factors are important, if not essential, for its development. The antral and fundal pangastritis of MIP (Fig. 142-23) is histologically like that seen in NUP and in the DAG antrum. As Stemmerman and Hayashi have shown, [53] the multifocal intestinalized gastritis in MIP begins along the lesser curvature at the junction between the antral pyloric glands and the fundal glands. It then spreads proximally and distally and is the most intestinalized and progressive of the HP gastritides. In some patients IM is the precursor of dysplasia which becomes gastric adenocarcinoma of the intestinal type. The risk of this cancer is proportional to the extent of the intestinal metaplasia. [54] In other patients the IM is associated with benign gastric ulcer that does not become malignant with the passage of time. [32] The HP gastritis and the benign gastric ulcers of MIP are reversed by HP eradication (see Fig. 142-23), but the IM and intestinal-type adenocarcinoma are not. [32] Claims that IM is reversible are probably based on inadequate sampling. When MIP is advanced and intestinal metaplasia is extensive, HP may disappear. Until long-term, prospective data with multiple gastric biopsies are available it will remain unknown whether early treatment by dietary modification or eradication of HP will prevent development of MIP with intestinal metaplasia, benign gastric ulcers, dysplasia, and intestinal-type adenocarcinoma.

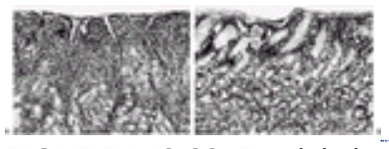

FIGURE 142-23. Fundal glands of the gastric body in multifocal intestinalized pangastritis (MIP) before (**A**) and 8 months after (**B**) *Helicobacter pylori* eradication. **A:** Note the inflammatory infiltrate in the upper half of the mucosa pushing the parietal and chief cells of the fundal glands basally. **B:** Note the complete restitution of normal fundal glands after *H pylori* eradication. Other focal areas of intestinal metaplasia (not illustrated) did not reverse their metaplasia after *H pylori* eradication.

We call the second type of pangastritis NUP because gastric ulcers, duodenal ulcers, and intestinal-type adenocarcinoma are rarely if ever seen and there is considerably less IM than in MIP. NUP has not been studied extensively because it is difficult to justify endoscopy with extensive biopsy in individuals with a normal upper endoscopy and few if any symptoms. Nevertheless, limited longitudinal data suggest that untreated NUP progresses more slowly than MIP and is present in many asymptomatic patients who are infected with HP. [3] The pangastritis of NUP is reversible after HP eradication. Whether NUP can ever evolve into MIP is unknown.

DCAG is not caused by HP and is confined to the fundal gland area. This is a genetically determined, progressive autoimmune destruction of the parietal cells, but the pyloric glands of the antrum are unaffected except for occasional concomitant HP antral gastritis. These patients develop an autoantibody directed to the proton pump on the canalicular membrane of the parietal cell. As the glands are progressively destroyed, they are usually replaced by intestinal metaplasia or less frequently by pyloric gland metaplasia, by pancreatic acinar metaplasia, or by nothing at all (atrophy). Destruction of the fundal glands results in achlorhydria and loss of intrinsic

factor, both of which are manufactured in the parietal cells. When gastric acid is low, gastrin secretion increases, predisposing to gastric enterochromaffin cell-like (ECL) hyperplasia and carcinoids. The lack of intrinsic factor prevents normal ileal absorption of vitamin B_{12} leading to the eventual development of pernicious anemia. As the gastritis progresses and the parietal and chief cells are destroyed, the extent of metaplasia or gland loss increases and the inflammatory infiltrate diminishes producing a picture sometimes referred to as gastric atrophy.

The older term “superficial” gastritis does not appear in the previous classification because it refers to a grade of gastritis rather than to a single histological pattern or nosological category; thus it has no independent clinical or histological significance. A substantial body of evidence indicates that HP is a causative agent also of acute epidemic gastritis with hypochlorhydria. We speculate that acute HP gastritis evolves into either DAG, MIP, or NUP, depending upon environmental influences. HP is also an important factor in the development of gastric or duodenal ulcer as well as intestinal-type adenocarcinoma. Only half of benign gastric ulcers are infected with HP in the United States and the other half are associated mostly with NSAIDs. One suspects that even when HP gastritis is present in gastric ulcers, many of these ulcers are caused by superimposed NSAID injury. The frequency of gastric ulcers caused by NSAIDs, and of duodenal ulcers caused by HP-DAG, is increasing in those areas where benign gastric ulcers and intestinal-type adenocarcinomas caused by HP-MIP are decreasing in frequency. Although all HP infected patients have gastritis, the majority will never develop an ulcer or cancer. The histological patterns of DAG, MIP, or NUP may be present without HP if the patient recently ingested an antibiotic because HP may disappear shortly after the start of an antibiotic.

To differentiate the four patterns of gastritis with certainty in our opinion requires at least six representative biopsies of the stomach. To save money, the six biopsies can be mounted three each in two blocks because most laboratories charge by the block. The biopsies in the first block should contain two from the lesser curvature of the antrum near the pylorus, and one from the mid-greater curvature of the stomach in an area of heavy folds. The three additional biopsies in the second block should contain one from the upper greater curvature, one from the middle of the crescentic fold that marks the angulus, and one from the lesser curvature 2.5 cm proximal to the angulus. Foci of intestinal metaplasia in four of the six biopsies suggest the diagnosis of MIP and differentiate it from NUP, which tends to have less than two biopsies (or no biopsies) with intestinal metaplasia. The DAG pattern is diagnosed when there is diffuse pyloric gland gastritis of the antrum and a lesser degree of gastritis in fundal glands that are *free of intestinal metaplasia.*

Although it has been said that 90% of patients with duodenal ulcers have DAG with HP infection, this number is decreasing because of widespread treatment of HP. Those patients with a duodenal ulcer and no HP-DAG most frequently are taking NSAIDs which causes an erosive, reactive gastroduodenopathy with ulcerations (duodenal ulcer and gastric ulcer). On rarer occasions, the duodenal ulcer is caused by Crohn’s disease, gastrinoma, or short bowel syndrome, or is idiopathic.

Intestinal metaplasia is subdivided histologically into two types: Complete intestinal metaplasia contains goblet cells, Paneth cells and absorptive cells with a well-developed brush border resembling those seen in the small bowel. Incomplete intestinal metaplasia also contains goblet cells, but the cells between the goblet cells may resemble gastric surface cells or altered absorptive cells that may secrete either sialomucins or predominantly sulfated mucins. Incomplete intestinal metaplasia that contains epithelium that secretes sulfated mucins may be more closely associated with increased risk of cancer, but this relationship has been challenged. We believe that subtyping of intestinal metaplasia need not be performed for diagnostic purposes.

A spiral bacterium that differs from HP may be found in the gastric mucosa of some patients with active gastritis. The organism originally named *Gastrospirillim hominis,* is longer and has more spirals than HP and appears to be a significant but uncommon cause of gastritis. Studies of the 16S ribosomal RNA gene sequences of this organism show that it is closely related to *Helicobacter,* and it is now called *H heilmannii.*

Gastropathies

Reactive, erosive gastropathies are caused by agents or factors that injure the gastric mucosa without producing a pronounced inflammatory infiltrate like that seen in the gastritides. NSAIDs are the most common cause. [55]

The mechanism by which reactive, erosive gastropathy develops is not completely understood; however, increasing evidence suggests that microvascular injury to the mucosa by NSAIDs plays an important role in addition to any direct effect by the NSAIDs on the epithelium. Early erosions are remarkably similar histologically to mild ischemic changes with superficial zonal necrosis and hemorrhage.

The lesion adjacent to an ulcer or erosion in reactive gastropathy is characterized by prominent foveolar hyperplasia with little or no gastritis and no HP infection. In foveolar cell hyperplasia there is elongation of the gastric pits with a “corkscrew” or serrated appearance of the longitudinal profiles of the pit lumen (Fig. 142-24). The individual hyperplastic foveolar cells may be normal, as they often are near the edge of an NSAID gastric ulcer, or they may have decreased amounts of mucus and large hyperchromatic nuclei especially in a healing erosion or ulcer.

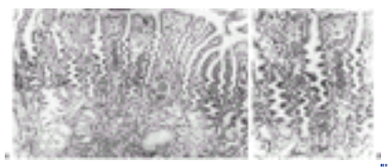

FIGURE 142-24. Reactive gastropathy near the edge of a gastric ulcer associated with ingestion of nonsteroidal antiinflammatory drugs. **A:** Notice the foveolar hyperplasia, as evidenced by the corkscrew or pleated configuration of the elongated gastric foveolae (*arrows*). The lack of gastritis is indicated by the paucity of polymorphonuclear leukocytes within the interfoveolar lamina propria. **B:** The foveolar hyperplasia with its lengthened corkscrew configuration is illustrated at higher magnification (*arrow*). Notice the nuclear crowding and decreased quantity of mucus in the foveolar epithelium, but maintenance of basal uniform-sized nuclei. No *H pylori* were present.

Many patients with reactive gastropathy also develop mucosal petechiae and erosions. In these patients, multiple subepithelial hemorrhages (petechiae) and hemorrhagic gastric erosions are seen endoscopically. Sometimes the entire mucosa may appear to be bleeding. Such an endoscopic appearance is caused by a reactive, erosive gastropathy not by an HP gastritis. Endoscopic biopsy may show histological evidence of widespread superficial subepithelial hemorrhage [56] in addition to erosions (Fig 142-25). Active inflammation is usually mild or absent. The erosions may have an appearance of superficial ischemic necrosis with pseudomembranes. Healing erosions may show foveolar hyperplasia with regenerative epithelial alterations. This pathological picture of erosions and superficial hemorrhage has multiple causes: injury from drugs, especially NSAIDs and alcohol; stress from burns, trauma, or severe illness; ischemia from shock or atherosclerotic narrowing of the celiac axis or its branches; and trauma from previous passage of tubes or endoscopes.

FIGURE 142-25. Gastric fundal mucosa in reactive, erosive gastropathy. **A:** There is a focus of erosion (*open arrows*) of the surface epithelium with fibrinopurulent exudate adherent to the mucosal surface. Notice the absence of active inflammation within the gastric mucosa proper. **B:** Adjacent to the erosion, a focus of subepithelial hemorrhage can be seen and is marked with *arrowheads* at higher magnification.

With the passage of time the hemorrhagic and necrotic erosions may extend through the muscularis mucosae into the submucosa to become an acute ulcer. These can bleed or perforate. An ulcer is differentiated from an erosion because it penetrates through the muscularis mucosae into the submucosa and an erosion does not. Unlike in ulcers, resolution of the erosive lesions results in complete, rapid healing of the mucosal defects. The rapid regeneration of the mucosa that occurs in

erosive, reactive gastropathy may produce foveolar hyperplasia with atypical nuclei, and this picture can be misinterpreted as dysplastic or malignant. For this reason, one should be very cautious about making the diagnosis of neoplasia in a biopsy that shows necrosis or granulation tissue. As Dr. Haggitt taught: parietal cells within a worrisome regenerative gastric epithelial process strongly suggest benignancy. Also, because subepithelial hemorrhage is an artifact frequently resulting from the trauma of taking a biopsy, such hemorrhage alone must involve a relatively large area of the mucosa in order to be suggestive of erosive, reactive gastropathy.

When present, gastric ulcers that develop on the background of reactive gastropathy are usually caused by NSAIDs and differ from those seen in patients with DAG and MIP because they are surrounded by HP-free, relatively uninflamed mucosa. In fact, any gastric ulcer surrounded by mucosa that has little or no inflammation should be considered an NSAID-associated ulcer until proven otherwise. Because the prevalence of HP infection is so high and because NSAID therapy is so widespread, it would be surprising if the two did not coexist. When this is the case, it is not possible to recognize an NSAID-induced gastric ulcer superimposed on a preexisting HP gastritis. An increasing proportion of benign gastric ulcers in the United States are probably caused by NSAIDS, whether or not there is an accompanying HP gastritis. There is controversy about whether or not HP gastritis predisposes to NSAID damage to the gastric mucosa.

The reactive gastropathy seen after gastrectomy is a special problem presumably caused by regurgitation into the gastric remnant of injurious bile containing alkaline pancreatic secretions. This “bile reflux gastropathy” is usually worse in the peristomal area. The enlarged and hyperchromatic nuclei in the hyperplastic stomal foveolae may be confused with dysplasia or even carcinoma. The stomal foveolae may penetrate into the submucosa and form dilated cystic spaces that are seen in the lumen as polyps. A misinterpretation of dysplasia or cancer may be avoided if one notes that nuclear polarity is maintained and there is no nuclear stratification. Dr. Haggitt thought that the presence of parietal cells in these atypical glands is an additional reassurance that the lesion is benign. The rest of the gastric remnant usually shows less severe reactive gastropathy than the stoma but often has severe HP gastritis; it is an inflammatory gastritis without multifocal intestinal metaplasia even after 20 or more years of postgastrectomy reflux. The fundal glands are still present and adenocarcinoma of the stomach is no more frequent than in the unoperated population of the U.S. [57] Except for reactive gastropathy, the fundal gland mucosa is completely normal in those patients without HP infection. Unfortunately, the severity of the gastropathy is not a useful predictor of which symptomatic patients with biliary reflux will improve after a surgical procedure to minimize reflux.

OTHER DIFFUSE GASTRIC DISEASES

Vascular Ectasias

Vascular ectasias of the gastric mucosa may bleed substantially. [58] They are seen more often in patients with cirrhosis or renal failure. Gastric vascular ectasias are composed of dilated mucosal capillaries some of which may contain a fibrin thrombus (Fig. 142-26). They owe their enlargement to increased arteriovenous anastomoses and other vascular abnormalities in the submucosa. [59] Gastric antral vascular ectasia (GAVE), also called watermelon stomach, is probably a related variant recognizable endoscopically as red, linear stripes on the crests of folds in the distal stomach radiating toward the pylorus; they may also show fibrin within dilated mucosal capillaries and features of reactive gastropathy; pronounced fibromuscular proliferation within the lamina propria may suggest mucosal prolapse. [60]

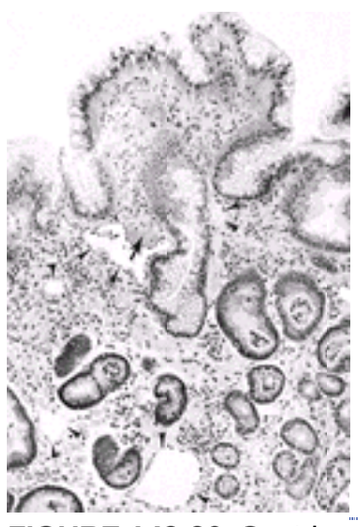

FIGURE 142-26. Gastric antral vascular ectasias (GAVE). Dilated capillaries are present in the lamina propria (*arrowheads*). In one of these, a small fibrin thrombus attached to the capillary wall can be seen (*arrow*).

Infectious Gastritis

Although gastric symptoms play a prominent role in acute gastroenteritis of presumed viral etiology, biopsies from these patients are rarely obtained because of the self-limited nature of most episodes. Other bacterial, fungal, and viral infections of the stomach occur primarily in immunocompromised patients and were described in the section “Esophageal Cancers” (see Fig. 142-14). CMV and *Candida* are the most commonly found infections diagnosed in gastric biopsies. The characteristic cytologic changes of HSV in the squamous epithelium of the esophagus are not seen in the columnar epithelium of the stomach and therefore the virus is more difficult to diagnose. The only acute gastritis caused by bacterial infection other than HP is phlegmonous gastritis. [61] It is rare and principally seen in alcoholics with a history of pharyngitis. This gastritis can be rapidly fatal. It is characterized by marked neutrophilic infiltration and edema, principally in the submucosa, but also affecting the mucosa. Phlegmonous gastritis is usually caused by *Streptococcus* and early treatment with antibiotics can be effective. The rare syphilitic gastritis [62] produces a nonspecific picture of dense plasmacytic infiltration of the mucosa with destruction of glands; if clinically suspected, spirochetes can be demonstrated by suitable stains. Tuberculosis is rare and is seen mostly in patients with AIDS and produces multiple large granulomas containing rare acid fast bacilli and, at times, caseation. Other histologically recognizable gastric pathogens in the immunosuppressed patient include *Cryptosporidium* and *Mycobacterium avium* complex.

Eosinophilic Gastroenteritis

Eosinophilic gastroenteritis is defined as an infiltration of the gastric wall or small intestinal wall with clumps of eosinophils infiltrating the epithelium of patients accompanied by a variety of GI symptoms (usually precipitated by eating) and no evidence of parasitic or extraintestinal disease. [63] An elevated peripheral blood eosinophil count is required for the diagnosis. It usually involves the stomach or small bowel but may also involve the colon or peritoneum. Eosinophils are present normally in the lamina propria of the GI tract; therefore their numbers must be excessive and they should occur in clumps before considering the diagnosis of eosinophilic gastritis. Eosinophilic infiltration is not confined to eosinophilic gastroenteritis but occurs in a wide variety of other inflammatory conditions. For this reason the diagnosis of eosinophilic gastroenteritis should not be made in the absence of an appropriate clinical history. The disease may be patchy; therefore multiple biopsies may be necessary to detect the lesion. If the patient has no GI mucosal involvement, the diagnosis cannot be made by endoscopic biopsy, as is the case with the rarer eosinophilic peritonitis.

Granulomatous Gastritis

Granulomatous inflammation of the stomach occurs in association with systemic granulomatous conditions such as tuberculosis and sarcoidosis, but is most often caused by Crohn’s disease (Fig. 142-27). It also may occur in association with neoplastic disease or with no apparent explanation in the very rare isolated granulomatous gastritis. [64]

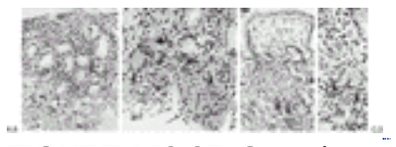

FIGURE 142-27. Granulomatous gastritis in a patient with Crohn's disease. **A:** A discrete epithelioid granuloma is seen (*arrows*) and (**B**) at higher magnification (*arrows*). This granuloma is composed of a compact aggregate of epithelioid histiocytes with a multinucleated giant cell (*arrowhead*). In the stomach such granulomas usually prove to be associated with Crohn's disease. Isolated multinucleated giant cells are present just to the left of the discrete granuloma (*arrowhead*). **C & D:** Multinucleated giant cells (*arrowheads*) can be seen in association with a few epithelioid histiocytes but no discrete granuloma. Giant cells like these in the stomach and duodenum are strongly suggestive of Crohn's disease, unlike in the colon.

A high percentage of patients with Crohn's disease of the small bowel and colon have focal, active inflammation in the stomach. [65] Only about 10% of these patients have epithelioid granulomas, and the majority are clinically asymptomatic. Isolated giant cells in the gastric and duodenal mucosa suggest Crohn's disease; in the colon giant cells are nonspecific and not diagnostic. Seemingly isolated involvement of the stomach by Crohn's disease will usually develop into Crohn's disease of more classic distribution in other organs with the passage of time.

An unusual reason for gastric granulomas is sarcoidosis; diagnosis of this disease requires evidence of a multisystem disease. As mentioned, multiple large granulomas containing necrotic centers suggest the possibility of tuberculosis and warrant a Ziehl-Neelsen stain for acid-fast bacilli.

Varioliform Gastritis

This so-called gastritis is characterized by multiple flat-topped nodules in the gastric mucosa that straddle the mucosal folds and often have an apical erosion. [66] They probably are identical to hyperplastic gastric polyps (described histologically in the section "Gastric Polyps"). This condition has been mislabeled "chronic erosive gastritis." It has been linked to lymphocytic gastritis when it became apparent that lymphocytic infiltration of the gastric surface and pit epithelium was characteristic of some of these lesions. Other patients present with large gastric folds as well as lymphocytic infiltration. The significance of this type of so-called gastritis is not clear, and additional information is needed to clarify whether it is a specific entity or several different ones.

Hypertrophic Gastropathies

Three diffuse gastric mucosal hypertrophies are recognized by their large folds at endoscopy:

- Ménétrier disease
- Zollinger-Ellison syndrome with gastrinoma
- hypertrophic hypersecretory gastropathy.

These lesions may be impossible to sample deeply enough at endoscopy for definitive histological diagnosis. The combination of the endoscopic picture with the clinical information usually makes it possible to arrive at a correct diagnosis.

At endoscopy, Ménétrier disease shows enlarged cerebriform folds covered with thick mucoid secretion especially in the fundus and body of the stomach. The essential histological lesion [67], [68] is foveolar cell hyperplasia, absent parietal and chief cells and a marked increase in mucosal thickness (Fig. 142-28). There is excess secretion of mucus that can cause hypoproteinemia. There can be cystic dilation of the gastric glands and strands of smooth muscle may extend from the muscularis mucosae into the lamina propria. The hyperplastic foveolar cells may have hyperchromatic enlarged nuclei and depletion of cytoplasmic mucus, and can be confused with dysplastic or malignant cells. The fact that adenocarcinoma can complicate Ménétrier disease makes it especially important to exclude this possibility even though it is very rare.

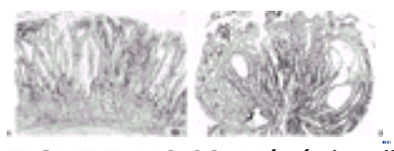

FIGURE 142-28. Ménétrier disease. **A:** A section of the gastric body taken from a resected stomach. The gastric pits or foveolae are strikingly elongated and irregular in configuration. This appearance is produced by a profound hyperplasia of the foveolar cells that extend into the glands and replace the parietal and chief cells so that the body mucosa is now composed exclusively of mucus-producing foveolar cells. **B:** An endoscopic biopsy specimen from a patient with Ménétrier disease. In addition to the marked foveolar hyperplasia that produces a corkscrew appearance of the pits, there are dilated glands, another feature of Ménétrier disease.

At endoscopy in Zollinger-Ellison syndrome with gastrinoma there are large folds and voluminous watery acidic secretion. Parietal and chief cells are proliferating rather than the foveolar cells and this accounts for the hyperchlorhydria, severe peptic ulcer disease, and steatorrhea that may be seen. [69] The presence of many parietal and chief cells rules out Ménétrier disease. Increased parietal and chief cell mass is impossible to quantitate in a biopsy specimen but requires a gastrectomy specimen. The elevation of the blood gastrin level makes the diagnosis.

The endoscopic appearance of the rare hypertrophic, hypersecretory gastropathy is very much the same as that seen in gastrinoma with excess watery acid secretion and numerous parietal and chief cells histologically. Protein loss is variable. Serum gastrin is not elevated and this differentiates this illness from a gastrinoma. These patients may well represent a severe duodenal ulcer diathesis and are probably at the high end of the acid secretory spectrum of duodenal ulcer.

GASTRIC POLYPS

Hyperplastic Polyps

The benign hyperplastic polyp used to be the most common gastric polyp (Fig. 142-29). Fundic polyps now far exceed them in the Western world. Hyperplastic polyps usually are recognized endoscopically because of their characteristic features: they are often less than 1 cm in diameter; sometimes multiple; and distributed on the crests of folds throughout the whole stomach. They may have a sessile, mesa-like shape with an erosion or healed dimple on their top. So-called varioliform gastritis may be a variant of the same lesion. When a solitary or larger hyperplastic polyp develops, it may be pedunculated. An inflammatory polyp is indistinguishable from a hyperplastic polyp except for its increased inflammation. If it is solitary with a smooth eroded surface it is often called a juvenile polyp. Histologically, foveolar hyperplasia, irregular-shaped cystic glands, and edema and inflammation of the lamina propria are typical. Pyloric glands may be present, but parietal and chief cells are infrequent or absent. The inflammatory atypia within the superficial erosion in a hyperplastic polyp may be misdiagnosed as dysplasia but attention to surface cytologic maturation should avoid this error. It is not necessary to remove all multiple small sessile hyperplastic polyps with a snare, but several should be sampled to establish their nature. Hyperplastic polyps that contain adenoma or adenocarcinoma are uncommon but how frequently this occurs is controversial. [70], [71] If a polyp enlarges with the passage of time, or is pedunculated, it should be removed completely for diagnosis.

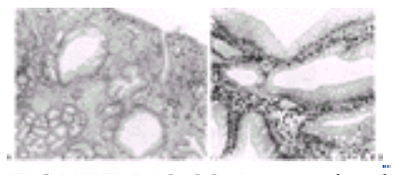

FIGURE 142-29. Hyperplastic gastric polyp. **A:** The low-power view shows the dilated, irregular-shaped gastric glands lined with hyperplastic foveolar cells. Note the inflamed, edematous stroma in which there are numerous dilated capillaries. **B:** Higher magnification shows the hyperplastic foveolar cells lining the glands composing the polyp.

Fundic Gland Polyps

Fundic gland polyps (FGPs) have become the most common gastric polyp in the Western world. They consist of small sessile mucosal elevations in the body of the stomach made up of fundal glands that contain normal parietal and chief cells. Cystically dilated glands are diagnostic; they are lined by parietal and chief cells or gastric surface mucous cells [72] (Fig. 142-30). Fundal gland polyps were first identified in association with familial adenomatous polyposis (FAP) of the colon [73] but those that occur sporadically are far more common; FGPs are benign but some may harbor surface or foveolar dysplasia that may be low or high grade; [74] how significant this is clinically is currently uncertain as gastric cancer is relatively uncommon in FAP. FGPs can disappear spontaneously. The increasing frequency of sporadic FGPs may be explained by the increased use of proton pump inhibitors. The usually small FGP can be totally excised for diagnosis with a large biopsy forceps. Those with dysplasia may require continued endoscopic biopsy surveillance and this surface dysplasia may be a marker for unsuspected FAP. [7], [73], [75]

FIGURE 142-30. Fundic gland polyp of the stomach. **A:** This endoscopic biopsy specimen from a small polyp shows dilated, irregular-shaped fundal glands. **B:** The nature of the cells lining these cystic glands can be recognized; they include parietal cells and chief cells. Although not seen in this particular field, foveolar cells may also be present.

Adenomatous Polyps

In the United States, adenomatous polyps of the stomach are uncommon, usually solitary, and most often located in the antrum. They closely resemble colonic adenomatous polyps histologically and may be tubular, villous, or tubulovillous. The mucosa surrounding gastric adenomas usually demonstrates pangastritis with intestinal metaplasia (MIP). Gastric adenomas are dysplastic precursors to intestinal-type adenocarcinoma and frequently contain a focus of early carcinoma; they may also be associated with carcinoma elsewhere in the same stomach. [7] Strangely, the adenomatous gastric polyps associated with FAP of the colon have a lower than 3% prevalence of carcinoma, unlike the FAP adenomas of the duodenum near the papilla of Vater that are frequently malignant, with a cancer prevalence higher than 12%. [76] Gastric adenomatous polyps should be excised to exclude the diagnosis of focal carcinoma and to prevent their future transformation into adenocarcinoma. Because precancerous dysplasia or early carcinoma may be present in the flat mucosa adjacent to the solitary adenoma, as well as within it, multiple biopsies of the apparently uninvolved surrounding mucosa are also indicated. If the polyp contains carcinoma, and especially if dysplasia is present in the surrounding mucosa, simple endoscopic polypectomy may not be adequate therapy. If an adenomatous polyp is confined to the antrum, as these polyps often are, antrectomy with careful frozen section evaluation of the resection margins may be required.

Polyposis Syndromes

Polyps with foveolar hyperplastic changes similar to those seen in gastric hyperplastic polyps are seen in three other intestinal polyposis syndromes which may involve the stomach: juvenile polyposis, Peutz-Jeghers syndrome, and Cronkhite-Canada syndrome.

Juvenile polyps of the stomach are indistinguishable from gastric hyperplastic polyps; they are part of a generalized *juvenile polyposis syndrome,* [77] in contrast to the *solitary juvenile polyp,* which is benign and confined characteristically to the colon. It is a hamartoma composed of various normal gastric tissues with an inflammatory infiltrate, predominant stromal elements, dilated glands, and usually no adenomatous changes, but a rare adenoma can become a carcinoma. *Peutz-Jeghers syndrome* (PJS) polyps contain an arborized pattern of muscle strands extending from the muscularis mucosae into the lamina propria (Fig. 142-31). Patients with PJS may rarely develop GI cancers, however, associated breast cancers are much more common. [78] *Cronkhite-Canada syndrome* is a rare, acquired, generalized, usually nonneoplastic GI polyposis syndrome. Histologically the polyps resemble juvenile polyps but they are not usually pedunculated; [79] the small bowel and stomach are almost always involved. The nonpolypoid mucosa may also be involved. Patients usually are in their 60s and have diarrhea, protein loss, alopecia, and characteristic skin and nail changes.

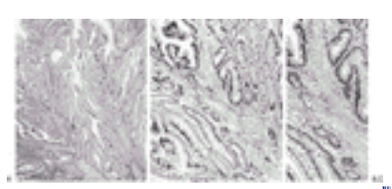

FIGURE 142-31. Small bowel hamartomatous polyp from a patient with Peutz-Jeghers syndrome. Similar polyps may be located in the stomach. **A:** The low-power view shows the complex, branched configuration of the hamartoma. **B & C:** The abnormal arrangement of normal tissue elements is seen. Normal-appearing epithelium lines tubules and covers villi. A thick band of smooth muscle divides the field diagonally. **C:** This smooth muscle band and the normal-appearing epithelium are shown at higher magnification.

Inflammatory Fibroid Polyps

Inflammatory fibroid polyps of the stomach may be pedunculated or sessile and may cause obstruction, [80] especially in the gastric antrum where they are most common and of large size. They are benign proliferations of spindle cells with localized infiltration by eosinophils without an elevated eosinophil count in the peripheral blood. Formerly they were erroneously called eosinophilic granulomas. Inflammatory fibroid polyps usually are asymptomatic until they grow to a size large enough to obstruct the gastric outlet. They are unrelated to eosinophilic gastritis.

Approach to Gastric Polyps

Short of total excision, gastric polyps may be difficult to diagnose histologically. Fortunately, gastric polyps are often pedunculated, and therefore can be easily excised for diagnosis with an electrocautery snare. Sessile lesions that cannot be excised as easily and safely with the endoscope may be diagnosable by multiple large biopsy specimens taken with a snare or by snare removal after they have been elevated from the underlying bowel wall by submucosal saline injection.

SMALL BOWEL CANCERS

Malignant small bowel tumors are the rarest seen in the GI tract (1.5%). Most of these are adenocarcinomas that are found in the duodenum and jejunum. Most have

no known precursor but others are sequelae of celiac sprue, long-term Crohn's disease, or a hereditary polyposis syndrome such as FAP or hereditary nonpolyposis colorectal cancer (HNPCC). Many small bowel carcinomas are periampullary and may be an extension of a pancreatic cancer. Rarely primary periampullary, well-circumscribed carcinomas are potentially curable. Distinguishing neoplasm from an inflammatory process histologically may be difficult in periampullary lesions. Rare lymphomas are associated with Crohn's disease. The malignancies associated with celiac sprue may be adenocarcinomas or T-cell lymphomas. Even more rarely, carcinomas may develop in ileostomies, pouches, and conduits. Primary lymphomas of the small bowel are more common in certain geographic areas including the Middle East where they were first described (see " Flat Lesions with Diagnostic Histology"). Other malignant small bowel tumors include mesenchymal tumors, carcinoids, and metastatic lesions. Duodenal carcinoids differ from other carcinoids because they frequently secrete hormones (gastrin, insulin, somatostatin). Immunocytochemical techniques performed on tissue sections can be helpful in assessing hormone production.

BENIGN SMALL BOWEL DISEASES

Duodenal Nodules

Three types of benign duodenal nodules are seen in the duodenal bulb: Brunner gland nodules, heterotopic gastric mucosa, and benign lymphoid nodules.

Brunner gland nodules are made up mostly of enlarged Brunner glands. They may also involve the mucosa and blunt or flatten overlying villi. Such nodules are frequent, may be multiple, are not adenomas, in that they are not dysplastic, and are usually of no clinical significance.

Heterotopic gastric mucosa in the duodenum is of three types:

1. Gastric surface cell metaplasia replacing some of the epithelium of the intestinal villi. This type is usually not polypoid and is most commonly a variant of normal. Extensive gastric surface cell metaplasia may be associated with excess acid production.
2. Pyloric gland metaplasia occurs as a nonspecific reaction to injury in the small bowel mucosa, particularly in Crohn's disease.
3. Nodules of heterotopic fundal glands may also be seen. They have no clinical significance.

Benign lymphoid nodules may be found in the duodenum and are often of no clinical significance, but they may be a reflection of benign lymphoid hyperplasia which is seen in common variable immunodeficiency (see " Flat Lesions with Diagnostic Histology"). In the latter entity, plasma cells that manufacture the immunoglobulins may be virtually or completely absent.

Small Bowel Adenomas

Adenomas predominate in the duodenum and are frequently periampullary, especially in FAP and HNPCC. They are similar histologically to colonic adenomas. Ideally all adenomas should be excised endoscopically because of their malignant potential. Snare removal of an adenoma of the pancreatic papilla may necessitate placement of a pancreatic stent to maintain drainage.

Other Polyposis Syndromes

Polyps seen elsewhere in the GI tract may also be seen in the small bowel (i.e., gastric-type hyperplastic polyps, adenomas, hamartomatous polyps, and inflammatory fibroid polyps). The Peutz-Jeghers hamartomatous polyps are most common in the small bowel (see Fig. 142-31).

The Proximal Duodenum

The bulb, second, and third portion of the duodenum are easily accessible to endoscopic biopsy. The normal duodenal bulb may differ histologically from the rest of the small bowel. [81] Even when normal, villi in the bulb may be distorted or absent, especially over areas of submucosal Brunner glands penetrating through the muscularis mucosae. The number of plasma cells in the normal bulbar lamina propria is often greater than in the rest of the small bowel. Flattening of villi located over Brunner glands in normal subjects can be differentiated from the abnormal flat biopsy characteristic of celiac sprue by the absence of diffuse chronic inflammation of the lamina propria and the lack of lymphocytic infiltration of the surface epithelium.

Nonspecific duodenitis of the bulb cannot be diagnosed reliably by endoscopic appearance; it requires biopsy to document the presence of polymorphonuclear leukocytes within the epithelium reflecting active injury (Fig 142-32). Nonspecific duodenitis may be a precursor or sequel to duodenal ulcer. Many of the diffuse reactions seen in the stomach are also seen in the bulb, including hemorrhagic erosive lesions, opportunistic and other infections, and eosinophilic or granulomatous inflammation.

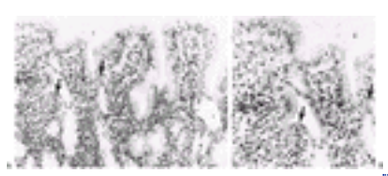

FIGURE 142-32. Active duodenitis. **A:** There are foci of neutrophilic infiltration of glands (*arrows*). Notice the gastric surface cell metaplasia involving most of the surface cell of this biopsy specimen. Two glands just to the right of center contain goblet cells and are normal intestinal glands, whereas the remainder of the glands are lined completely by cells that contain clear mucus and that represent gastric surface cells. Brunner glands that have extended into the lamina propria are seen below; such Brunner glands are also seen in normal biopsy specimens of the duodenal bulb. **B:** Higher magnification shows gastric surface cells more clearly and glands infiltrated by neutrophils, indicating active inflammation (*arrows*).

DIFFUSE PROXIMAL SMALL BOWEL LESIONS (Table 142-2)

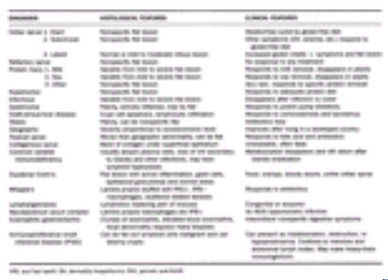

TABLE 142-2 Diffuse Proximal Small Bowel Lesions

Most disease of the small bowel is diffuse, proximal in location, and can be diagnosed by duodenal biopsy. [81] To diagnose these various kinds of disease, we strongly suggest that three or four biopsies be taken from the crest of the valvulae conniventes in the distal descending duodenal loop with a large biopsy forceps. This extra effort maximizes the diagnostic sensitivity for focal lesions and makes it easier to mount and orient the biopsies so that one can best assess villous architecture.

Distal duodenal or proximal jejunual biopsy specimens normally show tall villi in patients in the United States (Fig 142-33), but in some developing countries and in

several diseases, villi may be blunted or even completely absent (flat). The severity of villous architectural abnormality may be variable in the same patient or in different patients with the same disease. Villous abnormalities in some diseases can be diagnosed if they are accompanied by a specific diagnostic histological feature. Other diseases with only nonspecific villous abnormalities may be diagnosed by their response to a specific therapeutic regimen. Although a flat biopsy with absent villi is almost always caused by celiac sprue in the United States, there are a wide variety of much rarer diseases that may have the same histological picture, but which do not respond to a gluten-free diet. For example, the flat biopsy of severe tropical sprue may be indistinguishable from that seen in celiac sprue, but it responds to a combination of folic acid and antibiotics rather than to a gluten-free diet.

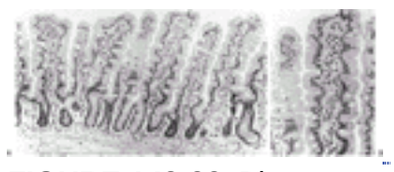

FIGURE 142-33. Biopsy specimen of normal distal duodenum. **A:** The tall, slender villi are about three times as tall as the crypts are deep. Epithelial cells covering the villi consist predominantly of absorptive cells with scattered goblet cells. The nuclei of the surface epithelial cells are lined up in a uniform, picket fence–like arrangement. The lamina propria contains small numbers of lymphocytes and plasma cells are normally seen. **B:** The normal surface epithelium and lamina propria at higher magnification. The slender, fusiform cells in the lamina propria represent smooth muscle cells normally found in the cores of small bowel villi.

Nonspecific Flat Lesions—Celiac Sprue

This disease is termed celiac sprue because in the past the same disease was traditionally called celiac disease in children and idiopathic sprue in adults. [82] There are other satisfactory names such as adult celiac disease and gluten-sensitive or gluten-induced enteropathy. We believe there are three types of celiac sprue: overt celiac sprue, subclinical celiac sprue, and latent celiac sprue.

Patients with *overt celiac sprue* have steatorrhea and it is the hallmark of serious malabsorption. There are two criteria for this diagnosis. The first is the presence of a characteristic but nonspecific flat duodenal mucosal biopsy with abortive or no villi and lymphatic infiltration of the surface epithelium (Fig. 142-34). The second criterion is a dramatic clinical response to a gluten-free diet [83] with a marked improvement in emotional status and a disappearance of steatorrhea. The diagnosis of overt celiac sprue strongly indicates the need for a diet free of wheat, barley, and rye products. This gluten-free diet is a difficult, lifelong undertaking.

FIGURE 142-34. Celiac sprue before and after treatment with a gluten-free diet. **A:** Observe the flat mucosal surface in this biopsy specimen. The villi are effaced, and the crypts have become elongated. The lamina propria contains a prominent infiltrate of chronic inflammatory cells, and lymphocytes infiltrate the surface epithelium. **B:** The appearance of this same patient's mucosa after an absolutely gluten-free period of 3 weeks, during which time the patient was fasting and on total parenteral nutrition. The villi have returned to a normal appearance and the inflammatory infiltrate within the lamina propria and surface epithelium has receded.

Patients with *subclinical celiac sprue* are free of overt malabsorption (steatorrhea). Their proximal small bowel biopsies may have a lesion of variable severity from complete loss of villi to moderate or mild villous abnormalities. The lesion may even be patchy and it is usually confined to the duodenum. The usual reason the small bowel is biopsied to reveal the lesion is the presence of one or several conditions known to be associated with celiac sprue (e.g., dermatitis herpetiformis, [84] diabetes mellitus with steatorrhea, autoimmune thyroiditis, iron deficiency anemia unresponsive to oral iron, hypocalcemia, tetany, osteoporosis, short stature, delayed onset of puberty, arthralgias, infertility, or a family history of celiac sprue). A positive serologic tissue transglutaminase test helps make the decision for biopsy. However, duodenal biopsy and response to a gluten-free diet remain the "gold standards" for diagnosis of celiac sprue.

Latent celiac sprue is very rare. [85] Such patients carry the gene for celiac sprue but the abnormal duodenal phenotype has not penetrated and they have a normal proximal small bowel. Patients with latent celiac sprue eventually may develop steatorrhea and a typical flat lesion after a large dietary gluten challenge of long enough duration. [85]

How does duodenal mucosal morphology guide treatment of celiac sprue? Patients with a flat lesion in overt celiac sprue should be treated by maintaining a lifelong gluten-free diet. Should subclinical celiac sprue with a flat lesion also be treated? In some instances treatment is clearly indicated because the associated disease process which had led to the biopsy is reversible by a sufficiently strict long-term gluten-free diet (e.g. dermatitis herpetiformis, [86] diabetic steatorrhea, and malabsorption of iron). Some clinicians treat because they believe that a gluten-free diet can prevent the dreaded complications of small intestinal lymphoma or adenocarcinoma. Others think that the mere presence of a flat lesion is a "ticking bomb" that may produce a variety of serious, preventable complications of the disease before the patient is aware of them (e.g., osteopenia). Treatment of subclinical sprue with milder duodenal abnormalities may be indicated if the patient has disabling symptoms which seem to respond to a gluten-free diet.

Another question is whether one has to test the efficacy of a gluten-free diet by repeating proximal small bowel biopsies on a gluten-free diet until they become normal. If the patient with treated overt celiac sprue is gaining weight on a diet with a normal caloric content and feels well we think rebiopsy is unnecessary and can be misleading. After institution of a gluten-free diet the mucosa normalizes gradually, beginning distally and progressing proximally. [87] Histological normalization of the proximal duodenum may be dramatic and occur in a short time but occasionally it may take months to years in some patients, and in the rare patient, may never become completely normal proximally. If the residual severe abnormality is delimited very proximally in the duodenum, the patient will be free of malabsorption. [84] The severity of malabsorption is related to the length of small bowel that has lost its normal villi. [84], [87] The reason why the lesion is not completely reversed in some patients is often obscure and one always suspects that the patient inadvertently or purposefully is not adhering to a strict gluten-free diet.

The nonspecific flat lesions of overt celiac sprue (see Fig. 142-34), refractory sprue, severe geographic abnormality (Fig. 142-35), and several other rarer conditions cannot be distinguished from one another histologically. Typically, the flat lesion shows abortive or absent villi, abnormal injured surface epithelium infiltrated by lymphocytes, and increased length of parallel, closely packed crypts with increased numbers of mitotic figures that extend from the crypts toward the surface. The lamina propria is heavily infiltrated predominantly by plasma cells, as well as lymphocytes, eosinophils, macrophages, mast cells, and a few neutrophils. Lymphocytic infiltration is most obvious in the surface epithelium but is present throughout the biopsy, especially within crypt epithelium. Marked active inflammation by neutrophils within crypts and crypt abscess formation is distinctly unusual in celiac sprue. Giant cells or epithelioid granulomas are not seen in celiac sprue and in a flat biopsy usually indicate a missed diagnosis of Crohn's disease (Fig. 142-36).

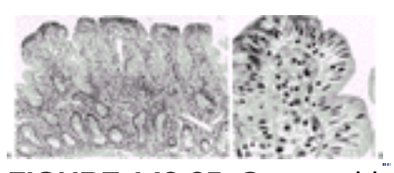

FIGURE 142-35. Geographic abnormality in small bowel morphology. **A:** The villi show a moderately severe nonspecific abnormality in architecture in this biopsy

specimen from an asymptomatic inhabitant of an extremely economically depressed country. **B:** Higher magnification. Note the blunted, shortened villus, the intraepithelial lymphocytes in the surface epithelium, and the prominent infiltrate of lymphocytes and plasma cells in the lamina propria.

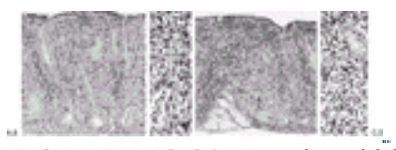

FIGURE 142-36. Duodenal biopsy specimen from a patient with Crohn's disease. **A:** Note the flat mucosal surface and markedly elongated crypts. **B:** The intense chronic inflammatory infiltrate with plasma cells and lymphocytes includes active inflammation with numerous neutrophils that have produced a crypt abscess, shown here at higher magnification. **C:** In another biopsy specimen from the same patient, a focus of granulomatous inflammation is present. A multinucleated giant cell is surrounded by macrophages and neutrophils (*arrow*). **D:** Higher-power magnification of the giant cell shown in **C** (*arrow*).

Nonspecific Flat Lesions—Apparently Unresponsive to Treatment

In *refractory sprue,* the proximal small bowel biopsy looks like untreated celiac sprue histologically, but it does not respond either histologically or clinically to exclusion of gluten from the diet, even when the patient is put on a totally parenteral regimen. Its etiology is unknown.

Some refractory patients with a flat lesion either have or develop *intestinal lymphoma* [88] or *idiopathic ulcerative ileojejunitis.* [89] The prognosis in both of these diseases is poor.

One exceedingly rare adult patient who seemed to us to have refractory sprue with a flat lesion was then found elsewhere to have injury by dietary protein other than gluten. [90] Removal of two proteins from the diet led to regression of the flat histological lesion and the restoration of health. Because the institution of an elimination diet to determine such a rare protein injury is very difficult and rarely rewarding, it should only be undertaken if the patient responds to a totally parenteral diet initially.

Other Nonspecific Flat Lesions With a Specific Therapeutic Response

There are a variety of conditions that have a variable villous abnormality, which may, when severe, be indistinguishable from the characteristic nonspecific flat lesion seen in untreated celiac sprue. *Milk protein injury* is the most common cause of the flat lesion in infants and children. *Soy bean protein* is the next most frequent protein that may be harmful to infants and children. Ingestion of soy bean protein in a susceptible patient leads to a flat lesion and its removal from the diet heals the intestinal lesion and relieves the symptoms. [91] Unlike in celiac sprue, these milk and soy proteins are no longer injurious in adult life. Diets deficient in high-quality protein can cause the disease *Kwashiorkor* with a nonspecific flat lesion, especially when the diet is otherwise adequate calorically. [92] These patients improve after addition of high quality protein to their diet even if gluten is included. In children, various *intercurrent infections* can cause a transient flat lesion. This lesion disappears when the infection recedes. The patchy abnormalities in *Zollinger-Ellison syndrome* [69] are caused by acid peptic injury secondary to excessive secretion of gastrin by a tumor. The injury in this disease is variable in severity but patchy complete loss of villi can occur in some areas of the small bowel with active inflammation and hemorrhage as well as erosions. The mucosa in these patients heals after treatment with proton pump inhibitors and they no longer have ulcers, diarrhea, or steatorrhea.

Acute *graft-versus-host disease* (GVHD) [93] can demonstrate lesions of varying severity from mild villous abnormality and focal lymphocytic and eosinophilic infiltration to complete denudation of the small bowel mucosa. A pathologist with special experience can recognize GVHD by apoptosis dropout, and lymphocytic infiltration of the crypt cells. This lesion may respond to large doses of corticosteroids and cyclosporine or tacrolimus, but severe GVHD can be a fatal disease. *Stasis syndrome* with bacterial overgrowth has a villous lesion of variable severity but it can be completely flat and indistinguishable from the lesion of celiac sprue. [94] Symptoms respond to intermittent antibiotic treatment or to relief of the obstruction if one is present.

The small bowel in inhabitants of some socioeconomically developing countries shows *geographically related abnormalities.* These are nonspecific villous abnormalities of varying severity including the flat lesion indistinguishable from that of celiac sprue with a prominent infiltrate of lymphocytes and plasma cells in the lamina propria and the surface epithelium (see Fig. 142-35). Generally, the severity of the abnormality correlates with the socioeconomic level of the developing country. The abnormality is environmental because it disappears after prolonged residence in a country with a higher socioeconomic level. Patients with *tropical sprue* in certain economically depressed countries tend to have an exaggeration of the nonspecific small intestinal abnormalities seen in their fellow inhabitants. [95] The severity of the villous abnormality varies. Such individuals may have a flat lesion indistinguishable from that of overt or subclinical celiac sprue but they do not respond to a gluten-free diet. Rather, the malabsorption and intestinal lesion respond to folate and antibiotics.

There are rare patients who appear to have *ulcerative colitis and proximal small bowel involvement, including duodenitis.* In these unusual patients, an extensive, nonspecific, flat, actively inflamed lesion of the whole small bowel was demonstrated with severe malabsorption. [96]

Flat Lesions With Diagnostic Histology

Rare cases of sprue are, or become, resistant to gluten elimination and may develop an abnormal band of subepithelial collagen (Fig. 142-37). This disease, *collagenous sprue,* [97] does not respond to a gluten-free diet or any other treatment and has a dismal prognosis. The subepithelial collagen band should be thicker than the diameter of two red cells. One must also avoid a misinterpretation of artifactual thickening of the collagen band caused by tangential sectioning. Other very helpful diagnostic features in collagenous sprue are incorporation of capillaries and inflammatory cells within the matrix of the collagen band and a spiculated interface at the junction of the collagen band with the underlying lamina propria. A number of reports of lymphocytic or collagenous colitis in association with celiac sprue have appeared, and a patient with both collagenous colitis and collagenous sprue has been described.

FIGURE 142-37. Collagenous sprue. **A:** The mucosal surface is flat, the villi are absent, and the crypts are elongated. An abnormally thick layer of collagen lies beneath the surface epithelium (*arrows*). Lymphocytes and plasma cells infiltrate the lamina propria, the collagen, and the surface epithelium. **B:** The markedly thickened subepithelial collagen table (*arrows*) can be better seen at higher power. Note the abnormal surface epithelium.

The flat lesion of *hypogammaglobulinemic sprue* can easily be confused with a nonspecific flat lesion (Fig. 142-38), except that at high-power magnification it is evident that the excess numbers of round cells in the lamina propria are mostly lymphocytes and that the usually numerous plasma cells are virtually or completely absent. [98] This is in marked contrast to the increase in the number of lamina propria plasma cells seen in most other flat lesions. The flat biopsy specimen of hypogammaglobulinemic sprue is usually infected with *Giardia lamblia* and represents one end of the spectrum of the small bowel lesions seen in *common variable immunodeficiency;* the other end is *benign lymphoid hyperplasia.* It too has virtual absence of plasma cells, but, in addition, exhibits large lymphoid nodules or polyps that may or may not have normal villi between them. The most common lesion in common variable immunodeficiency is a mixture of benign lymphoid hyperplasia and abnormal villous architecture of varying severity (Fig. 142-39A). The loss of villi in common variable immunodeficiency is caused by infection with *G lamblia,* [98] an organism easily missed in histological sections (see Fig. 142-39B, Fig. 142-39C) but well visualized in Giemsa-stained smears of mucus adherent to the biopsy specimens (see Fig. 142-39D). Pharmacological eradication of the *Giardia* cures the malabsorption in hypogammaglobulinemic sprue by restoring the villi, but the

humoral deficiency in immunoglobulin A (IgA) and other immunoglobulins remains because the plasma cells that manufacture the immunoglobulins are not restored and are still virtually absent (see Fig. 142-38).

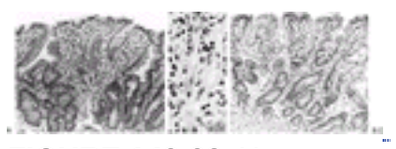

FIGURE 142-38. Hypogammaglobulinemic sprue (a variant of common variable immunodeficiency) before and after eradication of *Giardia lamblia.* **A:** This biopsy sample of the small intestine has a flat mucosal surface with marked elongation of the crypts and an inflammatory infiltrate within the lamina propria. At this magnification, it is indistinguishable from the flat biopsy specimens shown in Figure 142-34 from a patient with celiac sprue. **B:** At higher magnification inspection of the inflammatory infiltrate in the lamina propria reveals mostly lymphocytes with no plasma cells, a finding indicative of hypogammaglobulinemic sprue. *Giardia* organisms were identified in a Giemsa-stained smear made from the mucus adherent to the biopsy specimen. **C:** After treatment for giardiasis, the mucosal villi began to return, and malabsorption disappeared. Virtual absence of plasma cells from the lamina propria was not reversed by eradication of the *Giardia* organisms.

FIGURE 142-39. The typical appearance of a mixed lesion in a biopsy specimen of the small intestine in common variable immunodeficiency. **A:** The biopsy shows mild lymphoid hyperplasia (*arrows*) and an abnormality of the villous architecture. **B:** Longitudinal sections of *Giardia lamblia* are seen (*arrow*). **C:** Pear-shaped organisms (*arrow*) that have been sectioned en face in the paraffin-embedded biopsy. Intracytoplasmic detail is unclear. **D:** Giardiasis is detected most easily in Giemsa-stained smears of mucus adherent to the biopsy. The *arrow* points to a characteristic pear-shaped organism in which two nuclei are barely visible.

An unusual manifestation of *Crohn's disease* may be a flat duodenal mucosa indistinguishable from that seen in celiac sprue [99] (see Fig. 142-34). It is easily confused with celiac or refractory sprue especially if malabsorption is the main symptom and no other usual Crohn's involvement of the GI tract is found. Some indication of the unusual diagnosis of isolated duodenal Crohn's disease is the intensity of the active inflammatory response histologically with polymorphonuclear leukocytes, crypt abscesses, and destruction of glands. If present, the clinical story of fever, cramps, and bloody stools and the lack of response to a gluten-free diet favor the diagnosis of Crohn's disease. If the duodenum is biopsied repeatedly throughout its length endoscopically, epithelioid granulomas or giant cells, as well as focal areas of normal villi may eventually be detected, strongly suggesting Crohn's disease. Other rarer diseases with granulomas are sarcoidosis and tuberculosis or other unusual infections.

In *Whipple disease* [100] (Fig. 142-40), the mucosa is stuffed with macrophages that are negative for acid-fast bacilli but also contains dilated lacteals within the lamina propria. The mucosa may be flat or have residual villi. In the histologically similar disease with many macrophages with which it may be confused— *Mycobacterium avium complex* (MAC)—there are innumerable acid-fast bacilli present within macrophages but no dilated lacteals are seen (Fig. 142-41). MAC is an opportunistic AIDS infection.

FIGURE 142-40. Whipple's disease. **A:** Notice the loss of villi and heavy infiltration of the lamina propria by macrophages with abundant cytoplasm. The clear spaces represent dilated blocked lacteals filled with lipid. The surface epithelium remains essentially normal. **B:** Foamy macrophages at higher magnification are visible. By electron microscopic study, the abundant pale cytoplasm of these macrophages can be seen to contain bacilli and breakdown products of bacterial cell membranes within lysosomes. Note the completely normal surface epithelium (**upper right**).

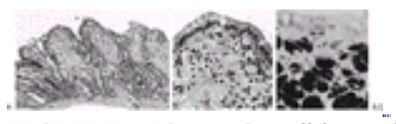

FIGURE 142-41. Small bowel biopsy specimen from a patient with acquired immunodeficiency syndrome (AIDS) who was infected with *Myobacterium avium* complex. **A:** Note the shortened, broad villi and the lamina propria packed full of macrophages. **B:** Macrophages within the lamina propria are seen at higher magnification. With a hematoxylin and eosin stain, this appearance is indistinguishable from that of Whipple's disease except for the absence of dilated lymphatics. **C:** Ziehl-Neelsen stain identifies the acid-fast nature of the bacilli within the macrophages and in the lamina propria.

Lymphangiectasia is usually a congenital disease but it may be secondary to various infiltrating mesenteric lesions. The biopsy varies from normal to flat because of multiple dilated lymphatics distorting the villous architecture (Fig. 142-42).

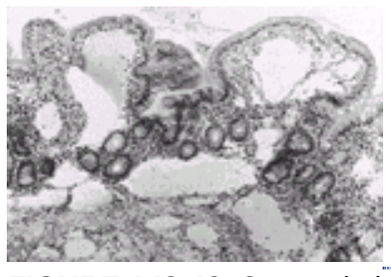

FIGURE 142-42. Congenital lymphangiectasia. The villous architecture in this small bowel biopsy specimen is distorted by dilated lymphatic channels that extend through the muscularis mucosae into the submucosa at the bottom of the photograph. Apart from the dilated lymphatics and distorted architecture, the mucosa is normal.

Eosinophilic gastroenteritis [101] /SUP>of the small bowel is a patchy and rare disease. The small bowel and distal stomach are the most common areas of involvement. There may be large areas of normal villi; thus it often requires multiple biopsies for detection. When the mucosa is free of disease and only the deeper layers of the small bowel are involved it will be impossible to diagnose by peroral biopsy. There are clumps of mucosal eosinophils in the involved areas and an elevated eosinophil count in the peripheral blood. This is a diagnosis of exclusion because collagen-vascular

disease, parasitic infection, and drug and other allergic reactions must be excluded. Also a variable number of eosinophils are present normally in the lamina propria or accompanying any type of inflammation.

Rarely, an *autoimmune enteropathy* can flatten the intestinal mucosa; in this entity, lymphocytes may be seen in the process of destroying crypts and villi; goblet cells may be absent and circulating antigoblet cell antibodies may be demonstrable. Associated thymoma has been reported.

Microvillus inclusion disease [102] is a rare hereditary enteropathy, present from birth onward and compatible with only a short life. Histologically it may resemble the flat celiac sprue lesion but electron microscopy reveals microvilli buried within the cytoplasm of the absorptive cells.

Primary intestinal lymphoma [103] is associated with malabsorption (Fig. 142-43) and is typically a marginal zone lymphoma. It is seen mostly in patients from developing countries and usually is confined to the intestine and its lymph nodes. When the villi are destroyed by this type of lymphoma, the biopsy may be confused at low power magnification with the nondiagnostic flat lesion of celiac sprue. Two features that should prevent this error are the replacement of many of the crypts with the lymphoid infiltrate within the lamina propria, suggesting an invasive process, and the malignant cytologic appearance of the lymphoid infiltrate [103] (see Fig. 142-43). Because in some patients the infiltrating cells may synthesize abnormal heavy chain immunoglobulin, it has been called immunoproliferative small intestinal disease (IPSID). [104] This disease may have a prelymphomatous stage in which there is diffuse infiltration of the mucosa by plasma cells without morphologically malignant lymphoid cells.

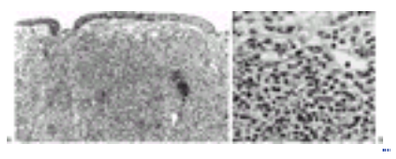

FIGURE 142-43. Primary small intestinal lymphoma (immunoproliferative small intestinal disease). **A:** The absence of crypts is noteworthy and probably reflects the destruction of the glandular architecture by the diffuse infiltrate of small lymphocytes. **B:** At higher magnification, the monotonous, atypical appearance of the lymphoid infiltrate can be readily seen.

Diagnostic Histological Features Despite Normal Villi

There are a few diseases with normal villous architecture that have other diagnostic histological abnormalities. *Amyloidosis* may be suspected when amorphous eosinophilic material is seen in the lamina propria within, or surrounding, vessel walls. This diagnosis is confirmed by staining with Congo red, which causes dichroic birefringence in polarized light (a green and reddish orange color).

The various *storage diseases* have a predilection for ganglion cells, macrophages, or endothelial cells; these cells usually are vacuolated and require special histochemical examination for diagnosis.

If the absence of plasma cells is not recognized, the small bowel biopsy in a patient with *X-linked immunodeficiency* and common variable immunodeficiency can easily be missed because the villi may be normal and it may not be appreciated that the round cells in the lamina propria are lymphocytes rather than plasma cells.

Small Bowel Infections

At least six potentially fatal opportunistic infections with specific histological features are seen in the small bowel of immunosuppressed patients: strongyloidiasis, cryptosporidiosis (Fig. 142-44), candidiasis, CMV, MAC (see Fig. 142-41), and *Isospora belli.* Other infections that may be seen in the duodenum of both immunologically competent and immunosuppressed patients include giardiasis, histoplasmosis, and cryptosporidiosis.

FIGURE 142-44. Cryptosporidiosis in an immunosuppressed individual. It can be seen in both the small and large bowel. **A:** The low-power view illustrates the shortened, blunted villi, the elongated crypts, and an inflammatory process in the mucosa. **B:** Infiltration of the lamina propria and surface epithelium by lymphocytes is evident. Cryptosporidia are barely visible at this magnification (*arrowheads*). **C:** The organisms are best seen in the high-power magnification. They appear as small, round, gray dots adherent to the apical surface of the absorptive cells. Cryptosporidia also may produce disease in immunocompetent persons, but the process is acute and self-limited rather than chronic, as it may be in acquired immunodeficiency syndrome (AIDS).

Other Rare Diseases

Other rare lesions with specific histological features include the abetalipoproteinemia with normal villi and fat-filled, vacuolated, fasting absorptive cells; *chronic granulomatous disease* with normal villi and typical clumps of vacuolated pigmented macrophages in the lamina propria; *Waldenström macroglobulinemia* with variably abnormal villi containing eosinophilic deposits of IgM-specific macroglobulin in the lamina propria; and *severe vitamin B_{12} or folate deficiency,* with its reversible epithelial macrocytosis and mild abnormalities of villous architecture.

TERMINAL ILEAL DISEASE

As experience with biopsy of the terminal ileum has accumulated, diagnostic usefulness has proven somewhat disappointing. It is of limited value in the diagnosis of Crohn's disease; although this disease frequently involves the terminal ileum, surprisingly, diagnostic epithelioid granulomas in this location are very unusual. In Crohn's disease ileal biopsy has proven less accurate than endoscopic, clinical, or surgical impressions. Ulcerative colitis, involving the entire colon, may be accompanied by "backwash" ileitis that can be impossible to differentiate from Crohn's disease. Also, one must avoid a diagnosis of chronic inflammation prompted by over-interpretation of the normal lymphoid aggregates and normal Peyer patches.

Tuberculosis may be diagnosed by histological demonstration of the organisms in granulomas with the Ziehl-Neelsen stain. *Yersinia* infection may be suspected because of its characteristic necrotizing lesions of the gut lymphoid tissue (Fig. 142-45), and PCR detection of this organism in paraffin tissue blocks of the small bowel has been described. [105] Partially obstructive *NSAID-induced diaphragms* of the ileum have been described; [106] characteristically, they are covered with ileal mucosa that has been eroded or ulcerated over the narrowed edge of the diaphragm, where there may also be crypt architectural distortion and active inflammation. Underlying the inflamed mucosa, there often is a shelf of fibromuscular and neural proliferation; transmural lymphoid aggregates that simulate Crohn's disease are also common. Seronegative arthritis may be accompanied by an acute or chronic inflammatory terminal ileal lesion and may resemble, or be associated with, Crohn's disease. [107]

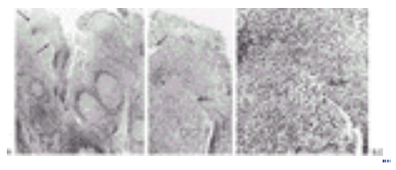

FIGURE 142-45. Yersiniosis of terminal ileum. **A:** The low-power view shows a Peyer patch in which there is erosion of the surface epithelium with adherent exudate (*arrows*). **B:** This erosion is shown at higher power. The margins of the eroded surface epithelium are indicated by the *arrows.* The inflammatory exudate extends into the underlying lymphoid tissue, in which there is a focus of necrosis (*arrowhead*). **C:** The focal necrosis (*arrowhead*) is better seen at higher power.

COLONIC TUMORS, BENIGN AND MALIGNANT

The diagnosis of benign and malignant tumors of the colon and the recognition of precancerous dysplastic lesions depend on endoscopic biopsy. A polyp is any projection from the colonic mucosal surface and is called an adenoma when composed of dysplastic tissue. In contrast, the dysplasia and early carcinoma in ulcerative colitis may not form a grossly visible lesion and is usually found in flat mucosa. The occasional adenocarcinoma seen in various polyposis syndromes also develops from dysplasia. Total excision of polypoid lesions for histological examination is the most reliable method of diagnosis and usually of treatment.

Colonic Polyps

There are four common kinds of colonic mucosal polyps:

- benign hyperplastic polyps
- adenomas with or without adenocarcinoma
- hamartomatous polyps
- inflammatory polyps. [108]

Hyperplastic polyps are the most common nonneoplastic colonic polyp. Only adenomas are neoplastic.

Hyperplastic polyps have no recognized relationship to adenocarcinoma except in the rare (0.3% incidence) *hereditary hyperplastic polyposis* (HHP). The usual hyperplastic polyp is not a marker of an increased probability of adenoma in the more proximal colon except in HHP. There are more than 20 hyperplastic polyps in the distal colon or rectum in HHP. Benign hyperplastic polyps usually are less than 0.5 cm in diameter and are often multiple. Only histological examination can differentiate hyperplastic polyps with certainty from adenomas. The frondlike, serrated, nonneoplastic epithelial surface of colonic hyperplastic polyps is diagnostic (Fig. 142-46). They are considered hyperplastic because they have an increased number of epithelial cells per unit length; the excess number of epithelial cells, squeezed into a shorter distance, buckles and causes serrated glandular profiles. Deeper in crypts, their epithelium may be crowded, depleted of mucus, and confused with an adenoma, especially in tangentially cut sections. The histological appearance of a colonic hyperplastic polyp is different from that of a gastric hyperplastic polyp. Occasionally colonic adenomas may grow in a hyperplastic polyp-like pattern; but their epithelium appears neoplastic and the significance of such lesions is the same as that for any other adenoma.

FIGURE 142-46. Hyperplastic colonic polyp. **A:** This section passes through the junction of normal mucosa and the hyperplastic polyp (*arrow*). The lumen of the normal crypts has a straight appearance whereas the crypt lumina within a hyperplastic polyp has a serrated or stellate profile. **B:** Inspection of the higher power magnification shows that this serrated appearance is caused by crowding of excess numbers of normal, nonneoplastic epithelial cells, causing them to form tufts that project into the lumen and from the lumenal surface.

The most common type of neoplastic polyp is the *adenoma.* Carefully done autopsy studies in which the entire surface of the colon is inspected with a hand lens have shown that the frequency of adenomas is around 50% for males and 40% for females in the United States. [109] Most endoscopic series that report colorectal adenomas, however, find 20% to 30% of individuals have them.

An adenoma is a grossly visible mass of dysplastic epithelium. Dysplasia is defined as neoplastic epithelium (an autonomous new growth of epithelium) that is still confined within the basement membrane of the crypt within which it arose. Because dysplastic epithelium is still confined within its basement membrane, it *cannot* metastasize. Note that all adenomas are, by definition, composed of dysplastic epithelium. Thus, it is redundant to say that “an adenoma has dysplasia.” Some pathologists grade the degree of dysplasia within an adenoma but we do not, nor did Dr. Haggitt, because grading has no prognostic significance regarding future development of additional adenomas or carcinomas.

There are three histological types of adenomas: most common are the tubular adenomas with branched tubules (Fig. 142-47); least common are the villous adenomas that are typically larger and sessile, made up of fingerlike villi and more likely to contain a focus of carcinoma invading the submucosa (Fig. 142-48). Of intermediate frequency are the tubulovillous adenomas that are a mixture of the two types. Histological typing of adenomas is not predictive of future adenomas or carcinomas, but a diameter of greater than 1 cm or an increase in the numbers of polyps is indicative of these. Thus, we see no point in grading the dysplasia of an adenoma or in determining its histological type. Excision is the appropriate treatment for all adenomas.

FIGURE 142-47. Colonic tubular adenoma. **A:** Note the characteristic tubular architecture. **B & C:** Cytologic features of the neoplastic cells lining the tubules. Nuclear enlargement, crowding and stratification, and focal diminution in mucus production are seen. These changes indicate the dysplasia seen in all adenomas.

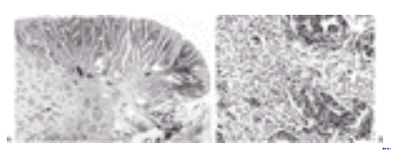

FIGURE 142-48. Electrocautery snare biopsy specimen of a sessile colonic adenoma with invasive carcinoma. **A:** Normal mucosa distorted by electrocautery artifact is shown (**lower right**). The

upper portion of the tissue is composed of villous adenoma, and (**lower left**) an invasive adenocarcinoma infiltrates below the muscularis mucosae into the submucosa. **B:** High-power view of the area marked with an *arrow* in **A** shows the prominent desmoplastic stromal reaction elicited by the invading carcinoma.

"Carcinoma in situ" in an adenoma is a term that should be abandoned. It is defined as cytologically malignant neoplastic epithelium confined within the basement membrane. Note that this definition is synonymous with HGD and thus "carcinoma in situ" *cannot* metastasize. Using the word "carcinoma" for a lesion that is not biologically malignant (because it cannot metastasize) is a great potential source of confusion. It could precipitate an unwarranted laparotomy with segmental resection or impair the patient's insurability. For this reason, most GI pathologists recommend not using the term "cancer" for such dysplastic changes. Rather, adenoma is accurate and sufficient.

"Intramucosal carcinoma" in an adenoma is defined as neoplastic epithelium that has invaded through the basement membrane into the lamina propria, but not through the muscularis mucosae. This is a rare event in the colon, but in the esophagus and stomach, invasion of the lamina propria is a meaningful diagnosis because it is associated with a low, but definite risk of nodal metastasis (approximately 3%). Nodal metastasis from carcinoma confined to the lamina propria in the colon *has never been reported.* Thus, colonic intramucosal carcinoma is *not* a biologically malignant lesion and the World Health Organization has correctly recommended abandoning this term for intramucosal colonic neoplasms. Our approach is not to look for the rare colonic adenoma with intramucosal invasion because it has no more significance than an adenoma alone and both lesions are cured by complete colonoscopic excision.

Adenocarcinoma in an adenoma is neoplastic epithelium that has invaded through the muscularis mucosae *into* the submucosa. Invasion into the submucosa is virtually always associated with a surrounding desmoplastic stromal reaction, which serves as a diagnostic hallmark for invasion and connotes a definite risk of nodal metastasis (see Fig. 142-48).

There are various logical steps in management of an endoscopically resected adenoma containing carcinoma: The first and most important step is to confirm that the lesion is invasive carcinoma. This is not as easy as it seems because "pseudocarcinomatous" invasion, or misplaced epithelium, is a fairly common finding. However, the desmoplastic stromal reaction mentioned previously as being such a characteristic feature of invasive carcinoma is the single most useful criterion to make this distinction (see Fig. 142-48). If the worrisome misplaced glands are surrounded by normal lamina propria, they are not invasive. Also helpful are the rounded appearance of the glands on low power in benign, misplaced epithelium versus the irregular, angulated and infiltrative appearance of invasive adenocarcinoma. Acellular mucin pools within the submucosa are of no differential diagnostic help because they may be associated with both benign misplaced and malignant invasive processes. After determining that the lesion is truly invasive, the pathologist then evaluates the depth of the invasion within the polypectomy specimen. A histological grade is then assigned and the lesion is scrutinized for evidence of lymphatic or blood vessel invasion. The completeness of resection is then evaluated by noting the status of the margin and finally, an estimate of the risk of metastasis can be provided based upon the foregoing information.

Nodal metastasis from a carcinoma in which the invasion extends into the submucosa of the head of a pedunculated polyp or at any level into its stalk is extremely rare, certainly less than 1%. The probability of having positive nodes from cancer arising in a pedunculated polyp becomes significant only with invasion into the level of the submucosa of the bowel wall proper.[110] Sessile adenomas containing invasive carcinoma are less likely to be cured by endoscopic excision because these cancers are more likely to extend into the submucosa of the underlying bowel. The data suggest that the overall risk of nodal metastasis is about 5% when invasion is limited to the underlying submucosa. Poorly differentiated tumors are more likely to have positive lymph nodes than well or moderately differentiated ones. The exact percentage of such cases with nodal metastasis is difficult to determine with certainty because of the small numbers of cases that are poorly differentiated and because of the frequent presence of confounding factors such as lymphatic invasion. If invasion of lymphatic channels is present, there is a high probability of positive nodes, but the exact percentage of such patients with positive nodes is difficult to determine because of small numbers of cases and the presence of confounding factors such as a poorly differentiated tumor. In fact, lymphatic invasion is unusual unless the tumor is poorly differentiated. The presence of carcinoma at the polypectomy margin, if the stalk was transected near its base, implies that invasion into the submucosa of the underlying bowel wall may be present and additional evaluation of the patient to determine whether there is residual tumor left or not needs to be carried out.

Thus, unless the patient has invasion of the tumor into the submucosa of the underlying bowel wall, a poorly differentiated tumor, or lymphatic invasion, resectional surgery appears not to be indicated because of the low risk of nodal metastasis that could potentially be cured by a resection. However, even if positive nodes are resected surgically, the 5-year survival for this Dukes C colon cancer is only approximately 40%. In fact, the 5% risk of nodal metastasis is low enough with invasion into the submucosa that an argument can be made for not proceeding with a surgical resection.

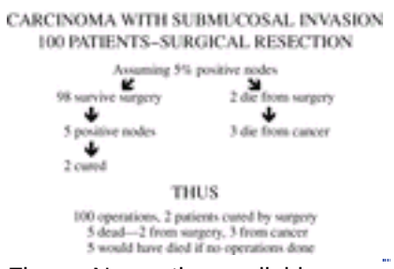

Figure. No caption available.

The status of the polypectomy margin, as an index of prognosis, is also controversial. Morson and colleagues[111] found that if the polypectomy margin was free of the tumor, regardless of the distance from the neoplasm to the electrocautery margin, the prognosis was uniformly favorable. When the carcinoma extended to the electrocautery margin, but when the endoscopist thought that the lesion was completely resected, the prognosis was again favorable. This probably reflects the destruction of an additional zone of 0.2 to 0.3 cm of tissue by the electrocautery during excision. In contrast, when the endoscopist thought that the excision was incomplete or questionable, the prognosis was less favorable. Others have suggested that extension of the carcinoma to within 0.2 cm or less of the margin is an unfavorable prognostic sign. Use of this criterion as an indication for colectomy produces a favorable outcome because the probability of nodal metastasis is distinctly low in this subset of individuals. In Dr. Rodger Haggitt's opinion, if one used extension of the tumor to within 0.2 cm or less of the margin as a criterion for surgical resection, one increased the rate of surgery unacceptably. Thus, careful communication with the endoscopist concerning his or her impression of the completeness of excision is a must in planning the optimal management for a patient with carcinoma arising in an adenoma.

The adenomas of FAP of the colon look the same as those of sporadic adenoma initially, but in the classic form of FAP they are multiple (=100), and in virtually all patients, they progress eventually to cancer if the colon is not removed prophylactically.[112] Patients with FAP have an APC gene mutation. *Hyperplastic polyposis* (HPP) is a far more rare disease than FAP; it is defined by the presence of 20 distal hyperplastic polyps.[113] HPP is often associated with adenomas, some of which contain cancer but there is no family history of colonic carcinoma. Carcinomas in HPP are found throughout the colon. Obviously, these patients need close colonoscopic surveillance. Whether they will need prophylactic colectomy as in FAP remains to be determined. *Hereditary nonpolyposis colorectal cancer* (HNPCC) is caused by a defect in mismatch repair genes. In certain kindreds there may be multiple organ cancers associated with the syndrome. The cancers in HNPCC arise in one or several adenomas rather than in multiple polyps.[113] More than 60% of the cancers are in the right colon and the tumors are both synchronous and metachronous. The colonic pathology is characteristic but not specific for HNPCC: the tumor is diploid, poorly differentiated, mucin producing, or of the signet cell type. There often are dense lymphoid infiltrates and a Crohn's like reaction. Endoscopic surveillance should begin early (5 years prior to the age of the earliest colon cancer in the family) because of the early onset of cancer and the suspicion that some of these tumors progress much more rapidly than sporadic carcinomas. Prophylactic colectomy is also a possible choice for prevention. Surveillance for extracolonic cancers in HNPCC should start at 25 to 30 years of age (uterus, stomach, ovary, etc.).

The hamartomas are an abnormal mixture of benign cells that only rarely contain cancer, and then only arising in dysplastic epithelium or adenomas. The most common hamartoma is the single pedunculated juvenile polyp in a child under 10 years of age (Fig. 142-49). It is less common in older children and adults. *Familial juvenile polyposis*[77] is hereditary and is seen in patients older than 10 years of age, may have multiple polyps involving the whole GI tract, and is associated with an increased risk of carcinoma. The juvenile polyposis of infancy may be fatal but fortunately is rare. The polyps of *Peutz-Jeghers syndrome* (PJS) are more common in the small bowel than in the colon and the polyps of Cowden disease are rare. Histologically, juvenile and Cowden polyps are more cystic and Peutz-Jeghers polyps contain highly ramified branching smooth muscle with a large benign epithelial component (see Fig. 142-31). All are genetic disorders. PJS may have other associated

malignancies, particularly breast cancer. [78]

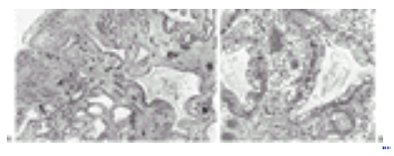

FIGURE 142-49. Juvenile polyp. **A:** The lesion is composed of dilated, irregular-shaped glands (**B**) lined by epithelium of an essentially normal appearance. The lamina propria is inflamed and edematous and contains numerous congested vessels.

Inflammatory or postinflammatory polyps, also incorrectly known as pseudopolyps, are common in areas of healed or partially healed ulcerative colitis or Crohn's colitis and may be found after ischemic or infectious colitis. These polyps often represent islands of variably inflamed residual mucosa in a "sea" of surrounding, previously sloughed, healed mucosa. Inflammatory polyps may have a filiform, sessile, or pedunculated configuration.

Colonic Carcinoma

Most sporadic colonic carcinoma originates in adenomas. Grossly, the lesser the depth of invasion and the absence of nodal metastasis, the better the prognosis (Dukes classification).

Extensive ulcerative colitis of more than 10 years duration predisposes to carcinoma. The coexistence of primary sclerosing cholangitis is an additional risk factor. [114] Such patients should be in an endoscopic biopsy surveillance program to detect dysplasia in nonpolypoid mucosa as an indication for prophylactic colectomy. Histologically, the degree of abnormality (atypia) within dysplastic epithelium forms a continuous spectrum from slight to marked. For clinical purposes, dysplasia is divided into low-grade and high-grade, based upon its degree of deviation from normal colonic epithelium. [6] The problems with dysplasia as a marker of cancer risk are: sampling error, observer variation in diagnosis, and differentiation from inflammatory reactive changes. Dysplasia may be localized to a single, small area of the mucosa, it may be multifocal, or it may be extensive and involve virtually the entire mucosa. We found that 33 biopsy samples are required to achieve 90% confidence that dysplasia would be detected if it were present. [115] We, therefore, recommend at least four biopsy specimens from each 10-cm segment of the colonic mucosa and every 5-cm segment in the rectum, based on the higher incidence of distal carcinoma in ulcerative colitis (i.e., at least one half of colorectal cancers arising in ulcerative colitis occur in the rectum and sigmoid colon). The biopsies from each segment are placed in the same bottle of fixative because we may wish to resample the same colonic area if the findings are equivocal. Thus, in a patient with a 70-cm colon, 32 biopsies would be taken and 8 blocks would be made.

Active inflammation in ulcerative colitis induces reactive and regenerative epithelial changes that can be difficult, and at times, impossible, to distinguish from dysplasia (see Fig. 142-2). For this reason, most experienced GI pathologists are reluctant to diagnose dysplasia in actively inflamed mucosa unless the abnormalities are overt. Reactive epithelium may have crowded, stratified, enlarged, hyperchromatic nuclei and abnormal mitotic figures, but the degree of nuclear atypism typically diminishes toward the lumenal surface—atypism extending onto the lumenal surface suggests dysplasia. In order to recognize abnormal epithelium involving the surface, well-oriented biopsy specimens are necessary. Abundant, eosinophilic cytoplasm is also characteristic of reactive or regenerative epithelial cells. When there is doubt about the significance of epithelial abnormalities, they should be designated as indefinite for dysplasia. If active inflammation is present, posttreatment biopsies may show a dramatic reduction in the intensity of inflammation and resolution of epithelial abnormalities, but dysplasia will be unaffected.

Flow cytometric analysis of DNA content of epithelial nuclei provides objective information about the ploidy status and cell cycle events in populations of cells. Suitable cells/nuclei for analysis can be derived readily from "jumbo" endoscopic forceps biopsies. Such biopsies measure approximately 4 to 5 mm in diameter and can be divided in half, with one half being used for flow cytometric analysis and one half for a histological control. We found aneuploid cells in 22 of 25 patients with dysplasia in ulcerative colitis, while 52 of 53 patients whose biopsies were negative for dysplasia contained only diploid populations of cells. Interestingly, 6 of 13 patients whose biopsies were indefinite for dysplasia contained aneuploid cell populations, and 5 of these patients progressed to develop dysplasia and multiple aneuploidies over relatively short follow-up. [115] The presence of multiple aneuploid populations of cells appears to be highly associated with carcinoma and may indicate high risk for progressing to carcinoma.

Other Colonic Tumors

Lipomas, fibromas, leiomyomas, and Kaposi sarcoma in AIDS are usually submucosal in location and are thus diagnosed less frequently by biopsy. Angiomas can be sampled by hot biopsy but there is some risk of bleeding. Benign, enlarged lymphoid nodules can be mistaken for neoplasms or polyposis; these nodules may be idiopathic or a reaction to infection, or they may be the benign lymphoid hyperplasia of common variable immunodeficiency.

Occasionally tumors metastatic to the colon may be recognizable as such if they retain the characteristic morphologic features of their primary site.

Carcinoids almost always are histologically invasive and have the characteristic appearance of monotonously similar cells with small round nuclei growing in cords, glands, or nests. Many colonic carcinoids that are smaller than 2 cm are indolent and can be resected endoscopically. If they recur or enlarge, segmental resection may be required.

INFLAMMATORY DISORDERS OF THE COLON (Table 142-3)

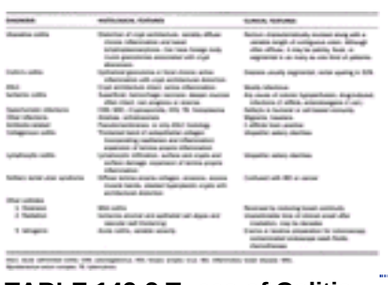

TABLE 142-3 Types of Colitis

An important function of biopsy is to establish the presence and type of colitis in various possible inflammatory disorders of the colon. It is important to emphasize that an isolated increase in the numbers of lymphocytes, plasma cells, and macrophages in the lamina propria is not by itself diagnostic of colitis. There is a wide normal variation in round cell content of the lamina propria. Numerous isolated eosinophils may be seen in the normal mucosa, especially in the right colon, in children, [116] or in patients from the southern latitudes, even within the U.S. [117] The diagnosis of colitis requires alterations in the epithelial cells or in the crypt architecture in addition to an increase in inflammatory cells in the lamina propria. These changes include infiltration of surface, or crypt epithelium by neutrophils, eosinophils, and lymphocytes; decreased mucus secretion (goblet cell depletion); distortion of crypt architecture and reactive nuclear changes, such as enlargement, vesicular chromatin, and prominent nucleoli.

Three causes of bloody diarrhea that may be confused with one another are idiopathic ulcerative colitis (Fig. 142-50), Crohn's disease (Fig. 142-51), and acute self-limited colitis (Fig. 142-52). The term idiopathic inflammatory bowel disease (IIBD) refers to both ulcerative colitis and Crohn's disease. These are both chronic diseases in contrast to acute self-limited colitis which is often of known infectious origin and is both relatively acute and self-limited. The most common reason to biopsy the colon is to help differentiate between these three diseases because their prognoses and responses

to treatment are very different.

FIGURE 142-50. Ulcerative colitis. **A:** The biopsy specimen shows disease in which the inflammatory infiltrate varies in intensity and the distortion of the crypt architecture is mild; the crypt seen in the center of the biopsy sample is branched and contains neutrophils infiltrating its epithelium and lumen. **B:** A biopsy specimen from a patient with severe disease with diffuse inflammation, distortion of the crypt architecture, and basal lymphoplasmacytosis. **C:** Atrophic mucosa in a patient with inactive ulcerative colitis; there is distortion of the crypt architecture but no inflammation.

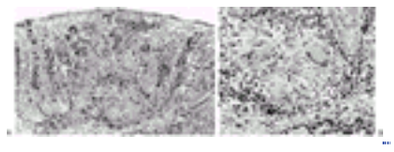

FIGURE 142-51. Crohn's disease of the colon. **A:** This biopsy specimen contains a discrete epithelioid granuloma (*arrows*) composed of multinucleated giant cells and epithelioid histiocytes. **B:** View at higher magnification power. Although inflammation surrounds the granuloma and involves some of the lamina propria, the mucosa at the left side of **A** is essentially normal, emphasizing that Crohn's colitis is commonly focal in distribution.

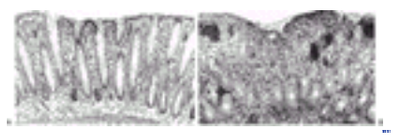

FIGURE 142-52. Acute self-limited colitis in a patient with salmonellosis. **A:** Normal mucosa for comparison with (**B**) the histological appearance of the mucosa in acute self-limited colitis. In acute self-limited colitis, there is no distortion of crypt architecture, and this differentiates it from idiopathic inflammatory bowel disease. The crypts remain as straight, evenly spaced tubules, but there is a mixed inflammatory infiltrate in the lamina propria in which neutrophils predominate over round cells. Accumulation of neutrophils within the crypt lumen to form crypt abscesses is evident, but this is a nonspecific finding seen in all kinds of colitis, as is the absence of mucus secretion in the actively inflamed mucosa.

While both ulcerative colitis and Crohn's disease undoubtedly are at some point in time truly "acute," at the time of onset of the bloody diarrhea there are virtually always histological features of chronicity, even when the duration of symptoms is quite short. [118] From a diagnostic standpoint, the presence of these features of chronicity is often more important than the acute neutrophilic inflammatory component since, in an acute inflammatory background, evidence of chronicity suggests an active IIBD (see Fig. 142-2) rather than an infectious acute self-limited colitis (see Fig. 142-52). In endoscopically normal or near-normal segments of bowel, the presence of histologically detected chronic IIBD changes (see Fig. 142-50) can be crucial to the distinction between ulcerative colitis and Crohn's disease, and thus these apparently "normal" areas must also be biopsied. Histologically a completely normal segment between abnormal segments is more suggestive of segmental Crohn's disease, whereas the presence of residual distortion of crypt architecture in endoscopically normal-appearing areas is in favor of ulcerative colitis which is more often a diffuse process.

The following features suggest chronicity (see Fig. 142-50): crypt architectural distortion, Paneth cell metaplasia (distal to the right colon), and basal lymphoplasmacytosis. Irregularities of crypt shape include branching, foreshortening, and loss of parallelism. Abnormal branching must be differentiated from normal innominate grooves. In the former, two branches of the crypt join to empty at the surface; in the latter, all crypts open independently on the surface of the innominate groove. An occasional branched gland in an otherwise normal mucosa is of no significance, as is branching adjacent to a lymphoid follicle or in proximity to the anus. Foreshortening refers to glands that fail to extend down to a level seen in normal mucosa (i.e., to nearly touch the muscularis mucosae). Loss of parallelism refers to the lack of a "rack of test tubes" architecture seen in optimally oriented normal colonic mucosa.

Marked variation in size and irregular spacing of glands are sometimes best appreciated on portions of specimens tangentially oriented parallel to the surface. Small cross-sectional glands immediately adjacent to mid- and large-size glands suggest crypt architectural distortion.

Crypt architectural distortion may accompany "diversion" colitis following mechanical diverting of the fecal stream and in "pouchitis" within an ileal pouch following ileoanal-pouch anastomosis after total proctocolectomy. Crypt distortion may also be seen in rare infections that linger for a prolonged period of time such as amoebic colitis, occasional cases of *Campylobacter jejuni, Salmonella, Shigella,* and in radiation (Fig. 142-53) or chemotherapeutic damage or chronic ischemia. A segmental colitis mimicking ulcerative colitis but limited to the sigmoid colon may occur in patients with sigmoid diverticular disease. [119] Thus, the pathologist should indicate that the changes likely represent an IIBD, but if the clinical context is not completely known, a "consistent with" modifier is appropriate. Finally, patients with IIBD are at increased risk of infection with the usual bacterial and viral pathogens. These are usually diagnosed clinically through appropriate stool pathogen cultures. One pathogen that may only be detected morphologically is CMV and it should be sought in all IIBD biopsies, particularly those with marked inflammatory activity.

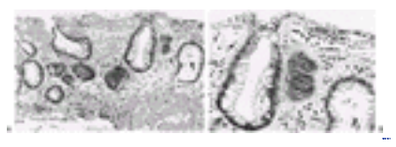

FIGURE 142-53. Colitis after therapeutic radiation. **A:** After radiation the chronic changes in the mucosa are atrophic and may resemble inactive ulcerative colitis. **B:** Fibrosis of the lamina propria and hyalinization of vascular walls are shown (*arrows*). These ectatic vessels with hyalinized walls serve to distinguish radiation colitis from inactive ulcerative colitis. The surface epithelium in this biopsy sample is eroded; this may be an artifact because it is not a consistent finding in radiation colitis. Acute radiation changes, not shown here, may be indistinguishable from ischemia.

Ulcerative Colitis

In the past ulcerative colitis was believed to virtually always affect the colonic mucosa in a diffuse and circumferential manner. The disease usually involves the rectum in continuity with a variable length of colon, and when the entire large bowel is inflamed, the appendix and terminal ileum may be affected as well. There are now numerous reported exceptions to the conventional wisdom regarding the diffuse nature of the disease in ulcerative colitis. In up to one third of cases, patients with well-documented ulcerative colitis may have patchy or focal segmental inflammation. [120], [121] (see Fig. 142-50A) This is usually seen in patients whose disease is or has at one time been in remission.

The pathological findings are essentially restricted to the mucosa and upper submucosa unless there is extensive ulceration, as in fulminant colitis, in which case the inflammation may extend through the bowel wall. Paradoxically, large and deep ulcers are not typical of ulcerative colitis except when the disease is fulminant. A pronounced chronic inflammatory infiltrate consisting of many plasma cells, lymphocytes, and variable numbers of macrophages and eosinophils occupies the lamina propria and may form aggregates near the base of the mucosa (basal lymphoplasmacytosis) (see Fig. 142-50B). In contrast, the neutrophils in active ulcerative colitis tend to be localized to crypt epithelium ("cryptitis") or lumen ("crypt abscess") or marginating in capillaries; they are not found in large numbers in the lamina propria unless crypts are perforated. Numerous eosinophils, however, may be found within the lamina propria or epithelium in IIBD. In active ulcerative colitis, the goblet cell mucin is usually markedly diminished or depleted, even in crypts that are not infiltrated by neutrophils. The crypt epithelium may be hyperplastic with large, hyperchromatic, crowded and stratified nuclei that may closely resemble those seen in dysplastic epithelium, except that the nuclear changes generally mature toward the surface (see Fig. 142-2).

During the resolution phase of active inflammation in ulcerative colitis, the inflammatory infiltrate within the lamina propria may resolve in an irregular manner so that the biopsy shows what appears to be a focal inflammatory process. [120], [121] With quiescence, the neutrophilic infiltrate disappears and the intensity of chronic inflammation gradually diminishes. Biopsies taken during the inactive stage may show a characteristic appearance referred to as "atrophy" and are characterized by reduced numbers of crypts with distorted architectures best recognized by branching (see Fig. 142-50C), or they may be normal, as previously mentioned. The base of the crypt may be separated from the underlying muscularis mucosae by a long distance, and the muscularis mucosae itself may be quite thickened and its bundles of muscle separated from each other by fibrosis. The picture of "atrophic" mucosa is highly characteristic of ulcerative colitis and is seldom seen in Crohn's disease. A closely similar picture may occur in healed ischemia and radiation (see Fig. 142-53) or chemotherapy-induced colitis, so that clinical correlation is important.

Frequently there is a lack of correlation between the endoscopic appearance of the mucosa and its histological appearance. For this reason, the endoscopist should be encouraged to biopsy endoscopically normal-appearing mucosa to document that it is, in fact, normal. In children with ulcerative colitis, focal inflammation or rectal sparing may be present at the initial diagnostic evaluation. [122] Patients presenting with fulminant ulcerative colitis may also have gross and histological rectal sparing. In cases with atypical distributions, particular attention should be given to *all* of the patient's biopsies, especially those from early on in their disease, as time or treatment may influence disease distribution in ulcerative colitis.

Crohn's Disease

Crohn's disease may affect any part of the GI tract from the mouth to the anus. The small bowel alone is affected in about 30% of cases and the small and large bowel together in about 55%. [123] The colon is said to be affected by itself in 15% of cases, but this is probably overestimated. In the most common pattern of involvement, the disease affects the terminal ileum and proximal colon together. Inflammatory lesions of the anus are particularly characteristic of Crohn's disease and may occur even in the absence of inflammation of the colon. In contrast to ulcerative colitis, which it may mimic clinically, Crohn's disease usually affects the colon and the rest of the bowel in a discontinuous manner (see Fig. 142-51). In about half of patients who have colonic involvement by Crohn's disease, the rectum is spared both endoscopically and histologically. Large, linear or serpiginous ulcers surrounded by normal-appearing mucosa are characteristic, as are small, punctate erosions surrounded by a zone of hyperemia (aphthae).

True epithelioid granulomas are the most dependable criterion for the diagnosis of Crohn's disease (see Fig. 142-51). Typically, they are relatively small, compact aggregates of epithelioid histiocytes that contain variable numbers of giant cells. Not infrequently, giant cells cannot be identified. In some granulomas, the epithelioid histiocytes do not form compact nodules but rather "loose" aggregates. Such loose granulomas are more difficult to recognize and are more often related to foreign material, such as mucus, than are compact granulomas. The reported frequency with which granulomas can be identified in biopsies varies markedly between various studies. Possible explanations for this marked variation include the definition of granulomas used, whether or not isolated giant cells are mislabeled granulomas, the number of biopsies obtained, and the number of sections examined. Serial sectioning will significantly increase the yield of granulomas. [124], [125] Microgranulomas are quite small, consisting only of a few histiocytes, and can be easily overlooked. The cytoplasm of epithelioid histiocytes has a typical ground-glass appearance revealed on fine focusing at high power; this appearance reflects the excess cytoplasmic smooth endoplasmic reticulum (E.R.) in epithelioid histiocytes revealed at the higher magnifications of electron microscopy. One must be cautious about what is diagnosed as an epithelioid granuloma in colorectal biopsies. Ruptured crypts release mucin into the lamina propria, and this commonly induces a granulomatous reaction. [126] Such mucin granulomas usually include mature macrophages and foreign body–type giant cells. They can be recognized because of their predominance of giant cells and their orientation about perforated crypts. Mucin granulomas are nonspecific, and they may be seen in Crohn's disease, ulcerative colitis, infections, diverticulitis, and adenomas, among other conditions. Isolated giant cells in the lamina propria occur in similar settings and should likewise not be considered as evidence of granulomatous inflammation in the colon; if located in the basal portion of the mucosa, they are characteristic of IIBD but may be seen in both ulcerative colitis and Crohn's disease. Biopsies from patients with Crohn's disease may also demonstrate aphthous lesions. In the absence of another known cause of aphthous lesions (e.g., *Yersinia enterocolitica*), their presence is very suggestive of the diagnosis of Crohn's disease.

Aphthous-like lesions may also be seen endoscopically in patients who have received sodium phosphates orally or by enema. Histologically, these are prominent lymphoid follicles with reactive changes in the adjacent mucosa. These do not indicate colitis and are a bowel preparation artifact.

Normal areas of colonic mucosa alternating with focal areas of inflammation favors Crohn's disease over ulcerative colitis but ulcerative colitis can become patchy over time, especially with remission and with new immunosuppressive therapies. A variant of untreated ulcerative colitis that must not be confused with Crohn's disease is the "cecal patch" of colitis in which the cecum is discontinuously involved. [121] Occasionally, clinically typical Crohn's disease shows diffuse inflammation that is indistinguishable from ulcerative colitis; unless typical epithelioid granulomas are present, this colitis must be classified as IIBD of indeterminate type. The latter is a diagnosis that we make infrequently. Correlation of clinical activity with biopsy appearance in Crohn's disease is even less dependable than that seen in ulcerative colitis and the typical atrophic appearance of quiescent ulcerative colitis is not often seen in Crohn's disease.

The pathology of postoperative ileoanal pouches in ulcerative colitis overlaps substantially with that of Crohn's disease, and Crohn's like complications are also seen. This does not mean that the original diagnosis of ulcerative colitis was incorrect.

Acute Self-Limited Colitis

Colorectal mucosal biopsy plays an important role in the management of patients with the acute onset of often bloody diarrhea. The endoscopic appearance of acute self-limited colitis is indistinguishable from that of ulcerative colitis. In marked contrast to the histological appearance of acute self-limited colitis (see Fig. 142-52), when a patient with the initial symptoms of IIBD is seen for the first time, even if symptoms have only been present for a few days, the characteristic distortion of crypt architecture and marked, chronic inflammation of the lamina propria are present in colorectal biopsies (see Fig. 142-2). [118], [127], [128] The absence of these key features signifies that the disease process will probably pursue an acute, self-limited course and is, therefore, most likely infectious. [118], [128] In the characteristic picture of acute self-limited colitis crypt architecture is well preserved, and polymorphonuclear leukocytes predominate in a lamina propria that lacks the prominent chronic inflammatory infiltration of IIBD. [118], [128] As in ulcerative colitis, goblet cell mucin may be diminished or absent, neutrophilic infiltration of the crypt epithelium (cryptitis) may be present, and crypt abscesses may be seen. Crypt abscesses associated with a prominent infiltrate of macrophages, producing the appearance of granulomatous inflammation, suggest *C jejuni* infection or *Salmonella.* Pseudomembranes are most commonly seen with *Clostridium difficile*-induced colitis, but are nonspecific and may be seen in other types of colitis, including *E coli* 0157:H7. [129], [130] The latter may produce an appearance that is indistinguishable from ischemic colitis (Fig. 142-54). The changes include, in addition to the other characteristic features of acute self-limited colitis, mucosal hemorrhage, capillary thrombosis, and necrosis.

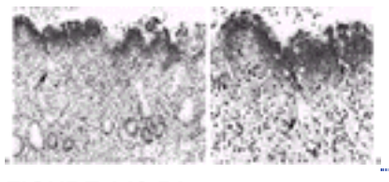

FIGURE 142-54. Ischemic colitis. **A:** The upper part of the mucosa is necrotic whereas the deeper portion remains viable. The *dark band* along the surface of the mucosa represents hemorrhage. The *arrow* points to a crypt basement membrane devoid of epithelial cells because they have become necrotic and sloughed off (crypt ghost). Notice the relatively mild inflammatory infiltrate. **B:** The eroded surface epithelium, superficial hemorrhage, and crypt ghosts (*arrow*) are seen at higher power.

In about half of patients who have acute self-limited colitis that is presumably infectious, the stool cultures and examination for ova and parasites are negative. Thus, the biopsy is quite helpful in determining whether the patient with acute bloody diarrhea has a self-limited process or chronic IIBD. In the general population of the United States, the most common enteric pathogens cultured in the acute self-limited colitides are *C jejuni, Salmonella* species, *E coli* 0157:H7, and *Shigella* species. Cultures are essential for planning appropriate treatment. When none of these pathogens is identified, the causative organism may be an invasive *E coli* that cannot be distinguished from nonpathogenic *E coli* without tissue culture assays or DNA probing.

Ischemic Colitis

Ischemic colitis from vascular hypoperfusion can have a wide variety of causes. [131], [132] Clinically its diagnosis can be difficult and biopsy often is required. In patients with acute ischemic injury, the bowel wall may be friable, but biopsy specimens can be taken safely if done carefully. Black or dusky bowel which may be infarcted should not be biopsied.

The histological hallmarks of ischemic colitis include necrosis of the upper part of the mucosa with sloughing of epithelial cells from the surface and upper regions of the crypts, while the epithelium in the deeper part of the mucosa may remain intact (see Fig. 142-54). A superficial zone of hemorrhage corresponds approximately to the region of necrosis of epithelial cells. Pericryptal basement membranes from which the epithelium has sloughed are called crypt ghosts. Capillary thrombi are prominent. Inflammatory cells, including mostly neutrophils but with a few mononuclear cells, are present but in relatively small numbers unless infection supervenes. The submucosal edema typical of ischemic colitis may produce a bleb that is occasionally removed like a polyp and submitted for histological examination. Such blebs are responsible for the “thumbprints” seen on roentgenograms. In patients with transitory mucosal ischemia, regeneration rapidly occurs and produces changes in the epithelial cells that can be misinterpreted as dysplasia or carcinoma. Ischemic colitis most commonly occurs in the absence of an occlusive vascular lesion. [133]

Patients who develop ischemic lesions of the intestine without vascular occlusion do so because of hypoperfusion of the mesenteric vasculature. [132], [134] The hypoperfusion can be due to a wide variety of causes, but the most common are cardiac failure or arrhythmias, digitalis toxicity, shock, and septicemia. [133] The physiology of the mesenteric circulation is such that a countercurrent exchange mechanism reduces oxygen content in the upper portion of the mucosa while it remains normal in the lower. [135] Consequently, the upper mucosa, which is relatively hypoxic under normal conditions, is particularly vulnerable to the effects of hypoperfusion and may become necrotic while the lower mucosa remains intact.

Ischemic lesions are more common in the colon than in the small bowel, probably because of the vast collateral network in the small bowel. Ischemic colitis may follow three courses depending upon the duration of the ischemic episode and the extent and depth of the lesion. If the ischemia is transitory, only superficial necrosis is produced, and spontaneous healing with no sequelae may occur. Following more prolonged ischemic episodes, the necrosis extends deeper into the bowel wall so that the muscularis propria may be affected. Recovery is usually accompanied by prominent submucosal fibrosis with stricture formation. After prolonged acute ischemia, necrosis of the full thickness of the bowel wall develops, and perforation, peritonitis, and sepsis result in a high mortality rate. Morphologically, one may see all stages at the same time in different biopsies, with necrosis restricted to the superficial mucosa, various depths of mucosal destruction, and ulcers.

Ischemic colitis developing in a younger person or in a patient without apparent predisposing factors, such as cardiac failure or arrhythmia, should suggest a vascular occlusive lesion, such as arteritis, intravascular coagulation, atheroembolism, amyloidosis, volvulus, or thromboangiitis obliterans. [136], [137] Several examples of ischemic colitis that were apparently induced by cocaine have been reported. [138] The mechanism of this ischemia is probably related to the potentiating action of cocaine on norepinephrine. Venous stenosis due to a spectrum of lesions ranging from various types of phlebitis to myointimal hyperplasia may also occur in young people. [139], [140] These mesenteric venoocclusive lesions are of unknown etiology and appear to have a favorable prognosis after resection. Ischemia in young persons taking NSAIDs has been described, as has ischemic colitis in young women taking oral contraceptive agents. In some young patients with ischemic colitis, no apparent cause for the decreased blood flow is ever found. A group of other colitides that probably arise on an ischemic basis and which morphologically resemble ischemic colitis includes: *E coli* 0157:H7, some forms of pseudomembranous colitis, uremic colitis, radiation colitis, and colitis that accompanies some obstructive lesions. [134], [141], [142] and [143]

Infectious Colitides

Sexually transmitted rectal infections include gonorrhea, herpes simplex, chlamydia, syphilis, amebiasis, and the various other enteric pathogenic bacteria. [144] The persisting opportunistic infections in AIDS each have characteristic organisms: Cryptosporidia which may involve the small and large bowel (see Fig. 142-44), MAC, and rotavirus in children. CMV, HSV, tubercle bacilli (Fig. 142-55), and *Histoplasma* species (Fig. 142-56) also are seen. Schistosomal colitis (Fig. 142-56) is an important and extremely common problem in the rest of the world but is only occasionally seen in foreigners or international travelers in the United States. It is diagnosed by finding the characteristic eggs, often within epithelioid granulomas. A section through the edge of a granuloma may miss the egg and be confused with Crohn’s colitis, but inspection of serial sections can overcome this problem. Amebic colitis (Fig. 142-57) in nonendemic areas of the U.S. is seen mostly in travelers, migratory workers, and homosexual men. When amebic colitis is suspected, special care must be taken while obtaining and handling the biopsy specimen to avoid removing adherent mucus containing the amebae. Do not orient such specimens; just drop them into fixative and let the technician attempt orientation after fixation.

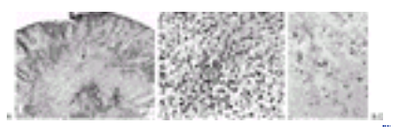

FIGURE 142-55. Colonic tuberculosis in a patient with acquired immunodeficiency syndrome (AIDS). **A:** The biopsy specimen shows inflammation of the colonic mucosa with multiple, large, confluent granulomas. The *arrow* points to a focus of necrosis within a granuloma. **B:** This shows the same biopsy specimen as in panel **A,** at higher power. Necrosis within granulomas should always suggest an infectious agent. **C:** Ziehl-Neelsen stain in which acid-fast bacilli of *Myobacterium tuberculosis* are evident. The tissue response to *Myobacterium avium* complex differs in that granulomas and diffuse granulomatous inflammation with discrete aggregates of epithelioid histiocytes are not seen; rather, there is diffuse infiltration of macrophages with foamy cytoplasm filled with acid-fast bacilli (see Fig. 142-41).

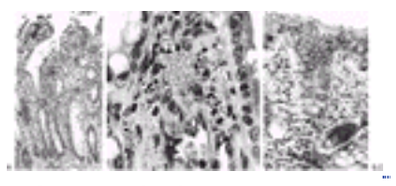

FIGURE 142-56. A: Colonic histoplasmosis in a patient with disseminated histoplasmosis in acquired immunodeficiency syndrome (AIDS). Distorted glandular architecture and an inflammatory infiltrate in the lamina propria are shown. **B:** Inspection of the inflamed lamina propria at higher power reveals a foamy or bubbly appearance caused by numerous *Histoplasma* organisms within the cytoplasm of macrophages. The organisms are seen as clear vacuoles, many of which contain a dark central dot representing the nucleus. **C:** Biopsy specimen from a patient with schistosomiasis. The ovum is clearly identified in the lamina propria and can be diagnosed as a schistosome, but speciation is not possible except in the rare circumstance in which the plane of section passes through a lateral spine of *Schistosoma mansoni.*

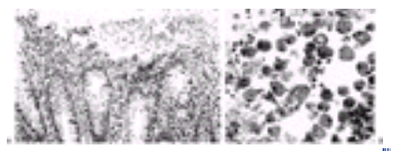

FIGURE 142-57. Amebic colitis. **A:** The mucosa shows diffuse inflammation of the lamina propria and an eroded surface. The material adherent to the surface includes erythrocytes and numerous amebic organisms. **B:** These are seen at high power. The organisms are large cells, measuring approximately 30 µm in diameter. They have a relatively small nucleus compared with the cytoplasmic volume. Phagocytized red blood cells are visible as black round structures within several of the amebae.

The most common antibiotic-related colitis is caused by overgrowth of *C difficile,* which secretes specific toxins. Although colitis caused by *C difficile* usually is called pseudomembranous colitis (Fig. 142-58), only 50% of patients have pseudomembranes at the time of biopsy and pseudomembranes are also seen in other colitides. With or without pseudomembranes, the histological pictures may be indistinguishable from acute self-limited colitis.

FIGURE 142-58. Pseudomembranous colitis caused by *Clostridium difficile.* **A:** This biopsy specimen shows features of an acute self-limited colitis with active inflammation of the lamina propria, erosion of the surface epithelium, and adherent fibrinopurulent exudate (pseudomembrane), but preservation of the crypt architecture. Most of the inflammatory cells are neutrophils. **B:** The erosion of the surface epithelium, the pseudomembrane, and the predominantly neutrophilic infiltration are well illustrated at high-power magnification. This picture has features also seen in ischemic colitis.

Collagenous Colitis

In collagenous colitis there is troublesome, often persistent watery diarrhea, for as long as 15 years, frequently in older women with a normal colonoscopy. [145] On biopsy there is a thickened collagen plate (=15 microns, the diameter of two red cells) located beneath the surface epithelium but not around the crypts (Fig. 142-59). It has a spiculated and irregular appearance due to an incorporation of capillaries and inflammatory cells within the collagen and due to projections of collagen into the underlying lamina propria. This produces a striking contrast to the normal basement membrane which has a uniform straight lower border. These qualitative changes in the collagen plate may be as important as the absolute thickness of the subepithelial collagen in establishing the diagnosis of collagenous colitis. [146] The collagen deposition may be diffuse or patchy and tends to be least prominent in the distal sigmoid and rectum. The superficial lamina propria may be expanded by increased numbers of round cells. Occasionally the rectum is not involved and colonoscopy with more proximal sampling may be necessary for diagnosis. The crypt architecture is almost always well preserved and about half of patients have a focal, mild neutrophilic infiltration of the crypt epithelium which tends to be observed early in the course of disease rather than later. If the neutrophils are prominent, bacterial infection must be ruled out. Lymphocytosis of the surface and crypt epithelium may be present and surface cell damage may be prominent. Collagenous colitis does not have enough inflammatory change or architectural distortion to resemble IIBD. Patchy collagenous thickening however can occasionally be seen in IIBD, ischemic colitis, and other conditions. Rarely amyloid may mimic collagenous colitis.

FIGURE 142-59. Collagenous colitis. **A:** This biopsy specimen shows normal glandular architecture, a possible increase in the number of plasma cells in the upper portion of the lamina propria, and a markedly thickened subepithelial collagen plate. The surface epithelium is abnormal and infiltrated by lymphocytes. **B:** These changes are seen better at higher power. Observe the lacy or reticulated appearance of the thickened subepithelial collagen table, which incorporates inflammatory cells and capillaries.

Lymphocytic Colitis

This is an entity in which patients have diarrhea and other symptoms and a normal appearance on colonoscopy. [147] It is remarkably similar symptomatically to collagenous colitis. However histologically there is no excessive collagen but rather a prominent lymphocytic infiltration of the surface and crypt epithelium with expansion of the lymphocytic content of the lamina propria. Like collagenous colitis, lymphocytic colitis is idiopathic and has preservation of the crypt architecture, so the histological picture is not likely to be confused with IIBD.

The term “microscopic” colitis [148] is applied to both collagenous and lymphocytic colitis as well as to patients with chronic diarrhea of unknown origin in general who have no endoscopic abnormalities. These patients may have mild inflammation on biopsy and are a heterogenous group that has only chronic diarrhea, normal endoscopy, and mild histological inflammation in common. Other causes are various drug-induced injuries. Thus, the term does not refer to a specific disease entity and should be replaced by more specific terminology whenever possible.

Patients with chronic, watery diarrhea and histological findings similar to lymphocytic colitis have been described in an epidemic of diarrhea traceable to a point source. [149] Thus, the histological features of lymphocytic and collagenous colitis seen in patients with chronic, watery diarrhea may reflect reaction patterns to a variety of injuries, rather than specific disease entities.

Other Colitides

There are a variety of other colitides. Diversion colitis usually is a mild colitis that occurs when the colon is excluded from the fecal stream. [150] Restoration of the fecal stream to the diverted colon reverses the process. Radiation colitis has an acute mucosal phase resembling acute ischemia and a more chronic mesenchymal phase with atypical fibroblasts, scarring, and telangiectatic capillaries that may have hyalinized walls. [151] It is an unpredictable process that may become clinically manifest at any time from months to years after irradiation.

Finally, a number of colitides are iatrogenic. Endoscopes may be inadequately rinsed after use of sterilizing solutions or wash water may be contaminated. For example, a microvesicular form of pneumatosis intestinalis can be caused by the accumulation of gas bubbles after inadvertent exposure to hydrogen peroxide (Fig. 142-60A) or by insufflation of gas into microtears in the mucosa during endoscopy. [152] Less dramatic are the changes produced by various methods of cleaning the colon in preparation for colonoscopy. Many cleansing enemas can cause active surface inflammation and edema. [153] A bisacodyl enema may produce a confusing picture that can easily be considered colitic (see Fig. 142-60B, Fig. 142-60C). [154] Gastrografin used as an enema instead of barium is hypertonic and highly irritating.

FIGURE 142-60. Pseudolipomatosis and other artifacts. **A:** Pseudolipomatosis of the lamina propria after inadvertent exposure to hydrogen peroxide left after cleaning the wash channel of the colonoscope. The bubbles in the lamina propria represent gas bubbles caused by the release of nascent oxygen when hydrogen peroxide contacted the mucosa. These bubbles can be differentiated from fat cells because they lack a cell membrane and vary markedly in size. **B:** The mucosa in a patient who has received a bisacodyl enema. The surface epithelium is flattened and contains focal infiltrates of neutrophils (*arrow*). **C:** Focal neutrophilic infiltration of the abnormal surface epithelium at higher power (*arrow*).

A variety of chemotherapeutic agents can cause extensive mucosal injury. This is especially well documented with 5-fluorouracil. [155] Colitis associated with bone marrow transplantation is complex; [93] the early lesion is caused by the preparatory irradiation and chemotherapy and the later lesions represent graft-versus-host reaction or opportunistic infections.

Focal active inflammation is not uncommonly identified in colonic biopsies. Surface apoptosis may also be prominent. The clinical significance of these findings is hard to assess and should probably not be referred to as colitis. These changes may very well be related to bowel preparation.

Solitary Rectal Ulcer Syndrome

Solitary rectal ulcer syndrome [156] (SRUS) is also known by other, perhaps better, terms such as mucosal prolapse and localized colitis cystica profunda. It is a distinct clinical entity and has a characteristic but nonspecific histological appearance (Fig. 142-61). The term is a misnomer because the lesions may be multiple, they may involve the sigmoid as well as the rectum, and usually have erosions rather than ulcers or may lack both. This entity may be confused clinically with the ulcers of Crohn's disease or with ulcerative carcinoma because of the hard lumpy tissue palpable around the ulcer. The thickened tissue may be filled with diffuse collagenous granulation tissue, [157] excess muscle bundles, and lengthened crypts with epithelial hyperplasia.

FIGURE 142-61. Solitary rectal ulcer syndrome. **A:** The biopsy specimen shows erosion of the surface epithelium, obliteration of the lamina propria by fibrosis, granulation tissue and smooth muscle, and gross distortion of the glandular architecture. Hyperplasia of the glandular epithelium also is seen. **B:** At higher power the eroded surface epithelium and obliteration of the lamina propria by collagenous granulation tissue and fibroblasts is better seen. Observe the epithelial hyperplasia that occasionally results in the misinterpretation of these lesions as adenomas or even cancer.

In 10% to 15% of patients, cystic glands penetrate into the submucosa to form so-called colitis cystica profunda; [156] these ectopic glands are benign and have no desmoplastic stromal reaction surrounding them to suggest invasive malignancy. [158] Histologically, there may be gross distortion of crypt architecture and erosion of the surface epithelium with adherent exudate producing a "pseudomembrane." The muscularis mucosae is markedly hypertrophic and disorganized and bundles of smooth muscle and collagen extend high into the lamina propria around the crypts. The epithelium may appear quite hyperplastic and atypical, but it matures as it approaches the surface where the cytologic features of neoplasia are not present.

Other Diagnoses

Other diagnoses by biopsy include melanosis coli, a benign collection of lipofuscin-containing macrophages caused by laxatives, and pneumatosis cystoides intestinalis, marked by gross polyps on colonoscopy that contain gas-filled spaces in the submucosa lined by giant and epithelioid cells. [152]

COST EFFECTIVENESS

Taking a sufficient number of adequately sized biopsy specimens and orienting and processing them in the manner suggested is more costly than routine biopsy. Currently, less comfortable, large-diameter operating endoscopes must be used to take large biopsies; but smaller-diameter endoscopes with larger biopsy channels will be generally available and should remedy this problem. An extra assistant is helpful to orient the specimens correctly without prolonging the procedure but the endoscopy may take longer if more specimens are needed for optimal diagnosis. Step-serial sectioning and special staining also require some extra effort and expense.

Is this compulsive approach worth the effort? We believe that it is for ulcerative colitis [115] and Barrett esophagus surveillance. [7] In the long run our approach may not be more costly. [159] This is because the surveillance interval may be extended in patients who are negative for dysplasia (the majority) because of the high level of diagnostic confidence our approach achieves. In our two teaching hospitals our approach has been self-supporting and has not required subsidization. Those who have been exposed to both approaches firmly believe that the more compulsive technique detects more focal lesions, may be more accurate in the diagnosis of dysplasia and early carcinoma, permits better evaluation of abnormalities of mucosal architecture, and is easier to read.

An incorrect diagnosis of cancer followed by an operation is costly, especially if complications or death occur. Similarly, it is difficult to estimate the cost and distress of the patients and their families that could be avoided if an early, curable cancer had been diagnosed in time. The same considerations apply to more accurate biopsy diagnosis of certain other diseases. The problem is that there may be no way to know which patients need this extra effort at the time of endoscopic biopsy. Ideally all biopsy specimens should be taken and processed as suggested to increase diagnostic accuracy and improve patient care.

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CHAPTER 143

Sugantha Govindarajan and Maurizio Bonacini

LIVER BIOPSY AND HISTOPATHOLOGICAL DIAGNOSIS

This chapter describes the morphologic changes of various disease processes as one attempts to interpret the needle biopsy of the liver. Attention is drawn to the most important changes one should be aware of while analyzing a liver biopsy to arrive at a diagnosis and differential diagnoses. Sections in other chapters deal with more detailed analysis of these disease processes.

The first step in liver biopsy interpretation is recognition of the lobular architecture differentiating normal from fibrosis, cirrhosis, and noncirrhotic nodular regeneration. Systematic analysis of the hepatic lobule starts with the examination of the portal tract, followed by terminal hepatic veins, and the assessment of the parenchyma and the sinusoidal spaces. Based on this analysis, the following differentiations are made: acute versus chronic; parenchymal versus biliary disease; list of possible etiologic factors; if neoplastic, benign versus malignant; and primary versus metastatic.

TECHNIQUE OF LIVER BIOPSY

The most common method of liver biopsy is transcutaneous (transthoracic or subcostal) with or without ultrasound or computed tomography (CT) guidance. [1,2] Occasionally, biopsies are taken via the transjugular route (useful in patients with severe coagulopathy) or during surgery. [2] Needles used for liver sampling rely either on suction (Menghini, Klatskin, and Jamshidi) or cutting technique (Tru-cut and Vim Silverman). [2] The latter technique is applied with a spring load mechanism (Quick-core or similar) that allows a specific length of tissue, usually 15 to 20 mm, to be retrieved with a minimum of needle manipulation in the liver parenchyma. [3] In cirrhotic livers, cutting techniques result in less fragmented specimens than the suction technique. Most percutaneous liver biopsies result in the retrieval of sufficient liver tissue for diagnosis after a single pass, but more than one pass is required in 4% to 12% of patients. [1,4] The use of real-time imaging (ultrasound more cost-effective than CT) or ultrasound X-marking of the entry site prior to biopsy has significantly decreased the percentage of patients who experience pain requiring analgesia (50%–35%) [1,2,4] as well as the chance of hospitalization. [1] A decrease in mortality has not been shown, [1,4] but the sample size required to show a difference would be exceedingly large.

Patients with coagulopathy (platelets < 60,000 or PT > 4 seconds above normal) can be biopsied transcutaneously after infusion of platelets and fresh frozen plasma, or via the transjugular route. [2] Patients with inherited bleeding disorders such as hemophiliacs can be biopsied transcutaneously, provided they are infused with the appropriate coagulation factors and are followed by experienced hematologists during the peribiopsy period. [5]

Complications of percutaneous liver biopsy that lead to hospitalization are rare and include severe pain (2%), hemoperitoneum (0.04%), hemobilia (0.01%), and sepsis (0.01%). [6,7] Hemo-, pneumo-, or hydrothorax was reported in 0.07% of cases. [6] Perforations of other organs such as gallbladder, lung, colon, kidney, or adrenal gland have all been described. [1,4,6] The vast majority of complications occur within the first 3 hours after biopsy, so a 4- to 6-hour monitoring period is usually carried out, with a friend or family member to take charge of the patient for 24 hours. [7] Death, reported in 1 per 10,000 cases, is always due to massive hemoperitoneum, and most often associated with cirrhosis or malignancy. [6]

The risk of bleeding appears to increase with the number of passes, so only two are customarily allowed. There is little difference in the complication rates that occur with trainees versus attendings [7] and with suction, cutting, or spring-load techniques. [1,4] The role of a lower needle gauge (14G vs. 16 or 18G) or the use of “plugging” devices in the chance of complications is unsettled. [2] As mentioned, the transjugular route is used by interventional radiologists in the most difficult cases, often with advanced liver diseases or bleeding diathesis. [2] Postprocedure complications appear to occur more often than with percutaneous biopsy.

PROCESSING OF LIVER BIOPSY TISSUES

Fixation and processing technique vary from laboratory to laboratory; however, the most commonly used fixative is 10% buffered formalin. For crisp nuclear details and rapid fixation, we use B5 solution containing 6 g of mercuric chloride, 2.074 g of hydrated sodium acetate dissolved in distilled water and mixed with 10 mL of 40% formaldehyde (pH 5.8) at time of use. Embedding in plastic resins such as araldite improves the ability to obtain thin (2–3 μm) sections with greater cytologic details, such as cellular mitochondria or microvesicular fat over paraffin embedding. [8] Besides regular hematoxylin and eosin (H & E) stains, the most useful routine stains are Masson trichrome for collagen (Fig. 143-1; see also Color Fig. 143-1), periodic acid-Schiff (PAS) for glucose, diastase-digested PAS for glycoprotein (i.e., a $_1$-antitrypsin) (Fig. 143-2; see also Color Fig. 143-2), iron stain (Fig. 143-3; see also Color Fig. 143-3), Shikata orcein stain for hepatitis B surface antigen (HB $_S$Ag) (Fig. 143-4), and copper binding proteins (Fig. 143-5), and reticulin stain for identification of cord sinusoidal structures. Other special stains helpful in diagnosis are those for acid-fast and fungal organisms in cases of granulomata, phosphotungstic acid-hematoxylin for fibrin in disseminated intravascular coagulation or necrosis in cases of Q fever, rubeanic acid for copper, and immunoperoxidase stains for viral proteins, a $_1$-antitrypsin, a-fetoprotein, factor VIII, and others.

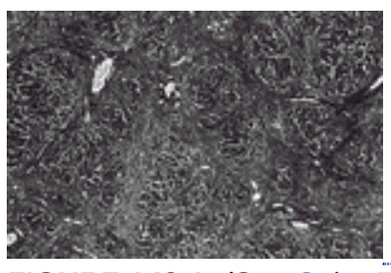

FIGURE 143-1. (See Color Fig. 143-1.) Cirrhosis of liver with fibrous septa and regenerative nodules. (Masson stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

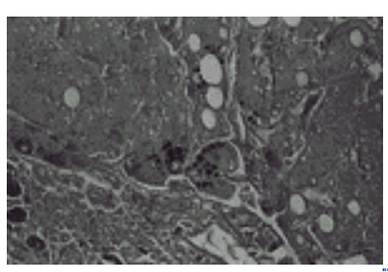

FIGURE 143-2. (See Color Fig. 143-2.) a $_1$-Antitrypsin globules in the periportal hepatocytes-diastase resistant PAS-positive. (Di-PAS stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

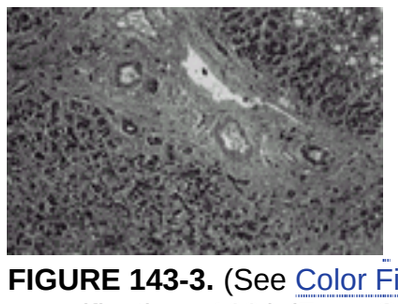

FIGURE 143-3. (See Color Fig. 143-3.) Perls iron stain demonstrating bright blue granules in hepatocytes and duct epithelial cells in hemochromatosis. (Original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

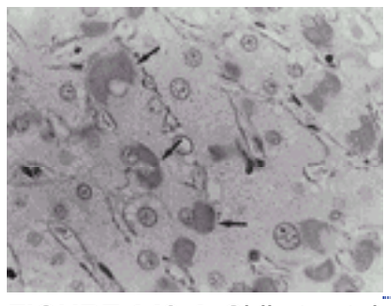

FIGURE 143-4. Shikata stain demonstrating the presence of HB $_S$Ag in the hepatocytes (*arrows*) in chronic hepatitis B virus. (Shikata stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

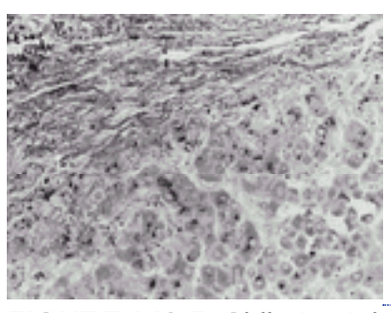

FIGURE 143-5. Shikata stain demonstrating dark black granules of copper binding protein in periseptal hepatocytes in Wilson disease. (Original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Special Requirements

Frozen sections of fresh or fixed tissue are needed to demonstrate fat with oil red “O” stain in cases of Reye syndrome, fatty liver of pregnancy, and fat-soluble vitamin A in Ito cells in hypervitaminosis A. For quantitative analysis of hepatic copper or iron, a part of the tissue needs to be saved either fixed or fresh frozen prior to processing for paraffin embedding. Electron microscopic examination is not a routine necessity and is used for special studies.

SYSTEMATIC APPROACH TO THE REVIEW OF LIVER BIOPSY

The biopsy section should be reviewed on low-power field to screen all fragments present in the section. At this time, an evaluation of the overall architecture is made. Regularly placed portal tracts and hepatic venules with their proper relationship constitute normal architecture. The portal areas can be expanded with fibrosis. When there are fibrous septa from portal to portal or to perivenular areas with formation of regenerative nodules, a diagnosis of cirrhosis is made (see Fig. 143-1). In nodular regenerative hyperplasia, the parenchyma exhibits regenerative nodules compressing the adjacent hepatic cords without fibrous tissue around them (Fig. 143-6). Collapsed reticulin fibers can be differentiated from true fibrosis by reticulin and trichrome stains. When there is confluent or submassive liver necrosis, there is loss of hepatocytes from hepatic cords resulting in a collapsed or compressed reticulum network. If this occurs in the perivenular region, it can mimic fibrosis (Fig. 143-7). Immediately following the overall assessment of the hepatic architecture, special attention is paid to each of the elements of the hepatic lobule.

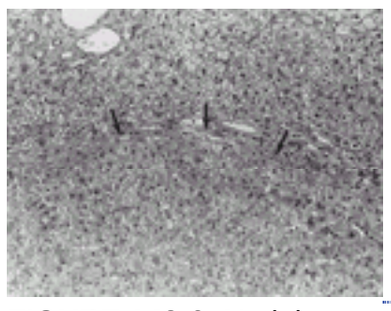

FIGURE 143-6. Nodular regenerative hyperplasia demonstrating regeneration of parenchyma (*arrows*) compressing the surrounding parenchyma without fibrous septa formation. (H & E stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

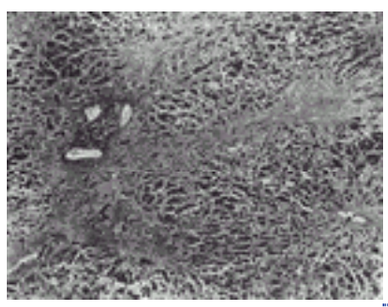

FIGURE 143-7. Submassive hepatic necrosis with collapsed perivenular reticulum network. (H & E stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Portal Areas

Enlargement of the portal tracts can be due to either cellular infiltration or fibrosis. Cellular infiltrate can be of inflammatory reactive changes, such as in a variety of viral, drug-induced, or autoimmune types of hepatitis, or neoplastic infiltrate, such as lymphomas. Inflammatory infiltrates are further categorized as acute with neutrophils, usually seen in acute cholangitis with orientation around the dilated ducts due to biliary obstruction (Fig. 143-8) or diseases such as oriental cholangiohepatitis. Predominant eosinophilic infiltrates, not oriented to the ducts, can be seen in allergic drug reactions (Fig. 143-9; see also Color Fig. 143-9), parasitic diseases, and acute rejection following liver transplantation (Fig. 143-10; see also Color Fig. 143-10). Among the inflammatory infiltrates of mononuclear type, predominant plasma cells are seen in autoimmune chronic active hepatitis (CAH) (Fig. 143-11), mixed mononuclear cells are seen in viral-induced chronic active hepatitis, and cells oriented around the bile ducts are seen in primary biliary cirrhosis. Portal tracts may contain granulomata due to various etiologies, as well as hypertrophic macrophages with phagocytized pigments, such as iron or foreign material. Polarizable talc crystals are often seen in intravenous drug abusers (Fig. 143-12; see also Color Fig. 143-12). The pattern of portal fibrosis varies with the type of disease. For example, in chronic biliary tract diseases, the fibrosis is often periductal, concentric, and lamellar (Fig. 143-13) in nature, while in chronic alcoholic liver disease and in CAH of non-A, non-B type, it is arachnoid with irregular extension into the parenchyma (Fig. 143-14; see also Color Fig. 143-14). Bile ducts are involved in acute as well as chronic disease processes. In acute cholangitis, usually due to mechanical obstruction, there is dilation of the duct with periductal edema and polymorphonuclear leukocytes infiltrating the duct epithelium and within the duct lumina (see Fig. 143-8). In chronic biliary obstruction, there is bile duct proliferation and biliary fibrosis with periductal orientation (Fig. 143-15). Periductal concentric fibrosis is also seen in sclerosing cholangitis (Fig. 143-16). There is chronic inflammatory reaction involving the duct epithelium in early stages of primary biliary cirrhosis (PBC) (Fig. 143-17) and, in the late stage, the ducts are absent. Similar changes are seen in chronic rejection of liver transplantation. Anomalous duct changes include Meyenburg complexes (Fig. 143-18) and cystic disease such as Caroli disease. Paucity of bile ducts is seen in intrahepatic biliary atresia while biliary fibrosis and bile duct proliferation are seen in extrahepatic biliary atresia (Fig. 143-19). Portal veins are thin-walled vascular structures, and are increased in number and dilated in portal hypertension (Fig. 143-20). There is inflammation and thrombosis of portal veins in pyelephlebitis. Primary or metastatic tumors can be seen in the portal venous structures. Hepatic artery can reveal hyalinization in diabetics. Polyarteritis nodosa and giant cell arteritis can produce typical inflammatory reaction involving the arterial wall (Fig. 143-21). There is arterial hypoplasia in congenital hepatic fibrosis. In Osler-Weber-Rendu syndrome there is an increase in abnormal vascular channels suggestive of arteriovenous shunting (Fig. 143-22). There is deposition of amyloid on the arterial wall in systemic amyloidosis.

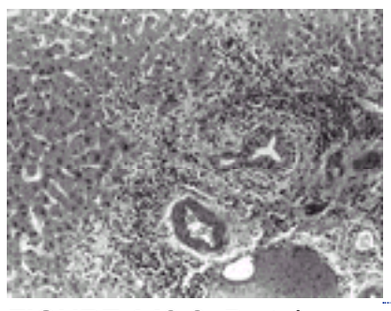

FIGURE 143-8. Portal area with prominent neutrophils in close proximity to the dilated interlobular bile duct in acute cholangitis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

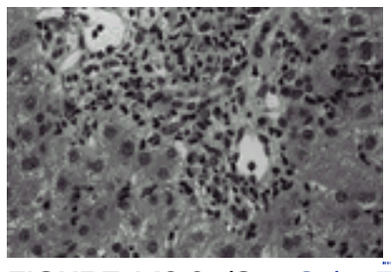

FIGURE 143-9. (See Color Fig. 143-9.) Portal area with prominent eosinophils among the inflammatory infiltrates in a case of Dilantin-induced hepatotoxicity. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

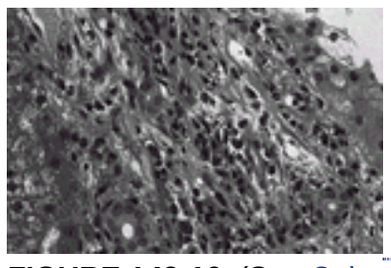

FIGURE 143-10. (See Color Fig. 143-10.) Portal area with increased number of eosinophils in a case of early rejection of orthotopic liver transplantation. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

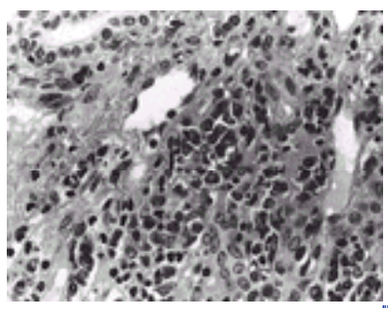

FIGURE 143-11. Prominent plasma cells among the infiltrates in the portal tract of autoimmune chronic active hepatitis. (H & E stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

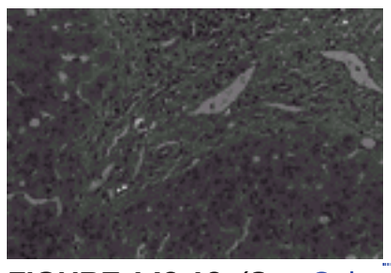

FIGURE 143-12. (See Color Fig. 143-12.) A portal area under polarizing light to demonstrate polarizable crystals in an intravenous drug user. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

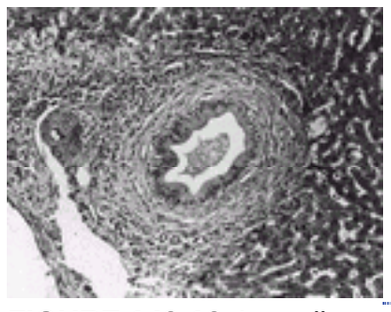

FIGURE 143-13. Lamellar periductal fibrosis in chronic bile duct obstruction. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

FIGURE 143-14. (See Color Fig. 143-14.) Arachnoid portal fibrosis with periportal extension of collagen in chronic alcoholic liver disease. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

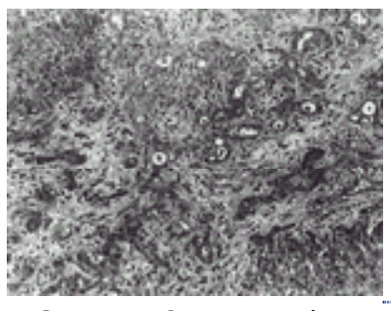

FIGURE 143-15. Portal area with marked cholangiolar proliferation in mechanical duct obstruction. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

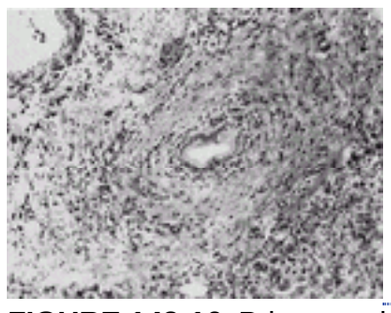

FIGURE 143-16. Primary sclerosing cholangitis with evidence of periductal fibrosis and chronic inflammatory infiltrate. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

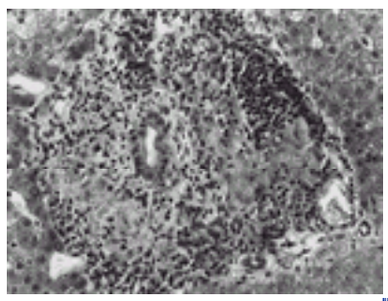

FIGURE 143-17. Primary biliary cirrhosis with granuloma. (Original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

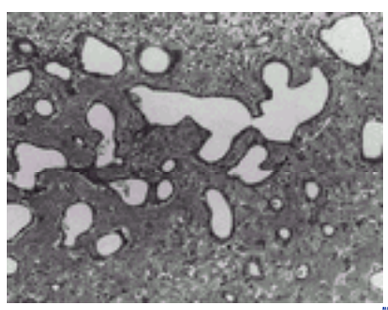

FIGURE 143-18. A few dilated duct structures with abnormal epithelium surrounded by loose collagen representing Meyenburg complex. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

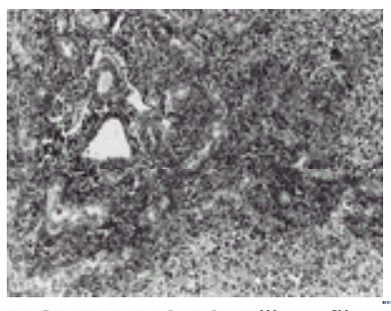

FIGURE 143-19. Biliary fibrosis and ductular proliferation in a 3-month-old infant with extrahepatic biliary atresia. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

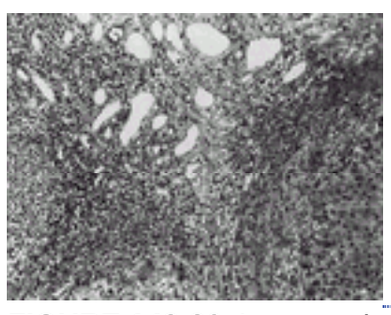

FIGURE 143-20. Increased number of thin-walled vascular structures representing portal venous radicles reflective of portal hypertension. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

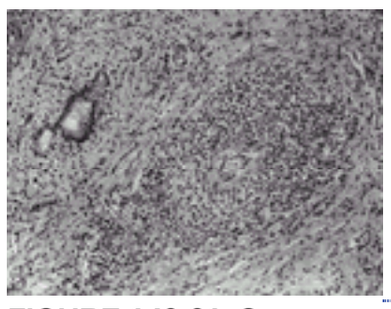

FIGURE 143-21. Severe necrotizing inflammatory reaction around hepatic arteriole in polyarteritis nodosa. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

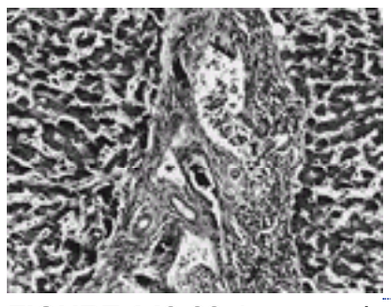

FIGURE 143-22. Increased number of abnormal vascular structures in a portal tract in Osler-Weber-Rendu syndrome. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Terminal Hepatic Venules

Terminal hepatic venules are thin, endothelial-lined vascular spaces without collagen deposition. There is perivenular fibrosis in most types of alcoholic liver disease (ALD) (Fig. 143-23). Endophlebitis with inflammatory reaction of terminal hepatic venules is seen in acute rejection of liver transplantation (Fig. 143-24). In Budd-Chiari syndrome, there is obliteration of the terminal hepatic veins along with hemorrhagic necrosis of the perivenular hepatocytes and dilation of sinusoids (Fig. 143-25).

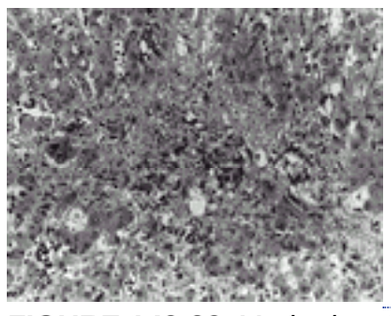

FIGURE 143-23. Marked perivenular fibrosis in alcoholic liver disease. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

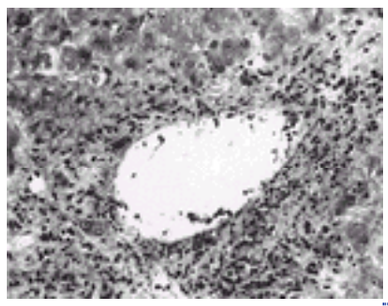

FIGURE 143-24. Endothelialitis showing inflammatory changes of a terminal hepatic venule in acute rejection of orthotopic liver transplantation. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

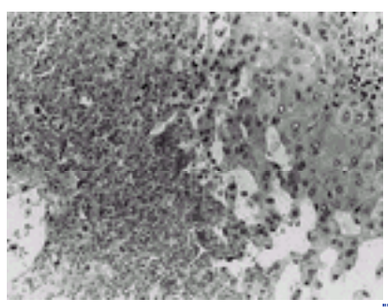

FIGURE 143-25. Budd-Chiari syndrome with perivenular hemorrhage, necrosis, and sinusoidal dilation. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Hepatic Parenchyma

The hepatic parenchyma should be evaluated with orientation to the three zones of Rappaport. Zone 3, or the perivenular zone, is most susceptible to anoxic changes and most drug-induced liver necrosis occurs here. Acute alcoholic injury, as well as acute viral hepatitis, reveals a predominance of zone 3 involvement. Periportal cell necrosis can be seen in some cases of acute type A viral hepatitis, in ferrous sulfate toxicity, and in toxemia of pregnancy. The term *spotty necrosis* is used when there are small scattered foci of hepatocytolysis replaced by a small group of hyperplastic Kupffer cells. Confluent necrosis occurs when a larger number of liver cells, such as an entire zone, is necrotic (Fig. 143-26). Submassive and massive necrosis are more severe forms. In submassive necrosis, entire lobules are involved with sparing of a few periportal hepatocytes, while in massive necrosis, no viable hepatocytes are present (Fig. 143-27). The term granulomatous necrosis is used when the area of necrosis is well circumscribed and is composed of a compact arrangement of macrophages, lymphocytes, and plasma cells (Fig. 143-28). These differ from true granulomata in lacking epithelioid cells and multinucleated giant cells. Coagulative necrosis denotes eosinophilic change of hepatocytes with loss of nuclei and maintenance of reticulin without inflammatory infiltration. This is seen in anoxia (see Fig. 143-26) or toxic necrosis due to halothane or acetaminophen (Fig. 143-29 and Fig. 143-30).

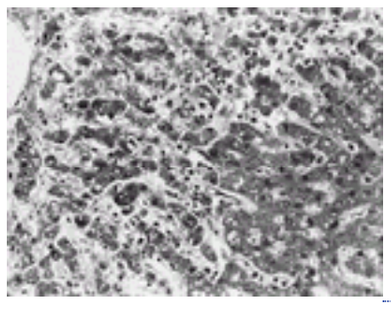

FIGURE 143-26. Confluent necrosis in the perivenular zone due to anoxia. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

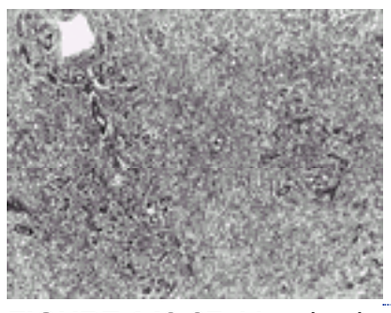

FIGURE 143-27. Massive hepatic necrosis involving the entire parenchyma with islands of portal tracts remaining. (H & E stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

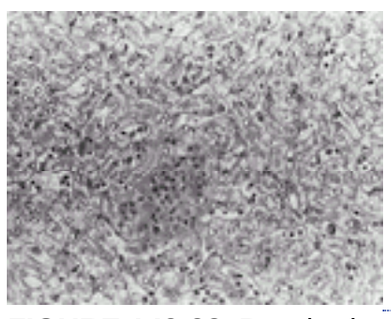

FIGURE 143-28. Punched out granulomatous necrosis of the parenchyma in mononucleosis due to Epstein-Barr virus. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

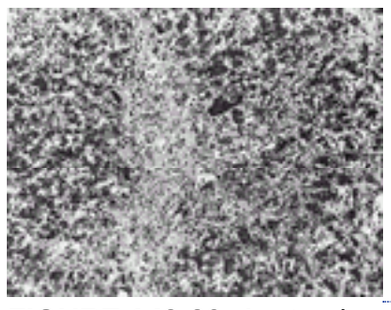

FIGURE 143-29. Acetaminophen toxicity resulting in perivenular coagulative necrosis without hepatocyte swelling. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

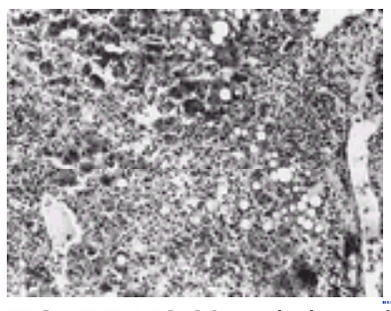

FIGURE 143-30. Halothane-induced perivenular and midzonal coagulative necrosis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Parenchymal Inclusions

The most common pigment seen in the liver cells is lipochrome or lipofuscin in the wear- and-tear pigment prominent in zone 3. Hemosiderin pigment is usually seen in periportal hepatocytes (see Fig. 143-3; see also Color Fig. 143-3). Bile can be seen in the hepatocytes and Kupffer cells but often it is associated with canalicular bile plugs. Mallory hyalin is seen as eosinophilic clumpy, ropy material in hepatocytes that are hydropic. It represents aggregates of intermediate filaments. In acute ALD, hyaline necrosis is associated with neutrophilic infiltrates usually in perivenular areas (Fig. 143-31; see also Color Fig. 143-31). Mallory bodies can be seen in periportal hepatocytes in chronic biliary diseases such as primary biliary cirrhosis or chronic obstruction (Fig. 143-32). Another eosinophilic inclusion body that is often spherical but of variable size is a $_1$-antitrypsin globule, which is seen in heterozygous or homozygous deficiency of a $_1$-antitrypsin resulting in an accumulation of the protein in hepatocyte. This is also seen in periportal hepatocytes. These globules are PAS-positive with resistance to diastase digestion (see Fig. 143-2; see also Color Fig. 143-2.). In some cases of ALD, giant mitochondria that are spherical or needle-shaped can be seen in hepatocytes (Fig. 143-33; see also Color Fig. 143-33). Among the viral inclusions, ground glass hepatocytes with excessive HB $_S$Ag in chronic hepatitis B virus (HBV) infection, nuclear and cytoplasmic inclusions of cytomegalovirus (CMV), and nuclear inclusion of herpes simplex are important.

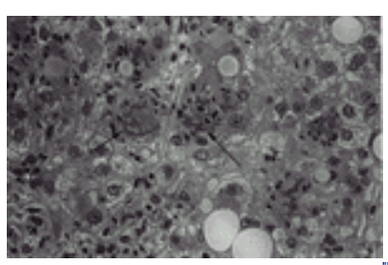

FIGURE 143-31. (See Color Fig. 143-31.) Perivenular hepatocytes containing Mallory hyaline (*arrows*) with neutrophilic reaction around them. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

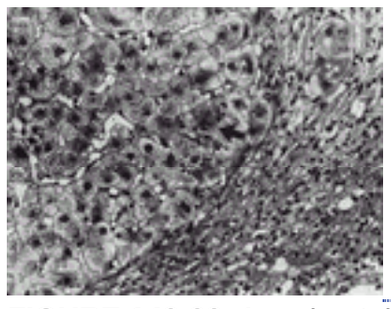

FIGURE 143-32. A periportal hepatocyte containing Mallory hyaline (*arrow*) in primary biliary cirrhosis. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

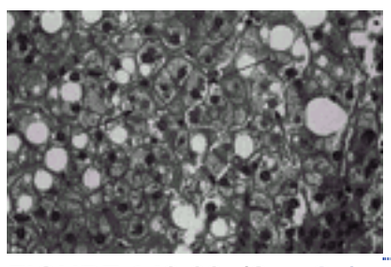

FIGURE 143-33. (See Color Fig. 143-33.) Hepatocytes containing spherical megamitochondria (*arrows*) in alcoholic liver disease. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Cord Pattern Hepatocytes are arranged in a radial fashion alternating with the sinusoidal blood spaces. In cases of diffuse cell swelling, this pattern is altered. When there is uniform cell enlargement with crisp nuclei and the cells are arranged in a cobblestone formation, it indicates focal regenerative activity (Fig. 143-34). Diffuse cobblestone pattern is seen in persistent viral hepatitis.

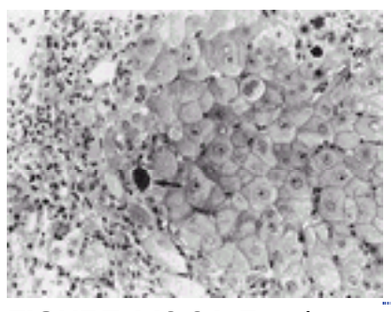

FIGURE 143-34. Focal regeneration with cobblestone arrangement of hepatocytes in chronic active hepatitis. *Arrow* points to acidophilic body. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Nuclear Changes Nuclear changes of hepatocytes include dysplasia, defined as enlargement of a group of cells with high nuclear cytoplasmic ratio (Fig. 143-35). Nuclear polyploidy means irregularity and enlargement of individual cell nuclei. Nuclear membrane invagination and glycogen vacuolization are other frequent changes (Fig. 143-36). Syncytial change represents multinucleated liver cells often seen with neonatal hepatitis (Fig. 143-37).

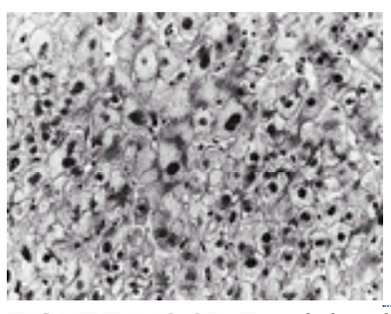

FIGURE 143-35. Focal dysplastic change consisting of enlarged cells, large nuclei in chronic active hepatitis B. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

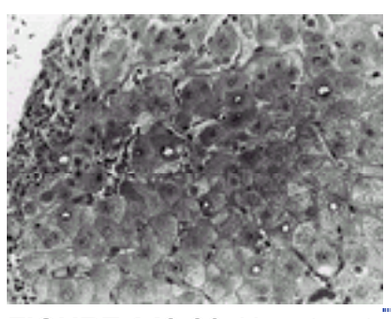

FIGURE 143-36. Hepatocytes with glycogen vacuolated nuclei. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

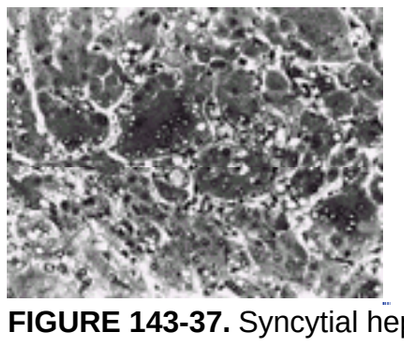

FIGURE 143-37. Syncytial hepatocytes in neonatal hepatitis. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In:

Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Sinusoidal Spaces and Lining Cells In chronic passive congestion and outflow obstruction, the sinusoids in zone 3 are dilated along with atrophy of hepatocytes (Fig. 143-38). In left-sided heart failure, red cells traverse the space of Disse and enter the hepatic trabecula (Fig. 143-39; see also Color Fig. 143-39). Focal sinusoidal dilation can be seen in space-occupying lesions in the adjacent parenchyma. Collagen deposition in the space of Disse eventually leads to narrowing and occlusion of sinusoidal spaces (Fig. 143-40; see also Color Fig. 143-40). This is commonly seen in ALD, hypervitaminosis A, chronic outflow obstruction including constrictive pericarditis, and inferior vena cava web lesions, as well as in methotrexate toxicity. In amyloidosis, there is deposition of amyloid either as a reticular type or globular amyloid in the space of Disse (Fig. 143-41 and Fig. 143-42). Hypertrophy of Kupffer cells is seen in many inflammatory diseases of the liver, but is most striking in salmonellosis (Fig. 143-43). Stellate cells or Ito cells are located in the space of Disse; they are the storage cells of retinoids (vitamin A metabolites). Through a process of activation, these cells become myofibroblasts causing sinusoidal fibrosis (Fig. 143-44). Peripheral circulating cells are visualized in sinusoidal blood spaces. Atypical cells of mononucleosis, leukemic cells, and cells of hairy cell leukemia can be seen (Fig. 143-45). Fibrin thrombi are seen packed within the sinusoids in disseminated intravascular coagulation and in toxemia of pregnancy (Fig. 143-46; see also Color Fig. 143-46). Sickled red blood cells are often seen in small clumps within the sinusoidals in sickle cell anemia (Fig. 143-47; see also Color Fig. 143-47).

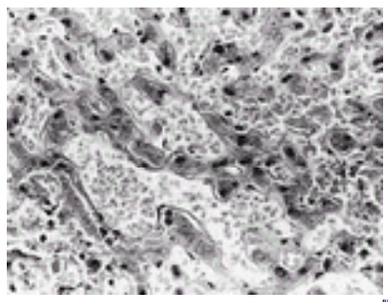

FIGURE 143-38. Chronic passive congestion causing perivenular sinusoidal dilation and atrophic hepatic cords. (Masson trichrome stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

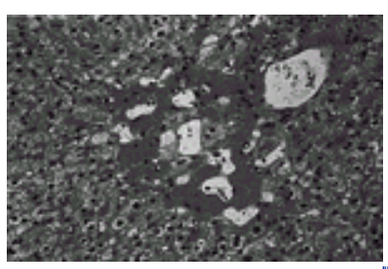

FIGURE 143-39. (See Color Fig. 143-39.) Perivenular hepatic parenchyma with dilated sinusoids and the presence of red blood cells within the hepatic cords in left-sided heart failure. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

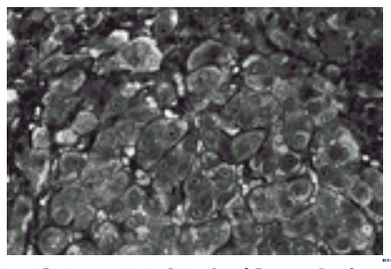

FIGURE 143-40. (See Color Fig. 143-40.) Collagen fibers along the sinusoids in the space of Disse in alcoholic liver disease. (Masson stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

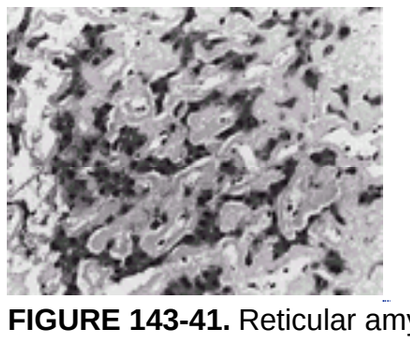

FIGURE 143-41. Reticular amyloid deposition in the space of Disse. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

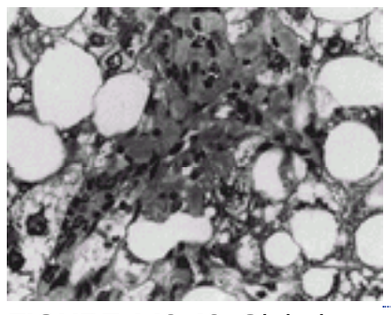

FIGURE 143-42. Globular amyloid deposition. (H & E stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

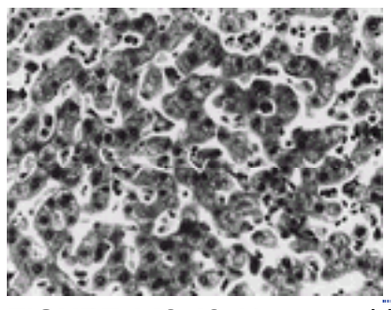

FIGURE 143-43. Hypertrophic Kupffer cells in salmonellosis. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

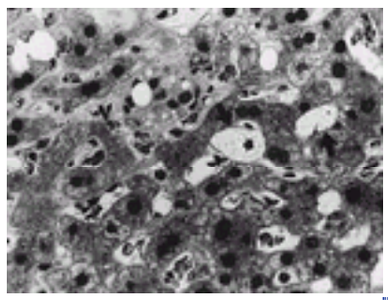

FIGURE 143-44. Ito cells with foamy fatty cytoplasm (*arrows*) along the sinusoidal surface in hypervitaminosis A. (H & E stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

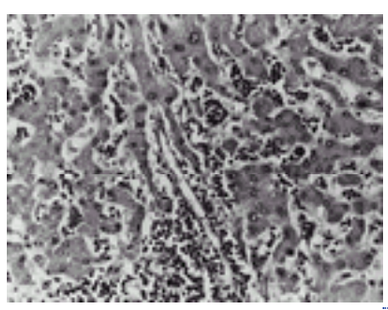

FIGURE 143-45. Leukemic cells in the sinusoidal blood space in a case of lymphocytic leukemia. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

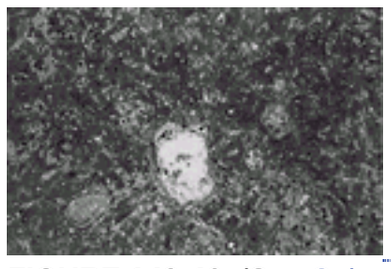

FIGURE 143-46. (See Color Fig. 143-46.) Periportal sinusoidal space filled with fibrin thrombi in toxemia of pregnancy. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

FIGURE 143-47. (See Color Fig. 143-47.) Clumps of sickled red blood cells packed in the sinusoidal spaces. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Cholestasis Bile stasis can be within the lobules in dilated biliary canaliculi or in periportal cholangioles or bile ducts. Cholestasis in zone 3 is often associated with biliary obstruction or drug-induced injury (Fig. 143-48), while periportal (zone 1) bile stasis is seen in late stages of primary biliary cirrhosis. Simple cholestasis with no associated morphologic changes of bile ducts is seen in conditions such as benign recurrent intrahepatic cholestasis, benign postoperative cholestasis, pregnancy, and administration of drugs such as estrogen or anabolic steroids, as well as in bacterial sepsis.

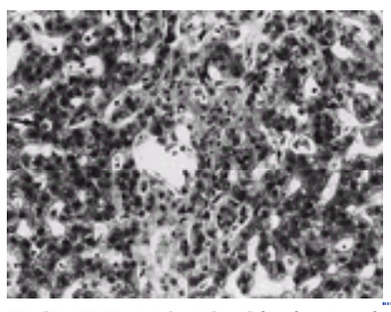

FIGURE 143-48. Cholestasis (*arrow*) in dilated canaliculi in zone 3 in chlorpromazine-induced liver disease. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Fat Two major types of fatty changes can occur in hepatocytes: macrovesicular and microvesicular types. Macrovesicular fat droplets appear as a large vacuole dislocating the nucleus and the cytoplasmic material to the periphery of the cell. Macrovesicular fatty change involving zone 3 is often seen in ALD (Fig. 143-49) and also in diabetes mellitus and in obesity. Steroid-induced fatty change involves all zones. Microvesicular fat, composed of small droplets of fat, sometimes appears as foamy change of the cytoplasm, without displacement of the hepatic nucleus. This is seen in alcoholic foamy degeneration, fatty liver of pregnancy, tetracycline

toxicity, Reye syndrome, and in valproic acid toxicity (Fig. 143-50; see also Color Fig. 143-50).

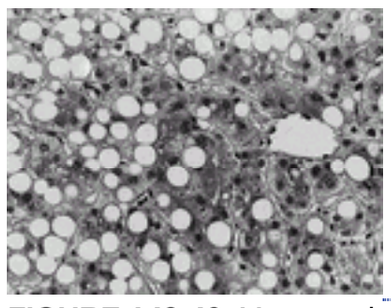

FIGURE 143-49. Macrovesicular fatty change of hepatocytes in alcoholic liver disease. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

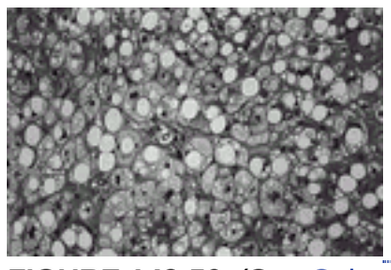

FIGURE 143-50. (See Color Fig. 143-50.) Diffusely enlarged hepatocytes with foamy fatty change in acute alcoholic liver disease. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Terminal Hepatic Venules and Venous Outflow The most common pathological change of terminal hepatic venule is perivenular and obliterative sclerosis seen in alcohol-induced liver injury (see Fig. 143-23). Similar fibrosis can also be seen in Budd-Chiari syndrome and other causes of outflow obstruction, such as constrictive pericarditis, inferior vena caval obstruction, or web lesions. Endothelialitis or inflammatory cell infiltration of terminal hepatic venules is a feature of acute rejection of liver transplantation (see Fig. 143-24).

SPACE-OCCUPYING LESIONS OF THE LIVER

Primary Tumors—Benign and Malignant

These can be neoplastic lesions, cysts, or abscesses. A CT or ultrasound-guided liver biopsy can result in a sample diagnostic of the lesion present. Cysts and abscesses are usually aspirated. Among the benign solid mass lesions, focal nodular hyperplasia is one of the most common and must be differentiated from liver cell adenoma. [9], [10] Liver cell adenoma does not have a capsule but compresses an adjacent parenchyma (Fig. 143-51). The tumor cells are benign with normal nuclear cytoplasmic ratios, and with lack of portal tracts and terminal veins in the lesions. There are prominent, thick-walled arteries present along the edge of the tumor (Fig. 143-52). Focal nodular hyperplasia has a typical central stellate scar with loose vascularized fibrous tissue containing atypical ductal elements along the periphery of the scar (Fig. 143-53). This lesion also lacks normal portal areas and terminal hepatic veins (Fig. 143-54). Among the primary malignant neoplasms of the liver, hepatocellular carcinoma (HCC) is the most common form. [11] Other forms include cholangiocarcinoma and angiosarcoma. The tumor cells of the majority of typical HCC have a trabecular pattern with increased numbers of cell thicknesses forming fingerlike projections into vascular spaces. These trabeculae are lined by endothelial cells and surrounded by vascular spaces (Fig. 143-55; see also Color Fig. 143-55). The tumor can have focal acinar changes with secretory material in the lumina. Sclerosing hepatic carcinoma is a variant with a dense collagenized stroma (Fig. 143-56; see also Color Fig. 143-56). The fibrolamellar variant has lamellar strands of collagen separating thin cords of large eosinophilic neoplastic hepatocytes [12] (Fig. 143-57; see also Color Fig. 143-57). Cholangiocarcinomas are composed of neoplastic ductal elements surrounded by dense fibrous stroma [11] (Fig. 143-58; see also Color Fig. 143-58). The tumor is clearly demarcated from the surrounding liver.

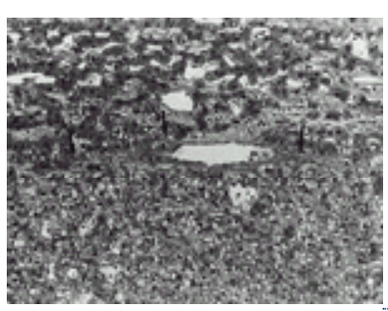

FIGURE 143-51. Liver cell adenoma with clear cells and the adjacent normal parenchyma (*arrows* pointing to the edge of the adenoma). (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

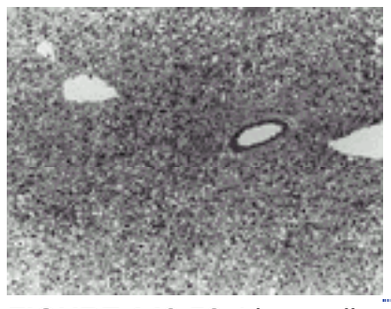

FIGURE 143-52. Liver cell adenoma with thick-walled vessels and lack of portal tracts. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

FIGURE 143-53. Focal nodular hyperplasia (FNH) with central stellate scar. (H & E stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

FIGURE 143-54. Focal nodular hyperplasia (FNH) with the scar exhibiting lack of bile ducts and presence of vascular structures. Liver cells are uniform and regenerative in appearance. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

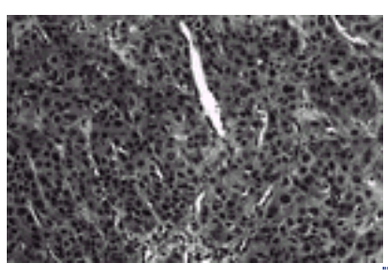

FIGURE 143-55. (See Color Fig. 143-55.) Well-differentiated trabecular hepatocellular carcinoma with endothelial lining. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

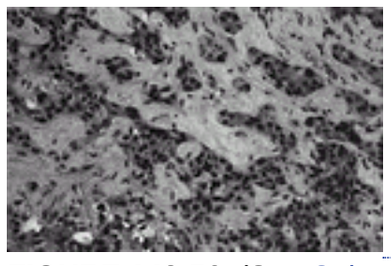

FIGURE 143-56. (See Color Fig. 143-56.) Sclerosing hepatic carcinoma with dense fibrous stroma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

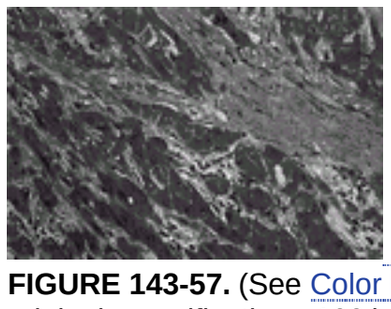

FIGURE 143-57. (See Color Fig. 143-57.) Eosinophilic neoplastic hepatocytes with lamellar fibrous stroma in fibrolamellar hepatocellular carcinoma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

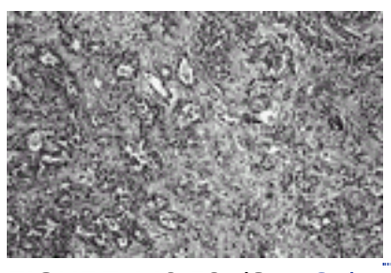

FIGURE 143-58. (See Color Fig. 143-58.) Neoplastic ductal structures with fibrous stroma in cholangiocarcinoma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Nodular Regenerative Hyperplasia (NRH)

NRH is a rare form of hyperplasia of liver cells with formation of nodules without fibrous septa. [13] Microscopically, the hyperplastic nodules are better identified on low power exhibiting compression by the surrounding parenchyma (see Fig. 143-6). A reticulum stain outlines these nodules, which can vary from small and microscopic to large, up to 23 cm. Patients with NRH may have evidence of portal hypertension.

Metastatic Tumors

Metastatic tumors in the liver can be easily diagnosed from an ultrasound-guided needle biopsy. The tumor cells usually grow into the adjacent sinusoids, compressing and separating the hepatic trabeculae (Fig. 143-59). Examination of the junction of the tumor and the nontumor is very useful in differentiating HCC from metastatic tumors. In HCC, the tumor cells merge into the hepatic cords where there is a transition from normal to neoplastic cells (Fig. 143-60; see also Color Fig. 143-60). In metastatic tumors, as mentioned earlier, the tumor grows into the sinusoidal spaces.

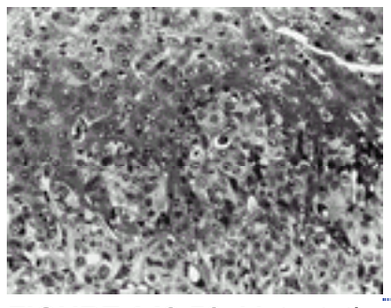

FIGURE 143-59. Metastatic, poorly differentiated adenocarcinoma infiltrating into the sinusoids. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

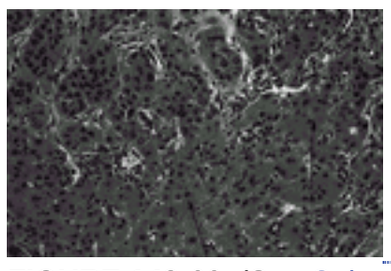

FIGURE 143-60. (See Color Fig. 143-60.) Junction of tumor and nontumor liver in hepatocellular carcinoma. The tumor cells grow into the hepatic cords (*arrows*). (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Granulomas Granulomas are identified in the liver at a rate of 25% of all liver biopsies in most of the large centers. [14] The most common cause of epithelioid granulomas in the liver is sarcoidosis (Fig. 143-61). Among infectious agents causing granulomas in the liver, *Mycobacterium tuberculosis, Mycobacterium avium intracellulare,* Q fever, and brucellosis are most common (Fig. 143-62, Fig. 143-63 and Fig. 143-64). Rare fungal infections such as blastomycosis and coccidiomycosis are the next most common causes. Parasitic infections such as schistosomiasis with hepatic granulomas are common in some endemic areas of the world. Epithelioid granulomas are well circumscribed and comprise multinucleated giant cells, epithelioid cells, and lymphocytes along the periphery. Granulomatous lesions, which are not true granulomas, are small punched out areas of hepatic necrosis with aggregates of macrophages and lymphocytes without epithelioid cell formation. These lesions are seen in viral infections such as CMV, Epstein-Barr virus (EBV), rickettsial infections, such as Q fever, and in liver injury caused by drugs such as Dilantin, allopurinol, Butazolidin, and sulfonamides. Some of these drugs can also elicit an epithelioid reaction with true granuloma formation (Fig. 143-65). Hepatic granulomas can also be associated with Hodgkin disease, postjejunoileal bypass, and PBC (see Fig. 143-17). Some unique features useful in the differential diagnosis of hepatic granulomas include: a central vacuolated space with a fibrin ring in granulomas [15] of Q fever representing the so-called “donut” lesions (see Fig. 143-64); demonstrations of acid-fast and fungal organisms using special stains; the presence of eosinophils in granulomas associated with drug reactions, parasitic diseases, and Hodgkin disease; the presence of remnants of ova in schistosomiasis (Fig. 143-66); and caseation necrosis, rarely occurring in tuberculous granulomas (see Fig. 143-62).

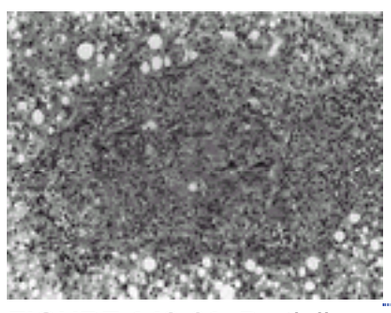

FIGURE 143-61. Partially segmented, exuberant epithelioid granuloma of sarcoidosis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

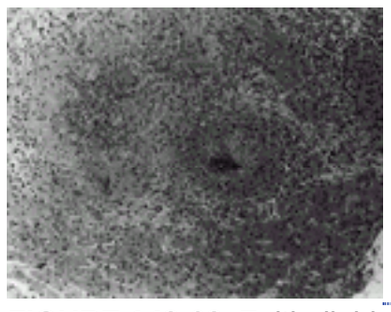

FIGURE 143-62. Epithelioid granuloma with Langhans giant cells and one showing central necrosis (**left**) in *Mycobacterium tuberculosis* of liver. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

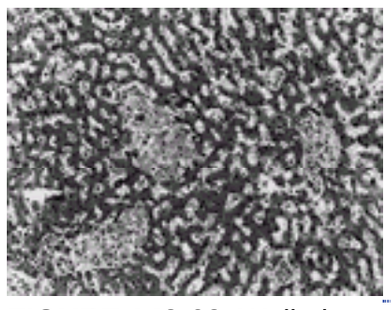

FIGURE 143-63. Well-circumscribed clusters of large foamy histiocytes in *Mycobacterium avium intracellulare* infection of the liver. These cells contain abundant acid-fast organisms on special stain (not shown). (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

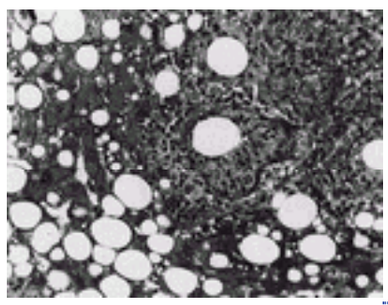

FIGURE 143-64. Granulomatous lesion with central vacuolization surrounded by a fibrin ring in Q fever. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

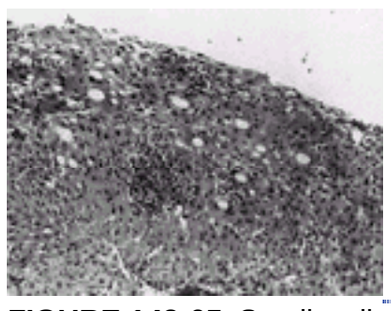

FIGURE 143-65. Small well-circumscribed granuloma in sulfonamide-induced hepatic necrosis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

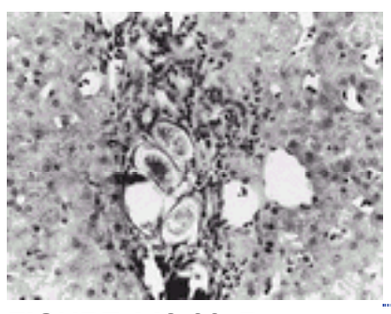

FIGURE 143-66. Remnants of ova of schistosomal organisms in a fibrous portal area. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

DIFFERENTIAL DIAGNOSIS OF CIRRHOSIS

Based on the morphologic features, cirrhosis of the liver can be classified as alcoholic, nonalcoholic, biliary, and cardiac cirrhosis. Based on the size of the regenerative nodules, a broad distinction is made between micronodular (<3-mm nodules) and macronodular (>3-mm nodules) cirrhosis. A typical alcoholic cirrhosis is a micronodular type diagnosed by the presence of dense, broad fibrous scars with small parenchymal nodules exhibiting sinusoidal collagenosis (see Fig. 143-1; see also Color Fig. 143-1.). Fat, alcoholic hyaline, and other acute changes due to alcohol can be superimposed. Mild chronic inflammatory reaction of nonspecific nature can be present in the fibrous septa without periseptal or parenchymal activity. Cirrhosis due to hemochromatosis, also a micronodular cirrhosis, is characterized by the presence of hemosiderin deposition in the parenchymal cells as well as in the fibrous septa and in bile duct epithelia. There is a striking lack of inflammatory process in this type of cirrhosis. Nonalcoholic cirrhosis, usually of macronodular type, is a broad category where the cirrhosis is the end result of CAH of various etiologies. Fibrous septa are relatively thin and vascularized and reveal prominent chronic inflammatory infiltration. The inflammation extends into the periseptal parenchyma, which exhibits a varying degree of necroinflammatory activity. In biliary cirrhosis, the fibrous septa reveal a characteristic lamellar pattern and extend from portal to portal areas leaving islands of parenchyma. This is best described as a jigsaw puzzle-like appearance (Fig. 143-67; see also Color Fig. 143-67). Cardiac cirrhosis, on the other hand, has a reverse lobular pattern, as the perivenular fibrous areas form septa leaving the portal areas relatively uninvolved.

Following a morphologic identification of the type of cirrhosis, an etiologic diagnosis is attempted. Morphology of alcoholic cirrhosis can also be seen in postjejunoileal bypass liver disease. Among the etiologies of nonalcoholic cirrhosis, chronic HBV infection can be diagnosed by the presence of ground-glass cells (Fig. 143-68) and significant dysplastic changes of the hepatocytes (see Fig. 143-35). Autoimmune hepatitis can be suspected as the etiology if there is a predominance of plasma cells among the portal and parenchymal infiltrates (see Fig. 143-11). In addition, areas of collapse may exist even in the cirrhotic stage in these cases. Wilson disease can be suggested if there is evidence of increased hepatic copper and copper binding protein (see Fig. 143-5). In cases of biliary cirrhosis, absence of bile duct elements with the presence of lymphoid infiltrates in the septa is suggestive of primary biliary cirrhosis while evidence of bile duct proliferation with or without acute cholangitis is suggestive of biliary obstruction leading to cirrhosis. Cirrhosis in infants and children due to a $_1$-antitrypsin deficiency, intra- or extrahepatic biliary atresia, and in cystic fibrosis involving liver also appears biliary in nature. Clinical, laboratory, and radiologic data are needed to arrive at a more accurate etiologic diagnosis of cirrhosis in most cases.

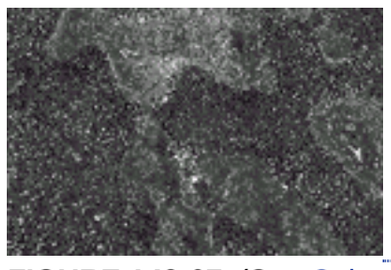

FIGURE 143-67. (See Color Fig. 143-67.) Jigsaw-puzzle appearance of biliary cirrhosis. (Masson stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

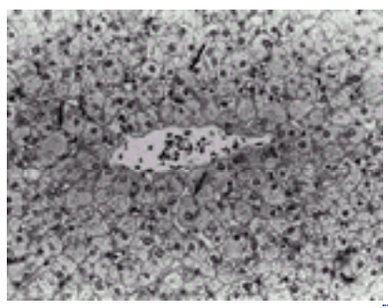

FIGURE 143-68. Scattered ground-glass cells (*arrows*) in chronic hepatitis B. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

SPECIFIC LIVER DISEASES

Necroinflammatory Diseases

Acute Viral Hepatitis Regardless of the etiology, typical perivenular hepatocytolysis, extreme hydropic swelling of the hepatocytes, marked Kupffer cell hyperplasia, and mononuclear exudative reaction in the parenchyma as well as the portal tracts are found in cases of acute viral hepatitis [16] (Fig. 143-69 and Fig. 143-70). Numerous acidophilic bodies are also present. The acidophilic bodies represent individual hepatocytes undergoing necrosis with cytoplasmic condensation and nuclear pyknosis. Cholestasis may or may not be present. The hepatocytes in zone 1 usually reveal uniform hydropic change suggestive of regeneration. Portal tracts are expanded with inflammation without fibrosis (see Fig. 143-70). Some of the following specific changes are associated with different viral agents: Acute type A hepatitis can exhibit a periportal accentuation of the necrosis. [17] Enteric non-A, non-B (type E) acute hepatitis tends to have prominent acinar changes with hepatocytes arranged around dilated biliary canaliculi resembling rosettes throughout the lobules [18] (Fig. 143-71). In acute hepatitis due to delta (type D) agent, one can demonstrate the presence of hepatitis D antigen (HDAg) by immunoperoxidase. When the necrosis in acute viral hepatitis (AVH) is severe, it involves the entire zone 3 and is called confluent necrosis. When the necrosis involves zones 2 and 3 and part of zone 1 with hepatocytes remaining viable in the periportal zones, it is classified as submassive necrosis. Massive necrosis is the term used when the necrosis involves all zones without any viable hepatocytes.

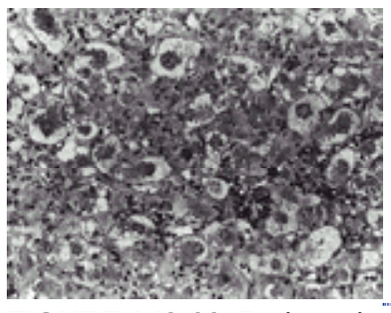

FIGURE 143-69. Perivenular zone in acute viral hepatitis demonstrating hydropic hepatocytes, hepatocytolysis, inflammatory exudate, and rare acidophilic bodies. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

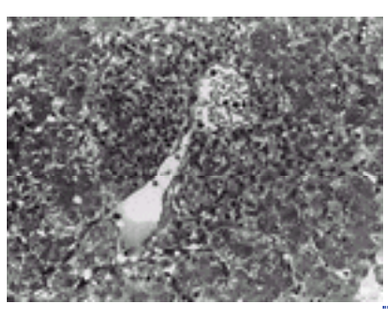

FIGURE 143-70. Portal area in acute viral hepatitis with mononuclear infiltration extending to the periportal regions. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

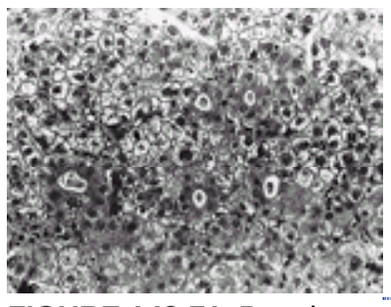

FIGURE 143-71. Prominent acinar transformation of hepatocytes in enterically transmitted acute hepatitis, type E. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Chronic Viral Hepatitis

Terminology. Chronic hepatitis is the universal terminology used for all liver biopsies from patients with virologic markers persistent for greater than 6 months with abnormal liver enzymes. Terms such as persistent hepatitis, chronic active hepatitis, chronic lobular hepatitis, and so forth are no longer used by pathologists. The diagnosis of “chronic hepatitis” is further elaborated by adding the name of the etiologic virus (i.e., type B, type C, or type D) and by adding the assessment of necroinflammation and fibrosis in a semiquantitative method, using grades (0–4) and stages (1–4) as proposed by Ludwig. [19] Detailed quantitation assessments have

included many scoring systems; the most commonly used are those devised by Knodell and colleagues, Ishak colleagues, and the Metavir scoring system. [20], [21] and [22] *Knodell scoring* has a total of 18 for necroinflammation including lobular, periportal, and portal inflammation, and a total of 4 for fibrosis (0–4). The *Ishak scoring system* has more details for necroinflammation (0–18) and expanded scores for fibrosis (0–6). *Metavir scoring* is simpler with a combination score for inflammation including periportal and lobular activity (A0–A3) and a fibrosis score on a scale of F0 to F4. The term persistent hepatitis (PH) has been used primarily in cases of chronic "B" infections when the liver biopsy reveals mild expansion of portal tracts with mononuclear inflammatory hyperplasia without periportal extension or fibrosis. The parenchyma reveals diffuse mild hydropic swelling, suggestive of a cobblestone pattern (Fig. 143-72). Ground- glass hepatocytes are usually scattered uniformly among the lobules. Rare foci of necrosis are seen and there is no Kupffer cell activity. This lesion is generally nonprogressive in type B. It is uncommon to have PH when there are chronic B and D co-infections. Rather, progressive chronic hepatitis is seen.

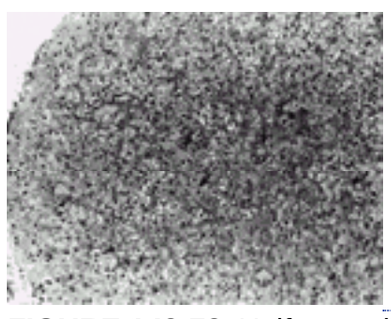

FIGURE 143-72. Uniform cobblestone appearance of parenchyma in chronic hepatitis, type B. A few ground-glass cells are seen (*arrows*). (H & E stain; original magnification ×200.) (Reprinted with permission from Govindarajan S. Liver Biopsy Interpretation. In Kaplowitz N. (ed.). Liver and Biliary Diseases, Second Edition. Baltimore, Williams & Wilkins, 1996.)

The most important morphologic characteristics are portal fibrosis, portal and periportal mononuclear inflammation, and necroinflammatory changes within the parenchyma with an irregular distribution between one lobule and another (Fig. 143-73). There is also regenerative activity in an irregular distribution. Lymphocytes and Kupffer cells are seen cuffing around the target hepatocytes (Fig. 143-74). The degree of necrosis depends on the disease activity, more severe in exacerbations or reactivation and less so in quiescent stages. Fibrosis progresses to cirrhosis and the periseptal inflammatory activity continues with cirrhosis. The term piecemeal necrosis is used to describe the periportal extension of mononuclear inflammation in association with hepatocytolysis involving the limiting plate. In chronic hepatitis B, there is significant nuclear dysplasia (see Fig. 143-35). In cases of chronic hepatitis C, the following unique features are seen in most of the cases: macrovesicular fatty change, mild sinusoidal collagenosis in the periportal areas, prominent lymphoid reaction of portal areas, and moderate to marked atypia of the bile duct epithelium [23] (Fig. 143-75 and Fig. 143-76). In cases of chronic hepatitis B one can stain for the viral proteins such as HB $_{c}$Ag, HB $_{s}$Ag, and HB $_{e}$Ag, using immunoperoxidase methodology. [24] HB $_{s}$Ag is also demonstrable using Shikata orcein stain or Victoria blue (see Fig. 143-4). HDAg is demonstrable by immunoperoxidase techniques in chronic hepatitis D virus infections [25] (Fig. 143-77).

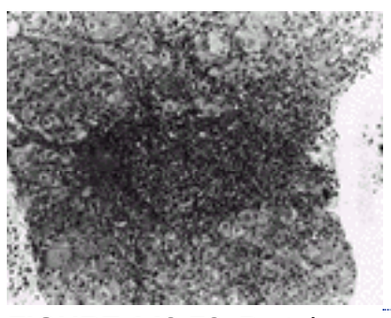

FIGURE 143-73. Portal areas with fibrosis and mononuclear inflammation extending to the parenchyma exhibiting piecemeal necrosis in chronic hepatitis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

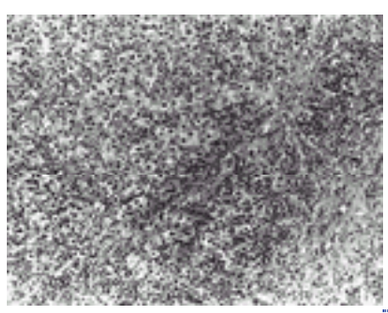

FIGURE 143-74. Inflammatory cells are seen cuffing around the hepatocytes in chronic hepatitis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

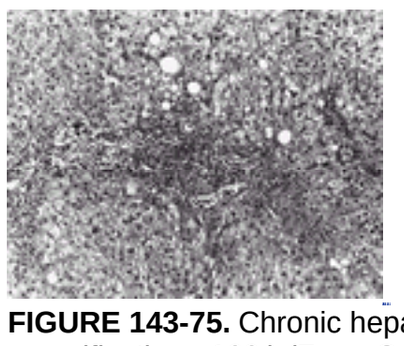

FIGURE 143-75. Chronic hepatitis C showing portal fibrosis and inflammation, macrovesicular fat, and inflammation in the adjacent parenchyma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

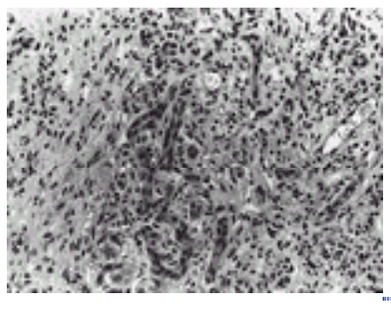

FIGURE 143-76. Atypical bile ducts in a portal tract of chronic hepatitis C. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

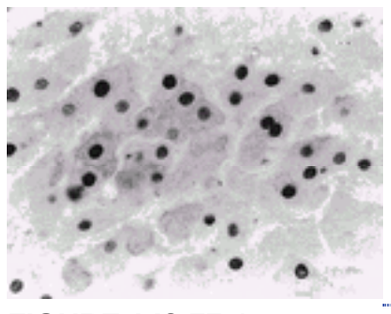

FIGURE 143-77. Immunoperoxidase stain demonstrating hepatitis D antigen in the nuclei of hepatocytes in chronic hepatitis D. (Original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Chronic Hepatitis—Autoimmune Etiology In addition to the features of chronic hepatitis already described, the changes unique to autoimmune chronic hepatitis (the term *lupoid hepatitis* is used by some, but should be discouraged as it is not related to systemic lupus erythematosis) are:

- the presence of significant number of plasma cells among the portal and the parenchymal infiltrates (see Fig. 143-11)
- evidence of collapse and striking degree of periportal as well as perivenular necrosis
- trapped hepatocytes in the fibrous portal or septal areas. [26]

Necroinflammatory changes are minimal in patients on steroid therapy with good response.

Other Viral Infections Mononucleosis pattern of hepatitis can be seen in hepatitis due to EBV, CMV, and some cases of hepatitis C virus (HCV). [27] In this form, the hepatocytes are not swollen and the cord pattern is maintained. There is striking sinusoidal lymphocytosis, many of these cells being atypical in nature (Fig. 143-78). There is portal infiltration by the same type of cells. In addition, there is multifocal punched out hepatocytolysis (see Fig. 143-28) without confluent or zonal necrosis. Very rarely, epithelioid granuloma can be seen. In immunocompromised individuals, the CMV infection results in an abundance of virally infected cells with typical nuclear inclusion bodies in the bile duct epithelium or reticuloendothelial cells. In postorthotopic liver transplantation, the CMV infection causes typical microabscess formation with the polymorphonuclear leukocytes (PMNs) surrounding the cells with viral inclusion bodies [28], [29] (Fig. 143-79; see also Color Fig. 143-79). Viral antigen in CMV hepatitis in these cases is demonstrable using immunochemical methods. Herpes simplex infection of the liver is rarely encountered in liver biopsies, since the prothrombin time is too prolonged for a biopsy in these patients. Large, discrete, irregular zones of coagulative necrosis of parenchymal cells with the presence of eosinophilic intranuclear inclusions in the viable hepatocytes at the margins of necrosis is the typical change seen in the liver biopsies [30] (Fig. 143-80).

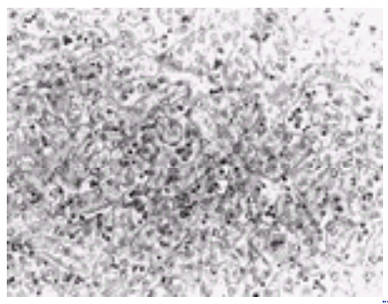

FIGURE 143-78. Striking sinusoidal lymphocytosis of atypical type in Epstein-Barr virus-induced mononucleosis. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

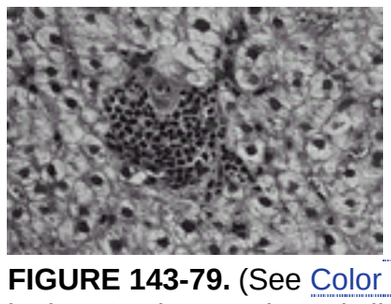

FIGURE 143-79. (See Color Fig. 143-79.) Intranuclear and cytoplasmic inclusions of cytomegalovirus (CMV) in a hepatocyte surrounded by polymorphonuclear leukocytes in an orthotopic liver transplant infected with CMV. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

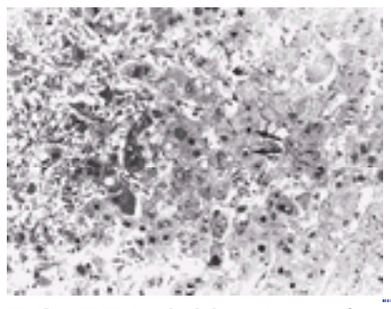

FIGURE 143-80. Intranuclear inclusions (*arrows*) of Cowdry type A of herpes simplex seen in hepatocytes. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Alcoholic Liver Disease

Changes in liver biopsies due to alcohol can be broadly divided into acute cellular damage and its characteristic features, and chronic slowly progressive changes with evidence of chronic liver disease.

Acute Changes The most common form of acute damage is seen as fatty change of macrovesicular type with or without cholestasis. The patients present with large fatty change with or without jaundice. Needle biopsy reveals large vacuoles of macrovesicular fat involving the perivenular hepatocytes (see Fig. 143-49). Occasionally, the entire lobule is uniformly involved. Cholestasis and sinusoidal fibrosis may be seen. Acute foamy fatty change (FFC) represents a variant of acute alcoholic hepatitis. [31] The perivenular hepatocytes are enlarged with foamy or microvesicular fat (see Fig. 143-50; see also Color Fig. 143-50). The nucleus is central in location and the cells may exhibit abnormal giant mitochondria visible as eosinophilic, round bodies (cherry bodies) (see Fig. 143-33; see also Color Fig. 143-33).

Sinusoidal collagen deposition can be prominent in the perivenular zones. FFC can also be seen in association with the classic acute sclerosing hyaline necrosis (ASHN) in patients with alcoholic hepatitis. [32] The morphologic hallmarks of ASHN are:

- hydropic hepatocytes with cytoplasmic Mallory hyaline bodies
- neutrophil reaction around these cells with hyaline necrosis
- sclerosis of the terminal hepatic venules with sinusoidal collagenosis (see Fig. 143-31; see also Color Fig. 143-31).

Other microscopic features, such as fatty change, cholestasis, and portal fibrosis with arachnoid extension, can also be part of this morphology. Other uncommon biopsy findings of acute ALD include lytic necrosis of liver cells in which the hepatocytes are extremely hydropic with lysis and fusion of adjacent cells and acute portal reaction in which there is prominent neutrophilic reaction within the portal tract without any orientation to the bile ducts. These portal tracts often have classic arachnoid fibrosis.

Chronic Alcohol-Induced Liver Damage This primarily manifests as fibrosis. Three forms of progressive fibrosis are seen in the liver. [33] They are:

- portal and perivenular fibrosis leading to fibrous septa formation, resulting in cirrhosis with small regenerative nodules (see Fig. 143-1; see also Color Fig. 143-1)
- diffuse interstitial fibrosis with collagen extending throughout the lobule encircling individual hepatocytes and without parenchymal regeneration or septal formation (Fig. 143-81; see also Color Fig. 143-81)

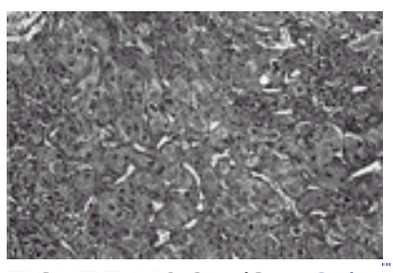

FIGURE 143-81. (See Color Fig. 143-81.) Diffuse interstitial fibrosis in chronic alcoholic liver disease. (Masson stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

- progressive perivenular fibrosis with dense scarlike collagen involving zone 3 with relatively minimal portal fibrosis (Fig. 143-82; see also Color Fig. 143-82).

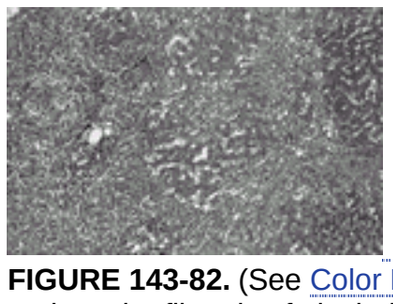

FIGURE 143-82. (See Color Fig. 143-82.) Marked perivenular fibrous scarring with mild portal fibrosis and lack of regenerative nodules in progressive perivenular fibrosis of alcoholic etiology. (Masson stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

In the early stages of development of cirrhosis the fibrous septa appear thin, with larger pseudolobules or parenchymal nodules. There is evidence of prominent intranodular sinusoidal fibrosis. As the disease progresses, the septa become denser and wider, the nodules become subdivided into smaller nodules, with continued sinusoidal fibrosis in the periseptal regions. In advanced stages, there is extensive scarring with islands of parenchymal nodules scattered within. Masson trichrome and PAS stains clearly outline this ratio of the fibrous tissue to the parenchyma. Following an established state of cirrhosis, if there is prolonged abstinence from alcohol, the regenerative parenchymal nodules enlarge, pushing the fibrous septa centrifugally with a remarkable reduction in the amount of collagen both within the parenchyma and the septa. At this inactive stage, it is difficult to identify alcohol as the initial etiologic agent causing cirrhosis. Any of the acute cellular changes described previously can occur superimposed on a cirrhotic or fibrotic liver. The distribution of Mallory hyaline, neutrophilic infiltrate, or fatty change may be irregular and focal, not uniform. When there are additional complications, such as anoxia due to variceal bleeding or general anesthesia, typical coagulative necrosis can be seen within the central zone of the cirrhotic nodules.

Differential Diagnosis Nonalcoholic steatohepatitis (NASH) or nonalcoholic fatty liver disease is unrelated to alcohol or any other recognized etiology for liver disease, but most often progressive liver disease with histological features resembling alcoholic liver disease. [34] These include micro- and macrovesicular steatosis, perivenular and periportal sinusoidal fibrosis with or without necroinflammation, and Mallory bodies. Degree of necroinflammation and ballooning changes (scale of 1–3) as well as fibrosis (1–4) have been graded by a system developed by Brunt and colleagues. [35] Differentiation is made based on clinical and laboratory data. Identical morphologic changes of ASHN can be seen in diabetes mellitus, liver disease secondary to jejunoileal bypass surgery for morbid obesity, and drug-induced reactions in patients receiving amiodorone. [36] Mallory bodies without accompanying neutrophilic reaction can be seen in a number of conditions such as Wilson disease, primary biliary cirrhosis, chronic biliary obstruction, and in livers bearing HCC either in the tumor or nontumor cells. Sinusoidal collagen deposition typical for ALD can be seen in a moderate degree in chronic HCV hepatitis, hypervitaminosis A, and methotrexate toxicity. A much more localized perivenular fibrosis can be seen in venous outflow obstruction such as Budd-Chiari syndrome. Studies of serologic testing for HCV have identified a high prevalence (>50%) of chronic HCV infection among those with ALD. In many of these patients, the liver biopsy reveals features of both ALD and chronic hepatitis of HCV type. There is an increased degree of chronic inflammatory infiltrate in the portal tracts or fibrous septa with spillover to the periseptal parenchyma.

Biliary Diseases

Mechanical Duct Obstruction This is often diagnosed without a liver biopsy. When the various procedures, such as ultrasound, endoscopic retrograde cholangiopancreatography and others, are inconclusive, a liver biopsy is performed. The changes can be divided into parenchymal and portal. Although the predominance of one change over the other varies from patient to patient, the earliest change might be parenchymal cholestasis in the perivenular hepatocytes. This is followed by canalicular dilation with bile plug formation. The hepatocytes become hydropic with a reticular rarefaction of the cytoplasm termed as feathery degeneration. The portal tracts reveal edema with loose fibrous tissue around dilated bile ducts. Portal inflammatory reaction includes PMNs and mononuclear cells. Orientation of PMN infiltration around the bile duct, and within the duct lumen, strongly suggests mechanical duct obstruction (see Fig. 143-8). Prolonged biliary obstruction results in marked cholangiolar proliferation as well as periductal fibrosis (see Fig. 143-13 and Fig. 143-15). Long-standing obstructions can result in disappearance of the ducts resembling PBC. During the acute stage, drug-induced cholangitic and cholestatic reaction, acute viral hepatitis of cholestatic type, impaired regeneration syndrome, changes due to parenteral hyperalimentation, and septic shock should be considered in the differential diagnosis. Biliary fibrosis and cirrhosis follow if there is continued duct obstruction (see Fig. 143-67).

Primary Biliary Cirrhosis (PBC) and Primary Sclerosing Cholangitis (PSC) These are discussed in detail in other chapters. Diagnosis of PBC based on a needle biopsy of liver might be difficult if adequate portal tracts are not present in the specimen. In addition to the classic destructive duct lesions with marked lymphoplasmacytic infiltrates, the presence of granulomas, absence of ducts in small portal tracts, hydropic changes of periportal hepatocytes containing Mallory hyaline and copper-binding protein (Shikata-positive granules), and periportal cholestasis are all helpful morphologic features in diagnosing PBC. [37] A staging system of 1 to 4 has been used by some; briefly, the histological hallmarks of each of these stages are as follows: Stage 1: destructive changes of interlobular bile ducts with or without granulomas.

Stage 2: proliferation of atypical ductules mostly along the periportal areas.

Stage 3: absence of interlobular bile ducts in most of the portal tracts and portal-to-portal bridging fibrosis.

Stage 4: cirrhosis.

However, in many patients, the biopsies reveal changes overlapping between different stages. [38] Diagnosis of PSC is much more difficult in needle biopsies of liver. A wide range of pathological changes from minimal portal fibrosis and inflammatory reaction, changes of biliary obstruction secondary to extrahepatic sclerosing cholangitis to typical, marked, concentric periductal fibrosis with chronic inflammation can be seen [39] (see Fig. 143-16). Radiologic studies are necessary for definitive

diagnosis.

Drug-Induced Liver Diseases

Liver biopsy interpretation should always consider the possibility of drug-induced changes as differential diagnosis. [40] A careful history of drug intake should be considered prior to final morphologic diagnosis.

Parenchymal Changes Parenchymal changes resembling acute viral hepatitis are seen in both isoniazid- and methyldopa-induced hepatic damage (Fig. 143-83). Changes resembling mononucleosis pattern are seen in diphenylhydantoin-, paraaminosalicylic acid-, and, occasionally, in sulfonamide-induced liver diseases (Fig. 143-84). Coagulative necrosis without much inflammatory reaction is seen in acetaminophen- and halothane-induced damage (see Fig. 143-29 and Fig. 143-30). The necrosis is typically in perivenular and midzonal location. Periportal coagulative necrosis is seen in ferrous sulfate poisoning. Fatty change of microvesicular type is caused by tetracycline and valproic acid. This is accentuated in zone 3 resembling alcoholic FFC. Macrovesicular fatty change is most commonly seen with corticosteroid and methotrexate use. Carbon tetrachloride and tritetrachlorethylene toxicity results in fatty change as well as liver cell necrosis without inflammatory changes. Hepatocyte necrosis with Mallory body formation can be seen with amiodarone, perhexilene maleate, and with synthetic estrogen preparation used for prostatic carcinoma. Parenchymal cholestasis in zone 3 without inflammation or necrosis can be seen with use of anabolic steroids and with oral contraceptives. Cholestasis with mild hepatocellular necrosis is associated with total parenteral hyperalimentation, antibiotics such as erythromycin, penicillin, sulfonamides, nitrofurantoin, oral hypoglycemic agents, tranquilizers such as chlorpromazine (see Fig. 143-48), antihypertensive agents, captopril, gold therapy, and some antineoplastic agents. Some of these drugs such as sulfonamides cause granulomatous necrosis in addition to cholestasis. Rare cases of allopurinol-induced submassive hepatic necrosis have been reported. Morphologic patterns of CAH can result from hepatotoxicity from nitrofurantoin, methyldopa, oxyphenisatin, dantrolene, papaverine, and, occasionally, isoniazid. A number of drugs can elicit granulomatous reactions in the liver. Phenylbutazone and sulfonamides cause granulomas resembling sarcoidosis, either portal or parenchymal. Allopurinol, Dilantin, quinidine, nitrofurantoin, methyldopa, Pronestyl, and, rarely, isoniazid can cause a granulomatous reaction, consisting of punched out areas of hepatocytolysis replaced by hyperplastic Kupffer cells, lymphocytes, and other inflammatory cells in a circumscribed fashion resembling a granuloma. Among the drugs causing hepatocyte inclusions, ground-glass appearance can be caused by cyanamide and proliferation of smooth endoplasmic reticulin (SER) resulting in similar appearance by phenobarbital, chlorpromazine, and Dilantin. [41]

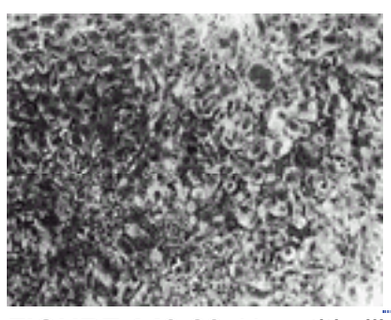

FIGURE 143-83. Hepatitis-like activity resembling acute viral hepatitis in Aldomet-induced hepatotoxicity. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

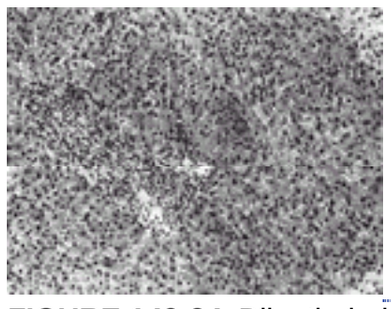

FIGURE 143-84. Dilantin-induced hepatic changes resembling mononucleosis. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Portal Tract Changes Oral hypoglycemic agents and tranquilizers mentioned previously, as well as allopurinol, can cause variable degrees of portal inflammation and bile duct epithelial abnormalities, resembling mechanical duct obstruction or PBC. Dilantin toxicity results in a mononucleosis pattern along with portal inflammatory reaction with predominance of eosinophils (see Fig. 143-2; see also Color Fig. 143-2.). Allopurinol causes portal eosinophilia with cholangitic reaction.
Vascular Changes These include sinusoidal dilation in oral contraceptive agents, anabolic steroids, and in azathroprine therapy and peliosis hepatis seen in anabolic steroid and methyl testosterone treatment (Fig. 143-85). Venous outflow occlusion with its hepatic morphologic changes can be seen as follows: perivenular sclerosis with sinusoidal collagenosis resembling alcohol is associated with hypervitaminosis A, methotrexate (Fig. 143-86), azathioprine, and certain antineoplastic drugs, such as mitomycin C. Venoocclusive disease with coagulative necrosis and severe congestion in zone 3 is seen in toxicity due to bush tea containing pyrrolizidine alkaloids as well as 6-thioguanine and azathioprine. Hepatic vein thrombosis leading to Budd-Chiari syndrome has been associated with prolonged use of oral contraceptives (see Fig. 143-25).

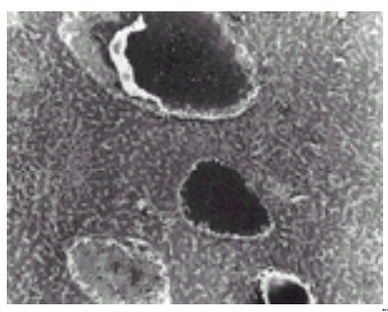

FIGURE 143-85. Peliosis hepatis with blood-filled spaces without endothelial lining. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

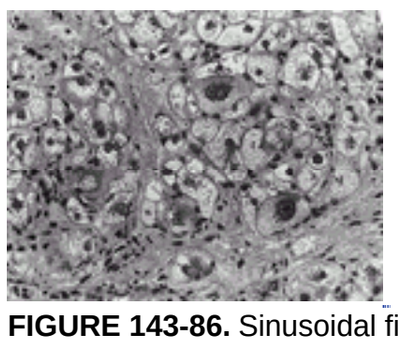

FIGURE 143-86. Sinusoidal fibrosis and nuclear dysplastic changes in methotrexate toxicity. (H & E stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Liver Biopsy Changes in Systemic Diseases

Hematopoietic System Lymphomas with hepatic involvement reveal monomorphic portal infiltrates of neoplastic lymphocytes (Fig. 143-87). Bile ducts and parenchymal cells are not involved. On the other hand, in Hodgkin disease, the portal infiltrate, including atypical or typical Reed-Sternberg cells, seems to produce destructive lesions of bile ducts (Fig. 143-88). In addition, epithelioid granulomas of nonneoplastic nature are seen in both portal and lobular zones. Cholestasis can also be a prominent feature in Hodgkin disease. Liver biopsies in leukemias reveal immature neoplastic white cells in the sinusoidal spaces as well as in the portal zones (see Fig. 143-45). Hairy cell leukemia exhibits typical leukemic cells in the sinusoidal spaces as well as infiltrating the parenchyma. Sickle cell disease can be diagnosed by identifying clumped red cells as well as sickled cells in the sinusoidal spaces (see Fig. 143-47; see also Color Fig. 143-47). Disseminated intravascular coagulation manifests as fibrin thrombi within the sinusoidal spaces.

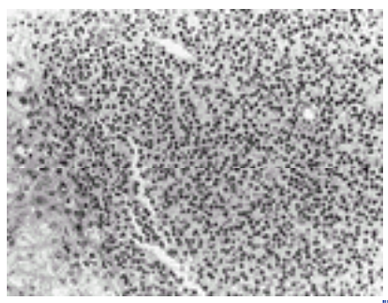

FIGURE 143-87. Portal infiltrate in non-Hodgkin lymphoma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

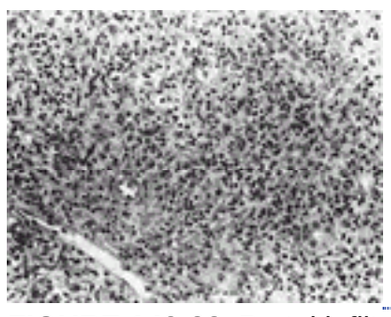

FIGURE 143-88. Portal infiltrate in Hodgkin lymphoma with an atypical Reed-Sternberg cell (*arrow*). (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Acquired Immunodeficiency Syndrome (AIDS) Liver biopsies in patients with AIDS should be examined for evidence of opportunistic infections commonly seen in these patients. In general, the portal areas are relatively lymphopenic, in spite of the inflammatory processes in the liver [42] (Fig. 143-89). *Mycobacterium avium intracellulare* (MAI) infection in these patients typically results in numerous clusters of bloated Kupffer cells, appearing foamy and containing numerous acid-fast stainable organisms (see Fig. 143-64). No true granulomas are seen. Similarly, fungal infections such as cryptococcosis reveal lack of granuloma formation in the presence of organisms identified by special stains and cultures. Viral inclusion bodies of CMV and herpes simplex involving either the reticuloendothelial cells or duct epithelium cells, or parenchymal cells can be seen. Cryptosporidiosis involving the biliary system can be identified by examining the luminal surface of the duct epithelial cells using special stain such as Giemsa (Fig. 143-90). Sclerosing cholangitis identified in patients with AIDS has been described as a sequela of CMV and cryptosporidiosis of the biliary system. [43] In patients with Kaposi sarcoma elsewhere in the body, liver biopsy can also exhibit the presence of the tumor [44] (Fig. 143-91). Since these patients are on a number of drugs on a long-term basis, hepatotoxic changes due to the drugs should also be considered in the differential diagnosis.

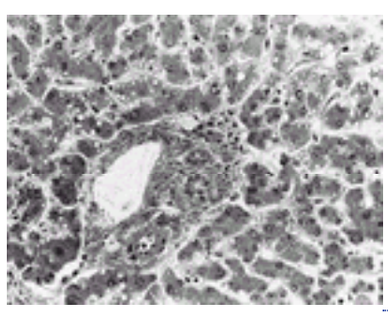

FIGURE 143-89. Portal area with lymphopenia in a patient with AIDS. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

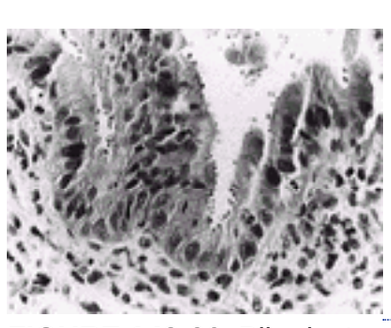

FIGURE 143-90. Bile duct epithelium along the luminal surface demonstrates the presence of cryptosporidiosis of 3- to 4-µm size. (H & E stain; original magnification ×400.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

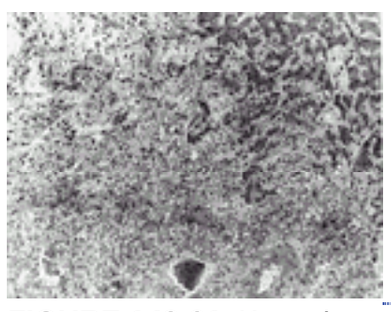

FIGURE 143-91. Kaposi sarcoma involving the liver. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed.

Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

PATHOLOGY OF TRANSPLANTATION

Allograft Rejection

Acute rejection is a more common event seen in varying degree in 70% of allografts; histological characteristics as described by Snover and colleagues [45] include portal inflammatory infiltration, inflammation and destructive changes of interlobular bile ducts, and endothelialitis involving hepatic and portal venous structures (Fig. 143-92). Portal inflammation consists of activated lymphocytes or “immunoblasts,” neutrophils, and eosinophils. These cells are seen infiltrating the epithelial cells of interlobular bile ducts. In addition, the duct epithelium reveals pyknosis of the nuclei, hydropic changes, and disruption of basement membrane. Endothelialitis is exhibited as lymphocytes attached to the venous endothelium and infiltrating through the subendothelium. The hallmark of chronic rejection consists of progressive destruction and eventual loss of bile ducts [46] (Fig. 143-93). In addition, there is perivenular cholestasis and multifocal hepatocytolysis. There is also evidence of obliterative vascular lesions involving hepatic arteries and arterioles. Chronic rejection is less common than acute rejection and occurs in about 4% of graft recipients.

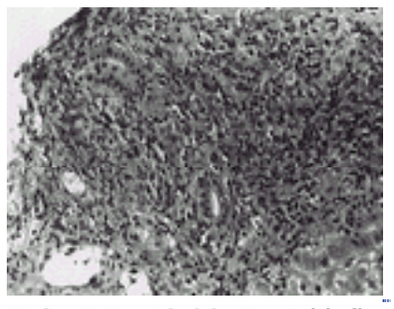

FIGURE 143-92. Portal inflammatory infiltrate, changes of interlobular bile ducts and endothelialitis of portal vein radicles in acute rejection. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

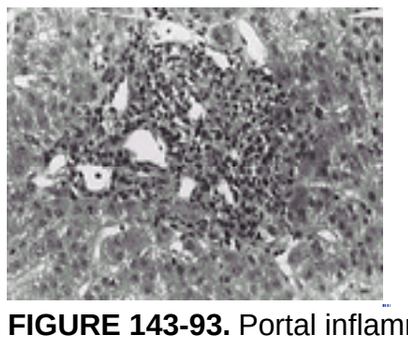

FIGURE 143-93. Portal inflammatory infiltrate with loss of bile ducts in chronic rejection. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Harvest or Preservation Injury

In the majority (54%) of orthotopic hepatic grafts, there is marked ballooning of the hepatocytes in acinar zone 3 (Fig. 143-94). This usually occurs in the first 2 weeks and in some patients as early as 48 hours. This is often associated with cholestasis and high serum ALT levels. Ischemia is considered one of the pathogenetic factors. Ballooning without associated necrosis has better prognosis and is not a sign of graft rejection. Among the other features included under preservation injury are microvesicular fatty change and mild acidophilic degeneration of the hepatocytes.

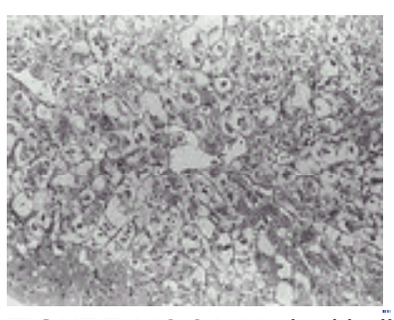

FIGURE 143-94. Marked ballooning of the hepatocytes in acinar zone 3 representing harvest injury. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

CMV Hepatitis

CMV hepatitis in allograft is often associated with parenchymal microabscesses seen as small aggregates of neutrophils in the hepatic lobule. Infected cells have large eosinophilic intranuclear inclusions. Immunoperoxidase stains demonstrate these inclusions clearly.

Recurrent Hepatitis B in Allograft

Liver allograft recipients who had chronic hepatitis B with cirrhosis invariably develop recurrence of hepatitis B infection of the graft. The recurrence usually develops after more than 3 months but can occur earlier in association with an accelerated course. The recurrent HBV infection is similar to that seen in nonliver graft patients. The acute process consists of lobular disarray, ballooning hepatocytes, with necroinflammatory changes (Fig. 143-95). Portal areas reveal inflammatory hyperplasia without any evidence of bile duct epithelial changes or vascular endothelial damage. At this early stage, HB $_{C}$Ag is demonstrable in the hepatocytes by immunoperoxidase stains. Although the acute events can be self-limited, viral infection is never cleared and invariably leads to chronic infection, with progressive increase in the expression of viral antigens in the liver. A unique variant of recurrent hepatitis in the allograft is termed *fibrosing cholestatic hepatitis.* [47] The characteristic pathological changes include periportal fibrosis with thin strands of pericellular collagen, cholestasis, and ballooning of hepatocytes with apoptotic necrosis. There is abundance of HB $_{S}$Ag and HB $_{C}$Ag demonstrable in hepatocytes. Lymphocytic infiltrates are minimal. This morphology has been associated with poor outcome. Development of chronic hepatitis and cirrhosis is the eventual outcome.

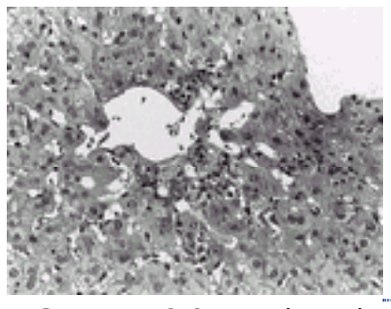

FIGURE 143-95. Perivenular zone with necroinflammatory changes in recurrent acute hepatitis B infection of allograft. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.)

Recurrent Hepatitis C

Reinfection of allografts is universal but the disease progression varies widely. Acute changes can be seen as early as 80 to 90 days. They include sinusoidal lymphocytosis, acidophic bodies, and lobular disarray. Portal lymphoid aggregates, progressive fibrosis, and changes similar to chronic HCV in nontransplant setting are seen in later stages. A small percentage (2%–10%) of patients with recurrent HCV develop severe fibrosing cholestatic hepatitis similar to that seen in recurrent HBV. [48]

Graft-Versus-Host Disease

Graft-versus-host disease is seen in association with bone marrow transplantation and less often with other solid organ transplantation. [49] In the acute stage, there is evidence of hepatocellular necrosis and endothelialitis. In chronic stages, there is extensive bile duct damage and the interlobular bile duct epithelium shows cytoplasmic vacuolization, nuclear hyperchromasia, and atypia. Destructive bile duct changes resemble PBC.

SUMMARY

Needle biopsy of liver represents a minute sampling of a large organ and, understandably, errors of sampling can lead to a negative conclusion. This is true in cases of focal lesions, such as tumors, and in chronic liver diseases, such as PSC or PBC with wide variations of morphologic changes within the liver. Ultrasound or CT scan-guided liver biopsies provide a direct sampling of localized lesions. The type of needle used can alter the size and amount of tissue depending on the degree of fibrosis or cirrhosis present. A Tru-cut needle is more helpful in obtaining an intact, nonfragmented sample in cases of advanced cirrhosis.

In a study of hepatic pathology, histological changes of biopsy sections should be recognized and an objective morphologic diagnosis should be made prior to examining the clinical and the laboratory data. However, it is much more difficult to arrive at an etiologic diagnosis without such data. Final interpretation of liver biopsy should include a correlation of these data to provide a meaningful pathological diagnosis.

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CHAPTER 144

John D. Long and Ralph A. Giannella

DIAGNOSTIC TESTS IN GASTROINTESTINAL INFECTIONS*

This chapter reviews the major tests generally used for microbiologic diagnosis in North America, provides guidelines for their use and interpretation in the evaluation of specific clinical syndromes, and discusses their limitations and cost-benefit issues. Nucleic acid hydridization and reverse transcriptase–polymerase chain reaction (RT-PCR) assays have been developed for most bacterial, protozoal, fungal, and viral pathogens. They are, in general, both sensitive and specific. These assays, however, have not yet found their way into common usage in hospital laboratories. The concept and technical aspects of these assays are discussed in Chapter 135.

MICROBIOLOGIC TECHNIQUES FOR DIAGNOSING CLASSES OF PATHOGENS

Bacteria

General Considerations Bacterial infections are most commonly diagnosed by either culture of the organisms or by the detection of specific bacterial antigens or toxins. Although microscopy or histopathological techniques may be useful in certain circumstances, several macroscopic observations increase the chances that a bacterial pathogen will be present in a body fluid. For example, the presence of grossly bloody diarrhea, especially if accompanied by fever or abdominal pain, increases the likelihood that a patient has a bacterial infection. [1] In body fluids that are normally sterile, microscopic examination often helps identify the organism. Staining of body fluids for organisms is especially valuable if patients have received antibiotics that might interfere with culture results. Stains are also helpful if a fastidious organism such as a fungus or mycobacterium is a potential etiologic agent (when culture results are likely to be either falsely negative or delayed), or when the contents of an abscess are aspirated. In contrast, microscopic examination of a body fluid that is not normally sterile, such as stool, rarely helps detect specific bacterial pathogens because of the polymicrobial composition.

Culture Bacterial culture usually involves placing an aliquot of the specimen onto solid media for stool or tissue, or into liquid media for blood or ascites. The media are incubated under conditions that favor or distinguish various pathogens. Subsequently biochemical tests and, if necessary, serotyping are performed to identify the genus and species. Bacterial cultures necessitate a minimum of overnight incubation, and for many organisms that infect the gastrointestinal (GI) tract, an additional 2 to 3 days might be required for growth and identification. Once the bacteria are identified antibiotic susceptibility can be determined, often within the next 24 hours. The most frequent bacterial test requested in GI practice is a stool culture, which in North America usually includes techniques to detect *Salmonella, Shigella,* and *Campylobacter jejuni.* [2] Greater than 95% of all clinical microbiology laboratories in the United States test for all three organisms routinely. *Escherichia coli* O157:H7, *Yersinia, Vibrio, Aeromonas* species, and *Plesiomonas shigelloides* are less regularly sought. [2] It is important for the gastroenterologist to know which pathogens will be sought routinely by the laboratory to which the specimen is sent. If clinical or epidemiologic factors suggest other pathogens, this should be communicated to the laboratory so that more specific optimal media and conditions can be employed. *Salmonella* and *Shigella* are usually detected using MacConkey *Salmonella-Shigella* agar plates or other selective media and an enrichment broth for *Salmonella.* These media are incubated aerobically and potentiate the growth of gram-negative flora while suppressing the growth of gram-positive flora. *Salmonella* and *Shigella* do not ferment lactose within 24 hours and thus do not acidify the media. An indicator dye in the agar fails to turn pink as a result and the colonies are colorless or white. Supplemental biochemical, functional, and serotyping studies are then performed to confirm the genus and assign species, a process that takes an additional 24 hours. *C jejuni* is usually detected using Campy agar plates which contain antibiotics to suppress other flora (for example, cefoperazone/vancomycin/amphotericin B). The culture process for *C jejuni* requires a microaerophilic environment of 5% oxygen. In addition the agar plates are incubated at 42°C rather than at 37°C. *E coli* 0157:H7, the most frequently isolated member of the enterohemorrhagic *E coli* (EHEC) group in the United States, can be distinguished from most other *E coli* because this serotype either slowly ferments sorbitol or does not ferment it at all. Thus *E coli* 0157:H7 are identified on sorbitol-MacConkey agar, on which they appear as colorless colonies. These colonies are then serotyped with commercially available antisera. [3] The yield of *E coli* 0157:H7 in North America equals or exceeds the yield for other classic bacterial enteric pathogens, yet in one large survey only 34% of microbiology laboratories in the U.S. test for this pathogen as part of a routine stool culture. [2] Other strains of *E coli* (enterotoxigenic, enteropathogenic, enteroinvasive, and enteroadhesive) cause acute diarrhea, but are not generally sought as the assays are only available in research laboratories. *Yersinia enterocolitica* and *Yersinia pseudotuberculosis* can be detected in stool specimens using a selective media containing cefsulodin, irgasan, and novobiocin (CIN). These agar plates need to be incubated between 25°C and 35°C. To detect *Vibrio* species, stool specimens are plated on thiosulfate-citrate-bile salt-sucrose (TCBS) agar. TCBS agar can distinguish *Vibrio cholerae* from noncholera vibrios, such as *Vibrio parahaemolyticus.* After the initial isolation on TCBS agar, biochemical and serologic tests are then performed to confirm the species and serotype. Detection methods for *Aeromonas* species and *P shigelloides* are not well established. *Clostridium difficile,* the predominant cause of antibiotic-associated pseudomembranous colitis, is recovered from stool specimens by the use of selective media such as cycloserine, cefoxitin, and fructose (CCFA) agar. Incubation is done under anaerobic conditions at 35°C and takes 48 to 72 hours. [4] Culture for *C difficile* has largely been replaced by assays to detect *C difficile* toxins in stool (see below).

Bacterial Antigens and Toxins Tests for detection of bacterial antigens and toxins provide several advantages over conventional culture techniques for certain pathogens, including increased sensitivity and much quicker time to diagnosis since most can be performed directly on stool specimens obviating the need for culture. These techniques are mainly applied to detecting infections with *E coli* O157:H7 and *C difficile.* [5], [6], [7], [8], [9], [10], [11], [12] and [13] Specific techniques employed include direct immunofluorescence, latex agglutination, enzyme immunoassays, and cytotoxicity assays. Immunofluorescence techniques involve application of labeled antibodies specific for cell wall products to prepared stool samples and identification of the organism using a fluorescent microscope. Two techniques, the latex agglutination test (LAT) and enzyme immunoassay (EIA), use latex strips or microtiter wells, respectively, which are coated with either monoclonal or polyclonal antibodies against the organism's cell wall antigens or toxins. Processed stool samples are then applied to the strips or wells. In LAT, a positive test results if agglutination of the bacteria is noted. In EIA the wells are incubated for 2 to 3 hours with antibodies conjugated with an enzyme (usually either horseradish peroxidase or alkaline phosphatase). Subsequently a substrate and a chromogen are added and if the antigen or toxin is present, a color change will result. The color change can be detected either visually or with a spectrophotometer. Cytotoxicity assays involve addition of processed stool samples to microtiter wells containing monolayers of mammalian cell lines such as MRC-5 (lung fibroblasts). These monolayers often require incubation for 24 to 48 hours to demonstrate the cytopathic effect, which is usually cell rounding. The assay is considered positive if the cytopathic effect is neutralized by antitoxin. The most common method for diagnosing *E coli* O157:H7 infection is the growth of sorbitol-negative colonies on sorbitol-MacConkey agar. *E coli* O157:H7 can also be identified by a direct immunofluorescence in less than 2 hours with 100% sensitivity and 99.7% specificity. [5] However this test is limited by its inability to detect non-O157:H7 strains that produce the Shigalike toxins Stx1 and Stx2, which are the primary mediators of disease associated with this infection. Various EIAs which rapidly detect these toxins are commercially available and have sensitivities from 82.4% to 100% and specificities of 99.7% to 100%. [6], [7] and [8] The gold standard in the U.S. for diagnosing *C difficile* has become the cytotoxicity assay which identifies toxin B and carries a sensitivity of 67% to 99%. [14] Early studies using an LAT to detect toxin A revealed sensitivities of 48% to 67% and specificities of 96% to 97%. [9], [12] Subsequently the specificity of the LAT was found to be as low as 70% to 80%. Commercially available EIAs to detect toxin A require 2 to 3 hours to complete and carry sensitivities of 64% to 99% and specificities of 93% to 100%. [9], [10], [11], [12] and [13] Unfortunately due to the low sensitivities of some of these assays, the ability to detect *C difficile* infection may be compromised if only one assay is used. [14] A negative EIA should not deter the clinician from making a

diagnosis of *C difficile* colitis and treating the patient in whom the clinical likelihood is very high.
Histopathology Histological examination of colonic biopsy specimens usually will not provide a specific bacterial diagnosis. However, histological features may be helpful in distinguishing an acute self-limited (i.e., infectious-type) colitis from a chronic colitis such as inflammatory bowel disease. In acute self-limited colitis crypt architecture is usually normal, and lamina propria inflammation is predominantly superficial and acute (polymorphonuclear cells) (Fig. 144-1). In contrast, inflammatory bowel disease, especially ulcerative colitis, is characterized by distorted crypt architecture, even early in the course of disease, and a lamina propria that contains acute and chronic (lymphocytes and plasma cells) inflammatory cells [15] (Fig. 144-2). Basilar plasmacytosis and lymphoid hyperplasia and aggregates are other histological features that are more frequent in inflammatory bowel disease and less common in acute self-limited colitis. Epithelial granulomas usually indicate Crohn's disease but can also be present in other less common conditions.

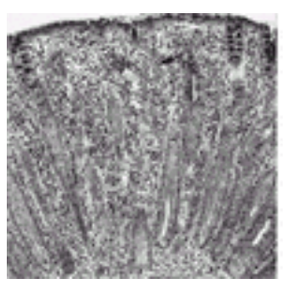

FIGURE 144-1. Biopsy specimen of colorectal mucosa from a patient with acute, self-limited, infectious-type colitis caused by *Campylobacter jejuni.* Notice the preserved crypt architecture, acute lamina propria inflammation, and cryptitis. (From Tarr PI, Surawicz CM, Clausen CR. Microbiologic studies. In: Yamada T, Alpers DH, Laine L, Owyang C, Powell DW, eds. Textbook of gastroenterology, 3rd ed. Philadelphia: Lippincott Williams & Wilkins, 1999.)

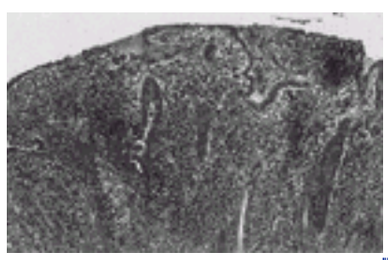

FIGURE 144-2. Biopsy specimen of colorectal mucosa from a patient with ulcerative colitis. Notice the distorted crypt architecture, chronic lamina propria inflammation, and basilar plasmacytosis. These histological features are typical of inflammatory bowel disease and rare in acute self-limited colitis. (From Tarr PI, Surawicz CM, Clausen CR. Microbiologic studies. In: Yamada T, Alpers DH, Laine L, Owyang C, Powell DW, eds. Textbook of gastroenterology, 3rd ed. Philadelphia: Lippincott Williams & Wilkins, 1999.)

Infections with *E coli* O157:H7 and *C difficile* may demonstrate certain histological features that support the diagnosis. For example, colitis from *E coli* O157:H7, which is often right-sided, may mimic ischemia clinically and demonstrate superficial necrosis histologically. [3] Colitis associated with *C difficile* is commonly associated with pseudomembranes which can be visualized on endoscopy but also demonstrated on biopsy by the presence of a layer of fibrin, neutrophils, and cellular debris emanating or erupting from the mucosa. Pseudomembranes may be present even in milder cases in which the mucosa is uninflamed. [4]

Protozoan and Parasites

General Considerations Modern air travel enables patients to acquire exotic infections that might never be considered as causative agents in the patient's home community. In this section the term *parasite* is used to include protozoan. Laboratory identification of parasites from stool specimens depends on observer competence and experience, which may be quite variable in different laboratories. Furthermore, the finding of a parasite or protozoan in a stool sample does not incriminate it as a cause of the patient's symptoms. Although parasitic infections are common, the range of diagnostic tests is limited and consists of examination of body fluids and tissues for ova, parasites, and parasitic antigens, and blood for serologic tests. Parasites are in general not identifiable by culture techniques.
Body Fluids Various stages of the life cycles of parasites can be detected by directly examining wet mounts or stains of fresh stool. A complete evaluation of stool for ova and parasites (O & P) should include an examination of both fresh and fixed specimens. Trophozoites are more likely to be detected in diarrheal or semisolid stools, whereas cysts may be detected in solid stools. If a stool specimen cannot be examined and processed immediately after collection, appropriate fixatives should be used including 10% formalin, polyvinyl alcohol, or merthiolate-iodine-formaldehyde. [16] Barium or bismuth may interfere with visualization by such methods and thus should not be administered to the patient for at least one week before examination. *Giardia lamblia* is most commonly diagnosed by the identification of cysts or, less frequently, trophozoites in fecal specimens that are stained with trichrome or iron hematoxylin. [17] Stool samples should be concentrated by formalin-ethyl acetate or zinc sulfate methods. The cysts are oval in shape and in general 11 to 15 μm in size. If motile trophozoites are detected by a direct examination of fresh stool using a wet mount, this may correlate better with symptomatic giardiasis. At best these methods, which depend largely on observer experience, are only 60% to 70% sensitive in confirming a diagnosis of giardiasis. [17] In patients with chronic diarrhea due to giardiasis, the results of stool examinations may be repeatedly negative despite high suspicion of this parasite. In such cases it may be necessary to sample the lumenal contents of the small bowel, obtained by either the string test or duodenal aspiration. For the string test, the patient swallows a capsule on the end of a string which moves to the jejunum where the trophozoites attach. After a minimum of 4 hours, the string is withdrawn and inspected for trophozoites. [17] Duodenal aspiration can also be accomplished during an upper endoscopy. Direct examination of unstained stool is not useful for the identification of some protozoa such as *Cryptosporidium, Isospora, Cyclospora,* and microsporidia, since these organisms are too small to be noted. The gastroenterologist should be aware of the practice of the laboratory when ordering a routine O & P examination, as many laboratories do not routinely perform the special staining techniques needed to detect these pathogens. *Cryptosporidium, Isospora,* and *Cyclospora* can be readily identified by a modified acid-fast stain. [18] The oocysts of *Cryptosporidium* are 2 to 6 μm in size, *Cyclospora* 8 to 10 μm, and *Isospora* 20 to 30 μm. Microsporidia spores can be identified in fecal specimens but they are hard to differentiate from bacteria and debris. Microsporidia spores can be identified using a modified trichrome stain. [19] *Entamoeba histolytica* can be diagnosed in fecal specimens by the identification of trophozoites using wet mounts or the trichrome stain. However traditional wet mount and stains are at best 60% sensitive for identifying *E histolytica,* mainly because this organism is identical in appearance to *Entamoeba dispar,* a nonpathogenic commensal protozoan. [20] Hematophagous amoebas (i.e., contain ingested red blood cells) provide better but not definitive proof of *E histolytica* infestation.
Parasite Antigens Some intestinal parasitic infections may escape detection despite multiple direct examinations of stool. To avoid the need to perform endoscopy for diagnosis in these patients, rapid antigen detection tests have been developed which can directly detect the parasite or parasite-specific molecules in the stool. Many antigen detection tests have been developed to diagnose *Giardia lamblia* and *Cryptosporidium parvum.* [21], [22], [23], [24], [25], [26], [27], [28], [29], [30] and [31] Direct immunofluorescence tests of fecal specimens have more than 99% sensitivity and 100% specificity in identifying *G lamblia* and more than 93% sensitivity and 100% specificity in identifying *C parvum*. [23], [24], [25], [26] and [27], [30] There are several commercially available EIAs that are useful for the diagnosis of *G lamblia* infection. All are directed at detecting the presence of a *Giardia*-specific antigen (GSA) that is present on both cysts and trophozoites. [17] These assays have more than 90% sensitivity and more than 96% specificity in identifying *G lamblia*. [21], [22], [23] and [24], [27], [28] Similarly, there are also several EIAs that can be used to diagnose infection with *C parvum;* these are directed at detecting the presence of a *Cryptosporidium*-specific antigen (CSA) that is present on oocysts. [31] These assays have more than 83% sensitivity and more than 96% specificity in identifying *C parvum*. [27], [28], [29], [30] and [31] As noted, distinguishing *E histolytica* from *E dispar* is difficult using conventional O & P examination of stool specimens. Only 10% of people with amebiasis suffer from symptomatic, invasive disease, whereas 90% are asymptomatic cyst passers. [20] Therefore, it is important to know whether the amoeba on a conventional O & P examination is truly *E histolytica.* One rapid EIA is available that can distinguish *E histolytica* from *E dispar* with a 95% sensitivity and 93% specificity. [32] Polymerase chain reaction (PCR) techniques, however, are the most sensitive ways to distinguish these two amoeba. [33]
Serology The local availability of parasite serologic tests varies, although most tests can be performed at the Center for Disease Control in Atlanta or in private reference parasitology laboratories. However, in many situations physicians may not be able to wait for the result of a serologic study before initiating medical or surgical therapy. Also, the clinician must determine the likelihood that a positive result explains the symptoms being evaluated. For example, the finding of antibodies to *E histolytica* in a patient with colitis or a liver abscess is helpful in confirming the presence of invasive amebiasis, [20] while the finding of antibodies to *Echinococcus* in a patient with a liver cyst might help avoid needle aspiration, which is contraindicated and could lead to anaphylaxis. [34] In contrast, the finding of antibodies to *E*

histolytica in an asymptomatic person or in a patient who has lived for many years in the tropics may not be as helpful. Negative results for parasite serologic study can be helpful in excluding *E histolytica* or *Echinococcus* in the patient with a liver abscess or cyst. Several EIAs are available for the detection of serum antibodies to *Strongyloides stercoralis.* Sensitivities vary from 80% to 90% and specificities from 90% to 100%. [35]

Histopathology Mucosal biopsy may aid in the diagnosis of parasitic infections when the stool examination or serology is negative. Parasites that may be detected by small intestinal biopsy include *G lamblia, C parvum, Isospora belli,* microsporidia, and *S stercoralis.* Parasites that can be detected by colonic biopsy include *E histolytica, S stercoralis,* and *Schistosoma* species. *G lamblia* trophozoites, which are pear-shaped, can be present on the lumenal surface of the small intestine (Fig. 144-3). *G lamblia* does not invade the mucosa and does not usually cause inflammatory or structural changes. However, in individuals who are immunodeficient, small bowel mucosa may show flattening of villi and chronic lamina propria inflammation. *Cryptosporidium* oocysts are also only present on the lumenal surface of enterocytes and on stained specimens appear as round, blue dots. [18] *C parvum* may be associated with blunting of the villi and increased eosinophilia of the lamina propria, especially in cases of massive infection. The oocysts of *I belli,* which are flask-shaped, can only be detected within the enterocytes (as inclusions). [18] Giemsa stains help identify these oocysts. Finally, microsporidia can be detected in biopsy specimens of the small bowel using routine light microscopy, although these are best detected on electron microscopy. [18] Microsporidia can be detected as inclusions at the apical surface of enterocytes.

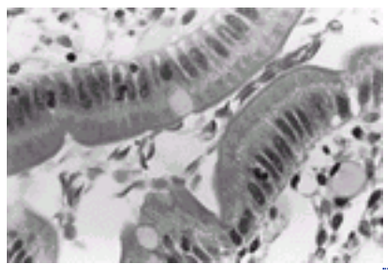

FIGURE 144-3. Biopsy specimen of duodenal mucosa from a patient with giardiasis. Notice the trophozoites of *Giardia lamblia* lining the lumenal surface of the enterocytes. (H & E stain; original magnification ×400). (Courtesy of Amy Noffsinger, M.D., Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine.)

E histolytica can be detected on mucosal biopsy specimens of the colon. The cecum, ascending colon, and rectosigmoid are most commonly involved. [36] The organisms are usually found in the superficial areas of ulcerated mucosa, such as in the surface exudate, but can also be found in the mucosa and submucosa (Fig. 144-4). The trophozoites are usually large in size (10–30 µm) and have foamy cytoplasm with small, round nuclei. They are best detected through biopsy of the shallow, flask-shaped ulcers caused by amebiasis. In general these ulcers occur over lymphoid follicles and are associated with minimal inflammatory reaction.

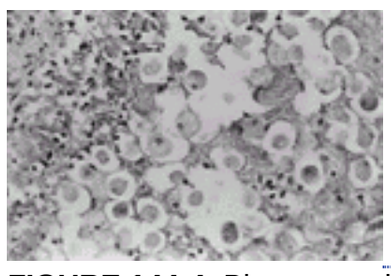

FIGURE 144-4. Biopsy specimen of colorectal mucosa from a patient with amebic colitis. Notice the abundant trophozoites of *Entamoeba histolytica* in the surface mucus. (H & E stain; original magnification ×400). (Courtesy of Amy Noffsinger, M.D., Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine).

Both schistosomiasis and strongyloidiasis can be diagnosed on colonic biopsy. *Schistosoma* ova can be detected and can produce foreign body granulomas. *S stercoralis* causes chronic inflammation and both the adult worms and oocysts can be often be detected in the mucosa. [37]

Mycobacteria

Only 20% of cases of *Mycobacterium tuberculosis* involving the GI tract are associated with active pulmonary tuberculosis. [38] In contrast, over 75% of patients with disseminated mycobacterium avium complex (MAC) will have GI or hepatic involvement. [39] *M tuberculosis* involves the jejunum, ileum, or cecum in more than 75% of cases, whereas MAC involves primarily the duodenum. [38], [40]

Smear and Culture *M tuberculosis* is rarely diagnosed without smear or cultures of mucosal biopsy specimens. [41] The highest yield for such an evaluation is obtained by colonoscopy, since the cecum and terminal ileum are commonly involved. Smears are usually examined by use of an acid-fast stain (e.g., Ziehl-Neelsen) but in general are very insensitive. Cultures of mucosal biopsy specimens using Lowenstein media are more sensitive but take 4 to 6 weeks for results. In the patient with human immunodeficiency virus (HIV) with fever and chronic nonbloody diarrhea, disseminated MAC is most readily diagnosed by blood or bone marrow culture. [38] A positive result may avoid an invasive GI evaluation. In some cases, however, stool, or endoscopic investigations may be required. Smears and mycobacterial culture of stool specimens have a moderate (approximately 70%) yield for MAC.

Histopathology The classic histological features of GI *M tuberculosis* infection include epithelioid granulomas with Langhans giant cells. [38] Central caseation is also a helpful sign, but is more often absent on biopsy because it is more likely found in surrounding lymph nodes (which are not sampled in a biopsy). These histological signs are present in less than 50% of definitive cases of *M tuberculosis* diagnosed by other means (e.g., culture). MAC infection is most apparent in biopsy specimens obtained from the duodenum. The typical features include the presence of multiple foamy macrophages located in the intestinal villi, an appearance similar to that of Whipple disease. [40] In MAC infection, granulomas are rarely observed.

Fungi

Fungal infections should be considered in the differential diagnosis of esophagitis, hepatic masses, and rarely diarrheal syndromes. [42], [43] and [44] In particular infection with a fungus should always be considered in immunosuppressed individuals. The identification of a commensal fungus, (e.g., *Candida*) from a fluid or tissue that is not normally sterile, in the absence of gross or histological evidence of abnormality, should be interpreted cautiously. In contrast, the isolation of a fungus from a fluid or tissue that is normally sterile, such as blood, should always be taken seriously. Viral infections such as herpes simplex virus (HSV) and cytomegalovirus (CMV) can coexist with a fungus, thus the presence of a mucosal fungal infection should not stop the search for additional viral pathogens. Because fungal infections may require prolonged therapy with toxic agents, every effort should be made to establish the specific diagnosis.

Smear and Culture A smear of the characteristic exudates of mucosal fungal infections, the most common being *Candida albicans,* are obtained usually by endoscopic brushing. Stains such as Gram stain, potassium hydroxide, and periodic acid-Schiff (PAS) are used to identify the fungus. Fungal cultures are obtained by placing the appropriate specimen on growth media. The rapidity with which positive cultures are reported usually depends on the size of the inoculum and the inherent growth characteristics of the specific fungus.

Histopathology *Candida esophagitis* is the most frequent fungal infection of the GI tract encountered in immunocompromised patients. In esophageal mucosal biopsies, pseudohyphae can be seen on routine light microscopy of hematoxylin and eosin (H & E) stained sections [42] (Fig. 144-5). Silver or PAS stains can improve detection in certain situations. The finding of a granuloma on histological examination suggests the presence of a fungal infection such as histoplasmosis and blastomycosis. Additionally in histoplasmosis, the lamina propria may be filled with macrophages containing the organism in the cytoplasm. [45] With appropriate staining, a specific identification based upon the morphology of the fungus can be made.

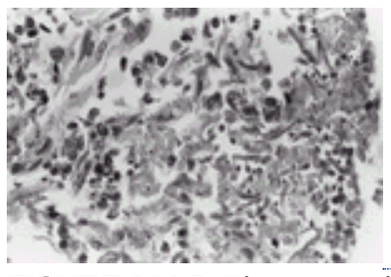

FIGURE 144-5. Biopsy specimen of esophageal mucosa from a patient with candidiasis. Notice the pseudohyphae and yeast forms present in the inflammatory exudate. (H & E stain; original magnification ×400). (Courtesy of Amy Noffsinger, M.D., Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine.)

Viruses

General Considerations A number of modalities are available for diagnosis of viral infections of the GI and hepatobiliary systems. These include viral growth in cell culture, detection of viral antigens in serum or stool, serology, electron microscopic imaging of the virus particle, nucleic acid hybridization technology, or demonstration of characteristic histological features or specific antigens in biopsy specimens.
Cultures Viral cultures of body fluids are only variably helpful in diagnosing GI infections. Virus isolation from stool, which is not normally sterile, is not useful in diagnosing acute enteric infection. The isolation of viruses from fluids and tissues that are normally sterile is more useful. For example, the recovery of CMV from blood or urine might suggest systemic CMV infection that can be associated with GI and hepatic manifestations. Viral cultures are more commonly obtained on biopsy and brushing specimens of mucosal ulcers in immunocompromised patients. The specimen should be placed in sterile viral transport medium which contains both antibacterial and antifungal agents that inhibit the growth of contaminants. Media that are supplied frozen should be kept frozen until immediately before use, but some commercially available viral transport media do not require frozen storage. A conventional viral culture entails inoculation of tissue culture cells. [46] This technique frequently uses a monolayer of cells in a shell (centrifuge) vial. Viral cultures are usually incubated for several weeks after inoculation before issuing a final negative result. HSV, adenovirus, and enteroviruses usually grow within 7 to 10 days of inoculation, while CMV usually grows within 10 to 21 days. One technique that has been developed to shorten the time to diagnosis is the application of specific monoclonal antibodies directed against viral antigens present in the inoculated tissue culture cells. This process may provide a presumptive diagnosis within 48 hours, well before any typical cytopathic effects are seen. [46]

Serology

Determining the presence and the class (i.e., immunoglobulin M or G) of antibodies directed against specific pathogens can be used to characterize whether the infection is acute versus persistent or resolved. In addition, the level of antibodies may be quantified by serial dilution or by enzyme immunoassay. [47] A significant (usually fourfold) rise in antibody levels against a specific virus, as determined by using paired acute and convalescent specimens, is presumptive evidence of acute systemic infection. The absence of antibodies to CMV viral antigens is highly predictive of the absence of mucosal disease as long as the infection has been present long enough to elicit an antibody response and the patient is able to mount an antibody response. Likewise the appearance of antibodies to HSV or CMV antigens in a previously seronegative, recently transfused person should be interpreted cautiously since these antibodies could have been obtained passively from a seropositive donor.

To properly interpret the presence or absence of antibodies to specific viruses, several qualifying circumstances must be considered. A false-positive result may result from a previous infection that has resolved and is thus no longer germane to the patient's symptoms. Other causes of false-positives include a cross-reactive antibody to a different pathogen or the polyclonal activation of the patient's B cells. A false-negative result may be caused by the fact that specific antibodies were either never produced or have not yet been formed.

Imaging by Electron Microscopy Electron microscopy is not commonly used in the clinical setting. Electron microscopic study of stool specimens can be used for diagnosing noncultivatable viruses such as the Norwalk agent or rotavirus, which have distinctive morphologic features that can be recognized. Immune electron microscopic study, which uses antibodies present in convalescent sera to enhance the morphologic features of viruses in stool obtained during the acute illness, is chiefly a research tool.
Histopathology Biopsy diagnosis of viral infection is used most frequently in evaluating esophagitis and colitis, especially in immunosuppressed adults. In immunocompetent individuals, HSV can cause esophagitis as well as a sexually transmitted, self-limited proctitis. Self-limited colitis due to CMV has also been described in immunocompetent individuals. [48] The presence of typical cytomegaloviral inclusions on routine H & E stains of mucosal biopsy specimens is considered the gold standard for diagnosing CMV infection. [49] The CMV-infected cell is large (25–35 µm) with a basophilic intranuclear inclusion surrounded by a clear halo giving rise to an "owl's eye" appearance. Intracytoplasmic inclusions are also present (Fig. 144-6). CMV typically infects endothelial cells, but glandular epithelial cells can also be infected, particularly in the stomach. The surrounding tissue may be normal or may exhibit nonspecific focal or diffuse inflammation or ulceration. Routine histological analysis for CMV, although the gold standard, is unfortunately less than 100% sensitive and in some cases the pathologist may have difficulty finding cells with typical features of CMV. Other methods that have been developed to help detect CMV in biopsy specimens include immunohistochemistry (immunoperoxidase or immunofluorescence methods), [50], [51] and [52] in situ DNA hybridization, [52], [53] and [54] and PCR. [55] All three methods are either as sensitive or more sensitive than light microscopy, with PCR being the most sensitive. [49] The only drawback of the higher sensitivity of PCR is that in patients with documented CMV mucosal lesions; 20% of biopsies taken from normal-appearing areas are positive as well. [55]

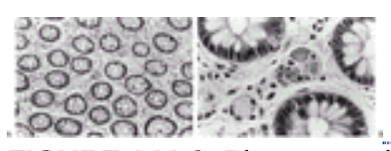

FIGURE 144-6. Biopsy specimen of colorectal mucosa from a patient with cytomegalovirus (CMV) infection. **A:** Low power (original magnification ×100) view showing scattered typical cytomegalovirus inclusions. **B:** Higher magnification (original magnification ×400) of a CMV-infected endothelial cell demonstrating the intranuclear inclusion. (H & E stains.) (Courtesy of Amy Noffsinger, M.D., Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine.)

Light microscopy examination of H & E-stained mucosal biopsy specimens is also considered the gold standard for diagnosing HSV infection since it produces characteristic intranuclear inclusions (Fig. 144-7). The intranuclear, or Cowdry type A, inclusion is smaller than the inclusion associated with CMV infection and thus may be more difficult to detect. Other histological features that suggest HSV infection are the presence of multinucleated giant cells and cells with ground-glass nuclei. [56]

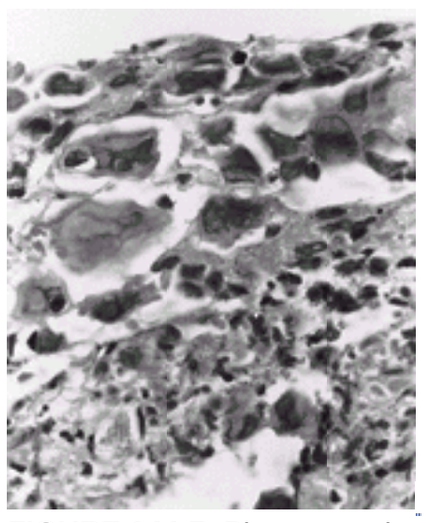

FIGURE 144-7. Biopsy specimen of esophageal mucosa from a patient with herpes esophagitis. Notice the typical viral inclusions of herpes simplex type 2 and an

associated multinucleated giant cell. (H & E stain; original magnification ×400.) (Courtesy of Amy Noffsinger, M.D., Department of Pathology and Laboratory Medicine, University of Cincinnati College of Medicine.)

TECHNICAL ASPECTS OF SPECIMEN COLLECTION

The office and procedure suites of gastroenterologists should be equipped with the materials needed to collect and transport specimens for microbiologic testing. The most important element in specimen collection and processing is rapid transport to the laboratory. Table 144-1 summarizes the materials and other factors required for transport and processing of specimens for each of the classes of pathogens.

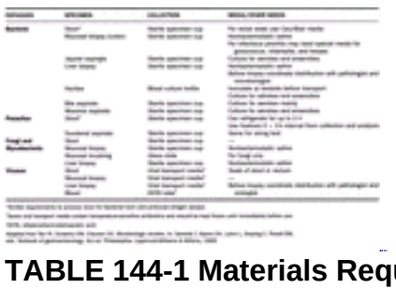

TABLE 144-1 Materials Required for Microbiologic Testing Performed By Gastroenterologists

INDICATIONS FOR MICROBIOLOGIC TESTING

Diarrhea

Acute Diarrhea Most cases of acute diarrhea in immunocompetent adults are self-limited and thus do not require investigation. However, stool cultures are obtained frequently even though their yield is notoriously low: 5.7% in combined data from 10 studies with over 90,000 stool specimens. [2], [57], [58], [59], [60], [61], [62], [63], [64] and [65] Attempts have been made to identify clinical factors that increase the yield of stool cultures. [1] Factors such as abrupt onset of diarrhea, greater than four stools per day, absence of vomiting before the onset of diarrhea, or the presence of fever, bloody diarrhea, tenesmus, abdominal pain, or raw seafood ingestion all increase the likelihood of recovering a bacterial pathogen from a stool culture. Recent American College of Gastroenterology guidelines for the evaluation and treatment of acute infectious diarrhea [1] suggest that bacterial stool culture be limited to patients with:

- severe diarrhea (>6 stools/24 hours, or >48 hours duration of illness, or diarrhea that results in clinical volume depletion)
- temperature >101.3°F
- dysentery (bloody stools)
- stools positive for occult blood, leukocytes, or lactoferrin
- immunosuppression (e.g., acquired immunodeficiency disease [AIDS], transplant, and cancer chemotherapy).

These guidelines also recognize the low yield of routine stool O & P examinations in patients with acute infectious diarrhea and suggest that indications for O & P be limited to:

- patients with acute diarrhea and recent travel to Russia, Nepal, or mountainous regions
- infants attending day care centers
- patients with AIDS or homosexual men
- those exposed to a possible community waterborne outbreak
- patients with bloody diarrhea but few or no fecal leukocytes. [1]

A spot analysis of stool samples for the presence of leukocytes has been advocated as a measure to increase the chance of isolating a bacterial pathogen. [1] Leukocytes may be detected by light microscopy using methylene blue staining. Bacterial pathogens are more likely to be isolated from samples that contain fecal leukocytes, although the absence of leukocytes does not accurately predict the absence of a bacterial pathogen. For example, in a summary of several studies that reported the prevalence of leukocytes in stool samples from patients with a documented bacterial pathogen, leukocytes were detected in 73% of samples with *Shigella,* 52% with *Salmonella,* 58% with *Campylobacter,* 54% with *E coli* O157:H7, and 42% with *C difficile*. [66] These results suggest that analysis of feces by light microscopy for the presence of leukocytes is not sensitive enough to be used as a screening test and thus certainly cannot be relied on to select patients for bacterial culture. Perhaps a more sensitive test to use for screening stool specimens for bacterial pathogens is the fecal lactoferrin assay. [66] Lactoferrin is a glycoprotein found in leukocytes and can be detected with a commercially available LAT. Two studies showed that the sensitivity of a positive lactoferrin assay in detecting *Salmonella, Shigella,* and *Campylobacter* ranged from 83% to 93%. [67], [68] Similarly one study [69] has shown that the lactoferrin assay was a more sensitive marker (75%) of *C difficile* infection compared to fecal leukocytes (40%), but this was not confirmed in a second study. [70] Two viruses are common causes of acute diarrhea, especially in the winter: rotavirus and Norwalk-like agent, in children and adults, respectively. Although EIAs are available to detect both in the stool, they are not generally used in adults as the illnesses are self-limited and no specific antiviral treatment is available. However, in pediatric practice EIAs for rotavirus are commonly used since a positive assay usually obviates the need to search for other causes.

Diarrhea in HIV-Infected Patients Although the usual bacterial pathogens such as *Salmonella, Shigella,* and *Campylobacter* are potential causes of diarrhea in HIV-infected patients, more common pathogens include protozoa (e.g., *Cryptosporidium*), viruses (e.g., CMV), and mycobacteria. [39] The likelihood of detecting an enteric pathogen in HIV-infected patients with diarrhea depends greatly on the CD4 lymphocyte count and whether endoscopy with mucosal biopsy is used in the diagnostic work-up. Guidelines published in 1996 from the American Gastroenterological Association [39] recommend the following evaluation in HIV-infected patients with chronic, nonbloody diarrhea: bacterial culture for enteric pathogens (including *Salmonella, Shigella,* and *Campylobacter*), *C difficile* toxin assay, modified acid-fast stain (to detect *Cryptosporidium* and *Isospora*), modified trichrome stain (to detect microsporidia), and blood culture for bacteria and mycobacteria (if fever is present). For patients in whom this initial evaluation fails to arrive at a diagnosis, especially in those with bloody diarrhea or CD4 counts less than 100, lower endoscopy with mucosal biopsies is recommended. In most cases a flexible sigmoidoscopy (rather than a full colonoscopy) is all that is necessary. To optimize the yield and minimize costs, colonic biopsies should be obtained from endoscopic lesions and not from normal mucosa. In addition, processing of biopsies in these patients should be limited to routine H & E staining since there is little evidence that the use of cultures or special stains increases the identification of viruses, mycobacteria, or fungi. [71] For example, one study of 121 HIV-infected patients who underwent endoscopic biopsy revealed that careful evaluation of H & E stains was 97% sensitive and 100% specific in diagnosing CMV. [71] In the same study, special stains used to identify CMV and HSV (immunoperoxidase), mycobacteria (acid-fast), and fungi (silver or PAS) did not increase the diagnostic yield over routine H & E. In a study of 290 HIV-infected patients with diarrhea who underwent colonoscopy with mucosal biopsies, the yield of culturing biopsy specimens for bacteria, mycobacteria, and fungi was found to be quite low. [72] The spectrum of causes of chronic diarrhea in HIV-infected patients has been changing over the last 5 years, and this trend may eventually impact the diagnostic approach suggested in the American Gastroenterological Association guidelines. [39] The change in spectrum is largely a result of the widespread use since 1996 of protease inhibitors and HAART (highly active antiretroviral therapy). These therapies, which significantly restore CD4 counts and lower viral load, have led to a significant reduction in the incidence of opportunistic infections. [73] For example, in a cohort of HIV-infected patients with chronic diarrhea, the prevalence of opportunistic infections fell from 53% in 1995 to 13% in 1997. [74] During this same time period the number of patients evaluated for chronic diarrhea did not change but those diagnosed with noninfectious causes increased significantly.

Nosocomial Diarrhea Diarrhea that develops in patients after 3 days of hospitalization is classified as nosocomial or hospital-acquired. Nosocomial diarrhea is rarely caused by community-acquired bacterial or parasitic pathogens. For example, 6 studies have compared the yield of bacterial stool culture obtained during the first 3 days of hospitalization (2%–12.6%) versus the yield after 3 days (0–1.5%). [2], [57], [60], [61], [63], [65] The yield in the first 3 days is 2 to 9 times higher. Similar results are obtained with the use of routine O & P examinations in patients with nosocomial diarrhea. [2], [57], [75] Despite this low yield, it has been shown that as much as 50% of all bacterial stool cultures submitted to microbiology laboratories are obtained from patients who have been hospitalized for more than 3 days, and this results in substantial costs per positive diagnosis. These issues have helped formulate the “3-day rule” which recommends that stool samples submitted for routine bacterial

culture be rejected if obtained from a patient hospitalized for more than 3 days. [66] Strict application of such criteria would result in significant reductions in costs through limiting the use of supplies and reagents and decreasing laboratory staff time. Unfortunately, a few cases of clinically important nosocomial diarrhea resulting from these bacterial pathogens could be missed. As a result, a modified 3-day rule has been advocated which allows cultures to be processed after 72 hours of admission in certain high-risk patients (age >65 with significant co-morbid illnesses, HIV infection, or neutropenia) or in cases in which a nosocomial outbreak of salmonellosis is suspected. [76] *C difficile* is the most common nosocomial infection of the GI tract and tests to detect this pathogen should be used in the evaluation of nosocomial diarrhea. Clinical factors which increase the yield of detecting *C difficile* include recent hospitalization (especially if within the last 2 weeks), antibiotic use within 30 days (particularly cephalosporins), the presence of abdominal pain or significant diarrhea, a white blood cell count more than 10,000/mm, and the presence of fecal leukocytes or lactoferrin. [70], [77], [78] Other frequent causes of diarrhea in hospitalized patients include medications, particularly elixirs that contain sorbitol, and tube feedings. In addition, antibiotics may cause diarrhea in the absence of *C difficile.*

Chronic Diarrhea Bacterial infections are rarely the cause of chronic diarrhea in immunocompetent patients. However, *Y enterocolitica, Aeromonas* species, *C difficile, Campylobacter,* and some *E coli* can cause chronic diarrhea. Thus, at least one routine stool culture should be obtained at some point during the evaluation of these patients, especially in patients with an elusive diagnosis in which an initial negative stool analysis did not include a bacterial culture. [79] In general stool should also be sent for routine O & P examination and for a *C difficile* toxin assay. [79] Some patients with chronic diarrhea may be suspected of having small intestinal bacterial overgrowth. Quantitative aerobic and anaerobic cultures of jejunal aspirates are still the most reliable methods to diagnose this syndrome. [79] Greater than 10 [6] aerobic or anaerobic organisms per mL of fluid is considered a positive culture. The most common aerobic bacteria isolated are *E coli* and *Streptococcus,* while the most common anaerobic are *Lactobacillus* and *Bacteroides.* [80] These tests are, however, cumbersome since they require intubation of the small bowel to obtain the fluid. Several noninvasive breath tests have been used in practice to detect small intestinal bacterial overgrowth, including ^{14}C (using radioactive xylose as a substrate) and hydrogen (using glucose or lactulose as a substrate) breath testing. [79] These tests are easy and safe to administer but suffer from poor sensitivity and specificity. [81] In some cases of chronic diarrhea, sampling of duodenal contents occasionally is warranted to diagnose parasitic infections such as giardiasis. The string test can be used to perform this task and can identify *G lamblia* and strongyloidiasis. Alternatively aspiration of duodenal fluid or mucosal biopsy, both accomplished using upper endoscopy, may be required to detect such infections. Other protozoa that can cause chronic diarrhea include *Cryptosporidium* and *E histolytica.*

Inflammatory Bowel Disease Bacterial enteric pathogens (*Campylobacter, Salmonella, Shigella,* EHEC, *Yersinia,* and *C difficile*), amebiasis, and CMV have all been associated with idiopathic inflammatory bowel disease (IBD). These infections may be associated with either the first attack of IBD or relapses in patients with well-established disease. [82], [83] For example, one study reported a 20% prevalence of bacterial pathogens including *C difficile* in first attacks of IBD. [82] In another study of 64 patients with IBD who suffered a relapse and in which stool cultures were obtained, 17% were found to be infected with an enteric pathogen. [83] *C difficile* and sometimes the toxin produced by this organism may be present in stools from patients with IBD, but in most cases *C difficile* does not appear to be a pathogen. However, in a patient with severe colitis or disease refractory to therapy who has cytotoxin in stools, therapy to eradicate *C difficile* should be strongly considered. Stool examination and serologic study for *E histolytica* should be done at least once in all patients with inflammatory bowel disease, particularly in those who lived or traveled in an area where amebiasis is prevalent. [84] CMV may be an important exacerbating factor in inflammatory bowel disease (particularly in patients with ulcerative colitis). Infection with CMV has been reported in patients with new-onset and flares of ulcerative colitis, [85] pouchitis, [86] and severe or refractory ulcerative or Crohn's colitis. [87] Some cases of CMV complicating ulcerative colitis involved patients who had never received steroids or other immunosuppressants. [88] Patients with both conditions often require colectomy and carry a high mortality rate. One case series found CMV on biopsy in 11% of 62 patients with severe colitis and in 37% of those refractory to steroid therapy. [87] Based on this information it seems reasonable to exclude superimposed enteric infections in patients with new-onset IBD. Furthermore, patients with IBD who develop either severe colitis refractory to medical therapy or toxic megacolon (and thus are approaching need for colectomy) might benefit from an evaluation for superimposed CMV infection since ganciclovir may be an effective therapy. [85] It remains to be determined whether it is clinically helpful and cost-effective to exclude a superimposed enteric infection in every patient with a mild-to-moderate relapse of IBD.

Infectious Esophagitis

Infectious esophagitis has been extensively reviewed in this text (Chapter 61) as well as in numerous journal articles. [42] Infectious esophagitis primarily occurs in the setting of immunosuppression, particularly in HIV-infected patients. The most frequent causes are *C albicans,* CMV, and HSV. Mixed infections can occur. Efforts should be made to establish the presence of these agents since all three are readily treatable. Since *C albicans* is by far the most common esophageal pathogen in HIV-infected patients, current guidelines recommend an empiric trial of appropriate antifungal therapy in these patients before endoscopic evaluation. [42] If therapy fails or if symptoms recur, the patient should be investigated. *C albicans* can be easily identified by mucosal brushings, which have a lower yield for identifying CMV and HSV. Biopsies of both the base and edges of an esophageal ulcer and use of H & E staining will be diagnostic of CMV and HSV. [42] Therefore it is generally not necessary to send biopsy material for fungal or viral cultures or use special techniques such as immunohistochemistry or in situ DNA hybridization in order to make the diagnosis. [71]

Cholecystitis and Cholangitis

Bile obtained from the gallbladder and the bile ducts is normally sterile in healthy individuals. [89] The prevalence of bacterial infection of bile correlates with the severity of the underlying disease. For example, patients with symptomatic cholelithiasis have a 20% prevalence while those with ascending cholangitis have over a 90% prevalence. [90] Cultures of infected bile are most often polymicrobial. The most common pathogens are *E coli, Enterococcus, Klebsiella,* and *Enterobacter,* while anaerobes are fairly uncommon. [89], [90]

Blood cultures should be obtained on febrile patients with suspected cholecystitis. Furthermore, cholecystitis or indolent ascending cholangitis should be suspected as a source of fever and bacteremia in patients without other symptoms or signs of biliary tract infection, in particular elderly patients. Samples of bile can be obtained by aspiration during procedures to decompress an obstructed biliary tree, such as percutaneous transhepatic cholangiography or endoscopic retrograde cholangiopancreatography. These specimens should be sent for aerobic culture.

Pancreatitis

Bacteria, parasites, and viruses can precipitate acute pancreatitis. [91] In order to qualify as an infectious cause of pancreatitis, patients need to have both clinical evidence of pancreatitis as well as documentation of infection by the pathogen (i.e., organism identified in the pancreatic parenchyma, duct, or juice or a fourfold-rise in serologic titers). Using these strict criteria Parenti and colleagues [91] identified 32 definite or probable cases in the literature of acute pancreatitis caused by infectious agents. The most common causes were CMV and *Ascaris lumbricoides.* The latter roundworm causes pancreatitis by direct obstruction of the common bile duct or pancreatic duct.

If the common precipitants of acute pancreatitis (e.g., alcohol, gallstones, drugs) have been excluded, it may be reasonable to consider an infectious cause although these are very uncommon. In a patient who is immunocompromised due to HIV or transplantation, CMV serology may be indicated. Likewise in a patient with risk factors for *A lumbricoides,* it would be reasonable to obtain stool for ova and parasite examination. Superimposed bacterial infection frequently complicates severe acute pancreatitis. [92]

Peritonitis

Infections of the peritoneum are almost always caused by bacteria or fungi. Spontaneous bacterial peritonitis (SBP) usually occurs in cirrhotics with preexisting ascites and complicates about 15% of such patients. [93] Over 90% of cases of SBP involve monomicrobial infections. About 70% are caused by aerobic gram-negative bacilli, the most common organism being *E coli.* Approximately 25% are caused by gram-positive organisms and only 5% are caused by anaerobes. [93] SBP is diagnosed by paracentesis and culture of the ascitic fluid. Optimal yield is obtained if the fluid is placed in a blood culture bottle at the bedside before transport to the microbiology laboratory.

For patients in which ascitic fluid culture yields a polymicrobial bacterial infection or a fungal pathogen, a perforation of a hollow abdominal organ should be suspected as spillage of both aerobic and anaerobic bacteria can occur. [94] Other indications to culture ascitic fluid for fungi include patients with peritonitis who have indwelling peritoneal dialysis catheters or patients who recently had abdominal surgery.

COST ISSUES IN MICROBIOLOGIC TESTING

The very low yield of nonselective bacterial stool culture and O & P examinations has not been subject to much scrutiny in clinical practice because these tests are easy to perform and involve little risk to the patient. The occasional positive result can lead to specific therapy and the avoidance of more expensive and risky diagnostic interventions such as endoscopy and biopsy. Despite these apparent benefits, the costs of such practices are significant with estimates of $1000 to $1300 per positive culture. The costs of performing a routine stool bacterial culture include charges for reagents, supplies, technologist time, and charges to the patient. Two surveys of microbiology laboratory practices in 668 hospitals in the United States revealed that the majority (>50%) did not have a policy that restricted the number of stool specimens that would be processed per patient. [2], [95] Furthermore, less than 25% of hospitals rejected specimens submitted after 3 days of hospitalization. Based on the fact that nearly 50% or more of stool specimens submitted to microbiology laboratories are from patients hospitalized for more than 3 days, it has been estimated that application of the “3-day rule” would save the average hospital per year $10,000 of reagent costs, over $70,000 of patient charges, and 730 hours of technologist time. [95] Extrapolation of these figures to the 6000 hospitals in the U.S. suggests that savings of $27 to $73 million a year could be achieved by implementation of such restrictions.

Another source of unnecessary costs associated with stool bacterial cultures and O & P examinations is the submission of multiple specimens from the same patient. It is not an uncommon practice to request the collection of three stool samples before the results of the first sample are known. Several studies have documented that over 90% of enteric bacterial and parasitic pathogens were detected on the first stool sample. [62] Guidelines suggest that to minimize the costs associated with multiple specimens, clinicians should submit an initial stool sample for analysis and then submit additional samples only if the first is negative. An alternative approach would be to order the initial sample, and if negative, to then send the second stool sample for the more rapid and sensitive tests such as immunofluorescence and enzyme immunoassays. For example, in a patient with chronic diarrhea with a recent history of travel to an area with a high prevalence of *G lamblia,* an initially negative routine O & P examination might be followed up with one of the EIAs that specifically detects *G lamblia* rather than another O & P analysis.

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*This chapter succeeds the chapter in previous editions by Tarr, Surawicz, and Clausen entitled "Microbiologic Studies." We wish to acknowledge that we have drawn heavily on the material from their chapter.

CHAPTER 145

Richard A. Kozarek

GASTROINTESTINAL DILATION AND STENT PLACEMENT

The primary goal of treatment of any stenosis is lumenal enlargement and amelioration of obstructive symptoms. Such symptoms depend on the stricture etiology and site and may include dysphagia, nausea and vomiting, abdominal pain and obstipation, or frank bowel obstruction.

Historically, the dilation of gastrointestinal (GI) strictures was limited to accessible anatomic areas, primarily the esophagus or anorectum. With the advent of endoscopically or radiographically placed polyethylene balloons, a variety of gastric, small bowel, and colonic strictures became amenable to such therapy. Likewise, prosthesis placement was initially limited to the esophagus and subsequently expanded into the pancreaticobiliary tree. Expandable metallic stent technology, however, has allowed treatment of previously inaccessible and more central stenoses.

Whereas all of the current dilating systems achieve efficacy by either stricture stretch or fracture, data are sparse regarding the mechanism of action with individual dilating systems. Nor are there good prospective data comparing endoscopically facilitated dilation with alternative treatment modalities. Finally, for central stenoses and chronic proton-pump inhibition for esophageal reflux stricture, there are few studies that compare the efficacy and side effects of various dilating systems. Similarly, despite their widespread use, there are few comparative studies randomizing expandable prostheses in the esophagus. Central placements, in turn, have been limited to small series and data comparing their placement to surgical intervention have not been published. This chapter compares the various technologies with regard to ease and site of application, patient tolerance, cost-benefit ratio, and available safety and efficacy data.

THEORETICAL CONSIDERATIONS

Dilation

The basic goals of stricture dilation include safe and efficacious lumenal enlargement plus prevention of restenosis. [1] The latter may include proton pump inhibitors after esophageal bougienage for a reflux-induced stricture, [2] placement of a prosthesis after dilation of an esophageal malignancy, or elective surgical resection, plasty, or bypass of a dilated area after dilation or stent placement for an obstructing rectosigmoid malignancy.

Although the exact mechanisms of lumenal enlargement remain uncertain, circumferential stretch and stricture split may be operative in stricture dilation. The former presupposes considerable elasticity in circumferential fibrous tissue and the latter an inherent rigidity in which dilation is effected by one or several longitudinal tears. It is unlikely that a pylorus of 3 mm diameter can be dilated to 10 or 15 mm without a significant laceration of scirrhous tissue and, potentially, of muscle. Relevant to this are the gross longitudinal tears and histological disruption of collagen and circular muscle that have been described after esophageal bougienage for fibrous stenoses and achalasia. [3] Most perforations associated with dilation, in turn, appear to be an extension of these tears.

Available dilating modalities can be divided into mercury bougies, guidewire-directed dilators, polyethylene balloons, and miscellaneous types [1] (Table 145-1, Fig. 145-1, Fig. 145-2, Fig. 145-3 and Fig. 145-4). Mercury-filled dilators, ranging in size from 3 to 20 mm (10F to 60F), can be subdivided into the original, blunt-tipped Hurst bougies and a tapered-tip variant called Maloney bougies. Originally passed without fluoroscopic control, these dilators are used infrequently by many practitioners.

Mercury Bougies
Blunt-tip (Hurst)
Tapered-tip (Maloney)
Guidewire-Directed
Metal olives (Jackson-Plummer; Eder-Puestow, including triple olive adaptation)
Hollow-core polyvinyl (Savary-Gilliard, American)
Neoplex stepped-diameter (Celeştin)
Spindle-shaped (Key-Med advanced dilator)
Polyethylene Balloons
TTS
Guidewire facilitated
CRE
Latex Balloons
Brown-McCardy
Mosher
Miscellaneous
Woven silk (Jackson, Phillips)
Graded, plastic oversheath
Tapered-tip endoscope
Balloon or tape affixed to endoscope shaft
Electronic-mechanical (Starck variant)

CRE, controlled radial expansion; TTS, through-the-scope.

TABLE 145-1 Dilators for Gastrointestinal Stenoses

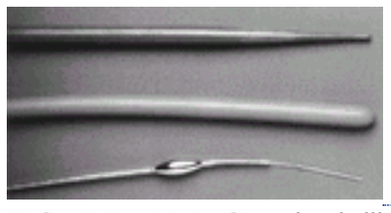

FIGURE 145-1. Standard dilating systems for esophageal bougienage: Maloney (**top**), Hurst (**middle**), Eder-Puestow (**bottom**). The latter two are obsolete but are still used occasionally.

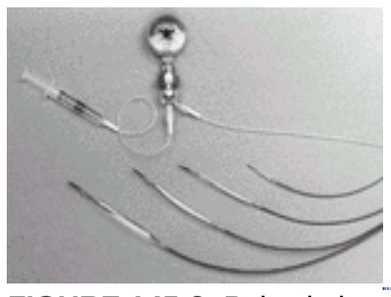

FIGURE 145-2. Polyethylene balloon dilating kit includes mercury manometer and injection syringe. Balloons are passed over an endoscopically or radiographically placed guidewire.

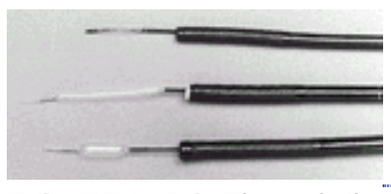

FIGURE 145-3. Through-the-scope polyethylene dilating balloons are available in various lengths and diameters. The biliary balloon (**top**) is a hybrid that passes through the scope but over a guidewire.

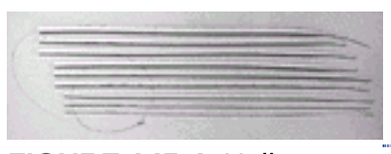

FIGURE 145-4. Hollow-core polyvinyl dilators. The Savary-Gilliard dilator is longer, more tapered, and less radiopaque.

Historically, guidewire-directed dilators have included Jackson-Plummer bougies and Eder-Puestow dilators, which include a triple olive variant in which multiple metal olives of increasing diameter are placed on the same dilating shaft. [4] Four additional wire-guided dilating systems have been marketed: the Key-Med advanced dilator, consisting of three spindle-shaped silicone bougies on stainless steel shafts; the stepped neoplex (Celestin) dilator; and two types of hollow-core polyvinyl systems, the Savary-Gilliard and the American. The Celestin dilating system consists of two tapered dilators that reach a maximum diameter of 12 and 18 mm, respectively. The Savary-Gilliard system consists of bougies ranging in size from 5 to 18 mm (15F to 54F). The American dilating system (Bard, Inc., Mentor, OH) ranges in size from 7 to 20 mm (21F to 60F). In contrast to the barium-impregnated American dilators, Savary-Gilliard dilators are longer and have a more gradually tapered tip.

Bougienage was first used by Fabricus al Acquadendente, who used a wax dilator for a food impaction. [5] The term *bougienage* is derived from the Algerian town of Bouginhay, the medieval capital of the wax candle trade. Cork and woven silk dilators have also been used, the latter since at least the 16th century, when Thomas Willis used a cork-tipped whalebone to treat a probable achalasia patient.

After wire-guided bougies, the second major advance was the development of polyethylene balloons for use in the GI tract. [6], [7] Ranging in diameter from 4 to 40 mm, [6] they allow dilation of previously inaccessible strictures in the stomach, small bowel, and colon. Dilating balloons are fixed on 5F to 7F catheter shafts that range between 100 and 200 cm in length. They can be passed over an endoscopically or radiographically placed guidewire or directly through-the-scope (TTS). A full dilation set includes balloons of variable length and diameter, 5- to 30-mL syringes, guidewires, and a manometer to delineate balloon pressure during inflation; a dilating gun to maintain pressure and stopcocks to ensure a constant pressure during inflation are optional. More recent advances in balloon technology are fabrication of low profile and high compliance balloons. The latter can withstand a dilating pressure three- to fourfold higher than previously marketed balloons and have improved results in recalcitrant strictures. A newer development is the controlled radial expansion (CRE) balloon. [8] These balloons can be passed over a guidewire and variable inflation pressure results in balloons of increasing diameter. A single CRE balloon, contingent upon the pressure used to inflate it, may increase in diameter from 6 to 8, 8 to 10, 10 to 12, 15 to 18, and 18 to 20 mm.

Stents

In contrast to dilation, prosthesis placement attempts to reestablish permanent continuity to the GI tract. The fact that such prostheses are placed primarily in the setting of malignancy suggests that the technology is imperfect: GI stents occlude, migrate, erode, and may allow reflux of potentially noxious GI contents contingent on their location. [9] Symonds is credited with the first prosthesis placement in the esophagus in 1887. Initial stents were fabricated of ivory or boxwood and held in place by a variety of external fixation devices until the development of the inverted funnel by Grootskin in the early 1900s. Since then, a variety of homemade and commercially available esophageal prostheses have been used, the latter usually constructed of latex or silicone molded over a stainless steel core (Table 145-2; Fig. 145-5). Inserted surgically or pushed into place using bougies, a small-caliber endoscope, or an expandable metal olive fixation device (Nottingham introducer), the design of these stents and insertion devices precluded placement across central or sharply angulated stenoses and their application was primarily limited to esophageal or gastroesophageal malignancies.

Conventional
Tygon tubing (homemade)
Silicone/latex-coated steel spiral*
Neoprene coated (Wilson-Cook, Inc.)
Expandable
Wallstent
Esophageal/colonic/enteral
Flamingo variant
Z Stent
21 mm/25 mm flanges
Gianturco-Rösch variant
Dua stent with antireflux valve
Song variant
Esophacoil
Ultraflex
Covered/uncovered

*Multiple types/manufacturers.

TABLE 145-2 Prostheses for Malignant Gastrointestinal Stenoses

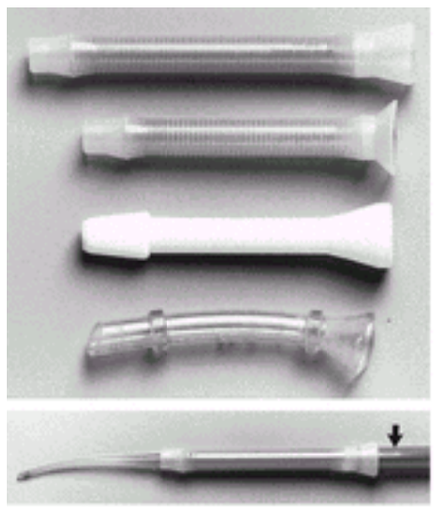

FIGURE 145-5. Conventional esophageal prostheses (**A)**. Conventional and low profile Wilson-Cook stents (**top)**, Atkinson prosthesis (**2nd from bottom**), and homemade stent fashioned from Tygon tubing. *Arrow* demonstrates pusher tube used to seat prosthesis, which is stabilized over a Savary dilator (**B**).

Currently, expandable metal prostheses have supplanted rigid stents in the esophagus and have also made gastroduodenal and colorectal stenoses amenable to prostheses [10] (Table 145-2, Fig. 145-6). Although different in expansible force and delivery systems, all of these prostheses share the feature of placement in a compressed state with subsequent spontaneous or balloon-assisted expansion. Four major types are marketed in the United States and variations of these stents are marketed around the world. The Wallstent (Microvasive, Inc., Natick, MA) was initially introduced as a two-layer stainless steel, double-dogbone prosthesis with an interposed layer of silicone. Subsequently marketed stents are uncovered or partially covered, range between 18 to 25 mm in diameter, and are constrained by an 18F delivery system. A heavily tapered version of this prosthesis (Flamingo stent) has also been marketed in Europe for esophagogastric junction tumors. [11] The Z stent (Wilson-Cook Medical, Winston-Salem, NC) is a 20-mm diameter urethane-covered prosthesis that flares to 22 to 25 mm at the ends. Ranging from 4 to 14 cm in length, the prosthesis must be backloaded into a 28F compression catheter at time of placement. [12] In contrast, the Z stent, marketed in Europe and Asia, has a different coating and delivery system and also has small barbs on the shaft to minimize migration. A variant of the Z stent (Windsock, Dua stent [Wilson-Cook, Winston-Salem, NC]) containing an antireflux valve attempts to minimize regurgitation when used for distal esophageal or cardia malignancies. [13] An additional variant, the Choo stent (Solco Intermed, Seoul, Korea) is marketed in Asia and Europe. The Ultraflex prosthesis (Microvasive, Natick, MA) consists of a 10- or 15-cm long, 18- to 25-mm diameter nitinol weave. The delivery system allows release by pulling a long suture used to constrain the stent on an insertion shaft. Finally, the Esophacoil (Medtronics, Inc., Eden Prairie, MN) consists of a spiral nitinol coil constrained on an insertion shaft by three separate trip wires.

FIGURE 145-6. Expandable prostheses (**left to right**): conventional Z stent, small intestinal mucosa covered Z stent (prototype device), Windsock Z, variably sized Esophacoil and covered Ultraflex prostheses, Flamingo, and Wallstent II.

All of these products differ not only in design and delivery system but also in physical characteristics. Although all are more malleable than conventional prostheses, Ultraflex stents and Esophacoils can often be used for angulated stenoses. Fully covered Z stents and Esophacoils can, at least in theory, be removed and may ultimately play a role in refractory benign strictures. Wallstents and Ultraflex prostheses imbed deeply into the esophageal wall and may have a role to play in patients with esophago-airway fistulae without a tumor shelf. The radial force generated by the prostheses is quite different and may help define the type of stent that is chosen (Esophacoil > Wallstent > Z stent ˜ covered Ultraflex stent). [14]

Advantages of expandable metal prostheses over their conventional counterparts in the esophagus include the ability to be inserted under combined endoscopic and fluoroscopic control and the limited need for dilation for placement. [10] The latter may decrease the risk of bleeding or perforation during insertion. Moreover their flexibility allows placement beyond acutely angulated malignancies and their ability to imbed into the esophageal wall should minimize subsequent stent migrations. Their use in central stenoses (e.g., gastric, duodenal, colorectal) offers a previously unavailable therapeutic option in patients with recurrent, metastatic, or unresectable obstructing malignancies. Examples are obstructing sigmoid cancer with widespread disease or advanced pancreatic cancer and gastric outlet obstruction. [15], [16] and [17]

What remains ill-defined, however, is the role this technology should play compared to conventional treatments such as surgery or percutaneous endoscopic gastrostomy/jejunostomy (PEG/J) placement. Moreover, where this technology fits into our therapeutic armamentarium of ablative therapies (neodymium:yttrium-aluminum garnet [Nd:YAG] laser, bipolar cautery, caustic therapy, argon plasma coagulation, photodynamic therapy) requires continued evaluation. [18]

TECHNICAL APPLICATION

Dilation

In general, radiologic- or endoscopic-directed bougienage should prove safer and more effective than blind dilation, [7], [19] particularly for sharply angulated, extremely tight, or proximal esophageal stenoses, and it should be an invariable rule with more central stenoses, including those in the stomach and colon. For dilation, these general principles must be balanced against the availability of fluoroscopy, the added cost of endoscopy or fluoroscopy, and the physician's previous experience with a particular dilating modality. Endoscopically facilitated guidewire placement and subsequent bougienage need not always require fluoroscopic control if copious guidewire has been placed into the stomach or a marked guidewire is used and attention is given to ensure that guidewire displacement does not occur with endoscope withdrawal. [1]

Of importance technically is the degree of lumenal enlargement that can be undertaken safely in a single dilating session. There remains a maxim in esophageal bougienage that one should increase a lumenal stenosis by no more than 2 mm (6F) in a single dilation session. [1] Although it is based on common sense and an attempt to avoid such complications as bleeding and perforation, this adage does not necessarily hold true for most rings or webs and some pliable, reflux-induced strictures. The degree of lumenal enlargement should be contingent not only on the stricture itself (i.e., membranous or fibrotic) but also on the degree of associated lumenal ulceration and the risks and benefits of alternative treatment modalities. These decisions cannot be made by a review of the scanty literature discussing side effects of bougienage but require a great deal of common sense on the part of the physician. [1], [19], [20]

Stent Placement

Stent placement requires stricture dilation to a size that admits a conventional prosthesis or the delivery system of an expandable stent. [9] More commonly, strictures are dilated to 30F to 36F even with small (18F) delivery systems. This precludes stent dislodgment when the delivery system is retrieved through a partially expanded prosthesis and may avoid the need for stent dilation with its potential for dislodgment. Perhaps the most important technical aspects of stent placement are adequate measurement of the stenosis with selection of a prosthesis that is 3 to 4 cm longer than the neoplasm and accurate placement. Whereas a subset of expandable prostheses can be placed either through or alongside an endoscope without x-ray control, [21] most endoscopists use concomitant fluoroscopy marking the proximal and distal stricture margins either with external markers or with injections of contrast material. The latter allows more precise prosthesis placement and may allow for repositioning of the prosthesis prior to full stent deployment.

Esophageal Strictures

Dilation Taking esophageal stenosis as the prototype, there are three basic dilation systems in widespread use: mercury bougies, guidewire-directed dilators, and polyethylene balloons. Most patients who present with dysphagia have had previous esophagoscopy, although a few with lower esophageal rings may have had barium studies alone. With rings, webs, and mild reflux stenosis, mercury bougienage after a 6-hour fast and pharyngeal anesthesia can often be done using a single 16 to 18 mm (48F to 54F) Maloney dilator. [22] Such dilators can be passed in the upright or lateral decubitus position. Although ideally done under fluoroscopic control to avoid kinking or retroflexion, many of these esophageal dilations can be performed safely without these capabilities. Long, angulated, or eccentric esophageal strictures, as well as severe (<7 mm) stenoses, are best handled with a guidewire dilating system. Patients require an initial endoscopy to define the stricture's cause and characteristics (e.g., length, diameter, pliability, eccentricity, and associated pseudodiverticula). Because a biopsy of the entire length of the stricture can be performed after dilation, I sometimes delay tissue sampling until immediately after the dilation. Use of polyvinyl dilators, which have supplanted Eder-Puestow metal olives in most centers, always requires guidewire placement. [1], [19], [23] This is usually done in conjunction with initial endoscopy, at which time a fluoroscopically monitored piano-style wire with a spring tip can be passed through the stricture and fed freely into the stomach. Alternatively, the wire can sometimes be passed radiographically without endoscopy. After baseline endoscopy and stricture sizing, a dilator approximately the size of the stricture is passed, making sure the guidewire is fixed and the head is bent forward. This is followed by one or two additional dilators, for an increment of up to 3 or 4 mm (10F to 12F), contingent on the stricture, before repeat endoscopy and stricture biopsy. Because of their gradual taper, polyvinyl dilators can pass through most stenoses with relative ease. The hesitation felt with these systems is related more to dilator friction over the guidewire than to the stricture itself. Moderate stenoses (7–13 mm) can be dilated using either mercury bougies under fluoroscopic control or polyvinyl dilators. The latter systems can be used without radiographic monitoring in some, although it is imperative that the guidewire is advanced far into the stomach and not inadvertently withdrawn at the time of endoscope removal. This is best done by feeding a marked guidewire forward simultaneously with endoscope or bougie withdrawal and having an assistant fix the wire at the level of the patient's mouth. [1] In addition, polyethylene balloons (TTS, CRE) have also been used to dilate moderate esophageal strictures. The technique is described in the section on "Nonesophageal Strictures."

Stent Placement Esophageal prosthesis placement requires adequate dilation both to allow passage of various delivery devices and to localize and measure the length of the neoplasm. [9] Necessary dilator sizes range from 48F to 51F for conventional prostheses and 24F to 36F for the expandable prostheses. Preoperative bronchoscopy may be required, particularly for bulky extrinsic neoplasms, as stent placement may be associated with airway compression and acute respiratory embarrassment. Tumor localization can be done by using external radiopaque markers taped to the patient's chest but is more commonly accomplished by injecting water or lipid-soluble contrast material into the proximal or distal tumor margins. [9] Alternatively, endoscopic clip placement can be used. Prosthesis placement requires fluoroscopic control although some of the smaller delivery systems (Ultraflex, single-braid Wallstent) allow simultaneous endoscopic visualization during delivery. [9] Stents should be 4 to 6 cm longer than the neoplasm, contingent on local anatomy (e.g., esophagogastric junction, cricopharyngeus, contralateral gastric wall; stricture angulation). Conventional prostheses are pushed into place over a guidewire using a variety of devices to stabilize the stent (e.g., small-caliber endoscope, Savary-type dilator, Nottingham expandable olive fixation shaft) and various types of pusher tubes. These devices have considerable resistance at the level of the cricopharyngeus and may require neck hyperextension for passage. Conventional stents are pushed into place followed by retrieval of the pusher tube and delivery system and an immediate repeat endoscopy to document correct prosthesis position. Expandable prostheses have variable delivery systems. [9] Z stents and Wallstents are delivered by pulling back a compression catheter. Ultraflex stents are constrained by a single long suture that can be pulled off the delivery shaft. The Esophacoil is compressed onto a delivery haft and released with three separate trip wires. [24] Although there is significant foreshortening of both Wallstents and Ultraflex prostheses, the Esophacoil foreshortens to half of its predelivery length, making proper placement crucial. Despite the plethora of prostheses now available, certain practices are followed after placement of all prostheses. The most important is the need for immediate endoscopy postinsertion to assure that the prosthesis has accomplished the treatment goal (e.g., correct location, full expansion, occlusion of tracheoesophageal fistula). Problems occur when the prosthesis is too long or short, abuts into the cricopharyngeus or contralateral gastric wall, or has inadequate radial force to open a tight neoplastic stricture. The latter may preclude retrieval of the delivery system. Airway compression and immediate migration at the time of placement are additional potential problems. Once expandable stents have fully deployed, only the coated Z stent has the potential to be repositioned. The Esophacoil and uncovered Ultraflex prostheses, in turn, can be removed, at least in theory. The endoscopist has to be prepared to deal with these problems. This requires access to balloon dilators as well as additional expandable and conventional prostheses to correct problems associated with inadequate length, acute angulation, or migration.

Nonesophageal Strictures

Dilation Most nonesophageal strictures require balloon dilation, although stenotic gastric stapling orifices and anastomotic strictures of the rectosigmoid can be enlarged with polyvinyl dilators or even electrocautery. [25], [26] and [27] Using pyloric stenosis as a representative stricture, I recommend using TTS or CRE balloons whenever possible. Advantages include direct stricture visualization, improved placement control, and immediate evaluation of the dilated stenosis. [6] The use of a TTS or CRE dilator requires endoscopic approximation of the stricture size and selection of a balloon that is 1 to 2 mm (3F to 6F) larger. Both balloon and dilator shaft should be coated with silicone, and negative pressure should be applied to the balloon using a 10- to 20-mL syringe. These measures, as well as avoiding excessive angulation of the endoscopic tip, allow dilator passage until all or part of the balloon is visualized. The balloon is centered in the stenotic pylorus under a combination of endoscopic and fluoroscopic control. The latter also helps prevent damage of the bulb apex or C loop wall related to excessive pressure of the balloon tip or extreme balloon angulation. Although air can be used for inflation, 10% to 25% contrast solution allows better visualization fluoroscopically and a more uniform balloon dilation. Obliteration of the balloon waist is required with pressures up to 12 atmospheres (atm). There is no evidence that 2 minutes of continued inflation is better than 15 seconds after the balloon waist has been effaced. [28] I usually use 30 seconds of dilation and redilate a second or third time after repositioning of the balloon. After dilation has been effected, complete evacuation of the balloon and straightening of the endoscope tip are required to allow retrieval. Additional, larger dilating balloons can then be used. However, the degree of lumenal enlargement in a single session remains a matter of common sense and is contingent on size of the initial stenosis, presence and degree of active ulceration, and patient discomfort with initial dilation. The ultimate goal is to dilate to 15 to 18 mm and follow up with complete endoscopic inspection of the pylorus and duodenum. This goal sometimes requires two or three dilating sessions separated by an interval of several days if the obstruction is acute or several weeks if it is chronic.

Stent Placement Expandable prostheses can be placed in the proximal stomach and distal colorectum using conventional delivery systems and techniques similar to those described for esophageal stenting. Duodenal, proximal jejunal, and more proximal colon strictures require either a longer insertion system or a stent mounted on a 3- to 4-mm shaft that can be placed through a large channel endoscope. [18] As of this writing, only uncovered Wallstents 18 to 24 mm in diameter and 6 to 9 cm in length are available for transendoscopic placement. These stents require balloon dilation of the malignant stenosis, delineation of the stricture length, localization with contrast injection at the tumor margins, and concomitant endoscopic and fluoroscopic control to ascertain proper placement and prosthesis expansion.

INDICATIONS

Dilation

The benefits and risks associated with lumenal dilation must be considered in relation to alternative treatment modalities (e.g., antireflux surgery or esophageal resection for a reflux stricture; vagotomy-pyloroplasty for pyloric stenosis; [29] endoscopic laser photoablation, electrocautery, or prosthesis placement [10]). Pharmacological implications of dilation also must be considered. Reflux-related esophageal strictures and many pyloric stenoses require long-term proton pump inhibition and possibly use of a prokinetic agent. Dilation of a Crohn's disease stenosis usually requires adjustment of corticosteroid dosage or initiation of immunosuppressive agents or infliximab to minimize the inflammatory response and stricture reformation. [30]

Indications for dilation are contingent on the anatomic area involved (Table 145-3). In the esophagus, symptoms are most often dysphagia and food impaction, although atypical chest pain, aspiration, and odynophagia may also be seen. [31], [32], [33] and [34] Indications for pyloric dilation are usually recurrent nausea and vomiting, weight loss, abdominal pain, and severe reflux. [35], [36] Small bowel and distal colonic stenoses may require dilation for intractable obstipation, progressive diminution in

stool size, pain, and recurrent bowel obstruction. [25], [26] and [27], [37] The causes of these and of a variety of other miscellaneous stenoses are listed in Table 145-3.

Esophagus
Ring or web
Reflux stricture
Malignancy
Miscellaneous (e.g., caustic ingestion, motility disorders)
Stomach
Pyloric stenosis
Anastomotic stricture
Gastric stapling/bypass stenosis
Miscellaneous (e.g., caustic ingestion, proximal malignancy)
Small Bowel
Duodenum (web, acid peptic stricture, anastomotic)
Ileum (Crohn's, NSAID-induced stenosis)
Colon
Anastomotic stenosis
Inflammatory stricture (IBD, diverticular, radiation-induced)
Miscellaneous
Stenotic gastrostomy, enterostomy, or colostomy stoma

IBD, inflammatory bowel disease; NSAID, nonsteroidal antiinflammatory drug.

TABLE 145-3 Indications for Gastrointestinal Dilation

Stent Placement

For the most part, prostheses should be used for obstructing or fistulizing neoplasms that are unresectable for cure or in potentially curable patients with prohibitive operative risk [9], [10] (Table 145-4). The most experience has accrued in the proximal gut for malignant dysphagia or esophago-airway fistulae, the latter a consequence of primary esophageal carcinoma, lung cancer, or mediastinal metastases. Gastric outlet obstruction in the setting of pancreatic cancer occurs in the setting of widespread disease. Traditionally treated by gastrojejunostomy or PEG/J in infirm patients, the latter might be ideal candidates for stent insertion into the C loop. [16], [38] Likewise, patients who present with malignant colon obstruction usually have far advanced disease (stage III [40%] or IV [60%]). [17] Traditionally treated with palliative resection or bypass, prosthesis placement is being evaluated both to allow adequate bowel preparation in some patients and as long-term palliation in a subset of patients with malignant ascites or liver metastases. [39]

Conventional stents
Esophagus
Malignant dysphagia (esophagus, esophagogastric junction, lung, mediastinal tumor)
Esophago-airway fistula
± Benign disease (e.g., caustic ingestion/anastomotic stricture or leak)
Expandable Prostheses
Esophagus
Malignant dysphagia
Esophago-airway fistula
Stomach/Duodenum (high surgical risk)
Malignant pyloric/C-loop obstruction
Gastric/pancreatic/biliary
Small Bowel/Colon (high surgical risk)
Obstructing/malignant stenosis
??? Benign strictures: radiation, Crohn's disease

TABLE 145-4 Indications for Prostheses

CONTRAINDICATIONS AND RISKS

Dilation

Contraindications to dilation include lack of informed consent, an acute abdomen, or a deeply ulcerated stenosis for which the risks of dilation outweigh the benefits. There may also be patient- and lesion-specific contraindications, such as coagulopathy or a colon stenosis associated with acute diverticulitis. Both the physician and the patient must be aware of alternative treatment modalities and the possible need for subsequent long-term ancillary measures such as proton pump inhibitors. Data on alternative treatment modalities are sparse but include a retrospective series on patients with pyloric stenosis treated with resective surgery, highly selective vagotomy, or vagotomy with pyloroplasty. [29] The combined morbidity was 32%, mortality was 6%, reoperation rate was approximately 10%, and the long-term symptom relief rate was not defined. Postoperative delayed gastric emptying was recorded in 25% of patients.

Risks of GI dilation include problems associated with all endoscopic procedures. The risks associated with the addition of dilation have usually been defined as an increased incidence of bleeding and perforation, [40] as well as bacteremia. [41], [42] With the exception of esophageal dilation, the incidence of these complications in various settings remains poorly defined.

Stent Placement

Prostheses are contraindicated in the good risk and potentially curable patient. [9], [18] Conventional prostheses should not be placed in the setting of a tracheoesophageal fistula unless there is an adequate shelf to seat the stent. Stents should not be used if placement of a dilator approximating the stent diameter results in significant airway compression by a bulky neoplasm. Stents may be contraindicated if placement cannot be undertaken without impingement on the cricopharyngeus muscle or anal sphincter, or if placement impacts on a contralateral lumenal wall thereby precluding adequate function. An absolute contraindication for prosthesis placement is an inability to endoscopically or radiographically define both the proximal and distal stricture margin. Nor should permanent stents be used for most benign esophageal, enteral, or colonic stenoses given long-term potential for erosion, occlusion, and migration.

Complications of prostheses are well defined in the esophagus but otherwise experience is limited. Acute procedure-related complications with conventional stents include perforation, bleeding, tracheal compression, and tube malposition. Total complications with conventional stent placement have approximated 20% with a mean procedure-related mortality of 8.6%. [9] Subacute complications include erosion with bleeding or fistula development, stent migration, food bolus impaction, and tumor overgrowth. Variable degrees of reflux are invariable if the esophagogastric junction has been stented and florid regurgitation or aspiration may be noted in patients who have delayed gastric emptying as a consequence of vagal denervation or gastric replacement by tumor.

Complications of expandable prostheses are variable and contingent on prosthesis design and endoscopist experience. Our group retrospectively reviewed 85 patients, 47 of whom had conventional and 37 a variety of expandable prostheses. [43] Insertion complications, prestent and poststent dysphagia scores, and complete tracheoesophageal fistula occlusion rates were comparable, but there was a 25% higher rate of subacute or chronic complications in patients receiving expandable prostheses. The latter included tumor ingrowth or overgrowth, migration, and aspiration pneumonia. Other studies have confirmed ingrowth through uncovered prostheses or stents that have delaminated. For instance, ingrowth occurred in 66% of 114 patients in whom an uncovered Ultraflex stent was placed. [44] Fully covered prostheses, on the other hand, appear to predispose to stent migration as noted in the U.S. Multicenter Z Stent Trial. [12]

Prior radiation and chemotherapy may predispose to significant complication, particularly with the barbed European Z stent. For instance, 23 late complications occurred in 22 (38%) of 59 patients treated with Gianturco-Rösch Z stents, including life-threatening complications in 9 (bleeding, 7; perforation, 1; tracheoesophageal fistula, 1). [46] Eight (36%) of 22 patients with prior radiation or chemotherapy had life-threatening complications as compared to only 1 (3%) of 37 without such prior therapy ($P = 0.005$). This increased risk was confirmed in a more recent publication. [47]

Metallic prostheses also have intrinsic problems. Early Wallstents delaminated, Esophacoils may deliver in a tangle, and there are multiple reports of Ultraflex fractures. [9] Moreover, the ability to place stents more proximally than with conventional prostheses [48] has been associated with complications such as cervical discitis

[49] and stent perforation into the common carotid artery. [50]

Data directly comparing expandable to conventional stent placements are sparse, although three reports [51], [52] and [53] suggest that expandable prostheses are easier to insert and potentially associated with fewer complications. Survival rates, in contrast, are not different.

ASSESSMENT OF RESULTS

Dilation

Esophagus Most data on GI dilation derive from esophageal bougienage with a combination of Eder-Puestow and mercury dilating systems. This combination, using sequential bougies over a period of days to months, is associated with 70% to 100% improvement in dysphagia in over 850 patients with benign esophageal strictures compiled from various series. [7], [22], [54] Reported complications in these patients included 16 (0.2%) perforations, 5 major bleeding episodes, 2 aspirations, and 1 death. This compares with the perforation rates of 0.4% for mercury dilators and 0.6% for metal olives reported in an American Society for Gastrointestinal Endoscopy (ASGE) survey. [7] Such perforations are usually in the cervical esophagus and relate to improper dilator introduction or in the thoracoabdominal esophagus just proximal to the stenosis related to dilator or guidewire kinking. Less frequently, the stricture itself splits, as may be seen with achalasia (1%–5% perforation rate). [1] A number of series assess balloon dilators for esophageal stenoses. In an early series, 93% of dilations of benign and malignant esophageal strictures in 88 patients were technically successful with a 3% minor complication rate and 91% symptomatic improvement over a mean of 10 months. [55] Additional series have been reported in adults and children, as have trials comparing balloon technology to Eder-Puestow or Savary dilators. [55], [56], [57] and [58] Data using the guidewire-directed dilating systems continue to accrue. In a randomized prospective trial comparing Eder-Puestow and Celestin dilators in 72 patients, there was no significant difference in long-term symptom relief, although the Celestin system was thought to be quicker and cause less pharyngeal trauma. A single perforation occurred with the Celestin system. [59] A second paper reviewed 302 dilations in 100 patients with benign esophageal strictures using this system. [60] The two reported perforations were medically managed and were thought to be related to esophageal laceration with the endoscope and not the dilator. A number of series review experiences with hollow-core polyvinyl dilating systems. [5], [57], [58], [61], [62] and [63] Dumon and colleagues [63] claimed efficacy in all 300 patients treated for benign or malignant esophageal stenosis, with only one perforation. Our group successfully dilated 432 patients with 716 courses of dilation therapy; 89% were dilated with polyvinyl dilators and only 8% required fluoroscopic monitoring. [61] Approximately 80% of the dilation sessions were undertaken with a single large dilator or employed incremental dilators more than 6F in a single session. There were no complications directly related to the bougienage. Additional large series have been published. [62]

Clinical implications. The ability to dilate a stenosis does not imply that dilation is the procedure of choice in every instance. Dilation should not be the sole treatment in individuals who require bougienage so frequently that cumulative risk and expense become prohibitive. In this setting, intralesional steroid injection might be tried or surgical resection may be more appropriate. [64] If a decision has been made that esophageal bougienage is appropriate, several considerations are involved in choosing the technique. If all types of dilators had equal efficacy and safety profiles for bougienage of routine esophageal stenoses (an unproven assumption), the physician would use the least costly system. [61] On the other hand, if inherent advantages are assumed for a particular system, there is a tendency to increase its use. The claimed advantages of balloon dilators over other systems have included ease of passage, dilation of the stricture alone, and radial as opposed to vector force applied to a stenotic wall. [1] Ideally, such balloons should be safer and more effective, claims that cannot be substantiated with available data. If fluoroscopy is used to ensure waist dilation, total costs using this technology in our institution are threefold higher than dilation with polyvinyl dilators and eightfold higher than mercury bougienage. [61] Given this and the fact that such balloons are marketed as one-time use devices, balloon technology is better limited to areas of the gut not accessible to other types of dilators. The exception to this statement is achalasia, in which polyethylene balloons have proven both effective and much easier to pass than conventional dilating systems.

Nonesophageal Strictures

Stomach. The only large series of gastric dilation is limited to polyethylene balloon dilation, and series with long-term follow-up report conflicting results. Since the initial description of successful hydrostatic dilation in a patient with pyloric stenosis and gastric outlet obstruction, [65] several series have been published using this technique. McLean and colleagues [66] radiographically dilated 94 GI strictures in 92 patients, with a mean follow-up of 389 days in 80 patients. Thirty-three of these patients had various forms of gastric stenosis. Technically, 25 of the 33 were successfully dilated, and 70% were symptom-free at 1 year. Perng and associates [35] reviewed 42 patients with benign gastric outlet obstruction. At a median follow-up of 2 years, two thirds of patients had sustained improvement and one third required surgery. The symptom-free rates at 1, 2, 3, and 4 years were 85%, 78%, 69%, and 69%, respectively, and the independent prognostic factor for failure was need for more than two courses of dilation for symptom relief. Lau and colleagues [36] reviewed 45 patients who had successful gastric outlet obstruction dilation, 4 of whom had rapid re-obstruction and were found to have malignancy. At a median follow-up of 39 months, more than 50% of patients required surgery, 18 for recurrent obstruction, 2 for interval perforations, and 1 for bleeding.

Small bowel. Because of the problems of access to the small intestine, there are few data regarding dilation of proximal small bowel strictures. [67] There are, however, multiple series using balloon technology to dilate ileal or anastomotic ileocolonic strictures in Crohn's disease. [30] Dilation of 27 anastomotic strictures in patients with Crohn's disease led to no complications, and after 7 to 38 months, 18 patients remained free of obstructive symptoms. [68] A prospective study of 55 patients with Crohn's disease with 59 ileocolonic strictures noted 90% technical success with complications in 11% (6 perforations). [37] Complete long-term symptom relief was noted in 34 (62%) patients. Corticosteroid injections into inflamed small bowel following anastomotic balloon dilation may decrease restenosis rates further. [69] As of this writing, however, results remain anecdotal and data should be placed into the perspective of surgical resection or stricturoplasty.

Colon. Most reported colonic dilations have been used for rectal or anastomotic stenoses. [25], [26] Linares and colleagues [70] dilated 33 anorectal strictures in patients with Crohn's disease with a variety of techniques; half experienced short- or long-term symptomatic relief. Pietropaolo and associates [71] dilated 42 patients with postoperative strictures with either balloons or bougies. There was no morbidity or mortality with a 2.4% failure rate. A combination of various types of dilators with endoscopic electrosection in 39 patients with benign anastomotic strictures resulted in no complications or symptoms in any patient at a mean follow-up of 25 months. [26] Finally, Virgilio and colleagues [72] used a 30- to 40-mm achalasia dilator in 18 patients with anastomotic strictures following resection for colorectal cancer. A total of 45 dilating sessions were performed in 17 of the patients. Long-term symptom relief was described in 16 patients (94%) and complications were limited to one episode of bleeding and a punctiform bowel perforation.

Clinical implications. The major question for nonesophageal stenoses is the appropriateness of stenosis dilation compared with alternative treatment modalities. Using pyloric stenosis as an example, most series are small and follow-up of dilated patients suboptimal. Nevertheless, a 60% to 65% good-to-excellent symptomatic response at 1 to 2 years has been reported, [35], [36] and this should be compared with the results of drainage or resective therapy for benign gastric outlet obstruction. [29] A cost analysis comparing pyloric balloon dilation with vagotomy and pyloroplasty at our institution [73] showed balloon dilation to be cost-effective in the short term at one tenth of the total costs for vagotomy and pyloroplasty. However, long-term acid suppression was not considered. Additional unanswered questions include the relative efficacy of balloons as opposed to polyvinyl dilators for rectosigmoid stenoses, and the rapidity with which stenoses at various sites can be safely dilated.

Stent Placement

Esophagus

Conventional. Conventional stents can be successfully placed in approximately 90% to 95% of patients with malignant esophageal obstructions, with mean patient survival of 2 to 4 months. [9] Terminal events frequently include tumor cachexia or subacute stent-related problems such as erosions into major vessels or aspiration.

Expandable prostheses. Numerous series defining long-term results of expandable stent placement for malignant dysphagia have been published. [9], [11], [52], [74], [75] As noted, these results are contingent, in part, both on stent design and initial successful palliation of dysphagia. Nevertheless, in most series, survival appears identical to that following conventional prosthesis insertion: 2 to 4 months. [9] For instance, placement of 138 Ultraflex stents in 114 patients led to improvement in dysphagia score (3.5 to 1.5), although tumor ingrowth occurred in two thirds of patients and 35 endoscopic interventions were required. [44] No significant differences in implantation success or efficacy were seen in a series using 3 types of expandable stents in 87 patients, but 22% of patients treated with Wallstents required early reintervention, compared with 37% in the Ultraflex group and 10% in the Z stent group. Subsequent reinterventions were required in 43%, 35%, and 10% in each group. [76] Comparable palliation and complication rates have been noted in studies comparing Z, Ultraflex and Flamingo prostheses. [77] Despite the invariable stent dysfunction noted in these series, expandable prostheses have also been used occasionally in benign disorders and may have a role to play for anastomotic leaks in a subset of patients with refractory benign stenoses. [78], [79]

Clinical implications. Patients with malignant dysphagia who require any form of endoscopic intervention generally have either widespread disease or an aggressive tumor biology that precludes prolonged survival. Our study comparing 85 patients in whom conventional or expandable prostheses were placed for malignant dysphagia documented comparable levels of prestent and poststent dysphagia (3[0.9]/0.4[0.9] vs. 2.7[1.1]/0.9[1.4]), fistula occlusion (13/15 vs. 13/14), and survival (87[72] vs. 90[80] days), respectively. [43] Other studies comparing conventional prostheses to Nd:YAG laser document a 2- to 4-month mean survival regardless of the treatment modality.

Nonesophageal Strictures

Stent placement. Data relating to stent placement in malignant stenoses of the small bowel and colon are limited, [15], [16], [80] and data in benign disease are almost nonexistent. Baron and colleagues [39] inserted a variety of expandable prostheses for obstructing colorectal cancer in 25 patients not thought to be operative candidates. Two thirds of these patients were adequately palliated, although there were three perforations and one procedure-related mortality. Four patients ultimately developed stent migration and one tumor ingrowth through the prosthesis. Our group placed a variety of expandable stents in nine patients with widespread obstructing malignancies. [78] Indications included afferent loop obstruction in three, colon obstruction in three, gastric outlet obstruction in two, and stenosis of an esophagojejunal interposition in one. There was a single perforation at the time of dilation prior to stent insertion. Additional problems included migration in one case and food bolus occlusion in one patient. Three additional patients developed tumor overgrowth or ingrowth through the stent that was treated with laser. Median survival approximated 9 months.

Clinical implications. The ability to insert an expandable prosthesis into a malignant stenosis does not imply that it should be done. Particularly disturbing are claims that this is first-line treatment for obstructing colorectal cancers in lieu of surgery or as a means to facilitate bowel preparation. Although this is theoretically attractive, as virtually all patients with obstructing colorectal cancer have stage III or IV disease, there are few data to support such claims. [10] It is hoped that prospective, randomized trials answer such questions in the future. Likewise, what role, if any, expandable stents should play in patients with benign central stenoses remains to be defined. Although potentially useful in a handful of high-risk patients who have pernicious stenoses (e.g., anastomotic, radiation), experience with stent-related problems in the esophagus in patients with limited survival suggests that use in individuals with the potential for several-year survival may prove to be ill-advised unless the stent is removable and placed for several weeks or months at most.

FUTURE APPLICATIONS

The future of GI dilation is contingent upon further experience with the newer dilating systems such as CRE balloons in comparison to polyvinyl dilators. Both expanded experience and controlled clinical trials should allow improved definition of the indications, benefits, per procedure costs, and risks for individual dilating systems and for specific stenotic lesions.

Additional technologic combinations using dilators and various thermal modalities for malignant stenoses are likely to become available. Just as bipolar tumor probes have conjoined Eder-Puestow dilators with multipolar electrocautery to treat concentric esophageal or rectal neoplasms, similar electrodes have been implanted on balloons or polyvinyl dilators. A new approach is to take a quartz laser fiber and surround the tip with a balloon. The balloon keeps the fiber tip in the center of the lumen. These catheters may ultimately allow treatment of nonresectable biliary, gastroesophageal, or rectal neoplasms by transmission of laser energy. Photodynamic therapy, in turn, may play an increasing role in the treatment of malignant stenoses and argon plasma coagulation may be used in lieu of Nd:YAG laser to open a lesion or facilitate stent placement.

The future of GI dilation may also include the development of dilators that can detect a fall in wall tension or resistance within a tissue during dilation. This fall in tension may predict that the dilation is adequate and that further lumenal enlargement may be unnecessary and hazardous. Devices such as these should improve safety.

Finally, as previously noted, dilation will be increasingly used in conjunction with expandable stent technology, potentially even in the setting of benign stenoses. These expandable stents, undergoing refinement both in the endoprostheses themselves and in their delivery systems, hold at least the potential of prolonged stricture patency, which would preclude the need for repeated dilation sessions. At a minimum, prostheses will be fashioned from newer materials, and prototypes testing both plastic and thermocoupled metals have been described. [81], [82]

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CHAPTER 146

Jeffrey L. Ponsky and Brian J. Dunkin

PERCUTANEOUS ENDOSCOPIC GASTROSTOMY

The percutaneous endoscopic gastrostomy (PEG) was introduced in 1980 as an alternative to operative gastrostomy for those patients requiring long-term intubation of the stomach. [1] It was devised as a safe and effective nonoperative means of providing long-term enteral nutrition. The method has become widely practiced, with about 150,000 PEGs being placed in patients in the United States annually.

The concept of a surgically created fistula between the stomach and abdominal wall was first conceived by Egeberg in 1837 and attempted by Sellidot in 1849. A variety of surgical approaches were developed, culminating in the more popular Janeway and Stamm methods. [2] Both were effective in creating a gastrocutaneous fistula, but use of these techniques was somewhat limited because the patients were frequently poor surgical candidates and these procedures required a laparotomy, often with general anesthesia.

Thus, delivery of enteral nutrition was most often by means of nasoenteric tubes. This was effective but associated with numerous frustrations and complications. These tubes were frequently large in caliber and constructed of stiff rubber or plastic, making them uncomfortable. Frequent plugging of the tubes necessitated replacement, and it was not unusual for them to become dislodged, accounting for up to 60% of tube removals. [3], [4] Esophagitis also developed from irritation of the esophageal mucosa by the tube, and gastroesophageal reflux was common because the tube rendered the esophagogastric junction incompetent. [5], [6] This reflux frequently led to the development of aspiration pneumonia. Long-term use of nasoenteric tubes also caused parotitis and erosion of the nasal cartilage.

In 1980, the technique of PEG was introduced. This method allowed the formation of a tube gastrostomy without laparotomy or general anesthesia. Safety of the procedure was ensured by endoscopic visualization of the tube insertion, and placement directly into the stomach reduced the risk for complications encountered with the nasoenteric tube. The original method described by Ponsky and Gauderer has been called the *pull technique*. [7] Two modifications of the original method have been introduced and have gained some popularity. These are referred to as the *push* or *Sacks-Vine technique* and the *introducer* or *Russell technique*. [8], [9] All of these methods have been shown to be safe and effective.

Complications of percutaneous gastrostomy, although not frequent, are known to occur and are well documented. Careful attention to patient selection and the details of the method may help to reduce the incidence of undesirable outcomes. The increasing ease by which enteral access is established with percutaneous gastrostomy has led to tremendous growth in the application and use of this procedure. Moral and ethical dilemmas arise as the technique is applied to an increasingly debilitated population of patients. Further study of the technique and its long-term outcome has provided answers to some of these problems.

METHODS

Patient Preparation

Proper preparation can help avoid complications. The patient should not be fed for 8 hours before the procedure. If an antibiotic with good gram-positive coverage is not already being administered, then a single intravenous dose should be given. The most common organisms cultured from PEG site infections are *Staphylococcus aureus* and ß-hemolytic streptococci. Cefazolin is very good at covering these and other commonly encountered organisms. [10] The patient should be positioned supine with the head elevated to reduce the risk for aspiration. Because it can be difficult to intubate the esophagus in this position, it is sometimes preferable to start the endoscopy in the left lateral decubitus position and then reposition the patient supine. Suction should be available to manage oral-pharyngeal secretions, and the patient should be maintained on supplemental oxygen. In addition, blood pressure and pulse oximetry should be monitored by a nurse in attendance throughout the procedure.

The Pull (Ponsky-Gauderer) Technique

Once positioned, the patient is given intravenous sedation, and the abdomen is washed and draped in a sterile fashion. The room lights are turned down as the gastroscope is passed into the stomach. Transillumination of the abdominal wall by the light of the scope is an indication that intervening structures have been displaced and that the stomach lies in contact with the abdominal wall. The site selected for placement of the gastrostomy should be one at which transillumination is well seen. Finger pressure by the assistant at this point produces a clear indentation of the gastric wall as seen by the endoscopist (Fig. 146-1). This indentation should be covered by an open polypectomy snare passed through the endoscope. To enhance the safety and accuracy of PEG placement, the use of the “safe tract” technique described by Foutch has been very useful. [29] Once the best site of finger indentation or transillumination is selected, the syringe with local anesthetic is advanced only a small distance beneath the skin. Suction is applied to the syringe barrel as it is advanced more deeply. The appearance of air in the syringe barrel and the visualization of the needle tip in the stomach by the endoscopist should occur simultaneously. Should air appear in the syringe barrel before the needle tip appears in the stomach, an alternative site should be selected because this usually indicates that an intervening air-containing viscus, such as colon or small bowel, has been punctured.

FIGURE 146-1. The endoscopist observes a distinct indentation of the gastric wall as the assistant applies finger pressure to the abdominal wall at the proposed site of puncture.

Local anesthetic is injected into the abdominal wall at the proposed area of puncture, and a 1-cm incision is made in the skin. An intravenous cannula is thrust through the incision and into the gastric lumen. If the polypectomy snare is correctly positioned, the thrusting needle will enter the center of its loop. If not, the snare is quickly looped around the cannula and tightened near the point where the cannula exits the gastric mucosa (Fig. 146-2). A long, stout suture or wire is passed through the needle into the gastric lumen, and the snare is moved from around the cannula to tighten around the suture itself (Fig. 146-3). The suture is pulled with the endoscope from the stomach and out of the mouth. The drainage end of the gastrostomy tube is affixed to the suture at the patient's mouth (Fig. 146-4). The tube is well lubricated, and pull is begun on the abdominal end of the long suture so that the gastrostomy tube moves down the esophagus and exits the abdominal wall (Fig. 146-5). The gastroscope is reinserted to check the position of the head of the tube, and care is taken to avoid excessive tension on the head of the catheter. The endoscope is removed, and an outer bolster is applied, which holds the gastric and abdominal walls in close contact.

FIGURE 146-2. The snare is looped around the cannula and tightened near the point where the cannula exits the gastric mucosa.

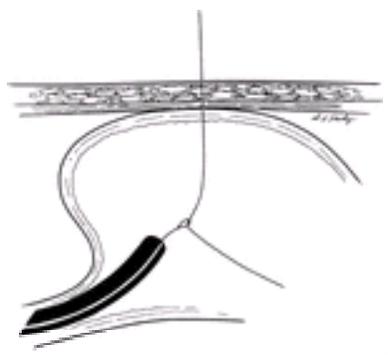

FIGURE 146-3. The snare is tightened around the suture and pulled out of the patient's mouth using the gastroscope.

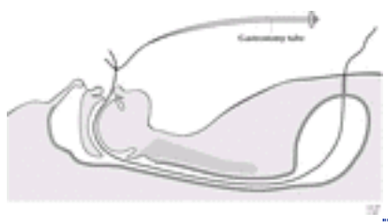

FIGURE 146-4. The gastrostomy tube is affixed to the suture at the patient's mouth.

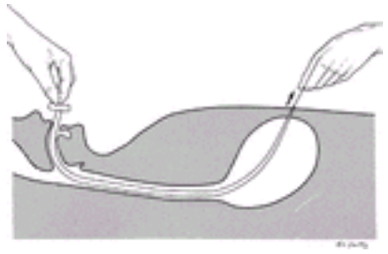

FIGURE 146-5. The assistant pulls on the abdominal end of the suture, and the gastrostomy tube proceeds down through the esophagus and stomach to exit the abdominal wall.

The Push (Sacks-Vine) Technique

The push method differs little from the pull technique. The patient is repared, the puncture performed, and a guidewire grasped and pulled from the patient's mouth in the same manner (Fig. 146-6). In this procedure, however, a special gastrostomy tube with a long tapered dilator end is pushed over the guidewire. Both ends of the wire must be held under tension as the tube is pushed down the esophagus and exits the abdominal wall (Fig. 146-7). Once the tube emerges from the abdominal wall, it may be grasped and pulled the rest of the way into its final position. The gastroscope is reinserted to ensure that the head of the catheter lies in contact with the gastric mucosa. An outer crossbar or faceplate is added to fix the abdominal and gastric walls together.

FIGURE 146-6. As in the pull technique, the cannula is inserted into the stomach, a guidewire is passed through it, and the wire is grasped and pulled out of the mouth using the gastroscope.

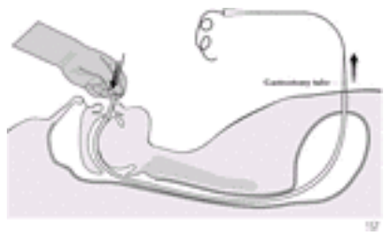

FIGURE 146-7. Both ends of the wire are held under tension while the gastrostomy tube is pushed down the esophagus and out the abdominal wall. Once the tube emerges from the abdominal wall, it is grasped and guided into position.

The Introducer (Russell) Technique

The percutaneous method for insertion of a central venous catheter as described by Seldinger has been adapted to the performance of gastrostomy placement in the introducer method. Developed by Russell and colleagues, [9] this method allows percutaneous placement of a gastrostomy tube without passing the tube through the oropharynx and with only a single insertion of the endoscope. The gastroscope is passed into the stomach, and the room lights are dimmed. Transillumination of the abdominal wall and finger pressure at the point of best transillumination are used to identify the proper site for puncture. Once the site for insertion has been established, the skin is anesthetized, and a small incision is made. A needle is introduced into the gastric lumen, and a guidewire is passed through the needle. The needle is removed, and an introducer with an outer, peel-away sheath is passed over the guidewire into the stomach (Fig. 146-8). The wire and introducer are removed, leaving only the sheath in the stomach. A balloon catheter is introduced through the sheath and inflated (Fig. 146-9). The sheath is removed, and the balloon of the catheter is pulled up to the gastric mucosa. Either the catheter is sutured to the skin, or an outer faceplate is applied to hold the gastric and abdominal walls in approximation.

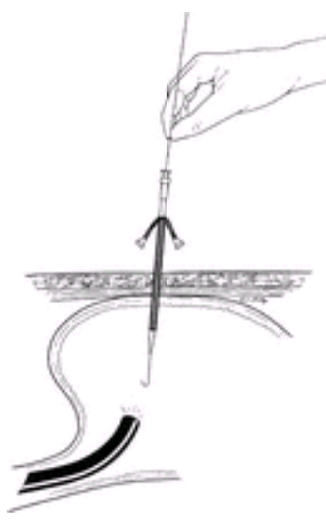

FIGURE 146-8. The introducer and its outer sheath are passed over the guidewire into the gastric lumen.

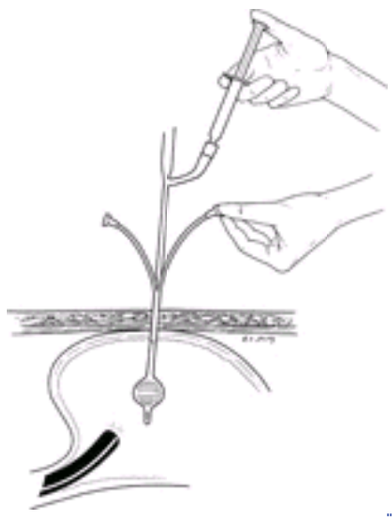

FIGURE 146-9. The introducer and guidewire are removed, and the balloon catheter is inserted through the sheath. The balloon is inflated, and the sheath is peeled away.

Comparison of Methods

All of these techniques for the performance of percutaneous gastrostomy have been widely used and found effective. Each provides benefits that should be considered in choosing a method for a particular patient. [11]

The pull technique provides a great deal of control for the physician. The puncturing needle is stabilized with the surrounding snare, ensuring that loss of air from the gastric lumen does not result in loss of the puncturing needle. The gastrostomy tube is pulled down the esophagus and out of the abdominal wall in a direction that apposes the gastric and abdominal walls. This should minimize leakage around the puncture site. The soft button of the pull PEG trailing free behind the tube may be more deformable than that of the push PEG, thus allowing it to traverse a narrowed esophagus more readily. The push method adds the stability of a guidewire to act as a trolley on which the tube can ride as it passes through the alimentary tract. Both of these methods have the potential of introducing oral bacteria or tumor cells from a lesion in the oropharynx or esophagus into the tissues of the abdominal wall. Thus, infection and metastasis of carcinoma are potential complications with

these techniques. Both require a second passage of the endoscope to ensure correct positioning of the catheter head against the gastric wall.

The introducer method enjoys the advantage of a direct puncture insertion of the tube. This avoids the potential risk for infection from oral flora and possibly reduces the risk for cancer seeding to the abdominal wall. The method permits insertion of a gastrostomy in patients with narrow lesions of the esophagus without the fear of disrupting such lesions while the catheter is positioned.

In the introducer technique, the endoscope serves only an observatory function. However, the forces of insertion with the introducer method push the stomach away from the abdominal wall and may lead to the creation of a submucosal tunnel in the stomach without puncture of the mucosa. Loss of control of the punctured stomach and leakage of gastric contents are also more likely to occur. Finally, the tubes used for this procedure are of small diameter and suffer from premature extrusion secondary to deflation of the balloon catheter.

In practice, each of these methods has been demonstrated to be effective and to have a low rate of complications. Experienced endoscopists are familiar with all of these techniques and select the one with which they are most comfortable and that best suits the needs of the patient.

Radiologically Placed Gastrostomy Gastrostomy tubes may be placed under radiologic guidance. A number of studies have examined these methods and found that they compare favorably with endoscopically guided insertion. [12] The best indications for radiologically guided PEG include esophageal stenosis due to malignant or benign stricture and the inability to place an endoscopically guided PEG owing to the inability to accurately localize a safe puncture site. Although this method of insertion is effective, the tubes used for the gastrostomy are frequently of small caliber and prone to clogging. Tube replacement may also be more involved after radiologic insertion.

INDICATIONS

The PEG was first conceived as a means of providing long-term enteral nutrition to those patients who are unable to ingest food by mouth but have a functioning gastrointestinal tract. Nutritional support should be initiated in all patients anticipated to be without nutrient intake for 2 or more weeks. If the patient has a functional gastrointestinal tract and is expected to require enteric feeding for less than 30 days, then a nasogastric or nasoenteric tube should be used. If it is anticipated that enteric feeds will be required for more than 30 days, however, then a tube gastrostomy or enterostomy should be considered. Such patients may have severe neurologic or developmental disorders, obstruction of the oropharynx from trauma or tumor, or a critical illness requiring prolonged endotracheal intubation.

As the safety and efficacy of the PEG has been demonstrated, the population of patients benefiting from it has expanded. PEGs have been placed in burn patients (even through burned skin) to facilitate enteric feeding of high caloric density formulas. [13] They also have been placed in patients about to receive neoadjuvant therapy for esophageal cancer [14] and head and neck carcinoma. [15], [16] Victims of severe maxillofacial injury have benefited from PEG placement as well. [17]

Non–alimentation-related applications also have been described. These include decompression of the stomach for gastric atony, uncorrectable obstruction, carcinomatosis, and gas bloat following Nissen fundoplication. [18], [19] and [20] The method also has been used for the administration of unpalatable medications to children [21] and the refeeding of bile to patients with external biliary fistula. [22] Finally, multiple PEGs have been placed in patients with paraesophageal gastric herniation and gastric volvulus as a method of gastropexy. [23]

CONTRAINDICATIONS

The contraindications to PEG placement can be grouped into three categories: absolute, relative, and potential. The absolute contraindications to PEG placement include uncorrectable coagulopathy, correctable intestinal obstruction, peritonitis, peritoneal dialysis, gastric varices, absence of a stomach, or any process that precludes endoscopic access to the stomach.

Relative contraindications are those situations traditionally considered absolutely contraindicated but that more recently have been successfully addressed using ultrasound imaging, better medical preparation of the patient, or the addition of a minor surgical procedure. The first of these is massive ascites. Patients with massive ascites are at major risk for complication from PEG placement. Successful placements have been described, however, using preprocedure treatment with beta-blockade, diuresis, nitrate administration, and paracentesis. [24], [25] The second relative contraindication to PEG placement is the inability to transilluminate the abdominal wall, usually because of morbid obesity or the presence of an intervening structure between the stomach and the abdominal wall. Endoscopic and transabdominal ultrasound have been used to ensure that no intervening structures are present and to guide the passage of a spinal needle through a thick abdominal wall. [26] Alternatively, a cut-down through the skin and subcutaneous tissue under local anesthesia has been used in an obese patient so that fascia could be visualized and a PEG safely placed. [27] Without these additional maneuvers, however, the inability to transilluminate the abdominal wall is a contraindication to PEG placement.

Finally, there are those situations where PEG placement is not contraindicated but should be performed with caution because there is a significant potential for a complication to occur. These include prior surgery, distended loops of small bowel, presence of a ventriculoperitoneal shunt, or severe cardiac disease. Previous abdominal surgery is not a contraindication to PEG placement if there is meticulous adherence to the details of the procedure, including transillumination of the abdominal wall. [28] A safe tract should be identified in these patients by looking for the aspiration of air from only the stomach during passage of the angiocatheter. [29] These same principles apply when treating a patient who has distended loops of small bowel. The presence of a ventriculoperitoneal shunt is not a contraindication to PEG placement as long as care is taken to avoid the catheter. [30] A preprocedure abdominal radiograph can be helpful. Severe underlying heart disease increases the risk for major complication in PEG placement, possibly related to the use of intravenous sedation. [31] Cappell and Iacovone [32] have shown, however, that a PEG can be safely placed within 1 month after myocardial infarction if it is considered essential and the patient is otherwise stable.

COMPLICATIONS

Large cumulative retrospective studies have reported that 10% to 16% of both adult and pediatric patients have at least one complication after PEG placement. [33] The overall mortality rate is 1%, with a major complication rate of 3% and a minor complication rate of 13%. [34] Wound infections account for up to 30% of all minor complications. These are caused by multiple organisms, but most commonly by *S aureus* and ß-hemolytic streptococci. Jain and colleagues [34a] have shown that a single prophylactic dose of cefazolin covers 72% of the organisms encountered in PEG infections and will reduce the rate of infection after PEG placement from 28.6% to 7.4%. In contrast, cefoxitin is only effective against 33% of the organisms encountered. Infection usually becomes evident as erythema around the gastrostomy tube site several days after the procedure, with local tenderness and slight edema of the skin. The patient often demonstrates a low-grade fever and leukocytosis. Early recognition and incision and drainage of the area under local anesthesia most often resolve the problem. Failure to identify and treat this problem at an early stage can result in necrotizing infections of the abdominal wall and death. Necrosis of the tissue of the abdominal wall that is interposed between the head of the tube and the outer bolster may play a major role in the occurrence of infection at a PEG site. Excessive tension applied to the bolster acts as a tourniquet, producing ischemia of the underlying tissue. Loose contact is all that is required between the outer bolster and skin. If a dressing is required, it should be placed over the bolster, not under it, for the same reason. It is a good idea to begin systemic antibiotics for all infections around the tube and to continue them until the infection is resolved. Also, an adequate skin incision in the abdominal wall allows room around the gastrostomy tube for the egress of bacteria.

Extrusion of the head of the tube from the gastric lumen into the subcutaneous tissue (the buried bumper syndrome) has been frequently encountered. This is almost certainly caused by excessive tension that has been applied to the tube. To prevent leakage of gastric contents and ensure adhesion of the stomach to the abdominal wall, the physician may pull up on the gastrostomy tube and push downward on the outer bolster. Although this fixes the stomach to the abdominal wall, it also produces ischemia of the intervening abdominal wall tissue, with subsequent necrosis. This can be avoided by ensuring that no tension is present when the head of the catheter is placed in contact with the gastric mucosa and that the outer bolster only loosely approximates the skin. Such approximation of the gastric and abdominal walls results in satisfactory adhesion without tissue necrosis.

Leakage of feedings into the peritoneal cavity may occur after percutaneous gastrostomy. This is usually the result of separation of the gastric and abdominal walls and often is due to necrosis of the abdominal wall tissue caused by excessive tension on the catheter. Patients who develop abdominal tenderness, fever, or leukocytosis should be evaluated for leakage and the resultant peritonitis. This may be done by instilling water-soluble contrast material into the gastrostomy tube under fluoroscopic guidance. Intraperitoneal extravasation indicates that something has gone awry. If the contrast study results indicate that the head of the tube remains in the stomach and that the extravasation is around the tube, the clinician may pull up on the tube, providing a bit more tension to seal the leak, and place the

tube to gravity drainage; intravenous fluids and antibiotics can be administered. If the contrast study results reveal complete separation of the gastric and abdominal walls with dislodgment of the tube from the stomach, the tube should be pulled from the abdominal wall, a nasogastric tube inserted to effect gastric drainage, and intravenous fluids and antibiotics begun. A patient whose PEG has been pulled out inadvertently less than 2 weeks from the time of insertion should be treated similarly. If at any time the patient's condition begins to deteriorate or signs of peritonitis worsen, exploratory laparotomy with operative repair should be performed.

Gastrocolic fistula has rarely been known to occur after percutaneous gastrostomy. This type of fistula may be caused by puncture of the colon at the time of gastrostomy or pinching of the colon between the gastric and abdominal walls with subsequent necrosis of the colonic wall and fistula formation. This complication usually becomes apparent after several weeks, with the development of severe diarrhea after feedings. It may be documented with an upper gastrointestinal series or barium enema. In nearly all cases, the condition may be treated by removing the gastrostomy tube. The fistula closes rapidly once the tube is removed.

Progressive enlargement of the gastrostomy stoma around the gastrostomy tube may occur in some patients. Although this, too, may be the result of excessive tension on the tube, it is also occasionally seen in patients in whom the gastrostomy tract is well established. Poor nutritional status may play a part in the development of this problem, as may excessive movement of the tube at the skin level. The temptation in addressing this problem is to remove the gastrostomy tube and replace it with a larger one that fills the hole. This solves the problem for a short time, but the tract soon enlarges again. A better solution is to remove the tube entirely and allow the tract to close. After it has closed a bit, a new, smaller tube may be inserted. Also, minimizing movement at the skin by securing the tube properly or placing it through a baby bottle nipple or a similar stabilizing device is effective.

Pneumoperitoneum is a frequent occurrence after percutaneous gastrostomy. This may be the result of air escaping around the puncturing needle. Routine radiographs of the abdomen are unwarranted. Air in the abdominal cavity has been shown to last for up to 5 weeks after PEG placement. Patients found to have pneumoperitoneum after gastrostomy must be clinically evaluated. In the absence of abdominal tenderness, leukocytosis, or fever, there is no need for further evaluation. However, the patient who demonstrates any of these should be evaluated using a water-soluble contrast study through the gastrostomy tube for signs of separation or intraperitoneal extravasation. One case was reported of tension pneumoperitoneum during PEG placement causing hemodynamic compromise, but this is exceedingly rare. [35]

Neoplastic seeding to the skin from both oropharyngeal and esophageal cancers has been reported after PEG placement. [36] If the gastrostomy is being placed for palliation, this may not be an issue. However, if placement is part of the patient's therapy, as in preparation for neoadjuvant radiation therapy for esophageal carcinoma, then this must be considered. There may be advantages to using an introducer technique in this setting, but this has not been tested.

Some clinicians question the need for repeat endoscopy once the PEG tube has been pulled (or pushed) into position. Endoscopic intubation of the esophagus can be difficult in a supine, often intubated, patient. It is important, however, to confirm the position of the PEG bumper because obstruction from malpositioned tubes at both the gastroesophageal junction and pylorus have been reported. Endoscopic examination also helps confirm proper tension between the gastric wall and the bumper and rules out any associated injury such as laceration of the esophagus from an improperly grasped wire (wire snared close to its tip). [37] Repeat passage of the endoscope can be simplified by attaching the end of the scope to the button of the PEG tube before it is pulled into the oropharynx. The button of the tube is snared on one side only with a polypectomy snare preloaded through the endoscope. With gentle pressure, the endoscope will follow the PEG tube as it is being pulled into position. Once in the esophagus, the button is released, and the endoscope is passed into the stomach under direct vision.

Good skin care is important after gastrostomy. It is common to see a foreign body reaction around the tube with some exudate or granulation tissue. This is usually easy to treat. The exudate is merely swabbed away with hydrogen peroxide. The site should be left open to the air. Granulation tissue may be cauterized with silver nitrate. Occlusive dressings should be avoided because they lead to maceration of the underlying skin.

FEEDING

Once the PEG has been placed without complication, the next issue is how to feed through it. Intermittent feeding is preferred because it is easy to administer (no pump required), well tolerated, and physiologic. The formula is allowed to flow in by gravity over a period of 30 to 60 minutes. Rapid bolus feeds should be avoided because they decrease lower esophageal sphincter pressure to incompetent levels and are associated with free gastroesophageal reflux. Patients should be placed in a semirecumbent position to reduce the risk for aspiration. Most endoscopy units start feeding 24 hours after PEG placement with the initiation of a continuous infusion of water at 50 mL/h. Once this has been tolerated for 4 hours, the infusion of a full-strength polymeric formula is begun at 50 mL/h and advanced by 25 mL/h every 12 hours as tolerated until the goal rate is reached. This method is anecdotal, however, and some clinicians have demonstrated that a continuous infusion of full-strength formula at 30 mL/h can be safely initiated only 3 hours after PEG placement. [38] The choice of formula is based on the patient's caloric needs, tolerance, and associated medical conditions. Fiber-containing formulas have not been shown to be of any advantage. Residual volumes should be checked on a regular basis until tolerance of the regimen has been demonstrated. If found to be more than 100 mL, this should prompt concern for intolerance but should not cause cessation of the tube feedings because a repeat measurement an hour later is often normal. [39] Residual volumes of more than 100 mL for successive measurements, however, should prompt cessation of feeding. Tubes smaller than 10 French in size have been shown to be unreliable for the determination of a residual volume. [40]

Aspiration is one of the most important complications associated with tube feeding. The prevalence is reported to be from 2% to 95%, depending on the definition and how it is measured. It has been shown that patients are at a higher risk for aspiration from nasogastric tube feedings in comparison with PEG feedings. [5] It is less clear if feeding into the jejunum decreases the risk for aspiration when compared with gavage feeding. Most studies addressing this issue are too small, are poorly designed, or do not document the position of the feeding tube. In general, direct jejunal feeding is not required unless a patient has a history of gastroesophageal reflux or has recurrent tube feeding aspiration through a PEG.

PERCUTANEOUS ENDOSCOPIC JEJUNOSTOMY

Percutaneous endoscopic jejunostomy is a modification of PEG intended to accomplish concomitant jejunal feeding and gastric decompression. The method is similar to that of PEG, with the gastric tube acting to anchor the apparatus and decompress the stomach while a longer enteric tube is passed through the PEG and positioned in the distal duodenum or proximal jejunum. [41] The proposed advantage of this system is to decrease the risk for aspiration, but early studies failed to demonstrate this advantage. The tips of the "jejunal" tubes used for these studies, however, were often only in the proximal duodenum. More recent studies have confirmed placement of the tube into the distal duodenum or proximal jejunum using an over-the-wire technique and have demonstrated a virtual elimination of aspiration associated with feeding. [42] The indications for percutaneous endoscopic gastrostomy or jejunostomy tube placement are tracheal aspiration, reflux esophagitis, gastroparesis, insufficient stomach from previous resection to perform PEG placement or to tolerate gavage feeding, or enteral access in patients with partially obstructing lesions from gastric or pancreatic carcinoma. Correct placement of these tubes has proved difficult. Traditionally, a suture is attached to the tip of a jejunal tube before passing it through the PEG and into the stomach. The suture is grasped with a biopsy forceps loaded through the endoscope and dragged under direct vision into the duodenum. Deep passage is often impossible, however, and the tube frequently drags back into the proximal duodenum or stomach during removal of the endoscope. A pediatric colonoscope or enteroscope may be used to ensure passage of the jejunal tube into the distal duodenum or proximal jejunum. More recently, various guidewire techniques have been described to assist in placement. Duckworth and colleagues [42, 43] described passing a guidewire through a 24-French PEG tube and grasping it with a biopsy forceps passed through the endoscope. The wire is dragged under direct vision into the distal duodenum or proximal jejunum. While still holding it with the forceps, tension is applied to the free end of the wire to straighten it. A 12-French jejunal feeding tube is then passed over the wire. The endoscope is removed by a rocking motion followed by removal of the guidewire. Tube placement is confirmed by abdominal radiograph. An initial placement success rate of 100% has been reported, but there is a 20% long-term tube failure rate owing to malpositioning or occlusion. Chaurasia and Chang [44] have described passing a pediatric bronchoscope through the PEG and into the distal duodenum to gain access to the bowel. A wire is passed through the scope and guided beyond the ligament of Treitz using fluoroscopy. The jejunal feeding tube is then passed over the wire and into position. Finally, Leichus and colleagues [45] have described passing the endoscope into the stomach and through an open polypectomy snare that has been passed into the stomach through the PEG. The tip of the endoscope is placed into the distal duodenum, and a guidewire passed through it and beyond the ligament of Treitz. The wire is kept in position while the endoscope is withdrawn, leaving the peroral wire to be grasped at the intragastric segment by the trans-PEG polypectomy snare. The snare is pulled so that the oral end of the wire is retrieved out the PEG. A jejunostomy tube is then passed over the wire and into position, confirmed by fluoroscopy.

To overcome some of the placement difficulty with percutaneous endoscopic gastrostomy and jejunostomy systems, Shike and colleagues [46] described placing a feeding tube directly into the jejunum using a similar technique to that of PEG. This was initially used only in patients with partial or total gastrectomy but has subsequently been used in patients without gastric resection. Such placement may require use of a push enteroscope or pediatric colonoscope, and fluoroscopic guidance is useful. When successful, this procedure obviates the problem of tube migration and ensures placement beyond the ligament of Treitz. It is also easier to place larger-caliber tubes that are less likely to fail from occlusion. When used in combination with a PEG, better decompression of the stomach can be obtained

because the PEG tube is not partially occluded by the percutaneous endoscopic gastrostomy and jejunostomy system. There is a greater potential for complications, however. The mobility of the jejunum and the higher likelihood for intervening structures between it and the abdominal wall make strict adherence to technique mandatory. The thin wall of the jejunum is also more likely to allow leakage of enteric contents around the tube. Finally, the direct percutaneous endoscopic jejunostomy tube can act as a “maypole” around which the small bowel can twist and obstruct.

REMOVAL AND REPLACEMENT OF PERCUTANEOUS ENDOSCOPIC GASTROSTOMIES

PEG tubes may require replacement when the tube has become frayed, obstructed, or develops a leak. Additionally, in some patients, an enlarging stoma may be the result of excessive tube motion at the skin level. In such cases, removal and replacement with a skin-level device may be advantageous. Elective tube removal should not be performed until a secure tract has formed, usually at least 10 days to 2 weeks following placement.

When PEG tubes are inadvertently removed prematurely, peritonitis is a threat. When discovered immediately, such patients can often be treated with nasogastric suction and intravenous administration of antibiotics. With close observation and steady improvement, the PEG may be repeated in about 1 week. Should the patient demonstrate signs of peritonitis or sepsis, urgent laparotomy is indicated.

Some PEG tube designs require endoscopic extraction of the intragastric portion at the time of tube replacement or removal. Designs are presently available that permit extraction by means of external traction without the need for endoscopy. [47] Although PEG tubes may be replaced with balloon-tip catheters, these tubes generally last only a short time and have been known to migrate and obstruct the pyloric channel. Use of a mushroom-tip catheter or a skin-level device is preferable. Gauderer and colleagues [48] developed the concept of a skin-level gastrostomy device. This device, with an antireflux valve, permits a cosmetic solution to gastrostomy feedings. It may be placed after removal of a previous gastrostomy tube or at the time of the creation of the initial PEG. A standard tube is often preferable to the skin-level device in patients who require custodial care because it is more easily accessed by nursing personnel.

Percutaneous gastrostomy is a simple procedure with few severe complications. Close attention to detail and rapid assessment of potential problems minimizes unsatisfactory outcomes.

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CHAPTER 147

Joseph J. Y. Sung and James Y. W. Lau

ENDOSCOPIC THERAPY FOR UPPER GASTROINTESTINAL VARICEAL HEMORRHAGE

ANATOMY OF THE PORTAL VENOUS SYSTEM AND GASTROESOPHAGEAL VARICES

It is important to understand the anatomy of the portal system and the vascular system around the gastroesophageal junction to rationalize the management of variceal bleeding. The portal vein is formed behind the neck of the pancreas by the confluence of the superior mesenteric vein, which drains blood vessels perfusing the small bowel, and the splenic vein. The short gastric veins, which drain the gastric fundus, enter the spleen above the origin of the splenic vein. The left gastric vein, also called the coronary vein, enters the portal venous system within a few centimeters from the confluence through the splenic vein or the portal vein itself. The portal circulation of a normal individual is about 1 to 1.2 L/min with a low pressure of about 7 mm Hg. It constitutes two thirds of the hepatic blood flow and provides 30% to 60% of oxygen consumed by the liver. Blood vessels in the gastroesophageal region drain into the azygos vein before returning to the systemic circulation. The normal azygos venous blood flow is 150 to 200 mL/min (Fig. 147-1).

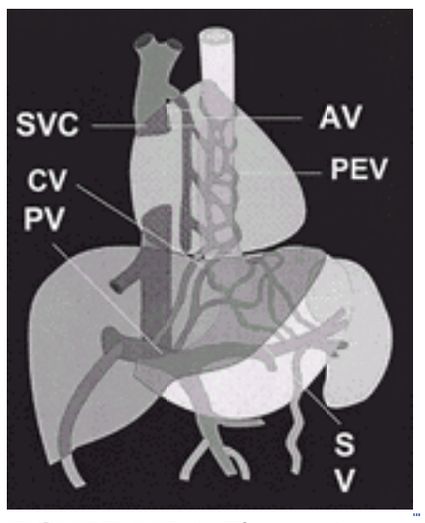

FIGURE 147-1. The anatomy of the portal venous system. AV, azygos vein; CV, coronary vein; PEV, paraesophageal vein; PV, portal vein; SV, splenic vein; SVC, superior vena cava.

Gastroesophageal varices arise from the lower esophagus and the gastric cardia. Based on radiologic imaging and corrosion casting study by Vianna and colleagues,[1] the vasculature at the esophagogastric junction can be divided into four distinct zones. The gastric zone, located just below the esophagogastric junction, is characterized by longitudinal veins draining into the short gastric and left gastric veins. The palisade zone extends 2 to 3 cm above the gastric zone and consists of uniformly distributed parallel veins running in four groups with multiple anastomoses lying in the lamina propria. Each of these groups corresponds to one esophageal mucosal fold. Blood flows in both caudad and cephalad directions in this zone depending on pressure above and below this level. The perforating zone lies above the palisade zone. It has lost the longitudinal structure but features venules penetrating through the muscle wall of the esophagus linking the submucosal vessels to the extraesophageal vessels. Above the perforating zone, the truncal zone consists of longitudinal veins and occasional perforating veins penetrating at irregular intervals.

Esophageal varices constitute a dilated venous plexus in the lamina propria of the lower esophagus. Obstruction of portal venous blood flow increases pressure in the system, which when above 12 mm Hg, causes dilation and high pressures in the periesophageal veins, rendering the valves in the perforating veins incompetent and allowing retrograde flow into the deep intrinsic veins and its tributaries. The dilated deep intrinsic veins displace the superficial venous plexus and assume a subepithelial position forming the large tortuous variceal columns in the esophageal lumen. [2] The high-pressure dilated vessels in the perforating zone force blood to flow caudally into the palisade zone where these thin parallel vessels run within the mucosal fold in a longitudinal manner. The esophageal plexus in this region is more superficial and has poor connective tissue support. The palisade zone is therefore the most common site of bleeding. Varices at the perforating and truncal zones are connected to periesophageal varices by the perforators. It has been suggested that these perforators are the cause of failed endoscopic treatment [3] and early recurrence of varices after endoscopic therapy. [4], [5] and [6]

NATURAL HISTORY OF VARICEAL BLEEDING

Thirty percent of patients with compensated cirrhosis and 60% of patients with decompensated cirrhosis have gastroesophageal varices at the time of presentation. [7], [8] Among cirrhotic patients who have no esophageal varices on diagnosis, the incidence of new varices ranges between 5% and 23% after 1 year of follow-up. [9], [10] and [11] In a recent prospective study, D'Amico and associates [12] followed 225 newly diagnosed cirrhotic patients who had no varices on first endoscopy. In the 10-year follow-up, during which patients received endoscopy every 1 to 3 years, the incidence of varices in newly diagnosed compensated cirrhosis was around 4.5% per

year, mortality rate in patients without varices at diagnosis 3% per year, and mortality rate from bleeding 0.5% per year. The risk for varices parallels the severity of liver disease as reflected by Child scoring. Platelet count also is suggested to predict development of varices. [12] Once varices have developed, they tend to increase in size. The rate of increase of variceal size from small to large is not well documented, with reported figures ranging between 5% and 31% per year. [10], [11], [12], [13] and [14] It appears that the rate of growth of varices from small to large in compensated patients is faster than the rate of de novo appearance of varices.

Among patients with cirrhosis and esophageal varices, the incidence of first variceal hemorrhage ranges from 20% to 40% within 2 years. A recent metaanalysis studying the effects of prophylactic beta-blocker treatment revealed that the mean weighted bleeding rate of the control group (without treatment) at 2 years is around 24%. [15] Interestingly, only one third of patients with esophageal varices bleed from their varices in their lifetime. [16] Thus, many patients with gastroesophageal varices may never bleed. As such, better understanding of risk factors for gastroesophageal variceal bleeding and the capability to predict which patients with varices will bleed are important in planning timely treatment.

Bleeding from gastroesophageal varices is now widely accepted as a phenomenon of “explosion”—a result of pressure inside the vascular structure. [17] Working on this theory, the risk for variceal rupture, according to LaPlace’s law, is related to the size of the varices, wall thickness, and intravariceal pressure. Several groups have confirmed that variceal size is the most important prognostic factor for variceal bleeding. [16], [18], [19] Red color signs, which include cherry-red spots and red wale markings, are also associated with a more advanced grade of varices and higher risk for hemorrhage. These signs are thought to represent focal weakness or “blowouts” in the variceal wall. Fibrin clots (“white nipple sign”) are occasionally seen over variceal columns that recently bled. [20], [21] Severity of liver disease also predicts outcome. Patients with decompensated liver disease, alcoholic hepatitis, and portal vein thrombosis fare worse than those with compensated diseases. [22], [23] The risk for bleeding from varices is correlated to the Child-Pugh score, not only because of more severe portal hypertension, but also because of other factors such as nutritional deficiency, coagulopathy, and increased fibrinolysis in advanced liver disease. [24], [25] Recently, an association between bacterial infection and failure to control bleeding has been observed. [26] Metaanalysis of prophylactic antibiotics in variceal bleeding in cirrhotic patients showed reduced morbidity and mortality. [27] A potential causal relationship between infection and bleeding has thus been put forward. [28]

A diurnal periodicity of variceal bleeding has been reported. Bleeding episodes occur more frequently in the early mornings and late evenings, [29] probably related to hyperdynamic blood flow in the portal system after a meal during these hours. [30], [31] Most variceal bleeding stops, at least temporarily, by the time the patient arrives at the hospital. Recurrent bleeding occurs in 30% to 40% of patients within the next 2 to 3 days and in up to 60% within 1 week. [32], [33] and [34] The risk for recurrent bleeding is likely related to changes in hemodynamics of the portal system, including an increase in portocollateral resistance after hypotension, increased splanchnic blood flow stimulated by blood in the gut, and an increase in portal venous pressure as a result of overzealous volume expansion during resuscitation. Bleeding-associated mortality is highest in the first 5 days after the index episode of variceal bleeding and returns to baseline levels by 3 to 4 months. [34], [35] This is the critical window for optimal treatment to improve the survival of variceal bleeders.

Several prognostic indexes have been developed to predict which patients with esophageal varices are likely to bleed. Factors incorporated include clinical and biochemical parameters, endoscopic features, and Doppler ultrasound findings. [16], [36], [37], [38] and [39] The most widely used index is the North Italian Endoscopic Club (NIEC) Index. [16] The index is based on three independent predictors of bleeding: (1) severity of liver disease (Child-Pugh class), (2) size of varices, and (3) red wale markings on the varices. Cirrhotic patients have been classified into six risk classes with a prediction rate of bleeding for each. The NIEC index has been prospectively validated in more than 1000 patients. [40], [41] and [42] The main limitation of the available prognostic indexes is that they can only identify a relatively small proportion of the patients who eventually bleed. In the NIEC index, less than 40% of the patients who eventually bled had been identified as high-risk patients. [16]

ENDOSCOPY IN VARICEAL BLEEDING

The objectives of endoscopy in gastrointestinal bleeding are to identify the source of bleeding, assess the risk for recurrent hemorrhage, and attempt to control the bleeding and prevent recurrent hemorrhage. About one third of patients with a known history of portal hypertension actually bleed from nonvariceal sources such as peptic ulcers, portal hypertensive gastropathy, and Mallory-Weiss tears. [43], [44] and [45] The use of balloon tamponade as a temporary measure without endoscopy is often futile and could be dangerous because it may aggravate an esophageal tear. The use of pharmacological methods is preferable to balloon tamponade. Somatostatin and its synthetic analogs have been shown to control bleeding from varices, providing a “dry field” for endoscopic examination. In patients with decompensated liver disease, light sedation is preferred to general anesthesia. The judicious use of endotracheal intubation is recommended during emergency endoscopy to prevent aspiration of blood. A therapeutic endoscope with a working channel of at least 3.2 mm allows for adequate suction. In the presence of a large quantity of blood and clots in the stomach obscuring the view, a large 6-mm channel endoscope can be used. In cases of blood or food material sticking to the mucosa, a water-pump system provides a forceful water jet for washing. Occasionally, an overtube in the stomach is needed for proper gastric lavage. Evacuating blood from the stomach not only allows for better vision but also helps reduce the subsequent risk for hepatic encephalopathy in cirrhotic patients with liver dysfunction.

ENDOSCOPIC THERAPIES: TECHNIQUES AND COMPLICATIONS

Endoscopic Injection Sclerotherapy

In 1939, Crafoord and Frenckner [46] pioneered endoscopic injection sclerotherapy (EST) using a rigid esophagoscope in the treatment of esophageal varices. The technique did not gain popularity until flexible endoscopy became available in the 1970s. To date, injection sclerotherapy remains the primary treatment of bleeding esophagogastric varices in many parts of the world. It is used both in the control of acute bleeding and in elective obliteration of varices. Despite its popularity, the mechanism of action of EST is not entirely clear. The hemostatic effect is unlikely to be due to acute venous thrombosis induced by sclerosant. Blood flow in varices is much faster than that in peripheral varicosities, and sclerosant injected intravariceally dissipates rapidly. [47] Furthermore, when sclerosants are injected around the varices, acute hemostasis can still be achieved at the same rate. [48] Autopsy studies indicate that venous thrombosis, mucosal ulceration, and acute inflammatory reactions can be found in the injection site as early as 2 days after sclerotherapy. A multitude of actions of EST may take effect in bringing the hemorrhage under control. [49], [50]

The technique of EST varies according to individual endoscopists. Differences include the choice of sclerosants (type, concentration, single or in combination), number of sessions, interval between sessions, site and volume of injection, and use of an overtube. Injections may be directed into the veins (intravariceal injection) or into the esophageal wall adjacent to the varices (paravariceal injection), but most endoscopists favor the former technique. When injections are made into the moving esophagus, more often than not a combination of intravariceal and paravariceal injections are given. Most endoscopists use a freehand method, injecting sclerosant into the source of bleeding or variceal column that shows a fibrin clot. Systematic injections to each variceal column hould then start from the esophagogastric junction, working upward to the midesophagus. Injections should be confined to the distal one third of the esophagus because this is the most likely area of variceal hemorrhage. Many sclerosants have been used, but there appears to be little difference in their hemostatic effects. Sclerosants are either chemical irritants such as fatty acids (e.g., sodium morrhuate, ethanolamine oleate, polidocanol) or dehydrating agents (e.g., sodium tetradecyl sulfate, ethanol, hypertonic glucose, phenol). There are few systematic comparisons between different sclerosants. The choice is largely related to personal preference and availability in different countries. For obliteration of varices, most endoscopists use 1.5% sodium tetradecyl sulfate, 5% sodium morrhuate, and 5% ethanolamine oleate. Depending on the size of the varix, 1- to 2-mL aliquots are injected into each column. A total volume of 20 ml can be injected at each session. Sodium tetradecyl may cause more severe esophageal ulceration and strictures compared with polidocanol [51] or ethanolamine oleate. [52]

Devices such as flexible overtubes and balloon catheters have been designed to provide variceal compression during EST. The Kitano tube has a tunnel running on its side for the insertion of the injection needle and a slit at the end of the injection channel. Esophageal tissue and varices invaginate through the slit into the lumen, facilitating injection. Although it confers protection against aspiration and facilitates sclerotherapy, the Kitano tube causes significant discomfort to patients and is not frequently used by experienced endoscopists. If there is no evidence of further bleeding, feeding can be resumed after several hours of observation. Recent studies support the use of prophylactic antibiotics to prevent bacteremia and associated peritonitis after EST. [26], [27] Patients may also be given sucralfate or acid-suppressive treatment after receiving EST to prevent extensive esophageal ulceration. Further injection is then continued at regular intervals until all varices are obliterated. The appropriate interval for follow-up EST after control of the initial hemorrhage remains somewhat arbitrary. Most centers adopt a weekly or biweekly treatment schedule.

Cyanoacrylate Injection

Endoscopic injection of varices with *N*-butyl-2-cyanoacrylate (Histoacryl) was first introduced in Germany. [53] The tissue adhesive coagulates almost instantaneously

when in contact with blood. To slow down the process of clotting and to allow fluoroscopic monitoring of the injection, cyanoacrylate is mixed with radiologic contrast (lipiodol) before injection. Before the injection, the working channel of the endoscope and the injector probe are flushed with 1 to 2 mL of lipiodol to prevent channel blockage. A mixture of 0.5 mL of cyanoacrylate and 0.7 mL lipiodol is injected intravariceally. The injected submucosal vessels harden, and bleeding ceases (Fig. 147-2A; see also Color Fig. 147-2A). A serpentine structure demonstrated on fluoroscopy confirms successful intravariceal injection (see Fig. 147-2B). The needle must be rinsed with distilled water before retraction. Suction should be prohibited at this time to avoid adhesive clogging the suction channel of the endoscope. A cast will slough off from the injection site within 1 to 2 weeks, leaving an ulcer. Cyanoacrylate is very effective in controlling actively spurting variceal hemorrhage. Nonrandomized studies have shown that it is superior to other sclerosants in acute hemostasis. [54], [55] In patients with hepatocellular carcinoma complicated by variceal bleeding, a single injection of cyanoacrylate has been shown to be as effective as weekly injection of sodium tetradecyl sulfate. [56] Cyanoacrylate also offers the best results in the treatment of gastric varices. [57] Serious complications, including thromboembolism of the pulmonary, coronary, and cerebral vessels, organ infarction, [58] and multiple septic emboli, [59] have been reported.

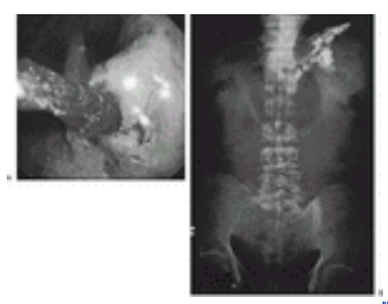

FIGURE 147-2. (See also Color Figure 147-2A.) Cyanoacrylate injection for gastric varices. **A:** The injected cyanoacrylate stops bleeding from a fundal varix instanteously. **B:** Vascular structure demonstrated on fluoroscopy confirming success of variceal obliteration.

Endoscopic Variceal Ligation

The concept of endoscopic variceal ligation (EVL) was derived from band ligation for hemorrhoids. The basic principle of variceal banding involves placement of elastic O-ring ligatures, causing strangulation of the varix. Stiegmann and Goff [60], [61] first introduced a ligation device for esophageal varices in 1986. The assembly consists of a friction-fit adaptor onto the end of the scope, an inner ligation cylinder with an elastic O-ring, and a trip wire. The varix is sucked into the inner cylinder. The trip wire is pulled to retract the inner cylinder and release the O-ring. In the treatment of actively bleeding varices, the bleeding point can be aspirated directly into the cylinder and a band applied. Varices with stigmata of hemorrhage, such as a fibrin clot, can be treated similarly. Variceal columns are banded at their most distal portions, usually just caudal to or at the esophagogastric junction. Because only a single band could be placed with the original ligation device, an overtube across the cricopharyngeus sphincter was required to facilitate multiple passages of the endoscope and its attached banding device. However, the overtube can be a cause of serious morbidity. Pinching injury of esophageal wall in the gap between the overtube and endoscope, causing esophageal laceration and perforation, has been described. The original design of the banding device also created a tunnel view and limited thorough examination. A number of modifications have since been made. Multi-fire band-ligating devices are now available, obviating the need for an overtube. The cylinder is now made of transparent plastic to improve field of vision. These multi-fire banding devices have largely replaced the single-fire banding device. They shorten the duration of each treatment session and reduce patient discomfort. [62], [63]

However, EVL still has its limitations. As varices become smaller and scarring of the esophageal wall starts to develop after a few sessions of EVL, further banding becomes difficult. In some cases, EVL alone fails to eradicate all residual varices without use of EST. The other potential problem with EVL is early recurrence of varices. Many studies have witnessed early recurrence of esophageal varices within 6 to 12 months after obliteration. [64], [65] and [66] EVL obliterates varices by pinching on the mucosa and submucosa of the esophageal wall. The periesophageal and paraesophageal varices and the perforating veins, which are the determinant factor for recurrence, are not occluded by EVL. [3], [4], [5] and [6]

Complications of Endoscopic Therapy

Both EST and EVL are associated with certain procedure-related complications (Table 147-1). Almost all patients experience some minor complications, such as chest pain, temporary dysphagia, and low-grade fever, after receiving EST. These adverse events, likely related to chemical irritation producing spasm and ulceration of the esophagus, are usually transient and self-limiting. The incidences of these complications are related to the nature, concentration, and volume of sclerosant injected. More serious complications (e.g., recurrent bleeding from esophageal ulcer, esophageal perforation, and mediastinitis) develop in up to 20% of patients after EST. Aspiration pneumonia is not uncommon in agitated patients with active bleeding. Bacteremia and bacterial peritonitis are increasingly recognized as frequent adverse events of EST. [67], [68] Severe esophageal ulcerations may heal with extensive fibrosis and stricture of esophagus requiring esophageal dilation. Rarely, portal vein thrombosis, brain abscess, spinal cord paralysis, and pericarditis have been reported. Acid-suppressing agents and sucralfate have been used to minimize the effect of acid reflux and to accelerate ulcer healing. [69], [70] Prophylactic antibiotics have been shown to reduce bacteremia and bacterial peritonitis. [26], [27], [71], [72] Judicious use of an overtube should be considered in struggling patients with active bleeding.

TABLE 147-1 Complications of Endoscopic Therapies for Variceal Hemorrhage

Compared with EST, EVL has been associated with fewer complications. This is related to the fact that EVL produces only mechanical strangulation of varices without much tissue inflammation and injury. Small ulcers are produced after the O-ring and the strangulated varix slough off from the esophageal wall. The ulcers are usually superficial and almost never produce stricture of esophagus. Complications associated with chemical irritation, such as fever, mediastinitis, and pleural effusion, are rare. The only serious complication associated with EVL is related to the use of an overtube: esophageal laceration, perforation, and pinching injury of the esophagus. As multiple banding devices are replacing single-band ligators, serious esophageal injury inflicted by overtube will become of historical interest.

Detachable Mini-Snare

The detachable snare employs the same concept as the original banding device. It was initially designed for endoscopic resection of elevated mucosal lesions to minimize the risk for bleeding. [73] The snare has a maximum diameter of 4 cm when fully opened. Yoshida and colleagues [74] used the detachable snare for the ligation of gastric varices and reported excellent results. Despite that, detachable snare ligation of gastric varices has not been very popular. Gastric varices are usually quite large, and positioning of the snare around the gastric varices is often difficult. With modification of the detachable mini-snare, it has recently been used for the treatment of esophageal varices. [75] This method of variceal ligation has the advantages of obviating the use of overtube, producing a clear endoscopic image, and offering an unlimited number of ligations. A transparent ligation chamber with a rim on the inside is fitted onto the tip of an endoscope. The detachable mini-snare, which is an oval-shaped nylon ring with a maximum diameter of 11 mm, is inserted into the working channel of the endoscope until it opens at the rim of the ligation chamber. The endoscope is positioned to obtain an en face view of a variceal column. The varix is then sucked into the ligation chamber, and the mini-snare is maneuvered to surround the varix. Ligation of the varices is performed by sliding the stopper forward. When the loop is tightened around the base of the varix, suction can be released and the mini-snare detached from the snare. The ligated portion of the varix appears as a “polyp” with a short string attached to the base (Fig.

147-3). Additional ligations are carried out by reloading the mini-snare with the endoscope remaining in the esophagus. A recent prospective randomized study showed that mini-snares performed comparably to a multi-fire variceal ligator. [76]

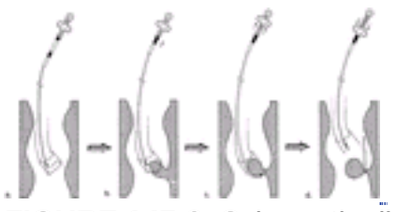

FIGURE 147-3. Schematic diagram on the mechanism of deployment of detachable mini-snare for the ligation of esophageal varices. **A:** An endoscope inside the esophagus loaded with the detachable mini-loop. **B:** An esophageal varix is sucked into the ligation chamber and the position of the mini-loop adjusted to surround the varix. **C:** The mini-loop is tightened by sliding the stopper forward. **D:** The mini-loop is detached from the snare when the loop is tightened. (From Sung JY, Chung SC. The use of a detectable mini-loop for the treatment of esophageal varices. Gastrointest Endosc 1998;47:178.)

Thrombin, Hemoclips, and Argon Plasma Coagulation

Both human and bovine thrombins have been used in the control of variceal hemorrhage. The injection of bovine thrombin (single intravariceal injection of 1000 U/mL) resulted in cessation of bleeding in 11 patients in a single series. [77] It has been proposed that thrombin is equally effective in controlling esophageal as well as gastric varices. The major advantage of using thrombin as a thrombotic agent to obliterate varices is that it appears to produce few complications in the absence of mucosal inflammation. Allergic reaction and infection transmission from bovine thrombin has always been a concern. Data from randomized study to confirm its efficacy is, so far, lacking.

Poly *N*-acetyl glucosamine, derived from marine microalgae, has been tested in the treatment of varices. [78] In a canine model study, it has been shown that intravariceal injection of 2.5% to 3.5% poly *N*-acetyl glucosamine has the ability to eradicate esophageal varices and achieve complete hemostasis in a single session. Further studies will be required before this gel can be considered in clinical practice.

Hemoclips were originally designed to control arterial bleeding at the base of peptic ulcers. In a Japanese study, hemoclips were used in the treatment of esophageal varices in 12 children. [79] Multiple hemoclips were applied from the lesser curve of the stomach to the lower esophageal. Although the authors claimed a 100% success rate, there is no further report to confirm efficacy.

Recently, argon plasma coagulation has been used to induce superficial burns and fibrosis on the esophageal mucosa after eradication of varices. [80] A circumferential burn at the lower esophagus is made to induce fibrosis in the submucosa to prevent recurrence of varices. Although the procedure appears safe in patients after all varices are obliterated, the long-term effect of argon plasma coagulation remains to be confirmed.

ENDOSCOPIC TREATMENT OF ACUTE VARICEAL HEMORRHAGE

In patients with acute variceal bleeding, the goals of treatment are to restore hemodynamic stability, stop hemorrhage, prevent complications, including early rebleeding, and decrease mortality. Many modalities of treating variceal bleeding have been devised, including pharmacological therapy, endoscopic treatment, surgical devascularization or bypass, and radiologic shunting.

Endoscopic Injection Sclerotherapy versus Sham or Balloon Tamponade

Although EST has been used as the standard treatment for decades, few data compare this treatment with sham to support its efficacy. The best evidence for the value of EST comes only recently from the Veterans Affairs Cooperative Variceal Sclerotherapy Group. [81] In this study, EST stopped hemorrhage from actively bleeding esophageal varices more effectively than sham EST with medical therapy (91% versus 60%) and significantly improved survival (75% versus 51%). EST was compared with medical treatment with or without balloon tamponade in four trials. Barsoum [82] randomized 100 patients and found a favorable effect of EST superior to balloon tamponade in reducing both in-hospital rebleeding and death. In the Copenhagen Esophageal Varices Sclerotherapy Project, 187 patients were randomized, and the authors reported no significant difference in the initial control of hemorrhage. [83] Rebleeding within the first 40 days was similar in either group. After 40 days, a significant reduction in rebleeding associated with EST was noted (15% versus 31%); thus, the beneficial effect of EST was seen in the long-term after eradication of varices. Paquet and Feussner [84] and Moreto and associates [85] recruited smaller numbers of patients and reported improvement in control of bleeding with EST. In summary, EST is superior to balloon tamponade in the acute control of bleeding and in the prevention of recurrent hemorrhage from esophageal varices.

Endoscopic Injection Sclerotherapy versus Vasoactive Agents

Two major classes of drugs used in the control of acute variceal hemorrhage are vasopressin and its analogs and somatostatin and its analogs. EST was compared with vasopressin in five studies, terlipressin in one study, somatostatin in four studies, and octreotide in three studies (Table 147-2). Soderlund and Ihre [86] randomized 107 unselected patients to receive either EST or vasopressin infusion supplemented by balloon tamponade. Initial control and hospital mortality were similar in both groups. With long-term follow-up, EST led to variceal eradication and less rebleeding. Larson and colleagues [87] randomized 82 patients to EST or conventional treatment, including balloon tamponade and vasopressin, and demonstrated that EST was clearly beneficial in reducing rebleeding episodes. Westaby and associates [88] compared EST with vasopressin plus nitroglycerin and reported a better control of active bleeding and fewer deaths due to variceal bleeding in the EST group. El-Zayadi and coauthors [89] and Alexandrino and colleagues [90] confirmed comparable efficacy of vasopressin and EST. The use of vasopressin is hampered by its frequent side effects due to nonselective vasoconstriction. The risk is highest among patients with cardiac ischemia and peripheral vascular diseases.

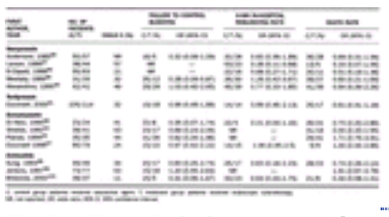

TABLE 147-2 Comparative Trials of Sclerotherapy with Vasoactive Agents in the Acute Control of Variceal Hemorrhage

In a single comparison study between terlipressin and EST, both treatments were found to have similar efficacy. [91] Side effects developed in 20% of patients receiving terlipressin and 30% of patients receiving EST, which gives the vasoactive agent a marginal superiority in safety profile over endoscopic treatment.

Unlike vasopressin, somatostatin and its analogs have selective vasoconstrictive activity in the splanchnic circulation, [92], [93] with a lower risk of systemic side effects. Four trials compared somatostatin [94], [95], [96] and [97] and three compared octreotide against endoscopic sclerotherapy in the treatment of acute bleeding esophageal varices. [98], [99] and [100] All studies showed similar rates of initial control of bleeding, recurrent bleeding, and death. In a recent metaanalysis combining data collected from 13 randomized trials, octreotide has been shown to improve control of esophageal variceal hemorrhage compared with all alternative therapies combined. Its

effect is comparable to immediate EST with fewer major complications than vasopressin or terlipressin. [101]

Endoscopic Injection Sclerotherapy versus Endoscopic Variceal Ligation

Since its development by Stiegmann and Goff in the late 1980s, more than 10 studies have compared EVL with EST. [64], [65] and [66], [102], [103], [104], [105], [106], [107], [108] and [109] (Table 147-3). Most studies reported that EVL is a safer and quicker technique with fewer episodes of rebleeding and complications. The early metaanalysis by Laine and Cook [110] demonstrated reduction in rebleeding rate (odds ratio [OR], 0.52; 95% confidence interval [95% CI], 0.37 to 0.74), overall mortality (OR, 0.67; 95% CI, 0.46 to 0.98) and mortality caused by rebleeding (OR, 0.49; 95% CI, 0.24 to 0.996) after EVL as compared with EST. The number of EVL sessions required to achieve variceal obliteration was also lower. The frequency of esophageal strictures was less with ligation. Following this metaanalysis, at least six more studies were published demonstrating the superiority of EVL over EST. Although attractive because of a faster eradication rate and fewer complications, EVL is associated with a higher variceal recurrence. The difference in variceal recurrence can be explained by the fact that the ulcers caused by banding are usually superficial and induce very limited scarring. Because the high portal pressure is not affected by endoscopic treatment and the perforating veins are not obliterated by EVL, the residual esophageal varices dilate over a period of weeks or months. In almost all studies comparing the long-term effects of EST and EVL, recurrence of esophageal varices was found more frequently in the EVL-treated patients (30% to 48%) compared with the EST-treated patients (8% to 30%). On the contrary, rebleeding from esophageal varices was significantly higher among those treated by EST than by EVL (37% versus 8%). The overall survival rates are similar in both treatment modalities and are determined by the hepatic function reserve rather than by the method of endoscopic hemostasis. A comparison between EST and EVL is summarized in Table 147-4. With the exception of the study by Gimson and colleagues, [103] EST produced a higher complication rate than EVL. In summary, EVL is at least as effective as EST in the control of acute variceal hemorrhage and has the clear advantage of producing fewer complications. Although EVL treatment is associated with more variceal recurrence, rebleeding from varices is paradoxically less common.

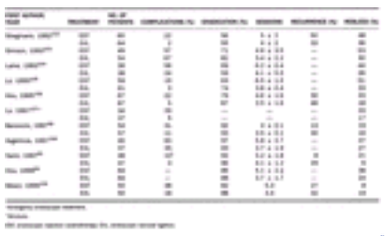

TABLE 147-3 Randomized Comparative Trials of Injection Sclerotherapy and Variceal Ligation

	SCLEROTHERAPY	VARICEAL LIGATION
Technique	Variable	Standardized
Control of active bleeding	>90%	>90%
Sessions required for obliteration	More	Less
Complications	Common	Uncommon
Recurrence of varices	Less common	Common
Effects on survival	No effect	May improve survival
Use in primary prophylaxis	Not recommended	Comparable to beta-blocker
Cost	Inexpensive	Expensive

TABLE 147-4 Comparison between Endoscopic Sclerotherapy and Endoscopic Variceal Ligation

Endoscopic Therapy plus Vasoactive Agents versus Endoscopic Therapy Alone

Because both endoscopic therapy and vasoactive drugs are effective in the acute control or prevention of recurrent bleeding in esophageal varices, it would be logical to combine modalities. Seven studies combined EST with vasoactive agents; somatostatin in two studies, [111], [112] octreotide in three studies, [113], [114] and [115] terlipressin in one study, [116] and vapreotide in one study. [117] In most studies, combination therapy of EST and vasoactive agents was shown to be more effective than EST alone.

Optimally, vasoactive drugs are given as early as possible in patients with suspected esophageal variceal bleeding. Terlipressin plus nitroglycerin started by the emergency medical team at the patient's home before arrival at the hospital significantly improved control of variceal hemorrhage. [116] Avgerinos and colleagues [111] (the ABOVE trial) and Cales and associates [117] commenced somatostatin and vapreotide, respectively, soon after hospitalization before endoscopy. In the ABOVE trial, active bleeding was seen less frequently in patients assigned to somatostatin. [111] EST was technically easier after bleeding was temporarily controlled with pharmacological therapy. Endoscopic treatment failure was less likely in the somatostatin group. Similar findings were noted in the study using vapreotide. As in the ABOVE Study, fewer patients had active bleeding during endoscopy if they had received prior vapreotide. [117] The lowering of portal venous pressure by early administration of vasoactive drugs facilitates subsequent endoscopic control of hemorrhage. In all three studies, there was significant improvement in the initial control of bleeding with combined pharmacological and endoscopic therapy. Mortality due to bleeding in the first 2 weeks was reduced with terlipressin given before endoscopic treatment. Despite the popularity of EVL, there are few studies in the literature combining EVL and vasoactive agents. Sung and coauthors [118] found that EVL plus octreotide led to significantly less rebleeding (9% versus 38%), as well as less balloon tamponade and endoscopic retreatment, than EVL alone. No effect on mortality was seen. Combination of somatostatin or its analog with either EST or EVL appears to offer the best treatment for control of acute hemorrhage and prevention of early rebleeding.

Combined Endoscopic Therapy versus Endoscopic Injection Sclerotherapy or Endoscopic Variceal Ligation Alone

EVL eradicates varices faster than EST and is associated with fewer side effects. EVL, however, has its disadvantages. After a few sessions of EVL, ligation of small residual varices becomes difficult owing to underlying fibrosis. Early recurrence of varices is often seen with EVL owing to persistence of perforating veins and paraesophageal collaterals. EVL has been combined with EST in an effort to include merits of both modalities and minimize complications. There are two approaches: a synchronous approach using both EVL and EST in the same session, or a metachronous approach using EVL to ligate varices initially, followed by EST for the small residual varices. The methodology for synchronous treatment varies: some endoscopists inject into a varix ligated at both its proximal and distal ends (sandwich technique), whereas others inject into the adjacent mucosa or ligate an injected varix. The following section summarizes randomized studies comparing combined EVL and EST to either EVL or EST alone.

Synchronous Approach Seven randomized trials compared combined therapy administered in the same session against monotherapy (five against EVL and two against EST). [119], [120], [121], [122], [123], [124] and [125] In an earlier trial, Laine and coauthors [119] observed a higher complication rate with the use of synchronous EVL and EST when compared with EVL alone (29% versus 10%), and more sessions of combined treatment were required to eradicate varices. There was little difference in the rates of eradication, mortality, and rebleeding. Saeed and associates [120] reported similar findings but observed a higher mortality rate with combined treatment. Umehara and colleagues [121] observed a significantly higher recurrence rate with EVL alone at 3 years when compared with combined EVL and EST (72% versus 21%) without a significant difference in rebleeding. On the contrary, Al Traif and colleagues [122] reported that EVL alone produces fewer recurrences of varices than combined therapy. Among the five trials comparing synchronous EVL and EST to EVL alone, all except that of Al Traif and colleagues reported a higher rate of complications associated with combined treatment (29% to 68% versus 10% to 38%). Synchronous EVL and EST took longer to eradicate varices, with equivocal benefit in reducing long-term recurrence and associated rebleeding. In the two trials comparing synchronous EVL and EST to EST alone, the rate of complications associated with EST alone was substantial (as high as 91% in one trial), suggesting that combined EVL and EST improved safety by limiting the volume of injected sclerosant. [124], [125]

Metachronous Approach Five randomized studies compared sequential EVL and EST to either EVL or EST alone. [126], [127], [128], [129] and [130] Bhargava and Pokharna [126] reported higher eradication (87% versus 24%) and lower rebleeding (19% versus 22%) with combination therapy, although the number of sessions required and the overall complication rate were higher when compared to EVL alone. Lo and colleagues [127] reported similar treatment sessions and eradication rate with EVL and EST versus EVL. The mortality rates (2.7% versus 8.6%), rebleeding rates (8% versus 31%), and variceal recurrence rates (14% versus 43%) were, however, lower with

combined treatment. In a similar study from Taiwan, Cheng and colleagues [128] noted that recurrence of varices was more common in patients receiving EVL alone. Garg and associates [129] found a higher complication rate with EST alone than with EVL and EST. Masumoto and co-workers [130] assigned patients to sequential EVL and EST (n = 21), EVL alone (n = 20), or EST alone (n = 18). Recurrence of varices was highest among those who received EVL alone (40%), and the use of EST alone was again associated with a high rate of complications (50%). In summary, synchronous EVL and EST cannot be recommended when compared with EVL alone. There may, however, be a role in the sequential use of EVL to reduce the size of varices followed by low-dose sclerotherapy for small residual varices in reducing long-term variceal recurrence and possibly rebleeding.

Endoscopic Therapy versus Surgery

Surgical treatments for variceal bleeding include direct esophageal devascularization of the lower esophagus plus the proximal stomach and a variety of surgical shunts. Simple surgical devascularization (i.e., surgical ligation of varices) with esophageal transection is an effective means of controlling acute bleeding, but long-term recurrence of varices and bleeding is common. Shunt operation can be classified as total, partial, or selective. Total shunts (side-to-side or end-to-side) divert all portal blood flow into the inferior vena cava, causing effective control in most cases of variceal bleeding but also severe hepatic encephalopathy in a significant proportion of patients. To reduce the incidence of this fatal complication of nonselected shunt, Sarfeh and associates [131] described an interposition side-to-side H-graft between the portal vein and the cava using an 8-mm graft. By maintaining partial hepatic perfusion, the incidence of encephalopathy was reduced. Its role in the prevention of recurrent esophageal bleeding has not been fully evaluated. Selective shunting refers to a shunt that selectively decompresses variceal flow while preserving portal blood flow. Distal splenorenal shunt (DSRS) anastomoses the splenic vein to the distal left renal vein, allowing the short gastric veins to drain into the systemic circulation. [132] It was designed as a selective shunt to avoid the high rate of encephalopathy seen with total shunts. However, with time, this shunt tends to reconnect with the portal vein through pancreatic branches of the splenic vein. They progressively enlarge and serve as outflow collateral for the portomesenteric system. DSRS then loses its selective quality and becomes a partial or even a total shunt. [133] A total disconnection of splenic vein from pancreas and division of splenocolic ligament (splenopancreatic disconnection) has been advocated as an adjunct at initial surgery. Because of differences in study designs, trials comparing shunt surgery to endoscopic therapy (mostly EST) must be analyzed separately in terms of short-term effects and long-term results.

In emergency control of variceal hemorrhage, EST has been compared with esophageal transections in four trials. [134], [135], [136] and [137] (Table 147-5). Except for the subgroup analysis from the study by Teres and colleagues, [136] all studies showed that surgery produced less rebleeding. However, high-risk patients treated by either EST or surgery had the same mortality rate of close to 50%. Rebleeding after esophageal transection and devascularization occurs in 31% in 4 years. It has been concluded that esophageal transection confers no benefit in long-term management of esophageal varices. Triger and associates [138] compared esophageal transection to EST in the long-term management of esophageal varices after initial stabilization with EST. Mortality and recurrence of varices with esophageal transection were no better than EST, indicating no long-term benefit of surgery over endoscopic therapy.

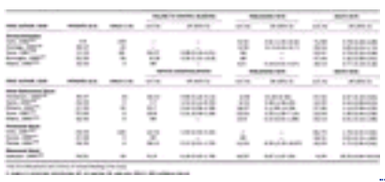

TABLE 147-5 Comparative Trials of Surgery and Endoscopic Sclerotherapy in the Control of Variceal Hemorrhage

For subsequent prevention of recurrent bleeding after the acute episode, long-term EST has been compared with DSRS in five studies. [139], [140], [141], [142] and [143] In a metaanalysis of four of these trials, the pooled relative risk for rebleeding was significantly reduced by DSRS (0.16; 95% CI, 0.10 to 0.27). [144] The overall risk for death following DSRS was not significantly decreased (0.78; 95% CI, 0.47-1.29); the relative risk for chronic encephalopathy was 1.86 (95% CI, 0.90 to 0.86). [144] Nonselective portacaval shunt was evaluated against EST in three trials [145], [146] and [147] and mesocaval shunt in one trial. [148] There is considerable heterogeneity between these studies, making comparisons difficult. In the overall group, shunt surgery was associated with significantly lower rebleeding rate. The incidence of hepatic encephalopathy after shunt surgery was significantly higher than that of sclerotherapy. Mortality rates were comparable.

Endoscopic Therapy versus Transjugular Intrahepatic Portosystemic Shunt

Transjugular intrahepatic portosystemic shunt (TIPSS) is a radiologic intrahepatic portacaval shunt. It has been compared with endoscopic therapy as a modality in the secondary prophylaxis of variceal bleeding in 11 trials involving 811 patients (Table 147-6). Endoscopic treatment in these trials are: sclerotherapy alone in five studies, [149], [150], [151], [152] and [153] EVL alone in three studies, [154], [155] and [156] and EST or EVL plus propranolol in three studies. [157], [158] and [159] In a metaanalysis pooling data from the above studies, variceal rebleeding was significantly more frequent with endoscopic therapy (47%) than with TIPSS (19%) (OR, 3.8; 95% CI, 2.8 to 5.2). [160] Despite high success in controlling hemorrhage, TIPSS has failed to demonstrate improvement in mortality (OR, 0.97; 95% CI, 0.71 to 1.34). One of the major problems with TIPSS is, as in all portosystemic shunts, the development of encephalopathy. Hepatic encephalopathy was much more frequent after TIPSS (34%) than after endoscopic therapy (19%) (OR, 0.43; 95% CI, 0.30 to 0.60). Currently, TIPSS cannot be recommended as a routine form of secondary prophylaxis. On the other hand, TIPSS is highly efficacious as a rescue procedure after failed endoscopic therapy. Therefore, TIPSS should be reserved for the subset of patients who continue to bleed or develop recurrent bleeding after endoscopic therapy. [161], [162] and [163]

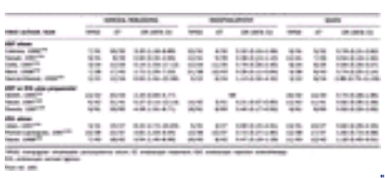

TABLE 147-6 Comparative Trials of TIPSS with Endoscopic Therapy in the Secondary Prophylaxis of Variceal Hemorrhage

Summary on Treatment of Acute Variceal Hemorrhage

Based on existing evidence, a clinical algorithm is presented in Figure 147-4. During initial resuscitation with fluid and blood replacement, cirrhotic patients with upper gastrointestinal bleeding should be given intravenous injection or infusion of a vasoactive agent: terlipressin, somatostatin, or octreotide. Emergency endoscopy should be done if the patient is hemodynamically unstable. After endoscopic confirmation of the source of bleeding, banding ligation, injection sclerotherapy, or both should be offered immediately. Vasoactive agents should be continued for 5 days to prevent early rebleeding, and prophylactic antibiotics against infectious complications should be given. When initial hemostasis is achieved, the patient can be scheduled for an endoscopic obliteration program with weekly or biweekly banding or sclerotherapy until all varices are obliterated. Patients who fail to respond to endoscopic therapy or suffer from recurrent bleeding during the acute phase should be offered balloon tamponade before a second session of therapeutic endoscopy. Vasoactive therapy should be continued. In patients who continue to bleed from their varices despite nonoperative means, the choice between TIPSS or surgical shunts is less well defined. Rosemurgy and associates [164] randomly assigned such patients to receive either TIPSS or surgical shunts using a small-diameter prosthetic H-graft (35 in each group). The use of TIPSS was associated with more frequent shunt failure and total occlusions, resulting in more rebleeding and death. In low-risk patients with compensated liver function unlikely to require liver replacement in due course, surgical shunts in the form of selective shunt (e.g., DSRS) should probably be offered. TIPSS is reserved for patients with limited hepatic reserve as a bridging procedure while awaiting liver transplantation.

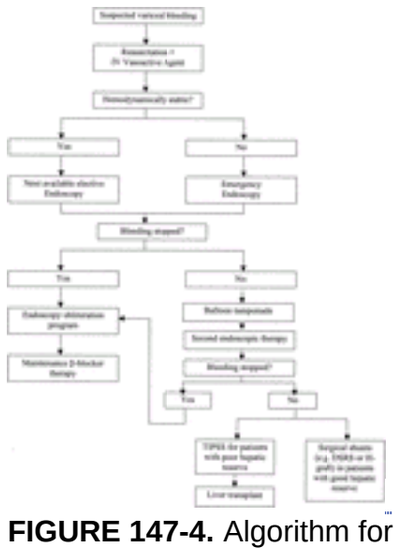

FIGURE 147-4. Algorithm for management of acute bleeding from esophageal varices.

PROPHYLACTIC TREATMENT OF ESOPHAGEAL VARICES

Beta-blockers reduce the risk for the first variceal hemorrhage, especially in patients with large esophageal varices. The protective effects of beta-blockers may be enhanced when co-prescribed with nitrates. However, only one third of patients respond to beta-blockers, and they are not tolerated by all patients. Controversy exists regarding the role of endoscopic treatment as primary prophylaxis.

Endoscopic Injection Sclerotherapy as Primary Prophylaxis to Prevent Variceal Hemorrhage

Prophylactic EST was used in the 1980s to prevent variceal bleeding. Discrepant results were found for the bleeding and mortality rates. Three of the four initial controlled trials reported that prophylactic EST significantly reduces the risk for first variceal bleeding and improved survival. [165], [166] and [167] However, in subsequent trials, prophylactic EST did not show a survival benefit. Sclerotherapy may even provoke bleeding that is difficult to control. Indeed, in two American studies, the mortality rate was higher in the EST group than in the control group. [168], [169] However, these studies were heavily criticized for patient selection. A metaanalysis revealed marked heterogeneity among these trials and showed that EST significantly reduced the risk for a first episode of bleeding, whereas the mortality rate was higher in those who received EST in certain studies. [170]

Endoscopic Variceal Ligation as Primary Prophylaxis to Prevent Variceal Hemorrhage

Although EST may produce more harm than good in patients with cirrhosis, EVL is associated with fewer procedure-related complications. Three trials [171], [172] and [173] compared EVL to no treatment in the prevention of the first bleeding, and two compared EVL to propranolol therapy [174], [175] (Table 147-7). These studies showed a significant decrease in the risk for bleeding, and the mortality rate was also significantly reduced in two studies. Sarin and colleagues [175] showed reduced bleeding with EVL as compared with propranolol (15% versus 43%). [175] The overall mortality rate related to variceal bleeding, however, was identical. It has been pointed out that the rebleeding rate of the propranolol-treated group was unusually high. [176] Beta-blockers should remain the first choice for prophylaxis, with EVL reserved for patients with contraindications or intolerance to these drugs. In a recently published metaanalysis based on four trials and 283 patients, EVL reduced the risk for variceal bleeding by 64% (relative risk [RR], 0.36; 95% CI, 0.26 to 0.50) and mortality by 45% (RR, 0.55; 95% CI, 0.43 to 0.71). [177] When compared with beta-blocker, EVL reduced the risk for variceal bleeding by 52% (RR, 0.48; 95% CI, 0.24 to 0.96) but failed to reduce mortality (RR, 0.95; 95% CI, 0.56 to 1.62).

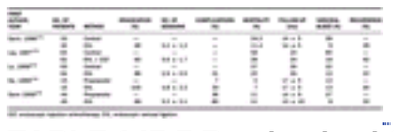

TABLE 147-7 Randomized Comparative Trials of Band Ligation or a Combination of Band Ligation and Injection Sclerotherapy for the Primary Prophylaxis of Variceal Bleeding

Summary on Primary Prophylaxis of Variceal Bleeding

All patients with a confirmed diagnosis of portal hypertension with or without cirrhosis should receive screening for varices by upper endoscopy. Based on natural history studies, [11], [12] patients with no varices need to have follow-up endoscopy every 3 to 4 years. Patients with esophageal varices should be assessed by a predictive scoring system, such as the NIEC score. Low-risk patients should be reexamined yearly for progression of esophageal varices. Moderate- to high-risk patients (e.g., NIEC score higher than 30) should be offered prophylactic therapy. Beta-blockers are recommended as the first-line prophylaxis. Patients with contraindications or intolerance to beta-blockers can be offered prophylactic banding ligation (Fig. 147-5).

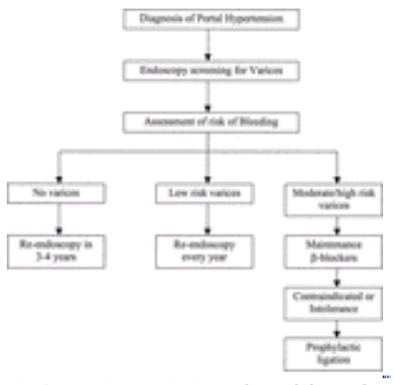

FIGURE 147-5. Algorithm for primary prophylaxis of variceal bleeding.

GASTRIC VARICES

Compared with esophageal varices, the natural history and optimal treatment of gastric varices are much less well defined. Gastric varices can occur in patients with portal hypertension or occlusion of splenic vein (primary), or after endoscopic treatment of esophageal varices, which produces back pressure on vessels in the gastric zone (secondary). Sarin and colleagues [178] prospectively studied 586 patients with portal hypertension. At first endoscopy, gastric varices were found in 20% of patients, and they developed in 9% of patients within 2 years after eradication of esophageal varices. Gastric varices tend to bleed less frequently than esophageal varices, but bleeding tends to be more severe. Mortality in those who bleed from gastric varices is substantial.

Gastric varices can be categorized into two groups according to their locations in the stomach and relationship with esophageal varices: gastroesophageal varices (GOV), found associated with esophageal varices; and isolated gastric varices (IGV), found independent of esophageal varices. Each of the subtypes can be further classified into type 1 and 2 based on their locations in the stomach (Fig. 147-6). Type 1 GOV are continuous, with esophageal varices extending 2 to 5 cm beyond the gastroesophageal junction along the lesser curvature of the stomach, whereas type 2 GOV extend into the fundus of the stomach. Type 1 IGV are isolated fundal gastric varices, and type 2 IGV are isolated "ectopic" varices occurring in the distal portion of stomach, including the duodenum.

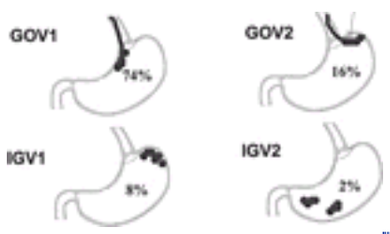

FIGURE 147-6. Classification of gastric varices and their relative frequencies. (Adapted from ref. [178].)

Sarin's classification of gastric varices has prognostic implications and may be helpful in management planning. [178] Type 1 GOV are the commonest type, accounting for 75% of cases. Their rate of bleeding was only 11.8%. Their treatment is identical to that of esophageal varices, and they are usually responsive to endoscopic therapy. Type 2 GOV tend to bleed more frequently (55%). Type 1 IGV have the highest rate of bleeding at 78% and carry substantial mortality. Sclerotherapy has been described in the treatment of bleeding gastric varices as an overall group, with initial control in about 80% of patients. [179] The reported rate of initial control of bleeding in type 1 IGV, however, was much lower. Active bleeding was arrested in only 26% of patients with IGV in one reported series. [180] In addition, rebleeding after endoscopic sclerotherapy for bleeding gastric varices is common, especially in type 1 IGV. EVL is less effective in bleeding gastric varices. Deployment of bands can be difficult with the endoscope in a retroflexed position. Band retention rate is low, leading to frequent early rebleeding.

Histoacryl injection may produce better results. [181] In a nonrandomized series, Oho and colleagues [54] compared injections using cyanoacrylate to ethanolamine in 53 patients. [54] Their study showed that the rate of initial control of bleeding is significantly higher in patients treated with cyanoacrylate (93% versus 67%). The in-hospital mortality rate was significantly higher in the ethanolamine-treated group (67% versus 38%). [54] In a recent randomized study comparing EVL with cyanoacrylate injection in 60 patients, cyanoacrylate injection had better initial hemostasis, less recurrent bleeding, fewer transfusions, and lower mortality. [182]

In general, the low rate of hemostasis and high rate of recurrent bleeding in IGV with endoscopic treatment would imply that alternate therapies such as surgery or TIPSS are sometimes indicated. The management of gastric varices requires further study.

ENDOSCOPIC ULTRASONOGRAPHY IN VARICEAL HEMORRHAGE

Endoscopic ultrasonography (EUS) has been applied in patients with variceal hemorrhage since 1990. [183] Using EUS, a large part of the portal venous system can be visualized, including the portal vein, splenic vein, superior mesenteric vein, and azygos vein. However, when used in detecting esophageal varices, EUS was found to be inferior to conventional upper endoscopy and gave little additional information. [184], [185] This was mainly due to the large-bore echoendoscope of the first generation, and the use of a water-filled balloon, which compresses variceal columns in the esophageal lumen. With improvement in technology, and the development of catheter probes using higher ultrasound frequency, [186], [187] EUS can detect vascular channels both inside and outside the esophageal lumen. EUS can map vessels in the connective tissue layer just outside the esophageal wall (periesophageal varices), collateral veins in the mediastinum running longitudinally alone with the esophagus (paraesophageal varices), and the perforators that connect the submucosal and peripheral vascular channels [188], [189] (Fig. 147-7). Paraesophageal varices and perforating veins are reported in about 80% of patients with esophageal varices, and their presence has been associated with poor response to endoscopic therapy and early recurrence of varices. [3], [4] and [5], [190], [191], [192] and [193] It has been suggested that patients undergoing EVL are more likely to have multiple paraesophageal varices of considerable size compared with those who received EST. [192] EUS-guided sclerotherapy using color Doppler has been attempted to improve results of EST. [194] It has also been suggested that after endoscopic therapy has achieved obliteration of submucosal varices, EUS examination of the lower esophagus is warranted to identify those who have perforating veins and periesophageal or paraesophageal varices. Small-volume injections of sclerosants may be given to seal these perforators to prevent early recurrence of varices and rebleeding. The development of color Doppler EUS has also permitted study of blood flow hemodynamics in the portal venous system. Cirrhotic patients have been found to have enlarged azygos veins and thoracic ducts. [195] Azygos vein blood flow has been found to be reduced after intravenous infusion of vasoactive agents [195] and after endoscopic therapy. [196] Recently, the measurement of azygos vein blood flow using EUS has been validated by thermodilution method. [197] The use of azygos blood flow measurement in the clinical management of variceal hemorrhage remains to be defined.

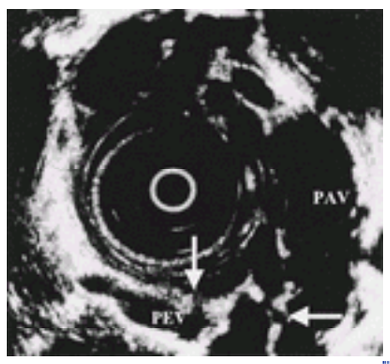

FIGURE 147-7. Periesophageal varices (PEV) and paraesophageal varices (PAV) together with perforators (*arrow*) as seen with endoscopic ultrasonography.

Unlike esophageal varices, EUS has been found to be very useful in detecting gastric varices because it can easily distinguish vascular structure from other submucosal lesions in the gastric fundus. [183], [184] EUS also is a useful tool in monitoring the effects of injection sclerotherapy because it confirms obliteration of vascular channels in the submucosa of the stomach. Cyanoacrylate injection monitored by EUS is reported to reduce recurrence of gastric varices. [198]

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CHAPTER 148

David J. Bjorkman

ENDOSCOPIC DIAGNOSIS AND TREATMENT OF NONVARICEAL UPPER GASTROINTESTINAL HEMORRHAGE

Upper gastrointestinal (GI) bleeding is a common clinical disorder. It is responsible for more than 250,000 to 300,000 hospital admissions and $2.5 billion in costs in the United States each year. [1] The most common cause of nonvariceal upper GI bleeding is peptic ulcer disease, but other disorders may have an identical presentation. [1] GI endoscopy remains both the diagnostic and therapeutic procedure of choice for upper GI bleeding. Despite progressive advances in the diagnosis, the mortality rate from acute upper GI bleeding remains 5% to 15%. [2], [3] Recent data demonstrate an age-related mortality rate that ranges from 4% in young patients to 9% in elderly patients. [4] It is possible that part of the failure to see a significant reduction in overall mortality is that patients presenting with upper GI bleeding today are often older and have significant co-morbid illnesses that affects the outcome of the bleeding episode. [2]

INITIAL EVALUATION AND PREPARATION FOR ENDOSCOPY

The initial evaluation of patients with acute GI bleeding should focus on the hemodynamic status. Hemoglobin concentration and hematocrit are unreliable markers of acute blood loss. Intravenous access and vigorous volume replacement should be immediately initiated in all patients with hemodynamically significant GI bleeding. [5] This requires large-bore vascular access, preferably in at least two sites, or through a central line. Immediate infusion of isotonic electrolyte solutions should be given until vital signs become stable. The use of blood products should be determined by the patient's clinical condition and the rapidity of blood loss. [6] Other diagnostic and treatment decisions should not take precedence over volume resuscitation.

ASSESSING THE LOCATION OF BLEEDING

Endoscopy is the optimal method for establishing the location of bleeding. A history of hematemesis with frank blood or coffee ground–like material is sufficient to confirm the level of bleeding to be above the ligament of Treitz. A prior history of bleeding ulcers, nonsteroidal antiinflammatory drug use, or variceal bleeding is suggestive but is not sufficient to establish the presence of an upper GI bleeding source. A history of melena is also suggestive of upper GI bleeding, but melena can occur with slow bleeding from the small bowel or, rarely, the proximal colon. Hematochezia is more suggestive of a bleeding site in the lower GI tract but may also result from vigorous upper GI bleeding in 10% of cases.

Nasogastric aspiration is 80% sensitive for an actively bleeding upper GI source when it produces fresh or altered blood. False-negative aspirates occur when the tube is improperly positioned or when the reflux of blood from a duodenal source is prevented by pylorospasm or obstruction. [7] When vigorous bleeding is identified, gastric lavage using a large-bore orogastric tube is often needed to clear the stomach of blood and clots before endoscopic evaluation and treatment. [8]

ROLE OF ENDOSCOPY IN DIAGNOSIS

After the patient has become hemodynamically stable, endoscopy is the diagnostic procedure of choice in the setting of upper GI bleeding. The contraindications to endoscopy are listed in Table 148-1. Absolute contraindications include refusal of the procedure by the patient, a situation in which the risks of the procedure outweigh the potential benefits of the procedure (such as shock), and a situation in which the procedure will not alter the outcome or care of the patient. [9] The relative contraindications can often be overcome by appropriate patient preparation. For example, airway protection may be achieved by endotracheal intubation before endoscopy, and coagulation disorders may be mitigated by the infusion of blood products.

Absolute contraindications
Refusal of informed consent
The risks of the procedure outweigh the potential benefits
Endoscopic information or therapy will not alter the outcome or care
Relative contraindications
Suspected gastroduodenal perforation
Hemodynamic instability
Combative, uncooperative patient
Severe coagulopathy
Unprotected airway

TABLE 148-1 Contraindications to Upper Gastrointestinal Endoscopy

Endoscopy has a sensitivity of 92% when the entire stomach and proximal duodenum can be carefully visualized and has a specificity that approaches 100%. [10] By comparison, barium radiography has a sensitivity of only 54%. Visualization of the bleeding lesion may be aided by local application of 3% hydrogen peroxide through the endoscope. [11], [12] The ability to perform biopsies to diagnose malignancy and the presence of *Helicobacter pylori* infection is not available with barium studies. Most importantly, barium radiography interferes with subsequent endoscopy, angiography, or surgery. As a result, barium radiography should be avoided in the setting of acute upper GI bleeding.

Not only will a careful endoscopic examination identify the source of upper GI bleeding, but also it can provide predictive information about the prognosis (probability of rebleeding, morbidity, and mortality) for the patient. Clinical prognostic factors include older age, shock, volume of bleeding (as determined by hemodynamic parameters and the volume of fluids needed to restore blood pressure and pulse to normal), need for transfusion, onset of bleeding in the hospital, and co-morbid conditions. [13], [14] Since the advent of endoscopy, however, it has been demonstrated that the endoscopic appearance of bleeding ulcers more accurately predicts the risk for rebleeding-associated morbidity and mortality. Shortly after the availability of endoscopy, Forrest and colleagues [15] recognized specific endoscopic findings that were associated with an increased risk for rebleeding. The endoscopic findings that correlate with the risk for rebleeding (in order of risk for rebleeding and increased mortality) are ulcers with a clean base; the presence of flat, pigmented, spots in the base; an adherent clot resistant to vigorous flushing; a nonbleeding visible vessel; and active bleeding (either oozing or vigorous bleeding) (Fig. 148-1, Color Fig. 148-1). This classification has been repeatedly shown to predict accurately the rate of rebleeding, morbidity, and mortality in multiple studies. [1], [16] A review of the data from various studies by Laine and Peterson [1] is presented in

Table 148-2, demonstrating that patients with high-risk stigmata for hemorrhage (active bleeding or a nonbleeding visible vessel) have a high rebleeding rate that is associated with correspondingly high morbidity and mortality rates. Patients without high-risk stigmata (those with a clean ulcer base or flat, pigmented, spots) have an extremely low rate of rebleeding and a negligible mortality rate. [1] In a large study in the United Kingdom, Rockall and colleagues [17] combined the previously noted clinical predictors with the endoscopic appearance of lesions into a scoring system to predict the outcome of acute upper GI bleeding (Table 148-3). This validated scoring system demonstrates the utility of endoscopy in predicting the outcome of acute upper GI bleeding. Patients with a score of 2 or less have a risk for rebleeding of less than 5% and virtually no mortality. [17] The addition of endoscopic findings to the other clinical criteria dramatically increased the accuracy of prognosis prediction based on clinical factors alone. In another study, Jaramillo and colleagues [18] evaluated 1567 consecutive patients with acute upper GI bleeding using multivariate analysis. None of the patients had endoscopic therapy. The rebleeding rate was 20%. The factor that best predicted the risk for rebleeding was the presence of endoscopic high-risk stigmata (odds ratio [OR], 3.1; $P < 0.001$).

ENDOSCOPIC FINDING	RISK FOR REBLEEDING (%)	MORTALITY (%)
Active bleeding	55	11
Visible vessel	43	11
Adherent clot	22	7
Flat spot	10	3
Clean base	5	2

Adapted from ref. 1.

TABLE 148-2 Association of Endoscopic Findings with Risk for Rebleeding and Mortality in Patients with Bleeding Ulcers

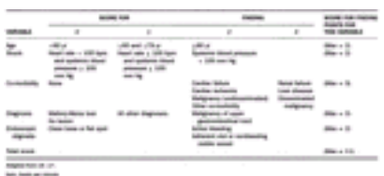

TABLE 148-3 Rockall Classification of Risk in Acute Upper Gastrointestinal Bleeding

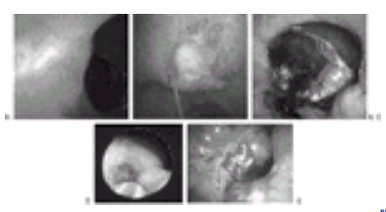

FIGURE 148-1. (See Color Fig. 148-1.) Endoscopic photos of stigmata of recent hemorrhage. **A:** Clean base. Duodenal ulcer with a clean base. No evidence of protruding or pigmented areas is present. The rebleeding rate in this lesion is near zero. **B:** Flat spot. Pyloric channel ulcer with a flat, black spot in the center. The risk for rebleeding in this lesion is less than 10%. **C:** Adherent clot. Duodenal ulcer with a large adherent clot in the base that could not be removed with vigorous irrigation. The nature of the lesion beneath the clot is unknown. The risk for rebleeding in this lesion is intermediate (up to 25%). **D:** Nonbleeding visible vessel. A prominent protruding visible vessel with a bipolar probe in the foreground. This lesion has a high risk for recurrent bleeding (40% to 50%) without endoscopic therapy. **E:** Active bleeding. A duodenal ulcer with blood vigorously flowing. This is the lesion with the highest risk for continued or recurrent bleeding (approaching 60%). Endoscopic hemostasis can control active bleeding in most patients and can reduce the risk for rebleeding.

Other endoscopic findings have been suggested to increase the risk for recurrent bleeding. These include the size and location of the ulcer. [19], [20] Both of these findings relate to the proximity of the base of the ulcer to a major artery or arteriole. Lesions in the posterior duodenal bulb may penetrate into the gastroduodenal artery. Likewise, lesions high on the lesser curvature of the stomach may involve the left gastric artery. Doppler ultrasound probes have been used in an attempt to identify large vessels in or near ulcers to predict the risk for rebleeding. These attempts have met with moderate success in targeting endoscopic therapy but have limited sensitivity and specificity. [21], [22] The technology has not been widely accepted.

Diagnostic Role of Urgent Endoscopy (Endoscopic Triage)

Although it may seem intuitively obvious that endoscopy would improve patient outcomes, randomized trials have indicated that *diagnostic* endoscopy alone does not improve mortality, rebleeding rates, the need for surgery, or hospital stay. [23], [24] and [25] Most patients (75% to 80%) with bleeding ulcers stop bleeding spontaneously, limiting the impact of early endoscopy for diagnosis. [16], [18], [25] Based on these data, patients are often initially admitted to the hospital for volume resuscitation and observation, then undergo diagnostic endoscopy electively. Because urgent endoscopy improves morbidity and mortality only in the subset of patients with persistent or recurrent bleeding, both the National Institutes of Health and the American Society for Gastrointestinal Endoscopy have recommended that endoscopy be urgently performed in the subset of patients for whom endoscopic therapy is anticipated. [26], [27]

Recent data have raised the possibility of endoscopically identifying patients with acute upper GI bleeding who are at low risk for rebleeding and require limited medical care. Laine and colleagues [28] demonstrated that patients with clean ulcer bases or Mallory-Weiss esophageal tears had only a 2% risk for rebleeding and could be safely fed and immediately discharged from the hospital. Rockall and colleagues [29] applied the previously mentioned scoring system to 2531 consecutive patients seen with acute upper GI bleeding in the United Kingdom. They identified 29.4% of the patients who had a score of 2 or less and a negligible risk for rebleeding or morbidity. Based on these data, the authors suggested that a large number of patients without clinical risks or endoscopic stigmata for rebleeding could be safely managed without hospital admission. They stated, "… hospital admission has usually been regarded as obligatory until the risk of further haemorrhage has receded. This policy means that some patients at low risk stay in hospital for longer than is necessary especially when diagnostic endoscopy is delayed." [17] Kodali and colleagues [30] demonstrated that 21% of patients with bleeding from peptic ulcer disease had a clean ulcer base at endoscopy. These patients had a 3% risk for rebleeding. These authors concluded that this group of patients could be safely managed as outpatients.

Two trials have retrospectively reviewed hospitalizations for upper GI bleeding, and then developed criteria for same-day discharge (endoscopic triage) that were prospectively applied with success. [31], [32] In a single-center study, Lee and associates [33] demonstrated a decrease in costs of care associated with decreased resource use in patients undergoing endoscopic triage for nonvariceal upper GI bleeding, with no increase in morbidity. Another similar-sized prospective multicenter study, however, demonstrated no difference in the length of stay or clinical outcomes of patients undergoing urgent (preadmission) endoscopy or elective endoscopy after admission. [34] The major difference between these studies was the number of patients discharged from the emergency department. In the first study, 46% of the patients randomized to receive urgent endoscopy were discharged from the emergency room by the endoscopist, whereas in the second study, the decision regarding admission was left to the attending physician (not the endoscopist), and only 4 of 47 eligible patients avoided hospitalization. These studies demonstrate the difference between efficacy and clinical effectiveness. In a situation in which the endoscopic findings drive subsequent clinical care, endoscopic triage can significantly decrease hospitalization and costs of care for patients at low risk for rebleeding. If the endoscopic findings do not affect subsequent care, then the information will not affect resource use.

Additional data regarding the effectiveness of endoscopic triage are provided by two studies by Hay and colleagues. [35], [36] In the first study, the authors developed a treatment guideline for upper GI bleeding retrospectively, demonstrating the potential for substantial cost savings using endoscopy and three other factors as predictors of low risk and possible discharge. [35] In the second study, they used an alternating time approach to evaluate implementation of the guideline. [36] During the intervention periods, they aggressively modified physician behavior in the emergency department, promoting the previously developed clinical guideline that included

preadmission endoscopy and discharge of patients at low risk for rebleeding. During the control periods, Hay and colleagues allowed the physicians to make the admission decisions. Compliance with the guideline was 70% during the period of aggressive promotion and only 30% during the other periods. Aggressive implementation of the guideline resulted in substantially decreased hospitalization and costs without compromising patient care or satisfaction. The cost savings were lost during the control periods.

It appears that urgent endoscopy can substantially reduce the costs of care for upper GI bleeding by identifying a group of patients who can be safely treated as outpatients only if the endoscopic findings, in conjunction with other clinical information, can alter the subsequent care of the patient.

ENDOSCOPIC THERAPY FOR BLEEDING PEPTIC ULCERS

As previously noted, about 20% of patients with nonvariceal upper GI bleeding have persistent or recurrent bleeding. There is compelling evidence that endoscopic therapy reduces both the morbidity and mortality in these patients. [26], [27] All available endoscopic techniques appear to have similar results, but they vary in their approach to sealing the bleeding vessel and maintaining hemostasis. The major methods may be divided into thermal coagulation, injection therapy, and mechanical compression.

Thermal Coagulation

Thermal therapy of bleeding peptic ulcer disease is based on the understanding that the ulcer erodes into a submucosal vessel. Therapeutic interventions using heat have the goal of coagulating or sealing this vessel to stop active bleeding and prevent rebleeding. [8]

Tissue has a characteristic response to thermal energy. When heated to 50° to 60°C, intracellular proteins become denatured, destroying the tissue and forming a white coagulum. This also produces a surrounding area with a zone of sublethal damage manifest by erythema and edema. [37] When heated to 100°C, the water in cells is vaporized, resulting in cavitations and tissue ablation. The goal of endoscopic therapy is to coagulate the bleeding lesion and create a ring of edema that reduces blood flow and promotes hemostasis without causing tissue vaporization or cavitation that may exacerbate the bleeding. There are multiple methods of delivering different types of energy capable of raising tissue temperature, including light energy (lasers), electrical energy (monopolar or bipolar [multipolar] electrocoagulation), and direct application of a heated surface.

Laser Therapy Laser therapy was the first endoscopic method of hemostasis extensively evaluated. Although different lasers produce wavelengths of light with specific effects on tissue, the most commonly used laser is the neodymium:yttrium-aluminum-garnet (Nd:YAG) laser. The light from this laser (1060-nm wavelength) can be delivered through a flexible quartz fiber that is introduced through the working channel of a standard endoscope (Fig. 148-2). As the light strikes the tissue, it is converted to thermal energy, creating the heat necessary for coagulation. Tissue contact is avoided while coagulating a ring of tissue surrounding the bleeding site. Other lasers (argon, KTP) commonly used in endoscopic therapy produce light of a shorter wavelength with more limited tissue penetration. As a result, they are less effective in the setting of acute upper GI bleeding. The Nd:YAG laser has been shown to stop bleeding lesions in the stomach and duodenum in more than 90% of cases and to reduce rebleeding to less than 20%. [38]

FIGURE 148-2. Laser fiber. A quartz laser fiber with covering plastic sheath. Fibers may also have a sheath that provides coaxial gas flow. The quartz fiber conducts the laser light but is flexible enough to be passed through an endoscope channel. Similar fibers are used for argon, Nd:YAG, and KTP lasers.

Disadvantages of laser therapy include variable control of the depth and type of tissue injury, with the potential for overheating, vaporization, and charring of tissue that exacerbates the bleeding. There is a small risk for perforation (less than 2%) when used in the stomach. When coaxial gas is used around the quartz fiber, abdominal distention can result. The equipment necessary for laser therapy is expensive and requires special expertise for its use and maintenance. Most lasers are not portable and require special safety precautions and training. Because of these logistic issues and the availability of equally effective alternatives, lasers are now rarely used in the setting of acute upper GI bleeding.

Electrocoagulation Electrocoagulation uses an electrical current passed through tissue to generate thermal energy to coagulate the target tissue. The earliest use of electricity used an electrified endoscopic biopsy forceps touched to the tissue. In this case, the electrical current flowed from the tip of the forceps through the patient to an electrical pad placed on the hip or thigh. In this method, the degree of tissue injury depends on electrical current density at the site of contact with the forceps and local tissue factors that are poorly controlled. The result is an unpredictable depth of injury. For this reason, monopolar electrocoagulation is usually avoided in the treatment of acute upper GI bleeding. [26] To avoid the problems associated with monopolar electrical current, endoscopic probes have been developed that have both the positive and negative electrical poles at the end of a catheter (Fig. 148-3). An electrical circuit is completed between a positive and negative electrode (bipolar electrocoagulation). To allow tangential application, probes have been designed with multiple pairs of positive and negative electrodes on the tip (often referred to as *multipolar electrocoagulation*) or alternating positive and negative electrodes circling the end of the probe. The result is limited delivery of electrical current within the diameter of the probe, using the electrical resistance of the tissue to heat only that area where the probe is placed. The depth and degree of tissue injury are well controlled. The probe also allows for direct pressure to be applied to the bleeding lesion to seal the vessel (coaptive coagulation). [39]

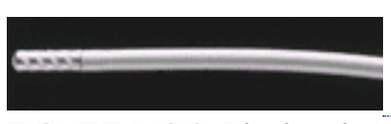

FIGURE 148-3. Bipolar electrocoagulation catheter. Electrodes circle the end of the catheter, and a central channel provides vigorous water irrigation. Some electrodes have a retractable central injection needle to allow combined electrocoagulation and injection therapy.

Multipolar electrocoagulation was evaluated by Laine [40] in a randomized prospective trial of patients with acute upper GI bleeding. Hemostasis was achieved in 90% of the patients treated with the multipolar probe, compared with 13% treated with medical therapy. [40] There was also a significant reduction in transfusion requirement, hospital stay, morbidity, and costs for the patients receiving endoscopic therapy. In a similar trial limited to patients with nonbleeding visible vessels, the rebleeding rate was reduced by more than 50% in the treatment group (18% compared with 41%). [41] There was a similar improvement in transfusion requirements, hospital stay, need for surgery, and cost in the treatment group. Multiple other trials have demonstrated similar results when using bipolar or multipolar electrocoagulation. [42], [43]

Heater Probe Direct application of thermal energy can be applied to bleeding lesions using a heater probe. This device uses a catheter with a heating element at the tip that can be passed through the endoscope channel (Fig. 148-4). The heating element delivers a prespecified amount of energy by adjusting the time of the energy burst. The tissue effect is limited to the area in contact with the probe. As with the bipolar electrode, pressure can be applied directly to the bleeding site, allowing coaptive coagulation of bleeding essels.

FIGURE 148-4. Heater probe. A flexible catheter tipped with a heating element that is placed directly on the tissue for coagulation. No electrical current passes through the patient. Vigorous water irrigation is possible through the central channel of the catheter.

The efficacy of this method is very similar to that of the bipolar probe. Initial randomized trials demonstrated that hemostasis could be achieved in actively bleeding lesions in 78% of patients, compared with none in a sham treatment group. [44] In a study limited to patients with nonbleeding visible vessels, there was, again, a greater than 50% reduction in the rebleeding rate (10% compared with 26%) in the treatment group. [45] As with bipolar electrocoagulation, multiple other studies have confirmed the ability of heater probe therapy to control active bleeding and reduce the risk for rebleeding from high-risk lesions. [8]

Argon Plasma Coagulation A more recently developed method of endoscopic hemostasis is the argon plasma coagulation (APC) device. This device uses a flow of ionized argon gas to conduct electricity from the power generator to the tissue (Fig. 148-5). It is a type of monopolar electrocautery and should not be confused with

the argon laser. Few controlled studies have evaluated the efficacy of this method of hemostasis. The distribution of tissue injury depends on the flow of the gas and density of the electrical current at the tissue surface. One small study of 41 patients found no significant difference in results comparing APC with heater probe therapy in acute upper GI bleeding. [46] This methodology, however, has not undergone the rigorous evaluation of other thermal hemostasis methods. [47], [48]

FIGURE 148-5. Argon plasma coagulation catheter. This hollow catheter produces a flow of electrically charged argon gas when activated. The catheter should not come in contact with the tissue. It allows rapid electrocoagulation of larger areas of tissue, or areas that are difficult to reach with a contact probe.

Technique of Thermal Therapy The technical approach to thermal treatment of bleeding lesions is different in contact methods (bipolar electrocoagulation, heater probe) and noncontact methods (laser, APC). Sapphire tips can also be attached to laser fibers, creating a light-heated tip for contact therapy. Bleeding in peptic ulcer disease occurs when the ulcer erodes into a vessel. Either the vessel wall or the clot in the vessel can be identified endoscopically as a "visible vessel" (Fig. 148-6, Color Fig. 148-6). In the noncontact method, the vessel feeding the bleeding site is cauterized below the surface by treating circumferentially around the clot or point of bleeding (Fig. 148-7). This has the advantage of not disrupting any clot that may have formed in the lesion, but it does not seal the vessel with coaptive coagulation. Contact probes, on the other hand, are used to apply direct pressure to the bleeding site, using the water-jet capability of the probes to clear away surrounding blood and debris. Multiple bursts of energy are applied to the lesion until hemostasis or adequate coagulation has been achieved (Fig. 148-8). Larger probes are more effective, as are longer pulses of lower energy. [49] The design of the contact probes also allows for tangential treatment of lesions that are in difficult locations. The disadvantage of contact probes is tissue adherence to the probe. When the probe is moved, the clot and coagulated tissue can be dislodged, leading to more vigorous bleeding that rarely requires surgical intervention. Vigorous bleeding was precipitated in 18% of patients treated with contact probe therapy in one randomized trial. [42]

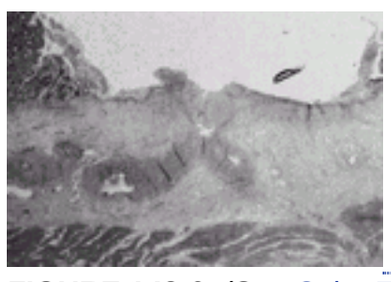

FIGURE 148-6. (See Color Fig. 148-6.) Visible vessel. This classic image demonstrates the histology of a bleeding vessel in the base of an ulcer. The submucosal vessel is seen extending to the ulcer surface, with a thrombus present in the "hole" in the arterial wall. (Courtesy of C. Paul Swain, M.D.)

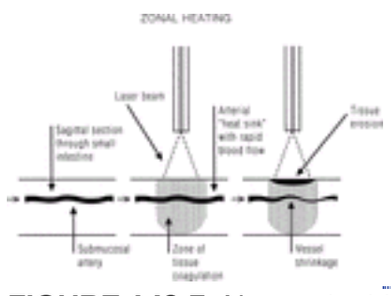

FIGURE 148-7. Noncontact therapy diagram. Coagulation of tissue with noncontact methods, such as laser therapy or argon plasma coagulation. The area surrounding the bleeding site is treated with the aim of coagulating the submucosal vessel. Even when the vessel is not coagulated, the resulting circumferential edema results in enough tamponade to control bleeding.

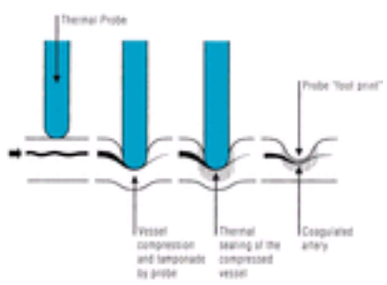

FIGURE 148-8. Contact therapy diagram. Treatment with contact probes focuses on coaptive coagulation of the bleeding site. Considerable pressure is applied with the probe to tamponade the bleeding vessel. Multiple bursts of energy are then delivered to coagulate the lesion and seal the bleeding vessel. Care must be taken to avoid dislodging coagulated tissue, which can adhere to the catheter tip. (From Johnston JH, Jensen DM, Auth D. Experimental comparison of endoscopic yttrium-aluminum-garnet laser, electrosurgery, and heater probe for canine gut arterial coagulation. Importance of compression and avoidance of erosion. Gastroenterology. 1987;92:1101.)

The approach to patients with adherent clots in the base of ulcers remains controversial. Adherent clots are those that cannot be dislodged by prolonged application of a vigorous water jet. Unless the clot is removed, the nature of the underlying lesion cannot be determined. Laine and colleagues [50] evaluated 46 patients with adherent clots. All received a directed water-jet wash for 5 minutes to try to dislodge the clot. After this time, 57% of the patients still had clots obscuring the bleeding site, 15% had active bleeding, and 13% had nonbleeding visible vessels. The remainder had clean ulcer bases or flat spots. Endoscopic therapy was applied to the high-risk lesions uncovered by the removal of the clot, but not in patients with clots that remained. This approach resulted in an excellent outcome. Only one patient (who had an actively spurting vessel) rebled, and there were no deaths. An alternative approach of cold snare guillotine removal of the clot and treatment of underlying lesions has been advocated by Jensen and colleagues. [51] In a pilot study, 11 patients with adherent clots were randomized to either medical therapy or snare removal of the clot with endoscopic therapy as appropriate. Rebleeding occurred in 40% of the patients treated medically and in none of the patients in whom the clot was removed. When clot removal is contemplated, it should be done only if the endoscopist is prepared to deal with any underlying lesion, including vigorous bleeding. Freeman [16] has advocated injection therapy of the base of the clot before attempted mechanical removal to reduce the risk for vigorous bleeding. It appears that the clinical outcome of patients with adherent clots found at endoscopy depends on the lesion under the clot and the stability of the hemostatic plug formed by the clot. Preliminary reports have suggested that the rebleeding rate in patients with nonbleeding adherent clots can be reduced from between 25% and 30% to between 0% and 5% with clot removal and endoscopic therapy of the underlying lesion. [52], [53] The removal of such clots should be determined by the patient's condition and the availability of appropriate therapy in the event that clot removal precipitates vigorous bleeding.

Injection Therapy

The least expensive method of endoscopic therapy for upper GI bleeding is the use of a simple needle catheter to inject the bleeding site with a compound selected to achieve and maintain hemostasis. There are different classes of materials used for injection therapy, including hemostatic agents, sclerosants, and thrombogenic agents.

The most commonly used material is diluted epinephrine, usually in the readily available 1:10,000 dilution. This is injected submucosally around the bleeding site in

aliquots of 1 to 2 mL to a total volume of 5 to 10 mL. On occasion, larger volumes may be required to achieve the desired tamponade effect. Hemostasis is postulated to occur as a result of a combined effect of vasoconstriction from the epinephrine and tamponade of the bleeding vessel by the volume effect of the injection. [54], [55] The efficacy of this method was demonstrated by Chung and colleagues [56]: 68 patients with actively bleeding ulcers were randomized to receive either injection therapy with 1:10,000 epinephrine or medical therapy. Initial hemostasis was achieved in all patients in the injection therapy group. There were fewer patients who required surgery (5 versus 14), fewer blood transfusions, and a shorter hospital stay in the epinephrine-treated group. These findings were validated in a larger prospective trial of 98 patients with acute upper GI bleeding. Subjects were randomized to receive either 1:10,000 epinephrine injected in 1- to 2-mL aliquots around the lesion and 1 to 2 mL of ethanolamine into the bleeding site, or medical therapy alone. The rebleeding rate was 17% in the injected group and 47% in the control group (*P* = 0.011). There was also reduced mortality, need for surgery, transfusion requirement, and the length of hospital stay in the endoscopic treatment group. [57]

Other agents have also been used for subcutaneous injection to tamponade bleeding upper GI lesions. These include normal saline, 3% saline, distilled water, and 50% glucose in water. These solutions have an effect not significantly different than that of 1:10,000 epinephrine. [58], [59]

Sclerosing agents include absolute ethanol, polidocanol, ethanolamine, and sodium tetradecyl sulfate. An important difference between these compounds and those that act by local tamponade is that the sclerosants cause tissue desiccation or destruction. As a result, much smaller volumes must be used (e.g., for absolute alcohol, 0.1 to 0.2 mL/injection with total volume of less than 2 mL). [56] Even with these volumes, there is a risk for extending tissue destruction and complications. [60], [61]

Initial trials demonstrated that ethanol injection could control active upper GI bleeding, [62] reduce the rate of rebleeding, and improve clinical outcomes [63] to a degree similar to that seen with epinephrine injection. Other sclerosants have mainly been used in conjunction with epinephrine injection. Studies have suggested that the combination of a sclerosant with epinephrine has no better outcome than epinephrine alone. [64], [65] and [66] One study randomized 64 patients with bleeding ulcers to receive epinephrine injection alone or epinephrine injection followed by alcohol injection. [67] The volumes of epinephrine used were similar in both groups. Initial therapy failed in one patient treated with epinephrine alone and in none who received dual-injection therapy. The rebleeding rate was lower when alcohol injection was added to epinephrine injection (16% versus 36%), but this was not statistically significant.

Thrombin acts to convert fibrinogen to fibrin, thus stabilizing the clot in a bleeding vessel. In a large European trial, [68] fibrinogen and thrombin were combined to form a fibrin-glue ubstance that was compared with polidocanol. There was no significant difference between the groups after a single injection, but there was a statistically significant improvement in the fibrin-glue group after repeated endoscopy with multiple injections. The need for multiple endoscopic sessions to achieve this result limits its utility in clinical practice. The one advantage of fibrinogen or thrombin glue may be the ability to achieve an effect similar to that of sclerosants without causing tissue damage. Thrombin alone has also been investigated but has not been widely adopted.

As the practice of endoscopic therapy progresses, it has become clear that the combination of injection therapy and thermal therapy can be very useful. In the setting of an actively bleeding lesion, initial therapy with injected 1:10,000 epinephrine or other agent around the bleeding site to tamponade the lesion helps slow the rate of bleeding and clears the field, facilitating subsequent thermal coagulation. [54] It has also been suggested that this injection pretreatment creates a cushion of fluid that reduces the complication of a transmural thermal injury. Chung and colleagues [55] performed a prospective trial of 270 patients randomized to epinephrine injection alone or epinephrine followed by heater-probe treatment. In actively bleeding lesions, there was a significant advantage in the group treated by both injection and thermal therapy, with a reduction in both rebleeding and the need for surgery. [69] In the same study, there was a statistically nonsignificant trend toward better outcomes (rebleeding and the need for surgery) in the group that received dual therapy compared with injection therapy alone. The failure of the difference in the rebleeding and surgery rates to achieve statistical significance was likely due to a beta error resulting from the small number of patients in the trial.

In summary, epinephrine injection given to slow or control active bleeding should be followed by a second treatment modality. The optimal role of epinephrine may be as an initial therapy, in preparation for thermal modalities or sclerosants to achieve more effective long-term hemostasis. On the other hand, as compared with a thermal method alone, injection of epinephrine plus a thermal method appears to have no significant advantage in terms of rebleeding.

Hemostatic Clips

The use of hemostatic clips was first reported in 1975 by Hayashi and colleagues. [70] The technique was based on the use of clips to seal specific vessels in both surgery and radiology (Fig. 148-9). As the logistics of clip placement were improved, the use of clips was shown to be effective in the treatment of active bleeding in uncontrolled studies. [71], [72] Binmoeller and colleagues [72] treated 88 patients with either active bleeding (n = 78) or nonbleeding visible vessel (n = 10) with endoscopic clips. Initial hemostasis was achieved in all patients with an average of three clips. With a mean follow-up of longer than 1 year, only 5 patients rebled, all of whom were successfully retreated with clips. In randomized studies comparing the use of clips to injection therapy, thermal therapy, and injection therapy plus clips, there was no significant difference in the outcomes among the three approaches. [73], [74] One study compared the use of endoscopic clips to heater probe in ulcers with major stigmata of hemorrhage. The rebleeding rate was significantly lower in the patients treated with clips (1.8% versus 21%). [75] The study has been criticized, however, because the rebleeding rate in the heater-probe group is at the high end of that reported in other studies, and the rebleeding rate in the clip-treated group is remarkably low. [76] Nonetheless, the same study noted that logistic issues prevent adequate treatment of lesions that are in specific locations (lesser curve of the stomach, posterior gastric wall), where it is difficult to achieve an en face endoscopic approach for clip placement. Rare complications have also been reported with clip placement. [77]

FIGURE 148-9. Hemostatic clip. A hemostatic clip with its delivery catheter is shown. The clip can be rotated to achieve optimal position, then closed and released from the delivery device. Multiple clips are often required to achieve the appropriate hemostatic effect.

Summary of the Efficacy of Endoscopic Therapy

It is clear that the application of endoscopic therapy to bleeding from peptic ulcer disease is a successful means of treating active bleeding and reducing the risk for recurrent bleeding. Two large metaanalyses combining studies of endoscopic hemostasis in acute upper GI bleeding demonstrate the efficacy of thermal therapies and injection techniques. [43], [78] Very similar outcomes are achieved regardless of the treatment method used (laser, bipolar, heater probe, or injection). [78] In one analysis, thermal endoscopic therapy reduced the rate of rebleeding (OR, 0.38), surgery (OR, 0.36), and mortality (OR, 0.55). The overall complication rate requiring surgery or causing perforation of therapy was less than 1% for each method. Subgroup analysis showed no significant differences in the outcomes among the various thermal methods of therapy. The benefit of therapy was limited, however, to patients with endoscopic stigmata suggesting a high risk for rebleeding (visible vessels, active bleeding), but was not seen in patients with low-risk stigmata (clean ulcer base, flat pigmented spots, adherent clots). This confirms the importance of endoscopic criteria in the selection of patients for therapy. As mentioned previously, rebleeding occurs in only 20% to 25% of patients. Those with a low risk for rebleeding do not require endoscopic therapy. In the subset of patients with a high risk for rebleeding, however, endoscopic therapy improves clinical outcomes.

NONULCER UPPER GASTROINTESTINAL BLEEDING

Upper GI bleeding may also occur from non–peptic ulcer sources. The most common of these is a tear at the gastroesophageal junction, called a Mallory-Weiss tear, which accounts for 5% to 14% of upper GI bleeding. [79], [80] Bleeding usually stops spontaneously, but when persistent bleeding is found, this lesion should be treated in a similar manner as bleeding from peptic ulcers. Patients who have self-limited bleeding have a good prognosis. [27], [81] Active bleeding responds to the same methods outlined for peptic ulcers, including thermal therapy, injection therapy, and endoscopic clips. [79], [82] Another method of hemostasis reported for therapy of a bleeding Mallory-Weiss tear is band ligation. [83], [84]

Dieulafoy lesion is an aberrant submucosal artery that erodes into the lumen of the stomach and often presents as intermittent massive upper GI bleeding. [85] Although

more common in the cardia, these lesions can occur anywhere in the stomach and have also been reported throughout the GI tract. The overall prevalence of Dieulafoy lesion in upper GI bleeding is probably less than 5%. [80] Because these lesions are not accompanied by ulceration, they are often difficult to identify unless seen while actively bleeding. The infrequent nature of Dieulafoy lesion has prevented randomized trials on different types of endoscopic therapy. Combining uncontrolled experience, it appears that endoscopic therapy is effective for initial hemostasis in 96% of patients and achieves permanent hemostasis in 85% to 90%. [85], [86] As with Mallory-Weiss tears, ligation of the lesion with either a rubber band [87] or endoscopically placed loop, [88] has been reported to be successful.

P>Malignant and benign tumors are rare (less than 1%) causes of acute upper GI bleeding. [88] They more commonly present with other symptoms and chronic low-grade blood loss. When the cause of acute bleeding, they may respond to endoscopic therapy as outlined for peptic ulcer disease. [7], [88]

Another rare cause of acute upper GI bleeding is a vascular ectasia. These lesions may occur as part of the hereditary hemorrhagic telangiectasia (Osler-Weber-Rendu) syndrome or sporadically. [89] The sporadic lesions are a common cause of lower GI bleeding but rarely present with hematemesis. These lesions also respond to standard thermal or injection therapy. [89], [90] An uncommon cause of acute upper GI bleeding is a diffuse antral form of vascular ectasia, often called a “watermelon stomach” or gastric antral vascular ectasia. [91] As with other vascular ectasias, chronic blood loss is a more common presentation of this lesion. Endoscopic laser therapy has been demonstrated to control acute bleeding and decrease transfusion requirements. [92] Other thermal methods have also been demonstrated to be effective. Contact methods (bipolar electrocoagulation, heater probe) may require more time to perform than noncontact methods (laser, APC). [93], [94], [95] and [96]

Bleeding from diffuse gastric erosions, as seen in critically ill patients, does not respond to endoscopic therapy and should be avoided by primary medical prevention. [94], [95], [96], [97] and [98]

REPEAT ENDOSCOPIC THERAPY

Some investigators have advocated routine second-look endoscopy in the setting of acute upper GI bleeding to assess the efficacy of the first therapy and retreat lesions that have persistent stigmata for rebleeding. [99] Multiple studies have randomized patients to receive second-look endoscopy or standard care. [100], [101], [102] and [103] Most have found a statistically significant difference in clinical outcomes of rebleeding or the need for surgery. [99], [100] and [101] Smaller studies have suggested a possible small difference in rebleeding rates. [102], [103] The number of additional endoscopic procedures, with the associated costs and risks, needed to prevent one episode of rebleeding appears to make this approach clinically inappropriate in most cases. [102]

It is clear that not all initial attempts at endoscopic therapy are equal. Patient condition, the presence of blood or clots in the stomach or duodenum, and other logistic factors can affect the efficacy of the treatment. The endoscopist often has a perception of how adequate the initial therapy was. In cases in which the therapy was thought to be suboptimal, a second procedure timed to provide optimal therapy may be appropriate. Routine second-look endoscopy for all patients, however, requires further evaluation before the benefits (e.g., fewer surgeries or deaths) can be demonstrated to outweigh the costs and risks to the patient.

On the other hand, it is clear that repeated endoscopic therapy is indicated in the setting of rebleeding. Lau and colleagues [104] randomized 92 patients who had recurrent bleeding after endoscopic thermal therapy to either endoscopic retreatment or surgery. The second endoscopic therapy session produced sustained hemostasis in 35 (73%) of the 48 patients randomized to endoscopy. Thirteen patients went to surgery, 11 of whom had a failure of the second endoscopic therapy and 2 of whom had a perforation resulting from the repeat endoscopy. Thirty-day mortality rates were similar in the two groups, but complications were less in the endoscopy group, of whom 7 patients (6 of whom ultimately went to surgery) had complications, compared with 16 of 44 patients in the surgery group. The authors concluded that a second attempt at endoscopic therapy can avoid surgery, with its associated complications, in many patients and does not increase mortality.

In most cases of rebleeding after endoscopic therapy, a second attempt at endoscopic therapy is indicated before resorting to surgical therapy. [105] Thermal therapy in this situation may be associated with a higher rate of perforation.

MEDICAL TREATMENT TO PREVENT REBLEEDING

The prevention of recurrent bleeding after endoscopic therapy has been the source of considerable controversy. It is clear that the ulcerogenic conditions that led to peptic ulcer bleeding should be ameliorated. In most cases, this means the discontinuation of nonsteroidal antiinflammatory drugs and treating *H pylori* infection. [106] There are no data, however, indicating that this improves the short-term (72-hour) outcome of the acute bleeding episode.

Normal hemostatic mechanisms are inhibited in the low pH of the stomach. Increasing the pH can improve coagulation and platelet aggregation. [107] In an attempt to increase gastric pH, intravenous histamine H_2 receptor antagonists have been routinely used in the setting of acute upper GI bleeding. Unfortunately, they have not been demonstrated to be better than placebo in reducing the rate of recurrent bleeding. [108] This is probably because of a decrease in the acid reduction effect of these agents over time. [109]

Despite the failure of histamine H_2 receptor antagonists, it is clear that profound acid suppression can reduce the rebleeding rate of peptic ulcers with high-risk stigmata. Khuroo and colleagues [110] randomized 220 patients to receive either oral omeprazole (40 mg twice daily for 5 days) or placebo. No endoscopic therapy was provided. Rebleeding occurred in 12 of 110 patients in the omeprazole group and in 40 of 110 patients in the placebo group ($p < 0.001$). Subgroup analysis showed the benefit of omeprazole to be limited to those patients with nonbleeding visible vessels. In a prospective study of patients with either active bleeding or nonbleeding visible vessels (high-risk stigmata), Lau and colleagues [111] randomized patients to either intravenous omeprazole (8 mg/hour for 72 hours) or placebo. All patients received endoscopic therapy before randomization. Rebleeding occurred within 72 hours in 5 of the patients receiving omeprazole and in 24 of the patients receiving placebo ($p < 0.001$). This difference persisted through the 30-day follow-up. Based on these data, it is clear that profound acid suppression using a proton pump inhibitor is an appropriate adjunct to endoscopic therapy of acute upper GI bleeding. High-dose constant proton pump inhibitor infusion is indicated as an adjunct to endoscopic therapy in patients with clinically significant bleeding and high-risk endoscopic stigmata.

Somatostatin, or its analog, octreotide, is standard therapy for bleeding esophageal varices. It has been suggested that the reduction in splanchnic blood flow caused by these agents could also control acute upper GI bleeding from other causes. In a metaanalysis, Imperiale and Birgisson [112] demonstrated a modest advantage of somatostatin or octreotide over histamine H_2 receptor antagonists in the prevention of recurrent bleeding from peptic ulcer disease. The authors suggested a possible role for these agents in patients awaiting endoscopy or when endoscopy is not available. It should be noted, however, that there was significant heterogeneity in the results of the metaanalysis, suggesting that combining the studies may not be appropriate. Limiting the analysis to rigorously done prospective double-blinded trials eliminated the heterogeneity but also suggested that there was no significant impact of somatostatin or octreotide on the outcome of the bleeding episode.

SUMMARY

Acute upper GI bleeding is a major medical problem in terms of both the burden of disease and the mortality of an individual episode. Endoscopy is the gold standard for diagnosis of the cause of bleeding and provides important prognostic information on the risk for continued or recurrent bleeding. Early endoscopy may also identify a group of patients at very low risk for rebleeding who can be treated safely as outpatients. Endoscopic therapy is effective in controlling active bleeding, reducing the risk for recurrent bleeding, and decreasing morbidity and mortality from bleeding peptic ulcers. It has a similar efficacy in bleeding from most nonulcer lesions in the upper GI tract. Endoscopic diagnosis and therapy is the standard of care for acute upper GI bleeding.

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CHAPTER 149

Douglas K. Rex and Emad Y. Rahmani

ENDOSCOPIC THERAPY FOR POLYPS AND TUMORS

The development of endoscopic methods for resection of polyps and tumors in the gastrointestinal tract has been an important advance and a useful complement to traditional surgical therapy. Endoscopic resection allows treatment without general anesthesia, provides shorter recovery time than open surgical resection, reduces costs by moving patients out of operating rooms and eliminating or reducing the need for hospitalization, and reduces the risk for resection relative to surgery. Palliative endoscopic therapy of advanced cancers similarly reduces costs and risks compared with surgical treatment. This chapter reviews methods of endoscopic therapy of polyps and tumors, the principles and goals of endoscopic therapy, selection of candidates for endoscopic treatment, and, finally, data from clinical trials on endoscopic treatment of specific lesions in the esophagus, stomach, duodenum, and colon. Endoscopic therapy for tumors in the pancreas and biliary tree is covered in Chapter 141.

METHODS OF ENDOSCOPIC THERAPY

Resection

Resection is the optimal method of endoscopic treatment of polyps and tumors because it provides the potential for curative removal. An immediate requirement for curative endoscopic resection is that a lesion can be reached with endoscopy and properly positioned in the endoscopic visual field to be treated in its entirety. Furthermore, it must be of a shape and sufficiently small size that endoscopic resection can be accomplished in a time frame that is practical and with a low to minimal risk for perforation. Given these features, curative endoscopic resection should be generally possible for disease confined to the mucosa and sometimes possible for mucosal disease extending into the submucosa or originating from and confined to the submucosa (Fig. 149-1). Disease extending into the muscularis propria or originating from it is usually not amenable to curative resection.

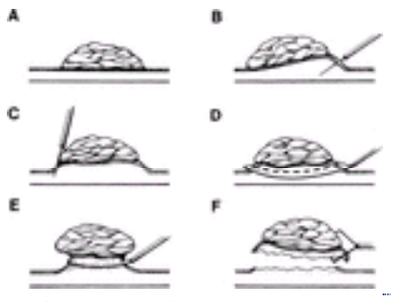

FIGURE 149-1. Mucosectomy technique. **A:** Sessile lesion limited to the mucosa. **B,C:** Physiologic saline is injected into the submucosa, creating a cushion effect. **D,E:** The lesion is resected using a loop snare. **F:** The entire specimen is retrieved.

A successful endoscopic resection is one resulting in complete removal of a polyp or tumor and retrieval of the pathological specimen without subsequent recurrence or complication. There are several general methods of endoscopic resection.

Cold Resection Cold resection refers to removal of polypoid tumors without electrocautery and can be performed with either biopsy forceps or polypectomy snares. Cold forceps removal is most readily applied to polyps of less than 4 mm in size, for which two to four bites is generally sufficient for apparent complete removal. Polyps of 1 to 2 mm generally appear to be completely removed in a single bite, although residual tissue may be left. [1] Minor immediate bleeding is very common and should not be treated with electrocautery. Delayed bleeding is rare because there is no cautery burn (delayed bleeding is associated with erosion of a cautery burn into a submucosal vessel). Cold snare resection has been applied to colon polyps of less than 7 mm in size. The polypectomy snare is placed around the base of a polyp, which is then guillotined by mechanical closure of the snare. Small amounts of immediate bleeding are typical. Like cold forceps resection, the major advantage appears to be a reduction in the risk for delayed hemorrhage. [2], [3] and [4] There may also be a reduction in the risk for perforation, although with small polyps, the risk for perforation is very low regardless of method. Safety of the technique for polyps larger than 7 mm has not been established. Initial evaluation suggests that cold snare resection is more effective than either hot or cold forceps at eliminating small polyps.([3])

Electrosurgical Resection

Diathermic snare. Endoscopic electrosurgical resection is typically accomplished using monopolar snares. A wire loop is placed around the base of a lesion with application of electrocautery followed by mechanical transection. The principal effect of electrocautery is tissue heating. Power sources for electrosurgery generate currents of typically more than 1,000,000 cycles per second (10 Hz). At these very high frequencies, the current alternates before sufficient time has transpired to depolarize muscle or nerve cells; thus, no muscle contraction occurs, and there is no danger to cardiac muscle. The current excites the molecules in the tissue, increasing their kinetic energy and raising tissue temperature. Electrosurgical resection typically produces temperatures of 45° to 100°C, with the degree of heating related to the high electrical resistance of the tissue. To obtain effective heating, the current must be concentrated through the smallest amount of tissue possible. This principle is referred to as *current density*. Current density is thus highly dependent on the tightness of closure of the diathermic snare. In fact, generated heat increases as the square of the current density. On the other hand, heat is directly proportional to the power setting and the length of time of power application. Thus, current density (snare closure) is the dominant determinant of heating. Heating results in tissue coagulation. Coagulation necrosis is visible endoscopically as tissue whitening and reflects denaturation of proteins and cell death. A second effect of heating is desiccation and resultant shrinkage. Desiccation facilitates subsequent mechanical tissue transection. Shrinkage results in blood vessel constriction and thrombosis and thus prevents hemorrhage. Properly applied, the controlled tissue injury results in easy mechanical transection, sealing of blood vessels in the base of the lesion, and an increased probability of complete excision by destruction of any abnormal cells in the lesion base. Concurrently, the injury should be sufficiently controlled that the depth of coagulation does not extend through the muscularis propria and the lateral spread of the burn is not unnecessarily beyond the borders of the lesion. After electrosurgical resection, there is typically an erythematous rim of tissue at the margin of the white coagulum in the lesion base. This reflects edema and vasodilation from radiant thermal injury, and subsequently, this tissue sloughs. Electrocautery power sources offer options of pure coagulation current, pure cutting current, or a mixture (blend). Coagulating current consists of intermittent high-voltage pulses. High voltage allows current passage through desiccated tissue, producing a wide coagulation zone. Cutting occurs primarily by mechanical

closure through the soft coagulum. Cutting current is low voltage but continuous flow. The low voltage prevents current passage through desiccated tissue, so that the heating effect remains local. Similar settings of cutting and coagulation on many power sources are such that cutting is more powerful. Thus, high power and localized heating results in vaporization of tissue near the wire loop, which in turn creates the cutting effect. The overall incidence of bleeding with cutting and coagulation current is similar, but cutting current is associated with more immediate bleeding and coagulation current with a tendency toward delayed bleeding. [5],[6] Monopolar electrocautery currents travel from the device tip to a grounding pad placed on the skin. These pads should not be placed near electrically sensitive devices such as pacemakers. Monopolar electrocautery should not be used at all in patients with defibrillating pacemakers. Electrocautery snares are available in multiple sizes (e.g., jumbo, large, standard, diminutive, micro) and shapes (e.g., oval, crescent, hexagonal). Availability of larger snares is essential for large lesions, but small snares are easier to place around small lesions. Choice of shape is made by personal preference. One rationale for shapes other than oval is that oval snares may not maintain an open oval with repeated use. This factor has become irrelevant with the progressive move toward nonreusable snares. Barbs or spikes on snares may prevent slippage during mucosectomy. Bipolar snares should limit the depth of coagulum and are theoretically safer than monopolar snares. However, they are inefficient for removal of larger polyps [7] and have no proven advantage.

Hot Biopsy Forceps Hot biopsy forceps can be used to remove polyps 5 mm or smaller in size. Proper placement involves grasping the polyp tip with the forceps and stretching or “tenting” the polyp to produce a stalk, then applying electrocautery. Endoscopically, the coagulum is seen to fan out from the forceps tip. When the coagulum appears to have destroyed the entire polyp, the forceps are withdrawn. The tip of the polyp is preserved in the forceps for histological assessment. Hot forceps are generally the easiest instrument with which to grasp a small polyp but the most difficult for controlling the coagulum. Thus, if too much tissue is grasped, if tension is greater on one side of the polyp, or if current is applied too long, the coagulum can spread easily into normal tissue. This can result in a larger, deeper ulcer than desired with a risk for delayed bleeding or perforation. Bipolar forceps provide the advantages of monopolar forceps with a potentially decreased risk for a deep burn, although greater safety remains unproven. Hot forceps are available in small (2.3 mm) and larger (3.2 mm) diameters. The small forceps are preferable because it is easier to grasp only the polyp tip. Anecdotal reports of serious delayed hemorrhage abound, but a risk for hemorrhage with monopolar hot forceps of 0.41% in a retrospective series [8] is similar to that reported with diathermic snares. For colon polyps, the risk for complications with hot forceps is five times greater with right than left colon polyps. [9] Some experts have nearly abandoned hot forceps for colon polyps in favor of diminutive snares, especially in the right colon. [10] These small snares can be readily placed around most small polyps.

Mucosectomy *Mucosectomy* refers to removal of sessile or flat neoplasms confined to the mucosa or penetrating but not fixing the submucosa. The method was initially referred to as *strip biopsy*. In Japan, the technique has been used to remove barely elevated, flat, or even “depressed” neoplasms. In the West, the principal application has been resection of large sessile colon polyps (saline-assisted polypectomy). Mucosectomy begins with injection of the submucosal space under the lesion with a cushion of normal saline. [11] A variceal injection catheter is generally used. Hypertonic saline is preferred by some. [12] Either solution can be supplemented with epinephrine, generally in a 1:10,000 dilution. [12] Only the submucosal space of the intestinal wall has sufficiently loose connective tissue to allow accumulation of saline. Volumes up to 10 to 50 mL may be needed to create an adequate submucosal “mound” (see Fig. 149-1). The lesions can then be removed using a diathermic snare in one or several pieces (see Fig. 149-1). Endoscopic mucosal resection with a cap (EMRC) is a variant useful in removal of flat lesions less than 20 mm in maximal diameter. [13] A variceal ligation hood is placed on the endoscope tip, and the raised lesion and surrounding normal mucosa are suctioned into the ligation hood. The lesion is then snare-resected using a crescent-shaped snare mounted on the hood (Fig. 149-2). An alternative is snare resection after placement of a ligation band over the suctioned tissue (Fig. 149-3). The band effectively ensures hemostasis and falls off the tissue several days later. Another variation of this type of mucosectomy is the use of a dual-channel endoscope, with a biopsy forceps through one channel to “lift” the lesion and a snare through the other channel (Fig. 149-4).

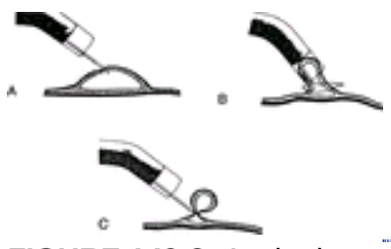

FIGURE 149-2. Aspiration mucosectomy using a snare. **A:** Physiologic saline is injected into the submucosal layers. **B:** The lesion-bearing mucosa is captured inside the cap attached to the endoscope tip under full suction. **C:** Mucosal lesion is snared tightly with the snare device passed through the biopsy channel of the endoscope.

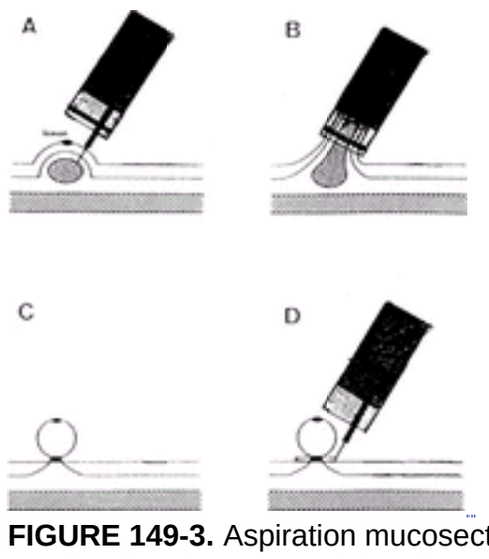

FIGURE 149-3. Aspiration mucosectomy using band ligation. **A:** Physiologic saline is injected into the submucosal layer. **B,C:** The lesion is suctioned into the cap-fitted endoscope and then ligated with a rubber band. **D:** The ligated “polypoid” lesion is snared and resected with electrocautery.

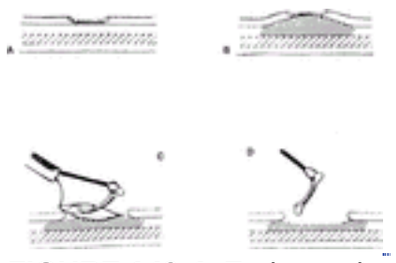

FIGURE 149-4. Endoscopic mucosectomy with the lift-and-cut technique. **A:** Flat lesion. **B:** Physiologic saline is injected into the submucosal layer. **C:** The lesion is slightly elevated using a biopsy forceps, and a snare is introduced through the second channel of the dual-channel endoscope. **D:** The lesion is resected and retrieved.

Mucosectomy has expanded the spectrum of endoscopically removable lesions. Very flat and depressed lesions cannot be removed by conventional snaring because the margin is not raised enough to enable grasping with the snare. Mucosectomy is preferred by some for sessile lesions that could be removed by conventional snaring because the saline cushion theoretically reduces the risk for a deep burn that might result in perforation, although this principle remains unproven. Mucosectomy should not be attempted for lesions that do not “raise” during submucosal injections. Failure to raise (the nonlifting sign) indicates fixation of the submucosa by tumor. [14] Such lesions should be surgically resected. However, the nonlifting sign is often present if there has been a previous attempt at removal; in this instance, it does not indicate the need for surgery. Japanese investigators have correlated the nonlifting sign with the depth of submucosal invasion in early colorectal cancer. [15]

Ablative (Destructive) Therapies

Ablation (tissue destruction) is indicated for relief of obstruction or control of bleeding when neither curative endoscopic resection nor surgical resection is feasible (Fig. 149-5). Ablation may also be considered when curative treatment is desired but endoscopic resection cannot be achieved and for any reason surgery is not selected. Ablation is generally less desirable than resection because it does not provide tissue for histological evaluation. Thus, focal cancer can go unrecognized when ablative therapy is used, but its presence might influence a decision regarding surgery. Alternatively, the risk for cancer may be so small that the cost-effectiveness of resection and submission of tissue for histological evaluation can be questioned. Such an approach has not been evaluated but may be applicable when techniques for real-time assessment of histology (e.g., high-magnification endoscopes combined with chromoscopy [16]) or light-induced fluorescence microscopy [17] are available. Ablation can be achieved by induction of coagulation necrosis (temperatures 45° to 100°C) or by vaporization (temperatures greater than 100°C).

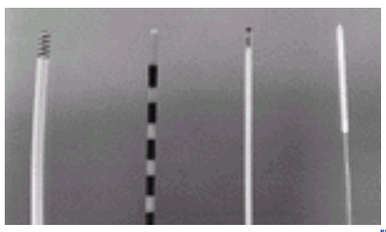

FIGURE 149-5. Probes used in mucosal ablation in the gastrointestinal tract. **A:** Bipolar probe. **B:** Argon plasma coagulation probe. **C:** Nd:YAG laser probe. **D:** Photodynamic therapy probe.

Bipolar Electrocoagulation In bipolar electrocoagulation (BEC), current flows out one electrode and back to another electrode, both of which are located in the device tip. No grounding plate is needed. During application, tissue desiccation occurs, and resistance rises, keeping the current path localized and limiting depth of injury. Bipolar probes are highly effective for endoscopic hemostasis. Relative to noncontact methods, bipolar probes are easy to use in the setting of active hemorrhage. They can be effectively applied to the surface of bleeding tumors to achieve hemostasis and have been used for eradication of residual flat neoplastic tissue that cannot be snare-resected and for ablation of very small polyps. Advantages of BEC probes are low cost of the cautery unit (although the probes are relatively expensive) and ease of application (en face or tangential). BEC probes are not useful for ablation of malignant tumors because of inadequate depth of injury and a tendency for tissue to stick to the device, requiring frequent cleaning. The principal use of this modality for tumor ablation has been the BICAP bipolar tumor probe (Circon, ACMI, Santa Barbara, CA). Superficial tissue destruction and tissue sticking have limited the usefulness of this device.
Heater Probe The heater probe uses a thermal element that heats the device tip and results in tissue coagulation. The power settings range from 5 to 30 joules, and the length of the pulse is determined by the power setting. Heat is transferred regardless of the degree of coagulation, so that the depth of injury tends to be deeper than with BEC probes. [18] The primary role of the heater probe has been in hemostasis rather than treatment of neoplasia.
Laser Laser (*l*ight *a*mplification by *s*timulated *e*mission of *r*adiation) delivered by endoscopic probes is an effective modality for relief of malignant obstruction, control of hemorrhage from tumor, and curative ablation of benign growths. The use of probe-delivered laser has declined for treatment of cancer and polyps because of purchase cost and need for repeated treatments. For malignant obstruction, stenting with self-expanding metal stents has increased in popularity relative to laser. For large benign sessile growths, mucosectomy and localized surgical resection (through transanal or laparoscopic route in the rectum and colon, respectively) have reduced the use of laser. Laser's lack of portability and depth of penetration have almost eliminated its use in hemostasis, so that endoscopic probe–delivered laser therapy has undergone a general decline. However, laser is used in the emerging field of photodynamic therapy (PDT), and, despite the previous comments, probe-delivered laser is effective and remains available in many centers. Laser depends on an active lasing medium. The medium is a material that, when illuminated, first excites electrons to higher energy states and then releases photons. Two media have been particularly useful for treatment of gastrointestinal tumors: the rare earth neodymium doped in yttrium-aluminum-garnet (Nd:YAG) and argon gas (Table 149-1). The lasing medium is housed in a nearly closed-space cylinder with mirrors at each end (Fig. 149-6). As photons are released, they are reflected and focused by the mirrors. Repeated excitation and decay of electrons in the lasing medium allows amplification of the process. An opening in one of the mirrors allows release of a monochromatic beam of photons whose wavelength is determined by the properties of the lasing medium.

LASERS	WAVELENGTH (nm)
Argon	458–514
KTP	532
Nd:YAG	1060

TABLE 149-1 Currently Available Thermal Lasers

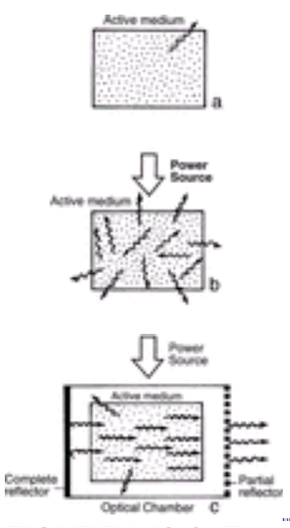

FIGURE 149-6. Essential components of lasers. **A:** Active medium of the laser in its nonactivated state. There are few, if any, excited atoms. **B:** A power source is applied to the active medium to produce a population inversion with many excited atoms. There is no directionality to the emitted photons and little amplification. **C:** The addition of an optical chamber with two reflective surfaces provides the directed output and feedback and amplification between the back reflector and the front partial reflector.

Laser effects on tissue depend on the extent of absorption of the laser beam. Absorption results in tissue excitation at a molecular level, resulting in heating and coagulation or vaporization. Absorption is a function of wavelength of the laser beam and the tissue content of water and hemoglobin, the principal absorption molecules. Scattered light may heat adjacent tissue, depending on the tissue-wavelength interaction of this tissue. If the tissue-wavelength interaction is not optimal for absorption, light is transmitted or reflected, resulting in no tissue damage. The Nd:YAG laser delivers an invisible light with wavelength in the near-infrared spectrum at 1.06 µm. The power of most clinically available units is up to 120 W. The light is pointed endoscopically at the target through a red aiming beam. The argon laser delivers a blue-green light of wavelength 0.50 µm. The depth of penetration using argon is less than BEC. [19] On the other hand, Nd:YAG penetration is five or more times greater than argon, and heating can readily reach a level (more than 100°C) allowing vaporization. [19] Thus, the Nd:YAG laser has been the most useful probe-delivered laser for treatment of gastrointestinal tumors. Tissue heating is affected by the power setting, pulse duration, and probe distance from the tumor, which determines the spot size of the beam. Continuous (nonpulsed) therapy of tumors using Nd:YAG laser leads to conduction of heat energy to surrounding tissue, increasing patient discomfort and increasing perforation risk. Pulsed therapy helps focus heating on more superficial tumor. The most commonly used laser probes are noncontact. The optimal spot size and tissue heating are achieved by a focal distance of about 10 mm. Noncontact probes generally allow the most efficient application. In very tortuous or narrow strictures, however, unintended probe contact can result in the need for repeated cleaning or even probe replacement.

Contact laser tips overcome this problem. Contact tips can be used to cut tumor tissue. [19], [20] In general, they require less power (12 to 20 W) and produce less smoke, but operators often find application less efficient than with noncontact probes.

Chemical Injection Small series have reported control of malignant obstruction and bleeding through endoscopic injection of agents that result in chemical necrosis. Relief of obstruction occurs by tissue sloughing, and hemorrhage is controlled by thrombosis, edema, and vasoconstriction. The technique is inexpensive and easily applied, and the necessary equipment and agents are widely available. Despite this, injection has not become popular, probably because of absence of an immediate tissue destructive effect that would guide and limit further therapy. Effective agents include sclerosants such as ethanol, sodium morrhuate, and polidocanol and chemotherapeutic drugs such as 5-fluorouracil.

Argon Plasma Coagulation Argon plasma coagulation (APC) is a noncontact thermal coagulation method in which current is applied by high-frequency electric current passed through argon gas. When the argon gas is passing current, an electrically insulating steam layer develops as soon as fluids in the tissue reach boiling point. This increases electrical resistance and can limit the depth of coagulation. [21] However, in experimental models, APC can readily produce transmural injury. [22] Depth of injury is a function of duration of application and secondarily of power setting. [22] In the cecum and right colon, power settings of 40 to 45 W and short bursts are most appropriate. In the rectum and stomach, settings of 60 to 75 W and more prolonged application are appropriate for benign tumor ablation. For palliation of cancers, power settings of 90 to 100 W and repeated application result in overt cavitation and marked tissue charring. APC has been used successfully in Europe and Canada for thermal hemostasis of gastrointestinal angiomata, [23] to treat gastrointestinal tumors, to treat tumor ingrowth or overgrowth, and to treat residual flat neoplasia after snare resection. [24], [25] and [26] It is effective in the treatment of radiation proctitis [27] and has been used to eradicate Barrett esophagus in combination with antisecretory therapy. [28]

Photodynamic Therapy PDT involves the use of a photosensitizer administered before photoradiation [29], [30] (Fig. 149-7). To activate the photosensitizer, photoradiation using a light of appropriate wavelength, good tissue penetration, and optimal absorption by the photosensitizer is applied using an optical fiber. The photosensitizer then destroys the dysplastic cells to which it is bound by production of cytotoxic singlet oxygen species.

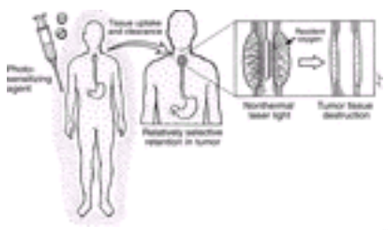

FIGURE 149-7. Method of photodynamic therapy (left to right). A light-sensitive drug is administered and accumulates selectively in the tumor. After a certain time, photoactivation of the drug by illumination with laser light initiates the photodynamic reaction, which kills the cells containing the specific drug.

Porfimer sodium (Photofrin), the only photosensitizer currently approved by the U.S. Food and Drug Administration, is indicated for palliation of patients with esophageal cancer. After an intravenous injection of porfimer sodium (2 mg/kg), clearance from a variety of tissues occurs over 40 to 72 hours, but tumors, skin, and organs of the reticuloendothelial system retain the drug longer. Illumination with 630-nm wavelength laser light initiates free radical production after porfimer sodium absorbs light to form a porphyrin-excited state. Spin transfer from porfimer sodium to molecular oxygen may then generate singlet oxygen (Fig. 149-8). Subsequent reactions can form superoxide and hydroxyl radicals, with subsequent cell death. Tumor death also occurs through ischemic necrosis secondary to vascular occlusion partly mediated by thromboxane A_2 release.

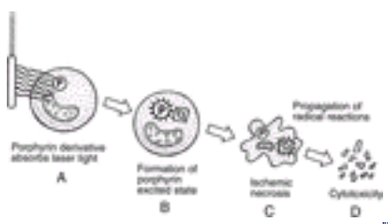

FIGURE 149-8. Photodynamic therapy: mechanism of action. Light-sensitive drug (*P*, porphyrin derivative) absorbs laser light (**A**). Energy is transferred from the excited triplet state of the sensitizer to molecular oxygen (**B**) to generate singlet oxygen (**C**), which oxidizes cell membranes; this results in ischemic necrosis and cell death (**D**).

PDT is contraindicated in pulmonary esophageal fistula and in patients with porphyria. Patients should avoid exposure of the skin and eyes to direct sunlight or bright indoor light, including unshaded light bulbs at close proximity, for 1 month or more. Some patients require a second laser light treatment 96 to 120 hours after the initial injection. PDT using 5-aminolevulinic acid (5-ALA) has been used in Canada and Europe for gastrointestinal cancers with promising results. [31], [32] It is administered orally, and the associated photosensitivity reaction is short lived and minimal. 5-ALA appears most useful for the treatment of very thin dysplastic or malignant lesions. The laser system used for PDT must be approved for delivery of a stable output (for porfimer sodium and 5-ALA at a wavelength of 630 nm). Light is delivered to the tumor by a cylindrical fiberoptic diffuser passed through the operating channel of the scope. Photoreactivation of the drug is controlled by the total light dose delivered. In the treatment of esophageal cancer, a light dose of 300 joules/cm of tumor length should be delivered. The cylindrical fiberoptic diffusers (OPTIGUIDE, Diomed, Andover, MA) are available in several lengths. The total power output at the fiber tip is set to deliver the appropriate light dose using exposure times of 12 minutes and 30 seconds. The major limitation of PDT is its cost, which is substantial both for the equipment and for the photosensitizers. New photosensitizers with more selectivity for the dysplastic and cancer cells and with fewer side effects will enhance the safety and effectiveness of the treatment.

Other Methods Palliative modalities that produce displacement (dilation and stent placement) are covered in Chapter 145.

ENDOSCOPIC TISSUE STAINING AND TATTOOING

Tissue staining and tattooing are adjuncts to the endoscopic therapy of polyps and tumors. *Tissue staining* refers to the topical application of chemical stains or pigment to alter tissue appearances in order to improve localization, characterization, or diagnosis.

Absorptive or vital stains identify specific epithelial types or cellular constituents by preferential entry. Contrast stains highlight tissue topography by entering mucosal depressions and crevices, a process called *chromoscopy*. Lugol solution is an iodine-based absorptive stain with affinity for glycogen in nonkeratinized squamous epithelium. When applied to normal esophagus, it turns a green-brown color within moments of application. Absence of staining indicates diminished or absent glycogen content, as seen in squamous cell cancer and dysplasia, [33], [34] Barrett epithelium, [35] and occasionally inflammatory esophagitis. Typically, 20 to 50 mL of a 1% to 3% Lugol solution is sprayed onto the mucosa during endoscopy.

Methylene blue enters the cytoplasm of absorptive tissues such as colonic and small bowel epithelia. Staining in the stomach and esophagus identifies intestinal metaplasia, [36], [37] whereas the absence of staining in the duodenum or colon usually indicates metaplastic, [38] neoplastic, [39] or inflammatory change. [40] After washing the mucous layer with 10% *N*-acetylcysteine or other mucolytics, a 0.05% to 1% solution of methylene blue is sprayed onto the mucosa. A few drops of methylene blue added to saline for submucosal injection during mucosectomy will sharply delineate the extent of the saline cushion as it develops and will sharply delineate the margin of flat colon polyps. Its use during colon polyp removal eliminates the need for chromoscopy to define the edges of the polyp.

Indigo carmine is a blue stain used for chromoscopy. [41], [42] It is sprayed as a 0.1% to 0.8% solution or ingested as a capsule. Chromoscopy using indigo carmine dye has been used to identify or enhance flat adenomas and can distinguish hyperplastic from neoplastic lesions, by observing the innominate grooves and pit pattern. [16], [43]

Tattooing is a means of labeling a site by intramural injection of a pigment for future identification during surgery or repeat endoscopy. India ink is particularly useful because it produces a black stain visible permanently from serosal and lumenal surfaces. [44], [45] India ink is a suspension of inert carbon particles in a variety of aqueous and nonaqueous stabilizers and diluents. Pure India ink is preferable because inclusion of other materials may evoke an immunologic response, although toxicity is exceedingly rare. A 1:10 or 1:100 ink-to-saline dilution can be autoclaved or filtered, [46] and commercial preparations ready-made for endoscopic injection are available. The tattoo is made by injections of 0.1 to 0.5 mL using a sclerotherapy needle inserted tangentially to make a submucosal bleb. Injection into four quadrants ensures easy intraoperative identification.

GOALS OF ENDOSCOPIC THERAPY

Tumors in the gastrointestinal tract can be approached with the objectives of curative resection or palliation of symptoms. These same general goals are applicable to endoscopic treatment of polyps and tumors. Palliation becomes the goal when staging of a malignant tumor shows it to be incurable or when a tumor is present that cannot be completely resected endoscopically, and when the patient is not a candidate for or is unwilling to undergo other types of therapy.

RATIONALE FOR ENDOSCOPIC THERAPY VERSUS OTHER MODALITIES

Optimal features of curative resection include a minimally invasive approach, low rate of complications, outpatient procedure (minimal need for hospitalization), limited or no need for sedation or anesthesia, minimal recovery time, recovery of the surgical specimen, and minimal need for repeated examinations or treatments. Compared with surgery, endoscopic therapy is generally superior in each regard, except the recovery of the specimen (occasional failures with endoscopic therapy) and the need for repeated examinations or treatments.

The objective of palliative therapy is usually maintenance of lumenal patency and nutrition or cessation of bleeding. The optimal features of palliative therapy are similar to those of curative resection, except that recovery of a specimen is less important. Endoscopic therapy is generally better tolerated than surgical therapy for palliation, except that it may necessitate multiple treatments.

PRECURSOR LESIONS

Most gastrointestinal malignancies arise from the mucosal lining. In some cases, a sequence of dysplasia progressing to cancer is documented. In the colon, this progression involves a benign growth (the adenomatous polyp) in nearly all cases of colorectal cancer arising in the United States and Europe. A careful endoscopic examination of the colon can identify these lesions and remove them with nearly complete protection from colorectal cancer mortality. Adenomatous polyps also arise in the stomach and duodenum. An adenoma-carcinoma sequence is less well established in these areas, particularly the stomach. Nevertheless, the association of adenomas with cancer in these sites is clear, and a general principle throughout the gastrointestinal tract is that endoscopically identifiable growths that are benign and dysplastic (adenomatous polyps) should be removed to prevent progression to cancer. Concepts regarding precursor lesions are critical to the endoscopist because endoscopy is superior to all other technologies in both their detection and removal.

A number of mucosal cancers in the gastrointestinal tract are not associated with an identifiable precursor that is appreciable by routine endoscopy. Examples include some colorectal cancers in ulcerative colitis and Crohn's colitis, and squamous cell and adenocarcinomas of the esophagus. A progressively important focus of endoscopic research is to identify dysplastic, flat mucosa and treat the area endoscopically or by surgical resection before carcinoma develops or spreads to an incurable stage. In some cases (e.g., Barrett esophagus and chronic ulcerative colitis), the area at risk is endoscopically apparent and can be subjected to systematic sampling using endoscopic biopsy forceps. Several methods are under investigation to allow sampling that is more directed to areas of possible dysplasia. One approach is to use dye spraying (chromoscopy) or staining. Areas of dysplasia can then be subjected to mucosectomy or surgical resection (stomach or esophagus), ablative therapy (esophagus), or regular intensive surveillance biopsies to rule out cancer (esophagus). Other approaches that can help direct biopsy to flat precursor lesions are high-magnification endoscopy [43] and light-induced autofluorescence. [17]

Japanese investigators have accumulated an extensive experience with chromoscopy, sometimes in combination with high-magnification endoscopy, to enhance detection of small, flat dysplastic colonic lesions, often referred to as *flat adenomas* and *flat cancers*. These lesions appear endoscopically as slightly raised, flat, or even depressed lesions, sometimes with an erythematous halo. Although an exact definition is not possible, they are usually defined as being less than 1 to 1.5 cm in size, and the total histological height of the lesions should not exceed twice that of the normal mucosa. Flat adenomas are said to have an increased probability of severe dysplasia relative to their size, although Japanese pathologists have been shown to vary widely in their interpretations of severe dysplasia. [47] Similarly, it is clear that lesions determined by Japanese pathologists to be cancer lesions would be termed high-grade dysplasia in the West. [48] However, a single pathologist who reviewed the histology of flat adenomas from both Tokyo and Stockholm found that flat adenomas in Japan are more likely than those in the West to contain high-grade dysplasia or invasive cancer. [49]

The prevalence of flat adenomas and cancer is relatively low, even in Japan. [42], [50] In the West, one colonoscopist found that 12% of adenomas met histological criteria for flat adenomas, but none had severe dysplasia or cancer. [51] In an autopsy study in Vancouver, 29 flat adenomas were found, of which 41% had high-grade dysplasia. [52] Three prospective studies have evaluated patients in Britain [53], [54] or the United States [55] using Japanese colonoscopic techniques to investigate the issue of flat colonic neoplasms. This technique in essence is to scan the colonic mucosa for subtle surface irregularities or color changes and then to spray dye, usually indigo carmine, on any such abnormalities. Rembacken and associates [53] studied 1000 British patients and found four very small depressed lesions, three of which had Dukes A cancer or high-grade dysplasia. It is uncertain whether these lesions would have been detected without chromoscopy. In another study, five small flat cancers were identified in 870 British patients. [54] These new data must be viewed in light of the National Polyp Study, in which conventional colonoscopy and resection of all polypoid lesions was essentially completely effective in preventing colorectal cancer mortality. [56] However, in the National Polyp Study, as well as in other large postpolypectomy studies, colonoscopy is not fully protective against development of incident cancers. [56] It is possible that failure to detect flat and depressed neoplasms accounts for some incident cancers after apparent clearing colonoscopy. [57]

PATIENT SELECTION FOR ENDOSCOPIC TREATMENT

When considering an attempt at curative endoscopic resection, the endoscopist first considers the likelihood that a lesion is malignant. Irregular surface contour, large size, ulceration, and firmness of the lesion with probing are all indications of malignancy. Careful staging is indicated before any attempt at resection of a lesion with these features because surgical resection offers the best chance for cure of intestinal cancers. In uncertain cases, submucosal injection of saline may be helpful because fixation of the submucosa by tumor will prevent elevation of the lesion, and endoscopic resection can be aborted. Injections should not be made directly through lesions that appear possibly malignant. In general, lesions involving the mucosa are resectable for cure by endoscopic methods, as are some lesions extending into the submucosa or involving only the submucosa (e.g., carcinoids). In the esophagus, which lacks a serosa, even superficial cancers are at times associated with lymph node metastasis. Factors such as surgical candidacy and the feasibility of endoscopic removal (size of the lesion) can influence the decision of an endoscopic attempt versus surgery. Similarly, transanal surgical resection of a rectal lesion may at times be preferable for a large benign rectal growth because the distal rectum is easily accessible to transanal surgery, and endoscopic resection or ablation, although feasible, might require many sessions. Lesions extending into the muscularis propria are seldom treated in a curative fashion by endoscopy.

At times, the need for surgical treatment is not evident until after endoscopic resection with curative intent. This need is best established for malignant pedunculated colon polyps, in which the presence of cancer at the histological resection line of an endoscopically removed polyp, poorly differentiated cancer, or vascular (lymphatic) invasion are predictors of metastasis and serve as a general guide indicating the need for segmental colon resection. In some centers, any cancer in a sessile colon polyp is an indication for surgical resection in a good surgical candidate.

Staging should generally be accomplished by computed tomography (CT) scan and endoscopic ultrasound (EUS). EUS is more accurate for T and N staging, and CT scan is more accurate for distant metastases. Patients with unresectable disease may be candidates for endoscopic palliation of symptoms, provided they have reasonable life expectancy (perhaps greater than 1 month).

FAILURE OF ENDOSCOPIC THERAPY

Curative endoscopic resection fails when a lesion is too large for removal, when a lesion recurs despite attempts at resection, when complications occur, when a lesion contains invasive cancer requiring resection, when a patient is lost to follow-up before completion of endoscopic resection, or when metastasis occurs after what was thought to be complete endoscopic removal. The likelihood of endoscopic failure should be carefully considered in cases in which both endoscopic and surgical resections are reasonable alternatives. Conversely, the patient is poorly served when surgery is performed unnecessarily to remove a section of the intestinal tract containing an endoscopically removable lesion.

ESOPHAGEAL LESIONS

For patients with localized esophageal cancer, surgical resection with or without preoperative chemoradiation is the preferred approach. Many patients with esophageal cancer are elderly and may not be good operative candidates even if staging demonstrates localized disease. Patients with T1 or T2 disease (TNM staging system) who are not surgical candidates or who refuse surgery may undergo an attempt at curative therapy by endoscopic methods such as PDT, [58] mucosectomy, [59] or both. Patients whose disease is not resectable for cure and those who are not operative candidates should be offered palliative therapy if their life expectancy is greater than 1 month. Because tumor stage and operative candidacy guide therapy, accurate staging is critical. Local staging and involvement of lymph nodes is determined by EUS, which is generally indicated when CT fails to demonstrate distant metastases. Palliation is directed toward maintenance of lumenal patency so that swallowing can continue. Palliative therapy does not generally lead to substantial prolongation of survival. Modalities for the palliation of esophageal cancer include surgery, radiotherapy, chemotherapy, and endoscopic therapy. Endoscopic palliative treatments include dilation, thermal modalities (Nd:YAG laser, BEC, argon coagulation), nonthermal ablation using PDT, endoscopic endoprosthesis, and intratumor chemical injections.

Screening programs for squamous cell carcinoma have used Lugol iodine solution to identify early cancers and flat precancerous lesions. Endoscopic mucosal resection with a cap-fitted endoscope has been performed after submucosal injection with saline and epinephrine solution. [59]

Endoscopically delivered Nd:YAG laser therapy may be used as primary palliation of dysphagia or to establish patency and improve nutritional status before initiating radiation or chemotherapy (Table 149-2). Favorable factors for successful laser therapy include exophytic tumor, tumor location in a straight segment of the middle or distal esophagus, and tumor less than 5 cm in length. Patients with noncircumferential tumors may be treated more effectively with Nd:YAG laser than with stent placement. Patients with tumors in the cervical esophagus or with submucosal tumors are poor candidates for laser therapy. A major disadvantage of Nd:YAG laser therapy is the need for repeated treatments at 4- to 8-week intervals. [60], [61], [62], [63], [64], [65], [66] and [67] Retrograde application is preferred, [66] and power settings of 90 to 100 W are commonly used.

TUMOR CHARACTERISTICS	STENT	LASER	BIPOLAR	PDT
Circumferential cancer	Yes	Yes	Yes	Yes
Noncircumferential cancer	Maybe	Yes	No	Yes
Exophytic	Yes	Yes	Yes	Yes
Intramural	Yes	No	Yes	Maybe
Cervical esophagus	No	Maybe	Yes	Yes
Gastroesophageal junction tumor	Maybe	Maybe	Yes	Yes
Angulated stricture	Yes	Maybe	No	Yes
Tracheoesophageal fistula	Yes	No	No	No
Recurrence after radiation	Maybe	Maybe	Maybe	Yes

PDT, photodynamic therapy

TABLE 149-2 Selection of Palliative Procedures for Esophageal Cancer

Contact probes [19] produce less smoke than noncontact methods and eliminate the concern about fiber tip damage during the procedure, although in a randomized comparison, the two fiber types had equal performance. [20] Nd:YAG laser produced fewer complications than plastic stents in the treatment of esophageal cancer, [68], [69] but the need for repeated treatments has led to replacement of Nd:YAG by self-expanding metal stents.

The bipolar tumor probe is now used infrequently for palliation of esophageal malignancy. [70], [71] and [72] The principal advantages are the relatively low cost and widespread availability of the power unit. After sufficient dilation, the probe is passed to the distal end of the tumor. [70] Probes with electrodes oriented circumferentially are used most frequently. Partly circumferential probes are available for treatment of partly circumferential tumors and must be maintained in correct radial orientation by endoscopic observation.

APC has been used in palliative tumor ablation, in stent overgrowth and ingrowth, and for treatment of tumors after laser therapy. [24], [73], [74] Sessler and associates [25] used APC in eight patients with T1 tumors of the esophagus who were not candidates for surgical resection. Follow-up for 9.45 ± 2.8 months showed no tumor in seven of eight patients. In another study, APC was used as an adjunct to PDT in 11 patients with high-grade dysplasia and Barrett esophagus. Short-term follow-up revealed effective eradication. [73]

A prospective 12-year follow-up study of PDT for esophageal malignancy revealed that PDT caused minimal complications and no procedure-related deaths. The length of palliation for patients having noncurative treatment was equal to or better than that reported historically for most other treatment regimens. For patients with Barrett esophagus and severe dysplasia or T1 carcinoma who are high surgical risks, it can be considered an alternative treatment. [75] Overholt and Panjehpour [76] treated 100 patients with Barrett esophagus and dysplasia (13 with early superficial cancer) with PDT. Patients were maintained on long-term acid-suppressive therapy. Follow-up endoscopy demonstrated ablation of the superficial cancer in 10 of 13 malignancies, [66] elimination of dysplasia in 78 of 100 patients, reduction of the extent of Barrett esophagus in all patients, and eradication of Barrett esophagus in 43 patients. Laukka and Wang [77] used low-dose PDT (1.5 mg/kg) in patients with Barrett esophagus and revealed a mean reduction of 2.4 ± 0.9 cm in the length of columnar epithelium. PDT was also effective in treating tumor ingrowth and overgrowth of an esophageal stent. [78], [79] A multicenter randomized trial comparing PDT and thermal ablation with Nd:YAG laser for palliation of esophageal cancer revealed that both methods are equally effective for palliation of dysphagia and objective tumor response rate. However, PDT was easier to use and was associated with fewer acute perforations than Nd:YAG laser therapy. [30]

Endoscopic mucosal resection (EMR) of high-grade dysplasia and superficial esophageal cancer has emerged as a potential alternative treatment. Endoscopic treatment can be considered for esophageal cancer if it is at an early stage, is intramucosal or microinvasive (involving only the most superficial aspect of the submucosal), and has no lymph node involvement by endoscopic ultrasound. The tumor should generally be less than 2 cm in diameter, and the patient should be a suboptimal candidate for surgery. [80], [81], [82], [83] and [84] Yoshida and colleagues [83] observed a 12.9% rate of immediate complications, of which mediastinal emphysema (usually asymptomatic) and bleeding were the most common. They also observed a 7% of late complications that were mainly stenoses. Inoue [82] had two severe complications (one perforation and one stenosis resistant to dilation) in a series of 142 patients.

GASTRIC LESIONS

Gastric polyps are usually adenomas or hyperplastic polyps. Adenomas are markers of an increased risk for adenocarcinoma, but an adenoma-carcinoma sequence is not fully established in the stomach. Adenomas are predominantly solitary and antral in location. Their size appears to be of great importance in predicting malignant potential. In polyps larger than 2 cm, there was a 24% incidence of malignancy; whereas in the polyps smaller than 2 cm, there was only a 4% incidence. [85] Carcinoma arising in adenomas is usually of the intestinal, well-differentiated type. [86] Hyperplastic polyps are more common than adenomas, are randomly distributed throughout the stomach, and typically are less than 2 cm in diameter. Endoscopically, these lesions may have surface inflammatory erosion and exudate. Giant gastric hyperplastic polyps occasionally undergo malignant transformation. [87] Fundic gland polyps were unrecognized in early series and often called “hyperplastic.” They now account for up to 50% of gastric epithelial polyps. Fundic gland polyps occur within the body or the fundus and are multiple. There is no association between fundic gland polyps and atrophic gastritis or adenocarcinoma. [88] Because biopsy specimens obtained by forceps have been shown to be unreliable in determining the type of polyp present, [89] gastric polyps larger than 1 cm should be removed endoscopically if feasible. [90], [91]

Gastric polyps smaller than 5 mm can be removed using biopsy forceps. Gastric polyps larger than 5 mm should be removed by snare excision. When the resected specimen shows carcinoma invading the polyp base or if poorly differentiated carcinoma is present, surgical resection should be considered. Cancer developed in 1.3% of 1177 patients after resection of gastric adenomas, and yearly follow-up was recommended. [92]

The technique of gastric polypectomy is similar to that for colonic polypectomy (see later). Intravenous glucagon may reduce peristalsis. Bleeding is more common after gastric polypectomy [89], [91], [93] than after colonic polypectomy, but the risk for perforation is very low. Polyps should be retrieved in a manner providing airway protection. An overtube or a Roth retrieval net should be considered for polyps that cannot be suctioned through the endoscope. Acid-suppressive therapy may help heal the cautery ulcer produced by polypectomy.

Endoscopic resection of gastric submucosal lesions may be performed using strip biopsy, [94] aspiration mucosectomy, [95] or endoscopic mucosal resection with a cap-fitted endoscope. [59] Aspiration mucosectomy produces larger specimens that contain a greater depth of submucosa compared with strip biopsy. [95]

Early gastric cancer is defined as carcinoma limited to the gastric mucosa or submucosa regardless of lymph node involvement. [96], [97] and [98] In Japan, mass screening programs and the availability of widespread endoscopy methods have increased the fraction of gastric cancer diagnosed at the early stage from 5% to more than 30% in 15 years. [98] Unfortunately, early gastric cancer is diagnosed in the United States in only 3% to 6% of the cases. [99] Several large series described EMR for gastric superficial carcinoma. [100], [101], [102] and [103] Depending on the technique and the characteristics of the lesion, the complete resection rate ranges between 43% and 100%. In one series, complete resection was achieved in 43% of cases, which increased to 87% when it was combined with laser ablation.

A band ligation device can be used to convert flat neoplasms to polypoid lesions. [104], [105] The resulting polypoid lesion is resected below the elastic ring using conventional polypectomy snare technique. Other successful treatments for early gastric cancer include PDT [106], [107] and intratumor injection of OK-432, a drug produced by treating the Su strain of *Streptococcus pyogenes* with penicillin. [108] Thus, endoscopic "curative" therapy for early gastric cancer is feasible in patients who are poor candidates for surgery. EUS staging should precede any attempt at endoscopic resection.

Nd:YAG laser has been useful as a palliative treatment of advanced gastric cancer in patients who are poor surgical candidates. A mean of three treatment sessions of about 6000 W/second per session was required to establish a lumenal opening, and 73% of patients could eat all foods. [109]

DUODENAL LESIONS

Adenomas are the most common duodenal neoplasms. Large, villous adenomas have an increased risk for containing adenocarcinoma, [110] and resection is generally indicated. [93], [111] Endoscopic resection of lesions on the medial wall of the duodenum is facilitated by use of a duodenoscope (side-viewing endoscope). Periampullary lesions should be staged by EUS before resection. [112] Endoscopic therapy of periampullary tumors may be accompanied by pancreatitis or bleeding. Snare polypectomy, [112] strip biopsy, [113] EMR, [114] Nd:YAG laser, [115], [116] and PDT with 5-ALA [31] or Photofrin [117] have all been used to resect or ablate duodenal adenomas or early cancer.

COLORECTAL POLYPS AND TUMORS

The goal of endoscopic treatment of benign colorectal neoplasms is curative resection. The evidence to support the effectiveness of colonoscopy and polypectomy in the prevention of colorectal cancer incidence and mortality includes three adenoma cohort studies [56], [118], [119] with reductions in incidence compared with reference populations, a small randomized controlled clinical trial, [120] a case-control study, [121] case-control studies of sigmoidoscopy and polypectomy, [122], [123] and a randomized controlled trial in which fecal occult blood testing reduced the incidence of colorectal cancer by identifying large polyps that were then removed by colonoscopic polypectomy. [124]

The standard therapy of colorectal cancer is surgical. However, the mean age of colorectal cancer diagnosis is 70 years, and incidence continues to rise with age. [125] If operative risk is substantial owing to age or co-morbid illness, endoscopic therapy may be preferable. Many patients with metastatic disease benefit from operative resection or diversion to avoid colonic obstruction. However, patients with widely metastatic disease and short life expectancy may opt for endoscopic palliation, as may those who are unwilling to have a stoma.

Pedunculated Polyps

Pedunculated (stalked) polyps are more common in the left colon than the right. They may be positioned in the sigmoid colon in areas of marked angulation. Repositioning the patient may improve exposure of the polyp head and stalk. A skilled endoscopist should be able to remove essentially all pedunculated colon polyps. Ensnaring a very large polyp is often facilitated by the use of an extra-large (jumbo) snare. The closed snare should be positioned near the polyp head or at least two thirds of the distance from the base of the stalk to the polyp head. This leaves residual stalk that can be snared if there is immediate bleeding. On the other hand, some have argued that if the polyp looks as though it may be malignant, it is preferable to close on the base of the stalk [126] in order to improve the chance of a clear resection margin. If the tip of the polyp head touches the contralateral colon wall, current may pass through that wall and produce a burn (Fig. 149-9). Gentle movement of the snare during application of cautery will dissipate this burn on the contralateral wall and eliminate the risk for perforation. Most experts hold the colonoscope shaft in position with their right hand during application of cautery, or pin the scope against the bed with their hip to prevent scope movement if manipulation of the snare during cautery is needed. An assistant generally closes the snare. If, before passing the snare down the instrument, a mark is placed on the snare handle at the point of closure where the snare tip just enters the plastic sheath, then the distance from the mark to the closing portion of the snare provides an estimate of the amount of tissue in the snare. If this distance is excessive (more than 1 to 1.5 cm), then careful inspection or remanipulation of the snare is prudent to ensure that no portion of the normal wall or the polyp head is in the snare.

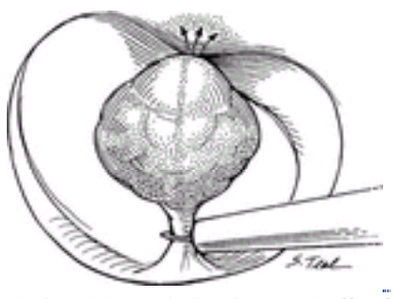

FIGURE 149-9. Current dissipation through the contralateral colonic wall. The electrosurgical snare is around the stalk of a pedunculated polyp. The polyp head is in contact with the contralateral wall. Leaking current during cautery may cause thermal injury to the contralateral wall (*arrows*). Significant injury is easily averted by moving the polyp head during application of electrocautery whenever the polyp head touches the contralateral wall.

The risk for perforation with pedunculated polyps is exceedingly low, so that application of current can usually be liberal and mechanical closure slow. A white coagulum should be endoscopically visible adjacent to the snare before mechanical closure begins. Current should be applied continuously because intermittent application allows cooling of the snare and may seal it to the tissue ("stuck snare").

The most common complication of endoscopic transection of pedunculated polyps is bleeding, which can be either immediate or delayed. Some experts preinject the stalk with epinephrine and saline. However, the risk for immediate bleeding overall is low, particularly if coagulation current is used. Thus, the use of an injection catheter and epinephrine routinely for colon polyps is not cost-effective. A time-honored approach to immediate hemorrhage is to regrasp the stalk with the snare and hold the stalk for 10 to 15 minutes. No additional cautery is applied, yet this method is consistently effective. Alternatively, epinephrine can be injected into the stalk if immediate bleeding occurs, followed by bipolar coagulation of the transection site on the stalk. A recent innovation, proved effective in a randomized trial, [127] that appears to prevent both immediate and delayed bleeding is a detachable snare, which is placed on the polyp stalk before transection (Fig. 149-10). The snare subsequently sloughs up to 1 week later. The detachable snare can also be placed on the stalk after transection. Attempted application to semi-pedunculated polyps before or after transection is often unsuccessful. An alternative is to place a metal clip across the base of the stalk.

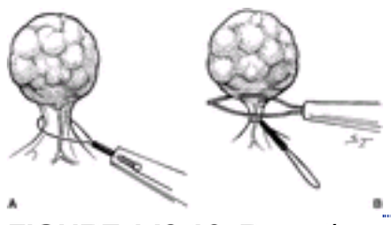

FIGURE 149-10. Resection of a pedunculated polyp with a detachable snare. **A:** The loop of a detachable snare is placed around the stalk of the polyp, tightened, and then detached from the handle. **B:** A cautery snare is placed above the loop and transects the stalk by electrocoagulation.

More than 95% of colon polyps can be successfully retrieved. Small pedunculated polyps can be suctioned through the colonoscope into a trap. If the polyp is not visible in the trap after suctioning, it is usually impacted in the umbilical cord of the instrument. Water can be suctioned from a syringe inserted into the suction or biopsy inlet of the instrument through the umbilical cord to flush the polyp into the trap. Larger pedunculated polyps are most easily retrieved by gently regrasping the transected polyp head. If 2 to 3 cm of the snare is left outside the colonoscope tip, then examination can continue while the polyp is being delivered. If additional small polyps are encountered, the large polyp is dropped in the lumen, the small polyps are removed and suctioned, the large polyp is regrasped, and the examination then continues. Combined cautery and retrieval snares have been recently described. [128]

Large pedunculated colon lesions with overlying normal mucosa are usually lipomas. The diagnosis is confirmed by the “cushion” or “pillow” sign of easy deformability with probing; by the yellow hue, which is sometimes present; and if necessary, by EUS. These tumors are usually asymptomatic but can produce intussusception. Endoscopic snare resection is often unsuccessful. Fat has low water content and resists the desiccation necessary for easy mechanical transection.

Endoscopic resection of a malignant pedunculated polyp is considered curative if (1) the cancer is well or moderately differentiated, (2) there is no vascular (lymphatic) invasion, and (3) the margin is clear. If these criteria are met, surgical resection is not advised. If the criteria are not met, the risk for metastasis is increased, with the extent of increase depending on the number and severity of the changes. [129], [130] and [131] If the estimated risk of operation is lower than the risk for metastasis, then surgical resection should be considered.

Diminutive Polyps

Polyps less than or equal to 5 mm in size are diminutive. Most of these polyps in the proximal colon are adenomas, and most in the rectosigmoid are hyperplastic. The concept of “clearing” colonoscopy includes removal of these polyps, although they rarely contain cancer. In some instances, they are so frequent in the rectosigmoid that only representative biopsy sampling is feasible. In these instances, biopsies generally demonstrate hyperplastic polyps. Whether it is cost-effective to remove diminutive polyps and send them for histology has not been evaluated.

Cold biopsy, cold snaring, hot forceps biopsy, and snare cautery are all acceptable approaches to diminutive polyps. Diminutive snares are almost as easy to use as forceps, [2] although flat 1- to 2-mm polyps can still reliably be grasped only with forceps. Snare cautery is the most reliable of the four methods for completely removing small polyps. Hot forceps are five times more likely to produce a complication in the right colon as in the left [9] (Fig. 149-11). Cold methods appear to be the safest. [4]

FIGURE 149-11. Hot biopsy forceps. **A:** Diminutive polyp. **B:** The forceps grasps the polyp (or even just the polyp tip), and the polyp is pulled way from the bowel wall as cautery is applied. Failure to “tent” the polyp by pulling it away from the wall may allow the burn to penetrate the wall deeply. **C:** Other errors with hot forceps include grasping too much tissue (i.e., including normal tissue) or tenting the polyp too much in one direction, which may cause the cautery burn to spread selectively down one side of the polyp.

Sessile Polyps

Sessile polyps larger than 5 mm are typically removed by snare cautery. Lesions smaller than 1.5 cm should be removed in a single piece if possible. Larger polyps may require removal in pieces. [132], [133] The snared polyp or portion of polyp is tented into the lumen to create an artificial stalk. In the right colon, lumenal deflation will thicken the wall before application of cautery.

Large sessile polyps that demonstrate surface irregularity or ulceration are usually malignant; biopsy should generally be performed to rule out cancer before attempting endoscopic resection. The rationale for biopsy is that sessile malignant lesions are a general indication for surgical resection. However, some experts consider endoscopic resection to be definitive therapy for sessile malignant polyps if they exhibit favorable histological criteria. [4] In this case, a sessile polyp with some malignant features could be resected endoscopically if it could be lifted on a submucosal saline mound, indicating that no submucosal fixation is present. In general, lesions covering more than one third of the colon circumference or having a longitudinal extent of more than two haustral folds should be considered for surgical resection as primary therapy. However, in the right colon and rectum where the lumenal diameter is relatively large, much larger sessile lesions may be amenable to endoscopic resection.

Submucosal saline injection should be considered for large or flat sessile polyps. Injection begins at the proximal edge or, for smaller polyps, is made directly into the polyp center. Only injection into the submucosa will raise a mound. Injection has not been associated with a risk for tumor seeding or peritonitis, although clearly, the needle may pass through the colon wall. During injection polypectomy, the snare may close over the saline mound without grasping tissue. This effect can be countered by barbed snares (Olympus Corp, Lake Success, NY) or needle-tipped snares (Wilson-Cook, Winston-Salem, NC) that hold the snare in place. Alternatively, a circumferential incision cut with electrocautery in the saline mound but outside the polyp forms a groove that seats the snare in position around the polyp. [134]

In some cases, sessile adenomas are so flat (so little elevated aspect) that the lesions are unsnarable even after submucosal injection. Potentially effective treatments in this instance include ablative therapy (see later) and EMRC. EMRC has the advantage of retrieving a histological specimen. In the colon, EMRC use is best confined to the thicker-walled left colon and rectum. EMRC is performed only after submucosal saline injection, and volumes of at least 30 to 40 mL are anecdotally important to reduce the risk for perforation associated with aspiration of deep wall layers into the cap. In our experience with an American population, EMRC is needed much less frequently in the colon than the stomach and especially toward the esophagus.

Visualization of the margin of very flat polyps can be facilitated by dye spraying with methylene blue, indigo carmine, or cresol violet, or by addition of small amounts of methylene blue to the saline before injection (see earlier). Sections that are flat and that cannot be removed by saline injection can be destroyed using Nd:YAG laser, BEC, or APC. The latter two techniques are preferred because they have a lower chance of perforation.

Endoscopic localization for possible subsequent surgery is not reliable unless the ileocecal valve is in view or the polyp is in the rectum or distal sigmoid. Between these sites, large polyps should be marked by tattooing with India ink.

If a sessile polyp is removed in piecemeal fashion and the fragments are too large to be retrieved by suctioning, retrieval can be facilitated by the Roth retrieval basket. This device consists of a net on a large snare, which is repeatedly opened and closed on the fragments until all are trapped in the net.

Histological criteria to assess the need for surgical resection are less clearly applicable to sessile malignant polyps. In a review of nine studies reporting the frequency of cancer at surgery or during follow-up after endoscopic resection of a sessile malignant adenoma, polyps with favorable histology had a 4.1% incidence of residual cancer, compared with 20.6% in polyps with unfavorable histology. [135] In another study, the incidence of metastasis from sessile malignant polyps was 15%. [136] Thus,

operative resection should be considered in any sessile malignant polyp because the risk for metastasis is several times that of pedunculated polyps with similar histological criteria.

Endoscopic resection of sessile polyps carries a risk for perforation, postpolypectomy syndrome, and immediate and delayed bleeding. Perforation should be managed operatively in most cases, particularly if it is recognized within the first few hours after resection. Perforations presenting later with well-localized peritoneal signs can sometimes be managed nonoperatively. [137] Postpolypectomy syndrome is the result of a serosal (transmural) burn without free perforation. Patients present with localized pain and tenderness, fever, and elevated white blood cell count, but no free air on abdominal radiographs. In some instances, a water-soluble contrast enema or CT scan may be necessary to rule out perforation. Treatment is bowel rest, antibiotics, and careful observation. Immediate bleeding is best managed by injection of epinephrine (1:10,000 in normal saline) into the polypectomy base or simply spraying the solution onto the polyp base. This can be followed by BEC or heater-probe treatment of the bleeding site. Monopolar cautery should not be reapplied. Most delayed bleeding stops spontaneously. If delayed bleeding remains active, repeat colonoscopy with epinephrine injection and bipolar cautery or heater probe treatment of the base is effective. [138] Application of metal clips is an inexpensive and effective alternative for both immediate and delayed bleeding from sessile polypectomy sites.

Recurrence rates of large sessile adenomas after endoscopic piecemeal polypectomy vary from 16.5% to 50%. [131], [139] Nearly one half of recurrences occur after one negative follow-up examination, and recurrences may develop more than 1 year after the initial resection. [139] It should be recalled that most current guidelines for postpolypectomy surveillance [140], [141] are derived primarily from the National Polyp Study. [142] However, sessile adenomas greater than or equal to 3 cm in size were excluded from the National Polyp Study, [142] and thus the conclusions derived from that study do not apply to these large polyps. Most recurrences can be successfully treated by additional endoscopic therapy. [139] Repeat examinations at 3- to 6-month intervals are indicated after piecemeal resection of large sessile adenomas. [139] After complete resection appears verified, another examination at 1 year is prudent. [139]

Several groups have reported experience with Nd:YAG laser ablation of large rectal polyps. [143], [144] and [145] A disadvantage is that tissue is obliterated and not available for histological evaluation, so that late presentation of metastatic cancer is possible. The overall success rate depends on the polyp size, with success rates as low as 56% for lesions occupying two thirds of the circumference, [145] and as many as 10 treatment sessions required. Complications include bleeding, rectal stenosis, and fever. Perforation is rare owing to the retroperitoneal location of much of the rectum. Snare cautery debridement before Nd:YAG laser therapy is superior to Nd:YAG therapy alone. [146], [147] Transanal or transsacral resection should also be considered, although recurrence rates of 6% to 42% and complications in 13% follow the transanal approach, and the transsacral approach is followed by fistula formation in up to 21% of patients. [148], [149] and [150]

PDT was reported to eradicate sessile villous adenomas in the rectosigmoid that had failed to respond to Nd:YAG laser. [151] PDT has been reported to debulk large sessile rectal adenomas before Nd:YAG therapy. [152] Debulking by snare excision, however, is the preferred approach. [146], [147]

When lesions compatible with flat adenomas or cancers are identified, mucosectomy is the preferred approach because it allows clarification that the submucosa is not fixed and gives an ideal histological specimen for ascertainment of tumor-free margins. [153], [154], [155] and [156]

Endoscopic Treatment of Colorectal Cancers

The treatment of choice for cure or palliation of colorectal cancer is surgical resection. Less than 5% of patients are not candidates for surgery because of widely metastatic disease, inability to tolerate surgery, or unwillingness to undergo surgery or ostomy. In these instances, management depends on local expertise. However, Nd:YAG laser, PDT, and placement of a colonic stent may all be considerations, depending on symptoms. The Nd:YAG laser is effective for relief of bleeding, obstruction, or both. [157], [158], [159], [160], [161], [162], [163] and [164] In rare instances in which surgery cannot be performed, Nd:YAG laser can be used to treat T1 or T2 lesions staged as N0, M0 with curative intent. [162], [163] and [164]

PDT was used for 21 patients with inoperable or recurrent colorectal cancer, and 16 had evidence of necrosis, 15 had increased lumenal diameter, and 10 had relief of obstructive symptoms. [165] However, the ease with which colonic stents can be placed and the need for repeated treatments with ablative therapies has made stent placement the treatment of choice for palliation of obstructing colorectal cancer.

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CHAPTER 150

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EVALUATION OF GASTROINTESTINAL MOTILITY: METHODOLOGIC CONSIDERATIONS

Abnormalities of gastrointestinal (GI) motor function contribute directly or indirectly to a number of common clinical problems. Advances in monitoring techniques allow relatively noninvasive recording of GI motility under physiologic conditions. This chapter emphasizes the developments in the application of esophageal manometry, electrogastrography (EGG), gastroduodenal manometry, electronic barostat, and anorectal manometry to evaluate GI motility.

ESOPHAGEAL MANOMETRY AND 24-HOUR pH MONITORING

Esophageal manometry and 24-hour pH testing are accepted clinical and investigative tools for the study of esophageal dysfunction. [1],[2] These procedures are most frequently performed in evaluating patients with dysphagia who have no evidence of mechanical obstruction and in patients with suspected noncardiac chest pain or gastroesophageal reflux. Generalized motor disorders of the GI tract, such as scleroderma and chronic intestinal pseudoobstruction, can affect the esophagus. Confirmation of the diagnosis frequently can be made by esophageal manometry.

Technical Aspects of Esophageal Manometry

Equipment Esophageal motility testing requires intubation of the esophagus with a recording catheter, which is connected to a physiograph. The clinician must choose between two major systems of recording catheters available: water perfused or solid state. The standard system in most esophageal motility laboratories uses a water-perfused catheter system connected indirectly to a physiograph through a series of transducers. [3],[4] Pressure is recorded at predetermined recording orifices; the signal from each orifice is changed into electrical current at external transducers and retransformed into a pen deflection at the physiograph. The major advantages of this system are the initial comparatively low cost, rapid repair of malfunctioning external transducers, and extensive experience with the system. The major disadvantage of the water-perfused system is variability in recording accuracy when measuring transient high-pressure events such as upper esophageal sphincter (UES) pressure and pharyngeal UES coordination. This disadvantage can be overcome by using low-compliance microcapillary tubing and perfusing with a low-friction pneumohydraulic pump, which allows recording of all intraesophageal events at acceptable perfusion rates. [5] Other disadvantages are the constant need for recalibration and the possible need for a separate pH recording system. The second system is a solid-state recording catheter with multiple, small-diameter intralumenal transducers that directly interface with the physiograph. The major advantages of the solid-state system are ease of use, minimal need for calibration, accuracy of recording, and the capability of performing simultaneous motility and pH recordings. Major disadvantages of the intralumenal transducer system are the high initial cost and the potential downtime for the system if one or more transducers are damaged. The major cause of transducer breakdown is transducer membrane breakdown with acid exposure. Another disadvantage is the fixed nature of the transducer position; solid-state recorders can be made to order, albeit at greater expense. Both systems can measure intraesophageal events with acceptable precision. Recent technical advances permit continuous monitoring of lower esophageal sphincter (LES) pressure. This led to examination of the relationship between acid reflux and transient spontaneous relaxation of the LES. The systems described previously cannot adequately measure LES pressure or relaxation continuously because of movement of this sphincter during respiration. To circumvent this problem, Dent [6] developed a 6-cm sleeve catheter to straddle the LES throughout its entire range of motion. This device monitors the maximal pressure sensed at any point on the sleeve. Although manometric sleeve catheters are available commercially, they are used predominantly in the research laboratory. The choice of the physiograph to record pressure events is also important because this represents the major cost in any manometric system. The physiograph should satisfy the following requirements: at least four separate pressure-recording channels, including an extra channel for swallow sensor or event marker; variable paper speeds from 1 to 10 mm/second; full-scale recording for pressures between 0 and 400 mm Hg; and a recording channel for pH testing.

Recording Procedure Before attempting intubation, all of the necessary accessories must be on hand. Nasal topical anesthesia should be used: topical cocaine and viscous lidocaine are the most common agents used. Tissue paper, an emesis container, and a 30-mL water cup should be available. Reassuring the patient and explaining the procedure step by step are prerequisites to obtaining consistently good recordings. Short-term oral intubation studies using a manometric catheter passed through a mouthpiece can be used in place of nasal studies and may be more comfortable for the patient. [7]

Lower Esophageal Sphincter Measurements After GI intubation, all recording sites should be positioned in the stomach. This can be confirmed by observation of a positive deflection during abdominal breathing or compression. LES pressure usually is measured first. Two techniques are used. The first is the rapid pull-through technique. [8] The catheter is withdrawn at a rate of 1 cm/second while the patient suspends respiration until the distal three recording sites give LES pressure readings. The three distal sites are again placed in the stomach, and the cycle is repeated two more times. Mean and peak pressures are recorded from these nine measurements as that pressure above intragastric pressure. The second approach is the station pull-through technique. This method involves withdrawing the catheter in 0.5- to 1-cm increments and leaving it in each position for a few seconds. LES pressure and relaxation can be measured by this technique. Values from rapid pull-through LES pressure are usually 2 to 3 cm higher than those obtained with the station pull-through technique. [9] Relaxation of the LES requires that a distal recorder be stationed in the LES and a more proximal recording orifice be stationed in the lower esophagus to measure completeness of relaxation and coordination of relaxation with esophageal peristalsis. Relaxation should be complete, with a drop in intragastric pressure, and coordinated for more than 90% of wet swallows.

Esophageal Body Measurements After LES manometry is completed, all recording sites are withdrawn into the esophagus. The most distal recording site is placed 2 cm above the upper border of the LES. Because recording sites are separated by 5 cm and three sites are usually used, the distal 15 cm of the esophagus can be recorded. This segment represents the smooth muscle portion of the esophagus. Water is given with each swallow because esophageal body pressure is most reliably recorded after a bolus. [10] A total of 10 wet swallows is used to measure distal esophageal, or smooth muscle, body pressure. The striated muscle portion and the junction of the striated and the smooth muscles can be measured by sequentially withdrawing the catheter in 3-cm intervals and recording 5 wet swallows and again withdrawing the catheter 3 cm and repeating 5 wet swallows. Measurements of the esophageal body contractions include amplitude of contraction, duration of contraction, and percentage of abnormal contractions. Esophageal peristalsis is then observed. Simultaneous contractions (contractions in multiple segments at the

same time), spontaneous activity (pressure waves in the esophagus without swallowing), or repetitive contractions (three peaks) are considered abnormal. Failure of peristalsis increases somewhat with age but usually occurs in less than 15% of all wet swallows.

Upper Esophageal Sphincter Measurements UES pressure and ability to relax are measured. Techniques for the UES are the same as those for the LES. Recording speed may have to be increased from the usual 2.5 mm/second to 5 to 10 mm/second to record UES relaxation adequately. If a question of incomplete relaxation is present, different recording sites placed in the UES may demonstrate more complete relaxation. Because measurement of the UES may require withdrawing the catheter almost completely, only two recording orifices should be used to measure UES pressure, and more swallows should be performed to give a reliable pressure measurement. Continuous monitoring of the UES has been reported. [11] UES pressure values recorded with this technique tend to be lower than the values obtained with the method described previously. Special circumstances, such as a suspicion of striated muscle disorders of the upper esophagus (e.g., dermatomyositis), may require more attention to the proximal esophagus, but most of the clinician's time is spent recording from the distal esophageal segment. At the completion of the standard manometric examination, provocative tests, such as bethanechol, edrophonium, or balloon distention, can be performed to induce chest pain in patients sent for the evaluation of noncardiac chest pain. The mechanism by which provocative agents induce chest pain is unknown, and the use of provocative agents is experimental. [12]

Technical Aspects of pH Studies

Standard Tests After standard manometry is completed, pH studies are obtained. The first test is acid clearance. A 15-mL bolus of 0.1 normal hydrochloric acid is injected into the esophagus 5 cm above the LES. The patient is asked to dry-swallow every 30 seconds until the pH is restored to the baseline level. This requires at least 15 dry swallows. Poor acid clearance is seen predominantly in conditions that disrupt peristalsis (e.g., achalasia, diffuse esophageal spasm, and scleroderma) or in disorders affecting salivation (e.g., scleroderma and the sicca syndrome) because these conditions can disturb bolus movement or esophageal acid neutralization. The Bernstein examination allows the clinician to determine whether acid in the esophagus induces chest pain or discomfort. Infusions of 0.1 normal hydrochloric acid or saline should be given in a blinded manner by using a three-way stopcock connected to both reservoirs that are located behind the patient. Saline infused at a rate of 120 drops/minute for 5 minutes should be the initial perfusate. Acid infusions at the same rate should then be started, and saline should be stopped. Acid infusion should continue until symptoms begin or until 30 minutes have elapsed. If acid, but not saline, produces chest pain or heartburn, gastroesophageal reflux is likely. If both acid and saline or saline alone induces pain, the patient may have a hypersensitive esophagus. If neither produces pain, acid reflux is not the source of chest pain. Saline washout, to evaluate pain relief after acid-induced pain, is no longer considered essential to the diagnosis of acid-induced chest pain. Simultaneous recording of esophageal body pressures may show dysmotility as a mechanism for acid-induced chest pain. To determine whether gastroesophageal reflux is present, standard acid reflux testing is performed. The pH probe is placed 5 cm above the LES, and the patient is placed in the supine, right-sided, left-sided, and prone positions. In each of these positions, the patient is asked to perform the Valsalva maneuver and to move into the knee-chest position. Both maneuvers increase intra-abdominal pressure and increase the possibility of overcoming a weak sphincter. These maneuvers are repeated in the basal state and after loading the stomach with 300 mL of 0.1 normal (N) hydrochloric acid. Reflux of acid, seen as a decrease in pH to less than 4.0, in any position in the basal state, or in two positions after acid loading, is considered pathological reflux. [13] Sensitivity of this examination in patients with chronic reflux symptoms approaches 90%. After esophageal manometry and pH testing are completed, the patient should be observed for 30 minutes. Heartburn usually responds to antacids. More serious complications such as bleeding or perforation are extremely rare.

Ambulatory 24-Hour pH Testing, Esophageal Topographic Analysis, and Esophageal Impedance Recording Ambulatory outpatient evaluation under true-life conditions gives a better determination of whether acid in the esophagus is associated with symptoms. [14] Six monitoring systems are available in the United States. [15] All have demonstrated reasonable durability, and the life expectancy of each recording catheter is five to eight examinations. [15] Glass or antimony electrodes are frequently used. Reference electrodes must be attached to the patient with a conducting jelly for proper measurements. Newer recording catheters contain the reference electrode, eliminating the errors often seen with dislodgment of the reference electrode. The pH electrode is placed 5 cm above the upper border of the LES. Until recently, localization of the LES required manometric testing before pH recording. Using a standard electrode to measure the step-up in esophageal pH while the recording catheter crosses the LES allows placement accuracy of within 1 to 2 cm, when compared with positioning guided by manometry. This pH recording technique may be an acceptable alternative to standard manometric placement for 24-hour pH recording in patients who cannot tolerate esophageal manometrics or in an office practice in which manometric equipment is too costly for routine use. The ideal duration of recording has not been established. To differentiate patients with reflux from normal subjects, recording times of 12 and 24 hours have been more successful than shorter recording periods of only 3 hours postprandially. [16], [17] An ambulatory pH profile is best interpreted by calculating the total time that the pH is less than 4.0 and the number of episodes lasting longer than 5 minutes; this method yields the best positive-negative predictive value. [16], [18] Symptom correlation with pH decreases is also crucial in defining acid reflux as a cause for chest pain. [17] Correlation is rarely 100%, and multiple indexes have been used. Association of chest pain with pH less than 4.0 in 75% or more of painful episodes is considered strong evidence for acid-induced chest pain. Esophageal topographic analysis and esophageal impedance are two new technologies that have increased our understanding of esophageal and gastroesophageal reflux, respectively. [17], [19], [20] By using multiple, closely spaced, and circumferentially placed intralumenal sensors, esophageal topographic methods allow us to examine both the spatial and temporal relationships of esophageal peristalsis simultaneously. This technique allows the generation of three-dimensional maps of esophageal peristalsis beginning in the pharynx and ending in the gastric fundus (Fig. 150-1). Esophageal peristalsis has been demonstrated by topographic methods to be composed of four separate esophageal segments with demonstrable segmental abnormalities in motility and different responses to pharmacological interventions. [17] The research potential of topography is substantial, although application to clinical practice is unproven.

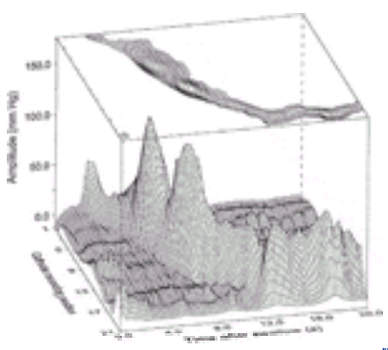

FIGURE 150-1. Three-dimensional topographic plots. The peristaltic wave proceeds from the upper through the lower esophageal sphincter shown in the foreground. Spatial distribution amplitude and duration are easily evaluated by multiple catheter recording sites separated by 1 cm over a 20-cm catheter length. The top of the figure has a contour plot to demonstrate three-dimensional amplitude data. Increasing amplitudes are represented by increasing concentric rings.

Concepts of gastroesophageal reflux and the antireflux barrier have been limited to measurement of acid and bile reflux and the measurement by manometry of common cavity phenomenon. [19], [20] Liquid reflux of gastric contents and gas reflux without change in pH could barely be detected. Using impedance measurement of bolus movement, gas or liquid reflux can be detected as a rise or drop in impedance, respectively. Initial studies have shown that reflux patients and controls do not differ in the total rate of reflux episodes but that acid reflux is higher in patients with reflux disease. Mixed reflux of both liquid and gas was seen in reflux patients and control subjects but pure liquid reflux and prolonged reflux were more common in reflux patients. One third of all reflux episodes are nonacidic. This technique will allow better definition of patterns of reflux in patients with esophagitis, patients with nonerosive reflux disease, and patients who fail proton pump inhibitor therapy despite good acid control. [20]

Indications, Contraindications, and Clinical Applications

Indications for esophageal manometry and pH monitoring are presented in Table 150-1. The primary indication is to evaluate esophageal dysfunction when anatomic causes (e.g., esophagitis, tumors, strictures, rings, webs, diverticula) are not present.

Indications for Esophageal Manometry
Patients with oropharyngeal dysphagia and normal anatomic studies
Cricopharyngeal achalasia (poor UES relaxation)
Pharyngeo–UES dyscoordination
Low-amplitude pharyngeal contractions
Early or later pharyngeal contraction
Early upper esophageal body contraction
Patients with esophageal dysphagia and normal anatomic studies
Primary esophageal body dysmotility
Achalasia
Diffuse esophageal spasm
Nutcracker esophagus
Hypertensive lower esophageal sphincter
Nonspecific esophageal dysmotility
Secondary esophageal body dysmotility
Collagen vascular diseases
Amyloidosis
Secondary achalasia
Chronic intestinal pseudoobstruction
Laser treatment for esophageal cancer
Sclerotherapy
Hypothyroidism
Patients with noncardiac chest pain
Primary esophageal body dysmotility
Indications for 24-hour pH Monitoring
Patients with noncardiac chest pain
Refractory acid reflux symptoms: evaluate treatment efficacy
Preoperative and postoperative evaluation for antireflux surgery
Patient with atypical presentations of acid reflux (ENT, pulmonary)

ENT, ear-nose-throat; UES, upper esophageal sphincter.

TABLE 150-1 Indications for Esophageal Manometry and 24-Hour pH Monitoring

Esophageal manometry is mandatory in patients with suspected achalasia, particularly in patients before pneumatic dilation or cardiomyotomy. Manometry has had less utility in patients with presumed diffuse esophageal spasm. Classic findings of esophageal spasm are not frequently seen in patients with esophageal chest pain.[10],[21],[22] Usually, esophageal manometry is performed to help exclude an esophageal source for chest pain. Motility disorders, other than diffuse esophageal spasm, that can cause chest pain and dysphagia include nutcracker esophagus and hypertensive LES. Although specific manometric criteria exist for each of these diagnoses, the physiologic basis for the symptoms in patients having these manometric findings remains to be elucidated. Position statements and a technical review for the clinical use of manometry have been published.[22]

Twenty-four-hour pH monitoring of the distal esophagus has helped to establish that excessive acid exposure is associated with chest pain. Young patients with no history of dysphagia or weight loss do not need pH testing to evaluate typical pyrosis or regurgitation. Twenty-four-hour pH testing is indicated in patients presenting with typical reflux symptoms who do not respond to treatment. Patients presenting with cervical dysphagia, cough, recurrent sore throat, or aspiration with a normal result from pulmonary and otorhinolaryngology evaluation also constitute a patient group in whom pH monitoring may define the cause for the symptoms.[22] Before antireflux surgery, all patients should undergo pH monitoring of the distal esophagus. This establishes the occurrence of acid reflux and whether the presence of acid in the distal esophagus is the source for symptoms because up to 10% of patients presenting for antireflux surgery do not have acid reflux.[23] In addition, this permits objective evaluation of the surgeon's ability to prevent acid reflux in the early and the late postoperative period. A technical review on the use of esophageal pH recording has been published recently.[23]

The use of esophageal function testing in patients with generalized GI motility disorders such as scleroderma and chronic intestinal pseudoobstruction is in its infancy. Testing should be confined to three groups of patients. The first group includes patients for whom the results of esophageal function testing will help the clinician in making a treatment decision. Although manometric findings are not highly specific for scleroderma, they are reasonably sensitive and occasionally can be helpful in supporting the diagnosis. Esophageal manometry and 24-hour pH monitoring also can be useful in evaluating patients in whom a therapeutic change will be forthcoming, depending on the findings. Examples include 24-hour pH monitoring in patients with scleroderma and recalcitrant esophagitis or patients with chronic idiopathic intestinal pseudoobstruction and dysphagia in whom pneumatic dilation is a treatment option. The last group includes patients in whom documentation of how the disease affects esophageal function may improve the physician's understanding of the mechanisms underlying symptoms and general knowledge of the disease process. These patients are the least likely to be candidates for monitoring and should be monitored only at established research motility laboratories.

Major contraindications for esophageal manometry are few and involve esophageal obstruction or large diverticula because of the risk for perforation. Barium study or endoscopy is prerequisite to manometric studies. The major reason for not performing esophageal manometry is if the information will not affect management. Esophageal manometry and pH testing are not required in all patients with typical reflux symptoms or noncardiac chest pain. A treatment course, coupled with careful exclusion of chest wall pain or panic attacks, should be attempted before manometry is performed. About 20% to 40% of patients referred with dysphagia or chest pain require manometric evaluations.[12]

GASTRODUODENAL MANOMETRY

Gastroduodenal manometry provides information regarding normal and abnormal contraction patterns in this region. The technique is helpful in diagnosing motility disturbances by localizing and characterizing abnormal motor patterns.

Gastroduodenal Motility in the Fasting and Fed States

Gastroduodenal manometry is used to assess the presence of interdigestive migratory motor complex (IMMC) activity in the fasting state and the occurrence of a typical fed pattern postprandially. In the fasting state, the stomach and small intestine demonstrate a cyclic, periodic contractile pattern referred to as the IMMC.[24] The IMMC is typically divided into four phases:

Phase 1: a period of quiescence
Phase 2: irregular motor activity
Phase 3: a brief period of regular contractions
Phase 4: a brief period of irregular activity, which is occasionally noticed before the quiescence of phase 1.

Figure 150-2A depicts examples of normal fasting intralumenal pressure patterns. Normal phase 2 activity is demonstrated, culminating in a phase 3 complex in the antrum and duodenum. Phase 3 contractions occur at a rate of 3 per minute in the antrum; they occur more rapidly, 11 to 12 per minute, in the duodenum. Occasionally, phase 3 activity begins in the proximal duodenum and not in the stomach, although this usually is preceded by strong antral phase 2 activity.

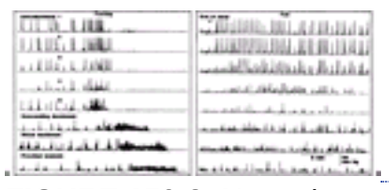

FIGURE 150-2. Normal gastrointestinal motility. **A:** Normal fasting interdigestive migrating motor complex (IMMC). Notice phase 2 activity in the antrum and duodenum culminating in phase 3 (IMMC) activity in the antrum, the pressure activity gradually propagating into the duodenum, and the gradual change in configuration of the waves while they move through the antroduodenal junction. **B:** Normal fed activity. Notice irregular but persistent phasic pressure activity in the distal antrum and proximal small bowel. (From ref. [36].)

After ingestion of nutrients, IMMC activity is inhibited and replaced by a fed pattern, the characteristics of which vary.[25] For example, liquid nutrients decrease the amplitude of contractions in the antrum and generate irregular motor activity in the small intestine, whereas ingestion of solid food produces high-amplitude contractions in the distal antrum and a motor pattern similar to that generated by liquid nutrients in the small intestine. Figure 150-2B shows the irregular but persistent phase 2 activity characteristically observed postprandially in the distal antrum and proximal small intestine. Repetitive pressure activity with an amplitude of more than 50 mm Hg is usually noticed in the antrum at a rate of 3 per minute, and persistent, irregular activity is noticed in the duodenum.

Technical Aspects

Gastroduodenal manometry requires upper GI intubation with a specially adapted polyvinyl catheter on which miniature pressure transducers are mounted; alternatively, manometric perfusion catheters are assembled together as a multilumen tube. The monitoring probe is guided into the proximal jejunum under fluoroscopic guidance. Manometric devices using miniature transducers or the pneumohydraulic perfusion system provide similar information on GI motor activity. Both techniques measure changes in intralumenal pressure, which is an indirect assessment of smooth muscle contractions. Occasionally, the recording probe must be positioned in the duodenum with an endoscope and manually advanced into the proximal jejunum.

Assessing gastroduodenal motility requires the capability to monitor pressure changes from 6 to 12 recording sites, including the antrum, pylorus, duodenum, and proximal jejunum. Manometric sites should be spaced 1 cm apart across the gastroduodenal junction and at 10-cm intervals in the small intestine. A commonly used arrangement involves positioning five closely spaced recording sites to monitor antral, pyloric, and proximal duodenal motility, whereas three sites are spaced more widely for assessment of distal duodenal and proximal jejunal motility. The manometric recording system includes a polygraph with sufficient channels to monitor each recording site. The pressure transducers must be calibrated at the beginning and end of each study. Recent studies suggest that 24-hour antroduodenal motility can be recorded reliably and reproducibly in the ambulatory setting using solid-state catheters. [26]

Indications and Contraindications

Mild to moderate GI symptoms are of limited value for predicting the outcome of manometric studies because patients with normal manometry results may report symptoms similar to those whose studies yield abnormal results. [27] The presence of GI symptoms and evidence of collagen-vascular, metabolic, urologic, neurologic, and psychiatric disorders increase the likelihood of abnormal manometric findings. [27] Gastroduodenal manometry is indicated in a patient with suggestive symptoms after mechanical obstruction has been ruled out with endoscopy or radiologic studies; appropriate consultations and workups have been performed to evaluate possible endocrine-metabolic, neurologic, and psychiatric disorders; and documentation of delayed gastric emptying or impaired small intestinal transit is available. Gastroduodenal manometry can be helpful in making the diagnosis of a motility disorder by demonstrating an abnormality in motor activity and localizing the involved region. In addition, the technique can be useful in monitoring the patient's course and response to therapy. These results rarely indicate the need for additional procedures, such as laparotomy, to rule out undetected anatomic abnormalities, or biopsy, to obtain a full-thickness intestinal specimen to rule out an infiltrative or degenerative process.

Contraindications to gastroduodenal manometry include anatomic abnormalities such as a diverticulum or fistula, respiratory diseases or a hypersensitive gag reflex that may lead to poor tolerance of the procedure, and prior GI surgeries, such as a Billroth II antrostomy, that can be difficult to evaluate because of manometric tube placement issues.

Data Interpretation

The typical recording session lasts about 5 hours. Three hours are devoted to recording motor activity in the fasted state, followed by ingestion of a standard meal and 2 hours of postprandial recording. Long-term (6 to 24 hours) records of small bowel motility, including evaluation during sleep, may enhance diagnostic accuracy. [28] The motility pattern normally observed during the fasting state, the IMMC, is often examined for the following: the mean cycle duration or period of time between phase 3 complexes; the duration of each phase and the amplitude and propagation velocity of phase 3 complexes in different regions of the stomach and small intestine; and the rate of contractions during phase 3 activity. [29], [30] Useful qualitative information is derived from inspection of the tracing to determine whether phase 3 activity is present; whether phase 3 activity originates from the gastric pacemaker or an ectopic site; whether the time intervals for the phases of the IMMC are appropriate; and whether the complex propagates in a normal (aboral) or abnormal (retrograde) manner.

The pressure activity pattern observed in the fed state is determined by the solid versus liquid composition of the meal and its nutrient content. Therefore, the meal must be standardized to exclude meal composition as a variable between studies. Usually, inhibition of IMMC activity occurs postprandially, replaced by phasic pressure waves in the antrum and irregular, phasic contractions in the small intestine. Pressure activity is often quantified in the form of a motility index, which reflects the amplitude and frequency of contractions for a defined period.

Interpretation of gastroduodenal manometric tracings is complicated by the intrinsic variability of the fasting and fed motor patterns. A number of IMMC parameters, including the duration of its constituent phases, the velocity of propagation of the phase 3 activity, and the duration of inhibition by food, demonstrate variability among patients as well as within the same individual when evaluated serially. [31], [32] A number of motor patterns, such as the presence of jejunal-clustered contractions, which have been proposed as a characteristic feature of the irritable bowel syndrome, apparently can be influenced by emotional and physical stress, including the anxiety associated with having the examination. [31], [33], [34] Abnormalities that have been reported in the fasting and fed motility patterns are summarized in Table 150-2.

Abnormalities Observed During the Fasting State
Bursts of phasic activity having an abnormal duration (>2 min), amplitude (>20 mm Hg), and frequency (10-12/min), which are nonpropagating and distinct from phase 3 activity
Sustained (>30 min) poorly coordinated phasic activity, isolated to one or more segments of the intestine
Low-amplitude contractions but otherwise normal
Absent, incomplete, or retrograde propagation of phase 3 complexes covering a distance of at least 30 cm
Prolonged (>3 min) increase in basal tone (>30 mm Hg) during phase 3 activity
Abnormalities Observed During the Fed State
Persistence of a fasting-like pattern after ingestion of a meal
Low-amplitude pressure waves in the antrum and small intestine
Bursts of phasic contractions that fail to propagate
Premature return of IMMC activity within 90 min after ingestion
Broad-based contractions occurring in the presence of increased tone, sometimes termed *minute clusters*

IMMC, interdigestive migratory motor complex.
Adapted from ref. [8].

TABLE 150-2 Abnormalities Reported in the Fasting and Fed Motility Patterns on Gastroduodenal Manometry

Clinical Situations that Affect Gastroduodenal Motility

GI manometry can be useful to distinguish motility patterns that are typically observed with intestinal myopathy versus enteric or extrinsic neuropathy. Diseases associated with an intestinal myopathy often demonstrate normal motor patterns but with abnormally low pressures. In contrast, diseases that affect the enteric or extrinsic neural pathways can demonstrate abnormalities in the configuration and propagation of the IMMC or failure to convert from the fasting to fed motor pattern postprandially.

Manometry often reveals a localized region of infrequent, weak contractions, particularly in the postprandial state in the presence of a relatively normal fasting motility pattern (Fig. 150-3). This pattern can be contrasted with the patient who also presents with a pseudoobstruction-like picture but whose disease affects the intrinsic or extrinsic neural pathways that regulate GI motility. [35] In this situation, abnormalities of fasting and postcibal motility are observed. In the fasting state, abnormalities in the configuration and propagation of phase 3 complexes are seen (Fig. 150-4).

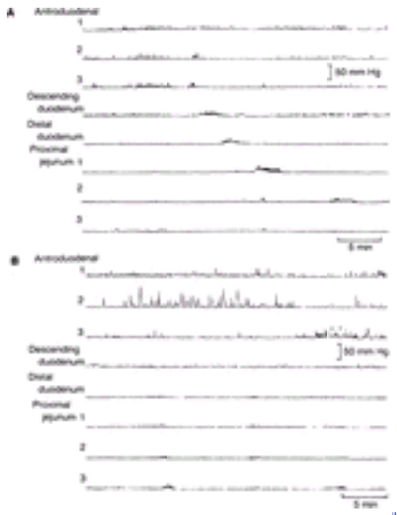

FIGURE 150-3. Myogenic chronic intestinal pseudoobstruction in a 71-year-old man with recurrent episodes of vomiting, abdominal pain, and constipation for 1 year. Radiologic examination revealed markedly dilated intestinal loops without mechanical obstruction. Full-thickness specimens taken from the terminal ileum and sigmoid colon at exploratory laparotomy showed smooth muscle degeneration with replacement of muscle fibers by fibrous tissue. **A:** Fasting tracing shows low-amplitude but normal propagation of migrating motor complex and virtual absence of activity in the most distal jejunal port. **B:** In the fed phase, antropyloric activity is mostly preserved, with waves of normal amplitude and frequency. In contrast, there is virtual absence of phasic pressure activity in the intestine. (From ref. [36].)

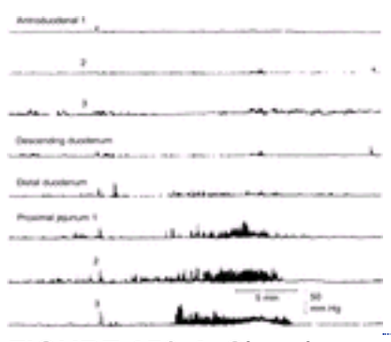

FIGURE 150-4. Chronic sensory and autonomic neuropathy with gut dysmotility in a 34-year-old woman with weakness in the extremities, speech impairment, and dysphasia for 10 years. At the time of study, she had episodes of diarrhea, abdominal pain, and labile blood pressure with hypertensive crises followed by hypotension. There was evidence of autonomic dysfunction with sympathetic and parasympathetic involvement, and a peripheral sensory neuropathy involved upper and lower extremities. Fasting tracing reveals simultaneous bursts of phase 3–like activity in the proximal small bowel. (From ref. [36].)

Gastroduodenal manometry is typically performed on patients who present with evidence of gastroparesis. During the fasting state, phase 3 activity may be absent in the antrum, but the most common abnormality is postcibal antral hypomotility: antral phasic pressure waves occur less frequently and with less force (i.e., decreased amplitude). This motility disorder often is associated with delayed gastric emptying of solid food. [36], [37] Antral hypomotility is observed in a wide range of clinical situations: diabetes mellitus, generally type 1 with evidence of autonomic neuropathy [38], [39]; use of adrenergic and dopaminergic agents [40], [41] or cholinergic agonists [42]; occasionally after surgical procedures, including fundoplication, [43] vagotomy alone or with pyloroplasty, [44], [45] and cardiomyotomy for achalasia [46]; and idiopathic gastroparesis. [47], [48]

Localized, uncoordinated bursts of tonic and phasic activities can occur in the small intestine that blur the distinction between fasting and fed motility patterns. Diseases that can present in this manner include early scleroderma [49], [50]; diabetes mellitus [38], [39]; amyloidosis [51], [52]; visceral autonomic neuropathies and degenerative conditions involving the autonomic system, such as idiopathic orthostatic hypotension, Shy-Drager syndrome, and pandysautonomias [53], [54]; chronic idiopathic constipation, that is, colonic inertia [55]; lesions involving the central nervous system, especially the brain stem [56], [57]; and neurodegenerative disorders involving the spinal cord, such as syringomyelia. [58] Patients with intestinal dysmotility may have normal gastric manometric patterns but demonstrate delayed gastric emptying, supporting the view that these two tests can be complementary.

After partial gastrectomy and gastroenteric anastomosis, occasional patients experience a profound motility disturbance involving fasting and fed activities. [59] Although no pathognomonic findings are seen on manometry, typically localized areas of intense activity alternate with regions of quiescence. In patients treated for postoperative gastric stasis with the Roux-en-Y procedure, the Roux limb may contribute to delayed emptying in some, perhaps because of loss of electrical synchrony in the implanted jejunal efferent limb. [60] In addition, patients with postcholecystectomy sphincter of Oddi dysfunction can demonstrate abnormalities in phases 2 and 3 of the IMMC. [61]

Patients with radiation enteritis can exhibit abnormal gastroduodenal manometric findings, especially localized, poorly coordinated, either high- or low-amplitude pressure activity. Antral hypomotility can be observed and, in the presence of uncoordinated small intestinal activity, may be associated with intractable vomiting. [62], [63] A number of infectious processes can present with pseudoobstruction-like symptoms and signs thought to be due in part to disturbances of motility. These disorders may affect intrinsic or autonomic neural pathways and include infectious mononucleosis, [64] herpes varicella-zoster virus, [65] botulism B, [66] secondary bacterial overgrowth in the small intestine, [67] and Guillain-Barré syndrome. [68]

Malignancies involving the posterior abdominal wall may interfere with the autonomic neural pathways to the GI tract, producing a chronic ileus. Extra-abdominal malignancies such as oat cell carcinoma of the lung can be associated with a paraneoplastic syndrome, which produces a clinical and manometric picture consistent with intestinal pseudoobstruction. [69], [70]

Metabolic abnormalities associated with disturbances of motility include hyperthyroidism and hypothyroidism and hyperparathyroidism and hypoparathyroidism. Some reports suggest that rapid gastric emptying and rapid intestinal transit occur in hyperthyroidism. [71], [72] Patients with hypothyroidism are at risk for pseudoobstruction, paralytic ileus, and megacolon. [73], [74] Patients afflicted with hyperparathyroidism frequently report symptoms of abdominal pain, nausea, vomiting, gastroesophageal reflux, and constipation possibly related to defective neurotransmission. [75], [76] Hypoparathyroidism can be associated with intestinal pseudoobstruction and regional areas of tetany. [77] Patients with malnutrition and anorexia nervosa frequently have delayed gastric emptying, especially of solid meals. [78] The etiology of the disorder is not known, but disturbances of autonomic function have been reported. [79] Patients who have experienced hypoxia and reperfusion can demonstrate abnormal motility patterns. [80] When evaluating patients with suspected colonic inertia, it is important to consider the possibility of a more generalized panenteric motility disorder. [81] Clinical situations associated with abnormalities of GI manometry are summarized in Table 150-3.

Disorders Presenting with Symptoms and Signs of Gastroparesis and Absent Phase 3 Activity or Postcibal Antral Hypomotility on Manometry
Diabetes mellitus, type I with autonomic neuropathy
Drugs: adrenergic agonists, cholinergic antagonists, and opiate agonists
Postoperatively: fundoplication, vagotomy ± pyloroplasty, and cardiomyotomy for achalasia
Idiopathic
Disturbances of Intestinal Motility Presenting Clinically as Chronic Intestinal Pseudoobstruction
Manometry reveals localized region of infrequent, weak contractions, particularly in the postcibal state in the presence of relatively normal fasting motility pattern
Myotonic dystrophia
Late progressive systemic sclerosis
Manometry reveals abnormalities in both fasting and postcibal motility; in the fasting state, abnormalities in the configuration and propagation of phase 3 complexes are observed, whereas in the small intestine, localized, poorly coordinated bursts of tonic and phasic activities are seen
Early scleroderma
Diabetes mellitus
Amyloidosis
Visceral autonomic neuropathies, degenerative conditions involving the autonomic nervous system, and lesions involving the central nervous system

TABLE 150-3 Clinical Situations Associated with Abnormalities on Gastroduodenal Manometry

ELECTROGASTROGRAPHY

Electrophysiologic Basis of Gastric Motility

Electrogastrography (EGG) measures gastric electrical activity noninvasively. The development of a gastric contraction requires that electrical events occur at the membrane of smooth muscle cells. The electrophysiologic properties of gastric smooth muscle cells vary, depending on the region of the stomach. Myoelectrical activity in the proximal one third of the stomach is characterized by a steady, nonfluctuating transmembrane potential associated with slow contractions occurring every 1 to 3 minutes. The electrical changes observed in this region are small in magnitude and usually not seen during EGG. The distal two thirds of the stomach and small intestine demonstrate slow, phasic changes in membrane potential. This cyclic change in electrical potential has been called by various names, including basal electrical rhythm (BER), slow waves, pacesetter potential, and control potential.

The BER appears to originate from the longitudinal muscle layer or nonneural boundary cells between the longitudinal and circular layers. [82], [83] An important feature of the BER is that it represents cyclic depolarization of the smooth muscle cells in the absence of associated peristaltic contractions. Rapid depolarization of the smooth muscle membrane is necessary for a contraction to occur. This event is referred to as an action potential or *electrical response activity* (ERA). Peristaltic contractions in the gastric body and proximal antrum are associated with an increase in the amplitude and duration of the depolarization plateau above the threshold for smooth muscle contraction. In the distal antrum and small intestine, contractions are associated with electrical spikes or action potentials superimposed on the depolarization plateau.

The maximal frequency that GI smooth muscle can contract is established by the frequency of the BER in that region. In the stomach, the frequency of the BER is established by a pacemaker site located in the proximal body along the greater curvature. [84] In humans, the BER propagates longitudinally and circumferentially from this site at a frequency of about 3 cycles per minute. In the duodenum, the frequency increases to 11 to 12 cycles per minute, which is established by a different pacemaker. Passage of the BER between the stomach and duodenum is prevented by the pylorus, which acts as a functional barrier. Propagation of the BER occurs myogenically by way of gap junctions between adjacent smooth muscle cells. Occurrence of peristaltic contractions is influenced by several factors, including the presence of a fed or fasted state, the neurotransmitter and hormonal milieu, and, most likely, the presence of local regulatory, or paracrine, substances. The presence of these regulatory substances appears to influence the sensitivity of smooth muscle cells to contract by decreasing or increasing the occurrence of plateau potentials. The force of contraction is determined by the amplitude and duration of the plateau potential.

Technical Aspects

Serosal, mucosal, and cutaneous EGGs have been used to record electrical activity of the stomach. This chapter emphasizes cutaneous EGG because it is noninvasive. Serosal recordings require a laparotomy for surgical placement of the recording electrodes. Mucosal recordings require intubation and attachment of the recording electrode to the gastric lining by suction or by using magnetic force to hold the electrode in position. [85], [86] Mucosal recordings reflect the true extracellular gastric electrical activity better than cutaneous recordings and appear to be less susceptible to interference from extragastric electrical signals from duodenal, colonic, cardiac, and respiratory sources. [83] Proper positioning of the recording electrodes is confirmed fluoroscopically. Optimally, at least three internal electrodes should be used, which permits assessment of the direction of electrical propagation: antegrade versus retrograde conduction. Respiratory movements are recorded concurrently with a pneumobelt transducer.

Cutaneous recording requires the placement of electrodes across the anterior abdominal wall, along the antral axis, which represents the imaginary line along the proximal to distal portion of the stomach. Monopolar recording devices (those with one active and one ground electrode) and bipolar recording devices (those with two active and one ground electrode) have been used. The quality of recordings obtained with the bipolar configuration appears to be better because of the advantageous signal-to-noise ratio it provides. [85] The electrical activity detected by the electrodes reflects potential differences over large areas of the stomach and, therefore, the summation of electric potentials generated by many gastric smooth muscle cells. [87] The signal represents the BER in the noncontracting stomach and the ERA in the contracting stomach. It correlates well with the frequency but not the amplitude of serosal recordings.

The signals monitored during cutaneous EGG usually require filtering to decrease undesired signals from other sites, such as the duodenum, colon, heart, or lungs. [88], [89] However, filtering can result in the loss of useful data. In addition, cutaneous monitoring may increase the likelihood of missing focal gastric electrical disturbances because they are hidden by the signal from the normally depolarizing gastric smooth muscle cells.

The normal gastric electrical signal in humans occurs at a frequency of about 3 to 4 cycles per minute. This is equivalent to 0.05 Hz. The typical EGG filter partially eliminates signals having frequencies of more than 0.2 Hz (12 cycles per minute) and less than 0.03 Hz (1.8 cycles per minute). In addition to filtering, the EGG signal is amplified and recorded along with other signals, such as manometric data, on a polygraph. [90], [91] Contemporary recording systems include an analog-to-digital converter, which allows electronic storage of data.

Mucosal and cutaneous recordings can be analyzed visually for information regarding frequency, amplitude, and configuration of waveforms. However, computer-based analytic techniques have become the norm because they analyze large amounts of data and generate useful information from recordings unsuitable for visual interpretation. Most methods used to analyze EGG signals employ some form of spectral analysis incorporating one of several available mathematical models such as the fast Fourier transformation, which transforms the signal from the time domain to a frequency spectrum. [92], [93]

A power spectrum of the recorded signals is then obtained (Fig. 150-5). The fundamental frequency, 3 cycles per minute or 0.05 Hz in humans, is represented with greater power because this frequency contributes a greater portion of the recorded signal frequencies. Other electrical frequencies originating from the duodenal, colonic, respiratory, or cardiac sources may be represented, but at a lower power. One drawback is loss of the ability to determine when specific events occurred during the recording period. For example, it is typically necessary to record data for several minutes to compute the transform, which generates the frequencies present during the recording interval and, therefore, hinders correlation of the EGG data with manometric data and the patient's symptoms. This limitation has been partially alleviated by the development of running spectral analysis. [94] Using this approach, the power spectra of brief overlapping stretches of the EGG signal are computed and presented as a function of time. Although easy to perform, this approach is susceptible to problems associated with inaccurate or erratic spectral estimates because of the recording time required to generate an accurate fast Fourier transformation. Dysrhythmias of brief duration will not be detected reliably using this approach. This issue is addressed in part by adaptive spectral analysis that improves the detection of short-duration dysrhythmias using adaptive autoregressive moving-average modeling. [95] Thus, the reduction of artifact in the EGG signal is an ongoing technical challenge. Discussion of these issues and recommendations regarding possible corrective measures have been published. [96], [97]

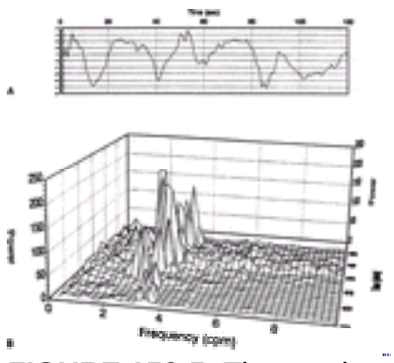

FIGURE 150-5. The results of an electrogastrographic study of a healthy volunteer. **A:** The raw slow-wave signal after ingestion of a 500-kcal mixed solid-liquid meal. The signal appears as a regular sinusoidal wave with a period of about 20 seconds. The raw signal can be analyzed by power spectral analysis to determine the dominant slow-wave frequency at any given time during the recording. **B:** The pseudo–three-dimensional plot from the power spectral analysis. With fasting, the slow wave was regular at 3 cpm, but of low intensity. With meal ingestion, at 15 minutes, there was a transient decrease in slow-wave frequency and a prolonged increase in signal amplitude at 3 cpm. (Courtesy of William L. Hasler, M.D., Ann Arbor, MI.)

Indications and Contraindications

The indication for performing EGG is similar to that for gastroduodenal manometry, namely, a patient who presents with a history of unexplained nausea, vomiting, and evidence of impaired gastric emptying. EGG is usually performed after documentation of abnormal gastric emptying or in conjunction with gastroduodenal manometry. EGG complements gastroduodenal manometry and gastric emptying studies by providing additional information that may reveal the basis for disturbed gastric motility.

Interpreting the Electrogastrogram

The normal gastric electrical signal is established by the gastric pacemaker at a frequency of 3-4 cycles per minute. The characteristic shape of the electrical waveform detected during EGG varies, depending on the location of the electrode: internal (mucosal or serosal) versus external (cutaneous) electrogastrogram. Abnormalities include both increased rates (tachygastria) and decreased rates (bradygastria) of electrical pacemaker activity. Tachygastria, defined as more than 4 cycles per minute, and bradygastria, defined as less than 2 cycles per minute, should be present for at least 1 minute. [98] Other electrical abnormalities include the presence of a dysrhythmia associated with an irregular resting membrane potential and abnormal wave configuration and a mixed or so-called tachybradyarrhythmia. [98] Tachygastria frequently develops at an ectopic site in the distal antrum and propagates in a retrograde manner. [99] Gastric contractions are often absent in the presence of tachygastria because of deformed BER, resulting in the failure of the plateau potential to reach threshold potential. [91], [100] In contrast, bradygastria arises in the corpus and antrum and propagates aborally. [101]

Clinical Situations that Affect the Electrogastrogram

The amplitude of slow-wave activity detected by cutaneous EGG increases during gastric contractions in the fasting and fed states. The EGG amplitude, but not the frequency, correlates significantly with gastric emptying. [102] Tachygastria and other dysrhythmias have been reported in unexplained nausea, bloating, and vomiting [99], [103] (Fig. 150-6); symptomatic ulcer [104]; nonulcer dyspepsia and delayed gastric emptying in children [105]; nausea during pregnancy [106] (Fig. 150-7); diabetes with chronic nausea, vomiting, and delayed gastric emptying in the fasting state [93]; chronic renal failure [107]; chronic intestinal pseudoobstruction [108]; Parkinson's disease [109]; systemic sclerosis [110]; and end-stage liver disease [111] and in normal subjects in whom motion sickness is induced experimentally. [112], [113] It is noteworthy that the EGG correlated poorly with nuclear scintigraphic gastric emptying studies in children with symptoms suggestive of gastric motility disorders. [114]

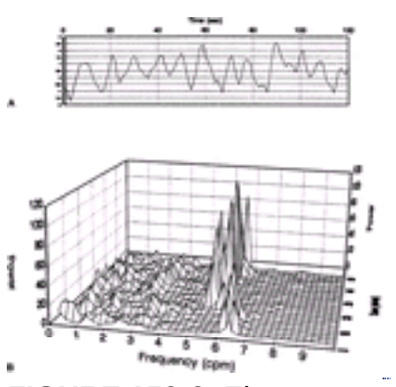

FIGURE 150-6. Electrogastrography (EGG) was used to evaluate a 28-year-old woman with persistent nausea, bloating, and fullness who had previously undergone a solid-phase gastric emptying scan, which yielded normal results. **A:** A low-amplitude raw EGG signal after ingestion of a 250-kcal liquid meal with a period of about 10 seconds. The raw EGG signal was evaluated with power spectral analysis. **B:** The pseudo–three-dimensional plot demonstrates the presence of a dominant slow-wave frequency that is higher (5 to 6 cpm) than in the healthy volunteer (see Fig. 150-5). This patient showed evidence of tachygastria, which was associated with antral hypomotility on a simultaneously performed manometric study. (Courtesy of William L. Hasler, M.D., Ann Arbor, MI.)

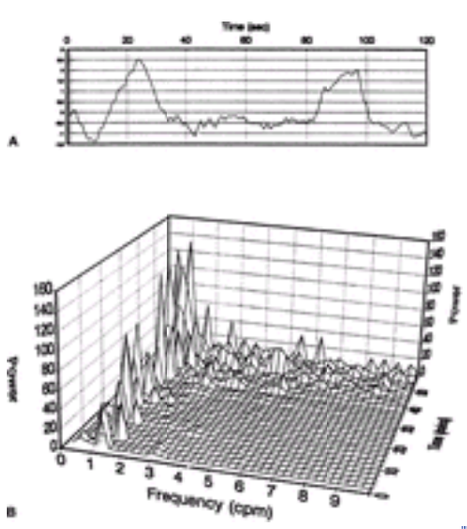

FIGURE 150-7. Women with nausea in the first trimester of pregnancy can exhibit marked slow-wave disturbances. Electrogastrography was performed on a 22-year-old woman in the 11th week of pregnancy. **A:** Large but infrequent slow waves are seen on the raw tracing. **B:** Pseudo–three-dimensional plotting of the power spectral analysis demonstrates that the bulk of the slow-wave signal is in the frequency range from 0.75 to 2 cpm. This pregnant patient exhibited bradygastria in association with her first-trimester nausea. (Courtesy of William L. Hasler, M.D., Ann Arbor, MI.)

In summary, EGG can be a useful diagnostic tool in the evaluation of patients with unexplained nausea, vomiting, and delayed gastric emptying. The EGG should be considered in individuals with delayed gastric emptying after metabolic and structural abnormalities have been ruled out. EGG can provide information regarding whether a gastric dysrhythmia presenting in the form of tachygastria, bradygastria, or a combined tachybradyarrhythmia is the cause of the motility disturbance. This information is not readily derived from manometry. The technique also may prove to be useful for following a patient's course and response to therapy. Technical advances will allow routine 24-hour ambulatory EGG in the future. [115]

ELECTRONIC BAROSTAT AND ITS APPLICATION IN EVALUATING GASTROINTESTINAL SENSORY AND MOTOR FUNCTION

Technical Aspects

The barostat is a research tool used by investigators interested in identifying the pathways underlying the physiology and pathophysiology of GI neuromuscular function. It has been used extensively to evaluate tone in the digestive tract and visceral sensitivity. The objective of barostat studies is to isolate a segment of the gut without interfering with its function and to follow the movements of the wall. [116] The barostat was designed to maintain constant pressure within an air-filled bag positioned in the lumen of the target organ. The barostat aspirates air to maintain a constant intrabag pressure during contraction and injects air during relaxation. The volume of air entering or leaving the bag is an indirect measurement of changes in the tone of the organ. Circumferential wall tension is defined by the Laplace law as the radius of the distended viscus multiplied by the transmural pressure. The transmural pressure is the difference between pressure within the distending bag and the pressure in the chest or abdomen outside the viscus.

The probe consists of a bag or a balloon connected by a catheter to the barostat. Latex balloons have been used widely and are characterized by a rapid increase in internal pressure for small-volume changes. When the pressure increases above the elastance threshold, the balloon becomes plastic and may accommodate large volumes with little increase in pressure. By contrast, polyethylene bags display an infinite compliance at low pressure or volume ranges, so that they may accommodate large volumes without an increase in pressure until the volume of air injected into the bag is equal to the volume of the bag itself. Further increases in volume will be associated with a rapid increase in internal pressure.

In distention studies, the function of the probe is to apply pressure on the wall of the gut in such a way that the pressure stimulus is equally distributed around the circumference of the organ over the whole segment to be distended. The probe should not interfere with the measurement of volumes needed to reach a given pressure. Many investigators use a bag instead of a balloon. The primary advantage of a latex balloon is its compact size, making it is easier to pass through the mouth or anus. The subject's position is important because the weight of adipose tissue overlying the organ and the tone of the abdominal wall directly influence barostat recordings. [117]

Investigators agree on some elements in distention study design: First, measurements of balloon pressure are less susceptible to intersubject and interlaboratory variability than measurements of balloon volume because the pressure scale compensates for differences in bag shape, smooth muscle compliance, and contractile activity. Second, both the pressure and volume that produce a sensation should be used to define sensory thresholds. Finally, the threshold for sensation tends to be higher when using a rapid inflation protocol compared with slow inflation. Additional information regarding distention protocols is available in a comprehensive review. [118]

When designing protocols to evaluate sensory thresholds, the investigator should be aware of the distinction between perceptual sensitivity and response bias. *Perceptual sensitivity* refers to the ability to detect distention, whereas *response bias* refers to the subjective aspects of reporting behavior—for example, the intensity of pain. Most protocols are designed to minimize response bias. Within practical limits, multiple distentions at each pressure or volume yield more reliable estimates of perceptual sensitivity.

Subjects should be asked to rate the intensity and unpleasantness of the sensations produced by distention separately and on scales, rather than simply indicating the presence or absence of the sensation because the latter is less sensitive. The rating scale should be an equal-interval or ratio scale, although many investigators use ordinal scales or visual analog scales. [119], [120]

Before barostat study, the investigator should be aware of potential anatomic abnormalities in subjects, such as diverticulosis or prior surgical procedures, that could complicate intubation. Patients who are gravely ill or experiencing persistent nausea, vomiting, or diarrhea are not likely to tolerate the technique.

Clinical Applications and Caveats

The electronic barostat has been used to measure changes in visceral tone in several regions of the GI tract. The technique has proved useful in several areas: (1) assessing sensory thresholds and reflex behavior in the GI tract, [121], [122] and [123] (2) establishing the role of altered visceral perception in functional bowel disorders, [124], [125], [126] and [127] and, (3) evaluating new therapeutic modalities. [128], [129] and [130]

When designing a protocol, the investigator should always be conscious of the likelihood of intraindividual and interindividual variability. Some authorities believe that phasic distentions are reproducible when repeated twice on the same day or even at 2-week intervals. [131] Others suggest that significant variation exists between results obtained in the same subject on different days. [128] These different opinions have resulted in the recommendation to include a baseline distention test before each attempt to evaluate the response to a therapeutic intervention or placebo. This allows the investigator to compare the response to distention during the therapeutic intervention (e.g., active drug or placebo) with the baseline distention performed on the same day. The differences between intervention and baseline distentions can be compared across sessions. Interindividual variability in sensory thresholds accounts for most of the large overlap between healthy controls and patients with functional bowel disorders when evaluating sensory thresholds.

The barostat also has been used to study gut reflex behavior, such as the gastrocolonic reflex and peristaltic reflex. [108] The ability of the barostat to record both increases and decreases in visceral tone reliably lends itself to the examination of the peristaltic reflex (e.g., ascending contraction and descending relaxation).

ANORECTAL MANOMETRY

Anorectal manometry is used in the clinical assessment of patients in whom a problem with defecation is suspected. The technique is particularly helpful in evaluating the most distal components of the anorectal sphincter mechanism: the internal and external sphincter muscles and the neural regulation of these muscles. [132], [133] and [134]

Technical Aspects

Direct pressure-sensitive microtransducers and perfused probes have been used to record anal sphincter pressure. The former eliminates the need for a fluid perfusion system by positioning the sensor device at the point at which pressure is to be monitored. Potential drawbacks include the relatively high cost and susceptibility of the solid-state microtransducer system to malfunction. The latter approach relies on the perfusion of a noncompressible fluid, such as water, at a constant rate through low-compliance catheters. Change in pressure is monitored as change in the resistance to flow that the sphincter offers to the constant flow.

Several technical issues must be kept in mind when performing anorectal manometry, particularly with the perfused catheter system. These include the potential for inducing recording artifacts because of probe rigidity, probe diameter, and the infusion rate. For example, a rigid probe is easier to position but can deform the anal canal and thereby produce an artifact. The diameter of the probe introduces some distortion of the anal canal by its very placement; therefore, it is advisable to use the smallest probe diameter (4 to 8 mm) that allows adequate contact with the opposing surfaces of the canal. From a practical standpoint, this is not an important issue unless dealing with a patient who has a markedly patulous sphincter. In this situation, it may be necessary to use a larger-diameter probe of 10 to 20 mm. Infusion rates usually are 0.3 to 1 mL/minute per channel. During a routine study taking 15 to 45 minutes, considerable fluid accumulation can occur within the rectum when using a multilumen probe with 3 to 12 channels per probe. This can alter the reproducibility of the results during the recording period.

The pressure profile of the anal sphincter is obtained using a station pull-out or continuous pull-out technique. The station pull-out approach involves positioning the probe several centimeters above the anal verge and measuring the pressure at various intervals, usually 1 cm, while removing the probe. Because moving the probe

induces changes in sphincter pressure, a stabilization period of 30 seconds is observed before recording the pressure at each interval. The advantage of this approach is the accuracy of the pressure recordings at each interval. This is potentially offset by the absence of the entire pressure profile, and failure to identify accurately the point of maximal squeeze pressure. With the continuous pull-out technique, the probe is positioned above the canal and subsequently pulled through the sphincter at a constant rate, producing a continuous pressure profile along the longitudinal axis of the anal canal. To help avoid recording artifacts, a mechanical pulling device must be used to ensure consistency and reproducibility. The probe should be rendered as frictionless as possible with the liberal use of lubricant.

An important modification of these recording techniques involves positioning and distention of a balloon within the rectum to examine the rectal-anal inhibitory reflex.[108] This is often referred to as *balloon reflex manometry*. Distention of the rectum by stool, or artificially with a balloon, should evoke relaxation of the internal anal sphincter. The balloon catheter assembly is positioned 10 to 15 cm above the anal verge and distended to a volume greater than 60 mL to initiate the inhibitory reflex. Either two small balloons or the perfused catheter system are positioned in the anal canal to monitor the internal and external sphincter response to distention of the rectal balloon. Relaxation should be observed only at the level of the internal sphincter. A typical perfusion apparatus used to perform balloon reflex manometry is shown in Figure 150-8. Normal proximal sphincter response to increased rectal pressure during balloon distention is depicted in Figure 150-9.

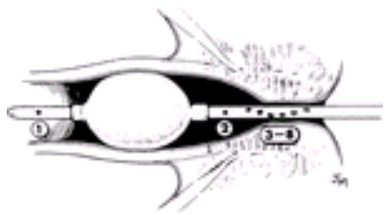

FIGURE 150-8. Perfused multilumen probe for balloon reflex manometric study. The balloon, positioned 10 to 15 cm above the anal verge, is capable of distending through at least 60 mL. Orifices 1 and 2 record intralumenal rectal pressure. Orifices 3 through 8 are located within the sphincter at 0.5-cm intervals. (From Coller JA. Clinical application of anorectal manometry. Gastroenterol Clin North Am 1987;16:27.)

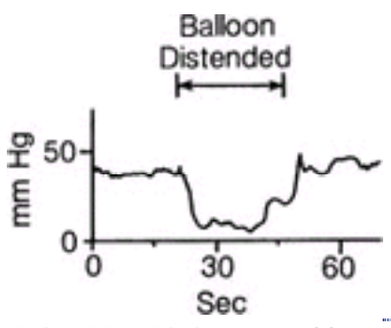

FIGURE 150-9. Normal internal anal sphincter relaxation response to balloon distention in the rectum. The balloon was located 15 cm above the anal verge and distended to 60 mL for the time shown. (Courtesy of Jeffrey L. Barnett, M.D., Ann Arbor, MI.)

Indications and Contraindications

Most patients complain of constipation or incontinence. A number of problems can be assessed adequately on the basis of clinical history, physical examination, and routine laboratory tests. Examples include irritable bowel syndrome, maldigestion, and thyroid dysfunction. A specific example involving the anal sphincter mechanism in which anorectal manometry would not be indicated is the presence of an acute anal fissure. This condition is associated with a deviation from the normal mean resting anal sphincter pressure,[135] but performance of manometry would not contribute any useful information beyond that obtained from the physical examination. Use of the examiner's finger as a pressure probe is highly inaccurate compared with objective measurements of sphincter function.[136]

When constipation is associated with the perception of increased pelvic or rectal pressure, incomplete evacuation, or difficulty initiating defecation, there is an increased likelihood that anorectal manometry may be a useful diagnostic aid. On the other hand, if resting sphincter tone appears normal or elevated in the presence of incontinence, balloon reflex manometry is indicated to evaluate the possibility of decreased threshold or increased sensitivity to rectal distention.

Data Interpretation

Manometric studies of the anal canal are typically plotted on an X-Y recorder, which presents a visual comparison of the various channels relative to one another. Unfortunately, subsequent quantitative analysis is often manually performed, which is laborious and time-consuming. However, software has been developed that uses an analog-to-digital converter to process and store data and to facilitate subsequent analysis of the pressure recordings. A typical microcomputer-generated tracing of the resting longitudinal anorectal sphincter pressure profile is depicted in Figure 150-10. The proximal end of the sphincter is that point at which the pressure is clearly above the intralumenal rectal pressure. This occurs when the perfusion orifices are withdrawn from the intralumenal rectum into the anal sphincter musculature. The normal sphincter length varies from 2.5 to 5 cm. A high-pressure zone is observed while the perfusion apparatus passes the distal portion of the internal sphincter muscle. The high-pressure zone is defined as that portion of the sphincter that demonstrates pressures greater than one half the maximal pressure. The point of maximal pressure is located in the distal portion of the high-pressure zone, about 1 to 1.5 cm from the distal end of the sphincter musculature. The normal average maximal resting pressure in the adult ranges from 65 to 85 mm Hg above rectal intralumenal pressure when measured with a perfused multichannel probe using the continuous pull-out technique. Usually, a modest lowering of the resting pressure occurs with aging.[137]

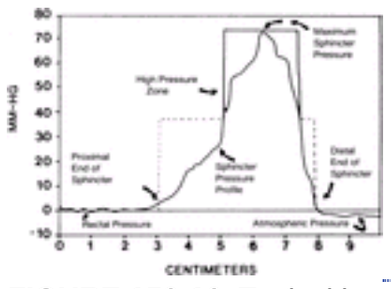

FIGURE 150-10. Typical longitudinal pressure profile of the resting anorectal sphincter. Pressures have been equated to a rectal pressure of zero. The pressures from an eight-channel multilumen probe during continuous resting pull-out have been averaged at each point along the sphincter by microcomputer. (From Coller JA. Clinical application of anorectal manometry. Gastroenterol Clin North Am 1987;16:20.)

Recent studies evaluating the longitudinal pressure profile of the anal canal with multichannel, radially positioned probes reveal evidence for radial pressure asymmetry. In the proximal sphincter, the posterior quadrant exhibits pressure predominance, whereas in the distal sphincter, the anterior quadrant demonstrates predominance.[138] Therefore, the longitudinal pressure profile reveals that the resting sphincter is a somewhat distorted cylinder with a gentle gradient of pressure, which shifts from posterior to anterior with movement from the rostral to the caudal end of the sphincter.

In addition to the resting sphincter pressure profile, patients undergoing anorectal manometry often have the maximal voluntary squeeze pressure evaluated. This test

is thought to reflect the contribution of the striated external sphincter. A voluntary squeeze should produce an increase of 50% to 100% over the average maximal resting pressure. Absolute normal values or ranges for voluntary squeeze pressure have not been agreed on in the literature, although women tend to have lower values than men. [137] The point of maximal pressure is located in the distal portion of the sphincter, similar to the status at rest. When evaluating external sphincter function with the voluntary squeeze maneuver, several technical issues should be kept in mind. The examiner must clearly tell the patient what is required to perform a voluntary squeeze. This is best achieved by placing a finger into the anal canal and instructing the patient regarding the appropriate voluntary squeeze effort. The squeezing effort results in movement of the anal canal because of the involvement of pelvic musculature; therefore, the probe should move freely with the sphincter to maintain the same position within the canal, which allows accurate comparison of resting and voluntary squeeze pressure at the same point. The external sphincter can sustain a voluntary squeeze for about 1 minute, after which fatigue occurs; therefore, the probe should be withdrawn within 30 to 40 seconds to obtain an accurate indication of the longitudinal pressure profile during voluntary squeeze.

Clinical Applications

Anorectal manometry is useful in the initial evaluation of patients with constipation and in the person with incontinence. The technique also may be useful to monitor progression of a disorder that affects anal sphincter function or the response to therapeutic interventions, such as biofeedback in the appropriate patient with incontinence.

Constipation When anorectal manometry reveals a normal or high average maximal resting pressure, the examiner should proceed with balloon reflex manometry to evaluate whether the internal sphincter relaxes in response to rectal distention. If rectal distention fails to initiate relaxation of the internal sphincter, the diagnosis of Hirschsprung disease should be considered [139] (Fig. 150-11). If the inhibitory reflex is present but demonstrates a high threshold for initiation, a problem with rectal compliance or afferent (sensory) pathways may be present. This pattern is observed in some patients with acquired megacolon or spinal cord injuries. [140] If the patient shows a deficient increase in voluntary squeeze pressure, abnormalities of sacral root innervation to the external sphincter should be considered. [141], [142]

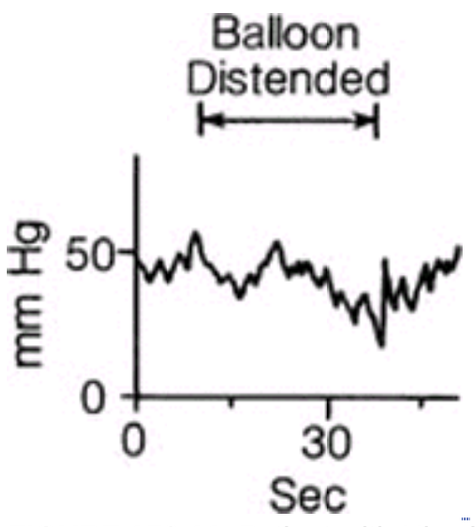

FIGURE 150-11. Patient with Hirschsprung disease. Tracing depicts failure of normal internal anal sphincter relaxation in response to rectal balloon distention. The balloon is positioned 15 cm above the anal verge and distended to 60 mL for the time shown. (Courtesy of Jeffrey L. Barnett, M.D., Ann Arbor, MI.)

Some patients with constipation demonstrate a low-average maximal resting pressure and normal rectal distention-mediated inhibition of the internal sphincter. In this scenario, the examiner should consider other anatomic abnormalities in the anorectal mechanism, such as accentuated puborectalis angulation or internal rectal prolapse, to explain the defecation problem. In addition, the constipated patient with low resting sphincter pressure deserves special mention because of the problems these patients face if an inappropriate sphincterotomy is performed. This procedure could potentially leave the patient with persistent constipation and iatrogenic loss of anal sphincter control.

Incontinence Assessment of resting and sphincter pressures, voluntary squeeze response, and balloon reflex manometry can be used to identify the location of abnormalities in anorectal function associated with incontinence. These findings can be helpful in directing therapy. For example, when the incontinent patient exhibits normal or elevated resting sphincter pressures, balloon reflex testing should be performed to rule out decreased threshold or increased sensitivity to rectal distention. Demonstration of these abnormalities suggests that a problem in rectal compliance underlies the incontinence. The typical patient with incontinence related to anorectal dysfunction and no history of trauma has low resting anal sphincter pressures. The voluntary squeeze response can help localize a problem in anal sphincter control to the internal or external sphincter. If a normal increase in tone is noticed with voluntary squeeze, this suggests that the problem resides in the internal sphincter, which can occur after hemorrhoid surgery or with diseases that affect the autonomic nervous system or smooth muscle cell function, such as diabetes mellitus and progressive systemic sclerosis. [143], [144] If a diminished or absent response to voluntary squeeze is observed, dysfunction of the external sphincter should be considered. This can be seen in patients with a neuropathy secondary to long-standing defecatory straining, sacral nerve impairment, and disorders affecting striated muscle. [145], [146] In patients with idiopathic fecal incontinence, documentation of increased pudenal nerve terminal motor latency is supportive that a neuropathy is present. However, a normal pudenal nerve terminal motor latency does not necessarily correlate with abnormalities in maximum squeeze pressure because this measurement does not exclude weakness of the pelvic floor. [147] Patients who receive radiation therapy for localized carcinoma of the prostate can demonstrate chronic heightened rectal sensitivity for at least 1 year after completion of therapy. [148], [149]

FUTURE APPLICATIONS

Novel technologies to evaluate GI neuromuscular function are being evaluated on a continuing basis. Manometry and barostat studies are invasive, technically challenging, and costly, which reduces their widespread clinical utility. Novel approaches to study gastric motility include magnetic resonance imaging (MRI), single photon emission computed tomography (SPECT), ultrasonography, tensostat, water and nutrient drink test, paracetamol absorption test, and ^{13}C-octanoic acid breath test. [150] Each of these tests has strengths and limitations. For example, the water and nutrient test has been proposed as a noninvasive alternative to sensory (satiety) testing with an intragastric balloon. The tensostat measures gastric wall tension that correlates with the perception of gastric distention. The paracetamol absorption test, ^{13}C breath tests, ultrasound, and MRI have been used to assess gastric emptying. MRI requires expensive equipment and ultrasound advanced training to perform and interpret the results. SPECT has been employed to assess gastric accommodation to a meal. This technology requires intravenous injection of 99mTc-pertechnetate and specialized imaging and analysis capability. Each of these technologies requires validation of their utility in the research and clinical settings.

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CHAPTER 151

William D. Chey and William Y. Chey

EVALUATION OF SECRETION AND ABSORPTION FUNCTIONS OF THE GASTROINTESTINAL TRACT

This chapter reviews the available gastric secretory function tests, pancreatic exocrine function tests, and breath tests for carbohydrate malabsorption or bacterial overgrowth. No attempt is made to review the physiology of gastric secretory and pancreatic exocrine function; instead, for detailed discussions of these topics, see Chapter 13 and Chapter 15.

TESTS OF GASTRIC SECRETORY FUNCTION

The clinical applications of gastric acid analysis and prolonged intragastric pH monitoring are limited. However, these tests remain critical for better understanding the effects of various disease states and pharmaceuticals on gastric secretory function. Gastric acid analysis and prolonged intragastric pH monitoring have played a pivotal role in our understanding of the link between *Helicobacter pylori* infection and changes in acid secretion documented in patients with peptic ulcer disease decades ago.[1] As the *H pylori* story has taught us, groundbreaking discoveries of the future will require an ongoing reevaluation of our understanding of gastric secretory function. As such, the methodology, strengths, and weaknesses of gastric acid analysis and prolonged intragastric pH monitoring continue to be worthy of the gastroenterologist's understanding.

Gastric Acid Analysis

Indications Traditionally, physicians have used gastric acid analysis to answer the following questions: (1) Does the stomach produce acid? (2) If so, what is the basal or unstimulated gastric acid secretion? (3) How much acid is secreted in response to physiologic or pharmacological stimulants? (4) Is the vagal innervation to the stomach intact?[2] In the past, gastric acid analysis served as an important tool in the evaluation of patients with a large number of disorders affecting the stomach. With the development of more effective diagnostic and therapeutic modalities for acid peptic disorders, however, the importance of tests to measure gastric acid secretion has decreased substantially. Gastric acid analysis remains the gold standard to evaluate gastric secretory function and is useful to today's gastroenterologist or gastrointestinal surgeon in several clinical situations (Table 151-1). Gastric acid analysis can be helpful in the evaluation of patients who develop recurrent ulcers after surgery for peptic ulcer disease. In this setting, gastric acid secretion can be quantified and hypersecretion excluded. In addition, the completeness of vagotomy can be determined by sham feeding that evaluates the vagally mediated cephalic phase of acid secretion. In the age of *H pylori* and proton pump inhibitors, however, the role of gastric analysis in the patient with postoperative ulcer recurrence will have to be redefined. With more effective medical therapies, fewer patients have chronic or recurrent peptic ulcer disease. Thus, fewer patients require operative intervention for this indication. In addition, most of the classic literature addressing postoperative ulcer recurrence was performed before the acceptance of *H pylori* as one of the major pathogenic factors in the development of peptic ulcer disease.[3] It is possible that *H pylori* eradication may alter postoperative gastric acid secretion or decrease the likelihood of peptic ulceration after surgery. As such, in the age of *H pylori,* it is unclear whether gastric acid analysis will retain the same importance in postoperative patients with ulcer recurrence.

I. Assess the patient with ulcer recurrence after surgery for peptic ulcer disease
a. Exclude acid hypersecretion
b. Assess the completeness of vagotomy (sham feeding)
II. Evaluate an elevated fasting plasma gastrin level
a. Assess physiologic response (atrophic gastritis)
b. Assess pathological response (gastrinoma, ulcerogenic tumor syndrome with nongastrin secretagogue)
III. Assess the adequacy of proton pump inhibitor therapy in patients with diseases of gastric acid hypersecretion

TABLE 151-1 Indications for Gastric Acid Analysis

Gastric analysis can help determine whether an elevated circulating gastrin level is physiologic (achlorhydria) or pathological (acid hypersecretory states). Gastric analysis can establish that hypergastrinemia is an appropriate consequence of the inability to produce acid, as seen in the setting of atrophy, pernicious anemia, or malignancy involving the body of the stomach.[4],[5] On the other hand, hypergastrinemia can be seen in diseases of excessive acid secretion such as Zollinger-Ellison syndrome.[6] The development of secretin stimulation testing and accurate imaging modalities, such as high-resolution computed tomography (CT), endoscopic ultrasound (EUS), and somatostatin receptor scintigraphy, has relegated gastric acid analysis to an adjunctive role in the diagnosis of this disease.[7],[8] Gastric analysis remains critical to the evaluation of the unusual patient with acid hypersecretion attributable to the ulcerogenic tumor syndrome, in which circulating gastrin levels are typically normal.[9],[10] Gastric analysis can be useful in determining the adequacy of acid suppressive therapy after the diagnosis of gastrinoma or the ulcerogenic tumor syndrome has been established. Gastric acid analysis remains an important means of better understanding the effect of various disease states and pharmaceuticals on gastric secretory function. For example, it has long been known that patients with duodenal ulcer have higher levels of basal acid secretion than controls.[11],[12] Studies using gastric acid analysis have shown that this abnormality is related to *H pylori* infection and that eradication of this infection leads to normalization of basal acid output (BAO). On the other hand, pentagastrin stimulated maximal acid output (MAO) in patients with duodenal ulcer remains elevated even after successful *H pylori* eradication. This suggests the presence of parietal cell hypersensitivity to gastrin or an increase in parietal cell mass that persists after eradication of *H pylori* infection.[1] As opposed to patients with duodenal ulcer, healthy volunteers with *H pylori* infection can have normal, enhanced, or diminished acid secretion.[13],[14] Investigators have also recently used gastric analysis to prove that proton pump inhibitors and histamine H_2 receptor antagonists more effectively suppress gastric acid secretion in individuals with *H pylori* infection than in noninfected individuals.[15],[16] It is now clear that gastric analysis does not accurately differentiate between functional versus ulcer-related dyspepsia because the overlap in acid secretion between the two groups is substantial[11],[12] (Fig. 151-1). There are a number of situations in which the role of gastric analysis has not been clearly defined. For example, the utility of gastric acid analysis in patients with peptic ulcer disease, but no evidence of *H pylori* infection or history of nonsteroidal antiinflammatory drug use, has yet to be determined. Preliminary data suggest that acid secretion may be abnormal in such patients.[17] Other patient populations in whom gastric acid analysis might be useful include those with a nonhealing ulcer despite appropriate acid-suppressive therapy and those with ulcer recurrence despite successful *H pylori* eradication.

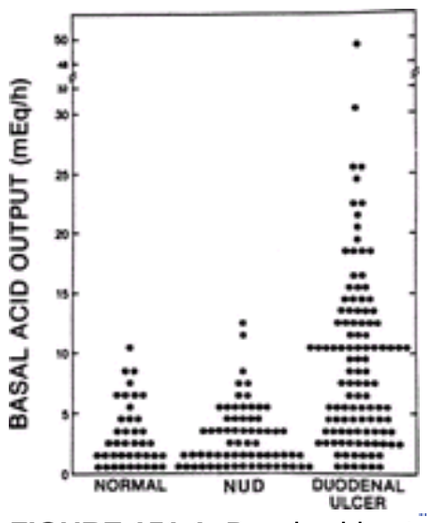

FIGURE 151-1. Basal acid output (BAO) in 40 healthy controls, 66 patients with nonulcer dyspepsia (*NUD*), and 114 patients with duodenal ulcer. There was no difference in mean BAO between normals (3.2 ± 2.7 mEq/h) and patients with NUD (2.9 ± 2.7 mEq/h). However, the mean BAO was significantly higher in patients with duodenal ulcer (9.1 ± 7.6 mEq/h) than in either normals or patients with NUD. (From ref. [11].)

Methodology The most widely practiced means of measuring acid secretion is the aspiration method in which gastric contents are withdrawn through a nasogastric tube at timed intervals under different test conditions. The hydrogen ion concentration in these samples can then be determined providing a quantitative assessment of basal (BAO) and stimulated (MAO, modified sham feeding) acid secretion. A point worthy of consideration is the reliability of gastric acid analysis. A great deal of variability in test results has been observed in studies of healthy individuals. Both BAO and MAO tend to be higher in males than females. Patients with duodenal ulcer tend to have greater acid secretion than healthy individuals [1], [11], [12] (see Fig. 151-1). Because of considerable overlap between populations, however, gastric analysis does not reliably discriminate between those with peptic ulcer disease and healthy volunteers. Even when Zollinger-Ellison syndrome is suspected, gastric analysis is not always a reliable single diagnostic test because of the overlap in results yielded by patients and controls. [4] Aside from the considerable interindividual variability, there is also intraindividual variability when gastric acid analysis is performed on different days. [18] Reproducibility of this test is dependent on several factors, including day-to-day variations in BAO, day-to-day variations in responsiveness of acid output to various secretagogues, and the completeness of gastric contents recovery. A review article reported a relatively poor coefficient of variation for intraindividual BAO data. [18] This is likely related to the multiple factors (time of day, mental state, duodenal losses of gastric contents, neutralization of acid with swallowed saliva, and neurohormonal effectors of acid secretion) that influence acid secretion and its measurement in the basal state. [19] In contrast to basal acid secretion, acid secretion stimulated by histamine or pentagastrin is much more reproducible from day to day. [18] The recovery of gastric contents can be facilitated by proper tube placement and patient position. The aspiration tube should be of sufficient caliber to allow recovery of the gastric contents but not so large as to be intolerable to the patient. In general, tubes in the range of 14 to 18 French fulfill these requirements. Testing should be performed in the seated position. The tip of the tube should be placed into the most dependent portion of the stomach, and proper placement should be confirmed with fluoroscopy. Even when test conditions are optimized, recovery of the gastric contents is often not complete. [20] The recovery of gastric contents during aspiration can be quantified by the infusion of a nonabsorbable marker such as polyethylene glycol or radiolabeled chromic chloride ($^{51}CrCl_3$). [2], [20] Although this is the ideal means of performing gastric acid analysis, the infusion and measurement of a nonabsorbable marker, particularly one that is radioactive, makes the procedure much more cumbersome and costly to perform. For this reason, most centers, including our center, perform gastric acid analysis without the infusion of a nonabsorbable marker.

Basal acid output. On the evening before undergoing gastric acid analysis, the patient should take a clear liquid dinner and consume nothing by mouth after midnight. Unless the purpose of the study is to assess the effect of a medication on acid secretion, dosing of agents that affect acid secretion should be modified before the test. Histamine H_2 receptor antagonists, antihistamines, anticholinergics, and cholinergic agonists should be discontinued at least 24 hours before gastric acid analysis. Proton pump inhibitors inactivate the hydrogen-potassium ATPase found on the parietal cell. [21] As such, proton pump inhibitors have prolonged effects on acid secretion [22] and should be withheld for at least 7 days before testing. For patients who suffer with recurrent symptoms once their proton pump inhibitor has been discontinued, a histamine H_2 receptor antagonist may be used until 24 hours before gastric acid analysis. On the day of gastric acid analysis, the patient is intubated, and tube placement is confirmed as described earlier. The study starts with a 30-minute washout period during which all gastric contents are aspirated and discarded. After this washout period, samples are collected at 15-minute intervals. Sample volume is recorded, and an aliquot is used to determine the titratable acidity. Titratable acidity (millimoles [mmol] per liter) is determined by adding a base, such as NaOH, to an aliquot of gastric juice until the pH reaches 7.0. Values are then corrected for the size of the total timed sample. The following equations can be used to determine the titratable acidity:

$$(\text{mL base to titrate to pH 7.0}) \times [\text{concentration of the base}] = \text{millimoles } H^+ \text{ per aliquot}$$

$$\frac{\text{volume of sample}}{\text{volume of aliquot}} \times \text{millimoles } H^+ \text{ per aliquot} = \text{millimoles } H^+ \text{ per sample}$$

BAO (millimoles H^+ per hour) is the sum of the acid output from the four 15-minute collections done in the unstimulated state. In healthy unoperated volunteers, BAO should be less than 15 mmol H^+ per hour. A BAO of greater than 5 mmol H^+ per hour is considered abnormal for a patient who has previously undergone partial gastrectomy or vagotomy. [4] In humans, BAO should be about 10% of the maximal secretory capacity of the stomach. [18]

Maximal and peak acid output. MAO and peak acid output (PAO) indirectly assess functional parietal cell mass through the measurement of acid secretion after the administration of maximally effective doses of agents that directly stimulate the parietal cell. [23], [24] Such stimulants of acid secretion include pentagastrin, histamine, and histamine analogs such as 3-ß-aminoethylpyrazole. Pentagastrin is the most widely used agonist and is usually administered subcutaneously (6 µg/kg). Pentagastrin can also be given intramuscularly (6 µg/kg) or intravenously (1 to 6 µg/kg/h). After pentagastrin administration, gastric juice is collected every 15 minutes for 1 to 1.5 hours. The titratable acidity (millimoles H^+ per sample) is then determined for each of the four to six samples obtained after pentagastrin administration. MAO represents the greatest sum of the titratable acid from four consecutive samples (millimoles H^+ per hour). PAO is the sum of the two highest acid outputs multiplied by two to yield results in millimoles H^+ per hour. MAO and PAO measurements vary widely in healthy subjects, ranging from 0 to 80 mmol H^+ per hour. MAO is influenced by a number of variables, including sex, age, and lean body mass. [25] MAO decreases by about 50% after truncal or proximal gastric vagotomy. [26], [27] In conjunction with the BAO, MAO measurement can suggest the presence of the ulcerogenic tumor syndrome, including gastrinoma. Because the presence of an elevated plasma gastrin concentration stimulates gastric acid output by parietal cells in the basal state, patients with gastrinoma typically have a BAO-to-MAO ratio of greater than 0.6. [4] Another approach to the study of gastric secretory function is to administer gastrin-releasing peptide (GRP) rather than pentagastrin or histamine. [1] GRP leads to the release of gastrin from the antral G cell and thereby indirectly stimulates gastric acid secretion by the parietal cell. [28], [29] In addition, GRP influences the release of counterregulatory hormones, such as cholecystokinin (CCK), secretin, gastric inhibitory peptide, vasoactive intestinal peptide, neurotensin, enteroglucagon, and somatostatin. [30], [31] and [32] Therefore, unlike pentagastrin or histamine, which assess only the parietal cell response to direct stimulation, GRP theoretically evaluates the sum of both stimulatory and inhibitory pathways responsible for the control of acid secretion. Investigators have used this approach to demonstrate abnormalities in the inhibitory control of acid secretion in patients with *H pylori* infection and duodenal ulcer. [1] At the current time, GRP is an interesting clinical investigative tool but cannot be considered part of routine gastric acid analysis.

Testing of vagally stimulated acid secretion. Since the turn of the 20th century, it has been known that the thought, sight, smell, and taste of food stimulate gastric acid secretion. Food-induced stimulation of the senses leads to activation of cerebral cortical sites, resulting in increased gastric acid secretion. The cephalic phase of gastric acid secretion is dependent on an intact vagus nerve and is cholinergically mediated. [18] In the past, vagally mediated acid secretion was assessed after insulin-induced hypoglycemia (Hollander test). [33] The absence of an increase in acid secretion with hypoglycemia suggested the presence of vagotomy. Unfortunately, this test was associated with a number of severe adverse reactions, including myocardial infarction, seizures, and, on rare occasions, death. [34], [35] For this reason, the Hollander test has been replaced by the sham feeding test. Most commonly, the sham feeding test is used to determine the completeness of vagotomy in patients with ulcer recurrence after surgery for peptic ulcer disease. [36] In humans, acid output in response to sham feeding is 50% to 65% of secretagogue-induced acid output. [18] In addition, some investigators have used this technique to assess the patency of vagal autonomic innervation to the stomach in the patients with diabetes mellitus. [37] During sham feeding, patients are allowed to see, smell, taste, and chew an appetizing meal. Food is then expectorated by the patient, thus avoiding the gastric phase of acid secretion. Gastric juice is collected through a nasogastric tube, and the titratable acidity is calculated as described previously. Feldman and colleagues [37] have shown that a sham acid output (SAO)-to-PAO ratio of less than 0.1 is abnormal in healthy volunteers. Therefore, an SAO-to-PAO ratio of more than 0.1 in a patient who has previously undergone peptic ulcer surgery suggests the presence of intact vagal innervation to the stomach. [36]

Ambulatory Intragastric pH Monitoring

Indications The role of ambulatory intragastric pH monitoring in clinical practice is limited. Like gastric acid analysis, however, the prolonged measurement of intragastric pH is an important clinical research tool. Prolonged intragastric pH monitoring has gained popularity as an investigative tool because it is better tolerated by patients, is less cumbersome to perform, and arguably gives more physiologic information than traditional gastric analysis. This technique has most often been used to evaluate the effects of duodenogastric reflux [38], [39] and drugs that suppress acid secretion (e.g., histamine H_2 receptor antagonists and proton pump inhibitors) on intragastric pH. [40], [41] Intragastric pH monitoring can also help to establish a link between pH events and gastrointestinal symptoms. [41] Recently, differences between patients with duodenal ulcer associated with *H pylori* infection and controls were demonstrated using ambulatory intragastric pH monitoring. [42], [43] Intragastric pH monitoring has also been used to identify differences in the effectiveness of proton pump inhibitor therapy between those infected and those not infected with *H pylori*. [44], [45] It is important to understand the differences in information yielded by gastric acid analysis and intragastric pH monitoring. Although gastric acid analysis provides a quantitative measure of gastric secretory function, intragastric pH monitoring yields only qualitative information. As has already been discussed, gastric acid analysis quantitates basal and maximal acid secretion in response to the administration of a specific secretagogue. On the other hand, intragastric pH monitoring allows only the determination of pH, albeit in a more physiologic setting than gastric acid analysis. How well results obtained with prolonged intragastric measurement correlate with traditional gastric acid analysis remains controversial. Several studies have eported a good correlation between data obtained from the two techniques. [40], [46] However, studies have questioned the validity of drawing conclusions regarding overall intragastric pH solely on the basis of measurements obtained with a single electrode. [39], [47], [48] and [49] Simultaneous recordings from the body and antrum have demonstrated significant differences in pH both in the interdigestive and postprandial periods. [39], [47], [48] and [49] In general, H^+ concentration is higher when measured by intragastric pH monitoring compared with gastric acid analysis. [48] The precise reasons for these differences are not clear, but biophysical phenomena, including environmental carbon dioxide (CO_2) concentration, temperature, and technical problems, such as electrode instability, artifact, or interference, may play a role. [48], [50]

Methodology There are a variety of ways to measure intragastric pH. The most widely employed method has been to place a catheter containing a single electrode transnasally into the body of the stomach. [51], [52] As mentioned previously, recent literature has questioned the practice of extrapolating data from a single electrode to the stomach as a whole. Obtaining pH measurements from more than one part of the stomach appears to yield more complete information. Whether this more complete information has practical relevance to the clinician or investigator remains to be determined. Proper placement should be confirmed fluoroscopically. Both antimony and glass pH electrodes have been used and appear to give equivalent results. [53] The catheter is connected to any of a number of commercially available computerized modules that can digitally record changes in intragastric pH. Careful attention should be paid to proper calibration of the pH monitoring equipment before each study. The probes used for intragastric pH monitoring reportedly become less accurate with repeated use. [50] Data are often expressed as the percentage of time above or below a certain pH. This means of data presentation is simple and easy to understand. However, differences in gastric secretory function are more difficult to appreciate using this means of data presentation because pH is the inverse log of H^+ concentration. As such, it is more precise to express data from intragastric pH monitoring as H^+ concentration. [48], [54]

TESTS OF PANCREATIC EXOCRINE FUNCTION

Traditionally, it has proved challenging to identify patients with mild to moderate chronic pancreatitis. The use of endoscopic retrograde pancreatography (ERP), CT, transcutaneous ultrasonography, and, more recently, EUS has facilitated the diagnosis of chronic pancreatic diseases. Unfortunately, despite evaluation with structural studies, a definitive diagnosis will not be apparent in a subset of patients with chronic pancreatitis. In addition, structural studies cannot quantitatively assess the degree of exocrine dysfunction. Although local expertise and availability influence the choice of diagnostic studies, functional testing can uncover previously unidentified abnormalities or complement information yielded by structural studies.

Tests of pancreatic exocrine function can be divided into four categories: (1) tests that quantitate pancreatic secretion through gastroduodenal intubation; (2) studies that assess exocrine function indirectly using an orally administered test substrate; (3) tests that rely on the measurement of fat or pancreatic enzyme activity in feces; and (4) a miscellaneous group of tests, including the measurement of serum pancreatic enzyme activity, plasma pancreatic polypeptide response to ingestion of a meal, and changes in plasma amino acid concentration after stimulation of the pancreas (Table 151-2).

Tests Requiring Gastroduodenal Intubation
Direct tests
Secretin test
Secretin-cholecystokinin (CCK) or CCK-8 test
CCK-8 or cerulein test
Indirect tests
Lundh test
Tubeless Pancreatic Function Tests
Oral tests
N-benzoyl-L-tyrosyl-paraamino benzoic acid (NBT-PABA) test
Fluorescein dilaurate (pancreolauryl) test
Dual-labeled Schilling test
Breath tests
Triolein and mixed triglyceride breath test
Histamine H_2 and [^{13}C]-labeled starch breath tests
Fecal tests
Fecal fat determination
Chymotrypsin
Lipase
Elastase
Miscellaneous tests
Serum trypsin-like immunoreactivity
Plasma pancreatic polypeptide test
Plasma amino acid consumption test

TABLE 151-2 Pancreatic Function Tests

Tests Requiring Gastroduodenal Intubation

These tests rely on the collection of pancreatic juice from the duodenum before and after the administration of secretagogues including secretin or CCK or its analogs (e.g., CCK-8, pancreozymin, cerulein). Tests that measure pancreatic secretion from duodenal contents provide an accurate, sensitive (67% to 90%), and specific (80% to 90%) means of assessing pancreatic exocrine function. [55], [56], [57], [58] and [59] These tests may be more sensitive in detecting pancreatic dysfunction than traditional imaging techniques including ERP [60] but are probably not as sensitive as EUS. [61] Although these tests are clearly superior to the so-called tubeless pancreatic function tests, they are labor intensive, expensive, and require specially trained personnel. As such, they are performed almost exclusively in large referral centers and research laboratories.

Secretin and Cholecystokinin Tests The secretin test was developed in the early part of the 20th century [55], [56] to measure exocrine pancreatic secretion in response to the intravenous injection of purified porcine secretin. The aspirate volume as well as the concentration of bicarbonate and pancreatic amylase [56], [57] are measured before and after the administration of secretin. To minimize contamination of the duodenal aspirate by gastric juice, a double-lumen tube should be used. With fluoroscopic guidance, gastric aspiration ports are positioned in the antrum and body while duodenal aspiration ports lie in the first, second, and third portion of the duodenum (Fig. 151-2). Gastric juice and duodenal contents are separately collected by continuous suction. Gastric contents are discarded while duodenal contents are retained for analysis.

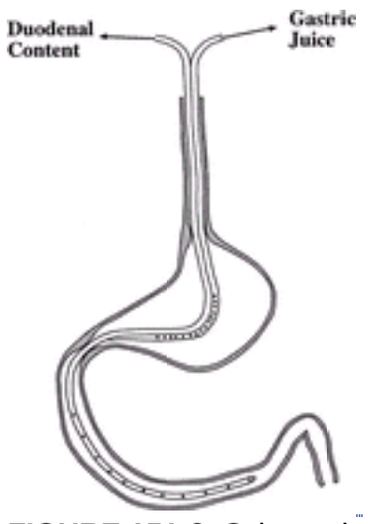

FIGURE 151-2. Schematic representation of the set-up for pancreatic function tests requiring gastroduodenal intubation. A double-lumen tube is placed into the stomach and duodenum with the assistance of fluoroscopy. Gastric and duodenal contents are then simultaneously aspirated during the basal state and after intravenous administration of pancreatic secretagogues, including secretin, cholecystokinin-8, or cerulein alone or in combination. Gastric contents are discarded, whereas duodenal contents are stored for subsequent analysis as described in the text.

Gastric aspiration is important because contamination of duodenal contents with gastric acid produces falsely low bicarbonate concentration and inaccurate volume measurements. Moreover, acid stimulates the release of secretin from the duodenal mucosa, which can affect test results. The pH of uncontaminated duodenal fluid should be greater than 7.5. In clinical conditions associated with gastric acid hypersecretion such as Zollinger-Ellison syndrome, the collection of uncontaminated duodenal fluid is virtually impossible. Once the tube is in place, duodenal fluid is collected every 15 minutes for 1 hour. Secretin in a dose of one clinical unit per kilogram of weight is administered as a bolus intravenous injection, and duodenal fluid is collected for an additional 1 to 1.5 hours. Except in patients with gallbladder dysfunction and those who have previously undergone cholecystectomy, the duodenal aspirate should lose its bile-stained appearance, becoming clear and colorless, after secretin injection. [59] Normal values for duodenal fluid after secretin injection are provided in Table 151-3. In patients with chronic pancreatitis, one of the four following patterns can be seen: (1) low maximum concentration of bicarbonate only; (2) low enzyme output only; (3) low bicarbonate concentration and low enzyme output; and (4) abnormally low volume, bicarbonate concentration, and amylase output. It is difficult to judge the significance of an isolated abnormality in duodenal juice volume because the adequacy of aspiration is often unclear. To avoid this technical error, a nonabsorbable marker can be infused into the proximal duodenum during the course of the study. Quantification of the marker in the samples collected allows determination of the completeness of duodenal aspiration. [62] When low volume is truly the only abnormality found, the clinician should consider the possibility of cancer in the head of the pancreas. When abnormalities in volume, bicarbonate concentration, and amylase output are all present, patients virtually always exhibit signs of advanced pancreatic disease, such as extensive calcification, steatorrhea, or pancreatic endocrine dysfunction. [63] The secretin test is sensitive enough to detect the presence of mild to moderate chronic pancreatitis. In such patients, low bicarbonate concentration may be the only abnormality. At the time of this writing, secretin is not commercially available in the United States. Recombinant human secretin is under review by the U.S. Food and Drug Administration. It is hoped that this agent will become available in the near future.

1. Secretin test* (mean ± 2 SD)	
Volume: 107–223 (mL/h)	
Bicarbonate concentration: 92–124 (mEq/L)	
Bicarbonate output: 14–21 (mmol/h)	
Amylase output: 174–1270 (U/h)	
2. Secretin + CCK (lower limits of normal)†	
Volume: 151 (mL/h)	
Bicarbonate concentration: 70 (mmol/L)	
Bicarbonate output: 11.3 (mmol/h)	
Amylase output: 131 (units during 30 min of CCK)	
3. CCK test‡ (range in 16 control subjects)	
Trypsin output: 25–54 (Ku/h)	
Lipase output: 77–322 (Ku/h)	
4. Lundh test§ (mean ± 2SD)	
Mean tryptic activity: 7–38 (IU/L)	

*Data from Dreiling DA, Hollander F. Studies in pancreatic function. II. A statistical study of pancreatic secretion following secretin in patients without pancreatic disease. Gastroenterology 1950;15:620.

†Data from Howat HT, Braganza JM. Assessment of pancreatic dysfunction in man. In: Howat HT, Sarles H, eds. The exocrine pancreas. Philadelphia: WB Saunders, 1979:129.

‡Data from ref. 65.

§Adapted from ref. 70.

CCK, cholecystokinin; Ku, kilounits; SD, standard deviation

TABLE 151-3 Normal Ranges for Results of Pancreatic Function Tests Requiring Gastroduodenal Intubation

Using the basic technique described previously, testing can be modified by adding the intravenous injection of CCK [58], [59] or its analogs. [64] However, there is no convincing evidence to suggest that the additional administration of CCK improves the sensitivity or specificity of the secretin test. Alternatively, some investigators have used CCK alone to stimulate pancreatic enzyme output. [65] Whenever CCK is used, measurements of trypsin and lipase output are performed. Normal values for the secretin and CCK tests are given in Table 151-3.

Lundh Test The Lundh test determines the pancreatic enzyme concentration of the duodenal contents in response to a test meal. After fluoroscopically guided placement of a catheter into the duodenum, the test subject ingests a standardized 300-mL meal composed of dried milk, vegetable oil, and dextrose. [66] Duodenal contents are aspirated at set intervals for measurement of pancreatic enzyme concentration. When chymotrypsin is measured, a concentration below 12.6 IU suggests the presence of pancreatic exocrine insufficiency. Because this test relies on secretin and CCK-stimulated pancreatic secretion and these hormones are released mainly from the duodenal mucosa, it is necessary for patients to have gastroduodenal continuity and normal duodenal mucosa. [67], [68] Although this test is simpler and less expensive than the secretin test, it allows determination of enzyme concentration only. The sensitivity of this test ranges from 66% to 94%. [69], [70] The secretin or CCK tests appear to be more sensitive than the Lundh test, particularly in patients without steatorrhea. [71], [72]

Tests that Do Not Require Gastroduodenal Intubation

Like studies that identify structural pancreatic disease, all of the tubeless pancreatic function tests are reasonably sensitive and specific in patients with severe pancreatic exocrine dysfunction. Unfortunately, the tubeless tests have yielded only marginal sensitivity and specificity in patients in whom an accurate test is most needed: those with mild to moderate exocrine dysfunction. Furthermore, recent data suggest that combining tubeless tests does little to improve their accuracy in patients with mild to moderate disease. [73] Because of their inadequacies, these tests have not gained wide acceptance in clinical practice. General information regarding the oral, fecal, and miscellaneous pancreatic function tests is provided in Table 151-4.

	WHAT IS MEASURED	SOURCE
Oral Tests		
NBT-PABA test	PABA	Urine, serum
Pancreolauryl test	Fluorescein	Urine, serum
Dual-labeled Schilling test	[^{58}Co]/[^{57}Co]	Urine
Breath tests		
Triolein	[^{14}C]O_2	Breath
Mixed triglyceride breath test	[^{13}C]O_2	Breath
[^{13}C]-labeled starch breath tests	[^{13}C]O_2 or H_2	Breath
Fecal Tests		
Fecal fat determination	Fat	Feces
Chymotrypsin	Chymotrypsin	Feces
Lipase	Lipase	Feces
Elastase	Elastase	Feces
Miscellaneous Tests		
Serum trypsin-like immunoreactivity	Trypsin-like activity	Serum
Plasma pancreatic polypeptide test	Pancreatic polypeptide	Plasma
Plasma amino acid consumption test	Amino acid levels	Plasma

Note: All of the tubeless tests are substantially better able to identify patients with severe pancreatic exocrine dysfunction (steatorrhea present) than those with earlier stages of disease.

TABLE 151-4 Tubeless Pancreatic Function Tests

Oral Pancreatic Function Tests

N-Benzoyl-L-tyrosyl-paraamino benzoic acid test. The *N*-benzoyl-L-tyrosyl-paraamino benzoic acid (NBT-PABA) or bentiromide test [74], [75] is a simple and noninvasive test of pancreatic exocrine function. After oral administration, NBT-PABA, a synthetic peptide, is specifically cleaved by chymotrypsin to yield free PABA. A dose of 500 to 1000 mg appears to provide the best separation between healthy subjects and patients with chronic pancreatitis. [76], [77] The necessity of giving a test meal to stimulate pancreatic secretion with the NBT-PABA is controversial. PABA is absorbed in the small intestine, conjugated by the liver, and excreted in the urine. Thus, the quantitation of PABA and its metabolites in serum or urine provides an indirect measure of duodenal chymotrypsin activity. The reported sensitivity of the NBT-PABA test is between 37% and 90%. [77] The specificity of the test has also been variable (39% to 100%). Tests using a timed urine collection have not been as reliable as serum testing. Perhaps the most important factor influencing the sensitivity and specificity of this test has been the severity of pancreatic disease in the population studied. When data from several studies using the secretin-CCK test as a gold standard were pooled, the sensitivity of the NBT-PABA test with a 6-hour urine collection was 71% in patients with severe exocrine dysfunction but only 46% in those with mild to moderate insufficiency. [77] In addition, the measurement of PABA and other arylamines can be confounded by certain clinical conditions (gastrointestinal surgery, small bowel disease, diabetes mellitus, severe liver disease and renal insufficiency), drugs (acetaminophen, phenacetin, benzocaine, lidocaine, procaine, chloramphenicol, sulfonamides, sulfonylurea, and thiazides), and foods (prunes and cranberries). Pancreatic enzyme supplements can cause false-negative results and should be discontinued at least 5 days before the test. The specificity of the NBT-PABA test can be improved by administering a marker that allows the determination of individual variability in extrapancreatic factors such as gastric emptying and intestinal, hepatic, or renal handling of PABA. To these ends, it is possible to administer free PABA [78] on a separate day or to coadminister *p*-aminosalicylic acid, [79], [80] carbon-14 (^{14}C)-PABA, [81] or carbon-13 (^{13}C)-PABA [82] with NBT-PABA to assess better the effects of extrapancreatic factors on the test.

Urine test. If assessing urinary excretion, a 6-hour urine collection appears to yield the greatest sensitivity in patients with pancreatic exocrine dysfunction. The lower limit of normal for a 6-hour urine collection is 50% excretion of the dose administered.

Serum test. The measurement of serum PABA is simpler, more reliable, and more accurate in assessing pancreatic exocrine function than a timed urinary collection. [82], [83], [84], [85] and [86] Blood samples are obtained before and 150 minutes after NBT-PABA administration. [83] The reported cutoff value for PABA that discriminates between healthy controls and patients with chronic pancreatitis is 20 nmol/mL. Using this cutoff value, the sensitivity and specificity of this test are 70% to 90% and 87% to 95%, respectively. [83], [84] In a variation on this study that used ^{13}C-PABA as a marker, [82] both PABA and ^{13}C-PABA in blood were measured using a detailed analysis including hydrolysis, extraction, separation by high performance liquid chromatography (HPLC), and methyl ester formation of the test substances before chromatography and mass spectrometry. The modified protocol appeared to discriminate better between healthy volunteers and patients with pancreatic insufficiency.

Fluorescein dilaurate test (pancreolauryl test). The fluorescein dilaurate test [83], [87] is similar in principal to the NBT-PABA test. Fluorescein dilaurate, a synthetic, relatively water-insoluble ester, is given orally with a standardized breakfast. Fluorescein dilaurate is specifically hydrolyzed by pancreatic arylesterases to lauric acid and water-soluble, free fluorescein. Fluorescein is readily absorbed by the small intestine, conjugated in the liver, and excreted in the urine. Urine or serum measurements of fluorescein can be performed. The pancreolauryl test performs in a manner similar to the NBT-PABA test, with a sensitivity of 55% to 100% and a specificity of 46% to 97%. [77] Like the NBT-PABA test, the pancreolauryl test is more sensitive in patients with severe pancreatic exocrine insufficiency than mild to moderate disease. The accuracy of the test can be negatively influenced by previous gastrointestinal surgery, small bowel mucosal diseases (e.g., celiac disease, Crohn's disease), severe liver disease, or bacterial overgrowth. Pancreatic enzyme supplements should be discontinued at least 5 days before the study.

Urine test. Urinary fluorescein is measured spectrophotometrically in 10-hour collections on two separate days. The recovery of fluorescein on the test day (T) is expressed as a percentage of the recovery of free fluorescein during a control study (C): [T/C × 100]. A T/C ratio of less than 20 is considered abnormal, whereas results between 20% and 30% fall into the equivocal range. [83], [87]

Serum test. A modified serum test [88] consisting of a standard breakfast with two capsules of 0.5 mmol fluorescein dilaurate was studied in a large number of patients with chronic pancreatitis confirmed by endoscopic retrograde cholangiopancreatography. [89] Patients received simultaneous intravenous infusions of metoclopramide (10 mg) and secretin (1 clinical unit/kg). Their modified protocol achieved a more rapid peak in serum fluorescein concentration after the test meal and improved the sensitivity of the test in patients with moderate chronic pancreatitis. A value below 2.5 µg/mL was thought to represent the optimum cutoff value for this test. In this large group of patients with varying degrees of pancreatic disease, the modified test yielded a sensitivity of 82% and specificity of 91% for chronic pancreatitis. In those with mild to moderate chronic pancreatitis, the test had a sensitivity of 70%. Another recent study confirmed the value of secretin administration before the test meal. [90]

Dual-labeled schilling test. Cobalamin (vitamin B_{12}) malabsorption occurs in a subset of patients with exocrine pancreatic insufficiency. R protein is a nonintrinsic factor cobalamin-binding protein present in the saliva, gastric juice, and other body fluids. Brugge and colleagues [91] reported a modified dual-labeled Schilling test that indirectly evaluates pancreatic exocrine function. The basis of the test is that duodenal pancreatic enzymes digest the cobalamin–R protein complex, allowing cobalamin to bind intrinsic factor (IF). The cobalamin-IF complex is then absorbed in the ileum. As such, patients with exocrine dysfunction absorb cobalamin bound to IF normally but are unable to absorb cobalamin bound to R protein. After the oral ingestion of a test solution containing human IF[^{57}Co]cobalamin (0.2 nmol), hog R protein [^{58}Co]cobalamin (0.2 nmol), free human IF (0.4 nmol), and cobinamide (200 nmol), an intramuscular injection of cobalamin is administered. Urine is then collected for 24 hours, and [^{58}Co] and [^{57}Co] are measured. The ratio of [^{58}Co] to [^{57}Co] is calculated. Chen and associates [92] have reported that an excretion ratio for [^{58}Co]/[^{57}Co] of less than 0.68 was abnormal. Among 26 patients who underwent ERP, 0 of 9 patients with a normal pancreatogram, 4 of 9 patients with mild to moderate ductal changes on pancreatogram, and 7 of 8 patients with advanced ductal abnormalities on pancreatogram showed a positive value lower than the ratio of 0.68. In 28 patients who underwent CCK-secretin or cerulein-secretin testing, 2 of 13 with a normal test, 6 of 9 with mild exocrine dysfunction, and 5 of 6 with definitive exocrine dysfunction had an abnormal dual-labeled Schilling test. The [^{58}Co]/[^{57}Co] ratio significantly correlated ($r = 0.73$) with the maximal bicarbonate concentration as determined by CCK-secretin testing. The authors suggested that the dual-labeled Schilling test may be useful for identifying patients with relatively mild disease as well as those with more severe exocrine dysfunction.

Breath tests. Various breath tests using labeled carbon in a fatty acid of triglyceride have been developed to assess intralumenal digestion of fat by pancreatic lipase activity. [77]

Triolein breath test. Triglycerides such as triolein can be labeled with [^{14}C] fatty acids. After oral administration, triglycerides undergo digestion by pancreatic enzymes in the small intestine. Free fatty acids are absorbed by the intestinal mucosa and hepatically metabolized to yield [^{14}C]O_2, which is subsequently exhaled in the breath. Patients with severe pancreatic dysfunction exhale low to nonexistent levels of [^{14}C]O_2 after the ingestion of labeled triolein. Because only those with pancreatic dysfunction severe enough to cause fat maldigestion are identified and steatorrhea occurs only when lipase output is less than 10% of normal, [65] the triolein breath test is relatively insensitive in the setting of mild to moderate disease. For this test truly to reflect pancreatic function, nonpancreatic factors involving gastric emptying, gastroduodenal anatomy, intestinal absorption, hepatic metabolism, and pulmonary function must be intact. Disease states affecting any of these functions can lead to misleading results. In an attempt to improve specificity, some have suggested repeating the test after the oral administration of pancreatic enzyme supplements. Those with pancreatic exocrine insufficiency should experience an improvement in test results with exogenous pancreatic enzyme supplementation, whereas those with nonpancreatic causes would not be expected to improve. [93] Although this modification likely does improve specificity, it does not improve sensitivity of this test.

Mixed triglycerides breath test. Vantrappen and colleagues [94] recently described the mixed triglycerides breath test that uses 1,3-distearyl,2[carboxyl- ^{13}C] octanoyl glycerol as a substrate. In the presence of pancreatic lipase, the two stearyl groups are cleaved from the glyceryl group. [^{13}C]octanoyl monoglyceride and [^{13}C]octanoate, a medium-chain fatty acid, are absorbed and rapidly metabolized to [^{13}C]O_2. Breath [^{13}C]O_2 production correlates well with lipase output as measured by duodenal intubation with CCK administration. The reported sensitivity of 89% and pecificity of 81% for the mixed triglyceride breath test are comparable to other noninvasive studies, such as the pancreolauryl test. The authors claimed that this test is much more sensitive in detecting impaired pancreatic exocrine

function than fecal fat determination. [94] Indeed, the test detected most of the patients with chronic pancreatitis based on decreased pancreatic lipase output by invasive testing but no steatorrhea. It appears that the mixed triglyceride breath test is a promising noninvasive method to assess duodenal pancreatic lipase activity. This method may also provide a useful means of monitoring the efficacy of pancreatic enzyme replacement therapy. A breath test using ^{13}C-cholesteryl octanoate as a substrate has been reported. [95] Like the triolein breath test, nonpancreatic factors, including previous gastroduodenal surgery and significant intestinal, hepatic, metabolic, or pulmonary disease, can affect test results.

Starch breath test. The excretion of [^{13}C]O_2 in the breath after the oral administration of [^{13}C]-labeled rice starch (amylase dependent) or [^{13}C]-labeled glucose (amylase independent) has been measured in patients with chronic pancreatitis. [96] One study reported that breath [^{13}C]O_2 excretion after starch was significantly less than after glucose in both healthy volunteers and patients with chronic pancreatitis. However, [^{13}C]O_2 excretion after rice starch was significantly less in patients with chronic pancreatitis than in healthy volunteers. Unfortunately, the sensitivity of this test was less than 50%. Others have reported improved sensitivity (50% to 71%) with test modifications, including the administration of 100 mg of rice starch and the measurement of breath hydrogen excretion rather than [^{13}C]O_2. [97], [98] As is the case with tubeless pancreatic function testing in general, the starch breath test has poor sensitivity in patients with mild to moderate pancreatic exocrine dysfunction. False-positive breath hydrogen measurements can occur in the setting of small bowel bacterial overgrowth. Theoretically, bacterial overgrowth of the stomach, as seen in patients with gastroparesis and in those taking medications such as proton pump inhibitors, could also cause false-positive results.

Fecal Fat and Enzyme Tests

Quantitative stool collection with fecal fat determination is the gold standard for the detection of steatorrhea. For this study, patients should be instructed to ingest a diet containing 100 g of fat per day starting at least 3 days before the 48- to 72-hour stool collection. Fecal fat excretion of more than 7 g per 24 hours is abnormal. [2] If a patient is ingesting a proper diet, qualitative analysis of random stool samples with Sudan staining also sensitively and specifically detects steatorrhea. Unfortunately, less than 10% of normal lipase output must be present before fat maldigestion from pancreatic disease can be detected. [65] In addition, many other gastrointestinal diseases (e.g., Crohn's disease, celiac disease, Whipple disease) can cause steatorrhea. Consequently, the detection of steatorrhea by stool testing is neither sensitive nor specific for pancreatic disease. The specificity of fecal fat testing can be improved by performing a two-stage procedure in which a repeat collection is done in conjunction with oral pancreatic enzyme replacement. A decrease in fecal fat excretion after enzyme replacement supports the presence of pancreatic exocrine dysfunction.

Fecal pancreatic enzyme determination has been used to identify patients noninvasively who have severe chronic pancreatitis. The enzymes most commonly measured for this purpose include chymotrypsin and elastase.

Chymotrypsin Fecal chymotrypsin determination has been used for many years to identify patients with chronic pancreatic disease. The test is typically performed on random stool samples, although prolonged collection may enhance accuracy. It is convenient because fecal chymotrypsin remains stable at room temperature for days, [77] and it is easy to perform now that rapid colorimetric techniques are available. [99] Fecal chymotrypsin determination has a sensitivity of 45% to 100% and a specificity of 49% to 90%. [77], [99], [100], [101], [102] and [103] Measurement of fecal chymotrypsin can be confounded by binding to insoluble debris in the intestine. [104] Other factors, including Billroth II gastrojejunostomy, severe hepatic disease, and gastrointestinal diseases that cause diarrhea or malabsorption can affect the accuracy of this test. [77] Pancreatic enzyme supplementation can cause falsely normal results and should be discontinued 5 days before stool collection (Fig. 151-3). As is the case with other forms of noninvasive pancreatic function testing, fecal chymotrypsin measurement is better able to identify those with advanced disease as defined by invasive testing. [105]

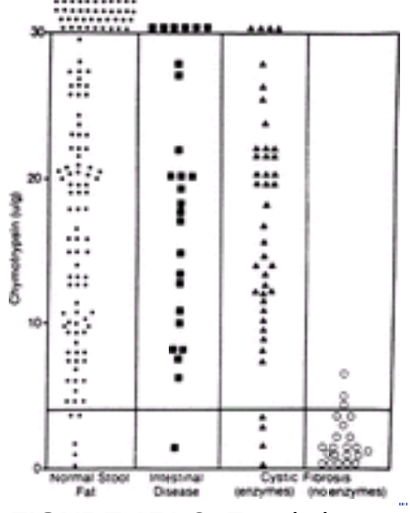

FIGURE 151-3. Fecal chymotrypsin activity in patients with normal stool fat excretion (127 cases), patients with steatorrhea due to intestinal disease (26 cases), patients with pancreatic insufficiency and steatorrhea due to cystic fibrosis but receiving pancreatic enzyme therapy (42 cases), and similar patients before enzyme replacement therapy (22 cases). (From ref. [101].)

The noninvasive nature, relatively low cost, and ease of performing and repeating fecal chymotrypsin determination are attractive features of this test. However, the marginal performance characteristics limit the usefulness of this test as a screening tool for chronic pancreatitis.

Elastase Human pancreatic elastase 1, a member of the acidic elastase family, was detected by Sziegoleit [106] as a new endoprotease and sterol-binding protein present in both human pancreatic secretion and feces. Elastase is measured with a new sandwich-type enzyme immunoassay. [107] Unlike chymotrypsin, quantitative studies using immunoelectrophoresis have shown that fecal elastase levels remain unaffected during intestinal transit. In fact, concentrations of elastase were found to be five to six times greater in stool than in pancreatic juice. [107], [108] Elastase has been found to be stable in stool samples for up to a week at room temperature. [109] Immunoreactive elastase cannot be detected in either porcine or bovine pancreatic enzyme preparations. Consequently, in contrast to fecal chymotrypsin determination, elastase measurement is not affected by oral pancreatic enzyme replacement therapy (Fig. 151-4). In addition, this test is not affected by previous gastrointestinal surgery, gastric dysmotility, or mucosal disease of the small intestine. [110]

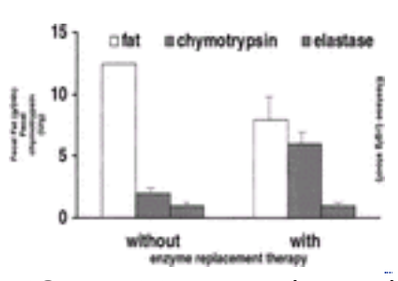

FIGURE 151-4. Fecal excretion of chymotrypsin (mean ± SD), fat, and immunoreactive elastase during a 24-hour collection period in 12 patients with cystic fibrosis and steatorrhea with and without enzyme replacement therapy. Fecal elastase content, unlike fecal fat and chymotrypsin content, was not influenced by pancreatic enzyme therapy. (From ref. [111].)

When patients with an abnormal secretin-pancreozymin test were divided into groups based on the presence or absence of steatorrhea, abnormal fecal elastase levels were found in 96% of patients with severe exocrine pancreatic insufficiency and steatorrhea and in 88% of those with pancreatic dysfunction but no steatorrhea. In the same group of patients, the sensitivity of fecal chymotrypsin measurement in those with steatorrhea was 91% but was only 56% in those without steatorrhea [111] (Table 151-5). The fecal elastase test was also compared with the pancreolauryl test in patients with chronic pancreatitis confirmed by ERP and CT. [110] Fecal elastase determination was as sensitive and more specific than the pancreolauryl test in detecting chronic pancreatitis.

TABLE 151-5 Fecal Elastase 1 Compared with Secretin-Pancreozymin Test and Fecal Chymotrypsin in Pancreatic Insufficiency with and without Steatorrhea

The fecal elastase test is a simple, noninvasive, relatively inexpensive functional test for patients with suspected chronic pancreatitis. Although reasonably sensitive in patients with severe disease, this test has not been found to be sufficiently sensitive in patients with mild or even moderate disease. [112], [113]

Miscellaneous Indirect Tests of Pancreatic Function

Serum Trypsin-like Immunoreactivity and Pancreatic Isoamylase Concentration The measurement of various pancreatic factors in peripheral blood as an indirect assessment of pancreatic function is an attractive prospect. Although serum trypsin-like immunoreactivity and pancreatic isoamylase concentration have been shown to be low in patients with severe pancreatic dysfunction, the sensitivity of these tests is probably less than 50% overall. [78] There are no reliable serum markers for severe pancreatic disease.

Plasma Pancreatic Polypeptide Concentration Pancreatic polypeptide (PP) is found in both the endocrine and exocrine portions of the pancreas. In normal volunteers, plasma PP concentration rises after stimulation of the pancreas and therefore may provide an indirect measure of exocrine function. An insufficient rise in plasma PP concentration after pancreatic stimulation with CCK, secretin, bombesin, or a test meal has a sensitivity of more than 90% in patients with severe pancreatic disease and steatorrhea. [78], [114] However, the sensitivity of this test in a population with less severe disease and no steatorrhea is likely less than 50%. [115]

Plasma Amino Acid Consumption Test In response to agents such as CCK, secretin, or cerulein, plasma amino acids are taken up by the pancreas and used in protein synthesis. The premise of the amino acid consumption test (AACT) is that a patient with severe pancreatic dysfunction should not deplete the plasma amino acid pool as efficiently as an individual with a normal pancreas. [116] Amino acid concentrations in timed venous blood samples obtained before and after pancreatic stimulation are compared. A fall in amino acid concentration of less than 12% after pancreatic stimulation with CCK is considered abnormal. [117] Although some investigators have reported sensitivity of the AACT to approach 90%, [116], [117] and [118] others have not been able to confirm these results. [119] In addition, the specificity of this test has been reported to be as low as 21%. [117] Given the need for multiple timed blood samples and the marginal performance characteristics, the AACT is of little value in clinical practice.

Comparison between Pancreatic Exocrine Function and Imaging in Chronic Pancreatic Disease

Transcutaneous ultrasound and high-resolution CT are widely accessible means of evaluating the pancreatic parenchyma. CT is more accurate than transcutaneous ultrasound in detecting small cysts and parenchymal calcifications. ERP has traditionally been regarded as the gold standard method to detect morphologic changes of the pancreatic duct. Unfortunately, ERP has a number of drawbacks, including relatively high cost and the potential for significant adverse events such as pancreatitis and even death. More recently, EUS has been shown to accurately detect abnormalities of the pancreatic duct and parenchyma. [120] EUS is clearly superior to transcutaneous ultrasound and CT and at least equivalent and arguably superior to ERP in detecting chronic pancreatitis. [61], [121], [122] One potential advantage of EUS over ERP is its lack of association with postprocedural pancreatitis. For all of the diagnostic tests for chronic pancreatitis, the challenge is not in the detection of severe disease that can usually be easily established with a limited evaluation. Rather, the difficulty lies in identifying patients with earlier stages of chronic pancreatitis. Unfortunately, none of the available structural or functional tests is always diagnostic in such patients. As such, a combination of structural and functional studies is often required to achieve diagnostic certainty in patients with mild to moderate disease. Among the structural and functional tests, ERP and invasive pancreatic function testing appear to be the best able to identify those with mild to moderate chronic pancreatitis.

There are relatively few studies that directly compare the ability of structural and functional tests to detect chronic pancreatitis. [123], [124], [125], [126] and [127] The best designed studies are those that evaluate functional or structural studies against a histological gold standard. For practical reasons, few such studies have been conducted. [128], [129] and [130] When direct comparisons have been made, invasive functional testing has been found to correlate more closely with histological findings than ERP in patients with chronic pancreatitis. [129], [130]

A number of published protocols have used one or more structural studies or invasive pancreatic function testing as a gold standard, making meaningful direct comparisons difficult, if not impossible. In one study that directly compared invasive pancreatic function testing (secretin-pancreozymin test) to ERP, Lankisch and colleagues [123] found that in most patients with chronic pancreatitis, there was a good correlation between the two tests. Of 202 patients, 129 (64%) had parallel secretin-pancreozymin and ERP test results. Twenty-one percent had abnormal results on both tests, but the degree of abnormalities observed was not consistent. Fifteen percent had discordant results, with one test abnormal and one test normal. In the group with discordant results, all but one patient had an abnormal ERP but normal secretin-pancreozymin result. Others have also observed discordant ERP and invasive functional test results in 12% to 29%. [124], [125], [126] and [127] Another recent study evaluated ERP, EUS, and the secretin test in 80 consecutive patients with recurrent pancreatitis and 25 controls. [61] The secretin test agreed with EUS 100% of the time in controls and patients with severe pancreatic disease. However, agreement was only 13% in those with mild and 50% in those with moderate chronic pancreatitis by EUS. There are several possible explanations for this observation. First, prior episodes of severe acute pancreatitis may have resulted in morphologic abnormalities of the pancreatic duct without long-term consequence to pancreatic function. [131] Second, morphologic changes of the pancreatic duct without corresponding changes in exocrine function are known to occur with aging. [132] Third, there are likely situations in which ERP films are thought to demonstrate changes suggestive of mild, chronic pancreatitis when in fact they do not. Finally, it may be that ERP is more sensitive than invasive functional testing or that the invasive functional testing is falsely normal in some patients with discordant results. This possibility seems less likely based on a recent study by Lambiase and associates, [127] who reported that 8 of 52 (15%) patients had divergent functional and ERP test results. Six of 8 patients had an abnormal functional test but normal ERP. Of the 6 patients with an abnormal functional test but normal ERP, 5 were subsequently found to have chronic pancreatitis. In contrast, neither of the 2 patients with an abnormal ERP and normal functional test was later found to have chronic pancreatitis. This study would suggest that invasive functional testing is better able to detect early chronic pancreatitis than ERP.

In summary, both structural and functional studies play an important role in establishing the diagnosis of chronic pancreatitis. This is especially true in patients with earlier stages of chronic pancreatitis. The information gained from such studies can prove independently diagnostic or complementary, particularly when assessing disease severity. Given the limitations of the available tests, perhaps the future of diagnostic testing for pancreatic disease will include the marriage of structural and functional studies. Recently, *intraductal secretin testing,* which involves the collection of pure intraductal pancreatic juice during ERP, has been described. Preliminary work suggests that this test yields performance characteristics comparable to traditional secretin testing. [133], [134] Hopefully, the future will bring the development of other novel tests for the diagnosis of chronic pancreatitis.

BREATH TESTS TO EVALUATE CARBOHYDRATE MALABSORPTION AND BACTERIAL OVERGROWTH

Measurements of the respiratory excretion of labeled CO_2 after oral administration and metabolism of carbon-labeled substrates, or of H_2 after administration of carbohydrates can be used to assess fat, carbohydrate, and bile salt malabsorption or bacterial overgrowth. [135] All breath tests that rely on the measurement of labeled CO_2, both those using radioactive ^{14}C and those using the stable isotope ^{13}C, are based on the idea that either intralumenal (bacterial) or cellular (intermediary) processes convert the substrate into CO_2, which is excreted in expired air. To calculate the dilution of tracer in the total CO_2 excreted, one must assume a constant CO_2 production of 9 mmol/kg body weight or 300 mmol/m^2 body surface area. The process is complicated further by the fact that it is impractical to measure CO_2 excretion continuously, and interval samples must be taken. The stable isotope (^{13}C) tests require mass spectrometric or infrared detection techniques and are subject to variability because of the natural abundance and inconsistency of ^{13}C in the diet.

Hydrogen breath tests can be used to study carbohydrate malabsorption or bacterial metabolism because the sole source of H_2 in the mammal is bacterial fermentation. The most commonly used breath tests are described in the following paragraphs.

Hydrogen Breath Test for Carbohydrate Malabsorption

In those individuals in whom a therapeutic trial of carbohydrate-restricted free diet is inconclusive or impractical, either breath hydrogen testing or lactose tolerance

testing may be indicated. [136], [137] Because hydrogen is produced only by bacterial fermentation of carbohydrates, increased breath hydrogen excretion after a carbohydrate challenge can be used to uncover small intestine bacterial overgrowth, disaccharidase deficiency, and even excess carbohydrate wastage that might occur with motility disorders. [138] Patients with small intestine mucosal disease and pancreatic exocrine insufficiency may also have mild carbohydrate malabsorption. Bacterial overgrowth of the small bowel may cause an early peak of increased hydrogen production within 2 hours of giving a carbohydrate meal. In disaccharidase deficiency, small intestine mucosal disease, or pancreatic insufficiency, the peak in hydrogen may come later, between 3 and 6 hours after ingestion, when the carbohydrate reaches the colonic bacteria [139], [140] and [141] (Fig. 151-5). The increase in hydrogen excretion by patients with pancreatic insufficiency can be reduced by concomitant administration of pancreatic enzymes. Therefore, depending on the type of carbohydrate malabsorption sought, an oral dose of lactose (0.25 to 1.0 g/kg body weight), glucose (50 g), lactulose (10 g), fructose (1 g/kg), or rice flour (100 g) may be given after an overnight fast. To test for lactose intolerance, a lactose dose of 25 g is frequently used, although a 50-g test dose may be more sensitive and a 12.5-g dose more specific. [136], [141] The test must be standardized for each carbohydrate and dose, but generally an increase of over 20 ppm in exhaled hydrogen over baseline values within the first 2 to 8 hours of ingestion is diagnostic. It is important to realize that the breath hydrogen test measures only carbohydrate malabsorption, and about 10% of people have a gut flora incapable of producing H_2. [142] In this circumstance, the measurement of breath methane excretion can be of value. Unfortunately, there are currently no standards established for what constitutes an abnormal rise in breath methane excretion in patients with suspected carbohydrate malabsorption.

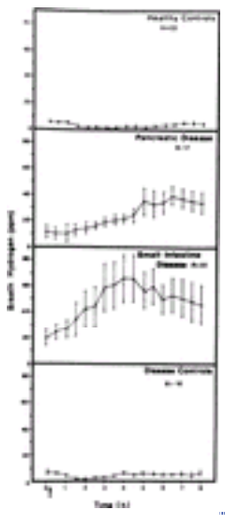

FIGURE 151-5. Breath histamine-2 (H_2) production following ingestion of 100 g of rice carbohydrate. Healthy and disease controls are compared with patients with pancreatic insufficiency and intestinal mucosal disease. The increased H_2 produced in patients with pancreatic insufficiency was corrected by addition of pancreatic extract to the rice pancake test meal. (From ref. [139].)

Hydrogen and Carbon-13 and -14 Breath Tests for Bacterial Overgrowth

The breath hydrogen test can be used to diagnose bacterial overgrowth and has a sensitivity of 62% to 90% and a specificity of 78% to 83% [143], [144] and [145] if breath H_2 increases are found within the first 2 hours of either glucose or rice flour ingestion. Because of its ready availability, glucose (50 to 75 g) is the most commonly used substrate. The combination of a high fasting breath hydrogen excretion rate and a significant increase in breath hydrogen after glucose (more than 10 to 12 ppm H_2 excretion over baseline) is present in about half of the patients with bacterial overgrowth. [146] An isolated increase in basal breath hydrogen excretion has a sensitivity for bacterial overgrowth of 4% to 29%. [147] Lactulose and rice flour have also been used as substrates for hydrogen breath testing, but sensitivity for bacterial overgrowth has been less than 50%. [148], [149] A number of factors can influence the accuracy of this test. Diseases affecting small intestinal absorption of the test substrate or with rapid intestinal transit can lead to false-positive results. In addition, bacterial overgrowth of the stomach, as can be seen in patients with gastric stasis and those taking a proton pump inhibitor, can also lead to false-positive results. Delayed gastric emptying in the absence of gastric bacterial overgrowth can cause false-negative results. About 10% of patients harbor non–H_2-producing enteric bacteria leading to false-negative results. As has already been mentioned, the measurement of breath methane excretion may be helpful in these patients, although studies to validate this suggestion have not been performed.

The ^{14}C-D-xylose breath test is said to be more specific for bacterial overgrowth, with more than 85% of patients having an increase in exhaled $^{14}CO_2$ within 60 minutes of ingesting 1 g of ^{14}C-D-xylose. [150], [151] Because most of the ^{14}C-labeled xylose is absorbed in the small intestine, less reaches the colon, where it can be metabolized to CO_2 by bacteria, confounding interpretation of the test. A version of the D-xylose breath test using the stable isotope ^{13}C has been used in children. [152] Although the test can be complicated by delayed gastric emptying, the champions of the test claim excellent specificity, sensitivity, and reproducibility for this test. Unfortunately, the available studies have typically involved small numbers of selected patients and have not universally endorsed this test. [153] A recent study in 46 consecutive patients with suspected bacterial overgrowth found the sensitivities for the ^{14}C-D-xylose and glucose hydrogen breath tests to be similar (42% versus 58%, respectively). [154]

The cholyl- ^{14}C-glycine breath test is based on the rationale that conjugated (glycine or taurine) bile acids are reabsorbed passively throughout the jejunum and actively absorbed in the terminal ileum. [155], [156] Bacterial overgrowth hydrolyzes the peptide bond, releasing the ^{14}C-labeled glycine, which is then absorbed, metabolized to $^{14}CO_2$, and exhaled in expired air. Unfortunately, the test is not specific, and it gives similar results in patients with terminal ileal disease or resection and may be misleading in severe mucosal disease.

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CHAPTER 152

Marc S. Levine, Stephen E. Rubesin, Hans Herlinger, and Igor Laufer

PLAIN AND CONTRAST RADIOLOGY

ABDOMINAL PLAIN FILMS

Abdominal plain films often play an important role in the evaluation of patients with abdominal pain or distention or with clinical signs of an acute abdomen. [1] The purpose of this section is to review plain film findings such as bowel dilation, pneumoperitoneum, and pneumatosis and to discuss their significance. However, computed tomography (CT) has increasingly been recognized as a more sensitive technique than abdominal plain films for diagnosing a host of conditions in patients with acute abdominal symptoms.

Technique

In patients with acute abdominal findings, both supine and upright plain films of the abdomen should be obtained. The upright films should be centered to include the diaphragms. This permits assessment not only of free intraperitoneal air beneath the diaphragms but also of air-fluid levels within the bowel that can be detected only on horizontal beam views. If patients are too sick or debilitated to stand, left lateral decubitus views should instead be obtained to detect free air between the liver and the right lateral abdominal wall.

Abnormalities

Bowel Dilation Bowel dilation is usually caused by obstruction or ileus. The small bowel is a more common site of obstruction than the colon. Other patients may develop an adynamic ileus without evidence of mechanical obstruction. These conditions are considered separately in the following sections.
Small Bowel Obstruction Because of the great increase in abdominal surgery in recent decades, most small bowel obstructions are caused by postoperative intraperitoneal adhesions. [1] Other, less common causes include incarcerated hernias, metastases, radiation, Crohn's disease, intussusception, and gallstone ileus. In a mechanical small bowel obstruction, supine abdominal plain films usually reveal multiple loops of dilated small bowel and a paucity of colonic gas (Fig. 152-1A); multiple air-fluid levels are almost always seen on upright or decubitus films (see Fig. 152-1B). Dilated small bowel can usually be differentiated from dilated colon by its more central location in the abdomen as well as the presence of tightly spaced folds or valvulae conniventes that completely traverse the diameter of the bowel.

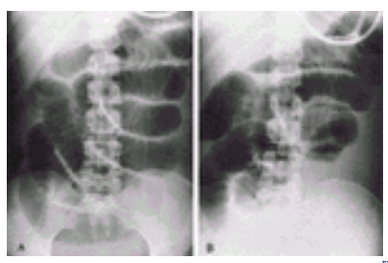

FIGURE 152-1. Small bowel obstruction due to postoperative adhesions. **A:** Supine abdominal film shows multiple loops of dilated small bowel and a paucity of colonic gas. **B:** Upright film shows multiple air-fluid levels in dilated small bowel. These findings are characteristic of small bowel obstruction. (From ref. [1].)

Although advanced small bowel obstructions are easily recognized on abdominal plain films, the diagnosis can be more difficult in patients who do not swallow a large amount of gas. In such cases, gaseous distention of bowel can be minimal or absent. Nevertheless, accumulation of fluid still occurs, and horizontal beam films may still demonstrate numerous air-fluid levels in nondilated bowel. Occasionally, a row of tiny gas bubbles or "string of pearls" may be seen on upright or decubitus films as a result of small amounts of gas trapped along the superior margin of small bowel loops almost completely filled with fluid. [2] In patients who swallow virtually no air at all, plain films may reveal a gasless abdomen with multiple fluid-filled loops of bowel appearing as tubular or sausage-shaped densities in the bowel that may be indistinguishable from true abdominal masses. [2] A simple mechanical obstruction usually does not cause bowel ischemia. However, a closed loop obstruction due to an incarcerated hernia, volvulus, or other cause may occasionally produce a strangulating obstruction with ischemia or necrosis of the involved segment of bowel. Recently, CT has been recognized as a sensitive technique for detecting strangulation of bowel in patients with closed loop obstruction. [3] It is important to recognize that advanced cecal carcinomas may obstruct the ileocecal valve, mimicking the plain film appearance of a distal small bowel obstruction. [1] More distal colonic obstructions may also be masked by an incompetent ileocecal valve that allows gas to reflux from the obstructed colon into the small bowel. A barium enema may be performed on patients with radiographic evidence of a distal small bowel obstruction to rule out an unsuspected colonic carcinoma, particularly when there is no history of prior abdominal surgery.
Colonic Obstruction In colonic obstruction, supine abdominal plain films usually reveal disproportionate colonic distention proximal to the obstructing lesion with air-fluid levels in the dilated bowel on upright films. The transition from dilated to nondilated bowel can often be recognized on the radiographs and most commonly occurs in the sigmoid colon because of an obstructing carcinoma. Primary colonic carcinoma accounts for 80% to 90% of all colonic obstructions, but diverticulitis, metastases, and volvulus are other, less common causes. [1] The most devastating complication of colonic obstruction is cecal perforation. Because of the high mortality associated with cecal perforation, colonic obstruction should be considered a surgical emergency. In general, a cecal diameter greater than 10 to 12 cm on

abdominal plain films is thought to be an indication for urgent colonic decompression because of the high risk for perforation. [4] Colonic volvulus should be distinguished from a simple colonic obstruction because it results from twisting of the colon around a fixed point on its mesentery, producing a closed loop obstruction. Most cases involve the sigmoid colon or cecum. [5] Sigmoid volvulus is an acquired condition that often occurs in elderly patients who have chronic constipation or a high-residue diet. [6] In contrast, cecal volvulus tends to occur in younger patients owing to congenital failure of retroperitoneal fixation of the cecum and ascending colon. [6] As a result, the right side of the colon has a persistent mesentery on which a volvulus can take place. In sigmoid volvulus, abdominal plain films usually reveal a massively dilated sigmoid colon that extends out of the pelvis into the upper abdomen with some degree of proximal colonic distention [1], [7] (Fig. 152-2). In contrast, cecal volvulus usually produces a massively dilated cecum that flips into the left upper quadrant with a single air-fluid level on upright or decubitus horizontal beam films. [1], [7] Because colonic volvulus is a closed loop obstruction, twisting of the mesentery may compromise the vascular supply of the bowel, leading to strangulation, infarction, and perforation of the involved loop. Early diagnosis is therefore essential, so that colonic volvulus can be treated before strangulation occurs.

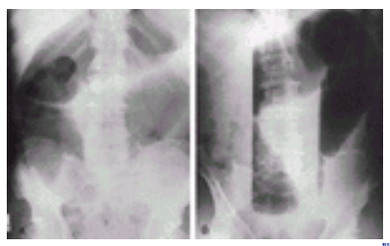

FIGURE 152-2. Sigmoid volvulus. **A:** Supine abdominal plain film shows a massively dilated sigmoid colon rising out of the pelvis into the abdomen. **B:** Right lateral decubitus film shows long air-fluid levels in both limbs of this dilated sigmoid loop. (From ref. [1].)

Because of the risk for cecal perforation in patients with colonic obstruction, a barium enema may be required as an emergent procedure to determine whether an obstruction is present and to delineate the nature and site of the obstructing lesion. Carcinoma of the colon typically manifests as an annular lesion with shelflike, overhanging borders, whereas diverticulitis produces a tapered area of narrowing with intact but distorted mucosal folds. In colonic volvulus, the barium enema reveals a typical “bird-beak” deformity at the site of the volvulus.

Adynamic Ileus In patients with an adynamic or paralytic ileus, there is interference with intestinal transport without an actual mechanical obstruction. As a result, a supine abdominal radiograph classically shows diffusely dilated small and large bowel. Although the degree of distention depends on the degree of air swallowing, horizontal beam films usually demonstrate air-fluid levels throughout the bowel. [8] For reasons that are unclear, however, abdominal plain films sometimes reveal an isolated small bowel or colonic ileus. In such cases, the findings may be indistinguishable radiographically from a mechanical small bowel or colonic obstruction. [1], [8] When the colon appears dilated, the presence of a significant amount of rectal gas on left lateral views of the rectum should favor an ileus. [9] If it is unclear whether the patient has a distal colonic obstruction or an ileus, a single-contrast barium enema should be performed to differentiate these conditions.

Pneumoperitoneum Upright chest and abdominal films and left lateral decubitus films of the abdomen are extremely sensitive for detecting free intraperitoneal air (i.e., pneumoperitoneum) and may demonstrate collections of intraperitoneal air as small as 1 mL. [10] In patients with an acute abdomen, the presence of free air almost always indicates a perforated viscus with subsequent peritonitis. The most common cause is a perforated duodenal ulcer. [1] Colonic perforation due to ischemic bowel disease, toxic megacolon, and diverticulitis is much less common but should be suspected in any patient with pneumoperitoneum and colonic distention on abdominal plain films. [11] Pneumoperitoneum can readily be documented on upright chest films or upright or left lateral decubitus films of the abdomen by the presence of free intraperitoneal air directly beneath the diaphragms (Fig. 152-3A) or between the liver and the right lateral abdominal wall. Unfortunately, some patients are too ill or debilitated to stand or lie on their side; supine abdominal radiographs may be the only films that are obtained in these patients. However, pneumoperitoneum can be recognized on supine films by the presence of air on both sides of the bowel wall (i.e., the Rigler sign) (see Fig. 152-3B), by linear or triangular collections of gas in the subhepatic space, or by air outlining the falciform ligament as a linear density in the right upper quadrant. [1], [2] In one study, one or more signs of free intraperitoneal air were present on supine films in 59% of patients with pneumoperitoneum. [12]

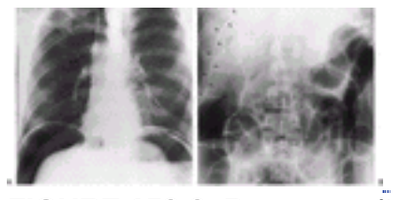

FIGURE 152-3. Pneumoperitoneum. **A:** Upright chest film shows large amounts of free intraperitoneal air beneath the diaphragm in this patient with a perforated duodenal ulcer. **B:** Supine abdominal plain film of a different patient shows an indirect sign of pneumoperitoneum with air on both sides of the bowel wall (the Rigler sign) (*arrows*) after inadvertent perforation at colonoscopy. (From ref. [1].)

Pneumatosis Intramural bowel gas or pneumatosis may be caused by ischemic or necrotic bowel but may also occur as a benign finding in asymptomatic patients. In patients with intestinal necrosis, gas may dissect into the wall of the bowel, producing mottled or linear gas shadows that have a characteristic plain film appearance [1], [13] (Fig. 152-4). CT has been recognized as a more sensitive technique than abdominal plain films for detecting pneumatosis in patients with necrotic bowel. In patients with intestinal necrosis, tiny, linear, peripherally branching gas shadows may occasionally be identified over the liver owing to gas within the portal venous system. [13] This finding usually indicates severe disease with an extremely ominous prognosis. However, portal venous gas should be distinguished radiographically from biliary gas (i.e., pneumobilia), which tends to collect centrally in the common bile duct because of the opposite direction of bile flow. Pneumobilia can result from a variety of causes, including gallstone ileus, emphysematous cholecystitis, and a surgical or spontaneous biliary-enteric fistula. [1]

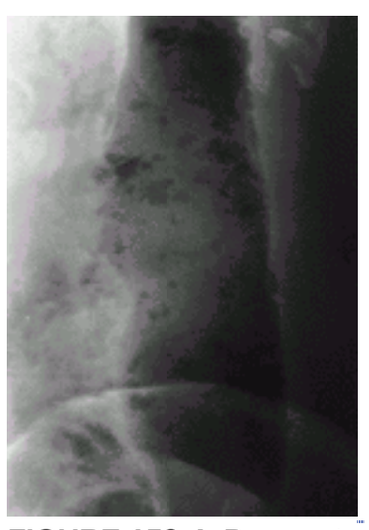

FIGURE 152-4. Pneumatosis due to an infarcted left colon after surgery. Close-up view from supine abdominal plain film shows tiny, mottled, and linear collections of gas in the wall of the descending colon.

Pneumatosis can also occur as a benign condition in which multiple gas-filled cysts or blebs are present in the wall of the bowel. [1], [13] These patients are rarely ill, and the condition is usually discovered accidentally on abdominal plain films obtained for other reasons. Radiographically, these gas-filled blebs are typically recognized as cystic or grapelike collections of gas that appear quite different from the linear gas shadows associated with intestinal necrosis.

CONTRAST STUDIES

Contrast studies of the gastrointestinal tract can be performed using single- or double-contrast technique. Single-contrast studies entail filling the lumen with a relatively low-density barium suspension and evaluating for contour abnormalities or filling defects in the barium pool. In contrast, double-contrast studies use a smaller amount of high-density barium and gas to evaluate the en face appearance of the mucosal surface. As a result, double-contrast techniques have dramatically

improved the radiologist's ability to diagnose a variety of inflammatory and neoplastic diseases throughout the gastrointestinal tract. A major advantage of these techniques is their ability to demonstrate superficial mucosal abnormalities that cannot easily be recognized on conventional single-contrast examinations. In other cases, double-contrast studies may detect lesions that are missed or misinterpreted at endoscopy. Double-contrast radiography is also less expensive and less invasive than endoscopy. Thus, it is a valuable technique for evaluating patients with suspected gastrointestinal disease. All radiologists should strive for excellent double-contrast studies to maximize the information available from the procedure.

PHARYNX

Indications

With increased survival of elderly people, pharyngeal disorders have become an increasingly frequent problem in modern medical practice. About 35% of nursing home patients have some form of swallowing dysfunction. [14] Aspiration pneumonia and choking are particularly common causes of morbidity and mortality. Radiographic examination of the pharynx is now recognized as a valuable tool in the diagnostic workup of this large group of patients with pharyngeal disorders.

Contrast studies of the pharynx are most frequently performed on patients who have difficulty swallowing. However, disorders of the pharynx also may manifest as respiratory and speech problems. Laryngeal penetration or overflow aspiration may lead to recurrent pneumonia, asthma, chronic bronchitis, coughing, or choking. In other patients, soft-palate insufficiency may result in nasal regurgitation or may give the voice a nasal quality. A pharyngoesophagram may be helpful in patients who have a wide spectrum of respiratory, speech, and swallowing difficulties. Barium studies are also useful in assessing pharyngeal function and morphology in patients with a history of neuromuscular disease, stroke, pharyngeal tumor, or prior head and neck surgery or radiation.

Normal Anatomy

The pharynx is a complex muscular tube suspended superiorly from the skull base and styloid process, posteriorly from the cervical spine, and anteriorly from the mandible and hyoid bone. At least 26 muscles and six cranial nerves participate in pharyngeal function. [15], [16]

The pharynx can be arbitrarily divided into three portions: the nasopharynx, oropharynx, and hypopharynx. The soft palate separates the nasopharynx from the oropharynx, and the pharyngoepiglottic fold separates the oropharynx from the hypopharynx. The tongue forms the anterior wall of the oropharynx. The larynx, with its associated epiglottic, thyroid, cricoid, and arytenoid cartilages, forms the anterior wall of the hypopharynx. This laryngeal complex often protrudes into the lower hypopharynx as an apparent mass.

The mucosal surface of the pharynx is thrown into a series of folds by underlying lymphoid and muscular tissue. The vertical surface of the base of the tongue often has a nodular appearance as a result of the circumvallate papillae and lingual tonsil. Nodular lymphoid tissue or linear webs may also interrupt the normally smooth surface of the valleculae. Although the anterior border of the hypopharynx usually has a smooth contour, close apposition of the longitudinal muscles of the pharynx to the overlying squamous mucosa results in longitudinal striations of the lateral and posterior walls of the hypopharynx. [16] Horizontal mucosal striations are seen in the redundant mucosa overlying the arytenoid processes and cricoid cartilages. [16]

Technique

A complete radiographic examination of the pharynx includes a cine or video pharyngoesophagram to evaluate motility and a series of spot films to evaluate morphology. [17], [18] and [19] In patients with suspected foreign body, fistula, or abscess, frontal and lateral plain films of the neck should also be obtained.

The barium study is initially performed with a high-density barium suspension for optimal visualization of the pharynx. The patient is asked to swallow barium in frontal, lateral, and oblique projections. A video recording of each swallow permits a frame-by-frame or slow motion analysis of the various parameters of deglutition. Movement of the tongue, soft palate, and epiglottis, as well as of the laryngeal closure and cricopharyngeal opening, are best evaluated in the lateral projection. However, symmetry of tongue motion, pharyngeal peristalsis, and epiglottal tilt are best evaluated in the frontal projection.

After individual swallows of barium, double-contrast spot films of the pharynx are obtained in frontal and lateral projections. The spot films are obtained during suspended respiration and during a modified Valsalva maneuver or phonation to distend the pharynx optimally. [20] The frontal view is best for demonstrating the contours of the valleculae and piriform sinuses, the lateral walls of the tonsillar fossae and hypopharynx, and the superior border of the base of the tongue. The lateral view is best for demonstrating the inferior border of the base of the tongue, the soft palate, the posterior pharyngeal wall, the anterior hypopharyngeal wall, the epiglottis, and the cricopharyngeus.

After the pharyngeal examination has been completed, upright double-contrast and prone single-contrast views of the esophagus are obtained to rule out associated esophageal disease.

Abnormalities

Laryngeal Penetration and Aspiration Laryngeal penetration occurs when barium enters the laryngeal vestibule during swallowing. Penetration may be limited to the region of the subepiglottic space or may extend as far as the true vocal cords or trachea. Laryngeal penetration occurs because of poor timing of oral and pharyngeal events associated with swallowing or pharyngeal dysmotility due to neuromuscular disorders such as amyotrophic lateral sclerosis, multiple sclerosis, or cerebrovascular accidents. Inflammatory or neoplastic diseases that restrict pharyngeal motility may also cause penetration. Aspiration occurs when barium enters the laryngeal vestibule during normal breathing. Aspiration results from stasis and retention of pharyngeal contents because of tumor, diverticula, or neuromuscular disease in the pharynx. Aspiration may also be caused by gastroesophageal reflux or reflux of esophageal contents above an obstructing esophageal lesion, such as a stricture or carcinoma. Penetration is primarily associated with dysmotility, and aspiration is associated with stasis.
Cricopharyngeal Prominence The pharyngoesophageal segment, the radiographic equivalent of the manometrically defined upper esophageal sphincter (UES), is formed by the inferior portion of the cricopharyngeus and possibly the proximal cervical esophagus. [21], [22] Although the UES is tonically contracted at rest, initiation of swallowing causes the sphincter to relax ahead of the oncoming bolus. The UES also acts as part of the pharyngeal peristaltic wave, functioning in sequence with the constrictor musculature. The pharyngoesophageal segment is best evaluated on a dynamic recording of the pharynx in a lateral projection. During swallowing, a prominent cricopharyngeus appears as a smooth, 1-cm in height, barlike protrusion of the posterior pharyngeal wall into the barium column on lateral projections (Fig. 152-5). The cricopharyngeus may show delayed opening, incomplete opening, or early closure. A prominent cricopharyngeus is detected on barium studies in about 5% of asymptomatic individuals. [23], [24] However, some patients with this finding complain of dysphagia. In symptomatic patients, a prominent cricopharyngeus is often associated with pharyngeal paresis or occurs as a compensatory response to gastroesophageal reflux or esophageal obstruction.

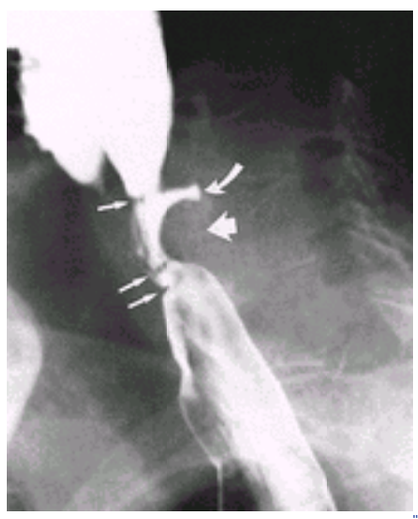

FIGURE 152-5. Incomplete opening of cricopharyngeus, manifested by posterior indentation on pharyngoesophageal segment (*large arrow*). Notice the associated

webs anteriorly (*small arrows*) and pseudo Zenker diverticulum posteriorly (*curved arrow*) that is seen because barium is trapped above a prominent cricopharyngeus. Cricopharyngeal dysmotility is often related to underlying gastroesophageal reflux or pharyngeal paresis.

Lateral Pharyngeal Pouches and Diverticula Lateral pharyngeal pouches are transient protrusions of the lateral pharyngeal wall at sites of anatomic weakness, such as the posterior thyrohyoid membrane and tonsillar fossae after a tonsillectomy. [25] These pouches are common findings, usually occurring as normal variants in asymptomatic patients. In contrast, lateral pharyngeal diverticula are persistent protrusions from the tonsillar fossae or region of the thyrohyoid membrane. These diverticula are much less common than pharyngeal pouches, occurring primarily in individuals who have markedly elevated pharyngeal pressure, such as glassblowers and tuba players. If stasis occurs in pharyngeal pouches or diverticula, the subsequent spillage of pouch contents into the hypopharynx may result in aspiration into the larynx or tracheobronchial tree. Stasis with delayed spill into the hypopharynx may also cause mild neck discomfort or dysphagia after swallowing. Diverticula may also manifest as neck masses and occasionally may be sites of ulceration or neoplasia. Lateral pharyngeal pouches appear on frontal views as transient, hemispheric protrusions of mucosa in the upper hypopharynx above the calcified edge of the thyroid cartilage. These pouches can be recognized on lateral views as ovoid barium collections or rings anteriorly in the upper hypopharynx just below the hyoid bone. In contrast, lateral pharyngeal diverticula appear as persistent protrusions in these areas.
Zenker Diverticulum Zenker diverticulum, or posterior hypopharyngeal diverticulum, is an acquired mucosal herniation through an area of anatomic weakness in the region of the cricopharyngeus (i.e., the Killian dehiscence). This area of anatomic weakness is located between the thyropharyngeus and cricopharyngeus or between the oblique and horizontal fibers of the cricopharyngeus. [21], [22] Most patients with Zenker diverticulum have an associated hiatal hernia or gastroesophageal reflux. Rarely, these diverticula are complicated by ulceration or malignancy. During swallowing, a Zenker diverticulum appears radiographically as a posterior bulging of the distal pharyngeal lumen above an anteriorly protruding cricopharyngeal bar (Fig. 152-6A). At rest, the barium-filled diverticular sac often extends below the level of the cricopharyngeus posterior to the proximal cervical esophagus (see Fig. 152-6B).

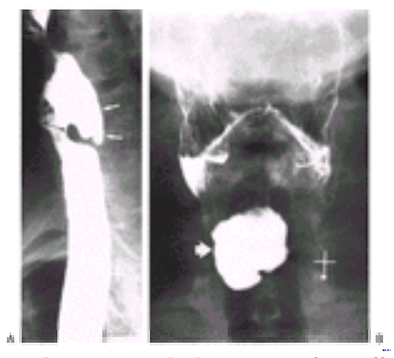

FIGURE 152-6. A: Zenker diverticulum seen as a posterior outpouching of the hypopharyngeal wall (*white arrows*) above a prominent cricopharyngeus (*black arrow*) during swallowing. **B:** A frontal view shows retention of barium in a diverticulum (*arrow*) at completion of the swallow.

Inflammatory Conditions Barium studies are of limited value in patients with viral, bacterial, or fungal infection of the pharynx. [26] Such patients usually have normal pharyngograms or nonspecific lymphoid hyperplasia of the palatine tonsil or base of the tongue. Occasionally, however, *Candida* species infection or herpes pharyngitis may manifest on double-contrast radiographs as plaques or ulcers in the pharynx, particularly in patients with acquired immunodeficiency syndrome (AIDS). Barium studies may also be helpful in a patient who has a chronic sore throat to determine whether there is underlying gastroesophageal reflux or reflux esophagitis.
Tumors Double-contrast pharyngography has an important role in the initial detection and subsequent workup of pharyngeal tumors. [27], [28] Double-contrast radiographs of the pharynx can accurately define the size, level, and extent of the lesion. The radiologic examination is particularly helpful in demonstrating regions of the pharynx (e.g., valleculae, lower hypopharynx, cricopharyngeus) that are difficult to visualize at endoscopy. It also enables detection of submucosal masses that are easily missed at endoscopy. Although its accuracy is limited in the region of the palatine tonsil, the double-contrast examination is capable of detecting more than 95% of all mucosal neoplasms in the pharynx below the level of the pharyngoepiglottic fold. [27] Squamous cell carcinoma is by far the most common malignant tumor of the pharynx. With an overall 5-year survival rate of about 20%, this tumor has a somewhat better prognosis than esophageal carcinoma. These lesions may manifest on double-contrast radiographs as an intralumenal mass, mucosal irregularity, or loss of distensibility. [29] An intralumenal mass may cause asymmetry or obliteration of the normal pharyngeal contour, barium-coated lines in unusual locations, or a superimposed radiodensity. Mucosal irregularity may manifest as an irregular, lobulated, nodular, or granular surface pattern. A loss of distensibility may be associated with fixation of pharyngeal structures by infiltrating tumor. When malignant lesions are detected in the pharynx, the esophagus should be carefully evaluated radiographically because of the increased incidence of synchronous esophageal cancers in these patients. [30]

UPPER GASTROINTESTINAL TRACT

The development of routine double-contrast techniques for examining the upper gastrointestinal tract has dramatically improved our ability to diagnose a variety of inflammatory and neoplastic diseases in the esophagus, stomach, and duodenum. Despite increasing acceptance of this technique, some investigators advocate endoscopy as the initial screening study in patients with dyspepsia or other upper gastrointestinal symptoms. Although endoscopy has been recognized as a highly accurate technique for examining the upper gastrointestinal tract, it is also an invasive technique with a small but measurable risk for gastric perforation or other complications. Furthermore, it is an expensive technique, costing three to four times more in the United States than double-contrast upper gastrointestinal examinations. Because barium studies are safer and less expensive than endoscopy, radiologic evaluation of the upper gastrointestinal tract remains a viable alternative as long as its accuracy approaches that of endoscopy for clinically significant disease. We think that a carefully performed double-contrast examination provides the best opportunity for radiology to be competitive with endoscopy as a diagnostic modality.

Technique

The routine double-contrast upper gastrointestinal examination should be performed as a biphasic study in which double-contrast and single-contrast views of the esophagus, stomach, and duodenum are obtained. [31] In the double-contrast portion of the study, a series of maneuvers is required to achieve adequate gaseous distention of the lumen while a thin layer of high-density barium is spread on the mucosa. The double-contrast examination is facilitated by the use of pharmacological agents (e.g., 0.1 mg of glucagon given intravenously) to induce gastric hypotonia. After the double-contrast portion of the study has been completed, prone or upright single-contrast views of the esophagus, stomach, and duodenum are obtained with a low-density barium suspension and various degrees of compression to supplement the double-contrast study. Because of its greater diagnostic yield, this biphasic study has been advocated as the best radiologic technique for examining the upper gastrointestinal tract.

Indications

Esophagus In patients with reflux symptoms, barium studies have traditionally been advocated to document the presence of a sliding hiatal hernia or gastroesophageal reflux, to detect complications such as strictures, and to rule out other abnormalities in the esophagus that can mimic reflux esophagitis. By permitting a more detailed assessment of the esophageal mucosa, double-contrast radiographic techniques have made it possible to detect superficial ulceration and other changes of esophagitis before the development of deep ulcers and strictures. Because modern medical care is prolonging the survival of patients who are immunosuppressed, infectious esophagitis has become an increasingly frequent problem. When infectious esophagitis is suspected on clinical grounds, double-contrast esophagography may be performed to confirm the diagnosis and differentiate the various underlying organisms. The radiologist's ability to differentiate fungal and viral esophagitis is particularly important for patients with AIDS because some gastroenterologists are reluctant to perform endoscopy on these individuals because of fear of contaminating their endoscopic instruments or exposing themselves to the human immunodeficiency virus (HIV). However, endoscopy should be performed if the radiographic findings are equivocal. Dysphagia is another important indication for performing barium studies. If the sensation of dysphagia is localized to the pharynx, a careful pharyngeal examination should be obtained. However, some lesions involving the distal esophagus or cardia may cause dysphagia referred to the upper esophagus or pharynx. Thus, the gastric cardia and esophagus should be carefully evaluated radiographically in all patients with unexplained pharyngeal dysphagia to rule out a distal lesion masquerading as a pharyngeal disorder.
Stomach and Duodenum The most common indications for performing a double-contrast examination of the stomach and duodenum include epigastric pain or discomfort, bloating, belching, early satiety, and signs or symptoms of upper gastrointestinal bleeding, such as hematemesis, melena, and guaiac-positive stool. If erosive gastritis or duodenitis, duodenal ulcers, or unequivocally benign-appearing gastric ulcers are diagnosed radiographically, the patient can be treated medically without need for endoscopic intervention. If, however, the double-contrast examination demonstrates a gastric ulcer or other lesion that is equivocal or suspicious for malignancy, endoscopy and biopsy should be performed for a more definitive diagnosis. If the double-contrast examination is normal, the decision for endoscopy

should be based on the severity of symptoms, age, and overall health of the patient.
Contraindications and Risks Because oral barium sulfate is contraindicated in patients with suspected esophageal or gastric perforation, water-soluble contrast media should be used if there are any clinical or radiographic signs of mediastinitis or peritonitis. Otherwise, the risks of the barium study are negligible. Nevertheless, a double-contrast examination may be difficult to perform on elderly or debilitated patients who cannot undergo the turning maneuvers required for this examination. These patients may be evaluated by conventional single-contrast barium studies.

Abnormalities

Gastroesophageal Reflux Disease In patients with suspected gastroesophageal reflux disease, barium studies may be performed not only to detect gastroesophageal reflux but also to look for the morphologic sequelae of reflux, including reflux esophagitis, peptic strictures, and Barrett esophagus. Conventional single-contrast esophagography has been considered an unreliable technique for diagnosing reflux esophagitis, with an overall sensitivity of only 50% to 75% reported in the literature. However, the use of double-contrast esophagography has increased the radiographic sensitivity to almost 90%. [32] In relatively mild reflux esophagitis, mucosal edema and inflammation may manifest on double-contrast radiographs as a granular or finely nodular mucosa. [32] Other patients may have shallow ulcers and erosions appearing as one or more tiny collections of barium in the distal esophagus near the gastroesophageal junction (Fig. 152-7). In more severe disease, the esophagus may have a grossly irregular contour, with serrated margins and decreased distensibility due to extensive ulceration, edema, and spasm. Scarring from reflux esophagitis may lead to the development of reflux-induced or peptic strictures, most commonly seen as tapered areas of concentric narrowing in the distal esophagus above a hiatal hernia.

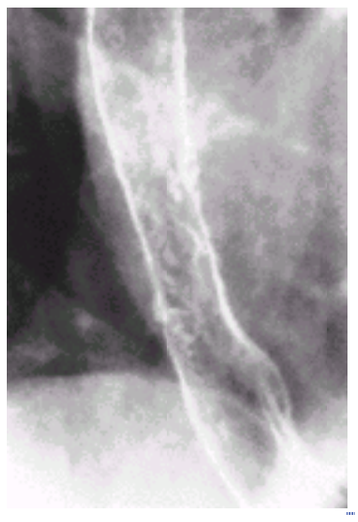

FIGURE 152-7. Reflux esophagitis with multiple areas of shallow ulceration seen en face and in profile in the distal esophagus. (From ref. [31].)

Barrett esophagus is a well-recognized complication of reflux esophagitis that is associated with a significantly increased risk for developing esophageal adenocarcinoma. Unfortunately, the classic radiologic features of Barrett esophagus (i.e., a high esophageal stricture or ulcer or a reticular mucosal pattern) occur in only a minority of patients. [33] However, data suggest that patients without reflux esophagitis or peptic strictures on double-contrast examinations rarely have Barrett esophagus at endoscopy. [34] Thus, double-contrast esophagography may be a useful screening study for Barrett esophagus to determine the relative need for endoscopy and biopsy in patients with reflux symptoms.
Infectious Esophagitis Esophagography has traditionally been considered an unreliable technique for diagnosing *Candida* esophagitis, with an overall sensitivity of less than 50%. [35] With double-contrast technique, however, esophagography has a sensitivity of about 90% in diagnosing *Candida* esophagitis. [36] The major advantage of this technique is its ability to demonstrate mucosal plaques that cannot easily be recognized on conventional single-contrast studies. These discrete, plaquelike lesions tend to be longitudinally oriented, appearing on double-contrast radiographs as linear or irregular filling defects with normal intervening mucosa (Fig. 152-8A).

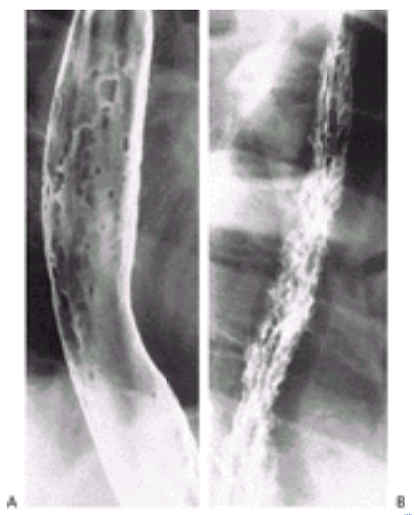

FIGURE 152-8. Candida esophagitis. **A:** Double-contrast esophagram shows discrete plaque-like defects with normal intervening mucosa. This appearance is characteristic of candidiasis. **B:** Severe Candida esophagitis in a patient with acquired immunodeficiency syndrome. Notice the shaggy contour of the esophagus, caused by multiple plaques, pseudomembranes, and ulcers. (**A,** from ref. [36].)

The AIDS epidemic has led to the development of a more fulminant form of *Candida* esophagitis, manifesting as a “shaggy” esophagus with a grossly irregular contour due to multiple plaques, pseudomembranes, and ulcers [37] (see Fig. 152-8B). Because this degree of esophagitis rarely occurs in other immunocompromised patients, the possibility of AIDS should be suspected when a shaggy esophagus is detected on barium studies, particularly in young homosexual men. Herpes and, less frequently, cytomegalovirus (CMV) esophagitis also occur in immunosuppressed patients with odynophagia, and these conditions should be suspected in the same clinical setting as *Candida* esophagitis. More than 50% of patients with herpes esophagitis have discrete, superficial ulcers on the esophageal mucosa that are readily detected on double-contrast radiographs [38] (Fig. 152-9). In contrast, CMV esophagitis, which occurs primarily in patients with AIDS, often manifests as one or more large, relatively flat ulcers in the esophagus. [37]

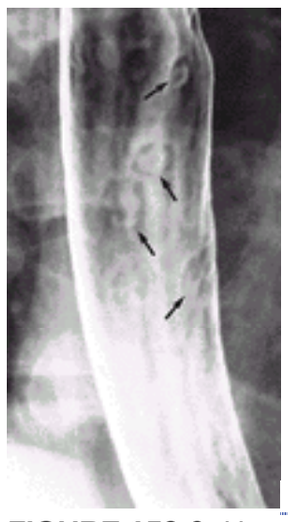

FIGURE 152-9. Herpes esophagitis with discrete superficial ulcers (*arrows*) on relatively normal background mucosa. Radiolucent halos of edematous mucosa surround the ulcers. (From ref. [37].)

HIV has also been associated with the development of giant esophageal ulcers in HIV-positive patients with odynophagia. The lesions typically appear radiographically as giant, flat ulcers indistinguishable from those caused by CMV. [39] Because HIV-related ulcers may respond dramatically to oral steroids, endoscopy is required to differentiate HIV from CMV infection in the esophagus before initiating treatment.

Esophageal Carcinoma Esophageal carcinoma is a deadly disease with an overall 5-year survival rate of only 5% to 10%. This dismal prognosis is primarily related to the advanced stage of the disease at the time of clinical presentation. Occasionally, esophageal cancer may be discovered at an early stage, and unlike advanced esophageal carcinoma, early esophageal cancer is a readily curable lesion, with reported 5-year survival rates approaching 90%. In Western countries, detection of these lesions is best accomplished by some form of radiologic or endoscopic surveillance of patients known to be at increased risk for developing esophageal cancer. Early esophageal cancers classically appear on double-contrast esophagrams as protruded lesions less than 3.5 cm in diameter. [40] They may be plaquelike lesions or small, sessile polyps with smooth or slightly lobulated contours. Other superficial spreading carcinomas may manifest radiographically as tiny coalescent nodules or plaques, causing localized nodularity or granularity of the mucosa. [40] In contrast, advanced esophageal carcinomas appear as polypoid, ulcerated, or infiltrating lesions with mass effect, ulceration, or irregular narrowing of the lumen. Some authors believe that endoscopy is warranted for all patients with dysphagia who have negative esophagrams because of the risk for missing esophageal cancer on barium studies. In a recent study, however, carcinoma of the esophagus or esophagogastric junction was diagnosed or suspected on double-contrast esophagography in 96% of patients with proven cancers. [41] In the same study, endoscopy was recommended to rule out malignant tumors of the esophagus or esophagogastric junction in only about 1% of all patients who had barium examinations. Thus, a high sensitivity can be achieved in the radiographic diagnosis of these tumors without exposing an inordinate number of patients to unnecessary endoscopy.
Erosive Gastritis Gastric erosions may be classified radiographically as complete or incomplete erosions. Most patients have complete or varioliform erosions in which a punctate or slitlike collection of barium is surrounded by a radiolucent halo of edematous mucosa [42] (Fig. 152-10A). Varioliform erosions are most commonly found in the gastric antrum and are often aligned on rugal folds. In contrast, incomplete or “flat” erosions appear as dots or streaks of barium without an edematous halo. Because the surrounding mucosa is normal, incomplete erosions have been extremely difficult to detect radiographically, accounting for less than 5% of all erosions seen on double-contrast studies.

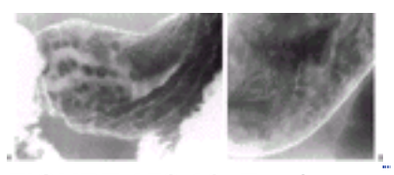

FIGURE 152-10. Erosive gastritis. **A:** Typical varioliform erosions in the gastric antrum. The erosions are aligned on the rugal folds. **B:** Linear and serpiginous erosions in the body of the stomach near the greater curvature are the result of recent ingestion of indomethacin.

Although erosions caused by aspirin or other nonsteroidal antiinflammatory drugs often are located in the gastric antrum, they sometimes occur in the gastric body, appearing as distinctive linear or serpiginous erosions that tend to be clustered together on or near the greater curvature [43] (see Fig. 152-10B). In such cases, it has been postulated that the erosions result from localized mucosal injury as the dissolving tablets collect by gravity in the most dependent portion of the stomach.
***Helicobacter pylori* Gastritis** *H pylori* gastritis can be diagnosed on barium studies by the presence of thickened folds or thickened, polypoid folds in the antrum, body, or, less commonly, fundus of the stomach. [44] It is important to recognize that fold thickening is a nonspecific radiographic finding. Nevertheless, *H pylori* gastritis should be a leading consideration in the differential diagnosis of thickened gastric folds in patients with dyspepsia, epigastric pain, or other upper gastrointestinal symptoms. Patients suspected of having peptic ulcer disease can be evaluated by a noninvasive *H pylori* test combined with a double-contrast upper gastrointestinal examination. If the barium study reveals a gastric or duodenal ulcer, a noninvasive *H pylori* test can be performed to determine whether or not antibiotic therapy should be initiated as well as conventional antiulcer therapy. Such an approach might avoid the need for endoscopy in many patients.
Gastric Ulcers In the past, some researchers advocated endoscopy and biopsy of all radiographically diagnosed gastric ulcers to rule out cancer in these patients. However, several studies have found that virtually all gastric ulcers with an unequivocally benign appearance on double-contrast upper gastrointestinal examinations are benign lesions. [45], [46] In those studies, about two thirds of all benign ulcers had a benign radiographic appearance. As a result, unnecessary endoscopy can be avoided in the initial evaluation of most gastric ulcers diagnosed on double-contrast examinations. Instead, typically, benign gastric ulcers could be followed radiographically to complete healing without need for endoscopic intervention. Because endoscopy is a considerably more expensive procedure than double-contrast radiography, the potential financial savings are enormous. Benign gastric ulcers classically appear en face as round or ovoid collections of barium, often associated with a smooth mound of edema or thin, straight folds that radiate directly to the edge of the ulcer crater. [46] When viewed in profile, benign ulcers project beyond the contour of the adjacent gastric wall and are often associated with an ulcer mound or collar. In contrast, malignant gastric ulcers classically appear en face as irregular ulcer craters within a discrete tumor mass, sometimes associated with nodularity or clubbing of adjacent folds owing to infiltration of the folds by tumor. When viewed in profile, malignant ulcers project inside the gastric lumen within a tumor mass that forms acute angles with the adjacent gastric wall rather than the obtuse, gently sloping angles expected for a benign mound of edema. Most gastric ulcers detected on double-contrast studies are smaller than 1 cm in diameter. [46] Although some benign ulcers are round and symmetric, others have a rod-shaped or linear appearance. Almost all benign ulcers occur in the antrum or body of the stomach, and most are located on the lesser curvature or posterior wall [46] (Fig. 152-11). Occasionally, benign gastric ulcers may be found on the greater curvature of the distal stomach. The latter ulcers are almost always caused by ingestion of aspirin. [46] As these aspirin-induced greater curvature ulcers enlarge, they have a tendency to penetrate inferiorly into the gastrocolic ligament, occasionally leading to the development of gastrocolic fistulas. [47]

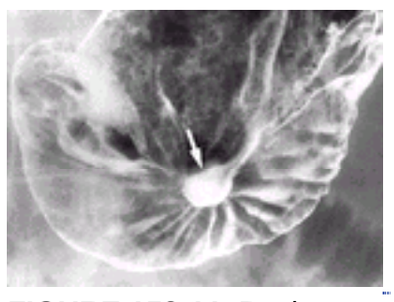

FIGURE 152-11. Benign gastric ulcer. Round, symmetric ulcer (*arrow*) is seen on the posterior gastric wall, with smooth folds radiating to the edge of the crater. This ulcer has the typical radiologic features of a benign gastric ulcer.

Ulcer healing may manifest radiographically as a decrease in the size of the ulcer and by a change in its shape. In most cases, ulcer healing produces a radiographically visible scar with a central pit or depression, radiating folds, or retraction of the adjacent gastric wall. [46]
Gastric Carcinoma Gastric carcinoma has a relatively bleak prognosis, with 5-year survival rates of only 10% to 30%. Advanced tumors may appear radiographically as polypoid or ulcerated lesions or, less frequently, as infiltrating lesions with diffuse narrowing of the stomach, producing a linitis plastica appearance. In contrast, early gastric cancer is a curable disease, with reported 5-year survival rates as high as 95%. The Japanese have developed an elaborate system for classifying these

tumors based on whether they are predominantly elevated, flat, or depressed lesions. Unfortunately, most patients in the United States with gastric carcinoma already have advanced lesions at the time of clinical presentation. As a result, early gastric cancer is unlikely to be detected on double-contrast studies or endoscopy as long as these examinations are performed predominantly on symptomatic patients. [48] The average sensitivity of single-contrast barium studies for the diagnosis of gastric carcinoma has only been about 75%. [49] Concern about missing gastric cancers on barium studies has therefore been used as a rationale for performing endoscopy as the initial diagnostic test in patients with upper gastrointestinal signs or symptoms. In a recent study, however, gastric carcinomas were diagnosed or suspected on double-contrast examinations in 96% of patients with proven lesions. [49] In a separate part of the study, only 3.5% of all patients who had double-contrast examinations during this period underwent endoscopy because of findings that were equivocal or suspicious for gastric carcinoma. Thus, a high sensitivity can be achieved in the radiographic diagnosis of gastric cancer while referring only a small percentage of symptomatic patients for endoscopy.

Gastric Lymphoma It is well recognized that chronic *H pylori* gastritis leads to the development of mucosa-associated lymphoid tissue (MALT) in the stomach. This lymphoid tissue is thought to be the precursor of low-grade B-cell gastric MALT lymphomas, which, if untreated, may undergo transformation to more high-grade lymphomas. [50] Gastric MALT lymphomas can sometimes be recognized on double-contrast upper gastrointestinal examinations by variably sized, rounded, often confluent nodules in the stomach [51] (Fig. 152-12). However, focal gastritis due to *H pylori* or other causes may produce similar findings, so that endoscopic biopsy specimens are required for a definitive diagnosis. In contrast, advanced gastric lymphoma may manifest on barium studies as thickened folds, multiple submucosal masses, centrally ulcerated bull's-eye lesions, or giant, cavitated lesions. [52]

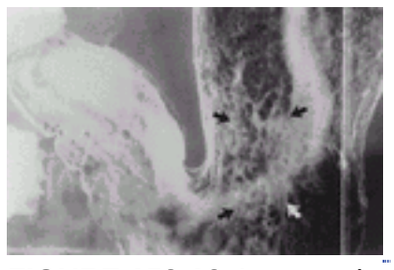

FIGURE 152-12. Low-grade gastric mucosa-associated lymphoid tissue (MALT) lymphoma. Multiple confluent nodules of varying sizes (*arrows*) are seen in gastric body. (From ref. [51].)

Other Gastric Tumors Leiomyomas are the most common benign submucosal tumors found in the stomach. These tumors usually appear on barium studies as smooth submucosal masses (with or without central ulceration) that form right angles or slightly obtuse angles with the adjacent gastric wall. Most leiomyomas are less than 3 cm in size. In contrast, malignant gastrointestinal stromal tumors usually appear as larger, more lobulated submucosal masses, often containing one or more ulcers or giant areas of cavitation. Ectopic pancreatic rests usually are located on the greater curvature of the distal antrum, appearing as small submucosal masses, sometimes with central umbilication or ulceration. Rarely, barium may reflux into rudimentary ductal structures. Gastric carcinoids may also manifest on barium studies as one or more small submucosal masses, occasionally containing central ulcers.

Duodenitis Duodenitis may manifest radiographically as thickened folds, mucosal nodules, erosions, or deformity of the bulb. [53] Duodenal erosions are detected less frequently than gastric erosions on double-contrast studies because of the difficulty in differentiating these lesions from normal mucosal pits. However, erosive duodenitis can be diagnosed when double-contrast radiographs reveal central barium collections surrounded by radiolucent halos of edematous mucosa.

Duodenal Ulcers Unlike gastric ulcers, which occur primarily on the lesser curvature or posterior wall of the stomach, as many as 50% of duodenal ulcers are located in the anterior wall. [31] Because most double-contrast radiographs are obtained with the patient in a supine or supine oblique position, the anterior wall of the duodenum is not optimally coated with barium, and anterior wall ulcers may be missed on the double-contrast portion of the study. For this reason, double-contrast views of the duodenum should be supplemented with prone compression views obtained with a low-density barium suspension to demonstrate ulcers on the anterior wall. Thus, the biphasic technique is particularly important for evaluating the duodenum.

SMALL BOWEL

The conventional small bowel follow-through examination usually consists of a series of overhead abdominal radiographs obtained at regular intervals after ingestion of barium. With this technique, the small bowel is evaluated fluoroscopically only if the overhead radiographs appear abnormal. Because this technique is extremely unreliable in detecting pathology in the small bowel, this form of examination should have no place in modern radiologic practice. Instead, adequate evaluation of the small bowel requires more detailed methods of examination, such as the small bowel meal (SBM) and enteroclysis. However, these techniques require time, effort, and interest by the radiologist. Barium studies of the small bowel should be requested only if there are good clinical indications of small bowel disease.

Techniques

Small Bowel Meal The "dedicated" or "fluoroscopic" SBM includes overhead films but uses fluoroscopy as the primary means of examining the small bowel. Unless contraindicated, metoclopramide (20 mg) is administered orally 20 minutes before the examination to accelerate transit of barium through the stomach and small bowel. The patient then ingests 500 to 600 mL of an appropriate 35% to 40% (weight/volume) suspension of barium sulfate. After a brief examination of the upper gastrointestinal tract, intermittent fluoroscopy is performed until all small bowel loops have been demonstrated. Periodic spot films are obtained to document the fluoroscopic findings.

Enteroclysis (Small Bowel Enema) A more detailed study of the entire small bowel is carried out by enteroclysis. [54] If not contraindicated, the patient swallows 20 mg of metoclopramide to accelerate transit. Surface anesthesia to the throat precedes intubation of the distal duodenum or proximal jejunum. Between 200 and 250 mL of a 75% to 80% suspension of barium is then injected, followed by infusion of 1 to 2 L of a 0.5% solution of methylcellulose in water. This biphasic examination first demonstrates the small bowel in single contrast and then in lumen-distended double contrast (Fig. 152-13). The radiologist must be present throughout the examination to assess individual bowel loops by graded compression and to document the fluoroscopic findings with periodic spot films. An overhead radiograph of the entire small bowel completes the examination.

FIGURE 152-13. Normal small bowel enema. **A:** Distal jejunum seen on a preliminary single-contrast examination. **B:** Same jejunal loops on double-contrast examination.

Application of Techniques

Small Bowel Meal The major advantage of the SBM is that it does not require intubation. Transit acceleration by oral metoclopramide is an important component of the examination because it decreases flocculation of barium and the time needed to perform the study. However, distention of the bowel lumen cannot be achieved by this technique. The SBM is an appropriate examination for evaluating established diseases of the small bowel, such as Crohn's disease. It is also adequate for investigating pathological conditions not associated with lumenal distention, such as radiation enteritis, ischemia, and hematomas. This technique can be used to demonstrate the level and degree of small bowel obstruction, although it is less successful in determining the cause.

Enteroclysis The major advantage of enteroclysis is that it combines transit acceleration with lumenal distention. Enteroclysis virtually prevents flocculation of barium. As a result, the double-contrast images permit assessment of mucosal surface detail, even through overlapping small bowel loops. The straightening of mucosal folds that occurs with lumenal distention permits detection of small, relatively subtle lesions. Normality becomes measurable in terms of wall thickness; fold thickness, shape, and height; and distance between folds. The major indications for enteroclysis include the following: *Small bowel obstruction, particularly intermittent obstruction*: enteroclysis often permits differentiation of the various causes of obstruction. [55]

Malabsorption states: enteroclysis can identify features of celiac disease, lymphangiectasia, Whipple disease, morphologic changes associated with bacterial overgrowth, and other conditions associated with malabsorption.

Crohn's disease: enteroclysis can delineate the full extent of disease, including skip lesions and fistulas, and can be used to distinguish fibrous strictures from stenotic

disease.
Small bowel tumors, particularly carcinoids, other primary and secondary malignancies, and lymphoma.
Meckel diverticulum: increased lumenal distention greatly improves its detection rate. [56]
Normality: enteroclysis is the most reliable radiologic examination for establishing morphologic normality of the small bowel.

Abnormalities

Small Bowel Obstruction Concern has been expressed regarding the possible inspissation of barium within obstructed small bowel. [57] Although animal studies and clinical experience have shown that such inspissation does not occur, [58] it may be advisable to replace barium-based techniques by CT or magnetic resonance imaging enteroclysis in patients who have high-grade or complete small bowel obstructions. [54] With lower grades of obstruction, barium enteroclysis remains the best method for delineating its site and cause, particularly in patients with intermittent obstruction. If a decompression tube has already been positioned beyond the duodenum, it should be used for the enteroclysis examination. A contrast study may also be needed when there is suspicion of small bowel perforation. In such cases, a water-soluble contrast agent is usually employed, although its high osmolality may cause an outpouring of fluid into the bowel lumen and dilution of its contrast density that degrades the quality of the examination.
Adhesions Adhesions account for almost 75% of small bowel obstructions. Enteroclysis can identify features that favor obstruction by a single band, multiple bands, or extensive adhesions. Single bands are more likely to produce high-grade obstruction. Closed-loop obstruction may be caused by prolapse of a bowel loop beneath an adhesive band. This type of obstruction is associated with twisting of bowel, with a high risk for ischemia and necrosis. Although barium studies may be helpful in the early stages of a closed loop obstruction, CT is essential for establishing the degree of vascular compromise in these patients. Multiple bands are almost always associated with adherence of the small bowel to anterior abdominal wall scars. Like extensive adhesions, they tend to produce lower-grade small bowel obstruction. Because of its ability to distend the lumen, enteroclysis draws attention to single bands as a cause of low-grade or intermittent obstruction, even when symptoms are minimal or absent.
Hernias Barium studies are not required for small bowel obstructions caused by clinically evident external hernias. However, enteroclysis is useful in detecting hernias of developmental or postsurgical origin, including peristomal, spigelian, or Richter hernias.
Malignancy In patients who have undergone laparotomy for abdominal malignancy, small bowel obstruction may be caused by adhesions, metastases, radiation, or residual tumor. These conditions can be differentiated by enteroclysis in more than 80% of patients. [54] This information can facilitate surgical management by providing a preoperative choice of resection of tumor, bypass of radiation-damaged bowel, or lysis of adhesions.
Malabsorption States Malabsorption is usually diagnosed by clinical and laboratory criteria. The purpose of barium studies is to determine the most likely cause of a clinically diagnosed malabsorption state or to document complications. In many conditions associated with malabsorption, abnormal accumulation of intralumenal fluid causes flocculation of barium on SBM. Because flocculated barium prevents accurate depiction of mucosal surface detail, enteroclysis is the preferred radiologic examination in these patients.
Celiac Disease The diagnosis of celiac disease must be firmly established to justify placing the patient on lifelong dietary restrictions. A confident diagnosis can be made in patients who have characteristic mucosal changes on duodenal biopsy and have shown a satisfactory response to gluten withdrawal. However, more than 50% of patients have atypical clinical presentations. As a result, the diagnosis may first be suggested by enteroclysis, which can demonstrate a measurably increased separation of folds in the distended proximal jejunum (Fig. 152-14). In one study, zero to three folds per inch of jejunum were found by enteroclysis in 73% of celiac patients but in only 2% of controls. [59] If coupled with the finding of “jejunization” of ileal folds, the enteroclysis recognition of celiac disease rises to 82%. Conversely, patients with five or more folds per inch of jejunum rarely had celiac disease.

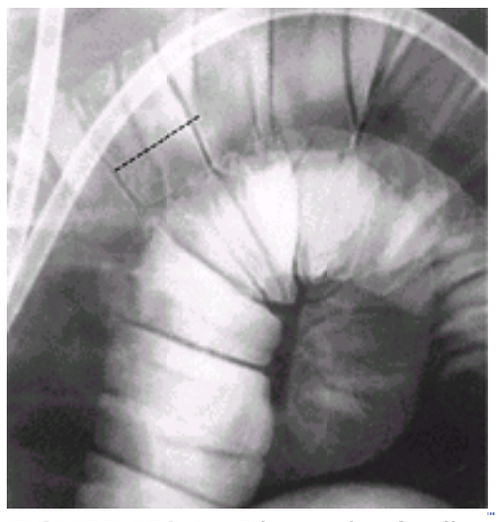

FIGURE 152-14. Diagnosis of celiac disease using a small bowel enema. The distended proximal jejunum shows an increased separation of folds, with only three folds per inch (*dotted line*).

Enteroclysis is especially important in celiac patients who relapse after an initial dietary response; enteroclysis can then distinguish between features due to dietary lapses and those of ulcerative jejunoileitis, T-cell lymphoma, and intestinal carcinoma.
Bacterial Overgrowth Syndrome Barium studies can accurately depict many structural abnormalities in the small bowel that cause stasis or contamination and lead to bacterial overgrowth. These abnormalities include strictures, blind pouch and blind loop syndromes, jejunoileal diverticulosis, and coloenteric fistulas. Other patients with bacterial overgrowth syndrome may have chronic intestinal pseudoobstruction due to scleroderma or other causes. Small bowel involvement by scleroderma may lead to intestinal dilation with crowded folds, producing the virtually pathognomonic “hide-bound” bowel sign. [60]
Intestinal Lymphangiectasia The primary and secondary forms of intestinal lymphangiectasia are manifested by nonspecific changes of fold thickening and fluid increase on conventional barium studies. With enteroclysis, it is possible to demonstrate 1- to 2-mm micronodules that represent villi distended by dilated lacteals. Whipple disease involving the small bowel may also manifest as tiny nodules due to villous distention by the enlarged lamina propria laden with foamy macrophages, bacilli, and periodic acid-Schiff–positive material. A similar nodular pattern may be demonstrated in patients with *Mycobacterium avium* complex enteritis, macroglobulinemia, immunoglobulin A deficiency, and other unusual conditions.
Crohn’s Disease Early radiologic findings of Crohn’s disease in the small bowel include a coarse villous pattern of the mucosa, thickened folds, and aphthous ulcers. The presence of linear ulcers along the mesenteric border of the bowel with pleating or sacculation of the uninvolved antimesenteric border is virtually diagnostic of Crohn’s disease (Fig. 152-15). With progression of disease, transaxial extension of linear ulcers and fissures produces a cobblestone appearance with narrowing of the lumen. Further progression of Crohn’s disease leads to complications such as fibrous strictures, fistulas, inflammatory masses, and abscesses.

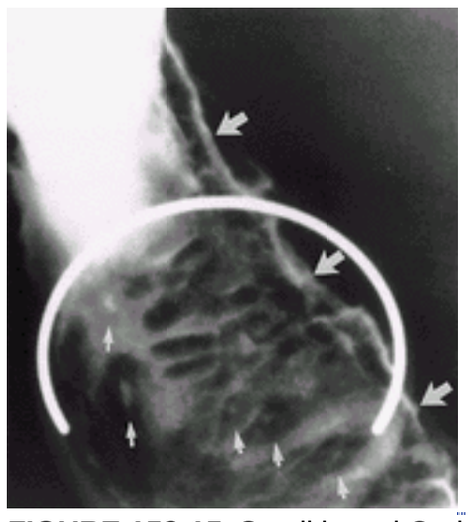

FIGURE 152-15. Small bowel Crohn’s disease. Spot film from enteroclysis examination shows a long, linear ulcer (*large arrows*) on the mesenteric border of the bowel with multiple aphthous ulcers (*small arrows*) and thickened, interrupted folds. The linear mesenteric border ulcer is characteristic of Crohn’s disease.

Many features of Crohn's disease can be demonstrated as effectively by the SBM as by enteroclysis. Disease of the terminal ileum is particularly well shown by combining the SBM with a peroral pneumocolon (i.e., introducing barium by mouth and air by the rectum). [61] However, enteroclysis is the optimal radiologic examination for detecting early lesions, particularly the more proximal skip lesions. It is also a better examination for distinguishing Crohn's disease from other pathological conditions such as lymphoma, tuberculosis, ischemia, and metastatic disease.

Malignant Tumors

Adenocarcinoma Most primary adenocarcinomas of the small bowel are advanced tumors with regional lymph node involvement at the time of clinical presentation. In the absence of obstruction, these tumors can be missed on conventional small bowel follow-through examinations. Enteroclysis is the best radiologic technique for demonstrating these lesions at a less advanced stage (Fig. 152-16).

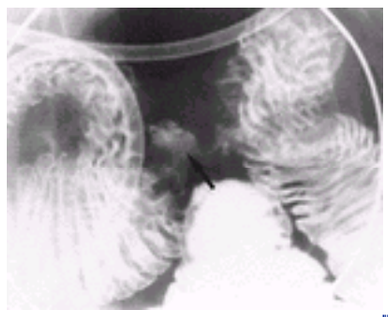

FIGURE 152-16. Annular adenocarcinoma of the proximal jejunum with a central ulceration (*arrow*).

Non-Hodgkin Lymphoma Primary small bowel lymphomas typically appear as cavitary lesions extending into the mesentery or as segmental infiltrating lesions. In other patients with disseminated lymphoma, barium studies may demonstrate submucosal nodules of various sizes. An enlarging mass of mesenteric nodal lymphoma can secondarily infiltrate a loop of small bowel. [54] CT can be used to delineate better the extent of mesenteric and lymph node involvement.

Other Malignancies

Small bowel carcinoids can be diagnosed by enteroclysis at the stage of incipient transmural extension of the primary tumor or tumors (Fig. 152-17). More advanced carcinoids are typically associated with distorted folds. Enlarging mesenteric metastases produce mass effect and secondary fibrotic alterations in adjacent bowel loops. Early carcinoids may appear as small, polypoid nodules, more often in the terminal ileum.

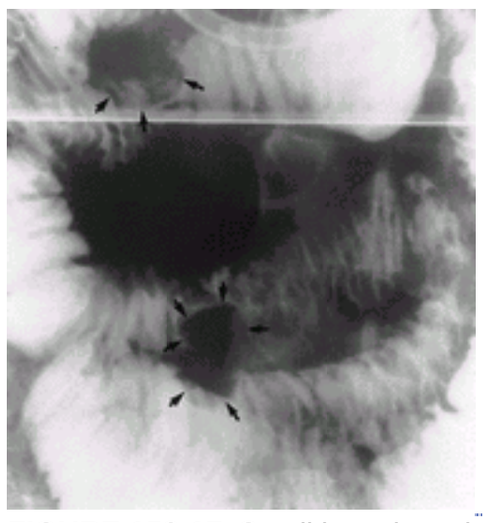

FIGURE 152-17. Small bowel carcinoids. Discrete submucosal masses (*arrows*) are seen in the mid small bowel. Also note the separation of small bowel loops caused by spread of tumor into the mesentery.

Both hematogenous and intraperitoneal-seeded metastases to the small bowel tend to occur as multiple lesions, often associated with signs of obstruction. Gastrointestinal stromal tumors may appear as excavated, exoenteric masses.

Meckel Diverticulum

Enteroclysis is the best radiologic technique for diagnosing Meckel diverticulum, with an overall detection rate comparable to that found at autopsy. [56] The diverticulum arises from the antimesenteric border of the distal ileum, forming a blind sac, with a characteristic mucosal fold pattern at its site of origin. Occasionally, it is possible to identify a defect in the diverticulum due to ectopic gastric mucosa. It is also possible to identify an invaginated Meckel diverticulum presenting with low-grade obstruction.

Obscure Blood Loss

In patients with unexplained gastrointestinal blood loss, enteroclysis is indicated when radiologic and endoscopic examinations of the upper gastrointestinal tract and colon fail to demonstrate a source of bleeding. Enteroclysis has a diagnostic yield of 21% in identifying the presumptive source of obscure gastrointestinal bleeding. [62] Most patients have tumors involving the small bowel; other diagnoses include ischemia, arteriovenous malformations, Meckel diverticulum, and celiac disease.

COLON

Since the 1920s, the barium enema has been used as the primary nonsurgical method for investigating the colon. Since the 1970s, colonoscopy has become an important diagnostic modality for evaluating the colonic mucosa and for removing polyps. Cross-sectional imaging techniques, particularly CT, have also become important for evaluating the extramucosal extent of disease. CT and ultrasound may also be used to assess bowel wall thickness. Because of its simplicity, low cost, safety, and accuracy, however, the barium enema remains a useful diagnostic technique for investigating the colon.

Single versus Double Contrast

Radiologic examination of the colon can be performed by a single- or double-contrast technique. In single-contrast studies, the entire colon is filled with a relatively low-density barium suspension. The examination is performed under fluoroscopic control, with extensive palpation and compression of the colon as it fills. A postevacuation radiograph is usually obtained to demonstrate additional mucosal detail. In double-contrast studies, a smaller volume of high-density barium is introduced into the colon, followed by insufflation of air. With this technique, the mucosal surface is coated by a thin layer of high-density barium, and the lumen is distended with air.

The double-contrast examination is considered to be the best radiologic technique for demonstrating fine mucosal lesions such as small polyps and the early changes of inflammatory bowel disease (IBD). [63], [64] It can also better demonstrate segments of the large bowel that are inaccessible to palpation, such as the rectum and the hepatic and splenic flexures. The single-contrast barium enema is the preferred technique when careful control of the flow of barium is required. A single-contrast

study should be performed in patients with suspected obstruction, diverticulitis, fistulas, or Hirschsprung disease. This technique is also frequently used in patients who are too old or debilitated to tolerate a double-contrast study.

Radiology versus Colonoscopy

There is ongoing controversy about the relative roles of radiology and endoscopy in investigating colonic disease. In general, the choice of technique depends on the clinical setting and the relative skill and experience of the examiners. Nevertheless, it should be recognized that colonoscopy is primarily of value for detecting mucosal lesions. It should not be used as the primary diagnostic modality when the patient's symptoms suggest an intramural or extrinsic lesion involving the bowel.

Colonoscopy probably is more accurate than radiology in demonstrating subtle mucosal abnormalities in the colon. When compared with high-quality, double-contrast barium enemas, this benefit applies primarily to the detection of polypoid lesions less than 1 cm in diameter and the early, preulcerative changes of IBD. However, this benefit must be balanced against the higher cost and complication rate of colonoscopy. The colonoscopist is unsuccessful in advancing the endoscope to the cecum in about 10% of patients, [65] but the cecum is almost always seen on barium enema studies.

We recommend contrast examination of the colon for patients with symptoms that could be caused by diseases of the colon (e.g., abdominal pain, constipation, change in bowel habit, and anemia). Colonoscopy is generally the examination of choice for the evaluation of uncertain radiologic findings, for removal or biopsy of lesions found at barium enema, and for the evaluation of patients who are at high risk for developing colorectal cancer or who are positive for fecal occult blood.

Indications

Although radiologic examination of the colon may be performed for a wide variety of indications, the most common include the following: detection of colorectal polyps and cancer; assessment of the type, extent, and severity of IBD; diagnosis of diverticular disease and its complications; evaluation of extrinsic mass lesions involving the colon; and evaluation of the rectum.

Colorectal Polyps and Cancer The barium enema is a valuable diagnostic test for patients with symptomatic colorectal cancer. It is a valuable technique for diagnosing the primary lesion and for detecting synchronous carcinomas elsewhere in the colon. The double-contrast barium enema has an overall accuracy of about 95% in diagnosing colorectal cancer. However, there appears to be an irreducible minimum error rate of about 5%, primarily the result of perceptive error. [66] The double-contrast barium enema is also used to evaluate patients who have overt rectal bleeding or occult blood in the stool on routine screening tests for colorectal cancer. In such cases, the goal is to detect invasive carcinomas and small adenomas that are precursors of colonic carcinoma (Fig. 152-18). These adenomatous polyps should be removed endoscopically to prevent the subsequent development of cancer. [67]

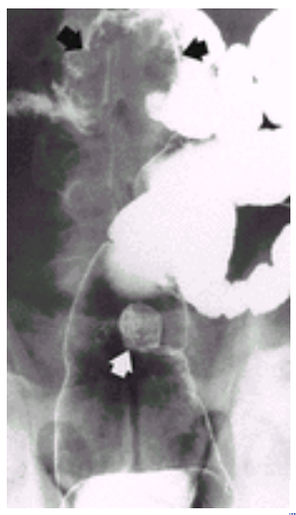

FIGURE 152-18. Example of a coexisting carcinoma and benign adenoma in the same patient. Notice the polypoid carcinoma (*black arrows*) in the transverse colon and small tubulovillous adenoma (*white arrow*) in the distal sigmoid colon.

The double-contrast barium enema also has a role in the surveillance of patients with conditions that are known to predispose patients to the development of colorectal carcinoma. Such high-risk patients include those with a personal or family history of colon cancer or polyps and those with a history of chronic ulcerative colitis. For these high-risk groups, we recommend a screening program that alternates colonoscopy and double-contrast barium enema to maximize the diagnostic yield. Contrast examination of the colon may also have a role in the routine screening of patients for colorectal cancer. Eddy and colleagues suggested that annual fecal occult blood testing and barium enema every 5 years may be the most cost-effective way of reducing the mortality from colorectal cancer. [68]
Inflammatory Bowel Disease Radiologic examination of the colon serves a variety of purposes in patients with known or suspected IBD. It can establish the presence of disease in patients who have not had prior sigmoidoscopy and in those who have had a egative sigmoidoscopy because the disease did not involve the rectosigmoid colon. It can also define the extent of disease in the colon. This information is particularly important in patients with ulcerative colitis because the risk for developing carcinoma is related to the extent of colonic involvement. Radiologic examination of the colon can differentiate ulcerative from granulomatous colitis. In typical cases, ulcerative colitis is characterized on double-contrast radiographs by a granular mucosa involving the rectum and extending proximally to a variable degree (Fig. 152-19A). In contrast, granulomatous colitis is characterized by a progression from discrete aphthous ulcers (see Fig. 152-19B) to transmural disease with deep ulcers, fissures, fistulas, and abscesses. The rectum is often spared, and colonic involvement tends to be discontinuous and patchy.

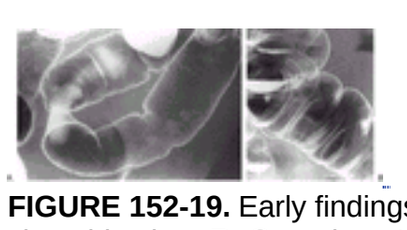

FIGURE 152-19. Early findings of inflammatory bowel disease detected by a double-contrast barium enema. **A:** Ulcerative colitis with typical granular mucosa in the sigmoid colon. **B:** Granulomatous colitis with discrete, aphthous ulcers in the transverse colon.

Radiologic examination also can detect the complications of chronic ulcerative or granulomatous colitis, including the development of strictures, abscesses, fistulas, and inflammatory or postinflammatory polyps. In patients who develop colonic carcinoma, the tumor can be diagnosed, and in some cases, the development of dysplasia or "precancer" can be recognized on double-contrast studies. [69]
Diverticular Disease Diverticular disease is one of the most common afflictions of Western society. The presence of massive diverticulosis poses a particular dilemma in interpreting double-contrast studies because the multiplicity of ring shadows makes it difficult to differentiate diverticula from polyps. As a result, single-contrast studies may be easier to interpret than double-contrast studies in patients with severe diverticular disease. Diverticulitis is a complication of diverticulosis in which a diverticular perforation leads to the formation of a pericolic abscess. Because barium may extravasate into the abscess, it is important to control the flow of barium into the colon in these patients. We therefore prefer single-contrast technique when a barium enema is performed for suspected diverticulitis (Fig. 152-20). In recent years, however, CT has increasingly been used as the primary radiologic modality in patients with suspected diverticulitis because of its relatively high sensitivity and specificity for diagnosing this condition. [70]

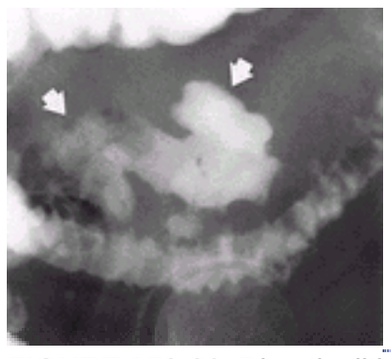

FIGURE 152-20. Diverticulitis with pericolic abscess. Barium has extravasated from a perforated diverticulum into an abscess (*arrows*). Notice the diverticula in the sigmoid colon.

Extrinsic Mass Lesions In patients with abdominal or pelvic masses, the barium enema is a useful technique for determining whether the colon is displaced, compressed, or actually invaded by these lesions. The barium enema can also detect intraperitoneal seeding of the colon by metastatic tumor, inflammatory lesions, or endometriosis. In many cases, the barium study must be correlated with CT or other cross-sectional imaging modalities to determine the true extent of disease in the abdomen.

Rectum When barium enemas were performed primarily by the single-contrast technique, the rectum was not considered to be in the province of radiology. With the use of the double-contrast technique, the rectum has become one of the simplest portions of the large bowel to evaluate radiographically. The anatomy of the rectum, including the columns of Morgagni and the valves of Houston, can be demonstrated in exquisite detail. [71] Radiologic evaluation of the rectum is particularly important for the diagnosis of rectal tumors, IBD, and other unusual conditions, such as the solitary rectal ulcer syndrome. [72] The double-contrast examination can also detect rectal carcinomas that are missed by digital rectal examination and proctoscopy [73] (Fig. 152-21). The routine double-contrast barium enema, therefore, should include a careful examination of the rectum.

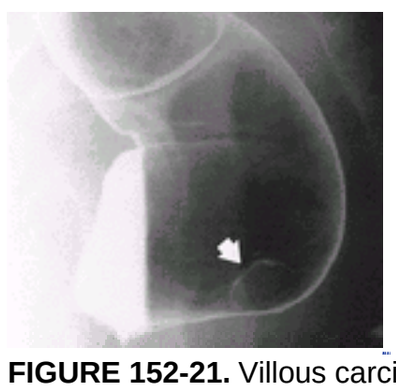

FIGURE 152-21. Villous carcinoma (*arrow*) of the posterior wall of the rectum that had been missed on prior digital and proctoscopic examinations.

Contraindications

The barium enema examination is generally a safe procedure, but there are several conditions for which the study is contraindicated or should be performed with extreme caution. These conditions include toxic megacolon, ischemic colitis, or other diseases in which the bowel wall is friable and more likely to perforate during the procedure. When a colonic perforation is suspected, the radiologic examination should be performed with water-soluble iodinated contrast material that can be reabsorbed from the peritoneal cavity if extravasation occurs.

Complications

The most feared complication of the barium enema is colonic perforation. [74] When this complication occurs, the perforation usually involves the intraperitoneal portion of the colon. Extravasated barium in the peritoneal cavity may be seen outlining loops of bowel. Surgical treatment is usually required. On some double-contrast barium enemas, a colonic perforation may manifest as only a pneumoperitoneum without extravasation of barium into the peritoneal cavity. If these patients are aggressively treated with intravenous fluids and antibiotics, surgery may be avoided in some cases.

Retroperitoneal perforations usually result from laceration of the rectum by an inflated retention balloon on the enema tip in cases of diffuse rectal disease, such as ulcerative or radiation proctitis. In such cases, barium may be observed radiographically in the rectal wall or in the perirectal soft tissues. Because of the risk for rectal laceration, it is our practice not to inflate the retention balloon routinely. When the balloon is required in patients who have poor anal sphincter tone, it should be inflated under careful fluoroscopic control after barium has been instilled into the rectum.

Despite occasional complications, the barium enema examination is a safe, simple, and relatively inexpensive procedure. It provides reliable information about the nature and extent of mucosal disease and about disease within and outside the bowel wall. Most patients undergo barium enema to rule out colonic neoplasm and IBD, and a double-contrast study is the best radiologic technique for evaluating these individuals. In other patients with suspected diverticulitis or colonic obstruction, a single-contrast barium enema may be performed. Whether single- or double-contrast technique is used, the barium enema examination has few contraindications, and the complications can be minimized by careful attention to the clinical history and examination technique. In complicated or difficult cases, correlation of the radiologic and endoscopic findings may be necessary to clarify the nature and extent of colonic disease.

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CHAPTER 153

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ULTRASONOGRAPHY

INTRODUCTION

Basic Principles of Diagnostic Ultrasound

Ultrasonography is complex and technically challenging. It requires knowledge of physics, instrumentation, ultrasound anatomy, pathology, and, most important, scanning technique. Small handheld scanners make the concept of ultrasonography as an extension of the physical examination a real possibility. Those wishing to use ultrasonography, however, must learn the technique adequately, or erroneous results are inevitable. At least 4 months of full-time intensive training is needed for those wishing to perform even the simplest ultrasonographic evaluation.

Ultrasound refers to frequencies of sound above the audible range—about 20 to 20,000 cycles per second (20 to 20,000 Hz). Diagnostic ultrasonography uses sound frequencies far higher than this, in the 1 to 20 million Hz range (i.e., 1 to 20 MHz). Diagnostic ultrasound systems transmit sound pulses into the body. The echoes reflected from tissue are then processed to produce ultrasound images. The heart of a modern transducer is a complex array of specially manufactured piezoelectric elements that convert electrical energy into sound waves when transmitting and then convert the faint returning sound into electrical signals that can be processed into images.

In sonograms, the depth of any tissue is determined by timing how long it takes an echo to return, then calculating depth (distance from the transducer) based on an assumed average velocity of sound in human tissue. This is the pulse-echo principle. The earliest returning echoes arise from tissue reflectors closer to the transducer; later echoes come from progressively deeper reflectors. The exact anatomic location of each reflector may be calculated from the known direction of the

sound beam in the body and the measured time of arrival of its echo. Repeated pulses are used to generate a two-dimensional image of the region interrogated.

Acoustic interfaces reflect sound and generate the echoes that make up the returning ultrasound information. Acoustic interfaces occur where two tissues of different acoustic impedance are in contact. Acoustic impedance is related to tissue density and the speed of sound in the tissues. The greater the acoustic impedance difference between the two tissues at an interface, the greater the percentage of incident sound reflected and the stronger the returning echo. This explains why ultrasound cannot “see through” gas or bone. The soft tissue–bone and soft tissue–gas interfaces have huge acoustic impedance differences, causing nearly all sound to be reflected. Clearly, if nearly all the sound beam is reflected, tissues deep to the bone or gas will not be imaged.

The amplitude (loudness) of echoes reflected from any given tissue is represented by different shades of gray. Most ultrasound images use a black background display. With this display convention, whiter shades of gray represent higher-amplitude echoes. Descriptively, echo-free structures are said to be *anechoic*. *Isoechoic* means identical in echogenicity to another tissue. *Hypoechoic* means lower in echogenicity than another tissue, whereas *hyperechoic* (or *echogenic*) means higher in echogenicity than another tissue.

The ultrasound beam is progressively attenuated (weakened) as it travels deeper into the body. Soft tissues are more attenuative than fluid-filled structures. Cystic structures cause little attenuation of sound, so that tissues deep to the gallbladder or liver cysts appear more echogenic (brighter echoes) than do neighboring areas at a similar depth. This is called *distal acoustic enhancement* or *through-transmission* of sound.

There is a tradeoff between the use of high-frequency and low-frequency transducers in ultrasound imaging. Low-frequency sound penetrates tissue effectively, facilitating good images of deep tissues (e.g., large livers). Higher-frequency transducers produce higher-resolution images but cannot penetrate sufficiently well to produce images deeper than relatively superficial structures. As a practical matter, abdominal organ ultrasonography is done with lower-frequency transducers (3 to 7 MHz). When the bowel itself (e.g., appendiceal ultrasound) is evaluated, higher-frequency transducers (5 to 10 MHz) are generally used. It is generally best to choose the highest-frequency transducer that allows adequate penetration.

Artifacts are a significant problem in ultrasound imaging. They may be related to equipment problems, but unavoidable artifacts caused by the interaction of sound and tissue are the most troublesome. Reverberation of sound between tissue and the face of the transducer can obscure detail, most notably in the near field. Blockage of sound results in acoustic shadowing—a “void” in the image deep to the structure causing shadowing. Acoustic shadowing may be complete or partial. Reflection, refraction, or absorption of the sound beam may cause acoustic shadowing. Shadowing occurs deep to obstructing objects (such as gallstones, bone, or gas) or deep to curved interfaces at the margin of cysts. Shadowing is one of the diagnostic criteria for gallstones; it can be useful in distinguishing gas or calcification from other highly echogenic reflectors. The mirror image and multipath reflection artifacts can cause tissue to appear at an incorrect depth or location, simulating, for example, liver tissue above the diaphragm, or an apparent double inferior vena cava (IVC). Side-lobe and partial thickness artifacts may lead to spurious hazy echoes that are most evident when superimposed on echo-free structures (e.g., the gallbladder). Comet-tail artifacts appear as highly echogenic “tails” deep to small intense reflectors, such as small air bubbles, surgical clips, small cholesterol crystals, and shotgun pellets.

Tissue harmonic imaging is a newer gray-scale technique that results in less image artifact. It uses the nonlinear sound produced by the ultrasound beam passing through tissue to produce images. This is distinct from conventional ultrasound, which produces images from echoes (reflected sound). Because of the decrease in artifact, tissue harmonics are becoming more popular as a way to obtain gray-scale images.

Doppler Ultrasonography

Doppler ultrasonography is a virtually routine component of many modern ultrasonographic examinations. It adds dynamic, real-time flow information to the morphologic images provided by gray-scale imaging. Flowing blood is detected by its Doppler frequency shift. Stationary soft tissue lacks a Doppler frequency shift; echoes without a Doppler shift are ignored in the production of flow images.

Color Doppler ultrasonography passively and automatically displays color-coded flow information superimposed on all or a selected portion of the gray-scale image. It detects flow and displays both flow direction and frequency shift (a rough estimate of flow velocity). Flow direction is usually displayed as red or blue; velocity estimates are depicted as different hues or a change in color saturation. Color Doppler ultrasonography is invaluable when flow detection or global anatomic information about blood flow is needed. The ability of color Doppler ultrasonography to display flow passively in real time minimizes the chance of missing flow in an unexpected area. It facilitates comparison of flow in different anatomic locations. Color Doppler ultrasonography has the advantage of displaying both flow and anatomic information in a real-time image, which is much easier to understand than a combined gray-scale and spectral “duplex Doppler” display.

Spectral Doppler acquires detailed flow information from a small area (the sample volume or range gate). Spectral Doppler is often erroneously called *duplex Doppler*. In fact, both color and spectral Doppler are duplex—both display flow and morphologic information simultaneously. Spectral Doppler is useful when detailed quantitative information about flow velocity or pulsatility is important (e.g., in liver transplantation). Spectral Doppler information is displayed graphically as a time-velocity waveform, scrolled out across the screen. Spectral Doppler information can also be processed into an audible signal that is very useful to experienced examiners. For example, arteries can be distinguished from veins audibly, and the higher-pitched sound of stenosis can be useful in detecting arterial narrowing. In many situations, color Doppler ultrasonography is first used to survey a vessel to facilitate fast and accurate spectral Doppler sample volume placement.

Ultrasound Contrast Agents

As of this writing, however, no ultrasound contrast agent has received U.S. Food and Drug Administration approval for radiologic imaging. Limited approval has been granted for cardiac ultrasonography. Although in its infancy in the United States, significant contrast research, as well as daily clinical use, is occurring worldwide. Use of these agents improves the diagnostic power of ultrasound imaging and may expand ultrasound’s current role into diagnosis of conditions for which computed tomography (CT), magnetic resonance imaging (MRI), and nuclear medicine are dominant. Most new ultrasound contrast agents are intravenously injected microbubbles.

Microbubble ultrasound contrast agents enhance the useful ultrasound signal reflected back to the ultrasound transducer. The increased signal is depicted as an increase in echogenicity [1] or increase in signal strength. Ultrasound contrast agents thus affect gray-scale imaging as well as color or spectral Doppler imaging.

Vascular enhancement may involve several types of improvement in examination quality. First, greater portions of vessels are seen, and smaller, deep vessels are visible. Second, spectral Doppler signal from a vessel is increased. Third, vascular signals are enhanced, so that flow, stenosis, and thrombus determinations are made with more confidence. [2]

Even though some ultrasound contrast agents do not enter the tissues, all traverse both large vessels and small capillaries. Thus, on gray-scale imaging, they can appear to enhance an entire organ rather than merely its large arteries and veins. Gray-scale enhancement is a new method of ultrasound imaging that may dramatically change ultrasonographic tumor evaluation and diagnosis. [3]

Gray-scale enhancement with harmonic imaging dramatically increases the conspicuity of the ultrasound signal from microbubbles. Harmonic imaging takes advantage of the resonance of the microbubbles during insonation. Benefits of harmonic imaging and other techniques include a significantly increased signal-to-noise ratio of the image.

Strengths and Weaknesses Compared with Other Modalities

When ultrasonography can image the area of clinical interest, it is a nearly ideal modality. The ability of diagnostic ultrasonography to display flow and soft tissue in real time is unique among imaging techniques. Its spatial resolution is superior to CT and MRI. It is safe and tolerated well by patients. When needed, ultrasound can be performed quickly and at the bedside. Unfortunately, there are many circumstances in which optimal, or even adequate, images cannot be obtained. The single most important problem is the inability of ultrasonography to see beyond gas–soft tissue and bone–soft tissue interfaces. This makes comprehensive survey scanning essentially impossible. CT, especially, and MRI are better survey modalities.

Many abnormalities displayed automatically on whole-body CT or MRI are either impossible or very difficult to visualize ultrasonographically. Finally, ultrasonography is technically challenging—scanning to obtain optimally diagnostic images is more of an art than a science. Producing acceptable diagnostic images

ultrasonographically is usually harder than obtaining acceptable CT or MRI images. Ultrasonography, however, is often superior to CT and MRI in patients who are uncooperative, unable to hold their breath, or unable to remain relatively still during examination. Ultrasonography is often superior in patients with little body fat. This makes ultrasonography very useful in pediatric imaging, for example. Patient factors that impede optimal ultrasonography include extreme obesity and limited cutaneous access, such as burns, incisions, dressings that cannot be removed, and cutaneous gastrointestinal (GI) enterostomies.

LIVER

Indications for Ultrasonography

Primary indications include suspected or known cirrhosis, suspected liver abscess, suspected liver tumor, suspected vascular abnormalities, trauma, and transplantation. Ultrasonography can also be used to characterize abnormalities found on other imaging examinations. Its safety and relatively modest cost make it an ideal means to assess the therapeutic response of known lesions, or to sequentially follow liver lesions of questionable significance. Ultrasonography is often a simple and effective way to guide percutaneous aspiration, drainage, biopsy, or tumor ablation.

Patient Preparation and Technique

Hepatic ultrasonography can be performed without special preparation. Because the other upper abdominal organs are often evaluated with the liver, an overnight fast or 8 to 12 hours of a clear liquid diet is recommended to decrease bowel gas and distend the gallbladder. It is difficult to image the entire liver, even in the best circumstances. Difficult areas include the superficial liver above the costal margin, the left tip of the lateral segment of the left lobe, and the ventral subdiaphragmatic regions. [4]

The liver is best imaged with the patient in the supine and left lateral decubitus positions, starting with 3- to 7-MHz curved linear array transducers. A subcostal acoustic window should be used first, supplemented with intercostal scans. Small sector transducers should be used to image areas inaccessible with the larger curved linear transducers. The liver surface (usually the ventral left lobe) should be evaluated for nodularity with a near-field optimized 5- to 12-MHz linear array transducer. It is easier to appreciate subtle nodularity during real-time examination than on hard-copy images. Routine color-flow imaging is useful in patients with suspected liver pathology. Optimal color-flow and spectral Doppler ultrasonography of the liver generally requires relatively low frequency (2- to 3-MHz) scanning and good acoustic access.

Normal Anatomy

The liver parenchyma is of uniform medium echogenicity, interspersed with vessels and fissures. The liver parenchyma is slightly less echogenic than the spleen, equal to or slightly more echogenic than the renal cortex, and isoechoic or somewhat less echogenic than the pancreas.

The liver has three lobes—the large right lobe, the left lobe, and the small caudate lobe. Ultrasonographic landmarks can be used to segregate the liver into lobes and segments (Fig. 153-1 and Fig. 153-2). Fissures and ligaments can be identified because they are echogenic, fat-containing structures. Portal veins, with their accompanying bile ducts and hepatic arteries, make up the portal triad. These triads are encased in echogenic fibrofatty tissue. This echogenic margin is, as a practical matter, the feature that allows differentiation from hepatic veins. Portal veins, hepatic veins, hepatic arteries, and bile ducts can also be identified with Doppler imaging. Within the porta hepatis, the left and right bile ducts lie anterior to the portal veins. This relationship is not constant more peripherally. A transverse sonogram through the porta hepatis is one of the simplest methods for determining the presence of intrahepatic biliary obstruction.

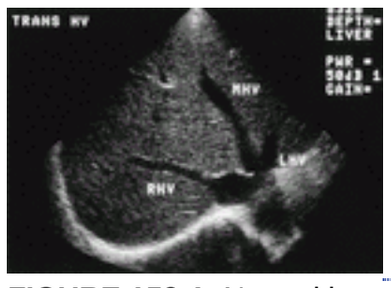

FIGURE 153-1. Normal hepatic veins. Transverse ultrasound of the upper portion of the liver shows the right hepatic vein *(RHV),* middle hepatic vein *(MHV),* and left hepatic vein *(LHV)* entering the inferior vena cava. Note the relative paucity of echogenic material around the hepatic veins, a characteristic that distinguishes them from portal veins, which have peripheral echogenic fibrofatty tissue. The middle hepatic vein separates the right and left lobes. The right hepatic vein and left hepatic vein separate the segments of the left and right lobes.

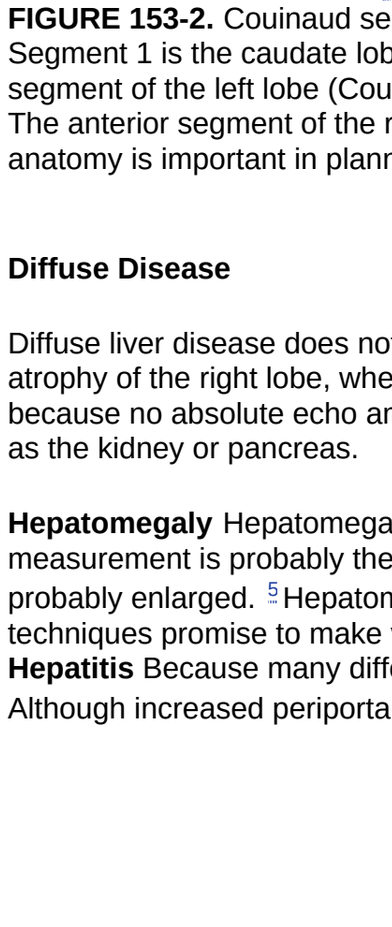

FIGURE 153-2. Couinaud segments. Transverse sonogram through the left and right portal vein, the axis through which the liver is divided into Couinaud segments. Segment 1 is the caudate lobe, bordered ventrally by the fissure for the ligament of venosum (*curved arrow*) interposed between the caudate lobe, and the lateral segment of the left lobe (Couinaud segments 2 and 3). The medial segment of the left lobe is composed of Couinaud segments 4a (cephalically) and 4b (caudally). The anterior segment of the right lobe is composed of segments 5 and 8 and the posterior segment of the right lobe of segments 6 and 7. Couinaud segmental anatomy is important in planning hepatic surgery.

Diffuse Disease

Diffuse liver disease does not always cause distortion of liver anatomy or architecture. This can make ultrasonographic detection difficult. Liver surface nodularity and atrophy of the right lobe, when present, can be useful signs of cirrhosis. Parenchymal echogenicity may be increased in diffuse disease but is difficult to evaluate because no absolute echo amplitude standard exists (analogous to CT attenuation numbers). Liver echogenicity is judged by comparison with adjacent organs, such as the kidney or pancreas.

Hepatomegaly Hepatomegaly can be difficult to diagnose objectively with ultrasonography. The normal span of the adult liver is 15 to 17 cm. The most reliable measurement is probably the sagittal dimension from the dome to the tip of the right lobe, measured at the midclavicular line. If this exceeds 15.5 cm, the liver is probably enlarged. [5] Hepatomegaly can be confidently diagnosed when the liver extends caudal to the right kidney and the left lobe is of normal size or larger. New techniques promise to make volumetric measurements feasible in the future.

Hepatitis Because many different diseases cause hepatic inflammation, ultrasonographic findings are variable. The liver is generally normal in acute viral hepatitis. Although increased periportal echoes coupled with decreased parenchymal echogenicity have been described, [6], [7] in a recent series, only 19 of 791 patients had this

pattern. [8] In the same study, there was no difference in ultrasound findings between a normal control group and patients with acute viral hepatitis. Striking irregular gallbladder wall thickening is sometimes present in patients with acute hepatitis, especially hepatitis A. [9] Direct inflammation and edema cause wall thickening, sometimes reaching 20 mm (normal, less than 3 mm). Hepatomegaly and inhomogeneous patchy or diffuse increased echogenicity are common in chronic hepatitis and are related to the amount of fatty infiltration and fibrosis present. The liver surface is smooth, unless cirrhosis is present. Enlarged arteries are noted on color Doppler ultrasonography because of increased arterial flow. This may cause a double-channel sign on gray-scale images that can potentially be confused with biliary dilation. Lymph nodes may be seen in the hepatoduodenal ligament. [10] Ultrasound findings in alcoholic hepatitis depend on the amount of fibrosis or fat present. Patchy, irregular, increased parenchymal echogenicity is usually present. The liver is often enlarged, but the surface is usually smooth, in contrast to the nodularity sometimes detected in cirrhosis. Similar to chronic hepatitis, the arteries may be enlarged.

Fatty Infiltration Severe fatty infiltration often results in an enlarged liver with diffuse increased echogenicity (Fig. 153-3). Acoustic penetration may be decreased, resulting in indistinctness of the diaphragm. The liver surface is smooth. Fatty infiltration is often patchy or focal. Focal fat appears as an area of increased echogenicity. A less affected region ("spared" area) may appear as a conspicuous hypoechoic area. Both focal fat and spared areas have a tendency to be pyramidal, with flame-shaped tapered margins. Frequent locations include the region of the porta hepatis, near the falciform ligament, the dorsal left lobe, and the caudate lobe. Although both focal fatty infiltration and focal sparing can simulate neoplasm, [11], [12] and [13] an appreciation of the usual appearance and location generally suffices to avoid confusion. Occasionally, noncontrast CT scanning, MRI, or biopsy may be required to clarify the diagnosis. Geographic fatty infiltration typically has well-defined margins between areas of greater and lesser involvement.

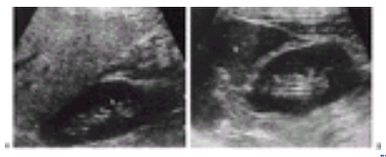

FIGURE 153-3. Fatty infiltration of the liver. Longitudinal sonogram (**A**) of the liver shows that the liver is much more echogenic than the right kidney (*k*). The echogenicity of the splenic parenchyma (*S*) and left renal cortex sonogram (**B**) are nearly equivalent. This internal comparison of the liver and spleen to adjacent kidneys allows a more reliable determination of the increased liver echogenicity associated with fatty infiltration.

Cirrhosis Ultrasonographic findings of cirrhosis include changes in the shape of the liver, parenchymal inhomogeneity, and nodularity of the liver, notably at the surface. Intrahepatic vessels may be indistinct. Unfortunately, these signs are both insensitive and insufficiently specific to diagnose cirrhosis reliably. The caudate and left lobes tend to be relatively less affected by cirrhotic scarring than the right lobe. This sometimes results in a small right lobe with left and caudate lobe hypertrophy, especially in hepatitis B. Ratios comparing the size or volume of the caudate to the shrunken right lobe have been used to diagnose cirrhosis. [14], [15] These ratios may not be practical, however, because a large series revealed a sensitivity of 43% and an accuracy of 79% for the caudate lobe–to–right lobe ratio. [16] Evaluation of the smoothness or nodularity of the liver surface using a high-resolution linear array transducer is useful. [17], [18] and [19] Surface nodularity may be the only ultrasonographic sign of irrhosis. Some studies have not confirmed the usefulness of evaluating surface nodularity. [19] We believe that the specificity for macronodular cirrhosis is good, although micronodular cirrhosis is often missed. The only other significant cause of surface nodularity is multiple subcapsular tumor nodules, usually from metastasis. Color Doppler ultrasonography may detect portal vein flow reversal or portal collaterals, prompting the diagnosis of portal hypertension. These may be the only findings indicating severe liver disease in an otherwise normal study. Enlarged tortuous arteries are sometimes imaged by color Doppler ultrasonography in cirrhotic livers. This finding, similar to "corkscrew" arteries seen angiographically, probably occurs because of truncation of arteries from liver atrophy related to cirrhosis, coupled with the increased arterial flow [20] that occurs when portal venous flow decreases. Focal fatty infiltration, hepatocellular carcinoma (HCC), regenerating nodules (adenomatous hyperplastic nodule), or other focal lesions may occur in a cirrhotic liver. A liver mass in a cirrhotic patient strongly suggests the possibility of HCC.

Hepatic Vascular Disease

Portal Hypertension Ultrasonographic evaluation of portal hypertension includes evaluation of the portal venous system and a search for portosystemic collaterals. The normal main portal vein measures slightly more than 1 cm in diameter. Although portal vein size is not useful, a lack of respiratory variation in size may be useful in diagnosing portal hypertension. [21] Normally, blood flows toward the liver (hepatopedal) in portal veins. Reversed (hepatofugal) flow, although often associated with collaterals, [20] may be the sole indication of portal hypertension. Other portal flow abnormalities include bidirectional flow and, rarely, nearly static blood flow. Ultrasound contrast agents can be useful in aiding the detection of flow in the main portal vein and other hepatic vessels. [22] Detection of portal collaterals may prompt diagnosis of unsuspected portal hypertension, or reinforce it when suspected. Portal collaterals should be sought with color Doppler ultrasonography because they are often invisible on gray-scale images. The most common collaterals are left gastric (coronary) and paraumbilical veins. Left gastric collaterals, although by far the most prevalent, are often difficult to image because of their deep location. Paraumbilical veins are easier to image because they are superficial. They arise from the ventral tip of the left portal vein and usually flow caudally through the ligamentum teres, where they communicate with superficial peritoneal collaterals. Other types of portal collaterals occur, including retroperitoneal, splenorenal, splenoretroperitoneal, short gastric, and omental.

Therapeutic Portosystemic Shunts Color Doppler ultrasonography often displays surgical shunts, [23] even when they are inapparent on preliminary gray-scale images. Transjugular intrahepatic portosystemic shunt stents can be evaluated by spectral and color-flow Doppler before, after, and sometimes during the procedure. Shunt thrombosis can be reliably diagnosed only when no anastomotic flow is seen with color Doppler ultrasonography. Only direct visualization of the shunt itself should be used to determine patency or thrombosis. Secondary signs of shunt thrombosis, such as hepatopedal intrahepatic portal flow, [23] may be misleading. [24]

Venous Thrombosis Ultrasonographically, intralumenal echoes suggest clot, but they are not diagnostic. When hypoechoic or anechoic, clot may be virtually invisible without color Doppler. Anechoic thrombus is rare in neoplastic invasion but is fairly common with bland clot. Color Doppler ultrasonography highlights clot by displaying flow around the clot and in adjacent patent vessels. [20] Portal venous thrombosis can result from neoplastic invasion, septic portal thrombosis (pylephlebitis), and pancreatitis. We believe that thrombosis in portal hypertensive patients [20], [25] is considerably more common than the 1% previously reported. [26] Color Doppler ultrasonography is unrivaled in diagnosing partially occluded vessels. Small residual flow channels are automatically displayed in color. Clot resolution or progression can be documented on serial examinations. Contrast agents may be useful in difficult cases. Cavernous transformation of the portal vein, resulting from main portal vein clot, consists of prominent hepatopedal portal collaterals. In acute portal vein thrombosis, color Doppler ultrasonography may reveal small acute hepatopedal portal collaterals, invisible on gray-scale ultrasonography. This may be a precursor to cavernous transformation of the portal vein.

Budd-Chiari Syndrome Ultrasonography, especially with color Doppler imaging, is the preferred screening technique for evaluating patients with suspected Budd-Chiari syndrome. [27] Gray-scale findings include hepatic vein abnormalities: absence, stenosis, dilation, irregularity, and abnormal or absent junction with the IVC. The IVC may itself be occluded or stenosed. Less specific changes include caudate hypertrophy, atrophy and enlargement of the other lobes and segments, and parenchymal inhomogeneity. Absent or reversed hepatic venous flow and intrahepatic hepatic vein–to–hepatic vein "spider web" collaterals are diagnostic of Budd-Chiari syndrome. A flat hepatic vein waveform, lacking the normal phasic fluctuation, indicates hepatic vein compression–a finding that, although nonspecific, supports the diagnosis of Budd-Chiari syndrome. Other collaterals seen in Budd-Chiari syndrome include hepatic vein–to–portal vein collaterals with portosystemic shunting (e.g., through enlarged paraumbilical veins, esophageal varices) and hepatic veins to systemic veins.

Other Vascular Diseases Ultrasonographic findings in hepatic venoocclusive disease are nonspecific changes related to fibrous obliteration of microscopic central hepatic lobular venules. These findings, not universally present, include thickened gallbladder wall, abnormal or reversed portal venous flow, [28] and increased hepatic artery peripheral resistance. [29] Hereditary hemorrhagic telangiectasia (Osler-Weber-Rendu disease), characterized by telangiectasias, arteriovenous (AV) fistulas, and aneurysms, involves the liver in about one third of patients. Gray-scale findings include enlarged hepatic arteries and increased echogenicity from fatty infiltration or cirrhosis. [30], [31] Color Doppler ultrasonography reveals enlarged, tortuous, arteries with high-velocity flow. Color Doppler ultrasonography clarifies the potentially confusing gray-scale appearance that may mimic biliary dilation. Visualization of hepatic AV fistulas is variable and probably depends on their size. Color Doppler ultrasonography can be used to diagnose and follow hepatic pseudoaneurysms and AV fistulas, to assess the results and adequacy of vascular interventional procedures without recourse to angiography, and to demonstrate portal and hepatic vein malformations. Thrombosis or occlusion of the hepatic artery are the most common causes of hepatic infarction. [32] Ultrasonographically, infarcts are usually peripherally located, hypoechoic, wedge-shaped lesions. Others are round and centrally located. In the acute phase, the infarcts are ill-defined, hypoechoic lesions that evolve into more anechoic or cystic areas. [33] After hepatic infarction, cystic collections of bile may develop (bile lakes).

Focal Liver Disease

All standard noninvasive imaging modalities are insensitive detectors of focal hepatic disease, with individual lesion detection rates ranging from 38% to 66% for MRI and CT. [34], [35] Focal hepatic lesions are frequently missed with one modality, then detected with another. Although ultrasonography is less sensitive than CT and MRI, modern ultrasonography can perform well in detecting focal liver disease. [36], [37] and [38] Arterial and portal venous phase imaging with ultrasound contrast agents can

increase both the detection and characterization of lesions in patients with cirrhosis or liver metastasis. [39], [40] Ultrasound contrast enhancement may prove useful in differentiating benign from malignant tumors [2], [3], [41], [42] and [43] (Fig. 153-4).

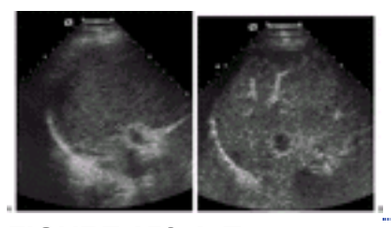

FIGURE 153-4. Transverse ultrasound image through the medial segment of the left lobe in a patient with prior right hepatectomy for metastatic disease from adenocarcinoma of unknown primary. **A:** Precontrast gray-scale image shows a mass that is difficult to see (*cursors*). **B:** Postcontrast gray-scale image shows a small hypoechoic metastasis (*cursors*). Note that the surrounding liver is hyperechoic, consistent with increased vascularity with respect to the metastasis.

The main strengths of ultrasonography are its ability to guide biopsy and to characterize common benign lesions (cysts, hemangiomas), its safety, and its low cost. Its weaknesses include its inability to image the entire liver in some patients and its inferiority to CT in detecting extrahepatic disease. Ultrasound-guided biopsy is more efficient and cost-effective, even when lesions are initially detected with some other modality. Ultrasonography may be used to evaluate resectability of primary or metastatic liver tumors. Color Doppler ultrasonography improves detection of focal lesions, especially anechoic or hypoechoic lesions. Some tumors are more conspicuous because of increased flow compared with normal liver: focal nodular hyperplasia (FNH), most HCCs, some metastases, and occasionally other masses. [44], [45] and [46] Color and spectral Doppler can be used to help discriminate among benign and various types of malignant liver tumors. [47], [48], [49] and [50]

Simple Cysts The prevalence of simple cysts increases with age. [51] When a cyst is found in a young patient, other diseases should be considered, including autosomal dominant polycystic disease, von Hippel–Lindau disease, and conditions simulating simple cysts, such as echinococcal disease, residua from trauma or infection, and cystic neoplasia. Ultrasonography is usually superior to CT and MRI for evaluating cysts. Simple cysts are usually well-defined, echo-free, round or oval lesions with imperceptible walls and good through-transmission (Fig. 153-5). Thin septations are frequent and should not suggest a different diagnosis. Thicker septations are generally a sign of a complicated cyst or of a cystic neoplasm.

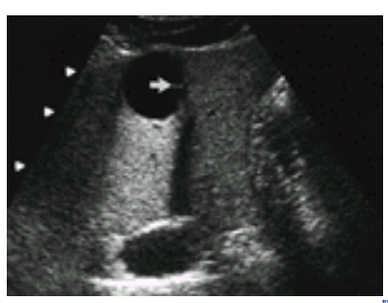

FIGURE 153-5. Simple liver cyst. This 2.5-cm cyst is anechoic, has imperceptible walls, and has prominent through-transmission (*(whiter area,* deep to the cyst). There is a thin septation within the cyst (*arrow*). Thin (but not thick) septations are perfectly compatible with diagnosis of a classic simple cyst.

Hepatic Polycystic Disease Ultrasonographic findings include multiple, often contiguous simple cysts. Irregular shape and septae are common. Hemorrhage or infection may cause debris-like echoes within one or more cysts. [52] About one half of patients have renal cysts. Von Meyenburg complex is a rare disorder characterized by multiple small cystic hamartomas of the bile ducts that may be detected with CT or ultrasonography.

Other Cystic Lesions Acquired cysts may occur as a result of trauma or infection. [53], [54] and [55] Neoplasms that simulate simple cysts are unusual; metastases from squamous cell carcinoma, GI stromal tumors, and cystic adenocarcinomas occasionally do this. Hepatobiliary cystadenoma is a rare neoplasm probably arising from bile duct epithelium. These lesions are predominantly multicystic with varying amounts of solid tissue. [56], [57] and [58] The liver is the most frequently involved organ in hydatid (echinococcal) disease. A spectrum of ultrasonographic findings from purely cystic to solid-appearing pseudotumors occurs. Internally, wavy bands of delaminated endocyst (the “water lily” sign) may be noted. [59] Daughter cysts, sometimes surrounded by echogenic debris (“matrix”) are frequent. Calcifications, varying from tiny to massive, are often present. A densely calcified cyst usually indicates a dead, inactive lesion. Previously considered contraindicated, percutaneous drainage and treatment are gaining wider acceptance.

Pyogenic Liver Abscess When liver abscess is suspected clinically, ultrasonography is the preferred screening modality. Ultrasonographically, pyogenic liver abscesses have a variable appearance (Fig. 153-6). Typical features include irregular margins and a primarily hypoechoic mass. Irregular areas of increased echogenicity are frequent. On occasion, a diffusely hyperechoic appearance may be noted, owing to microbubbles from gas-forming organisms. Diffuse microabscesses, often associated with biliary obstruction, may cause a confusing ultrasonographic pattern of increased irregular hepatic echogenicity. Clusters of small low-echogenicity lesions may also be noted. [60] Septic portal venous thrombosis (pylephlebitis), which may be imaged ultrasonographically, may lead to liver abscess. [61]

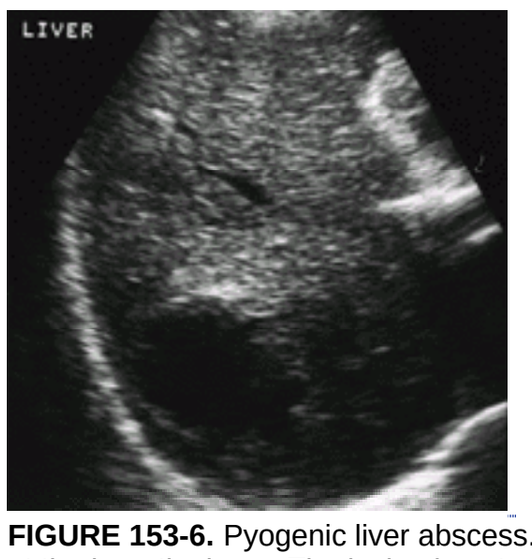

FIGURE 153-6. Pyogenic liver abscess. This transverse sonogram of the right lobe of the liver shows a mixed echogenicity, predominantly hypoechoic liver abscess at the hepatic dome. The lesion is not well defined medially, a feature of pyogenic liver abscess on ultrasonography.

Amebic Liver Abscess Amebic liver abscesses tend to have a round or oval shape and hypoechoic appearance with fine, homogeneous, low-level echoes throughout. Image findings alone are rarely sufficient to distinguish amebic from pyogenic liver abscesses. [62] Amebic liver abscess can have ultrasonographic patterns that are bizarre, including diffuse increased echogenicity, debris levels, and prominent heterogeneity. When it is unclear clinically if an abscess is amebic or pyogenic, percutaneous diagnostic aspiration should be performed (necessary in 15% of patients in our institution). In contrast to pyogenic liver abscesses, percutaneous drainage of amebic liver abscess is rarely indicated. [63] Therapeutic aspiration provides no objective benefit when compared with the use of oral amebicidals alone. [64]

Candidiasis Candidiasis, occurring in leukemic and other immunocompromised patients, causes multifocal hypoechoic lesions typically smaller than 2 cm. Four distinct ultrasonographic patterns of hepatic microabscesses from candidiasis have been described by Pastakia and associates. [65] These include discrete hypoechoic nodule, a bull’s-eye configuration, a discrete echogenic focus with variable degrees of acoustic enhancement, and a wheel-within-a-wheel appearance. Internal echogenic foci produce the target or wheel-within-a-wheel appearance. [66], [67] Echogenic lesions and lesions larger than 2 cm are much less common.

Benign Neoplasms

Cavernous Hemangioma Cavernous hemangioma, the most common benign hepatic neoplasm (prevalence, 1% to 4%), rarely causes clinical symptoms. Although ultrasonographic findings are often classic, hemangiomas must often be evaluated further to exclude more significant lesions, especially in patients with malignancy. Small hemangiomas are usually well-defined, echogenic lesions with enhanced through-transmission [68] (Fig. 153-7). Larger hemangiomas frequently diverge from this pattern. Mixed-echogenicity lesions and even hypoechoic hemangiomas may occur.

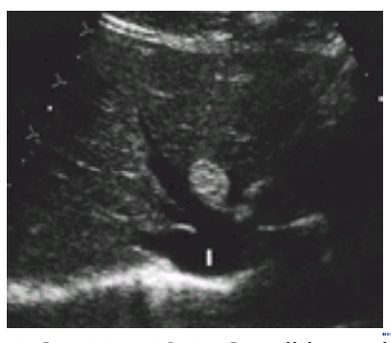

FIGURE 153-7. Small hepatic hemangioma. This transverse sonogram at the level where the hepatic veins enter the inferior vena cava (*I*) reveals a well-defined, white, echogenic focus that is virtually diagnostic for hepatic hemangioma.

Typical small hemangiomas in a patient without evidence of malignancy should be followed by ultrasound in 3 to 4 months. Hemangiomas rarely change on serial exams. [69] Lesions in patients with malignancy should be evaluated further with T2-weighted MRI or 99mTc-tagged red cell blood pool single photon emission computed tomography scanning. Preliminary results suggest that addition of ultrasound contrast agents may be sufficient to diagnose incidentally detected hemangiomas at ultrasound confidently [3], [70] (Fig. 153-8). CT and MRI may fail to identify small (less than 1 cm) hemangiomas detected with ultrasound. If needed, thin-needle biopsy is a reasonable and safe way to evaluate hemangiomas, [71], [72] provided skilled cytologic consultation is available.

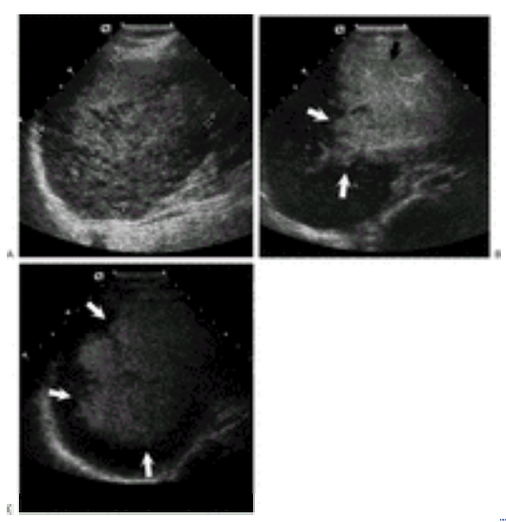

FIGURE 153-8. Atypical hemangioma (stable for 11 years): transverse gray-scale ultrasound image through the right lobe. **A**: Precontrast shows large ill-defined heterogenous mass (*cursors*). **B**: Postcontrast, normal liver (*black arrow*) and the medial wall of the hemangioma (*white arrow*) enhance. **C**: At 90 seconds postcontrast administration, a larger volume of tissue with delayed enhancement is seen (*arrows*).

Focal Nodular Hyperplasia and Liver Cell Adenoma FNH is a clinically insignificant lesion, whereas liver cell adenoma (LCA) can cause morbidity and mortality because of hemorrhage and, occasionally, malignant degeneration. The ultrasonographic features of FNH and LCA are variable. There is a tendency for FNH to be more homogeneous than LCA. FNH often has mildly increased echogenicity compared with normal parenchyma, whereas LCAs are usually hypoechoic and more inhomogeneous. [73], [74], [75], [76] and [77] On color-flow imaging, FNH usually has markedly increased flow, sometimes with vessels radiating peripherally from a central feeding artery. [42]

Malignant Neoplasms

Hepatocellular Carcinoma Advanced HCC is almost always multifocal, making it difficult to distinguish from metastatic disease (Fig. 153-9). Small HCCs (less than 5 cm) are often (75%) hypoechoic. [78] As HCCs grow, they tend to develop hypoechoic peripheral rims. [79] With further progression, lesions become more numerous and heterogeneous. Fatty metamorphosis may cause increased echogenicity and confusion with hemangioma. [80] Hepatic or portal venous invasion should suggest the diagnosis of HCC, although other liver tumors may invade veins. [81] Rarely, HCC invades the bile duct. About three fourths of patients with HCC have identifiable internal color flow, compared with one third of those with metastases. Most HCCs show very abnormal corkscrew tumor vessels both at the center and at the periphery with ultrasound contrast agents, a pattern distinct from benign tumors. [3] Fibrolamellar HCC, which accounts for 2% of HCCs but 25% to 50% of HCCs in young adults, is typically a single well-circumscribed lesion in an otherwise normal liver. Other features include a “central scar” and calcification, both of which also occur in other subtypes of HCC and lesions of other histology. [77]

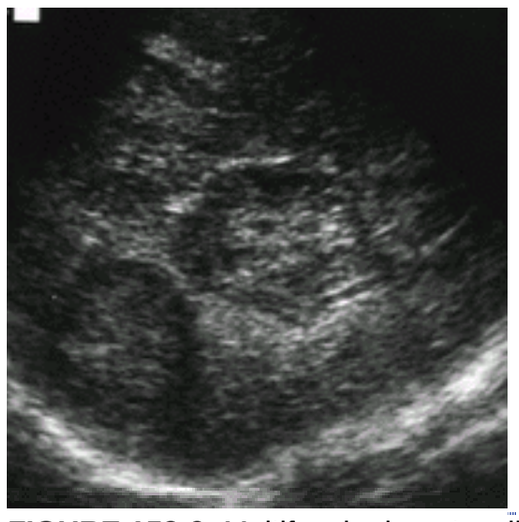

FIGURE 153-9. Multifocular hepatocellular cancer. A transverse sonogram shows multiple tumor nodules in the right lobe of the liver. Some of these are hypoechoic, and others have echogenic centers with hypoechoic rims.

Hepatic Metastasis Metastatic disease, the most common liver malignancy in North America, is multifocal in 90% of patients. Virtually any ultrasonographic appearance may occur. Metastatic lesions, whether diffuse or focal, are usually heterogeneous. Hypoechoic halos or target or bull’s-eye patterns with rings of varying echogenicity are common. Ill-defined infiltration with focal nodularity is another frequent pattern. Metastases that simulate simple cysts or classic hemangiomas are uncommon. Predominantly fluid-filled, presumably necrotic metastases occur most frequently with squamous cell carcinoma, sarcomas, and ovarian and testicular carcinoma. Calcified lesions, especially mucinous adenocarcinoma, may occur. Although ultrasonographic appearance is a poor predictor of the primary tumor, certain patterns are suggestive. Large to moderate-sized hyperechoic metastases should suggest the possibility of a colonic primary. Lesions with fluid-fluid levels

(representing intralesional necrosis and hemorrhage) are often found with metastatic GI stromal tumors. Distinction between metastasis and HCC occasionally requires ultrasonographically guided biopsy. Invasion of the portal or hepatic veins suggests HCC rather than metastasis. Even though portal venous invasion in metastatic disease has been reported in as many as 8% of patients, [82] our experience suggests a lower prevalence.

Human Immunodeficiency Virus Infection The liver is often involved by neoplasm (non-Hodgkin lymphoma, Kaposi sarcoma) or by opportunistic infection in patients with human immunodeficiency virus (HIV) infection. Ultrasonographic findings of uncertain cause, such as decreased periportal echogenicity or focal hyperechoic areas, are frequent. High- and intermediate-grade HIV-related B-cell lymphomas cause focal liver lesions more frequently than other lymphomas. These are usually hypoechoic masses that may exhibit increased peripheral color flow. Kaposi sarcoma usually infiltrates the liver, causing little or no ultrasonographic abnormality. Kaposi sarcoma may cause focal masses and increased periportal echogenicity. [83] Multiple echogenic foci, often imaged more clearly ultrasonographically than with CT, may give a starry-sky pattern with disseminated *Pneumocystis carinii* infection. [84] This pattern also occurs with mycobacterial or cytomegalovirus infection. [85] On occasion, disseminated *Pneumocystis* infection results in larger calcifications or large, hypoechoic lesions.

Other Focal Lesions Hepatic lymphoma and leukemia are usually microscopic. Only about 5% cause ultrasonographically detectable focal liver lesions. [86] The lesions are usually anechoic or hypoechoic, although other patterns occur. [87] Chloroma of the liver, associated with myelogenous leukemia, can be hypoechoic or echogenic. HIV-related lymphomas cause more frequent and conspicuous focal lesions than other lymphomas. Hepatic sarcomas are rare. Angiosarcomas and undifferentiated embryonal sarcomas are usually large, internally inhomogeneous masses. [88] Hepatic epithelioid hemangioendothelioma, a rare vascular sarcoma, [89] starts as a multinodular tumor, coalescing as it grows. It is usually hypoechoic compared with normal liver. Hypertrophy of the uninvolved normal liver may occur. Adenomatous hyperplastic nodules in cirrhotic livers (regenerating nodules) are premalignant lesions that, when visualized, are usually hypoechoic and may have a thin echogenic rim. [90] Intrahepatic extramedullary hematopoiesis may cause inhomogeneous, mainly hypoechoic, masses. [91] Mesenchymal hamartoma of the liver is usually a large, mainly cystic mass with septae of variable thickness and solid internal tissue. [92] Intrahepatic hematomas may cause ultrasonographically visible masses.

Trauma

CT is the mainstay for evaluating hemodynamically stable patients with suspected blunt hepatic trauma. Ultrasonography can be used effectively to detect hemoperitoneum, but it has a rather limited role in the initial diagnostic evaluation of hepatic trauma. A high percentage of liver and other parenchymal injuries are missed ultrasonographically. [93] Ultrasonography may be valuable in the follow-up of nonoperated patients and in diagnosing posttraumatic complications, including biloma, vascular pseudoaneurysms and AV fistulas, and abscesses.

Liver Transplantation

Ultrasonography, especially Doppler imaging, is important in evaluating liver transplantation candidates and potential donors. [94] Ultrasonography can be useful in defining portal hypertension–related collaterals, detecting portal vein clot, delineating spontaneous or surgical portosystemic shunts, and detecting HCC in the native liver.

After transplantation, ultrasonography can screen for hepatic arterial occlusion. [95] Absent flow on color Doppler and decreased peripheral resistance, measured by resistive index, [96] are signs of arterial compromise. The use of ultrasound contrast agents may improve the accuracy and speed of examination in transplant recipients. When abnormalities are noted, arteriographic evaluation and intervention are often indicated. [97] Hypoechoic infarcts or intrahepatic fluid collections may result when arterial occlusion occurs. Cholangiography is generally needed to detect bile duct injuries reliably because ultrasonography may miss 50% of bile duct complications. [98] Posttransplantation fluid collections include abscess, biloma, seroma, lymphocele, and hematoma. [99]

Overview of Hepatic Imaging

The main strengths of hepatic ultrasonography are its ability to characterize common benign lesions (cysts, hemangiomas) and to guide percutaneous procedures, its safety, its excellent patient tolerance, and its low cost. In the near future, the application of ultrasound contrast agents to the liver may extend the utility of ultrasound from a screening examination to a lesion-detection and diagnosis modality. Its weaknesses include inability to image the entire liver in some patients and inferiority to CT in detecting extrahepatic disease. When liver abscess is suspected clinically, ultrasonography. The ability of ultrasonography to image in any oblique plane often makes it superior to CT and MRI in localizing lesions to an anatomic hepatic segment when evaluating resectability of a liver tumor.

In experienced hands, ultrasonographically guided liver biopsy is often quicker and easier than CT guidance. Ultrasonography can directly visualize the needle tip as it is placed in the lesion, facilitating biopsy of small lesions and lesions in uncooperative patients. Ultrasound-guided biopsy is more efficient and cost-effective, even when lesions are initially detected with some other modality. Despite the advantages of ultrasonography, CT-guided biopsy is often more popular with radiologists because it almost always shows needle location. Ultrasonographic needle visualization may be difficult or impossible when the liver is echogenic or when acoustic access is imperfect. Newer ultrasonographic techniques that enhance needle-tip visualization and improved biopsy guides promise to make ultrasonographically guided biopsies easier.

Ultrasound contrast enhancement also may be useful in delineating residual tumor after ablation. Viable tumor can be identified and targeted for retreatment with ultrasound contrast during or after ethanol [100] or radiofrequency ablation. Interestingly, US failed to demonstrate flow in residual tumor before contrast enhancement, an indication of the potential usefulness of ultrasound contrast agents in these patients.

GALLBLADDER

Indications for Ultrasonography

Ultrasonography is the imaging method of choice for the initial evaluation of all suspected diseases of the gallbladder. It is particularly valuable in patients with acute right upper quadrant pain and possible acute cholecystitis. [101], [102] Ultrasonography is highly reliable in detecting tiny gallstones and is useful for evaluating focal or diffuse abnormalities of the gallbladder wall. [103] Biliary ultrasonography has several distinct advantages compared with scintigraphy and CT. It is less expensive and can be performed rapidly without patient preparation or contrast agents. Unlike biliary scintigraphy, ultrasonography is not organ specific and may provide important diagnostic information regarding the liver, pancreas, and peritoneal cavity. Ultrasonography can readily be performed in patients with abnormal liver function that may preclude scintigraphy. Finally, ultrasonography may be used to guide percutaneous cholecystostomy in critically ill patients at the bedside.

Patient Preparation and Technique

In patients undergoing ultrasonography for acute right upper quadrant pain and possible acute cholecystitis, no prior preparation is necessary, and patients can be imaged directly from the emergency room. Patients undergoing elective ultrasonography to evaluate the gallbladder should be fasting for a minimum of 6 hours. This will ensure adequate distention of the gallbladder to visualize small stones and facilitate imaging of the gallbladder wall.

Normal Anatomy

Except in the rare instance of agenesis, the normal gallbladder can be visualized in virtually all fasting patients [104], [105] as a pear-shaped, fluid-filled structure along the inferomedial aspect of the right lobe of the liver. The gallbladder is typically less than 12 cm in length and is up to 3 cm in anteroposterior diameter. The normal gallbladder wall measures less than 3 mm. In addition to agenesis, other rare anomalies of the gallbladder that may be diagnosed with ultrasonography include duplication and septations between the fundus and body (Phrygian cap). [106]

Gallstone Disease

Stones as small as 3 to 5 mm can be easily visualized ultrasonographically. The characteristic ultrasonographic appearance is an intralumenal echogenic focus casting a distal acoustic shadow (Fig. 153-10). It may be difficult to document acoustic shadowing in all patients with small gallstones. By varying the patient's position, mobility of the gallstones may be demonstrated in a high percentage of patients. Demonstrating mobility helps to differentiate small stones from polyps

arising from the gallbladder wall that will not change in position.

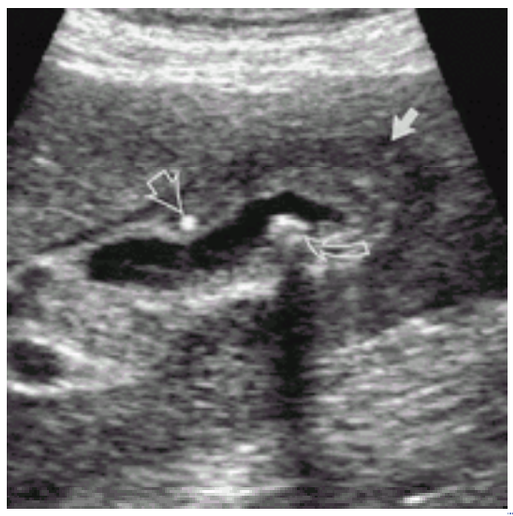

FIGURE 153-10. Gallstones in a patient with adenomyomatosis. Note the intralumenal gallstone *(curved open arrow).* There is an intramural gallstone (*straight open arrow)* within a diffusely thickened gallbladder wall (*arrow*) as a result of adenomyomatosis.

Although ultrasonography is accurate for detecting stones within the gallbladder, once stones migrate into the cystic duct, they are often very difficult to visualize. [107] It is important to perform parasagittal images of the neck, gallbladder, and cystic duct region in patients with right upper quadrant pain. The identification of impacted cystic duct stones strongly suggests cystic duct obstruction.

Echogenic abnormalities within or adjacent to the gallbladder at times may ultrasonographically mimic gallstones. These include polyps, tumefactive sludge, surgical clips, and air bubbles. Gallbladder polyps are echogenic mucosal lesions that do not shadow. Sludge within the gallbladder is never a normal finding. It generally indicates lack of gallbladder emptying with precipitation of cholesterol or calcium bilirubinate crystals. At times, sludge crystals may adhere together as “tumefactive” sludge and mimic a gallstone or mucosal mass. Gas within the gallbladder may be due to surgically created biliary-enteric anastomoses, endoscopic biliary procedures, or gas-forming infection from emphysematous cholecystitis.

Acute Cholecystitis

The most important ultrasonographic findings to confirm the diagnosis of acute cholecystitis are gallstones, gallbladder wall thickening, and a positive ultrasonographic Murphy sign. [101], [102] The ultrasonographic Murphy sign differs significantly from the clinical sign. It refers to focal, unequivocal tenderness directly over the visualized gallbladder when pressure is applied by the transducer. In a prospective study of 497 patients with acute right upper quadrant pain, the combination of gallstones and a positive Murphy sign had a 92% positive predictive value for acute cholecystitis. [101] Gallbladder wall thickening (greater than 3 mm) and pericholecystic fluid in the appropriate clinical setting are secondary findings of acute cholecystitis, but both nonspecific findings may be due to a variety of other disorders. Biliary scintigraphy should be performed whenever the ultrasonographic findings are equivocal.

In a small percentage of patients (5% to 10%), acute cholecystitis develops in the absence of gallstones or cystic duct obstruction. [108] Conditions implicated in acalculous cholecystitis include trauma, major surgical procedures, hyperalimentation, burns, sepsis, and other severe intercurrent illnesses. The ultrasonographic findings in early acalculous cholecystitis are often subtle and nonspecific. In a small percentage of patients, the gallbladder may be normal early in the clinical course. [109] Generalized symmetric thickening of the gallbladder wall is frequently not due to cholecystitis and must be interpreted with caution. It may be due to such diverse conditions as hepatitis, fluid overload, portal hypertension, or hypoalbuminemia. In the appropriate clinical setting, gallbladder wall thickening with a positive ultrasonographic Murphy sign is suggestive of acute acalculous cholecystitis. The sensitivity of ultrasonography for the diagnosis of acalculous cholecystitis, however, is substantially less than with calculous cholecystitis, in the range of 63% to 67%. [110] Interval follow-up studies performed at 24 to 48 hours may be valuable in demonstrating progressive gallbladder wall edema or the development of fluid.

In patients without acute right upper quadrant pain, stones within a contracted gallbladder often indicate chronic cholecystitis. Shrinking of the gallbladder wall is a common associated finding. Marked contraction of the gallbladder around a large stone results in the wall-echo shadow sign, and there is prominent acoustic shadowing from the gallbladder fossa.

There are several ultrasonographic features that, in the appropriate clinical setting, may suggest gangrenous cholecystitis. These include pericholecystic fluid, intralumenal membranes (due to sloughed mucosa or fibrinous debris), and asymmetric thickening of the gallbladder wall [110] (due to intramural microabscesses or hemorrhage). A high percentage of patients with gangrenous cholecystitis do not have a positive ultrasonographic Murphy sign. [111] In critically ill patients with suspected gangrenous cholecystitis, ultrasonography may be useful to guide percutaneous cholecystostomy even at the bedside.

Perforation of the gallbladder with pericholecystic abscess is a major cause of sepsis and morbidity from acute cholecystitis. [112] Pericholecystic abscesses appear ultrasonographically as complex fluid collections in direct contiguity with the gallbladder. Mural necrosis may be identified ultrasonographically as a focal loss of the normal reflectivity of the gallbladder wall. Inflamed omental and pericholecystic fat has been noted adjacent to pericholecystic abscesses. These fatty structures “wall-off” the abscesses from the greater peritoneal cavity. Hyperemia of the inflamed omental fat may be demonstrated with color Doppler imaging. [113]

Gallbladder Tumors

Most benign neoplasms of the gallbladder originate from the mucosal surface of the gallbladder and are either adenomatous or cholesterol polyps [114], [115] (Fig. 153-11). Small polyps are often incidental findings that, when less than 5 mm in diameter, are of no cause for clinical concern. Enlarging gallbladder polyps on serial sonograms or polyps greater than 1 cm in diameter are usually an indication for cholecystectomy owing to the slight increased risk for carcinoma.

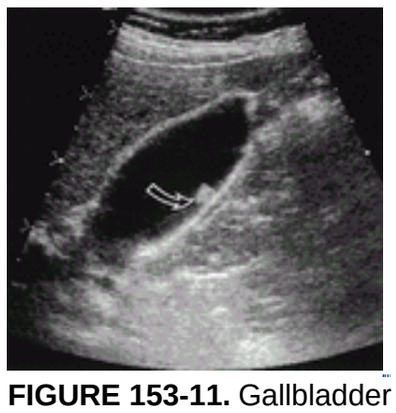

FIGURE 153-11. Gallbladder polyp. Note the echogenic mucosal mass without an acoustic shadow (*arrow*).

The ultrasonographic findings of gallbladder carcinoma are variable and include a polypoid mucosal lesion or focal or irregular thickening of the gallbladder wall, sometimes associated with an extramural mass. [116] In advanced cases of gallbladder carcinoma, patients may present with a large mass arising from the gallbladder fossa. [116] Direct hepatic invasion and nodal metastases may also be demonstrated ultrasonographically. However, in many instances, CT is required to delineate more precisely the extent of tumor and to detect distant spread of the disease.

Other Conditions

Adenomyomatosis and cholesterolosis are benign disorders of the gallbladder that are classified as hyperplastic cholecystoses. [114], [115] Cholesterolosis typically results in either single or multiple small mucosal polyps. It is due to the subepithelial accumulation of cholesterol-laden histiocytes that ultimately extend to the mucosal surface. Adenomyomatosis causes either focal or diffuse hypertrophy of the normal Rokitansky-Aschoff sinuses and smooth muscle within the gallbladder wall. Crystals and debris may be trapped within the sinuses of the hypertrophied wall and produce echogenic foci with echogenic comet-tail artifacts.

BILE DUCTS

Indications for Ultrasonography

The ability of ultrasonography to detect dilated bile ducts and the level of biliary obstruction makes it the technique of choice for evaluating jaundiced patients. Ultrasonography can also detect the cause of obstruction, albeit with less accuracy. Infectious cholangitis and conditions such as HIV cholangiopathy are indications for ultrasonography. Ultrasonography can detect and assess, with the help of color Doppler, resectability of cholangiocarcinoma and other tumors affecting the bile ducts.

Patient Preparation and Technique

Biliary ultrasonography is best performed after an overnight fast or 8 to 12 hours of a clear liquid diet. Because distention of the gallbladder is desirable, fatty foods should be avoided. The biliary tree is best imaged with the patient in the supine and right anterior oblique positions, starting with 3- to 7-MHz curved linear array transducers. A subcostal acoustic window should be used first, supplemented with intercostal scans. Sector or other small-footprint transducers are often useful to image the extrahepatic bile duct and the cystic duct of the gallbladder. Transverse compression views of the pancreatic head region with large-footprint curved linear transducers are useful to visualize the intrapancreatic bile duct, where about 90% of symptomatic common duct stones are located. [117] Color-flow imaging is useful to differentiate the bile ducts from vessels and to detect hyperemia in and near the gallbladder.

Normal Anatomy

Ultrasonography routinely displays the normal intrahepatic and extrahepatic ducts. The normal extrahepatic bile duct (common duct) runs ventral to the main portal vein (Fig. 153-12) and to the left of the proper hepatic artery. The internal diameter of the normal extrahepatic bile duct is 5 mm or less, although elderly patients and patients with cholelithiasis may have 6- to 9-mm internal-diameter ducts. The distal common duct is usually seen within the pancreatic head and has a somewhat smaller caliber. The intrahepatic bile ducts may be seen ventral to the left and right portal veins. The normal internal diameter of the main left and right intrahepatic bile ducts is about 1 mm.

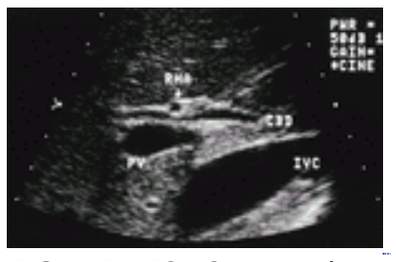

FIGURE 153-12. Normal common bile duct. This long-axis view of the normal common bile duct *(CBD)* shows the relationship of the bile duct to the portal vein (*PV*). Note that the right hepatic artery *(RHA)* passes ventrally (above) the bile duct. This occurs in 10% of patients. In 90% of patients, the right hepatic artery passes between the portal vein and the bile duct. The common bile duct enters the pancreatic parenchyma. IVC, inferior vena cava.

Bile Duct Obstruction

Stones are the most common cause of distal bile duct obstruction. Tumors are a more frequent cause of proximal bile duct obstruction. Ultrasonographically, dilated intrahepatic bile ducts are tubular anechoic structures that demonstrate posterior acoustic enhancement. Centrally, in the porta hepatis, dilated ducts are usually ventral to the portal veins. Peripheral ducts have a variable relationship to the portal veins. The “too many tubes” sign is the classic pattern indicative of intrahepatic biliary dilation. Color Doppler is useful to distinguish veins and enlarged arteries from dilated bile ducts, simplifying detection of abnormal ducts and preventing misidentification of vessels as bile ducts.

Surprisingly, normal bile duct size is controversial. Obstruction is rarely present when the extrahepatic bile duct is 5 mm or less in maximal internal diameter. Nondilated obstruction may be seen in cholangiocarcinoma or sclerosing cholangitis. Bile ducts with maximal internal diameters of 10 mm or more are almost always obstructed. In most patients, 6- to 9-mm ducts (“gray-zone”–sized ducts) are not obstructed, but rather are that size because of breakdown of the elastic fibers within the duct wall. Patients older than 50 years of age are given 1 mm per decade in duct diameter. For example, a 7-mm duct should be considered normal in a 70-year-old patient. Patients with cholelithiasis often have 6- to 9-mm-diameter ducts. We postulate that this is caused by degeneration of elastic tissue within the duct wall from transient obstruction caused by recurrent passage of stones. This is analogous to the increased residual diameter of a deflated balloon that has been repeatedly blown up and deflated. Rarely, an unobstructed duct 10 mm or larger may be found. Administration of a fatty meal may aid in evaluating gray-zone–sized ducts. A decrease of 2 mm in duct size after a fatty meal indicates normal bile dynamics and excludes obstruction. If the extrahepatic duct enlarges after the fatty meal, obstruction is likely.

Distal bile duct obstruction causes extrahepatic bile duct dilation and often intrahepatic bile duct dilation. Intrahepatic duct dilation is almost always a sign of obstruction, although rare exceptions are seen. Thus, intrahepatic dilation is specific for obstruction, but it is less sensitive than extrahepatic bile duct dilation. Obstruction at sites other than the distal duct results in proximal dilation, with a normal-caliber distal duct.

Choledocholithiasis Choledocholithiasis occurs in at least 15% of patients with cholelithiasis. Conversely, 95% of patients with common duct stones also have gallstones. [118] The prevalence of choledocholithiasis in patients with gallstones increases with age. [119] A shadowing echogenic focus in the bile duct is virtually diagnostic of cholelithiasis (Fig. 153-13). Shadowing may be difficult to demonstrate in some impacted stones. Potential pitfalls include the Mirizzi syndrome, air in the bile duct, extrabiliary calcifications, or surgical clips. On occasion, a completely stone-filled duct may be confused with a gas-containing segment of bowel. Extrahepatic biliary dilation is present in 70% to 94% of patients with choledocholithiasis. [120], [121] Although stones in normal-sized ducts can be identified, these are probably harder to detect than stones in dilated ducts.

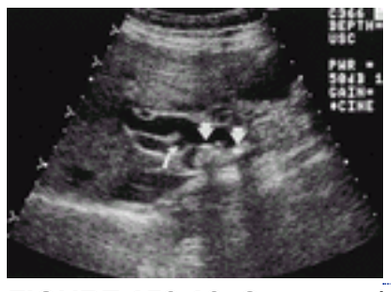

FIGURE 153-13. Common duct stone. This long-axis view of the intrahepatic and extrahepatic bile ducts reveals two echogenic common duct stones (*arrows*). Note the cystic duct (*curved arrow*) entering the common duct dorsally.

Bile duct stones are harder to detect than gallstones, with optimal reported sensitivity of 75% to 80%. [122] The difficulty relates to gas sometimes obscuring the distal duct and the lack of echo-free bile around impacted stones. About 90% of proximal, suprapancreatic duct stones (about 10% of cases) are detected. [123] Meticulous technique is necessary to achieve optimal sensitivity. A prospective Finnish trial reported detection rates of only 22% for ultrasonography and 25% for CT in patients with choledocholithiasis. [123]

Infectious Cholangitis

Contrast CT is probably the best screening examination in patients with severe cholangitis. [124] Ultrasonography is the study of choice in patients with clinically less severe cholangitis. Diffuse bile duct wall thickening is often present. [125] Debris in the bile duct suggests the presence of pus and, hence, infectious cholangitis. Liver abscesses can be identified.

Recurrent Pyogenic Cholangitis Recurrent pyogenic cholangitis (also known as Oriental cholangiohepatitis) is characterized by recurrent bacterial cholangitis promoted by strictures and primary bile duct stones that may occur in any part of the biliary tree. Gallstones and liver abscesses are frequent. [126] Ultrasonography is the preferred initial imaging modality. [127], [128] Most patients have huge stones with dramatic shadowing that can obscure the biliary tree. The soft, mudlike stones are often of medium echogenicity, however, and may lack acoustic shadowing. This appearance can mimic sludge, pus, blood, or even neoplasm within the biliary tree. Markedly dilated intrahepatic and extrahepatic ducts are the rule. It is not unusual for duct size to exceed 2 cm. [129] Ultrasonography is limited in patients with prior biliary enteric anastomosis because intraductal gas obscures both the ducts and intrahepatic calculi. Gas may also make detection of liver abscesses difficult.
Human Immunodeficiency Virus–related Cholangiopathy HIV-related cholangiopathy is a diffuse biliary tract disease affecting the gallbladder and the bile ducts. More than 80% of patients have alkaline phosphatase levels greater than twice normal, but bilirubin is usually normal or minimally elevated. [129], [130] and [131] Ultrasonography, although less sensitive than endoscopic retrograde cholangiopancreatography, is useful in evaluating patients suspected of having HIV-related cholangiopathy. [131] Smooth or irregular thickening of the bile duct wall (45%) and biliary dilation (about 70%) are frequent abnormalities. [132] Biliary dilation often occurs in association with ampullary stenosis. An echogenic nodule at the end of the common duct, thought to represent an edematous ampulla of Vater, has been described. Striking thickening of the gallbladder wall with prominent wall striations suggestive of acute acalculous cholecystitis may occur. [129], [131], [133]

Tumors

Cholangiocarcinoma Primary bile duct tumor, cholangiocarcinoma, has two forms: peripheral cholangiocarcinoma and hilar cholangiocarcinoma (Klatskin tumor). [134], [135] In the United States, peripheral cholangiocarcinoma is three times more common than hilar cholangiocarcinoma. In Japan, hilar and peripheral cholangiocarcinoma occur in equal numbers. [136] Peripheral cholangiocarcinoma is usually a large tumor. Hilar cholangiocarcinomas present earlier with ductal obstruction and jaundice and are more difficult to image because the tumors are smaller. Cholangiocarcinoma is usually hypoechoic (Fig. 153-14), but isoechoic and hyperechoic tumors may occur. [137]

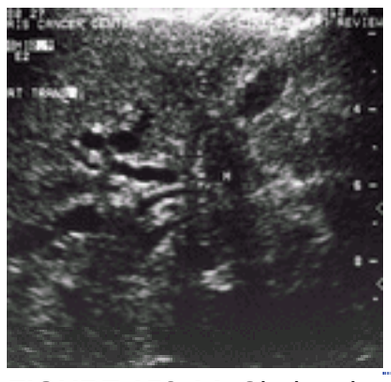

FIGURE 153-14. Cholangiocarcinoma. Transverse sonogram in the region of the porta hepatis shows biliary dilation and a hypoechoic mass (*M*).

Color and spectral Doppler ultrasonography are useful in determining whether cholangiocarcinoma is resectable. [138], [139] Cholangiocarcinoma is unresectable when one of the following is present in both the left and right lobes: lobar atrophy, vascular invasion, or obstruction of two or more sectorial (segmental) ducts. Vascular invasion, narrowing, and tumor contiguity are readily demonstrated with color Doppler ultrasonography. Duct involvement can be diagnosed when a duct is obstructed by a mass or when wall thickening or an intraductal mass is identified. When two or more ducts peripheral to the left or right hepatic duct are involved by the mass, sectorial (segmental) duct involvement is present. Sectorial ducts are those that arise from different liver segments and join to form the main left and right hepatic ducts. *The ultrasonographic evaluation of the bile ducts should be performed before stenting, if at all possible.* Stenting may cause mural and periductal hemorrhage or edema that can obscure or simulate tumor. It may also interfere with identification of dilated sectorial ducts by decompressing them.
Biliary Cystic Neoplasm Biliary cystic neoplasms (formerly called biliary cystadenoma and cystadenocarcinoma) are rare. Lesion morphology ranges from completely cystic with a few thin septations to cystic with thick septations and large amounts of internal soft tissue. Thick, sometimes papillary septations and larger amounts of internal soft tissue characterize the mesenchymal variant, which has a greater potential for malignant behavior.

Other Conditions

Biliary Parasites Biliary parasites such as *Ascaris* species or *Clonorchis sinensis* may infest the biliary tree and cause secondary biliary tract obstruction. [140] Ultrasonographically, the adult *Ascaris* organism may be visualized as a tubular structure within the bile ducts or gallbladder. The flukes of *C sinensis* are small and not generally visible with ultrasonography. However, aggregates or clumps of parasites may be identified within the extrahepatic bile duct or within the gallbladder.
Mirizzi Syndrome Mirizzi syndrome is biliary obstruction caused by a gallstone impacted in the cystic duct or gallbladder neck. Extrinsic compression and inflammation obstruct the extrahepatic bile duct. Ultrasonography may suggest the diagnosis when a stone is impacted in the neck of the gallbladder, proximal bile duct dilation is present, and the distal common bile duct is normal.
Choledochal Cysts Of the four main types of choledochal cysts, about 90% of cases involve dilation of the extrahepatic bile duct (type I). Ultrasonographically, there is obvious dilation of the extrahepatic duct, often associated with varying degrees of intrahepatic dilation. Dilation may be cystic (IA), focal (IB), or fusiform (IC). Other types include choledochal diverticula (type II), choledochocele (type III), and multiple intrahepatic or extrahepatic cysts (type IV). [141]
Caroli Disease Caroli disease is cystic dilation of the intrahepatic bile ducts. Some cases are associated with a choledochal cyst (Todani type 4A). Ultrasonographically, Caroli disease is characterized by multiple small cystic areas that may or may not appear to communicate with the unaffected bile ducts. A central vessel identified with Doppler ultrasonography supports the diagnosis. Other findings include renal cysts, cirrhosis, or liver abscess.
Bilomas Bilomas are encapsulated bile collections caused by rupture, usually traumatic, of the biliary tree. [142] Large asymptomatic bilomas are not rare. Ultrasonographically, bilomas are usually well-defined anechoic collections with striking through-transmission of sound. Detection of a lesion with this appearance 2 to 6 weeks after liver trauma strongly suggests the diagnosis. [143] Occasionally, bilomas are more complex, with internal debris and septations and less pronounced through-transmission, which are features that may also occur with abscesses. In these cases, needle aspiration can exclude infection and confirm the presence of bile by laboratory analysis for bilirubin. Bilomas discovered incidentally in asymptomatic patients generally do not require treatment; they resolve spontaneously within a few weeks. Imaging studies alone cannot always distinguish bilomas from abscesses, chronic hematomas, or hepatic cysts. Percutaneous drainage, when required, is often an effective alternative to surgery. [142], [144]

PANCREAS

Indications for Ultrasonography

Although CT remains the most sensitive means of evaluating pancreatic disease, modern ultrasound technology and new scanning techniques (oral contrast, compression scanning) are reestablishing ultrasonography as a useful and clinically relevant pancreatic imaging technique. Ultrasonography is indicated in all patients with acute pancreatitis, not to evaluate the pancreas itself but rather to detect gallstones and biliary dilation. CT is superior to ultrasonography in evaluating potential complications of pancreatitis, but ultrasonography may be used to follow pancreatitis-associated fluid collections and can guide biopsy, drainage, and aspiration of selected pancreatic lesions. Ultrasonography is the primary imaging method to screen patients with jaundice or abdominal pain. Thus, ultrasonography may detect acute or chronic pancreatitis or reveal pancreatic masses. Ultrasonography may be more effective than CT in determining whether a lesion is pancreatic or contiguous to the pancreas. Ultrasonography is occasionally useful to characterize abnormalities noted on CT, determining, for example, whether a lesion is cystic or solid.

Patient Preparation and Technique

Pancreatic ultrasonography is easier after the patient has fasted overnight or for at least 6 hours. This improves visualization of the gallbladder and minimizes GI gas. The spatial resolution of ultrasonography is better than that of CT and MRI when conventional scanning parameters are used. In some patients, however, CT is needed to visualize the pancreas adequately. With ultrasonography, optimal pancreatic visualization requires that intervening GI content (gas and fluid) be displaced by compression or by filling the gut with water or GI contrast material. For the head and body, we prefer compression, using 3- to 7-MHz curved array transducers. Compression results in better resolution—the transducer is closer to the pancreas. The left lateral decubitus position may be useful, with visualization of the pancreatic head through the gallbladder or liver. Oral administration of water may be helpful if compression is unsuccessful.

The tail of the pancreas is more difficult to image. Routine pancreatic scanning should include coronal and axial views through the spleen and left kidney. The normal tail is usually not seen, but lesions invisible on other views, especially pseudocysts, are occasionally delineated. Water or contrast in the stomach is useful for imaging the tail. Right anterior oblique positioning is useful to view the tail though the water-containing stomach. Color-flow imaging can assess vascular complications of pancreatitis and delineate vascular relationships to pancreatic neoplasms.

Normal Anatomy

The normal pancreatic parenchyma is finely homogeneous and isoechoic to hyperechoic compared with liver parenchyma. The pancreas is a retroperitoneal organ that lies draped over the spine, aorta, and IVC. The pancreatic head is medial to and intimately related to the sweep of the second and third portions of the duodenum. The head is ventral to the IVC and medial to the superior mesenteric artery and vein (Fig. 153-15). In cross section, the round and sonolucent common bile duct is seen in the posterior head, sweeping caudally and to the right as it passes into the duodenum. The body lies ventral to the aorta, splenic vein, and superior mesenteric artery, behind the stomach (with the collapsed potential space of the lesser peritoneal sac between). The pancreatic tail nestles into the splenic hilus and is closely related to the upper pole of the left kidney.

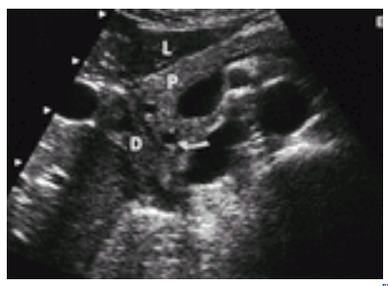

FIGURE 153-15. Normal pancreatic head. The pancreas (*P*) is more echogenic than the hepatic parenchyma (*L*). Note the anatomic relationship of the duodenum (*D*) to the pancreatic head. The rounded anechoic areas within the pancreatic head are the gastroduodenal artery (anteriorly) and the common bile duct (*curved arrow*).

Acute Pancreatitis

Evaluation with ultrasonography is mandatory for all patients, even alcoholic patients, during their first attack of acute pancreatitis. Ultrasonography in patients with acute pancreatitis focuses on the gallbladder and bile ducts—the gallbladder for stones (as a cause of pancreatitis) and the bile duct for choledocholithiasis and obstruction.

The reported prevalence of acute pancreatitis–associated abnormalities varies widely—33% to 90% of patients. Pancreatic echogenicity typically decreases in acute pancreatitis because of interstitial edema but can be considered abnormally hypoechoic only when echogenicity is less than liver parenchyma. Because the normal pancreas may be more echogenic than the liver, echogenicity is normal in many patients with acute pancreatitis. In patients with echogenic livers from fatty infiltration, the normal pancreas may be relatively hypoechoic, simulating the findings of pancreatitis. This is called *pseudopancreatitis*. Rarely, echogenicity may actually increase with acute pancreatitis, owing to hemorrhage, necrosis, or fat saponification. With newer ultrasonographic systems, inhomogeneity of the parenchyma is often evident in acute pancreatitis.

Enlargement of the pancreas in acute pancreatitis is probably almost universal. Unfortunately, enlargement may be difficult to judge because pancreatic size before the onset of pancreatitis is unknown and varies widely from individual to individual. It is not unusual to detect extrapancreatic inflammatory changes even when the pancreas appears normal. Thus, extrapancreatic abnormalities may be the only ultrasonographic findings that suggest the diagnosis of acute pancreatitis. Hypoechoic inflammation is often seen around the pancreas, in the anterior retroperitoneal spaces bilaterally, in the transverse mesocolon, around splanchnic vessels, and elsewhere. [145] Acute fluid collections, pseudocysts, and abscesses may be imaged. Color and spectral Doppler ultrasonography may be useful in detecting pancreatitis-associated vascular complications, such as pseudo- aneurysms and portal thrombosis.

Acute Pancreatic Fluid Collections and Pseudocysts Acute pancreatic fluid collections and pseudocysts are the most common complications of pancreatitis. Pancreatic fluid collections consist of a combination of pancreatic juice, edema fluid, hemorrhage, and necrotic tissue resulting from acute pancreatic inflammation, enzymatic autolysis, and pancreatic ductal disruption. Two features distinguish a pancreatic pseudocyst from an acute fluid collection: persistence past 4 to 6 weeks, and a well-defined wall (fibrous pseudocapsule). [146] Acute fluid collections occur in about 30% to 50% of patients with severe pancreatitis, but more than half of these regress spontaneously. CT is more sensitive than ultrasonography in diagnosing acute pancreatic fluid collections and pseudocysts. [145] Once an acute pancreatic fluid collection is identified with CT or ultrasonography, serial studies should be performed to document either resolution or persistence as a pseudocyst. If ultrasonography shows the pancreatic fluid collection well, it is the most cost-effective method for serial evaluation. CT should be performed immediately before any surgical drainage of a pseudocyst, to identify any lesions not detected by ultrasonography. Acute fluid collections and pseudocysts have a variable ultrasonographic appearance depending on the associated internal hemorrhage and debris. Acute fluid collections may be anechoic to hyperechoic but are generally hypoechoic with internal debris. Enhanced through-transmission is almost always present but is variable in degree. Color Doppler ultrasonography should be used to ensure that all discrete cystic-appearing areas are not pseudoaneurysms.

Pancreatic Abscess and Infected Pancreatic Necrosis The two major infectious complications of acute pancreatitis are infected pancreatic necrosis and pancreatic abscess. Pancreatic abscess is a circumscribed collection of pus that contains little or no necrosis. Infected pancreatic necrosis is a region of infected nonviable pancreatic parenchyma. Pancreatic abscess is associated with a lower mortality rate (7% to 33%) than is infected pancreatic necrosis (about 40%). [147] Necrosis is diagnosed by contrast CT—no enhancement of the pancreatic parenchyma corresponds to necrosis. Currently, ultrasound cannot diagnose pancreatic necrosis. Contrast CT, because of its superiority to ultrasonography as a survey modality, is preferred if infectious complications are sought. Unfortunately, image findings alone cannot distinguish infected from uninfected collections, [148] so that aggressive use of CT- or ultrasound-guided needle aspiration is mandatory to diagnose

infection. The treatment of infectious complications of pancreatitis is controversial. Localized pancreatic abscess responds well, albeit imperfectly, to percutaneous drainage. Van Sonnenberg and colleagues [149] reported successful cure in 69% of patients with pancreatic abscess by percutaneous drainage alone.
Hemorrhage and Vascular Complications Erosion of small vessels is the most frequent cause of pancreatitis-associated hemorrhage. It may result in bleeding into the pancreatic bed or the pancreatic duct or in a pseudocyst. Peripancreatic pseudoaneurysm [150] is rare but potentially catastrophic. Pseudoaneurysms occur most often in the splenic artery (42%), gastroduodenal artery (22%), and small peripancreatic arteries (25%). Contrast CT and Doppler ultrasonography can identify pseudoaneurysms, but smaller pseudoaneurysms (i.e., less than 1 cm) are more reliably diagnosed angiographically. Inflammation from acute pancreatitis may cause venous occlusion. The splenic vein is most commonly affected and may result in gastric varices. The superior mesenteric or portal veins are also occasionally involved. Color Doppler ultrasonography and contrast CT may be very useful to diagnose pancreatitis-related thrombosis in the portal venous system.

Chronic Pancreatitis

Image findings are poor predictors of the clinical severity of chronic pancreatitis, [151] although patients with severe exocrine insufficiency are more likely to have abnormal pancreatic size and contour irregularity. [152] Multiple calcified stones located within the main or branch pancreatic ducts are the hallmark of chronic pancreatitis, usually alcohol related. Ultrasonography is less sensitive than CT in detecting pancreatic calcifications. On occasion, the pancreas is diffusely hyperechoic. This may be difficult to judge ultrasonographically because the pancreas is normally more echogenic than adjacent parenchymal organs like the liver. Bolondi [153] believes that dilation of the main pancreatic duct is the most reliable sign of chronic pancreatitis. The double-duct sign (dilation of pancreatic and bile ducts) may occur not only in pancreatic carcinoma but also in chronic pancreatitis. [154] Glandular atrophy or fatty infiltration may occur, making evaluation of the pancreas difficult ultrasonographically. Chronic asymptomatic pseudocysts may be found in patients with chronic pancreatitis.

Focal masses that can simulate neoplasm occur in about one third of patients with chronic pancreatitis. Calcification within the mass is strongly suggestive, but not definitive, of chronic pancreatitis. Uncalcified isoechoic or hypoechoic masses may require endoscopic retrograde cholangiopancreatography or biopsy to exclude malignancy. [151]

Neoplasms

Ultrasonography is the primary imaging method to screen patients with jaundice or abdominal pain. Thus, ultrasonography often detects pancreatic masses. Ultrasonography may be useful to characterize the internal architecture of masses, information that may be useful in classifying neoplasms. Ultrasonography may detect a carcinoma as an area of decreased echogenicity within the gland when no morphologic or attenuation abnormality is noted on CT. [155]

Ductal Adenocarcinoma Ultrasonographically, pancreatic carcinoma is typically a hypoechoic mass that deforms the gland's morphology (Fig. 153-16). Occasionally, pancreatic carcinoma is echogenic. A few scattered calcifications or cystic areas may occur occasionally (about 5% each). [156] Secondary findings of carcinoma include ductal dilation (biliary and pancreatic; see Fig. 153-16), vascular and extraglandular invasion, and metastatic disease. Pseudocysts, related to obstruction of a pancreatic duct, have been reported in as many as 11% of patients, [157] although in our experience, carcinoma-related pseudocysts are less frequent.

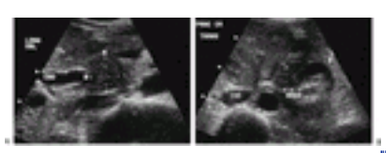

FIGURE 153-16. Pancreatic carcinoma with obstruction of the bile duct. **A**: The long-axis view of the common bile duct shows a 3.5-cm mass (highlighted by calipers A and B) that obstructs the common bile duct (*CBD*). **B**: A transverse sonogram obtained with the patient in the right anterior oblique position shows the mass (*arrows*). The mass is hypoechoic, compared with the normal pancreatic parenchyma (*p*). IVC, inferior vena cava; LRV, left renal vein.

Most recent discussions of imaging in pancreatic and periampullary neoplasms have almost totally ignored ultrasonography [158], [159] despite a 1988 study, which showed that gray-scale ultrasonography was useful in assessing resectability. [160] We and others have shown that color Doppler ultrasonography can effectively assess resectability, specifically vascular involvement by tumor, with accuracy comparable to that of CT. [161], [162], [163] and [164]
Ductal Adenocarcinoma Variants Many histological variants of pancreatic ductal adenocarcinoma are indistinguishable on images from tumors with the usual histological features. Acinar center cell carcinoma and pleomorphic giant cell carcinoma can be larger and exhibit central necrosis. [165] Mucinous ductectatic carcinoma and mucin hypersecreting carcinoma are variants of ductal adenocarcinoma. [165], [166] These lesions have markedly dilated, mucin-filled, pancreatic branch ducts, usually in the pancreatic head. Ultrasonographically, mucinous ductectatic carcinomas show multiple sonolucent, dilated ducts. A discrete mass may not be evident. Mucin hypersecreting carcinoma usually has more extensive ductal dilation.
Cystic Pancreatic Lesions The most common cystic pancreatic neoplasms are serous cystic neoplasm (microcystic adenoma) and mucinous cystic neoplasms. [167] Morphologically, microcystic adenomas comprise many tiny cysts, most smaller than 2 cm. The most distinctive feature, present in about one half of tumors, is a central radial fibrotic scar that frequently calcifies. Ultrasonographically, microcystic adenomas are echogenic in regions where there are many tiny cysts. Through-transmission is frequent. Hypoechoic, larger cysts may be seen. Calcifications are imaged more often with CT than ultrasonography. Microcystic adenomas are almost universally benign. [168] Mucinous cystic neoplasms consist of large cysts, usually easily imaged with CT or ultrasonography. Most are located in the tail or body of the pancreas and are much more common in female patients. Calcifications occur in about 20% of these neoplasms (compared with about one half of serous and microcystic neoplasms). Ultrasonography usually reveals a mass with internal cysts of variable size. Unilocular lesions may occur. The septations may be few or many, thin or thick, and polypoid. The internal architecture is often shown to better advantage by ultrasonography than CT. Mucinous cystic neoplasms may be confused with rarer cystic neoplasms such as papillary cystic tumor, cystic islet cell tumor, lymphangioma, cystic metastases, or nonneoplastic lesions, such as pseudocyst, abscess, and echinococcal disease. [169] Solid and papillary epithelial neoplasm, a low-grade malignancy found most frequently in young female patients, tends to be large and well encapsulated. Tumors can be mostly solid, mixed cystic and solid, or almost entirely cystic. Less common cystic neoplastic tumors include papillary cystic tumor, cystic islet cell tumors, and acinar cell cystadenocarcinoma. Simple cysts of the pancreas are uncommon. They occur sporadically or in association with von Hippel–Lindau syndrome and, rarely, in renal polycystic disease.
Endocrine Tumors It is difficult to image functional islet cell tumors; they are usually small when the patient presents with hormonal abnormalities. Insulinomas and gastrinomas are frequently less than 2 cm in diameter. In contrast, "nonfunctional" islet cell tumors can be large at the time of presentation. Ultrasonographic detection rates are 25% to 60% for insulinomas [170] and about 20% for gastrinomas. [171] Ultrasonographically, islet cell tumors are usually hypoechoic, well-defined, and round or oval in shape. Intraoperative ultrasonography (IOUS) is useful in localizing occult neoplasms.

GASTROINTESTINAL TRACT

Indications for Ultrasonography

The most common indication for GI tract ultrasonography is evaluation of the patient with right lower quadrant pain and possible appendicitis. Ultrasonography may also be useful in the diagnosis of diverticulitis, small bowel obstruction, and bulky mesenteric or GI neoplasms. One of the more useful applications of ultrasonography of the GI tract is endolumenal ultrasound to stage the depth of GI tract neoplasms. This is discussed in Chapter 154.

Patient Preparation and Technique

Patients undergoing ultrasonographic evaluation for possible GI tract abnormalities typically require no special preparation. Patients with right lower quadrant pain and possible appendicitis can be imaged immediately after initial assessment in the emergency room. Evaluation of the stomach and duodenum may be facilitated by having the patient drink sufficient water to distend the upper GI tract. A water enema may be useful in selected patients to evaluate pelvic masses.

Graded compression ultrasonography is the most important technique for evaluation of possible appendicitis and other focal GI tract abnormalities. [172] This technique takes advantage of the fact that lesions infiltrating the bowel wall alter its compressibility. The normal GI tract can be readily compressed by the sonologist when moderate pressure is applied with the transducer. However, inflammatory or neoplastic disorders of the bowel alter its compressibility and render it relatively noncompressible. [173], [174]

Ultrasonography has a number of important advantages compared with CT in evaluating the GI tract. Real-time observation of bowel peristalsis is possible with ultrasonography. Delineation of layers of the bowel wall (most importantly, the echogenic submucosal layer) is also readily apparent with ultrasonography. Rapid multiplanar imaging can be performed and vascular flow can be assessed without contrast agents with the use of color and power Doppler (the most sensitive form of color Doppler ultrasonography). [175], [176] Finally, ultrasonography is an interactive technique, and the patient can be scanned at the site of maximal pain and tenderness to correlate symptoms with anatomic abnormalities. It should be noted, however, that ultrasonography is often unable to image all the lumenal GI tract mesentery and subperitoneal space reliably because of bowel gas.

Graded compression is generally performed with a high-resolution linear array transducer (5 to 10 MHz) depending on the depth of the structures to be imaged. For most other GI studies, a 5-MHz curved array transducer yields the best results.

Normal Anatomy

Standard abdominal transducers do not delineate all five layers of the bowel wall. The echogenic submucosal layer, however, is clearly visible and serves as a constant anatomic feature that is an extremely useful landmark to identify an intra-abdominal structure as a bowel loop. Pathological processes that cause ulceration and necrosis of the bowel lead to either focal or global loss of visualization of the echogenic submucosa. [177] Primary neoplasms involving the bowel wall may result in focal bowel wall thickening, referred to as the *target sign* or *pseudokidney sign*. [178] Tumor infiltrating the bowel wall appears as a hypoechoic mass (mimicking the cortex of the kidney). The echogenic mucosal surface lumen is preserved, mimicking the fat-containing hilum of the kidney. Gas trapped within ulcerating lesions involving bowel wall may result in high-amplitude echoes with acoustic reverberation artifacts within the submucosal layers of the bowel wall.

Color Doppler ultrasonography may be a useful adjunct to gray-scale imaging in evaluation of patients with focal bowel wall thickening. [175], [176] Increased arterial flow within the involved segment suggests either inflammation or infection; diminished or absent flow suggests intramural hemorrhage, ischemia, or infarction. The vascularity of GI tract tumors is variable, but in general, adenocarcinomas are hypovascular.

Acute Appendicitis

In patients with an uncertain clinical diagnosis of appendicitis, ultrasonography is preferred to CT in women of childbearing age, pediatric patients, and thin, adult male patients. CT is preferred in patients who are obese or if appendiceal perforation is suspected. [179] The ultrasonographic criteria for diagnosing acute appendicitis include a noncompressible appendix measuring 7 mm or more in greatest anteroposterior diameter [180] (Fig. 153-17). An appendix measuring 5 mm or less is virtually always normal. Appendices measuring 5 to 6 mm are equivocal for appendicitis, and clinical observation, repeated ultrasonography, or both should be considered. [180] In a patient with right lower quadrant pain, the presence of an appendicolith within a noncompressible appendix generally indicates acute appendicitis regardless of the size of the appendix. In some patients, early acute appendicitis is confined to the distal tip of the appendix. [181] It is therefore important always to visualize the entire length of the appendix to evaluate for distal appendicitis.

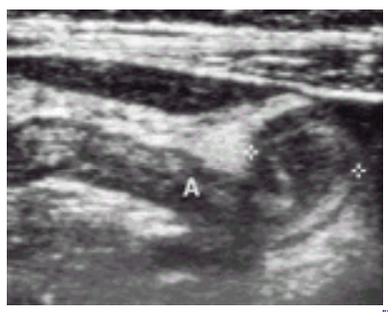

FIGURE 153-17. Acute appendicitis. Note distended appendix (*A*) with dilated tip (*cursors*).

Diverticulitis

In most patients, CT is the imaging method of choice to evaluate patients who present with left lower quadrant pain and possible sigmoid diverticulitis. [182] This is particularly true in septic patients with possible paracolonic abscesses that may require percutaneous or surgical drainage. [183] Nevertheless, ultrasonography may play a useful role as a screening examination in patients with pelvic pain and unclear signs and symptoms of diverticulitis. The ultrasonographic findings in acute diverticulitis include focal mural thickening of the bowel and increased echogenicity of the mesenteric fat with hyperemia on color Doppler imaging. [184] In some patients, the infected colonic diverticulum may be identified as an echogenic intramural focus due to either gas or echogenic pus within the diverticulum. [184] On occasion, ectopic gas bubbles may be identified in the sigmoid mesentery due to their characteristic high-amplitude linear echoes and distal reverberation artifacts.

Enteritis and Colitis

The ultrasonographic findings of enteritis or colitis are often nonspecific. [185] Typical findings include focal areas of mural thickening of the bowel, often with associated adjacent adenopathy. There is lack of compressibility of the involved segment with graded compression. Increased color-flow Doppler may be demonstrated with high-frequency probes. Fibrofatty mesenteric masses may be identified as focal echogenic areas in Crohn's disease. In general, CT is superior to ultrasonography in identifying mesenteric abscesses, colovesicular fistulas, and retroperitoneal abnormalities that accompany Crohn's disease. [185]

Mesenteric Adenitis

In the presence of a normal appendix or ultrasonography, the identification of enlarged mesenteric lymph nodes is highly suggestive of mesenteric adenitis. There may also be associated thickening of the terminal ileum. [186] Once this diagnosis is suggested, patients should be observed clinically without surgery intervention. Symptoms of mesenteric adenitis typically resolve spontaneously without specific therapy. Color Doppler may be useful to highlight the hypoechoic lymph nodes adjacent to the flow in mesenteric vessels.

Intestinal Obstruction

Ultrasonography and CT may play an important role in the patient with possible small bowel obstruction. [187] The characteristic ultrasonographic features are dilated fluid-filled loops with increased peristalsis on real-time examination. Fluid-filled levels and echogenic intralumenal debris may also be seen within the dilated, obstructed bowel. In any patient with possible obstruction, a specific attempt should be made to identify a transition zone between dilated and nondilated bowel. This is often the most specific finding to confirm the diagnosis of obstruction. On occasion, ultrasonography may identify GI tract tumors, mesenteric masses, abscesses, and other intra-abdominal pathology that caused the obstruction. The appearance of intussusception is characteristic on ultrasonography because there is a target appearance of the invaginated intussusceptum. A crescent-shaped intralumenal area of echogenic fat representing invaginated mesentery is identified at the level of the lead point.

Gastric outlet obstruction may be diagnosed whenever marked dilation of the stomach is noted with a collapsed duodenum. In selected patients, however, gastroparesis may have a similar appearance. Closed-loop obstruction characteristically results in a single U-shaped aperistaltic loop of dilated bowel (Fig. 153-18). Hyperperistaltic bowel is noted proximal to this loop.

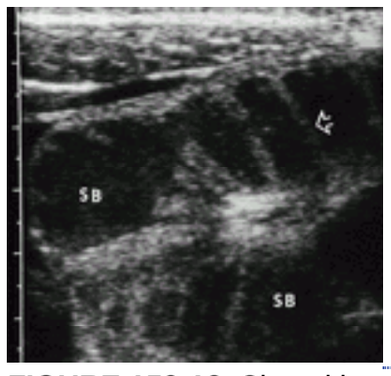

FIGURE 153-18. Closed loop small bowel obstruction. Dilated U-shaped loop of small bowel (*SB*) represents closed loop obstruction.

Small Bowel Infarction

In most patients with suspected mesenteric ischemia and small bowel infarction, CT, mesenteric angiography, or both are performed emergently. However, in selected patients, ultrasonography may be performed owing to confusing signs and symptoms on clinical presentation. Mural thickening of the small bowel wall due to either intramural hemorrhage or edema may be visualized as a noncompressible area of mural thickening. [175], [176] In the appropriate clinical setting, absence of vascular flow on color and, more specifically, power Doppler suggests the diagnosis of ischemia.

Gastrointestinal Tract Tumors

In patients with palpable abdominal masses, ultrasonography is often valuable in identifying the organ of origin of the lesion. As previously noted, the pseudokidney sign may be due to tumors infiltrating through muscular elements of the bowel wall (Fig. 153-19). Caution must be observed, however, in that focal inflammatory or ischemic processes may mimic neoplasms on ultrasonography. Endoscopy, barium studies, and CT are often required to establish a more precise diagnosis. Both lymphoma and GI stromal tumors result in bulky, exophytic masses that are typically hypoechoic on ultrasonography. Areas of liquefied hemorrhage or necroses are more typical of GI stromal tumor.

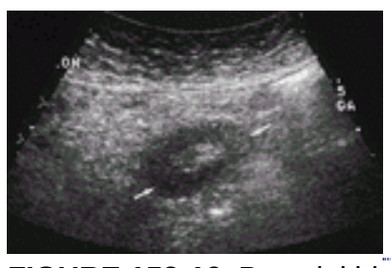
FIGURE 153-19. Pseudokidney sign of gastrointestinal tumor. Sagittal sonogram of midabdomen demonstrating a mass (*arrows*) that resembles a kidney.

PERITONEAL CAVITY

Indications for Ultrasonography

The most common indication to evaluate the peritoneal cavity with ultrasonography is to search for intraperitoneal fluid collections such as ascites, abscesses, or hemorrhage. Ultrasonography is useful not only to identify intraperitoneal fluid collections but also to guide percutaneous needle aspiration for definitive diagnosis. Solid peritoneal masses representing either primary or metastatic tumors may also be detected on occasion with ultrasonography. However, CT is the preferred imaging modality in patients with a clinical suspicion for peritoneal metastases.

Patient Preparation and Technique

Scanning of the pelvic peritoneal spaces is easier when the bladder is full. The distended bladder provides an excellent acoustic window to evaluate the cul-de-sac or retrovesical recess. However, the remainder of the peritoneal cavity can readily be imaged with little or no patient preparation. In general, a curved 3.5- or 5-MHz transducer provides adequate penetration for a general abdominal survey to detect intraperitoneal fluid in the upper peritoneal compartments. Where access is difficult, sector transducers may be useful. High-resolution linear arrays (5 to 7.5 MHz) are best for evaluation of superficial peritoneal implants and omental abnormalities.

Intraperitoneal Fluid Collections

Ultrasonography excels in demonstrating even minute quantities of intraperitoneal fluid (Fig. 153-20). With endovaginal scanning, small amounts of intraperitoneal fluid may routinely be demonstrated in the cul-de-sac as a normal finding in ovulating women. In the upper abdomen, free intraperitoneal fluid first collects in the hepatorenal fossa, which is the most dependent portion of the supramesocolic peritoneal cavity. [188] As fluid increases within the peritoneal cavity, it involves the subphrenic, subhepatic, and intraloop compartments. [189] Massive ascites typically displaces bowel loops centrally unless tethered by adhesions. [190]

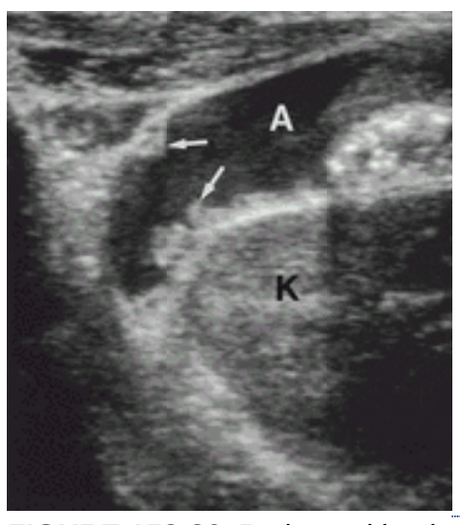

FIGURE 153-20. Peritoneal implants from mesothelioma. Transverse scan of right lower quadrant demonstrates ascites (*A*) and plaquelike peritoneal implants (*arrows*). K, right kidney.

Uncomplicated serous fluid collections are typically anechoic. On ultrasonography, complex fluid characteristically demonstrates internal debris, septations, and low-level echoes. The differential diagnosis of complex intraperitoneal fluid includes hemorrhage, infection, and carcinomatosis. On occasion, long-standing bland ascites may contain internal septations and be loculated. However, ascites that is not infected generally has no mass effect and passively conforms to its intraperitoneal components. Intraperitoneal abscesses, on the other hand, typically exhibit mass effect and may displace adjacent bowel or adjacent solid organs. It is not always possible to differentiate loculated bland ascites from an abscess by ultrasonographic criteria alone. Therefore, diagnostic needle aspiration is often required.

Intraperitoneal Abscesses

CT and ultrasonography have greatly facilitated the diagnosis of intra-abdominal abscesses. [191], [192], [193] and [194] With both modalities, the predominant finding is a localized fluid collection with mass effect. Gas bubbles and an air-fluid level may be present in some patients but are not identified in most intra-abdominal abscesses. Ultrasonography is the imaging method of choice for hepatic, pericholecystic, and pelvic abscesses. CT is superior to ultrasonography in patients with pancreatic, retroperitoneal, paraspinous, intraloop, or gas-forming abscess. [192] In the postoperative setting, CT is often preferred to ultrasonography owing to frequent problems with access as a result of open wounds and surgical drains.

The ultrasonographic appearance of intra-abdominal abscesses is variable and depends on the presence or absence of septations and solid debris. [194] Also, there is considerable overlap with the ultrasonographic appearance of other complex intraperitoneal fluid collections, such as lympho- celes, bilomas, pancreatic pseudocysts, postoperative seromas, and hematomas. Because of the attenuation of sound by proteinaceous debris, many intra-abdominal abscesses demonstrate little if any enhanced through-transmission of sound. Thus, an intra-abdominal abscess may ultrasonographically mimic a solid hypoechoic mass. [194] This pitfall should always be kept in mind in a febrile patient. Color Doppler may be of value in resolving this dilemma because liquefied abscesses lack internal vascularity. Intra-abdominal enteric abscesses are often walled off by adjacent mesenteric or omental fat. In some patients, it may be possible with ultrasonography to identify adjacent abnormal bowel to identify the cause of the abscess (i.e., appendicitis or diverticulitis). Both CT and ultrasonography may be valuable for guidance of percutaneous needle aspiration for confirmation of an abscess and subsequent catheter insertion for definitive therapy. [191]

Tumors of the Peritoneum, Mesentery, and Omentum

The most common intraperitoneal neoplasms are metastatic carcinomas. Most of these lesions are tiny, plaquelike implants on the surface of the bowel or mesentery that cannot be imaged with any current diagnostic techniques. In the presence of ascites, peritoneal implants may be detected by ultrasonography as solid masses arising from the peritoneum or mesentery surrounded by peritoneal fluid (see Fig. 153-20). Thickened septations may also be demonstrated ultrasonographically. Diffuse infiltration of the omentum is a characteristic finding in patients with ovarian metastases, carcinomatosis from GI tract malignances, and primary omental tumors. Primary neoplasms involving the omentum, such as mesotheliomas, can be imaged with graded compression ultrasonography using high-frequency linear array transducers. [195] Diffuse omental infiltration (“omental caking”) typically results in a broad, bandlike mass anterior to the transverse colon. Because of its superficial location, these abnormalities are often best imaged with a high-resolution linear array transducer.

Pseudomyxoma peritonei, an unusual form of peritoneal carcinomatosis caused by mucinous adenocarcinomas of the appendix, ovary, or colon, [196] results in gelatinous intraperitoneal masses that cause subtle scalloping of solid viscera, such as the liver and spleen. Echogenic intraperitoneal masses containing debris may be detected ultrasonographically.

Mesenteric masses may often be best imaged with ultrasonography using a graded compression technique and color Doppler imaging. Adenopathy with the dorsal and ventral leaves of the mesentery may be noted to encase the mesenteric vessels highlighted by color Doppler. Primary mesenteric and peritoneal tumors such as mesothelioma cannot be distinguished from the more common metastatic peritoneal tumors.

INTRAOPERATIVE ULTRASONOGRAPHY

IOUS is an important and rapidly developing diagnostic imaging technique. It provides indispensable information that influences clinical management and choice of surgical procedure at the time of surgery. [197] Several recent technical advances have made IOUS even more effective, including the miniaturization of transducers, the use of spectral and color Doppler, and, most recently, the development of laparoscopic ultrasonography.

Indications

In gastroenterology, IOUS is most often used in evaluating patients who are candidates for surgical resection of primary or metastatic hepatic malignancies. [198] It is essential for optimal detection of all liver lesions and is far superior to all preoperative imaging, including MRI and CT portography. It is even better than surgical inspection and palpation. [199], [200] and [201] IOUS is also important in intraoperative pancreatic imaging–searching for small occult tumors, assessing tumor extension, and detecting metastatic disease in draining lymph nodes and the liver. Laparoscopic ultrasonography is important in gallbladder surgery, replacing intraoperative cholangiography in some centers. Laparoscopic ultrasonography has also been used to stage patients with bowel malignancies, particularly gastric tumors.

IOUS guidance facilitates more accurate and safer biopsy of deep-seated nonpalpable lesions and small lesions adjacent to critical vascular structures. It can effectively guide drainage of cysts, pseudocysts, and other fluid collections encountered intraoperatively. IOUS guidance is very useful for tumor ablation with cryosurgery, alcohol injection, or hyperthermic ablation with radiofrequency, laser, or microwave energy sources.

Patient Preparation and Technique

IOUS requires no patient preparation beyond that required for the surgical procedure itself. For the liver, the optimal frequency is 5 MHz, and the optimal probe design is a T-shaped probe, which images perpendicularly to the electrical cord (side-fire probe). Transducers of 5 MHz achieve sufficient penetration to scan the entire liver from the anterior surface. Anterior surface scanning is best because it is faster and allows the best acoustic coupling. [202] The pancreas is scanned with high-resolution 7.5-MHz end-fire transducers (i.e., the imaging surface is inline with the electrical cord). These higher-frequency transducers provide adequate penetration because the pancreas is much thinner than the liver. Similarly, an end-fire 7.5-MHz probe is suitable for evaluation of the gallbladder and common bile duct.

Some transducers are designed to tolerate sterilization using ethylene oxide, obviating transducer covers. Transducers are sometimes cleaned by immersion in glutaraldehyde for 30 to 60 minutes. Most practitioners, however, use sterile sheaths to cover the probes and their cords. A small amount of sterile gel is used as a coupling agent. If needed, sterile saline can be used to moisten the surface of the organ scanned.

Liver

The principal uses of liver IOUS include detection of occult primary or metastatic tumors; demonstration of lobar and segmental anatomy and of vascular supply and drainage, including accessory and anomalous vasculature; assessment of resectability and planning appropriate lobar, segmental, or nonsegmental resection; and guidance for biopsy, aspiration, or therapeutic ablation.

The superb spatial resolution of IOUS allows for consistent imaging of cysts as small as 1 to 3 mm and solid nodules as small as 3 to 5 mm. This far surpasses the usual lower limit of about 1 cm for detection of solid lesions by preoperative imaging techniques. IOUS detects 25% to 35% more lesions in the liver than preoperative imaging—most of these are 1 cm or smaller in size. [199], [200] Although some of these lesions are on the surface and can be seen or palpated by the surgeon, most additional lesions are detected only by IOUS. CT arterial portography can demonstrate 85% to 90% of the IOUS-detected lesions, [201] but there is an attendant decrease in specificity, with false-positive results owing to tiny hepatic cysts and hemangiomas, as well as perfusion artifacts. [203], [204] Experience with liver imaging is crucial to evaluate tumor extent accurately and to avoid misinterpretation of other incidentally discovered but benign lesions and pseudolesions as malignancy. Malignant tumors of similar size tend to look very much alike in each individual patient. Therefore, if two distinctly different-appearing nodules of similar size are present, one should strongly consider alternative diagnoses. Coexistent hemangiomas, for example, are common.

Hepatic pseudolesions are common, frequently caused by focal fatty infiltration or focal spared areas within a diffusely fatty liver. Pseudolesions often occur in

predictable locations such as at the porta hepatis and bare area of the liver adjacent to the gallbladder. They frequently have geometric shapes, such as triangular or rectangular configurations.

IOUS evaluation of the hepatic vasculature is often important in determining resectability. IOUS can accurately assess tumor invasion, as commonly occurs with HCC. IOUS can readily and completely delineate tumor thrombi, which generally render the patient inoperable. Periportal and celiac adenopathy, another finding that renders the patient inoperable, also can be demonstrated. The presence of an inferior accessory right hepatic vein should always be determined. This accessory hepatic vein can be torn during mobilization of the liver. Its presence can allow for a more extensive left trisegmentectomy than would otherwise be an option.

With experience, IOUS-guided biopsy (performed freehand) is feasible, even for small, deeply situated lesions. IOUS guidance is indispensable in hepatic cryosurgical tumor ablation, both for accurate placement of the cryoprobes and for real-time assessment of the expanding cryolesion. IOUS monitoring of the expanding cryolesion is critical to ensure complete treatment of the entire tumor and tumor-free margins extending into the adjacent hepatic parenchyma. [205] Both thermal ablation and alcohol ablation also require accurate ultrasound guidance, whether performed percutaneously or intraoperatively.

Pancreas

IOUS is the most accurate technique for localizing islet cell tumors of the pancreas. With spatial resolution of 3 to 5 mm, high-resolution IOUS scanning can detect even small nonpalpable lesions. Most islet cell tumors are homogenous and therefore relatively hypoechoic when compared with the more highly echogenic pattern of the pancreas. [170] These lesions are also usually hypervascular, and color-flow imaging can be helpful in lesion detection. Insulinomas are usually solitary, benign, and hypoechoic, [206] but in any IOUS study for islet cell tumors, the pancreas and peripancreatic and periportal lymph nodes should be evaluated for metastatic disease. Gastrinomas are often multiple and frequently extrapancreatic in location, often being found as intramural nodules in the duodenum. Gastrinomas are also found in tissue near the pancreas and duodenum and in adjacent lymph nodes.

IOUS is useful to assess the resectability of pancreatic ductal adenocarcinoma. Invasion of the superior mesenteric vein is often found, even after negative preoperative imaging, and precludes resection of even small pancreatic head tumors. IOUS, with gray-scale and color-flow imaging, can delineate these findings effectively. IOUS may demonstrate unanticipated hepatic metastases, peripancreatic and portal lymphadenopathy, or other sites of metastatic disease, leading to surgical bypass or other palliative procedures. There is a growing experience and enthusiasm for using laparoscopic ultrasonography for local staging of cancer of the head of the pancreas, particularly to evaluate vascular invasion. [207], [208]

IOUS can also assess cystic neoplasms of the pancreas. In particular, IOUS can generally diagnose microcystic cystadenoma (serous cystic neoplasm), which is virtually always benign. If the cyst cavities are consistently very small (a few millimeters in diameter) and there are no solid components, nodules, or irregular septal thickening, a relatively confident diagnosis of microcystic adenoma can be made. This may obviate pancreatic resection and its attendant morbidity.

Some authors believe IOUS is useful in patients undergoing surgery for pancreatitis with ductal obstruction or for pancreatic pseudocyst drainage. [209] The size, extent, internal contents, and site of origin of pseudocysts and other peripancreatic fluid collections can easily be demonstrated. Real-time guidance may facilitate drainage of these fluid collections. If the pancreatic duct is obstructed proximal to a pseudocyst, then mere cyst drainage may not be sufficient, and drainage or stenting of the pancreatic duct may also be required for complete therapeutic resolution. Color-flow imaging may be important in evaluating fluid collections for the presence of pancreatitis-associated pseudoaneurysms.

In chronic pancreatitis patients with focal masses and biliary obstruction, IOUS assessment of the pancreas may allow for a definition of “a mass within the mass.” This finding is suggestive of concurrent pancreatic carcinoma. IOUS may allow accurate biopsy of small tumor sites surrounded by a fibrotic mass of chronic pancreatitis. Conversely, the lack of identification of any discrete mass within a fibrotic or calcified area of chronic pancreatitis may suggest that the biliary obstruction is benign in nature. If biopsy has also failed to demonstrate cancer, a more conservative biliary bypass procedure may be elected.

Biliary Tract

Intraoperative assessment of the gallbladder is rarely necessary. Laparoscopic ultrasonography has been advocated as an effective method to demonstrate possible anatomic anomalies accurately, in hope of preventing unanticipated surgical complications. [210], [211] IOUS and laparoscopic ultrasonography can effectively evaluate the extrahepatic bile ducts for choledocholithiasis. [212] IOUS can accurately assess tumors of the gallbladder and biliary tract, just as it does tumors of the liver and pancreas. [213] It may be difficult to determine the precise site of duct obstruction intraoperatively by inspection and palpation. IOUS can provide indispensable information regarding tumor resectability and can help determine which type of biliary bypass is feasible. If the central location of the tumor prevents resection and hepaticojejunostomy, IOUS can be used to identify substantially dilated peripheral bile ducts that may be suitable for use in internal drainage.

Laparoscopic Ultrasonography

Laparoscopic ultrasonography has shown promise in evaluation and staging of tumors in the liver, pancreas, biliary tract, stomach, and colon. [214] Laparoscopic staging can identify patients with advanced or metastatic disease, obviating inappropriate lap- arotomies in patients who have limited life expectancy. Further technological improvement will likely lead to more widespread and effective use of laparoscopic ultrasonography in the future.

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CHAPTER 154

Michael B. Kimmey and Peter Vilmann

ENDOSCOPIC ULTRASONOGRAPHY

Gastrointestinal endoscopy has revolutionized the diagnosis and treatment of upper and lower digestive tract mucosal diseases, but endoscopic evaluation of abnormalities of the walls of hollow gastrointestinal organs and their surrounding structures has been limited. Endoscopy may suggest the presence of a mass within or outside the gastrointestinal wall, but it cannot elucidate the character or obtain tissue to allow pathological diagnosis of the mass.

The spatial resolution of extracorporeal imaging techniques has limited the use of those technologies. Transcutaneous ultrasound (US) and computed tomography (CT) cannot differentiate the causes of increased wall thickness or evaluate the depth or localization of a specific abnormality of the gastrointestinal wall. Magnetic resonance imaging (MRI) still requires prolonged imaging times, producing significant motion artifacts that impair resolution.

These deficiencies in conventional imaging techniques prompted the development of endoscopy combined with ultrasound (endoscopic ultrasound or EUS) in the early 1980s in an effort to obtain more information about diseases of the gastrointestinal tract and its surroundings. [1], [2] Further technical advances resulted in the capability of EUS-guided tissue sampling to help differentiate malignant from benign lesions and more recently the provision of EUS-guided therapy. This chapter outlines the principles and indications for EUS and reviews the types of EUS equipment. We review the role of EUS and EUS-guided tissue sampling in the management of patients with digestive diseases, giving particular attention to how EUS compares with other imaging modalities.

H3>TECHNICAL CONSIDERATIONS

Ultrasound Physics and Image Interpretation

Principles of US imaging are outlined in Chapter 153. A few concepts that are pertinent to the combination of US with endoscopy should be emphasized. Increasing the frequency of a US system augments the ability of the US beam to resolve tissue structure, however, tissue penetration of the US beam is reduced with higher frequencies. Placing the US transducer next to the mucosa of the gastrointestinal tract avoids the necessity of having a US beam penetrate far into the tissue and avoids bone and gas-filled organs that reflect US. Higher US frequencies can be used (5 to 30 MHz) to resolve two points as close as 0.2 mm from each other in the direction of the US beam. [3], [4] US waves require the presence of a liquid or soft tissue medium between the US transducer and the target tissue. EUS transducers must be placed directly against the mucosa, or there must be fluid between the transducer and the mucosa. Placing the transducer directly against the mucosa is the simpler of the two alternatives and often is used when extramural structures are being examined. This technique limits examination of the gastrointestinal wall, however, because echoes from structures close to the transducer may not be in the optimal focal zone of the US transducer, and the superficial wall layers may be compressed. [5] By putting water directly into the lumen or into a balloon around the transducer, the mucosa and adjacent structures can be imaged clearly.

Knowledge of what generates echoes within tissue is helpful in interpreting EUS images. Echoes are produced when acoustic energy is reflected back to the transducer. An echo occurs when there is nonhomogeneity within a tissue or when the acoustic wave encounters a change in the acoustic impedance between adjacent tissue types. [6] Tissue inhomogeneities are responsible for most of the echoes coming from a homogeneous organ, such as the liver or pancreas. The microscopic structure of these organs determines the magnitude of the echoes. In many tissues, fat and collagen are responsible for the brightest echoes. For example, a fatty liver is more echogenic than a normal liver. Within the gastrointestinal tract, collagen and fat are found primarily in the submucosa and subserosal or mesenteric fat, probably accounting for the echogenicity of these tissue layers. The overall layered appearance of the gastrointestinal wall is a composite of echoes from the tissue layers and the echoes produced at the boundary or interface between tissue layers.

The US image of the normal gastrointestinal wall usually consists of five layers [6] (Fig. 154-1). However, the number of layers is dependent on the ultrasonic frequencies used. Frequencies from 5 to 12 MHz produce 5 layers while higher frequencies like 20 or 30 MHz depict 7 to 9 layers. Most commonly, there are 3 echogenic layers separated by 2 echo-poor layers (Fig. 154-2). The first echogenic layer, beginning at the mucosal surface, is thin and is produced by the interface between lumenal fluid or the balloon around the transducer and the mucosa. The second layer is echo poor and represents the remainder of the mucosa. This layer was formerly attributed to the muscularis mucosae; however, the normal muscularis mucosae is too thin to account for all of the second layer. [6] The muscularis mucosae usually is obscured by an echo occurring at its interface with the lamina propria so that the location of the muscularis mucosae corresponds to the most superficial part of the third (submucosal) layer. If the muscularis mucosae is thicker than the interface echo, a separate thin hypoechoic layer is seen between this interface echo and the underlying submucosal layer. [7], [8] The third US layer is the easiest layer to recognize because it is the most echogenic. This layer corresponds to the submucosa but is thicker because it also includes the interface between the submucosa and muscularis propria. [6] The fourth layer is echo poor and corresponds to the muscularis propria. In areas with a well-developed inner circular and outer longitudinal muscle component, a small amount of connective tissue between the muscle layers may produce a line of echoes within the muscularis propria. [6], [7] The fifth layer is echogenic but of variable thickness. If no subserosal fat or inflammation is present, this echogenic layer corresponds to the serosa and the interface between the serosa and the surrounding tissue. The layer can be thick in the rectum, where there is often abundant echogenic perirectal fat.

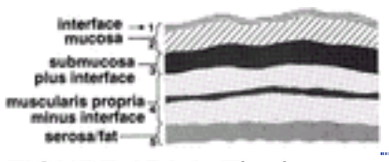

FIGURE 154-1. The layers of the normal gastrointestinal wall as seen on ultrasound images. The ultrasound image comprises echoes arising from the anatomic layers and from the acoustic interfaces between tissue layers.

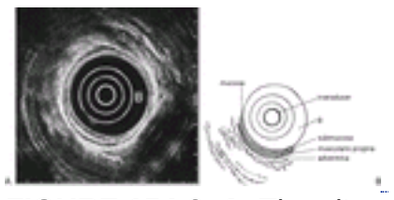

FIGURE 154-2. A: The ultrasound image of the normal gastroesophageal junction made with a 12-MHz mechanical sector scanning ultrasound endoscope. **B:** All the anatomic layers, including mucosa, submucosa, muscularis propria, and adventitia with surrounding structures, are seen in this image made through a water-filled balloon (*B*).

The appearance of organs in the mediastinum and abdomen is roughly similar to that seen with transcorporeal US. The higher resolution of EUS however often provides much greater detail. When the transducer is in the esophagus, the great vessels, parts of the heart, the posterior mediastinum, aortopulmonary window, and subcarinal region can be evaluated. The liver and spleen are homogeneous and of intermediate echogenicity; the normal pancreas has a characteristic “salt and pepper” appearance. The liver usually can be differentiated from the spleen by the bright echoes adjacent to intrahepatic portal venous branches. The kidneys are easily recognized by the characteristic appearance of the urinary collecting system. In most patients, the left adrenal gland can be seen cephalad to the upper pole of the kidney as a seagull-shaped, echo-poor structure. Large arteries and veins in the upper abdomen are key landmarks for orienting EUS images and include the aorta, celiac, and superior mesenteric arteries and the inferior vena cava, portal, splenic, and superior mesenteric veins.

Many efforts have been made to differentiate malignant from benign lymph nodes by analysis of US features such as size, shape, echogenicity, internal echo pattern, and the character of their outer margin. [9], [10] and [11] Malignant lymph nodes are usually echo poor, are similar in echogenicity to the primary neoplasm, and have sharply defined boundaries with adjacent tissue. The internal echoes from the nodes can be homogeneous or inhomogeneous. Benign nodes tend to have poorly defined boundaries with the surrounding tissue and may be hyperechoic. When nodes are over 1 cm in diameter, hypoechoic, and have rounded distinct borders, there is an 80% to 100% probability of malignant involvement. [12], [13] It has become increasingly clear, however, that the diagnosis of a malignant lymph node by EUS image appearance has significant limitations, which has led to the increased use of EUS-guided needle aspiration for pathological confirmation of suspicious nodes. [14], [15]

Instrument Types

US endoscopes can be classified according to the type of endoscope and the type of US system (Table 154-1). The largest worldwide experience has been with the use of a mechanical sector scan on the end of a forward oblique-viewing endoscope. Development of this instrument began in 1979 and has progressed through several generations of improved devices that produce a 360-degree radial scan with US frequencies varying between 5 and 20 MHz. The US transducer at the end of the endoscope is rotated by a motor mounted on the proximal shaft of the endoscope. Optical viewing is achieved through a forward-oblique lens mounted on the side of the endoscope near the transducer. A small biopsy channel allows aspiration or biopsy, but the catheter path does not coincide with the imaging plane, limiting its use for EUS-guided tissue sampling and intervention. [16], [17] Mechanical sector scanning endoscopes have been modified in several ways for special circumstances. A longer forward-viewing variation was developed for use in the colon by omitting a 30-degree sector of the US image to allow passage of fiber bundles to the endoscope tip. [18] Another modification is the removal of viewing optics and the addition of a wire-guided, tapered tip to allow use within tight esophageal strictures. [19]

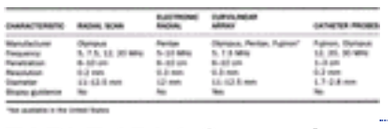

TABLE 154-1 Comparison of Endoscopic Ultrasound Systems

Electronic curved-array US endoscopes are used increasingly as indications for fine-needle aspiration and interventional EUS have been clarified. [17], [20], [21] The US array is mounted along the long axis of the tip of a forward-oblique viewing endoscope. The array of transducer elements is slightly convex to the endoscope, and their signals are processed electronically to produce a 100-degree sector scan that is aligned with the endoscopic image, allowing direct visualization of needles during tissue sampling. Switchable US frequencies of 5 and 7.5 MHz are available. Another advantage of this system is color Doppler capability, which allows the direction of blood flow within vessels to be displayed. [22] This technology is helpful in delineating retroperitoneal and other blood vessels and as an adjunct to guided tissue sampling to avoid unintended puncture of vessels. [14] An electronic radial scanning endoscope has been made. It produces a 270-degree image with a variable frequency between 5 and 10 MHz.

Another method of combining US and endoscopy is to place a US probe through the channel of conventional endoscopes. [23], [24], [25], [26] and [27] These probes contain a US transducer element at the tip of a catheter that rotates or is mechanically moved in a linear direction along the gastrointestinal wall. These probes vary in frequency from 12 to 30 MHz, producing high-resolution images with limited tissue penetration and are most useful for imaging small, endoscopically visible intramural lesions and small mucosal lesions such as early cancer. Placing a water-filled balloon around the transducer may improve image resolution in some situations. [28] Although these systems have been used in the pancreatic and bile ducts, clear indications for intraductal imaging have not yet been established. [29]

Imaging Technique

Detailed descriptions of imaging techniques and the proper positioning of a US endoscope have been published and are beyond the scope of this chapter. [30], [31] Patients are prepared for the procedure as they would be for a standard upper endoscopy. After an overnight fast, topical anesthesia to the oropharynx and intravenous sedation are administered. The US endoscope then is passed blindly through the mouth and into the stomach. The endoscope is directed to the area of clinical interest by the use of a combination of endoscopic views and the US image. Esophageal imaging is accomplished by withdrawing the instrument to the position of interest and imaging performed through the water-filled balloon surrounding the transducer.

The US endoscope is visually directed into the duodenal bulb and descending duodenum for imaging the ampulla of Vater, pancreatic head, distal common bile duct, gallbladder, right lobe of the liver, and retroperitoneal lymph nodes. The water-filled balloon method is used most commonly in the duodenum, although mucosal polypoid structures may be imaged better by placing water in the duodenal lumen. US visualization of the common bile and pancreatic ducts, aorta, inferior vena cava, and portal vein are important landmarks to guide imaging in the duodenum (Fig. 154-3). Imaging from within the stomach also is directed by prior knowledge of the area of clinical interest. Lesions of the gastric wall are identified first by endoscopic vision. The US transducer then is directed over the abnormality. Usually, water is

placed through the endoscope's channel into the lumen for imaging in the stomach.

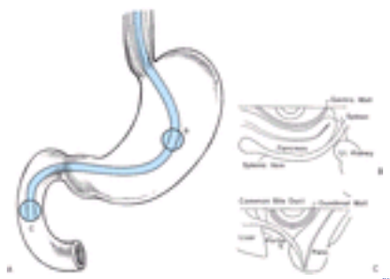

FIGURE 154-3. Position of the ultrasound endoscope (**A**) when imaging from the stomach (**B**) and the duodenum (**C**) is shown. The relative positions of the abdominal organs to the ultrasound transducer when it is in the stomach and the duodenum are also diagrammed.

The body of the pancreas is imaged through the antrum and body of the stomach. The pancreatic tail can be imaged through the fundus of the stomach. Pancreatic imaging is aided by identifying the superior mesenteric artery and vein and the splenic vein posterior to the pancreas and by following the course of the pancreatic duct (see Fig. 154-3). The spleen, left adrenal, left kidney, and left lobe of the liver also can be imaged through the gastric fundus.

Transrectal US is best performed after full colonic purge to avoid artifacts from retained feces. The US endoscope is inserted into the rectum and guided with endoscopic vision into the sigmoid colon or to the level of an obstructing neoplasm. Imaging is accomplished through a water-filled balloon as the probe is withdrawn. Areas of particular interest within the rectum also can be scanned by infusing water into the lumen. Use of a curvilinear array instrument will often allow passage to the splenic flexure. [32]

Optimal performance of EUS requires specific training and substantial experience. The degree of difficulty of EUS is comparable to that of therapeutic endoscopic retrograde cholangiopancreatography (ERCP). [33] Attending EUS courses and studying cross-sectional anatomy and EUS atlases are helpful but should be supplemented by supervised, hands-on training. [33]

Fine-Needle Aspiration

Since EUS imaging is not equivalent to histological analysis, tissue sampling is being increasingly used to confirm the diagnosis. Because EUS can visualize even minute lesions that are not visible with other imaging modalities, the combination of EUS and guided biopsy has the potential to obtain conclusive diagnostic information from such lesions. It is also advantageous to obtain tissue from lesions that are visible with other imaging modalities but are located in areas that are not readily accessible. Several publications have demonstrated that EUS-guided biopsy from imaged lesions is possible in most cases. [34], [35], [36], [37], [38], [39], [40] and [41] Both lesions in the gastrointestinal wall itself as well as lesions lying adjacent to the wall may be punctured through the esophagus, stomach, duodenum, or rectum.

Detailed descriptions of EUS-guided fine-needle aspiration (FNA) technique have been described in several publications. [38], [39], [42], [43] Several different aspiration needles are commercially available including reusable and disposable varieties. A full-length steel needle with a stylet is recommended to provide sufficient stiffness to penetrate hard lesions. After the target is outlined ultrasonically by direct transducer contact with the gastrointestinal wall, the needle with the stylet is introduced through the biopsy channel of the endoscope. The needle then is advanced out of the housing catheter so that reflections from the needle tip are seen ultrasonically.

The stylet then is withdrawn a few millimeters, and the needle is pushed into the lesion under ultrasonic guidance before the stylet is removed completely. A vacuum is applied to the needle by means of a 10-mL syringe, and the needle is advanced further into the lesion and moved back and forth under ultrasonic guidance. [44] The vacuum is equilibrated while the tip of the needle is in the lesion. An alternate technique for sampling lymph nodes without suction has been reported to provide adequate diagnostic material that is less bloody. [15] The needle finally is removed, and the aspirated material is expelled and smeared onto glass slides. The aspiration is repeated until sufficient material is obtained; the presence of a cytopathologist is helpful in making this differentiation. [40] Between two and five passes usually are required to obtain an adequate specimen.

Normally, the aspiration procedure lasts 5 to 20 minutes. The time used depends on the location of the puncture target. FNA from the esophagus is usually fast, whereas FNA of hard lesions in the head of the pancreas may require more time. Patient discomfort in connection with the procedure is limited to that experienced as a result of the EUS examination itself. The overall complication rate for FNA of solid masses (e.g., bleeding, infection, perforation) is less than 1%. [40] Infections following aspiration of cystic lesions occur in up to 14% of cases when prophylactic antibiotics are not given. [40] Antibiotics are not used routinely but should be considered when performing transrectal biopsy and when aspirating cystic lesions. [45] Major bleeding, pancreatitis, and air embolism are rare events. [17], [46], [47] Tumor seeding of a needle tract is a theoretical complication, but has not yet been described following EUS-guided FNA.

The indications for EUS-guided biopsy continue to evolve. The procedure should be considered in patients when lesions suspicious for malignancy are detected on conventional imaging modalities that are within or adjacent to the gastrointestinal tract. [39], [40] Also, lesions suggestive of malignancy that are detected only by US and not by conventional modalities may be biopsied if an aspirate positive for malignancy could change patient management. Indications for EUS-guided FNA are listed in Table 154-2.

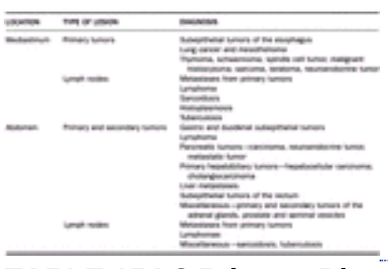

TABLE 154-2 Primary Diagnoses That May Be Obtained by Endoscopic Ultrasound–Fine-Needle Aspiration (EUS-FNA)

ENDOSCOPIC ULTRASONOGRAPHY OF THE UPPER GASTROINTESTINAL TRACT

Focal Intramural and Extramural Mass Lesions

Focal abnormalities of the upper gastrointestinal wall often are suspected based on the results of radiographic and endoscopic examinations. Contrast radiographs may reveal a smooth, rounded defect impinging on the lumen, or the endoscopist may encounter a bulge covered with normal mucosa. The clinical significance of these masses can be difficult to ascertain but may be facilitated by knowing whether the mass is intramural or extramural and whether it is benign or malignant. EUS can be useful in defining the location of the mass and can direct FNA to determine whether it is malignant.

Intramural masses arise from a well-defined US layer that corresponds to one of the histological layers of the gastrointestinal wall. [48], [49] Extramural masses may compress the outer echogenic layer of the gut wall or, if malignant, may disrupt normal layer structure. In a series of 139 patients with suspected intramural or extramural masses of the upper gastrointestinal tract, EUS correctly identified the mass as intramural (80 cases) or extramural (59 cases) in every patient. [48]

Gastrointestinal stromal tumors (GIST) are the most commonly encountered focal intramural masses in the upper gastrointestinal tract. They are differentiated ultrasonographically by their presence within the fourth US layer, corresponding to the muscularis propria. They are hypoechoic and usually expand the hypoechoic fourth layer but do not disrupt the boundaries between the third and fourth or fourth and fifth layers (Fig. 154-4). Occasionally, GISTs are seen as well-marginated hypoechoic masses between the second and third layers. In these instances, the tumor probably has arisen from the muscularis mucosae. Most small GISTs are round and homogenous. Although EUS cannot distinguish reliably between benign and malignant GISTs, features that suggest malignancy include diameter greater than 3 cm, irregular outer borders, central ulceration, and the presence of enlarged regional lymph nodes [50], [51] and [52] (Fig. 154-5). Other causes of focal intramural lesions can be imaged with EUS. Duplication cysts are identified most commonly as smooth, anechoic masses within the distal esophagus, but they can occur anywhere in the gastrointestinal tract. [53] Esophageal and gastric carcinoid tumors cause well-defined hypoechoic nodules in the second or third US layers. Lipomas are differentiated by being echogenic and clearly marginated and usually are located in the third (submucosal) layer. [48] Ectopic pancreas, submucosal cysts (Fig. 154-6), gastric duplications, granular cell myoblastoma, metastatic cancer, and eosinophilic granuloma also have been imaged with EUS. [48], [49] Endometriomas may be imaged within or adjacent to the colon and rectum.

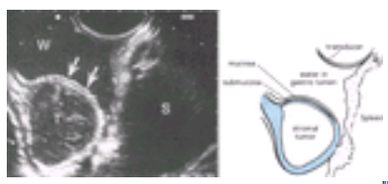

FIGURE 154-4. A gastric stromal tumor (*T*), formerly called a leiomyoma, is imaged through water (*W*) in the gastric lumen using a curvilinear array ultrasound endoscope. The overlying mucosal and submucosal layers (*arrows*) show that this tumor is contiguous with the muscularis propria layer. The spleen (*S*) is also seen adjacent to the stomach wall.

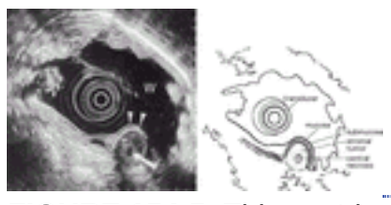

FIGURE 154-5. This gastric stromal tumor (*T*) imaged with a radial scanning echoendoscope through water (*W*) in the gastric lumen is covered by mucosa and submucosa (*arrowheads*) and has a central hypoechoic area (*arrow*), probably due to necrosis.

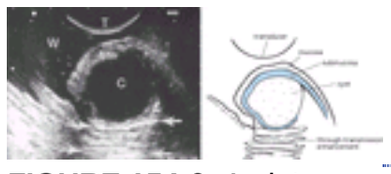

FIGURE 154-6. An intramural cyst (*C*) is imaged with a curvilinear array echoendoscope after putting water (*W*) in the gastric lumen. There is normal overlying mucosa (*m*) and submucosa (*sm*). Note the enhancement of echoes from the area underneath the cyst (*between the arrows*). This phenomenon (termed *through transmission*) occurs because the fluid-filled cyst causes less impedance to the ultrasound beam than does the surrounding tissue.

The origins of extramural mass lesions often can be deduced by identification of contiguous structures. When enlarged or of normal size, the aorta, liver, gallbladder, spleen, and splenic artery or vein may compress the gastrointestinal tract. [54] Hepatic cysts and pancreatic cystic neoplasms or pseudocysts also are identified by their surrounding organs. [55] Malignant extramural lesions such as bronchogenic carcinoma, hepatic and pancreatic malignancies, and malignant lymph nodes may compress or invade the outer layers of the gastrointestinal wall.

EUS-guided biopsy of lesions within the gastrointestinal wall have a lower diagnostic value and higher rate of inconclusive biopsies than EUS-guided biopsy of extramural lesions. [40], [56] In a multicenter study of 554 lesions taken by biopsy in 457 patients, 115 lesions were located within the gastrointestinal wall. [40] The accuracy of EUS-guided FNA cytology of 12 submucosal tumors was 50%, with one half of the biopsy specimens being inadequate. In cases of GISTs, although cytology demonstrated spindle cells in four of seven cases, the single case of a malignant GIST was not diagnosed. EUS FNA was performed in 103 lesions arising in the gastrointestinal wall, exclusive of stromal tumors with a sensitivity, specificity, positive predictive value (PPV), negative predictive value (NPV), and accuracy of 61%, 79%, 100%, 76%, and 67%, respectively.

Esophageal Carcinoma

The diagnosis of esophageal carcinoma is usually not difficult with esophagoscopy and biopsy. Staging this tumor to plan appropriate therapy is more difficult. Endoscopy or radiography defines the longitudinal extent of tumor growth but does not define depth of invasion. CT and EUS have been used to evaluate the depth of invasion into the esophageal wall, to detect spread to mediastinal and celiac lymph nodes, and to detect invasion of other mediastinal and thoracic structures. EUS is the only imaging modality that can differentiate among mucosal, submucosal, and muscular involvement with esophageal carcinoma, and it can accurately determine the depth of invasion in more than 80% of early cases [57], [58], [59], [60], [61] and [62] (Table 154-3). Most patients have spread of esophageal carcinoma outside the muscularis propria at presentation. EUS and CT accurately detect about 90% of cases with mediastinal invasion. [57], [58], [59], [60], [61] and [62] EUS is more sensitive than CT in detecting aortic (Fig. 154-7), pericardial, and diaphragmatic involvement with carcinoma. [57], [58], [59], [60], [61], [62] and [63] The dynamic nature of EUS, with the ability to detect moving structures in real time, may contribute to the accuracy of this technique in detecting invasion of these viscera. Patients with extraesophageal organ involvement with cancer (T4 stage) detected by EUS have similar survival rates when treated surgically or by nonoperative methods. [63]

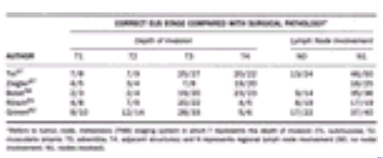

TABLE 154-3 Endoscopic Ultrasound Staging of Esophageal Carcinoma

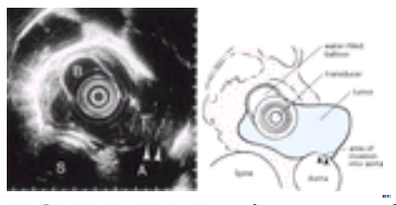

FIGURE 154-7. A large esophageal cancer (*T*) invades the tissue around the esophagus and invades the descending aorta (*A*) at the *arrows.* The thoracic spine (*S*) is posterior to the mass. Imaging has been aided by filling a balloon around the radial scanning transducer with water (*B*).

EUS is more accurate than CT in detecting lymph node involvement in patients with esophageal cancer. [57], [58], [59], [60], [61], [62] and [63] The limitations of CT in detecting enlarged mediastinal lymph nodes in patients with esophageal cancer are well known; about 50% of cases of esophageal cancer are understaged by CT. Lymph nodes as small as 5 mm in diameter can be visualized with EUS. The accuracy of EUS in detecting mediastinal lymph node involvement is about 70% to 80% (see Table 154-3). [57], [58], [59], [60], [61] and [62] Limitations of EUS in detecting lymph node involvement include false-positive results caused by enlarged inflammatory nodes and, to a lesser degree, false-negative results caused by microscopic metastases in normal-sized nodes. CT correctly identifies the presence or absence of mediastinal lymph node involvement in only 20% to 50% of cases, with good specificity but limited sensitivity. [57], [58], [59], [60], [61] and [62] CT and mediastinoscopy, however, can detect some groups of lymph nodes that are not imaged with EUS, such as those separated from the esophagus by the lung or trachea. The number of abnormal lymph nodes detected by EUS correlates with 5-year survival following esophagectomy. [64]

EUS-guided biopsy of lymph nodes should be used whenever possible to confirm the presence of malignant involvement. [65], [66] Avoidance of the primary tumor in the needle path is important to avoid contaminating material from the node with the primary neoplasm. Confirmation of celiac lymph node involvement is especially important as these represent metastases in patients with middle and upper esophageal cancers. [66] EUS-FNA of lymph nodes in esophageal cancer has been reported to change management in 60% of patients. [65] The sensitivity, specificity, PPV, and NPV of EUS-FNA for the detection of malignancy in lymph nodes in a large multicenter study was 93%, 100%, 56%, and 92%, respectively. [40]

EUS and CT are complementary in the detection of distant metastases from esophageal carcinoma. EUS has a sensitivity of greater than 90% in detecting celiac lymph node metastases when it is possible to pass the endoscope through the malignant stricture. [57] In comparing the overall accuracy of EUS and CT, the accuracy of both modalities is about 70% to 80%, because the US endoscope cannot pass through 20% to 50% of malignant strictures. [57], [58], [59], [60], [61] and [62], [67], [68] and [69] The ability to pass through malignant esophageal strictures however has been improved with the development of smaller diameter US endoscopes with more tapered tips. [66] CT is also more sensitive than EUS in detecting hepatic metastases, especially in the right lobe.

EUS is more accurate than CT in predicting nonresectability of esophageal cancer: 91% for EUS versus 39% for CT. [70] Prediction of resectability by EUS may be higher for adenocarcinoma than for squamous cell carcinoma because of the difficulty in recognizing microscopic submucosal spread of disease in the latter. Detection of local anastomotic recurrence by EUS is sensitive but relatively nonspecific when only thickening is used as a criterion; the false-positive rate is 5% to 20%. [71]

The role of EUS before and after neoadjuvant chemoradiation is evolving. Imaging alone does not distinguish between residual tumor and scar following chemoradiation so imaging findings should be confirmed with FNA whenever possible. [72] The presence of celiac adenopathy and T4 stage on EUS imaging should not preclude the patient receiving preoperative chemoradiation since tumor can be sterilized from these locations in some patients. [73] A reduction of the maximum cross-sectional area of the primary tumor measured with radial EUS imaging before and following chemoradiation has been reported to correlate with survival following surgery. [74], [75]

Gastric Carcinoma

The greatest experience with EUS in the staging of gastric cancer comes from Japan, where the incidence of this tumor is high. EUS has the potential for detecting the depth of invasion of gastric neoplasms because of the clear layers seen on US images of the gastric wall. Because gastric cancer confined to the mucosa and submucosa has a better outcome with surgical resection than does more advanced disease, EUS can contribute significantly to the management of patients who have early gastric cancer. For example, EUS findings are used to select candidates for endoscopic mucosal resection. [76] The depth of invasion is seen as a disruption in the normal layer structure or diffuse thickening of the normal layers by infiltrating scirrhous carcinomas. [76], [77], [78] and [79] EUS accurately predicts the depth of malignant invasion into the gastric wall in about 80% of patients [61], [62], [77], [78] and [79] (Table 154-4). Accuracy rates of over 90% are achieved in differentiating early (i.e., confined to the mucosa or submucosa) from advanced (i.e., invasion of the muscularis propria or deeper) gastric cancer (Fig. 154-8 and Fig. 154-9). Cell type and the degree of differentiation do not significantly affect the staging accuracy of EUS. The depth of invasion may be overestimated if there is an ulcer scar or inflammatory reaction below the cancer. [77] Underestimation of the depth of invasion occurs when microscopic invasion of deeper layers occurs or the serosal interface echo cannot be well seen.

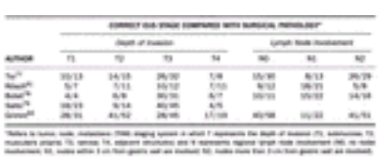

TABLE 154-4 Endoscopic Ultrasound (EUS) Staging of Gastric Carcinoma

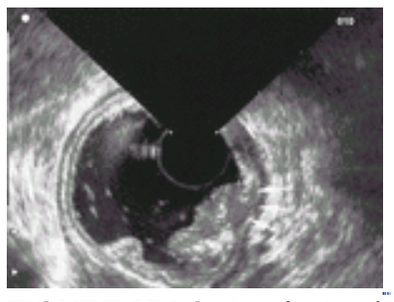

FIGURE 154-8. An electronic radial scanning endoscope has been used to image this early gastric cancer. The cancer invades the submucosa (*arrows*) making this a T1 tumor.

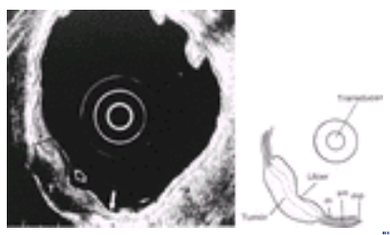

FIGURE 154-9. This advanced gastric cancer has raised borders and a central ulceration (*open arrow*). The tumor mass (*T*) arises from the normal wall (*arrow*), where layers corresponding to mucosa (*m*), submucosa (*sm*), and muscularis propria (*mp*) can be seen. The cancer invades the fat and other tissue around the gastric wall.

The accuracy of EUS in detecting gastric cancer in extraintestinal lymph nodes is less well established. About 75% to 80% of malignant regional lymph nodes are detected by EUS, but false-positive results are seen in as many as 50% of benign nodes. [9], [78], [80] False-negative results occur most commonly in cases of microscopic metastases in normal-sized nodes. The role of EUS-FNA in patients with gastric cancer is similar to that discussed previously for esophageal cancer. [40], [56], [81] EUS can stage the depth of invasion and detect lymph node metastases more accurately than high-quality CT scanning. [78] Compared with pathological evaluation of the resected specimen, EUS correctly predicts depth of invasion and lymph node involvement in 78% to 91% and 50% to 78% of cases, respectively. [77], [78] CT scanning correctly predicted depth of invasion and lymph node involvement in 42% and 48% of cases, respectively. [78] Overall staging was significantly more accurate with EUS (73% correct) than with CT (45% correct). [78]

The invasion of gastric cancer into surrounding structures may be detected with EUS, and malignant involvement of the pancreas and liver has been described. [77] The accuracy of predicting resectability in gastric cancer is higher than CT, reaching about 95%. Because of the limited penetration of EUS, however, CT scans are necessary for the detection of most hepatic metastases. [77], [78]

Gastric Lymphoma

Lymphomas involving the stomach can present as an infiltrating or polypoid mass lesion or as a gastric ulcer. [82], [83] and [84] Endoscopic biopsy may reveal the diagnosis, but biopsies may be nondiagnostic when the bulk of the tumor is below the mucosa. EUS can detect the extent of lymphomatous involvement of the gastric wall in infiltrating and ulcerative gastric lymphomas [83], [84] (Fig. 154-10). The depth of wall invasion, involvement of surrounding organs such as the pancreas or liver, and the presence of enlarged lymph nodes can be assessed by EUS. Reduction of tumor size after chemotherapy also has been documented by EUS imaging. [83], [84] EUS appears to be more sensitive than CT scanning in detecting the presence of intramural invasion, extragastric spread, and regional lymph node involvement.

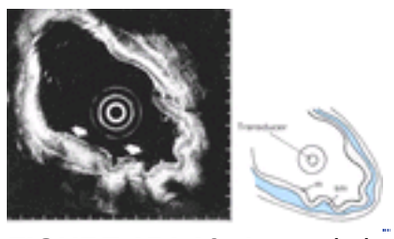

FIGURE 154-10. A gastric lymphoma has caused thickening of the ultrasound layers corresponding to mucosa (*m*) and submucosa (*sm*) in the region under the *arrows.*

Preliminary studies suggest that EUS can help in the decision of whether to treat gastric mucosa-associated lymphoid tissue (MALT) lymphoma with antibiotics for *Helicobacter pylori* infection or systemic chemotherapy; patients with thickening confined to the mucosa and submucosa on EUS usually respond to antibiotics alone. [85], [86]

Ampullary Carcinoma

Carcinoma of the ampulla of Vater occurs infrequently compared with pancreatic cancer, but it is important to recognize the tumor because patients often present with jaundice when the tumor is resectable. Five-year survival after surgical resection of ampullary cancer can be expected for about 40% of patients. Surgery is radical and carries a mortality rate of up to 10%. It would be useful to have preoperative assessment of those patients most likely to benefit from the surgery. EUS offers more accurate staging than other imaging studies in the preoperative staging of these tumors. [87], [88], [89], [90], [91], [92] and [93]

EUS detects the normal five layers of the duodenal wall adjacent to the papilla. The common bile duct and pancreatic duct converge at the papilla, with the portal vein lying immediately adjacent to the bile duct. EUS can detect tumor masses as small as 8 mm in diameter within the papilla. EUS also correctly predicts the depth of invasion of the tumor with respect to involvement of the muscularis propria or subserosa in as many as 80% of patients. [87], [88], [89], [90], [91], [92] and [93] Invasion of the pancreatic parenchyma and the existence of lymph node metastases also can be detected with EUS. The limitations of EUS in the staging of ampullary carcinoma include failure to detect very small tumors, failure to visualize liver metastases, and false-positive interpretation of inflammatory lymph nodes. [87], [88], [89], [90], [91], [92] and [93] If therapeutic plans are changed by the presence of malignant lymph nodes, EUS-FNA should be used to confirm the presence of malignancy. [94]

Most ampullary neoplasms can be diagnosed accurately by a combination of endoscopy with biopsy and ERCP. The role of EUS is in demonstrating the size and depth of invasion of the tumor, the presence of involved regional lymph nodes, and the presence of portal or superior mesenteric vein invasion. [87], [88], [89], [90], [91], [92] and [93] Larger studies with long-term follow-up are needed to determine whether the results of EUS imaging predict which patients will have long-term survival after surgical resection.

Upper Gastrointestinal Wall Thickening

EUS can detect a thickened gastrointestinal wall. The upper limit of normal wall thickness has not been defined rigorously but is probably about 3 to 4 mm. The degree of lumenal distention and balloon compression can affect layer visualization and wall-thickness measurements. [5] There are several causes of gut wall thickening, some of which can be differentiated by using EUS imaging. The first two US layers, corresponding to the mucosa, are thickened in severe esophagitis and gastritis, superficially growing MALT lymphomas, foveolar hyperplasia, Ménétrier disease, Kaposi sarcoma, and gastrinoma. The fourth layer, corresponding to the muscularis propria, may be thickened in response to distal obstruction. [6] All four layers may be thickened in the stomach adjacent to an ulcer or with gastric lymphoma or linitis plastica. [83] A few studies have addressed the role of EUS in the diagnostic approach to large gastric folds. EUS has a nearly 100% specificity but a lower sensitivity for detecting malignancy. [95], [96]

Portal Hypertension

The course and mural location of esophageal and gastric varices can be determined by EUS. Varices are imaged within the submucosa and in the periesophageal or perigastric soft tissue. [97] Small submucosal varices are easily compressed and may be missed when imaging with the water-filled balloon method. After sclerotherapy, submucosal varices are obliterated, but periesophageal collaterals may not be affected. [98], [99] EUS detects gastric varices and may prove useful in differentiating gastric varices from large rugal folds. Large extragastric venous collaterals in patients with portal hypertension can also be detected by EUS.

Peptic Ulcer Disease

The depth of mucosal lesions may be difficult to gauge by routine endoscopic examination but can be examined with EUS. The second US layer is thickened by edema around erosions, but the third layer is not penetrated. Ulcers may extend into the third or fourth layer or completely into the subserosal region. The gastric wall may be diffusely thickened in the region around an ulcer, and inflammatory changes may obscure the normal layered appearance of the surrounding wall. EUS also may aid in differentiating benign from malignant gastric ulcers, but false-positive and false-negative results limit its use; mucosal biopsy is always recommended to exclude malignancy.

ENDOSCOPIC ULTRASONOGRAPHY OF ORGANS ADJACENT TO THE UPPER GASTROINTESTINAL TRACT

Lung Cancer

EUS can detect and, with FNA, provide a tissue diagnosis in patients with lung cancer. Primary tumors adjacent to the esophagus can be best approached via the transesophageal route which can provide a tissue diagnosis even when bronchoscopic methods fail. [100], [101], [102], [103] and [104]

An emerging role for EUS-guided FNA is in the staging of non–small-cell lung cancer. [105], [106], [107] and [108] The presence of cytologically confirmed malignancy in subcarinal and contralateral mediastinal lymph nodes precludes curative surgery and therefore reduces costs associated with mediastinoscopy and thoracotomy [108], [109] (Fig. 154-11). A Japanese study of 132 lung cancer patients demonstrated a sensitivity of 54% and a specificity of 98% for EUS in the evaluation of mediastinal lymph node metastases. [105] The sensitivity increased to 81% when right superior mediastinal lymph nodes (lymph nodes located anterior to the trachea) were excluded from calculations. Another study demonstrated an accuracy of lymph node evaluation by EUS of 72% but a sensitivity of only 67%. The lower sensitivity was explained by the high proportion of patients with anthracosilicosis. [108] In the latter study, CT demonstrated only 47% of the lymph nodes, and 67% of these lymph nodes were malignant. EUS is regarded as more accurate than CT in preoperative staging of lung cancer, especially when imaging results are confirmed by FNA.

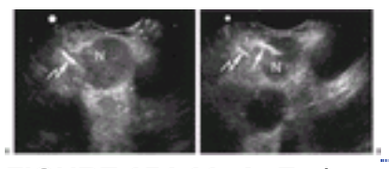

FIGURE 154-11. A: Endoscopic ultrasound (EUS) from the esophageal lumen identified an 18-mm subcarinal lymph node (*N*) in a patient with non–small-cell lung cancer. The trachea (*curved arrow*) is seen at the left of the image. **B:** EUS-guided fine-needle aspiration (*straight arrow*) obtained malignant cells.

EUS-FNA is complementary to mediastinoscopy for staging non–small-cell lung cancer. If enlarged lymph nodes in areas adjacent to the esophagus are imaged on CT, EUS-FNA is more cost-effective than mediastinoscopy in confirming malignancy within the nodes. [109], [110]

Other Thoracic Extraesophageal Structures

EUS may detect other mediastinal or pulmonary abnormalities as incidental findings or because they compress the esophagus. EUS can reveal the cause of these compressions and exclude a primary esophageal lesion. Causes of esophageal compression other than lung cancer that have been detected by EUS include mediastinal cyst, bronchogenic cyst, aberrant right subclavian artery, thoracic aortic aneurysm, teratomas, and mediastinal pseudocysts. Hodgkin disease, tuberculosis, histoplasmosis, sarcoidosis, metastatic cancer, and neuroendocrine and neurogenic tumors can also be diagnosed by EUS-guided-biopsy. [111], [112] and [113]

Acute Pancreatitis

EUS has been used in the setting of suspected gallstone pancreatitis and is more sensitive than transabdominal US for the detection of common bile duct stones in this setting. [114] EUS can also be useful in defining the cause of unexplained acute pancreatitis. Microlithiasis, occult common bile duct stones, small pancreatic neoplasms, and pancreas divisum can be found in up to 75% of this population. [115], [116] Due to the lower risk of complications, EUS should be considered as an alternative to ERCP in this group of patients.

Chronic Pancreatitis

The diagnosis of chronic pancreatitis is not difficult in the patient with steatorrhea, glucose intolerance, and pancreatic calcification on plain abdominal radiographic films. Early cases, however, may be more subtle, defying diagnosis using conventional diagnostic tests. EUS has been proposed as a sensitive, specific imaging tool for the diagnosis of chronic pancreatitis. High-resolution US images of the pancreas can be made through the posterior gastric wall and through the medial wall of the duodenum. Because there is no intervening intestinal gas, EUS is able to image the entire pancreas in most cases; the head and tail of the pancreas are seen more often with EUS than with transcutaneous US. The normal pancreas has a homogeneous, fine granular appearance when imaged with EUS. The pancreatic duct has clearly defined walls with an overall diameter of 2 to 3 mm. Endosonographic features of chronic pancreatitis include ductal dilation with or without calculi, echogenic duct walls, pseudocysts, and a heterogeneous pancreatic parenchyma with focal hypoechoic and other echogenic areas [117], [118] and [119] (Fig. 154-12). Early chronic pancreatitis may be diagnosed by EUS at a stage when other modalities are unable to demonstrate any structural signs of disease. However, the specificity of these findings is uncertain since many of the EUS features of chronic pancreatitis can be seen in alcoholics without clinical evidence of pancreatitis. [120]

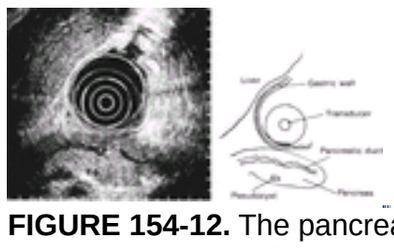

FIGURE 154-12. The pancreas of a patient with chronic pancreatitis is imaged through the posterior gastric wall with a 7.5-MHz ultrasound endoscope. A dilated pancreatic duct (*pd*) and a small pancreatic pseudocyst (*c*) are seen as well as a heterogeneous echo texture within the pancreatic parenchyma.

Mass lesions within a chronically inflamed pancreas may be inflammatory or malignant. It may be impossible to differentiate between these possibilities with transcutaneous US, CT, and ERCP. Although evaluation of EUS with rigorous, blinded, histologically confirmed studies is not available, there are endosonographic features that may help to differentiate between these causes of a pancreatic mass. [121], [122] Inflammatory lesions are favored by the presence of a homogeneous hypoechoic mass with smooth boundaries. Malignant lesions are suggested when a heterogeneous mass with irregular contours, often in a lobulated pattern, is imaged. The sensitivity and specificity of these imaging findings however are not sufficient to differentiate reliably between malignancy and focal chronic pancreatitis; therefore EUS-guided biopsy should be used whenever possible to confirm the presence of suspected malignancy.

Pancreatic Carcinoma

One of the first targets for EUS imaging was pancreatic cancer. [121] The early detection of this usually lethal neoplasm has eluded other diagnostic modalities. The increased spatial resolution of EUS may allow detection of smaller, resectable neoplasms in a few patients whose symptoms are vague. [121] More recently, the use of

EUS to detect premalignant changes in members of pancreatic cancer families has also been described. [123]

EUS has been used to image malignant pancreatic masses, to investigate whether malignant lymph nodes are present, and to detect the presence of vascular invasion that would preclude surgical resection. [41], [87], [88], [121], [124], [125] and [126] EUS is more sensitive than transcutaneous US, axial CT, and angiography in detecting masses smaller than 3 cm in diameter, and it is more accurate than the other imaging modalities in assessing tumor size. Masses as small as 10 mm in diameter can be imaged by EUS. [121] Most pancreatic neoplasms are hypoechoic, but a few may be hyperechoic on EUS imaging. EUS can also detect cystic pancreatic neoplasms and metastases to the pancreas. [55], [127], [128] Enlargement of lymph nodes around the porta hepatis, aorta, celiac trunk, and splenic artery can be detected by EUS. Accurate prediction of lymph node status in 70% to 80% of patients undergoing surgery has been reported. [41], [87], [88] EUS is more sensitive than CT in detecting regional lymph node involvement; [88] however, because of false-positive lymph node diagnoses in up to 50% of cases by EUS imaging, malignant involvement should be confirmed with FNA if therapeutic management might be affected. [41], [94], [129], [130] and [131] Approximately 30% of patients with pancreatic carcinoma undergoing EUS will be found to have FNA-positive malignant lymph nodes. [94]

Major vessel invasion by a pancreatic neoplasm precludes curative resection. Morbidity and mortality resulting from pancreaticoduodenectomy could be prevented in many patients if a reliable method of detecting vascular invasion were available. EUS can detect portal vein and splenic vein involvement by pancreatic neoplasms [87], [88], [126] (Fig. 154-13). EUS is more sensitive than angiography and axial CT for detecting neoplastic involvement of the portal vein, including the region of its confluence with the superior mesenteric vein. [88] The accuracy of locoregional cancer staging with newer helical CT scanners may be comparable to staging with EUS. [92], [93], [132], [133] and [134] EUS is not as reliable for detecting superior mesenteric vein invasion or major arterial (e.g., celiac, hepatic, splenic, superior mesenteric) encasement by tumor. [88], [135] The radial and curvilinear array instruments appear to be comparable in staging accuracy for pancreatic cancer. [136] EUS-FNA is increasingly used to obtain a tissue diagnosis in patients with pancreatic cancer (Fig. 154-14). The diagnostic values of FNA for pancreatic masses in a large multicenter study were a sensitivity of 86%, a specificity of 94%, a PPV of 100%, an NPV of 86%, and an accuracy of 88%. [40] Other studies have also described the use of EUS-guided FNA in managing patients with pancreatic cancer. [41], [56], [126], [129], [130] and [131], [137], [138], [139], [140], [141] and [142] The detection of unresectable masses, malignant nodes outside the area of planned resection, and liver metastases avoid the morbidity and cost of major surgery. [130], [131]

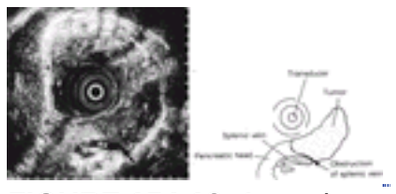

FIGURE 154-13. A carcinoma of the body of the pancreas (*T*) is shown (*arrow*) to obstruct the splenic vein (*sv*) in this transgastric endoscopic ultrasonographic image.

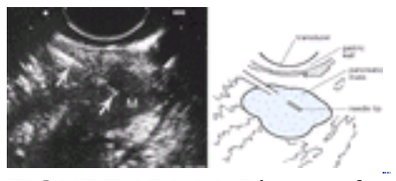

FIGURE 154-14. Biopsy of a pancreatic mass (*M*) with a needle (*arrows*) passed through the stomach wall into the mass under ultrasound guidance.

Islet Cell Neoplasms

The anatomic localization of hormone-producing pancreatic tumors is important for guiding their surgical resection. Most of these neoplasms arise from islet cells within the pancreas, although a significant number are found elsewhere in the abdomen. Conventional transcorporeal imaging with US and CT detects fewer than one third of cases, probably because of the small size of the tumors. MRI and selective arteriography also may fail to localize the neoplasm. Careful examination of the pancreas with EUS by experienced endosonographers detects about 75% of islet cell neoplasms within the pancreas not found on CT scanning. [143], [144] and [145] Islet cell neoplasms are usually round, homogeneous, and hypoechoic compared with the surrounding pancreas. Masses as small as 5 mm in diameter can be imaged by EUS. [143], [144] and [145] All patients who have a clinical diagnosis of an islet cell neoplasm that has not been localized with other imaging techniques should undergo preoperative EUS to localize the neoplasm and facilitate surgical planning. Octreotide scanning and EUS appear to be complementary in the preoperative localization of gastrinomas. [146] Although EUS-guided FNA can confirm the diagnosis of an islet cell neoplasm, this is not required in the setting of a well-defined clinical syndrome and documented excessive hormone secretion.

Gallbladder and Bile Ducts

The close proximity of the common bile duct and gallbladder to the duodenum and distal stomach allows EUS imaging of these organs. The detection of choledocholithiasis and the characterization and staging of bile duct and gallbladder neoplasms have been studied by EUS.

Transcutaneous US detects more than 95% of gallbladder stones but detects only approximately 30% of common duct stones. EUS may detect stones in the bile duct by imaging from the descending duodenum or duodenal bulb. [147], [148], [149] and [150] Common bile duct stones can be identified when EUS is used to evaluate the patient with a dilated bile duct or jaundice of unknown cause. If a common duct stone is suspected from transcutaneous imaging procedures or on clinical grounds, ERCP should be undertaken rather than EUS because it allows removal of the stone; however, EUS has comparable sensitivity to that of ERCP in detecting bile duct stones and represents a noninvasive diagnostic test without the risk of pancreatitis that accompanies ERCP. [114], [148], [149] and [150] Intraductal catheter probes can also aid in the detection of small stones missed by cholangiography. [151], [152] A French study of patients with choledocholithiasis demonstrated a sensitivity for stone detection of 97% for EUS compared with 25% for abdominal ultrasonography and 75% for CT. [149] Preliminary studies also demonstrated that microlithiasis may be diagnosed by EUS with a sensitivity as high as 95% in patients with negative abdominal US but in whom there is a strong clinical suspicion of gallstone disease. [153], [154]

EUS has been used to stage carcinoma of the bile duct and gallbladder. As with pancreatic carcinoma, the detection of major portal venous invasion by the neoplasm is probably the most clinically relevant role for EUS. Small-diameter catheter probes placed in the bile duct at percutaneous transhepatic cholangiography or ERCP also may prove useful for this indication. [29], [155] EUS-guided FNA can also facilitate obtaining a tissue diagnosis in the majority of patients with hilar cholangiocarcinoma. [156] Prospective studies are needed to define the roles of EUS, CT, and angiography in staging bile duct and gallbladder neoplasms.

Liver

EUS has not been used extensively in imaging the liver. Although portions of the left lobe are imaged from the stomach and part of the right lobe from the stomach and duodenum, inadequate penetration prevents imaging of the entire liver. Transcutaneous US and CT provide a more complete examination of the liver than does EUS, although no prospective comparisons of these modalities in the detection of focal liver abnormalities are available. The better resolution of EUS does allow detection of small metastases in parts of the liver immediately adjacent to the stomach and duodenum that cannot be imaged with extracorporeal imaging techniques. [157], [158] EUS-FNA of these metastases has yielded a tissue diagnosis in nearly all cases and often leads to a change in patient management. [40], [157], [158]

TRANSRECTAL ULTRASONOGRAPHY

US imaging from within the rectal lumen has been applied to the diagnosis of benign and malignant diseases of the rectum. The diagnosis of inflammatory bowel disease and its complications has been investigated in a few studies, but most experience has been accumulated in the staging of colorectal neoplasms. Most of these imaging studies have been performed without the use of endoscopic viewing, but the same examination can be done using a US endoscope with similar results.

Inflammatory Bowel Disease

Transcutaneous ultrasonography has been used to detect wall thickening and define the location and extent of intestinal involvement in patients with Crohn's disease and ulcerative colitis. [159] An in vitro study, however, showed that changes in the US layers of the intestinal wall do not reliably differentiate ulcerative colitis from Crohn's disease; [160] it is unlikely, therefore, that even transrectal US would be able to differentiate these two entities. Catheter probe ultrasonography has also been used to demonstrate wall thickening in patients with ulcerative colitis; however, the clinical use of this examination is not clear. [161] The detailed images of pararectal tissues may allow detection of abscesses or fistulae. Abscesses appear as hypoechoic or anechoic areas adjacent to the rectal lumen. Fistulous tracts, with connections to the skin, anus, and vagina, also can be identified by EUS; [162] however, MRI may be a better method in the evaluation of complex fistulae. [163]

Rectal Carcinoma

Transrectal ultrasonography has been evaluated for preoperative staging and the detection of postoperative recurrence of rectal cancer. The depth of cancer invasion into the rectal wall and surrounding tissue and the presence of malignant perirectal lymph nodes have been evaluated using transrectal US imaging (Fig. 154-15). The accuracy of this modality has been compared with CT imaging, but no rigorous studies have been done to show that use of transrectal US changes patient management or influences survival from rectal cancer. Because preoperative radiotherapy may downstage T4 cancers and neoadjuvant chemotherapy may improve survival in patients with T3 cancers, rectal ultrasonography should be used in the preoperative evaluation of these patients. Confirmation of T1 distal rectal cancer is also important if transanal or endoscopic mucosal resection is being considered.

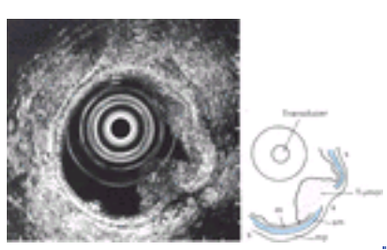

FIGURE 154-15. A polypoid rectal cancer (*T*) invades the submucosa (*sm*) and muscularis propria (*mp*) but does not extend into the subserosal fat (*s*). The boundary between the muscularis propria and the subserosal fat is not disrupted under the tumor (*arrows*).

Most studies of transrectal US staging of rectal cancer have a predominance of extensive lesions, especially those with involvement beyond the muscularis propria [164], [165], [166], [167], [168], [169], [170], [171] and [172] (Table 154-5). Transrectal US correctly predicts invasion beyond the muscularis propria in about 85% of cases. The accuracy of detecting the extent of invasion within the rectal wall is only about 70%. Overstaging the extent of invasion within the wall may be caused by distortion of the US image by inflammation in the tissue layer beneath the cancer. [164] Lymph node involvement by rectal cancer has been studied by using transrectal US which is accurate in detecting lymph node involvement in about 75% of cases [164], [168], [169] and [170] (see Table 154-5). As with esophageal, gastric, and pancreatic cancer, the US appearance of malignant lymph nodes is nonspecific; malignancy within suspicious nodes should be confirmed with EUS-FNA whenever possible. [40], [56] Lymph nodes smaller than 5 mm in diameter usually are missed on trans- rectal ultrasonography. [173]

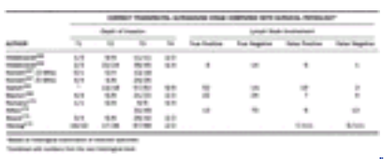

TABLE 154-5 Endoscopic Ultrasound Staging of Rectal Cancer

Transrectal US has been used in an effort to detect early recurrences after the resection of rectal cancer. Postoperative recurrences usually are detected as mass lesions but also may have an infiltrating appearance. [32], [174], [175], [176] and [177] Anastomotic and extrarectal recurrences may be detected by transrectal US when CT scans are negative or equivocal. [32], [174] A false-positive US diagnosis of recurrence may be the result of inflammatory or fibrotic tissue. EUS-guided biopsy of extralumenal masses should allow differentiation of recurrent cancer from fibrosis. [32], [176], [177] Transrectal US staging of rectal cancers has been compared with clinical staging schemes that are largely based on the result of a digital rectal examination. [178] US staging is more accurate than clinical staging, especially in detecting invasion beyond the muscular wall and in staging tumors proximal to the reach of the examining finger.

Rectal cancer staging by transrectal US also has been compared with staging by CT and MRI. Transrectal US and CT are both accurate in determining whether the cancer is confined within or has spread beyond the muscularis propria in 65% to 90% of cases; [168], [170], [178] however, CT cannot detect lesser degrees of wall invasion. [178] CT is also less accurate in detecting enlarged perirectal lymph nodes. [170] These differences in the accuracy of staging by transrectal US and CT are not clinically significant if only major surgical resection is contemplated. However, if less invasive therapies, including transanal excision and endoscopic mucosal resection, are used, the additional information provided by US could aid significantly in management planning. [179], [180] MRI may prove to be as accurate as rectal US for staging rectal cancer. [181] The ability of MRI in differentiating fibrosis from neoplasm is a particular advantage. The availability of rectal surface coils for MRI may allow even further improvements in resolution. [182]

Anal Sphincter Defects

The internal anal sphincter as well as the external anal sphincter, puborectalis sling, and pelvic muscles can be visualized by EUS or blind rectal US. This method is useful in visualizing anal sphincter defects and planning therapy for patients who have fecal incontinence. [183], [184]

INTERVENTIONAL ENDOSONOGRAPHY

Endoscopic US scanning has become an integral part of modern gastrointestinal endoscopy. With the aid of high frequency ultrasonic transducers, it is possible to image a wide range of different disorders located inside and adjacent to the gastrointestinal tract. This has stimulated interest in monitoring endoscopic therapeutic procedures. EUS-guided therapy can either be performed as an EUS-directed procedure with simultaneous endosonographic monitoring, or as an EUS-assisted procedure in which the endoscopic therapeutic procedure follows the EUS examination.

EUS-assisted therapy can be performed either with radial scanning transducers, longitudinally scanning transducers, or with miniprobes. EUS has been used prior to endoscopic cystogastrostomy, [185] submucosal tumor resection, [186], [187] and [188] mucosectomy, [189], [190], [191] and [192] treatment of vascular lesions, [193], [194] and [195] and steroid injection into resistant esophageal strictures. [196] Examples of EUS-directed therapy with simultaneous monitoring include pancreatic pseudocyst drainage, [197], [198], [199] and [200] abscess drainage, [199], [200] celiac plexus nerve block in patients with pain due to chronic pancreatitis or pancreatic cancer, [201], [202] and [203] injection of vessels

such as esophageal varices and local vascular malformations, [204], [205] botulinum toxin injection in patients with achalasia, [206] and cytoimplant placement in patients with advanced pancreatic cancer. [207]

ESTABLISHED INDICATIONS

The ability of EUS to examine the detailed structure of the bowel wall and adjacent organs has made it a useful tool in the investigation of many gastrointestinal disorders. More than many other techniques, EUS has been evaluated extensively, but its clinical value in routine practice continues to evolve. Although the accuracy of EUS in staging gastrointestinal and pancreatic malignancy is firmly established, debate continues as to the impact of this information on patient outcome. The American Endosonography Club published a prospective multicenter study on the impact on management in 428 patients. [208] It concluded that EUS findings alter clinical management in 75% of patients, with a change of major consequence in approximately one third. The recommendations and guidelines from several international groups on endoscopic ultrasonography have also been published and provide a concise review of accepted indications for EUS [209], [210] and [211] (Table 154-6).

DIAGNOSTIC	INTERVENTIONAL
Subepithelial mass definition	Endoscopic mucosal resection
Mediastinal mass definition*†	Treatment of vascular lesions
Lung cancer staging*†	Transmural pseudocyst drainage
Large gastric folds	Celiac plexus neurolysis
Esophageal cancer: local and regional staging*	
Gastric cancer: local and regional staging*	
Ampullary cancer: local and regional staging	
Pancreatic cancer	
Diagnosis of small masses*†	
Evaluation of resectability*†	
Pancreatic endocrine tumors: preoperative localization†	
Other abdominal masses*† (adrenal, lymphoma)	
Chronic pancreatitis: diagnosis†	
Choledocholithiasis: diagnosis†	
Rectal cancer: local staging	
Anal sphincter defects (incontinence)	

*EUS-guided fine-needle aspiration may be helpful in selected situations.
†EUS should be used in conjunction with other imaging tests.

TABLE 154-6 Indications for Endoscopic Ultrasonography (EUS)

FUTURE TRENDS

EUS is a relatively new imaging modality that is still evolving. The technique has been limited to relatively few expert centers throughout the world. New instruments that allow EUS-directed tissue sampling are expected to expand the clinical value of this procedure. In the future, instruments with biplane transducers allowing for an ultrasonic examination in two planes may increase its value even further. Color Doppler, duplex scanning, and pulsed Doppler capabilities are available today but need further evaluation. [22] Three-dimensional EUS is presently under development, [212], [213] although its clinical relevance is not known. More studies on EUS-guided aspiration biopsy and comparison with other techniques are awaited to define the exact indications for this technique. Also, development of new needle types that obtain core biopsies for histological analysis is expected. New endoscopes with larger working channels will allow new applications such as transintestinal placement of large drainage catheters in pancreatic pseudocysts or abscesses. [197], [199] Additional well-designed studies are needed to evaluate the impact of EUS and EUS-guided biopsy on patient management and outcome.

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CHAPTER 155

Alec J. Megibow

APPLICATIONS OF COMPUTED TOMOGRAPHY TO THE GASTROINTESTINAL TRACT

Computed tomography (CT) scanning is widely accepted by the medical community; virtually every practicing physician, and patient for that matter, has familiarity with the appearance of CT images, and their applications throughout the body. The significance of this breakthrough in diagnostic imaging was recognized by the entire scientific community by awarding Godfrey N. Hounsfield and A.M. Cormack with the Nobel Prize in 1979. It was estimated in 1997 that 87% of practicing radiologists interpret CT images; only plain film radiography and fluoroscopy being more widely performed, according to the American College of Radiology (*ACR Bulletin,* February 1997, Volume 53). Continuous motion (helical or spiral) CT has replaced conventional sequential acquisitions, and now this is being replaced by multidetector row (multislice) systems. [1] These systems permit the acquisition of a continuous stream of image data which allows for:

- significantly shortened acquisition times
- the real beginning of increased reliance on three-dimensional data.

Perhaps the most important consequence of this wide dissemination and technologic advancements is the increasing use of CT as a routine method of x-ray examination, challenging the role of the plain film and intraluminal studies. [2]

COMPLICATIONS

CT scanning is so widely disseminated and routinely used in clinical practice it becomes increasingly difficult to be mindful that there are inherent risks associated with the procedure which should be considered before any patient is referred for scanning. Radiation exposure and intravenous contrast administration account for the two broad categories of complications.

Radiologists have expressed concern over increasing radiation dose to patients (particularly children) directly resulting from increasing use of CT scanning. [3] The average effective doses for the abdomen are 12.85 and 19.62 mSv for spiral and conventional CT, respectively. Typical multiple-scan average doses are in the range of 10 to 40 mGy for body scans. [4] Dose can be decreased by lowering tube current without loss of diagnostic quality. Calculation of effective dose will allow investigators to optimize acquisitions across body habitus, types of contrast, and clinical application. [5] Radiation doses are increased when multislice CT scanners are used.

Complications associated with intravenous contrast administration are grouped as:

- minor reactions (rashes, urticaria, nausea, and vomiting)
- serious reactions (anaphylaxis, cardiorespiratory compromise, or contrast-induced renal failure).

The incidence of minor reactions is clearly diminished by using low osmolar agents. Although serious reactions can occur with low osmolar agents, they are given in a higher risk population of patients and are probably safer. However, these agents offer no statistically verifiable safety from fatal complications [6] and have no proven safety in patients with impaired renal function. One survey documented that intravenous contrast material is administered for 90% (median) of abdominal and pelvic CT examinations; nonionic contrast material is used in 68%. [6a] We believe in selective use of low osmolar agents as provided in the guidelines developed by the American College of Radiology.

THE HOLLOW VISCERA OF THE GASTROINTESTINAL TRACT

Techniques

CT abnormalities in the lumenal gastrointestinal tract are recognized by identifying a thickened bowel wall. [7], [8] The suspicious bowel loop must be distended before confident, accurate recognition of pathological wall thickening can be made. Orally and rectally administered agents provide either positive (a “white” bowel using orally or rectally administered 1.5% to 2% water soluble contrast or 2% barium suspension) or negative contrast (gray or black bowel using plain water, gas-producing granules, or insufflated air). In practice, a combination is used. When a clinician reviews a CT scan, the first parameter he or she should evaluate is the adequacy of bowel opacification. Unfilled loops can be mistaken for pathological masses, and significant abnormalities may be obscured. High-density compounds, such as metal and barium, cause severe, image degrading artifacts. If these materials are in the region of imaging interest, the scans should be delayed until they can be cleared. Preliminary localizing radiographs should identify monitoring lines which can be moved. When planning multiple imaging tests for an individual patient, CT should be scheduled *before* barium studies because it may take several days for the barium to clear sufficiently to allow adequate CT examination.

Using intravenous contrast-enhanced images expands the diagnostic range of CT. Essential in the evaluation of the solid abdominal viscera, intravenous contrast enhancement improves conspicuity of pathological processes in evaluation of the alimentary canal by increasing the level of confidence in diagnosing a thickened loop as truly diseased. This refines the differential diagnosis by highlighting attenuation differences *within* the thickened loop, or by accentuating the “vascularity” of a bowel-related mass thereby allowing assessment of histological type and neoplastic potential. [9] Variations in blood supply of pathological processes demand a tailored approach to intravenous contrast administration protocols. Hepatic studies in cirrhotics looking for hepatocellular carcinoma are given intravenous contrast in a different manner than patients with colon cancer in whom the presence or absence of liver metastases is questioned. Pancreas studies are performed so that a rendered angiographic display can be generated from the source data and presented with the axial images.

Esophagus

For evaluation of masses, CT scanning aids in assessing local extension, locoregional lymphadenopathy, and thoracic and abdominal metastases. Conversely, secondary effects of mediastinal processes on the esophagus may be easily imaged aiding in surgical planning. In patients with suspected esophageal perforation, with or without thoracic trauma, CT can localize effusions and evaluate the status of adjacent lung. [10]

Neoplasms All esophageal neoplasms appear on CT as focal, asymmetric, soft tissue attenuation masses arising from the wall or as concentric wall thickenings. These findings are nonspecific and can be seen in benign and malignant tumors and in inflammatory conditions. [11]
Malignant neoplasms. Carcinoma of the esophagus typically appears on CT as a focal eccentric mass or as concentric thickening of the esophageal wall, neither of which is a specific finding. The role of CT in esophageal carcinoma includes staging the extent of disease (highest use in detecting extraesophageal disease), evaluating patients after treatment for therapy responses, and evaluating the complications of surgical therapies. Therapy success or failure can be based on documentation of the development of enlarged mediastinal or upper abdominal lymphadenopathy, lung nodules, or liver and adrenal metastases. CT criteria for extensive esophageal cancer include demonstration of local invasion (aorta, diaphragm, tracheobronchial tree), distant lymphadenopathy, and distant metastases. Local extension is the most difficult parameter to accurately diagnose. Contact angles of more than 45 degrees have been suggested to accurately predict aortic invasion, although several studies have shown this finding to result in frequent overstaging. [12] Pathological lymphadenopathy can be more reliably determined when regional adenopathy of more than 5 mm in the short axis is used as a criterion. [13] CT has been shown to be as accurate as fiberoptic bronchoscopy in assessing tracheobronchial invasion. If minimal disease is present, procedures such as endoscopic ultrasound (EUS) may be necessary for accurate preoperative staging. [14] Postsurgical complications can be detected by demonstrating anastomotic leaks, which occasionally are seen only on CT scans and are not detected by standard contrast esophagography. Although the site of the leak may be demonstrated, CT provides no histological specificity; squamous cell carcinoma, adenocarcinoma, lymphoma, and metastatic disease to the esophagus have similar CT appearances. Esophageal stromal tumor should be considered when there is a prominent exophytic component to the mass. [15]
Varices Esophagogastric varices are easily detected with contrast-enhanced CT and diagnosis is improved using helical CT techniques. CT has additional use in that it can show extraserosal varices, which may be underestimated by EUS. Thickening of the esophageal wall, a scalloped contour, and enhancing intralumenal protrusions after a contrast bolus injection are seen. [16] Low attenuation in the esophageal wall in patients with other stigmata of portal hypertension should suggest a history of sclerotherapy.
Esophagitis Esophagitis may produce thickening of the distal esophageal wall. CT is neither sensitive nor specific for this finding. CT does not aid in the differentiation of distal abnormalities caused by either carcinoma or severe esophagitis. CT may be useful in recognizing distal esophageal changes in immunocompromised patients who can go on to endoscopic evaluation for specific etiology.

Stomach

CT can evaluate the position of the stomach, the lumenal contour, the gastric wall, surrounding lymph nodes, peritoneal ligaments, and relation to surrounding viscera. The distended gastric wall should measure less than 5 mm. At the esophagogastric junction and the pyloric canal, the wall may be up to 1-cm thick and still be normal. The wall displays a uniform enhancement pattern following intravenous contrast administration. Use of negative contrast allows assessment of the distribution of the rugal fold pattern in the proximal stomach. Changes in positioning may be necessary to maximally distend regions of the stomach where the wall is questionably thickened. There have been initial investigations of three-dimensional imaging capabilities in improving evaluation of the stomach and perigastric relationships. [17]

Neoplasms Gastric carcinoma appears as a region of focal, variably thickened gastric wall. Polypoid and scirrhous forms (with or without ulceration) are encountered. Scirrhous tumors (linitis plastica) can be difficult to diagnose as the degree of wall thickening is less than in polypoid tumors. [18] Gastric cancer spreads along the perigastric peritoneal reflections through intramural lymphatics, by peritoneal seeding, and hematogenously (Fig. 155-1). Lymphatic submucosal spread into the esophagus occurs in approximately 30% of patients with tumors in the gastric fundus. Extension into the serosa allows tumor access to the supporting mesenteries of the stomach providing pathways into omentum, kidney, liver, pancreas, transverse colon, and spleen. Proximal lesions can extend into the diaphragm and the aorta.

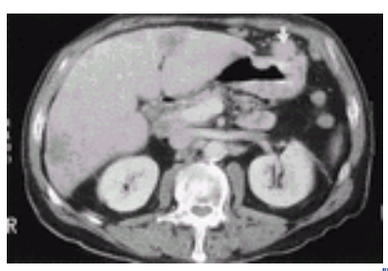

FIGURE 155-1. Gastric carcinoma—patterns of spread. A gastric carcinoma is identified by a thickened wall in the proximal stomach. A nodular density is seen along the greater curvature aspect of the lesion representing peritoneal seeding (*arrow*). Multiple hepatic metastases are evident in the enhanced liver, indicative of hematogenous spread.

With single-slice helical CT, using thin sections and intravenous contrast, resultant density discrimination improves staging accuracy over conventional CT. In a Japanese study, overall T-staging was correct in 66% (27 of 41), despite a large number of “early” carcinomas that might bias the sample against CT accuracy, [19] implying that results should be improved in populations where patients present with more advanced disease. Dedicated imaging protocols have yielded detection of lesions in greater than 95% of patients and detection of lymph node disease in approximately 71%. [20] Gastric lymphomas produce significantly greater wall thickness than carcinoma, although this finding is of limited value for differential diagnosis in individual cases. Following intravenous contrast enhancement, lymphoma masses minimally enhance and remain homogeneous, as opposed to the more frequently heterogeneous adenocarcinoma. In cases presenting with fold hypertrophy, there may be demonstrable clefts between the thickened folds extending through the wall into the extraserosal tumor. [21] CT cannot be used to screen for mucosa-associated lymphoid tissue (MALT) in patients with chronic *Helicobacter pylori* infections. [22] CT is accurate for detecting complications such as perforation and fistulization. Metastatic disease to the stomach may be indistinguishable from primary adenocarcinoma. Particular attention should be focused on the gastric antrum in patients with carcinoma of the breast, where a linitis plastica appearance resulting from breast metastases has been reported.
Stromal Tumors A wide variety of stromal tumors occur in the stomach. Spindle cell lesions are the most common. [23], [24] CT is useful in displaying the predominant tissue (particularly in recognizing fat). Malignant lesions are larger and present as huge abdominal masses. The attachment site may be quite small considering the size of the mass. The CT appearance correlates with biologic behavior of the mass (i.e., large malignant lesions with wide areas of liquefaction are easily recognized on CT whereas, because of scant viable tissue, numbers of mitoses may be limited and true malignant potential of a lesion may be less well appreciated). [25], [26] An exophytic growth pattern is unusual, although it has been reported for adenocarcinoma.
Inflammatory Diseases CT scanning has no primary indication for the evaluation of inflammatory gastric diseases. However, they may result in a focal or diffuse wall thickening. The thickened wall will display some degree of low attenuation, reflecting edema in the gastric wall. Only scattered reports of cases of neoplasm display this finding. [27]

Small Intestine

The key to evaluation of the small intestine lies in adequate opacification with orally administered contrast material. Typically, 500 to 600 mL of dilute contrast material should be administered 1 to 2 hours before examining the abdomen to provide opacification of the small bowel and avoid mistaking fluid-filled or nondistended loops for abnormal masses. Oral metoclopramide aids in gastric emptying and filling of the terminal ileum. There has been growing interest in the use of negative contrast agents in small bowel evaluation engendered by the increasing use of three-dimensional imaging.

Neoplasms Two large categories of benign lesions include adenomatous polyps and stromal tumors. The latter are more commonly encountered at imaging. Leiomyoma, the most common stromal lesion, appears as a rounded, soft tissue attenuation with an exophytic growth pattern. Lipomas can be definitively diagnosed by the recognition of fat attenuation within the mass. Adenocarcinoma of the small intestine produces soft tissue attenuation wall thickening and irregular narrowing of the lumen which can lead to obstruction. This lesion is most frequently seen in the periampullary duodenum, and may be a cause of biliary obstruction. Malignant lesions demonstrate exophytic or intramural mass, central necrosis, and ulceration. [28] The radiologic manifestations of lymphoma of the small intestine are classified as aneurysmal, constrictive, nodular, ulcerative, mesenteric, and sprue forms. The morphologic patterns of nodular, ulcerative, and aneurysmal disease are best visualized with CT. [29], [30] Acquired immunodeficiency syndrome (AIDS)-related lymphomas display two main patterns on CT: single or multiple segments with homogeneous, circumferential wall thickening, or single or multiple cavitary lesions. The gross morphologic features, distribution pattern, degree of wall thickening, and length of involvement are similar in AIDS-related and non–AIDS-related lymphomas. [31] Carcinoid tumor results in a well-described, specific CT appearance resulting from the intense desmoplasia incited by the tumor. The primary small bowel lesion often goes undetected. The CT appearance is characterized by radially oriented linear densities in the perienteric fat. A lymph node mass (frequently calcified) is present centrally. Bowel loops are drawn toward the center. [32] The mesenteric desmoplasia may produce ischemia thickening the involved loops. Metastatic disease to the small bowel results from blood-borne (embolic) metastases, peritoneal seeding, or from direct extension from a contiguous lesion. Each has a recognizable CT appearance. Embolic metastases are commonly seen from

melanoma, Kaposi sarcoma, and breast carcinoma [33] (Fig. 155-2). Discrete masses eccentrically localized along the bowel circumference are present. Tethering of the mesenteric border of individual bowel loops in the presence of changes of peritoneal carcinomatosis is indicative of seeded metastases. Variable degrees of partial obstruction are common. Nodular thickening of the mesenteric reflections is observed. These findings are seen in metastatic ovarian, colonic, breast, and lung carcinoma.

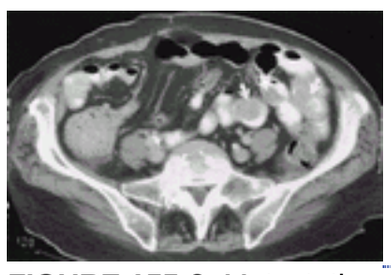

FIGURE 155-2. Metastatic melanoma multiple soft tissue attenuation masses are readily identified in the wall of the jejunum (*arrows*).

Nonneoplastic Diseases

Crohn's disease. One of the earliest applications of CT in the alimentary tract was the evaluation of Crohn's disease. [34] Axial display and markedly expanded contrast resolution compared to plain film and barium radiography make CT ideal in displaying the transmural inflammation, effects in local fat, and global assessment of disease. [35] The affected bowel wall displays variable degrees of mural thickening. When intravenous contrast is administered, submucosal hypoattenuation, reflecting submucosal edema, may be seen. This "target sign," although nonspecific for Crohn's disease, was first described in this entity [36] (Fig. 155-3). This appearance may be mimicked in long-standing disease from deposition of submucosal fat. The bowel becomes "isolated" in a "sea" of hypertrophied fat. CT definitively establishes this fatty proliferation as a cause of separation of bowel loops. The fatty proliferation is seen with hypervascularity in the local mesentery. [37] Linear nonvascular densities are correlated with the presence of sinus tracts or fistulae. When these are identified on CT, barium studies are necessary to precisely map their relationships. Local lymphadenopathy (generally not greater than 15 mm) is associated with these findings. Increased density in the fat reflects inflammatory exudate and when contained, frank abscess. Intra-abdominal abscess is not uncommon, with severe, steroid-refractory relapses of Crohn's disease being present in about one third of patients. CT is superior to indium 111 leukocyte scintigraphy, which is sensitive to inflammation but poor in documenting extent. Indium 111 leukocyte scintigraphy is the most specific method to image "active inflammation."

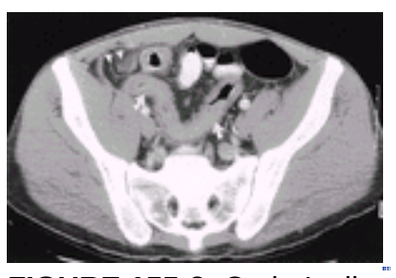

FIGURE 155-3. Crohn's disease—ileum. A loop of ileum possesses a thickened wall (*arrows*). Note the stratification of density in the wall. Central lucency represents edema. Increased density in the adjacent mesenteric fat results from the transmural inflammatory process (*arrowheads*).

CT findings simulating Crohn's disease have been described in a variety of infectious processes including: *Yersinia, Shigella, Salmonella,* [38] and *Mycobacterium tuberculosis.* Specific radiologic diagnosis is impossible; findings must be interpreted in context of patient presentation. In patients with AIDS, cytomegalovirus (CMV) infection frequently produces distal ileitis resembling Crohn's disease.

Vascular diseases. Using helical and multislice CT, enhancement levels of normal bowel reach levels between 110 to 120 Hounsfields. [39] A wide variety of conditions produce abnormalities at all levels of the small intestinal vasculature. The most severe forms of ischemia result from emboli to the superior mesenteric artery. These patients rarely come to CT. "Small vessel" diseases are most likely to manifest on CT. These include diseases such as lupus erythematosis, graft-versus-host disease, polyarteritis nodosa, and radiation enteritis. All result in relatively short segments of thickened intestinal wall with prominent submucosal edema. Intramural hemorrhage, whether secondary to vasculitis, excessive anticoagulation, or trauma, results in high-attenuation wall thickening. [40] Variable degrees of obstruction may be present from the compression of the lumen secondary to the intramural hematoma. Frank infarction is difficult to diagnose clinically and by CT. The infarcted portion generally displays a thin wall and fluid-filled lumen. [41] In advanced cases, linear pneumatosis may be present in the bowel wall (Fig. 155-4). Pneumatosis may be seen in the absence of infarction.

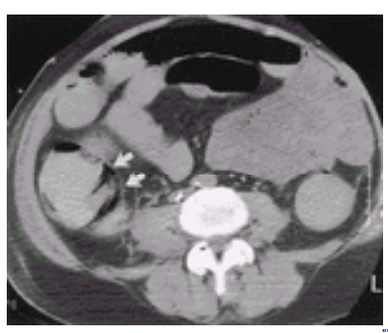

FIGURE 155-4. Bowel infarction—visualized bowel loops are distended, fluid-filled, and airless. Linear streaks of intramural air (*arrows*) are identified in the cecum. The patient expired despite attempt at surgical resection.

Trauma. CT is widely used in the evaluation of blunt abdominal trauma. [42] Solid organ injury is relatively easily detected; the significance of subtle findings indicative of severe mesenteric injuries are becoming more frequently recognized. [43] If mesenteric infiltration, bowel wall thickening, extravasation of vascular or enteric contrast agent, and free air are visualized, sensitivity, accuracy, and specificity are 62%, 82%, and 97%, respectively. [44] CT can contribute to early surgical management which may have been delayed if only clinical signs were incorporated into decision making. [45]

Small bowel obstruction. CT findings of obstruction are similar to traditional radiographic findings: disparate dilation of proximal bowel loops compared with more distal ones. Comparative studies have shown that CT is superior to plain film radiography in detecting intestinal obstruction and in determining the cause of obstruction. [46] Studies have shown CT sensitivities ranging from 90% to 95% in detecting obstruction, with no false-positive examinations (Fig. 155-5). CT establishes the etiology in a majority of cases. [47] The major contribution of CT in these patients is the ability to detect signs of strangulation. These signs include thickening and edema in affected loops (ischemic signs) and mesenteric clouding, ascites, or infiltration (mesenteric signs) [48] (Fig. 155-6). The combination of multiple findings has improved reliability in predicting obstruction as compared to any single sign in isolation. [49]

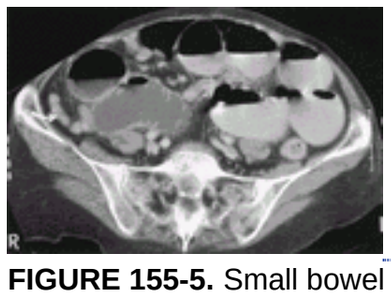

FIGURE 155-5. Small bowel obstruction—computed tomography findings. Dilated small bowel loops occupy most of the abdomen. Collapsed distal ileal loops are recognized (*curved arrow*). This pattern is diagnostic of small bowel obstruction.

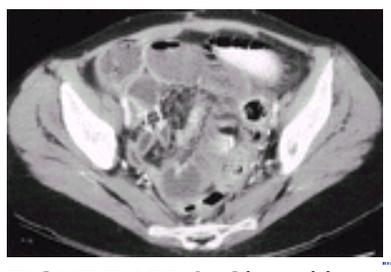

FIGURE 155-6. Closed loop obstruction—high-grade obstruction is evident. Note the converging of two adjacent narrowed loops at the transition to collapsed lumen (*curved arrows*). This appearance is diagnostic of a closed loop strangulated small bowel obstruction and considered as an indication for immediate surgical decompression.

CT can change management by accelerating surgical treatment in obstructed patients resulting in verifiable improvements in outcome. [50], [51] CT efficacy decreases in patients with chronic obstructive symptoms and low-grade obstruction. For these patients, barium radiography, especially by enteroclysis, is the method of choice. [52]

Colon

CT is useful in the evaluation of many colonic disorders. Because CT shows both the colonic wall (generally <3 mm) and surrounding pericolic fat, extension of pathological processes into the surrounding pericolic fat offers improved staging of tumors and preoperative assessment of complex inflammatory reactions in a single examination, thereby facilitating both diagnosis and treatment decisions.

Neoplasms

Malignant neoplasms. A thorough preoperative evaluation is essential in selecting appropriate operative therapy and for sequencing surgery with available adjuvant treatments. [53] These decisions are based on the anatomic location of the primary tumor(s) and the presence or absence of metastatic disease. An additional element in rectal cancer patients is assessment of transmural penetration and the distance from the anal verge. Although EUS excels in local staging of accessible lesions, CT provides the most efficient methodology by which *all* of these components can be comprehensively evaluated. Results from a multi-institutional study reported 74% accuracy for CT assessment of wall invasion; the sensitivity in evaluating lymph node metastases was 48%. CT demonstrated an 85% accuracy and 97% specificity in detection of liver metastases. [54] The detection of colorectal carcinoma depends on the degree of colonic distention and adequacy of the preparation. [55] In the unprepped bowel, all but the largest lesions will be overlooked. Adenocarcinoma appears as a soft tissue attenuation focal mass, which focally or circumferentially thickens the bowel wall. Using air insufflation and a colon preparation, detection was increased from 68% to 95% [55] (Fig. 155-7).

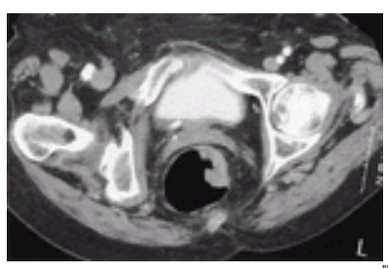

FIGURE 155-7. Rectal carcinoma. An ulcerated mass is visualized along the left wall of the rectum. No obvious extension into perirectal fat is seen.

Transmural invasion is suggested when streaky, spikelike soft tissue attenuation changes are seen in the peritumoral fat. The main limitation of CT staging is understaging the extent of local disease, with few overstaging problems. CT is useful in identifying complications of colorectal cancer such as perforation (Fig. 155-8), obstruction, and synchronous lesions. These may be clinically silent but their presence impacts on long-term survival. The negative predictive value of CT has been estimated at 90%. [56] Although most surgeons have come to rely on preoperative CT scanning to survey the abdomen and pelvis, there is no routine agreement regarding widespread use. [57]

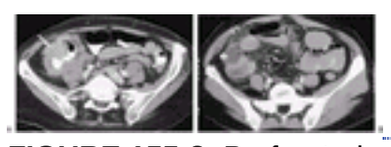

FIGURE 155-8. Perforated cecal carcinoma. **A:** A bulky mass is present in the cecum (*arrow*). Note the spikelike borders typical of an invasive lesion. **B:** An abscess collection (*arrow*) is identified. Note the peritoneal reaction (*arrowheads*); it is impossible to differentiate between local peritonitis or peritoneal seeding.

CT plays an adjunctive role in evaluating recurrent disease after abdominoperineal resection or colonic resection. CT is capable of demonstrating locally recurrent pelvic masses, lymphadenopathy, and liver metastases. Locally recurrent rectal cancer appears as spiculated densities or a fibrous soft tissue layer around the rectum; when the abnormal tissue is attached to the perirectal fascia or extends beyond it, positive predictive value is high. [58] CT or ultrasound (US) imaging combined with colonoscopy has been shown to detect recurrent colon cancer at a lower total cost per patient than routine use of carcinoembryonic antigen (CEA), physical examination, and chest x-ray. [59] However, use may be greater in tumors arising in the abdomen as opposed to the pelvis.

CT colonography (virtual colonoscopy). The ability of helical CT to acquire a continuous volumetric data set, three-dimensionally, allows application of rendering techniques which result in a display of the lumenal (mucosal) surface of the colon, a process known as CT colonography or “virtual colonoscopy”. [60] Extensive evaluation of virtual colonoscopy has occurred since the late 1990s. [61], [62] Uniform distension, multiple positions, and meticulous bowel preparation are absolutely necessary to achieve diagnostic results. Patient table time is less than 3 minutes, more time is necessary for processing of the data. Most investigators have abandoned the “fly-through” movie relying on two-dimensional multiplanar reformatted images and use three-dimensional movies as a static problem-solving technique. [63], [64] Virtual colonoscopy will not compete with colonoscopy, however it *will* compete with barium enema, and investigators have used this technique to “finish” incomplete colonoscopy [65] (Fig. 155-9). The accuracy of polyp detection depends on size of the lesion. In a widely quoted study, virtual colonoscopy identified all 3 cancers, 20 of 22 polyps that were 10 mm or more in diameter (91%), 33 of 40 that were 6 to 9 mm (82%), and 29 of 53 that were 5 mm or smaller (55%). [62] Of the 69 adenomatous polyps, 46 of the 51 that were 6 mm or more in diameter (90%) and 12 of the 18 that were 5 mm or smaller (67%) were correctly identified. [62] It is hoped that virtual colonoscopy will eventually make full colonic evaluation accessible to a greater number of patients than presently take advantage of colorectal cancer screening programs and aid in polyp detection.

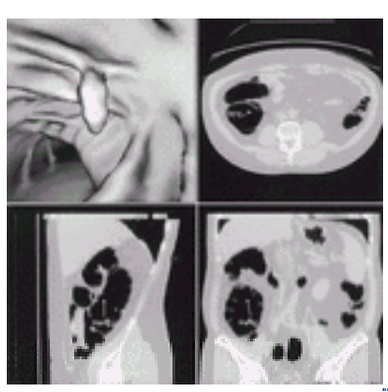

FIGURE 155-9. A 1-cm pedunculated adenoma in the ascending colon. This computed tomography (CT) colonoscopic view (**upper left panel**) identified this

pedunculated polyp. Reformatted “conventional” CT images in axial, coronal, and sagittal planes are included in the data display (**other panels**). The patient was referred following an incomplete colonoscopy, taking advantage of the patient preparation. Repeat colonoscopy was successful to the cecum and the polyp was removed.

Nonneoplastic Diseases

Diverticulitis. In those patients in whom the clinician requires imaging clarification or confirmation of a diagnosis of diverticulitis, CT is the primary, and in most instances, the only necessary test. [66], [67] Although CT and lumenal radiography have equal accuracy in *establishing* the diagnosis, CT is clearly superior in determining whether abscess is present and in documenting the extent of the inflammatory mass [68] (Fig. 155-10). When abscess is present, resection of the perforated segment with anastomosis is desirable, however if the field is contaminated by pus, temporary colostomy may be necessary. Percutaneous abscess drainage reduces surgical morbidity and allows a primary anastomosis. [69]

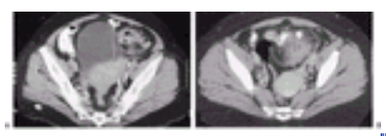

FIGURE 155-10. Diverticular abscess (**A**). Increased density in the fat of the sigmoid mesocolon indicates diverticulitis (*open arrow*). **B:** An abscess (*arrow*) is defined within the dense inflammatory response.

CT is excellent in diagnosing diverticulitis as a cause of fistulae arising from the colon, particularly those to the urinary bladder or the vagina. [70] The major limitation of CT is the inability to assess the integrity of the colonic mucosa, and perforated neoplasm may be overlooked; [71] this is estimated to be a problem in approximately 10% of cases. In these individuals, lumenal evaluation with either endoscopic or contrast radiography is indicated. [72]

Appendicitis. CT has emerged as the primary imaging modality in patients suspected of acute appendicitis. [73] Outcome-based studies [74], [75] have documented an impact on negative appendectomy rates in patients who have undergone CT imaging. CT has similar accuracy to US, although the negative predictive value of CT is significantly higher. [76] Local expertise and body habitus (thinner patients for US and larger patients for CT) should dictate the use of US or CT. The diagnosis is dependent on visualizing an abnormal appendix. Defining criteria include a dilated, thick-walled tubular structure, which brightly enhances following intravenous contrast [77] (Fig. 155-11). Periappendiceal inflammatory changes may be minimal, even when the appendix is found to be gangrenous at surgery. Although it is possible to recognize findings of acute appendicitis on unenhanced studies, [78] we *strongly* believe that intravenous contrast and oral contrast and acquisition using thin sections [79] are essential in patients *referred* for evaluation of right lower quadrant pain.

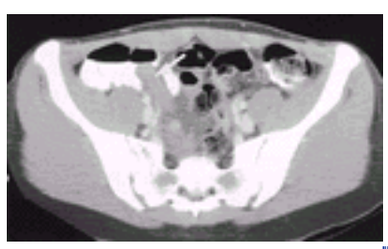

FIGURE 155-11. Appendicitis. No periappendiceal inflammation is present. The appendix is recognized as a tubular structure with a thick, hyperdense wall and a fluid-filled lumen (*arrow*). Cecal opacification with orally administered contrast is essential.

Differential diagnosis of right lower quadrant inflammatory diseases includes cecal diverticulitis, [80] epiploic appendagitis, [81] and right-sided colitis. CT may not be able to predict neoplasm obstructing the appendiceal orifice.

Colitis. For most patients with a known diagnosis of acute colitis, CT offers little information. CT is most useful in those with nonspecific symptoms, directing the workup to an unsuspected colitis. [82] As in small bowel diseases, submucosal lucency representing edema can be seen on enhanced scans. In an attempt to suggest a specific diagnosis, one evaluates mural thickness and enhancement pattern, distribution of bowel involvement, and associated mesenteric and small bowel disease. The mean colon wall thickness in Crohn’s colitis is significantly greater than in ulcerative colitis. Exclusive involvement of the right colon and small bowel is more frequent with Crohn’s and infectious colitis. Abscess is associated almost exclusively with Crohn’s colitis. [83] An accordion-like appearance of the colon can be seen in a variety of acute colitides. [84] CT is not indicated in workup of patients with ulcerative colitis. The disease is recognized by a continuously thickened segment of intestinal wall, displaying a targetlike appearance. Fat hypertrophy in the presacral region is seen. In patients with long-standing colitis, fat deposition within the wall may occur. [85] CT is not sensitive enough to monitor these patients for the development of carcinoma. In Crohn’s colitis, CT has additional use in that pericolic disease is frequent. CT has unique application in the evaluation of perianal complications. [86] CT findings in pseudomembranous (*Clostridium difficile*) colitis produces dramatic wall thickening, as an almost uniform accompaniment of pseudomembranous colitis [87] (Fig. 155-12). The diagnostic sensitivity decreases without the use of intravenous contrast. [88] CMV colitis has a similar appearance to pseudomembranous colitis. Ischemic colitis appears as a focal area of wall thickening with intramural lucency. [89] In most cases, local ascites is present. Care should be taken to rule out a distal obstructing carcinoma. [90] Similar symptoms may be produced by hemorrhagic *Escherichia coli* infection. [91] Neutropenic colitis has a predilection for the terminal ileum and right colon.

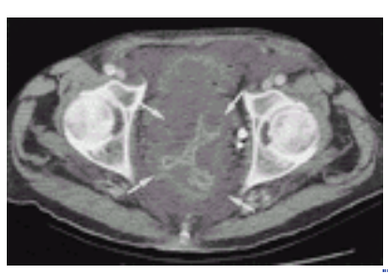

FIGURE 155-12. *Clostridium difficile* proctocolitis. There is massive thickening of the colonic wall; the serosal margin is identified by the arrows. The central Y-shaped density represents a markedly hyperemic mucosa; the wall thickness is entirely edematous. This patient presented with a postoperative fever; no diarrhea or rectal bleeding was noted.

THE SOLID ORGANS OF THE GASTROINTESTINAL TRACT

Liver

Techniques The technique of examination of the liver is based on whether the study is to be performed to:

- exclude any amount of hepatic disease
- determine the precise disease burden for preoperative planning
- characterize known masses seen on other imaging studies.

Separating these imaging tasks allows the radiologist to optimize contrast administration, acquisition speed, and timing. Hepatic parenchymal enhancement following a rapid bolus infusion of *intravenous* contrast administration is characterized by three distinct phases: a hepatic artery phase, a portal phase, and an equilibrium phase. The hepatic artery phase, useful in detection of hypervascular lesions and hepatocellular carcinoma (HCC), will last between 30 to 50 seconds following the initiation of injection. All hepatic lesions are supplied by the hepatic artery and it is during this time that these lesions are brightly enhanced because background parenchyma has not reached peak enhancement. At approximately 70 to 90 seconds, peak hepatic enhancement is achieved (portal phase). Peak enhancement maintains itself for approximately 30 seconds and then slowly decays. Most scanning is performed during this portal phase because this is the time when there are maximal differences in attenuation between normal liver parenchyma and most masses. Delayed scanning during the “equilibrium phase” will result in decreased

lesion conspicuity. [92] Software modifications are available to time the beginning of scanning with peak enhancement. [93] Unenhanced scans are important in patients with suspected “hypervascular” lesions, including metastases from renal, thyroid, breast, islet-cell, and carcinoid tumors and HCC. [94] In patients with cirrhosis in whom HCC is suspected, noncontrast scans are especially important due to the changes in hepatic artery/portal vein hepatic supply. The clinician should communicate a specific clinical history or suspicion of these diagnoses to the radiologist for proper examination. [95] With helical/spiral CT, images of the entire liver can be acquired in a single breath hold (20–30 seconds depending on the length of the liver). Using multislice CT, the liver can be scanned in 12 to 15 seconds allowing better use of hepatic artery phase imaging. Following the hepatic arterial images, the liver can be immediately rescanned to capture peak parenchymal enhancement during the portal vein phase. [96] This technique is referred to as “dual-phase” helical CT. [97] Many perform a “three-phase” study recognizing the role of the unenhanced scans (Fig. 155-13). In order to achieve a diagnostic arterial phase image, contrast must be power injected at a rate exceeding 4 mL/s. We reserve this technique almost exclusively for detection of HCC. [98], [99] Intravenous portal-phase, contrast-enhanced helical CT has been reported as sensitive (91%) for detecting malignant hepatic tumors measuring larger than 1 cm but relatively insensitive (56%) for tumors smaller than 1 cm. [100] The availability of multislice CT facilitates simultaneous lesion characterization and detection because both phases of hepatic enhancement are accessible from the same dose of intravenous contrast. [101] Additionally, three-dimensional display improves localization. These results are slightly superior to results of nonhelical CT, although lesions smaller than 1 cm remain elusive.

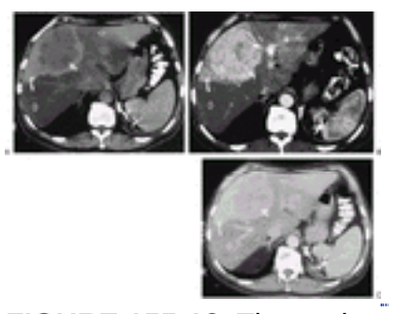

FIGURE 155-13. Three-phase liver examination. This figure illustrates several basic principles of liver imaging with helical computed tomography. A lesion seen on the noncontrast image (**A**) (*straight arrow*) is hypodense to surrounding hepatic parenchyma (*curved arrow*). This segment of parenchyma is denser than the majority of the fatty infiltrated liver (*open arrows*). **B:** An “arterial phase” image resulting in bright enhancement of the lesion with less enhancement of the background liver parenchyma. **C:** The portal phase reveals the lesion to be almost equal in density to the surrounding “normal” liver parenchyma. This perilesional parenchyma is brighter than the maximally enhanced fatty liver due to perilesional circulatory effects.

If documenting the exact number and location of liver metastases is imperative, such as before contemplated resection of apparently isolated metastases, angiographic-assisted CT improves sensitivity. CT-portography (CTAP) has the highest reported sensitivity for detecting focal liver masses, approaching that of intraoperative US. [102] The improved sensitivity may lead to better patient selection for resection of metastases. [103] The volumetric data set from helical and particularly multislice acquisitions allows for angiographic and three-dimensional volume-rendered data displays; these aid in lesion localization and surgical planning. [104] Improvements in contrast sensitivity afforded with magnetic resonance imaging (MRI) have significantly reduced the use of CTAP in most clinical practice today compared to the late 1990s. In our practice, which includes a large number of transplant patients in whom hepatic surgery is dependent on accurate demonstration of entire tumor burden, CTAP is almost never performed.

Diffuse Diseases

Fatty infiltration. Fatty infiltration of the liver may occur in a diffuse or focal distribution. This intracellular fat lowers the liver’s CT attenuation reversing the relation to splenic enhancement as judged on unenhanced scans. [105] Focal fatty deposition can be confused with hepatic space-occupying lesions. Lack of displacement of adjacent vessels, location, and the rapid change with time can be helpful in differentiation. [106] When problematic, the presence of focal fat can be established by biopsy or “chemical shift” MRI.

Cirrhosis. Characteristic CT features can be seen in patients with advanced cirrhosis. The right lobe and medial segment of the left lobe are often decreased in size, but the lateral segment of the left lobe and caudate lobe are enlarged. [107] Additionally, cirrhotic livers demonstrate irregular, nodular liver margins’ apparent widening of the intrahepatic fissures and the porta hepatis as the liver retracts and shrinks. Associated changes of portal hypertension, with varices and venous collaterals, commonly occur. [16] Enhancement of the cirrhotic liver is similar to normal liver during the arterial phase, but may be slightly less dense during the portal phase. [108] A normal CT does not exclude the diagnosis of cirrhosis as fibrosis may still be severe on liver biopsy. The distorted liver appearance caused by cirrhosis obscures many small tumors on CT scans. Considering the worldwide frequency of hepatitis C, the number of cases is expected to increase. Using conventional CT imaging, Miller and associates [109] reported detecting HCC in only 68% of cirrhotic patients with tumor. Incorporating arterial phase helical CT scanning improves to an additional 30% to 40% of HCC tumor nodules over portal venous phase imaging alone. [110] Correlation with subsequent transplant specimens has shown that while arterial phase imaging for detecting HCC in cirrhotic patients is a significant improvement over conventional or portal venous contrast imaging, lesions less than 2 cm in size still go undetected more often than not. [111] While the significance of dysplastic nodules remains a controversial issue, it is known that CT is poor at detecting dysplastic nodules in cirrhotic livers. [112] Sarcoidosis may simulate macronodular cirrhosis. Associated retroperitoneal adenopathy and nodular changes in the spleen suggest the diagnosis. [113]

Other diffuse diseases. Iron deposition and hemochromatosis, whether primary or secondary, increase the attenuation of the liver and spleen on CT scans. [114] CT is valuable in evaluating long-term iron deposition in the reticuloendothelial system from chronic transfusion therapy. The hepatic density may also be increased in patients receiving amiodarone therapy, those with glycogen storage disease, and following gold therapy for rheumatoid arthritis. Despite high-serum copper levels, CT scan of the liver does not show significant elevations over normal attenuation measurements in patients with Wilson disease.

Infections Although CT is not indicated for the primary evaluation of hepatitis, several reports indicate findings suggesting its presence. These should be recognized so that diagnosis and therapy may be expedited. A thickened gallbladder wall with periportal edema may suggest the diagnosis. In patients with cirrhosis due to chronic hepatitis, perihepatic lymphadenopathy is common. [115] Periportal edema may cause this sign in patients with congestive heart failure and secondary liver congestion, hepatitis, or enlarged lymph nodes and tumors in the porta hepatis which obstruct lymphatic drainage. This CT sign has also been observed in liver transplants (probably secondary to disruption and engorgement of lymphatic channels) and in recipients of bone marrow transplants who might develop liver edema from microvenous occlusive disease. [116] Hepatic tuberculosis may appear as a focal mass (tuberculous pseudotumor), an abscess, or result in diffuse liver disease. Hepatic involvement is associated with low attenuation lymphadenopathy and splenic disease. [117] Bacillary angiomatosis results in innumerable dilated sinusoids; high attenuation lymphadenopathy may be seen. A similar appearance has been described in cat-scratch fever. [118] Diffuse calcification throughout the liver has been observed in patients with extrapulmonary *Pneumocystis carinii* infection. [119]

Masses

Cysts. Cysts appear as homogeneous, low attenuation (0–10 Hounsfields) masses having smooth, sharp margins without a perceptible wall. Cysts less than 1 cm may cause concern, particularly in patients with cancer because of artifactually higher attenuation measurements from volume averaging. Assessment of multiplicity and comparison with prior studies is necessary. [120]

Benign neoplasms. Hepatic hemangioma is the most common tumor of the liver. Taking advantage of the improved contrast and temporal sensitivity of helical CT, hemangioma may be accurately and definitively diagnosed in over 90% of cases by CT alone. [121] Criteria include:

- globular enhancement
- lack of continuity of enhancing tissue
- iso-, or hyperdense enhancement relative to the aorta
- peripheral enhancement.

Globular enhancement, isodense with the aorta, is 67% sensitive and 100% specific in differentiating cavernous hemangiomas and hepatic metastases (Fig. 155-14). When demonstrated, no further workup is needed.

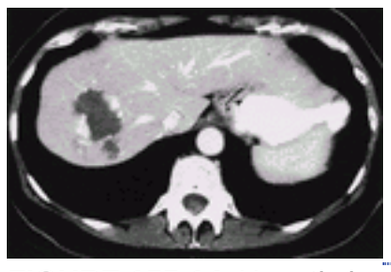

FIGURE 155-14. Hepatic hemangioma. Globular, peripheral enhancement, isodense with the aorta is diagnostic of hepatic hemangioma. When these features are imaged, no further workup is necessary.

Hepatic adenomas and focal nodular hyperplasia (FNH) are vascular lesions that can present with a variety of CT appearances that are often indistinguishable, unless there is hemorrhage (indicating adenoma). [122] Both lesions usually demonstrate homogeneous, increased attenuation as viewed during the hepatic arterial phase of enhancement, rapidly becoming isodense or slightly hypodense as peak portal enhancement is achieved. The identification of a central, stellate, low-density area is helpful in the diagnosis of FNH, seen only in a minority of patients. [123] Patients with fibrolamellar HCC may demonstrate a similar, central low density. Calcification, while rare in FNH, is frequent in fibrolamellar HCC. [124]

Malignant neoplasms. HCC can appear on CT scans as a focal, solitary mass; a multifocal process; or as a diffuse, infiltrating process. These lesions appear hypodense or isodense with normal liver on noncontrast-enhanced images; after contrast administration, larger lesions are predominantly hypodense during portal imaging. Smaller lesions may be isodense during the portal phase and only appreciated during early (hepatic arterial phase) images obtained after rapid (>4 mL/s) contrast bolus. [125] Characteristic appearances of HCC include fibrous capsules, fibrous septa, and mosaic appearances. The fibrous capsules of HCCs are readily detected by CT and MRI. [126] The mosaic pattern is a result of variable tissue composition of HCC. In most patients, enhancing nodules indicate viable tumor cells, and low attenuation areas represent necrosis, fibrosis, or hemorrhage. [127] With the increase in prevalence and incidence of hepatitis B and C worldwide, interest in imaging surveillance of these high-risk populations is gaining momentum. There is considerable interest in regional therapy for HCC, including chemoembolization and radiofrequency ablation. CT imaging aids in localization of foci of tumor for which angiographic administered chemoembolization can be applied. [128] Smaller lesions are amenable to percutaneous alcohol ablation. [129] Liver metastases demonstrate a variety of CT appearances. The majority typically appear hypodense compared with adjacent enhanced liver parenchyma, although vascular lesions can appear hypodense, isodense, or hyperdense, depending on the timing of contrast administration and scanning. Hypervascular metastatic lesions, as are found in renal cell carcinoma, islet-cell tumors, leiomyosarcoma, or thyroid carcinoma, may be better demonstrated on noncontrast-enhanced CT (Fig. 155-15). Metastatic lesions do not have a pathognomonic appearance and can be simulated by HCC, hemangiomas, abscesses, complicated cysts, or hematomas. Analysis of the hepatic artery enhancement patterns of metastases reveal that a complete ring enhancement pattern is a highly specific sign of metastatic liver disease regardless of primary site. [101], [130]

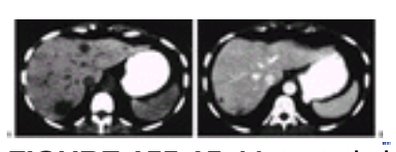

FIGURE 155-15. Metastatic breast cancer. Value of noncontrast scans. **A:** Multiple low-attenuation lesions are seen in an unenhanced image. **B:** Following intravenous contrast, most of the lesions have become inapparent. Care must be taken to insure that consistent contrast protocols are adhered to in oncology patients.

Abscesses. The diagnosis of liver abscess depends heavily on radiologic imaging, particularly US and CT scanning. [131] CT has become the screening procedure of choice for detecting pyogenic liver abscesses, with detection rates as high as 97%. The typical CT features of a pyogenic abscess include a mass hypodense with normal liver parenchyma, often with a contrast-enhanced rim. Gas collections are seen in approximately 20% of cases; internal septations may be seen. The mass usually is of higher density than simple cysts, but overlap in these densities can make it difficult to differentiate a simple cyst from an abscess. [132] Some malignant lesions can simulate liver abscess. Clinical correlation and aspiration are necessary to differentiate the entities. Fungal abscesses usually present on CT as multiple, small, low-density, nonenhancing lesions spread throughout the liver. Although CT is sensitive for detecting these lesions, active disease can evade CT visualization. [133] Almost always caused by *Candida albicans,* biopsy is the only way to establish the specific etiologic diagnosis. Amebic abscesses appear similar to pyogenic lesions as a low-density lesion, usually solitary, but with 20% of patients demonstrating multiple lesions. [134] Echinococcal infection (i.e., hydatid cysts) also appears as low-density cysts, often with internal septations and daughter or inclusion cysts, creating the appearance of a cyst within a cyst. Calcification may be seen in the cyst wall or septations.

Vascular Diseases Increasing awareness of imaging findings secondary to altered hepatic perfusion is the direct result of increasing use of helical CT. Hepatic perfusion disorders are related to:

- portal vein inflow obstruction
- hepatic venous outflow obstruction
- mediastinal or thoracic venous inflow obstruction
- focal liver lesions
- inflammatory processes
- normal anatomic variants
- altered hemodynamics following intrahepatic portosystemic shunts
- idiopathic processes.

General imaging characteristics are those of hyperattenuation on hepatic artery images becoming isodense on portal vein phase images. [135]

Portal vein thrombosis. Portal vein thrombosis can be detected on CT scans by visualizing decreased intralumenal portal vein attenuation values, accompanied on dynamic contrast scans with ringed enhancement from opacification and thickening of the vasovasorum. Chronically extensive collateral formation involving the porta hepatis is called cavernous transformation. [136] Intrahepatic portal vein thrombi produce slow flow peripherally resulting in paradoxical hyperattenuation in the periphery. The changes appear similar whether caused by a primary vascular abnormality with intrinsic thrombosis or secondary tumoral obstruction of the portal venous system, such as HCC. Zonal perfusion abnormalities may be seen in radiation hepatitis. The radiated liver demonstrates decreased perfusion in the portal venous phase. In fatty livers, the radiated portions appear to have normal attenuation because the damaged hepatocytes cannot undergo fatty metamorphosis. [137] Similar changes may be seen in hepatic parenchyma surrounding masses which alter the regional portal vein flow (see Fig. 155-13).

Hepatic venoocclusive disease. CT reveals a variety of findings in hepatic venoocclusive disease (i.e., Budd-Chiari syndrome). Although CT, MRI, and Doppler US can suggest the diagnosis, venography remains the gold standard for evaluation and diagnosis. Because of their noninvasive nature and optimal demonstration of vessel morphology and flow without the use of intravenous contrast, MRI and Doppler US are preferred for screening examinations of suspected patients. Noncontrast-enhanced CT findings include hepatomegaly with a hypodense parenchyma that spares the caudate lobe and perihilar regions of the left lobe. After intravenous contrast administration, the liver parenchyma demonstrates a mottled enhancement pattern. Initially, the greatest contrast enhancement is seen in the caudate lobe and the periportal regions of the left lobe, but a reversal of this pattern is seen on later images. Delayed images generally show a homogeneous enhancement pattern of the liver. Although direct visualization of thrombus within the hepatic vein or inferior vena cava can be demonstrated, in most cases, the hepatic veins are not visualized and may be the clue to the diagnosis. A similar mottled enhancement pattern is present in patients with chronic passive congestion, regardless of etiology. [138]

Hepatic trauma. Abdominal CT scanning makes nonoperative management of liver injury possible. Despite extensive clinical experience, it is not always possible to predict those patients in whom delayed complications may occur. [139] CT findings of liver trauma include foci of low or high attenuation indicating hemorrhage, usually in contact with some portion of the liver surface. If there has not been a prior peritoneal lavage, the presence of hemoperitoneum can be seen on CT as high attenuation intraperitoneal fluid accumulations. Low-attenuation collections diffusely surrounding portal venous branches (termed *periportal tracking*) can be indicative of periportal extension of blood or lymphedema, both sequelae of severe trauma. [140] However, there is not uniform agreement in the significance of this finding, as it can be seen in patients who have been overhydrated during a serious resuscitation. Because of the lack of specificity of this finding, caution is therefore recommended in relying on this finding as a sole indicator of traumatic injury to the liver. The presence of hemoperitoneum and clinical assessment of hemodynamic stability must be considered in conjunction with the CT scan before making an appropriate decision for nonoperative management.

Pancreas

Pancreatic CT requires thin collimation (3–5 mm), overlapped reconstruction, bolus injection of iodinated contrast material (best performed with a power injector), and rapid scanning to improve tumor detection, visualize relationships of peripancreatic vessels, and display findings three-dimensionally for optimized surgical and therapeutic planning (Fig. 155-16). Properly timed contrast-enhanced studies will image the pancreas when its parenchyma is maximally enhanced and the peripancreatic vessels bright white. [141] Accuracy in depicting variations in peripancreatic arterial anatomy, as well as clear depiction of vessel encasement, has resulted in the elimination of invasive angiography. [142], [143] Helical CT facilitates the use of dual-phase imaging where the earlier arterial-based enhancement of the pancreatic parenchyma can be isolated from the generalized organ opacification resulting from widespread distribution of the intravenous contrast dose. [144]

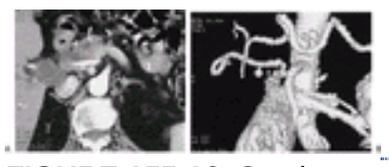

FIGURE 155-16. Carcinoma of pancreas and aberrant right hepatic artery. **A:** *Curved arrow* points to a low-attenuation mass in the head of the pancreas. A dense linear structure is seen posteriorly (*thick arrow*). **B:** A shaded-surface, three-dimensional rendering identifies this vessel as a replaced right hepatic artery arising from the superior mesenteric artery.

Anatomic Variants Pancreas divisum is the most common congenital anomaly of the pancreas. Although this anomaly most often goes unrecognized on CT scans, the pancreatic head can appear enlarged and simulate a mass lesion. Magnetic resonance cholangiopancreatography (MRCP) may be a simple noninvasive method for diagnosis. [145] Agenesis of the dorsal pancreatic duct is identified with CT by visualizing only the head of the pancreas without a body or tail portion, and the condition may be confused with a mass in the head of the pancreas with proximal pancreatic atrophy. [146] Annular pancreas is an anomaly embryologically formed by the ventral anlage of the pancreas as the duodenum rotates, mainly due to hypertrophy of the left ventral bud. According to the fusion pattern of the ventral and dorsal pancreatic ducts, annular pancreas can be divided into three types: type I, divisional annular pancreas; type II, branch annular pancreas; and type III, main duct annular pancreas. Final diagnosis is based on the evidence of ERCP combined with CT. The CT diagnosis is suggested by visualizing a soft tissue collar surrounding the descending duodenum. [147] Thin-section CT may allow visualization of the aberrant pancreatic duct.

Neoplasms

Adenocarcinoma. The major role of imaging in pancreatic carcinoma is detection of a suspected neoplasm and determination of potential surgical resectability. Nonresectability is defined as the presence of either major arterial encasement, hepatic (or other distant) metastases, or lymph node metastases. [148] Realistically, at the time of initial diagnosis, 50% of patients have distant metastases to the liver or peritoneal surface, and more than 80% of the remaining patients have locally advanced tumors. The most common CT finding in adenocarcinoma is a focal mass isodense with the pancreas on noncontrast images and hypodense after intravenous contrast administration. [149] This is more obvious using dual phase helical CT protocols. [150] Segmental pancreatic duct obstruction is a crucial finding and may be the only indicator of an obstructing mass. Detecting evidence of spread to the lymph nodes, around vascular structures, or to the liver may also assist in making the diagnosis. Potentially resectable lesions frequently show minimal findings. [151] Helical CT data provide angiographic information and are useful for surgical planning (Fig. 155-17).

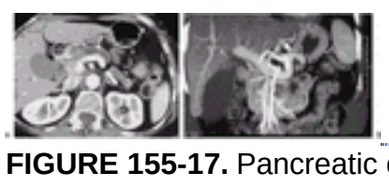

FIGURE 155-17. Pancreatic carcinoma. **A:** A soft-tissue attenuation mass is present in the neck of the pancreas (*straight arrow*). A dilated pancreatic duct is seen proximal to the lesion (*curved arrow*). The normal-caliber common bile duct can be seen immediately posterior to the mass surrounded by normally enhanced pancreatic parenchyma. **B:** Angiographic rendering from the same data set allows appreciation of narrowing without encasement of the superior mesenteric vein (*arrow*). Portions of the superior mesenteric artery and hepatic arteries are also incorporated in this projection.

CT angiographic assessment of pancreatic adenocarcinoma can be reliably accomplished with well-performed, thin-section single, or more optimally multislice CT. CT has been shown to be superior to catheter angiography [142], [143], [152] and slightly better than dedicated MRI [153] in evaluation of vascular invasion. The patterns of collateral formation around the peripancreatic branches of the superior mesenteric vein have improved prediction of tumor extension into this vessel. [154], [155] False-positive CT diagnosis of pancreatic adenocarcinoma has been reported in approximately 8% of patients; patients with focal chronic pancreatitis, metastatic lesions, normal pancreas, lymphoma, and islet-cell carcinoma account for most misdiagnoses. [156] In a large prospective series of 189 patients suspected with pancreatic adenocarcinoma the accuracy of CT in predicting nonresectability was 73%. The negative predictive value of CT (resectable by CT, resectable by pathology) was 28%. The positive predictive value of CT (nonresectable at surgery or by biopsy confirmation) was 89%. [157] Prediction of resectability has slightly improved with implementation of newer technology. [158]

Islet-cell tumors. The vast majority of islet-cell tumors are hypervascular lesions enhancing to a brighter level than enhanced pancreatic parenchyma. This helps differentiate these lesions from adenocarcinoma which are almost always hypodense. There are scattered case reports of cystic islet-cell tumors. [159] Although the nonfunctioning (endocrine quiescent) tumors typically attain large sizes, functioning islet-cell tumors are usually less than 2 cm in diameter. Multiple radiologic techniques can be used for tumor localization, including preoperative and intraoperative US, EUS, CT, MRI, radionuclide scanning, angiography, and venous sampling. [160] There are conflicting claims as to the relative accuracies of these procedures, and many of these investigations are difficult to justify because of their high cost, degree of invasiveness, or lack of precise anatomic information that is obtained. [161] If surgical resection of a neuroendocrine tumor is planned, intraoperative US is the best method to detect nonpalpable masses and to discern the relationship of the tumor to vital adjacent pancreatic ductal anatomy. The choice of preoperative imaging is more controversial, and depends on the clinical problem, local expertise, and availability of imaging techniques. US and contrast-enhanced helical CT using two-phase helical CT in both the arterial and the parenchymal phase are the most commonly used preoperative imaging methods [162] because of their relatively low cost and widespread availability. Radionuclide scanning with ^{111}In DTPA-D- octreotide may be valuable in patients with symptoms of tumor recurrence.

Cystic neoplasms. Most of these lesions are incidentally discovered. [163] The earliest pathological differentiation of the microcystic and macrocystic lesions remains a useful framework, but considerable variation and overlap is becoming more obvious. [164], [165] The ability of CT to definitively differentiate between these two classes of cystic neoplasms is limited. [166] Most published series suggest that while this diagnosis can be suggested on CT, surgical management must be considered as the standard of care. [167], [168] Cyst size is accurately determined by imaging and is the finding with the highest accuracy in classifying these lesions. [169], [170] Microcystic (cysts =1 cm) lesions are almost always serous. These lesions appear as focal masses with innumerable cysts interspersed within a dense fibrous honeycomb. Central calcification occurs in approximately 30% of the lesions. [171] Serous, microcystic cystadenoma is considered benign; there are scattered reports of metachronous adenocarcinoma arising at a distant site in the pancreas of the same patient. [172] Mucinous lesions present as macrocystic (=2 cm) unilocular cysts or as intraductal neoplasms. Peripheral soft tissue nodules, peripheral calcification (in <5% of lesions), and minimal septation aid in radiologic differentiation from microcystic adenoma. Scattered case reports of serous macrocystic adenomas reflect the lack of precision of absolute diagnosis from imaging alone. [173] Using thin collimation, mural nodules can be resolved in malignant lesions (Fig. 155-18). Imaging criteria of benign from malignant mucinous lesions is impossible. Most authorities recommend needle aspiration of the cyst fluid for definitive preoperative diagnosis. [174]

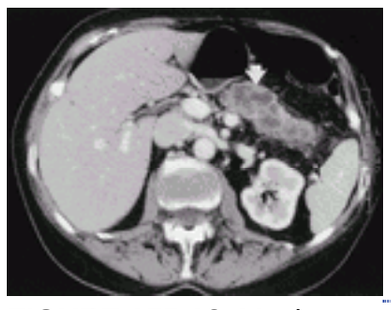

FIGURE 155-18. Mucinous cystadenocarcinoma. A focal lesion in the body of the pancreas (*arrow*) results in proximal pancreatic duct dilation. The lesion has soft-tissue attenuation peripheral projections typical of papillary-mucinous masses.

Intraductal mucinous pancreatic neoplasms are becoming increasingly recognized. [175] These lesions have a wide range of clinical presentation, from small foci of ductectasia with hyperplastic epithelium to frank carcinoma. The most frequent feature of intraductal papillary mucinous tumors is a lobulated multilocular cystic mass located in the uncinate process in contiguity with the dilated main pancreatic duct. [176] In some patients, a bulging papilla and papillary projections in the ducts, which were specific findings, were visualized on CT. When these lesions are branch type, under 2.5 cm, and have no solid components, clinical behavior appears nonaggressive. [177] Main duct-type lesions are virtually always malignant. The possibility of a sequential change from nonpapillary and papillary hyperplasia, via adenoma, to carcinoma in intraductal papillary-mucinous neoplasms associated with mucinous ductal ectasia has been speculated. [178]

Pancreatitis

Acute pancreatitis. Acute pancreatitis presents clinically and to imaging with a variety of forms. Interstitial edematous pancreatitis and necrotizing pancreatitis are the most frequent clinical manifestations; pancreatic pseudocyst and pancreatic abscess are late complications after necrotizing pancreatitis, developing after 3 to 5 weeks. [179] CT has become integrated into the evaluation of virtually all patients suspected of acute pancreatitis, and is considered mandatory in patients suspected of pancreatic necrosis. [180] CT scanning offers excellent anatomic and morphologic representation of the pancreas and peripancreatic tissue. [181], [182] The value of CT is based on its ability to show the presence and distribution of pancreas-related fluid collections (Fig. 155-19) and the presence or absence of pancreatic necrosis (Fig. 155-20). These two prognostic factors are evaluated in the Balthazar severity index. The presence of acute pancreatitis is graded as normal (grade A), gland enlargement (grade B), enlarged gland with peripancreatic edema (grade C), or single (grade D) or multiple (grade E) fluid collections. [183] Pancreatic necrosis on CT is recognized by regional nonenhancement on contrast-enhanced CT scans. [184], [185] The effect of degree of necrosis, combined with CT grading of the appearance of the gland and fluid collections, has greater prognostic implication than either manifestation alone. [186]

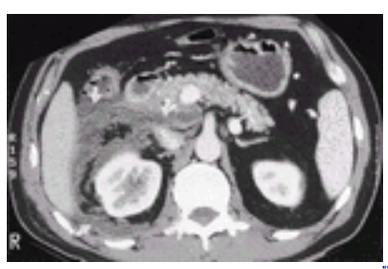

FIGURE 155-19. Acute pancreatitis. Peripancreatic fluid extends from the posterior aspect of the gland into the right pararenal and peritoneal retroperitoneal compartments (*arrows*). Note the uniform enhancement of the gland.

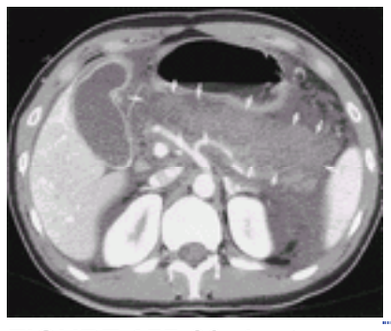

FIGURE 155-20. Acute pancreatitis with pancreatic necrosis. A “ghost” of the pancreas can be discerned (*arrows*). There is no normally enhancing tissue in this case of necrotizing pancreatitis.

The detection of necrosis is one of the primary goals of the imaging study. Intravenous contrast is required to make this assessment. In laboratory rats, intravenous iodinated contrast may increase cellular damage and potentially increase the severity of acute pancreatitis by promoting further necrosis and higher mortality. [187] These findings are not reproducible in other animal models. [188] Most clinicians believe that the importance of establishing the presence of necrosis justifies the preferred use of intravenous contrast. Complications from acute pancreatitis are present in the adjacent fat, [189] biliary tree, [190] peripancreatic veins [191] and arteries, [192] and adjacent bowel. [193] Pseudoaneurysm formation occurs in 2% to 5% of cases. Intravenous contrast and dynamic or helical scanning is required for detection; detection is improved with helical CT supplemented by angiographic renderings. Treatment is aimed at drainage of periarterial fluid collections and possibly embolization of the affected vessel. CT in patients with documented chronic pancreatitis reveals findings in over 90% of cases. These findings, in decreasing order of frequency, include: dilation of the main pancreatic duct, parenchymal atrophy, pancreatic calcifications, fluid collections, focal pancreatic enlargement, biliary ductal dilation, and alterations in peripancreatic fat or fascia. [194] Focal masslike chronic pancreatitis can result in pancreaticoduodenectomy for presumed neoplasm. In a Mayo Clinic series, [194a] 22 of 603 pancreaticoduodenectomies performed between 1956 and 1990 ultimately proved to be for chronic pancreatitis. When a patient with a hypervascular pancreatic mass has a history of alcoholism and pancreatitis, and normal serum levels of CA 19-9, mass-forming pancreatitis should be kept in mind as a differential diagnosis of pancreatic carcinoma. [195] Pseudocyst formation is a well-recognized sequela of chronic pancreatitis. Percutaneous catheter drainage of both infected and noninfected pancreatic fluid has a reported average success rate of 80% and a complication rate of about 15%. [196] Cyst “maturity” is difficult to diagnose by imaging. A visible wall and lack of change over several weeks indicate the patient is most likely a candidate for successful cystogastrostomy.

Biliary Tract

CT performs well at documenting the presence and extent of biliary tract malignant disease and, because of its use as a general survey tool in the abdomen varying forms of bile duct disease are frequently encountered on abdominal CT examinations. US and MRI are superior in the evaluation of calculous disease.

Using current CT technology and intravenous contrast enhancement, normal intrahepatic bile ducts may be visualized in over 40% of normal patients; these should not be confused with ductal dilation. [197] The extrahepatic bile ducts are identified normally in 80% of patients at CT and usually have diameters of 4 to 10 mm. [198] The extrahepatic biliary tree may be up to 10 mm in diameter in patients who have undergone cholecystectomy.

Comparative studies have shown that CT is comparable to US in its ability to detect biliary obstruction and generally exceeds US in the ability to predict the level and cause of biliary obstruction, although some controversy still exists. [199], [200] and [201] If a high degree of suspicion for obstruction exists and if malignancy is strongly suspected, CT is the examination of choice, because it allows the staging of extent of disease in a standard, reproducible fashion with excellent depiction of all regions surrounding the duct and distant sites as well.

Three-dimensional helical CT cholangiography following intravenous infusion of a biliary contrast agent that rivals direct cholangiography for displaying biliary tract anatomy and pathology has been reported. [202], [203] This technique, using postprocessing reconstructions (maximum intensity and shaded surface display projections), has not achieved widespread use due to the invasive nature of intravenous biliary contrast agents and the availability of noninvasive MRCP.

Congenital Anomalies Congenital anomalies of the biliary tract are diagnosed definitively with direct cholangiography, but CT and US can suggest their presence. Congenital anomalies of the gallbladder are rare, consisting mostly of positional variants. The gallbladder can be seen in the left abdomen or in suprahepatic, intrahepatic, and other rare locations. CT can identify these anomalies if the gallbladder is not seen in its usual location at US. Congenital cystic diseases of the biliary tree, including choledochal cysts, choledochoceles, [204] and Caroli disease, can often be diagnosed by demonstrating the dilated biliary tree with appropriate patterns of dilation. Choledochal cysts demonstrate focal dilation of the extrahepatic bile ducts that may be mild and simulate a dilated duct or may be as large as 15 cm in diameter. [205]

Gallbladder Disease CT is an acceptable method of demonstrating gallbladder wall abnormalities. Thickening of the gallbladder wall, although nonspecific, can be a clue to underlying pathology. Unusual right upper quadrant abdominal calcifications can be clarified with CT. CT can document the location of these as stones within the gallbladder, within the gallbladder wall (e.g., porcelain gallbladder), or extrinsic to the gallbladder. CT is able to demonstrate gallstones in 74% to 79% of patients with gallstones; the remainder are isoattenuating with bile and cannot be delineated. [206], [207] This lower sensitivity limits the usefulness of CT in screening for gallbladder disease.

Neoplasms Gallbladder carcinoma is usually far advanced when patients are first seen by a physician, and the CT scans reflect these changes. “Early” gallbladder cancer has been classified on the basis of CT appearance. [208] In the more commonly seen advanced disease, total replacement of the gallbladder lumen by a soft tissue-density mass usually invading adjacent hepatic parenchyma characterizes this lesion. Less common appearances include focal gallbladder wall thickening or polypoid intralumenal masses, similar to those seen with US. [209] The extent of tumor can be depicted on CT in the common regions of spread, including liver, lymph node, and peritoneal metastases and invasion of adjacent organs such as liver and duodenum (Fig. 155-21).

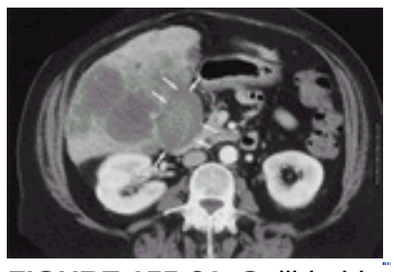

FIGURE 155-21. Gallbladder carcinoma. A bulky mass is present in the gallbladder fossa (*arrows*). The mass widely invades the hepatic parenchyma

The gallbladder can be secondarily involved by tumor from other organs. Direct invasion from adjacent organs is usually by means of porta hepatis metastases, most often from gastric or pancreatic carcinoma. Blood-borne metastases to the gallbladder are uncommon but can be seen most frequently in patients with melanoma.

Cholecystitis The most common CT findings in cholecystitis are gallbladder wall thickening (>3 mm) and cholelithiasis. [210] These findings are neither specific nor sensitive indicators and are also found with gallbladder carcinoma and hyperplastic cholecystosis. Other CT findings suggestive of the diagnosis include an increased attenuation of the bile (>20 Hounsfields) and loss of clear definition of the gallbladder wall (Fig. 155-22). Increased attenuation in the adjacent hepatic parenchyma is a useful indicator of acute inflammation. [211] With more advanced cases, streaky inflammatory changes infiltrating adjacent pericholecystic fat can be seen. Air within the gallbladder wall or lumen in the absence of a history of prior enteric anastomosis or sphincterotomy is virtually pathognomonic of complicated cholecystitis. [212] A low attenuation halo around the gallbladder may indicate edema or minimal fluid collections and is a useful clue in differentiating complicated cholecystitis from carcinoma on CT scans. [213]

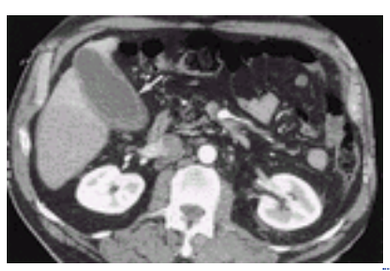

FIGURE 155-22. Acute cholecystitis. The gallbladder wall is thickened (*arrow*). Increased density is present in the hepatic parenchyma adjacent to the inflamed viscus.

Bile Duct Diseases

Neoplasms. Primary bile duct cancers take three forms, mass-forming, periductal infiltrating, and intraductal. [214] Hilar cholangiocarcinoma usually presents centrally near the porta hepatis producing obstruction of the main hepatic ducts and intrahepatic branches. Peripheral intrahepatic cholangiocarcinoma will present as a hypodense hepatic lesion with peripheral enhancement, biliary dilation, capsular retraction, and contrast enhancement on delayed images. [215] All hilar cholangiocarcinomas should be seen on well-performed arterial phase images, and most will be seen during portal venous phase imaging. A characteristic contrast CT feature of cholangiocarcinoma is the delayed retention of iodine diffusely throughout the tumor during equilibrium phase imaging at 8 to 20 minutes following contrast initiation. [216] This retention is seen in tumors with a marked fibrous stroma and can be helpful in delineating the extent of hilar cholangiocarcinoma and in differentiating from HCC. [217] CT is useful in the preoperative assessment of the extent of disease and resectability of cholangiocarcinoma; CT sensitivity for resectability is approximately 60%. [218]

Inflammatory diseases. Acute cholangitis is usually found in patients with underlying biliary tract obstruction. Most such patients demonstrate dilated intrahepatic and extrahepatic bile ducts. Suppurative material within the bile ducts may be seen on CT images as high attenuation debris within the duct lumen. Bile duct wall thickening may be present and appears diffuse and concentric, often demonstrating marked contrast enhancement. [198] Gas can be seen as low-attenuation collections within the biliary tree in cases of infection with gas-forming organisms. Acute suppurative cholangitis can progress to frank liver abscesses, demonstrated on CT scans as low-attenuation areas in the liver in contiguity with the biliary tree. The characteristic findings seen by means of cholangiography in primary and secondary sclerosing cholangitis can also be demonstrated on CT images. Intrahepatic duct stenoses, dilated peripheral ducts with no apparent connection to the central ducts, and irregular intrahepatic duct dilation with a beaded appearance are characteristic CT findings of sclerosing cholangitis. [219] CT findings in the extrahepatic ducts in this condition include duct wall thickening and irregularity, as well as duct wall enhancement after intravenous contrast. Primary sclerosing cholangitis should be considered as premalignant, with more than 15% of patients at risk. [220] Careful search for masses, delayed contrast enhancement, progressive biliary dilation, and thickening of the bile duct wall is required to recognize complicating cancer. [221] Other less common causes of infection can produce characteristic CT images. Recurrent pyogenic cholangitis (also known as Oriental cholangiohepatitis) is found in patients of lower socioeconomic status from Asia. The CT findings include marked intrahepatic and extrahepatic bile duct dilation, sharp tapering of peripheral extrahepatic bile ducts with loss of arborization, giant ductal calculi, and intraductal debris. Organisms such as CMV and *Cryptosporidium* can cause inflammation of the biliary tract in patients with AIDS, resulting in changes on CT, US, and cholangiography similar to those of sclerosing cholangitis, with bile duct wall thickening, multiple strictures, and duct wall contrast enhancement. [222]

Choledocholithiasis. The noninvasive diagnosis of common bile duct stone disease has been simplified with the development of MRCP which has a sensitivity and specificity similar to ERCP. [223] Most patients with obstructive jaundice will undergo US, with CT used as a problem-solving modality. [224] The reported CT sensitivity for common duct stone detection varies from 45% to 90%. [225], [226] The ability to use thin collimation and overlapping image reconstruction available with volume data acquisition of helical CT scanners improves stone detection rates. [227] Noncontrast CT may improve the recognition of choledocholithiasis to more than 90%. [228] The higher figures reflect series with selected patient populations, often patients with known dilated ducts in whom it is easier to visualize stones. The CT appearance of common duct stones parallels that of gallstones and depends on the chemical composition for each stone. Densely calcified stones can be seen as high-attenuation structures within the duct lumen, but most duct stones are isoattenuating with soft-tissue or bile on CT scans. [229] Several processes can simulate duct stones on CT, and care must be taken to avoid misdiagnosis. Critically placed pancreatic calculi, oral contrast in an adjacent duodenal diverticulum, residual contrast material from a prior cholangiogram, and papillary ductal neoplasms can simulate choledocholithiasis.

PERITONEUM

Most gastrointestinal diseases for which CT has use have associated peritoneal findings. Many of these are subtle; recognition of abnormalities requires a systematic evaluation of the peritoneal structures based on understanding of the spread of disease processes within the abdominal cavity. The majority of ligaments and mesenteries in the abdomen are formed from remnants of the ventral and dorsal mesenteries, which suspend the primitive gut. The pelvic ligaments are mainly formed by reflections of peritoneum over the pelvic organs or structures. The mesenteries and ligaments form the boundaries of the peritoneal spaces. Accurate localization

of fluid collections and detection of neoplasms is facilitated when the pathway of spread through adjacent ligaments and mesenteries is understood. [230] For purposes of radiologic diagnosis, the peritoneal cavity can be thought of as an “end organ,” in that the response to a pathological process is similar regardless of etiology.

Neoplasms

Radiologic appearances of the more common peritoneal abnormalities may be thought of in three basic patterns:

- solid but relatively well-defined masses
- cystic-appearing masses
- ill-defined or infiltrative processes.

The most common solid masses to affect these anatomic regions are secondary neoplasms. The various cystic-appearing masses (including cystic lymphangioma, cystic mesothelioma, teratoma, and loculated ascites) and infiltrating masses (such as peritoneal mesothelioma, retractile mesenteritis, desmoid, and carcinoid) must be differentiated on the basis of clinical findings and additional imaging findings (e.g., CT depiction of fat and calcium in teratomas and the radiating appearance of carcinoid). [231]

Peritoneal carcinomatosis is a common finding in patients with abdominal cancer. This form of metastatic disease is most frequent with primary lesions in the ovary, large bowel (including appendix), and stomach; however any primary malignancy may disseminate through the peritoneal cavity. Peritoneal lymphoma (not unique to AIDS patients), [232] peritoneal leiomyosarcoma, [233] and melanoma are reported (Fig. 155-23). Loculated ascites is the most common finding. Bowel wall thickening and irregularity with or without obstruction may occur, the most common site of involvement being at the rectosigmoid junction.

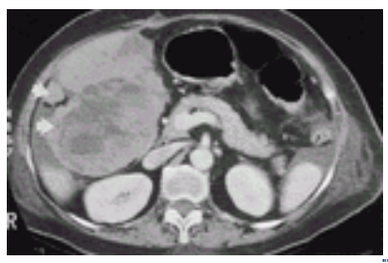

FIGURE 155-23. Metastatic leiomyosarcoma, peritoneum. A bulky mass is present in the Morison pouch (*posterior arrow*). A second mass is present just anteriorly (*anterior arrow*). Ascites is evident.

Tumor involvement of the omentum is visible as soft-tissue attenuation nodular or reticular densities within the peritoneal fat. This can coalesce into an omental cake. [234] Implants may be present without ascites and careful inspection of the omentum [235] and sites of peritoneal ligamentous attachments is necessary to detect these subcentimeter nodules. [236] Pseudomyxoma peritonei has a unique appearance of large, low-attenuation masses distributed within the peritoneal cavity. Calcifications may occur along the periphery. When pseudomyxoma peritonei is suspected, careful attention should be given to the appendix or ovaries as the potential source. Primary peritoneal tumors include a variety of benign mesenteric cysts, stromal neoplasms, lymphoid neoplasms (such as Castelman disease), mesothelioma, and desmoid tumors.

Nonneoplastic Peritoneal Diseases

Enteric inflammatory processes will cause local increased attenuation within the adjacent peritoneal fat; [237] however, one should not diagnose acute generalized peritonitis unless there is evidence of free fluid. In chronic peritonitis, such as in *M tuberculosis* peritonitis, one may see retraction of the parietal peritoneum from the abdominal wall and apparent enhancement of the peritoneal surfaces. [238] In all other ways, the findings may be identical to peritoneal carcinomatosis.

CT diagnosis in patients with an acute abdomen depends on recognition of free air or hemoperitoneum. CT has a higher sensitivity for the detection of pneumoperitoneum than plain x-rays, including upright chest films. [239] When subhepatic free air tracks into the porta hepatis intrahepatic reflections of the peritoneal cavity, it may resemble biliary air. [240] The prevalence of pneumoperitoneum in the postoperative period based on CT findings is greater than that previously reported. Fifty percent of patients may demonstrate free intraperitoneal air 6 days following surgery. [241] In the setting of blunt abdominal trauma, pneumoperitoneum does not necessarily indicate hollow viscus injury. It frequently is secondary to other etiologies, especially dissection of interstitial air from the chest. [242]

Hemoperitoneum is seen most frequently in trauma, [243] acute gynecologic disease, or in hemorrhage from neoplasm. [244] The CT attenuation of hemoperitoneum may be falsely low in the upper abdomen, because the blood products have settled to the dependent portion of the peritoneum (the “hematocrit effect”). In patients with blunt abdominal trauma, scanning through the lower pelvis, regardless of the site of injury, frequently detects hemoperitoneum in the cul-de-sac.

HERNIAS

The diagnosis of abdominal wall hernia is aided by CT. [245] Although most hernias are diagnosed clinically, they may be occult, particularly in obese patients, and a source of abdominal pain. A wide variety of abdominal wall hernias have been described including lumbar, spigelian, obturator, and perineal. When a hernia is suspected as the cause of bowel obstruction, it is important to be able to identify a transition zone within the hernia; if both limbs are dilated, then a “downstream” etiology should be sought. Complications of hernias include incarceration and strangulation. The CT findings are similar to findings seen in strangulated small bowel obstruction. [246]

CT findings of right paraduodenal hernia are encapsulation of small bowel loops in the right midabdomen with looping of arterial and venous jejunal branches behind the superior mesenteric artery. The findings of left paraduodenal hernia are less specific and involve encapsulation of bowel loops at or above the level of the ligament of Treitz with intermittent dilation. [247]

Traumatic diaphragmatic hernia may cause diagnostic difficulty. Left-sided diaphragmatic hernia is easier to diagnose than right. [248] CT can supplant barium studies and nuclear scans. Continuity of the diaphragm should be assessed to aid in differentiation of ectopic organ position due to hernia or diaphragmatic paralysis. Careful history of trauma should be sought.

FUTURE DIRECTIONS

The role of CT continues to expand into that of a “routine” procedure. CT colonography is at the verge of becoming an alternative to barium enema for screening colorectal cancer. As technology allows faster imaging times, improved reconstruction time, and interactive data, a CT scan begins to rival plain film examinations in terms of ease and speed yet surpasses them in terms of information. The increasing availability of volume data and more powerful workstations has resulted in increasing preoperative planning, and more aggressive uses of minimally invasive surgery.

Radiologists are continuously mindful of the cost of CT scanning applied over a larger population. However, the comprehensive nature of the examination, and higher level of diagnostic surgery at the completion of the test improves patient care and may decrease use of more expensive hospital and therapeutic resources.

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CHAPTER 156

Eric K. Outwater and Donald G. Mitchell

MAGNETIC RESONANCE IMAGING

Since the development of magnetic resonance (MR) imaging about 18 years ago, the applications for MRI of the gastrointestinal tract have expanded rapidly. MR imaging had been hampered somewhat in the evaluation of the abdomen primarily because of artifacts associated with respiratory and bowel motion. [1] The development of newer fast imaging techniques, however, has overcome these effects of motion, enabling examination of structures and organs that were previously not reliably imaged. These new techniques improve the results of MR imaging of the abdomen, particularly when compared with computed tomography (CT) scanning.

MR imaging has certain advantages over CT. CT relies on the single variable of x-ray attenuation for tissue contrast. All image contrast with CT relies on attenuation, and vascularity of structures can be inferred by the changes in attenuation imparted by intravenous contrast agents. Basic tissue differences seen include: air, calcification and bone, soft tissue, fat, and changes in soft tissue attenuation imparted by intravenous contrast agents. MR imaging, however, has several parameters which distinguish pathological tissues. These include T1, T2, lipid content, magnetic susceptibility imparted by metal ions, such as iron in the liver, and specific characteristics of flowing blood. In addition, various types of contrast agents, some of them targeted receptor agents, have been developed for imaging the abdomen. Finally, newer techniques such as diffusion and perfusion imaging have only begun to be developed for imaging the abdomen. Compared with CT, MR imaging has a range of tissue characteristics with which to build the image, and therefore has a greater potential for tissue characterization.

The risks of CT derive from the radiation dose and from risks of nephrotoxicity of intravenous contrast. Risks of radiation dose may be significant, particularly in younger patients. The probability of a radiation-induced cancer in a young adult undergoing CT of the abdomen and pelvis is about 1/2000 or greater. [2], [3], [4] and [5] The cost of this one-time radiation dose has been estimated at $30/mSv, or $300 per abdominal CT. [2], [5] These considerations lead one author to suggest that MR imaging in younger patients may be cost-effective compared to CT based on considerations of radiation risk alone. [2] Costs associated with irradiation of unsuspected pregnancies have not been assessed.

CT of the abdomen routinely employs iodinated contrast agents for delineation of vascular anatomy, and to increase the tissue contrast between tumors and normal organs. Iodinated contrast agents pose three dangers to patients. These are anaphylactic reaction, renal nephrotoxicity, and osmotic load. Certain patients, such as those with diabetic nephropathy or renal insufficiency from any cause, are at increased risk from renal nephrotoxicity from iodinated contrast agents. It is safer and cost-effective for these patients to be imaged with MR imaging. [6] MR imaging poses significant risk only to patients with certain types of implanted devices such as cochlear implants, cerebral aneurysms clips, or cardiac pacemakers. The risk of anaphylactic reaction associated with available gadolinium agents is very low. These agents have no known cross-reactivity with the iodinated contrast agents used for CT scanning, so these patients may be safely imaged with MR imaging.

In this chapter, we explain some of the tissue characteristics and basic principles of imaging and discuss specific applications and techniques in imaging. A glossary of the terms used in this chapter is supplied in Table 156-1.

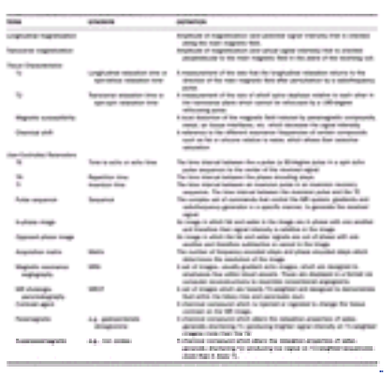

TABLE 156-1 Glossary of Magnetic Resonance (MR) Terms

BASIC PRINCIPLES

Differences in tissue contrast with CT are measured by units called Hounsfield units, which are a measure of x-ray attenuation. In MR imaging, the difference in tissue contrast (white, gray, black) depicted on the image is termed *signal intensity*. High signal intensity refers to structures that are white on the image and low signal intensity refers to structures that are dark on the image. Unlike Hounsfield units in CT, signal intensity units are arbitrary and have no real meaning as absolute numbers.

The terms *T1* and *T2* refer to specific tissue properties that describe the way protons (mostly in water and lipids) behave after being excited by a radiofrequency pulse in a strong magnetic field. [7], [8] Each type of tissue will show a typical T1 and T2. When the image tissue contrast is based mostly on the differences in T1 between the tissues, the image is termed a T1-weighted image (Fig. 156-1). T1 is termed the *longitudinal relaxation rate* and T2 is termed the *transverse relaxation rate* or *spin-relaxation rate*. Structures or fluids that are bright on T1-weighted images have a short (lower) T1 because of greater longitudinal magnetization, and images that are dark have a long T1. Structures containing no water or fat, such as bone or air, also will appear black on a T1-weighted image (see Fig. 156-1).

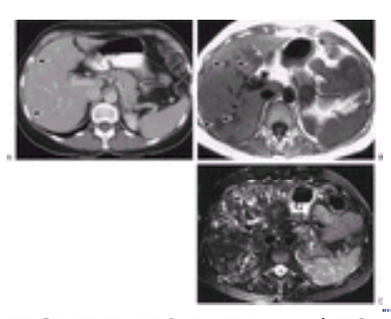

FIGURE 156-1. T1- and T2-weighted images of the abdomen in a patient with islet cell tumor of the pancreas. **A:** Computed tomography (CT) scan of the upper abdomen shows faint hypoattenuating lesions (*arrowheads*) in the liver. The pancreatic body is full but there is no tissue contrast between the parenchyma and the

pancreatic mass (*arrow*). **B:** T1-weighted image shows basic tissue contrast not dissimilar from the CT image. That is, the greatest tissue contrast is between fat and soft tissue and air. However, the intrinsic tissue contrast between the liver lesions (*arrowheads*) and the hepatic parenchyma is greater than the CT. Also, the tissue contrast between the pancreatic mass (*arrow*) and the surrounding pancreatic parenchyma is greater. This allows the discrete pancreatic mass to be visualized. **C:** T2-weighted image shows the liver as lower signal intensity (*darker*) than the hepatic metastases (*arrowheads*). The spleen (*S*) has higher signal intensity than the liver and the fluid in the stomach (*G*) is high signal intensity (*bright*).

The specific parameters chosen to acquire the image on the MR magnet system determine whether an image is T1-weighted or T2-weighted or some other type of image.[7],[8] TE and TR refer to the *echo time* and *repetition time,* respectively. They are parameters chosen by the MR operator to acquire a T1-weighted or T2-weighted image. The specific parameters chosen by the MR technician that determine exactly what kind of image is obtained are the *pulse sequence* or sequence. This sequence therefore describes a set of images covering a specific anatomic area, with identical imaging parameters such as TE, TR, and resolution. *T1-weighted images* have a short TE and a relatively short TR.

Images designed to show contrast based mostly on the T2 values of the tissues are called *T2-weighted.* Such images generally have a long TE and a long TR. Structures that are high signal intensity (white) on the T2-weighted image generally are fluid-containing structures such as cysts, intestinal contents, or gallbladder bile (see Fig. 156-1). Most tissues in the abdomen, such as liver, pancreas, and intestinal wall, have low signal intensity (they are dark) on T2-weighted images.[9] Structures that have a high fluid content or contain more fluid than normal tissues are brighter on the T2-weighted image.[9] Examples of this are cysts and hemangiomas of the liver. Structures such as malignant tumors contain a higher water content than normal tissues, and therefore show higher signal intensity than normal tissues (see Fig. 156-1). T2-weighted images, because of the long repetition time (2–3 seconds), usually take longer to acquire than T1-weighted images and therefore are somewhat more susceptible to motion artifact.

Within a strong magnetic field, protons (hydrogen atoms in water or lipid) have a physical property termed *spin* (angular momentum). These spins can be perturbed by a radiofrequency pulse at a specific frequency, the resonance frequency of the spins.[10],[11] and[12] After this excitation pulse or tip, the spins will emit a radiofrequency wave that can be registered by a receiver called the *receiver coil*. A radiofrequency pulse of sufficient magnitude to tip the spins 90 degrees away from the main magnetic field is referred to as a *90-degree pulse*. Similarly, a radiofrequency pulse that tips the spin 180 degrees is called a *180-degree pulse* or an *inversion pulse*. After tipping the spin 90 degrees, the spins emit a signal termed the *free-induction decay* (FID). This radiowave signal is the basis of the MR signal.

Because it occurs soon after the initial radiofrequency pulse, the FID is not received normally. The FID rapidly decays in signal amplitude because of dephasing of the spins. This means the spins precess out of phase relative to each other because of small differences in the magnetic field strength in the tissues and therefore have no net signal. This effect of signal amplitude decay, because of dephasing, can be overcome by the application of the 180-degree pulse after the 90-degree pulse in order to refocus the spins and bring the spins back into phase with a corresponding rapid increase in signal amplitude. This process is termed the *spin echo* and is used in spin echo sequences, which are the most frequently used sequences in MR imaging.[8] Spin echo sequences can be T1-weighted, T2-weighted, or intermediate-weighted. Intermediate-weighted images use a fairly short TE and a long TR, and are sometimes called proton density-weighted images. They generally have limited usefulness for abdominal imaging.

The signal from the spin echo is localized in space within the body by applying magnetic field gradients. These gradients change the resonance frequency and phase of the spins linearly across the body so that the source of the radio signal can be localized. These gradients are termed the *frequency-encoding* and *phase-encoding gradients*. A map of the location of the signal and the strength of the signal linearly across the body is the image. The complex radio signal received by the receiver coil from the body is decoded into the spatial location of the signals and the strength of the signals by a mathematic algorithm called the *Fourier transform*.

Another way of refocusing the signal after the 90-degree pulse is by the application of bipolar gradients, which are gradients that are switched on in one direction and then are switched on rapidly in the opposite direction.[13] This will cause the refocusing of the FID signal in a manner appropriate for spatial localization of the signal. These sequences are termed *gradient refocused echoes* or *gradient echo sequences.*[14] The advantage of gradient echo sequences is that they are acquired very rapidly, allowing acquisition within a breath-hold period.[15],[16] Gradient echo images designed specifically to image blood flow are termed *magnetic resonance angiography sequences,* because flowing blood appears bright on these sequences.

The resonant frequencies of water differ from the protons in lipid molecules such as adipose tissue or fat. Because of this difference in resonant frequencies, these two populations of protons in the body can be imaged separately, sometimes termed *chemical shift imaging.* The most frequently used method is the application of a radiofrequency pulse which saturates the signal selectively from fat or water.[17],[18] If this is performed at the resonance frequency of fat, it is called a *fat saturation sequence* or *fat saturation image*.

On gradient echo images, fat and water may be out of phase with one another and thus cancel their signal. Whether the fat and water spins are out of phase with one another depends upon the TE.[19] If fat and water are out of phase relative to each other, small volumes of tissue containing both fat and water (such as fatty infiltrated liver) will cancel their signal and appear dark on the image.[19] This type of image is called an opposed-phase image. In contrast, an in-phase image shows the lipid and water protons in the image as bright. In-phase and opposed-phase images are very sensitive to the presence of a small amount of lipid in tissues.[20] These sequences are used to detect fatty liver and the lipid that commonly occurs in adrenal adenomas.[21],[22],[23] and[24]

Magnetic resonance angiography (MRA) refers to a family of pulse sequences that image flow within blood vessels.[25],[26] These pulse sequences take advantage of the fact that flowing blood is bright (high signal intensity) on certain gradient echo sequences (Fig. 156-2). Blood flowing into a slice being imaged is fully unsaturated (i.e., has greater longitudinal magnetization) because it has not experienced prior excitation pulses. This effect renders the blood flowing into the slice as very high signal intensity on the image. Blood flowing through the slice shows less signal intensity because it experiences the excitation pulses used to acquire the data for the image. If thin slices are obtained continuously, preferably with the blood vessel of interest in cross-section, the resulting data can be reconstructed into a data set representing the entire volume of tissue, and then projected into images that resemble conventional angiograms.[25],[26]

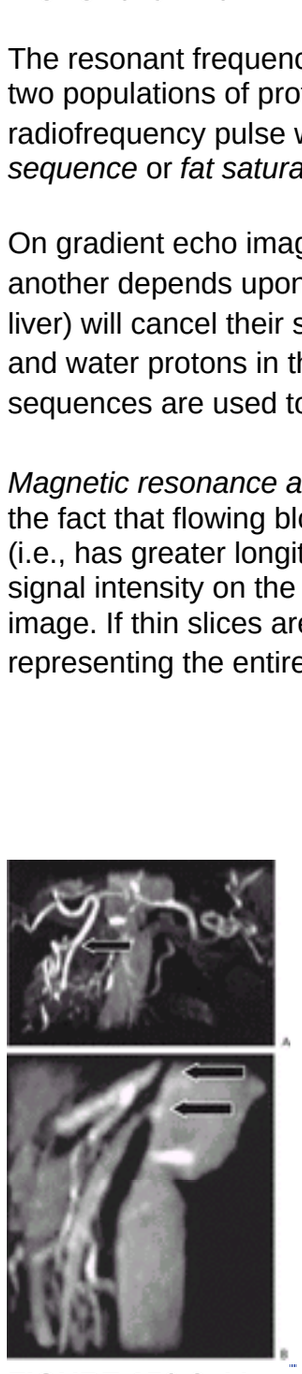

FIGURE 156-2. Magnetic resonance (MR) angiography with gadolinium-enhanced fast three-dimensional gradient echo sequences. **A:** Volume reconstruction images show multiple collateral vessels in and around the pancreatic head and duodenum (*arrow*). **B:** Projection MR angiogram in the lateral projection shows the stenoses of the celiac axis and superior mesenteric artery (*arrows*).

The principle described here is called *time-of-flight MRA.*[25],[26] There are other forms of MRA that rely on different physical principles such as phase contrast angiography, bolus tracking, black blood techniques, and three-dimensional, gadolinium-enhanced MRA (see Fig. 156-2).[25],[27],[28] The description of these

techniques is beyond the scope of this chapter. However these techniques have in common the principle of imaging flowing blood, and not necessarily imaging the vascular structures themselves. They all are susceptible to certain artifacts which give the impression of occluded or stenotic vessels such as slowly flowing blood, turbulent flow, and artifacts from adjacent structures such as metallic surgical clips. The basic goal of these sequences is to increase the contrast between the vessel and surrounding structures dramatically. Thus, the vessels are selectively displayed with markedly higher signal intensity than surrounding stationary tissue (see Fig. 156-2).

An MR technique that simulates MRA, but differs in principle, is the *magnetic resonance cholangiopancreatography* (MRCP) study.[29], [30] The goal of MRCP is to image selectively fluid within the biliary tree and pancreatic duct (Fig. 156-3). Very heavily T2-weighted images are used to achieve this effect. With the very long TEs that are used for these heavily T2-weighted images, virtually all solid tissue has low signal (is dark on the image). By virtue of its long T2, fluid within the biliary tree, the gallbladder, and the pancreatic duct retains signal intensity and therefore appears bright on the image. Like MRA studies, multiple projections in a manner similar to conventional cholangiography or endoscopic retrograde cholangiopancreatography (ERCP) can be obtained.

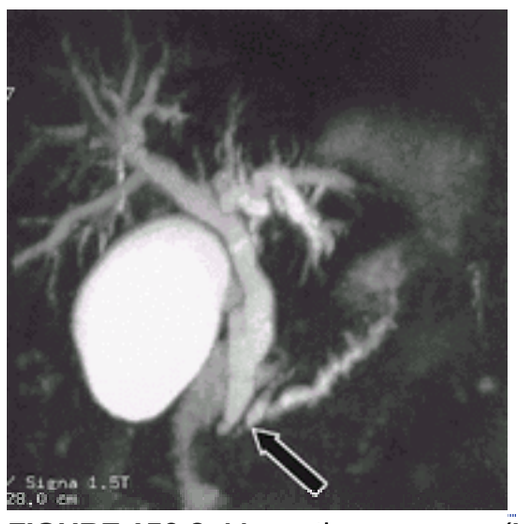

FIGURE 156-3. Magnetic resonance (MR) cholangiography in a patient with pancreatic carcinoma. Projection MR cholangiogram in the slight right anterior oblique view shows a dilated pancreatic duct with dilated side branches, as well as the dilated biliary tree. Both ductal systems are dilated into the pancreatic head and have distal strictures (*arrow*). The MR cholangiogram is made up of heavily T2-weighted thin slices which are then reconstructed into a volume projection by means of a computer algorithm (maximum intensity projection).

Current pulse sequences for imaging the liver are designed for very fast acquisition of images so that motion artifacts are limited or absent. These very fast images generally require improved (high performance) gradient systems. For T1-weighted images, fast gradient echo sequences are used (Fig. 156-4). T1-weighted images that acquire the whole liver as a volume (three-dimensional imaging) can be used for dynamic perfusion studies (see Fig. 156-4) or as MRA studies (see Fig. 156-2) or both.[31], [32] For T2-weighted images, subsecond pulse sequences have been developed so that motion-free images are obtained, whether or not the patient is holding his or her breath.[33] These sequences are called *HASTE, EXPRESS,* or *single-shot fast spin echo sequences,* depending on the manufacturer of the MR system. Echo planar imaging is not a new development but has been adapted to average clinical scanners with improved gradient systems and can also acquire subsecond images.

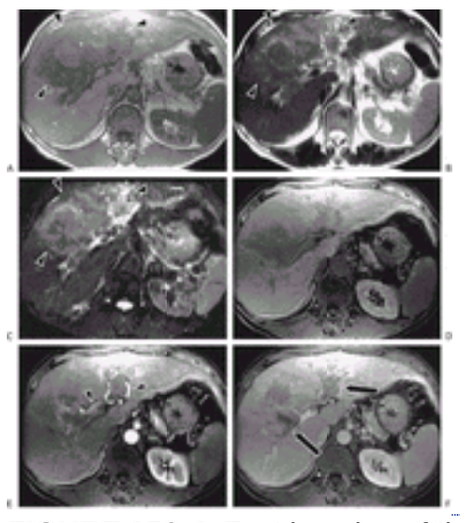

FIGURE 156-4. Fast imaging of the liver in a 56-year-old man with hepatocellular carcinoma. All of these sequences (**A–F**) image the entire liver in a breath-hold period. **A:** T1-weighted gradient echo image shows a large mass (*arrowheads*) in the liver with some areas of high signal intensity. **B:** T2-weighted single-shot fast spin echo image shows the mass as higher signal intensity than the rest of the liver. **C:** T2-weighted echo-planar image with fat saturation also shows the mass as high signal intensity. **D:** T1-weighted three-dimensional gradient echo image before injection of gadolinium chelate. **E:** T1-weighted three-dimensional gradient echo image during the arterial phase of gadolinium chelate enhancement shows hepatic arterial enhancement (*arrowheads*) and patchy enhancement of the tumor. **F:** T1-weighted three-dimensional gradient echo image during the portal phase of gadolinium-chelate enhancement shows tumor thrombus in the right and left portal veins (*arrows*).

Iodinated compounds are used as in CT scanning as injected *contrast agents* that help identify vascular structures and identify areas of abnormality within organs by differences in their vascularity. Injected contrast agents identify vascular structures and identify areas of abnormality within organs by differences in their vascularity.[34] Contrast agents also are available for MR imaging scanning that have functions similar to the iodinated contrast agents used in CT scanning. The most frequently used agent is gadopentetate dimeglumine (see Fig. 156-4).[35] This agent fills the arteries and capillary beds and rapidly diffuses into the interstitium and the sinusoids in the liver.[36] Because of this sequence of events, it is important to rapidly image abdominal organs to capture this temporal progression. In the liver, for example, it is advantageous to image an arterial phase, followed by a portal phase, followed by a delayed phase [37] (see Fig. 156-4). This imaging of the arterial and venous phases is important in the differential diagnosis of liver tumors. Specifically, hypervascular tumors such as hepatocellular carcinomas will be seen better on an arterial phase and may be invisible on delayed phases.[38], [39] and [40] Similarly, other liver tumors such as hemangiomas can be characterized accurately based on their temporal sequence of enhancement with the contrast agent [41], [42] and [43] (Fig. 156-5). Gadolinium-chelate enhanced images are important to detect and characterize vascular abnormalities (see Fig. 156-5).

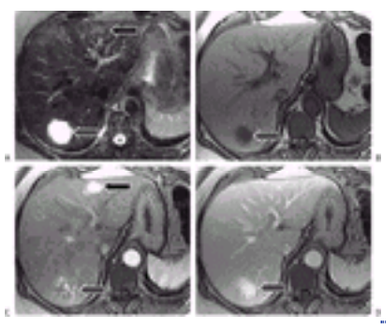

FIGURE 156-5. Contrast agents for imaging the liver: extracellular paramagnetic gadolinium agents. **A:** T2-weighted image of the liver shows a large, very hyperintense (bright) lesion in the posterior segment of the right lobe of the liver (*hatched arrow*). Faintly seen in the anterior aspect of the left hepatic lobe is a slightly hyperintense lesion, which is focal nodular hyperplasia (*black arrow*). Dynamic enhancement patterns of these tumors help in the differential diagnosis. Breath-hold, T1-weighted gradient echo images before (**B**) and after administration of an interstitial contrast agent (gadopentetate dimeglumine). Images acquired during the arterial phase of liver enhancement (**C**) and during the portal phase of liver enhancement (**D**) show differential enhancement patterns of these lesions. The hemangioma (*hatched arrow*) shows nodular peripheral enhancement in (**C**) with progression to complete contrast fill-in (**D**). On the other hand, the small focal nodular hyperplasia shows rapid arterial enhancement (**C**) denoted by the *black arrow* with rapid fading of the contrast to isointensity with the remainder of the liver (**D**).

Agents that are accumulated selectively by hepatocytes and that undergo some biliary excretion have been developed. [44], [45] and [46] These agents show prolonged liver enhancement on T1-weighted images and increase the signal intensity difference between metastases and liver. [44], [45] and [46]

Superparamagnetic iron oxide agents have been approved by the U.S. Food and Drug Administration (FDA) for imaging the liver. [47], [48] and [49] These agents are taken up by normal Kupffer cells in the liver and spleen. Most tumors, such as metastases, contain no Kupffer cells and therefore do not accumulate the agent. Thus, the signal intensity difference between liver and tumors is increased, making tumors more conspicuous on T2-weighted images (Fig. 156-6). Greater sensitivity to metastases using these agents has been reported. [48], [50], [51] and [52] Hepatocellular tumors, such as focal nodular hyperplasia and hepatic adenoma, take up these agents providing a valuable diagnostic tool for evaluating liver tumors. [47], [53], [54]

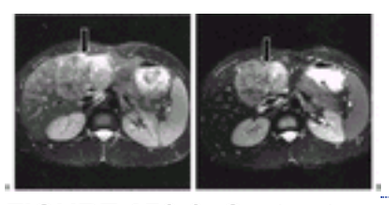

FIGURE 156-6. Contrast agents for imaging the liver: superparamagnetic iron oxides. **A:** T2-weighted image of the liver shows a hyperintense mass in the left lobe of the liver (*arrow*) in a 25-year-old man thought to have focal nodular hyperplasia. **B:** T2-weighted image performed after administration of superparamagnetic iron oxides, which are taken up by reticuloendothelial cells in the liver and cause T2-shortening (low signal intensity) in the liver. The signal intensity of the liver is lower in **B** than in **A,** as is the spleen to a lesser extent. The hepatic tumor is not lower signal intensity, which indicates that it is not likely to represent focal nodular hypoplasia. Resected specimen showed the fibrolamellar hepatocellular carcinoma. Low signal intensity bands in **A** and **B** represent fibrous bands typical of this tumor.

LIVER

Most research on MR imaging in the abdomen has concentrated on focal liver masses. Liver imaging is a frequent examination because many of the CT appearances and ultrasound appearances of focal liver masses are nonspecific, and because MRI has some intrinsic advantages over ultrasound and CT. One of these advantages is that intrinsic tissue contrast on MR imaging is high. What this means is that tumors can be easily detected and characterized without the addition of intravenous contrast materials. [55] Various tissues of liver tumors have different characteristics on T1- and T2-weighted sequences allowing better characterization with MR imaging. The chief disadvantage of MR imaging compared with CT and ultrasound has been that MR images are very sensitive to the presence of motion, such as the movement of the liver with respiratory motion. This disadvantage has been overcome in the last few years by the introduction of rapid pulse sequences for imaging the liver during suspended respiration.

Two characteristics of hepatic imaging of focal liver masses are important. The first is sensitivity, that is, the ability to detect liver lesions of clinical significance. For most patients, this means sensitivity for detection of metastases. Most studies have shown that MR imaging is superior to CT for the detection of metastases. [56], [57], [58], [59], [60] and [61] MR imaging is probably inferior for the detection of metastases when compared with CT arterial portography, [60], [62], [63], [64] and [65] however, it is more specific. [60], [66] MR imaging performed with specialized contrast agents, such as superparamagnetic iron oxides, improves the detection of metastases compared with unenhanced MR imaging. [50], [51] and [52], [58], [65], [67], [68] and [69] The second important characteristic of liver imaging is lesion characterization. The most frequent benign lesions in the liver are hemangiomas and cysts, and these must be differentiated from malignant lesions. MR imaging is superior to CT or ultrasound in lesion characterization because of the multiple tissue parameters that can be evaluated. [55]

One of the fundamental ways in which MR imaging can differentiate liver tumors is by virtue of their behavior on T2-weighted sequences. [55] Both hemangiomas, which are largely blood-filled, and cysts have a high fluid content and therefore a long T2. [42], [70], [71] Metastases and other solid liver tumors have a shorter T2. This difference in T2 values can be exploited by MR imaging to differentiate hemangiomas and cysts from metastases. [42], [70], [71] Specifically, T2-weighted sequences with a long TE (heavily T2-weighted sequences) show that hemangiomas and cysts maintain high signal intensity, and metastases are lower signal intensity. [70] Hepatocellular carcinomas tend to have even lower signal intensity on heavily T2-weighted sequences than metastases.

The enhancement pattern of hemangiomas and cysts also is distinctly different from that of metastases and other solid liver lesions. [72], [73] Because hemangiomas are largely cavernous vascular spaces, the perfusion of contrast agents through these spaces is slow. Typically, hemangiomas show enhancement starting with a peripheral, nodular pattern, and gradually fill in with the contrast agent (see Fig. 156-5). [41], [42] and [43], [74], [75] Metastases and hepatocellular carcinomas, on the other hand, enhance rapidly in the viable portions of the tumor or show poor enhancement in the nonperfused parts of the tumor (Fig. 156-7). Metastases from gastrointestinal primary tumors are often hypovascular in the sense that much of the tumor is made up of small areas of necrosis, desmoplastic reaction, and other hypovascular areas of the tumor.

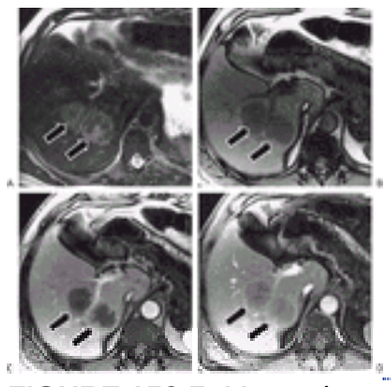

FIGURE 156-7. Magnetic resonance (MR) imaging characterization of malignant hepatic masses. **A:** T2-weighted image shows two metastases from a colorectal tumor in the right lobe of the liver (*arrows*). One can confidently predict that these represent malignant liver lesions based on their heterogeneity and the signal intensity lower than hemangiomas or cysts (compare the degree of signal intensity with the hemangioma in Figure 156-5). T1-weighted gradient echo images performed before (**B**) and after (**C, D**), intravenous injection of gadopentetate dimeglumine show patchy enhancement of these liver tumors with a pattern very different from the enhancement pattern shown for the hemangioma in Figure 156-5. The lack of "fill-in" of the masses with contrast on the delayed enhanced image in (**D**), should be compared to the delayed image of the hemangioma shown in Figure 156-5D.

Most types of malignant lesions in the liver cannot be distinguished from one another on MR imaging. They generally are lower signal intensity than the liver on T1-weighted images and higher signal intensity on T2-weighted images. [76], [77] and [78] An exception is hepatocellular tumors which tend to display characteristic appearances on MR imaging. [79], [80] and [81] The most frequent hepatocellular tumors are regenerating nodules in patients with cirrhosis, and they are slightly hyperintense on T1-weighted images and hypointense to cirrhotic liver on T2-weighted images [82], [83] (Fig. 156-8). In patients with cirrhosis, these nodules are separated by fine strands of fibrovascular tissue which are high signal intensity on T2-weighted images (see Fig. 156-8). [82], [84] MR imaging provides a method for directly visualizing these changes caused by cirrhosis. Larger regenerative nodules in cirrhosis, called *macroregenerative nodules*, are visible as hyperintense masses on T1-weighted images in patients with cirrhosis. These also generally are hypointense to the rest of the liver on T2-weighted images. [82] In addition, their blood supply is derived from the portal vein and enhances during the portal phase of enhancement on dynamic gadolinium-enhanced images.

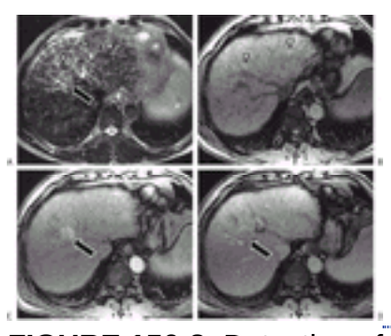

FIGURE 156-8. Detection of small hepatocellular carcinoma in a patient with chronic active hepatitis and cirrhosis. **A:** T2-weighted image shows an abnormal appearance of the liver. Small lower signal intensity nodules (*white arrowheads*) with a network of stranding throughout the liver caused by the fibrovascular septae separating the regenerative nodules of cirrhosis. A faint hyperintense lesion (*black arrow*) is seen adjacent to the middle hepatic vein. **B:** T1-weighted gradient echo image performed in a breath-hold shows faint hyperintensity to the regenerating nodules and cirrhosis. **C:** T1-weighted image performed during the arterial phase of liver enhancement shows rapid enhancement of the small hepatocellular carcinoma (*black arrow*). **D:** Rapid equilibrium of the contrast agent throughout the hepatocellular carcinoma and the remainder of the liver makes the lesion difficult to detect. Imaging during the brief window of arterial phase enhancement is important for the detection of small hepatocellular carcinomas because these malignancies adopt an arterial supply rather than a venous supply.

Hepatocellular carcinomas are frequent complications of cirrhosis. Similar to macroregenerative nodules, they often appear as hyperintense masses on T1-weighted images. [85] However, unlike macroregenerative nodules, they are typically hyperintense on T2-weighted sequences (see Fig. 156-8). [81], [85], [86] They also adopt an arterial supply and enhance during the arterial phase of enhancement on dynamic gadolinium-enhanced images (see Fig. 156-8). [38], [85], [87] Other helpful signs for the diagnosis of hepatocellular carcinomas are the presence of a pseudocapsule and venous invasion. Many hepatocellular carcinomas show steatosis providing a relatively specific diagnostic clue. MR imaging is more sensitive to small hepatocellular carcinomas in those patients with cirrhosis than CT scans. [87]

MR imaging has a greater ability to diagnose and characterize certain forms of diffuse liver disease. These include hepatic steatosis, iron storage disorders, and cirrhosis. Other liver disorders such as hepatitis, drug-induced disorders, alcoholic hepatitis, and Wilson disease do not have specific MR imaging features. Fatty infiltration of the liver from any cause can be detected with specific MR imaging sequences. As discussed previously under "Basic Principles," comparison of in-phase to opposed-phase T1-weighted images is a sensitive measure for the presence of lipid in tissue [22], [88] (Fig. 156-9). These sequences can definitively detect focal or diffuse fatty infiltration of the liver. [22], [89] These sequences also are important for evaluating focal abnormalities discovered on ultrasound or CT because focal fatty infiltration can mimic space-occupying lesions in the liver on these modalities (see Fig. 156-9). [20], [88]

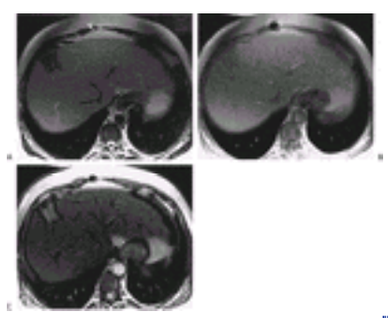

FIGURE 156-9. Opposed phase characterization of a mass suspected on computed tomography. **A:** T2-weighted image shows a masslike area of abnormality in the right lobe of the liver. **B:** In-phase image only faintly shows the abnormality. **C:** Opposed phase gradient echo image shows that the liver is diffusely low signal intensity (compare to in-phase image in **B**), indicating diffuse fatty infiltration. In addition there is slight loss of signal in the area (*arrow*) indicating that this represents a relatively fatty spared area.

Excess iron in the liver can result from primary iron storage disorders (hemochromatosis), iron overload states (e.g., thalassemia), or disordered iron absorption and deposition in cirrhosis (siderotic nodules). In hemochromatosis and iron overload states the iron is deposited diffusely in the liver, giving diffuse and profound low signal intensity on T2-weighted or gradient echo images. [90], [91] However, iron overload states cause severe splenic and liver low-signal abnormality, reflecting the distribution of reticuloendothelial macrophages which take up the excess circulating iron. [92], [93] The spleen is unaffected in primary hemochromatosis, thus allowing for differentiation from iron overload states. [90], [94] Siderotic nodules, which are common in cirrhosis from any cause, result in small nodular low signal intensity foci, distinct from hemochromatosis. [83]

BILIARY TREE

MR imaging has evolved to become a valuable imaging tool for the pancreas and biliary tree (Fig. 156-10).

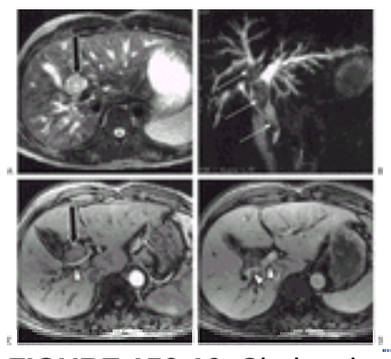

FIGURE 156-10. Cholangiocarcinoma. **A:** T2-weighted image shows a mass (*arrow*) which is hyperintense and lies anterior to the portal vein. The portal vein shows a flow void (black) because of flow within it. **B:** The magnetic resonance cholangiogram shows intrahepatic ductal dilation with separate obstruction of the right and the left ducts. There is an extensive fill-in defect within the common hepatic duct, right hepatic duct, and common bile duct (*arrows*) denoting the extent of the tumor. **C:** Arterial phase dynamic contrast-enhanced T1-weighted image shows the mass (*black arrow*) anterior to the right hepatic artery (*arrowhead*). The demarcation of this vessel argues against extension of the tumor to the porta epatis. **D:** Portal phase contrast-enhanced scan shows the right and the left portal veins (*arrowheads*), again without evidence of invasion. At surgery the ductal wall confined the cholangiocarcinoma. There was no evidence of invasion beyond the duct.

Applications in this area have been aided greatly by the developments in MRA and MR cholangiography, which allow selective visualization of the vascular structures and biliary and pancreatic ducts. MR cholangiography has been shown to be useful for the detection of biliary obstruction, calculi, and normal variants of biliary anatomy that have surgical importance. [30], [95], [96] and [97] The bile ducts appear as high signal intensity with this technique. Calculi appear as low signal intensity filling defects (Fig. 156-11). [98] Unlike CT scanning, where the attenuation of calculi is variable and may approximate that of surrounding soft tissue, calculi identified by MR imaging are almost universally low signal intensity on T2-weighted images. MR cholangiography has been shown to be accurate for the detection of calculi in the common bile duct. [98], [99]

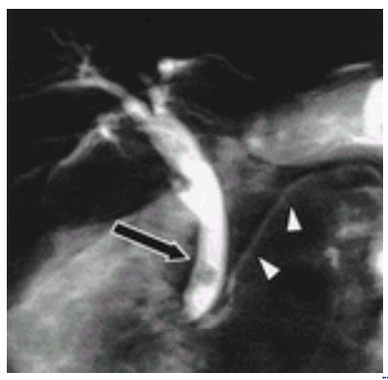

FIGURE 156-11. Magnetic resonance (MR) cholangiographic demonstration of choledocholithiasis. MR cholangiogram in the anterior projection shows a dilated common bile duct containing a calculus (*arrow*). Also displayed is the nondilated pancreatic duct (*arrowheads*).

Because of its ability to image bile ducts, vascular structures, and soft tissue of the hepatic parenchyma in one examination, MR imaging is useful for the detection of cholangiocarcinoma. Cholangiocarcinoma shows a characteristic pattern of enhancement on dynamic-enhanced images. It shows little uptake of contrast on the arterial and portal phases and fairly marked increase in enhancement on delayed images. [100], [101] With hilar cholangiocarcinomas (Klatskin tumors), MR cholangiography can show the multiple obstructed ductal segments. Unlike percutaneous transhepatic cholangiography, which requires separate injection of the different ductal segments if they are completely obstructed, MR cholangiography visualizes all the obstructive ductal segments simultaneously (see Fig. 156-10). Therefore, MR cholangiography can assist in the treatment planning for these tumors. [100], [101]

PANCREAS

The normal pancreas has greater signal intensity than liver on T1-weighted images which is most apparent on fat-saturated T1-weighted images. [102], [103] and [104] This characteristic aids in the detection of small tumors, particularly islet-cell tumors, as well as diffuse diseases such as chronic pancreatitis. Focal tumors of the pancreas appear as low signal intensity on T1-weighted images. [102], [103] Diffuse diseases of the pancreas, such as chronic pancreatitis, also cause diminished signal intensity on T1-weighted images. Typical T2-weighted images are of limited value in evaluating for pancreatic abnormalities because the most frequent types of pancreatic abnormalities (pancreatic carcinoma and chronic pancreatitis) have a prominent fibrotic component and therefore appear as low signal intensity on T2-weighted images, much like the normal pancreatic parenchyma. [102], [103], [105]

MR imaging depicts chronic pancreatitis as a change in signal intensity on T1-weighted images. T2-weighted images are helpful for showing peripancreatic edema and fluid collection such as pseudocysts in acute pancreatitis. On MRCP sequences, pseudocysts, and ductal abnormalities in patients with chronic pancreatitis are visualized. [106], [107] and [108] MRCP has been shown to be a fairly accurate technique for the detection of strictures, pancreatic duct filling defects from plaques or calculi, and ductal dilation which are the hallmarks of chronic pancreatitis. [107], [108] In addition, normal variants such as pancreas divisum and annular pancreas are identified. [106], [109] Fat saturated T1-weighted images are sensitive to the diminished signal intensity that occurs when the gland is replaced by fibrosis in chronic pancreatitis. [110], [111]

In contrast, pancreatic carcinomas show very little difference in signal intensity between tumor and normal pancreas on T2-weighted images (Fig. 156-12). Islet-cell tumors generally appear as high signal intensity on T2-weighted images. [112], [113] Most islet-cell tumors will be fairly conspicuous on T2-weighted images and will stand out from the remainder of the pancreatic parenchyma. Dynamic gadolinium-enhanced studies can be used to assist in identifying small tumors, particularly the insulin-secreting islet-cell tumors. [114] Insulinomas may be particularly difficult to identify on CT or ultrasound.

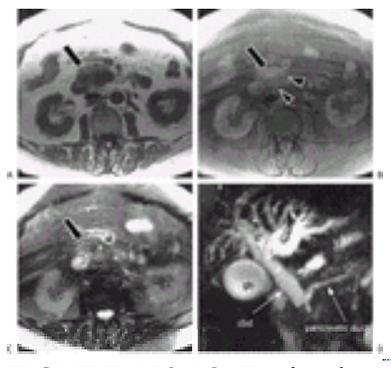

FIGURE 156-12. Evaluation of pancreatic carcinoma. T1-weighted spin-echo image (**A**) shows the pancreatic head (*arrow*), but a discrete mass is difficult to see. **B:** T1-weighted spin-echo fat-saturation image shows that the mass (*arrowheads*) has distinctly lower signal intensity than normal pancreas (*arrow*). T2-weighted fast spin-echo image (**C**) shows that there is heterogeneity to the mass (*arrow*), which has signal intensity characteristics similar to normal pancreatic parenchyma. **D:** Heavily T2-weighted cholangiogram shows dilation of the pancreatic duct in the body and tail. There is also marked dilation of the entire biliary tree because of distal common bile duct (*cbd*) obstruction. Strictures of the common bile duct and pancreatic duct in close proximity comprise the "double duct sign," which is predictive of pancreatic carcinoma.

Several studies have shown that some tumors other than pancreatic adenocarcinomas also are conspicuous on T2-weighted images. [115], [116] This is particularly true of those tumors that have a high mucin content which leads to very high intensity on T2-weighted images. These mucinous and microcystic tumors are recognized by characteristic morphology. [115] T2-weighted images show very high signal intensity of a cluster of septated cysts. MRCP images also show the ductal involvement of ductectatic mucinous tumors. [117]

INTESTINE

In the past, MR imaging had limited value in evaluating the bowel when compared with CT and conventional barium studies. This was mostly because motion of the bowel, either caused by respiratory motion or peristalsis, degraded the MR images. Newer techniques such as single-shot fast spin echo have corrected these problems to a great extent. [118] Antiperistaltic agents also are helpful for reducing bowel-related motion artifacts. Now MR imaging has great potential for evaluating the small bowel and colon, particularly for judging the extent of wall involvement with various pathological processes (Fig. 156-13). MR imaging has been shown to be valuable in assessing the severity and extent of ulcerative colitis and Crohn's disease. [119], [120] and [121]

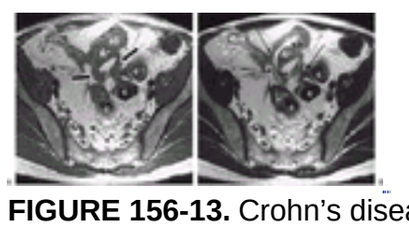

FIGURE 156-13. Crohn's disease with enterocolic fistulae in a 55-year-old man. **A:** Axial T1-weighted image shows a network of fistula tracts (*arrows*) from sigmoid colon (*c*) to ileum (*sb*). **B:** Axial T2-weighted fast spin echo image shows focal breaks in the bowel wall (*thin arrows*) at entrance of the fistula tracts. Note wall thickening in involved portions of the sigmoid colon.

MR imaging is useful for showing the extent of pelvic fistulae from any cause. [122], [123] and [124] T2-weighted images display the muscularis propria of the bowel as low signal intensity. Breaks in the muscularis from fistulae are seen as discontinuities in this layer. [122] Fistulae can be seen without the administration of contrast material (see Fig. 156-13). The high signal intensity of granulation tissue is seen surrounded by low signal intensity fibrosis. Because of the intrinsic soft tissue contrast of T2-weighted images, MR imaging has shown promise for staging gastrointestinal tract tumors, particularly rectal carcinoma. [125], [126] and [127] With high resolution, layers of the rectal wall can be visualized discretely. The muscularis propria appears as a very low signal intensity, while the submucosa appears as higher signal intensity. This has led to preliminary attempts to locally stage rectal carcinoma, with good results reported. [125], [126] and [127] MR imaging is an excellent method to evaluate the pelvis in general and to determine the local extent of tumors.

In addition, MR imaging can be used to evaluate for recurrent tumor after surgery or after radiation therapy. Radiation edema and radiation fibrosis are differentiated from tumor on T2-weighted images and dynamic gadolinium-enhanced images. [72], [73] Fibrosis appears as low signal intensity on T2-weighted images. Recurrent tumor shows nodules of higher signal tissue. [72], [73] Recurrent tumor enhances rapidly during dynamic injection of gadolinium while post-radiation changes enhance more slowly. [72]

IMAGING RECOMMENDATIONS

A wide range of MR imaging systems are available which, both in terms of hardware and software capabilities, provide clinicians with a corresponding range of diagnostic capabilities. In addition, there is a greater range of technical expertise and experience among radiologists in the performance and interpretation of MR imaging studies compared with CT. For these reasons, specific recommendations about the appropriateness of MR imaging examinations are difficult to make. Types of examinations that are well suited to MR imaging and which most radiologists in the community can perform are as follows:

Characterization of a liver mass identified on ultrasound, CT, or other study. Most frequently this would involve distinguishing hemangiomas or cysts from solid liver masses, although MR imaging diagnosis of other liver masses, such as focal nodular hyperplasia or hepatocellular carcinoma, is possible.
Evaluation of the complications of cirrhosis. Specific techniques can identify ascites, collaterals and varices, vascular occlusions, and hepatocellular carcinoma.
Identification of diffuse liver disease such as steatosis or iron deposition. Transfusional iron overload can be distinguished from hemochromatosis.
Identification of small pancreatic masses or exclusion of tumor in the large or bulbous pancreas.
Identification and characterization of a pelvic fistula, specifically perianal fistula and the degree of levator muscle involvement.
Identification of biliary calculi.
Evaluation of any disorders of the liver, biliary tree, or pancreas when the patient is allergic to iodinated contrast medium or is at risk from nephrotoxic effects from iodine (e.g., creatinine > 1.5, diabetic nephropathy).

Clinical situations in which CT may be more effective than routine MR imaging include:

- Routine evaluation of the abdomen and pelvis in a patient with cancer to search for metastases of the liver, lymph nodes, and so forth
- Initial evaluation of obstructive jaundice
- Evaluation of primarily peritoneal or bowel abnormalities
- Abdominal trauma.

FUTURE DIRECTIONS

In the future, refinements of MR imaging in the abdomen and pelvis will focus on more specific contrast agents for hepatic imaging, and the exploration of specialized pulse sequences for use in the abdomen.

The development of specific contrast agents in MR imaging has concentrated on liver-specific agents. A number of these agents are or will be commercially available and FDA-approved. These include superparamagnetic agents that are taken up by Kupffer cells in the liver. [128], [129] These help identify non-Kupffer cell-bearing

tumors such as metastases and most hepatocellular carcinomas with higher contrast, and therefore increased conspicuity, compared with nonenhanced images. Other liver-specific agents are under development, including agents with biliary excretion.

Relatively large volumes or amounts of the iodinated contrast agent are necessary for visualization on routine CT scans. By contrast, MR imaging is very sensitive to small amounts of the contrast agent. For this reason, the development of receptor-specific contrast agents is possible. [130], [131] Receptor-specific agents have been developed for imaging the liver [132], [133] and the pancreas. [131] Other types of superparamagnetic agents have been developed for imaging the lymph nodes. [134], [135] Injected intravenously, these agents are taken up by the lymph nodes over 24 to 48 hours. This may allow imaging of normal- sized, but tumor-bearing nodes because of defects within the contrast-enhanced lymph nodes. These contrast agents afford a degree of tissue characterization that CT scanning does not offer.

Forms of MR imaging used in the brain include diffusion imaging, perfusion imaging, functional imaging, and spectroscopic imaging and are being adapted to the liver and other organs. [136], [137] For example, preliminary evidence suggests that diffusion imaging may predict tumor response to chemotherapy in patients with liver metastases. MR spectroscopy can be used to probe the metabolic state of the liver in diffuse systemic and hepatic disease. [136]

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CHAPTER 157

>Uri Ladabaum and Satoshi Minoshima

POSITRON EMISSION TOMOGRAPHY

Positron emission tomography (PET) allows noninvasive imaging of physiological parameters within the body, such as metabolic activity and perfusion, using tracers labeled with positron-emitting radioisotopes. In contrast with imaging techniques that reflect primarily anatomic structure, such as computed tomography (CT) and magnetic resonance imaging (MRI), PET reflects biologic functions in tissue. In PET, the spatial distribution and concentration of administered positron-emitting compounds are used to generate cross-sectional images through the body that reflect pathological states or that elucidate physiological processes, leading to applications in clinical practice as well as in research. The principal clinical uses of PET in gastroenterology are in the area of oncology. The potential uses of PET in research are greatly varied. Functional imaging of the central nervous system with PET has been applied in research on visceral pain and perception.

PRINCIPLES AND TECHNICAL CONSIDERATIONS

PET relies on detecting the radiation produced by a systemically administered radiotracer labeled with a positron emitter and determining the distribution of the positron emitter within the subject under study. [1], [2] A small number of proton-rich radioisotopes can decay by emitting a positron (sometimes referred to as a positive electron or an antielectron), whose existence is fleeting. After emission, a positron travels a short path and ultimately combines with an ambient electron. This combination of a positron and electron undergoes a process referred to as annihilation, during which the masses of the positron and electron are converted into electromagnetic radiation in the form of two photons (gamma rays) of equal energy (approximately 511 keV) that travel in nearly opposite directions to each other (Fig. 157-1). PET scanners are designed to detect the photons produced by the annihilation of emitted positrons. Using a computer-assisted image reconstruction method, this information is ultimately used to construct images depicting the concentration of the positron emitter within the object of interest.

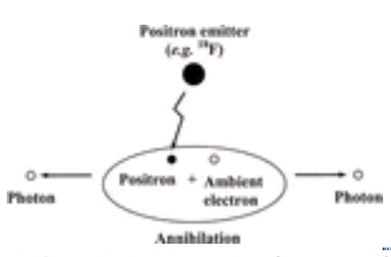

FIGURE 157-1. Positron emission and annihilation. Certain radioisotopes decay by emitting a positron, which travels a short path in tissue and ultimately combines with an ambient electron. This combination of particles undergoes annihilation, during which the masses of the positron and electron are converted into electromagnetic radiation in the form of two photons of equal energy that travel in nearly opposite directions to each other.

Radionuclide Tracers

The first step in PET imaging consists of labeling a suitable molecule with a positron-emitting radionuclide. Positron-emitting isotopes exist for carbon, nitrogen, and oxygen, three prominent atoms in biochemistry. The radionuclides ^{11}C, ^{13}N, and ^{15}O are commonly used in PET imaging in what is sometimes called “isotopic labeling.” [3] ^{11}C-methionine, ^{13}N-ammonia, and ^{15}O-water are examples of PET tracers labeled with these isotopes. In contrast, hydrogen, another important constituent of biochemical molecules, has no positron-emitting isotope. However, positron-emitting isotopes of halogens, most commonly fluorine 18 (^{18}F), can be substituted for a hydroxyl group in what is termed “analogical labeling.” [3] The PET tracer most commonly used in clinical applications is a glucose analog, 2-deoxy-2-[^{18}F]fluoro-D-glucose (^{18}F-fluorodeoxyglucose or ^{18}F-FDG).

Examples of radionuclides used in PET and their properties are listed in Table 157-1. [4] Due to the relatively short half-lives of these compounds, a small amount of tracer with high specific activity can be administered, translating into relatively low radiation exposure during the imaging session. [3] Because the quantity of tracer used is very small, there is no risk of pharmacological activity or chemical toxicity. Preparation of PET tracers requires the generation of a positron-emitting radionuclide followed by incorporation of the radionuclide into a molecule of choice. In theory, an unlimited variety of molecules could be labeled. Hundreds of tracers have been labeled with positron emitters, including simple molecules used to measure blood volume and blood flow, amino acids to study protein synthesis, fatty acids and sugars to study metabolic pathways, and specific receptor ligands including pharmaceuticals to map receptor distribution. Positron emitters are generated in accelerators, commonly small medical cyclotrons. [4] The positron-emitting atoms must be incorporated quickly into a tracer molecule as a purified, sterile, isotonic, pyrogen-free solution. [3] This underscores the importance of cooperation among experts in multiple disciplines including physics, chemistry, computer science, imaging, and medicine in the original development of PET and in the maintenance of an active PET program. The short half-life of many radiotracers mandates that they be produced on site, as with ^{15}O-water. However, the comparatively longer half-life of ^{18}F-FDG allows it to be produced at a regional facility, possibly in close proximity to several PET centers. Currently, ^{18}F-FDG is commercially available and distributed for PET imaging in the United States.

RADIONUCLIDE	HALF-LIFE (MIN)	% POSITRON DECAY	DAUGHTER ATOM AFTER DECAY
^{11}C	20.4	99.8	^{11}B
^{13}N	9.96	100	^{13}C
^{15}O	2.07	99.9	^{15}N
^{18}F	109.7	96.9	^{18}O
^{68}Ga	68.1	90	^{68}Zn
^{75}Br	101	76	^{75}Se (radioactive)

Adapted from ref. 4.

TABLE 157-1 Examples of Radionuclides Used in Positron Emission Tomography

2-deoxy-2-[^{18}F]fluoro-D-glucose (^{18}F-fluorodeoxyglucose or ^{18}F-FDG) The use of ^{18}F-FDG for PET applications in oncology rests on the high demand for

glucose by many tumors [5] and the similarities in the initial cellular metabolism of glucose and ^{18}F-FDG. [6], [7] Cells take up glucose and ^{18}F-FDG through specific transporters, and both sugars undergo phosphorylation by hexokinase inside the cell. Whereas glucose-6-phosphate can be a substrate for subsequent reactions in the glycolytic pathway, deoxyglucose-6-phosphate and fluorodeoxyglucose-6-phosphate cannot be metabolized further and are trapped in the cell, where the activity of the reverse enzyme glucose-6-phosphatase is limited. The contrast in ^{18}F-FDG uptake between tumors and metastases compared to surrounding tissue is often detectable.

Data Acquisition and Processing

The second key step in PET imaging comprises detection of photons emitted by the annihilation of positrons, followed by processing of these data to form cross-sectional images. The fact that positron annihilation produces photons traveling in nearly opposite directions serves as a sort of natural collimation and forms the basis for PET detector design and arrangement.

The typical PET scanner design includes multiple pairs of photon detectors oriented opposite to each other in a circular arrangement around a field of view where the subject is placed. [7] "Coincidence detection" refers to the method of accepting photons as indicative of a true positron annihilation only when a pair of photons is detected nearly simultaneously (current time windows are approximately 12 × 10 $^{-9}$ seconds) by an oppositely aligned pair of detectors (Fig. 157-2). Such coincident detection indicates that the photons originated somewhere near the imaginary line connecting the pair of detectors. Current PET scanners contain up to 48 adjacent rings of some 500 detector elements in each ring. Each detector is connected in coincidence with an arc of detectors on the opposite side of the ring. In addition, rings can be connected in coincidence with each other. Standard two-dimensional image acquisition treats individually each pair of adjacent rings, or each "slice" through the subject. More recently, three-dimensional acquisition has been developed, in which all possible coincident detections across rings are used in generating image volume data. Axial fields of view of current scanners are on the order of 15 to 16 cm, but whole body scanning can be performed clinically with axial movement of the scanner table.

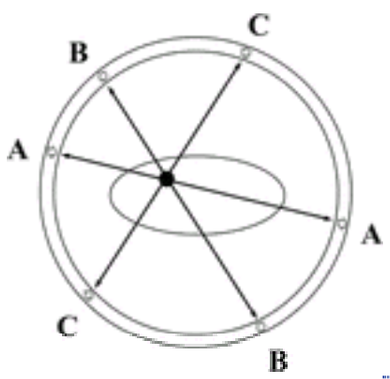

FIGURE 157-2. "Coincidence detection" is the basis for positron emission tomography (PET) detector design. "Coincidence detection" refers to the method of accepting photons as indicative of a true positron annihilation only when a pair of photons is detected nearly simultaneously by an oppositely aligned pair of detectors. In this schematic, accumulation of a PET radiotracer within a lesion (*black circle*) in the PET subject (*oval*) produces three pairs of coincident detections (*A, B* and *C*) in one ring of detectors surrounding the subject.

Corrections to coincident detection counts are often performed before image generation to account for a number of factors, including random coincidences at detector pairs and attenuation of photons inside the subject. [7] Attenuation refers to the scattering or absorption of emitted gamma rays within the subject's body. In present day scanners, attenuation correction is achieved by directly measuring tissue attenuation through the subject using an external radiation source equipped in the scanner. External rotating rods of germanium 68 (^{68}Ge) which are shielded when not in use, are commonly used to perform a "transmission scan" before PET tracer administration. This transmission scan is equivalent to a CT scan that visualizes differences in tissue attenuation within the body for diagnostic purposes.

Computerized mathematical algorithms can be applied for image reconstruction. These usually rely on filtered back projection, the same technique used in radiographic CT. The image produced by this technique represents the "solution" to the question of what spatial and temporal distribution of positron emitter can account for the pattern of coincident detections recorded by the PET scanner. An iterative image reconstruction method based on statistical modeling and segmented transmission data has become available for commercial scanners. This method generally produces better image quality, particularly when count rates are low. Due to a variety of factors, including the short path that positrons can travel before annihilation and the slight deviations in photon direction that can result from residual angular momentum in a positron or electron before annihilation, the intrinsic resolution of modern PET is on the order of 4 to 6 mm.

CLINICAL APPLICATIONS OF PET IN GASTROENTEROLOGY

In recent years, the recognized clinical uses of PET have expanded, particularly in the imaging of tumors. Regarding gastrointestinal cancers, the United States Health Care Financing Administration (HCFA) first announced Medicare coverage in 1999 for ^{18}F-FDG PET in the evaluation of patients with colorectal cancer and rising carcinoembryonic antigen (CEA) levels. In December 2000, HCFA issued a decision memorandum broadening the indications for ^{18}F-FDG PET covered by Medicare to include diagnosis, staging and restaging of non–small-cell lung cancer, colorectal cancer, esophageal cancer, lymphoma, melanoma, and head and neck (excluding thyroid and brain) cancers; myocardial viability studies; and presurgical evaluation of refractory seizures. [8] Additional applications in gastroenterology for which PET has been investigated include imaging of the pancreas, the liver, and neuroendocrine tumors. A survey [9] of 22 institutions found no adverse reactions in over 80,000 positron-emitting radiopharmaceutical administrations, highlighting the extraordinary safety record of PET.

Because anatomic information is often necessary in evaluating patients, including determining the potential resectability of tumor, ^{18}F-FDG PET cannot be considered a substitute for anatomic imaging. However, it can provide important additional information in a variety of clinical situations. In interpreting the values for sensitivity and specificity of PET in the settings discussed subsequently, it is important to recognize that these can be affected by a clinical trial's design, including patient selection criteria and the method for determining the reference gold standard (e.g., conventional imaging, histological confirmation, or clinical follow-up).

Colorectal Cancer

Increased ^{18}F-FDG uptake in hepatic metastases from colorectal cancer was demonstrated at a very early stage in PET technology development. [10] Later studies suggested that ^{18}F-FDG PET could be useful in differentiating recurrent rectal cancer from surgical scar or a nonmalignant mass. [11], [12] When anatomic imaging techniques could not reliably distinguish malignant from nonmalignant tissue, the enhanced glycolytic activity of malignant tissue could be detected by ^{18}F-FDG PET. Subsequently, ^{18}F-FDG PET has been investigated in the evaluation of patients with suspected colorectal cancer recurrence, as well as in preoperative staging of patients with known recurrence and in staging of primary disease. Figure 157-3 shows an example of ^{18}F-FDG PET in a patient with rectal cancer metastatic to the liver.

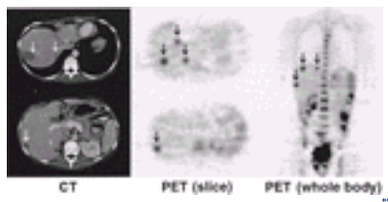

FIGURE 157-3. Imaging with contrast computed tomography (CT) and with ^{18}F-FDG PET in a patient with rectal cancer metastatic to the liver. Contrast CT shows at least three hepatic hypovascular lesions (*white arrows*). Corresponding positron emission tomography (PET) slice images show a total of four hepatic lesions (*black arrows*). Whole body PET shows at least three of the hepatic lesions (*black arrows*). A large focus of uptake in the pelvis is excreted ^{18}F-FDG in the bladder. Uptake is also visible in the right kidney next to the lumbar spine and in the descending colon.

Table 157-2 summarizes studies that have reported on the sensitivity and specificity of ^{18}F-FDG PET in suspected metastatic or recurrent colorectal cancer. [13], [14], [15], [16], [17], [18], [19], [20], [21], [22], [23] and [24] A metaanalysis of 11 studies yielded overall estimates (and 95% confidence intervals) for the sensitivity of whole body ^{18}F-FDG PET in recurrent colorectal cancer of 97% (95%–99%), and specificity of 76% (64%–88%). [25] For local/pelvic disease, the estimated sensitivity was 95% (91%–98%) and the estimated specificity was 98% (96–99.7%). For hepatic involvement, the estimated sensitivity was 96% (94–99%) and the estimated specificity was 99% (98–100%). While in many investigations ^{18}F-FDG PET images were interpreted visually, some authors have reported higher signal intensity in malignant compared to benign lesions using semiquantitative methods such as the standardized uptake value (SUV) or SUV corrected for lean body mass (SUL), [26] which normalizes signal intensity by radioactive dose injected and body weight, and the target-to-background ratio, which compares signal intensity within and outside a region of interest. [14], [17], [21], [24]

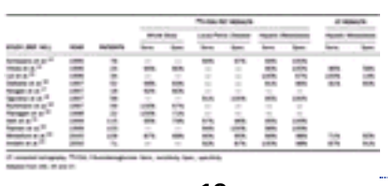

TABLE 157-2 ^{18}F-FDG Positron Emission Tomography (PET) in Suspected Metastatic or Recurrent Colorectal Cancer, with Comparison to CT for Hepatic Metastases

Patients may be suspected of having colorectal cancer recurrence on the basis of symptoms, physical findings, rising CEA levels, or anatomic imaging studies. In a study of 22 patients with previous colorectal cancer resection who had abnormal CEA but normal anatomic imaging studies, 15 patients were ultimately considered to have recurrent disease. Abnormalities were detected with ^{18}F-FDG PET in 17 patients, including the 15 with proven recurrence, yielding a positive predictive value of 89% and a negative predictive value of 100%. [20] The additional value of ^{18}F-FDG PET was examined in 103 patients with suspected or proven colorectal cancer recurrence who also underwent conventional imaging including CT. [22] In 12 of 60 patients considered to have resectable disease on conventional imaging, ^{18}F-FDG PET detected additional disease in 9 and excluded disease in 3, yielding additional diagnostic value in 20%. In 13 patients with inconclusive conventional imaging or isolated elevated CEA, ^{18}F-FDG PET was of additional diagnostic value in 62%.

Accurate staging is of paramount importance to guide potential surgical intervention, including resection of hepatic metastases. In the identification of hepatic metastases of colorectal tumors, ^{18}F-FDG PET has been compared with anatomic imaging as illustrated by studies listed in Table 157-2. [27] An important contribution of ^{18}F-FDG PET appears to be demonstration of hepatic and extrahepatic foci of disease not identified on conventional imaging. However, a study directly comparing ^{18}F-FDG PET results with surgical resection specimens found that ^{18}F-FDG PET detected only 25% of hepatic lesions smaller than 1 cm, compared to 85% of lesions larger than 1 cm. [28]

One study has assessed ^{18}F-FDG PET in the staging of primary colorectal cancer. [29] The sensitivity and specificity of ^{18}F-FDG PET for the primary tumor were 100% and 43%, respectively. For liver metastases, the sensitivity and specificity were 88% and 100%, respectively, compared to 38% and 97%, respectively, for CT. For lymph node metastases, the sensitivity and specificity of ^{18}F-FDG PET were 29% and 96%, respectively, compared to 29% and 85%, respectively, for CT. Thus, the use of ^{18}F-FDG PET appears greater in detecting distant metastasis than local nodal involvement.

The ultimate value of a diagnostic test is manifest in its impact on patient outcomes. Although no data are available comparing outcomes such as life expectancy or quality of life between patients studied or not studied with ^{18}F-FDG PET, several investigators have reported the impact of ^{18}F-FDG PET on medical decision making. In the metaanalysis by Huebner and colleagues, [25] it was estimated that ^{18}F-FDG PET affected management in 29% (95% confidence intervals, 25%–34%) of patients undergoing evaluation for recurrent colorectal cancer. Subsequent studies have reported similar results, with ^{18}F-FDG PET affecting management in 23% of patients undergoing evaluation for possible resection of hepatic metastases [28] and 21% of patients suspected of having metastatic or recurrent colorectal cancer. [24]

The abilities to monitor response to therapy or to assess a patient's candidacy for chemotherapy based on tumor characteristics are potentially of great clinical relevance. Decreases in ^{18}F-FDG uptake after chemotherapy have been shown to correlate with response to chemotherapy. [30], [31] In contrast, because inflammatory tissue may show similar glycolytic activity to that of tumor, ^{18}F-FDG PET may not be able to accurately assess the response to radiation therapy in the short term. [32], [33] However, one pilot study has suggested that ^{18}F-FDG PET might be of incremental value in selecting patients with rectal cancer for sphincter-preserving surgery after preoperative radiation and chemotherapy. [34]

In patients with unresectable liver metastases, PET has been performed with ^{18}F-fluorouracil before 5-fluorouracil chemotherapy to determine its potential role in predicting prognosis. [35] Overall, patients with higher values for ^{18}F-fluorouracil uptake were more likely to achieve stabilization of disease with chemotherapy, but trapping of ^{18}F-fluorouracil varied among metastases, even within the same patient. Quantitative modeling of ^{18}F-fluorouracil uptake has been investigated as a way to optimize treatment schedules for individual patients. [36] It remains to be determined whether these applications will find a role in clinical oncology.

Esophageal Cancer

Figure 157-4 shows an ^{18}F-FDG PET in a patient with a tumor at the gastroesophageal junction, and Table 157-3 presents the results of studies that have compared ^{18}F-FDG PET and CT for staging of esophageal cancer. [37], [38], [39], [40] and [41] The specificity of the two tests appears to be comparable in the evaluation of nodal disease, while the sensitivity of ^{18}F-FDG PET appears to be superior. For both techniques, detecting local nodal involvement is problematic. In one study, the overall accuracy in determining resectability was 88% for ^{18}F-FDG PET and 65% for CT ($P = 0.04$), but neither modality could assess the extent of wall invasion. [39] Differences in accuracy of similar magnitude have been found in other studies. [37], [40], [41] As with colorectal cancer staging, an important contribution of ^{18}F-FDG PET appears to be its ability to detect distant metastases not visible on anatomic imaging, and thus alter management. One group [42] has reported additional diagnostic value in 22% of patients, with upstaging in 15% and downstaging in 7%. Another study [37] reported a change in management based on ^{18}F-FDG PET results in 17% of patients.

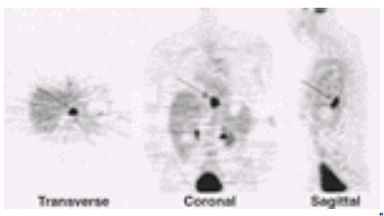

FIGURE 157-4. Whole body ^{18}F-FDG PET in a patient with a tumor at the gastroesophageal junction. A large focus of ^{18}F-FDG uptake due to the tumor (*arrows*) is visible in three orthogonal planes from the whole body PET scan. No regional lymph node metastases were identified, which contributed to pretreatment staging of disease. A large focus of uptake in the pelvis is excreted ^{18}F-FDG in the bladder. Uptake is also visible in both kidneys.

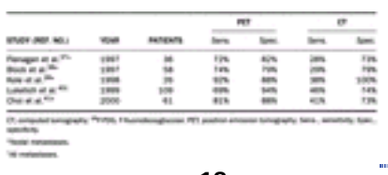

TABLE 157-3 ^{18}F-FDG PET Compared to CT in the Evaluation of Metastases in Esophageal Cancer

In staging potentially operable esophageal cancers, ^{18}F-FDG PET has been compared to the combination of CT and endoscopic ultrasound (EUS). [42] EUS demonstrated a higher sensitivity than ^{18}F-FDG PET for local nodal involvement (81% vs. 33%, $P = 0.027$). In evaluating regional and distant nodal involvement, ^{18}F-FDG PET was more specific than CT and EUS combined (98% vs. 90%, $P = 0.025$), with both testing stra- tegies having similar sensitivity (43% vs. 46%).

The combination of ^{18}F-FDG PET and ^{11}C-choline PET, which relies on uptake of choline by tumors for integration into phospholipids, has been reported to improve the accuracy of PET in staging esophageal cancer. [43] The ^{11}C-choline PET had a sensitivity of 88% for mediastinal lymph nodes, compared to 34% for ^{18}F-FDG PET. However, ^{11}C-choline PET demonstrated 0% sensitivity in the upper abdomen due to high ^{11}C-choline uptake by the liver, compared to 79% for ^{18}F-FDG PET. The combination of both modalities detected 85% of all lymph node metastases.

One study [40] has reported differences in survival based on stratification by initial ^{18}F-FDG PET results. Survival at 30 months was 60% in patients with localized disease compared to 20% in patients with distant disease ($P = 0.01$), suggesting that ^{18}F-FDG PET results may be of prognostic significance. It is not clear how much of this difference may be due to the effects of treatment.

Pancreatic Cancer

The HCFA decision memorandum of December 2000 did not include pancreatic imaging as an approved indication for ^{18}F-FDG PET under Medicare. However, pancreatic imaging is considered a recognized application of ^{18}F-FDG PET by some experts, as reflected by the recommendations of a German interdisciplinary consensus conference. [27]

Table 157-4 presents test performance data from studies that have evaluated ^{18}F-FDG PET in the diagnosis of pancreatic cancer and its differentiation from chronic pancreatitis. [44], [45], [46], [47], [48], [49], [50] and [51] Using ^{18}F-FDG PET may be beneficial in assessing pancreatic masses that are indeterminate for malignancy on conventional imaging. Beyond visual interpretation of images, semiquantitative analysis has demonstrated differences between the signal intensity, such as SUV, in tumor and chronic pancreatitis. [45], [46], [49], [51], [52] In one study, [50] ^{18}F-FDG PET suggested potential alterations in clinical management in 43% of patients.

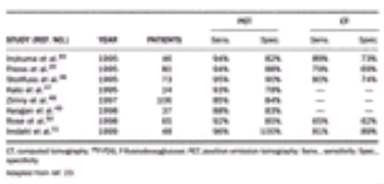

TABLE 157-4 ^{18}F-FDG PET Compared to CT in the Diagnosis of Pancreatic Cancer

Two important factors may affect the results of ^{18}F-FDG PET in pancreatic imaging. First, hyperglycemia can result in false-negative studies in pancreatic cancer. [48], [53] The cancer itself may cause endocrine insufficiency, and hyperglycemia results in greater competition by glucose against ^{18}F-FDG for uptake into cells. In patients with normal serum glucose, for instance, the sensitivity and specificity of ^{18}F-FDG PET in diagnosing pancreatic cancer were reported as 98% and 84%, respectively, compared to 63% and 86%, respectively, in hyperglycemic patients. [48] Second, inflammation can produce ^{18}F-FDG uptake as intense as that seen in tumor, either in the setting of pancreatic masses without clear evidence of acute pancreatitis, or during and after attacks of acute pancreatitis. [54]

Multiple imaging modalities can be used in evaluating patients with suspected pancreatic malignancy. One study [44] reported sensitivity and specificity for four different modalities in patients with suspected pancreatic cancer: 94% and 82%, respectively, for ^{18}F-FDG PET, 89% and 73%, respectively, for CT, 97% and 64%, respectively, for EUS, and 89% and 45%, respectively, for abdominal ultrasound. A subsequent study found sensiti- vity for detecting pancreatic cancer of 87% for ^{18}F-FDG PET, 53% for CT, and 93% for EUS. [55] Overall accuracy values (defined as area under receiver operating characteristic curves, with 1 reflecting a perfect test) for differentiating malignant from benign pancreatic masses have been reported for various imaging strategies. [53] Overall accuracy was 0.86 for ^{18}F-FDG PET, 0.93 for endoscopic retrograde cholangiopancreatography (ERCP), 0.82 for CT, and 0.95 for the combination of ^{18}F-FDG PET and ERCP. In the subgroup of euglycemic patients with contradictory or indeterminate CT and ERCP results, ^{18}F-FDG PET was accurate in 84% of cases.

The role of ^{18}F-FDG PET has also been studied for staging of metastatic disease. For lymph node staging in pancreatic cancer, one study [53] reported ^{18}F-FDG PET sensitivity and specificity of 49% and 63%, respectively, and for hepatic metastases 70% and 95%, respectively. As in colorectal cancer, ^{18}F-FDG PET may miss smaller metastases (<1 cm). A study designed specifically to assess the detection of hepatic metastases of pancreatic cancer found sensitivity of 90% and specificity of 91% for ^{18}F-FDG PET, compared to 69% and 100%, respectively, for CT, and 82% and 100%, respectively, for abdominal ultrasound. [56] In another investigation, the sensitivity of ^{18}F-FDG PET for hepatic and extrahepatic disease was 78% compared to 33% for CT. [55]

Monitoring patients with ^{18}F-FDG PET after therapy has been employed. [57], [58] Survival in patients without measurable pancreatic ^{18}F-FDG uptake 1 month after chemotherapy was improved compared to patients with residual ^{18}F-FDG uptake (319 compared to 139 days, $P = 0.034$). As in colorectal cancer, it remains to be determined whether ^{18}F-FDG PET will find routine clinical applications in determining prognosis or response to treatment, which is likely to depend on whether such determinations influence patient management.

Hepatic Imaging

The role of ^{18}F-FDG PET has been evaluated in hepatocellular carcinoma and metastatic disease to the liver. The sensitivity of ^{18}F-FDG PET for hepatoma is poor. In one study, [59] only 50% of sonographically detectable hepatomas demonstrated increased ^{18}F-FDG uptake, whereas 79% were detectable by CT. None of the well-differentiated tumors were visualized by ^{18}F-FDG PET, compared to 88% of the moderately or poorly differentiated tumors. Similar results were reported in a second study, [60] with ^{18}F-FDG PET showing a sensitivity of 55% in the diagnosis of hepatoma, compared to 90% for CT.

In contrast to poor results in the diagnosis of hepatoma, ^{18}F-FDG PET can be useful in evaluating metastatic disease to the liver. This application for staging specific primary gastrointestinal tumors has already been discussed. One study [61] of patients with various primary tumors and possible liver involvement found sensitivity of 97% and specificity of 88% for ^{18}F-FDG PET compared to 93% and 75% for CT (not statistically different). The use of ^{18}F-FDG PET yielded new information for 23% of patients, including demonstrating liver metastases in 17% when other imaging was equivocal or negative. In patients undergoing CT-guided fine-needle aspiration (FNA) of liver lesions, ^{18}F-FDG PET was positive for malignancy in 96% of cases in which malignancy was diagnosed by FNA, and was negative in 86% of cases where FNA was negative. [62]

The role of ^{18}F-FDG PET has been examined in the setting of hepatic lesions requiring further characterization. In a series of 110 patients with hepatic lesions 1 cm or larger on CT, all liver metastases from adenocarcinomas and sarcomas and all cholangiocarcinomas showed increased ^{18}F-FDG uptake values, whereas only 70% of hepatomas showed increased uptake. [63] All benign lesions had poor uptake, and abscesses showed equivocal or increased uptake. A series of 78 patients with various liver tumors has confirmed that ^{18}F-FDG uptake tends to be lower in hepatocellular carcinoma compared to metastatic disease or cholangiocarcinoma. [64] Case reports have suggested that ^{18}F-FDG PET might be useful in detecting cholangiocarcinoma in patients with primary sclerosing cholangitis. [65], [66] Monitoring the response to chemoembolization is another use of ^{18}F-FDG PET. [67]

Neuroendocrine Tumors

The experience with PET in detecting primary and metastatic neuroendocrine tumors is limited. Carcinoid tumors synthesize serotonin from 5-hydroxytryptophan (5-HTP). An early study with 7 patients suggested that hepatic metastases from carcinoid tumors could be imaged with ^{11}C-5-HTP PET. [68] In 22 patients with neuroendocrine tumors, ^{11}C-L-dihydroxyphenylalanine (^{11}C-L-dopa) PET detected tumor in 50%. [69] In those imaged with both ^{11}C-L-dopa and ^{11}C-5-HTP, results were similar. One study has reported similar overall results for ^{11}C-5-HTP PET and CT in 16 patients with gut carcinoids and 2 with endocrine pancreatic tumors. [70] Although ^{11}C-5-HTP detected more lesions than CT in 10 patients, it is not clear that this is of clinical significance.

Somatostatin receptor scintigraphy (SS-R) is commonly used for imaging neuroendocrine tumors. In 15 patients with neuroendocrine tumors, SS-R was found to be more useful overall than ^{18F}-FDG PET, although SS-R was negative and ^{18}F-FDG PET was positive in 2 patients with less differentiated tumors. [71] A second study with 16 patients found an overall sensitivity of 85% for SS-R in neuroendocrine tumor imaging. Even though it was able to image aggressive tumors, ^{18}F-FDG PET did not seem to provide much additional value. [72] Notably, in this study, CT scans were abnormal in all patients with abnormal ^{18}F-FDG PET and in all but one patient with abnormal SS-R.

The difficulty in detecting pancreatic islet-cell tumors is highlighted by a study in which the sensitivity of ^{18}F-FDG PET was 53%, compared to 53% for ultrasonography, 50% for CT, and 53% for magnetic resonance imaging (MRI). [73] In all cases ^{18}F-FDG PET was negative or equivocal where the other modalities were negative.

APPLICATIONS OF PET IN GASTROENTEROLOGY RESEARCH

The ability of PET to use a theoretically unlimited variety of molecules radiolabeled with different positron emitters to image biologic processes in vivo opens numerous possibilities for PET applications in research. The following studies illustrate the broad range of questions that have been addressed in PET research studies relevant to gastroenterology. An anticolorectal carcinoma monoclonal antibody labeled with copper 64 (^{64}Cu) (radioimmunoPET) has shown 71% sensitivity for imaging tumor foci. [74] ^{11}C-methionine uptake by the pancreas has been investigated by PET as a possible reflection of pancreatic exocrine function. [75] Biliary bicarbonate secretion in humans has been studied after intravenous injection of sodium ^{11}C-bicarbonate. [76] In cholestatic conditions, an impaired response to secretin was observed. Glucose use has been studied with ^{18}F-FDG PET in a patient with hepatoma and hypoglycemia. [77] Model parameters of glucose use did not differ between tumor and liver, and the authors concluded that hypoglycemia was attributable to insulin-like growth factor II-like proteins causing glucose uptake by skeletal muscle and suppression of glucose production. The mechanism and kinetics of percutaneous ethanol injection of hepatomas has been studied with ^{11}C-ethanol. [78]

Neurological abnormalities can accompany various forms of liver disease. The role of ^{18}F-FDG PET of the brain in patients with Wilson disease has demonstrated decreases in regional cerebral glucose metabolism in the cerebellum, striatum, cortex, and thalamus compared to controls. [79] In this study, striatal metabolism was most affected in those patients who had received therapy for the shortest time. Altered cerebral blood flow and metabolism have been observed in patients with severe liver disease and low-grade encephalopathy. [80] Normalization of brain glucose metabolism has been observed following liver transplantation. [81] In patients with hepatic encephalopathy, central benzodiazepine receptor binding has been studied with ^{11}C-flumazenil, with significant increases shown in the thalamus, cerebellum and pons, but not in the cortex or whole brain, when modeling corrected for altered hepatic metabolism. [82]

In animal studies, liver regeneration has been assessed with ^{11}C-thymidine PET, [83] the trafficking of radiolabeled tumor cells in vivo has been assessed with PET, [84] ^{11}C-octanoate PET has been investigated for assessing liver function, [85] and ^{11}C-labeled cholecystokinin receptor antagonists have been synthesized and evaluated. [86]

Functional Brain Imaging and Visceral Sensation

One application that has received intense interest in recent years is functional imaging of the central nervous system as a tool for studying visceral sensation, particularly in the context of visceral hypersensitivity in functional gastrointestinal disorders. [87] In brain activation studies with PET, regional cerebral blood flow (assessed with ^{15}O-water) during a task of interest is compared to regional cerebral blood flow during baseline to identify cerebral regions with increased activation during the particular task. This widely used technique is based on the assumption that regional blood flow is coupled with regional neuronal activity under physiological conditions. In visceral sensation research, the stimulation “task” has consisted most often of visceral distension with a balloon.

In an early study, [88] rectal distension in healthy volunteers led to activation in the pre- and postcentral gyrus and thalamus. In the first study of functional brain imaging in patients with functional gastrointestinal disease, rectal distension in controls but not patients with irritable bowel syndrome (IBS) produced activation of the anterior cingulate cortex, a region implicated in the affective response to pain. [89] A subsequent study with functional MRI reported activation in the anterior cingulate cortex (ACC), prefrontal cortex, insular cortex, and thalamus in most IBS and control subjects, with increased ACC activation in IBS subjects compared to controls. [90] The reasons for the conflicting results in these studies remain to be fully explained. Compared to female subjects with IBS, male subjects with IBS have demonstrated increased activation in the insula, considered a visceral sensory cortex. [91] In this study, insular activation was associated most strongly with distending pressure, whereas anterior cingulate activation was associated more with subjective discomfort.

The cerebral representation of esophageal distension has also been studied with ^{15}O-water PET. [92] During nonpainful stimulation, activation was seen bilaterally along the central sulcus, in the insula, and in the frontal/parietal operculum. Painful stimulation led to greater activation in the same regions, with additional activation of the right anterior insula and the anterior cingulate gyrus, leading the authors to suggest that these latter structures are involved in esophageal pain processing.

Cerebral activation has been investigated with ^{15}O-water PET during gastric distension producing bloating, pain, and nausea. [93] Paralleling increases in distension stimulus and symptoms, progressive increases in activation were observed in the thalami, insulae, anterior cingulate cortex, caudate nuclei, brainstem periaqueductal

gray matter, cerebellum, and occipital cortex. This activation pattern showed important similarities to activation patterns reported in studies of painful somatic stimulation. Due to the high correlation among symptom scores for bloating, pain, and nausea, the specific regions activated during the experience of each individual sensation could not be determined.

FUTURE DIRECTIONS

PET has been called the most sensitive and specific means for studying, through imaging, molecular processes in humans in vivo. [94] The development of a microPET scanner for small animals, combined with rapidly growing experimental capabilities in molecular biology, presents the opportunity to carry out molecular imaging assays in vivo, including studies of gene expression. [95] The kinetics of ^{18}F-labeled oligonucleotides, for instance, have been studied with PET in baboons. [96]

In the clinical arena, the number of clinical PET facilities has grown over time, and PET is becoming an essential diagnostic tool for the management of certain patients with cancer. Given the ability to produce ^{18}F-FDG off-site and its commercial availability in the United States, not all clinical PET centers will require an on-site cyclotron. This may facilitate further the widespread use of PET. Newer tracers, including ^{18}F-labeled radiopharmaceuticals that can be prepared and delivered commercially owing to their relatively long half-life, may ultimately find clinical applications. [97]

The precise role of functional imaging with respect to anatomic imaging remains to be clarified in many clinical conditions. It is conceivable that a well-defined sequence of tests may produce the best information for clinical decision making. [98] New generation PET scanners combined with CT capability (PET-CT scanners) already can provide both functional and anatomic information simultaneously with minimal incremental burden to patients. Sites of abnormal ^{18}F-FDG uptake detected by PET in the abdomen, for example, can be readily localized by simultaneous CT. For correlating functional and anatomic imaging, PET-CT scanners are likely to prove clinically useful.

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CHAPTER 158

Arnold F. Jacobson

APPLICATIONS OF RADIONUCLIDE IMAGING IN GASTROENTEROLOGY

Nuclear medicine is a specialty devoted to diagnostic and therapeutic applications of radioactive isotopes (radionuclides) in medicine. The most important attribute of nuclear medicine imaging is its capability for minimally invasive study of organ and organ system physiology and function. This chapter provides an overview of the major clinical applications of radionuclide imaging in gastroenterology, including studies for diagnosis of acute cholecystitis and diseases of the hepatobiliary system; gastrointestinal (GI) bleeding; alimentary tract motility disorders; cavernous hemangiomas of the liver; and inflammatory, infectious, and malignant processes involving the liver, stomach, bowel, and abdominal cavity. Positron-emission tomography (PET), a radionuclide technique that has experienced dramatic growth during the past decade, is discussed briefly in this chapter and more fully in Chapter 157.

THEORETICAL CONSIDERATIONS

Radionuclide imaging is performed after administration of a radiolabeled compound (radiopharmaceutical) that localizes to a specific organ system or the location of an ongoing physiological process. Most radiopharmaceuticals are administered intravenously, although oral, intrathecal, intradermal, and intraperitoneal routes are used in association with some nuclear medicine procedures. Only those structures in which the radiolabeled compound accumulates can be seen externally, with alterations in the normal biodistribution of the administered material, seen as decreased or increased accumulation of the labeled compound, providing information concerning abnormal anatomic or physiological processes. Depending upon rates of uptake and excretion and the physical characteristics of the radionuclides, nuclear medicine imaging is performed as early as a few seconds to as late as 5 to 7 days after radiopharmaceutical administration.

Radionuclides

The radionuclides used in nuclear medicine are chosen based upon the characteristics of the emitted radiation, and either the biologic properties of an element which allow its use in vivo, such as iodine for thyroid imaging, or the physical and chemical properties of an element which allow it to be linked to appropriate compounds or pharmaceuticals, typically as a chelate in a complex. In order to be usable for imaging, an internally administered radioisotope must produce photons (either ?-ray or x-ray) of sufficient energy that the majority will travel through and escape from the body unattenuated. Isotopes with primarily lower energy photons (<60 keV) or particulate radiation such as ß-particles (electrons) are potentially useful for therapeutic pharmaceuticals but are generally avoided for diagnostic radiopharmaceuticals, since these emissions increase the radiation dose to the patient without contributing to imaging information. Isotopes with high energy photons are also avoided because most of these photons pass through the crystal in typical nuclear medicine imaging systems without interacting, resulting in low detection efficiency and statistically low quality images. While radionuclides with photons of 60 to 400 keV are used in conventional nuclear medicine imaging [1] (Table 158-1), there is noticeable deterioration in image quality for studies employing isotopes with energies greater than 200 keV. However, many newer general-purpose nuclear medicine systems have been designed specifically to have the capability for detecting the 511 keV annihilation photons used in PET.

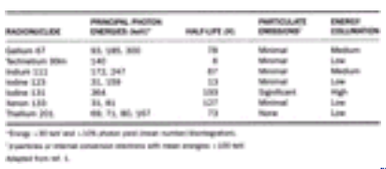

TABLE 158-1 Radionuclides in Common Use in Nuclear Medicine

Radiopharmaceuticals

Nuclear medicine imaging procedures are most effective when the radiopharmaceutical used successfully targets the organs or the physiological process(es) of interest. While most radiopharmaceuticals at best only approach the ideal properties for specific imaging applications, development work continues to produce newer compounds with increased radiochemical binding efficiency or targeted biologic extraction which results in improved imaging characteristics.

Technetium has proven particularly suitable for incorporation into stable complexes and chelates, with all necessary chemical reactions usually taking place in a single vial that contains the compound to be labeled and various reducing agents and stabilizers. Technetium 99m (99mTc), supplied in solution as the pertechnetate anion () from a device called a generator, is added, and after thorough mixing, greater than 95% of the radionuclide is typically bound to the desired compound or complex.

Side effects resulting from administration of nuclear medicine radiopharmaceuticals are extremely rare, with a reported occurrence of minor nonspecific or possible allergic reactions in 2/100,000 doses. [2] Strict adherence to proper techniques for handling blood products also has minimized incidents associated with reinjection of radiolabeled blood components. Even the potentially more antigenic radiolabeled proteins (of both human and mouse origin) in newer approved and investigational radiopharmaceuticals have generally shown excellent safety profiles, similar to those of previous generations of nuclear medicine compounds. [3], [4]

Radiation Dose

The internal radiation dosimetry from a nuclear medicine procedure depends upon the biodistribution and biologic half-life of the radiopharmaceutical, and the physical half-life and characteristics of the radiation emissions of the radionuclide. Compounds that are relatively rapidly cleared from the target organ and the body,

and are labeled with shorter-lived photon-emitting radionuclides, result in lower patient radiation doses. The dominant role of ^{99m}Tc in nuclear medicine, while attributable in part to the element's favorable chemical properties, is also due to its emission of a single ?-photon with energy suitable for imaging, minimal co-emissions of low-energy electrons, and short half-life (see Table 158-1). Target organ doses from nuclear medicine procedures are generally comparable to those received from examinations such as barium x-ray studies or contrast computed tomography (CT).

TECHNICAL APPLICATIONS

Imaging Systems

The ?-camera has been the central component of most clinical nuclear medicine imaging systems for the past four decades. The principal elements of this device are a large, flat crystal of sodium iodide in which incoming ?- and x-ray photons are absorbed and converted into flashes of light; a matrix of photomultiplier tubes that detect and amplify these scintillations; and electronic circuitry which determines the location (X-Y coordinates) of the scintillation events and their intensity, reflecting the energy deposited in the interactions. [1]

An exact image of the radiotracer distribution in a region can be obtained if only those photons emitted perpendicular to the desired imaging plane are detected. While absolute selectivity cannot be achieved due to the isotropic character of ?-photon emissions (in all directions with essentially equal probability), limitation of photon detection adequate for most diagnostic imaging purposes is accomplished through use of a collimator (in essence a sheet of lead containing a large number of small holes) placed in front of the camera face. Low-, medium-, and high-energy collimators are used for different isotopes, depending on the highest energy photon with significant abundance (see Table 158-1), with greater thicknesses of lead required to achieve adequate absorption of scatter and off-angle photons as the energy of the primary photons able to be imaged increases.

The completed scintigraphic image reflects the distribution of scintillation events (counts) detected across the ?-camera crystal during the course of an acquisition. An acquisition may be performed with the camera stationary, producing a spot image of a selected body region, or with either the camera or imaging table moving, resulting in a single image of the total-body distribution of the administered radiopharmaceutical.

While planar ?-camera imaging may be considered analogous to conventional radiography, cross-sectional imaging analogous to x-ray CT or magnetic resonance (MR) imaging also can be done using radionuclide techniques. This method is commonly referred to as single photon emission computed tomography (SPECT). SPECT is typically performed using a rotating ?-camera and either a "step-and-shoot" acquisition, with 32 to 128 planar views obtained while the camera traverses a 180- or 360-degree rotational arc around the patient, or an acquisition obtained during a continuous rotation over the same angular domain. SPECT data are processed using reconstruction algorithms similar to those used in CT or MR, and images are typically displayed in three standard planes: axial (cross-sectional), sagittal, and coronal.

While SPECT systems can have 1, 2, or 3 camera heads, 2-head systems are by far the most commonly used. In-plane spatial resolution with a single-head SPECT system is typically on the order of 10 to 15 mm, while resolution of 7 to 10 mm can be achieved using a dual-head system. Many newer 2-head systems also are capable of performing coincidence detection for imaging positron emitters.

Computers

The computer is an integral part of all modern nuclear medicine imaging systems, providing the capability for quantitation that distinguishes nuclear medicine from most other diagnostic imaging techniques. Each photon detected by the ?-camera is recorded in computer memory as a count in a digital matrix of picture elements (pixels). Each pixel encompasses a range of X-Y coordinates, depending upon the matrix size selected (typically 64 × 64 to 256 × 256 pixels for spot, 256 × 1024 for whole-body images).

Quantitative data are obtained from the stored digital images by determination of the number of counts detected within user-defined regions of interest (ROIs). Such data allow quantitation of organ ratios that reflect the biodistribution of the administered radiotracer. In addition, dynamic acquisitions, with typical image frame rates ranging from 0.25 seconds to 1 minute per image, can be stored in the computer memory and then displayed in a cine format to demonstrate the pattern of change in activity distribution over time. ROIs also can be drawn to generate time-activity curves for determination of uptake, filling, and emptying rates of physiological interest.

IMAGING APPLICATIONS

Liver

Many nuclear medicine procedures provide diagnostic information concerning the functional status of the liver or its involvement by pathological processes. These include examinations that are specific for liver function evaluation (e.g., hepatobiliary scintigraphy) and those that are used to examine the total body for infection (gallium 67 [^{67}Ga], labeled leukocytes) or malignancy (^{67}Ga, labeled peptides and monoclonal antibodies, PET). Specific radiologic imaging of the liver is now mostly the domain of ultrasound, CT, and MR imaging, with traditional radionuclide methods (e.g., colloid scintigraphy) and methods used primarily for research (e.g., cell function and receptor studies) only chosen for highly selected patients.

The conventional radionuclide liver-spleen scan, although important in the early progress of nuclear medicine imaging, is of little relevance in current clinical practice. Taking advantage of the ability of the hepatic Kupffer cells to phagocytose appropriately sized particulate material injected intravenously, the scan is performed following injection of a colloidal preparation of sulfur or albumin labeled with ^{99m}Tc. [5] Following intravenous injection, these labeled colloids are rapidly cleared from the circulation (half-time [T 1/2], 2–3 minutes) by the liver (80%–85% extraction), spleen (10%–15% extraction), and bone marrow (1%–5%). Significant liver disease results in impaired hepatic uptake of colloid and proportionate increases in uptake in the spleen and bone marrow. [6]

Because of its simplicity as an imaging procedure, colloid scintigraphy may occasionally be of use in situations where other imaging modalities are limited, such as for obese patients in whom sonography is inadequate and whose weight exceeds the limit of the CT or MR imaging tables, or for noninvasive evaluation of lesions suspected of being either focal nodular hyperplasia, which normally show colloid uptake, [7] or adenomas, which usually do not. [8] The need to use degree of shift of colloid uptake to the spleen and marrow as an indication of liver cirrhosis severity [6] is rarely encountered in current practice. Colloid imaging, particularly using SPECT, still finds occasional use as a means for identifying residual or accessory splenic tissue in patients after splenectomy, which can be of particular value when other imaging modalities have demonstrated potential sites of infection or tumor, neither of which should show uptake. Comparably specific splenic imaging can also be performed using heat-denatured ^{99m}Tc labeled red blood cells. [9]

Specialized examinations for assessing selected aspects of liver function are primarily used in centers with interest in the study of particular liver diseases. Investigators have taken advantage of the high solubility of xenon in fat to allow estimation of fat content in cirrhotic livers (as determined on biopsy) based upon the amount of retained xenon activity on anterior abdominal images acquired during the washout phase after a period of equilibrium breathing of xenon 133 (^{133}Xe) gas. [10] The investigational compound ^{99m}Tc galactosyl-neoglycoalbumin, whose binding to hepatic binding protein is directly related to hepatic function, [11], [12] also continues to be studied for its potential use in quantifying hepatic functional reserve and prognosis in cirrhotic patients.

Cavernous Hemangiomas

Cavernous hemangiomas are the most common benign liver tumor, and as such they are often observed as incidental findings on abdominal ultrasound and CT examinations. Lesions that display typical echogenicity on sonography or the expected pattern of delayed enhancement on contrast CT usually require no further workup. For lesions with atypical appearance on sonography or CT, or those in patients whose history or clinical findings make it necessary to exclude metastatic involvement of the liver, ^{99m}Tc labeled red blood cell (RBC) imaging of the liver provides an excellent means for demonstrating the greater blood pool within the hemangioma than in the surrounding liver parenchyma. Similar to the findings on contrast CT, the characteristic scintigraphic finding for hemangiomas is mildly decreased or normal labeled-RBC activity immediately after injection, followed by progressive increase in activity over the next 20 to 30 minutes. [13], [14] While observation of this pattern of enhancement on planar imaging is virtually pathognomonic for hemangioma, in general only larger lesions (=2 cm) can be reliably detected in this manner. Smaller lesions, and those that are located deeply within the liver parenchyma, are visualized with increased sensitivity using SPECT, typically performed 1 to 2 hours after the labeled RBC injection to maximize lesion enhancement. Hemangiomas appear as distinct focal areas of increased activity

compared with the surrounding liver parenchyma [15], [16] (Fig. 158-1). For lesions larger than 2 cm, sensitivity of SPECT RBC imaging for hemangioma identification exceeds 90%, which is equivalent to the performance of contrast CT and MR imaging. [17], [18] Hemangiomas down to 1.4 cm can be reliably identified on SPECT with a multihead camera, [19] and some as small as 1 cm can also be imaged. [17], [20] Nevertheless, verification of the etiology of lesions smaller than 1 to 1.5 cm is probably best accomplished with CT or MR.

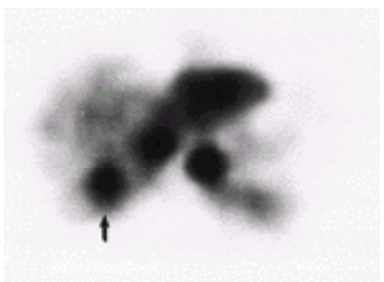

FIGURE 158-1. A 48-year-old man with a new lung neoplasm and an equivocal liver lesion on computed tomography and magnetic resonance imaging underwent single photon emission computed tomography imaging approximately 1 hour after injection of 26 mCi technetium 99m labeled red blood cells (RBCs). Axial image shows a discrete 2.5- to 3-cm focus of increased RBC accumulation in the posterior aspect of the right lobe of the liver (*arrow*), corresponding to the finding on the other cross-sectional modalities and consistent with a liver hemangioma. The centrally located areas of intense activity represent the aorta and inferior vena cava, respectively.

Hepatobiliary System

The most widely used nuclear medicine study examining hepatocyte function and the hepatobiliary system employs ^{99m}Tc labeled iminodiacetic acid (IDA) compounds. [21] Correctly described as hepatobiliary scintigraphy or cholescintigraphy, this procedure is still often referred to as a HIDA scan, in reference to an early IDA compound which was long ago supplanted by superior substituted IDA agents.

IDA compounds enter the hepatocyte via a carrier-mediated pathway, with a mechanism very similar to that of bilirubin. Uptake of the compounds in current use (disofenin, mebrofenin) is considerably less affected by competitive inhibition in the presence of significantly elevated serum bilirubin levels compared to earlier IDA derivatives such as HIDA and PIPIDA. These improved physiological characteristics allow adequate liver uptake and tracer excretion at bilirubin levels as high as 20 to 30 mg/dL. [21]

Hepatobiliary scintigraphy is performed following intra- venous administration of typically 3 to 5 mCi of a ^{99m}Tc labeled IDA compound, although up to 10 mCi may be used when bilirubin levels are significantly elevated. Anterior abdominal images are most often acquired dynamically (one image per minute) over the next 60 minutes, although acquisition of a series of static views (typically every 5 minutes) is an acceptable alternative. A normal study shows rapid uptake of the radiotracer into the liver, with virtually all cardiac blood pool activity cleared by 5 minutes after injection, followed by the liver excretory phase, with sequential visualization of intrahepatic bile ducts, the common hepatic duct, the cystic duct, gallbladder, common bile duct, and small bowel (Fig. 158-2). All of these structures typically are seen by 30 minutes after injection, although visualization of activity in the gallbladder and small bowel within 60 minutes is considered normal. While relative visualization time of the gallbladder and small bowel are not definitely correlated with most hepatobiliary pathology, small bowel visualization prior to the gallbladder likely reflects low basal sphincter of Oddi pressure, with the majority of hepatic bile excreted directly into the small intestine. In contrast, up to 20% of normal individuals have a relatively high resting sphincter of Oddi tone, which potentiates early filling of the gallbladder and results in absence of bowel visualization during the first 60 minutes of a hepatobiliary scan. [22] For such individuals, normalcy of the hepatobiliary system can be demonstrated through stimulation of the gallbladder, either by a fatty ingestion, which releases endogenous cholecystokinin (CCK), or an exogenous-administered cholecystagogue such as sincalide (Kinevac, Bristol-Myers Squibb, New York, NY), the terminal octapeptide of CCK, which produces gallbladder contraction and sphincter of Oddi relaxation, with resultant visualization of radiolabeled bile entering the small bowel.

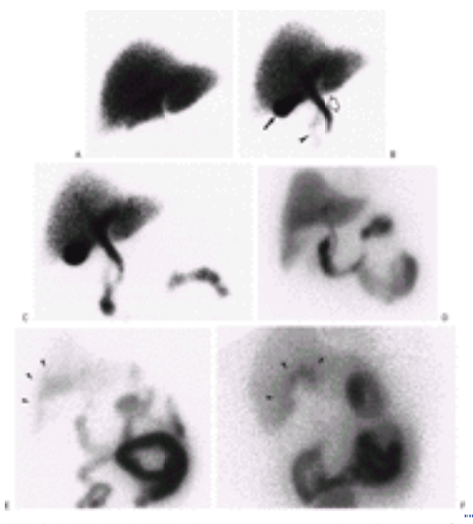

FIGURE 158-2. Images **A–C** are from a 76-year-old man with atypical right upper quadrant pain and fever to 104°F. Ultrasound showed multiple stones but no pericholecystic fluid or gallbladder wall thickening. Technetium 99m (^{99m}Tc) mebrofenin image at 4 minutes postinjection (**A**) shows prompt normal homogeneous uptake throughout the liver. At 10 minutes (**B**), there is visualization of activity in the gallbladder (*arrow*), common bile duct (*open arrow*), and small bowel (*arrowhead*). At 15 minutes (**C**), further transit of small bowel activity is evident. These results exclude the diagnosis of acute cholecystitis with high negative predictive value. For comparison, images **D–F** are from hepatobiliary scintigraphy in a 61-year-old man with a 2-day history of severe right upper quadrant abdominal pain, sonography showing irregular gallbladder wall thickening and shadowing suggestive of either a polyp or stone, temperature of 101.1°F and a white blood cell count of 20.1. Anterior image at 30 minutes (**D**) shows satisfactory uptake of the ^{99m}Tc mebrofenin by the liver and normal appearance of tracer activity in the small bowel. The gallbladder was not visualized to 60 minutes, at which time 4 mg morphine sulfate was administered intravenously; the gallbladder was not seen during an additional 30 minutes of imaging. Increased activity in the liver adjacent to the gallbladder fossa can be seen on the last postmorphine image (**E**) and even more clearly on a subsequent right anterior oblique image (**F**) (*arrowheads*), reflecting the rim sign. At subsequent surgery, necrotizing and suppurative cholecystitis was found, with extension of inflammation into the liver bed.

Hepatobiliary scintigraphy can be used to evaluate both parenchymal liver function and the structural integrity and patency of the various parts of the hepatobiliary tree. Among applications of this method are studies of infants with suspected biliary atresia, [23] patients with primary biliary cirrhosis [24], [25] and sclerosing cholangitis, [26], [27] and individuals who have undergone liver transplantation. [28], [29] Time-activity curves for selected regions of interest in the liver and bile ducts can be used to estimate rates of hepatic uptake and clearance and bile flow. Abnormal bile flow kinetics may be an indication of intrahepatic and extrahepatic biliary obstruction, rejection in transplant patients, and congenital or acquired ductal anomalies. In particular, common bile duct obstruction results in a typical pattern of intrahepatic cholestasis and gallbladder nonvisualization, particularly if the obstruction is more than 24 hours in duration. [30], [31]

Hepatobiliary scintigraphy is most frequently used to evaluate for the presence of acute cholecystitis, reflected by mechanical or functional obstruction of the cystic duct resulting in absence of gallbladder visualization. In the presence of signs and symptoms such as right upper quadrant tenderness, elevated white blood cell count, fever, and gallstones or wall thickening on gallbladder sonography, nonvisualization of the gallbladder by 60 minutes has a sensitivity greater than 95% for the

diagnosis of acute cholecystitis. [32], [33], [34] and [35] The most common pathological reason for a false-positive scan is chronic cholecystitis, which is often associated with low rates of bile flow into the gallbladder and therefore early nonvisualization. Although in patients with chronic cholecystitis there is often sufficient bile flow into the gallbladder to demonstrate visualization by 2 to 4 hours postinjection, current practice is to administer intravenous low-dose morphine sulfate (typically 0.04 mg/kg to a maximum of 4 mg) if the gallbladder is not visualized by 60 minutes. Morphine causes contraction of the sphincter of Oddi, increased intraductal pressure, and increased bile flow into the gallbladder, and results in gallbladder visualization within 30 minutes in the majority of patients with chronic but not acute cholecystitis. Continued absence of gallbladder visualization during this time further supports the diagnosis of acute cholecystitis (see Fig. 158-2). Another indicator of acute cholecystitis seen in some patients is the “rim sign,” a persistent region of activity in the liver adjacent to the gallbladder fossa attributed to edema and localized inflammation in the liver causing impaired clearance of labeled bile. This sign, although only modestly specific, is most often noted in association with more complicated cholecystitis with transmural involvement, such as gangrenous or perforated gallbladder [36], [37] and [38] (see Fig. 158-2).

Morphine can occasionally convert a true-positive into a false-negative study, [39] presumably as a result of the increased intraductal pressure either dislodging a stone or overcoming the high resistance of an edematous but still patent gallbladder neck. Nevertheless, the reported sensitivity of gallbladder nonvisualization on morphine-augmented cholescintigraphy in patients suspected of acute cholecystitis (93%–95%) [40], [41] is only slightly lower than results published in the “pre-morphine” era. It has been suggested that false-positive (for acute rather than chronic cholecystitis), morphine-augmented cholescintigraphy may reflect failure of the pharmacological agent to increase bile flow through the cystic duct, [42] which could conceivably occur if either sphincter of Oddi constriction or rate of production and excretion of radiolabeled hepatic bile were inadequate to produce a detectable change in rate of gallbladder filling.

Hepatobiliary scintigraphy, similar to sonography, must be performed in the fasting state, since postprandially, the gallbladder is contracted and bile flow is diverted almost entirely into the small bowel, producing a false-positive finding of gallbladder nonvisualization. [43] A minimum fast of 4 hours is usually recommended prior to initiation of a study. However, false-positive studies can also occur after prolonged fasting, such as in patients in intensive care units and those on total parenteral nutrition, presumably because the gallbladder becomes atonic and contains concentrated viscous, sludgy bile, which limits entry of new hepatic bile even though the cystic duct is patent. [44], [45] One option in patients who have taken nothing orally for extended periods (>24–48 hours) is to administer a dose of intravenous sincalide 1 to 2 hours prior to beginning the scintigraphic study, thereby potentially producing sufficient emptying of the gallbladder to allow filling to occur following injection of the radiotracer. [46] Reports indicating that regular administration of sincalide can reduce sludge formation in patients on prolonged periods of intravenous alimentation [47] also provide support for the potential use of sincalide pretreatment in diagnostic imaging.

Hepatobiliary scintigraphy can be used to evaluate congenital or acquired structural anomalies of the biliary tree or to examine for abnormalities resulting from operative or traumatic injury. Choledochal cysts are visualized as isolated areas of increased retention of tracer corresponding to the involved region of the bile duct, [48], [49] while sclerosing cholangitis results in multiple scattered areas of retention secondary to ductal stenoses. [26], [27] Visualization of tracer outside the normal ductal pathways is usually indicative of bile leakage, and scintigraphy can therefore be helpful in evaluating patients suspected of bile peritonitis or postoperative or posttraumatic liver or bile duct injury. [50] In postgastrectomy patients, the pattern of bile flow can be examined to assess the functional characteristics of the efferent and afferent limbs, the latter often better demonstrated scintigraphically than by x-ray studies with oral contrast.

A commonly performed, although somewhat controversial, functional evaluation in hepatobiliary imaging is the quantitative measurement of gallbladder response to stimulation by endo- genous or exogenous CCK or its analogs. Following adminis- tration of the ^{99m}Tc labeled IDA compound and sufficient time thereafter to allow liver clearance and maximum gallbladder filling, the oral meal or the intravenous agent is administered. Dynamic images in the anterior or occasionally the anterior oblique projection are acquired on computer as the gallbladder gradually contracts, and these images are then analyzed to estimate the reduction in volume represented by the gallbladder ejection fraction (GBEF). GBEF values are typically compared against results for normal controls studied under the same test conditions. [51], [52] and [53]

The ready availability, ease, and reproducibility of administration of intravenous sincalide have made this agent the most commonly used stimulus for gallbladder contraction in clinical and research examinations. The total dose of sincalide used is typically 0.01 to 0.04 µg/kg, administered as either a short-duration (1 to 3 minutes) bolus or a slow continuous infusion over 15 to 60 minutes. The dose most commonly employed is 0.02 µg/kg, with a higher dose of 0.04 µg/kg only occasionally needed to elicit a response in selected patients. In comparison to the bolus technique, sincalide administered as a continuous infusion produces more complete gallbladder contraction, has better reproducibility, produces fewer symptoms, and is more physiological, which is analogous to the sustained elevation of endogenous CCK that occurs after a meal. In a comparison of 0.02 µg/kg bolus and 30-minute infusion protocols in 23 normal volunteers, the mean GBEFs were 56% and 70%, respectively, with the infusion GBEF higher in 21 subjects (91%). [54]

The value of sincalide-augmented hepatobiliary imaging and more generally the role of GBEF determinations as a guide for therapeutic decision making has long been a subject of controversy. Although early investigators noted a definite relationship between decreased gallbladder contractility and acalculous cholecystitis, later studies often yielded disparate results, with some showing a definite correlation between low GBEF and symptom relief following cholecystectomy, [55], [56] and [57] while others found no correlation between the presurgical GBEF and either severity of gallbladder histopathological changes [58] or relief of symptoms following cholecystectomy. [59] Given that the scintigraphic GBEF can only be determined if the gallbladder is visualized, which is typically not the case in patients with cholecystitis, a study showing that sincalide can be used reliably when the gallbladder is only seen postmorphine is notable. [60] In that study, of 11 patients with abnormal GBEF (defined as <40%, although all had GBEF <20%), all had cholecystitis, including 5 with acalculous disease. Nevertheless, some asymptomatic subjects with normal gallbladder sonography also have low GBEFs, [52], [54] indicating that although sincalide-augmented cholescintigraphy is reliable as a measurement technique, application of its results to therapeutic decision making must be done with care, particularly in judging the clinical significance of biliary dyskinesia.

Scintigraphy has proven useful for examining patients suspected of biliary pain on the basis of sphincter of Oddi dysfunction. Delayed excretion of radiolabeled bile and decreased gallbladder contraction in response to sincalide are both associated with sphincter of Oddi dysfunction documented manometrically. [61], [62] Sphincter dysfunction may also be associated with a paradoxic response to sincalide (i.e., increased sphincter pressure resulting in increased activity in the common bile duct after administration of the cholecystagogue). Several studies have shown significant improvement in bile flow dynamics following sphincterotomy or sphincteroplasty, with symptom relief also correlated with abnormal preoperative emptying kinetics. [62], [63] and [64]

Gastric Mucosa

Radionuclide imaging of the stomach and gastric mucosa is generally limited to investigations of children and young adults with abdominal pain and GI bleeding to identify the presence of ectopic gastric mucosa often associated with Meckel diverticula. Technetium 99m pertechnetate is actively extracted by the mucous-secreting cells in gastric mucosa, thereby providing a means for locating sites of ectopic gastric mucosa. [65], [66] For a Meckel scan, 30 to 100 µCi/kg of ^{99m}Tc pertechne- tate is administered, followed by serial anterior imaging of the abdomen for the next 30 to 60 minutes. [67] Just as the circulating radiotracer is progressively extracted by the gastric mucosa in the stomach, a Meckel diverticulum that contains gastric mucosa will show enhancement of activity over time. Appearance of a focus of pertechnetate uptake outside of the stomach within 10 to 20 minutes, typically in the right lower quadrant of the abdomen, usually is indicative of a Meckel diverticulum. Oblique views of the abdomen and views with the bladder blocked with lead or drained via an indwelling catheter are sometimes done to enhance identification of small focal accumulations in the lower abdomen. Intravenous administration of pentagastrin (6 µg/kg 15 to 20 minutes before pertechnetate injection) or an H2-receptor antagonist (300 mg cimetidine or 50 mg ranitidine 1 hour before pertechnetate injection; pretreatment can also be done using oral preparations for 2 days prior to the study) may result in improved visualization of Meckel diverticula via increasing gastric mucosal uptake, and glucagon (50 µg/kg IV 10 minutes after pertechnetate injection) may produce the same outcome via delaying local clearance of the radiotracer. [67], [68] and [69] Even without use of such pharmacological augmentation, sensitivity and specificity for detection of Meckel diverticula based upon surgical correlation in patients presenting with rectal bleeding and other symptoms are on the order of 85% and 95%, respectively. [65]

Gastrointestinal Bleeding

While most nuclear medicine studies are routine outpatient diagnostic examinations, GI bleeding studies are often performed on an urgent or emergent basis on sometimes hemodynamically unstable patients. The underlying principle of GI bleeding scintigraphy is that an intravascular tracer in circulation at the time of active bleeding will extravasate into the bowel lumen, thereby allowing identification of the bleeding site. The most well-characterized radiopharmaceuticals for this application are ^{99m}Tc labeled colloid and ^{99m}Tc labeled RBCs. These two agents have dramatically different imaging characteristics, in that the former is rapidly cleared from the circulation following intravenous administration, while the latter has prolonged residence in the vascular space. The advantages and disadvantages

of the physiological characteristics of these two agents are discussed subsequently.

The theoretical advantage of a radiotracer agent for GI bleeding studies that is rapidly cleared from circulation is that the initial extravasation of activity into the bowel and its subsequent intraluminal movement can be identified with great precision because of the low tissue background. [70] In the case of radio- labeled colloid, activity rapidly accumulates in the liver and spleen, with the remainder of the abdomen having minimal background counts within a few minutes postinjection. Rates of bleeding as low as 0.05 mL/min have been identified using sulfur colloid imaging in an animal model in which a catheter was used to introduce radiotracer into the bowel at a controlled rate. [71] The major disadvantage of labeled colloid is that the same rapid vascular clearance that allows high-contrast imaging also results in only a few minutes of opportunity for identification of the bleeding site. The intermittent nature of many episodes of GI hemorrhage and the variable interval between the initial presentation with bleeding complaints and the referral of the patient for a scintiscan makes it highly unlikely that active bleeding will be occurring during any short period of study. In two comparative studies with labeled RBCs involving 127 patients with GI bleeding, a bleeding site was identified on colloid scintigraphy in only 7 (5%). [72], [73] Even though multiple repeated colloid injections can be done to improve sensitivity, this is usually not practical as a routine procedure. Ultimately, the intermittent nature of GI bleeding limits the use of colloid as a study agent.

A radiotracer with prolonged blood pool residence provides the best capability for detection of both active and intermittent GI bleeding, and ^{99m}Tc labeled RBCs are most commonly used for this application. [74], [75], [76] and [77] Cells are labeled by a method that does not involve intravenous administration of ^{99m}Tc pertechnetate (either a modified in vitro or totally in vitro technique) to avoid possible uptake and secretion of free pertechnetate by the gastric mucosa in the absence of active bleeding. Once reinjected, the labeled RBCs remain in circulation for as long as the cells are viable, which permits repeated imaging as necessary limited only by the physical half-life of ^{99m}Tc.

Following injection of 20 to 30 mCi of ^{99m}Tc labeled RBCs, anterior abdominal imaging is typically performed for 60 to 120 minutes, depending upon the initial findings and the relative stability of the patient. A continuous dynamic acquisition is definitely preferable to serial static imaging (typically at 5-minute intervals), with a frame duration of at most 60 seconds; rates as rapid as 10 seconds per frame have been reported useful in detecting rapid intermittent bleeding. [78], [79] and [80] Anterior oblique and lateral views are sometimes also obtained to aid in bleeding site localization. In the presence of active bleeding, labeled RBCs enter the bowel lumen and are seen as a focus of increased activity. The two critical features of a positive bleeding study are this initial identification of a focal accumulation of blood, and subsequent observation of its intralumenal movement to more specifically localize the region of bowel that contains the bleeding site (Fig. 158-3). If the patient is too unstable to remain for an extended period in the nuclear medicine department, or if angiographic or surgical procedures to stop the bleeding are pending, it may be necessary to terminate the study as soon as a positive finding is identified, but under most circumstances, further images demonstrating movement of labeled blood allow more accurate discrimination of small from large bowel sources. The dynamically acquired images viewed as a continuous cine loop are particularly effective for demonstrating passage of intralumenal labeled RBCs through the bowel. [67], [79]

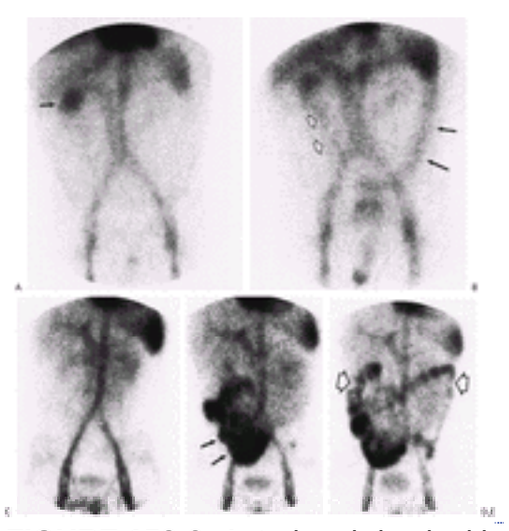

FIGURE 158-3. Anterior abdominal images from two labeled red blood cell gastrointestinal bleeding studies are shown. **A:** Focal accumulation of activity is seen in the right upper quadrant (*arrow*) at 10 minutes after injection at the site of an anastomotic bleed in the right colon, with (**B**) labeled red blood cells seen in the ascending (*open arrows*), transverse, and descending colon (*arrows*) on a later image at 60 minutes as a result of both retro- and antegrade movement of blood. **C:** The images on the right show no definite extravasation at 60 minutes, but (**D**) diffuse intralumenal blood is seen in the distal small bowel (*arrows*) at 4 hours and (**E**) throughout the colon (*open arrows*) at 5 hours. Angiography demonstrated active bleeding at the site of duodenal ulcers seen previously at endoscopy. All scintigraphic images show normal blood pool activity in the liver, heart, and spleen (left to right at the top of each image) and in the major abdominal blood vessels.

The appearance of an intralumenal accumulation of labeled RBC activity, either on initial imaging or during acquisition of a consecutive series of images at a later time, identifies bleeding sites in the stomach, small bowel, or colon with an accuracy exceeding 90%. When bleeding is slow, the initial site visualized may be a location where a sufficient quantity of radiolabeled blood has pooled to allow it to be seen above the abdominal blood pool background. Although this initial visualized site may therefore occasionally be a few centimeters proximal or distal to the primary bleeding location, the discrepancy rarely interferes with correlation of scan findings with those of other procedures (e.g., angiography, endoscopy, surgery).

Active bleeding sites are typically seen during the first 1 to 2 hours of imaging in up to one- third of patients studied. [72], [75], [81], [82] For the remaining patients, options at this point include proceeding with other investigations (endoscopy, angiography, surgery), or obtaining later delayed views. The results on later delayed views indicate whether active bleeding has occurred during the interval between imaging sessions, and several sequences of images can be obtained over the approximately 24-hour period after the original labeled RBC injection during which imaging with acceptable acquisition times is feasible (see Fig. 158-3). As “old” blood which entered the bowel prior to the injection of the labeled cells will not produce a positive scan finding, any intralumenal labeled RBCs can only have entered the bowel subsequent to the start of the study. Stool or blood passed per rectum can also be examined for the presence of ^{99m}Tc using either the ?-camera or a well counter, providing an extremely sensitive method for identifying even trace amounts of extravasated blood which may not have been evi- dent on the abdominal images.

Later delayed images demonstrate intralumenal labeled RBCs in 20% to 46% of patients whose early images are negative, overall about one-third of all patients studied. [74], [81], [82] and [83] However, the location(s) of activity seen on delayed images often does not reflect the site where the labeled blood entered the bowel. While the bleeding site is unlikely to be significantly distal to the location where blood is seen (allowing for the possibility of some retrograde movement of bowel contents), it could be present anywhere proximal to that location. It is relatively common to see diffuse activity in the colon on 18- to 24-hour delayed images, but nevertheless the identified source of bleeding in as many as half of such patients is in the upper GI tract. [82] As a prognostic indicator, positive delayed images (denoting continued bleeding during the study interval) are seen more often in patients with more complicated courses, including those who require more blood transfusions and more frequent intervention by angiography or surgery. [74], [82]

In clinical practice, only early imaging results from radionuclide GI bleeding studies are likely to influence patient management decisions, with later delayed images, although of demonstrated diagnostic and prognostic value, usually considered worth obtaining in only selected cases. A strongly positive early finding might result in prompt angiographic or surgical follow-up, while a subtle positive finding might not alter a preexisting plan to proceed with bowel cleansing followed by colonoscopy. The latter course of action is also typically followed in patients with negative early images. Given that bowel cleansing removes the evidence of interval extravasation of labeled RBCs, there is in fact no value in obtaining late imaging in this situation, and such imaging could even be misinterpreted as false negative. While it is potentially helpful to be able to identify individuals who are most likely to have late-positive imaging, features such as serum blood urea nitrogen (BUN) to creatinine ratios of =25 unfortunately do not translate into a significantly greater probability of bleeding site identification. [84]

The sensitivity for detection of bleeding using labeled RBCs is about 0.1 to 0.2 mL/min, [73] compared with 0.5 mL/min for angiography. Even preceded by a positive bleeding scan, angiograms typically show contrast extravasation in less than half the patients studied. [74], [81], [85] This reflects both the intermittent nature of the majority

of GI bleeding, and the fact that even in the most well-organized imaging department, bleeding may stop in the interval, regardless of the length, between the completion of the bleeding scan and the initiation of angiography. Adjunctive approaches that have been used in an attempt to improve bleeding source identification include scintigraphic studies in association with intravenous heparin to increase the likelihood of bleeding, [86] and intraarterial injections of radiolabeled material through angiographic catheters in patients with negative angiograms.

Gastrointestinal Tract Motility Studies

Gastric Emptying Test meals containing small amounts of radionuclides can be used to study both rates and characteristics of the transit of materials through any portion of the tubular alimentary tract. Of the various methods that have been developed, those for studying gastric emptying are in the widest general use. Both liquid and solid gastric emptying can be examined via serial abdominal imaging in anterior or posterior projections for 60 to 180 minutes following ingestion of a suitable labeled material after an overnight fast. [87], [88] and [89] Images are acquired either as a dynamic sequence at one frame per minute, or as a series of static views typically every 15 minutes, although through use of mathematical models of gastric emptying fewer measurement points may be needed (e.g., images at 0, 1, 2, and 3 hours), reducing the imaging burden on both the patient and the nuclear medicine clinic. [90] Acquisition of both anterior and posterior views (such as is readily achieved with a dual-head camera) allows calculation of the geometric mean tracer activity at each time point. ROIs are routinely drawn around the gastric silhouette to obtain the time-activity curve for emptying, and smaller ROIs may be used to isolate portions of the stomach (e.g., fundus, antrum) to assess peristaltic behavior that may contribute to delays in emptying. The emptying half-time (T 1/2) for the whole stomach is typically determined and compared against results for normal subjects with the same test meal (Fig. 158-4). The time-activity curve can also be analyzed to assess the pattern of emptying, including determination of the duration of emptying delay associated with gastric processing of solids (lag phase).

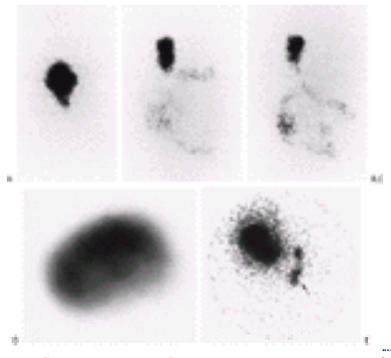

FIGURE 158-4. A 54-year-old man with history of gastroesophageal reflux disease refractory to medical therapy underwent a gastric emptying study to evaluate for a possible gastric motility disorder. Posterior abdominal images shown are from the 2nd minute (**A**), the 25th minute (**B**), and the 60th minute (**C**) of the study. There is gradual progressive clearance of tracer activity from the stomach throughout the study, with an estimated clearance half-time of 26 minutes, within the normal range. For comparison, images **D** and **E** are from liquid gastric emptying studies performed on a 56-year-old man with pyloric stenosis, the first scan before and the second three days after pyloric dilation. The initial examination (**D**) shows no emptying during the first 60 minutes of imaging (posterior image shown). Following dilation, there is substantial improvement in emptying, with activity immediately seen in the duodenum on a 1-minute posterior image (**E,** *arrow*) and gastric emptying half-time of 26 minutes.

Liquid-phase gastric emptying studies are relatively simple to perform, with 100 to 300 µCi of ^{99m}Tc-labeled colloid or indium 111 (^{111}In) diethylenetriamine pentaacetic acid (DTPA) administered in 150 to 300 cc of water, juice, or another suitable liquid, followed by imaging for 30 to 60 minutes. Normal T 1/2 is in the range of 15 to 45 minutes, but may be as short as 5 to 10 minutes. When liquid-phase emptying is abnormal, no further study may be necessary, but in most cases, examination of solid emptying is also appropriate. A variety of test meals have been used for solid gastric emptying studies, typically labeled with 300 to 1000 µCi of ^{99m}Tc colloid. As it is important that the labeled compound not dissociate or separate from the solid test meal in the stomach, one commonly used method involves injecting labeled colloid into an egg through the shell and then cooking the egg, thereby binding the radiotracer in the coagulated protein. A recent multicenter trial used a slice of liver injected at multiple sites with colloid, then fried on both sides, cut up and mixed into chicken stew for ingestion. [90] Liver pate, beef stew, and scrambled eggs have also been used in test meals. [91], [92] Solid gastric emptying studies are performed in essentially the same manner as for liquids, although an imaging acquisition of typically 60 to 120 minutes and occasionally to 3 to 4 hours may be required. Normal curves and derived half-times for gastric emptying depend on the composition (e.g., calories, fat content) and total mass of the test meal. Although efforts continue to standardize test meals (cooked egg and meat stew being most often used), sites that perform sufficient numbers of gastric emptying studies are still best served by establishing normal ranges locally with the specific meal being used to evaluate patients. Mean T 1/2 for solid gastric emptying for different size test meals ranges from 68 to 277 minutes, [90], [92] with T 1/2 usually less than 120 minutes for the smaller meals in common clinical use. [92] Simultaneous examination of liquid and solid gastric emptying can be accomplished using a dual-isotope technique, with the solid labeled with ^{99m}Tc colloid and the liquid labeled with ^{111}In DTPA. The dual-isotope technique is somewhat more physiological in its allowance for ingestion of solids accompanied by some liquid, and permits a more complete analysis of gastric motility in the same length of time required for a standard solid-only emptying study. [89], [92] As a standardized quantitative technique, serial gastric emptying studies are readily used for evaluating the effectiveness of interventions, such as procedures to relieve mechanical obstruction or medications to treat gastroparesis. [93] A baseline study is done prior to initiation of therapy, and a second study is performed subsequent to the procedure or achievement of the optimum response to drugs such as metoclopramide [94] (see Fig. 158-4). Studies using stimulators of the motilin receptors in the stomach such as erythromycin may also be useful in documenting effects of delayed gastric emptying in postsurgical patients, such as those status-postesophagectomy with gastric pull-up, [95] in whom conventional barium swallow x-rays often demonstrate no evidence of mechanical obstruction.

Gastroesophageal Reflux Gastroesophageal (GE) reflux is often identified qualitatively on the images from a conventional gastric emptying study, and a semiquantitative estimate of reflux severity can be derived from determination of the amount of activity in an esophageal region of interest. GE reflux can be quantified in a more systematic manner using an inflatable abdominal binder. Following ingestion of 300 µCi of ^{99m}Tc colloid in 300 mL acidified orange juice in the upright position, baseline images of the chest and upper abdomen are obtained to verify initial esophageal clearance of tracer activity. The patient then is fitted with the abdominal binder, positioned supine under the ?-camera, and imaged as the binder is inflated in stepwise fashion to produce incremental increases in intra-abdominal pressure. Intra-abdominal pressure required to induce GE reflux has been quantified in normal volunteers, and the control data have been used to evaluate results in symptomatic individuals. [87] In patients suspected of pulmonary aspiration secondary to GE reflux, a radiolabeled meal can be consumed in the evening before bedtime. Following a normal night of sleep, the patient is imaged to determine if radiotracer is present in the tracheobronchial tree or the lungs. Detection of activity in the respiratory system above background is objective evidence of the occurrence of aspiration. [96] This study can be repeated following medical or surgical intervention to reduce or eliminate GE reflux and aspiration to assess the effectiveness of those measures.

Esophageal Motility Nuclear medicine techniques can be used to examine esophageal motor function and esophageal transit. The patient is typically instructed to take a small quantity of ^{99m}Tc-labeled liquid into his or her mouth and then to swallow while dynamic ?-camera images of the anterior chest are obtained at a high frame rate, typically 1 to 3 images per second. Additional 15 second per frame imaging for up to 10 minutes, with a dry swallow at the beginning of each frame, may also be performed. Time-activity curves then are obtained for three segments of the esophagus (proximal, middle, and distal), and the timing and coordination of the passage of the tracer is examined. Neuromuscular and collagen-vascular diseases produce characteristic patterns of delayed tracer clearance, while diffuse esophageal spasm results in discoordinate changes in regional activity. Patient study results are commonly compared with data from normal volunteers. [97]

Bowel Motility While motility studies of the stomach and esophagus are technically straightforward because these structures are readily identified on scintigraphic images and therefore discrete regions of interest can be drawn to perform quantitative assessments, similar examinations of the small bowel and colon have proved considerably more difficult. The complex and variable anatomy and spatial orientation of the small bowel make it virtually impossible to isolate specific bowel segments with sufficient clarity to permit detailed analysis of transit times and peristaltic function. For the colon, although the anatomy is more straightforward, it may be difficult to obtain images for analysis that are not seriously compromised by interference from overlapping loops of small bowel. Investigators have nevertheless sought means to employ tracer techniques for assessing bowel motility disorders and have achieved modest success despite the technical limitations. The most well validated of these techniques use various quantitative measures of bowel transit. [89] Several different approaches have been investigated to quantify the movement of intralumenal contents between specific locations in the gut. A standard gastric emptying study can be continued for an additional few hours, and an estimate made of the arrival time of activity in the terminal ileum or cecum. Alternatively, to eliminate dispersal of labeled food throughout the bowel, tracer can be formulated in a nondigestible medium that passes unaltered through the alimentary tract, or it can be placed in a time-release capsule designed to dissolve in the distal small bowel. This latter technique uses an analytic scheme involving determination of the geometric center of colonic activity based upon the relative activity in each of four regions of the colon (ascending, transverse, descending, rectosigmoid) at specific times after ingestion of the radiotracer. From these data, “whole-gut” transit can be evaluated by comparison with results from control populations. [98] As colon or “whole-gut” motility studies usually require more than one day to complete, the

longer-lived isotope ^{111}In is typically used. [99]

Salivary Glands

The physiological behavior of the salivary glands can be studied using the radiotracer ^{99m}Tc pertechnetate, [100], [101] which is taken up by the secretory cells of the salivary glands similar to chloride or iodide anions. Following injection of 5 to 10 mCi, anterior and lateral images of the head and neck are acquired to document the location and relative function of the various glands. Dynamic acquisitions allow the generation of time-activity curves for regions of interest over the salivary glands in order to estimate rates of saliva production and excretion. Saliva secretion can also be stimulated with an acidic substance such as lemon juice to evaluate for possible effects of ductal anomalies, stones, or malignancy on gland clearance.

Inflammatory and Infectious Diseases

Labeled leukocytes are the scintigraphic agent of choice for investigating inflammatory processes involving the bowel. [102], [103] and [104] Mixed leukocyte populations are typically separated from 30 to 60 cc of blood, labeled in vitro with either ^{99m}Tc or ^{111}In, and then reinjected. However, because of differences in labeling efficiency and biodistribution, imaging techniques are usually individualized for the two radionuclides.

Imaging for ^{111}In oxine labeled leukocytes is usually performed 2 to 24 hours postinjection. Delayed imaging at 18 to 24 hours allows time for clearance of normal intravascular activity, particularly noticeable in the lungs early after reinjection, and greater accumulation of labeled leukocytes at abnormal sites, which are typically defined more clearly on later views. Imaging at 2 to 6 hours postinjection is of acceptable quality for abdominal sites where blood pool background is not a major constraint. [102]

There is increased focal uptake of labeled leukocytes at sites of infection or inflammation, the intensity of which is sometimes scored relative to activity at sites of normal uptake such as the liver or bone marrow. For increased activity seen in the bowel, it is necessary to ascertain whether the leukocyte accumulation is in the bowel wall, such as might occur in active inflammatory bowel disease or pseudomembranous colitis, or in the bowel lumen, either sloughed from a local inflammatory process, or transported from a more proximal site in the bowel or from locations such as the nasopharynx or the respiratory tract. GI bleeding or swallowed blood from hemoptysis also can result in intralumenal labeled leukocyte activity. Thus, when initial images demonstrate activity in a region of bowel, additional views are usually obtained a few hours or a day later; intralumenal activity will change its location, configuration, and intensity over time, while inflammation or infection in the bowel itself will display a relatively fixed distribution (Fig. 158-5). Bowel cleansing also can also be used to aid in determining whether activity is intralumenal or intramural.

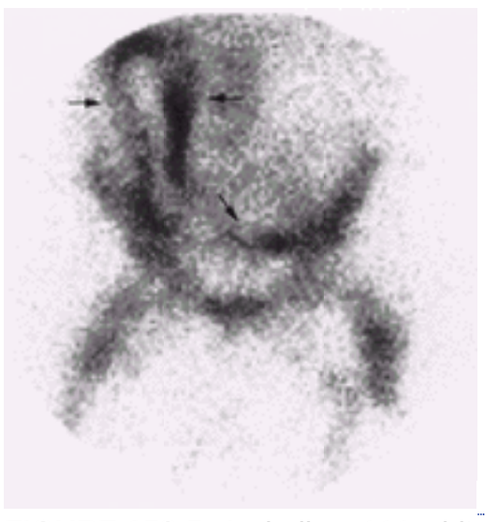

FIGURE 158-5. An indium 111 white blood cell scan was performed on a 50-year-old quadriplegic man to investigate for a source of persistent fever, bacteremia, and elevated white blood cell count. Anterior abdominal image at 24 hours postinjection unexpectedly revealed diffuse increased tracer activity throughout a somewhat redundant colon (*arrows*), which remained essentially unchanged on a follow-up image several hours later (not shown). Sigmoid colon biopsies obtained at colonoscopy several days later revealed chronic active colitis consistent with idiopathic inflammatory bowel disease.

The most common means to label leukocytes with ^{99m}Tc involves use of the brain imaging agent ^{99m}Tc exametazime (Ceretec, Amersham Health, Princeton, NJ), a lipophilic substance which crosses cell membranes with relative ease. [104], [105] The leukocyte labeling efficiency for ^{99m}Tc exametazime is lower than for ^{111}In, but the higher total dose of ^{99m}Tc that can be used compensates for this. Using 10 mCi of ^{99m}Tc exametazime, an average labeling efficiency of 60% results in a considerable gain in detected count rate compared to an injected dose of 0.3 to 1.0 mCi ^{111}In-labeled cells. Technetium 99m exametazime leukocyte imaging is usually performed 1 to 4 hours after injection, although positive scans can sometimes be obtained within 30 minutes or less. Because hepatobiliary excretion of exametazime results in the presence of nonspecific bowel activity in most patients imaged beyond 6 hours postinjection, such later delayed views are generally avoided. Results with early ^{99m}Tc exametazime-leukocyte imaging of inflammatory bowel disease may be superior to those for ^{111}In leukocytes, [106] particularly for less severe disease, where a low sensitivity for the latter agent has been reported. [107] Results for early diagnosis of acute appendicitis in patients with equivocal clinical presentations have also been promising. [108]

Other applications of labeled leukocytes in the abdomen include the identification of abscesses, both intra-abdominal and retroperitoneal, with SPECT sometimes useful for correlation with CT and MR findings. Labeled leukocyte imaging can also confirm the diagnosis of acute cholecystitis in patients with chronic or acalculous gallbladder disease.

Gallium 67 is a radionuclide which avidly binds to iron-binding proteins such as transferrin, and localizes at sites of inflammation and infection primarily due to increased capillary permeability. [109], [110] Once a common nuclear medicine imaging agent, gallium is now typically limited to studies for detecting opportunistic infections (including in the bowel) in immune-compromised patients (e.g., human immunodeficiency infection, organ transplant).

Oncologic Applications

The rapid growth of oncologic PET during the past decade has dramatically altered the role of radionuclide methods in tumor imaging. [111] Whereas nuclear medicine imaging traditionally played only a peripheral, supplementary role in evaluating patients for the presence and extent of malignancies, PET has now taken on a central role in many assessments for staging newly diagnosed tumors and evaluating patients suspected of recurrent disease. With the growth of PET, there has been a decline in use of other radionuclide procedures with established use in oncologic imaging, including gallium scans and newer studies employing radiolabeled monoclonal antibody agents. While gastroenterologic applications of PET are described in Chapter 157, the following discussion of the other still-available and potentially useful oncologic nuclear medicine procedures includes mention of those circumstances where PET would likely be the superior imaging choice.

The most commonly employed adjunctive nuclear medicine scan in oncology has historically been the ^{99m}Tc diphosphonate bone scan, which after four decades in clinical use remains the most effective imaging method for assessing the entire skeleton for malignant involvement (primary or metastatic). [112], [113] Although skeletal involvement by GI tract malignancies is less common than for tumors of the lung, breast, and genitourinary tract, the need to evaluate patients with GI tumors and signs or symptoms potentially reflecting involvement of bone periodically arises. Patients presenting with bone pain, with or without elevation of tumor markers such as carcinoembryonic antigen (CEA) or alkaline phosphatase, will typically undergo radiographic examination of the symptomatic area. Total-body bone scintigraphy, being more sensitive than plain radiographs for detection of metastases, [112], [114] is often obtained next if the radiographic study is negative or equivocal. Even in the circumstance where the radiographic or even an MR imaging examination shows definite evidence of metastatic disease, the bone scan can be helpful for assessing the extent of metastatic involvement throughout the skeletal system. In a review of a 14-year bone scintigraphy clinical database, 114a among 86 patients with history

of colorectal cancer and 37 patients with esophageal cancer evaluated primarily because of bone pain or suspicious radiographs, metastases were identified in 15 (17%) and 10 (27%) patients respectively (Fig. 158-6). These results indicate that in appropriately selected patients, the bone scan remains a useful procedure for identifying or confirming the presence of metastatic disease.

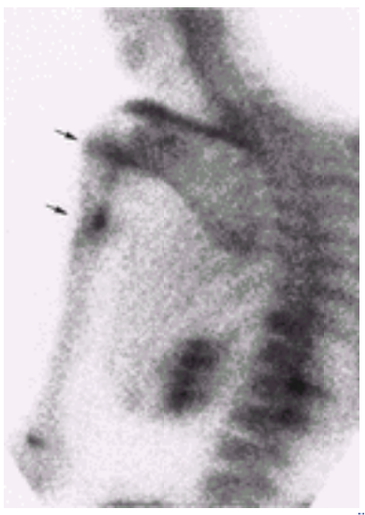

FIGURE 158-6. Bone scan images from a patient with esophageal carcinoma. Posterior oblique image of the left humerus in a 61-year-old man with persistent left shoulder and arm pain and negative radiographs of the area demonstrates foci of increased uptake at the left humeral neck and toward the medial posterior aspect of the proximal left humeral shaft (*arrows*). Metastatic esophageal carcinoma was subsequently confirmed following a pathological fracture of the proximal left humerus requiring surgical fixation.

Another compound with a long history as a tumor imaging agent is ^{67}Ga citrate, whose GI applications have included identification of hepatocellular carcinoma [115] and assessment of the extent of abdominal involvement by lymphoma. [116] Widespread availability of 2-deoxy-2-[^{18}F]fluoro-D-glucose (^{18}F-FDG) PET has substantially reduced the role of gallium imaging, despite occasional reports of tumors with increased gallium and normal FDG uptake. [117] While gallium remains an option for sites without PET capability, it is likely that its use will continue to decline, with limitations on supply becoming more severe as production is decreased in parallel with demand.

Another category of radiopharmaceuticals for imaging of malignancy, the monoclonal antibodies, [118], [119] has not had the impact which developers at one time envisioned. In the best multicenter trials, these agents directed against specific cellular antigens have typically been able to identify primary and metastatic lesions with sensitivity and specificity less than 80%. This mediocre performance can be attributed to a number of factors, including the difficulty of producing antibodies of high specificity, the heterogeneity of tumor tissues with respect to cellular antigens, and the sometimes limited access of intravenously administered substances to viable tumor tissue. Although PET has already superceded monoclonal antibody imaging in most large centers, the two U.S. Food and Drug Administration (FDA)-approved agents with applications to gastroenterology are briefly described subsequently.

Antibody agents for diagnostic usage fall into two broad categories, whole antibodies and antibody fragments. The former have a relatively long intravascular residence time, and are primarily cleared by the liver and excreted via the hepatobiliary system. An approved agent in this category with GI applications was satumomab (Oncoscint, Cytogen, Princeton, NJ), an anti-TAG (tumor-associated glycoprotein)-72 antibody labeled with ^{111}In which was indicated for detection of colorectal carcinoma (sensitivity and specificity 69% and 77%, respectively). [3] However, distribution of this agent was discontinued at the end of 2002. Antibody fragments (usually Fab) are smaller protein structures that are cleared more rapidly from the circulation and eliminated primarily via the kidneys, although some liver uptake and gut excretion may occur. The approved agent in this category of relevance to gastroenterology is the ^{99m}Tc labeled anti-CEA antibody arcitumomab (CEA-Scan, Immunomedics, Morris Plains, NJ), a compound with reported sensitivity for recurrent or metastatic colorectal carcinoma of 78% for the abdomen outside the liver and 73% for pelvic lesions. [4] For metastases in the liver, anti-CEA antibody imaging may demonstrate foci of increased or decreased uptake, with the photopenic defects (sometimes surrounded by a rim of increased activity) seen more often with larger lesions, likely as a result of inadequate delivery of the agent to hypoperfused or necrotic tissue. [4], [119] Reports of discordant CEA-Scan and ^{18}F-FDG imaging (one positive, the other negative) leave open a possible continuing role for antibody imaging in tumors with low metabolic activity.

A category of radiopharmaceuticals that has moved into the forefront of development for new nuclear medicine imaging agents is the radiolabeled peptides. As these compounds are of nonbiologic origin, they have several advantages over monoclonal antibodies, including greater flexibility in the molecular design of targeted compounds and lower risk for potential immune-mediated complications. Peptide agents can be designed to bind to specific cell surface receptors, creating possibilities for not only targeted diagnostic applications, but also therapeutic applications. The peptide-imaging agent of particular interest in gastroenterologic oncology is ^{111}In pentetreotide (OctreoScan, Mallinckrodt, Hazelwood, MO), whose primary component is octreotide, a somatostatin analog which binds to cell surface somatostatin receptors.

OctreoScan is used for imaging neuroendocrine tumors whose cells possess somatostatin receptors, including endocrine pancreatic tumors such as gastrinomas, insulinomas, islet cell tumors, glucagonomas, and carcinoid tumors. [120], [121], [122], [123], [124], [125], [126], [127], [128], [129] and [130] Somatostatin receptor imaging has demonstrated sensitivities as high as 90% in patients with gastrinomas and 80% to 100% in patients with carcinoid tumors [131] (Fig. 158-7). Octreotide imaging has proven particularly valuable for detection of functioning occult metastases, frequently outperforming conventional anatomic imaging methods such as ultrasound, CT, and MR imaging in these often difficult localizations. [126], [129], [130] As a further aid in this process, an intraoperative ?-detector can be used to localize sites of increased labeled octreotide uptake, thereby facilitating surgical removal of small deposits of tumor tissue. [132], [133]

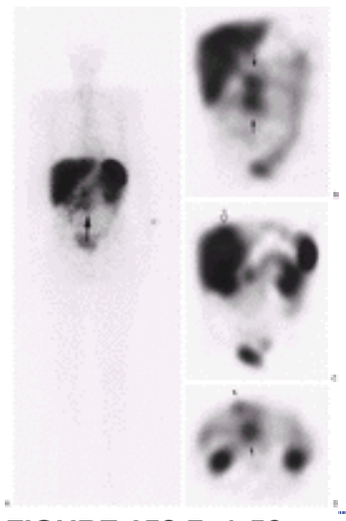

FIGURE 158-7. A 53-year-old man with symptoms suggestive of a carcinoid tumor, including progressively worsening diarrhea and facial and extremity flushing, and abnormalities on abdominal computed tomography (CT), underwent indium 111 octreotide scintigraphy as part of preoperative evaluation. Anterior whole-body image (**A**) shows normal physiological uptake in the liver, spleen, kidneys, colon, and bladder, and abnormal uptake in the right mid-abdomen (*arrow*). This abnormality is better defined on coronal single photon emission computed tomography (SPECT) image (**B**), and correlated with mesenteric deposits of tumor seen on CT. A slightly

more posterior coronal image (**C**) demonstrates a focus of increased uptake superiorly in the liver (*open arrow*), while an axial SPECT image (**D**) shows a second liver abnormality anteriorly in the right lobe (*arrowhead*), as well as the mesenteric mass (*arrow*). The two liver foci corresponded to hypervascular lesions seen on CT. At exploratory laparotomy, a cecal carcinoid with liver and mesenteric metastases was found.

NONIMAGING USES OF RADIOTRACERS

While most routine uses of radiotracers in nuclear medicine involve imaging of internal organ anatomy and physiology, some nonimaging uses of radiotracers can also provide valuable diagnostic information, several of which relate to the GI tract.

The Schilling test is used to ascertain whether vitamin B_{12} deficiency is the result of intrinsic factor deficiency or intestinal malabsorption of other causes. In part I of the test, Vitamin B_{12} labeled with an isotope of cobalt is administered orally in a capsule, and the presence of adequate intrinsic factor in the stomach is judged based upon the amount of labeled B_{12} that is absorbed and excreted in the urine. In an individual with normal secretion of intrinsic factor by the gastric parietal cells, more than 9% of an ingested dose of labeled B_{12} will be excreted in the urine in 24 hours. If a low amount of labeled B_{12} is measured in a 24-hour urine specimen, especially if the amount is less than 4% of the ingested dose, part II of the test is performed, which involves a second 24-hour urine collection following ingestion of radiolabeled B_{12} and intrinsic factor together. If B_{12} malabsorption reflects pernicious anemia/intrinsic factor deficiency, there will be normal B_{12} excretion on the second part of the test. If, on the other hand, B_{12} deficiency is secondary to malabsorption, such as due to bacterial overgrowth or a mucosal deficiency in the terminal ileum, the amount of labeled B_{12} excreted in the 24-hour urine collection will remain low. Both parts of the Schilling test can also be done in one day via simultaneous administration of two different forms of B_{12}—one labeled with cobalt 58 and with no intrinsic factor, the other labeled with cobalt 57 and also containing intrinsic factor. The 24-hour urine collection is performed, and dual isotope measurements are made. The part I Schilling test is sometimes repeated after treatment of the presumed cause of B_{12} deficiency to determine if the mucosal effects of this deficiency have been reversed. This follow-up study is sometimes called a Schilling test part III.

Bacteria that produce high levels of urease, such as *Helicobacter pylori,* can result in the breakdown of ingested C-14 urea, with production of C-14-labeled carbon dioxide which can be quantified in exhaled gases. The C-14 urea breath test is now widely used to assess for the presence and severity of *H pylori* infection.[134] The commercial test kit consists of a capsule containing 1 µCi C-14 labeled urea, which is ingested by the patient, and a Mylar balloon for collecting a sample of exhaled air (PYtest, Ballard Medical/Kimberly Clark, Draper, UT). C-14 activity is determined by counting the sample in a liquid scintillation counter, with a high value (typically =200 disintegrations/min) strongly indicative of the presence of *H pylori*.

SUMMARY

Nuclear medicine applications in gastroenterology take advantage of the quantitative and physiologically relevant information provided by this relatively noninvasive modality. Future improvements in ?-cameras and SPECT systems, advances in radiopharmaceutical development, and expanded availability of PET should insure that nuclear medicine remains at the forefront in the imaging of physiological processes and the new field collectively described as molecular medicine.

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CHAPTER 159

Kyung J. Cho

ANGIOGRAPHY

Since the introduction of the newer imaging modalities (including ultrasound [US], computed tomography [CT], and magnetic resonance [MR] imaging), diagnostic angiography has been used less frequently in the evaluation of gastrointestinal (GI) disease. Angiography is currently used to establish a specific diagnosis of mesenteric vascular disease, to localize GI bleeding, to evaluate portal hypertension, to assess tumor extension, to demonstrate traumatic arterial injury, and to obtain the necessary vascular information before percutaneous or surgical intervention. This chapter reviews the vascular anatomy as well as the techniques and role of angiography in the diagnosis and treatment of GI disease.

TECHNICAL CONSIDERATIONS

Conventional film-screen angiography has been largely replaced by digital subtraction angiography (DSA) in GI angiography. DSA requires a smaller amount of dilute contrast medium, causes less discomfort to patients, and reduces the risks for renal toxicity and fluid overload. DSA images may be viewed on a video monitor during and after injection of contrast material.

Two types of iodinated contrast material have been used for angiography: the conventional high-osmolar (ionic) contrast agent has been replaced by the low-osmolar (nonionic) contrast agent. The nonionic contrast agents are more expensive but cause less pain and burning sensation. Patients with a history of contrast media allergy and renal failure benefit from the use of low-osmolar contrast media. [1], [2] and [3]

Carbon dioxide (CO_2) gas has been used as a contrast agent. CO_2 is about 20 times more soluble than air, allowing the intravascular injection of the gas without causing clinically significant embolism. CO_2 angiography is useful for patients with a history of contrast allergy or renal failure. [4] CO_2 can be used as a contrast agent for visualization of the abdominal aorta; renal, mesenteric, and peripheral arteries; and portal and hepatic veins. CO_2 is used as a contrast agent for the diagnosis of GI bleeding, mesenteric arterial occlusive disease, and wedged hepatic venography for visualization of the portal vein. CO_2 is also used as a contrast agent to guide a variety of vascular interventions. [5], [6] and [7] Gadolinium-based contrast medium is used as an alternative contrast to iodinated contrast material for patients with a history of contrast hypersensitivity or renal failure. [8], [9]

Percutaneous Catheterization

Angiography is performed using the percutaneous technique introduced by Seldinger in 1953. [10] The arteries used for GI angiography are the femoral, axillary, and brachial. Of these, the femoral approach is preferable. When the needle is introduced into the artery, the guidewire is inserted through the needle into the abdominal aorta, and a diagnostic catheter is introduced over the guidewire. The technique used for femoral vein catheterization is similar to that used for arterial introduction. The guidewire is inserted into the inferior vena cava, and the catheter is advanced over the guidewire. If thrombosis has occurred in the femoral veins or inferior vena cava, the catheter is introduced by way of an antecubital, brachial, or jugular vein.

General Angiographic Approach

On the day of the procedure, the patient is allowed to take fluids by mouth. Patients usually receive conscious sedation with a narcotic analgesic (Fentanyl) and a benzodiazepine central nervous system depressant (Versed) immediately before and during the procedure. Biplane aortography is performed using a pigtail catheter for the evaluation of the origins of the celiac and superior mesenteric arteries. Otherwise, visceral angiography begins with catheterization of the celiac and superior mesenteric arteries. Selective catheterization of the branches of the visceral arteries is performed as needed: splenic, dorsal pancreatic, and gastroduodenal arterial catheterization for the evaluation of pancreatic disease, and left gastric arterial catheterization for gastric bleeding.

Visualization of the portal venous system is essential in angiographic study for evaluating intra-abdominal masses, vascular diseases, and portal hypertension. The presence or absence of a venous abnormality plays an important role in determining whether the arterial abnormality is neoplastic or arteriosclerotic in nature. Arterial involvement by a neoplasm is usually associated with narrowing or occlusion of the adjacent vein. Visualization of the portal vein requires the injection of a large volume of contrast medium into the superior mesenteric or splenic artery following the intra-arterial injection of 25 to 50 mg of the vasodilator tolazoline.

Under normal conditions, puncture site hemostasis can be achieved with manual compression for 5 to 10 minutes after percutaneous transarterial angiography. No pressure dressing is required. The patient may resume a normal diet after the procedure and ambulate after 4 hours of bed rest.

Transhepatic Portal Vein Catheterization

Percutaneous transhepatic portal vein catheterization [11] is a useful method for evaluating portal hemodynamics, localizing occult islet cell tumors, controlling gastroesophageal variceal bleeding, and performing balloon angioplasty of the portal vein stenosis and recanalization of occluded portal vein. Before puncture of the portal vein, portal vein patency is verified by US, CT, or MR angiography. The venous phase of a celiac or superior mesenteric angiogram is also useful in visualizing the portal vein. Wedged hepatic venography with CO_2 may be performed to visualize the portal vein. After the procedure has been completed, the catheter track is sealed near the hepatic capsule with a gelatin sponge or coil to arrest bleeding from the puncture site.

Transjugular Portal Vein Catheterization

The transjugular approach to the portal venous system is commonly used to create a transjugular intrahepatic portosystemic shunt (TIPS). [12], [13] The right internal jugular vein is punctured under US guidance. A 10-French vascular sheath is advanced into the right atrium. A 16-gauge Colapinto needle is used to puncture the right portal vein near the portal vein bifurcation under fluoroscopic control. When the portal vein is entered, a guidewire is introduced into the portal vein. Over the guidewire a 5-French catheter is advanced into the portal vein, and manometry is performed. For a TIPS procedure, the parenchymal tract is dilated with an angioplasty balloon catheter and covered with metallic stents.

Balloon Occlusion Hepatic Venography

The balloon occlusion method is useful in hepatic venography, facilitating measurement of the wedged hepatic venous pressure. Injection of contrast medium (4 to 5 mL/second for a total volume of 10 to 20 mL) or CO_2 (30 to 50 cc) into the hepatic vein distal to balloon occlusion results in visualization of the portal vein. [14], [15] and [16]

Splenoportography

Since the advent of the imaging modalities and the development of indirect portography (arterial portography), splenoportography is rarely performed. CO_2 is a useful contrast agent for splenoportography. Because of the low viscosity, the gas can be injected using a 22- or 25-gauge needle. Because of the relative safety of the small needle and the lack of the nephrotoxicity of the CO_2, CO_2 splenoportography should be useful in pediatric patients.

RISKS AND CONTRAINDICATIONS

The overall complication rates of transfemoral and transaxillary angiography are 1.73% and 3.29%, respectively. [17] The complications of transfemoral angiography include puncture site complications, complications related to catheter manipulation, contrast material reactions, contrast material toxicity (renal failure), and systemic complications (cardiac and neurologic). Brachial nerve injury is the most serious complication of transaxillary artery catheterization. Nerve injury is less likely to occur with a brachial artery puncture because the brachial plexus is not as close to the brachial artery as it is to the axillary artery. The use of intra-arterial nitroglycerin and calcium-channel blocking drugs is helpful in preventing arterial spasm. Mortality related to the angiographic procedures other than reactions to contrast agents is extremely rare.

The overall mortality associated with the intravenous use of the ionic contrast agents is 1 in 40,000. [18] The risk factors involved in the use of contrast material are renal failure, a history of previous reactions (major and minor), and allergic diathesis. Before the administration of contrast medium for angiography, risk factors for contrast nephropathy should be identified. These include preexisting renal insufficiency, diabetes mellitus, intravascular volume depletion, congestive heart failure, repeated contrast procedures, and multiple myeloma. [19] Recommendations for prevention of contrast nephropathy include use of alternative contrast such as gadolinium-based contrast or CO_2, discontinuation of potential nephrotoxic agents 48 to 72 hours before the procedure, adequate hydration before and after the procedure, use of low-osmolar or iso-osmolar contrast material, and use of minimal volume of contrast material. Pretreatment with 32 mg of methylprednisolone 12 and 2 hours before contrast administration has been advocated to prevent reaction to contrast agents for patients with a history of this complication.

There are no absolute contraindications to GI angiography. Relative contraindications include severe coagulopathy, recent myocardial infarction, congestive heart failure, renal failure, and pregnancy. Patients undergoing brachial or axillary artery puncture or a percutaneous transhepatic procedure are at increased risk for hemorrhagic complications in the presence of coagulopathy or hypertension. Depending on the urgency and nature of the procedure, coagulopathies should be reversed with appropriate treatment.

VASCULAR ANATOMY OF THE ABDOMINAL VISCERA

A thorough knowledge of vascular anatomy of the abdominal viscera is essential in performing and interpreting an angiogram and in planning therapeutic intervention for GI disease.

Arterial Vasculature

The abdominal viscera receive their blood supply from the celiac, superior, and inferior mesenteric arteries. The superior mesenteric artery arises from the ventral aspect of the abdominal aorta 1 to 2 cm below the celiac and above the renal artery. It courses anterior to the third portion of the duodenum and the left renal vein into the mesentery. The celiac artery gives off the left gastric, splenic, and common hepatic arteries (Fig. 159-1). Occasionally, the inferior phrenic or dorsal pancreatic artery originates from the celiac axis. The inferior phrenic artery may arise from the left gastric artery or renal artery, or directly from the aorta. One or more branches of the celiac artery may originate from sources other than the celiac axis: the left gastric artery from the aorta, the splenic artery from the superior mesenteric artery, and the hepatic artery from the aorta, superior mesenteric artery, gastroduodenal artery, or left gastric artery.

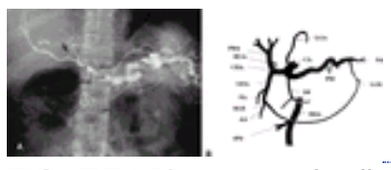

FIGURE 159-1. Normal celiac trunk. **A:** Celiac arteriogram showing standard arterial anatomy. The left gastric (*LGA*), splenic (*SA*), and common hepatic (*CHA*) arteries arise from the celiac trunk (*CA*). The proper hepatic artery (*PHA*) divides into the right and left hepatic arteries. The middle hepatic artery (*arrow*) arises from the right hepatic artery. The right gastric artery (*RGA*) arises from the PHA. **B:** Line drawing of branches of the celiac trunk. *DP,* dorsal pancreatic artery; *PM,* pancreatica magna artery; *LGE,* left gastroepiploic; *PA,* posterior arcade; *AA,* anterior arcade; *RGE,* right gastroepiploic; *IPD,* inferior pancreaticoduodenal; *SMA,* superior mesenteric artery; *GDA,* gastroduodenal artery.

The common hepatic artery divides into the proper hepatic and gastroduodenal arteries. The proper hepatic artery ascends in the hepatoduodenal ligament for a variable distance and divides into a right and a left hepatic artery. The right hepatic artery usually courses behind the common hepatic duct and divides into an anterior and posterior segmental artery. Each of the segmental arteries gives off arterial branches to the superior and inferior subsegments of the liver. The left hepatic artery divides into a medial segmental (middle hepatic) and a lateral segmental artery. Each segmental artery gives rise to superior and inferior subsegmental branches. The middle hepatic artery may arise from a right hepatic, a left hepatic, or a proper hepatic artery. The proper hepatic artery also gives off arterial branches to the bile duct, the portal vein (vasa vasorum), and the subcapsular branches. These nonparenchymal branches may provide collateral circulation to the liver in hepatic arterial occlusion distal to the gastroduodenal artery.

In about half of cases, one or more branches of the hepatic artery arise from sources other than the celiac-hepatic artery. Two types of aberrant hepatic artery may occur: replaced (the origin of the artery is “replaced” and the aberrant artery serves as a substitute), or accessory (additive to the celiac-hepatic artery). According to Michels’ dissection of 200 cadavers, [20] aberrant right hepatic arteries occur in 26% of patients (replaced, 18%; accessory, 8%), most frequently originating from the superior mesenteric artery (17%); aberrant left hepatic arteries occur in 27% of patients (replaced, 15.5%; accessory, 11.5%), most frequently from the left gastric artery (13%).

The gastroduodenal artery usually originates from the common hepatic artery. In 25% of the population, it may have an aberrant origin. The three main branches originating from the gastroduodenal artery are the posterior superior pancreaticoduodenal (posterior arcade), anterior superior pancreaticoduodenal (anterior arcade), and right gastroepiploic arteries. The posterior and anterior arcade arteries join inferomedially and anastomose with the inferior pancreaticoduodenal artery, a branch of the superior mesenteric artery. The gastroduodenal artery supplies arterial blood to the stomach, duodenum, pancreas, and bile duct. In the presence of a celiac artery occlusion, the pancreaticoduodenal arcade arteries function as the major collateral pathway to the liver from the superior mesenteric artery. Normally, the gastroduodenal blood flows away from the liver (hepatofugal), but with stenosis or occlusion in the celiac or common hepatic artery, the blood flow is reversed (hepatopetal). Preoperative recognition of flow reversal in the gastroduodenal artery is important in planning resection of pancreatic head tumors and for placement of a hepatic artery infusion catheter for chemotherapy.

The superior mesenteric artery supplies the pancreas, duodenum, small intestine, cecum, ascending colon, and proximal half of the transverse colon. The inferior pancreaticoduodenal and occasionally the dorsal pancreatic arteries originate from the proximal portion of the superior mesenteric artery. The other branches of the superior mesenteric artery are the middle colic, jejunal, ileal, right colic, and ileocolic arteries (Fig. 159-2). A true right colic artery is an inconstant branch (present in 13% of people), and the ascending colon often receives its blood supply from a paracolic arcade fed from the middle and ileocolic arteries. [21] However, Michels and colleagues [22] asserted that any branch proximal to the paracolic arcade that supplies the ascending colon is a right colic artery. With this definition, the right colic artery arises from the superior mesenteric artery in 38%, from a common right colic–middle colic trunk in 52%, and from an ileocolic–right colic trunk in 8% of individuals. The right colic artery is absent in 2% of individuals, and an accessory right colic artery is present in 8% of individuals.

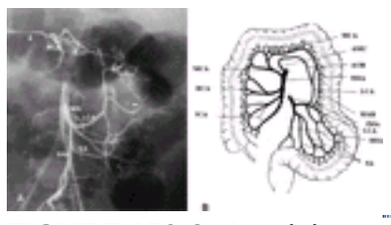

FIGURE 159-2. Arterial supply to the colon. **A:** Arterial phase of an inferior mesenteric angiogram. The inferior mesenteric artery (*IMA*) gives off the left colic (*LCA*), sigmoidal (*SA*), and superior hemorrhoidal (*SHA*) arteries. The middle colic (*MCA*) branch of the superior mesenteric artery anastomoses with the left colic branch of the inferior mesenteric artery through the marginal artery of Drummond (*arrowheads*). Opacification of the middle colic artery is due to forceful injection of contrast medium into the inferior mesenteric artery. **B:** Line drawing of the arterial supply to the colon. *RCA,* right colic artery; *ICA,* ileocolic artery; *AOR,* arc of Riolan; *MCA,* middle colic artery; *SMA,* superior mesenteric artery; *LCA,* left colic artery; *IMA,* inferior mesenteric artery; *SHA,* superior hemorrhoidal artery; *SA,* sigmoidal artery; *AMC,* accessory middle colic artery; *MAR,* marginal artery of Drummond.

The inferior mesenteric artery arises from the anterolateral aspect of the aorta near the level of the L3-4 interspace and courses caudally and to the left for up to 5 cm before giving off the left colic artery (see Fig. 159-2). The ascending branch of the left colic artery supplies the descending colon and a variable amount of the splenic flexure. The inferior mesenteric artery gives off several additional branches to the descending and sigmoid colon before terminating in the superior hemorrhoidal artery. The inferior mesenteric artery communicates through the left colic–middle colic anastomosis to the superior mesenteric artery. It also communicates through the superior hemorrhoidal–middle and inferior hemorrhoidal arterial anastomoses to the internal iliac artery. They function as collaterals in mesenteric arterial or distal aortic occlusion.

Portal Venous System

The portal vein is formed by the junction of the splenic and superior mesenteric veins behind the head of the pancreas (Fig. 159-3) and ascends toward the hepatic hilus in the hepatoduodenal ligament, dorsal and to the left of the bile duct, and to the right of the hepatic artery. The portal vein is joined by the left gastric, right gastric, posterior superior pancreaticoduodenal, and cystic veins. These tributaries provide the portosystemic collaterals in patients with cirrhosis and portal hypertension. The right branch of the portal vein divides into the anterior and posterior segmental branches. The left portal branch divides into the superior and inferior subsegmental branches after giving off branches to the caudate and quadrate lobes. The umbilical vein joins the left portal vein at its bifurcation into the subsegmental branches. It is normally obliterated but may be recanalized in the presence of portal hypertension. The inferior mesenteric vein joins the splenic vein or, less frequently, the superior mesenteric vein. Portosystemic collaterals develop from the inferior mesenteric vein through the superior hemorrhoidal vein to the branches of the internal iliac vein and through the retroperitoneal vein to the inferior vena cava.

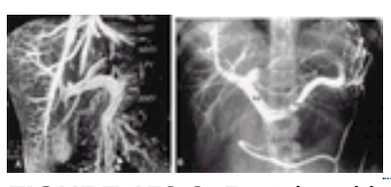

FIGURE 159-3. Portal and hepatic veins. **A:** Magnetic resonance (MR) angiography and three-dimensional contrast MR venography. The celiac and superior mesenteric arteries were normal (not shown). Imaging of the upper abdomen in the coronal plane was performed after intravenous administration of gadolinium. The superior mesenteric, portal, and hepatic veins are visualized. *SMV,* superior mesenteric vein; *MPV,* main portal vein; *RPV,* right portal vein; *LPV,* left portal vein; *RHV,* right hepatic vein; *LHV,* left hepatic vein; *MHV,* middle hepatic vein. **B:** Percutaneous transhepatic splenoportogram with injection of contrast medium into the proximal splenic vein. *LG,* left gastric vein; *GE,* gastroepiploic vein; *SV,* splenic vein; *PV,* portal vein. Venous anastomoses are seen between the short gastric and left

gastric veins and between the left and right gastroepiploic veins.

The portal venous system receives venous blood from the GI tract, the pancreas, the spleen, the gallbladder, and the omentum. Normally, the portal vein blood flows toward the liver (hepatopetal). Reversal in flow (hepatofugal) in any tributaries of the portal venous system indicates the presence of portal hypertension; visualization of the coronary vein or inferior mesenteric vein in the portal venous phase of a superior mesenteric angiogram indicates reversal in flow of these veins functioning as portosystemic collaterals. Reversal of portal vein flow may be partial or complete. Partial reversal of portal flow may occur in the extrahepatic or intrahepatic branches of the portal venous system. Complete reversal of the intrahepatic portal flow may occur in some patients with severe cirrhosis, hepatic vein obstruction, surgical portosystemic shunts, and TIPS. In such cases, the portal vein is the principal outflow conduit of the liver, and the hepatic artery blood leaves the liver through the portal vein. Blood flow direction in the intrahepatic and extrahepatic portal veins can be assessed by Doppler US, direct transhepatic portal vein catheterization, superior mesenteric and celiac angiography, and wedged hepatic venography. When the main portosystemic collateral develops from the umbilical vein, the extrahepatic portal blood flow can be hepatopetal in the presence of hepatofugal intrahepatic portal flow.

CT, US, and MR venography can be used to visualize accurately the portal venous system. [23], [24] and [25] The portal and hepatic venous imaging can be obtained immediately after MR angiography with gadolinium enhancement for arterial examination (see Fig. 159-3). The method is useful when a TIPS procedure, liver transplantation, or resection of intra-abdominal tumors is contemplated.

Hepatic Veins

The hepatic veins begin in the center of the hepatic lobules as intralobular veins. These veins join together to form sublobular veins. The hepatic veins are intersegmental or interlobar in course: the right hepatic vein lies in the intersegmental fissure of the right hepatic lobe, dividing it into the anterior and posterior segments; the middle hepatic vein lies in the interlobar fissure; and the left hepatic vein lies between the medial and lateral segments of the left hepatic lobe. The hepatic veins converge posteriorly and run near the hepatic capsule before emptying into the inferior vena cava (see Fig. 159-3).

The accessory hepatic veins, which originate from the right hepatic and caudate lobes, are small and empty into the inferior vena cava between the main hepatic and renal veins. They function as collaterals in hepatic vein occlusion and occlusion of the hepatic portion of the inferior vena cava. Diagnostic imaging of the hepatic veins may be obtained by spiral CT scanning, [26] US, [27] or MR angiography. [28]

ARTERIAL DISEASE

Arteriography remains the gold standard in the diagnosis of visceral arterial disease. It can provide a specific diagnosis and information on the vascular anatomy and arterial hemodynamics.

Acute Mesenteric Ischemia

Angiography is the most useful diagnostic examination for acute mesenteric ischemia and should be performed urgently (Fig. 159-4). After a lateral aortogram with a pigtail catheter has been obtained, celiac and superior mesenteric arteries are catheterized and injected with contrast medium. When colonic ischemia is suspected, an inferior mesenteric arteriogram is performed. When the celiac, superior, and inferior mesenteric arteries are occluded, the collateral blood supply to these arteries comes primarily from the pelvic branches of the internal iliac arteries through the inferior and middle hemorrhoidal–superior hemorrhoidal arterial anastomoses. Collaterals to the superior hemorrhoidal artery from the middle and inferior hemorrhoidal arteries can be demonstrated by injecting contrast medium into the distal abdominal aorta or internal iliac arteries.

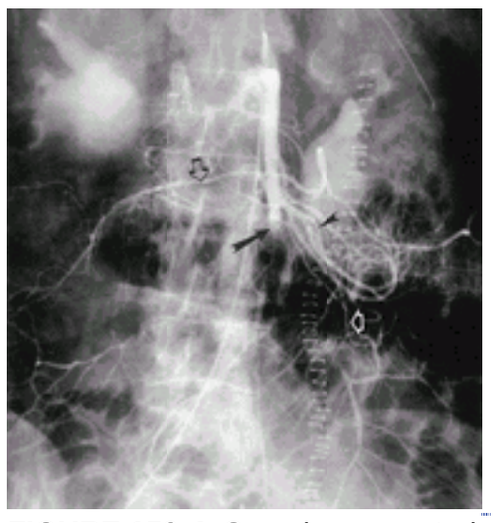

FIGURE 159-4. Superior mesenteric artery embolus. Superior mesenteric angiogram in an elderly woman with atrial fibrillation and abdominal pain. There is an embolus in the superior mesenteric artery (*black arrow*) distal to the second jejunal artery. A small embolus is seen in the jejunal artery (*arrowhead*). The distal mesenteric branches reconstitute through collaterals from the middle colic (*open black arrow*) and jejunal arteries (*open white arrow*).

Acute mesenteric ischemia may be occlusive or nonocclusive in type. The etiology of occlusive mesenteric ischemia includes embolism, atherosclerotic plaques, aortic dissection, neoplasms, and vasculitis. Arteriography can accurately identify the level of occlusion and evaluate collateral circulation. In superior mesenteric artery embolism, the occlusion may be proximal or distal to the middle colic artery. Arteriosclerosis usually involves the origin of the superior mesenteric artery, allowing collateral development through the marginal artery from the inferior mesenteric artery. If the inferior mesenteric artery is occluded, collateral circulation may originate from the middle and inferior hemorrhoidal arteries of the internal iliac arteries.

Nonocclusive mesenteric ischemia is caused by a significant reduction in mesenteric blood flow secondary to cardiac failure or hypovolemic shock. The typical angiographic findings include diffuse mesenteric arterial constriction, slowing of mesenteric arterial flow, and decreased intestinal mucosal staining. Intravenous glucagon or intra-arterial infusion of papaverine into the superior mesenteric artery through a percutaneous catheter is used to relieve mesenteric vasoconstriction.

Colonic Ischemia

Colonic ischemia may be caused by decreased perfusion to the bowel in a low-flow state or by surgical ligation of the inferior mesenteric artery during abdominal aortic reconstruction or abdominoperineal resection surgery. [29] The angiographic findings may be nonspecific: in the early stage, the mesenteric arteries are constricted, and blood flow is slowed with decreased parenchymal vascularity; in the late stage, the colon may appear hypervascular with prominent intramural arteries and increased accumulation of contrast material in the wall of the bowel. Preoperative angiography is necessary to assess the mesenteric circulation because ligation of the inferior mesenteric artery in the patient with superior mesenteric artery stenosis may result in mesenteric ischemia.

Intestinal Angina

At least two of the three splanchnic arteries usually have significant occlusive disease before the syndrome of intestinal angina occurs. [30] Duplex US with spectral analysis is used for screening patients thought to have chronic mesenteric artery occlusive disease. Gadolinium-enhanced three-dimensional (3D) MR angiography is sensitive in assessing patency of the origins of the celiac and superior mesenteric arteries, but its resolution is inadequate for evaluation of mesenteric artery branch stenosis. [31] Demonstration of a patent superior mesenteric artery or an insignificant stenosis of the superior mesenteric artery with patent celiac artery should exclude

the diagnosis of chronic mesenteric ischemia. [32]

Celiac Axis Compression

Celiac axis compression syndrome (median arcuate compression syndrome) is controversial as a cause of abdominal pain. The celiac axis may be narrowed or, in severe cases, occluded by the median arcuate ligament of the diaphragm. Surgical correction of the lesion by decompression or reconstruction of the vessel usually ameliorates the pain. Because balloon dilation is not helpful in eliminating the compression, stent placement is required. [33] Celiac axis stenosis frequently is an incidental finding, with a reported prevalence of 12% to 49%. [34], [35] Lateral aortogram demonstrates a concave impression on the cranial aspect of the celiac axis. The compression usually is accentuated during deep expiration. A superior mesenteric angiogram demonstrates collateral circulation to the hepatic and splenic arteries through the gastroduodenal and pancreatic arcade arteries from the superior mesenteric artery.

Superior Mesenteric Artery Syndrome

The superior mesenteric artery can compress the third portion of the duodenum, causing duodenal obstruction. Such compression is most often found in patients with chronic immobilization with weight loss and a body cast. [36], [37] The upper GI barium examination shows a characteristic oblique indentation toward the right lower quadrant on the third portion of the duodenum, corresponding to the course of the superior mesenteric artery. A narrow aortomesenteric angle corresponding to the site of obstruction is usually found on a lateral aortogram.

Vasculitis

The angiographic findings vary with the type of vasculitis. In polyarteritis nodosa, celiac and superior mesenteric angiograms demonstrate occluded small- and medium-sized arteries and microaneurysms (Fig. 159-5). In ergot or digitalis toxicity, the mesenteric artery branches are narrowed or occluded with collaterals. Angiography is the most sensitive method of visualizing microaneurysms. The angiographic examination should include the renal, celiac, and mesenteric arteries.

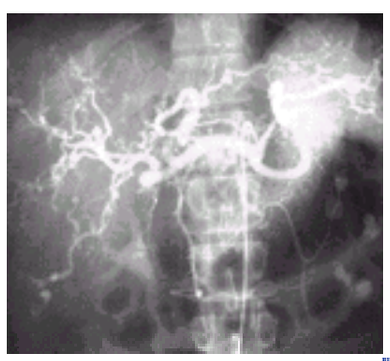

FIGURE 159-5. Multiple microaneurysms in polyarteritis nodosa. Arterial phase of a celiac angiogram shows numerous aneurysms involving the hepatic and pancreatic arteries.

Splanchnic Artery Aneurysms

Splanchnic artery aneurysms and pseudoaneurysms are rare and often found incidentally during angiographic studies for other indications. Angiography usually is necessary to identify the exact site of the aneurysm and its relationship to adjacent vascular structures as well as to plan surgical resection or embolization to prevent catastrophic hemorrhage. [38] Aneurysms occur in virtually all splanchnic arteries.

Splenic artery aneurysms account for 60% of all visceral artery aneurysms. [39] They occur four times more frequently in women than in men and are multiple in 20% of patients. Causes include atherosclerosis, medial fibrodysplasia, multiple pregnancies, pancreatitis, portal hypertension, polyarteritis nodosa, Ehlers-Danlos syndrome, and trauma. In portal hypertension, aneurysms tend to occur at the bifurcation of the intrasplenic branches of the splenic artery. Most atherosclerotic splenic artery aneurysms are asymptomatic, and calcified aneurysms less than 2 cm in diameter require no treatment. On the other hand, aneurysms occurring in pregnant women and those associated with pancreatitis require treatment because of their propensity to bleed. Splenic artery aneurysms can be treated with embolization by the transcatheter approach or percutaneous puncture.

The hepatic artery is the second most common site for splanchnic aneurysm, accounting for 20% of all splanchnic aneurysms. [39] Most hepatic pseudoaneurysms are traumatic in origin and are secondary to blunt or penetrating abdominal trauma, liver biopsy, or the placement of a transhepatic biliary drainage catheter or hepatic arterial infusion catheter. Most spontaneous aneurysms occur in the common hepatic or right hepatic artery. The potential complications of the hepatic artery aneurysms are bleeding into the bile duct or peritoneal cavity and rupture into the portal vein.

Mesenteric aneurysms are arteriosclerotic and may cause GI bleeding or mesenteric ischemia. Celiac axis aneurysms account for 4% of all visceral aneurysms and should be differentiated from pseudocysts when encountered during US or unenhanced CT scanning. A lateral aortogram is necessary to confirm the diagnosis of aneurysms arising from the origins of the celiac and superior mesenteric artery.

Gastroduodenal and pancreatic arterial aneurysms are associated with pancreatitis and pseudocysts. They may cause GI and intraperitoneal hemorrhage. Aneurysms rarely rupture into the pancreatic duct or a pseudocyst. Contrast-enhanced CT usually differentiates pseudocysts and aneurysms. Angiography is required to delineate the vascular anatomy and the exact origin of the aneurysm before surgical treatment or transcatheter embolotherapy.

Abdominal Trauma

Most traumatized patients require radiologic studies to assess the extent of the injury. CT is the initial study used for the evaluation of abdominal trauma. Angiography is indicated to determine the exact site and extent of vascular injury after CT has demonstrated evidence for vascular injury or when a penetrating injury has occurred in the vicinity of major vessels.

VENOUS DISEASE

Occlusion of the portal, mesenteric, splenic, and hepatic veins may be asymptomatic or associated with ascites, hepatic failure, or GI hemorrhage. The newer imaging modalities can diagnose occlusion of the portal and splenic vein by demonstrating intra-abdominal varices and aneurysms in the portal venous system. [24], [40] Duplex scanning is useful in the assessment of patency and direction of blood flow of the portal vein and portosystemic shunts. The portal venous phases of the celiac and superior mesenteric angiograms are used to evaluate portal vein occlusion and aneurysm. An inferior venacavogram and hepatic venogram are performed in patients with suspected Budd-Chiari syndrome.

Portal Vein Occlusion

Angiographic findings include large collateral veins originating from the superior mesenteric vein, ascending in the hepatoduodenal ligament in the venous phase of the superior mesenteric angiogram (Fig. 159-6). The collateral vessels reconstitute the patent intrahepatic portal vein. If the intrahepatic portal venous branches are occluded, the collateral vessels continue to run along the intrahepatic bile ducts, giving the appearance of railroad tracks. [41] In mesenteric vein thrombosis, numerous tiny collateral veins are demonstrated throughout the mesentery without visualization of the superior mesenteric vein in the venous phase of the superior mesenteric

angiogram.

FIGURE 159-6. Portal vein occlusion. Portal venous phase of a superior mesenteric angiogram (oblique view) demonstrates tortuous venous collaterals (*arrow*) running along the hepatoduodenal ligament adjacent to the occluded portal vein. The collateral veins reconstitute intrahepatic portal venous branches. (From Reuter SR, Redman HC, Cho KJ. Gastroenterology angiography. 3rd ed. Philadelphia: WB Saunders, 1986:121.)

The angiographic abnormality of portal vein occlusion due to neoplasms varies with the type of neoplasm. Pancreatic and biliary cancers cause localized narrowing or occlusion of the portal vein without associated tumor vessels. In contrast, portal vein invasion by hepatocellular carcinoma has a characteristic appearance, with abnormal vascular channels coursing within the portal vein branches in the vicinity of the tumors. Arteriovenous shunting is often associated with portal vein invasion by tumors. [42]

Splenic Vein Occlusion

Splenic vein occlusion usually is silent clinically but may cause hypersplenism or gastric variceal bleeding. [43], [44] The diagnosis can be made by dynamic CT, duplex US, and 3D contrast MR venography. Angiography should be performed when the noninvasive studies are inconclusive or when additional vascular information is needed. A celiac or a splenic arteriogram is performed for the diagnosis; the angiographic findings include splenoportal collaterals through the short gastric-coronary and gastroepiploic veins as well as nonopacification of the splenic vein during the venous phase.

Budd-Chiari Syndrome

In Budd-Chiari syndrome, a significant portion of the hepatic venous system is obstructed. CT usually is used as the initial test and may reveal nonuniform contrast enhancement of the liver parenchyma and enlarged caudate lobe. Increased central venous pressure from right heart failure may produce a contrast enhancement pattern similar to that of Budd-Chiari syndrome. MR imaging is useful in identifying the underlying lesions, such as hepatic and vena caval thrombi, and congenital membrane.

Angiography is the most important procedure for the diagnosis of the Budd-Chiari syndrome. Superior mesenteric and celiac angiograms are obtained to see the portal vein and exclude hepatic neoplasms. An inferior venacavogram is obtained to exclude occlusion of or membrane in the hepatic portion of the inferior vena cava. Demonstration of hepatic vein patency excludes the diagnosis of Budd-Chiari syndrome. When the hepatic vein is occluded, the catheter is wedged into the occluded hepatic vein, and contrast medium is injected to visualize the collateral channels (Fig. 159-7). If the right hepatic vein cannot be entered, catheterization of the accessory hepatic vein should be performed. If any of the hepatic veins cannot be catheterized, a 22-gauge needle is inserted into the liver parenchyma using the same technique used for percutaneous transhepatic portal vein catheterization. Injection of iodinated contrast medium or CO_2 usually demonstrates hepatic vein occlusion and collateral veins. [45]

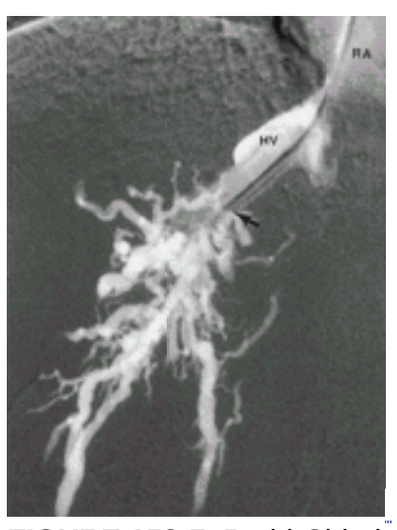

FIGURE 159-7. Budd-Chiari syndrome. Digital subtraction hepatic venogram was performed with contrast injection into the right hepatic vein (*HV*), demonstrating numerous collateral veins pathognomonic of hepatic vein occlusion. The *arrow* indicates the level of hepatic vein occlusion. *RA,* right atrium.

Percutaneous translumenal angioplasty and stent placement has been used to treat Budd-Chiari syndrome caused by obstruction of the hepatic segment of the inferior vena cava. [46], [47] Restenosis or reocclusion occurs frequently at the angioplasty site. Patency of the vein may be improved with use of metallic stents after unsuccessful angioplasty. The transhepatic approach may be used for angioplasty and stent placement for right hepatic vein occlusion causing Budd-Chiari syndrome. [48] The TIPS procedure has been used to treat intractable ascites and bleeding varices from hepatic venous occlusive disease and as an effective bridge to transplantation for hepatic failure associated with Budd-Chiari syndrome. [49]

GASTROINTESTINAL DISEASE

When evaluating GI disease, angiography is indicated for the diagnosis of vascular disease, visualization of vascular anatomy, and diagnosis and treatment of GI bleeding. If active GI bleeding is diagnosed, angiography can be urgently performed for localization of the bleeding site as well as control of the bleeding. The rate of bleeding should be at least 0.5 mL/minute to be detected by selective angiography. Angiography is insensitive in detecting capillary or venous bleeding because of the dilution of the contrast material. A recent study showed that helical CT angiography with intra-arterial injection of contrast medium into the abdominal aorta at the level of celiac axis is useful in detecting GI bleeding of obscure origin and facilitates angiographic localization of the bleeding. [50] Angiography is useful in identifying vascular malformations and vascular neoplasms. A catheter may be placed in the mesenteric branch supplying a vascular malformation preoperatively to facilitate intraoperative localization of the lesion.

Upper Gastrointestinal Bleeding

Once upper GI bleeding is diagnosed, celiac and superior mesenteric angiograms are obtained. If a bleeding site is identified, the bleeding artery is selectively catheterized to control the bleeding. If a bleeding site has not been demonstrated, left gastric and gastroduodenal arteriograms are obtained. If the source of bleeding still has not been identified and nasogastric aspiration reveals dark blood, a catheter may be placed in the left gastric artery for the next 6 to 12 hours. The bleeding may be precipitated by intra-arterial administration of heparin, vasodilators, or thrombolytic agents, allowing accurate localization of the bleeding site and prompt treatment. [51], [52] Prophylactic embolization (left gastric artery embolization for gastric bleeding and gastroduodenal artery embolization for duodenal bleeding) is an acceptable alternative when the massive bleeding has ceased at the time of angiography. [53]

Angiography usually is not helpful in determining the cause of upper GI hemorrhage. Hemorrhagic gastritis is the most common cause of capillary bleeding and may demonstrate a hypervascular stomach with dilated gastric arteries and increased parenchymal staining. Peptic ulceration is the most common cause of arterial bleeding and usually produces no angiographic abnormality. Gastric bleeding usually arises from the left gastric artery and occasionally from the short gastric, right gastric, or gastroepiploic artery. Duodenal bleeding may originate from the celiac or superior mesenteric artery or from both arteries. Angiographically, the arterial bleeding appears as a localized accumulation of contrast material extravasation during the arterial phase of the angiogram. The escaped contrast material persists throughout the venous phase and may outline the mucosa. Selective arterial embolization is an effective means of controlling bleeding from peptic ulceration.

Upper endoscopy should be obtained in all patients suspected of bleeding from the biliary tract (hemobilia) and pancreatic duct (hemosuccus pancreaticus). Once bleeding from the ampulla of Vater is documented, angiography is performed to demonstrate the underlying lesion and rarely contrast extravasation into the bile duct. Hemobilia can be treated by embolizing the feeding artery. Bleeding into the pancreatic duct is amenable to selective arterial embolization of the associated arterial lesions. Aortoenteric fistulae rarely cause GI bleeding, and the duodenum is the most common site of fistulae. Biplane aortography may show a false aneurysm at the aortic anastomotic site.

Percutaneous transhepatic coronary vein embolization may be used to treat variceal bleeding when endoscopy is not available or fails to control the bleeding. Variceal embolization may be necessary if the varices continue to fill after creation of TIPS. TIPS is an effective method of controlling variceal bleeding and is indicated when the bleeding is unresponsive to endoscopic therapy. [54], [55] and [56]

Lower Gastrointestinal Bleeding

Once lower GI bleeding is diagnosed, a superior mesenteric angiogram is performed to detect the bleeding from the small intestine and the right side of the colon. If no bleeding site is seen, a celiac arteriogram is obtained because gastroduodenal bleeding may present as lower GI bleeding. If a radionuclide scan has localized the bleeding to the left side of colon, inferior mesenteric arteriograms are obtained with a coverage area from the rectum to the splenic flexure of the colon. Active bleeding shows a localized accumulation of escaped contrast material during the arterial phase of the mesenteric angiogram, which usually persists through the venous phase (Fig. 159-8).

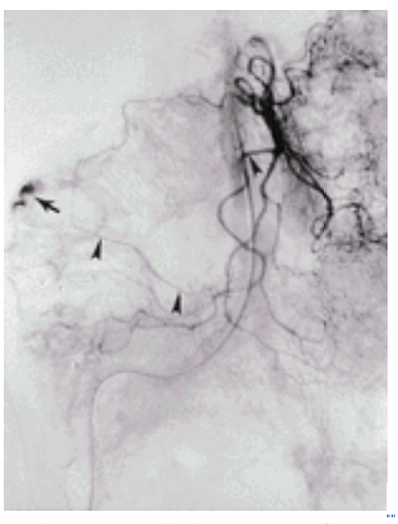

FIGURE 159-8. A superior mesenteric angiogram in an elderly man who developed massive colonic bleeding. Extravasation of contrast medium is seen from the hepatic flexure of the colon (*arrow*). The bleeding was controlled with embolization of the bleeding branch from the right colic artery (*arrowheads*).

Selective infusion of vasopressin is usually effective in controlling the bleeding in up to 80% of patients, but the recurrence rate is as high as 50%. [57] Patients with coronary artery disease should not be treated with vasopressin because of the risk for myocardial infarction. Selective embolization is an acceptable alternative when vasopressin is contraindicated. Superselective catheterization and embolization of the bleeding artery can be achieved with use of the 3-French coaxial catheter system with minimal risk for intestinal necrosis. [58], [59]

Angiodysplasia Angiodysplasia may occur in a variety of pathological types: vascular ectasia, arteriovenous malformations, and capillary telangiectasia. [60] The clinical presentation and angiography allow identification of each type of angiodysplasia. Vascular ectasia usually occurs in the cecum and ascending colon. Most patients are older than 60 years of age. Angiography is useful in identifying the bleeding source. The angiographic findings are usually diagnostic and include a small vascular cluster and blush in the wall of the colon and early, dense opacification of the draining vein [61] (Fig. 159-9). Normal mesenteric angiography usually is accepted for exclusion of vascular ectasia, although data supporting this assumption are not available. Two or more vascular ectasias are frequently present at microscopic examination of the injected specimen, although most patients demonstrate a single vascular lesion at angiography. Endoscopy is superior to angiography in identifying vascular ectasia. If active extravasation is demonstrated, selective arterial embolization is an effective treatment. Arteriovenous malformations are of developmental origin and usually affect people younger than 50 years of age. The angiographic findings include tortuous, dilated arteries with early, prominent veins. The lesions frequently are found in the small intestine and vary in size. Vascular malformations should be differentiated from vascular tumors, such as leiomyomas.

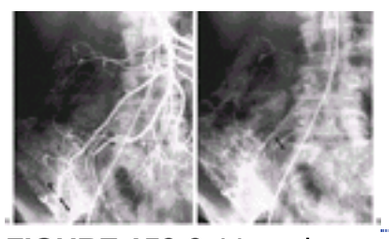

FIGURE 159-9. Vascular ectasia of the cecum. **A:** Arterial phase of superior mesenteric angiogram in a 70-year-old woman with recurrent lower gastrointestinal bleeding. A small vascular lesion (*open arrow*) is demonstrated in the antimesenteric border of the cecum with simultaneous opacification of an entering artery (*arrow*) and a draining vein (*arrowhead*). **B:** Capillary phase of the same angiogram. Notice the early, densely opacified draining vein (*arrow*). The veins from other parts of the bowel have not been opacified.

Capillary telangiectasia may occur in any part of the GI tract and may be associated with Osler-Weber-Rendu disease. The angiographic findings usually include tiny areas of blush without arteriovenous shunting. Hepatic angiography may demonstrate dilated hepatic arteries and increased hepatic parenchymal staining.
Meckel Diverticulum Angiography is obtained only when active bleeding occurs. Angiographically, a Meckel diverticulum appears as a short segment of bowel with increased contrast accumulation supplied by an abnormal branch from the superior mesenteric artery. [62]
Rectal Trauma Inferior mesenteric angiography is performed for localization and treatment of the bleeding. Infusion of vasopressin into the inferior mesenteric artery

is effective in controlling the bleeding. If the patient has coronary artery disease or if vasopressin fails, selective embolization is performed with Gelfoam, Ivalon particles, or microcoils. [63]

Intestinal Varices Scintigraphy and abdominal CT are useful in detecting large varices. Angiography provides the necessary information regarding the location and extent of the varices. The varices are demonstrated in the venous phase of a superior mesenteric angiogram. Percutaneous transhepatic portal vein catheterization may be necessary for evaluation of portal hemodynamics and embolization. TIPS is effective in treating stomal or rectal varices in patients with cirrhosis and portal hypertension. [64]

Neoplasms of the Small Intestine

Leiomyomas Angiography is useful in diagnosing small bowel leiomyomas (Fig. 159-10). The tumors usually are hypervascular with abundant tumor vessels and dense blush. The draining vein usually is densely opacified. Angiographic differentiation between benign and malignant tumors is not possible unless venous invasion or metastasis is demonstrated. [65]

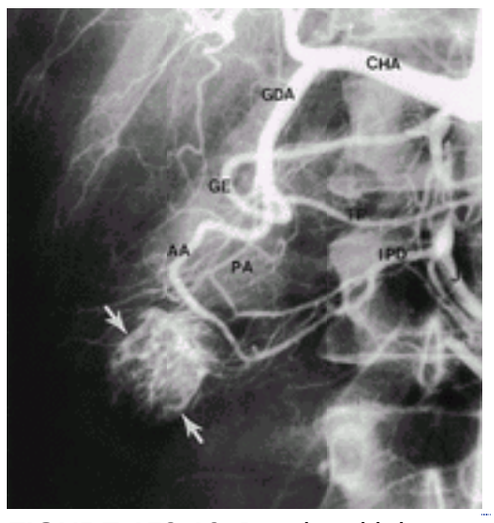

FIGURE 159-10. Duodenal leiomyoma. Late arterial phase of a gastroduodenal arteriogram in a young woman with upper gastrointestinal hemorrhage. A vascular mass (*arrows*) in the second portion of the duodenum receives blood supply from the dilated anterior arcade artery (*AA*). *CHA,* common hepatic artery; *GDA,* gastroduodenal artery; *GE,* gastroepiploic; *PA,* posterior arcade artery; *IPD,* inferior pancreaticoduodenal; *J,* jejunal; *TP,* transverse pancreatic. Surgical removal of the mass confirmed the diagnosis.

Carcinoid Tumors Angiography may be used for diagnosis and staging of carcinoid tumors. The angiographic findings include retraction, kinking, and occlusion of the mesenteric arteries. Carcinoid tumors metastatic to the liver usually are hypervascular with abundant tumor vessels. Venous sampling with hormone assay may be necessary for localizing occult carcinoid tumors, especially those occurring in the ovary and lungs. When the patient with liver involvement has severe carcinoid syndrome, hepatic artery embolization is an acceptable palliative treatment. [66]

Other Tumors Hemangiomas of the small bowel are an uncommon cause of GI bleeding. Angiography may be used to identify small hemangiomas. The angiographic findings include small areas of abnormal vessels with contrast accumulation in the capillary phase, with or without early draining veins. Angiography is not useful in diagnosing adenomas, lipomas, carcinomas, or Kaposi sarcoma.

PANCREATIC DISEASE

Dynamic contrast spiral CT is used to identify a pancreatic mass and assess the extent of the tumor. [67] Angiography is performed to obtain a specific diagnosis, to determine the extent of the tumor, and to demonstrate vascular anatomy. In patients with pancreatitis or pseudocysts, angiography is necessary when surgery is contemplated.

Adenocarcinoma

Conventional and helical CT may be performed to detect the extension of the tumor and involvement of the peripancreatic vessels. [68], [69] Angiography is useful in assessing the extent of the tumor. When any major peripancreatic artery (splenic, hepatic, or superior mesenteric artery) or vein (superior mesenteric, splenic, or portal vein) is involved, the tumor may be unresectable (Fig. 159-11).

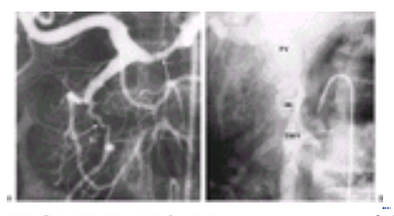

FIGURE 159-11. Unresectable pancreatic cancer. **A:** Hepatic arteriogram (magnification technique). The common hepatic (*a*) and proximal gastroepiploic (*b*) arteries are encased by a tumor in the head of the pancreas. **B:** Portal venous phase of a superior mesenteric angiogram (oblique view). The junction of the superior mesenteric and portal veins is invaded by the tumor (*arrow*). *PV,* portal vein; *SMV,* superior mesenteric vein. (From Shields JJ, Porter DJ, Brady TM, Cho KJ. Angiography of pancreatic disease. In: Dent TL, ed. Pancreatic disease. New York: Grune & Stratton, 1981:93.)

Cystic Tumors

US and abdominal CT can provide the diagnosis of a cystic pancreatic tumor; therefore, angiography is reserved for determining the extent of the tumor and vascular anatomy before surgical resection. Angiographically, microcystic adenomas usually are hypervascular with abundant tumor vessels and blush, whereas mucinous cystic tumors are hypovascular with sparse tumor vessels mimicking a pseudocyst.

Pancreatic Endocrine Tumors

Angiography is infrequently used for localization of islet cell adenomas. Angiographically, islet cell tumors appear as a localized area of contrast material accumulation (tumor blush) with or without tumor vessels (Fig. 159-12). Malignant islet cell tumors may grow into the portal vein. Most hepatic metastases from islet cell carcinomas are hypervascular and are readily detected by hepatic angiography. Angiography is sensitive in localizing insulinomas (accuracy, 60% to 90%), but it is less sensitive in detecting gastrin-producing tumors. Other endocrine tumors, such as vasoactive intestinal peptide–producing tumors (VIPoma), glucagonoma, and somatostatinoma, are usually hypervascular with abundant tumor vessels and can be demonstrated by CT.

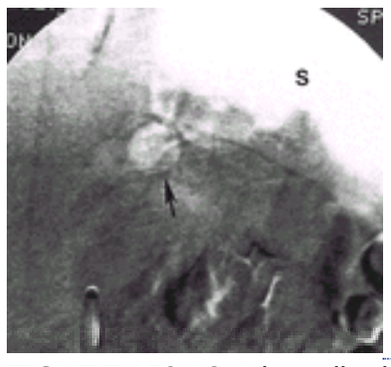

FIGURE 159-12. Islet cell adenoma in a patient with hyperinsulinism. Parenchymal phase of a splenic angiogram (digital subtraction technique) demonstrates a 1-cm diameter tumor blush (*arrow*) in the distal body of the pancreas. *S,* splenic parenchymal staining.

Percutaneous transhepatic venous sampling with hormone assay is a useful method for localizing occult islet cell tumors. This method may be used to localize the source of abnormal hormone secretion to the head, body, and tail of the pancreas as well as to the liver. This is helpful in avoiding blind resection of the pancreas. [70] Simultaneous arterial and venous sampling from the splenic, superior mesenteric, and portal veins can detect a localized elevation of hormone value near the tumor site; adenomas of the body and tail of the pancreas cause a step-up of hormone concentration in the splenic vein, and those of the pancreatic head cause step-ups in the superior mesenteric and portal veins. Simultaneous blood sampling from the hepatic and portal veins helps to determine the presence of hepatic metastases from gastrinomas. [71]

Selective intra-arterial injection of secretin with assay of hepatic venous gastrin has been useful in localizing gastrinomas and hepatic metastases. [72] It involves injection of 30 units of secretin into the splenic, gastroduodenal, proper hepatic, and superior mesenteric arteries as well as blood sampling from the hepatic vein to measure the changes of gastrin concentration. An increase (more than 50%) in gastrin concentration in the hepatic vein after secretin injection localizes the tumor to the part of the pancreas supplied by the artery injected: the gastroduodenal and superior mesenteric artery for the head of the pancreas and the splenic artery for the body or tail. The selective arterial secretin injection test is more sensitive than transhepatic portal venous sampling for localizing occult gastrinomas. [73] An increase in hepatic venous gastrin concentration of at least 25% at 20 seconds or 50% at 30 seconds after hepatic arterial injection of secretin indicates the presence of hepatic metastases. [74] In patients with suspected insulinomas, calcium gluconate is injected intra-arterially, and the level of insulin is measured in the hepatic vein for localization of insulinomas.

Pancreatic Arteriovenous Malformations

These rare vascular lesions may bleed directly into the intestine or into the pancreatic duct. Portal hypertension and varices associated with the malformation may be the cause of the bleeding. Angiography is important in the preoperative diagnosis and localization of the lesion. The lesion may mimic chronic pancreatitis or a vascular tumor.

Vascular Lesions Associated with Pancreatitis

Angiography has little diagnostic use in patients with pancreatitis because the diagnosis is well established clinically. When vascular complications occur, angiography is necessary for the diagnosis of vascular involvement and treatment. In about half of cases of pseudocyst, peripancreatic venous narrowing or occlusion is found. Bleeding associated with pancreatitis may be venous or arterial in origin. Venous bleeding usually results from gastric varices associated with splenic or portal vein occlusion. Venous bleeding rarely results from duodenal varices caused by superior mesenteric or portal vein occlusion. Arterial hemorrhage is a rare but life-threatening complication of pancreatitis, usually secondary to rupture of intrapancreatic or peripancreatic aneurysms. Arteriography is indicated for the diagnosis of arterial bleeding and pseudoaneurysm, and selective embolization should be used as a definitive or preoperative temporizing procedure. [75]

HEPATIC DISEASE

CT is the most useful screening method for hepatic lesions and can be used to distinguish a cystic from solid hepatic lesion. Bolus dynamic CT with CT-arterial portography is the most sensitive imaging modality for detecting hepatic metastases. [76] Delayed CT with the intra-arterial injection of iodized oil into the hepatic artery is useful in detecting small hepatocellular carcinomas. [77] Doppler US and color-flow Doppler imaging are complementary procedures and provide useful information on the vascularity and hemodynamics of liver tumors. Angiography still is important in evaluating liver tumors for the specific diagnosis, for resectability of the tumor, and for therapeutic hepatic artery embolization.

Knowledge of hepatic circulation is important in the performance and interpretation of hepatic angiograms and in planning for hepatic arterial chemotherapy. The liver receives a dual blood supply: about 75% of the total hepatic blood flow comes from the portal vein, and the remaining 25% comes from the hepatic artery. The relationship between the portal vein and hepatic artery is reciprocal: a decrease in portal blood flow results in immediate compensatory increase in the hepatic arterial flow. When portal blood flow is diminished in cirrhosis or obstructive jaundice, hepatic arterial embolization for bleeding or tumors must be undertaken cautiously because the liver in these conditions depends primarily on the arterial blood.

Cavernous Hemangiomas

Cavernous hemangiomas, the most common benign hepatic tumors, are composed of large sinusoidal spaces with fibrosis and thrombosis. CT, MR imaging, and ^{99m}Tc-scintigraphy with the use of single photon emission computed tomography are useful for obtaining a specific diagnosis. [78], [79] The angiographic findings are characteristic, with normal feeding arteries and dense persistent pooling of contrast material through the venous phase of a hepatic angiogram.

Adenoma

The findings of hepatic adenomas on CT, US, and MR imaging are nonspecific. Angiographically, hepatic adenomas usually are vascular, with abnormal vessels and tumor blush. The definitive diagnosis cannot be established on the basis of the angiographic abnormality. When an adenoma bleeds, angiography is necessary to evaluate the extent of bleeding and identify the tumor. If active contrast extravasation is identified, selective arterial embolization is an acceptable alternative treatment. When the patient becomes stable, resection should be performed. Adenomas associated with bleeding may become hypovascular or avascular, making their angiographic localization difficult.

Focal Nodular Hyperplasia

Angiographically, lesions are hypervascular; abundant abnormal vessels show granular contrast agent accumulation, and multiple septa radiate from the central area of scar.

Biliary Cystadenoma

Angiography is performed to define hepatic arterial anatomy and determine the extent of the tumor before surgery. The angiographic findings usually are nonspecific and may demonstrate subtle neovascularity and rimlike staining in the wall of the tumor, mimicking a hepatic abscess, an echinococcal cyst, or a congenital hepatic cyst.

Hepatocellular Carcinoma

Angiography is necessary to define the extent of the tumor, blood supply, and portal venous anatomy. Most hepatomas are hypervascular angiographically, with coarse tumor vessels (Fig. 159-13). Involvement of the extrahepatic portal vein by hepatoma indicates that the tumor is unresectable.

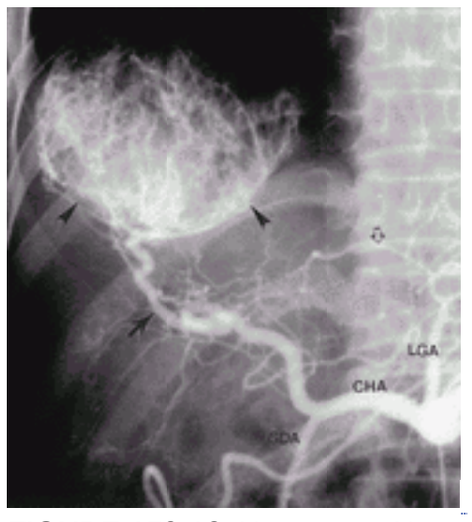

FIGURE 159-13. Hepatoma. Hepatic arteriogram shows a hypervascular mass (*arrowheads*) with tumor blush. Numerous tumor vessels supplied by a dilated right hepatic artery branch (*arrow*) are seen. The left hepatic artery (*open arrow*) arises from the left gastric artery (*LGA*). *CHA,* common hepatic artery; *GDA,* gastroduodenal artery.

Cholangiocarcinoma

Angiography has a limited role in the diagnosis but may be performed to provide a vascular road map and evaluation of portal vein patency when surgical resection is contemplated. The angiographic findings include encasement of the hepatic arterial branches, which may be associated with encasement of the portal vein.

Metastatic Hepatic Neoplasms

Hepatic metastases from renal cell carcinomas, islet cell carcinomas, carcinoid tumors, and medullary thyroid carcinomas are hypervascular, and other metastases are less vascular. Angiography is sensitive in detecting hypervascular metastases. Angiography is performed to determine resectability of hepatic metastases and the presence of the aberrant hepatic arteries before the surgical placement of a hepatic arterial catheter for chemotherapy.

PANHEPATIC ANGIOGRAPHY

Increased resistance to the flow of portal vein blood is the principal cause of portal hypertension. The location of the blockage in portal blood flow may be intrahepatic or extrahepatic. Cirrhosis is the most common cause of the intrahepatic sinusoidal block. Hepatic vein (postsinusoidal) and portal vein (presinusoidal) occlusion produce an extrahepatic block to portal vein flow. An increase in portal vein blood flow secondary to splenomegaly or arterioportal fistulas rarely causes portal hypertension (hyperkinetic).

The term *panhepatic angiography* is used for the angiographic study performed for the evaluation of portal hypertension. It includes celiac, superior mesenteric, hepatic, and splenic angiograms; wedged hepatic and left renal venograms; and manometry. Panhepatic angiography determines hepatic arterial variations, assesses portal hemodynamics, and excludes hepatoma. Visualization of aberrant hepatic arteries is important because accidental ligation of the hepatic artery in the presence of portal hypertension may result in hepatic necrosis. A right hepatic artery that originates from the superior mesenteric artery poses technical difficulty for portacaval shunt surgery.

The hepatic angiogram is necessary to exclude hepatomas and helps determine the hemodynamics of the intrahepatic portal flow. In advanced cirrhosis with reversed portal flow, the portal vein may be visualized in the venous phase of the hepatic arteriogram. High-dose superior mesenteric and splenic angiograms are necessary to see the superior mesenteric, splenic, and portal veins, as well as the portosystemic collaterals. In alcoholic cirrhosis, a pressure measurement from the catheter wedged in the hepatic vein reflects portal vein pressure and is useful in determining whether the location of the obstruction is intrahepatic or extrahepatic. However, wedged hepatic vein pressure is not a useful determinant for the selection of the type of shunt surgery. It is also a poor predictor of prognosis and survival after shunt surgery. Wedged hepatic venography is the most accurate means of assessing morphologic features of the hepatic sinusoids and parenchyma. Manometry of the right atrium, inferior vena cava, and left renal vein is important because the presence of significant elevation of caval pressure may result in incomplete decompression of portal hypertension after portacaval shunt surgery. Likewise, if the left renal vein pressure is elevated, the surgeon may decide to place another type of shunt rather than a distal splenorenal shunt.

TRANSCATHETER THERAPY

Diagnostic angiography should be performed before any trans- catheter intervention for GI disease. Interventional radiologic procedures are reviewed in detail in Chapter 160.

Percutaneous Translumenal Angioplasty and Stent Placement

Surgical therapy (endarterectomy and bypass surgery) for superior mesenteric artery obstruction has significant mortality and recurrence rates. Percutaneous translumenal angioplasty is a safe and effective alternative to surgery in treating intestinal angina resulting from arteriosclerotic occlusive disease of the visceral arteries. Stent placement is required for ostial stenosis or complete occlusion of the celiac and superior mesenteric arteries, failed percutaneous translumenal angioplasty because of elastic recoil, and mesenteric arterial compression by the false lumen of an aortic dissection. [80], [81], [82] and [83] Celiac axis stenosis caused by median arcuate ligament compression is not amenable to angioplasty and thus requires surgery.

Percutaneous translumenal angioplasty is effective in treating Budd-Chiari syndrome caused by a membrane in the inferior vena cava and a stenosis of the hepatic vein. [84], [85] The method may not be possible technically in patients with diffuse hepatic vein thrombosis and sclerosis. Percutaneous transhepatic angioplasty and stent placement has been used to treat portal vein stenosis and postoperative stricture of the portal vein complicating liver transplantation. [86], [87] and [88]

Transcatheter Embolization

Transcatheter embolization is an acceptable alternative to surgery in treating massive arterial bleeding, and aneurysms of the mesenteric, hepatic, and splenic arteries. [89], [90] and [91] Occlusion of the hepatic artery by embolization is safe and well tolerated because the hepatic sinusoidal perfusion can be maintained by collateral circulation and the portal venous blood. Selective hepatic arterial embolization is a safe and effective treatment for traumatic hepatic bleeding, hemobilia, and symptomatic arterioportal fistula. However, hepatic artery embolization in patients with symptomatic hepatic arteriovenous malformations associated with hereditary hemorrhagic telangiectasia is associated with a high risk for hepatic infarction and death. [92] The spleen receives adequate collateral blood flow through the pancreatic arteries after occlusion of the extrasplenic part of the splenic artery. Steel coil is the most frequently used occluding agent and produces arterial occlusion equivalent to surgical ligation. Injection of thrombin into an aneurysm promotes clot formation. A 3-French coaxial catheter system is extremely useful in catheterization and embolization of a mesenteric branch artery. If an aneurysm cannot be catheterized transarterially, a 22-gauge needle is used for percutaneous puncture, and embolization of the lesion is accomplished with microcoils and thrombin.

Thrombolytic Therapy

Thrombolytic therapy may be used for superior mesenteric artery embolism and portal vein thrombosis. A report of thrombolytic therapy for superior mesenteric artery

embolism indicates that the method is safe and effective, with technical success in 90% and clinical success in 70%, [93] but the treatment requires at least 6 to 8 hours of infusion. A percutaneous transhepatic or transjugular approach is necessary for thrombolysis of mesenteric and portal vein thrombosis. The technique involves placement of a catheter into the embolus or thrombus and infusion of a lytic agent. During thrombolysis, the patient should be monitored for bleeding and evidence of bowel infarction in the intensive care unit. Reteplase and alteplase are alternative fibrinolytic agents for catheter-directed thrombolysis of peripheral arterial occlusion. [94], [95] The transcatheter use of a mechanical thrombectomy device, such as AngioJet (Possis Medical Inc, Minneapolis, MN), has been shown to be effective in removing clots.

Hepatic Arterial Infusion Chemotherapy

Regional delivery of antineoplastic agents through a selectively placed arterial catheter produces higher tumor response than systemic infusion. [96], [97] The success of hepatic arterial chemotherapy depends on accurate evaluation of the hepatic arterial anatomy and hemodynamics, and correct catheter placement. To ensure total liver perfusion and drug delivery to the tumor site, the hepatic arterial anatomy is meticulously evaluated by celiac and superior mesenteric arteriograms. Multiple hepatic arteries can be converted to a single hepatic artery using the transcatheter embolization technique to facilitate catheter placement. For example, if an aberrant left hepatic from the left gastric artery is embolized, the left hepatic lobe receives blood supply through intrahepatic collaterals from the right hepatic artery. If the infusion catheter cannot be placed in the proper hepatic artery, the gastroduodenal artery may be occluded by embolization to allow infusion of drugs with a catheter placed in the common hepatic artery.

Hepatic arterial catheters can be placed through a percutaneous approach from the femoral or brachial artery, or surgically. The introduction of the totally implantable pump has improved regional chemotherapy with surgical catheter placement. [98] Radionuclide flow study using ^{99m}Tc macroaggregated serum albumin is the most accurate means of assessing hepatic perfusion pattern after catheter placement. The complications of percutaneous catheter placement are brachial artery thrombosis, hepatic artery thrombosis, catheter displacement, and puncture-site bleeding. The complications of surgical catheter placement are hepatic artery thrombosis, incorrect catheter placement, catheter occlusion, and pump malfunction. Complications related to the toxicity of chemotherapeutic agents are chemical hepatitis, biliary sclerosis, chemical cholecystitis, and gastroduodenal inflammation and ulceration.

Hepatic Tumor Embolization

Hepatic artery embolization is used to palliate hepatic tumors (hepatoma, metastatic neuroendocrine tumors, cholangiocarcinoma, and metastatic colon cancer) when the tumors are unresponsive to hepatic arterial chemotherapy. [99] The patients usually are not candidates for surgical resection. Before embolization, CT of the liver must be reviewed for evaluation of location of tumor, extent of disease, and any associated findings (portal vein occlusion, biliary obstruction). Adequate portal venous flow should be available to maintain hepatic parenchymal vitality. If the intrahepatic portal flow is reversed (hepatofugal), embolization should not be performed or limited to the subsegment containing tumor.

Hepatic arterial embolization results in selective destruction of the tumors exclusively dependent on arterial supply while maintaining the vitality of the normal liver with portal vein blood. Small particles, such as polyvinyl alcohol (Ivalon, 150–250 μm), are used to occlude peripheral arteries to decrease the development of collateral blood flow. Hepatic artery embolization can effectively control symptoms related to abnormal hormone production in most patients with carcinoid and islet cell tumors metastatic to the liver. [100] In patients with hepatocellular carcinoma with portal vein tumor invasion and arterioportal shunt, hepatic artery embolization may result in acute hepatic failure. Embolization of arterioportal shunts may be performed to reverse the portal blood flow from hepatofugal to hepatopetal in direction. Tumor embolization can then be performed with less risk for hepatic failure. [101]

Chemoembolization is the combination of intra-arterial infusion of chemotherapeutic agents and arterial embolization of the tumor vascular bed. [102] The rationale for this method is that concurrent embolization increases the effect of the drug by prolonging exposure of the tumor to the drug and producing tumor anoxia while minimizing systemic drug effects. The occluding materials currently in use include Gelfoam, Ivalon, and iodized oil. Mitomycin C, doxorubicin, cisplatin, and floxuridine are among drugs used for chemoembolization. Hepatic arterial chemoembolization, with Ivalon particles of 150 to 250 μm before and after selective injection of cisplatin, has been shown to be effective in treating hepatic metastases. [103] Chemoembolization material for hepatocellular carcinoma may consist of 10 mg mitomycin C, 50 mg doxorubicin, and 100 mg cisplatin dissolved in contrast medium and sterile water. This is mixed with Ethiodol and Ivalon particles. [104] This emulsion is injected into the hepatic artery until hepatic arterial flow is significantly reduced. Injection of iodized oil into the hepatic artery results in selective accumulation of the oil within most vascular hepatic tumors. This phenomenon has been exploited to deliver antineoplastic agents and radioactive isotopes selectively to hepatic cancers, with intra-arterial injection of a mixture of iodized oil and antineoplastic agents or radioactive iodized oil solution. [105], [106] Chemoembolization is effective in patients with hepatic metastases of neuroendocrine tumors. Intra-arterial injection of iodized oil and doxorubicin into tumor vessels is effective in palliating patients with hepatic islet cell or carcinoid tumor metastases. This method causes less morbidity than embolization of particulate material and permits repeat embolization because the supplying arteries remain patent after embolization. [107], [108] The chemoembolic mixture is composed of 40 to 60 mg of doxorubicin dissolved in 10 mL of contrast medium and 20 mL of iodized oil.

Hepatic artery embolization may be associated with the postembolization syndrome: abdominal pain, nausea, vomiting, and fever lasting 2 to 7 days. Mildly abnormal liver tests are common. Other complications include hepatic necrosis, gallbladder infarction, and gastroduodenal ulcers. Follow-up CT is obtained at 3-month intervals, and repeated embolizations are performed for recurrent tumors.

FORTHCOMING ADVANCES

Noninvasive imaging modalities (spiral CT, 3D contrast MR angiography, color Doppler imaging, and duplex US) will continue to play a primary role in the evaluation of suspected GI diseases. Angiography will be used less frequently but will continue to be used in the preoperative evaluation of vascular disease, portal hypertension, neoplasms of the pancreas and liver, and GI hemorrhage. Therapeutic applications of angiography will expand in the treatment of lower GI bleeding (small and large bowels), vascular abnormalities (arteriovenous fistula and aneurysm), hepatic neoplasms (chemoembolization for primary and metastatic malignancies), mesenteric vascular obstruction (acute and chronic), variceal bleeding unresponsive to endoscopic therapy, intractable ascites, and Budd-Chiari syndrome.

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CHAPTER 160

Kyung J. Cho and Grace H. Elta

INTERVENTIONAL RADIOLOGY

The field of interventional radiology has evolved since the introduction of the percutaneous transarterial technique devised by Seldinger in 1953. [1] Interventional radiologists perform a variety of diagnostic and interventional procedures of the circulatory system and nonvascular tracts under imaging guidance. The imaging guidance includes fluoroscopy, ultrasound (US), computed tomography (CT), magnetic resonance (MR) imaging, and conventional and digital subtraction angiography. The interventional radiologic procedures can be divided into vascular and nonvascular procedures. The vascular procedures include infusion, occlusion, dilation, stent placement, and foreign body retrieval. The nonvascular procedures include biopsy, drainage, dilation, and ostomy formation.

VASCULAR INTERVENTION

Control of Arterial Gastrointestinal Bleeding

Angiographic management is indicated for endoscopic failures, especially in poor operative candidates, and for inaccessible sites of bleeding, such as hemobilia. [2] Angiographic therapy for massive gastrointestinal (GI) bleeding includes intra-arterial infusion of vasopressin and selective arterial embolization. The success of angiographic treatment depends on accurate localization of the bleeding site and selective catheterization of the bleeding artery.

Vasopressin Infusion Vasopressin is effective for bleeding from Mallory-Weiss tears, hemorrhagic gastritis, and small bowel and colonic bleeding, [3], [4] but it is less effective for bleeding from a peptic ulcer because ulcers rupture larger arteries. Vasopressin infusion can control lower GI tract bleeding in 80% to 90% of patients, but 40% to 50% of these patients rebleed and eventually require surgery. Therefore, vasopressin infusion is infrequently used for control of lower GI bleeding. When the bleeding site has been demonstrated, a catheter is selectively placed in the bleeding artery. Vasopressin is infused into the left gastric artery for gastric bleeding, into the superior mesenteric artery for small bowel and right colonic bleeding, and into the inferior mesenteric artery for left colonic and rectosigmoid bleeding. Vasopressin infusion may be initiated at the rate of 0.2 U/minute. After 20 minutes of infusion, repeat arteriogram is performed to assess treatment effectiveness. If no bleeding is demonstrated and vasoconstriction is moderate, the infusion is continued for the next 24 hours at the same rate. When the bleeding has stopped, the rate should be reduced to 0.1 U/minute and continued for an additional 6 to 12 hours before termination of the infusion. If the bleeding is not controlled after vasopressin infusion of 0.4 U/minute for 20 minutes, increasing the rate is not recommended; embolization or surgery should be considered. Serious side effects from vasopressin infusion occur infrequently. Most side effects are dose dependent. They are coronary ischemia, arrhythmia, bradycardia, hypertension, bowel ischemia, peripheral vascular ischemia, and oliguria. In patients with a history of coronary artery disease, the infusion should be done with caution and concomitant administration of nitroglycerin.

Selective Arterial Embolization Selective arterial embolization is an effective method of therapy for control of massive upper GI hemorrhage when endoscopic therapy has failed. Selective occlusion of the bleeding artery with embolic agents such as gelatin sponge, polyvinyl alcohol (Ivalon), and microcoils results in prompt cessation of the bleeding, and it usually obviates the need for surgical intervention (Fig. 160-1). It is important to localize the bleeding site before embolization. Prophylactic embolization of the left gastric or gastroduodenal artery for upper GI bleeding can be performed, but endoscopic localization of the bleeding is necessary before embolization. Correction of coagulopathy before and after the embolization reduces the risk for bleeding recurrence. [5] The use of coils as the only embolic agent and concurrent coagulopathy in patients with upper GI bleeding are associated with a high risk for early recurrent hemorrhage. [6] The stomach and duodenum have sufficient collateral pathways; hence, infarction is unlikely to occur after embolization. On rare occasions, patients with previous surgery or those receiving vasopressin therapy after the embolization may develop infarction of the stomach. Duodenal ischemia, duodenal stenosis, and pancreatic necrosis are the potential complications of gastroduodenal artery embolization.

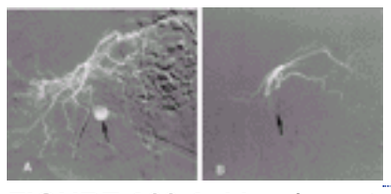

FIGURE 160-1. Massive arterial bleeding from a branch of the left gastric artery. **A:** Superselective left gastric arteriogram (digital subtraction technique). Active bleeding is seen from a branch of the left gastric artery in the body of the stomach. The extravasated contrast medium has formed a pseudoaneurysm (*arrow*). **B:** The bleeding has stopped following microcoil embolization (*arrow*).

Selective arterial embolization is a safe and effective method of therapy for control of lower GI bleeding. [7], [8] Gelfoam pledgets and Ivalon particles are the commonly used embolization agents. The use of a 3-French (3-Fr) coaxial catheter allows catheterization of the extravasating arteria recta for embolization with microcoils. [9] Rectal hemorrhage is usually diagnosed and treated by interventional endoscopy. When the rectum is filled with fresh blood, endoscopic identification of the bleeding site may be difficult. Regardless of the underlying etiology, embolization can be effective in controlling superior hemorrhoidal arterial hemorrhage. [10]

Variceal Embolization

In 1974, Lunderquist and Yang [11] reported the technique of percutaneous transhepatic coronary vein embolization as a means of controlling gastroesophageal variceal bleeding. Portal vein catheterization can also be done using the transjugular, umbilical, and intestinal veins after minilaparotomy. In the presence of massive ascites, coagulopathy, or liver tumors, the transjugular approach is used. Coronary vein embolization can effectively control variceal hemorrhage in 80% of cases, but recurrent bleeding is common because of the development of new gastric and esophageal varices. Coronary vein embolization is infrequently performed because endoscopic therapy is more effective in preventing recurrent hemorrhage. [12]

Rarely, bleeding may occur from the portosystemic collaterals at unusual sites, including the small bowel, colon, rectum, and enterostomy. [13], [14] If the bleeding is unresponsive to conservative therapy, variceal embolization can be performed through the transhepatic or transjugular approach [15], [16] (Fig. 160-2). Transjugular intrahepatic portosystemic shunt (TIPS) usually is preferable for the treatment of bleeding from varices at unusual sites owing to the risk for rebleeding with embolization alone. [17], [18] and [19]

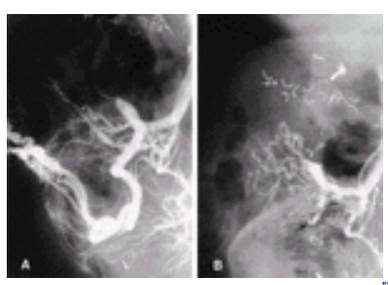

FIGURE 160-2. Transhepatic embolization of stomal varices. Percutaneous transhepatic mesenteric venogram in a 79-year-old woman with a 2-year history of recurrent bleeding from ileostomy varices. The bleeding began 1 year after total colectomy and ileostomy for colon carcinoma. **A:** Transhepatic catheterization of the ileal vein. Contrast medium fills the ileal vein, draining toward the stomal varices (*arrow*). **B:** Transhepatic mesenteric venogram after occlusion of the ileal vein with ethanol and coils. The ileal vein and collateral veins are occluded (*arrow*). The patient required three embolization procedures for recurrent bleeding.

Another reported unusual indication for variceal embolization is chronic recurrent hepatic encephalopathy. [20], [21] Embolization of portosystemic shunts can decrease diversion of portal blood flow through the shunts, resulting in an increase in portal blood flow. Portosystemic shunts amenable to such embolic therapy are splenorenal shunts, gastrorenal shunts, hepatic portal vein–hepatic vein shunts, and ileal vein–gonadal vein communications. Embolization can be done through the transfemoral or transhepatic approach, depending on the site of shunts. Balloon-occluded retrograde transvenous injection of a sclerosing agent is a safe and effective treatment of gastric varices and encephalopathy. [22] The technique involves the retrograde catheterization of the portosystemic collateral vein through the left renal and adrenal veins from the femoral vein. An occlusion balloon is wedged in the collateral vein, and a sclerosing agent is injected through the shunt into the gastric varices. If complete obliteration of the varices is not possible with the retrograde approach, the varices can be embolized by the transhepatic approach or the TIPS if present.

Budd-Chiari Syndrome

Percutaneous translumenal balloon angioplasty (PTA) is an effective therapy in patients with Budd-Chiari syndrome secondary to obstruction of the inferior vena cava (IVC) or hepatic vein. However, recurrence is common, and therefore, stent placement is usually performed. [23], [24] and [25] The transfemoral or transjugular approach is used for IVC obstruction and the percutaneous transhepatic approach for hepatic vein occlusion (Fig. 160-3). When the hepatic venous outflow obstruction extends into the peripheral hepatic veins, requiring portal decompression, TIPS is a viable alternative to surgical shunting and provides a bridge to liver transplantation. [26], [27] When portal, splenic, or mesenteric venous thrombosis is associated with Budd-Chiari syndrome, the thrombi can be dissolved by intrathrombic infusion of a fibrinolytic agent or removed by a mechanical thrombectomy device before placement of a TIPS. The TIPS stent may be extended into the splenic or mesenteric vein to achieve adequate inflow. [28]

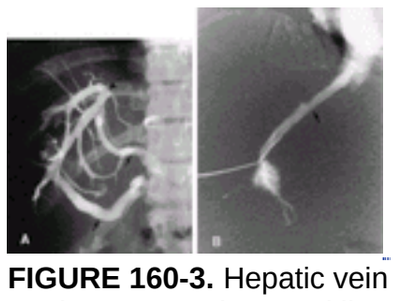

FIGURE 160-3. Hepatic vein stent placement for Budd-Chiari syndrome. Transhepatic hepatic venogram in a 23-year-old woman with right upper quadrant pain and tenderness, ascites, and liver failure. **A:** Transhepatic hepatic venogram. The hepatic vein is occluded (*arrowhead*). The contrast medium fills hepatic venous collaterals (*arrows*), draining toward the inferior vena cava. **B:** Transhepatic hepatic venogram after recanalization of the hepatic vein and placement of a 10-mm diameter Wallstent. The hepatic vein is patent with excellent blood flow (*arrow*). The venous collaterals are no longer filled. The hepatic vein pressure decreased from 29 mm Hg to 7 mm Hg after stent placement. One year later, the patient underwent orthotopic liver transplantation because of occlusion of the stent.

Portal Vein Stenosis

PTA and stent implantation using the percutaneous transhepatic and transjugular approaches are safe and effective in the therapy of portal vein stenosis, including cases complicating liver transplantation. [29], [30] and [31] Although most portal vein stenosis or occlusion is amenable to percutaneous techniques, their long-term efficacy is not known; therefore, follow-up with imaging studies is needed to detect restenosis of the treated lesion. [32]

Mesenteric Vein Thrombosis

Infusion of thrombolytic agents has been used for treatment of portal and mesenteric venous thrombosis. A thrombolytic agent may be administered by peripheral intravenous route or directly into the mesenteric vein through the transjugular [33] or transhepatic [34], [35] approach. Mechanical thrombectomy using a thrombectomy device or a Fogarty catheter can be used for acute portal vein thrombosis. This technique has been used for recanalization of acute portal vein thrombosis complicating TIPS. [36]

Portal vein reconstruction with recanalization and stent placement may be performed in patients with symptomatic chronic splenomesenteric-portal venous thrombosis. [37]

Visceral Arterial Stenosis

Atherosclerotic stenoses of the celiac and superior mesenteric arteries are a common cause of chronic mesenteric ischemia in elderly patients. Surgical revascularization is the standard treatment for clinically significant stenoses of the celiac and superior mesenteric arteries. PTA is a safe and effective alternative to surgical treatment. In a review of 83 reported patients who had PTA of visceral arteries, the initial technical success was 88%; the initial clinical success, 92%; the recurrence rate, 24%; primary long-term clinical success, 76%; successful repeat PTA, 92%; secondary long-term clinical success, 92%; and major complications, 6%. [38] Stent placement has been shown to be safe and clinically effective as an adjunctive treatment to PTA or as a primary therapy in patients with chronic mesenteric ischemia caused by calcified ostial stenoses, high-grade eccentric stenoses, chronic occlusions, and flow-limiting dissection of mesenteric arteries. [39]

PTA and stent placement are usually done through the femoral approach. If the femoral artery cannot be used, the left axillary approach is used. Before the procedure, lateral aortography and selective mesenteric arteriography are performed to evaluate the arterial lesions and visceral arterial circulation. The origin of the artery to be dilated is then catheterized, and the stenosis is then crossed using either a soft-tipped guidewire or a hydrophilic guidewire. The patient is given an intravenous bolus of 3000 to 5000 units of heparin. Over a 0.035-inch (0.89-mm) heavy-duty guidewire, a PTA balloon catheter of appropriate balloon size is placed across the stenosis and dilated to pressures of 5 to 7 atm (Fig. 160-4). If a gradient is greater than 10 mm Hg after balloon dilation or if a hemodynamically significant dissection is seen, a metallic stent is placed. Systemic anticoagulation is not usually necessary after the procedure. PTA should have a limited role in the patient with acute mesentericischemia.

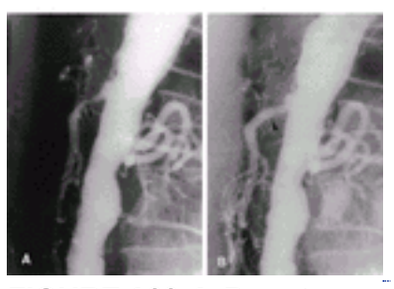

FIGURE 160-4. Percutaneous translumenal angioplasty for chronic mesenteric ischemia caused by mesenteric artery stenoses in a 78-year-old woman. **A:** Lateral aortogram shows occlusion of the celiac and inferior mesenteric arteries and high-grade stenosis of the superior mesenteric artery (*arrowhead*). **B:** After angioplasty, the stenosis (*arrowhead*) has been markedly decreased. Her symptoms markedly improved. She remained asymptomatic for 2 years after angioplasty.

Superior Mesenteric Artery Embolism

Embolism of the superior mesenteric artery is an important cause of acute intestinal ischemia with high mortality rates. Survival is often directly related to early diagnosis by emergency mesenteric arteriography. If an embolus is found in the proximal superior mesenteric artery, surgical treatment usually is indicated. In selected patients with an early diagnosis of peripheral embolism of the mesenteric arteries, intra-arterial thrombolysis may be a therapeutic alternative. [40], [41]

Splanchnic Artery Aneurysm

Selective arterial embolization is safe and effective in treating visceral arterial aneurysms in poor surgical risk patients, particularly those with pancreatitis-associated aneurysms and pseudoaneurysms. [42], [43] The embolization is usually done using the transfemoral approach. Celiac and superior mesenteric arteriograms are performed to evaluate the lesion and arterial anatomy. A 5-Fr catheter is advanced into the feeding artery as close to the lesion as possible. A 3-Fr coaxial catheter system is usually required to place the tip of the catheter close to or in the aneurysm. Coils are the most commonly used embolic agent. Thrombin may be added to the coils to promote clot formation.

If transarterial access to the aneurysm cannot be obtained owing to unfavorable arterial anatomy, the aneurysm is punctured percutaneously using a 22-gauge, skinny needle under ultrasonic or angiographic guidance. After confirmation of the position of the needle in the lesion, microcoils are deposited through the needle using a 0.018-inch guidewire, or thrombin is injected into the aneurysm. [44], [45]

Vascular lesions are uncommon complications of laparoscopic cholecystectomy. Most bleeding complications that occur intraoperatively are treated during the procedure. Any delayed vascular complications, such as hepatic artery pseudo- aneurysm and arteriobiliary fistula, [46] can be treated by selective embolization.

TRANSJUGULAR INTRAHEPATIC PORTOSYSTEMIC SHUNT

TIPS is invaluable and has largely replaced surgical shunts in patients with variceal bleeding who fail medical and endoscopic treatment. [47]

TIPS is a nonsurgical method of achieving portal decompression in patients with portal hypertension and variceal hemorrhage. The indications for TIPS are variceal bleeding unresponsive to endoscopic therapy, recurrent variceal bleeding despite endoscopic therapy, portal hypertensive gastropathy, refractory ascites, hepatic hydrothorax, Budd-Chiari syndrome, stomal variceal hemorrhage, and portal venous thrombosis. [48], [49], [50], [51], [52] and [53] TIPS may also be a treatment option for liver transplant recipients with refractory variceal bleeding and intractable ascites. [54] There are no absolute contraindications for creating TIPS. The relative contraindications include elevated right heart pressure, heart failure, fulminant hepatic failure, severe hepatic encephalopathy, uncontrolled systemic infection or bacteremia, unrelieved biliary obstruction, multiple hepatic cysts, extensive primary or metastatic hepatic malignancy, and severe uncorrectable coagulopathy.

Technique

The transjugular approach to the portal venous system and creation of a portosystemic shunt between the hepatic vein and portal vein was developed by Rösch in animals [55] and in humans by Colapinto. [56] Before a TIPS procedure, patency of the portal vein should be demonstrated by imaging methods. US is the most commonly used method for evaluation of portal vein patency. The other methods include CT arterial portography, three-dimensional contrast MR venography, indirect portography (arterial portography), and carbon dioxide (CO_2) wedged hepatic venography.

Preprocedure fresh-frozen plasma or platelet transfusion is given if needed. The procedure can be performed under conscious sedation or a general anesthesia for uncooperative patients. The patient lies on his or her back, and the neck is slightly extended backward and turned to the left during the procedure. The right internal jugular vein is catheterized using a 21-gauge, single-wall puncture needle under real-time US guidance. A 10-Fr vascular sheath is advanced into the right atrium. A 5-Fr catheter is then introduced coaxially, and pressures are measured of the right atrium, IVC, and free and wedged hepatic veins. CO_2 digital subtraction wedged

hepatic venography is performed to see the portal vein.

A 16-gauge Colapinto needle (Fig. 160-5) is advanced over a 0.035-inch Amplatz, superstiff guidewire into the right hepatic vein. The guidewire is removed, and the needle is advanced with fluoroscopic guidance into the expected position of the right main portal vein 3 cm from the portal vein bifurcation. The needle is withdrawn until blood can be aspirated, and contrast medium is injected to see the vascular structure entered.

TABLE cellSpacing=0 cellPadding=0 align=left border=0 hspace="10" vspace="5">

FIGURE 160-5. Colapinto needle used in transjugular intrahepatic portosystemic shunt (TIPS) procedure.

When the portal vein is entered, a guidewire is introduced into the superior mesenteric or splenic vein. The 9-Fr catheter is advanced over the Colapinto needle and guidewire into the portal vein. The needle is removed, and a 5-Fr catheter is advanced into the portal vein. Pressure of the portal vein is measured, and a portal venogram is performed (Fig. 160-6) to see the portal venous system and portosystemic collaterals. The parenchymal tract is dilated with an 8-mm PTA balloon catheter, and a 10-mm diameter metallic stent (see Fig. 160-6) is placed in the tract bridging the distance from the portal to the hepatic vein. Sometimes, more than one stent must be used to cover the tract. The stents are then dilated with a 10-mm PTA balloon to ensure their maximal expansion. Portal and systemic venous pressures are measured, and another portal venogram is obtained (see Fig. 160-6). The residual pressure gradient, the angiographically determined flow into portosystemic collaterals, such as gastroesophageal varices, gastrorenal or splenorenal shunts, and the patient's bleeding status, determine whether embolization is required.

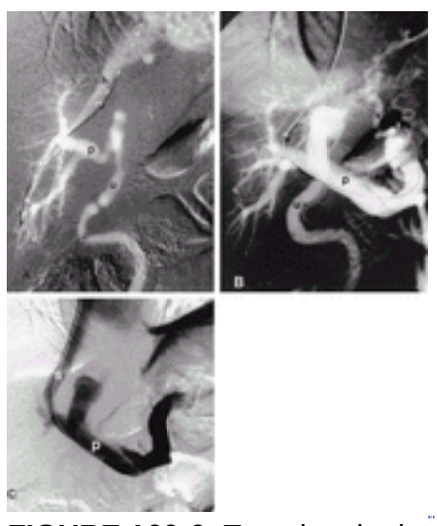
FIGURE 160-6. Transjugular intrahepatic portosystemic shunt (TIPS) in a patient with portal hypertension and variceal bleeding unresponsive to endoscopic therapy. **A:** Carbon dioxide wedged hepatic venogram shows hepatic (*h*), portal (*p*), and recanalized umbilical (*u*) veins. **B:** Transjugular catheterization of the portal vein. The transjugular portal vein catheter has been introduced into the splenic vein from the right hepatic vein. Contrast medium fills the portal (*p*) and umbilical (*u*) veins and esophageal varices (*v*). The portal venous blood flows toward the liver (hepatopetal). **C:** After creation of a 10-mm diameter TIPS shunt, the portal blood (*p*) flows through the shunt (*s*) directly into the right atrium. The intrahepatic portal and umbilical veins are no longer seen, indicating that the blood flows toward the shunt. The bleeding has stopped.

A portosystemic gradient of less than 12 mm Hg after creation of a TIPS is deemed satisfactory. The hemodynamic alterations caused by a TIPS are similar to those caused by H-mesocaval and side-to-side portacaval shunts, but there appears to be slightly better preservation of hepatic portal perfusion following the TIPS. The flow through the shunt can be controlled by modifying the size of the shunt. Portal vein blood flow is usually reversed after creation of a TIPS, and when injected into the portal vein, contrast medium will flow directly toward the shunt into the IVC. The intrahepatic portal venous branches will not fill. No benefit in terms of shunt patency and survival has been demonstrated by using a 12-mm stent. [57] The radial force of the 10-mm stent is greater than that of the 12-mm stent.

Intrahepatic portosystemic shunts can be performed using transmesenteric-transfemoral [58] and transfemoral [59] approaches. The transmesenteric-transfemoral technique involves catheterization of the portal vein through a mesenteric vein through a minilaparotomy [60] and percutaneous transfemoral access to the hepatic vein. The procedure is usually performed in the angiography suite. Through a midline incision, a mesenteric vein branch is cannulated by a 19-gauge needle, a 0.035-inch guidewire is advanced into the portal vein, and an 8-Fr vascular sheath is introduced. A curved blunt direction needle is inserted from the mesenteric side, and a 20-gauge needle is advanced from the portal vein to the hepatic vein in which a radiopaque basket is placed for a target. Once the needle is within the basket, a 0.018-inch, 260-cm long nitinol guidewire is advanced through the needle and retrieved through the femoral sheath. Over the guidewire, a 7-Fr, 100-cm long van Andel catheter is introduced from the femoral side. The liver tract is dilated with an angioplasty balloon catheter, and a 10-mm diameter metallic stent is deployed from the portal vein to the hepatic vein from the mesenteric site to create a shunt.

The transfemoral approach is used in patients with severely stenotic, angulated, or thrombotic hepatic veins, which make the standard jugular approach impossible. The procedure involves the placement of loop snares in the right portal vein and hepatic segment of the IVC and puncture of the IVC from the portal vein side using a sheathed needle. A guidewire is then advanced into the IVC and is snared and withdrawn through the transjugular sheath. A sheathed, 16-gauge Colapinto needle is advanced over the guidewire into the right portal vein. The parenchymal tract is dilated, and a stent is deployed from the main portal vein to the IVC.

TIPS Surveillance

Doppler US is an effective noninvasive screening tool for assessing TIPS patency and detecting increased portosystemic gradient. US examination after placement of a TIPS is obtained before discharge from the hospital and 3, 6, 9, and 12 months after the shunt. If US indicates shunt stenosis or occlusion, portal venography and pressure measurements are performed through the right internal jugular vein. Midshunt velocities of less than 60 cm/second, main portal vein velocities of less than 40 cm/second, or interval change from hepatofugal to hepatopetal intrahepatic portal flow at US examination usually indicates shunt stenosis. [61], [62] Because US screening of TIPS is not 100% sensitive or specific, angiographic evaluation may be required if clinical suspicion for stenosis is high. Helical CT angiography has been used to identify both normal and abnormal TIPS and to image focal or diffuse stenoses of the stents and hepatic vein. [63]

Therapeutic procedures can be performed easily on a failing TIPS in the outpatient setting. Patients whose shunts are occluded at US should undergo revision of their shunts. The shunt is catheterized from the jugular vein, and pressures are measured at the right atrium, IVC, hepatic vein, shunt, and portal vein. A portosystemic gradient of greater than 15 mm Hg usually indicates a malfunctioning shunt, and a portal venogram will disclose a stenosis anywhere from the portal vein to the hepatic vein. An angioplasty balloon catheter with the same size of the shunt can be used to dilate a narrowed shunt. If this is unsuccessful, a metallic stent is placed across the lesion.

Complications

The risks of TIPS vary greatly with the patient's bleeding status at the time of shunt creation. Data suggest that the 30-day survival of patients undergoing TIPS procedure is about 95%. In patients who are actively bleeding and who undergo TIPS, however, a 55% mortality rate has been seen. [64] Major complications include hemoperitoneum, gallbladder puncture, stent malposition, hemobilia, hepatic infarction, renal failure requiring dialysis, hepatic artery injury, accelerated liver failure, severe encephalopathy, cardiac perforation, intraperitoneal hemorrhage, and death. [65], [66], [67] and [68] The reported incidence of new or worsened encephalopathy after

TIPS is 20%. [49], [50] In almost all cases, the encephalopathy occurring after TIPS creation can be controlled medically. A balloon catheter can be used to occlude the shunt temporarily in a patient who develops refractory hepatic encephalopathy. Balloon occlusion of the shunt for more than 12 hours produces shunt thrombosis and improves encephalopathy [69] (Fig. 160-7). If the patient develops variceal bleeding after TIPS occlusion using an occlusion balloon catheter, shunt recanalization can be performed. The flow through the shunt can also be reduced by placing a Wallstent with constriction. [70] Minor complications include transient contrast-induced renal failure, encephalopathy controlled by medial therapy, transient pulmonary edema, and puncture site hematoma.

FIGURE 160-7. Temporary balloon occlusion of transjugular intrahepatic portosystemic shunt (TIPS) for refractory hepatic encephalopathy complicating TIPS placement. Balloon occlusion of the shunt (*arrow*) for 24 hours improved hepatic portal perfusion and encephalopathy. The portal venogram shows hepatopetal blood flow in the right portal vein (*open arrow*). Subsequently, a smaller shunt (7-mm diameter) was placed. One month later, the patient underwent successful hepatic transplantation.

Outcome and Prognosis

Patients with TIPS require intensive follow-up for at least 2 years after shunt placement, with shunt revision frequently being required. Primary shunt patency at 1 year after placement is only 25% to 60% [48], [71], [72]; however, with careful monitoring and treatment of shunt stenoses with balloon dilation, additional stent placement, or both, a primary-assisted patency rate of 83% to 96% can be achieved 1 year after placement. Some patients require more than one reintervention.

Acute and subacute TIPS thrombosis occurs in up to 10% of cases. Rarely, TIPS thrombosis may extend into the portal, splenic, and mesenteric veins. Percutaneous management in such cases includes thrombolysis, balloon embolectomy, suction embolectomy, and basket extraction of clot. Long-term results of each of the treatments are unknown, but a combination of methods may be required to restore mesenteric blood flow through the TIPS. [73]

Inadvertent traversal of bile ducts during creation of the intrahepatic tract results in biliary-to-TIPS fistulae that result in intimal hyperplasia and eventual shunt occlusion. Testing the parenchymal tract with injections of contrast material is useful in detecting inadvertent traversal of the bile duct before stent placement. If stent occlusion is associated with biliary fistula, stent grafts covered with polytetrafluoroethylene can be used to isolate the shunt from the fistula and to help prevent reocclusion. [74], [75]

The long-term patency rate of TIPS remains to be determined. Unless the stenosis rate of TIPS can be reduced, either with covered stents or pharmacological therapy, TIPS will have to be thought of as a temporary treatment of portal hypertension. Even as a temporary treatment, TIPS can effectively control variceal bleeding and stabilize patients who will undergo liver transplantation. As a method to control bleeding in Child's class C patients when other therapy has failed, TIPS is useful. In these patients, who have short life expectancies, a durable procedure is not required.

Operative shunts are clearly more durable than TIPS; thus, it must be determined whether TIPS with frequent follow-up and reintervention or surgical shunts result in longer survival or better quality of life in patients with longer life expectancies who, for medical or social reasons, are not transplantation candidates. Likewise, the role of endoscopic therapy and TIPS in elective treatment of variceal bleeding remains to be determined. TIPS is more effective in preventing recurrent bleeding than endoscopic therapy but produces encephalopathy. Both procedures require repetitive reintervention, and survival rates are similar.

BILIARY INTERVENTION

Percutaneous Transhepatic Cholangiography

Diagnostic information about the biliary tree, bile leakage, or intraductal lesions should first be obtained by noninvasive tests, CT, US, MR imaging, biliary scintigraphy or endoscopic US. When further diagnostic information or the opportunity for therapy is required, the invasive procedures, endoscopic retrograde cholangiopancreatography (ERCP) or percutaneous transhepatic cholangiography (PTC), are indicated. The choice between ERCP and PTC depends on local expertise, indication for the procedure, and the patient's anatomy.

The indications for PTC include (1) differentiation of obstructive from nonobstructive jaundice, (2) identification of the cause of ductal obstruction, (3) delineation of congenital anomalies, (4) determination of the site of biliary leakage, (5) documentation of communication of hepatic abscess with the biliary tree, and (6) visualization of ductal anatomy before percutaneous transhepatic biliary drainage (PTBD).

Contraindications include (1) uncorrectable coagulopathy, (2) history of severe reaction to iodinated contrast medium, (3) uncooperative patient (may require general anesthesia), (4) massive ascites, and (5) the presence of a vascular tumor or arteriovenous malformations. Risks include bleeding, sepsis, bile leakage, and death, with a 3% morbidity rate and a 0.1% mortality. [76]

Technique Coagulation studies are obtained on all patients, and significant abnormalities are corrected by the administration of fresh-frozen plasma, platelets, or both. Because of the high incidence of bacterial colonization of the biliary tree in patients with biliary obstruction, [77] broad-spectrum intravenous antibiotics are administered before and for 48 hours after the procedure. The imaging studies should be reviewed for the presence of intrahepatic masses and the distribution of intrahepatic biliary dilation. The puncture site is chosen in the midaxillary line midway between the costophrenic angle and the lower margin of the liver. Selection of a puncture site caudal to the 10th rib usually helps avoid the pleural space. After the puncture site is selected, the area is washed with povidone-iodine (Betadine) and isolated with drapes. The skin and planned puncture tract are anesthetized with 2% lidocaine. The skin nick is made in the caudal portion of an intercostal space to avoid the intercostal vessels and nerves. Under fluoroscopic control, a 22-gauge, skinny needle is directed, in a plane parallel to the table top, to a position 3 cm cephalad to the hilum of the liver. The position of the hilum is estimated fluoroscopically by observing the position of the dome of the liver and the air in the first and second portions of the duodenum. The stylet is removed, and the needle is connected through a connecting tube to a contrast-filled syringe. The needle is slowly withdrawn while small amounts of nonionic contrast medium are injected. When the needle tip is in the hepatic parenchyma, the contrast agent remains adjacent to the needle tip or opacifies the hepatic sinusoids. When the tip is in a hepatic artery or portal vein, the contrast agent washes out promptly to the liver periphery. When the tip is in a hepatic vein, the contrast agent washes out promptly toward the IVC. When the tip is in a biliary duct, the contrast material flows away slowly to reveal branching ducts as well as the central bile ducts (Fig. 160-8). When the tip is in a lymphatic duct, the contrast material enhances small channels that run toward the liver hilum.

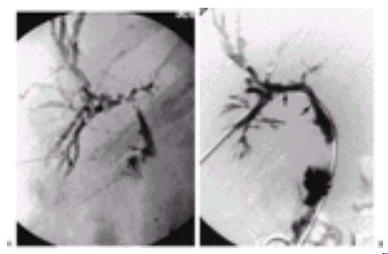

FIGURE 160-8. Percutaneous transhepatic cholangiography and cholangioplasty in a 46-year-old woman who 9 years earlier had undergone orthotopic liver transplantation for primary sclerosing cholangitis. **A:** Contrast medium was injected into a peripheral branch of the right hepatic duct through a 22-gauge needle (*arrowhead*) showing multiple intrahepatic and hilar biliary strictures. Contrast medium flows through the choledochoenterostomy (*arrow*) into the bowel. **B:** Cholangioplasty with a 6-mm diameter balloon catheter improved biliary strictures (*arrows*). The left hepatic duct has been occluded.

If no bile ducts have been encountered by the time the needle has been withdrawn to within 2 to 3 cm of the liver edge, the stylet is reinserted and the needle readvanced to a slightly different position. With dilated ducts, the biliary tree can be punctured almost always within several needle passes. [78] When the intrahepatic ducts are not dilated, additional needle passes frequently are required. For optimal visualization of the left hepatic ducts, or if a left-sided drainage is anticipated, a skin puncture site is chosen below the xiphoid process with the help of real-time US. A peripheral location in a duct of the lateral segment of the left lobe is chosen to be the puncture site so that if drainage is desired, the catheter will have a long segment with multiple side holes within the hepatic duct above the obstruction. The needle is angled parallel to the duct in the axial plane toward the hilum of the liver and at a 30- to 50-degree angle to the table top. The needle is advanced under real-time US guidance, and its position is confirmed with injection of contrast medium. If the site of obstruction has previously been determined by cross-sectional imaging and biliary drainage is to be performed, only a small amount of contrast agent is injected to assist guidewire placement. If a diagnostic study is to be performed, first an attempt is made to aspirate as much bile as possible, although typically, little or no bile can be aspirated through the fine needle because of its viscosity. Any bile obtained is sent for a Gram stain and culture and sensitivity tests. Iodinated contrast medium is slowly injected, and the contrast medium can be seen to flow into the most dependent ducts. Distention of the biliary tree is minimized to avoid causing bacteremia. By tilting the table, the radiologist takes advantage of the higher specific gravity of contrast material, allowing it to flow to opacify ducts not seen with the patient in the supine position. Radiographs are obtained in multiple obliquities, and the needle is removed. The patient is placed on bed rest for 6 hours, and vital signs are monitored. Fine-needle PTC is successful in 98% of patients with dilated ducts and in 70% of patients with nondilated ducts. [76] Major complications of PTC are sepsis, cholangitis, bile leak, hemorrhage, or pneumothorax, occurring in 2% of cases. [77]

Percutaneous Transhepatic Biliary Drainage

Nonoperative biliary drainage can be performed percutaneously or endoscopically in patients with biliary obstruction. Endoscopic techniques should be used when possible because the morbidity of endoscopic approach is lower than that of PTBD. ERCP has a higher failure rate with biliary obstruction at or above the hilar level, making PTBD frequently necessary in these patients (see Fig. 160-8). In technically difficult situations in which endoscopic access has failed, a guidewire can be placed into the duodenum transhepatically to facilitate repeat ERCP and intervention. These “combined procedures” are associated with increased complications but have high success rates.

The indications for PTBD include (1) palliation of obstruction of the biliary tree by tumor, (2) treatment of cholangitis, (3) nonoperative removal of common bile duct or intrahepatic stones when ERCP is unsuccessful, (4) access for dilation of benign biliary strictures, and (5) diversion of bile for treatment of biliary leaks or fistulae.

An uncorrectable coagulopathy is a contraindication for PTBD. Prominent ascites is a relative contraindication because the liver is displaced from the abdominal wall. This displacement can cause buckling of guidewires and catheters between the abdominal wall and liver, making the procedure difficult. Also, in patients with ascites, a risk exists of creating a peritoneal-cutaneous fistula with chronic leakage of ascitic fluid. Paracentesis can be performed before PTBD to minimize these problems.

The complications of PTBD include bleeding, sepsis, cholangitis, bile leakage, hemorrhage, pancreatitis, peritonitis, and death. Immediate significant complications are seen in 5% to 10% of patients. [78], [79] Patients who have chemotherapy-induced biliary sclerosis and obstructive jaundice have a higher incidence of bleeding complications with development of central pseudo- aneurysm following PTBD. [80] After access to the bile duct has been made, the tract may be injected with contrast medium to exclude communication with large vascular structures. If vascular communication is seen, the access should be abandoned, and a new puncture of a peripheral bile duct is made. This technique reduces the incidence of hemobilia. [81] The overall mortality rate reported with PTBD is 1.7%.

Technique The puncture of a bile duct is the same as that for PTC. After a small injection of contrast medium confirms the needle position in a bile duct, a 0.018-inch (0.46-mm) wire is passed through the needle into the bile duct. Over the guidewire, a 6.3-Fr introducing catheter with a stiffening cannula is advanced into the common bile duct. The stiffening cannula and the wire are removed, and bile is aspirated. After the biliary tree has been decompressed, a cholangiogram is performed with injection of contrast medium in the bile duct near the obstruction. The introducer is then exchanged for a 50-cm catheter with an angled tip. The catheter is then advanced to the point of obstruction, and a hydrophilic guidewire is manipulated through the obstruction into the duodenum. The catheter is advanced into the duodenum over the guidewire. The parenchymal tract is then dilated using sequential dilators over a 0.035-inch (0.89-mm) stiff guidewire, and an 8.5-Fr (outer diameter of 2.8 mm), internal-external drainage catheter is advanced into the duodenum, its distal retention loop is formed, and the drawstring is locked at the catheter hub to prevent catheter displacement. The drainage catheter is connected to a drainage bag to allow maximal drainage, which reduces the risk for sepsis. On the second day, the catheter is capped to allow internal drainage only. Once the internal drainage has been established, placement of a biliary endoprosthesis, if desired, is performed. If the guidewire does not traverse the occlusion, an external drainage catheter is placed above the obstruction, and the catheter is allowed to drain externally for 2 to 3 days. After a period of decompression, the obstruction usually can be crossed, and an internal-external drain can be placed. If the drainage is being performed for the treatment of biliary sepsis, no attempt is made to traverse the obstruction. Rather, an external drainage catheter is placed above the obstruction, and a more definitive procedure is performed in several days. Catheter and guidewire manipulation in purulent bile should be minimized because of the profound bacteremia that can ensue. The success rate of PTBD is high, and most malignant biliary obstructions can be crossed. In patients with malignant biliary obstruction, bilirubin levels fall quickly (2 mg/day) after decompression, and patients drained for sepsis defervesce within 24 hours. [82]

Percutaneous Transhepatic Cholangioplasty

An angioplasty balloon catheter can be used to dilate benign biliary strictures through endoscopy, a transhepatic access (see Fig. 160-8), a mature T-tube tract, or a percutaneous transjejunal route. [83] Endoscopic dilation is generally preferred by the patient owing to the lack of an external appliance and has long-term success rates of 70% to 85%, although it requires two to five ERCP sessions. [84], [85] The usefulness of temporary stent placement after cholangioplasty is debated for both the endoscopic and percutaneous routes, and even among those who favor using stents, the stent size and appropriate duration of having the stents in place is controversial. Long-term patency rates of percutaneous cholangioplasty are 67% to 93% for anastomotic strictures, [86], [87] and [88] 76% to 88% for iatrogenic strictures, [86], [87] and 42% to 54% for strictures of primary sclerosing cholangitis. [86], [89] Balloon dilation works well, is less expensive than placing stents, and will not interfere with a future operation, as might a metallic stent. Maccioni and associates [90] reported a 69% 3-year patency rate in a series of patients with anastomotic stenoses and iatrogenic bile duct injuries.

Transhepatic balloon dilation is a safe and effective treatment for biliary stricture in liver transplant recipients. [91] The dilation procedure usually requires multiple sessions (one to six sessions; mean, three). Balloon sizes to be used depend on the estimated size of the duct and are usually 25% to 30% larger than the duct. Balloon size is increased by 1 to 2 mm at each session, and the balloon is inflated two to three times to a pressure of 10 to 12 atm for 10 to 20 minutes. After dilation, an 8.5- to 14-Fr drainage catheter is placed.

Percutaneous Transhepatic Biliary Stent Placement

Internal stents (endoprostheses) can be placed at ERCP or PTC to provide biliary drainage without an external catheter and thus improve quality of life. The problem with internal stents is limited patency, which ranges from 4 months to 1 year. [92], [93], [94], [95] and [96] Endoscopically placed metallic biliary stents have longer patency than plastic stents owing to larger diameters but are not indicated in benign disease or in patients with operable tumors because they are permanent. [95] Endoscopic placement of internal stents is preferable to the percutaneous route because it can be achieved in a single session without the need for temporary percutaneous drainage. Percutaneous endoprosthesis placement is usually reserved for patients who have failed ERCPs or who already have percutaneous access. [97]

Hilar biliary stricture formation is a common complication of orthotopic liver transplantation, occurring in 10% of transplantations. [98] Intrahepatic strictures unresponsive to balloon dilation have been treated with metallic stents with relatively poor long-term patency. Culp and colleagues [99] reported on placement of 61 metallic stents in 36 liver transplant recipients with biliary stricture. Their primary patency rate was 44% at 3 years, with a secondary patency rate of 88%. Major stent-related complications occurred in 26% of their patients. Obstructed stents often require percutaneous drainage and additional stent placement to maintain patency. [100] Therefore, metallic stents play a limited role in the treatment of biliary stricture in the liver transplant recipient. Metallic stents are not generally used for treatment of other benign biliary strictures. [101] In patients with malignant biliary strictures, a significant decrease in bilirubin level occurs in 83% following metallic stent implantation, and 30% of patients develop recurrent jaundice after an average of 97 days. [102]

A variety of plastic and metal stents are in clinical use. Plastic stents require 12- to 14-Fr sheaths for placement, whereas metal stents can be placed through 7-Fr sheaths (Fig. 160-9, Fig. 160-10 and Fig. 160-11). Stent placement is usually done through the existing percutaneous transhepatic tract (Fig. 160-12) or endoscopically. The most common size of metallic stents is 10 mm in diameter and 4 to 8 cm in length, longer than the 10- to 14-Fr plastic stents. After the stent has been placed, an external drainage catheter is left above the stent for 48 hours and withdrawn after demonstration of stent patency.

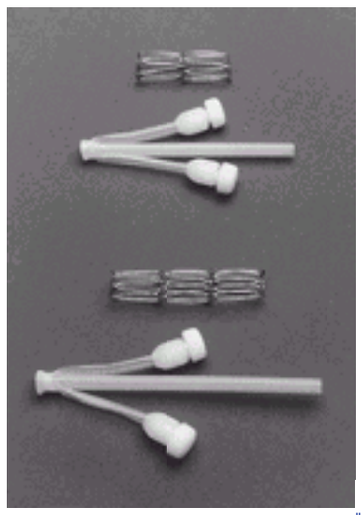

FIGURE 160-9. Two- and three-body Gianturco-Rösch stents and their loading devices.

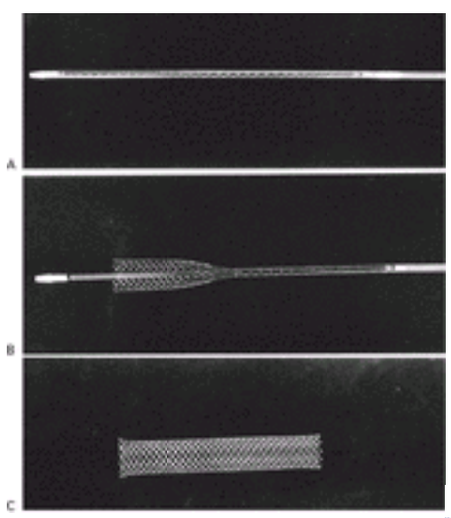

FIGURE 160-10. The Wallstent (**A**) in its constrained, elongated form, between two coaxial catheters and (**B**) partially deployed with the outer catheter partially pulled back over the inner catheter. **C:** The fully deployed stent.

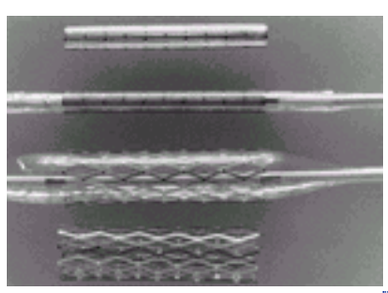

FIGURE 160-11. The Palmaz stent. From *top* to *bottom:* the stent; the stent mounted on an angioplasty balloon; the stent expanded to the size of the balloon on balloon inflation; and the stent after removal of the balloon.

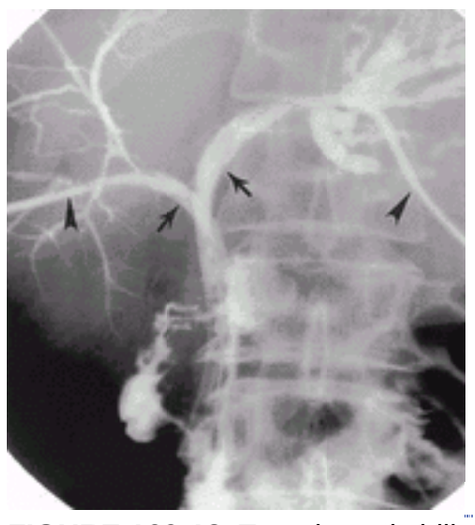

FIGURE 160-12. Transhepatic biliary stent placement in a 61-year-old man with Klatskin tumor. After bilateral percutaneous transhepatic drainage catheter placement for 6 months for hilar duct obstruction, 8-mm diameter Wallstents (*arrows*) were deployed simultaneously in both hilar ducts. The percutaneous catheters (*arrowheads*) were removed after demonstration of patency of the stents.

The common complications of endoprostheses are occlusion, migration, and cholangitis. The occlusion is usually caused by either tumor overgrowth or ingrowth or clogging of the stent by sludge or debris. [103] Plastic-covered Wallstents may reduce the rate of tumor ingrowth in patients with malignant biliary obstruction, but their application should be limited to the common bile duct to avoid obstruction of the hepatic ducts. [104]

Combined therapy with stent placement and brachytherapy with iridium-192 wire in patients with malignant biliary obstruction appears to extend stent patency to a mean of 19.5 months for cholangiocarcinoma and 4.8 months for metastatic disease. [105] The combination of arterial chemoembolization and brachytherapy after stent placement may result in additional palliation for biliary obstruction. Covered Gianturco stents have been used for malignant biliary obstruction, achieving a longer patency rate without increased complication rates. [106]

In patients with a Hutson loop (afferent limb of a Roux-en-Y choledochojejunostomy or hepaticojejunostomy sutured to the anterior abdominal wall), repeat access to the bile duct can be done for biliary drainage, cholangioplasty, or placement of biliary endoprostheses. [107], [108] and [109]

Percutaneous Cholecystostomy

Percutaneous cholecystostomy (PC) has become an effective technique in the management of acute cholecystitis. The indications for PC include (1) suspected cholecystitis in severely ill patients unlikely to tolerate cholecystectomy, (2) sepsis of unknown cause and a distended gallbladder in critically ill patients, [110] (3) acalculous cholecystitis, [111] (4) obstructive jaundice because of distal common bile duct obstruction when PTBD and endoscopic drainage are unsuccessful, [112] (5) diagnostic cholecystocholangiography in patients without dilated intrahepatic ducts or in whom PTC and ERCP are unsuccessful, and (6) gallstones requiring percutaneous removal or dissolution, in selected patients. The relative contraindications for PC include the visualization of a necrotic or perforated gallbladder wall on US examination and an uncorrectable bleeding diathesis.

Once the PC tract is matured, percutaneous techniques—including contact dissolution using methyl-tert-butyl ether or monoctanoin, mechanical lithotripsy, electrohydraulic lithotripsy, and laser lithotripsy—can be used to remove gallstones. Aside from the cost and multiple sittings required for many of these procedures, the main failing is that the gallbladder remains in situ to reform stones in 20% to 50% of patients, [113] with an increased risk for gallbladder carcinoma. [114] The rapid evolution of laparoscopic cholecystectomy has supplanted these percutaneous techniques in most circumstances because laparoscopic cholecystectomy does not leave the gallbladder in place. PC should be performed in severely ill patients who may not tolerate laparoscopic cholecystectomy.

Technique Intravenous antibiotics are given and coagulopathies are corrected before the procedure. Ideally, the procedure is performed in the angiography suite with a combination of US guidance for gallbladder puncture and fluoroscopy for guidewire and catheter placement. Extremely ill patients may be treated at their bedside with the entire procedure guided by US. The procedure is performed under local anesthesia. PC initially was performed by means of a transhepatic approach in an attempt to puncture the gallbladder through the bare area of the liver and avoid the possible complication of bile peritonitis. [115] Transhepatic puncture does not ensure puncture through the bare area, however, and transhepatic transperitoneal punctures are common. [116] Subhepatic puncture has been shown to be safe and avoids the risk for bleeding from hepatic injury. [117], [118] and [119] A variety of techniques are used for PC, including the standard Seldinger technique with needle puncture, guidewire placement, removal of the needle, tract dilation, and advancement of a drainage catheter over the guidewire into the gallbladder. Another technique more suitable for use at the bedside involves the use of a trocar. A tapered catheter with an inner stiffener and a central mandril with a sharp point are advanced into the gallbladder under US guidance. Another technique appropriate for bedside use involves a long, 22-gauge needle with a 6-Fr Teflon catheter loaded on the back of the needle. The needle is placed into the gallbladder under US guidance, and its mandril is removed. Injection of a small volume of saline and observation with US can be used to confirm the intralumenal position of the needle tip if the bile is too viscous to be aspirated through the long 22-gauge needle. A 0.046-cm guidewire is advanced through the needle, and the needle tip is positioned in the center of the gallbladder. The catheter is advanced over the needle-wire unit into the gallbladder. The needle and guidewire are removed, and the monofilament suture present within the catheter is tightened to accordion the catheter into a Z-shaped self-retaining configuration. Bile is obtained for aerobic and anaerobic bacterial cultures, and the catheter is secured to the skin. Although a small amount of contrast medium may be injected to confirm catheter position, if the PC is being performed for cholecystitis, a cholecystogram should not be performed at this time to reduce the risk for gallbladder distention and bacteremia. The catheter then is connected to gravity drainage. The use of a self-retaining catheter is essential to reduce the risk for catheter dislodgment and bile leakage. A diagnostic cholecystocholangiogram can be performed to evaluate cystic duct patency and cholelithiasis after 48 hours and after the patient's condition stabilizes. Options for definitive therapy include cholecystectomy, percutaneous stone removal, or, if the patient had acalculous cholecystitis and cystic duct patency is confirmed, eventual removal of the catheter with no further treatment. The catheter cannot be removed until the tract is mature or bile leakage can occur. Patients with transhepatic PC catheters have mature tracts at 20 days. [119] If a question exists about the maturity of the tract, it can be evaluated under fluoroscopy with injection of contrast medium. For patients with severe nonbiliary problems, the catheter frequently is left in place until other medical problems resolve.

Complications The complication rate is 5% to 8%; complications includes bile leakage, vasovagal reaction, catheter dislodgment, hemorrhage, and a 0.5% procedure-related mortality rate. [120] In comparison, emergency surgical cholecystostomy has a mortality rate of 8%. [121]

Results PC for presumed cholecystitis in critically ill patients is useful for diagnosis and treatment. Although initially it was thought that Gram stain and culture of aspirated gallbladder contents would be useful to diagnose cholecystitis, many of these patients are already on broad-spectrum antibiotics. The 48% sensitivity rate of Gram stain and 38% sensitivity rate of bile culture are sufficiently low to make gallbladder aspiration a procedure of limited value. [120] However, PC is useful as a clinical diagnostic test. Essentially all patients who have cholecystitis without a necrotic gallbladder wall will defervesce and normalize their leukocyte count after drainage. [111], [122] Severely ill patients who have ultrasonographic evidence of a necrotic gallbladder wall are less likely to respond to PC and should undergo immediate operation or a trial of PC with operation to follow if no clinical improvement is seen in 24 hours, depending on the patient's risk for surgery. [123] In patients without a necrotic gallbladder wall and no response to PC, cholecystitis is excluded, and the search for a nongallbladder source of sepsis should continue.

Biliary Duct Biopsy

The most difficult diagnostic problem of bile duct lesions is differentiating between malignancy and inflammatory disease in the patient with biliary strictures. Bile aspiration yields only 35% positive cytology. Brush biopsy, clamshell forceps biopsy, and shave biopsy are somewhat more sensitive, with sensitivities of 44% to 67% reported. [124], [125] and [126] Endoscopically, a combination of tissue sampling techniques, such as direct forceps biopsy, brush cytology, and aspiration cytology, has the highest yield. [127] An advantage of endoscopic over percutaneous tissue sampling is that it can all be performed during the initial procedure.

For the percutaneous route, in the absence of bacteremia or hemobilia following drainage, biopsy can be performed 1 to 2 days after the procedure. [128] Broad-spectrum antibiotics are administered before the procedure. The transhepatic drainage catheter is exchanged over a guidewire for an introducer, which is advanced distal to the stricture or to the duodenum, and a second safety guidewire is placed into the duodenum. A 3-mm bronchoscopy brush or a removable ureteral biopsy brush is advanced through the sheath or through an introducing catheter beyond the lesion. After brushing of the lesion has been performed, the brush is pulled into the introducing catheter or the sheath before withdrawal. The brush is cut from the wire and submitted for cytologic examination.

Another technique for biliary biopsy involves the use of forceps. After placing a safety wire in the duodenum, a 7-Fr, peel-away sheath is placed immediately proximal to the lesion, a 5.2-Fr or a 7-Fr flexible clamshell forceps is advanced to the lesion, and biopsy is performed. Clamshell forceps biopsies can also be performed through a choledochofiberscope.

Laparoscopic Injuries to the Biliary System

Both endoscopic and interventional radiologic management techniques play an important role in the treatment of patients with bile duct injuries from laparoscopic cholecystectomy. The injuries include biliary obstruction from bile duct transection or stricture, biliary leakage with formation of bilomas, and retained stones. US or CT is the initial imaging method used for the diagnosis of the biliary complications. ERCP is then performed to obtain the necessary anatomic information for therapy. If ERCP is unsuccessful or unable to cross the obstruction secondary to inadvertent ligation of the bile duct, a drainage catheter must be placed above the obstruction.

Biliary strictures that are not amenable to surgical intervention may be treated with percutaneous or endoscopic balloon dilation. Before the procedure, the patient is given broad-spectrum antibiotics. The procedure can be done on an outpatient basis with conscious sedation and local anesthetic. After placing a 0.035-inch guidewire across the stricture, a balloon catheter (usually 8 to 10 mm in diameter) is placed over the guidewire and inflated two to three times for 3 to 5 minutes. A

drainage catheter, such as an 8.5-Fr Cope-loop catheter, is left across the stricture for external drainage, and the treatment effectiveness is then assessed in 1 to 2 weeks. If a satisfactory response is demonstrated by a follow-up cholangiogram, a catheter is placed above the stricture and capped to allow internal drainage for 1 to 2 weeks before withdrawal of the catheter. Metallic stents should be reserved for patients with a recurrent biliary stricture following multiple balloon dilations who are not candidates for surgery.

Bile leak is treated with temporary diversion of bile through endoscopic biliary intubation or transhepatic stent placement. After 3 to 4 weeks of bile diversion, the plastic stent is removed over a guidewire, and a cholangiogram is performed through a sheath to show healing of the leak. If no leak is demonstrated, a drainage catheter is placed above the area of leakage and capped for 1 to 2 weeks to reassess bile duct leak before removal of the catheter. If the leak is associated with significant bile collection, percutaneous drainage is performed under CT or US guidance.

Retained common bile stones may be found after laparoscopic cholecystectomy. Endoscopic extraction after sphincterotomy is the preferred method in such patients. If an endoscopic approach is unsuccessful, a percutaneous transhepatic approach may be used. Stone extraction should be deferred 4 to 6 weeks to allow the tract to mature after placement of a transhepatic catheter. Alternatively, combined ERCP and PTC, with the PTC wire facilitating endoscopic access, may be used.

Biliary Manometry

Biliary manometry is used to assess bile pressure and flow dynamics. This is different from the sphincter of Oddi manometry performed at ERCP, which measures sphincter muscle function. Measurement of bile pressure at PTC has been shown to be useful in predicting long-term success after treatment of biliary strictures with balloon cholangioplasty. [129]

The patient is treated with prophylactic antibiotics before the examination. In patients with transhepatic biliary intubation, the indwelling tube is removed over a guidewire and an end-hole catheter is placed above the treated stricture and connected to a manometer. Pressure is measured at the level of the midaxillary line. Continuous pressure measurements are obtained before and during perfusion of 30% contrast medium at rates of 5 and 10 mL/minute for 5 minutes and at rates of 15 and 20 mL/minute for 2 minutes. The perfusion should be discontinued if the patient experiences right upper abdominal pain or chills, or if the biliary pressure increases to more than 30 cm H_2O. In the absence of a significant stricture, the biliary pressure will remain less than 20 cm H_2O during the perfusion.

Percutaneous Biliary Stone Extraction

Endoscopic sphincterotomy with stone extraction is the primary mode of treatment for choledocholithiasis. However, if a T-tube tract or transhepatic access is available, radiologically guided stone extraction may be performed. Intrahepatic stones occasionally are inaccessible at ERCP and require the percutaneous transhepatic route. Choledoscopy facilitates stone extraction. [130] Percutaneous stone extraction is a radiologically guided procedure for the treatment of biliary stones not amenable to surgical, endoscopic, or extracorporeal shock wave therapy. [131], [132] Stone removal can be done through the preexisting mature transhepatic biliary drain tracts, T-tube tracts, cholecystostomy tube tracts, and hepaticocutaneous enterostomy.

Basket removal of retained stones through the T-tube tract is a safe and simple method of removing retained stones. With the advent of the steerable catheter system, stone extraction has become a technically easy procedure. The Dormia-type basket is most commonly used for stone extraction. The steps for stone extraction are (1) removal of the T-tube over a guidewire, (2) insertion of a steerable catheter through the T-tube tract, and (3) extraction of the stone using a wire basket. After the stone has been extracted, a drainage catheter is placed in the common bile duct. When no residual stones are present, the catheter is capped for 1 to 2 weeks of “clinical trial” before withdrawal of the catheter. Large stones must be fragmented using crushing baskets or electrohydraulic lithotriptor before extraction. [133] Extracorporeal shock wave lithotripsy is effective in fragmenting bile duct stones but often requires extraction of fragments. [134], [135] Percutaneous techniques may be used for removal of gallstones in patients with high surgical or general anesthetic risk. [136] Because the percutaneous method is a gallbladder-sparing procedure, it carries a risk for recurrence of stone (40%) and symptoms (12%).

Percutaneous Management of Biliary Fistula

Biliary fistulae that originate in the liver may result from trauma, surgery (cholecystectomy, resection of hepatic tumor, and liver transplantation), or local infection, such as hydatid disease. Small biliary fistulae may close spontaneously, and no specific treatment is required. When a biliary fistula is associated with distal biliary obstruction, however, biliary decompression is usually required by using endoscopic or percutaneous transhepatic intervention. [137], [138], [139] and [140]

The nonoperative methods for managing biliary fistulae include external biliary drainage, nasobiliary catheter drainage, bile duct stent placement, and sphincterotomy. Selective embolization through a percutaneous transhepatic or endoscopic approach may be used for the treatment of biliary fistulae unresponsive to biliary decompression. [141]

PERCUTANEOUS BIOPSY

Percutaneous image-guided sampling of abdominal lesions is a safe and effective method of obtaining material for cytologic or histological analysis and for microbiologic and chemical analysis of fluid collections. Advances in CT and US have allowed accurate localization of smaller and more subtle lesions, and experience has demonstrated the safety of percutaneous biopsy through windows previously thought to be unsafe.

Technique

The optimal imaging modality for guiding a needle biopsy depends on the position and visualization of the lesion, the patient’s body habitus, the ability to introduce radiopaque contrast medium adjacent to the lesion, and the experience of the radiologist. Patients undergoing biopsy have their prothrombin time, activated partial thromboplastin time, and platelet count checked and corrected if necessary. Patients who are to receive intravenous contrast medium are hydrated and have their blood urea nitrogen and creatinine levels checked. Most patients do not require a sedative, but those who are unusually anxious or find it painful to lie in the required position are given midazolam, fentanyl, or both.

Fluoroscopy in multiple planes, most frequently with a C-arm, provides easy and efficient guidance for biopsy of large, superficially located lesions. Fluoroscopic landmarks, such as distance from a particular bony structure or position adjacent to a radiopaque stent, can be used to position the needle. If an indwelling biliary catheter is present, contrast medium can be introduced to allow fluoroscopically guided biopsy of biliary duct lesions. Fluoroscopy also is useful for biopsy of abnormal lymph nodes identified at lymphangiography that still contain lymphangiographic contrast medium.

Real-time US has many advantages for intra-abdominal biopsy. Perhaps its greatest advantage is that, unlike CT, it allows continuous monitoring of the course of the needle while it is being advanced, allowing instant readjustment of the needle path as necessary. US guidance may be applied when the lesion is ultrasonographically visible and a needle path that avoids gas-filled bowel exists because the soft tissue–gas interfaces reflect most of the sound waves and preclude visualization of the needle tip and the lesion. US also is advantageous when the ideal needle path is at a steep angle to the axial plane because such an angle makes CT guidance more difficult. However, US guidance is difficult in obese patients because fat degrades the US image.

CT is most frequently used to guide biopsies that cannot be performed reliably with fluoroscopy or US. CT usually is the imaging modality of choice for small, deep-seated lesions. Oral, intravenous, and rectal contrast medium is administered as needed for lesion visualization. To perform CT-guided biopsies, a localizing radiopaque grid is placed on the skin covering the region of the skin entry site as estimated from prior scans, and a scan at the level of the lesion is repeated. A needle path is chosen, preferably in the axial plane, and the skin puncture site is marked on the skin. The depth of the lesion and the angle between the needle path and a vertical line through the lesion are measured.

The types of needles available for percutaneous biopsy are nearly as numerous as the types of diseases for which biopsy is performed. Needles can be classified into those that obtain material for cytologic analysis and those that obtain tissue for histological analysis. Needles from 25-gauge to 14-gauge are used: the smaller sizes predominantly for aspiration cytologic study and the larger ones for obtaining a core of tissue. Aspiration cytologic study with a skinny needle (20 to 25 gauge) is the most frequently performed percutaneous biopsy. When a tissue core is desirable, the spring-loaded, mechanically triggered biopsy needles (guns) are used. These needles have a Tru-Cut–type design, and once in position, the inner or outer parts (depending on the manufacturer) of the needle are advanced rapidly forward by a

preloaded spring mechanism. These guns offer the benefit of producing less pain than the Tru Cut needle, and some 18-gauge guns produce a specimen as good as that obtained with a 14-gauge Tru Cut needle. [142]

A needle path is chosen that avoids lung, gallbladder, and spleen and, if possible, bowel and vascular structures. Passing 21-gauge needles through the stomach and small and large bowel is safe. [143] Some institutions are routinely placing 18-gauge needles through bowel without complication. It appears judicious to avoid puncturing bowel in immunocompromised patients and those with ascites.

Local anesthetic and a small skin nick precede needle placement. The needle may then be advanced directly into position. If a larger needle is being used, a skinny needle may be first positioned within the lesion, then the larger needle may be placed parallel to the smaller needle. This allows the needle repositioning common with CT guidance to take place with the smaller needle and allows the larger needle to be properly positioned on the first pass. A coaxial needle technique also is useful for small or difficult lesions. The technique involves placement of a larger needle up to the lesion with the biopsy performed through a smaller inner needle. This allows simple, repetitive sampling by removing only the smaller inner needle, assessing its yield, and if necessary, replacing the inner needle and repeating the biopsy.

Indications

In most cases, an aspiration biopsy is adequate to make the diagnosis of malignancy, although specific tumor characterization may require a core of tissue. Aspiration biopsy is useful in diagnosing acute liver transplant rejection, but core biopsy is more useful in diagnosing chronic rejection. [144] Core soft tissue should be obtained routinely whenever the cellular architecture is important for microscopic diagnosis (e.g., diffuse liver disease, likely benign focal liver lesions, lymphoma).

Contraindications and Risks

A patient who will not lie still is an absolute contraindication for image-guided needle biopsy. It is impossible to target a moving lesion, and there is the risk for visceral injury. For biopsy of abdominal lesions, patients must be capable of holding their breath for a minimum of 10 seconds so that respiratory excursion of the lesion is suspended during needle placement. A bleeding diathesis is a relative contraindication depending on the severity of the diathesis, the location of the lesion, lesion vascularity, and the size of the needle required. If a coagulopathy would otherwise contraindicate biopsy and the lesion is intrahepatic, transvenous biopsy or biopsy with embolization of the needle tract (plugged biopsy) can be performed. Ascites does not affect the major or minor complication rate of image-guided hepatic biopsy. [145]

Suspected hemangiomas have been listed as a contraindication for needle biopsy; however, after unintended aspirations of such lesions were reported, several authors reported series of safe biopsy of these two entities. [146], [147] With use of a 20- gauge cutting needle, a percutaneous hepatic biopsy can be safely performed for histological diagnosis of hemangioma. [147] A needle path traversing normal liver before reaching the lesion is recommended to reduce the possibility of hemorrhage.

Percutaneous needle biopsy is a relatively safe, invasive procedure with low morbidity and mortality. Severe pancreatitis can occur after pancreatic biopsy, and the incidence of pancreatitis increases when biopsy is performed on a normal pancreas. Needle tract tumor seeding after intra-abdominal biopsy is a rare occurrence. Traversing normal parenchyma before entering a lesion may reduce the risk for seeding. Hemorrhage is the most common complication of liver biopsy. Hemorrhage may occur as intra-abdominal bleeding or hemobilia after creation of an arteriobiliary fistula. Severe hemobilia after needle biopsy is best managed with selective embolization of the bleeding artery. Severe intra-abdominal bleeding may be more common if the needle does not initially traverse normal parenchyma.

Results

Image-guided hepatic biopsy using skinny needles is accurate in the diagnosis of malignancy in more than 90% of patients. [148] Accurate diagnosis of hemangioma can be made 100% of the time using a 20-gauge coring needle. [147] Skinny needle biopsy of the pancreas has been reported to be accurate 60% to 90% of the time. The coexistence of pancreatic carcinoma and pancreatitis may result in biopsy of the inflammatory area, which frequently cannot be distinguished by CT or US from the neoplastic area. The possibility of sampling error suggests that an initial negative result from percutaneous pancreatic biopsy should not be accepted, and either a repeat percutaneous biopsy or an operative biopsy should be performed as warranted by clinical circumstances.

Percutaneous image-guided aspiration biopsy of lymph nodes has been performed more often than large-core biopsy for evaluation of lymphadenopathy. Fine-needle aspiration of lymph nodes using cytologic examination for detection of metastatic carcinoma is highly accurate. [149] The accuracy of detecting lymphoma is about 80%. [149] Accurate typing of lymphoma by fine-needle aspiration remains problematic, but advances in immunocytochemistry [150] and flow cytometry may prove helpful. The accuracy of percutaneous core biopsies for classifying lymphomas remains to be determined.

Alternative Techniques to Biopsy

A number of catheter-based techniques have evolved that are useful in specific situations when percutaneous needle biopsy is difficult or contraindicated.

Transjugular Liver Biopsy If coagulopathy or marked ascites is present, transjugular liver biopsy provides an alternative to percutaneous biopsy because the needle does not transverse the liver capsule, avoiding hemorrhage. [151] An 18-gauge, 60-cm long automated biopsy device has been designed for obtaining liver histological samples through a jugular venous approach. The technique is similar to that used for the TIPS procedure. The right internal jugular vein is punctured using a 21-gauge, single wall access needle under US guidance. A 7-Fr, 49-cm long, curved catheter is advanced into the right hepatic vein, and contrast medium is injected to confirm the catheter position in the right hepatic vein. Through this sheath, a 14-gauge stiffening cannula is advanced into the right hepatic vein and directed anteriorly against the wall of the vein. Liver biopsy is performed near the IVC (Fig. 160-13). Biopsy from a peripheral hepatic vein may result in perforation of the hepatic capsule and increases the possibility of bleeding. This technique is successful in more than 90% of patients. [151], [152] Major complications occur in 2% of patients and include cardiac arrhythmias and intraperitoneal hemorrhage from inadvertent capsular puncture.

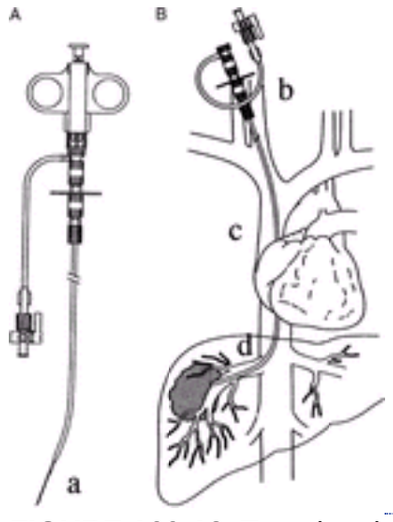

FIGURE 160-13. Transjugular liver biopsy. **A:** Liver access set assembled with Quick-Core biopsy needle. **B:** The preassembled liver access set has been introduced into the right hepatic vein over the guidewire from the internal jugular vein. The directional arrow on the stiffening cannula's hub is used to direct the needle anteriorly, and gentle pressure is applied to the wall of the central hepatic vein. The Quick-Core biopsy needle is advanced through the sheath into the liver tissue. The stylet is then advanced to expose the specimen notch (*a*), and the cutting cannula is fired to obtain tissue within the specimen notch. The biopsy needle is withdrawn, and the tissue specimen is removed from the notch and placed in formalin. *b,* Right internal jugular vein; *c,* superior vena cava; *d,* right hepatic vein.

Transhepatic Liver Biopsy with Tract Embolization When a histological core of liver tissue is needed and coagulopathy or marked ascites preclude standard percutaneous liver biopsy, embolization of the needle tract within the liver can prevent bleeding from the liver capsule puncture site. Embolization of the tract is

performed with a Gianturco coil [153] or with gelatin sponge plugs. The occluding materials may be introduced through the outer part of a Tru Cut–type needle after the inner obturator and specimen have been removed, or through a 4-Fr (1.3 mm diameter) vascular sheath fitted over the needle before biopsy. [154] Introducing the embolus through the vascular sheath is useful because the sheath is much more flexible than the needle cannula and is less likely to cause injury while the patient breathes and the liver moves. The potential disadvantage of using the sheath is that a larger hole is made in the liver capsule than would be made by the needle alone. Plugged biopsy is faster and usually less costly, and it does not require the same level of technical skill and experience required for transvenous biopsy. Plugged biopsy can be performed with imaging guidance to obtain a core sample of a focal lesion. Transvenous biopsy is not useful for most focal hepatic lesions because of the inability to steer the transvenous needle precisely. The published experience with plugged liver biopsy is too small to allow an accurate comparison of the risks of plugged liver biopsy with transvenous biopsy. Transvenous biopsy is considered safer for patients with severe coagulation disorders.

PERCUTANEOUS ABSCESS AND FLUID COLLECTION DRAINAGE

Percutaneous abscess drainage (PAD) is effective in treating abdominal abscesses and fluid collections. [155] Percutaneous needle aspiration is helpful in determining the nature of any intra-abdominal fluid collections, including abscess, biloma, lymphocele, seroma, loculated ascites, hematoma, and urinoma. Percutaneous catheter drainage is indicated for the treatment of infected fluid collections (e.g., hepatic and diverticular abscesses) and symptomatic fluid collections (e.g., pancreatic pseudocyst and lymphocele). Contraindications include absence of a safe access route to the fluid collection and coagulopathy.

Technique

Percutaneous needle aspiration and catheter drainage usually require CT or US guidance. Real-time US is most useful if the fluid collection is easily seen and a safe access route can be found, such as retroperitoneal, hepatic, or splenic drainage. C-arm–supported CT fluoroscopy is useful in PAD procedures. [156] CT guidance is preferred for drainage of deep fluid collections and is helpful in differentiating bowel loops from abscess. All patients undergoing abscess drainage receive intravenous broad-spectrum antibiotics before the catheter placement, and in anticipation of possible drain placement, bleeding diatheses are corrected.

The methods in general use for percutaneous drainage of the intra-abdominal abscesses include the Seldinger method, using guidewires, dilators, and a drainage catheter, and the trocar method, using a drainage catheter with an inner stiffener and central pointed trocar. The trocar catheter is advanced in a single pass, parallel to the initial needle, and to the same depth. The trocar technique obviates guidewire exchange and tract dilation and also theoretically reduces the risk for leakage of pus into a contaminated space during guidewire exchange. When the acceptable window of access is narrow, the Seldinger technique may be preferred, to avoid unintentional passage of the trocar catheter into a vital structure.

Single- or double-lumen (sump) catheters (8 to 16 Fr) may be used. Single-lumen catheters are connected to gravity drainage. Sump catheters may be left to gravity or low suction. The catheter is flushed with 10 mL of saline every 4 to 6 hours for the first day and once daily after the first day. The effectiveness of the drainage should be assessed with a repeat scan. The determination of successful therapy is made on the basis of abatement of symptoms and signs of infection and less than 10 mL/day of drainage, radiographic evidence of cavity closure with contrast injection, and absence of fluid collection by a repeat scan.

Hepatic Cysts and Abscess

Congenital cysts may be solitary or multiple and may be associated with polycystic kidney disease. Cysts that cause symptoms such as pain or jaundice may require treatment. Percutaneous drainage and injection of sclerosing agent into the cyst is an effective alternative to surgical treatment. [157], [158] The technique consists of percutaneous puncture of the cyst under US guidance and insertion of a 5-Fr pigtail catheter into the cyst using the Seldinger technique. After complete aspiration of the cyst, the cyst is filled with 95% ethanol in a volume of 30% of the cystic fluid aspirated. After 20 minutes of sclerosis with changes of the patient's position, the residual cystic fluid is aspirated.

Hepatic abscesses may be bacterial, echinococcal, or amebic in origin. When the diagnosis of pyogenic abscess is established, parenteral broad-spectrum antibiotic coverage is required 1 hour before percutaneous aspiration or catheter drainage. Sedation and use of analgesia with appropriate monitoring of vital signs are required during the procedure. Percutaneous drainage is safe and succeeds in about 90% of patients. [155] Effective drainage can be achieved in a loculated hepatic abscess. CT or US is required for accurate puncture of the lesion. The pleural reflection extends to the 10th intercostal space laterally. To avoid transgression of the pleural space, an angled approach, guided by US and CT, should be used for drainage of high lesions.

The traditional treatment of hydatid cysts has been surgical because of the risk for percutaneous puncture (e.g., peritoneal seeding and anaphylaxis). Although hydatid cysts were previously considered a contraindication for percutaneous aspiration, safe and successful drainage has been achieved with the use of transhepatic puncture and injection of alcohol or hypertonic saline into the cyst after aspiration. [159], [160]

The role of percutaneous aspiration or catheter drainage for the treatment of hepatic amebic abscess remains controversial. Nearly all hepatic amebic abscesses can be cured by antimicrobial drugs alone, although percutaneous drainage may be useful for patients with abscesses larger than 6 cm and for those who have failed one course of medical therapy. After a drainage catheter is placed, the catheter is irrigated with normal saline every 8 hours. The volume of irrigating saline is about 25% of the volume of the original cavity. Excessive amounts of fluid should be avoided to prevent rupture of the cavity and leakage of the fluid. Follow-up imaging of the abscess is performed at 5 to 7 days to evaluate for the effectiveness of the drainage and communication with bile ducts or bowel. When the patient is afebrile with normal white blood cell count, the 24-hour drainage volume is less than 10 mL, and a cavity has no communication with a bile duct, the tube is removed. The patients should receive antibiotics for 2 to 6 weeks.

Pancreatic Abscess and Fluid Collections

Pseudocysts, whether infected or not, are cured by percutaneous drainage in 70% to 90% of patients. [161], [162], [163] and [164] The response rate may be higher in infected than in noninfected pseudocysts. [163] Indications for percutaneous drainage include pseudocyst infection, pain associated with a pseudocyst greater than 4 to 5 cm, and a pseudocyst causing bile duct or gastric outlet obstruction. An asymptomatic pseudocyst larger than 10 cm also warrants percutaneous drainage to avoid serious complications. Somatostatin analogs reduce output from pseudocyst drainage catheters and may prove useful in reducing the duration of catheter drainage. The technique for percutaneous drainage of pancreatic pseudocysts is the same as that for other intra-abdominal fluid collections. Various access routes to pseudocysts have been used, including transgastric, transhepatic, transsplenic, and transhepatic-transduodenal approaches.

Long-term external drainage is often required for percutaneous transgastric decompression of large pancreatic pseudocysts. The initial external catheter can be converted to internal drainage by placing an internal double-mushroom stent after 14 days, eliminating the need for an external catheter. [165] When early catheter occlusion occurs, replacement or insertion of a second internal stent is done through the gastrostomy tract. When the cyst has resolved, the internal stent is retrieved by endoscopic snare.

The technique for the percutaneous cystogastrostomy involves insertion of a nasogastric tube, percutaneous transgastric puncture of the pancreatic fluid collection under CT guidance, and placement of a 10-Fr, percutaneous, transgastric catheter through an 11-Fr, peel-away sheath into the cyst. The percutaneous gastrostomy is left to gravity drainage for 2 to 3 days and, when no leakage is seen with injection of contrast medium, the tube is capped. After 14 days of external-internal drainage, the percutaneous drainage is converted to an internal drainage with placement of a stent between the cyst and the stomach under lateral fluoroscopy. Initial results in 19 patients showed a low complication and recurrence rate. [165]

Diverticular Abscess

PAD plays an important role in treating certain patients with complications of diverticulitis. [166], [167] Patients with phlegmonous changes around the colon can be managed medically. Patients with small pericolic abscesses can be treated with en bloc resection at the time of their colectomy, and patients with peritonitis require a two-stage operation. Patients who have larger pericolic, pelvic, or retroperitoneal abscesses may benefit from PAD. This subset of patients would require staged operations if PAD were not available. PAD provides for resolution of septic complications and healing of the abscess such that a delayed single-stage operation can be performed. As with all cases of PAD, if signs of sepsis do not improve within 24 to 48 hours of PAD and intravenous antibiotics, immediate operation should be undertaken unless catheter repositioning or replacement has a high likelihood of effecting improvement.

Abscesses with Fistulae

The identification of fistulae associated with an abdominal or pelvic abscess frequently is not made on imaging studies before drainage or even at the time of drainage. [168] After several days, if the quantity of drainage remains more than 50 mL/day or if the character of the drainage changes and visually or chemically resembles stool, small bowel secretions, pancreatic juice, bile, or urine, then identification of the fistula can be attempted by injecting contrast medium into the abscess cavity under fluoroscopic observation. If this is unsuccessful, an attempt can be made to identify the fistula from its other end by filling the suspected organ of origin with contrast medium; for example, a barium enema is performed if a colonic origin is suspected, or a PTC or ERCP if biliary communication is present.

The presence of a fistula does not preclude definitive PAD in most circumstances. If a downstream stenosis is present in the hollow viscus or if a foreign body or neoplasm is present at the site of perforation, then closure of the fistula will not occur without surgical intervention. [169] Low-output fistulae (less than 200 mL/day) appear to be more likely to heal than high-output fistulae. High-output fistulae may be treated more effectively by converting them to low-output fistulae with nasoenteric intubation and suction, total parenteral nutrition, and histamine H_2 blocker therapy. Catheterization of the fistula itself and advancement of a drainage catheter into the viscus is extremely useful in diverting drainage from the fistulous tract and allowing resolution. [170]

When a fistula is present in association with a percutaneously drained abscess, the duration of catheter drainage usually is longer than if no fistula is present. Drainage for several months may be necessary; however, most of these patients can be managed as outpatients. If a patient's social situation precludes outpatient treatment, operative treatment should be considered to avoid the costs and risks associated with prolonged hospitalization.

Abscesses and Crohn's Disease

The role of PAD in Crohn's disease has been debated, [171] but several points are clear. Postoperative abscesses with or without fistulae and hepatic abscesses in patients with Crohn's disease respond well to PAD. [172], [173] Abscesses in association with transmural disease of the bowel but without visible fistulae also appear to respond. [174] It is unclear what the response rate is for abscesses with fistulae from transmural disease. Nevertheless, these patients may benefit from palliation with PAD before definitive operation. PAD may control septic complications and allow improvement of nutritional status with a course of parenteral nutrition before operation. As in treatment of diverticular disease, PAD may allow conversion of a two-stage operation into a single-stage operation.

PERCUTANEOUS GASTROINTESTINAL INTERVENTION

Both endoscopic and interventional radiology techniques are currently used for gastrostomy, gastrojejunostomy, and direct jejunostomy placement. Morbidity is low, and the success rate is high for both approaches; hence, the choice between techniques depends on local expertise and availability.

Percutaneous, radiologically guided ostomy formation is performed in both adults and children with use of conscious sedation and local anesthesia. General anesthesia is required in uncooperative patients. The procedure is performed on a C-arm fluoroscopic table. Before gastrostomy tube placement, US may be used to identify the liver and spleen.

Percutaneous Gastrostomy and Gastrojejunostomy

Indications for percutaneous radiologic gastrostomy (Fig. 160-14) and gastrojejunostomy (Fig. 160-15) include (1) inadequate oral intake due to esophageal obstruction or neurologic disorders, (2) decreased gastric motility (for gastrojejunostomy), and (c) decompression in chronic small bowel obstruction.

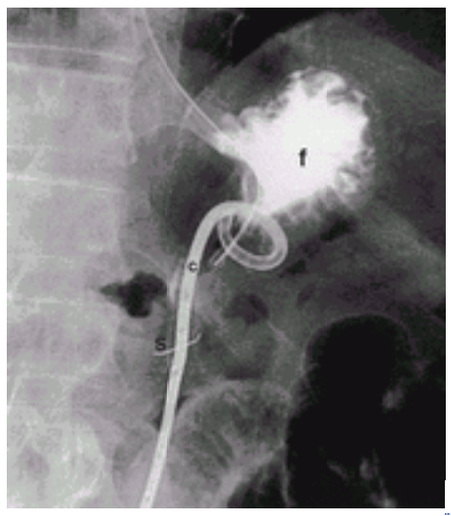

FIGURE 160-14. Percutaneous gastrostomy. A 12-Fr Cope loop gastrostomy tube (*c*) was placed after placement of a Cope suture anchor (*s*), and injection of contrast medium confirms the catheter loop in the fundus of the stomach (*f*). The nasogastric tube has been introduced into the stomach for air insufflation during puncture of the stomach.

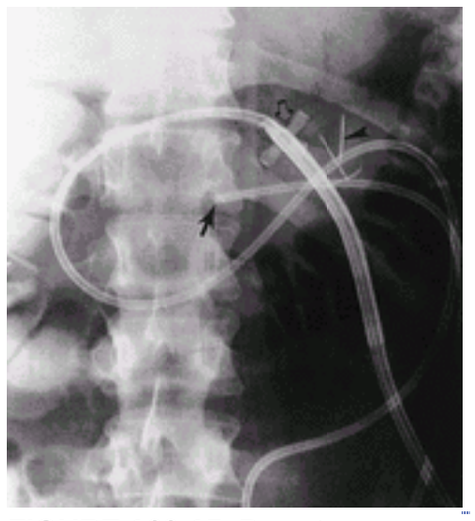

FIGURE 160-15. Percutaneous gastrojejunostomy. After gastrostomy, the pylorus was crossed using a guidewire and catheter. The tract was dilated, and a 16.5-Fr Carey-Alzate-Coons gastrojejunostomy catheter was introduced through a peel-away sheath. The distal catheter tip is positioned in the jejunum (*arrow*) beyond the ligament of Treitz, allowing enteral feeding immediately following tube placement. Three anchor sutures (*arrowhead*) have been placed in the stomach before tract dilation. The friction-lock Malecot tip (*open arrow*) prevents tube dislodgment.

The contraindications for gastrostomy or gastrojejunostomy tube placement include (1) uncorrectable bleeding diathesis; (2) inaccessible stomach from prior gastric pull-up, total gastrectomy, or partial gastrectomy; (3) clinical instability; (4) limited life expectancy; and (5) presence of a ventriculoperitoneal shunt. Ascites has been

considered a contraindication for gastrostomy, although successful placement in patients with ascites has been reported. [175] Pericatheter ascitic leak may be avoided with preprocedural paracentesis and, if necessary, postprocedural paracentesis to promote skin healing. Gastropexy sutures are used in patients with marked ascites to prevent catheter dislodgment by preventing increasing amounts of ascites from displacing the stomach from the anterior abdominal wall. Two to three suture anchors may be placed before tract dilation. The suture may be left in place for 1 to 2 weeks, or the suture may be cut after placement of a gastrostomy tube. Bacterial seeding of the ascitic fluid remains a potential problem.

Technique A nasogastric tube is placed the evening before the procedure, or, if a nasoenteric feeding tube is present, it is left in place. The patient is given nothing by mouth or through the nasogastric tube except for medications beginning at midnight before the procedure. Glucagon is administered immediately before the needle puncture of the stomach to reduce the egress of air from the stomach. Distention of the stomach brings its anterior wall closer to the anterior abdominal wall, displaces the small and large bowel, and brings the stomach out from under the costal margin to facilitate puncture. Inflation also allows fluoroscopic visualization of the stomach. If no nasoenteric tube can be passed blindly because of esophageal obstruction, in nearly all cases, a small-bore tube or catheter can be placed through the obstruction using standard angiographic techniques. For patients who cannot be transported to the angiography suite or who are unable to remain in supine position, percutaneous gastrostomy can be performed at the bedside under portable real-time US guidance. [176] The radiologic technique in general use for ostomy formation is the Seldinger method, using a guidewire, needle, dilators, and catheters. The puncture site is chosen fluoroscopically so that the needle enters the body of the stomach away from the greater curvature, avoiding the gastroepiploic vessels, epigastric vessels, the colon, and the left lobe of the liver. Oblique fluoroscopy is useful to confirm that the stomach abuts the anterior abdominal wall without intervening structures at the puncture site. The chosen puncture site is anesthetized with lidocaine, and a skin incision, large enough to admit the selected tube, is made. Gastropexy sutures may be placed with a modified 18-gauge needle. [177] Under fluoroscopic guidance, an 18-gauge needle attached to a 10-mL syringe half-filled with water-soluble contrast medium is advanced into the stomach with a brisk motion during aspiration with the syringe. Entry into the stomach is observed by free aspiration of air and is confirmed by injection of contrast medium and visualization of rugal folds. A 0.038-inch (0.97-mm) guidewire is advanced into the stomach, and the tract is dilated. If a gastrojejunostomy tube is to be placed, the catheter and a long, torqueable guidewire are used to traverse the pylorus and advance the guidewire through the duodenum into the proximal jejunum. The gastrojejunostomy catheter then is advanced over the guidewire into the jejunum through a peel-away sheath. Catheters from 2.64 to 8.58 mm (8 to 26 Fr) are used for gastrostomy, with the smallest catheters used in the pediatric population for feeding and the largest catheters used in adults for gastric decompression. Retention devices include balloons, Cope loops, and Malecot tips. Injection of contrast medium confirms the final position of the gastrostomy or gastrojejunostomy tube.

Large-bore gastrostomy tubes and endoscopic-type gastrostomy tubes may be safely placed using the same radiologic technique. [178] The gastrostomy or gastrojejunostomy tube is secured at the skin with a Molnar disk or friction fitting to stoma adhesive. If for any reason there is a high risk for catheter dislodgment, the tube may be sutured to the skin. The patient is observed for 6 hours for signs of bleeding or peritoneal irritation. If a gastrostomy tube has been placed, it is left open to gravity drainage overnight, and tube feedings begin the following morning. If a gastrojejunostomy has been placed, tube feedings may begin immediately. The stitches (and gastropexy fasteners, if used) are removed after 10 days. The tract usually is mature enough by 7 to 10 days that, if the tube is accidentally dislodged, the tract can be recatheterized and a de novo gastrostomy avoided if replacement is performed within 24 hours.

Feeding Gastrostomy versus Feeding Gastrojejunostomy Because adequate data are not available, radiologists remain divided into two camps: those who place gastrostomy tubes in most patients and gastrojejunostomy tubes only when gastroesophageal reflux is proved, and those who place gastrojejunostomy tubes in all patients. The most serious complication of tube feedings is aspiration pneumonia. Some evidence exists that surgical gastrostomies induce reflux; however, percutaneous radiologic gastrostomy does not induce reflux. No study has demonstrated that endoscopic or radiologic percutaneous gastrojejunostomy results in less aspiration than percutaneous gastrostomy. Gastrostomy tubes are superior for most patients. Most of the aspiration in these patients is due to oropharyngeal aspiration. Gastrojejunostomy tubes require greater maintenance than do gastrostomy tubes (see Fig. 160-15). Gastrojejunostomy tubes have occluded more frequently than gastrostomy tubes because the jejunostomy portion of the tubes has been limited in size to 3.3 to 3.96 mm (10 to 12 Fr). Poorly crushed pill fragments or concretions of tube feedings may block the smaller tube and necessitate tube replacement. The greater length of gastrojejunostomy tubes may also contribute to this problem. Gastrojejunostomy tubes are harder to manage at home or at chronic care facilities because they require mechanical pump infusion to achieve controlled infusion rates in the small intestine. In contrast, gastrostomy tubes can be infused with intermittent boluses or by gravity drips. If gastroesophageal reflux and aspiration occur in a patient with a gastrojejunostomy tube or in a patient with intractable gastroparesis, a better result may be achieved with two tubes: one placed into the jejunum for feeding and one placed into the gastric fundus for decompression.

Complications and Results A compilation of four major published series of radiologic gastrostomy and gastrojejunostomy tube placement [179] in a total of 635 patients revealed a procedure-related mortality rate of 1% and a major complication rate of 5%. Major complications included peritonitis requiring laparotomy, bleeding requiring transfusion, external leakage, aspiration, gastric or duodenal perforation, and deep abdominal wall infection. Minor complications occur in 5% of patients. A comparison of radiologic with endoscopic gastrostomy and surgical gastrostomy revealed that radiologic gastrostomy is associated with a higher success rate than is percutaneous endoscopic gastrostomy and less morbidity than either percutaneous endoscopic gastrostomy or surgery. [180] Patients were not randomized in this study, however, making comparisons difficult. Percutaneous radiologic gastrostomy or gastrojejunostomy can be performed in essentially all patients in whom the procedure is not contraindicated. Large-bore 20- to 24-Fr gastrostomy and gastrojejunostomy tubes can be placed under fluoroscopy. The technical success, morbidity, and mortality rates are similar to those of tubes placed surgically or endoscopically. [181] In pediatric patients with neurologic or oropharyngeal impairment, fluoroscopically guided gastrostomy provides a safe and effective means for enteral nutrition and gastric decompression. Colonic interposition, microgastria, and hepatosplenomegaly are the cause of access failure of 1.2%. [182] The procedure can be performed with use of sedation and local anesthesia in nearly 90% of children.

Percutaneous Jejunostomy

When percutaneous gastrostomy is not possible because of previous gastrectomy or an abnormal stomach position, direct percutaneous jejunostomy can be performed for enteral feeding. [183], [184] Under fluoroscopic guidance, a nasogastric tube is passed into the stomach and through the pylorus. One milligram of glucagon is administered intravenously to decrease peristalsis. The small bowel is then filled with room air. Using C-arm fluoroscopy, a loop of bowel adjacent to the anterior abdominal wall is identified, a 21-gauge needle is inserted into the jejunal lumen, and injection of contrast medium confirms the needle position within the jejunal lumen. A 0.018-inch (0.046-cm) guidewire is inserted, and a 6.3-Fr introducer catheter is advanced into the jejunum. Over a 0.038-inch (0.097-cm) guidewire, the tract is dilated, and an 8-Fr Cope-loop nephrostomy catheter is placed in the jejunal loop. Pericatheter leakage is the main complication. The use of Cope suture anchors facilitates a feeding tube insertion by securing the jejunal loop to the anterior abdominal wall.

In the absence of a previous surgical jejunostomy scar, this procedure is considered risky and is not widely performed. In patients who have undergone esophagectomy and a previous surgical jejunostomy, the jejunal loop that has been surgically fixed to the anterior abdominal wall can be punctured using the metal clips as a guide. [185] The skin mark or a dimple at the site of the previous surgical jejunostomy is used to guide the needle puncture into the bowel loop.

Percutaneous Cecostomy

Percutaneous cecostomy is an alternative to surgical and endoscopic decompression of the cecum in cases of colonic obstruction and pseudoobstruction (Ogilvie syndrome). [186], [187] Small 2.31- to 2.97-mm (7- to 9-Fr) catheters can be placed through the peritoneum with a Seldinger or trocar technique for decompression of the colon, and large 7.92- to 9.9-mm (24- to 30-Fr) catheters are placed for drainage of liquid stool after placement of anchoring sutures, as used for gastrostomy. This technique is applicable to patients with Ogilvie syndrome in whom endoscopic decompression fails and to patients with colonic obstruction whose medical condition precludes operation.

Balloon Dilation and Stent Placement for Gastrointestinal Obstruction

Balloon dilation or metallic stents may be used as alternatives to surgery for malignant and benign strictures in the esophagus and GI tract (stomach, pylorus, duodenum, colon, and rectum) using radiologic or endoscopic techniques. [188] Generally, balloon dilation is used for benign strictures, whereas stents are reserved for patients with malignant strictures.

The radiology-guided technique is similar to that used for PTA. It can be performed on outpatients under sedation and local anesthetic. The strictured site is identified by injection of contrast medium or endoscopy. After placing a 0.035-inch (0.89-mm) guidewire across the stricture, an angioplasty balloon catheter is advanced over the guidewire, placed across the stricture, and inflated two to three times for 3 to 5 minutes. Water-soluble contrast medium should be injected after dilation to rule out any perforation.

Metallic stents have been used for treatment of esophageal obstruction, gastric outlet stenosis, duodenal stenosis, and malignant colorectal stricture. [189], [190], [191], [192], [193], [194] and [195] The technique for stent placement is similar to that used for angioplasty. Use of metallic stents in patients with colonic obstruction caused by malignant neoplasms eliminates the need for emergency surgical intervention on unprepared bowel, reducing mortality and morbidity. [196] The procedure can be performed on an outpatient basis or requires minimal hospitalization, permitting a single-stage surgery. The procedural steps for colonic stent placement are as follows: the patient is

placed in the supine position on a fluoroscopic table, and the combination of a 0.038-inch (0.97-mm), stiff-angled hydrophilic guidewire and a catheter is advanced through the rectosigmoid colon under fluoroscopy to the area of obstruction. After identification of the obstruction by injecting water-soluble contrast material, the catheter and guidewire are advanced through the lesion. The length of the obstruction is measured, and a self-expanding Wallstent is deployed over a guidewire or through an endoscope if desired. The correct position of the stent and its patency is assessed by a barium examination. Complications occur in 10% of patients, including perforation, migration, hemorrhage, and sepsis.

Malignant Tracheoesophageal Fistula

A fistula may develop between the esophagus and the tracheobronchial tree from invasion of mediastinal structures by esophageal carcinoma. This may lead to pneumonia, lung abscess, and death. Covered esophageal stents can provide effective palliation of symptoms caused by the tracheoesophageal fistula. Simultaneous placement of a tracheal stent may be required for tracheal compression by the stent and tumor invasion. [197], [198] Although radiologic placement has been reported, most of these stents are placed endoscopically.

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CHAPTER 161

Nathaniel J. Soper

LAPAROSCOPY AND LAPAROTOMY

Although laparoscopy for examination of the peritoneal surfaces and diagnosis of intra-abdominal pathology has been used since the turn of the 20th century, its popularity has waxed and waned over the years. Application of laparoscopic technology and techniques has recently been stimulated by the introduction of many therapeutic laparoscopic operations. [1] This resurgence of therapeutic laparoscopy has stimulated a renewed interest in the possibilities of diagnostic laparoscopy. For decades, this modality had been largely neglected as major advances were made in diagnostic imaging techniques; well-controlled comparison studies are now proving that diagnostic laparoscopy holds many potential advantages over these imaging techniques. [2]

Similarly, the enthusiasm with which the surgical community greeted laparoscopic cholecystectomy led to a plethora of other therapeutic procedures being performed using laparoscopic guidance. The unbridled zeal of some of the laparoscopic enthusiasts has occasionally led surgeons to perform laparoscopic operations that may be better performed with a standard laparotomy. [1] The results of outcome studies will show the proper role of many of these therapeutic laparoscopic procedures with regard to morbidity, mortality, and cost. Although laparoscopy and other “minimally invasive” technologies will likely continue to expand, a number of patients will still require laparotomy for diagnosis and therapy. This chapter summarizes the current status of diagnostic laparoscopy, therapeutic laparoscopy, and laparotomy.

HISTORY OF LAPAROSCOPY

The technique of accessing the abdomen through small incisions to diagnose and treat abdominal disease is today commonly referred to as laparoscopy (Greek *lapara,* the flank; *skopein,* to view). In 1901, G. Kelling performed the first minimally invasive examination of the abdomen using a cystoscope in a dog. [3], [4] A decade later, H. Jacobaeus reported the safe utilization of laparoscopy in patients to diagnose syphilis, tuberculosis, cirrhosis, and malignancy. [5] The German hepatologist H. Kalk introduced the oblique-viewing laparoscope for diagnosis of liver diseases and the concept of using accessory ports for biopsy. [6] Laparoscopic access was simplified by the introduction of the Veress spring-loaded needle in 1938, which was used to insufflate the abdomen with air. [7] Subsequently, carbon dioxide (CO_2) was used to create the pneumoperitoneum necessary to have a working space. Fiberoptics brightly illuminated the abdominal cavity with superior lighting, and as instruments for grasping, cutting, and cauterization became available, simple therapeutic procedures were possible.

The modern era of video laparoscopy began with the development of the computer-chip television in the 1980s. With the laparoscope attached to a television camera, the resulting images could be transmitted to a video monitor, and the whole operating team could visualize the operative field. No longer was the surgeon restricted to “scope holder.” With an assistant delegated as camera holder, the surgeon was free to operate with both hands and perform more difficult procedures. P. Mouret

performed the first laparoscopic cholecystectomy in France in 1987, [8] followed by Reddick and Olsen [9] and McKernan and Saye [10] in the United States in 1988.

The appeal of small incisions and claims of less pain, shorter hospitalization, and speedier recuperation after laparoscopy were portrayed by the media as "bellybutton," "Band-Aid," "Nintendo," and "minimally invasive" surgery. [11] Before randomized prospective studies could verify the proposed benefits of laparoscopy, general surgeons embraced the new methods, and other operations were performed using laparoscopic guidance. Hospitals invested in costly laparoscopes, monitors, and instruments. Surgical societies, journals, and even postgraduate fellowships were established to teach, validate, and credential surgeons in emerging laparoscopic procedures. Today, detailed outcome studies are being performed and reported to justify the outcomes and costs of these new procedures relative to alternative treatment strategies. Thus far, only laparoscopic cholecystectomy has succeeded in becoming the gold standard approach, [12] but many other laparoscopic abdominal procedures are rapidly gaining acceptance (Table 161-1).

Accepted
Diagnostic laparoscopy
Laparoscopic cholecystectomy
Operations for gastroesophageal reflux
Splenectomy
Adrenalectomy
Nephrectomy
Gaining Acceptance
Exploration of common bile duct
Appendectomy
Repair of inguinal hernia
Operations for peptic ulcer disease
Resection of colon
Esophagomyotomy
Operations for morbid obesity
Anecdotal Experience
Biliary bypass procedures
Pancreatic resection
Esophageal resection
Ablation of hepatic tumors

TABLE 161-1 Current Status of Laparoscopic Procedures

DIAGNOSTIC LAPAROSCOPY

Advantages

Diagnostic laparoscopy has many potential advantages over other diagnostic studies and traditional open laparotomy. Compared with other imaging modalities, diagnostic laparoscopy more realistically characterizes a lesion's color and contour; and enhanced resolution and magnification may detect lesions smaller than 1 mm, a size below the resolution of both computed tomography (CT) scan and magnetic resonance imaging (MRI). In addition to improving diagnostic accuracy, laparoscopy also allows directed biopsy with a reduced risk for bleeding. Furthermore, overall cost of the procedure may be less than that of multiple noninvasive tests.

Compared with exploratory laparotomy, diagnostic laparoscopy leads to less postoperative pain and intestinal ileus. Small trocar puncture sites are also cosmetically more appealing than large laparotomy incisions. By avoiding the morbidity of a laparotomy, otherwise healthy patients can usually be discharged from the hospital within 24 hours of a diagnostic laparoscopy and return to normal activity within a few days.

Disadvantages

Diagnostic laparoscopy also has several disadvantages. Patients should be acceptable candidates for general anesthesia. Even if laparoscopy is performed under local anesthesia, inadvertent perforation of a viscus or uncontrolled hemorrhage may occur and require formal laparotomy. Two-dimensional video systems lack depth perception, distort anatomic relationships, and hinder task performance. Elongated laparoscopic instruments allow the transfer of only minimal haptic information, and structures in the abdomen and retroperitoneum may not be palpated. Instead, the laparoscopist relies on visualizing the surfaces within the abdominal cavity. Some patients are not suitable candidates for laparoscopy because of intra-abdominal adhesions that are too extensive to allow visual exploration using a laparoscope. Adequate pneumoperitoneum and exposure of the operative field may be difficult to maintain because of continuous gas leaks, unrecognized perforated viscera, or excessive irrigation. Intraoperative complications are also more challenging to manage laparoscopically than during open surgery, especially control of brisk hemorrhage, and the laparoscopy suite should be equipped to deal with these rare emergencies. Still, the major concern with diagnostic laparoscopy is whether pathology is being overlooked. To minimize this problem, just as with traditional laparotomy, the laparoscopist needs to develop a systematic approach to examining the entire abdomen.

Technological advances may solve many of the current limitations of laparoscopy. Three-dimensional video systems restore depth perception, perhaps enhancing the safety and accuracy of laparoscopic examination and biopsies compared with two-dimensional systems. Intraoperative laparoscopic ultrasonography permits directed evaluation of organ parenchyma beyond the visible surface. Mechanical and robotic camera holders free the hands of the surgical team members to perform other tasks. [13] Alternatively, the pneumoperitoneum with its potential risks can be avoided using various abdominal wall retractors. [14], [15] and [16]

TECHNIQUE OF LAPAROSCOPIC ABDOMINAL EXPLORATION

Preoperative Care and Anesthesia

Ideally, patients are fasted from midnight before the operation. Patients without other major medical problems are admitted to the hospital the morning of the operation and given preoperative sedatives. If the possibility of intraoperative contamination exists, a single dose of intravenous antibiotics is administered. Gastric distention is diminished by administering both intravenous metoclopramide and a histamine H_2 receptor antagonist. Sequential compression stockings are placed on both legs to avoid pooling of blood in the lower extremities. In patients with known malignancy and in those at risk for venous thromboembolism, minidose subcutaneous heparin may be used. After induction of anesthesia, a bladder catheter and orogastric tube may be placed to decompress these hollow organs before inserting laparoscopic trocars. For more emergent exploration, an orogastric tube is placed and the stomach drained before induction to prevent aspiration. The abdomen is prepared and draped sterilely.

Local Anesthesia

Bupivacaine (0.5%) is injected at the initial port site and is used to a maximum dose of about 2.5 mg/kg. The skin and peritoneum surrounding the site of incision should be adequately infiltrated with anesthetic before inserting the laparoscopic trocar. All patients receive oxygen by nasal cannula or mask. As an adjunct, intravenous sedation or inhaled nitrous oxide may be added. Intravenous agents for sedation and amnesia may include a benzodiazepine, commonly midazolam, and a short-acting opioid such as fentanyl. Nitrous oxide also has analgesic properties, but inhalational agents are contraindicated if the patient has eaten within 6 hours of operation because of the high risk for aspiration with pneumoperitoneum. Routine patient monitoring would include blood pressure, pulse oximetry, and electrocardiography. Nord and Boyd [17] reported 357 patients examined by diagnostic laparoscopy under local anesthesia. None required intubation, general anesthesia, or ventilatory support. Transient vasovagal reactions occurred in 3% of patients, but hypotension generally responded promptly to atropine. When performing diagnostic laparoscopy under local anesthesia, one should consider using nitrous oxide, rather than CO_2, for creation of the pneumoperitoneum. This agent has anesthetic properties and is less irritating to the peritoneum. Because local anesthesia is safe for most patients, cost-containment issues will probably encourage more procedures to be performed under local anesthesia as an outpatient rather than under general anesthesia in the operating room.

General Anesthesia

General anesthesia allows pain-free probing of inflamed tissues and biopsy of peritoneal lesions, and when necessary, it facilitates rapid conversion to open laparotomy. Adequate depth of anesthesia, complete muscle relaxation, and administration of antiemetics are important considerations for optimal anesthetic management. Postoperative pain is lessened by preincisional subcutaneous injection at the port sites with a long-acting local anesthetic, such as 0.5% bupivacaine. [18]

Instrumentation

The laparoscopes may be 2 to 10 mm in diameter with a forward-viewing (0-degree) or obliquely angled lens. Smaller laparoscopes are more commonly used in the emergency room or office setting. Larger laparoscopes have the advantage of supplying more light and a wider visual field. The 30- to 45-degree angled lenses diminish illumination but permit a more thorough examination of the extremes of the peritoneal cavity, whereas a straight scope only views objects straight ahead. Other standard instruments for diagnostic laparoscopy include scissors, grasping devices, liver retractor, blunt probe, Babcock clamp, hook cautery, cupped forceps, biopsy forceps, uterine retractor, and a hollow suction-irrigator probe. The patient lies supine or in the lithotomy position on a table that has multiplanar movement to maximize gravitational retraction of intra-abdominal organs.

Access and Pneumoperitoneum

A working space (pneumoperitoneum) in the abdominal cavity is established by insufflating gas using a high-flow insufflator. The pneumoperitoneum is established by either a closed or an open technique. [19]

Closed Technique of Establishing Pneumoperitoneum The patient is placed in a 10- to 15-degree Trendelenburg position, and a small skin incision is made into the subcutaneous tissue of the infraumbilical skin fold. In patients who have had previous abdominal surgery, an alternative site distant from previous incisions is chosen for initial access. With an upper midline scar, a right or left lower quadrant insertion site is chosen two thirds of the distance from the umbilicus to the iliac crest lateral to the rectus sheath. Adhesions from a lower midline scar may be avoided by an initial puncture in the right or left upper abdomen at the lateral edge of the rectus muscle. The positions of the liver, spleen, and bladder relative to the port site must be ascertained before needle insertion to avoid iatrogenic injury. The lower abdominal wall is grasped to elevate the fascia safely away from the intra-abdominal organs before needle puncture. The Veress needle is then inserted at a right angle to the abdominal wall, usually at a 45-degree angle off the vertical axis toward the pelvis but away from the aortic bifurcation and iliac arteries (Fig. 161-1). Two or three clicks of the spring-loaded obturator may be heard as the needle passes through the fascia and peritoneum into the peritoneal cavity. Absence of blood, urine, or stool on aspiration and rapid flow of sterile saline through the needle by gravity (a positive drop test) corroborate the correct needle position. Elevating the lower abdominal wall during the drop test will decrease intra-abdominal pressure and enhance free flow. The abdominal cavity is then insufflated with the appropriate gas to a pressure of 8 to 15 mm Hg, beginning at a flow rate of 1 L/minute.

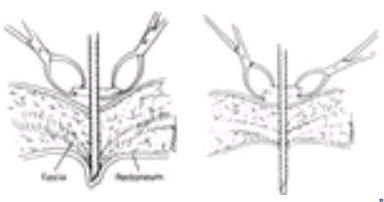

FIGURE 161-1. In the closed insertion technique, a Veress needle is blindly inserted at the umbilicus through the fascial layers of the anterior abdominal wall. (From ref. [19].)

After the abdominal cavity is distended, the Veress needle is removed, and a trocar is inserted carefully into the abdominal cavity while manually exerting anterior countertraction on the abdominal wall. The resistance of the fascia and peritoneum is apparent as the trocar slowly penetrates the abdominal wall. The sharp inner trocar is removed, and the sheath may be secured at the skin level with adhesive patches, screw threads, internal balloons, or suture. Blood-stained fluid and frank blood after trocar insertion are ominous signs. If trocar insertion is followed by brisk return of blood and hemodynamic compromise, a laparotomy should be performed immediately while leaving the trocar in place. Delay in converting to formal laparotomy has resulted in mortality from injury to a major blood vessel.

Open Technique of Establishing Pneumoperitoneum The closed technique is applicable in most patients; for patients who have undergone previous abdominal surgery, however, and for patients who are pregnant or have distended bowel, we generally use the open technique. Open port insertion is similar to the cut-down procedure used for catheter insertion during diagnostic peritoneal lavage (Fig. 161-2). A vertical or horizontal 1.5-cm skin incision is made in the infraumbilical skin fold. Blunt dissection of the subcutaneous tissue is performed deep to the skin until the umbilical raphe's insertion into the fascia is clearly visible. A clamp is applied to the fascia, which is elevated, and a small vertical incision is made into the peritoneal cavity. A finger or curved Kelly clamp is carefully introduced into the incision, and any loose adhesions may be swept away. A blunt-tipped (Hasson) port is then placed under direct vision through the fascial incision into the peritoneal cavity. The pneumoperitoneum is established only after ensuring safe access. The Hasson port is secured with sutures on both sides of the fascial incision.

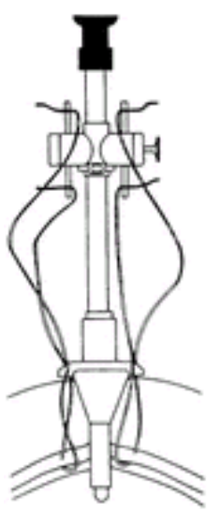

FIGURE 161-2. In the open technique, a wedge-shaped Hasson trocar is placed after peritonotomy and secured with sutures prior to insufflation with CO_2. (From ref. [19].)

Open techniques take a few minutes longer than the closed technique for initial port insertion; however, open insertion is especially advantageous in patients with previous abdominal incisions and in situations in which the Veress needle position is in doubt. At the conclusion of the operation, all trocar entry sites larger than 5 mm should be closed to avoid the risk for postoperative herniation. [20]

Gases Used for the Pneumoperitoneum For diagnostic laparoscopy, several gases may be used to insufflate the pneumoperitoneum, which creates the space for viewing and working (Fig. 161-3). Many laparoscopists use nitrous oxide during short cases and during cases performed with local anesthesia because it is inexpensive and associated with a lower incidence of cardiac arrhythmias than CO_2. Nitrous oxide also does not irritate the peritoneum (as does CO_2) and, when absorbed, does not cause metabolic abnormalities. However, nitrous oxide will support combustion and is therefore contraindicated when the use of electrocautery is anticipated. [21] Nitrous oxide is also less soluble in blood than CO_2 and may be more likely to cause gas embolism. CO_2, on the other hand, is commonly used during therapeutic laparoscopy because it is noncombustible. CO_2 is eliminated rapidly from the systemic circulation and will not form gas emboli under experimental conditions unless infused into a systemic vein at a rate greater than 1 L/minute. [22] However, CO_2 absorption may lead to hypercarbia and respiratory acidosis in patients with chronic obstructive pulmonary disease. [23] CO_2 also may cause postoperative discomfort because of referred diaphragmatic pain as it is converted on the moist peritoneal surfaces to carbonic acid.

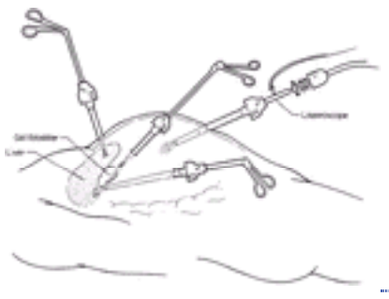

FIGURE 161-3. Cross section of abdomen after insufflating the pneumoperitoneum. Laparoscopic instruments are inserted through accessory ports. (From ref. [19].)

Gas embolism may cause right ventricular outflow obstruction signaled by a decrease in end-tidal CO_2, hypotension, or a "millwheel" heart murmur. When this life-threatening complication is suspected, the patient is placed immediately in the Trendelenburg (head-down) position with the left side down, and the gas is promptly aspirated through a central venous catheter. [24] Other problems encountered during laparoscopy include hypotension because of decreased venous return and diminished cardiac output, bradycardia due to vagal reactions, and acidosis secondary to hypercarbia. Fortunately, most of these adverse conditions improve or resolve by deflating the pneumoperitoneum. They also may be circumvented altogether by external retraction devices, which create working space by lifting the abdominal wall without pneumoperitoneum. [14], [15] and [16]

Intraoperative Ultrasonography

In the past, a criticism of diagnostic laparoscopy was that only the surfaces of organs could be visualized, and parenchymal disease was overlooked. Intraoperative laparoscopic ultrasonography improves the sensitivity of abdominal exploration when performed by laparoscopists skilled in ultrasonographic interpretation. [25] This technology has proved particularly useful in the staging of malignancy. [25], [26], [27], [28] and [29] With experience, laparoscopists have become quite facile with this technology, and we are using ultrasonography for more and more applications. [2]

Endoscopic Transillumination

Laparoscopically assisted panenteroscopy has been performed in select patients. Patients with obscure gastrointestinal bleeding or inaccessible small bowel lesions, who otherwise would require open laparotomy, may benefit from this novel approach. With the help of the laparoscopist, the endoscopist can quickly advance an enteroscope for intralumenal examination. In an animal model, Bleau and colleagues [30] used laparoscopically assisted panenteroscopy to reach the ileocecal valve in about 30 minutes without perforation. Moreover, distention and endoscopic transillumination of the bladder, stomach, and colon can sometimes aid in diagnosis during laparoscopy. [31] Transillumination of the duodenum and small bowel delineates tumors, vascular malformations, and other lesions that may be present.

Tissue Sampling

Laparoscopically directed biopsy of lesions is more accurate than blind percutaneous biopsy. In a review of several studies, Nord [32] found a false-negative rate of 24% (range, 1% to 67%) for blind percutaneous biopsy of the liver and an average false-negative rate of only 9% (range, 4% to 18%) for laparoscopically guided biopsy. Directed transabdominal ultrasonographically guided biopsy is comparable to laparoscopy in focal and diffuse disease of the liver; however, ultrasonography and CT scan are relatively insensitive to metastatic disease. [17]

For most solid hepatic tumors, a Tru-Cut biopsy is sufficient to make the diagnosis. Several biopsy instruments are available that can be passed under laparoscopic guidance through the abdominal wall or introduced through an accessory port. Bleeding is usually controlled by direct pressure. Most small exophytic lesions are ideally suited for the punch forceps. This instrument has a cup-shaped blade that may be used to remove a small tissue sample without tearing adjacent structures.

If larger tissue samples are required, an incisional biopsy is made, and the specimen is retrieved in an entrapment sac. Control of hemostasis should be considered before biopsy. Good exposure should be achieved, and hemostatic agents (e.g., cellulose, collagen, or thrombin) should be readily available. Most surface bleeding is controlled by monopolar electrocautery. Laparoscopic suturing may be necessary, and the laparoscopist should develop expertise in both intracorporeal and extracorporeal suturing techniques.

Advanced laparoscopic procedures may be indicated during diagnostic laparoscopy. For large tissue specimens, morcellation of an organ may be performed to retrieve the tissue through a small skin incision. The spleen, for example, after laparoscopic splenectomy may be placed in a sac and divided into smaller fragments until the contents can be retrieved through a 10-mm incision. Morcellation destroys tissue margins; hence, its use for malignancy is limited.

Cysts may be needle aspirated, but at the risk of determining that the lesion is in fact a hemangioma. These lesions are notorious for uncontrolled hemorrhage after biopsy of any kind. Needle aspiration also may be used to confirm gallbladder cancer by cytologic examination of bile. One method to prevent contamination of the peritoneal cavity is to introduce the needle through the liver rather than by direct puncture of the gallbladder.

The laparoscopic suction-irrigator is designed to aspirate fluid. Large quantities of ascitic fluid are easily collected by this means. Similarly, peritoneal washing may be done by injecting 20 to 30 mL of sterile saline and then aspirating the contents into a collection container.

Postoperative Care

Patients are either observed in the hospital overnight or discharged later the same day after diagnostic laparoscopy. Common problems encountered in the immediate postoperative period include urinary retention and nausea. Antiemetics and analgesics are given as needed. Most patients tolerate clear liquids in the immediate postoperative period and resume a regular diet the next day. Nausea and mild shoulder pain due to diaphragmatic irritation may occur in the early postoperative period and may be diminished by the routine administration of ketorolac. [33] Patient activity is not restricted, and abdominal tenderness usually subsides by the second or third postoperative day. Patients discharged home the same day should be advised about these problems and should live relatively near the hospital, preferably with another person. Within 1 week, patients are usually ready to resume their normal level of activity.

ABDOMINAL INSPECTION

Whenever a laparoscope is inserted into the abdomen, the entire abdominal cavity and pelvis should be explored systematically and thoroughly. For diagnostic exploration, most laparoscopists recommend a 5-mm laparoscope, whereas for therapeutic laparoscopy, a 10-mm laparoscope is usually used. A second trocar placed in the flank may be used to divide adhesions if necessary to facilitate a thorough exploration. A third port may be necessary to allow a second working instrument to help retract, "run" the bowel, or obtain biopsy samples. After accessing the abdomen, the abdominal viscera and retroperitoneum immediately posterior to the initial port are first viewed to ensure that no injury has resulted from inserting the trocar. If access is uneventful, the abdomen is examined systematically. The following discussion highlights just a few of the more common findings that the laparoscopist encounters during abdominal inspection.

Pelvis

The pelvic viscera are examined for pathological abnormalities before evaluating the upper abdomen. The patient is placed in the Trendelenburg position. In women, the ovaries, fallopian tubes, and uterus should be inspected. If pelvic pathology is expected, a uterine manipulator is used to elevate the uterus and increase access to the adnexa, cul de sac, and bladder. Mature ovaries are white and almond shaped and usually measure about 2 × 3 × 3 cm. Benign functional cysts have a characteristic appearance and may be left alone because they will resolve spontaneously, whereas other cysts are neoplastic and may require oophorectomy. If an ovarian malignancy is suspected, peritoneal washings are generally required. The fallopian tubes may be red and inflamed from salpingitis or an early ectopic pregnancy. During the first 2 to 6 weeks of an ectopic pregnancy, the tubes are dilated to 3 to 4 cm and may later rupture, causing life-threatening hemorrhage. Uterine leiomyomas are usually gray, firm masses that may be microscopic in size or fill the abdominal cavity. If the ovary, fallopian tube, or uterus is abnormal at laparoscopy, we generally request the presence of a gynecologist to assist in diagnosis. Whenever gynecologic disorders are suspected as the primary problem by history and physical examination, diagnostic laparoscopy is preferably performed by a gynecologist.

The pelvic floor should be inspected for inguinal or femoral hernias as well as tumors of the bladder and colon. Bowel or bladder attached to the pelvic floor may represent an incarcerated hernia, adhesions, or tumor extension. The sigmoid colon may reveal diverticular disease, abscess, or tumor. Typically, diverticula are herniations 1 mm to several centimeters in diameter and are located between the mesenteric and antimesenteric teniae of the colon.

Midabdomen

The anterior surfaces of the intestines, omentum, and stomach are examined for abnormalities. Following the teniae of the cecum proximally will locate the appendix. In the early stages of inflammation, the appendix is erythematous with engorged surface vessels or covered by a fibrinopurulent material; later, green-black foci of necrosis typify gangrenous appendicitis and herald impending perforation. Crohn's disease generally occurs in sharply demarcated segments of bowel that have a rubber-hose consistency surrounded by creeping fat. The duodenum may have a sealed patch of omentum covering a perforated ulcer, which can be missed altogether by cursory examination. If portal hypertension is present, the veins of the omentum and abdominal wall will be dilated. The intestine may be ischemic and appear blue because of volvulus or vascular infarction. With intestinal obstruction, the proximal bowel is generally distended and the bowel distal to the obstructive lesion decompressed, especially with a complete obstruction secondary to an adhesive band from a previous operation.

Right Upper Quadrant

To examine the right upper abdomen, the patient is placed in a reverse Trendelenburg position of 30 to 40 degrees while the table is rotated to the patient's left by 15 to 20 degrees. This maneuver allows the colon and duodenum to fall away from the liver's edge. Normal liver is reddish-brown and has a smooth surface. The falciform ligament and both lobes of the liver are closely examined for pathology. Adhesions on the anterior liver surface may be due to inflammation from Fitz-Hugh-Curtis syndrome. Primary carcinoma of the liver may appear as small nodular lesions with widespread peritoneal metastases. Fatty degeneration imparts a yellow color to the liver, whereas nodularity of the liver suggests cirrhosis. Extrahepatic biliary obstruction from periampullary cancer may cause cholestasis, and the liver will generally be tense and appear green hued owing to bile discoloration. Most metastatic liver lesions appear as yellow, gray, or white nodules that feel solid. Metastatic melanoma appears as brownish-black spots. Solid masses should be examined by biopsy using either a cutting needle, cupped forceps, or cautery scissors, or such masses may be sampled by needle aspiration to determine histology and cytology. Bluish cystic hepatic lesions, however, should never be examined by biopsy because hemangiomas can cause severe bleeding, which may be difficult to control laparoscopically. Hepatic, pancreatic, and other suspicious lesions may be further evaluated by laparoscopic ultrasonography.

Usually, the gallbladder can be seen protruding beyond the inferior edge of the liver, but sometimes, it is not visible without carefully elevating the liver or taking down adhesions. The normal gallbladder has a shiny bluish-green color. In acute cholecystitis, the gallbladder may be tense, edematous, fiery red, and covered with a fibrinosuppurative exudate. Extensive adhesions surrounding a thickened, gray-white, tough gallbladder wall are usually due to chronic inflammation. Distal extrahepatic obstruction may cause a distended (Courvoisier) gallbladder. Transcholecystic cholangiography or laparoscopic ultrasono- graphy [34] may be used to diagnose stones in the gallbladder or bile ducts. A pale or opaque gallbladder may represent chronic cholecystis or tumor.

Left Upper Quadrant

The normal spleen is usually not seen in the left upper quadrant; when visible, splenomegaly is often apparent. Further inspection of the spleen is best achieved by rotating the patient to the right in the head-up position. Although splenic biopsy has been performed, the risk for severe hemorrhage usually warrants caution. The left hemidiaphragm can be inspected for hiatal hernia and esophageal varices. If the diaphragm bows inward, a pneumothorax should be suspected.

INDICATIONS FOR DIAGNOSTIC LAPAROSCOPY

Diagnostic laparoscopy may be used to optimize the workup of abdominal pain and masses, triage trauma, and stage malignancy. Some of the current indications for diagnostic laparoscopy are listed in Table 161-2.

Accepted
Assessment of liver
Acute abdominal pain
Ascites of unknown etiology
Gaining Acceptance
Staging of malignancy
Blunt and penetrating trauma
Chronic abdominal pain
Anecdotal Experience
Intensive care unit setting
Fever of unknown origin
Second-look procedures

TABLE 161-2 Common Indications for Diagnostic Laparoscopy

Evaluation of Abdominal Pain

Acute Pain Acute right lower quadrant pain that may be due to acute appendicitis is ideally evaluated by laparoscopy. Traditionally, appendicitis was a clinical diagnosis, for which a 20% error rate with a normal appendix was considered acceptable because of the morbidity and mortality caused by appendiceal perforation from a delay in operation. With the use of laparoscopy, several studies have shown a markedly decreased rate of negative laparotomy results for questionable appendicitis. [35] Especially in young women, who experience gynecologic problems that may mimic appendicitis, diagnostic laparoscopy has reduced the unnecessary laparotomy rate by one third. [36], [37], [38] and [39] In a prospective randomized trial of 100 patients with the clinical diagnosis of appendicitis, 50 underwent open laparotomy, and 50 underwent diagnostic laparoscopy. In the latter group, 19 patients (38%) did not require appendectomy; moreover, there were no complications from laparoscopy. [40] When appendicitis is present, the appendix may be removed laparoscopically. If the appendix is perforated, laparoscopy allows thorough irrigation of the abdominal cavity. Several randomized prospective studies have suggested that laparoscopic appendectomy decreases postoperative pain and accelerates return to full activity compared with conventional appendectomy. [40], [41], [42] and [43] Other causes of right lower quadrant pain also may be better evaluated by a laparoscope. Laparoscopic treatment of tubal pregnancy is as safe as treatment with laparotomy and has the benefit of decreased length of hospital stay, lower cost, and earlier return to full activity. [44], [45] Laparoscopy has also been suggested as the definitive diagnostic modality for "pelvic inflammatory disease" because no reliable clinical methods exist that accurately establish such a diagnosis. [46] Children, newborn to 15 years of age, with ill-defined abdominal pain have been evaluated by laparoscopy and found to have problems including appendicitis, adhesions, cysts, Meckel diverticula, or unappreciated hernias. [47] Many of these problems can be treated laparoscopically. In the intensive care unit setting, diagnostic laparoscopy has been used to assess the critically ill patients with suspected abdominal catastrophe. In a study of 25 such patients, 13 (52%) laparoscopies yielded negative findings, and nontherapeutic operations were avoided. [48]
Chronic Pain The cause of chronic abdominal pain may be unclear even after extensive preoperative diagnostic evaluation. Diagnostic lap- aroscopy yielded positive findings in 47% of chronic abdominal pain cases. [49] Laparoscopy has the advantage of adding therapeutic possibilities to its diagnostic value; for example, when adhesions are detected, laparoscopic adhesiolysis may relieve the patient's symptoms.

Trauma

Laparoscopy may have a role in evaluating blunt and penetrating abdominal trauma. After trauma, a surgical dictum has been to explore all penetrating wounds of the abdomen. This practice results in many negative laparotomies when operation is based on clinical impression alone. Hemodynamically stable patients with significant abdominal trauma are traditionally evaluated by peritoneal lavage or CT scan. With a 5-mm laparoscope inserted under local anesthesia in the emergency room, in 150 blunt abdominal trauma cases, diagnostic laparoscopy predicted or excluded the need for laparotomy in more than 90% of patients. [50] Sosa and colleagues [51] used laparoscopy in six patients with tangential gunshot wounds to the abdomen to exclude fascial penetration and avoid laparotomy. Diagnostic laparoscopy is also efficacious as an adjunct in patient selection after CT scan has demonstrated solid organ injury; in 15 patients with splenic and liver lacerations, 8 (53%) were managed conservatively without laparotomy. [52] Diagnostic laparoscopy in 33 stable patients with penetrating trauma and no clinical evidence of intraperitoneal injury identified peritoneal penetration in 10 patients, of whom 9 had intraperitoneal injury at laparotomy; 23 patients were spared a negative exploratory laparotomy. [53]

Laparoscopy in 182 hemodynamically stable patients after abdominal trauma averted laparotomy in 44% after penetrating trauma and in 47% after blunt trauma. [54]

Diagnostic laparoscopy for trauma remains controversial despite its potential to avoid negative laparotomy because retroperitoneal wounds and hollow-organ perforations may not be recognized and go untreated. Diagnostic laparoscopy was better than peritoneal lavage in predicting the need for laparotomy for stab wounds but not after blunt trauma in a prospective study of 75 patients. [55] After penetrating abdominal trauma in a series of 100 patients, diagnostic laparoscopy was accurate in assessing for hemoperitoneum, solid-organ injuries, diaphragmatic lacerations, and retroperitoneal hematomas. For injuries to the hollow viscera, laparoscopy was 100% specific but only 18% sensitive. Injuries to the flank and epigastric region were frequently missed, thereby limiting its usefulness. [56] Laparoscopic findings in 32 patients before laparotomy correlated (97%) with the need for laparotomy, but injuries to the liver, pancreas, stomach, duodenum, small bowel, intestinal mesentery, ureter, and urinary bladder were missed by laparoscopy. [57] In the future, perhaps a combined approach using laparoscopy to examine the peritoneum and CT scan to image the retroperitoneum may increase diagnostic accuracy after abdominal trauma.

Liver Lesions and Ascites

Inspection of the liver may be the most common indication for diagnostic laparoscopy. Blind percutaneous biopsy of the liver may cause inapparent injury and will miss focal disease such as small metastases, whereas laparoscopy improves the accuracy of biopsy. [32] In addition, laparoscopic criteria may be predictive of outcome. In cirrhosis, although the interpretation of laparoscopic findings is largely subjective, the size and pattern of regenerative nodules, formation of lymphatic vesicles, and presence of an enlarged spleen all predict a poor prognosis. [58] Laparoscopy is indicated in the evaluation of ascites after transabdominal ultrasonography, CT scan, and paracentesis fail to determine its etiology. [59] Because of the 12- to 15-fold magnification and excellent ability to view the entire peritoneal surface, laparoscopy is particularly sensitive for small peritoneal implants. If the size of the miliary nodules is uniform, the nodules are more likely to be benign because malignant nodules usually are of varying sizes. [60] Biopsy for histological confirmation is essential to the diagnosis, and ascitic fluid should be sent routinely for cytology and microbiology. In a study of 129 patients with ascites of unknown origin, laparoscopy in combination with biopsy established the etiology of ascites in 86%. [61]

Staging of Malignancy

When performing laparoscopy to stage abdominal malignancy, preoperative consideration should be given to the accessibility to specific viscera, tissue samples required for adequate staging, and range of palliative interventions possible endoscopically and laparoscopically if the lesion proves unsuitable for curative operation. After initial port placement and primary visual survey of the abdomen, accessory ports are placed under direct vision in appropriate locations. Of 25 patients referred for evaluation, laparoscopy was 100% accurate in the diagnosis or exclusion of various intra-abdominal malignant neoplasms. [62]

Liver Cancer

Of patients thought to have resectable disease by preoperative transabdominal ultrasonography or CT scan, 40% to 70% of patients with primary or metastatic hepatic malignancy are found to have unresectable disease at operation. [63] Therefore, diagnostic laparoscopy before laparotomy is recommended to evaluate the extent of cancer and degree of cirrhosis.

Laparoscopy accurately diagnoses metastatic tumors to the liver. Lightdale and colleagues, [64] in a study of 65 metastatic lesions, obtained tissue samples in 60. Histology was 92% sensitive and 100% specific. However, 40% to 50% of hepatomas less than 3 cm in diameter are not visible on the surface of the liver. For these small tumors, laparoscopic ultrasonography improves the sensitivity to 88%, whereas for tumors larger than 3 cm, the sensitivity by laparoscopic ultrasonography is 100%. [26] Small hepatic cysts may be confused with metastases by surface imaging, whereas cysts usually appear as round, echo-free lesions with posterior enhancement by laparoscopic ultrasonography. [28]

Ovarian Cancer

Diagnostic laparoscopy has been used extensively to evaluate gynecologic malignancy. [65], [66] Laparoscopic evaluation of 819 adnexal masses had a sensitivity for the detection of malignancy of 100%, specificity of 97%, and negative predictive value of 100%. [66] However, the positive predictive value was only 41% because 27 benign tumors were incorrectly thought to be malignant. Ovarian cancer metastatic to the diaphragm is best identified by laparoscopy, [67], [68] and in one study, 11 of 14 patients were upstaged by diagnostic laparoscopy. [67]

Gastric Cancer

With cancer of the stomach, laparoscopic examination can prevent 40% to 50% of unnecessary laparotomies. [27], [69], [70] Laparoscopy determines serosal infiltration, tumor fixation, and metastases to the liver and peritoneum. Laparoscopy in 40 patients with gastric carcinoma detected distant metastases in 5 patients and locally advanced unresectable cancer in 11 patients (27%). [69] The overall diagnostic accuracy was 92%. Similarly, laparoscopy in 360 patients with gastric cancer was 89% accurate in detecting peritoneal spread and 97% accurate in identifying liver metastases. [70] Intraoperative ultrasonography is complementary, identifying small lymph nodes, gastric wall involvement, and deep hepatic metastases. [25], [27]

Gallbladder Cancer

If gallbladder cancer is suspected, diagnostic laparoscopy can confirm local or distant spread, precluding resection in 85% of patients. [71], [72] Fortunately, gallbladder cancer is rare. In the author's personal series of more than 1700 laparoscopic cholecystectomies, only 3 patients were initially diagnosed intraoperatively with cancer of the gallbladder, none of whom were resectable for cure.

Cancer of the Pancreas

Diagnostic laparoscopy was recognized as early as 1911 by Bernheim [73] as a valuable procedure to exclude pancreatic cancer metastases; however, Meyer-Burg [74] reported the first series using diagnostic laparoscopy for the staging of cancer of the pancreas. Since then, several different approaches have been proposed. Cuschieri and associates [75] examined the pancreas through an infragastric opening into the lesser sac and was able to see the entire gland in 60% of patients. Ishida, [76] using a supragastric approach, visualized only 32% of tumors in the head of the pancreas but accurately identified 85% of cancers in the body of the pancreas. Diagnostic laparoscopy is particularly helpful in patients who have no evidence of metastatic disease on preoperative imaging studies. In these patients, staging laparoscopy has identified metastases in more than 40% of cases. [73], [77] More recently, laparoscopic contact ultrasonography has enhanced the ability to stage cancer of the pancreas laparo- scopically. John and colleagues [29] reported that of 40 patients with suspected carcinoma of the head of the pancreas considered preoperatively to have resectable disease, 35% had advanced disease at laparoscopy. The addition of intraoperative laparoscopic ultrasonography identified another 10 patients (25%) with unresectable cancer. In addition to hepatic metastases, ultrasonography reveals lymphadenopathy, local tumor infiltration, portal vein or mesenteric arterial invasion, and anomalous anatomy.

At our institution, staging laparoscopy with laparoscopic ultrasonography is routinely used to stage cancer of the liver, biliary tree, and pancreas before laparotomy. We have shown that more than 40% of patients with tumors thought to be resectable by conventional imaging tests are proved unresectable using these studies. [2] Patients unresectable at laparoscopy are considered for laparoscopic biliary bypass or gastrojejunostomy, maintaining the goal of minimal invasion. Patients without evidence of metastases or vascular invasion by tumor are converted to a laparotomy for curative resection.

Lymphoma

Laparoscopy completely stages lymphoma by liver biopsy, nodal sampling, and complete visualization of the abdominal cavity. [78] During diagnostic laparoscopy, Salky [49] obtained adequate tissue samples in 12 of 12 patients after percutaneous biopsy was unsuccessful. Childers and Surwit [79] have reported laparoscopic paraaortic lymph node biopsy for non-Hodgkin lymphoma. Whether laparoscopic splenectomy is necessary for full staging remains controversial.

Esophageal Cancer

Diagnostic laparoscopy has been advocated for staging esophageal cancer in those patients considered suitable for resection by CT scan. Laparoscopy before laparotomy identified 14% of patients with metastases to the liver, peritoneum, omentum, and lymph nodes, precluding resection. [80]

Second-Look Procedures

After initial treatment of abdominal malignancy, assessment of response by second look may be better done laparoscopically than by a laparotomy. If early tumor recurrence is suggested by history, physical examination, or rising tumor markers, but imaging studies are negative, laparoscopy may be indicated. Imaging studies may be equivocal because of scar formation, altered anatomy from a previous operation, or reactive lymphadenopathy. Although abdominal exploration by laparoscopy also may be limited by adhesions, the finding of histologically confirmed distant metastases or peritoneal carcinomatosis may obviate further evaluation. In the future, radionuclide probes that detect tumors may improve the accuracy of diagnostic laparoscopy for this purpose.

Absolute and Relative Contraindications

Preoperative evaluation should determine the presence of confounding medical problems that may adversely affect the outcome of laparoscopy. Absolute contraindications to laparoscopy include the inability to tolerate general anesthesia or lap- arotomy, uncorrectable coagulopathy, a “frozen” abdomen (obliteration of the celomic cavity, usually caused by multiple prior operations), or a hemodynamically unstable patient (Table 161-3). Relative contraindications are as dependent on the experience of the laparoscopist as on any particular attribute of the patient.

Absolute
Poor risk for general anesthesia
Inability to tolerate a laparotomy
Uncorrected coagulopathy
"Frozen" abdomen
Hemodynamic instability
Relative
Prior abdominal surgery
Peritonitis; previous or acute
Obesity
Pregnancy
Unreducible abdominal/inguinal hernia
Severe pulmonary disease
Intestinal obstruction

TABLE 161-3 Contraindications to Laparoscopy

With experience, most of these conditions can be successfully managed. For example, morbidly obese patients may require longer trocars to traverse the anterior abdominal wall and higher insufflation pressures to obtain an adequate working space. Appendicitis and cholecystitis may occur during pregnancy, and laparoscopy is often helpful for diagnosing and treating this subset of patients with abdominal pain in whom additional radiologic studies are undesirable. Although laparoscopic cholecystectomies have been performed safely during pregnancy, [81] the prolonged exposure to CO_2 pneumoperitoneum may have untoward effects on the fetus. Hyperventilation and monitoring of end-tidal CO_2 minimize maternal hypercarbia and fetal acidosis. Insufflation pressures are kept low, preferably below 12 mm Hg, to obviate respiratory problems, and monitoring of fetal heart sounds should be done in consultation with an obstetrician. [82], [83]

Conclusions

Diagnostic laparoscopy has rapidly been embraced by clinicians around the world. With minimal morbidity, lesions too small to be seen by CT scan, MRI, and abdominal ultrasonography can be accurately described and examined by biopsy using laparoscopic techniques. Malignancy can be staged, and vague abdominal complaints may be elucidated. Certainly, many unnecessary and morbid laparotomies will be avoided. For these reasons, diagnostic laparoscopy is being incorporated into modern diagnostic and therapeutic algorithms for more and more disease processes. Occasionally, anatomic or physiological considerations will preclude the laparoscopic approach, and conversion to an open operation reflects sound surgical judgment and should not be considered a complication. The exact role for diagnostic laparoscopy in the elective and emergent situation is still evolving as laparoscopists become more technically facile and as newer “competing” technology improves.

THERAPEUTIC LAPAROSCOPY

Laparoscopic Cholecystectomy

Laparoscopic cholecystectomy has rapidly become the gold standard for removing the gallbladder. [12] In addition to reports from multiple large clinical series, several randomized studies have clearly demonstrated the benefits of laparoscopic cholecystectomy compared with open cholecystectomy. [83], [84] Laparoscopic cholecystectomy is indicated for symptomatic cholelithiasis, biliary dyskinesia (acalculous cholecystitis), and select patients with asymptomatic gallstones (e.g., immunosuppressed status, *Salmonella typhi* carriers, porcelain gallbladder [85], [86]). At our institution, more than 95% of elective cholecystectomies are performed laparoscopically. Although the operation was originally reserved for young thin patients, today elderly [87] and obese [88] patients also benefit from surgery through small incisions.

Several variations of the operation have been proposed; we favor the “American” technique. The 5- or 10-mm viewing port is placed at the umbilicus. Two 5-mm working ports are right subcostal, and one working port is epigastric. With the gallbladder retracted over the liver by the assistant, the gallbladder infundibulum and porta hepatis become accessible to the operating surgeon (Fig. 161-4). Antegrade dissection beginning on the gallbladder facilitates isolation of the cystic artery and cystic duct within the hepatocystic triangle (bounded by the gallbladder, liver, and cystic duct). This critical view must be clearly demonstrated before clipping or dividing any structure to avoid inadvertent injury to the common bile duct or right hepatic artery [89] (Fig. 161-5).

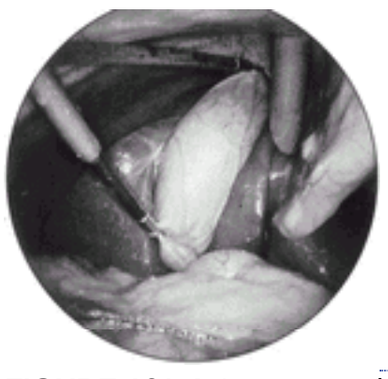

FIGURE 161-4. Laparoscopic view of the gallbladder during cholecystectomy.

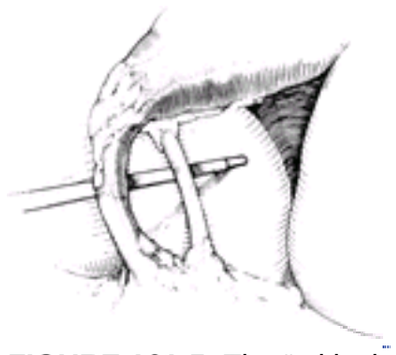

FIGURE 161-5. The “critical view” of safety during laparoscopic cholecystectomy after dissection of the neck of the gallbladder away from its bed. The cystic duct is seen on the left, and the cystic artery is on the right. This dissection should prevent bile duct injuries during laparoscopic cholecystectomy. (From ref. [89].)

Before dividing the cystic duct, a clip is placed across the junction of the cystic duct and gallbladder, and a small ductotomy is made distal to the clip. An atraumatic dissecting forceps is then used to palpate the cystic duct for stones and to milk potential stones out of the cystic duct. Cystic duct stones should be sought in all patients. Their presence predicts choledocholithiasis [90] and could lead to retained stones or the postcholecystectomy syndrome if left untreated.

Intraoperative cholangiography should be considered whenever there is a suspicion of choledocholithiasis (Fig. 161-6) or bile duct injury or when the anatomy is unclear intraoperatively. Fluoroscopic cholangiography may be performed more rapidly than static cholangiography. [90], [91] Using fluoroscopy, the surgeon can interact with findings (stones, air bubbles, aberrant anatomy) in real time. Although selective application of cholangiography will detect more than 90% of common bile duct stones, advocates for routine cholangiography argue that the best preoperative indicators of common bile duct stones are not adequately sensitive to detect all stones. [91] In the era of cost containment, however, a policy of routine cholangiography is difficult to justify. In the future, new technologies such as laparoscopic ultrasonography may delineate biliary anatomy and discover common bile duct stones less invasively and expensively.

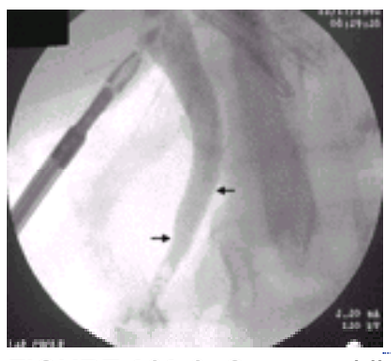

FIGURE 161-6. Common bile duct stones identified by intraoperative digital fluorocholangiography during laparoscopic cholecystectomy. The stones can be viewed as filling defects in the contrast within the distal common bile duct (*arrows*).

After the critical view is clearly demonstrated and the common bile duct is cleared, the cystic duct and artery are clipped and divided. As dissection is continued, tension is maintained on the gallbladder wall to help define the plane between the gallbladder and liver bed. Electrocautery is usually used to dissect the gallbladder from the hepatic fossa. The gallbladder is removed through the umbilical port incision.

Laparoscopic cholecystectomy decreases postoperative pain, shortens hospital stay from 1 week to less than 24 hours, and returns the patient to full activity within 1 week compared with 1 month after open cholecystectomy. [12], [83], [84] In addition to lower morbidity and mortality, laparoscopic cholecystectomy also may be less expensive than open cholecystectomy. [92]

During the learning curve for laparoscopic cholecystectomy, however, bile duct injuries increased to about 0.5% or greater; one state-wide survey estimated the frequency of this potentially devastating complication to be seven or eight times higher than with open cholecystectomy. [93] Certain anatomic situations also have been found to predispose to bile duct injury: with absent or extremely short cystic ducts, the common bile duct may be mistaken for the cystic duct; clips of insufficient length may not occlude the dilated cystic duct and lead to a cystic duct stump leak; and aberrant origin of the right hepatic duct may not be recognized and result in biliary injury. Insult is added to injury whenever ductal injuries are not determined until days later; a straightforward repair is converted into a very complex repair of a high-grade injury. [89] With incomplete dissection, the right hepatic artery also may be mistaken for the cystic artery, and the divided right hepatic artery may cause hepatic or bile duct ischemia. Over time, the technical complications of Laparoscopic cholecystectomy, especially bile duct injuries, seem to have decreased. [94] When injuries occur, the gastroenterologist plays an important role in diagnostic evaluation and patient management. [90], [95]

Laparoscopic Appendectomy

Gynecologist K. Semm was the first to perform laparoscopic appendectomy in 1981. [96] A decade later, Attwood and colleagues [40] reported a randomized prospective study from the United Kingdom showing laparoscopic appendectomy to decrease pain, expedite hospital discharge, and cause fewer infections compared with open appendectomy. Other theoretical advantages include fewer postoperative adhesions and a lower frequency of subsequent intestinal obstruction. [97] When compared with the view afforded by a small right lower quadrant incision, a more thorough abdominal exploration is also possible laparoscopically. In women, for example, the adnexae are inspected, and many gynecologic problems that may account for pain in the right lower quadrant can be excluded. During laparoscopic appendectomy, the appendiceal stump may be isolated and closed with a preformed loop ligature or alternatively ligated and divided with a stapler. The appendix is usually placed in a plastic pouch to prevent contamination before it is removed through a port. Despite the shorter hospital stay, laparoscopic appendectomy appears to offer no overall medical cost savings. [98] Several prospective randomized trials of laparoscopic versus open appendectomy have been performed, with varying results. [40], [41], [42] and [43] Because of these discrepancies and because many appendectomies are performed at times of day when operating room staffing may not include individuals who are comfortable handling laparoscopic equipment, laparoscopic appendectomy has not been embraced for routine use.

Advanced Procedures

Advanced laparoscopic procedures rely on more complex techniques, such as using variable endoscopic viewpoints and suturing. These operations have not been used as frequently as the “basic” procedures mentioned earlier, and additional clinical experience is needed to clarify their role.

Laparoscopic Management of Choledocholithiasis

The optimal treatment of common bile duct stones is controversial. Although many methods for treating choledocholithiasis have been reported, the appropriate therapy depends on the patient’s condition and the relative local expertise in laparoscopy, endoscopy, and interventional radiology. [99] Management strategies depend not only on the patient’s medical condition but also on whether the stones are detected before, during, or after cholecystectomy. The algorithm we propose assumes that the surgeon has advanced laparoscopic biliary experience and excellent endoscopic and radiologic support (Fig. 161-7). The common goal of therapy must be to achieve ductal clearance with the fewest number of interventions, lowest cost, and least morbidity [91], [100], [101], [102], [103], [104], [105], [106], [107], [108], [109] and [110] (Table 161-4).

FIGURE 161-7. Algorithm for the management of common bile duct stones. (From ref. [100].)

METHOD	SUCCESS (%)	POSTOPERATIVE HOSPITALIZATION (days)	RTW (days)
Transcystic duct extraction	80–95	1–2	7–10
Laparoscopic CBDE	85–100	4–7	4–30
Open CBDE	90–100	5–10	20–42
ERC/ES	85–95	0–3	1–10

CBDE, common bile duct exploration; ERC, endoscopic retrograde cholangiography; ES, endoscopic sphincterotomy; RTW, return to work.

TABLE 161-4 Results of Therapy for Choledocholithiasis [91], [99], [100], [101], [102], [103], [104], [105], [106], [107], [108], [109] and [110]

Preoperative endoscopic retrograde cholangiography (ERC) with sphincterotomy and stone extraction should be performed in those patients with deep jaundice, cholangitis, severe pancreatitis, or an indication for biliary drainage. [106], [107], [108] and [109] In patients with frank jaundice and a dilated common bile duct by ultrasonography, ERC is performed to exclude malignancy or other conditions requiring therapy. If patients cannot be successfully managed endoscopically, transhepatic biliary decompression or operative therapy may become necessary. After stone extraction for acute gallstone pancreatitis, laparoscopic cholecystectomy is usually performed during the index admission. [111]

During laparoscopic cholecystectomy, 5% to 10% of patients are found to have unsuspected common bile duct stones. [99], [101], [102] and [103] When discovered by intraoperative cholangiography or ultrasonography, most stones in the common bile duct may be removed laparoscopically. Small stones can be cleared from the common bile duct by advancing the cholangiocatheter and irrigating forcefully with saline solution after relaxing the ampullary sphincter with 1 to 2 mg of intravenous glucagon. If irrigation is unsuccessful, baskets with soft, radiopaque, filiform tips may be passed under fluoroscopic guidance and used to extract stones. [101] If these attempts fail to retrieve the stones, the guidewire should be left in place to facilitate insertion of the choledochoscope.

Transcystic choledochoscopy directly visualizes the biliary tract and therefore is highly effective at clearing the common bile duct of stones [102], [103] and [104] (Fig. 161-8). A 7- to 10-French choledochoscope may be passed over the guidewire into the common bile duct after cystic duct dilation using balloon dilators. The endoscope is equipped with a biopsy channel for introducing a 3-French stone basket. Under visual guidance, the basket is advanced beyond the stone and opened. As the basket is pulled backward and rotated, the stone is ensnared. The stone and scope are removed under direct vision through the ductotomy. Multiple passes are made until the duct is clear. After stone removal, completion cholangiogram should be performed to demonstrate conclusively the clearance of the duct at the end of the procedure. [112] Ideal candidates for transcystic extraction are patients with relatively few, small (less than 8 mm) stones located in the distal common duct.

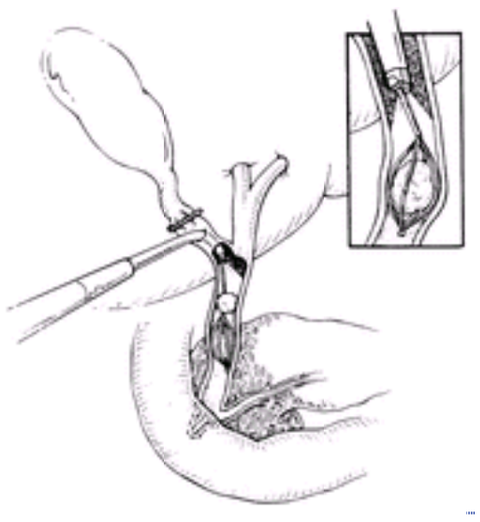

FIGURE 161-8. A flexible choledochoscope is passed through the cystic duct, and a basket is used to extract the stone from the common bile duct (From ref. [100].)

Larger or impacted stones and those located in the proximal bile ducts may require a choledochotomy and placement of a T tube. Laparoscopic choledochotomy has been performed safely and effectively in experienced hands with shorter hospitalization and disability when compared historically with open common bile duct exploration. [104], [105] If choledochotomy is reserved for failures of the transcystic technique, laparoscopic choledochotomy is needed only infrequently.

The major disadvantages of laparoscopic common bile duct exploration are that it is technically difficult, is time consuming, and requires special equipment. Because many surgeons have not yet acquired these skills, they either convert to open laparotomy or rely on ERC with sphincterotomy and stone extraction. [113] Open choledochotomy may be performed to clear the common duct in a single operation if the surgeon lacks the expertise needed to manipulate the choledochoscope and suture the choledochotomy closed.

Stones intentionally left for postoperative endoscopic clearance should be small and considered unlikely to obstruct the bile duct completely. Relying on postoperative ERC subjects the patient to at least two procedures with their additive morbidity, and possibly a second operation if ERC and stone extraction fail to clear the duct. Even in experienced hands, stone extraction has a 5% to 10% chance of failure. [106] Relative contraindications to leaving ductal stones to be managed by postoperative ERC and stone extraction include a distal common duct stricture, duodenal diverticulum, previous Billroth II reconstruction, and stones greater than 1 cm in size, and these patients may be best managed at the time of surgery. The surgeon also can leave a cholangiocatheter in the common bile duct to facilitate subsequent ERC and stone extraction. [114] Intraoperative ERC also may be considered, but this approach is difficult because of patient positioning, personnel coordination, and the tendency toward intestinal distention by endoscopic insufflation. Postoperative ERC with stone extraction is the procedure of choice for retained stones in the absence of a T tube; however, it is unwise to rely on postcholecystectomy ERC and stone extraction at centers with less than a 90% endoscopic duct clearance rate.

Laparoscopic Management of Gastroesophageal Reflux Disease

A recent randomized prospective study showed that conventional open Nissen fundoplication resulted in more effective treatment of complicated gastroesophageal reflux disease (GERD) than did medical management with histamine H_2 receptor antagonists. [115] However, few patients were treated operatively owing to the perceived morbidity of surgery until the first report of laparoscopic Nissen fundoplication was published in 1991. [116] The preoperative evaluation may include radiographic imaging, endoscopy, pH monitoring, and esophageal manometry. [117] A markedly shortened esophagus is a contraindication to standard Nissen repair and necessitates an esophageal lengthening procedure. If the propulsive force of the esophagus is diminished, a partial, rather than a total, wrap (200- to 270-degree

partial fundoplication) is preferred by many surgeons. [118], [119]

Laparoscopic Nissen fundoplication is usually performed through five ports with the patient in the reverse Trendelenburg (head-up) position. The laparoscope is placed through a port in the left midrectus 12 to 15 cm inferior to the xiphoid. The remaining ports are typically placed in an arc across the upper abdomen. This port arrangement allows access to the hiatus and permits comfortable suturing.

The Nissen fundoplication is performed by wrapping the gastric fundus 360 degrees around the distal esophagus. [120] A retractor placed through the right subcostal port elevates the left lobe of the liver, and a Babcock forceps, through the right midrectus port, pulls the stomach and epiphrenic fat pad inferiorly. After dividing the phrenoesophageal ligament, both crura and the anterior vagus nerve may be clearly identified. If a hiatal hernia is present, it is reduced into the abdominal cavity with gentle traction after cutting all adhesions to the hernia sac. By retracting the right crus laterally, the right side of the esophagus is dissected to expose the aortoesophageal groove and posterior vagus nerve. Similarly, the left crus is dissected from the esophagus and fundus to its point of origin from the right crural leaflet.

The fundus is fully mobilized by dividing the proximal gastrosplenic ligament (Fig. 161-9). Dissection begins about 8 to 10 cm distal to the esophageal junction on the greater curve. The short gastric vessels are placed on traction, and a window is created into the lesser sac. The short gastric vessels are then serially divided by clipping and cutting them or by serial application of ultrasonic coagulating shears. Division of the short gastric vessels may diminish early postoperative dysphagia. [121] All posterior retroperitoneal adhesions to the fundus are divided to mobilize the proximal stomach completely.

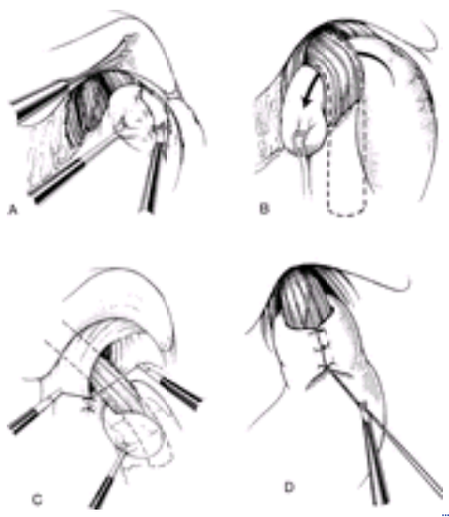

FIGURE 161-9. Laparoscopic Nissen fundoplication. **A:** Mobilization of fundus and division of short gastric vessels. **B:** Fundus of stomach is wrapped posterior to the esophagus around a 60-French bougie. **C:** The hiatus may be closed posteriorly. **D:** Appearance of completed fundoplication as viewed by the laparoscope. (From ref. [120].)

The fundus near the insertion of the short gastric vessels is pulled from left to right posterior to the esophagus (see Fig. 161-9). After serially dilating the esophagus to a 50- to 60-French Maloney bougie, the wrap can be formed without excessively narrowing the esophagus. The hiatus should be closed in most patients because of the hiatal dissection. Several sutures are placed to reapproximate the right and left crura.

The goal is a “short, floppy” Nissen fundoplication. Generous seromuscular bites of fundus to the left of the esophagus, the anterior esophageal wall away from the anterior vagus nerve, and the fundus to the right are all incorporated in sutures securing the 360-degree fundoplication. The esophageal wall is incorporated to prevent slippage of the wrap around the body of the stomach or into the thoracic cavity. The wrap should be only 1.5 to 2 cm in length because longer wraps may cause a higher incidence of postoperative dysphagia. [122]

Laparoscopic Nissen fundoplication has demonstrated excellent results in several large clinical series. [118], [119], [123], [124] The patient is usually discharged within 2 days, and more than 90% of patients report complete relief of symptoms up to 5 years after surgery. Long-term outcome data will be necessary to demonstrate the effectiveness of laparoscopic antireflux operations.

Laparoscopic and Thoracoscopic Esophagocardiomyotomy for Achalasia

Recent application of minimally invasive techniques to myotomy have stimulated a reemergence of esophageal myotomy in the treatment of this disease. Despite a previously published prospective, randomized trial demonstrating that esophageal myotomy yielded better results than balloon dilation, [125] the morbidity attendant on the necessary thoracotomy or laparotomy dampened enthusiasm for the operation. The incisional trauma related to the operation has been substantially decreased by minimally invasive procedures. The first technique reported was a thoracoscopic myotomy without an antireflux procedure. [126] This operation is performed using four or five trocars arranged in a diamond-shaped pattern around the lower portion of the left chest. A long myotomy can be performed using the thoracoscopic approach. Short-term postoperative results have been excellent, although 24-hour pH monitoring suggests that abnormal gastroesophageal reflux occurs in 60% of these patients postoperatively. [126]

Laparoscopic esophagomyotomy has been described more recently, subsequent to the extensive experience with the laparoscopic approach to the esophageal hiatus. Using the laparoscopic approach, a partial fundoplication can be added to help prevent postoperative gastroesophageal reflux. [127], [128] This may take the form of an anterior 180-degree (Dor) fundoplication [128] or a posterior 180-degree (Toupet) fundoplication. [127] A 360-degree Nissen fundoplication should not be added to the esophagomyotomy because it would likely cause postoperative dysphagia.

All laparoscopic approaches facilitate extending the myotomy onto the stomach and thus may prevent recurrence of postoperative dysphagia. [127], [128] However, it is more difficult to extend the myotomy as high cephalad as can be done using the thoracoscopic approach. Thus, the laparoscopic approach should probably not be used in the rare patients who require proximal extension of the myotomy to the midesophagus. As is typical of minimally invasive surgery, the hospital stay and recovery time are shortened by at least 50% compared with the standard open myotomies. Long-term follow-up results of the minimally invasive procedures are not yet available, but the initial outcomes are sufficiently promising to recommend this form of treatment by experienced surgeons in uncomplicated cases.

Laparoscopic Inguinal Herniorrhaphy

The initial laparoscopic inguinal herniorrhaphies simply plugged the fascial defect of the groin floor with a wad of prosthetic mesh or directly closed the defect under tension. Consequently, these approaches had higher recurrence rates than standard methods and are no longer performed. Many surgeons then adapted laparoscopic techniques to perform a tension-free Stoppa repair using prosthetic material. [129] Using a transabdominal approach, the peritoneal cavity is entered using three trocars placed at the level of the umbilicus. The peritoneum overlying the hernia is incised, and the inguinal floor defect is covered with mesh that is secured with staples to the Cooper ligament, the iliopubic tract, and the transverse arch (Fig. 161-10). The peritoneum is then closed to diminish formation of adhesions between the mesh and bowel. By resurfacing the inguinal floor with prosthetic mesh, good preliminary results were achieved. Large reports with short-term follow-up have shown an acceptable recurrence rate and an earlier return to full physical activity with this technique. [130], [131] Several randomized prospective studies comparing early results of open and laparoscopic herniorrhaphy confirmed these proposed advantages. [132], [133], [134] and [135] The major disadvantage of this approach compared with an inguinal incision is that the laparoscopic repair is more costly and nearly always requires general anesthesia, adding to the risks of the procedure and limiting its indications.

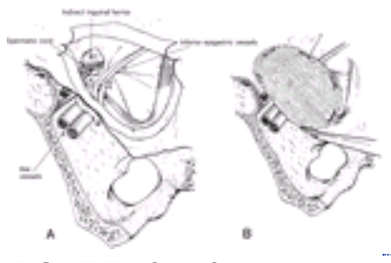

FIGURE 161-10. Laparoscopic inguinal herniorrhaphy. **A:** The peritoneum is incised to identify clearly the hernia and anatomy. **B:** Mesh is secured with staples to the Cooper ligament, iliopubic ligament, and transverse arch. (From ref. [1].)

More recently, many surgeons have begun performing a totally extraperitoneal laparoscopic hernia repair. [136] With the patient in the Trendelenburg position, the operation is begun with a 1.5-cm incision placed just below the umbilicus. An inflatable balloon is inserted into the properitoneal space and filled with saline to dissect the peritoneum from the posterior surface of the abdominal wall and create a working space in the groin. The working space is maintained by CO_2 insufflation, and two additional 5- and 10-mm ports are inserted. Mesh is laid between the fascial defect and peritoneum and may be fixed in place with staples. The advantage of the totally extraperitoneal approach is that the peritoneum is never violated. Therefore, the patient should avoid any complications of the transperitoneal herniorrhaphy related to adhesions and subsequent obstruction, intra-abdominal abscesses, or iatrogenic perforation injuries. The hernia repair also may be performed under regional anesthesia, thereby avoiding a general anesthetic.

Laparoscopic hernia repairs seem appropriate in certain settings. Many surgeons feel that laparoscopic herniorrhaphies are most beneficial for healthy patients with bilateral defects who would be incapacitated with bilateral groin incisions. Recurrent hernias previously repaired conventionally may be better repaired the second time laparoscopically from behind through “virgin” tissue rather than scar. Other patients who need to return to 100% physical activity with minimal delay are also suitable for a laparoscopic repair, which does not rely on suture strength to hold the repair together in the early postoperative period. Before laparoscopic herniorrhaphy gains full acceptance, the repair will need to become less costly, and long-term follow-up will need to prove its relative benefit.

Laparoscopic Management of Peptic Ulcer Disease

Ulcer surgery is rarely required in the current era because of the recent advances in medical therapy. The rare ulcer that is refractory to medical therapy and perforated ulcers have been treated laparoscopically with good results. Most commonly, perforations are closed primarily and then patched with omentum. [137] Laparoscopic truncal vagotomy with pyloric dilation or variations of proximal gastric vagotomy also boast nearly complete ulcer healing and few complications. [138] To date, most reports of laparoscopic ulcer surgery are anecdotal and have minimal follow-up data.

Laparoscopic Colectomy

Laparoscopic small and large bowel resections have been performed to treat benign and malignant disease. Using laparoscopic techniques, the colon is mobilized and the mesentery divided. The segment of colon may be exteriorized through a small incision, resected, and anastomosed before being dropped back into the abdominal cavity (coined *laparoscopy-assisted colectomies*). Alternatively, resection and anastomosis may be performed totally intracorporeally with transanal removal of the specimen. The extent of tissue margins and the number of lymph nodes included in the resection specimen suggest that laparoscopic techniques do not compromise an en bloc cancer operation, [139] although long-term follow-up will be necessary for confirmation. Patients usually are discharged from the hospital within 4 days, rather than the 5 to 7 days required after open operation. However, potential savings resulting from shorter hospitalization are offset by longer operative times and expensive laparoscopic equipment. [140], [141]

Laparoscopic resection of potentially curable colorectal malignancy is very controversial. A number of case reports of tumor metastases to trocar sites after laparoscopy-assisted surgery have raised concern that this approach may spread tumor mechanically. [142], [143], [144] and [145] In an animal model, pneumoperitoneum of 10 mm Hg for 10 minutes more than doubled tumor cell implantation at trocar sites. [146] Several multiinstitutional randomized trials comparing laparoscopic with open colectomy for resectable colon cancers are underway. For these reasons, laparoscopic colonic resection for cancer should be performed only on protocol.

Newer Procedures

Minimally invasive techniques may hold promise for many other procedures. We and others have had laboratory and clinical experience with adrenalectomy, nephrectomy, splenectomy, pancreatectomy, gastrectomy, and biliary bypass. Other surgeons have performed esophagectomy [147] and pancreaticoduodenectomy in humans. [148] However, just because an operation can be performed with a laparoscope does not mean that it should be. In our hands, laparoscopic performance of pancreaticoduodenectomy (the Whipple procedure) may have a higher complication rate and does not seem justified. [149] Randomized prospective outcome studies with longer periods of postoperative follow-up, as well as cost-benefit analyses, will be needed to settle the issues in an era of fiscal responsibility.

Emerging Technology

State-of-the art technology from the National Aeronautics and Space Administration and the defense industry is being redirected to solve many of the current limitations of laparoscopy. Several types of three-dimensional color video systems have been developed to restore the surgeon’s visual depth of field and possibly improve laparoscopic task performance for complex maneuvers such as suturing. [150] Flexible-tipped scopes and articulating instruments compensate for ports fixed to the abdominal wall. Biologic adhesives and new technology such as fibrin glue [151] and laser welding [152] are being investigated as alternatives to time-consuming suturing or costly stapling for anastomoses. The harmonic scalpel uses ultrasonic energy to coagulate vessels without desiccation and without obscuring vision with smoke. Dissecting balloons are creating small spaces in which to operate in the neck, axilla, and retroperitoneum. [136], [153] External retractors are used to lift the abdominal wall [14], [15] and avoid the potentially harmful local and systemic effects of a CO_2 pneumoperitoneum. Mechanical camera holders steady the camera and free the assistant to perform other tasks. Advances in simulation technology for pilot training are being applied to realistic laparoscopic surgical simulators to train surgeons, who may otherwise have limited laboratory and clinical experience with laparoscopy.

Laparoscopy has expanded the vision of general surgery, and the application of new technology is advancing rapidly. Robotic arms can precisely guide the laparoscope in response to the surgeon’s eye movements or voice commands. [12] Furthermore, slave robotic operating systems are being used, which can eliminate the surgeon’s tremor and allow ultraprecise surgical movements. [154] Interactive tactile feedback may be programmed into graphics to allow surgeons to “feel” what they are seeing on video. Three-dimensional images may be projected, not on a video monitor, but in the operating suite directly over the patient or superimposed on the patient to color-paint abnormalities for better identification during dissection. In time, applications of virtual reality may have surgeons wear Datasuits (Greenleaf Medical, Palo Alto, CA) and “fly” within the patient’s simulated abdomen to probe organs, just as architects today walk through simulated buildings testing structural properties. Teleproctoring may allow expert laparoscopic surgeons to guide novice surgeons safely along their learning curve as well as to establish a mode for credentialing and recertifying laparoscopic skills. Telepresence technology may even enable military surgeons safe behind-the-lines access to operate on patients on the battlefield. [155]

Compared with open operation, laparoscopy seems to offer most patients less postoperative pain, more rapid recovery, and better cosmesis. Patients are often discharged from the hospital earlier and return to active employment sooner. As the costs of technology decrease and the value of each health dollar is scrutinized, abdominal surgery will undoubtedly continue the evolution toward minimally invasive therapy.

LAPAROTOMY

The term laparotomy (Greek *lapara,* the flank; *otomy,* to cut into) is used loosely to indicate any operation in which the abdomen is cut open widely. An exploratory laparotomy is a surgical procedure performed through an abdominal incision for the diagnosis of intra-abdominal pathology. This exploration also forms a part of most

therapeutic abdominal operations, in which the primary objective is surgical correction of a previously defined problem. The number of exploratory laparotomies performed solely for the purpose of diagnosis is impossible to ascertain, but such procedures are relatively rare today. Medical advances, including improvements in diagnostic laparoscopy, flexible intralumenal endoscopy, imaging, and interventional treatment techniques, have reduced the necessity for laparotomy, but an appropriately directed abdominal exploration remains the most reliable method for diagnosing and treating obscure or acute catastrophic intra-abdominal illness. Particularly with the rapid acceptance of diagnostic laparoscopy by general surgeons, laparotomy has gained an increasingly worse reputation as being among the most costly, invasive, and morbid of diagnostic techniques available. In weighing the value of exploratory laparotomy as a diagnostic procedure, the accuracy and cost efficiency of diagnosis, corollary therapeutic benefit, and an accurate estimate of risk in the individual patient must all be assessed. Unfortunately, most of these end points are difficult to ascertain prospectively in the individual patient.

Surgical Stress Response

Laparotomy, with its surgical trauma, is always associated with an acute injury response. The degree of response is loosely associated with duration of anesthetic, degree of tissue injury, and metabolic derangements. Recovery after laparotomy occurs in stages similar to recovery from other types of traumatic injury. [156] Multiple abnormalities are seen in the perioperative interval in the hormonal milieu, [157] and cellular [158] and humoral [159] immunity are impaired. Furthermore, postoperative ileus is another reliable physiological consequence of laparotomy; although small bowel motility may be largely unaffected, the stomach and colon usually exhibit an ileus pattern of varying duration. [160] Recent research has suggested that virtually all of these aforementioned derangements occur to a lesser degree after laparoscopic procedures than after similar operations performed by laparotomy. [161], [162], [163], [164] and [165]

Laparotomy also increases myocardial oxygen demand while inducing physiological changes that potentially decrease oxygen supply to the heart. These alterations increase the risk for myocardial injury and arrhythmias for a period of up to 1 week postoperatively. [166] Laparotomy alone, particularly with incisions extending into the upper abdomen, remains an independent risk factor associated with a 15- to 25-fold increase in cardiac morbidity. [167] Upper abdominal incisions likewise cause major decreases in pulmonary function, particularly in functional residual capacity and lung compliance. [168] The diminution in abdominal wall trauma reduces the cardiopulmonary risks in patients undergoing laparoscopic procedures [169], [170] compared with the larger incisions used for operations performed by laparotomy.

PROCEDURE

Most exploratory laparotomies are performed using general endotracheal anesthesia with adequate muscle relaxation. The type of incision is dictated by patient factors such as prior incisions, intestinal stomas or wounds, and the indication for operation. Oblique or transverse incisions may work well for access to specific discrete lesions or organs (e.g., the right upper quadrant) while minimizing postoperative pain and pulmonary problems. [171] However, in the patient with unclear pathology or diffuse symptoms, as is the case with most patients undergoing true exploratory laparotomy, a midline incision has the greatest versatility for a complete abdominal exploration.

During exploratory laparotomy, all peritoneal and retroperitoneal organs are assessed in a systematic fashion. All available peritoneal surfaces are visually inspected with manual palpation of retroperitoneal structures. It is the use of the surgeon's hand with its trained haptic sensation that enhances the diagnostic capabilities of laparotomy above those of exploratory laparoscopy. The other primary advantage of laparotomy over laparoscopy is the ability to staunch the flow of major hemorrhage in a timely fashion. Laparoscopy is hindered by inadequate visualization when blood splashes on the lens or aggressive suctioning of blood evacuates the CO_2 pneumoperitoneum. It is also easier to control hemorrhage by direct pressure using one's hand placed through a laparotomy incision. Biopsy samples of abnormal tissues and aspirates of peritoneal fluid are sent for histology and, where necessary, cytology, bacterial culture, or chemical analysis. A systematic approach to exploration of the peritoneum is important to attain the full value of the procedure.

After the diagnostic phase, therapeutic maneuvers should be performed to achieve the goal set out preoperatively, as modified by the exploratory findings. In all patients, care must be taken to maintain normal temperature and hemostasis and to protect the tissues from drying or contamination. Upon completion of the procedure, the abdominal fascia must be closed in a secure fashion to prevent postoperative dehiscence or evisceration.

Indications for Laparotomy

Laparotomy is indicated for diagnostic purposes when a calculation has been made that the benefits of surgical intervention in a given patient outweigh the potential risk, complications, and costs of the procedure, when no other technique provides equal safety and accuracy of diagnosis, and when there is a reasonable likelihood that effective surgical treatment for the condition is possible. Exploratory laparotomy is the gold standard with which all other, less direct diagnostic techniques for abdominal pathology, including laparoscopy, must be compared. The range of specific diagnoses and procedures performed in or through the peritoneal space numbers in the hundreds. However, surgical pathology of the abdomen can be organized into several common categories (Table 161-5).

INDICATION	OBJECTIVES OF SURGICAL MANAGEMENT
Acute abdomen	Diagnosis and treatment
Visceral abdominal trauma	Diagnosis and treatment
Acute hemorrhage	Control intra-abdominal hemorrhage Define and control gastrointestinal source
Infection	Diagnosis Drainage or debridement
Inflammation	Diagnosis Ulcer disease: vagotomy and gastric drainage, repair perforation Cholecystitis/biliary colic: cholecystectomy Inflammatory bowel disease: resection or bypass Intestinal ischemia: revascularization, resection
Intestinal obstruction	Mechanical: repair Dysmotility syndrome: diagnosis, intestinal resection
Neoplasm	Diagnosis, staging Palliative bypass Resection

TABLE 161-5 Indications for Laparotomy

Medical and surgical advances, including laparoscopy and conventional imaging techniques, have reduced the necessity for laparotomy, and many of the indications for laparotomy listed in Table 161-5 may be performed using laparoscopic techniques. Several studies have emphasized the diagnostic value of laparotomy when there is a significant risk for missed or incorrect diagnoses of abdominal pathology, which can result in increased mortality. [172], [173] In contrast, blind abdominal exploration in the septic, gravely ill intensive care unit patient without specific evidence for an abdominal process has not been shown to influence survival. [174] Patients presenting with an "acute abdomen" (physical signs of peritonitis) are among those who benefit most from concomitant surgical treatment of their condition. Laparotomy is often the first and only diagnostic procedure for these patients other than physical examination. Although most patients who traditionally were thought to have indications for laparotomy may now be approached using laparoscopic techniques, in some cases initial laparotomy without consideration of laparoscopy is mandated. These situations would include patients with diffuse peritonitis of unclear etiology in whom there is a sense of urgency for diagnosis and treatment, those patients with an uncorrectable coagulopathy, those with hypotension and hemodynamic instability (in whom the pneumoperitoneum may cause vascular collapse), [175] and those with a "frozen" abdomen who have no true celomic cavity because of multiple prior operations with dense adhesions.

Conversion from Laparoscopy to Laparotomy

In many surgeons' practices, the aforementioned conditions are relatively rare, and most exploratory laparotomies occur as a result of conversion from an attempted laparoscopic procedure. The act of electively converting from a laparoscopic to an open operation should not be considered a complication but sound surgical judgment. Conversion generally occurs when the laparoscopic approach does not allow the surgeon to complete the objectives of the operation. These shortcomings would include the following: (1) inability to access the involved area, (2) inadequate laparoscopic visualization of the operative field, (3) inability to manipulate or intervene in a satisfactory manner, (4) lack of timely progression of the operation, (5) inability to treat a recognized complication by laparoscopic means.

Contraindications and Risks of Laparotomy

The contraindications to a laparotomy are, for the most part, relative rather than absolute. They primarily reflect the inherent risks of anesthesia, the surgical stress response, and the surgical wound. It is rare that the underlying health of a patient is so severely compromised or the risks of the procedure so prohibitive that he or she is absolutely ruled out as a candidate for laparotomy. If the indication for laparotomy is compelling and no good alternative treatment exists, the combination of preoperative resuscitation, modern anesthesia, expedient surgery, and postoperative intensive care can bring most patients through the stress of an operation. On the other hand, improvements in medical, endoscopic, laparoscopic, and interventional radiologic treatment now offer many alternatives that can achieve the same goals and may obviate the necessity for laparotomy, at least until the patient's status has improved.

Complications of Laparotomy

Numerous perioperative and postoperative complications may accompany laparotomy. These include cardiopulmonary complications, [176] complications of anesthesia and analgesia, [177] abnormal wound healing with wound infections or dehiscence, [178] venous thromboembolism, [179] and bowel obstruction. Abdominal surgery inevitably results in an injury to the peritoneal membrane. Exuberant healing of injured, ischemic peritoneum after surgery is the most common (80%) cause of abdominal adhesions in humans. [180] Adhesive small bowel obstruction occurs after 3% of all laparotomies, 20% of these within 1 month of operation, and a further 10% within the first year. Unfortunately, the risk for obstruction is lifelong. [181] Procedures centered inferior to the transverse mesocolon, such as colonic, small bowel, appendiceal, or pelvic procedures, have a higher risk for small bowel obstruction than upper abdominal operations. [182] There is a possibility of specific pharmacotherapy for adhesions in the future. [181], [183] Some early data suggest that laparoscopic procedures result in fewer adhesions than do their open counterparts. [97]

Results of Laparotomy

Conventional measures of efficacy for any procedure include morbidity, mortality, and long-term outcome from the disease process requiring laparotomy. More recently, other, less traditional measures of outcomes have assumed greater importance, including cost of therapy, length of stay, duration of disability, and quality of life. The very act of measuring cost and outcomes data has led to changes in the way physicians practice. These changes in physicians' habits have occurred simultaneous to the introduction of laparoscopy, rendering most retrospective comparisons between laparoscopic and open operations to be factitious. Surgery is a technical art, and the results of surgical therapy depend on skillful technique. Operator-dependent factors, such as excessive tension on tissues, inadequate hemostasis, technically poor anastomosis, excessive manipulation or inadequate margins in cancer surgery, and prolonged operative time, increase the risks for cancer recurrence, [184] complications, and death. [185] The skill and experience of the individual surgeon and the ancillary resources of the institution are crucial factors in patient outcome.

Conclusions

Exploratory laparotomy has been used for decades in patients with obscure or acute catastrophic intra-abdominal illness. Medical advancements, including improvements in flexible intralumenal endoscopy and radiologic imaging, have reduced the frequency with which direct or endoscopic imaging of the peritoneal cavity is necessary to establish a diagnosis. A number of studies suggest that when laparoscopy can be performed, it results in lower morbidity and probably less cost than laparotomy. Combined with the ability to perform diagnostic laparoscopy is the burgeoning field of therapeutic laparoscopic surgery, an area that remains in evolution today. A limited number of patients, however, because of acuity of illness or various confounding factors, are best served by a timely laparotomy performed in a skillful fashion. The choice between these techniques in the individual patient remains inherent to the art and craft of surgery.

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CHAPTER 162

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EXPERIMENTAL THERAPIES: HEPATOCYTE TRANSPLANTATION, GENE THERAPY, AND LIVER ASSIST DEVICES

Hepatocyte transplantation, liver-directed gene therapy, and artificial liver-assist devices are being developed in a parallel but interdependent manner. For example, hepatocyte transplantation can be used as a vehicle for liver-directed gene therapy. On the other hand, gene transfer can be used to expand hepatocytes in culture for transplantation and abrogation of immune response. This chapter provides a short overview of advances and the remaining hurdles in these areas.

HEPATOCYTE TRANSPLANTATION

Orthotopic liver transplantation has significantly improved the prognosis for patients with acute or end-stage liver failure or potentially lethal inherited metabolic disorders. Because of the expense, complexity, and associated morbidity and mortality of this procedure, hepatocyte transplantation is being explored as an alternative. Hepatocyte transplantation is performed by percutaneous or transjugular injection into the portal vein, spleen, or peritoneal cavity and therefore is less invasive than liver transplantation. As the host liver remains intact, the loss of graft function should not worsen liver function. Although intact livers must be transplanted shortly after procurement, isolated liver cells may be cryopreserved for later use. [1] The success of hepatocyte transplantation in experimental animals has encouraged the clinical trial of this procedure at several institutions. Potential indications for hepatocyte transplantation include acute or chronic liver failure and inherited metabolic disorders. Hepatocytes could be used as a vehicle for ex vivo gene therapy of inherited metabolic disorders. Hepatocyte transplantation is also being considered for rescuing patients from radiation-induced liver damage after radiotherapy for liver tumors.

Liver-Based Metabolic Diseases

Hepatocyte transplantation has been shown to correct several liver-based metabolic disorders in animals. The Gunn rat, an animal model of Crigler-Najjar syndrome type 1, lacks bilirubin-UDP-glucuronosyltransferase (UGT1A1) activity and consequently exhibits toxic levels of hyperbilirubinemia. [2] Gunn rats and Nagase genetically analbuminemic (NAR) rats have been studied extensively in hepatocyte transplantation experiments. Transplantation of normal donor hepatocytes has ameliorated the metabolic abnormality in both animal models. [3] In the absence of cirrhosis, hepatocytes injected into the splenic pulp or infused into the portal vein engraft in the liver, where they integrate into the hepatic cords and function throughout life. [4], [5] Hepatocyte transplantation has resulted in partial correction of metabolic disorders in low-density lipoprotein receptor-deficient [6] Watanabe heritable hyperlipidemic rabbits (an animal model of familial hypercholesterolemia) and in the Long-Evans Cinnamon rat (an animal model for Wilson disease). [7]

Although many biologically active liver proteins are present in excess, the expectation that liver-based metabolic deficiencies could be corrected by the transplantation of a very small number of hepatocytes did not materialize. Repeated hepatocyte infusions can improve the response to transplantation, [8] but more dramatic results have been obtained by progressive repopulation of the host liver with engrafted hepatocytes.

Massive Repopulation of the Liver Studies in fumarylacetoacetate hydrolase (FAH) mutant mice (a model of hereditary tyrosinemia type I) showed that transplanted normal hepatocytes can spontaneously repopulate the liver of animals in which hepatocytes are dying because of a metabolic disorder. [9] A small number of hepatocytes from normal mice, engrafted into the liver of FAH mutant mice, almost completely replace the native liver cells. [9] However, the transplant recipients remain susceptible to the development of hepatocellular carcinomas, despite extensive repopulation of their livers with normal hepatocytes. Therefore, risk for cancer or recurrent disease should be an important consideration in the clinical application of hepatocyte transplantation for a given disease. For metabolic diseases, in which host hepatocytes do not die spontaneously, strategies are being developed to inhibit host hepatocyte regeneration while providing a proliferative stimulus to the transplanted cells by partial hepatectomy, induction of Fas-mediated apoptosis of host hepatocytes, or administration of pharmacological doses of thyroid hormone. [10], [11] The plant alkaloid retrorsine blocks the hepatocyte cell cycle and has been used for this purpose. [12] Because retrorsine is potentially carcinogenic, X-irradiation of the liver has been used as a part of the preparative regimen. [13], [14] Transplantation of normal hepatocytes from congeneic donor rats into Gunn rats that had undergone partial hepatectomy and hepatic irradiation resulted in complete normalization of serum bilirubin levels.

Inherited Metabolic Disorders Patients with ornithine transcarbamylase (OTC) deficiency, a $_1$-antitrypsin deficiency, and Crigler-Najjar syndrome type I have been transplanted with allogeneic hepatocytes. Although one child with OTC deficiency showed some evidence of enzyme activity on a liver biopsy, she ultimately died shortly thereafter from hyperammonemia. Unequivocal evidence of function of transplanted human hepatocytes was obtained in one patient with Crigler-Najjar syndrome type I (UGT1A1 deficiency). [15], [16] After transplantation, the serum bilirubin level decreased by 50%, hepatic bilirubin glucuronidating activity increased from essentially unmeasurable pretransplantation levels to about 5% of normal, and bilirubin glucuronides were excreted in bile. Allograft rejection was prevented with standard tacrolimus therapy. The procedure was not associated with any significant complications, but the extent of replacement of hepatic UGT1A1 activity was not sufficient to eliminate the need for phototherapy.

Acute Liver Failure

Conceptually, hepatocyte transplantation should be particularly suitable for treating acute liver failure because the liver remains architecturally normal and retains considerable restorative potential. Hepatocyte transplantation can rescue rodents with acute liver failure induced by hepatotoxins, liver ischemia, or 90% hepatectomy [17], [18] and prevent the development of intracranial hypertension in pigs with acute, ischemic liver failure. [19] However, the outcome of hepatocyte transplantation in patients with acute liver failure has been disappointing, except, when used as a bridge to liver transplantation.

In the first clinical study, seven patients with fulminant liver failure were injected with human fetal hepatocytes into the peritoneal cavity. Some of the recipients recovered from encephalopathy, [20] and survival of patients with grade III hepatic encephalopathy was superior to that observed in the untransplanted control group. Because long-term survival of hepatocytes following direct injection into the peritoneum has not been shown, investigators in the United States have used other sites. Because of ethical concerns, adult hepatocytes, rather than fetal cells, have been used. In one study, seven transplantation candidates received hepatocyte infusion through the splenic artery; five of these survived to undergo orthotopic liver transplantation. [21] In other studies, patients who were not candidates for organ transplantation were transplanted with hepatocytes by infusion into the portal vein for engraftment in the liver. [22], [23] Engrafted hepatocytes were found in the spleen and liver of some of the transplant recipients, and there are anecdotal reports of improvement in ammonia, prothrombin time, encephalopathy, cerebral perfusion pressure, and cardiovascular stability. Complications have been few but include transient hemodynamic instability during intraportal hepatocyte infusion, sepsis, and

embolization of hepatocytes into the pulmonary circulation. [24] Portal hypertension as a result of transplantation through the portal vein has been generally transient. Although the results suggested that the transplanted hepatocytes provided some benefit, convincing evidence for engraftment and function of the transplanted cells has not been obtained. Part of the problem could be that relatively small numbers of hepatocytes were ransplanted in many of the patients.

In contrast to the situation in animal models of acute liver failure, liver regeneration is significantly inhibited in patients with acute liver failure. [25] Because injection of bone marrow cells and hepatocyte lysates have also been shown to affect animal survival, hepatocyte transplantation in the animal models might have helped by enhancing the regeneration of the host liver rather than through functional engrafted cells. Based on studies on a recently developed mouse model of acute liver failure that more closely parallels the clinical situation, [26] it is likely that multiple hepatocyte infusions over time may be required for improved patient survival.

Chronic Liver Failure

Studies in Experimental Animals Because of the difficulty in generating animal models of stable and irreversible liver cirrhosis causing hepatic failure, most studies on evaluating hepatocyte transplantation in treating chronic liver failure have been performed in other animal models of hepatic encephalopathy. Intrasplenic transplantation of hepatocytes in rats with end-to-side portacaval shunt resulted in improvement of behavioral score, partial correction of the amino acid imbalance, [27] and prevention of hepatic coma induced by ammonium chloride administration. [28] The engrafted hepatocytes proliferated in the spleen, organizing into structures resembling hepatic chords. More recently, hepatocyte transplantation has been shown to improve many of the physiological abnormalities associated with decompensated liver failure in rats with end-stage liver cirrhosis, induced by chronic administration of phenobarbital and carbon tetrachloride. Hepatocyte transplantation also prolonged the survival of these animals. [29] Transplantation of hepatocytes into cirrhotic liver results in a severe and prolonged increase in portal pressure. [30] Although some transplanted hepatocytes migrate into cirrhotic nodules after intraportal infusion, it is unlikely that enough cells can engraft in the cirrhotic liver to affect liver function significantly. Moreover, it is unclear whether the transplanted cells could function within the cirrhotic nodules. For these reasons, in the studies on cirrhotic rats, hepatocytes were engrafted in the spleen.
Clinical Experience The initial clinical trial was performed on 10 Japanese patients with liver cirrhosis to test the hypothesis that the functional failure of hepatocytes within the cirrhotic liver is due to the deranged architecture of the organ. Ten million to six hundred million hepatocytes harvested from their own left lateral liver segments were injected directly into the spleen, or infused into the splenic artery or the portal vein. In one patient, ascites and encephalopathy eventually resolved, and engrafted hepatocytes in the spleen were detected by ^{99m}TC (technetium-9mm)- radioisotope uptake 11 months after transplantation. [31] Subsequently, in the United States, infusion of up to 10 [10] allogeneic donor hepatocytes into the splenic artery in patients with decompensated chronic liver disease was shown to be well tolerated and associated with improvement in encephalopathy, hepatic protein synthesis, and renal function. [24] Nuclear scanning demonstrated the presence of engrafted hepatocytes in the spleen. If extrahepatic hepatocyte transplantation proves to be effective in improving liver function, it could be performed in combination with transjugular intrahepatic portosystemic shunt (TIPS) to relieve portal hypertension. However, the risk for hepatocellular carcinoma would persist, and routine surveillance would still be needed. Unfortunately, the improvement of liver function after hepatocyte transplantation in chronic liver failure patients has been relatively poor, compared with that observed in animal models. The discrepancy may have resulted from the fact that hepatocytes were transplanted by injection into the splenic pulp in rats, whereas they were infused in the splenic artery of the patients. [24] In fact, hepatocytes infused in the arterial bed of rats are lost rapidly. [32] Similarly, in patients, only a few hepatocytes were found in the spleen after infusion into the splenic artery.

Ongoing Issues

Technically, the number of hepatocytes that can be transplanted at any one time is limited. Using present techniques, less than 30% of transplanted cells engraft in the liver. A better understanding of the factors that allow hepatocyte engraftment might reduce the need for repeated cell infusions and multiple donors. The most important issue limiting the application of hepatocyte transplantation is the availability of donor livers, which is reduced by competition for use in whole-organ transplantation. The ability to cryopreserve and bank hepatocytes might allow pooling of cells from multiple donors, increasing cell availability for transplantation. Unfortunately, cryopreserved liver cells have not yet been shown to engraft as well as fresh hepatocytes, [33] and viability of human hepatocyte cryopreserved by current methods is variable.

Despite some progress in growing primary hepatocytes in culture, [34] currently, only the hepatocytes that have been immortalized by gene transfer are capable of long-term growth and correcting metabolic abnormalities in liver failure after transplantation. [28], [35] An alternative solution for the scarcity of human donor cells is the transplantation of xenogeneic hepatocytes. [36] However, xenogeneic hepatocytes are susceptible to immunologic processes that are not active after allotransplantation. [36], [37] Xenograft survival in humans has been of such short duration that whether organs or tissues from another species will function adequately to replace human organ function remains uncertain. [38] It is also a concern that xenogeneic liver cells could transfer infectious agents to the recipient and, potentially, beyond. [39] It is possible, however, that transplanted nonhuman hepatocytes could be resistant to recurrence of infection by human-specific viruses.

Finally, it is difficult to detect early rejection of the transplanted hepatocytes, and new noninvasive approaches to the detection of donor hepatocytes are needed.

LIVER-DIRECTED GENE THERAPY

Goals

Targets of liver-directed gene therapy include both inherited metabolic disorders and acquired conditions, such as infectious and neoplastic diseases, cirrhosis of the liver, and immune rejection of transplants. Liver-directed gene therapy could be used for diverse purposes. Gene products that are missing in inherited disorders could be replaced by transferring transcription units expressing those proteins. In other cases, certain genes could be overexpressed for specific therapeutic benefits, such as treatment of cirrhosis by expression of metalloproteases. The large size of liver and its capability to secrete proteins to the plasma could be exploited to generate extrahepatic proteins, such as coagulation factors or hormones. Proteins that are normally expressed at extrahepatic sites could be expressed in the liver for specific purposes. For example, the catalytic subunit of the apolipoprotein B messenger RNA editing enzyme (APOBEC-1), which is expressed in the intestinal epithelial cells, could be expressed ectopically in the liver to switch the hepatic apolipoprotein production from apo B100 to apo B48, thereby reducing the generation of low-density lipoproteins. [40] Similarly, PDX, a homeobox protein that is responsible for pancreatic differentiation, could be expressed in the liver, to switch the hepatocyte phenotype to an insulin-generating phenotype. [41] Gene transfer to the liver could be used to generate pharmacological gene products, such as vaccines, single-chain antibodies, dominant negative proteins, immunomodulatory substances, and proapoptotic or antiapoptotic proteins. On the other hand, nucleic acids may be used to inhibit the expression of deleterious proteins, such as viral proteins or mutant a $_1$-antitrypsin. This could be accomplished by transferring synthetic antisense RNAs or ribozymes. Genes could be transferred to express antisense RNAs, [42] ribozymes, or dominant negative proteins. [43] Finally, technologies are being developed to correct mutations within the endogenous genes in intact organisms. This could be accomplished by targeted replacement of the defective gene [44] or by site-directed correction of a target genomic sequence. [45] These newer strategies permit the repaired gene to remain under the control of the endogenous promoter, whereby physiological regulation is retained. A partial list of inherited and acquired disorders targeted for liver-directed gene therapy is provided in Table 162-1.

I. Inherited Liver Disorders
Crigler-Najjar syndrome type I
Familial hypercholesterolemia and other lipid metabolic disorders
Maple syrup urine disease
Progressive familial intrahepatic cholestasis
Phenylketonuria
Tyrosinemia
Mucopolysaccharidosis VII
α_1-Antitrypsin deficiency
Ornithine transcarbamylase deficiency
Wilson disease
Glycogen storage diseases, e.g., von Gierke disease and Pompe disease
II. Inherited Systemic Disorders
Hemophilia A and B
Oxalosis
III. Acquired Disorders
Infectious diseases, e.g., hepatitis B and C
Malignant neoplasms: hepatomas, cholangiocarcinomas, metastatic tumors
Extrahepatic tumors (inhibition of neovascularization)
Cirrhosis of the liver
Allograft or xenograft rejection

TABLE 162-1 Some Hepatic Disorders that are Currently Targeted for Gene Therapy

Cancer Like other therapeutic approaches, the target is to eliminate the tumor cells while minimizing the damage to normal cells. Gene transfer could be used to kill the tumor cells or inhibit their growth, reduce the vascular supply to tumors, generate immune response against tumor cells, or augment the effect of chemotherapy and radiotherapy. Enhancement of apoptosis of the tumor cells could be promoted by expressing *p53,* a sentinel gene of the cell cycle. [46] Tumor killing is attempted by transferring "suicide genes," such as the herpes simplex virus thymidine kinase (*HSV-TK*), which converts a prodrug, ganciclovir, to its active phosphate derivative, [47] or cytosine deaminase and purine nucleoside phosphorylase (which converts fludarabine to a diffusible toxic metabolite). [48] The activated toxin, such as ganciclovir phosphate, may diffuse to neighboring cells, killing them by "by-stander effect." Toxicity to normal cells could be reduced by tumor-specific delivery by tagging the DNA to a monoclonal antibody, such as that directed against AF-20, a 180-kd tumor-specific cell-surface glycoprotein, expressed in hepatoma cell lines, [49] and by using tumor-specific promoters (e.g., alpha-fetoprotein or carcinoembryonic antigen) to drive the expression of transgene. In an alternative approach, E1B-mutant adenoviruses that are capable of replicating in tumor cells deficient in functional *p53* have been used in an attempt to kill tumor cells, [50] although the replication of these mutant adenoviruses may not necessarily depend on the absence of *p53*. Direct killing of the tumor cells by topical expression of toxic gene products does not eliminate those at metastatic sites. Because tumor requires neovascularization, inhibition of new blood vessel formation by the expression of angiostatin or endostatin may retard the growth of both primary and metastatic tumors. [51], [52] Permanent elimination of the tumor cells may require host immune response against tumor-specific "neoantigens" that are not expressed by normal cells. Specific tumor antigens, such as those from melanoma cells, may permit DNA-based tumor vaccination. Host immune response against the neoantigens may be feeble because of inadequate presentation of the antigens by antigen-presenting cells (APCs). Genetic manipulation of APCs may augment the host immune response against tumor cells. Secretion of cytokines, such as transforming growth factor-ß or interleukin-10, by large tumors may suppress immune response. [53] "Debulking" the tumor by surgery, radiotherapy, chemotherapy, or gene therapy may enhance the immune response. Currently, gene therapy for cancer has the greatest potential as an adjunct to chemotherapy or radiotherapy. Irradiation enhances the number of tumor cells transducible by recombinant viruses. Transgene expression could be driven by promoters that are stimulated by irradiation. [54] On the other hand, inhibition of the expression in the tumor cells of radioprotective proteins, such as ATM (mutated in ataxia telangiectasia), could increase their radiosensitivity.

Infectious Diseases Nucleic acid–based approaches are being explored both for prophylaxis and treatment of hepatic viral infections. DNA vaccination [55] eliminates the possibility of contamination of the vaccines by microbes or other organic material. The injected DNA itself serves as an adjuvant, enhancing the immune response. For viral antigens that are presented poorly, the antigenic peptides may be expressed directly in APCs by gene transfer. Gene transfer may also be used to express cytokines that promote the APC proliferation and maturation of T cells to helper T cells. In a different approach, termed *intracellular vaccination,* single-chain antibodies or antibody fragments may be expressed within the cells to make them resistant to viral infections. [56] Gene therapy can also be used to interfere with the viral life cycle by introducing synthetic antisense RNAs, ribozymes, or DNA ribonucleases (see later). Ribozymes, antisense RNA, or dominant negative proteins [57] can also be expressed within the target cells by gene transfer.

Inherited Disorders Because many inherited disorders are caused by the abnormality of a single gene, the effect of gene therapy can be evaluated directly and precisely in these conditions. For this reason, although the inherited disorders are much less common than acquired diseases, single gene abnormalities continue to be important targets of liver-directed gene therapy. Table 162-1 contains a partial list of inherited disorders that are currently targets of gene therapy.

Methods of Gene Transfer to the Liver

Genes may be delivered to the liver through isolated hepatocytes, termed ex vivo gene therapy, or by delivery of the gene into the intact liver. Hepatocyte transplantation-based methods were discussed previously. In this section, we consider available methods of delivering genes to the intact organism, using replication-deficient recombinant viruses, or nonviral vectors. For gene transfer to hepatocytes, important considerations include the attainable infectious titer, ability of the virus to infect nondividing cells, integration into the host genome, repeatability of administration, and safety of the vector system. Frequently used methods of gene transfer to the liver are listed in Table 162-2. Some commonly used recombinant viral vectors are described below.

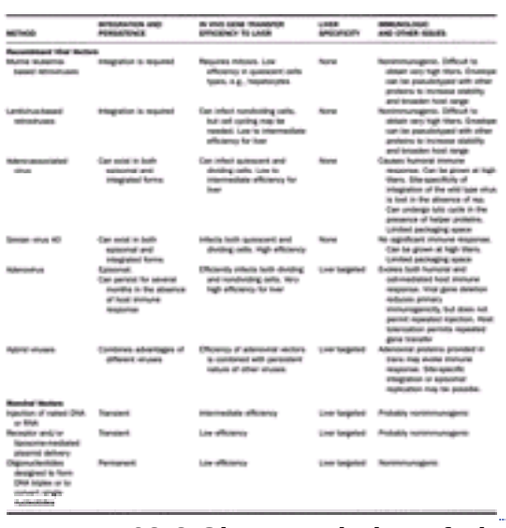

TABLE 162-2 Characteristics of Liver-directed Gene Therapy Methods

Vectors Based on Recombinant Viruses

Retrovirus-based vectors. Complementary DNA generated from the RNA genome of retroviruses are integrated into the host genome and are transmitted to the progeny of the transduced cells. [58] Murine leukemia viruses, particularly Moloney murine leukemia virus (MoMuLV), are used commonly for generating recombinant vectors (Fig. 162-1). The viral genes are replaced by target transgenes, but the packaging signal (*?*) is retained. [59] The viral proteins are provided in *trans* by "packaging cell lines," generating a replication-deficient recombinant virus (Fig. 162-2). The range of species that recombinant retroviruses can infect is determined by the envelope protein. The host range can be expanded by engineering the viral envelope to contain proteins, such as the G protein of the vesicular stomatitis virus (VSV). Inside the cell, the RNA genome is reverse-transcribed into double-stranded DNA, which is transported to the nucleus and is integrated into the host genome. This process requires mitosis of the host cell, which is infrequent in hepatocytes of the adult liver. To overcome this obstacle, retroviruses of the lentivirus group, which form preintegration complexes that can be translocated into nondividing nuclei, are being explored for liver-directed gene therapy. However, lentivirus-based gene transfer *in vivo* may also require the hepatocytes to be in cell cycle. [60]

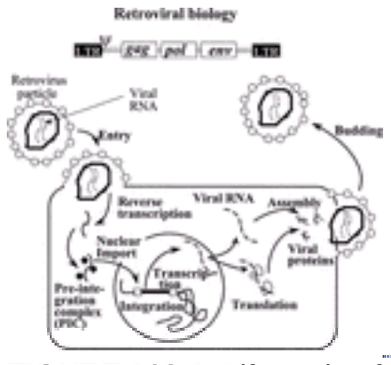

FIGURE 162-1. Life cycle of wild-type Moloney murine leukemia virus. The RNA genome of the virus consists of the retroviral genes, *gag, pol,* and *env,* flanked on the 5' and 3' ends by the long terminal repeats (LTRs), consisting of a promoter, enhancers, and polyadenylation signals. Sequences upstream to the 5' LTR contain the packaging signal (*?*) that is needed for packaging of the viral RNA genome into viral particles. After entry into the infected cell, the RNA genome undergoes reverse transcription, and the DNA transcript is incorporated into a preintegration complex (PIC), which is imported into the nucleus. Dissolution of the nuclear envelope, which occurs during mitosis, may be needed for this step. The proviral DNA is then integrated into the host genome. The entire unspliced transcript provides the new viral genome, which is packaged with the viral proteins expressed from the integrated proviral DNA. The assembled virus buds out and is ready to infect another cell.

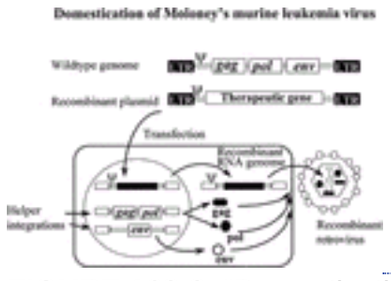

FIGURE 162-2. "Domestication" of the Moloney murine leukemia virus. The packaging cell is stably transfected with multiple DNA segments expressing different viral proteins (gag, pol, and env). A DNA version of the viral genome is cloned into a plasmid. All viral genes are replaced by target transgene, but the packaging signal (') is left intact. The recombinant plasmid is transfected into the packaging cell, which provides the viral proteins in *trans*. The viral RNA genome can be assembled with the viral proteins because of the presence of the packaging signal, '. The assembled recombinant virus buds out of the packaging cell into the culture medium. The recombinant virus can infect other cells but is replication incompetent owing to the lack of the viral genes.

Recombinant adenovirus. Adenoviruses are large linear double-stranded DNA viruses. Adenovirus types 5 and 2 are commonly used for preparing gene transfer vectors. Recombinant adenoviruses can be generated at high titers, can infect both dividing and quiescent cells, and upon intravenous administration in rodents, can localize preferentially to the liver. [61] The adenoviral receptor, CAR, is highly concentrated in rodent liver, but less so in the human liver. Thus, it is unclear whether the high gene transfer efficiency observed in the rodent liver will be replicated in human liver. Adenovirus vectors are generated by inserting the transgene into the early region-1 (*E1*) gene, which encodes transcription factors required for the expression of adenoviral genes. The recombinant virus is generated in packaging cells that provide the *E1* gene products in *trans*. All adenoviral genes may be deleted to increase the "stuffing space" and to prevent any possibility of expression of viral proteins. [62] These "gene-deleted" vectors require replication competent helper adenoviruses to provide the structural proteins. The helper virus is removed during purification. Strong immunogenicity of the viral antigens limits the clinical application of adenovectors. Neutralizing antibodies generated after the first injection block gene transfer following subsequent administrations of the vector. Cytotoxic lymphocytes against adenoviral antigens attack the adenovirally infected host cells, causing liver damage and rapid loss of the transgene after secondary gene transfer. [63] The helper-dependent, gene-deleted ("gutless") adenovectors, in which all viral genes are deleted, retain their immunogenicity, [64] thereby frustrating the attempts at repeated gene transfer. Immunosuppressive genes have been incorporated into adenovectors to inhibit host immune response to adenoviral proteins. [65] Development of effective immune response requires costimulatory interaction between antigen-presenting cells and cytotoxic lymphocytes by the B7-CD28 and CD40-CD40 ligand interactions. Injection of CTLA4-Ig, a soluble inhibitor of B7-CD28 costimulation, prolongs the expression of transgenes introduced by adenoviral vectors but, used alone, does not permit repeated administration of adenovectors. [66] However, coexpression of CTLA4-Ig and the target transgene permits multiple administration of the recombinant adenovirus. [67] Alternatively, host tolerance toward adenoviral proteins can be induced by injecting recombinant adenoviruses in utero into newborn rats, [61] inoculating adenoviral proteins into the thymus of young adult rats, [68] or orally administering small doses of adenoviral proteins. [69] However, safety concerns persist regarding tolerization of humans to adenoviral antigens because wild-type adenoviruses are pathogenic to humans.

Herpes simplex virus type 1. Herpes simplex virus type 1 (HSV-1) is a 150-kb double-stranded DNA virus with a broad host range [70] and ability to infect nondividing cells. However, long-term gene expression in the liver has not been achieved with the available HSV vectors.

Recombinant baculovirus. The *Autographa californica* nuclear polyhedrosis virus (AcNPV) is usually employed for generating recombinant proteins in insect cells. [71] Interestingly, among mammalian cells, hepatocytes can be infected by baculovirus vectors, and the transgene expression can be driven by mammalian promoters. Hybrid viruses, consisting partly of baculoviral sequences, are being evaluated for in vivo application.

Recombinant adeno-associated virus. Adeno-associated virus type 2 (AAV-2) is a small (4.7-kb) single-stranded DNA virus of the parvovirus family. The wild-type virus integrates preferentially on the q13.4-ter arm of human chromosome 19. [72] The AAV remains latent in the cell genome for long periods but can cause lytic infection following infection with a "helper virus," such as adenovirus or herpes simplex virus, or by genotoxic stimuli, such as ultraviolet light or irradiation. [73] Binding of AAV-2 to a heparin sulfate proteoglycan at the cell surface is required for its internalization. The AAV genome is flanked by 145-bp inverted terminal repeats (ITRs) that are needed for integration into the host genome. After introduction into cells, AAV vectors may persist as episomes or integrate into the host genome over several weeks. The viral Rep protein directs the site specificity of AAV integration into chromosome 19. Recombinant vectors lacking this gene lose site specificity of integration and in fact may remain in the nucleus as episomes [74] (Fig. 162-3).

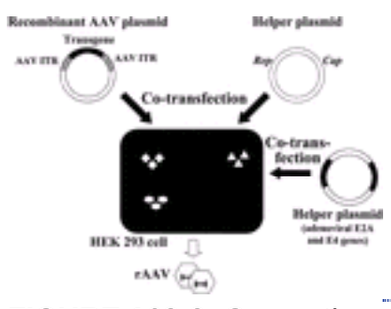

FIGURE 162-3. Generation of recombinant adeno-associated viruses (AAVs). To generate AAV vectors, two plasmids, one containing the transgene, flanked by the inverted terminal repeats (ITRs) and the other encoding rep, cap, and adenoviral proteins are cotransfected into 293 cells, which provide adenoviral E1A in *trans*. The recombinant virus is "rescued" by infection with a helper adenovirus (E1A-deleted) or, more commonly, by transfecting a plasmid that expresses adenoviral proteins.

Initial studies of intramuscular administration of AAV-based vectors for delivering coagulation factor IX in patients with hemophilia B resulted in low levels of protein production, [75] but infusion of high doses of the recombinant virus into the portal vein of factor IX–deficient dogs resulted in the appearance of 5% of normal levels of factor IX activity in plasma. [76] Efficiency of AAV-mediated cancer gene therapy could be augmented when used in combination with conventional tumor therapies, such as irradiation or chemotherapy. [77] Recombinant AAV causes a humoral immune response, which may be a problem if the vector needs to be readministered.

Simian virus 40–based vectors. Simian virus 40 (SV40) is a nonenveloped virus of the papova family with a 5.2-kb double-stranded DNA genome. The large (Tag) and small (tag) T antigens are transcription factors required for the expression of the viral structural genes, *VP1, VP2,* and *VP3.* The recombinant vector is generated by replacing the *Tag* genes of the viral genome by the target transgene. Various lengths of the structural genes can also be deleted to make additional stuffing space. The recombinant viral genome is transfected into a helper cell line (e.g., COS cells that provide Tag in *trans*). Absence of the *Tag* gene makes the recombinant virus replication deficient and markedly reduces its immunogenicity. Up to 4.7 kb of exogenous DNA can be inserted into SV40 vectors, which can be generated at 10^{9} infectious units (IU)/mL and concentrated to 10^{12} IU/mL. [78], [79] Recombinant SV40 integrates into the genome of both dividing and nondividing cells. The range of target cells is broad and includes hepatocytes and bone marrow cells. [78], [79]

Nonviral Vectors

Liposomes, lipoplexes, and polyplexes. Nonviral vectors comprise (1) lipid-based delivery systems, such as lipid microcapsules (liposomes) or lipid-nucleic acid complexes (lipoplex); (2) polycations, such as poly-L-lysine (PLL), polyethylenimine (PEI), polyglucosamines, lipopolyamines, and cationic peptides; and (3) polycation-lipid hybrids (lipopolyplex). DNAs are usually compacted with polycations before lipid encapsulation into liposomes or forming complexes with lipids. Lipopolyplex particles are smaller than are the liposomes or lipoplexes and provide greater protection of the nucleic acids from degradation by nucleases. Cationic lipid transfecting agents (e.g., lipofectamine, DOTAP), and cationic polymers, (e.g., PLL and PEI), provide relatively effective transfection [80] but are inefficient in the presence of plasma and too toxic for use in vivo. For targeting the gene delivery to hepatocytes, the gene transfer vehicles have been modified to include ligands that are endocytosed by hepatocyte-specific receptors, such as the asialoglycoprotein receptor (ASGPr). Galactose-terminated peptides (e.g., asialoorosomucoid) or galactose can be conjugated to polylysine or PEI for hepatocyte-specific gene delivery. [81] Similarly, liposomes composed of galactocerebrosides serve as ligands for the ASGPr through the galactose residues. [82] Incorporation of nuclear localization signal peptide motifs on plasmid constructs enhances their translocation to the nucleus. [83] Nucleic acid transfection generally results in transient effect in vivo, which may be sufficient when using synthetic antisense RNAs or ribosymes. Long-term effect is possible if the DNA integration can be enhanced by some mechanism, such as using the transposon-transposase system, or if the transfected nucleic acid triggers a permanent change, such as site-directed gene repair. Some specific applications of nonviral gene transfer methods are discussed below.

Ribozymes, antisense RNA, and DNA ribonucleases. Ribozymes are RNA enzymes that hybridize to complementary RNA sequences and catalyze endoribonucleolytic cleavage, causing rapid degradation of the RNA molecule. [84] For hairpin ribozymes, the presence of a guanosine residue immediately downstream to the cleavage site is essential, but the effect is enhanced by a GUC sequence. Hammerhead ribozymes require only a UA, UC, or UU dinucleotide for cleavage. Ribozymes can be synthesized and packaged for cellular uptake or expressed in cells from transfected DNA. Hammerhead [84] and hairpin [85] ribozymes have been shown to inhibit the production of hepatitis B and hepatitis C viruses in cells. Antisense oligonucleotides targeted to 5'-untranslated region of hepatitis C virus were transferred to a differentiated human hepatoma cell line HuH7 using ASGPr-polylysine complexes, resulting in specific inhibition of the hepatitis C–directed protein synthesis in the cells. [86] Multiple sites within the hepatitis C RNA genome may be targeted simultaneously using a series of ribozymes expressed from a single vector, thereby inhibiting the development of drug-resistant mutants. [87] Hepatitis B virus, a partially double-stranded DNA virus, replicates through a pregenomic RNA intermediate, which could also be a target for ribozyme-based RNA cleavage. [85] DNA ribonucleases are synthetic single-stranded DNAs comprising a 15-nucleotide catalytic domain, flanked by two RNA-binding domains, that catalyze RNA cleavage with a greater efficiency than ribozymes. [88] DNA ribonucleases against the hepatitis C viral genome can specifically cleave the viral RNA. DNA ribonucleases designed to cleave hepatitis B RNA transcript substantially inhibit viral gene expression. [89]

Homologous recombination. Site-specific recombination of DNA could be a precise means of repairing mutated or damaged DNA. Several proteins, including homologs of yeast recombination proteins rad51 or rad52, are important in homologous recombination in higher eukaryotes. [90] Although used extensively in cultured cells, the process is inefficient and has been rarely successful in vivo. [91] As homologous recombination is cell cycle–regulated, it occurs extremely infrequently in quiescent cells, such as hepatocytes. [92]

Triplex DNA. This method is based on the formation of a triple DNA by the binding of an exogenous single-stranded nucleic acid to the major grove of a homopurine region of the double-stranded genomic DNA. [93] The polypurine regions must be guanine rich and 12 to 14 nucleotides in length for adequate triplex formation to occur. Conventionally, cross-linking agents, such as psoralen or other mutagens, are covalently attached to the triplex forming oligonucleotide. [93] After intercalation of the psoralen at the target 5'ApT3' site, ultraviolet irradiation causes cross-linking of the thymines in the two strands. This substrate is then repaired by endogenous cellular DNA repair mechanisms, producing the characteristic T:A to A:T transversions. Single-strand DNAs with cross-linking agents can also be used to promote insertions and deletions at target through the excision repair pathway. [94] The target sequence constraints currently limit the application of this method in vivo. Bifunctional oligonucleotides comprising regions that form triple-helical structures as well as conventional Watson and Crick base pairs have partly overcome this hurdle in cultured cells. [95]

Single nucleotide modification. A method that uses the cells' mismatch repair machinery to correct single base mutations has been introduced recently. A synthetic oligonucleotide is generated that is complementary to the targeted genomic DNA, except at a single mismatched base. Most of the successful studies have used a DNA/RNA chimera. The RNA component, consisting of 2'-O-methyl ribonucleic acid residues, is included to increase the strength of hybridization with the targeted DNA sequence. The mismatch between the oligonucleotide and the genomic DNA triggers the mismatch repair system of the cell, so that the genomic mutation is permanently corrected. In *Escherichia coli* strains deficient in specific repair proteins, [96] this approach was shown to require RecA and MutS, proteins involved in DNA pairing and mismatch repair, respectively, suggesting the involvement of both the homologous recombination and mismatch repair pathways. Both recombination and mismatch repair pathways are evolutionarily conserved. Extracts of human hepatoma cells (HuH-7) containing the mismatch repair protein hMSH2 can complement MutS-deficient *E. coli* in converting the mutant aminoglycoside resistance gene. Wild-type *p53* may inhibit the initial pairing step in this repair process by inhibiting the pairing activity of RecA and its human homolog Rad51. [97] In intact rats, RNA-DNA chimeric oligonucleotides were delivered to the liver by receptor-mediated endocytosis, using lactosylated PEI or galactocerebroside-containing liposomes. In initial studies, a Ser365Arg conversion of the rat factor IX gene was achieved with significant frequency. [98] Subsequently, the guanosine base deletion in the bilirubin-UDP-glucuronosyltransferase (*UGT1A1*) gene in jaundiced Gunn rats was repaired at a frequency that was sufficient to express detectable enzyme activity in the liver and reduce serum bilirubin levels significantly. [99] Nucleotide insertion was verified by differential hybridization, Southern blot, and Western blot analysis. Tagalakis and associates [100] have demonstrated a high level of conversion of the gene-expressing human apolipoprotein E2 in a transgenic mouse to apolipoprotein E3 after intraperitoneal injection of a RNA-DNA chimeric oligonucleotide. In addition to hepatocytes, the RNA-DNA chimera has been used successfully for site-directed gene conversion of the apolipoprotein *A2* gene in human lymphocytes, [100] carbonic anhydrase II in nude mouse primary kidney tubular cells, [101] the missense mutation in the tyrosinase gene in albino mouse melanocytes, [102] and the dystrophin gene of myocytes. [103]

Transposon-based gene delivery. Transposable genetic elements (transposons) are ubiquitous in eukaryotic genomes and are capable of moving from one chromosomal location to another by a "cut-and-paste" mechanism. Radice and associates [104] characterized five transposable elements from fish, which were similar in structure to the Tc1 transposon of the nematode *Caenorhabditis elegans*. These transposons of the Tc1/*mariner* superfamily consist of short inverted repeats that flank an open reading frame, encoding a transposase. Ivics and associates [105] generated a synthetic transposable element from defective copies of an ancestral Tc1-like fish transposon. This transposon was termed "Sleeping Beauty" because it had remained dormant for millions of years during evolution and became functional only after correcting mutations that had accumulated over many generations. It is a 1.6-kb element, flanked by 250-bp terminal inverted repeats. DNA sequences, flanked by the terminal inverted repeats, can efficiently integrate into mammalian cellular genome, provided the transposase is expressed in the same cell. The integration occurs at TA dinucleotide sites, which are duplicated upon insertion of the transposable element. [105] The coding region of human factor IX has been inserted into the genome of hepatocytes of factor IX–deficient hemophilic mice by cotransfection of one plasmid containing the factor IX transcription unit flanked by the inverted repeats and another plasmid expressing the Sleeping Beauty transposase. [106]

EXTRACORPOREAL LIVER ASSIST DEVICES

The fact that some patients with acute liver failure recover spontaneously led to the idea that patient survival could be improved by providing metabolic support until the liver had time to regenerate. During the 1960s and early 1970s, ex vivo and in situ cross-circulation with animal and human livers was shown to improve clinical and biochemical parameters in animal models of liver failure and in uncontrolled human trials. [107] However, without the option of transplantation, there was no evidence of increased survival. In the 1980s, the life-saving effects of liver transplantation in fulminant hepatic failure was shown, resulting in expanding demand for a finite number of donor organs, increasing the difficulty of obtaining suitable organs in a timely manner. Thus, the concept of temporary liver support as a bridge to successful liver transplantation emerged. Because coagulopathy, renal failure, encephalopathy, and cerebral edema negatively affect the prognosis after liver transplantation, survival could improve if these complications could be held at bay while waiting for a suitable organ. Liver-assist devices have been considered also for use in acute exacerbation of chronic liver failure to alleviate encephalopathy following gastrointestinal bleeding or to prevent hepatorenal syndrome. Because many of these patients are not transplantation candidates, the potential for liver-assist devices in improving survival is limited.

Although many liver support systems are in the development stage, only a few have been tested in animal models. Although the survival benefit of these devices in patients with acute liver failure is yet to be shown in controlled, randomized trials, much progress has been made in the bioengineering of artificial liver support systems. Hepatic support systems comprise three basic groups, namely (1) cell-free liver assist devices; (2) entirely biologic systems, such as extracorporeal liver perfusion; and (3) hybrid bioartificial devices that include both nonbiologic and biologic components. A brief discussion of these devices follows.

Cell-free Liver Assist Devices

To substitute for the detoxification and excretion functions, hemodialysis, [108] exchange transfusion, [109] and hemoperfusion [110] through charcoal or resins have been attempted during the past 40 years, without consistent beneficial outcomes. Currently, two such systems are available for clinical use. In the BioLogic-DT plasma filter system, patient plasma comes in contact with a powdered charcoal suspension. [111] Many putative toxins that are thought to contribute to the complications of hepatic failure bind to these columns in vitro. Clinical trials have suggested improvement in patients with decompensated chronic liver disease, but studies in acute hepatic

failure did not show any benefit over standard clinical management.

The second system, named the *molecular absorbent recirculating system* (MARS), is a cell-free system for selective removal of albumin-bound substances (Fig. 162-4). As standard dialysis is included in the circuitry, some water-soluble substances are also removed. [112] An albumin-enriched dialysate is passed through an anion exchange resin column and a charcoal column and recirculated through the dialysis unit. This system effectively reduces plasma bilirubin and bile acid concentrations in patients with cholestasis. Use in more than 200 patients has suggested that treatment with this system may improve encephalopathy scores and the hemodynamic status, thereby increasing urine output in hepatorenal syndrome reducing ascites. Significant improvement in cases of acute-on-chronic decompensation was documented in controlled trials. [112] Patients with fulminant hepatic failure have been bridged to transplantation, and four cases of primary graft hypofunction have been supported until recovery with the MARS system. [113] Although these results are encouraging, the cell-free plasma detoxification systems cannot substitute for the metabolic and synthetic functions of the liver. Therefore, other liver support systems, including liver tissues or liver-derived cells, are being developed.

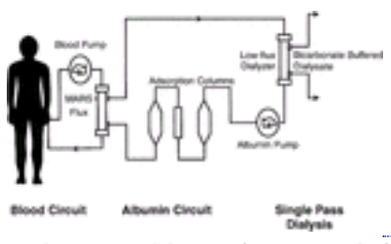

FIGURE 162-4. The MARS (molecular adsorbent recirculating system) unit, a cell-free liver assist device. An albumin-enriched dialysate is passed through a series of adsorption columns, including an anion exchange resin column and a charcoal column, and recirculated through the dialysis unit.

Extracorporeal Liver Perfusion

Otto and colleagues [114] in 1958 demonstrated that allogeneic extracorporeal liver perfusion could reduce plasma ammonia in hyperammonemic dogs. Eiseman and associates [115] applied this concept in patients in the 1960s using both human and xenogeneic livers. By 1994, 87 cases treated with extracorporeal perfusion using porcine liver or human cadaver livers were reported. [116] More recently, at the University of Nebraska, 14 patients received extracorporeal liver perfusion using eight human and eight pig livers. Of the 10 patients who were considered suitable for transplantation, 9 were “bridged” to liver transplantation. [117] In one study, livers from transgenic pigs expressing human CD55 and CD59 to limit hyperacute rejection were used for the management of two patients with fulminant hepatic failure. [118] Although controlled trials of extracorporeal liver perfusion have not been carried out, the complete recovery of three patients and the prolonged survival of some patients suggest improvement over conventional treatment. [118] However, even the best results using extracorporeal liver perfusion are not equal to that of liver transplantation. Membranes, filters, and tubing included in the circuitry may activate leukocytes, leading to cytokine release. Because immunosuppression is avoided to reduce the risk for sepsis, humoral and cellular rejection can occur. Finally, perfusion of the extracorporeal organ with systemic, rather than the portal, blood may reduce hepatic function. Logistic hurdles to extracorporeal perfusion include the limited availability of human donor organs and difficulty of procuring porcine livers, unless an institution maintains its own colony. Concerns over safety and efficacy of porcine organs persist.

Hybrid Bioartificial Liver Support Devices

Standardized machinery, including bioreactors containing metabolically active hepatocytes, have been in development in recent years. Initially, Matsumara and colleagues [119] used a device containing liver cell suspension to treat one patient, but it is now recognized that hepatocytes function more effectively when anchored to a surface. In a clinical trial on patients with acute liver failure, perfusion of patient plasma over porcine hepatocytes attached to Biosilon microcarriers, 37 of the 59 patients recovered, but only 25% of those with grades 3 and 4 encephalopathy survived. [120] The hepatocytes lost function after a few hours of application. Other systems that use hepatocytes cultured onto stacked flat surfaces [121] or three-dimensional structures [122] have been designed but not yet tested in clinical studies. The largest clinical experience is with systems consisting of a large number of cellulose-acetate hollow fibers packaged into plastic cartridges. The intralumenal and the extralumenal compartments can be perfused independently. In all devices presently undergoing clinical trial, the intralumenal compartment is used for perfusion of blood or plasma, and the hepatocytes are inoculated into the extralumenal compartment, although Nyburg and associates [123] have designed an experimental system in which gel-entrapped hepatocytes are packed within the intralumenal compartment.

Sussman and co-workers employed C3A cells, a human hepatoblastoma derived cell line, grown to confluence in the extralumenal compartment of hollow fiber cartridges in their Extracorporeal Liver-Assist Device (ELAD). [124] Whole blood from patients was perfused through the intralumenal compartment. Although initial uncontrolled observations in 11 patients with acute hepatic necrosis were encouraging, a pilot control trial demonstrated no difference in outcome compared with a control group receiving conventional supportive therapy. [125] In this study, the survival rate was surprisingly high in both the treatment (78%) and the control group (75%), probably because of the inclusion of patients with very early stages of acute liver failure.

Rozga and associates [126] used cryopreserved primary pig hepatocytes, attached to collagen-coated microcarriers in a device, named HepatAssist (Fig. 162-5). In this device, hepatocytes are introduced into the extralumenal compartment of hollow fiber cartridges, and the intralumenal compartment is perfused with patient plasma, which has been previously circulated through a charcoal column to remove toxins in the “hepatic failure” plasma. Each cartridge functions for 6 hours only, but the course may be repeated several times. The ongoing controlled trial of HepatAssist has not been published at the time of this writing.

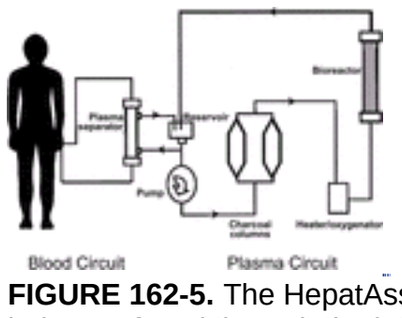

FIGURE 162-5. The HepatAssist device, an example of a hybrid liver assist device. Plasma is separated from blood cells and passed through a charcoal column being perfused through the intralumenal compartment of a hollow-fiber bioreactor. The extralumenal space of the bioreactor is packed with cryopreserved primary pig hepatocytes, attached to collagen-coated microcarriers.

A third hybrid hollow fiber system, Modular Extracorporeal Liver Support (MELS), uses woven polyether-sulphone capillaries that exit from the bioreactor in discrete bundles, so that different bundles can serve separate functions such as plasma flow and oxygenation. [127] The three-dimensional structure is claimed to improve the function of the cultured hepatocytes. CellModule bioreactors containing primary porcine hepatocytes were used to treat eight patients with liver failure, all of whom were carried through to successful transplantation. More recently, primary human hepatocytes obtained from cadaver donor organs were used, leading to encouraging results in six patients with acute failure (I.M. Sauer and J.C. Gerlach, personal communication, 2001).

Hurdles that Need to be Overcome for the Development of Extracorporeal Liver Support

Currently, porcine livers are the main source of the biologic component of artificial liver devices. Limitations of using xenogeneic sources include hyperacute or

delayed immune response to the cells or their products and concerns regarding the presence of infectious organisms, such as the porcine endogenous retrovirus. The use of human hepatocytes would be logical but is limited by donor organ shortage. Therefore, more progress is needed for generating human hepatocytes that can be expanded in vitro. The potential sources, including embryonic stem cells, hepatocyte progenitors, fetal hepatocytes, and genetically engineered immortalized hepatocytes are being explored actively.

Preclinical evaluation of extracorporeal liver-assist devices has been limited by the lack of animal models of fulminant hepatic failure that fully represent the clinical course of acute liver failure in patients. The relatively low incidence of acute liver failure makes it difficult to perform a detailed clinical evaluation of a hepatic support system at any one center, on the other hand, multicenter trials add to the variables to be considered in the trial design. The wide spectra of etiology, severity at presentation, and rapidity of progression of acute liver failure complicate the standardization of entry criteria and randomization. The level of encephalopathy in patients is difficult to quantify. Because liver transplantation is known to improve survival after fulminant hepatic failure, the extracorporeal hepatic support may be applied only until a donor organ becomes available, making the data analysis difficult. However, even if the present generations of devices do not prove sufficiently efficacious, the experience gained will undoubtedly help in the future development of an effective extracorporeal liver support.

Acknowledgment

We acknowledge the important contributions of many investigators in the fields of hepatocyte transplantation, liver-directed gene therapy, and extracorporeal liver-assist devices that were not cited because of limited space.

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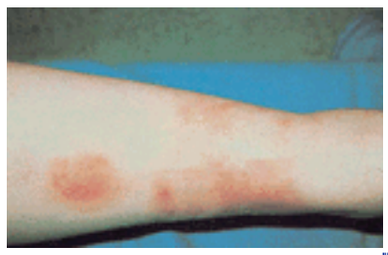
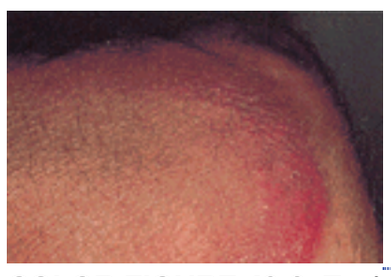
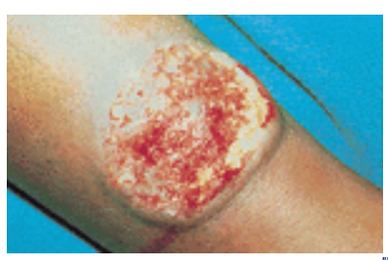
Color Plates



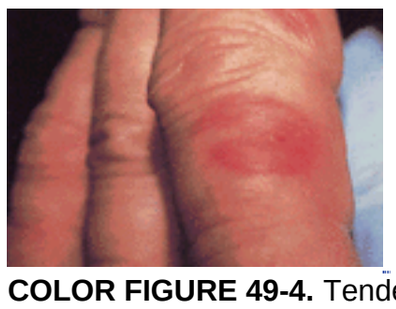
COLOR FIGURE 49-1. The tender nodules of erythema nodosum occurred in association with ulcerative colitis. (This figure is printed in black and white as Figure 49-1.)



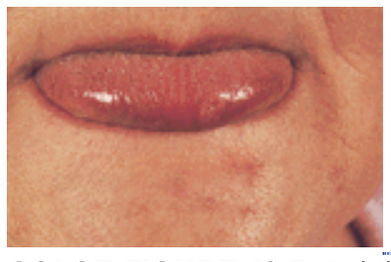
COLOR FIGURE 49-2. Typical lesions over joints in a patient with erythema elevatum diutinum. (This figure is printed in black and white as Figure 49-2.)



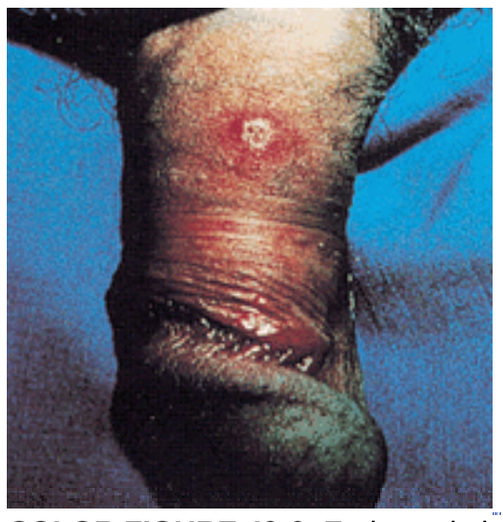
COLOR FIGURE 49-3. Pyoderma gangrenosum in a patient with Crohn's disease. (This figure is printed in black and white as Figure 49-3.)



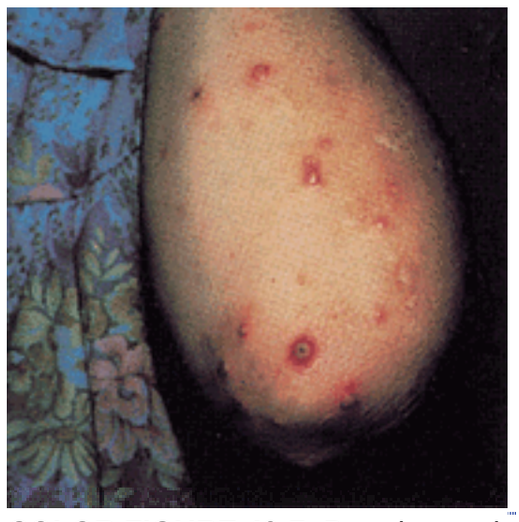
COLOR FIGURE 49-4. Tender erythematous papules and plaques on the upper extremities in a patient with Sweet syndrome. (This figure is printed in black and white as Figure 49-4.)



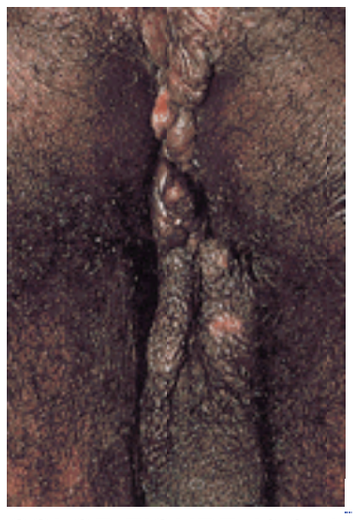
COLOR FIGURE 49-5. Aphthous ulcer involving the tongue in a patient with Behçet disease. (This figure is printed in black and white as Figure 49-5.)



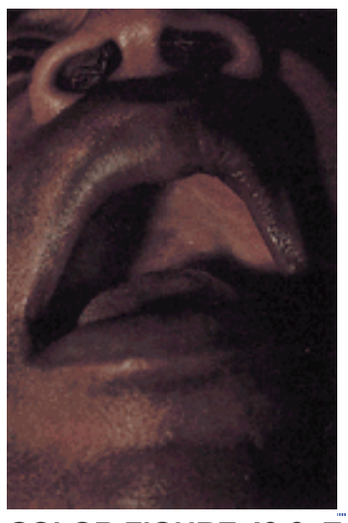
COLOR FIGURE 49-6. Early genital aphtha showing features of pustular vasculitis in a patient with Behçet disease. (This figure is printed in black and white as Figure 49-6.)



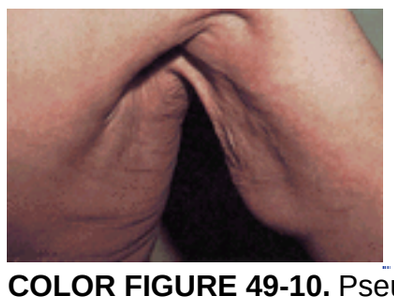
COLOR FIGURE 49-7. Bowel-associated dermatosis-arthritis syndrome. Pustular vasculitis lesions occurred in a patient with a blind loop after Billroth II surgery. (This figure is printed in black and white as Figure 49-7.)



COLOR FIGURE 49-8. Amyloidosis. Note the perirectal amyloid nodules in this patient with multiple myeloma. (This figure is printed in black and white as Figure 49-8.)

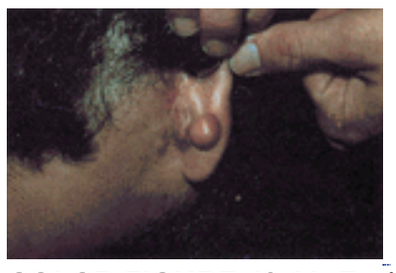


COLOR FIGURE 49-9. Typical telangiectasia of Osler-Weber-Rendu disease. (This figure is printed in black and white as Figure 49-9.)

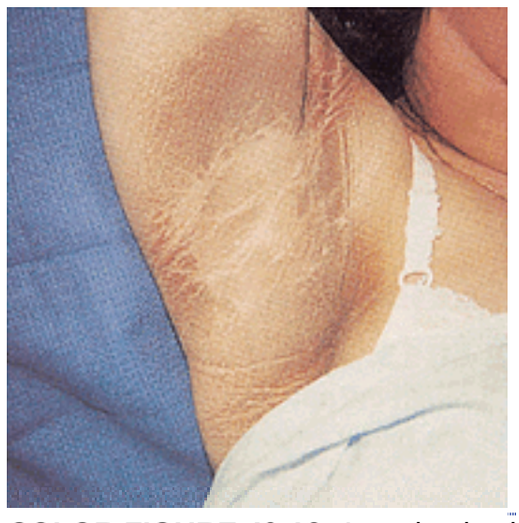


COLOR FIGURE 49-10. Pseudoxanthoma elasticum. Note the chicken skin–like appearance of the axillary skin. (This figure is printed in black and white as Figure

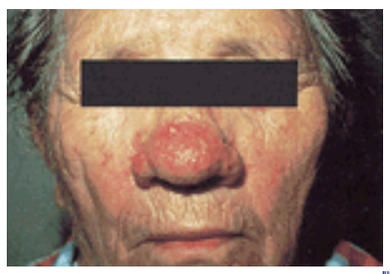
49-10.)



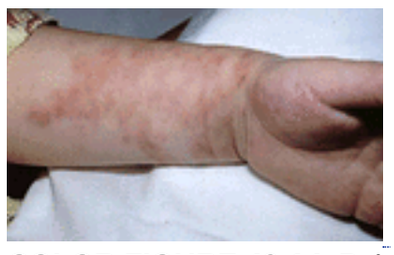
COLOR FIGURE 49-11. Typical epidermal inclusion cyst of Gardner syndrome. This patient had many other cystic nodules, particularly on the scalp. (This figure is printed in black and white as Figure 49-11.)



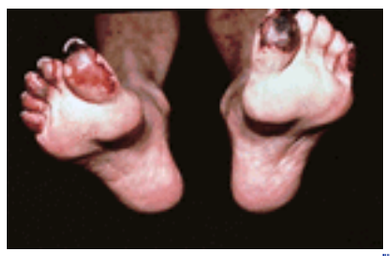
COLOR FIGURE 49-12. Acanthosis nigricans in a nonobese adult patient. (This figure is printed in black and white as Figure 49-12.)



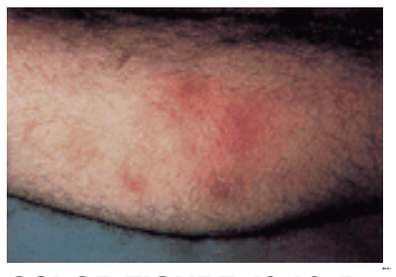
COLOR FIGURE 49-13. Characteristic telangiectasias, acneiform papules, and rhinophyma in a patient with rosacea. (This figure is printed in black and white as Figure 49-13.)



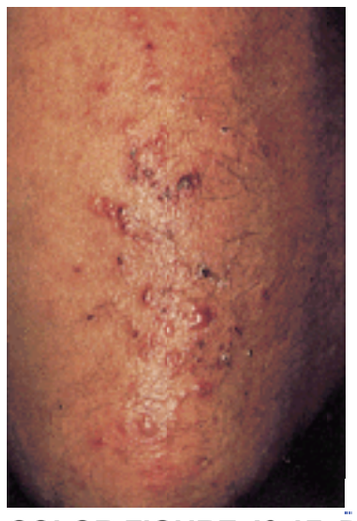
COLOR FIGURE 49-14. Polyarteritis nodosa with characteristic livedo reticularis and tender nodules. (This figure is printed in black and white as Figure 49-14.)



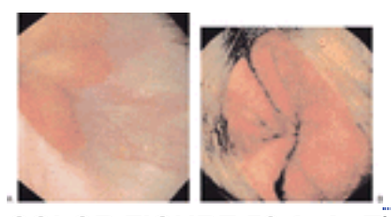
COLOR FIGURE 49-15. Cryoglobulinemia often results in acral vasculitic infarcts. (This figure is printed in black and white as Figure 49-15.)



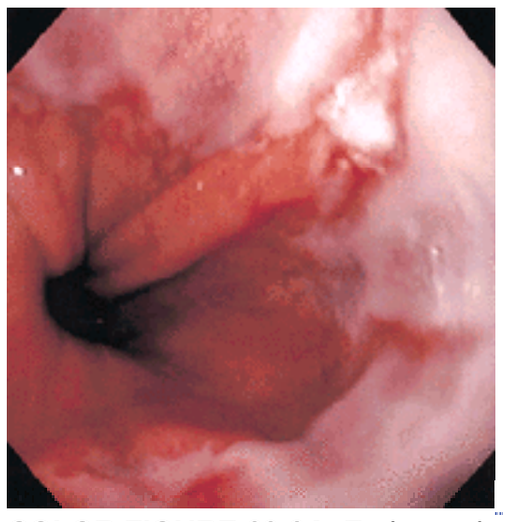
COLOR FIGURE 49-16. Panniculitis presented as tender erythematous nodules in a patient with pancreatitis. (This figure is printed in black and white as Figure 49-16.)



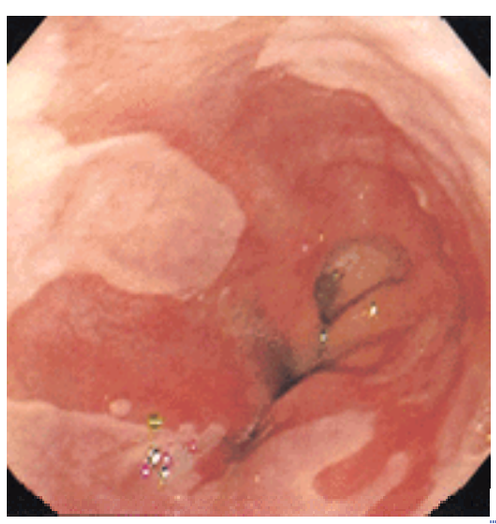
COLOR FIGURE 49-17. Dermatitis herpetiformis. Both elbows are involved. Note the intact vesicles and multiple crusted (i.e., excoriated) lesions. (This figure is printed in black and white as Figure 49-17.)



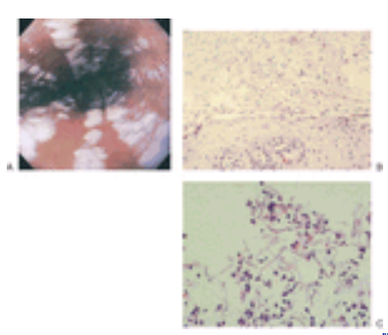
COLOR FIGURE 58-1. A: This view of the normal squamocolumnar junction reveals a slightly serrated contour, a distinct color difference between the two types of mucosa, and linear esophageal vessels that disappear at the mucosal junction. The slightly elevated gastric mucosal folds contact the squamous margin. **B:** The squamocolumnar junction has been better defined by dark Lugol iodine staining of the glycogen-containing squamous cells. This normal junction is located just below the diaphragmatic hiatus. (This figure is printed in black and white as Figure 58-1.)



COLOR FIGURE 60-9A. Endoscopic signs of esophagitis. Two linear erosions extend proximally from the squamocolumnar junction at the proximal border of a hiatal hernia. This would be classified as grade II esophagitis by the Savary-Miller and Hetzel systems and as grade B by the Los Angeles scale. (This figure is printed in black and white as Figure 60-9.)

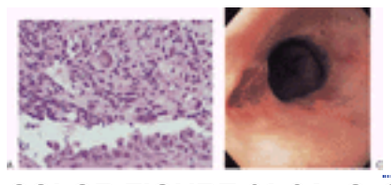


COLOR FIGURE 60-12A. Barrett esophagus. Reddish pink columnar mucosa with tonguelike projections 3 to 4 cm into the tubular esophagus. (This figure is printed in black and white as Figure 60-12.)

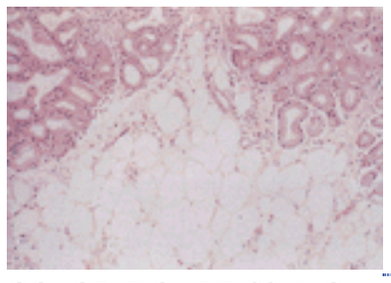


COLOR FIGURE 61-1. *Candida* esophagitis. **A:** Endoscopic photograph shows multiple raised plaques involving the esophagus with normal intervening mucosa. **B:** Desquamated squamous epithelial cells admixed with fungi and inflammatory cells adherent to the mucosa. The underlying squamous epithilium appears normal. **C:**.

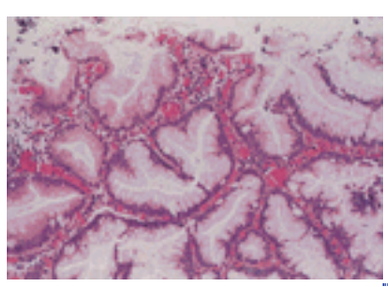
Close-up view of the plaque material demonstrates mycelia and spores typical of *Candida.* (This figure is printed in black and white as Figure 61-1.)



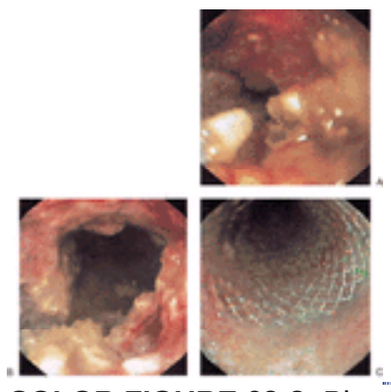
COLOR FIGURE 61-3A, C. Cytomegalovirus esophagitis. **A:** Biopsies from an ulcer base demonstrate granulation tissue with one large cell with intranuclear and intracytoplasmic inclusions diagnostic for cytomegalovirus. **C:** Endoscopic photograph shows the two ulcers, which are well circumscribed and have some depth, and the surrounding mucosa is normal. (This figure is printed in black and white as Figure 61-3.)



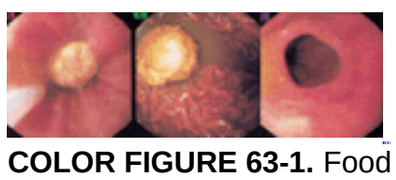
COLOR FIGURE 62-4. Specialized-type Barrett esophagus. The epithelium shows intestinal-type absorptive cells, goblet cells, and mucinous cells in a villiform pattern. (Courtesy of Robert Odze, MD.) (This figure is printed in black and white as Figure 62-4.)



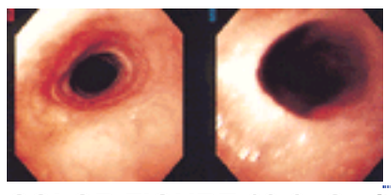
COLOR FIGURE 62-5. High-grade dysplasia in Barrett esophagus. The epithelium shows architectural complexity, atypia, pleomorphism, and nuclear stratification. (Courtesy of Robert Odze, MD.) (This figure is printed in black and white as Figure 62-5.)



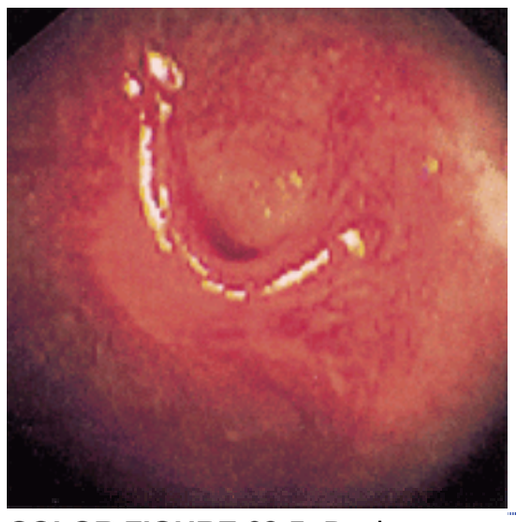
COLOR FIGURE 62-8. Photodynamic therapy of esophageal cancer with laser after administration of porfimer sodium, a photosensitizer. Light of 630 nm from a laser acts on cells that accumulate the photosensitizer (**A**). After 6 days, there is some decrease in the mass size (**B**). After 12 days, the mass is diminished in size significantly, and a metallic endoprosthesis is endoscopically inserted (**C**). (Courtesy of Norman Nishioka, MD.) (This figure is printed in black and white as Figure 62-8.)



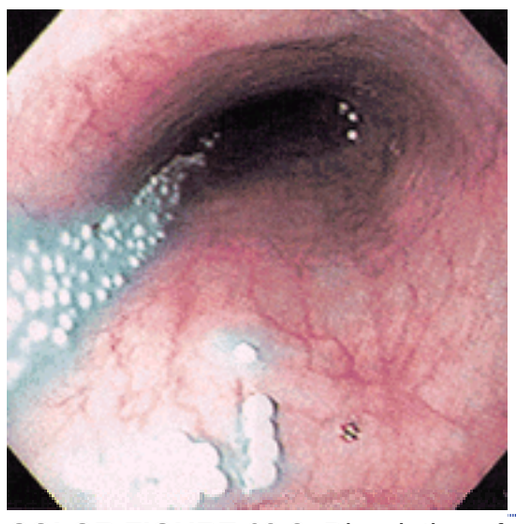
COLOR FIGURE 63-1. Food impaction in the distal esophagus (**A**). When cleared by advancing the bolus into the stomach (**B**), a Schatzki ring is evident (**C**). (This figure is printed in black and white as Figure 63-1.)



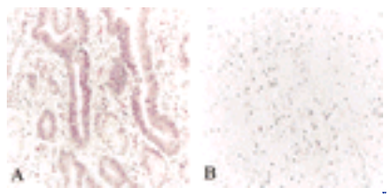
COLOR FIGURE 63-3. Graft-versus-host disease. Multiple fine mucosal webs are present in the esophagus. (This figure is printed in black and white as Figure 63-3.)



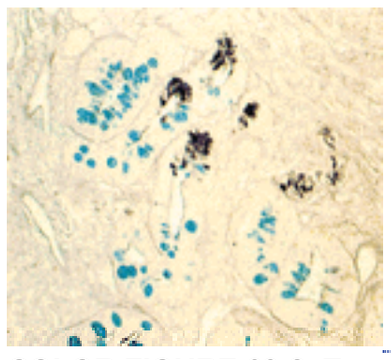
COLOR FIGURE 63-5. Benign mucous membrane pemphigoid. Also known as cicatricial pemphigoid, this endoscopic photograph depicts a tight esophageal stricture. (This figure is printed in black and white as Figure 63-5.)



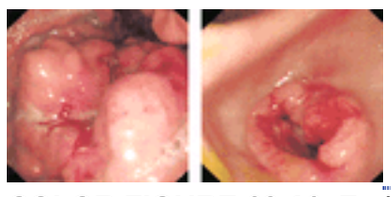
COLOR FIGURE 63-8. Dissolution of a medication in the midesophagus. (This figure is printed in black and white as Figure 63-8.)



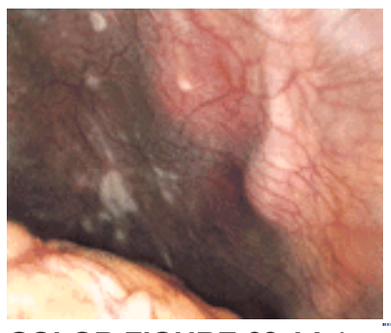
COLOR FIGURE 69-6. Lauren classification of gastric adenocarcinoma. **A:** Well-differentiated intestinal-type gastric cancer. **B:** Diffuse-type cancer with infiltrative and discohesive tumor cells. (Courtesy of Dr. K. F. To, Chinese University of Hong Kong, Prince of Wales Hospital, Hong Kong.) (This figure is printed in black and white as Figure 69-6.)



COLOR FIGURE 69-8. Type III intestinal metaplasia of stomach. The presence of sulfomucin in type III intestinal metaplasia is illustrated by the high iron diamine (*HID*)/Alcian blue staining. (This figure is printed in black and white as Figure 69-8.)

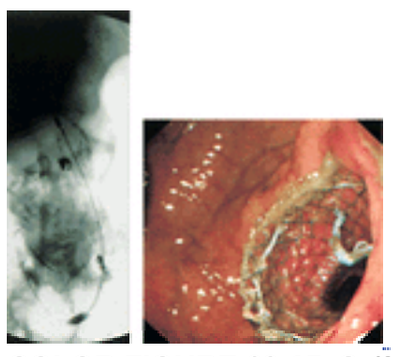


COLOR FIGURE 69-10. Endoscopic appearance of gastric cancer. (This figure is printed in black and white as Figure 69-10.)

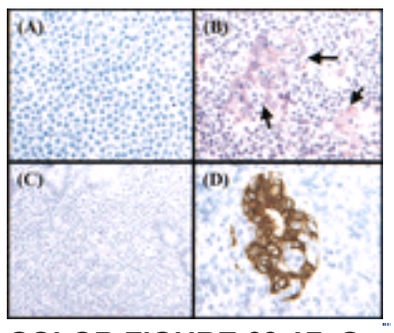


COLOR FIGURE 69-14. Laparoscopic view of a patient with gastric carcinoma with peritoneal metastasis (whitish nodule). (This figure is printed in black and white as

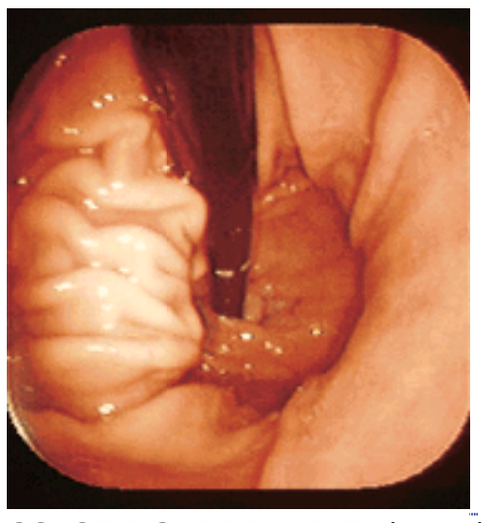
Figure 69-14.)



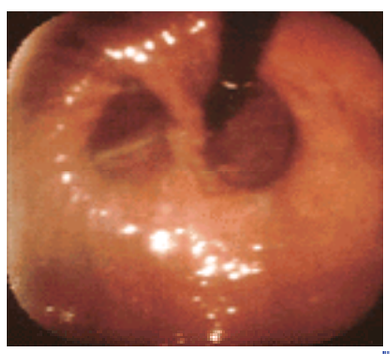
COLOR FIGURE 69-15. Self-expandable metal stent in patient with recurrent gastric carcinoma causing gastric outlet obstruction after subtotal gastrectomy. (This figure is printed in black and white as Figure 69-15.)



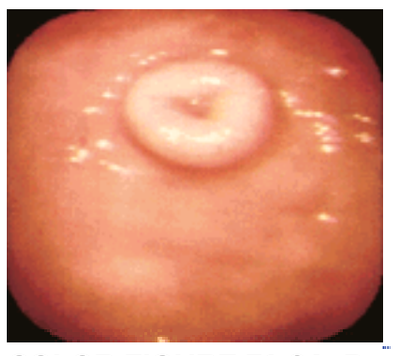
COLOR FIGURE 69-17. Gastric mucosa-associated lymphoid tissue (MALT) lymphoma: characteristics features of low-grade MALT lymphoma. (**A**) Centrocyte-like cells (hematoxylin and eosin, ×200). (**B**) Lymphoepithelial lesions (marked by *arrows*) (hematoxylin and eosin, ×200). (**C**) Plasmacytic differentiation (hematoxylin and eosin, ×100). (**D**) Lymphoepithelial lesions (immunostain for cytokeratin, ×200). (Courtesy of Dr. Wing Y. Chan, Chinese University of Hong Kong, Prince of Wales Hospital, Hong Kong.) (This figure is printed in black and white as Figure 69-17.)



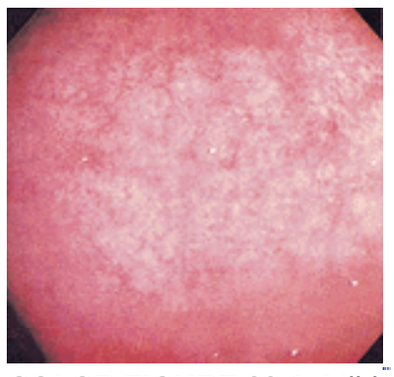
COLOR FIGURE 71-1A. Endoscopic view of a type I hiatal hernia while in gastric retroflexion. (This figure is printed in black and white as Figure 71-1.)



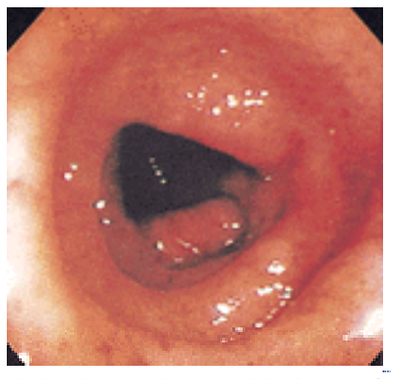
COLOR FIGURE 71-2. Endoscopic view of a paraesophageal hernia while in gastric retroflexion. The fundus is herniating into the thoracic cavity alongside a concomitant type I hernia. (This figure is printed in black and white as Figure 71-2.)



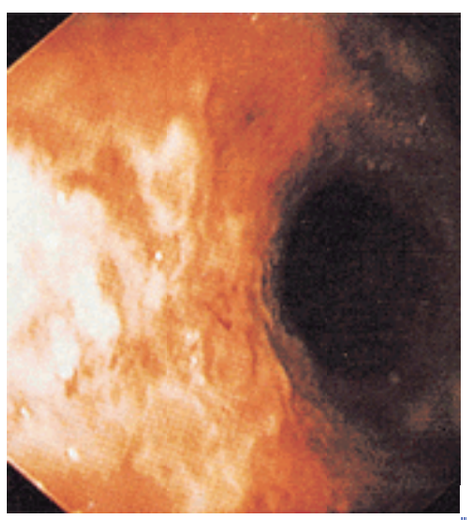
COLOR FIGURE 71-8A. Pancreatic rest identified in the antrum during upper endoscopy. (This figure is printed in black and white as Figure 71-8.)



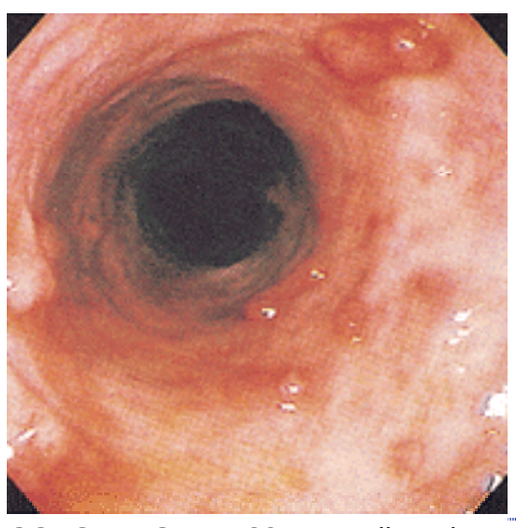
COLOR FIGURE 83-1. Mild ulcerative colitis with erythema, blurring of the vascular pattern, and granularity. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-1.)



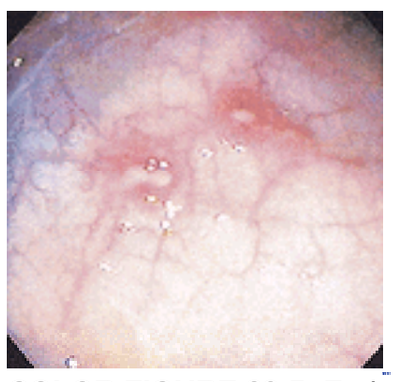
COLOR FIGURE 83-2. Moderate ulcerative colitis with prominent edema, blunting of the haustral pattern, diffuse granularity, and loss of the vascular pattern. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-2.)



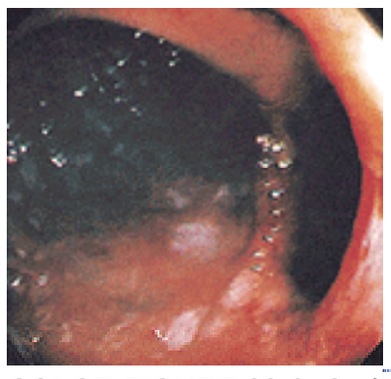
COLOR FIGURE 83-3. Moderate-to-severe ulcerative colitis with coarse granularity, marked edema with early nodule formation, and superficial ulcerations. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-3.)



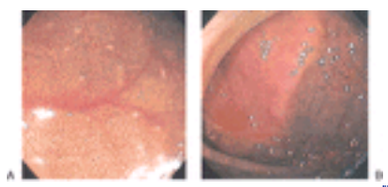
COLOR FIGURE 83-4. Healing ulcerative colitis with residual edema, absent vasculature, and postinflammatory polyps in a tubular, ahaustral lumen. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-4.)



COLOR FIGURE 83-5. Early Crohn's disease with aphthoid ulcers. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-5.)

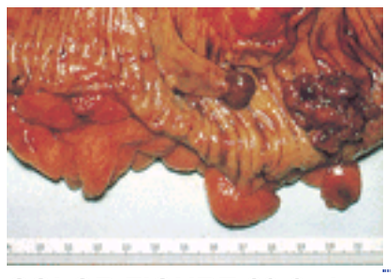


COLOR FIGURE 83-6. Crohn's ileocolitis with rounded, punched-out ulcers around the ileocecal valve. (Courtesy of the Crohn's and Colitis Foundation of America, New York, NY.) (This figure is printed in black and white as Figure 83-6.)

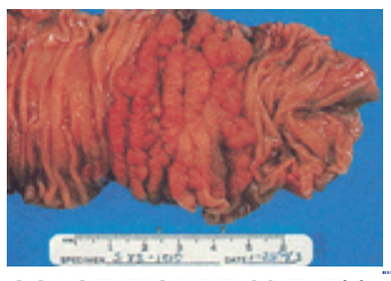


COLOR FIGURE 85-3. Endoscopic photographs of melanosis coli. **A:** The reticulated, alligator-skin appearance is characteristic of this condition. **B:** Melanosis coli with a sharp line of demarcation at the small bowel–colonic anastomosis. (This figure is printed in black and white as Figure 85-3.)

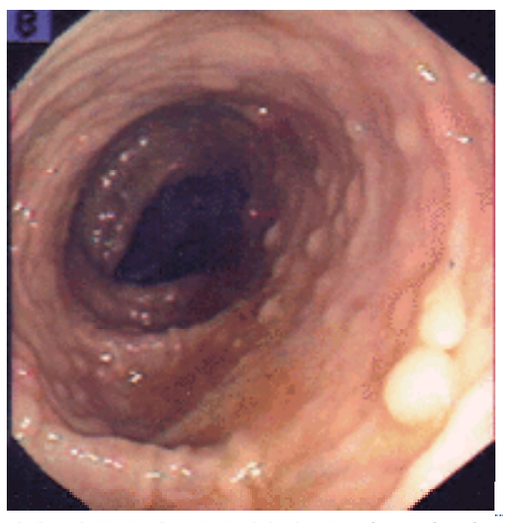
COLOR FIGURE 89-2. Macroscopic appearance of a pedunculated tubular adenoma. The forceps mark the base and the neck of the stalk. (This figure is printed in black and white as Figure 89-2.)



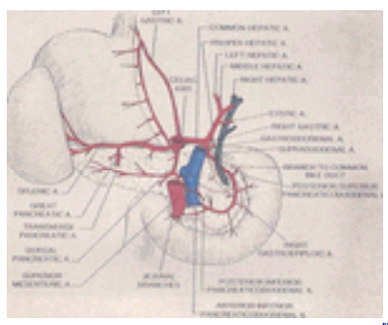
COLOR FIGURE 89-3. A synchronous, pedunculated, tubular adenoma adjacent to a sessile, tubulovillous adenoma. Synchronous adenomas not infrequently are found adjacent to each other. In this instance, a 1-cm, pedunculated, tubular adenoma is found about 5 cm from a 3-cm, sessile, tubulovillous adenoma. Note the smooth surface of the tubular adenoma and the smooth, lobulated surface of the tubulovillous adenoma. (This figure is printed in black and white as Figure 89-3.)



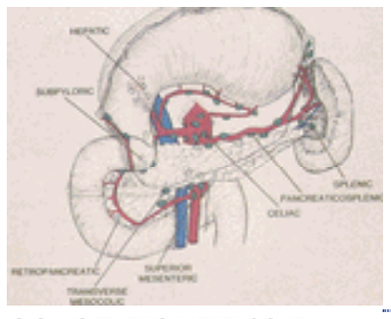
COLOR FIGURE 89-7. This cecal villous adenoma is virtually circumferential, with submucosal carcinomatous invasion. The lesion is sessile in nature and has a cauliflower appearance with a shaggy, frondlike, friable surface that contrasts with the smoother, lobulated surface seen in Color Figure 89-2. (This figure is printed in black and white as Figure 89-7.)



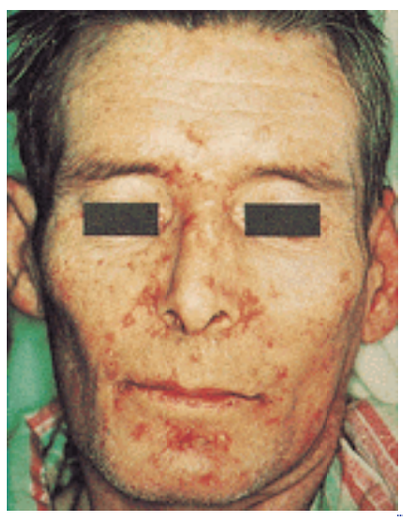
COLOR FIGURE 90-2. Early colonic polyposis in familial adenomatous polyposis. (This figure is printed in black and white as Figure 90-2.)



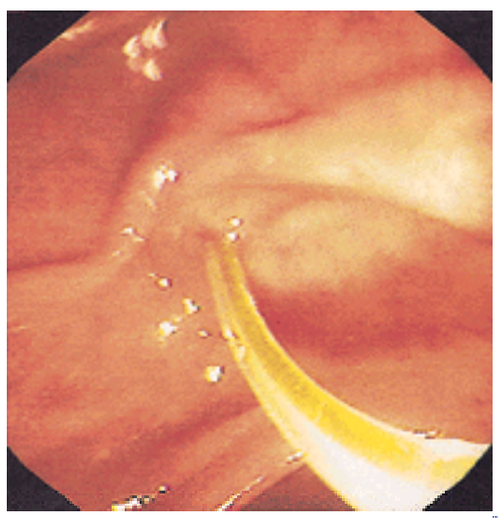
COLOR FIGURE 93-3. Blood supply of the pancreas. The pancreas, duodenum, stomach, and spleen are viewed from their posterior aspects. (This figure is printed in black and white as Figure 93-3.)



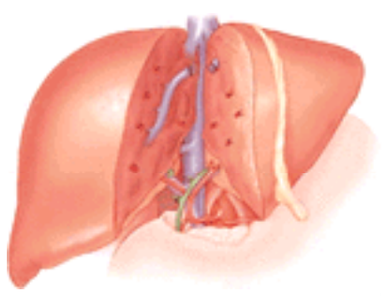
COLOR FIGURE 93-5. Lymphatic drainage of the pancreas. The pancreas is viewed from its anterior aspect. The gastrocolic ligament has been divided along the greater curvature of the stomach, which has been retracted anterosuperiorly. The transverse mesocolon has been detached from the peritoneum of the posterior abdominal wall. Labels indicate representative lymph nodes in the major regional nodal groups. (This figure is printed in black and white as Figure 93-5.)



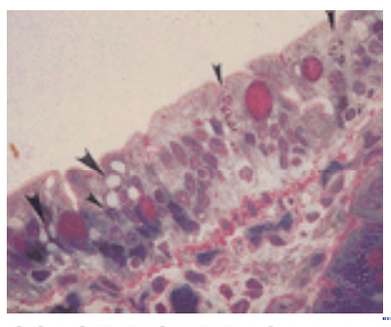
COLOR FIGURE 97-2. Migratory necrolytic erythema involving the face in a patient with metastatic glucagonoma. These typical skin lesions usually start on the extremities or intertriginous or periorificial sites. The lesions initially are erythematous and scaly, later become raised and bullous, and finally become crusty as is evident in this patient. Healing results in hyperpigmentation. Angular cheilitis, a common feature in patients with glucagonoma, is also present in a mild form in this patient. The patient also demonstrates loss of the buccal fat pad and temporal muscle wasting indicative of the generalized wasting these patients characteristically develop. (This figure is printed in black and white as Figure 97-2.)



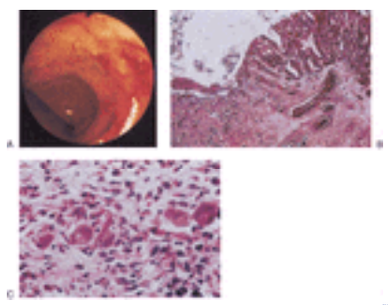
COLOR FIGURE 104-2. The duct entered during sphincter of Oddi manometry can be identified by aspirating the catheter. Dark-colored fluid (*top right*) signifies entry into the bile duct, whereas clear fluid indicates pancreatic duct entry. (This figure is printed in black and white as Figure 104-2.)



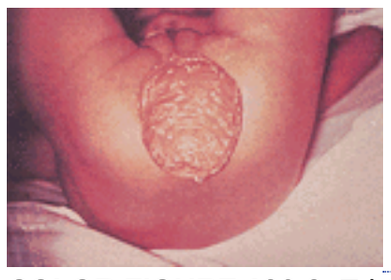
COLOR FIGURE 117-2. Illustration of technique of living donor right hepatic lobectomy. (Courtesy John P. Roberts, M.D., Department of Surgery, University of California, San Francisco.) (This figure is printed in black and white as Figure 117-2.)



COLOR FIGURE 124-1. A: Densely stained microsporidial spores are detected in the cytoplasm of several epithelial cells by light microscopic examination of an intestinal biopsy from a patient with microsporidiosis (semi-thin plastic section; methylene blue–azure II and basic fuchsin stain, original magnification × 630). (Courtesy of Dr. Jan M. Orenstein.) (This figure is printed in black and white as Figure 124-1.)

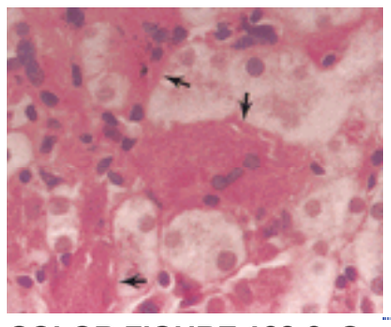


COLOR FIGURE 124-2. A: Endoscopic visualization of mucosal ulceration and inflammation in a patient with cytomegalovirus colitis. **B:** Light microscopic view of a colon biopsy from the same patient shows ulceration and hemorrhage. **C:** A higher magnification shows infiltration by inflammatory cells, many containing cytomegalic inclusions. (**B, C,** hematoxylin and eosin; **B,** original magnification × 30; **C,** original magnification × 62). (From ref. [211].) (This figure is printed in black and white as Figure 124-2.)

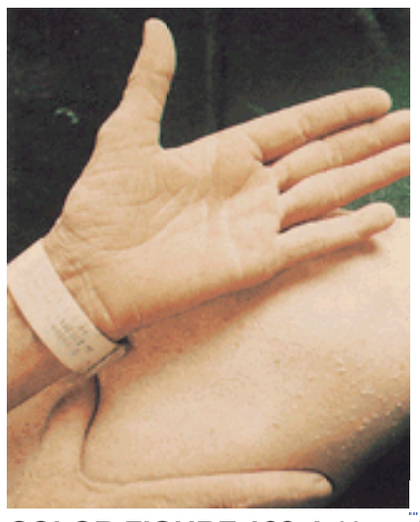


COLOR FIGURE 126-2. *Trichuris trichiura* associated with rectal prolapse in a child. Adult *T trichiura* are seen as white threads on the mucosal surface. (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.) (This figure is printed in black and white as Figure 126-2.)

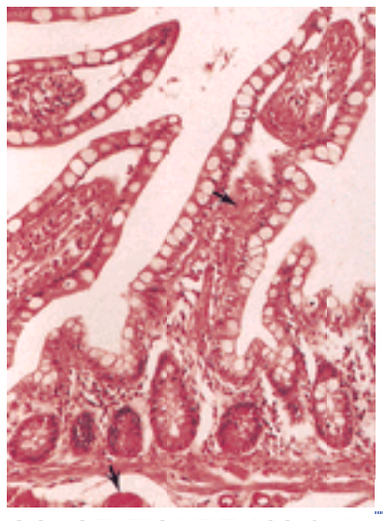
COLOR FIGURE 126-4. Intestinal obstruction caused by a mass of adult *Ascaris lumbricoides* is seen in this autopsy specimen. Intestinal obstruction is an unusual complication of heavy *Ascaris* infection. (From Smith JW, Ash LR, Thompson JH Jr, et al. Intestinal helminths. In: Atlas of diagnostic medical parasitology series. Chicago: American Society of Clinical Pathologists, 1984.) (This figure is printed in black and white as Figure 126-4.)



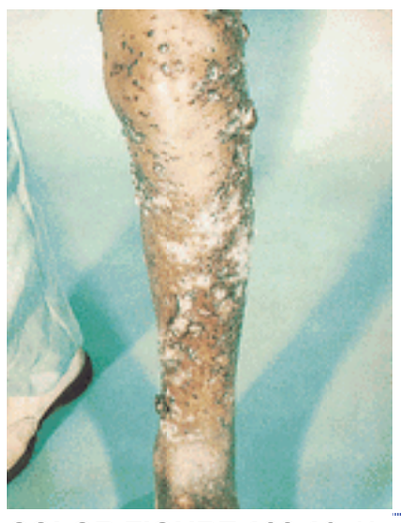
COLOR FIGURE 128-2. Gaucher disease of the liver. Multinucleated Gaucher cells (*arrows*) are present. (Hematoxylin and eosin stain; original magnification × 400.) (This figure is printed in black and white as Figure 128-2.)



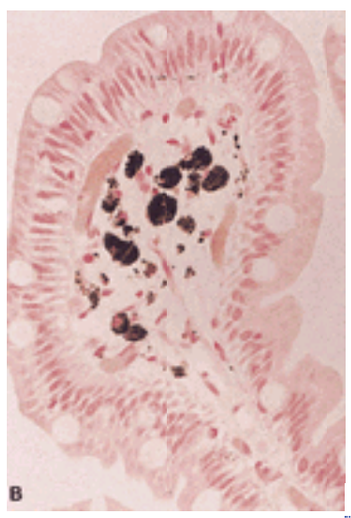
COLOR FIGURE 128-4. Hyperkeratosis of the palm and leg may be indicative of tylosis. (This figure is printed in black and white as Figure 128-4.)



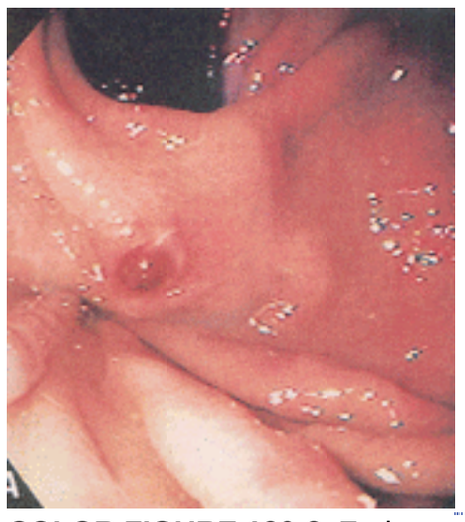
COLOR FIGURE 128-6. Amyloid infiltration of the small intestine (*arrows*). (Congo red stain; original magnification ×200.) (This figure is printed in black and white as Figure 128-6.)



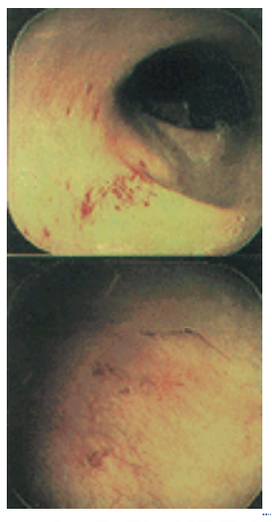
COLOR FIGURE 128-10. Kaposi sarcoma of the lower extremities. (This figure is printed in black and white as Figure 128-10.)



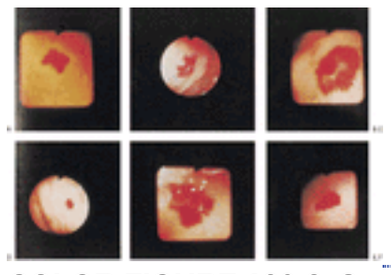
COLOR FIGURE 128-12. Duodenal biopsy specimen from a patient with pseudomelanosis duodeni shows the deposition of pigment within lamina propria macrophages. (This figure is printed in black and white as Figure 128-12.)



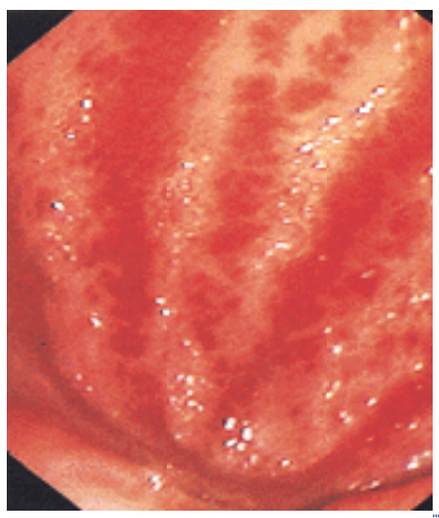
COLOR FIGURE 130-2. Endoscopic appearance of Dieulafoy lesion. Videophotograph shows a gastric cardial Dieulafoy lesion as a small, pigmented protuberance, which represents the vessel stump, with minimal surrounding erosion and no ulceration. (From: Lee JG, Leung JWC. Stigmata of recent hemorrhage in Dieulafoy's lesion. Gastrointest Endosc 2000;51:191.) (This figure is printed in black and white as Figure 130-2.)



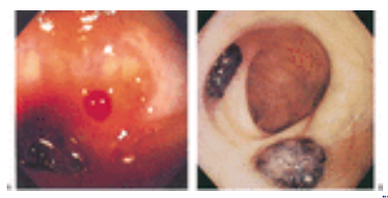
COLOR FIGURE 130-5. Endoscopic photo of cluster of angiodysplasia. **A:** Colonoscopic videophotography demonstrates two adjunct clusters of angiodysplasia in the descending colon in an elderly female with iron deficiency anemia; colonoscopy revealed no other angiodysplasia in the rest of the colon. **B:** Close-up views demonstrate the characteristic endoscopic appearance of angiodysplasia: an intensely red color, an intricate reticulonodular structure, and communication with prominent feeding or draining vessels. (From ref. [69].) (This figure is printed in black and white as Figure 130-5.)



COLOR FIGURE 130-8. Spectrum of endoscopic appearance of upper gastrointestinal angiodysplasia. The characteristic endoscopic findings of angiodysplasia are intense erythema, well-demarcated margin, irregular stellate (fern-like) margin, fine internal reticular (fern-like) structure, and macular mucosal location. Lesions range from 2 × 3 mm to 5 × 7 mm in size. All angiodysplasia are from the stomach, except for **A,** which is from the duodenum. (From ref. [80].) (This figure is printed in black and white as Figure 130-8.)

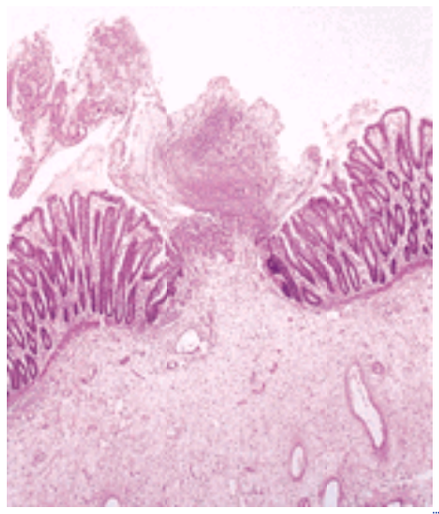


COLOR FIGURE 130-10. Videophotograph shows the characteristic endoscopic finding in gastric antral vascular ectasia (GAVE) of linear, intensely erythematous lesions at the apices of longitudinal antral folds radiating to the pylorus. The alternative lesion name of *watermelon stomach* derives from the resemblance of these erythematous linear streaks to the stripes on a watermelon rind. (From Clouse RE. Vascular ectasias, tumors, and malformations. In: Yamada T, Alpers DH, Laine L, Owyang C, Powell DW, eds. Atlas of gastroenterology. 2nd ed. Philadelphia: Lippincott Williams & Wilkins, 1999:550.) (This figure is printed in black and white as Figure 130-10.)

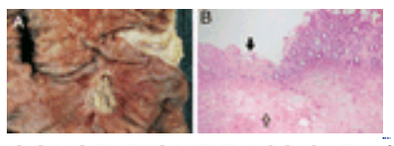


COLOR FIGURE 130-12. Endoscopic appearance of colonic hemangiomas. **A:** Videophotograph of a 3-mm colonic capillary hemangioma (bottom left) adjacent to a

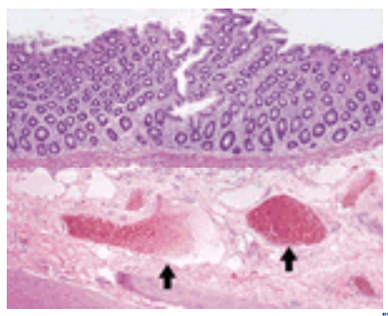
mid-sigmoid diverticulum (center). **B:** Videophotograph from the colon shows the characteristic endoscopic appearance of a cavernous hemangioma as a bluish, round, smooth, and well-circumscribed sessile polypoid lesion. (**A,** Courtesy of T.G. Tietjen, M.D., Petoskey, Michigan; **B,** From ref. [206] .) (This figure is printed in black and white as Figure 130-12.)



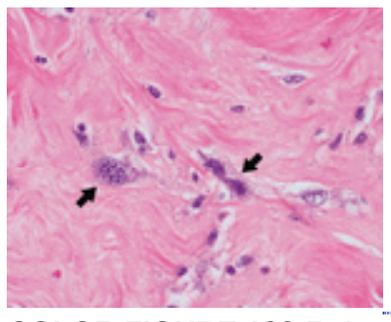
COLOR FIGURE 131-7. Intestinal ischemic damage. Well-delimited ulceration of the mucosa, with formation of a pseudomembrane. The remaining mucosa is normal. The submucosa has a marked edema and discrete infiltration by inflammatory cells. (This figure is printed in black and white as Figure 131-7.)



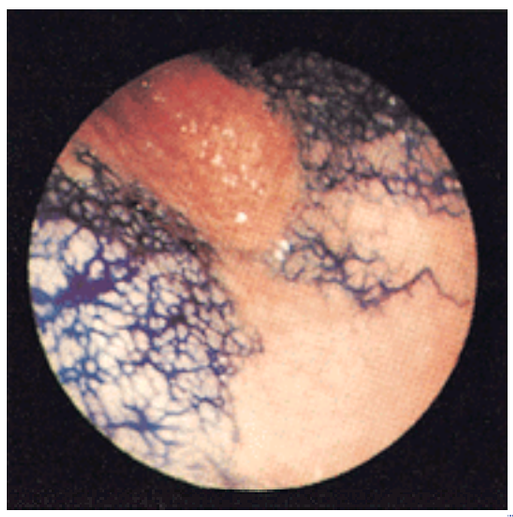
COLOR FIGURE 132-2. Radiation-induced ulceration in the colon. **A:** Gross appearance of a well-demarcated ulcer present in the rectum years after external radiation for an adjacent neoplasm. **B:** Histological appearance with chronic ulceration, mucosal necrosis (*solid arrow*), and dense submucosal fibrosis (*open arrow*) similar to that seen in ischemic injury. The lesion is notable for the absence of a prominent inflammatory infiltrate. (H&E, original magnification × 20.) (Courtesy of Dr. Christopher Moskaluk.) (This figure is printed in black and white as Figure 132-2.)



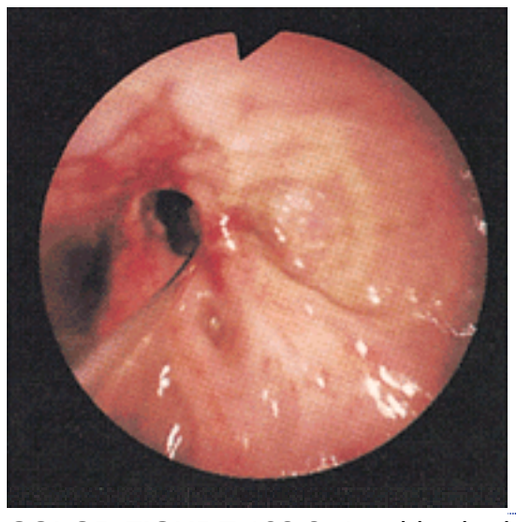
COLOR FIGURE 132-4. Submucosal telangiectasias (*arrows*) are seen in delayed radiation injury. Dilated venules and lymphatic channels are seen in the submucosa underlying relatively normal-appearing colonic epithelium. (H&E, original magnification × 40.) (Courtesy of Dr. Christopher Moskaluk.) (This figure is printed in black and white as Figure 132-4.)



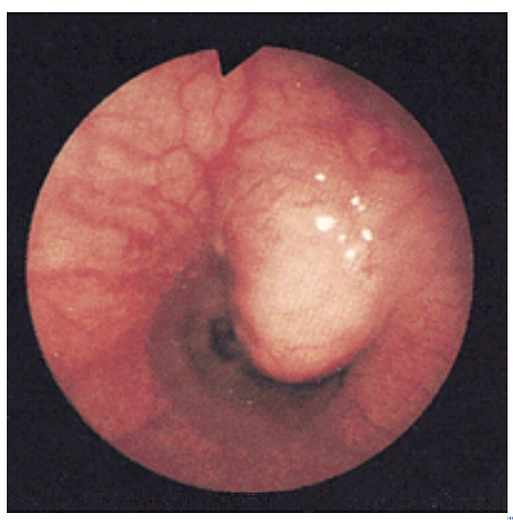
COLOR FIGURE 132-5. Atypical fibroblasts in radiation injury (*arrows*). Bizarre-appearing, pyknotic fibroblasts are frequently seen in the delayed phase of radiation injury in the alimentary tract. Although these atypical fibroblasts are frequently observed, their presence is not specific for radiation injury. (H&E, original magnification × 4000.) (Courtesy of Dr. Christopher Moskaluk.) (This figure is printed in black and white as Figure 132-5.)



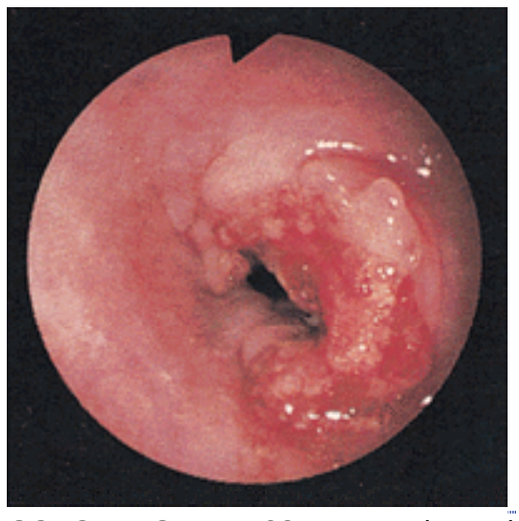
COLOR FIGURE 138-4. Gluten enteropathy with total villous atrophy. Mosaic appearance of the mucosal lining, accentuated by methylene blue entering deep mucosal crevices. (This figure is printed in black and white as Figure 138-4.)



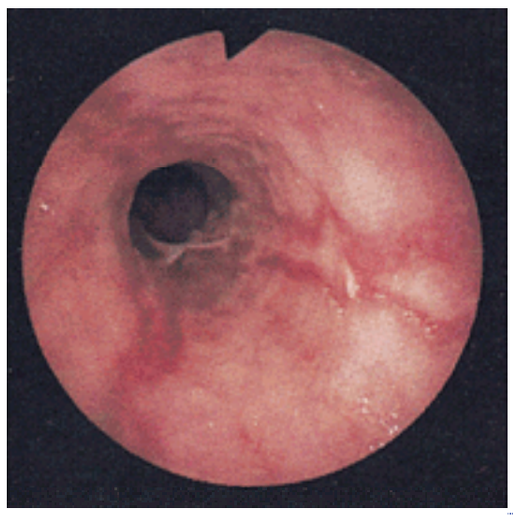
COLOR FIGURE 138-8. A guidewire has been passed through this eccentric, reflux-induced stricture. (This figure is printed in black and white as Figure 138-8.)



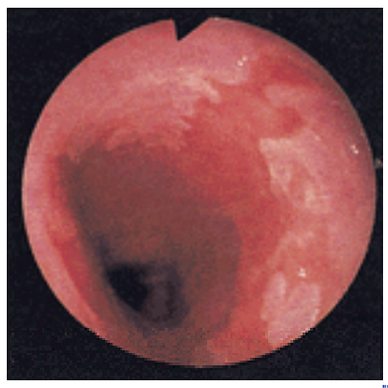
COLOR FIGURE 138-10. Esophageal leiomyoma. (This figure is printed in black and white as Figure 138-10.)



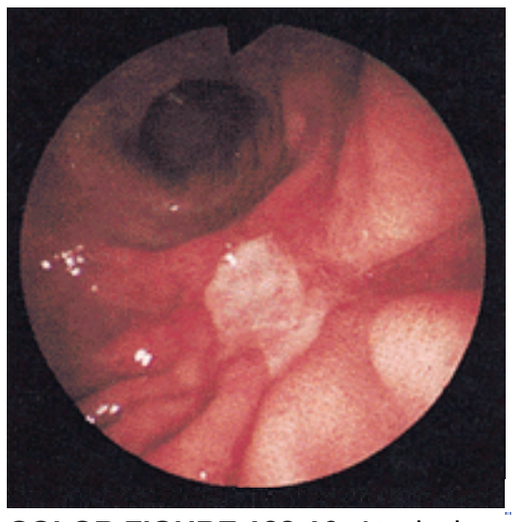
COLOR FIGURE 138-11. Esophageal carcinoma obstructs the esophagus. (This figure is printed in black and white as Figure 138-11.)



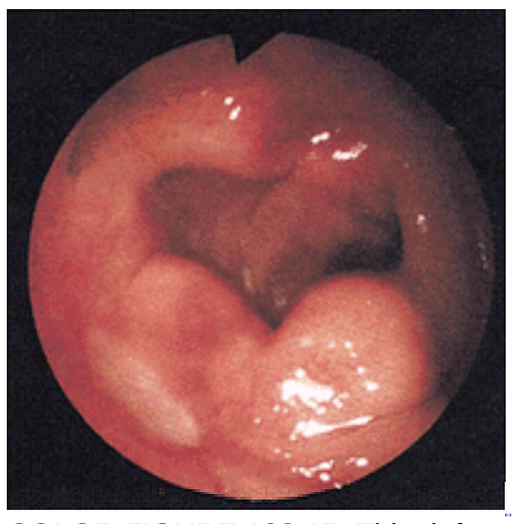
COLOR FIGURE 138-12. Reflux esophagitis with confluent but noncircumferential breaks or erosions. (This figure is printed in black and white as Figure 138-12.)



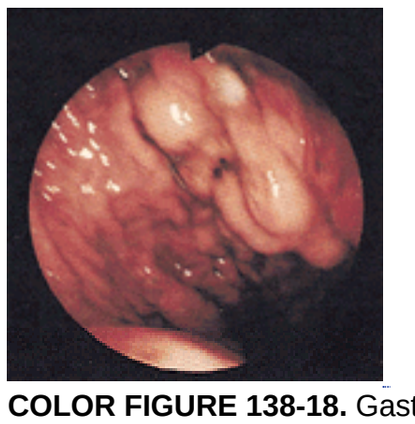
COLOR FIGURE 138-13. A columnar mucosa–lined esophagus, also known as Barrett esophagus. (This figure is printed in black and white as Figure 138-13.)



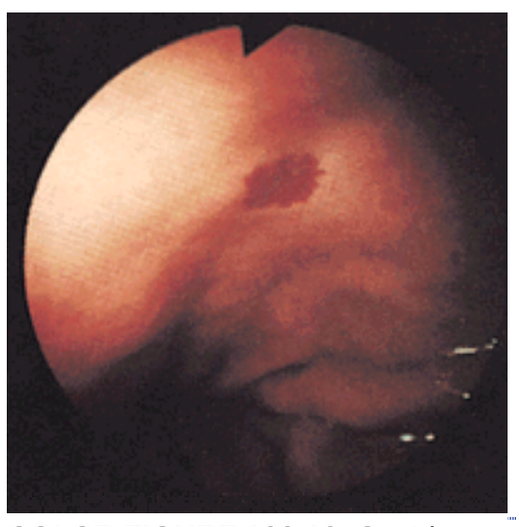
COLOR FIGURE 138-16. Atypical gastric ulceration along the greater curvature was presumably induced by nonsteroidal antiinflammatory drugs. (This figure is printed in black and white as Figure 138-16.)



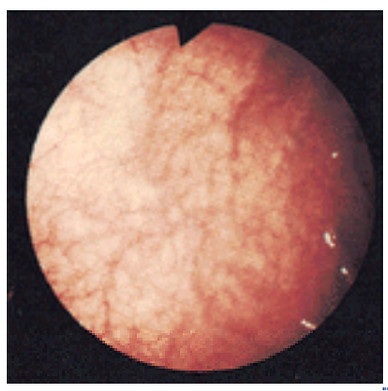
COLOR FIGURE 138-17. This deformed bulb was caused by scarring after prior ulceration and recurrent duodenal ulcer. (This figure is printed in black and white as Figure 138-17.)



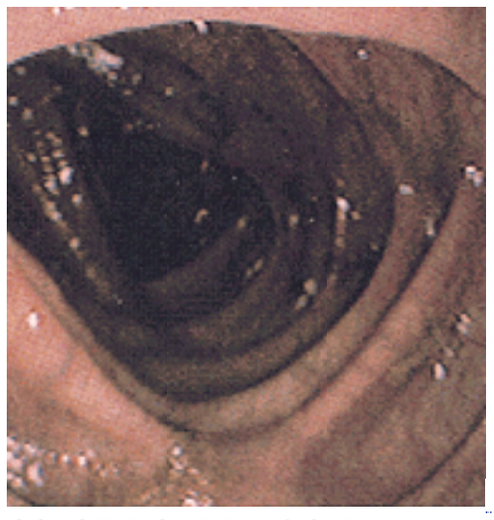
COLOR FIGURE 138-18. Gastric fundus varices. (This figure is printed in black and white as Figure 138-18.)



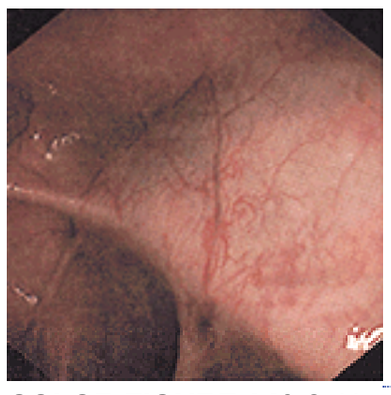
COLOR FIGURE 138-19. Gastric mucosal vascular ectasia. (This figure is printed in black and white as Figure 138-19.)



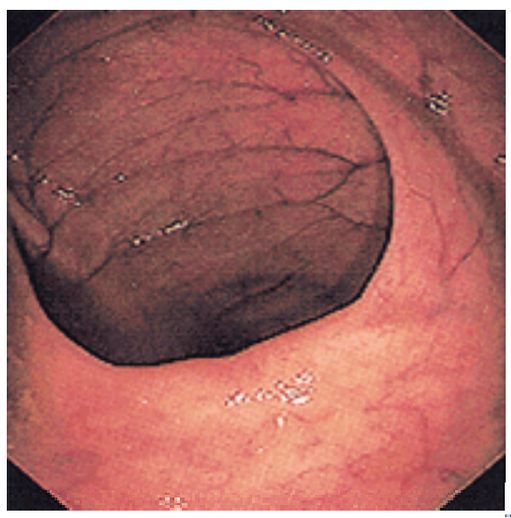
COLOR FIGURE 138-21. Atrophic gastritis. (This figure is printed in black and white as Figure 138-21.)



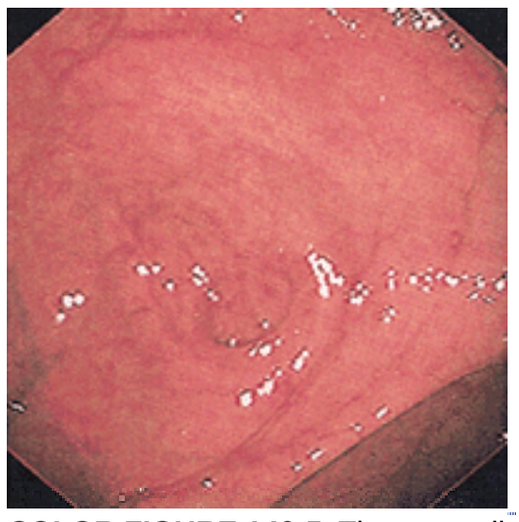
COLOR FIGURE 140-2. Transverse colon shows the characteristic triangular outline resulting from the relative thickness of the three longitudinal teniae coli. The circular muscle is much thinner. (This figure is printed in black and white as Figure 140-2.)



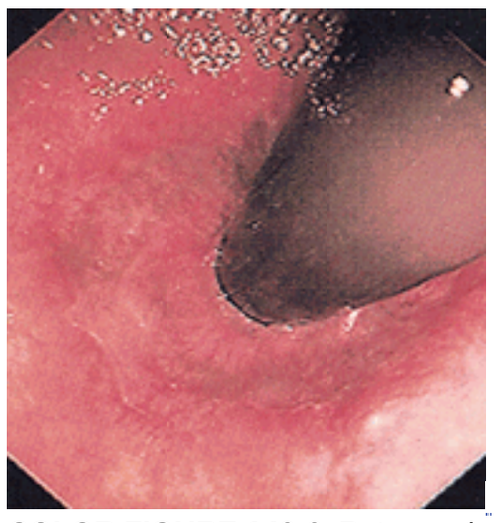
COLOR FIGURE 140-3. Hepatic flexure shows the dark impression of the liver. Similar discoloration can be caused by other extracolonic viscera at the splenic flexure or in the distal colon. Note the longitudinal impression of a tenia and the transverse haustral folds. (This figure is printed in black and white as Figure 140-3.)



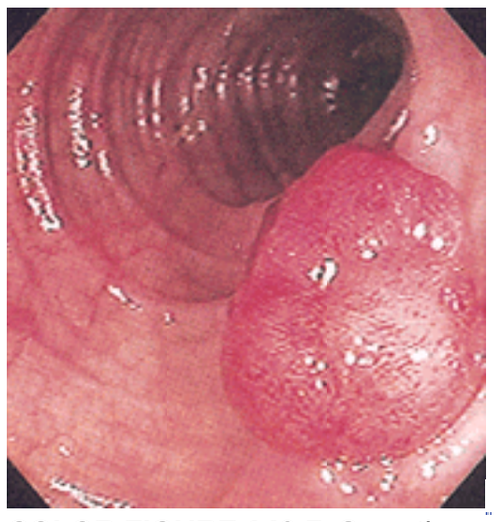
COLOR FIGURE 140-4. Ileocecal fold shows the characteristic notch on a flattened fold. The notch denotes the superior lip of the ileocecal valve. The opening is just proximal to and below the notch, and cannot be seen without marked angulation of the instrument tip. (This figure is printed in black and white as Figure 140-4.)



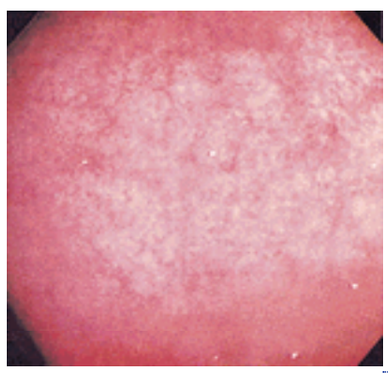
COLOR FIGURE 140-5. The appendix orifice is variable in configuration. Here, the orifice is round, but it may also be seen as a crescent-shaped slit. It is usually rather insignificant in appearance. The internal aspect remains the same after appendectomy. (This figure is printed in black and white as Figure 140-5.)



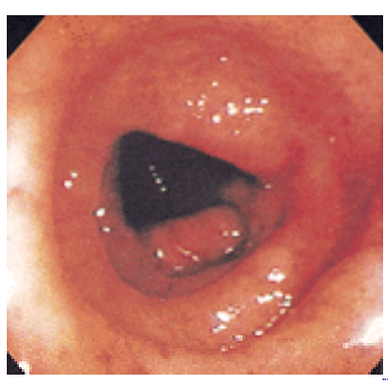
COLOR FIGURE 140-6. Retroversion of the endoscope in the rectum is a useful maneuver to find or remove small polyps in the distal part of the rectal ampulla. The dentate (pectinate) line is serpiginous at the junction of the squamous mucosa of the anal canal and the columnar rectal mucosa. (This figure is printed in black and white as Figure 140-6.)



COLOR FIGURE 140-7. Smooth sessile adenoma is located in the descending colon. (This figure is printed in black and white as Figure 140-7.)

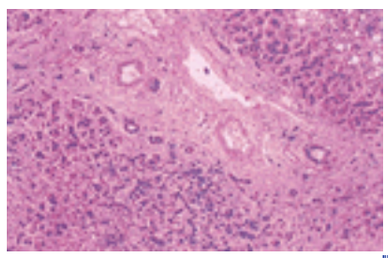


COLOR FIGURE 143-1. Cirrhosis of liver with fibrous septa and regenerative nodules. (Masson stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-1.)

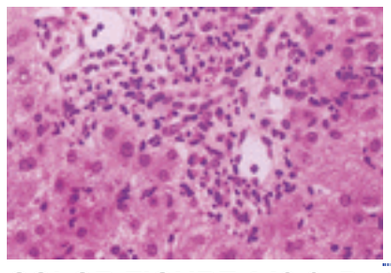


COLOR FIGURE 143-2. a $_1$-Antitrypsin globules in the periportal hepatocytes-diastase resistant PAS-positive. (Di-PAS stain; original magnification ×200.) (From

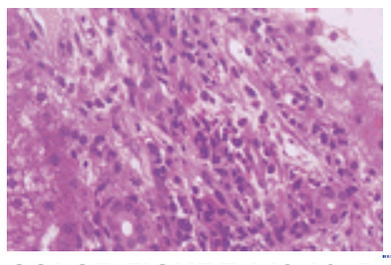
Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-2.)



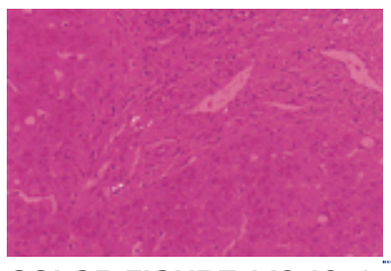
COLOR FIGURE 143-3. Perls iron stain demonstrating bright blue granules in hepatocytes and duct epithelial cells in hemochromatosis. (Original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-3.)



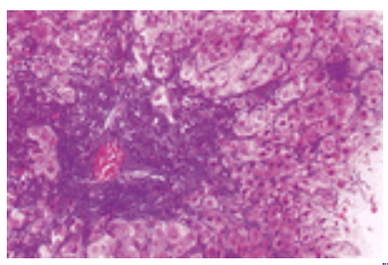
COLOR FIGURE 143-9. Portal area with prominent eosinophils among the inflammatory infiltrates in a case of Dilantin-induced hepatotoxicity. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-9.)



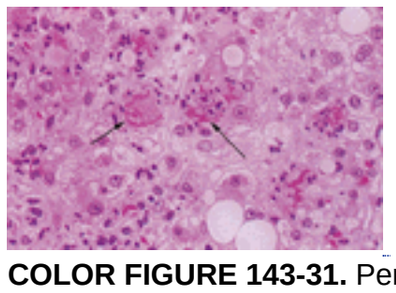
COLOR FIGURE 143-10. Portal area with increased number of eosinophils in a case of early rejection of orthotopic liver transplantation. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-10.)



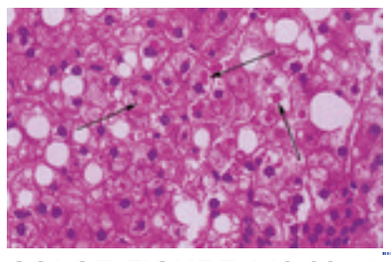
COLOR FIGURE 143-12. A portal area under polarizing light to demonstrate polarizable crystals in an intravenous drug user. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-12.)



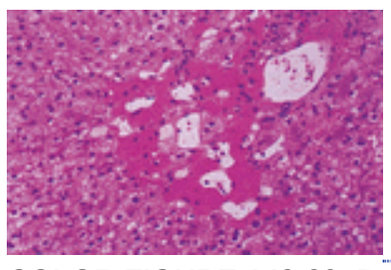
COLOR FIGURE 143-14. Arachnoid portal fibrosis with periportal extension of collagen in chronic alcoholic liver disease. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-14.)



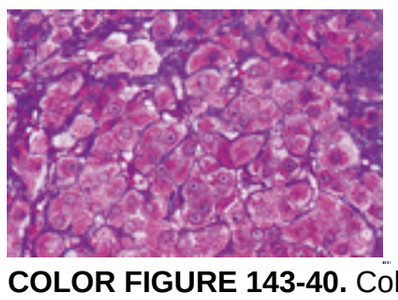
COLOR FIGURE 143-31. Perivenular hepatocytes containing Mallory hyaline (*arrows*) with neutrophilic reaction around them. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-31.)



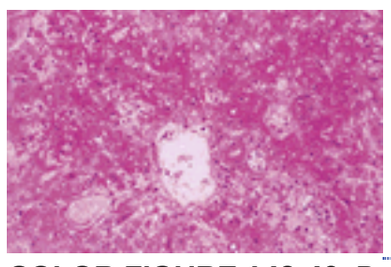
COLOR FIGURE 143-33. Hepatocytes containing spherical megamitochondria (*arrows*) in alcoholic liver disease. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-33.)



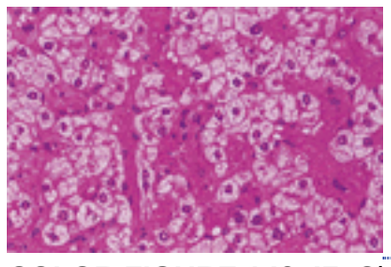
COLOR FIGURE 143-39. Perivenular hepatic parenchyma with dilated sinusoids and the presence of red blood cells within the hepatic cords in left-sided heart failure. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-39.)



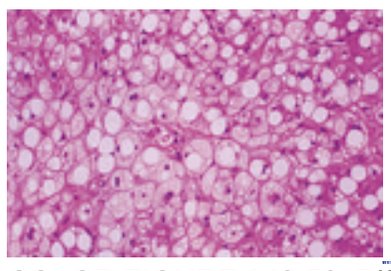
COLOR FIGURE 143-40. Collagen fibers along the sinusoids in the space of Disse in alcoholic liver disease. (Masson stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-40.)



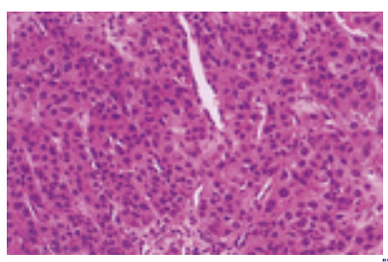
COLOR FIGURE 143-46. Periportal sinusoidal space filled with fibrin thrombi in toxemia of pregnancy. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-46.)



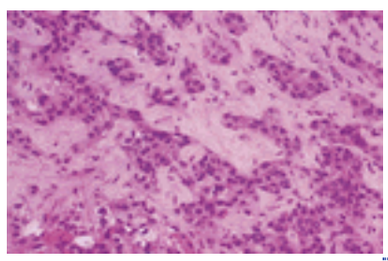
COLOR FIGURE 143-47. Clumps of sickled red blood cells packed in the sinusoidal spaces. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-47.)



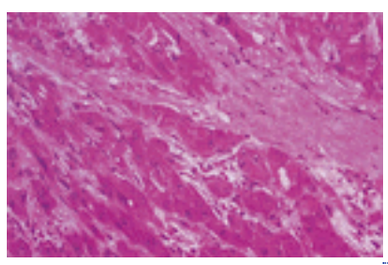
COLOR FIGURE 143-50. Diffusely enlarged hepatocytes with foamy fatty change in acute alcoholic liver disease. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-50.)



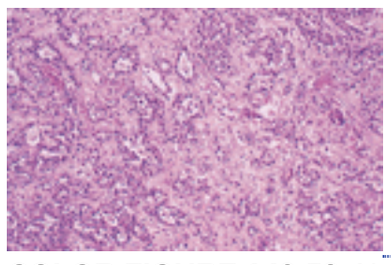
COLOR FIGURE 143-55. Well-differentiated trabecular hepatocellular carcinoma with endothelial lining. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-55.)



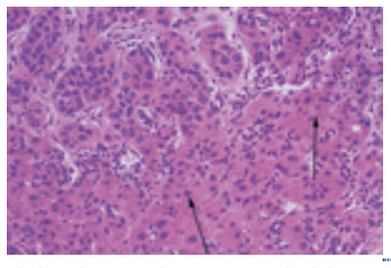
COLOR FIGURE 143-56. Sclerosing hepatic carcinoma with dense fibrous stroma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-56.)



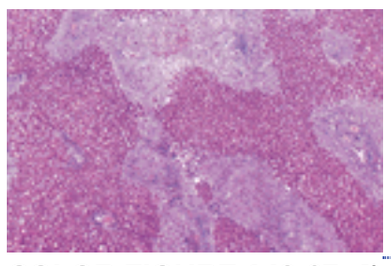
COLOR FIGURE 143-57. Eosinophilic neoplastic hepatocytes with lamellar fibrous stroma in fibrolamellar hepatocellular carcinoma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-57.)



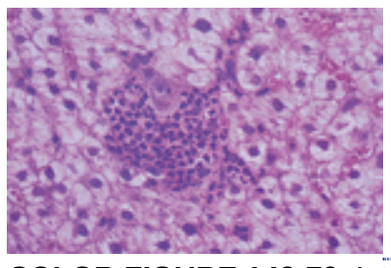
COLOR FIGURE 143-58. Neoplastic ductal structures with fibrous stroma in cholangiocarcinoma. (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-58.)



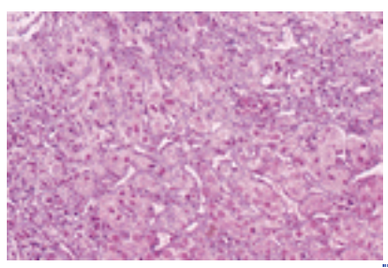
COLOR FIGURE 143-60. Junction of tumor and nontumor liver in hepatocellular carcinoma. The tumor cells grow into the hepatic cords (*arrows*). (H & E stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-60.)



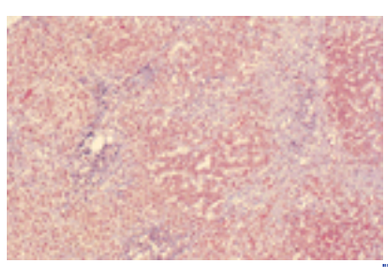
COLOR FIGURE 143-67. Jigsaw-puzzle appearance of biliary cirrhosis. (Masson stain; original magnification ×40.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-67.)



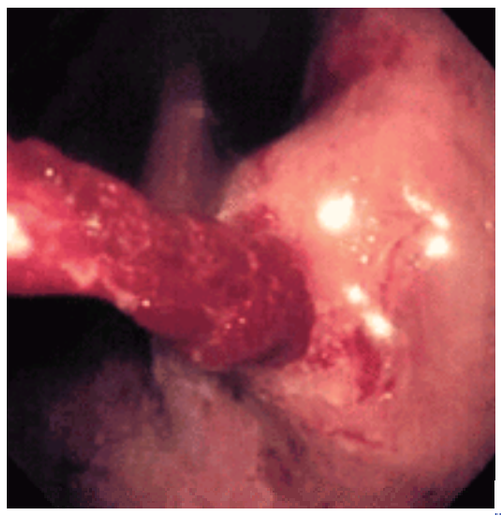
COLOR FIGURE 143-79. Intranuclear and cytoplasmic inclusions of cytomegalovirus (CMV) in a hepatocyte surrounded by polymorphonuclear leukocytes in an orthotopic liver transplant infected with CMV. (H & E stain; original magnification ×200.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-79.)



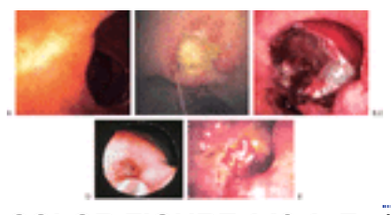
COLOR FIGURE 143-81. Diffuse interstitial fibrosis in chronic alcoholic liver disease. (Masson stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-81.)



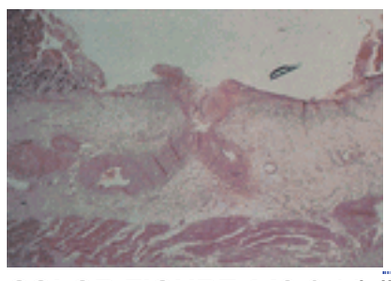
COLOR FIGURE 143-82. Marked perivenular fibrous scarring with mild portal fibrosis and lack of regenerative nodules in progressive perivenular fibrosis of alcoholic etiology. (Masson stain; original magnification ×100.) (From Govindarajan S. Liver biopsy interpretation. In: Kaplowitz N, ed. Liver and biliary diseases, 2nd ed. Baltimore: Williams & Wilkins, 1996.) (This figure is printed in black and white as Figure 143-82.)



COLOR FIGURE 147-2A. Cyanoacrylate injection for gastric varices. The injected cyanoacrylate stops bleeding from a fundal varix instanteously. (This figure is printed in black and white as Figure 147-2.)



COLOR FIGURE 148-1. Endoscopic photos of stigmata of recent hemorrhage. **A:** Clean base. Duodenal ulcer with a clean base. No evidence of protruding or pigmented areas is present. The rebleeding rate in this lesion is near zero. **B:** Flat spot. Pyloric channel ulcer with a flat, black spot in the center. The risk for rebleeding in this lesion is less than 10%. **C:** Adherent clot. Duodenal ulcer with a large adherent clot in the base that could not be removed with vigorous irrigation. The nature of the lesion beneath the clot is unknown. The risk for rebleeding in this lesion is intermediate (up to 25%). **D:** Nonbleeding visible vessel. A prominent protruding visible vessel with a bipolar probe in the foreground. This lesion has a high risk for recurrent bleeding (40% to 50%) without endoscopic therapy. **E:** Active bleeding. A duodenal ulcer with blood vigorously flowing. This is the lesion with the highest risk for continued or recurrent bleeding (approaching 60%). Endoscopic hemostasis can control active bleeding in most patients and can reduce the risk for rebleeding. (This figure is printed in black and white as Figure 148-1.)



COLOR FIGURE 148-6. Visible vessel. This classic image demonstrates the histology of a bleeding vessel in the base of an ulcer. The submucosal vessel is seen

extending to the ulcer surface, with a thrombus present in the “hole” in the arterial wall. (Courtesy of C. Paul Swain, M.D.) (This figure is printed in black and white as Figure 148-6.)